

**A new species of *Dasyhelea* from Brazilian Amazonas
and the description of the male of *D. paulistana*
(Diptera: Ceratopogonidae)**

Florentina DIAZ^{1,4}), Sharlene R. S. TORREIAS²), Gustavo R. SPINELLI¹)
& María M. RONDEROS¹)

¹) División Entomología, Museo de La Plata, UNLP, Paseo del Bosque s/n, B1900WA La Plata, Buenos Aires, Argentina; e-mail: mfdiaz@fcnym.unlp.edu.ar, spinelli@fcnym.unlp.edu.ar

²) Av. Torquato Tapajós, João Bosco, Bloco 8 Flores, CEP. 69048-660, Manaus, Amazonas, Brazil; e-mail: rtorreias@gmail.com

³) CEPAVE, CONICET, 2 n° 584, B1902CHX, La Plata, Buenos Aires, Argentina; e-mail: ronderos@fcnym.unlp.edu.ar

⁴) corresponding author

Abstract. A new species, *Dasyhelea pseudopollinosa* Díaz & Ronderos sp. nov., is described from Brazil, illustrated and photographed as pupae and male and female adults. The male of *D. paulistana* Forattini & Rabello, 1957 is described for the first time, and the pupa and female adult are redescribed. Pupae of both species were collected from mats of floating fern leaves (Salviniaceae and Azollaceae) in Brazil and Argentina, and exhibit a respiratory organ which is thick at the base and tapering to a sharp apex, and segment 9 with elongate, slender apicolateral process. Differences between the two species are established, and they are also compared with Nearctic congeners that have the same kind of pupa: *D. pollinosa* Wirth, 1952, *D. traversae* Thomsen, 1935, and *D. chani* Wirth & Linley, 1990.

Key words. Diptera, Ceratopogonidae, *Dasyhelea pseudopollinosa* sp. nov., *D. paulistana*, adult, pupa, Argentina, Brazil, Neotropical Region

Introduction

Dasyhelea Kieffer, 1911 is a large and complex genus of Ceratopogonidae with diverse morphology and biology which occurs worldwide in a variety of small aquatic habitats (WAUGH & WIRTH 1976). Taxonomically, the recognition of subgenera and/or species groups is still incipient and generally has been applied intermittently only to various regional faunas (DÍAZ et al. 2009). BORKENT (2014) listed 67 species for the Neotropics, of which seven belong to the *grisea* species-group as defined by WAUGH & WIRTH (1976). Of these, three species are

known also from their immatures, each one with a different kind of breeding site: *D. necrophila* Spinelli & Rodriguez, 1999 inhabits artificial containers and rock holes containing water, *D. correntina* Ronderos & Díaz, 2004 is found in mud, and *D. paulistana* Forattini & Rabello, 1957 is associated with leaves of aquatic ferns of the families Salviniaceae and Azollaceae.

The purpose of this paper is to describe from adult and pupal stages a new species from Amazonas, and to describe for the first time the male and redescribe the pupa and the adult female of *D. paulistana*. The studied material was recently collected in the vicinities of Manaus, Brazil, as well as in northeastern Argentina.

Material and methods

The pupae of both species were collected with a pipette from mats of floating aquatic ferns (*Salvinia auriculata* Aubl., Salviniaceae, and *Azolla filiculoides* Lam., Azollaceae) in Lago Grande, Lago Camaleão on Ilha da Marchantaria, Município de Iranduba, Amazonas, Brazil, and pupae of *D. paulistana* were also sampled from flooded areas along a road margin near Bella Vista, Corrientes province, Argentina, and in a temporary pond in the Estación Biológica Corrientes (EBCO), located 20 km south of Corrientes city. Collected pupae were isolated in a vial with a drop of water and observed daily until adult emergence. Adults were allowed to harden for 24 hours before being preserved to ensure their pigmentation was complete.

Pupal exuviae and adults were mounted in Canada balsam and observed with a compound microscope following the technique described by BORKENT & SPINELLI (2007). Illustrations were made with pen and ink using an attached camera lucida. Photomicrographs were taken with a digital camera Micrometrics SE Premiun, through a Nikon Eclipse E200 microscope.

Terms for structures of adults follow those in the Manual of Central American Diptera (BROWN et al. 2009). For terminology of pupa see BORKENT (2012), with the addition of the following abbreviations of measurements: DAL, dorsal apotome length, and DAW, dorsal apotome width.

The material examined is deposited in the following collections:

INPA Instituto Nacional de Pesquisas da Amazônia, Manaus, Brazil;
MLPA División Entomología, Museo de La Plata, Argentina.

Taxonomy

Dasyhelea pseudopollinosa Díaz & Ronderos sp. nov.

(Figs 1–32, 62)

Type material. HOLOTYPE: ♂ (with 'ex pupa') on microscope slide, labelled: "HOLOTYPE" *Dasyhelea pseudopollinosa* Díaz & Ronderos. BRAZIL: AMAZONAS: Iranduba, Ilha da Marchantaria, Lago Grande, 03°14'43.9"S 59°58'54.2" W, 17.v.2011, Torreias-Ferreira-Keppler lgt., (INPA). ALLOTYPE: ♀ (with 'ex pupa'), same data as holotype. PARATYPES: 5 ♂♂ (with 'ex pupa') (4 in INPA, 1 in MLPA) and 1 ♀ (with 'ex pupa'), same date as holotype (MLPA); 1 ♂ (with 'ex pupa'), same data except: Lago Camaleão, 03°13'14.9"S; 59°56'52.6"W, 20.v.2011 (MLPA).

Diagnosis. Only Neotropical species of the *grisea* species-group with the following combination of characters: male with posterolateral arms of aedeagus bearing a subapical, outer,

pointed process; female with legs uniformly pale brown and subgenital plate represented by a slender transverse band. Pupa with respiratory organ thick at the base and distal half tapering to a sharp apex, with 18–20 apical pores, and terminal processes as long as length of body of segment 9.

Description. Male (Figs 1–4, 21–24). Similar to female with the usual sexual differences. Flagellum as in Fig. 1. Palpus (Fig. 2) with segment 3 bearing scattered sensilla on surface; PR 2.00–2.60 (2.45, $n = 6$). Scutellum with 6–7 strong, 2 thinner setae. Wing (Fig. 3) length 0.93–0.99 (0.95, $n = 4$) mm, width 0.24–0.30 (0.27, $n = 4$) mm, CR 0.47–0.51 (0.48, $n = 4$). Genitalia (Figs 4, 21): tergite 9 tapering distally, distal margin rounded; apicolateral process short, stout, with apical seta; cercus small, with 1–2 setae; sternite 9 (Figs 4, 22) $0.3\times$ longer than greatest width, distal margin slightly convex. Gonocoxite stout, $1.5\times$ longer than greatest width, with blunt anteromedian process; gonostylus $0.8\times$ longer than gonocoxite, narrow base, nearly straight, tip pointed, spur-like. Paramere and gonocoxal apodemes forming an asymmetrical structure (Figs 4, 23); gonocoxal apodeme stout, curved, not in contact with paramere, latter elongate, straight, tapering to slender blunt apex. Aedeagus (Figs 4, 24) nearly as long as greatest width, basal arch slightly concave, low, extending to 0.15 of total length; basal arms stout, directed anterolaterally; posterolateral arms stout, slightly convergent, each tapering to pointed tip with subapical, outer pointed process.

