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Source: Integrative Systematics: Stuttgart Contributions to Natural History, 6(1) : 51-58

Published By: Stuttgart State Museum of Natural History

URL: <https://doi.org/10.18476/2023.462484>

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RESEARCH ARTICLE

Description of *Tonnoira chuki* sp. n. (Diptera: Psychodidae) from Ecuador, with an updated identification key for the genus *Tonnoira*

SANTIAGO JAUME-SCHINKEL

Abstract

Tonnoira chuki sp. n. is described based on specimens collected in a pre-mountain rainforest in Pichincha Province, northeastern Ecuador. Illustrations and photographs of the new species are provided. The identification key to males of the genus is updated. Furthermore, the first published DNA barcodes (*COI*) for the genus are provided.

Keywords: dark taxa, DNA barcoding, moth flies, new taxa, Psychodinae, taxonomy.

Zusammenfassung

Tonnoira chuki sp. n. wird anhand von Exemplaren beschrieben, die in einem Vorgebirgsregenwald der Provinz Pichincha im Nordosten Ecuadors gesammelt wurden. Die neue Art wird anhand Illustrationen und Fotos dargestellt. Der Bestimmungsschlüssel für die Männchen der Gattung wird aktualisiert. Zudem werden die ersten DNA-Barcodes (*COI*) für die Gattung zur Verfügung gestellt.

Introduction

The Neotropical genus *Tonnoira* Enderlein, 1937 is currently present in ten countries, not including the Caribbean islands, ranging from Nicaragua to Brazil (BRAVO et al. 2008; SANTOS & CURLER 2014). The genus was erected based on a single female, *Tonnoira pelliticornis* Enderlein, 1937, and to date, the male sex of the type species remains unknown. Since then, a total of 27 extant species have been described (ENDERLEIN 1937; WAGNER 1981; QUATE 1996, 1999; BRAVO & CHAGAS 2004; QUATE & BROWN 2004; BRAVO et al. 2008, 2020; CHAGAS-VIEIRA 2012; SANTOS & CURLER 2014; JAUME-SCHINKEL 2022).

The present work describes a new species from male individuals collected in Ecuador, and the first DNA barcodes (*COI* gene) for this genus are provided. Moreover, an updated identification key to the known males of *Tonnoira* species of the world is given.

Material and methods

Study area. Cantón Pedro Vicente Maldonado is located in Pichincha Province in the northeastern part of Ecuador (0.1667N, -79.0000E), with an average altitude of 600 m. a.s.l. Pedro Vicente Maldonado experiences a tropical climate characterized by warm to hot temperatures throughout the year. Average highs range from 25 °C to 30 °C, while average lows range from 18 °C to 22 °C. The wet season typically extends from December to May, with the highest rainfall occurring from January to April and an average annual precipitation of 4,341 mil-

limeters. The region includes lowland rainforests, cloud forests, and the foothills of the Andes, but the main vegetation is pre-mountain rainforest (HPPC 2015).

General morphology follows CUMMING & WOOD (2017) and KVIFTE & WAGNER (2017).

Measurements. The length of the wing was measured from its base, at the start of the costal node, to its apex, while its width was roughly measured with an imaginary line crossing the wing at the apex of vein CuA₂. The width of the head was measured at its widest part, roughly above the insertion of the antennal scape, and its length was measured from the vertex to the lower margin of the clypeus. Palpal proportions are provided using the first segment's length as a unit of measurement (1.0).

All examined material was collected using double Malaise traps with 96% ethanol as a killing and preserving medium, and temporarily stored in 96% ethanol for DNA extraction. Whole specimens were used for DNA extraction and processed at Museum Koenig (ZFMK; previously known as Zoologisches Forschungsmuseum Alexander Koenig) in Bonn (Germany). Lysis and PCR were performed at ZFMK following the protocol by ASTRIN & STÜBEN (2008) and the primers from FOLMER et al. (1994). After the PCR, samples were sent to BGI Group (formerly Beijing Genomics Institute) for bidirectional sequencing. Raw data were curated manually using Geneious (v. 7.1.9). Final *COI* sequences were 658 bp long. All sequences are publicly available in GenBank (<https://www.ncbi.nlm.nih.gov/genbank/>) and/or BOLD (<https://www.boldsystems.org>).

