# Cytospora: an important genus of canker pathogens

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Abstract: *Cytospora* species have commonly been reported as important plant pathogenic fungi with wide host ranges and geographic distributions. With the increase in the number of cryptic species being described, a comprehensive global taxonomic revision of the genus *Cytospora* is required. The present study includes 399 isolates from 32 countries. These isolates were subjected to DNA sequence analysis for five genomic loci (ITS, *act1, rpb2, tef1-a* and *tub2*). Based on these data, it could be confirmed that *Cytospora, Leucostoma, Valsa, Valsella* and *Valseutypella* are congeneric. Furthermore, 111 species of *Cytospora* could also be reassessed, 44 species and four combinations newly introduced, and new typifications proposed for a further three species. Three asexual morphological groups (including 13 asexual morphological types) and three sexual morphological groups (including eight sexual morphological types) were designated. The present study explored the species diversity of *Cytospora* and re-evaluated the identity of all cultures in the Westerdijk Fungal Biodiversity Institute (Utrecht, The Netherlands) that were deposited as either *Cytospora* or as one of its related genera. This is the most comprehensive phylogenetic analysis thus far conducted on *Cytospora* and the results contribute to an increased understanding of the taxonomy of these important fungi. It is also hoped that the findings will lead to improved management strategies for diseases associated *Cytospora* species.

Key words: Cytosporaceae, Diaporthales, multi-gene phylogeny, taxonomy.

Taxonomic novelties: New species: Cytospora acericola X.L. Fan & C.M. Tian, C. adamsii Jami, Crous & M.J. Wingf., C. beijingensis L. Lin & X.L. Fan, C. betulae Jami, Crous & M.J. Wingf., C. brabeji Jami, Crous & M.J. Wingf., C. castaneicola L. Lin & X.L. Fan, C. cerebriformis L. Lin & X.L. Fan, C. conceptaculata L. Lin & X.L. Fan, C. crataegina X.L. Fan & C.M. Tian, C. deginensis L. Lin & X.L. Fan, C. digingensis L. Lin & X.L. Fan, C. eastringensis L. Lin & X.L. Fan, C. elaeagnina L. Lin & X.L. Fan, C. fraxinea X.L. Fan & C.M. Tian, C. guyuanensis L. Lin & X.L. Fan, C. jiufengensis L. Lin & X.L. Fan, C. lauricola L. Lin & X.L. Fan, C. Ihasaensis L. Lin & X.L. Fan, C. Iijiangensis L. Lin & X.L. Fan, C. Ivxinensis L. Lin & X.L. Fan, C. malvicolor X.L. Fan & C.M. Tian, C. multiseriata L. Lin & X.L. Fan, C. nanyangensis X.L. Fan & C.M. Tian, C. polyspora X.L. Fan & C.M. Tian, C. pseudochrysosperma L. Lin & X.L. Fan, C. ginghaiensis L. Lin & X.L. Fan, C. gingshuiensis L. Lin & X.L. Fan, C. sanbaensis L. Lin & X.L. Fan, C. shaanxiensis L L. Lin & X.L. Fan, C. sidaohensis L. Lin & X.L. Fan, C. sinensis L. Lin & X.L. Fan, C. songshanensis L. Lin & X.L. Fan, C. suecica Jami, Crous & M.J. Wingf., C. syringina L. Lin & X.L. Fan, C. tenebrica L. Lin & X.L. Fan, C. tetraspora L. Lin & X.L. Fan, C. tongzhouensis X.L. Fan, & C.M. Tian, C. uniloculata L. Lin & X.L. Fan, C. washingtonensis Jami, Crous & M.J. Wingf., C. xiaolongmenensis L. Lin & X.L. Fan, C. yinchuanensis L. Lin & X.L. Fan, C. yuduensis L. Lin & X.L. Fan, C. yulinensis L. Lin & X.L. Fan. New combinations: Cytospora auerswaldii (Nitschke) L. Lin & X.L. Fan, C. multicollis (Checa et al.) L. Lin, X.L. Fan & Crous, C. tristicha (De Not.) L. Lin, X.L. Fan & Crous, C. weiriana (Petr.) X.L. Fan & Crous. New replacement names: Cytospora desmazieri L. Lin, X.L. Fan & Crous, C. fuckeliana L. Lin, X.L. Fan & Crous, C. hoffmannii L. Lin, X.L. Fan & Crous, C. massarii L. Lin, X.L. Fan & Crous, C. nitschkeana L. Lin, X.L. Fan & Crous, C. saccardoi L. Lin, X.L. Fan & Crous. New synonyms: Cytospora ampulliformis Norph., Bulgakov, T.C. Wen & K.D. Hyde, C. brevispora (G.C. Adams & Jol. Roux) G.C. Adams & Rossman, C. cenisia Sacc., C. ceratospermopsis C.M. Tian & X.L. Fan, C. cotini Norph., Bulgakov & K.D. Hyde, C. ershadii Zafari & Hanifeh, C. erumpens Norph., Bulgakov, T.C. Wen & K.D. Hyde, C. fraxinigena Senan., Camporesi & K.D. Hyde, C. galegicola Q.J. Shang, E. Camporesi & K.D. Hyde, C. granati D.P. Lawr., L.A. Holland & Trouillas, C. hippophaicola Spetik, Eichmeier, Gramaje, Stuskova & Berraf-Tebbal, C. massariana Sacc., C. nivea (Hoffm.) Sacc., C. parakantschavelii Norph., Bulgakov, T.C. Wen & K.D. Hyde, C. parasitica Norph., Bulgakov & K.D. Hyde, C. paratranslucens Norph., Bulgakov, T.C. Wen & K.D. Hyde, C. pini Desm., C. populicola D.P. Lawr., L.A. Holland & Trouillas, C. predappioensis Q.J. Shang, Norph., Camporesi & K.D. Hyde, C. quercicola Senan., Camporesi & K.D. Hyde, C. rosae Senan., Camporesi & K.D. Hyde, C. salicella Sacc., C. vinacea D.P. Lawr., Travadon & Pouzoulet, Valsa germanica Nitschke, V. massariana De Not., V. nivea (Hoffm.) Fr., Valsella salicis Fuckel, Sphaeria nivea Hoffm. Typification: Lecto- and epitypifications (basionyms): Sphaeria chrysosperma Pers., Valsa eucalypti Cooke & Harkn., Valsella salicis Fuckel.

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# INTRODUCTION

The genus *Cytospora* includes fungal species that occur mostly on woody plants including angiosperms and gymnosperms. Species of *Cytospora* (sexual morphs including *Leucostoma*, *Valsa*, *Valsella* 

and *Valseutypella*) have a cosmopolitan distribution and are most likely endophytes or latent pathogens in healthy plant tissues. They can also become important pathogens when their host trees are under stress (Schoeneweiss 1981, 1983). In this pathogenic phase, most *Cytospora* species cause canker diseases (Sinclair

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*et al.* 1987, Farr *et al.* 1989, Fan *et al.* 2020). The sporocarps of these fungi are then found on the dying wood associated with these cankers.

The family *Cytosporaceae* was introduced by Fries (1825) but later placed in synonymy under *Valsaceae* (1861), which was introduced as "*Valsarum*" by Tulasne & Tulasne (1863). Von Höhnel (1917, 1918) subsequently placed its genera in the subfamily "*Valseen*" (*Diaporthaceae*), which comprised six allantoid-spored genera, *i.e.*, *Leucostoma*, *Peroneutypa*, *Scoptria*, *Valsa*, *Valsella* and *Valseutypella*. Nannfeldt (1932) elevated *Valseen* to ordinal level as *Valsales*, but Gilman *et al.* (1957) considered these taxa as the subfamily *Valseae* within *Diaporthaceae*, which was followed by Kobayashi (1970). Wehmeyer (1975) separated *Valsaceae* from *Diaporthaceae* with *Leucostoma*, *Glomerella*, *Scoptria*, *Valsa* and *Valsella*.

Dennis (1978) merged almost all allantoid-spored genera (including Leucostoma, Valsa and Valsella) into the Diatrypaceae. Barr (1978) recognised the connection of Valsaceae and Diaporthaceae based on the ascomatal characters and treated Diaporthaceae as synonym of Valsaceae, including 15 genera (Amphiporthe, Apioplagiostoma, Clypeoporthella, Cryphonectria, Cryptodiaporthe, Diaporthe, Hypospilina, Leucostoma, Linospora, Ophiovalsa, Plagiosphaera, Plagiostoma, Pleuroceras, Valsa and Valsella). However, many of them were considered as individual families in Diaporthales based on an LSU phylogeny, and only three genera (Leucostoma, Valsa and Valsella) were retained in Valsaceae as an individual family (Castlebury et al. 2002, Rossman et al. 2005). Adams et al. (2005) treated the asexual genus Cytospora in Valsaceae and accepted the hypothesis proposed in previous studies (Défago 1942, Urban 1957, 1958, Barr 1978, 1990), which considered the sexual genera (Leucocytospora, Leucostoma, Valsella and Valseutypella) as synonyms of Valsa without checking type materials. Senanayake et al. (2017) regarded Cytospora, Pachytrype, Paravalsa, Waydora and Xenotypa as genera of Cytosporaceae, of which only the genera Cytospora, Pachytrype and Waydora had DNA data available for phylogenetic comparison.

The genus *Cytospora* was described by Ehrenberg (1818). Donk (1964) designated *Cytospora chrysosperma* (syn.: *Naemaspora chrysosperma*), which had been recorded from *Populus nigra* in Europe, as the lectotype species. *Cytospora* species are best known as asexual morphs of *Valsa* (1825). The generic name *Valsa* was first introduced by Adanson (1763), but Fries (1849) placed the species of *Valsa* Adans. in *Diatrype* Fr. and simultaneously introduced the other group of fungi as *Valsa* Fr. with *Valsa ambiens* (Pers.) Fr. as type species. Tulasne & Tulasne (1863) proposed that *Cytospora* and *Valsa* represent two forms of the same organism.

Three other genera, *Leucostoma, Valsella* and *Valseutypella*, were described as having *Cytospora* asexual morphs. However, there was no clear evidence to show that these genera were truly distinct from *Valsa* (Gilman *et al.* 1957, Vasilyeva 1988). *Leucostoma* (1917) was first introduced as a subgenus of *Valsa* by Nitschke (1870), but was later elevated to generic level by von Höhnel (1917) based on *Leucostoma massarianum* (De Not.) Höhn. *Leucostoma* had been distinguished from *Valsa* based on the presence of a black zone line produced between the ascoma and the host tissue. However, the black zone was not produced by all *Leucostoma* species, and occasionally present in some *Valsa* species (Spielman 1980). Vasilyeva (1988, 1994) believed that this morphological feature was insufficient to distinguish taxa at generic level. *Valsella* (1870) was initially introduced as subgenus *Leucostoma*, but it produced more than eight ascospores per ascus

(Barr 1978). Therefore, Vasilyeva (1994) re-described *Valsella* as a distinct genus based on *Valsella salicis*. *Valseutypella* (1919) was introduced by von Höhnel (1918) to accommodate *Diatrype tristicha*, which has perithecia entirely surrounded by a stoma of pseudoparenchymatous (or sclerotial) cells. Vasilyeva (1994) placed *Valseutypella tristicha* into the genus *Valsa* as *V. tristicha*. Rossman *et al.* (2015) rechecked the ITS and LSU rDNA sequences of *Valseutypella* spp., and accepted this hypothesis.

In the past, morphological features have played an important role in the classification of the sexual morphs in Cytospora. Saccardo (1882) treated Valsa in two groups, Macrosporae and Microsporae, based on the size of the ascospores and an intermediate group Mesosporae was added by Ellis & Everhart (1892). However, Spielman (1985) introduced sections that varied based on the morphological features of the stromata. The three sections Cypri, Monostichae and Valsa were applied to the genus Valsa as well as to three subgenera Leucostoma, Valsella and Valseutypella (Munk 1953, Gilman et al. 1957, Gvritishvili 1982, Adams et al. 2005). Species in section Valsa have large asci, ascospores and perithecia that are circinately arranged. Section Monostichae has small asci, ascospores and perithecia, while section Cypri has large asci and ascospores but few large perithecia. The subgenus Leucostoma has large asci and ascospores and few perithecia that are circinately arranged and the subgenus Valsella is similar to Leucostoma but its asci are polysporous. The subgenus Valseutypella, however, has perithecia surrounded by pseudoparenchyma. Barr (1990) retained four distinct genera, namely Leucostoma, Valsa, Valsella and Valseutvpella.

Morphological features have also been used in the taxonomy of the asexual morphs of *Cytospora*. Von Höhnel (1914–1923) proposed six distinct asexual genera including *Cytospora*, *Cytophoma*, *Cytosporopsis*, *Lamyella*, *Leucocytospora* and *Torsellia*, based on the shape of their locules. *Cytophoma* and *Cytosporopsis* have individual globose locules but a stromatic pillar formed in the locule of *Cytosporopsis*. *Cytospora* and *Leucocytospora* have complex and labyrinthiform locules while *Leucocytospora* has a black-shaped conceptacle, which distinguishes it from *Cytospora*. *Torsellia* and *Lamyella* both have multiple locules with each locule having an independent wall, but *Torsellia* has a single ostiole while *Lamyella* has several ostioles (Adams *et al.* 2005).

Morphological characters have failed to provide robust criteria to distinguish species of Cytospora. For example, Urban (1958) and Spielman (1985) investigated the correlation between ascomatal forms and conidiomatal locule types because they believed that there would be a correlation between the morphologies of sexual and asexual morphs. However, their conidiomatal morphologies overlapped with several sections of sexual morphs. Adams et al. (2005) rejected the notion of a correlation of features between the sexual and asexual morphs in Cytospora. Based on morphological features and rDNA-ITS sequence analyses Adams et al. (2005) thus reduced the genera Leucostoma, Valsella and Valseutypella to synonymy under Valsa or Cytospora, depending on the presence or absence of a known sexual morph, respectively. Following the end of dual nomenclature for pleomorphic fungi (Wingfield et al. 2012, Crous et al. 2015), the older and more commonly encountered genus Cytospora (1818) was chosen to be placed on the list of protected fungi over its sexual morphs Valsa (1825), Valsella (1870), Leucostoma (1917), Valseutypella (1919) and Leucocytospora (1927) (Fan et al. 2015a, b, Rossman et al. 2015).

Various Cytospora species cause important plant diseases, the most common being stem cankers of woody hosts. Only some

of the more recent records are included here. For example, they include C. palmoides on Cotinus coggygria (Zhang et al. 2014), C. carbonacea, C. pruinopsis and C. ribis on Ulmus spp. (Yang et al. 2015), C. atrocirrhata, C. chrysosperma, C. gigalocus, and C. sacculus on Juglans regia (Fan et al. 2015a), C. notastroma on Populus tremuloides (Kepley et al. 2015) and C. chrysosperma on more than 80 hosts (Fan et al. 2020, Farr & Rossman 2023). Cytospora sacculus has also been documented as initiating other pathologies such as stem and root rot on Ziziphus jujuba (Du et al. 2013, Chen et al. 2016). Infection of Punica granatum and Salix alba by C. punicae and C. salicicola respectively can result in branch dieback and collar rot (Palavouzis et al. 2015, Triki et al. 2015, Li et al. 2016), a leaf spot disease of Lumnitzera racemosa is caused by C. lumnitzericola and branch canker of Xylocarpus is caused by C. thailandica and C. xylocar (Norphanphoun et al. 2017). In South Africa, C. carpobroti causes dieback of Carpobrotus edulis (Jami et al. 2018), and C. euphorbiicola has been associated with dieback of Euphorbia mauritanica (Marincowitz et al. 2023). On conifers, C. beilinensis, C. bungeanae, C. juniperina, C. kunzei, C. platycladi, C. platycladicola, and C. pini have been associated with cankers on Juniperus, Platycladus and Pinus in China, Japan, South Africa, Switzerland and the USA (Kobayashi 1970, Adams et al. 2005, Kavak 2005, Fan et al. 2020).

