PENSOFT.



Phylogeny of genus *Sichuana* Shen & Yin, 2020 (Orthoptera, Tettigoniidae, Tettigoniinae) with four new species from Sichuan, China

Jun-Jie Gu^{1*}, Chengjie Zheng^{1*}, Su-Rong Jiang¹, Yanli Yue¹

1 College of Agronomy, Sichuan Agricultural University, Chengdu 611130, Sichuan, China

https://zoobank.org/1292EF4A-6967-45A5-8732-2F980482DB00

Corresponding author: Yanli Yue (14332@sicau.edu.cn)

Academic editor: Claudia Hemp ◆ Received 3 May 2023 ◆ Accepted 2 August 2023 ◆ Published 29 September 2023

Abstract

Four new species of *Sichuana* Shen & Yin, 2020 are described based on morphological comparison and molecular analysis: *S. planicercata* **sp. nov.**, *S. curvicercata* **sp. nov.**, *S. longilamina* **sp. nov.** and *S. magnicerca* **sp. nov.** Specimens showed some intraspecific variation of male tegmina and subgenital plates. The genes *COI* and 16S were used to analyze the genetic distance between species and *COI* was used to analyze the phylogenetic relationship of *Sichuana*.

Key Words

Drymadusini, genetic distance, revision, variation, veins

Introduction

The genus *Sichuana* Shen & Yin, 2020 (Tettigoniinae, Drymadusini) is endemic to Sichuan, China (Yin et al. 2020). It is comprised of two species, *S. cryptospina* Shen & Yin, 2020 and *S. feicui* He, 2020, in Wenchuan County and Mao County of western Sichuan (He et al. 2020). It is characterized by the following character states: lateral carina distinct in the metazona but faintly indicated on the prozona; prosternum with a pair of spines; female tegmina slightly longer than the length of the pronotum; male cerci strongly incurved in the middle, the apices are acute, and there is an inner tooth in the basal area of the male cerci.

After morphological comparison, we revised the diagnosis of *Sichuana* and described four new species: *S. planicercata* sp. nov., *S. curvicercata* sp. nov., *S. longilamina* sp. nov. and *S. magnicerca* sp. nov. We analyzed the genetic distances among species using the mitochondrial genes *COI* and 16S and did a phylogenetic analysis of the genus based on *COI*.

Materials and methods

Specimens and equipment

The specimens were collected in western Sichuan, China. They (including holotypes) are deposited in the collection of the Department of Plant Protection of Sichuan Agricultural University (**SICAU**), Chengdu, China (Jun-Jie Gu, Curator).

Photographs were taken using a SZX16 microscope system (Olympus, Tokyo, Japan), or a Cannon D550 with a 50 mm lens, composing by cellSens Dimension 3.2 software. The images were digitally stacked composites of approximately 20 focal planes using Helicon Focus 6 (http://www.heliconsoft.com accessed on 12 May 2022).

Anatomical abbreviations

Wing venation nomenclature is based on the interpretation of Béthoux and Nel (2002) and Chivers et al. (2017): CuA, anterior cubitus; CuP, posterior cubitus; CuPaa, an-

Copyright Jun-Jie Gu et al. This is an open access article distributed under the terms of the Creative Commons Attribution License (CC BY 4.0), which permits unrestricted use, distribution, and reproduction in any medium, provided the original author and source are credited.

^{*} These authors contributed equally to this work.

terior branch of first posterior cubitus; CuPa β , posterior branch of first posterior cubitus; CuPb, second posterior cubitus; MA, anterior media; MP, posterior media; RA, anterior radius; RP, posterior radius; ScA, anterior subcosta; ScP, posterior subcosta. "Handle" describes the strong crossvein appearing as a main vein between the origin of CuA + CuPa α and CuPa β .

DNA extraction and amplification.

Genomic DNA was extracted from the muscles of one hind leg using a CWBIO Universal Genomic DNA Kit by the manufacturer's instructions. The molecular markers selected for this paper were mitochondrial cytochromec o xidase subunit I gene (*COI*) and the mitochondrial large-subunit rRNA gene (16S). The primers used are shown in Table 1 (Xiong and Kocher 1991; Pan et al. 2006). GenBank accession numbers are showed in Table 2.

Table 1. Primer sequences.

Target genes	Sequences				
COI	COBL TYTCAACAAAYCAY AARGATATTGG				
	COBU TAAACTTCWGGRTGWCCAAARAATCA				
16S	16Sa CGCCTGTTTATCAAAAACAT				
	16Sb CTCCGGTTTGAACTCAGATCA				

The *COI* and 16S sequences isolated from all samples were used for phylogenetic assay (Table 2) (Yin et al. 2020; Liu et al. 2018). All fragments were trimmed using Seqmen in DNAstar (Echeverry et al. 2017), and were aligned in MEGA11 by ClustalW (Tamura et al. 2021).

Genetic distance analysis

COI and 16S were used for this analysis (Table 2). Estimation of genetic distance was conducted by MAGE11.

Table 2. GenBank accession number.

Taxon	COI	168	Reference
Sichuana magnicerca	OQ799533	OQ801125	this study
sp. nov. 1			
S. magnicerca sp. nov. 2	OQ799537	OQ801126	this study
S. magnicerca sp. nov. 3	OQ799534	OQ801127	this study
S. magnicerca sp. nov. 4	OQ799532	OQ801128	this study
S. magnicerca sp. nov. 5	_	OQ801129	this study
S. longilamina sp. nov.	OQ799531	OQ801133	this study
S. feicui	OQ799536	OQ801124	this study
S. planicercata sp. nov. 1	OQ799525	OQ801130	this study
S. planicercata sp. nov. 2	OQ799526	OQ801131	this study
S. planicercata sp. nov. 3	OQ799527	OQ801132	this study
S. planicercata sp. nov.4	OQ799528	-	this study
S. curvicercata sp. nov.1	OQ799529	OQ801121	this study
S. curvicercata sp. nov.2	OQ799530	OQ801122	this study
S. curvicercata sp. nov. 3	-	OQ801123	this study
S. cryptospina 1	MT161464	-	Yin et al. 2020
S. cryptospina 2	MT161465	_	Yin et al. 2020
Mongolodectes huangxinleii	MT161460	-	Yin et al. 2020
Atlanticus fengyangensis	MG787210	_	Liu et al. 2018

We selected "compute pairwise distances" for comparing *Sichuana* species and used the "bootstrap method" with 1000 bootstrap replicates in the variance estimation method, then analyzed the data by the Kimura-2-parameter model.

Molecular phylogenetic analysis

COI was used for this analysis (Table 2). *M. huangxinleii* Liu, Xu & Zhang, 2015 and *A. fengyangensis* Liu, 2013 were used as outgroups. In all three datasets, the species of *Sichuana* Yin & Shen, 2020 were treated as ingroups. Phylogenetic analysis was conducted using MrBayes 3.2.7 (Ronquist et al. 2012) for Bayesian inference (BI) and IQ-tree 1.6.2 (Nguyen et al. 2015) for maximum-like-lihood (ML). In the BI analysis, four chains were run for 1,000,000 generations, sampling every 1,000 generations and taking the first 25% of the trees as burn-in. In the ML analysis, the best model obtained by IQ-tree1.6.2 was TVM+F+I, a total of 10000 ultrafast bootstrap replicates were executed before a thorough ML search.

Species distribution map

The satellite maps were sourced from the National Platform for Common Geospatial Information Services (https://www.tianditu.gov.cn/) and edited with ArcGIS 10.8 (Esri 2011). The distribution of *Sichuana* species is shown in Fig. 1.

