

# Papéis Avulsos de Zoologia

Museu de Zoologia da Universidade de São Paulo

Volume 53(27):359-372, 2013

www.mz.usp.br/publicacoes  
http://portal.revistasusp.sibi.usp.br  
www.scielo.br/paz

ISSN impresso: 0031-1049  
ISSN on-line: 1807-0205

## FIRST INSTAR LARVA OF *ATRACTOCERUS BRASILIENSIS* (LEPELETIER & AUDINET-SERVILLE, 1825) (LYMEXYLIDAE, ATRACTOCERINAE)

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### ABSTRACT

*The first instar larva of Atractocerus brasiliensis (Lepetelier & Audinet-Serville, 1825) from state of Goiás, Brazil, and reared in laboratory, is described and illustrated. This is the second species with known first instar larva for the genus and the third for the family. Comparisons with mature larva of this species and with the other known first instar larvae are presented. Illustrations of first instar and mature larvae are also included.*

KEY-WORDS: Coleoptera; *Hylecoetus*; Immature; Mature larva; Neotropical region.

### INTRODUCTION

The first instar larvae of Coleoptera are poorly known, especially due to the difficulty of maintaining adults capable of copulating within a laboratory, or to the difficulty of finding fertilized females. Usually it is easier to collect and keep mature larvae in laboratory until reaching adult stage. First instar larvae have been described for only two species of Lymexylidae: one belonging to the genus *Atractocerus* Palisot de Beauvois, 1801, and the other to *Hylecoetus* Latreille, 1806.

Fulmek (1930) described the first instar larva of *Atractocerus emarginatus* Castelnau, 1836, obtained from eggs of one female collected in Medan, North Sumatra, province of Indonesia – “Medan (Deli, Sumatra)”.

Simmonds (1956), in treating the biological control of *Melittomma insulare* Fairmaire, 1893,

presented the biological data of the first instar and mature larvae, as well as pupa, of *Atractocerus brasiliensis* from Trinidad. He illustrated the eggs, first instar and mature larvae, and pupa; and further illustrated male and female heads and terminal abdominal segments.

Grandi (1962) described and illustrated the “larva neonata” of *Hylecoetus dermestoides* (L., 1761), reared from eggs laid after collecting male and female in Val di Fassa, near Pian di Trevisan, northeast Italy.

In addition, Costa Lima (1953), in treating Lymexylidae, presented three illustrations (*l.c.* Fig. 137) of “larvae of *Atractocerus* sp.” that look like first instar larva.

Casari & Teixeira (2011) described the mature larva of *Atractocerus brasiliensis* collected in *Pinus oocarpa* Schiede ex Schltld. (Pinaceae). After the publication, the first instar larvae of this species were obtained in laboratory by the second author of this

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paper (FFA) and is hereby described, illustrated, and compared with other known first instar larvae and mature larva of this species.

## MATERIAL AND METHODS

The junior author (FFA) collected one female attracted by home light in the city of Goiânia, state of Goiás. The female was wounded between abdominal segments 6 and 7, exposing many eggs; but even while injured it laid more than one hundred eggs. The emergence occurred 20 days after oviposition and one day after the four surviving larvae were fixed.

The studied material is housed at the Museu de Zoologia da Universidade de São Paulo (MZSP).

Scanning Electron Micrographs (SEM) were prepared at the MZSP laboratory with Carl Zeiss microscopy LEO 440, controlled by technician Lara Maria Guimarães. As we are not aware of any reference for Lymexylidae chaetotaxy, the topology of setae follows Ratcliffe & Skelley (2011) and Vanin *et al.* (2012).

The mites collected on the abdomen of *Atractocerus brasiliensis* were treated with lactic acid; each specimen was placed on a slide and coverslip with Royal solution and examined in a Leica DM 5000B optic microscope. The pictures presented here originate from 10 (Fig. 50) and 3 (Fig. 51) pictures merged in Helicon Focus 5.3 and taken with Leica Application Suite 4.1.0.

Mite identification and classification follow Krantz & Water (2009).

## RESULTS

### *Atractocerus brasiliensis*

*Eggs* (Fig. 42): length: 1.8-2.0 mm; width 0.6-0.8 mm.

Cream, elliptical, without ornamentation.

