PENSOFT. SENCKENBERG world of biodiversity Leibniz Association

Integrating morphology and DNA barcodes for identification of *Delia sanctijacobi* (Diptera: Anthomyiidae): new host and new records in South America

Lucas Roberto Pereira Gomes[1], Maria Aparecida Cassilha Zawadneak[2], Magda Clara Vieira da Costa-Ribeiro[2], Tiago Miguel Jarek[3], Claudio José Barros de Carvalho[1]

1 *Universidade Federal do Paraná, Departamento de Zoologia, Av. Cel. Francisco H. dos Santos, 100 - Jardim das Americas, Curitiba - PR, 81530-000, Brazil*

2 *Universidade Federal do Paraná, Departamento de Patologia Básica, Brazil*

3 *Universidade Federal do Paraná, Departamento de Fitotecnia e Fitossanitarismo, Brazil*

http://zoobank.org/634A7EB8-0E64-495D-A0AA-939E51A32866

Corresponding author: Lucas Roberto Pereira Gomes (lucaspergos@gmail.com)

Received 27 February 2022
Accepted 08 July 2022
Published 27 September 2022

Academic Editors Anna Hundsdörfer, Klaus-Dieter Klass

Citation: Gomes LRP, Zawadneak MAC, da Costa-Ribeiro MCV, Jarek TM, de Carvalho CJB (2022) Integrating morphology and DNA barcodes for identification of *Delia sanctijacobi* (Diptera: Anthomyiidae): new host and new records in South America. Arthropod Systematics & Phylogeny 80: 511–522. https://doi.org/10.3897/asp.80.e82831

Abstract

Delia sanctijacobi is critically assessed and given a revised description using data from scanning electronic microscopy (SEM) and DNA barcode analysis. This species is recorded for the first time in Brazil and Peru. We provide a morphological identification key (with figures) for *Delia* species from Brazil, a molecular identification based on COI (cytochrome C oxidase subunit I) barcode sequences and an updated distributional map. We also report the first occurrence of *D. sanctijacobi* feeding on *Brassica* species in Brazil. This potential pest was observed in broccoli roots (*Brassica oleracea* var. *italica*; Brassicaceae) in União da Vitória, Paraná, southern Brazil, in August and September of 2017. The infested plants displayed reduced growth due to damage to the stem base or death if severely attacked.

Keywords

Brassicaceae, cruciferous vegetables, DNA barcode, insect pest, Neotropical Region, plant–insect interaction, root maggot flies

1. Introduction

Root maggot flies (*Delia* Robineau-Desvoidy, 1830) (Diptera: Anthomyiidae) are polyphagous, and some species, such as *D. platura* Meigen 1826, are considered important agricultural pests in innumerable plant species of commercial interest, causing important crop losses (Griffiths 1986; Gouingauen and Städler 2006; Meraz-Álvarez et al. 2020). *Delia* is a diverse and unclear genus whose species are found mainly in subalpine and subarctic areas of the Palearctic and Nearctic regions (Griffiths 1986). Three agricultural pest species, *Delia antiqua* Meigen, 1826, *D. platura* and *D. radicum* Linnaeus, 1758, are found in Brazil (de D'Araujo e Silva et al. 1968; de Carvalho and Couri 2018); however, all of these records could be cases of misidentification and need to be revised. Here, we present the first records of *D. sanctijacobi* (Bigot, 1885) from southern Brazil (25 records) and Peru (a record from Cuzco). Other *Delia* species, such as *D. radicum* and *D. floralis,* have a wide host range, including most cultivated crucifers, and wild crucifers such as stinkweed (*Thlaspi arvense* Linnaeus, 1753) are potential hosts (Strickland 1938; Griffiths 1991).

Delia sanctijacobi is native to South America and is suspected to have been introduced, probably through human dispersion with Chilean vegetables, to coastal localities in western Alaska (Cold Bay and Unalakleet) (Griffiths 1993). *D. sanctijacobi* larvae are commonly known as "potato worms", as the adults are attracted by decaying fruits and lay their eggs near cuts in potatoes or seeds (Hamity and Roman 1987). This species attacks the seeds of corn, bean (cotyledons), pumpkin, melon, wheat, flax, garlic, sunflower, and cauliflower (*Brassica* cv.), as well as the seedlings of tomato, eggplant, and onion (Molinari 1942; Quintanilla 1969; Griffiths 1993). The largest population is found in the spring (Hamity and Roman 1987). Some *D. sanctijacobi* specimens used in this study were found feeding on broccoli (*Brassica oleracea* L. var. *italica*), an important vegetable crop of the family Brassicaceae (Kumar and Srivastava 2016).

Delia sanctijacobi and *D. platura* are similar species (Hamity and Roman 1987; Griffiths 1993). Hennig (1955) considered *D. sanctijacobi*, previously known as the subspecies *D. platura sanctijacobi*, to be a South American lineage of *D. platura*, indicating that *D. platura* probably does not occur in South America. Griffiths (1993) described some male morphological differences in the chaetotaxy of the legs and terminalia; however, differences between *D. platura* and *D. sanctijacobi* females have not been established (Griffiths 1993). Wang et al. (2014) presented scanning electronic microscope (SEM) images of different structures of *D. platura*, including male and female genitalia. Gomes et al. (2018) presented a key to "Muscoidea" species from Juan Fernández Archipelago, including *D. sanctijacobi* and *D. platura*. Gomes et al. (2021) presented a phylogenetic analysis of Anthomyiidae based on molecular data, including three species of *Delia*, and recovered *D. radicum* + (*D. platura* + *D. sanctijacobi*).

Delia pest species can be difficult to identify without extensive training, mainly when dealing with females or immature specimens, which lack diagnostic features (Savage et al. 2016). In addition, *Delia* species are very similar and genital characters are necessary to make a correct identification (Griffiths 1993). Savage et al. (2016) provided keys (with images) to the different life stages of *Delia* pests from Canada (including the three species also found in Brazil, namely, *D. antiqua*, *D. platura* and *D. radicum*). These authors also provided a reference DNA barcode for each of these species.

In this present contribution, we report the first record of *Delia sanctijacobi* in Brazil and Peru. We provide a morphological redescription of *D. sanctijacobi*, an updated distributional map of *Delia* species and an identification key for adults. Additionally, we provide DNA barcode sequences as an undoubtedly molecular taxonomy tool for the identification of this economically important species. Finally, we report the first occurrence of *D. sanctijacobi* feeding on cultivated *Brassica* species in Brazil.

2. Material and methods

2.1. Material

We examined 1064 specimens (573 males and 491 females) from Argentina, Brazil, Chile, Peru and Uruguay, including 46 locations, deposited in **DZUP** – Department of Zoology, Universidade Federal do Paraná and **MNRJ** – Museu Nacional, Universidade Federal do Rio de Janeiro.

2.2. Methods

2.2.1. Dissections and terminology

For the examination of the terminalia, the abdomen was removed from a dry specimen and placed in cold potassium hydroxide (KOH) 10% for 24 h to soften and lighten the parts. The abdomen was transferred to acetic acid, and then to glycerin. Examination and illustration of the structures were performed using a microscope and a stereomicroscope with an attached camera lucida. Dissected terminalia were placed in glycerin in microvials pinned beneath the respective specimens. For the examination of terminalia by SEM, the terminalia was removed from each specimen, dehydrated using 99.5% ethanol, glued on cooper tape and coated with gold-palladium. The dissected parts were placed in a plastic microvial with glycerin and pinned with the respective specimen. The morphological terminology followed Cumming and Wood (2017).

2.2.2. Images and measurements

Images were stacked using an auto-montage setup acquired by the Taxonline project (UFPR – http://www.taxonline.bio.br). Scanning electronic microscope (SEM) images were taken using a JEOL JSM 6360-LV at Centro de Microscopia Eletrônica, Curitiba, Paraná, Brazil (UFPR – http://www.cme.ufpr.br/). The QGIS software (available in: http://www.qgis.org/en/site) was used to create maps of distribution. Geographical coordinates were found at Google Maps (https://www.google.com.br/maps/) based on the place name (location) available on the labels and in the literature. The locations from Argentina based on Hamity and Roman (1987) (Santa Fe, Tucumán, Córdoba, Cuyo and Jujuy), have no location-specific information, and their estimated coordinates were placed at each state center.

Table 1. Details of *Delia* species and outgroup processed in the study along with the BOLD/GenBank numbers. *Newly collected COI sequences; [B]BOLD accession number; [G]GenBank acession number.

