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Article



A new species of *Bucculatrix* Zeller (Lepidoptera: Bucculatricidae) associated with *Baccharis salicifolia* (Asteraceae) in northern Chile

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Abstract

Male, female and immature stages of *Bucculatrix mirnae* **sp. n.**, from the Azapa Valley, coastal desert of northern Chile, are described and illustrated, under optical and scanning electron microscopy. The immature stages are associated with the shrub *Baccharis salicifolia* (Ruiz & Pav.) Pers. (Asteraceae). The larva is hypermetamorphic, having five instars and two feeding habits. The first, second and third instars are leaf miners, and the fourth and fifth instars are leaf skeletonizers. This is the first species of Bucculatricidae described from Chile.

Key words: leaf-mining moths, Neotropical region, hypermetamorphosis, molting cocoon, pupal cocoon

Introduction

Bucculatricidae are plant-mining Lepidoptera, with nearly 250 species known around the world (Davis & Robinson 1998). Their larvae undergo a hypermetamorphic development, most being leaf miners in early instars and either leaf skeletonizers, stem borers or gall-makers in later ones (Friend 1927; Braun 1963; Davis & Robinson 1998; Davis *et al.* 2002; Kobayashi *et al.* 2009).

In the Nearctic region they are represented only by the genus *Bucculatrix* Zeller, 1848, for which more than 100 species are recognized in North America (Braun 1963; Rubinoff & Osborne 1997). On the other hand, only seven species of *Bucculatrix* were listed by Davis and Miller (1984) for the Neotropical region. Among them is the cotton pest *Bucculatrix thurberiella* Busck, 1914, whose type locality is located in the United States but that has a distributional range extending to many South American countries. Of the six remaining Neotropical species, one has its type locality in Bermuda, three in Mexico and two in the Virgin Islands. Subsequently, Davis *et al.* (2002) described two other Neotropical species for this genus, one from Belize and another from the Galapagos Islands, both with host plants in the genus *Cordia* (Boraginaceae). They provided a detailed description of the immature stages of *Bucculatrix caribbea* Davis and Landry, illustrated with scanning electron microscopy.

Chilean micro-moths, especially those of the coastal northern desert, are poorly known, mostly because of the poor collecting efforts in the past. This region, in conjunction with the coastal desert of southern Peru, is very interesting for taxonomic studies regarding many insect groups, and some endemic taxa have been described (e.g., Porter 1985; Vargas & Landry 2005; Vargas & Parra 2005; Howden 2008). Records of Bucculatricidae were previously unknown from Chile. As part of a survey of indigenous Lepidoptera associated with native vegetation in the Azapa Valley, Arica Province in northern Chile, adults of an undescribed species of *Bucculatrix* were obtained from larvae collected on the shrub *Baccharis salicifolia* (Ruiz & Pav.) Pers. (Asteraceae). Herein, we describe and illustrate all the life stages of this new species, and provide a preliminary characterization of its life history.

Materials and methods

Specimens used in the study were reared in small plastic vials at room temperature, in the entomology laboratory of the Facultad de Ciencias Agronómicas, Universidad de Tarapacá, Arica city, from larvae collected on B. salicifolia in the Azapa Valley, northern Chile. They were fixed with Dietrich's fluid and preserved in 75% ethanol. At least five specimens were used for the descriptions of each life stage or instar. For observations on gross morphology, the specimens were cleared in a 10% potassium hydroxide solution (KOH) and slide-mounted in either glycerin jelly or Canada balsam. Observations were performed with the aid of a Leica M125 stereomicroscope, where structures selected to be drawn were photographed with an attached Sony® DSC-H10 digital camera. Vectorized line drawings were then made with the software CorelDraw[®] X4, using the corresponding digital images as a guide. Color drawings were similarly accomplished adopting the stippling technique under high magnification (greater than 6,400%) of the computer screen, using digital images taken from live specimens as references. Measurements were made with an attached ocular micrometer; unless noted, values are presented as mean \pm standard deviation. For scanning electron microscope examination, the specimens were dehydrated in a Bal-tec CPD030 critical-point dryer, mounted with double-sided tape on metal stubs, and coated with gold in a Bal-tec SCD050 sputter coater. They were then examined and photographed in a JEOL JSM5800 scanning electron microscope at the Centro de Microscopia Eletrônica (CME) of the Federal University of Rio Grande do Sul (UFRGS), Porto Alegre, state of Rio Grande do Sul. Brazil.

