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***Conotrachelus*<sup>1</sup> Species of Agricultural and Quarantine Importance for Mexico**

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**Abstract.** The genus *Conotrachelus* Dejean is a large, diverse group in which some pest species are associated with fruit with high commercial value. Nevertheless, there are few morphological studies focused on differentiating the species. In Mexico, the species *Conotrachelus perseae* Barber, *Conotrachelus aguacatae* Barber, *Conotrachelus dimidiatus* Champion, *Conotrachelus copalensis* Salas and Romero, *Conotrachelus crataegi* Walsh, and *Conotrachelus eburneus* Champion cause damage and loss in fruit of economic importance. For this reason, this paper describes the most distinctive morphological characteristics of the species, considering easily observed external traits, as well as the aedeagus and Sternite VIII, and provides an illustrated taxonomic key.

**Resumen.** El género *Conotrachelus* es un grupo grande y diverso, donde algunas especies plaga se encuentran asociadas a frutales de alto valor comercial. A pesar de esto existen pocos trabajos morfológicos dirigidos a la diferenciación de estas especies. En México se presentan las especies *Conotrachelus perseae* Barber, *Conotrachelus aguacatae* Barber, *Conotrachelus dimidiatus* Champion, *Conotrachelus copalensis* Salas and Romero, *Conotrachelus crataegi* Walsh, y *Conotrachelus eburneus* Champion causando daños económicos en los cultivos de aguacate, guayaba, tejocote, y nanche rojo, respectivamente. Debido a esto, se presenta una diagnosis de las características morfológicas más distintivas de estas especies, para lo cual se tomaron en cuenta caracteres externos de fácil observación, además del edeago y el esternito VIII. Adicionalmente se ofrece una clave taxonómica ilustrada.

### Introduction

According to Morrone (2014), the superfamily Curculionoidea includes approximately 62,000 species and 5,800 genera in seven families. In Mexico, 603 genera and 3,594 species have been described. About 85% of the species of weevils (family Curculionidae) in Mexico are classified into the subfamilies Baridinae, Brachycerinae, Cossoninae, Curculioninae, Cyclominae, Dryophthorinae, Entiminae, Molytinae, Platypodinae, and Scolytinae.

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The genus *Conotrachelus* Dejean contains 1,200 described species found on the American continent (Pinzón-Navarro et al. 2010, Castañeda-Vildózola et al. 2014). Most occur in the neotropical region and feed on reproductive and vegetative structures of a broad diversity of dicotyledons (Anderson 2002, Marvaldi et al. 2002, Marvaldi and Lanteri 2005).

In Mexico, 86 species of the genus are reported (O'Brien and Wibmer 1982); of these species, six are major pests associated with fruit with high commercial value. *Conotrachelus aguacatae* Barber and *C. perseae* Barber perforate fruit of avocado (*Persea americana* Mill) and feed on the seeds. This reduces yield and can restrict exporting (Domínguez et al. 2015); *C. dimidiatus* Champion and *C. copalensis* Salas and Romero attack fruit of guava (*Psidium guajava* L.), causing production losses of 60%, and limit transportation of the fruit into zones free of the pests (Tafoya et al. 2010, Sánchez 2011, Salas and Romero 2012, Aragón et al. 2015). *C. crataegi* Walsh bore into fruit of Mexican hawthorn (*Crataegus* spp.) and cause 40 to 100% less yield (Muñiz et al. 2012). *C. eburneus* Champion attacks and affects the appearance and commercial value of fruit of red nanche (*Malphigia mexicana* Juss.) (Castañeda-Vildózola et al. 2014).

Identification of *Conotrachelus* species of agricultural interest, even the most abundant species, can be difficult without previous training, and the problem is greater when only female specimens are available. This is important because correct identification of insect species in agriculture is necessary for decision-making when implementing a program of pest management or, in the case of quarantined pests, avoiding closing markets because of incorrect determination.