After DNA extraction, whole specimens were put back into 96% ethanol and then further dehydrated in absolute ethanol (100%) for five minutes, transferred to clove oil for 10 minutes, and mounted on microscope slides using Euparal.

The material examined for this study is deposited in the following natural history institutions, referred to in the text with the following acronyms:

INABIO: Instituto Nacional de Biodiversidad, Quito, Ecuador.
ZFMK: Museum Koenig, Bonn, Germany.

In the material examined section and at the end of each record, the holding institution is indicated in square brackets ([]). In the description of type labels, the contents of each label are enclosed in double quotation marks (" "), and a double forward slash (//) separates the individual lines of data.

Taxonomic account

Genus *Tonnoira* Enderlein, 1937

Type species: *Tonnoira pelliticornis* Enderlein, 1937: 106; type locality: Peru, Callanga.

Important references: ENDERLEIN (1937: 106; original description); QUATE (1963: 189; diagnosis); QUATE (1996: 33; revised description); QUATE & BROWN (2004: 25; revised description); SANTOS & CURLER (2014: 464; updated diagnosis); BRAVO et al. (2020: 4; species list; identification key); JAUME-SCHINKEL (2022: 2–3; updated distribution map and species list).

To date, there are 28 described species in the genus, including the herein newly described species.

Tonnoira chuki sp. n.

(Figs. 1–2)

Differential diagnosis

The aedeagal shape of *Tonnoira chuki* sp. n. resembles those of *Tonnoira rapiformis* Quate & Brown, 2004 and *T. bitenacula* Quate, 1996, but both these species can be separated from *Tonnoira chuki* sp. n. by the number of tenacula present on the epandrial appendages (surstyli): one in *Tonnoira chuki* sp. n. and two in *T. rapiformis* and *T. bitenacula*. Furthermore, *Tonnoira chuki* sp. n. can be differentiated from *T. castanea* Quate & Brown, 2004 by the shape of the aedeagus (symmetrical and spear-shaped in *Tonnoira chuki* sp. n., asymmetrical and not spear-shaped in *T. castanea*).

Type locality

Ecuador, Pichincha Prov., Parroquia Pedro Vicente Maldonado.

Type material

Holotype: "Ecuador, Pichincha Prov. // Parroquia Pedro Vicente // Maldonado, roadway to // Pachijal. // 0.1156110000000001, // -78.95805359999996. 750 m. // 1-9. February.2022 // ZFMK-TIS-2637090 // Leg. Kilian, Isabel" "SJS-00811" "ZFMK-DIP-00097120" "Psychodidae // Tonnoira // chuki" "HOLOTYPE [red]" [INABIO].

Paratypes: 2 ♂♂, same label information except: SJS-00824, ZFMK-DIP-00097121; SJS-00837, ZFMK-DIP-00097122 [INABIO]; 4 ♂♂, same label information except SJS-00838, ZFMK-DIP-00097123; SJS-00847, ZFMK-DIP-00097124; SJS-00852, ZFMK-DIP-00097125; SJS-881, ZFMK-DIP-00097126 [ZFMK]; 1 ♂, "Ecuador, Pichincha Prov., // Parroquia Pedro Vicente // Maldonado, 0.118626, -78.95802240, 770m. // 25-

28.January.2020. // Leg. Kilian, Isabel." "ZFMK-DIP-00081674" "Psychodidae // Tonnoira // chuki // det. Jaume-Schinkel, Santiago" "PARATYPE [yellow]" [ZFMK]; 1 ♂, same label information except "ZFMK-DIP-00081650" [ZFMK].