Species	Accession Number
Botanophila spinidens (Malloch, 1920)[G]	KM627639; KM625462; KM631112; KM857756; KM634039
Delia antiqua (Meigen, 1826)[B]	BUICD289-15; BUICD300–15; BUICD312-15; BUICD328-15; BUICD549-15; BUICD365-15
Delia floralis (Fallen, 1824)[B]	BUICD216-15; BUICD217-15; BUICD218-15; BUICD219-15
Delia florilega (Zetterstedt, 1845)[B]	BUICD066-15; BUICD206-15; BUICD292-15; BUICD340-15; BUICD355-15; BUICD626-15; PAMA333-13; PAMA335-13
Delia platura (Meigen, 1826)[B]	AOTW075-14; BUICD194-15; BUICD262-15; BUICD265-15; BUICD204-15; BUICD270-15; BUICD274-15; BUICD277-15; BUICD279-15; BUICD308-15; BUICD475-15; BUICD541-15; BUICD593-15; BUICD607-15; UICD191-15; UICD250-15
Delia radicum (Linnaeus, 1758)[B]	BUICD293-15; BUICD307-15; BUICD332-15; BUICD379-15; BUICD559-15
Delia sanctijacobi (Bigot, 1885)[G]	MT808064.1*; MT808207.1*; MT808208.1*; MT808209.1*

Table 2. Pairwise of sequence divergent within *Delia* species and outgroup based on Kimura-2-parameter model (K2P).

Species	n	Bspi	Dant	Dfls	Dflg	Dpla	Drad	Dsan
Bspi	5	**0.011**						
Dant	6	0.123	**0.000**					
Dfls	4	0.112	0.094	**0.000**				
Dflg	8	0.124	0.060	0.081	**0.005**			
Dpla	16	0.125	0.072	0.088	0.058	<u>**0.047**</u>		
Drad	5	0.119	0.090	0.067	0.084	0.092	**0.000**	
Dsan	4	0.115	0.060	0.088	<u>0.048</u>	0.055	0.090	**0.002**

The numbers of intraspecific distances are shown in boldface for clarity. Numbers underlined indicate the highest intraspecific distance and the lowest interspecific distance. n = Number of sequences. Bspi = *Botanophila spinidens*; Dant = *Delia antiqua*; Dfls = *Delia floralis*; Dflg = *Delia florilega*; Dpla = *Delia platura*; Drad = *Delia radicum*; Dsan = *Delia sanctijacobi*.

2.2.3. DNA extraction, amplification, sequencing and barcode analysis

DNA extraction was conducted through a nondestructive method using the whole specimen with body perforations and the GenElute™ Blood Genomic DNA Kit. The complete sequence primers used were LCO-1490f: 5’ GGT CAA CAA ATC ATA AAG ATA TTG G 3’, and HCO-2198r: 5’ TAA ACT TCA GGG TGA CCA AAA AAT CA 3’ (Folmer 1994). The MyTaq™ DNA Polymerase kit (Bioline Reagentes Ltd, the United Kingdom) was used for PCR amplification. The 25 µL reaction consisted of 17.5 µL of sterilized ultrapure water, 5 µL 5x MyTaq Reaction Buffer (comprising 15 mM MgCl2 and 5 mM dNTPs), 0.5 µL of each primer, 0.2 µL of MyTaq DNA Polymerase, and 1.0 µL of DNA extracted from the specimen. The PCR amplification conditions were as follows: 94°C for 5 min, 39 cycles at 94°C for 45 s, 51°C for 50 s and 72°C for 2 min, and a final extension at 72°C for 10 min. The PCR products were visualized on a 1.0% agarose gel. PCR products were sent to WenSeq Pesquisa e Desenvolvimento Ltda. (Curitiba) for sequencing.

Neighbor-joining (NJ; Saitou and Nei 1987) was chosen as the best-performing method for low sample sizes among the available phylogenetic, simple distance-based and supervised statistical classification methods (Austerlitz et al. 2009). The sequencing was aligned using the ClustalW procedure (Thompson et al. 1994) using MEGA7 (Kumar et al. 2016). The NJ and ML trees were constructed using the Kimura-2-Parameter model (K2P) (Kimura 1980) by MEGA7, with uniform rates among sites. The bootstrap consensus tree was generated with 1000 replicates (Felsenstein 1985). The ML heuristic method was nearest-neighbor-interchange (NNI). For both analyses the ingroup was composed of 39 COI sequences (ranging from 595 to 658 base pairs) from five *Delia* species, all of which were deposited in the BOLD database (The Barcode of Life Data System, http://www.barcodinglife.org) and previously used by Savage et al. (2016), and four newly collected COI sequences from *D. sanctijacobi* from two locations (Palmas and União da Vitória). We used five sequences of *Botanophila spinidens* (Malloch, 1920) (all previously available in GenBank) as the outgroup. All sequences used in this study are presented in Table 1. Pairwise distances within and between species were calculated using Kimura’s 2-parameter (K2P) distance model in MEGA7 (Kumar et al. 2016) (Table 2).

2.3. Abbreviations

Morphology. **a** – anterior surface, **d** – dorsal surface, **p** – posterior surface, **v** – ventral surface, **ad** – anterodorsal surface, **av** – anteroventral surface, **pd** – posterodorsal surface, **pv** – posteroventral surface, **dm-m** – discal medial crossvein, Terminalia: **cerc** – cercus, **distph** – distiphallus, **epand** – epandrium, **epiph** – epiphallus, **epiprct** – epiproct, **hypd** – hypandrium, **hyprct** – hypoproct, **pgt** – postgonite, **phapod** – phallapodeme, **pregt** –

pregonite, **spmth** – spermatheca, **st** – sternite, **sur** – surstylus, **tg** – tergite.

Depositories. DZUP – Department of Zoology, Universidade Federal do Paraná; **MNRJ** – Museu Nacional, Universidade Federal do Rio de Janeiro; **USNM** – National Museum of Natural History, Washington, USA.

3. Results

3.1. DNA Barcode analyses

To confirm the validity of *D. sanctijacobi*, we used 39 mitochondrial COI gene sequences from five *Delia* species from BOLD Systems (www.boldsystems.org/) and included four newly collected COI sequences from *D. sanctijacobi*. Five sequences from *Botanophila spinidens* from GenBank (https://www.ncbi.nlm.nih.gov/genbank/) were used as the outgroup (Table 1). Similar topologies were observed in both COI sequence analyses. The only difference was related to *D. florilega*, which was recovered as sister group of *D. sanctijacobi*, and *D. platura* on ML and recovered as sister group of *D. antiqua* on NJ (Fig. 1). According to the pairwise distances values of K2P, *D. sanctijacobi* has a low intraspecific distance (0.002) and a high interspecific divergence (0.048–0.090). Among the *Delia* species, *D. sanctijacobi* has the highest similarity to *D. florilega* (0.048). The conspecific K2P divergence of *Delia* species ranges from 0.000 to 0.047. The sequence divergence between species ranges from 0.048 to 0.094 (Table 2).

3.2. Taxonomy

Delia Robineau-Desvoidy, 1830

Diagnosis. Eye usually bare; frons usually narrow in male and broad in female; with pairs of inter frontal setae; legs black or yellow; fore tibia with 0–1 ad, 1–2 medial p or 1–2 pv and 1 apical pv; mid tibia with 1 pd; hind femur without pv row or apical pv; hypopygium with fine or weak setae; surstylus longer than cercus, not bifurcate apically; aedeagus slender, apical part with distiphallus in most species. Female: tergum 5 usually without discal setae (Huckett 1971; Du and Xue 2017, listed by various authors).

3.3. Identification key

Key to male *Delia* species from Brazil (modified from Savage et al. 2016)

1 Prealar setae at least as long as notopleural setae; ventral surface of costal vein (C) setulose, with hairs reaching at least the insertion point of vein R_1 .... ***D. radicum*** (Linnaeus, 1758)

1’ Prealar setae absent or very short, never as long as notopleural setae (Fig. 2A); ventral surface of costal vein (C) bare or with a few sparse hairs distal from the insertion point of the subcostal vein (Sc) (Fig. 2B) **2**

2 Hind tibia with 7–15 short erect posteroventral setae; frontal vitta visible at narrowest point of frons (Fig. 2C); parafacial broad in lateral view (Fig. 2D) .... ***D. antiqua*** (Meigen, 1826)

2’ Hind tibia with >18 short erect posteroventral setae (Fig. 2E, F); frontal vitta usually obliterated at narrowest point of frons (Fig. 3A); parafacial narrow in lateral view (Fig. 2A) **3**

3 First tarsomere of midleg without a brush of long dorsal bristles, only with regular setae (Fig. 2G); row of pv setae on hind tibia with similar length of tibia width (Fig. 2E) .... ***D. platura*** (Meigen, 1826)

3’ First tarsomere of midleg with a brush of long dorsal setae (Fig. 2H); row of pv setae on hind tibia longer than tibia width (Fig. 2F) .... ***D. sanctijacobi*** (Bigot, 1885)

3.4. *Delia sanctijacobi* (Bigot, 1885)

Figs 2–5

Delia sanctijacobi (Bigot, 1885:296; *Anthomyia*). Lectotype male, UMO. Type-locality: “Chile”.

