



# Susceptibility of eucalyptus species and hybrids to the gall wasp *Leptocybe invasa* (Hymenoptera: Eulophidae) in northern Misiones, Argentina

Edgar R. Eskiviski<sup>1</sup>, Maria E. Schapovaloff<sup>1</sup>, Delia M. Dummel<sup>1</sup>, Margarita M. Fernandez<sup>2</sup>, and Fernando L. Aguirre<sup>3</sup>

<sup>1</sup>INTA-EEA Montecarlo, Av. El Libertador 2472, CP 3384, Montecarlo, Misiones, Argentina. <sup>2</sup>FCF-UNaM, Bertoni 124, CP 3380, Eldorado, Misiones, Argentina. <sup>3</sup>ARAUCO, CP 3374, Puerto Libertad, Misiones, Argentina.

## Abstract

**Aim of study:** To analyze the susceptibility of *Eucalyptus* and hybrids species to *Leptocybe invasa* through field assays.

**Area of study:** The north of the Argentine province of Misiones (Colonia Delicia).

**Material and methods:** A total of 11 *Eucalyptus* species and 2 hybrids were surveyed for damage and severity of *L. invasa* infestation. Six evaluations were made during an annual period.

**Main results:** The susceptibility ranking to *L. invasa* from highest to lowest was *E. tereticornis* > *E. propinqua* > *E. dunnii* > *E. camaldulensis* > *E. grandis* > *E. major* > *E. longistrata* > *E. grandis* × *E. camaldulensis*. However, *E. moluccana*, *E. urophylla* × *E. grandis* and *E. urophylla* were tolerant to *L. invasa*.

**Research highlights:** This study suggests that in Misiones, *E. tereticornis* is the most sensible eucalyptus species to gall wasp attack, whereas other species and hybrids presented low damage levels or tolerance to *L. invasa* region.

**Additional key words:** forest plantation; exotic pest; damage infestation.

**Abbreviations used:** ADI (average damage index); DI (damage index).

**Authors' contribution:** Conceived and designed the experiments, and wrote the paper: ERE and MES. All authors performed the experiments, read and approved the final manuscript.

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**Correspondence** should be addressed to Edgar Eskiviski: [eskiviski.edgar@inta.gob.ar](mailto:eskiviski.edgar@inta.gob.ar)

## Introduction

*Eucalyptus* is the most widely planted exotic species in the tropics, and play important roles in reforestation and production of timber, pulp, potential bioenergy feedstock, and other forest products (Wyle & Floyd, 1998; Rejmánek & Richardson, 2011). In Argentina, *Eucalyptus* spp. plantations occupy about 261,000 ha, mainly in the northeast region (MAGyP, 2015).

The eucalyptus gall wasp, *Leptocybe invasa* Fisher & La Salle (Hymenoptera: Eulophidae), native from Australia, is a worldwide pest in *Eucalyptus* plantations. *L. invasa* has expanded to more than 29 countries in Asia, Europe, Africa and the Americas (Mendel *et al.*, 2004; Zheng *et al.*, 2014). This insect was first reported in Argentina in 2009 (Aquino *et al.*, 2011). The female *L. invasa* lays her eggs in plant tissues causing

the formation of galls on the leaf midribs and petioles and on the stem of new shoots, eventually leading to leaf-curling and premature aging of the leaves. Eggs overloading might cause death of young shoots, while severe attacks lead to leaf fall, stunted growth and may seriously weaken the tree (Mendel *et al.*, 2004). Gall development occurs as response to a physiological disorder caused by a specific relationship between plant and insect. It is highly dependent on specific small changes in morphological and phenological characteristics of the plant, which determines the degree of susceptibility and coevolution between plant and insect (Stone & Schönrogge, 2003; Zheng *et al.*, 2014).

*Eucalyptus* species show different susceptibility to *L. invasa* attacks (Mendel *et al.*, 2004) with *Eucalyptus grandis* W. Hill, *Eucalyptus camaldulensis* Dehnh and *Eucalyptus tereticornis* Smith being the most susceptible

ones (Thu *et al.*, 2009). However, the susceptibility can be strongly influenced by local environmental factors, and therefore further susceptibility tests are needed in locations where eucalyptus trees are cultivated. The objective of the present study was to evaluate the susceptibility of different *Eucalyptus* species and hybrids to *L. invasa* planted in the field in Argentina.

