











## Research article

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**Description of three new species of frog-biting midges (Diptera: Corethrellidae) from the Central Brazilian Amazon**

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**Abstract.** Three species of *Corethrella* Coquillett, 1902 from the state of Amazonas, Brazil are described as new to science based on female adult specimens. *Corethrella cabocla* Feijó, Belchior, Marialva & Pessoa sp. nov. possesses four large setae on the frons between the ventromedial area of ommatidia, a

wide clypeus with 1–4 setae, a wing with the apex of  $R_2$  basal to the apex of  $M_2$  and with a midlength band, and with the abdomen entirely dark brown. *Corethrella ielemdei* Feijó, Ramires, Lima & Pessoa sp. nov. possesses an elongated coronal suture, four large setae on the frons between the ventromedial area of ommatidia, a clypeus squarish with 42–43 setae, a wing with the apex of  $R_2$  basal to the apex of  $M_1$  and with a midlength band and dark scales on the basal and subbasal areas of the anterior margin, legs with dark scales, and with the abdomen entirely dark brown. *Corethrella menini* Feijó, Picelli, Ríos-Velásquez & Pessoa sp. nov. possesses wings with the apex of  $R_2$  basal to the apex of  $M_2$  and a midlength band, with darker basal scales along all veins, basal band dark scales on C, Sc, R, M, and Cu and the abdomen entirely dark brown. With the addition of the new species, the numbers of frog-biting midges described in the Amazon basin, Brazil and in Neotropical region are now 31, 49 and 80 species, respectively.

**Keywords.** Culicomorpha, taxonomy, hematophagous dipterans, Neotropical region.

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## Introduction

The monogeneric family Corethrellidae Edwards, 1932 is a widely distributed family in the tropical and subtropical areas (Borkent 2008) where species of the genus *Corethrella* Coquillett, 1902 locate their anuran hosts by sound instead of chemical cues (McKeever 1977; McKeever & Hartberg 1980; Toma *et al.* 2005; Bartlett-Healy *et al.* 2008; Borkent 2008; Bernal & de Silva 2015; Camp & Irby 2017). Female adults are ectoparasitic, feed on host blood and are documented vectors of trypanosomatids (Johnson *et al.* 1993; Bernal & Pinto 2016; Meuche *et al.* 2016). Recently, it was shown that members of this genus are involved in the transport of chytrid fungus, which is one of the main causes of the recent decline and extinction of amphibians (Toledo *et al.* 2021).

To date, the genus *Corethrella* includes 115 extant species and 10 extinct species. These insects have a semi cosmopolitan distribution with vast majority of extant species restricted to the tropics and subtropics (Kvifte & Bernal 2018; Amaral *et al.* 2019; Baranov *et al.* 2019). The Neotropical region possesses the highest richness of species of *Corethrella* with 77 registered species (Amaral 2018; Amaral *et al.* 2019), of which 28 species are from the Amazon basin (Borkent 2014; Amaral *et al.* 2019).

There are few studies about *Corethrella* from the Brazilian Amazon, with 20 recorded species (Borkent 2014; Amaral *et al.* 2019), out of which only five have been registered for the state of Amazonas.

In an entomological survey undertaken in a degraded forest area near Manaus, the administrative capital of the state of Amazonas, several hundred frog-biting midges were collected and some new species were found. This study expands our knowledge of species of the genus *Corethrella* in the state of Amazonas, Brazil, by describing three new species based on female specimens.

## Material and methods

Adult females were collected in a degraded forest from the experimental farm of the Universidade Federal do Amazonas (FAEXP-UFAM), located at km 38 of BR-174 highway (02°37'17.1" – 02°39'41.4" S, and 60°03'29.1" – 60°07'57.5" W), with an area of 3000 ha. This area is designated as secondary Amazonian upland 'terra-firme' forest (Fig. 1) and is characterized by floristic heterogeneity that includes palm trees, bromeliads, lianas, and other medium and large trees of heights between 35 to 40 m (Ribeiro

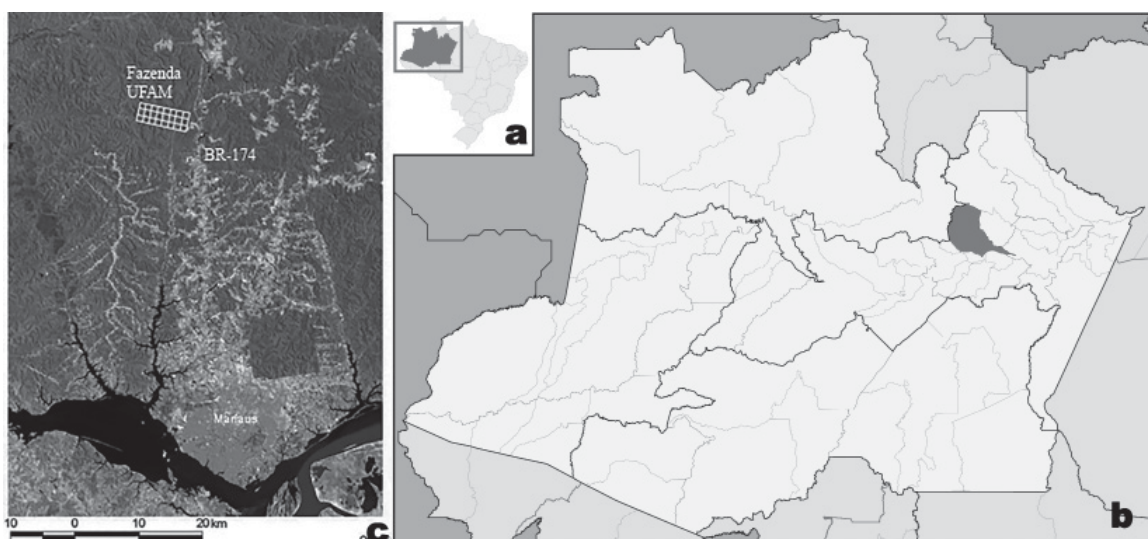
1998). The forest has a fairly dense canopy and an understory with low light, characterized by the abundance of palm trees (Guillaumet & Kahn 1982). Periodically, the extensive area of FAEXP-UFAM is flooded, due to the presence of large streams and an unregulated topography, with an altitude ranging from 42 to 130 m (Rojas-Ahumada *et al.* 2012).

Monthly air temperature variation is very low, with average temperatures between 24.6°C and 26.9°C. Daily relative air humidity ranges from 75% during relatively dry days up to 92% in the rainy season, and average annual rainfall is 2,362 mm (Araújo *et al.* 2002).

The collections were carried out on two trails in the farm over two days, in the months of April, May, and July 2019. Frog-biting midges were collected using four HP model light traps, two with and two without light bulbs (Pugedo *et al.* 2005); with overnight ‘frog call traps’ speakers attached, as described by McKeever & Hartberg (1980) (Fig. 2). The recorded frog sounds were from the ‘sapoteca’ frog library (<https://ppbio.inpa.gov.br/sapoteca/paginainicial>) of the Instituto Nacional de Pesquisas da Amazônia (Lima *et al.* 2012) (Supp. file 1).

