Review of the Leafhopper Genus Phlogotettix Ribaut (Hemiptera: Cicadellidae: Deltocephalinae) with Description of a New Species from India

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Source: Florida Entomologist, 98(1) : 229-236

Published By: Florida Entomological Society

URL: https://doi.org/10.1653/024.098.0139

Review of the leafhopper genus *Phlogotettix* Ribaut (Hemiptera: Cicadellidae: Deltocephalinae) with description of a new species from India

Naresh M. Meshram, N. S. Chandra Bose R. and V. V. Ramamurthy

Abstract

The leafhopper genus *Phlogotettix* Ribaut (Hemiptera: Cicadellidae: Deltocephalinae) is reviewed, and to the 10 species known so far, one new species *Phlogotettix subhimalayanus* **sp. nov.** is added and described. The latter has been analyzed for its mtCOI. *Phlogotettix indicus* Rao is redescribed. An annotated checklist and key to the species is also provided.

Key Words: leafhoppers, *Phlogotettix* spp., Scaphoideini, checklist, key, mtCOI

Resumen

Se revisa el género del insecto saltahojas, *Phlogotettix* Ribaut (Hemiptera: Cicadellidae: Deltocephalinae) y las 10 especies ya conocidas, además se añade y describe una nueva especie *Phlogotettix subhimalayanus* **sp. nov.** Esta última especie ha sido analizada por su mtCOI. Se re-describe *Phlogotettix indicus* Rao. También se presentan una clave y una lista revisada de las especies.

Palabras Clave: saltahojas, chicharritas, *Phlogotettix* spp., Scaphoideini, lista de verificación, lista revisada, clave, mtCOI

The genus *Phlogotettix* (Hemiptera: Cicadellidae: Deltocephalinae) was established by Ribaut (1942) for the Palaearctic *Jassus cyclops* Mulsant & Rey which is also the type species. Ribaut (1952) and Ishihara (1953) redescribed it from France and Japan, respectively. Then, Rao (1989) described *P. indicus* from Khasi hills, Meghalaya, India. Zhang and Wang (1998) added four species from China. Li and Dai (2003) described another species from Taiwan. Gnezdilov (2003) described its eighth species from Eastern China, and Kamitani et al. (2007) added two more species from Japan. With the new species described herein from the Indian subcontinent, the genus will now have 11 species known.

This genus was earlier placed in the tribe Platymetopiini (Oman et al., 1990) of Deltocephalinae. Recently, Zahniser & Dietrich (2013) based on the molecular and morphological aspects of Deltocephalinae placed it in the tribe Scaphoideini. It is distinguished from its related genera by the following characters: generally yellow in color; symmetrical aedeagus with large ventral process or pair of 5 to 6 processes arising caudoventrally, with or without spines or processes, fused to aedeagus; the anterior arms of the connective being strongly divergent forming an obtuse angle, and the subgenital plate with or without sclerotized brace sometimes terminating in a process.

The present study reviews this genus, and provides description of a new species *Phlogotettix subhimalayanus* **sp. nov.** along with redescription of *P. indicus* Rao. An annotated checklist and a key to the species is provided. Also, the new species has been analyzed for its mtCOI and the DNA barcode generated, was deposited in NCBI GenBank (Accession number: KM047668).Colored versions of the Figs. 8-19, 20 and 22-36 can be seen online in Florida Entomologist 98(1) (March 2015) at http://purl.fcla.edu/fcla/entomologist/browse .

Materials and Methods

Leafhopper specimens were collected using sweepnets from Mizoram and presented for studies. Line diagrams were drawn using a drawing tube attached to a Leica DM500 phase contrast compound microscope. Photographs were taken with a Leica DFC 425C digital camera attached on toa Leica M205FA stereozoom microscope with automontage. Male genitalia dissections were carried out as described by Oman (1949) and Knight (1965). Type material is deposited in the National Pusa Collection, Division of Entomology, Indian Agricultural Research Institute, New Delhi, India (NPC).