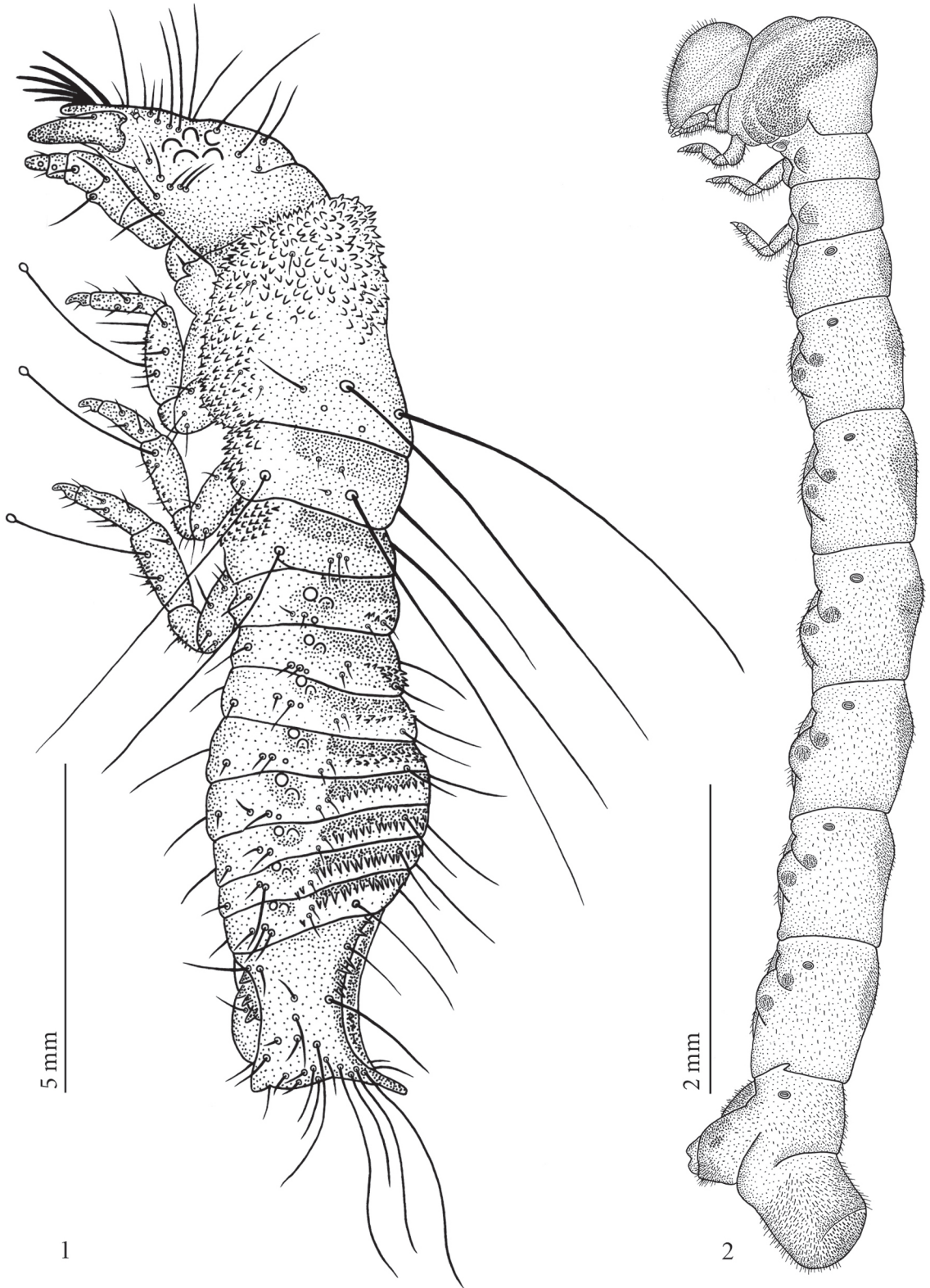
The eggs were laid in two batches of about 80 eggs each, among a gumming whitish substance. The majority of eggs did not eclose and 23 were fixed. After eclosion the larvae stayed for a short time moving among the egg shells and the gumming substance (Figs. 43-45).

*First instar larva* (Figs. 1, 3-15, 25-41, 43-47): length: 2.0-2.1 mm

Elongate, cylindrical, slightly flattened, with apex narrowed and upwardly directed. General coloration cream (Figs. 43-47); head yellowish and

mandibles brownish with black apices; labrum brownish; dark-brown transverse band at base of pronotum, at middle of meso- and metanotum and on segments I-VIII (some divided at middle); abdominal segments with a dark-brown lateral patch on each side; segment IX very sclerotized, ferruginous with darker apex; stemmata black (Figs. 45-46). General pubescence yellowish and very long; microspines in specific areas of thorax and abdomen (Figs. 1, 25-26, 28, 45-47).

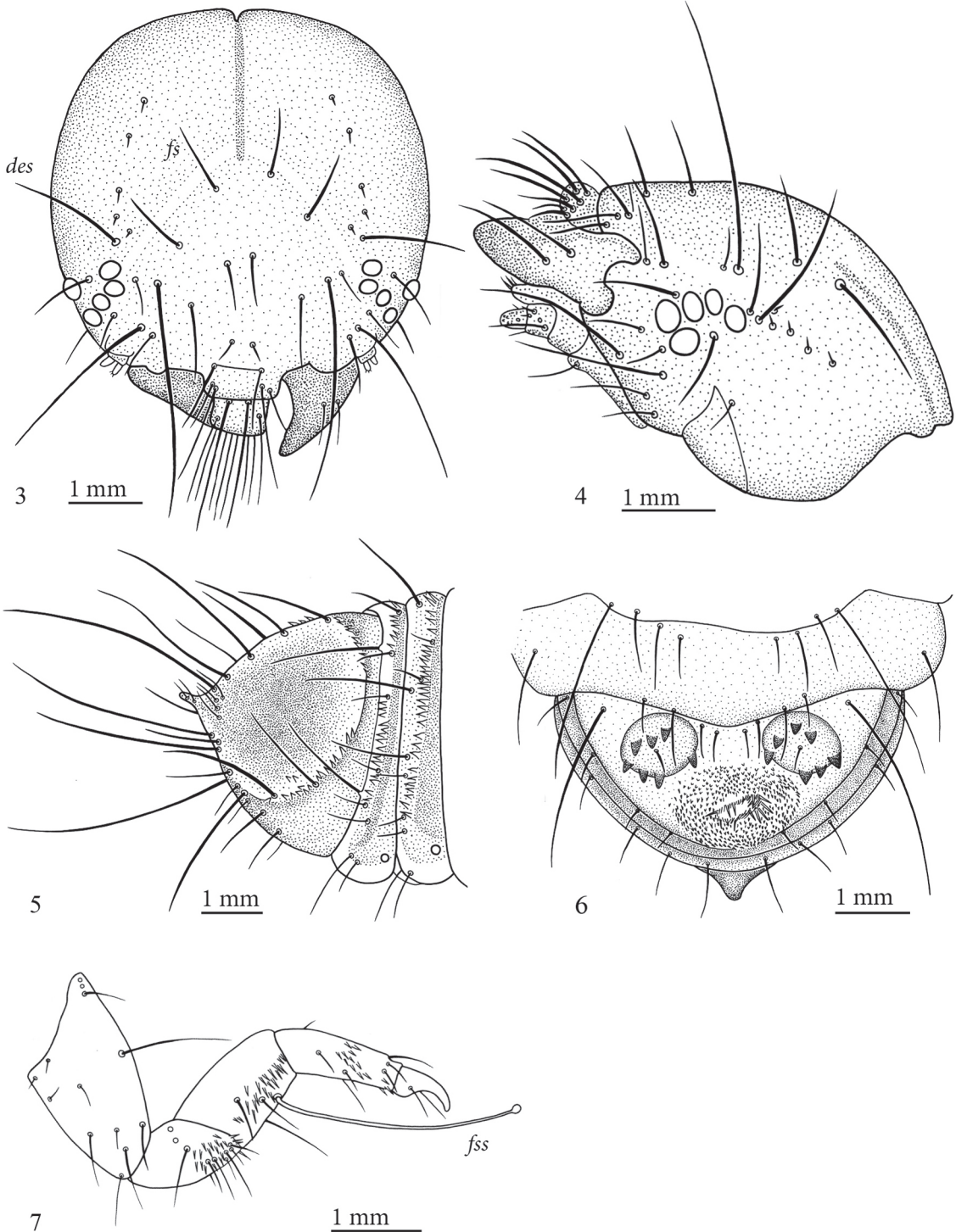
Head (Figs. 3-4, 25, 27, 29, 43, 45-47) hypognathous partially retracted into pronotum, visible dorsally; oval, heavily sclerotized. Coronal suture long, straight; endocarina as long as coronal suture; frontal arms not well defined, visible like a pale band with undefined end. Frons with ten pairs of frontal setae (*fs*) of varied sizes, some longer than half of head length; dorsally each side with a row of five short dorso epicranial setae (*des*) and two longest near stemmata. Five black stemmata on each side, behind antennal base: two basally located and three in a semi-circle above anterior marginated by several setae. Antennae (Figs. 3, 10-11, 25, 27, 