The material examined is deposited in the collections listed below:

- **MNNC** Museo Nacional de Historia Natural de Santiago, Santiago, Chile.
- IDEA Colección Entomológica de la Universidad de Tarapacá, Arica, Chile.
- **LMCI** Laboratório de Morfologia e Comportamento de Insetos, Universidade Federal do Rio Grande do Sul, Porto Alegre, Brazil.

Results

Bucculatrix mirnae Vargas & Moreira, new species Fig. (1–13)

Type material. Data are listed as they are found in the labels; dates correspond to adult emergences. HOLOTYPE, σ , CHILE: Azapa, Arica, Chile, July 2009, H.A. Vargas coll., reared from larva on *Baccharis salicifolia* (MNNC). PARATYPES: 2 $\varphi \varphi$, same data as holotype (MNNC); 1 σ Azapa, Arica, Chile, August 2008, H.A Vargas coll., reared from larva on *Baccharis salicifolia*, July 2008 (MNNC); 3 $\sigma \sigma$, 3 $\varphi \varphi$ Azapa, Arica, Chile, September 2008, H.A Vargas coll., reared from larva on *Baccharis salicifolia*, July 2008 (MNNC); 3 $\sigma \sigma$, 3 $\varphi \varphi$ Azapa, Arica, Chile, September 2008, H.A Vargas coll., reared from larva on *Baccharis salicifolia*, August 2008 (MNNC); 1 σ , 3 $\varphi \varphi$ Azapa, Arica, Chile, July 2010, H.A Vargas coll., reared from larva on *Baccharis salicifolia*, June 2010 (IDEA); 1 σ , 3 $\varphi \varphi$ Azapa, Arica, Chile, August 2011, H.A Vargas coll., reared from larva on *Baccharis salicifolia*, July 2011 (IDEA); 1 σ Azapa, Arica, Chile, September 2011, H.A Vargas coll., reared from larva on *Baccharis salicifolia*, August 2011 (IDEA); 1 σ Azapa, Arica, Chile, September 2011, H.A Vargas coll., reared from larva on *Baccharis salicifolia*, August 2011 (IDEA); 1 σ Azapa, Arica, Chile, September 2011, H.A Vargas coll., reared from larva on *Baccharis salicifolia*, August 2011 (IDEA) Other specimens examined. Immature stages collected by the senior author at the type locality on the same host plant, August 2010, fixed in Dietrich's fluid and stored in 70% ethanol. All deposited in the LMCI collection, under accession numbers as follows: 5 eggs (LMCI 161-1), 6 first-instar larvae (LMCI 161-2), 26 second-instar larvae (LCMI 161-3), 15 third-instar larvae (LMCI 161-4), 12 fourth-instar larvae (LMCI 161-5), 13 fifth-instar larvae (LMCI 161-6), 6 pupae (LMCI 161-7).

Diagnosis. The male genitalia of *B. mirnae* resemble those of *B. dominatrix* Rubinoff & Osborne, described from Florida, United States and with immature stages that are associated with *Baccharis pilularis* (de Candolle) (Asteraceae) (Rubinoff & Osborne 1997). In the adult male, the unique, broad juxta of *B. mirnae* clearly separates the two species. The female of *B. mirnae* can also be easily distinguished from that of *dominatrix* by the distinct, broader area of the signum, composed of multiple small, spine-like elements, the corresponding lines being distally interspersed with single, larger spines.

