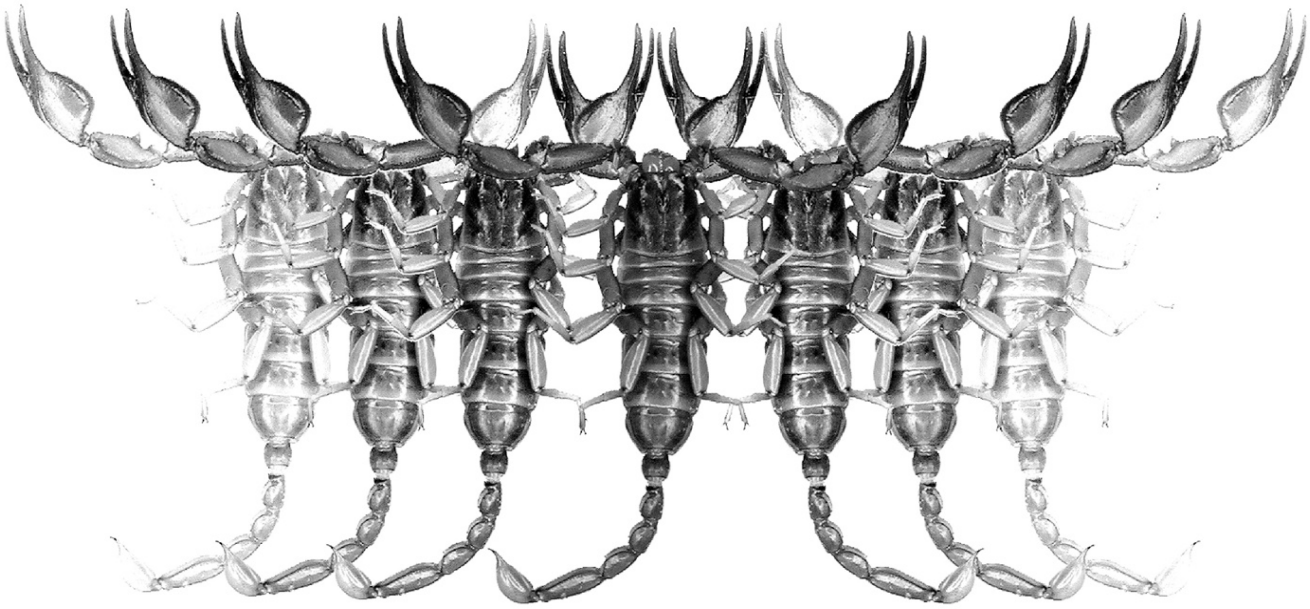
Euscorpius

Occasional Publications in Scorpiology



A new cryptic species of *Scorpiops* Peters, 1861 (Scorpiones: Scorpiopidae) from the northern Western Ghats, India

Shauri Sulakhe, Shubhankar Deshpande, Nikhil Dandekar, Makarand Ketkar, Anand Padhye & Deshabhushan Bastawade

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Occasional Publications in Scorpiology

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Euscorpius — Occasional Publications in Scorpiology. 2020, No. 327

A new cryptic species of *Scorpiops* Peters, 1861 (Scorpiones: Scorpiopidae) from the northern Western Ghats, India

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http://zoobank.org/urn:lsid:zoobank.org:pub:4C223102-4CE6-4440-910A-EDFD6A731715

Summary

A new cryptic species of *Scorpiops* (Scorpiopidae) is described from northern Western Ghats of India with integrated taxonomic approach. *Scorpiops telbaila* **sp**. **n**. is closely related to *S. tenuicauda* and differs from all species of *Scorpiops* in morphological features and raw genetic divergence of 5.4-14.1 %.

Introduction

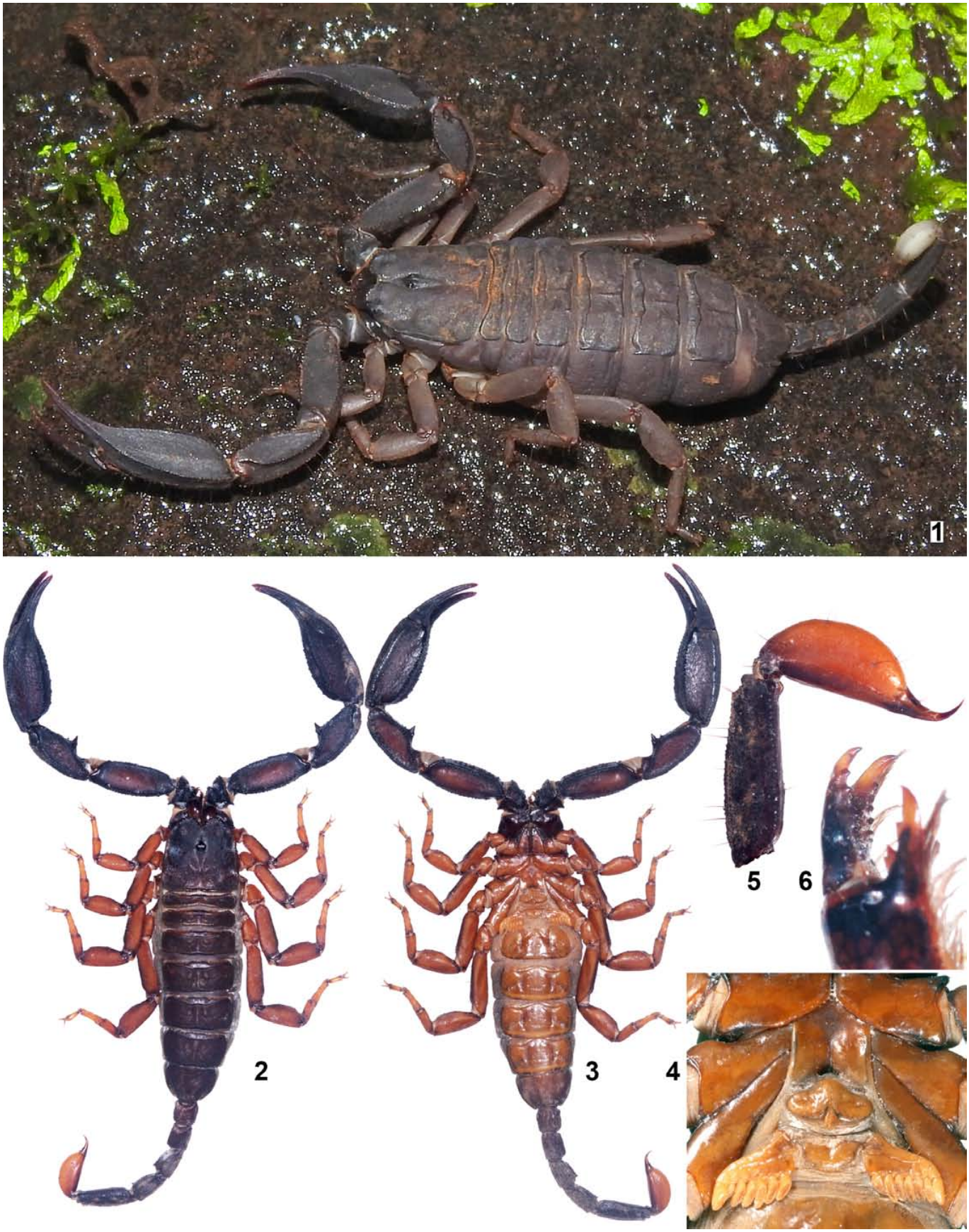
Recently, in a major taxonomic revision of family Scorpiopidae Kraepelin, 1905, all the genera, subgenera, and species were critically reviewed by Kovařík et al. (2020). In this study, genera *Neoscorpiops* Vachon, 1980, *Alloscorpiops* Vachon, 1980, *Dasyscorpiops* Vachon, 1974, *Euscorpiops* Vachon, 1980, *Plethoscorpiops* Lourenço, 2017, and *Vietscorpiops* Lourenço & Pham, 2015, and subgenus *Alloscorpiops* (*Laoscorpiops*) Lourenço, 2013, were all synonymized under *Scorpiops* Peters, 1861, based on detailed study of characters, variations in trichobothriotaxy and supported by strong statistical analysis. All main characters used for morphological comparisons are reviewed across genera and show congruence with the published DNA and cytogenetic analysis (Šťáhlavský et al., 2020).

After this revision, genus *Scorpiops* consists of 95 species distributed in Southeast Asia, out of which 22 are found in India.