For these reasons, the objective of this work was to use diagnosis and comparative illustrations of diagnostic structures to provide a comprehensive resource for facilitating identification of adults of each species of *Conotrachelus* weevil important to agriculture and subject to quarantine in Mexico. We also developed a taxonomic key for adults and descriptions of male and female genitalia that could serve as a tool for differentiation or characterization of the six species.

## Materials and Methods

Adult *Conotrachelus* specimens were captured directly from hosts at different locations and dates in Mexico. Additional material was examined from entomological collections at Colegio de Postgraduados (CP), and Instituto Nacional de Investigaciones Forestales, Agrícolas y Pecuarias (INIFAP) campus Celaya.

In a laboratory, the insects collected were killed with ethyl acetate and mounted on entomological pins or fixed in 70% alcohol for use in later studies. The adult weevils were identified using the keys of Schoof (1942), Whitehead (1979), Salas and Romero (2012), and Domínguez et al. (2015). The terminology for describing adults used the format of Lyal (2017).

Photographs of lateral and dorsal views of each species and distinctive characteristics were taken with a Carl Zeiss Tessovar photomicroscope. To extract genitalia of each species, the abdomen was separated and placed into an Eppendorf tube containing 10% KOH for maceration at 80°C in a thermoblock (AccuBlock, Digital Dry Bath) for 20 to 30 minutes. The abdomens were placed in Petri dishes with 70% alcohol to extract the genital structures (Sternites VIII and spermatheca of females and aedeagus and spicula of males). The structures were fixed with gel in Petri dishes and covered with alcohol to prevent dehydration. The genital structures were photographed by using a Carl Zeiss photomicroscope III.

Combine ZP software (Hadley 2010) was used for optimal focus of the photographs. The examined material was archived in the insect collection of the Agricultural Entomology Program of the Colegio de Postgraduados, campus Montecillo, State of Mexico.

## Results

A diagnostic, taxonomic key and images for identification of six *Conotrachelus* species of agricultural importance for Mexico were created. The diagnosis and key are based on easily observed external morphological traits.

***Conotrachelus perseae*** (Fig. 1 A-B): Piliform scales with white, pink, orange, or red coloring. Pronotum from dorsal view with lateral edges with a soft constriction at the middle part of the segment (Fig. 2A). Humeral region is one fourth wider than the prothorax base; rounded-tipped angular humeri, a distinctive characteristic of the species (Fig. 3A).

***Conotrachelus aguacatae*** (Fig. 1 C-D): Piliform-type scales fewer, but with coloring similar to *C. perseae*. Prothorax in dorsal view with lateral edges that decrease toward the apex with no evident constriction (Fig. 2B). Humeral region narrower than in *C. perseae*; rounded humeri (Fig. 3B).

***Conotrachelus dimidiatus*** (Fig. 1 E-F): Piliform scales of varying color: reddish brown, yellow, and white, some forming patches. Pronotum in dorsal view with a strip of white pubescence on the hind edge, inverted V- or U-shaped. Lateral edges of the pronotum with a soft constriction in the middle of the segment (Fig. 2C). Humeral region one fourth wider than the base of the pronotum; angular humeri rounded at the tip (Fig. 3C).

***Conotrachelus crataegi*** (Fig. 1 G-H): Reddish brown to dark brown with bands of lighter scales. Dense layer of piliform scales covering the entire body. Strip of white pubescence on the pronotum with appearance of an inverted V or U. Lateral margins of the prothorax with a notable abrupt constriction on the middle part of the segment (Fig. 2E). Humeri angular, sharp tips (Fig. 3D). Elytral crests discontinuous and prominent with thin, semi-erect brown scales.

***Conotrachelus copalensis*** (Fig. 1 I-J): General dark brown coloring. Scales broad at the apex and sharp at the insertion point, mixed with a smaller quantity of piliform, semi-erect, white scales. Pronotum, dorsal view, has lateral margins with well-marked constriction at mid-segment (Fig. 2D). Squared and rounded humeri. The dividing line between pronotum and elytra is almost straight. Elytral slope is rounded and pronounced (Fig. 2E).