Diagnosis. Male. First tarsomere of midleg with a brush of long setulae (longer than tibia width) on its dorsal surface. Row of 32–45 pv setae on hind tibia longer than tibia width. Sternite 5 processes each with 2–3 truncate spinules on inner apical corner. Cercus approximately 3 times as long as wide with 6 long apical setae. *D. sanctijacobi* genitalia is quite similar to *D. platura*, except postgonite slightly longer than in *D. platura* (similar to postgonite length) and distally narrowed in lateral view. Surstylus 1/3 more enlarged than *D. platura*.

Redescription. Measurements: Male. Body length 4.5–5.0 mm; wing length 4.7–5.3 mm (Fig. 3B). — ***Head***: Eye sparsely and shortly setulose; frontal vitta dark brown; eyes separated by less than width of anterior ocellus (Fig. 3A); frontal setae 5–8 pairs; short interfrontal setae usually present; fronto-orbital plate and parafacial brownish or yellowish-orange, with gray pruinosity; antenna dark-brown, postpedicel approximately 2.0 times longer than wide; arista short pubescent; gena with gray pruinosity, genal height approximately 3 times

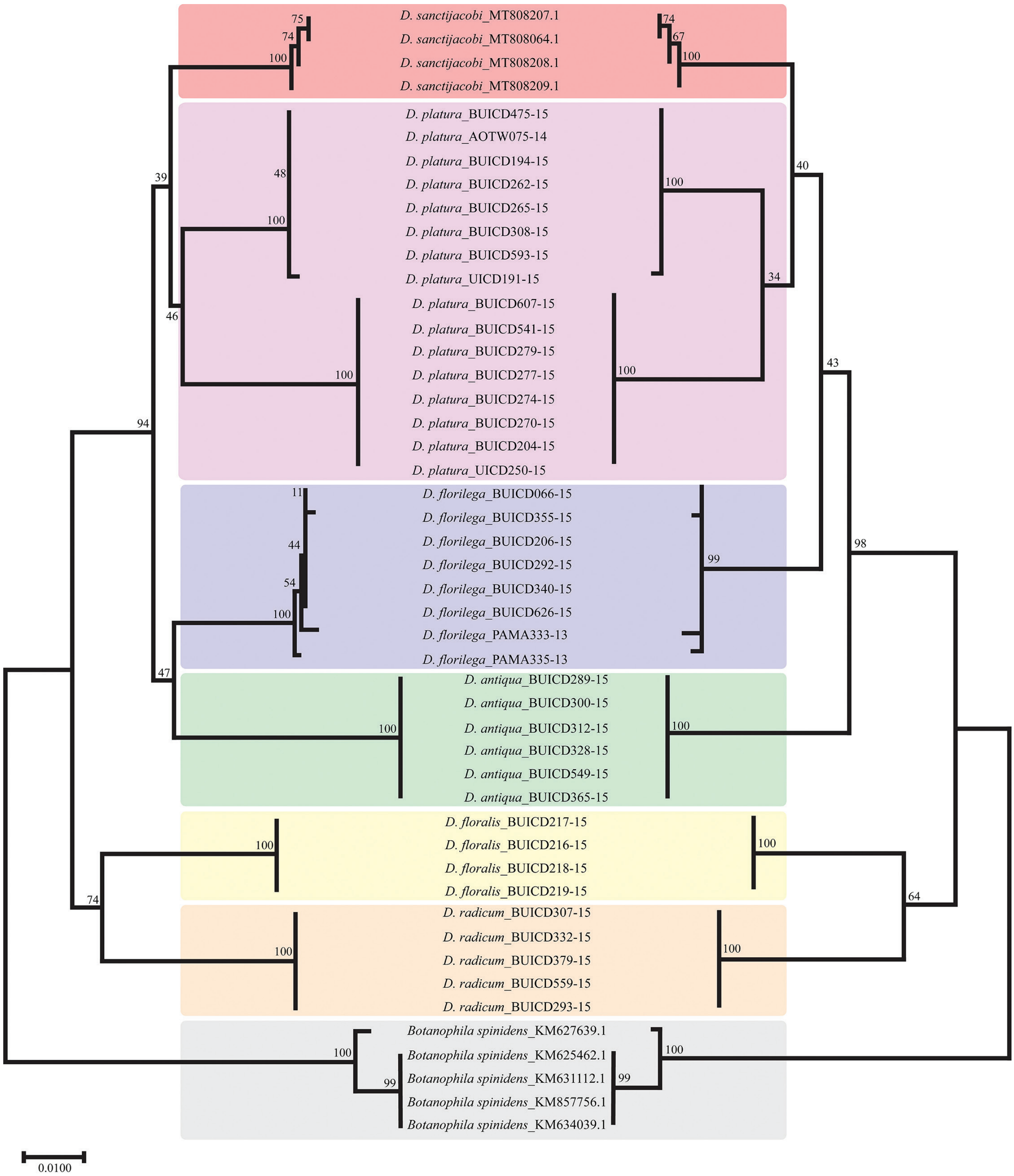

Figure 1. Neighbour joining (NJ) and Maximum likelihood (ML) tree of Delia based in COI sequences. Numbers on branches indicate the bootstrap support values (1000 replicates). *D. sanctijacobi* – red box; *D. platura* – violet box; *D. florilega* – blue box; *D. antiqua* – green box; *D. radicum* – orange box; *Botanophila spinidens* – gray box. Evolutionary distance divergence scale bar: 0.01.

the pedicel length (Fig. 3B); anterior margin of gena with 2–4 upcurved subvibrissal setae; palpus dark-brown. — ***Thorax***: Light-brown with gray pruinosity; scutum distinct with 3 black vittae visible at acrostichals region and among dorsocentral and intra-alars (Fig. 3C); acrostichal setae 3+6–9, dorsocentral setae 2+3, intra-alar setae 1+2, prealar seta about half length of posterior notopleural seta; scutellum light-brown, ventral margins with white hairs apically, basal scutellar setae and apical scutellar setae developed; notopleuron with two same length setae and without covered setulae; prosternum, anepimeron, meron and katepimeron bare; katepisternal setae 1+2. — ***Legs***: Entirely dark-brown; fore tibia with 1 medial pv, preapical d with half-length of first tarsomere, and apical seta on v; mid femur with a row of setae on ad and av, a row of long (longer than femur width) pv in basal half; mid tibia with 1 pd, 1–2 pv and 1 submedian ad, long (longer than tibia width) apical setae on a, d, pv, v, first tarsomere of midleg with a brush of long setulae (longer than tibia width) on its dorsal surface; hind femur with a

