



BIOLOGICAL SCIENCES

***Schizonyxhelea thomsenae* (Wirth), description of the pupa and first records from Argentina, Brazil and Peru (Diptera: Ceratopogonidae)**

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Abstract: The pupa of *Schizonyxhelea thomsenae* (Wirth) is described and illustrated from material collected in Misiones province, Argentina. A key for the four known species of pupae of *Schizonyxhelea* is given, a diagnosis and photographs of the adult are also provided, and the geographic distribution of the species in the Neotropical region is enlarged including first records from Argentina, Brazil and Peru based mainly on adult specimens.

Key words: Neotropical region, predaceous midges, pupa, new records, *Schizonyxhelea*.

INTRODUCTION

The small predaceous midges of the worldwide genus *Schizonyxhelea* Clastrier 1984 are currently represented by 15 species (Borkent 2016, Huerta & Grogan 2017). This genus was restricted to only two Neotropical species: *S. forattinii* Wirth and Grogan 1988 and *S. guyana* Clastrier 1984 until Borkent (2000, 2014), after a deep study of the male features and the striking similarities between the pupae of *Schizonyxhelea* and *Stilobezzia* Kieffer 1911 redefined the genus and transferred eight species previously placed in *Stilobezzia*: *Schizonyxhelea brevicostalis* (Das Gupta & Wirth 1968), *S. bulla* (Thomsen 1935), *S. caribe* (Lane & Forattini 1958), *S. diminuta* (Lane & Forattini 1958), *S. obscura* (Lane & Forattini 1958), *S. panamensis* (Lane & Forattini 1958), *S. thomsenae* (Wirth 1953) and *S. scutata* (Lane & Forattini 1961). Posteriorly, more species revised by Clastrier (1991) and considered to be related to *Stilobezzia insolita* Das Gupta and Wirth 1968 were also recognised as members of *Schizonyxhelea* by Borkent (2015), on the basis

of a transverse sclerite in their male genitalia and/or the single, distinctive, basally bent spermatheca of the female: *Schizonyxhelea afra* (Clastrier 1991), *S. afrotropica* (Clastrier 1991), *S. corneti* (Clastrier 1991), *S. amazonica* (Clastrier 1991), *S. gallica* (Clastrier 1991) and *S. insolita* (Das Gupta & Wirth 1968). Finally, Huerta & Grogan (2017) described a new species from Mexico, *Schizonyxhelea zoologica* and provided a key to identification for adult specimens, as well as new records for the genus from the Neotropical region. Regarding the immatures, currently the larva and pupae of only two species of *Schizonyxhelea* are known: *Schizonyxhelea forattinii* and *S. bulla* (Borkent 2014).

During a field sampling in Misiones province, Argentina, two pupae of *Schizonyxhelea* were collected. The adults posteriorly emerged were identified as *Schizonyxhelea thomsenae* (Wirth). Besides, the specimens of *Schizonyxhelea* deposited in the collection of the División Entomología, Museo de La Plata were studied and allowed us to identify several adult specimens

of this species from Neotropical localities. The purpose of this contribution is to describe and illustrate the pupa of *Schizonyxhelea thomsenae*, to provide a key of pupae of the four known species, as well as to give a diagnosis of the adult and illustrate it. We also enlarge the geographic distribution of this species providing the first records from Argentina, Brazil and Peru.

MATERIALS AND METHODS

The pupae were collected with kick net from a flooded, grassy pond next to the route with an average of water temperature of 29.8°C, pH of 9-10 and conductivity of 243.8 µS/cm. They were carried back to the laboratory and conditioned individually in vials with water. Observations were done daily until adult emergence three days later and when needed, a small amount of water was added to the vial to keep the sample from drying. Adults were allowed to harden for 24 hours before being preserved to ensure its pigmentation was complete.

For detailed examination with phase-contrast microscope pupal exuviae and adults were mounted in Canada balsam following the technique described by Borkent & Spinelli (2007). Photographs were taken with a digital camera Micrometrics SE Premium, through Nikon Eclipse E200 microscope and digital camera Leica EC3, through Leica DM 500. Ink illustrations were drawn with Genius Ultra-slim tablet from photographs. Measurements were taken using binocular microscope (BCM). For terms of pupae see Borkent (2014). For terms of adult see the Manual of Central America Diptera (Borkent et al. 2009). Studied specimens are deposited in the collection of the División Entomología, Museo de La Plata (MLPA), Argentina except as noted.