***Conotrachelus eburneus*** (Fig. 1 K-L): Dense layer of piliform scales covering the entire body, middle part of the elytra with a wide line of lighter scales. Prothorax and femurs with a line of white scales. Robust rostrum as long as or shorter than the prothorax. Postocular lobule absent. Square humeri with rounded edges.

### Key for Identification of Six *Conotrachelus* Species of Agricultural and Quarantine Importance for Mexico

1. Whitish scales covering the body (Fig. 1 K-L). Associated with red nanche (*Malphigia mexicana*).....**C. eburneus (Fig. 1 K-L)**
- 1'. Brown scales covering all or most of the body.....**2**
2. Dividing line between pronotum and elytra almost straight (Fig. 3C and 3E).....**3**

- 2'. Dividing line between pronotum and elytra sinuous or wavy, with or without macules (Fig. 3A, 3B, and 3D).....4
3. Elytral slope round and pronounced; wide scales at the apex and sharp at the insertion point, mixed with rows of piliform scales (Fig. 2D); scarcely developed elytral crests; square humeri with rounded edges (Fig. 3E). Associated with guava (*Psidium guajava*).....**C. copalensis (Fig. 1 I-J)**
- 3'. Elytral slope continuous; abundant piliform scales, elytral crests developed, but not prominent; angular humeri with rounded tips (Fig. 3C); white strip of pubescence on the hind margin of the pronotum, inverted V- or U-shaped (arms of the V pointing toward back) (Fig. 2C). Associated with guava (*Psidium guajava*).....**C. dimidiatus (Fig. 1 E-F)**
4. Elytral crests discontinuous and prominent; macules absent in the lobules of articulation of the elytra (Fig. 3D); angular humeri sharp at the tip; abundant piliform scales, lateral margins of the prothorax with a notable abrupt constriction in the middle part of the segment; white stripe of pubescence on the pronotum with an inverted V or U shape (Fig. 2E). Associated with hawthorn fruit (*Crataegus* spp.).....**C. crataegi (Fig. 1 G-H)**
- 4'. Elytral crests barely developed, generally macules present in the lobules of articulation of the elytra; piliform scales less dense (Fig. 1A-D).....5
5. Lateral margins of the prothorax with a soft constriction at the middle of the segment; well-marked strip of bare cuticle on the longitudinal middle dorsal line of the prothorax (Fig. 2A); angular rounded-tipped humeri (Fig. 3A). Associated with avocado (*Persea americana*).....**C. perseae (Fig. 1 A-B)**
- 5'. Lateral margins of the prothorax narrow toward the apex with no evident constriction; strip of bare cuticle hardly perceptible on the longitudinal middle dorsal line of the prothorax (Fig. 2B); rounded humeri; distance between humeri shorter than in *C. perseae* (Fig. 3B). Associated with avocado (*Persea americana*).....**C. aguacatae (Fig. 1 C-D)**

### Description of Male and Female Genitalia

**Conotrachelus perseae.** Aedeagus with almost parallel margins, dorsally with a rectangular appearance, slightly widened at the distal tip and a little narrower at the basal tip; seen laterally, the apex bends ventrally at a 90° angle. Tip of the margins rounded with fine, short pubescence. Aedeagus apodemes short, from a lateral view with marked ventral curvature (Fig. 4A). Sternite VIII inverted V-shaped, medium short stem, symmetric divergent lateral arms ending in a flat rounded tip with several long setae; mid part of the arms widened forming a semicircular membrane (Fig. 5A).

**Conotrachelus aguacatae.** Aedeagus with parallel edges that converge into a point at the apex, giving the appearance of a “V”; lateral view, apex curved ventrally from which a middle membranous lobe protrudes; apodemes not curved ventrally, but are longer than those of the previous species (Fig. 4B). Inverted Y-shaped Sternite VIII; medium short stem and hind tip spatula-shaped; lateral arms curved outward in the first third of their length, later converging toward the tip (Fig. 5C).