Female (Figs 5–11, 25–27). Head dark brown (Fig. 5) Eyes contiguous by width of 2 ommatidia. Antenna with flagellum (Fig. 5) brown; AR 0.94–0.97 (0.95, $n = 2$). Frontal sclerite semicircular (Fig. 25), with long, slender, posteromesally directed projection. Clypeus (Fig. 6) with three pairs of setae. Palpus (Figs 5, 7) whitish, segment 5 slightly infuscated; segment 3 with 6–7 capitate sensillae, PR 1.86 ($n = 2$).

Thorax (Fig. 8). Scutum brown, scutellum pale, with 6–7 strong, 1–2 thinner setae. Legs (Fig. 9) uniformly pale brown, tarsomeres 5 infuscated; apex of hind tibia with 5 spines; foreleg TR 2.37–2.50 (2.43, $n = 2$), midleg TR 2.41–2.44 (2.42, $n = 2$), hind leg TR 2.31–2.36 (2.33, $n = 2$). Wing (Fig. 10), length 0.96 ($n = 2$) mm, width 0.36–0.39 (0.37, $n = 2$) mm, CR 0.50–0.56 (0.53, $n = 2$); membrane hyaline, densely covered with macrotrichia; cubital fork at level of first radial cell. Halter knob whitish.

Abdomen. Pale brown. Subgenital plate (Figs 11, 26) represented by slender transversal band, slightly arised anteromesally. Spermatheca ovoid (Figs 11, 27), heavily sclerotized, diameter 252 μm , neck short, sclerotized, measuring 12 μm .

Female pupa (Figs 13–19, 28–32). Total length 2.88 mm. General coloration of exuviae pale brown. Dorsal apotome (Fig. 13) $2.11\times$ broader than long, surface smooth with wrinkles on margins, anterior margin slightly concave, posterior margin rounded; dorsal apotome sensilla with one short, thin seta, one campaniform sensillum (Fig. 13); DAL 0.10–0.12 (0.11, $n = 2$) mm; DAW 0.240 ($n = 2$) mm; DAW/DAL 2.00–2.22 (2.11, $n = 2$). Cephalothorax surface with very small rounded tubercles, length 0.99–1.02 (1.00, $n = 2$) mm, width 0.69–0.75 (0.72, $n = 2$) mm. Cephalothoracic sensilla as follows: three dorsolateral cephalic sensilla (Figs 14, 28), two minute setae, one campaniform sensilla; three anterolateral sensilla (Figs 14, 28), one long, thin seta, one medium-sized seta, one coeloconica sensillum; two anteromedial sensilla, one medium-sized seta, one short seta (Figs 14, 28); dorsals (Figs 15, 29): D-1-T,

D-2-T, D-4-T absent, D-3-T campaniform sensillum, D-5-T short, thin seta, supraalar (SA-2-T) campaniform sensillum. Respiratory organ (Fig. 16) thick at base, distal half tapering to a sharp apex, $4.75\times$ longer than broad, surface smooth, with 18–20 apical pores; RO length 0.23–0.25 (0.24, $n = 2$) mm, RO width 0.05 ($n = 2$) mm; pedicel (P) pale brown, smooth, short, pedicel length 0.024 ($n = 2$) mm, P/RO 0.100–0.105 (0.1025, $n = 2$) mm. Two clypeal/labral sensilla (Figs 17, 30), CL-1-H long, thin seta, CL-2-H short seta; two ocular sensilla, one long, thin seta, one campaniform sensillum (Figs 17, 30). Abdominal segments covered with small spinules. Abdominal segment 1 (Figs 18, 31) with sensilla as follows: D-2-I peg; D-4-I, D-7-I pores, D-8-I short seta; 3 lateral sensilla, L-1-I, L-3-I short setae, L-2-I minute seta. Second abdominal segment similar to first. Segment 4 with sensillar pattern (Fig. 32) as follows: D-2-IV peg; D-4-IV, D-7-IV, D-8-IV without setae, all located on flattened tubercles; L-1-IV medium-sized, thin seta on triangular tubercle, L-2-IV campaniform sensillum, L-3-IV, L-4-IV minute setae, all located on triangular tubercles; two ventral sensilla, V-5-IV without seta, V-6-IV short, stout seta, both located on bifid tubercles. Segment 9 (Fig. 19) $1.45\times$ longer than width, ventral surface with many spinules; length 0.348–0.360 (0.354, $n = 2$) mm, width 0.204–0.240 (0.222, $n = 2$) mm. Terminal process (Fig. 19) straight, as long as length of body of segment 9, tip pointed, base broad with two setae, one long, thin, hyaline seta on small rounded base, other medium-sized, stout seta on rounded tubercle; length 0.180 ($n = 2$) mm.

Male pupa (Figs 12, 20). Similar to female with usual sexual differences: Total length 2.52–2.64 (2.61, $n = 6$) mm. Exuviae pale brown. Dorsal apotome with DAL 0.12–0.14 (0.130, $n = 6$) mm; DAW 0.24–0.26 (0.25, $n = 6$) mm, DAW/DAL 1.85–2.10 (1.98, $n = 6$). Respiratory organ (Fig. 12), RO length 0.22–0.24 (0.23, $n = 7$) mm, RO width 0.05 ($n = 7$); pedicel length 0.024–0.030 (0.025, $n = 7$) mm, P/RO 0.10–0.13 (0.107, $n = 7$). Cephalothorax (Fig. 12) length 0.99–1.07 (1.03, $n = 6$) mm, width 0.66–0.74 (0.70, $n = 6$) mm. Segment 9 (Fig. 20) length 0.34–0.39 (0.35, $n = 7$) mm, width 0.19–0.24 (0.22, $n = 7$) mm; terminal process (Fig. 20) length 0.17–0.24 (0.19, $n = 7$) mm.