Results

Systematics

Orthoptera: Tettigoniidae: Tettigoniinae: Drymadusini

Genus Sichuana Shen & Yin, 2020

Emended diagnosis. Differs from all other Drymadusini genera in China by its male cercus being strongly incurved at or after its middle with acute apex, with a basal inner tooth (Figs 2, 6, 9, 12); median carina faintly indicated in prozona and absent in metazona; male tegmen about twice as long as pronotum; and female tegmen equal to or slightly longer than pronotum. Furthermore, Sichuana differs from Uvarovina Ramme, 1939, Ptosoproctus Shen, Yin & He, 2021, and Eulithoxenus Bey-Bienko, 1951 by its prosternum with a pair of spines (Figs 4, 8, 11, 14). Differs from Atlanticus Scudder, 1894, Eulithoxenus, Ptosoproctus, and Uvarovina by its ovipositor being decurved (Figs 2, 6, 9, 12). Differs from Atlanticus, Eulithoxenus, Mongolodectes Bey-Bienko, 1951, Paratlanticus Ramme, 1939, Ptosoproctus, and Uvarovina by its lateral carina being distinct in metazona and faintly indicated on prozona (Figs 2, 6, 9, 12). Differs from Kansua Uvarov, 1933, Paratlanticus, Ceraeocercus Uvarov, 1910 by dorsal side of protibia only



🔲 S. magnicerca sp. nov. 🔲 S. cryptospina 📃 S. longilamina sp. nov. 🔵 S. planicercata sp. nov. 🌑 S. curvicercata sp. nov. 🛆 S. feicui

Figure 1. A. Distribution of *Sichuana* species in Sichuan Province, China; B, C. Enlarged version of *Sichuana* species distribution; B. *S. planicercata* sp. nov. and *S. curvicercata* sp. nov.; C. *S. longilamina* sp. nov. and *S. magnicerca* sp. nov. (circles, squares and triangles represent the three clades on the phylogenetic tree).

with external spurs, and dorsal side of mesotibia with spurs on both sides. Differs from *Kansua* Uvarov, 1933 by its smooth pronotum (Heller and Liu 2016; Liu 2013; Liu 2015; Liu et al. 2019; Shen et al. 2021).

Type species. Sichuana cryptospina Shen & Yin, 2020.

Sichuana planicercata Gu, Zheng & Yue, sp. nov.

https://zoobank.org/B79B73BD-FB84-485C-B8CA-B4A8E54FAAE4

Material examined. *Holotype:* \mathcal{J} , Xiaojin County, Ngawa Tibetan and Qiang Autonomous Prefecture, Sichuan Province, China, (30°59'31"N, 102°21'39"E, alt., ca. 2700 m), coll. Cheng-Jie Zheng and Yuan Wei, VIII-2022. *Paratypes*:10 \mathcal{J} 5 \mathcal{Q} , same data as in holotype.

Diagnosis. Differs from all other *Sichuana* species by its male tenth abdominal tergite with a pair of very short round projections at posterior margin (Fig. 2F); male cercus without distinct curve upward or downward (Fig. 2E, F, H), inner tooth far above the top of cercus in lateral view (Fig. 2 H); and lacuna of female tenth abdominal tergite rounded and deep, reaching to or near posterior margin of ninth abdominal tergite (Fig. 2G). The related species *S. curvicercata* sp. nov. with narrower lateral field of male tegmen and simple male tenth abdominal tergite, thus being similar to *S. planicercata* sp. nov. (Fig. 6I, F).

Etymology. The specific epithet is derived from a combination of the Latin '*plani*' meaning flat, and '*cercus*', describing the male cerci not bent ventrally or dorsally. Chinese name: 平尾川螽.

Measurements (mm). Body (head to tip of abdomen): 32.4–33.86♂, 34.22–35.83♀; pronotum: 8.26–8.98♂, 7.86–8.46♀; tegmen: 15.39–16.21♂, 7.96–9.35♀; mirror

of right tegmen (from fore to hind): 4.23-4.423; hind wing: 6.12-6.903, 5.43-5.762; protibia: 8.86-9.363, 8.85-10.312; profemur: 7.68-8.043, 7.92-8.532; mesotibia: 9.18-10.583, 9.96-11.282; mesofemur: 8.47-9.013, 8.48-9.4222; metatibia: 21.09-22.713, 22.39-23.2422; metafemur: 21.61-22.713; 22.98-23.7622; ovipositor: 20.82-21.43.

Description. Male. Body, large. Frons flat, slightly oblique. Frontal fastigium and clypeofrontal sulcus black. Face light-colored. Occiput convex. Vertical fastigium broad, slightly wider than scape. Median ocellus visible. Compound eyes broadly round and bulging outwards, surrounded by black coloration that extends backward to form a band. Antennae inserted at the inner sides of the compound eyes, scapus robust, much thicker than pedicel, flagellum tapers toward the apex, covered with short setae (Fig. 2A–D).

Pronotum saddle-shaped, smooth, nearly equal to profemur in length. Disc of prozona with a broadly obtuse concavity in the middle of each side, anterior margin of pronotum slightly concave and posterior margin blunt, median carina faintly indicated in prozona, absent in metazona, lateral carina distinct in metazona, faintly indicated in prozona. Lateral lobe of pronotal length greater than depth, with a light-colored stripe along the lateral margin, sometimes not obvious, humeral sinus obvious (Fig. 2A-D). Prosternum with a pair of small cone-shaped spines (Fig. 4E). Mesosternum with a pair of triangular lobes, nearly equal in width to height. Metasternum with a pair of rounded triangular lobes, width distinctly greater than height (Fig. 4E). Thoracic auditory spiracle elongated and elliptical, partially covered by lateral lobe of pronotum.



Figure 2. A–D. Body of *Sichuana planicercata* sp. nov. A, B. Male holotype; C, D. Female paratype; A, C. Dorsal view; B, D. Lateral view; E. Male terminal abdomen with artificially unfurled cerci in dorsal view for showing inner tooth; F. Male terminal abdomen in dorsal view; G. Female terminal abdomen in dorsal view; H. Male terminal abdomen in lateral view; I. Male left tegmen in lateral view.

Tegmen approximately equal to, or slightly shorter than, twice length of pronotum, with clear longitudinal and cross veins. Tegmen folded downward along the M+CuA, flat dorsal field with a transverse lacuna in the middle. Tegmen almost the same width as the metazona disc from base to middle, then gradually narrowing in dorsal view. Lateral field of the tegmen slightly broadened (Fig. 2I). ScA weak and short, very close to anterior margin, ended at or before the middle of the anterior margin. ScP strong, with 4–6 branches. R forked to RA and RP very distally or without distinct dichotomy (Fig. 3B), sometimes distally fused with ScP then separated immediately (Fig. 3A). M+CuA separated to M and CuA after the origin of the handle, but the position of their separation is unstable. M forked to MA and MP distally (Fig. 3A–F). Stridulatory file with about 20 teeth (Fig. 4G). Mirror on right tegmen pentagonal, its length greater than its width (Fig. 3B, D, F). Hind wing rudimentary.

Legs. Prothoracic leg: genicular lobes of pro- and mesothoracic leg usually unarmed on both sides, sometimes armed with 1–2 spinules; dorsal procoxa with a long spine; profemur with 0–2 external black spinules and 1–4 internal black spinules ventrally; protibia with a slit-like tympanum on both sides; protibia with 2–3 external spurs dorsally, with 4–5 spurs on each side ventrally; protibia with an external apical spur dorsally and a pair of apical



Figure 3. A-F. Tegmina of Sichuana planicercata sp. nov. in dorsal view. A, C, E. Left tegmina; B, D, F. right tegmina.

4mm

spurs ventrally. Mesothoracic leg: mesofemur with 1-4 external black spinules and 0-2 internal black spinules ventrally; mesotibia with 2-4 external spurs and 3-4 internal spurs dorsally, with 5-6 spurs on each side ventrally; mesotibia with an internal apical spur dorsally and with a pair of apical spurs ventrally. Metathoracic leg: metafemur with sparse black spinules on each side ventrally; metatibia with a row of spines of different sizes on each side dorsally, with a row of sparse tiny spurs on each side ventrally, progressively denser toward apex; metatibia with a pair of apical spurs dorsally, with two pairs of apical spurs ventrally, one pair distinctly larger.

