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A newly detected exotic ambrosia beetle in Argentina: *Euwallacea interjectus* (Coleoptera: Curculionidae: Scolytinae)

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In Argentina, poplar (*Populus* spp.: Salicaceae) cultivation has been developing since the late 19th century, particularly in the lower Paraná Delta region. The poplar-forested area in the country was estimated to be around 14,500 has by 2012 (Signorelli & Gaute 2012). Poplars provide lumber for various uses and are important in the manufacture of reconstituted board products. Also, they represent the basis for the pulp and paper industry. Although some poplar species may have fairly extensive geographic ranges, they are restricted in occurrence and often exhibit their best development in riparian areas. Following a significant drought event in Dec 2008, some scolytine species (Coleoptera: Curculionidae: Scolytinae) have appeared regularly in poplar forests in the region (Landi et al. 2017). Today, the rate of introduction and establishment of non-native insect species is increasing worldwide as a result of human activity, mainly due to the transport of goods in international trade (Brockerhoff et al. 2006). In the presence of suitable conditions, the establishment of exotic species may be favored, and may become invasive by attacking living trees (Liebhold et al. 1995).

Among the ambrosia-feeding scolytines, the Xyleborini includes approximately 1,200 species in 30 genera that are distributed in forests throughout the world (Cognato et al. 2011). Besides their xylomycectophagy, the Xyleborini are biologically defined by extreme inbreeding and the presence of a haplodiploid genetic system wherein haploid males are flightless and often dwarfed (Hulcr et al. 2007).

Euwallacea (Hopkins) (Coleoptera: Curculionidae) is a large, cosmopolitan genus that contains over 50 species, mostly native from Asia (Wood & Bright 1992). Only a few species are introduced in the Americas (e.g., *Euwallacea fornicatus* Eichhoff, *Euwallacea validus* Eichhoff, and *Euwallacea interjectus* Blandford) (Rabaglia et al. 2006; Cognato et al. 2015), none of which is reported to occur presently in Argentina.

The purpose of this article is to communicate the detection of the Asian ambrosia beetle *E. interjectus* in Argentina and South America for the first time. Both traditional morphology-based taxonomy and molecular diagnosis through DNA barcoding were combined in order to identify the species.

In Argentina, *E. interjectus* was first found in Apr 2009 from a 10-yr-old poplar plantation (34.139332°S, 58.795332°W). Healthy trees of *Populus deltoides* W. Bartram cv 'Deltoides' (Salicaceae) attracted

attention due to the presence of sap exuding from beetle entrance holes. They were cut into 30 cm logs and taken to the laboratory, where they were maintained in order to capture the adult beetles as they emerged; the beetles were preserved in 96% ethanol. Afterwards, the logs were chopped, immature stages were collected, and wood damage (i.e., galleries, wood stained by symbiotic fungi) was confirmed. Later, between Nov 2012 and Feb 2013, 12-unit multifunnel traps were located in poplar plantations as part of a surveillance program. Traps were baited with ethanol lures (low rate of release), and wet collection cups filled with propylene glycol (150 mL) were used as the killing and preserving agent. Consequently, a second collection was achieved. In both cases, adults were deposited at the Instituto Nacional de Tecnología Agropecuaria (Instituto de Recursos Biológicos, Buenos Aires, Argentina), and samples were selected for both morphological and molecular study.

Taxonomic identification was based on structural characters observed under stereomicroscope, and photographs of diagnostic characters were taken by means of scanning electron microscopy. As Cognato et al. (2015) pointed out, *E. interjectus* shares very similar morphological traits with *E. validus*. In general, these species can be distinguished by the shape of the declivity, the punctures, and placement of the tubercles on the second declivital interstriae. In *E. interjectus*, the declivity is gradually sloped from base to apex, punctures of the declivital striae are shallow, giving the declivity a smooth appearance, and tubercles on the second declivital interstriae extend from the base to the apex (Fig. 1).

Sequences of cytochrome oxidase subunit I (COI) were acquired from 2 of the specimens sampled from poplar plantations in Argentina in order to verify the species identification with molecular evidence. The primers used allowed us to amplify and sequence almost an entire COI fragment that includes both the 5' region, the standardized DNA "barcode," as well as the 3' region of COI, which also is widely used for species identification purposes. The protocols and conditions for DNA isolation, PCR amplification, and sequencing of the COI gene were conducted as in Pereyra et al. (2016). Sequences then were checked and compared with others available in GenBank through the Basic Local Alignment Search Tool