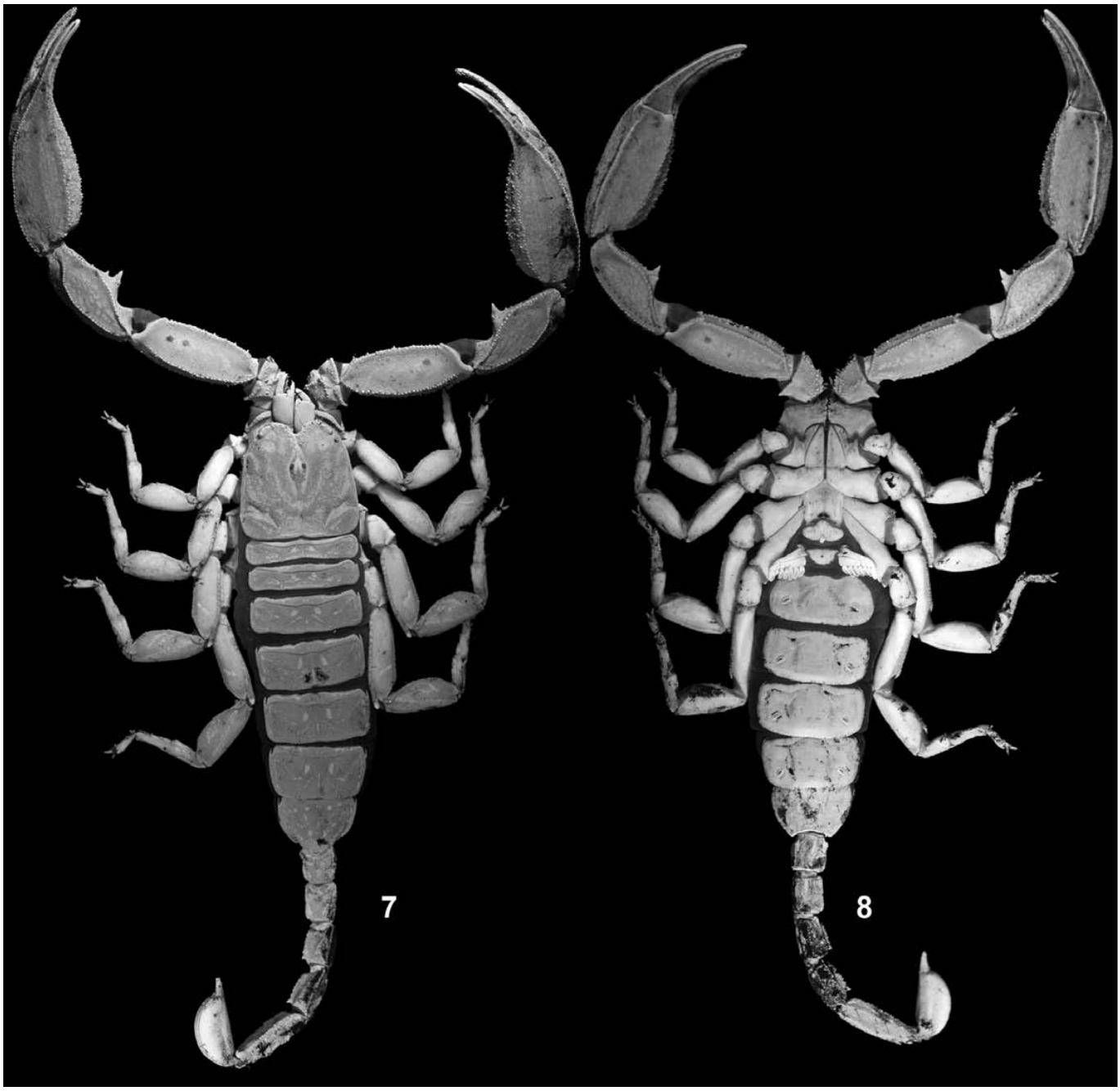
Only five species of this genus are found in the state of Maharashtra, including the recent description of *Scorpiops phaltanensis* Sulakhe et al., 2020 from Phaltan, on the eastern fringes of Western Ghats of India. The remaining four species of *Scorpiops* from Maharashtra are: *S. deccanensis* Tikader & Bastawade, 1983 (type locality: Sinhagad Fort, Pune District), *S. maharashtraensis* Mirza et al., 2014 (Shidi Ghat near Wadali Village, Aurangabad District). *S. satarensis* Pocock, 1900 (Mahabaleshwar, Satara District), and *S. tenuicauda* Pocock, 1894 (Matheran, Raigad District). Only one more species, *Scorpiops pachmarhicus* Bastawade, 1992, is found in Pachamarhi, Madhya Pradesh, while all the remaining Indian species of *Scorpiops* are from the Himalayas. Doubts were raised by Mirza et al. (2014) regarding the taxonomic validity of *S. deccanensis*, however Sulakhe et al. (2020c) established taxonomic validity of *Scorpiops* species found in Maharashtra, based on broad morphological comparisons and preliminary molecular phylogeny with integrated taxonomic approach. In this study, we describe a new cryptic species of *Scorpiops* from the northern Western Ghats, India with integrated taxonomic approach.

Methods, Material & Abbreviations

Sampling was carried out in Saltar Khind Pass, near Ambawne Village (18.58°N 73.36°E, 743 m a. s. l.), Pune District, Maharashtra State, India. Specimens were located with the help of ultra violet light (AmiciVision 18w 100 LED UV Torch), and collected. A total of 8 specimens were collected (5 males and 3 females). Photographs of holotype and paratype were taken using Nikon D500, 105mm F2.8 micro lens and R1C1 flash kit. Specimens were euthanized and preserved in absolute ethanol, and later transferred to 70% ethyl alcohol in collection jars for long term preservation. Examination and morphological measurements were done using LEICA EZ4HD microscope with LEICA application suite. Morphometry was performed following Stahnke (1971); morphological terminology follows Hjelle (1990). Measurements were taken (in mm) for 47 morphological characters (Table 1). The trichobothrial



Figures 1–**6**: *Scorpiops telbaila* **sp**. **n**. **Figure 1**. Female, paratype, INHER-SC-126, in vivo habitus. **Figures 2**–**6**. Male, holotype, dorsal (2) and ventral (3) views, sternopectinal area (4), metasoma V and telson in lateral view (5), and movable finger of chelicera, ventral fang, internal view (6).



Figures 7–8. *Scorpiops telbaila* **sp. n.**, male, holotype in dorsal (7) and ventral (8) views under UV light.

terminology follows Vachon (1974). Hemispermatophore was dissected using scalpel, pointed needles and was treated with 5% KOH and cleaned with clove oil. Hemispermatophore terminologies follow Monod et al. (2017) and Lamoral (1979). Specimens collected and studied are deposited in the museum collection of Bombay Natural History Society (BNHS), Mumbai and Institute of Natural History Education and Research (INHER), Research laboratory, Pune, Maharashtra, India.

Comparative material examined.

Data used for comparison, affinities and statistical analysis of *S. deccanensis*, *S. tenuicauda*, *S. maharashtraensis*, *S satarensis* and *S. phaltanensis* has been sourced from Sulakhe et al. (2020c). Data used for comparison of morphological characters of other *Scorpiops* species from Himalayas (India) and *S. pachmarhicus* has been sourced from Kovařík (2000, 2005, 2020), Mirza & Gowande (2016), and Kovařík et al. (2020).

Statistical analysis.

A Discriminant Function Analysis (DFA) using Principal Component Analysis (PCA) factors was conducted to assess the degree of morphological differentiation among the new species and their closest relatives. For multivariate PCA, a total of 19 characters of both sexes were transformed to their ratios to carapace median length (CML). Multivariate normality of

the size corrected variable was checked following Doornik and Hansen (2008) omnibus. Sets of 19 predictor variables were generated from PCA and all PCA factor scores were used as input variables for DFA to determine the classification success of our samples (Sulakhe et al., 2020a). PCA and DFA were performed using the statistical software PAST 3.25 (Hammer et al., 2001).

Following characters were used for statistical analysis: L (Length), W (Width), D (Depth), Carapace (W), Mesosoma VII (L), Metasomal Segment I, II, III, IV and V (L), Femur and Patella (L/W), Pedipalp Chela (L), Pedipalp Manus (W), Telson (L), Pectine, and Genital Operculum (L/W).

Molecular analysis

DNA extraction, *amplification and sequencing.*
Protocol as per Sulakhe et al. (2020a) was followed. Whole genomic DNA was extracted from preserved (ethanol 99.9%) muscle tissue (leg fragment) of *Scorpiops telbaila* **sp**. **n**. (Voucher numbers of specimens used for DNA analysis are mentioned in Table 4 and Figs. 51, 52) with the help of MACHEREY-NAGEL NucleoSpin® DNA Insect kit as per manufacturer's protocols. A 550-600 base pair (bp) fragment of the cytochrome c oxidase subunit I (*COI*) mitochondrial gene was amplified by polymerase chain reaction (PCR) using the primers as per Table 2. A 25 µl PCR reaction (TaKaRa Taq™ DNA Polymerase) was set containing 1 unit of Taq DNA polymerase (0.2µL), 2.5µL of 10x buffer, 2 µl of dNTPs (2.5mM each), 2 µl (5mM) of each primer, 2µl template DNA, and 14.3 µl of water, carried out with an Miniamp Thermal Cycler. Thermal cycler profiles used for amplification were as follows: 95° C for 3 min (denaturation temperature 95° C for 30 seconds, annealing temperature 50° C for 30 seconds, elongation temperature 72° C for 1 minute) x 35 cycles, 72° C for 7 minutes, hold at 4° C. PCR product was cleaned through column purification method with Qiagen PCR Cleanup Kit and sequenced with a 3730 DNA Analyzer. The sequencing primers were the same as those used in the PCRs. All sequences were deposited in the GenBank® nucleotide sequence database (http://www.ncbi.nlm.nih.gov) under accession numbers as per Table 4.