The apical area of the tenth abdominal tergite with a wide and pileous lacuna in the middle, covered with many tiny granular protrusions; posterior margin of tenth abdominal tergite with a shallow notch at the middle of the posterior margin, sides of which form a pair of very short round projections (Fig. 2F). Cercus conical and pileous, strongly incurved after the middle of itself, apex acute, not bent toward dorsal or ventral side. Presence of a hook-like and incurved inner tooth at base of cercus, tapering and curving from base to apex; inner teeth far

above top of cercus in lateral view (Fig. 2E, F, H). Subgenital plate length greater than width, slightly beyond cercus; with lateral carinae, middle part of posterior margin with a deep notch; width of notch varies; stylus slender and longer than notch (Fig. 4A–C). Epiproct triangular. Titillator L-shaped, with 2-3 rows of larger denticles, gradually decreasing from base to apex in apical portion (Fig. 4F).

Female. Generally similar to male, but body slightly larger. Tegmen slightly longer or shorter than pronotum, extending to the third abdominal tergum (Fig. 2B, D). Hindwing micropterous, longer than half of pronotum. Cercus conical and pileous. Tenth abdominal tergite with a wide rounded deep lacuna in the middle, reaching to or near posterior margin of ninth abdominal tergite (Fig. 2G). Subgenital plate nearly trapezoid, its width greater than length, middle of posterior margin with a wide notch (Fig. 4D). Ovipositor slightly shorter than metafemur, straight or slightly decurved distally (Fig. 2B, D).

Remarks. S. planicercata sp. nov., can be assigned to Sichuana Shen & Yin, 2020 by its median carina faintly indicated in the prozona and absent in the metazona;

4mm



Figure 4. *Sichuana planicercata* sp. nov. A–C. Male subgenital plate; D. Female subgenital plate; E. Thorax in ventral view; F. Titillator; G. Stridulatory file on underside of male left tegmen.

lateral carina distinct in metazona, faintly indicated in prozona; prosternum with a pair of spines; male tegmina mesopterous, far exceeding pronotum; male cerci conical, strongly incurved at middle, apex acute, and with an inner tooth placed in basal area.

S. planicercata sp. nov. is similar to *S. curvicercata* sp. nov., but differs distinctly by: male cerci not bending ventrally or dorsally, while those of *S. curvicercata* sp. nov. are curved ventrally with apex pointing dorsally; a hook-like inner tooth that tapers and curves from base to apex, extending far above the top of cerci in lateral view, while *S. curvicercata* sp. nov. having almost the same thickness overall and suddenly sharp and incurved at apex, slightly above the top of the cerci in lateral view; the posterior margin of the male tenth abdominal tergite has a shallow notch at the middle of the posterior margin, and its sides form a pair of very short round projections, while that of *S. curvicercata* sp. nov. is without protrusion, only a wide and shallow notch at the middle (Figs 2E, F, 6E, F); denticles on the apical portion of the titillator of *S.*

planicercata sp. nov. are fewer and sparser than those of *S. curvicercata* sp. nov. and are relatively larger (Figs 4F, 8F); *S. planicercata* sp. nov. has slightly more stridulatory teeth than *S. curvicercata* sp. nov., and the spacing of the teeth of *S. planicercata* sp. nov. is slightly narrower than that of *S. curvicercata* sp. nov. (Figs 4G, 8G); and female tenth abdominal tergite has a wide rounded deep lacuna in the middle that reaches the posterior margin of the ninth abdominal tergite, while that of *S. curvicercata* sp. nov. has a wide trapezoidal projection at the posterior margin (Figs 2G, 6G).

S. planicercata sp. nov. differs from *S. feicui* He, 2020 and *S. cryptospina* Shen & Yin, 2020 by the following character states: the lateral field of the male tegmina is only slightly broadened; in the male tegmina, M+CuA is separated to M and CuA after the origin of the handle; the male cerci are strongly incurved after their middle; a hook-like inner tooth is far above the top of the cerci in lateral view; and the pair of projections at the posterior margin of the male's tenth abdominal tergite is very short

and inconspicuous. These two species also differ from *S. planicercata* sp. nov. in the shape of the inner teeth, the denticles on the titillator, the morphology of the female tenth abdominal tergite, the shape of the stridulatory file, and the number of stridulatory teeth.

Sichuana curvicercata Gu, Zheng & Yue, sp. nov. https://zoobank.org/EA8A23D6-8FDD-46B0-9CC0-C6FF514353C6

Material examined. *Holotype*: \Diamond , Yonghong village, Dawei town, Xiaojin County, Ngawa Tibetan and Qiang Autonomous Prefecture, Sichuan Province, China (30°58'6"N, 102°38'16"E, alt., ca. 2800 m), coll. Cheng-Jie Zhengand Yuan Wei, VIII-2022. Paratypes: 10 \Diamond 1 \bigcirc , same data as in holotype (Fig. 5).

Diagnosis. Differs from all other *Sichuana* species by male tenth abdominal tergite without projections at posterior margin (Fig. 6F); male cercus gradually curved ventrally with apex pointing dorsally (Fig. 6E, F, H), inner tooth thick and nearly straight, suddenly sharp and incurved at apex (Fig. 6E); female tenth abdominal tergite with a wide trapezoidal projection at posterior margin (Fig. 6G).

Etymology. The specific epithet is derived from a combination of the Latin '*curvi*' meaning curved and 'cercus', describing the male cerci curved ventrally with the apex pointing dorsally. Chinese name: 弯尾川螽.

Measurements (mm). Body (head to tip of abdomen): 25.36-26.77, 27.28, pronotum: 6.98-7.26, 7.44; tegmen: 11.70-12.42, 7.08; mirror of right tegmen (from fore to hind): 3.88-3.96; hind wing: 4.39-4.62, 4.79; protibia: 6.24-7.23, 7.6; profemur: 6.45-6.86, 7.28; mesotibia: 7.24-7.72, 9.32; mesofemur: 6.26-7.10, 8.16; metatibia: 16.33-17.19, 19.92; metafemur: 17.56-17.8; 20.96; ovipositor: 17.64.

Description. Male. *Body* size medium. Frons flat, slightly oblique. Frontal fastigium and clypeofrontal sulcus black. Face light-colored. Occiput convex. Vertical fastigium broad, slightly wider than scape. Median ocellus visible. Compound eyes broadly round and bulging outwards, surrounded by black coloration that extends backward to form a black band. Filiform antennae inserted at the inner side of the compound eyes, scapus robust, much thicker than pedicel, flagellum taper toward the apex, covered with short setae (Fig. 6A–D).

Pronotum saddle-shaped, smooth, nearly equal to profemur in length. Disc of prozona with a broadly obtuse concavity in the middle of each side, anterior margin of pronotum slightly concave and posterior margin blunt, median carina faintly indicated in prozona, absent in metazona, lateral carina distinct in metazona, faintly indicated in prozona. Lateral lobe of pronotal length greater than depth, with a light-colored stripe along the lateral margin, sometimes not obvious, humeral sinus obvious (Fig. 6A– D). Prosternum with a pair of small cone-shaped spines



Figure 5. Sichuana curvicercata sp. nov. A. Adult female; B-D. Adult male.



Figure 6. A–D. Body of *Sichuana curvicercata* sp. nov. A, B. Male holotype; C, D. Female paratype; A, C. Dorsal view; B, D. Lateral view; E. Male terminal abdomen with artificially unfurled cerci in dorsal view for showing inner tooth; F. Male terminal abdomen in dorsal view; G. Female terminal abdomen in dorsal view; H. Male terminal abdomen in lateral view; I. Male left tegmen in lateral view.

