



Identification and characterization of *Colletotrichum* species associated with ornamental plants in Southern China

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Abstract

Colletotrichum is an important plant pathogenic genus with a wide range of hosts. *Colletotrichum* species can infect various plants and cause diseases, leading to serious economic losses. Ornamental plants are an important commercial crop with high aesthetic value and emerging diseases have become a serious problem threatening the ornamental plant industry. However, little is known about the fungi and fungal species associated with ornamental plants in China especially latent pathogens such as *Colletotrichum* spp. In the present study, 45 *Colletotrichum* isolates were obtained from 17 ornamental plants with typical symptoms including leaf spot and stem blight. These isolates were further identified based on morphological analysis, multigene molecular phylogenetic analysis of the internal transcribed spacer regions (ITS), actin (*act*), partial sequences of the chitin synthase 1 (*chs-1*), glyceraldehyde 3-phosphate dehydrogenase (*gapdh*), and β -tubulin 2 (*tub2*) genes, and pairwise homoplasy index (PHI) analysis. Based on multigene phylogenetic analysis and morphology 11 species of *Colletotrichum* were identified belonging to five species complexes: *C. acutatum*, *C. boninense*, *C. gloeosporioides*, *C. orchidearum* and *C. truncatum*. Among these complexes, one species was described as new species namely, *C. chrysalidocarpi*. In addition, *C. dimorphum*, and *C. nanhuaense* are reduced as synonyms of *C. gloeosporioides* and *C. orientale* and *C. radermacheriae* are reduced as synonyms of *C. fioriniae*. Furthermore, 18 new host records were identified and described. This is the first comprehensive study on *Colletotrichum* species associated with ornamental plants in South China. Our results suggested a high *Colletotrichum* species diversity on ornamental plants. These findings enhance the current knowledge of *Colletotrichum*, and its diversity and expand the host range. In addition, these results will help to early diagnose, and control diseases caused by *Colletotrichum* species.

Keywords – 1 new species – 18 new host records – multi-locus phylogenetic analysis – taxonomy

Introduction

Ornamental plants have high economic value and aesthetic value. They include cut flowers, cut foliage crops, potted plants, bulb and corm crops, and woody ornamentals (Lecomte et al. 2016). China is one of the largest producers of ornamental plants. The export value of ornamental flowers accounted for 4.77 hundred billion USD in China in 2022 (Wu et al. 2023). Guangdong, Yunnan, and Fujian provinces are the largest ornamental plant exporters in China, with 69.96% of the total export value being from these three provinces (Wu et al. 2023). However, with the expansion of global trade and the growing cultivation area, diseases are becoming a serious limiting factor in the ornamental industry. In the last few years, many destructive diseases have been reported from various countries on ornamental plants, such as Fusarium wilt or rot caused by *Fusarium* and allied fusarioid taxa (Kamali-Sarvestani et al. 2022, Zhang et al. 2022, Chen et al. 2023), anthracnose caused by *Colletotrichum* spp. (Guarnaccia et al. 2021), and leaf blight and crown rot caused by *Calonectria* spp. (Aiello et al. 2022). These diseases pose a significant threat to ornamental plant production.

Colletotrichum (Glomerellaceae, Sordariomycetes, Ascomycota) is one of the most common and important phytopathogenic genera. This genus was listed in the top 10 fungal pathogens worldwide (Dean et al. 2012). Species of *Colletotrichum* have a wide host range, including fruit trees (Huang et al. 2013, Lima et al. 2013), vegetables (Than et al. 2008), and ornamental plants (Diao et al. 2017, Guarnaccia et al. 2021, Zakaria 2021, Manova et al. 2022). Many *Colletotrichum* species are well-known causal organisms for most destructive diseases (Yan et al. 2015, Diao et al. 2017, Manova et al. 2022). They can infect aerial plant tissues and cause leaf spots, stem blight, and fruit rot. Especially, during the ripening stage, they can infect fruits and lead to high yield losses (Zakaria 2021). Most tropical and subtropical fruit crops, such as mango, dragon fruit, litchi, papaya, avocado, grape, and apple are susceptible to this genus (Zakaria 2021). For example, grape ripe rot, which is a notorious disease, is caused by over ten *Colletotrichum* species belonging to three *Colletotrichum* species complexes (*C. gloeosporioides*, *C. acutatum*, and *C. boninense*) worldwide (Yan et al. 2015, Echeverrigaray et al. 2020, Batista et al. 2023, Ye et al. 2023). In some countries the disease incidence rate can be over 30%, or even up to 90% (Lei et al. 2022, Batista et al. 2023), causing huge economic losses. Furthermore, anthracnose caused by *Colletotrichum* species is one of the most damaging diseases in vegetable crop production. For instance, 14 *Colletotrichum* species from *Capsicum* spp. and six species from Solanaceous crops have been reported to be associated with this disease (Than et al. 2008, Diao et al. 2017, Manova et al. 2022).

Apart from phytopathogens, *Colletotrichum* species also occur as endophytes and saprobes. In the last 20 years, many studies have focused on endophytic *Colletotrichum*, especially those species involved in medical plants or commercial crops (Lima et al. 2012, Peng et al. 2012, Vieira et al. 2014, Rai et al. 2014, Ma et al. 2018, Zhang et al. 2023). In recent years, number of new *Colletotrichum* species have been introduced as endophytes (Tao et al. 2013, Ma et al. 2018, Liu et al. 2023b, Zhang et al. 2023). It has been mentioned that these endophytic *Colletotrichum* species play an important role in promoting plant growth, protecting the host from adverse environmental conditions and or sometimes as a potential pathogen (Photita et al. 2004).

Studies on *Colletotrichum* species associated with ornamental plants are limited. Few studies have been focused on pathogenic *Colletotrichum* species (Guarnaccia et al. 2019, Silva-Cabral et al. 2019, Guarnaccia et al. 2021, Yu et al. 2022a). Guarnaccia et al. (2019) studied anthracnose causing *Colletotrichum* species diversity in ornamental *Lamiaceae* plants and identified nine species belonging to three species complexes. Guarnaccia et al. (2021) isolated and identified seven *Colletotrichum* species belonging to four species complexes from symptomatic ornamental plants in northern Italy. From *Orchidaceae* hosts in China, Ma et al. (2018) isolated and identified 10 species with five new taxa and Yang et al. (2011) described eight endophytic *Colletotrichum* species with one novel taxa. Further, 17 endophytic *Colletotrichum* species were isolated, and seven species were introduced as novel species from *Bletilla ochracea* (*Orchidaceae*) (Tao et al. 2013).

Even though *Colletotrichum* species are frequently isolated from various hosts, identification and species delineation are still challenging. With the implementation of the polyphasic approaches in species delineation, new species have been continuously discovered and the number of species in *Colletotrichum* has been continuously updated (Jayawardena et al. 2021a, Yu et al. 2022b, Armand et al. 2023, Liu et al. 2023c, Zhang et al. 2023). For example, 119 species were accepted by Cannon et al. (2012), 189 species were accepted by Jayawardena et al. (2016), 248 species by Jayawardena et al. (2021a) and 280 species by Liu et al. (2022). Since then, more than 20 novel taxa have been described (Yu et al. 2022b, Armand et al. 2023, Liu et al. 2023b, c, Peng et al. 2023, Zhang et al. 2023). However, it is necessary to consider the higher within-species diversity of *Colletotrichum* species (Mahmodi et al. 2014, Liu et al. 2023a) while introducing a novel species.

The present study was initiated to understand the diversity of *Colletotrichum* species in various ornamental plants grown in South China. Diseased samples with typical symptoms such as leaf spot, and stem blight were collected from 2020 to 2023 in South China. Fungal species were identified based on multigene phylogeny and morphological characteristics. In addition, updated multigene phylogenetic trees for five *Colletotrichum* species complexes are given. Our results will provide a baseline for the diagnosis and control of various fungal diseases on ornamental plants, especially those caused by *Colletotrichum* species.

Materials & Methods

Sample collection and isolation

From 2020 to 2023, plant tissues showing leaf spot and stem blight were collected from different locations in Guangdong Provinces, Yunnan Provinces, Hunan Provinces, and Shanghai City (Fig. 1). Photographs of symptoms were taken, and sample information including habitat, host, collection site, collector, and collection date was recorded. All samples were taken back to the laboratory for further study.

Fungi were isolated using the tissue isolation method (Senanayake et al. 2020). Small tissue pieces, cut from the margins of the symptoms to include both healthy and diseased parts, were surface sterilized for 10 to 15 seconds using 70% ethanol and then for 30 to 40 seconds using 1% sodium hypochlorite (NaClO). The pieces were then washed in sterile water and dried on sterile filter paper placed on plates of potato dextrose agar (PDA), and then incubating the plates at 25 °C. Pure cultures were obtained by single-spore and single-hyphal tip methods (Senanayake et al. 2020). All living cultures were deposited in the culture collection of Zhongkai University of Agriculture and Engineering (ZHKUCC) and dried cultures were deposited in the herbarium (MHZU).

Morphological characteristics observation

Mycelial plugs (5 diam.) of representative strains (5 replicates of each) were placed on fresh PDA plates and incubated at 25 °C under 12 h light/12 h dark regime. After seven days, colony diameter was measured, and the growth rate was calculated. Colony colour and textures were observed and recorded (Rayner 1970). Asexual structures (conidiomata, conidiophores and conidia) and sexual structures (ascmata, asci and ascospores) were photographed and recorded. Synthetic nutrient-poor agar (SNA) was used for those strains which did not produce conidia on PDA. Slide culture techniques were used to induce appressoria (Johnston & Jones 1997, Cai et al. 2009). Conidia were inoculated onto the edge of 10 mm² PDA plugs, which were then covered with a sterile coverslip and placed in a Petri dish. The shape, colour, and size of the appressoria were recorded. Micro-morphological structures were examined using a Nikon Eclipse 80i microscope (Nikon, Tokyo, Japan) and measured using NISElements BR 3.2. Fifty spores (conidia and/or ascospores) and 30 appressoria for each isolate were measured. The Cnoptec SZ650 (Chongqing Optec Instrument Co., Chongqing, China) series stereomicroscope was used to observe macro-morphological characteristics.

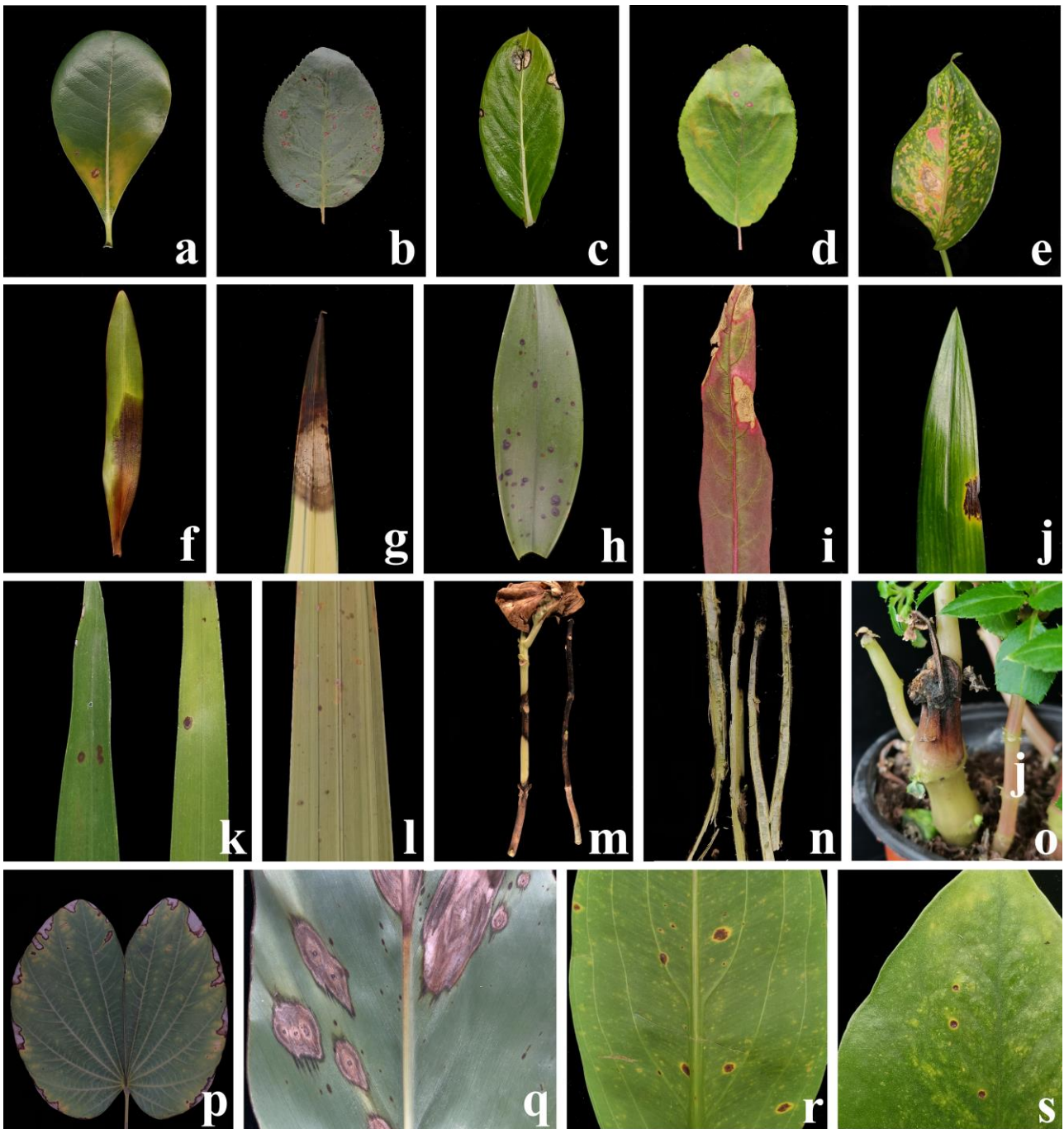


Figure 1 – Field symptoms on leaves and stems of diseased plants collected in this study. a *Pittosporum tobira*. b *Rosa chinensis*. c *Catharanthus roseus*. d *Malus spectabilis*. e *Aglaonema* sp. f *Dendrobium nobile*. g *Cymbidium sinense*. h *Dendrobium nobile*. i *Celosia cristata*. j *Cymbidium sinense*. k, l *Chrysalidocarpus lutescens*. m, n Stems of *Hydrangea macrophylla*. o Stem of *Impatiens balsamina*. p *Bauhinia blakeana*. q *Alpinia zerumbet*. r Leaf of *Thalia dealbata*. s *Epipremnum aureum*.

DNA extraction and PCR amplification

Mycelia derived from five-days-old cultures were used for DNA extraction. Genomic DNA was extracted according to the manufacturer's instructions given in the genomic DNA Extraction Kit (Aidlab Biotechnologies Co., Beijing, China). Five loci, the internal transcribed spacer regions (ITS), actin (*act*), partial sequences of the chitin synthase 1 (*chs-1*), glyceraldehyde 3-phosphate dehydrogenase (*gapdh*), and β -tubulin 2 (*tub2*), were selected for PCR amplification. The primers of each locus were ITS4 and ITS5 for ITS (White 1990), *act*-512F and *act*-783R (Carbone & Kohn

1999), 79F and 345R for *chs-1* (Carbone & Kohn 1999), T1 (O'Donnell & Cigelnik 1997) and Bt2b (Glass & Donaldson 1995). The PCR was amplified following the methods of Damm et al. (2009, 2012a, b, 2019), Jayawardena et al. (2016) and Liu et al. (2022). The PCR products were sequenced by Guangzhou Tianyi Science and Technology Co. (Guangzhou, China). All sequence data generated in this study were submitted to NCBI GenBank (Supplementary Table 1).

Phylogenetic analysis

For the phylogenetic analysis, reference sequences of *Colletotrichum* species were downloaded from NCBI following Jayawardena et al. (2021a), Liu et al. (2022), and Zhang et al. (2023). Analysed sequences were aligned using MAFFT v. 7 (<https://mafft.cbrc.jp/alignment/server/>). Alignments were checked and improved manually when necessary, using BioEdit 7.0.5.2 (Hall 1999). Phylogenetic trees were constructed for concatenated datasets of ITS, *gapdh*, *chs-1*, *act* and *tub2* sequences. Phylogenetic analyses used maximum likelihood (ML) and Bayesian posterior probability analysis (BYPP) on the CIPRES science gateway platform (<http://www.phylo.org>). The ML analysis was performed using the RAxML-HPC2 on XSEDE (8.2.12) (Stamatakis et al. 2008). The GTR + I + G evolution model was used with 1000 non-parametric bootstrapping iterations. The BI analysis was performed in MrBayes (v3.0b4) (Ronquist & Huelsenbeck 2003). Bayesian analysis was performed with six simultaneous Markov chains run for 2,000,000 generations. Trees were sampled every 100 generations. The phylogram was viewed in FigTree v. 1.4.0 and edited in Adobe Illustrator CC 2019 software (Adobe Systems Inc.).

Pairwise homoplasy index (PHI)

The pairwise homoplasy index (PHI index) test was conducted for new taxa identification using SplitsTree4 v. 4 (Huson & Bryant 2006). The PHI was calculated to evaluate the relationship between our new taxa and closely related species. The PHI over 0.05 ($P > 0.05$) indicated no significant recombination in the dataset. The concatenated dataset of ITS, *gapdh*, *chs-1*, *act* and *tub2* was used for the analyses. Relationships between novel species and their closely related taxa were visualized as split graphs via Log-Det transformation and split decomposition options.

Results

Forty-five isolates of *Colletotrichum* were obtained from 18 host species. Preliminary species identification was done based on BLASTn results of all gene regions. Based on the BLASTn results single gene phylogenetic trees were constructed and confirmed that our isolates belonged to five species complexes namely *C. acutatum*, *C. boninense*, *C. gloeosporioides*, *C. orchidearum* and *C. truncatum*. For the taxonomic treatment of *Colletotrichum*, we followed Jayawardena et al. (2021a), Liu et al. (2022) and Zhang et al. (2023). Updated phylogenetic trees and species descriptions are given below under each species complex.

Colletotrichum acutatum species complex

Phylogenetic analysis

Phylogenetic trees were generated using combined ITS (542 bp), *gapdh* (234 bp), *chs-1* (251 bp), *act* (224 bp), and *tub2* (493 bp) sequence data. The tree topologies generated by ML and Bayesian analysis were similar. The best-scoring ML tree is shown in Fig. 2. The sequence alignment of the *C. acutatum* species complex comprised 67 taxa of representative strains, including five isolates obtained from this study. *Colletotrichum orchidophilum* (CBS 632.80 and CBS 631.80) were used as the outgroup. The best-scoring ML tree had an optimization likelihood value of -7206.139499. The matrix had 524 distinct alignment patterns with a 6.03% proportion of gaps and completely undetermined characters. Estimated base frequencies were as follows: A = 0.232812, C = 0.301166, G = 0.240003, T = 0.226019; substitution rates: AC = 1.648307, AG = 5.190572, AT = 1.442542, CG = 0.742996, CT = 7.975627, GT = 1.000000; gamma distribution

shape parameter $\alpha = 0.813331$. Incomplete portions at the ends of the sequences were excluded from the analysis. Five isolates from this study formed two distinct clades.

Two isolates from our collection constituted a sister relationship to *C. eriobotryae* with 100% ML and 1.00 Bayesian posterior probabilities (BYPP) support. The other three isolates developed a cluster with *C. radermacheriae*, *C. fiorinia* and *C. orientale* (Fig. 2). Within the same cluster, these three species and our isolates developed distinct evolutionary lengths. Based on phylogenetic analysis, morphological comparisons and pairwise nucleotide comparisons, our collections were identified as one novel species and a new record of *C. fiorinia* in *C. acutatum* species complex. Species descriptions with illustrations are given below.

