Description of larva and pupa of the weevil *Hybolabus amazonicus* Voss (Attelabidae: Attelabinae), a leaf roller on Brazil nut (*Bertholletia excelsa*)

Sergio Antonio Vanin¹ & Daniela de Cássia Bená²

- ¹ Universidade de São Paulo (USP), Instituto de Biociências (IB-USP), Departamento de Zoologia. São Paulo, SP, Brasil. ORCID: http://orcid.org/0000-0002-2471-3991. E-mail: savanin@ib.usp.br
- ² Universidade de São Paulo (USP), Museu de Zoologia (MZUSP). São Paulo, SP, Brasil. ORCID: http://orcid.org/0000-0002-5205-2684. E-mail: danicoleoptera@gmail.com

Abstract. The last instar larva and the pupa of *Hybolabus amazonicus* Voss, 1925 are described and illustrated, based upon adults and immatures collected in the Amazonian Region (Acre and Amazonas, Brazil). The larvae live and develop inside a leaf-roll made by the female weevil. Although the species has already been reported damaging leaves of the Brazil nut (*Bertholletia excelsa* Humb. & Bonpl., Lecythidaceae) in the states of Acre and Amazonas, the preimaginal stages have not been formerly described. These are the first descriptions of larva and pupa of a species of the genus *Hybolabus* Jekel, 1860.

Key-Words. Amazonian Region; Natural history; Neotropics; Larva; Pupa.

INTRODUCTION

Some years ago, the senior author (SAV) was asked to identify adults and larvae of a leaf-rolling attelabid species associated with *Bertholettia excelsa* Humb. & Bonpl. (Lecythidaceae), the Brazilian nut. The weevil (Coleoptera: Curculionoidea) was identified as *Hybolabus amazonicus* Voss, 1925 (Fig. 1G) and two papers on the occurrence and damage caused by that species in plantations of Brazilian nut were published (Fazolin & da Silva, 1995; Garcia *et al.*, 1997).

Hybolabus Jekel, 1860 is a Neotropical genus of Attelabidae, ranging from Southern Mexico to Uruguay (Alonso-Zarazaga & Lyal, 1999). The genus was revised by Voss (1925), who also described two new species from Brazil (Voss, 1938). To date, *Hybolabus* is represented in Brazil by eleven species (Wibmer & O'Brien, 1986). In the collection of the "Museu de Zoologia da Universidade de São Paulo" we found several specimens of *Hybolabus* that could not be determined using Voss' identification key (Voss, 1925), indicating the need for a revision.

Since larvae of *Hybolabus* have never been formally described, herein we provide the descriptions to larvae and pupa of *H. amazonicus*.

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MATERIAL AND METHODS

Material

Adults, larvae and one leaf-roll of *H. amazonicus* were collected by Ricardo Andreazze (INPA, Manaus, AM, presently at University of Rio Grande do Norte, RN) and Ana M.R.S. Pamplona (Embrapa, Manaus, AM) during their research studies on insects associated with the Brazil nut, conducted in the states of Acre and Amazonas (Brazil). The species was identified using the key of Voss (1925) and by comparison with determined specimens of the MZSP collection. Adults and immatures are deposited in the entomological collection of the MZSP.

Methods

The larvae of last instar were dissected in water, under a Wild M5A stereomicroscope. Temporary slides were prepared using 100% glycerin as medium. Drawings were done with the aid of a camera lucida coupled to the stereomicroscope or to a Leitz Microscope SM-Lux. All images were processed with the computer program

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Adobe Photoshop CS3. Measurements were taken with an eyepiece micrometric scale in the stereomicroscope; values were given in millimeters. The terminology and abbreviations used in the larva and pupa descriptions follow May (1993; see also Marvaldi, 1999).

RESULTS

Hybolabus amazonicus Voss, 1925 (Figs. 1B-5E)

Description of last larval instar (Figs. 2A-4D): About length: 5 mm; head capsule width about 1.8 mm; body C-shaped, subcircular in cross section, with sparse, short and fine setae, not placed on tubercles.