**Conotrachelus dimidiatus.** Aedeagus formed by a curved lobule with almost parallel edges; dorsal view rectangular appearance; lateral view, aedeagus with well-marked ventral curve. Apodemes almost as long as the aedeagus, with a small fold in the middle part (Fig. 4E). Sternite VIII inverted Y-shaped, medium short stem, at the back end there is a slight dilatation; lateral arms curve outward and

later converge toward the middle plane, widened at the place of maximum separation; hind tips are rounded, membranous with numerous long setae (Fig. 5B).

***Conotrachelus copalensis*.** Aedeagus formed by a flat lobule, but with several ventral curvatures; from a dorsal view, it extends abruptly near the apex; from lateral view is a slight curvature near the apodemes; apex curved ventrally, from which a membranous middle lobe stands out. Long apodemes, approximately half the length of the aedeagus (Fig. 4C). Sternite VIII inverted Y-shaped, middle stem is one third of its length; lateral arms curved outward the last third of their length and later converge toward the middle plane, their hind tips are rounded, slightly thickened, and with many setae (Fig. 5E).

***Conotrachelus crataegi*.** Apodemes with ventral curvature as long as the mean length of the aedeagus (Fig. 4D). Sternite VIII inverted Y-shaped, long middle stem, approximately a fourth as long as the sclerite. Its fore tip has a slight translucent dilatation; lateral arms curved outward and later converge toward the middle plane, the last quarter of its length thickened slightly, its hind tips are membranous, rounded, and with numerous long setae (Fig. 5D).

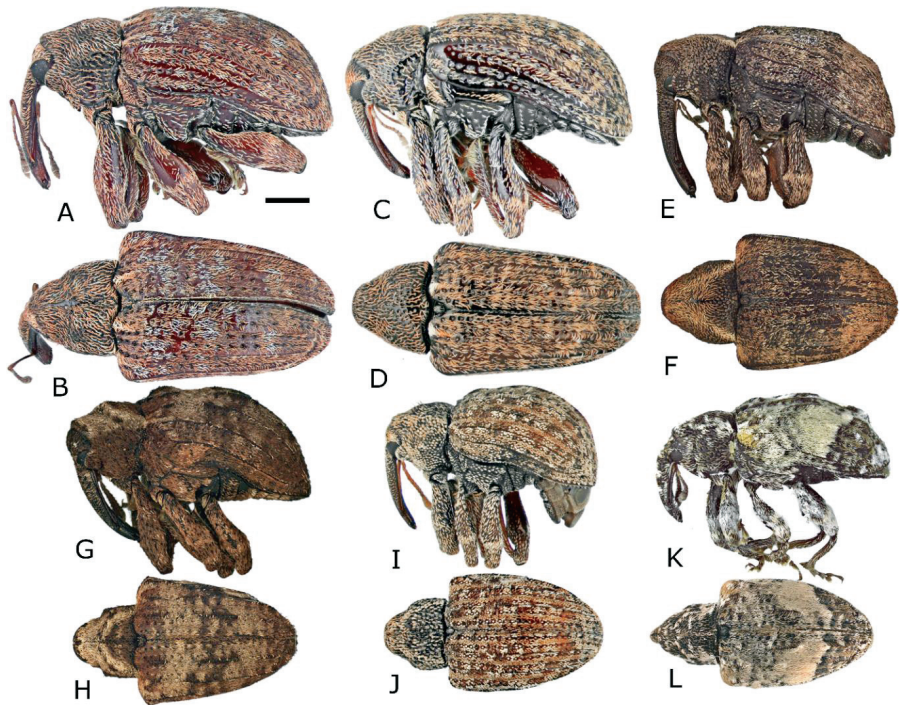


Fig. 1. Lateral and dorsal view of male. A and B) *C. perseae*; C and D) *C. aguacatae*; E and F) *C. dimidiatus*; G and H) *C. crataegi*; I and J) *C. copalensis*; K and L) *C. eburneus*. Scale bar 1 mm.