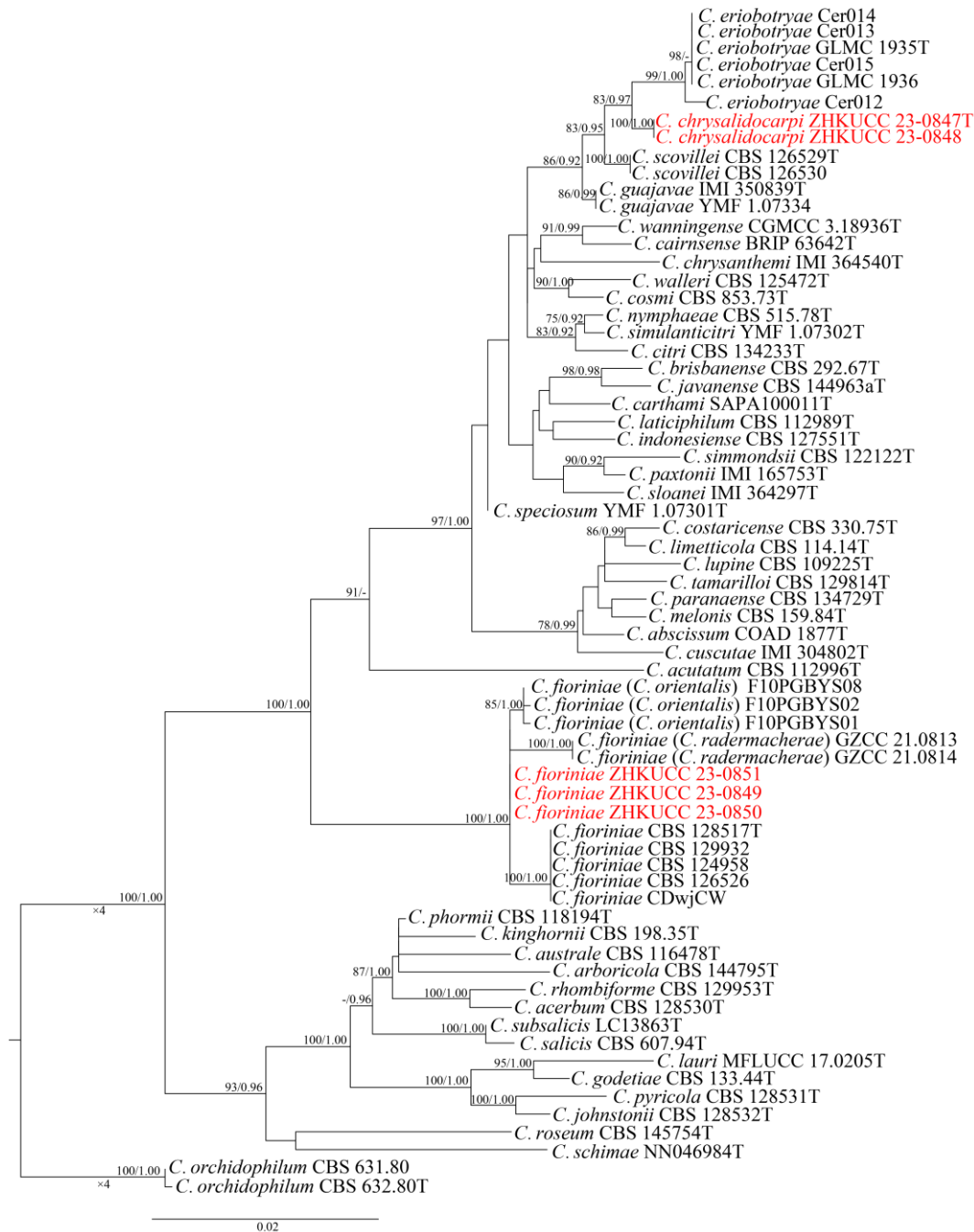


Figure 2 – Maximum likelihood tree of the *Colletotrichum acutatum* species complex. *Colletotrichum orchidophilum* (CBS 632.80 and CBS 631.80) were selected as the outgroup. At the nodes, bootstrap support values for ML ($\geq 75\%$) and BYPP (≥ 0.95) are displayed (ML/PP). Some branches were shortened to fit them to the page, Ex-type isolates are marked with “T”, and the new isolates from this study are in red. The scale bar indicates 0.02 nucleotide changes per site.

Taxonomy

Colletotrichum chrysalidocarpi Y.X. Zhang, J.W. Chen & Manawas., sp. nov.

Fig. 3

Index Fungorum number: IF900955; Facesoffungi number: FoF 14668

Etymology – Named after the host genus, *Chrysalidocarpus*, on which it was found.

Holotype – MHZU 23-0206

Associated with leaf spot of *Chrysalidocarpus lutescens*. Sexual morph: Not observed. Asexual morph: *Mycelium* 1–1.5 μm diam., hyaline to pale brown, smooth-walled, septate, branched. *Conidiomata* dark grey, scattered or in groups. *Setae* not observed. *Conidiophores* hyaline to pale brown, smooth-walled to verruculose, septate, branched. *Conidiogenous cells* 10–14(–17) \times 3–5 μm (\bar{x} = 13 \times 4 μm , n = 50), hyaline, smooth-walled, clavate. *Conidia* 13–17 \times 4–6 μm (\bar{x} = 15 \times 5 μm , n = 50), hyaline, smooth-walled, aseptate, straight, clavate, ends rounded. *Appressoria* 6–10(–14) \times 4–6(–8) μm (\bar{x} = 9 \times 6 μm , n = 30), single or in loose groups, medium to dark brown, smooth-walled, ovoid to ellipsoidal, outline entire.

Culture characteristics: Colonies on PDA 50 mm diam. after 7 days of growth at 25 °C. The colony is grey-white, the edge regular, rounded, mycelium lush, velvet, reverse dark green in the center, conidial masses orange.

Material examined – China, Guangdong Province, Guangzhou City, on leaf spot of *Chrysalidocarpus lutescens* H. Wendl (*Arecaceae*), April 2022, Yanhong Lin, holotype dried culture MHZU 23-0206; ex-type, living culture ZHKUCC 23-0847.

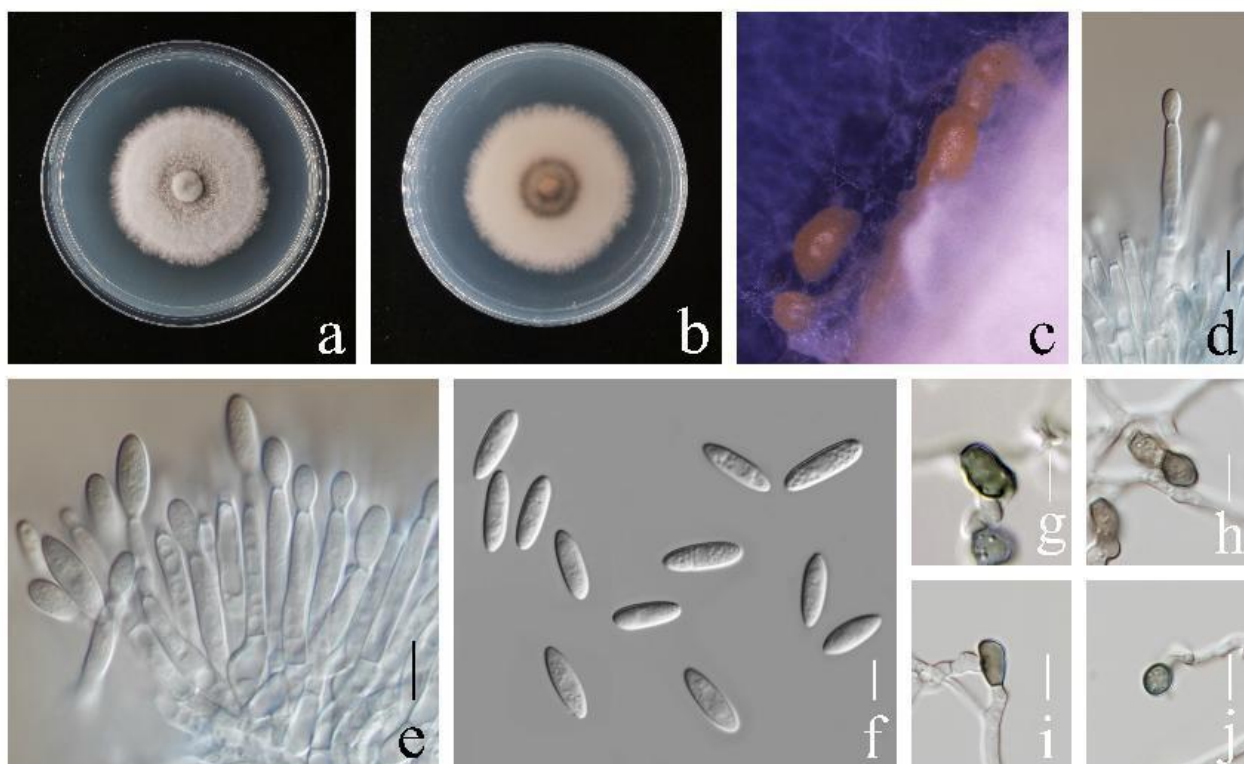


Figure 3 – *Colletotrichum chrysalidocarpi* (MHZU 23-0206, holotype). a, b Upper and reverse view on PDA (7 d). c Conidial masses. d, e Conidiogenous cells. f Conidia. g–j Appressoria. Scale bars: d–j = 10 μm .

Note – In the phylogenetic analysis, two isolates developed a distinct lineage sister to *C. eriobotryae* (83% in ML, 0.97 in BYPP) and *C. scovillei* (83% in ML, 0.95 in BYPP) (Fig. 2). Morphologically, *C. chrysalidocarpi* has larger conidia (13–17 \times 4–6 μm , \bar{x} = 15 \times 5 μm) than either *C. eriobotryae* (12–16 \times 3–4 μm , \bar{x} = 13.4 \times 4.0 μm) or *C. scovillei* (13–15 \times 3.5–4 μm , \bar{x} = 13.7 \times 3.8 μm) (Damm et al. 2012a, 2020). The nucleotide differences between *C. eriobotryae*

(GLMC 1935) and *C. chrysalidocarpi* (ZHKUCC 23-0848) are ITS: 0.55% (3/542 bp), *gapdh*: 0.91% (2/220 bp), *chs-1*: 2.39% (6/251 bp), *act*: 0.90% (2/221 bp), and *tub2*: 0.00% (0/490 bp) excluding gaps. The nucleotide differences between *C. scovillei* (CBS 126529) and *C. chrysalidocarpi* (ZHKUCC 23-0848) are ITS: 0.37% (2/534 bp), *gapdh*: 3.65% (8/219 bp), *chs-1*: 0.40% (1/251 bp), *act*: 0.45% (1/221 bp), and *tub2*: 0.00% (0/490 bp) excluding gaps. The nucleotide differences between *C. eriobotryae* (GLMC 1935) and *C. scovillei* (CBS 126529) are ITS: 0.19% (1/540bp), *gapdh*: 2.39% (6/251 bp), *chs-1*: 0.35% (5/282 bp), *act*: 1.20% (3/246 bp), and *tub2*: 0.00% (0/490 bp) excluding gaps. The PHI test revealed that there is no significant recombination ($P = 0.3314 > 0.05$) between *C. chrysalidocarpi* and its closely related taxa (Fig. 4). Considering morphology, phylogeny and sequence data, we introduce *C. chrysalidocarpi* as a new species.

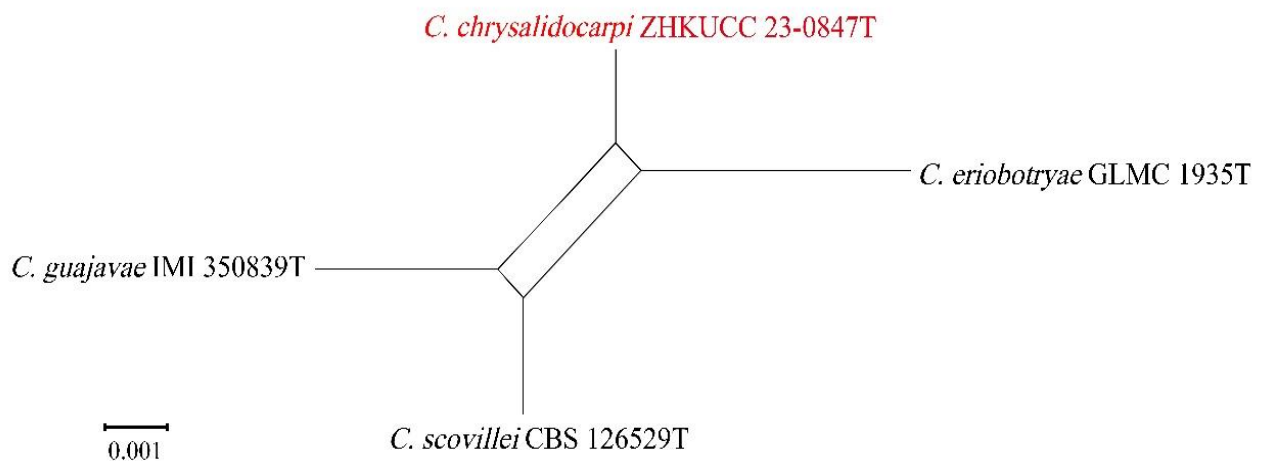


Figure 4 – The split graphs show the results of the PHI test of *Colletotrichum chrysalidocarpi* (ZHKUCC 23-0847) and their most closely related species using Log-Det transformation and split decomposition options. The PHI test result indicated ($P > 0.05$) that there is no evidence for significant recombination within the dataset. The new taxon is shown in red.

Colletotrichum fioriniae (Marcelino & Gouli) Pennycook, Mycotaxon 132(1): 150 (2017) [2016]

Fig. 5

Index Fungorum number: IF553097; Facesoffungi number: FoF 02891

New synonyms:

Colletotrichum orientale Dandan Fu & G.Y. Sun [as ‘orientalis’], in Chen et al., Journal of Fungi 8(7, no. 740): 10 (2022)

Colletotrichum radermacherae Y. Feng, Q. Zhang, Yong Wang bis & K.D. Hyde, in Zhang, et al., Mycosphere 14(2): 11 (2023)

Associated with leaf spots of *Malus spectabilis*. Sexual morph: Not observed. Asexual morph: *Setae* not observed. *Vegetative hyphae* 3–6.5 μm diam., hyaline to pale brown, smooth-walled, septate, branched. *Conidiomata* not observed. *Conidiophores* hyaline to pale brown, smooth-walled, septate, branched. *Conidiogenous cells* 10–14 \times 3–5 μm ($\bar{x} = 12 \times 4 \mu\text{m}$, $n = 30$), hyaline, ampulliform, smooth-walled. *Conidia* mostly fusiform with both ends acute, hyaline, aseptate, 12–15 \times 5–6 μm ($\bar{x} = 14 \times 6 \mu\text{m}$, $n = 50$); some short-cylindric with both ends round, 9–14 \times 4–7 μm ($\bar{x} = 12 \times 6 \mu\text{m}$, $n = 50$). *Appressoria* 9–14 \times 6–8 μm ($\bar{x} = 11 \times 6.5 \mu\text{m}$, $n = 30$), dark brown, clavate to irregular in outline.

Culture characteristics – Colonies on PDA 55 mm diam. after 7 days at 25 °C, flat, pink to yellow in the center, white at the margin, reverse pink to white, with pink concentric bands, conidial masses orange.

Material examined – China, Shanghai City, Pudong District, on leaf spot of *Malus spectabilis*

(Ait.) Borkh (*Rosaceae*), June 2022, Yanhong Lin, dried culture MHZU 23-0207; living culture ZHKUCC 23-0849.

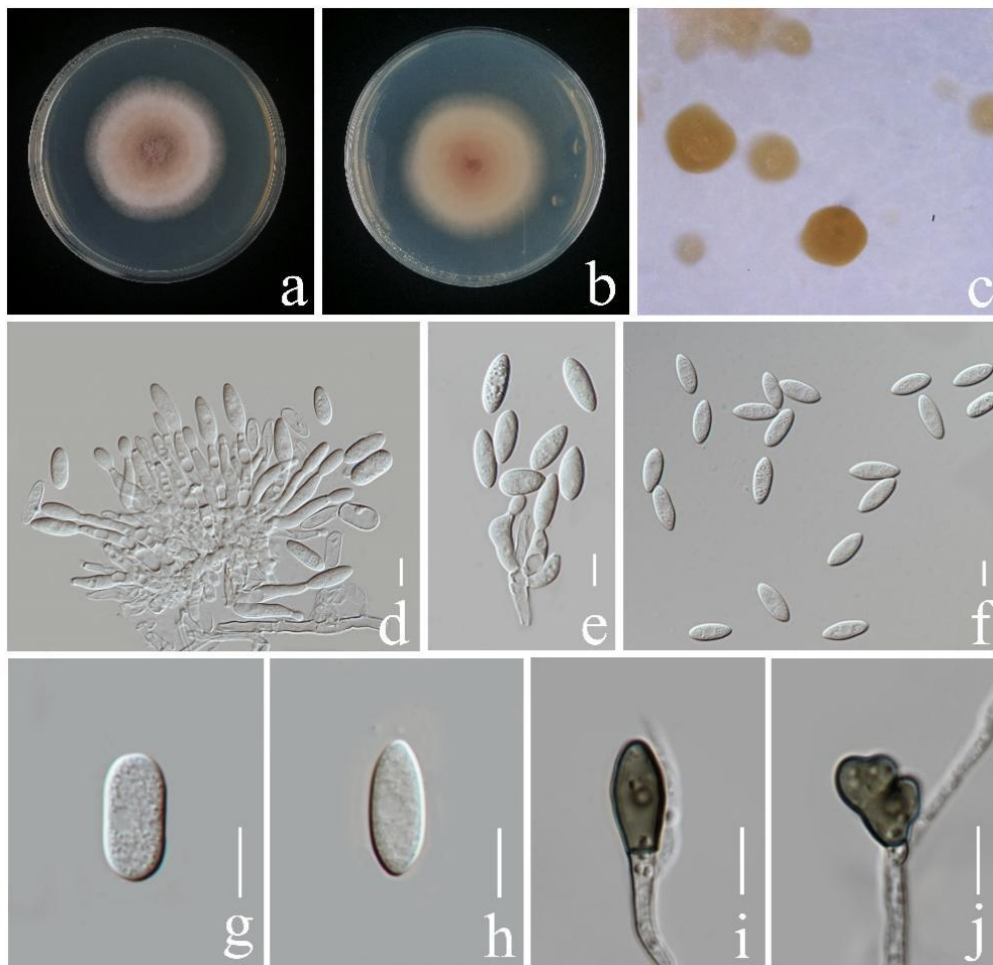


Figure 5 – *Colletotrichum fioriniae* (ZHKUCC 23-0849). a, b Upper and reverse view on PDA (7 d). c Conidial masses. d, e Conidiogenous cells. f–h Conidia. i, j Appressoria. Scale bars: d–k = 10 μ m.

Notes – In the phylogenetic analyses, three isolates from this study developed a distinct clade with *C. fioriniae*, *C. orientalis* and *C. radermacherae* from other known species in *Colletotrichum acutatum* species complex (100% ML bootstrap support and 1.00 BYPP; Fig. 2). Our isolates produced two shapes of conidia, fusiform and cylindrical, while *C. fioriniae* and *C. radermacherae* only have fusiform conidia. The fusiform conidia in this study ($\bar{x} = 14 \times 6 \mu\text{m}$) are smaller than *C. fioriniae* ($\bar{x} = 15.0 \times 4.5$), *C. orientalis* ($\bar{x} = 15.0 \times 4.5$) and *C. radermacherae* ($\bar{x} = 17.4 \times 5.3 \mu\text{m}$) (Damm et al. 2012a, Chen et al. 2022, Zhang et al. 2023) (Table 1). The nucleotide differences between our isolate (ZHKUCC 23-0849) and *C. fioriniae* (CBS 128517) are ITS: 0.00% (0/534 bp), *gapdh*: 0.91% (2/219 bp), *chs-1*: 1.21% (3/247 bp), *act*: 0.90% (2/221 bp), and *tub2*: 0.41% (2/491 bp) excluding gaps. The nucleotide differences between our isolate (ZHKUCC 23-0849) and *C. orientalis* (F10PGBYS8) are ITS: 0.00% (0/534 bp), *gapdh*: 0.00% (0/221 bp), *chs-1*: 1.22% (3/245 bp), *act*: 0.00% (0/221 bp), and *tub2*: 0.38% (2/530 bp) excluding gaps. The nucleotide differences between our isolate (ZHKUCC 23-0849) and *C. radermacherae* (GZCC 21-0813) are ITS: 0.40% (2/506 bp), *gapdh*: 0.93% (2/216 bp), *chs-1*: 1.63% (4/245 bp), *act*: 0.91% (2/220 bp), and *tub2*: 0.47% (3/641 bp) excluding gaps. The nucleotide differences between *C. fioriniae* (CBS 128517) and *C. radermacherae* (GZCC 21-0813) are ITS: 0.39% (2/506 bp), *gapdh*: 1.85% (4/216 bp), *chs-1*: 1.22% (3/245 bp), *act*: 1.82% (4/220 bp), and *tub2*: 1.01% (5/491 bp) excluding gaps. The nucleotide differences between *C. fioriniae* (CBS 128517) and *C. orientale*

(F10PGBYS8) are ITS: 0.00% (0/540 bp), *gapdh*: 0.79% (2/254 bp), *chs-1*: 0.71% (2/282 bp), *act*: 0.45% (1/221 bp), and *tub2*: 0.81% (4/491 bp) excluding gaps. Based on the morphology, *C. fioriniae*, *C. orientalis*, *C. radermacheriae* and our isolates have conidial dimensions which are only 1-2 microns different (Table 1). Furthermore, our isolate only differs by fusiform and cylindrical whereas the other three species are only fusiform. Therefore, we proposed these morphological variations are not enough to separate these strains as three different species. This is further confirmed by the nucleotide comparisons, in which three species have less than 2% variation among each other for all gene regions. Therefore, based on this evidence we reduce, *C. orientalis*, *C. radermacheriae* as synonyms of *C. fioriniae*. Here the present study provides a new host record of *C. fioriniae* associated with *Malus spectabilis*.

***Colletotrichum boninense* species complex**

Phylogenetic analyses

Phylogenetic trees were generated using combined ITS (563 bp), *gapdh* (277 bp), *chs-1* (255 bp), *act* (258 bp) and *tub2* (508 bp) sequence data. The tree topologies generated by ML and Bayesian analyses were similar and the best-scoring ML tree is shown in Fig. 6. The sequence alignment comprised 47 taxa of representative strains, including five isolates obtained in this study. *Colletotrichum truncatum* (CBS 151.35) was used as the outgroup taxon. The best-scoring ML tree had an optimization likelihood value of -9255.467896. The matrix had 746 distinct alignment patterns with an 8.90% proportion of gaps and completely undetermined characters. Estimated base frequencies were as follows: A = 0.229116, C = 0.298644, G = 0.249510, T = 0.222730; substitution rates: AC = 1.473821, AG = 3.346968, AT = 1.099222, CG = 0.954941, CT = 5.003071, GT = 1.000000; gamma distribution shape parameter $\alpha = 0.373360$. Incomplete portions at the ends of the sequences were excluded from the analysis. Nine isolates from this study formed three distinct clades, ZHKUCC 23-0852, ZHKUCC 23-0853 and ZHKUCC 23-0854 clustered with *C. cymbidiicola* with 98% ML and 1.00 BYPP support, ZHKUCC 23-0855 and ZHKUCC 23-0856 clustered with *C. chamaedoreae* with 100% ML and 1.00 BYPP support, and ZHKUCC 23-0857, ZHKU CC 23-0858, ZHKUCC 23-0859 and ZHKUCC 23-0850 constituted a sister relationship to *C. karsti* with 100% ML and 1.00 BYPP support (Fig. 6).

