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Diolcogaster flammeus sp. nov. from Brazil, a new Microgastrinae wasp (Hymenoptera: Braconidae) of importance in biological control

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ABSTRACT

A new species of *Diolcogaster* (Ashmead, 1900) (Hymenoptera: Braconidae) is described and illustrated. Additionally, a key to all New World species formally published of the *xanthaspis* species-group (to which the new species belongs) is provided. The solitary larval parasitoid *Diolcogaster flammeus* sp. nov. was collected in Viçosa, Minas Gerais State, Brazil. This natural enemy was reared from caterpillars of *Agaraea minuta* (Schaus, 1892) (Lepidoptera: Erebidae) feeding on plants of spiked spiralflag ginger, *Costus spicatus* (Jacq.) Sw. and ginger spiral, *Costus spiralis* (Jacq.) Roscoe var. *spiralis* (Costaceae). The fauna of the *xanthaspis* group in the New World included just three published species prior to this publication. *Diolcogaster flammeus* sp. nov. is the only yellow-orange species of the *xanthaspis* group recorded in the New World thus far.

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The subfamily Microgastrinae (Hymenoptera: Braconidae) comprises 2700+ described species (Yu et al., 2016; Whitfield et al., 2018) and many thousand additional undescribed ones (Rodriguez et al., 2013; Fernández-Triana and Boudreault, 2016; Moghaddam and Mokhtari, 2017). Microgastrinae is the single most important group of parasitoid wasps attacking caterpillars (Avila et al., 2013; Fiaboe et al., 2017), with many species used or being considered as biocontrol agents against lepidopteran pests in agriculture and forestry (Smith et al., 2013; Pinto et al., 2014). The genus *Diolcogaster* (Ashmead, 1900) is currently the sixth largest of Microgastrinae, with 98 species described worldwide (Zeng et al., 2011; Fernández-Triana, 2015) and hundreds awaiting description. This genus has 17 species recorded from the New World, 10 being from the Neotropical region, and four in Brazil (Gupta and Fernández-Triana, 2014, 2015; Yu et al., 2016).

Nixon (1965) included six *Diolcogaster* species in the *xanthaspis* species-group: *Diolcogaster xanthaspis* (Ashmead, 1900) from the West Indies (Saint Vincent and Grenada) and five other species from the Old World tropics (the Philippines). Later, Choi (2005) also considered two additional species from the New World: *Diolcogaster bakeri* (Muesebeck, 1922) and *Diolcogaster iridescens* (Cresson, 1865) to belong to that group. Based on specimens seen in collections worldwide, many more species remain undescribed (in fact, Choi, 2005 described six additional New World species for the group, but unfortunately his Ph.D. thesis has never been published and thus those species names are not valid – and are not considered here).

Diolcogaster parasitizes early larval stages of Lepidoptera (Yeargan and Braman, 1986) including *Agaraea minuta* (Schaus, 1892) (Lepidoptera: Erebidae) (synonym = *Agoraea minuta* Schaus, 1892), a major pest of spiked spiralflag ginger, *Costus spicatus* (Jacq.) Sw. and ginger spiral, *Costus spiralis* (Jacq.) Roscoe var. *spiralis* (Costaceae) plants (Tavares et al., 2012). It is unknown other host plant species of *A. minuta*. *Costus spicatus* is native to the Caribbean, including Dominica, Guadeloupe, Hispaniola, Martinique, and Puerto Rico (Keller et al., 2009), while *C. spiralis* is present in tropical South America (de Araújo and Oliveira, 2007). *Costus spicatus* and *C. spiralis* are economically significant due to their pharmacological and medicinal properties (Sulakshana et al., 2013; Pawar and Pawar, 2014; Alonso-Castro et al., 2016) as well as ornamental value (Castro et al., 2011; Skinner, 2016). *Agaraea minuta* is found from Mexico to Brazil (Vincent and Laguerre, 2014).