(Fig. 8E). Mesosternum with a pair of triangular lobes, nearly equal in width to height. Metasternum with a pair of rounded triangular lobes, width distinctly greater than height (Fig. 8E). Thoracic auditory spiracle elongated and elliptical, partially covered by lateral lobe of pronotum.

Tegmen approximately equal to or slightly shorter than twice length of pronotum, with clear longitudinal and cross veins. Tegmen folded downward along M+CuA, dorsal field flat, with a transverse lacuna in the middle. Tegmen almost the same width with disc of metazona from basic until middle, and then gradually narrowing in dorsal view. Lateral field of tegmen slightly broadened (Fig. 6I). ScA weak and short, very close to anterior margin, ending at or before middle of anterior margin. ScP strong, with 5–7 branches. R without distinct dichotomy. M+CuA separated into M and CuA after intersection of handle and M+CuA, slightly after middle of tegmen. M separated into MA and MP after origin of handle but the position of their separation is unstable (Fig. 7A–F). Stridulatory file with about 18 teeth (Fig. 8G). Mirror on right tegmina pentagonal, length greater than width (Fig. 7B, D, F). Hind wing rudimentary.

Legs. Prothoracic leg: genicular lobes armed with 1–2 internal spinules and externally unarmed. Dorsal side of procoxa with a long spine. Profemur with 2–4 internal black spinules ventrally; protibia with a slit-like auditory tympanum on both sides; protibia with 0–3 external spurs dorsally, with 4–5 spurs on each side ventrally; protibia with an external apical spur dorsally and with a pair of apical spurs ventrally. Mesothoracic leg:



Figure 7. A-F. Tegmina of Sichuana curvicercata sp. nov. in dorsal view. A, C, E. Left tegmina; B, D, F. Right tegmina.

genicular lobes armed with 0-2 spinules on each side. Mesofemur with 2–4 external black spinules and 0-2 internal black spinules ventrally; mesotibia with 2–4 external spurs and 3–4 internal spurs dorsally, with 5–6 spurs on each side ventrally; mesotibia with an internal apical spur ventrally and a pair of apical spurs dorsally. Metathoracic leg: genicular lobes unarmed; metafemur with sparse black spinules on each side ventrally; metatibia with a row of spines of different sizes on each side ventrally, progressively denser toward apex; metatibia with a pair of apical spurs dorsally and two pairs ventrally, one of which is distinctly larger.

The apical area of the tenth abdominal tergite with a wide and pileous lacuna in the middle, covered with many tiny granular protrusions. The tenth abdominal tergite with a wide and shallow notch in the middle of the posterior margin (Fig. 6F). Cercus conical and pileous, strongly incurved at middle; cercus curved ventrally after the middle, with the apex pointing dorsally. With a thick and nearly straight inner tooth at the base of the cercus, almost the same thickness overall, its apex abruptly thin and incurved; inner tooth slightly above the top of cercus in lateral view (Fig. 6E, F, H). Subgenital plate length greater than width, with lateral carinae, middle part of posterior margin with a deep notch, width of notch varies; stylus slender and longer than notch (Fig. 8A–C). Epiproct triangular. Titillator L-shaped, with 5–6 rows of dense denticles or more, gradually decreasing from base to apex on apical portion (Fig. 8F).

Female. Generally similar to male, but body slightly larger. Tegmen shorter than pronotum, extending to the third abdominal tergum (Fig. 6B, D). Hindwing micropterous, longer than half of pronotum. Cercus conical and pileous. The posterior margin of tenth abdominal tergite with a wide trapezoidal projection (Fig. 6G). Subgenital plate nearly trapezoid, width nearly equal to length, middle of posterior margin with a wide notch (Fig. 8D). Ovipositor slightly shorter than metafemur, slightly decurved distally (Fig. 6B, D).

Remarks. S. curvicercata sp. nov. is similar to S. planicercata sp. nov., but is distinct by: male cerci gradually



Figure 8. *Sichuana curvicercata* sp. nov. A–C. Male subgenital plate; D. Female subgenital plate; E. Thorax in ventral view; F. Titillator; G. Stridulatory file on underside of male left tegmen.

curved ventrally with the apex pointing dorsally, while those of S. planicercata sp. nov. do not bend ventrally or dorsally; the inner tooth is nearly straight and almost fo same thickness overall and is suddenly sharp and incurved at apex, while that of S. planicercata sp. nov. is tapering and curving from base to apex and is far above the top of the cerci in lateral view; the posterior margin of the male tenth abdominal tergite only with a wide and shallow notch in the middle of the posterior margin, while that of S. planicercata sp. nov. has a pair of very short and inconspicuous projections (Figs 2E, F, 6E, F); denticles on the apical portion of titillator of S. planicercata sp. nov. are fewer and sparser than those of S. curvicercata sp. nov. and are relatively larger (Figs 4F, 8F); S. curvicercata sp. nov. with slightly fewer stridulatory teeth than S. planicercata sp. nov., and the spacing of the teeth of S. curvicercata sp. nov. is slightly wider than that of S. planicercata sp. nov. (Figs 4G, 8G); female tenth abdominal tergite with a wide trapezoidal projection at the posterior margin, while that of S. planicercata sp. nov. have a wide rounded deep lacuna in the middle (Figs 2G, 6G).

S. curvicercata sp. nov. differs from S. feicui He, 2020 and S. cryptospina Shen & Yin, 2020 by: the lateral field

of the male tegmina is slightly broadened; in male tegmina, M+CuA separate to M and CuA after the origin of the handle; the posterior margin of the male tenth abdominal tergite is without projections; male cerci are curved ventrally with the apex pointing dorsally. Furthermore, *S. curvicercata* sp. nov. differs from *S. feicui* by its male cerci strongly incurved at the middle. These two species also differ from *S. curvicercata* sp. nov. in the shape of the inner teeth, the denticles on the titillator, the morphology of the female tenth abdominal tergite, the shape of the stridulatory file, and the number of stridulatory teeth.

Sichuana longilamina Gu, Zheng & Yue, sp. nov.

https://zoobank.org/0B23FBDC-D40A-4AE2-957A-982FB4F64E4D

Material examined. *Holotype*: ∂, Guergou, Li County, Ngawa Tibetan and Qiang Autonomous Prefecture, Sichuan Province, China, (31°30'29"N, 102°58'35"E, alt., ca. 2400 m), coll. Cheng-Jie Zhengand Yuan Wei, VIII-2022. *Paratypes*: 1♀, same data as in holotype.

Diagnosis. Differs from all other *Sichuana* species by notch of tenth abdominal tergite of male trapezoidal (Fig.



Figure 9. A–D. Body of *Sichuana longilamina* sp. nov. **A**, **B**. Male holotype; **C**, **D**. Female paratype; **A**, **C**. Dorsal view; **B**, **D**. Lateral view; **E**. Male terminal abdomen with artificially unfurled cerci in dorsal view for showing inner tooth; **F**. Male terminal abdomen in dorsal view; **G**. Female terminal abdomen in dorsal view; **H**. Male terminal abdomen in lateral view; **I**. Male left tegmen in lateral view.

9 F); male cercus strongly incurved at an acute angle and pointing dorsally (Fig. 9E, F, H), inner tooth pointing dorsally (Fig. 9E); apex of male subgenital plate elongate, the long styli about one-third of length of subgenital plate (Figs 9H, 11C); notch of female tenth abdominal tergite trapezoidal (Fig. 9G).

Etymology. The specific epithet is derived from a combination of the Latin 'longus' meaning long and 'lamina' meaning plate, to describe its male subgenital plate which is distinctly longer than the cerci. Chinese name:长板川螽.