Description

Male. Measurements in mm (n=5) Wing length 2.20 (2.3–2.1), width 1.05 (1.1–0.9); head length 0.52 (0.56–0.50), width 0.52 (0.55–0.49); antennal segments: scape 0.12 (0.13–0.18), pedicel 0.07 (0.09–0.05), flagellomeres 1–5: 0.22 (0.25–0.20); palpomere 1: 0.08 (0.1–0.07), palpomere 2: 0.18 (0.20–0.17), palpomere 3: 0.18 (0.20–0.18), palpomere 4: 0.17 (0.19–0.16).

Head. Slightly longer than wide; eye bridge separated by less than one eye facet diameter, with four rows of facets, interocular suture as an inverted "Y"; frontal patch of alveoli almost divided in two but joined in the middle, upper margin M-shaped, lower margin rounded, with a concavity in the middle. Antennal scape about two times the length of the pedicel, almost cylindrical; pedicel spherical; flagellomeres cylindrical, at least four times longer than wide, with scattered setae on the surface, apical flagellomeres absent in the examined material, maximum number of flagellomeres present five; ascoids indistinguishable in the examined material. Palpal segments cylindrical, palpal proportions: 1.0:2.0:2.0:1.9; labium without any strong sclerite; labella bulbous, with seven setae scattered between the middle and the apical margin.

Thorax without allurement organs, with a single patch of alveoli on the paratergite and antepronotum; all coxae with a stripe of three to five rows of alveoli. Wing length about two times its width; wing membrane brown-infuscated; subcostal vein short, ending at level of origin of R₄; junction of R₂₊₃ basal to M₁₊₂, stem of R₂₊₃ very short; R₅ ending slightly below the wing apex; CuA₂ ending at wing margin.

Terminalia (Figs. 1D, E, 2A–C). Hypandrium a distinct band connecting the gonocoxites, narrow and arch-like; gonocoxites about the same length as gonostyli; gonostyli lightly incurved, with a sudden lateral and digitiform narrowing as in Fig. 2A, B, covered with scattered alveoli; ejaculatory apodeme shorter than aedeagus, rounded; aedeagus spear-tip-shaped, formed of two triangular and elongated phallomeres, joined at the apex; aedeagus with two digitiform parameres, parameres narrowing towards apex, resembling an inverted V; below the aedeagal complex is a triangular, semi-sclerotized structure that resembles an aedeagal sheath, here considered to be the subepandrial sclerite; epandrium about two times wider than long; hypoproct triangular, longer than epandrium and covered in small setulae; epiproct shorter than hypoproct; epandrial appendages (surstyli) conical, tapering towards the apex and curved dorsally, each with one apical tenaculum, tenacula with rounded apex.

Female. Unknown.