Cytospora species have various life strategies. Some species have been isolated from asymptomatic bark, xylem and from leaves of many woody trees as endophytes (Bettucci & Saravay 1993, Fisher et al. 1993, Bills et al. 1996). They can be "xylotropic" (xylem-inhabiting) endophytes (Chapela 1989), saprophytic (Christensen 1940) or epiphytic (Mcintyre et al. 1996). They typically cause disease when their hosts are subjected to abiotic stress that negatively impacts on tree health. Drought and temperature stresses (Gibbs 1957, Bertrand & English 1976, Schoeneweiss 1983, Guyon et al. 1996), nutrient deficiency (Abebe et al. 1990, Burks et al. 1998), frost (Reich & van der Kamp 1993) and fire (Dearness & Hansbrough 1934) contribute to the susceptibility of trees to Cytospora infection. For example, studies on the effect of drought, flooding and defoliation on the development of Cytospora species found that drought and warmer seasonal temperatures resulted in larger cankers on poplar and apple trees (Bloomberg 1962a, b, Schoeneweiss 1983, Guyon et al. 1996, Worrall et al. 2010).

*Cytospora* species have a world-wide distribution and have been reported from angiosperms, gymnosperms, woody plants and non-woody plants such as *Carpobrotus edulis*, *Triticum aestivum* and *Saccharum officiarum*. More than 600 hosts have been recorded for these fungi from over 60 different countries (Farr & Rossman 2023). Synonymies and frequent name changes within *Cytospora* have caused much confusion for plant pathologists and mycologists. Thus, a systematic account of the genus *Cytospora* that considers both morphology and phylogenetic analyses is required. Several recent papers have provided updated phylogenies for the genus based on multigene data using ex-type or reference strains (Norphanphoun *et al.* 2017, 2018, Lawrence *et al.* 2018, Fan *et al.* 2020). However, it remains unclear whether *Cytospora* is monophyletic or not.

Therefore, the aims of this study were to 1) re-evaluate strains in the Westerdijk Fungal Biodiversity Institute, Utrecht, The Netherlands (CBS culture collection) identified as cytosporalike, which have been collected from 32 countries; 2) identify 260 newly collected specimens to explore the species diversity of *Cytospora*; 3) resolve the position of the type species of *Cytospora*, *Leucostoma*, *Valsa*, *Valsella*, and *Valseutypella*; and 4) provide a



new framework for *Cytospora* including morphological characters and phylogenetic inference. The overall intention has been to provide a comprehensive and current document that can serve as a foundation for future studies of related genera and species.

### MATERIALS AND METHODS

### Isolates

A total of 399 fungal strains from 32 countries were either newly collected or obtained from the CBS culture collection and the working collection of Pedro W. Crous (CPC) housed at the Westerdijk Institute (Table S1). In addition, new collections were made for type species evaluation and possible epitypification in China and Europe. Fungarium acronyms are according to Thiers (2024). Cultures were grown on 2 % (w/v) malt extract agar (MEA; Biolab) or 1.8 % (w/v) potato dextrose agar (PDA) and incubated at 25 °C. Fresh specimens collected in China were deposited in the Museum of the Beijing Forestry University (BJFC), American Type Culture Collection (ATCC) and the fungarium of Westerdijk Fungal Biodiversity Institute (CBS H). Cultures were deposited in the China Forestry Culture Collection Centre (CFCC; http://www.cfcc-caf.org. cn/) and the Westerdijk Fungal Biodiversity Institute (CBS; https:// wi.knaw.nl/).

### DNA isolation, amplification and sequencing

Total genomic DNA was extracted from cultures growing on MEA or PDA using the Wizard<sup>®</sup> Genomic DNA Purification Kit (Promega, USA) based on the manufacturer's instructions. Five loci were used for comparison-based phylogenetic analyses to determine the identities of the isolates. These included the internal transcribed spacer region of the nuclear ribosomal RNA gene (ITS) using primers ITS5 and ITS4, the partial actin (*act1*) using primers ACT512F and ACT728R, the partial RNA polymerase II second largest subunit (*rpb2*) using primers RPB2-5F and RPB2-7cR, the partial translation elongation factor 1- $\alpha$  (*tef1-\alpha*) gene amplified with primers Bt2a and Bt2b. Primer sequences and corresponding references can be found in Chen *et al.* (2011) and Videira *et al.* (2016).

The PCR amplifications were performed on a GeneAmp PCR System 9700 (Applied Biosystems, Foster City, CA, USA). The PCR mixture for all genes consisted of 1 µL genomic DNA, 3 mM MgCl<sub>2</sub>, 20 µM of each dNTP, 0.2 µM of each primer and 0.25 U BIOTAQ DNA polymerase (Bioline). Conditions for PCR of ITS and  $tef1-\alpha$ genes constituted an initial denaturation step of 2 min at 95 °C, followed by 35 cycles of 30 s at 95 °C, 30 s at 48 °C and 1 min at 72 °C and a final denaturation step of 8 min at 72 °C. Conditions for act1 and tub2 genes constituted an initial denaturation step of 2 min at 95 °C, followed by 35 cycles of 30 s at 95 °C, 30 s at 55 °C and 1 min at 72 °C and a final denaturation step of 8 min at 72 °C. For the rpb2 gene, the amplification consisted of five cycles of 45 s at 95 °C, 45 s at 56 °C and 2 min at 72 °C, then five cycles with 53 °C annealing temperature and 30 cycles with 50 °C annealing temperature. The PCR products were sequenced in both directions using the PCR primers and the BigDye Terminator v. 3.1 Cycle Sequencing Kit (Applied Biosystems, Foster City, CA, USA) and performed with an ABI Prism 3730xl DNA Analyzer (Applied Biosystems) according to the manufacturer's instructions.

### **Phylogenetic analyses**

DNA sequencing electropherograms generated by each primer combination were assembled using Seqman v. 7.1.0 in the DNASTAR Lasergene core suite software (DNASTAR Inc., Madison, WI, USA). Reference sequences were selected based on ex-type sequences available from relevant published literature (Adams *et al.* 2005, Lawrence *et al.* 2017, 2018, Norphanphoun *et al.* 2017, 2018, Fan *et al.* 2020, Hanifeh *et al.* 2022, Lin *et al.* 2023b) (Table S1). *Diaporthe vaccinii* (CBS 160.32) was selected as outgroup. All sequences were aligned using MAFFT v. 7 (Katoh & Standley 2013) with default settings (http://mafft.cbrc.jp/alignment/server/index.html) and edited manually using MEGA v. 6 (Tamura *et al.* 2013). Some characters were excluded from both ends of the alignments to approximate the size of our sequences to those included in the dataset.

Phylogenetic analyses for all the datasets were performed using Maximum Likelihood (ML) and Bayesian Inference (BI) analyses. The ML analysis was performed by PhyML v. 3.0 with a GTR site substitution model, including a gamma distributed rate heterogeneity and a proportion of invariant sites (Guindon et al. 2010). The BI analysis was performed using the bestfit evolutionary models for each partitioned locus estimated in MrModeltest v. 2.3 (Posada & Crandall 1998) following the Akaike Information Criterion (AIC), with a Markov Chain Monte Carlo (MCMC) algorithm in MrBayes v. 3.1.2 (Ronquist & Huelsenbeck 2003). Two MCMC chains were run from random trees for 10 M generations and stopped when average standard deviation of split frequencies fell below 0.01. Trees were saved each 1 000 generation. The first 25 % of trees were discarded as the burnin phase of each analysis and the posterior probabilities (BPP) were calculated to assess the remaining trees (Rannala & Yang 1996). The branch support from ML analyses were evaluated with a bootstrapping (BS) method of 1 000 replicates (Hillis & Bull 1993). Phylograms were shown using FigTree v. 1.3.1 (Rambaut & Drummond 2010). Novel sequences generated in the present study were deposited in GenBank (Table S1) and the aligned matrices used for phylogenetic analyses in TreeBASE (www. treebase.org; accession number: S31068).

# Morphology

Species descriptions were based on morphological features on host material where possible, and cultures. The macro-morphological descriptions on host tissues included size and arrangement of stromata; presence or absence of special structures such as the conceptacle and central column; number and diameter of ostioles per ectostromatic disc; shape and size of discs and number of locules. Micro-morphological observations included size and shape of conidiophores and conidia (asci and ascospores).

To induce sporulation, cultures were inoculated onto sterilized pine needles, twigs of *Salix* sp. or *Ulmus* sp. placed on the surface of CHA (cherry decoction agar; cherry extract 200 mL, water 800 mL, agar 15 g), MEA, OA (oatmeal agar; oatmeal extract 1 L, agar 15 g) and PDA, and incubated at 25 °C. A minimum of 10 conidiomata/ascomata, 20 conidiophores and conidiogenous cells/ asci (when possible) and 30 conidia/ascospores were measured for the isolates chosen to represent holotypes for each putative new species, and the ranges and averages were computed. Observations and measurements were recorded using Nikon SMZ1500 dissecting microscope and with a Nikon Eclipse Ni compound microscope, using a DS-Ri2 digital camera (Nikon,

Tokyo, Japan) and accompanying NIS-Elements imaging software v. 4.20. Colony morphology was described, and the colours determined using the Rayner (1970) colour charts.

# RESULTS

### **Phylogenetic analyses**

Phylogenetic analyses were performed based on individual gene datasets, and a combined dataset of the available ITS, *act1*, *rpb2*, *tef1-a* and *tub2* sequences (Figs S1–S5). The multi-gene analyses include 666 *Cytospora* ingroup strains with a total of 3 659 characters including gaps (677 characters for ITS, 422 for *act1*, 736 for *rpb2*, 992 for *tef1-a* and 832 for *tub2*), of which 1 584 characters are constant, 241 variable characters are parsimony-uninformative, and 1 834 characters are variable and parsimony-informative. The ITS, *act1*, *rpb2*, *tef1-a* and *tub2* individual analyses include 666, 539, 533, 535 and 493 *Cytospora* ingroup strains with a total of 673, 419, 732, 974 and 839 characters are constant, 62, 41, 27, 55 and 53 variable characters are parsimony-uninformative, and 256, 243, 296, 550 and 470 characters are variable and parsimony-informative, respectively.

In the ML analysis based on the combined gene dataset, the matrix had 2 655 distinct alignment patterns. Estimated base frequencies are as follows: A = 0.244484, C = 0.287439, G = 0.238641, T = 0.229436; substitution rates: AC = 1.301057, AG = 3.491193, AT = 1.438134, CG = 0.929235, CT = 6.320021, GT = 1.000000; gamma distribution shape parameter:  $\alpha$  = 0.458372. For BI analysis, the general time reversible model with inverse gamma rates (GTR + I + G) was determined to be the best for ITS, act1, rpb2 and tef1- $\alpha$  genes by MrModeltest, while the most appropriate model of the tub2 genes were Hasegawa-Kishino-Yano with inverse gamma rates model (HKY + I + G). The results of the Bayesian analyses were similar to the ML tree. The ML bootstrap support values (BS) equal to or above 60 % and the significant Bayesian posterior probabilities (BPP) equal to or above 0.80 are shown above the branches in Fig. 1. The phylogenetic analyses divide the genus Cytospora into 209 clades, of which 111 clades are relevant to the isolates in this study (Fig. 1).

# Morphology

Three asexual morphological groups and 13 asexual morphological types were designated as Group AI to AIII and type a1 to a13 (Fig. 2). Three sexual morphological groups and eight sexual morphological types were designated as Group SI to SIII and type s1 to s8 (Fig. 3). The types were characterised by the features of the conidiomata (the number of locules; presence or absence of black conceptacle, central column and beak; and locules subdivided by shared walls or individual walls) and ascomata (the number of ascospores per ascus; presence or absence of black conceptacle; the number of perithecia; and perithecia arranged each with its own ostiole or with a shared ostiole) *in vivo*.

# Asexual morphological groups and types (Fig. 2):

**Group AI** = single locule

**Type a1: unilocular** = single locule, conceptacle absent. Conidiomatal stromata not delimited by black marginal lines



**Fig. 1.** Phylogram of *Cytospora* resulting from a maximum likelihood analysis based on combined ITS, *act, rpb2, tef1-a*, and *tub2* genes. Numbers above the branches indicate ML bootstrap values (ML-BS  $\ge$  60%) and Bayesian Posterior Probabilities (BPP  $\ge$  0.8). Ex-type isolates are in bold. Reference strains are marked with\*. Isolates from the present study are marked in blue.

(conceptacle). Single locule with or without disc of ectostroma but lacking ring-like ectostroma encircling the ostiole.

**Type a2: leucounilocular** = single locule, conceptacle prominent.

Conidiomatal stromata delimited by black marginal lines (conceptacle). Single locule with or without disc of ectostroma but

lacking ring-like ectostroma encircling the ostiole.

**Type a3: cyclounilocular** = single locule with a central column, conceptacle absent.

Conidiomatal stromata not delimited by black marginal lines (conceptacle). Single locule with a central column, with or without disc of ectostroma.



Fig. 1. Continued).

**Type a4: rostrunilocular** = single locule, with a hard rostriform ostiole wall, conceptacle absent.

Conidiomatal stromata not delimited by black marginal lines (conceptacle). Single locule with a thorn-like ostiolar beak.

**Type a5: cytophomoid** = single locule with a wing-like ring of ectostroma, conceptacle absent.

Having an undivided locule and a distinctive ring of ectostroma that was wing-like in longitudinal median cross section encircling

the ostiole. Conidiomatal stromata not delimited by black marginal lines (conceptacle). Conidiomata of discrete simple undivided locules with ring-like ectromata encircling the ostiolar beak (wing-like in longitudinal median cross section).

Group All = multiple locules sharing common walls

**Type a6: labyrinthine cytosporoid** = multilocules sharing common walls, labyrinthine-like, conceptacle absent.

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#### Fig. 1. Continued).

Having a single locule subdivided by invaginations into several chambers, labyrinthine-like. Conidiomatal stromata not delimited by black marginal lines (conceptacle). Conidiomata of multichambered locules, subdivided by invaginations, sharing common walls. Single or multiple ostioles per disc.

Type a7: labyrinthine leucocytosporoid = multilocules sharing

common walls, labyrinthine-like, conceptacle prominent.

Having a single locule subdivided by invaginations into several chambers, labyrinthine-like. Conidiomatal stromata delimited by black marginal lines (conceptacle). Conidiomata of multichambered locules, subdivided by invaginations, sharing common walls. Single or multiple ostioles per disc.



Fig. 1. Continued).

**Type a8: cyclocytosporoid** = multilocules sharing common walls with a central column, conceptacle absent.

Having a single locule subdivided by invaginations into several chambers. Conidiomatal stromata not delimited by black marginal lines (conceptacle). Conidiomata of multi-chambered locules, subdivided by invaginations, sharing common walls, with a central column.

**Type a9: rosette cytosporoid** = multilocules sharing common walls, rosette-like, conceptacle absent.

Having a single locule subdivided by invaginations into several chambers, rosette-like. Conidiomatal stromata not delimited by black marginal lines (conceptacle). Conidiomata of multi-chambered locules, subdivided by invaginations, sharing common walls. Single or multiple ostioles per disc.



Fig. 1. Continued).

**Type a10: rosette leucocytosporoid** = multilocules sharing common walls, rosette-like, conceptacle prominent.

Having a single locule subdivided by invaginations into several chambers, rosette-like. Conidiomatal stromata delimited by black marginal lines (conceptacle). Conidiomata of multi-chambered locules, subdivided by invaginations, sharing common walls. Single or multiple ostioles per disc.

#### **Group AllI** = multiple locules with individual walls

**Type a11: lamyelloid** = multiple locules with separate walls and multiple ostioles, conceptacle absent.

Having groups of simple undivided locules. Locules not sharing common walls. Conidiomatal stromata not delimited by

black marginal lines (conceptacle). Ostioles multiple, converging independently to shared discs, or to surface.

