New species, hyper-diversity and potential importance of Calonectria spp. from Eucalyptus in South China

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ABSTRACT: Plantation forestry is expanding rapidly in China to meet an increasing demand for wood and pulp products globally. Fungal pathogens including species of Calonectria represent a serious threat to the growth and sustainability of this industry. Surveys were conducted in the Guangdong, Guangxi and Hainan Provinces of South China, where Eucalyptus trees in plantations or cuttings in nurseries displayed symptoms of leaf blight. Isolations from symptomatic leaves and soils collected close to infected trees resulted in a large collection of Calonectria isolates. These isolates were identified using the Consolidated Species Concept, employing morphological characters and DNA sequence comparisons for the β-tubulin, calmodulin, histone H3 and translation elongation factor 1-alpha gene regions. Twenty-one Calonectria species were identified of which 18 represented novel taxa. Of these, 12 novel taxa belonged to Sphaero-Naviculate Group and the remaining six to the Prolate Group. Southeast Asia appears to represent a centre of biodiversity for the Sphaero-Naviculate Group and this fact could be one of the important constraints to Eucalyptus forestry in China. The remarkable diversity of Calonectria species in a relatively small area of China and associated with a single tree species is surprising.

Key words: Calonectria, Cylindrocladium leaf blight, Eucalyptus, Soil, Taxonomy.

Taxonomic novelties: New species: Calonectria acordiniallis L. Lombard, Crous & S.F. Chen, C. arbusta L. Lombard, Crous & S.F. Chen, C. expansa L. Lombard, Crous & S.F. Chen, C. folicola L. Lombard, Crous & S.F. Chen, C. guangxiensis L. Lombard, Crous & S.F. Chen, C. hainanensis L. Lombard, Crous & S.F. Chen, C. hainae L. Lombard, Crous & S.F. Chen, C. magnispora L. Lombard, Crous & S.F. Chen, C. microconidialis L. Lombard, Crous & S.F. Chen, C. papillata L. Lombard, Crous & S.F. Chen, C. parakyotensis L. Lombard, Crous & S.F. Chen, C. pluramosa L. Lombard, Crous & S.F. Chen, C. pseudokyotensis L. Lombard, Crous & S.F. Chen, C. seminaria L. Lombard, Crous & S.F. Chen, C. sphaeropedunculata L. Lombard, Crous & S.F. Chen, C. terestris L. Lombard, Crous & S.F. Chen, C. tetraramosa L. Lombard, Crous & S.F. Chen, C. turangicola L. Lombard, Crous & S.F. Chen.

Published online 23 January 2015: http://dx.doi.org/10.1016/j.stimyc.2014.11.003. Hard copy: March 2015.

INTRODUCTION

Eucalyptus plantation forestry has grown rapidly during the course of the past two decades in China. This is due to the country being the world’s leading consumer of wood products (Turnbull 2007) and a growing global forest products market. In order to service this market, large-scale plantations of fast-growing trees and especially Eucalyptus spp. have been established in South and Central China. The area spans 19 provinces (Chen et al. 2011a, b, c, d, Zhou & Wingfield 2011) and the aim is to establish 13.3 M ha by 2015 (Turnbull 2007). As is true in other parts of the world, pests and diseases represent a significant challenge to reaching this goal (Zhou et al. 2008, Wingfield et al. 2010, Wingfield et al. 2013).

A recent survey of commercial Eucalyptus plantations and nurseries in the Guangdong, Guangxi, Yunnan and Hainan Provinces resulted in the identification of several important Eucalyptus pathogens. These included leaf pathogens belonging to the genera Mycosphaerella (Burgess et al. 2007), Quambalaria (Zhou et al. 2007) and Teratosphaeria (Burgess et al. 2006). Stem pathogens found included species of Botryosphaeriaceae (Chen et al. 2011a), Celoperthe (Chen et al. 2011b), Ceratocystis (Chen et al. 2013), Chrysosporthe (Chen et al. 2010) and Teratosphaeria (Chen et al. 2011c). In eucalypt nurseries, only isolates belonging to the genus Calonectria (as Cylindrocladium) were found and these were shown (Lombard et al. 2010d) to represent two novel taxa, C. cericiana and C. pseudoreteaudii, and the well-known Eucalyptus nursery pathogen, C. pauciramosa (Kolke et al. 1999, Poluzzi & Crous 1999, Schoch et al. 1999, Crous 2002, Lombard et al. 2010a, d). A more recent survey of Eucalyptus leaves showing symptoms of Calonectria Leaf Blight (CLB) in the Fujian Province resulted in the identification of three novel taxa, C. crousiana, C. fujianensis and C. pseudocolhouiini, and the first record of C. pauciramosa as plantation pathogen (Chen et al. 2011d). Pathogenicity test showed that all four Calonectria species are aggressive pathogens of two important Eucalyptus hybrid clones extensively deployed in plantations (Chen et al. 2011d).

The genus Calonectria accommodates well-known pathogens of various agricultural, horticultural and forestry crops, worldwide (Crous 2002, Lechat et al. 2010, Lombard et al. 2010a, b, c, 2011). Diseases associated with these fungi include cutting and root rot, stem cankers as well as leaf and shoot blight (Crous 2002, Lombard et al. 2010a, b, 2011). In Asia, several Calonectria species have been reported on Eucalyptus trees grown in plantations with most species associated with CLB (Sharma et al. 1984, Booth et al. 2000, Kang et al. 2001a, Crous 2002, Old et al. 2003, Crous et al. 2004b, Chen et al. 2011d). Of these species, members of the C. reteaudii complex (Lombard et al. 2010d) have most frequently been found on Eucalyptus trees, especially in

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Studies in Mycology 80: 151–188
tropical regions of Asia (Booth et al. 2000, Kang et al. 2001a, b, Crous 2002, Old et al. 2003, Lombard et al. 2010d).

Studies by Lombard et al. (2010d) and Chen et al. (2011d) suggested a high level of diversity of Calonectria species associated with Eucalyptus in plantations and nurseries in Southeast China. The aim of this study was to undertake surveys to further assess the limits of diversity of Calonectria in a relatively small area of China associated with Eucalyptus plantations.

MATERIALS AND METHODS

Isolates

An extensive survey for Calonectria species was conducted in Eucalyptus plantations in the Guangdong, Guangxi and Hainan Provinces, China in 2008 and 2009. Where present, leaves of Eucalyptus trees showing symptoms were collected in these plantations. In addition, soil samples were collected associated with the symptomatic trees and these baited with germinating Medicago sativa (alfalfa) seeds using the technique described by Crous (2002). Eucalyptus cuttings showing CLB symptoms were also collected in the nursery of the China Eucalypt Research Centre (CERC) in Guangdong Province.

Plant samples were incubated in moist chambers at room temperature for up to 14 d and inspected daily for fungal structures. Isolations were made directly from these structures onto malt extract agar (2 % w/v; MEA; Biolab, Midrand, South Africa) and incubated for 7 d at 24 °C under continuous near-ultraviolet light. From these primary isolations, single structures were transferred with a sterile loop and grown onto malt extract agar (MEA incubated at 25 °C and the colour charts of Rayner (1970). Isolations were made directly from these structures and incubated for 7 d. Gross morphological characteristics of the asexual structures were determined for all taxonomically informative characters.

DNA sequence comparisons

Total genomic DNA was extracted from 7-d-old cultures established from single-conidial propagules, grown on MEA at room temperature, using the UltraClean™ Microbial DNA isolation kit (Mo Bio Laboratories, Inc., California, USA) following the protocols provided by the manufacturer. Partial gene sequences were determined for β-tubulin (tub2), calmodulin (cmdA), histone H3 (his3), and the translation elongation factor 1-alpha (tef1) regions using the primers and protocols described by Lombard et al. (2010b). To ensure the integrity of the sequences, the amplicons were sequenced in both directions using the same primers used for amplification. Consensus sequences for each locus were assembled in MEGA v. 5.1 (Tamura et al. 2011) and compared with representative sequences from Lombard et al. (2010b) and Alfenas et al. (2015). Subsequent alignments for each locus were generated in MAFFT v. 7.110 (Katoh & Standley 2013) and the ambiguously aligned regions of both ends were truncated.

Phylogenetic analyses were based on both Bayesian inference (BI) and Maximum Parsimony (MP). For BI, the best evolutionary model for each locus was determined using MrModeltest (Nylander 2004) and incorporated into the analyses. MrBayes v. 3.2.1 (Ronquist & Huelsenbeck 2003) was used to generate phylogenetic trees under optimal criteria for each locus. A Markov Chain Monte Carlo (MCMC) algorithm of four chains was initiated in parallel from a random tree topology with the heating parameter set at 0.3. The MCMC analysis lasted until the average standard deviation of split frequencies was below 0.01 with trees saved every 1 000 generations. The first 25 % of saved trees were discarded as the “burn-in” phase and posterior probabilities (PP) were determined from the remaining trees.

For MP, analyses were done using PAUP (Phylogenetic Analysis Using Parsimony, v. 4.0b10; Swofford 2003) with phylogenetic relationships estimated by heuristic searches with 1 000 random addition sequences. Tree-bisection-reconnection was used, with branch swapping option set on “best trees” only. All characters were weighted equally and alignment gaps treated as fifth state. Measures calculated for parsimony included tree length (TL), consistency index (CI), retention index (RI) and rescaled consistency index (RC). Bootstrap analyses (Hillis & Bull 1993) were based on 1 000 replications.

Phylogenetic analyses were conducted on two separate sequence datasets. Datasets were separated based on morphological characteristics into the Prolate Group and Sphaero-Naviculate Group as defined by Lombard et al. (2010b), making it possible to reduce the number of ambiguously aligned regions for the loci analysed. The dataset representing the Prolate Group of species was rooted to C. hongkongensis (CBS 114711 & CBS 114828) and the dataset representing the Sphaero-Naviculate Group was rooted to C. pauciramosa (CMW 5683 & CMW 30823).

Taxonomy

Axenic cultures were sub-cultured onto synthetic nutrient-poor agar (SNA; Nirenburg 1981) and incubated at room temperature for 7 d. Gross morphological characteristics of the asexual morphs were studied by mounting the structures in 85 % lactic acid and 30 measurements were made at ×1 000 magnification for all taxonomically informative characters.

Axenic cultures of Calonectria species of unknown identity and identified based on DNA sequence analyses were crossed among themselves in all possible combinations. Crosses were made on minimal salt agar (MN) with sterile toothpicks placed on the agar surface as described by Lombard et al. (2010b, d). Isolates were crossed with themselves as controls, thus making it possible to distinguish between heterothallic and homothallic mating systems of the isolates. The plates were stacked in plastic containers and incubated at 20 °C for 6–8 wk. Crosses were regarded as successful when isolate combinations produced ascomata extruding viable ascospores.

