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Diversity of *Lygistorrhina* (*Probolaeus*)  
Williston, 1896 (Diptera: Keroplatidae,  
*Lygistorrhinae*) of Mitaraka (French  
Guiana), with descriptions  
of three new species

Vladimir BLAGODEROV & Marc POLLET





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View: hill top site along trail C, 24 February 2015 (photo: Marc Pollet). In medallion: Habitus of *Lygistorrhina mitarakensis* n. sp. (photo: Vladimir Blagoderov).

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# Diversity of *Lygistorrhina* (*Probolaeus*) Williston, 1896 (Diptera: Keroplatidae, Lygistorrhinae) of Mitaraka (French Guiana), with descriptions of three new species

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## ABSTRACT

Three new species of *Lygistorrhina* (*Probolaeus*) Williston, 1896 are described, *Lygistorrhina maculipennis* n. sp., *Lygistorrhina conica* n. sp. and *Lygistorrhina mitarakensis* n. sp. The material was collected mainly by Malaise and SLAM traps in Mitaraka, southwestern French Guiana, during the “Our Planet Revisited” expedition in 2015. *Lygistorrhina cerqueirai* Lane, 1958 and *Lygistorrhina urichi* Edwards, 1912 are illustrated and their descriptions emended. A key to all known species of the subgenus *Lygistorrhina* (*Probolaeus*) is provided.

## RÉSUMÉ

*Diversité de Lygistorrhina* (*Probolaeus*) Williston, 1896 du Mitaraka (Guyane), avec la description de trois nouvelles espèces.

Trois nouvelles espèces de *Lygistorrhina* (*Probolaeus*) Williston, 1896 sont décrites, *Lygistorrhina maculipennis* n. sp., *Lygistorrhina conica* n. sp. et *Lygistorrhina mitarakensis* n. sp. Le matériel a été collecté principalement avec des pièges Malaise et SLAM sur le massif du Mitaraka, dans le sud-ouest de la Guyane, pendant l’expédition “La Planète Revisitée” en 2015. *Lygistorrhina cerqueirai* Lane, 1958 et *Lygistorrhina urichi* Edwards, 1912 sont illustrées et leur description émanée. Une clé de toutes les espèces connues du sous-genre *Lygistorrhina* (*Probolaeus*) est fournie.

## KEY WORDS

Neotropics,  
Amazon basin,  
Malaise trap,  
SLAM,  
fungus gnats,  
new species.

## MOTS CLÉS

région néotropical,  
bassin amazonien,  
piège Malaise,  
SLAM,  
mycétophilides,  
espèces nouvelles.

## INTRODUCTION

Lygistorrhinae Edwards, 1925 is a small group of fungus gnats comprising 51 species in 16 genera, of which 16 are fossil species in nine genera and 35 extant species in eight genera (Blagoderov 2020). They are distributed globally in tropical and warm-temperate areas but quite rare in collections, despite being sometimes very abundant in the field (Thompson 1975; Bertone 2018). Their biology is practically unknown, except for some field observations of nectarophagy and possible pollination (Bertone 2018). Although previously lygistorrhinids were considered as a separate family in the superfamily Sciaroidea Billberg, 1820, the latest molecular phylogenetic study presented the group as a well-supported monophyletic clade within Kero-platidae Rondani, 1856 (Mantić *et al.* 2020).

The largest genus of the subfamily, *Lygistorrhina* Skuse, 1890, is subdivided into two subgenera, *L. (Lygistorrhina)* with 14 described species and *L. (Probolaus)* Williston, 1896, with 11 known species, distributed in the Old World and New World respectively (Grimaldi & Blagoderov 2001). Both subgenera have fossil representatives in the early Eocene Cambay amber from India and the Miocene amber from the Dominican Republic (Grund 2012; Stebner *et al.* 2017). Although the monophyly of the nominative subgenus is not confirmed, *L. (Probolaus)* seems to be monophyletic with all species of the subgenus sharing one apomorphic feature, i.e., the absence of the external mid tibial spur. Unfortunately, not all species, particularly those described before 1975, are sufficiently known; material of those was rather limited, and some, for example *L. barretto* Lane, 1947 and *L. edwardsi* Lane, 1947, were described on the basis of female specimens only. Moreover, since *Lygistorrhina* demonstrates significant sexual dimorphism (Thompson 1975), species identification and delimitation in this genus remains very difficult. Descriptions of new species in this genus from recent years (Huerta & Ibanez-Bernal 2008; Huerta *et al.* 2019) suggest that the group might be megadiverse.

In 2015, the “Our Planet Reviewed” or “La Planète revisitée” Guyane 2014–2015 expedition, also known as the “Mitaraka 2015 survey”, was conducted in French Guiana as the 5<sup>th</sup> edition of a large scale biodiversity survey undertaken by the Muséum national d’Histoire naturelle in Paris and the NGO Pro-Natura international (both in France) (Pollet *et al.* 2014, 2015, 2018; Pascal *et al.* 2015; Touroult *et al.* 2018). Basic arthropod taxonomy and species discovery were at the heart of the survey, although forest ecology and biodiversity distribution modelling were also project topics. The expedition was conducted in the Mitaraka Mountains, a largely unknown and uninhabited area in the southwesternmost corner of French Guiana, directly bordering Surinam and Brazil (Krolow *et al.* 2017). It is part of the Tumuc Humac mountain chain, extending east in Amapá region (Brazil) and west in southern Surinam. The area consists primarily of tropical lowland rain forest with scattered inselbergs, i.e., isolated hills that stand above the forest plains.

The survey produced extensive material of most groups of Diptera, including three new species of *Lygistorrhina*, which are described in the present paper. In addition, type material of most of the species of the subgenus *Lygistorrhina (Probolaus)*

was studied and descriptions of *L. cerqueirai* Lane, 1958 and *L. urichi* Edwards, 1912 are emended below. A key to known species of *L. (Probolaus)* is provided as well.

## MATERIAL AND METHODS

Between 22 February and 27 March 2015, two consecutive equal-sized teams (of about 30 researchers) explored the Mitaraka area, including more than 10 invertebrate experts. A third smaller team returned to the site from 12 to 20 August 2015. Marc Pollet was coordinator of the collected Diptera and was also the only Diptera worker actively involved in the field work. Invertebrate sampling was carried out near the base camp, on the drop zone (an area near the base camp that had been clear-cut entirely to allow helicopters to land) and, in particular, along 4 trails of about 3.5 km that started from the base camp in four different directions (see Krolow *et al.* 2017; Touroult *et al.* 2018). During the first period (22 February–11 March 2015) over 21 different collecting methods were applied, with a total of 401 traps operational within a perimeter of 1 km<sup>2</sup>. This array consisted primarily of pan traps (n=280), Charax butterfly traps (n=50), square Malaise traps (SLAM) (n=32), Flight Intercept Traps (FIT, n=13) and Butterfly banana traps (n=12), but also a light trap (Pollet *et al.* 2018; Touroult *et al.* 2018). In the subsequent periods, pan traps were no longer operational. A total of 223 invertebrate samples (often pooled yields of different traps of the same type) were examined, including 94 sweep net samples. Fourteen samples, most collected with the 6 m Malaise trap (n=6) and SLAMs (n=5), contained 81 lygistorrhines. Of the pan traps employed, only blue traps produced two specimens. None of the sweep net samples comprised any specimens due to the fact that Marc Pollet focused mainly on capturing Dolichopodidae during active collecting.

The collected material was stored in 70% alcohol immediately after being collected. Some specimens were later dissected and mounted on microscopic slides in Euparal and Dimethyl Hydantoin Formaldehyde (DMHF). An attempt to extract DNA from several specimens was unsuccessful. Images were taken using Leica 205C stereomicroscope with Canon EOS 7D camera attached and Olympus BX51 compound microscope with Olympus UC30 camera; extended depth of field images was obtained with Helicon Focus v. 6 software. Morphological nomenclature is based on Grimaldi & Blagoderov (2001), Hippa *et al.* (2005) and Cumming & Wood (2017); wing venation nomenclature is also according to Cumming & Wood (2017). Measurements (in millimetres), when multiple specimens are available, are provided in Table 1 as Range, Min-Max, followed by Mean and measurement for holotype in square brackets.