This study aims to describe a new species of *Diolcogaster* from Brazil. This previously undescribed species had already been found in a study on biological interactions, where it was found to attack *A. minuta* (according to Tavares et al., 2012, parasitism of *Diolcogaster* sp. on *A. minuta* was 90.0% in the municipality of Viçosa, Minas Gerais State, Brazil). Herein we describe the new taxon and place

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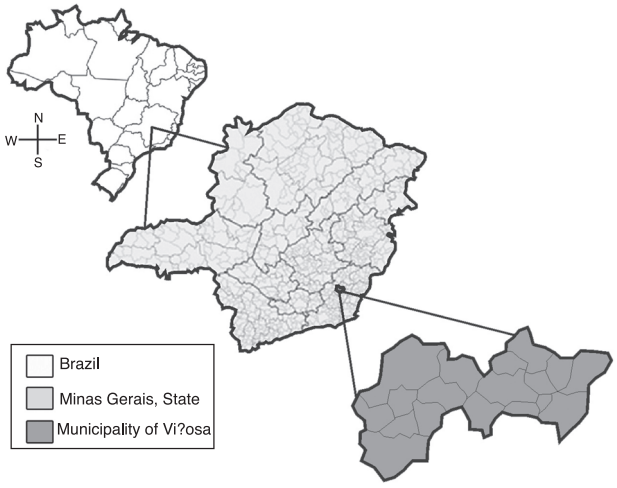

Fig. 1. Geographical location of the Brazilian municipality of Viçosa (Minas Gerais State, Brazil).

it into a taxonomic context by providing a key to all New World species formally published of the *xanthaspis* species-group.

Materials and methods

A plant of *C. spicatus* and another of *C. spiralis* var. *spiralis* were sampled in May 2011. Plants sampled were approximately five years old in the herbarium of the Federal University of Viçosa (UFV) in Viçosa, Minas Gerais State, Brazil (20° 45′ S × 42° 51′ W, 651 m above sea level) (Fig. 1). One hundred last instar caterpillars of *A. minuta* were collected, 50 from *C. spicatus* and another 50 from *C. spiralis*.

The occurrence of defoliating caterpillars of *A. minuta* was monitored in the plants of *C. spicatus* and *C. spiralis* var. *spiralis* in May 2011 due to the occurrence of these insects during that month in 2009 and 2010 (Tavares et al., 2012).

The caterpillars were collected with a fine-tipped brush, placed in 500 mL plastic containers lined with filter paper and with the lid aerated (20 caterpillars per plastic container in two containers and another one with 10 caterpillars) with leaves ad libitum of the respective host plants supplied as food and substrate for pupation (Tavares et al., 2012). They were brought to the Laboratory of Biological Control of Insects – UFV in Viçosa, Minas Gerais State, Brazil where they were kept at 25 ± 1 °C, 12:12 h (L:D) photoperiod and 70 ± 10% RH. The plastic containers were opened and cleaned when needed (Tavares et al., 2012). The leaf petioles were moistened to avoid desiccation (Tavares et al., 2012).

Some pupae of *A. minuta* were obtained and individualized in a 50 mL plastic container until the emergence of adults of this insect, which were mounted with entomological pins and four individuals (two males and two females) sent to Dr. Vitor Osmar Becker, of the Uiraçu Institute in Camacan, Bahia State, Brazil, to identify the species. Adults of the parasitoid, obtained from *A. minuta* caterpillars, were transferred to plastic Eppendorf™ vials with 70% ethanol and mounted with entomological pins. Four male and four female of the parasitoid were deposited in the Regional Entomological Museum – UFV. Adults of the parasitoid were identified by the second author of this study (JLF-T). The specimens used to describe the parasitoid in the present study were part of the ones used for the biological study by Tavares et al. (2012).

A wealth of information on *A. minuta*, including additional pictures (eggs, larvae, pupae, cocoons, and adults of *A. minuta* as well as damage caused by its larvae) was provided by Tavares et al. (2012).