Morphological characteristics of the sexual morphs were studied by mounting ascomata in tissue freezing medium (Leica Biosystems, Nussloch, Germany) and cutting sections with a Leica CM1100 cryostate (Leica Biosystems, Nussloch, Germany). The 10 μm sections were mounted in 85 % lactic acid and 3 % KOH. The 95 % confidence levels were calculated for the conidia and ascospores with extremes provided in parentheses. For all other fungal structures measured, only the extremes are provided. Colony colour was assessed using 7-d-old cultures on MEA incubated at 25 °C and the colour charts of Rayner (1970).
| Species | Isolate nr. | Substrate | Locality | GenBank Accession no. |
|---------|-------------|-----------|----------|----------------------|
| Calonectria aconidialis | CBS 136088; CMW 35174; CERC 1860 | Soil in Eucalyptus plantation | Hainan, China | – KJ463017 KJ463133 KJ462785 |
| | CBS 136091; CMW 35384; CERC1886 | Soil in Eucalyptus plantation | Hainan, China | – – KJ463134 KJ462786 |
| C. arbusa | CBS 136079; CMW 31370; CERC1705 | Soil in Eucalyptus plantation | Guangxi, China | KJ462904 KJ463018 KJ463135 KJ462787 |
| | CBS 136098; CPC 23519; CMW07981; CERC 1944 | Soil in Eucalyptus plantation | Guangxi, China | – KJ463019 KJ463136 KJ462788 |
| | CPC 23481, CMW 31369; CERC1704 | Soil in Eucalyptus plantation | Guangxi, China | KJ462905 KJ463020 KJ463137 KJ462789 |
| | CMW 31367; CERC 1702 | Soil in Eucalyptus plantation | Guangxi, China | KJ462907 KJ463022 KJ463139 KJ462791 |
| C. asiatica | CBS 134855 | Soil | Teresina, Piauí, Brazil | KM395969 KM396056 KM396139 KM395882 |
| | CBS 134856 | Soil | Teresina, Piauí, Brazil | KM395970 KM396057 KM396140 KM395883 |
| C. canadania | CBS 110817; CPC 499 | Leaf litter | Thailand | AY725613 AY725738 AY725655 AY725702 |
| C. candelabra | CPC 1675; CMW 31000 | Eucalyptus sp. | Brazil | FJ972426 GQ267367 FJ972476 FJ972525 |
| | CMW 31001 | Eucalyptus sp. | Brazil | FJ972427 GQ267368 GQ267246 GQ267246 |
| C. cerciana | CBS 123693, CMW 25309 | Eucalyptus cutting | Zhanjiang, China | FJ918510 GQ267369 FJ918528 FJ918559 |
| | CBS 123695; CPC 1944 | Eucalyptus leaf | Zhanjiang, China | FJ918511 GQ267370 FJ918529 FJ918560 |
| C. chinensis | CBS 112744; CPC 4104 | Soil | Hong Kong, China | AY725618 AY725746 AY725661 AY725710 |
| | CBS 114827; CPC 4101 | Soil | Hong Kong, China | AY725619 AY725747 AY725662 AY725710 |
| C. colhounii | CBS 112711; CPC 3898 | Leaf litter | Thailand | AY725613 AY725738 AY725655 AY725702 |
| | CBS 114073; CPC 3900 | Leaf litter | Thailand | AY725616 AY725741 AY725658 AY725705 |
| C. brasiliensis | CBS 220.51; CPC 2390 | Anacardium sp. | Brazil | GQ267241 GQ267421 GQ267259 GQ267262 |
| | CBS 114257; CPC 1944 | Eucalyptus leaf | Brazil | GQ267242 GQ267422 GQ267246 GQ267246 |
| C. brassiana | CBS 112220; CPC 723 | Soil | La Selva, Brazil | GQ267207 AY725748 AY725662 AY725711 |
| | CBS 112221; CPC 724 | Soil | La Selva, Brazil | AY725620 AY725749 AY725663 AY725712 |
| C. crousiana | CBS 127198; CMW 27249 | E. grandis | Fujian, China | HQ285794 – HQ285808 HQ285822 |
| | CBS 127199; CMW 27253 | E. grandis | Fujian, China | HQ285795 – HQ285809 HQ285823 |
| C. curvispora | CBS 116159; CPC 765 | Soil | Tamatave, Madagascar | AF333394 GQ267374 AY725664 AY725702 |
| C. cylindrospora | CBS 110668; CPC 496 | Soil | USA | FJ918509 GQ267423 FJ918527 FJ918557 |
| | CBS 119670; CPC 12766 | Pistacia lentiscus | Italy | DO251600 – DO251602 DO421797 |
| C. eucalypticola | CBS 134846 | Eucalyptus leaf | Eunápolis, Bahia, Brazil | KM395963 KM396050 KM396133 KM396876 |
| | CBS 134847 | Eucalyptus seedling | Santa Bárbara, Minas Gerais, Brazil | KM395964 KM396051 KM396134 KM396877 |
| C. expansa | CBS 136078; CMW 31441; CERC 1776 | Soil in Eucalyptus plantation | Guangdong, China | KJ462913 KJ463028 KJ463145 KJ462797 |
| Species           | Isolate nr. | Substrate                        | Locality                        | GenBank Accession no. |
|-------------------|-------------|----------------------------------|----------------------------------|-----------------------|
| **CBS 136247; CMW 31392; CERC 1727** | Soil in Eucalyptus plantation | Guangxi, China                  | KJ462914 KJ463029 KJ463146 KJ462798 |
| C. folicola       |             | Soil in Eucalyptus plantation    | Guangxi, China                  | KJ462915 KJ463030 KJ463147 KJ462799 |
| **CBS 136641; CMW 31393; CERC 1728** | E. urophylla × E. grandis clone leaf | Guangxi, China                  | KJ462916 KJ463031 KJ463148 KJ462800 |
| CMW 31394; CERC 1729 | E. urophylla × E. grandis clone leaf | Guangxi, China                  | KJ462917 KJ463032 KJ463149 KJ462801 |
| CMW 31395; CERC 1730 | E. urophylla × E. grandis clone leaf | Guangxi, China                  | KJ462918 KJ463033 KJ463150 KJ462802 |
| C. fujianensis    |             | Soil in Eucalyptus plantation    | Guangxi, China                  | KJ462919 KJ463034 KJ463151 KJ462803 |
| **CBS 136248**    | Soil in Eucalyptus plantation | Guangxi, China                  | HQ285791 – HQ285805 HQ285819 |
| **CBS 13852**     | Soil        | Guangxi, China                  | HQ285792 – HQ285806 HQ285820 |
| **CBS 136092; CMW 35409; CERC 1900** | Soil in Eucalyptus plantation | Guangxi, China                  | KJ462919 KJ463034 KJ463151 KJ462803 |
| **CBS 136094; CMW 35411; CERC 1902** | Soil in Eucalyptus plantation | Guangxi, China                  | KJ462920 KJ463035 – KJ462804 |
| C. glaeboicola    |             | Soil in Eucalyptus plantation    | Guangxi, China                  | KJ462919 KJ463034 KJ463151 KJ462803 |
| **CBS 134852**    | Soil        | Guangxi, China                  | KM395966 KM396053 KM396136 KM395879 |
| C. guangxiensis   |             | Soil in Eucalyptus plantation    | Guangxi, China                  | KJ462919 KJ463034 KJ463151 KJ462803 |
| **CBS 136248; CMW 35187; CERC 1863** | Soil in Eucalyptus plantation | Guangxi, China                  | KJ462919 KJ463034 KJ463151 KJ462803 |
| C. hainanensis    |             | Soil in Eucalyptus plantation    | Guangxi, China                  | KJ462919 KJ463034 KJ463151 KJ462803 |
| C. hawksworthii   | **CBS 111870; CPC 2405; MUCL 30866** | Soil in Eucalyptus plantation | KJ462919 KJ463034 KJ463151 KJ462803 |
| C. hodgesii       | **CBS 133609; LPF 245** | E. urophylla × E. grandis clone leaf | Guangxi, China                  | KJ462919 KJ463034 KJ463151 KJ462803 |
| **CBS 133610; LPF 261** | E. grandis | Guangxi, China                  | KJ462919 KJ463034 KJ463151 KJ462803 |
| C. hongkongensis  | **CBS 114711; CPC 467** | Soil in Eucalyptus plantation | KJ462919 KJ463034 KJ463151 KJ462803 |
| **CBS 114828; CPC 4670** | Soil in Eucalyptus plantation | Guangxi, China                  | KJ462919 KJ463034 KJ463151 KJ462803 |
| **CBS 136080; CMW 31443; CERC 1778** | Soil in Eucalyptus plantation | Guangxi, China                  | KJ462919 KJ463034 KJ463151 KJ462803 |
| **CBS 136246; CMW 31374; CERC 1709** | Soil in Eucalyptus plantation | Guangxi, China                  | KJ462919 KJ463034 KJ463151 KJ462803 |
| CPC 23478; CMW 31438; CERC 1773 | Soil in Eucalyptus plantation | Guangxi, China                  | KJ462919 KJ463034 KJ463151 KJ462803 |
| CPC 23480; CMW 31414; CERC 1749 | Soil in Eucalyptus plantation | Guangxi, China                  | KJ462919 KJ463034 KJ463151 KJ462803 |
| CPC 23499; CMW 35175; CERC 1851 | Soil in Eucalyptus plantation | Guangxi, China                  | KJ462919 KJ463034 KJ463151 KJ462803 |
| CPC 23877; CERC 1932 | Soil in Eucalyptus plantation | Guangxi, China                  | KJ462919 KJ463034 KJ463151 KJ462803 |
| CPC 23878; CMW 37973; CERC 1936 | Soil in Eucalyptus plantation | Guangxi, China                  | KJ462919 KJ463034 KJ463151 KJ462803 |
| **CMW 31375; CERC 1710** | Soil in Eucalyptus plantation | Guangxi, China                  | KJ462919 KJ463034 KJ463151 KJ462803 |
| **CMW 31377; CERC 1712** | Soil in Eucalyptus plantation | Guangxi, China                  | KJ462919 KJ463034 KJ463151 KJ462803 |
| **CMW 31382; CERC 1717** | Soil in Eucalyptus plantation | Guangxi, China                  | KJ462919 KJ463034 KJ463151 KJ462803 |
| **CMW 31383; CERC 1718** | Soil in Eucalyptus plantation | Guangxi, China                  | KJ462919 KJ463034 KJ463151 KJ462803 |
| **CMW 31384; CERC 1719** | Soil in Eucalyptus plantation | Guangxi, China                  | KJ462919 KJ463034 KJ463151 KJ462803 |
| **CMW 31385; CERC 1720** | Soil in Eucalyptus plantation | Guangxi, China                  | KJ462919 KJ463034 KJ463151 KJ462803 |
| **CMW 31387; CERC 1722** | Soil in Eucalyptus plantation | Guangxi, China                  | KJ462919 KJ463034 KJ463151 KJ462803 |
| Species               | Isolate nr.¹ | Substrate                              | Locality                                      | GenBank Accession no.² |
|----------------------|--------------|----------------------------------------|------------------------------------------------|------------------------|
|                       |              | tub2 | cmdA                         | his3 | tef1 |
| CMW 31388; CERC 1723 | Soil in Eucalyptus plantation | Fangchenggang, Guangxi, China            | KJ462935 | KJ463051 | KJ463167 | KJ462820 |
| CMW 31399; CERC 1734 | Soil in Eucalyptus plantation | Shiling, Zhanjiang, Guangdong, China     | KJ462936 | –         | KJ463168 | KJ462821 |
| CMW 31400; CERC 1735 | Soil in Eucalyptus plantation | Shiling, Zhanjiang, Guangdong, China     | KJ462937 | KJ463052 | KJ463169 | KJ462822 |
| CMW 31401; CERC 1736 | Soil in Eucalyptus plantation | Shiling, Zhanjiang, Guangdong, China     | KJ462938 | KJ463053 | KJ463170 | KJ462823 |
| CMW 31404; CERC 1739 | Soil in Eucalyptus plantation | Shiling, Zhanjiang, Guangdong, China     | KJ462939 | KJ463054 | KJ463171 | KJ462824 |
| CMW 31432; CERC 1767 | Soil in Eucalyptus plantation | Shiling, Zhanjiang, Guangdong, China     | KJ462940 | KJ463055 | KJ463172 | KJ462825 |
| CMW 31433; CERC 1768 | Soil in Eucalyptus plantation | Shiling, Zhanjiang, Guangdong, China     | KJ462941 | KJ463056 | KJ463173 | KJ462826 |
| CMW 31434; CERC 1769 | Soil in Eucalyptus plantation | Shiling, Zhanjiang, Guangdong, China     | KJ462942 | KJ463057 | KJ463174 | KJ462827 |
| CMW 31442; CERC 1777 | Soil in Eucalyptus plantation | Fangchenggang, Guangxi, China           | KJ462943 | KJ463058 | KJ463175 | KJ462828 |
| CMW 35186; CERC 1862 | Soil in Eucalyptus plantation | Hainan, China                           | KJ462944 | KJ463059 | KJ463176 | KJ462829 |
| CMW 35188; CERC 1864 | Soil in Eucalyptus plantation | Hainan, China                           | KJ462945 | KJ463060 | KJ463177 | KJ462830 |
| CMW 35189; CERC 1865 | Soil in Eucalyptus plantation | Guangxi, China                          | KJ462946 | KJ463061 | KJ463178 | KJ462831 |
| CMW 35192; CERC 1867 | Soil in Eucalyptus plantation | Guangxi, China                          | KJ462947 | KJ463062 | –         | KJ462832 |
| CMW 35371; CERC 1874 | Soil in Eucalyptus plantation | Guangdong, China                        | KJ462948 | KJ463063 | KJ463179 | KJ462833 |
| CMW 35376; CERC 1880 | Soil in Eucalyptus plantation | Guangdong, China                        | KJ462949 | KJ463064 | KJ463180 | KJ462834 |
| CMW 35381; CERC 1883 | Soil in Eucalyptus plantation | Hainan, China                           | KJ462950 | KJ463065 | KJ463181 | KJ462835 |
| CMW 35401; CERC 1898 | Soil in Eucalyptus plantation | Guangxi, China                          | KJ462951 | KJ463066 | KJ463182 | KJ462836 |
| CMW 35404; CERC 1895 | Soil in Eucalyptus plantation | Guangxi, China                          | KJ462952 | KJ463067 | KJ463183 | KJ462837 |
| CMW 35414; CERC 1905 | Soil in Eucalyptus plantation | Guangxi, China                          | KJ462953 | KJ463068 | KJ463184 | KJ462838 |
| CMW 36270; CERC 1928 | Soil in Eucalyptus plantation | Hainan, China                           | KJ462954 | KJ463069 | KJ463185 | KJ462839 |
| CBS 190.50; CMW 30998; IMI 29938 | Solanum tuberosum | Bogor, Indonesia | AY725631 | AY725764 | AY725676 | AY725726 |
| CBS 115897; CPC 493; UFV 108 | Anacardium sp. | Brazil | AY725647 | GQ267403 | GQ267256 | AY725729 |
| CBS 112283; CPC 4508 | Soil | Warambunga, Indonesia | AY725623 | AY725756 | AY725666 | AY725718 |
| CBS 112840; CPC 4554 | Syzygium aromaticum | Indonesia | AY725625 | AY725758 | AY725670 | AY725720 |
| CBS 114558; CPC 786 | Soil | Tamatave, Madagascar | AF210861 | GQ267389 | FJ918526 | FJ918556 |
| CBS 114559; CPC 954 | Soil | Tamatave, Madagascar | AF210862 | GQ267390 | FJ918525 | FJ918555 |
| CBS 413.67; CPC 2391; IMI 299577 | Paphiopedilum callosum | Celle, Germany | GQ267208 | GQ267379 | GQ267248 | GQ267307 |
| CBS 170.77; IMI 299388 | Ilexis polycarpa | Auckland, New Zealand | GQ267209 | GQ267380 | GQ267249 | GQ267308 |
| CBS 136629; CMW 31412; CERC 1474 | Soil in Eucalyptus plantation | Fangchenggang, Guangxi, China | KJ462955 | KJ463070 | KJ463186 | KJ462840 |
| CBS 109166; CPC 2385; ATCC 64824 | Leucothoe axillaris | Gainesville, Florida, USA | FJ918508 | GQ267392 | FJ918523 | FJ918553 |
| CBS 136249; CMW 35184; CERC 1860 | Soil in Eucalyptus plantation | Guangxi, China | KJ462956 | KJ463071 | KJ463187 | KJ462841 |
| CBS 112710; CPC 3899 | Leaf litter | Thailand | AY725626 | AY725759 | AY725671 | AY725721 |
| CBS 112752; CPC 4223 | Soil | Sumatra, Indonesia | AY725627 | AY725760 | AY725672 | AY725722 |
| CBS 134811 | Eucalyptus sp. | Açaílandia, Maranhão, Brazil | KM395948 | KM396035 | KM396119 | KM396861 |
| CBS 134812 | Eucalyptus sp. | Açaílandia, Maranhão, Brazil | KM395949 | KM396036 | KM396119 | KM396862 |

(continued on next page)
| Species | Isolate nr. | Substrate | Locality | GenBank Accession no. |
|---------|------------|-----------|----------|----------------------|
|         |            |           |          | **tub2** | **cmdA** | **his3** | **tef1** |
| C. metrosideri | CBS 133604; LPF 103 | Metrosidea polymorpha | Viçosa, Brazil | KM395952 | KM396039 | KM396122 | KM395865 |
|          | CBS 133605; LPF 104 | M. polymorpha | Viçosa, Brazil | KM395953 | KM396040 | KM396123 | KM395866 |
| C. microconidialis | CBS 136633, CMW 31471; CERC 1806 | E. urophylla × E. grandis clone seedling leaf | CERC Nursery, Zhanjiang, Guangdong, China | KJ463957 | KJ463072 | KJ463188 | KJ462842 |
|          | CBS 136634, CMW 31472; CERC 1806 | E. urophylla × E. grandis clone seedling leaf | CERC Nursery, Zhanjiang, Guangdong, China | KJ463958 | KJ463073 | KJ463190 | KJ462843 |
|          | CBS 136635, CMW 31473; CERC 1807 | E. urophylla × E. grandis clone seedling leaf | CERC Nursery, Zhanjiang, Guangdong, China | KJ463959 | KJ463074 | KJ463192 | KJ462844 |
|          | CBS 136636, CMW 31474; CERC 1808 | E. urophylla × E. grandis clone seedling leaf | CERC Nursery, Zhanjiang, Guangdong, China | KJ463960 | KJ463075 | KJ463193 | KJ462845 |
|          | CBS 136637, CMW 31475; CERC 1809 | E. urophylla × E. grandis clone seedling leaf | CERC Nursery, Zhanjiang, Guangdong, China | KJ463961 | KJ463076 | KJ463194 | KJ462846 |
| C. nemuricola | CBS 134837 | Soil | Araponga, Minas Gerais, Brazil | KM395979 | KM396086 | KM396149 | KM395892 |
|          | CBS 134838 | Soil | Araponga, Minas Gerais, Brazil | KM395980 | KM396087 | KM396150 | KM395893 |
| C. nymphaeae | CBS 131802; HGUP 100003 | Nymphaea tetragona | Guizhou, China | JN984864 | – | – | JK559273 |
| C. pacifica | CBS 109063, CPC 2534; IMI 354526 | Araucaria heterophylla | Hawaii, USA | QQ267213 | QQ267255 | QQ267254 |
| C. papillata | CBS 136084, CMW 35165; CERC 1841 | Soil in Eucalyptus plantation | Guangdong, China | KJ463062 | KJ463077 | KJ463193 | KJ462847 |
|          | CBS 136096, CMW 37972; CERC 1935 | Soil in Eucalyptus plantation | Guangdong, China | KJ463063 | KJ463078 | KJ463194 | KJ462848 |
|          | CBS 136097, CMW 37976; CERC 1939 | Soil in Eucalyptus plantation | Guangdong, China | KJ463064 | KJ463079 | KJ463195 | KJ462849 |
|          | CBS 136251, CMW 37971; CERC 1934 | Soil in Eucalyptus plantation | Guangxi, China | KJ463065 | KJ463080 | KJ463196 | KJ462850 |
| C. parakyotensis | CBS 136085, CMW 35169; CERC 1845 | Soil in Eucalyptus plantation | Guangdong, China | KJ463081 | KJ463197 | KJ462851 |
|          | CBS 136095, CMW 35413; CERC 1904 | Soil in Eucalyptus plantation | Guangxi, China | KJ463082 | KJ463198 | KJ462852 |
| C. pauciramosa | CMW 5683 | E. grandis | South Africa | FJ185154 | FJ185314 | FJ185565 |
|          | CMW 30823 | E. grandis | South Africa | FJ185155 | FJ280404 | FJ185566 |
| C. pentasepata | CBS 133349 | Eucalyptus hybrid | Bavi, Hanoi, Vietnam | JX855942 | – | JX855946 | JX855968 |
|          | CBS 133351 | Macadamia sp. | Bavi, Hanoi, Vietnam | JX855944 | – | JX855948 | JX855960 |
|          | CBS 136087, CMW 35177; CERC 1853 | Eucalyptus leaf | Hainan, China | KJ462966 | KJ463083 | KJ463199 | KJ462853 |
|          | CBS 136089, CMW 35377; CERC 1879 | Eucalyptus leaf | Hainan, China | KJ462967 | KJ463084 | KJ463200 | KJ462854 |
|          | CBS 136250, CMW 35451; CERC 1923 | Eucalyptus leaf | Guangdong, China | KJ462968 | KJ463085 | KJ463201 | KJ462855 |
|          | CBS 136646, CMW 35436; CERC 1908 | Eucalyptus leaf | Guangdong, China | KJ462969 | KJ463086 | KJ463202 | KJ462856 |
|          | CMW 31332, CERC 1667 | Eucalyptus clone U6 leaf | Shiling, Zhanjiang, Guangdong, China | KJ462970 | KJ463087 | KJ463203 | KJ462857 |
|          | CMW 31333, CERC 1668 | Eucalyptus clone U6 leaf | Shiling, Zhanjiang, Guangdong, China | KJ462971 | KJ463088 | KJ463204 | KJ462858 |
|          | CMW 31336, CERC 1671 | Eucalyptus clone U6 leaf | Shiling, Zhanjiang, Guangdong, China | KJ462972 | KJ463089 | KJ463205 | KJ462859 |
### Table 1. (Continued).