**Type a12: torsellioid** = multiple locules with separate walls and a single shared ostiole, conceptacle absent.

Having groups of simple undivided locules. Locules not sharing common walls. Conidiomatal stromata not delimited by black marginal lines (conceptacle). Ostioles converging to a shared single ostiole.

**Type a13: leucotorsellioid** = multiple locules with separate walls and a single shared ostiole, conceptacle prominent.

Having groups of simple undivided locules. Locules not sharing common walls. Conidiomatal stromata delimited by black marginal lines (conceptacle). Ostioles converging to a shared single ostiole.





Fig. 2. Asexual morphological groups and types of Cytospora.

### Sexual morphological groups and types (Fig. 3):

#### Group SI = numerous ascospores per ascus

**Type s1: polysporous leucocircinate** = numerous ascospores per ascus; pseudostromata delimited by conceptacles; few large perithecia circinately arranged, each with its own ostiole.

**Type s2: polysporous leucocyprious** = numerous ascospores per ascus; pseudostromata delimited by conceptacles; few large perithecia circinately arranged with a single shared ostiole.

Group SII = eight ascospores per ascus

**Type s3: octosporous circinate** = eight ascospores per ascus; pseudostromata not delimited by conceptacles; few large perithecia circinately arranged, each with its own ostiole.

**Type s4: octosporous cyprious** = eight ascospores per ascus; pseudostromata not delimited by conceptacles; few large perithecia circinately arranged with a single shared ostiole.

**Type s5: octosporous leucocircinate** = eight ascospores per ascus; pseudostromata delimited by conceptacles; few large perithecia circinately arranged, each with its own ostiole.



Fig. 3. Sexual morphological groups and types of Cytospora.

**Type s6: octosporous leucocyprious** = eight ascospores per ascus; pseudostromata delimited by conceptacles; few large perithecia circinately arranged with a single shared ostiole.

**Type s7: octosporous monostichous** = eight ascospores per ascus; pseudostromata not delimited by conceptacles; numerous small perithecia upright to inclined crowded, each with its own ostiole.

#### Group SIII = four ascospores per ascus

**Type s8: tetrasporous circinate** = four ascospores per ascus; pseudostromata not delimited by conceptacles; few large perithecia circinately arranged, each with its own ostiole.

### TAXONOMY

*Cytosporaceae* Fr. [as 'Cytisporei'], Syst. Orb. Veg. (Lundae) 1: 118. 1825.

Synonym: Valsaceae Tul. & C. Tul. [as 'Valsarum'], Select. Fung. Carpol. (Paris) 1: 180. 1861.

Ascomata solitary to grouped, immersed in bark. Stromatic tissues delimited by a black marginal line (conceptacle) or not. Ectostromatic disc usually surrounded by tightly ostiolar necks. Perithecial



ascostroma inclined to upright, flask-shaped to spherical, arranged circularly or irregularly. *Paraphyses* present or deliquescent. *Asci* free floating, ellipsoid to clavate, apical ring chitinoid, refractive. *Ascospores* hyaline, elongate-allantoid, aseptate, thin-walled, biseriate to irregularly multiseriate, 4, 8 or more per ascus. *Conidiomata* stromatic or pycnidial, immersed in bark. *Stromatic tissues* delimited by a black marginal line (conceptacle) or not. *Ectostromatic disc* prominent or lacking, one to more ostioles per disc. *Locules* single, undivided to multiple chambered with invaginations. *Conidiogenous cells* enteroblastic, phialidic. *Conidia* hyaline, allantoid, aseptate, small and narrow.

#### Type genus: Cytospora Ehrenb., Sylv. Mycol. Berol. (Berlin): 28. 1818.

*Notes: Cytosporaceae* (1825) was treated as the synonym of *Valsaceae* (1861) prior to the currently accepted unitary nomenclature for fungi. Because *Cytospora* (as oldest name) has priority over *Valsa* and related genera, *Cytosporaceae* (again as oldest family name) has priority over *Valsaceae* (Fan *et al.* 2015a, b, Rossman *et al.* 2015). Several genera were in *Cytosporaceae* but many of them have since been separated into different families of *Diaporthales* (Wehmeyer 1975, Barr 1978, Castlebury *et al.* 2002, Senanayake *et al.* 2017, 2018). Senanayake *et al.* (2017,

2018) listed five genera (*Cytospora*, *Pachytrype*, *Paravalsa*, *Waydora* and *Xenotypa*) in *Cytosporaceae*, of which only *Cytospora*, *Pachytrype* and *Waydora* have DNA data available for phylogenetic comparison. The present study re-evaluated *Cytospora* and related genera, showing them to cluster in a well-supported monophyletic clade in *Cytosporaceae* (Fig. 1).

Cytospora Ehrenb., Sylv. Mycol. Berol. (Berlin): 28. 1818.

Synonyms: Valsa Fr., Syst. orb. veg. (Lundae) 1: 107. 1825; Typus: Valsa ambiens (Pers.) Fr. 1849.

*Valsella* Fuckel, Jb. nassau. Ver. Naturk. 23–24: 203. 1870; *Typus*: *Valsella salicis* Fuckel 1870.

Leucostoma (Nitschke) Höhn., Ber. dt. bot. Ges. 35: 637. 1917; Basionym: Valsa subgen. Leucostoma Nitschke 1870; Typus: Leucostoma massarianum (De Not.) Höhn. 1917.

Valseutypella Höhn., Ann. Mycol. 16(3/6): 224. 1919; *Typus:* Valseutypella tristicha (De Not.) Höhn. 1920.

Leucocytospora (Höhn.) Höhn., Mitt. bot. Inst. tech. Hochsch. Wien 4(2): 73. 1927; Basionym: Cytospora subgen. Leucocytospora Höhn. 1918; Typus: Leucocytospora corni (Westend.) Höhn. 1927.

Plant pathogenic, saprobic and endophytic, mostly causing canker and dieback diseases. Ascomata solitary to grouped, immersed in vascular plant tissues, slightly to strongly erumpent through the bark surface. Stromatic tissues prosenchymatous or pseudoparenchymatous, sometimes delimited by a black marginal line (conceptacle). Ectostromatic disc usually surrounded by tightly ostiolar necks. Perithecial ascomata inclined to upright, in valsoid or diatrypelloid configurations, immersed, usually embedded in ectostromatic disc, with beaks converging at surface. Ostioles numerous per disc, periphysate; walls of perithecia bilayered, narrow, outer layer of textura epidermoidea to textura angularis. Paraphyses present but usually deliquescent at maturity, often collapsed and broad. Asci free floating, narrow, ellipsoid to clavate, apical ring chitinoid, refractive. Ascospores hyaline, allantoid, aseptate, thin-walled, smooth, biseriate to irregularly multiseriate, 4, 8 or polysporous per ascus. Conidiomata stromatic or pycnidial, ostiolate, immersed in vascular plant tissues, slightly to strongly erumpent through the bark surface, sometimes delimited by a black marginal line (conceptacle). Ectostromatic disc prominent or lacking, one to few ostioles per disc. Locules single, undivided to multiple chambered with invaginations, globoid to flattened toroid, in ectostroma or embedded in entostroma, sometimes with a column; wall bilayered, outer layer prosenchymatous, ultimately sclerenchymatous. Conidiophores borne along the locules, hyaline, branched or not, thin-walled, normally embedded in a gelatinous layer. Conidiogenous cells enteroblastic, phialidic, subcylindrical to cylindrical, tapering towards apices. Conidia hyaline, allantoid, eguttulate, smooth, aseptate, thin-walled, relatively small and narrow (adapted from Adams et al. 2005, Fan et al. 2020).

*Cytospora abyssinica* G.C. Adams *et al.*, Stud. Mycol. 52: 79. 2005. (Fig. 1: Clade 182)

*Typus*: **Ethiopia**, Sidama Region, Wondo Genet, Forestry College Compound, on dead twig of *Eucalyptus saligna*, Sep. 2002, *A. Gezahgne* (**holotype** MSC 380700, ex-holotype culture CBS 116819 = CMW 10181).

Description: See Adams et al. (2005).

*Additional material examined*: **Ethiopia**, Sidama Region, Wondo Genet, Forestry College Compound, on dead twig of *Eucalyptus globulus*, 2002, *A. Gezahgne* (MSC 380702, culture CBS 117004).

*Notes*: *Cytospora abyssinica* has a black conceptacle around the conidiomata and multiple locules with separate walls and a single ostiole (Adams *et al.* 2005). Adams *et al.* (2005) described this species based on ITS sequences of three Ethiopian isolates, namely CBS 116819, 117004 and 117605 with CBS 116819 as ex-holotype culture. However, isolate CBS 117605 differs from isolate CBS 116819 in ITS (27/504), act1 (23/243), rpb2 (44/650) and *tef1-a* (68/536) genes and groups with the ex-holotype of *C. nitschkei.* Therefore, isolates CBS 116819 and 117004 are identified as *C. abyssinica* while isolate CBS 117605 is identified here as *C. nitschkei.* 

*Cytospora acericola* X.L. Fan & C.M. Tian, *sp. nov.* MycoBank MB 850139. Fig. 4. (Fig. 1: Clade 171)

Etymology: Named refers to the host genus Acer.

*Typus*: **China**, Shaanxi Province, Xi'an City, Chang'an District, Mount Wutai, 34°2'40.23"N, 108°53'46.21"E, on branches of *Acer davidii*, 18 Jun. 2021, *C. Peng & C.M. Tian* (**holotype** BJFC-S2106, ex-holotype culture CFCC 55994; **paratype** BJFC-S2107, ex-paratype culture CFCC 55995).

Description: Conidiomata Group All (type a6), scattered or serried, immersed in bark, erumpent through the bark surface, discoid, with multiple locules. Conceptacle absent. Ectostromatic disc amber, circular, (350–)400–480(–540) µm diam, with one ostioles per disc. Ostiole grey to black, (110–)130–220(–300) µm diam. Locules numerous, subdivided frequently with independent walls, (1 490–) 1 750–1 950(–2 190) µm diam. Conidiophores borne along the locules, hyaline, branched at the base, in the middle or occasionally unbranched, (14–)14.5–17.5(–21) × 1–2 µm, embedded in a gelatinous layer. Conidiogenous cells enteroblastic, phialidic, subcylindrical to cylindrical. Conidia hyaline, allantoid, thin-walled, eguttulate, aseptate, smooth, 2.5–3.5(–4) × 1–1.5 (av. = 3.1 ± 0.3 × 1.3 ± 0.2, n = 30) µm.

*Culture characteristics*: Colonies are white, growing fast and entirely covering the 9-cm-diam Petri dish after 4 d, with felty aerial mycelium, compact at the centre and sparse at the margins.

Additional material examined: **China**, Beijing City, Changping District, Liu County, Wangjiayuan Town, 40°10'23"N, 116°4'9"E, on branches of *Koelreuteria paniculata*, 24 Aug. 2022, *X.L. Fan & L. Lin* (BJFC-S1964, culture CFCC 58486).

*Notes: Cytospora acericola* is associated with canker disease of *Acer davidii* in China. It represents a unique *Cytospora* isolated from this host in China and can be identified by its multiple locules with independent walls and small conidia  $(2.5-3.5 \times 1-1.5 \mu m)$ . Additionally, it is revealed in the multi-gene phylogram as a distinct clade with full support (ML/BI = 100/1). *Cytospora acericola* forms a group close to *C. platycladi* but differs in the conidial length (2.5-3.5 vs 4.5-5 µm in the latter) (Fan *et al.* 2020). Phylogenetically, *C. acericola* (CFCC 55994) differs from *C. platycladi* (CFCC 50504) in ITS (1/503), *act1* (24/239), *rpb2* (8/726), *tef1-a* (13/505) and *tub2* (13/433) genes.

Cytospora adamsii Jami, Crous & M.J. Wingf., sp. nov. MycoBank MB 850140. Fig. 5. (Fig. 1: Clade 92)

*Etymology*: Named in honour of the United States mycologist Gerard C. Adams, in recognition of his contributions to *Cytospora* systematics.



Fig. 4. Cytospora acericola (BJFC-S2106). A, B. Habit of conidiomata on twig. C. Transverse section through conidioma. D. Longitudinal section through conidioma. E. Conidiophores and conidiogenous cells. F. Conidia. Scale bars: A = 1 mm; B–D = 500 μm; E, F = 10 μm.

*Typus*: **Unknown locality**, on *Pinus sylvestris*, unknown date and *collector* (**holotype** CBS H-22972, ex-holotype culture CBS 179.70).

Description: Conidiomata produced on OA in 2–4 wk, globose, stromata black, up to 400  $\mu$ m diam, semi-immersed, unilocular, exuding a creamy conidial mass. Conidiophores hyaline, smooth, subcylindrical, 0–2-septate, acropleurogenous, branched with terminal and lateral conidiogenous cells. Conidiogenous cells embedded in a continuous gelatinous matrix, phialidic, subcylindrical, tapering towards the apex, collarettes minute, 3–9 × 1.5–2  $\mu$ m. Conidia hyaline, eguttulate, allantoid, aseptate, apex subobtuse (4–)4.5–5.5(–7) × 1–2.5  $\mu$ m.

*Culture characteristics*: Colonies covering the Petri dish after 10 d at 24 °C, with aerial mycelium flat, moderately sparse. On MEA grey on the surface and vinaceous in reverse.

*Notes*: The isolate CBS 179.70 was identified as *C. friesii* (= *C. pinastri*) by Adams *et al.* (2005), but it was not closely related to *C. pinastri* in the present study based on multigene sequences.

The isolates CBS 179.70 and CFCC 50493 (the ex-holotype of *C. beilinensis*) form a clade with high support (ML/BI = 99/1) but differs by ITS (7/507 bp) and *tub2* (41/454 bp) sequences. Despite the inability to evaluate the sequence differences of the *act1*, *rpb2*, and *tef1-a* genes due to a lack of data, we propose that the isolate CBS 179.70 is introduced as a new species, *C. adamsii*. Additional fresh specimens need to be collected to improve the phylogenetic placement and clarify the geographical distribution of *C. adamsii*.

*Cytospora ailanthicola* X.L. Fan & C.M. Tian, Persoonia 45: 13. 2020. (Fig. 1: Clade 53)

*Typus*: **China**, Ningxia Province, Zhongwei City, Zhongning County, Qukou, on branches of *Ailanthus altissima*, 3 Jun. 2012, *X.L. Fan* (**holotype** BJFC-S550, ex-holotype culture CFCC 89970).

Description: See Fan et al. (2020).

Materials examined: China, Beijing City, Mentougou District, Baihuashan National Nature Reserve, 39°50'31"N, 115°26'52"E, on twigs and branches of *Populus beijingensis*, 22 Aug. 2018, H.Y. Zhu & X.L. Fan



Fig. 5. Cytospora adamsii (CBS 179.70). A. Conidiomata on MEA. B, C. Conidiophores and conidiogenous cells. D. Conidia. Scale bars: A = 100 μm; B, C = 10 μm; D = 5 μm.



