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ARTICLE

The puparium of *Cyphomyia wiedemanni* Gerstaecker, 1857 (Diptera: Stratiomyidae) found in cassava (*Manihot esculenta*) roots

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Abstract. The puparium of *Cyphomyia wiedemanni* Gerstaecker, 1857 is hereby described for the first time, based on 14 puparia from specimens collected buried in soil and feeding off the roots of cassava (*Manihot esculenta* Crantz) plants in an area of industrial cassava plantation in Distrito Federal (15°35'30"S, 47°42'30"W), Brazil and, reared in controlled lab conditions until the emergence of adults. Unprecedented data on the occurrence of *Cyphomyia wiedemanni* in Distrito Federal (new record), and the relationship of the larvae with the cassava plants are discussed.

Keywords. Clitellariinae; Cerrado vegetation; Immature forms; Soldier fly; Taxonomy.

INTRODUCTION

The genus *Cyphomyia* Wiedemann, 1819 is composed of 86 species, 73 of which are neotropical, being the second largest neotropical genus in Stratiomyidae, with 32 species registered for Brazil (Woodley, 2001, 2011, 2014; Hauser *et al.,* 2017). The larvae of *Cyphomyia* develop under the bark of rotting logs and in damaged plant material where they feed on microorganisms associated with the decomposition process of fluidified plant matter (Iide, 1963; McFadden, 1967; Woodley, 2001; Xerez *et al.,* 2004; Torres-Toro & Wolff, 2019). Larvae may be found in different kinds of rotting plant material, such as damaged or rotting cacti, rotting papaya tree trunks, viscous material from Agave, fluidified resin and in rotting tree logs (Table 1) (James, 1957; Iide, 1963, 1967; McFadden, 1967; McFadden & James, 1969; Xerez *et al.,* 2004; Torres-Toro & Wolff, 2019).

Even considering the large number of species of *Cyphomyia,* the history of studies over the past decades, few recent studies have focused on the species of this genus (James, 1940, 1957; Iide, 1963, 1967; McFadden & James, 1969; Torres-Toro & Wolff, 2019). Of the 86 species of *Cyphomyia,* only nine have their larvae or puparium known and described, viz. *C. albitarsis* Fabricius, 1805 (Torres-Toro & Wolff, 2019: 557), *C. aurifrons* Wiedemann, 1830 (Xerez *et al.,* 2004: 80), *C. bicarinata* Williston, 1900 (McFadden, 1967: 28), *C. erecta* McFadden, 1969 (McFadden & James, 1969: 315), *C. leucocephala* Wiedemann, 1819 (Iide, 1963: 25), *C. marginata* Loew, 1866 (McFadden, 1967: 27), *C. picta* Schiner, 1868 (Xerez *et al.,* 2004: 81), *C. pilosissima* Gerstaecker, 1857 (James, 1957: 639) e *C. souzalopesi* Iide, 1967 (Iide, 1967: 226) (Table 1).

Cyphomyia wiedemanni Gerstaecker, 1857 is widely distributed in the Neotropical region (Fig. 1) (Woodley, 2001; Fachin & Assis-Pujol, 2016). Adults can be found on flowers, but are much more often found sitting on sunny leaves. Like most *Cyphomyia* species, they display a strong sexual dimorphism, with dichoptic females having, generally, a yellow head, and the holoptic males have a dark head (Fig. 2). Here the puparium of *C. wiedemanni* is described based on 14 puparia and represents the tenth species of the genus with known immature forms (larvae and pupae), and the first associated with cassava roots (Table 1).

MATERIAL AND METHODS

Seventy one immature specimens of *C. wiedemanni* were found and collected in cas-

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Table 1. Species of *Cyphomyia* Wiedemann: instars and development time, natural substrate breeding and geographical distribution.

SPECIES	INSTAR/DEVELOPMENT	BREEDING SUBSTRATE	DISTRIBUTION[6,7,8,10]
C. albitarsis[11]	Puparia/34 days	Decomposing papaya tree trunks (*Carica papaya* L.)	Bolivia, **Brazil**, Colombia, Costa Rica, Ecuador, Guyana, Honduras, Mexico, Panama, Paraguay, Peru, Trinidad, Venezuela
C. aurifrons[9]	Puparia/44-94 days	Unidentified decaying tree trunks	Argentina, **Brazil**, Ecuador
C. bicarinata[4,5]	Larvae and Puparia/Not described	Damaged or decomposing cacti (*Opuntia* sp.); Decomposing papaya tree trunks (*Carica papaya* L.)	Mexico
C. erecta[5]	Larvae and Puparia/Not described	Damaged or decomposing cacti (*Platyopuntia* sp.)	Mexico, USA
C. leucocephala[2]	Larvae/Not described	Decomposing papaya tree trunks (*Carica papaya* L.)	Argentina, **Brazil**, Ecuador, Paraguay, Peru
C. marginata[4]	Puparia/Not described	Decomposing papaya tree trunks (*Carica papaya* L.)	Costa Rica, Cuba, Dominican Republic, Haiti, Jamaica, Puerto Rico, USA.
C. picta[9]	Puparia/69 days	Unidentified decaying tree trunks	**Brazil**, Costa Rica, Ecuador, Panama
C. pilosissima[1,4]	Larvae and Puparia/54-84 days	Slimy material from Agave (*Agave* sp.)	Mexico
C. souzalopesi[3]	Larvae and Puparia/41 days	Fluidized resin	**Brazil**
C. wiedemanni	Larvae and Puparia/85-123 days	Cassava roots (*Manihot esculenta* Crantz)	Argentina, Bolivia, **Brazil**, Colombia, Costa Rica, Ecuador, French Guiana, Guyana, Mexico, Nicaragua, Panama, Paraguay, Peru, Suriname, Trinidad and Tobago, Venezuela

References: James, 1957[1]; Iide, 1963[2], 1967[3]; McFadden, 1967[4]; McFadden & James, 1969[5]; Woodley, 2001[6], 2011[7], 2014[8]; Xerez *et al.,* 2004[9]; Hauser *et al.,* 2017[10]; Torres-Toro & Wolff, 2019[11].