For mtCOI analysis, the DNA was extracted from a single whole specimen according to the manufacturer protocol, QIAGEN QIAamp® DNA Investigator Kit. The isolated DNA was stored at -20 °C until required. The PCR protocol is after Folmer et al (1994). The DNA extractions were amplified to get PCR products using universal primers LCO1490:5'-ggtcaacaaatcataaagatattgg-3'; HCO2198-5'-taaacttcagggtgaccaaaaaatca-3' which target the mitochondrial cytochrome oxidase subunit I (mtCOI) gene. The 25 µL total volumes of PCR were heated at 94 °C for 4 minutes followed by 35 cycles of 30 s at 94 °C denatuarion, 60 seconds at 47 °C annealing, 50 s at 72 °C extension and a final extension 72 °C for 8 min in a C1000™ Thermal Cycler. The reactions were combined (as described by KOD FX puregene™ manufacturer protocol)

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Supplementary material for this article in Florida Entomologist 98(1) (March 2015), including color-coded nucleotide sequences and color images of leafhoppers, is online at http://purl.fcla.edu/fcla/entomologist/browse.

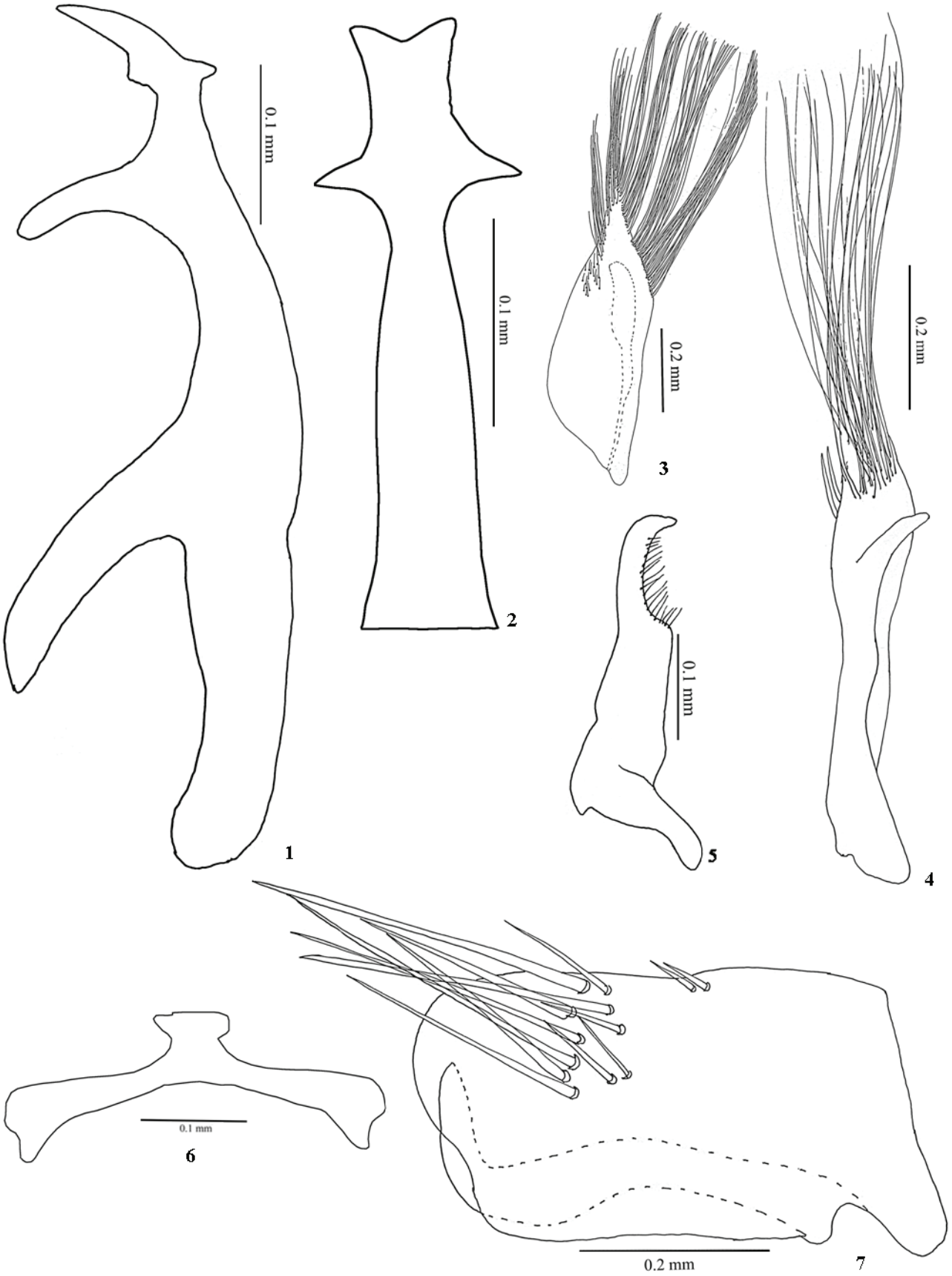

Fig. 1–7. Male genitalia of *Phlogotettix subhimalayanus* **sp. nov.** 1, 2. Aedagus, lateral & caudal view; 3, 4. Subgenital plate, dorsal & lateral view; 5. Style ventral view; 6. Connective; 7. Pygofer.