| Species | Isolate nr. | Substrate | Locality | GenBank Accession no. |
|---------|-------------|-----------|----------|-----------------------|
| C. pseudometrosideri | CMW 31340, CERC 1675 | E. urophylla × E. grandis clone seedling leaf | CERC Nursery, Zhanjiang, Guangdong, China | KJ462973 KJ463090 KJ465206 KJ462860 |
| | CMW 31343, CERC 1678 | E. urophylla × E. grandis clone seedling leaf | CERC Nursery, Zhanjiang, Guangdong, China | KJ462973 KJ463091 KJ465207 KJ462861 |
| | CMW 31344, CERC 1679 | E. urophylla × E. grandis clone seedling leaf | CERC Nursery, Zhanjiang, Guangdong, China | KJ462975 KJ463092 KJ465208 KJ462862 |
| | CMW 31345, CERC 1680 | E. urophylla × E. grandis clone seedling leaf | CERC Nursery, Zhanjiang, Guangdong, China | KJ462976 KJ463093 KJ465209 KJ462863 |
| | CMW 31346, CERC 1681 | E. urophylla × E. grandis clone seedling leaf | CERC Nursery, Zhanjiang, Guangdong, China | KJ462977 KJ463094 KJ465210 KJ462864 |
| | CMW 31347, CERC 1682 | E. urophylla × E. grandis clone seedling leaf | CERC Nursery, Zhanjiang, Guangdong, China | KJ462978 KJ463095 KJ465211 KJ462865 |
| | CMW 31348, CERC 1683 | E. urophylla × E. grandis clone seedling leaf | CERC Nursery, Zhanjiang, Guangdong, China | KJ462979 KJ463096 KJ465212 KJ462866 |
| | CMW 31355, CERC 1690 | E. urophylla × E. grandis leaf | Heping, Guangxi, China | KJ462980 KJ463097 KJ465213 KJ462867 |
| | CMW 31356, CERC 1691 | E. urophylla × E. grandis leaf | Heping, Guangxi, China | KJ462981 KJ463098 KJ465214 KJ462868 |
| | CMW 31357, CERC 1692 | E. urophylla × E. grandis leaf | Heping, Guangxi, China | KJ462982 KJ463099 KJ465215 KJ462869 |
| | CMW 31358, CERC 1693 | E. urophylla × E. grandis leaf | Heping, Guangxi, China | KJ462983 KJ463100 KJ465216 KJ462870 |
| | CMW 31359, CERC 1694 | E. urophylla × E. grandis leaf | Heping, Guangxi, China | KJ462984 KJ463101 KJ465217 KJ462871 |
| | CMW 31363, CERC 1698 | E. urophylla × E. grandis leaf | Heping, Guangxi, China | KJ462985 KJ463102 KJ465218 KJ462872 |
| | CMW 31422, CERC 1757 | Eucalyptus clone U6 leaf | Shiling, Zhanjiang, Guangdong, China | KJ462986 KJ463103 KJ465219 KJ462873 |
| | CMW 31497, CERC 1832 | E. urophylla × E. grandis clone seedling leaf | CERC Nursery, Zhanjiang, Guangdong, China | KJ462987 KJ463104 KJ465220 KJ462874 |
| | CMW 35385, CERC 1887 | Soil in Eucalyptus plantation | Hainan, China | KJ462988 KJ463105 KJ465221 KJ462875 |
| | CMW 35437, CERC 1909 | Eucalyptus leaf | Guangdong, China | KJ462989 KJ463106 KJ465222 KJ462876 |
| | CMW 35442, CERC 1914 | Eucalyptus leaf | Guangdong, China | KJ462990 KJ463107 KJ465223 KJ462877 |
| | CMW 35452, CERC 1924 | Eucalyptus leaf | Guangdong, China | KJ462991 KJ463108 KJ465224 KJ462878 |
| | CMW 35453, CERC 1925 | Eucalyptus leaf | Guangdong, China | KJ462992 KJ463109 KJ465225 KJ462879 |
| | CMW 35454, CERC 1926 | Eucalyptus leaf | Guangdong, China | KJ462993 KJ463110 KJ465226 KJ462880 |
| C. pisuiensis | CBS 134850 | Soil | Teresina, Piauí, Brazil | KM395973 KM396060 KM396143 KM395866 |
| | CBS 134851 | Soil | Teresina, Piauí, Brazil | KM395974 KM396061 KM396144 KM395867 |
| C. plurinamosa | CBS 136976; CMW 31440; CERC 1775 | Soil in Eucalyptus plantation | Fangchengegang, Guangxi, China | KJ462995 KJ463112 KJ465228 KJ462882 |
| C. polizzi | CBS 125270, CMW 7804 | Callistemon citrinus | Messina, Sicily, Italy | FJ972417 HQ267461 FJ972436 FJ972486 |
| | CBS 125271, CMW 10151 | Arbutus unedo | Catania, Sicily, Italy | FJ972418 HQ267462 FJ972437 FJ972487 |
| C. propagincola | CBS 134815 | Eucalyptus cutting | Santana, Pará, Brazil | KM395953 KM396040 KM396123 KM395866 |
| | CBS 134820 | Used planting substrate | Santana, Pará, Brazil | KM395956 KM396043 KM396126 KM395869 |
| | CBS 134821 | Used planting substrate | Santana, Pará, Brazil | KM395957 KM396044 KM396127 KM395870 |
| C. pseudocerciana | CBS 134824 | Eucalyptus seedling | Santana, Pará, Brazil | KM395962 KM396049 KM396132 KM395875 |
| C. pseudocolochnii | CBS 127195; CMW 27209 | E. dunnii | Fujian, China | HQ285788 – HQ285802 HQ285816 |
| | CBS 127196; CMW 27213 | E. dunnii | Fujian, China | HQ285789 – HQ285803 HQ285817 |
| C. pseudohodgesii | CBS 134818 | Azadirachta indica | Viçosa, Minas Gerais, Brazil | KM395905 KM395991 KM396079 KM395817 |
| | CBS 134819 | A. indica | Viçosa, Minas Gerais, Brazil | KM395906 KM395992 KM396080 KM395818 |
| C. pseudokytosensis | CBS 137332; CMW 31439; CERC 1774 | Soil in Eucalyptus plantation | Fangchengegang, Guangxi, China | KJ462994 KJ463111 KJ465227 KJ462881 |
| C. pseudometrosideri | CBS 134843 | Soil | Viçosa, Minas Gerais, Brazil | KM395907 KM395993 KM396081 KM395819 |
| CBS 134845 | Soil | Maceió, Alagoas, Brazil | KM395909 KM395995 KM396083 KM395821 |

(continued on next page)
| Species                  | Isolate nr. | Substrate                                    | Locality                      | GenBank Accession no. |
|-------------------------|-------------|----------------------------------------------|-------------------------------|----------------------|
|                         |             | tub2                                         | cmdA                          | his3                 | tefl                |
| *C. pseudoretarda*      | CBS 123694; CMW 25310 | Eucalyptus hybrid cutting                    | Guangdong, China              | FJ918504             | GQ267411            | FJ918519 | FJ918541 |
|                         | CBS 123696; CMW 25292 | Eucalyptus hybrid cutting                    | Guangdong, China              | FJ918505             | GQ267410            | FJ918520 | FJ918542 |
| *C. pseudoscoparia*     | CBS 125256; CMW 15216 | *E. grandis*                                | Pichincha, Ecuador            | GQ267228             | GQ267440            | GQ267277 | GQ267348 |
|                         | CBS 125257; CMW 15218 | *E. grandis*                                | Pichincha, Ecuador            | GQ267229             | GQ267441            | GQ267278 | GQ267349 |
| *C. pseudopathulata*    | CBS 134840   | Soil                                         | Araponga, Minas Gerais, Brazil | KM395982             | KM396069            | KM396152 | KM395895 |
|                         | CBS 134841   | Soil                                         | Araponga, Minas Gerais, Brazil | KM395983             | KM396070            | KM396153 | KM395896 |
| *C. queenslandica*      | CBS 112146; CPC 3213 | *E. urophylla*                              | Australia                     | AF389835             | GQ267415            | FJ918521 | FJ918543 |
|                         | CBS 112155; CPC 3210 | *E. petitta*                                | Australia                     | AF389834             | GQ267416            | DQ190687 | FJ918544 |
| *C. retaeda*            | CBS 112143; CPC 3200 | *E. camaldulensis*                          | Vietnam                       | GQ240642             | GQ267418            | DQ190660 | FJ918542 |
| *C. seminaria*          | CBS 136630; CMW 31448; CERC 1781 | *E. urophylla × E. grandis* clone seedling leaf | CERC Nursery, Zhanjiang, Guangdong, China | KJ462996             | KJ463113            | KJ463229 | KJ462863 |
|                         | CBS 136631; CMW 31449; CERC 1784 | *E. urophylla × E. grandis* clone seedling leaf | CERC Nursery, Zhanjiang, Guangdong, China | KJ462997             | KJ463114            | KJ463230 | KJ462864 |
|                         | CBS 136632; CMW 31450; CERC 1824 | *E. urophylla × E. grandis* clone seedling leaf | CERC Nursery, Zhanjiang, Guangdong, China | KJ462998             | KJ463115            | KJ463315 | KJ462865 |
|                         | CBS 136639; CMW 31489; CERC 1833 | *E. urophylla × E. grandis* clone seedling leaf | CERC Nursery, Zhanjiang, Guangdong, China | KJ462999             | KJ463116            | KJ463320 | KJ462866 |
|                         | CBS 136648; CMW 37970; CERC 1833 | Eucalyptus leaf                            | Guangxi, China                | KJ463000             | KJ463117            | KJ463233 | KJ462867 |
|                         | CPC 23486; CMW 31447; CERC 1792 | *E. urophylla × E. grandis* clone seedling leaf | CERC Nursery, Zhanjiang, Guangdong, China | KJ463001             | KJ463118            | KJ463234 | KJ462868 |
|                         | CPC 23487; CMW 31448; CERC 1793 | *E. urophylla × E. grandis* clone seedling leaf | CERC Nursery, Zhanjiang, Guangdong, China | KJ463002             | KJ463119            | KJ463235 | KJ462869 |
| *C. silvicola*          | CBS 134836   | Soil                                         | Araponga, Minas Gerais, Brazil | KM395975             | KM396062            | KM396145 | KM395888 |
| *C. sphaeropedunculata* | CBS 136681; CMW 31390; CERC 1725 | Soil in Eucalyptus plantation               | Guangxi, China                | KJ463003             | KJ463120            | KJ463236 | KJ462890 |
| *C. sulawesiensis*      | CBS 125248; CMW 14857 | Eucalyptus sp.                              | Sulawesi, Indonesia           | GQ267223             | GQ267435            | GQ267272 | GQ267343 |
|                         | CBS 125253; CMW 14879 | Eucalyptus sp.                              | Sulawesi, Indonesia           | GQ267220             | GQ267432            | GQ267269 | GQ267340 |
| *C. sumatrensis*        | CBS 112829; CPC 4518 | Soil                                         | Sumatra, Indonesia            | AY725649             | AY725751            | AY725696 | AY725733 |
|                         | CBS 112934; CPC 4516 | Soil                                         | Indonesia                     | AY725651             | AY725757            | AY725798 | AY725735 |
| *C. terrae-reginae*     | CBS 112151; CPC 3202 | *E. urophylla*                              | Queensland, Australia         | FJ918506             | GQ267451            | FJ918522 | FJ918545 |
|                         | CBS 112534; CPC 4233 | Xanthorrhoea australis                      | Victoria, Australia           | FJ918507             | GQ267452            | DQ190668 | FJ918546 |
| *C. terrestris*         | CBS 136642; CMW 35180; CERC 1896 | Soil in Eucalyptus plantation               | Guangdong, China              | KJ463004             | KJ463121            | KJ463237 | KJ462891 |
|                         | CBS 136643; CMW 35364; CERC 1868 | Soil in Eucalyptus plantation               | Guangdong, China              | KJ463005             | KJ463122            | KJ463238 | KJ462892 |
|                         | CBS 136644; CMW 35366; CERC 1870 | Soil in Eucalyptus plantation               | Guangdong, China              | KJ463006             | KJ463123            | KJ463239 | KJ462893 |
|                         | CBS 136645; CMW 35178; CERC 1854 | Soil in Eucalyptus plantation               | Guangdong, China              | KJ463007             | KJ463124            | KJ463240 | KJ462894 |
|                         | CBS 136647; CMW 35447; CERC 1919 | Eucalyptus leaf                            | Guangdong, China              | KJ463008             | KJ463125            | KJ463241 | KJ462895 |
|                         | CBS 136651; CMW 37974; CERC 1937 | Soil                                         | Guangdong, China              | KJ463009             | KJ463126            | KJ463242 | KJ462896 |
|                         | CBS 136653; CMW 37980; CERC 1943 | Soil                                         | Guangxi, China                | KJ463010             | KJ463127            | KJ463243 | KJ462897 |
RESULTS

Isolates

A total of 278 isolates were collected of which 162 were from the Guangdong Province (44 isolates from soil; 45 isolates from Eucalyptus leaves on trees; 73 from cuttings in a single nursery). 87 isolates from Guangxi Province (63 from soil; 24 from Eucalyptus leaves in plantations), and 29 isolates from the Hainan Province (27 from soil; two from Eucalyptus leaves in plantations). One hundred and twenty of these isolates were selected for further study (Table 1) based on preliminary phylogenetic analysis of the cmdA and tub2 gene region sequences (results not shown).

DNA sequence comparisons

Approximately 500–550 bases were determined for the four gene regions used in this study. For the Bayesian analyses, a HKY+I+G model was selected for cmdA, tef1 and tub2 and the GTR+I+G model for his3. These models were incorporated for each of the datasets analysed. The Bayesian consensus trees for both datasets confirmed the tree topologies obtained from the MP analyses, and therefore, only the MP trees are presented with bootstrap support values (BS) and posterior probabilities (PP) shown for well-supported nodes.

The dataset for the Prolate Group isolates included 127 ingroup taxa, with C. hongkongensis (CBS 114711 & CBS 114828) as the outgroup taxon. The sequence dataset consisted of 2 018 characters, including alignment gaps. Of these, 1 308 were constant, 73 were parsimony-uninformative and 637 parsimony-informative. The MP analysis yielded 1 000 trees (TL = 1 512; CI = 0.612; RC = 0.538; RI = 0.952) of which the first is presented (Fig. 1). The majority of the isolates included in this dataset clustered in the clade (BS < 50; PP = 1.00) representing C. pentaseptata (ex-type CBS 133439) with five isolates (CBS 136633, CBS 136634, CBS 136636, CBS 136638 & CBS 136640) forming a sister clade (BS < 50; PP = 0.96) to the C. pentaseptata clade. A clade (BS = 75; PP = 0.99) incorporating seven isolates (CBS 136630, CBS 136631, CBS 136632, CBS 136639, CBS 136640, CPC 23486 & CPC 23487), with an additional two isolates (CBS 136635 & CBS 136637) forming a sister clade (BS = 81; PP = 1.00), clustered close but separate from C. pauciramosa (ex-type CMW 5683) and C. polizzi (CBS 125270 & CBS 125271). A further seven isolates (CBS 136642, CBS 136643, CBS 136644, CBS 136645, CBS 136647, CBS 136651 & CBS 136653) formed a clade (BS = 60; PP = 1.00) close but separate to C. carciaina (ex-type CBS 123693) with four isolates (CBS 136604, CBS 136606, CBS 136609 & CBS 136251) forming a sister clade (BS = 72; PP = 1.00) to these seven isolates. Three isolates (CBS 136641, CMW 31394 & CMW 31395) formed a clade (BS = 100; PP = 1.00) close but separate from C. brasiliensis (ex-type CBS 230.51) and C. sulawesiensis (CBS 125248 & CBS 125253).