RESULTS

Schizonyxhelea thomsenae (WIRTH 1953)

Stilobezzia (Stilobezzia) thomsenae Wirth 1953: 83 (male, female; USA); Wirth 1965: 134 (in Nearctic catalog north of Mexico; distribution); Wilkening et al. 1985: 526 (Florida records); Borkent 1995: 98 (interpretation of aedeagal characters); Borkent & Wirth 1997: 112 (in World catalog); Borkent 2000: 866 (discussion of adult characters); Borkent & Spinelli 2000: 55 (in New World catalog south of the USA; distribution); Huerta 2002: 5 (female, male, in key, figures, distribution); Borkent & Spinelli 2007: 87 (in Neotropical catalog; distribution); Borkent & Grogan 2009: 22 (in Nearctic catalog north of Mexico; distribution); Grogan et al. 2010: 41 (Florida, Jamaica records; comparison with *S. bulla* Thomsen); Grogan et al. 2013: 10 (Guadeloupe records).

Stilobezzia scutata Lane & Forattini 1961: 92 (Panama).

Schizonyxhelea thomsenae (Wirth); Borkent 2014: 66 (combination); Borkent 2016: 134 (in online World catalog); Huerta & Grogan 2017: 404 (in key, Guatemala, Mexico records).

Adult diagnosis (Fig. 1a-e). The only New World species of *Schizonyxhelea* with the following combination of characters: distal 1/2 of hind femur with a dark brown stripe, distal apex of hind tibia dark brown and wing membrane without macrotrichia beyond costa to wing tip; female with abdominal terga 2-6 with two anterior small white spots, 2-6 dark brown except pale mid area which is much broader in the tergite 4; male gonostylus with a basal thumb-like appendage.

Description female pupa: (Figs. 2a-b, d-h, 3a-c, e). Habitus as in Fig. 2a. Total length 1.97 mm. Exuviae general coloration yellowish, except cephalotorax pale brown. **Head:** Dorsal apotome (Fig. 2b) with surface smooth, lateral margins with row of thick spines, posterior margin