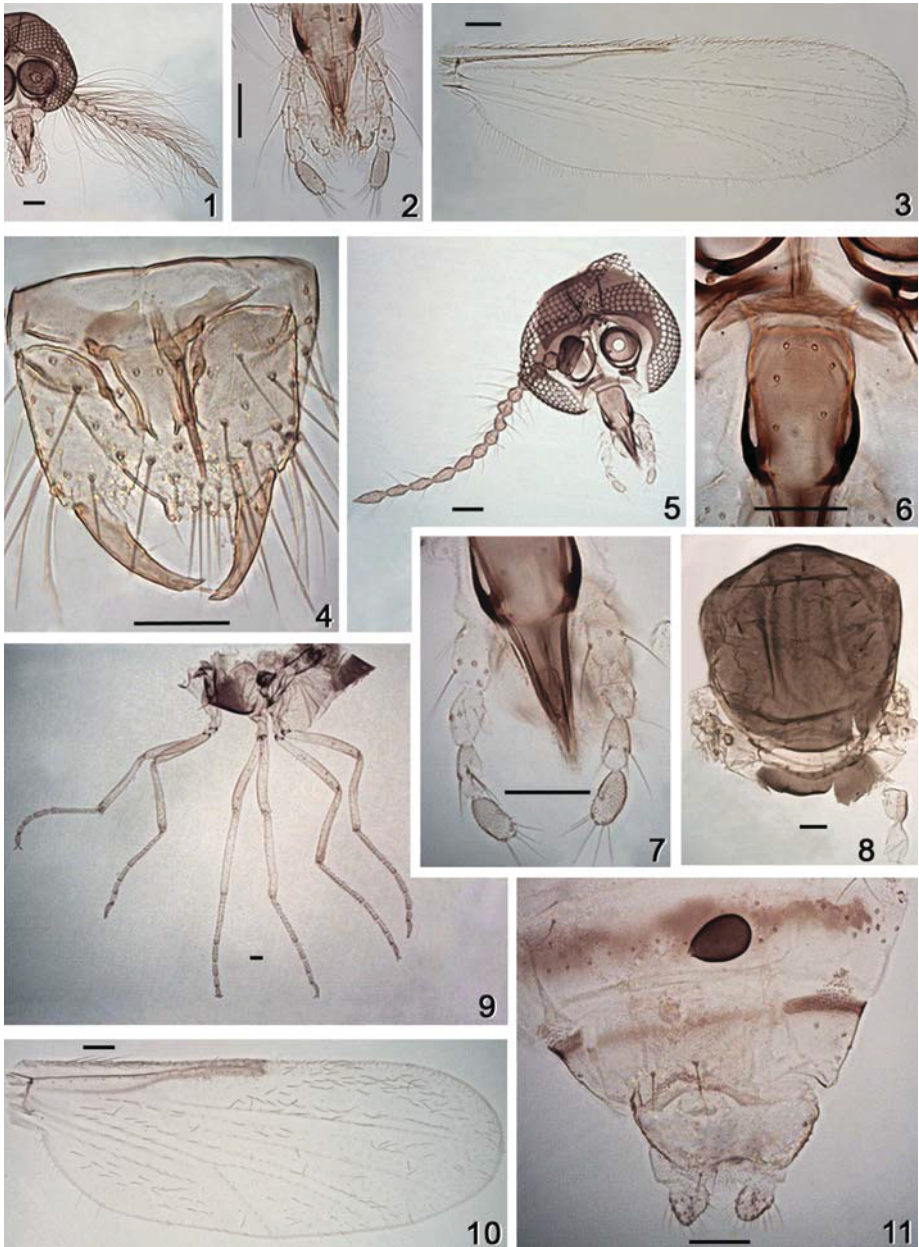
Etymology. The species name *Dasyhelea pseudopollinosa* refers to the similarity with its congener, Nearctic *Dasyhelea pollinosa* Wirth, 1952.

Distribution. Brazil (Amazonas).

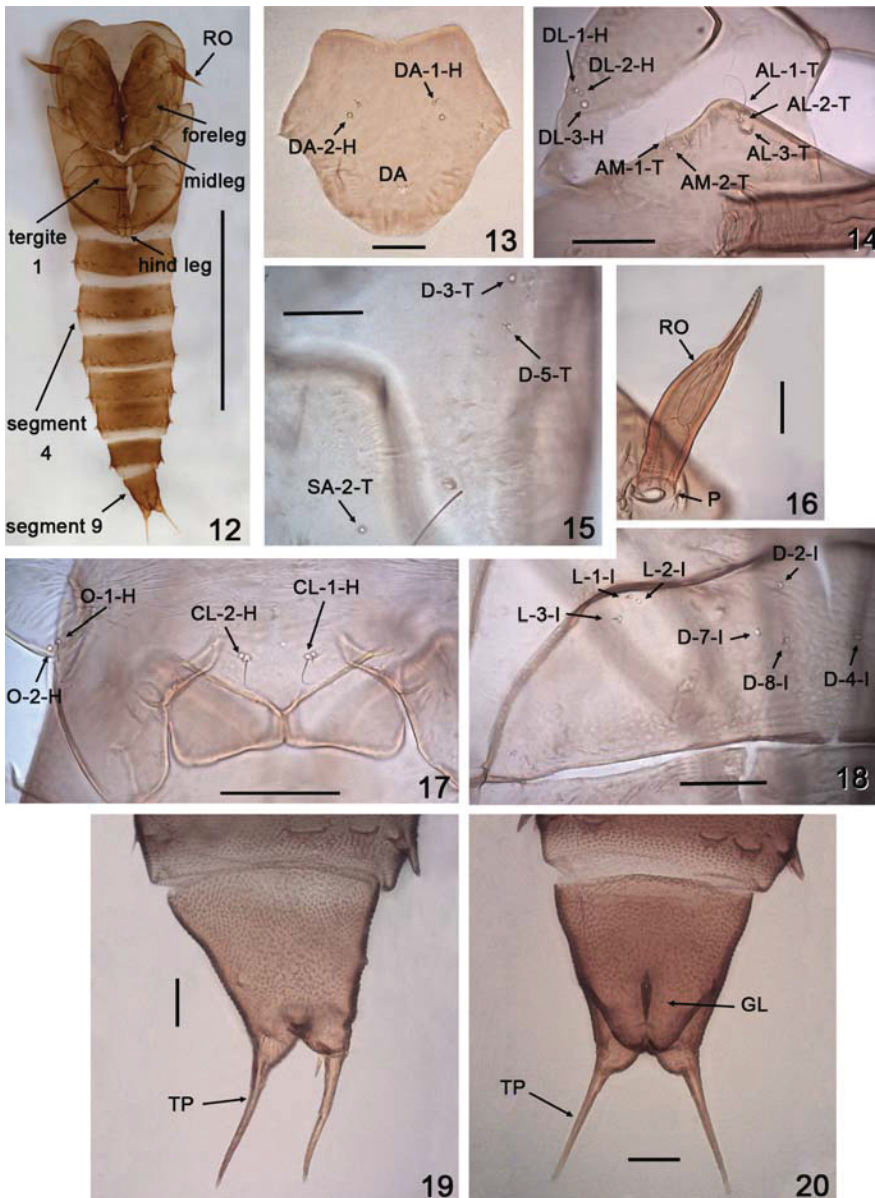
Discussion. WAUGH & WIRTH (1976) proposed the *cincta*, *grisea*, *leptobranchia* and *mutabilis* species-groups for the Nearctic *Dasyhelea* species, working in an exclusively phenetic framework. Most of the Neotropical species fit within these groups or are in the *brevicosta* species-group proposed by GROGAN & WIENERS (2006) and the *patagonica* species-group proposed by DÍAZ et al. (2010).

We provisionally place this new species within the *grisea* group by virtue of the well developed frontal sclerite and sinuate paramere. On the other hand, the uniformly pale brown legs, the female subgenital plate represented by a transversal sclerotized band without anterior triangular projection and the posterolateral arms of aedeagus bearing an outer subapical pointed process, are not characters otherwise present in species of the *grisea* group.