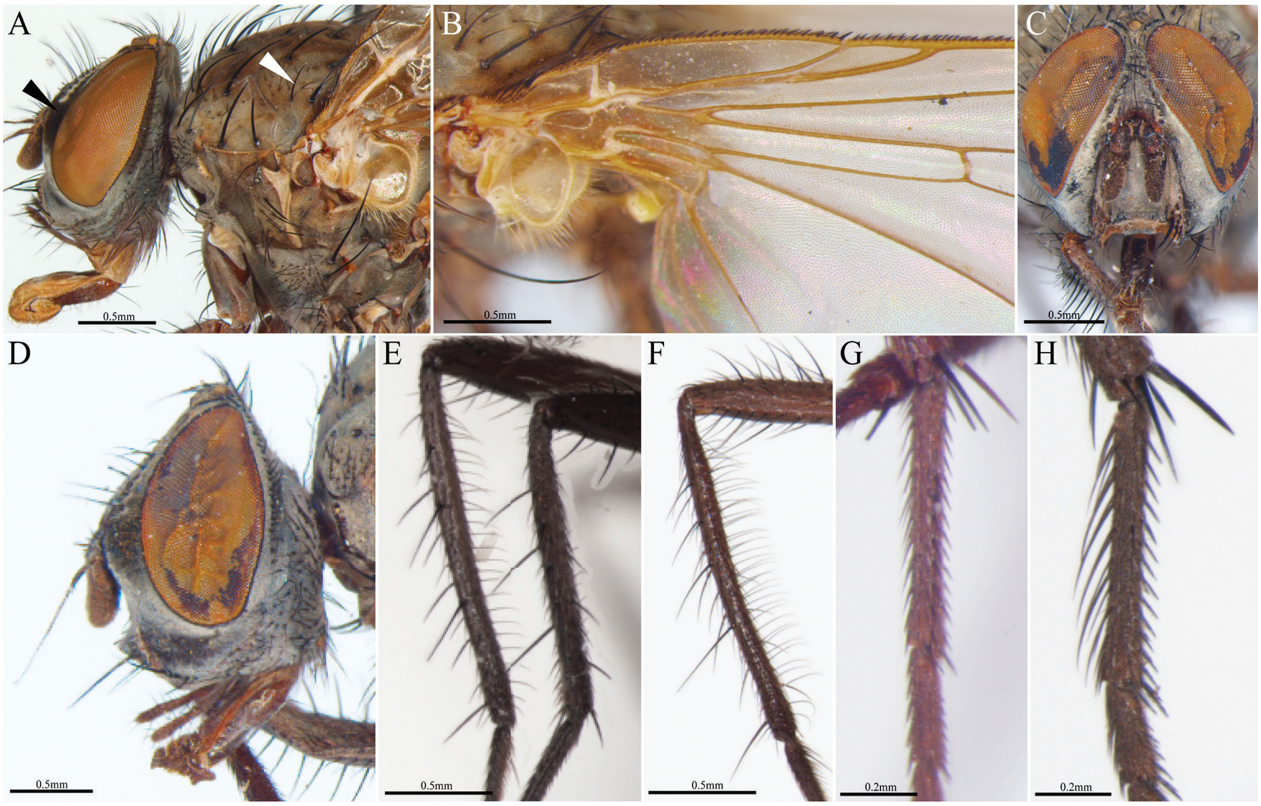

Figure 2. *Delia sanctijacobi*. **A** parafacial (black arrow), prealar (white arrow), lateral view. **B** wing, ventral view; *Delia antiqua*. **C** head, frontal view. **D** head, lateral view; *Delia platura*. **E** hind tibia; *Delia sanctijacobi*. **F** hind tibia; *Delia platura*. **G** first tarsomere of the midleg; *Delia sanctijacobi*. **H** first tarsomere of the midleg.

complete row on ad becoming long apically, a basal half row on pv, an apical half row on av and d; hind tibia with 3–5 av, 7–11 ad, 4 pd, becoming longer apically, 32–45 pv with slightly bent tips in largely duplicated rows, a preapical d, apical setae on av. Claws black and pulvillus white. — ***Wing***: Veins bare, except costal; costal with all spinules short (pair before distal break twice longer), ventrally bare; dm-m slightly sinuate; calypteres yellow, marginal hairs long and light yellow, lower calypter developed, but smaller than upper; halter yellow. — ***Abdomen***: Slender, with gray pruinosity and a dark stripe dorsally, dense and long setulose, marginal setae longer (approximately 1/3 of abdomen length) (Fig. 3C); sternite 1 with long dense setulae (longer than sternite length); sternite 4 with a pair of long setae (longer than sternite length); sternite 5 processes projecting, with outer lateral setae becoming longer distally, each with 2–3 truncate spinules on inner apical corner (Fig. 4A). — ***Terminalia***: pregonite with 2 setae; postgonite with 1 seta; cercus approximately 3 times as long as wide, elongate-ovoid (broadest on distal half) with 6 long setae distally and 4 long lateral setae on each side; surstyli slender and almost twice longer than cercus, phallapodeme straight; epiphallus short, similar to postgonite length; distiphallus long, three times longer than pregonite, bifurcate distally (Figs 4C; 5A, B).

Female. Measurements: Body length 4.0–5.5 mm; wing length 4.5–5.5 mm. Similar to male, except: Frons approximately 1/3 of head width (Fig. 3D); frontal vitta approximately 5 times as wide as fronto-orbital plate; a pair of interfrontal setae; region among interfrontal setae and lunula yellowish-orange; 5 pairs of frontal setae; outer vertical seta long and divergent; inner vertical seta long and convergent; ocelar, outer vertical seta and inner vertical seta with similar length; fronto-orbital plate and parafacial with brownish-gray pruinosity; body with brownish-gray pruinosity (Fig. 3E, F); fore tibia with 1 medial ad; mid tibia with 2 median ad, pd and pv; pre-apical setae on ad, av, pd and pv; hind tibia with 2–3 av and 5–7 ad; pv bare; abdomen not slender (Fig. 3E, F). — ***Terminalia***: tergites 6–8 reduced to pairs of lateral plates; spiracles 6 and 7 situated on posterior half of segment 6; sternite 6 and 7 long and narrow, bearing a pair of long and few shorter setae posteriorly; sternite 8 represented by a pair of small plates, each bearing 5 setae; epiproct trapezoid, with a long distal pair of setulae; hypoproct triangular covered by setulae; cerci setulose, twice longer than epiproct; three round spermathecae (Figs 4E, F; 5C–F).

Remarks. The female of *D. sanctijacobi* is morphologically indistinguishable from *D. platura*. Their identity must be confirmed by DNA barcoding. The ultrastructural morphology of male and female genitalia revealed a high similarity between the species. No difference was observed in the SEM images of *D. sanctijacobi* presented here (Fig. 3) and the images of *D. platura* presented by Wang et al. (2014), except that the *D. sanctijacobi* epiphallus is twice as long as that of *D. platura* (Fig. 3B).

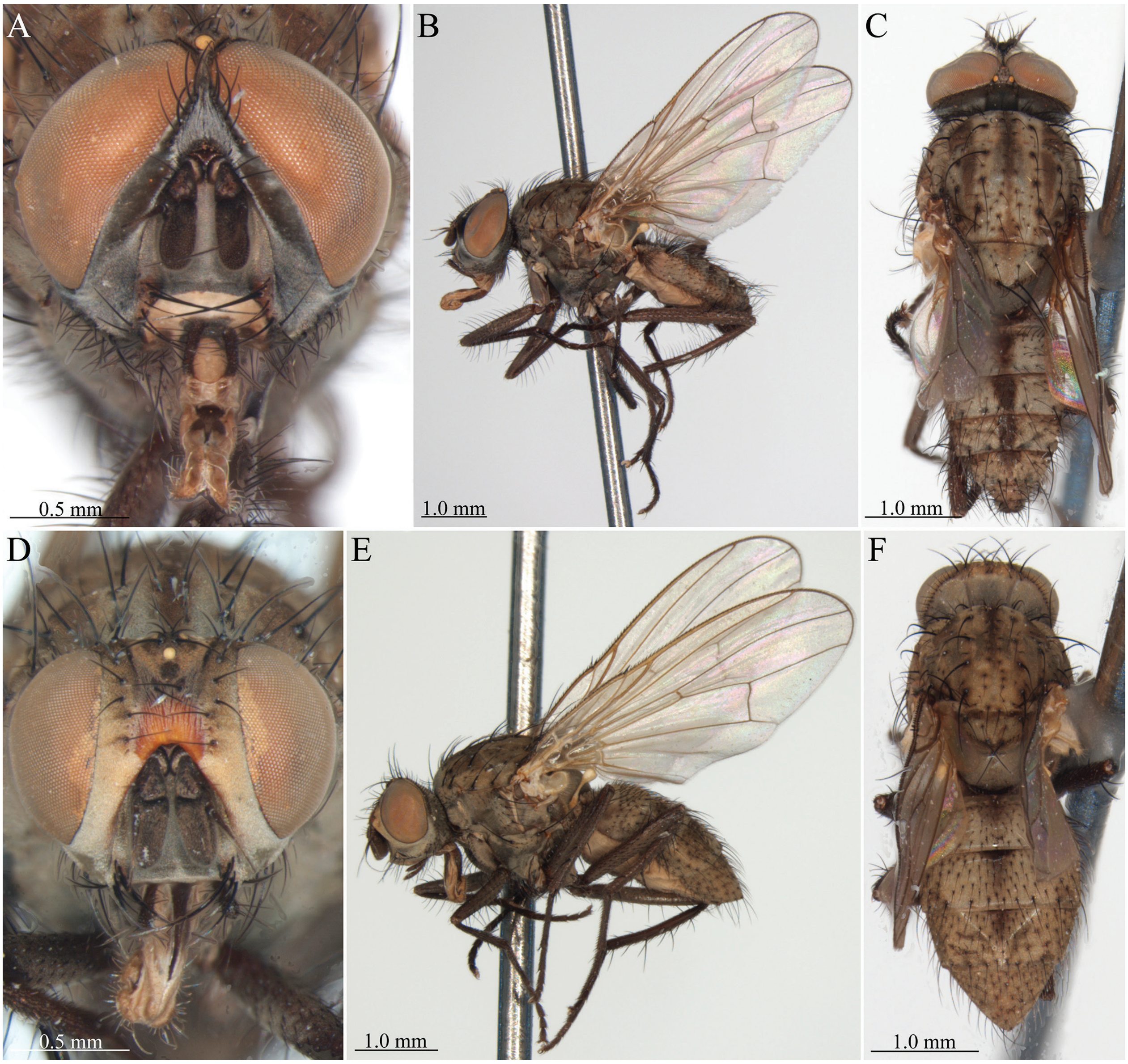

Figure 3. Male *Delia sanctijacobi* (Bigot, 1885). **A** head, anterior view. **B** lateral view. **C** dorsal view. Female *Delia sanctijacobi* (Bigot, 1885). **D** head, anterior view. **E** lateral view. **F** dorsal view.

