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Two *Cryphonectriaceae* species from *Eucalyptus* leaves in North Sumatra and their stem inoculation outcomes

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Abstract: Cryphonectria canker, caused by a number of genera and species in the family *Cryphonectriaceae* (*Diaporthales*, *Ascomycota*), is an important disease affecting woody shrubs and trees in the *Melastomataceae* and *Myrtaceae* (*Myrtales*). Initially reported as a stem canker disease on *Eucalyptus* in Cuba, it has since emerged as an important threat, particularly in tropical regions and the Southern Hemisphere where *Eucalyptus* is extensively cultivated for commercial forestry. Many species of *Cryphonectriaceae* can occur as asymptomatic endophytes in different plant parts, causing disease only when the hosts are exposed to biotic or abiotic stresses. During 2018, isolations from healthy *Eucalyptus* leaf tissues adjacent to symptoms caused by *Elsinoe necatrix* in North Sumatra, Indonesia resulted in two isolates resembling species of the *Cryphonectriaceae*. Multi-locus DNA sequence comparisons (ITS+LSU+*TUB1*+*TUB2*+*TEF1*+*RPB2*), as well as morphological observations, showed that these fungi represented a novel *Celoporthe* (*Cel.*) species, described here as *Cel. endophylla sp. nov*. and the new genus *Aureofoliicola eucalypti gen. et sp. nov.* Inoculation trials on 4-mo-old plants of two *Eucalyptus* genotypes showed that *Cel. endophylla* can cause lesions significantly different to those from the controls, while *A. eucalypti* did not produce lesions on inoculated stems. These findings underscore a biosecurity risk posed by apparently latent fungal pathogens that persist undetected within asymptomatic plant tissues.

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INTRODUCTION

In Southeast Asia, short-rotation plantation forestry has experienced significant expansion over the past few decades, driven by an increasing demand and production capacity of the regional pulp and paper industry (Harwood & Nambiar 2014, Healey *et al.* 2022). These plantations now span over 7 M hectares and are dominated by fast-growing, non-native Australian tree species (Harwood & Nambiar 2014). In Indonesia, the widespread decline of *Acacia mangium* (Tarigan *et al.* 2011) has led to the increasing establishment of *Eucalyptus* plantations. These primarily consist of *E. pellita*, *E. grandis* and *E. urophylla*, along with their interspecific hybrid genotypes (Harwood & Nambiar 2014). This large-scale conversion to *Eucalyptus* monocultures has introduced substantial new challenges, notably the emergence of pests and diseases that threaten the sustainability of the local forestry industry (Wingfield *et al.* 2008, 2015, Hurley *et al.* 2016).

The *Cryphonectriaceae* (*Diaporthales*, *Ascomycota*) includes numerous fungal taxa that are important pathogens of woody plants and shrubs globally (Gryzenhout *et al.* 2009, Wingfield *et al.* 2025). The most notable of these is *Cryphonectria parasitica*, the causal agent of chestnut blight, responsible for the near destruction of *Castanea dentata* (*Fagales*) in North America (Anagnostakis 1987, Hepting 1974, Rigling & Prospero 2018). While early research focused primarily on *Cryphonectriaceae* species in the Northern Hemisphere associated with hosts in the *Fagales*, the discovery of Cryphonectria canker on *Eucalyptus* in Cuba (Bruner 1917) signalled a broader geographic and host range for these fungi. This finding prompted more intensive surveys in tropical and Southern Hemisphere regions, leading to the discovery of many novel species in new genera including *Chrysoporthe*, *Celoporthe*, *Holocryphia, Xanthoporthe* and *Cryptometrion*, many of which are associated with cankers on *Myrtales*, including *Eucalyptus* (Gryzenhout *et al.* 2004, 2009, Wingfield *et al.* 2025).

In Indonesia, several species of *Cryphonectriaceae* have been identified over the past two decades, some of which are of particular concern to plantation forestry. *Chrysoporthe deuterocubensis* has been frequently isolated from diseased *Eucalyptus* stems and from other shrubs belonging to the *Myrtaceae* and *Melastomataceae*, and it has been shown to cause disease on planted *Eucalyptus* trees in pathogenicity trials (Tarigan *et al.* 2024). In North Sumatra and Riau, two *Celoporthe* spp. have been isolated from native *Syzygium* species, namely *Cel. indonesiensis* (Chen *et al.* 2011), and *Cel. foliorum* (Chen *et al.* 2022), but their potential to cause disease on *Eucalyptus* has not been fully evaluated. Additionally, *Cryptometrion aestuescens*, a novel species described by Gryzenhout *et al.* (2010), was reported to cause severe stem cankers on *E. grandis* clonal hedges in North Sumatra.

Recent studies have shown that species of the *Cryphonectriaceae* can naturally exist as endophytes in healthy

tissues of trees and woody shrubs in the *Myrtales* (Gryzenhout *et al.* 2006d, Mausse-Sitoe *et al.* 2016, Grannados *et al.* 2020, Roux *et al.* 2020, Chen *et al.* 2022, Wingfield *et al.* 2025). These fungi typically do not produce disease symptoms unless the host plant is subjected to environmental or biotic stress. Endophytic *Cryphonectriaceae* species can be found inhabiting various parts of trees, including stems (Beier *et al.* 2017), branches (Mausee-Sitoe *et al.* 2016, Granados *et al.* 2020), shoots (Bissegger *et al.* 1994), bark (Coelho *et al.* 2021) and sprouts (Guérin & Robin 2003). Their latent presence within asymptomatic plant tissue poses significant biosecurity risks, as they may escape detection and later emerge as aggressive pathogens under favourable host and environmental conditions (Mausse-Sitoe *et al.* 2016, Granados *et al.* 2020, Wingfield *et al.* 2025).

During 2018 surveys of the fungal community associated with Eucalyptus scab and shoot malformation caused by *Elsinoe necatrix* in North Sumatra, Indonesia (Pham *et al.* 2021), two isolates resembling members of the *Cryphonectriaceae* were isolated from asymptomatic *Eucalyptus* leaf tissues. The objectives of this study were to identify these isolates based on multi-locus DNA sequence data and morphological characteristics, to assess their relative pathogenicity on different *Eucalyptus* genotypes, and thus to determine whether they pose a potential risk to commercial forestry in Indonesia.

MATERIALS AND METHODS

Sample collection and fungal isolations

Fresh branchlets of a *E. grandis* × *urophylla* clone, bearing leaves with scab-like necrotic spots caused by *Elsinoe necatrix* (Pham *et al.* 2021) were collected in Tele, North Sumatra, placed in sealable plastic bags and transferred to a laboratory for fungal isolations. Leaf samples were surface disinfected with 70 % ethanol and then washed with sterile distilled water. Apparently healthy leaf tissue adjacent to the necrotic scabs was cut into 1 mm^2 pieces and placed onto 2 % malt extract agar (MEA; Biolab, Midrand, South Africa) supplemented with streptomycin sulphate (100 ppm) and incubated at 25 °C for 3–7 d. Emerged fungal colonies resembling the *Cryphonectriaceae* were transferred to fresh MEA plates. Primary isolations were incubated for 3–7 d at 25°C for fungal growth. Single hyphal tips of fungal colonies were sub-cultured to new MEA plates to obtain pure isolates. The isolates were deposited in the research culture collection (CMW) of the Forestry and Agricultural Biotechnology Institute (FABI) and the publicly accessible culture collection (CMW-IA) of Innovation Africa at the University of Pretoria, South Africa, and the culture collection (CBS) of the Westerdijk Fungal Biodiversity Institute, Utrecht, the Netherlands. Dried cultures representing the holotype of the novel taxa were deposited in the H.G.W.L. Schweickerdt herbarium (PRU), University of Pretoria, South Africa.

DNA extraction, PCR amplification, and sequencing

Genomic DNA was isolated from 7-d-old cultures grown on 2 % MEA at 25 °C using Prepman® Ultra Sample Preparation Reagent (Thermo Fisher Scientific, Waltham, MA, USA), following the manufacturer's instructions. Amplification of the nuclear internal transcribed spacer regions 1 and 2 (ITS), including the 5.8S rRNA gene, was performed using primers ITS1 and ITS4 (White *et al.* 1990), the nuclear large subunit (LSU) of rRNA using LR0R and LR5 (Vilgalys & Hester 1990, Rehner & Samuels 1994), two regions of β-tubulin gene (*TUB1* and *TUB2*) using Bt1a/Bt1b and Bt2a/Bt2b (Glass & Donaldson 1995), the second largest subunit of RNA polymerase II (*RPB2*) with RPB2-5F2 and fRPB2-7cR (Liu *et al.* 1999, Sung *et al.* 2007) and the translation elongation factor 1-alpha (*TEF1*) using EF1-728F and EF1-986R (Carbone & Kohn 1999). The PCR amplifications were conducted in 13 µL reaction volumes containing 1 µL of genomic DNA, 5× MyTaq buffer (Bioline, London, UK), 0.25 µL of MyTaq DNA polymerase, 0.5 µL of each primer (10 µM), and sterile deionised water. Amplified products were cleaned using ExoSAP-IT™ (Thermo Fisher Scientific) and sequenced in both directions. Sequence assembly and editing were conducted in Geneious Prime v. 2025.1.2 (https://www.geneious.com).