## Material and methods

This study was conducted from March to November 2014 in Colonia Delicia, Misiones, Argentina (26°10'16" S, 54°33'15"W). The climate is subtropical without dry seasons. *Eucalyptus* seedlings, less than one year old, were planted in a field experiment without irrigation. *L. invasa* damages were evaluated in a 1.35 ha field plot, with a spacing between plants of 3.0 × 2.5 m (1,333 plants/ha). The evaluated species were *E. camaldulensis*, *E. grandis*, *E. major*, *E. propinqua*, *E. tereticornis*, *E. dunnii*, *E. moluccana*, *E. longirostrata*, *E. urophylla* and the hybrids *E. grandis* × *E. camaldulensis* and *E. urophylla* × *E. grandis* (Table 1), planted in October 2013.

The experiment was designed as a complete randomized block, with 4 blocks and 5 replications. In each block, 11 experimental plots of each species with 5 plants were randomly established, and all trees in these plots were surveyed for damage and severity of *L. invasa* infestation. Six evaluations were made during 2014 between March and November, evaluating the damage index (DI) and average damage index (ADI). DI scored the proportion of leaves and twigs damaged per tree, as follows: 0, no damage; 1 < 25%; 2, 25-50%; 3, 51-75%; 4 >75% (Thu *et al.*, 2009). DI was calculated as the percentage of trees affected. ADI was calculated according to:

$$ADI = \sum ni \cdot vi / Ni$$

where  $ni$  is the number of trees infected at  $DI=i$ ,  $vi$  the damage index at level  $i$ , and  $Ni$  is the number of trees assessed per species. Based on maximum ADI values, a damage severity level was defined (Thu *et al.*, 2009): nil (ADI = 0), low (ADI: 0-1.0), medium (ADI: 1.1-2.0), severe (ADI: 2.1-3.0) and very severe damage (ADI: 3.1-4.0).

Statistical analysis was performed using analysis of variance (ANOVA) after checking that normality and homoscedasticity were fulfilled, and means were compared with the Tukey test ( $p < 0.05$ ), using InfoStat statistical software V 2016 (Di Rienzo *et al.*, 2016).

## Results and discussion

Damage incidence by *L. invasa* significantly differed among the tree species ( $df=18$ ,  $F=28.42$ ,  $p < 0.0001$ ). The highest damage incidence was observed on *E. tereticornis* and the lowest in *E. urophylla*, *E. urophylla* × *E. grandis* and *E. moluccana*. *E. grandis* × *E. camaldulensis*, *E. longirostrata*, *E. major*, *E. grandis*, *E. camaldulensis*, *E. dunnii* and *E. propinqua* showed medium damage incidence (Table 2).

Based on ADI, in the six evaluations between March and November 2014, the resistance of eucalyptus species differed widely. *E. urophylla* and the hybrid *E. urophylla* × *E. grandis* seemed tolerant to *L. invasa* and no galls were produced. Six species (*E. moluccana*, *E. longirostrata*, *E. major*, *E. grandis*, *E. camaldulensis*, *E. dunnii*) and the hybrid *E. grandis* × *E. camaldulensis* showed low damage severity. Medium damage was observed in one species, *E. propinqua* and severe damage occurred in *E. tereticornis* (Table 2).

**Table 1.** Provenances of *Eucalyptus* species selected for the field trial.

Seedlot	Section	Series	Species	Collection location
ECA90	Exsertaria	Rostratae	<i>E. camaldulensis</i>	Kenia
EDU64	Maidenaria	Globulares	<i>E. dunnii</i>	Buenos Aires, Argentina
EGC91	Transversaria	Salignae	<i>E. grandis</i>	São Paulo, Brazil
EGC91			<i>E. grandis</i> × <i>E. camaldulensis</i>	São Paulo, Brazil
EUG92			<i>E. grandis</i> × <i>E. urophylla</i>	São Paulo, Brazil
ELO119	Transversaria	Punctatae	<i>E. longirostrata</i>	South Africa
EMJ110	Transversaria	Punctatae	<i>E. major</i>	Queensland, Australian
EMO33	Adnataria	Moluccanae	<i>E. moluccana</i>	Queensland, Australia
EPR103	Transversaria	Punctatae	<i>E. propinqua</i>	Unumgar, Australia
ETE63	Exsertaria	Tereticornes	<i>E. tereticornis</i>	Buenos Aires, Argentina
EUR94	Transversaria	Resiniferae	<i>E. urophylla</i>	São Paulo, Brazil

**Table 2.** Damage incidence (DI) and average damage index (ADI) of *Leptocybe invasa* infestation on *Eucalyptus* species and hybrids in the field.