30-32) minute, posterolateral to base of mandibles, in a groove that comes frontally from anterior stemmata (Figs. 25, 27, 23, 32); invaginated and surrounded by an antennifer-like structure with very long distal projections (Figs. 30-32), visible only in SEM; antennifer membranous, well developed; two antennomeres: basal wider than long, bearing at apex a well-developed membranous sensorial appendix (*sa*), two long setae (one longest) and two trichobothria; distal antennomere narrow, shorter than sensorial appendix, inserted laterally at apex of basal antennomere, side by side with sensorial appendix, bearing four or five long setae at apex (two widest) and one trichobothrium visible only in SEM. Frontoclypeal (*fcbs*) suture (Figs. 3, 9, 27) well visible only at middle between two basal setae of clypeus. Clypeus (*cl*) narrow, transverse with fore angles rounded, bearing three pairs of long setae (one pair longest). Labrum (*lbr*) (Figs. 3, 9, 27) narrow, transverse, proximal margin as wide as distal margin, distal margin slightly rounded, bearing ten very long setae, more than twice as long as labrum length: six at anterior margin, two near fore angles and two at base. Epipharynx (Fig. 12) slightly prominent ventrally, becoming slightly raised longitudinally at middle; distal margin slightly rounded with three long setae (one sinuous) on each side and four at middle; each side of raised area with microsetose band convergent medially, continuous with longitudinal median sclerite. Mandibles (Figs. 13-15) symmetrical, heavily sclerotized, broad, short with wide apex and subapical rounded lobe; molar area weakly developed with rows of irregular teeth (*mit*); two lateral setae.



