NEW SPECIES OF THE GENUS *Trigonospila* POKORNY (DIPTERA: TACHINIDAE) PARASITISES ADULT *Compsus* sp. SCHOENHERR (COLEOPTERA: CURCULIONIDAE) IN COLOMBIA*

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Abstract

Trigonospila unicaldasi new specie, is described for the first time for science. In order to differentiate it from other rarely occurring species in the Americas, key characteristics of the adult were identified through scanning electron microscopy. The species was found parasitising adult weevils of the genus *Compsus* sp. (Coleoptera: Curculionidae) associated with citrus crops in the central coffee-growing region of Colombia. The interesting symbiotic phenomenon of control described in this paper is considered unusual in the biology of Tachinidae given that only 5-10% of the family parasitise their hosts at the adults stage.

Key words: parasitism, adults, scanning electron microscope, description.

NUEVA ESPECIE DEL GÉNERO *Trigonospila* POKORNY (DIPTERA: TACHINIDAE) PARA COLOMBIA, PARASITANDO ADULTOS DE *Compsus* sp. SCHOENHERR (COLEOPTERA: CURCULIONIDAE)

Resumen

Trigonospila unicaldasi nueva especie, es registrada por primera vez para la ciencia con el propósito de diferenciarla de otras especies que son de escasa ocurrencia en las Américas. Para lograr este objetivo se consideraron las estructuras clave del adulto utilizando la técnica de Microscopía Electrónica de Barrido (MEB) para su completa descripción. La especie se encontró parasitando adultos de Compsus sp. (Coleoptera: Curculionidae) asociados a cultivos de cítricos en la zona cafetera central de Colombia. El interesante fenómeno simbiótico de control mencionado en este documento se considera un caso poco usual en la biología de los Tachinidae debido a que solo del 5% al 10% de estos parasitan especies en estado adulto.

Palabras clave: parasitismo, adultos, Microscopía Electrónica de Barrido, descripción.

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INTRODUCTION

Trigonospila Pokorny 1886 (Tachinidae: Blondeliini) is a cosmopolitan genus, present in the Old World in Eurasia, Australia, Oriental and Afrotropical region. In the New World is represented for three from the north of Mexico (O'HARA & WOOD, 2004) and four in South America, of which, two were found on larvae of Crambidae and Pyralidae (Lepidoptera), reared in Guanacoste, Costa Rica (JANZEN & HALLWACHS, 2005). Recently, FLEMING et al. (2015) describe three new species of the genus Trigonospila, and describe the male of Trigonospila panamensis (Townsend), all reared from wild-caught caterpillars collected from Área de Conservación Guanacaste, in northwestern Costa Rica. BYUN & HAN (2010) differentiated 18 nominal species (3 Palaearctic, 3 Oriental, 5 Australasian, 4 Afrotropical, and 3 Nearctic), with two species of the families Oecophoridae and Gelechiidae (Lepidoptera) parasitised by Trigonospila brevifacies (Crosskey 1973), and the Asian beetle Plesiophthalmus nigrocyaneus (Motschulsky) (Tenebrionidae) as host to Trigonospila vittigera (Coquillett). T. brevifacies was introduced to Australia and northern New Zealand to control the tortricid pest *Epiphyas postvittana* (Walker) between 1967 and 1973, and in southern New Zealand in 1999 (SHAW et al., 2001). The species is known to oviposit directly between the head and thorax of lepidopteran larvae, where the newly hatched larvae of the parasitoid complete their whole life cycle (GREEN, 1984). Parasitism on adult insect hosts is poorly known in Tachinid flies (BROWN et al., 2010). T. unicaldasi represents a promising parasitoid for the control of Compsus sp. (ARNETT et al., 2002), an important economic species, detected since 1939, mainly in citrus crops in the departments of Quindío, Caldas, Valle, Tolima, Antioquia and Risaralda in Colombia, causing considerable damage to foliage and tree roots (PÁSSARO, 2012).

This study describes a new species of the genus *Trigonospila* found parasitising adults of the citrus weevil, *Compsus* sp. in Colombia. Using images obtained from scanning electron microscopy we describe the male holotype and differential female characteristics which allow the species to be described as *Trigonospila unicaldasi*.

MATERIALS AND METHODS

The description follows the format of BYUN & HAN (2010) which includes several morphometric ratios: frons-head ratio (frons/head width); eye ratio (length/width); arista-antenna ratio (arista length/antenna length excluding arista); wing-thorax ratio (wing length/thorax length); abdomen ratio; genital differences. The structures morphological in this study was based conform to the keys found in WOOD (1985) and WOOD & ZUMBADO (2010).

