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New records of *Pachycrepoideus vindemmiae* (Hymenoptera: Pteromalidae) associated with *Drosophila suzukii* (Diptera: Drosophilidae) in cherry and berry crops from Argentina

Nuevos registros de *Pachycrepoideus vindemmiae* (Hymenoptera: Pteromalidae) asociados con *Drosophila suzukii* (Diptera: Drosophilidae) en cultivos de cerezas y bayas de Argentina

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Abstract: New records of the ectoparasitoid *Pachycrepoideus vindemmiae* (Rondani) (Hymenoptera: Pteromalidae) associated with *Drosophila suzukii* Matsumura (Diptera: Drosophilidae) in the provinces of Tucumán, Río Negro and Neuquén are cited. Detections occurred in blueberry, raspberry and cherry crops. The records from Río Negro and Neuquén constitute the southernmost report of the species. Diagnosis and prevalence of the parasitoid are briefly discussed.

Keywords: Biological control, Small fruits, Spotted-wing *Drosophila*, Parasitoid.

Resumen: Se citan nuevos registros del ectoparásitoide *Pachycrepoideus vindemmiae* (Rondani) (Hymenoptera: Pteromalidae) asociado a *Drosophila suzukii* Matsumura (Diptera: Drosophilidae) en las provincias de Tucumán, Río Negro y Neuquén. Las detecciones ocurrieron en cultivos de arándano, frambuesa y cereza. Los registros de Río Negro y Neuquén constituyen el reporte más austral de la especie. Se comentan brevemente la diagnosis y la prevalencia del parasitoide.

Palabras clave: Control biológico, Frutas finas, Mosca de las alas manchadas, Parasitoide.

From about 1,500 *Drosophila* Fallén species known worldwide (Markow & O'Grady, 2006), *D. suzukii* Matsumura (Diptera: Drosophilidae), known as "cherry drosophila" in Japan and "spotted-wing drosophila" (SWD) worldwide, and *D. pulchrella* Sturtevant (also native to Japan) are the only species that oviposit in healthy fruits, with the consequent economic damage. The rest of the *Drosophila* species attacks only damaged or overripe fruit (Sasaki & Sato, 1996). This characteristic, together with its high fertility rate, wide host range, high dispersal capacity and adaptation to different climatic conditions, make SWD a high-risk pest.

Drosophila suzukii is native to the southeast of the Palearctic region, but in the last decade invaded Europe and North America (Walsh et al., 2011; Cini et al., 2012, 2014; Asplen et al., 2015). More recently, it was found in regions with different climatic characteristics of South America (Bitner-Mathé et al., 2014; Deprá et al., 2014; dos Santos, 2014; Paula et al., 2014; Vilela & Mori, 2014). In Argentina, it was initially found in raspberry crops in the province of Río Negro (Cichón et al., 2015) and in blueberry crops in the province of Buenos Aires (Santadino et al., 2015). In the province of Entre Ríos (northeast region), the pest was detected in traps in the summer 2014-15 (Díaz et al., 2015), and in the province of Tucumán (northwest region), it was found both in traps and in fruit in blueberry crops, in 2015 (Funes et al., 2018). Considering that the northeast and northwest regions are the main blueberry producers and exporters of the country, SWD presence represents an imminent economic threat for northern Argentina.

In view of the described above, and due to a limited availability of insecticides authorized to control SWD, there is a need to deepen the development of biorational alternatives for its control. Biological control is one of them, and its implementation would reduce the use of pesticides, avoiding resistance phenomena and other secondary risks derived from improper pest management (Guédez et al., 2008). A first step to assess the possibility of implementing biological control would be the survey of potential natural enemies, and in this sense, parasitoids associated with local drosophilids play a fundamental role (Funes et al., 2019)



Fig. 1. Side view of *Pachycrepoideus vindemmiae* collected in Argentina.
Bar = 0.5 mm

Frugivorous drosophilids are attacked mainly by larval and pupal parasitoids. Pupal parasitoids are ecto or endoparasitoids and tend to be generalist, attacking a wide range of hosts. Larval parasitoids, on the other hand, are endoparasitoids that interact with the host's immune system and tend to have a narrow host range (Carton et al., 1986; Fleury et al., 2009). Around 16 genera and 30 species of parasitoid wasps develop in *Drosophila* spp. in the world, being *Asobara* (Braconidae), *Ganaspis*, *Leptopilina* (Figitidae), *Pachycrepoideus* (Pteromalidae), and *Trichopria* (Diapriidae) the most relevant genera. *Pachycrepoideus* and *Trichopria* are worldwide distributed; *Asobara* species were found in Asia, Europe and North America, while *Ganaspis* and *Leptopilina* are spread in Asia, Europe, and North and South America (Carton et al., 1986; Asplen et al., 2015; Kirschbaum et al., 2020).