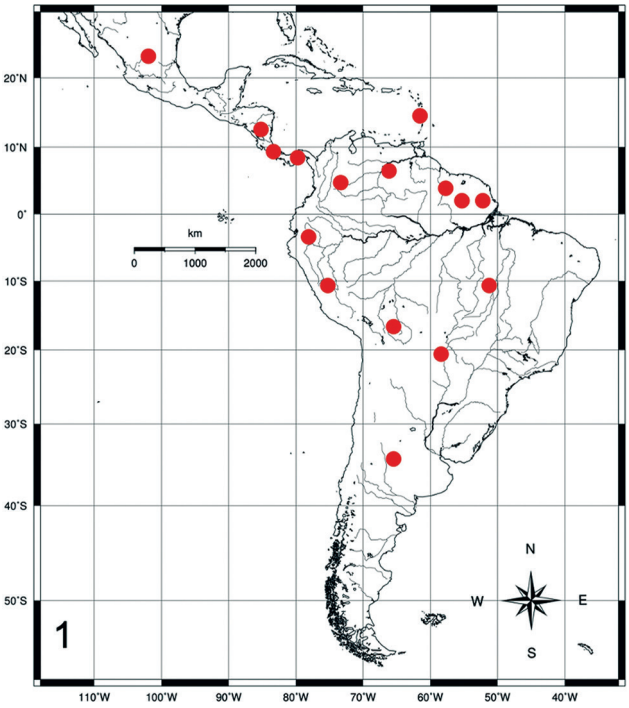

Figure 1. *Cyphomyia wiedemanni* Gerstaecker. Range of geographical distribution in the Neotropical Region.

sava roots (*Manihot esculenta* Crantz) at an industrial cassava crop of the Empresa Brasileira de Pesquisa Agropecuária (Embrapa) (15°35'30"S, 47°42'30"W). The larvae were reared in semi-controlled conditions (relative humidity 75%-80% and the temperature 22°C-24°C) in Petri dishes with cassava mass for feeding. After pupation, the specimens were transferred to clean Petri dishes until the emergence of the adults. The adults were pinned, and the puparia fixed in microtubes containing a (1:1) solution of 70% alcohol and glycerol. In total, 14 adults of *Cyphomyia wiedemanni* (6♂ and 8♀) emerged. All specimens are housed in the Coleção Entomológica do Departamento de Zoologia da Universidade de Brasília (DZUB).

Photographs of the puparia and adults were obtained with the Leica M205C© stereomicroscope equipped with the Leica DFC295© digital camera. Measurements, in millimeters (mm), were made with the Leica LAS-V3-8© software. Micrographs of the puparia were obtained with Scanning Electron Microscopy (SEM). The samples were fixed in osmium tetroxide for one hour. They were first washed in distilled water, and afterwards dehydrated in different concentrations of acetone (30%, 50%, 70%, 90% e 100%), for 10 minutes in each concentration. Then, the samples were critical point dried with CO_2, assembled on stubs, sputter coated with gold and examined in a scanning electron microscope JEOL JSM 7001F©.

The terminology adopted for this study follows Rozkošný (1982). The following abbreviations were used: (a) antenna; (Ad) anterodorsal seta; (Ap) apical seta; (as) anterior spiracle; (asl) anal slit; (Cf) clypeofrontal seta; (D) dorsal seta; (Dl) dorsolateral seta; (ep) eye prominence; (L) lateral seta; (Lb) labral seta; (lb) labrum; (mm) mandibular-maxillary complex; (mo) molar area (inner surface of the mandible); (mxp) maxillary palpus; (ph) pupal respiratory horn; (po) pore; (pso) posterior spiracular opening; (Sa) subapical seta; (Sl) sublabral seta; (sl) sensilla; (spa) sternal patch; (V) ventral seta; (prm) prementum; (Vl) ventrolateral seta.

The acronyms of the collections cited in the text are given below:

DZUB – Coleção Entomológica do Departamento de Zoologia da Universidade de Brasília, Brasília, Brazil;
NHMUK – The Natural History Museum, London, United Kingdom;
NMW – Naturhistorisches Museum Wien, Vienna, Austria;
SMF – Forschungsinstitut Naturmuseum Senckenberg, Frankfurt/Main, Germany;
ZMHB – Museum für Naturkunde, Berlin, Germany;
ZIL – Museum of Zoology, Lund, Sweden.

RESULTS

Cyphomyia wiedemanni Gerstaecker, 1857 (Figs. 3-7)

Cyphomyia flavispinis Macquart, 1855: 60. ST 1♂, 1♀ [NHMUK]. "Amérique méridionale". Syn. by Gerstaecker (1857: 302).

Cyphomyia wiedemanni Gerstaecker, 1857: 302. ST 1♂, 1♀ [ZMHB]: Brazil, Pará; ST 1♂, 1♀ [ZMHB]: French Guiana, Cayenne; ST ? [ZMHB]: Surinam; ST ♀ [ZMHB]: Venezuela; ST 1♂, 1♀ [ZIL]: Brazil; ST ♂♀ [NMW]: Brazil, "south Brazil".

Cyphomyia albispinis Costa, 1866: 39. *Nomen nudum*. Likely referring to *Cyphomyia flavispinis* Macquart, 1855.

Cyphomyia cyanispis Bigot, 1874: 86. *Nomen nudum*.

Cyphomyia cyanispis Bigot, 1876: 487. HT ♂ [NHMUK]: Brazil, "Amazon".

Cyphomyia wiedemanni ssp. *peruana* Lindner, 1933: 329. HT ♀ [SMF]: Peru, Iquitos, Roqué.

Cyphomyia wiedemanni ssp. *burmeisteri* Lindner, 1949a: 786. *Nomen nudum*.

Cyphomyia wiedemanni ssp. *burmeisteri* Lindner, 1949b: 863. ST 2♂ [NHMUK]: Brazil, "Amazon".

Cyphomyia viedemanni James, 1953: 311. Incorrect subsequent spelling.

Cyphomyia cyanispis. Woodley, 2001: 163. Incorrect subsequent spelling.