The sequences were also checked in BLAST (Altschul et al., 1990) tool to find the closest available sequences in the GenBank® and the related ones were downloaded for analysis.

Sequence alignment. Generated sequences were cleaned manually in MEGA 7 (Kumar et al., 2016) using chromatograms visualised in Chromas V.2.6.5 (Technelysium PTY. Ltd.). Cleaned and downloaded sequences were aligned using MUSCLE (Edgar, 2004) implemented in MEGA 7 (Kumar et al., 2016) using default parameters. The final alignment contained 16 sequences each of 519 bp length including 2 sequences of *Euscorpius phrygius* Bonacina, 1980 (Table 4) used as out-group to route the phylogenetic tree. This alignment was used in molecular phylogenetic analyses.

Molecular phylogenetic analysis.
Maximum Likelihood (ML) and Bayesian Inference (BI) methods of phylogenetic analysis were implemented. The COI region was partitioned per codon position and the best substitution model for phylogenetic analysis was determined using PartitionFinderV.1.1.1 (Lanfear et al. 2012). Model search was performed during the Bayesian Information Criterion (BIC) with a greedy search algorithm (Schwarz, 1978). Maximum Likelihood analysis was performed in raxmlGUI (Silvestro & Michalak, 2011) under the GTR + G + I model of sequence evolution, 1000 non-parametric bootstrap pseudo-replicates with rapid ML search were performed. Bayesian trees were generated using MrBayes V.3.2.6 (Ronquist et al., 2012). The models of sequence evolution were as follows: - F81+I for codon position 1, HKY+G for codon position 2 and HKY+I for codon position 3. Two simultaneous, independent analyses were run starting from different random trees. Three heated and one cold chain was used in the analysis. Markov chains were sampled every 500 generations for 5 million generations. At the end of the run we ensured convergence of the two MCMC runs by ensuring that the standard deviation of split frequencies was less than 0.002 and by checking the trace plots using Tracer v. 1.7 (Rambaut et al., 2003). We also ensured that the ESS values for all the parameters were above 200. A total of 25% trees were discarded as burn-in. The tree representing the best evolutionary hypothesis was selected using a 50% majority consensus rule. Un-corrected pairwise genetic divergence "*p*-distance" was calculated in MEGA 7 (Kumar et al., 2016).

Species delimitation analysis.
Species delimitation analysis was performed on the BI tree using Bayesian Poisson Tree Process using 500000 Markov chain Monte Carlo (MCMC) generations with thinning parameter of 100 and burn-in of 0.1 (Zhang et al. 2013).

Systematics

Family Scorpiopidae Kraepelin, 1905
Scorpiops Peters, 1861
(Figures 1–52, Tables 1–4)
http://zoobank.org/urn:lsid:zoobank.org:act:45E3D60F-43C5-4655-9675-E8C72D771112

Type species. *Scorpiops hardwickii* Gervais, 1843

***Scorpiops telbaila* sp. n.**
(Figures 1–34, 40, 46–49, Tables 1–4)
http://zoobank.org/urn:lsid:zoobank.org:act:D907B140-9AA0-4572-AC3B-3853EDB431C8

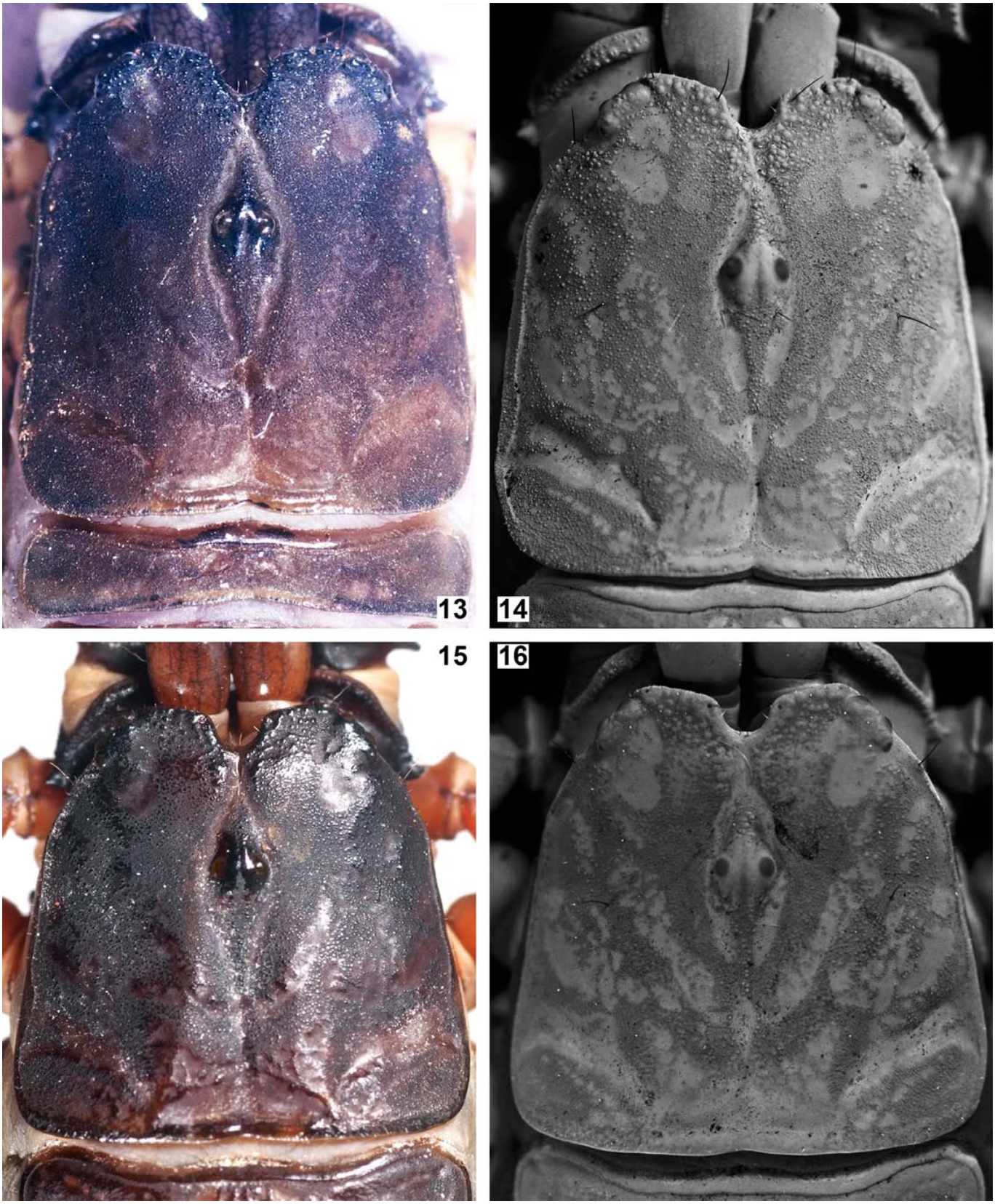
Type locality and type repository. **India**, Maharashtra State, Pune District, Saltar Khind Pass, near Ambawne Village, 18°34'38"N 73°21'25"E, 743 m a. s. l.; BNHS.

Dimensions (mm)		*Scorpiops telbaila* sp. n.				
		♂ holotype BNHS SC 175	♂ paratype BNHS SC 176	♂ paratype BNHS SC 177	♂ paratype INHER 123	♂ paratype INHER 223
Carapace	L / W	7.3 / 8.1	6.5 / 7.0	5.4 / 4.9	6.5 / 7.0	7.3 / 7.3
Mesosoma	L	19.2	18.2	15.6	15.4	19.6
Tergite VII	L / W	3.2 / 5.3	3.5 / 5.4	2.9 / 4.4	3.5 / 5.4	4.3 / 5.8
Metasoma and telson	L	22.5	20.9	15.5	20.1	22.4
Segment I	L / W / D	1.9 / 2.2 / 2.2	2.0 / 2.5 / 2.3	1.5 / 1.9 / 1.7	2.1 / 2.5 / 2.2	2.2 / 2.7 / 2.1
Segment II	L / W / D	2.4 / 2.2 / 2.0	2.3 / 2.3 / 2.0	1.6 / 1.7 / 1.4	2.2 / 2.2 / 1.9	2.4 / 2.3 / 1.9
Segment III	L / W / D	2.7 / 2.1 / 1.9	2.5 / 2.1 / 1.9	1.9 / 1.6 / 1.5	2.4 / 2.0 / 1.9	2.7 / 2.2 / 2.0
Segment IV	L / W / D	3.1 / 2.0 / 2.0	2.9 / 1.9 / 1.9	2.0 / 1.4 / 1.4	2.7 / 1.9 / 1.9	3.0 / 2.0 / 2.0
Segment V	L / W / D	5.1 / 1.9 / 1.9	4.9 / 1.8 / 1.8	3.7 / 1.4 / 1.4	4.7 / 1.9 /1.8	5.3 / 2.0 / 1.9
Telson	L / W / D	6.5 / 2.1 / 2.4	6.3 / 2.1 / 2.3	4.9 / 1.4 / 1.4	6.1 / 2.0 / 2.2	6.9 / 2.1 / 2.3
Pedipalp	L	32.6	31.4	24.0	30.9	33.7
Femur	L / W / D	9.0 / 3.4 / 1.6	8.7 / 3.1 / 1.4	6.1 / 2.5 / 1.2	8.1 / 3.2 / 1.5	8.8 / 3.5 / 1.7
Patella	L / W / D	7.7 / 4.2 / 2.1	7.4 / 3.9 / 2.0	5.8 / 3.1 / 1.5	7.2 / 3.9 / 2.0	7.7 / 3.4 / 2.2
Chela	L	15.9	15.3	12.2	15.6	17.2
Manus	W / D	4.7 / 2.9	4.3 / 2.6	3.1 / 1.8	4.1 / 2.6	4.8 / 3.3
Movable finger	L	7.6	7.2	5.6	7.0	8.2
Pectine	L / W	2.6 / 1.5	2.6 / 1.3	2.3 / 1.1	2.6 / 1.2	3.0 / 2.4
Genital Operculum	L / W	1.5 / 2.8	1.4 / 2.6	1.2 / 2.4	1.6 / 2.6	1.7 / 2.8
Total	**L**	**47.6**	**45.6**	**36.4**	**42.5**	**49.4**
Pectinal teeth count	PTC	7 / 7	7 / 7	8 / 8	7 / 8	7 / 6
Trichobothria count	TPV	15 / 14	15 / 16	15 / 16	15 / 16	16 / 16
Trichobothria count	TPE	24 / 25	25 / 27	26 / 26	26 / 26	23 / 24
Chelicera	DVC	6 / 6	6 / 6	5 / 6	8 / 8	6 / 6