All specimens of the three newly described species originated from the Mitaraka survey, in southernmost French Guiana. This information is not repeated in the species records of the paratypes, with usually the following format: number of specimens per sex; site code (description); latitude, longitude; altitude; collecting period/date; collecting method; name of collector; sample code; preservation method; specimen collection code; repository. Site codes are explained in Pollet *et al.* (2018). All the



FIG. 1. — Habitus of new species of *Lygistorrhina* (*Probolaeus*) Williston, 1896: **A**, *Lygistorrhina maculipennis* n. sp., with detached head in separate box; paratype MNHN-ED-ED10673; **B**, *Lygistorrhina conica* n. sp., paratype NMS-10003795; **C**, *Lygistorrhina mitarakensis* n. sp., paratype NMS-10003799. Scale bar: 1 mm.

specimens collected benefited from the Access and benefit sharing agreement of the Our Planet Reviewed program (APA 973-1).

#### ABBREVIATIONS

##### Collecting method

MT(6 m) Malaise trap with length of 6 meters;  
SLAM Sea, land and air malaise trap.

##### Morphology

C costal vein;  
CuA cubital anterior vein;  
M<sub>1</sub> first medial vein;  
M<sub>2</sub> second medial vein;  
M<sub>3+4</sub> third/fourth medial vein;  
R<sub>1</sub> first radial vein;  
R<sub>5</sub> fifth radial vein;  
Sc subcostal vein.

##### Repositories

CUMZ Cambridge University, Museum of Zoology, Cambridge;  
MNHN Muséum national d'Histoire naturelle, Paris;  
MZUSP Museu de Zoologia, Universidade de São Paulo, São Paulo;  
NHMUK Natural History Museum, London;  
NMS National Museums Scotland, Edinburgh.

#### SYSTEMATICS

Family KEROPLATIDAE Rondani, 1856  
Subfamily LYGISTORRHININAE Edwards, 1925

Genus *Lygistorrhina* Skuse, 1890

TYPE SPECIES. — *Lygistorrhina insignis* Skuse, 1890: 600.

Subgenus *Lygistorrhina* (*Probolaeus*) Williston, 1896.

TYPE SPECIES. — *Probolaeus singularis* Williston, 1896: 261.

*Lygistorrhina* (*Probolaeus*) *maculipennis* n. sp.  
(Figs 1A, 2A, 3A, B, 4A, B, Table 1)

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TYPE MATERIAL. — **Holotype**. French Guiana • ♂; Mitaraka, different sites nr base camp and along trails; tropical moist forest (different sites); 14.III.2015; SLAM; Julien Touroult & Eddy Poirier leg.; MITARAKA/191; mounted in Euparal on slide; MNHN-ED-ED10672; MNHN.

**Paratypes**. French Guiana • ♂; same data as holotype; 25.III.2015; SLAM; Julien Touroult & Eddy Poirier leg.; MITARAKA/190; in alcohol; MNHN-ED-ED10673; MNHN • 1 ♂; nr MIT-A-RBF1 (river); 24.III.2015; MT(6 m); Julien Touroult & Eddy Poirier leg.; MITARAKA/193; in alcohol; NMS-10003793; NMS.

**DIAGNOSIS**. — Wing with a small dark spot at the tip; Sc ends free. Thorax brown with pronotum and dorsal part of katepisternum yellow; fore coxa yellow, mid and hind coxa brown; metepimeron longer than hind coxa. Abdominal tergites and sternites 1-6 with a pale-yellow apical band; gonostyli slightly expanded at the tip.

**ETYMOLOGY**. — The name of this species refers to its infusate wing apex.

#### DESCRIPTION

##### Male

**General coloration**. Dark brown (Fig. 1A); measurements, see Table 1.

**Head**. Rounded, dichoptic. Vertex slightly convex. Rounded ommatidia with subequal diameter, interocular setae subequal to ommatidial diameter. Three ocelli arranged almost in a



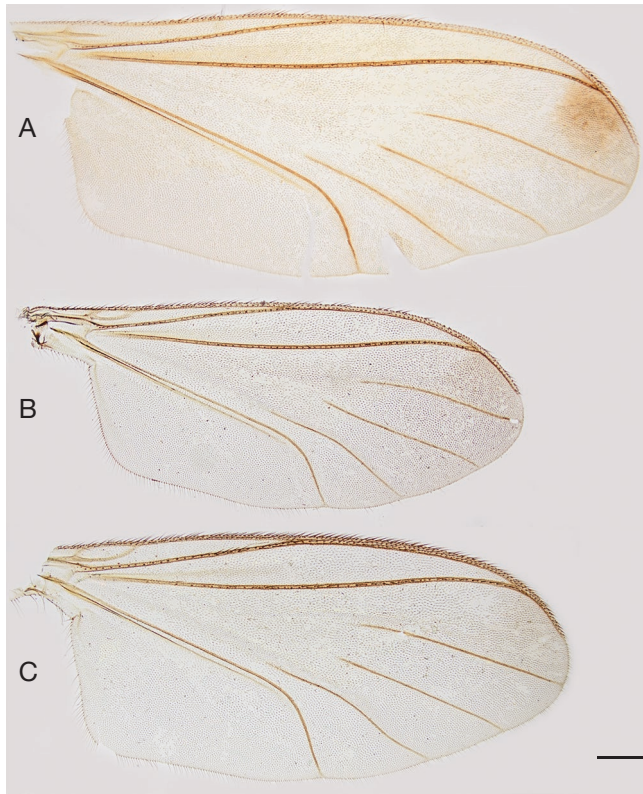


FIG. 2. — Wings of new species of *Lygistorrhina* (*Probolaeus*) Williston, 1896. **A**, *Lygistorrhina maculipennis* n. sp., holotype MNHN-ED-ED10672; **B**, *Lygistorrhina conica* n. sp., paratype MNHN-ED-ED10681; **C**, *Lygistorrhina mitarakensis* n. sp., holotype MNHN-ED-ED10691. Scale bar: 0.2 mm.

straight line, with median ocellus slightly anterior of two lateral ocelli. Antenna light brown, flagellomeres 1-6 yellow (Fig. 1A); antenna shorter than hind femur, length: 0.9-1.1 mm; 14 flagellomeres, with strong dorsal setae (flagellomeres 1-6: one pair, flagellomeres 7-14: two pairs), subcylindrical, gradually tapering; length of flagellomeres 1-6 1.3-1.6× their width, and 2-4× in flagellomeres 7-14. Face longer than wide. Clypeus with straight ventral edge. Palpus length 1.1 mm, tapering towards apex, with a simple row of long setae. Hypopharynx length 1.5 mm; labellum slightly longer.

**Thorax.** Brown, with anteprepronotum, prosternum, anepimeron, dorsal parts of anepisternum and katepisternum, and anterodistal part of mesonotum yellow. Scutum irregularly setose. Scutellum rounded, with 4-6 posterior setae. Anteprepronotum and proepisternum with 4-5 setae each. Laterotergite lobed, with a row of 13-14 setae. Hind coxa shorter than both metepimeron and laterotergite.

**Legs.** Fore coxa pale-yellow, in some specimens with dark brown basal part; mid and hind coxa completely dark brown, mid coxa with indistinct light brown spot in the centre. Remainder of fore and mid legs yellow; hind femur pale-yellow basally, dark brown in distal 1/3-1/2; hind tibia brown in distal half; hind tarsus brown. Tibiae irregularly setose. Claws of fore and

mid legs curved, apically blunt, with a small incision at apex; claw of hind leg setiform, straight and pointed.

**Wing** (Fig. 2A). Membrane hyaline, densely covered with microtrichia; with indistinctly rounded infusate area at the tip posterior of  $R_5$ , not reaching  $M_1$ . Sc short, ending free;  $R_1$  at the level of CuA tip; C extending to half the distance between tips of  $R_5$  and  $M_1$ . C,  $R_1$  and  $R_5$  with dark setae, rest of the veins bare;  $M_1$ ,  $M_2$  and  $M_{3+4}$  almost straight; CuA curved posteriorly, sometimes inconspicuously sinusoid. Anal lobe well developed. Halter white, same length as first abdominal segment.

**Abdomen.** Dark brown. Segments 1-6 with pale-yellow transversal band in posterior part, occupying 20-30% of segment length; last two segments and terminalia entirely dark brown.

**Terminalia** (Figs 3A, B, 4A, B). Tergite 9 parallel-sided, apex somewhat rounded, with dense unequal short spines. Sternite 9 oval, parallel-sided, with evenly rounded tip. Gonostyli expanded at tip; apical tooth scoop-shaped. Aedeagal complex (tegmen *sensu* Huerta *et al.* 2019) moderately sclerotised, rounded, wider than long. Aedeagal apodeme short, directed laterally, not extending width of aedeagal complex. Spines on 9<sup>th</sup> tergite and setae on gonostylus gradually denser towards apex.