Taxonomy

Colletotrichum chamaedoreae F. Liu, W.P. Wu & L. Cai, in Liu et al., Studies in Mycology 101: 17 (2022) Fig. 7

Index Fungorum number: IF841374; Facesoffungi number: FoF 15257

Associated with leaf spot of *Chrysalidocarpus lutescens*. Sexual morph: *Ascomata* 130–180 (–220) × 100–150 (–180) μm ($\bar{x} = 160 \times 130 \mu\text{m}$, $n = 30$), formed after 4 weeks, black, globose to subglobose, semi-immersed or immersed. *Asci* 50–70(–84) × 11–16(–18) μm ($\bar{x} = 63 \times 14 \mu\text{m}$, $n = 30$), 6–8 spores, hyaline, clavate or cymbiform, apex and base acute. *Ascospores* 18–23(–25) × 5–8 μm ($\bar{x} = 21 \times 7 \mu\text{m}$, $n = 50$), uni- or biserially arranged, aseptate, hyaline, fusiform, slightly curved. Asexual morph: *Vegetative hyphae* hyaline, smooth-walled, septate, branched. *Setae* 46–71 μm long, 1–7 septate, dark brown, base cylindrical, tip broadly acute. *Conidiomata* black, scattered or in groups. *Conidiogenous cells* 9–16(–20) × 4–7 μm ($\bar{x} = 12 \times 5 \mu\text{m}$, $n = 50$), hyaline or pale brown, smooth, cylindrical. *Conidia* 17–21 × 6–9 μm ($\bar{x} = 19 \times 8 \mu\text{m}$, $n = 50$), aseptate, smooth-walled, hyaline, mostly both ends round, sometimes one end with a prominent truncate scar. *Appressoria* 9–13(–15) × (6–)8–12(–15) μm ($\bar{x} = 11 \times 10 \mu\text{m}$, $n = 50$), dark brown, irregular.

Culture characteristics – Colonies on PDA 60 mm diam. after 7 days at 25 °C. Colony green-grey, white edges, flat, regular, mycelium flocculent, reverse orange, conidial masses orange.

Material examined – China, Guangdong Province, Guangzhou City, on leaf spot of *Chrysalidocarpus lutescens* H. Wendl (*Arecaceae*), October 2021, Yunxia Zhang and Jingwen Chen (living culture ZHKUCC 23-0855, new host record).

Note – In the multigene phylogenetic tree, the strains in this study clustered with *C. chamaedoreae* with 100% ML and 1.00 BYPP value (Fig. 6). Morphologically our isolates were similar to those of *C. chamaedoreae* (Liu et al. 2022). Therefore, we identified our isolates as *C. chamaedoreae*. The holotype of this species was isolated from *Chamaedorea erumpens* of the same host family *Arecaceae* (Liu et al. 2022). This is the first report of *C. chamaedoreae* associated with *Chrysalidocarpus lutescens*.

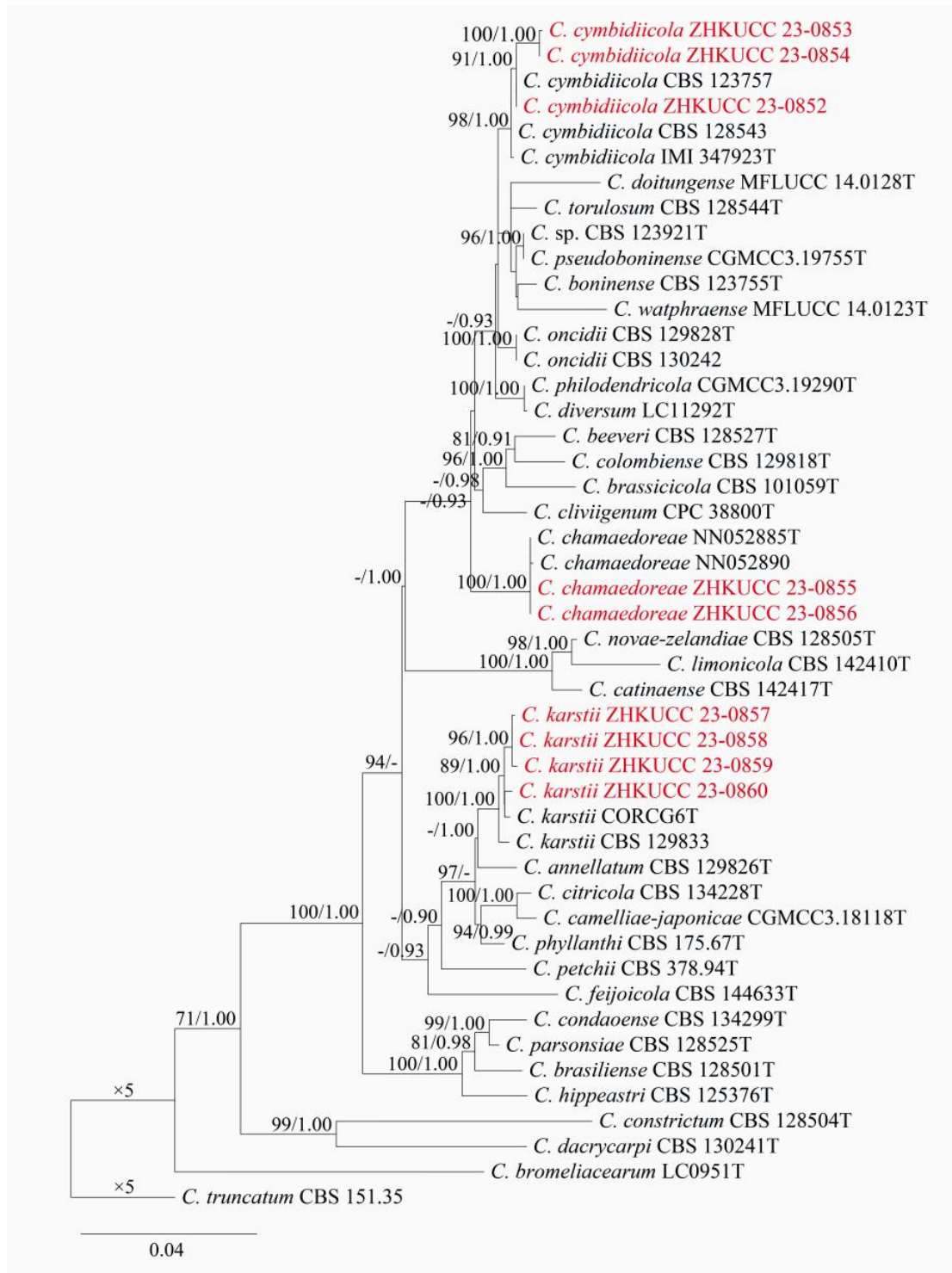


Figure 6 – Maximum likelihood tree of the *Colletotrichum boninense* species complex. *Colletotrichum truncatum* (CBS 151.35) was selected as the outgroup. At the nodes, bootstrap support values for ML ($\geq 70\%$) and BYPP (≥ 0.90) are displayed (ML/PP). Some branches were

shortened to fit them to the page, Ex-type isolates are marked with “T”, and new isolates from this study are marked in red font. The scale bar indicates 0.04 nucleotide changes per site.

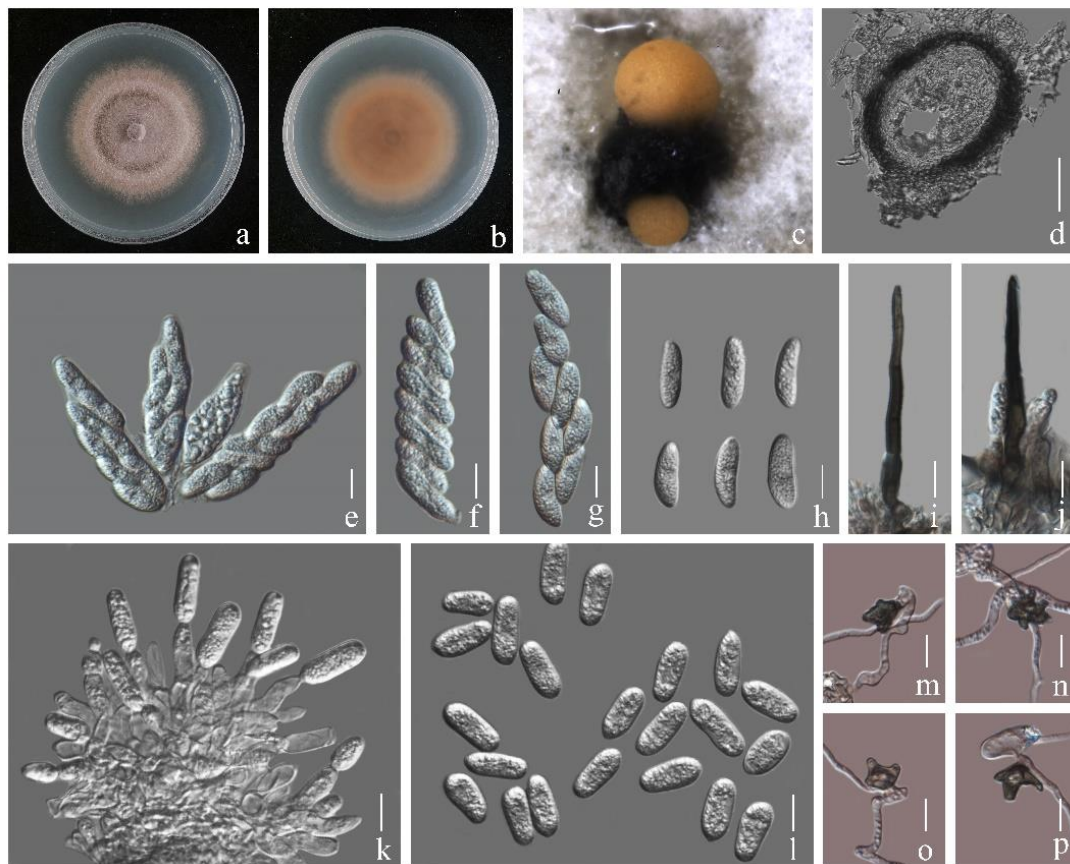


Figure 7 – *Colletotrichum chamaedoreae* (ZHKUCC 23-0855). a, b Upper and reverse view on PDA (7 d). c Conidial masses. d Ascomata. e–g Asci. h Ascospores. i, j Setae. k Conidiogenous cells. l Conidia. m–p Appressoria. Scale bars: d = 20 μ m, e–p = 10 μ m.

Colletotrichum cymbidiicola Damm, P.F. Cannon, Crous, P.R. Johnst. & B.S. Weir, in Damm et al., *Studies in Mycology* 73: 19 (2012) Fig. 8

Index Fungorum number: IF560740; Facesoffungi number: FoF 15258

Associated with leaf spot of *Dendrobium nobile*. Sexual morph: Not observed. Asexual morph: *Conidiomata* not observed. *Conidiophores* not developed *Conidiogenous cells* formed from mycelium directly. *Conidia* 17–21 \times 6–8 μ m (\bar{x} = 19 \times 7 μ m, n = 50), hyaline, smooth-walled, aseptate, cylindrical, both ends rounded, with a prominent scar, contents granular. *Appressoria* 9–14(–20) \times 7–11 μ m (\bar{x} = 13 \times 9 μ m, n = 50), single, medium to dark brown, plum-shaped to irregular, outline variable, margin lobate.

Culture characteristics – Colonies on PDA 85–90 mm diam. after 7 days at 25 $^{\circ}$ C, flat, with entire margin, white, aerial mycelium dense, flocculent, reverse pink in the center, white towards margin.

Material examined – China, Guangdong Province, Zhaoqing City, on leaf spot of *Dendrobium nobile* Lindl. (*Orchidaceae*), July 2021, Yunxia Zhang and Jingwen Chen (living culture ZHKUCC 23-0852, new host record)

Additional material – China, Guangdong Province, Meizhou City, on leaf spot of *C. sinense* (Jack. ex Andr.) Willd. (*Orchidaceae*), October 2021, Yunxia Zhang and Jingwen Chen (living culture ZHKUCC 23-0853, new host record).

Note – Based on the multigene phylogenetic tree, three isolates clustered together with *C. cymbidiicola* with 98% support in MP and 1.00 in BYPP (Fig. 6). Our isolates were similar to

the original description of *C. cymbidiicola* (Damm et al. 2012b). Based on morphology and multigene phylogenetic analyses, our isolates were identified as *C. cymbidiicola*. *Colletotrichum cymbidiicola* was described from *Cymbidium* sp. (Damm et al. 2012b), but it has a wide host range, including orchids such as *Bulbophyllum hirtum*, *Coeloyne elata*, *Dendrobium fimbriatum*, *Liparis longipes*, *Eria* sp. and *Oncidium sphacelatum*, as well as *Citrus grandis* and *Cassia fistula*. However, there is no previous record of *C. cymbidiicola* on *Cymbidium sinense*.

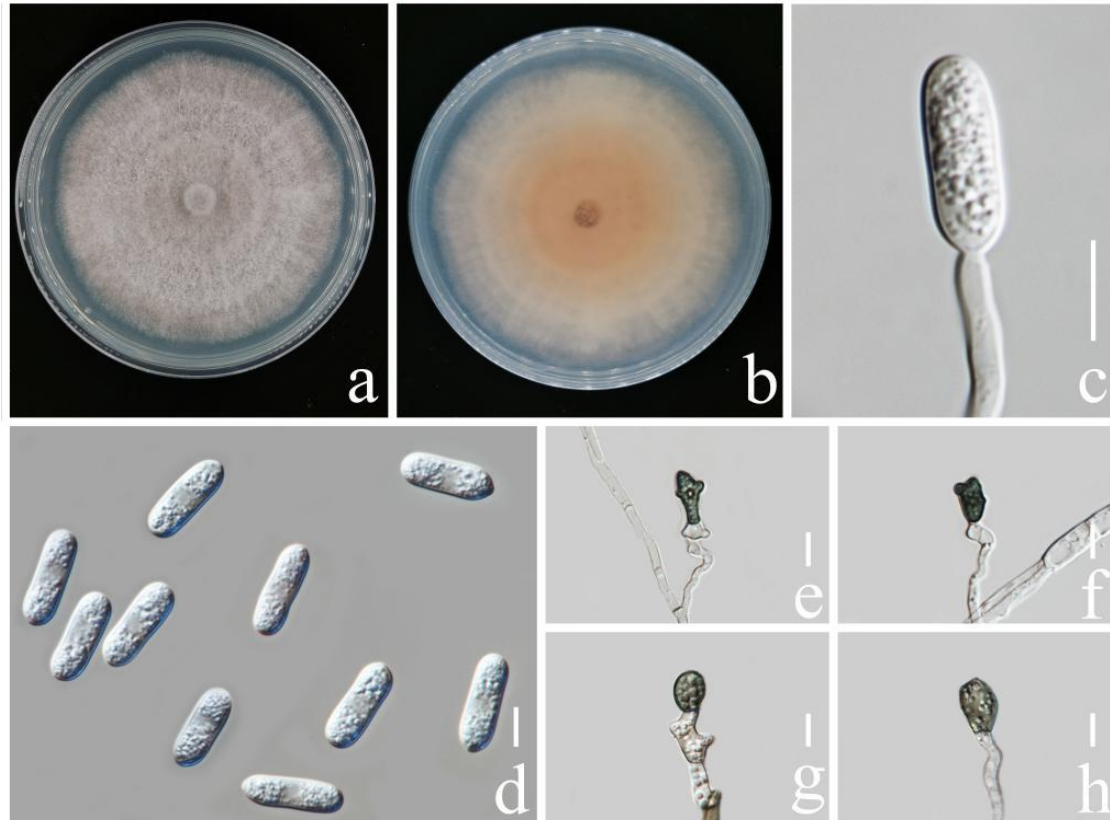


Figure 8 – *Colletotrichum cymbidiicola* (ZHKUCC 23-0852). a, b Upper and reverse view on PDA (7 d). c Conidia forming from mycelium. d Conidia. e–h Appressoria. Scale bars: c–k = 10 μ m.

Colletotrichum karsti You L. Yang, Zuo Y. Liu, K.D. Hyde & L. Cai [as ‘karstii’], in Yang et al., Cryptog. Mycol. 32(3): 241 (2011) Fig. 9

Index Fungorum number: IF581687; Facesoffungi number: FoF 09442

Associated with leaf spot of *Cymbidium sinense*. Sexual morph: *Ascomata* 140–190 \times 120–150(–180) μ m (\bar{x} = 168 \times 139 μ m, n = 30), formed after 4 weeks, solitary, superficial or immersed, black, subglobose to obpyriform, ostiolate. *Asci* (48–)53–70 \times 10–14 μ m (\bar{x} = 58 \times 12 μ m, n = 50), smooth-walled, cylindrical to clavate, tapering to apex and base, 8-spored. *Ascospores* 15–18(–21) \times 6–8 μ m (\bar{x} = 17 \times 7 μ m, n = 50), uni- or biserially arranged, aseptate, hyaline, fusiform to ovoid, slightly curved. Asexual morph: *Vegetative hyphae* 1–5 μ m diam., hyaline, smooth-walled, septate, branched. *Conidiomata* black, scattered. *Setae* not observed. *Conidiophores* 100 μ m long, hyaline to pale brown, smooth, septate, branched. *Conidiogenous cells* 9–20 \times 3–5 μ m (\bar{x} = 16 \times 4 μ m, n = 30), hyaline or pale brown, smooth, cylindrical to elongate-ampulliform. *Conidia* 13–16(–21) \times 5–7 μ m (\bar{x} = 15 \times 6 μ m, n = 30), straight, hyaline, smooth-walled, aseptate, cylindrical, both ends rounded, contents granular. *Appressoria* 8–13 \times 5–7(–9) μ m (\bar{x} = 10 \times 6 μ m, n = 30), single or in groups, medium to dark brown, navicular to bullet-shaped, with entire edge.

Culture characteristics – Colonies on PDA 70 mm diam. after 7 days at 25 $^{\circ}$ C, circular, regular, white-pink, mycelium lush, flocculent, reverse orange, producing black ascomata and orange conidial masses.

Material examined – China, Guangdong Province, Meizhou City, on leaf spot of *Cymbidium sinense* (Jack. ex Andr.) Willd. (*Orchidaceae*), October 2021, Yunxia Zhang and Jingwen Chen (living culture ZHKU 23-0857, new host record).

Additional material – China, Guangdong Province, Meizhou City, on leaf spot of *Aglaonema* sp. (*Araceae*), October 2021, Yunxia Zhang and Jingwen Chen (ZHKUCC 23-0858, new host record). China, Hunan Province, Changsha City, on leaf spot of *Rosa chinensis* Jacq. (*Rosaceae*), August 2022, Chao Chen (living culture ZHKUCC 23-0860, new host record).

Note – In the phylogenetic tree, our strains grouped with *C. karsti* with 100% MP and 1.00 BYPP support (Fig. 6). Morphologically, our isolates were similar to *C. karsti* (Yang et al. 2011). Based on morphology and multigene analyses, isolates from *Aglaonema* sp., *Cymbidium sinense* and *Rosa chinensis* were identified as *C. karsti*. The holotype of *C. karsti* was isolated from *Vanda* sp. (Yang et al. 2011). It has a wide range of hosts, including fruits (*Annona cherimola*, *Carica papaya*, *Citrus* sp., *Mangifera indica*) and ornamental plants (*Anthurium* sp., *Clivia miniata*, and *Ficus microcarpa*) (Damm et al. 2012b, Jayawardena et al. 2016, Liu et al. 2022). However, this is the first report of *C. karsti* isolated from *Aglaonema* spp., *Cymbidium sinense* and *Rosa chinensis*.