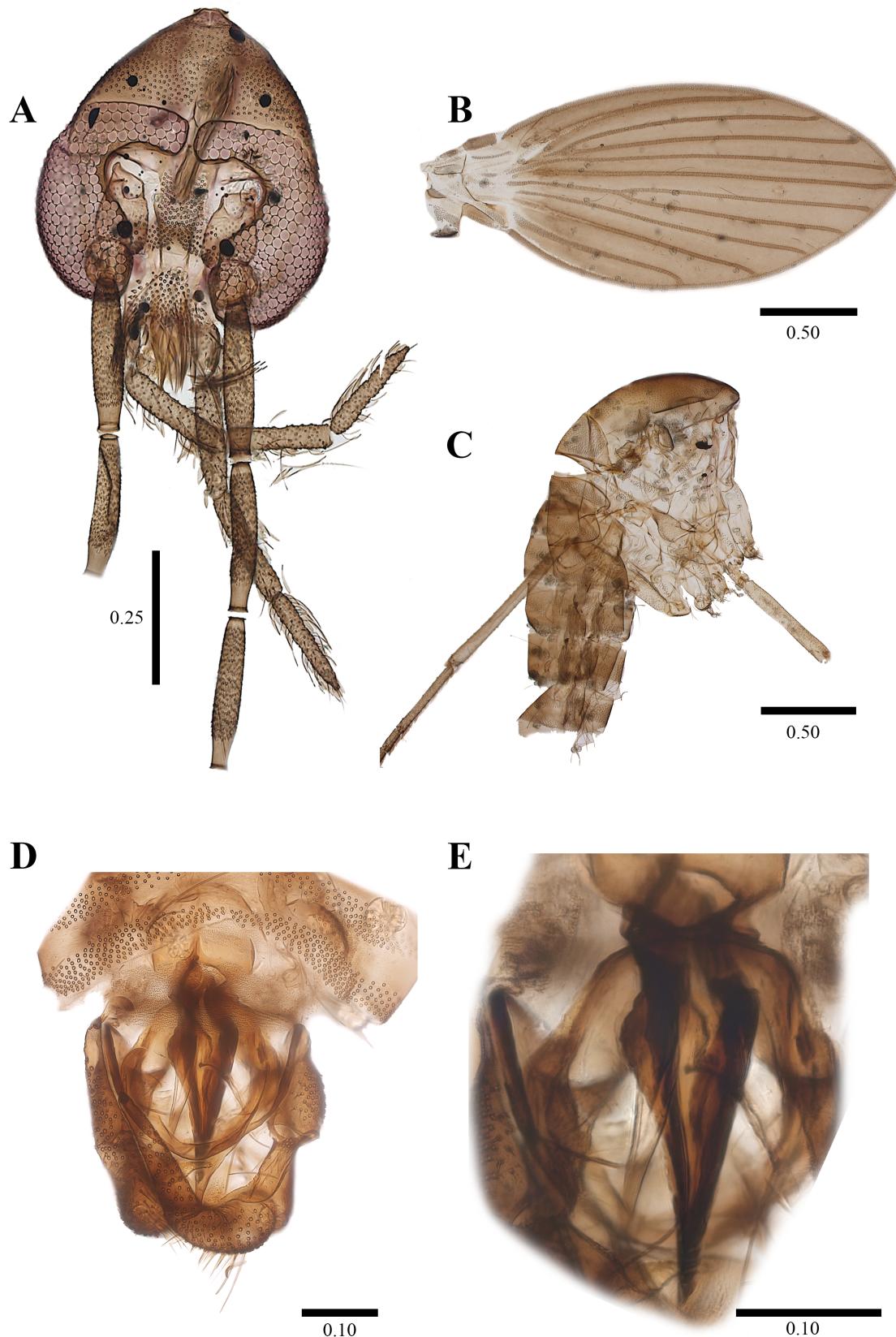


Fig. 1. *Tonnoira chuki* sp. n., male holotype. **A.** Head. **B.** Wing. **C.** Thorax and abdomen. **D.** Genitalia. **E.** Aedeagus. Scales in mm.