#### COMPARISON

Free Sc is present as well in *L. brasiliensis* Edwards, 1932, *L. barretto* Lane, 1947, *L. sanctaecatherinae* Thompson, 1975, and *L. cerqueirai* Lane, 1958. However, the first three species have wings completely hyaline, without infuscation, and also differ from *L. maculipennis* n. sp. in the pattern of abdominal markings. *L. cerqueirai* has abdominal segments 1-6 with pale apical bands similar as the new species, but has a very different pattern of wing spots. In addition, *L. cerqueirai* has a single dorsal seta on each flagellomere, Costa extends to almost 2/3 of distance between  $R_5$  and  $M_1$ , laterotergite and metepimeron are shorter than hind coxa, and  $M_1$  and CuA are conspicuously bent anteriorly and sinusoid.

#### REMARK

Specimen NMS-10003793 is darker, with thorax and coxae completely dark brown except for extreme tip of fore coxa; abdominal segment 6 has a slightly lighter coloration than the rest of the segment, but the genitalia are of the same structure as the other specimens included in the type series.

*Lygistorrhina* (*Probolaeus*) *conica* n. sp.  
(Figs 1B, 2B, 3C, D, 4C, D, Table 1)

<https://doi.org/10.21203/rs.3.rs-10003793/v1>

**TYPE MATERIAL.** — **Holotype.** French Guiana • ♂; Mitaraka, different sites nr base camp and along trails, tropical moist forest (different sites); 14.III.2015; SLAM; Julien Touroult & Eddy Poirier leg.; MITARAKA/191; mounted in Euparal on slide; MNHN-ED-ED10674; MNHN.

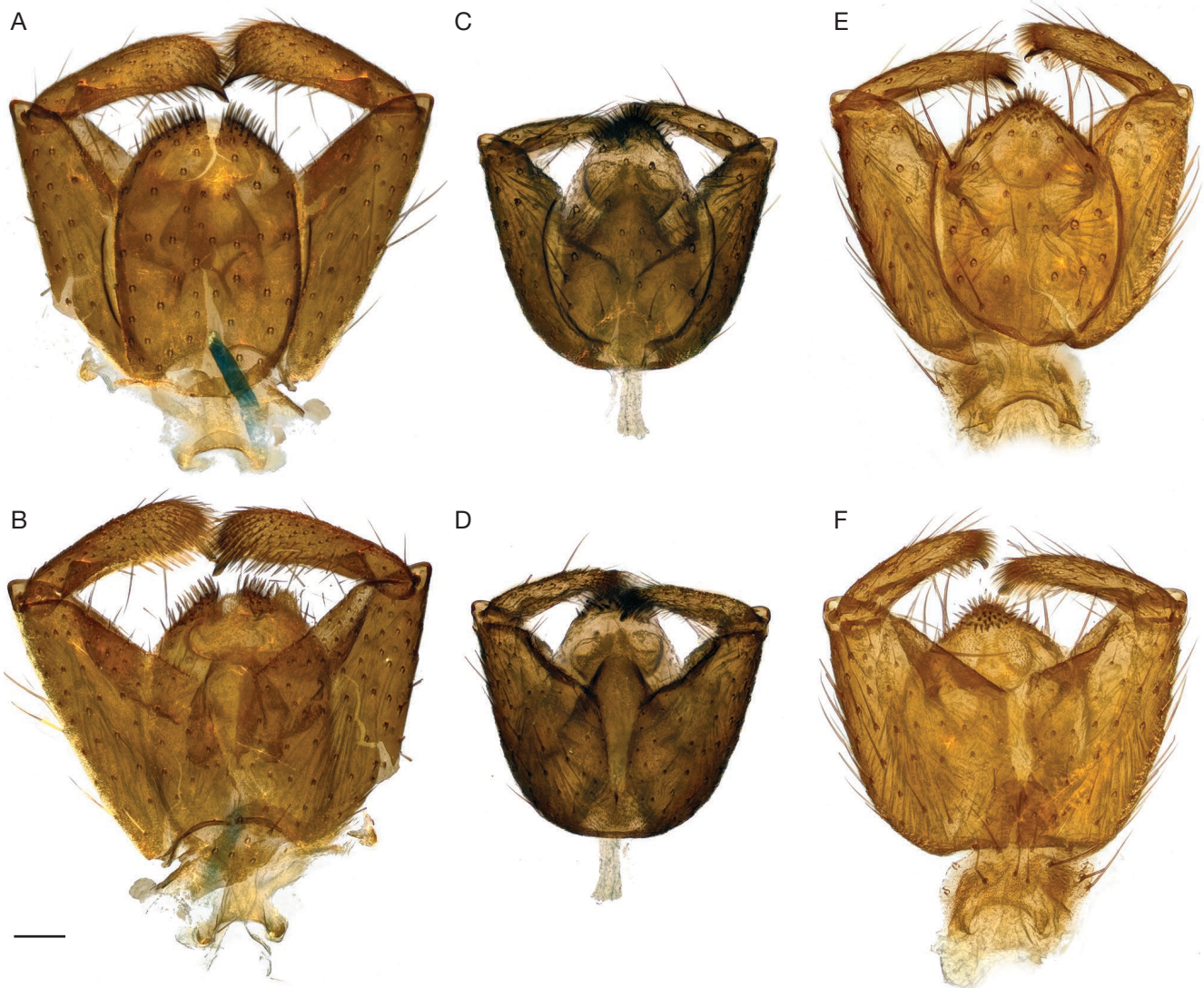


FIG. 3. — Male genitalia of new species of *Lygistorrhina* (*Probolaus*) Williston, 1896: **A, B**, *Lygistorrhina maculipennis* n. sp., holotype [MNHN-ED-ED10672](#); **C, D**, *Lygistorrhina conica* n. sp., holotype [MNHN-ED-ED10674](#); **E, F**, *Lygistorrhina mitarakensis* n. sp., [MNHN-ED-ED10691](#); **A, C, E**, dorsal view; **B, D, F**, ventral view. Scale bar: 50  $\mu$ m.

**Paratypes. French Guiana** • 6  $\sigma$ ; same data as holotype; in alcohol; [MNHN-ED-ED10675](#), [MNHN-ED-ED10676](#), [MNHN-ED-ED10677](#), [MNHN-ED-ED10678](#), [MNHN-ED-ED10679](#), [MNHN-ED-ED10680](#); MNHN • 4  $\sigma$ ; nr MIT-A-RBF1 (river); 24.III.2015; MT(6 m); Julien Touroult & Eddy Poirier leg.; MITARAKA/193; in alcohol (genitalia of one 1  $\sigma$  mounted in DMHF); [MNHN-ED-ED10681](#), [MNHN-ED-ED10682](#), [MNHN-ED-ED10683](#), [MNHN-ED-ED10684](#); MNHN • 1  $\sigma$ ; MIT-C-RBF2 (tropical wet forest, bas fond); 02°14'03.4"N, 54°26'53.0"W; 299 m; 2-10.III.2015; SLAM; leg. Marc Pollet; MITARAKA/138; in alcohol; [MNHN-ED-ED10685](#); MNHN • 1  $\sigma$ ; different sites nr base camp and along trails; tropical moist forest (different sites); 25.III.2015; SLAM; Julien Touroult & Eddy Poirier leg.; MITARAKA/190; in alcohol; [MNHN-ED-ED10686](#); MNHN • 1  $\sigma$ ; different sites nr base camp and along trails; tropical moist forest (different sites); 1.III.2015; SLAM; Julien Touroult & Eddy Poirier leg.; MITARAKA/194; in alcohol; [MNHN-ED-ED10687](#); MNHN • 3  $\sigma$ ; MIT-E-savane roche 2 (open/partially opened areas); 02°13'59.8"N, 54°27'46.5"W; 471 m; 13-20.VIII.2015; MT(6 m); leg. Pierre-Henri Dalens; MITARAKA/230; in alcohol; [MNHN-ED-ED10688](#), [MNHN-ED-ED10689](#), [MNHN-ED-ED10690](#); MNHN • 1  $\sigma$ ;

different sites nr base camp and along trails; tropical moist forest (different sites); 14.III.2015; SLAM; Julien Touroult & Eddy Poirier leg.; MITARAKA/191; in alcohol; NMS-10003794; NMS • 1  $\sigma$ ; nr MIT-A-RBF1 (river); 24.III.2015; MT(6 m); Julien Touroult & Eddy Poirier leg.; MITARAKA/193; in alcohol; NMS-10003795; NMS • 1  $\sigma$ ; nr MIT-A-RBF1 (river); 25.III.2015; MT(6 m); Julien Touroult & Eddy Poirier leg.; MITARAKA/189; in alcohol; NMS-10003796; NMS • 1  $\sigma$ ; MIT-E-savane roche 2 (open/partially opened areas); 02°13'59.8"N, 54°27'46.5"W; 471 m; 13-20.VIII.2015; MT(6 m); Pierre-Henri Dalens leg.; MITARAKA/230; in alcohol; NMS-10003797; NMS.