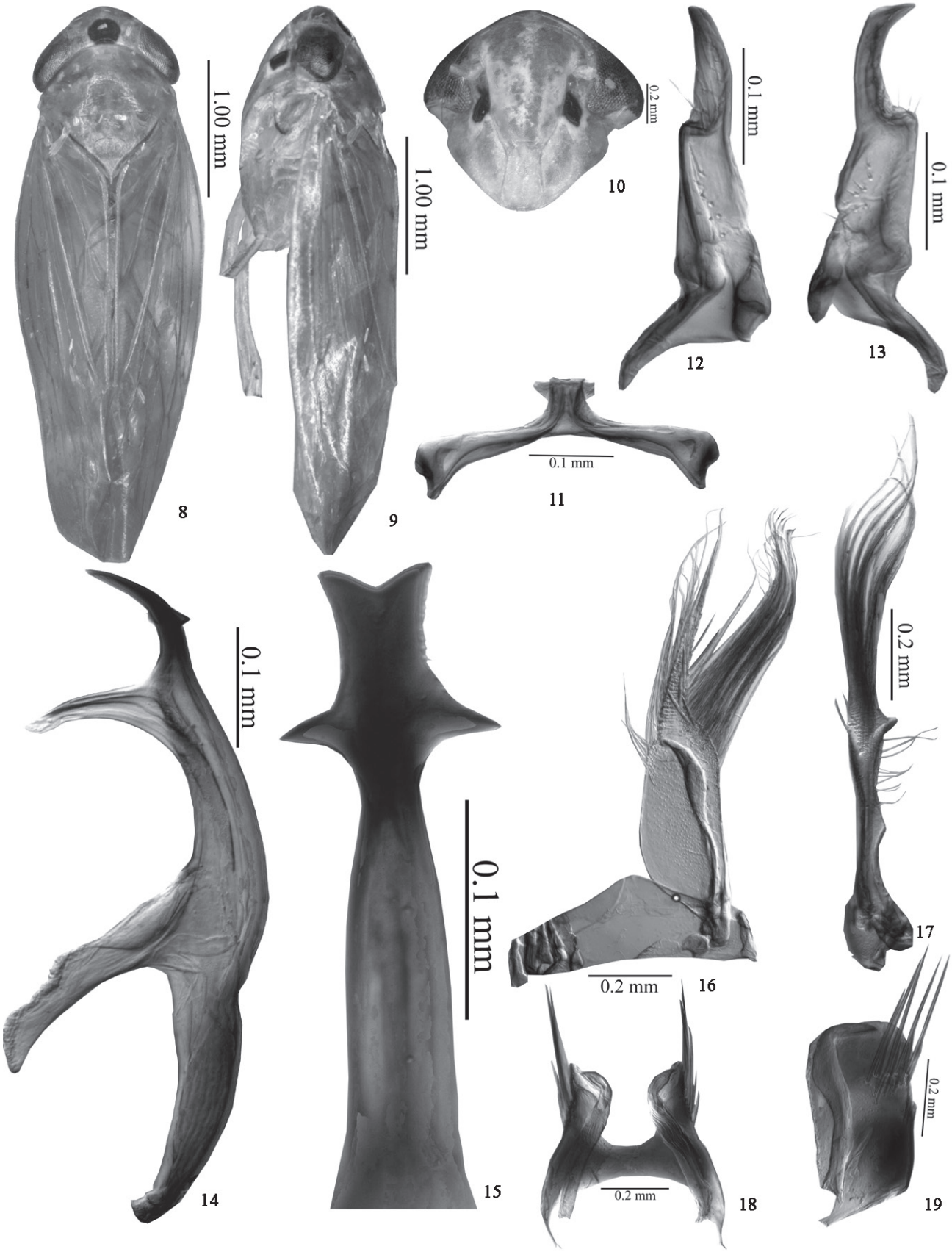

Fig. 8–19. Habitus, face and male genitalia of *Phlogotettix subhimalayanus* **sp. nov.** male 8. Habitus, dorsal view; 9. Habitus, lateral view; 10. Face; 11. Connective; 12. Style, dorsal view; 13. Style ventral view; 14. Aedagus, lateral view; 15. Aedeagus, caudal view; 16. Subgential plate, dorsal view; 17. Subgenital plate, lateral view; 18. Pygofer, lateral view; 19. Pygofer, ventral view. A colored version of this figure can be seen online in Florida Entomologist 98(1) (March 2015) at http://purl.fcla.edu/fcla/entomologist/browse .

of DNA template 4 µL,2× PCR buffer 12.5 µL,2mM dNTP 10 µL, TAQ (KOD FX) enzyme 1unit, and forward and reverse primers were 0.3 µM each at final concentration to the reaction. The products were checked on 2% agarose gel and visualized under UV using Alphaview® software version 1.2.0.1. The amplified products were sequenced at ScigenomicPvt. Ltd. (Cochin, India) The quality sequences were assembled with BioEdit version 7.0.0 and deposited in NCBI GenBank (Accession number: KM047668).

Key to the species of *Phlogotettix* (males)

1. Subgenital plate with longitudinal sclerotized brace (Figs. 16 and 17) 2

—. Subgenital plate without longitudinal sclerotized brace (Kamitani et al. 2007: Figs. 17 and 18) 5

2. Pygofer ventral process posteriorly directed, exceeding posterior margin and distally looped (Li & Wang 1998: Fig. 20); apophysis of style slender, elongate distally hooked . . . . . *tibetensis*

—. Pygofer ventral process dorsally curved at distal one third; not looped; apophysis of style not strongly hooked 3

3. Aedeagal shaft ventral process bifid apically with lateral triangular projection before forking (Figs. 1 and 2 & 14 and 15) . . . . . *subhimalyanus* **sp. nov.**

—. Aedeagal shaft ventral process deeply forked, lacking lateral triangular expansions before the fork 4

4. Aedeagal shaft ventral process forked near base close to divergence from shaft (Li & Wang, 1998: Fig. 4) . . . . . *monozoneus*

—. Aedeagal shaft ventral process forked in apical one third distant from point of divergence from shaft (Figs. 21 and 22) . . . . . *indicus*

5. Aedeagal shaft ventral process as long as or shorter than length of shaft beyond divergence 6

—. Aedeagal shaft ventral process distinctly longer than length of shaft beyond divergence from process 7