(BJFC CF20191225, culture CFCC 54066); Haidian District, Beijing Botanical Garden, 116°12'29.4"E, 40°0'10.72"N, on branches of Populus sp., 18 Aug. 2017, H.Y. Zhu (BJFC CF20191233, culture CFCC 54069); Yanging District, on twigs and branches of Salix sp., 8 Jul. 2022, Y.K. Bai & X.L. Fan (BJFC-S1976, culture CFCC 59073; BJFC-S1975, culture CFCC 59068; BJFC-S1974, culture CFCC 59060; BJFC-S1977, culture CFCC 59080; BJFC-S1973, culture CFCC 59057); Ningxia Hui Autonomous Region, Zhongwei City, Shapotou National Nature Reserve, on branches of Populus simonii, 3 Jun. 2012, X.L. Fan (BJFC CF20191211, culture CFCC 54062); Qinghai Province, Haidong City, Huzhu Tu Autonomous County, on branches of Populus alba var. pyramidalis, 8 Aug. 2012, X.L. Fan (BJFC CF20191243, culture CFCC 54074); Shaanxi Province. Xianyang City, Wugong county, on branches of Populus sp., unknown date and collector (BJFC CF20191206, culture CFCC 54059); Tibet Autonomous Region, Shannan City, Jiacha County, on branches of Populus sp., unknown date, X.L. Jiang (BJFC CF20191213, culture CFCC 54063, CFCC 54064); Yunnan Province, Kunming City, Panlong District, Southwest Forestry University, 102°45'22"E, 25°3'43.8"N, on branches of Populus × canadensis, L. Lin & Z.Q. Wu, 11 Aug. 2022 (BJFC-S1968, culture CFCC 58226; (BJFC-S1971, culture CFCC 58237; BJFC-S1972, culture CFCC 58238); ibid., Dali City, Xiaguan District, 25°38'49"N, 100°10'8"E, on branches of Populus yunnanensis, 10 Jul. 2022, L. Lin & M. Lin (BJFC-S1969, culture CFCC 58229); ibid., 25°39'9"N, 100°10'50"E, on branches of Populus × canadensis, 10 Jul. 2022, L. Lin & M. Lin (BJFC-S1970, culture CFCC 58230).

Notes: Cytospora ailanthicola was described by Fan et al. (2020) from Ailanthus altissima in China. It has a labyrinthine cytosporoid conidiomata without a black conceptacle (Group All, type a6) (Fan et al. 2020). Lin et al. (2023a) tested the pathogenicity of C. ailanthicola on Populus alba var. pyramidalis. Cytospora ailanthicola has been recorded to cause canker disease on Populus spp. in many provincial administrative regions of China, *i.e.*, Beijing, Ningxia, Qinghai, Shaanxi, Tibet and Yunnan (Fan et al. 2020, Lin et al. 2023a). The currently available 17 isolates and CFCC 89970 (the ex-holotype of C. ailanthicola) form a distinct clade with maximum support (ML/BI = 100/1) and have the following nucleotide differences from the sequences for C. salicacearum, C. melnikii and C. tritici: in ITS: 1/502 bp, 1/501 bp and 0/501 bp, respectively; in act: 6/195 bp, 2/224 bp and 10/225 bp, respectively; in *rpb2*: 10/726 bp, 9/726 bp and 9/650 bp, respectively; in *tef1-a*: no tef1- $\alpha$  available for C. salicacearum, 23/492 bp and 19/492 bp, respectively; in tub2: no tub2 available for C. salicacearum, 14/415 bp and 11/415 bp, respectively.

*Cytospora alba* L. Lin & X.L. Fan, J. Fungi 8 (4, no. 377): 6. 2022. (Fig. 1: Clade 101)

*Typus*: **China**, Gansu Province, Lanzhou City, Yongdeng County, on branches of *Salix matsudana*, 20 Oct. 2020, *X.L. Fan, N. Jiang & C. Peng* (holotype BJFC CF20201001, ex-holotype culture CFCC 55462; paratype BJFC CF20201011, ex-paratype culture CFCC 55463).

Description: See Lin et al. (2022).

*Materials examined*: **China**, Yunnan Province, Diqing Tibetan Autonomous Prefecture, Shangri-La City, Sanba Naxi Town, East Ring Road, 27°34'26''N, 100°1'5''E, on branches of *Populus adenopoda*, 9 Aug. 2022, *L. Lin & M. Lin* (BJFC-S1978, culture CFCC 58253; BJFC-S1979, culture CFCC 58254; BJFC-S1980, culture CFCC 58259); *ibid.*, Deqin County, Shengping Town, 28°29'0''N, 98°55'1''E, on branches of *Yulania denudata*, 9 Aug. 2022, *L. Lin & M. Lin* (BJFC-S1981, culture CFCC 58472); *ibid.*, 28°28'51''N, 98°55'1''E, on branches of *Populus haoana*, 9 Aug. 2022, *L. Lin & M. Lin* (BJFC-S1982, culture CFCC 58479; BJFC-S1983, culture CFCC 58480); Gansu Province, Lanzhou City, Yuzhong County, Guantangou, 35°53'17''N, 103°56'35''E, on branches of *Populus simonii*, 10 Jul. 2022, *L. Lin & X.H. Wang* (BJFC-S1984, culture CFCC 58261); *ibid.*, 35°53'36"N, 103°56'24"E, on branches of *Populus simonii*, 10 Jul. 2022, *L. Lin & X.H. Wang* (BJFC-S1985, culture CFCC 58262); *ibid.*, 35°53'17"N, 103°56'35"E, on branches of *Populus* sp., 10 Jul. 2022, *L. Lin & X.H. Wang* (BJFC-S1986, culture CFCC 59079).

*Notes*: Lin *et al.* (2022) described *C. alba* in Gansu, China and confirmed its pathogenicity to *Salix matsudana*. It can be recognised by the white ectostromatic disc and the large conidia (av. =  $8.2 \times 0.7 \mu$ m; Lin *et al.* 2022). The present study expands its host range to include *Yulania denudata* and *Populus* spp. The geographical distribution is now extended to Yunnan Province, China.

*Cytospora albodisca* X.L. Fan & C.M. Tian, Front. Plant Sci. 12 (no. 636460): 3. 2021. (Fig. 1: Clade 118)

*Typus*: **China**, Beijing City, Mentougou District, Mount Dongling, Xiaolongmen Forestry Centre, on dead branches of *Platycladus orientalis*, 17 Aug. 2017, *H.Y. Zhu & X.L. Fan* (**holotype** BJFC CF2019908, **isotype** BJM 240516, ex-holotype culture CFCC 53161).

Description: See Pan et al. (2021).

*Materials examined*: **China**, Beijing City, Mentougou District, Xiaolongmen Forestry Centre, 39°50'8"N, 115°34'35"E, on twigs and branches of *Ulmus macrocarpa*, 24 Aug. 2022, *L. Lin & X.L. Fan* (BJFC-S1987, culture CFCC 58440); *ibid.*, Yanqing District, Duck Lake Wetland Park, 40°24'56.88"N, 115°49'55.9"E, on twigs and branches of *Ulmus pumila* 'Jinye', 3 Jul. 2022, *Y.K. Bai & X.L. Fan* (BJFC-S1988, culture CFCC 59077).

*Notes: Cytospora albodisca* was described by Pan *et al.* (2021) from *Platycladus orientalis* in Beijing, China, which clusters in a sister phylogenetic clade with *C. corylina*. However, *Cytospora albodisca* (CFCC 53161) differs from *C. corylina* (CFCC 54684) in ITS (0/502), *act* (4/236), *rpb2* (10/726), *tef1-a* (23/488) and *tub2* (7/325) genes. The specimens BJFC-S1987 and BJFC-S1988 have octosporous leucocyprious ascomata (Group SII, type s6) with white discs, which is consistent with the description of Pan *et al.* (2021). Its host range is extended here to include *Ulmus pumila* and *Ulmus macrocarpa*. Neither Pan *et al.* (2021) nor this study observed asexual conidiomata on the host and for the present, it is known only based on the sexual morph.

*Cytospora annulata* Ellis & Everh., Proc. Acad. Nat. Sci. Philad. 45: 160. 1893. (Fig. 1: Clade 24)

Synonym: Valsa ambiens subsp. leucostomoides (Peck) Spielman, Canad. J. Bot. 63(8): 1361. 1985.

*Typus*: **USA**, Brookings, on dead branches of *Acer negundo*, Oct. 1871, *T.A. Williams* (N. Am. Fung. 2770; not examined).

Description: See Spielman (1985).

*Materials examined*: **USA**, New Jersey, on *Acer rubrum*, unknown date, *L. Spielman* (culture CBS 116809 = ATCC 52279 = CUP 06952279132); Illinois, on *Acer negundo*, unknown date, *L. Spielman* (culture CBS 116810 = ATCC 52282 = CUP 60135); New York, on *Acer rubrum*, unknown date and *collector* (culture CBS 118089 = ATCC 52280 = CUP 60133).

*Notes: Cytospora annulata* has a cytosporoid conidiomata (Group AII, type a6) and octosporous monostichous ascomata (Group SII, type s7) with white or grey discs (Spielman 1985). Isolate CBS 116810 = CUP 60135 was identified as *C. annulata* by Spielman (1985), who linked the asexual morph *C. annulata* to *V. ambiens* 

subsp. *leucostomoides*. Consequently, the name *C. annulata* is applied to CBS 118089 that was previously identified as *V. ambiens* subsp. *leucostomoides*. The DNA phylogenies place *C. annulata* in a distinct clade (ML/BI = 100/1) within a larger clade (ML/BI = 86/1) that includes *C. crataegina* and *C. malvicolor* (Fig. 1). It has the following nucleotide differences from the sequences for *C. crataegina* and *C. malvicolor*: in ITS: 43/492 bp and 45/489 bp, respectively; in *act1*: 15/244 bp and 19/245 bp, respectively; in *rpb2*: 32/650 bp and 29/650 bp, respectively; in *tef 1-a*: 35/482 bp and 38/453 bp, respectively; in *tub2*: 22/397 bp and 18/410 bp, respectively.

*Cytospora atrocirrhata* Gvrit., Mikol. Fitopatol. 7: 547. 1973. (Fig. 1: Clade 156)

Typus: Georgia, on branch of Salix sp. (not examined).

Description: See Fan et al. (2015a).

*Material examined*: **China**, Yunnan Province, Diqing Tibetan Autonomous Prefecture, Shangri-La City, Sanba Naxi Town, East Ring Road, 27°34'26"N, 100°1'5"E, on branches of *Populus adenopoda*, 9 Aug. 2022, *L. Lin & M. Lin* (BJFC-S1989, culture CFCC 59056).

Notes: Cytospora atrocirrhata was described on the branches of Salix in Georgia (Gvritishvili 1973). Its hosts include Salix, Populus and Juglans (Fan et al. 2020). An ex-holotype isolate for *C. atrocirrhata* does not exist and epitypification will be required to resolve its taxonomy. *Cytospora atrocirrhata* is sister to *C. tenebrica* and *C. yuduensis* (Fig. 1). However, *C. atrocirrhata* (CFCC 89615) differs from *C. tenebrica* (CFCC 56269) in ITS (36/527), act (68/234), rpb2 (33/487), tef1- $\alpha$  (135/503) and tub2 (97/410) genes and from *C. yuduensis* (CFCC 57539) in ITS (35/523), act (75/246), rpb2 (52/726), tef1- $\alpha$  (114/320) and tub2 (95/413) genes. They are morphologically similar in the undivided single-locule surrounded by a black conceptacle. However, *C. atrocirrhata* can be distinguished from the other two species by a slightly red pigment secreted into the PDA medium.

*Cytospora auerswaldii* (Nitschke) L. Lin & X.L. Fan, *comb. nov.* MycoBank MB 850141. (Fig. 1: Clade 15)

*Basionym: Valsa auerswaldii* Nitschke, Pyrenomyc. Germ. 2: 225. 1870.

Synonym: Leucostoma auerswaldii (Nitschke) Höhn., Mitt. Bot. Inst. Tech. Hochsch. Wien 5: 55. 1928.

*Typus*: In need of lecto- and epitypification; syntypes: **Germany**, Aulhausen, on *Fagus sylvatica*, L. Fuckel, Fungi Rhen. 1980; bei Östrich, on *Rhamnus frangula*, L. Fuckel Fungi Rhen. 603; Westfalen, Jägerhäuschen I, on *Malus*, Apr. 1867, *T. Nitschke* (B); Westfalen, Jägerhäuschen II, on *Malus*, *T. Nitschke* (B); Westfalen, near Erdmanns, on *Betula*, *T. Nitschke* (B).

Description: See Nitschke (1870), Ellis & Ellis (1997).

*Culture characteristics*: Colonies covering the Petri dish after 10 d at 24 °C, with aerial mycelium flat, sparse to moderate. On MEA surface honey and fawn on the reverse.

*Material examined*: **Russia**, unknown host and date, N.A. Naumov (CBS H-22971 dried culture, culture CBS 153.29).

*Notes*: The identity of the culture CBS 153.29, for which no data on the host are available, remains somewhat tentative, as the typification of the species (and therefore the species concept) has not yet been resolved. When describing the species, Nitschke (1870) did not designate a type, but cited the specimen Fuckel, Fungi Rhen. 1980, from *Fagus*, in the protologue next to the

species name, indicating that he considered his new species to be primarily based on this collection; however, he subsequently also cited the exsiccatum Fuckel, Fungi Rhen. 603 (from Rhamnus frangula) and his own collections from two localities (Erdmanns, from Betula; Jägerhäuschen, from Malus). In their publication on the type collections of Nitschke extant in Herbarium B, Gerhardt & Hein (1980) listed four syntypes (Fungi Rhen. 1980 and three collections from Erdmanns and Jägerhäuschen; see above); however, all other extant copies of Fungi Rhen. 603 and 1980 in other herbaria are syntypes as well, and therefore qualify for lectotypification. To stabilise the species concept and circumscription, an appropriate lectotype needs to be chosen, which should be epitypified with a recent collection from the same host as the lectotype. However, as no collection and culture matching the protologue and type hosts was available for sequencing, we currently refrain from lecto- and epitypification.

Ellis & Ellis (1997) re-described Valsa auerswaldii with 2-8 perithecia per pseudostroma with black conceptacle, 8-spores per ascus, saprobic, on dead branch. Adams et al. (2005) provided ITS sequence data of the V. auerswaldii isolate CBS 153.29 and recorded that this species had a leucotorsellioid type of conidioma. In the present study, the name V. auerswaldii is combined in Cytospora. It clusters in a sister phylogenetic clade with C. ginghaiensis and C. tanaitica (Fig. 1). Cytospora auerswaldii (CBS 153.29) differs from C. qinghaiensis (CFCC 50026) in ITS (1/506 bp), act1 (3/241 bp), rpb2 (23/650 bp), tef1-α (3/481 bp) and tub2 (0/412 bp) genes and from C. tanaitica (MFLUCC 141057) in ITS (0/494 bp), act1 (15/226 bp); no *rpb2*, *tef1-\alpha* and *tub2* genes available for *C*. *tanaitica*. We have not treated these three species as conspecific, even though their sequences are similar, because the support value of the combined large branch is inordinately low (ML/BI = 54/-). The current classification is tentative, and further study of type materials, as well as collections of additional fresh specimens, are required.

Cytospora austromontana G.C. Adams & M.J. Wingf., Stud. Mycol. 52: 114. 2005. (Fig. 1: Clade 195)

*Typus*: **Australia**, New South Wales, Perisher Valley, on dead cankered branch of *Eucalyptus pauciflora*, 2001, *M.J. Wingfield* (**holotype** MSC 380693, ex-holotype culture CBS 116820).

Description: See Adams et al. (2005).

*Material examined*: **Australia**, New South Wales, Perisher Valley, on dead cankered branch of *Eucalyptus pauciflora*, 2001, *M.J. Wingfield* (culture CBS 116821).

Notes: Cytospora austromontana was described from dead cankered branches on *Eucalyptus pauciflora* in Australia (Adams *et al.* 2005). It has conidiomata in Group AII (type a7), which can be produced on dead branches of *Eucalyptus* and autoclaved *Eucalyptus* leaves (Adams *et al.* 2005). The sexual morph of this species has not been observed. In ITS, *act1, rpb2, tef1-a* and *tub2* gene trees, the ML and BI phylogenies all place it as a distinct clade.

*Cytospora beijingensis* L. Lin & X.L. Fan, *sp. nov.* MycoBank MB 850142. Fig. 6. (Fig. 1: Clade 104)

Etymology: Name refers to Beijing City where it was collected.