Dimensions (mm)		*Scorpiops telbaila* sp. n.			*S. montanus*
		♀ paratype BNHS SC178	♀ paratype INHER 124	♀ paratype INHER 126	♂ INHER 168
Carapace	L / W	6.9 / 7.1	6.4 / 6.5	4.7 / 4.7	7.1 / 4.5
Mesosoma	L	21.9	16.6	14.6	17.6
Tergite VII	L / W	4.0 / 6.0	3.1 / 5.5	2.3 / 4.3	3.7 / 5.2
Metasoma and telson	L	20.5	18.0	13.21	23.7
Segment I	L / W / D	2.0 / 2.3 / 2.0	1.7 / 2.1 / 1.7	1.2 / 1.7 / 1.5	2.3 / 2.6 / 2.2
Segment II	L / W / D	2.2 / 2.2 / 1.8	2.0 / 2.0 / 1.6	1.4 / 1.5 / 1.3	2.5 / 2.3 / 2.2
Segment III	L / W / D	2.4 / 2.0 / 2.0	2.3 / 1.7 / 1.6	1.6 / 1.5 / 1.3	2.6 / 2.3 / 2.2
Segment IV	L / W / D	2.8 / 1.8 / 1.9	2.4 / 1.6 / 1.7	1.9 / 1.4 / 1.3	3.0 / 2.1 / 2.2
Segment V	L / W / D	4.8 / 1.7 / 1.7	4.5 / 1.6 / 1.6	3.4 / 1.4 / 1.2	5.8 / 1.9 / 1.9
Telson	L / W / D	6.3 / 1.7 / 1.9	5.1 / 1.6 / 1.7	3.7 / 1.2 / 1.2	7.4 / 2.3 / 2.3
Pedipalp	L	30.5	28.2	19.9	33.8
Femur	L / W / D	8.2 / 3.0 / 1.6	7.5 / 2.8 / 1.6	5.1 / 2.1 / 1.0	9.2 / 3.6 / 2.0
Patella	L / W / D	7.3 / 3.9 / 2.4	6.9 / 3.7 / 1.9	4.7 / 2.7 / 1.5	7.9 / 3.4 / 2.8
Chela	L	15.0	13.7	10.1	16.8
Manus	W / D	4.5 / 3.3	3.9 / 2.5	2.8 / 1.7	4.8 / 3.3
Movable finger	L	7.0	6.4	4.7	7.7
Pectine	L / W	2.8 / 1.5	2.2 / 1.4	1.8 / 0.7	4.2 / 2.4
Genital Operculum	L / W	1.7 / 3.1	1.3 / 2.9	0.9 / 2.2	1.9 / 2.8
Total	**L**	**49.4**	**47.6**	**32.6**	**48.4**
Pectinal teeth count	PTC	6 / 6	7 / 7	6 / 6	8 / 8
Trichobothria count	TPV	16 / 16	14 / 16	16 / 16	15 / 16
Trichobothria count	TPE	25 / 25	27 / 27	25 / 26	17 / 17
Chelicera	DVC	6 / 7	6 / 6	6 / 6	6 / 6

Table 1. Comparative measurements of adults of *Scorpiops telbaila* **sp**. **n**. Abbreviations: length (L), width (W, in carapace it corresponds to median width), depth (D), holotype (HT), paratype (PT), pectinal teeth count (PTC), trichobothria on pedipalp patella ventral count (TPV), trichobothria on pedipalp patella external count (TPE), dentition on ventral fangs of chelicera count (DVC).

Figures 9–12. *Scorpiops telbaila* **sp**. **n**., female, paratype, BNHS SC 178, in dorsal (9) and ventral (10) views, sternopectinal area (11), and metasoma V and telson in lateral view (12).



Figures 13–16: *Scorpiops telbaila* **sp. n. Figures 13**–**14**. Male, holotype, carapace under white (13) and UV (14) light. **Figures 13–14**. Female, paratype, BHNS SC 178, carapace under white (15) and UV (16) light.

Type material. **India**, Maharashtra State, Pune District, Saltar Khind Pass, near Ambawne Village, 18°34'38"N 73°21'25"E, 743 m a. s. l., 1♂ (holotype, BNHS SC 175), 16 August 2019, 3♂ (paratypes, INHER-123, BNHS SC 176, 177), 8 June 2020, 1♂ (paratype, INHER-223), 3♀ (paratypes, INHER-124, 126, BNHS SC 178), 16 August 2019. All specimens collected by S. Sulakhe, S. Deshpande & M. Ketkar.

Etymology. The species epithet is a noun in opposition, named after a famous hill fort "Telbaila", used as a watch tower by Maratha warriors from India. It is located in Pune District, northern Western Ghats, very close to the type locality of the new species.

Diagnosis (♂♀). Total length 32–50 mm. Base color uniformly dark brownish to blackish. Pectinal teeth number 6–8 in both sexes, fulcra reduced to absent. Pectines have three marginal lamellae and three middle lamellae present. Patella of pedipalp with 23–27 (5 *eb*, 2 *esb*, 2 *em*, 9–11 *est*, 5–7 *et*) external and 14–16 ventral trichobothria. Chela of pedipalp with 4 ventral trichobothria located on ventral surface. Chelal trichobothrium Eb_3 is located in proximal half of manus between trichobothria *Dt* and *Db*. Fingers of pedipalps strongly undulate in male and margins undulate in female. Chela length to width ratio 3.6–4.0 in males. Pedipalp movable finger with ca 55–60 IAD, which form second row, parallel with MD (ca 80 in number); there are also 4–5 ID and 11–12 OD present. Tarsomere II of legs with 4–6 stout median ventral spinules and two pairs of flanking setae. Metasoma I with ten and metasoma II–IV with eight carinae. Telson elongate and smooth, length to depth ratio 2.7–3.5; annular ring developed.

Description (♂ holotype, measurements in Table 1).

Coloration (in preservation) (Figs. 2, 3, 9, 10). Overall body color dark brownish to blackish. Legs uniformly brownish. Telson brownish orange on vesicle and dark brown on aculeus. Ventral portion of body yellowish brown.Carapace and fingers of manus blackish. Pedipalps dark brown, darker on carinae. Chelicera basal segment blackish brown. Fingers of chelicera dark brown.

Carapace (Figs. 13–17). Anterior margin of carapace with deep emargination in the middle. Entire surface of carapace mixed with fine and coarse granules. Anterior margin of carapace with strong tuberculate granules. Lateral ocular tubercles granular with three pairs of lateral eyes. Anterior two pairs larger and third pair smaller in size. Median ocular tubercle granular on dorsal portion with a pair of median eyes situated in the ratio of 1:2.1 (ratio of median eyes to anterior margin and median eyes to posterior margin).

Chelicerae (Fig. 6). Proximal portion with reticulated mosaic design. Fixed finger of chelicera with 3 large triangular teeth on inner margin. Ventral fang of movable finger with a row of 6 minute teeth on inner margin. Dorsal fang of movable finger with 4 teeth on inner margin.

Pedipalp (Figs. 20–26, 34). Femur and patella dorsoventrally flattened and evenly carinated. Intercarinal space finely and almost evenly granular. Internal surface of patella with one large posterio-ventral tubercle (Fig. 19, tubercle 1) and with one small, thick adjacent bulge, one adjacent small posterio-dorsal tubercle (Fig. 19, tubercle 1a) and one small, anterio-ventral tubercle (Fig. 19, tubercle 2). Manus elongated, carinated and intercarinal space coarsely granular on inner and outer surface. Dorsal exterior carina evenly granular, running anteriorly up to the base of fixed finger. Both fingers with two rows of dentitions, scalloped deeply at the base. Trichobothrial pattern typical of the genus.