In Argentina, six genera of parasitoids were registered associated with *D. suzukii*: *Ganaspis*, *Leptopilina*, *Hexacola* and *Dieucoila* (Figitidae), *Trichopria* (Diapriidae) and *Pachycrepoideus* (Pteromalidae) (Escobar et al., 2018; Gallardo et al., 2018), confirming the presence of the species *Leptopilina clavipes* Hartig, *Ganaspis hookeri* Crawford (Lue et al., 2017), and *L. boulandi* Barbotin, Carton & Kelner-Pillaut (Garrido et al., 2018).

In recent years, in the Patagonian region and almost simultaneously, in the province of Tucumán (in blueberry crops), the frequency of occurrence of a species belonging to the Pteromalidae family allowed inferring its possible relevance in SWD trophic relationships. In this context, the objective of this work was to identify taxonomically specimens of the Pteromalidae family found in Argentina, and to study their prevalence in berry agroecosystems.

In the province of Tucumán, a SWD trapping network was established in representative locations of the blueberry producing area, such as the departments of Lules, Monteros and Chichigasta. In each of them, a farm was selected as a study unit, where three lots were chosen. In each lot, three to four traps were placed on a diagonal (transect), totaling ten traps

per farm, during 2016 harvesting season (August 3rd and December 7th). Between December 16th and March 31st (2017), the trap number *per farm* was reduced to four, and relocated to the surroundings of the farms, with the purpose of knowing the behavior of the pest in the period in which blueberry plants were not producing fruit.

Modified Mc Phail traps were used for the capture of small-sized insects (mesh with 0.5 mm diameter holes), which were placed at the height of the fruiting canopy and protected from direct sunlight. As a food attractant, a solution consisting of apple cider vinegar (250 mL) + water (100 mL) and two drops of detergent, was used to break the surface tension. Traps were weekly or biweekly revised and serviced, during the spring-summer season and the autumn-winter period, respectively.

In the provinces of Río Negro and Neuquén, a cherry and a raspberry orchard were selected, respectively. Sampling was conducted from January to May 2017. For cherries, the harvest season had finished already but not for raspberries. Trap revision and service was made biweekly. Traps were placed in plants located in the central part of the orchard, in the middle part of the plant and facing north. Each trap consisted of a 1.5 L transparent, plastic bottle with twelve 0.5 mm-diameter holes in the center, containing a 350 mL-solution of apple cider vinegar and water in equal parts.

The content of the traps, in both regions, was filtered, washed and subsequently preserved in 70% ethanol. Then they were labeled indicating date, crop and sampling site, and transferred to INTA facilities in Famaillá (Tucumán) and in Alto Valle (Río Negro), for observation by light microscopy and material classification.

The parasitoids obtained were individually conditioned in eppendorf tubes with 70% ethanol, and sent to La Plata Museum for taxonomic identification. Keys of Gibson et al. (1997) were used. Specimens were compared with the reference material deposited in the Entomology Division of the La Plata Museum, with the original description (Rondani, 1875) and with the diagnosis of the species (Sureshan, 2003). Photographs were taken using a Cannon Power Shot G9 camera, with a G7 adapter tube attached to a Zeiss Stemi DV4 40 x binocular loupe and a Leica DFC295 attached to a stereoscopic microscope Leica S8APO. Voucher copies were deposited in the collection of the Entomology Division of the La Plata Museum.

Pachycrepoideus vindemmiae (Fig. 1) diagnosis (Modified from Sureshan, 2003): Body length variable, 1.9-2.3 mm. General body color black with some metallic green to blue reflections. Head and mesosoma reticulate with very short and quite appressed thin and sparse pubescence, metasoma dorsally smooth. Head a little wider than high and wider than the greatest width of thorax. Antennae concolorous, inserted slightly below ventral end of eyes line, scape reaches the anterior border of median ocellus; scape, pedicel brown to light brown, first two ring-joints light brown to yellow, rest of funicular segments and clava brown, darker than scape and pedicel; third anellus as long as first and second anelli combined; rest of funicular segments longer than wide. Coxae

concolorous, legs variable, meso and metafemur brown, profemur a little darker, tibiae brown with apex light brown, tegulae brown; wings hyaline, forewing venation almost light brown, except marginal vein brown. Notauli very weak; propodeum with a weak median carinae and plicae.