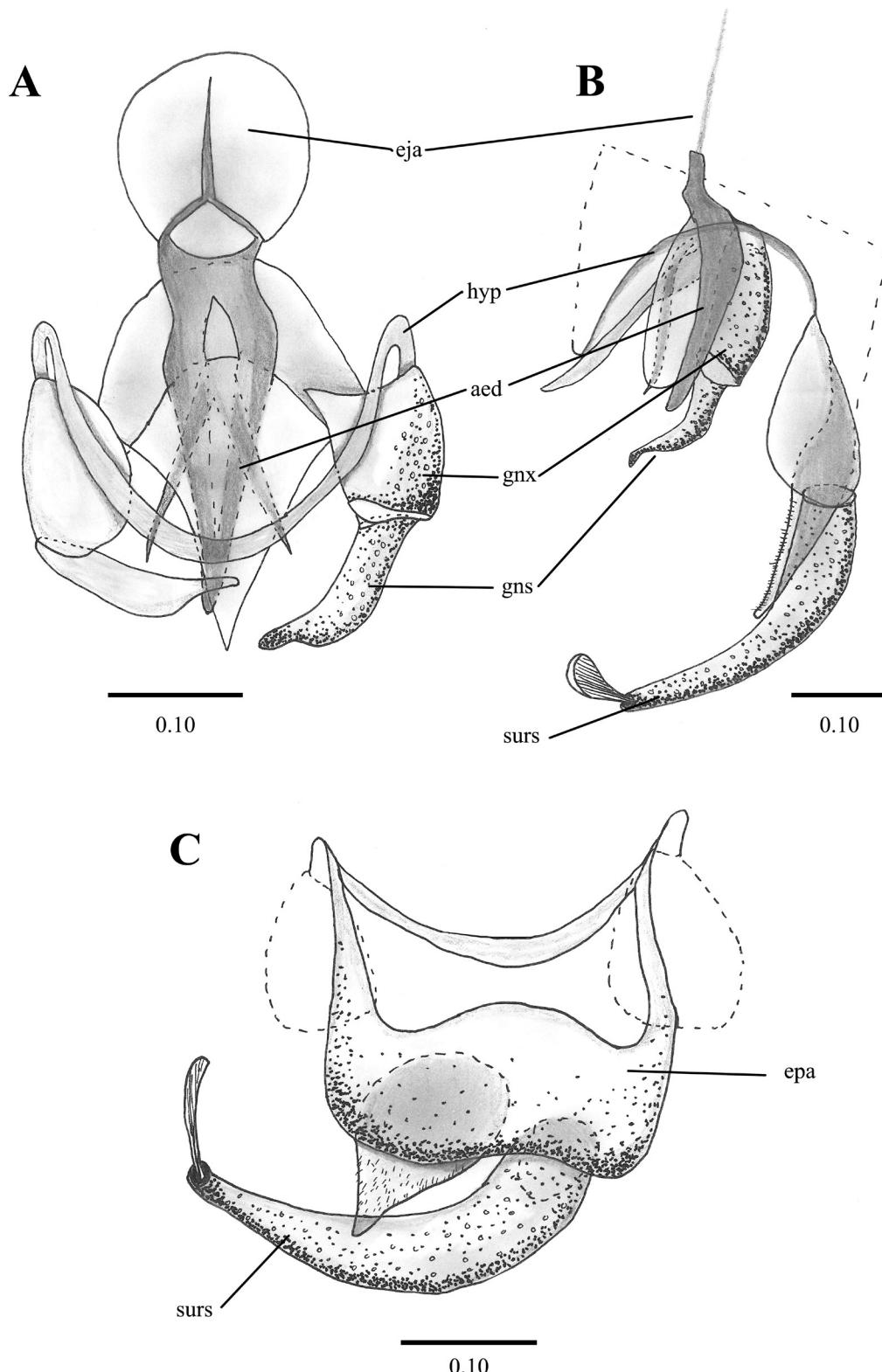


Fig. 2. *Tonnoira chuki* sp. n., male paratype. **A.** Dorsal view of aedeagus, gonocoxites and gonostyli. **B.** Lateral view of terminalia. **C.** Ventral view of epiproct, hypoproct, epandrium, and surstyli. Abbreviations: aed: aedeagus, ej: ejaculatory apodeme, epa: epandrium, gns: gonostylus, gnx: gonocoxites, hyp: hypandrium, surs: epandrial appendage (surstylus). Scales in mm.

Etymology

The specific epithet is derived from the Quechuan word “chuki” meaning spear, in reference to the spear-shaped aedeagus.

Distribution

Only known from the type locality in Ecuador.

Genetics

Nine specimens were successfully sequenced: ZFMK-DIP-00097120, ZFMK-DIP-00097121, ZFMK-DIP-00097122, ZFMK-DIP-00097123, ZFMK-DIP-00097124, ZFMK-DIP-00097125, ZFMK-DIP-00097126, ZFMK-DIP-00081674, and ZFMK-DIP-00081650. The maximum intraspecific uncorrected pairwise distance between sequences was 1.82% or 12 bp. GenBank accession numbers are: ZFMK-DIP-00097120 (ZFMK-TIS-2637090): OQ685791; ZFMK-DIP-00097121 (ZFMK-TIS-2637103): OQ685797; ZFMK-DIP-00097122 (ZFMK-TIS-2637116): OQ685794; ZFMK-DIP-00097123 (ZFMK-TIS-2637117): OQ685792; ZFMK-DIP-00097124 (ZFMK-TIS-2637126): OQ685795; ZFMK-DIP-00097125 (ZFMK-TIS-2637131): OQ685793; ZFMK-DIP-00097126 (ZFMK-TIS-2637160): OQ685796; BOLD sequence accession numbers are: ZFMK-DIP-00081674 (ZFMK-TIS-2629869): GDIP21965-23; ZFMK-DIP-00081650 (ZFMK-TIS-2629888): GDIP21976-23.