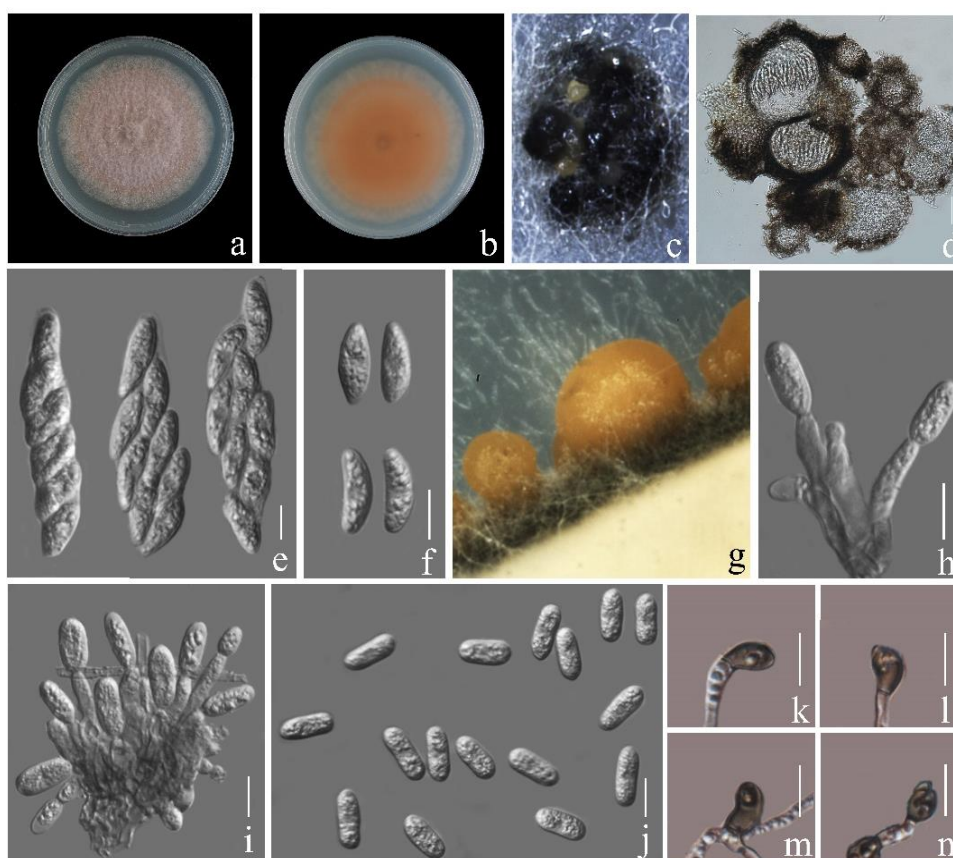


Figure 9 – *Colletotrichum karsti* (ZHKUCC 23-0858). a, b Upper and reverse view on PDA (7 d). c, d Ascomata. e Asci. f Ascospores. g Conidial masses. h, i Conidiogenous cells. j Conidia. k–n Appressoria. Scale bars: d = 20 μ m, d–f, h–n = 10 μ m.

Colletotrichum gloeosporioides species complex

Phylogenetic analysis

Phylogenetic trees were generated using combined ITS (542 bp), *gapdh* (253 bp), *chs-1* (244 bp), *act* (239 bp) and *tub2* (696 bp) sequence data. The tree topologies generated by ML and Bayesian were similar and the best-scoring ML tree is shown in Fig. 10. The sequence alignment comprised 111 taxa of representative strains of the *C. gloeosporioides* species complex, including 19 isolates from this study. *Colletotrichum boninense* (CBS 123755) and *C. brasiliense* (CBS

128501) were used as the outgroup taxa. The best-scoring ML tree had an optimization likelihood value of -11730.698033. The matrix had 900 distinct alignment patterns with a 13.20% proportion of gaps and completely undetermined characters. Estimated base frequencies were as follows: A = 0.227302, C = 0.303998, G = 0.238628, T = 0.230072; substitution rates: AC = 1.017549, AG = 3.255408, AT = 1.009193, CG = 0.891395, CT = 4.677941, GT = 1.000000; gamma distribution shape parameter α = 1.228568. Incomplete portions at the ends of the sequences were excluded from the analysis.

In the present study, we obtained a total of 19 isolates belonging to *Colletotrichum gloeosporioides* species complex. Six isolates from this study (ZHKUCC 23-0832, ZHKUCC 23-0833, ZHKUCC 23-0834, ZHKUCC 23-0835, ZHKUCC 23-0836 and ZHKUCC 23-0837) grouped with *C. siamense*, four isolates (ZHKUCC 23-0828, ZHKUCC 23-0829, ZHKUCC 23-0830 and ZHKUCC 23-0831) grouped with *C. fructicola* with 94% ML and 0.99 BYPP support. Three isolates constituted a sister relationship to *C. nanhuaense* with 67% ML and 0.96 BYPP support (Fig. 10) while developing a distinct cluster together with *C. gloeosporioides*, *C. dimorphum* and *C. yunjiangenses*. However, this particular relationship only could be observed when we added more *C. gloeosporioides* strains to the phylogenetic analysis (Fig. 10). The final tree (Fig. 10) suggests that *C. nanhuaense*, *C. dimorphum* and *C. yunjiangenses* and our isolated could be the same species; *C. gloeosporioides*. To confirm this, we compared morphology and pairwise nucleotide variations among these four species. A similar clustering pattern was observed for the remaining six isolated from this study. Three isolates constituted a sister relationship to *C. endophyticum* with 81% ML and 0.99 BYPP support and the other three isolates grouped separately with *C. endophyticum* type strain (81% ML and 0.99 BYPP support). For all identified taxa, species descriptions and illustrations are given below.

Taxonomy

Colletotrichum endophyticum Manamgoda, Udayanga, L. Cai & K.D. Hyde [as ‘endophytica’], in Manamgoda, Udayanga, Cai, Chukeatirote & Hyde, *Fungal Diversity* 61: 110 (2013) Fig. 11

Index Fungorum number: IF565248; Facesoffungi number: FoF 15259

Associated with leaf spot of *Bauhinia blakeana*. Sexual morph: Not observed. Asexual morph: *Conidiomata* scattered, dark brown. *Setae* not observed. *Conidiophores* hyaline, unbranched. *Conidiogenous cells* (4–)6–8 × 3–4 μm (\bar{x} = 6.5 × 3.9 μm , n = 30), hyaline, cylindrical. *Conidia* 14–18 × 4–6 μm (\bar{x} = 16 × 5 μm , n = 50), hyaline, smooth-walled, cylindrical, one end rounded and one end acute, producing conidial anastomosis tubes. *Appressoria* 7–11 × 5–8 μm (\bar{x} = 9 × 6 μm , n = 30), pale brown, ellipsoidal to oval in outline.

Culture characteristics – Colonies on PDA 67 mm diam. after 7 days at 25 °C, circular, regular margin, white, aerial mycelium dense and raised, villous, reverse white to grey, concentric ring in the center, conidial masses orange.

Material examined – China, Guangdong Province, Guangzhou City, on leaf spot of *Bauhinia blakeana* Dunn (*Leguminosae*), February 2022, Jiachun Wang, (MHZU 23-0200 dried culture); living culture ZHKUCC 23-0841.

Additional material examined – China, Guangdong Province, Shenzhen City, on leaf spot of *Acacia confuse* Merr. (*Fabaceae*), November 2020, Jingwen Chen, (ZHKU 23-0197 dried culture); living cultures ZHKUCC 23-0838, ZHKUCC 23-0839 and ZHKUCC 23-0840.

Notes – In the phylogenetic analysis, six isolates from the present study cluster together with *Colletotrichum endophyticum* strains (Fig. 10). Our isolates produced conidial anastomosis tubes, but this phenomenon was not observed in *C. endophyticum* (as *endophytica*) (Manamgoda et al. 2013). The nucleotide differences between our representative strain (ZHKUCC 23-0841) and *C. endophyticum* (MFLUCC 13.0418) are ITS: 0.21% (1/477 bp), *gapdh*: 0.46% (1/218 bp), and *act*: 2.16% (5/232 bp) excluding gaps. Furthermore, since our isolates grouped in two clades, we also compared nucleotide differences between ZHKUCC 23-0841 and ZHKUCC 23-0838, which are ITS: 0.37% (2/541 bp), *gapdh*: 0.92% (2/218 bp), *chs-1*: 1.23% (3/243 bp), *act*: 3.02% (7/232

bp), and *tub2*: 2.21% (15/680 bp) excluding gaps. Based on morphological and sequence comparisons, we identified six strains belonging to this study as *C. endophyticum*. This is the first report of *C. endophyticum* associated with *Bauhinia blakeana*.

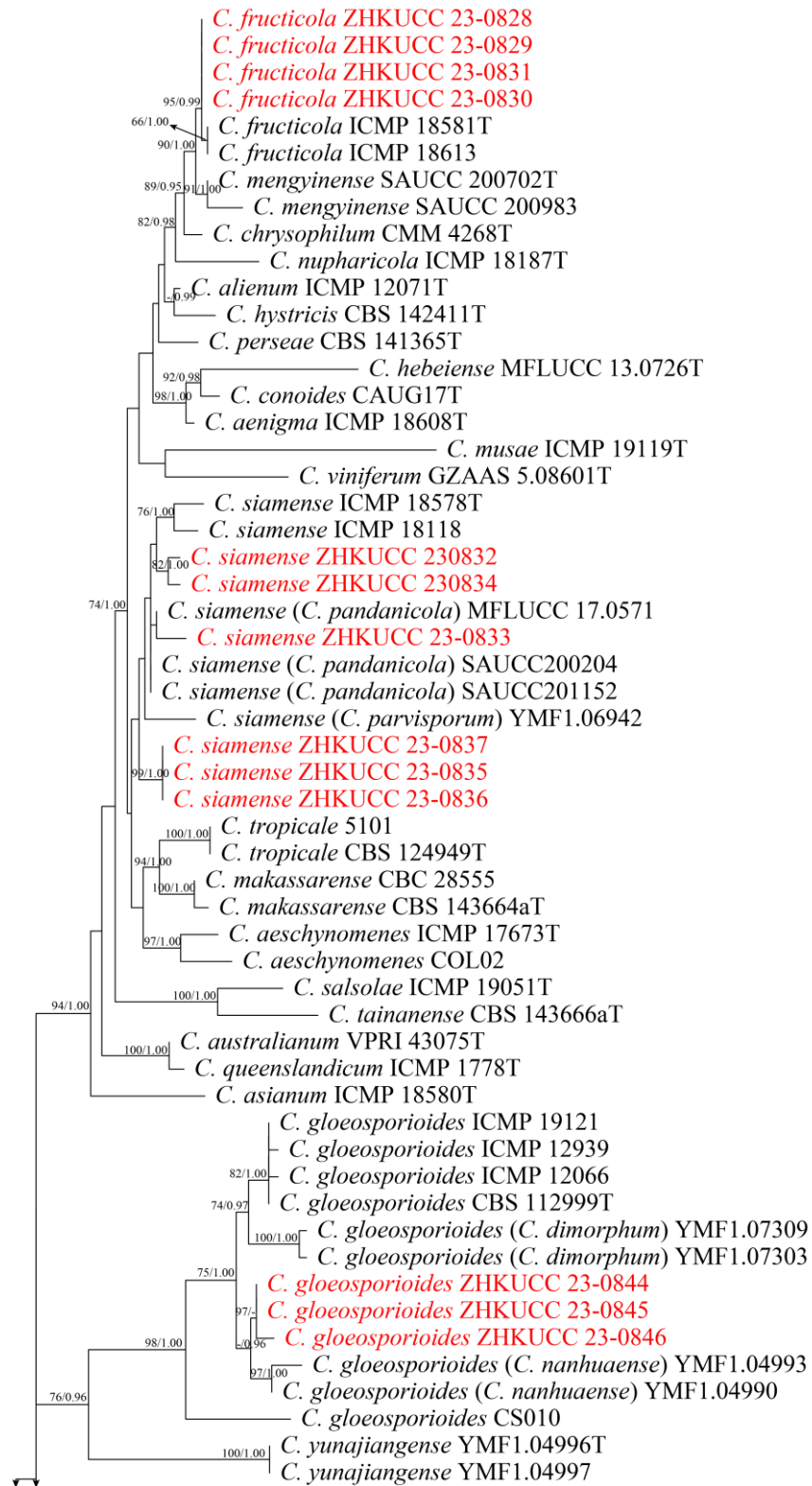


Figure 10 – Maximum likelihood tree of the *Colletotrichum gloeosporioides* species complex. *C. boninense* (CBS 123755) and *C. brasiliense* (CBS 128501) were used as the outgroup. At the nodes, bootstrap support values for ML ($\geq 70\%$) and BYPP (≥ 0.95) are displayed (ML/PP). Some branches were shortened to fit them to the page, Ex-type isolates are marked with “T”, and isolates from this study are in red. The scale bar indicates 0.03 nucleotide changes per site.

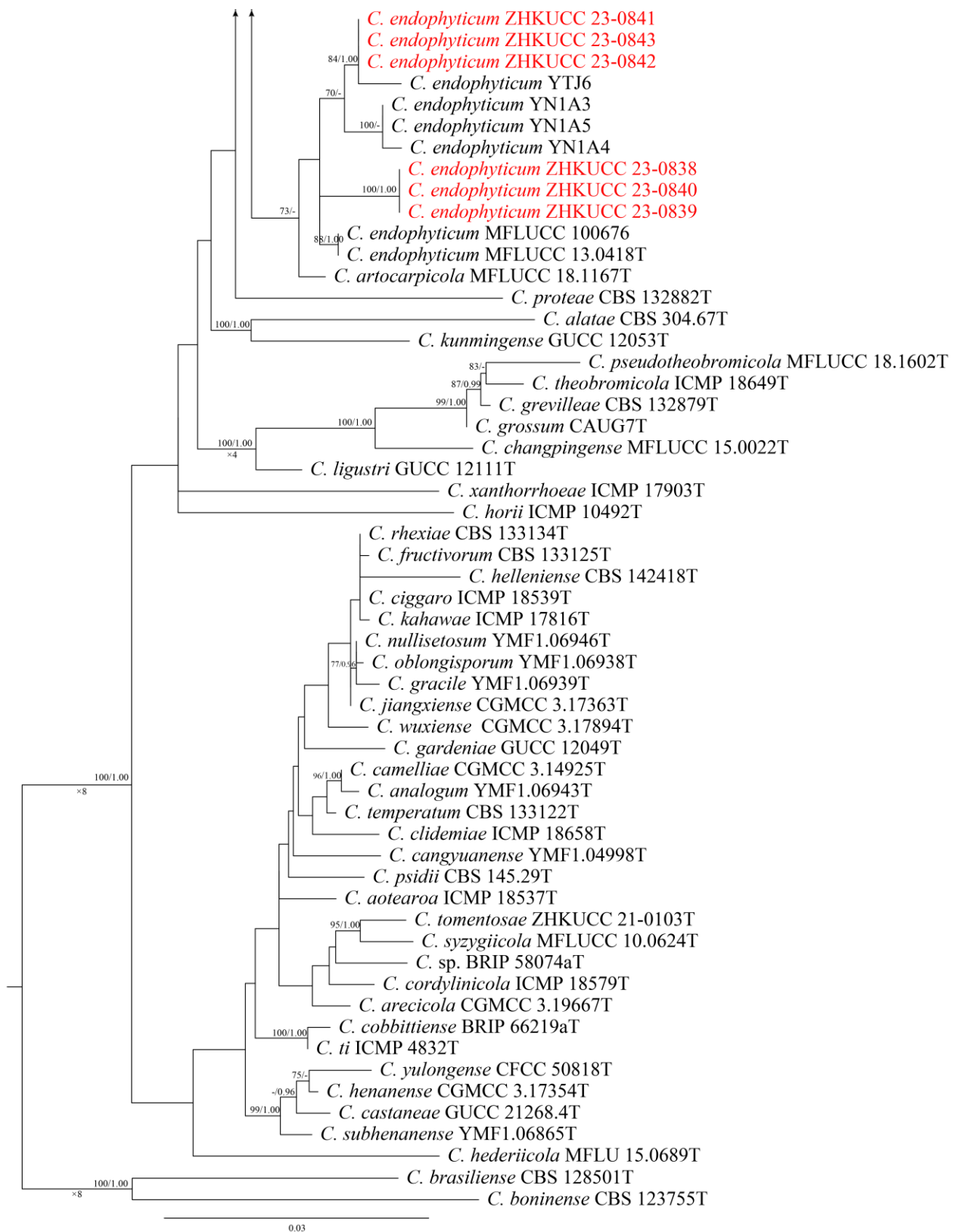


Figure 10 – Continued.

Colletotrichum fructicola Prihast., L. Cai & K.D. Hyde, in Prihastuti, Cai, Chen, McKenzie & Hyde, Fungal Diversity 39: 96 (2009) Fig. 12

Index Fungorum number: IF515409; Facesoffungi number: FoF 06767

Associated with leaf spot of *Celosia cristata*. Sexual morph: *Ascomata* (70–)80–100(–140) × 50–80(–120) μm (\bar{x} = 95 × 78 μm, n = 30), formed after 4 weeks, pale brown to brown, globose to

subglobose, semi-immersed to immersed. *Asci* 43–68(–80) × 8–12(–14) μm (\bar{x} = 59 × 11 μm, n = 30), thin-walled, 6–8 spored, clavate or cymbiform, apex and base acute. *Ascospores* 16–20(–22) × 4–6 μm (\bar{x} = 18.5 × 5 μm, n = 50), uni- or biserially arranged, aseptate, hyaline, guttulate, curved. Asexual morph: *Vegetative hyphae* hyaline, smooth-walled, septate, branched. *Conidiogenous cells* (4–)5–9(–13) × 2–5 μm (\bar{x} = 7 × 4 μm, n = 50), hyaline or pale brown, smooth, cylindrical to elongate-ampulliform. *Conidia* 14–17 × 5–7 μm (\bar{x} = 15 × 6 μm, n = 50), aseptate, smooth-walled, hyaline, cylindrical, both ends obtuse, or one end rounded and one end acute. *Appressoria* 8–12(–15) × 5–8(–9) μm (\bar{x} = 11 × 6 μm, n = 50), mostly formed from mycelia, brown to dark brown, ovoid, clavate to irregular.

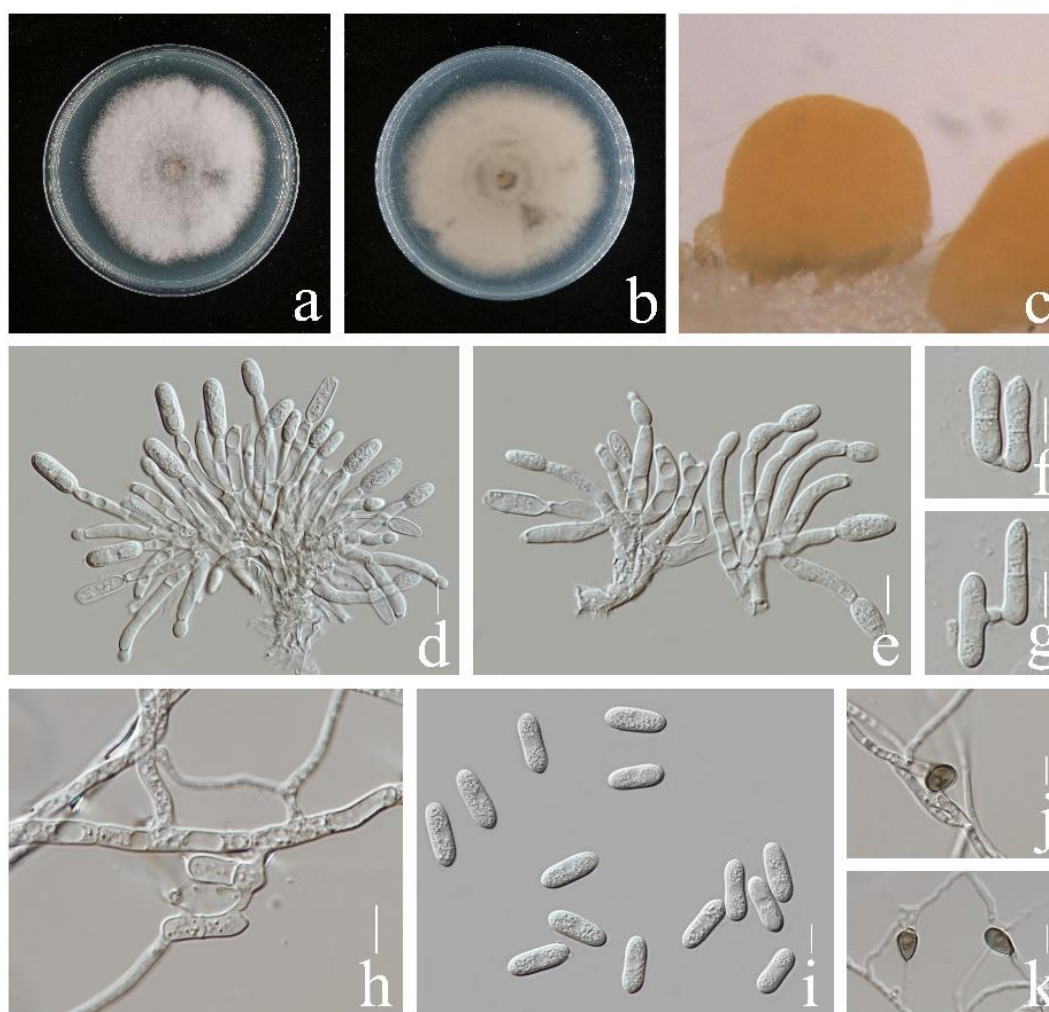


Figure 11 – *Colletotrichum endophyticum* (ZHKUCC 23-0841). a, b Upper and reverse view on PDA (7 d). c Conidial masses. d, e Conidiogenous cells. f–h Conidial anastomosis fusion. i Conidia. j, k Appressoria. Scale bars: d–k = 10 μm.

Culture characteristics – Colonies on PDA 80 mm diam. after 7 days at 25 °C, grey to dark grey with white margin, reverse dark grey in the centre and white towards margin with white concentric bands, conidial masses orange yellow.

Material examined – China, Guangdong Province, Guangzhou City, on leaf spot of *Celosia cristata* Linn. (*Amaranthaceae*), October 2021, Yunxia Zhang and Jingwen Chen (living culture ZHKUCC 23-0829, new host record).

