Morphological and phylogenetic characterisation of novel Cytospora species associated with mangroves

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Abstract
Mangroves are relatively unexplored habitats and have been shown to harbour a number of novel species of fungi. In this study, samples of microfungi were collected from symptomatic branches, stem and leaves of the mangrove species Xylocarpus granatum, X. moluccensis and Lumnitzera racemosa and examined morphologically. The phylogeny recovered supports our morphological data to introduce three new species, Cytospora lumnitzericola, C. thailandica and C. xylocarpi. In addition, a combined multi-gene DNA sequence dataset (ITS, LSU, ACT and RPB2) was analysed to investigate phylogenetic relationships of isolates and help in a more reliable species identification.

Keywords
3 new species, Cytosporaceae, Lumnitzera racemosa, Mangroves, Phylogeny, Taxonomy, Xylocarpus granatum, Xylocarpus moluccensis
Introduction

Mangroves are forests established in tropical and subtropical backwaters, estuaries, deltas and lagoons. These forests play a major role in the ecology of coastal tropical/subtropical waters, as they serve as hatchery and nursery habitats for marine organisms and protect coastlines from catastrophic events such as storms and tidal surges (Hyde and Jones 1988, Fisher and Spalding 1993, Hyde and Lee 1995, Hyde et al. 1998). The greatest diversity of mangrove species occurs in the mangroves of Indonesia, Malaysia and Thailand (Alias and Jones 2009, Alias et al. 2010).

Reports of fungi associated with mangroves are relatively few and data on diseases of mangroves are uncommon (Cribb and Cribb 1955, Kohlmeyer and Kohlmeyer 1979, Hyde and Jones 1988). So far, a number of fungi collected from mangroves are either saprobes (e.g. Swe et al. 2008a, b, Devadatha et al. 2018, Li et al. 2018) or endophytes (e.g. Liu et al. 2012, Doilom et al. 2017). One early species documented from mangroves is that of Stevens (1920) who reported a species of Anthostomella that was found from a leaf spot in red mangroves (Rhizophora mangle) in Puerto Rico. Later, McMillan (1964) reported Cercospora which caused leaf spot on red mangroves in Florida and Kohlmeyer (1969) documented an undescribed Cytospora species on R. mangle in Hawaii. Cytospora rhizophorae has also been reported as a marine fungus from Rhizophora mangle in southwest Puerto Rico (Wier et al. 2000). Later, Shivas et al. (2009) reported a serious disease, caused by Pseudocercospora avicenniae, on leaves of Avicennia marina in Cape Tribulation, Queensland.

Cytospora was introduced by Ehrenberg (1818) and belongs to the family Cytosporaceae in Diaporthales (Wijayawardene et al. 2018). Cytospora species are phytopathogens or saprobes (Wehmeyer 1975, Barr 1978, Eriksson 2001, Castlebury et al. 2002, Wijayawardene et al. 2018). Cytospora has a worldwide distribution and is an important pathogenic genus, causing canker and dieback disease on branches of a wide range of plants (Adams et al. 2005, 2006, Hyde et al. 2017, Norphanphoun et al. 2017). Currently, there are 614 epithets for Cytospora (Index Fungorum 2018, 14 June 2018) with an estimated 110 species in Kirk et al. (2008). Recently, fourteen new species were introduced to this genus by Norphanphoun et al. (2017). In this study, we report on three novel species of Cytospora associated with mangroves in Thailand. Detailed descriptions and illustrations of all the species identified are provided in this paper.

Material and methods

Sample collection and examination of specimens

Samples collected were dead branches of Xylocarpus granatum K.D. Koenig, X. molucensis (Lam.) M. Roem. and leaf spots of Lumnitzera racemosa Willd. from Phetchaburi and Ranong provinces, Thailand in 2016. Specimens were returned to the laboratory in paper bags, examined and described following Norphanphoun et al.
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Morphological characters of ascomata and conidiomata were examined using a Motic SMZ 168 dissecting microscope. Hand sections were mounted in water and examined for morphological details. Micro-morphology was studied using a Nikon Ni compound microscope and photographed with a Canon EOS 600D digital camera fitted to the microscope. Photo-plates were made using Adobe Photoshop CS6 Extended version 13.0 × 64 (Adobe Systems, USA), while Tarosoft (R) Image Frame Work programme v. 0.9.7 was used for measurements.

Cultures were obtained by single spore isolation method outlined in Chomnunti et al. (2014). Single germinating spores were observed and photographed using a Nikon Ni compound microscope fitted with Canon EOS 600D digital camera. Geminated spores were transferred aseptically to 2% malt extract agar (MEA, malt extract agar powder 32 g in 1000 ml water) and incubated at room temperature (18–25 °C). A tissue isolation method was used for isolation of taxa from leaf spots of Lumnitzera racemosa. Leaves with leaf spots were cut into small pieces (0.5 × 0.5 cm²) using a sterilised blade and surface was sterilised using 70% ethanol for 1 minute, followed by three rinses with sterile distilled water, 1 minute in 3% sodium hypochlorite (NaOCl) and rinsed with sterile water for 1–2 minutes and dried by blotting on sterile filter paper. Four to five segments including the edge of the leaf spot were placed on water agar (WA) plates, supplemented with 100 mg/ml streptomycin. The dishes were incubated at 27 °C ± 2 °C for 7–10 days. Fungal colonies were transferred using single hyphal tips on to potato dextrose agar (PDA) plates throughout a 2-week period. Pure cultures were maintained for further studies on PDA (Bharathidasan and Panneerselvam 2011). The specimens/dried cultures and living cultures are deposited in the Herbarium Mae Fah Luang University (MFLU) and culture collection Mae Fah Luang University (MFLUCC), Chiang Rai, Thailand and duplicated in the International Collection of Micro-organisms from Plants (ICMP). Facesoffungi numbers were registered as in Jayasiri et al. (2015). New taxa are established based on recommendations as outlined by Jeewon and Hyde (2016).

DNA extraction, PCR amplification and sequencing

Genomic DNA was extracted from fresh fungal mycelia growing on MEA at room temperature (18–25 °C) for three weeks using a E.Z.N.A.TM Fungal DNA MiniKit (Omega Biotech, CA, USA) following the manufacturer's protocols. Polymerase chain reactions (PCR) were carried out using primer pairs of ITS1 (5’-TCCGTAGGTGAACCTGCGG-3’) and ITS4 (5’-TCCTCCGCTTATTGATATGC-3’) to amplify the ITS region (White et al. 1990), primer pairs of NL1 (5’-GCATATCAATAAGCGGAGGAAAAG-3’) and NL4 (5’-GGTCCGTGTTTCAAGACGG-3’) to amplify part of the large subunit rDNA (28S, LSU) (O’Donnell 1993), the partial ACT region was amplified using primers ACT512F (5’-ATGTGCAAGGCCGGTTTCGC-3’) and ACT783R (5’-TACCAGTCCCTTCTGGCCCAT-3’) (Carbone and Kohn 1999) and the partial RPB2 region was amplified using primers bRPB2-6F (5’-TGCGGGYGATGTTNGYCCYGC-3’) and bRPB2-7.1R (5’-CCCATRGCYTTGMCCATDGC-3’) (Matheny 2005).
The amplification reactions were carried out with the following protocol: 50 μl reaction volume containing 2 μl of DNA template, 2 μl of each forward and reverse primers, 25 μl of 2 × Bench Top™Taq Master Mix (mixture of Taq DNA Polymerase (recombinant): 0.05 units/μl, MgCl₂: 4 mM and dNTPs (dATP, dCTP, dGTP, dTTP): 0.4 mM) and 19 μl of double-distilled water (ddH₂O) (sterilised water) using the thermal cycle programme in Norphanphoun et al. (2017). Purification and sequencing of PCR products with the same primers mentioned above were carried out at Life Biotechnology Co., Shanghai, China.

Phylogenetic analysis

The sequences were assembled by GENEIOUS Pro v. 11.0.5 (Biomatters) and BLAST searches were made to retrieve the closest matches in GenBank and multiple alignment also included recently published sequences (Norphanphoun et al. 2017, Hyde et al. 2017, 2018). Combined analyses of ITS1, 5.8S, ITS2, LSU, RPB2 and ACT sequence data of 86 taxa were performed under different optimality criteria (MP, ML, BI). *Diaporthe eres* (AFTOL-ID 935) was used as the outgroup taxon. In order to obtain a better picture of the phylogenetic relationships amongst our strains and closely related strains, a separate ITS1+ITS2 phylogeny was inferred, because only ITS sequences were available for many strains in that group and because less ambiguously aligned (and excluded) positions are expected in a dataset with narrower taxonomic coverage. Nineteen strains were selected for this analysis based on preliminary analyses and results from the multigene phylogeny. All sequences were aligned separately using the MAFFT v.7.110 online programme (http://mafft.cbrc.jp/alignment/server/; Katoh and Standley 2013) and Gblocks v. 0.91b was used to exclude ambiguously aligned positions in the ITS and ACT alignments (Castresana 2000, Talavera and Castresana 2007). A partition homogeneity test (PHT) was performed with PAUP 4.0b10* (Swofford 2002) to determine whether the individual datasets were congruent and could be combined. The combined sequence alignments were obtained from MEGA7 version 7.0.14 (Kumar et al. 2015), missing data were coded as question marks (?) and further manual adjustments were made wherever necessary in BioEdit 7.2.3 (Hall 1999). The combined sequence alignment was converted to NEXUS file for maximum parsimony analysis using ClustalX v. 2 (Larkin et al. 2007). The NEXUS file was prepared for MrModeltest v. 2.2 (Nylander 2004) in PAUP v.4.0b10 (Swofford 2002).