*Typus*: **China**, Beijing City, Huairou District, Forestry Centre of Beijing University of Agriculture, 40°57'56.65"N, 116°29'44.59"E, on branches





**Fig. 6.** *Cytospora beijingensis* (BJFC-S2182). **A, B.** Habit of pseudostromata on twig. **C.** Transverse section through pseudostroma with ascomata. **D.** Longitudinal sections through pseudostromata with ascomata. **E.** Asci. **F.** Ascospores. Scale bars: A = 1 mm; B = 250 μm; C–D = 500 μm; E–F = 10 μm.

of *Corylus heterophylla*, 18 Jun. 2021, *X.L. Fan* & *Y.K. Bai* (holotype BJFC-S2182, ex-holotype culture CFCC 55836); ex-isotype culture CFCC 55835; *ibid.*, Yanqing District, Songshan National Nature Reserve, 40°29'33.28"N, 115°46'48.96"E, on twigs and branches of *Corylus heterophylla*, 4 Aug. 2021, *Y.K. Bai* (paratype BJFC-S2183, ex-paratype culture CFCC 56705).

Description: Pseudostromata with ascomata Group SII (type s5), immersed in the bark, erumpent through the bark surface, scattered, 950–1 170(–1 310) µm diam, with 4–6 perithecia arranged circularly or irregularly. Conceptacle present. Ectostromatic disc grey to isabelline, usually surrounded by tightly ostiolar necks, irregularly, (220–)243–340(–356) µm diam, with 4–6 ostioles regularly arranged in disc. Ostioles dark mouse grey to black, at the above level as the disc, concentrated, 75–115(–122) µm diam. Perithecia dark grey to black, flask-shaped to spherical, arranged circularly, (205–)265–380(–450) µm diam. Asci free, clavate to elongate-obovoid, 8-spored, (24–)31.5–39(–40) × (7.5–)10.5–12.5(–13) µm. Ascospores hyaline, aseptate, elongate-allantoid, thin-walled, (8–)10–13(–14) × 2–3 (av. =  $11.5 \pm 1.3 \times 2.5 \pm 0.3$ , n = 30) µm. Asexual morph not observed.

*Culture characteristics*: Colonies on PDA are initially white, becoming olivaceous buff above, growing fast, entirely covering the 9-cm-diam Petri dish after 3 d, flat, with a uniform texture. Sterile.

Additional materials examined: China, Beijing City, Yanqing District, Songshan National Nature Reserve, 40°29'48.11"N, 115°46'52.63"E, on twigs and branches of *Quercus mongolica*, 4 Aug. 2021, *X.L. Fan & Y.K. Bai* (BJFC-S2184, culture CFCC 56684); *ibid.*, Mentougou District, Xiaolongmen Forestry Centre, 39°57'61"N, 115°25'62"E, on twigs and branches of *Populus* × *beijingensis*, 25 Aug. 2022, *L. Lin & X.L. Fan* (BJFC-S1992, culture CFCC 58263).

Notes: Cytospora beijingensis was associated with canker disease of Corylus heterophylla and Quercus mongolica. The phylogenetic analysis showed it as a distinct clade (ML/BI = 100/1) within a

larger clade (ML/BI = 100/1) containing *C. kunsensis*. However, *Cytospora beijingensis* (CFCC 55836) has the following nucleotide differences from the sequences for *C. kunsensis* (CFCC 59570): in ITS: 1/509 bp, *act*: 17/227 bp, *rpb2*: 1/499 bp, *tef1-a*: 22/282 bp, and in *tub2*: 4/402 bp. Morphologically, it can be distinguished from *C. kunsensi* by different size of asci (31.5–39 × 10.5–12.5 µm vs 45–70 × 8.5–10.5 µm) and ascospores (10–13 × 2–3 µm vs 7–8 × 2–2.5 µm). The sexual morph of this species is leucostoma-like in its leucostomoid stromata and ascomata, delimited by a dark conceptacle (Adams *et al.* 2005).

Cytospora berkeleyi G.C. Adams, Stud. Mycol. 52: 118. 2005. (Fig. 1: Clade 192)

*Typus*: **USA**, California, Palo Alto campus of Stanford University, on dead cankered branches of *Eucalyptus globulus*, 2001, *G.C. Adams* (**holotype** MSC 380710, ex-holotype culture CBS 116823).

Description: See Adams et al. (2005).

Additional materials examined: **USA**, California, Palo Alto campus of Stanford University, on dead cankered branches of *Eucalyptus globulus*, 2001, *G.C. Adams* (MSC 380711, culture CBS 117005); *ibid.*, Berkeley campus of University of California, on dead twigs of *E. globulus*, 8 Jun. 2001, *G.C. Adams & H. Hallen* (MSC 380709, culture CBS 116824). **New Zealand**, on stem and branches canker of *Castanea sativa*, unknown date, *M. Braithwaite* (culture CBS 101714).

*Notes*: *Cytospora berkeleyi* was described from dead cankered branches of *Eucalyptus globulus* in the USA (Adams *et al.* 2005), and clusters in a sister clade with *C. diatrypelloidea*. However, *C. berkeleyi* (CBS 117005) differs from *C. diatrypelloidea* (CBS 120062) in ITS (0/502), *act* (0/243), *rpb2* (18/650), *tef1-a* (16/509) and *tub2* (5/415) loci. Conidiomata of *C. berkeleyi* can have both a cytosporoid rosette and torsellioid features (Adams *et al.* 2005). Currently, this species has been found in the USA and New Zealand on *Eucalyptus* spp. and *Castanea sativa*.



**Fig. 7.** *Cytospora betulae* (CBS 141622). **A.** Exuding conidial mass from conidioma on MEA. **B.** Conidiomata on MEA. **C–F.** Conidiophores and conidiogenous cells. **G.** Conidia. Scale bars: B = 100 μm; C–F = 10 μm; G = 5 μm.

Cytospora betulae Jami, Crous & M.J. Wingf., sp. nov. MycoBank MB 850143. Fig. 7. (Fig. 1: Clade 13)

Etymology: Name refers to the host genus, Betula.

*Typus*: **USA**, New York, Syracuse, Oakwood cemetery, on twig of *Betula* papyrifera, 6 Jun. 2007, *L.C. Mejía* LCM109 (**holotype** BPI 881503, ex-holotype culture CBS 141622 = CPC 28412 = LCM 109.01; **isotype** CBS H-22936, dried culture).

Description: Conidiomata produced on MEA after 3–5 wk; stromata globose to irregular, black, up to 300  $\mu$ m diam, semi-immersed, multilocular, exuding an orange conidial mass. Conidiophores hyaline, smooth, subcylindrical, 0–2-septate, acropleurogenous, branched at the base. Conidiogenous cells embedded in a continuous gelatinous matrix, phialidic, with periclinal thickening, subcylindrical, tapering towards the apex, collarettes minute, 5–12 × 1–1.5  $\mu$ m. Paraphyses intermingled among conidiophores, hyaline, smooth, branched, aseptate, up to 20  $\mu$ m long, 1–1.5  $\mu$ m diam, at times developing fertile terminal conidiogenous cells. Conidia hyaline, eguttulate, allantoid, aseptate, apex subobtuse, 3.5–5 × 1–1.5  $\mu$ m.

*Culture characteristics*: Colonies covering the Petri dish after 10 d at 24 °C, with aerial mycelium flat, sparse to moderate. On MEA buff colour on the surface and reverse honey.

Notes: Cytospora betulae is described from twigs of Betula papyrifera in USA, and clusters in a strongly supported clade with Cytospora washingtonensis (CBS 141616, CBS 141619 and CBS 141620). However, Cytospora betulae (CBS 141622) differs from C. washingtonensis (CBS 141619) in ITS (2/506), act1 (7/246), rpb2 (6/657), tef1- $\alpha$  (16/495) and tub2 (3/340) genes. Morphologically, C. betulae has smaller conidia (3.5–5 × 1–1.5 µm), compared to conidia of C. washingtonensis (4–7 × 1–2 µm).

Cytospora brabeji Jami, Crous & M.J. Wingf., sp. nov. MycoBank MB 850144. Fig. 8. (Fig. 1: Clade 202)

Etymology: Name refers to the host genus, Brabejum.

*Typus*: **South Africa**, Western Cape Province, on twig of *Brabejum stellatifolium*, 2000, *S. Marincowitz* (**holotype** PREM 59524, ex-holotype culture CBS 119207 = CMW 20020).

Description: Conidiomata produced on twigs on MEA after 2–4 wk, stromata multilocular, convoluted, semi-immersed, globose to irregular, black, up to 1 mm diam, exuding white to a pale luteous conidial cirrhi or conidial masses. Conidiophores hyaline, smooth, branched, 0–3-septate, 6–30 × 1–2 µm, embedded in a gelatinous layer. Conidiogenous cells phialidic, with periclinal thickening, subcylindrical, tapering towards apices, collarettes minute, 7–12 × 0.9–2 µm; arranged in rosettes of up to five, or lateral along branching conidiophores with terminal stipe extensions



Fig. 8. Cytospora brabeji (CBS 119207). A. Conidiomata on MEA. B. Tangential section of conidiomata. C, D. Conidiophores and conidiogenous cells. E. Conidia. Scale bars: A, B = 100 μm; C, D = 10 μm; E = 5 μm.



that resemble paraphyses, but at times also fertile. *Paraphyses* intermingled among conidiophores, hyaline, smooth, branched, septate, up to 40  $\mu$ m long, 1.5–2  $\mu$ m diam, at times developing fertile lateral cells. *Conidia* hyaline, smooth, guttulate, allantoid or straight, apex rounded, aseptate, 3.5–5.5 × 0.9–2  $\mu$ m.

*Culture characteristics*: Colonies covering the Petri dish after 10 d at 24 °C, with aerial mycelium flat, moderately sparse. On MEA grey colour on the surface and reverse dark grey.

*Notes*: Isolate CBS 119207 was identified as *C. pruinosa* based on ITS sequence data (Marincowitz *et al.* 2008). However, in the present study it groups separately based on multigene data and a new name, *Cytospora brabeji*, is therefore introduced to accommodate it. In the ITS, *act1*, *rpb2*, *tef1-α* and *tub2* gene trees, the BI and ML phylogenies all place it as a distinct clade.

*Cytospora californica* D.P. Lawr. *et al.*, IMA Fungus 9: 349. 2018. Fig. 9. (Fig. 1: Clade 186)

*Typus*: **USA**, California, Lake County, from wood canker of *Juglans regia*, 14 Mar. 2014, *T.J. Michailides* (**holotype** BPI 910651, ex-holotype culture CBS 144234).

Description: Conidiomata produced on MEA after 2–4 wk, solitary, globose, black, immersed, unilocular, 100–300 µm diam, exuding a creamy conidial mass. Conidiophores hyaline, smooth, subcylindrical, 1–3-septate, branched with terminal and lateral conidiogenous cells, 15–35 × 1.5–3 µm. Conidiogenous cells hyaline, smooth, embedded in a continuous gelatinous matrix, 4–12 × 1.5–2 µm, phialidic, subcylindrical, tapering towards the apex, 1 µm diam, with flaring collarette, 1–2 µm long. Conidia hyaline, eguttulate, allantoid, apex subobtuse, base truncate, 0.5–1 µm diam, aseptate, (5–)5.5–6.5(–7) × (1–)1.5(–2) µm.

*Culture characteristics*: Colonies covering the Petri dish after 10 d at 24 °C, with aerial mycelium flat, moderately sparse. On MEA buff coloured on both surface and reverse with white tufts and patches on the colony surface.

*Material examined*: **USA**, Washington, U.S. Route 101, near the Lake Crescent, on *Rubus* sp., unknown date and *collector* (culture CBS 141612 = CPC 28373 = DMW 198.1).

*Notes*: *Cytospora californica* was described from cankers on *Juglans regia* in California by Lawrence *et al.* (2018). It clusters closely with *C. eucalypti* but can be distinguished by its shorter conidia on PDA ( $4.5-5.5 \times 1.2-1.6 \mu m vs 5.4-6.5 \times 1.2-1.6 \mu m$ ) and slower growth rate (58.8 mm vs 85 mm in 7 d) (Lawrence *et al.* 2018). Phylogenetically, *C. californica* (CBS 141612) differs from *C. eucalypti* (CBS 116815) in ITS (3/488), *act1* (16/242), *rpb2* (19/650) and *tef1-a* (46/547) loci (no *tub2* available for *C. californica*). The present study adds morphological descriptions on MEA, which shows that conidial size in this species varies on different media.

*Cytospora castaneicola* L. Lin & X.L. Fan, *sp. nov.* MycoBank MB 850145. Fig. 10. (Fig. 1: Clade 203)

*Etymology*: Name refers to the host genus, *Castanea*.

*Typus*: **China**, Shaanxi Province, Ankang City, Xiangxidong Forest Park, 32°40'33"N, 109°18'57"E, on stem bark of *Castanea mollissima*, 1 Jul. 2017, *N. Jiang* (**holotype** BJFC-S1704, ex-holotype culture CFCC 52454; **paratype** BJFC-S1705, ex-paratype culture CFCC 52455).

Description: Conidiomata Group AIII (type a11), pycnidial, immersed in bark, erumpent through the bark surface, discoid to conical, with multiple locules. Conceptacle absent. Ectostromatic disc black, with one ostiole per disc. Ostiole inconspicuous. Locules multiple, arranged irregularly with individual walls. Conidiophores borne along the locules, hyaline, unbranched or occasionally branched at the base, reduced to conidiogenous cells, 9.5–16.5 × 1–2 (av. =  $15.3 \pm 1.5 \times 1.5 \pm 0.2$ , n = 30) µm. Conidia hyaline, allantoid, thinwalled, eguttulate, aseptate, smooth,  $3.5-5.5 \times 1-1.5$  (av. =  $4.6 \pm 0.5 \times 1.3 \pm 0.1$ , n = 50) µm.

Culture characteristics: See Jiang et al. (2020b, as C. myrtagena).

*Notes*: Jiang *et al.* (2020b) provisionally treated isolates CFCC 52454 and 52455 as *C. myrtagena*. Although the isolates CFCC 52454 and 52455 from *Castanea mollissima* are closely related to *C. myrtagena* and *C. tibouchinae*, they differ in 22 bp from *C. myrtagena* (CBS 116843) and 6 bp from *C. tibouchinae* (CBS 141324) in ITS. In this study, we therefore treat this clade as a new species, *C. castaneicola*.

*Cytospora ceratosperma* (Tode) G.C. Adams & Rossman, IMA Fungus 6: 147. 2015. Figs 11, 12. (Fig. 1: Clade 187)



Fig. 9. Cytospora californica (CBS 141612). A. Conidiomata on MEA. B–D. Conidiophores and conidiogenous cells. E. Conidia. Scale bars: A = 100 μm; B–D = 10 μm; E = 5 μm.



Fig. 10. *Cytospora castaneicola* (BJFC-S1704). A. Conidioma on host issue. B. Tangential section through conidiomata. C. Longitudinal section through conidioma. D, E. Conidiophores and conidiogenous cells. F. Conidia. Scale bars: A–C = 200 µm; D–F = 10 µm.

*Basionym*: Sphaeria ceratosperma Tode, Fung. Mecklenb. Sel. (Lüneburg) 2: 53. 1791.

Synonyms: Diatrype ceratosperma (Tode) Fr., Summa veg. Scand., Sectio Post. (Stockholm): 385. 1849.

Valsa ceratosperma (Tode) Maire, Publ. Inst. Bot. Barcelona 3(no. 4): 20. 1937.

*Cytospora sacculus* (Schwein.) Gvrit., Mikol. Fitopatol. 3: 207. 1969.

Cytospora vinacea D.P. Lawr. et al., Pl. Pathol. 66: 720. 2017.

*Cytospora predappioensis* Q.J. Shang *et al.*, Mycosphere 9: 376. 2018.

*Cytospora ceratospermopsis* C.M. Tian & X.L. Fan, Persoonia 45: 19. 2019.

*Typus*: **Europe**, unknown collection details [**neotype** Stirp. Vog.-Rhen. 567 (CUP) (as *Sphaeria ceratosperma*) (*fide* Hubbes 1960; not examined)].

Descriptions: See Hayova & Minter (1998a), Adams *et al.* (2005) and Fan *et al.* (2020).