**DIAGNOSIS.** — Small dark fly. Wing hyaline, Sc ends at C, C extends beyond  $\frac{1}{2}$  distance between  $R_5$  and  $M_1$ ;  $M_1$  sinuous. Tergite 1 brown, abdominal segments 2-4 with pale apical fringe. Tergite 9 ovoid, with very dense patch of equal spines concentrated on apex; gonostyli tapering, with dense patch of apical setae as long as apical tooth.

**ETYMOLOGY.** — The species epithet refers to the shape of its aedeagal complex, and should be considered as an adjective.



TABLE 1. — Measurements (length, in millimetres) for new species of *Lygistorrhina* (*Probolaeus*) Williston, 1896.

Species	<i>Lygistorrhina maculipennis</i> n. sp.	<i>Lygistorrhina conica</i> n. sp.	<i>Lygistorrhina mitarakensis</i> n. sp.
# of specimens	3	18	34
Sex	M	M	M
Total	5.45–6.45, 6.1 [6.45]	2.9–4.45, 3.8 [3.55]	3.5–5.7, 4.11 [4.1]
Wing	2.7–3.0, 2.85 [2.8]	1.6–2.2, 2.0 [2.1]	1.95–2.75, 2.24 [2.25]
Coxa 1	0.75–0.85, 0.82 [0.85]	0.44–0.56, 0.5 [0.48]	0.42–0.62, 0.51 [0.54]
Coxa 2	0.65–0.7, 0.68 [0.7]	0.38–0.5, 0.42 [0.38]	0.34–0.54, 0.42 [0.42]
Coxa 3	0.4–0.5, 0.45 [0.4]	0.26–0.4, 0.3 [0.26]	0.26–0.4, 0.31 [0.3]
Femur 1	[1.15]	0.56–0.84, 0.7 [0.68]	0.6–0.86, 0.69 [0.76]
Femur 2	1.35	0.76–0.94, 0.82 [0.76]	0.64–0.9, 0.79 [0.84]
Femur 3	1.4	1.0–1.44, 1.11 [1.06]	0.84–1.52, 1.14 [1.22]
Tibia 1	[1.3]	0.6–1, 0.73 [0.66]	0.56–0.94, 0.75 [0.7]
Tibia 2	1.95	0.76–1.08, 0.96 [0.96]	0.82–1.08, 0.95 [1.0]
Tibia 3	2.1	1.4–1.7, 1.53 [1.52]	1.4–2, 1.62 [1.78]
Basitarsus 1	[1.7]	0.5–0.8, 0.7 [0.74]	0.46–0.84, 0.64 [0.46]
Basitarsus 2	1.75	0.7–1.1, 0.89	0.54–0.94, 0.72 [0.63]
Basitarsus 3	1.1	0.7–0.96, 0.8 [0.8]	0.62–0.98, 0.8, [0.9]

## DESCRIPTION

*Male*

**General coloration.** Dark brown (Fig. 1B).

**Measurements.** See Table 1.

**Head.** Rounded, dichoptic. Vertex slightly convex. Rounded ommatidia with similar diameter, interocular setae length subequal to ommatidium diameter. Three ocelli arranged on a straight line, with median ocellus round, half the diameter of lateral ocelli, and two semi-circular lateral ocelli, directed laterally. Antenna light brown at the basis, gradually becoming yellow towards apex (Fig. 1B); 14 flagellomeres, with strong dorsal setae (flagellomeres 1–3 with several irregular setae, and flagellomeres 4–13 with one pair), subcylindrical, gradually tapering, about 2× as long as wide. Face longer than wide. Clypeus rounded apically. Palpus length 0.9 mm, tapering towards apex, with a simple row of long setae. Hypopharynx length 1.3–1.5 mm; labellum slightly longer.

**Thorax.** Thorax brown. Scutum irregularly setose. Scutellum rounded, with 6–8 posterior setae. Antepronotum and proepisternum with 4–5 setae each. Laterotergite lobed, with a row of 10–14 setae. Hind coxa slightly longer than either metepimeron or laterotergite.

**Legs.** Fore coxa pale-yellow; mid and hind coxa completely dark brown. Remainder of fore and mid legs entirely yellow; hind femur pale-yellow basally, dark brown in distal 1/2; hind tibia light brown basally, dark brown in distal 1/2; hind tarsus entirely brown. Tibiae irregularly setose. Dorsal setae of hind tibia shorter than tibial width. Claws of fore and mid legs curved, apically blunt, with a small incision at apex; claw of hind leg setiform, straight and pointed.

**Wing (Fig. 2B).** Membrane hyaline, densely covered with microtrichia; slightly darker at the tip posterior of  $R_5$ . Sc short, ending on C;  $R_1$  slightly proximad the level of CuA tip;

C extending to more than half the distance between tips of  $R_5$  and  $M_1$ ; C,  $R_1$  and  $R_5$  with dark setae, remaining veins bare;  $M_1$ ,  $M_2$ , and  $M_{3+4}$  approximately equal in length.  $M_1$  bent anteriorly, sinuous,  $M_2$  almost straight,  $M_{3+4}$  and CuA curved posteriorly. Anal lobe well developed. Halter yellow, as long as first abdominal segment.

**Abdomen.** Dark brown. Sternite 1 in some specimens with pale transversal posterior band; segments 2–4 with pale-yellow posterior band, occupying 20–25% of segment length; segments 5–7 and terminalia dark brown.

**Terminalia (Figs 3C, D; 4C, D).** Tergite 9 slightly ovoid, with very dense patch of spines of equal length concentrated on apex. Gonostyli tapering, as long as apical tooth, with dense patch of apical setae. Aedeagal complex heavily sclerotised, conical, with rounded apex. Aedeagal apodemes long, straight, unbranched, directed medially.

## COMPARISON

Although similar in coloration to *L. borkenti*, *L. conica* n. sp. differs from this species by the following features (character status in *L. borkenti* given in brackets): its smaller size (Table 1); a very light infusate wing apex, occupying area between  $R_5$  and  $M_1$  (distinct spot);  $M_1$  sinuous (curved posteriorly); fore coxa yellow except its very base (completely brown); aedeagus sclerotised, tapering, almost triangular (membranous, rounded).

*Lygistorrhina* (*Probolaeus*) *mitarakensis* n. sp.  
(Figs 1C, 2C, 3E, F, 4E, F, Table 1)

[url:lsid:zoobank.org:act:20F21808-86A0-4AB8-A10A-75050FD763C9](https://doi.org/10.21203/rs.3.rs-1000000/v1)

**TYPE MATERIAL.** — **Holotype.** French Guiana • ♂; Mitaraka, different sites nr base camp and along trails; tropical moist forest (different sites); 14.III.2015; Julien Touroult & Eddy Poirier leg.; SLAM; MITARAKA/191; mounted in Euparal on slide; MNHN-ED-ED10691; MNHN.



