The Colletotrichum gigasporum species complex

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Abstract In a preliminary analysis, 21 Colletotrichum strains with large conidia preserved in the CBS culture collection clustered with a recently described species, C. gigasporum, forming a clade distinct from other currently known Colletotrichum species complexes. Multi-locus phylogenetic analyses (ITS, ACT, TUB2, CHS-1, GAPDH) as well as each of the single-locus analyses resolved seven distinct species, one of them being C. gigasporum. Colletotrichum gigasporum and its close allies thus constitute a previously unknown species complex with shared morphological features. Five of the seven species accepted in the C. gigasporum species complex are described here as novel species, namely C. arxii, C. magnisporum, C. pseudomajus, C. radialis and C. vietnamense. A species represented by a single sterile strain, namely CBS 159.50, was not described as novel species, and is treated as Colletotrichum sp. CBS 159.50. Furthermore, C. thailandicum is reduced to synonymy with C. gigasporum.

INTRODUCTION

Colletotrichum gigasporum was originally reported from healthy leaves of Centella asiatica in Madagascar and Stylosanthes guianensis in Mexico, as well as from Coffea arabica in Colombia (Rakotominina et al. 2013). It has an endophytic growth habit and could be isolated from various host plants occurring in geographically distant areas.

The most distinctive morphological feature of C. gigasporum is the long straight conidia (up to 32 μm long, av. length 26 μm). Rakotominina et al. (2013) discussed the morphological differences between C. gigasporum and other species that produce large conidia, e.g. C. crassipes, C. echinatum, C. macrosorum, C. taiwanense and C. vinosum. Based on phylogenetic analyses of ITS and TUB2 sequence data, they showed C. gigasporum to belong to a distinct clade, distant from other currently accepted Colletotrichum species.

Numerous Colletotrichum isolates detected in a blastn search on GenBank have similar ITS sequences to that of the ex-type strain of C. gigasporum, e.g. isolates from Coffea arabica in Vietnam (Nguyen et al. 2010), Hibiscus rosa-sinensis in Thailand (Noireung et al. 2012), Magnolia illifera in Thailand (Promputtha et al. 2007), Taxus chinesis var. mairei in China (Wu et al. 2013) and Theobroma cacao, Trichilia tuberculata and Virola surinamensis in Panama (Rojas et al. 2010). In our preliminary ITS analysis, 21 isolates retrieved from the CBS collection clustered with C. gigasporum, but showed considerable genetic variability, suggesting further species belonging to a previously unreported species complex.

The objectives of this study are to clarify the genetic and taxonomic relationships of Colletotrichum strains from various hosts and geographic areas thought to be closely related to C. gigasporum, and to describe the new species from this complex.

MATERIALS AND METHODS

Isolates

Colletotrichum isolates with large conidia were obtained from the culture collection of the CBS-KNAW Fungal Biodiversity Centre, Utrecht, the Netherlands (CBS). All descriptions are based on ex-type cultures. Features of other strains are added if deviant. Cultures of additional isolates used for morphological and phylogenetic analyses are maintained in the CBS culture collection (Table 1).

Morphological analysis

To enhance sporulation, 5-mm-diam plugs from the margin of actively growing cultures were transferred to the centre of 9-cm-diam Petri dishes containing synthetic nutrient-poor agar medium (SNA) (Nirenberg 1976) amended with autoclaved filter paper and double-autoclaved stems of Anthriscus sylvestris placed onto the agar surface. Strains were also studied after growth on oatmeal agar (OA). Cultures were incubated for 10 d at 20 °C under near UV light with a 12 h photoperiod. Measurements and photographs of characteristic structures were made according to methods described by Liu et al. (2012). Appresoria on hyphae were observed on the reverse side of colonies grown on SNA plates. Microscopic preparations were made in clear lactic acid, with 30 measurements per structure, and observed with a Nikon Eclipse 80i microscope using differential interference contrast (DIC) illumination. Colony characters and pigment production on SNA and OA incubated at 20 °C were noted after 10 d. Colony colours were scored according to Rayner (1970). Growth rates were measured after 7 and 10 d.

Phylogenetic analyses

Genomic DNA of the isolates was extracted using the method of Damm et al. (2008). Eight loci including the 5.8S nuclear ribosomal gene with the two flanking internal transcribed spacers...
| Species | Accession number¹ | Host / Substrate | Locality |
|---------|------------------|-----------------|----------|
| C. acutatum | CBS 112966, ATCC 56186* | Carica sp. | Australia |
| C. anthropodispora | CBS 125353* | Anthriscus sylvestris | Netherlands |
| C. arxii | CBS 169.59, IMI 304050, IMI 309371 | Oncidium excavatum | Netherlands |
| C. boninense | CBS 123511, MAFF 305972* | Paphiopedilum sp. | Germany |
| C. boninense | CBS 123755 | Crinum asiaticum var. sinicum | Japan |
| C. brevisporum | BCC 38376* | Neogalaga sp. | Thailand |
| C. chlorophyti | IMI 103806* | Chlorophyti sp. | India |
| C. circinans | CBS 111.21 | Allium cepa | USA |
| C. clavata | CBS 221.81 | Allium cepa | Germany |
| C. coccodes | CBS 125375, CSSK4* | Clivia miniata | China |
| C. cordoae | CBS 164.49 | Solanum tuberosum | Thailand |
| C. croceum | CBS 130355, FMR 6728 | Homo sapiens | Brazil |
| C. dactyloides | CBS 12497 | Theobroma cacao | Panama |
| C. dactyloides | CBS 125835, E2452 | Virola surinamensis | Panama |
| C. dactyloides | CBS 125475, L330a(T4) | Coffea sp. | Vietnam |
| C. dactyloides | CBS 125476, L330b(B2) | Coffea sp. | Vietnam |
| C. dactyloides | CBS 125730, 3386 | Coffea sp. | Vietnam |
| C. dactyloides | CBS 125731, E2452 | Theobroma cacao | Panama |
| C. dactyloides | CBS 132881, CPC 12084 | Acaica auriculiformis | Thailand |
| C. dactyloides | CBS 132884, CPC 1623 | Musa sp. | Mexico |
| C. dactyloides | CBS 133386, MUCU 44947* | Gentella acutiloba | Madagascar |
| C. dactyloides | CBS 159.75 | Musa sp. | India |
| C. dactyloides | CBS 181.52 | Theobroma cacao | East Africa |
| C. dactyloides | CBS 181.52, MUCU 44947* | Gentella acutiloba | Madagascar |
| C. drosophilae | CBS 953.97 | Citrus sinensis | Italy |
| C. drosophilae | CBS 130836, M 1.001* | Zea mays | USA |
| C. ferrugineum | CBS 132134, 3.14194* | Vanda sp. | China |
| C. flavoviride | CBS 523.97 | Phaseolus coccinus | Costa Rica |
| C. flavoviride | CBS 144.31* | Phaseolus vulgaris | Germany |
| C. flavoviride | CBS 125339 | Apiosegna | Czech Republic |
| C. flavoviride | CBS 125337* | Apiosegna | Czech Republic |
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| C. flavoviride | CBS 125337* | Apiosegna | Czech Republic |
The DNA sequences obtained from forward and reverse primers were used to obtain consensus sequences using MEGA v. 5.1 (Tamura et al. 2011), and subsequent alignments were generated using MAFFT v. 6 (Katoh & Toh 2010), and manually edited using MEGA v. 5.1.

