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

L. Lombard[1], S.F. Chen[2,3*], X. Mou[2,3], X.D. Zhou[2,3], P.W. Crous[1,2,4], and M.J. Wingfield[2]

[1]*CBS-KNAW Fungal Biodiversity Centre, Uppsalalaan 8, 3584 CT Utrecht, The Netherlands;* [2]*Department of Microbiology and Plant Pathology, Tree Protection Co-operative Programme, Forestry and Agricultural Biotechnology Institute, University of Pretoria, Pretoria 0002, South Africa;* [3]*China Eucalypt Research Centre (CERC), Chinese Academy of Forestry (CAF), ZhanJiang 524022, GuangDong Province, China;* [4]*Microbiology, Department of Biology, Utrecht University, Padualaan 8, 3584 CH Utrecht, The Netherlands*

*Correspondence: S.F. Chen. shuaifei.chen@gmail.com

**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 aconidialis* L. Lombard, Crous & S.F. Chen, *C. arbusta* L. Lombard, Crous & S.F. Chen, *C. expansa* L. Lombard, Crous & S.F. Chen, *C. foliicola* L. Lombard, Crous & S.F. Chen, *C. guangxiensis* L. Lombard, Crous & S.F. Chen, *C. hainanensis* L. Lombard, Crous & S.F. Chen, *C. lateralis* 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. pluriramosa* 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. terrestris* L. Lombard, Crous & S.F. Chen, *C. tetraramosa* L. Lombard, Crous & S.F. Chen, *C. turangicola* L. Lombard, Crous & S.F. Chen.

Published online ; http://dx.doi.org/10.1016/j.simyco.2014.11.003. Hard copy: xxx.

## 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), *Celoporthe* (Chen *et al.* 2011b), *Ceratocystis* (Chen *et al.* 2013), *Chrysoporthe* (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. cerciana* and *C. pseudoreteaudii*, and the well-known *Eucalyptus* nursery pathogen, *C. pauciramosa* (Koike *et al.* 1999, Polizzi & 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. pseudocolhounii*, 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

Peer review under responsibility of CBS-KNAW Fungal Biodiversity Centre.





Please cite this article in press as: Lombard L, et al., New species, hyper-diversity and potential importance of *Calonectria* spp. from *Eucalyptus* in South China, Studies in Mycology (2015), http://dx.doi.org/10.1016/j.simyco.2014.11.003

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 conidial cultures were prepared on MEA and these are maintained in the culture collection (CMW) of the Forestry and Agricultural Biotechnology Institute (FABI), University of Pretoria, Pretoria, South Africa, the CBS-KNAW Fungal Biodiversity Centre (CBS), Utrecht, The Netherlands, the research collection of P.W. Crous (CPC) maintained at CBS, and the culture collection of CERC, Zhanjiang, Guangdong Province, China.

### 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).



Please cite this article in press as: Lombard L, et al., New species, hyper-diversity and potential importance of *Calonectria* spp. from *Eucalyptus* in South China, Studies in Mycology (2015), http://dx.doi.org/10.1016/j.simyco.2014.11.003

**Table 1.** *Calonectria* spp. used in phylogenetic analyses.

| Species | Isolate nr.[1] | Substrate | Locality | GenBank Accession no.[2] | | | |
|---|---|---|---|---|---|---|---|
| | | | | *tub2* | *cmdA* | *his3* | *tef1* |
| *Calonectria aconidialis* | **CBS 136086**; CMW 35174; CERC 1850 | Soil in *Eucalyptus* plantation | Hainan, China | – | *KJ463017* | *KJ463133* | *KJ462785* |
| | CBS 136091; CMW 35384; CERC1886 | Soil in *Eucalyptus* plantation | Hainan, China | – | – | *KJ463134* | *KJ462786* |
| *C. arbusta* | **CBS 136079**; CMW 31370; CERC1705 | Soil in *Eucalyptus* plantation | Guangxi, China | *KJ462904* | *KJ463018* | *KJ463135* | *KJ462787* |
| | CBS 136098; CPC 23519; CMW37981; 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* |
| | CPC 23483; CMW 31371; CERC 1706 | Soil in *Eucalyptus* plantation | Guangxi, China | *KJ462906* | *KJ463021* | *KJ463138* | *KJ462790* |
| | CMW 31367; CERC 1702 | Soil in *Eucalyptus* plantation | Fangchenggang, Guangxi, China | *KJ462907* | *KJ463022* | *KJ463139* | *KJ462791* |
| | CMW 31368; CERC 1703 | Soil in *Eucalyptus* plantation | Guangxi, China | *KJ462908* | *KJ463023* | *KJ463140* | *KJ462792* |
| *C. asiatica* | 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 230.51**; CPC 2390 | *Anacardium* sp. | Brazil | GQ267241 | GQ267421 | GQ267259 | GQ267328 |
| | CBS 114257; CPC 1944 | *Eucalyptus* leaf | Brazil | GQ267242 | GQ267422 | GQ267260 | GQ267329 |
| *C. brassiana* | **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 | | Canada | AF348212 | AY725743 | AF348228 | GQ267297 |
| *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; CMW 25290 | *Eucalyptus* cutting | Zhanjiang, China | FJ918511 | GQ267370 | FJ918529 | FJ918560 |
| *C. chinensis* | CBS 112744; CPC 4104 | Soil | Hong Kong, China | AY725618 | AY725746 | AY725660 | AY725709 |
| | **CBS 114827**; CPC 4101 | Soil | Hong Kong, China | AY725619 | AY725747 | AY725661 | AY725710 |
| | CBS 136082; CMW 35367; CERC 1871 | Soil in *Eucalyptus* plantation | Guangdong, China | *KJ462909* | *KJ463024* | *KJ463141* | *KJ462793* |
| | CBS 136083; CMW 35179; CERC 1855 | Soil in *Eucalyptus* plantation | Guangdong | *KJ462910* | *KJ463025* | *KJ463142* | *KJ462794* |
| | CBS 136088; CMW 35376; CERC 1878 | Soil in *Eucalyptus* plantation | Hainan, China | *KJ462911* | *KJ463026* | *KJ463143* | *KJ462795* |
| | CBS 136090; CMW 35379; CERC 1881 | Soil in *Eucalyptus* plantation | Hainan, China | *KJ462912* | *KJ463027* | *KJ463144* | *KJ462796* |
| *C. colhounii* | CBS 293.79 | *Camellia sinensis* | Bandung, Indonesia | DQ190564 | GQ267373 | DQ190639 | GQ267301 |
| | CBS 114704 | *Arachis pintoi* | Australia | DQ190563 | GQ267372 | DQ190638 | GQ267300 |
| *C. colombiensis* | **CBS 112220**; CPC 723 | Soil | La Selva, Brazil | GQ267207 | AY725748 | AY725662 | AY725711 |
| | CBS 112221; CPC 724 | *Eucalyptus grandis* | 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 | GQ267302 |
| *C. cylindrospora* | CBS 110666; CPC 496 | | USA | FJ918509 | GQ267423 | FJ918527 | FJ918557 |
| | CBS 119670; CPC 12766 | *Pistacia lentiscus* | Italy | DQ521600 | – | DQ521602 | GQ421797 |
| *C. eucalypticola* | CBS 134846 | *Eucalyptus* leaf | Eunápolis, Bahia, Brazil | KM395963 | KM396050 | KM396133 | KM395876 |
| | **CBS 134847** | *Eucalyptus* seedling | Santa Bárbara, Minas Gerais, Brazil | KM395964 | KM396051 | KM396134 | KM395877 |
| *C. expansa* | CBS 136078; CMW 31441; CERC 1776 | Soil in *Eucalyptus* plantation | Guangdong, China | *KJ462913* | *KJ463028* | *KJ463145* | *KJ462797* |

(*continued on next page*)



Please cite this article in press as: Lombard L, et al., New species, hyper-diversity and potential importance of *Calonectria* spp. from *Eucalyptus* in South China, Studies in Mycology (2015), http://dx.doi.org/10.1016/j.simyco.2014.11.003

**Table 1.** (Continued).

| Species | Isolate nr.[1] | Substrate | Locality | GenBank Accession no.[2] | | | |
|---|---|---|---|---|---|---|---|
| | | | | *tub2* | *cmdA* | *his3* | *tef1* |
| | **CBS 136247**; CMW 31392; CERC 1727 | Soil in *Eucalyptus* plantation | Guangxi, China | *KJ462914* | *KJ463029* | *KJ463146* | *KJ462798* |
| | CMW 31413; CERC 1748 | Soil in *Eucalyptus* plantation | Guangxi, China | *KJ462915* | *KJ463030* | *KJ463147* | *KJ462799* |
| *C. foliicola* | **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* | CBS 127200; CMW 27254 | *E. grandis* | Fujian, China | HQ285791 | – | HQ285805 | HQ285819 |
| | **CBS 127201**; CMW 27257 | *E. grandis* | Fujian, China | HQ285792 | – | HQ285806 | HQ285820 |
| *C. glaeboicola* | **CBS 134852** | Soil | Martinho Campos, Minas Gerais, Brazil | KM395966 | KM396053 | KM396136 | KM395879 |
| | CBS 134853 | Soil | Bico do Papagaio, Tocantins, Brazil | KM395967 | KM396054 | KM396137 | KM395880 |
| *C. guangxiensis* | **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. hainanensis* | **CBS 136248**; CMW 35187; CERC 1863 | Soil in *Eucalyptus* plantation | Hainan, China | – | *KJ463036* | *KJ463152* | *KJ462805* |
| *C. hawksworthii* | **CBS 111870**; CPC 2405; MUCL 30866 | *Nelumbo nucifera* | Mauritius | AF333407 | GQ267386 | DQ190649 | FJ918558 |
| *C. hodgesii* | **CBS 133609**; LPF 245 | *Anadenanthera peregrina* | Viçosa, Brazil | KC491228 | KC491222 | – | KC491225 |
| | CBS 133610; LPF 261 | *Azadirachta indica* | Viçosa, Brazil | KC491229 | KC491223 | – | KC491226 |
| *C. hongkongensis* | CBS 114711; CPC 686 | Soil | Hong Kong, China | AY725621 | AY725754 | AY725666 | AY725716 |
| | **CBS 114828**; CPC 4670 | Soil | Hong Kong, China | AY725622 | AY725755 | AY725667 | AY725717 |
| | CBS 136080; CMW 31443; CERC 1778 | Soil in *Eucalyptus* plantation | Fangchenggang, Guangxi, China | *KJ462921* | *KJ463037* | *KJ463153* | *KJ462806* |
| | CBS 136246; CMW 31374; CERC 1709 | Soil in *Eucalyptus* plantation | Fangchenggang, Guangxi, China | *KJ462922* | *KJ463038* | *KJ463154* | *KJ462807* |
| | CPC 23478; CMW 31438; CERC 1773 | Soil in *Eucalyptus* plantation | Shiling, Zhanjiang, Guangdong, China | *KJ462923* | *KJ463039* | *KJ463155* | *KJ462808* |
| | CPC 23480; CMW 31414; CERC 1749 | Soil in *Eucalyptus* plantation | Fangchenggang, Guangxi, China | *KJ462924* | *KJ463040* | *KJ463156* | *KJ462809* |
| | CPC 23499; CMW 35175; CERC 1851 | Soil in *Eucalyptus* plantation | Hainan, China | *KJ462925* | *KJ463041* | *KJ463157* | *KJ462810* |
| | CPC 23877; CERC 1932 | Soil in *Eucalyptus* plantation | Hainan, China | *KJ462926* | *KJ463042* | *KJ463158* | *KJ462811* |
| | CPC 23878; CMW 37973; CERC 1936 | Soil in *Eucalyptus* plantation | Guangdong, China | *KJ462927* | *KJ463043* | *KJ463159* | *KJ462812* |
| | CMW 31375; CERC 1710 | Soil in *Eucalyptus* plantation | Fangchenggang, Guangxi, China | *KJ462928* | *KJ463044* | *KJ463160* | *KJ462813* |
| | CMW 31377; CERC 1712 | Soil in *Eucalyptus* plantation | Fangchenggang, Guangxi, China | *KJ462929* | *KJ463045* | *KJ463161* | *KJ462814* |
| | CMW 31382; CERC 1717 | Soil in *Eucalyptus* plantation | Fangchenggang, Guangxi, China | *KJ462930* | *KJ463046* | *KJ463162* | *KJ462815* |
| | CMW 31383; CERC 1718 | Soil in *Eucalyptus* plantation | Fangchenggang, Guangxi, China | *KJ462931* | *KJ463047* | *KJ463163* | *KJ462816* |
| | CMW 31384; CERC 1719 | Soil in *Eucalyptus* plantation | Fangchenggang, Guangxi, China | *KJ462932* | *KJ463048* | *KJ463164* | *KJ462817* |
| | CMW 31385; CERC 1720 | Soil in *Eucalyptus* plantation | Fangchenggang, Guangxi, China | *KJ462933* | *KJ463049* | *KJ463165* | *KJ462818* |
| | CMW 31387; CERC 1722 | Soil in *Eucalyptus* plantation | Fangchenggang, Guangxi, China | *KJ462934* | *KJ463050* | *KJ463166* | *KJ462819* |



Please cite this article in press as: Lombard L, et al., New species, hyper-diversity and potential importance of *Calonectria* spp. from *Eucalyptus* in South China, Studies in Mycology (2015), http://dx.doi.org/10.1016/j.simyco.2014.11.003

**Table 1.** (Continued).

| 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; CERC1739 | Soil in *Eucalyptus* plantation | Shiling, Zhanjiang, Guangdong, China | *KJ462939* | *KJ463054* | *KJ463171* | *KJ462824* |
| | CMW 31432; CERC1767 | Soil in *Eucalyptus* plantation | Shiling, Zhanjiang, Guangdong, China | *KJ462940* | *KJ463055* | *KJ463172* | *KJ462825* |
| | CMW 31433; CERC1768 | Soil in *Eucalyptus* plantation | Shiling, Zhanjiang, Guangdong, China | *KJ462941* | *KJ463056* | *KJ463173* | *KJ462826* |
| | CMW 31434; CERC1769 | Soil in *Eucalyptus* plantation | Shiling, Zhanjiang, Guangdong, China | *KJ462942* | *KJ463057* | *KJ463174* | *KJ462827* |
| | CMW 31442; CERC1777 | Soil in *Eucalyptus* plantation | Fangchenggang, Guangxi, China | *KJ462943* | *KJ463058* | *KJ463175* | *KJ462828* |
| | CMW 35186; CERC1862 | Soil in *Eucalyptus* plantation | Hainan, China | *KJ462944* | *KJ463059* | *KJ463176* | *KJ462829* |
| | CMW 35188; CERC1864 | Soil in *Eucalyptus* plantation | Hainan, China | *KJ462945* | *KJ463060* | *KJ463177* | *KJ462830* |
| | CMW 35190; CERC1865 | Soil in *Eucalyptus* plantation | Guangxi, China | *KJ462946* | *KJ463061* | *KJ463178* | *KJ462831* |
| | CMW 35192; CERC1867 | Soil in *Eucalyptus* plantation | Guangxi, China | *KJ462947* | *KJ463062* | – | *KJ462832* |
| | CMW 35371; CERC1874 | Soil in *Eucalyptus* plantation | Guangdong, China | *KJ462948* | *KJ463063* | *KJ463179* | *KJ462833* |
| | CMW 35378; CERC1880 | Soil in *Eucalyptus* plantation | Hainan, China | *KJ462949* | *KJ463064* | *KJ463180* | *KJ462834* |
| | CMW 35381; CERC1883 | Soil in *Eucalyptus* plantation | Hainan, China | *KJ462950* | *KJ463065* | *KJ463181* | *KJ462835* |
| | CMW 35401; CERC1892 | Soil in *Eucalyptus* plantation | Guangxi, China | *KJ462951* | *KJ463066* | *KJ463182* | *KJ462836* |
| | CMW 35404; CERC1895 | Soil in *Eucalyptus* plantation | Guangxi, China | *KJ462952* | *KJ463067* | *KJ463183* | *KJ462837* |
| | CMW 35414; CERC1905 | Soil in *Eucalyptus* plantation | Guangxi, China | *KJ462953* | *KJ463068* | *KJ463184* | *KJ462838* |
| | CMW 36270; CERC1928 | Soil in *Eucalyptus* plantation | Hainan, China | *KJ462954* | *KJ463069* | *KJ463185* | *KJ462839* |
| *C. ilicicola* | **CBS 190.50**; CMW 30998; IMI 299389 | *Solanum tuberosum* | Bogor, Indonesia | AY725631 | AY725764 | AY725676 | AY725726 |
| | CBS 115897; CPC 493; UFV 108 | *Anacardium* sp. | Brazil | AY725647 | GQ267403 | GQ267256 | AY725729 |
| *C. indonesiae* | **CBS 112823**; CPC 4508 | Soil | Warambunga, Indonesia | AY725623 | AY725756 | AY725668 | AY725718 |
| | CBS 112840; CPC 4554 | *Syzygium aromaticum* | Indonesia | AY725625 | AY725758 | AY725670 | AY725720 |
| *C. insularis* | **CBS 114558**; CPC 768 | Soil | Tamatave, Madagascar | AF210861 | GQ267389 | FJ918526 | FJ918556 |
| | CBS 114559; CPC 954 | Soil | Tamatave, Madagascar | AF210862 | GQ267390 | FJ918525 | FJ918555 |
| *C. kyotensis* | CBS 413.67; CPC 2391; IMI 299577 | *Paphiopedilum callosum* | Celle, Germany | GQ267208 | GQ267379 | GQ267248 | GQ267307 |
| | CBS 170.77; IMI 299388 | *Idesia polycarpa* | Auckland, New Zealand | GQ267209 | GQ267380 | GQ267249 | GQ267308 |
| *C. lateralis* | **CBS 136629**; CMW 31412; CERC 1747 | Soil in *Eucalyptus* plantation | Fangchenggang, Guangxi, China | *KJ462955* | *KJ463070* | *KJ463186* | *KJ462840* |
| *C. leucothoes* | **CBS 109166**; CPC 2385; ATCC 64824 | *Leucothoe axillaris* | Gainsville, Florida, USA | FJ918508 | GQ267392 | FJ918523 | FJ918553 |
| *C. magnispora* | **CBS 136249**; CMW 35184; CERC 1860 | Soil in *Eucalyptus* plantation | Guangxi, China | *KJ462956* | *KJ463071* | *KJ463187* | *KJ462841* |
| *C. malesiana* | CBS 112710; CPC 3899 | Leaf litter | Thailand | AY725626 | AY725759 | AY725671 | AY725721 |
| | **CBS 112752**; CPC 4223 | Soil | Sumatra, Indonesia | AY725627 | AY725760 | AY725672 | AY725722 |
| *C. maranhensis* | **CBS 134811** | *Eucalyptus* sp. | Açailândia, Maranhão, Brazil | KM395948 | KM396035 | KM396118 | KM395861 |
| | CBS 134812 | *Eucalyptus* sp. | Açailândia, Maranhão, Brazil | KM395949 | KM396036 | KM396119 | KM395862 |

(*continued on next page*)



Please cite this article in press as: Lombard L, et al., New species, hyper-diversity and potential importance of *Calonectria* spp. from *Eucalyptus* in South China, Studies in Mycology (2015), http://dx.doi.org/10.1016/j.simyco.2014.11.003

