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Description of the last larval instar and pupa of *Chlorota paulistana* Ohaus, 1912 (Coleoptera: Melolonthidae: Rutelinae: Rutelini)

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ABSTRACT

The first immature description for a South American species of the genus *Chlorota* Burmeister, 1844 (Melolonthidae: Rutelinae) is presented. The last larval instar and female pupa of *Chlorota paulistana* Ohaus, 1912 are described. Illustrations and diagnostic characters are also presented, along with additional characters for the genus, and a new state record for Pernambuco is added. With this study, two species of immatures of *Chlorota* are now known and their diagnostic characters and differences are here discussed.

Introduction

The Anticheirina (Melolonthidae: Rutelinae: Rutelini) are leaf chafers with 44 genera and nearly 550 species and subspecies (Moore et al., 2014). Anticheirine scarabs include many brightly colored, metallic beetles, ranging in size from 05 to 40 mm; they are characterized by the frontoclypeal suture indistinct at middle, clypeus without contractions on the posterolateral angles, pronotum without posterior marginal bead, and metatibiae with spine-like apical setae (Moore et al., 2014; Morón, 1997; Soula, 2005).

Knowledge about Anticheirina life history is scarce, and immature stages are known for only a handful of species (Moore et al., 2014). Indeed, within Rutelini, only 23 genera and 34 species have their larvae described, and six genera and 14 species have their pupae described (Albertoni et al., 2014; Bento et al., 2018). About Anticheirina, only five genera and 14 species have larvae described, and four genera and nine species have pupae described (see Albertoni et al., 2014). Immatures are frequently found in rotten wood, sometimes in association with adults, but they are rarely preserved or described (Morón and Paucar-Cabrera, 2003).

The genus *Chlorota* Burmeister, 1844 (Anticheirina) includes about 35 species distributed from southern Mexico to southeastern Brazil (Soula, 2002, 2006, 2008). Adult specimens of the genus are diurnal, and usually collected with fermented fruit traps in Brazil (Jameson and Morón, 2001). In the Brazil nine species are known (Grossi and Vaz-de-Mello, 2019). Regarding the taxonomic knowledge of their immature stages, only the larvae and pupae of *Chlorota cincticollis* Blanchard, 1850 have been described (Jameson and Morón, 2001).

This paper aims to contribute with the taxonomic knowledge of the immature stages of *Chlorota*. Pupa and last larval instar of the Brazilian species *Chlorota paulistana* Ohaus, 1912 are described. Diagnostic characters and illustrations are presented along with a discussion presenting new diagnostic larval characters for the genus, and a new state record for Pernambuco is added.

Material and methods

Larvae of *C. paulistana* were collected in urbanized Atlantic Forest area located at the private woods Oficina de Cerâmica Francisco Brennand, Várzea, Recife – Pernambuco, −8.050811°S/−34.979714°W, 55 m, Northeastearn Brazil. Three larvae were found inside a rotten trunk about 20 cm in diameter in October 2016, and they were maintained in laboratory inside plastic pots containing the crushed rotten wood, that they were found in, for three months. At the end of this period, one last instar larva, one female pupa and one male adult were obtained. The immatures were killed in hot water and fixed in alcohol 80%. The adult was killed in ethyl acetate, its genitalia was extracted