The dataset representing the Sphaero-Naviculate Group of isolates included 85 ingroup taxa, with C. pauciramosa (CMW 5683 & CMW 30823) as the outgroup taxon. This dataset consisted of 2 016 characters, of which 1 369 were constant, 127 were parsimony-uninformative and 520 were parsimony-informative. The MP analysis yielded 1 000 trees (TL = 1 512; CI = 0.612; RC = 0.538; RI = 0.952) of which the first is presented (Fig. 1). The majority of the isolates included in this dataset clustered in the clade (BS < 50; PP = 1.00) representing C. hongkongensis (ex-type CBS 230.51) as the outgroup taxon. This dataset consisted of 2 018 characters, of which 1 369 were constant, 127 were parsimony-uninformative and 520 were parsimony-informative. The MP analysis yielded 1 000 trees (TL = 1 512; CI = 0.612; RC = 0.538; RI = 0.952) of which the first is presented (Fig. 1). The majority of the isolates included in this dataset clustered in the clade (BS < 50; PP = 1.00) representing C. hongkongensis (ex-type CBS 114828) with four isolates (CBS 136077, CBS 136093, CBS 136652 & CMW 35383) forming a sister clade (BS = 78; PP = 1.00) to the C. hongkongensis clade. A single isolate (CBS 136629) formed a basal sister lineage to both these clades. Four isolates (CBS 136635, CMW 35383; CERC 1885; CBS 136093; CMW 35410; CERC 1901) were constant, 73 were parsimony-uninformative and 637 were parsimony-informative. The MP analysis yielded 1 000 trees (TL = 1 512; CI = 0.612; RC = 0.538; RI = 0.952) of which the first is presented (Fig. 1). The majority of the isolates included in this dataset clustered in the clade (BS < 50; PP = 1.00) representing C. pentaseptata (ex-type CBS 133439) with five isolates (CBS 136633, CBS 136634, CBS 136636, CBS 136638 & CBS 136640) forming a sister clade (BS < 50; PP = 0.96) to the C. pentaseptata clade. A clade (BS = 75; PP = 0.99) incorporating seven isolates (CBS 136630, CBS 136631, CBS 136632, CBS 136639, CBS 136640, CPC 23486 & CPC 23487), with an additional two isolates (CBS 136635 & CBS 136637) forming a sister clade (BS = 81; PP = 1.00), clustered close but separate from C. pauciramosa (ex-type CMW 5683) and C. polizzi (CBS 125270 & CBS 125271). A further seven isolates (CBS 136642, CBS 136643, CBS 136644, CBS 136645, CBS 136647, CBS 136651 & CBS 136653) formed a clade (BS = 60; PP = 1.00) close but separate to C. carciaina (ex-type CBS 123693) with four isolates (CBS 136604, CBS 136606, CBS 136609 & CBS 136251) forming a sister clade (BS = 72; PP = 1.00) to these seven isolates. Three isolates (CBS 136641, CMW 31394 & CMW 31395) formed a clade (BS = 100; PP = 1.00) close but separate from C. brasiliensis (ex-type CBS 230.51) and C. sulawesiensis (CBS 125248 & CBS 125253).

All descriptions, illustrations and nomenclatural data were deposited in MycoBank (Crous et al. 2004a).

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1 ATCC: American Type Culture Collection, Virginia, USA; CBS: CBS-KNAW Fungal Biodiversity Centre, Utrecht, The Netherlands; CERC: China Eucalyp Research Centre, Zhanjiang, Guangdong Province, China; CMW: culture collection of the Forestry and Agricultural Biotechnology Institute (FABI), University of Pretoria, Pretoria, South Africa; CPC: Pedro Crous working collection housed at CBS; HGUP: Plant Pathology Herbarium of Guizhou University, Guiyang 550025, China; IMI: International Mycological Institute, CABI-Bioscience, Edinburgh, UK; LPI: Laboratoire de Patologie Forestale, Université Libre de Bruxelles, Brussels, Belgium; UFV: Universidade Federal de Viçosa, Viçosa, Brazil. Isolates obtained during the survey are indicated in grey blocks.

2 cmdA = calmodulin, his3 = histone H3, tef1 = translation elongation factor 1-alpha. Ex-type isolates indicated in bold. Sequences generated in this study indicated in italics.
Fig. 1. One of 1 000 equally most parsimonious trees obtained from a heuristic search with 1 000 random taxon additions of the combined cmdA, his3, tef1 and tub2 sequence alignments of the Prolate group. Scale bar shows 5 changes. Bootstrap support values and Bayesian posterior probability values are shown at the nodes. The tree was rooted to C. hongkongensis (CBS 114711 & CBS 114878). Ex-type strains are indicated in bold.

LOMBARD ET AL.

160
Fig. 2. One of 1 000 equally most parsimonious trees obtained from a heuristics search with 1 000 random taxon additions of the combined cmdA, his3, tef1 and tub2 sequence alignments of the Sphaero-Naviculate Group. Scale bar shows 10 changes. Bootstrap support values and Bayesian posterior probability values are shown at the nodes. The tree was rooted to C. pauciramosa (CMW 5683 & CMW 30823). Ex-type strains are indicated in bold.
136082, CBS 136083, CBS 136088 & CBS 136090) clustered in a clade (BS = 100; PP = 1.00) with C. chinensis (ex-type CBS 114827). Sixteen isolates clustered near the C. kyotensis (ex-type CBS 413.67) clade (BS = 99; PP = 1.00) of which three isolates (CBS 136081, CBS 136976 & CMW 31439) formed single lineages. The remaining isolates clustered in four separate clades, three of which were well supported (BS = 99; PP = 1.00, BS = 76; PP = 1.00 & BS = 54; PP = 1.00, respectively). Of the remaining four isolates, two (CBS 136248 & CBS 136249) formed single lineages and two (CBS 136092 & CBS 136094) formed a unique clade (BS = 81; PP = 1.00).

**Taxonomy**

Morphological observation supported by phylogenetic inference showed that the majority of strains included in this study belonged to C.chinensis, C. hongkongensis and C. pentaseptata (Figs 1 and 2; Table 1). The remaining strains are shown to represent several distinct taxa that are provided names in Calonectria. Important morphological characters are summarised in Table 2.

**Calonectria aconidialis** L. Lombard, Crous & S.F. Chen, sp. nov. MycoBank MB809043. Fig. 3.

Etymology: Name refers to an absence of macroconidia in the fungus.

Ascomata perithecial, solitary or in groups of two, orange, becoming orange-brown with age; in section, apex and body orange, base red-brown, subglobose to ovoid, 357–444 μm high, 276–391 μm diam, body turning dark orange to red, and base dark red-brown in 3 % KOH; ascomatal wall rough, consisting of two thick-walled layers; outer layer of textura globulosa, 37–66 μm thick, cells becoming more compressed towards the inner layer of textura angularis, 18–20 μm thick, cells becoming thin-walled and hyaline towards the centre; outermost cells 34–71 × 34–55 μm, cells of inner layer 23–32 × 6–9 μm; ascomatal base up to 137 μm wide, consisting of dark red, angular cells, merging with an erumpent stroma; cells of the outer wall layer continuous with the pseudoparenchymatous cells of the erumpent stroma. Asci 8-spored, clavate, 97–119 × 16–19 μm, tapering into a long thin stalk. Ascospores aggregated in the upper third of the ascus, hyaline, gutiltate, fusoid with rounded ends, straight to slightly curved, 1-septate, constricted at the septum, (30–)35–41(–43) × 5–7(–8) μm (av. 38 × 7 μm). Homothallic. Macroconidiophores consist of a stipe bearing a penicillate arrangement of fertile branches, and a stipe extension terminating in a vesicle; stipe septate, hyaline, smooth, 40–133 × 6–10 μm; stipe extension septate, straight to flexuous, 134–196 μm long, 3–6 μm wide at the apical septum, terminating in a sphaeropodenduculate vesicle, 7–13 μm diam; lateral stipe extensions (90° to main axis) abundant. Conidiogenous apparatus 58–151 μm wide, and 54–108 μm long; primary branches aseptate, 18–42 × 5–8 μm; secondary branches aseptate, 10–27 × 4–7 μm; tertiary branches aseptate, 9–18 × 3–6 μm; quaternary branches and additional branches (–5) aseptate, 10–20 × 3–6 μm, each terminal branch producing 2–6 phialides; phialides doliiform to reniform, hyaline, aseptate, 9–15 × 2–4 μm, apex with minute periclinal thickening and inconspicuous collarette. Macroconidia cylindrical, rounded at both ends, straight, (41–)42–48(–52) × 4–6 μm (av. 45 × 5 μm), 1-septate, lacking a visible abscission scar, held in parallel cylindrical clusters by colourless slime. Mega- and microconidia not observed.

Culture characteristics: Colonies fast growing at 24 °C on MEA, producing abundant white to pale luteous aerial mycelium with profuse sporulation on the medium surface; reverse sienna to amber after 7 d; chlamydospores extensive throughout the medium forming microsclerotia.

Specimens examined: China, Guangxi Province, from soil collected in a Eucalyptus plantation, Mar. 2009, X. Zhou, G. Zhao & F. Han (holotype CBS H-21482, living ex-type culture CBS 136079 = CPC 23492 = CMW 31370 = CERC 1705), CPC 23491 = CMW 31369 = CERC 1704, CPC 23493 = CMW 31371 = CERC 1706, CMW 31368 = CERC 1703; Guangxi Province, Fangchenggang, from soil collected in a Eucalyptus plantation, Mar. 2009, X. Zhou, G. Zhao & F. Han, CMW 31367 = CERC 1702.

Note: Calonectria arbusta produces a larger conidiogenous apparatus than C. kyotensis and the ascospores and macroconidia of C. arbusta are also larger than those of C. kyotensis (Table 2).
Table 2. Morphological characteristics of Calonectria spp. included in this study.

| Species                        | Perithecia | Asci | Ascospores | Conidiogenous apparatus | Stipe extension | Vesicle | Macroconidia | Reference                  |
|--------------------------------|------------|------|------------|--------------------------|-----------------|---------|--------------|----------------------------|
|                                | Size (μm)  | Shape | Size (μm)  | Branches (μm)            | Diam (μm)       | Shape   | Size (μm)    |                            |
| **Calonectria reteaudii species complex** |            |       |            |                          |                 |         |              |                            |
| C. microconidialis             | 26–92 × 35–95 | 3    | 175–441 × 4–7 | 3–7 Narrowly clavate | (69–78) × 99(113) × 4–6(7) | 4–6(7) | This study   |                            |
| C. pentatseptata               | 23–90 × 70–99 | 3    | 168–350 × 3–6 | 2–6 Narrowly clavate | (75–87) × 109(115) × 5(–8) | 5(–8) | Crous et al. (2012) |                            |
| C. pseudoreticulata            | 26–82 × 45–103 | 3    | 193–313 × 5–6 | 3–5 Narrowly clavate | (61–65) × 73(–76) × 4–6(7) | 4–6(7) | Lombard et al. (2010d) |                            |
| C. queenslandica               | 27–68 × 39–64 | 3    | 105–156 × 4–5 | 3–4 Narrowly clavate | (61–65) × 73(–76) × 4–6(7) | 4–6(7) | Lombard et al. (2010d) |                            |
| C. reteaudii                   | 350–450 × (50–75) × 100 × (4–5–6–7) | 70–150 × 7–20 | (50–75) × 60 | 3–6 Clavate |          |         |              |                            |
| C. terra-reginae               | 33–48 × 35–54 | 4    | 127–235 × 4–6 | 3–5 Narrowly clavate | (60–83) × 87(–7) × 4–6(7) | 4–6(7) | Lombard et al. (2010d) |                            |
| **Calonectria candelabra species complex** |            |       |            |                          |                 |         |              |                            |
| C. brassiata                  | 50–135 × 50–80 | 3    | 90–172 × 2–3 | 3–7 Ellipsoid to narrowly obpyriform | (35–50) × 56(–65) × 3–5 | 1     | Alfenas et al. (2015) |                            |
| C. candelabra                  | 350–450 × 300–350 | 70–130 × 7–15 | (40–45) × 50(–60) × 5–6 | 3–8 Ellipsoid to narrowly obpyriform | (45–58) × 68(–80) × 4–5(–6) | 1     | Crous (2002) |                            |
| C. eucalypticola              | 45–75 × 35–62 | 3    | 145–170 × 2–4 | 5–7 Ellipsoid to obpyriform | (43–49) × 52(–55) × 3–5 | 1     | Alfenas et al. (2015) |                            |
| C. gleboicola                 | 25–40 × 27–45 | 2    | 100–165 × 2–4 | 3–6 Ellipsoid to narrowly obpyriform | (45–50) × 52(–55) × 3–5 | 1     | Alfenas et al. (2015) |                            |
| C. metrosideri                | 60–75 × 40–65 | 4    | 90–170 × 2–4 | 5–9 Spathulate to obpyriform | (40–44) × 46(–51) × 3–5 | 1     | Alfenas et al. (2013a) |                            |
| C. mossambicensis             | 37–87 × 19–59 | 3    | 91–203 × 2–6 | 2–8 Obpyriform to ellipsoidal | (35–38) × 46(–50) × 3–6 | 1     | Crous et al. (2013) |                            |
| C. nemerica                   | 50–80 × 40–60 | 4    | 150–205 × 6–12 | 7–13 Obpyriform | (40–44) × 46(–50) × 3–5 | 1     | Alfenas et al. (2015) |                            |
| C. pauciramosa                | 250–400 × 170–300 | 70–140 × 8–25 | (30–33) × 38(–40) × 6–7(–8) | 1     | 20–50 × 35–85 | 3    | 120–230 × 2–3 | 5–11 Obpyriform to ellipsoidal | (30–45) × 55(–60) × 3–5(–4) | 1     | Schoch et al. (1999) |                            |
| C. plinensis                  | 35–80 × 20–60 | 2    | 95–130 × 2–3 | 3–7 Ellipsoid to narrowly obpyriform | (38–47) × 52(–60) × 3–5 | 1     | Alfenas et al. (2015) |                            |
| C. polizzi                    | 28–51 × 27–57 | 3    | 111–167 × 5–6 | 6–9 Obpyriform to ellipsoidal | (31–32) × 42(–49) × 3–5 | 1     | Lombard et al. (2010a) |                            |
| C. pseudoscytophora           | 52–74 × 34–87 | 4    | 124–201 × 4–6 | 6–10 Obpyriform to ellipsoidal | (41–45) × 51(–52) × 3–3 | 1     | Lombard et al. (2010b) |                            |

(continued on next page)
Table 2. (Continued)

| Species                  | Perithecia | Asci | Ascospores | Conidiogenous apparatus | Stipe extension | Vesicle | Macroconidia | Reference          |
|--------------------------|------------|------|------------|--------------------------|-----------------|---------|--------------|-------------------|
|                          | Size (μm) | Shape | Size (μm) | Size (μm) | Branches | Size (μm) | Branches | Shape | Size (μm) | Diam (μm) | Shape | Size (μm) | Septation |
|                          |           |       |           |           |          |           |          |       |           |           |       |           |           |
| C. pseudopathulata       | 60–100    | × 30–70 | 3         | 145–190    | × 2–4    | 7–10     | Obpyriform | (35–)41–44(–50) | × 3–5 | 1 Aflenas et al. (2015) |
| C. seminaria             | 31–155    | × 36–72 | 3         | 105–185    | × 4–7    | 6–11     | Obpyriform to ellipsoidal | (42–)45–49(–52) | × 3.5–4.5(–7) | 1 This study |
| C. sylvicola             | 45–105    | × 35–90 | 3         | 130–195    | × 3–4    | 7–10     | Obpyriform | (30–)40–42(–50) | × 3–5 | 1 Aflenas et al. (2015) |
| C. tetramorosa           | 54–95     | × 36–75 | 4         | 102–253    | × 3–6    | 4–10     | Obpyriform | (45–)46.5–49.5(–51) | × (4–)5.5–5.5(–6) | 1 This study |
| C. zulensii              | 292–394   | × 170–295 | 92–140    | (26–)29–34(–38) | × 4–5 | 37–70     | × 35–67 | 3 | 110–171    | × 5–8 | 6–10 | Ellipsoid to obpyriform | (31–)34–38(–40) | × 3–5 | 1 Lombard et al. (2010a) |
| Cabonectria cylindrospora species complex |            |       |           |           |          |           |          |       |           |           |       |           |           |
| C. brasiliensis          | 81–103    | × 58–90 | 3         | 204–286    | × 6–7    | 7–11     | Ellipsoid to obpyriform | (35–)36–40(–41) | × 3–5 | 1 Lombard et al. (2010a) |
| C. ceratina              | 62–113    | × 70–98 | 4         | 148–222    | × 5–6    | 8–13     | Fusiform to obpyriform | (37–)41–46(–49) | × 5–8 | 1 Lombard et al. (2010d) |
| C. cylindrospora         | 280–520   | × 280–400 | 75–100    | (24–)30–40(–49) | × (4–)5–6(–8) | 60–100   | × 80–110 | 6 | 150–200    | × 3–4 | 6–8 | Ellipsoid to pyriform or clavate | (40–)42–50(–66) | × 3–4–5 | 1 Crous (2002) |
| C. fuciformis            | 76–180    | × 59–130 | 7         | 140–215    | × 4–6    | 6–13     | Obpyriform to ellipsoidal | (41–)44–50(–52) | × (3–)4–5(–6) | 1 This study |
| C. hawksworthii          | 40–90     | × 65–100 | 4         | 150–250    | × 2–3    | 6–9      | Ellipsoid to clavate | (38–)50–60(–76) | × 4–5 | 1 Crous (2002) |
| C. hodgesii              | 81–72     | × 45–65 | 3         | 136–196    | × 2–4    | 6–11     | Pyriform to ellipsoidal or ovoid to sphaeropedunculata | (44–)49–51(–55) | × 3–5 | 1 Aflenas et al. (2013b) |
| C. insulans              | 350–450   | × 300–350 | 70–125    | (27–)30–36(–42) | × 5–6(–7) | 45–90     | × 45–80 | 6 | 110–250    | × 4–5 | 4–13 | Obpyriform to broadly ellipsoidal | (33–)40–50(–60) | × 3.5–4 | 1 Crous (2002) |
| C. leucothoes            | 25–50     | × 50–80 | 6         | 160–250    | × 3–6    | 6–11.5   | Ellipsoid to obpyriform | (45–)58–78(–97) | × (4–)5–5.5(–8.5) | (1–)3(–6) | 1 Crous (2002) |
| C. maranhensis           | 45–65     | × 45–71 | 3         | 125–190    | × 3–5    | 7–11     | Ellipsoid, obpyriform to sphaeropedunculata | (50–)58–68(–85) | × (3–)5(–6) | 1 Aflenas et al. (2015) |
| C. mexicana              | 400–450   | × 350–450 | 70–120    | (35–)40–55(–65) | × 5–6(–7) | 25–60     | × 40–70 | 3 | 160–250    | × 2–3 | 7–12 | Broadly ellipsoidal with papillate apex | (35–)40–48(–52) | × 3–6–4.5 | 1 Crous (2002) |
| C. papillata             | 425–455   | × 345–395 | 106–112   | (27–)32–40(–46) | × 5–6(–7) | 45–114    | × 33–82 | 4 | 163–218    | × 4–7 | 8–14 | Ellipsoid to ellipsoidal with papillate apex | (40–)43–47(–50) | × (3–)3–4(–5) | 1 This study |
| C. propagincola          | 40–75     | × 31–85 | 4         | 130–250    | × 2–5    | 5–12     | Ellipsoid, obpyriform to sphaeropedunculata | (40–)48–51(–55) | × 3–5 | 1 Aflenas et al. (2015) |
Table 2. (Continued)