The samples were stored in 70% alcohol. The frog-biting midges were separated from the other families of dipterans, and morphologically identified using the identification key by Borkent (2008). Then, female samples were dissected and mounted on slides in phenol-balsam (Wirth & Marston 1968). The diagnostic characteristics were photomicrographed using a digital system (Synoptics™, Cambridge, UK) coupled to an optical microscope (Leica DMTM1000, Frankfurt, Germany). The Helicon Soft High Assembly program was used to obtain the final images. For species descriptions, the morphological terms proposed by Borkent (2008) were used.

The holotypes were deposited in the entomological collection of the Instituto Leônidas e Maria Deane - Fiocruz Amazônia (ILMD) (Manaus, Brazil). The paratypes were housed in the entomological collections of the ILMD, Zoological collection of Instituto Nacional de Pesquisas da Amazonia (INPA) (Manaus, Brazil), Entomological collections of Instituto Oswaldo Cruz (CEIOC) (Rio de Janeiro, Brazil), and Zoological collection of Prof. Paulo Bührnheim (ZCPB) (Manaus, Brazil).



**Fig. 1.** Type locality of *C. cabocla* Feijó, Belchior, Marialva & Pessoa sp. nov., *C. ielemdei* Feijó, Ramires, Lima & Pessoa sp. nov., and *C. menini* Feijó, Picelli, Ríos-Velásquez & Pessoa sp. nov. **a.** Map of Brazil highlighting Amazonas state. **b.** Municipality of Manaus. **c.** Experimental farm of the Universidade Federal do Amazonas with access to BR-174.

We proposed an identification key for the new species, based on the species diagnoses in Borkent (2008), Amaral & Pinho (2015) and Amaral *et al.* (2019).

## Results

A total of 1270 female *Corethrella* were collected. In the modified light trap, 1106 individuals were captured: 395 *C. cabocla* sp. nov., 31 *C. ielemdei* sp. nov., 497 *C. menini* sp. nov. and 183 *C. manaosensis* (Lane & Cerqueira, 1958). In the light trap, 164 frog-biting midges were collected: 25 *C. cabocla* sp. nov., 60 *C. ielemdei* sp. nov. and 79 *C. manaosensis*.

## Taxonomic descriptions

Class Insecta Linnaeus, 1758  
Order Diptera Linnaeus, 1758  
Family Corethrellidae Edwards, 1932  
Genus *Corethrella* Coquillett, 1902

*Corethrella cabocla* Feijó, Belchior, Marialva & Pessoa sp. nov.  
urn:lsid:zoobank.org:act:A37CF95C-DF0F-452C-A9DC-D71F1DA6CE9D

Figs 3–4

## Diagnosis

The species can be distinguished from other species of the genus by the following characters the female adult. Four large setae on frons between ventromedial area of ommatidia; clypeus wide, with one or four setae; antenna with flagellomere one and two darker brown while the others are light brown, flagellomere I with four sensilla coeloconica, flagellomere II with one sensillum coeloconicum, flagellomeres VIII–XIII each with a single sensillum coeloconicum; posterior portion of the dorsocentral row with a group of about six elongate setae; wing with apex of  $R_2$  basal to apex of  $M_2$  and with midlength band; abdomen entirely dark brown.



**Fig. 2.** Method of *Corethrella* collection. **a.** CDC light trap with a speaker attached playing looping of frog species songs from the ‘sapoteca’ frog library (<https://ppbio.inpa.gov.br/sapoteca/paginainicial>). **b.** Cage with dozens of *Corethrella* collected.

**Table 1.** Distribution of sensilla coeloconica along the flagellar segments of the new species of *Corethrella* Coquillet, 1902. \*Arabic numerals = number of sensilla; \*\*Roman numerals = flagellomere number.

Species	Distribution of sensilla coeloconica in flagellomeres		
<i>Corethrella cabocla</i> sp. nov.	4* (I)**	1 (II)	1 (VIII–XIII)
<i>C. ielemdei</i> sp. nov.	2 (I)	1 (II)	2 (IX–XIII)
<i>C. menini</i> sp. nov.	4 (I)	1 (II)	1 (IX–XIII)

**Table 2.** Wing length measurements of the new female species of *Corethrella* Coquillet, 1902 in mm.

Species	Number	Wing length	Mean	1.5SD	R <sub>1</sub> / Wing length	Mean	1.5SD
<i>Corethrella cabocla</i> sp. nov.	10	1.1–1.25	1.17	0.050	0.587–0.70	0.663	0.038
<i>C. ielemdei</i> sp. nov.	10	1.45–1.62	1.5	0.062	0.583–0.92	0.648	0.130
<i>C. menini</i> sp. nov.	10	1.05–1.15	1.125	0.038	0.556–0.70	0.667	0.046

### Etymology

The name ‘*cabocla*’ refers to the local name for native Amazonians, feminine gender, a mixture of Amerindian, European and African origins, resulting in a rich heritage and culture.

### Material examined

#### Holotype

BRAZIL • ♀ adult; Amazonas State, Manaus, experimental farm of the Universidade Federal do Amazonas FAEXP-UFAM, BR-174, km 38; 02°38'44" S, 60°03'13" W; 4 Apr. 2019; H. Lima and A. Ramires leg.; from tropical forest and collected with light trap; ILMD 0001-45.

#### Paratypes

BRAZIL • 7 ♀♀; same collection data as holotype; ILMD 0001-46-50 • 2 ♀♀; same collection data as holotype; INPA - DIP - 004619 • 2 ♀♀; same collection data as holotype; CEIOC 81296 • 2 ♀♀; same collection data as holotype; CZPB - DP - 008266, CZPB - DP - 008267.

### Description

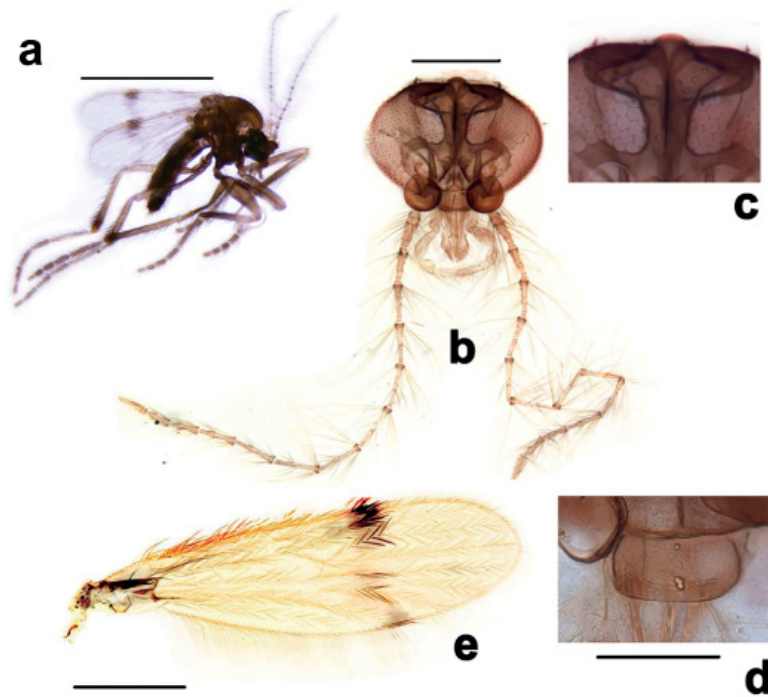
#### Adult female (n = 10)

HABITUS. As shown in Fig. 3a.