Additional materials – China, Guangdong Province, Zhongshan City, on leaf spot of *Dendrobium nobile* Lindl (*Orchidaceae*), October 2021, Yunxia Zhang and Jingwen Chen (living culture ZHKUCC 23-0828, new host record). China, Guangdong Province, Meizhou City, on leaf

spot of *Cymbidium sinense* (Jack. ex Andr.) Willd. (*Orchidaceae*), October 2021, Yunxia Zhang and Jingwen Chen (living culture ZHKUCC 23-0830 and ZHKUCC 23-0831, new host record).

Note – In the multigene phylogenetic tree, our strains clustered with *C. fructicola* with 95% support in ML and 0.99 in BYPP (Fig. 10). Conidial dimensions of our isolates were similar to *C. fructicola* (Prihastuti et al. 2009). Therefore, our isolates were identified as *C. fructicola*. This species has a wide range of reported hosts, mainly including plants in *Fabaceae*, *Leguminosae*, *Lauraceae*, *Magnoliaceae*, *Rutaceae*, and *Ficus habrophylla* (Peng et al. 2012, Liu et al. 2016, 2022). However, our study provides the first report of *C. fructicola* isolated from *Celosia cristata*, *Dendrobium nobile* and *Cymbidium sinense*.

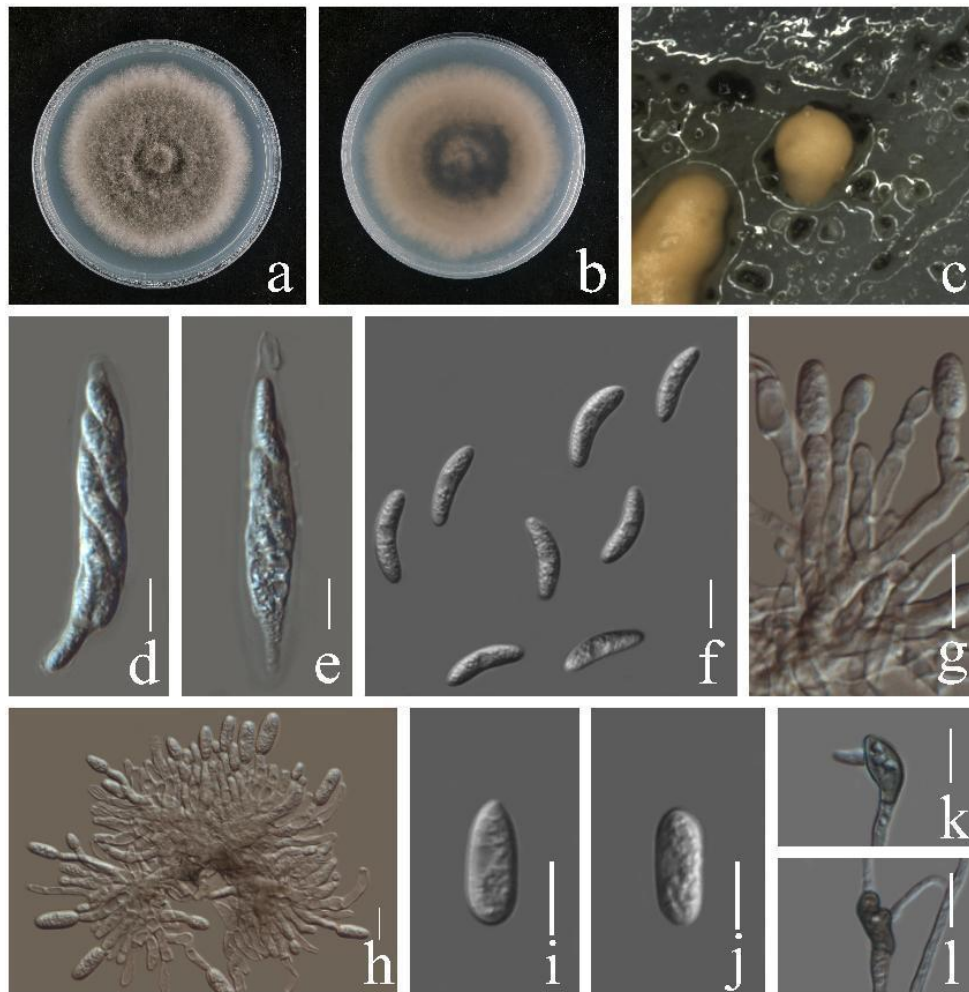


Figure 12 – *Colletotrichum fructicola* (ZHKUCC 23-0829). a, b Upper and reverse view on PDA (7 d). c Conidial masses. d, e Asci. f Ascospores. g, h Conidiogenous cells. i, j Conidia. k, l Appressoria. Scale bars: d–l = 10 μ m.

Colletotrichum gloeosporioides (Penz.) Penz. & Sacc., Atti Inst. Veneto Sci. lett., ed Arti, Sér. 6 2(5): 670 (1884) Fig. 13

Index Fungorum number: IF158410; Facesoffungi number: FoF 09424

New synonyms:

Colletotrichum nanhuaense Z.F. Yu, in Yu et al., Journal of Fungi 8(2, no. 185): 17 (2022)

Colletotrichum dimorphum Z.F. Yu, in Yu et al., Journal of Fungi 8(2, no. 185): 14 (2022)

Associated with leaf spot of *Pittosporum tobira*. Sexual morph: Not observed. Asexual morph: *Conidiomata* dark grey, scattered. *Setae* not observed. *Conidiophores* hyaline to pale brown, aseptate, branched. *Conidiogenous cells* 4–8 \times 3–4 μ m (\bar{x} = 6.5 \times 4 μ m, n = 30), hyaline, cylindrical, smooth-walled. *Conidia* 15–19 \times 4–7 μ m (\bar{x} = 17 \times 6 μ m, n = 50), hyaline, smooth-

walled, aseptate, straight, cylindrical, one end rounded and one end acute, contents granular. *Appressoria* 6–9(–11) × 4–8 μm (\bar{x} = 8 × 6 μm, n = 30), dark brown, ellipsoidal to oval, or irregular in outline with entire margin

Culture characteristics – Colonies on PDA 75 mm diam. after 7 days at 25 °C, circular, regular at the margin, white, villous, aerial mycelium dense and raised, reverse dark green in the center, pink to white towards margin, conidial masses orange.

Material examined – China, Yunnan Province, Kunming City, on leaf spot of *Pittosporum tobira* (Thunb.) W. T. Aiton (*Pittosporaceae*), July 2022, Yunxia Zhang, (MHZU 23-0203 dried culture); living cultures ZHKUCC 23-0844, ZHKUCC 23-0845 and ZHKUCC 23-0846.

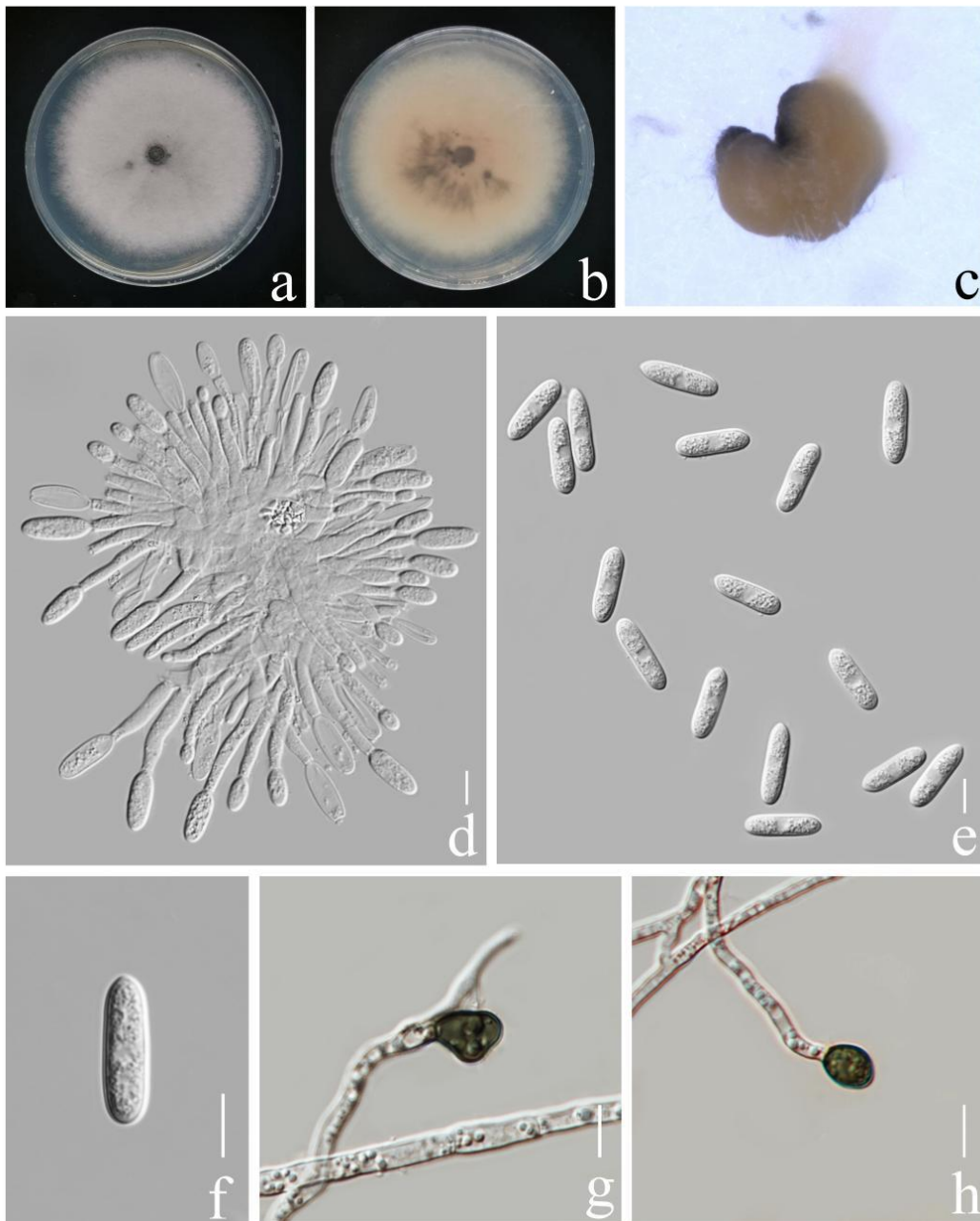


Figure 13 – *Colletotrichum gloeosporioides* (ZHKUCC 23-0844). a, b Upper and reverse view on PDA (7 d). c Conidial masses. d Conidiogenous cells. e, f Conidia. g, h Appressoria. Scale bars: d–h = 10 μm.

Notes – In the phylogenetic tree, isolates from our study developed a distinct clade closely related to *C. nanhuaense*, *C. gloeosporioides* and *C. dimorphum* (Fig. 10). We compared the

morphology of these two species with our isolates (Table 1). Our isolates have longer conidia ($15\text{--}19 \times 4\text{--}7 \mu\text{m}$, $\bar{x} = 17 \times 6$) than *C. nanhuaense* ($10.5\text{--}16 \times 4.5\text{--}6 \mu\text{m}$, $\bar{x} = 14 \times 5.4 \mu\text{m}$). The conidia of our isolates also differ in shape (cylindrical, one end rounded and one end acute) from those of *C. nanhuaense* (oblong to narrowly ovoid, rounded at both ends) (Yu et al. 2022b). In addition, an in-depth morphological comparison has been given for these three species in Table 1. The nucleotide differences between our isolate (ZHKUCC 23-0844) and *C. gloeosporioides* (CBS 112999) are ITS: 0.00% (0/536 bp), *gapdh*: 3.21% (7/218 bp), *act*: 0.00% (0/232 bp), and *tub2*: 0.00% (0/488 bp) excluding gaps. Nucleotide differences between our isolate (ZHKUCC 23-0844) and *C. nanhuaense* (YMF1.04990) are ITS: 0.00% (0/541 bp), *gapdh*: 0.00% (0/217 bp), *chs-1*: 0.44% (1/225 bp), *act*: 0.43% (1/232 bp), and *tub2*: 0.28% (1/361 bp) excluding gaps. Nucleotide differences between our isolate (ZHKUCC 23-0844) and *C. dimorphum* (YMF1.04990) are ITS: 0.00% (0/540 bp), *gapdh*: 1.86% (4/215 bp), *chs-1*: 2.86% (6/210 bp), *act*: 0.43% (1/231 bp), and *tub2*: 0.49% (2/409 bp) excluding gaps. *Colletotrichum gloeosporioides* (CBS 112999) and *C. nanhuaense* (YMF1.04990) have nucleotide different in ITS: 0.00% (0/536 bp), *gapdh*: 3.23% (7/217 bp), *act*: 0.43% (1/232 bp), and *tub2*: 0.85% (2/361 bp) excluding gaps. *Colletotrichum gloeosporioides* (CBS 112999) and *C. dimorphum* (YMF1.04990) have nucleotide different in ITS: 0.00% (0/536 bp), *gapdh*: 1.21% (3/248 bp), *act*: 0.39% (1/255 bp), and *tub2*: 0.49% (2/409 bp) excluding gaps. *C. nanhuaense* (CBS 112999) and *C. dimorphum* (YMF1.04990) have nucleotide different in ITS: 0.18% (1/568 bp), *gapdh*: 1.61% (4/248 bp), *act*: 2.38% (5/210 bp), *act*: 0.78% (2/255 bp), and *tub2*: 0.00% (0/361 bp) excluding gaps. Based on morphology, phylogeny, and sequence data, here in we introduce our isolates as *C. gloeosporioides* associated with *Pittosporum tobira*. In addition to that due to lack of morphological and sequence variations, we reduced *C. dimorphum* and *C. nanhuaense* as synonyms of *C. gloeosporioides*. In depth explanation for taxonomic treatments of this section is given in the discussion.

Colletotrichum siamense Prihast., L. Cai & K.D. Hyde, in Prihastuti, Cai, Chen, McKenzie & Hyde, Fungal Diversity 39: 98 (2009) Fig. 14

Index Fungorum number: IF515410; Facesoffungi number: FoF 03599

Associated with leaf spot of *Thalia dealbata*. Sexual morph: not observed. Asexual morph: *Vegetative hyphae* hyaline, smooth-walled, septate, branched. *Setae* not observed. *Conidiomata* dark brown, scattered. *Conidiogenous cells* $5\text{--}9\text{--}10 \times 3\text{--}4 \mu\text{m}$ ($\bar{x} = 8 \times 3 \mu\text{m}$, $n = 30$), hyaline, cylindrical. *Conidia* $15\text{--}18 \times 5\text{--}6 \mu\text{m}$ ($\bar{x} = 17 \times 5 \mu\text{m}$, $n = 50$), hyaline, aseptate, smooth-walled, straight, cylindrical with both ends rounded. *Appressoria* $8\text{--}10 \times 6\text{--}8 \mu\text{m}$ ($\bar{x} = 9 \times 7 \mu\text{m}$, $n = 50$), ellipsoidal to ovoid or clavate with entire margin.

Culture characteristics – Colonies on PDA 40 mm diam. after 7 days, white, circular, regular at the margin, mycelium dense and raised, villous, felt, reverse white, conidial masses orange.

Material examined – China, Guangdong Province, Guangzhou City, on leaf spot of *Alpinia zerumbet* (Pers.) Burt & Sm. (*Zingiberaceae*), February 2022, Jiachun Wang (living culture ZHKUCC 23-0832, new host record).

Additional materials – China, Guangdong Province, Guangzhou City, on leaf spot of *Epipremnum aureum* (Linden & André) G. S. Bunting (*Araceae*), June 2022, Jingwen Chen (living culture ZHKUCC 23-0833, new host record). China, Hunan Province, Changsha City, on leaf spot of *Hydrangea macrophylla* (Thunb.) Ser. (*Hydrangeaceae*), August 2022, Chao Chen (living culture ZHKUCC 23-0834, new host record). China, Guangdong Province, Guangzhou City, on leaf spot of *Thalia dealbata* Fraser (*Marantaceae*), June 2022, Jingwen Chen (living culture ZHKUCC 23-0835, new host record).

Note – In the multigene phylogenetic tree, the strains clustered together with *C. siamense* with 1.00 BYPP support (Fig. 10). Morphologically, our isolates were similar to *C. siamense* (Prihastuti et al. 2009). Therefore, we introduce our isolates as *C. siamense*. The type species of *C. siamense* were isolated from *Coffea arabica* (Prihastuti et al. 2009). This species has a wide range of hosts, including trees (*Bauhinia variegata*, *Chrysalidocarpus lutescens* and *Ficus elastica*) and commercial crops (*Citrus*, *Mangifera indica*, *Rosa chinensis* and *Psidium guajava*) (Liu et al.

2017, Sharma et al. 2015, Chou et al. 2019, Feng et al. 2019). However, this is the first report of *C. siamense* on *Alpinia zerumbet*, *Epipremnum aureum*, *Hydrangea macrophylla* and *Thalia dealbata*.

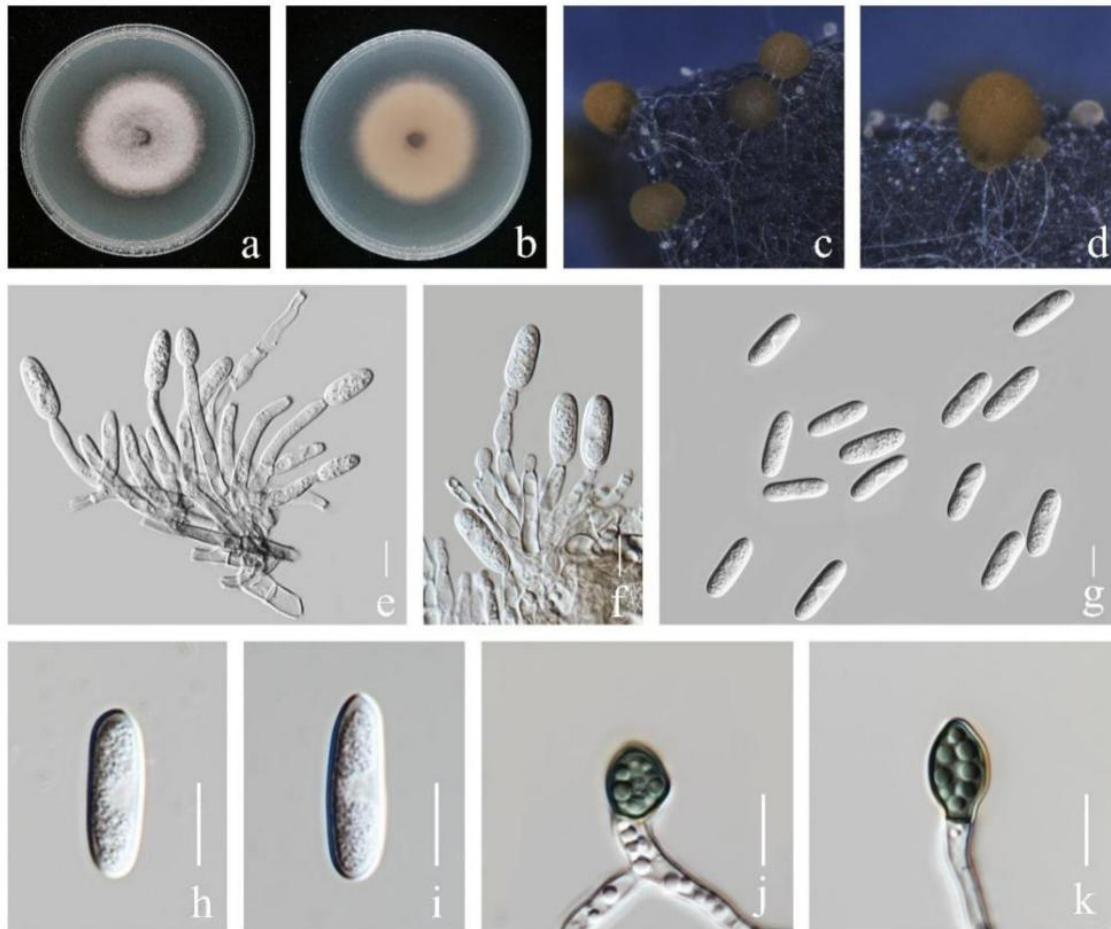


Figure 14 – *Colletotrichum siamense* (ZHKUCC 23-0835). a, b Upper and reverse view on PDA (7 d). c, d Conidial masses. e, f Conidiogenous cells. g–i Conidia. j, k Appressoria. Scale bars: e–k = 10 μ m.

Colletotrichum orchidearum species complex

Phylogenetic analysis

Phylogenetic trees were generated using combined ITS (542 bp), *gapdh* (212 bp), *chs-1* (265 bp), *act* (241 bp) and *tub2* (540 bp) sequence data. The tree topologies generated by ML and Bayesian were similar and the best-scoring ML tree is shown in Fig. 15. The sequence alignment comprised 32 taxa of representative strains, including nine isolates obtained in this study. *C. brevisporum* (BCC 38876) and *C. magnum* (CBS 519.97) were used as the outgroup taxa. The best-scoring ML tree had an optimization likelihood value of -4673.875725. The matrix had 268 distinct alignment patterns with a 2.79% proportion of gaps and completely undetermined characters. Estimated base frequencies were as follows: A = 0.220760, C = 0.308184, G = 0.256151, T = 0.214905; substitution rates: AC = 1.215559, AG = 3.534129, AT = 0.716204, CG = 0.711797, CT = 5.819183, GT = 1.000000; gamma distribution shape parameter α = 0.857303. Incomplete portions at the ends of the sequences were excluded from the analysis. In the resulted phylogenetic tree, ZHKUCC 23-0861, ZHKUCC 23-0862, ZHKUCC 23-0863, ZHKUCC 23-0864, ZHKUCC 23-0865, ZHKUCC 23-0866, ZHKUCC 23-0867, ZHKUCC 23-0868 and ZHKUCC 23-0869) grouped with *C. plurivorum*.