Life history. According to the label information, some specimens were found in soy, onion and broccoli. Here, we report some larvae feeding on the roots of broccoli (*Brassica oleracea* L. var. *italica* (Brassicaceae).

Distribution. Alaska (Cold Bay and Unalakleet), Argentina (Jujuy, Tucumán, Córdoba, Santa Fe, Mendoza, Neuquén and Río Negro), Brazil (Paraná, Santa Catarina, Rio Grande do Sul), Chile (Juan Fernández Islands, Los Lagos, Valparaíso and Araucanía), Peru (Cuzco) and Uruguay.

Material examined. ARGENTINA: La Martona, Prov. B. aires, xi.1957, H. S. Lopes, 1♂ and 3♀♀, (MNRJ); BRAZIL: Paraná: Cambé, 29.ix.1981, Hamada, 2♂♂ and 3♀♀ (DZUP); Campo do Tenente, 19.x.1985, [no col.], 1♂ and 4♀♀; Castro, 11.v.1961, N. L. Marston, 1♀; Colombo, BR476, KM20, 01.x.1986, Lev. Ent. PROFAUPAR, 1♂ and 3♀♀; same locality and collector: 01.xi.1986, 3♂♂; 01.xii.1986, 2♂♂; 02.ix.1986, 3♂♂ and 35 ♀♀; 02.x.1986, 4♂♂ and 2♀♀; 02.xi.1986, 5♂♂; 02.xii.1986, 1♂; 03.ix.1986, 6♂♂ and 7♀♀; 03.viii.1986, 1♂; 03.x.1986, 6♂♂ and 19♀♀; 03.xi.1986, 7♂♂ and 2♀♀; 04.ix.1986, 13♂♂ and 45♀♀; 04.viii.1986, 1♂; 04.x.1986, 4♂♂ and 10♀♀; 04.xi.1986, 4♂♂ and 1♀; 05.ix.1986, 1♀; 05.x.1986, 7♂♂ and 6♀♀; 06.viii.1986, 2♂♂; 08.xi.1981, B. B. Santos, 2♀♀ (DZUP); Curitiba, 19.x.1982, R. Misiuta, 1♂; same locality: 29.xi.1970, P. Moure, 1♂; 19.i.1984, M.L. Pilato Silva & A.C. Saad, onion decay, 1♀; Curitiba, C. B. Jesus, x.1980, 1♂ and 1♀; same locality and collector: i.1981, 1♂ and 3♀♀; ii.1981, 1♀; iv.1981, 2♀♀; v.1981, 1♀; vii, 1980, 1♀; x.1980, 1♂ and 1♀; xii.1980, 4♀ (DZUP); Curitiba, Uberaba, Imbiriba [col.], 21.i.1976, 1♂ (DZUP); Guarapuava, Est. Águas Sta. Clara, Lev. Ent. PROFAUPAR, 01.ix.1986, 1♂; 01.x.1986, 1♂ and 3♀♀; 01.xi.1986, 6♂♂ and 8♀♀; 02.ix.1986, 31♂♂ and 28♀♀; 02.x.1986, 17♂♂ and 2♀♀; 02.xi.1986, 1♂ and 2♀♀; 02.xii.1986, 1♀; 03.ix.1986, 51♂♂ and 16♀♀; 03.viii.1986, 24♂♂ and 16♀♀; 03.x.1986, 6♂♂ and 2♀♀; 03.xi.1986, 4♂♂ and 1♀; 03.xii.1986, 1♀; 04.ix.1986, 2♂♂; 04.viii.1986, 1♂ and 14♀♀; 04.x.1986, 1♂ and 4♀♀; 04.xi.1986, 3♂♂ and 2♀♀; 05.ix.1986, 8♀♀; 05.viii.1986, 31♂♂ and 25♀♀; 05.x.1986, 8♂♂ and 10♀♀; 06.ix.1986, 12♂♂ and 3♀♀; 06.vii.1986, 5♂♂;

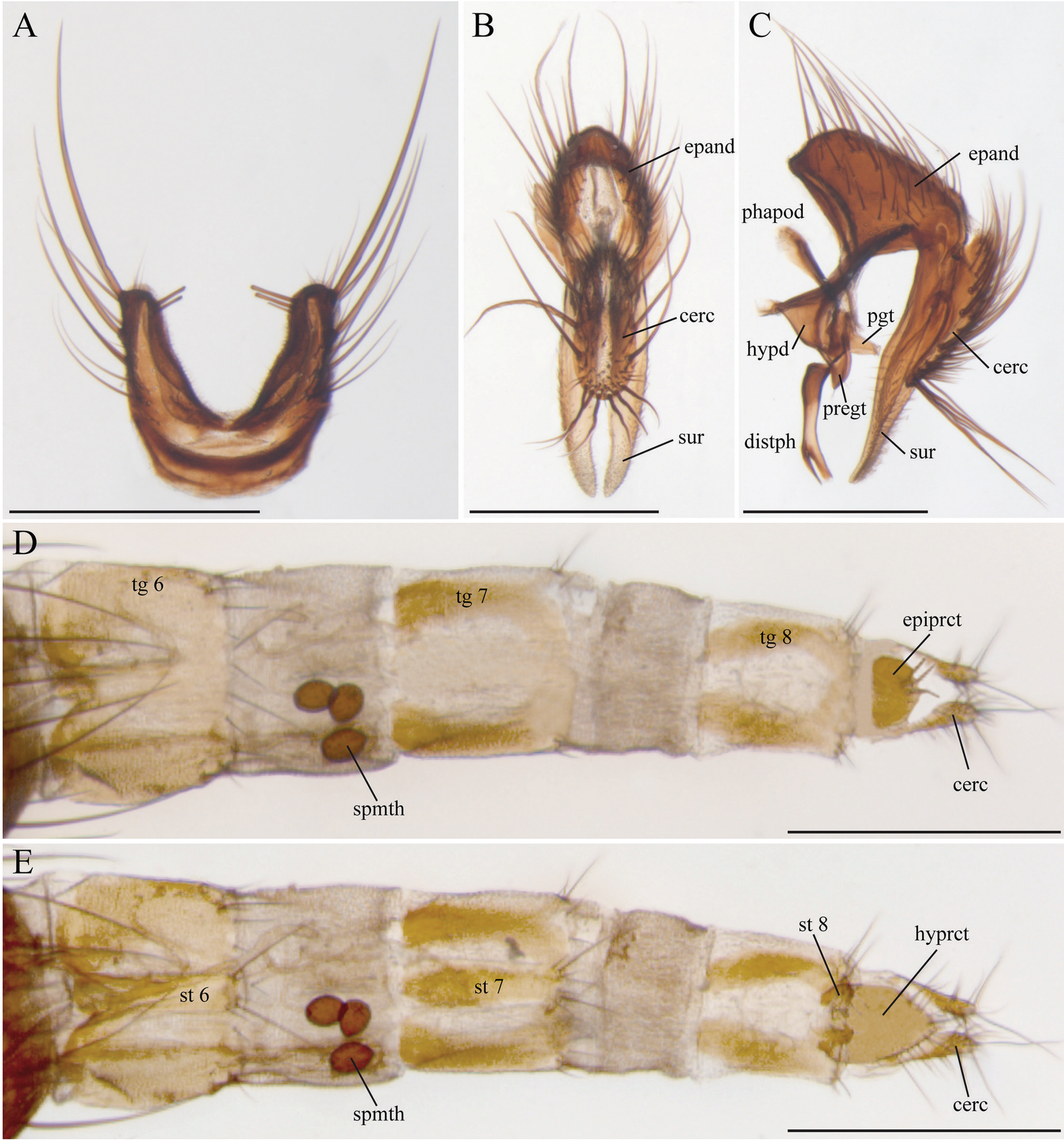

Figure 4. Male *Delia sanctijacobi* (Bigot, 1885). **A** sternite 5, dorsal view. **B** epandrium, cercus and surstyli, dorsal view. **C** epandrium, cercus and surstyli, lateral view. **D** aedeagus and associated structures, lateral view. Female *Delia sanctijacobi* (Bigot, 1885). **E** ovipositor, dorsal view. **F** ovipositor, ventral view (Scale: 0.5 mm). Abbreviations: cerc – cercus, distph – distiphallus, epand – epandrium, epiprct – epiproct, hyprct – hypoproct, pgt – postgonite, phapod – phallapodeme, pregt – pregonite, spmth – spermathecae, st – sternite, sur – surstylus, tg – tergite.