Collection of entomological material: Twenty insects were taken from hosts *Citrus* limon (lemon) trees, Rubus glauca (andean blackberry) bushes and Passiflora molissima (curuba) plants, grown in an experimental plot of the Granja Tesorito, belonging to the Universidad de Caldas. The university farm is located 6 km along the highway between Manizales and Bogotá in Valle de Maltería. The specimens were collected using the umbrella method, which consists of putting a plastic under each plant and then shaking it so that the insects fall on it facilitated by *Compsus* sp. adults thanatosis behaviour, or apparent death some of which had previously been parasitised by T. unicaldasi. The specimens were collected in plastic containers, measuring 20 x 15 x 6 cm, and labelled with the following information: date, host plant, altitude above sea level, quantity per host plant and name of collector. The material was taken to the Entomological Laboratory of the Universidad de Caldas (LEUC) to be disinfected (washed with distilled water, 3% sodium hypochlorite and 75% ethanol for 5 minutes). Fifteen specimens of Compsus sp. adults were placed on sand and filter paper inside sterilised plastic containers measuring 15 x 15 x 10 cm. From this moment on the parasitised specimens were monitored every 48 hours in order to obtain immature stages of T. unicaldasi. Pupae of T. unicaldasi were transferred to a cage designed for manipulating live insects (BIOQUIP® reference 1450TC). Small recipients with cotton wool soaked in water and 10% honey solution were installed for flies to feed on until adult specimens were obtained. Twenty individuals were chosen from the whole population (10 males and 10 females) for analysis with scanning electron microscopy.

Scanning electron microscope analysis: Specimens of *T. unicaldasi* were taken to the Electron Microscope Laboratory of the Universidad de Caldas where samples were placed on plates and photographed with an FEI- QUANTA 200 electron microscope and processed in Microsoft Office 2010. Dimensions given for the anatomical structures are the mean of measurements taken from 10 male and 10 female specimens, obtained via scanning electron microscopy. Ten of the specimens were deposited in the Laboratory of Entomology at the Faculty of Agricultural Sciences, Universidad de Caldas, Manizales, Colombia.

RESULTS

Trigonospila unicaldasi Vinasco, Vallejo and Soto, sp. nov. (Figs. 1-11)

Type material: Holotype labelled as "Colombia, department of Caldas, municipality of Manizales, Granja Tesorito, Valle de Maltería, Universidad de Caldas, 2280 m, January 20, 2011, in adults of *Compsus* sp. feeding on *Citrus limon* L. (lemon), Alberto Soto Giraldo". Original label in Spanish: "Colombia, departamento de Caldas, Municipio de Manizales, Granja Tesorito, Valle de Maltería, Universidad de Caldas 2280 m, enero 20 de 2011, en adultos de *Compsus* sp. alimentándose de hojas *Citrus limon* L. (limón), Alberto Soto Giraldo".

Holotype deposited at the Entomology Laboratory Collection, Universidad de Caldas (LEUC), Manizales, Colombia.

Description of the male holotype

Diagnosis: Terminology used by BYUN & HAN (2010) and the morphological description by Wood (1985) and WOOD & ZUMBADO (2010) of members of the genus *Trigonospila* is based on a combination of the following characters: transverse body bands, compound eyes with proclinate and reclinate orbital setae, upper frontal setae, ocellar setae, parafacial setae, facial ridge, vibrissae and subvibrissae, epistomal suture, gena, antennae, flagellum, flagellomere, arista, thoracic segments, wing venation, distribution of setae on legs (Fig. 1). Measurements are given in millimetres.



Figure 1. Trigonospila unicaldasi, adult male. Lateral view.

Male: Body pale yellow with vertical brown bands on thorax and horizontal bands on abdomen; wing length: 1.12-1.54; thorax length: 1.09-0.91; body length, from cephalic capsule to posterior end of abdomen 3.0-3.82; mid abdominal zone 1.14-1.53. Semicircular compound eyes with minute setae 0.01in length and alternating with ommatidia. Eye ratio 0.47-0.82, arista-antenna ratio 0.53-0.86; gena-eye ratio 0.09; exterior setae present; occelar seta 0.29, as long as outer vertical seta; postocellar

seta 0.07; paravertical seta short but different to anterior, almost 0.14, as long as ventral outer seta; ocellar triangle; bare lunule; orbital plate yellow, same shade as dorso, with wide, long setae; orbital seta markedly smaller than frontal setae; parallel setae at least twice as long as inner; divergent ocellar setae; dark, bare frontal vitta; dark brown arista with 3 aristomere; scape very short and wide, pedicel slender, first flagellomere markedly wide and long; facial carina wide, concave, as long as length of first flagellomere.

