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First record of the eucalypt gall-wasp *Leptocybe invasa* (Hymenoptera: Eulophidae) from Uruguay

Primer reporte de la avispa agalladora del eucalipto *Leptocybe invasa* (Hymenoptera: Eulophidae) para Uruguay

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SUMMARY

Leptocybe invasa is a recently introduced forest pest, responsible for significant economic loss in *Eucalyptus* spp. plantations worldwide. The objective of this work is to report its presence in Uruguay, providing data of its distribution and biology. A survey for galls was conducted from 2013 to 2015. The presence of galls was confirmed in 15 out of 35 surveyed points and four out of 157 traps. *Eucalyptus benthamii* was recorded as a new host. Three periods of emergence were observed. Life cycle was completed only in *E. tereticornis*, *E. camaldulensis* and *E. grandis*.

Key words: gall-insects, *Eucalyptus*, forestry pests.

RESUMEN

Leptocybe invasa es una plaga forestal de reciente introducción, responsable de importantes pérdidas económicas en plantaciones de *Eucalyptus* spp. a nivel mundial. El objetivo de este trabajo fue reportar su presencia para Uruguay, aportar datos de distribución y biología. Se observó la presencia de agallas de 2013 a 2015, confirmada en 15 de 35 puntos muestreados y en cuatro de 157 trampas. *Eucalyptus benthamii* se registra como nuevo hospedero. Fueron observados tres períodos de emergencia de adultos. El ciclo de vida fue completado solamente en *E. tereticornis*, *E. camaldulensis* y *E. grandis*.

Palabras clave: insectos agallícolas, *Eucalyptus*, plagas forestales.

INTRODUCTION

Commercial forestry with *Eucalyptus* species reaches 80 % of the forested area in Uruguay, leading, in 2013, to more than 590 million dollars in export goods (DIEA 2014). *Eucalyptus globulus* Labill and *E. grandis* W. Hill ex Maiden are the most planted species, while red gum trees (*E. tereticornis* Sm. and *E. camaldulensis* Dehnh.) have been extensively used as shade and shelter forest in cattle breeding. Paine *et al.* (2011) suggest a correlation between the increase of global area planted with *Eucalyptus* spp. and the arrival of new insects and diseases. This phenomenon has also been observed in Uruguay, particularly during the last decade (Martínez 2010).

The last wave of world invasive forest pests has been characterized by small, cryptic insects, notably gall forming species (Paine *et al.* 2011). The *Eucalyptus* gall-wasps *Ophelimus maskelli* Ashmead 1900, *Selitrichodes globulus* La Salle and Gates 2009 and *Leptocybe invasa* Fischer and La Salle 2004 are well known because of their economic importance, though the latter is particularly relevant due to the aggressiveness of the attack and its rapid dispersion (Paine *et al.* 2011).

The Australian gall wasp *L. invasa* was first recorded from South America in 2008; in Brazil in 2008, Argentina in 2010 and Chile in 2014 (Wylie and Speight 2012, SAG 2014). This insect has become a relevant pest for eucalypt forestry worldwide. Life cycle occurs almost entirely inside the gall. In temperate regions, the developmental cycle lasts 130 days on average leading to the occurrence of 2-3 generations annually (Mendel *et al.* 2004, Aquino *et al.* 2011), reaching to 5-6 generations in tropical areas (Zhu *et al.* 2012). Thelytokous parthenogenesis is the most common reproductive mechanism in *L. invasa* (Mendel *et al*. 2004). The objective of this work is to report the presence of *L. invasa* in Uruguay and the first mention of *Eucalyptus benthamii* as its host.

METHODS

Occurrence of galls on eucalypt was recorded for the first time in April 2011 in Montevideo during an expert visit (34°50'10.96" S; 56°13'11.77" W). From August 2013 to August 2015 we conducted a survey of galls on eucalypt trees in plantations and shade and shelter forests. Thirty-five sites were selected following two criteria: (1) presence of *Eucalyptus* species susceptible to *L. invasa* and (2) proximity to wood transport routes. All sites were inspected monthly to observe gall development. Adult wasps for identification were obtained from two sites located on *E. tereticornis* x *E. camaldulensis* hybrids and *E. grandis* at INIA Experimental Station (Tacuarembó). Additional points of occurrence were obtained from the review of 157 yellow sticky traps belonging to the National Monitoring System (Martínez *et al.* 2014).