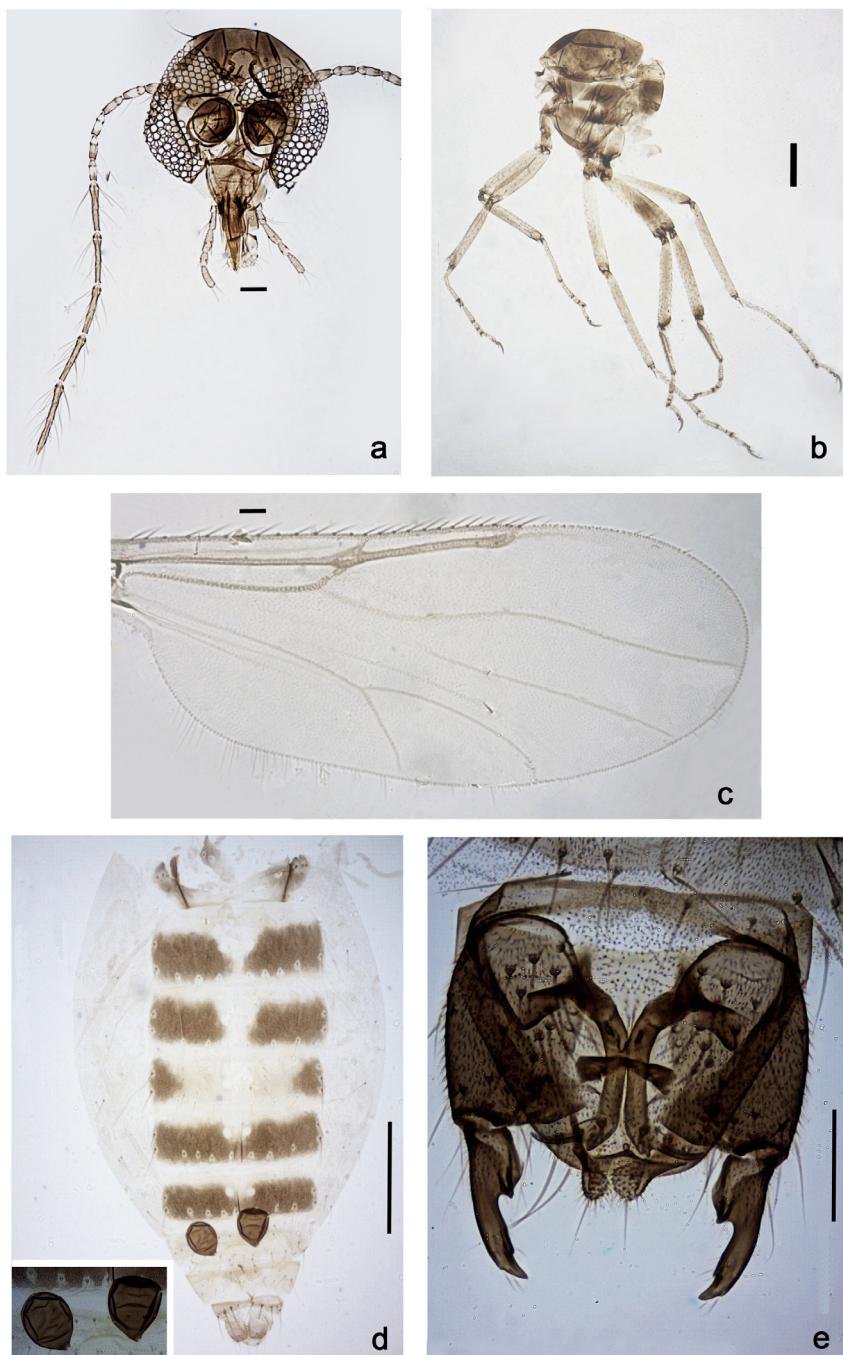


Figure 1. Photographs of *Schizonyxhelea thomsenae* (Wirth) (a-e) adult, (a-d) female, (e) male. (a) Head capsule with flagellum, dorsal view. (b) Thorax. (c) Wing (d) Abdomen with detail of spermathecae. (e) Genitalia.