Maximum Parsimony (MP) analysis was performed using PAUP (Phylogenetic Analysis Using Parsimony) v. 4.0b10* (Swofford 2002) with 1000 bootstrap replicates using a heuristic search with random stepwise addition and tree-bisection reconnection (TBR), as detailed by Jeewon et al. (2002) and Cai et al. (2005). Maxtrees was set to 1000, branches of zero length were collapsed. The following descriptive tree statistics were calculated: parsimony tree length [TL], consistency index [CI], retention index [RI], rescaled consistency index [RC] and homoplasy index [HI].
Table 1. GenBank accession numbers of the sequences used in phylogenetic analyses.

| No | Taxon                   | Strain | Host          | Origin             | ITS     | LSU     | RPB2    | ACT     | References                     |
|----|-------------------------|--------|---------------|--------------------|---------|---------|---------|---------|--------------------------------|
| 1  | *Cytospora abyssinica*  | CMW 10181<sup>T</sup> | *Eucalyptus globulus* | Wondo Genet, Ethiopia | AY347353 | –       | –       | –       | Adams et al. (2005)            |
| 2  | *C. acaciae*            | CBS 468.69 | *Ceratonia siliqua* | Spain, Mallorca    | DQ243804 | –       | –       | –       | Adams et al. (2006)            |
| 3  | *C. ampulliformis*      | MFLUCC 16-0583<sup>f</sup> | *Sorbus intermedia* | Russia             | KY417726 | KY417760 | KY417794 | KY417692 | Norphanphoun et al. (2017)     |
| 4  | *C. atroirrhata*        | HMBF156 | *Eucalyptus pauciflora* | Australia          | –       | –       | –       | –       | –                              |
| 5  | *C. austromontana*      | CMW 6735<sup>T</sup> | *Eucalyptus pauciflora* | China              | AY347361 | –       | –       | –       | Adams et al. (2005)            |
| 6  | *C. berberidis*         | CFCC 89927<sup>f</sup> | *Berberis dasystachya* | China              | KR045620 | KR045702 | KU710948 | KU710990 | Liu et al. (2015)              |
| 7  | *C. berkeleyi*          | StanfordT3<sup>T</sup> | *Eucalyptus globulus* | California, USA    | AY347350 | –       | –       | –       | Adams et al. (2005)            |
| 8  | *C. brevisspora*        | CBS 116829 | *Eucalyptus grandis* | Venezuela          | AF192321 | –       | –       | –       | Adams et al. (2005)            |
| 9  | *C. carbonacea*         | CFCC 89947 | *Ulmus pumila* | Qinghai, China     | K045622  | KP310812 | KU710950 | KP310842 | Yang et al. (2015)             |
| 10 | *C. centrevillosa*      | MFLUCC 16-1206<sup>f</sup> | *Sorbus domestica* | Italy              | MF190122 | MF190068 | MF377600 | –       | Senanayake et al. (2017)       |
| 11 | *C. centosperma*        | MFLUCC 16-0625 | *Acer platanoides* | Russia             | KY563246 | KY563248 | KY563244 | KY563242 | Tibpromma et al. (2017)        |
| 12 | *C. chrysosperma*       | HMBF151 | *Eucalyptus globulus* | Chile              | AY347377 | –       | –       | –       | –                              |
| 13 | *C. cinereostroma*      | CMW 5700<sup>T</sup> | *Eucalyptus globulus* | Russia             | KX430142 | KX430143 | KX430144 | –       | Norphanphoun et al. (2017)     |
| 14 | *C. cotini*             | MFLUCC 14-1050<sup>f</sup> | *Cotinus coggyria* | Russia             | KY417728 | –       | –       | –       | –                              |
| 15 | *C. curvata*            | MFLUCC 15-0865<sup>T</sup> | *Salix alba* | Russia             | KY417728 | –       | –       | –       | Norphanphoun et al. (2017)     |
| 16 | *C. cypri*              | CBS 201.42<sup>T</sup> | *Syringa sp.* | Switzerland        | DQ243801 | –       | –       | –       | Adams et al. (2006)            |
| 17 | *C. diatrypelloidea*    | CMW 8549<sup>T</sup> | *Eucalyptus globulus* | Orbost, Australia  | AY347368 | –       | –       | –       | Adams et al. (2005)            |
| 18 | *C. disciformis*        | CMW6509 | *Eucalyptus globulus* | Russia             | AY347374 | –       | –       | –       | Adams et al. (2005)            |
| 19 | *C. donetzica*          | MFLUCC 16-0574<sup>f</sup> | *Rosa sp.* | Russia             | KY417731 | KY417765 | KY417799 | KY417697 | Norphanphoun et al. (2017)     |
| 20 | *C. elaeagni*           | CFCC 89632 | *Elaeagnus angustifolia* | Ningxia, China     | K045626  | K045706  | KU710955 | KU710995 | Fan et al. (2015b)             |
| No | Taxon          | Strain  | Host                | Origin            | GenBank accession numbers | ITS          | LSU          | RPB2          | ACT          | References                              |
|----|----------------|---------|---------------------|-------------------|---------------------------|-------------|-------------|--------------|-------------|-----------------------------------------|
| 21 | C. erumpens    | MFLUCC 16-0580 | Salix × fragilis | Russia            | KY417733                  | KY417767    | KY417801    |               |             | Norphanphoun et al. (2017)              |
| 22 | C. eriobotryae | IMI136523 | Eriobotrya japonica | India             | 4D806                      | AL347327    |             | AL347340    | AL347305    | Adams et al. (2005)                     |
| 23 | C. eucalypti   | MFLUCC 17-0534 | Sequoiadendron grados | California, USA   | AY473357                  | AY473600    |             | AY473518    |             | Norphanphoun et al. (2017)              |
| 24 | C. eucalyptina | LSEQ113.81 | Eucalyptus grandis | Cali, Columbia    | AY473275                  |             |             |             |             | Norphanphoun et al. (2017)              |
| 25 | C. fabianae    | CXY1322  | Eucalyptus         | Cali, Columbia    | AY473275                  |             |             |             |             | Norphanphoun et al. (2017)              |
| 26 | C. friesii     | MFLUCC 16-0624 | Picea abies  | Norway            | AY473275                  |             |             |             |             | Norphanphoun et al. (2017)              |
| 27 | C. gelida      | MFLUCC 16-0634 | Cotinus coggyria | Russia            | KY417734                  | KY417768    | KY417802    | KY417700    |             | Norphanphoun et al. (2017)              |
| 28 | C. germanica   | MFLUCC 16-0634 | Elaeagnus angustifolii | China           | KY417734                  | KY417768    | KY417802    | KY417700    |             | Norphanphoun et al. (2017)              |
| 29 | C. gigesu      | CFCC 896434 | Salix psammophylla | China            | KY417734                  | KY417768    | KY417802    | KY417700    |             | Norphanphoun et al. (2017)              |
| 30 | C. gigaspora   | CFCC 89636 | Tamarix parviflora | China            | KY417734                  | KY417768    | KY417802    | KY417700    |             | Norphanphoun et al. (2017)              |
| 31 | C. gigalocus   | MFLUCC 17-0882 | Juniperus communis | Japan             | MF190125                  | MF190072    |             |             |             | Norphanphoun et al. (2017)              |
| 32 | C. gigaspora   | MFLUCC 17-0882 | Juniperus communis | Japan             | MF190125                  | MF190072    |             |             |             | Norphanphoun et al. (2017)              |
| 33 | C. gigaspora   | MFLUCC 17-0882 | Juniperus communis | Japan             | MF190125                  | MF190072    |             |             |             | Norphanphoun et al. (2017)              |
| 34 | C. gigaspora   | MFLUCC 17-0882 | Juniperus communis | Japan             | MF190125                  | MF190072    |             |             |             | Norphanphoun et al. (2017)              |
| 35 | C. gigaspora   | MFLUCC 17-0882 | Juniperus communis | Japan             | MF190125                  | MF190072    |             |             |             | Norphanphoun et al. (2017)              |
| 36 | C. gigaspora   | MFLUCC 17-0882 | Juniperus communis | Japan             | MF190125                  | MF190072    |             |             |             | Norphanphoun et al. (2017)              |
| 37 | C. gigaspora   | MFLUCC 17-0882 | Juniperus communis | Japan             | MF190125                  | MF190072    |             |             |             | Norphanphoun et al. (2017)              |
| 38 | C. gigaspora   | MFLUCC 17-0882 | Juniperus communis | Japan             | MF190125                  | MF190072    |             |             |             | Norphanphoun et al. (2017)              |
| 39 | C. gigaspora   | MFLUCC 17-0882 | Juniperus communis | Japan             | MF190125                  | MF190072    |             |             |             | Norphanphoun et al. (2017)              |
| 40 | C. gigaspora   | MFLUCC 17-0882 | Juniperus communis | Japan             | MF190125                  | MF190072    |             |             |             | Norphanphoun et al. (2017)              |
| 41 | C. gigaspora   | MFLUCC 17-0882 | Juniperus communis | Japan             | MF190125                  | MF190072    |             |             |             | Norphanphoun et al. (2017)              |
| 42 | C. gigaspora   | MFLUCC 17-0882 | Juniperus communis | Japan             | MF190125                  | MF190072    |             |             |             | Norphanphoun et al. (2017)              |
| No | Taxon       | Strain       | Host                   | Origin     | GenBank accession numbers | References                        |
|----|-------------|--------------|------------------------|------------|---------------------------|-----------------------------------|
| 43 | C. multicolor | CBS 105,889 | Quercus decurrens, Quercus suber, Quercus robur | Spain      | DQ243467-68                | Adams et al. (2006)               |
| 44 | C. myrtagena | HiloTib1     | Tibouchina urvilleana   | Hawaii     | FJ765666                   | Adams et al. (2005)               |
| 45 | C. nitschkii | MFLUCC15-0857| Eucalyptus globulus    | Ethiopia   | JN111959                   | Norphanphoun et al. (2017)        |
| 46 | C. nivea    | MFLUCC 15-0860| Salix arctostaphyloides | Russia     | KY417777                   | Adams et al. (2005)               |
| 47 | C. palmaris | CXY12802     | Cotinus coggygria      | China      | JN111959                   | Norphanphoun et al. (2017)        |
| 48 | C. paratranslucens | T28.1 | Prunus persicae    | China      | JN111959                   | Zhang et al. (2014)               |
| 49 | C. paratranslucens | MFLUCC 16-0506 | Malus domestica | Russia     | KY417740                   | Adams et al. (2005)               |
| 50 | C. paratranslucens | MFLUCC 16-0506 | Malus domestica | Russia     | KY417740                   | Adams et al. (2005)               |
| 51 | C. paratranslucens | MFLUCC 16-0506 | Malus domestica | Russia     | KY417740                   | Adams et al. (2005)               |
| 52 | C. paratranslucens | MFLUCC 16-0506 | Malus domestica | Russia     | KY417740                   | Adams et al. (2005)               |
| 53 | C. paratranslucens | MFLUCC 16-0506 | Malus domestica | Russia     | KY417740                   | Adams et al. (2005)               |
| 54 | C. paratranslucens | MFLUCC 16-0506 | Malus domestica | Russia     | KY417740                   | Adams et al. (2005)               |
| 55 | C. paratranslucens | MFLUCC 16-0506 | Malus domestica | Russia     | KY417740                   | Adams et al. (2005)               |
| 56 | C. paratranslucens | MFLUCC 16-0506 | Malus domestica | Russia     | KY417740                   | Adams et al. (2005)               |
| 57 | C. paratranslucens | MFLUCC 16-0506 | Malus domestica | Russia     | KY417740                   | Adams et al. (2005)               |
| 58 | C. paratranslucens | MFLUCC 16-0506 | Malus domestica | Russia     | KY417740                   | Adams et al. (2005)               |
| 59 | C. paratranslucens | MFLUCC 16-0506 | Malus domestica | Russia     | KY417740                   | Adams et al. (2005)               |
| 60 | C. rosii     | MFLUCC 16-0506 | Malus domestica | Russia     | KY417740                   | Adams et al. (2005)               |
| 61 | C. rosea     | MFLUCC 16-0506 | Malus domestica | Russia     | KY417740                   | Adams et al. (2005)               |
| 62 | C. rosae     | MFLUCC 16-0506 | Malus domestica | Russia     | KY417740                   | Adams et al. (2005)               |
| 63 | C. rosae     | MFLUCC 16-0506 | Malus domestica | Russia     | KY417740                   | Adams et al. (2005)               |
| No | Taxon          | Strain* | Host              | Origin        | GenBank accession numbers | References                      |
|----|---------------|---------|-------------------|---------------|---------------------------|---------------------------------|
| 64 | C. rostrata   | CFCC 89909<sup>T</sup> | *Salix cupularis* | Gansu, China | KR045643, KR045722, KU710974, KU711009 | Unpublished                     |
| 65 | C. rusnovii   | MFLUCC 15-0854<sup>T</sup> | *Salix babylonica* | Russia        | KY417744, KY417778, KY417812, KY417710 | Norphanphoun et al. (2017)      |
| 66 | C. secularis  | HMBF281 |                   |               | KF225615, KF225629, –, KF498678 | Fan et al. (2015a)              |
| 67 | C. salicacearum | MFLUCC 16-0509<sup>T</sup> | *Salix alba*     | Russia        | KY417746, KY417780, KY417814, KY417712 | Norphanphoun et al. (2017)      |
| 68 | C. saliccola  | MFLUCC 14-1052<sup>T</sup> | *Salix alba*     | Russia        | KU982636, KU982635, –, KU982637 | Li et al. (2016)                |
| 69 | C. salicina   | MFLUCC 15-0862<sup>T</sup> | *Salix alba*     | Russia        | KY417750, KY417784, KY417818, KY417716 | Norphanphoun et al. (2017)      |
| 70 | C. schulzeri  | CFCC 50040 | *Malus domestica* | Ningxia, China | KR045649, KR045728, KU710980, KU711013 | Unpublished                     |
| 71 | C. sibiraeae  | CFCC 50045<sup>T</sup> | *Siberaea angustata* | Gansu, China | KR045651, KR045730, KU710982, KU711015 | Liu et al. (2015)               |
| 72 | C. sorbi      | MFLUCC 16-0631<sup>T</sup> | *Sorbus aucuparia* | Russia        | KY417752, KY417786, KY417820, KY417718 | Norphanphoun et al. (2017)      |
| 73 | C. sorbicola  | MFLUCC 16-0584<sup>T</sup> | *Acer pseudoplatanus* | Russia        | KY417755, KY417789, KY417823, KY417721 | Norphanphoun et al. (2017)      |
| 74 | C. sordida    | HMBF159 |                   |               | KF225613, KF225627, –, KF498676 | Fan et al. (2015a)              |
| 75 | C. sophorae   | CFCC 50047 | *Styphnobium japonicum* | Shanxi, China | KR045653, KR045732, KU710984, KU711017 | Fan et al. (2014)               |
| 76 | C. sophoricola | CFCC 89596 | *Styphnobium japonicum* | Gansu, China | KR045656, KR045735, KU710987, KU711020 | Unpublished                     |
| 77 | C. tanaitica  | MFLUCC 14-1057<sup>T</sup> | *Betula pubescens* | Russia        | KT459411, KT459412, –, KT459413 | Ariyawansa et al. (2015)        |
| 78 | C. thailandica | MFLUCC 17-0262 | *Xylocarpus moluccensis* | Ranong, Thailand | MG975776, MH253463, MH253455, MH253459 | In this study                  |
| 79 | C. thailandica | MFLUCC 17-0263 | *Xylocarpus moluccensis* | Ranong, Thailand | MG975777, MH253464, MH253456, MH253460 | In this study                  |
| 80 | C. tibouchinae | CPC 26333<sup>T</sup> | *Tibouchina semidecandra* | La Reunion, France | KX228284, KX228335, –, – | Unpublished                     |
| 81 | C. translucens | 35 |                   |               | EF447403, –, –, – | Fotouhifar et al. (2010)        |
| 82 | C. ulmi       | MFLUCC 15-0863<sup>T</sup> | *Ulmus minor*    | Russia        | KY417759, KY417793, KY417827, KY417725 | Norphanphoun et al. (2017)      |
| No | Taxon                  | Strain*        | Host            | Origin              | GenBank accession numbers | References                  |
|----|-----------------------|----------------|-----------------|---------------------|---------------------------|-----------------------------|
|    |                       |                |                 |                     |                           |                             |
| 83 | *C. valsoidea*        | CMW 4309      | *Eucalyptus grandis* | Sibisa, North Sumatra | AF192312, –, –, –       | Adams et al. (2005)         |
| 84 | *C. variostromatica*  | CMW 6766      | *Eucalyptus globulus* | Australia           | AY347366, –, –, –      | Adams et al. (2005)         |
| 85 | *C. vinacea*          | CBS 141585    | *Vitis* sp.     | New Hampshire, USA  | KX256256, –, –, –     | Lawrence et al. (2017)      |
| 86 | *C. xylocarpi*        | MFLUCC 17-0251| *Xylocarpus granatum* | Ranong, Thailand   | MG975775, MH253462, MH253454, MH253458 | In this study |
| 87 | *Diaporthe eros*      | AFTOL-ID 935  |                  |                     | DQ491514, –, DQ470919, – | Spatafora et al. (2006)     |
| 88 | *C. "rhizophorae"*    | A761           | *Morinda officinalis* | China               | KU529867, –, –, –     | Unpublished                 |
| 89 | *C. "rhizophorae"*    | HAB16R13      | *Cinnamomum porrectum* | Malaysia            | HQ336045, –, –, –    | Harun et al. (2011)         |
| 90 | *C. "rhizophorae"*    | M225           | *Rhizophora mucronata* | Philippines         | KR056292, –, –, –    | Unpublished                 |
| 91 | *C. "rhizophorae"*    | MUCC302        | *Eucalyptus grandis* | Australia           | EU301057, –, –, –    | Unpublished                 |