Photos were prepared using a Keyence VHX-1000 Digital Microscope (Keyence Corporation of America; Illinois, United States of America), using a lens with a range of 10–130×. Multiple images were taken of a structure through the focal plane and then combined to produce a single in-focus image using the software associated with the Keyence System. Plates were prepared using Microsoft PowerPoint 2010.

Results

Diolcogaster flammeus **Salgado-Neto & Fernández-Triana sp. nov**.

Description. Diagnosis. This is the only yellow-orange species of the *xanthaspis* group recorded in the New World thus far. It is possible that more yellow-orange New World species will be discovered in a near future. **Female** (Fig. 2). Body color entirely yellow to orange yellow, except for dark brown to black on flagellomeres, parts of the scape, and most of the interocellar area, and most of T2–T4 brown. Fore wing with pterostigma and veins brown. Head mostly with striate sculpture on face, frons, gena and vertex; occiput strongly concave and smooth. Mesosoma with anteromesoscutum and scutellar disk rugose-punctate, scutellar disk posteriorly with a keel; propodeum with strong and complete median carina, and additional transverse rugosity on posterior 0.6 of propodeum. Metasoma with T1 slightly widening toward posterior margin and with strong and deep median sulcus running throughout the entire tergite; T2 subtriangular, with relatively strong sulcus delimiting lateral margins and a central area that is slightly raised and delimited laterally by punctures; T3+ smooth; ovipositor sheaths with a few setae on apical 0.3. Metacoxa relatively large, more than half the length of metasoma. Measurements (all in mm). Body length: 3.50. Fore wing length: 3.4. Metacoxa length: 0.96. Metafemur length/width: 1.12/0.31. Metatibia length: 1.22. Length of inner/outer spurs of metatibia: 0.53/0.30. Length of first segment of metatarsus: 0.70. Ovipositor sheath length: 0.25. T1 length: 0.60; T1 width at anterior margin/maximum width 0.20/0.28. T2 length/width at posterior margin: 0.24/0.50. T3 length: 0.36. Length of flagellomeres: F1/F2/F3/F14/F15/F16: 0.26/0.26/0.26/0.15/0.15/0.16.

Male (Fig. 3). Identical to female.

Remarks. Accounting for the new species from Brazil being described in this study, the fauna of the *xanthaspis* group in the New World now includes four species (Table 1). A key to all New World species is provided below.

1- Body almost entirely yellow to orange-yellow (except for flagellomeres, part of scape and most of interocellar area black, and most of T2–T4 partially brown) . . . *Diolcogaster flammeus* Salgado-Neto & Fernández-Triana sp. nov.
- Body mostly to partially dark brown to black . . . 2
2(1) Female antenna relatively short, as long as fore wing length, and with apical 8–9 flagellomeres with only one placode rank; metacoxa yellow or white-yellow . . . *Diolcogaster iridescens* (Cresson, 1865)
- Female antenna longer than fore wing, most flagellomeres with two ranks of placodes; metacoxa black to dark brown . . . 3
3(2) Second submarginal cell of fore wing triangular and relatively large in size, without defined vein RS + r – m; tergite 2 0.5 × as long as tergite 3. . . . *Diolcogaster bakeri* (Muesebeck, 1922)
- Second submarginal cell of fore wing relatively small in size, slit-like, with short vein RS + r – m; tergite 2 0.6–0.7 × as long as tergite 3 . . . *Diolcogaster xanthaspis* (Ashmead, 1900)

Type material. Holotype. Female, "Museu Regional de Entomologia – UFV", 20° 45′ S × 42° 51′ W, 651 m above sea level, 18.V.2011, Wagner de Souza Tavares & José Cola Zanuncio. Holotype locality: Viçosa, Minas Gerais State, Brazil. Holotype label: WAG01.18.05.2011. Host plants: *C. spicatus* and *C. spiralis* var. *spiralis*. Host caterpillar: *A. minuta*. **Paratypes.** Same locality as for holotype, five females and five males (all in "Museu Regional de Entomologia – UFV", except one female and one male in Canadian National Collection of Insects, Arachnids, and Nematodes (CNC),

Fig. 2. (A) Female paratype. (B) Head, ventral view. (C) Head, frontal view. (D) Fore and hind wings. (E) Propodeum and metasomal tergites, dorsal view. (F) Mesosoma, dorsal view. (G) Miasma, lateral view. (H) Metasoma, dorsal view of *Diolcogaster flammeus* sp. nov. (Hymenoptera: Braconidae).