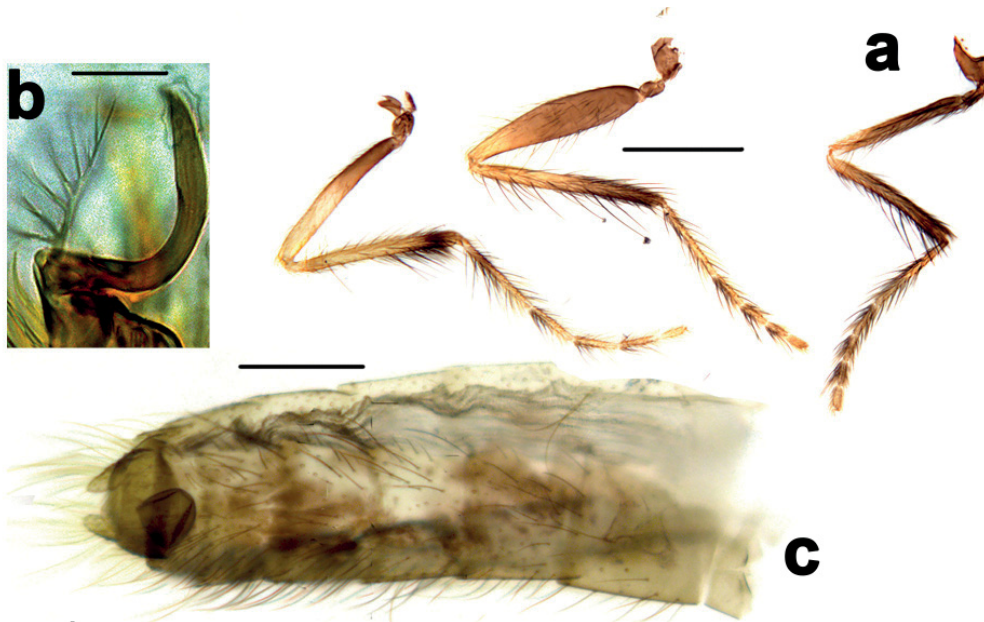
HEAD (Fig. 3b). Laterally expanded in dorsal view. Coronal suture elongate, reaching the area between ventral margin of eye bridge (Fig. 3c). Four large setae on frons between ventromedial area of the ommatidia.

ANTENNA (Fig. 3b). Darker to light brown, with flagellomeres I and II darker brown, while others are light brown; flagellomere XIII apically bifurcated; sensilla coeloconica distributed according to Table 1.

CLYPEUS (Fig. 3d). Wide, with one to four setae. Mandible with small pointed teeth. Palpus light brown; segment lengths: range; mean; 1.5 SD (in  $\mu\text{m}$ ): 1–(9.6–16.5; 11.5; 2.12), 2–(12.4–26.3; 18.4; 3.82), 3–(28.2–37.4; 36.1; 3.5), 4–(16.2–26.7; 21.7; 3.9), 5–(38.5–48.6; 43.2; 3.61); average width: range; mean; 1.5 SD (in  $\mu\text{m}$ ): 3–(10.5–17.4; 15.7; 2.74), 4–(8.8–15.7; 12.9; 2.45), 5–(7.1–15.1; 11.5; 2.74), third palpal segment slightly expanded at midlength.



**Fig. 3.** Microphotography of *Corethrella cabocla* Feijó, Belchior, Marialva & Pessoa sp. nov. ♀, holotype (ILMD 0001-45). **a.** Habitus. **b.** Head. **c.** Head with coronal suture elongate. **d.** Clypeus. **e.** Wing. Scale bars: a = 1 mm; b = 100 µm; c–d = 50 µm; e = 200 µm.



**Fig. 4.** Microphotography of *Corethrella cabocla*. Feijó, Belchior, Marialva & Pessoa sp. nov. ♀, holotype (ILMD 0001-45). **a.** Legs. **b.** Tarsal claw with empodium. **c.** Abdomen. Scale bars: a = 100 µm; b = 10 µm; c = 100 µm.

**THORAX.** Dark brown with scutum, scutellum medium brown, pale sclerites around base of wing. Posterior portion of dorsocentral row with group of approximately six elongate setae. Prescutal suture short, not extending to dorsocentral row of setae. Anterior anepisternum medium brown, divided diagonally suture, dorsal portion about equal to ventral portion. Posterior anepisternum dark brown, divided by transversal suture, with ventral portion triangular.

**WING** (Fig. 3e; Table 2). Apex of  $R_2$  basal to apex of  $M_2$ . Anterior margin with differently and discretely pigmented scales, with midlength band, some with darker more basal scales restricted to veins C, R (except  $R_4$ ), and M; veins with well-developed scales. Halter light brown, lighter than scutellum.

**LEGS** (Fig. 4a). Dark to light brown with slender setae dark brown. Foreleg with dark brown femur to about  $\frac{3}{10}$  of total light brown structure in apex region, tibia dark brown, approximately  $\frac{1}{10}$  of the total light brown structure in the base region, tarsomeres 1 to 5 light brown. Midleg with dark brown femur, approximately  $\frac{3}{10}$  of total light brown apically, tibia light brown from base to about  $\frac{5}{10}$  of total length, other half dark brown, extending to apex, tarsomeres one to five light brown. Hindleg with dark brown femur from base to about  $\frac{4}{10}$  of length, apical  $\frac{1}{10}$  light brown, tibia mostly light brown, about apical  $\frac{3}{10}$  dark brown, tarsomeres 1 to 5 light brown. Tarsal claws equal, inserted apically; each claw without an inner tooth. Empodia (Fig. 4b) with similar thickness to apical setae of last tarsomere, longer than average diameter of last tarsomere, with five or six bifid branches.

**ABDOMEN.** Uniformly dark brown. One ovoid spermatheca. Cercus, equally dark brown (Fig. 4c).

#### **Male**

Unknown

#### **Distribution**

This species is known only from its type locality in the experimental farm of the Universidade Federal do Amazonas FAEXP-UFAM, Manaus, Amazonas State, Brazil.