* AFTOL-ID Assembling the Fungal Tree of Life; CBS CBS-KNAW Fungal Biodiversity Centre, Utrecht, The Netherlands; CFCC China Forestry Culture Collection Center; IMI International Mycological Institute, CABI-Bioscience, Egham, Bakeham Lane, UK; CPC Culture collection of Pedro Crous, housed at CBS; MFLU Mae Fah Luang University Herbarium Collection; MFLUCC Mae Fah Luang University Culture Collection, Chiang Rai, Thailand; Ex-type and ex-epitype cultures.
For both Maximum Likelihood and Bayesian analyses, a partitioned analysis was performed with the following six partitions: ITS1+ITS2, 5.8S, LSU, ACT-exons, ACT-introns and RPB2. Maximum-likelihood (ML) analysis was performed with RAxML (Stamatakis 2006) implemented in the CIPRES Science Gateway web server (RAxML-HPC2 on XSEDE; Miller et al. 2010), 25 categories, 1000 rapid bootstrap replicates were run with the GTRGAMMA model of nucleotide evolution. Maximum likelihood bootstrap values (MLBS) equal or greater than 50% are given above each node.

Bayesian Inference (BI) analysis was performed using the Markov Chain Monte Carlo (MCMC) method with MrBayes 3.2.2 (Ronquist et al. 2012). The best-fit nucleotide substitution model for each dataset was separately determined using MrModeltest version 2.2 (Nylander 2004). GTR+I+G was selected as the best-fit model for the ITS1+ITS2, LSU, ACT (ACT-exons and ACT-introns) and RPB2 datasets and K80 for 5.8S. The MCMC analyses, with four chains starting from random tree topology, were run for 5,000,000 or 10,000,000 generations for the combined dataset or the ITS1+ITS2 dataset. Trees were sampled every 100 generations. Tracer v. 1.5.0 was used to check the effective sampling sizes (ESS) that should be above 200, the stable likelihood plateaus and burn-in value (Rambaut et al. 2013). The first 5000 samples were excluded as burn-in.