Head (Figs. 2-3). Yellow-brown in colour, dimensions: 1.25 frontal width, and 0.96 laterally. Parafacial bare, paler than other regions around compound eye; postocular wide in relation to eye margin; occiput with small setae; postcranium with one row of black setae, longer than postocular row, rest of the postcranium covered with small, pale setae. Postgena short, 0.08 in height, moderately swollen; genal setae differentiated and genal dilation not well developed; prominent vibrissa, markedly wide at base, sufficiently long to overlap, and project over epistome (Fig. 2), supravibrissa also evident. Supravibrissal setulae absent; 4-6 subvibrissal setae; mouthparts with lightly clavate palps and with small setae (Fig. 3); prementum dark brown, with small setae; labellum paler and covered with abundant setae.

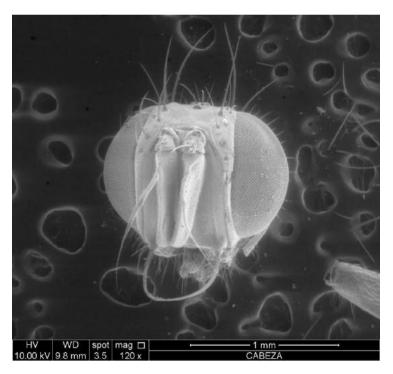


Figure 2. Head of male *Trigonospila unicaldasi*. Frontal view (note appearance of lunule).

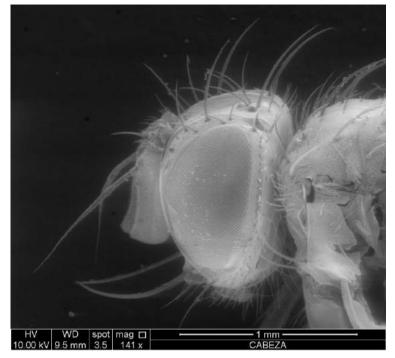


Figure 3. Head of male Trigonospila unicaldasi. Lateral view.

Thorax (Fig. 4). Yellow with 4 dark longitudinal bands on the scutellum; prosternum bare; anepisternum with one anterior-dorsal seta; anepimeron bare; proepisternum with 2 large setae projected upwards; proepimeron wide and bare; katepisternum with 2 wide, prominent setae, katepimeron bare, semi-rectangular; katatergite bare, anatergite bare, wide meron, with a row of 4 prominent setae, the first smaller, second and third joined, and fourth slightly separated from the group. 2 + 3 acrostichal; 2 + 3 dorsocental; 2 + 3 intraalar; 2 + 4 supraalar; postpronotum with 2 setae; scutellum with 4 scutellar setae, 2 of these apical, subapical scutellar setae 1.52-1.64 times as long as scutellum length, lateral scutellar setae 0.42 - 0.55 times as long as subapical scutellar setae; mediotergite dark brown with pale yellow pruinosity. Semicircular spiracle 0.05 in height by 0.06 wide, with densely pilose edges.

Legs (Figs. 5-6). All three pairs are dark brown in colour, with dark setae arranged longitudinally; proleg coxa with 8-10 strong antedorsal setae; femur 1.07 in length, strong with 7-8 setae on each edge, tibia elongated, cylindrical, 1.03 in length, with rows of 3-5 anterodorsal, 2-3 posterodorsal and 1 ventral setae, distal corona made up of 6-7 long, slender spines, tarsi with 5 long tarsomeres, similar in shape, the first twice as long as the second, the second a third longer than the third and the fourth longer than the pretarsus. Foretarsus with double row of setae on each edge, tarsal claws short, with pulvillus (Fig. 6) unpronounced.



Figure 4. Thorax of male Trigonospila unicaldasi. Lateral view.

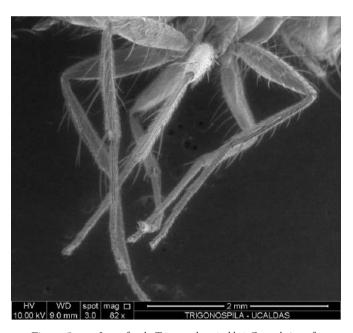


Figure 5. Legs of male *Trigonospila unicaldasi*. General view of the pro, meso and meta legs.