Measurements (mm). Body (head to tip of abdomen): 27.7, 31.38; pronotum: 8.36, 8.6; tegmen: 17.42,

8.76 \bigcirc ; mirror of right tegmen (from fore to hind): 4.34 \Diamond ; hind wing: 7.82 \Diamond , 5.54 \bigcirc ; protibia: 7.58 \Diamond , 10 \bigcirc ; profemur: 7.4 \Diamond , 8.5 \bigcirc ; mesotibia: 9.42 \Diamond , 10.94 \bigcirc ; mesofemur: 8.52 \Diamond , 9.62 \bigcirc ; metatibia: 21.12 \Diamond , 26.28 \bigcirc ; metafemur: 20.98 \Diamond ; 25.64 \bigcirc ; ovipositor: 23.34.

Description. Male. *Body* size medium. Frons flat, slightly oblique. Frontal fastigium and clypeofrontal sulcus black. Face light-colored. Occiput convex. Vertical fastigium broad, slightly wider than scape. Median ocellus visible. Compound eye broadly round and bulging outwards, surrounded by black coloration extending backward to form a band. Filiform antennae inserted on the inner sides of the compound eyes, scapus robust,



Figure 10. A, B. Tegmina of Sichuana longilamina sp. nov. in dorsal view. A. Left tegmen; B. Right tegmen.

much thicker than pedicel, flagellum tapers toward the apex, covered with short setae (Fig. 9A–D).

Pronotum saddle-shaped, smooth, nearly equal to profemur in length. Disc of prozona with a broadly obtuse concavity in the middle of each side, anterior margin of pronotum slightly concave and posterior margin blunt, median carina faintly indicated in prozona, absent in metazona, lateral carina distinct in metazona, faintly indicated in prozona. Lateral lobe of pronotal length greater than depth, with a light-colored stripe along the lateral margin, sometimes not obvious, humeral sinus obvious (Fig. 9A-D). Prosternum with a pair of longer cone-shaped spines (Fig. 11A). Mesosternum with a pair of acute triangular lobes, height greater than width. Metasternum with a pair of rounded triangular lobes, width distinctly greater than height (Fig. 11E). Thoracic auditory spiracle elongated and elliptical, partially covered by lateral lobe of pronotum.

Tegmen slightly shorter than twice the length of pronotum, with clear longitudinal and cross veins. Tegmen folded downward along the M+CuA, the dorsal field flat, with a transverse lacuna in the middle. Tegmen almost the same width as disc of metazona from base to the middle, then gradually narrowing in dorsal view. Lateral field of the tegmen distinctly broadened (Fig. 9I). ScA weak, very close to anterior margin, ending at the middle of the anterior margin or fused with branch of ScP. ScP strong, with 3-5 branches. R forked to RA and RP distally, RA very close to ScP, sometimes distally fused with ScP (Fig. 10B). M+CuA branched to M and CuA before intersection of handle and CuA, slightly before middle of tegmen. M forked to MA and MP before the origin of the handle, but position of their separation unstable (Fig. 10A, B). Stridulatory file with about 34 teeth (Fig. 11E). Mirror on right tegmina pentagonal, length greater than width (Fig. 10B). Hind wing rudimentary.

Legs. Prothoracic leg: genicular lobes armed with 1–2 internal spinules and externally unarmed. Dorsal procoxa with a long spine. Profemur with 2–4 internal black spinules ventrally; protibia with a slit-like auditory tympanum on both sides; protibia with 2 external spurs dorsally, with 5 spurs on each side ventrally; protibia with an external apical spur dorsally and with a pair of apical

spurs ventrally. Mesothoracic leg: genicular lobes armed with 0–1 external spinule and 1 internal spinule; mesofemur with 2–3 external black spinules and 0–2 internal black spinules ventrally; mesotibia with 2 external spurs and 3 internal spurs dorsally, with 5 spurs on each side ventrally; mesotibia with an internal apical spur dorsally and with a pair of apical spurs ventrally. Metathoracic leg: genicular lobes unarmed; metafemur with sparse black spinules on each side ventrally; metatibia with a row of spines of different sizes on each side dorsally, with a row of sparse tiny spurs on each side ventrally, progressively denser toward the apex; metatibia with a pair of apical spurs dorsally, with two pairs of apical spurs ventrally, one pair distinctly larger.

The apical area of the tenth abdominal tergite with a wide lacuna in the middle covered with many tiny granular protrusions. The posterior margin of the tenth abdominal tergite with a trapezoidal notch at the middle, its sides forming a pair of round, blunt lobes (Fig. 9E, F). Cercus conical and pileous, strongly incurved at an acute angle after its middle and points dorsally, apex acute; with a hook-like, incurved inner tooth placed at base of cercus, tapering and curving from the base to the apex and pointing dorsally (Fig. 9E, F, H). Subgenital plate length greater than width, with lateral carinae, middle posterior margin with a deep notch, apex of subgenital plate elongate, far beyond cercus; stylus slender, about one-third the length of subgenital plate, longer than notch (Fig. 11C). Epiproct triangular. Titillator L-shaped, with 2-3 rows of denticles, and denticles on upper part of the apical portion distinctly larger than those on the lower part (Fig. 11D).

Female. Generally similar to male, but body slightly larger. Tegmen shorter than pronotum, extending to the third abdominal tergum (Fig. 9C, D). Hindwing micropterous, longer than half of the pronotum. Cercus conical and pileous. The apical area of the tenth abdominal tergite with a wide depression in the middle, near the posterior margin of ninth abdominal tergite. Tenth abdominal tergite with a U-shaped excision in the middle of posterior margin, sides of excision form a pair of round blunt projections (Fig. 9G). Subgenital plate nearly trapezoid, width greater than length, middle of posterior margin



Figure 11. *Sichuana longilamina* sp. nov. A. Thorax in ventral view; B. Female subgenital plate; C. Male subgenital plate; D. Titillator; E. Stridulatory file on underside of male left tegmen.

with a wide notch (Fig. 11B). Ovipositor slightly shorter than metafemur, slightly decurved distally (Fig. 9C, D).

Remarks. S. longilamina sp. nov. differs from S. feicui He, 2020, S. cryptospina Shen & Yin, 2020, S. planicercata sp. nov. and S. curvicercata sp. nov. by the following: pair of cone-shaped spines on prosternum more slender; pair of lobes on mesosternum acutely triangular, height greater than width (Fig. 11 A); male cerci strongly incurved at an acute angle slightly after their middle and gradually curved dorsally (Fig. 9E, F, H); apex of male subgenital plate extended far beyond cerci (Figs 9H, 11C); styli longer, about one-third the length of subgenital plate (Fig. 11C). Furthermore, S. longilamina sp. nov. differs from S. planicercata sp. nov. and S. curvicercata sp. nov. by: tenth abdominal tergite with a pair of round blunt projections on posterior margin; in male tegmina M+CuA branching to M and CuA before origin of handle; lateral field of male tegmina distinctly broadened.

These four species also differ from *S. longilamina* sp. nov. in the shape of the inner teeth, the denticles on the titillators, the morphology of the tenth abdominal tergite, the shape of the stridulatory file, and the number of stridulatory teeth.

Sichuana magnicerca Gu, Zheng & Yue, sp. nov. https://zoobank.org/532D02B3-2225-46C3-9DC9-2EB1918373A2

Material examined. *Holotype*: ♂, Zagunao town, Li County, Ngawa Tibetan and Qiang Autonomous Prefecture, Sichuan Province, China, (31°27'33"N, 103°10'52"E, alt., ca. 2000 m), coll. Cheng-Jie Zheng and Yuan Wei, VIII-2022. *Paratypes*: 8♂ 11♀, same data as in holotype.

Diagnosis. Differs from all other *Sichuana* species by notch of male tenth abdominal tergite U-shaped; large and long male cercus beyond subgenital plate (Fig. 12F, H), inner tooth small, inserted in the most basal part of cercus (Fig. 12E); notch of female tenth abdominal tergite V-shaped (Fig. 12G). The related species *S. cryptospina* Shen & Yin, 2020 with a pair of projections covering the inner tooth at male tenth abdominal tergite, male cercus at an obtuse angle, and broader lateral field of male tegmen, thus being similar to *S. magnicerca* sp. nov. (Fig. 12).