#### **Taxonomic comments**

In the Neotropical key by Borkent (2008) *C. cabocla* sp. nov. keys to *C. lepida* Borkent, 2008. However, it is distinguished from this species by having the antennal flagellomeres dark brown, whereas in *C. lepida* the antennal flagellomeres I–IV are light brown. Furthermore, *C. cabocla* sp. nov. has the first flagellomere with four sensilla coeloconica, whereas in *C. lepida*, the first flagellomere has been described with two sensilla, and the scutum of *C. cabocla* sp. nov. is dark brown and the scutellum light brown, distinct from *C. lepida*, in which the color of the scutum and scutellum ranges from medium to light brown. The posterior portion of the dorsocentral thorax in *C. cabocla* sp. nov. has a group of six elongate setae, whereas in *C. lepida* five elongate setae are present. The wing of *C. cabocla* sp. nov. has the apices of  $R_2$  and  $M_2$  at approximately the same level, whereas in *C. lepida*  $R_2$  and  $M_1$  are nearly at the same level. In *C. lepida*, a pale brown band has been described on each of tarsomeres 2–4 of the mid and hind leg; tarsomeres are uniformly pigmented in *C. cabocla* sp. nov. The abdomen of *C. cabocla* sp. nov. is entirely dark brown, whereas in *C. lepida* only segments VIII and IX are dark brown. *Corethrella caribbeana* Borkent, 2008 shares some characteristics with *C. cabocla* sp. nov. and *C. lepida*, but *C. caribbeana* is distinguished from *C. cabocla* sp. nov. based on several features. In *C. caribbeana* the flagellomeres I–III are short, whereas in *C. cabocla* sp. nov. they are elongate. In addition, *Corethrella caribbeana* has two sensilla distributed on each of flagellomeres IX–XII, whereas in *C. cabocla* sp. nov. there is only one on each of flagellomeres VIII–XIII. The clypeus of *C. caribbeana* is squarish, different from that of *C. cabocla* sp. nov., which is wide. The abdomen of *C. caribbeana* is light to dark brown, with sternites I–II slightly darker and segment IX dark brown, whereas in *C. cabocla* sp. nov. the abdomen is entirely dark brown.

The holotype has a set of characteristics that do not fit within the existing species groups of *Corethrella* (Borkent 2008). The only apparent synapomorphy that *C. cabocla* sp. nov. shares with the groups *fulva*, *appendiculata*, *brakeleyi* and *wirthi* is found on the posterior dorsocentral region of the thorax, where a group of more than three elongated setae is present. The lack of knowledge of the immature stages and adult males of *C. cabocla* sp. nov., makes it difficult to associate the new species with a specific species group as defined by Borkent (2008).

**We suggest to incorporate the following emendation to the *Corethrella*. Key by Borkent (2008) to identify adult females of *C. cabocla* sp. nov.**

46. Abdomen uniformly dark brown; cercus dark brown; flagellomere I with four sensilla coeloconica.  
 ..... *C. cabocla* Feijó, Belchior, Marialva & Pessoa sp. nov.  
 – Abdomen medium to dark brown, with segments VIII and IX dark brown; cercus medium brown; flagellomere I with one to two sensilla coeloconica ..... 47
47. Flagellomere I–III elongate; flagellomeres IX–XIII each with only a single sensillum coeloconicum ..... *C. lepida* Borkent, 2008  
 – Flagellomere I–III short; female flagellomeres IX–XII each with two sensilla coeloconica .....  
 ..... *C. caribbeana* Borkent, 2008

*Corethrella ielemdei* Feijó, Ramires, Lima & Pessoa sp. nov.  
 urn:lsid:zoobank.org:act:B2522305-03E9-4BA7-9792-696AC4A34802  
 Figs 5–6

### Diagnosis

The species can be distinguished from other species of the genus by the following characteristics of the female adult. Coronal suture elongate, four large setae on frons between ventromedial area of ommatidia, clypeus squarish, with 42–43 setae; antenna uniformly medium brown, flagellomere I with two sensilla coeloconica, flagellomere II with one sensillum coeloconicum, flagellomeres IX–XIII with two sensilla coeloconica; posterior portion of dorsocentral row with a group of about eight elongate setae; wing with apex of  $R_2$  basal to apex of  $M_1$  and with midlength band and dark scales on basal and subbasal areas of anterior margin; foreleg, midleg, and hindleg with femora and foretibia with dark scales.

### Etymology

The name ‘*ielemdei*’ is based on the acronym of the Instituto Leônidas e Maria Diane - Fiocruz Amazônia, an important research facility for training and providing professional experiences for biologists, including taxonomists for the Amazon region. To be treated as a noun in apposition.

### Material examined

#### Holotype

BRAZIL • ♀ adult; Amazonas State, Manaus, experimental farm of the Universidade Federal do Amazonas FAEXP-UFAM, BR-174 km 38; 02°38'44" S, 60°03'13" W; 7 Jul. 2019; H. Lima and A. Ramires leg.; from tropical forest and collected with modified light trap (‘frog call traps’); ILMD 0001-51.



### Paratypes

BRAZIL • 7 ♀♀; same collection data as holotype; ILMD 0001-52-56 • 2 ♀♀; same collection data as holotype; INPA - DIP - 004618 • 2 ♀♀; same collection data as holotype; CEIOC 81297 • 2 ♀♀; same collection data as holotype; CZPB - DP - 008268, CZPB - DP - 008269.

### Description

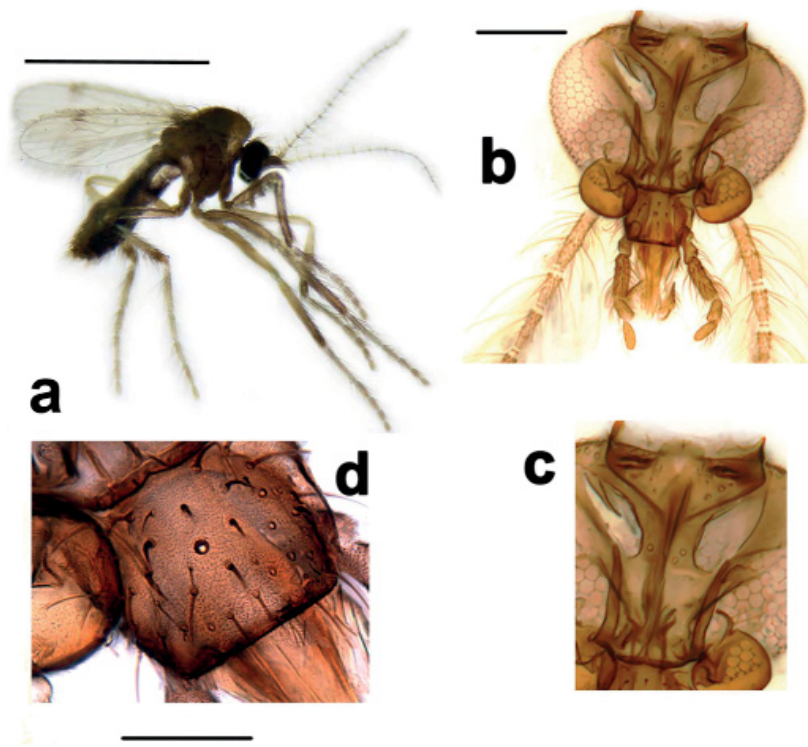
#### Adult female (n = 10)

HABITUS. As shown in Fig. 5a.