FIGURES 1-2: *Atractocerus brasiliensis* (Lepelletier & Audinet-Serville, 1825). Larva lateral: (1) first instar; (2) mature (from Casari & Teixeira, 2011).

Maxilla (Figs. 8, 33-34) elongate; cardo (*crd*) subtriangular bearing three moderately long setae; stipes elongate bearing one short seta at base and eight long

setae (two represented by punctures); palpifer with one long palpiferal seta (*pf*); mala elongate, longer than palpus, with five long malar setae (*ms*) at apex;

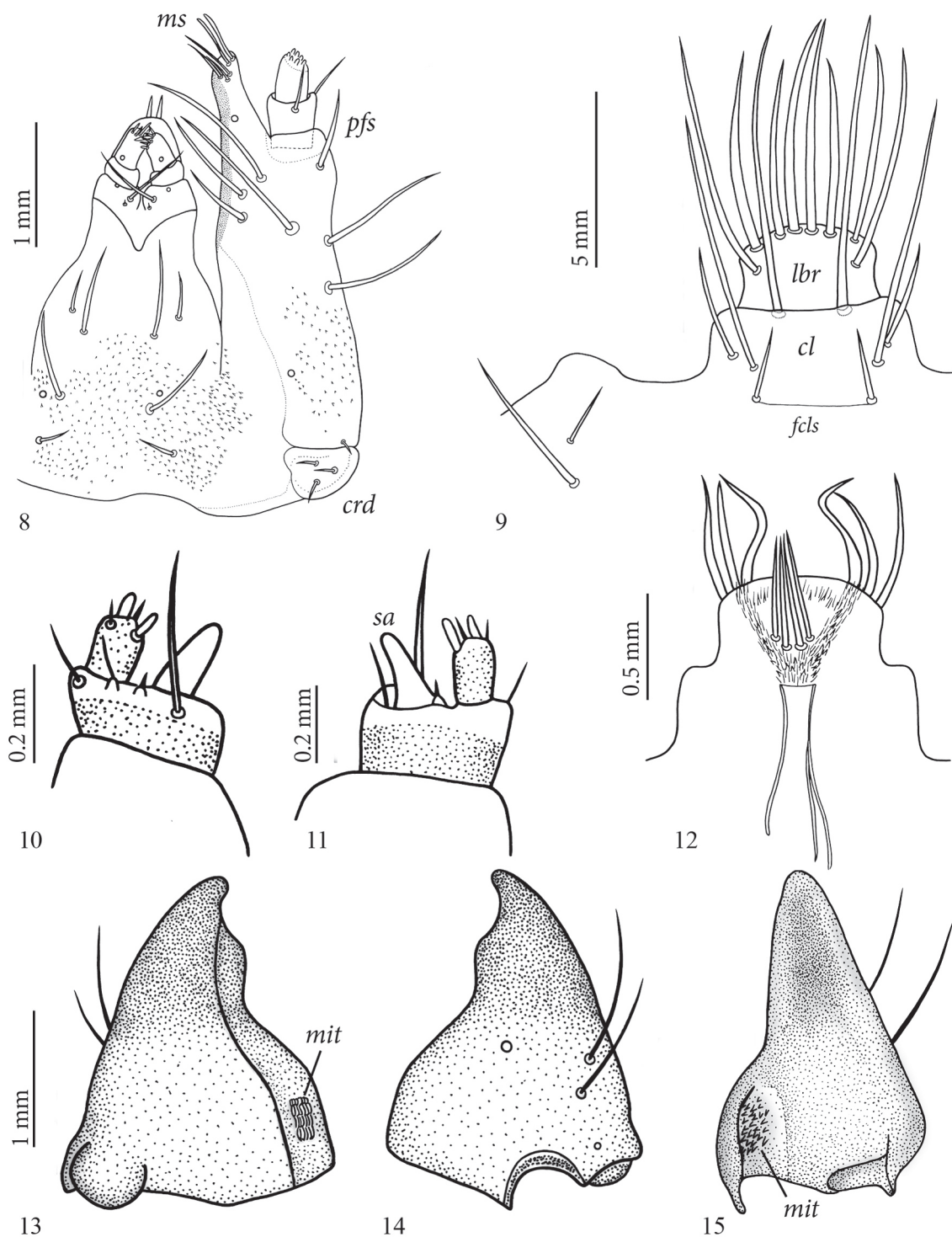


**FIGURES 3-7:** *Atractocerus brasiliensis* (Lepelletier & Audinet-Serville, 1825). First instar larva: (3-4) head (dorsal, lateral); (5) segments VI-IX (dorsolateral); (6) segments VIII-X (ventral); (7) metathoracic leg (lateral). *des* = dorso epicranial setae, *fs* = frontal setae, *fss* = femoral spatulate seta.

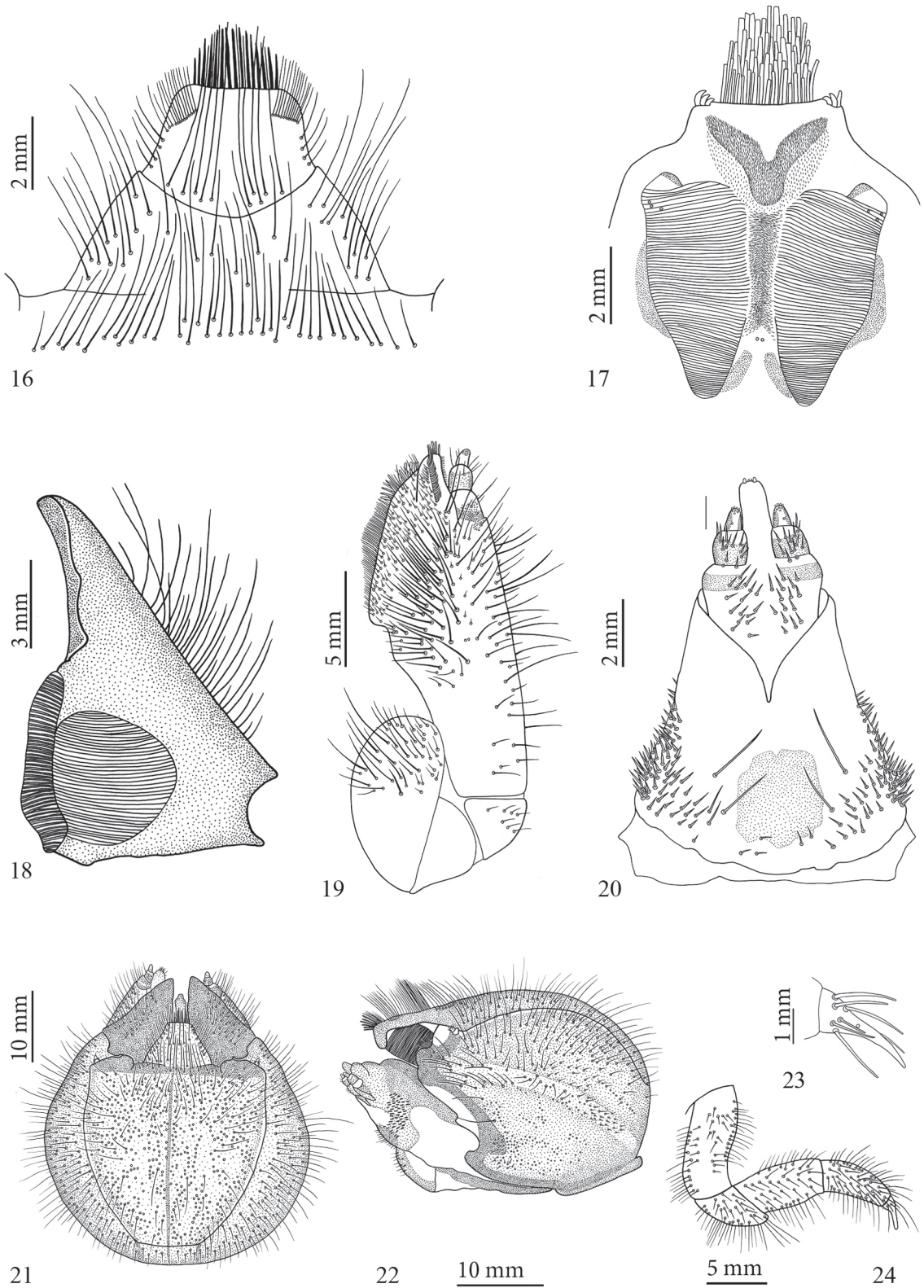


palpus with two palpomeres: basal as wide as long with two long setae, distal elongate, narrower than basal with several peg-like sensilla at apex. Labium

(Fig. 8) elongate; prementum short, transverse, basal margin strongly prominent at middle, making basal half triangular; palpiger with one long and one tiny