Legs (Figs. 2, 3, 7, 8, 27–30). Femur and patella carinated, intercarinal space almost smooth. Tarsomere I provided with three rows of spinules and tarsomere II with single ventral row of spinules.

Sternum, **genital operculum and pectines** (Figs. 4, 11). Broad, pentagonal and finely granular only on anterior middle portion. Genital operculum with a pair of strongly protruding genital papillae. Basal piece with slight depression on middle portion. Pectines with 7/7 pectinal teeth.

Mesosoma (Figs. 2, 3, 7–10). All tergites finely granular, with median carina absent on segment I. Tergite VII additionally with a pair of lateral granular carina present only on half posterior portion. All sternites entirely smooth. Sternite VII finely granular only on lateral portion.

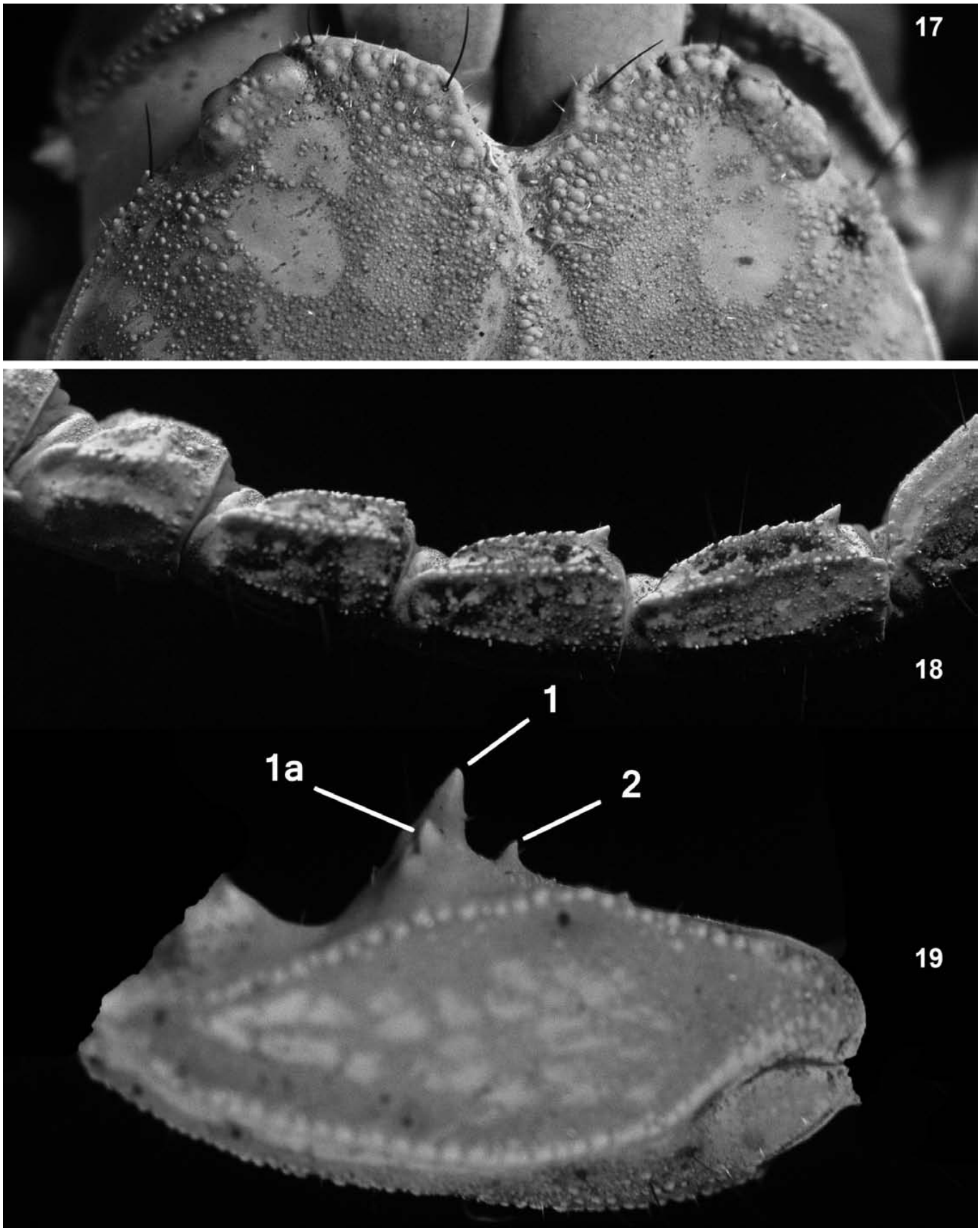
Metasoma (Figs. 2–5, 7–10, 18, 40). Metasomal segments I-V with 10-8-8-8-5 carinae. Intercarinal space densely granular. Dorsal carination on segments III and IV ending posteriorly into a strong tuberculate spine, more pointed on segment IV. Anal rim of segment V evenly crenulated. A pair of dorsolateral granules of anal rim weakly tuberculate.

Telson (Figs. 5, 12). Elongated and almost entirely smooth. A prominent depression present in between vesicle and at the base of aculeus.

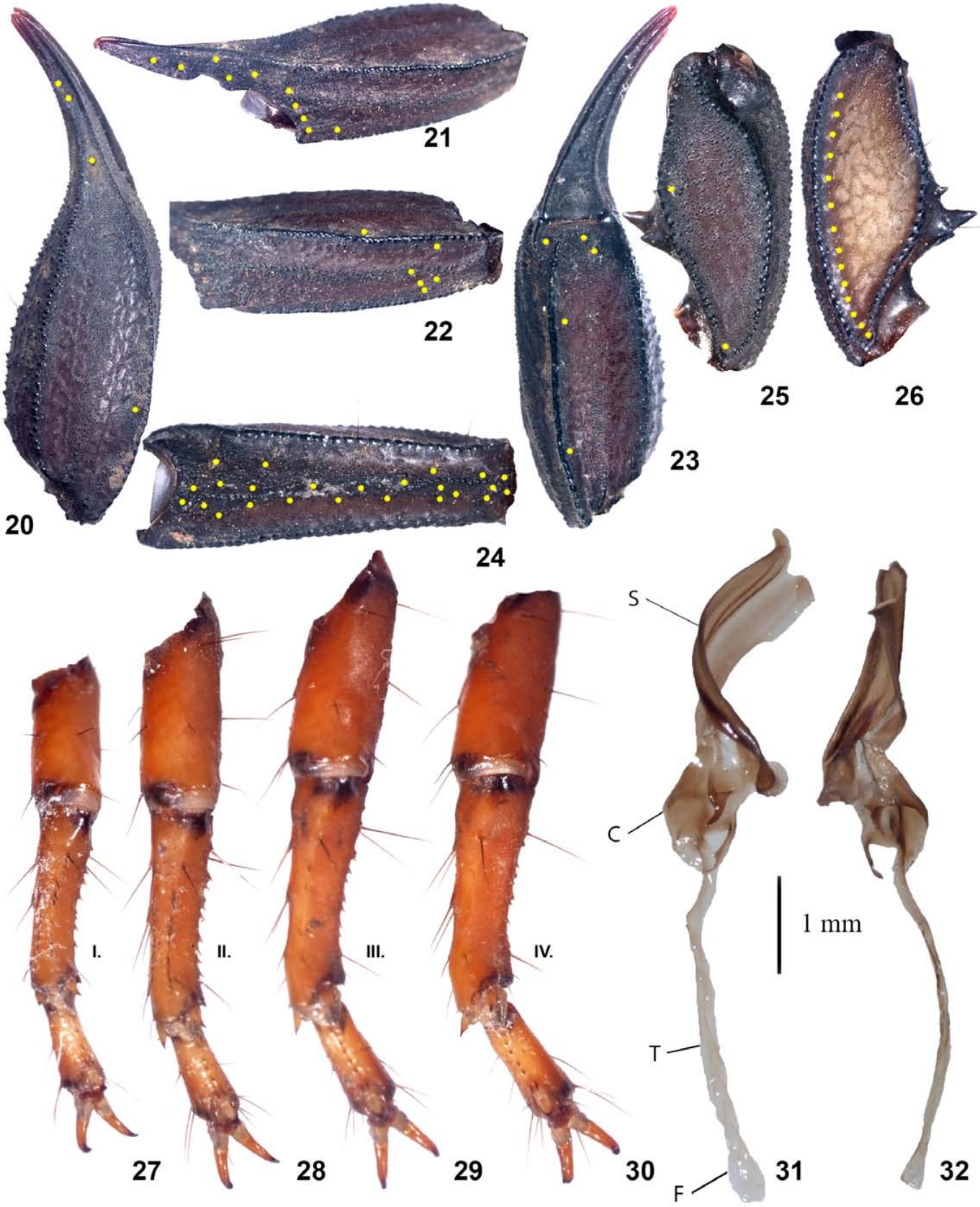
Hemispermatophore (Figs. 31–32). Lamelliform; lamina well sclerotized, curved inwardly and roundish on distal portion. Outer margin of lamina evenly notched on middle portion. Capsular portion appears to be simple and bulged. Trunk is narrow with an exterior sclerotized margin and capped with a ridge. Pedicel (foot) region short and transparent. Pedicel 0.49 mm long and 0.36 mm wide; stem 2.85 mm long and 0.22 mm wide; capsule 1.48 mm long and 0.57 mm wide; stalk 2.39 mm long and 0.78 mm wide.