The observation of specimens was made under a stereoscopic microscope Zeiss Stemi DV4 and by Scanning Electron Microscope (SEM) JEOL 5900 Low Vacuum. We followed Mendel *et al.* (2004) to determine the adult wasps and to describe gall morphology and we compared the individuals against identified material kept in the Forest Entomological Collection of INIA in Tacuarembó, Uruguay.

RESULTS

Gall and adult sampling. Gall occurrence was confirmed on 15 out of 35 surveyed points (figure 1). Emergency was confirmed on *E. tereticornis* x *E. camaldulensis* and *E. grandis* in three periods: August-September, mid-December and March-April. Only four out of 157 sticky traps checked contained adult wasp of *L. invasa* within the same three periods observed in the field survey (figure 1).

Material examined. Uruguay, Tacuarembó: INIA Experimental Station (31°44'23.84" S; 55°58'44.74" W) *ex Eucalyptus tereticornis* Sm. (three female adults).

Leptocybe invasa is a small wasp (1.1-1.4 mm) brownish in color with blue green metallic shine on the thorax (figure 2A). Specimens were ascribed to *L. invasa* based on the antennal morphology, thoracic morphology and wing venation (figure 2B-D). Detailed diagnosis of adult female was provided by Mendel *et al.* (2004). Scape longer than the *pedicellum* with broadened middle section. *Flagellum* with six segments separated by four *annelli*, the basal segments constitute the *funiculum* and the three apical segments form the antennal mass (figure 2B). Short prothorax; well-developed mesothorax; *scutellum* divided into three

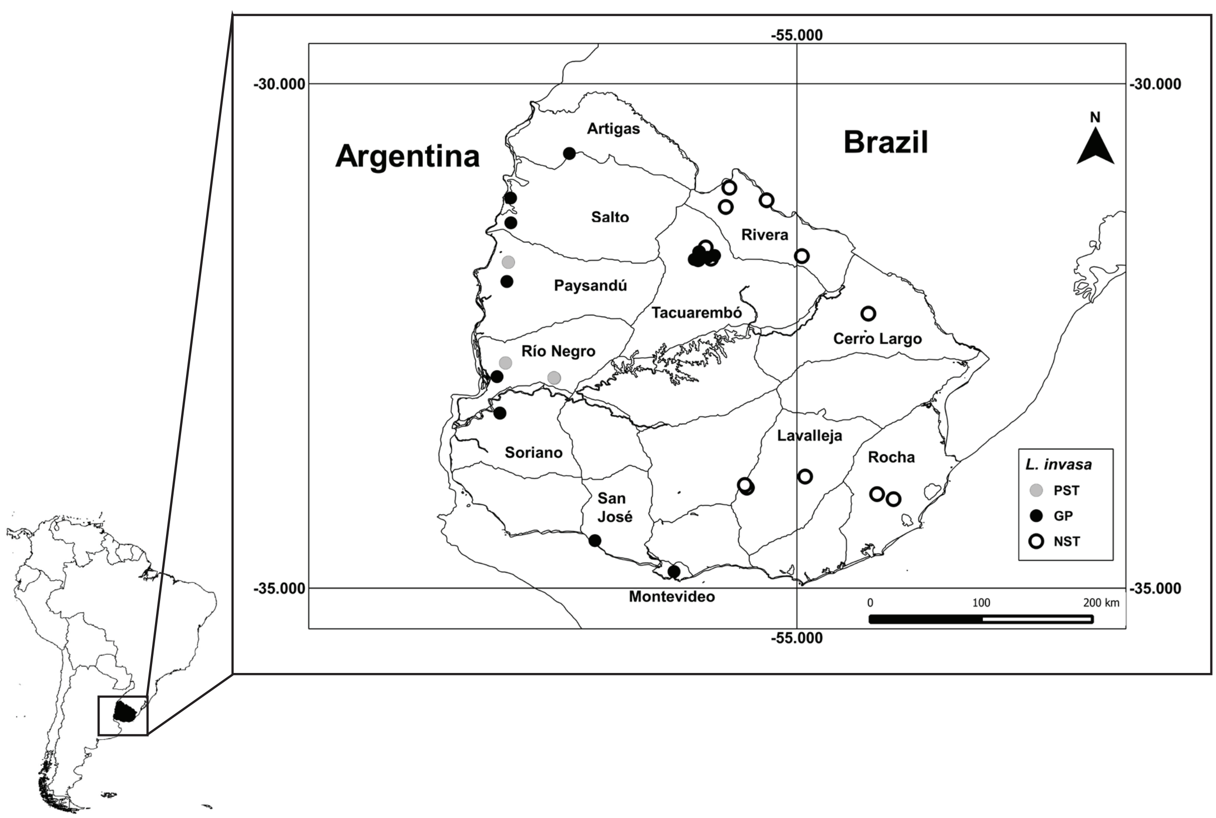

Figure 1. Occurrence of *L. invasa* in Uruguay. On yellow sticky cards: grey dots = presence; white dots = absence; black dots = presence of galls on survey points.