<i>Eucalyptus</i>	DI (%)	ADI						Damage severity
		March	April	May	July	August	November	
<i>E. urophylla</i>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	nil
<i>E. urophylla</i> × <i>E. grandis</i>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	nil
<i>E. moluccana</i>	2 <sup>a</sup>	0 <sup>a</sup>	0.20 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	low
<i>E. grandis</i> × <i>E. camaldulensis</i>	11 <sup>ab</sup>	0 <sup>a</sup>	0.25 <sup>a</sup>	0.40 <sup>ab</sup>	0.25 <sup>ab</sup>	0.05 <sup>a</sup>	0.15 <sup>a</sup>	low
<i>E. longistrata</i>	26 <sup>bc</sup>	0.10 <sup>a</sup>	0.60 <sup>a</sup>	0.50 <sup>ab</sup>	0.55 <sup>ab</sup>	0.30 <sup>a</sup>	0.30 <sup>a</sup>	low
<i>E. major</i>	29 <sup>c</sup>	0.15 <sup>a</sup>	0.75 <sup>a</sup>	0.85 <sup>ab</sup>	0.75 <sup>ab</sup>	0.35 <sup>a</sup>	0.75 <sup>ab</sup>	low
<i>E. grandis</i>	29 <sup>c</sup>	0 <sup>a</sup>	0.40 <sup>a</sup>	0.70 <sup>ab</sup>	0.95 <sup>ab</sup>	0.45 <sup>a</sup>	0.35 <sup>ab</sup>	low
<i>E. camaldulensis</i>	34 <sup>c</sup>	0.45 <sup>a</sup>	0.65 <sup>a</sup>	0.70 <sup>ab</sup>	0.80 <sup>ab</sup>	0.60 <sup>a</sup>	0.60 <sup>ab</sup>	low
<i>E. dunnii</i>	38 <sup>c</sup>	0.45 <sup>a</sup>	0.75 <sup>a</sup>	0.90 <sup>ab</sup>	0.95 <sup>ab</sup>	0.30 <sup>a</sup>	0.65 <sup>ab</sup>	low
<i>E. propinqua</i>	41 <sup>c</sup>	0.11 <sup>a</sup>	0.83 <sup>a</sup>	1.33 <sup>b</sup>	1.39 <sup>bc</sup>	0.78 <sup>a</sup>	1.33 <sup>b</sup>	medium
<i>E. tereticornis</i>	72 <sup>d</sup>	1.55 <sup>b</sup>	2.60 <sup>b</sup>	2.65 <sup>c</sup>	2.30 <sup>c</sup>	1.80 <sup>b</sup>	2.30 <sup>c</sup>	severe

Mean values followed by the same letter within a column are not significantly different according to Tukey test ( $p < 0.05$ ).

Differences in susceptibility to the gall-forming insects of eucalyptus tested may indicate that genetic factors are involved in both, attractiveness to *L. invasa* for oviposition and suitability for *L. invasa* larval development. Similarly, synchronization of gall-forming species with host plant phenology is behind many cases of susceptibility as consequence of coevolution between eucalyptus plants and *L. invasa*. These factors open the chance of coping with this pest through the development of resistant and productive genetic stock, better than through chemical means (Dungey *et al.*, 2000; Guerreiro *et al.*, 2015). In this context, the order of susceptibility to *L. invasa* in Misiones was *E. tereticornis* > *E. propinqua* > *E. dunnii* > *E. camaldulensis* > *E. grandis* > *E. major* > *E. longistrata* > *E. grandis* × *E. camaldulensis*. Other studies have also showed susceptibility to *L. invasa* of these species (Mendel *et al.*, 2004; Thu *et al.*, 2009; Javaregowda & Prabhu, 2010; Zhu *et al.*, 2012). On the other hand, *E. moluccana*, *E. urophylla* × *E. grandis* and *E. urophylla* resulted tolerant to *L. invasa*, as previously shown by Thu *et al.* (2009), Nyeko *et al.* (2009) and Guerreiro *et al.* (2015).

This study suggests that in Misiones, *E. tereticornis* is the most sensible eucalyptus species to gall wasp attack, whereas other species and one hybrid of regional economic importance like *E. dunnii*, *E. camaldulensis*, *E. grandis* and *E. grandis* × *E. camaldulensis* presented low damage levels. The hybrid *E. urophylla* × *E. grandis* showed tolerance to *L. invasa*. More studies are necessary to determine the tolerance to *L. invasa* of different eucalyptus species and hybrids cultivated in the northern Argentine region.

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