Male adult (Figs. 1a–2)

Head. Frontoclypeus (Figs. 2a, 3a) with creamy white, broad scales with blunt apex. Vertex with narrow, slender scales, projected in all directions (Fig. 2a), mostly creamy white, some grayish brown in middle. Antennae

(Fig. 1a) filiform, nearly 2/3 of forewing length; scape dorso-ventrally expanded (Figs. 2b, 3a), covering dorsal area of compound eye, creamy white, mostly covered by broad scales, with some long scales projected ventrally until near middle of compound eye; pedicel annular, short, creamy white; first flagellomere with dorsal notch (Fig. 2c), each flagellomere with single row of scales, alternating creamy white with grayish brown. Proboscis (Figs. 2d, 3a) without scales, short, nearly length of prothoracic coxa, with about 10 pairs of distally located styloconic sensilla. Labial palpus (Figs. 2e, 3a) short, apex reaching near base of proboscis, with creamy white scales on distal third, remaining portion without scales.



FIGURE 1. Bucculatrix mirnae sp. n. adults: (A) male; (B) female. Scale bars = 1 mm.



FIGURE 2. Scanning electron micrographs of *Bucculatrix mirnae* **sp. n.** adult head and thorax: (A) female head, antero-lateral view; (B) antennal cap (indicated by asterisk), ventro-posterior view; (C) basal section of male antenna, lateral view, showing notch on the first flagellomere (indicated by solid arrow); (D) partially coiled proboscis, anterior view, with styloconic sensilla (indicated by solid arrow); (E) labial palpus, ventro-posterior view; (F) hind leg, anterior view (open and solid arrows indicate basal and distal spur pairs respectively). Scale bars = 100, 50, 100, 50, 20, 250 µm, respectively.



FIGURE 3. *Bucculatrix mirnae* **sp. n.** adult morphology: (A) female head, anterior view; (B) fore- and hind wing venation; (C) male genitalia, ventral view (aedeagus removed); (D) aedeagus, lateral view; (E) female genitalia, ventral view. Scale bars = $300, 500, 100, 100, 100 \mu m$, respectively.

Thorax. Grayish brown dorsally, some scales with dark gray apex; lateral surface covered by creamy-white scales; ventral arm of tegula with group of creamy-white, piliform scales reaching anal margin of hind wing. Foreleg with creamy-white and dark brown scales mixed; epiphysis arising near base of foretibia. Midleg with color pattern similar to that of foreleg, with one pair of asymmetrical spurs at apex. Hindleg creamy-white with sparse grayish-brown scales on tarsus; tibia (Fig. 2f) with long hair-like scales distally projected, and with two pairs of

asymmetrical spurs, one arising near base, another at apex. Forewing (Figs. 1, 3b) (length: 2.78–3.33 mm) lanceolate, dorsally covered by light-reddish-brown scales, some with dark-grayish-brown apex, mostly concentrated at apex; fringe and ventral surface creamy white. Venation (Fig. 3b): Sc broadened basally, ending basal to midpoint of wing margin; R with five branches; basal half of R1 stalked with margin of discal cell; R5 stalked with M1; M2, CuA1 and CuP as a fold, poorly differentiated; M2 and CuA1 almost parallel, arising separately near apex of discal cell; CuP reaching wing margin near apex of CuA1; 1A+2A well developed, reaching wing margin near its midpoint. Hindwing dorsal and ventral surfaces and fringe creamy white. Venation (Fig. 3b): Sc broadened basally, expanded near basal third; Rs not reaching wing apex; M1 and M2 well separated; CuA ending near midpoint of wing margin; A as a fold ending near basal third of wing margin. Abdomen creamy white.

Male genitalia (Figs. 3c–d, 4a–c). Uncus absent; tegumen broad, not separated from saccus; socii well developed, divergent, with setae at apex; gnathos absent; juxta well developed, broad at base, narrow at apex; valva elongated, with costal margin slightly excavated distally, medial surface with hair-like setae (Figs. 4a–b) near midpoint, and peg-like setae (Figs. 4a, c) near apex; phallus elongated, narrow and straight; vesica without cornuti.

Female adult (Figs. 1b, 3a, 3e, 4d–g)

Similar to male, but antenna with first flagellomere without notch (Fig. 3a), hindwing with two frenular setae.

Female genitalia (Figs. 3e, 4d–g). Tergum VIII with spine-like microtrichia (Figs. 4d–e); papillae anales with spine-like microtrichia interspersed with long hair-like setae; ductus bursae cylindrical, narrow, lightly sclerotized; corpus bursae spherical (Figs. 3e, 4g), with signum formed by many small, spine-like, aligned elements that radiate from its proximal portion; distal lines interspersed with single, larger spines of similar shape; lamella postvaginalis as band laterally, continuous with tergum 8; anterior apophyses absent; posterior apophyses short.