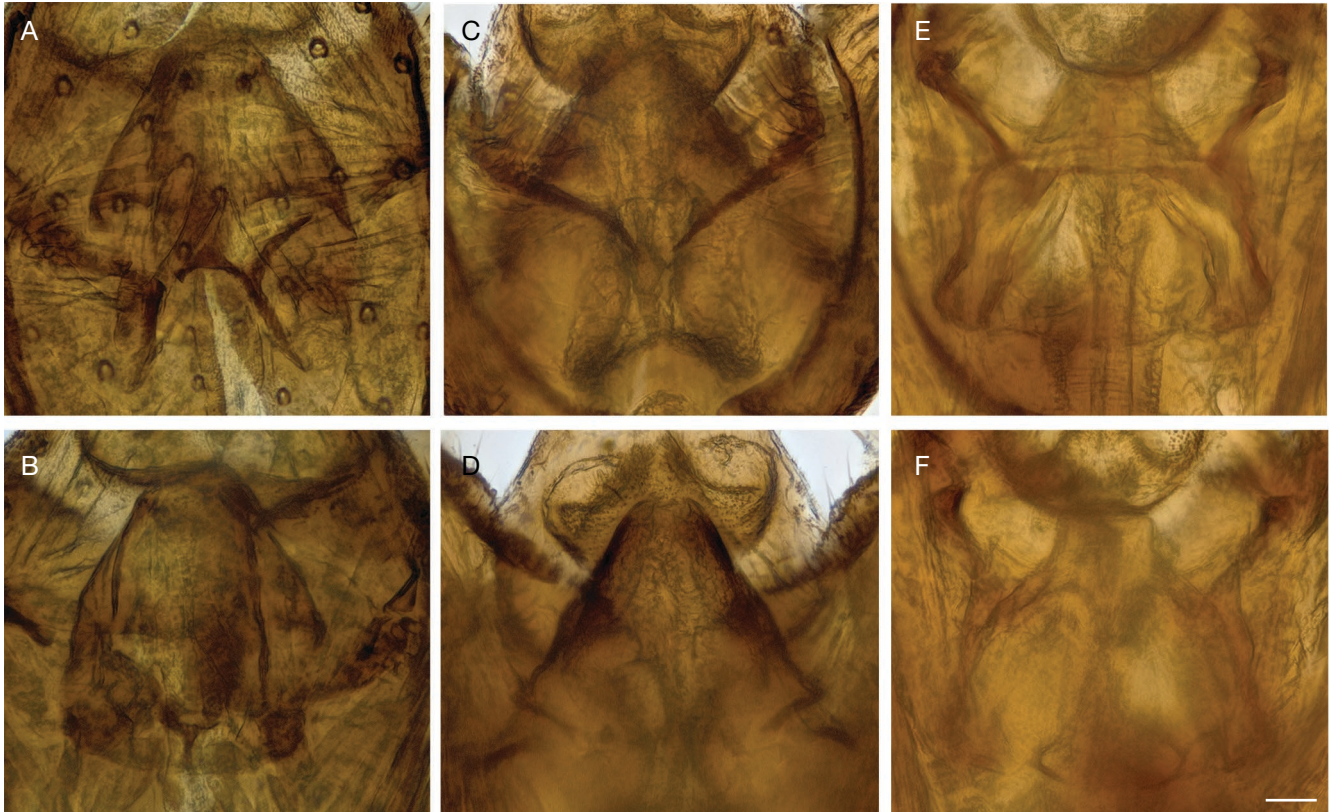


FIG. 4. — Aedeagal complex of new species of *Lygistorrhina* (*Probolaeus*) Williston, 1896: **A, B**, *Lygistorrhina maculipennis* n. sp., holotype MNHN-ED-ED10672; **C, D**, *Lygistorrhina conica* n. sp., holotype MNHN-ED-ED10674; **E, F**, *Lygistorrhina mitarakensis* n. sp., holotype MNHN-ED-ED10691; **A, C, E**, dorsal view; **B, D, F**, ventral view. Scale bar: 20  $\mu$ m.

**Paratypes. French Guiana** • 26 ♂; different sites nr base camp and along trails; tropical moist forest (different sites); 14.III.2015; Julien Touroult & Eddy Poirier leg.; SLAM; MITARAKA/191; in alcohol; MNHN-ED-ED10694 mounted in DMHF on slide; MNHN-ED-ED10691, ED10692, ED10693, ED10694, ED10695, ED10696, ED10697, ED10698, ED10699, ED10700, ED10701, ED10702, ED10703, ED10704, ED10705, ED10706, ED10707, ED10708, ED10709, ED10710, ED10711, ED10712, ED10713, ED10714, ED10715, ED10716, ED10722; MNHN • 1 ♂; different sites nr base camp and along trails; tropical moist forest (different sites); 1-6.III.2015; SLAM; Julien Touroult & Eddy Poirier leg.; MITARAKA/195; in alcohol; MNHN-ED-ED10717; MNHN • 1 ♂; different sites nr base camp and along trails; tropical moist forest (different sites); 25.III.2015; SLAM; Julien Touroult & Eddy Poirier leg.; MITARAKA/190; in alcohol; MNHN-ED-ED10718; MNHN • 2 ♂; MIT-DZ2 (tropical moist forest, plateau-slope); 02°14'02.6"N, 54°27'01.7"W; 296 m; 20-25.III.2015; SLAM; Julien Touroult & Eddy Poirier leg.; MITARAKA/206; in alcohol; MNHN-ED-ED10719, MNHN-ED-ED10721; MNHN • 1 ♂; MIT-DZ1 (tropical moist forest, plateau-slope); 02°14'01.4"N, 54°27'00.2"W; 304 m; 1-8.III.2015; SLAM; leg. Marc Pollet; MITARAKA/169; in alcohol; MNHN-ED-ED10720; MNHN • 2 ♂; MIT-DZ2 (tropical moist forest, plateau-slope); 02°14'02.6"N, 54°27'01.7"W; 296 m; 1-8.III.2015; SLAM; leg. Marc Pollet; MITARAKA/173; in alcohol; NMS-10003798, NMS-10003799; NMS • 1 ♂; MIT-E-savane roche 2 (open/partially opened areas); 02°13'59.8"N, 54°27'46.5"W; 471 m; 13-20.VIII.2015; MT(6 m); leg. Pierre-Henri Dalens; MITARAKA/230; in alcohol; NMS-10003800; NMS • 3 ♂; different sites nr base camp and along trails; tropical moist forest (different sites); 14.III.2015; SLAM; Julien Touroult & Eddy Poirier leg.; MITARAKA/191; in alcohol; NMS-10003801-NMS-10003803; NMS.

**DIAGNOSIS.** — Antenna entirely yellow, flagellomeres each with one prominent dorsal seta. Wing hyaline; Sc ends at C. Thorax, mid and hind coxa brown; fore coxa yellow. Abdominal segment 1 completely pale-yellow; segments 2-5 with a pale-yellow posterior band; gonostyli of subequal width along their entire course; aedeagus membranous.

**ETYMOLOGY.** — The species epithet refers to the type locality.

#### DESCRIPTION

##### Male

**General coloration.** Dark brown (Fig. 1C).

**Measurements.** See Table 1.

**Head.** Rounded, dichoptic. Eyes appear larger than head height, with vertex appearing slightly depressed. Rounded ommatidia with similar diameter, interocular setae subequal in length to ommatidial diameter. Three ocelli arranged almost on a straight line, semi-circular, with median ocellus 1/3× the size of lateral ocelli. Antenna yellow, strongly tapering, shorter than hind femur, with length 0.8-0.9 mm; 14 flagellomeres, each with one strong dorsal seta, subcylindrical, gradually tapering; flagellomeres 1-9 1.5-2× as long as wide, flagellomeres 10-14 2-4× as long as wide. Face longer than wide. Clypeus with rounded apex, setose. Palpus length 1.2 mm, tapering towards apex, with a simple row of very long setae (at least 5× as long as palpus width). Hypopharynx length 1.5 mm; labellum slightly longer.

**Thorax.** Thorax completely brown. Scutum irregularly setose. Scutellum rounded, with 6 posterior setae. Anteprenotum and proepisternum with 4-5 setae each. Laterotergite lobed, with one row of 14-20 setae. Hind coxa slightly longer than both metepimeron and laterotergite.

**Legs.** Fore coxa pale-yellow; mid and hind coxa completely dark brown. Remainder of fore and mid legs yellow; hind femur pale-yellow basally, dark brown in distal 1/3; hind tibia brown in distal 1/4; hind tarsus entirely brown. Tibiae irregularly setose. Dorsal setae of hind tibia longer than tibial width. Fore basitarsus 1 shorter than fore tibia. Claws of fore and mid leg curved, apically blunt, with a small incision at apex; claw of hind legs setiform, straight and pointed.

**Wing (Fig. 1C).** Membrane hyaline, densely covered with microtrichia. Sc relatively long, ending on C; R<sub>1</sub> slightly proximal to the level of CuA tip; C extending to half the distance between tips of R<sub>5</sub> and M<sub>1</sub>; C, R<sub>1</sub> and R<sub>5</sub> with dark setae, rest of the veins bare; M<sub>1</sub> longer than M<sub>2</sub> and M<sub>3+4</sub>, latter subequal in length. M<sub>1</sub> and M<sub>2</sub> almost straight; M<sub>3+4</sub> curved anteriorly and slightly sinuous; CuA curved posteriorly, sometimes inconspicuously sinusoid. Anal lobe well developed. Halter yellow, with the same length as first abdominal segment.

**Abdomen.** Mainly dark brown. Segment 1 completely pale; segments 2-5 with pale-yellow posterior band, occupying 20-30% of segment length; segments 6 and 7 and terminalia dark brown (sometimes sternite 6 with inconspicuous pale apical spot).

**Terminalia (Figs 3E, F; 4E, F).** Tergite 9 rounded at base, somewhat triangular at tip, with very long setae near apex (longer than on remainder of tergite) and a patch of sparse short setae at tip. Sternite 9 with rectangular base wider than long and square extension caudally. Gonostyli of equal width, with very dense apical and medioventral patch of setae at tip; apical tooth scoop-shaped. Aedeagal complex membranous, bell-shaped with short parallel-sided apex; gonocoxal apodemes heavily sclerotised distally; aedeagal apodeme sinusoidally curved.