Geographical Distribution: Argentina, Bolivia, Brazil (Acre*, Alagoas*, Amazonas, Bahia*, Distrito Federal*, Espírito Santo*, Goiás, Mato grosso, Mato Grosso do Sul, Minas Gerais, Pará, Paraná*, Pernambuco*, Rio Grande do Sul*, Roraima and São Paulo*), Colombia, Costa Rica, Ecuador, Guiana, French Guiana, Mexico, Nicaragua, Panama, Paraguay, Peru, Surinam, Trinidad & Tobago, Venezuela. **[*New record]**

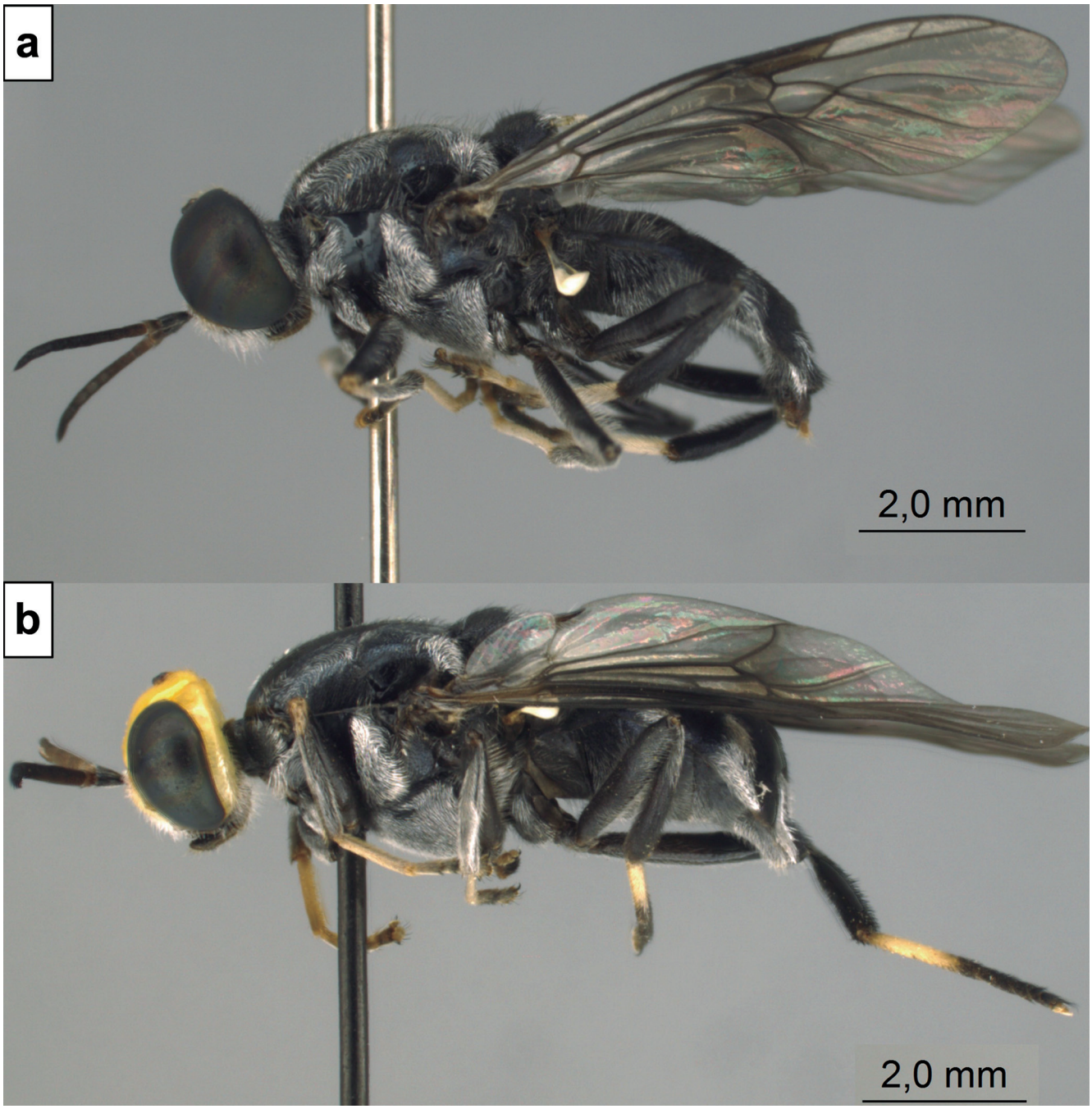

Figure 2. *Cyphomyia wiedemanni* Gerstaecker. Habitus, lateral view. (a) Male; (b) Female.