Presencia de *L. invasa* en Uruguay. En trampas amarillas: puntos grises = presencia; puntos blancos = ausencia; puntos negros = presencia de agallas en puntos inspeccionados.

separated zones by sublateral lines (figure 2C). Hyaline wings completely covered with setae, slight wing venation (figure 2D). Post marginal vein is shorter than stigmal vein. Submarginal vein presents 2-5 setae, visible on dorsal view. Identified material was deposited in the Forestry Entomological Collection of INIA in Tacuarembó, Uruguay.

Galls of *L. invasa* are always located in the central rib or in leaf petioles (figure 3A). Early attack is observed in the form of rectilinear wounds on the epidermis, which are inflicted by females during oviposition (figure 3B). Induced galls can cause deformation on leaves and apical shoots (figure 3C). Severe attacks can produce tissue necrosis and defoliation (figure 3D).

Table 1 reviews eucalypt species reported as susceptible to *L. invasa*. We observed for the first time the occurrence of galls on *E. benthamii*. Additionally, we observed

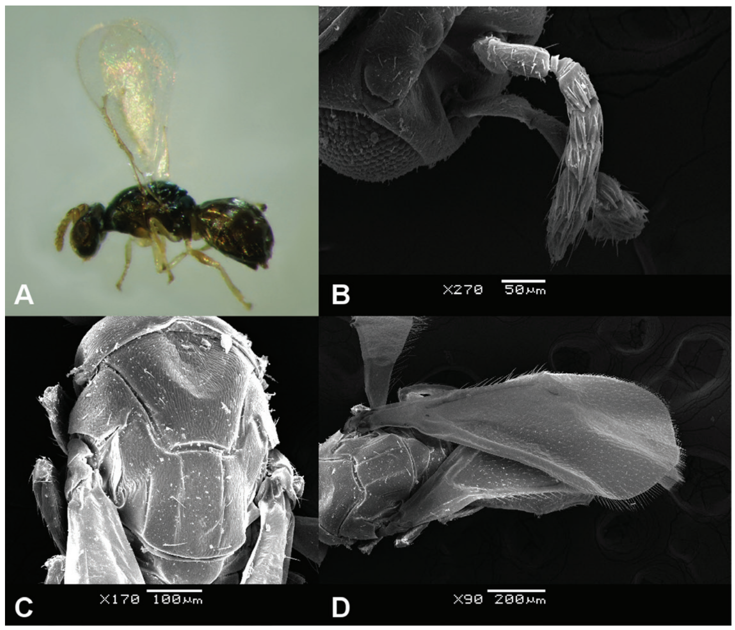

Figure 2. Adult female of *Leptocybe invasa*: A) Lateral view, body length 1.2 mm. B) Antenna. C) Thorax. D) Wings.

Hembra adulta de *L. invasa*: A) Vista lateral, longitud del cuerpo1,2 mm. B) Antena. C) Tórax. D) Alas.