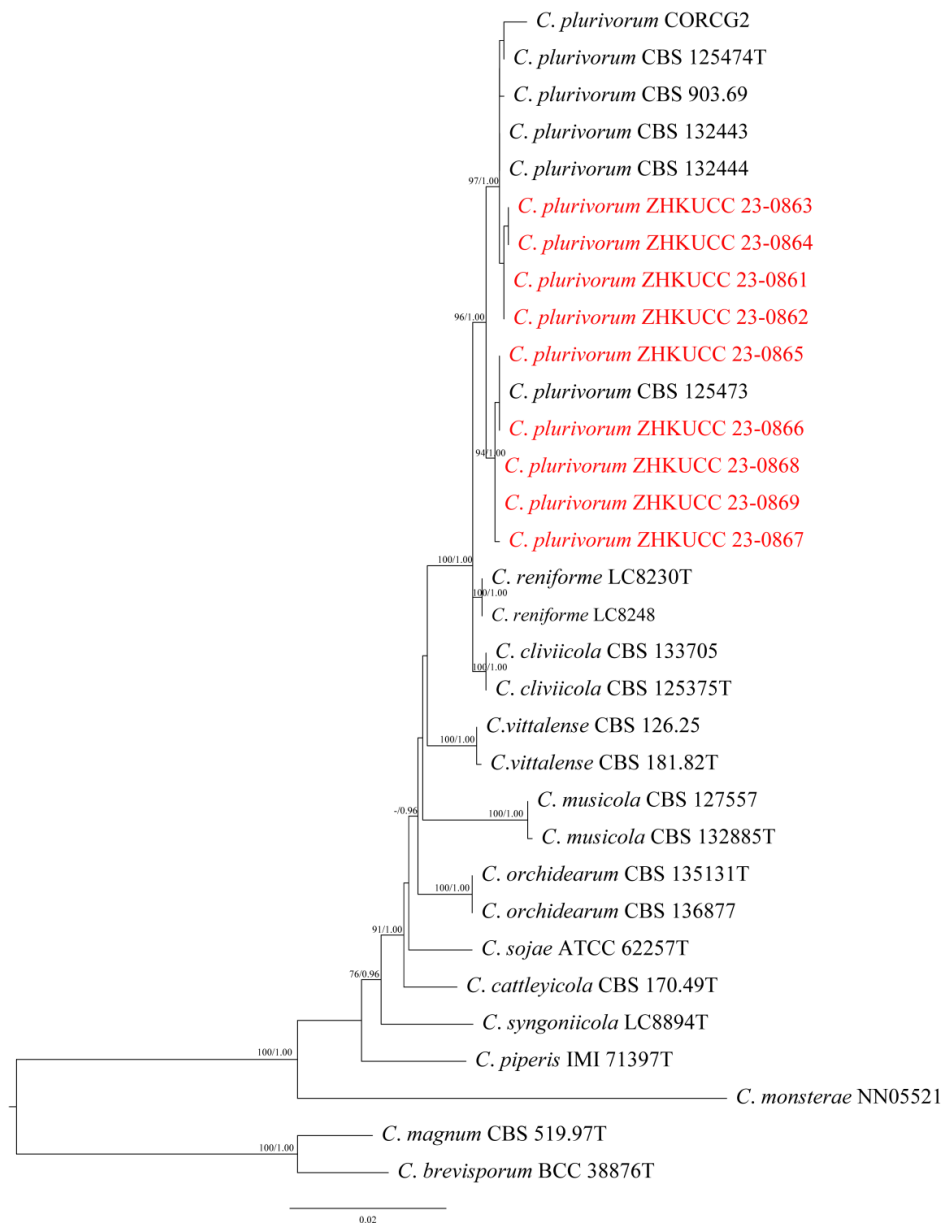


Figure 15 – Maximum likelihood tree of the *Colletotrichum orchidearum* species complex. *C. brevisporum* (BCC 38876) and *C. magnum* (CBS 519.97) were selected as the outgroup. At the nodes, bootstrap support values for ML ($\geq 75\%$) and BYPP (≥ 0.95) are displayed (ML/PP). Ex-type isolates are marked with “T” and the isolates in the study are marked in red. The scale bar indicates 0.02 nucleotide changes per site.

Taxonomy

Colletotrichum plurivorum U. Damm, Alizadeh & T. Sato, in Damm et al., *Studies in Mycology* 92: 31 (2018) Fig. 16

Index Fungorum number: IF824228; Facesoffungi number: FoF 10691

Associated with leaf spot of *Cymbidium sinense*. Sexual morph: *Ascomata* 120–200(–280) × (80–)110–160(–210) μm (\bar{x} = 180 × 140 μm , n = 30), formed after 3 weeks on SNA, solitary, superficial or immersed, black, subglobose to pyriform. *Asci* 45–65(–70) × 9–13 μm (\bar{x} = 54 × 11 μm , n = 50), cylindrical or clavate, broadly truncate at base, 8 spored. *Ascospores* 18–24 × 5–7 μm (\bar{x} = 21 × 6 μm , n = 50), biserially arranged, aseptate, hyaline, fusiform, with both ends rounded. Asexual morph not observed.

Culture characteristics – Colonies on PDA 90 mm diam. After 7 days, white, flat at the entire

margin, aerial mycelium lush, cottony, reverse dark green to brown, ascomata black.

Material examined – China, Guangdong Province, Zhongshan City, on leaf spot of *Cymbidium sinense* (Jack. Ex Andr.) Willd. (*Orchidaceae*), August 2021, Yunxia Zhang and Jingwen Chen (living cultures ZHKUCC 23-0861 and ZHKUCC 23-0862, new host record).

Additional materials – China, Guangdong Province, Zhongshan City, on leaf spot of *Paphiopedilum* sp. (*Orchidaceae*), August 2021, Yunxia Zhang and Jingwen Chen (living culture ZHKUCC 23-0863 and ZHKUCC 23-0864, new host record), China, Guangdong Province, Meizhou City, on leaf spot of *Cymbidium sinense* (*Orchidaceae*), October 2021, Jingwen Chen (living culture ZHKUCC 23-0865 and ZHKUCC 23-0866). China, Guangdong Province, Zhongshan City, on the stem of *Impatiens balsamina* Linn. (*Balsaminaceae*) October 2021, Jingwen Chen (living culture ZHKUCC 23-0867, ZHKUCC 23-0868 and ZHKUCC 23-0869, new host record).

Note – In the phylogenetic tree, our isolates clustered with *C. plurivorum*, as two distinct clusters. ZHKUCC 23-0861-65 clustered with *C. plurivorum* (CBS.125473) whereas ZHKUCC 23-0866-69 separately with 96% support in ML and 1.00 support in BYPP (Fig. 15). Morphologically, our isolates were similar to *C. plurivorum* (Damm et al. 2019). Thus, based on morphology and phylogenetic evidence our isolates were identified as *C. plurivorum*. This species has a wide host range, including commercial crops such as *Astragalus memranaceus*, *Arachis hypogaea*, *Coffea* sp., *Lycium chinense* and *Glycyrrhiza uralensis* (Fu et al. 2019, Damm et al. 2019, Liu et al. 2022). However, this is the first report of *C. plurivorum* isolated from *Cymbidium sinense*, *Paphiopedilum* sp., and *Impatiens balsamina*.

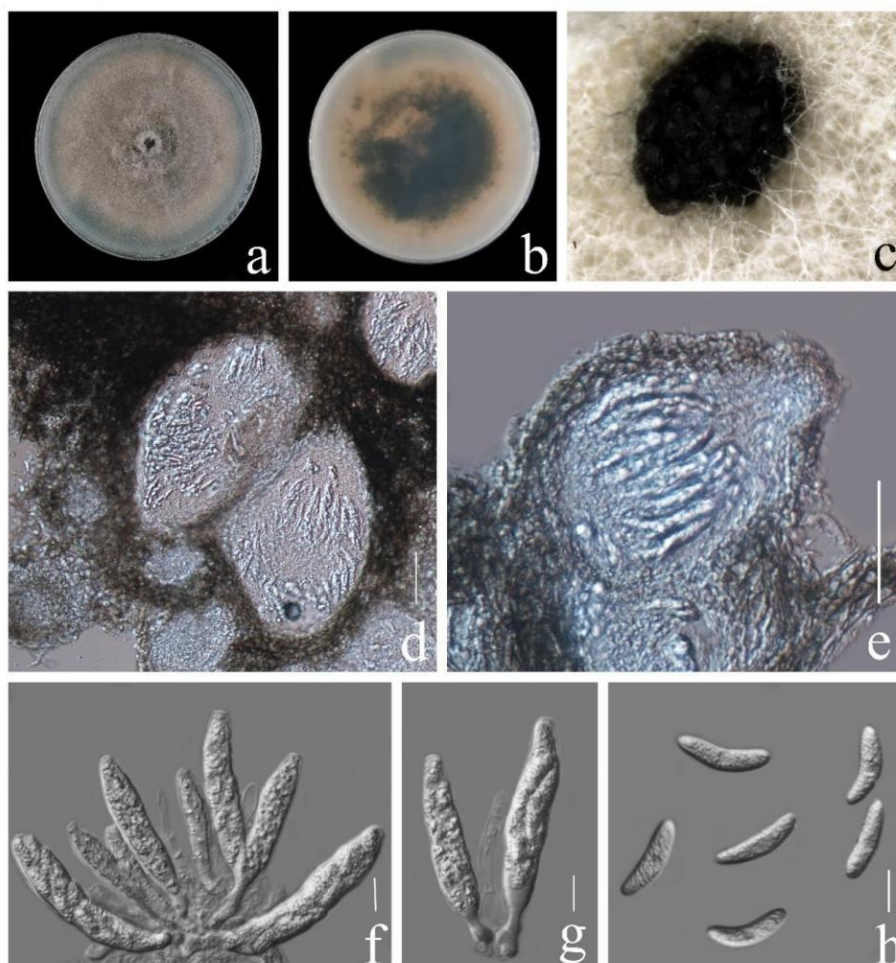


Figure 16 – *Colletotrichum plurivorum* (ZHKUCC 23-0861). a, b Upper and reverse view on PDA (7 d). c Ascomata on SNA. d, e Ascomata in longitudinal section. f, g Asci. h Ascospores. Scale bars: d = 20 μ m, e–j = 10 μ m.

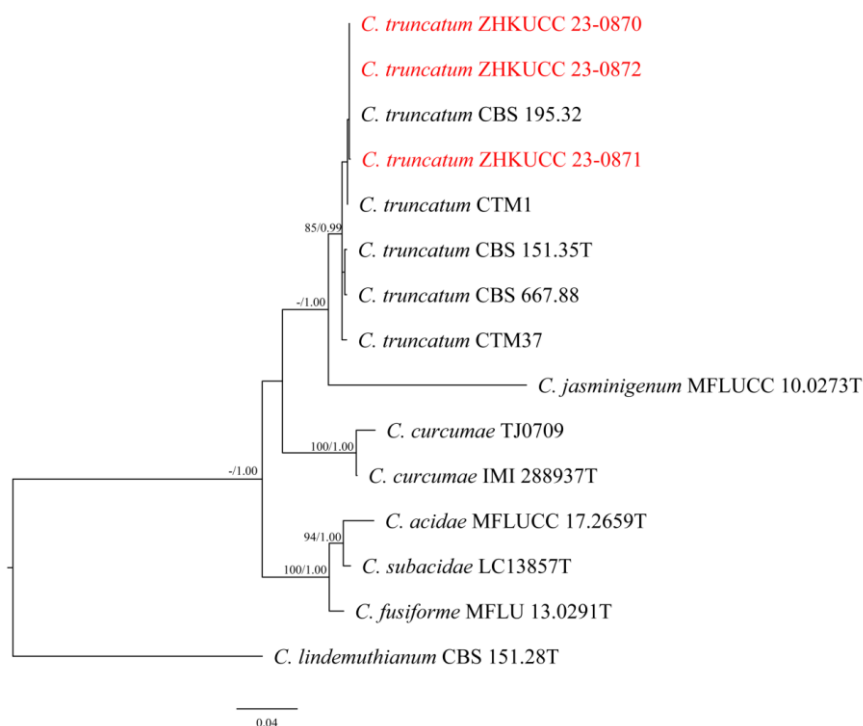


Figure 17 – Maximum likelihood tree of the *Colletotrichum truncatum* species complex. *C. lindemuthianum* (CBS 151.28) was selected as the outgroup. At the nodes, bootstrap support values for ML ($\geq 75\%$) and BYPP (≥ 0.95) are displayed (ML/PP). Ex-type isolates are marked with “T”, and the isolates in the study are marked in red. The scale bar indicates 0.04 nucleotide changes per site.

Colletotrichum truncatum species complex

Phylogenetic analysis

Phylogenetic trees were generated using combined ITS (518 bp), *gapdh* (284 bp), *chs-1* (244 bp), *act* (239 bp) and *tub2* (460 bp) sequence data. The tree topologies generated by ML and Bayesian were similar and the best-scoring ML tree is shown in Fig. 17. The sequence alignment comprised 15 taxa of representative strains, including three isolates obtained in this study. *Colletotrichum lindemuthianum* (CBS 151.28) was used as the outgroup taxon. The best-scoring ML tree had an optimization likelihood value of -5624.573762. The matrix had 365 distinct alignment patterns with a 6.21% proportion of gaps and completely undetermined characters. Estimated base frequencies were as follows: A = 0.240746, C = 0.282131, G = 0.236701, T = 0.240422; substitution rates: AC = 0.893367, AG = 2.930533, AT = 1.286451, CG = 0.7974581, CT = 4.114733, GT = 1.000000; gamma distribution shape parameter $\alpha = 0.614621$. Incomplete portions at the ends of the sequences were excluded from the analysis. Our isolates ZHKUCC 23-0870, ZHKUCC 23-0871 and ZHKUCC 23-0872 grouped with *C. truncatum* with 85% ML support and 0.99 BYPP support (Fig. 17).

Taxonomy

Colletotrichum truncatum (Schwein.) Andrus & W.D. Moore, *Phytopathology* 25: 121 (1935)

Fig. 18

Index Fungorum number: IF280780; Facesoffungi number: FoF 03827

Basionym: *Vermicularia truncata* Schwein. 1832

Saprobic on diseased leaves of *Catharanthus roseus*. Asexual morph: *Vegetative hyphae* hyaline, septate, branched, 1–8 μm diam. *Conidiomata* black, scattered or in groups. *Setae* (62–

)76–99(–141) × 5–8 μm (\bar{x} = 93 × 7 μm, n = 30), 3–4-septate, medium to dark brown, base cylindrical, tip broadly truncate. *Conidiophores* hyaline to pale brown, septate, branched, densely clustered, up to 90 μm long. *Conidiogenous cells* 6–20 × 2.5–4 μm (\bar{x} = 13 × 3.5 μm, n = 30), hyaline to pale brown, cylindrical. *Conidia* (20–)26 × 31(–34) × 4–5 μm (\bar{x} = 28 × 5 μm, n = 50), hyaline, smooth-walled to verruculose, aseptate, curved, ending abruptly at the round and truncate base, while tapering towards the acute and more strongly curved apex, contents granular. *Appressoria* 11–14(–17) × 7–11 μm (\bar{x} = 13 × 9, n = 30), solitary, in groups or dense clusters, pale to medium brown, entire edge to lobed, outline roundish to ellipsoidal or clavate, contact point of hyphae often above the appressorium.

Culture characteristics – Colonies on PDA 60 mm diam. after 7 days, flat with entire margin, aerial mycelium lush, cottony, yellow-white, hyaline towards margin, with concentric zonation, reverse dark vinaceous buff. Colonies on SNA 55 mm diam. after 7 days, white, conidial masses pale-yellow.

Material examined – China, Guangdong Province, Meizhou City, on leaf spot of *Catharanthus roseus* (L.) G. Don (*Apocynaceae*), October 2021, Jingwen Chen, (MHZU 23-0209 dried culture) living cultures ZHKUCC 23-0870; ZHKUCC 23-0871 and ZHKUCC 23-0872.

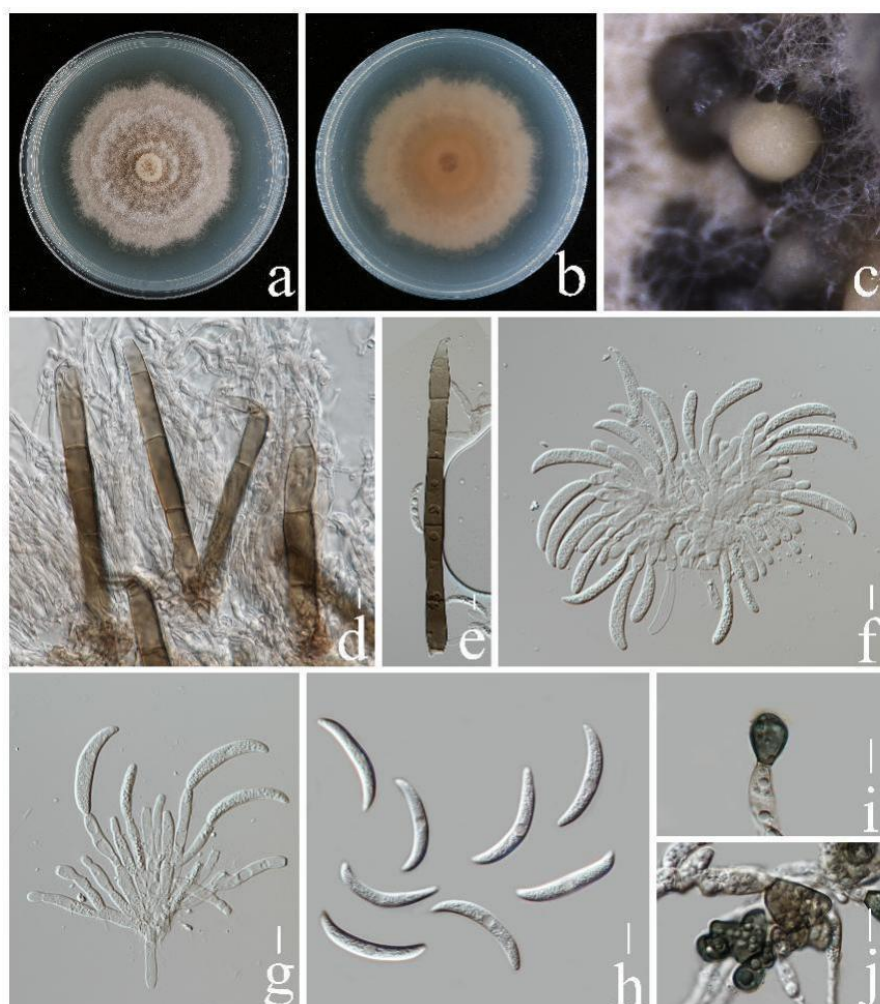


Figure 18 – *Colletotrichum truncatum* (ZHKUCC 23-0870). a, b Upper and reverse view on PDA (7 d). c Conidial masses on SNA. d, e Setae. f, g Conidiogenous cells. h Conidia. i, j Appressoria. Scale bars: d–j = 10 μm.

Note – In the multigene phylogenetic analysis of ITS, *gapdh*, *chs-1*, *act* and *tub2* sequence data, three isolates from this study formed a clade with *C. truncatum* with 85% in ML and 0.99 in BYPP values (Fig. 17). Our isolates differ from *C. truncatum* by faster growth (55 mm vs. 14 mm

diam. on SNA after 7 days) and larger conidia ($\bar{x} = 30 \times 5 \mu\text{m}$ vs $\bar{x} = 22 \times 4 \mu\text{m}$) (Damm et al. 2009). The nucleotide differences between *C. truncatum* (CBS 151.35) and our isolate (ZHKUCC 23-0870) are ITS: 0.19% (1/515 bp), *gapdh*: 1.64% (4/244 bp), *chs-1*: 2.07% (5/242 bp), *act*: 2.21% (5/226 bp), and *tub2*: 0.00% (0/450 bp) excluding gaps. Based on morphology, phylogeny, and sequence comparisons, three strains in this study are identified as *C. truncatum* associated with *Catharanthus roseus*.

Discussion

We isolated and identified 11 *Colletotrichum* species associated with various diseases on ornamental plants from South China. Our collection includes one new species and two new synonyms in *C. acutatum* species complex, two new synonyms for *C. gloeosporioides* and 18 new host records. These results showed that *Colletotrichum* species have a high diversity in ornamental plants in China. In addition, our study is a new addition to the number of *Colletotrichum* species which is now over 300 species. We predict that with the expansion of new hosts and locations, more species will be added to this genus in the future. Accurate species delimitation is important for disease control (Jayawardena et al. 2021b) thus, we assume our finding will facilitate ornamental plant diseases management in the future. This knowledge of fungal diversity can be expanded to understand the evolution which could help to understand fungal host relationships to address specific control measures (Jayawardena et al. 2021b; Manawasinghe et al. 2021). However, defining what is the species is mostly challenging for cryptic species such as *Colletotrichum* species and therefore, polyphasic approaches play a vital role in delineation of cryptic species (Manawasinghe et al. 2021). Therefore, methods combining morphological analysis and multi-loci phylogenetic analysis have increasingly been employed to define species boundaries in *Colletotrichum* (Jayawardena et al. 2020).