**Table 1.** (Continued).

| Species | Isolate nr.[1] | Substrate | Locality | GenBank Accession no.[2] | | | |
|---|---|---|---|---|---|---|---|
| | | | | *tub2* | *cmdA* | *his3* | *tef1* |
| | CBS 134858 | Soil | Urbano Santos, Maranhão, Brazil | KM395951 | KM396038 | KM396121 | KM395864 |
| | CBS 134829 | Soil | Urbano Santos, Maranhão, Brazil | KM395952 | KM396039 | KM396122 | KM395865 |
| *C. metrosideri* | **CBS 133604**; LPF 103 | *Metrosideros polymorpha* | Viçosa, Brazil | KC294314 | KC294305 | KC294308 | KC294311 |
| | CBS 133605; LPF 104 | *M. polymorpha* | Viçosa, Brazil | KC294315 | KC294306 | KC294309 | KC294312 |
| *C. microconidialis* | CBS 136633; CMW 31471; CERC 1806 | *E. urophylla* × *E. grandis* clone seedling leaf | CERC Nursery, Zhanjiang, Guangdong, China | *KJ462957* | *KJ463072* | *KJ463188* | *KJ462842* |
| | CBS 136634; CMW 31473; CERC 1808 | *E. urophylla* × *E. grandis* clone seedling leaf | CERC Nursery, Zhanjiang, Guangdong, China | *KJ462958* | *KJ463073* | *KJ463189* | *KJ462843* |
| | CBS 136636; CMW 31475; CERC 1810 | *E. urophylla* × *E. grandis* clone seedling leaf | CERC Nursery, Zhanjiang, Guangdong, China | *KJ462959* | *KJ463074* | *KJ463190* | *KJ462844* |
| | **CBS 136638**; CMW 31487; CERC 1822 | *E. urophylla* × *E. grandis* clone seedling leaf | CERC Nursery, Zhanjiang, Guangdong, China | *KJ462960* | *KJ463075* | *KJ463191* | *KJ462845* |
| | CBS 136640; CMW 31492; CERC 1827 | *E. urophylla* × *E. grandis* clone seedling leaf | CERC Nursery, Zhanjiang, Guangdong, China | *KJ462961* | *KJ463076* | *KJ463192* | *KJ462846* |
| *C. nemuricola* | **CBS 134837** | Soil | Araponga, Minas Gerais, Brazil | KM395979 | KM396066 | KM396149 | KM395892 |
| | CBS 134838 | Soil | Araponga, Minas Gerais, Brazil | KM395980 | KM396067 | KM396150 | KM395893 |
| *C. nymphaeae* | **CBS 131802**; HGUP 100003 | *Nymphaea tetragona* | Guizhou, China | JN984864 | – | – | KC555273 |
| *C. pacifica* | **CBS 109063**; CPC 2534; IMI 354528 | *Araucaria heterophylla* | Hawaii, USA | GQ267213 | AY725762 | GQ267255 | AY725724 |
| | CBS 114038; CPC 10717 | *Ipomoea aquatica* | Auckland, New Zealand | AY725630 | GQ267402 | AY725675 | GQ267320 |
| *C. papillata* | CBS 136084; CMW 35165; CERC 1841 | Soil in *Eucalyptus* plantation | Guangdong, China | *KJ462962* | *KJ463077* | *KJ463193* | *KJ462847* |
| | CBS 136096; CMW 37972; CERC 1935 | Soil in *Eucalyptus* plantation | Guangdong, China | *KJ462963* | *KJ463078* | *KJ463194* | *KJ462848* |
| | **CBS 136097**; CMW 37976; CERC 1939 | Soil in *Eucalyptus* plantation | Guangdong, China | *KJ462964* | *KJ463079* | *KJ463195* | *KJ462849* |
| | CBS 136251; CMW 37971; CERC 1934 | Soil in *Eucalyptus* plantation | Guangxi, China | *KJ462965* | *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 | FJ918514 | GQ267405 | FJ918531 | FJ918565 |
| | CMW 30823 | *E. grandis* | South Africa | FJ918515 | GQ280404 | FJ918532 | FJ918566 |
| *C. pentaseptata* | **CBS 133349** | *Eucalyptus* hybrid | Bavi, Hanoi, Vietnam | JX855942 | – | JX855946 | JX855958 |
| | 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* |



Please cite this article in press as: Lombard L, et al., New species, hyper-diversity and potential importance of *Calonectria* spp. from *Eucalyptus* in South China, Studies in Mycology (2015), http://dx.doi.org/10.1016/j.simyco.2014.11.003

**Table 1.** (Continued).

| Species | Isolate nr.[1] | Substrate | Locality | GenBank Accession no.[2] | | | |
|---|---|---|---|---|---|---|---|
| | | | | *tub2* | *cmdA* | *his3* | *tef1* |
| | CMW 31340; CERC 1675 | *E. urophylla* × *E. grandis* clone seedling leaf | CERC Nursery, Zhanjiang, Guangdong, China | *KJ462973* | *KJ463090* | *KJ463206* | *KJ462860* |
| | CMW 31343; CERC 1678 | *E. urophylla* × *E. grandis* clone seedling leaf | CERC Nursery, Zhanjiang, Guangdong, China | *KJ462974* | *KJ463091* | *KJ463207* | *KJ462861* |
| | CMW 31344; CERC 1679 | *E. urophylla* × *E. grandis* clone seedling leaf | CERC Nursery, Zhanjiang, Guangdong, China | *KJ462975* | *KJ463092* | *KJ463208* | *KJ462862* |
| | CMW 31345; CERC 1680 | *E. urophylla* × *E. grandis* clone seedling leaf | CERC Nursery, Zhanjiang, Guangdong, China | *KJ462976* | *KJ463093* | *KJ463209* | *KJ462863* |
| | CMW 31346; CERC 1681 | *E. urophylla* × *E. grandis* clone seedling leaf | CERC Nursery, Zhanjiang, Guangdong, China | *KJ462977* | *KJ463094* | *KJ463210* | *KJ462864* |
| | CMW 31347; CERC 1682 | *E. urophylla* × *E. grandis* clone seedling leaf | CERC Nursery, Zhanjiang, Guangdong, China | *KJ462978* | *KJ463095* | *KJ463211* | *KJ462865* |
| | CMW 31348; CERC 1683 | *E. urophylla* × *E. grandis* clone seedling leaf | CERC Nursery, Zhanjiang, Guangdong, China | *KJ462979* | *KJ463096* | *KJ463212* | *KJ462866* |
| | CMW 31355; CERC 1690 | *E. urophylla* × *E. grandis* leaf | Hepu, Guangxi, China | *KJ462980* | *KJ463097* | *KJ463213* | *KJ462867* |
| | CMW 31356; CERC 1691 | *E. urophylla* × *E. grandis* leaf | Hepu, Guangxi, China | *KJ462981* | *KJ463098* | *KJ463214* | *KJ462868* |
| | CMW 31357; CERC 1692 | *E. urophylla* × *E. grandis* leaf | Hepu, Guangxi, China | *KJ462982* | *KJ463099* | *KJ463215* | *KJ462869* |
| | CMW 31358; CERC 1693 | *E. urophylla* × *E. grandis* leaf | Hepu, Guangxi, China | *KJ462983* | *KJ463100* | *KJ463216* | *KJ462870* |
| | CMW 31359; CERC 1694 | *E. urophylla* × *E. grandis* leaf | Hepu, Guangxi, China | *KJ462984* | *KJ463101* | *KJ463217* | *KJ462871* |
| | CMW 31363; CERC 1698 | *E. urophylla* × *E. grandis* leaf | Hepu, Guangxi, China | *KJ462985* | *KJ463102* | *KJ463218* | *KJ462872* |
| | CMW 31422; CERC 1757 | *Eucalyptus* clone U6 leaf | Shiling, Zhanjiang, Guangdong, China | *KJ462986* | *KJ463103* | *KJ463219* | *KJ462873* |
| | CMW 31497; CERC 1832 | *E. urophylla* × *E. grandis* clone seedling leaf | CERC Nursery, Zhanjiang, Guangdong, China | *KJ462987* | *KJ463104* | *KJ463220* | *KJ462874* |
| | CMW 35385; CERC 1887 | Soil in *Eucalyptus* plantation | Hainan, China | *KJ462988* | *KJ463105* | *KJ463221* | *KJ462875* |
| | CMW 35437; CERC 1909 | *Eucalyptus* leaf | Guangdong, China | *KJ462989* | *KJ463106* | *KJ463222* | *KJ462876* |
| | CMW 35442; CERC 1914 | *Eucalyptus* leaf | Guangdong, China | *KJ462990* | *KJ463107* | *KJ463223* | *KJ462877* |
| | CMW 35452; CERC 1924 | *Eucalyptus* leaf | Guangdong, China | *KJ462991* | *KJ463108* | *KJ463224* | *KJ462878* |
| | CMW 35453; CERC 1925 | *Eucalyptus* leaf | Guangdong, China | *KJ462992* | *KJ463109* | *KJ463225* | *KJ462879* |
| | CMW 35454; CERC 1926 | *Eucalyptus* leaf | Guangdong, China | *KJ462993* | *KJ463110* | *KJ463226* | *KJ462880* |
| *C. piauiensis* | **CBS 134850** | Soil | Teresina, Piauí, Brazil | KM395973 | KM396060 | KM396143 | KM395886 |
| | CBS 134851 | Soil | Teresina, Piauí, Brazil | KM395974 | KM396061 | KM396144 | KM395887 |
| *C. pluriramosa* | **CBS 136976**; CMW 31440; CERC 1775 | Soil in *Eucalyptus* plantation | Fangchenggang, Guangxi, China | *KJ462995* | *KJ463112* | *KJ463228* | *KJ462882* |
| *C. polizzi* | CBS 125270; CMW 7804 | *Callistemon citrinus* | Messina, Sicily, Italy | FJ972417 | GQ267461 | FJ972436 | FJ972486 |
| | CBS 125271; CMW 10151 | *Arbustus unedo* | Catania, Sicily, Italy | FJ972418 | GQ267462 | FJ972437 | FJ972487 |
| *C. propaginicola* | **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. pseudocolhounii* | **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. pseudokyotensis* | **CBS 137332**; CMW 31439; CERC 1774 | Soil in *Eucalyptus* plantation | Fangchenggang, Guangxi, China | *KJ462994* | *KJ463111* | *KJ463227* | *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*)



Please cite this article in press as: Lombard L, et al., New species, hyper-diversity and potential importance of *Calonectria* spp. from *Eucalyptus* in South China, Studies in Mycology (2015), http://dx.doi.org/10.1016/j.simyco.2014.11.003

**Table 1.** (Continued).

| Species | Isolate nr.[1] | Substrate | Locality | GenBank Accession no.[2] | | | |
|---|---|---|---|---|---|---|---|
| | | | | *tub2* | *cmdA* | *his3* | *tef1* |
| *C. pseudoreteaudii* | **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. pseudospathulata* | 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. pellita* | Australia | AF389834 | GQ267416 | DQ190667 | FJ918544 |
| *C. reteaudii* | **CBS 112143**; CPC 3200 | *E. camaldulensis* | Vietnam | GQ240642 | GQ267418 | DQ190660 | FJ918536 |
| | CBS 112144; CPC 3201 | *E. camaldulensis* | Vietnam | AF389833 | GQ267417 | DQ190661 | FJ918537 |
| *C. seminaria* | CBS 136630; CMW 31446; CERC 1781 | *E. urophylla* × *E. grandis* clone seedling leaf | CERC Nursery, Zhanjiang, Guangdong, China | *KJ462996* | *KJ463113* | *KJ463229* | *KJ462883* |
| | CBS 136631; CMW 31449; CERC 1784 | *E. urophylla* × *E. grandis* clone seedling leaf | CERC Nursery, Zhanjiang, Guangdong, China | *KJ462997* | *KJ463114* | *KJ463230* | *KJ462884* |
| | **CBS 136632**; CMW 31450; CERC 1785 | *E. urophylla* × *E. grandis* clone seedling leaf | CERC Nursery, Zhanjiang, Guangdong, China | *KJ462998* | *KJ463115* | *KJ463231* | *KJ462885* |
| | CBS 136639; CMW 31489; CERC 1824 | *E. urophylla* × *E. grandis* clone seedling leaf | CERC Nursery, Zhanjiang, Guangdong, China | *KJ462999* | *KJ463116* | *KJ463232* | *KJ462886* |
| | CBS 136648; CMW 37970; CERC 1933 | *Eucalyptus* leaf | Guangxi, China | *KJ463000* | *KJ463117* | *KJ463233* | *KJ462887* |
| | CPC 23486; CMW 31447; CERC 1782 | *E. urophylla* × *E. grandis* clone seedling leaf | CERC Nursery, Zhanjiang, Guangdong, China | *KJ463001* | *KJ463118* | *KJ463234* | *KJ462888* |
| | CPC 23487; CMW 31448; CERC 1783 | *E. urophylla* × *E. grandis* clone seedling leaf | CERC Nursery, Zhanjiang, Guangdong, China | *KJ463002* | *KJ463119* | *KJ463235* | *KJ462889* |
| *C. silvicola* | CBS 134836 | Soil | Araponga, Minas Gerais, Brazil | KM395975 | KM396062 | KM396145 | KM395888 |
| | **CBS 135237** | Soil | Araponga, Minas Gerais, Brazil | KM395978 | KM396065 | KM396148 | KM395891 |
| *C. sphaeropedunculata* | **CBS 136081**; 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 | AY725771 | AY725696 | AY725733 |
| | CBS 112934; CPC 4516 | Soil | Indonesia | AY725651 | AY725773 | AY725798 | AY725735 |
| *C. terrae-reginae* | **CBS 112151**; CPC 3202 | *E. urophylla* | Queensland, Australia | FJ918506 | GQ267451 | FJ918522 | FJ918545 |
| | CBS 112634; CPC 4233 | *Xanthorrhoea australis* | Victoria, Australia | FJ918507 | GQ267452 | DQ190668 | FJ918546 |
| *C. terrestris* | **CBS 136642**; CMW 35180; CERC 1856 | 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* |



Please cite this article in press as: Lombard L, et al., New species, hyper-diversity and potential importance of *Calonectria* spp. from *Eucalyptus* in South China, Studies in Mycology (2015), http://dx.doi.org/10.1016/j.simyco.2014.11.003

**Table 1.** (Continued).

| Species | Isolate nr.[1] | Substrate | Locality | GenBank Accession no.[2] | | | |
|---|---|---|---|---|---|---|---|
| | | | | *tub2* | *cmdA* | *his3* | *tef1* |
| *C. tetraramosa* | **CBS 136635**; CMW 31474; CERC 1809 | *E. urophylla* × *E. grandis* clone seedling leaf | CERC Nursery, Zhanjiang, Guangdong, China | *KJ463011* | *KJ463128* | *KJ463244* | *KJ462898* |
| | CBS 136637; CMW 31476; CERC 1811 | *E. urophylla* × *E. grandis* clone seedling leaf | CERC Nursery, Zhanjiang, Guangdong, China | *KJ463012* | *KJ463129* | *KJ463245* | *KJ462899* |
| *C. turangicola* | **CBS 136077**; CMW 31411; CERC 1746 | Soil in *Eucalyptus* plantation | Fangchenggang, Guangxi, China | *KJ463013* | – | *KJ463246* | *KJ462900* |
| | CBS 136093; CMW 35410; CERC 1901 | Soil in *Eucalyptus* plantation | Guangxi, China | *KJ463014* | *KJ463130* | *KJ463247* | *KJ462901* |
| | CBS 136652; CMW 37977; CERC 1940 | Soil | Guangxi, China | *KJ463015* | *KJ463131* | *KJ463248* | *KJ462902* |
| | CMW 35383; CERC 1885 | Soil in *Eucalyptus* plantation | Hainan, China | *KJ463016* | *KJ463132* | *KJ463249* | *KJ462903* |

[1] ATCC: American Type Culture Collection, Virginia, USA; CBS: CBS-KNAW Fungal Biodiversity Centre, Utrecht, The Netherlands; CERC: China Eucalypt 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, Egham, Bakeham Lane, UK; LPF: Laboratório de Patologia Florestal, Universidade Federal de Viçosa, Viçosa, Brazil; MUCL: Mycothèque, Laboratoire de Mycologie Systématique st Appliqée, l'Université, Louvian-la-Neuve, Belgium; UFV: Universidade Federal de Viçosa, Viçosa, Brazil. Isolates obtained during the survey indicated in grey blocks.

[2] *tub2* = β-tubulin, *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*.

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

## 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 133349) 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 133639, CBS 1336640, 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. polizzii* (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. cerciana* (ex-type CBS 123693) with four isolates (CBS 136084, CBS 136096, CBS 136097 & 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 100 trees (TL = 1 264; CI = 0.672; RC = 0.633; RI = 0.942) of which the first is presented (Fig. 2). In this tree, 35 isolates clustered within the clade (BS = 97; 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



Please cite this article in press as: Lombard L, et al., New species, hyper-diversity and potential importance of *Calonectria* spp. from *Eucalyptus* in South China, Studies in Mycology (2015), http://dx.doi.org/10.1016/j.simyco.2014.11.003

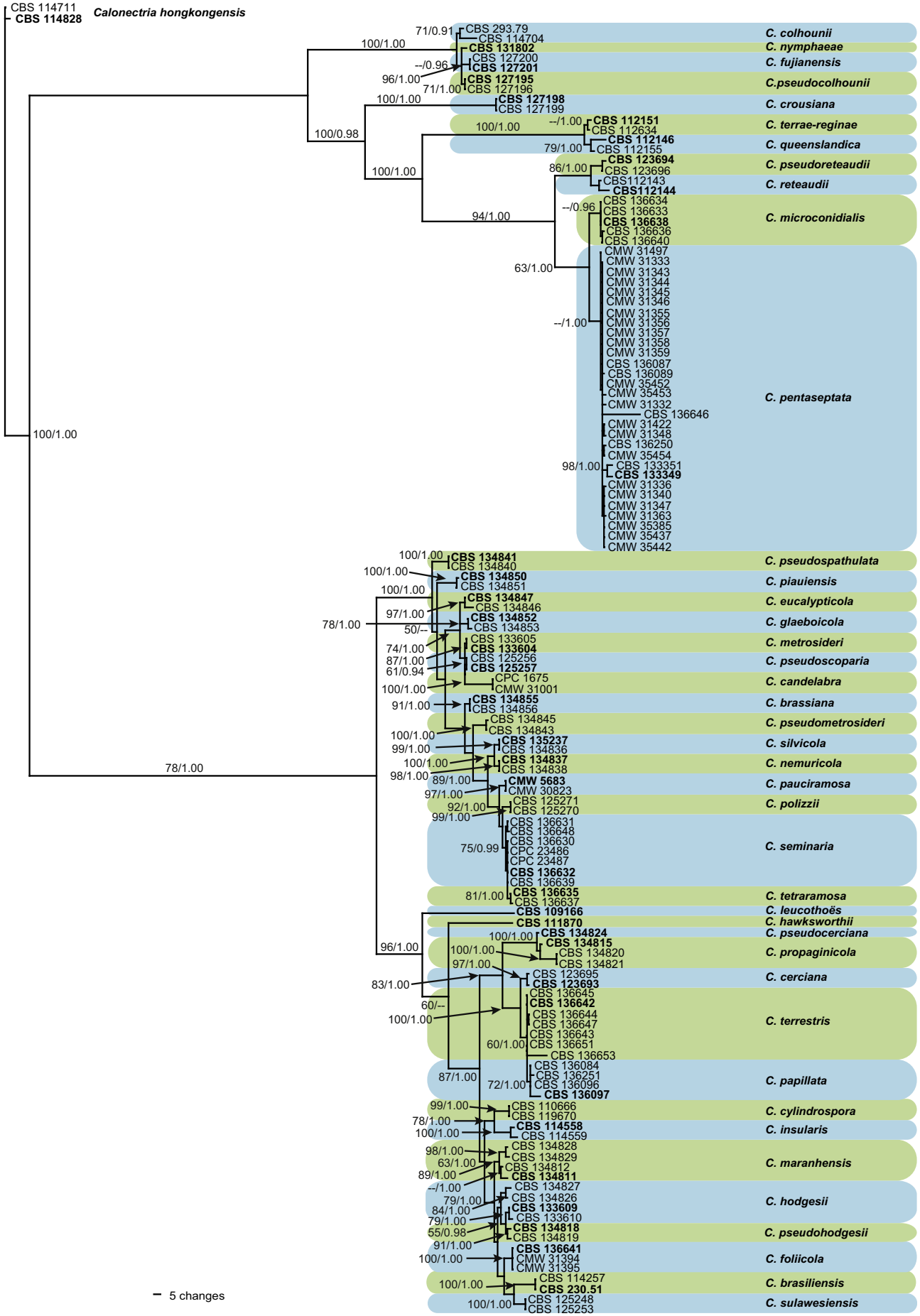


**Fig. 1.** One of 1 000 equally most parsimonious trees obtained from a heuristis search with 1 000 random taxon additions of the combined *cmdA*, *his3*, *tef1* and *tub2* sequence alignments of the Prolate group. Scale bar shows 5 changes. Bootstrap support values and Bayesian posterior probability values are shown at the nodes. The tree was rooted to *C. hongkongensis* (CBS 114711 & CBS 114878). Ex-type strains are indicated in **bold**.