The phylogram was visualised in FigTree v1.4.0 (http://tree.bio.ed.ac.uk/software/figtree/; Rambaut 2014) and edited in Adobe Illustrator CC and Adobe Photoshop CS6 Extended version 13.1.2 x 64. Newly generated sequences in this study are deposited in GenBank. The finalised alignment and tree were deposited in TreeBASE, submission ID: 22942 (combined sequence alignment) (Reviewer access URL: http://purl.org/phylo/treebase/phylows/study/TB2:S22942;x-access-code=f9115cf637b0e4171aab1c980eb15830&format=html) and (Reviewer access URL: http://purl.org/phylo/treebase/phylows/study/TB2:S22943;x-access-code=92a782825ac069b3fd761aff21fa2bf4&format=html) 22943 (ITS sequence alignment) (http://www.treebase.org).

**Results**

**Phylogenetic analysis of combined ITS, LSU, ACT and RPB2 sequences**

The combined alignment of ITS, LSU, ACT and RPB2 sequences comprised 86 taxa, including our strains, with Diaporthe eres (CBS 183.5) as the outgroup taxon. The total length of the dataset was 2037 characters including alignment gaps (1–199, 200–357, 358–518, 519–1056, 1057–1296 and 1297–2037 corresponding to ITS1, 5.8S, ITS2, LSU, ACT and RPB2, respectively). The combined dataset contained 1426 constant, 144 parsimony uninformative and 467 parsimony informative characters. The result from the partition homogeneity test (PHT) was not significant (level 95%), indicating that the individual datasets were congruent and could be combined. The combined dataset was analysed using MP, ML and Bayesian analyses. The trees generated under different optimality criteria were essentially similar in topology and did not differ sig-
Figure 1. Phylogenogram generated from maximum parsimony analyses based on analysis of combined ITS, LSU, ACT and RPB2 sequence data. The tree is rooted to *Diaporthe eres* (AFTOL-ID 935). Maximum parsimony and maximum likelihood bootstrap values ≥50%, Bayesian posterior probabilities ≥0.90 (MPBS/MLBS/PP) are given at the nodes. The species obtained in this study are in blue font. Ex-type taxa from other studies are in black bold.
The descriptive statistics of the phylogram generated from MP analysis based on the combined dataset of ITS, LSU, ACT and RPB2 (Fig. 1) were TL = 2418, CI = 0.375, RI = 0.650, RC = 0.244, HI = 0.625. The best scoring likelihood tree selected with a final value for the combined dataset = -14466.797686. The aligned sequence matrix of the ITS1+ITS2 dataset comprising 19 taxa had 279 constant, 23 parsimony uninformative and 57 parsimony informative characters. The descriptive statistics of the most parsimonious tree (Fig. 2) were TL = 2418, CI = 0.375, RI = 0.650, RC = 0.244, HI = 0.625. The best scoring likelihood tree obtained for the ITS1+ITS2 dataset had a log-likelihood of= -1276.782916.

**Taxonomy**

*Cytospora lumnitzericola* Norphanphoun, T.C. Wen & K.D. Hyde, sp. nov.
Index Fungorum number: IF554778; Facesoffungi number: FoF 04603

**Etymology.** Refers to the host where the fungus was isolated.

**Holotype.** MFLU 18-1227

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**Figure 2.** Maximum parsimony phylogenetic tree inferred from ITS1 and ITS2 sequence data. Maximum parsimony and maximum likelihood bootstrap values ≥50%, Bayesian posterior probabilities ≥0.90 (MPBS/MLBS/BIPP) are given at the nodes. The species obtained in this study are in blue font. Ex-type taxa from other studies are in black bold.
Isolated from leaf spot of *Lumnitzera racemosa*. **Culture characteristic:** Colonies on MEA reaching 5–6 cm diameter after 2 days at room temperature, colonies circular to irregular, medium dense, flat or effuse, slightly raised, with edge fimbriate, fluffy to fairly fluffy, white to grey from above, light yellow to green from below; not producing pigments in agar. **Asexual morph:** *Conidiogenous cells* (8–)8.5–14 × 0.6–1.4(–1.6) μm ($\bar{x} = 8.4 \times 1.4, n = 15$), blastic, enteroblastic, flask-shaped, phialidic, hyaline and smooth-walled. *Conidia* (3.7–)4–4.5 × 1–1.3(–1.5) μm ($\bar{x} = 4 \times 1.2$ μm, $n = 30$), unicellular, subcylindrical, hyaline, smooth-walled.

**Material examined.** THAILAND, Phetchaburi Province, the Sirindhorn International Environmental Park, on leaf spot of *Lumnitzera racemosa*, 30 November 2016, Norphanhoun Chada NNS23-2a (MFLU 18-1227 dried culture, holotype; PDD, isotype); ex-type-living culture, MFLUCC 17-0508, ICMP.
Notes. Based on the multigene phylogeny, *Cytospora lumnitzericola* is closely related to *Cytospora thailandica* (Fig. 1). Although conidial sizes of both species are similar, they have significant differences in nucleotides: ITS (26 nt), ACT (22 nt), and RPB2 (53 nt) (Table 5). The phylogeny derived from the ITS regions depicts *C. lumnitzericola* as an independent lineage close to *C. brevispora* CBS 116829 and *C. eucalyptina* CMW5882 (Fig. 2). In future, more collections are needed to confirm whether *C. lumnitzericola* can exist as a saprobe or endophyte as well as performing tests to confirm its pathogenicity.

*Cytospora thailandica* Norphanphoun, T.C. Wen & K.D. Hyde, sp. nov.  
Index Fungorum number: IF554779; Facesoffungi number: FoF 04605  
Figure 4  

Etymology. refers to the country where the fungus was collected.  
Holotype. MFLU 17-0709  

Associated with twigs and branches of *Xylocarpus moluccensis*. Sexual morph: Stromata immersed in bark. *Ascostromata* 400–1000 × 70–250 μm diameter, semi-immersed in host tissue, scattered, erumpent, uni- or multi-loculate, with ostiolar neck. *Ostiole* 70–150 μm diameter, numerous, dark brown to black, at the same level as the disc, occasionally area below disc a lighter entostroma. *Peridium* comprising several layers of cell of *textura angularis*, with innermost layer thick, brown, outer layer dark brown. *Hamathecium* comprising long cylindrical, cellular, anastomosed paraphyses. *Asci* (21–)23–25 × 4.1–4.7(–5) μm (x̄ = 22 × 4.3 μm, n = 15), 6–8-spored, unitunicate, clavate to elongate obovoid, with a J-, refractive apical ring. *Ascospores* (5.6–)6–6.8 × 1.3–1.5(–2) μm (x̄ = 6.6 × 1.5 μm, n = 20), biseriate, elongate-allantoid, unicellular, hyaline, smooth-walled. Asexual morph: *Conidiomata* 400–1200 × 180–380 μm diameter, semi-immersed in host tissue, solitary, erumpent, scattered, discoid, circular to ovoid, with multi-loculate, pycnidial, embedded in stromatic tissue, with ostiole. *Ostioles* 230–300 μm long, with an ostiolar neck. *Peridium* comprising few layers of cells of *textura angularis*, with innermost layer thin, pale brown, outer layer brown to dark brown. *Conidiophores* unbranched or occasionally branched at the bases, formed from the innermost layer of pycnidial wall, with conidiogenous cells. *Conidiogenous cells* (3.3–)6–9.1 × 1–1.3(–1.7) μm (x̄ = 6 × 1.3 μm, n = 15), blastic, enteroblastic, flask-shaped, phialidic, hyaline and smooth-walled. *Conidia* (3.3–)3.8–4 × 1–1.3(–1.5) μm (x̄ = 3.8 × 1.3 μm, n = 30), unicellular, subcylindrical, hyaline, smooth-walled.  

Material examined. THAILAND, Ranong Province, Ngao Mangrove Forest, on branches of *Xylocarpus moluccensis*, 6 December 2016, Norphanphoun Chada NG02a (MFLU 17-0709, holotype; PDD, isotype); ex-type-living cultures, MFLUCC 17-0262, MFLUCC 17-0263, ICMP.  

Notes. *Cytospora thailandica* was collected from branches of *Xylocarpus moluccensis*. The new species resembles some other *Cytospora* species, but is characterised by uni- or multi-loculate ascomata/conidiomata with unicellular, subcylindrical and hyaline spores in both morphs. *Cytospora* species associated with *Xylocarpus granatum* is also
Figure 4. Cytospora thailandica (MFLU 17-0709, holotype). a Xylocarpus moluccensis b Branch of Xylocarpus moluccensis c Ascostromata on host substrate d, e Surface of ascomata f Transverse sections through ascostroma to show distribution of locules g–h Longitudinal sections through ascostroma to show distribution of locules i Peridium j Ostiolar neck ka–kd n Asci l, m Apical ring oa–of Ascospores p Surface of conidioma q Transverse sections through conidioma to show distribution of locules r, s Longitudinal sections through conidioma to show distribution of locules t Peridium u Ostiolar neck va–vc w Conidiogenous cells with attached conidia x, y Conidia za, zb Colonies on MEA (za-from above, zb-from below). Scale bars: d = 1000 μm, e–g = 400 μm, h, j, p–s = 200 μm, i, u = 100 μm, ka–kd, n = 10 μm, l, m = 2 μm, oa–of, va–vc, w = 5 μm, t = 50 μm, x, y = 4 μm.
reported in this study as *C. xylocarpi* (MFLUCC 17-0251, Fig. 5). *Cytospora xylocarpi* is similar to *C. thailandica* in its conidiomata being multi-loculate and in the length of conidia in the asexual morph (*C. xylocarpi*: conidia 3 × 1.1 μm versus 3.8 × 1.3 μm in *C. thailandica*). However, *C. thailandica* differs from *C. xylocarpi* in having shorter ostiolar necks and larger asci and ascospores (Table 2). Phylogenetic analysis of our combined gene also reveals *C. thailandica* is closely related to *C. lumnitzericola* (Fig. 1), but there are nucleotide differences as mentioned in notes of *C. lumnitzericola*. The individual ITS1+ITS2 phylogenetic tree also indicates that *C. thailandica* is distinct with good support (Fig. 2).