Sequences of the 21 Colletotrichum strains studied in this paper as well as sequences of 50 reference strains (Table 1) downloaded from GenBank (www.ncbi.nlm.nih.gov/Genbank/) and NIAS GenBank (www.gene.affrc.go.jp/about_en.php) were included in individual alignments and eight single gene phylogenies were generated using a distance-based method. The ITS alignment included a further 22 sequences that were found in blast searches in GenBank in addition to those in Table 1. Distance matrixes of the aligned sequences were calculated using the Kimura 2-parameter model (Kimura 1980), and analysed with the Neighbour-joining (NJ) algorithm (Saitou & Nei 1987) using MEGA v. 5.1, excluding positions with gaps. The reliability of the inferred trees was estimated by bootstrap analyses with 1 000 replicates.

A maximum parsimony analysis was performed on the multi-locus alignment including five of the eight loci (ACT, CHS-1, GAPDH, ITS, TUB2) of a total of 71 strains (Table 1) using PAUP v. 4.0b10 (Swofford 2002). Ambiguously aligned regions were excluded from all analyses. Unweighted parsimony (UP) analysis was performed. Trees were inferred using the heuristic search option with TBR branch swapping and 1 000 random sequence additions. Maxtrees were unlimited, branches of zero length were collapsed and all multiple parsimonious trees were saved. Clade stability was assessed in a bootstrap analysis with 1 000 replicates, each with 10 replicates of random stepwise addition of taxa. A second phylogenetic analysis of the concatenated alignment using a Markov Chain Monte Carlo (MCMC) algorithm was done to generate trees with Bayesian posterior probabilities in MrBayes v. 3.2.1 (Ronquist & Huelsenbeck 2003). Nucleotide substitution models were determined using MrModeltest v. 2.3 (Nylander 2004) for each gene region and included in the analyses. Two analyses of four MCMC chains were run from random trees for 10 000 000 generations and sampled every 1 000 generations. The first 25 % of trees were discarded as the burn-in phase of each analysis and posterior probabilities determined from the remaining trees. Monilochaetes infuscans strain CBS 869.96 was used as outgroup in all analyses. Sequences derived in this study were lodged in GenBank, the multi-locus alignment and tree in TreeBASE (http://www.trebase.org/trebase-web/searchstudySearch.html) (S15175), and taxonomic novelties in MycoBank (www.MycoBank.org; Crous et al. 2004a).

RESULTS

Phylogenoy

The eight NJ trees derived from the single gene sequence alignments (ACT, CAL, CHS-1, GAPDH, GS, HIS3, ITS, TUB2)
confirmed that the 21 CBS isolates and the ex-type and other strains of *C. gigasporum* constituted a monophyletic lineage, distant from other known major clades of the genus *Colletotrichum* recognised by Cannon et al. (2012). The NJ trees are not shown in this study except for the phylogeny based on ITS data (Fig. 1). Isolates studied in this paper (marked with red squares) are separated into seven subclades, which could also be confirmed with the other seven single gene phylogenies. The multi-locus phylogenetic analysis included 70 ingroup strains, with *Monilochaetes infuscans* (CBS 869.96) as outgroup. The dataset of five loci comprised 1,512 characters including informative, 85 parsimony-uninformative and 728 constant.

- **AB738653** G. septospora MAFF 243176 Citrus Japan
- **AB738654** G. septospora MAFF 243177 Citrus Japan
- **HM852073** C. gloeosporioides ET100a Paulinia Ecuador
- **EF672326** Colletotrichum sp. Vega508 Coffea Colombia
- **GQ379686** C. incarnatum unknown
- **GQ396900** C. incarnatum unknown
- **JN196428** Colletotrichum sp. Z51 Tarus China
- **GU803511** C. septospora C07001 Capsicum Korea
- **GU935910** G. septospora C07005 Capsicum Korea
- **AB738850** G. septospora MAFF 242912 Homo sapiens Japan
- **JN050242** C. thailandicum Hibiscus Thailand
- **CBS 132884** Musa Mexico
- **CBS 109355** Homo sapiens Brazil
- **CBS 124947** Theobroma Panama
- **CBS 181.52** Theobroma East Africa
- **CBS 125385** Virola Panama
- **CBS 125387** Theobroma Panama
- **CMSP34** Alocasia Thailand
- **CBS 125476** Coffea Vietnam
- **AM982797** C. gigasporum Centella Madagascar
- **CBS 125730** Theobroma Panama
- **CBS 125731** Trichilia Panama
- **CBS 101881** Solanum New Zealand
- **AB738651** G. septospora MAFF 243031 Hederia Japan
- **AY438543** C. acutata IMI377313 Guipia Guyana
- **AY442184** C. orbiculare IMI377084 Mora Guyana
- **AY438553** C. orbiculare IMI377083 Mora Guyana
- **GU935907** G. septospora C030981 Capsicum Korea
- **CBS 132861** Acacia Thailand
- **JF602427** C. crassipes 1-98 Linder China
- **GU935909** G. septospora C07002 Capsicum Korea
- **GU935908** G. septospora C07001 Capsicum Korea
- **AB738648** G. septospora MAFF 238783 Chrysanthemum Japan
- **AB738649** G. septospora MAFF 242967 Diospyros Japan
- **AB738582** G. septospora MAFF 243173 Vanilla Japan
- **CBS 159.75** air and stored grain India
- **CBS 159.50** Derris Indonesia
- **CBS 571.88** Camellia Taiwan
- **CBS 125477** Coffea Vietnam
- **CBS 125478** Coffea Vietnam
- **CBS 529.93** root Costa Rica
- **CBS 398.84** unknown unknown
- **CBS 132511** Piptopodiokum Germany
- **CBS 169.89** Oncidium Netherlands
- **CBS 120709** C. truncatum
- **CBS 151.35** C. truncatum
- **CBS 953.97** C. gloeosporioides
- **CORC35** C. gloeosporioides
- **CBS 123755** C. boninense
- **CBS 128526** C. boninense
- **BRIP 28519** C. simmondsii
- **CBS 110996** C. acutatum
- **CBS 130836** C. graminicola
- **IMI 45525** C. verruculosum
- **CBS 164.49** C. coccodes
- **CBS 369.75** C. coccodes
- **BCC 38877** C. tropicola
- **BCC 38876** C. brevisporum
- **CBS 125375** C. citrata
- **CBS 144.31** C. lindenmuthianum
- **CBS 869.96** Monilochaetes infuscans