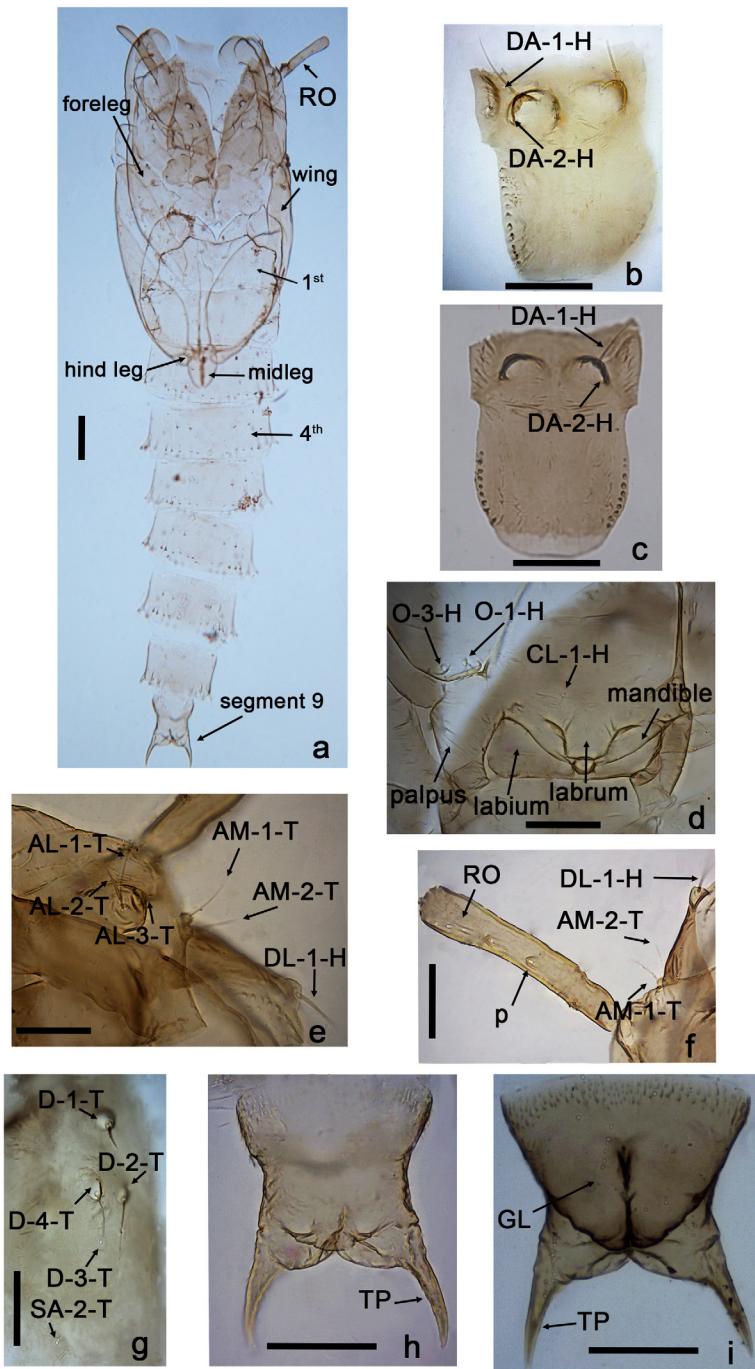


Figure 2. Photographs of *Schizonyxhelea thomsenae* (Wirth) (a-b, d-h) female pupa, (c, i) male pupa. (a) Entire pupa, ventral view. (b-c) Dorsal apotome. (d) Clypeal/labral sensillum and ocular sensilla (ventral view). (e) Cephalothoracic sensilla (dorsal view); (f) Respiratory organ. (g) Dorsal seta. (h-i) Segment 9 (ventral view). Anterolateral sensilla (AL-1-T, AL-2-T, AL-3-T); anteromedial sensilla (AM-1-T, AM-2-T); clypeal/labral sensillum respiratory organ (RO); supraalar sensillum (SA-2-T); terminal process (TP). (CL-1-H); dorsal apotome sensilla (DA-1-H, DA-2-H); dorsolateral cephalic sclerite sensilla (DL-1-H); dorsal seta (D-1-T, D-2-T, D-3-T, D-4-T); genital lobe (GL); ocular sensilla (O-1-H, O-3-H); pedicel (p);

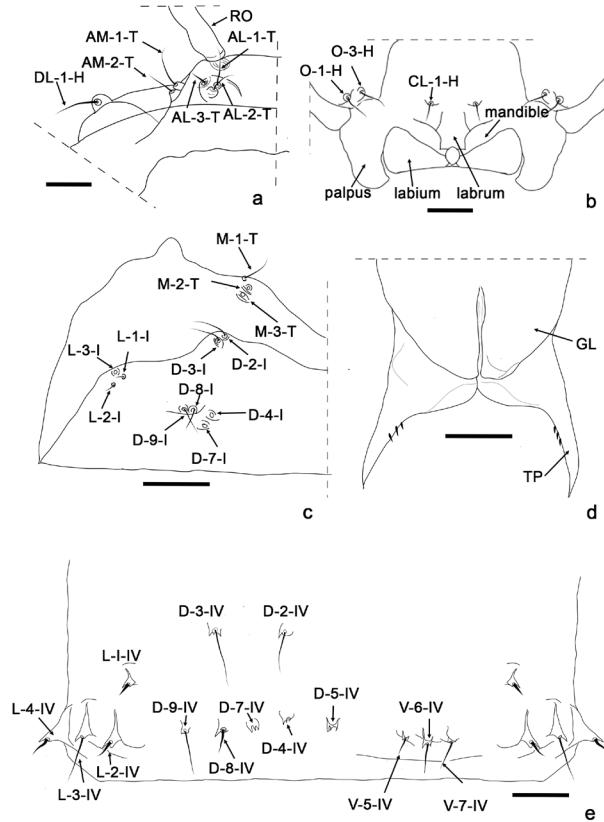


Figure 3. *Schizonyhelea thomsenae* (Wirth) (a-c, e) female pupa, (d) male pupa. (a) Cephalothoracic sensilla (dorsal view). (b) Clypeal/labral sensillum and ocular sensilla (ventral view). (c) Metathorax and tergite 1. (d) Segment 9. (e) Segment 4.