6. Crown with brown triangular markings enclosing pale areas; aedeagal shaft straight, not hooked apically (Li & Dai, 2003: Fig. 3) *nigriveinus*

—. Crown with large round black spot; aedeagal shaft curved and hooked apically (Gnezdilov, 2003: Figs. 6 and 7) . . . . . *polyphemus*

7. Pygofer ventral process well developed arising at base and extending dorsally beyond length of pygofer 8

—. Pygofer ventral process small reaching the mid-length of on posterior margin 9

8. Pygofer posterior margin truncate with ventral process going around margin making almost right turn at mid-length (Li & Wang 1998: Fig. 8) . . . . . *luridus*

—. Pygofer posterior margin rounded or triangular, ventral process not curved at right angle at mid-length (Li & Wang 1998: Fig. 14) . *grimeus*

9. Adeagal shaft ventral process fork deep almost near base of divergence from shaft (Kamitani et al. 2007: Figs. 33 and 34) . . . . . *longicornis*

—. Aedeagal shaft ventral process fork shallow distant from base of divergence from shaft 10

10. Gena with circular black spot below antennae (Kamitani et al, 2007: Figs. 9 and 10) . . . . . *cylops*

—. Gena without circular black spot below antennae (Kamitani et al, 2007: Figs. 14–16) . . . . . *cirrhocephalus*

B. Annotated Checklist (Modified from Zahniser 2007)

***Phlogotettix* Ribaut, 1942a: 262. Type species *Jassus cyclops* Mulsant & Rey 1855: 227**

1. *cirrhocephalus* Kamitani, Hayashi & Yamada 2007	Japan
2. *cyclops* (Mulsant & Rey) 1855: 227 *Jassus cyclops* Mulsant & Rey 1855:227 *Phlogotettix cyclops* (Mulsant & Rey): Ribaut 1952, 57: 307	Austria, Belgium, China, Czech Republic, France, Germany, Hungary, Japan, Korea, Romania, Serbia, Slovakia, Taiwan, Eastern Russia.
3. *grimeus* Li & Wang 1998: 374	China
4. *indicus* Rao 1989: 77	India (Meghalaya)
5. *longicornis* Kamitani, Hayashi & Yamada 2007: 371	Japan
6. *lurideus* Li & Wang 1998: 374	China
7. *monozoneus* Li & Wang 1998: 373	China
8. *nigriveinus* Li & Dai 2003: 9	Taiwan
9. *polyphemus* Gnezdilov 2003: 16	China
10. *subhimalayanus* **sp. nov.**	India (Mizoram); Nepal
11. *tibetensis* Li & Wang 1998: 376	China

C. DESCRIPTIONS

i. *Phlogotettix subhimalayanus* **sp. nov.** Meshram & Ramamurthy (Figs. 1–19)

Yellowish. Crown with circular piceous spot in posterior half and touching posterior margin. Face with piceous spot on genal area below antenna. Ocelli transparent, eyes black. Wing veins brown.

Head as wide as pronotum; vertex subtriangular; medial length of vertex 0.27x as long as width including eyes (Fig. 8); ocelli situated on anterolateral margin of vertex between vertex and frons, at distance equal to diameter of ocelli (Fig. 10); eyes large; coronal suture on posterior half of crown short, as long as median length of piceous spot. Frontal suture extending onto vertex, terminating laterad of ocelli. Frontoclypeus elongate, gradually widened towards apex; transclypeal suture distinct; clypellus slightly narrowed near base and gradually widened to apex. Antennae situated somewhat at level with upper margin of eye in facial view. Scutellum 1.1x as long as pronotum, transverse depression distinct and nearly reaching lateral margin. Pronotum 0.43x longer than broad and 1.6x longer than vertex (Fig. 8).