Fig. 1 Neighbour-joining tree of ITS sequences from 21 isolates generated in this study and 43 isolates from other studies, retrieved from GenBank. The tree was constructed using MEGA v.5.1 software. The Kimura-2-parameter method was used. Bootstrap support values (1,000 replicates) above 50% are shown at the nodes. Ex-type cultures are emphasised in bold, and include the taxonomic name as originally described. Our isolates are marked with a red square, and the strain number is followed by host and country of origin. Stars indicate reported pathogens, triangles indicate reported endophytes, GenBank accessions are followed by taxonomic name as originally identified, strain number, host and country of origin. The tree is rooted with *Monilochaetes infuscans*. The alignment gaps, of which 699 characters were parsimony-informative, 85 parsimony-uninformative and 728 constant. Parsimony analysis resulted in 94 most parsimonious trees, one of them (length = 3417, CI = 0.438, RI = 0.798, RC = 0.349, HI = 0.562) is shown in Fig. 2, where the 21 strains studied belong to a major clade consisting of seven subclades. More than half of the strains clustered in the largest subclade (*C. gigasporum*) with a high bootstrap support and Bayesian posterior probability value (100/1.00). The Bayesian tree confirmed the tree topology of the trees obtained with maximum parsimony.
Based on the results of the single and multi-locus phylograms, we accept seven species within the *Colletotrichum* species complex, including six species that are new to science. In addition, two recently described species are shown to be synonymous. All novel species are characterised and illustrated below except for a species which is represented by a single strain, CBS 159.50. Since this strain is sterile, we designate it as *Colletotrichum* sp. CBS 159.50.

### Taxonomy

**Colletotrichum arxii** F. Liu, L. Cai, Crous & Damm, sp. nov. — MycoBank MB807164; Fig. 3

**Etymology.** Named after Josef Adolf von Arx for his very substantial contribution to the classification of the genus *Colletotrichum*.

On *Anthriscus* stem. Vegetative *hyphae* hyaline, smooth-walled, septate, branched. *Conidiomata* acervular, conidiophores and setae formed on a cushion of roundish to angular brown cells. *Setae* pale to medium brown, smooth-walled to verruculose, 1–5-septate, 80–260 μm long, base cylindrical, 3.5–6 μm

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**Fig. 2** One of 206 most parsimonious trees obtained from a heuristic search of combined ACT, CHS-1, GAPDH, ITS and TUB2 gene sequences of *Colletotrichum* species. Bootstrap support values (1 000 replicates) above 50 % and Bayesian posterior probability values above 0.95 are shown at the nodes. Numbers of ex-type strains are emphasised in **bold**. Strain numbers studied are followed by host and country of origin. The tree is rooted with Monilochaetes infuscans.
diam, tip acute to obtuse. Conidiophores pale brown, septate, branched. Conidiogenous cells pale brown, cylindrical to clavate, 17.5–24 × 5–7 μm, opening 1–2.5 μm diam. Conidia hyaline, aseptate, smooth-walled, cylindrical to slightly curved, both ends rounded, 21–32 × 5.5–7.5 μm, av. ± SD = 28.1 ± 2.6 × 6.8 ± 0.5 μm, L/W ratio = 4.1; the other isolate CBS 169.59 forms relatively shorter conidia, 20–26.5 × 5.5–7.5 μm, av. ± SD = 23.1 ± 2 × 6.4 ± 0.5 μm, L/W ratio = 3.6.

On SNA. Vegetative hyphae hyaline to medium brown, smooth-walled, septate, branched. Conidiomata acervular. Setae pale to medium brown, smooth-walled to verruculose, 1–3-septate, 120–180 μm long, base cylindrical to inflated, 4.5–7.5 μm diam, tip acute. Conidiophores hyaline to pale brown, septate, branched. Conidiogenous cells hyaline to pale brown, cylindrical to clavate, 10–21.5 × 5.5–7.5 μm, opening 1.5–3 μm diam. Conidia hyaline, aseptate, smooth-walled, cylindrical to slightly curved, both ends rounded, (20–)24.5–30 × 5.5–7.5 μm, av. ± SD = 27.0 ± 1.8 × 6.7 ± 0.5 μm, L/W ratio = 4; the other isolate CBS 169.59 forms relatively shorter conidia, 15.5–24 × 5–7.5 μm, av. ± SD = 21.4 ± 2 × 6.3 ± 0.5 μm, L/W ratio = 3.4. Appressoria (few observed) pale brown, aseptate, solitary, with an ellipsoidal to irregular outline and a crenate or

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**Fig. 3** Colletotrichum arxii (CBS 132511). a, b. Acervuli; c, d. tips of setae; e–g. conidiophores; h, i. basal parts of setae; j–o. appressoria; p, q. conidia (a, d, f–g, i, q: from Anthriscus stem; b, c, e, h, j–p: from SNA. – a, b: DM; c–q: DIC). — Scale bars: a = 100 μm (applies to a, b); e = 10 μm (applies to c–q).
lobed margin, 4–11.5 × 4–9 μm, av. ± SD = 8.5 ± 2.5 × 6.0 ± 1.5 μm, L/W ratio = 1.4.

Culture characteristics — Colonies on OA flat with undulate margin, surface white, aerial mycelium lacking; reverse white; colonial diam 54–63 mm in 7 d, > 90 mm in 10 d. Colonies on SNA flat with erose or dentate margin, medium hyaline, buff around Anthuriscus stem, aerial mycelium lacking; colonial diam 68–77 mm in 7 d, > 90 mm in 10 d.

Specimens examined. GERMANY, Berlin, glasshouse, on living leaves of Paphiopedilum sp., Dec. 2010, U. Damm (holotype CBS H-21492, culture ex-type CBS 132511 = Paphi 2-1). — NETHERLANDS, Baarn, Cantonspark, on Oncidium excavatum, unknown collection date and collector (isolated by J.A. von Arx in 1956), culture CBS 169.59 = IMI 304050 = IMI 309371.

Notes — Although there are many Colletotrichum species reported from orchids, which include C. boninense (s.lat.), C. cinctum, C. cliviæ, C. cassinis, C. gloeoporioides (s.lat.), C. liriæ, C. lujae, C. macrosporum, C. ondici, C. orchidearum, C. orchidophilum, C. siamense, C. stanhopeae, C. vanillae (Stoneman 1898, Allescher 1902, Patel et al. 1953, von Arx 1957, Sutton 1980, Li 1999, Moriwaki et al. 2003, Talubnak & Soytong 2010, Yang et al. 2011, Damm et al. 2012a), C. arxi can be distinguished from these species either from phylogenetic data or morphological characteristics. Colletotrichum arxi is phylogenetically distinct from the C. acutatum, C. boniense and C. gloeoporioides complexes, as well as C. cliviæ and C. liriæ (Fig. 2), and could be morphologically distinguished from the other species that presently still lack molecular data.