FIGURE 4. Scanning electron and light microscopy micrographs of *Bucculatrix mirnae* **sp. n.** genitalia: (A) male valve, mesal view; (B) male valva basal setae (enlarged view of left square in A); (C) male valve, distal sensilla (enlarged view of right rectangle in A); (D) terminal portion of female abdomen, lateral view; (E) microtrichia on 8th segment of female (area marked with asterisk in D, in detail); (F) detail of setae and microtrichia of female anal papillae (enlarged view of square in D); (G) signum of female corpus bursae. Scale bars = 50, 10, 5, 50, 5, 10, 30 μ m, respectively.



FIGURE 5. Habitat, larval host plant and life history of *Bucculatrix mirnae* **sp. n.** (A) type locality, Azapa Valley, Arica municipality, Chile, showing scattered larval host plants, *Baccharis salicifolia*; (B) plant of *B. salicifolia* in close view; (C) *B. salicifolia* leaves in detail (enlarged view of rectangle area in B); (D) egg on adaxial surface of leaf; (E) mine on leaf; (F) egg chorion remaining at the beginning of a mine (indicated by open arrow in E); (G) detail of orifice left by third-instar larva at the mine end (indicated by closed arrow in E); (H) molting cocoon on leaf, showing larval exuvia by transparency; (I) leaf damage caused by fifth- instar larva; (J) pupal cocoon, lateral view, with anterior portion indicated by closed seta). Scale bars = 0.5, 5, 1, 1, 0.5, 1, 1 mm, from D to J, respectively.

Immature stages

Egg (Figs. 5d, 5f, 6). Flat, slightly ellipsoid; chorion translucent, larva may be observed before emergence, ornamented with irregular, poorly defined cells (Fig. 6a); aeropyles on vertex of cells near margin of chorion (Figs. 6c, 6d); micropylar area (Fig. 6b) finely reticulated, with smooth central region.

Larva. Hypermetamorphic, with five instars.

First instar (Figs. 8a, 8d, 9). Body length = 0.863 ± 0.061 mm; head capsule width = 0.067 ± 0.006 mm; n = 5. Prognathous, body cylindrical, apodal, setae extremely reduced. Head yellowish brown, smooth; frontoclypeus triangular (Fig. 8a), F1 present; stemmata absent; antennae greatly reduced; chewing mouthparts (Fig. 9a), labrum (Fig. 9c) bilobed with four small setae on external surface, epipharynx with four spines, mandible (Fig. 9b) well developed with four cusps along distal margin and one small seta basally on external surface, maxilla (Fig. 9e) with palpus and galea well developed, spinneret (Fig. 9d) tubular, long, nearly 1.5 x length of maxillary palpus, with multi-lobed apex, labial palpi absent. Thorax and abdomen white; prothoracic shield (Fig. 8a) as a fine transverse stripe near posterior margin of segment, with two antero-lateral expansions; circular spiracles (Fig. 9f) laterally on T1, A1–8.

Second instar. Similar in morphology to first instar, but with dorsal shield of prothorax as in third instar, and also larger in size (head capsule width = 0.121+0.006 mm, n = 6).



FIGURE 6. Scanning electron micrographs of *Bucculatrix mirnae* **sp. n.** egg chorion: (A) general view on adaxial surface of leaf; (B) micropilar area (location indicated by seta in A); (C) close view of marginal area, showing aeropiles (open setae) and cement substance (asterisk); (D) aeropile in detail. Scale bars = 100, 5, 10, 2 μ m, respectively.