HEAD (Fig. 5b). Laterally expanded in dorsal view. Coronal suture elongate, reaching the area between ventral margin of eye bridge (Fig. 5c). Four large setae on the frons between the ventromedial area of ommatidia.

ANTENNA (Fig. 5b). Uniformly medium brown; flagellomere XIII apically bifurcated; sensilla coeloconica distributed according to Table 1.

CLYPEUS (Fig. 5d). Squarish, with 42–43 setae. Mandible with small pointed teeth. Palpus dark brown; segment lengths: range; mean; 1.5 SD (in  $\mu\text{m}$ ): 1–(7.6–13.8; 10.8; 2.2), 2–(15.4–22.1; 17.8; 2.6), 3–(31.8–38.2; 35.4; 2.4), 4–(11.4–22; 16.1; 3.1), 5–(21.8–28.7; 23.7; 2.7); average width: range; mean; 1.5 SD (in  $\mu\text{m}$ ): 3–(11.1–17; 15.5; 2.3), 4–(8.1–14.5; 11.2; 2.3), 5–(6.5–11.3; 9.4; 1.6), third palpal segment of constant width.



**Fig. 5.** Microphotography of *Corethrella ielemdei*. Feijó, Ramires, Lima & Pessoa sp. nov., ♀, (ILMD 0001–51). **a.** Habitus. **b.** Head. **c.** Head with coronal suture elongate. **d.** Clypeus. Scale bars: a = 1 mm; b = 100  $\mu\text{m}$ ; c–d = 50  $\mu\text{m}$ .

**THORAX.** Dark brown, scutum, scutellum, pale sclerites around the base of the wing. Posterior portion of dorsocentral row with a group of approximately eight elongate setae. Prescutal suture short, not extending to dorsocentral row of setae. Anterior anepisternum dark brown, divided by diagonal suture, dorsal portion about equal to ventral portion. Posterior anepisternum dark brown, divided by transversal suture, with ventral portion triangular.

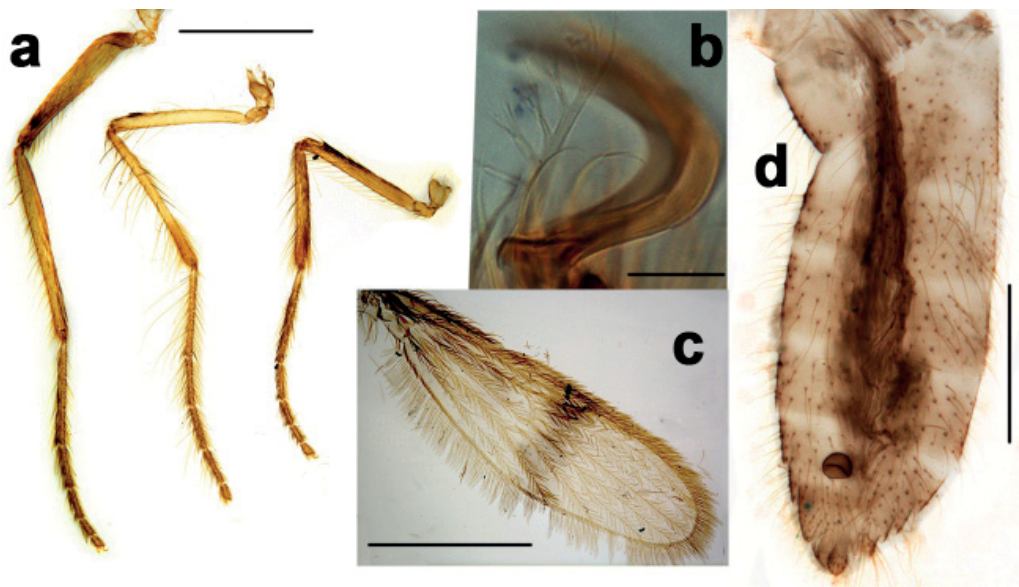
**WING** (Fig. 6c; Table 2). Apex of  $R_2$  basal to apex of  $M_1$ . Anterior margin with differently and discretely pigmented scales, with midlength band, with darker more basal scales along all veins (except  $A_1$ ); some scattered dark scales on basal and subbasal areas of anterior margin; veins with well-developed scales. Halter dark brown, darker than scutellum.

**LEGS** (Fig. 6a). Dark to light brown and slender setae light brown. Foreleg with femur entirely dark brown and thin dark scales on upper margin of medial portion extending near apex of femur; tibia dark brown, with thin dark scales on lower margin extending from the base to middle of the structure, first tarsomere dark brown and about  $\frac{2}{10}$  of the total light brown structure in base region, and other tarsomeres dark brown. Midleg with dark brown femur with a tuft of thin dark scales near apex region of the structure, tibia, and tarsomeres entirely dark brown. Hindleg with entirely light brown femur with a tuft of dark scales on posterior margin near apex of the structure, tibia predominantly light brown but about  $\frac{3}{10}$  dark brown in the apex region, tarsomeres 1 to 5 dark brown; tarsal claws equal, inserted apically; each claw without inner tooth. Empodia (Fig. 6b). With thickness larger than apical bristles of last tarsomere, longer than average diameter of last tarsomere, with seven bifid branches.

**ABDOMEN** (Fig. 6d). Uniformly dark brown. One ovoid spermatheca. Cercus dark brown.

#### Male

Unknown



**Fig. 6.** Microphotography of *Corethrella ielemdei* Feijó, Ramires, Lima & Pessoa sp. nov. ♀, (ILMD 000-51). **a.** Legs. **b.** Tarsal claw with empodium. **c.** Wings. **d.** Abdomen. Scale bars: a = 100 µm; b = 10 µm; c–d = 500 µm.

### Distribution

The species is known only from its type locality in the experimental farm of the Universidade Federal do Amazonas FAEXP-UFAM, Manaus, Amazonas State, Brazil.

### Taxonomic comments

The characters present in *C. ielemdei* sp. nov., as distribution of sensilla coeloconica, eight setae in the dorsocentral region of the thorax, and dark brown cercus and halter, are shared with *C. squamifemora* Borkent, 2008 and *C. hirta* Borkent, 2008. However, they can be distinguished from these species by the presence of an elongated coronal suture (in *C. squamifemora* and in *C. hirta*, they are shorter), four long setae between the ommatidia (in *C. squamifemora* and in *C. hirta*, there are just two), and squarish clypeus (in *C. squamifemora* and in *C. hirta*, they are broadly roundish). The presence of an elongate coronal suture, a dark antenna, and the wing with the apex of  $R_2$  basal to apex of  $M_1$  in *C. ielemdei* sp. nov. are also shared with *C. albicoxa* Borkent, 2008, *C. appendiculata* Grabham, 1906, *C. calathicola* Edwards, 1930, and *C. melanica* Lane & Aitken, 1956. However, the new species can be distinguished from these by the dark brown palpus in comparison to the pale palpus in *C. albicoxa*, *C. appendiculata*, *C. calathicola*, and *C. melanica*. The third palpal segment is evenly broad throughout its length in *C. ielemdei* sp. nov., whereas in *C. albicoxa*, *C. appendiculata*, *C. calathicola*, and *C. melanica* it is somewhat swollen in the middle of this segment. In addition, the pattern and distribution of dark bands and scales along the wing are different in *C. ielemdei* sp. nov., *C. albicoxa*, *C. appendiculata*, *C. calathicola*, and *C. melanica*.