Colletotrichum arxi differs from C. macrosporum, a species from an orchid from Brazil, by forming narrower conidia (C. mackrosporum 28–32 × 8–10 μm) (Saccardo 1896). Although C. orchidearum was originally described by Allescher (1902) from Munich, Germany, the same location as our strain CBS 132511, they can be differentiated from each other based on conidial size, with C. arxi forming significantly longer conidia than C. orchidearum (C. orchidearum (13.5–)15.5–19.5 × 5–6 μm, av. ± SD = 17.2 ± 1.6 × 5.5 ± 0.3 μm) (Damm et al. 2012a).

Colletotrichum cinctum (Berk. & M.A. Curtis) Stoneman was originally described from orchids, Oncidium sp. and Maxillaria sp. (Stoneman 1898) and also identified from Paphiopedilum insignis (specimen BPI 397219) in the USA (collected by J. Rubinger on 14 July 2021, unpubl.). Colletotrichum stanhopeae was described from Stanhopea sp. in Brazil (Hennings 1908), C. vanillae from Vanilla odorata in Italy (Saccardo 1906) and C. lujae from Luja in Belgium (Verplancke 1935). However, the conidia of these four species, C. cinctum (12–15 × 3–4 μm), C. stanhopeae (10–16 × 3.5–4 μm), C. vanillae (18–21 × 5.5–7 μm), C. lujae (9.3–10.5 × 2–3.1 μm) are significantly smaller than those of C. arxi (20–30 × 5.5–7.5 μm).

Closest match in a blastn search with the ITS sequence of strain CBS 132511 (with 99 % identity, 8 bp differences) was an endophytic isolate (DQ780412) from Magnolia lilifera probably in Thailand (Promputtha et al. 2007) and an endophytic isolate (FJ205460) from an orchid in Taiwan (Wang et al. unpubl. data). The closest match with the TUB2 sequence (with 97 % identity, 16 bp differences) was isolate MUCL 41702 from Orchis in Singapore (FN599826; Rakotoniaina & Munaut, unpubl. data).

Colletotrichum gigasporum E.F. Rakotoniaina & Munaut, Mycol. Progr. 12: 407. 2013

= Colletotrichum thailandicum Phouli., Noireung, L. Cai & K.D. Hyde, Cryptog. Mycol. 33: 354. 2012.

Notes — Colletotrichum gigasporum is characterised by large conidia ((22–)25–29–(32) × (6–)7–9 μm). Phylogenetic analyses by Rakotoniaina et al. (2013) based on the ITS and TUB2 sequences placed it in a distinct clade far from the currently accepted Colletotrichum species. Another species with large conidia (27–30 × 9–10 μm), C. thailandicum, was described from diseased Alocasia sp. and Hibiscus rosa-sinensis from Thailand (Noireung et al. 2012). Colletotrichum thailandicum is morphologically similar to C. gigasporum; the ITS and β-tubulin sequences of both fungi are identical or near-identical (differed in two nucleotide position in β-tubulin). In addition, phylogenetic analyses of single locus data, including ITS (Fig. 1), and multi-locus data (Fig. 2), show that the ex-type strains of the two species cluster together in one strongly supported clade. Since C. gigasporum was published online earlier (8 August 2012) than C. thailandicum (September 2012), we regard C. thailandicum as a synonym of C. gigasporum.

Strain CBS 109355, isolated from a phaeohyphomycotic cyst from a Brazilian man, was originally identified as C. crassipes, mainly based on morphology of the appressoria with crenate or deeply lobed margins and its size of conidia (Castro et al. 2001). In addition, strain CBS 159.75 and IMI 302450, which were deposited as C. crassipes in the CBS and IMI culture collections, were compared morphologically with CBS 109355 by Castro et al. (2001). However, strains CBS 159.75 and CBS 109355 were reidentified as C. gigasporum in the present study (Fig. 2). Hitherto, the taxonomic status of C. crassipes as well as the genetic relationship between C. gigasporum and C. crassipes remain unclear due to the lack of an ex-type culture and DNA sequence data. Thus, an epitype is needed to stabilise the nomenclature of C. crassipes.

In addition to being a disease-causing agent of humans, C. gigasporum is also associated with Musa sp. (Fig. 1, 2), the anthracnose of which is commonly considered to be caused by C. musae that belongs to the C. gloeoporioides species complex (Weir et al. 2012). However, C. gigasporum is phylogenetically distinct from C. musae, and its conidia are significantly larger than those of C. musae. Additional Colletotrichum species associated with Musa spp. include C. cavendishii, C. liukiensis and C. paxonii. Colletotrichum gigasporum differs from C. liukiensis (Sawada 1959), a species on leaves of M. liukiensis in Taiwan, and C. cavendishii (Petrak 1925), a species on living leaves of M. cavendishii by producing larger conidia (20.5–25.5 × 6–9 μm vs 12–14 × 4.8–5.5 μm and 10–19 × 4.5–7 μm, respectively). Colletotrichum paxonii, a species associated with banana in St. Lucia, belongs to the C. acutatum complex (Johnston & Jones 1997, Damm et al. 2012a) and is therefore not closely related to C. gigasporum.

Our 5-locus phylogram shows that several strains from diverse countries and hosts cluster with C. gigasporum (syn. C. thailandicum). Based on our blastn search in GenBank, the results of which are included in the ITS phylogeny, 22 additional ITS sequences from GenBank cluster with the ex-type strain of C. gigasporum, including sequences derived from strains isolated from plants as endophytes or pathogens and even strains that were isolated from human tissue (Fig. 1). This is in accordance with the conjecture that ecologically C. gigasporum can occur as either endophyte or pathogen (Rakotoniaina et al. 2013). The isolates from which most of these GenBank sequences were generated had been previously identified as C. crassipes, C. gloeoporioides, C. incarnatum, C. orbiculare or C. tawanense (sexual morph Glomerella septospora) (Fig. 1). The ascospores and conidia of C. gigasporum resemble those of C. tawanense with respect to their size. However, C. gigasporum produces asceptate conidia and 0–1-septate ascospores (Rakotoniaina et al. 2013), while the conidia of C. tawanense may become 1–5-septate with age and ascospores are mostly 3-septate and may become up to 6- or 8-septate when old (Sivanesan & Hsieh 1993). Colletotrichum tawanense, originally described from Styrrax formosanus in Taiwan, is currently poorly characterised using molecular methods (Hyde et al. 2009, 2012, 2013, Sivanesan & Hsieh 1993).
Fig. 4 *Colletotrichum magnisporum* (CBS 398.84). a, b. Acervuli; c, d. conidiophores; e, i, j. setae; f–h. conidia (a, d, g–j: from *Anthriscus* stem; b, c, e, f: from SNA. – a, b: DM; c–m: DIC). — Scale bars: a = 100 μm (applies to a, b); f = 10 μm (applies to c–j).
Cannon et al. 2012). Unfortunately, a subculture from the ex-
type isolate of C. taiwanense (IMI 353024) is contaminated; the
original strain could not be recovered. Several plant pathogen-
ics strains from various hosts (none of them from Syrinx) that were
previously identified as C. taiwanense were reidentified as
Colletotrichum based on the ITS-rDNA phylogram in this study
(Fig. 1). Colletotrichum gibbsorum differs from C. incarnatum
(Zimmermann 1901), a species first described from Coffea
liberica in Java, by producing larger conidia (20.5–25.5 × 6–9
μm vs 14–19 × 5 μm).