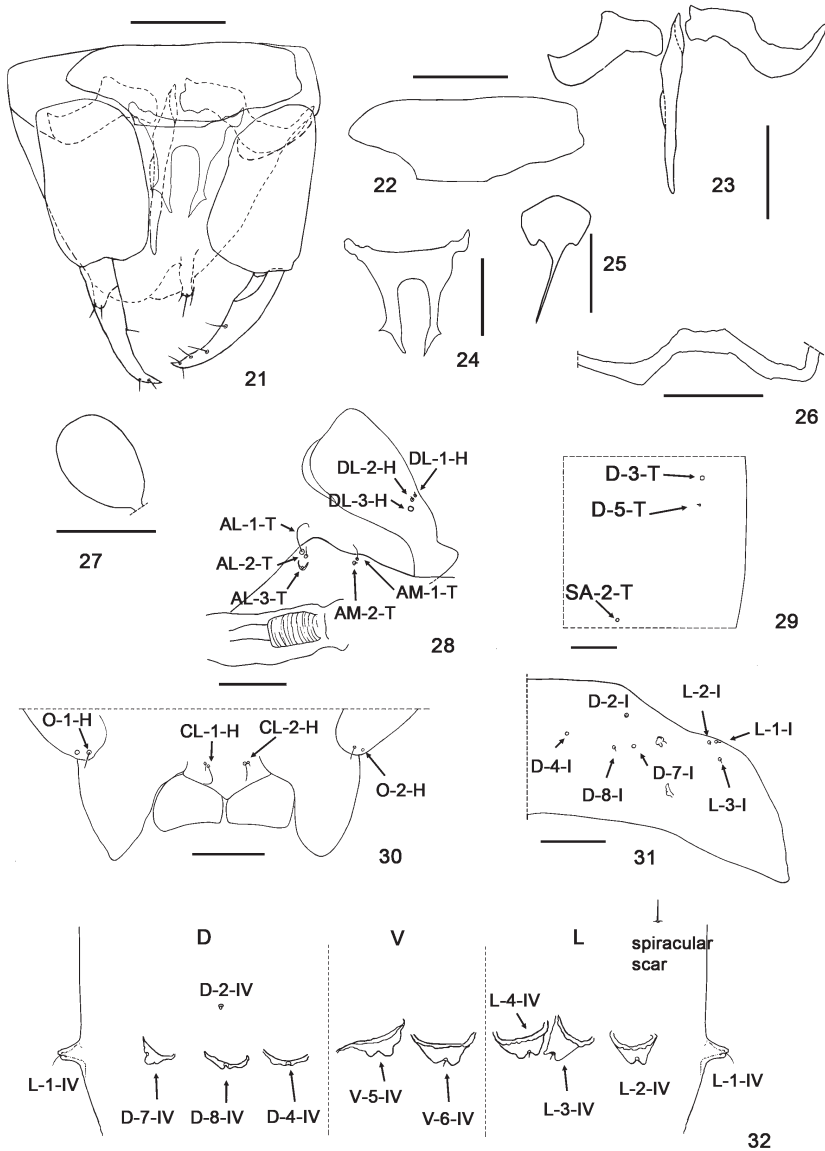
The pupa of *Dasyhelea paulistana* is very similar to this new species by virtue of the respiratory organ thick at base with its distal half tapering to a sharp apex, and by the slender and elongate terminal processes of the segment 9 but in *D. paulistana* the terminal processes are



Figs 1–11. *Dasyhelea pseudopollinosa* Díaz & Ronderos sp. nov., adult. 1–4 – holotype, male; 5–11 – allotype, female. 1, 5 – flagellum, anterior view; 2, 7 – palpus; 3, 10 – wing; 4 – genitalia, ventral view; 6 – clypeus; 8 – thorax; 9 – legs; 11 – abdominal segments 8–10, ventral view.



Figs 12–20. *Dasyhelea pseudopollinosa* Díaz & Ronderos sp. nov., pupa. 12, 20 – male pupa; 13–19 – female pupa. 12 – entire pupa; 13 – dorsal apotome; 14 – cephalothoracic chaetotaxy (dorsal view); 15 – dorsal sensilla and supraalar; 16 – respiratory organ; 17 – clypeal/labral sensilla and ocular sensilla; 18 – abdominal segment 1 chaetotaxy; 19–20 – segment 9. Abbreviations: AL-1-T, AL-2-T, AL-3-T – anterolateral sensilla; AM-1-T, AM-2-T – anteromedial sensilla; CL-1-H, CL-2-H – clypeal/labral sensilla; DA – dorsal apotome; DA-1-H, DA-2-H – dorsal apotomal sensilla; DL-1-H, DL-2-H, DL-3-H – dorsolateral cephalic sclerite sensilla; D-3-T, D-5-T – dorsal sensilla; D-2-I, D-4-I, D-7-I, D-8-I, L-1-I, L-2-I, L-3-I – abdominal segment 1 chaetotaxy; GL – genital lobe; O-1-H, O-2-H – ocular sensilla; P – pedicel; RO – respiratory organ; SA-2-T – supraalar; TP – terminal process.



Figs 21–32. *Dasyhelea pseudopollinosa* Díaz & Ronderos sp. nov. 21–24 – holotype, male; 25–27 – allotype, female; 28–32 – female pupa. 21 – genitalia; 22 – sternite 9; 23 – paramere and gonocoxal apodemes; 24 – aedeagus; 25 – frontal sclerite; 26 – subgenital plate; 27 – spermatheca; 28 – cephalothoracic chaetotaxy (dorsal view); 29 – dorsal sensilla and supraalar; 30 – clypeal/labral sensilla and ocular sensilla; 31 – abdominal segment 1 chaetotaxy; 32 – segment 4 chaetotaxy. Scale bars = 0.05 mm. Abbreviations: AL-1-T, AL-2-T, AL-3-T – anterolateral sensilla; AM-1-T, AM-2-T – anteromedial sensilla; CL-1-H, CL-2-H – clypeal/labral sensilla; D-3-T, D-5-T – dorsal sensilla; DL-1-H, DL-2-H, DL-3-H – dorsolateral cephalic sclerite sensilla; D-2-I, D-4-I, D-7-I, D-8-I, L-1-I, L-2-I, L-3-I – abdominal segment 1 chaetotaxy; D-2-IV, D-4-IV, D-7-IV, D-8-IV, L-1-IV, L-2-IV, L-3-IV, L-4-IV, V-5-IV, V-6-IV – segment 4 chaetotaxy; O-1-H, O-2-H – ocular sensilla; SA-2-T – supraalar.

very much longer, twice the body length of segment 9. The adult of *D. paulistana* is readily distinguished from the new species by the ovoid female subgenital plate, the elongate and sinuate paramere hooked at tip, and by the aedeagus with V-shaped basal arch.

The pupae of the Nearctic species *D. pollinosa* and *D. traverae* Thomsen, 1935 are similar to this new species. However, both species can be distinguished from *D. pseudopollinosa* by several characters offered by the adults, as follows: in *D. pollinosa* the legs are somewhat banded, the gonocoxal apodemes are fused with the paramere, and the female subgenital plate exhibits a conspicuous anterior, triangular projection. *Dasyhelea traverae* is a larger species (female wing length 1.21–1.27 mm) with a dark brown scutellum, male tergite 9 rounded, posterior margin of male sternite 9 nearly straight, paramere J-shaped and not in contact with the gonocoxal apodemes, aedeagus with the basal arch narrowed mesally and with straight lateral arms, each one bearing a slender, inner, posteriorly directed process with mesal tooth, and the female subgenital plate exhibits an elongate, slender, anteriorly directed projection.

Dasyhelea paulistana Forattini & Rabello, 1957

(Figs 33–62)

Dasyhelea paulistana Forattini & Rabello, 1957: 245 (pupa, female, Brazil).