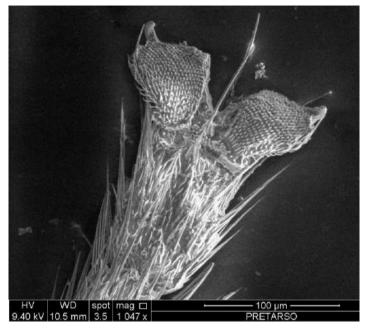


Figure 6. Pulvillus of male Trigonospila unicaldasi.

Wings (Fig. 7). Transparent, light brown in colour; distance between base and apex 3.41; costal vein interrupted close to the Sc apex; C-Sc ratio 0.85-1.20, bifurcation of R_{2+3} and R_{4+5} with a prominent dorsal seta, 0.31 in length, ratio 2.63-3.24; DM-Cu transverse vein straight; tegula darker colour than rest of wing; calypter yellow, bare, with edges covered in dense minute setae (microtrichia).

Abdomen (Figs. 8-9). Five metamera, pale yellow with brown bands, metamera 1 and 2 fused, with double, very prominent, long, bright setae on the upper region of each tergum, 1 prominent, long, shiny seta on the lateral segment of the tergum 1+2, 2 long, prominent, shiny setae on the third, 3 on the fourth, and 2 on the fifth.

Male genitalia (Figs. 10-11). Large, dark brown-yellow; sternite 5 with a medium U-shaped cleft; cerci heavily sclerotised with pilose surface, shaped like curved hooks with an internal concavity, ending in a point; surstylus, wide, concave, distal edge surrounded by conspicuous points, rounded apex with minute and fine setae; elongated epiphallus, sclerotised, short and denser than the rest of the structure, pregonite elongated, concave throughout, ends in a distinctive, sharp projection, phallus bifid, covered in a fine layer of minute spines from base until the extreme distal end of internal face; epandrium globulose with 10-12 large, thick setae on external surface; hypandrium concave, wide with large setae on mid and apical surface.



Figure 7. Wings of male Trigonospila unicaldasi.

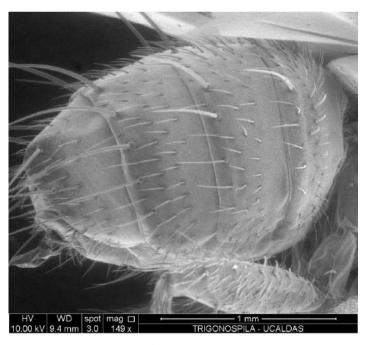


Figure 8. Abdomen of male *Trigonospila unicaldasi*. Lateral view.

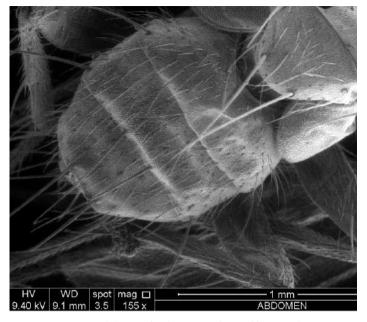


Figure 9. Abdomen of male Trigonospila unicaldasi. Dorsal view.

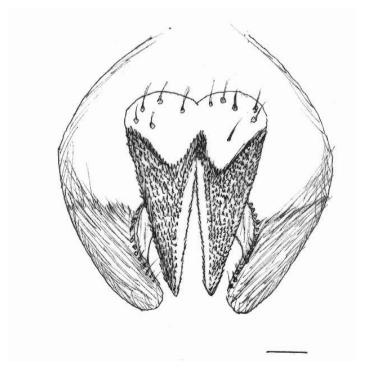


Figure 10. Epandrial complex, sternite 5 of male *Trigonospila unicaldasi*. Caudal view.

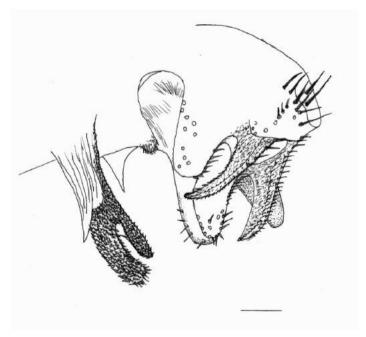


Figure 11. Epandrial complex, sternite 5 of male *Trigonospila unicaldasi*. Lateral view.