Colletotrichum species are classified into 16 species complexes (Jayawardena et al. 2021a). Among these complexes, the *C. acutatum*, *C. boninense* and *C. gloeosporioides* species complexes comprise more species with a wider range of hosts than other species complexes (Liu et al. 2022, Zhang et al. 2023). Coincidentally, most strains (33/45) collected in this study belong to these three species complexes. Nine out of 11 species belong to the *C. acutatum* species complex (including one new taxa), *C. boninense* (three species) and *C. gloeosporioides* species complexes (four species). Furthermore, 18 new host records in ornamental were found in this study and 14 new hosts were from these three species complexes. There are eight new host records in *C. gloeosporioides* complex; *Celosia cristata*, *Cymbidium sinense* and *Dendrobium nobile* are the new hosts of *C. fructicola*, while *Alpinia zerumbet*, *Epipremnum aureum*, *Hydrangea macrophylla* and *Thalia dealbata* are new hosts of *C. siamense*, *Bauhinia blakeana* is the new host of *C. endophyticum*. There are six new host records in *C. boninense* complex; *Dendrobium nobile* and *Cymbidium sinense* are the new hosts of *C. cymbidiicola*. *Chrysalidocarpus lutescens* is the new host of *C. chamaedoreae*. *Aglaonema* sp., *Cymbidium sinense*, and *Rosa chinensis* are the new hosts of *C. karsti*. There are three new host records in the *C. orchidearum* complex; *Cymbidium sinense*, *Impatiens balsamina* and *Paphiopedilum* sp. are the new hosts of *C. plurivorum*.

Colletotrichum siamense in the *C. gloeosporioides* species complex is the most common species (Liu et al. 2022), and a controversial and challenging taxon. Sharma et al. (2013) supported *C. siamense* as a species complex using *apn2* and *Mat1-2* gene (ApMat) and the *tef 1- α* gene analysis, including seven species (Sharma et al. 2015). Liu et al. (2016) proved it was a single species rather than a species complex based on genealogical concordance phylogenetic species recognition and coalescent methods. Zhang et al. (2023) synonymized *C. menglaense*, *C. pandanicola* and *C. parvisporum* with *C. siamense* as these four species cannot be well separated based on phylogeny. Six isolates from the present study clustered into two subclades in the *C. siamense* clade, which developed a distinct clade in the phylogenetic tree of *C. gloeosporioides* species complexes. Here we introduce our isolates as *C. siamense*.

In the present study, we also observed taxonomic uncertainties in several species complexes. In *Colletotrichum gloeosporioides* species complex our isolates showed a close relationship to

C. nanhuaense, they share a particular phylogenetic relationship with *C. gloeosporioides*, *C. dimorphum*, and *C. yunajiangenses* (Fig. 10). *Colletotrichum gloeosporioides* is the most diverse species in the *C. gloeosporioides* species complex. When we add more representative strains to the analysis, the taxonomic status of *C. dimorphum*, *C. yunajiangenses* and *C. nanhuaense*, becomes questionable. Morphologically these four species have overlapping characters (Table 1) and variations in sequence data are also lower than 1%. This suggests that these three species (*C. dimorphum*, *C. yunajiangenses* and *C. nanhuaense*) including our isolates could be possible genotypes of *C. gloeosporioides* thus we reduced *C. dimorphum*, and *C. nanhuaense* as synonyms of *C. gloeosporioides* and further studies are required to assure the taxonomic status of *C. yunajiangenses*.

Similar clustering pattern was also observed in the *C. acutatum* species complex. In this complex three isolates from our study were group with *C. fioriniae*, *C. orientalis* and *C. radermacherae*. *Colletotrichum orientalis* was introduced by Chen et al. (2022) and while introducing this species authors have provided only evidence from phylogenetic tree and the PHI analysis. However, in our study, it has been shown that *C. fioriniae* and *C. orientalis* share similar morphology (Table 1) as well there are no significant sequence differences between the type species. Subsequently, Zhang et al. (2023) introduced *C. radermacherae* to the same clade with lower sequence variations (with 99%–97% sequence similarities with *C. fioriniae*) and morphological variations are based on the size conidiogenous cells and conidia (Zhang et al. 2023). Furthermore, Zhang et al. (2023) has not accepted *C. orientalis* as a separate species while introducing *C. radermacherae*. Damm et al. (2012a) identified two well-separated clades *C. fioriniae* which was separated as *C. fioriniae* and *C. orientalis* by Chen et al. (2022). Thus, in our study, we observed four distinct clades in *C. fioriniae* including our isolates. Hence in our study, we accept the species concept of Damm et al. (2012a) and define *C. fioriniae* as well separated clades. We do not agree to introduce these clades as separate species due to the low variations in sequencing and morphology. These two cases in *C. acutatum* species complex and *C. gloeosporioides* species complex reflect the importance of considering inter-species diversity when we introduce a new species into *Colletotrichum*.

In addition, we observed an interesting cluster pattern in the *C. orchidearum* complex. In this complex (Fig. 3) *C. plurivorum* species developed four distinct groups. The important fact is none of these groups is either host-specific or locality species, for example, one group consists of isolates from *Cymbidium hookerianum* and *Coffea* from China and Brazil. Even though these species develop distinct groups, the average tree length for each group is similar. Based on the observation in this study, we propose that introducing novel *Colletotrichum* species requires following several important aspects including proper taxon sampling in the tree with all gene regions. However future studies are necessary to understand how much sequence variation and whether we need additional gene regions such as mating type gens to introduce new species into this complex. Furthermore, in the present study, we used five loci sequence analyses coupled with morphological characteristics, sequence data and PHI index in species delineation. However, there are several species and species complexes still difficult to identify using these sequence-based approaches. Therefore, whole genome analyses were proposed for further in-depth study on fungal classification (Liu et al. 2022, Zhang et al. 2023). Furthermore, we would like to propose additional gene regions such as mating type gene regions to incorporate in species delineation.

In conclusion, a significant number of *Colletotrichum* species associated with ornamental plant diseases in Southern China. The most are represented by the *C. acutatum* species complex, followed by the *C. boninense* and *C. gloeosporioides* species complexes. With the expansion of collection sites and hosts, we believe in the future there will be more novel species added to these most specious *Colletotrichum* complexes. Even though these species were isolated from diseased samples, the exact pathogenicity mechanisms are yet to be discovered. Therefore, future studies are required to understand the pathogenicity mechanisms of these pathogens as well as their species diversity.

Table 1 Morphological comparison of *Colletotrichum* species obtained in this study with their closely related species.

Species name	Conidia		Culture	References
	Shape	Size/ μm		
<i>C. gloeosporioides</i>	subcylindrical with bluntly rounded ends and slightly flattened base, some slightly constricted in the middle	12–17(–23.5) \times 4.5–6 \bar{x} = 14.4 \times 5.6	26.5 mm/d on PDA at 25 °C	Cannon et al. (2008)
<i>C. nanhuaense</i>	cylindrical, oblong to narrowly ovoid, obtuse at the base, rounded at the apex	10.5–16 \times 4.5–6 mean \pm SD = 14 \pm 1.1 \times 5.4 \pm 0.4	76–79 mm diam. on PDA at 25 °C in 7 days	Yu et al. (2022b)
<i>C. dimorphum</i>	cylindrical to oblong, attenuate at the base, rounded at the apex	10.5–19 \times 4–6 mean \pm SD = 14.6 \pm 2 \times 4.8 \pm 0.7	occupied the whole plate in 7 days at 25 °C	Yu et al. (2022b)
<i>C. yunajiangense</i>	cylindrical, obtuse at both ends or slightly acute at one end	10–14 \times 4–6 mean \pm SD = 12 \pm 0.9 \times 5.2 \pm 0.5	reaching 80 mm diam. on PDA in 7 days at 25 °C	Yu et al. (2022b)
<i>C. gloeosporioides</i>	cylindrical, one end rounded and one end acute	15–19 \times 4–7 \bar{x} = 17 \times 6	75 mm diam. on PDA at 25 °C after 7 days	This study
<i>C. fioriniae</i>	two different shape. mostly fusiform with both ends acute, some short-cylindric with both ends round	fusiform conidia, 12–15 \times 5–6; \bar{x} = 14 \times 6 short-cylindric conidia, 9–14 \times 4–7; \bar{x} = 12 \times 6	55 mm on PDA after 7 days at 25 °C	This study
<i>C. fioriniae</i>	fusiform, straight, fusiform to cylindrical with both ends acute	(10–)13.5–16.5(–19.5) \times 4–5(–5.5) mean \pm SD = 15.0 \pm 1.6 \times 4.5 \pm 0.3	22.5–23 mm on SNA in 7 days at 20 °C	Damm et al. (2012a)
<i>C. radermacheriae</i>	fusiform to cylindrical with both ends acute	14–21 \times 4.5–6 mean \pm SD = 17.4 \pm 1.5 \times 5.3 \pm 0.3	50 mm on PDA in 7 days at 25 °C	Zhang et al. (2023)
<i>C. orientalis</i>	fusiform or cylindrical with both ends acute	(12.8–)14–16(–18.5) \times (3.9–)4–5(–5.5) mean \pm SD = 15.1 \pm 1.2 \times 4.5 \pm 0.38	45–51 mm on PDA after 7 d	Chen et al. (2022)
<i>C. chrysalidocarpi</i>	straight, clavate, ends rounded	13–17 \times 4–6, \bar{x} = 15 \times 5	50 mm diam on PDA after 7 days growth at 25 °C	This study
<i>C. eriobotryae</i>	straight, fusiform to cylindrical, with both ends acute	(9–)11.5–15.5(–21.5) \times (3–)3.5–4(–4.5) mean \pm SD = 13.4 \pm 2.0 \times 4.0 \pm 0.3	25.5–30 mm in 7 days (36.5– \geq 40 mm in 10 days on SNA 20 °C	Damm et al. (2020)
<i>C. scovillei</i>	straight, cylindrical to clavate with one end round and one end \pm acute	(10.5–)12.5–15(–16.5) \times (3–)3.5–4(–4.5) mean \pm SD = 13.7 \pm 1.3 \times 3.8 \pm 0.3	22–22.5 mm in 7 days (33.5–35 mm in 10 d) on SNA 20 °C	Damm et al. (2012a)

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Supplementary Table 1 GenBank accession numbers used in this study.

Species	Strains	GenBank accession numbers				
		ITS	<i>gapdh</i>	<i>chs-1</i>	<i>act</i>	<i>tub2</i>
<i>C. abscissum</i>	COAD 1877	KP843126	KP843129	KP843132	KP843141	KP843135
<i>C. acerbum</i>	CBS 128530	JQ948459	JQ948790	JQ949120	JQ949780	JQ950110
<i>C. acidae</i>	MFLUCC 17-2659	MG996505	MH003691	MH003694	MH003697	MH003700
<i>C. acutatum</i>	CBS 112996	JQ005776	JQ948677	JQ005797	JQ005839	JQ005860
<i>C. aenigma</i>	ICMP 18608	JX010244	JX010044	JX009774	JX009443	JX010389
<i>C. aeschynomenes</i>	ICMP 17673	JX010176	JX009930	JX009799	JX009483	JX010392
<i>C. aeschynomenes</i>	COL02		MK792457			MK792460
<i>C. alatae</i>	CBS 304.67	JX010190	JX009990	JX009837	JX009471	JX010383
<i>C. alienum</i>	ICMP 12071	JX010251	JX010028	JX009882	JX009572	JX010411
<i>C. analogum</i>	YMF1.06943	OK030860	OK513663	OK513559	OK513599	OK513629
<i>C. annellatum</i>	CBS 129826	JQ005222	JQ005309	JQ005396	JQ005570	JQ005656
<i>C. aotearoa</i>	ICMP 18537	JX010205	JX010005	JX009853	JX009564	JX010420
<i>C. arboricola</i>	CBS 144795	MH817944	MH817950		MH817956	MH817962
<i>C. arecicola</i>	CGMCC 3.19667	MK914635	MW557464	MK935541	MK935374	MK935498
<i>C. artocarpicola</i>	MFLUCC 18-1167	MN415991	MN435568	MN435569	MN435570	MN435567
<i>C. asianum</i>	ICMP 18580	JX010196	JX010053	JX009867	JX009584	JX010406
<i>C. australe</i>	CBS 116478	JQ948455	JQ948786	JQ949116	JQ949776	JQ950106
<i>C. australianum</i>	VPRI 43075	MG572138	MG572127	MW091987	MN442109	MG572149
<i>C. beeveri</i>	CBS 128527	JQ005171	JQ005258	JQ005345	JQ005519	JQ005605
<i>C. boninense</i>	CBS 123755	JQ005153	JQ005240	JQ005327	JQ005501	JQ005588
<i>C. boninense</i>	CBS123755	JQ005153	JQ005240	JQ005327	JQ005501	JQ005588
<i>C. brasiliense</i>	CBS 128501	JQ005235	JQ005322	JQ005409	JQ005583	JQ005669
<i>C. brasiliense</i>	CBS128501	JQ005235	JQ005322	JQ005409	JQ005583	JQ005669
<i>C. brassicicola</i>	CBS 101059	JQ005172	JQ005259	JQ005346	JQ005520	JQ005606
<i>C. brevisporum</i>	BCC 38876	JN050238	MK862122	KF687760	JN050216	JN050244
<i>C. brisbanense</i>	CBS 292.67	JQ948291	JQ948621	JQ948952	JQ949612	JQ949942
<i>C. bromeliacearum</i>	LC0951	MZ595832	MZ664077	MZ799267	MZ664130	MZ673956
<i>C. cairnsense</i>	BRIP 63642	KU923672	KU923704	KU923710	KU923716	KU923688
<i>C. camelliae</i>	CGMCC:3.14925	KJ955081	KJ954782		KJ954363	KJ955230
<i>C. camelliae-japonicae</i>	CGMCC3.18118	KX853165	KX893584		KX893576	KX893580
<i>C. cangyuanense</i>	YMF1.04998	OK030865	OK513668	OK513564	OK513604	OK513634
<i>C. carthami</i>	SAPA100011	AB696998				AB696992
<i>C. castaneae</i>	GUCC 21268.4	OP722991	OP737973	OP715778	OP715812	OP720868
<i>C. catinaense</i>	CBS 142417	KY856400	KY856224	KY856136	KY855971	KY856482
<i>C. cattleyicola</i>	CBS 170.49	MG600758	MG600819	MG600866	MG600963	MG601025
<i>C. chamaedoreae</i>	NN052885	MZ595890	MZ664084	MZ799274	MZ664188	MZ674008

Supplementary Table 1 Continued.

Species	Strains	GenBank accession numbers				
		ITS	<i>gapdh</i>	<i>chs-1</i>	<i>act</i>	<i>tub2</i>
<i>C. chamaedoreae</i>	NN052890	MZ595891	MZ664086	MZ799275	MZ664189	MZ674009
<i>C. chamaedoreae</i>	ZHKUCC 23-0855	OR272087	OR493866	OR493838	OR493810	OR426927
<i>C. chamaedoreae</i>	ZHKUCC 23-0856	OR272088	OR493867	OR493839	OR493811	OR453353
<i>C. changpingense</i>	MFLUCC 15-0022	KP683152	KP852469	KP852449	KP683093	KP852490
<i>C. chrysalidocarpi</i>	ZHKUCC 23-0848	OR287501	OR493925	OR493908	OR493891	OR453377
<i>C. chrysalidocarpi</i>	ZHKUCC 23-0847	OR287500	OR493924	OR493907	OR493890	OR453376
<i>C. chrysanthemi</i>	IMI 364540	JQ948273	JQ948603	JQ948934	JQ949594	JQ949924
<i>C. chrysophilum</i>	CMM4268	KX094252	KX094183	KX094083	KX093982	KX094285
<i>C. ciggaro</i>	ICMP 18539	JX010230	JX009966	JX009800	JX009523	JX010434
<i>C. citri</i>	CBS 134233	KC293581	KC293741	KY856138	KY855973	KC293661
<i>C. citricola</i>	CBS 134228	KC293576	KC293736	KC293792	KC293616	KC293656
<i>C. clidemiae</i>	ICMP 18658	JX010265	JX009989	JX009877	JX009537	JX010438
<i>C. cliviicola</i>	CBS 125375	MG600733	MG600795	MG600850	MG600939	MG601000
<i>C. cliviicola</i>	CBS 133705	MG600732	MG600794	MG600849	MG600938	MG600999
<i>C. cliviigenum</i>	CPC 38800		MZ078178	MZ078161	MZ078143	MZ078260
<i>C. cobbittiense</i>	BRIP 66219a	MH087016	MH094133	MH094135	MH094134	MH094137
<i>C. colombiense</i>	CBS 129818	JQ005174	JQ005261	JQ005348	JQ005522	JQ005608
<i>C. condaoense</i>	CBS 134299	MH229914	MH229920	MH229926		MH229923
<i>C. conoides</i>	CAUG17	KP890168	KP890162	KP890156	KP890144	KP890174
<i>C. constrictum</i>	CBS 128504	JQ005238	JQ005325	JQ005412	JQ005586	JQ005672
<i>C. cordylinicola</i>	ICMP 18579	JX010226	JX009975	JX009864	HM470234	JX010440
<i>C. cosmi</i>	CBS 853.73	JQ948274	JQ948604	JQ948935	JQ949595	JQ949925
<i>C. costaricense</i>	CBS 330.75	JQ948180	JQ948510	JQ948841	JQ949501	JQ949831
<i>C. curcumae</i>	IMI 288937	GU227893	GU228285	GU228383	GU227991	GU228187
<i>C. curcumae</i>	J0709	MF278791	MF278793	MF278794	MF278796	MF278795
<i>C. cuscutae</i>	IMI 304802	JQ948195	JQ948525	JQ948856	JQ949516	JQ949846
<i>C. cymbidiicola</i>	IMI 347923	JQ005166	JQ005253	JQ005340	JQ005514	JQ005600
<i>C. cymbidiicola</i>	CBS 123757	JQ005168	JQ005255	JQ005342	JQ005516	JQ005602
<i>C. cymbidiicola</i>	CBS 128543	JQ005167	JQ005254	JQ005341	JQ005515	JQ005601
<i>C. cymbidiicola</i>	ZHKUCC 23-0852	OR272084	OR493863	OR493835	OR493807	OR453350
<i>C. cymbidiicola</i>	ZHKUCC 23-0853	OR272085	OR493864	OR493836	OR493808	OR453351
<i>C. cymbidiicola</i>	ZHKUCC 23-0854	OR272086	OR493865	OR493837	OR493809	OR453352
<i>C. dacrycarpi</i>	CBS 130241	JQ005236	JQ005323	JQ005410	JQ005584	JQ005670
<i>C. dimorphum</i>	YMF1.07303	OK030866	OK513669	OK513565	OK513605	OK513635
<i>C. dimorphum</i>	YMF1.07309	OK030867	OK513670	OK513566	OK513606	OK513636
<i>C. diversum</i>	LC11292	MZ595844	MZ664081	MZ799272	MZ664142	MZ673965

Supplementary Table 1 Continued.

Species	Strains	GenBank accession numbers				
		ITS	<i>gapdh</i>	<i>chs-1</i>	<i>act</i>	<i>tub2</i>
<i>C. doitungense</i>	MFLUCC 14-0128	MF448524	MH049480		MH376385	MH351277
<i>C. endophyticum</i>	MFLUCC 13-0418	KC633854	KC832854		KF306258	
<i>C. endophyticum</i>	MFLUCC 100676	KF242123	KF242181		KF157827	
<i>C. endophyticum</i>	YN1A3	KU251559	KU252013	KU251907	KU251640	KU252167
<i>C. endophyticum</i>	YN1A4	KU251561	KU252015	KU251909	KU251642	KU252169
<i>C. endophyticum</i>	YN1A5	KU251560	KU252014	KU251908	KU251641	KU252168
<i>C. endophyticum</i>	YTJ6	/	OK562584	/	OK562583	OK562585
<i>C. endophyticum</i>	ZHKUCC 23-0838	OR285930	OR493915	OR493898	OR493881	OR453367
<i>C. endophyticum</i>	ZHKUCC 23-0839	OR285931	OR493916	OR493899	OR493882	OR453368
<i>C. endophyticum</i>	ZHKUCC 23-0840	OR285932	OR493917	OR493900	OR493883	OR453369
<i>C. endophyticum</i>	ZHKUCC 23-0841	OR285939	OR493918	OR493901	OR493884	OR453370
<i>C. endophyticum</i>	ZHKUCC 23-0842	OR285940	OR493919	OR493902	OR493885	OR453371
<i>C. endophyticum</i>	ZHKUCC 23-0843	OR285941	OR493920	OR493903	OR493886	OR453372
<i>C. eriobotryae</i>	GLMC 1935	MF772487	MF795423	MN191653	MN191648	MF795428
<i>C. eriobotryae</i>	GLMC 1936	MF772488	MF795424	MN191654	MN191649	MF795429
<i>C. eriobotryae</i>	Cer012	MN197905	MN206737	MN206755	MN206749	MN206743
<i>C. eriobotryae</i>	Cer013	MN197906	MN206738	MN206756	MN206750	MN206744
<i>C. eriobotryae</i>	Cer014	MN197907	MN206739	MN206757	MN206751	MN206745
<i>C. eriobotryae</i>	Cer015	MN197908	MN206740	MN206758	MN206752	MN206746
<i>C. feijoicola</i>	CBS 144633	MK876413	MK876475		MK876466	MK876507
<i>C. fioriniae</i>	CBS 128517	JQ948292	JQ948622	JQ948953	JQ949613	JQ949943
<i>C. fioriniae</i>	CBS 129932	JQ948295	JQ948625	JQ948956	JQ949616	JQ949946
<i>C. fioriniae</i>	IMI 324996	JQ948301	JQ948631	JQ948962	JQ949622	JQ949952
<i>C. fioriniae</i>	CBS 126526	JQ948323	JQ948653	JQ948984	JQ949644	JQ949974
<i>C. fioriniae</i>	CBS 124958	JQ948306	JQ948636	JQ948967	JQ949627	JQ949957
<i>C. fioriniae</i>	ZHKUCC 23-0849	OR285933	OR493926	OR493909	OR493892	OR453378
<i>C. fioriniae</i>	ZHKUCC 23-0850	OR285934	OR493927	OR493910	OR493893	OR453379
<i>C. fioriniae</i>	ZHKUCC 23-0851	OR285935	OR493928	OR493911	OR493894	OR453380
<i>C. fructicola</i>	ICMP 18581	JX010165	JX010033	JX009866	FJ907426	JX010405
<i>C. fructicola</i>	ICMP 18613	JX010167	JX009998	JX009772	JX009491	JX010388
<i>C. fructicola</i>	ZHKUCC 23-0828	OR272047	OR493853	OR493825	OR493797	OR453340
<i>C. fructicola</i>	ZHKUCC 23-0829	OR272048	OR493854	OR493826	OR493798	OR453341
<i>C. fructicola</i>	ZHKUCC 23-0830	OR272049	OR493855	OR493827	OR493799	OR453342
<i>C. fructicola</i>	ZHKUCC 23-0831	OR272050	OR493856	OR493828	OR493800	OR453343
<i>C. fructivorum</i>	CBS 133125	JX145145				JX145196
<i>C. fusiforme</i>	MFLU 13-0291	KT290266	KT290255	KT290253	KT290251	KT290256
<i>C. gardeniae</i>	GUCC 12049	OP722995	OP737963	OP715766	OP715801	OP720858

Supplementary Table 1 Continued.