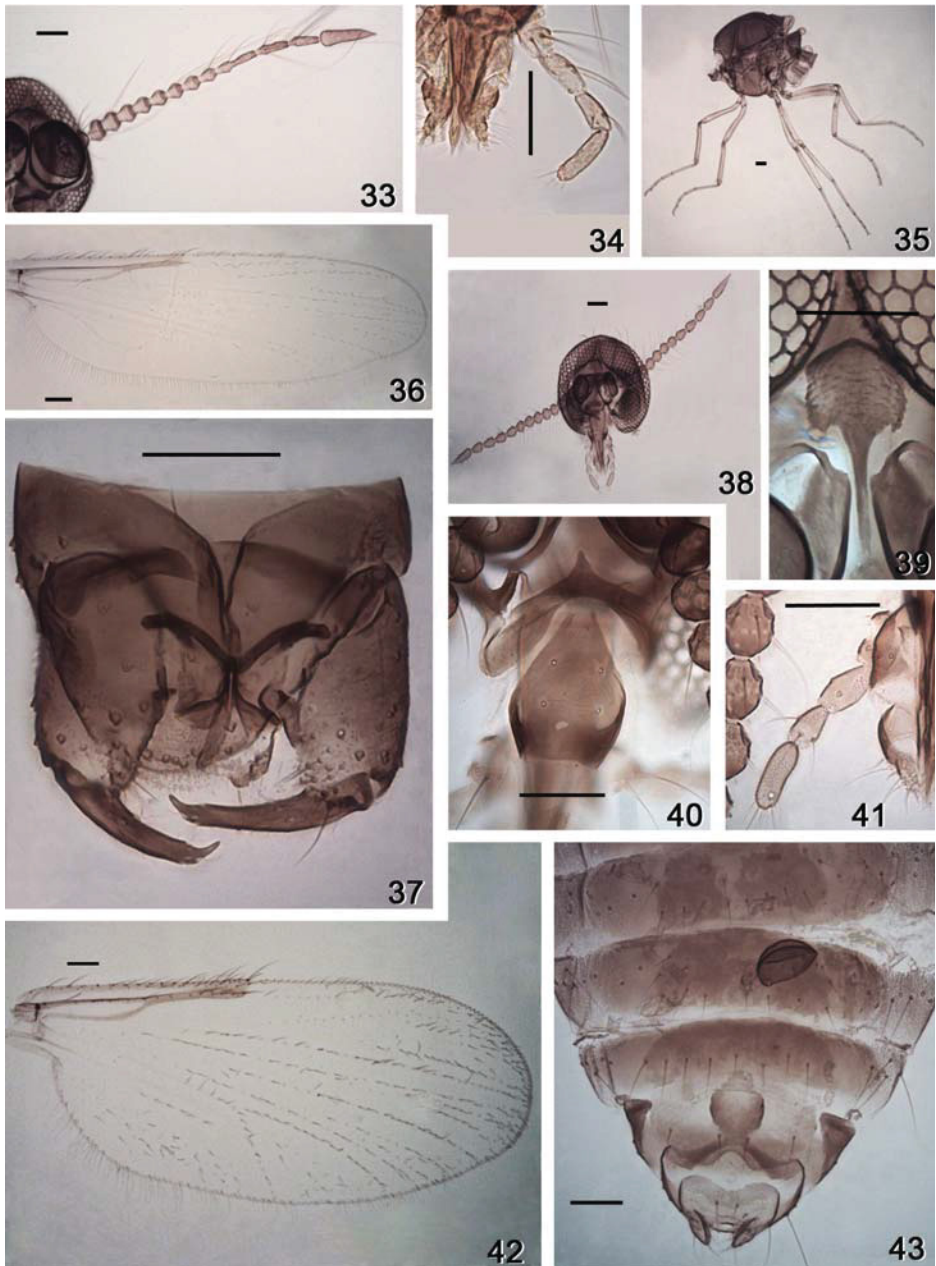
Dasyhelea paulistana: WIRTH (1974): 17 (catalog of species south of the USA); BORKENT & WIRTH (1997): 57 (World catalog); BORKENT & SPINELLI (2000): 25 (catalog of species south of the USA); BORKENT & SPINELLI (2007): 61 (Neotropical catalog); BORKENT (2014): 69 (online catalog).

Type material. HOLOTYPE: ♀ (with ‘ex pupa’), BRAZIL: ESTADO DE SÃO PAULO: cidade de São Paulo, Parque da Agua Funda, 5.xi.1954, O. P. Forattini & E. X. Rabello lgt. (MNRJ).

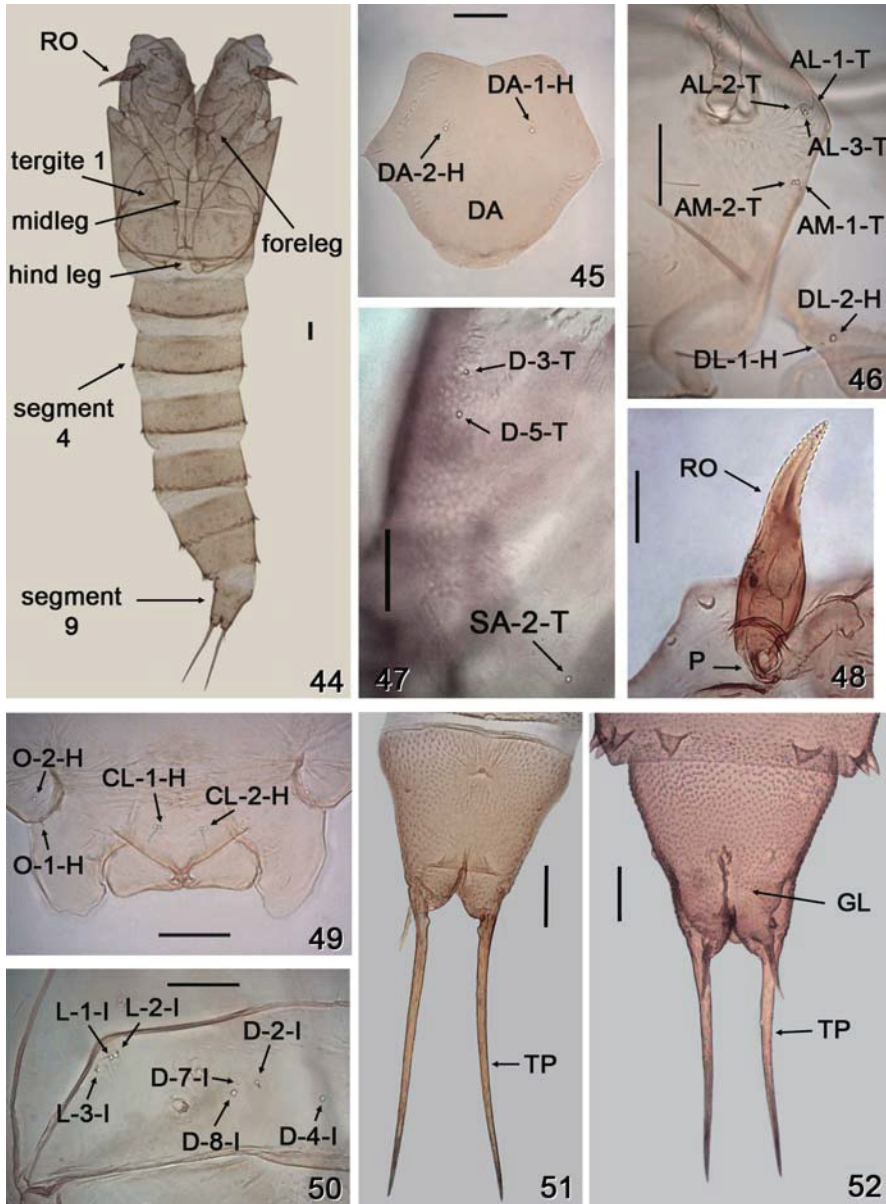
Additional material examined. BRAZIL: AMAZONAS: 3 ♂♂ 2 ♀♀ (all ‘ex pupa’), Iranduba, Ilha da Marchantaria, 28.vi.2005, D. Carrasco-Ferreira-Keppler lgt. (INPA); 2 ♂♂ 2 ♀♀ (all with ‘ex pupa’), Lago Grande, 03°14’43.9”S 59°58’54.2”W, 17.v.2011, Torreias-Ferreira-Keppler lgt. (INPA). ARGENTINA: CORRIENTES: 1 ♀, arroyo Pay Ubre, 29°01’41.2”S, 58°10’26.6”W, 66 m, 12.iii.2010, G. Spinelli lgt., sweep net (MLPA); 1 ♂ (with ‘ex pupa’), Bella Vista, ruta Prov. 27 Km 54, 28°34’30.7”S, 59°02’11.5”W, 16.ix.2010, F. Díaz lgt. (MLPA); 1 ♂ 1 ♀, rio Batel, 28°17’44.1”S, 58°01’51.2”W, 78 m, 8–9.xii.2010, G. Spinelli lgt., sweep net (MLPA); 2 ♂♂ 2 ♀♀ (all with ‘ex pupa’), Estación Biológica Corrientes, esterito, 27°32’51.8”S, 58°40’44.8”W, 52 m, 9.xi.2011, Díaz-Marino lgt. (MPLA); 2 ♂♂ 1 ♀ (all with ‘ex pupa’), same date except: 10.xi.2011 (MPLA).