Third instar (Figs. 8b, 8e, 10). Body length = 3.135 ± 0.678 mm; head capsule width = 0.225 ± 0.005 mm; n = 7. Prognathous, body cylindrical, translucent setae well developed, thoracic legs and abdominal prolegs present (Fig. 8e). Head pale brown, smooth; five pairs of stemmata (Fig. 10c); chewing mouthparts (Fig. 10a); labrum (Fig. 10d) bilobed with four setae on external surface; epipharynx with four spines and expanded laterally; mandible (Fig. 10b) with four cusps along distal margin and one seta basally on external surface; maxilla (Fig. 10f) with palpus and galea well developed; spinneret tubular, length similar to that of maxillary palpus; labial palpi (Fig. 10e) well developed, nearly 0.5 x length of spinneret, with two setae. Thorax and abdomen light yellowish, covered with small spine-like microtrichia; prothorax with dorsal shield (Fig. 8b) well developed, dark brown, with three forward-projecting arms, median arm nearly twice length of lateral arms, thoracic legs with simple pretarsal claw (Fig. 10g), not modified as in some other *Bucculatrix* species; tarsus on meso- and metathorax with two spatulate setae (Fig. 10h) at apex; short prolegs on A3–6 and A10, each with 2 crochets (Fig. 10i) at apex; circular spiracles laterally on T1, A1–8.

Fourth instar. Similar in morphology to fifth instar, but smaller in size (head capsule width = 0.287 ± 0.013 mm, n = 6).

Fifth instar (Figs. 8c, 8f, 11). Body length = 5.585 ± 0.136 mm; head capsule width = 0.376 ± 0.008 mm; n = 6. Hypognathous, body cylindrical, thoracic legs and abdominal prolegs well developed (Figs. 8c, 8f). Head pale brown (0.32-0.34 mm wide); five pairs of stemmata (Fig. 11c); chewing mouthparts (Fig. 11a); labrum (Figs. 11a, 11b) bilobed with four setae on external surface; epipharynx expanded laterally, partly concealing mandibles, with four spines; mandible with four cusps along distal margin and one seta basally on external surface; maxilla (Fig.

11d) with palpus and galea well developed; spinneret (Fig. 11e) tubular, length similar to that of maxillary palpus; labial palpi (Fig. 11e) well developed, nearly 0.5 x length of spinneret (Fig. 11e), with two setae. Thorax and abdomen olive green; setae long, black, with white circular area surrounding basis of setae (Fig. 8c, 8f); thoracic legs well developed, with simple pretarsal claw (Fig. 11h) at apex, not modified as in some *Bucculatrix* species; tarsus on meso- and metathorax with two spatulate setae (Fig. 11i) at apex; prolegs (Fig. 11j) on A3–6 and A10, tubular, elongated, with 3–4 crochets at apex; circular spiracles (Figs. 11f–g) laterally on T1, A1–8.



FIGURE 7. Scanning electron micrograph of *Bucculatrix mirnae* **sp. n.** larval leaf damage and cocoons: (A) feeding scars on adaxial surface of leaf lamina; (B) molting cocoon weaving pattern in close view; (C) weaving pattern of pupal cocoon anterior portion, in dorsal view (asterisk indicates anterior third, where ridges are interrupted); (D) weaving pattern of pupal cocoon posterior portion, in lateral view. Scale bars = 100, 10, 200, 200 μ m, respectively.

Chaetotaxy of fifth instar (Figs. 11c, 11f). Head with 14 pairs of setae: F group unisetose; C group bisetose, C1 small; AF group bisetose; A group trisetose; P group unisetose; L group unisetose; S group unisetose with S1 and S3 absent, SS group trisetose; three pairs of microsetae: CD1, CD2, MG1; and four pairs of pores: AFa, Pa, Sb, MGa. Labrum with four pairs of macrosetae: LA1, LA3, M1, M3; and one pair of pores: Ma. Prothorax with 12 pairs of macrosetae; XD group bisetose, XD1 anterodorsal to XD2; D group bisetose, D2 more than two x length of D1; SD group bisetose, SD1 closer to XD2; L group trisetose; SV group bisetose; V group not found; circular spiracle posterior to L3. Meso-metathorax with D group bisetose, D1 greatly reduced, dorsal to D2; SD and L group bisetose; SV group unisetose; V group unisetose; U group bisetose, D1 similar to D2, SD2 greatly reduced; L group trisetose; SV and V group unisetose; L3, SV1 and V1 in straight line; circular spiracle between SD1 and L2. A3-6 similar to segments A1, 2, 7, but SV group trisetose, laterally on proleg; V1 on medial face of proleg. A8 similar to segments A1, 2, 3, but with circular spiracle between SD1 and L1. A9 with D2 group bisetose; SD, L, SV and V group unisetose. A10 with D and SD group bisetose; inne setae on proleg: PP1, L1-3, SV1-4 and V1.