Key to known males of extant species of *Tonnoira*

[modified from BRAVO et al. (2020) and JAUME-SCHINKEL (2022)]

- 1 Gonostylus simple, not bifurcated 9
- Gonostylus bifurcated 2
- 2 Gonostylus with lateral and mesal branches of the same length; branches crossed (see SANTOS & CURLER 2014: fig. 4) *T. brisolaii* Santos & Curler, 2014
- Gonostylus with lateral and mesal branches of different lengths; branches subparallel or divergent 3
- 3 Lateral branch of gonostylus shorter than mesal branch; mesal branch of gonostylus longer than gonocoxite (see BRAVO & CHAGAS 2004: fig. 8) 6
- Lateral branch of gonostylus longer than mesal branch; mesal branch of gonostylus shorter than gonocoxite (see SANTOS & CURLER 2014: fig. 12) 4
- 4 Eye bridge with 5 facet rows; epandrial appendages (surstyli) with one tenaculum..... *T. protuberata* Quate & Brown, 2004
- Eye bridge with 4 facet rows; epandrial appendages (surstyli) with two tenacula 5
- 5 Eyes separated by 0.5 facet diameter; parameres symmetrical (see SANTOS & CURLER 2014: fig. 12) *T. andradei* Santos & Curler, 2014
- Eyes separated by almost 1.0 facet diameter; parameres asymmetrical (see BRAVO et al. 2020: fig. 9) *T. igrapuensis* Bravo, Vilarinho & Chagas, 2004
- 6 Lateral branch of gonostylus very short, 0.2 times the length of mesal branch (see SANTOS & CURLER 2014: fig. 8)..... *T. ferreirai* Santos, 2014