Diagnosis. Only Neotropical species of the *grisea* species-group with the following combination of characters: adults with female subgenital plate ovoid, and males with elongate, sinuate paramere hooked at tip, and aedeagus with V-shaped basal arch. Pupa with respiratory organ thick at the base and distal half tapering to a sharp apex, with 21–22 apical pores, and terminal processes twice longer than length of body of segment 9.

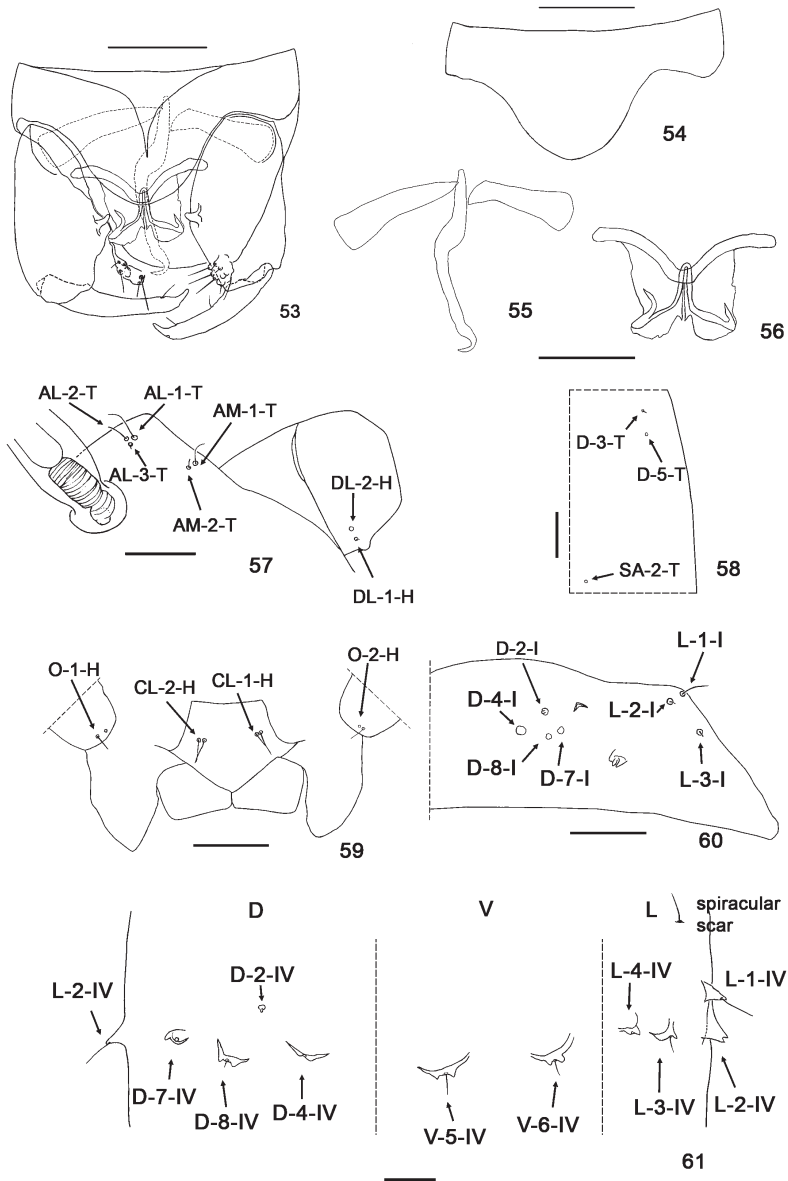
Description. **Male** (Figs 33–37, 53–56). Similar to female with the usual sexual differences. Flagellum as in Fig. 33. Palpus (Fig. 34) with segment 3 bearing scattered sensilla on surface; PR 1.83–2.16 (2.00, n = 9). Scutellum with 6 strong setae. Thorax, legs as in Fig. 35. Wing (Fig. 36) length 0.90–1.20 (1.03, n = 8) mm, width 0.24–0.36 (0.32, n = 8) mm, CR 0.39–0.40 (0.39, n = 8). Genitalia (Figs 37, 53): tergite 9 slightly tapering distally, not extending to level of apex of gonocoxite; apicolateral process stout, rounded, with subapical seta; cercus very small with 3–4 small setae; sternite 9 (Figs 37, 54) 0.4× longer than greatest width, posteromedian margin convex, broadly rounded. Gonocoxite stout, 1.9× longer than greatest width, with elongate, heavily sclerotized anteromedian process; gonostylus



Figs 33–43. *Dasyhelea paulistana* Forattini & Rabello, 1957, adult. 33–37 – male; 38–43 – female. 33, 38 – flagellum; 34, 41 – palpus; 36, 42 – wing; 35 – thorax and legs; 37 – genitalia; 39 – frontal sclerite; 40 – clypeus; 43 – abdominal segments 8–10.



Figs 44–52. *Dasyhelea paulistana* Forattini & Rabello, 1957, pupa. 44–48, 51 – female pupa; 49–50, 52 – male pupa. 44 – entire pupa; 45 – dorsal apotome; 46 – cephalothoracic chaetotaxy (dorsal view); 47 – dorsal sensilla and supraalar; 48 – respiratory organ; 49 – clypeal/labral sensilla and ocular sensilla; 50 – abdominal segment 1 chaetotaxy; 51–52 – segment 9. Abbreviations: AL-1-T, AL-2-T, AL-3-T – anterolateral sensilla; AM-1-T, AM-2-T – anteromedial sensilla; CL-1-H, CL-2-H – clypeal/labral sensilla; DA – dorsal apotome; DA-1-H, DA-2-H – dorsal apotomal sensilla; DL-1-H, DL-2-H – dorsolateral cephalic sclerite sensilla; D-3-T, D-5-T – dorsal sensilla; D-2-I, D-4-I, D-7-I, D-8-I, L-1-I, L-2-I, L-3-I – abdominal segment 1 chaetotaxy; GL – genital lobe; O-1-H, O-2-H – ocular sensilla; P – pedicel; RO – respiratory organ; SA-2-T – supraalar; TP – terminal process.