Anterolateral sensilla (AL-1-T, AL-2-T, AL-3-T); anteromedial sensilla (AM-1-T, AM-2-T); dorsolateral cephalic sclerite sensillum (DL-1-H); genital lobe (GL); metathoracic sensilla (M-1-T, M-2-T, M-3-T); segment 4 sensilla (D-2-IV, D-3-IV, D-4-IV, D-5-IV, D-7-IV, D-8-IV, D-9-IV, L-1-IV, L-2-IV, L-3-IV, L-4-IV, V-5-IV, V-6-IV, V-7-IV); tergite 1 sensilla (D-2-I, D-4-I, D-7-I, D-8-I, D-9-I, L-1-I, L-2-I, L-3-I); terminal process (TP).

hyaline; DAL 0.13 mm; DAW 0.14 mm; DAW/DAL 0.92. Sensilla: dorsal apotomal sensilla (Fig. 2b): DA-1-H elongate, stout seta, DA-2-H campaniform sensillum; dorsolateral cephalic sclerite sensillum: DL-1-H long, stout seta (Figs. 2e-f and 3a); clypeal/labral sensillum (Figs. 2d and 3b): CL-1-H long, thin seta; ocular sensilla (Fig. 2d): O-1-H, O-3-H long, thin setae. Cephalothorax with surface smooth, length 0.97 mm, width 0.57 mm. **Thorax:** respiratory organ length/width= 7.50,

elongate, slender, smooth with 6-7 apical pores closely abutting apex, arranged in single curved row and 4-5 lateral pores, ROL 0.19 mm, ROW 0.025 mm, pedicel yellowish, length 0.035 mm (Figs. 2a-f and 3a); P/RO 0.19. Sensilla as follows: two anteromedial sensilla (Figs. 2e and 3a): AM-1-T, AM-2-T long, stout setae, AM-1-T thicker than AM-2-T, separated on a single tubercle; three anterolateral sensilla (Figs. 2e and 3a): AL-1-T, AL-2-T long, thin setae, AL-3-T medium-sized, stout

seta, 4 dorsal setae (Fig. 2g): D-1-T thicker than others, D-2-T, D-4-T long setae, D-2-T stouter than D-4-T, D-3-T campaniform sensillum posterior to D-4-T, D-5-T absent, supraalar SA-2-T campaniform sensillum (Fig. 2g); 3 metathoracic sensilla (Fig. 3c): M-1-T long, thin seta, M-2-T, M-3-T campaniform sensilla. **Abdomen:** without pigmentation pattern, segment 2 as wide than segment 3 with undivided, thick setae, with serrate, short tubercles. Sensilla: tergite 1 (Fig. 3c) with two anterior sensilla, D-2-I, D-3-I long, thin setae; 4 posterior sensilla, D-4-I, D-7-I campaniform sensilla, D-8-I, D-9-I long, thin setae; three lateral sensilla, L-1-I, L-2-I short, thin setae, L-3-I campaniform sensillum; segment 4: (Fig. 3e): D-2-IV, D-3-IV long setae, D-3-IV longer, both on small serrate tubercles, D-4-IV, D-5-IV, D-7-IV campaniform sensilla, D-8-IV medium-sized, thick seta, D-9-IV long, thick seta, all on small serrate tubercles; lateral setae thicker; L-1-IV, L-2-IV, L-4-IV medium-sized, thick setae, L-3-IV long, thick seta, all located on serrate tubercles, V-5-IV, V-7-IV medium-sized, thick setae, V-6-IV long, thick seta all on stout, serrate tubercles. Segment 8 without D-3-VIII, L-1-VIII. Segment 9 (Fig. 2a and 2h) ventral surface covered with small spinules, length 0.28-0.30 (0.29, n=3) mm, width 0.17-0.19 (0.18, n=3) mm; terminal processes with basal inner margin slightly convex, each projecting posterolaterally, tapering to pointed apex, with a posterior row of 3-4 thick spines (Fig. 2h), with D-5-IX, D-6-IX campaniform sensilla; length 0.16-0.18 (0.17, n=3).