Pupa (Figs. 12, 13). Body length = 3.253 ± 0.093 mm; n = 6. Pale brown; head with frontal process (Figs. 12a–b, 13a–c) utilized for cutting the cocoon at adult emergence; antennae narrow, long, apex near forewing apex; proboscis short, not reaching apex of prothoracic legs; metathoracic legs reaching beyond forewing apex, concealing genital scars; prothorax a narrow transverse band between head and mesothorax; prothoracic spiracle partly concealed (Fig. 13d); hindwings concealed by forewings; abdominal segments A3–7 with transverse band of spines (Figs. 12b–c, 13f), near anterior margin of each segment; circular spiracle (Figs. 13e, g) laterally on A2–7, spiracle on A8 greatly reduced; A10 with two lateral processes (Fig. 13h) each with acute apex, and with two small spines dorsally (Fig. 13i).

Molting cocoon (Figs. 5h, 7b). Circular or ellipsoid, flat, smooth, pale whitish; spun by third and fourth instars previous to molting.

Pupal cocoon (Figs. 5j, 7c–d). Ellipsoid, long, with both ends rounded, with longitudinal ridge pattern broken near anterior third; surrounded by ellipsoidal ring formed by long silk filaments. Spun by fifth-instar larva prior to pupation.



FIGURE 8. Schematic representation of variation in *Bucculatrix mirnae* **sp. n.** larval color pattern: (A-C) head and prothorax, dorsal view (first, third and fifth instars, respectively); (D-F) body, lateral view (first, third and fifth instars, respectively). Scale bars = 25, 100, 200 μ m and 0.1, 0.25, 0.5 mm, from A to C and from D to F, respectively.

Etymology. *Bucculatrix mirnae* is named in honor of our friend, Prof. Dr. Mirna Martins Casagrande, Departamento de Zoologia, Universidade Federal do Paraná, Curitiba, Brazil, an outstanding Brazilian lepidopterist, for her great contribution to the development of lepidopterology in the Neotropics.

Host plant (Fig. 5b, 5c). The only host plant known for the immature stages of *B. mirnae* is the shrub *Baccharis salicifolia* (Ruiz & Pav.) Pers. (Asteraceae).

Distribution. *Bucculatrix mirnae* is known only from the type locality, the Azapa Valley (Fig. 5a), Atacama Desert, northern Chile.



FIGURE 9. Scanning electron micrographs of *Bucculatrix mirnae* **sp. n.** first larval instar. (A) head, antero-lateral view; (B) mandible and antenna, antero-lateral view; (C) labrum, antero-ventral view; (D) spinneret, antero-lateral view; (E) maxilla; (F) prothoracic spiracle. Scale bars = $20, 5, 5, 5, 5, 2 \mu m$, respectively.



FIGURE 10. Scanning electron micrographs of *Bucculatrix mirnae* **sp. n.** third larval instar. (A) head, lateral view; (B) mandible and antenna, lateral view; (C) stemmata, antero-lateral view; (D) labrum, ventral view; (E) spinneret, lateral view; (F) maxilla, anterior view; (G) foreleg, posterior view; (H) metathoracic tarsus, posterior view; (I) fourth abdominal proleg, ventral view. Scale bars = 50, 10, 10, 10, 10, 10, 10, 20 μ m, respectively.



FIGURE 11. Scanning electron micrographs of *Bucculatrix mirnae* **sp. n.** fifth larval instar. (A) head, anterior view; (B) labrum and antenna, antero-lateral view; (C) stemmata, lateral view; (D) maxilla, anterior view; (E) spinneret, lateral view; (F) prothoracic spiracle; (G) third abdominal spiracle; (H) prothoracic leg, posterior view; (I) metathoracic tarsus, postero-dorsal view; (J) fifth abdominal proleg, lateral view. Scale bars = 100, 20, 25, 20, 20, 10, 10, 20, 10, 50 μ m, respectively.