Figs 53–61. *Dasyhelea paulistana* Forattini & Rabello, 1957. 53–56 – adult male; 57–61 – female pupa. 53 – genitalia; 54 – sternite 9; 55 – paramere and gonocoxal apodemes; 56 – aedeagus; 57 – cephalothoracic chaetotaxy, dorsal view; 58 – dorsal sensilla and supraalar; 59 – clypeal/labral sensilla; 60 – abdominal segment 1 chaetotaxy; 61 – segment 4 chaetotaxy. Scale bars = 0.05 mm. Abbreviations: **AL-1-T**, **AL-2-T**, **AL-3-T** – anterolateral sensilla; **AM-1-T**, **AM-2-T** – anteromedial sensilla; **CL-1-H**, **CL-2-H** – clypeal/labral sensilla; **D-3-T**, **D-5-T** – dorsal sensilla; **DL-1-H**, **DL-2-H** – dorsolateral cephalic sclerite sensilla; **D-2-I**, **D-4-I**, **D-7-I**, **D-8-I**, **L-1-I**, **L-2-I**, **L-3-I** – abdominal segment 1 chaetotaxy; **D-2-IV**, **D-4-IV**, **D-7-IV**, **D-8-IV**, **L-1-IV**, **L-2-IV**, **L-3-IV**, **L-4-IV**, **V-5-IV**, **V-6-IV** – segment 4 chaetotaxy; **GL** – genital lobe; **O-1-H**, **O-2-H** – ocular sensilla; **SA-2-T** – supraalar.

0.8× longer than gonocoxite, narrow at base, nearly straight, slightly curved distally, with spur-like tip. Paramere and gonocoxal apodemes forming an asymmetrical structure (Figs 37, 55); gonocoxal apodemes stout, one narrowly connected with paramere, other separate; paramere elongate, sinuate, with hooked tip. Aedeagus (Figs 37, 56) 0.8× longer than greatest width; with slender, heavily sclerotized, V-shaped basal arch, from which arises pair of stout posterior, rounded expansions, each with heavily sclerotized inner margin, these margins pointed subapically and joined anteromesally, forming a slender U-shaped arch, basal arch extending to 0.3 of total length.

Female (Figs 38–43). Head dark brown. Eyes contiguous by width of 1–2 ommatidia. Antenna with flagellum (Fig. 38) pale brown; AR 0.96–1.03 (1.00, n = 6). Frontal sclerite subspherical with posteromesal slender projection (Fig. 39). Clypeus (Fig. 40) with 3 pairs of setae. Palpus (Fig. 41) pale brown; segment 3 with 6–7 subbasal capitate sensilla on anterior inner margin, PR 1.70–2.00 (1.78, n = 8).

Thorax. Scutum dark brown, scutellum pale with 6 strong setae. Legs pale brown; femora with mesal broad slightly infuscated band, knees dark, apex of hind tibia with 6 spines; foreleg TR 2.17–2.79 (2.35, n = 9), midleg TR 2.20–2.59 (2.41, n = 8), hind leg TR 2.16–2.50 (2.38, n = 8). Wing (Fig. 42), length 0.75–0.93 (0.85, n = 8) mm, width 0.33–0.45 (0.37, n = 8) mm, CR 0.39–0.46 (0.42, n = 8); membrane hyaline, densely covered macrotrichia; cubital fork at same level of the second radial cell. Halter pale brown.

Abdomen. Dark brown. Subgenital plate ovoid, as in Fig. 43; posterolateral arms slender, angulate. Spermatheca rounded (Fig. 43) strongly pigmented, diameter 48 µm, neck oblique, stout, measuring 12 µm.

Female pupa (Figs 44–48, 51). Total length 2.12–2.46 (2.32, n = 6) mm. General coloration of exuvia pale brown. Dorsal apotome (Fig. 45) 2.11× broader than long, surface smooth with wrinkles on its margins, anterior margin concave, posterior margin rounded with subapical rounded tubercle; dorsal apotome sensilla (Fig. 45), one short, thin seta, other campaniform sensillum; DAL 0.096–0.120 (0.10, n = 6) mm; DAW 0.216–0.240 (0.228, n = 6) mm; OW/OL 2.00–2.50 (2.29, n = 6). Cephalothorax surface smooth, length 0.780–0.960 (0.860, n = 8) mm, width 0.570–0.650 (0.60, n = 9) mm. Cephalothoracic sensilla as follows: two dorsolateral cephalic sclerite sensilla (Figs 46, 57), one short seta, one campaniform sensillum; three anterolateral sensilla (Figs 46, 57), one medium-sized, thin, hyaline seta, one short seta, one peg; two anteromedial sensilla (Figs 46, 57), one long, thin seta, other short seta; dorsals (Figs 47, 58): D-1-T, D-2-T, D-4-T absent, D-3-T short, thin seta, D-5-T campaniform sensillum, supraalar (SA-2-T) campaniform sensillum. Respiratory organ (Figs 44, 48) pale brown, thick at base, distal half tapering to a sharp apex, 3.7× longer than broad, surface smooth, with 21–22 apical pores; RO 0.132–0.160 (0.152, n = 8) mm, RO width 0.036–0.048 (0.038, n = 8) mm; pedicel (Fig. 48) yellowish, smooth, pedicel length 0.020–0.024 (0.023, n = 9) mm, P/RO 0.090–0.166 (0.148, n = 8). Two clypeal/labral sensilla, CL-1-H one long, thin seta, CL-2-H short seta (Fig. 59); two ocular sensilla, O-1-H medium-sized, thin seta, O-2-H campaniform sensillum (Fig. 59). Abdominal segments covered with very small spinules. Abdominal segment 1 (Fig. 60) with sensilla as follows: D-2-I peg; D-4-I, D-7-I, D-8-I campaniform sensilla; 3 lateral sensilla, L-1-I medium-sized seta, L-2-I, L-3-I short, thin setae. Second abdominal segment similar to first. Segment 4 with sensilla pattern (Fig.

61) as follows: D-2-IV peg; D-4-IV, D7-IV campaniform sensilla, D-8-IV medium-sized, thin seta, all located on flattened tubercles; L-1-IV, L-3-IV medium-sized, thin setae, L-2-IV, L-4-IV campaniform sensilla, all located on triangular tubercles; two ventral sensilla, V-5-IV medium-sized, thin seta, V-6-IV short, thin seta, both located on flattened tubercles. Segment 9 (Figs 44, 51) $2.75\times$ longer than width, ventral surface with many spinules; length 0.40–0.43 (0.41, $n = 9$) mm, width 0.14–0.19 (0.16, $n = 9$) mm. Terminal process (Fig. 51) straight, $2\times$ longer than length of body of segment 9, tip pointed, base wide with two setae, one long, thin, hyaline seta on small rounded base, other medium-sized, stout seta on rounded tubercle; length 0.23–0.28 (0.25, $n = 9$) mm.