Please cite this article in press as: Lombard L, et al., New species, hyper-diversity and potential importance of *Calonectria* spp. from *Eucalyptus* in South China, Studies in Mycology (2015), http://dx.doi.org/10.1016/j.simyco.2014.11.003

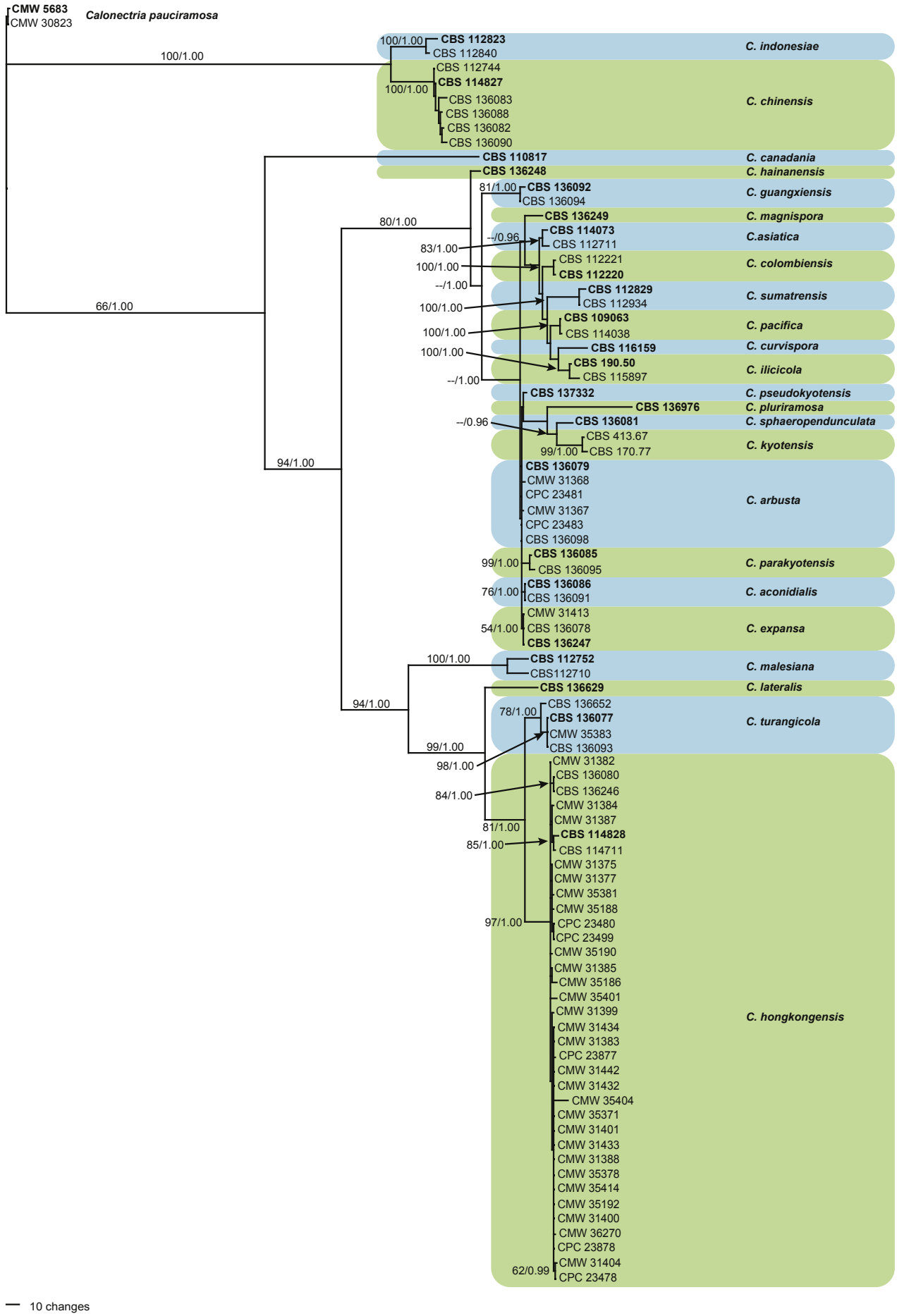


**Fig. 2.** One of 1 000 equally most parsimonious trees obtained from a heuristis search with 1 000 random taxon additions of the combined *cmdA*, *his3*, *tef1* and *tub2* sequence alignments of the Sphaero-Naviculate Group. Scale bar shows 10 changes. Bootstrap support values and Bayesian posterior 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**.



Please cite this article in press as: Lombard L, et al., New species, hyper-diversity and potential importance of *Calonectria* spp. from *Eucalyptus* in South China, Studies in Mycology (2015), http://dx.doi.org/10.1016/j.simyco.2014.11.003

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, 297–366 µm high, 232–304 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*, 37–75 µm thick, cells becoming more compressed towards the inner layer of *textura angularis*, 16–23 µm thick, cells becoming thin-walled and hyaline towards the centre; outermost cells 14–45 × 12–35 µm, cells of inner layer 10–25 × 3–7 µm; ascomatal 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. *Asci* 8-spored, clavate, 111–113 × 15–18 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, (28–)32–40(–44) × 5–7 µm (av. 36 × 6 µm). Homothallic. *Mega-*, *macro-* and *microconidia* not observed.

*Culture characteristics*: Colonies moderately fast growing at 24 °C on MEA with mycelium immersed in medium with no sporulation on the medium surface; surface and reverse white to pale luteous after 7 d.

*Specimens examined*: **China**, Hainan Province, from soil collected in a *Eucalyptus* plantation, Aug. 2009, X. Mou & S.F. Chen (**holotype** CBS H-21481, culture ex-type CBS 136086 = CMW 35174 = CERC 1850), CBS 136091 = CPC 23504 = CMW 35384 = CERC 1886.

*Notes*: All attempts to induce the asexual morph of *C. aconidialis* failed. Ascomata formed readily within 16 d on MEA, SNA and MSA, either on the surface or immersed in the medium, exuding viable ascospores after 20 d.

***Calonectria arbusta*** L. Lombard, Crous & S.F. Chen, **sp. nov.** MycoBank MB809045. Fig. 4.

*Etymology*: Name refers to a plantation and the environment from which this fungus was collected.

*Ascomata* perithecial, solitary, 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, guttulate, 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 sphaeropedunculate 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 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-21482, living ex-type culture CBS 136079 = CPC 23482 = CMW 31370 = CERC 1705), CPC 23481 = CMW 31369 = CERC 1704, CPC 23483 = 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).



Please cite this article in press as: Lombard L, et al., New species, hyper-diversity and potential importance of *Calonectria* spp. from *Eucalyptus* in South China, Studies in Mycology (2015), http://dx.doi.org/10.1016/j.simyco.2014.11.003

**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) | Size (μm) | Septation | Size (μm) | Branches | (μm) | Diam (μm) | Shape | Size (μm) | Septation | |
| ***Calonectria reteaudii* species complex** | | | | | | | | | | | | | |
| *C. microconidialis* | | | | | | 26–92 × 35–95 | 3 | 175–441 × 4–7 | 3–7 | Narrowly clavate | (69–)78–98(–113) × 7–9(10) | 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–)6–8(–10) | 5(–8) | Crous *et al.* (2012) |
| *C. pseudoreteaudii* | | | | | | 26–82 × 45–103 | 3 | 193–313 × 5–6 | 3–5 | Narrowly clavate | (61–)65–73(–78) × (4–)5–6(–7) | 4–6 | Lombard *et al.* (2010d) |
| *C. queenslandica* | | | | | | 27–68 × 39–64 | 3 | 105–156 × 4–5 | 3–4 | Narrowly clavate | (61–)65–73(–78) × (4–)5–6(–7) | 4–6 | Lombard *et al.* (2010d) |
| *C. reteaudii* | 350–450 × 250–350 | Subglobose to ovoid | 70–150 × 7–20 | (50–)65–85(–100) × (4–)5–6(–7) | (1–)3(–5) | 20–70 × 80–100 | 6 | 150–380 × 2.5–3.5 | 3–6 | Clavate | (50–)75–95(–120) × (5–)6–7 | (1–)5(–6) | Crous (2002) |
| *C. terrae-reginae* | | | | | | 33–48 × 35–54 | 4 | 127–235 × 4–6 | 3–5 | Narrowly clavate | 60–83(–87) × (4–)5–7(–8) | 4–6 | Lombard *et al.* (2010d) |
| ***Calonectria candelabra* species complex** | | | | | | | | | | | | | |
| *C. brassiana* | | | | | | 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 | Subglobose to ovoid | 70–130 × 7–15 | (40–)45–50(–60) × 5–6 | 1 | 30–70 × 50–80 | 5 | 100–220 × 3–3.5 | 5–8 | 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. glaeboicola* | | | | | | 25–40 × 27–45 | 2 | 100–165 × 2–4 | 3–5 | 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. nemuricola* | | | | | | 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 | Subglobose to ovoid | 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–)4–5 | 1 | Schoch *et al.* (1999) |
| *C. piauiensis* | | | | | | 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. polizzii* | | | | | | 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. pseudoscoparia* | | | | | | 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*)



Please cite this article in press as: Lombard L, et al., New species, hyper-diversity and potential importance of *Calonectria* spp. from *Eucalyptus* in South China, Studies in Mycology (2015), http://dx.doi.org/10.1016/j.simyco.2014.11.003

Please cite this article in press as: Lombard L, et al., New species, hyper-diversity and potential importance of *Calonectria* spp. from *Eucalyptus* in South China, Studies in Mycology (2015), http://dx.doi.org/10.1016/j.simyco.2014.11.003

**Table 2.** (Continued)

| Species | Perithecia | | Asci | Ascospores | | Conidiogenous apparatus | | Stipe extension | Vesicle | | Macroconidia | | Reference |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | Size (µm) | Shape | Size (µm) | Size (µm) | Septation | Size (µm) | Branches | (µm) | Diam (µm) | Shape | Size (µm) | Septation | |
| *C. pseudospathulata* | | | | | | 60–100 × 30–70 | 3 | 145–190 × 2–4 | 7–10 | Obpyriform | (35–)41–44(–50) × 3–5 | 1 | Alfenas *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. silvicola* | | | | | | 45–105 × 35–90 | 3 | 130–195 × 3–4 | 7–10 | Obpyriform | (30–)40–42(–50) × 3–5 | 1 | Alfenas *et al.* (2015) |
| *C. tetraramosa* | | | | | | 54–95 × 36–75 | 4 | 102–253 × 3–6 | 4–10 | Obpyriform | (45–) 46.5–49.5(–51) × (4–)4.5–5.5(–6) | 1 | This study |
| *C. zuluensis* | 292–394 × 170–285 | Subglobose to ovoid | 92–140 × 10–16 | (26–)29–34(–38) × 4–5 | 1 | 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) |
| ***Calonectria cylindrospora* species complex** | | | | | | | | | | | | | |
| *C. brasiliensis* | | | | | | 81–103 × 58–90 | 3 | 204–266 × 6–7 | 7–11 | Ellipsoid to obpyriform | (35–)36–40(–41) × 3–5 | 1 | Lombard *et al.* (2010a) |
| *C. cerciana* | | | | | | 62–113 × 70–98 | 4 | 148–222 × 5–6 | 8–13 | Fusiform to obpyriform | (37–)41–46(–49) × 5–6 | 1 | Lombard *et al.* (2010d) |
| *C. cylindrospora* | 280–520 × 280–400 | Globose to subglobose | 75–100 × 8–15 | (24–)30–40(–49) × (4–)5–6(–8) | 1 | 60–100 × 60–110 | 6 | 150–200 × 3–4 | 6–8 | Ellipsoid to pyriform or clavate | (40–)42–50(–66) × 3–4(–5) | 1 | Crous (2002) |
| *C. foliicola* | | | | | | 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* | | | | | | 61–72 × 45–65 | 3 | 136–196 × 2–4 | 6–11 | Pyriform to ellipsoidal or ovoid to sphaeropedunculate | (44–)49–51(–55) × 3–5 | 1 | Alfenas *et al.* (2013b) |
| *C. insularis* | 350–450 × 300–350 | Subglobose to ovoid | 70–125 × 7–18 | (27–)30–36(–42) × 5–6(–7) | 1 | 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. leucothoës* | | | | | | 25–50 × 50–80 | 6 | 160–250 × 3–6 | 6–11.5 | Ellipsoid to obpyriform | (45–)68–78(–97) × (4–)5–5.5(–6.5) | (1–)3(–6) | Crous (2002) |
| *C. maranhensis* | | | | | | 45–65 × 45–71 | 3 | 125–190 × 3–5 | 7–11 | Ellipsoid, obpyriform to sphaeropedunculate | (50–)56–58(–65) × (3–)5(–6) | 1 | Alfenas *et al.* (2015) |
| *C. mexicana* | 400–450 × 350–450 | Subglobose to ovoid | 70–120 × 10–20 | (35–)40–55(–65) × 5–6(–7) | | 25–60 × 40–70 | 3 | 160–250 × 2–3 | 7–12 | Broadly ellipsoid with papillate apex | (35–)40–48(–52) × 3–4(–4.5) | 1 | Crous (2002) |
| *C .papillata* | 425–455 × 345–395 | Subglobose to ovoid | 106–112 × 16–20 | (27–)32–40(–46) × 5–6(–7) | 1 | 45–114 × 33–82 | 4 | 163–218 × 4–7 | 8–14 | Obpyriform to ellipsoidal with papillate apex | (40–)43–47(–50) × (3–)4–5 | 1 | This study |
| *C. propaginicola* | | | | | | 40–75 × 31–85 | 4 | 130–250 × 2–5 | 5–12 | Ellipsoid, obpyriform to sphaeropedunculate | (40–)48–51(–55) × 3–5 | 1 | Alfenas *et al.* (2015) |

Please cite this article in press as: Lombard L, et al., New species, hyper-diversity and potential importance of *Calonectria* spp. from *Eucalyptus* in South China, Studies in Mycology (2015), http://dx.doi.org/10.1016/j.simyco.2014.11.003

**Table 2.** (Continued)

| Species | Perithecia | | Asci | Ascospores | | Conidiogenous apparatus | | Stipe extension | Vesicle | | Macroconidia | | Reference |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | Size (µm) | Shape | Size (µm) | Size (µm) | Septation | Size (µm) | Branches | (µm) | Diam (µm) | Shape | Size (µm) | Septation | |
| *C. pseudocerciana* | | | | | | 50–90 × 40–95 | 3 | 130–190 × 2–5 | 7–12 | Obpyriform to sphaeropedunculate | (35–)43–46(–55) × 3–5 | 1 | Alfenas *et al.* (2015) |
| *C. pseudohodgesii* | | | | | | 50–90 × 40–95 | 3 | 130–190 × 2–5 | 7–12 | Obpyriform to sphaeropedunculate | (35–)43–46(–55) × 3–5 | 1 | Alfenas *et al.* (2015) |
| *C. sulawesiensis* | | | | | | 43–81 × 41–79 | 5 | 113–262 × 5–7 | 5–7 | Broadly clavate to ellipsoid | (41–)45–51(–54) × (3–)4(–6) | 1 | Lombard *et al.* (2010b) |
| *C. terrestris* | | | | | | 35–89 × 35–102 | 4 | 147–228 × 4–7 | 5–12 | Obpyriform to pyriform to broadly clavate | (33–)36–40(–41) × (3–)4–5 | 1 | This study |
| ***Calonectria kyotensis* species complex** | | | | | | | | | | | | | |
| *C. aconidialis* | 297–366 × 232–304 | Subglobose to ovoid | 111–113 × 15–18 | (28–)32–40(–44) × 5–7 | 1 | | | | | | | | This study |
| *C. arbusta* | 357–444 × 276–391 | Subglobose to ovoid | 97–119 × 16–19 | (30–)35–41(–43) × 5–7(–8) | 1 | 58–151 × 54–108 | 5 | 134–196 × 3–6 | 7–13 | Sphaeropedunculate | (41–)42–48(–52) × 4–6 | 1 | This study |
| *C. asiatica* | 280–400 × 200–350 | Subglobose to ovoid | 70–120 × 12–20 | (28–)30–38(–40) × (5–)6(–7) | 1 | 40–80 × 40–90 | 5 | 200–280 × 3–7 | 12–17 | Sphaeropedunculate | (42–)48–55(–65) × (4–)5(–5.5) | 1 | Crous *et al.* (2004b) |
| *C. canadania* | | | | | | | 3 | 100–180 × 3–4 | 6–10 | Pyriform to sphaeropedunculate | (38–)48–55(–65) × 4(–5) | 1 | Kang *et al.* (2001b) |
| *C. chinensis* | | | | | | 40–60 × 40–60 | 3 | 120–150 × 2.5–3.5 | 6–9 | Sphaeropedunculate | (38–)41–48(–56) × (3.5–)4(–4.5) | 1 | Crous *et al.* (2004b) |
| *C. colombiensis* | 200–350 × 200–300 | Subglobose to ovoid | 90–150 × 11–23 | (28–)30–35(–40) × (4–)5(–6) | 1 | 25–60 × 40–60 | 5 | 130–200 × 3–4 | 7–12 | Sphaeropedunculate | (33–)48–58(–60) × (4–)4.5(–5) | 1(–3) | Crous *et al.* (2004b) |
| *C. curvispora* | | | | | | 15–30 × 35–50 | 3 | 110–150 × 2–3 | 5–10 | Sphaeropedunculate | (45–)55–65(–70) × (4–)5–6 | 1(–3) | Crous (2002) |
| *C. expansa* | 310–520 × 270–435 | Subglobose to ovoid | 107–146 × 16–21 | (33–)36–41(–44) × (4–)5–7 | 1 | 26–116 × 45–82 | 5 | 124–216 × 3–7 | 8–16 | Sphaeropedunculate | (44–)48–52(–57) × 4–6 | 1 | This study |
| *C. guangxiensis* | 295–435 × 265–355 | Subglobose to ovoid | 83–146 × 15–23 | (23–)32–40(–42) × 5–7(–8) | 1 | 31–95 × 55–85 | 4 | 175–193 × 5–7 | 11–14 | Sphaeropedunculate | (42–)45–49(–52) × 4–6 | 1 | This study |
| *C. hainanensis* | 300–455 × 230–385 | Subglobose to ovoid | 91–110 × 15–22 | (24–)30–38(–42) × (4–)5–7 | 1 | 54–119 × 41–80 | 5 | 112–186 × 5–9 | 7–14 | Sphaeropedunculate | (41–)43–49(–52) × 4–6 | 1 | This study |
| *C. hongkongensis* | 350–550 × 300–450 | Subglobose to ovoid | 80–140 × 14–20 | (25–)28–35(–40) × (4–)5–6(–7) | 1 | 70–120 × 70–100 | 8 | 100–200 × 3–4 | 8–14 | Sphaeropedunculate | (38–)45–48(–53) × 4(–4.5) | 1 | Crous *et al.* (2004b) |
| *C. ilicicola* | 300–550 × 280–400 | Subglobose to ovoid | 90–140 × 12–19 | (30–)37–50(–65) × (4–)5–6.5(–7) | 1(–3) | 25–100 × 55–100 | 3 | 120–140 × 3–4 | 6–12 | Sphaeropedunculate | (45–)70–82(–90) × (4–)5–6.5(–7) | (1–)3 | Crous (2002) |
| *C. indonesiae* | | | | | | 60–80 × 40–60 | 5 | 110–160 × 2.5–3 | 7–9 | Sphaeropedunculate | (40–)45–55(–60) × (3–)4 | 1 | Crous *et al.* (2004b) |
| *C. kyotensis* | 280–550 × 210–425 | Subglobose to ovoid | 70–140 × 13–22 | (18–)28–40(–48) × (4–)5–6(–7) | 1 | 40–100 × 40–90 | 5 | 100–200 × 3–4 | 6–12 | Sphaeropedunculate | (35–)45–50(–55) × 3–4(–5) | 1 | Crous (2002) |