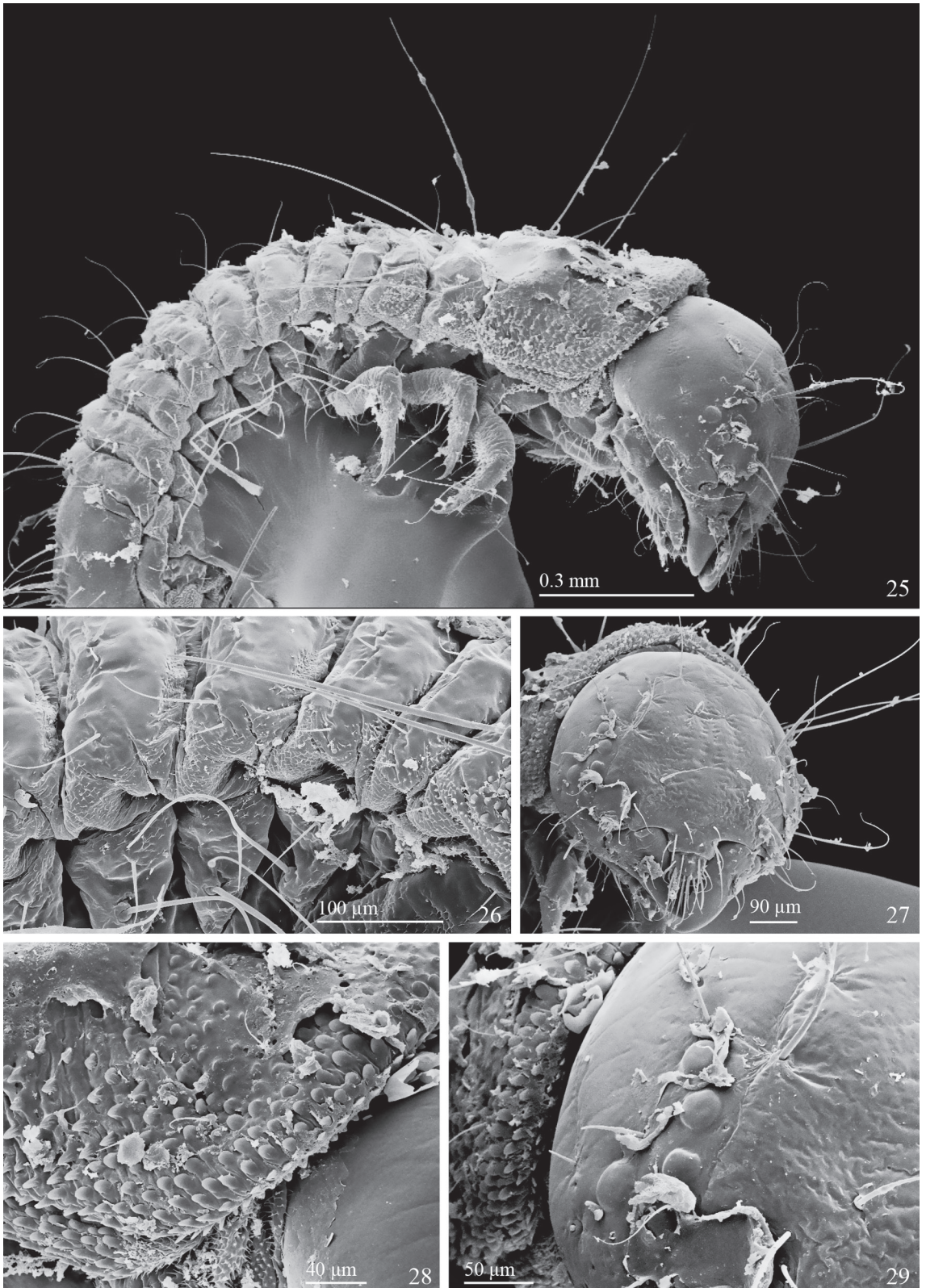


**FIGURES 8-15:** *Atractocerus brasiliensis* (Lepelletier & Audinet-Serville, 1825). First instar larva: (8) maxillo-labial complex; (9) clypeus and labrum; (10-11) right antenna (dorsal, ventral); (12) epipharynx; (13-15) right mandible (ventral, dorsal, mesal); *cl* = clypeus, *crd* = cardo, *fcls* = fronto clypeal suture, *lbr* = labrum, *mit* = molar irregular teeth, *ms* = malar setae, *pfs* = palpifer setae, *sa* = sensorial appendix.



**FIGURES 16-24:** *Atractocerus brasiliensis* (Lepeletier & Audinet-Serville, 1825). Mature larva (from Casari & Teixeira, 2011): (16) clypeus and labrum; (17) epipharynx; (18) mandible (dorsal); (19) maxilla (ventral); (20) labium (ventral); (21-22) head (dorsal, lateral); (23) prothoracic tarsal claw; (24) metathoracic leg (lateral).





**FIGURES 25-29:** *Atractocerus brasiliensis* (Lepelletier & Audinet-Serville, 1825). First instar larva (SEM): (25) habitus (lateral); (26) first abdominal segments (lateral); (27) head (frontal); (28) fore angle of pronotum with sharpened and rounded microspines; (29) head (fronto-lateral).

seta and one campaniform sensillum; palpus with two palpomeres: basal transverse; distal elongate with one campaniform sensillum near base and several elongate setae at apex; ligula elongate, trapezoidal, longer than palpi, with two setae at apex; mentum elongate, narrowed on distal third; distal margin strongly notched at middle with four pairs of long setae and one pair of campaniform sensilla.

Prothorax (Figs. 1, 25, 28, 45-46) longer than meso- and metathorax together; pronotum with sharpened and rounded microspines and sparse short setae on anterior and lateral irregular bands; dark-brown band at basal third; each side of darker band with two very long and three moderately short setae (two short represented by punctures) (Fig. 1). Meso- and metathorax similar: transverse, band-like, each with one dark-brown transverse dorsal band and one patch of microspines on each side; mesothorax with one long and three short dorsolateral setae and laterally one very long and one short setae; metathorax with three short dorsolateral setae and one very long lateral. Each side of mesothorax with one ventrolateral anterior spiracle. Legs (Figs. 1, 7, 40-41) increasing in size from anterior to posterior, inserted distant from middle line of venter; coxa elongate with several setae of varied sizes; trochanter subtriangular; femur and tibia elongate; femur wider and as long as tibia; trochanter, femur and tibia with setae of varied sizes and microspines near ventral region; one very long femoral spatulate seta (*fs*); pretarsus claw-like with two setae near base.

Segments I-VIII (Figs. 1, 25-26, 45-47) transverse, band-like, dorsally with dark-brown transverse dorsal band and one patch each side; dorsally with microspines distributed in different ways, increasing in size to apex direction; each side with dorsolateral rounded spiracle, barely or little visible, near a crescent-like scar and two or three short setae (some represented by punctures) below them; each side with one lateroventral short seta and one longest ventral seta. Segments I-IV dorsally with microspines in a transverse irregular band at middle, with two short setae each side and one longest pair dorsally near middle (except segment I). Segments V-VIII dorsally with transverse, narrow, sclerotized, dark-brown band with microspines at distal margin; with two short setae each side and one longest dorsal pair near middle of darker band. Segment IX (Figs. 1, 5, 35-38, 47) strongly sclerotized, concave with apex darker, tooth-like upwardly directed; concavity marginate by microspines and long setae; two pairs of moderately long setae dorsally, inside concavity, near apex. Segment X (Figs. 6, 38-39) rounded, ventral, globose, with two

lobes, each marginated by seven teeth; one distal sclerotized microspined patch, surrounding anal opening.