Male genitalia. Pygofer quadrangular in lateral view, ventral process sharply curved dorsally with acute apex and not reaching the dorsal of margin (Figs. 7, 18 and 19). Subgenital plate long, terminated by narrow fingerlike process, with numerous long hairs; plate dorsally with longitudinal sclerotized brace (Figs. 3 and 4 & 16 and 17). Style slender, apophysis acute at apex with well developed preapical lobe laterally (Figs. 5& 12 and 13). Anterior arms of the connective strongly divergent forming an obtuse angle (Figs. 6,11). Aedeagal shaft with its ventral process bifid apically with lateral triangular projection before forking. (Figs. 1 and 2 & 14 and 15).

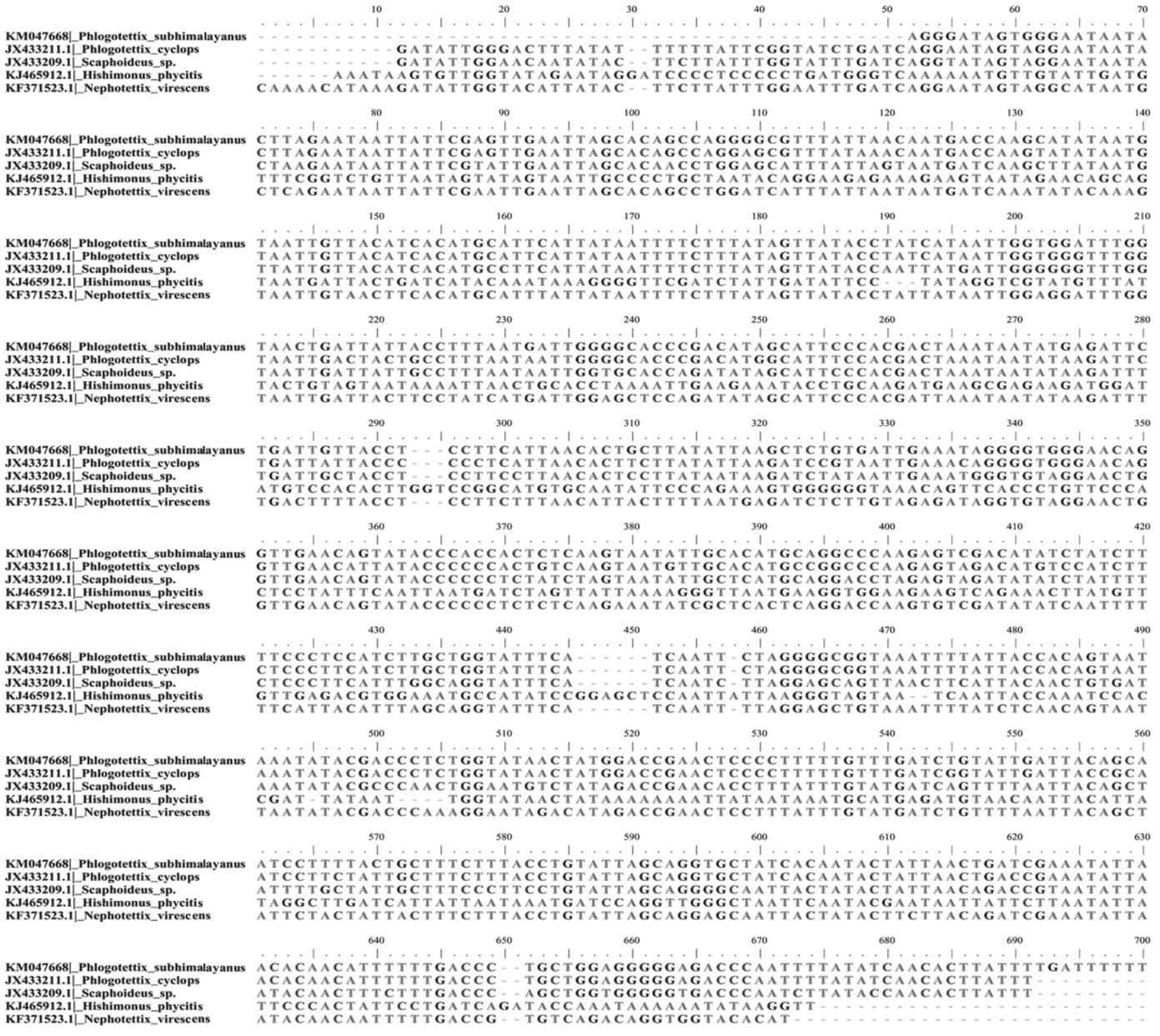

Fig. 20. Nucleotide sequence map of mtCOI of *Phlogotettix subhimalayanus* **sp. nov.** A colored version of this figure can be seen online in Florida Entomologist 98(1) (March 2015) at http://purl.fcla.edu/fcla/entomologist/browse .

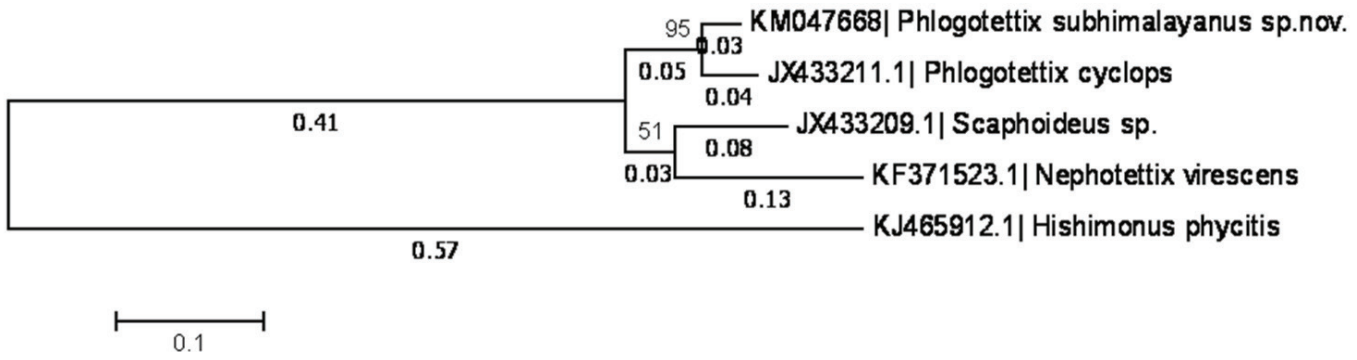