**Cytospora xylocarpi** Norphanphoun, T.C. Wen & K.D. Hyde, sp. nov.

Index Fungorum number: IF554810; Facesoffungi number: FoF 04604

Figure 5

**Etymology.** refers to the host genus that fungus was collected.

**Holotype.** MFLU 17-0708

Associated with *Xylocarpus granatum* branches. **Sexual morph:** Stromata immersed in bark. *Ascstromata* 230–600 × 90–250 μm diameter, semi-immersed in host tissue, scattered, erumpent, multi-loculate, with ostiolar neck. *Ostiole* 160–200 μm diameter, numerous, dark brown to black, at the same level as the disc, occasionally area surrounded with white hyphae. *Peridium* comprising several layers of cells of *textura angularis*, with innermost layer thick, pale brown, outer layer dark brown to black. *Hamathecium* comprising long cylindrical, cellular, anastomosed paraphyses. *Asci* (22–)24–28.8 × 3.6–4.8(–5.1) μm (\(\bar{x} = 26 \times 4 \) μm, n = 15), 6–8-spored, unitunicate, clavate to elongate obovoid, with a refractive, J-, apical ring. *Ascospores* (5.5–)6–6.5 × 1.7–1.8(–2) μm (\(\bar{x} = 5.7 \times 1.8 \) μm, n = 20), biseriate, elongate-allantoid, unicellular hyaline, smooth-walled. **Asexual morph:** *Conidiomata* 700–1200 × 400–480 μm diameter, semi-immersed in host tissue, solitary, erumpent, scattered, multi-loculate, with ostiole. *Ostioles* 200–250 μm long, with 1–2 ostiolar necks. *Peridium* comprising several layers of cells of *textura angularis*, with innermost layer brown, outer layer dark brown to black. *Conidiophores* unbranched or occasionally branched at the bases, formed from the innermost layer of pycnidial wall, with conidiogenous cells. *Conidiogenous cells* (6.3–)7.9–10 × 0.9–1.4(–1.6) μm (\(\bar{x} = 8.5 \times 1.4 \) μm, n = 15), blastic, enteroblastic, flask-shaped, phialidic, hyaline and smooth-walled. *Conidia* (2.4–)3–3.1 × 0.8–1(–1.2) μm (\(\bar{x} = 3 \times 1 \) μm, n = 30), unicellular, subcylindrical, hyaline, smooth-walled.

**Material examined.** THAILAND, Ranong Province, Ngao Mangrove Forest, on branches of *Xylocarpus granatum*, 6 December 2016, Norphanphoun Chada NG09b (MFLU 17-0708, holotype; PDD); ex-type-living cultures, MFLUCC 17-0251, ICMP.

**Notes.** The asexual morph of *C. xylocarpi*, studied here, is most similar to *C. rhizophorae* from dead roots of *Rhizophora mangle* L. in Guatemala, in having multi-loculate conidiomata and allantoid, slightly curved, hyaline and 3–6 × 1.1–1.5 μm
Morphological and phylogenetic characterisation of novel *Cytospora* species...

**Figure 5.** *Cytospora xylocarpi* (MFLU 17-0708, holotype). **a** *Xylocarpus granatum* **b** Branch of *Xylocarpus granatum* **c** Ascostromata on host substrate **d** Surface of ascomata **e** Transverse sections through ascostroma to show distribution of locules **f, g** Longitudinal sections through ascostroma to show distribution of locules **h** Peridium **i–l, n** Asci **m, o** Ascospores **p** Germinating spore **q, r** Colonies on MEA (q-from above, r-below) **s** Transverse sections through conidioma to show distribution of locules **t** Longitudinal sections through conidioma to show distribution of locules **u, v** Conidiogenous cells with attached conidia **w** Mature conidia. Scale bars: **c** = 2000 μm, **d–f** = 500 μm, **g** = 200 μm, **h** = 20 μm, **i, p** = 10 μm, **j–o, u–w** = 5 μm, **s, t** = 400 μm.
Table 2. Synopsis of species of *Cytospora* discussed in the paper.

| Taxon                  | Sexual morph | Asexual morph | References |
|------------------------|--------------|---------------|------------|
|                        | Ascostoma    | Ostiolar neck | Ascospores | Conidiomata | Ostiolar neck | Conidiogenous cell | Conidia |            |
| *C. lumnitzericola*    | –            | –             | –          | –           | –             | 8.4 x 1.4          | 4 x 1.2  | In this study |
| *C. rhizophorae*       | –            | –             | –          | –           | 370–500 x 100–310 | 30 x 10–25  | 13–20 x 1–1.8 | 3–6 x 1.1–1.5 | Kohlm. and Kohlm. (1971) |
| *C. thailandica*       | 400–1000 x 70–250 | 70–150       | 22 x 4.3   | 6.6 x 1.5   | 400–1200 x 180–380 | 230–300    | 6 x 1.3     | 3.8 x 1.3     | In this study |
| *C. xylocarpi*         | 230–600 x 90–250 | 160–200      | 26 x 4     | 5.7 x 1.8   | 700–1200 x 400–480 | 200–250    | 8.5 x 1.4   | 3 x 1         | In this study |

Table 3. GenBank BLAST search from ITS1 and ITS2 of *Cytospora xylocarpi* (MFLUCC 17-0251) with sequence from GenBank identified as *Cytospora rhizophorae*.

| Taxon                  | Strain  | Host              | Country | Accessions | ITS1 | ITS2 | ITS1+ITS2 | Identities (I), Query cover (QC) | References |
|------------------------|---------|-------------------|---------|------------|------|------|----------|----------------------------------|------------|
| *C. "rhizophorae"*     | HAB16R13 | Cinnamomum porrectum | Malaysia | HQ336045  | 213/215 | 167/169 | 380/384 | I=98.9%, QC=99% | Harun et al. (2011) |
| *C. "rhizophorae"*     | M225    | Rhizophora mucronata | Philippines | KR056292 | 213/217 | 167/169 | 380/386 | I=98.4%, QC=100% | Unpublished |
| *C. "rhizophorae"*     | A761     | Morinda officinalis | China    | KU529867  | 213/217 | 166/169 | 379/386 | I=98.2%, QC=100% | Unpublished |
| *C. "rhizophorae"*     | MUCC302  | Eucalyptus grandis | Australia | EU301057  | 213/217 | 164/169 | 377/386 | I=97.7%, QC=100% | Unpublished |
| *C. rhizophorae*       | ATCC38475 | Rhizophora mangle | LA, USA | DQ996040  | 187/202 | 156/166 | 343/368 | I=93.2%, QC=100% | He et al. (2003) |
Table 4. Nucleotide differences in the ITS1+ITS2 of *Cytospora xylocarpi* (MFLUCC 17-0251) with sequence from GenBank identified as *Cytospora rhizophorae*.

| Taxon          | Strain            | ITS1 |   |   |   |   |   |   |   |   |   |   |   |   |   |   |
|----------------|-------------------|------|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| *C. xylocarpi* | MFLUCC 17-0251    | - G  | A | C | C | C | G | G | G | C | G | C | T | T | C | A | G |
| *C. rhizophorae* | ATCC38475        | G   | A | C | T | G | A | T | A | T | T | T | A | T | - | C | T | - | T |
| *C. "rhizophorae"* | HAB16R13      | ?   | G | A | C | C | C | G | G | G | C | G | C | T | T | C | A | T |   |
| *C. "rhizophorae"* | M225             | ?   | G | A | T | C | C | G | G | G | C | G | C | T | T | C | A | T |   |
| *C. "rhizophorae"* | A761             | ?   | G | A | T | C | C | G | G | G | C | G | C | T | T | C | A | T |   |
| *C. "rhizophorae"* | MUCC302          | ?   | G | A | T | C | C | G | G | G | C | G | C | - | T | C | A | T |   |

| Taxon          | Strain            | ITS2 |   |   |   |   |   |   |   |   |   |   |   |   |   |   |   |
|----------------|-------------------|------|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| *C. xylocarpi* | MFLUCC 17-0251    | C   | C | A | T | - | T | T | C | A | A | C | T |   |   |   |   |   |
| *C. rhizophorae* | ATCC38475        | T   | T | - | T | - | - | T | C | G | T | A | T |   |   |   |   |   |
| *C. "rhizophorae"* | HAB16R13      | C   | T | A | T | - | T | T | C | A | A | C | C |   |   |   |   |   |
| *C. "rhizophorae"* | M225             | C   | T | A | T | - | T | T | C | A | A | C | T |   |   |   |   |   |
| *C. "rhizophorae"* | A761             | C   | T | A | T | T | - | T | C | A | A | C | T |   |   |   |   |   |
| *C. "rhizophorae"* | MUCC302          | C   | T | A | - | - | - | - | T | A | A | C | T |   |   |   |   |   |
Table 5. Nucleotides differences in the ITS, ACT and RPB2 sequences of *Cytospora lumnitzericola*, *C. thailandica* and *C. xylocarpi*.