Sexual dimorphism. Male genital operculum partially exposed on posterior portion, from which a pair of small genital papillae is seen. In females, the genital operculum is separated with a median suture covering the female genital opening. Movable finger of chela in males with a curved scallop on the internal margin (Figs 4, 11).

Affinities. *Scorpiops telbaila* **sp**. **n**. (which has trichobothrial counts on patella: 23–27 external, 14–16 ventral) differs from all other species of *Scorpiops* from India (which have a range of trichobothrial counts on patella 22–29 external, 12–19 ventral) by a raw genetic divergence of 5.4-14.1 % (Table 3). It is also distinguished from its congeners based on the following key of morphological characters



Figures 17–19. *Scorpiops telbaila* **sp. n.**, male, holotype, anterior margin of carapace (17), metasoma I-IV lateral (18), and pedipalp patella dorsal view with internal aspect tubercles (19) under UV light.



Figures 20–32: *Scorpiops telbaila* **sp**. **n**. **Figures 20–26**. Male, holotype, pedipalp chela dorsal (20), dorsoexternal (21, 22) and ventral (23), patella external (24), dorsal (25), and ventral (26) views. Trichobothrial pattern indicated by yellow circles. **Figures 27–30**. Left legs I-IV, retrolateral aspect. **Figures 31–32**. Male paratype, INHER-223, right hemispermatophore internal (31) and external (32) views. Abbrevations: F, foot/pedicel; T, trunk; C, capsule; S, stalk/lamina. Scale bar: 1 mm (31–32).

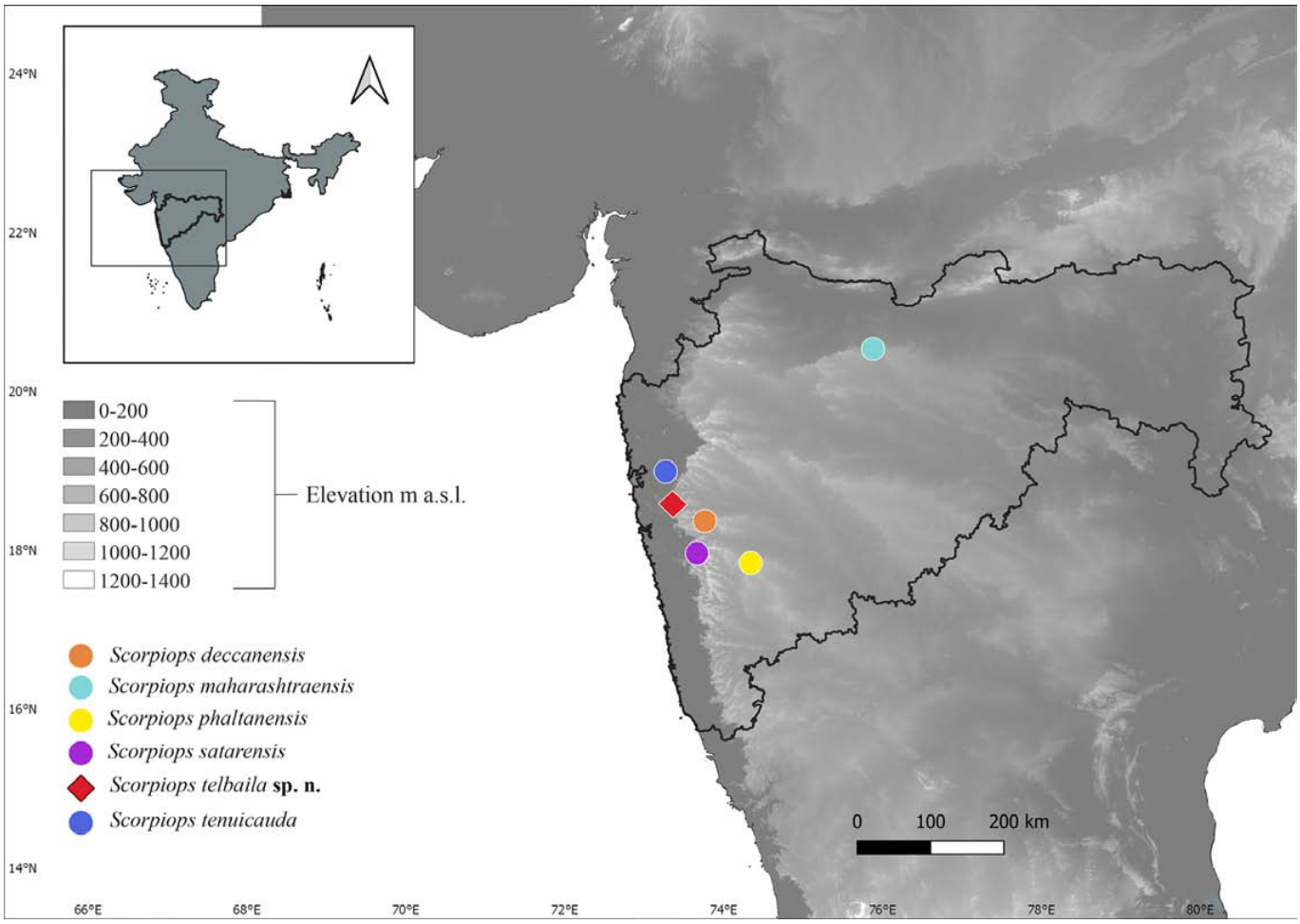


Figure 33. Distribution of *Scorpiops* species from Maharashtra (India).

Key to *Scorpiops* species from northern Western Ghats and northern Maharashtra, India.

1. Trichobothria on patella ventral 15–19. 2
– Trichobothria on patella ventral 12–16. 4

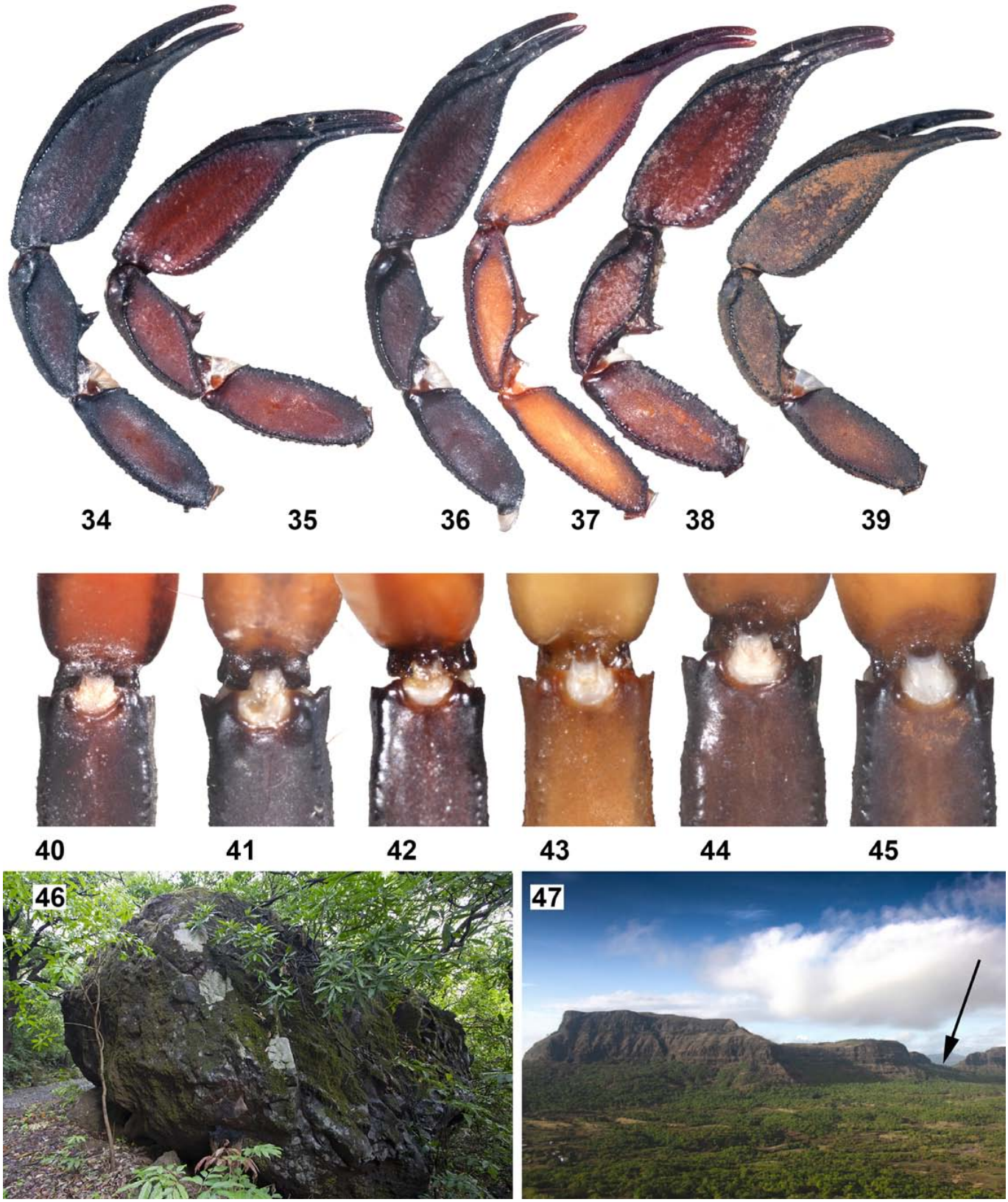
2. Trichobothria on patella ventral 17–19; anterio-ventral tubercle on interior surface of patella medium to almost equal compared to posterior-ventral tubercle. .... ***S. phaltanensis*** (Sulakhe et al., 2020)
– Trichobothria on patella ventral 15–17; anterio-ventral tubercle on interior surface of patella very small compared to posterior-ventral tubercle. 3