Head (Figs. 2A-2C): Prognathous, moderately retracted, sclerotized, yellowish brown; head capsule slightly wider than long, lateral margins parallel-sided. Epicranial suture distinct, approximately 0.65 times as long as head capsule. Frontal suture distinct, complete, weakly curved, V-shaped. Median endocarina present, about 0.15 times as long as head capsule. One stemma present on each side (stm) (Fig. 3B). Hypopharyngeal bracon absent (Fig. 3C). Antennae exposed (Figs. 2B, 3B), two segmented; basal segment ring-like with 4 medial setae; distal segment conical and about two times longer than wide; base of antennae with one conical sensillum and one small rounded sensillum. Head capsule (Figs. 2B, 2C) with five pairs of minute posterior epicranial setae (pes1-5), almost aligned vertically; five pairs of dorsal epicranial setae (des1-5); five pairs of subequal frontal setae (fs1-5); three pairs of lateral epicranial setae (les1-3) with similar length; two pairs of ventral epicranial setae (ves), ves2 slightly longer than ves1. Epistomal and frontoclypeal sutures slightly arcuate. Clypeus (Fig. 3A) transverse, lateral margins converging anteriorly to straight anterior margin, with two pairs of subequal clypeal setae (cls1-2), and one pair of basal sensilla. Clypeolabral suture distinct. Labrum (Fig. 3A) transverse, anterior margin trilobed, with four pairs of labral setae (Irms1-4) and one pair of basal sensilla. Epipharynx (Fig. 3C) with one pair of anteriomedian setae (ams), three pairs of anterolateral setae (als1-3), and three pairs of median spines (ms) between vertical labral rods, anterior sensilla larger than posterior ones; labral rods (Ir) long, extended backwardly beyond epistoma, H-shaped, arms curved; epipharyngeal sensory spots absent. Maxillae (Figs. 3D, 3E) with cardo transverse and slender, cuneiform; stipes elongate with one seta on dorsal side and two setae on ventral side; mala rounded, slightly inward extended, without spines, with about 15 marginal dorsal spatulate setae more or less aligned along anterior outer margin (Fig. 3E); maxillary palpus three-segmented, proximal palpomere transverse, wider than distal segments, with one seta and two sensilla, medial palpomere unisetose, distal palpomere with some minute apical sensilla. Labium (Fig. 3D): labial palpi two-segmented, palpomeres elongate and with similar length, basal palpomere slightly wider than distal one, distal palpomere with ventral sensillum; prementum with one pair of setae and one pair of sensilla close to palpus insertion; submentum with three pairs of setae, posterior pair longest; ligula short, not reaching tip of labial palpi, with 2 pairs of setae and one pair of sensilla. Mandibles symmetrical (Figs. 3F, 3G), stout, apically bidentate, cutting edge dentate; dorsally with two sensilla and two aligned setae (*mds*), hind seta slightly longer than distal one.

Thorax (Figs. 2A, 4A): Prothorax narrower than meso-, and meso- narrower than metathorax. Prothorax with patches of short setae; pedal lobe with six pedal setae (*pdas1-6*); ventropleural lobe with two setae (*vpls1-2*); each side of meso- and metathorax with similar number of setae: prodorsum with two prodorsal setae (*prs*), post-dorsum with eight dorsolateral setae (*dls1-8*); pedal lobe with three setae (*pdas1-3*); ventropleural lobe with two setae (*vpls1-2*). Thoracic spiracle in intersegmental fold, bicameral (Fig. 3H), slightly oblique, air-tubes with nine annuli.

Abdomen (Figs. 2A, 4B, 4C): With eight pairs of lateral spiracles (Figs. 3I, 3J); spiracles bicameral, air-tubes backwardly directed and with nine annuli; segments I-VIII with two dorsal folds; segment IX without dorsal fold, broadly rounded in lateral view; segment X reduced, ventrally directed, anus subterminal. Segments I-VII with similar chaetotaxy (Figs. 4B, 4C): prodorsum with 12 setae on each side (prs), nine of which more or less aligned and forming a transverse row; postdorsum with 10 setae on each side (pds), nine of which more or less aligned and forming a transverse row; spiracular area asetose; dorsopleural lobe with two setae (dpls1-2); ventropleural lobe with three setae (vpls1-3); laterosternal lobe with two setae (Ists1-2); mediosternal lobe with two setae (msts1-2). Chaetotaxy of segment VIII similar to segments I-VII, but with less prodorsal setae (prs).

Alimentary canal (Fig. 4D): Lacking mycetomes; posterior ventriculus (*pov*) three coiled, with about 14 short rod-like gastric caeca (*gcc*), nearly contiguous, arranged in two more or less aligned rows, located on either side of median ventricular coil. Six cryptonephric Malpighian tubules, not thickened, arranged 3 + 3.