Description male pupa: (Figs. 2c, i and 3d). Similar to female with usual sexual differences. Total length 2.22 mm. Exuvium pale brown, except cephalotorax brown. Dorsal apotome (Fig. 2c) with DAL 0.12 mm; DAW 0.15 mm, DAW/DAL 1.25. Respiratory organ: RO length 0.20 mm, RO width 0.025 mm; pedicel length 0.035 mm. Cephalotorax: length 0.92 mm, width 0.57 mm. Segment 9 (Figs. 2i and 3d) length 0.22

mm, width 0.19 mm; terminal processes with a posterior row of 3 thick spines (Fig. 3d), with D-5-IX, D-6-IX campaniform sensilla present at base; length 0.13 mm, genital lobe short (Figs. 2i and 3d) not extending beyond of posterior margin of segment.

Distribution: Argentina (Buenos Aires, Chaco, Córdoba, Corrientes, Formosa and Misiones provinces), Brazil (Bahia), Guadeloupe, Guatemala, Jamaica, Mexico (Guerrero, Jalisco, Tabasco, Veracruz and Yucatan states), Panama, Perú (Cuzco) and USA (Florida).

Type material: Holotype male on microscope slide, labeled "USA, Everglades City, Collier County, Fla, 7-II-1950, Davidson, light trap" (USNM).

Material examined: Jamaica, Westmoreland Parish Negril, Crystal Waters Tropical hammock, 20-XI-1968, R. E. Woodruft, 2 males, 2 females, blacklight trap (USNM). **New Peru records:** Cuzco, Kirigueti, VIII-2004, 2 males, 1 female, J. Williams, at light; same data except Pagoreni, 11°42'21.9" S 72°54'21.9" W, 1 male. **New Brazil records:** Bahia, Bahia Itabuna, Fazenda Almirante, 6-VIII-1993, 1 male, J. Winder (USNM); same data except 29-XII-1989, 2 males; same data except 3-VI-1988, 1 female; same data except 3-I-1990, 1 female; same data except 7-II-1990, 2 males; same data except 19-II-1990, 1 male. **New Argentina records:** Buenos Aires: Reserva Natural Integral Punta Lara, 34°47'28" S 57°59'49" W, marginal forest, 10/11-XII-2014, 3 males, 4 females, Cazorla-Campos, CDC light trap; same data except 3/4-II-2015, 3 females; Chaco, Parque Nacional Chaco, 26°48'20.1" S 59°36'32" W, 24/27-IX-2009, 3 males, 1 female, Cazorla-Melo, Malaise trap; Córdoba, La Higuera, 28-II-2000, 2 males, 2 females, D. Carpintero, at light; Corrientes, Colonia Pellegrini, 12-XI-2007, 1 male, G. Spinelli, sweep net; Estancia San Nicolás, 26°07'41.0"S 57°26'3.5"W, 16-IX-2009, 1 female, G. Spinelli; Formosa, Estancia La Marcela, 35 km E

El Colorado, 27/28-VII-2003, 2 males, 3 females, J. Williams, at light; same data except V-2005, 1 male, F. Brusa, at light; Ing. Juarez, V-2005, 1 male, H. Calandra, at light; Misiones, charca, RP10 (15 km de Apóstoles); 27°54'55.2" S 55°38'21.1" W, 128 m, 10-XII-09, 1 male (with pupal exuvium), A. Siri, kick net; same data except Garabí, (RP94); 28°13'44.9" S 55°48'34.4" W; 80 m, 1 female (with pupal exuvium); same data except sweep net, 1 male; Posadas, 9-I-2002, 2 males, G. Spinelli, CDC light trap; Puerto Iguazú, arroyo Mbocay, 24-VIII-2008. 1 male, H. Walantus, CDC light trap.