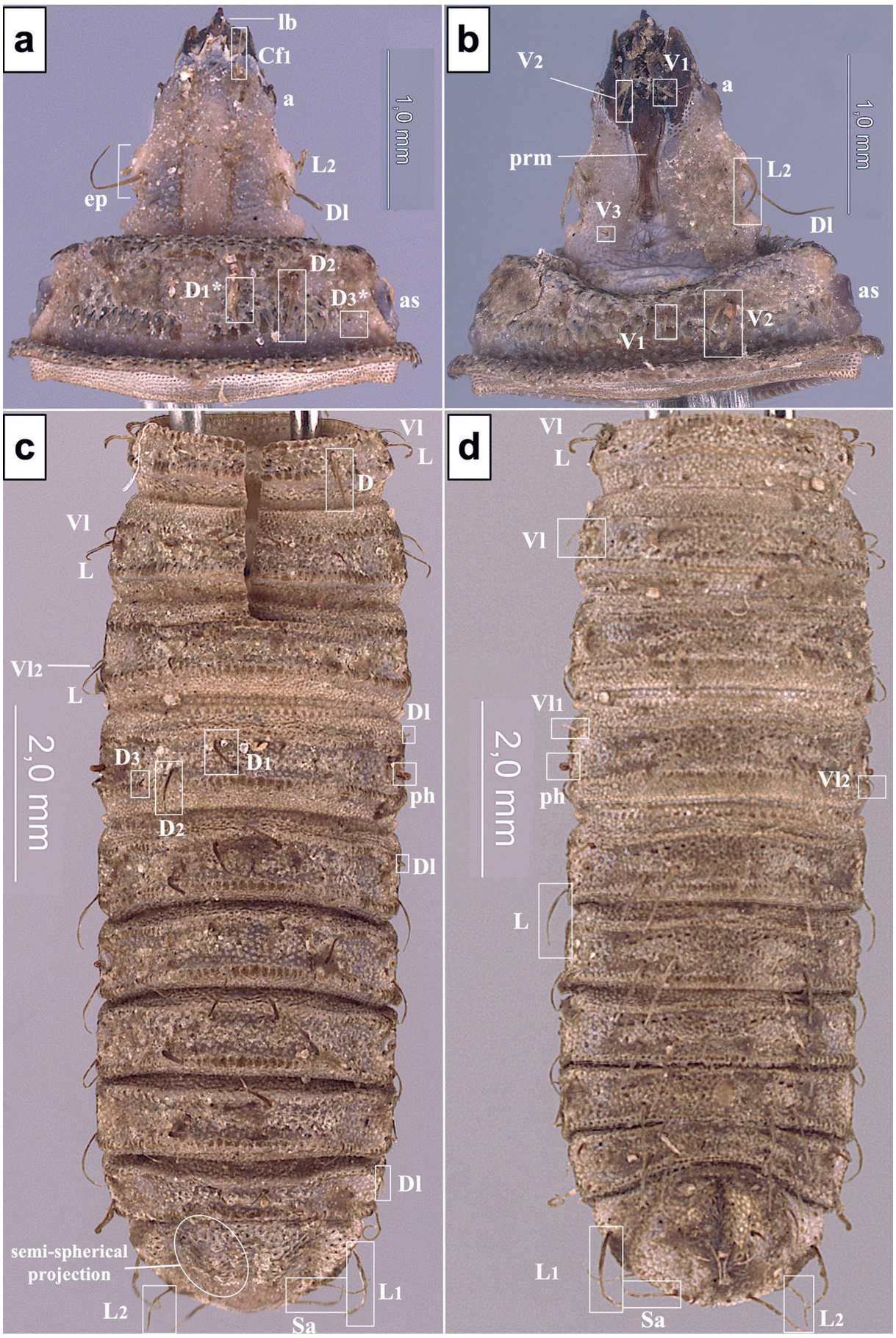

Figure 3. *Cyphomyia wiedemanni* Gerstaecker. Puparium. (a) Head and thoracic segment 1, dorsal view; (b) Head and thoracic segment 1, ventral view; (c) Thoracic segments 2-3 and abdominal segments 1-8, dorsal view; (d) Thoracic segments 2-3 and abdominal segments 1-8, ventral view. **[* Structure lost or not evident in the figure]**

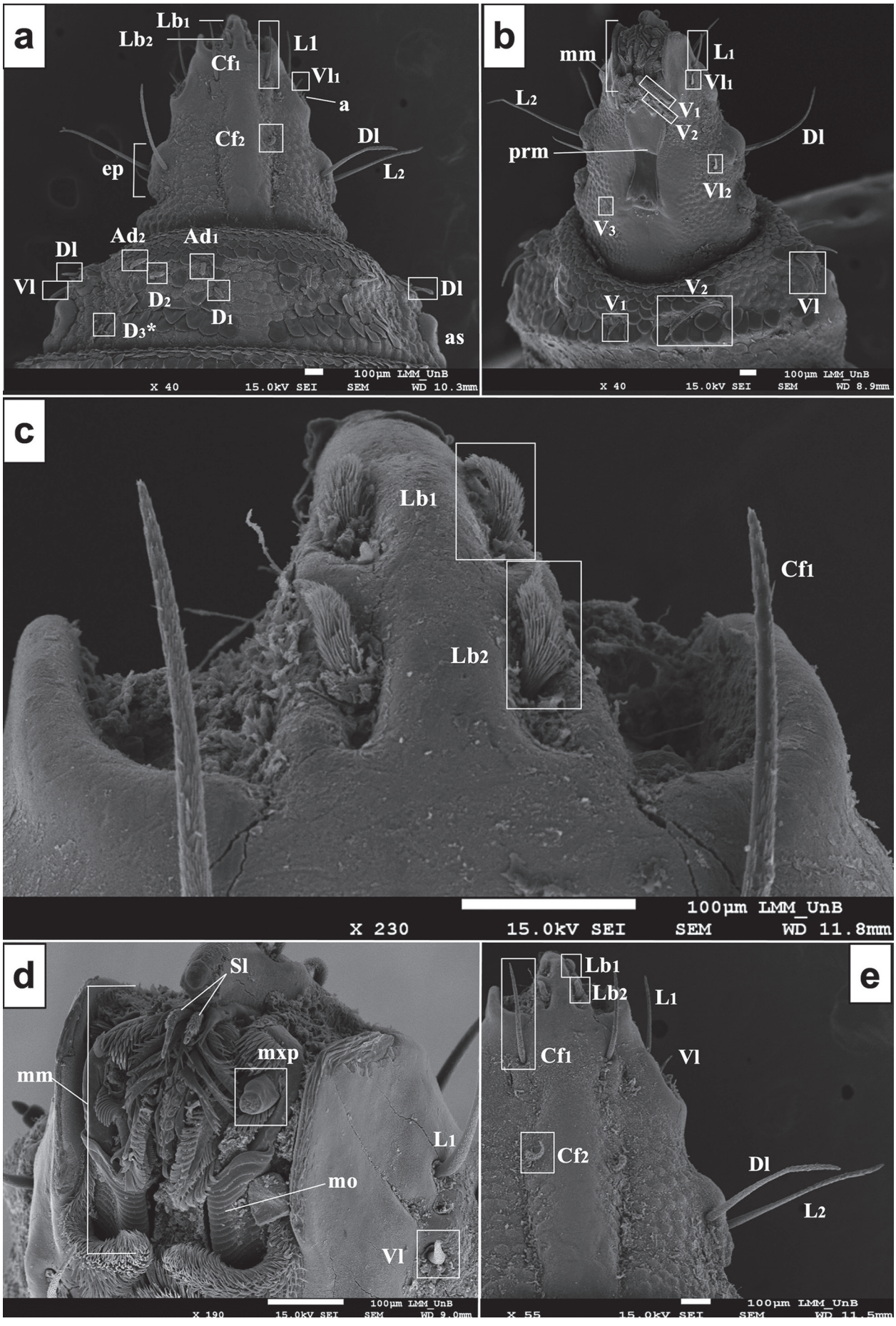

Figure 4. *Cyphomyia wiedemanni* Gerstaecker. Puparium. (a) Head and thoracic segment 1, dorsal view; (b) Head and thoracic segment 1, ventral view; (c) Labrum and labral setae, dorsal view; (d) mandibular-maxillary complex, ventral view; (e) Head, dorsal view. **[* Structure lost or not evident in the figure]**