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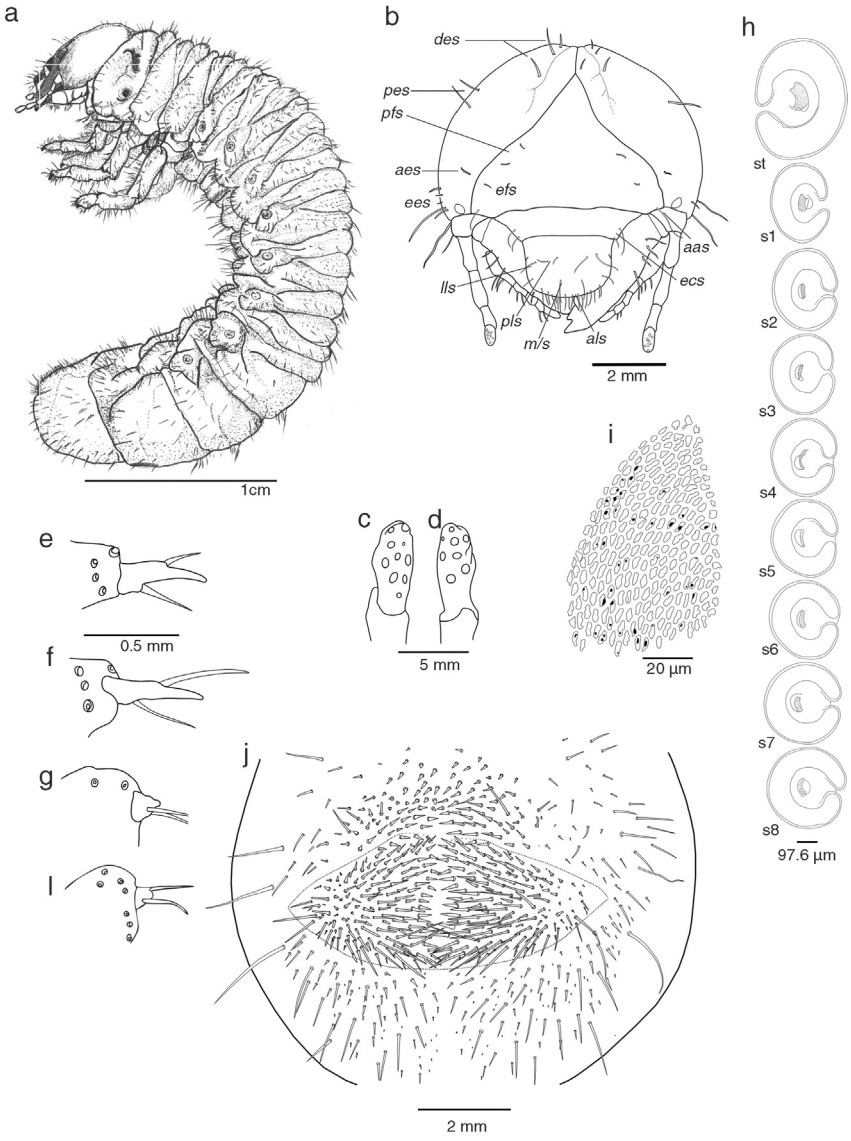

Fig. 1. *Chlorota paulistana* Ohaus, 1912; third larval instar. (a) Lateral; (b) head; (c–d) antennomere IV (dorsal, ventral); (e–g) external pro-, meso- and metapretarsus; (h) spiracles; (i) detail of the dorsal arm of abdominal spiracle I; (j) raster. Chaetotaxy (italic) on the text; st, thoracic spiracle; s1–8, abdominal spiracles I–VIII. *Macraspis festiva* Burmeister, 1844. (l) External metapretarsus.

and the specimen was pinned. All specimens were deposited in CERPE (Coleção Entomológica da Universidade Federal Rural de Pernambuco, Recife, Brazil - Paschoal Grossi curator).

Illustrations were made using a stereomicroscope OLYMPUS SZX12 coupled with camera lucida and treated with the software GIMP version 2.8. Spiracles' illustrations were made using a microscope OLYMPUS BX41 coupled with camera lucida. Measurements were obtained with a pachymeter and a micrometer.

Terminology of larvae follows Böving (1936) and Lawrence (1991), head chaetotaxy and pupa terminology follow Sousa et al. (2018).

Results

Chlorota paulistana *Ohaus, 1912*

Third larval instar (Figs 1 and 2)

Description (Fig. 1a). *Total length:* 37.35 mm. *Color:* Yellowish white. *Head* (Fig. 1b): Length: 6.20 mm, width: 5.45 mm. Color reddish brown. Posterior half of frons reddish brown, anterior half brown, surface densely rugopunctate. Epicranial suture present, each side of epicranium with 3–4 dorsoepicranial setae (*des*), 1–2 posteroepicranial setae (*pes*), 3–7 externoepicranial setae (*ees*), 1 anteroepicranial seta (*aes*), 1–2 posterofrontal setae (*pfs*), 1 externofrontal seta (*efs*), 1 anterofrontal angle seta (*aas*); anterofrontal setae absent. Stemmata present. Clypeus trapezoidal, densely rugopunctate, each side with 2 externoclypeal setae (*ecs*); anteroclypeal setae absent. Labrum strongly rugose, lobate, densely punctate, each side with 5 anterolabral setae (*als*), 2–3 laterolabral setae (*lls*), 1 mediolabral seta (*mls*), 2 posterolabral setae (*pls*). Epipharynx (Fig. 2j) oval and wider than long. *Corypha* (Fig. 2l) with epizygum and clithra absent. *Haptomerum* with 19 spine-like setae and 12 sensilla, zygum crossbar-like and slightly sclerotized, heli absent. *Paria*, acroparia with 6–7 setae on each side, acanthoparia with 9 short setae on left side, 11 on right side. Plegmatia and proplegmatia absent. Right chaetoparia with 26 sensilla and 21 spine-like setae, left chaetoparia with 12 sensilla and 8 spine-like setae, phobae absent. *Pedium* oval. *Haptolachus* with 2 setae on right side and 12 setae on left side, sensorial cone prominent and acuminate, sclerotized plate indistinct, crepis indistinct. Laeotorma