Description (BJFC-S2126): Conidiomata Group AIII (type a11), scattered or serried, immersed in bark, erumpent through the bark surface, conical, with multiple locules. Conceptacle absent. Ectostromatic disc amber, circular,  $(240-)270-420(-480) \mu m$  diam, with one ostioles per disc. Ostiole grey to black,  $(60-)85-130(-140) \mu m$  diam. Locules numerous, subdivided frequently with independent walls,  $(320-)430-710(-820) \mu m$  diam. Conidiophores borne along the locules, hyaline, branched at the base or occasionally unbranched,  $(13.5-)15-28(-30) \times 1-2 \mu m$ , embedded in a gelatinous layer. Conidiogenous cells enteroblastic, phialidic, subcylindrical to cylindrical. Conidia hyaline, allantoid, thin-walled, eguttulate, aseptate, smooth,  $(4-)4.5-6(-6.5) \times 1-1.5$  (av. = 5 ± 0.4 × 1.3 ± 0.2, n = 30) µm.

*Culture characteristics* (BJFC-S2126, culture CFCC 56503): Colonies white, thick, growing up to 9-cm-diam after 5 d, with



felty aerial mycelium and regular edges. Colonies becoming grey olivaceous (reverse) and smoky (surface).

Description (BJFC-S2165): Conidiomata Group AIII (type a11), immersed in bark, scattered, erumpent through the surface, with multiple locules. Conceptacle absent. Ectostromatic disc sulphur yellow to slight brown, conspicuous, circular to ovoid, 555–780(– 1 180) µm diam, with one ostiole per disc. Ostiole in the centre of the disc, black, inconspicuous, at the same level as the disc surface, (60–)75–120(–135) µm diam. Locules numerous, subdivided frequently by invaginations with independent walls, (1 070–)1 100– 1 280(–1 455) µm diam. Conidiophores borne along the locules, hyaline, unbranched or occasionally branched at base, middle, thin-walled, (7.5–)8.5–18.5 × 1.5–2 µm, embedded in a gelatinous layer. Conidiogenous cells enteroblastic, phialidic, subcylindrical to cylindrical, 8.5–12 × 1.5–2 µm, tapering towards apices. Conidia hyaline, allantoid, eguttulate, smooth, aseptate, thin-walled, (4.5–) 5–5.5(–6.5) × 1–1.5 (av. = 5.22 ± 0.3 × 1.4 ± 0.2, n = 30) µm.

*Culture characteristics* (BJFC-S2165, culture CFCC 55991): Colonies initially white, entirely covering the 9-cm-diam Petri dish after 4 d, becoming smoke grey with olivaceous pigment, flat with a uniform texture, with sparse aerial mycelium. Sterile.

Additional materials examined: Austria, Kärnten, St. Margareten im Rosental, on *Quercus petraea*, 18 Mar. 2000, *W. Jaklitsch*, WJ 1425 (BPI 748459, culture CBS 109777 = AR 3426). **Canada**, Quebec, Beauport, on twig of *Ulmus* sp., 2006, *L.C. Mejia* (CBS H-22938, culture CBS 141624 = CPC 28414 = LCM 07.02). **China**, Beijing City, Mentougou District, Xiaolongmen Forestry Centre, 39°57'41"N, 115°25'42"E, on twigs and branches of *Juglans mandshurica*, 25 Aug. 2022, *L. Lin & X.L. Fan* (BJFC-S1993, culture CFCC 58252); *ibid.*, 39°50'5"N, 115°34'35"E, on twigs and branches of *Juniperus chinensis*, 24 Aug. 2022, *L. Lin & X.L. Fan* (BJFC-S1994, culture CFCC 58461); *ibid.*, 39°58'46.27"N, 115°26'58.43"E, *on branches of Cotinus coggygria*, 23 Aug. 2021, *X.L. Fan* (BJFC-S2126, living CFCC 56503; BJFC-S2127, culture CFCC 58946); Henan Province, Nanyang City, Baotianman National Nature Reserve, 33°31'3.46"N, LIN ET AL.



Fig. 11. Cytospora ceratosperma (BJFC-S2126). A, B. Habit of conidiomata on twig. C. Transverse section through conidioma. D. Longitudinal section through conidioma. E. Conidiophores and conidiogenous cells. F. Conidia. Scale bars: A = 1 mm; B–D = 500 µm; E, F = 10 µm.

111°55'10.12"E, on twigs and branches of *Lindera reflexa*, 6 Oct. 2021, *C. Peng & C.M. Tian* (BJFC-S2165, culture CFCC 55991; BJFC-S2166, culture CFCC 58945). **Italy**, Milano, on trunk of *Castanea sativa*, 1980, *M. Bisiach* (culture CBS 217.81). **Russia**, on *Salix* sp., 30 May 2000, *L. Vasilyev* (culture CBS 141608 = CPC 28368 = AR 3418). **Sweden**, Stockholm, on *Fagus sylvatica*, Aug. 1976, unknown *collector* (CBS H-19146, culture CBS 512.76). **Switzerland**, on *Taxus baccata*, unknown date and *collector* (culture CBS 192.42); on *Vitis vinifera*, unknown date, *G. Défago* (culture CBS 397.36). **USA**, on twig of *Salix lucida*, 2008, *L.C. Mejía* (CBS H-22932, culture CBS 141611 = CPC 28371 = LCM 437.02).

*Notes: Cytospora ceratosperma* is a common but taxonomically confused species with a wide host range and worldwide distribution. It has a long, complicated taxonomic history and many synonyms (Spielman 1985). Adams *et al.* (2005) narrowed the species concept using ITS sequence data to solve the overlapping morphology of many specimens named *C. ceratosperma*. However, the species boundary remains unclear due to lack of type material and the

limitation of an ITS-generated phylogeny. In the original description, Tode (1791) provided no host for *Sphaeria ceratosperma*. Hubbes (1960) checked the neotype of *S. ceratosperma* [Mougeot & Nestler 1818, Stirp. Vog.-Rhen. 567 (ILT) (CUP)], which was collected from *Acer campestre*, and an epitype should be selected from that host to stabilize the application of the name (Spielman 1985).

A total of 21 isolates including *C. ceratosperma*, *C. ceratospermopsis*, *C. predappioensis*, *C. vinacea* and new strains obtained in the present study cluster in a large clade with high support value for each gene (in ITS, *act1*, *rpb2*, *tef1-a* and *tub2* gene trees, ML/BI = 77/1, 97/1, 100/1, 100/1 and 98/1, respectively). They can be distinguished from each other in the tree of the *tef1-a* gene (Fig. S4). However, the phylogenies for the other genes do not resolve them from one another (Figs S1–3, 5). Morphologically, *C. ceratosperma* (BJFC-S774), *C. ceratospermopsis* (BJFC-S567), the specimen BJFC-S2126 and the specimen BJFC-S2165 have the same asexual morph type (Group AIII, type a11) and overlapping



Fig. 12. Cytospora ceratosperma (BJFC-S2165). A, B. Habit of conidiomata on twig. C, D. Transverse sections through conidiomata. E. Longitudinal section through conidioma. F. Conidiophores and conidiogenous cells. G. Conidia. Scale bars: A = 1 mm; B-E = 500 µm; F, G = 10 µm.

conidial sizes (Lawrence *et al.* 2017, Fan *et al.* 2020). Therefore, *Cytospora ceratospermopsis* (CFCC 89626), *C. predappioensis* (MFLUCC 17-2458) and *C. vinacea* (CBS 141585) are reduced to synonymy with *C. ceratosperma*.

*Cytospora cerebriformis* L. Lin & X.L. Fan, *sp. nov.* MycoBank MB 850146. Fig. 13. (Fig. 1: Clade 111)

Etymology: Name after the country where it was collected, China.

*Typus*: **China**, Gansu Province, Qingyang City, Heshui County, 35°47'04.66"N, 107°59'37.03"E, on branches of *Prunus persica*, 13 Jul. 2013, *X.L. Fan* (**holotype** BJFC-S918, ex-holotype culture CFCC 50020); Beijing City, Fengtai District, Beigong National Forest Park, 39°51'42"N, 116°7'8"E, on twigs and branches of *Prunus triloba*, Aug. 2022, *Y.K. Bai* & *M. Pan* (**paratype** BJFC-S2054, ex-paratype culture CFCC 59061).

Description: Conidiomata Group SII (type a7), immersed in bark, erumpent when mature, discoid to conical, 700–1 150  $\mu$ m diam,

with multi-locule. *Conceptacle* prominent. *Ectostromatic disc* buff to honey-coloured, circular, 130–270 µm diam, with single ostiole per disc in the centre. *Ostiole* circular to ovoid, black, 55–75 µm diam. *Locules* multiple, subdivided with common walls. *Conidiophores* hyaline, unbranched or occasionally branched, 11.5–17.5 × 1–1.5 µm (av. =  $13.7 \pm 1.7 \times 1.3 \pm 0.1 \mu$ m, n = 30). *Conidiogenous cells* enteroblastic, phialidic, subcylindrical to cylindrical, 13.5–16.5 × 1–2 µm (av. =  $15.2 \pm 1.1 \times 1.6 \pm 0.1 \mu$ m, n = 30). *Conidia* hyaline, unicellular, eguttulate, elongate-allantoid, 4.5–6 × 1–1.5 µm (av. =  $5.2 \pm 0.3 \times 1.35 \pm 0.1 \mu$ m, n = 50).

Culture characteristics: See Fan et al. (2020, as C. leucostoma).

Additional materials examined: China, Beijing City, Yanqing District, Duck Lake Wetland Park, 40°24'39.060"N, 115°51'23.774"E, on twigs and branches of *Platycladus orientalis*, 3 Jul. 2022, Y.K. Bai & X.L. Fan (BJFC-S2053, culture CFCC 59051); *ibid.*, Changping District, Mangshan National Forest Park, 40°15'14.029"N, 116°18'25.639"E, on twigs and branches of *Prunus davidiana*, 1 Jul. 2022, Y.K. Bai & M. Pan



Fig. 13. Cytospora cerebriformis (BJFC-S918). A, B. Habit of conidiomata on twig. C. Transverse section through conidioma. D. Longitudinal section through conidioma. E. Conidiophores and conidiogenous cells. F. Conidia. Scale bars: A = 2 mm; B–D = 200 µm; E, F = 10 µm.



(BJFC-S2055, culture CFCC 59064); *ibid.*, Fangshan District, Xiayunling National Forest Park, 39°44'6"N, 115°43'44"E, on twigs and branches of *Prunus davidiana*, 3 Aug. 2022, *Y.K. Bai & M. Pan* (BJFC-S2056, culture CFCC 59076).

Notes: Specimen BJFC-S918 (culture CFCC 50020) was treated as C. leucostoma by Fan et al. (2020). In this study, clade 122 is applied to C. leucostoma and this clade (111) is described as a new species, C. cerebriformis. See notes on C. leucostoma (Clade 122) for details. Cytospora cerebriformis has been recorded in Beijing City, Shanxi Province, Gansu Province, Qinghai Province and the Ningxia Hui Autonomous Region in China. Phylogenetically, C. cerebriformis and C. donetzica form sister clades with maximum support (ML/BI = 100/1). The type specimen of *C. donetzica* was collected from dead and dying branches of Rosa sp. in Russia (Norphanphoun et al. 2017). Fan et al. (2020) treated C. donetzica as a synonym of *C. cerebriformis*. These two species are not clearly differentiated with ITS (100.0 % identity, with 0 bp difference), act1 (98.5 % identity, with 3 bp differences) and rpb2 (98.6 %, with 7 bp differences). Morphologically, C. cerebriformis is characterised by a black conceptacle (Fan et al. 2020). However, a black conceptacle is not specifically mentioned in the description of C. donetzica, and it is also not readily apparent in the figures (Norphanphoun et al. 2017).

*Cytospora chrysosperma* (Pers.) Fr., Syst. Mycol. (Lundae) 2(2): 542. 1823. Fig. 14. (Fig. 1: Clade 63)

Basionym: Sphaeria chrysosperma Pers., Neues Mag. Bot. 1: 82. 1794.

Synonyms: Naemaspora chrysosperma (Pers.) Pers., Observ. Mycol. (Lipsiae) 1: 80. 1796.

Valsa sordida Nitschke, Pyrenomyc. Germ. 2: 203. 1870.

Cytospora populicola D.P. Lawr. et al., IMA Fungus 9: 363. 2018.

*Typus*: **Sweden**, on *Populus* sp. **[isotype** Fries, Scler. Suec. 154 (BPI); **lectotype** designated here Herb. Univ. Upsaliensis (F-117599) 289268, MBT 10019183]. **UK**, unknown specific locality, on twig of *Populus tremula*, 1962, *Peace* (**epitype** designated here CBS 197.50, culture preserved in metabolically inactive state, MBT 10019184).

Descriptions: See Hayova & Minter (1998d), Adams *et al.* (2005) and Lawrence *et al.* (2018).

*Materials examined*: **Netherlands**, Utrecht, Baarn, Eemnesserweg, on dead branches of *Populus balsamifera*, 1982, *H.A. van der Aa* (culture CBS 120.83); *ibid.*, on dead branches of *Populus* sp., 1988, *H.A. van der Aa* (culture CBS 604.88); North Brabant, Tilburg, Oirschot, on dead trunk of *Populus* sp., unknown date and *collector* (culture CBS 133.46); North Holland, Castricum, on *Populus* sp., 1988, *H.A. van der Aa* (CBS H-19390,

culture CBS 195.49); Unknown specific locality, on *Populus robusta*, unknown date and *collector* (culture CBS 290.52). **Unknown country and location**, on *Fraxinus americana*, unknown date and *collector* (culture CBS 134.25).

*Notes: Cytospora chrysosperma* is the type species of *Cytospora* (Donk 1964). This fungus was described from *Populus* in Europe (Persoon 1794) and also in the sanctioning publication (Fries 1823) only *Populus* is mentioned as host. We have been unable to trace a type specimen designated by Persoon, but a copy of Scleromycetes Sueciae 154 mentioned in the sanctioning publication by Fries (1823) in BPI was chosen as an isotype (Spielman 1985). Another copy of Scleromycetes Sueciae 154, Herb. Univ. Upsaliensis (F-117599) 289268, is selected as a lectotype here. Isolate CBS 197.50 collected on the same host genus (*Populus*) and same continent (Europe) as the lectotype is therefore here designated as an epitype of *C. chrysosperma*.

Cytospora chrysosperma clusters in a strongly supported clade with *C. sophoriopsis*. However, *C. chrysosperma* (CBS 197.50) differs from *C. sophoriopsis* (CFCC 89600) in ITS (1/513), act1 (6/255), rpb2 (7/650), tef1-a (13/490) and tub2 (10/411) genes. The isolate CBS 144240, described by Lawrence et al. (2018) as *C. populicola*, clusters with *C. chrysosperma* in the present study. Therefore, *C. populicola* is treated as a synonym of *C. chrysosperma*. CBS 195.49 and CBS 134.25 has been identified as *C. hariotii* and *C. minuta* respectively by Adams (2006), but results of the present study show that both of these two isolates represent *C. chrysosperma*.

*Cytospora cinereostroma* (G.C. Adams & M.J. Wingf.) G.C. Adams & Rossman, IMA Fungus 6: 147. 2015. (Fig. 1: Clade 194) *Basionym: Valsa cinereostroma* G.C. Adams & M.J. Wingf., Stud. Mycol. 52: 73. 2005.

*Typus*: **Chile**, unknown specific locality, on dead branches of *Eucalyptus globulus*, 2000, *M.J. Wingfield* (**holotype** of *Valsa cinereostroma* MSC 375220, ex-holotype culture CBS 117081).

Description: See Adams et al. (2005).

Additional materials examined: Chile, unknown specific locality, on *Eucalyptus nitens*, unknown date, *M.J. Wingfield* (culture CBS 117082). South Africa, on fruit of *Mangifera indica*, 1998, *C. Roux* (culture CBS 116830);Barberton,WhiteRiver,JessievaleStateForest,on*Eucalyptus nitens*, 1988, *P.W. Crous* (cultures CBS 116831, 116832 and 116833).