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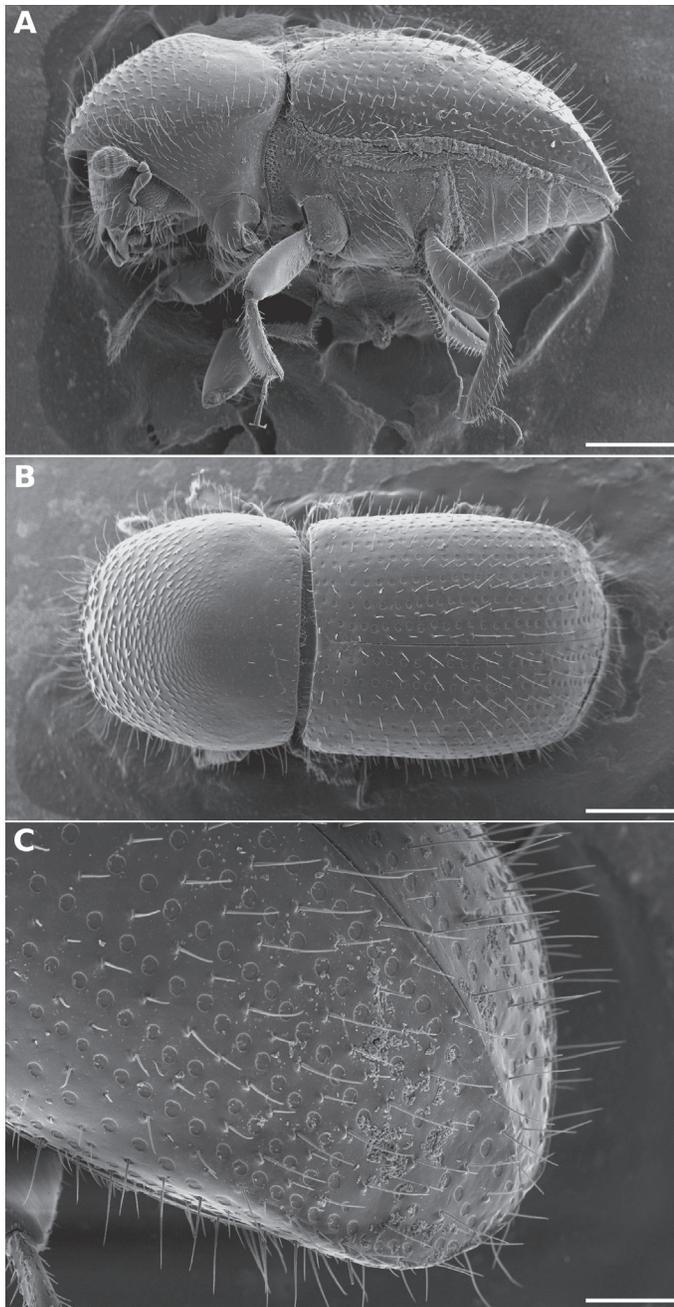


Fig. 1. *Euwallacea interjectus* female. (A) Lateral view, (B) Dorsal view, (C) Elytral declivity gradually sloped to the apex, showing tubercles on the second declivital interstriae. Scale bars: (A, B) 500 μ m, (C) 200 μ m.

(BLAST). BLAST searches gave significant alignments with *E. interjectus*, with 100% - 99% identity values with specimens from Japan and USA sequenced in previous studies (e.g., Cognato et al. 2015). Voucher specimens are deposited in the Entomology collection of the Museo de Ciencias Naturales de La Plata (La Plata, Buenos Aires, Argentina). The new sequences of *E. interjectus* from Argentina are deposited in GenBank under accession numbers MH105073 and MH105074.

The occurrence of genus *Euwallacea* in South America has been detected at least once during the current decade. In 2011, SAG (Servicio Agrícola Ganadero de Chile) intercepted *E. validus* in a ship with containers filled with stamped wooden packing coming from the US. The

presence of *E. interjectus* in poplar cultivations of Argentina represents a new find and is cause for concern due to the potential of this species to become invasive and spread rapidly (Cognato et al. 2015). The geographical expansion of the species is probably in progress, favored by both the shipment of timber products globally and the characteristics of the mating system of many Scolytinae (Kirkendall & Faccoli 2010; Rassati et al. 2018). Potential risks associated with this species include not only direct wood damage but also the spread of different species of fungi (O'Donnell et al. 2015). *Euwallacea interjectus* is among the relatively few species that attack living trees. Although it has been detected only affecting stressed plants so far, under suitable conditions it may attack healthy trees (Knížek & Beaver 2004). Consequently, *E. interjectus* could become economically relevant if it damages healthy stands of poplar monocultures in the Paraná Delta region. The genus *Euwallacea* has been reported to attack a significant number of hosts even in riparian areas (Boland 2016). Previously, *E. interjectus* had not been observed affecting poplar trees outside its natural range of distribution (Wood & Bright 1992). This article constitutes the first report of the species not only in South America and Argentina, but also affecting cultivated poplars. *Euwallacea interjectus* is now abundant in the Delta of Paraná River region, and several incidences of mass attack on live water-stressed poplars have been observed. Thus, there is an urgent need for study and monitoring of this exotic species as it may become a serious problem.

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Summary

An exotic species of ambrosia beetle (Coleoptera: Curculionidae: Scolytinae) of Asian origin has been detected recently in Argentina. *Euwallacea interjectus* (Blandford) has been collected infesting cultivated poplars in the Paraná Delta region. Identification was based on morphology and on DNA sequences of the COI gene. This finding adds to the increasing number of recent introductions of exotic ambrosia beetles into South America.

Key Words: Scolytinae; Xyleborini; DNA; new record; geographical distribution; poplars

Sumario

Recientemente se detectó por primera vez en Argentina una especie exótica de escarabajo de Ambrosía (Coleoptera: Curculionidae: Scolytinae) de origen asiático. *Euwallacea interjectus* (Blandford) fue colectada afectando cultivos de álamo en la región del Delta del Río Paraná. Su identificación se basó en la morfología y en las secuencias de ADN del gen COI. Este hallazgo se suma al número cada vez mayor de introducciones recientes de escarabajos de Ambrosía exóticos en Sudamérica.

Palabras Clave: Scolytinae; Xyleborini; ADN; nuevo registro; distribución geográfica; álamos

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