Phylogenetics analyses

Sequences generated in this study were aligned with reference sequences of closely related *Cryphonectriaceae* taxa (Table S1) using MAFFT v. 7 (Katoh & Standley 2013; http://mafft.cbrc.jp/alignment/server/). The resulting alignments were manually checked, trimmed and concatenated using Geneious Prime. Maximum likelihood phylogenetic analyses were performed using W-IQ-TREE (Trifinopoulos *et al.* 2016). Partition-specific substitution models were chosen using ModelFinder (Kalyaanamoorthy *et al.* 2017), and branch support was evaluated using 1000 Ultrafast bootstrap replicates (UFBoot; Hoang *et al.* 2018). Additionally, the Shimodaira–Hasegawa-like approximate likelihood ratio test (SH-aLRT) was conducted with 1000 replicates to further assess node support (Guindon *et al.* 2010). *Diaporthe eres* (LC3198) and *Cytospora ailanthicola* (CFCC 89970) were included as outgroup taxa. The resulting phylogenetic tree was visualised using FigTree v. 1.4.5 (https://github.com/rambaut/figtree).

Morphology

Fungal fruiting structures were induced in the laboratory by transferring a piece of fungal culture onto water agar (WA, 20 g Difco Bacto agar in 1 L water) with autoclaved *Eucalyptus* twigs and leaves placed on the agar surface. The plates containing the twigs and leaves were incubated at 25 °C in the dark for 4 weeks. When fruiting bodies developed on the plant tissue, they were dissected with a scalpel to expose the inner structures including spores. The structures were initially mounted on slides in water and later replaced with 85 % lactic acid, in which they were studied, measured, and images captured. Observations were made using a microscope (Nikon SMZ18, Eclipse N*i*, Tokyo, Japan) mounted with a camera (Nikon DS-Ri2) and an imaging software (NIS-Elements BR 4.30.00). Up to fifty measurements were made depending on availability. Dimensions were presented as minimum–maximum (average ± standard deviation) (n = number of observations). The pieces of twig and leaf containing fungal fruiting structures were sectioned vertically using a cryomicrotome (Leica Biosystems CM1520, Germa-

ny) to study their position in the substrate tissue. They were cut to 10–12 µm thickness, and the sections were mounted in 85 % lactic acid for observation.

A growth study was carried out on 2 % MEA in 90 mm Petri dishes. Five replicate plates were incubated at seven temperatures ranging from 5–35°C, at 5 °C intervals. Plugs of mycelium (5 mm) taken from 7-d-old cultures were placed at the centres of Petri dishes with mycelial side down, and the plates were maintained in incubators at the various growth temperatures in the dark. The study was terminated when the mycelial growth of the isolates at the optimum temperature reached the edges of the plates. Two diameter measurements perpendicular to each other were taken for each culture, and the averages for the replicate plates were computed. Culture characteristics were noted when the isolates were 14-d-old, and culture colours were described following the ISCC-NBS system.

Pathogenicity tests

Two isolates (CMW 55025 and CMW 55026) were selected for a pathogenicity trial to evaluate their relative aggressiveness on *Eucalyptus*. Inoculations were performed on 16-wk-old plants of a *E. grandis* × *pellita* (GP) and a *E. pellita* (EP) genotype, grown in 25-cm-diam. plastic planting bags and maintained in a greenhouse at approximately 28 °C. For each clone, 20 plants were inoculated per isolate, and an equal number of trees were included as negative controls.

A sterile scalpel was used to excise a 1 cm bark flap approximately 10–15 cm above the soil line to expose the cambial tissue. A 5-mm-diam. mycelial plug taken from the actively growing margin of a 7-d-old culture on potato dextrose agar (PDA; BD Difco, NJ, USA) was inserted into each wound with the mycelium facing the cambium. A sterile PDA disc was used for the control inoculations that were conducted in the same manner. Inoculation sites were sealed with Parafilm®M (Amcor, Zürich, Switzerland) to prevent desiccation and contamination. Lesion development was assessed after 6 wk by removing the bark at each wound site and measuring the length of the discoloured cambial tissue. Re-isolations were made from fruiting structures that formed on the bark and cambium after incubation in a moist chamber for seven days so as to verify the presence of the inoculated fungi.

Lesion length data were analysed using GraphPad Prism v. 10.4.1. Outliers were detected using the robust regression and outlier removal (ROUT) method with a coefficient value (Q) of 10 % (Motulsky & Brown 2006). The normality of the data was assessed using the Shapiro-Wilk test (Shapiro & Wilk 1965), and homogeneity of variances was tested using the Brown-Forsythe test (Brown & Forsythe 1974). A Kruskal-Wallis test was then applied, followed by Dunn's post-hoc test for pairwise comparisons at a 5 % significance level (Dunn 1964).

RESULTS

Fungal isolates, DNA sequencing and phylogenetic analysis

Isolations from apparently healthy *Eucalyptus* leaf tissue associated with *Elsinoe necatrix* symptoms resulted in a collection of approximately 150 fungal cultures (data not shown). Among these, two isolates resembling *Cryphonectriaceae* species were obtained. Amplicons of approximately 600 bp for the ITS, 850 bp for LSU, 500 bp for *TUB1*, 500 bp for *TUB2*, 650 bp for *TEF1*, and 900 bp for *RPB2* were generated for all the isolates. The combined sequence dataset for the six regions used for phylogenetic analysis included 100 ingroup taxa and contained 4256 characters, including alignment gaps. Based on the results of ModelFinder, a TIM2e+I+G4 model was selected for ITS and LSU, the HKY+F+I+G4 for *TUB1* and *TUB2,* the TPM2+F+I+G4 for *TEF1*, and the TN+F+G4 for *RPB2*, and these models were applied to individual partitions in the concatenated dataset for the ML analyses. The ML tree with UFBoot values and SH-aLRT branch supports is presented in Fig. 1. The isolate CMW 55025 grouped together with species of *Celoporthe*, but formed a lineage clearly distinct from other species in this genus. The isolate CMW 55026 formed a unique lineage, representing a sister lineage to *Pseudocryphonectria* and is introduced here as a novel genus in the *Cryphonectriaceae*.

Taxonomy

Celoporthe endophylla N.Q. Pham, Marinc., M.J. Wingf., ***sp. nov.*** MB 862245. Fig. 2.

Etymology: Name refers to its apparently endophytic lifestyle.

Diagnosis: Morphologically distinguishable by the absence of paraphyses and long attenuated apices of its conidiogenous cells.

Typus: **Indonesia**, North Sumatra, Tele, leaf of *Eucalyptus* sp. infected with *Elsinoe necatrix*, Apr. 2018, *M.J. Wingfield & N.Q. Pham* [**holotype** PRU(M) 4641, dried culture and twigs with artificially induced conidiomata; ex-holotype cultures CMW-IA 7269, CMW 55025, CBS 147167]. GenBank: PX727429 (ITS); PX727433 (LSU); PX733386 (*TUB1*); PX733388 (*TUB2*); PX733390 (*TEF1*); PX733392 (*RPB2*).

Description: On *Eucalyptus* twigs and leaves, where conidiomata were artificially induced. *Sexual morph* not observed. *Conidiomata* immersed, becoming erumpent, stromatic, uni- or multi-locular, convoluted, 85–404 (228.9 ± 86.61) µm long (n = 29), 87–407 (185.7 ± 73.09) µm wide (n = 29). *Conidiomatal walls* pseudoparenchymatous, textura angularis, 6–36 (19.2 ± 6.99) µm thick (n = 32), consisting of a few layers or thick-walled, brown cells in outer stratum, becoming thinner, paler, more flattened towards inner stratum. *Conidiophores* hyaline, borne along inner walls of locules, cylindrical, simple or branched, septate or aseptate, 4–13 (7.6 ± 2.91) µm long (n = 25), 1–3 (2 ± 0.34) µm wide (n = 25). *Paraphyses* not observed. *Conidiogenous cells* hyaline, enteroblastic, determinate, discrete or integrated, flask-shaped with attenuated apices, occasionally as aperture borne just below septum, 5–9 × 2–3 (7.0 ± 1.11 × 2.0 ± 0.27) µm (n = 25). *Conidia* hyaline, aseptate, fusiform to allantoid with pointed base, straight or slightly curved, exudating in cirrhus, yellowish in mass, 3–5 × 2 (4.1 ± 0.43 × 1.8 ± 0.15) µm (n = 50).