06.viii.1986, 31♂♂ and 14♀♀; 06.x.1986, 1♂; 07.viii.1986, 14♂♂ and 4♀♀; 25.viii.1986, 1♀; 30.xi.1986, 2♂♂ and 1♀; 31.x.1986, 2♂♂ and 3♀♀ (DZUP); Jundiaí do Sul, Fazenda Monte Verde, Lev. Ent. PROFAUPAR, 04.x.1986, 1♂ (DZUP); Lapa, 24.x.1981, E. Silveira, in soy, 3♂♂ and 1♀ (DZUP); Palmas, Refúgio de Vida Silvestre dos Campos de Palmas, Cerro Chato, Adriana C. Pereira, malaise trap, 09.ix.2014, 4♂♂ and 4♀♀ (DZUP); Palmas, Refúgio de Vida Silvestre dos Campos de Palmas, Pinus area, Adriana C. Pereira, malaise trap, 21.ix.2012, 4♂♂ and 4♀♀ (DZUP); Ponta Grossa, 02.xii.1986, [no collector], 6♂♂ and 5♀♀ (DZUP); Ponta Grossa, Vila Velha, Reserva IAPAR BR376, Lev. Ent. PROFAUPAR, 01.xi.1986, 6♂♂ and 2♀♀; same locality and collector: 02.xi.1986, 14♂♂ and 6♀♀; 03.viii.1986, 8♂♂; 03.xi.1986, 2♂♂; 04.viii.1986, 1♂; 04.xi.1986, 4♂♂; 05.viii.1986, 1♂; 10.xi.1986, 1♀; 31.x.1986, 64♂♂ and 66♀♀; 01.xii.1986, 1♂ and 2♀♀; 02.x.1986, 1♂ and 1♀; 04.x.1986, 1♂ and 3♀♀; 05.ix.1986, 3♂♂ (DZUP); Ponta Grossa, Vila Velha (IAPAR), Ganho & Marinoni, malaise, 04.x.1999, 2♂♂; 06.ix.1999, 1♀; 08.xi.1999, 2♂♂; 11.x.1999, 1♂ and 1♀; 20.ix.1999, 1♂ and 1♀; 22.xi.1999, 2♂♂; 27.ix.1999, 1♀; 28.viii.2000, 3♂♂ and 3♀♀; 29.xi.1999, 1♂ (DZUP); S. Antonio da Platina, N. Papavero, vi.1965, 1♀ (DZUP); S. José dos Pinhais, Serra do Mar Br277, Km 54, Lev. Ent. PROFAUPAR, 01.x.1986, 2♀♀; same locality and collector: 01.xi.1986, 6♂♂ and 5♀♀; 01.xii.1986, 2♂♂ and

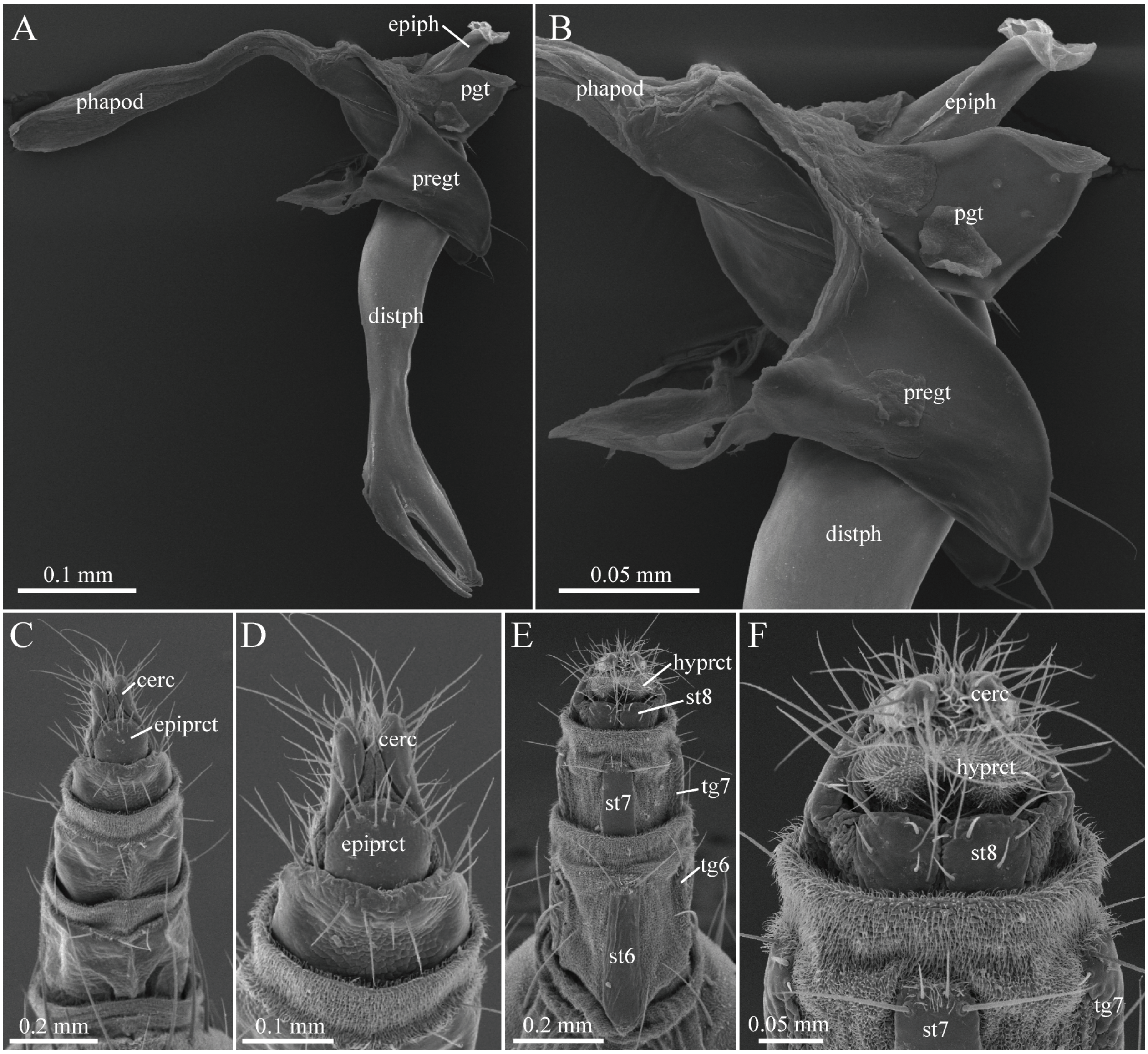

Figure 5. *Delia sanctijacobi* (Bigot, 1885). SEM micrographs: Male. **A** aedeagus and associated structures, lateral view (phallapodema curved by technical artefacts). **B** detailed pregonite and postgonite, lateral view. Female. **C** ovipositor, dorsal view. **D** ovipositor, detailed epiproct, dorsal view. **E** ovipositor, ventral view. **F** ovipositor, detailed hypoproct, ventral view. Abbreviations: cerc – cercus; distph – distiphallus; epiph – epiphallus; epiprct – epiproct; hyprct – hypoproct; pgt – postgonite; phapod – phallapodema; pregt – pregonite; st – sternite; tg – tergite.