Female. Similar to males except for genital structure. Body length from cephalic capsule to posterior end of abdomen 3.41-5.0; mid abdominal zone 1.34-1.82; wing length 3.445; thorax length 1.199; frons-head ratio 0.70-0.94; gena-eye ratio 0.174; from 9 to 10 frontal setae; 3 orbital setae, 0.50-0.81 times as long as inner vertical seta; 1 reclinate orbital seta, 0.54-0.75 times as long as inner vertical seta. Thorax and abdomen as described for male except for female terminalia.

Material examined. Male holotype: Colombia, Caldas, km 6 on Manizales – Bogotá Highway, in Valle de Maltería, 5°06′ N, 75°33′ W, 2160, immature stages of *Trigonospila* sp collected from *Compsus* sp. in plantations of *Passiflora mollisima*, *Rubus glauca* aned *Citrus limon* x *limon* 1, 2011, col. A. Soto. G (LEUC). Paratypes: same as holotype: 5 adult males, 5 adult females.

Etymology. The specific epithet 'unicaldasi' is in homage to the Universidad de Caldas, institution supporting this research and where the authors are based. Furthermore, the parasi.0toid was found for the first time within the university farms.

Geographic distribution. Colombia, Caldas, Manizales. Pre-montane Forest (bh-PM) according to the classification of life zones by HOLDRIDGE (1967).

Host. Compsus sp. (Schoenherr) (Coleoptera: Curculionidae).

Biology. Adult *Compsus* sp. parasitised by *T. unicaldasi* shows slow behaviour and a dark colouration at the end of its abdomen (Fig. 9). At an average temperature of 18° C and relative humidity of 75%, adult females survive for an average of 35 ± 10 days (n = 20) and adult males for an average of 25 ± 11 days (n = 23). Sex ratio in adults is 1:2 (females:males) for 78% (n = 47) of emergent adults.

Associated plants. Although it is known that *Compsus* sp. attack several species of plants, especially citrus fruits, the specimens were collected mainly on *P. mollisima*, where the largest number of parasitised adults was found, possibly due to the availability of nectar from its flowers serving as a food source for adults of *T. unicaldasi*.

Observations. As stated above, the genus *Trigonospila* is poorly known in the Neotropical region and is not comprehensively described in literature. As a result, species are very difficult to identify with certainty. *T. unicaldasi* has been compared with some *Trigonospila* species housed at the Canadian National Collection of Insects Agriculture and Agri-Food. Dr James O'Hara, (Systematic Entomology, Eastern Cereal and Oil Seed Centre, Ottawa, Canada) identified and placed these samples within the genus after comparisons with specimens from the abovementioned collection. In order to put a name to this potential biological control agent, both its identification by O'Hara and the presence of this species in the Neotropics is sufficient to be able to name this a new species to science.

DISCUSSION

The genus *Trigonospila* has been poorly studied around the world, with most studies concerning the Neartic, Paleartic and Asia. The first record for Colombia comes from adult citrus weevils, *Compsus* sp., parasitised by *T. unicaldasi*, in a pre-montane forest. The weevils were observed to reduce their movement before mature larvae of the parasitoid emerged through the exterior anal canal or through the cervical membrane, decapitating the host insect. Most of the time, parasitoid larvae emerge after the weevil has died and move slowly until finding a suitable place in which to pupate (SOTO, 2002). The parasitoids obtained belong to the Diptera order, Tachinidae family, Exoristinae subfamily and Blondellini tribe (SOTO & OCAMPO, 2011).

T. unicaldasi represents a promising biological control agent for adult Compsus (Coleoptera: Curculionidae) given that this coleopteran has become a limiting pest for the citrus fruit industry due to inappropriate management by farmers in certain agricultural areas of the country. The pest has caused a reduction in vigour and productivity of citrus trees; increased their susceptibility to radicular fungal attacks;

and, in many cases, their death (CORRALES, 2002). Furthermore, is it unusual to find a Tachinid parasitising coleopteran adults, rather, it is more common to find them parasitising immature stages, thus this represents a novel and interesting relationship between parasitoid and host in the field (BROWN *et al.*, 2010).

This study contributes to knowledge of the family Tachinidae and principally, the genus *Trigonospila*, poorly known in Latin America. Also, it aims to resolve the confusion arisen between the genera *Trigonospila* and *Lyxophaga*, especially concerning differences between the two, such as their parasitism mechanisms, morphotypical dimensions, antennal segments, genital differences and phenotypic characteristics. These characteristics have been observed using scanning electron microscopy, which guarantees the veracity of, and complements, morphological studies.

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