Fig. 21. Relationships of *P. subhimalayanus* **sp. nov.** and *P. cyclops* inferred using by Maximum Likelihood method and the kimura 2-parameter distances of mitochondrial COI sequences. Bootstrap values are shown next to the branches.

Measurements: Male 4.5 mm long, 1.1 mm wide across eyes, 0.9 mm wide across hind margin of pronotum. Female 4.6 mm long, 1.1 mm wide across eyes, 0.9 mm wide across hind margin of pronotum

TYPE MATERIAL

HOLOTYPE male, INDIA: Mizoram: Kolasib (23° 18′ 0″ N 92° 49′ 48″ E, 888 m), 26.iv.2012, mercury vapor light, Roni, K. PARATYPES, 2 males & 4 females, data same as holotype (NPC).

REMARKS

Phlogotettix subhimalayanus **sp. nov.** resembles *Phlogotettix cyclops* (Mulsant & Rey), but differs in following characters: Aedeagus ventral process very shallowly forked and with well developed subapical lateral angular projections in caudal view. It was observed that, the subapical projection of *P. cylops* from Europe and that of East Asia are very short (Courtesy: Dr. Satoshi Kamitani, Japan). Subgenital plate with dorsal sclerotized brace-like process directed caudodorsally which is absent in *P. cyclops.*