*Material examined:* BRAZIL. Goiás: Goiânia, Jardim Balneário Meia Ponte (16°36'37.84"S, 49°17'40.68"W). 14.vii.2011, N.A. da Silva & F.F. Albertoni cols., 1 adult, 23 eggs, 4 first instar larvae (one dissected and one gold covered for SEM) (MZSP), 8 deutonymphs mites (Uropodoidea). São Paulo: Salesópolis, Estação Biológica de Boracéia, (23°39'15,63"S, 45°53'22,38"W) 850 m, 25.ii.2012, F.F. Albertoni leg, 1 adult, 5 deutonymphs mites (Uropodoidea) (MZSP).

### Biological notes

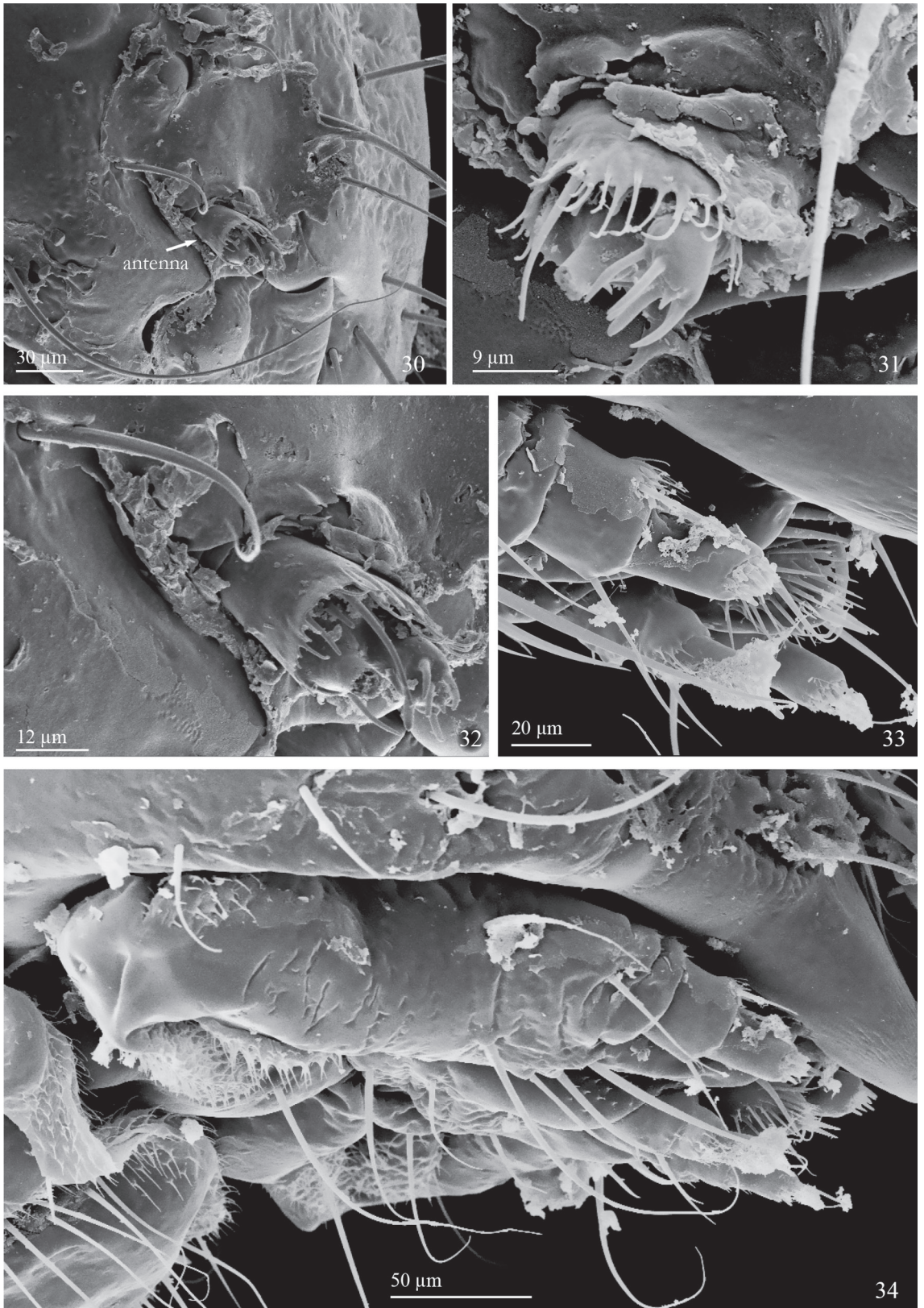
As already observed by Fulmek (1930), the eggs and just eclosed first instar larvae, under laboratory conditions, remained among the whitish viscous substance; they were very difficult to individualize or clean. This is evident in some photos, where the larvae are very dirty (Fig. 47).

Simmonds (1956) reported that the eggs were laid in freshly felled mango logs; they were laid in batches with a very "viscous fluid", which also exuded at times from the female abdomen when no eggs were being laid. According to him, this fluid remained extremely viscous for at least two weeks, even after the eggs had hatched.

After the eggs had hatched, the first instar larvae remained together with the eggshells (Figs. 43-45). According to Simmonds (*l.c.*), they ate the empty eggshells in the first 24 hours, becoming very active after that. According to Wheeler (1986), during the contact of the larvae with siblings and eggs, transference of fungal spores onto the integument would occur. The female would emerge bearing spores in vaginal pouches, eventually depositing them in the slimy matrix with eggs.

According to Lyngnes (1958) (*apud* Grandi, 1962), the first instar larva of *Hylecoetus dermestoides* feeds on fungi that grow at tunnel walls, especially Ascomycete, *Endomyces hylecoeti* Neger. He stated that segment IX of the first instar larva is used to push the frass outside of the tunnel. This protects the larva against insect attacks and also transports small particles, like fungi spores, to inside the tunnel. The modified segment IX, as in mature larva, could be used for the same purpose, as well as to excavate when the larva retreats. Casari & Teixeira (2011) observed mature larvae of *Atractocerus brasiliensis* pushing frass outside of the tunnel.