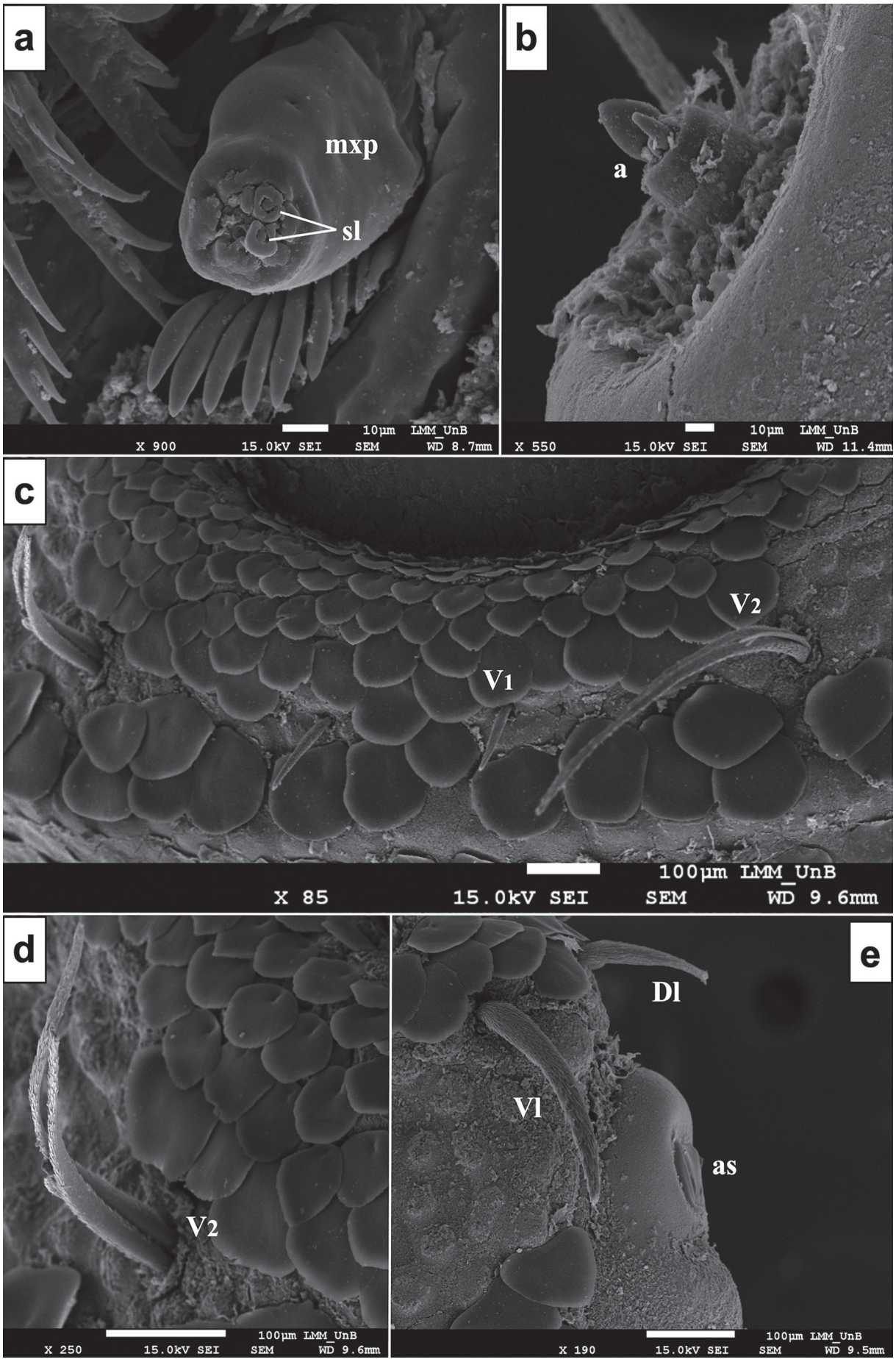

Figure 5. *Cyphomyia wiedemanni* Gerstaecker. Puparium. (a) Maxillary palp and sensilla, ventral view; (b) Antenna, dorsal view; (c) Thoracic segment 1, ventral setae, ventral view; (d) Thoracic segment 1, ventral seta 2, ventral view; (e) Anterior spiracle and dorsolateral and ventrolateral setae, ventral view.

Description of the puparium

Diagnosis: The puparium of *Cyphomyia wiedemanni* distinguishes itself from other *Cyphomyia* species for exhibiting: two pairs of minute labral setae (Lb_1, Lb_2) with a barbed appearance, similar to brushes; one sole ocular tubercle fused to the eye prominence (ep), with a reduced ridge between the eyes and ocular tubercles; a pair of long sublabral setae (Sl) inserted below the apex of the labrum, with the basal ½ barbed and the distal ½ smooth; first thoracic segment with 2 pairs of anterodorsal setae (Ad_1, Ad_2) and 3 pairs of dorsal setae (D_1, D_2, D_3), with D_3 being minute and practically imperceptible; abdominal segments with the first pair of ventral setae (V_1) significantly smaller than the rest; sternal patch with ellipsoidal shape longitudinally elongated; abdominal segment 8 with a pair of semi-spherical projections on the dorsal surface, where the dorsal setae (D) are inserted; anal slit (asl) long, almost the same length as the 8th abdominal segment.