| Species              | Perithecia | Asci | Ascospores | Conidiogenous apparatus | Stipe extension | Vesicle | Macroconidia | Reference                  |
|----------------------|------------|------|------------|--------------------------|-----------------|---------|--------------|---------------------------|
|                      | Size (μm)  | Shape| Size (μm)  | Size (μm)                | Diam (μm)       | Shape   | Size (μm)    | Reference                  |
| C. pseudocerciana    | 90–90 × 40 | –    | 30–60      | 30–60                    | 150–190 × 2-5   | 7–12    | (35–43–46–55) × | 3–5                        |
|                      | 90–90 × 40 | –    | 30–60      | 30–60                    | 150–190 × 2-5   | 7–12    | (35–43–46–55) × | 3–5                        |
| C. pseudohodgesii    |            |      |            |                          | 150–190 × 2-5   | 7–12    | (35–43–46–55) × | 3–5                        |
| C. sulawesiensis     |            |      |            |                          | 150–190 × 2-5   | 7–12    | (35–43–46–55) × | 3–5                        |
| C. terrestris        |            |      |            |                          | 150–190 × 2-5   | 7–12    | (35–43–46–55) × | 3–5                        |
| Calonectria kyotensis species complex | |       |            |                          | 150–190 × 2-5   | 7–12    | (35–43–46–55) × | 3–5                        |
| C. aconitidis        | 297–366    | 111–113 | 58–151     | 56–151                   | 134–196 × 3–6   | 7–13    | (41–42–48–52) × | 4–6                        |
| C. articula          | 357–444    | 97–119 | 58–151     | 56–151                   | 134–196 × 3–6   | 7–13    | (41–42–48–52) × | 4–6                        |
| C. asiatica          | 280–400    | 70–120 | (28–40–40) | 40–80                    | 200–280 × 3–7   | 12–17   | (42–48–55–65) × | 4–5                        |
| C. canadensis        | 200–350    | 110–150 | (28–40–40) | 40–80                    | 200–280 × 3–7   | 12–17   | (42–48–55–65) × | 4–5                        |
| C. chiensis          | 350–500    | 100–140 | (28–40–40) | 40–80                    | 200–280 × 3–7   | 12–17   | (42–48–55–65) × | 4–5                        |
| C. curvispora        | 300–500    | 110–150 | (28–40–40) | 40–80                    | 200–280 × 3–7   | 12–17   | (42–48–55–65) × | 4–5                        |
| C. expansa           | 310–520    | 107–146 | 58–151     | 56–151                   | 134–196 × 3–6   | 7–13    | (41–42–48–52) × | 4–6                        |
| C. guangxiensis      | 295–435    | 83–146 | (28–40–40) | 40–80                    | 200–280 × 3–7   | 12–17   | (42–48–55–65) × | 4–5                        |
| C. hainanensis       | 300–500    | 91–110 | (28–40–40) | 40–80                    | 200–280 × 3–7   | 12–17   | (42–48–55–65) × | 4–5                        |
| C. hongkongensis     | 350–500    | 80–140 | (28–40–40) | 40–80                    | 200–280 × 3–7   | 12–17   | (42–48–55–65) × | 4–5                        |
| C. ilicicola         | 300–500    | 90–140 | (30–37–50) | 30–150                   | 120–140 × 3-4   | 6–12    | (33–43–46–55) × | 4–5                        |
| C. indonesiae        | 280–400    | 100–140 | (30–37–50) | 30–150                   | 120–140 × 3-4   | 6–12    | (33–43–46–55) × | 4–5                        |
| C. kyotensis         | 280–550    | 70–140 | (30–37–50) | 30–150                   | 120–140 × 3-4   | 6–12    | (33–43–46–55) × | 4–5                        |
|                      | 210–425    | 13–22 | (30–37–50) | 30–150                   | 120–140 × 3-4   | 6–12    | (33–43–46–55) × | 4–5                        |

(continued on next page)
| Species          | Perithecia  | Asci          | Ascospores    | Conidiogenous apparatus | Stipe extension | Vesicle        | Macroconidia | Reference                  |
|------------------|-------------|---------------|---------------|--------------------------|-----------------|----------------|--------------|---------------------------|
|                  | Size (μm)   | Shape         | Size (μm)     | Shape                    | Branches (μm)   | Diam (μm)     | Shape        | Size (μm)            | Septation |
| C. lateralis     | 43–138 × 41–104 | 6             | 150–225 × 4–6 | 9–13 Sphaeropedunculate | 35–37–41–44) × 4–5 | 1              | This study   |
| C. magnispora    | 280–550 × 210–425 | 91–125 × 14–17 | [33–]36–44–49) × 5–7(–8) | 1 | 47–95 × 47–80 | 4 | 161–278 × 4–7 | 9–18 Sphaeropedunculate | (46–)49–55(–)60 × 4–6(–)7 | 1 | This study |
| C. malesiana     | 30–80 × 50–60 | 6             | 120–200 × 3–4 | 8–15 Sphaeropedunculate to globose | (34–)45–52(–)55 × 3–4 | 1 | Crous et al. (2004b) |
| C. pacifica      | 20–60 × 30–80 | 3             | 150–250 × 3–4 | 7–15 Sphaeropedunculate | (38–)45–65(–)75 × 4–5 | 1 | Crous (2002) |
| C. parakyoitensis| 49–98 × 41–84 | 4             | 135–210 × 4–6 | 10–14 Sphaeropedunculate | (39–)42–46(–)49 × 4–5(–)6 | 1 | This study |
| C. pluriramosa   | 76–177 × 59–127 | 7             | 140–215 × 4–6 | 6–13 Sphaeropedunculate | (41–)44–50(–)52 × 3–4(–)5(–)6 | 1 | This study |
| C. pseudokyoitensis| 43–103 × 76–109 | 4             | 145–320 × 5–7 | 10–13 Pyriform to sphaeropedunculate | (43–)45–51(–)53 × 5–7 | 1 | This study |
| C. sphaeropedunculata | 470–575 × 345–468 | 82–144 × 11–23 | [31–]33–40–42) × 5–7(–8) | 1 | 63–144 × 40–111 | 6 | 152–253 × 4–8 | 10–14 Sphaeropedunculate | (40–)43–47(–)49 × 4–6 | 1 | This study |
| C. sumatrensis   | 40–60 × 50–60 | 3             | 180–260 × 3–4 | 8–13 Sphaeropedunculate | (45–)55–65(–)70 × 4–5(–)6 | 1 | Crous et al. (2004b) |
| C. turangicola   | 48–110 × 35–86 | 5             | 133–195 × 4–6 | 8–12 Sphaeropedunculate | (40–)42–46(–)47 × 3–5 | 1 | This study |

*Note: All measurements are in micrometers (μm).*
**Calonectria expansa** L. Lombard, Crous & S.F. Chen, sp. nov. MycoBank MB809046. Fig. 5.

*Etymology:* Name refers to Guangxi Province, the “Western Expanse”, where this fungus was first collected.

Ascomata perithecial, solitary, orange, becoming orange-brown with age; in section, apex and body orange, base red-brown, subglobose to ovoid, 310–520 μm high, 270–435 μm diam, body turning dark orange to red, and base dark red-brown in 3 % KOH; ascocatal wall rough, consisting of two thick-walled layers; outer layer of textura globulosa, 37–64 μm thick, cells becoming more compressed towards the inner layer of textura angularis, 13–25 μm thick, cells becoming thin-walled and hyaline towards the centre; outermost cells 13–31 × 9–20 μm, cells of inner layer 9–18 × 3–5 μm; ascocatal base up to 150 μm wide, consisting of dark red, angular cells, merging with an erumpent stroma; cells of the outer wall layer continuous with the pseudoparenchymatous cells of the erumpent stroma. Ascii 8-spored, clavate, 107–146 × 16–21 μm, tapering into a long thin stalk. Ascospores aggregated in the upper third of the ascus, hyaline, guttulate, fusoid with rounded ends, straight to slightly curved, 1-septate, sometimes constricted at the septum, (33–)36–41(–44) × (4–)5–7 μm (av. 39 × 6 μm). Homothallic. *Macroconidiophores* consist of a stipe bearing a penicillate arrangement of fertile branches, and a stipe extension terminating in a vesicle; stipe septate, hyaline, smooth, 61–169 × 5–10 μm; stipe extension septate, straight to flexuous, 124–216 μm long, 3–7 μm wide at the apical septum, terminating in a sphaeropedunculate vesicle, 8–16 μm diam; lateral stipe extensions (90° to main axis) abundant. *Conidiogenous apparatus* 26–116 μm wide, and 45–82 μm long; primary branches aseptate, 18–29 × 5–7 μm; secondary branches aseptate, 12–22 × 4–7 μm; tertiary branches aseptate, 9–16 × 3–6 μm; quaternary branches and additional branches (～5) aseptate, 12–18 × 3–5 μm, each terminal branch producing 2–6 phialides; phialides doliform to reniform, hyaline, aseptate, 10–18 × 3–5 μm, apex with minute periclinal thickening and inconspicuous collarette. *Macroconidia* cylindrical, rounded at both ends, straight, (44–)48–52(–57) × 4–6 μm (av. 52 × 5 μm), 1-septate, lacking a visible abscission scar, held in parallel cylindrical clusters by colourless slime. *Mega- and microconidia* not observed.

*Culture characteristics:* Colonies fast growing at 24 °C on MEA, producing abundant white aerial mycelium with profuse sporulation on the medium surface; reverse sienna to umber after 7 d; chlamydospores extensive throughout the medium forming microsclerotia.

Specimens examined: China. Guangxi Province, from soil collected in a Eucalyptus plantation, Mar. 2009, X. Zhou, G. Zhao & F. Han (holotype CBS H-21483, living ex-type culture CBS 136247 = CPC 23485 = CMW 31392 = CERC 1727); Guangxi Province, Fangchenggang, from soil collected in a Eucalyptus plantation, Mar. 2009, X. Zhou, G. Zhao & F. Han, CBS 136078 = CMW 31441 = CERC 1776, CMW 31413 = CERC 1748.

*Note:* Calonectria expansa can be distinguished from *C. arbusta* and *C. kyotensis* by its larger macroconidia and longer stipe extension (Table 2).

**Calonectria foliicola** L. Lombard, Crous & S.F. Chen, sp. nov. MycoBank MB809047. Fig. 6.

*Etymology:* Name refers to the natural habitat of this species, being a foliar pathogen.

Ascomata not observed. *Macroconidiophores* consist of a stipe bearing a penicillate arrangement of fertile branches, and a stipe extension terminating in a vesicle; stipe septate, hyaline, smooth, 47–190 × 6–12 μm; stipe extension septate, straight to flexuous, 140–215 μm long, 4–6 μm wide at the apical septum,
terminating in an obovate to ellipsoidal vesicle, 6–13 μm diam. 

Conidiogenous apparatus 76–180 μm wide, and 59–130 μm long; primary branches aseptate, 17–37 × 5–8 μm; secondary branches aseptate, 16–30 × 4–7 μm; tertiary branches aseptate, 11–23 × 4–6 μm; quaternary and additional branches (–7) aseptate, 9–20 × 3–6 μm, each terminal branch producing 2–6 phialides; phialides doliiform to reniform, hyaline, aseptate, 8–13 × 3–5 μm, apex with minute periclinal thickening and inconspicuous collarette. Macroconidia cylindrical, rounded at both ends, straight, (41–)44–50(–52) × (3–)4–5(–6) μm (av. 47 × 5 μm), 1-septate, lacking a visible abscission scar, held in parallel cylindrical clusters by colourless slime. Mega- and microconidia not observed.

Culture characteristics: Colonies fast growing at 24 °C on MEA, producing abundant white aerial mycelium and sporulating moderately on the medium surface; reverse sienna to umber after 7 d; chlamydospores formed abundantly throughout the medium, forming microsclerotia.

Specimen examined: China, Guangxi Province, from *E. urophylla* × *E. grandis* clone leaf, Mar. 2009, X. Zhou & G. Zhao (holotype CBS H-21472, living ex-type culture CBS 136641 = CPC 23491 = CMW 31393 = CERC 1728), CPC 23492 = CMW 31394 = CERC 1729, CMW 31395 = CERC 1730.

Notes: *Calonectria foliicola* is closely related to *C. brasiliensis* and *C. sulawesiensis* and can be distinguished from these species by the formation of up to seven levels of conidiophore branches.
branches. The macroconidia of *C. foliicola* are larger than those of *C. brasiliensis* but slightly smaller than those of *C. sulawesiensis* (Table 2).

**Calonectria guangxiensis** L. Lombard, Crous & S.F. Chen, sp. nov. MycoBank MB809049. Fig. 7.

**Etymology:** Name refers to the Guangxi Province of China where the fungus was first collected.

Ascomata perithecial, solitary or in groups of two, orange, becoming orange-brown with age; in section, apex and body orange, base red-brown, subglobose to ovoid, 295–435 μm high, 265–355 μm diam, body turning dark orange to red, and base dark red-brown in 3 % KOH; ascomatal wall rough, consisting of two thick-walled layers; outer layer of **textura globulosa**, 32–80 μm thick, cells becoming more compressed towards the inner layer of **textura angularis**, 14–22 μm thick, cells becoming thin-walled and hyaline towards the centre; outermost cells 13–26 × 10–15 μm, cells of inner layer 11–15 × 4–5 μm; ascomatal base up to 175 μm wide, consisting of dark red, angular cells, merging with an erumpent stroma; cells of the outer wall layer continuous with the pseudoparenchymatous cells of the erumpent stroma. Asci 8-spored, clavate, 83–146 × 15–23 μm, tapering into a long thin stalk. Ascospores aggregated in the upper third of the ascus, hyaline, guttulate, fusoid with rounded ends, straight to slightly...
curved, 1-septate, constricted at the septum, (23–) 32–40(-42) × 5–7(-8) μm (av. 36 × 6 μm). Homothallic. Macroconidiophores consist of a stipe bearing a penicillate arrangement of fertile branches, and a stipe extension terminating in a vesicle; stipe septate, hyaline, smooth, 91–182 × 7–9 μm; stipe extension septate, straight to flexuous, 175–193 μm long, 5–7 μm wide at the apical septum, terminating in a sphaeropedunculate vesicle, 11–14 μm diam; lateral stipe extensions (90° to main axis) rare. Conidiogenous apparatus 31–95 μm wide, and 55–85 μm long; primary branches aseptate, 17–26 × 4–7 μm; secondary branches aseptate, 10–19 × 3–6 μm; tertiary branches aseptate, 9–17 × 2–5 μm; quaternary branches aseptate, 12–16 × 3–5 μm, each terminal branch producing 2–6 phialides; phialides doliiform to reniform, hyaline, aseptate, 8–19 × 3–7 μm, apex with minute periclinal thickening and inconspicuous collarette. Macroconidia cylindrical, rounded at both ends, straight, (42–) 45–49(-52) × 4–6 μm (av. 47 × 5 μm), 1-septate, lacking a visible abscission scar, held in parallel cylindrical clusters by colourless slime. Mega- and microconidia not observed.

Culture characteristics: Colonies fast growing at 24 °C on MEA, producing abundant white to cream-coloured aerial mycelium and sporulating profusely on the medium surface at the edge of the colony; reverse sienna to umber after 7 d; chlamydospores formed abundantly throughout the medium, forming microsclerotia.