*Corethrella ielemdei* sp. nov. shares characteristics with species in the *appendiculata* group (Borkent 2008), including scales on the femora and tibiae, and having a group of more than three elongated setae in posterior dorsocentral region of the thorax. The color of the medial and posterior femur distinguishes *C. ielemdei* sp. nov. from the other species of this group. The description of males and immature stages will probably confirm the inclusion of this species in the *appendiculata* group.

**We suggest to incorporate the following couplets in the *Corethrella*. Key by (Borkent 2008) to identify adult females of *C. ielemdei* sp. nov.**

47. Midfemur dark brown, contrasting with hindfemur which is entirely light brown .....  
..... *C. ielemdei* sp. nov.  
– Base of the midfemur with distinct color; base of the hindfemur ranging from light brown to dark brown ..... 48

*Corethrella menini* Feijó, Picelli, Ríos-Velásquez & Pessoa sp. nov.  
urn:lsid:zoobank.org:act:D2973CA5-CC66-4EFA-A223-8697D048D1AE  
Figs 7–8

### Diagnosis

The species can be distinguished from other species of the genus by the following characteristics of the female adult. Coronal suture elongate, two large setae on the frons between ventromedial area of ommatidia, clypeus squarish, with three setae; antenna dark brown, flagellomere I with four sensilla coeloconica, flagellomere II with one sensillum coeloconicum, flagellomeres IX–XIII with one sensillum coeloconicum; posterior portion of dorsocentral row with a group of about six elongate setae; wing with apex of  $R_2$  basal to apex of  $M_2$  and midlength band, with darker more basal scales along all on veins, basal band dark scales on C, Sc, R, M, and Cu; abdomen, completely dark brown.

### Etymology

*Corethrella menini* sp. nov. is named in honor of Dr. Marcelo Menin as a tribute to his important work as a herpetologist, focused on ecology and biology of anurans. Marcelo was a professor of zoology who inspired dozens of zoology students, a beloved friend. He passed away too young during the COVID pandemic.

### Material examined

#### Holotype

BRAZIL • ♀ adult; Amazonas State, Manaus, experimental farm of the Universidade Federal do Amazonas FAEXP-UFAM, BR-174, km 38; 02°38'38' S, 60°03'13' W; 14 Jul. 2019; H. Lima and A. Ramires leg.; from tropical forest and collected with modified light trap ('frog call traps') ILMD 0001-57.

#### Paratypes

BRAZIL • 7 ♀♀; same collection data as holotype; ILMD 0001-58-62 • 2 ♀♀; same collection data as holotype; INPA - DIP - 004617 • 2 ♀♀; same collection data as holotype; CEIOC 81298 • 2 ♀♀; same collection data as holotype; CZPB - DP - 008264, CZPB - DP - 008265.

### Description

#### Adult female (n = 10)

HABITUS. As shown in Fig. 7a.

HEAD (Fig. 7b). Laterally expanded in dorsal view. Coronal suture elongate, reaching the area between ventral margin of eye bridge (Fig. 7c). Two large setae on frons between ventromedial area of ommatidia.

ANTENNAE. Dark brown, with flagellomere XIII without apical bifurcation; sensilla coeloconica distributed according to Table 1.

CLYPEUS (Fig. 7d). Squarish with three setae. Mandible with small pointed teeth. Palpus dark brown; segment lengths: range; mean; 1.5 SD (in  $\mu\text{m}$ ): 1–(5.3–10.9; 8.7; 1.9), 2–(9.9–16.8; 13.2; 2.7), 3–(29.7–43.3; 34.5; 4.7), 4–(11.7–23.7; 19.5; 3.8), 5–(27.3–44.7; 41; 5.9); average width: range; mean; 1.5 SD (in  $\mu\text{m}$ ): 3–(10.6–15.9; 12.11; 1.9), 4–(9.4–15.6; 10.65; 2.4), 5–(6.8–15.8; 11.3; 3.1), and the third palpal segment was nearly constant and swollen at or near midlength.

THORAX (Fig. 8a). Dark brown, scutum, scutellum, pale sclerites around base of wing. Posterior portion of dorsocentral row with a group of six elongate setae. Prescutal suture short, not extending to dorsocentral row of setae. Anterior anepisternum dark brown, divided by diagonal suture, dorsal portion about equal to ventral portion. Posterior anepisternum dark brown, divided by transversal suture, with ventral portion triangular.

WING (Fig. 7e) (Table 2). Apex of  $R_2$  basal to apex of  $M_2$ . Anterior margin with differently and discretely pigmented scales, midlength band, with darker more basal scales along all veins, basal dark scales on C, Sc, R, M, and Cu; veins with well-developed scales. Halter dark brown, equal to the scutellum.

LEGS (Fig. 8a). Dark brown and slender setae dark brown. Foreleg with femur, tibia, and tarsomeres entirely dark brown. Midleg with femur, tibia, and tarsomeres entirely dark brown. Hindleg with femurs dark brown to about  $\frac{3}{10}$  of the total light brown structure in apex region, tibia dark brown but apical and basal extremities light brown, tarsomeres 1 to 5 light brown. Tarsal claws equal, inserted apically; each claw without an inner tooth. Empodia (Fig. 8b) with a thickness equal to apical bristles of last tarsomere, smaller than average diameter of last tarsomere, with one bifid branch.

ABDOMEN (Fig. 8c). Uniformly dark brown. One ovoid spermatheca. Cercus equally dark brown.