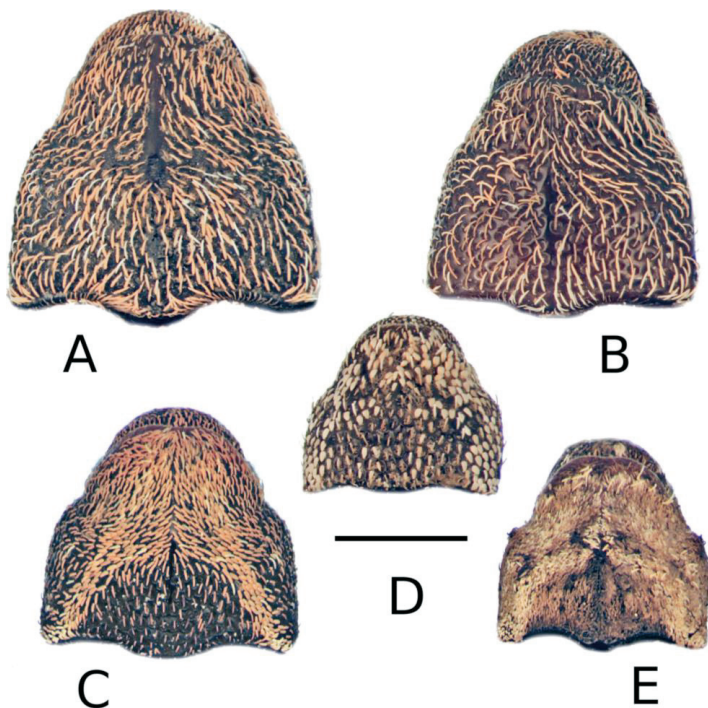


Fig. 2. Pronotum dorsal view. A) *C. perseae*, B) *C. aguacatae*, C) *C. dimidiatus*, D) *C. copalensis*, E) *C. crataegi*.

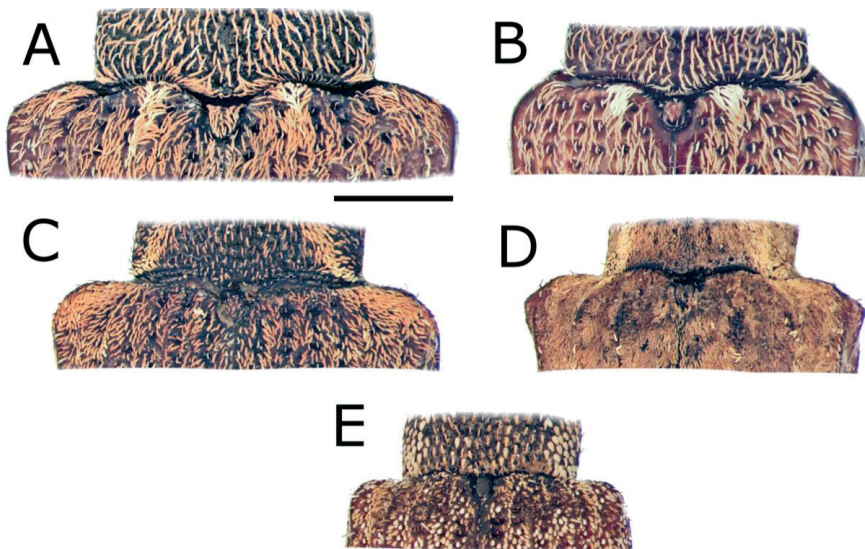


Fig. 3. Humeri and dividing line between pronotum and elytra. A) *C. perseae*, B) *C. aguacatae*, C) *C. dimidiatus*, D) *C. crataegi*, E) *C. copalensis*.