Ottawa, Ontario. Labels: Brazil, Minas Gerais, Viçosa-Minas Gerais, 2012, ex. *Agarea minuta*, coll. G. Salgado-Neto, voucher codes: CNC649489, CNC649490).

Etymology. The specific epithet, *flammeus*, comes from Latin, referring to the species color and means "flame".

Biology. A solitary parasitoid, reared from *A. minuta*. The information available suggests that it is extremely efficient as a natural enemy. Last larval instar of the parasitoid emerged from *A. minuta* caterpillars and pupated at the bottom of the plastic container on filter paper in the laboratory. Parasitoid larvae produced a cocoon made of white colored silk. The silk was wrapped around the leaves causing them to fold and become cone-shaped. Parasitized caterpillars survived only long enough to build an extra protection layer for the parasitoids and then died. Caterpillars of different ages were found in a same sampling which suggests ovipositions at different times and overlapping generations.

Distribution. So far only found in Viçosa, Minas Gerais State, Brazil; the municipality is inland, 300 km north of Rio de Janeiro State and west of the coast by 250 km.

Discussion

The fauna of the *xanthaspis* group in the New World now is represented by four species, including *D. bakeri*. A study recording the *D. bakeri* from Peru is questionable (sensu Fernández-Triana et al., 2009) because it was reported previously as a North American parasitoid species (Muesebeck, 1922). *Diolcogaster* has been at times divided in species-groups (e.g., Saeed et al., 1999; Whitfield et al., 2009; Zeng et al., 2011). One of such groups, the *xanthapsis* group (sensu Saeed et al., 1999), is characterized by having the head, anteromesoscutum and scutellar disk rugose-punctate, mediotergite 1 parallel-sided and 2.0–3.0 × as long as maximum width, mediotergite 2 with more or less distinct median field indicated by grooves (if not, then median field smooth and convex, distinguished by longitudinal pits), vannal lobe of hind wing convex with marginal setae, and postocellar vertex concave in dorsal view (Nixon, 1965; Mason, 1981). This group was originally erected by Nixon (1965: 240), who considered it "an artificial segregation within a large complex of species". Morphological differences and