2♀♀; 02.x.1986, 2♀♀; 02.xi.1986, 7♂♂ and 2♀♀; 02.xii.1986, 8♂♂ and 8♀♀; 03.viii.1986, 1♂; 03.x.1986, 6♂♂ and 2♀♀; 03.xii.1986, 1♂ and 1♀; 04.ix.1986, 1♂; 04.viii.1986, 1♀; 04.xi.1986, 11♂♂ and 3♀♀; 05.viii.1986, 1♀; 05.x.1986, 2♂♂ and 1♀; 29.xi.1986, 14♂♂ and 4♀♀; 31.x.1986, 10♂♂ and 4♀♀; 17–24.ix.1984, 1♀; 21–27.viii.1984, 2♂♂ and 1♀; 24.ix–01.x.1984, 1♀ (DZUP); União da Vitória, in broccoli, T. M. Jarek, 25.viii.2017, 3♂♂ and 1♀ (DZUP); Rio Grande do Sul: Arroio Grande, Distrito Mauá, 04.x.2002, R. F. Krüger, malaise trap, 1♂ (DZUP); Canela, 12–16.i.1984, Hoffmann, M., malaise trap, 1♀ (DZUP); Quaraí, Estância S. Roberto, J. R. Cure, 19–20.xi.1985, 1♂ and 4♀♀; same locality and collector: 22.xi.1985, 1♀; 21.xi.1985, 2♂♂ and 6♀♀ (DZUP); Santa Catarina: Ituporanga, 13.x.1988, M. J. Carissimi, 3♂♂ (DZUP); Ituporanga, 20.ix.1978, O. Nakano, 2♂♂ and 3♀♀ (DZUP); Lages, 01.viii.2012, Rosa, J. M. da, 5♂♂ and 4♀♀ (DZUP); Lages, 10.xii.1984, H. Kalvelage, 2♂♂ and 2♀♀ (DZUP); Nova Teutônia, 300–500 m, Fritz Plaumann, x.1967, 1♂ and 3♀♀; same locality and collector: xi.1967, 4♂♂ and 1♀ (DZUP); São Joaquim, Margem Rio Lava-tudo, 1191m, 02.xi.2006, A.J.C. Aguiar, A. Martins, L.R.R. Faria Jr., 1♂; São Joaquim, Pericó, 1174 m, 02.xi.2006, A.J.C. Aguiar, A. Martins, L.R.R. Faria Jr., 1♂ and 2♀♀ (DZUP); CHILE: Angol, Malleco, 16–21.xi.1970, T. Cekalovic, malaise trap, 2♂♂ and 1♀; same locality and collector: 23–28.xi.1970, 1♂ and 1♀ (DZUP); PERU: Cuzco: Carretera Manum dirt side road, ravine, WPB 34, 3296 m, A. L. Norrbom, G. J. Steck & B. D. Sutton, 28.i.2013, 1♂ (USNM); URUGUAY: Colonia, La Estanzuela, S. Laroca, 27.i.1992, 1♂ (DZUP); Montevideo, Parker Berry Silveira, 5.viii.1943, 1♂ (MNRJ).

3.5. Damage on crops

We report the first occurrence of the *Brassica* root fly, *Delia sanctijacobi*, larvae feeding on cultivated *Brassica* species in Brazil. This potential pest was observed in August and September of 2017 in União da Vitória

Figure 6. Geographic distribution of *Delia sanctijacobi* (Bigot, 1885). Symbols: white circle – new records; black circle – literature records.

(–26.212381; –51.077950), Paraná, southern Brazil. The specimens were collected from broccoli roots (*Brassica oleracea* L. var. *italica* (Brassicaceae).

3.6. Distribution

The type-locality of *D. sanctijacobi* and other localities lacking specific information were not used in assembling the distributional map. The Hamity and Roman (1987) localities (Córdoba, Cuyo, Jujuy, Santa Fé and Tucumán) were estimated by placement in each state center. The map was assembled using 48 localities, including 30 new records. In addition to new records from Argentina, Chile and Uruguay, the first records from Brazil (Paraná, Santa Catarina and Rio Grande do Sul) and Peru (Cuzco) are shown (Fig. 6). Specimens from DZUP and MNRJ were analyzed, and a large number of them had been misidentified as *D. platura* instead *D. sanctijacobi*; therefore, all *D. platura* records in South America require review.

The 48 locations indicated a distribution restricted to the cold regions of South America. The lower latitude area corresponds to Cuzco (Peru), with an altitude of 3296 m. We also present one new record each for Argentina and Chile, three new records for Uruguay and 25 new records for southern Brazil, indicating a wide distribution in this Brazilian region of agricultural importance (Fig. 6). Some changes in the *Delia* identification presented by Gomes et al. (2018; 2019) have resulted from this work. We reanalyzed the specimens of Gomes et al. (2018) from Juan Fernández Archipelago previously identified as *D. platura* and updated the identification to *D. sanctijacobi*. Also, we reanalyzed the specimens of Gomes et al. (2019) checklist (identified as *Delia* sp.) from Palmas (Paraná, Brazil) and updated the identification as *Delia sanctijacobi*.

4. Discussion

4.1. DNA Barcode analysis

COI sequences analyses using NJ and ML indicated that *D. sanctijacobi* is closely related to *D. florilega* and *D.*

platura (interspecific distances of 0.048 and 0.055, respectively). The topology using NJ was consistent with the analysis of Savage et al. (2016). The only difference among the topologies constructed with NJ and ML was for *D. florilega* (Fig. 1, blue box), which was recovered as sister group of *D. platura* (Fig. 1, violet box) + *D. sanctijacobi* (Fig. 1, red box) on ML and recovered as sister group of *D. antiqua* (Fig. 1, green box) on NJ. In both analyses, *D. sanctijacobi* was recovered as sister group of *D. platura* with bootstrap values of 39 and 40 in NJ and ML, respectively (Fig. 1). The bootstrap values for phylogenetic relationships among *D. florilega*, *D. platura* and *D. sanctijacobi* are low (less than 47), but all species clusters had bootstrap values of 100. The NJ topology is the same as obtained by Savage et al. (2016).

The pairwise distance values from the K2P distance model suggest that *D. sanctijacobi* is a valid species, as indicated by high interspecific divergence (0.048-0.090) among other species of the genus. The high intraspecific distance of *D. platura* (0.047) could be interpreted as evidence that we are dealing with a species complex (Table 2), as also indicated by Heyden et al. (2020).

4.2. Damage on crops

The first occurrence of *Brassica* root flies, *Delia sanctijacobi,* feeding on cultivated *Brassica* species in Brazil and the resulting damage was observed in 2017; however, our review of specimens from the DZUP and MNRJ collections indicated that this species has occurred in Brazil at least since 1961, as indicated by a male specimen from Castro (Paraná, Brazil), and deposited in DZUP. This specimen is probably a neotropical species often misidentified as *D. platura*. The infested plants showed reduced growth due to damage of the stem base. Due to damage to the vascular system, the plants wilt, and if they are severely attacked, they can die. Damage symptoms were more evident at times of higher temperature and insolation.

4.3. Distribution

Delia sanctijacobi is native to southern South America and was described based on two males from Chile; however, more specific geographic information about the type-locality is absent (Bigot 1885). Few authors reported distributional information about *D. sanctijacobi*, and extant distributional knowledge was restricted to Argentina (Córdoba, Jujuy, Mendoza, Neuquén, Río Negro, Santa Fe, Tucumán); Chile (Araucanía, Juan Fernández Islands, Los Lagos and Valparaíso) and Uruguay (Montevideo) (Malloch 1934; Ruffinelli and Carbonell 1954; Hennig 1955; 1957; Hamity and Roman 1985; Griffiths 1993). Also, *D. sanctijacobi* has been found in coastal locations in western Alaska (Cold Bay and Unalakleet), suspected to have been introduced with Chilean vegetables (Griffths 1993).

5. Conclusions

New morphological and molecular data for *Delia sanctijacobi* allow the appropriate identification of this economically important species. *D. sanctijacobi* larvae can cause severe damage to cruciferous crops. Although the potato worm was recorded as new to Brazil in 2017, our review of specimens from collections indicates this species has occurred in several regions of South America for at least five decades, and it is probably an endemic neotropical species of *Delia*. For this reason, previous South American records of *D. platura*, which is a species complex, should be reviewed. The correct identification and updating of *D. sanctijacobi* distribution in South America are important to infestation control of this species in crops.

6. Authors' contributions

LRPG wrote the main text, reviewed the specimens deposited in the collections, prepared the images, extracted, amplified and analysed the COI sequences. MACZ and TMJ collected the specimens. MCVCR helped with extraction and amplification of DNA sequences. CJBC helped with the COI sequence analysis and taxonomy. All authors contributed to improve the manuscript and approved the final manuscript.