Some strains from Mora excelsa in Guyana had been previ-
ously identified as C. orbiculare (Lu et al. 2004) and grouped
with C. gibbsorum in our ITS tree. However, C. orbiculare was
recently redefined and shown to belong to a different species
complex together with C. lindenuthianum (Damm et al. 2013).

Although the ITS-rDNA phylogram revealed that C. gibbsorum
strains formed two subclades (Fig. 1), the bootstrap values are
too low to support two distinct species, which could also be
verified by the multi-locus phylogram (Fig. 2).

**Colletotrichum magnisporum** F. Liu, L. Cai, Crous & Damm, sp. nov. — MycoBank MB807165; Fig. 5

**Eymology.** Referring to the large size of its conidia.

On Anthriscus stem. Vegetative hyphae hyaline to brown,
smooth-walled, septate, branched. *Conidiozymate* acervular,
conidiophores and setae formed on a cushion of angular
brown cells. Setae medium to dark brown, smooth-walled to
verruculose, 0–4–septate, 42.5–105 μm long, base cylindrical
to inflated, 5.5–11.5 μm diam, tip acute to obtuse. *Conidio-
phores* hyaline to brown, septate, branched. *Conidigenous
cells* hyaline to medium brown, cylindrical or clavate, 18–33.5
× 5.5–10 μm, opening 1.5–2.5 μm diam. *Conidia* hyaline, aseptate,
smooth-walled, cylindrical with rounded ends, 28–39 × 8.5–
10.5 μm, av. ± SD = 33.8 ± 4.1 × 9.9 ± 0.6 μm, L/W ratio = 3.4. Not observed.

On SNA. Vegetative hyphae hyaline to medium brown, smooth-
walled, septate, branched. *Conidiozymate* acervular. Setae me-
dium to dark brown, smooth-walled to verruculose, 1–4–septate,
91.5–230.5 μm long, base cylindrical to inflated, 5–12.5 μm
diam, tip ± acute. *Conidiozymate* hyaline to medium brown,
septate, branched. *Conidigenous cells* hyaline to pale brown,
cylindrical to clavate, 17.5–26.5 × 7.5–9.5 μm, opening 1.5–2.5
μm diam. *Conidia* hyaline, aseptate, smooth-walled, cylindrical with rounded ends, 28.5–40.5 × 8.5–11 μm, av. ± SD = 34.3 ±
2.7 ± 9.7 ± 0.5 μm, L/W ratio = 3.5. *Appressoria* not observed.

**Culture characteristics** — Colonies on OA flat with entire
margin, surface iron-grey with a white margin, aseptate, septate
lacking; reverse olivaceous-grey to iron-grey; colonial diam
56–60 mm in 7 d, > 90 mm in 10 d. Colonies on SNA flat with
entire margin, medium hyaline, buff around Anthriscus stem,
aeriel mycelium lacking; colonial diam 64–65 mm in 7 d, > 90
mm in 10 d.

*Specimen examined.* Unknown collection details (deposited in CBS cul-
ture collection in June 1984) (holotype CBS H-21491, culture ex-type CBS
398.84).

Notes — Although *C. magnisporum* is represented by only
a single strain in this study, it could be distinguished from the
related species *C. arxii* based on its phylogenetic distance
(Fig. 2) and its morphology. The two species differ by 40 bp
differences in five genes totally, as well as a long insertion
(174 bp) in GAPDH sequences in *C. arxii* that is missing in
*Colletotrichum magnisporum*. In addition, the conidia of *C. arxii*
(24.5–30 × 5.5–7.5 μm, av. = 27 × 6.7 μm) are shorter and narrower than
*C. magnisporum* (28.5–40.5 × 8.5–11 μm, av. = 34.3 × 9.7 μm). For other comments see C. radicus.

The closest matches in a blastn search in GenBank with the
ITS sequence of strain CBS 398.84 were with 100 % identity
EF672323 from the endophytic isolate VegaE4-36 from Coffea
arabica from Hawaii, USA (Vega et al. 2010), EU686812
from an endophytic isolate from Rhipidocladum racemiflorum
from Panama (Higgins et al. 2011), as well as KF436311 from
the endophytic isolate TK780 from a tropical woody plant from
Panama (Higginbotham et al. 2013). The closest match with
the TUB2 sequence (with 96 % identity, 16 bp differences) was
isolated MUC41702 from *Orchis* in Singapore (FN599826;
Rakotoiniarina & Munaut unpubl. data).

**Colletotrichum pseudomajus** F. Liu, L. Cai, Crous & Damm, sp. nov. — MycoBank MB807165; Fig. 5

**Eymology.** Referring to its morphology, which resembles that of Glome-
rella major.

On OA. Vegetative hyphae medium brown, smooth-walled,
septate, branched. *Conidiozymate* acervular, conidiophores and
setae formed on a cushion of roundish brown cells. Setae me-
dium to dark brown, smooth-walled to verruculose, 0–3–septate,
100–215 μm long, base inflated to cylindrical, 4–8 μm diam,
tip acute. *Conidiozymate* hyaline to medium brown, septate,
septate, branched. *Conidigenous cells* hyaline to pale brown,
cylindrical to clavate, 12–18 × 4–6 μm, opening 1.5–2 μm diam. *Conidia*
hyaline, aseptate, smooth-walled, cylindrical with rounded ends,
occasionally slightly curved, 21.5–27 × 6–9 μm, av. ± SD = 24.3
± 1.5 × 7.8 ± 0.6 μm, L/W ratio = 3.1.