Notes: Cytospora cinereostroma can be distinguished from the other leucostomoid species on Eucalyptus based on morphology.



Fig. 14. The lectotype designated here of Cytospora chrysosperma (Herb. Univ. Upsaliensis (F-117599) 289268).

Conidiomata of *C. cinereostroma* are Group AII (type a10), while other species on *Eucalyptus* have leucotorsellioid conidiomata (Group AIII, type a12) (Adams *et al.* 2005). Phylogenetically, the available six isolates of *C. cinereostroma* form a distinct clade from other species. The phylogenies of ITS, *tef1-a* and *tub2* gene place it as a distinct clade (ML/BI = 73/0.62, 95/1 and 92/1, respectively) inside a larger clade containing *C. austromontana*, *C. berkeleyi*, *C. carpobroti* and *C. diatrypelloidea*. The phylogenies for the *act1* gene places it in a distinct clade (ML/BI = 79/1) within a larger clade containing *C. berkeleyi* and *C. diatrypelloidea*. The *rpb2* gene phylogeny places it as a distinct clade (ML/BI = 62/0.86) within a larger clade containing *C. austromontana*, *C. berkeleyi* and *C. diatrypelloidea*.

*Cytospora conceptaculata* L. Lin & X.L. Fan, *sp. nov.* MycoBank MB 850148. Fig. 15. (Fig. 1: Clade 139)

*Etymology*: Name refers to its prominent black conceptacle (conceptaculum).

*Typus*: **Netherlands**, unknown specific locality, on dead wood, 2019, *N. Jiang* (**holotype** BJFC-S2102, ex-holotype culture CFCC 54019; **paratype** BJFC-S2103, ex-paratype culture CFCC 54020).

Description: Conidiomata Group AIII (type a12), immersed in bark, erumpent when mature, discoid to conical, 600–850 µm diam, with multi-locule. Conceptacle prominent. Ectostromatic disc buff to honey, circular to triangular, 250–310 µm diam, with single ostiole per disc in the centre. Ostiole circular to ovoid, hazel to black, 75–95 µm diam. Locules 9–11, subdivided with individual walls. Conidiophores hyaline, unbranched or occasionally branched, 9.5–17.5 × 1.5–2 µm (av. =  $13.5 \pm 2.0 \times 1.8 \pm 0.1$  µm, n = 30). Conidiogenous cells enteroblastic, phialidic, subcylindrical to cylindrical,  $3.5-8.5 \times 1.5-2.5$  µm (av. =  $6.0 \pm 1.0 \times 1.9 \pm 0.2$  µm, n = 30). Conidia hyaline, unicellular, eguttulate, elongate-allantoid,  $5-7 \times 1.5-2$  µm (av. =  $6.2 \pm 0.4 \times 1.65 \pm 0.1$  µm, n = 50).

*Culture characteristics*: Cultures on PDA initially white, growing up to 4 cm diam after 3 d, entirely covering the 6-cm-diam Petri dish after 4 d and becoming honey-coloured at the centre after 10 d, flat with a uniform texture.

*Notes*: Phylogenetically, *C. conceptaculata* is closely related to *C. pruni-mume* (Fig. 1). However, the isolates CFCC 54019 and 54020 form a distinct clade with full support (ML/BI = 100/1) and differ from CFCC 53180 (the ex-holotype of *C. pruni-mume*) by ITS (2/512 bp), *act1* (10/247 bp), *rpb2* (2/697 bp), *tef1-a* (10/494 bp) and *tub2* (1/406 bp). Morphologically, *C. conceptaculata* differs from *C. pruni-mume* in having a black conceptacle.

*Cytospora crataegina* X.L. Fan & C.M. Tian, *sp. nov.* MycoBank MB 850150. Fig. 16. (Fig. 1: Clade 21)

*Etymology*: Name refers to the host genus, *Crataegus*.

*Typus*: **China**, Beijing City, Huairou District, Forestry Centre of Beijing University of Agriculture, 40°52'50.47"N, 116°26'23.13"E, on branches of *Crataegus pinnatifida* var. *major*, 18 Jun. 2021, *X.L. Fan* & *Y.K. Bai* (**holotype** BJFC-S2128, ex-holotype culture CFCC 56027; **paratype** BJFC-S2129, ex-paratype culture CFCC 56028).

Description: Conidiomata Group AII (type a6), immersed in bark, scattered, erumpent through the surface of bark, with multiple locules. Conceptacle absent. Ectostromatic disc buff to dark, ovoid to circular,  $(430-)460-650(-700) \mu m$  diam, with a solitary ostiole per disc. Ostiole black, protruding above the disc surface,  $105-140(-165) \mu m$  in diam. Locules numerous, arranged regularly with common walls,  $(1\ 220-)1\ 425-1\ 850(-2\ 120) \mu m$  in diam. Conidiophores hyaline, branched at base, in the middle or unbranched, thin walled, filamentous,  $12.5-21.5(-24.5) \times 1.5-2 \mu m$ . Conidiogenous cells enteroblastic, phialidic. Conidia hyaline, allantoid, eguttulate, smooth, aseptate, thin-walled,  $3.5-4.5(-5) \times 1-1.5$  (av. =  $3.8 \pm 0.2 \times 1.2 \pm 0.1$ , n = 30)  $\mu m$ .

*Culture characteristics*: Colonies on PDA initially white, becoming honey, growing fast and entirely covering the 9-cm-diam Petri dish after 7 d, becoming hazel to isabelline after 14 d, felty with a uniform texture, without aerial mycelium, sterile.

Additional material examined: **China**, Beijing City, Huairou District, Forestry Centre of Beijing University of Agriculture, 40°52'50.47"N, 116°26'23.13"E, on branches of *Crataegus pinnatifida* var. *major*, 18 Jun. 2021, *X.L. Fan & Y.K. Bai* (BJFC-S2130, culture CFCC 56029).



**Fig. 15.** *Cytospora conceptaculata* (BJFC-S2102). **A**, **B**. Habit of conidiomata on twig. **C**. Transverse section through conidioma. **D**. Longitudinal section through conidioma. **E**, **F**. Conidiophores and conidiogenous cells. **G**. Conidia. Scale bars: A = 1 mm; B–D = 200 μm; E–G = 10 μm.





Fig. 16. Cytospora crataegina (BJFC-S2128). A, B. Habit of conidiomata on twig. C. Transverse section through conidioma. D. Longitudinal section through conidioma. E. Conidiophores and conidiogenous cells. F. Conidia. Scale bars: A = 1 mm; B–D = 500 µm; E, F = 10 µm.

Notes: Cytospora crataegina was associated with a canker disease on Crataegus pinnatifida var. major. Phylogenetically, C. crataegina is sister to C. malvicolor (Fig. 1). However, C. crataegina (CFCC 56027) differs from C. malvicolor (CFCC 56577) in the ITS (14/522), act1 (7/243), rpb2 (31/457), tef1- $\alpha$  (23/488) and tub2 (13/399) loci. Morphologically, C. crataegina differs from C. malvicolor due to the absence of a conceptacle and by its smaller conidial size [3.5–4.5(– 5) × 1–1.5 µm vs (6–)6.5–7.5(–8) × 1.5–2 µm].

*Cytospora deqinensis* L. Lin & X.L. Fan, *sp. nov.* MycoBank MB 850153. Fig. 17. (Fig. 1: Clade 82)

*Etymology*: Name refers to Deqin County, Diqing Perfecture where it was collected.

*Typus*: **China**, Yunnan Province, Diqing Tibetan Autonomous Prefecture, Deqin County, Shengping Town, Hexiang Middle Road, 28°28'51"N, 98°55'1"E, on branches of *Populus haoana*, 9 Aug. 2022, *L. Lin & M. Lin* (**holotype** BJFC-S2000, ex-holotype culture CFCC 58626); *ibid.*, 28°28'30"N, 98°55'5"E, elevation ~ 3 300 m, on branches of *Populus haoana*, 9 Aug. 2022, *L. Lin & M. Lin* (**paratype** BJFC-S1996, ex-paratype culture CFCC 58467).

Description: Conidiomata Group AII (type a6), immersed in bark, erumpent when mature, discoid to conical, 700–980 µm diam, with multi-locule. Conceptacle absent. Ectostromatic disc isabelline to olivaceous, circular to ovoid, 155–265 µm diam, with single ostiole per disc in the centre. Ostiole circular to ovoid, brown to black, 75–115 µm diam. Locules numerous, divided with shared walls. Conidiophores hyaline, unbranched or occasionally branched at the lower middle,  $19.5-30 \times 1-2 \mu m$  (av. =  $24.3 \pm 3.3 \times 1.6 \pm 0.2 \mu m$ , n = 30). Conidiogenous cells enteroblastic, phialidic, subcylindrical to cylindrical,  $3.5-9 \times 1-2 \mu m$  (av. =  $6.1 \pm 1.7 \times 1.5 \pm 0.2 \mu m$ , n = 30). Conidia hyaline, unicellular, eguttulate, elongate-allantoid,  $3.5-5.5 \times 1-2 \mu m$  (av. =  $4.5 \pm 0.5 \times 1.5 \pm 0.2 \mu m$ , n = 50).

Culture characteristics: Cultures on PDA initially white, growing up

to 3.5 cm diam after 3 d, entirely covering the 6-cm-diam Petri dish after 5 d and becoming sulphur yellow after 20 d, with felty aerial mycelium and irregular edge.

Additional materials examined: China, Yunnan Province, Diqing Tibetan Autonomous Prefecture, Deqin County, Shengping Town, Hexiang Middle Road, 28°28'56"N, 98°55'7"E, on branches of *Populus haoana*, 9 Aug. 2022, *L. Lin & M. Lin* (BJFC-S1997, culture CFCC 58468); *ibid.*, 28°28'50"N, 98°55'23"E, on branches of *Populus haoana*, 9 Aug. 2022, *L. Lin & M. Lin* (BJFC-S1998, culture CFCC 58469); *ibid.*, 28°28'30"N, 98°55'5"E, on branches of *Populus haoana*, 9 Aug. 2022, *L. Lin & M. Lin* (BJFC-S1999, culture CFCC 58625); Shangri-La City, Sanba Naxi Town, East Ring Road, 27°36'18"N, 100°1'19"E, on branches of *Populus szechuanica*, 9 Aug. 2022, *L. Lin & M. Lin* (BJFC-S1995, culture CFCC 58192).

*Notes: Cytospora deqinensis* is represented by isolates from branches of *Populus haoana* and *Populus szechuanica* exhibiting canker disease symptoms. It is phylogenetically clearly distinct from all species of the genus *Cytospora*. Morphologically, it can be distinguished by its colony on PDA with felty aerial mycelium and irregular edge. Therefore, it is described as a new species associated with poplar canker disease.

Cytospora desmazieri L. Lin, X.L. Fan & Crous, nom. nov. MycoBank MB 853111. (Fig. 1: Clade 163)

Replaced synonym: Sphaeria pini Alb. & Schwein., Consp. Fung. (Leipzig): 20. 1805, non Cytospora pini Desm., Nouv. Not.: 46. 1843.

Synonyms: Cytospora pini Desm., Nouv. Not.: 46. 1843.

Valsa pini (Alb. & Schwein.) Fr., Summa veg. Scand., Sectio Post. (Stockholm): 412. 1849.

Cytospora pinicola Westend., Pl. Crypt. Exsicc. 19-20: 933. 1854.

*Etymology*: Named in honour of the France mycologist Desmaziere, in recognition of his contributions to this species.

Typus: Requires typification.



Fig. 17. Cytospora deginensis (BJFC-S2000). A, B. Habit of conidiomata on twig. C. Transverse section through conidioma. D. Longitudinal section through conidioma. E. Conidiophores and conidiogenous cells. F. Conidia. Scale bars: A = 1 mm; B = 200 µm; C, D = 500 µm; E, F = 10 µm.

Description: See Saccardo (1884) (as Cytospora pini).

Material examined: Switzerland, Valais, Finger, on Pinus sylvestris, unknown date, G. Défago (culture CBS 197.42).

Notes: Cytospora desmazieri L. Lin, X.L. Fan & Crous is published here as an explicit substitute ("nom. nov.") for the legitimate name Cytospora pini Desm. (Nouv. Not.: 46. 1843). According to Saccardo (1884), Cytospora pini Desm. and C. pinicola Westend. are the asexual morphs of Valsa pini. In the protologue of C. pini, Sphaeria pini is not mentioned. Therefore, C. pini cannot be treated as a new combination and the earlier Sphaeria pini, which has priority, cannot be combined in Cytospora as the epithet is occupied. Adams et al. (2005) identified CBS 197.42 as Cytospora pini. It clusters in a sister phylogenetic clade with C. kunzei (CBS 118093) but differs from that species in the ITS (29/519 bp) gene sequence. As this isolate was obtained from the same host and continent as that for the original record (France, Belgium, from Pinus sylvestris) (Saccardo 1884), we recommend treating CBS 197.42 as a reference strain for C. desmazieri. This will first require detailed type studies to determine whether the heterotypic names are conspecific.

*Cytospora diatrypelloidea* G.C. Adams & M.J. Wingf., Stud. Mycol. 52: 121. 2005. (Fig. 1: Clade 193)

*Typus*: **Australia**, Victoria, Orbost, on dead branches of *Eucalyptus globulus*, 2000, *M.J. Wingfield* (holotype MSC 380719, ex-holotype culture CBS 116826 = CMW 8549).

Description: See Adams et al. (2005).

Additional materials examined: Australia, Perth, Kings Park, on leaves of *Eucalyptus* sp., 2005, *A. van Iperen* (CBS H-22981 dried culture, culture CBS 120062 = CPC 12453). **South Africa**, Western Cape Province, Cape Town, on dead twigs of *Eucalyptus* sp., 1995, *C. Roux* (culture CBS 116822).

Notes: Adams *et al.* (2005) introduced *C. diatrypelloidea* from *Eucalyptus globulus* in Australia. It can be distinguished from other species by its hemispherical diatrypelloid stromata with individual locules (Adams *et al.* 2005). *Cytospora diatrypelloidea* is sister to *C. berkeleyi* but differs phylogenetically (see notes on *C. berkeleyi*). In this study, South Africa is added to the geographical range of this species.

Cytospora diqingensis L. Lin & X.L. Fan, sp. nov. MycoBank MB 850155. Fig. 18. (Fig. 1: Clade 128)

*Etymology*: Name refers to Diqing Tibetan Autonomous Prefecture where it was collected.

*Typus*: **China**, Yunnan Province, Diqing Tibetan Autonomous Prefecture, Shangri-La City, Sanba Naxi Town, East Ring Road, 27°36'18"N, 100°1'19"E, on branches of *Populus szechuanica*, 9 Aug. 2022, *L. Lin & M. Lin* (**holotype** BJFC-S2083, ex-holotype culture CFCC 58245, ex-isotype culture CFCC 58242); *ibid.*, 27°36'13"N, 100°1'29"E, on branches of *Populus szechuanica*, 9 Aug. 2022, *L. Lin & M. Lin* (**paratype** BJFC-S2082, ex-paratype culture CFCC 58244).

Description: Pseudostromata Group SII (type s5), immersed in bark, scattered, with 5–8 perithecia arranged irregularly. *Conceptacle* prominent. *Ectostromatic disc* salmon to umber, usually surrounded by ostiolar necks, circular to ovoid, 285–415 µm diam, with 4–8 ostioles irregularly arranged ostioles. *Ostioles* numerous, buff to black when mature, 40–65 µm diam. *Perithecia* black when mature, flask-shaped to spherical, irregularly arranged, 150–200 µm diam. *Asci* hyaline, with chitinoid, refractive ring, clavate to elongate-obovoid, 35–50 × 7.5–10.5 µm (av. = 41.5 ± 4.3 × 8.9 ± 0.8 µm, n = 30), 8-spored. *Ascospores* hyaline, biseriate to multiseriate, elongate-allantoid, aseptate, 8.5–12.5 × 1.5–3 µm (av. = 10.2 ± 0.9 × 2.2 ± 0.2 µm, n = 50).