Male pupa (Figs 49–50, 52). Similar to female with sexual differences: Total length 2.05–2.61 (2.41, $n = 4$) mm. Exuviae pale brown. Dorsal apotome with DAL 0.11–0.13 (0.12, $n = 6$) mm; DAW 0.24–0.27 (0.25, $n = 6$) mm, OW/OL 1.82–2.20 (2.02, $n = 6$). Respiratory organ, RO length 0.16–0.19 (0.17, $n = 7$) mm, RO width 0.04 ($n = 7$) mm; pedicel length 0.024 ($n = 8$) mm, P/RO 0.12–0.15 (0.13, $n = 7$). Cephalothorax length 0.93–1.23 (1.02, $n = 7$) mm, width 0.66–0.75 (0.68, $n = 7$) mm. Clypeal/labral sensilla and oculars as in Fig. 49. Segment 1 as in Fig. 50. Segment 9 (Fig. 52) length 0.41–0.47 (0.44, $n = 8$) mm, width 0.14–0.22 (0.20, $n = 8$) mm; terminal process (Fig. 51) length 0.23–0.30 (0.27, $n = 8$) mm.

Distribution. Brazil (São Paulo, Amazonas), Argentina (Misiones, Corrientes).

Discussion. *Dasyhelea paulistana* perfectly fits in the *grisea* species-group as it was defined by WAUGH & WIRTH (1976), and is very similar to the Nearctic species *D. chani* Wirth & Linley, 1990. However, the latter species can be distinguished from *D. paulistana* by the male with sternite 9 exhibiting a concave posteromedian projection, paramere with a subapical tuft of setae and the basal arch of aedeagus with caudomedian expansions from which arises a slender, bifid, wing-like plate. The female differs in the rectangular subgenital plate with anterior margin concave and in the spermatheca with oblique neck. Finally, the pupal respiratory organ is amber in colour, with a hyaline tip and bearing 16 pores.

Bionomics

The pupa of *D. pseudopollinosa* sp. nov. was collected associated with aquatic ferns together with *Dasyhelea paulistana*, *Stilobezzia* (*Stilobezzia*) *punctulata* Lane, 1947, *Stilobezzia* (*Stilobezzia*) *rabelloi* Lane, 1947, and *Stilobezzia* (*Eukraiohelea*) *elegantula* (Johannsen, 1907) from Marchantaria Island. *Dasyhelea paulistana* was also collected in Argentina in similar environments, coexisting with *Allaudomyia schnacki* Spinelli & Wirth, 1983, *Stilobezzia* (*Stilobezzia*) *punctulata* and *Bezzia pulchripes* Kieffer, 1917.

When placed in cotton-stopper vials individually, *D. pseudopollinosa* completes its development in three days at a lab temperature of 9.7°C – 29°C , and *D. paulistana* in seven days at a lab temperature of 16.1°C – 21.5°C (19.5°C , $n = 7$) and humidity of 69–89 % (78 %, $n = 7$).

As was pointed out by WIRTH & LINLEY (1990), BORKENT & CRAIG (2001) and CAZORLA & MARINO (2004), the pointed respiratory organ of the two species herein described, as well as the ones of the Nearctic species *D. chani*, *D. pollinosa* and *D. traverae* are likely adapted to obtain air from the submerged roots of aquatic plants.

Acknowledgments

We are grateful to PIP 0381, CONICET and to INPA, MCTI PRJ 12.272 for the support during the field trip. Our gratitude to Nélide Caligaris for technical assistance. We also acknowledge one anonymous reviewer and Dr. Art Borkent acting as journal reviewers for their appropriate suggestions on the manuscript.

References

- BORKENT A. 2012: The pupae of the families of Culicomorpha—Morphology and a new phylogenetic tree. *Zootaxa* **3396**: 1–98.
- BORKENT A. 2014: World species of biting midges (Diptera: Ceratopogonidae). 236 pp. URL: <http://www.inhs.uiuc.edu/research/FLYTREE/CeratopogonidaeCatalog.pdf>. Last update Jan. 20, 2014.
- BORKENT A. & CRAIG D. A. 2001: Submerged *Stilobezzia rabelloi* Lane (Diptera: Ceratopogonidae) pupae obtain oxygen from the aquatic fern *Salvinia minima* Baker. *Proceedings of the Entomological Society of Washington* **103**: 655.
- BORKENT A. & SPINELLI G. R. 2000: Catalog of the New World biting midges south of the United States of America (Diptera: Ceratopogonidae). *Contributions on Entomology International* **4(1)**: 1–107.
- BORKENT A. & SPINELLI G. R. 2007: Neotropical Ceratopogonidae (Diptera: Insecta). Pp. 1–198. In: ADIS J., ARIAS J. R., RUEDA-DELGADO G. & WANTZEN K. M. (eds.): *Aquatic Biodiversity in Latin America. Vol. 4*. Pensoft, Sofia-Moscow, 198 pp.
- BORKENT A. & WIRTH W. W. 1997: World species of biting midges (Diptera: Ceratopogonidae). *Bulletin of the American Museum of Natural History* **233**: 1–257.
- BROWN B. V., BORKENT A., CUMMING J. M., WOOD D. M., WOODLEY N. E. & ZUMBADO M. A. 2009: *Manual of Central American Diptera. Volume 1*. NRC Research Press, Ottawa, Ontario, Canada, 744 pp.
- CAZORLA C. G. & MARINO P. I. 2004: The pupa of *Stilobezzia punctulata* Lane from Peruvian Amazonia (Diptera: Ceratopogonidae). *Amazoniana* **17(1–2)**: 75–80.
- DÍAZ F., SPINELLI G. R. & RONDEROS M. M. 2009: Biting midges of the *Dasyhelea cincta* group from Patagonia (Diptera: Ceratopogonidae). *Deutsche Entomologische Zeitschrift* **56**: 149–156.
- DÍAZ F., SPINELLI G. R. & RONDEROS M. M. 2010: A revision of the species allied to *Dasyhelea patagonica* Ingram & Macfie (Diptera: Ceratopogonidae). *Journal of Natural History* **44**: 2825–2849.
- FORATTINI O. P. & RABELLO E. X. 1957: Dados sobre *Dasyhelea* Kieffer, 1911 do Brasil (Diptera: Ceratopogonidae). *Revista Brasileira de Entomologia* **7**: 241–250.
- GROGAN W. L. & WIENERS J. A. 2006: A new species of the biting midges genus *Dasyhelea* Kieffer (Diptera: Ceratopogonidae) from the Bahamas. *Proceedings of the Entomological Society of Washington* **108**: 467–473.
- WAUGH W. T. & WIRTH W. W. 1976: A revision of the genus *Dasyhelea* Kieffer of the eastern United States north of Florida (Diptera: Ceratopogonidae). *Annals of the Entomological Society of America* **69**: 219–247.
- WIRTH W. W. 1974: Family Ceratopogonidae. In: *A catalog of the Diptera of the Americas South of the United States. Fasc. 14*. Museu de Zoologia, Universidade de São Paulo, 89 pp.
- WIRTH W. W. & LINLEY J. R. 1990: Description of *Dasyhelea chani* new species (Diptera: Ceratopogonidae) from leaves of the water lettuce (*Pistia stratiotes*) in Florida. *Florida Entomologist* **73**: 273–279.