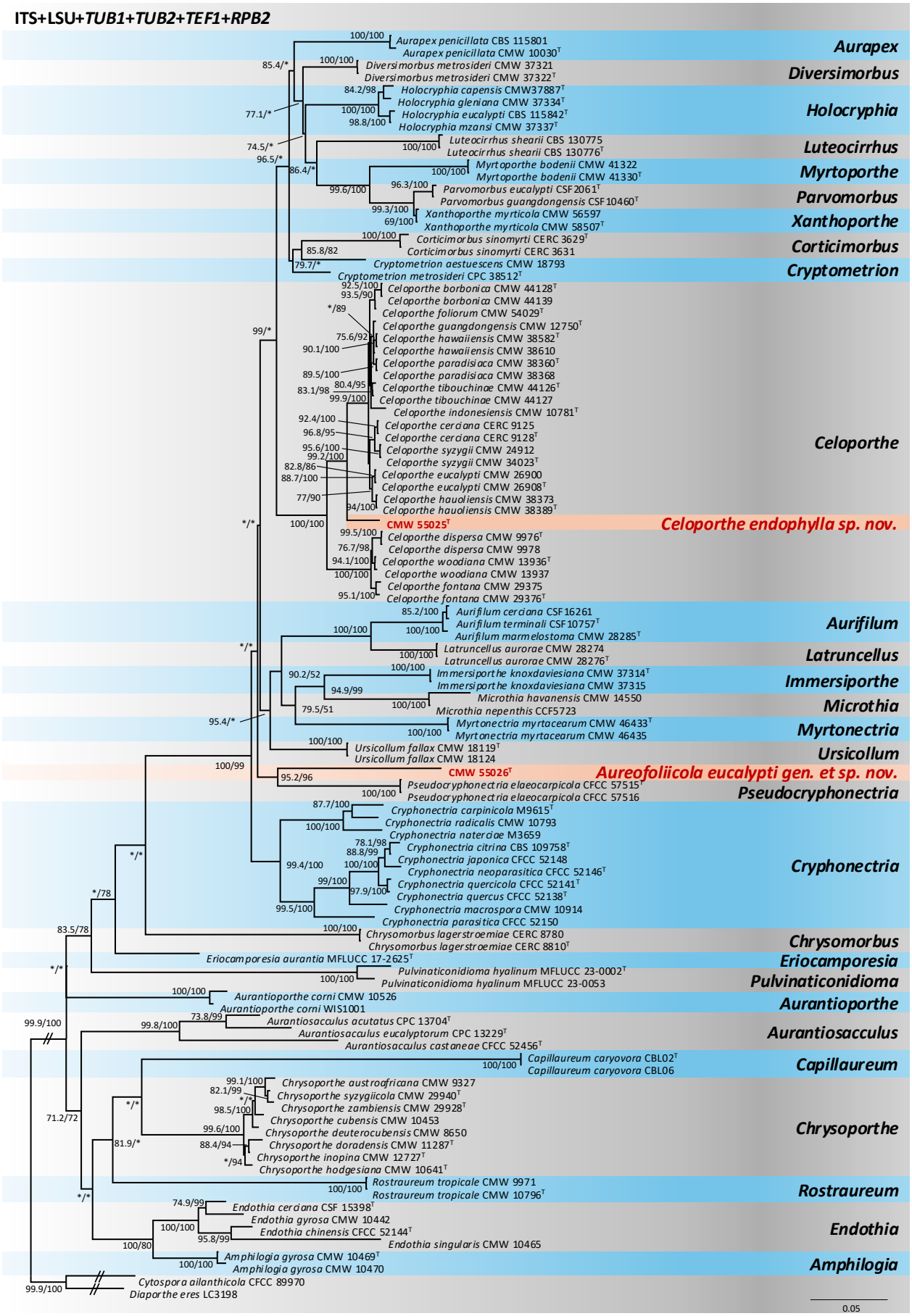

Fig. 1. Phylogenetic tree based on a Maximum Likelihood (ML) analysis of a combined DNA data set of ITS, LSU, *TUB1*, *TUB2, TEF1* and *RPB2* sequences for *Cryphonectriaceae* species. Isolates sequenced in this study are presented in red and boldface. SH-aLRT branch support and UFBoot values ≥ 70 % for ML analyses are indicated at the nodes as SH-aLRT/UFBoot. Branch support values < 70% are marked with "*". Isolates representing ex-type cultures are marked with a "T". *Diaporthe eres* (LC3198) and *Cytospora allanthicola* (CFCC 89970) represent the outgroup taxa.

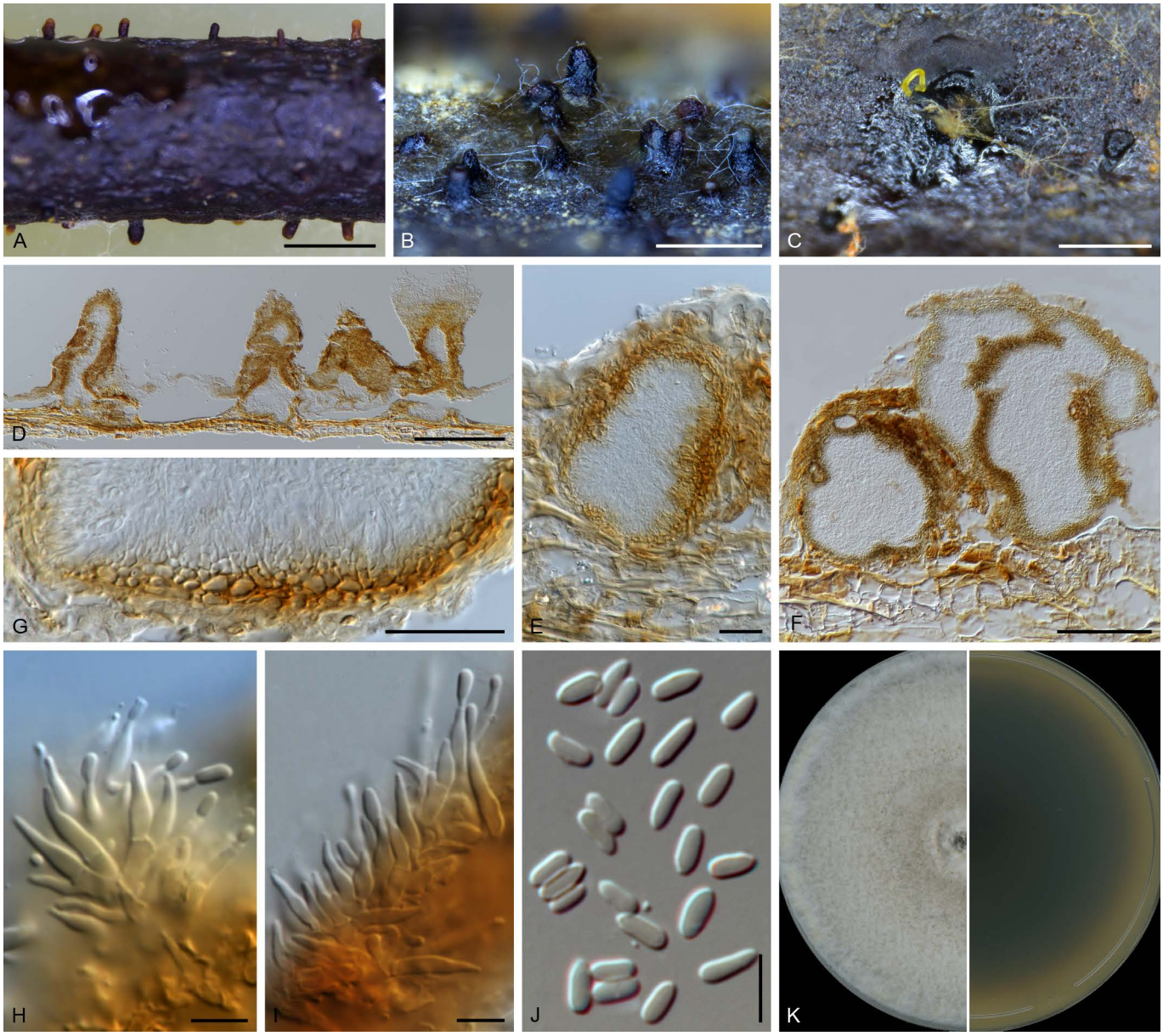

Fig. 2. Micrograph of *Celoporthe endophylla sp. nov.* [Holotype PRU(M) 4641; ex-holotype culture CMW-IA 7269, CMW 55025, CBS 147167]. **A, B.** Conidiomata formed on *Eucalyptus* twig in the laboratory. **C.** Conidia oozing in a cirrhus. **D–F.** Configuration of conidioma in twig (D, F) and leaf (E). **G.** Conidiomatal wall and inner layers producing conidiophores. **H, I.** Conidiophores and conidiogenous cells. **J.** Conidia. **K.** Colony at optimum temperature (25 °C) on 2 % MEA in the dark for 14 d (left: upper, right: under). Scale bars: A = 1 mm; B, C = 500 µm; D, G = 250 µm; F = 100 µm; E = 25 µm; H–J = 5 µm.