***Conotrachelus ebuneus*.** Aedeagus formed by a translucent lobule of almost parallel margins seen dorsally, thicken slightly, giving the general appearance of a rectangle; from a lateral view, it has a slight ventral curvature at the middle of its length, apex with slight ventral curvature; from a dorsal view, the apex is rounded. Apodemes short and thin, hardly perceptible (Fig. 4F). Sternite VIII has an inverted Y shape; long middle stem, approximately one third of the length, flat, wide, and with a slight dilatation in its hind part; lateral arms curve outward and then converge toward the middle plane; in the last third of its length, it thickens and forms a rounded membrane with numerous long setae (Fig. 5F).

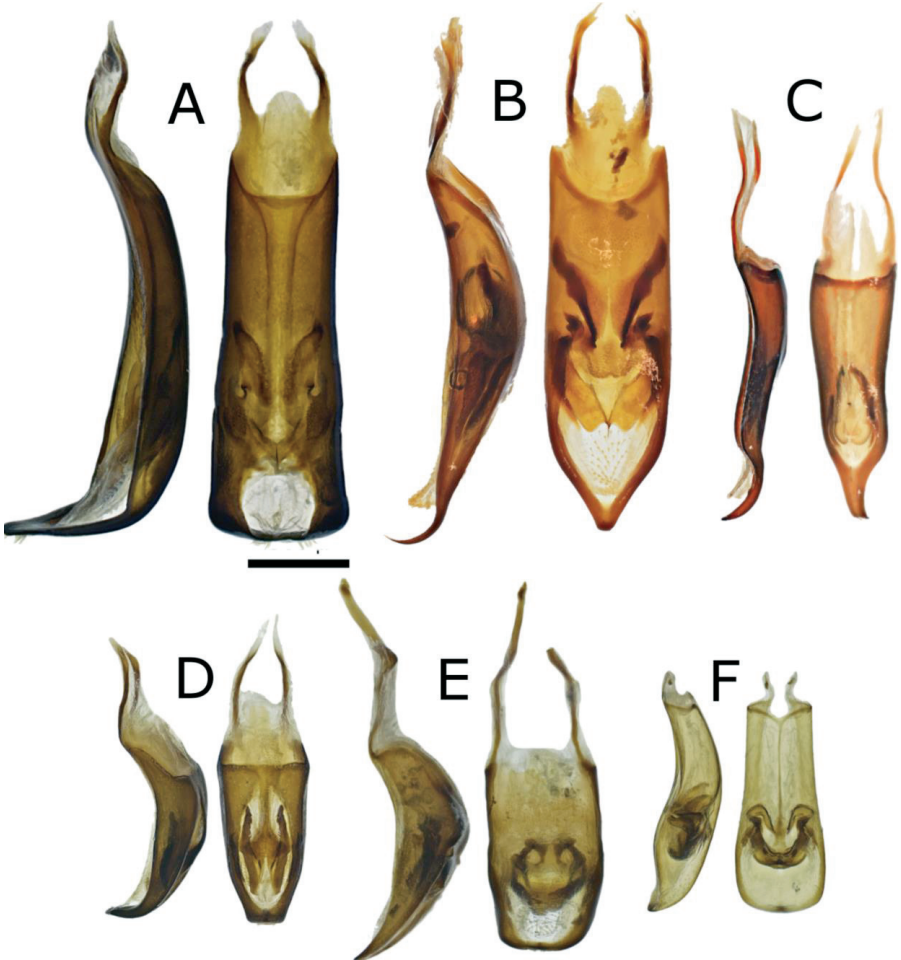


Fig. 4. Lateral and dorsal view of aedeagus. A) *C. perseae*, B) *C. aguacatae*, C) *C. copalensis*, D) *C. crataegi*, E) *C. dimidiatus*, F) *C. ebuneus*.