Locality (province)	Geographic coordinates	Crop	Abundance (number of specimens)				Prevalence of <i>P. vindemmiae</i> (%)
			<i>D. suzukii</i> (♀ ♂)	Other Drosophilidae	Total Drosophilidae	<i>P. vindemmiae</i>	
La Reducción (Tucumán)	26° 56' 51" S 65° 20' 53" W	Blueberry	241	1138	1379	25	1.78
Orán (Tucumán)	27° 11' 37" S 65° 32' 43" W	Blueberry	175	671	846	2	0.24
El Molino (Tucumán)	27° 19' 44" S 65° 42' 07" W	Blueberry	131	315	446	14	3.04
Plottier (Neuquén)	38° 58' 00" S 68° 13' 59" W	Raspberry	638	413	1051	101	8.76
		Cherry	1401	248	1649	61	3.57
Allen (Río Negro)	38° 58' 40" S 67° 49' 40" W	Cherry	15	350	365	37	9.20
Gral. Roca (Río Negro)	39° 05' 00" S 67° 34' 30" W	Raspberry	37	1661	1698	18	1.05

Table I. Locations, crops, abundance of *Drosophila suzukii*, and abundance and prevalence of *Pachycrepoideus vindemmiae*, during the period December 2016-May 2017.

Biology

Pachycrepoideus vindemmiae is considered a polyphagous pupal parasitoid of a large number of dipterans, although it also behaves as a facultative hyperparasitoid.

Hosts

Anthomyiidae, Calliphoridae, Cecidomyiidae, Drosophilidae, Lonchaeidae, Muscidae, Phoridae, Piophilidae, Sarcophagidae, Sphaeroceridae, Stratiomyidae, Syrphidae and Tephritidae (Noyes, 2019).

Geographical distribution

Cosmopolitan species. In Argentina, it has been registered mainly in crops infested with tephritids (Ovruski, 1995; Purcell, 1998; Ovruski et al., 2000). The present work reports the first records of *P. vindemmiae* associated with *D. suzukii* in cherry and berry crops in Argentina. The records of Río Negro and Neuquén are the southernmost report of the species (Table I)

Pachycrepoideus vindemmiae is a generalist parasitoid of a wide range of cycloraphic dipterans and a facultative hyperparasitoid (Van Alphen & Thunissen, 1983; Wang & Messing, 2004). Tephritidae hosts include *Anastrepha fraterculus* Wiedemann, *A. ludens* Loew, *A. mombinpraeoptans* Sein, *A. obliqua* Macquart, *A. sororcula* Zucchi, *A. suspensa* Loew, *Ceratitis capitata* Wiedemann, *C. rosa* Karsch, *Dacus ciliatus* Loew, *D. cucurbitae* Colett, *D. dorsalis* Hendel, *D. oleae* Gmelin, *D. passiflorae* Frogg, *Myiopardalis pardalina* Bigot, *Rhagoletis cingulata* Loew, *R. fausta* Osten Sacken, *R. indifferens* Curran, and *Terellia fuscicornis* Loew (Noyes, 2019). Although *P. vindemmiae* is often raised in bulk for release against tephritid pests, it is likely that it prefers other hosts, such as drosophilids. Many of the oldest introductions against tephritid pests may have involved releases in areas where *P. vindemmiae* was already present but it was not detected because it did not attack the target pest. It was introduced in Hawaii to control *C. capitata* and in several locations in America to control *Anastrepha* spp. and *C. capitata* (Purcell, 1998; Ovruski et al., 2000).

In laboratory studies, *P. vindemmiae* easily parasitized another four parasitoids of tephritid flies commonly used in biological control programs (Wang & Messing, 2004). *Pachycrepoideus vindemmiae* showed no preference for non-parasitized tephritid hosts and their offspring were equally effective controlling *Drosophila melanogaster* Meigen and *C. capitata*. However, adult females showed a preference for the smaller host. Some authors warn against the use of *P. vindemmiae* in traditional biological control applications, due to its generalist tendencies and hyperparasitoid behavior, generating an expansion of the host range including non-target species (Wharton, 1989).

For the aforementioned, although these new records of the species allow knowing its distribution and adding one more potential resource to regulate SWD populations naturally, it is necessary to deepen studies on the tritrophic relationships in each evaluated region, in order to prevent possible imbalances that could lead to a decrease of *P. vindemmiae* efficacy as a potential SWD biocontroller in these agroecosystems.

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