Sexual morph developed on OA. *Ascomata* globose, sometimes
subconical, black, surrounded with brown hairs, 95–165 μm
diam; ostiolate; neck, when present, 35–60 μm long; outer wall
composed of angular brown cells, 6–20 μm diam. *Interascal
tissue* composed of paraphyses, thin-walled, hyaline, septate,
the apex rounded. *Asci* cylindrical, 93–123.5 × 10.5–12.5 μm,
8-spored. *Ascospores* uni- or biseriately arranged, hyaline,
aseptate, smooth-walled, lunate, tip ± acute, 20–27.5 × 5–7
μm, av. ± SD = 24.2 ± 1.6 × 6.2 ± 0.4 μm, L/W ratio = 3.9.

On Anthriscus stem. Remaining sterile.

On SNA. Vegetative hyphae hyaline to medium brown, smooth-
walled, septate, branched. *Conidiozymate* acervular. Setae medium
to dark brown, smooth-walled to verruculose, 0–3–septate, 125–190
μm long, base cylindrical to inflated, 5.5–8 μm diam, tip acute. *Conidio-
phores* pale brown, septate, branched. *Conidigenous cells* pale brown,
cylindrical, clavate to bullet-shaped, 14.5–18 × 4–8 μm, opening 1.5–2 μm diam. *Conidia* hyaline, aseptate, smooth-walled, cylindrical with rounded ends, 22–30.5 × 6.5–
9.5 μm, av. ± SD = 26.3 ± 1.7 × 8.1 ± 0.5 μm, L/W ratio = 3.2. *Appressoria* not observed.

Sexual morph developed on SNA. *Ascomata* globose, subconi-
cal to obpyriform, black, surrounded with hyaline to medium
brown hairs, 260–360 μm diam, ostiolate; neck when present,
60–200 μm long; outer wall composed of angular brown cells,
5–15 μm diam. *Interascal tissue* composed of paraphyses, thin-walled, hyaline, septate,
the apex rounded. *Asci* cylindrical, 73.5–98.5 × 10–12.5 μm, 8-spored. *Ascospores* uni-
or biseriately arranged, hyaline, aseptate, smooth-walled, lunate,
tip ± acute, 18.5–25 × 4.5–7.5 μm, av. ± SD = 21.2 ± 1.5 × 6.0
± 0.7 μm, L/W ratio = 3.5.

**Culture characteristics** — Colonies on OA umbonate with
entire margin, surface iron-grey to greenish black, white aeral
mycelium; reverse olivaceous-grey; colonial diam 42–45 mm in
7 d, 65–68 mm in 10 d. Colonies on SNA flat with entire margin,
medium hyaline; colonial diam 40–47 mm in 7 d, 66–74 mm
in 10 d.

*Specimen examined.* TAUIN, on twig of *Camellia sinensis*, unknown col-
lection date and collector (isolated by J. Chen) (holotype CBS H-21493, cul-
ture ex-type CBS 571.88).
Fig. 5 Colletotrichum pseudomajus (CBS 571.88). a, f. Acervuli; b, c. tips of setae; d, l. conidiophores; e. paraphyses; g, h. basal parts of setae; j. outer surface of peridium; k, l. conidia; m, q, r. ascospores; n. ascomata; o, p. asci (a, b, d, e, g, j, k, m, n, p: from OA; c, f, h, i, l, o, q, r: from SNA. – a, f, n: DM; b–e, g–m, o–r: DIC). — Scale bars: f = 100 μm (applies to a, f, n); k = 10 μm (applies to b–e, g–m, o–r).
Weir et al. (2012) clarified the taxonomic status of *C. camelliae* f. sp. *camelliae* based on molecular analysis and pathogenicity tests, showing it to belong to the *G. gloeosporioides* complex. The phylogenetic analysis shows that strain CBS 571.88 (here referred to as *C. pseudomajus*) is phylogenetically distinct from the *G. gloeosporioides* complex. Additionally, *C. pseudomajus* differs from *G. cingulata* f. sp. *camelliae* in producing much larger conidia and ascospores (*C. pseudomajus*: conidia 22–30.5 × 6.5–9.5 μm and ascospores 18.5–25 × 4.5–7.5 μm vs *G. cingulata* f. sp. *camelliae*: conidia 11.3–21.8 × 3.5–6.9 μm and ascospores 10–13 × 3.5–4.5 μm) (Dickens & Cook 1989).

The name *C. camelliae*, although not listed by Hyde et al. (2009) and Cannon et al. (2012), is widely used for the causal agent of the brown blight disease of tea (Sosa de Castro et al. 2001, Muraleedharan & Baby 2007). However, the status of *C. camelliae* and its taxonomic relationship with *G. cingulata* f. sp. *camelliae* remain unresolved (Weir et al. 2012). There are 11 ITS sequences of *Colletotrichum* sp. from in GenBank (EF006386, FJ151507, EU723732, FJ216456, HQ382377, JQ089665, HQ382801, AB548281, AB218993, GQ916544, HE655519), of which sequence HQ382801 associated strain nested within the *C. boninense* complex in the ITS phylogenetic tree, while the others belong to several clades within the *G. gloeosporioides* complex (data not shown). Appropriate fresh collections associated with brown blight symptoms of tea from Sri Lanka are needed for epitypification to clarify the phylogenetic relationships of this taxon. *Colletotrichum pseudomajus* can be distinguished from *C. camelliae* by its significantly larger conidia (22–30.5 × 6.5–9.5 μm vs 15–17 × 4–5 μm).

*Colletotrichum pseudomajus* is morphologically similar to *G. major* except for the presence of paraphyses and the shape of its ascospores. Paraphyses were reported to be absent in *G. major*, but thin-walled, hyaline and septate paraphyses are present in *C. pseudomajus*; ascospores of *G. major* are ellipsoidal, not allantoid, with obtuse or subacute tips (Tunstall 1935), while those of *C. pseudomajus* are lunate, with more or less acute tips (Fig. 5). Currently, the phylogenetic position of *G. major* is unresolved due to the lack of an ex-type isolate. Thus, an epitope is needed to stabilise the nomenclature of *G. major* and to clarify the relationship between *C. pseudomajus* and *G. major*.

The closest matches in a blastn search with the ITS sequence of CBS 571.88 with 100 % identity were JX009424, the sequence generated from the same isolate by Weir et al. (2012), and JQ089667 from the endophytic isolate JD08-18 from *Camellia sinensis* in China (Fang et al. 2013), as well as JN418782 from the endophytic isolate E10202g from *Otoba parvifolia* in Ecuador (Barba et al. unpubl. data). Closest match with the TUB2 sequence (with 93 % identity, 32 bp differences) was isolate MUCL 41702 from *Orchis* in Singapore (FN599826; Rakotoniriana & Munaut unpubl. data). The blastn search with the GPDH sequence of CBS 571.88 showed similarity with JN050231 (85 % identity, 34 bp differences) from isolate BCC 38879 from *Hibiscus rosa-sinensis* in Thailand (Noireung et al. 2012) which is here referred to *C. gigasporum*, and JX009422 (99 % identity, 1 bp difference), a sequence generated from the same isolate. The only base difference in the end of the sequence was due to sequencing error by Weir et al. (2012).