Specimen examined: China, Guangxi Province, from soil collected in a Eucalyptus plantation, Aug. 2009, X. Mou & R. Chang (holotype CBS H-21484, culture ex-type CBS 136092 = CPC 23506 = CMW 35409 = CERC 1900), CBS 136094 = CPC 23507 = CMW 35411 = CERC 1902.

Notes: Calonectria guangxiensis can be distinguished from other species in the C. kyotensis complex by having fewer conidiophore branches and rarely forming lateral stipe extensions. The macroconidia of C. guangxiensis are slightly smaller than those of C. expansa and C. kyotensis and slightly larger than those of C. arbusta (Table 2).

Calonectria hainanensis L. Lombard, Crous & S.F. Chen, sp. nov. MycoBank MB809050. Fig. 8.

Etymology: Name refers to the Hainan Province of China where the fungus was first collected.

Ascomata perithecial, solitary, orange, becoming orange-brown with age; in section, apex and body orange, base red-brown,
subglobose to ovoid, 300–455 μm high, 230–385 μm diam, body turning dark orange to red, and base dark red-brown in 3 % KOH+; ascomatal wall rough, consisting of two thick-walled layers; outer layer of textura globulosa, 30–64 μm thick, cells becoming more compressed towards the inner layer of textura angularis, 10–16 μm thick, cells becoming thin-walled and hyaline towards the centre; outermost cells 16–42 × 13–42 μm, cells of inner layer 23–39 × 8–10 μm; ascomatal base up to 262 μm wide, consisting of dark red, angular cells, merging with an erumpent stroma; cells of the outer wall layer continuous with the pseudoparenchymatous cells of the erumpent stroma. Asci 8-spored, clavate, 91–110 × 15–22 μm, tapering into a long thin stalk. Ascospores aggregated in the upper third of the ascus, hyaline, guttulate, fusoid with rounded ends, straight to slightly curved, sometimes constricted at the septum, (24–)30–38(–42) × (4–)5–7 μm (av. 34 × 6 μm). Homothallic. Macroconidiophores
consisting of a stipe bearing a penicillate arrangement of fertile branches, and a stipe extension terminating in a vesicle; stipe septate, hyaline, smooth, 66–106 × 8–14 μm; stipe extension septate, straight to flexuous, 112–186 μm long, 4–11 μm wide at the apical septum, terminating in a sphaeropedunculate vesicle, 7–14 μm diam; lateral stipe extensions (90° to main axis) abundant. Conidiogenous apparatus 54–119 μm wide, and 41–80 μm long; primary branches aseptate, 18–28 × 5–9 μm; secondary branches aseptate, 12–21 × 5–8 μm; tertiary branches aseptate, 10–19 × 3–6 μm; quaternary branches and additional branches (–5) aseptate, 9–15 × 3–5 μm, each terminal branch producing 2–6 phialides; phialides doliiform to reniform, hyaline, aseptate, 10–17 × 3–5 μm, apex with minute periclinal thickening and inconspicuous collarette. Macroconidia cylindrical, rounded at both ends, straight, (41–)43–49(–52) × 4–6 μm (av. 46 × 5 μm), 1-septate, lacking a visible abscission scar, held in parallel cylindrical clusters by colourless slime. Mega- and microconidia not observed.
Culture characteristics: Colonies fast growing at 24 °C on MEA, producing abundant white to pale luteous aerial mycelium and sporulating profusely on the medium surface; reverse sienna toumber after 7 d; chlamydospores formed abundantly throughout the medium, forming microsclerotia.

Specimen examined: China, Hainan Province, from soil collected in a Eucalyptus plantation, Aug. 2009, X. Mou & S.F. Chen (holotype CBS H-21480, culture ex-type CBS 136248 = CPC 23505 = CMW 35187 = CERC 1863).

Notes: Based on morphological characteristics, C. hainanensis closely resembles C. malesiana. However, C. hainanensis readily produces fertile ascomata in culture, a feature not observed for C. malesiana (Crous et al. 2004b). Furthermore, C. hainanensis has fewer conidiophore branches than reported for C. malesiana, and the macroconidia of C. hainanensis are slightly smaller than those of C. malesiana (Table 2).

**Calonectria lateralis** Lombard, Crous & S.F. Chen, sp. nov. MycoBank MB809051. Fig. 9.

*Etymology:* Name refers to the lateral stipe extensions on its macroconidiophores.

Ascomata not observed. **Macroconidiophores** consisting of a stipe bearing a penicillate arrangement of fertile branches, and a stipe extension terminating in a vesicle; stipe septate, hyaline, smooth, 55–185 × 4–8 μm; stipe extension septate, straight to flexuous, 150–225 μm long, 4–6 μm wide at the apical septum, terminating in a sphaeropedunculate vesicle, 9–13 μm diam; lateral stipe extensions (90° to main axis) abundant. **Conidiogenous apparatus** 43–138 μm wide, and 41–104 μm long; primary branches aseptate, 17–28 × 4–7 μm; secondary branches aseptate, 11–26 × 3–7 μm; tertiary branches aseptate, 8–20 × 3–6 μm; quaternary and additional branches (~6) aseptate, 8–17 × 3–5 μm, each terminal branch producing 2–6 phialides; phialides doliiform to reniform, hyaline, aseptate, 7–13 × 2–4 μm, apex with minute periclinal thickening and inconspicuous collarette. **Macroconidia** cylindrical, rounded at both ends, straight, (35–)37–41(–44) × 4–5 μm (av. 39 × 4 μm), 1-septate, lacking a visible abscission scar, held in parallel cylindrical clusters by colourless slime. **Mega-** and **microconidia** not observed.

**Culture characteristics:** Colonies fast growing at 24 °C on MEA, producing abundant white to sienna aerial mycelium with profuse...
sporulation on the medium surface; reverse sienna to umber after 7 d; chlamydospores extensive throughout the medium forming microsclerotia.

Specimen examined: China, Guangxi Province, Fangchenggang, from soil collected in a Eucalyptus plantation, Aug. 2009, X. Zhou, G. Zhao & F. Han (holotype CBS H-21469, living ex-type CBS 136629 = CMW 31412 = CERC 1747).

Notes: Calonectria lateralis is closely related to C. hongkongensis and can be distinguished by having smaller macroconidia as compared to C. hongkongensis, and the stipe extensions of C. lateralis being longer than those of C. hongkongensis (Table 2).

**Calonectria magnispora** L. Lombard, Crous & S.F. Chen, sp. nov. MycoBank MB809052. Fig. 10.

Etymology: Name reflects the characteristically large ascospores produced by this fungus.

Ascomata perithecial, solitary, orange, becoming orange-brown with age; in section, apex and body orange, base red-brown, subglobose to ovoid, 390–495 μm high, 315–410 μm diam, body turning dark orange to red, and base dark red-brown in 3 % KOH+; ascomatal wall rough, consisting of two thick-walled layers; outer layer of textura globulosa, 53–91 μm thick, cells becoming more compressed towards the inner layer of textura...
Calyptopera, 16–20 μm thick, cells becoming thin-walled and hyaline towards the centre; outermost cells 16–28 × 10–18 μm, cells of inner layer 9–20 × 3–6 μm; ascomatal base up to 166 μm wide, consisting of dark red, angular cells, merging with an erumpent stroma; cells of the outer wall layer continuous with the pseudoparenchymatous cells of the erumpent stroma. Asci 8-spored, clavate, 91–125 × 14–17 μm, tapering into a long thin stalk. Ascospores aggregated in the upper third of the ascus, hyaline, guttulate, fusoid with rounded ends, straight to slightly curved, 1-septate, not constricted at the septum, (33–) 36–44(–49) × 5–7(–8) μm (av. 40 × 6 μm). Homothallic. Macroconidiophores consisting of a stipe bearing a penicillate arrangement of fertile branches, and a stipe extension terminating in a vesicle; stipe septate, hyaline, smooth, 57–139 × 7–11 μm; stipe extension septate, straight to flexuous, 161–278 μm long, 4–7 μm wide at the apical septum, terminating in a sphaeropedunculate vesicle, 9–18 μm diam; lateral stipe extensions (90° to main axis) moderately formed. Connidiogenous apparatus 47–95 μm wide, and 47–80 μm long; primary branches aseptate, 18–35 × 5–9 μm; secondary branches aseptate, 13–23 × 3–7 μm; tertiary branches aseptate, 10–19 × 3–5 μm; quaternary branches aseptate, 12–16 × 3–5 μm, each terminal branch producing 2–6 phialides; phialides doliform to reniform, hyaline, aseptate, 8–16 × 3–5 μm, apex with minute periclinal thickening and inconspicuous collarette. Macroconidia cylindrical, rounded at both ends, straight, (46–)49–55(–60) × 4–6(–7) μm (av. 52 × 5 μm), 1-septate, lacking a visible abscission scar, held in parallel cylindrical clusters by colourless slime. Macroconidiophores simple with some lateral branching, comprising a stipe and a penicillate or subverticillate arrangement of fertile branches. Stipe septate, hyaline, smooth, 53–86 × 7–8 μm; primary branches aseptate, straight, 19–26 × 4–5 μm, terminating in 1–3 phialides that are cylindrical to allantoid, 12–27 × 4–5 μm; apex with minute periclinal thickening and collarette. Microconidia cylindrical, straight, rounded at the apex, flattened at the base, (23–)31–47(–58) × 4–6(–7) μm (av. 39 × 5 μm), 1–3-septate, held in fascicles by colourless slime. Megaconidia not observed. 

Culture characteristics: Colonies fast growing at 24 °C on MEA, producing abundant white aerial mycelium and sporulating profusely on the medium surface, forming white to amber colonies with irregular margins; reverse sienna to umber after 7 d. Chlamydospores not observed. 

Specimens examined: China, Guangdong Province, Zhanjiang, CERC nursery, on E. urophylla × E. grandis clone seedling leaf, Mar. 2009, G. Zhao (holotype CBS H-21473, culture ex-type CBS 136638 = CMW 31487 = CERC 1822), CBS 136640 = CMW 31492 = CERC 1827 (Herb. CBS H-21474); Guangdong Province, Zhanjiang, CERC nursery, on E. urophylla × E. grandis clone seedling leaf, Mar. 2009, G. Zhao, CBS 136633 = CMW 31471 = CERC 1806, CBS 136634 = CMW 31473 = CERC 1810, CBS 136636 = CMW 31475 = CERC 1810. 

Notes: Calonectria microconidialis resides in the C. reteaudii complex (Lombard et al. 2010d, Crous et al. 2012). The ability of C. microconidialis to produce macroconidiophores and microconidia in culture distinguishes it from C. pentaseptata, C. queenslandica and C. terrae-reginae (Lombard et al. 2010d, Crous et al. 2012). The micro- and macroconidia of C. microconidialis are slightly larger than those of C. reteaudii but slightly smaller than those of C. pseudoreteaudii (Table 2). 

Calonectria papillata L. Lombard, Crous & S.F. Chen, sp. nov. MycoBank MB809054. 

Etymology: Name refers to the papillate apices of the stipe vesicles. 

Ascomata perithecial, solitary, orange, becoming orange-brown with age; in section, apex and body orange, base red-brown, subglobose to ovoid, 425–455 μm high, 345–395 μm diam, body turning dark orange to red, and base dark red-brown in 5 % K0H; ascostomal wall rough, consisting of two thick-walled layers; outer layer of textura globulosa, 49–57 μm thick, cells becoming more compressed towards the inner layer of textura angularis, 21–22 μm thick, cells becoming thin-walled and hyaline towards the centre; outermost cells 25–52 × 21–38 μm, cells of inner layer 11–21 × 5–9 μm; ascostomal base up to 200 μm wide, consisting of dark red, angular cells, merging with an erumpent stroma; cells of the outer wall layer continuous with the pseudoparenchymatous cells of the erumpent stroma. AscI 8-spored, clavate, 106–112 × 16–20 μm, tapering into a long thin stalk. Ascospores aggregated in the upper third of the ascus, hyaline, guttulate, fusoid with rounded ends, straight to slightly curved, 175–441 μm long, 4–7 μm wide at the apical septum, terminating in a narrowly clavate vesicle, 3–7 μm diam. Connidiogenous apparatus 26–92 μm wide, and 35–95 μm long; primary branches aseptate or 1-septate, 23–34 × 5–7 μm; secondary branches aseptate, 16–28 × 3–6 μm; tertiary branches aseptate, 14–24 × 3–6 μm, each terminal branch producing 1–3 phialides; phialides cylindrical to allantoid, hyaline, aseptate, 12–25 × 3–5 μm, apex with minute periclinal thickening and inconspicuous collarette. Macroconidia cylindrical, rounded at both ends, straight, (69–)78–98(–113) × 7–9(–10) μm (av. 88 × 8 μm), 4–6(7)-septate, lacking a visible abscission scar, held in parallel cylindrical clusters by colourless slime. Macroconidiophores simple with some lateral branching, comprising a stipe and a penicillate or subverticillate arrangement of fertile branches. Stipe septate, hyaline, smooth, 53–86 × 7–8 μm; primary branches aseptate, straight, 19–26 × 4–5 μm, terminating in 1–3 phialides that are cylindrical to allantoid, 12–27 × 4–5 μm; apex with minute periclinal thickening and collarette. Microconidia cylindrical, straight, rounded at the apex, flattened at the base, (23–)31–47(–58) × 4–6(–7) μm (av. 39 × 5 μm), 1–3-septate, held in fascicles by colourless slime. Megaconidia not observed. 

Culture characteristics: Colonies slow growing at 24 °C on MEA with mycelia immersed in the media, sporulating profusely on the medium surface, forming white to amber colonies with irregular margins; reverse sienna to umber after 7 d. Chlamydospores not observed. 

Notes: Calonectria magnispora can be distinguished from C. arbusa, C. expansa, C. guangxiensis, C. hainanensis and C. kyotensis by having larger ascospores and macroconidia. The stipe extensions of C. magnispora are also longer than observed in these species (Table 2).
curved, 1-septate, constricted at the septum, (27–)32–40(–46) × 5–6(–7) μm (av. 36 × 6 μm). Homothallic. 

Macroconidiophores consisting of a stipe bearing a penicillate arrangement of fertile branches, and a stipe extension terminating in a vesicle; stipe septate, hyaline, smooth, 54–245 × 6–11 μm; stipe extension septate, straight to flexuous, 163–218 μm long, 4–7 μm wide at the apical septum, terminating in a obpyrifrom to ellipsoidal vesicle with a papillate apex, 8–14 μm diam. Conidiogenous apparatus 45–114 μm wide, and 33–82 μm long; primary branches aseptate, 18–32 × 5–9 μm; secondary branches aseptate, 11–25 × 4–7 μm; tertiary branches aseptate, 9–12 × 3–4 μm; quaternary branches aseptate, 8–19 × 2–5 μm; periclinal branches aseptate, 9–12 × 3–4 μm, each terminal branch producing 2–4 phialides; phialides doliiform to reniform, hyaline, aseptate, 7–16 × 3–4 μm, apex with minute periclinal thickening and inconspicuous collarette. Macroconidia cylindrical, rounded at both ends, straight, (40–)43–47(–50) × (3–)4–5 μm (av. 45 × 4 μm), 1-septate, lacking a visible abscission scar, held in parallel cylindrical clusters by colourless slime. 

Mega- and microconidia not observed.

Culture characteristics: Colonies fast growing at 24 °C on MEA, producing abundant white aerial mycelium and sporulating profusely on the medium surface; reverse sienna to umber after 7 d; chlamydospores abundant throughout the medium, forming microsclerotia.

Specimens examined: China. Guangdong Province, from soil collected in a Eucalyptus plantation, Aug. 2009, X. Mou & R. Chang (holotype CBS H-21487, living ex-type CBS 136097 = CPC 23517 = CERC 1939), Guangdong Province, from soil collected in a Eucalyptus plantation, Aug. 2009, X. Mou & R. Chang, CBS 136084 = CPC 23497 = CMW 35165 = CERC 1841, CBS 136096 = CPC 23515 = CMW 37972 = CERC 1935, Guangxi Province, from soil collected in a Eucalyptus plantation, Aug. 2009, X. Mou & R. Chang, CBS 136251 = CPC 23514 = CMW 37971 = CERC 1934.

Notes: Calonectria papillata can be distinguished from both C. cerciana and C. terrestris by the papillate apices of the terminal vesicles on the stipe extension. This species is also homothallic, which is not the case for C. cerciana (Lombard et al. 2010d) or C. terrestris.

Calonectria parakyotensis L. Lombard, Crous & S.F. Chen, sp. nov. MycoBank MB809055. Fig. 13.
Etymology: Name refers to fact that this species has an asexual morph that is very similar to that of *C. kyotensis*.