Life history. Eggs (Fig. 5d) are deposited mostly on the adaxial surface of leaves, adhered by a cement substance that typically seems to surround the egg. Larvae are leaf miners during the first three instars (Fig. 5e). Hatching occurs through the surface of the egg adhered to the leaf (Fig. 5f); the first-instar larva enters progressively into the leaf, loading feces into the chorion, because initially the posterior part of the body remains within the chorion. The first two molts occur in the mine. When the third instar completes its feeding, the larva exits the mine (Fig. 5g) and spins the first molting cocoon (Fig. 5h), mostly on a leaf. Fourth and fifth instars are external feeders on the same plant, skeletonizing leaves (Fig. 5i, 7a). The second molting cocoon, which is constructed by the fourth-instar larva, is also in most cases adhered to a leaf. Pupation occurs inside the pupal cocoon (Fig. 5j), which is constructed by the fifth-instar larva, mostly on either twigs or dry leaves of the host plant. During adult emergence, the anterior portion of the pupal cocoon is split by the frontal process of the pupa. In most cases, after adult emergence, the anterior half of the pupal exuvium (head and thorax) is found protruding to the outside, while the posterior half remains in the pupal cocoon.



FIGURE 12. Bucculatrix mirnae sp. n. pupal morphology, in ventral (A), lateral (B) and dorsal (C) views. Scale bar = 250 µm.

Discussion

As already mentioned, the male genitalia of *B. mirnae* resemble those of *B. dominatrix*. This species was included by Rubinoff and Osborne (1997) in Braun's (1963) section II of *Bucculatrix*, which comprises the majority of the North American species associated with Asteraceae. Thus, based on comparison of genital morphology and hosts, *B. mirnae* would also be included in this section on a preliminary basis. This is the first species of Bucculatricidae described from Chile. Future fieldwork in neighbouring Neotropical areas may reveal other undescribed species for the genus that are more closely related to *B. mirnae*, and in that case the current taxonomic placement should be reevaluated.

Another Neotropical species of Bucculatricidae with immature stages described in detail is *Bucculatrix carib*bea Davis & Landry (Davis et al. 2002). This species, however, is not closely related to *B. mirnae*, judging by the conspicuous differences in the morphology of the male and female genitalia. Moreover, the host plant of *B. caribbea* is *Cordia sebestena* L. (Boraginaceae).

The gross morphologies of the molting and pupal cocoons of *B. mirnae* pupae resemble those of *B. caribbea*. However, the larval chaetotaxy of *B. mirnae* is more complete in comparison to that of *B. caribbea*. Some morphological features also differ between the two species, for example regarding the prothoracic pretarsal claw, which is highly modified in *B. caribbea*. This species also has more crochets on the prolegs than does *B. mirnae* (Davis *et al.* 2002).

Asteraceae is one of the most common host-plant families reported for larvae of Bucculatricidae (Braun 1963; Rubinoff & Osborne 1997). The association of *B. mirnae* larvae with *B. salicifolia* agrees with this pattern. Valleys located in the coastal Atacama Desert, including that of Azapa, where the type locality of *B. mirnae* is located, are under strong pressure from agricultural expansion. As a consequence, the unique native vegetation of these valleys, including populations of *B. salicifolia*, has been progressively removed to make way for new farm fields. By making formally available the name of a new species of *Bucculatrix* from that region, we expect also to contribute indirectly to justify the establishment of appropriate and much-needed conservation policies.



FIGURE 13. Scanning electron micrographs of *Bucculatrix mirnae* **sp. n.** pupa. (A) head, ventral view; (B) head, lateral view; (C) cocoon cutter in detail, lateral view; (D) prothoracic spiracle, lateral view; (E) abdominal terga, general view; (F) tergal spines in detail, dorsal view (indicated by seta in E); (G) fifth abdominal spiracle; (H) terminal portion of abdomen, ventral view; (I) tergal spines, dorso-lateral view. Scale bars = 100, 100, 50, 20, 100, 50, 10, 100, 20 μ m, respectively.

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