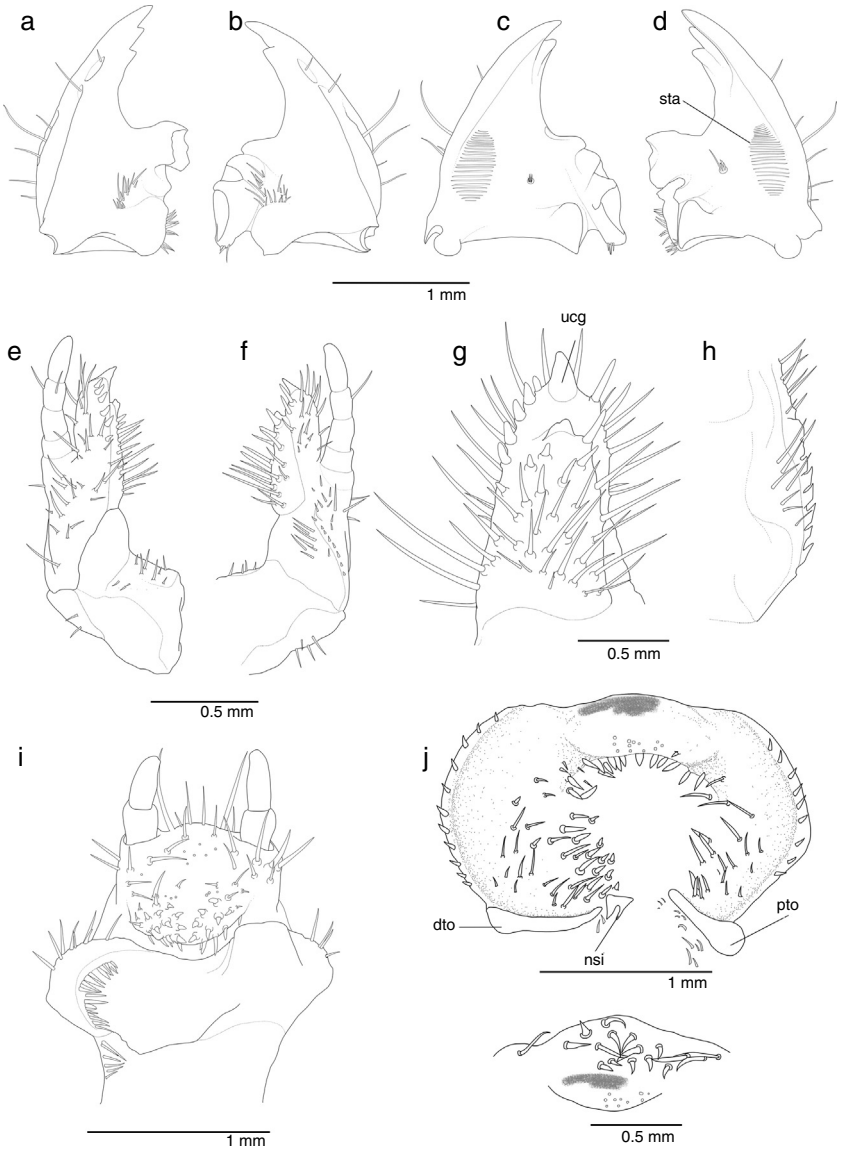

Fig. 2. *Chlorota paulistana* Ohaus, 1912; third larval instar; head appendages. (a–b) Mandibles, dorsal (left, right), (c–d) mandibles, ventral (right, left); (e–f) maxilla (ventral, dorsal), (g) mala, internal, (h) stipes stridulatory teeth detail; (i) labium, dorsal; (j) epipharynx. dto, dexiotorma, (l) detail of the corypha; nsi, sensorial cone; pto, pterotorma; sta, stridulatory area; ucg, uncus.