Puparium: Length varying from 8-14 mm, including head, three thoracic segments and eight abdominal seg-

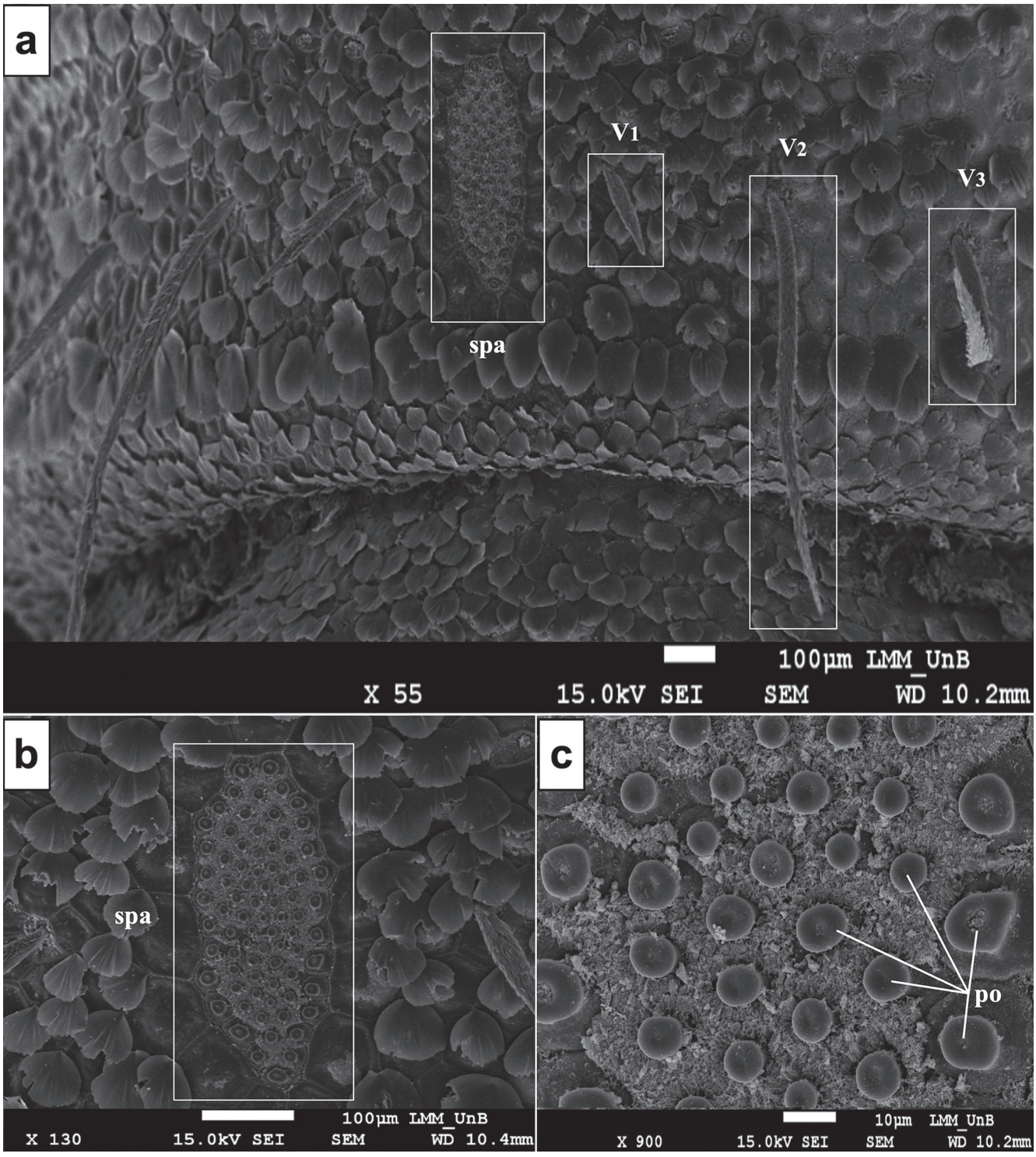

Figure 6. *Cyphomyia wiedemanni* Gerstaecker. Abdominal segment 6, ventral view. (a) Sternal patch and ventral setae; (b) Sternal patch detail; (c) Sternal patch pores.