Culture characteristics: *Growth* on 2 % MEA in the dark, optimal growth at 25 °C, reaching 71 mm diam. in 7 d, followed by 20 °C (52 mm), 30 °C (38 mm), 15 °C (29 mm), and 10 °C (9.8 mm). No growth at 5 °C and 35 °C. *Culture morphology* on 2 % MEA in 14 d in the dark, showing circular growth, even edge; *mycelia* fluffy texture, medium dense; *colours* at 10 °C very dark red becoming paler towards edges, covered with white aerial mycelia, reverse dark brown; 15 °C on black background, covered with light yellowish brown to deep orange yellow aerial hyphae, reverse black; 20 °C on black background with greyish yellow to dark greyish yellow, reverse black; 25 °C on black background with light greyish yellowish brown, paler towards edges, pigmented water droplets or exudates present, reverse black; 30 °C on black background with moderate yellowish brown centre becoming paler towards edges, edges often white to yellowish white, raised, reverse black.

Notes: *Celoporthe endophylla* is the 15th species of the most species-rich genus in the *Cryphonectriaceae*. It was isolated from an asymptomatic *Eucalyptus* leaf tissue, in contrast to *Cel. foliorum*, another foliicolous species in the genus, isolated from a leaf spot on a *Syzygium* sp. (*Myrtaceae*) (Chen *et al.* 2022). Both species occurring on leaf tissues lack paraphyses, which separate them from other *Celoporthe* species, which are corticolous and have paraphyses. *Celoporthe endophylla* has a larger conidial range (3–5 × 2 µm, av. 4.1 × 1.8 µm) than *Cel. foliorum* (3–4 × 1–2 µm, av. 3.6 × 1.5 µm). Conidiogenous cells (phialides) of *Cel. endophylla* have attenuated apices that are more elongated than those of *Cel. foliorum*.

Aureofoliicola N.Q. Pham, Marinc., M.J. Wingf., ***gen. nov.*** MB 862249.

Etymology: Name refers to its yellow spore masses and foliicolous lifestyle.

Type species: *Aureofoliicola eucalypti* N.Q. Pham, Marinc., M.J. Wingf.

Description: *Conidiomata* stromatic, unilocular. *Conidiophores* filiform, septate. *Conidiogenous cells* enteroblastic, determinate, integrated. *Conidia* hyaline, fusiform to cylindrical.

Aureofoliicola eucalypti N.Q. Pham, Marinc. M.J. Wingf., ***sp. nov.*** MB 862253. Fig. 3.

Etymology: Name refers to its host plant, *Eucalyptus* sp.

Diagnosis: Morphologically identifiable by its filiform conidiophores and lack of paraphyses.

Typus: **Indonesia**, North Sumatra, Tele, leaf of *Eucalyptus* sp. infected with *Elsinoe necatrix*, Apr. 2018, *M.J. Wingfield & N.Q. Pham* [**holotype** PRU(M) 4642, dried culture and twigs with artificially induced conidiomata; ex-holotype culture CMW-IA 7270, CMW 55026, CBS 147168]. GenBank: PX727430 (ITS); PX727434 (LSU); PX733387 (*TUB1*); PX733389 (*TUB2*); PX733391 (*TEF1*); PX733393 (*RPB2*).

Description: On *Eucalyptus* twigs and leaves, where conidiomata were artificially induced. *Sexual morph* not observed. *Conidiomata* stromatic, unilocular, convoluted, 182–587 (372.0 ± 100.48) µm long (n = 25), 153–478 (315.0 ± 82.98) µm wide (n = 25). *Conidiomatal wall* pseudoparenchymatous, 16–39 (25.8 ± 6.62) µm thick (n = 25), composed of three strata, outer stratum consisting of a few layers of thick-walled, brown cells, often mixed with plant tissue, mid-stratum consisting of thick-walled, sub-hyaline, compressed cells, inner stratum consisting of a few thin-walled, hyaline and highly compressed cells. *Conidiophores* hyaline, filiform, septate, simple or scarcely branched near

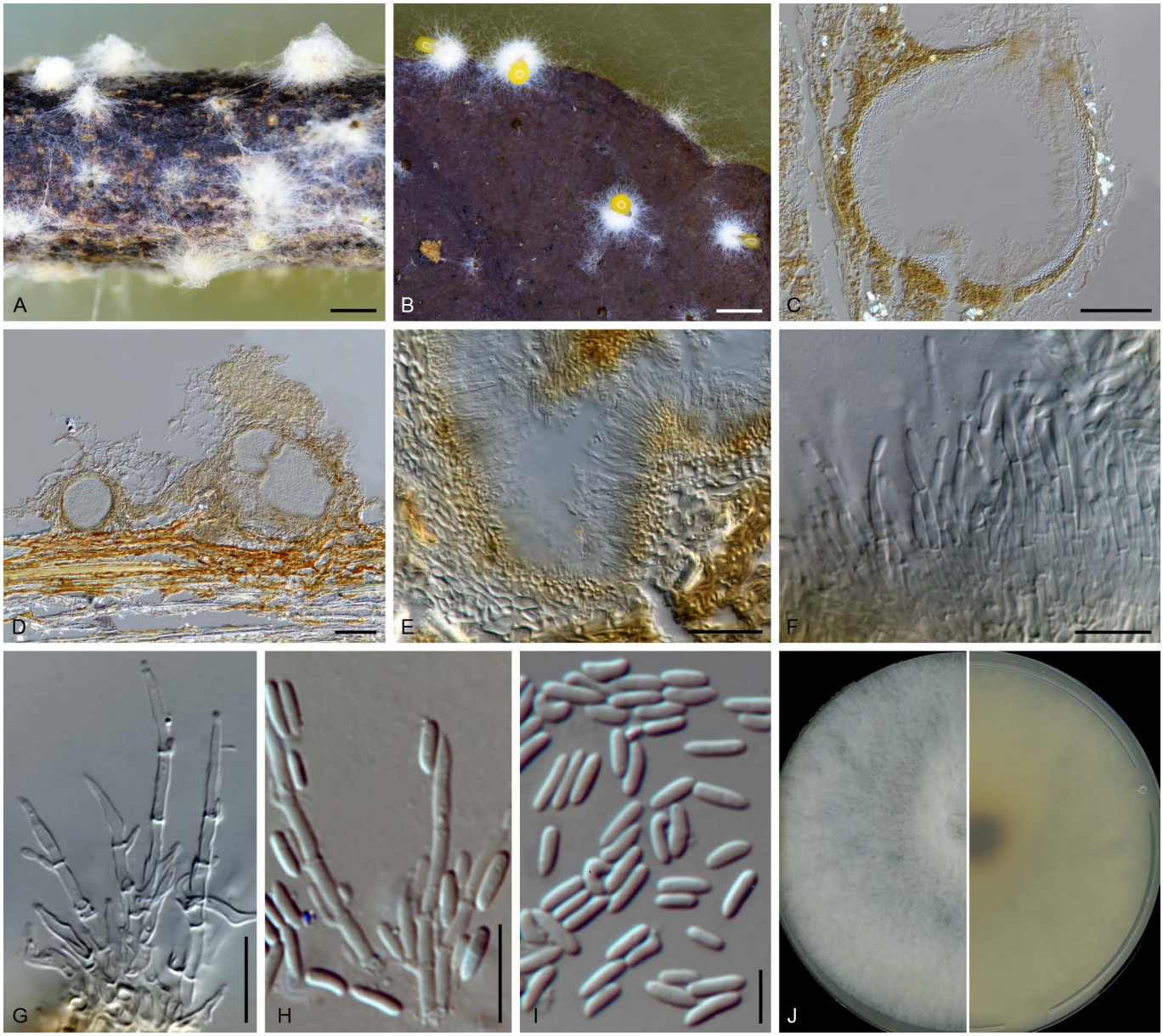

Fig. 3. Micrograph of *Aureofoliicola eucalypti gen. et. sp. nov*. [Holotype PRU(M) 4642; ex-holotype culture CMW-IA 7270, CMW 55026, CBS 147168]. **A, B.** Conidiomata formed on *Eucalyptus* twig (A) and leaf (B) in the laboratory. **C, D.** Vertical section of conidioma on leaf (C) and twig (D). **E.** Conidiomatal wall and conidiophores borne inside locule. **F.** Close-up of conidiophores born on the inner wall. **G, H.** Filiform conidiophores and integrated conidiogenous cells, apical or lateral beneath the septum. **I.** Conidia. **J.** Colony at optimum temperature (25 °C) on 2 % MEA in the dark for 14 d (left: upper, right: under). Scale bars: A, B = 1 mm; C, D = 100 µm; E = 25 µm; F–H = 10 µm; I = 5 µm.