3. Carapace with deep U shaped emargination. ....***S. deccanensis*** Tikader & Bastawade, 1977
– Carapace with moderate U shaped emargination. .... ***S. maharashtraensis*** (Mirza et al., 2013)

4. Trichobothria on patella ventral 12–14; a pair of dorsolateral granules of anal rim strongly tuberculate. .... ***S. satarensis*** Pocock, 1900
– Trichobothria on patella ventral 14–16; a pair of dorsolateral granules of anal rim weakly tuberculate. 5

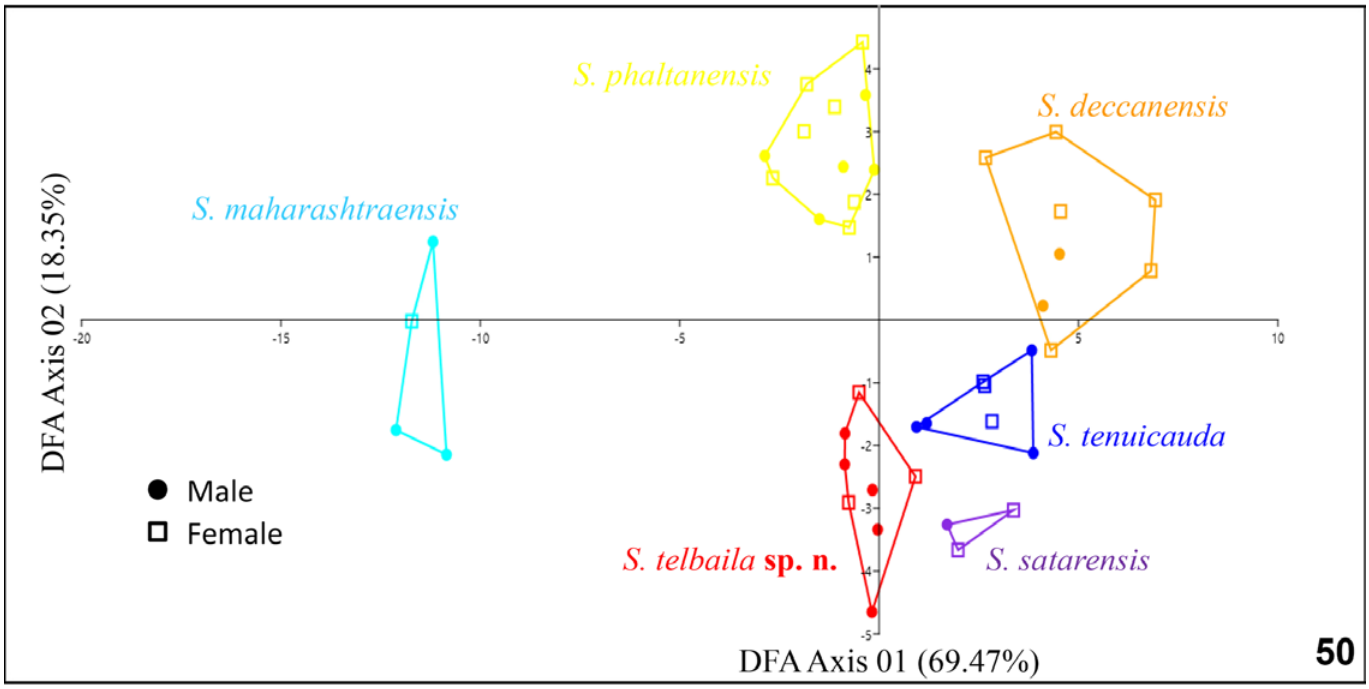
5. Anterior margin of carapace with strong tuberculate granulation; telson with annular ring at juncture between vesicle and aculeus. ..... ***S. telbaila* sp. n.**
– Anterior margin of carapace with moderate granulation; telson without an annular ring at juncture between vesicle and aculeus. ..... ***tenuicauda*** Pocock, 1894

Distribution and ecology. Presently, *S. telbaila* **sp. n.** is only known from its type locality, Saltar Khind Pass, near Ambawne Village, Pune District, Maharashtra State, India. The specimens were observed in the rock crevices in the semi-evergreen forest patches around this pass. A few specimens were also observed in the rock crevices on the basaltic rock walls on top of Telbaila fort. The type locality (Saltar Khind) is surrounded by a mountain range with basaltic rock cliffs where the species may be occurring, however this needs to be confirmed with further surveys. Individuals were found to be active at night sitting at the openings of very narrow rock crevices on the edges of the road and also on the boulders inside the thick forest. Individuals were less active during rains, mostly hiding inside the crevices, and ambushing for prey with pedipalps projecting outside the crevices during the dry time. The ecology and morphology of the new species is congruent with the lithophilic scorpions. Co-occurring scorpions observed at the type locality of *S. telbaila*



Figures 34–47: **Figures 34–39**: Comparison of left pedipalps. **Figures 40–45**: Comparison of anal rim of metasoma V in dorsal view. **Figures 34**, **40**. *Scorpiops telbaila* **sp**. **n**., male, holotype. **Figures 35**, **41**. *S. phaltanensis*, male, holotype. **Figures 36**, **42**. *S. deccanensis*, male, INHER-SC-86. **Figures 37**, **43**. *S. maharashtraensis*, male, INHER-SC-180. **Figures 38**, **44**. *S. satarensis*, male, INHER-SC-213. **Figures 39**, **45**. *S. tenuicauda*, male, INHER-SC-214. **Figures 46–47**. Type locality of *S. telbaila* **sp**. **n**., basaltic boulder inside the dense semi-evergreen forest from where the type series is collected (46), view of Saltar khind (pass) and surrounding mountain range, exact locality of collection marked with black arrow on the image (47).

Figures 48-49: **Figure 48**. View of the Telbaila fort after which the new species is named **Figure 49**. *Scorpiops telbaila* **sp**. **n**. on the Telbaila fort under UV light (specimen not collected).



51

Scorpiops telbaila **sp. n.**

Figures 50-51: **Figure 50**. Discriminant function analyses projection on first two factor planes explaining 87.82% of variation among the five species. **Figure 51**. Maximum Likelihood phylogenetic tree (ML) for *Scorpiops*. Values along the nodes are bootstraps for 1000 iterations.