Etymology. The specific epithet is derived from a combination of the Latin 'magnus' meaning huge and 'cercus', to describe its male cerci, large and longer than the subgenital plate. Chinese name: 巨钩川螽.



Figure 12. A–D. Body of *Sichuana magnicerca* sp. nov. A, B. Male holotype; C, D. Female paratype; A, C. Dorsal view; B, D. Lateral view; E. Male terminal abdomen with artificially unfurled cerci in dorsal view for showing inner tooth; F. Male terminal abdomen in dorsal view; G. Female terminal abdomen in dorsal view; H. Male terminal abdomen in lateral view; I. Male left tegmen in lateral view.

Measurements (mm). Body (head to tip of abdomen): 30.78–33.02 $^{\circ}$, 35.1–37.34 $^{\circ}$; pronotum: 8.16–9.04 $^{\circ}$, 8.82–9.7 $^{\circ}$; tegmen: 15.84–17.57 $^{\circ}$, 8.14–8.92 $^{\circ}$; mirror of right tegmen (from fore to hind): 4.39–4.64 $^{\circ}$; hind wing: 8.23–8.39 $^{\circ}$, 5.28–5.78 $^{\circ}$; protibia: 8.12–9.14 $^{\circ}$, 9.5–10.58 $^{\circ}$; profemur: 7.68–8.34 $^{\circ}$, 8.04–8.98 $^{\circ}$; mesotibia: 9.42–10.12 $^{\circ}$, 10.52–11.74 $^{\circ}$; mesofemur: 8.44– 9.39 $^{\circ}$, 9.1–10.18 $^{\circ}$; metatibia: 22.24–24.43 $^{\circ}$, 24.36– 27.76 $^{\circ}$; metafemur: 23.02–24.88 $^{\circ}$; 24.48–27.54 $^{\circ}$; ovipositor: 21.48–24.39.

Description. Male. *Body* size medium. Frons flat, slightly oblique. Frontal fastigium and clypeofrontal sulcus black. Face light-colored. Occiput convex. Vertical fastigium broad, slightly wider than scape. Median ocellus visible. Compound eyes broadly round and bulging outwards, surrounded by black coloration that extending backward to form a band. Filiform antennae inserted at inner sides of the compound eyes, scapus robust, much thicker than pedicel, flagellum tapering toward apex, covered with short setae (Fig. 12A–D).

Pronotum saddle-shaped, smooth, nearly equal to profemur in length. Disc of prozona with a broadly obtuse concavity in the middle of each side, anterior margin of pronotum slightly concave and posterior margin blunt, median carina faintly indicated in prozona, absent in metazona, lateral carina distinct in metazona, faintly indicated in prozona. Lateral lobe of pronotal length greater than depth, with a light-colored stripe along the lateral



Figure 13. A-D. Tegmina of Sichuana magnicerca sp. nov. in dorsal view. A, C. Left tegmina; B, D. Right tegmina.

margin, sometimes not obvious, humeral sinus obvious (Fig. 12A–D). Prosternum with a pair of cone-shaped spines (Fig. 14). Mesosternum with a pair of triangular lobes, nearly equal in width to height. Metasternum with a pair of rounded triangular lobes, width distinctly greater than height (Fig. 14E). Thoracic auditory spiracle elongated and elliptical, partially covered by lateral lobe of pronotum.

Tegmen approximately equal to or slightly shorter than twice the length of pronotum, with clear longitudinal and cross veins. Tegmen folded downwards along M+CuA, the dorsal field flat, with a transverse lacuna in middle. Tegmen almost same width as disc of metazona from base to middle, and then gradually narrowing in dorsal view. Lateral field of tegmen distinctly broadened (Fig. 12I). ScA weak, very close to anterior margin, ending at or before middle of anterior margin. ScP strong, with 5-6 branches. R usually forked to RA and RP after middle of tegmen, in a few examples very distally (for example, Fig. 13D). M+CuA forked to M and CuA before origin of handle. M usually very close to RP (Fig. 13A, C, D), or fused with RP then separateing immediately (Fig. 13B). Stridulatory file with about 33 teeth (Fig. 14G). Mirror on right tegmen pentagonal, length greater than width (Fig. 13B, D). Hind wing rudimentary.

Legs. Prothoracic leg: genicular lobes armed with 1–2 internal spinules, unarmed externally. Dorsal surface of procoxa with a long spine; profemur with 3–5 internal black spinules ventrally; protibia with a slit-like auditory tympanum on both sides; protibia with 2–4 external spurs dorsally, with 5 spurs on each side ventrally; protibia with an external apical spur dorsally and a pair of apical spurs ventrally. Mesothoracic leg: genicular lobes armed with

1–2 spinules on each side; mesofemur with 2–3 external black spinules and 0–2 internal black spinules ventrally; mesotibia with 2–3 external spurs and 3–5 internal spurs dorsally, with 5 spurs on each side ventrally; mesotibia with an internal apical spur dorsally and a pair of apical spurs ventrally. Metathoracic leg: genicular lobes unarmed. Metafemur with sparse black spinules on each side ventrally; metatibia with a row of spines of different sizes on each side dorsally, with a row of sparse tiny spurs on each side ventrally, progressively denser toward the apex; metatibia with a pair of apical spurs dorsally, with two pairs of apical spurs ventrally, one pair of which distinctly larger than the other.

Apical area of the tenth abdominal tergite with a slight pileous lacuna covered with many tiny granular protrusions. Posterior margin of tenth abdominal tergite with U-shaped notch in middle, sides of notch which form a pair of round blunt projections (Fig. 12E, F). Cercus large and long, extending beyond subgenital plate, conical and pileous, strongly incurved after middle, apex acute and slightly upturned. With a small, hook-like and incurved inner tooth at basal-most part of the cercus, tapering and curving from base to apex, invisible in lateral view (Fig. 12E, F, H). Subgenital plate length greater than width, with lateral carinae, middle part of posterior margin with a deep notch, width of notch varying among individuals; stylus slender and longer than notch (Fig. 14A-C). Epiproct triangular. Titillator L-shaped, with only one row of denticles, gradually getting larger from base to apex on apical portion (Fig. 14F).

Female. Similar to male, but body slightly larger. Tegmen shorter than pronotum, extending to the third abdominal tergum (Fig, 12B, D). Hindwing micropterous,



Figure 14. *Sichuana magnicerca* sp. nov. A–C. Male subgenital plate; D. Female subgenital plate; E. Thorax in ventral view; F. Titillator; G. Stridulatory file on the underside of the male left tegmen.

longer than half of pronotum. Cercus conical and pileous. Tenth abdominal tergite depressed downward in middle, and with a V-shaped notch at middle of posterior margin, its sides forming a pair of round blunt projections (Fig. 12G). Subgenital plate nearly trapezoid, nearly equal in width to length, middle of posterior margin with a wide notch (Fig. 14D). Ovipositor slightly shorter than metafemur, slightly decurved distally (Fig. 12B, D).

