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Notes and Comments

First detection of *Gonipterus platensis* (Coleoptera: Curculionidae) and its parasitoid *Anaphes nitens* (Hymenoptera: Mymaridae) in eucalyptus plantations in Minas Gerais, Brazil

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The area with forest plantations in Brazil is one of the largest in the world, 9.93 million hectares with 75.8% (7.53 million ha) of eucalyptus plantations with a mean annual productivity of 38.9 m^3/ha. These plantations are used to produce raw materials, mainly for the bioenergy, pulp and paper and laminate flooring sectors. Minas Gerais is the Brazilian state with the largest area of eucalyptus plantations, more than 2 million hectares, followed by the Mato Grosso do Sul and São Paulo states (IBÁ, 2022). Insect pests, especially exotic ones, damage these plantations.

Gonipterus platensis Marelli (Coleoptera: Curculionidae), native to Australia, is one of the main defoliator beetles of *Eucalyptus* spp. in the world (Schröder et al., 2020). In Brazil, reports of *Gonipterus* spp., date from the 1950s. *Gonipterus platensis* was first detected in 1979 in Curitiba, Paraná state, and spread to Rio Grande do Sul, Santa Catarina, and São Paulo (Wilcken and Oliveira, 2015). The last major territorial expansion of this insect in Brazil was in the early 2000s, damaging more than 50,000 hectares of *Eucalyptus grandis* x *Eucalyptus urophylla* ('urograndis') plantations in the Espírito Santo state (Wilcken and Oliveira, 2015) followed by its occurrence in the Bahia state (Nanini et al., 2022).

Adults and larvae of *G. platensis* feed on young leaves, mainly in the upper third of the plant, with larvae causing most of the damage. First and second instar larvae scrape the leaf surface without perforating the opposite epidermis and, in the third and fourth instars, feed on the entire leaf blade, causing complete defoliation of the tree crown, productivity losses, stag-horned or witches broom appearance (Schröder et al., 2020).

In Brazil, *G. platensis* damaged *Eucalyptus camaldulensis, E. dunnii*, *E. globulus*, *E. saligna*, *E. viminalis*, *E. grandis*, *E. urophylla*, *E. platyphylla* and hybrids of *E. grandis* x *E. urophylla* ('urograndis') and *E. grandis* x *E. dunnii* (Wilcken et al., 2008; Souza et al., 2016). The damage by this pest reduced the mean annual increment (MAI) of *E. grandis* in 10.4% and that of *E. grandis* x *E. dunnii* hybrids in 42.8% (Souza et al., 2016) in Brazil and more than 85% in volume at the end of the cycle in *E. globulus* plantations in Portugal (Reis et al., 2012).

The main management strategy for *G. platensis* is biological control with the egg parasitoid *Anaphes nitens* Girault (Hymenoptera: Mymaridae) (Schröder et al., 2020). This parasitoid was introduced and established in all regions of Brazil where this pest occurs (Ribeiro et al., 2023).

The objective is to report a new occurrence of *G. platensis* and its parasitoid in eucalyptus plantations in the Minas Gerais state, Brazil.

Forty adults and twenty egg capsules of *G. platensis* were collected in 9 months old *E. grandis* x *E. urophylla* plantations with low damage caused by adult feeding on the edge of the leaves in the municipalities of Uberlândia and Indianópolis (geographic coordinates -19.037930, -48.040757) and sent for taxonomic and molecular identification at the School of Agriculture of the São Paulo State University (FCA/UNESP). These materials were deposited in the entomological collection of the Plant Protection Department of the FCA/UNESP.

The head and pronotum of adult beetles were macerated and homogenized in a solution of 80 µL of 10% Chelex100 resin (Bio-Rad Laboratories, USA), and 8 µL of proteinase K (20 µg/mL) (Bioline, USA). The solutions' samples were transferred to an Infinigen thermocycler (model TC-96CG) and incubated at 95ºC for 20 minutes for DNA extraction.

The mitochondrial cytochrome c oxidase I (COI) gene was amplified by polymerase reaction (PCR) using specific primers to identify *G. platensis* and *Gonipterus pulverulentus* Lea (Coleoptera: Curculionidae) (Nanini et al., 2022) (Table 1).

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PCR reactions used the standards: 12.5µL of Taq DNA Polymerase (NeoBio); 3.5 µl of milliQ water; 1.5 µL of each Primer (10 µM) and 3.0 µL of subject DNA, totaling 25 µL per reaction. Each PCR cycle had 95ºC for 3 minutes; 35 cycles of 95ºC for 30s, 58ºC for 30s and 72ºC for 2 minutes, with final polymerization at 72ºC for 10 minutes. The positive control had *G. platensis* individuals reared at the laboratory and *G. pulverulentus* provided by Prof. Alberto S. Correa from the Luiz de Queiroz College of Agriculture of the University of São Paulo (ESALQ/USP). The column of the negative DNA control sample was replaced with miliQ water. PCR amplicons were visualized in a 1% agarose gel containing 80 mL of TBE buffer solution; 0.8g of agarose (Neo3Bio) and 0.4µl of DNA GelRed intercalator (NeoBio), in UV light transilluminator (Major Science).