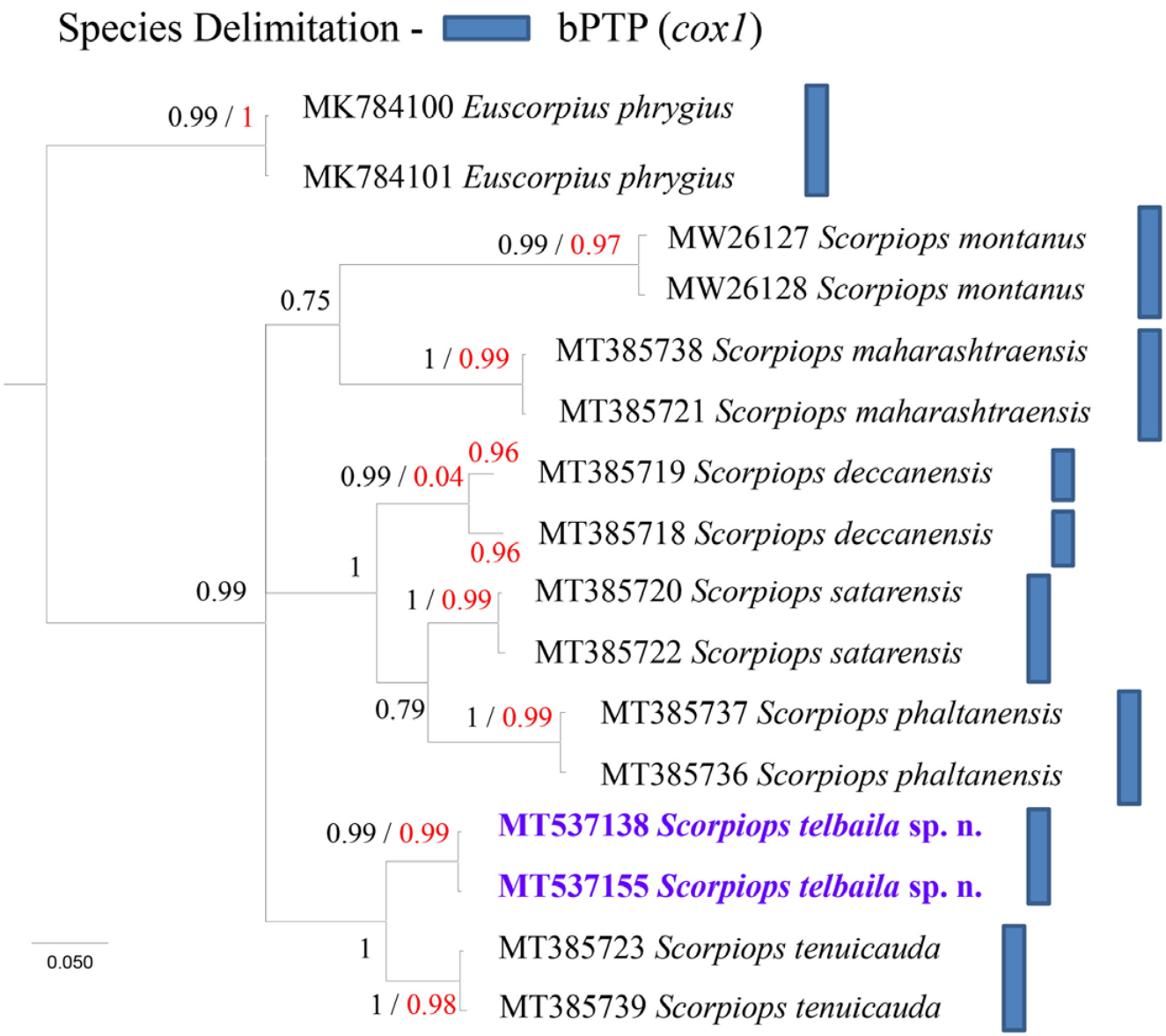


Figure 52. Bayesian phylogenetic tree for *Scorpiops*. Values along the nodes are Bayesian posterior probabilities for Bayesian Inference and Bayesian Poisson Tree Process (bPTP) respectively.

sp. **n**. were *Hottentota* sp., *Isometrus tamhini* Sulakhe et al., 2020 and *Hetrometrus* spp. (Figs. 46–49).

Statistical Analysis

Size corrected morphometric data was not significantly different from multivariate normal (Doornik and Hansen omnibus, within group Ep = 145.8, P <0.0001). First five PCA factors with eigenvalues more than 1.0 explained 90.37% of variation among the species. The DFA using all the PCA factors as input resulted in 100% individuals being classified into their respective species. The four discriminant functions with eigenvalues greater than 1.0 explained 97.54% of variation among these species, all the species formed distinct clusters on the factor plane using the first two DFA axes (Fig 50) (PCA data available from the authors).

Molecular Analysis

Maximum Likelihood and Bayesian analysis generated trees with minor differences in topologies. The two sequences of *Scorpiops telbaila* **sp**. **n**. clustered together (ML bootstrap values=99, BI posterior probabilities=0.99) and were recovered sister to *S. tenuicauda* (ML bootstrap values=94, BI posterior probabilities=1) (Figs. 51, 52). Nevertheless *S. deccanensis* was recovered as sister to *S. satarensis* in ML tree (Fig. 51) whereas *S. satarensis* was recovered sister to *S. phaltanensis* in BI tree (Fig. 52) although the relationship was not very well supported (ML bootstrap values=37, BI posterior probabilities=0.79). Interestingly *S. montanus* was recovered as sister to *S. maharashtraensis* in ML tree as well as BI tree with moderate support values (ML bootstrap values=56, BI posterior probabilities=0.75).

Primers- Cytochrome *c* Oxydaise I	5' –3' Primer Sequence	Source
HCO2198	TAAACTTCAGGGTGACCAAAAAATCA	Folmer et al. (1994)
HCOoutout	GTAAATATATGRTGDGCTC	Folmer et al. (1994)
LCO1490	GGTCAACAAATCATAAAGATATTGG	Folmer et al. (1994)
Nancy	CCCGGTAAAATTAAAATATAAACTTC	Simon et al. (1994)
Chelicerate F1	TACTCTACTAATCATAAAGACATTGG	Barrett & Hebert (2005)
Chelicerate R1	CCTCCTCCTGAAGGGTCAAAAAATGA	Barrett & Hebert (2005)
Chelicerate R2	GGATGGCCAAAAAATCAAAATAAATG	Barrett & Hebert (2005)

Table 2. Primers used for PCR amplification and sequencing of (COI) mitochondrial gene.

Species	SD	ST	SMH	SS	SP	STB	SM	EP
S. deccanensis (SD)	(0-2.1)							
S. tenuicauda (ST)	11.2-11.8	(0-0.2)						
S. maharashtrensis (SMH)	11.9-12.1	11.8-11.9	(0)					
S. satarensis (SS)	6.9-7.7	11.6-11.9	12.7-12.9	(0-0.2)				
S. phaltanensis (SP)	8.9-9.1	11.4-11.8	12.1-12.3	6.4-6.7	(0-0.2)			
S. telbaila **sp. n.** (STB)	10.6-11.2	5.4-5.6	11.2	10.8-11.0	10.8-11.0	(0)		
S. montanus (SM)	13.9-14.3	13.3-13.7	11.9-12.1	12.1-12.5	13.5-13.9	13.9-14.1	(0-0.4)	
Euscorpius phrygius (EP)	14.1-14.8	14.6-14.8	15.2	15.8-16.0	15.0-15.2	14.5	17.3-17.5	(0)

Table 3. Pairwise uncorrected raw distances (%) expressed as minimum–maximum based on COI gene sequence for *Scorpiops* species. Values in brackets are intra-clade distances.

Species	Voucher	GeneBank Accession Number	Source
Scorpiops telbaila **sp. n.**	INHER 123*	MT537138	Type material
Scorpiops telbaila **sp. n.**	INHER 126*	MT537155	Type material
S. phaltanensis	INHER 220	MT385736	Sulakhe et al. (2020b)
S. phaltanensis	INHER 221	MT385737	Sulakhe et al. (2020b)
S. deccanensis	INHER 85	MT385719	Sulakhe et al. (2020b)
S. deccanensis	INHER 86	MT385718	Sulakhe et al. (2020b)
S. tenuicauda	INHER 101	MT385723	Sulakhe et al. (2020b)
S. tenuicauda	INHER 102	MT385739	Sulakhe et al. (2020b)
S. maharashtraensis	INHER 179	MT385738	Sulakhe et al. (2020b)
S. maharashtraensis	INHER 180	MT385721	Sulakhe et al. (2020b)
S. satarensis	INHER 211	MT385722	Sulakhe et al. (2020b)
S. satarensis	INHER 213	MT385720	Sulakhe et al. (2020b)
S. montanus	INHER 168*	MW266127	Comparative material
S. montanus	INHER 166*	MW266128	Comparative material

Table 4: Voucher numbers and GenBank accession numbers for the sequence data used for the phylogenetic analysis.

Scorpiops telbaila **sp. n.** differs from *S. montanus* by a raw genetic distance of 13.9-14.1 %, from *S. maharashtraensis* by 11.2%, from *S. satarensis* and *S. phaltanensis* by 10.8-11.0%, from *S. deccanensis* by 10.6-11.2% and from *S. tenuicauda* by 5.4-5.6% (Table 3).

Bayesian Poisson Tree Process (bPTP) result supported the distinctness of *Scorpiops telbaila* **sp. n.** (posterior probability 0.99) (Fig. 52).

Discussion

This study has elevated the number of species of *Scorpiops* found in the Maharashtra State, India to six. The phylogenetic analysis based on *CO1* mitochondrial gene in this study is limited to *Scorpiops* species found in Maharashtra, India, with only *S. montanus* from the Himalayas. Further molecular studies are essential to understand the interrelationships within *Scorpiops* species from India and the placement of species from Maharashtra in the larger phylogenetic tree of Indian *Scorpiops*. Considering the overall low dispersal ability of lithophilic scorpions and local endemism due to limited biotic interchange within Western Ghats (Bossuyt et al., 2004), it can be arguably predicted that there could be point endemic populations of scorpions in this genus with several cryptic species. Some scorpion studies in the past have supported this hypothesis (Gantenbein & Keightley, 2004). Recent studies from northern Western Ghats with integrated taxonomic approach and with molecular tools have revealed the same (Mirza et al., 2019; Mirza, 2020; Sulakhe et al. 2020a, 2020b, 2020c). With this high level of endemicity of fauna in Western Ghats a more localised conservation plan is necessary for its protection.

It is important to also note that all the *Scorpiops* populations in northern Western Ghats are found in unprotected or informal protected areas. *Scorpiops telbaila* **sp**. **n**. is found very close to Tamhini Wildlife Sanctuary. This emphasizes the need to draft conservation strategies considering not only protected habitats but also informal protected areas surrounded by matrix of cultivated land (Bhagwat et al., 2005).

The recent descriptions of many new species of scorpions from India is a growing evidence that India and especially the Western Ghats are a hub of scorpion diversity and endemism, nonetheless, species diversity in several genera of this region still remains poorly explored. Therefore, large-scale integrated studies based on extensive sampling and multiple specimens are still necessary to understand their existing diversity in the Western Ghats global biodiversity hotspot.

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