base, borne along inner layers of locules, 18–45 (27.4 ± 7.05) µm long (n = 25). *Conidiogenous cells* enteroblastic, apical or lateral beneath septum, integrated, determinate, hyaline, cylindrical, tapered to apex when terminal, 8–13 (10.6 ± 1.49) µm long (n = 25) when apical, 5–8 (6.9 ± 0.86) µm long (n = 25) when lateral, 1–2 (1.6 ± 0.13) µm wide (n = 25). *Conidia* hyaline, fusiform to cylindrical, aseptate, straight or slightly curved, apex round, base pointed, exudating in cirrhus, yellowish in mass, 4–6 × 1–2 (5.0 ± 0.58 × 1.6 ± 0.16) µm (n = 50).

Culture characteristics: *Growth* on 2 % MEA, optimal growth at 25 °C reaching 71 mm in 7 d, followed by 20 °C (54 mm), 15 °C (30 mm), 30 °C (23 mm), 10 °C (11 mm), and no growth at 5 °C and 35 °C. *Culture morphology* on 2 % MEA in 14 d in dark, showing circular growth, even edge; *mycelia* texture fluffy texture, medium sparse density; *colours* at 10 °C yellowish white, sparse dense becoming less dense towards edges, reverse pale yellow with dark olive brown limited to centre; 15 °C centre yellowish grey aerial hyphae towards edges yellowish white, reverse pale yellow background with dark red brown limited to centre; 20 °C yellowish white, yellowish grey aerial hyphae over colony with dark olive brown dotted structures, reverse pale yellow background with dark red limited to centre; 25 °C mixture of aerial hyphae of medium grey, moderate olive brown, dark greyish yellow in concentric zones, reverse pale yellow background with dark reddish brown limited to centre or as random patches; 30 °C dark olive brown to deep yellowish brown becoming paler towards edges, reverse the same.

Notes: *Aureofoliicola eucalypti* is phylogenetically closely related to the monospecific *Pseudocryphonectria. Pseudocryphonectria elaeocarpicola* causes a stem blight on *Elaeocarpus* sp. (*Elaeocarpaceae*, *Oxalidales*) (Huang *et al.* 2022). They are, however, morphologically distinct from each other. *Pseudocryphonectria* has both obclavate and cylindrical conidia, and conidiophores that are often reduced to ampulliform conidiogenous cells. In contrast, *Aureofoliicola* has conidia that are all fusiform and septate filiform conidiophores, which distinguishes it from other genera in the *Cryphonectriaceae*.

Pathogenicity tests

Six weeks after inoculation, *Cel. endophylla* (CMW 55025) produced brown to dark brown lesions around the inoculation points on the tested plants (Fig. 4). The lesion lengths associated with each treatment showed heterogeneity of variance ($F_{(3,76)} = 77.59$, $p < 0.0001$) and did not follow a normal distribution ($p < 0.05$). No outlier was detected using the ROUT method. A significant difference in lesion length among different treatments was seen in the Kruskal-Wallis test ($H_{(3)} = 76.24$; $p < 0.0001$). Post-hoc Dunn's tests revealed significant differences between lesion length caused by CMW 55025 on clone EP ($\bar{x} = 343.00$ mm) compared to clone GP ($\bar{x} = 36.05$ mm), and these were longer than those in the controls ($p < 0.05$). *Celoporthe endophylla* was re-isolated from lesions on the inoculated trees but never from the controls. In contrast to *Cel. endophylla*, no lesions developed in association with inoculations using *A. eucalypti* (CMW 55026).

DISCUSSION

Two novel taxa in the *Cryphonectriaceae*, a family that includes numerous important canker pathogens of woody plants, were discovered in this study. Multi-locus DNA sequence comparisons for six regions, as well as morphological observations, showed that the isolates represented a new species of *Celoporthe*, described here as *Celoporthe endophylla sp. nov*., and a new genus and species, *Aureofoliicola eucalypti gen. et sp. nov*. The discovery of these two taxa highlights the hidden diversity of *Cryphonectriaceae* on *Myrtales* hosts and suggests that others remain to be discovered in Southeast Asia.

To date, six foliicolous species, including two novel species in this study, are reported in the family *Cryphonectriaceae*, where the remaining 70 species are corticolous, and most of them cause stem canker diseases (Wingfield *et al*. 2025). Among the foliicolous species, three were isolated from leaf spots: *Aurantiosacculus* (*Aur.*) *acutatus*, *Aur. eucalyptorum* (Crous *et al.* 2012) and *Cel. foliorum* (Chen *et al.* 2022), and the remaining species, *Microthia nepenthis* (Koukol *et al*. 2018), *Cel. endophylla* and *A. eucalypti*, were from

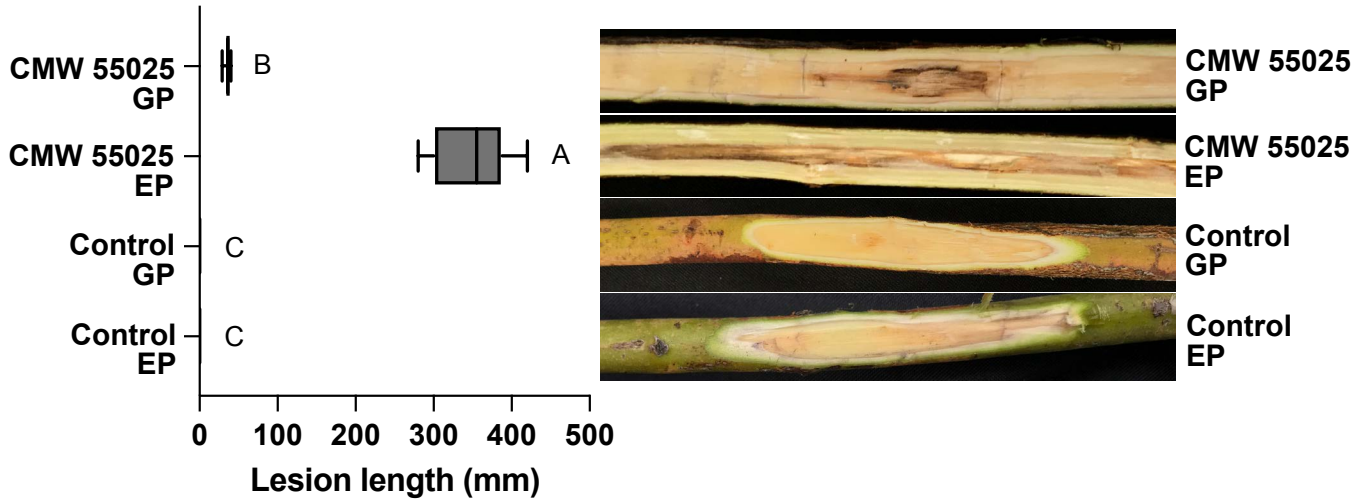

Fig. 4. Lesion length (mm) of *Celoporthe endophylla* (CMW 55025) on a *E. pellita* (EP) and a *E. grandis* × *pellita* (GP) genotype. Different letters indicate significant differences in pathogenicity based on the Dunn's post-hoc test ($p < 0.05$). Whiskers indicate min–max values. *Aureofoliicola eucalypti* (CMW 55026) was not included in the graph as it did not produce lesions on inoculated stems and was not different from the controls.

necrotised or asymptomatic leaves. Foliicolous species of the *Cryphonectriaceae* are relatively rare, occupy an unusual ecological niche, which is poorly understood, and not typically associated with disease symptoms. They could, however, be pathogens and provide inoculum for stem infections; a question that was considered in this study.

The genus *Celoporthe* was originally established by Nakabonge *et al.* (2006) to accommodate fungi closely related to *Chrysoporthe*, but distinguishable on the basis of DNA sequence data and morphology (Gryzenhout *et al.* 2009, Chen *et al.* 2022). Five *Celoporthe* spp. are known from Africa (Nakabonge *et al.* 2006, Vermeulen *et al.* 2013, Ali *et al.* 2018), three from the Pacific region (Roux *et al.* 2020), and six from Southeast Asia and South China (Wang *et al.* 2018, Chen *et al.* 2011, 2022). The description of *Cel. endophylla* brings the number of species in *Celoporthe* to 15, all of which occur on trees and shrubs in the *Myrtales* (Chen *et al.* 2022). Notably, *Cel. endophylla* described here, and one other species *Cel. foliorum,* are the only foliicolous species in the genus and both occur in Indonesia.