**Colletotrichum radicis** F. Liu, L. Cai, Crous & Damm, sp. nov.

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**Etymology.** Referring to the root organ, from which it was isolated.

On *Anthriscus* stem. *Vegetative hyphae* hyaline to medium brown, smooth-walled, septate, branched. *Conidiomata* acervular, conidiophores and setae formed on a cushion of angular brown cells. *Setae* brown, smooth-walled, 0–3-septate, 77–192 μm long, base cylindrical to inflated, 5.5–6.5 μm diam, tip acute to obtuse. *Conidiophores* hyaline to brown, septate, branched. *Conidiogenous cells* hyaline to medium brown, cylindrical to clavate, 14–23 × 5.5–8.5 μm, opening 1.5–2 μm diam. *Conidia* hyaline, aseptate, smooth-walled, cylindrical to slightly curved, both ends rounded, 15.5–28 × 5.5–9.5 μm, av. ± SD = 22.6 ± 3.4 × 7.8 ± 0.7 μm, L/W ratio = 2.9.

On SNA. *Vegetative hyphae* hyaline to medium brown, smooth-walled, septate, branched. *Chlamydospores* not observed (but see below). *Conidiomata* acervular. *Setae* medium to dark brown, smooth-walled, 0–3-septate, 43–230 μm long, base cylindrical to inflated, 3.5–8.5 μm diam, tip acute to obtuse. *Conidiophores* brown, septate, branched. *Conidiogenous cells* medium brown, cylindrical to clavate, 11.5–24 × 5–9 μm, opening 1–2.5 μm diam. *Conidia* hyaline, aseptate, smooth-walled, cylindrical to slightly curved, 25.5–32.5 × 6.5–9.5 μm, av. ± SD = 28.2 ± 1.7 × 7.9 ± 0.6 μm, L/W ratio = 3.6. *Appressoria* not observed on the underside of the medium, but in old cultures appressoria-like structures that possibly function as chlamydospores were observed within the medium; these are small or in single dense clusters, light to medium brown, smooth-walled, globose, subglobose, elliptical to clavate in outline, with an entire or undulate margin, 4–8.5 μm diam.

Culture characteristics — Colonies on OA flat with entire margin, aerial mycelium lacking; colonial diam 64–71 mm in 7 d, > 90 mm in 10 d. Colonies on SNA flat with entire margin, aerial mycelium lacking, medium hyaline, buff around *Anthriscus* stem; colonial diam 64–75 mm in 7 d, > 90 mm in 10 d.

**Specimen examined.** COSTA RICA, La Selva, host plant unknown (isolated from a plant root), unknown collection date and collector (isolated by G. Weber in Mar. 1993) (holotype CBS H-21494, culture ex-type CBS 529.93).

**Notes** — *Colletotrichum radicis* is phylogenetically close to but clearly differentiated from *C. magnisporum* based on multi-locus and single gene phylogenetic analyses (Fig. 1, 2). Furthermore, *C. radicis* produces relatively short and narrow conidia (25.5–32.5 × 6.5–9.5 μm, av. = 28.2 × 7.9 μm) compared to those of *C. magnisporum* (28.5–40.5 × 8.5–11 μm, av. = 34.3 × 9.7 μm). In addition, many conidia of *C. radicis* are slightly curved, while those of *C. magnisporum* are straight.

The closest match in a blastn search with the ITS sequence of CBS 529.93 was FJ205460 (with 97 % identity, 18 bp differences) from isolate CBS 169.59 from *Oncidium excavatum* in the Netherlands, which is here referred to as *C. arxii* (Munaut et al. unpubl. data) and FN599826 (with 95 % identity, 23 bp differences; Rakotoniriana & Munaut unpubl. data) from isolate MUCL 41702 from *Orchis* in Singapore.

**Colletotrichum vietnamense** F. Liu, L. Cai, Crous & Damm, sp. nov.

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**Etymology.** Referring to the country where the fungus was collected.

On *Anthriscus* stem. *Vegetative hyphae* hyaline to medium brown, smooth-walled, septate, branched. *Conidiomata* acervular, conidiophores and setae formed on a cushion of angular...
brown cells. **Setae** medium to dark brown, smooth-walled to verruculose, 1–3-septate, 100–180 μm long, base cylindrical to inflated, 6–9.5 μm diam, tip subacute to rounded. **Conidiophores** hyaline to brown, septate, branched. **Conidiogenous cells** hyaline to medium brown, cylindrical, clavate to pyriform, 17–26.5 × 7–9.5 μm, opening 2–3.5 μm diam, collarette (few observed) 0.5 μm long. **Conidia** hyaline, aseptate, smooth-walled, cylindrical, occasionally slightly curved, both ends rounded, 19.5–40 × 8–10.5 μm, av. ± SD = 32.3 ± 4.9 × 9.5 ± 0.6 μm, L/W ratio = 3.4.

On SNA. **Vegetative hyphae** hyaline to medium brown, smooth-walled, septate, branched. **Conidiomata** acervular. **Setae** medium to dark brown, smooth-walled to verruculose, 1–7-septate, 118–176 μm long, base cylindrical to inflated, 7.5–9.5 μm diam, tip subacute. **Conidiophores** hyaline to brown, septate, branched. **Conidiogenous cells** hyaline to medium brown, cylindrical, clavate, to pyriform, 13–20.5 × 7.5–10 μm, opening 2–3 μm diam, collarette 0.5 μm long. **Conidia** hyaline, aseptate, smooth-walled, cylindrical, occasionally slightly curved, both ends curved.

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**Fig. 6 Colletotrichum radicis** (CBS 529.93). a, b. Acervuli; c, i. basal parts of setae; d, g, h. tips of setae; e. conidiogenous cells with conidia; f. conidiophores; j, k. appressoria-like structures; l, m. conidia (a, f–i, m: from *Anthriscus* stem; b–e, j–l: from SNA. — a, b: DM; c–m: DIC). — Scale bars: b = 100 μm (applies to a, b); m = 10 μm (applies to c–m).
rounded, 24–39 × 7.5–11.5 μm, av. ± SD = 31.2 ± 3.6 × 9.6 ± 0.7 μm, L/W ratio = 3.3. Appressoria (only few observed) pale brown, solitary, irregular outline with crenate or lobed margin, 9–17 × 5.5–12.5 μm, av. ± SD = 13.2 ± 2.7 × 9.1 ± 2.7 μm, L/W ratio = 1.2.