7. Acknowledgements

The authors are very grateful to Allen Norrbom (USNM) for sending us part of specimens used in this contribution. We thank Taxonline – Rede Paranaense de Coleções Biológicas (UFPR) for specimen photographs and Centro de Microscopia Eletrônica (UFPR) for Scanning electronic microscope (SEM) images. Thanks to the Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPq) for the provided support (CJBC project # 309873/2016–9, LRPG project # 141030/2018–6). Thanks to "Divisão de Vigilância Prevenção e Controle de Pragas-DPCP, Ministério da Agricultura, Pecuária e Abastecimento" (MAPA) for permits (project # 21034.009554/2019-73). Wiley Services edited the manuscript.

8. References

D'Araujo e Silva AG, Gonçalves CR, Galvão DM, Gonçalves JL, Gomes J, do Nascimento Silva M, de Simoni L (1968) Quarto Catálogo dos Insetos que Vivem nas Plantas do Brasil seus Parasitos e Predadores. Ministério da Agricultura, Rio de Janeiro, Brasil, 622 pp.

Austerlitz F, David O, Schaeffer B, Bleakley K, Olteanu M, Leblois R, Veuille M, Laredo C (2009) DNA barcode analysis: a comparison of phylogenetic and statistical classification methods. BMC Bioinformatics 10: 1–10. https://doi.org/10.1186/1471-2105-10-S14-S10.

Bigot JMF (1885) Diptères nouveaux ou peu connus. 25e partie. XXXIII. Anthomyzides nouvelles. Annales de la Société Entomologique de France 4 [1884]: 263–304.

de Carvalho CJB, Couri MS (2018) Anthomyiidae. Catálogo Taxonômico da Fauna do Brasil. PNUD http://fauna.jbrj.gov.br/fauna/faunadobrasil/90563 [accessed on 25.08.2018].

Cumming JM, Wood DM (2017) 3. Adult morphology and terminology. In: Kirk-Spriggs AH, Sinclair BJ (Eds) Manual of Afrotropical Diptera, vol. 1. Introductory chapters and keys to Diptera families. Suricata 4, SANBI Graphics & Editing, Pretoria, 89–133.

Du J, Xue W (2017). Four new species of the genus *Delia* Robineau-Desvoidy in the Yunnan Province of China (Diptera, Anthomyiidae), Zookeys 693: 141–153. https://doi.org/10.3897/zookeys.693.12965

Felsenstein J (1985) Confidence limits on phylogenies: An approach using the bootstrap. Evolution 39: 783–791. https://doi.org/10.1111/j.1558-5646.1985.tb00420.x.

Folmer O, Black M, Hoeh W, Lutz R, Vrijenhoek R (1994) DNA primers for amplification of mitochondrial cytochrome c oxidase subunit I from diverse metazoan invertebrates – Molecular Marine Biology and Biotechnology 3: 298–299.

Gomes LRP, Couri MS, de Carvalho CJB (2018) Anthomyiidae, Fanniidae and Muscidae (Diptera) from the Juan Fernández Archipelago (Chile): 60 years after Willi Hennig's contributions. Zootaxa 4402: 373–389. https://doi.org/10.11646/zootaxa.4402.2.9

Gomes LRP, Fogaça JM, Bortolanza M, Pereira AC (2019) New records of the Brazilian Anthomyiidae (Diptera) and a checklist of species from Palmas Grasslands Wildlife Refuge. Check List 15: 93–103.

Gomes LRP, Souza DS, de Carvalho CJB (2021) First insights into the evolution of neotropical anthomyiid flies (Diptera: Anthomyiidae). Systematics and Biodiversity 19: 724–737. https://doi.org/10.1080/14772000.2021.1914765.

Gouinguené SP, Städler E (2006) Oviposition in *Delia platura* (Diptera, anthomyiidae): The role of volatile and contact cues of bean. Journal of Chemical Ecology 32: 1399–1413. https://doi.org/10.1007/s10886-006-9058-3.

Griffiths GCD (1986) Relative abundance of the root maggots *Delia radicum* (L.) and *D. floralis* (Fallén) (Diptera; Anthomyiidae) as pests of canola in Alberta. Questiones Entomologicae 22: 253–260.

Griffiths GCD (1991) Anthomyiidae [part]. In: Griffiths GCD (Ed) Flies of the Nearctic Region, 8(2), 7, E. Schweizerbart, Stuttgart, Germany, 953–1048.

Griffiths GCD (1993) Anthomyiidae [part]. In: Griffiths GCD (Ed) Flies of the Nearctic Region, 8(2), 10, E. Schweizerbart, Stuttgart, Germany, 1417–1632.

Heyden HV, Fortier A-M, Savage J (2020) A HRM Assay for Rapid Identification of Members of the Seedcorn Maggot Complex (*Delia florilega* and *Delia platura*) (Diptera: Anthomyiidae) and Evidence of Variation in Temporal Patterns of Larval Occurrence. Molecular Entomology 113: 2920–2930. https://doi.org/10.1093/jee/toaa230.

Hamity MGA, Roman LEN (1987) Anthomyiidae de interes Agricola en distintos cultivos de altura de Jujuy. Aparicion Estacional Y mayor abundancia. Revista de la Sociedad Entomológica Argentina 44: 411–418.

Hennig W (1955) Los Insectos de Las Islas Juan Fernandez. Revista Chilena de Entomologia 4: 21–34.

Hennig W (1957) Los Insectos de Las Islas Juan Fernandez. Revista Chilena de Entomologia 5: 409–412.

Huckett HC (1971) The Anthomyiidae of California exclusive of the subfamily Scatophaginae (Diptera). Bulletin of the California Insect Survey 12: 1–121.

Kimura M (1980) A simple method for estimating evolutionary rate of base substitutions through comparative studies of nucleotide sequences. Journal of Molecular Evolution 16: 111–120.

Kumar S, Stecher G, Tamura K (2016) MEGA7: Molecular Evolutionary Genetics Analysis version 7.0 for bigger datasets. Molecular Biology and Evolution 33: 1870–1874.

Kumar P, Srivastava DK (2016) Biotechnological advancement in genetic improvement of broccoli (*Brassica oleracea* L. var. *italica*), an important vegetable crop. Biotechnology Letters 38: 1049 pp. https://doi.org/10.1007/s10529-016-2080-9.

Malloch JR (1934) Muscidae. In: Diptera of Patagonia and South Chile. Part VII. Fascicle 2. Muscidae. British Museum (Natural History), London, 171–346.

Meraz-Álvarez R, Bautista-Martínez N, Illescas-Riquelme CP, González-Hernández H, Valdez-Carrasco JM, Savage J (2020) Identification of Delia spp. (Robineau-Desvoidy) (Diptera, Anthomyiidae) and its cruciferous hosts in Mexico. ZooKeys 964: 127–141.

Molinari OC (1942) Entomologia Agrícola. Ed. D'Accurzio, Mendoza, 575 pp.

Quintanilla RH (1969) Principales causas adversas de origen animal, en los cultivos de la República Argentina. Actas de las I Jornadas Fitosanitarias Argentinas, Universidad Nacional La Plata, 35–52.

Ruffinelli A, Carbonell CS (1954) Segunda lista de insectos y otros artrópodas de importancia económica en el Uruguay. Revista de la Asociación de Ingenieros Agronomos, Montevideo 94: 33–82.

Saitou N, Nei M (1987) The neighbor-joining method: a new method for reconstructing phylogenetic trees. Molecular Biology and Evolution 4: 406–425. https://doi.org/10.1093/oxfordjournals.molbev.a040454.

Savage J, Fortier A, Fournier F, Bellavance V (2016) Identification of *Delia* pest species (Diptera: Anthomyiidae) in cultivated crucifers and over other vegetable crops in Canada. Canadian Journal of Arthropod Identification 29: 1–40. https://doi.org/10.4039/Ent83109-5

Strickland EH (1938) An annotated list of the Diptera (flies) of Alberta. Canadian Journal of Research, Section D, Zoological Sciences 16: 175–219.

Thompson JD, Higgins DG, Gibson TJ (1994) "CLUSTAL W: improving the sensitivity of progressive multiple sequence alignment through sequence weighting, position-specific gap penalties and weight matrix choice". Nucleic Acids Research 22: 4673–4680.

Wang QK, Yang YZ, Liu MQ, Zhang D (2014) Fine Structure of *Delia platura* (Meigen) (Diptera: Anthomyiidae) Revealed by Scanning Electron Microscopy. Microscopy Research and Technique 77: 619–630. https://doi.org/10.1002/jemt.22380