Aureofoliicola represents the 29th genus recognised in *Cryphonectriaceae*. It is clearly morphologically distinct from other genera, producing only a single conidial type and septate, filiform conidiophores, rather than the dimorphic conidia and reduced conidiophores characteristic of the sister genus, *Pseudocryphonectria*. Unlike the majority of *Cryphonectriaceae*, which are typically bark-inhabiting fungi associated with stem cankers, *Aureofoliicola* was isolated from *Eucalyptus* leaves.

Both *Cel. endophylla* and *A. eucalypti* were described here from single isolates. While descriptions based on limited material require cautious interpretation, this is a common situation in fungal systematics, especially for endophytes and other rare fungi that occur sporadically or at low abundance in host tissues. In such cases, the integration of multi-locus phylogenetic data with morphological evidence from culture provides sufficient resolution to justify the recognition of novel taxa. The distinct phylogenetic positions of both isolates, coupled with consistent diagnostic features, support their formal description despite the restricted sample size. Moreover, deposition of the ex-type cultures in international collections ensures accessibility for further study, enabling future work to verify and refine their taxonomic placement as additional isolates are discovered.

Pathogenicity trials conducted on young *Eucalyptus* seedlings revealed a marked difference between *Cel. endophylla and A. eucalypti*. Only the former species showed evidence of pathogenicity on stems, unlike the tissues from which it was isolated. Whether it is a pathogen under natural conditions was not determined. Yet it remains interesting that it might have the capacity to cause disease, as is known for many other apparently endophytic fungi (Tanney *et al.* 2025).

The international movement of infected *Eucalyptus* planting material is rising (Wingfield *et al.* 2001, 2008, 2015, Burgess & Wingfield 2017). Many fungi, including members of the *Cryphonectriaceae*, are now known to persist asymptomatically as endophytes, allowing them to evade detection during routine phytosanitary inspections (Kemler *et al.* 2013, Tanney *et al.* 2025, Wingfield *et al.* 2025). Historical introductions of *Cryphonectriaceae* species into new environments illustrate how such hidden infections can give rise to serious canker diseases on *Eucalyptus* utilised in plantation forestry (Burgess & Wingfield 2017, Wingfield *et al.* 2025). In this context, the isolation of viable cultures was particularly valuable, as it made a pathogenicity test possible and thus offered some evidence of whether these fungi represent a potential threat to the commercial forestry industry.

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REFERENCES

Ali DB, Marincowitz S, Wingfield MJ, *et al.* (2018). Novel *Cryphonectriaceae* from La Réunion and South Africa, and their pathogenicity on *Eucalyptus*. *Mycological Progress* **17**: 953–966. https://doi.org/10.1007/s11557-018-1408-3

Anagnostakis SL (1987). Chestnut blight: the classical problem of an introduced pathogen. *Mycologia* **79**: 23–37. https://doi.org/10.1080/00275514.1987.12025367

Begoude ADB, Gryzenhout M, Wingfield MJ, *et al.* (2010). *Aurifilum*, a new fungal genus in the *Cryphonectriaceae* from *Terminalia* species in Cameroon. *Antonie van Leeuwenhoek* **98**: 263–278. https://doi.org/10.1007/s10482-010-9467-8

Beier GL, Hokanson SC, Bates ST, *et al.* (2015). *Aurantioporthe corni gen. et comb. nov*., an endophyte and pathogen of *Cornus alternifolia*. *Mycologia* **107**: 66–79. https://doi.org/10.3852/14-004

Bissegger M, Sieber TN (1994). Assemblages of endophytic fungi in coppice shoots of *Castanea sativa*. *Mycologia* **86**: 648–655. https://doi.org/10.1080/00275514.1994.12026464

Brown MB, Forsythe AB (1974). Robust tests for the equality of variances. *Journal of the American Statistical Association* **69**: 364–367. https://doi.org/10.2307/2285659

Bruner SC (1917). *Una enfermedad gangrenosa de los eucaliptos*. Estación Central Agronómica de Cuba, Santiago de las Vegas, Cuba.

Burgess TI, Wingfield MJ (2017). Pathogens on the move: a 100-year global experiment with planted eucalypts. *Bioscience* **67**: 14–25. https://doi.org/10.1093/biosci/biw146

Carbone I, Kohn LM (1999). A method for designing primer sets for speciation studies in filamentous ascomycetes. *Mycologia* **91**: 553–556. https://doi.org/10.1080/00275514.1999.12061051

Chen Q, Bakhshi M, Balci Y, *et al.* (2022). Genera of phytopathogenic fungi: GOPHY 4. *Studies in Mycology* **101**: 417–564. https://doi.org/10.3114/sim.2022.101.06

Chen SF, Gryzenhout M, Roux J, *et al.* (2011). Novel species of *Celoporthe* from *Eucalyptus* and *Syzygium* trees in China and Indonesia. *Mycologia* **103**: 1384–1410. https://doi.org/10.3852/11-006

Chen SF, Liu QL, Li GQ, *et al.* (2018). A new genus of *Cryphonectriaceae* isolated from *Lagerstroemia speciosa* in southern China. *Plant Pathology* **67**: 107–123. https://doi.org/10.1111/ppa.12723

Chen SF, Wingfield MJ, Li GQ, *et al.* (2016). *Corticimorbus sinomyrti gen. et sp. nov.* (*Cryphonectriaceae*) pathogenic to native *Rhodomyrtus tomentosa* (*Myrtaceae*) in South China. *Plant Pathology* **65**: 1254–1266. https://doi.org/10.1111/ppa.12507

Chen SF, Wingfield MJ, Roets F, *et al.* (2013a). A serious canker disease caused by *Immersiporthe knoxdaviesiana gen. et sp. nov*. (*Cryphonectriaceae*) on native *Rapanea melanophloeos* in South Africa. *Plant Pathology* **62**: 667–678. https://doi.org/10.1111/j.1365-3059.2012.02671.x

Chen SF, Wingfield MJ, Roux J (2013b). *Diversimorbus metrosiderotis gen. et sp. nov*. and three new species of *Holocryphia* (*Cryphonectriaceae*) associated with cankers on native *Metrosideros angustifolia* trees in South Africa. *Fungal Biology* **117**: 289–310. https://doi.org/10.1016/j.funbio.2013.02.005

Chungu D, Gryzenhout M, Muimba-Kankolongo A, *et al.* (2010). Taxonomy and pathogenicity of two novel *Chrysoporthe* species from *Eucalyptus grandis* and *Syzygium guineense* in Zambia. *Mycological Progress* **9**: 379–393. https://doi.org/10.1007/s11557-009-0646-9

Coelho V, Nunes L, Gouveia E (2021). Short and long term efficacy and prevalence of *Cryphonectria parasitica* hypovirulent strains released as biocontrol agents of chestnut blight. *European Journal of Plant Pathology* **159**: 769–781. https://doi.org/10.1007/s10658-021-02200-3

Cornejo C, Hauser A, Beenken L, *et al.* (2021). *Cryphonectria carpinicola sp. nov*. associated with hornbeam decline in Europe. *Fungal Biology* **125**: 347–356. https://doi.org/10.1016/j.funbio.2020.11.012

Crane C, Burgess TI (2013). *Luteocirrhus shearii gen. sp. nov*. (*Diaporthales*, *Cryphonectriaceae*) pathogenic to *Proteaceae* in the South Western Australian Floristic Region. *IMA Fungus* **4**: 111–122. https://doi.org/10.5598/imafungus.2013.04.01.11

Crous PW, Osieck ER, Jurjević Ž, *et al.* (2021). Fungal Planet description sheets: 1284–1382. *Persoonia* **47**: 178–374. https://doi.org/10.3767/persoonia.2021.47.06

Crous PW, Summerell BA, Alfenas AC, *et al.* (2012). Genera of diaporthalean coelomycetes associated with leaf spots of tree hosts. *Persoonia* **28**: 66–75. https://doi.org/10.3767/003158512X642030

Dunn OJ (1964). Multiple comparisons using rank sums. *Technometrics* **6**: 241–252. https://doi.org/10.1080/00401706.1964.10490181

Ferreira MA, de Oliveira MES, Silva GA, *et al.* (2019). *Capillaureum caryovora gen. sp. nov.* (*Cryphonectriaceae*) pathogenic to pequi (*Caryocar brasiliense*) in Brazil. *Mycological Progress* **18**: 385–403. https://doi.org/10.1007/s11557-018-01461-3