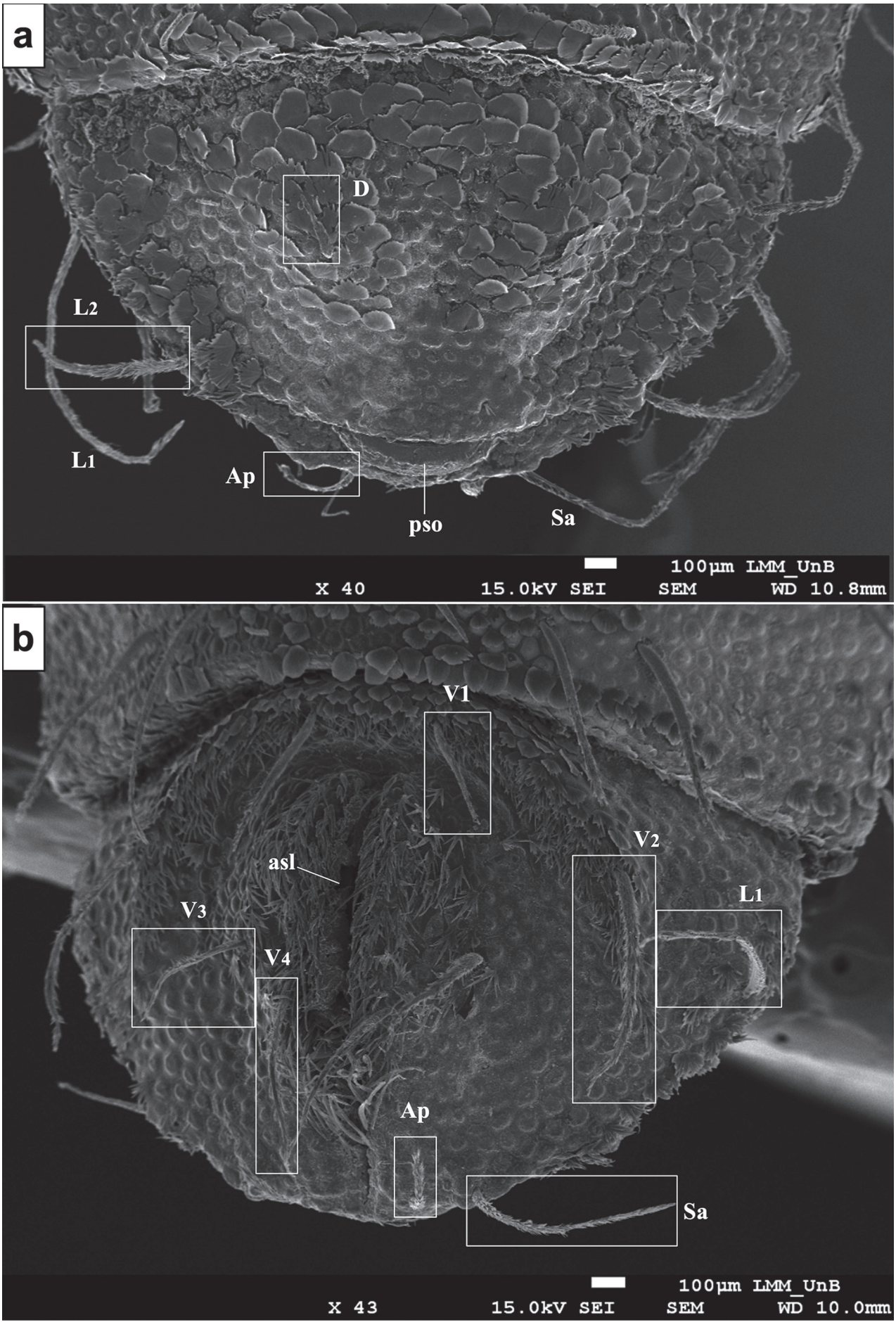

Figure 7. *Cyphomyia wiedemanni* Gerstaecker. Abdominal segment 8. (a) Dorsal view; (b) Ventral view.

ments (Fig. 3); cuticle brownish, with erect dorsal setae. **Head** (Figs. 3-4). Subtriangular, longer than wide and flattened dorsoventrally, without plates or scales; labrum (lb) triangular; antennae (a) short emerging dorsolaterally on the anterior portion of the head; dorsal surface smooth with median line extending from the base of the head up to the labrum (lb); eye prominence (ep) with ocular tubercles positioned on the posterior ½ of the head with inconspicuous eyes. Ventral surface smooth, without plates or scales, with prementum (prm) evident; mandibular-maxillary complex (mm) well developed and housed in clefts; maxillary palpus (mxp) inserted in the anterior portion of the molar area (mo). Chaetotaxy of the dorsal surface: 2 pairs of minute labral setae (Lb_1, Lb_2) with a barbed aspect, similar to brushes, positioned on the anterior extremity of the clipeus; 2 pairs of clipeofrontal setae (Cf_1, Cf_2), the anterior pair (Cf_1) significantly longer than the posterior (Cf_2); 1 pair of dorsolateral seta (Dl) above the eyes; 2 pairs of lateral setae (L_1, L_2), the first (L_1) inserted shortly after the labrum and before the antennae, and the second (L_2) inserted in the space between the eyes and ocular tubercles. Chaetotaxy of the ventral surface: 1 pair of long sublabral setae (Sl) below the apex of the labrum, with the basal ½ barbed and the distal ½ smooth; 3 pairs of short ventral setae (V_1, V_2, V_3) located right below the mandibular clefts; 2 pairs of ventrolateral setae (Vl_1, Vl_2), with the first pair (Vl_1) inserted shortly before the alignment of the antenna, and the second pair (Vl_2) aligned with the eye prominence (ep). **Thorax** (Figs. 3, 5). Coated by plates or scales with elliptical shape. Prothorax subretangular in shape, with prominent anterior spiracles (as); mesothorax and metathorax with lateral edges curved; T-shaped suture visible on the metathorax and mesothorax. Chaetotaxy of the dorsal surface: prothorax with 2 pairs of anterodorsal setae (Ad_1, Ad_2) and 3 pairs of dorsal setae (D_1, D_2, D_3), with minute D_1 and D_3, D_3 being shorter than D_1 and, in general, harder to visualize; 1 pair of dorsolateral setae (Dl) inserted before the anterior spiracle (as); meso and metathorax with 3 pairs of dorsal setae (D_1, D_2, D_3), the second pair (D_2) longer than the rest; 1 pair of lateral setae (L) on the anterior portion of each segment. Chaetotaxy of the ventral surface: prothorax, mesothorax and metathorax similar, with 1 pair of ventrolateral setae (Vl) and 2 pairs of ventral setae (V_1, V_2), with V_1 short and V_2 bipartite, long and with a short accessory seta (V_2). **Abdomen** (Figs. 3, 6-7). Segments 1-7 with similar shape; 1 pair of dorsolateral pupal respiratory horns (ph) in each of the segments 2-5; sternal patch (spa) on the ventromedian portion of segment 6, with longitudinally elongated elliptical shape, composed by cylindrical cells with distinct pores (po) (Fig. 6); segment 8 with semicircular shape; anal slit (asl) surrounded by two perianal grooves. Chaetotaxy of segments 1-7 in dorsal view: 3 pairs of dorsal setae (D_1, D_2, D_3), D_1 being the longest and D_1, D_3 the shortest; a pair of lateral setae (L) near the end of each segment; 1 pair of short dorsolateral setae (Dl) near the start of each segment. Chaetotaxy of segments 1-7 in ventral view: 2 pairs of ventrolateral setae (Vl_1, Vl_2) with the first pair (Vl_1) inserted at the top of each segment and being slightly longer than the second (Vl_2) inserted near the end of each segment; 3 pairs of ventral setae (V_1, V_2, V_3), V_1 significantly shorter than the rest. Chaetotaxy of segment 8 in dorsal view: 1 pair of dorsal setae (D) emerging from the top of semispherical projections; 2 pairs of long barbed lateral setae (L_1, L_2). Chaetotaxy of segment 8 in ventral view: 4 pairs of ventral setae (V_1, V_2, V_3, V_4); 1 pair of subapical setae (Sa) and 1 pair of apical setae (Ap).