The molecular data are given in Figs. 20 and 21, MEGA V6.0 (Tamura et al. 2013) was used to calculate the Kimura 2- parameter model (Kimura 1980) for mtCOI sequence. This revealed that the percent of sequence variation between *P. subhimalayanus* **sp. nov.** and *P. cyclops* is at least 6.5 % which was confirmed from the mtCOI sequences. The maximum likelihood tree was constructed including the near available species. Out groups were taken from NCBI Genbank and accession numbers are denoted with 2000 bootstraps and the node length depicted in the tree (Figs. 20 and 21).

ii. *Phlogotettix indicus* Rao (Figs. 22–36)

Yellowish. Head, pronotum, scutellum and legs with darker shade of yellowish. Crown with circular piceous spot in posterior half touching posterior margin. Face with or without semicircular piceous spot on genal area below antenna. Ocelli transparent, eyes black. Wing veins brown (Figs. 22–27).

Head as wide as pronotum; vertex subtriangular; medial length of vertex 0.28x as long as width including eyes (Fig. 24); ocelli situated on anterolateral margin of vertex between vertex and frons, at distance equal to diameter of ocelli (Figs. 26 and 27); eyes large, elongate covering half of entire dorsal area; coronal suture on posterior surface of vertex short, as long as median length of piceous spot on vertex. Frontal suture extending onto vertex, terminating laterad of ocelli. Frontoclypeus elongate, gradually widened towards apex; transclypeal suture distinct; clypellus slightly narrowed near base and gradually widened to apex. Antennae situated somewhat at level with upper margin of eye in facial view. Scutellum 1.2 x as long as pronotum, transverse depression distinct and nearly reaching lateral margin. Pronotum 0.43 x longer than broad and 1.7 x longer than vertex (Fig. 24).

Male genitalia. Pygofer with rounded outer margin, ventral process with acute apex and not reaching dorsal margin (Figs. 35 and 36). Subgenital plate long terminated in narrow fingerlike process with numerous long hairs; plate with dorsal with longitudinal sclerotized brace (Figs. 33 and 34). Style slender, apophysis fingerlike with well developed preapical lobe laterally (Figs. 29 and 30). Anterior arms of the connective strongly divergent forming an obtuse angle (Fig. 28). Aedeagal shaft ventral process forked in apical one third distance from point of divergence from shaft (Figs. 31 and 32).

Measurements: Male 4.6–4.7 mm long, 1.1 mm wide across eyes, 0.88 mm wide across hind margin of pronotum, Female 4.7 mm long, 1.1 mm wide across eyes, 0.9 mm wide across hind margin of pronotum.

MATERIAL EXAMINED

HOLOTYPE male, INDIA: Meghalaya, Ranichor, 7.xii.1977, I/HC 102 K.R. Rao Coll. Paratype: female, same as holotype except I/HC 103 (Zoological Survey of India). 4 male Mizoram: Kolasib (23°18′0″N 92°49′48″E, 888 m), 26.iv.2012, mercury vapor light, Roni, K.

REMARKS

The species is redescribed and illustrated adding to its diagnostics. The facial piceous spot below antennal base are variable, and are absent in the specimens from Meghalaya and Mizoram but present in the holotype.

Acknowledgments

The authors are grateful to Prof. C. A. Viraktamath for his comments on an earlier draft of the manuscript; also thanks Dr. Satoshi Kamitani for discussion on the Japanese species of *Phlogotettix*. The financial support from the ICAR XII Plan Network Project on Insect Biosystematics (NPIB) is acknowledged.

Fig. 22–36. Habitus, face and male genitalia of *Phlogotettix indicus* Rao 22 & 23 female 22–23. Habitus, dorsal & lateral view; 24–36 male 24–25. Habitus, dorsal & lateral view; 26 –27 Face; 28. Connective; 29–30. Style, dorsal & lateral view; 31. Aedagus, lateral view; 32. Aedeagus, caudal view; 33–34. Subgential plate, dorsal & lateral; 35–36. Pygofer, lateral & ventral view. A colored version of this figure can be seen online in Florida Entomologist 98(1) (March 2015) at http://purl.fcla.edu/fcla/entomologist/browse .

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