short, pterotorma rounded, dexiotorma elongate. Mandibles asymmetrical, dark, surface rugose and punctate. Left mandible (Fig. 2d), incisor with three teeth; dorsal surface (Fig. 2a) with 2 longitudinal lateral furrows, 7 dorsolateral setae, molar with 3 tubercles, distal tubercle bigger than other ones, acia indistinct, brustia with 11 setae. Right mandible (Fig. 2c), incisor with 2 teeth; dorsal surface (Fig. 2b) with 2 longitudinal lateral furrows, 6 dorsolateral setae, molar with 3 tubercles, basal tubercle bigger than other ones, brustia with 3 setae. Stridulatory area oval, formed by 24–28 transverse carinae, with a group of setae beside it. Maxillae symmetrical (Fig. 2e–h), mala setose and with a weak dorsal separation between galea and lacinia. Galea with a conical uncus, dorsal surface with 15 setae and a longitudinal row of 6 robust spine-like setae, ventral surface with 13 setae. Lacinia with a truncate uncus, dorsal surface with 32 setae. Palpus 4-segmented: III with 1 seta, IV with several minute apical sensilla. Stipe with stridulatory area bearing a longitudinal row of 7 short and truncate teeth. Labium, each side of submentum with 3–4 setae. Each side of mentum with 1 long posteromedial seta. Prementum with 10 apical setae and 2 posterolateral setae on each side; each side of ligula (Fig. 2i) with 15–19 posterior spine-like setae and 9 lateral long setae, anteromedial area with 6 sensilla, posterior area with 3 sensilla on each side, right side with 1 lateral sensillum. Hypopharynx with hypopharyngeal sclerome asymmetrical with right tooth truncate and short, right lobe with 4 setae in anterior margin, left lobe with 8 setae in anterior margin and a row of 27 setae. Antennae with 4 antennomers, distal article (Fig. 1c and d) with 10 dorsal sensorial spots and 8 ventral sensorial spots. *Thorax:* Prothorax with tergum and laterotergites pubescent, simply lobed. Thoracic spiracle (Fig. 1h–i) with C-shaped respiratory plate, distance of respiratory plate arms shorter than the dorso-ventral diameter of bulla. Meso- and metathorax dorsally and ventrally with a transverse row of setae. *Legs:* Surface densely setose, all pretarsi (Fig. 1e–g) with two long setae, metapretarsus relatively reduced and weakly sclerotized. *Abdomen:* 10-segmented, segments I–VI with 3 dorsal minutely pubescent lobes, some setae short or long, segments VII–X simply lobed, almost smooth, segments IX–X enlarged. Spiracles (Fig. 1h) smaller than thoracic, C-shaped. Raster (Fig. 1j), each palidium with about 25 acute pali, each side of tegillar area with more than 190 spine-like setae. Anal

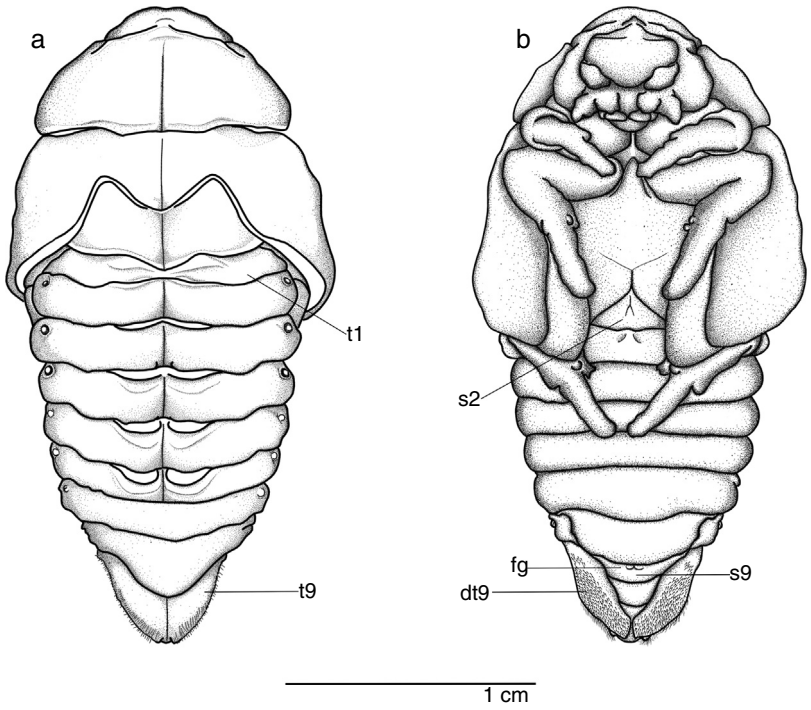

Fig. 3. *Chlorota paulistana* Ohaus, 1912; female pupa. (a) Dorsal, (b) ventral. dt9, ventral part of tergite IX; fg, female genital ampulla; s1–9, abdominal sternite I–IX; t1–9, abdominal tergite I–IX.

ventral lobe with about 157 small and long setae. Anal opening transverse.