The egg capsules of *G. platensis* were maintained in 5 cm diameter acrylic plates in a biochemical oxygen demand (B.O.D.) chamber at 25±1ºC; 60% RH and photoperiod 12:12h (light: dark) until parasitoid emergence.

The morphological and molecular analysis confirmed the adult insects collected as *G. platensis* and its detection in new productive areas of Eucalyptus in Brazil reveals its continue dispersion and adaptation to regions with different climatic conditions (Figure 1). The *E. urophylla* and the *E. grandis* x *E. urophylla* hybrids, among the most susceptible hosts to *G. platensis* (Oliveira et al., 2022) and widely planted in Brazil with high productivity, favor the dispersal of this insect.

The egg capsules of *G. platensis* collected were parasitized by *A. nitens*, confirming the joint dispersal of *G. platensis* and its natural enemy. However, outbreaks of this pest indicate the need to include other natural enemies along with *A. nitens* (Ribeiro et al., 2023), such as the native predatory stinkbug *Podisus nigrispinus* Dallas (Hemiptera: Pentatomidae) (Nascimento et al., 2017) to manage this pest. Commercial products using entomopathogenic fungi such as *Beauveria bassiana* (Jordan et al., 2021) have been registered at the Brazilian Ministry of Agriculture and entomopathogenic nematodes have been studied in the laboratory to manage this pest (Damascena et al., 2020)

Exotic pests such as *Glycaspis brimblecombei* Moore (Hemiptera: Aphalaridae), *Leptocybe invasa* Fisher and LaSalle (Hymenoptera: Eulophidae) and *Thaumastocoris peregrinus* Carpintero and Dellapé (Hemiptera: Thaumastocoridae) already reduce productivity of eucalyptus plantations in the Minas Gerais state (Wilcken et al., 2010), where the establishment of *G. platensis* is a cause of concern. The most significant area planted with Eucalyptus in Brazil is located on north of Minas Gerais. Control measures as biological control agent releases should be adopted to reduce and/or to prevent productivity losses caused by *G. platensis* and to restrict its dispersal.

Table 1. Primers used for the molecular identification of *Gonipterus platensis* and *Gonipterus pulverulentus* (Coleoptera: Curculionidae) (Nanini et al., 2022).

Primers	Sequence (5'-3')	Amplified species
Gpul_F	GGTATAGATGTAGATACGCGG	*G. pulverulentus*
Gpul_R	CGAGTACCGTGCAGGTATTCCTCTTAAT	*G. pulverulentus*
Gpla_R	TGAATATCGTCGAGGTATACCTCTTAAC	*G. platensis*
Guni_F	CATGATACTTATTATGTAGTTGCTC	*G. pulverulentus* and *G. platensis*

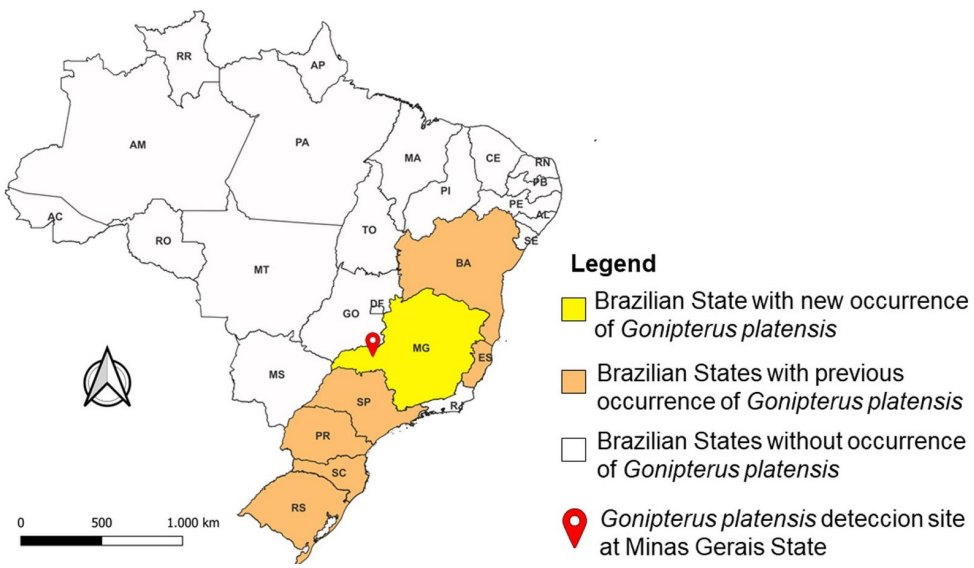

Figure 1. Geographic distribution of *Gonipterus platensis* (Coleoptera: Curculionidae) in Brazil with emphasis on the new record in Minas Gerais State.

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