Species	Strains	GenBank accession numbers				
		ITS	<i>gapdh</i>	<i>chs-1</i>	<i>act</i>	<i>tub2</i>
<i>C. gloeosporioides</i>	CBS 112999	JQ005152	JQ005239		JQ005500	JQ005587
<i>C. gloeosporioides</i>	CBS 19121	JX010148	JX010054	JX009903	JX009558	
<i>C. gloeosporioides</i>	CS010	MK215704	/	LC660190	LC660210	LC628909
<i>C. gloeosporioides</i>	ICMP 12939	EU149938	JX009931	JX009747	JX009462	/
<i>C. gloeosporioides</i>	ICMP 12066	JX010158	JX009955	JX009888	JX009550	/
<i>C. gloeosporioides</i>	ZHKUCC 23-0844	OR285942	OR493921	OR493904	OR493887	OR453373
<i>C. gloeosporioides</i>	ZHKUCC 23-0845	OR285943	OR493922	OR493905	OR493888	OR453374
<i>C. gloeosporioides</i>	ZHKUCC 23-0846	OR285944	OR493923	OR493906	OR493889	OR453375
<i>C. godetiae</i>	CBS 133.44	JQ948402	JQ948733	JQ949063	JQ949723	JQ950053
<i>C. gracile</i>	YMF1.06939	OK030868	OK513671	OK513567	OK513607	OK513637
<i>C. grevilleae</i>	CBS 132879	KC297078	KC297010	KC296987	KC296941	KC297102
<i>C. grossum</i>	CAUG7	KP890165	KP890159	KP890153	KP890141	KP890171
<i>C. guajavae</i>	IMI 350839	JQ948270	JQ948600	JQ948931	JQ949591	JQ949921
<i>C. guajavae</i>	YMF 1.07334	OK030896	OK513697		OK513627	
<i>C. hebeiense</i>	MFLUCC13-0726	KF156863	KF377495	KF289008	KF377532	KF288975
<i>C. hedericola</i>	MFLU 15-0689	MN631384		MN635794	MN635795	
<i>C. helleniense</i>	CBS 142418	KY856446	KY856270	KY856186	KY856019	KY856528
<i>C. henanense</i>	CGMCC 3.17354	KJ955109	KJ954810		KM023257	KJ955257
<i>C. hippeastri</i>	CBS 125376	JQ005231	JQ005318	JQ005405	JQ005579	JQ005665
<i>C. horii</i>	ICMP 10492	GQ329690	JX009964	JX009752	JX009438	JX010450
<i>C. hystricis</i>	CBS 142411	KY856450	KY856274	KY856190	KY856023	KY856532
<i>C. indonesiense</i>	CBS 127551	JQ948288	JQ948618	JQ948949	JQ949609	JQ949939
<i>C. jasminigenum</i>	MFLUCC 10-0273	HM131513	HM131499		HM131508	HM153770
<i>C. javanense</i>	CBS 144963a	MH846576	MH846572	MH846573	MH846575	MH846574
<i>C. jiangxiense</i>	CGMCC 3.17363	KJ955201	KJ954902		KJ954471	KJ955348
<i>C. johnstonii</i>	CBS 128532	JQ948444	JQ948775	JQ949105	JQ949765	JQ950095
<i>C. kahawae</i>	ICMP17816	JX010231	JX010012	JX009813	JX009452	JX010444
<i>C. karsti</i>	CORCG6	HM585409	HM585391	HM582023	HM581995	HM585428
<i>C. karsti</i>	CBS 129833	JQ005175	JQ005262	JQ005349	JQ005523	JQ005609
<i>C. karsti</i>	ZHKUCC 23-0857	OR286087	OR493868	OR493840	OR493812	OR453354
<i>C. karsti</i>	ZHKUCC 23-0858	OR286088	OR493869	OR493841	OR493813	OR453355
<i>C. karsti</i>	ZHKUCC 23-0859	OR286089	OR493870	OR493842	OR493814	OR453356
<i>C. karsti</i>	ZHKUCC 23-0860	OR286090	OR493871	OR493843	OR493815	OR453357
<i>C. kinghornii</i>	CBS 198.35	JQ948454	JQ948785	JQ949115	JQ949775	JQ950105
<i>C. kunmingense</i>	GUCC 12053	OP722975	OP737965	OP715769	OP715804	OP720861
<i>C. laticiphilum</i>	CBS 112989	JQ948289	JQ948619	JQ948950	JQ949610	JQ949940
<i>C. lauri</i>	MFLUCC 17-0205	KY514347	KY514344	KY514341	KY514338	KY514350

Supplementary Table 1 Continued.

Species	Strains	GenBank accession numbers				
		ITS	<i>gapdh</i>	<i>chs-1</i>	<i>act</i>	<i>tub2</i>
<i>C. ligustri</i>	GUCC 12111	OP722988	OP737968	OP715773	OP740216	OP720864
<i>C. limetticola</i>	CBS 114.14	JQ948193	JQ948523	JQ948854	JQ949514	JQ949844
<i>C. limonicola</i>	CBS 142410	KY856472	KY856296	KY856213	KY856045	KY856554
<i>C. lindemuthianum</i>	CBS 151.28	GU227800	GU228192	GU228290	GU227898	GU228094
<i>C. lupine</i>	CBS 109225	JQ948155	JQ948485	JQ948816	JQ949476	JQ949806
<i>C. magnum</i>	CBS 519.97	MG600769	MG600829	MG600875	MG600973	MG601036
<i>C. makassarensis</i>	CBS 143664a	MH728812	MH728820	MH805850	MH781480	MH846563
<i>C. makassarensis</i>	CPC 28555	MH728816	MH728822	MH805847	MH781477	MH846560
<i>C. melonis</i>	CBS 159.84	JQ948194	JQ948524	JQ948855	JQ949515	JQ949845
<i>C. mengyinense</i>	SAUCC200702	MW786742	MW846240	MW883686	MW883695	MW888970
<i>C. mengyinense</i>	SAUCC200983	MW786642	MW876476	MW883691	MW883700	MW888975
<i>C. monsterae</i>	NN055214	MZ595897	MZ664121	MZ799351	MZ664195	MZ674015
<i>C. musae</i>	ICMP19119	HQ596292	HQ596299	JX009896	HQ596284	HQ596280
<i>C. musicola</i>	CBS 132885	MG600736	MG600798	MG600853	MG600942	MG601003
<i>C. musicola</i>	CBS 127557	MG600737	MG600799	MG600854	MG600943	MG601004
<i>C. nanhuaense</i>	YMF1.04990	OK030871	OK513674	OK513570	OK513610	OK513640
<i>C. nanhuaense</i>	YMF1.04993	OK030870	OK513673	OK513569	OK513609	OK513639
<i>C. novae-zelandiae</i>	CBS 128505	JQ005228	JQ005315	JQ005402	JQ005576	JQ005662
<i>C. nullisetosum</i>	YMF1.06946	OK030872	OK513675	OK513571	OK513611	OK513641
<i>C. nupharicola</i>	ICMP 18187	JX010187	JX009972	JX009835	JX009437	JX010398
<i>C. nymphaeae</i>	CBS 515.78	JQ948197	JQ948527	JQ948858	JQ949518	JQ949848
<i>C. oblongisporum</i>	YMF1.06938	OK030874	OK513677	OK513573		OK513643
<i>C. oncidii</i>	CBS 129828	JQ005169	JQ005256	JQ005343	JQ005517	JQ005603
<i>C. oncidii</i>	CBS 130242	JQ005170	JQ005257	JQ005344	JQ005518	JQ005604
<i>C. orchidearum</i>	CBS 135131	MG600738	MG600800	MG600855	MG600944	MG601005
<i>C. orchidearum</i>	CBS 136877	MG600739	MG600801	MG600856	MG600945	MG601006
<i>C. orchidophilum</i>	CBS 632.80	JQ948151	JQ948481	JQ948812	JQ949472	JQ949802
<i>C. orchidophilum</i>	CBS 631.80	JQ948152	JQ948482	JQ948813	JQ949473	JQ949803
<i>C. orientalis</i>	F10PGBYS1	KF772134	KF772104	KF772074	KF772044	KF772164
<i>C. orientalis</i>	F10PGBYS2	KF772135	KF772105	KF772075	KF772045	KF772165
<i>C. orientalis</i>	F10PGBYS8	KF772139	KF772109	KF772079	KF772049	KF772169
<i>C. pandanicola</i>	MFLUCC 17-0571	MG646967	MG646934	MG646931	MG646938	MG646926
<i>C. pandanicola</i>	SAUCC201152	MW786746	MW876478	MW883693	MW883702	MW888977
<i>C. pandanicola</i>	SAUCC200204	MW786641	MW846239	MW883685	MW883694	MW888969
<i>C. paranaense</i>	CBS 134729	KC204992	KC205026	KC205043	KC205077	KC205060
<i>C. parsonsiae</i>	CBS 128525	JQ005233	JQ005320	JQ005407	JQ005581	JQ005667
<i>C. parvisporum</i>	YMF1.06942	OK030876	OK513679	OK513575	OK513613	OK513645

Supplementary Table 1 Continued.

Species	Strains	GenBank accession numbers				
		ITS	<i>gapdh</i>	<i>chs-1</i>	<i>act</i>	<i>tub2</i>
<i>C. paxtonii</i>	IMI 165753	JQ948285	JQ948615	JQ948946	JQ949606	JQ949936
<i>C. perseae</i>	CBS 141365	KX620308	KX620242		KX620145	KX620341
<i>C. petchii</i>	CBS 378.94	JQ005223	JQ005310	JQ005397	JQ005571	JQ005657
<i>C. philodendricola</i>	CGMCC3.19290	MH105257	MH105261	MH105265	MH105273	MH105277
<i>C. phormii</i>	CBS 118194	JQ948446	JQ948777	JQ949107	JQ949767	JQ950097
<i>C. phyllanthi</i>	CBS 175.67	JQ005221	JQ005308	JQ005395	JQ005569	JQ005655
<i>C. piperis</i>	IMI 71397	MG600760	MG600820	MG600867	MG600964	MG601027
<i>C. plurivorum</i>	CBS 125474	MG600718	MG600781	MG600841	MG600925	MG600985
<i>C. plurivorum</i>	CORCG2	HM585397	HM585380	HM582024	HM581985	HM585422
<i>C. plurivorum</i>	CBS 125473	MG600717	MG600780	MG600840	MG600924	MG600984
<i>C. plurivorum</i>	CBS 132443	MG600719	MG600782	MG600842	MG600926	MG600986
<i>C. plurivorum</i>	CBS 132444	MG600720	MG600783	MG600843	MG600927	MG600987
<i>C. plurivorum</i>	ZHKUCC 23-0861	OR286370	OR493872	OR493844	OR493816	OR453358
<i>C. plurivorum</i>	ZHKUCC 23-0862	OR286371	OR493873	OR493845	OR493817	OR453359
<i>C. plurivorum</i>	ZHKUCC 23-0863	OR286372	OR493874	OR493846	OR493818	OR453360
<i>C. plurivorum</i>	ZHKUCC 23-0864	OR286373	OR493875	OR493847	OR493819	OR453361
<i>C. plurivorum</i>	ZHKUCC 23-0865	OR286374	OR493876	OR493848	OR493820	OR453362
<i>C. plurivorum</i>	ZHKUCC 23-0866	OR286375	OR493877	OR493849	OR493821	OR453363
<i>C. plurivorum</i>	ZHKUCC 23-0867	OR286376	OR493878	OR493850	OR493822	OR453364
<i>C. plurivorum</i>	ZHKUCC 23-0868	OR286377	OR493879	OR493851	OR493823	OR453365
<i>C. plurivorum</i>	ZHKUCC 23-0869	OR286378	OR493880	OR493852	OR493824	OR453366
<i>C. proteae</i>	CBS 132882	KC297079	KC297009	KC296986	KC296940	KC297101
<i>C. pseudoboninense</i>	CGMCC3.19755	MK796540	MK796573		MK796547	MK796554
<i>C. pseudotheobromicola</i>	MFLUCC 18-1602	MH817395	MH853675	MH853678	MH853681	MH853684
<i>C. psidii</i>	CBS 145.29	JX010219	JX009967	JX009901	JX009515	JX010443
<i>C. pyricola</i>	CBS 128531	JQ948445	JQ948776	JQ949106	JQ949766	JQ950096
<i>C. queenslandicum</i>	ICMP 1778	JX010276	JX009934	JX009899	JX009447	JX010414
<i>C. radermacherae</i>	GZCC 21-0813	OP723052	OP737966	OP715771	OP715806	OP720862
<i>C. radermacherae</i>	GZCC 21-0814	OP723053	OP737967	OP715772	OP715807	OP720863
<i>C. reniforme</i>	LC8230	MZ595847	MZ664110	MZ799290	MZ664145	MZ673968
<i>C. reniforme</i>	LC8248	MZ595850	MZ664111	MZ799295	MZ664148	MZ673971
<i>C. rhexiae</i>	CBS 133134	JX145128				JX145179
<i>C. rhombiforme</i>	CBS 129953	JQ948457	JQ948788	JQ949118	JQ949778	JQ950108
<i>C. roseum</i>	CBS 145754	MK903611	MK903603		MK903604	MK903607
<i>C. salicis</i>	CBS 607.94	JQ948460	JQ948791	JQ949121	JQ949781	JQ950111
<i>C. salsolae</i>	ICMP 19051	JX010242	JX009916	JX009863	JX009562	JX010403
<i>C. schimae</i>	NN046984	MZ595885	MZ664105	MZ799347	MZ664183	MZ674003

Supplementary Table 1 Continued.

Species	Strains	GenBank accession numbers				
		ITS	<i>gapdh</i>	<i>chs-1</i>	<i>act</i>	<i>tub2</i>
<i>C. scovillei</i>	CBS 126529	JQ948267	JQ948597	JQ948928	JQ949588	JQ949918
<i>C. scovillei</i>	CBS 126530	JQ948268	JQ948598	JQ948929	JQ949589	JQ949919
<i>C. siamense</i>	ICMP 18578	JX010171	JX009924	JX009865	FJ907423	JX010404
<i>C. siamense</i>	ICMP 18118	JX010163	JX009941	JX009843	JX009505	JX010402
<i>C. siamense</i>	ZHKUCC 23-0832	OR272065	OR493857	OR493829	OR493801	OR453344
<i>C. siamense</i>	ZHKUCC 23-0833	OR272066	OR493858	OR493830	OR493802	OR453345
<i>C. siamense</i>	ZHKUCC 23-0834	OR272067	OR493859	OR493831	OR493803	OR453346
<i>C. siamense</i>	ZHKUCC 23-0835	OR272068	OR493860	OR493832	OR493804	OR453347
<i>C. siamense</i>	ZHKUCC 23-0836	OR272069	OR493861	OR493833	OR493805	OR453348
<i>C. siamense</i>	ZHKUCC 23-0837	OR272070	OR493862	OR493834	OR493806	OR453349
<i>C. simmondsii</i>	CBS 122122	JQ948276	JQ948606	JQ948937	JQ949597	JQ949927
<i>C. simulanticitri</i>	YMF1.07302	OK030878	OK513680	OK513577	OK513615	
<i>C. sloanei</i>	IMI 364297	JQ948287	JQ948617	JQ948948	JQ949608	JQ949938
<i>C. sojae</i>	ACC 62257	MG600749	MG600810	MG600860	MG600954	MG601016
<i>C. sp.</i>	CBS 123921	JQ005163	JQ005250	JQ005337	JQ005511	JQ005597
<i>C. sp.</i>	BRIP 58074a	MK469999	MK470017	MW091975	MK470089	MK470053
<i>C. speciosum</i>	YMF1.07301	OK030881				
<i>C. subacidae</i>	LC13857	MZ595846	MZ664068	MZ799307	MZ664144	MZ673967
<i>C. subhenanense</i>	YMF1.06865	OK030883	OK513684	OK513581	OK513618	OK513647
<i>C. subsalicis</i>	LC13863	MZ852849		MZ799346	MZ664128	MZ673953
<i>C. syngoniicola</i>	LC8894	MZ595863	MZ664117	MZ799296	MZ664161	MZ673982
<i>C. syzygiicola</i>	MFLUCC 10-0624	KF242094	KF242156		KF157801	KF254880
<i>C. tainanense</i>	CBS 143666a	MH728818	MH728823	MH805845	MH781475	MH846558
<i>C. tamarilloi</i>	CBS 129814	JQ948184	JQ948514	JQ948845	JQ949505	JQ949835
<i>C. temperatum</i>	CBS 133122	JX145159				JX145211
<i>C. theobromicola</i>	ICMP 18649	JX010294	JX010006	JX009869	JX009444	JX010447
<i>C. ti</i>	ICMP 4832	JX010269	JX009952	JX009898	JX009520	JX010442
<i>C. tomentosae</i>	ZHKUCC 21-0103	OL708422	OL855850	OL855860	OL855870	OL855887
<i>C. torulosum</i>	CBS 128544	JQ005164	JQ005251	JQ005338	JQ005512	JQ005598
<i>C. tropicale</i>	CBS 124949	JX010264	JX010007	JX009870	JX009489	JX010407
<i>C. tropicale</i>	5101	GU994331		JX009870		JX010407
<i>C. truncatum</i>	CBS 151.35	GU227862	GU228254	GU228352	GU227960	GU228156
<i>C. truncatum</i>	CBS 151.35	GU227862	GU228254	GU228352	GU227960	GU228156
<i>C. truncatum</i>	CBS 667.88	GU227891	GU228283	GU228381	GU227989	GU228185
<i>C. truncatum</i>	CBS 195.32	GU227865	GU228257	GU228355	GU227963	GU228159
<i>C. truncatum</i>	CTM1	JX971124	KC109579	KC109539	JX975356	KC109459
<i>C. truncatum</i>	CTM37	JX971160	KC109615	KC109575	JX975392	KC109495

Supplementary Table 1 Continued.

Species	Strains	GenBank accession numbers				
		ITS	<i>gapdh</i>	<i>chs-1</i>	<i>act</i>	<i>tub2</i>
<i>C. truncatum</i>	ZHKUCC 23-0870	OR285936	OR493929	OR493912	OR493895	OR453381
<i>C. truncatum</i>	ZHKUCC 23-0871	OR285937	OR493930	OR493913	OR493896	OR453382
<i>C. truncatum</i>	ZHKUCC 23-0872	OR285938	OR493931	OR493914	OR493897	OR453383
<i>C. viniferum</i>	GZAAS5.08601	JN412804	JN412798		JN412795	JN412813
<i>C. vittalense</i>	CBS 181.82	MG600734	MG600796	MG600851	MG600940	MG601001
<i>C. vittalense</i>	CBS 126.25	MG600735	MG600797	MG600852	MG600941	MG601002
<i>C. walleri</i>	CBS 125472	JQ948275	JQ948605	JQ948936	JQ949596	JQ949926
<i>C. wanningense</i>	CGMCC 3.18936	MG830462	MG830318	MG830302	MG830270	MG830286
<i>C. watphraense</i>	MFLUCC 14-0123	MF448523	MH049479		MH376384	MH351276
<i>C. wuxiense</i>	CGMCC 3.17894	KU251591	KU252045	KU251939	KU251672	KU252200
<i>C. xanthorrhoeae</i>	ICMP 17903	JX010261	JX009927	JX009823	JX009478	JX010448
<i>C. yulongense</i>	CFCC 50818	MH751507	MK108986	MH793605	MH777394	MK108987
<i>C. yunajiangense</i>	YMF1.04996	OK030885	OK513686	OK513583	OK513620	OK513649
<i>C. yunajiangense</i>	YMF1.04997	OK030886	OK513687	OK513584	OK513621	OK513650

The strains and sequences in this study are in bold.