#### Male

Unknown

#### Distribution

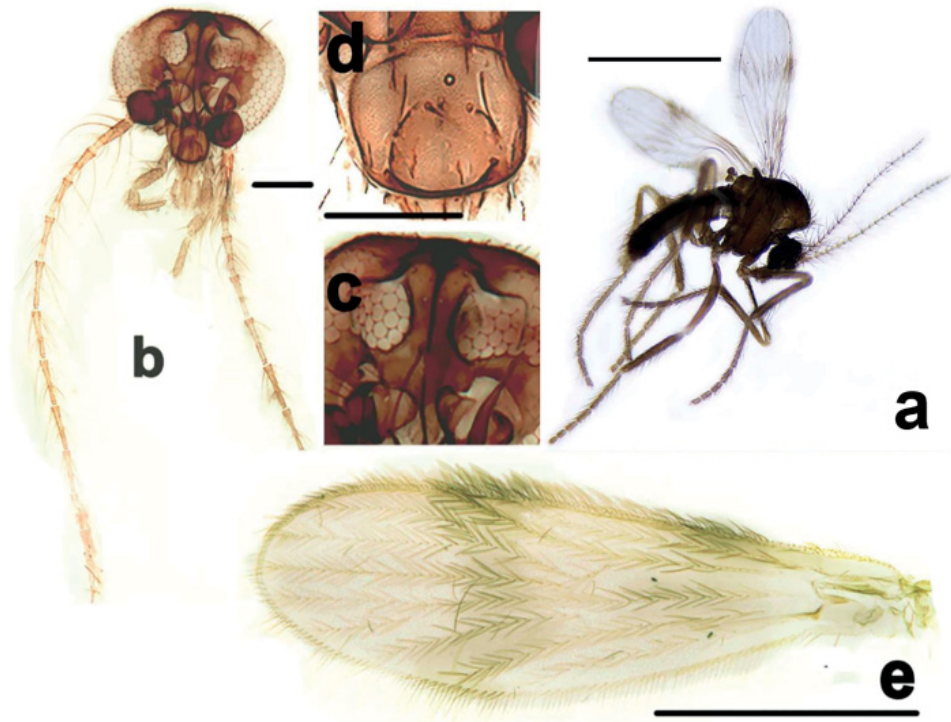
This species is known only from its type locality in the experimental farm of the Universidade Federal do Amazonas FAEXP-UFAM, Manaus, Amazonas State, Brazil.

#### Taxonomic comments

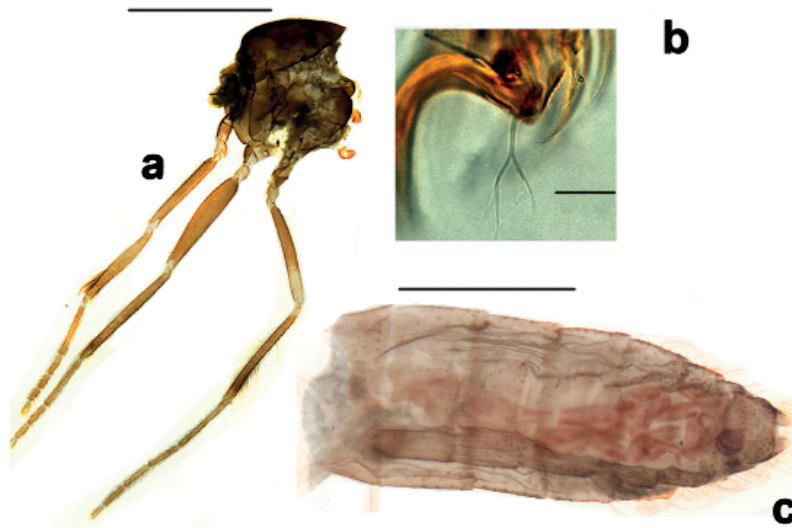
The structures present in *C. menini* sp. nov., such as the elongated coronal suture, the presence of two long frontal bristles between ommatidia, the third palpal segment being almost constant in width, and the group of six bristles in the thoracic dorsocentral region are characteristic for species in the *brakeleyi* group: *C. hispaniolensis* Borkent, 2008, *C. puella* Shannon & Del Ponte, 1928, *C. condita* Borkent, 2008, *C. brakeleyi* (Coquillett, 1902), and *C. longituba* Belkin, Heinemann & Page, 1970. However, *C. menini* sp. nov. can be distinguished from these based on the color of the palpus (dark brown in *C. menini* sp. nov., pale in *C. hispaniolensis* and *C. longituba*, and medium brown in *C. puella*, *C. condita*, and *C. brakeleyi*), the number of sensilla coeloconica on the flagellomeres, the band pattern and scales along the wing, and the color of the halter (dark brown in *C. menini* sp. nov. and pale in *C. hispaniolensis*, *C. puella*, *C. condita*, *C. brakeleyi*, and *C. longituba*). The squarish clypeus and the dark brown color of the antenna, halter, and cercus are characteristics shared by *C. menini* sp. nov., *C. curta* Borkent, 2008, and *C. ranapungens* Borkent, 2008. However, the thoracic dorsocentral region of *C. curta* and *C. ranapungens* have a group of three and four elongated bristles, respectively as opposed to six setae in *C. menini* sp. nov.; in addition, *C. menini* sp. nov. has the apex of  $R_2$  basal to the apex of  $M_2$ , whereas in *C. curta* and *C. ranapungens*, the apex of  $R_2$  is basal to the apex of  $M_1$ . Other characters, such as the color pattern of the thorax and palpus, can also be used to distinguish the species *C. menini* sp. nov. from *C. curta* and *C. ranapungens*. The new species likely belongs to the *brakeleyi* group, which includes eight species. However, male characters are still necessary to support this hypothesis because the occurrence of wings with apical bands, a characteristic that defines the group, is more commonly found in adult males.

#### We suggest to add the following couplets to the *Corethrella* key by Borkent (2008) to identify adult females of *C. menini* sp. nov.

90. Flagellomeres IX–XII each with two sensilla coeloconica.....*C. incompta* Borkent, 2008  
– Flagellomeres IX–XII each with a single sensillum coeloconicum ..... 91
91. Flagellomere I with one or two sensilla coeloconica; dorsocentral region of the thorax with a group of 4 elongate setae; wing with apex of  $R_2$  basal to apex of  $M_1$ ..... *C. ranapungens* Borkent, 2008  
– Flagellomere I with four sensilla coeloconica; dorsocentral region of the thorax with a group of 6 elongate setae; wing with apex of  $R_2$  basal to apex of  $M_2$ ..... *C. menini* sp. nov.



**Fig. 7.** Microphotography of *Corethrella menini* Feijó, Picelli, Ríos-Velásquez & Pessoa sp. nov. ♀, holotype (ILMD 0001-57). **a.** Habitus. **b.** Head. **c.** Head with coronal suture elongate. **d.** Clypeus. **e.** Wing. Scale bars: a = 1 mm; b = 100 µm; c–d = 50 µm; e = 500 µm.



**Fig. 8.** Microphotography of *Corethrella menini* Feijó, Picelli, Ríos-Velásquez & Pessoa sp. nov. ♀, holotype (ILMD 0001-57). **a.** Thorax and legs. **b.** Tarsal claw with empodia. **c.** Abdomen. Scale bars: a–c = 500 µm; b = 10 µm.

Identification key to female Amazon basin species of *Corethrella*.

The species *C. maculata* Lane, 1939, *C. shannoni* Lane, 1939 and *C. munteantaroku* Amaral, Mariano & Pinho, 2019 are not included as females of these species have not been described yet.