(*continued on next page*)

Please cite this article in press as: Lombard L, et al., New species, hyper-diversity and potential importance of *Calonectria* spp. from *Eucalyptus* in South China, Studies in Mycology (2015), http://dx.doi.org/10.1016/j.simyco.2014.11.003



**Table 2.** (Continued)

| Species | Perithecia | | Asci | Ascospores | | Conidiogenous apparatus | | Stipe extension | Vesicle | | Macroconidia | | Reference |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | Size (µm) | Shape | Size (µm) | Size (µm) | Septation | Size (µm) | 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 | Subglobose to ovoid | 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. parakyotensis* | | | | | | 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. pseudokyotensis* | | | | | | 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 × 345–465 | Subglobose to ovoid | 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–)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 |

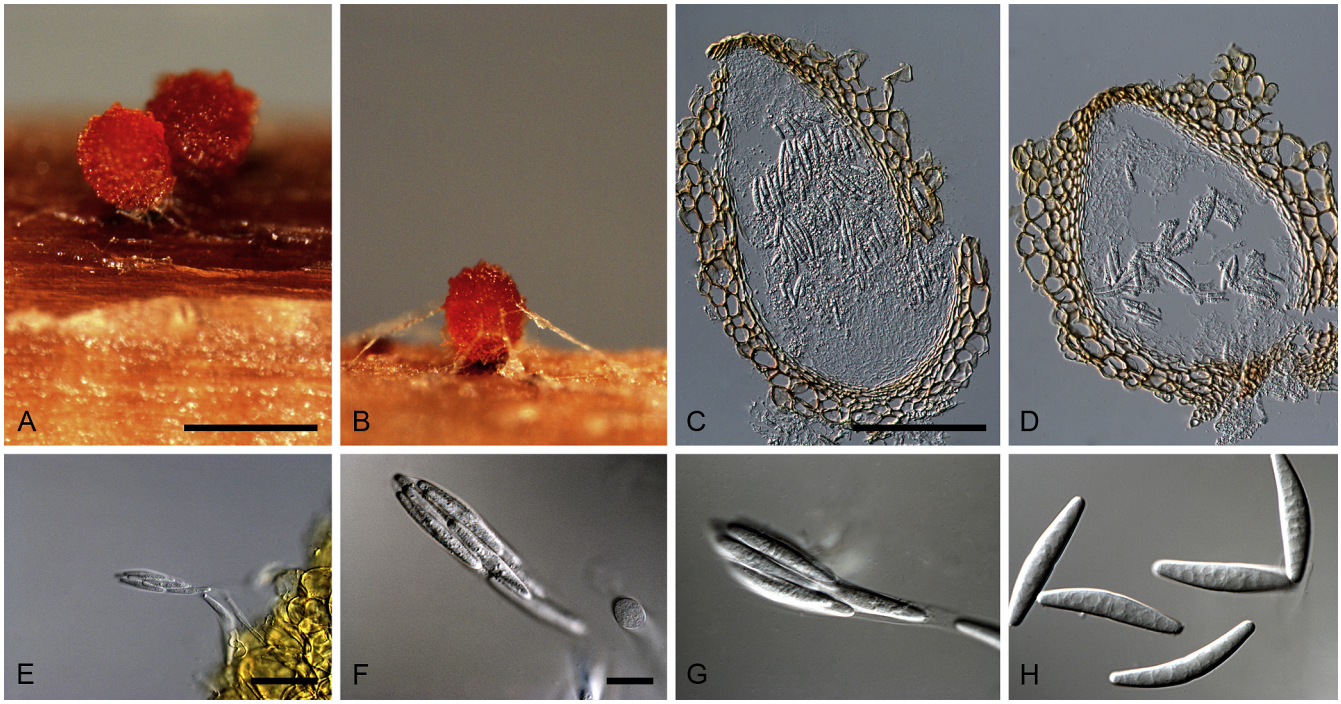


**Fig. 3.** *Calonectria aconidialis* (ex-type CBS 136086). A–B. Ascomata. C–D. Vertical section through ascomata, showing wall structure. E–G. Asci. H. Ascospores. Scale bars: A = 500 µm; C = 100 µm (apply to D); E = 50 µm; F = 10 µm (apply to G–H).

***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+; ascomatal 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; ascomatal 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. *Asci* 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 doliiform 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,



Please cite this article in press as: Lombard L, et al., New species, hyper-diversity and potential importance of *Calonectria* spp. from *Eucalyptus* in South China, Studies in Mycology (2015), http://dx.doi.org/10.1016/j.simyco.2014.11.003

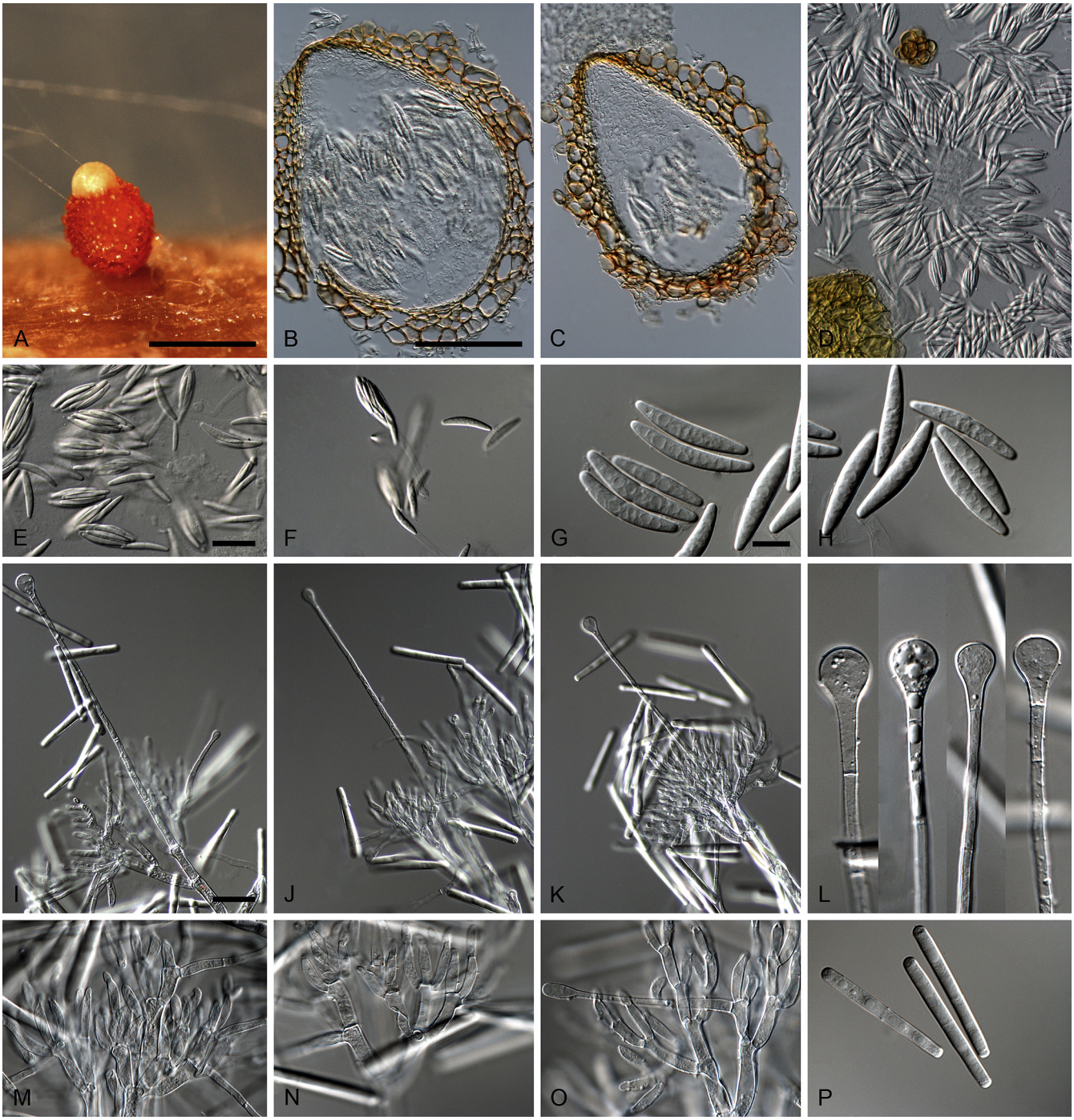


**Fig. 4.** *Calonectria arbusta* (ex-type CBS 136079). A. Ascoma. B–C. Vertical section through ascomata, showing wall structure. D–F. Asci. G–H. Ascospores. I–K. Macroconidiophores. L. Sphaeropedunculate vesicles. M–N. Conidiogenous apparatus with conidiophore branches and doliiform to reniform phialides. O. Conidiogenous apparatus with lateral stipe extension. P. Macroconidia. Scale bars: A = 500 µm; B = 100 µm (apply to C–D); E = 50 µm (apply to F, I–K), G = 10 µm (apply to H, L–P).

terminating in a obpyrifrom 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



Please cite this article in press as: Lombard L, et al., New species, hyper-diversity and potential importance of *Calonectria* spp. from *Eucalyptus* in South China, Studies in Mycology (2015), http://dx.doi.org/10.1016/j.simyco.2014.11.003

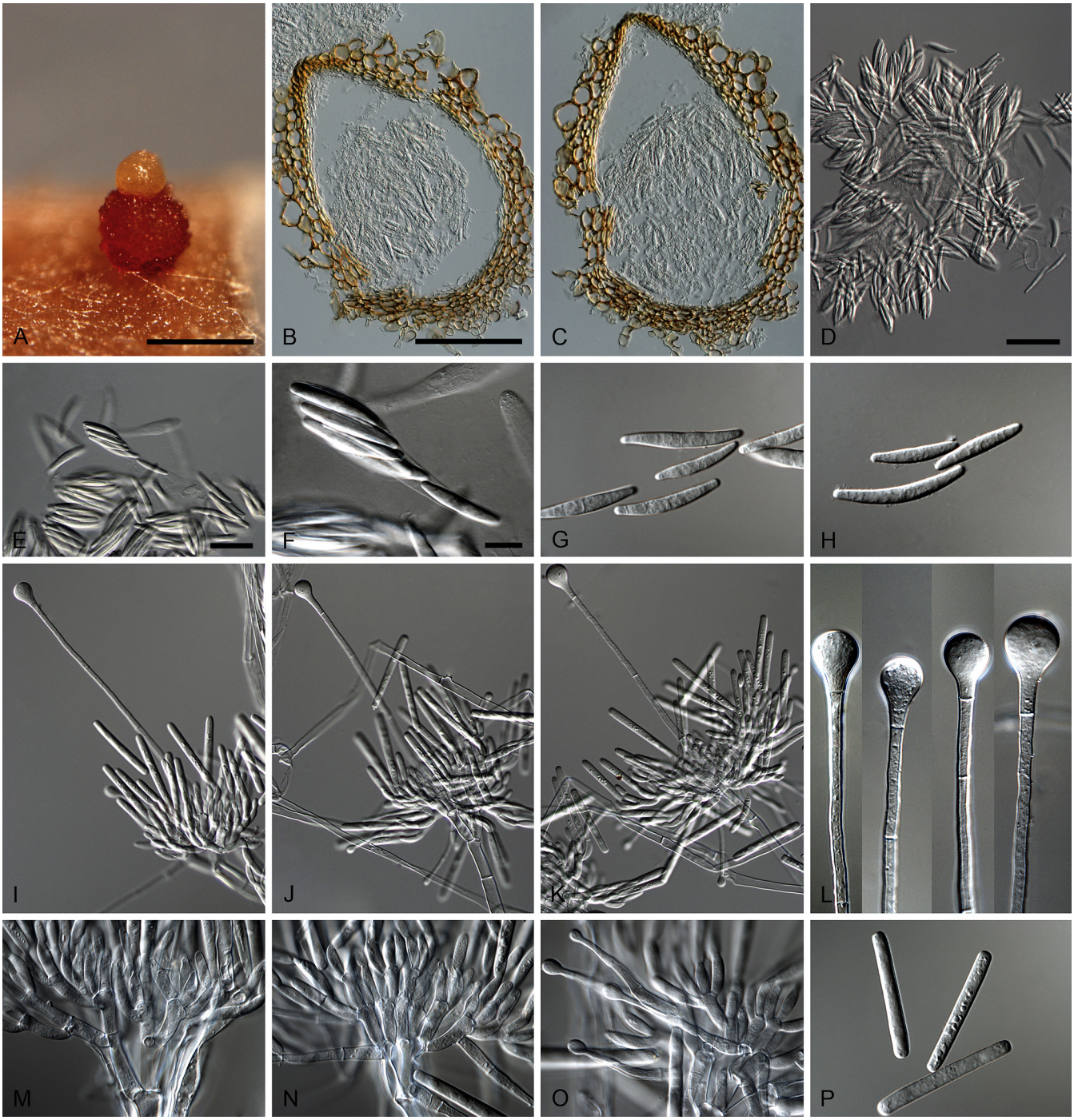


**Fig. 5.** *Calonectria expansa* (ex-type CBS 136247). A. Ascoma. B–C. Vertical section through ascomata, showing wall structure. D–F. Asci. G–H. Ascospores. I–K. Macroconidiophores. L. Sphaeropedunculate vesicles. M–N. Conidiogenous apparatus with conidiophore branches and doliiform to reniform phialides. O. Conidiogenous apparatus with lateral stipe extension. P. Macroconidia. Scale bars: A = 500 µm; B, D = 100 µm (apply to C); E = 50 µm (apply to I–K), F = 10 µm (apply to G–H, L–P).

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



Please cite this article in press as: Lombard L, et al., New species, hyper-diversity and potential importance of *Calonectria* spp. from *Eucalyptus* in South China, Studies in Mycology (2015), http://dx.doi.org/10.1016/j.simyco.2014.11.003

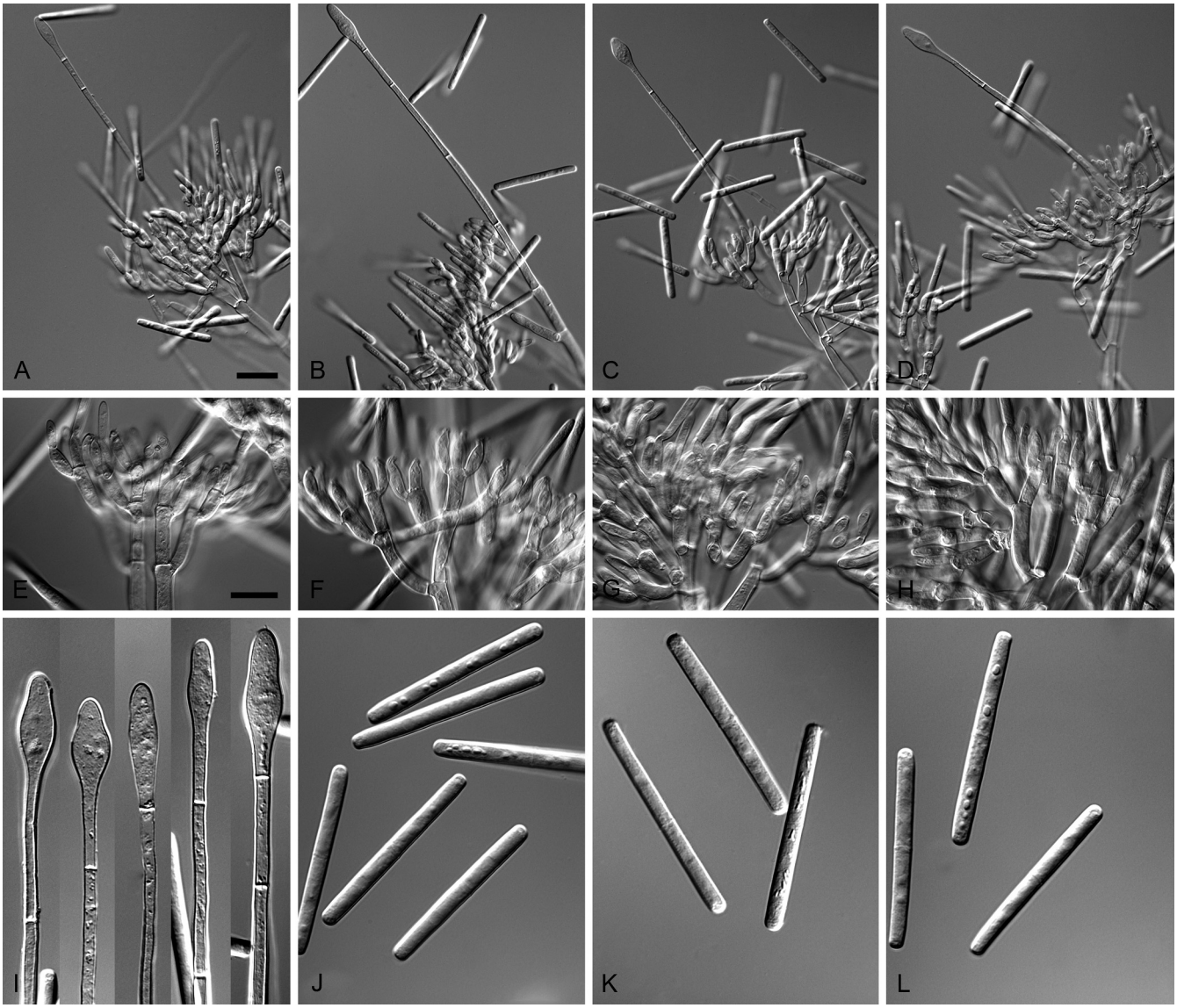


**Fig. 6.** *Calonectria foliicola* (ex-type 136641). A–D. Macroconidiophores. E–H. Conidiogenous apparatus with conidiophore branches and doliiform to reniform phialides. I. Obpyriform to ellipsoidal vesicles. J–L. Macroconidia. Scale bars: A = 50 µm (apply to B–D); E = 10 µm (apply to F–L).