Glass NL, Donaldson GC (1995). Development of primer sets designed for use with the PCR to amplify conserved genes from filamentous ascomycetes. *Applied and Environmental Microbiology* **61**: 1323–1330. https://doi.org/10.1128/aem.61.4.1323-1330.1995

Granados GM, McTaggart AR, Rodas CA, *et al.* (2020). Species of *Cryphonectriaceae* occupy an endophytic niche in the *Melastomataceae* and are putative latent pathogens of *Eucalyptus*. *European Journal of Plant Pathology* **156**: 273–283. https://doi.org/10.1007/s10658-019-01887-9

Gryzenhout M, Glen HF, Wingfield BD, *et al.* (2005a). *Amphilogia gen. nov*. for Cryphonectria-like fungi from *Elaeocarpus* spp. in New Zealand and Sri Lanka. *Taxon* **54**: 1009–1021. https://doi.org/10.2307/25065485

Gryzenhout M, Myburg H, Hodges CS, *et al.* (2006a). *Microthia*, *Holocryphia* and *Ursicollum*, three new genera on *Eucalyptus* and *Coccoloba* for fungi previously known as *Cryphonectria*. *Studies in Mycology* **55**: 35–52. https://doi.org/10.3114/sim.55.1.35

Gryzenhout M, Myburg H, Rodas CA, *et al.* (2006b). *Aurapex penicillata gen. sp. nov*. from native *Miconia theaezans* and *Tibouchina* spp. in Colombia. *Mycologia* **98**: 105–115. https://doi.org/10.3852/mycologia.98.1.105

Gryzenhout M, Myburg H, van der Merwe NA, *et al.* (2004). *Chrysoporthe*, a new genus to accommodate *Cryphonectria cubensis*. *Studies in Mycology* **50**: 119–142.

Gryzenhout M, Myburg H, Wingfield BD, *et al.* (2005b). *Chrysoporthe doradensis sp. nov*. pathogenic to *Eucalyptus* in Ecuador. *Fungal Diversity* **20**: 39–57.

Gryzenhout M, Myburg H, Wingfield BD, *et al.* (2005c). *Rostraureum tropicale gen. sp. nov*. (*Diaporthales*) associated with dying *Terminalia ivorensis* in Ecuador. *Mycological Research* **109**: 1029–1044. https://doi.org/10.1017/S0953756205003291

Gryzenhout M, Myburg H, Wingfield BD, *et al.* (2006c). *Cryphonectriaceae* (*Diaporthales*), a new family including *Cryphonectria*, *Chrysoporthe*, *Endothia* and allied genera. *Mycologia* **98** : 239–249. https://doi.org/10.1080/15572536.2006.11832696

Gryzenhout M, Rodas CA, Mena Portales J, *et al.* (2006d). Novel hosts of the *Eucalyptus* canker pathogen *Chrysoporthe cubensis* and a new *Chrysoporthe* species from Colombia. *Mycological Research* **110**: 833–845. https://doi.org/10.1016/j.mycres.2006.02.010

Gryzenhout M, Tarigan M, Clegg PA, *et al.* (2010). *Cryptometrion aestuescens gen. sp. nov.* (*Cryphonectriaceae*) pathogenic to *Eucalyptus* in Indonesia. *Australasian Plant Pathology* **39**: 161–169. https://doi.org/10.1071/AP09077

Gryzenhout M, Wingfield BD, Wingfield MJ (2009). *Taxonomy, phylogeny, and ecology of bark-inhabiting and tree-pathogenic fungi in the Cryphonectriaceae*. APS Press, St. Paul, Minnesota, USA

Guérin L, Robin C (2003). Seasonal effect on infection and development of lesions caused by *Cryphonectria parasitica* in *Castanea sativa*. *Forest Pathology* **33**: 223–235. https://doi.org/10.1046/j.1439-0329.2003.00329.x

Guindon S, Dufayard JF, Lefort V, *et al.* (2010). New algorithms and methods to estimate maximum-likelihood phylogenies: assessing the performance of PhyML 3.0. *Systematic Biology* **59**: 307–321. doi:10.1093/sysbio/syq010

Harwood CE, Nambiar EKS (2014). Productivity of acacia and eucalypt plantations in Southeast Asia. 2. Trends and variations. *International Forestry Review* **16**: 249–260. https://doi.org/10.1505/146554814811724766

Healey MA, Warman R, Mohammed C, *et al.* (2022). Acacia and eucalypt plantation biosecurity in Southeast Asia – a history, and directions for future research and engagement. *Australian Forestry* **85**: 146–160. https://doi.org/10.1080/00049158.2022.2145721

Hepting GH (1974). Death of the American chestnut. *Journal of Forest History* **18**: 60–67.

Hoang DT, Chernomor O, von Haeseler A, *et al.* (2018) UFBoot2: Improving the Ultrafast Bootstrap Approximation. *Molecular Biology and Evolution* **35**: 518–522. https://doi.org/10.1093/molbev/msx281

Huang HY, Huang HH, Zhao DY, *et al.* (2022). *Pseudocryphonectria elaeocarpicola gen. et sp. nov*. (*Cryphonectriaceae*, *Diaporthales*) causing stem blight of *Elaeocarpus* spp. in China. *MycoKeys* **91**: 67-84. https://doi.org/10.3897/mycokeys.91.86693

Hurley BP, Garnas J, Wingfield MJ, *et al.* (2016). Increasing numbers and intercontinental spread of invasive insects on eucalypts. *Biological Invasions* **18**: 921–933. https://doi.org/10.1007/s10530-016-1081-x

Hyde KD, Dong Y, Phookamsak R, *et al.* (2020). Fungal diversity notes 1151–1276: taxonomic and phylogenetic contributions on genera and species of fungal taxa. *Fungal Diversity* **100**: 5-277. https://doi.org/10.1007/s13225-020-00439-5

Jiang N, Fan XL, Tian CM (2019). Identification and pathogenicity of *Cryphonectriaceae* species associated with chestnut canker in China. *Plant Pathology* **68**: 1132–1145. https://doi.org/10.1111/ppa.13033

Jiang N, Fan XL, Tian CM, *et al.* (2020). Reevaluating *Cryphonectriaceae* and allied families in *Diaporthales*. *Mycologia* **112**: 267–292. https://doi.org/10.1080/00275514.2019.1698925

Jiang N, Fan XL, Yang Q, *et al.* (2018). Two novel species of *Cryphonectria* from *Quercus* in China. *Phytotaxa* **347**: 243–250. https://doi.org/10.11646/phytotaxa.347.3.5

Katoh K, Standley DM (2013). MAFFT multiple sequence alignment software version 7: improvements in performance and usability. *Molecular Biology and Evolution* **30**: 772–780. doi: 10.1093/molbev/mst010.

Kalyaanamoorthy S, Minh BQ, Wong TKF, *et al.* (2017). ModelFinder: fast model selection for accurate phylogenetic estimates. *Nature Methods* **14**: 587–589. https://doi.org/10.1038/nmeth.4285

Kemler M, Garnas J, Wingfield MJ, *et al.* (2013). Ion Torrent PGM as tool for fungal community analysis: a case study of endophytes in *Eucalyptus grandis* reveals high taxonomic diversity. *PLoS ONE* **8**: e81718. https://doi.org/10.1371/journal.pone.0081718

Koukol O, Hrabětová M, Srba M, *et al.* (2018). *Microthia nepenthis*, a new combination for *Zythia nepenthis*. *Czech Mycology* **70**: 91–98. https://doi.org/10.33585/CMY.70107

Liu YJ, Whelen S, Hall BD (1999). Phylogenetic relationships among ascomycetes: evidence from an RNA polymerse II subunit. *Molecular Biology and Evolution* **16**: 1799–1808. https://doi.org/10.1093/oxfordjournals.molbev.a026092

Mausse-Sitoe SND, Rodas CA, Wingfield MJ, *et al.* (2016). Endophytic *Cryphonectriaceae* on native *Myrtales*: Possible origin of Chrysoporthe canker on plantation-grown *Eucalyptus*. *Fungal Biology* **120**: 827–835. https://doi.org/10.1016/j.funbio.2016.03.005

Motulsky HJ, Brown RE (2006). Detecting outliers when fitting data with nonlinear regression – a new method based on robust nonlinear regression and the false discovery rate. *BMC Bioinformatics* **7**: 123. https://doi.org/10.1186/1471-2105-7-123

Myburg H, Gryzenhout M, Heath R, *et al.* (2002). Cryphonectria canker on *Tibouchina* in South Africa. *Mycological Research* **106**: 1299–1306. https://doi.org/10.1017/S095375620200669X