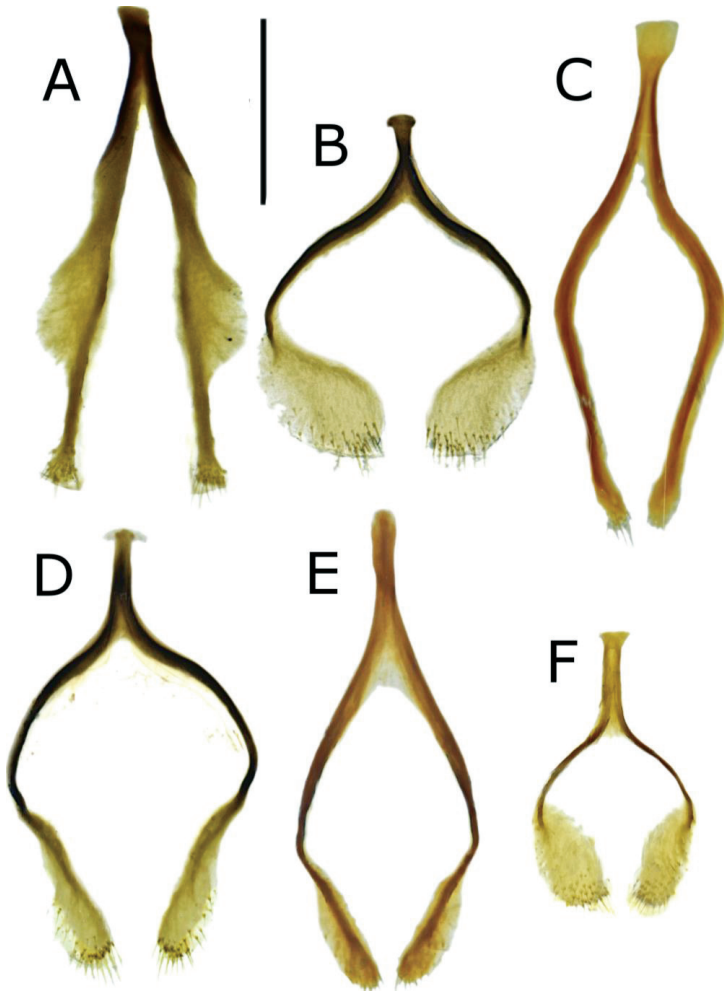


Fig. 5. Sternite VIII. A) *C. perseae*, B) *C. dimidiatus*, C) *C. aguacatae*, D) *C. crataegi*, E) *C. copalensis*, F) *C. eburneus*.

### Discussion

*Conotrachelus perseae* and *C. aguacatae* were described by Barber (1919, 1923). The specimens were studied and redescribed by Muñiz (1970) who did the first complete morphological study of the species in Mexico. However, he did not describe the female genitalia. Whitehead (1979) published a key to differentiate three species of *Conotrachelus* that attack avocado in Mexico, however he referred to the schemes of genitalia published by Barber (1923). Domínguez et al. (2015) reported a detailed morphological study of *C. perseae* that included the genitalia of both sexes and mentioned characteristics to differentiate it from *C. aguacatae*.

Studies of *C. dimidiatus*, *C. crataegi*, and *C. copalensis* mention short descriptions of the aedeagus and external morphology of the adults, but omit female genitalia (Schoof 1942, Muñiz and Gonzalez 1982, Salas and Romero 2012). Champion (1904) described the *C. eburneus* adult, but did not include a description of the genitalia.

The six *Conotrachelus* species studied can be differentiated by external morphology (Fig. 1). Traits for differentiation are the lateral margins of the prothorax and humeral area. However, results showed that identification using male and female genitalia were more effective. The aedeagus and Sternite VIII were the structures notably different among species, coinciding with a report by Castañeda-Vildózola et al. (2007) for weevils of the *Heilipus* Germar genus associated with avocado. Female genitalia constitute an additional taxonomic tool for separating *Conotrachelus* species and can be used with certainty in the absence of male specimens; our result was supported by Gaiger and Vanin (2008).

The six species exhibited sexual dimorphism: rostrum length and antennal insertion constitute the main intraspecific differentiation traits. However, there were no marked differences between sexes of *C. copalensis*. Color may be distinctive of each species but can vary because of intrinsic or extrinsic factors. Therefore, the genitalia was, in the end, the most reliable element for identification of species.

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