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,



Please cite this article in press as: Lombard L, et al., New species, hyper-diversity and potential importance of *Calonectria* spp. from *Eucalyptus* in South China, Studies in Mycology (2015), http://dx.doi.org/10.1016/j.simyco.2014.11.003

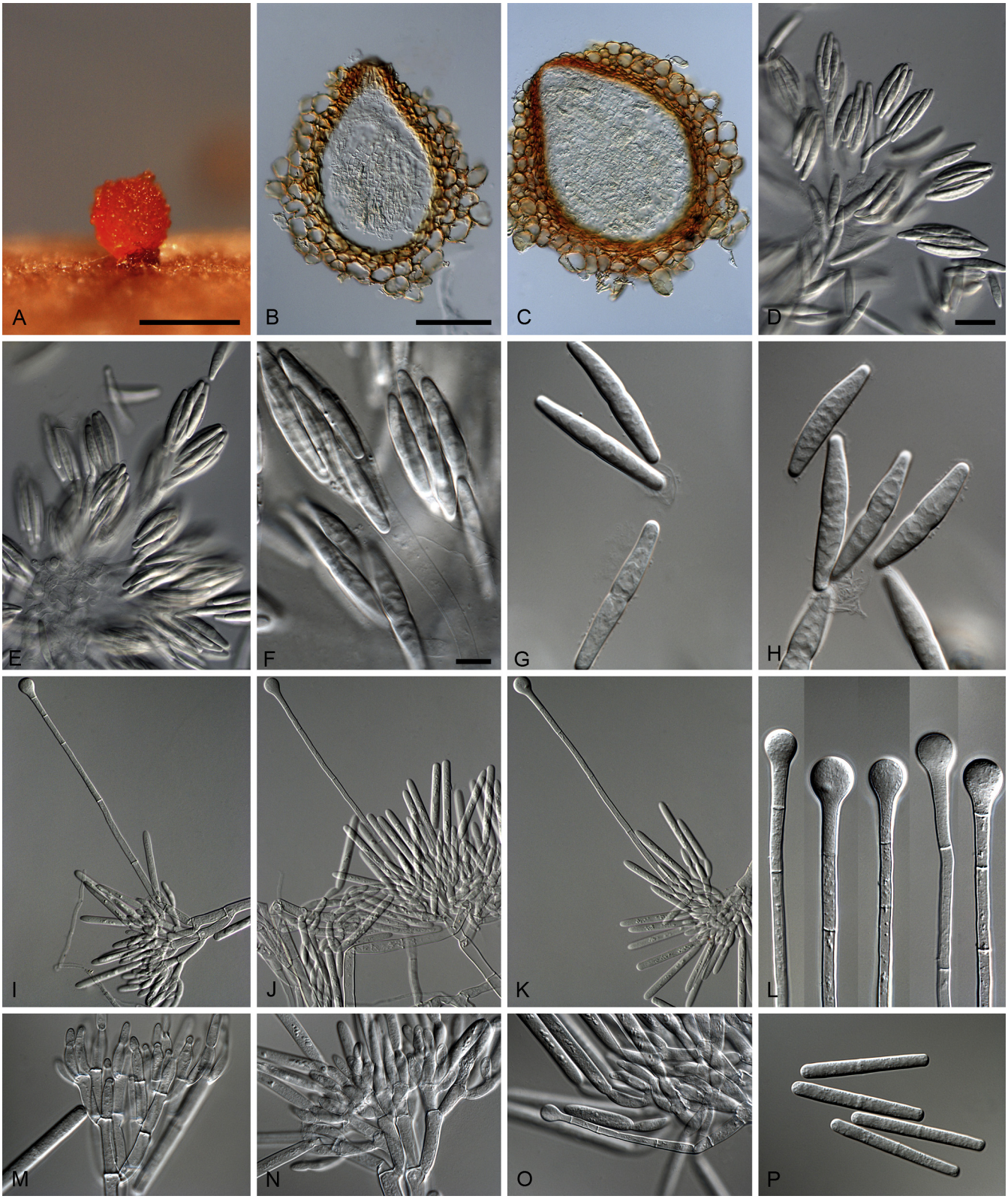


**Fig. 7.** *Calonectria guangxiensis* (ex-type CBS 136092). A. Ascoma. B–C. Vertical section through ascomata, showing wall structure. D–F. Asci. G–H. Ascospores. I–K. Macroconidiophores. L. Sphaeropedunculate vesicles. M–N. Conidiogenous apparatus with conidiophore branches and doliiform to reniform phialides. O. Conidiogenous apparatus with lateral stipe extension. P. Macroconidia. Scale bars: A = 500 µm; B = 100 µm (apply to C); D = 50 µm (apply to E, I–K), F = 10 µm (apply to G–H, L–P).

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, 1-septate, sometimes constricted at the septum, (24–)30–38(–42) × (4–)5–7 µm (av. 34 × 6 µm). Homothallic. *Macroconidiophores*



Please cite this article in press as: Lombard L, et al., New species, hyper-diversity and potential importance of *Calonectria* spp. from *Eucalyptus* in South China, Studies in Mycology (2015), http://dx.doi.org/10.1016/j.simyco.2014.11.003


**Fig. 8.** *Calonectria hainanensis* (ex-type CBS 136248). A. Ascoma. B–C. Vertical section through ascomata, showing wall structure. D–F. Asci. G–H. Ascospores. I–K. Macroconidiophores. L. Sphaeropedunculate vesicles. M–N. Conidiogenous apparatus with conidiophore branches and doliiform to reniform phialides. O. Conidiogenous apparatus with lateral stipe extension. P. Macroconidia. Scale bars: A = 500 µm; B = 100 µm (apply to C); D = 50 µm (apply to I–K), E = 10 µm (apply to F), G = 10 µm (apply to H, L–P).

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.



Please cite this article in press as: Lombard L, et al., New species, hyper-diversity and potential importance of *Calonectria* spp. from *Eucalyptus* in South China, Studies in Mycology (2015), http://dx.doi.org/10.1016/j.simyco.2014.11.003

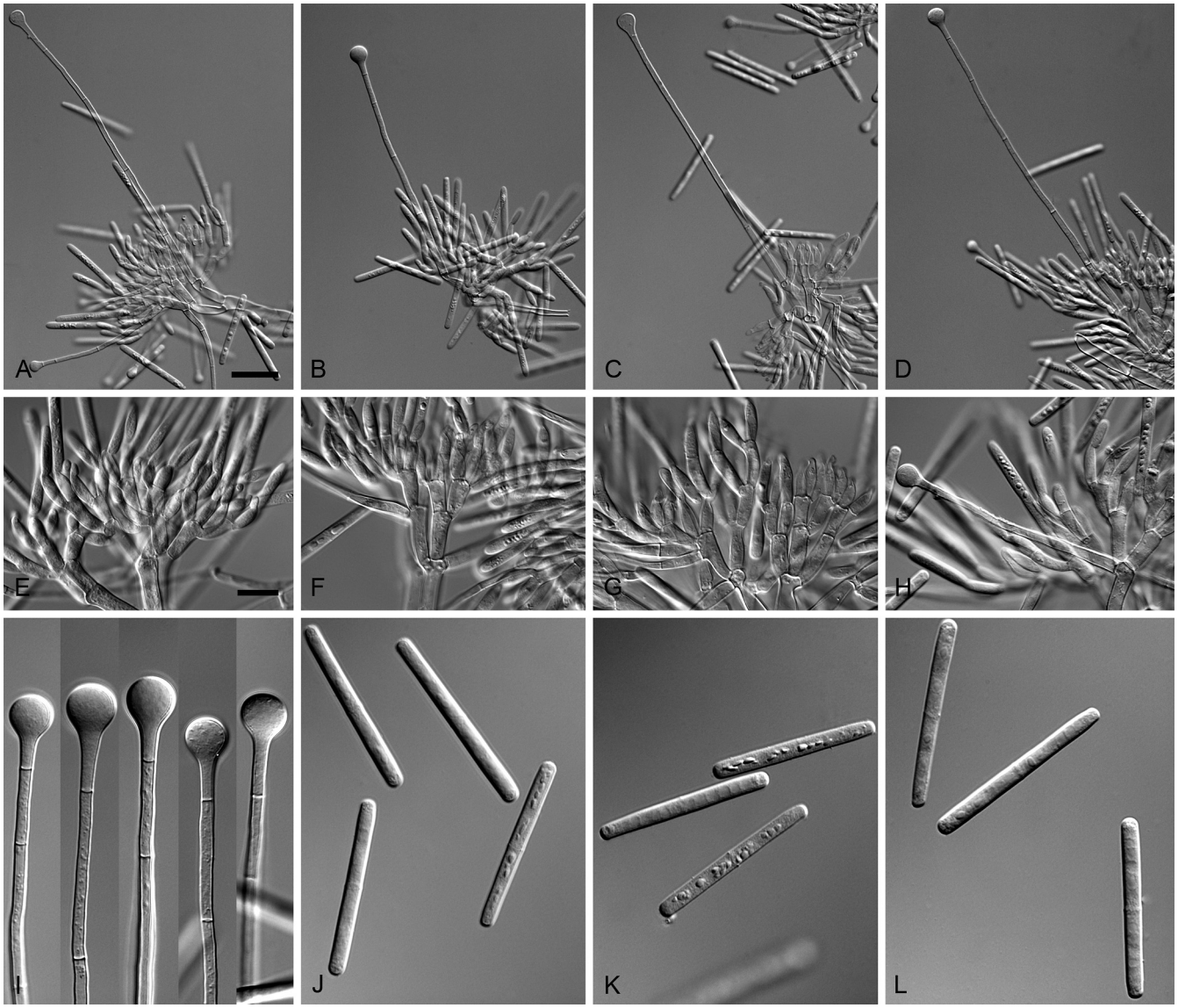


**Fig. 9.** *Calonectria lateralis* (ex-type CBS 136629). A–D. Macroconidiophores. E–H. Conidiogenous apparatus with conidiophore branches, doliiform to reniform phialides and lateral stipe extensions. I. Sphaeropedunculate vesicles. J–L. Macroconidia. Scale bars: A = 50 µm (apply to B–D); E = 10 µm (apply to F–L).

*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 to umber 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*** L. 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



Please cite this article in press as: Lombard L, et al., New species, hyper-diversity and potential importance of *Calonectria* spp. from *Eucalyptus* in South China, Studies in Mycology (2015), http://dx.doi.org/10.1016/j.simyco.2014.11.003

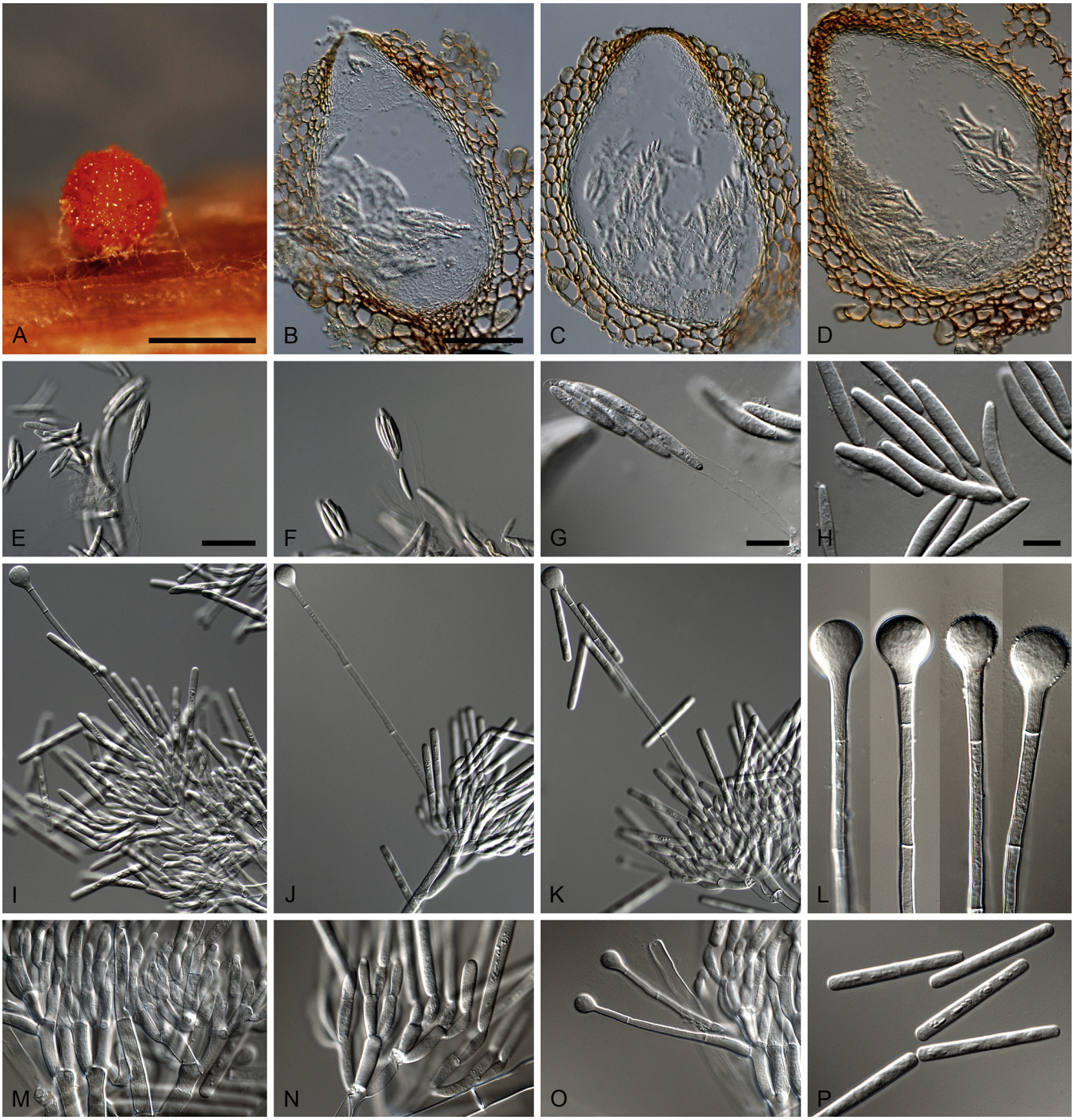


**Fig. 10.** *Calonectria magnispora* (ex-type CBS 136249). A. Ascoma. B–D. Vertical section through ascomata, showing wall structure. E–G. Asci. H. Ascospores. I–K. Macroconidiophores. L. Sphaeropedunculate vesicles. M–N. Conidiogenous apparatus with conidiophore branches and doliiform to reniform phialides. O. Conidiogenous apparatus with lateral stipe extension. P. Macroconidia. Scale bars: A = 500 µm; B = 100 µm (apply to C–D); E = 50 µm (apply to F, I–K), G = 20 µm, H = 10 µm (apply to L–P).

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*



Please cite this article in press as: Lombard L, et al., New species, hyper-diversity and potential importance of *Calonectria* spp. from *Eucalyptus* in South China, Studies in Mycology (2015), http://dx.doi.org/10.1016/j.simyco.2014.11.003

*angularis*, 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. *Conidiogenous 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 doliiform 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. *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 formed abundant 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-21471, living ex-type culture CBS 136249 = CPC 23509 = CMW 35184 = CERC 1860).

*Notes*: *Calonectria magnispora* can be distinguished from *C. arbusta*, *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).

***Calonectria microconidialis*** L. Lombard, Crous & S.F. Chen, **sp. nov.** MycoBank MB809053. Fig. 11.

*Etymology:* Name refers to the microconidial state that is readily 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, 59–195 × 7–11 µm; stipe extension septate, straight to flexuous, 175–441 µm long, 4–7 µm wide at the apical septum, terminating in a narrowly clavate vesicle, 3–7 µm diam. *Conidiogenous 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. *Microconidiophores* 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.

*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 1808, 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 microconidiophores 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. Fig. 12.