#### COMPARISON

The new species is most similar to *L. singularis* (Williston, 1896) but differs by (character status in *L. singularis* given in brackets): flagellomeres longer (0.7-1.3× as long as wide); Costa shorter, not extending half the distance between R<sub>5</sub> and M<sub>1</sub> (extending); M<sub>3+4</sub> curved anteriorly (posteriorly); fore basitarsus shorter than fore tibia (longer); abdominal segment 5 with pale apical band (completely brown).

*Lygistorrhina (Probolaeus) cerqueirai* Lane, 1958  
(Fig. 5)

*Lygistorrhina cerqueirai* Lane, 1958: 209.

#### REMARK

The species was described by J. Lane based on a single male specimen. During the senior author's visit to MZUSP in 2010, the holotype of the species was studied and photographed. Below is an emended description.

**MATERIAL EXAMINED.** — **Holotype.** Brazil • ♂; Amazonas, Igarapé de Mariano; 21.XI.1955; Elias and Rappe coll.; MZUSP.

#### EMENDED DESCRIPTION

##### Male

**Head.** Palpus  $\frac{3}{4}$ × as long as proboscis, rather densely setose; setae about as long as proboscis width. Median ocellus slightly anterior of lateral ocelli, half the diameter of the latter; lateral ocelli semicircular. Flagellomeres bacilliform, about 3× as long as wide, each with single prominent dark seta near apex.

**Thorax (Fig. 5C, D).** Laterotergite with 16-17 setae; mediotergite with 6 setae; anteprenotum with 5 setae; proepisternum with 6 setae. Mesonotum with short setae forming indistinct dorsocentral and acrostichal rows; with long setae in supra-alar rows and in posterior quarter of mesonotum.

**Wing (Fig. 5B).** Costa extends beyond R<sub>5</sub> to more than  $\frac{1}{2}$  of distance between R<sub>5</sub> and M<sub>1</sub>; M<sub>1</sub> and CuA sinuous.

**Terminalia (Fig. 5E, F).** Tergite 9 ovoid, entirely setose, with dense patch of strong spines on apex. Gonostyli parallel-sided, slightly curved inwards, with an apical scoop-shaped tooth. Gonocoxal apodemes heavily sclerotised.

*Lygistorrhina (Probolaeus) urichi* Edwards, 1912  
(Figs 6, 7)

*Lygistorrhina urichi* Edwards, 1912: 204.

#### REMARK

The species was described by F. W. Edwards based on a series of 8 specimens (7 males and one female), collected by H. Scott (Cambridge, UK). While the original paper specifies that the type was presented to the British Museum (now Natural History Museum, NHMUK, London), one male and one female specimen were located in this collection. Five male specimens were deposited in the collection of the Museum of Zoology, Cambridge University (UK). Edwards did not specify the gender of the type specimens deposited in the NHMUK, so the entire type series should be considered as syntypes. Discovery of additional syntypes allows us to emend Edward's description (only additional or misinterpreted characters being mentioned).

**MATERIAL EXAMINED.** — **Syntypes.** Trinidad • 1 ♂; Diego Martin; 22.III.1912; H. Scott, herbage by stream; BMNH(E)254352; NHMUK • 1 ♀; III.1912, H. Scott; BMNH(E)254351; NHMUK • 2 ♂; III.1912, H. Scott; CUMZ.



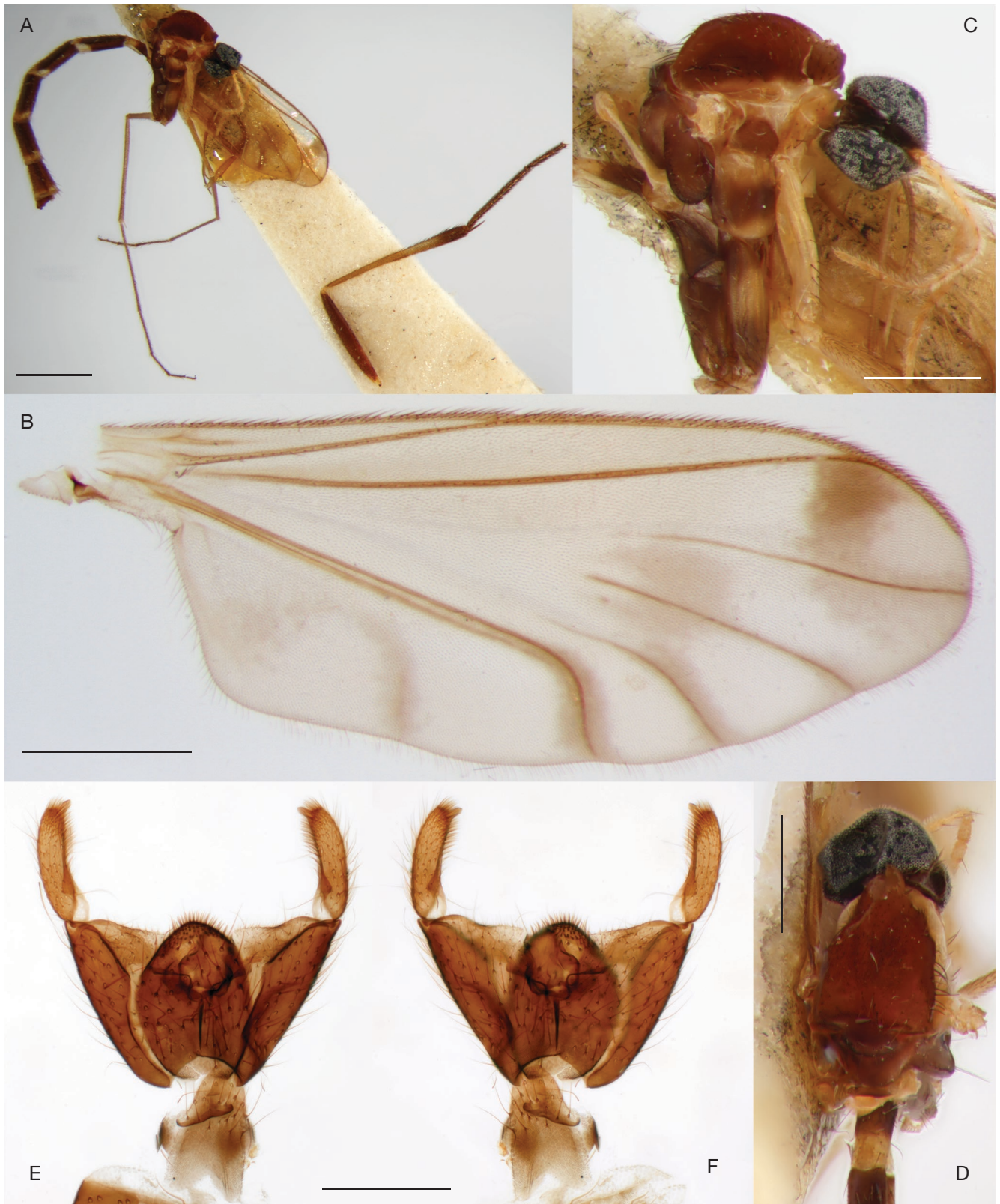


FIG. 5. — *Lygistorrhina (Probolaeus) cerqueirai* Lane, 1958, holotype: **A**, habitus; **B**, wing; **C**, **D**, thorax; **E**, **F**, male genitalia; **C**, lateral view; **D**, **E**, dorsal view; **F**, ventral view. Scale bars: A, 1 mm; B-D, 0.5 mm; E, F, 0.2 mm.