- Lateral branch of gonostylus 0.4–0.6 times the length of mesal branch (see BRAVO & CHAGAS 2004: fig. 8) 7
- 7 Aedeagus bipartite, basally U-shaped, with one of the branches longer than the other (see QUATE & BROWN 2004: fig. 69 a) *T. bifurcata* Quate & Brown, 2004
- Aedeagus bipartite, with neither of the branches basally U-shaped, or tripartite 8
- 8 Lateral and mesal branches of gonostylus with the same degree of sclerotization; hypandrium narrower than gonocoxite, stripe-like (see QUATE & BROWN 2004: fig. 70) *T. didyma* Quate & Brown, 2004
- Lateral branch of gonostylus appearing more sclerotized than mesal branch; hypandrium wider than gonostylus (see BRAVO & CHAGAS 2004: fig. 6) *T. bifida* Bravo & Chagas, 2004
- 9 Epandrial appendages (surstyli) with 2 or 3 tenacula 14
- Epandrial appendages (surstyli) with 1 tenaculum 10
- 10 Base of wing vein R₃ with a cluster of black “granules” (see QUATE & BROWN 2004: fig. 63) *T. psacadoptera* Quate & Brown, 2004
- Base of wing vein R₃ normal, without a cluster of black “granules” 11
- 11 Hypandrium broad, of same length as width of gonostylus; gonocoxite 0.8 times as long as gonostylus (see CHAGAS-VIEIRA 2012: fig. 16) *T. tripennis* Chagas-Vieira, 2012
- Hypandrium narrow, stripe-like, shorter than width of gonostylus; gonocoxite about as long as gonostylus 12
- 12 Aedeagus tripartite with a short spur/like branch, a sickle-shaped lateral branch, and a broad, twisted and paddle-shaped central branch (see QUATE & BROWN 2004: fig. 68) *T. sicilis* Quate & Brown, 2004
- Aedeagus bipartite, branches may be joined at apex 13
- 13 Aedeagus bipartite, one branch with basal three-quarters broad and asymmetrically narrowing at distal one-quarter, other branch digitiform, blunt, about half as long as other branch (see QUATE & BROWN 2004: fig. 67a) *T. castanea* Quate & Brown, 2004
- Aedeagus with two triangular sclerites joining towards the apex, resembling the tip of a spear (Figs. 1D, E, 2A), with two additional spine-like parameres *T. chuki* sp. n.
- 14 Epandrial appendages (surstyli) with 2 tenacula 16
- Epandrial appendages (surstyli) with 3 tenacula 15
- 15 Aedeagus ending at same level as apex of gonostylus; ejaculatory apodeme shorter than gonostylus (see WAGNER 1981: fig. 3) *T. mirabilis* Wagner, 1981
- Aedeagus ending beyond apex of gonostylus; ejaculatory apodeme longer than gonostylus (see CHAGAS-VIEIRA 2012: fig. 12) *T. spina* Chagas-Vieira, 2012
- 16 Aedeagus symmetrical or asymmetrical, long, subcylindrical, pointed or rounded at apex; parameres present 18
- Aedeagus symmetrical, base broad, apex sagittate; parameres absent (see QUATE 1996: fig. 13 b; QUATE & BROWN 2004: fig. 71) 17
- 17 Hypandrium present as a single stripe-like structure; gonocoxites as wide as long (see QUATE 1996: fig. 13 b) *T. bitemacula* Quate, 1996
- Hypandrium vestigial, sclerotization interrupted and broken in the middle; gonocoxites longer than wide (see QUATE & BROWN 2004: fig. 71) *T. rapiformis* Quate & Brown, 2004
- 18 Aedeagus ending far beyond apex of gonostylus (see BRAVO & CHAGAS 2004: fig. 1) *T. longipennis* Bravo & Chagas, 2004
- Aedeagus ending near or just beyond apex of gonocoxites, never beyond apex of gonostylus 19

- 19** Aedeagus sickle-shaped; ejaculatory apodeme triangular and shorter than aedeagus (see QUATE & BROWN 2004: fig. 77) *T. rectilata* Quate, 1999
- Aedeagus straight or slightly curved; ejaculatory apodeme sub-oval, longer than wide, of same length or longer than aedeagus **20**
- 20** First flagellomere cylindrical **24**
- First flagellomere fusiform **21**
- 21** Gonocoxal apodeme expanded posteriorly, with a pair of slender, acute processes flanking the aedeagus and extending nearly to tip of aedeagus (see QUATE 1999: fig. 5k).....
..... *T. bitalea* Quate, 1999
- Gonocoxal apodeme not expanded posteriorly, without processes flanking the aedeagus **22**
- 22** Hypandrium wide, posteriorly bilobed (see BRAVO & CHAGAS 2004: fig. 23) *T. magna* Bravo & Chagas, 2004
- Hypandrium narrow, posteriorly without lobes **23**
- 23** Eyes separated by 0.5 facet diameters; gonostyli shorter than gonocoxites; ejaculatory apodeme shorter than gonocoxites (see BRAVO et al. 2008: figs. 11, 16).....
..... *T. robusta* Bravo, Alves & Chagas, 2008
- Eyes separated by 0.2 facet diameters; gonostyli longer than gonocoxites; ejaculatory apodeme longer than gonocoxites; hypandrium with triangular projections at posterolateral margin; aedeagus not bipartite, with two parameres (see SANTOS & CURLER 2014: fig. 16).....
..... *T. galatiae* Santos & Curler, 2014
- 24** Aedeagus bipartite, with one branch abruptly narrowing to acute apex, other branch digitiform, evenly narrowing towards the apex; aedeagus without parameres (see QUATE & BROWN 2004: fig. 74) *T. fusiformis* Quate & Brown, 2004
- Aedeagus not bipartite, evenly narrowing to an acute apex... **25**
- 25** Eyes separated by one facet diameter; aedeagus extending to middle of gonostyli, with two parameres ending at the same level as apex of aedeagus (see BRAVO et al. 2008: fig. 6).....
..... *T. distincta* Bravo, Alves & Chagas, 2008
- Eyes separated by less than one facet diameter; aedeagus extending to apex of gonocoxite or more, but never close to middle of gonostyli, one or two parameres present **26**
- 26** Aedeagus extending a little beyond apex of gonocoxite; gonostyli digitiform, tapering towards apex, incurved; aedeagus with one paramere (see QUATE & BROWN 2004: fig. 72)
..... *T. cavernicola* Quate & Brown, 2004
- Aedeagus extending to apex of gonocoxites; gonostyli conical, straight; aedeagus with two parameres (see JAUME-SCHINKEL 2022: figs. 2c, 3).....
..... *T. conistylus* Jaume-Schinkel, 2022