Remarks. *S. magnicerca* sp. nov. is similar to *S. cryp-tospina* Shen & Yin, 2020, but differs distinctly by: male cerci large and long, extending beyond subgenital plate, while those of *S. cryptospina* are relatively shorter and smaller, not extending beyond subgenital plate; the inner tooth is small and placed at the basal-most part of the cerci, while that of *S. cryptospina* is relatively larger and longer, and is placed at the sub-basal area of the cerci (Fig. 12E, F, H); the apical portion of the titillator of *S. magnicerca* sp. nov. has only 1 row of denticles that are aligned

are aligned diagonally (Fig. 14F); and the female tenth abdominal tergite is depressed downward in the middle and has a V-shaped notch, while that of *S. cryptospina* has a U-shaped notch (Fig. 12G). *S. magnicerca* sp. nov. differs from *S. feicui* He, 2020, *S. planicercata* sp. nov., *S. longilamina* sp. nov. and

vertically, while that of S. cryptospina has 1–2 rows that

S. planicercata sp. nov., *S. longilamina* sp. nov. and *S. curvicercata* sp. nov. by its large and long male cerci, which extend beyond the subgenital plate. Furthermore, *S. magnicerca* sp. nov. differs from *S. planicercata* sp. nov. and *S. curvicercata* sp. nov. by: tenth abdominal tergite with a pair of round blunt projections on posterior margin; in male tegmina M+CuA branching to M and CuA before origin of handle; and lateral field of male tegmina distinctly broadened. It differs from *S. feicui* by its male cerci strongly incurved after middle. It differs from *S. longilamina* sp. nov. by male subgenital plate not extended beyond cerci, and male cerci incurved at an obtuse an-

gle. These four species also differ from *S. magnicerca* sp. nov. in shape of the inner teeth, denticles on the titillators, morphology of female tenth abdominal tergite, shape of stridulatory file, and the number of stridulatory teeth.

Genetic distanceanalysis

The mean of the sequence divergences for *COI* among species of *Sichuana* Shen & Yin, 2020 ranged from a low of 2.208% to a high of 15.688% (Table 3). The sequence divergences for *COI* between most species are greater than 8%. While only *S. planicercata* sp. nov. and *S. curvicercata* sp. nov. show low sequence divergences between them (2.208%), this is still significantly greater than the intraspecific mean of sequence divergence. Relative to *COI*, the genetic distance analysis using 16S shows lower sequence divergence at all levels (Table 4).

Molecular phylogenetic results

The results of the ML and BI analyses are almost identical (Fig. 15). *S. feicui* He, 2020 appears at the earliest position and is sister of all other *Sichuana* species. The rest consist of two clades. The clade containing *S. magnicerca* sp. nov., *S. cryptospina* Shen & Yin, 2020 and *S. longilamina* sp. nov. is sister of the clade containing *S. planicercata* sp. nov. and *S. curvicercata* sp. nov. *S. longilamina* sp. nov. is the sister group of the clade containing

 Table 3. Sequence divergences for COI among species of Sichuana.

Species	COI sequence divergences (%)					
	S. planicercata	S. magnicerca	S. longilamina	S. feicui	S. curvicercata	S. cryptospina
S. planicercata	0.586					
S. magnicerca	13.620	0.847				
S.longilamina	13.150	10.738	_			
S .feicui	15.391	15.688	14.407	-		
S. curvicercata	2.208	10.671	13.103	14.393	0.305	
S. cryptospina	13.563	8.760	11.000	17.214	12.686	0.152

 Table 4. Sequence divergences for 16S among species of Sichuana.

Species	16S sequence divergences (%)					
	S. planicercata	S. magnicerca	S. longilamina	S. feicui	S. curvicercata	
S. planicercata	0.127					
S. magnicerca	3.989	0.000				
S. longilamina	3.177	2.126	-			
S. feicui	5.828	4.945	3.719	-		
S. curvicercata	0.830	3.652	3.245	5.900	0.254	

S. magnicerca sp. nov. and *S. cryptospina*. In the ML tree, samples of *S. curvicercata* sp. nov. resolved as the sister of the clade containing *S. planicercata* sp. nov. and other samples of *S. curvicercata* sp. nov. In contrast, these two species are well resolved in the BI tree.

Discussion

Morphological variation within species

Variation in wing shape and venation is documented in various groups of fossil orthopterans and their relatives (Zessin 1987; Béthoux 2008; Gu et al. 2010, 2011, 2014, 2021). However, this is rarely addressed in extant orthoptera. The venation of Sichuana shows different degrees of intraspecific variation. For instance, the end of ScA, the dichotomous position of M, the dichotomous position of R and the fusion of some longitudinal veins are unstable among individuals, even in the left and right tegmina of an individual (Figs 3, 7, 10, 13), including structures associated with R. For instance, in S. planicercata sp. nov., R sometimes fuses distally with ScP and is separated immediately (Fig. 3A), R usually forks into RA and RP distally, but sometimes R is unbranched (Fig. 3B). In S. magnicerca sp. nov. RP sometimes fuses with M and separated immediately (Fig. 13B). M also shows significant variation between individuals, mainly in the position of its forking to MA and MP. However, the bifurcation of R and M is usually stable in fossil ensiferans (Gu et al. 2009, 2014, 2021; Wang et al. 2016). Therefore, more documentation on wing venation within species of extant orthopterans would help us better understand wing morphology and provide a reference for classification of extinct orthopterans.

The male subgenital plate of *Sichuana* also shows different degrees of variation between individuals. The shape and width of the notch on its posterior margin vary between individuals, especially in *S. planicercata* sp. nov. (Figs 4, 8, 11, 14).

Molecular analysis

Genetic distance analysis shows that sequence divergences for *COI* between most species of *Sichuana* are greater than 8% (Table 3). While the sequence divergence between *S. planicercata* sp. nov. and *S. curvicercata* sp. nov. is significantly smaller, about 2.208%, it's still significantly greater than the intraspecific mean of sequence divergence. Generally, the genetic distance within a species is much smaller than between species (Hebert et al. 2003b). Previous studies have shown that *COI* intraspecific divergences are rarely greater than 2% and most are less than 1% in insects (Avise 2000; Hebert et al. 2003a, b). Although there is a small amount of difference between the results from the BI and ML methods, the molecular phylogenetic analysis resolved these samples as six lineages with a high supporting rate. This is



0.04

Figure 15. The gene trees of Sichuana. A. BI phylogenetic tree; B. ML phylogenetic tree.

consistent with results from morphological comparison. Therefore, assigning these specimens to four new species of *Sichuana* is justified.

Conclusion

Based on the morphological and molecular analysis above, we described four new species of *Sichuana* Shen & Yin, 2020, *S. planicercata* sp. nov., *S. curvicercata* sp. nov., *S. longilamina* sp. nov. and *S. magnicerca* sp. nov., and refined the diagnosis of the genus. This large sample suggests that variation in wing venation and the male subgenital plate is common within species in this genus.

Acknowledgements

We are grateful to the editor and the reviewers for providing us with valuable suggestions and comments to improve this manuscript. We thank Wei Yuan for his help in the specimen collecting and data analysis. We thank Dr Bruce Archibald from the Beaty Biodiversity Museum, Vancouver, Canada for improving the language. We thank Rong Huang for her help in the DNA extraction. We thank Huilai Zhang for his help in the map editing. This work was supported by the National Natural Science Foundation of China (grant numbers 41872020). We acknowledge the support of the Museum für Naturkunde Berlin.

