New Disease Reports

First report of *Corynespora cassiicola* causing severe leaf blight on *Eucalyptus* in Brazil

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Eucalyptus species are the most widely cultivated forestry species in Brazil, currently covering over 5 million hectares. Recently, a new form of severe leaf blight was observed on all plants (about 2,880) of hybrid clones of *Eucalyptus* (*E. grandis* x *E. urophylla* and *E. grandis* x *globulus*), *E. benthamii* and *E. dunnii* grown in a glasshouse located at the Campus of the Universidade Federal de Viçosa (Viçosa, state of Minas Gerais, Brazil). So far, the disease has not been observed in the field. Lesions started on the leaves as small, circular to elliptical, light brown spots surrounded by a purple circle and a water-soaked area (Fig. 1). In addition, necrosis sometimes appeared on the shoot tips and developed into stem dieback. A dematiaceous hyphomycete fungus resembling *Corynespora* was frequently found sporulating on necrotic tissues.

To isolate and identify the causal agent of the disease, infected leaf tissues were surface sterilised in 70% ethanol for one minute, disinfected in 1% NaOCl for one minute, rinsed in sterile distilled water, plated on potato dextrose agar (PDA), and incubated at 28 ± 1°C for seven days. The fungus was characterised by gray to dark gray colonies; long, cylindrical brown conidiophores, 4.5-7.5 µm wide (Fig. 2); fusiform, obclavate to sub-cylindrical, straight or curved conidia, 51-165 µm x 5-9.5 µm, which formed singly, in acropetal chains, or through percurrent annelidic proliferations and were sub-hyaline to brown and 3-13 distoseptate, with a flat, darkened and thickened hilum (Fig. 2); features typical of *Corynespora cassiicola* (Berk. & Curt.) Wei (Ellis, 1971).

To confirm the identity of the fungus, genomic DNA was extracted using the Wizard® Genomic DNA Purification Kit (Promega, USA) and used as a template for amplification of the internal transcribed spacer (ITS) region of the rDNA with the primer pair ITS1/ITS4. The resulting sequence was deposited (GenBank Accession No. KF258591) and had 100% nucleotide identity with corresponding sequences of *C. cassiicola* (JQ801302, JX087447 and JQ595296). This species has a broad worldwide distribution in warm climates and has been reported from over 300 hosts, including tree species of the genus *Tabebuia* (Ferreira & Alfenas, 1980). The species includes populations with a high degree of host specificity, which have been recognised at the level of *forma specialis* (Pereira *et al*., 2003). A representative specimen was deposited in the local herbarium at the Plant Disease Clinic of the Universidade Federal de Viçosa (VIC39663). A type culture was also deposited in the local culture collection (COAD 1107).

Thirty-day-old healthy plants of the hybrid clone *E. grandis* x *E. urophylla* were spray-inoculated until run-off with a spore suspension (1 x 10 spores/ml) prepared from seven-day-old cultures of *C. cassiicola* grown on PDA. Uninoculated plants sprayed with distilled water served as controls. After inoculation, plants were kept in the dark in a mist chamber with intermittent irrigation for 24 h at 25°C and subsequently transferred to a glasshouse. After seven days, leaf spots were observed on inoculated plants but not on water-sprayed controls. Only *C. cassiicola* was isolated from necrotic tissues, fulfilling Koch's postulates. To date, there has only been one previous report of *C. cassiicola* on eucalyptus (*Eucalyptus grandis*), associated with 'water-soaked spots in India' (Wilson & Rema Devi, 1966). The work presented here is the first report of leaf spot caused by *C. cassiicola* on *Eucalyptus* in Brazil.

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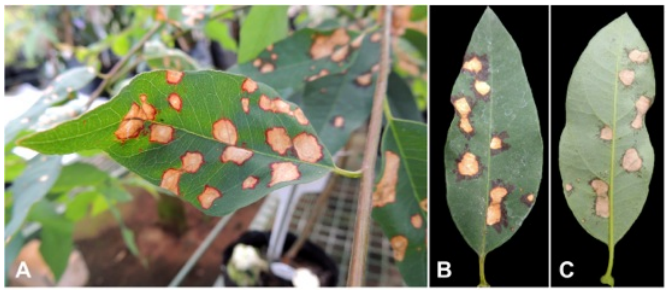

Figure 1

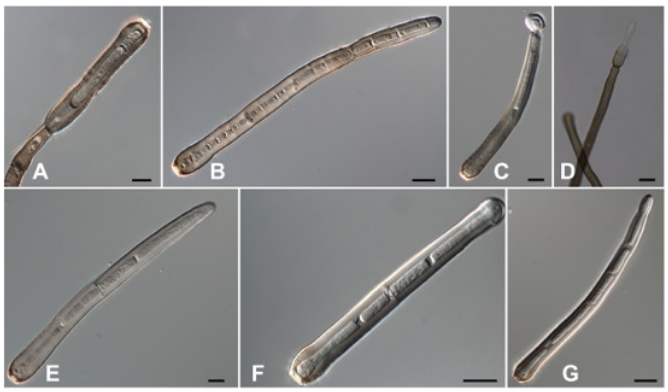

Figure 2

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