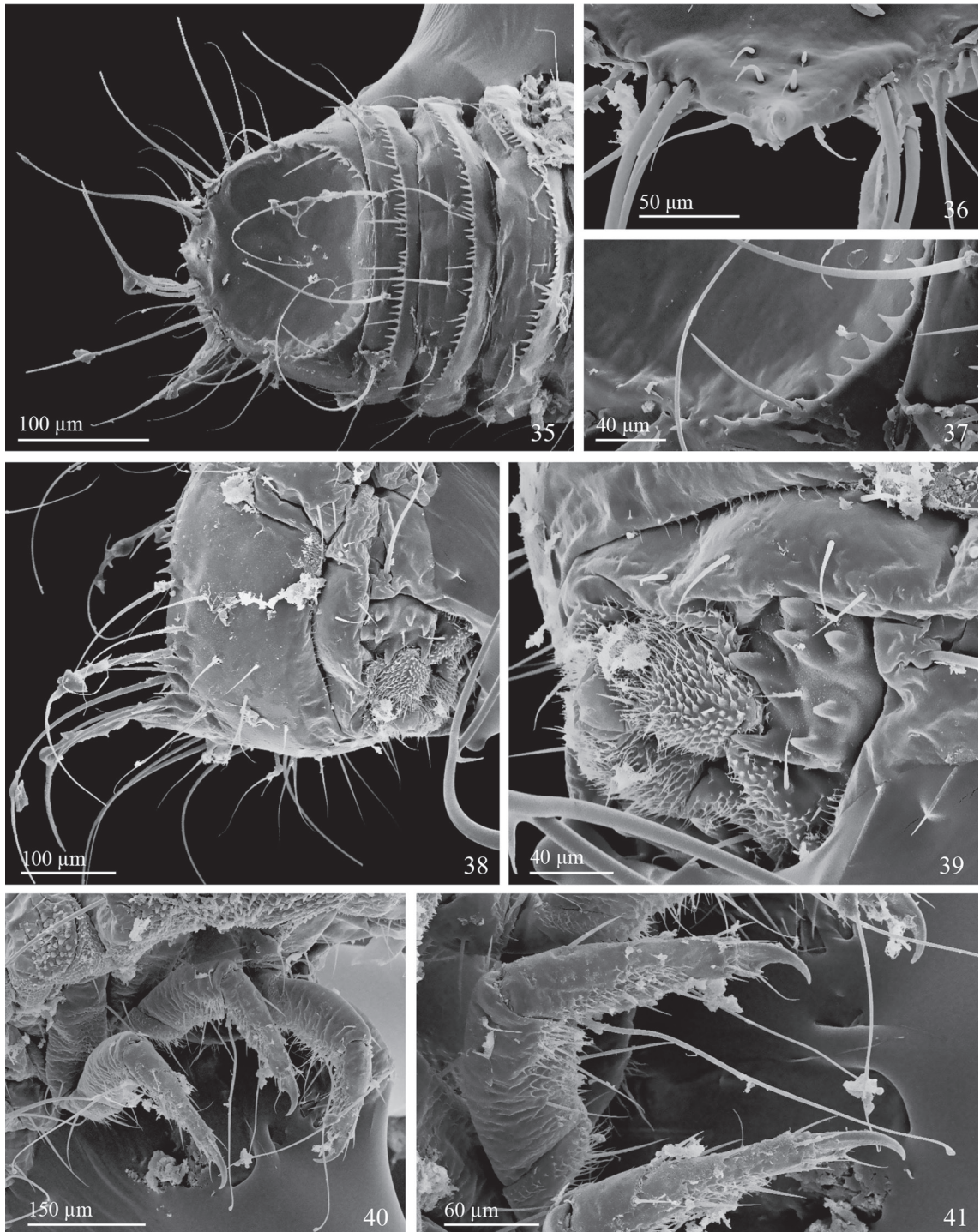


**FIGURES 30-34:** *Atractocerus brasiliensis* (Lepeletier & Audinet-Serville, 1825). First instar larva (SEM): (30) head (latero-posterior); (31-32) antenna (dorso-anterior, latero-posterior); (33) right maxillary palp (lateral); (34) right maxilla (lateral).



FFA observed a curious behavior in an adult collected in Estação Biológica de Boraceia (Salesópolis, São Paulo). When touched or held, the adult curved his abdomen upwards and expanded the pleura of

visible segments 4 and 5, as shown in Fig. 49. The same adult also would rest with abdomen suspended, (Fig. 52) a similar behavior that was observed and illustrated by Casari & Teixeira (2011) when the adult



**FIGURES 35-41:** *Atractocerus brasiliensis* (Lepelletier & Audinet-Serville, 1825). First instar larva (SEM): (35) segments VI-IX (dorsal); (36) apex of segment IX (dorsal); (37) detail of lateral margin of segment IX (dorsal); (38) segments IX-X (ventro-lateral); (39) segment X (ventro-lateral); (40) right legs; (41) median right leg with spatulate seta.



was observed resting upside down on the ceiling of a cage with the abdomen apex downwardly directed.

Several mites (Fig. 50) were found on the abdominal tergites of both specimens collected in Goiânia and Salesópolis. The mites are deutonymphs of the

superfamily Uropodoidea, presumably of the family Trematuridae. The deutonymphs were attached to the beetle integument by the abdomen apex. Lindquist *et al.* (2009) suggested that Trematuridae possibly feed on fungi and organic debris of insects galleries. Near



**FIGURES 42-46:** *Atractocerus brasiliensis* (Lepeletier & Audinet-Serville, 1825). (42) eggs; (43-45) eggs, eggshells and newly eclosed larvae; (46) first instar larva (lateral).



the body of one mite from Goiânia were found spores that resemble ascomycetes conidia (Fig. 51, red arrow) (Larissa Trierweiler, pers. com.) and all analyzed mites

from Goiânia had a distinct fungi spore of another species (Fig. 51, black arrow) (Larissa Trierweiler, personal communication).