Female pupa (Fig. 3)

Description (Fig. 3a and b). *Overall form*: Oval shaped. *Total length:* 24.2 mm. *Width:* 9.5 mm. *Color:* Yellowish. *Head:* Vertex visible dorsally, epicranial suture weakly defined. Clypeus and labrum rectangular (frontal view). Mandibles, maxillae, labial and maxillary palpi tubercle-like. Labium rounded. Antennae triangular. *Thorax:* Prothorax convex, trapezoid, ecdysial longitudinal line well-marked, anterior and posterior margin sinuous. Thoracic spiracle presents laterally between the anterior and medial legs and the hypomeron. Scutellar area weakly delimited. Metanotum slightly backwardly projected. Thoracic ventrite process well defined and produced between mesocoxae. Legs with meso- and metatibial with two tubercle-like spurs. Pterothecae curved ventrally. *Abdomen*: With 9 segments visible dorsally, sternite I constrict and coalesced, sternite II–X visible ventrally, sternite IX of females bearing a small and 2-tuberculate genital ampulla, tergite IX with lateral lobes pubescent. Dioneiform organs present between segments I–II, II–III, III–IV, IV–V, V–VI. Spiracles: I–IV large and with sclerotized peritreme, V–VIII small and represented by a cuticular invagination, spiracle I concealed by pterotheca.

Remarks. Male pupae unknown.

Discussion

Among the known Anticheirina larvae, those of *Chlorota* are similar to those of *Lagochile* Hoffmannsegg, 1817 and *Macraspis* MacLeay, 1819. Larvae of these three genera have the epipharynx without clithra, left mandible with incisor bearing three teeth, lacinia with reduced uncus and raster with palidia and septula present in ventral anal lobe (Morón and Paucar-Cabrera, 2003). However, *Chlorota* and *Lagochile* larvae are separated by the presence of acuminate metapretarsi, while *Macraspis* present the pointless metapretarsi and poorly sclerotized in relation to the pro- and mesothoracic pretarsi (Fig. 1l). *Chlorota* and *Macraspis* are distinguished by the ligula with short and stout setae (slender and moderately long in *Lagochile*).

The larvae of two species of *Chlorota* are known, and *C. paulistana* can be differentiated as follows (opposition to *C. cincticollis* between brackets): stemmata present (absent), frons densely rugopunctate (punctate), antennomere IV with 10 dorsal sensorial spots (4 dorsal spots) and 8 ventral sensorial spots (3 ventral spots), molar with parabolic tooth (truncate tooth). *Chlorota cincticollis* was described with epipharynx without zygum and with 10 heli (Jameson and Morón, 2001), but following Boving's terminology (see discussion in Albertoni et al., 2014, p. 42), the present study interpreted *C. paulistana* having zygum (bar-like and slightly sclerotized) and both species without heli but with some stout spine-like setae.

Soula (2002) organized *Chlorota* in four groups of species: *Chlorota aulica*, *C. limbaticollis*, *C. terminata* and *C. caucana* groups. *Chlorota paulistana* belongs to the *terminata* group, while *C. cincticollis* belongs to the *limbaticollis* group. Is possible that when more species have their immatures described, characters of the immatures stages shall provide important information for a clearer delimitation of these groups.

According to Morón (1993), Rutelini pupae are characterized by the presence of five pairs of dioneiform organs, spiracles I–IV with sclerotized peritreme and spiracles V–VIII as cuticular invagination, as well as the absence of urogomphi, and tergo-lateral tubercles absent. The pupa of *C. cincticollis* is similar to *Macraspis*, but in *Macraspis* tarsi and posterior margin of the mesonotum are more elongated (Jameson and Morón, 2001). *Chlorota paulistana* pupa is very similar to the *C. cincticollis* pupa, but former species can be distinguished by (opposition to *C. cincticollis* between brackets): sternite VIII and IX well defined (coalesced); pubescence of ventral fold of tergite IX present in all lateral extension (pubescent restricted to posterior half). Bento et al. (2018) presented a key to the known Neotropical Rutelini pupae. To include the present study's results, that key should by emended as follows:

6. Abdominal ventrite VI slightly longer than I–V combined..... *Rutela dorcyi*

6′. Abdominal ventrite VI much shorter than I–V combined..... *Chlorota*... A

A. Pubescence of ventral fold of abdominal tergite IX present in all lateral extension.....*C. paulistana*

A'. Pubescence of ventral fold of abdominal tergite IX restricted to posterior half.....*C. cincticollis*

Conflicts of interest

The authors declare no conflicts of interest.

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