1. Wing plain or mostly plain ..... 2
  - Wing with distinct banding ..... 9
2. Wing completely plain ..... 3
  - Mostly plain, with very light poorly defined midlength band, anterior margin of wing uniformly pigmented ..... *C. blanda* Dyar, 1928
3. Tergites II–VIII with most of the posterior portion with dark band, tergites I, IX probably uniformly light brown ..... *C. alticola* Lane, 1989
  - Abdominal tergites uniformly pigmented ..... 4
4. Palpus pale; clypeus with 1 seta ..... *C. unisetosa* Borkent, 2008
  - Palpus darker, clypeus with one or more than one seta ..... 5
5. Thorax light brown. Scutum light brown with lateral vitta distinctly dark brown .....
  - Thorax medium to dark brown ..... *C. manaosensis* (Lane & Cerqueira, 1958) 6
6. Thorax almost uniformly medium to dark brown but with the scutellum lighter laterally and with a median, longitudinal, dark stripe ..... 7
  - Scutellum without a median, longitudinal, dark stripe ..... 8
7. Antennal flagellomere I–II pale, abdomen light brown, with sternite II darker .....
  - Antenna uniformly brown, abdomen with segments medium brown ..... *C. tarsata* Lane, 1942
  - Antenna uniformly brown, abdomen with segments medium brown ..... *C. redacta* Borkent, 2008
8. Thorax uniformly dark brown contrasting with a pale halter, the midfemur with darker pigmentation at its base, and hind leg femur and tibia uniformly pigmented ..... *C. peruviana* Lane, 1939
  - Thorax uniformly medium brown, the halter as dark as the scutellum, with flagellomeres I–III elongate, foretrochanter pale and contrasting with dark forefemur ..... *C. amazonica* Lane, 1939
9. Wing with four transverse bands ..... 10
  - Wing with three or at least one band ..... 12
10. Halter dark brown (similar to the scutellum) ..... *C. quadrivittata* Shannon & Del Ponte, 1928
  - Halter pale or light brown (lighter than the scutellum) ..... 11
11. Abdomen and cercus medium brown ..... *C. vittata* Lane, 1939
  - Abdomen dark brown with, in some, segments VIII, IX slightly darker, cercus dark brown ..... *C. edwardsi* Lane, 1942
12. Wing with basal and midlength band of dark scales ..... *C. menini* sp. nov.
  - Wing without basal band but with a discreet or well-developed midlength and dark scales ..... 13
13. Two large setae on frons between ventromedial area of ommatidia ..... 14
  - Up to four large setae on frons between ventromedial area of ommatidia ..... 24

14. Flagellomeres I and II short with flagellomere I nearly globular, flagellomere III elongate.....	<i>C. bicolor</i> Borkent, 2008
– Flagellomeres I–III otherwise.....	15
15. Thorax dark brown, halter and scutellum equally dark brown.....	16
– Thorax dark brown, halter pale, much lighter than scutellum.....	18
16. Midfemur and hind tibia uniformly dark brown.....	<i>C. travassosi</i> Lane, 1942
– Midfemur dark brown, base of hind tibia pale, similar to the apex of hind femur.....	17
17. Flagellomeres IX–XIII each with one sensillum coeloconicum.....	<i>C. ranapungens</i> Borkent, 2008
– Flagellomeres IX–XII each with two sensilla coeloconica.....	<i>C. incompta</i> Borkent, 2008
18. Primarily medium to dark brown palpus.....	19
– Primarily pale or light brown palpus.....	20
19. Clypeus wide, few setae (less than 5).....	<i>C. fuscipalpis</i> Borkent, 2008
– Clypeus elongate, more than 5 to 10 setae.....	<i>C. puella</i> Shannon & Del Ponte, 1928
20. Flagellomere III without a sensillum coeloconicum.....	21
– Flagellomere III with a sensillum coeloconicum.....	23
21. Presence of 2 sensilla coeloconica on flagellomeres X–XII.....	<i>C. ananacola</i> Dyar, 1926
– Absence of 2 sensilla coeloconica on flagellomeres X–XII.....	22
22. Coronal suture short.....	<i>C. dicosimoae</i> Borkent, 2008
– Coronal suture elongate.....	<i>C. melanica</i> Lane & Aitken, 1956
23. Coronal suture elongate, extending ventrally past ommatidia.....	<i>C. inca</i> Lane, 1939
– Coronal suture short, extending ventrally about midway along the area between ommatidia.....	<i>C. selvicola</i> Lane, 1939
24. Clypeus squarish, with more than 10 setae.....	<i>C. ielemdei</i> sp. nov.
– Clypeus wide, with up to four setae.....	25
25. Thorax with a group of eight setae.....	<i>C. appendiculata</i> Grabham, 1906
– Thorax with a group of five to six setae.....	26
26. Legs without any well-defined pigmentation pattern (hind tibia slightly darker apically and hind femur slightly less pigmented distally).....	<i>C. yanomami</i> Amaral, Mariano & Pinho, 2019
– Legs with any well-defined pigmentation pattern.....	27
27. Antenna with flagellomere I and II dark brown and the others light brown; flagellomere I with four sensilla; scutum and scutellum medium brown; abdomen entirely dark brown....	<i>C. cabocla</i> sp. nov.
– Antenna with flagellomere I–IV light brown and V–XIII progressively dark; flagellomere I with 2 sensilla; scutum and scutellum light to medium brown; abdomen light brown to medium brown with segments VIII and IX darker.....	<i>C. lepida</i> Borkent, 2008



## Discussion

The possible preference for the calls of certain frog species seems to be a relevant factor that influences the diversity of frog-biting midges captured (Ambrozio-Assis *et al.* 2019). In our study, it was not possible to evaluate the preference of species of *Corethrella* to a specific type of frog call, because we used a database with more than 80 frog sounds. Most of the individuals were collected in the modified trap, with speakers; similar results were also found in studies of Caldart *et al.* (2016). Some other studies carried out in Brazil (Amaral & Pinho 2015; Amaral *et al.* 2019; Caldart *et al.* 2016), using same collection methods, captured less samples. Partly, this can be explained by the differences in collection methods, frog and toad mosquito fauna, and biomes.

Of the four species of frog-biting midges collected at FAEXP-UFAM, three are new to science. Collections of *Corethrella* are being carried out in several regions of the Brazilian Amazon and soon we hope to obtain data in order to expand the distribution of frog-biting midges, especially in areas with no existing records, but with favorable conditions for their establishment. The uncertainty of associating the new species to the groups described by Borkent (2008), exemplified the need to collect immature stages and males to robustly evaluate the phylogenetic position of Corethrellidae species.

The Amazon Basin is a vast heterogeneous area that constitutes half of the Brazilian territory, but very few surveys have been conducted for *Corethrella* in this region. Extensive ecological and biodiversity studies, especially focussing on areas with geographical barriers, e.g., areas between large rivers, are necessary to understand the overall distribution and abundance of these species in Brazil.

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### **Supplementary file**

**Supp. file 1.** List of song of frogs, found in ‘sapoteca’ frog library, with (<https://ppbio.inpa.gov.br/sapoteca/paginainicial>). <https://doi.org/10.5852/ejt.2021.778.1577.5473>

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