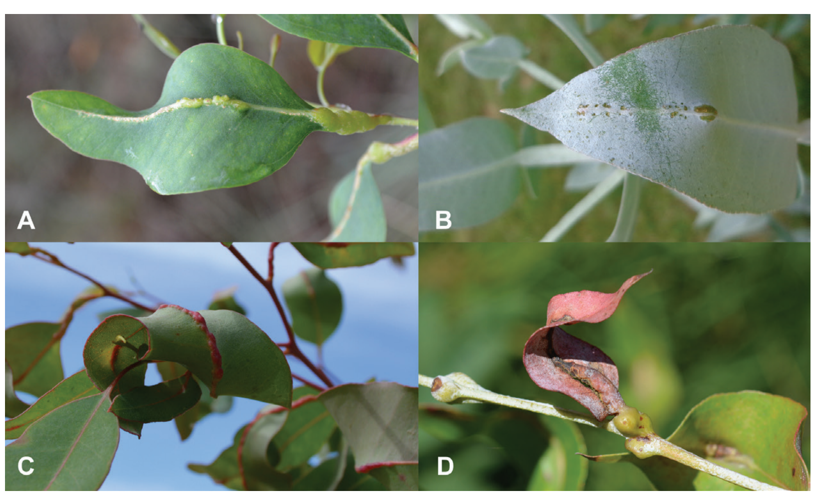

Figure 3. A) Shape and typical position of galls of *Leptocybe invasa* in *Eucalyptus tereticornis* x *E. camaldulensis*. B) Oviposition marks in *E. globulus*. C) Anomalous development on young plant in *E. grandis*. D) Necrosis and death of leaves and stems as a consequence of the attack by *L. invasa* in *E. globulus*.

A) Forma y posición típica de agallas de *L. invasa* en *E. tereticornis* x *E. camaldulensis*. B) Marcas de oviposición en *E. globulus*. C) Desarrollo anómalo en plantas jóvenes de *E. grandis.* D) Necrosis y muerte de hojas y ramas a consecuencia del ataque de *L. invasa* en *E. globulus.*

Table 1. Reported hosts for *Leptocybe invasa*. N.S.: Not specified.

Hospederos reportados para *Leptocybe invasa*. N.S.: No especificado.

Species	Countries/Sources
Subgenus: *Idiogenes*	
E. cloeziana F. Muell	N.S. (Nadel and Slippers 2011)
Subgenus: *Monocalyptus*	
E. pilularis Sm.	Vietnam (Quang Thu *et al*. 2009), N.S. (Nadel and Slippers 2011)
Subgenus: *Nothocalyptus*	
E. microcorys Muell.	Vietnam (Quang Thu *et al.* 2009), N.S. (Nadel and Slippers 2011), China (Zhu *et al.* 2012)
Subgenus: Symphyomyrtus	
Eucalyptus alba Reinw. *ex* Blume	N.S (Nadel and Slippers 2011)
E. botryoides Sm.	Israel (Mendel *et al*. 2004), N.S. (Nadel and Slippers 2011)
E. bridgesiana Baker	Israel (Mendel *et al*. 2004), N.S. (Nadel and Slippers 2011), N.S. (FAO 2012)
E. camaldulensis Dehnh.	Israel (Mendel *et al*. 2004), Uganda (Nyeko *et al*. 2010), Vietnam (Quang Thu *et al*. 2009), Argentina (Aquino *et al*. 2011, Nadel and Slippers 2011), N.S (FAO 2012), China (Zhu *et al.* 2012, Luo *et al*. 2014), Brazil (Cegatta and Villegas 2013), Mozambique (Chirinzane *et al*. 2014) Chile (SAG 2014)
E. cinerea F. Muell. ex Benth.	N.S (FAO 2012)
E cladocalyx Muell.	N.S (Nadel and Slippers 2011)
E. coolabah Blakely *et* Jacobs	Vietnam (Quang Thu *et al*. 2009)
E. dunnii Maiden	Kenya (Nyeko *et al*. 2010), N.S. (Nadel and Slippers 2011), N.S. (FAO 2012), China (Zhu *et al*. 2012)
E. excerta Muell.	China (Zhu *et al*. 2012)
E. globulus Labill.	Israel (Mendel *et al*. 2004), Uganda (Nyeko *et al*. 2009), N.S (Nadel and Slippers 2011), N.S. (FAO 2012) Chile (SAG 2014)
E. gomphocephala D.C.	N.S. (Nadel and Slippers(2011)
E. grandis Hill *ex* Maiden	Israel (Mendel *et al*. 2004), Vietnam (Quang Thu *et al*. 2009), Kenya and Uganda (Nyeko *et al*. 2010), N.S. (Nadel and Slippers 2011), N.S (FAO 2012)
E. gunnii Hook. F.	Israel (Mendel *et al*. 2004)
E. leucoxylon Muell.	N.S. (Nadel and Slippers 2011)
E. maidenii Muell.	Uganda (Nyeko *et al*. 2009), Vietnam (Quang Thu *et al*. 2009), N.S. (Nadel and Slippers 2011), N.S. (FAO 2012)
E. moluccana Roxb.	Vietnam (Quang Thu *et al*. 2009)
E. nicholii Maiden *et* Blakely	N.S. (FAO 2012)
E. nitens Deane *et* Maiden	N.S. (Nadel and Slippers 2011)
E. pellita Muell.	Vietnam (Quang Thu *et al*. 2009), N.S. (Nadel and Slippers 2011)
E. propinqua Deane *et* Maiden	China (Zhu *et al*. 2012)
E. pulverulenta Sims	N.S. (FAO 2012)
E. robusta Sm.	Israel (Mendel *et al*. 2004), Uganda (Nyeko *et al*. 2010), Vietnam (Quang Thu *et al*. 2009), N.S. (Nadel and Slippers 2011), N.S. (FAO 2012), China (Zhu *et al*. 2012)
E. rudis Sm.	N.S. (FAO 2012)
E. saligna Sm.	Israel (Mendel *et al*. 2004),Vietnam (Quang Thu *et al*. 2009), Kenya and Uganda (Nyeko *et al*. 2010), N.S. (Nadel and Slippers 2011), China (Zhu *et al*. 2012), Mozambique (Chirinzane *et al*. 2014)
E. sideroxylon Cunn.	N.S (Nadel and Slippers (2011))
E. smithii Baker	Vietnam (Quang Thu *et al*. (2009)), N.S (Nadel and Slippers (2011))
E. tereticornis Sm.	Israel (Mendel *et al*. (2004)), Vietnam (Quang Thu *et al*. (2009)), Kenya (Nyeko *et al*. (2010)), N.S (Nadel and Slippers (2011)), China (Zhu *et al*. (2012), Luo *et al*. (2014))
E. urophylla Blake	Vietnam (Quang Thu *et al*. (2009)), Kenya (Nyeko *et al*. (2010)), N.S (Nadel and Slippers (2011)), N.S (FAO (2012)), China (Zhu *et al*. (2012))
E. viminalis Labill.	Israel (Mendel *et al*. (2004)), N.S (Nadel and Slippers (2011))