Description of pupa (Figs. 5A-5D): About length: 3.2 mm. Adecticous and exarate. Coloration cream-colored, with short stiff setae. Head completely covered by pronotum in dorsal view; each side with one supra-orbital seta (*sos*) and one orbital seta (*os*) located near inferior margin of eye; rostrum broad, wider than long, lateral sides slightly enlarged apicad, apex nearly straight; rostrum with one pair of short post antennal setae (*pas*) and one pair of rostral setae (*rs*). Prothorax trapezoidal, lateral margins sinuous; anterolateral margin with a row of nine setae (*Js*) on each side; posterolateral margin with a row of 10 setae (*pls*) on each side. Mesothorax with two transverse rows of six anterior and 14 posterior erect ter-

gal setae, rows interrupted at middle. Metathorax with two transverse rows of four anterior and 12 posterior tergal setae, rows interrupted at middle. Each leg with two femoral setae (*fes1-2*). Pterothecae extending up to third ventrite. Abdomen: segments I-VII with 20 tergal setae forming a transverse row located close to posterior margin; segments narrowed to posteriad; each side of segment VIII with a lateroventral seta and an acute posteriolateral spine-like process (not articulated, pseudocerci or urogomphi); segment IX small. Segment X with gonotheca not divided in males (Fig. 5D), with two oval bulges in females (Fig. 5E). Seven annular spiracles placed at lateral sides of segments I to VII, visible at lateral view.

Material examined: BRAZIL: Acre, BR 317, km 75, III.1994 EMBRAPA, 6 adults, 1 leaf-roll (MZSP). Amazonas,



Figure 1. Bertholettia excelsa Humb. & Bonpl. (A) tree; (B-C) female of *H. amazonicus* cutting leaf to make nidus; (D) nidus or leaf roll; (E) branches with leaves and flowers; (F) open fruit with seeds. (G) adult *H. amazonicus* (dorsal view), from Acre.

Manaus, IV.1994, CPAA A. Pamplona col., 7 adults; same locality, Rod. AM 010, km 24.6, 06.VIII.1995, EMBRAPA CPAA Projeto Shift, Andreazze, R. col., 29 larvae (two of them dissected, mounted on 3 slides), 3 pupae (MZSP).

Host Plant: *Bertholetia excelsa* Humb. & Bonpl. ("castanheira-do-brasil", "castanha-do-pará", Brazil nut, Lecythidaceae), a plant native to the Amazonian Region, is one of the most important non-timber producing tree



Figure 2. *Hybolabus amazonicus Voss,* third instar larva. (A) habitus of larva (lateral view); (B) head capsule (frontal view); (C) head capsule (posterior view). Legends: *cls* = clypeal setae; *des* = dorsal epicranial setae; *fs* = frontal setae; *les* = lateral epicranial setae; *pes* = posterior epicranial setae; *vs* = ventral epicranial setae.

species for sustainable extractive exploration of almonds (Fig. 1F), used raw or roasted as human food, or for obtaining oils used in aviation and in the manufacture of cosmetics (Garcia *et al.*, 1997). The tree reaches 50 m in height (Fig. 1A), the leaves are simple and glabrous (Fig. 1E), reaching 25 to 35 mm long (Lorenzi, 2000). Occurs mainly in Brazil, in the states of Amazonas, Acre, Rondônia, Pará and north of Mato Grosso, Tocantins and



Figure 3. *Hybolabus amazonicus Voss,* third instar larva. (A) clypeus and labrum; (B) antenna and stemma; (C) epipharynx; (D) maxillo-labial complex (ventral view); (E) maxillo-labial complex (dorsal view); (F) mandible (ventral view); (G) mandible (dorsal view); (H) prothoracic spiracle; (I) abdominal spiracle I; (J) abdominal spiracle VIII. Legends: *als* = anterolateral setae; *ams* = anteromedian setae; *cls* = clypeal setae; *mds* = mandibular setae; *lr* = labral rods; *lrms* = labral setae.

Maranhão, but there are also a few records from Bolivia, Peru, Venezuela, Guyana, Suriname and French Guyana (Mori & Prance, 1990).