*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 3 % KOH+; ascomatal 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; ascomatal 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



Please cite this article in press as: Lombard L, et al., New species, hyper-diversity and potential importance of *Calonectria* spp. from *Eucalyptus* in South China, Studies in Mycology (2015), http://dx.doi.org/10.1016/j.simyco.2014.11.003

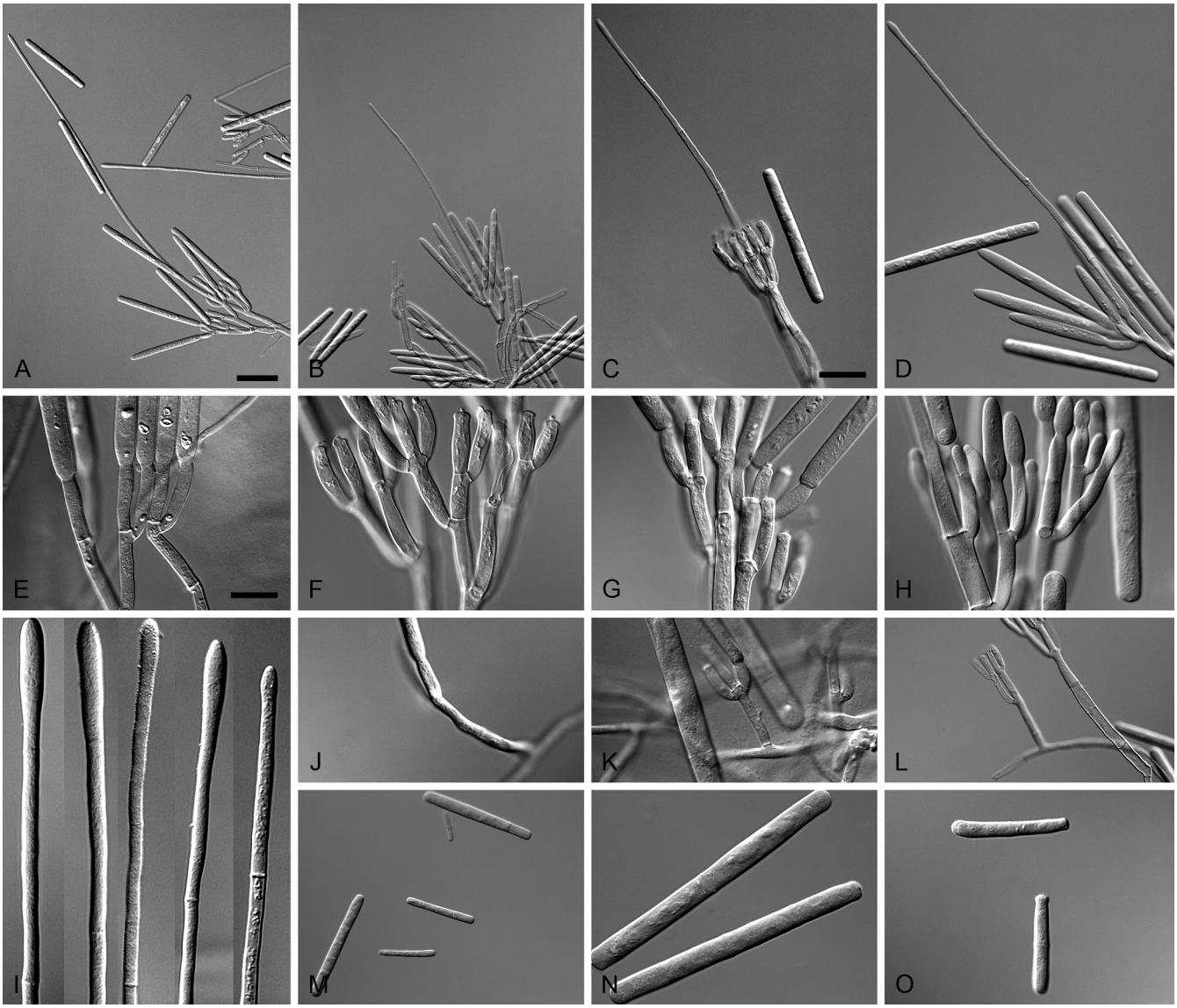


**Fig. 11.** *Calonectria microconidialis* (ex-type CBS 136638). A–D. Macroconidiophores. E–H. Conidiogenous apparatus with conidiophore branches and cylindrical to allantoid phialides. I. Narrowly clavate vesicles. J–L. Microconidiophores. M–O. Macro- and microconidia. Scale bars: A = 50 µm (apply to B, M); C = 20 µm (apply to D, L); E = 10 µm (apply to F–K, N–O).

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, 8–19 × 2–5 µm; quaternary 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 culture CBS 136097 = CPC 23517 = CMW 37976 = 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.



Please cite this article in press as: Lombard L, et al., New species, hyper-diversity and potential importance of *Calonectria* spp. from *Eucalyptus* in South China, Studies in Mycology (2015), http://dx.doi.org/10.1016/j.simyco.2014.11.003

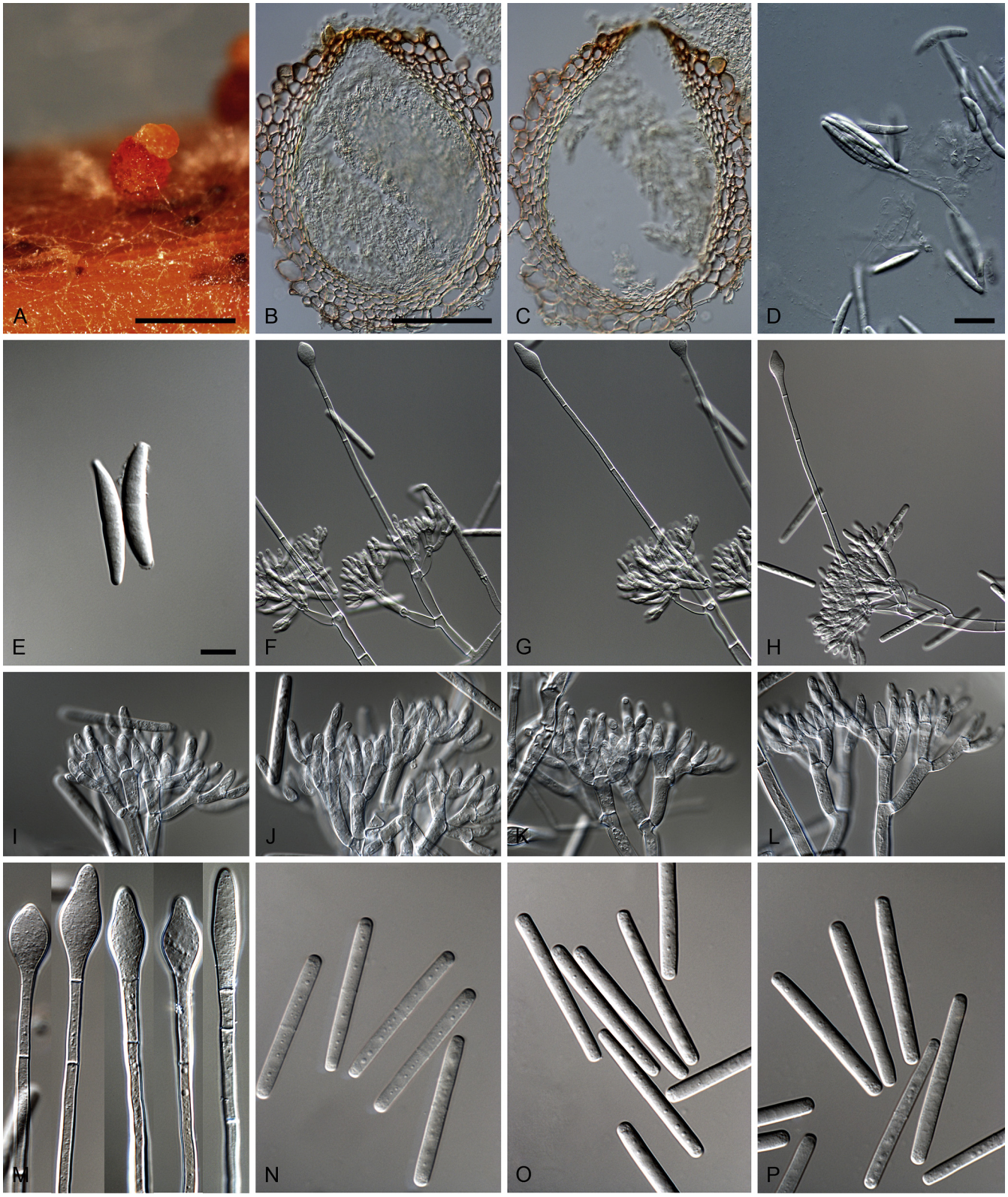


**Fig. 12.** *Calonectria papillata* (ex-type CBS 136097). A. Ascoma. B–C. Vertical section through ascomata, showing wall structure. D. Asci. E. Ascospores. F–H. Macroconidiophores. I–L. Conidiogenous apparatus with conidiophore branches and doliiform to reniform phialides. M. Obpyriform to ellipsoid 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).

*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,



Please cite this article in press as: Lombard L, et al., New species, hyper-diversity and potential importance of *Calonectria* spp. from *Eucalyptus* in South China, Studies in Mycology (2015), http://dx.doi.org/10.1016/j.simyco.2014.11.003

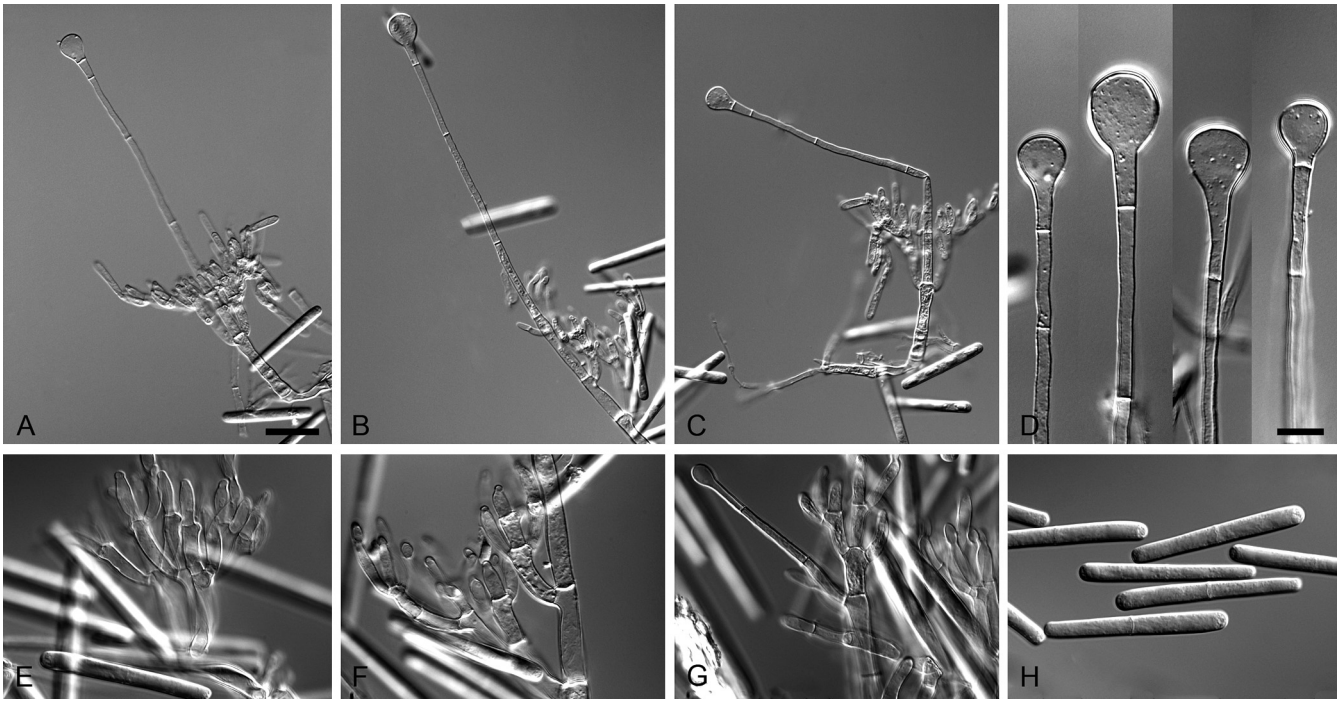


**Fig. 13.** *Calonectria parakyotensis* (ex-type CBS 136085). A–C. Macroconidiophores. D. Sphaeropedunculate vesicles. E–G. Conidiogenous apparatus with conidiophore branches, doliiform to reniform phialides and lateral stipe extensions. H. Macroconidia. Scale bars: A = 50 µm (apply to B–C); D = 10 µm (apply to E–H).

11–18 × 4–6 µm, each terminal branch producing 2–6 phialides; phialides doliiform 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(–49) × 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 a *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 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 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, 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*.



Please cite this article in press as: Lombard L, et al., New species, hyper-diversity and potential importance of *Calonectria* spp. from *Eucalyptus* in South China, Studies in Mycology (2015), http://dx.doi.org/10.1016/j.simyco.2014.11.003

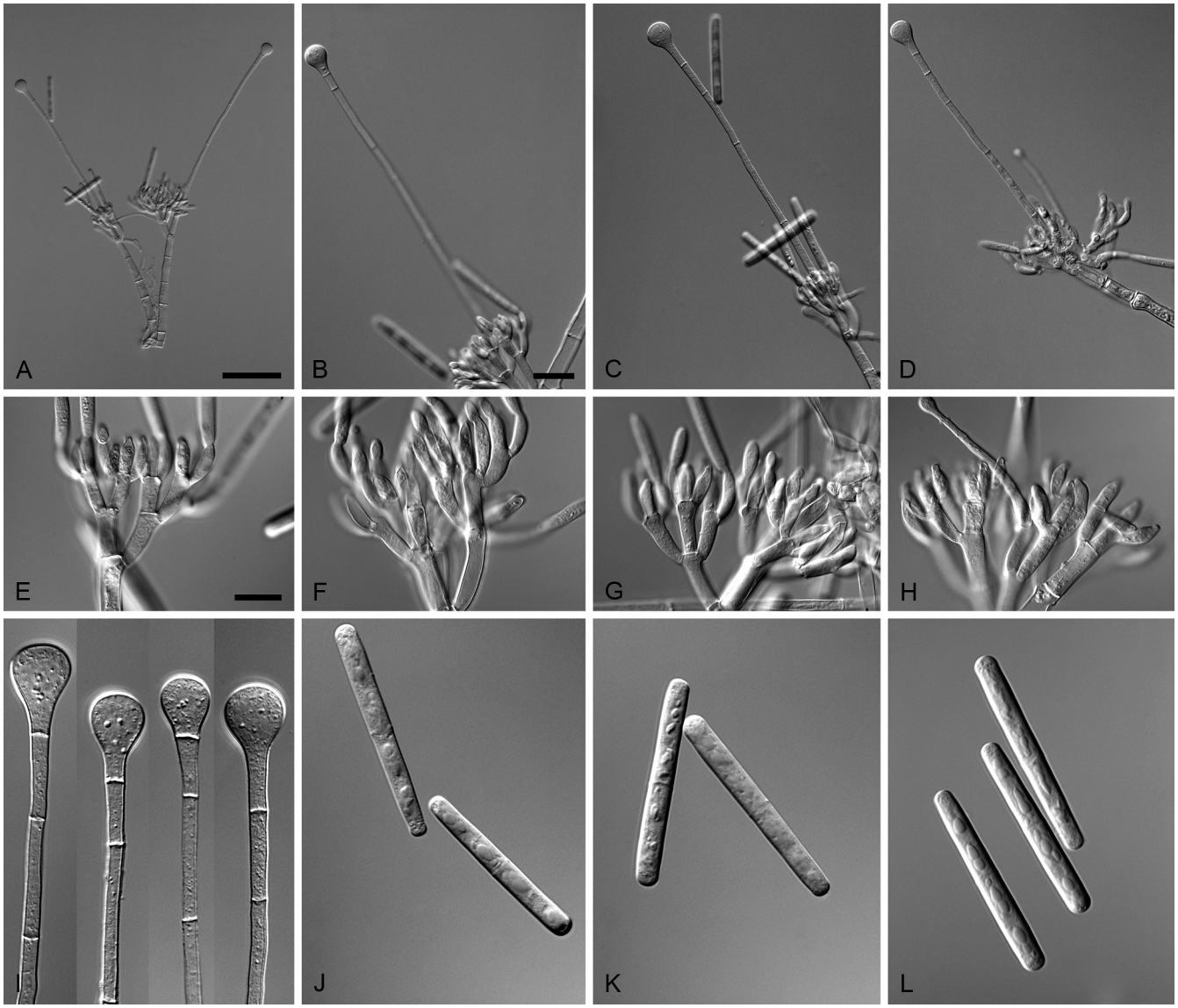


**Fig. 14.** *Calonectria pluriramosa* (ex-type CBS 136976). A–D. Macroconidiophores. E–H. Conidiogenous apparatus with conidiophore branches, doliiform to reniform phialides and lateral stipe extensions. I. Sphaeropedunculate vesicles. J–L. Macroconidia. Scale bars: A = 100 µm; B = 20 µm (apply to C–D); E = 10 µm (apply to F–L).

*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



Please cite this article in press as: Lombard L, et al., New species, hyper-diversity and potential importance of *Calonectria* spp. from *Eucalyptus* in South China, Studies in Mycology (2015), http://dx.doi.org/10.1016/j.simyco.2014.11.003

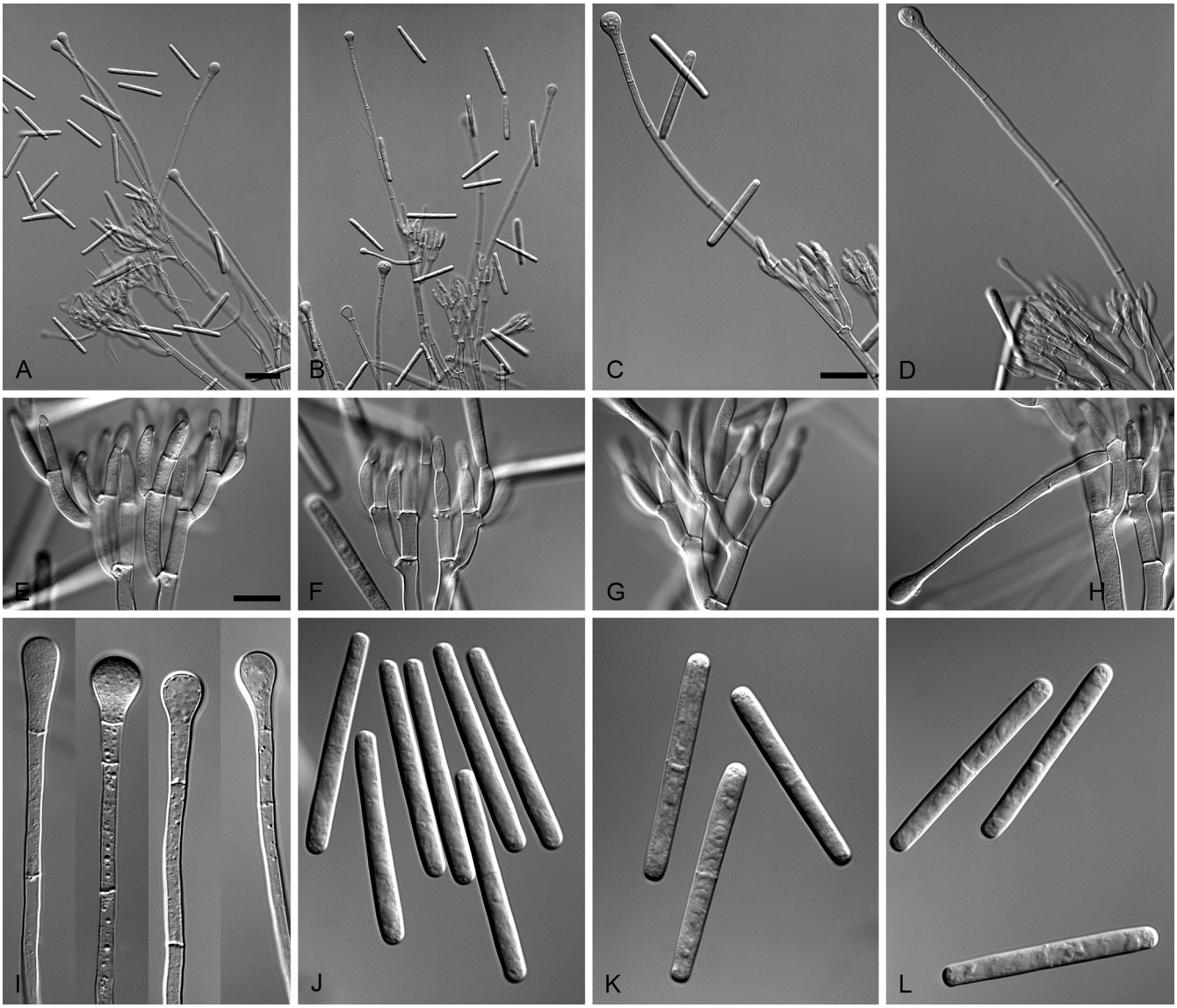


**Fig. 15.** *Calonectria pseudokyotensis* (ex-type CBS 137332). A–D. Macroconidiophores. E–H. Conidiogenous apparatus with conidiophore branches, doliiform to reniform phialides and lateral stipe extensions. I. Pyriform to sphaeropedunculate vesicles. J–L. Macroconidia. Scale bars: A = 50 µm (apply to B); C = 20 µm (apply to D); E = 10 µm (apply to F–L).