Discussion

The distribution of *Tonnoira* is restricted to the Neotropical Region, ranging from Nicaragua to Brazil (BRAVO et al. 2008, 2020; SANTOS & CURLER 2014; JAUME-SCHINKEL 2022). Currently, Brazil has the highest recorded diversity with 18 species, followed by Suriname with five species, and Ecuador, now with four species. This discrepancy in the numbers of recorded and/or described species throughout the Neotropics highlights the lack of taxonomic surveys in this biogeographic realm, and there is no doubt that new species are still waiting to be described.

The general biology and larval stages of *Tonnoira* remain greatly understudied. Three species have been found to be associated with caves (BRAVO et al. 2020; JAUME-SCHINKEL 2022); nonetheless, for the majority of species, the micro-habitat preferences and life cycle remain unknown.

Tonnoira is placed in the tribe Maruinini (see KVIFTE 2018). To date, the only publicly available *COI* barcodes of other Maruinini genera belong to *Alepidia viatrix* Jaume-Schinkel, Kvifte, Weele & Mengual, 2022 and *Platyplastinx ibanezbernalii* Jaume-Schinkel & Kvifte, 2022 (JAUME-SCHINKEL et al. 2022; JAUME-SCHINKEL & KVIFTE 2022). The maximum intraspecific uncorrected pairwise distance for *COI* sequences in *Alepidia viatrix* is 5.71%, while specimens of *Platyplastinx ibanezbernalii* present a maximum intraspecific uncorrected pairwise distance of 1.35%; therefore, the intraspecific distance of 1.82% in *Tonnoira chuki* sp. n. does not differ greatly from the intraspecific uncorrected pairwise distances reported for related genera. Nonetheless, further DNA barcodes from different genera and species are required to properly assess the intraspecific and interspecific uncorrected pairwise distances for the tribe.

Acknowledgements

The present results are part of the Marco contract entitled “Diversidad de moscas florícolas (Insecta: Diptera) del Ecuador” (MAAEDBI-CM-2021-0167), issued by the Ecuadorian Ministerio del Ambiente y Agua. I am thankful to RALPH PETERS and VERA RDUCH (both ZFMK) for their multiple contributions to the GBOL III: Dark Taxa project, and to BJÖRN MÜLLER (ZFMK) for his help during the DNA barcoding process. I extend my gratitude to JANA THORMANN and BJÖRN RULIK (both ZFMK) for their help uploading the sequences to BOLD. I wish to extend my special thanks to ISABEL KILIAN (ZFMK) for collecting the specimens that resulted in the description of the new species. Finally, I thank XIMO MENGUAL (ZFMK), GUNNAR M. KVIFTE (Nord University), DANIEL WHITMORE (Staatliches Museum für Naturkunde Stuttgart) and two anonymous reviewers for their comments, which substantially improved the manuscript.

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ZooBank registration: <https://zoobank.org/References/C26DC4E7-1E81-437F-BAC5-22FC199713AB>

Manuscript received: 02.XI.2022; accepted: 13.VI.2023.