Biological notes on natural history: Fazolin & da Silva (1995) and Garcia *et al.* (1997) reported adults and larvae of *H. amazonicus* in the states of Acre and Amazonas, respectively, damaging old or new leaves of *Bertholletia excels.* Adults scrape the upper epidermis of the leaves and the continuous scraping activity by weevils can cause tears in the leaves. Females cut out the leaves near the apical third to the midrib or more (Figs. 1B, 1C) and roll the cut part forming a cigar-like structure, called nidus (Fig. 1D), inside which they lay from 1 to 20 eggs. Most leaf-rolls of *H. amazonicus* fall to the ground after being

built and do not remain attached to the tree. The larvae feed on the leaf blade and use the leaf-roll as a shelter to turn into pupae. One to 14 adults were observed emerging from a single nidus. When the infestation is high, these weevils can cause serious damages, more pronounced in younger than older trees, due to the smaller foliage mass (Garcia *et al.*, 1997).

Garcia *et al.* (1997) also reported the occurrence of another species of *Hybolabus*, *H. columbinus* (Erichson, 1848), also damaging leaves of the Brazil nut in the states of Acre and Amazonas. It should be noted that *H. columbinus* was previously only known from Colombia and Guyana (Wibmer & O'Brien, 1986) and Garcia *et al.* (1997) published the first record of that species for Brazil. *Hybolabus amazonicus* and *H. columbinus* are very similar



Figure 4. *Hybolabus amazonicus Voss,* third instar larva. (A) thoracic segments (lateral view); (B) abdominal segments I-III (lateral view); (C) abdominal segments VII-X (lateral view); (D) alimentary canal. Legends: Ab = abdominal segment; anv = anterior ventriculus; dls = dorsolateral setae; dpls = dorsopleural setae; gcc = gastric setae; lsts = laterosternal setae; Mgt = Malphigian tubules; msts = mediosternal setae; phx = pharynx; pdas = pedal lobe setae; pds = prodorsal setae; pov = posterior ventriculus; Th = thoracic segment; vpls = ventropleural setae.



Figure 5. *Hybolabus amazonicus Voss,* pupa. Habitus: (A) entral view; (B) orsal view; (C) lateral view; (D) ventral abdominal tip, male; (E) ventral abdominal tip, female. Legends: *ds* = discal setae; *fs* = femoral setae; *ls* = lateral setae; *os* = orbital setae; *pas* = postantennal setae; *pls* = posterolateral setae; *pc* = pseudocerci; *rs* = rostral setae; *sas* = superapical setae; *sos* = superorbital setae; *SV* = sternite; *Th* = thoracic tergite; *T* = abdominal tergite.

and share the reddish-brown antennae. They are easily distinguished by their body color: *H. amazonicus* has an entirely black body, whereas the elytra of *H. columbinus* are dark blue, and the venter is reddish-brown (Voss, 1925).

DISCUSSION

Bionomical knowledge about host-plant associations of Hybolabus species is scant and usually restricted to a few plants species that have no economic importance, and the association of H. amazonicus with the Brazil nut is an exception. Bondar (1937, 1947) reported the behavior of four species of Hybolabus, all from the state of Bahia (Brazil), which develop inside leaf-rolls of different families of plants: H. ater (Olivier, 1789) from Inga edulis Martius ("ingá-cipó", Fabaceae), H. bryanti Voss, 1938 from Arapatiella psilophylla (Harms) Cowan ("faveca", Fabaceae), an endemic plant of South Bahia, H. collaris Voss, 1938 from Cariana sp. ("jequitibá", Lecythidaceae), and H. azuripennis Voss, 1925 from Terminalia catappa L. (Indian almond, sea almond, "amendoeira", "chapéu de sol", Combretaceae), an exotic plant introduced in the Americas and extensively used in Brazilian urban landscaping.

These are the first descriptions of larva and pupa published for a species of the genus Hybolabus and, as far as we know, the first described immature of a Brazilian attelabid species. The immatures agree very well to the descriptions and illustrations of other larvae of Attelabinae published by Emden (1938), May (1993), Lee & Morimoto (1988), Riedel (2014), and Fenili (1952); the latter also describes the pupa of Attelabus nitens (Scopoli, 1763). The main differences are related to chaetotaxy, the presence or absence of some setae on the head capsule, epipharynx, maxillary mala, thorax and abdomen. Other distinguishing features refer to the number of stemmata and to the presence or absence of an obtuse spine near the middle of the inner margin of the maxillary mala. The labral rods are short or long, subparallel or curved inwards at middle in all previously described Attelabinae, but it is H-shaped in H. amazonicus. Further studies are needed to discover and describe the immatures of other species of Hybolabus and of other genera of Attelabinae to allow a more detailed comparison.

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