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 obpyrifrom 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, straight, (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 H-21475, living ex-type culture 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



Please cite this article in press as: Lombard L, et al., New species, hyper-diversity and potential importance of *Calonectria* spp. from *Eucalyptus* in South China, Studies in Mycology (2015), http://dx.doi.org/10.1016/j.simyco.2014.11.003

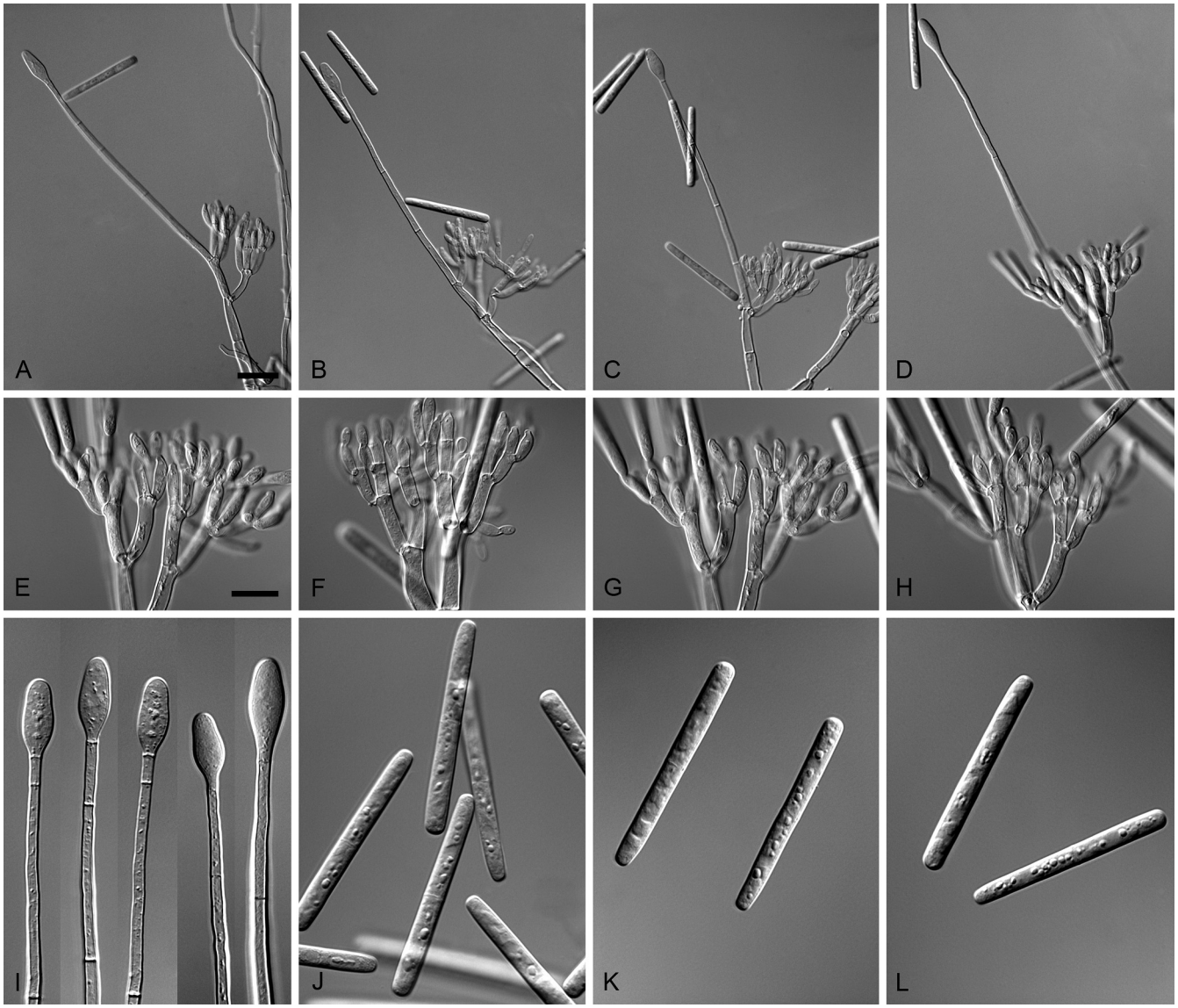


**Fig. 16.** *Calonectria seminaria* (ex-type CBS 136632). A–D. Macroconidiophores. E–H. Conidiogenous apparatus with conidiophore branches and doliiform to reniform phialides. I. Obpyriform to ellipsoid vesicles. J–L. Macroconidia. Scale bars: A = 100 µm (apply to B–D); E = 10 µm (apply to F–L).

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 sphaeropedunculate 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 doliiform 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).



Please cite this article in press as: Lombard L, et al., New species, hyper-diversity and potential importance of *Calonectria* spp. from *Eucalyptus* in South China, Studies in Mycology (2015), http://dx.doi.org/10.1016/j.simyco.2014.11.003

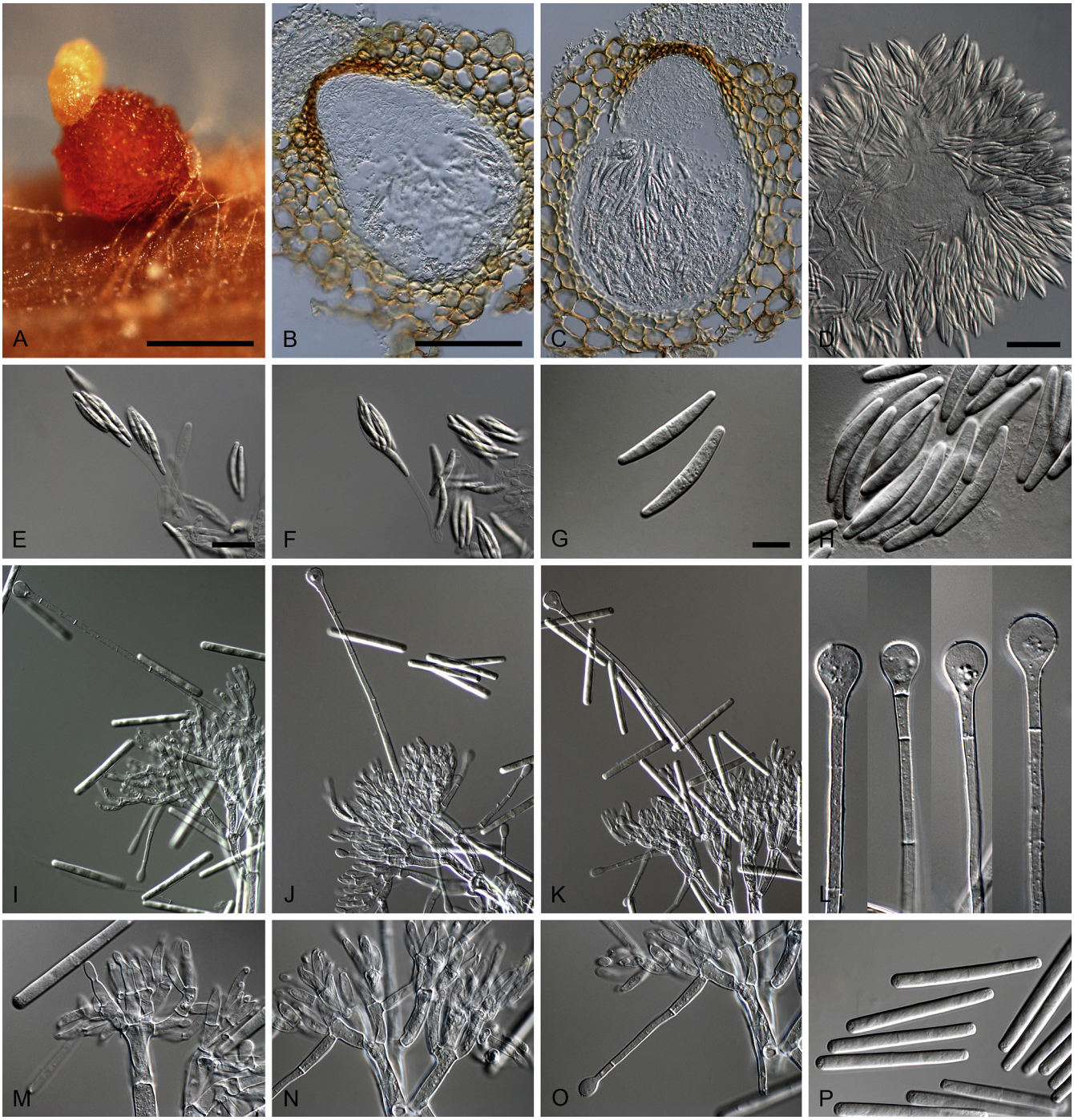


**Fig. 17.** *Calonectria sphaeropedunculata* (ex-type CBS 136081). A. Ascoma. B–C. Vertical section through ascomata, showing wall structure. D–F. Asci. G–H. Ascospores. I–K. Macroconidiophores. L. Sphaeropedunculate vesicles. M–O. Conidiogenous apparatus with conidiophore branches, doliiform to reniform phialides and lateral stipe extension. P. Macroconidia. Scale bars: A = 500 µm; B = 100 µm (apply to C); D = 50 µm (apply to I–K), E = 20 µm (apply to F), G = 10 µm (apply to H, L–P).

*Notes*: *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 obpyrifrom 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



Please cite this article in press as: Lombard L, et al., New species, hyper-diversity and potential importance of *Calonectria* spp. from *Eucalyptus* in South China, Studies in Mycology (2015), http://dx.doi.org/10.1016/j.simyco.2014.11.003

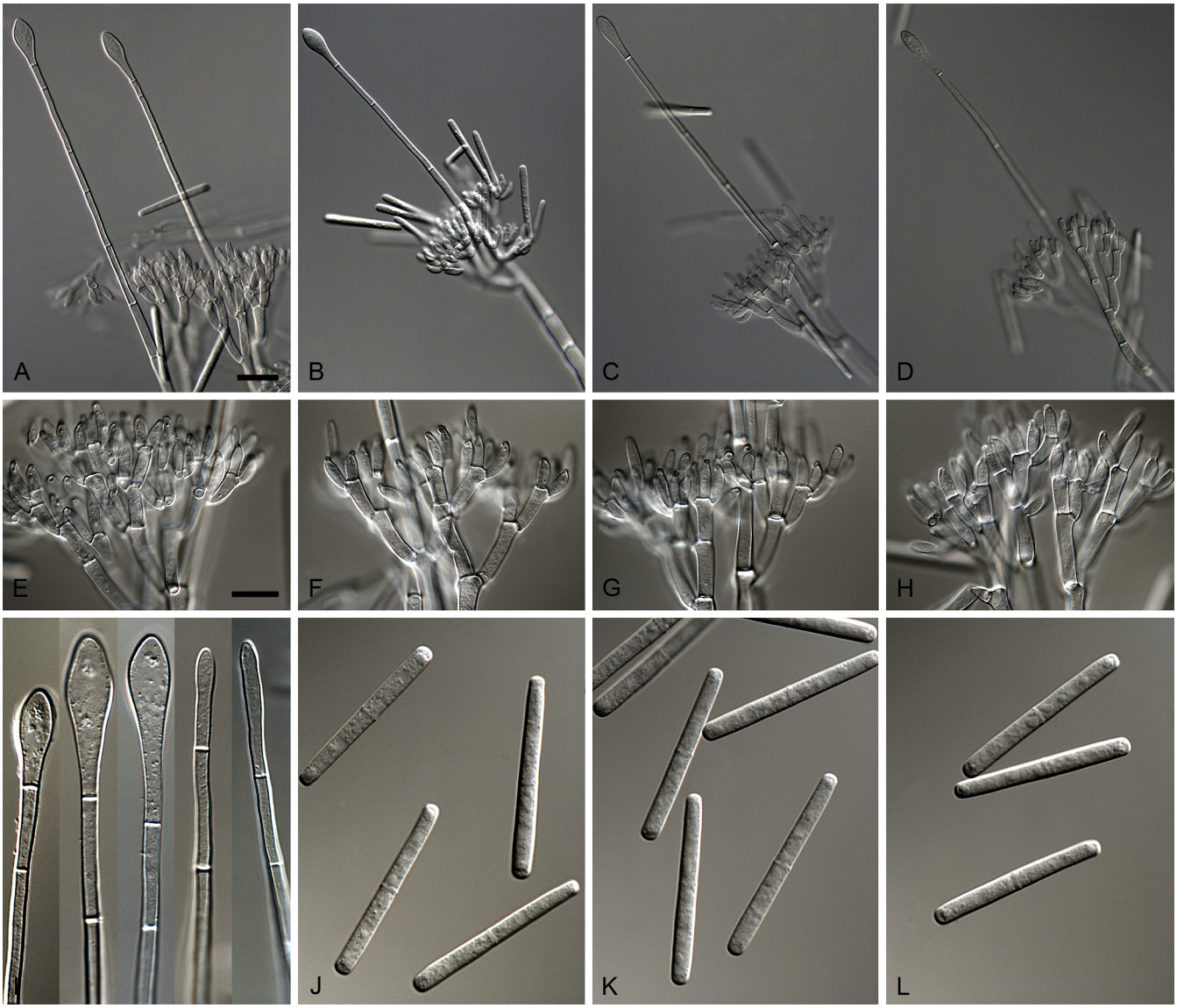


**Fig. 18.** *Calonectria terrestris* (ex-type CBS 136642). A–D. Macroconidiophores. E–H. Conidiogenous apparatus with conidiophore branches and doliiform to reniform phialides. I. Obpyriform to pyriform to broadly clavate vesicles. J–L. Macroconidia. Scale bars: A = 50 µm (apply to B–D); E = 10 µm (apply to F–L).

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 to umber 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 23493 = CMW 35364 = CERC 1868, CBS 136644 = CPC 23494 = CMW 35366 = CERC 1870, CBS 136645 = CPC 23496 = CMW 35178 = CERC 1854, CBS 136651 = CPC 23516 = 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 obpyrifrom 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 a obpyrifrom vesicle, 4–10 µm diam. *Conidiogenous apparatus* 54–95 µm wide, and 36–75 µm long; primary branches aseptate, 15–29 × 4–7 µm; secondary branches aseptate, 10–20 × 3–6 µm; tertiary branches aseptate, 9–15 × 3–6 µm; quaternary branches aseptate, 10–13 × 3–4 µm, each terminal branch producing 2–6 phialides; phialides elongate doliiform to reniform, hyaline, aseptate,



Please cite this article in press as: Lombard L, et al., New species, hyper-diversity and potential importance of *Calonectria* spp. from *Eucalyptus* in South China, Studies in Mycology (2015), http://dx.doi.org/10.1016/j.simyco.2014.11.003

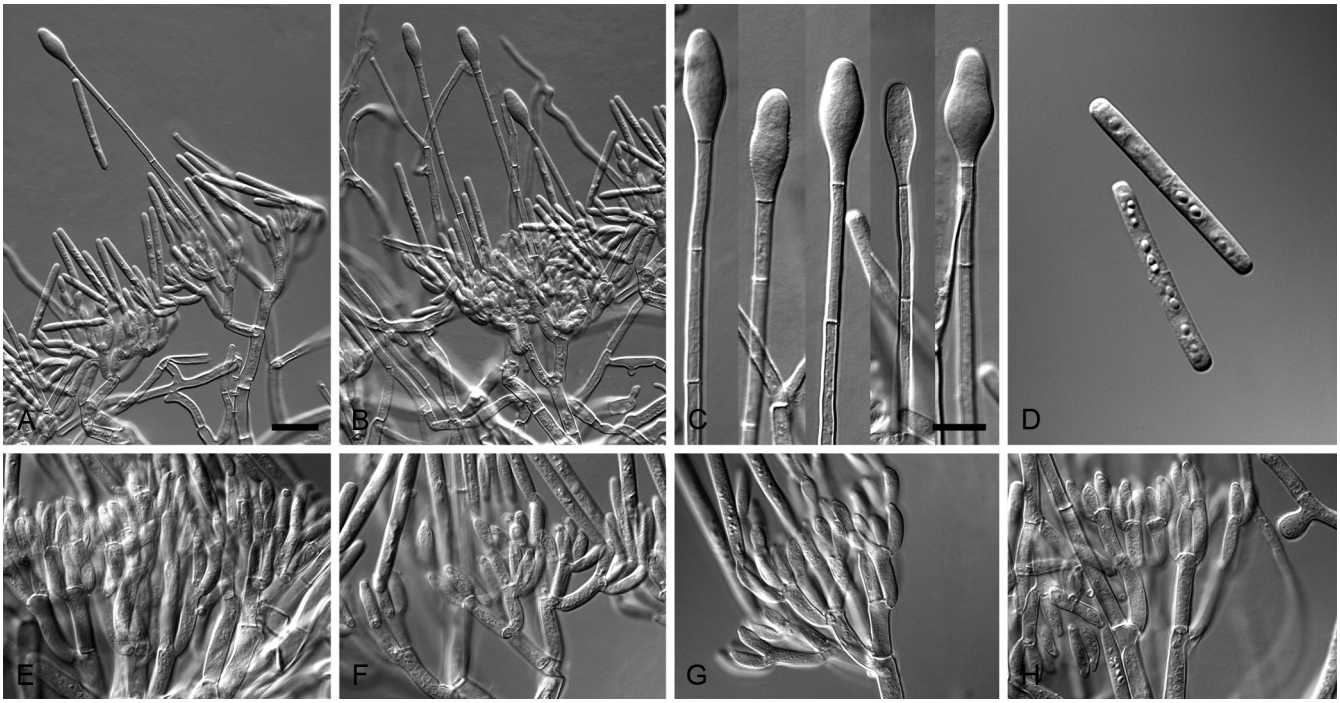


**Fig. 19.** *Calonectria tetraramosa* (ex-type CBS 136635). A–B. Macroconidiophores. C. Obpyriform vesicles. D. Macroconidia. E–H. Conidiogenous apparatus with conidiophore branches and doliiform to reniform phialides. Scale bars: A = 50 µm (apply to B); C = 10 µm (apply to D–H).

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 31476 = CERC 1811.

*Notes*: *Calonectria tetraramosa* 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. tetraramosa* 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. tetraramosa* 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 (Tǔrǎng), 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. Zhou, G. Zhao & F. Han (**holotype** CBS H-21488, culture ex-type CBS 136077 = CPC 23479 = CMW 31411 = CERC 1746); Guangxi Province, from soil collected in a *Eucalyptus* plantation, Aug. 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.