Material examined. Planaltina, [Brasília,] DF [Distrito Federal], Brasil, 05.ii.2020 (EMBRAPA [CERRADOS]), Pujol/Mousinho/Morgado [cols.], 15°35′30″S, 47°42′30″W, Raiz de mandioca [cassava root], [emergence] 03.iv.2020 (1♂, UNB-238098); [emergence] 09.iv.2020 (1♂, UNB-238113); [emergence] 13.iv.2020 (2♀, UNB-237985, UNB-238077); [emergence] 14.iv.2020 (2♂, UNB-237975, UNB-238049); [emergence] 20.iv.2020 (1♂, UNB-238023); [emergence] 24.iv.2020 (1♀, UNB-238066); [emergence] 25.iv.2020 (1♀, UNB-238092); [emergence] 27.iv.2020 (1♂, UNB-238075); [emergence] 28.iv.2020 (1♀, UNB-238041); [emergence] 30.iv.2020 (1♀, UNB-238005); [emergence] 04.v.2020 (1♀, UNB-238116); [emergence] 05.vi.2020 (1♀, UNB-238043).

Comments: The development time of *Cyphomyia wiedemanni,* from collection until the emergence of the adults, varied from 85 to 123 days. The larvae and puparia of *C. wiedemanni* are morphologically similar. A few differences are noticeable, all related to different development stages. We were unable to find features on the surface of the body unique to the larvae or puparia. Both larvae and puparia are flattened dorsoventrally (more pronounced in the larvae than the puparia). The dorsal setae 3 (D3) is always present in the larvae, but is hardly ever observable in the puparia, which are minute, delicate and break easily. In the larvae, the abdominal spiracles are observable dorsally in the segments 2-5, while in the puparia the respiratory horns are present, positioned dorsolaterally.

The general shape of the puparia, as well as the chaetotaxy, have proven conservative across the species of *Cyphomyia*. Even so, *C. wiedemanni* exhibits some distinct differences in comparison to the other species, such as: (1) presence of only one ocular tubercle immediately posterior to the eye, shared with *C. albitarsis* and *C. leucocephala,* but with a slightly less pronounced sulcus between the eyes and tubercles. (2) Chaetotaxy of thoracic segment 1, similar to that of *C. albitarsis,* with 2 pairs of anterodorsal setae (Ad_1, Ad_2) and 3 pairs of dorsal setae (D_1, D_2, D_3). (3) Chaetotaxy of abdominal segments also like *C. albitarsis.* (4) Anal slit length diverges significantly among species, being longer in *C. wiedemanni,* extending for almost the entire length of the 8th abdominal segment. (5) Semispherical projections with the dorsal setae (D) on the dorsal surface of the 8th abdominal segment, as in *C. leucocephala* e *C. albitarsis*.

In addition to the record from this study for the Distrito Federal, the distribution of *Cyphomyia wiedemanni* was expanded to all regions of Brazil based on records from iNaturalist (https://www.inaturalist.org/taxa/294692-Cyphomyia-wiedemanni): North (Acre), Northeast (Alagoas,

Bahia, Pernambuco), Southeast (Espírito Santo, São Paulo) and South (Paraná, Rio Grande do Sul).

DISCUSSION

The terrestrial larvae of Stratiomyidae are cryptic and are generally associated with dim and humid locations like soil, leaf litter, rotten trunks, dung and even in animal carcasses, feeding on the microorganisms associated with the decaying organic matter. An exception to these habits is the larvae of the Chiromyzinae brown soldier-flies genera *Chiromyza* Wiedemann, 1820 and *Inopus* Walker, 1950, which live in soil and have adaptations like tubercles on the head and cylindrical bodies that allow for the excavation of soil with a degree of efficiency. Beyond that, they also possess well-developed and functional mandibles that are used to cut plant roots for feeding (Pujol-Luz & Vieira, 2000). Some species cause damage to crops, like *Inopus rubriceps* (Macquart, 1847) and *Inopus flavus* (James, 1968), considered sugar cane (*Saccharum* spp.) pests in Australia (Bull, 1976). In Brazil, the larvae of *Chiromyza vittata* Wiedemann, 1820 feed on the roots of coffee plants (*Coffea arabica* Linnaeus), in depths that vary from 30 to 40 cm in soil (D'Antonio, 1991; Pujol-Luz & Vieira, 2000). In the other Stratiomyidae, the mouth apparatus is composed of a mandibular-maxillary complex used for the filtering of microorganisms present in different kinds of substrate (McFadden, 1967).

As in the previous study by Pujol-Luz *et al.* (2023) with *Hermetia teevani* Curran, 1934, the larvae of *C. wiedemanni* were collected buried in soil feeding on the roots of cassava. *Hermetia teevani* and *C. wiedemanni* belong to distinct subfamilies, Hermetiinae and Clitellariinae respectively, and do not possess any adaptation or morphological modification related to the excavation of soil or the processing of solid material, as is the case for the immature forms of Chiromyzinae. The presence of larvae of *C. wiedemanni* in the roots of cassava was probably enabled by previous damage caused by other organisms, as found by Pujol-Luz *et al.* (2023) for *H. teevani*. Where, the presence of larvae of *H. teevani* in the roots of cassava would be a consequence of damage caused by the larvae of cassava borer beetles (*Eubulus:* Coleoptera, Curculionidae). The cassava borer larvae make galleries in the root's interior and allow for the entry of microorganisms that cause the decaying of the roots (Oliveira *et al.,* 2019). This kind of opportunistic relationship has also been recorded for *Cyphomyia*. McFadden (1967) collected larvae of *C. bicarinata* feeding on fluids from damaged parts or decaying tissue from cacti (*Opuntia* sp.), and James (1957) collected larvae of *C. pilosissima* in fluid found at the base of Agave plants (*Agave* sp.), in both cases there was no evidence that the larvae had caused the damage to the plants. McFadden & James (1969) collected larvae of *C. erecta* in a rodent's nest made in a cavity in a cactus (*Platyopuntia* sp.). As in previous cases, the presence of the larvae associated with the rodent's nest inside a cavity in the cactus, it is likely due to an opportunistic behavior, since the larvae alone could not be capable of causing damage to the cactus. As such, similar to the findings by Pujol-Luz *et al.* (2023) for *H. teevani, C. wiedemanni* would be considered a secondary invader, taking advantage of an opportunity to feed on the damaged roots attacked by the cassava borer (*Eubulus*).

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