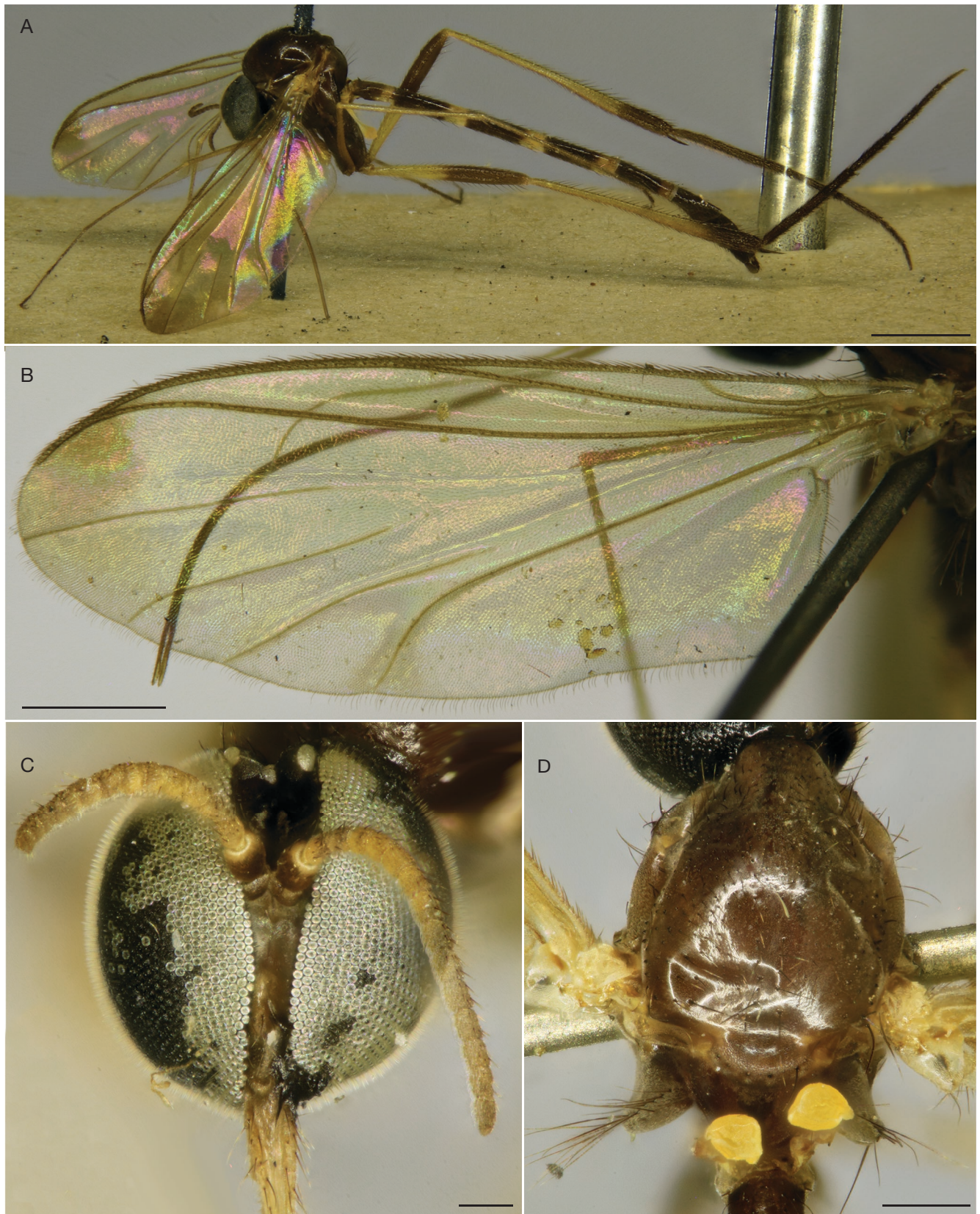


FIG. 6. — *Lygistorrhina (Probolaeus) urichi* Edwards, 1912, syntype male: **A**, habitus; **B**, wing; **C**, head; **D**, thorax, dorsal view. Scale bars: A, 1 mm; B, 0.5 mm; C, 0.1 mm; D, 0.2 mm.



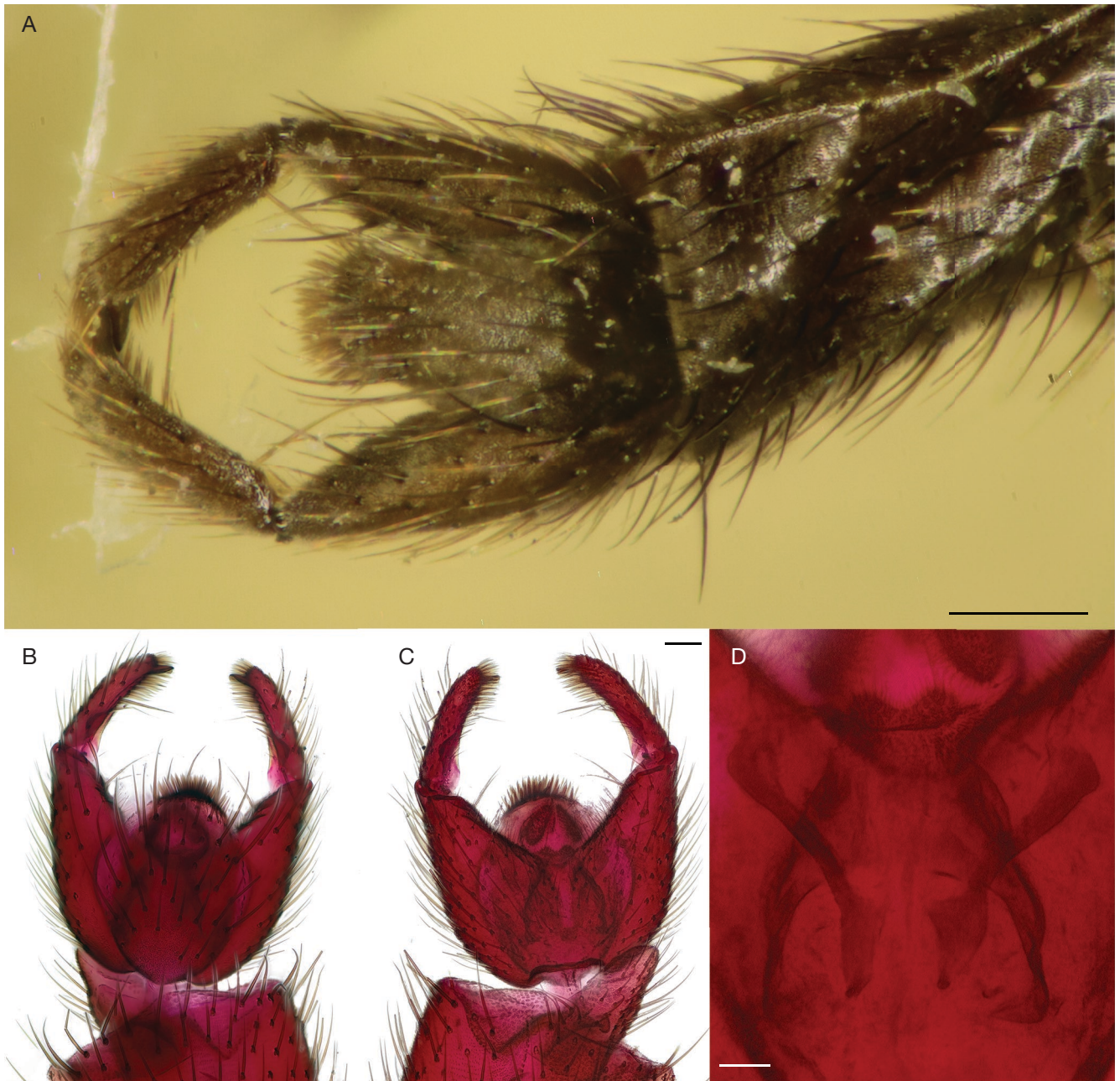


FIG. 7. — *Lygistorrhina* (*Probolaeus*) *urichi* Edwards, 1912, syntype male: **A–C**, genitalia; **D**, aedeagal complex; **A, B**, dorsal view; **C, D**, ventral view. Scale bars: A, 1 mm; B, C, 50 µm; D, 20 µm.

#### EMENDED DESCRIPTION

##### Male

**Head.** Scape and pedicel brown, flagellum uniformly pale, as long as thorax; flagellomeres as long as wide or slightly shorter, each with a few irregular dark dorsal setae. Median ocellus round,  $\frac{1}{2}\times$  the diameter of the lateral ocelli, latter semi-circular.

**Thorax.** Thorax and coxae uniformly dark brown. Antepronotum with three setae, proepisternum with six setae. Laterotergite with 18–19 setae, and mediotergite with 6–8 setae; laterotergite, metepisternum and hind coxa subequal in size. Mesonotum

irregularly setose except for posterior  $\frac{1}{3}$ , latter with indistinct dorsocentral rows of setae.

**Wing.**  $M_1$  straight;  $M_2$  and  $M_{3+4}$  curved and converging;  $M_{3+4}$  slightly and CuA conspicuously sinuous.

**Abdomen.** Tergite 1 of some specimens completely dark, but sternite 1 always with pale transversal posterior band.

**Terminalia** (Fig. 7). Male genitalia narrower than in other species. Tergite 9 oval, almost parallel-sided,  $1.5\times$  as long as

wide. Gonostyli appearing laterally flattened, slightly wider at apex; apical tooth scoop-shaped, with very dense patch of setae apico-medially. Aedeagus wider than long, bell-shaped, with flat apex; aedeagal apodeme long, curved; gonocoxal apodeme heavily sclerotised.