TAXONOMIC DISCUSSION

Adult specimens of *Schizonyxhelea thomsenae* strongly resemble those of the Neotropical species *Schizonyxhelea zoologica* in size, fused flagellomeres 5-9 or 5-10 and dark coloration of the hind femur, however the later species can be distinguished by the male genitalia with the gonostylus bearing three distinctive crenulations on the mesal surface, and the parameres elongated, slender and rod-like.

Schizonyxhelea bulla, *S. forattinii* and *S. panamensis* are the only species of the genus whose pupae are currently known. The pupa of *S. forattinii* is well described and illustrated, while the description of the pupa of *S. bulla* is briefer. Both species share with *S. thomsenae* the following characters: presence of thick spines on the lateral margin of the dorsal apotome, dorsal apotomal sensilla (DA-1-H) represented by an elongate and stout seta, D-1-T much stouter than the other dorsal setae, and short terminal processes. However, *S. bulla* can be distinguished by the presence of 2 clypeal/labrals and the respiratory organ with 4-7 apical and 3-4 lateral pores, while *S. forattinii* differs by the short anteromedial setae, the respiratory organ with 5-6 apical and 4 lateral pores and the V-6-IV medium-sized, thin seta. The pupal description of *S.*

panamensis is even briefer and incomplete, and it differs from *S. thomsenae* by the respiratory organ with 7 apical and 2 lateral separated pores on mid portion, and the terminal process which is similar in shape but apparently lack the posterior row of thick spines.

Borkent (2014) in his analysis of the phylogenetic relationships between genera, studied pupal exuviae of *S. forattinii* and *S. bulla* and interpreted some of the similarities between them as synapomorphies of the genus, as follows: 1, D-1-T much more stout than other dorsal setae; 2, tergite 1 with 1-2 lateral setae; 3, dorsal surface of abdominal segment 9 with two short medial tubercles and 4, terminal process with a posterior row of thick spinules. Our study of the pupal exuviae of *S. thomsenae* confirms that character states 1 and 4 are shared (both character states are apomorphic conditions), while the character state 3, medial tubercles of segment 9 (apomorphic conditions), is difficult to see in the two studied specimens. Finally, the tergite 1 of *S. thomsenae* presents 3 lateral setae, which is an apomorphic condition shared with most of the species of the subfamily Ceratopogoninae, unlike the other two species of this genus. Further studies on more species of *Schizonyxhelea* and with a larger number of specimens are necessary to reveal if the number of lateral setae of the tergite 1, as well as the number of clypeal/labrals sensilla are variable features in this genus.

It is also worth mentioning that the environments in which the immatures were found are similar: *S. bulla* was obtained from mud from the margins of a marsh, from the grassy margins of a swamp and from weeds of a pond and *Sphagnum* L.; the larva of *S. forattinii* was collected from wet mud in a small seep which flowed into the outflow of a larger spring,

and the pupae of *S. thomsenae* were collected in a flooded, grassy pond.

Finally, it should be noted that Anjos-Santos et al (2017) in their key for Neotropical pupae of the genus *Stilobezzia* Kieffer 1911 included by mistake *Schizonyxhelea panamensis* as *Stilobezzia panamensis*.

KEY TO PUPAE OF *Schizonyxhelea*

1- Terminal process segment 9 with a posterior row of thick spines; respiratory organ with at least 3 lateral pores**2**

1' - Terminal process of segment 9 without a posterior row of thick spines; respiratory organ with 2 lateral pores***S. panamensis***

2- Two minute clypeal/labral sensilla; tergite 1 with 1-2 lateral setae.....**3**

2'- One medium-sized clypeal/labral sensillum; tergite 1 with 3 lateral setae.....***S. thomsenae***

3- Terminal process with basal inner margin convex and dark tips; respiratory organ with 4-7 apical and 3-4 lateral pores***S. bulla***

3'- Terminal process with basal inner margin straight and pale tips; respiratory organ with 5-6 apical and 4 lateral pores***S. forattinii***

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Author contributions

CC identified the species, mounted the material and wrote the manuscript; CC and FD redescribed the specimens, FD made up the plates; CC and FD reviewed the final manuscript.