| Taxon                | Strain             | ITS          | ACT          |
|----------------------|--------------------|--------------|--------------|
|                      |                    | 29 88 91 92 93 94 96 97 99 101 102 103 104 105 106 107 108 111 | 439 468 485 487 488 74 78 80 92 95 96 97 107 122 125 129 136 137 | 139 146 147 148 149 150 152 159 165 198 209 210 212 215 216 217 218 223 |
| *C. lumnitzericola*  | MFLUCC 17-0508     | T C T T T T T C T C G G A C T A T A G | A A G C T C C G T C T C G A A A C A | |
| *C. thailandica*     | MFLUCC 17-0262     | T T T T T T T C T C A G - A C G C | A G - - T T T T T T T T C A A A - C A | |
| *C. thailandica*     | MFLUCC 17-0263     | T T T T T T T C T C A G - A C G C | A G - - T T T T T T T T C A A A - C A | |
| *C. xylocarpi*       | MFLUCC 17-0251     | C C C C - C C C C C G G G - G C G G | G G - - T T T T T T T T C A A T G - A - | |
| Taxon                  | Strain                     | ACT | RPB2 |
|------------------------|----------------------------|-----|------|
|                        |                            | 4   | 16   |
|                        |                            | 224 | 225  |
|                        |                            | 231 | 234  |
|                        |                            | 242 | 245  |
|                        |                            | 246 |      |
|                        |                            | 18  | 33   |
|                        |                            | 42  | 57   |
|                        |                            | 84  | 85   |
|                        |                            | 96  | 102  |
|                        |                            | 108 | 120  |
| C. lumnitzericola      | MFLUCC 17-0508             | C   | T    |
|                        |                            | G   | C    |
|                        |                            | -   | -    |
| C. thailandica         | MFLUCC 17-0262             | T   | T    |
|                        |                            | C   | T    |
|                        |                            | G   | T    |
|                        |                            | G   | T    |
|                        |                            | C   | A    |
|                        |                            | T   | C    |
|                        |                            | T   | C    |
|                        |                            | C   | T    |
|                        |                            | T   | C    |
|                        |                            | G   | A    |
| C. thailandica         | MFLUCC 17-0263             | T   | T    |
|                        |                            | C   | T    |
|                        |                            | G   | T    |
|                        |                            | G   | T    |
|                        |                            | C   | A    |
|                        |                            | T   | C    |
|                        |                            | T   | C    |
|                        |                            | T   | C    |
|                        |                            | C   | A    |
|                        |                            | G   | A    |
| C. xylocarpi           | MFLUCC 17-0251             | T   | T    |
|                        |                            | A   | C    |
|                        |                            | G   | T    |
|                        |                            | A   | C    |
|                        |                            | T   | C    |
|                        |                            | C   | C    |
|                        |                            | T   | C    |
|                        |                            | C   | A    |
|                        |                            | A   | A    |
|                        |                            | G   | A    |
|                        |                            | C   | A    |
|                        |                            | T   | T    |
|                        |                            | C   | C    |
|                        |                            | T   | C    |
|                        |                            | C   | C    |
|                        |                            | A   | A    |
|                        |                            | A   | G    |
|                        |                            | C   | A    |
|                        |                            | T   | T    |
|                        |                            | C   | C    |
|                        |                            | T   | C    |
|                        |                            | G   | A    |
|                        |                            | C   | A    |
|                        |                            | T   | C    |
|                        |                            | A   | C    |
|                        |                            | C   | T    |
|                        |                            | T   | T    |
|                        |                            | G   | G    |
|                        |                            | T   | C    |
|                        |                            | G   | A    |
|                        |                            | C   | A    |
|                        |                            | T   | T    |
|                        |                            | G   | G    |
|                        |                            | C   | A    |
|                        |                            | T   | T    |
|                        |                            | A   | C    |
|                        |                            | C   | T    |
|                        |                            | T   | T    |

All isolates are new taxa in this study; “-” gap (insertion/deletion); “?” missing data.
conidia (Kohlmeyer and Kohlmeyer 1971). However, the phylogenies, generated herein, show that *C. xylocarpi* is distinct from *C. rhizophorae* (ATCC 38475), a strain from *Rhizophora mangle* that was identified by Kohlmeyer, the author of the species (Fig. 2). The two species also differ by 25 substitutions in ITS1+ITS2 and were collected from different hosts. Therefore, the collection in the present study is designated as a new species.

Our phylogeny also indicates a close relationship to unpublished sequences from GenBank (Figs 1, 2). Given that no morphological descriptions are available for these, the similarity in the ITS1 and ITS2 sequence between our strain and the sequences from GenBank (HAB16R13, M225, A761, MUCC302) are presented in Table 3. Those strains were collected from different hosts (Table 3) and, together with our strain, show substantial variation in ITS1 and ITS2 (Table 4). More collections are needed to further study morphological and genetic variation in this group.

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**References**

Adams GC, Surve-Iyer RS, Iezzoni A (2002) Ribosomal DNA sequence divergence and group I introns within *Leucostoma* species, *L. cinctum*, *L. persoonii*, and *L. parapersoonii* sp. nov., ascomycetes that cause *Cytospora* canker of fruit trees. Mycologia 94: 947–967. https://doi.org/10.2307/3761863

Adams GC, Wingfield MJ, Common R, Roux J (2005) Phylogenetic relationships and morphology of *Cytospora* species and related teleomorphs (Ascomycota, Diaporthales, Valsaceae) from Eucalyptus. Studies in Mycology 52: 1–144.

Adams GC, Roux J, Wingfield MJ (2006) *Cytospora* species (Ascomycota, Diaporthales, Valsaceae): introduced and native pathogens of trees in South Africa. Australasian Plant Pathology 35: 521–548. https://doi.org/10.1071/AP06058
Alias SA, Jones EBG (2009) Marine fungi from mangroves of Malaysia. Institute Ocean and Earth Sciences, University Malaya, 108.

Alias SA, Zainuddin N, Jones EBG (2010) Biodiversity of marine fungi in Malaysian mangroves. Botanica Marina 53: 545–554. https://doi.org/10.1515/bot.2010.066

Ariyawansa HA, Hyde KD, Jayasiri SC, Buyck B, Chethana KWT, Dai DQ, Dai YC, Daranagama DA, Jayawardena RS, Lucking R, Ghobad-Nejhad M, Niskanen T, Thambugala KM, Voigt K, Zhao RL, Li GJ, Doilom M, Boonmee S, Yang ZL, Cai Q, Cui YY, Bahkali AH, Chen J, Cui BK, Chen JJ, Dayarathne MC, Dissanayake AJ, Ekanayaka AH, Hashimoto A, Hongsanan S, Jones EBG, Larsson E, Li WJ, Li QR, Liu JK, Luo ZL, Maharachchikumbura SSN, Mapook A, McKenzie EHC, Norphanphoun C, Konta S, Pang KL, Perera RH, Phookamsak R, Phukhamsakda C, Pinruan U, Randrianjohany E, Singtripop C, Tanaka K, Tian CM, Tibpromma S, Abdel-Wahab MA, Wanasinghe DN, Wijayawardene NN, Zhang JF, Zhang H, Abdel-Aziz FA, Wedin M, Merkelberg AM, Ammirati JE, Bulgakov TS, Lima DX, Callaghan TM, Callac P, Chang CH, Coca LF, Dal-Forno M, Dollhofer V, Fliegerova’ K, Greiner K, Griffith GW, Ho HM, Hofstetter V, Jeewon R, Kang JC, Wen TC, Kirk PM, Kyoto’vori I, Lawrey JD, Xing J, Li H, Liu ZY, Liu XZ, Liimatainen K, Thorsten Lumbsch H, Matsumura M, Moncada B, Nuankaew S, Parmen S, Santiago ALCMA, Sommai S, Song Y, de Souza CAF, de Souza- Motta CM, Su HY, Suetrong S, Wang Y, FongWS YH, Zhou LW, Re’blová’ M, Fournier J, Camporesi E, Luangsa-ard JJ, Tasanathai K, Khonsanit A, Thanakitpipattana D, Somrithipol S, Diederich P, Millanes AM, Common RS, Stadler M, Yan JY, Li XH, Lee HW, Nguyen TTT, Lee HB, Battistin E, Marsico O, Vizzini A, Vila J, Ercole E, Eberhardt U, Simonini G, Wen HA, Chen XH, Miettinen O, Spirin V, Hernawati (2015) Fungal diversity notes 111–252 taxonomic and phylogenetic contributions to fungal taxa. Fungal Diversity 75: 27–274. https://doi.org/10.1007/s13225-015-0346-5

Barr ME (1978) The Diaporthales in North America: with emphasis on Gnomonia and its segregates. Mycologia memoir 7: 1–232. https://doi.org/10.1002/fedr.19800910313

Bharathidasan R, Panneerselvam A (2011) Isolation and identification of endophytic fungi from Avicennia marina in Ramanathapuram District, Karankadu, Tamilnadu, India. European Journal of Experimental Biology 1: 31–36.

Cai L, Jeewon R, Hyde KD (2005) Phylogenetic evaluation and taxonomic revision of Schizothecium based on ribosomal DNA and protein coding genes. Fungal Diversity 19: 1–17.

Carbone I, Kohn LM (1999) A method for designing primer sets for speciation studies in filamentous ascomycetes. Mycologia 91: 553–556. https://doi.org/10.2307/3761358

Castlebury LA, Rossman AY, Jaklitsch WJ, Vasilyeva LN (2002) A preliminary overview of Diaporthales based on large subunit nuclear ribosomal DNA sequences. Mycologia 94: 1017–1031. https://doi.org/10.1080/15572536.2003.11833157

Castresana J (2000) Selection of conserved blocks from multiple alignments for their use in phylogenetic analysis. Molecular Biology and Evolution 17: 540–552. https://doi.org/10.1093/oxfordjournals.molbev.a026334

Chomnunti P, Hongsanan S, Aguirre-Hudson B, Tian Q, Peršoh D, Dhami MK, Hyde KD (2014) The sooty moulds. Fungal Diversity 66: 1–36. https://doi.org/10.1007/s13225-014-0278-5
Cribb AB, Cribb JW (1955) Marine fungi from Queensland-I. Papers of the University of Queensland. Department of Botany 3: 78–81.

Devadatha B, Sarma VV, Jeewon R, Wanasinghe DN, Hyde KD, Jones EBG (2018) Thyridariella, a novel marine fungal genus from India: morphological characterization and phylogeny inferred from multigene DNA sequence analyses. Mycological Progress, 1–14. https://doi.org/10.1007/s11557-018-1387-4

Doilom M, Manawasinghe IS, Jeewon R, Jayawardena RS, Tibpromma S, Hongsanan S, Meepol W, Lumyong S, Jones EBG, Hyde KD (2017) Can ITS sequence data identify fungal endophytes from cultures? A case study from Rhizophora apiculata. Mycosphere 8: 1869–1892. https://doi.org/10.5943/micosphere/8/10/11

Ehrenberg CG (1818) Sylvae Mycologicae Berolinenses. Formis Theophili Bruschcke, Berlin, Germany. [In Latin]

Eriksson OE (2001) Outline of Ascomycota. Myconet 6: 1–27.