References

- Avise JC (2000) Phylogeography: the history and formation of species. Harvard university press. Cambridge. https://doi.org/10.2307/j.ctv1nzfgj7
- Béthoux O, Nel A (2002) Venation pattern and revision of Orthoptera sensu nov. and sister groups. Phylogeny of Palaeozoic and Mesozoic Orthoptera sensu nov. Zootaxa 96(1): 1–88. https://doi. org/10.11646/zootaxa.96.1.1
- Béthoux O (2008) Groundplan, nomenclature, homology, phylogeny, and the question of the insect wing venation pattern. Alavesia 2: 219–232.
- Chivers BD, Béthoux O, Sarria-S FA, Jonsson T, Mason AC, Montealegre-Z F (2017) Functional morphology of tegmina-based stridulation in the relict species Cyphoderris monstrosa (Orthoptera: Ensifera: Prophalangopsidae). Journal of Experimental Biology 220(6): 1112–1121. https://doi.org/10.1242/jeb.153106
- Echeverry DF, Deason NA, Makuru V, Davidson J, Xiao H, Niedbalski J, Yu XY, Stevenson JC, Bugoro H, Aparaimo A, Reuben H, Cooper R, Burkot TR, Russell TL, Collins FH, Lobo NF (2017) Fast and robust single PCR for *Plasmodium* sporozoite detection in mosquitoes using the cytochrome oxidase I gene. Malaria Journal 16: 1–8. https://doi.org/10.1186/s12936-017-1881-1
- Esri R (2011) ArcGIS desktop: release 10. Environmental Systems Research Institute, CA.
- Gu J, Zhao YY, Ren D (2009) New fossil Prophalangopsidae (Orthoptera, Hagloidea) from the Middle Jurassic of Inner Mongolia, China. Zootaxa 2004(1): 16–24. https://doi.org/10.11646/zootaxa.2004.1.2
- Gu JJ, Qiao GX, Ren D (2010) Revision and new taxa of fossil Prophalangopsidae (Orthoptera: Ensifera). Journal of Orthoptera Research 19(1): 41–56. https://doi.org/10.1665/034.019.0110
- Gu JJ, Qiao GX, Ren D (2011) A exceptionally-preserved new species of *Barchaboilus* (Orthoptera: Prophalangopsidae) from the Middle Jurassic of Daohugou, China. Zootaxa 2909(1): 64–68. https://doi. org/10.11646/zootaxa.2909.1.7
- Gu JJ, Yue YL, Wen WC, Zong LY, Ren D (2014) A review of researches on Palaeozoic insects in China. Acta Entomologica Sinica 57(1): 123–132. https://doi.org/10.16380/j.kcxb.2014.01.006
- Gu JJ, Xu Z, Huang R, Wang HJ, Yue YL, Ren D (2021) Systematic significance of wing morphology in extinct Prophalangopsidae (Insecta, Ensifera) revealed by geometric morphometrics and description of two new species. Journal of Systematic Palaeontology 19(22): 1587–1599. https://doi.org/10.1080/14772019.2022.2067491
- He ZQ, Gong P, Hu TH, Yin ZX, Shen ZH, Wang BQ, Li K (2020) A new species of genus *Sichuana* from Sichuan, China (Orthoptera: Tettigoniidae: Tettigoniinae). Zootaxa 4877(1): 195–200. https://doi. org/10.11646/zootaxa.4877.1.10
- Hebert PDN, Cywinska A, Ball SL, de Waard JR (2003a) Biological identifications through DNA barcodes. Proceedings of the Royal Society of London. Series B, Biological Sciences 270(1512): 313–321. https://doi.org/10.1098/rspb.2002.2218
- Hebert PDN, Ratnasingham S, De Waard JR (2003b) Barcoding animal life: Cytochrome c oxidase subunit 1 divergences among closely related species. Proceedings of the Royal Society of London, Series B, Biological Sciences 270: S96–S99. https://doi.org/10.1098/ rsbl.2003.0025
- Heller GK, Liu CX (2016) The East Palaearctic Genus Uvarovina (Orthoptera: Tettigoniidae): Taxonomic Remarks and Bioacoustics. Journal of Orthoptera Research 25(2): 107–112. https://doi. org/10.1665/034.025.0201

- Liu CX (2013) Review of *Atlanticus* Scudder, 1894 (Orthoptera: Tettigoniidae: Tettigoniinae) from China, with description of 27 new species. Zootaxa 3647(1): 1–42. https://doi.org/10.11646/zootaxa.3647.1.1
- Liu CX (2015) New species from the genera Kansua and Anatlanticus (Orthoptera: Tettigoniidae) in China. Zootaxa 3925(2): e291. https://doi.org/10.11646/zootaxa.3925.2.10
- Liu CX, Xu WJ, Zhang CT (2015) A new species of *Mongolodectes* (Orthoptera: Tettigoniidae) from Alashan plateau in China. Zootaxa 4052(2): 246–250. https://doi.org/10.11646/zootaxa.4052.2.11
- Liu CX, Chen GY, Sun BX, Qiu XF, He ZQ (2018) A systematic study of the genus *Atlanticus* Scudder, 1894 from Zhejiang, China (Orthoptera: Tettigoniidae: Tettigoniinae). Zootaxa 4399(2): e170. https://doi.org/10.11646/zootaxa.4399.2.2
- Liu F, Chobanov DP, Chen LS, Liu CX (2019) Ceraeocercus Uvarov, a genus recorded in China for the first time (Orthoptera, Tettigoniidae; Tettigoniinae; Drymadusini). Zootaxa 4608(3): e586. https://doi. org/10.11646/zootaxa.4608.3.12
- Nguyen LT, Schmidt HA, Von Haeseler A, Minh BQ (2015) IQ-TREE: a fast and effective stochastic algorithm for estimating maximum-likelihood phylogenies. Molecular biology and evolution 32(1): 268–274. https://doi.org/10.1093/molbev/msu300
- Pan CY, Hu J, Zhang X, Huang Y (2006) The DNA barcoding application of mtDNA COI gene in seven species of Catantopidae (Orthoptera). Entomotaxonomia 28: 103–110. https://doi.org/10.3969/j.issn.1000-7482.2006.02.004
- Ramme W (1939) Beiträge zur Kenntnis der palaearktischen Orthopteren fauna (Tettig. u. Acrid.) III. Mitteilungen aus dem Zoologischen Museum in Berlin 24: 41–150. https://doi.org/10.1002/mmnz.19320180308
- Ronquist F, Teslenko M, Van Der Mark P, Ayres DL, Darling A, Höhna S, Larget B, Liu L, Suchard MA, Huelsenbeck JP (2012) MrBayes 3.2: Efficient Bayesian phylogenetic inference and model choice across a large model space. Systematic Biology 61(3): 539–542. https://doi.org/10.1093/sysbio/sys029
- Shen ZH, Yin ZX, Lee M, Liu YJ, He ZQ, Wang ZF, Wang TX (2021) Ptosoproctus gen. nov., a new genus with two new species of Shield-back Katydid, with the first record of genus Eulithoxenus Bey-Bienko, 1951 from China (Orthoptera: Tettigoniidae: Tettigoniinae: Drymadusini). Zootaxa 5067(4): 548–568. https://doi.org/10.11646/zootaxa.5067.4.4
- Tamura K, Stecher G, Kumar S (2021) MEGA 11: Molecular Evolutionary Genetics Analysis Version 11. Molecular Biology and Evolution. https://doi.org/10.1093/molbev/msab120
- Wang H, Zheng D, Hou X, Lei X, Zhang Q, Wang B, Fang Y, Jarzembowski EA, Zhang H (2016) The Early Cretaceous orthopteran *Par-ahagla sibirica* Sharov, 1968 (Prophalangopsidae) from the Jiuquan Basin of China and its palaeogeographic significance. Cretaceous Research 57: 40–45. https://doi.org/10.1016/j.cretres.2015.07.014
- Xiong B, Kocher TD (1991) Comparison of mitochondrial DNA sequences of seven morphospecies of black flies (Diptera: Simuliidae). Genome 34(2): 306–311. https://doi.org/10.1139/g91-050
- Yin ZX, He ZQ, Shen CZ, Shen ZH (2020) A new genus with a new species of Shield-back Katydid, with comments on the phylogeny and diagnosis of the genus *Kansua* Uvarov and the tribe Drymadusini (Orthoptera: Tettigoniidae: Tettigoniinae) from China. Zootaxa 4786(3): 369–380. https://doi.org/10.11646/zootaxa.4786.3.3
- Zessin W (1987) Variabilität, Merkmalswandel und Phylogenie der Elcanidae im Jungpaläozoikum und Mesozoikum und die Phylogenie der Ensifera (Orthopteroida, Ensifera). Deutsche Entomologische Zeitschrift 34(1–3): 1–76. https://doi.org/10.1002/mmnd.19870340102