Myburg H, Gryzenhout M, Wingfield BD, *et al.* (2004). Phylogenetic relationships of *Cryphonectria* and *Endothia* species, based on DNA sequence data and morphology. *Mycologia* **96**: 990–1001. https://doi.org/10.1080/15572536.2005.11832899

Myburg H, Wingfield BD, Wingfield MJ, *et al.* (2003). Conspecificity of *Endothia eugeniae* and *Cryphonectria cubensis*: a re-evaluation based on morphology and DNA sequence data. *Mycoscience* **44**: 187–196. https://doi.org/10.1007/S10267-003-0101-8

Nakabonge G, Gryzenhout M, Roux J, *et al.* (2006). *Celoporthe dispersa gen. et sp. nov*. from native *Myrtales* in South Africa. *Studies in Mycology* **55**: 255–267. https://doi.org/10.3114/sim.55.1.255

Pham NQ, Marincowitz S, Solís M *et al.* (2001) *Eucalyptus* scab and shoot malformation: A new and serious foliar disease of Eucalyptus caused by *Elsinoe necatrix sp. nov. Plant Pathology* **70**: 1230–1242. https://doi.org/10.1111/ppa.13348

Rauf MRBA, McTaggart AR, Marincowitz S, *et al.* (2020). Pathogenicity of *Chrysoporthe deuterocubensis* and *Myrtoporthe bodenii* gen. et sp. nov. on *Eucalyptus* in Sabah, Malaysia. *Australasian Plant Pathology* **49**: 53–64. https://doi.org/10.1007/s13313-019-00676-6

Rehner SA, Samuels GJ (1994). Taxonomy and phylogeny of *Gliocladium* analysed from nuclear large subunit ribosomal DNA sequences. *Mycological Research* **98**: 625–634. https://doi.org/10.1016/S0953-7562(09)80409-7

Rigling D, Prospero S (2018). *Cryphonectria parasitica*, the causal agent of chestnut blight: invasion history, population biology and disease control. *Molecular Plant Pathology* 19: 7–20. https://doi.org/10.1111/mpp.12542

Rodas CA, Gryzenhout M, Myburg H, *et al.* (2005). Discovery of the *Eucalyptus* canker pathogen *Chrysoporthe cubensis* on native *Miconia* (*Melastomataceae*) in Colombia. *Plant Pathology* **54**: 460–470. https://doi.org/10.1111/j.1365-3059.2005.01223.x

Roux J, Nkuekam GK, Marincowitz S, *et al.* (2020). *Cryphonectriaceae* associated with rust-infected *Syzygium jambos* in Hawaii. *MycoKeys* **76**: 49–79. https://doi.org/10.3897/mycokeys.76.58406

Shapiro SS, Wilk MB (1965). An analysis of variance test for normality (complete samples). *Biometrika* **52**: 591–611. https://doi.org/10.1093/biomet/52.3-4.591

Silva GA, Oliveira MES, Rêgo GMS, *et al.* (2024). *Chrysoporthe brasiliensis sp. nov*. pathogenic to *Melastomataceae* in southeast Brazil. *Fungal Biology* **128**: 1790–1799. https://doi.org/10.1016/j.funbio.2024.04.001

Sung GH, Sung JM, Hywel-Jones NL, *et al.* (2007). A multi-gene phylogeny of *Clavicipitaceae* (*Ascomycota*, *Fungi*): identification of localized incongruence using a combinational bootstrap approach. *Molecular Phylogenetetics and Evolution* **44**: 1204–1223. https://doi.org/10.1016/j.ympev.2007.03.011

Tang X, Lu YZ, Dissanayake LS, *et al.* (2023). Two new fungal genera (*Diaporthales*) found on *Dipterocarpaceae* in Thailand. *Frontiers in Microbiology* **14**: 1169052. https://doi.org/10.3389/fmicb.2023.1169052

Tanney JB, Kemler M, Vivas M, *et al.* (2025). Silent invaders: the hidden threat of asymptomatic phytobiomes to forest biosecurity. *New Phytologist* **247**: 533–545. https://doi.org/10.1111/nph.70209

Tarigan M, Roux J, van Wyk M, *et al.* (2011). A new wilt and die-back disease of *Acacia mangium* associated with *Ceratocystis manginecans* and *C. acaciivora sp. nov.* in Indonesia. *South African Journal of Botany* **77**: 292–304. https://doi.org/10.1016/j.sajb.2010.08.006

Tarigan M, Wingfield MJ, Jami F, *et al.* (2024). Pathogenicity of *Chrysoporthe deuterocubensis* on eucalypts in Indonesia. *Southern Forests: a Journal of Forest Science* **86**: 42–52. https://doi.org/10.2989/20702620.2023.2279054

Trifinopoulos J, Nguyen LT, von Haeseler A, *et al.* (2016). W-IQ-TREE: a fast online phylogenetic tool for maximum likelihood analysis. *Nucleic Acids Research* **44**: W232–W235. https://doi.org/10.1093/nar/gkw256

van der Merwe NA, Gryzenhout M, Steenkamp ET, *et al.* (2010). Multigene phylogenetic and population differentiation data confirm the existence of a cryptic species within *Chrysoporthe cubensis*. *Fungal Biology* **114**: 966–979. https://doi.org/10.1016/j.funbio.2010.09.007

Venter M, Myburg H, Wingfield BD, *et al.* (2002). A new species of *Cryphonectria* from South Africa and Australia, pathogenic to *Eucalyptus*. *Sydowia* **54**: 98–117.

Vermeulen M, Gryzenhout M, Wingfield MJ, *et al.* (2011). New records of the *Cryphonectriaceae* from southern Africa including *Latruncellus aurorae gen. sp. nov*. *Mycologia* **103**: 554–569. https://doi.org/10.3852/10-283

Vermeulen M, Gryzenhout M, Wingfield MJ, *et al.* (2013). Species delineation in the tree pathogen genus *Celoporthe* (*Cryphonectriaceae*) in southern Africa. *Mycologia* **105**: 297–311. https://doi.org/10.3852/12-206

Vilgalys R, Hester M (1990). Rapid genetic identification and mapping of enzymatically amplified ribosomal DNA from several *Cryptococcus* species. *Journal of Bacteriology* **172**: 4238–4246. https://doi.org/10.1128/jb.172.8.4238-4246.1990

Wang W, Chen SF (2023). Identification and pathogenicity of *Aurifilum* species (*Cryphonectriaceae*, *Diaporthales*) on *Terminalia* species in Southern China. *MycoKeys* **98**: 37–58. https://doi.org/10.3897/mycokeys.98.104719

Wang W, Li GQ, Liu QL, *et al.* (2020). *Cryphonectriaceae* on *Myrtales* in China: phylogeny, host range, and pathogenicity. *Persoonia* **45**: 101–131. https://doi.org/10.3767/persoonia.2020.45.04

Wang W, Liu QL, Li GQ, *et al.* (2018). Phylogeny and pathogenicity of *Celoporthe* species from plantation *Eucalyptus* in southern China. *Plant Disease* **102**: 1915–1927. https://doi.org/10.1094/PDIS-12-17-2002-RE

White TJ, Bruns T, Lee S, *et al*. (1990). Amplification and direct sequencing of fungal ribosomal RNA genes for phylogenetics.

In *PCR protocols: a guide to methods and applications* (Innis MA, Gelfand GD, Sninsky JJ, *et al.* eds), Academic Press, San Diego, CA, USA: 315–322. https://doi.org/10.1016/0307-4412(91)90165-5

Wingfield MJ, Brockerhoff EG, Wingfield BD, *et al.* (2015). Planted forest health: the need for a global strategy. *Science* **349**: 832–836. https://doi.org/10.1126/science.aac6674

Wingfield MJ, Pham NQ, Marincowitz S, *et al.* (2025). *Cryphonectriaceae*: Biodiverse and threatening tree pathogens in the tropics and Southern Hemisphere. *Annual Review of Phytopathology* **63**: 553–575. https://doi.org/10.1146/annurev-phyto-121823-030316

Wingfield MJ, Slippers B, Hurley BP, *et al.* (2008). Eucalypt pests and diseases: growing threats to plantation productivity. *Southern Forests: a Journal of Forest Science* **70**: 139–144. https://doi.org/10.2989/SOUTH.FOR.2008.70.2.9.537

Wingfield MJ, Slippers B, Roux J, *et al.* (2001). Worldwide movement of exotic forest fungi, especially in the tropics and the Southern Hemisphere. *BioScience* **51**: 134–140. https://doi.org/10.1641/0006-3568(2001)051[0134:Wmoeff]2.0.Co;2

Supplementary Material: http://fuse-journal.org/

Table S1. Collection details and GenBank accessions of isolates included in the phylogenetic analyses.