*Lygistorrhina (Probolaeus) barrettoi* Lane, 1947

*Lygistorrhina barrettoi* Lane, 1947: 346.

*Lygistorrhina coxata* Lane, 1947: 345, misidentification of *Manota coxata* (Enderlein, 1911).

REMARK

Lane (1947) described a female of *Lygistorrhina*, believing it to be a specimen of *Aphanizophleps coxata* Enderlein. However, the species described by Enderlein belongs to *Manota*. The name was treated as available by Papavero (1978), though it is a clear case of misidentification and was ignored subsequently (Oliveira & Amorim 2012). Examination of Lane's specimens in MZUSP by the senior author revealed that they were identical to *Lygistorrhina barrettoi* Lane, 1947, except for being slightly darker.

MATERIAL EXAMINED. — **Brazil** • 1 ♀; S. Paulo, Jaraguá; VIII.1945; J. Lane coll.; labelled "ALOTIPO"; MZUSP.

KEY TO EXTANT AND FOSSIL SPECIES OF SUBGENUS *LYGISTORRHINA (PROBOLAEUS)* WILLISTON 1896

1. Wing membrane entirely hyaline ..... 2
- Wing membrane with dark spots ..... 9
2. Sc ending free ..... 3
- Sc ending in C ..... 4
3. Antenna light brown; each flagellomere with two or more prominent dorsal setae; C extending to half distance between  $R_5$  and  $M_1$ ; coxa 1 pale with dark basal part; laterotergite with 13-14 setae; abdominal segments 2-5 (sometimes 2-4) with pale posterior band ..... *Lygistorrhina santaecatharinae* Thompson, 1975. [eastern USA: Alabama, Georgia, North Carolina, South Carolina, Tennessee, Virginia, West Virginia]
- Antenna with basal 4-5 flagellomeres pale yellow, remaining brown; each flagellomere with one prominent dorsal seta; fore coxa entirely yellow; laterotergite with 18-20 setae; abdominal segments 1-5 with pale posterior band ..... *Lygistorrhina brasiliensis* Edwards, 1932 [Brazil: São Paulo, Minas Gerais].
4. First abdominal tergite entirely dark (brown) (sometimes first sternite with narrow pale band) ..... 5
- First abdominal tergite pale or with pale posterior band ..... 6
5. Antenna light brown at base, gradually becoming yellow towards apex;  $M_1$  bent anteriorly and slightly sinusoid; fore coxa entirely pale; abdominal segments 2-4 with pale posterior band ..... *Lygistorrhina conica* n. sp. [French Guiana].
- Antenna entirely dark;  $M_1$  almost straight; fore coxa entirely dark; abdominal segments 2-5 with pale posterior band ..... *Lygistorrhina barrettoi* Lane, 1947 [Brazil: Goiás, Mato Grosso, Mato Grosso do Sul]
6. Abdominal segments 1-5 with pale posterior band ..... 7
- First abdominal tergite entirely pale and segments 2-4 with pale posterior band ..... 8
7. Flagellomeres 1-5 reddish brown, the rest dark brown; fore coxa entirely yellow; fore basitarsus longer than fore tibia; laterotergite with 13-14 setae;  $M_1$  bent posteriorly ..... *Lygistorrhina edwardsi* Lane, 1947 [Brazil: São Paulo, Minas Gerais].
- Antenna unicolourous (true colour unknown, amber inclusion); fore coxa entirely dark; fore basitarsus shorter than fore tibia; laterotergite with 9-10 setae;  $M_1$  bent anteriorly and sinusoid ..... *Lygistorrhina caribbiana* Grund, 2012 [fossil, Miocene: Dominican amber].
8. Costa extending to more than half distance between  $R_5$  and  $M_1$ ;  $M_{3+4}$  curved posteriorly; fore basitarsus longer than fore tibia; abdominal segments 1-4 with pale posterior band ..... *Lygistorrhina singularis* (Williston, 1896) [St. Vincent].
- Costa extending to half distance between  $R_5$  and  $M_1$ ;  $M_{3+4}$  curved anteriorly; fore basitarsus shorter than fore tibia; abdominal segments 1-5 with pale posterior band ..... *Lygistorrhina mitarakensis* n. sp. [French Guiana].
9. Sc ending free; segment 6 with pale posterior band (sometimes inconspicuous) ..... 10
- Sc ending in C; segment 6 entirely dark, without a pale posterior band ..... 11
10. Wing with pigmented spots in the middle of the wing and along veins;  $M_1$  bent anteriorly and sinusoid; C extending beyond half distance between  $R_5$  and  $M_1$  ..... *Lygistorrhina cerqueirai* Lane, 1958 [Brazil: Amazonas].



- Wing with pigmented spot at wing tip only;  $M_1$  bent posteriorly; C extending to half distance between  $R_5$  and  $M_1$  ..... *Lygistorrhina maculipennis* n. sp. [French Guiana].
- 11. Wing with triangular pigmented spot restricted to anterior part of cell  $R_5$  only ..... 12
- Wing with pigmented spot extending between  $R_1$  and  $M_2$  ..... 13
- 12. Abdominal segment 1 and 5 completely dark, segments 2-4 with pale posterior band;  $M_1$  and CuA bent posteriorly; laterotergite with 17 setae ..... *Lygistorrhina borkenti* Huerta & Ibanes-Bernal, 2008 [Mexico: Morelos].
- Abdominal segments 2-5 (and at least sternite of segment 1, but often also tergite 1) with pale posterior band;  $M_1$  almost straight, CuA conspicuously sinusoid; laterotergite with 18-20 setae ..... *Lygistorrhina urichi* Edwards, 1912 [Trinidad].
- 13. Mediotergite with a pair of pre-apical setae; fore coxa pale yellow with dark brown spot in centre ..... *Lygistorrhina acahuizotla* Huerta *et al.*, 2019 [Mexico: Guerrero].
- Mediotergite without setae; fore coxa completely pale yellow ..... *Lygistorrhina alexi* Huerta & Ibanes-Bernal, 2008 [Mexico: Jalisco, Guerrero, Yucatan].

## DISCUSSION

Lygistorrhinidae are usually considered to be quite rare flies, although they can occasionally be collected in large numbers (Thompson 1975). The examination of mass collected material produced by large-scale projects, such as the Mitaraka expedition, may reveal that lygistorrhinids are, in fact, more common and diverse than previously believed. Unfortunately, processing and sorting large numbers of samples (or large samples) is not a trivial task (Karlsson *et al.* 2020). As a result, many taxa often remain unavailable for further study. The phenomenon of multiple related co-existing species is very common in larger genera of fungus gnats and other insects and may be another evidence of hidden diversity within the group. It would be extremely interesting to re-visit the taxonomy of *Lygistorrhina sanctaecatharinae* Thompson, 1975, a species with a distribution range that apparently covers almost the entire eastern USA.

The best collecting method for Lygistorrhinidae is passive mass flight trapping, such as Malaise, SLAM, and flight interception traps. Surprisingly, compact traps, such as SLAM, produce very good results, probably because they can be installed in more secluded sites, such as understory, probably corresponding to lygistorrhinid microhabitats. Another interesting observation is the significant number of Lygistorrhinidae collected on rocky outcrops (“savane roche”) in Mitaraka, which might be due to the fact that a Malaise trap has been in operation here in August 2015. However, this also concurs with observations of the senior author in the Dominican Republic, where *Lygistorrhina* can often be collected along mossy stony walls. It is therefore possible that mosses and associated substrates are potential breeding habitats for lygistorrhinid larvae, but this should be further investigated.

## Acknowledgements

All specimens of the newly described Lygistorrhininae (Diptera) were collected during the “Our Planet Reviewed” Guyane-2015

expedition in the Mitaraka range, in the core area of the French Guiana Amazonian Park, organized by the MNHN and Pro-Natura international. The expedition was funded by the European Regional Development Fund (ERDF), the Conseil régional de Guyane, the Conseil général de Guyane, the Direction de l’Environnement, de l’Aménagement et du Logement and by the Ministère de l’Éducation nationale, de l’Enseignement supérieur et de la Recherche. It was realized in collaboration with the Parc amazonien de Guyane and the Société entomologique Antilles-Guyane. The junior author participated to this expedition as member of the first team (22 February-11 March 2015), hereby supported financially by MNHN and Pro-Natura international. Thanks are also due to the referees for their helpful comments and suggestions on an earlier draft of this paper.

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