Ascomata not observed. *Macroconidiophores* consisting of a stipe bearing a penicillate arrangement of fertile branches, and a stipe extension terminating in a vesicle; stipe septate, hyaline, smooth, 42–125 × 5–9 μm; stipe extension septate, straight to flexuous, 135–210 μm long, 4–6 μm wide at the apical septum, terminating in a sphaeropedunculate vesicle, 10–14 μm diam; lateral stipe extensions (90° to main axis) rare. *Conidiogenous apparatus* 49–98 μm wide, and 41–84 μm long; primary branches aseptate, 15–34 × 5–8 μm; secondary branches aseptate, 10–17 × 4–7 μm; tertiary branches aseptate, 9–17 × 3–6 μm; quaternary branches aseptate,

**Fig. 12.** *Calonectria papillata* (ex-type CBS 136097). A. Ascoma. B–C. Vertical section through ascomata, showing wall structure. D. Asci. E. Ascospores. F–H. *Macroconidiophores*. I–L. Conidiogenous apparatus with conidiophore branches and doliiform to reniform phialides. M. Obpyriform to ellipsoid vesicles with papillate apex. N–P. *Macroconidia*. Scale bars: A = 500 μm; B = 100 μm (apply to C); D = 50 μm (apply to F–H); E = 10 μm (apply to I–P).
11–18 × 4–6 μm, each terminal branch producing 2–6 phialides; phialides doliform to reniform, hyaline, aseptate, 10–18 × 2–6 μm, apex with minute periclinal thickening and inconspicuous collarette. Macroconidia cylindrical, rounded at both ends, straight, (39–)(42–46) × 4–5(–6) μm (av. 44 × 5 μm), 1-septate, lacking a visible abscission scar, held in parallel cylindrical clusters by colourless slime. Mega- and microconidia not observed.

Culture characteristics: Colonies fast growing at 24 °C on MEA, producing abundant white aerial mycelium with profuse sporulation on the medium surface; reverse sienna to cinnamon after 7 d; chlamydospores extensive throughout the medium forming microsclerotia.

Specimens examined: China. Guangdong Province, from soil collected in a Eucalyptus plantation, Aug. 2009, X. Mou & R. Chang (holotype CBS H-21470; living ex-type CBS 136085 = CPC 23498 = CMW 35169 = CERC 1845); Guangxi Province, from soil collected in an Eucalyptus plantation, Aug. 2009, X. Mou & R. Chang, CBS 136095 = CPC 23508 = CMW 35413 = CERC 1904.

Note: Calonectria parakyotensis can be distinguished from other closely related species in the C. kyotensis complex by having fewer conidiophore branches and the fact that it rarely forms lateral stipe extensions.

Calonectria pluriramosa L. Lombard, Crous & S.F. Chen, sp. nov. MycoBank MB809056. Fig. 14.

Etymology: Name refers to the numerous conidiophore branches formed by this species.

Ascomata not observed. Macroconidiophores consisting of a stipe bearing a penicillate arrangement of fertile branches, and a stipe extension terminating in a vesicle; stipe septate, hyaline, smooth, 47–185 × 6–12 μm; stipe extension septate, straight to flexuous, 140–215 μm long, 4–6 μm wide at the apical septum, terminating in a sphaeropedunculate vesicle, 6–13 μm diam; lateral stipe extensions (90° to main axis) rare. Conidiogenous apparatus 76–177 μm wide, and 59–127 μm long; primary branches aseptate or 1-septate, 17–37 × 5–8 μm; secondary branches aseptate, 16–30 × 4–7 μm; tertiary branches aseptate, 11–23 × 4–6 μm; quaternary branches and additional branches (–7) aseptate, 9–20 × 3–6 μm, each terminal branch producing 2–6 phialides; phialides doliform to reniform, hyaline, aseptate, 8–13 × 3–5 μm, apex with minute periclinal thickening and inconspicuous collarette. Macroconidia cylindrical, rounded at both ends, straight, (41–)44–50(–52) × (3–)4–5(–6) μm (av. 47 × 5 μm), 1-septate, lacking a visible abscission scar, held in parallel cylindrical clusters by colourless slime. Mega- and microconidia not observed.

Culture characteristics: Colonies fast growing at 24 °C on MEA, producing abundant white to sienna aerial mycelium with profuse sporulation on the medium surface; reverse sienna to umber after 7 d; chlamydospores extensive throughout the medium forming microsclerotia.

Specimen examined: China. Guangxi Province, Fangshenggang, from soil collected in a Eucalyptus plantation, Mar. 2009, X. Zhou, G. Zhao & F. Han (holotype CBS H-21485; living ex-type culture CBS 136976 = CMW 31440 = CERC 1775).

Notes: Calonectria pluriramosa is closely related to C. kyotensis and C. pseudokyotensis but can be distinguished by having a greater number of conidiophore branches. The macroconidia of C. pluriramosa are larger than those of C. kyotensis (Table 2). Unlike the latter two species, C. pluriramosa also failed to produce viable ascomata in culture.

Calonectria pseudokyotensis L. Lombard, Crous & S.F. Chen, sp. nov. MycoBank MB809057. Fig. 15.

Etymology: Name refers to the morphological similarity to the asexual morph of C. kyotensis.
Ascomata not observed. Macroconidiophores consisting of a stipe bearing a penicillate arrangement of fertile branches, and a stipe extension terminating in a vesicle; stipe septate, hyaline, smooth, 85–205 × 6–10 μm; stipe extension septate, straight to flexuous, 145–320 μm long, 5–7 μm wide at the apical septum, terminating in a pyriform to sphaeropedunculate vesicle, 10–13 μm diam; lateral stipe extensions (90° to main axis) moderate. Conidiogenous apparatus 42–103 μm wide, and 76–109 μm long; primary branches aseptate or 1-septate, 24–40 × 5–8 μm; secondary branches aseptate, 14–32 × 5–7 μm; tertiary branches aseptate, 13–25 × 4–6 μm; quaternary branches aseptate, 14–24 × 4–6 μm, each terminal branch producing 2–6 phialides; phialides elongate doliiform to doliiform or reniform, hyaline, aseptate, 10–20 × 3–5 μm, apex with minute periclinal thickening and inconspicuous collarette. Macroconidia cylindrical, rounded at both ends, straight, (43–) 45–51(–53) × 5–7 μm (av. 48 × 6 μm), 1-septate, lacking a visible abscission scar, held in parallel cylindrical clusters by colourless slime. Mega- and microconidia not observed.

Culture characteristics: Colonies fast growing at 24 °C on MEA, producing abundant white aerial mycelium with profuse sporulation on the medium surface; reverse sienna to umber after 7 d; chlamydospores extensive throughout the medium forming microsclerotia.

Specimen examined: China. Guangxi Province, Fangchenggang, from soil collected in a Eucalyptus plantation, Mar. 2009, X. Zhou, G. Zhao & F. Han (holotype CBS H-21774, living ex-type culture CBS 137332 = CMW 31439 = CERC 1774).

Notes: Calonectria pseudokyotensis has fewer fertile branches than C. kyotensis. Furthermore, the stipe extensions of C. pseudokyotensis are longer than those of C. kyotensis, terminating in pyriform to sphaeropedunculate vesicles, not observed in C. kyotensis (Table 2).

Calonectria seminaria L. Lombard, Crous & S.F. Chen, sp. nov. MycoBank MB809058. Fig. 16.

Etymology: Name refers the fact that this species was collected in a nursery.

Ascomata not observed. Macroconidiophores consisting of a stipe bearing a penicillate arrangement of fertile branches, and a stipe extension terminating in a vesicle; stipe septate, hyaline, smooth, 85–205 × 6–10 μm; stipe extension septate, straight to flexuous, 145–320 μm long, 5–7 μm wide at the apical septum, terminating in a pyriform to sphaeropedunculate vesicle, 10–13 μm diam; lateral stipe extensions (90° to main axis) moderate. Conidiogenous apparatus 42–103 μm wide, and 76–109 μm long; primary branches aseptate or 1-septate, 24–40 × 5–8 μm; secondary branches aseptate, 14–32 × 5–7 μm; tertiary branches aseptate, 13–25 × 4–6 μm; quaternary branches aseptate, 14–24 × 4–6 μm, each terminal branch producing 2–6 phialides; phialides elongate doliiform to doliiform or reniform, hyaline, aseptate, 10–20 × 3–5 μm, apex with minute periclinal thickening and inconspicuous collarette. Macroconidia cylindrical, rounded at both ends, straight, (43–) 45–51(–53) × 5–7 μm (av. 48 × 6 μm), 1-septate, lacking a visible abscission scar, held in parallel cylindrical clusters by colourless slime. Mega- and microconidia not observed.

Culture characteristics: Colonies fast growing at 24 °C on MEA, producing abundant white aerial mycelium with profuse sporulation on the medium surface; reverse sienna to umber after 7 d; chlamydospores extensive throughout the medium forming microsclerotia.

Specimen examined: China. Guangxi Province, Fangchenggang, from soil collected in a Eucalyptus plantation, Mar. 2009, X. Zhou, G. Zhao & F. Han (holotype CBS H-21774, living ex-type culture CBS 137332 = CMW 31439 = CERC 1774).

Notes: Calonectria pseudokyotensis has fewer fertile branches than C. kyotensis. Furthermore, the stipe extensions of C. pseudokyotensis are longer than those of C. kyotensis, terminating in pyriform to sphaeropedunculate vesicles, not observed in C. kyotensis (Table 2).

Calonectria seminaria L. Lombard, Crous & S.F. Chen, sp. nov. MycoBank MB809058. Fig. 16.
extension terminating in a vesicle; stipe septate, hyaline, smooth, 39–101 × 6–10 μm; stipe extension septate, straight to flexuous, 105–185 μm long, 4–7 μm wide at the apical septum, terminating in an obovate to ellipsoid vesicle, 6–11 μm diam. Conidiogenous apparatus 31–155 μm wide, and 36–72 μm long; primary branches aseptate or 1-septate, 13–27 × 3–6 μm; secondary branches aseptate, 8–19 × 2–5 μm; tertiary branches aseptate, 10–20 × 2–6 μm, each terminal branch producing 2–6 phialides; phialides doliiform to reniform, hyaline, aseptate, 7–14 × 2–4 μm, apex with minute periclinal thickening and inconspicuous collarette. Macroconidia cylindrical, rounded at both ends, (42–) 45–49(–52) × 3.5–4.5(–7) μm (av. 47 × 5 μm), 1-septate, lacking a visible abscission scar, held in parallel cylindrical clusters by colourless slime. Mega- and microconidia not observed.

**Culture characteristics:** Colonies fast growing at 24 °C on MEA, producing abundant white aerial mycelium and sporulating profusely on the medium surface; reverse amber to sepia-brown after 7 d; chlamydospores formed extensively in the media, forming microsclerotia.

Specimens examined: China, Guangdong Province, Zhanjiang, CERC nursery, on E. urophylla × E. grandis clone seedling leaf, Mar. 2009, G. Zhao (holotype CBS 136632 = CPC 23488 = CMW 31450 = CERC 1785); Guangdong Province, Zhanjiang, CERC nursery, on E. urophylla × E. grandis clone seedling leaf, Mar. 2009, G. Zhao, CBS 136630 = CMW 31446 = CERC 1781, CBS 136631 = CMW 31449 = CERC 1784, CPC 23486 = CMW 31447 = CERC 1782, CPC 23487 = CMW 31448 = CERC 1783, CBS 136639 = CMW 31489 = CERC 1824; Guangxi Province, on leaf of Eucalyptus in plantation, Aug. 2009, X. Mou & R. Chang, CBS 136648 = CMW 37970 = CERC 1933.

**Notes:** Calonectria seminaria belongs to the C. candelabra species complex (Schoch et al. 1999, Lombard et al. 2010a; see Lombard et al. 2015), closely related to C. pauciramosa and C. polizzii. The macroconidia of C. seminaria are slightly smaller than those of C. pauciramosa, and larger than those of C. polizzii (Table 2).

**Calonectria sphaeropedunculata** L. Lombard, Crous & S.F. Chen, sp. nov. MycoBank MB809059. Fig. 17.

**Etymology:** Name refers to the sphaeropedunculate vesicles produced by this species.

Ascomata perithecial, solitary or in groups of two, orange, becoming orange-brown with age; in section, apex and body
orange, base red-brown, subglobose to ovoid, 470–575 μm high, 345–465 μm diam, body turning dark orange to red, and base dark red-brown in 3% KOH+; ascomatal wall rough, consisting of two thick-walled layers; outer layer of textura globulosa, 40–80 μm thick, cells becoming more compressed towards the inner layer of textura angularis, 14–21 μm thick, cells becoming thin-walled and hyaline towards the centre; outermost cells 22–36 × 14–22 μm, cells of inner layer 13–32 × 6–8 μm; ascomatal base up to 216 μm wide, consisting of dark red, angular cells, merging with an erumpent stroma; cells of the outer wall layer continuous with the pseudoparenchymatous cells of the erumpent stroma.

Asci 8-spored, clavate, 82–144 × 11–23 μm, tapering into a long thin stalk. Ascospores aggregated in the upper third of the ascus, hyaline, guttulate, fusoid with rounded ends, straight to slightly curved, 1-septate, not constricted at the septum, (31–) 33–40(–42) × 5–7(–8) μm (av. 37 × 6 μm). Homothallic. macroconidiophores consisting of a stipe bearing a penicillate arrangement of fertile branches, and a stipe extension terminating in a vesicle; stipe septate, hyaline, smooth, 62–183 × 7–12 μm; stipe extension septate, straight to flexuous, 152–253 μm long, 4–8 μm wide at the apical septum, terminating in a sphaero-opedunculate vesicle, 10–14 μm diam; lateral stipe extensions (90° to main axis) formed moderately. Conidiogenous apparatus 63–144 μm wide, and 40–111 μm long; primary branches aseptate or 1-septate, 18–36 × 4–10 μm; secondary branches aseptate, 11–29 × 5–9 μm; tertiary branches aseptate, 14–23 × 5–8 μm; quaternary and additional branches (–6) aseptate, 9–19 × 4–7 μm, each terminal branch producing 2–6 phialides; phialides doliform to reniform, hyaline, aseptate, 9–17 × 3–5 μm, apex with minute periclinal thickening and inconspicuous collarette. Macroconidia cylindrical, rounded at both ends, straight, (40–) 43–47(–49) × 4–6 μm (av. 46 × 5 μm), 1-septate, lacking a visible abscission scar, held in parallel cylindrical clusters by colourless slime. Mega- and microconidia not observed.

Culture characteristics: Colonies fast growing at 24 °C on MEA, producing abundant white to cinnamon aerial mycelium with profuse sporulation on the medium surface; reverse sienna to umber after 7 d; chlamydospores extensive throughout the medium forming microsclerotia.

Specimen examined: China, Guangxi Province, from soil collected in a Eucalyptus plantation, Mar. 2009, X. Zhou, G. Zhao & F. Han (holotype CBS H-21486, culture ex-type CBS 136081 = CPC 23484 = CMW 31390 = CERC 1725).
Calonectria sphaeropedunculata produces longer stipe extensions than those of C. kyotensis and C. pluriramosa, but shorter extensions than those of C. pseudokyotensis. The macroconidia of C. sphaeropedunculata are also smaller than those of C. kyotensis, C. pluriramosa and C. pseudokyotensis (Table 2).

Calonectria terrestris L. Lombard, Crous & S.F. Chen, sp. nov. MycoBank MB809060. Fig. 18.

Etymology: Name refers to the fact that this fungus was isolated from soil.

Ascomata not observed. Macroconidiophores consisting of a stipe bearing a penicillate arrangement of fertile branches, and a stipe extension terminating in a vesicle; stipe septate, hyaline, smooth, 35–185 × 6–10 μm; stipe extension septate, straight to flexuous, 147–228 μm long, 4–7 μm wide at the apical septum, terminating in an obpyriform to pyriform to broadly clavate vesicle, 5–12 μm diam. Conidiogenous apparatus 35–89 μm wide, and 35–102 μm long; primary branches aseptate, 21–35 × 5–8 μm; secondary branches aseptate, 15–27 × 4–7 μm; tertiary branches aseptate, 10–18 × 4–6 μm; quaternary branches aseptate, 9–14 × 3–6 μm, each terminal branch producing 2–4 phialides; phialides doliiform to reniform, hyaline, aseptate, 8–12 × 3–5 μm, apex with minute periclinal thickening and inconspicuous collarette. Macroconidia cylindrical, rounded at both ends, straight, (33–)36–40(–41) × (3–) 4–5 μm (av. 38.5 × 4.5 μm), 1-septate, lacking a visible
abscission scar, held in parallel cylindrical clusters by colourless slime. *Mega-* and *microconidia* not observed.

**Culture characteristics:** Colonies fast growing at 24 °C on MEA, producing abundant white to pale luteous aerial mycelium and sporulating profusely on the medium surface; reverse sienna toumber after 7 d; chlamydospores formed abundant throughout the medium, forming microsclerotia.