Fan XL, Hyde KD, Liu M, Liang YM, Tian CM (2015a) Cytospora species associated with walnut canker disease in China, with description of a new species C. gigalocus. Fungal Biology 119: 310–319. http://dx.doi.org/10.1016/j.funbio.2014.12.011

Fan XL, Hyde KD, Yang Q, Liang YM, Ma R, Tian C (2015b) Cytospora species associated with canker disease of three antidesertification plants in northwestern China. Phytotaxa 197: 227–244. http://dx.doi.org/10.11646/phytotaxa.197.4.1

Fisher P, Spalding M (1993) Protected areas with mangrove habitat. World Conservation Centre, Cambridge, UK.

Fotouhifar KB, Hedjaroude GA, Leuchtmann A (2010) ITS rDNA phylogeny of Iranian strains of Cytospora and associated teleomorphs. Mycologia 102: 1369–1382. https://doi.org/10.3852/10-034

Hall TA (1999) BioEdit: a user-friendly biological sequence alignment editor and analysis program for Windows 95/98/NT. Nucleic Acids Symposium Series 41: 95–98.

Harun A, James RM, Lim SM, Abdul Majeed AB, Cole AL, Ramasamy K (2011) BACE1 Inhibitory Activity of Fungal Endophytes from Malaysian Medicinal Plants. BMC Complementary and Alternative Medicine 11: 79. https://doi.org/10.1186/1472-6882-11-79

He H, Janso JE, Williamson RT, Yang HY, Carter GT (2003) Cytosporacin, a highly unsaturated polyketide: application of the ACCORD-ADEQUATE experiment to the structural determination of natural products. The Journal of Organic Chemistry 68(16): 6079–82. https://doi.org/10.1021/jo030067f

Hyde KD, Jones EBG, LeañoStephen E, Pointing B, Poonyth AD, Vrijmoed LLP (1998) Role of fungi in marine ecosystems. Biodiversity & Conservation 7: 1147–1161. https://doi.org/10.1023/A:1008823515157

Hyde KD, Norphanphoun C, Abreu VP, Bazzicalupo1 A, Chethana KWT, Clericuzio M, Dayaratne MC, Dissanayake AJ, Ekanayaka1 AH, He MQ, Hongsanan S, Huang SK, Jayasiri SC, Jayawardena RS, Karunarathna A, Konta S, Kusan I, Lee H, Li J, Lin CG, Liu NG, Lu YZ, Luo ZL, Manawasinghe IS, Mapook A, Perera RH, Phokamsak R, Phukhamsakda C, Siedlecki I, Soares AM, Tennakoon DS, Tian Q, Tibpromma S, Wanasinghe DN, Xiao YP, Yang J, Zeng XY, Abdel-Aziz FA, Li WJ, Senanayake IC, Shang QJ, Daranagama DA, de Silva NI, Thambubala KM, Abdel-Wahab MA, Bahkali AH, Berbee ML, Boonmee S, Bhat DJ, Bulgakov TS,
Morphological and phylogenetic characterisation of novel *Cytospora* species...

Buyck B, Camporesi E, Castaneda-Ruiz RF, Chomnunti P, Doilom M, Dovana F, Gibertoni TB, Jadan M, Jeewon R, Jones EBG, Kang JC, Karunarathna SC, Lim YW, Liu JK, Liu ZY, Plautz Jr. HL, Lumyong S, Maharachchikumbura SSN, Matocec N, McKenzie EHC, Mesic A, Miller D, Pawlowska J, Pereira OL, Promputtha I, Romero AL, Ryvarden L, Su HY, Sutrong S, Tkalcev Z, Vizzini A, Wen TC, Wisitrasameewong K, Wrzosek M, Xu JC, Zhao Q, Zhao RL, Mortimer PE (2017) Fungal diversity notes 603–708: taxonomic and phylogenetic notes on genera and species. Fungal Diversity 87: 1–235. https://doi.org/10.1007/s13225-017-0391-3

Hyde KD, Chaivan N, Norphanphoun C, Boonmee S, Camporesi E, Chethana KWT, Dayaratne MC, de Silva NI, Dissanayake AJ, Ekanayaka AH, Hongsanan S, Huang SK, Jayasiri SC, Jayawardena R, Jiang HB, Karunarathna A, Lin CG, Liu JK, Liu NG, Lu YZ, Luo ZL, Maharachchimbura SSN, Manawasinghe IS, Prem D, Perera RH, Phukhamsakda C, Samarackoon MC, Senwanna C, Shang QJ, Tennakoon DS, Thambugala KM, Tibpromma, S, Wanasinghe DN, Xiao YP, Yang J, Zeng XY, Zhang JF, Zhang SN, Bulgakov TS, Bhat DJ, Cheewangkoon R, Goh TK, Jones EBG, Kang JC, Jeewon R, Liu ZY, Lumyong S, Kuo CH, McKenzie EHC, Wen TC, Yan JY, Zhao Q (2018) Mycosphere notes 169–224. Mycosphere 9: 271–430. https://doi.org/10.5943/mycosphere/9/2/8

Hyde KD, Jones EBG (1988) Marine mangrove fungi. Marine Ecology 9: 15–33. https://doi.org/10.1111/j.1439-0485.1988.tb00196.x

Hyde KD, Lee SY (1995) Ecology of mangrove fungi and their role in nutrient cycling: What gaps occur in our knowledge?. Hydrobiologia 295: 107–118. https://doi.org/10.1007/BF00029117

Index Fungorum (2018) www.indexfungorum.org

Jayasiri SC, Hyde KD, Ariyawansa HA, Bhat J, Buyck B, Cai L, Dai YC, Abd-Elsalam KA, Ertz D, Hidayat I, Jeewon R, Jones EBG, Bahkali AH, Karunarathna SC, Liu JK, Luangs-a-ard JJ, Lumbsch HT, Maharachchikumbura SSN, McKenzie EHC, Moncalvo JM, Ghobad-Nejhad M, Nilsson H, Pang KA, Pereira OL, Phillips AJL, Raspé O, Rollins AW, Romero AI, Etayo J, Selçuk F, Stephenson SL, Sutrong S, Taylor JE, Tsui CKM, Vizzini A, Abdel-Wahab MA, Wen TC, Boonmee S, Dai DQ, Daranagama DA, Dissanayake AJ, Ekanayaka AH, Fryar SC, Hongsanan S, Jayawardena RS, Li WJ, Perera RH, Phookamsak R, de Silva NI, Thambugala KM, Tian Q, Wijayawardene NN, Zhao RL, Zhao Q, Kang JC, Promputtha I (2015) The Faces of Fungi database: fungal names linked with morphology, phylogeny and human impacts. Fungal Diversity 74: 3–18. https://doi.org/10.1007/s13225-015-0351-8

Jeewon R, Liew ECY, Hyde KD (2002) Phylogenetic relationships of *Pestalotiopsis* and allied genera inferred from ribosomal DNA sequences and morphological characters. Molecular phylogenetics and evolution 25: 378–392. https://doi.org/10.1016/S1055-7903(02)00422-0

Jeewon R, Hyde KD (2016) Establishing species boundaries and new taxa among fungi: recommendations to resolve taxonomic ambiguities. Mycosphere 7: 1669–1677. https://doi.org/10.5943/mycosphere/7/11/4

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

Kirk PM, Cannon PF, Minter DW, Stalpers JA (2008) Ainsworth & Bisby’s dictionary of the fungi, 10th edn. CABI, Wallingford. https://doi.org/10.1079/9780851998268.0000
Kohlmeyer J (1969) Marine fungi of Hawaii including the new genus *Helicascus*. Canadian Journal of Botany 47: 1469–1487. https://doi.org/10.1139/b69-210

Kohlmeyer J, Kohlmeyer E (1971) Marine fungi from tropical America and Africa. Mycologia 63: 831–861. https://doi.org/10.2307/3758050

Kohlmeyer J, Kohlmeyer E (1979) Marine mycology. The higher fungi. Academic Press, New York, USA, 1–690.

Kumar S, Stecher G, Tamura K (2015) MEGA7: Molecular Evolutionary Genetics Analysis version 7.0. Molecular Biology and Evolution 33: 1870–1874. https://doi.org/10.1093/molbev/msw054

Larkin MA, Blackshields G, Brown NP, Chenna R, McGettigan PA, McWilliam H, Valentin F, Wallace IM, Wilm A, Lopez R, Thompson JD, Gibson TJ, H PA iggins DG (2007) Clustal W and Clustal X version 2.0. Bioinformatics 23: 2947–2948. https://doi.org/10.1093/bioinformatics/btm404

Lawrence DP, Travadon R, Pouzoulet J, Rolshausen PE, Wilcox WF, Baumgartner K (2017) Characterization of *Cytospora* isolates from wood cankers of declining grapevine in North America, with the descriptions of two new *Cytospora* species. Plant Pathology 66: 713–725. https://doi.org/10.1111/ppa.12621

Li J, Jeewon R, Phookamsak R, Bhat DJ, Mapook A, Chukeatirote E, Hyde KD, Lumyong S, McKenzie EHC (2018) * Marinophialophora garethjonesii* gen. et sp. nov.: a new hyphomycete associated with *Halocyphina* from marine habitats in Thailand. Phytotaxa 345: 1–12. http://doi.org/10.11646/phytotaxa.345.1.1

Liu AR, Chen SC, Jin WJ, Zhao PY, Jeewon R, Xu T (2012) Host specificity of endophytic *Pestalotiopsis* populations in mangrove plant species of South China. African journal of microbiology research 6: 6262–6269. https://doi.org/10.5897/AJMR12.766