**FIGURES 47-53:** (47-49, 52-53) *Atractocerus brasiliensis* (Lepelletier & Audinet-Serville, 1825). (47) first instar larva (dorsolateral). Adult female: (48) dorsal; (49, 53) lateral; (52) abdomen (dorsolateral) with Uropodoidea deutonymphs presumably Trematuridae (ellipses). (50) deutonymph of Uropodoidea mite with fungi spores (black arrows); (51) conidium (red arrows) nearby deutonymph and fungi spores (black arrows).



## DISCUSSION

The eggs of *Atractocerus brasiliensis* are similar to that of *Hylecoetus dermestoides* described by Grandi (1962).

The first instar larva (Fig. 1) of *Atractocerus brasiliensis* differs from the mature larva (Fig. 2) especially by (mature larva parenthesized): body slightly flattened (cylindrical), robust and does not present the large hoodlike pronotum; head visible dorsally (head partially visible dorsally); spiracles rounded (elliptical); pretarsus (Fig. 7) with two basal setae (densely setous (Fig. 23)); segment IX dorsally concave with apex tooth-like and upwardly directed (Figs. 1, 5, 35-37) (cylindrical, bulbous and upwardly directed (Fig. 2)); segment X globose, with two lobes marginate by teeth and distal sclerotized microspined patch (Fig. 6, 38-39) (pygopod-like with two lobes with spicules at apex). The pubescence is very long and moderately sparse in first instar and very short, except head and legs, and dense in mature larva (Figs. 1-4, 7, 21-24). The differences observed in the head and mouthparts are (mature larva parenthesized): head dorsally with 13 pairs of setae of varied sizes (Figs. 3-4) (many setae approximately of same size (Figs. 21-22)); stemmata present (Figs. 3-4, 27, 29, 45-46) (absent); frontal arms marked by a pale band (Fig. 3) (frontal arms well visible (Fig. 21)); frontoclypeal suture incomplete, visible at middle (incomplete, visible only laterally (Fig. 21)); clypeus (Fig. 9) rectangular (trapezoidal (Fig. 16)); labrum (Fig. 9) with eight long setae near anterior margin and two near base (ten long setae near base and a row of setae near each fore angle (Fig. 16)); epipharynx (Fig. 12): three long setae on each fore angle and four setae at middle (three short setae on each fore angle and a bundle of wide setae at anterior margin (Fig. 17)); mandible (Figs. 13-15): two lateral setae, molar area with small irregular rows of small teeth (densely setous laterally, molar area with many rows of irregular teeth, with dorsal striate rounded area (Fig. 18)); maxilla (Fig. 8) with a few long setae, mala with five very long setae, palpi with two palpomere (maxilla densely setous dorsally and ventrally, mala partially divided and densely setous, palpi with three palpomeres (Fig. 19)); labium (Fig. 8): prementum with two long and two tiny setae, mentum with four pairs of long setae, lateral margins glabrous (prementum with many moderately short setae, mentum with many setae, more concentrated laterally near base (Fig. 20)). The femur of first instar larva presents a very long spatulate seta, not presented in mature larva.

During larval development, Grandi (*l.c.*) also observed considerable modifications on segment

IX, but he did not compare both larval instars in details.

According to Wheeler (1986, 1991) the neonatal larvae of *Atractocerus brasiliensis* present stemmata, but they are lost in later instars; the first instar larvae differs from later instars in body shape and proportions, in having stemmata and sometimes bearing trichobothria.

Based on the description and illustrations, the first instar larva of *Atractocerus emarginatus* is very similar to first instar larva of *Atractocerus brasiliensis*, except for segment IX with apex two-toothed in the former.

Comparing the first instar larva of *Atractocerus brasiliensis* with the description and illustrations of *Hylecoetus dermestoides*, it is possible to observe (*H. dermestoides* parenthesized): labrum rectangular with anterior margin slightly rounded (anterior margin strongly rounded); labrum with a row of eight very long setae near anterior margin and two long setae near base (eight moderately long setae near anterior margin, six basal setae); epipharynx with three pairs of long setae at fore angles and two pairs at middle (different number of short or moderately long setae); clypeus with three pairs of setae (four pairs); antennae with two antennomeres (three antennomeres); mala narrow with five long setae (mala wide, bilobed with many setae); mala with setae only at apex (setae also at margin of basal lobe); maxillary palpus with two palpomeres (three palpomeres); legs with long setae and microspines (only moderately long setae); femur with one very long and spatulate seta (trochanter and femur, each with one very long seta, not spatulate). Additionally, segment IX is very different in both genera: concave dorsally with apex narrow and upwardly directed in *Atractocerus*, and apex rounded and randomly dentate in *Hylecoetus*.

## RESUMO

*A larva de primeiro instar de Atractocerus brasiliensis (Lepetelier & Audinet-Serville, 1825) do estado de Goiás, Brasil, e criada em laboratório, é descrita e ilustrada. Esta é a segunda espécie com larva de primeiro instar conhecida para o gênero e a terceira para a família. Comparações com a larva madura dessa espécie e com as demais larvas de primeiro instar conhecidas são apresentadas. Ilustrações das larvas de primeiro instar e madura também são incluídas.*

PALAVRAS-CHAVE: Coleoptera; *Hylecoetus*; Imaturo; Larva Madura; Região; Neotropical.

## ACKNOWLEDGEMENTS

We are grateful to the following: Our friend Neusa A. da Silva for the field helping; Fundação de Amparo a Pesquisa do Estado de São Paulo (FAPESP 2012/02441-1) for financial support to FFA; Michel P. Valim (MZSP), Shaun Bennett (Plant Health & Environment Laboratory, New Zealand) and Hans Klompen (Museum of Biological Diversity, Columbus, Ohio) for helping with mite proceedings and identification; Larissa Trierweiler Pereira (Universidade Federal do Rio Grande do Sul) for comments on fungi spores of mites; Marcelo Duarte (MZSP) for accessing MZSP's facilities; and two anonymous reviewers for suggestions.

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Aceto em: 22/08/2013  
Impresso em: 30/09/2013

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**Publisher:** Museu de Zoologia da Universidade de São Paulo. Avenida Nazaré, 481, Ipiranga, CEP 04263-000, São Paulo, SP, Brasil.

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