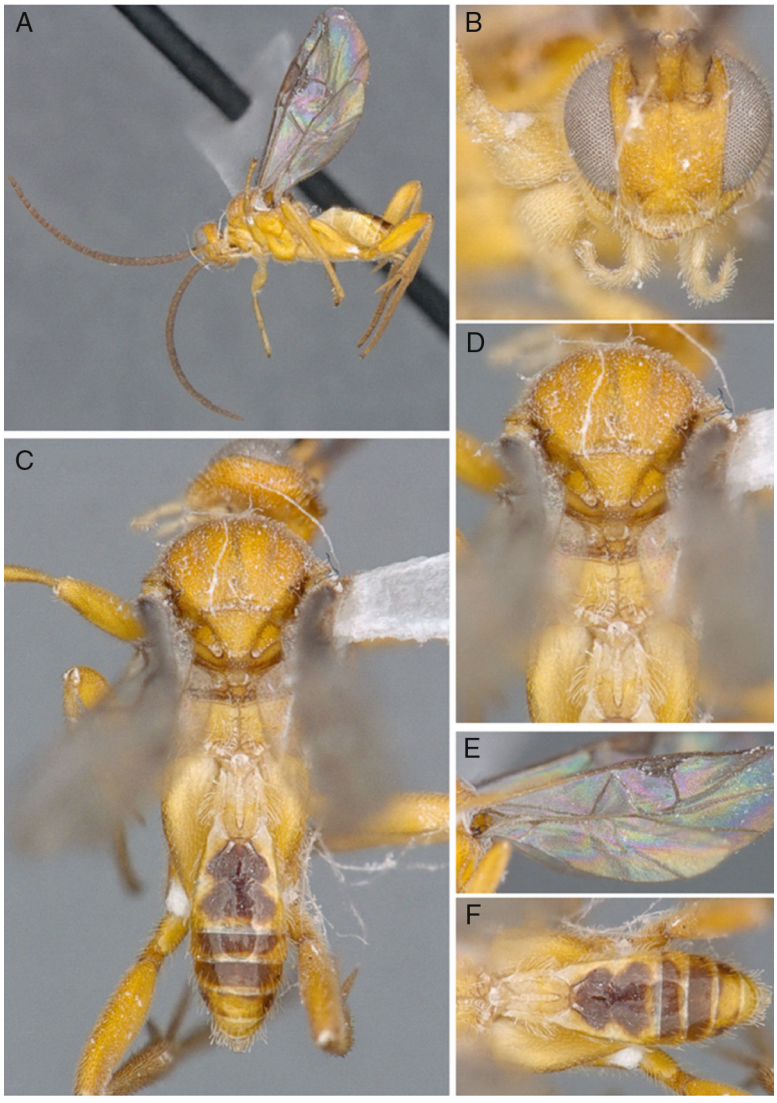

Fig. 3. (A) Male paratype. (B) Head, frontal view. (C) Habitus, dorsal view. (D) Propodeum and metasomal tergite, dorsal view. (E) Fore wing. (F) Metasoma, dorsal view of *Diolcogaster flammeus* sp. nov. (Hymenoptera: Braconidae).

Table 1
Host caterpillar (Lepidoptera) (author; family) and its geographical distribution, host plant (author; family), and reference of species of *Diolcogaster* (Hymenoptera: Braconidae) (author).

Species of *Diolcogaster*	Host caterpillar	Geographical distribution	Host plant	Reference
D. flammeus Salgado-Neto & Fernández-Triana sp. nov.	*Agaraea minuta* (Schaus, 1892; Erebidae)	Mexico to Brazil	*Costus spicatus* (Jacq.) Sw. and *C. spiralis* (Jacq.) Roscoe var. *spiralis*; Costaceae	Present study
D. iridescens (Cresson, 1865)	*Asciodes gordialis* Guenée, 1854; Crambidae	Cuba and southern USA (Florida)	Species of *Bougainvillea*; Nyctaginaceae	Patton (1958)
D. bakeri (Muesebeck, 1922)	*Ponometia candefacta* (Hübner, [1831]) and *P. erastrioides* (Guenée, 1852; Noctuidae)	Canada (Ontario, Quebec and Saskatchewan), USA (Arkansas, Florida, Georgia, Illinois, Iowa, Kansas, Louisiana, and Texas)	Asteraceae including species of *Aster* and *Ambrosia*	Pogue (2010) and Stojanović et al. (2011)
D. xanthaspis (Ashmead, 1900)	Unknown	Saint Vincent and the Grenadines	Unknown	Ashmead (1900)

the disparate arrange of hosts known for the described species, indeed suggest that this group is far from monophyletic (Whitfield et al., 2009), and it is very likely that it will be split in the future when more studies of the world fauna of *Diolcogaster* are done. *Diolcogaster flammeus* sp. nov. is extremely efficient as a natural enemy, as it was recorded parasitizing 90% of the *A. minuta* caterpillar pests collected in 2010 and 2011 (Tavares et al., 2012). In the field, pupation of the parasitoid occurred on the abaxial surfaces of the leaves of *C. spicatus* and *C. spiralis* var. *spiralis* (Tavares et al., 2012). *Agarea minuta* is a severe pest of *Costus* species because it can reduce plant biomass (stem heights, leaves per stem, largest width and greater length of leaves) (Tavares et al., 2012).

Conflicts of interest

The authors declare no conflicts of interest.

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