Liu JK, Hyde KD, Jones EBG, Ariyawansa HA, Bhat DJ, Boonmee S, Maharachchikumbura SSN, McKenzie EHC, Phookamsak R, Phukhamsakda C, Shenoy BD, Abdel-Wahab MA, Buyck B, Chen J, Chethana KWT, Singtripop C, Dai DQ, Dai YC, Danaramaga DA, Dissanayake AJ, Doliom M, D’souza MJ, Fan XL, Goonasekara ID, Hirayama K, Hongsanan S, Jayasiri SC, Jayawardena RS, Karunarathna SC, Li WJ, Mapook A, Norphanphoun C, Pang KL, Perera RH, Peršoh D, Pinruan U, Senanayake IC, Somrithipol S, Suatrung S, Tanaka K, Thambugala KM, Tian Q, Tibpromma S, Udayanga D, Wijayawardene NN, Wanasinghe D, Wisitrassameewong K, Zeng XY, Abdel-Aziz FA, Adamčík S, Bahkali AH, Boonyuen N, Bulgakov T, Callac P, Chomnunti P, Greiner K, Hashimoto A, Hofstetter V, Kang JC, Lewis D, Li XH, Liu ZY, Liu ZY, Matumura M, Mortimer PE, Rambold R, Randrianjohany E, Sato G, Indrasutdhi VS, Tian CM, Verbeke A, Brackel W, Wang Y, Wen TC, Xu JC, Yan JY, Zhao RL, Camporesi E (2015) Fungal Diversity Notes 1-100: Taxonomic and phylogenetic contributions to fungal species. Fungal Diversity 72: 1–197. https://doi.org/10.1007/s13225-015-0324-y

Matheny PB (2005) Improving phylogenetic inference of mushrooms with RPB1 and RPB2 nucleotide sequences (Inocybe; Agaricales). Molecular Phylogenetics and Evolution 35: 1–20. https://doi.org/10.1016/j.ympev.2004.11.014

McMillan Jr RT (1964) Studies of a recently described *Cercospora* on *Rhizophora mangle*. Plant Disease Reporter 48: 909–911.
Morphological and phylogenetic characterisation of novel *Cytospora* species...

Miller MA, Pfeiffer W, Schwartz T (2010) Creating the CIPRES Science Gateway for inference of large phylogenetic trees. Proceedings of the Gateway Computing Environments Workshop (GCE), 14 Nov. 2010, New Orleans, LA, 1–8. https://doi.org/10.1109/GCE.2010.5676129

Norphanphoun C, Doilom M, Daranagama DA, Phookamsak R, Wen TC, Bulgakov TS, Hyde KD (2017) Revisiting the genus *Cytospora* and allied species. Mycosphere 8: 51–97. https://doi.org/10.5943/mycosphere/8/1/7

Nylander JAA (2004) MrModeltest 2.0. Program distributed by the author. Evolutionary Biology Centre, Uppsala University.

O’Donnell K (1993) *Fusarium* and Its Near Relatives. In: Reynolds DR, Taylor JW (Eds) The Fungal Holomorph: Mitotic, Meiotic and Pleomorphic Speciation in Fungal Systematic, CAB International, Wallingford, 225–233.

Rambaut A, Suchard MA, Xie D, Drummond AJ (2013) Tracer v1.6, http://beast.bio.ed.ac.uk/software/tracer/ [accessed June 2018]

Rambaut A (2014) FigTree v1.4: Tree figure drawing tool. http://tree.bio.ed.ac.uk/software/figtree/

Ronquist F, Teslenko M, van der Mark P, Ayres DL, Darling A, Höhna S, Larget B, Liu L, Suchard MA, Huelsenbeck JP (2012) MrBayes 3.2: efficient Bayesian phylogenetic inference and model choice across a large model space. Systematic Biology 61: 539–542. https://doi.org/10.1093/sysbio/sys029

Senanayake IC, Crous PW, Groenewald JZ, Maharachchikumbura SSN, Jeewon R, Phillips AJL, Bhat JD, Perera RH, Li QR, Li WJ, Tangthirasunun N (2017) Families of Diaportheales based on morphological and phylogenetic evidence. Studies in Mycology 86: 217–296. https://doi.org/10.1016/j.simyco.2017.07.003

Spatafora JW, Sung GH, Johnson D, Hesse C, O’Rourke B, Serdani M, Spotts R, Lutzoni F, Hofstetter V, Miadlikowska J, Reeb V, Gueidan C, Fraker E, Lumbsch T, Lucking R, Schmitt I, Hosaka K, Aptroot A, Roux C, Miller AN, Geiser DM, Hafellner J, Westmark G, Arnold AE, Budel B, Rauhut A, Hewitt D, Untereiner WA, Cole MS, Scheidegger C, Schultz M, Sipman H, Schoch CL (2006) A five-gene phylogeny of Pezizomycotina. Mycologia 98: 1018–1028. https://doi.org/10.3852/mycologia.98.6.1018

Stamatakis A (2006) RAxML-VI-HPC: maximum likelihood - based phylogenetic analyses with thousands of taxa and mixed models. Bioinformatics 22: 2688–2690. https://doi.org/10.1093/bioinformatics/btl446

Stevens FL (1920) New or noteworthy Puerto Rican fungi. Bot Gaz 70: 399–402. https://doi.org/10.1086/332764

Swe A, Jeewon R, Pointing SB, Hyde KD (2008a) Taxonomy and phylogeny of *Arthrobotrys mangrovispora*, a new marine nematode-trapping fungal species: Botanica Marina 51: 331–338. https://doi.org/10.1515/BOT.2008.043

Swe A, Jeewon R, Hyde KD (2008b) Nematode-trapping fungi from mangrove habitats. Cryptogamie Mycologie 29: 333–354.

Swofford DL (2002) PAUP: phylogenetic analysis using parsimony, (*and other methods). Version 4.0 b10. Sinauer Associates, Sunderland, MA. https://doi.org/10.1111/j.0014-3820.2002.tb00191.x
Talavera G, Castresana J (2007) Improvement of phylogenies after removing divergent and ambiguously aligned blocks from protein sequence alignments. Systematic Biology 56: 564–577. https://doi.org/10.1080/10635150701472164

Tibpromma S, Hyde KD, Jeewon R, Maharachchikumbura SSN, Liu JK, Bhat DJ, Jones EBG, McKenzie EHC, Camporeesi E, Bulgakov TS, Doilom M, de Azevedo Santiago ALCM, Das K, Manimohan P, Gibertoni TB, Lim YW, Ekanayaka AH, Thongbai B, Lee HB, Yang JB, Kirk PM, Sysouphanthong P, Singh SK, Boonmee S, Dong W, Raj KNA, Latha KPD, Phookamsak R, Phukhamsakda C, Konta S, Jayasiri SC, Norphanphoun C, Tennakoon DS, Li JF, Dayaratnhe MC, Perera RH, Xiao Y, Wanasinghe DN, Senanayake IC, Goonasekara ID, de Silva NI, Mapook A, Jayawardena RS, Disanayake AJ, Manawasinghe IS, Chethana KWT, Luo ZL, Hapurarchchi KK, Baghela A, Soares AM, Vizzini A, Meiras-Ottoni A, Mešić A, Dutta AK, de Souza CAF, Richter C, Lin CG, Chakrabarty D, Daranagama DA, Lima DX, Chakraborty D, Ercole E, Wu F, Simonini G, Vasquez G, da Silva GA, Plautz HL Jr, Ariyawansa HA, Lee H, Kušan I, Song J, Sun JZ, Karmakar J, Hu K, Semwal KC, Thambugala KM, Voigk G, Acharya K, Rajeshkumar KC, Ryvarden L, Jadan M, Hosen MI, Mikšík M, Samarakoone MC, Wijayawardene NN, Kim NK, Matočec N, Singh PN, Tian Q, Bhatt RP, de Oliveira RJV, Tulloss RE, Aamir S, Kaewchai S, Svetasheva STY, Nguyen TTT, Antonín V, Li WJ, Wang Y, Indoliya Y, Tkalcčec Z, Elgorban AM, Bahkali AH, Tang AMC, Su HY, Zhang H, Promputtha I, Luangsa-ard J, Xu JC, Yan J, Chuan KJ, Mortimer PE, Chomnunti P, Zhao Q, Phillips AJL, Nontachaiyapoom S, Wen TC, Karunarathna SC (2017) Fungal diversity notes 491–602: taxonomic and phylogenetic contributions to fungal taxa. Fungal Diversity 83: 1–261. https://doi.org/10.1007/s13225-017-0378-0

Wehmeyer LE (1975) The pyrenomycetous fungi. Mycologia memoir 6: 1–250.

White T, Bruns T, Lee S, Taylor J (1990) Amplification and direct sequencing of fungal ribosom- nal RNA genes for phylogenetics. In: Innis MA, Gelfand DH, Sninsky JJ, White TJ (Eds) PCR Protocols: A Guide to Methods and Applications, Academic Press, Inc., New York, 315–322. http://doi.org/10.1016/b978-0-12-372180-8.50042-1

Wier AM, Tattar TA, Klekowski EJ (2000) Disease of red mangrove (Rhizophora mangle) in southwest Puerto Rico caused by Cytospora rhizophorae. Biotropica 32: 299–306. https://doi.org/10.1646/0006-3606(2000)032[0299:DORMRM]2.0.CO;2

Wijayawardene NN, Hyde KD, Lumbsch HT, Liu JK, Maharachchikumbura SSN, Ekanayaka AH, Tian Q, Phookamsak R (2018) Outline of Ascomycota: 2017. Fungal Diversity 88: 167–263. https://doi.org/10.1007/s13225-018-0394-8

Yang Q, Fan ZL, Crous PW, Liang YM, Tian CM (2015) Cytospora from Ulmus pumila in Northern China. Mycological Progress 14: 74. https://doi.org/10.1007/s11557-015-1096-1

Zhang QT, He M, Lu Q, Liang J, Zhang XY (2013) Morphological and molecular identification of Cytospora germanica causing canker on Populus spp. in China. Plant Disease 97: 846. http://dx.doi.org/10.1094/PDIS-11-12-1065-PDN

Zhang QT, Lu Q, He M, Decock C, Zhang XY (2014) Cytospora palm sp. nov. (Diaporthales, Ascomycota), a canker agent on Cotinus coggyria (Anacardiaceae) in Northern China. Cryptogamie Mycologie 35: 211–220. https://doi.org/10.7872/crmy.v35.iss3.2014.211