galls on red gums (*E. tereticornis*, *E. camaldulensis* and hybrids of these two species), *E. dunnii*, *E. grandis* and *E. globulus*. We only observed emergency holes on red gums and *E. grandis* in different growing stages in the field, we did not inspect greenhouses or nurseries.

DISCUSSION

We observed galls on six species already reported as susceptible, with the addition of *E. benthamii*. Emergence period observed in our study suggests that the duration of the life cycle in Uruguay could be on average 120 days, similar to other temperate regions (Aquino *et al*. 2011). The presence of emergency holes solely on *E. grandis* and red gums suggests that not all species are equally suitable to complete the cycle. However, from the point of view of forestry, gall formation *per se* may hamper the development of the tree stands implying additional costs for tree production.

We assume that *L. invasa* colonized Uruguay from Argentina. Data suggest that, as first reports were closer to the Uruguayan border (provinces of Buenos Aires and Entre Rios) than to Brazil (Bahia and São Paulo States (Costa *et al*. 2008), and no reports have been made yet by foresters from the East region of Uruguay.

We collected additional individuals from a network of yellow sticky traps already established for monitoring other forestry pests. Although sticky traps have been used in other countries for collecting *L. invasa* (Kumari *et al*. 2010, Zhu *et al*. 2012), identification of the captured micro-hymenopteran insects has proven a difficult task due to the conservation status of the specimens.

The report of *L. invasa* in the country poses a threat to Uruguayan commercial forestry, hence research should focus on the development of management strategies for this pest. Chemical control has been tested in Brazil and India (Kumari *et al*. 2010, Cegatta and Villegas 2013), although more than 80 % of the commercial stands are certified under ISO or FSC standards in Uruguay, turning this option unlikely, or at least restricted to nurseries. Alternatively, plant breeding seems to be a promising long term control tool for this pest (Kulkarni 2010, Dittrich-Schröder *et al*. 2012, Zhu *et al*. 2012). Since we observed variation in susceptibility to *L.invasa* among *Eucalyptus* species and hybrids, future studies should focus on screening resistance to attack from *L. invasa* in national genetic stock. Biological control programs are currently being implemented in Israel (Kim *et al*. 2008), South Africa (Kelly *et al*. 2012, Dittrich-Schröder *et al*. 2014), Chile (SAG 2014) and Brazil (de Sá *et al*. 2015). National studies may focus on the identification of the most reliable biocontrol candidates for our ecological context.

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