Please cite this article in press as: Lombard L, et al., New species, hyper-diversity and potential importance of *Calonectria* spp. from *Eucalyptus* in South China, Studies in Mycology (2015), http://dx.doi.org/10.1016/j.simyco.2014.11.003

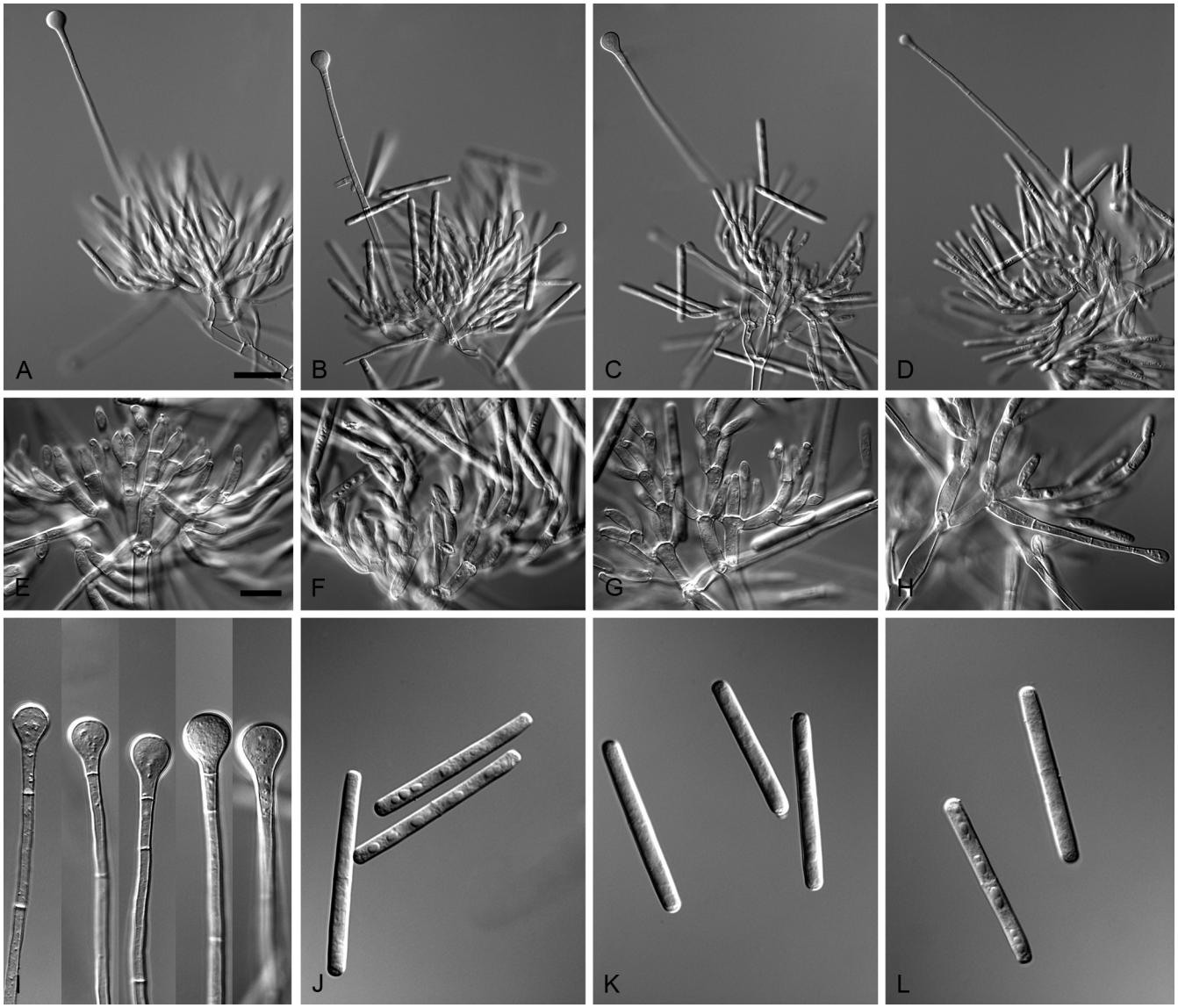


**Fig. 20.** *Calonectria turangicola* (ex-type CBS 136077). A–D. Macroconidiophores. E–H. Conidiogenous apparatus with conidiophore branches, doliiform to reniform phialides and lateral stipe extension. I. Sphaeropedunculate vesicles. J–L. Macroconidia. Scale bars: A = 50 µm (apply to B–D); E = 10 µm (apply to F–L).

*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. terrae-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 (Pitkethley 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



Please cite this article in press as: Lombard L, et al., New species, hyper-diversity and potential importance of *Calonectria* spp. from *Eucalyptus* in South China, Studies in Mycology (2015), http://dx.doi.org/10.1016/j.simyco.2014.11.003

*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, Guarnaccia *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 macroconidiophore 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 sphaeropedunculate 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 distinguished 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. sphaeropedunculata*. 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 Guangxi 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. sphaeropedunculata* 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. sphaeropedunculata* 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 ascomata. 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 *Calonectria* 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



Please cite this article in press as: Lombard L, et al., New species, hyper-diversity and potential importance of *Calonectria* spp. from *Eucalyptus* in South China, Studies in Mycology (2015), http://dx.doi.org/10.1016/j.simyco.2014.11.003

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 South Africa and China, and we are grateful for the funding via projects 2010KJCX015-03 (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 Scientific 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 Cooperative Programme (TPCP). We thank Arien van Iperen, Trix Merkx and Dr Seonju Marincowitz for their invaluable assistance with cultures.

## REFERENCES

Alfenas RF, Pereira OL, Ferreira MA, *et al.* (2013a). *Calonectria metrosideri*, a highly aggressive pathogen causing leaf blight, root rot, and wilt of *Metrosideros* spp. in Brazil. *Forest Pathology* **43**: 257–265.

Alfenas RF, Pereira OL, Jorge VL, *et al.* (2013b). A new species of *Calonectria* causing leaf blight and cutting rot of three forest tree species in Brazil. *Tropical Plant Pathology* **38**: 513–521.

Alfenas RF, Lombard L, Pereira OL, *et al.* (2015). Diversity and potential impact of *Calonectria* species in *Eucalyptus* plantations in Brazil. *Studies in Mycology* **80**. http://dx.doi.org/10.1016/j.simyco.2014.11.002.

Billiard S, López-Villavicencio M, Hood ME, *et al.* (2012). Sex, outcrossing and mating types: unsolved questions in fungi and beyond. *Journal of Evolutionary Biology* **25**: 1020–1038.

Bolland L, Tierney JW, Tierney BJ (1985). Studies on leaf spot and shoot blight of Eucalyptus caused by *Cylindrocladium quinqueseptatum*. *European Journal of Forest Pathology* **15**: 385–397.

Booth TH, Jovanovic T, Old KM, *et al.* (2000). Climatic mapping to identify high-risk areas for *Cylindrocladium quinqueseptatum* leaf blight on eucalypts in mainland South East Asia and around the world. *Environmental Pollution* **108**: 365–372.

Burgess TI, Andjic V, Hardy GESJ, *et al.* (2006). First report of *Phaeophleospora destructans* in China. *Journal of Tropical Forest Science* **18**: 144–146.

Burgess TI, Barber PA, Sufaati S, *et al.* (2007). *Mycosphaerella* spp. on *Eucalyptus* in Asia: new species, new hosts and new records. *Fungal Diversity* **24**: 135–157.

Chen SF, Gryzenhout M, Roux J, *et al.* (2010). Identification and Pathogenicity of *Chrysoporthe cubensis* on *Eucalyptus* and *Syzygium* spp. in South China. *Plant Disease* **94**: 1143–1150.

Chen SF, Pavlic D, Roux J, *et al.* (2011a). Characterization of *Botryosphaeriaceae* from plantation-grown *Eucalyptus* species in South China. *Plant Pathology* **60**: 739–751.

Chen SF, Gryzenhout M, Roux J, *et al.* (2011b). Novel species of *Celoporthe* from *Eucalyptus* and *Syzygium* trees in China and Indonesia. *Mycologia* **103**: 1384–1410.

Chen SF, Barnes I, Chungu D, *et al.* (2011c). High population diversity and increasing importance of the *Eucalyptus* stem canker pathogen, *Teratosphaeria zuluensis*, in South China. *Australasian Plant Pathology* **40**: 407–415.

Chen SF, Lombard L, Roux J, *et al.* (2011d). Novel species of *Calonectria* associated with *Eucalyptus* leaf blight in Southeast China. *Persoonia* **26**: 1–12.

Chen SF, Van Wyk M, Roux J, *et al.* (2013). Taxonomy and pathogenicity of *Ceratocystis* species on *Eucalyptus* trees in South China, including *C. chinaeucensis* sp. nov. *Fungal Diversity* **58**: 267–279.

Crous PW (2002). *Taxonomy and pathology of Cylindrocladium (Calonectria) and allied genera*. APS Press, St. Paul, Minnesota, USA.

Crous PW, Alfenas AC, Wingfield MJ (1993). *Calonectria scoparia* and *Calonectria morganii* sp. nov., and variation among isolates of their *Cylindrocladium* anamorphs. *Mycological Research* **97**: 701–708.

Crous PW, Gams W, Stalpers JA, *et al.* (2004a). MycoBank: an online initiative to launch mycology into the 21st century. *Studies in Mycology* **50**: 19–22.

Crous PW, Groenewald JZ, Risède J-M, *et al.* (2004b). *Calonectria* species and their *Cylindrocladium* anamorphs: species with sphaeropedunculate vesicles. *Studies in Mycology* **50**: 415–430.

Crous PW, Shivas RG, Wingfield MJ, *et al.* (2012). Fungal Planet description sheets: 128–153. *Persoonia* **29**: 146–201.

Crous PW, Wingfield MJ, Guarro J, *et al.* (2013). Fungal Planet description sheets: 154–213. *Persoonia* **31**: 188–296.

Guarnaccia V, Aiello D, Polizzi G, *et al.* (2014). Emergence of plochloraz-resistant populations of *Calonectria pauciramosa* and *Calonectria polizzii* in ornamental nurseries of southern Italy. *Plant Disease* **98**: 344–350.

Heitman J, Sun S, James TY (2013). Evolution of fungal sexual reproduction. *Mycologia* **105**: 1–27.

Hillis DM, Bull JJ (1993). An empirical test of bootstrapping as a method for assessing confidence in phylogenetic analysis. *Systematic Biology* **42**: 182–192.

Kang J-C, Crous PW, Old KM, *et al.* (2001a). Non-conspecificity of *Cylindrocladium quinqueseptatum* and *Calonectria quinqueseptata* based on beta-tubulin gene phylogeny and morphology. *Canadian Journal of Botany* **79**: 1241–1247.

Kang J-C, Crous PW, Schoch CL (2001b). Species concepts in the *Cylindrocladium floridanum* and *Cy. spathiphylli* complexes (Hypocreaceae) based on multi-allelic sequence data, sexual compatibility and morphology. *Systematic and Applied Microbiology* **24**: 206–217.

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

Koike ST, Crous PW (2001). First report of a root and crown rot disease of myrtle in California caused by *Cylindrocladium pauciramosum*. *Plant Disease* **85**: 448.

Koike ST, Henderson DM, Crous PW, *et al.* (1999). A new root and crown rot disease of heath in California caused by *Cylindrocladium pauciramosum*. *Plant Disease* **83**: 589.

Lechat C, Crous PW, Groenewald JZ (2010). The enigma of *Calonectria* species occurring on leaves of *Ilex aquifolium* in Europe. *IMA Fungus* **1**: 101–108.

Lombard L, Crous PW, Wingfield BD, *et al.* (2010a). Multigene phylogeny and mating tests reveal three cryptic species related to *Calonectria pauciramosa*. *Studies in Mycology* **66**: 15–30.

Lombard L, Crous PW, Wingfield BD, *et al.* (2010b). Phylogeny and systematics of the genus *Calonectria*. *Studies in Mycology* **66**: 31–69.

Lombard L, Crous PW, Wingfield BD, *et al.* (2010c). Species concepts in *Calonectria* (*Cylindrocladium*). *Studies in Mycology* **66**: 1–14.

Lombard L, Merwe NA van der, Groenewald JZ, *et al.* (2015). Generic concepts in *Nectriaceae*. *Studies in Mycology* **80**. http://dx.doi.org/10.1016/j.simyco.2014.12.002.



Please cite this article in press as: Lombard L, et al., New species, hyper-diversity and potential importance of *Calonectria* spp. from *Eucalyptus* in South China, Studies in Mycology (2015), http://dx.doi.org/10.1016/j.simyco.2014.11.003

Lombard L, Polizzi G, Guarnaccia V, *et al*. (2011). *Calonectria* spp. causing leaf spot, crown and root rot of ornamental plants in Tunisia. *Persoonia* **27**: 73–79.

Lombard L, Zhou XD, Crous PW, *et al*. (2010d). *Calonectria* species associated with cutting rot of *Eucalyptus*. *Persoonia* **24**: 1–11.

Nirenburg HI (1981). A simplified method for identifying *Fusarium* spp. occurring on wheat. *Canadian Journal of Botany* **59**: 1599–1609.

Nylander JAA (2004). *MrModeltest v. 2. Programme distributed by the author*. Evolutionary Biology Centre, Uppsala University.

Old KM, Wingfield MJ, Yuan ZQ (2003). *A manual of diseases of eucalypts in South-East Asia*. Center for International Forestry Research, Jakarta, Indonesia.

Pitkethley RN (1976). *Cylindrocladium quinqueseptatum* on myrtaceous tree seedlings. *Australian Plant Pathology Society Newsletter* **5**: 57.

Polizzi G (2000). Prime esperience di lotta chimica nei confrontidel marciume del colletto e delle radici di *Polygala myrtifolia* causato da *Cylindrocladium pauciramosum*. *Informatore Fitopatologico* **11**: 39–47.

Polizzi G, Catara V (2001). First report of leaf spot caused by *Cylindrocladium pauciramosum* on *Acacia retinodes*, *Arbutus unedo*, *Feijoa sellowiana* and *Dodenaea viscosa* in Southern Italy. *Plant Disease* **85**: 803.

Polizzi G, Crous PW (1999). Root and collar of milkwort caused by *Cylindrocladium pauciramosum*, a new record for Europe. *European Journal of Plant Pathology* **105**: 407–411.

Polizzi G, Grasso FM, Vitale A, *et al*. (2007). First occurrence of *Calonectria* Leaf Spot on Mexican Blue Palm in Italy. *Plant Disease* **91**: 1057.

Polizzi G, Vitale A (2001). First report of the prevalence of benzimidazole-resistant isolates in a population of *Cylindrocladium pauciramosum* in Italy. *Plant Disease* **85**: 1210.

Polizzi G, Vitale A, Aiello D, *et al*. (2006). First record of crown and root rot caused by *Cylindrocladium pauciramosum* on California Lilac in Italy. *Plant Disease* **90**: 1459.

Polizzi G, Vitale A, Aiello D, *et al*. (2009). First record of crown and root rot caused by *Cylindrocladium pauciramosum* on bush cherry in Italy. *Plant Disease* **93**: 547.

Rayner RW (1970). *A mycological colour chart*. Commonwealth Mycological Institute, Kew, Surrey. British Mycological Society.

Rodas CA, Lombard L, Gryzenhout M, *et al*. (2005). *Cylindrocladium* blight of *Eucalyptus grandis* in Colombia. *Australasian Plant Pathology* **34**: 134–149.

Ronquist F, Huelsenbeck JP (2003). MrBayes 3: Bayesian phylogenetic inference under mixed models. *Bioinformatics* **19**: 1572–1574.

Rowe RC, Beute MK (1975). Ascospore formation and discharge by *Calonectria crotalariae*. *Phytopathology* **65**: 393–398.

Schoch CL, Crous PW, Polizzi G, *et al*. (2001). Female fertility and single nucleotide polymorphism comparisons in *Cylindrocladium pauciramosum*. *Plant Disease* **85**: 941–946.

Schoch CL, Crous PW, Wingfield BD, *et al*. (1999). The *Cylindrocladium candelabrum* species complex includes four distinct mating populations. *Mycologia* **91**: 286–298.

Sharma JK, Mohanan C (1991). Pathogenic variation in *Cylindrocladium quinqueseptatum* causing leaf blight of *Eucalyptus*. *European Journal of Forest Pathology* **21**: 210–217.

Sharma JK, Mohanan C (1992). Relative susceptibility of *Eucalyptus* provenances to *Cylindrocladium* leaf blight in Kerala, India. *European Journal of Forest Pathology* **22**: 257–265.

Sharma JK, Mohanan C, Maria Florence EJ (1984). Nursery diseases of *Eucalyptus* in Kerala. *European Journal of Forest Pathology* **14**: 77–89.

Swofford DL (2003). *PAUP*. Phylogenetic analysis using parsimony (*and other methods), v. 4.0b10*. Computer programme. Sinauer Associates, Sunderland, Massachusetts, USA.

Tamura K, Peterson D, Peterson N, *et al*. (2011). MEGA5: molecular evolutionary genetics analysis using maximum likelihood, evolutionary distance, and maximum parsimony methods. *Molecular Biology and Evolution* **28**: 2731–2739.

Turnbull JW (2007). *Development of sustainable forestry plantations in China: a review*. In: *Impact assessment series Report No. 45*. Australian Centre for International Agricultural Research, Canberra, Australia.

Vitale A, Aiello D, Castello I, *et al*. (2009). Severe outbreak of crown rot and root rot caused by *Cylindrocladium pauciramosum* on strawberry tree in Italy. *Plant Disease* **93**: 842.

Vitale A, Crous PW, Lombard L, *et al*. (2013). *Calonectria* diseases on ornamental plants in Europe and the Mediterranean basin: an overview. *Journal of Plant Pathology* **95**: 463–476.

Wingfield MJ, Roux J, Slippers B, *et al*. (2013). Established and new technologies reduce increasing pests and pathogen threats to eucalypt plantations. *Forest Ecology and Management* **301**: 35–42.

Wingfield MJ, Slippers B, Wingfield BD (2010). Novel associations between pathogens, insects and tree species threaten world forests. *New Zealand Journal of Forestry Science* **40**(Suppl.): S95–S103.

Wright LP, Davis AJ, Wingfield BD, *et al*. (2010). Population structure of *Cylindrocladium parasiticum* infecting peanuts (*Arachis hypogaea*) in Georgia, USA. *European Journal of Plant Pathology* **127**: 199–206.

Wright LP, Wingfield BD, Crous PW, *et al*. (2006). Isolation and characterization of microsatellite loci in *Cylindrocladium parasiticum*. *Molecular Ecology Notes* **6**: 110–112.

Wright LP, Wingfield BD, Crous PW, *et al*. (2007). Isolation and characterization of microsatellite loci in *Cylindrocladium pauciramosum*. *Molecular Ecology Notes* **7**: 343–345.

Xu J-J, Qin S-Y, Hao Y-Y, *et al*. (2012). A new species of *Calonectria* causing leaf disease of water lily in China. *Mycotaxon* **122**: 177–185.

Zhou XD, Beer ZW De, Xie Y, *et al*. (2007). DNA-based identification of *Quambalaria piterka* causing severe leaf blight of *Corymbia citriodora* in China. *Fungal Diversity* **25**: 245–254.

Zhou XD, Xie YJ, Chen SF, *et al*. (2008). Diseases of eucalypt plantation in China: challenges and opportunities. *Fungal Diversity* **32**: 1–7.

Zhou X, Wingfield MJ (2011). Eucalypt diseases and their management in China. *Australasian Plant Pathology* **40**: 339–345.



Please cite this article in press as: Lombard L, et al., New species, hyper-diversity and potential importance of *Calonectria* spp. from *Eucalyptus* in South China, Studies in Mycology (2015), http://dx.doi.org/10.1016/j.simyco.2014.11.003