**Specimens examined:** China, Guangdong Province, from soil collected in a *Eucalyptus* plantation, Aug. 2009, X. Mou & R. Chang (holotype CBS H-21478, culture ex-type CBS 136642 = CMW 35180 = CERC 1856), CBS 136643 = CPC 23494 = CMW 35364 = CERC 1857, CBS 136644 = CPC 23496 = CMW 35366 = CERC 1858, CBS 136651 = CPC 23497 = CMW 37974 = CERC 1937 (CBS H-21479); Guangdong province, from leaf of *Eucalyptus*, Aug. 2009, X. Mou & R. Chang, CBS 136647 = CPC 23510 = CMW 35447 = CERC 1919; Guangxi Province, from soil collected in a *Eucalyptus* plantation, Aug. 2009, X. Mou & R. Chang, CBS 136653 = CPC 23518 = CMW 37980 = CERC 1943.

**Notes:** *Calonectria terrestris* can be distinguished from *C. cerciana* and *C. papillata* by its obpyriform to pyriform to broadly clavate vesicles rather than the fusiform to obpyriform vesicles of *C. cerciana*, and obpyriform to ellipsoidal vesicles with papillate apex of *C. papillata*. The macroconidia of *C. terrestris* are also slightly smaller than those of *C. cerciana* and *C. papillata* (Table 2).

### *Calonectria tetraramosa* L. Lombard, Crous & S.F. Chen, sp. nov. MycoBank MB809061. Fig. 19.

**Etymology:** Name refers to the four levels of fertile branches produced by this species.

Ascomata not observed. **Macroconidiophores** consisting of a stipe bearing a penicillate arrangement of fertile branches, and a stipe extension terminating in a vesicle; stipe septate, hyaline, smooth, 47–109 × 6–9 μm; stipe extension septate, straight to flexuous, 102–253 μm long, 3–6 μm wide at the apical septum, terminating in an obpyriform vesicle, 4–10 μm diam. **Conidiogenous apparatus** 54–95 μm wide, and 36–75 μm long; primary branches asceptate, 15–29 × 4–7 μm; secondary branches asceptate, 10–20 × 3–6 μm; tertiary branches asceptate, 9–15 × 3–6 μm; quaternary branches asceptate, 10–13 × 3–4 μm, each terminal branch producing 2–6 phialides; phialides elongate doliiform to reniform, hyaline, asceptate.
8–14 × 3–5 μm, apex with minute periclinal thickening and inconspicuous collarette. Macroconidia cylindrical, rounded at both ends, straight, (45–)46.5–49.5(–51) × (4–)4.5–5.5(–6) μm (av. 48 × 5 μm), 1-septate, lacking a visible abscission scar, held in parallel cylindrical clusters by colourless slime. Mega- and microconidia not observed.

Culture characteristics: Colonies fast growing at 24 °C on MEA, producing abundant white aerial mycelium and sporulating profusely on the medium surface; reverse amber to sepia-brown after 7 d; chlamydospores formed extensively in the media, forming microsclerotia.

Specimen examined: China, Guangdong Province, Zhanjiang, CERC nursery, on E. urophylla × E. grandis clone seedling leaf, Mar. 2009, G. Zhao (holotype CBS H-21477, living ex-type culture CBS 136635 = CPC 23489 = CMW 31474 = CERC 1809), CBS 136637 = CMW 37977 = CERC 1811.

Notes: Calonectria tetramosa is closely related to C. pauciramosa, C. polizzii and C. seminaria in the C. candelabra complex (Schoch et al. 1999, Lombard et al. 2010a). It can be distinguished from these three species by quaternary branches in the conidiogenous apparatus, which are not found in the other species. Furthermore, the macroconidia of C. tetramosa are slightly smaller than those of C. pauciramosa, larger than those of C. polizzii, but similar to those of C. seminaria. The stipe extensions of C. tetramosa are also longer than those of C. pauciramosa, C. polizzii and C. seminaria (Table 2).

Calonectria turangicola L. Lombard, Crous & S.F. Chen, sp. nov. MycoBank MB809062. Fig. 20.

Etymology: Name refers to the Chinese word for soil (Tūrāng), the substrate from which this fungus was first isolated.

Ascomata not observed. Macroconidiophores consisting of a stipe bearing a penicillate arrangement of fertile branches, and a stipe extension terminating in a vesicle; stipe septate, hyaline, smooth, 45–122 × 6–9 μm; stipe extension septate, straight to flexuous, 133–195 μm long, 4–6 μm wide at the apical septum, terminating in a sphaeropedunculate vesicle, 8–12 μm diam; lateral stipe extensions (90° to main axis) abundant. Conidiogenous apparatus 48–110 μm wide, and 35–86 μm long; primary branches aseptate, 16–30 × 4–7 μm; secondary branches aseptate, 10–18 × 3–6 μm; tertiary branches aseptate, 9–17 × 3–5 μm; quaternary and additional branches (–5) aseptate, 10–16 × 3–5 μm, each terminal branch producing 2–6 phialides; phialides doliiform to reniform, hyaline, aseptate, 8–16 × 3–7 μm, apex with minute periclinal thickening and inconspicuous collarette. Macroconidia cylindrical, rounded at both ends, straight, (40–)42–46(–47) × 3–5 μm (av. 44 × 4 μm), 1-septate, lacking a visible abscission scar, held in parallel cylindrical clusters by colourless slime. Mega- and microconidia not observed.

Culture characteristics: Colonies fast growing at 24 °C on MEA, producing abundant white to sienna aerial mycelium with profuse sporulation on the medium surface; reverse sienna to umber after 7 d; chlamydospores extensive throughout the medium forming microsclerotia.

Specimens examined: China, Guangxi Province, Fangchenggang, from soil collected in a Eucalyptus plantation, Mar. 2009, X. Mou & R. Chang, CBS 136093 = CMW 35410 = CERC 1901, CBS 136652 = CMW 37977 = CERC 1940; Hainan Province, from soil collected in a Eucalyptus plantation, Aug. 2009, X. Mou & S.F. Chen, CMW 35383 = CERC 1885.

Fig. 19. Calonectria tetramosa (ex-type CBS 136635); A–B. Macroconidiophores. C. Obpyriform vesicles. D. Macroconidia. E–H. Conidiogenous apparatus with conidiophore branches and doliiform to reniform phialides. Scale bars: A = 50 μm (apply to B); C = 10 μm (apply to D–H).
Note: The macroconidia of *C. turangicola* are slightly smaller than those of *C. hongkongensis* but larger than those of *C. lateralis* (Table 2).

**DISCUSSION**

A surprisingly large number of *Calonectria* species were collected from soils and *Eucalyptus* tissue in a relatively small area of southern China. Phylogenetic inference was used to define the species boundaries but these were in most cases also well-supported by morphological features. The 18 new species described in this study add to the eleven species previously recognised in the Southern provinces of China (Crous et al. 2004b, Lombard et al. 2010d, Chen et al. 2011d, Xu et al. 2012).

Most of the isolates obtained from *Eucalyptus* leaves displaying symptoms of CLB were identified as *C. pentaseptata*, which was recently described in the *C. reteaudii* complex from Vietnam (Crous et al. 2012), making this the first report of the fungus from China. *Calonectria pentaseptata* was collected in all three provinces sampled, including the sampled nursery surveyed, with a single isolate obtained from soil collected in Hainan Province. The collection data suggest that this fungus could be amongst the more important *Eucalyptus* leaf and shoot pathogens but this hypothesis will need testing experimentally.

*Calonectria microconidialis*, which was collected from *Eucalyptus* leaves in the nursery, resides in the *C. reteaudii* species complex, which now includes six species (Lombard et al. 2010d). The only other species in this complex known from China is *C. pseudoreteaudii* (Lombard et al. 2010d, Chen et al. 2011d). *Calonectria microconidialis* produces microconidiophores in culture, a characteristic shared with *C. reteaudii* and *C. pseudoreteaudii*, but distinguishing it from *C. pentaseptata*, *C. queenslandica* and *C. terre-reginae* (Lombard et al. 2010d, Crous et al. 2012). Species of the *C. reteaudii* complex are well-known causal agents of CLB in Australia, South America and Southeast Asia (Pilkethley 1976, Bolland et al. 1985, Sharma & Mohanan 1991, 1992, Booth et al. 2000, Kang et al. 2001a, Crous 2002, Rodas et al. 2005, Lombard et al. 2010d), but the pathogenicity of *C. microconidialis* and *C. pentaseptata* will need to be tested experimentally.

*Calonectria seminaria* and *C. tetraramosa* were found together with *C. microconidialis* in the nursery sampled in this study. These species represent new members of the
C. candelabra complex (Schoch et al. 1999, Lombard et al. 2015), which includes several well-known nursery pathogens (Schoch et al. 1999, Koike et al. 1999, Polizzi & Crous 1999, Polizzi 2000, Koike & Crous 2001, Polizzi & Catara 2001, Polizzi & Vitale 2001, Crous 2002, Polizzi et al. 2006, 2007, 2009, Vitale et al. 2009, Lombard et al. 2010a, d, Vitale et al. 2013, Guaraccia et al. 2014, Alfenas et al. 2015). The C. candelabra complex now includes 16 species (Schoch et al. 1999, Crous 2002, Lombard et al. 2010a, Alfenas et al. 2013a, 2015) and has the highest diversity of species found in South America (Schoch et al. 1999, 2001, Alfenas et al. 2015, see this volume). Although C. pauciramosa has been regarded as the dominant Eucalyptus nursery pathogen in previous studies (Schoch et al. 1999, Crous 2002, Lombard et al. 2010a), it was not isolated here. Although it has previously also been found in China (Lombard et al. 2010d) it is clearly not as common as it is elsewhere in the world such as in South America and Southern Africa.

This study included the description of three new species, C. foliicola, C. papillata and C. terrestris in the C. cylindrospora complex (Crous et al. 1993, Schoch et al. 1999, 2001, Lombard et al. 2010d, Alfenas et al. 2013b, 2015; see Lombard et al. 2015), which displays a similarly high level of species diversity in South America (Alfenas et al. 2013b, 2015). Calonectria papillata and C. terrestris are sibling species of C. cERCiana, but can be distinguished by their characteristic terminal vesicles and the morphology of their macroconidia. Calonectria papillata is also homothallic, a feature not known in C. cERCiana (Lombard et al. 2010d) nor in C. terrestris described in this study. Both C. papillata and C. terrestris were isolated from soils collected in Guangdong Province, but only a single isolate of C. terrestris was obtained from a Eucalyptus leaf collected from the same province. Calonectria foliicola, isolated from Eucalyptus leaves collected in Guangxi Province, is closely related to C. brasiliensis and C. sulawesiensis, but can be distinguished from those species based on its macroconidiphore morphology. Although some members of the C. cylindrospora complex are well-known pathogens (Crous 2002, Lombard et al. 2010c), nothing is known regarding the pathogenicity of C. foliicola, C. papillata and C. terrestris.

Most isolates of Calonectria spp. baited from soils in this study belonged to C. hongkongensis, a member of the C. kyotensis complex (Crous et al. 2004b) and the Sphaero-Naviculate Group (Lombard et al. 2010b). This fungus is characterised by its sphaeropecunulate terminal vesicles, a common feature for all members of the C. kyotensis complex (Crous et al. 2004b), and they also all have up to eight conidiophore branches (Crous et al. 2004b). Like C. hongkongensis, its sibling species C. turangicola and C. lateralis described here, were also isolated exclusively from soil. These species can be distinguish- ed from C. hongkongensis by having fewer conidiophore branches and from each other based on the morphology of their macroconidia.

Results of this study add 10 species to the C. kyotensis complex, which includes C. aconidialis, C. arbusta, C. expansa, C. guangxiensis, C. hainanensis, C. magnispora, C. parakyotensis, C. pseudokyotensis, C. pluriramosa and C. sphaeropecunulate. All 10 species were isolated from soils collected in all three provinces, although nothing is thus far known regarding their pathogenicity.

Calonectria aconidialis produced only its sexual morph in this study, despite many attempts to stimulate the production of conidiophores and conidia. However, sibling species such as C. arbusta and C. expansa formed both morphs in cultures derived from single conidia, and were thus homothallic. Calonectria parakyotensis, also a sibling species of C. aconidialis, failed to produce a sexual morph during this study. Different mating systems in species within related groups of Calonectria spp. are well-known and have been reported for members of the C. candelabra (Lombard et al. 2010a) and C. kyotensis complexes (Crous et al. 2004b). Calonectria aconidialis was found only in samples from the Hainan Province, and C. arbusta only in Guangxi, whereas both C. expansa and C. parakyotensis were found in soils collected in Guangdong and Guanxi provinces. Whether these species are restricted geographically would be interesting but more intensive and structured sampling would be needed to resolve this question.

Calonectria pseudokyotensis, C. pluriramosa and C. sphaeropecunulate are closely related to C. kyotensis and are easily distinguished from each other and C. kyotensis based on morphological features and phylogenetic inference. All of these novel species were isolated from soils collected in the Guangxi Province. Only C. sphaeropecunulate displayed a homothallic mating system, a feature shared with C. kyotensis (Crous 2002, Crous et al. 2004b), whereas C. pseudokyotensis and C. pluriramosa did not produce any sexual morphs in culture during this study.

The large ascospores and macroconidia of C. magnispora distinguish this novel species from the other members of the C. kyotensis complex. This species, along with C. guangxiensis, was isolated from soils collected in the Guangxi Province. Together with C. hainanensis, isolated from soil collected in the Hainan Province, these novel species readily formed their sexual morphs in culture, and are homothallic. Several isolates were also identified as C. chinensis based on phylogenetic inference and morphological features. This species, known only from China (Crous et al. 2004b), belongs to the C. kyotensis complex, and nothing is known regarding its ability to infect plants.

The greatest diversity of species found in this study came from baiting of soils collected in the Guangxi Province, followed by the Guangdong and Hainan Provinces. Of the 29 Calonectria species now known from China, 16 belong to the Sphaero-Naviculate Group, and 13 to the Prolate Group as defined by Lombard et al. (2010b). Ten species in the former group have a homothallic mating system and the remaining six species are more likely to be heterothallic because single conidial isolates mated in culture did not produce ascocarps. Interestingly, all homothallic species originated exclusively from a soil habitat, while those thought to be heterothallic were from both soil and plant material. It is possible that homothallism in these Calo-nectria species represents an adaptation to the soil environment where only short-distance spread is required, as ascospores are extremely susceptible to desiccation (Rowe & Beute 1975). The majority of the putative heterothallic Calonectria species in this study were isolated from leaves of different Eucalyptus clones displaying CLB symptoms. Since heterothallism results in sexual outcrossing and the generation of genetic diversity (Billiard et al. 2012, Heitman et al. 2013), such a mating system would be beneficial to fungi that infect plants where sexual outcrossing would facilitate the process of overcoming host resistance.

To better understand the genetic variation within these homothallic and putative heterothallic Calonectria species more knowledge of the population structure of these species is
required. Relatively few studies have focused on the population dynamics of Calonectria species (Wright et al. 2006, 2007, 2010), and therefore limited knowledge is available on the population structure, distribution of genetic diversity, gene flow, centres of origin and the role of mating strategies for these fungi. Population studies on these fungi, especially those associated with CLB in China would better facilitate our understanding of the epidemiology, and in turn, the management of CLB in Eucalyptus plantations in China. These studies would also allow the prediction of efficacy of host plant resistance to these fungi, necessary for the establishment of future commercial plantations in China.

Although several Calonectria species were isolated from Eucalyptus leaves displaying symptoms of CLB in this study, relatively little is known about their pathogenicity, and their roles as potential pathogens can only be assumed based on the symptoms they are associated with. Therefore, pathogenicity tests need to be done experimentally to determine whether these species are pathogenic to Eucalyptus and if they are host specific. These studies would help identify which Calonectria species are important to commercial Eucalyptus forestry in China. The high diversity of Calonectria species in a relatively small area of southern China, and especially in virgin soils, implies that more Calonectria species remain to be discovered as sampling is extended to more provinces in China, which would also have to be tested as possible threats to Eucalyptus production in China.

ACKNOWLEDGEMENTS

This study was initiated through the bilateral agreement between the Governments of South Africa and China, and we are grateful for the funding via projects 2010JKCY015-03 (Forestry Science and Technology Innovation Project of Guangdong Province of China), 2012DFG31830 (International Science & Technology Cooperation Program of China), 31400546 (National Natural Science Foundation of China) and the NWO Joint Chinese Thematic Research Programme – Joint Research Projects 2012 ALW file number 833.13.005 “Building the fungal quarantine & quality barcode of life database to ensure plant health”. We also appreciate the financial support of members of the Tree Protection Co-operative Programme (TCPWC). We thank Arien van Iperen, Tit Merx and Dr Seonju Marincowitz for their invaluable assistance with cultures.

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