Culture characteristics — Colonies on OA flat with entire margin, rosy-buff pigmented, aerial mycelium white to grey, sparse; reverse olivaceous-grey; colonial diam 56–61 mm in 7 d, > 90 mm in 10 d. Colonies on SNA flat with entire margin, medium hyaline, buff around Anthriscus stem, aerial mycelium lacking; colonial diam 61–63 mm in 7 d, > 90 mm in 10 d.

Specimens examined. Vietnam, Lam Dong Province, Dalat, from anthracnose on leaf of Coffea sp., unknown collection date, P. Nguyen & E. Lijeroth (holotype CBS H-21512, culture ex-type CBS 125478 = LD16(L2)); Dak Lac Province, Buon Ma Thout, from anthracnose on leaf of Coffea sp., unknown collection date, P. Nguyen & E. Lijeroth, culture CBS 125477 = BMT25(L3).

Notes — Anthracnose of Coffea sp. can be caused by various Colletotrichum species, e.g., C. acutatum (Damm et al. 2012a), C. asianum (Prihastuti et al. 2009), C. coffeaeum (Noack 1901), C. coffeophilum (Spegazzini 1919), C. costaricense (Damm et al. 2012a), C. fructicola (Prihastuti et al. 2009), C. incarnatum (Zimmermann 1901), C. kahawae (Waller et al. 1993), C. queenslandicum (Weir et al. 2012), C. siamense (Prihastuti et al. 2009) and C. walleri (Damm et al. 2012a). The newly described species C. vietnamense is morphologically and phylogenetically different from these species. Colletotrichum asianum, C. fructicola, C. kahawae, C. queenslandicum and C. siamense, belong to the C. gloeosporioides complex, and C. acutatum, C. costaricense and C. walleri, belong to the C. acutatum complex, all of them have much smaller conidia (Shivas & Tan 2009, Damm et al. 2012a, Weir et al. 2012).

Colletotrichum coffeaeum was characterised by 1–2-septate setae; pyriform hyaline conidiophores, 18–20 × 4 μm; smooth,
oblong with rounded ends, often curved conidia, 12–18 × 4–5 μm (Noack 1901). Colletotrichum coffeaphilum produces aspetate setae, 25–50 × 4–6 μm; conidia ellipsoidal and hyaline, 1-guttulate, 13–15 × 6–8 μm (Spiegazzini 1919). Colletotrichum incarnatum has dark brown setae, flat tipped, base cylindrical or somewhat swollen, 85 × 4–5 μm; conidia oblong, 14–19 × 5 μm (Zimmermann 1901). In contrast, C. vietnamense differs from these three species in forming much larger conidia and longer setae.

Another species known to occur on Coffea sp. from Vietnam in this complex is C. gigasporum (CBS 125476 and CBS 125475), which can be distinguished from C. vietnamense by each of the eight genes used in this study, including ITS (Fig. 1).

The closest matches with the ITS sequence of CBS 125478 were FJ968584 (with 100 % identity), a sequence generated from the same isolate by Nguyen et al. (2010), and EF672327 (with 100 % identity) from the endophytic isolate PR61F2, also from Coffea arabica, but from coffee berries in Puerto Rico, a country in Central America (Vega et al. unpubl. data). Closest match with the TUB2 sequence was KC293665 (with 96 % identity, 20 bp differences) from isolate gnqczg15 from China (Huang et al. unpubl. data).

DISCUSSION

Many of the strains included in the present study were deposited in the CBS culture collection as C. crassipes (Speg.) Arx. However, C. crassipes is a species with uncertain taxonomic status. There is significant confusion regarding its morphology in the literature. Spiegazzini (1878) originally described this fungus as Gloeosporium crassipes from Vitis vinifera from Conegliano, Italy with conidia measuring 20–30 × 7–8 μm. Subsequently, von Arx (1957) combined Gloeosporium crassipes in Colletotrichum as C. crassipes along with 17 synonyms. The conidial size of C. crassipes was reported as 22–31 × 6–8 μm, broadly matching the original description; and the appressoria as irregular, usually lobed, measuring 8–12 μm (von Arx 1957). Sutton (1980) presented a different morphological concept of C. crassipes, which was characterised by conidia measuring 10–15 × 4.5–6.5 μm, long clavate or circular appressoria with crenate or deeply divided edges, 10.5–14 × 7–9.5 μm, and reduced another two names to synonymy with it. However, when Sutton summarised an accepted taxa list of Colletotrichum species, C. crassipes was characterised with conidia again with a different size (14–28 × 5–7 μm), and he suspected that this species may consist of a number of separate taxa (Sutton 1992). Moreover, several isolates identified as C. crassipes that have sequences lodged in GenBank actually belong to C. gloeosporioides s.l. (Weir et al. 2012). Recollecting and epitypification of this taxon is required to stabilise the phylogenetic position of C. crassipes.

Although morphological features are not stable and change under different growth conditions and with repeated subculturing, species of the C. gigasporum species complex form larger conidia than most of the other species in the genus Colletotrichum, which provides a valuable character for species complex level diagnosis. Conidia of two other species with large conidia, C. euphoriae and C. sansevieriae, differ in shape; they are slightly clavate with a round apex tapering to a truncate to slightly acute base (Nakamura et al. 2006, Crous et al. 2013). These two species do not belong to the C. gigasporum complex. While single gene data, especially ITS data, are usually not sufficient for species recognition in most of the Colletotrichum species complexes or groups (Cannon et al. 2012) and multi-locus phylogenies are therefore now routinely used as the primary basis on which to describe new Colletotrichum species (Damm et al. 2012a, b, Weir et al. 2012, Liu et al. 2013a, b), species of the C. gigasporum species complex can be easily distinguished from each other using the individual gene data included in this study (Fig. 1).

Colletotrichum gigasporum appears to have a wide host range and geographic distribution. Isolates treated in this paper and those deposited in GenBank originate mainly from Africa (East Africa, Madagascar), Central and South America (Brazil, Chile, Columbia, Ecuador, Guyana, Mexico, Panama), Asia (China, India, Japan, Korea, Thailand, Vietnam) and New Zealand (Fig. 1). Besides, this species is associated with various host plants as pathogens and endophytes, from air and stored grain, indicating that it is not host-specific and apparently has different life styles. This character is not unique to C. gigasporum, many other Colletotrichum species have been reported as both pathogens and endophytes, e.g. C. boninense, C. karstii and C. liniipes (Yang et al. 2011, Damm et al. 2012b, Tao et al. 2013). For instance, C. boninense causes diseases of Crinum asiaticum var. sinicum and Solanum lycopersicum, and is also an endophyte of Bletilla ochracea and Dacrycarpus dacro-dioiides (Damm et al. 2012b, Tao et al. 2013). The relationship between plant endophytic and pathogenic isolates of the same Colletotrichum species needs more research, as some endophytes may be latent pathogens (Lu et al. 2004).

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