*Culture characteristics*: Cultures on PDA are initially white, growing up to 3 cm diam after 10 d, entirely covering the 6-cm-diam Petri





Fig. 18. Cytospora diqingensis (BJFC-S2083). A, B. Habit of pseudostromata on twig. C. Transverse section through pseudostroma with ascomata. D. Longitudinal section through pseudostroma with ascomata. E–G. Asci. H. Ascospores. Scale bars: A = 2 mm; B–D = 200 µm; E–H = 10 µm.

dish after 6 d and becoming honey after 10 d. The colonies are isabelline to black after 20 d, flat with a uniform texture and regular edge.

Notes: Phylogenetically, *C. diqingensis* is closely related to *C. elaeagnina* (Fig. 1). In ITS, *act1, rpb2, tef1-a* and *tub2* gene trees, the BI and ML phylogenies all place the three *C. diqingensis* isolates in a distinct clade with high support (ML/BI = 100/1) within a larger clade containing *C. elaeagnina*. However, *C. diqingensis* (CFCC 58246) differs from *C. elaeagnina* (CFCC 56018) in ITS (9/509), *act1* (8/242), *rpb2* (15/726), *tef1-a* (20/256) and *tub2* (20/413) loci. Morphologically, it differs from *C. elaeagnina* based on its disc colour; the ectostromatic disc is salmon to umber in *C. diqingensis*, and white to buff in *C. elaeagnina*. In the present study, *C. deqinensis* and *C. sophoriopsis* were also collected from *Populus szechuanica*. However, they are clearly distinct from *C. diqingensis* in the phylogram (Fig. 1).

*Cytospora disciformis* G.C. Adams & M.J. Wingf., Stud. Mycol. 52: 111. 2005. (Fig. 1: Clade 200)

*Typus*: **Uruguay**, Los Ceibos, on dead branch of *E. globulus*, 1999, *M.J. Wingfield* (**holotype** MSC 368323, ex-holotype culture CBS 116827 = CMW 6509).

Description: See Adams et al. (2005).

Additional material examined: Australia, Canberra, on dead branch of *E. globulus*, 2001, *M.J. Wingfield* (cultures CBS 116828, CBS 118083).

Notes: In act1, rpb2, tef1- $\alpha$  and tub2 gene trees, the BI and ML phylogenies all place *C. disciformis* in a distinct clade with high support (ML/BI = 100/1). This species forms conidiomata arranged in rosettes (Group AII, a7) and its conidioma is circular with a regular chamber shape and no compression among them, invaginations are regular, short and extend less than halfway through the chambers (Adams *et al.* 2005). The Australian isolates differ in their DNA sequences and additional investigations are necessary to resolve their species status.

*Cytospora donglingensis* M. Pan & X.L. Fan, Front. Plant Sci. 12 (no. 636460): 8. 2021. (Fig. 1: Clade 75)

*Typus*: **China**, Beijing City, Mentougou District, Mount Dongling, Xiaolongmen Forestry Centre, on branches of *Platycladus orientalis*, 17 Aug. 2017, *H.Y. Zhu & X.L. Fan* (**holotype** BJFC CF2019884, ex-holotype culture CFCC 53159).

Descriptions: See Pan et al. (2021), Lin et al. (2023a).

Materials examined: China, Beijing City, Mentougou District, Xiaolongmen Forestry Centre, 39°57'61"N, 115°25'62"E, on twigs and branches of Populus × beijingensis, 25 Aug. 2022, L. Lin & X.L. Fan (BJFC-S2002, culture CFCC 58206); ibid., 39°57'41"N, 115°25'42"E, on twigs and branches of Populus × beijingensis, 25 Aug. 2022, L. Lin & X.L. Fan (BJFC-S2003, culture CFCC 58207); ibid., 39°50'49"N, 115°33'12"E, on twigs and branches of Populus × beijingensis, 24 Aug. 2022, L. Lin & X.L. Fan (BJFC-S2007, culture CFCC 58436); Yunnan Province, Dali City, Xiaguan District, 25°36'58"N, 100°12'37"E, on branches of Populus × canadensis, 4 Aug. 2022, L. Lin & M. Lin (BJFC-S2004, culture CFCC 58215); ibid., 25°39'9"N, 100°10'50"E, on branches of Populus × canadensis, 4 Aug. 2022, L. Lin & M. Lin (BJFC-S2005, culture CFCC 58218); Diqing Tibetan Autonomous Prefecture, Shangri-La City, Sanba Naxi Town, East Ring Road, 27°36'18"N, 100°1'19"E, on branches of Juglans regia, 9 Aug. 2022, L. Lin & M. Lin (BJFC-S2006, culture CFCC 58220); ibid., Weixi Lisu Autonomous County, Yezhi Town, 27°49'37"N, 99°2'38"E, on branches of Populus × canadensis, 10 Aug. 2022, L. Lin & M. Lin (BJFC-S2008, culture CFCC 58491); ibid., Badi Town, 27°49'37"N, 99°1'58"E, on branches of Populus × canadensis, 10 Aug. 2022, L. Lin & M. Lin (BJFC-S2009, culture CFCC 58494).

Notes: Cytospora donglingensis was described from *Platycladus* orientalis in China, with a description of its sexual morph on branches and asexual morph on PDA medium (Pan *et al.* 2021). In the *act1*, *rpb2*, *tef1-a* and *tub2* gene trees, the BI and ML phylogenies all place it in a distinct clade, with high support (ML/BI = 85/1, 90/1, 95/1 and 97/1, respectively). Lin *et al.* (2023a) added the asexual morph of *C. donglingensis* from *Populus* spp. and tested the pathogenicity to *Populus alba* var. *pyramidalis*, but no symptoms were observed. However, *C. donglingensis* was isolated from branches with canker symptoms (Pan *et al.* 2021, Lin *et al.* 2023a, this study). It could be saprobic, or a weak pathogen (Lin *et al.* 2023a).

*Cytospora eastringensis* L. Lin & X.L. Fan, *sp. nov.* MycoBank MB 850166. Fig. 19. (Fig. 1: Clade 66)

*Etymology*: Name refers to the East Ring Road, the location where the holotype was collected.

*Typus*: **China**, Yunnan Province, Diqing Tibetan Autonomous Prefecture, Shangri-La City, Sanba Naxi Town, East Ring Road, 27°34'26"N, 100°1'5"E, on branches of *Populus adenopoda*, 9 Aug. 2022, *L. Lin & M. Lin* (**holotype** BJFC-S2038, ex-holotype culture CFCC 58222).

Description: Conidiomata Group AII, type a6, immersed in bark, erumpent when mature, discoid to conical, 900–1 085 µm diam, with multi-locule. Conceptacle absent. Ectostromatic disc buff to black, circular to ovoid, 300–385 µm diam. Ostiole inconspicuous. Locules divided with shared walls. Conidiophores hyaline, unbranched or occasionally branched at the bases, 13–27.5 × 1–2 µm (av. = 18.4 ± 2.9 × 1.3 ± 0.2 µm, n = 30). Conidiogenous cells enteroblastic, phialidic, subcylindrical to cylindrical, 3–4.5 × 1–1.5 µm (av. = 3.6 ± 0.5 × 1.2 ± 0.18 µm, n = 30). Conidia hyaline, unicellular, eguttulate, elongate-allantoid, 4–5.5 × 1.5–2 µm (av. = 4.7 ± 0.3 × 1.5 ± 0.1 µm, n = 50).

*Culture characteristics*: Cultures on PDA initially white, growing up to 3 cm diam after 3 d, entirely covering the 9-cm-diam Petri dish after 6 d and becoming olivaceous after 10 d. Colonies are greenish on surface and reverse dark mouse grey after 20 d, flat with a uniform texture.

Notes: Phylogenetically, C. eastringensis forms a distinct branch in the C. chrysosperma complex in the rpb2 and tef1- $\alpha$  phylogenies (no sequence are available for act1 and tub2 genes). It can be distinguished from other species in the C. chrysosperma complex by its cultures that become olivaceous after 10 d. In the present study, C. alba, C. atrocirrhata, C. sanbansis, C. shangrilaensis and C. translucens were also collected from Populus adenopoda. Of these, C. alba is characterised by its leucocytosporoid conidiomata and white ectostromatic discs, C. atrocirrhata by its leucounilocular conidiomata (Group AI, type a2), C. sanbaensis in having four ascospores per ascus, C. shangrilaensis by its leucotorsellioid conidiomata (Group AIII, type a12) and C. translucens by its leucocytosporoid conidiomata (Group AII, type a7). *Cytospora elaeagni* Allesch., Hedwigia 36 (Beibl.): 162. 1897. (Fig. 1: Clade 42)

*Typus*: **Germany**, on branches of *Elaeagnus angustifolia* (**holotype** Sydow, Mycoth. March. 4497, not examined).

Description: See Fan et al. (2015b).

*Material examined*: **China**, Beijing City, Mentougou District, Xiaolongmen Forestry Centre, 39°59'23.58"N, 115°27'05.00"E, on twigs and branches of *Ulmus pumila*, 22 Aug. 2022, *L. Lin & X.L. Fan* (BJFC-S2010, culture CFCC 58241).

*Notes*: *Cytospora elaeagni* has been found on branches of *Elaeagnus angustifolia* in China, Germany and North America (Sydow 1897, Zhuang 2005). *Cytospora elaeagni* clusters in a sister clade to *C. carbonacea*. However, *Cytospora elaeagni* (CFCC 89632) differs from *C. carbonacea* (CFCC 89947) in the ITS (4/511), *act1* (0/244), *rpb2* (3/726), *tef1-a* (46/479) and *tub2* (2/417) loci. In the present study, *Ulmus* is added as a new host record for *C. elaeagni*.

Cytospora elaeagnina L. Lin & X.L. Fan, sp. nov. MycoBank MB 850156. Fig. 20. (Fig. 1: Clade 129)

Etymology: Name refers to the host genus, Elaeagnus.

*Typus*: **China**, Gansu Province, Gannan City, Yeliguan National Forest Park, 34°57'20.38"N, 103°36'12.55"E, on branches of *Elaeagnus pungens*, 24 Oct. 2020, *C. Peng & C.M. Tian* (**holotype** BJFC-S2137, exholotype culture CFCC 56017; **paratype** BJFC-S2138, ex-paratype culture CFCC 56018).

Description: Pseudostromata immersed in the bark, erumpent through the bark surface, 165–2 540(–2 760)  $\mu$ m diam, with 2–6 perithecia irregularly arranged. Conceptacle conspicuous. Ectostromatic disc white to buff, circular to ovoid, disc dark yellow to brown, (610–)685–985(–1 100)  $\mu$ m diam, with 2–6 ostioles arranged circularly in the disc. Ostioles dark brown to black, at the same level as the disc, occasionally the area below disc of a lighter entostroma, concentrated, circularly arranged in a disc, (128–) 145–215(–266)  $\mu$ m diam. Perithecia dark brown, flask-shaped



Fig. 19. Cytospora eastringensis (BJFC-S2038). A, B. Habit of conidiomata on twig. C. Transverse section through conidioma. D. Longitudinal section through conidioma. E. Conidiophores and conidiogenous cells. F. Conidia. Scale bars: A = 1 mm; B = 200 µm; C, D = 500 µm; E, F = 10 µm.





Fig. 20. Cytospora elaeagnina (BJFC-S2137). A–C. Habit of pseudostromata on twig. D. Transverse section through pseudostroma with ascomata. E. Longitudinal section through pseudostroma with ascomata. F. Asci. G. Ascospores. Scale bars: A = 2 mm; B–E = 500 µm; F, G = 10 µm.

to spherical, circularly arranged, (325–)390–490(–525) µm diam. Asci free, clavate to elongate-obovoid, (29.5–)33.5–39.5(–40.5) × (6.5–)7–9(–10) µm, 8-spores. Ascospores elongate-allantoid, thinwalled, hyaline, aseptate, (7.5–)8.5–9.5(–10.5) × 1.5–2 (av. = 9.1 ± 0.4 × 1.8 ± 0.2, n = 30) µm.

*Culture characteristics*: Colonies initially white and entirely covering the 9-cm-diam Petri dish after 5 d, becoming olivaceous buff to slight helical after 2 wk. The colonies are flat, felt-like, thin with a uniform texture. Conidiomata produced after 2 wk.

Notes: Cytospora elaeagnina resides in a sister clade to *C. diqingensis* but has nucleotide differences and disc colour (see notes on *C. diqingensis*). Many *Cytospora* species associated with *Elaeagnus* sp. have been reported in previous studies, *i.e. C. elaeagni, C. elaeagnicola, C. nitschkeana, C. yulinensis* (Zhang *et al.* 2013, Fan *et al.* 2014b, 2015b, Zhang *et al.* 2019). *Cytospora elaeagnina* is characterised by its ascomata with 2–6 ostioles and 8.5–9.5 × 1.5–2 µm ascospores, which sets it apart from *C. elaeagni, C. elaeagnicola* and *C. nitschkeana*. It also differs from *C. yulinensis* by having smaller ascospores (8.5–9.5 × 1.5–2 µm vs 10.7–14.8 × 2.4–2.9 µm) (Fan *et al.* 2014b, as *C. nivea*).

*Cytospora eriobotryae* Curzi & Barbaini, Arch. Labor. Bot. Critt. Univ. Pavia: 174. 1927. (Fig. 1: Clade 94)

Typus: Italy, on leaves of Eriobotrya japonica.

Description: See Fan et al. (2020) (as C. leucosperma).

*Material examined*: **India**, Saharanpur, *Eriobotrya japonica*, unknown date and *collector*, deposited by *G.C. Adams* (culture CBS 116846 = IMI 136523).

*Notes*: Fan *et al.* (2020) treated the isolates CFCC 89622 and 89894 as *C. leucosperma* based on a related host and morphology, and proposed to further study European material from the type host. However, CFCC 89622 and 89894 cluster together with CBS 116846 and were distinct from *C. leucosperma* (Clade 11) in the present study. Adams *et al.* (2005) identified CBS 116846 as *C. eriobotryae* and provided ITS sequence data. However, there is no ex-type isolate available for *C. eriobotryae*. We thus retain this name for this isolate pending future collections and typification.

Wang *et al.* (2020) used the name "*Cytospora pyri*" (sensu *V. mali* var. *pyri*) to represent this clade as it referred to a pathogenetic species on apples and pears. However, the name *Cytospora pyri* Fuckel 1860 is already occupied, even though it is regarded as synonym of *Discula pyri* (Fuckel) Höhn. Therefore, *C. eriobotryae* is the best name to apply to this clade. In ITS, *act1*, *rpb2*, *tef1-a* and *tub2* gene trees, the BI and ML phylogenies all place it as a distinct clade (ML/BI = 100/0.998, 87/0.82, 100/1, 100/1 and 100/1, respectively), within a larger clade containing *C. mali*.

Cytospora eucalypti (Cooke & Harkn.) D.P. Lawr. et al., IMA Fungus 9: 353. 2018. (Fig. 1: Clade 185)

Basionym: Valsa eucalypti Cooke & Harkn., Grevillea 9(51): 85. 1881.

Synonyms: Engizostoma eucalypti (Cooke & Harkn.) Kuntze, Rev. Gen. Plant. 3(2): 474. 1884.

Valsa eucalypti var. myrti Rolland, Bull. Soc. Mycol. France 21: 22. 1905.

Leucostoma sequoiae Bonar, Mycologia 20: 295. 1928.

*Typus*: **USA**, on dead branches of *Eucalyptus globulus*, **isotypes** UM 15128, MSC 11471 (**lectotype** designated here MSC 11471, MBT 10017464); California, Mount Tamalpais, on fallen cankered branch of *Sequoia sempervirens*, 1995, *G.C. Adams* (**epitype** designated here MSC 380714, MBT 10017457, ex-epitype culture CBS 116814).

Descriptions: See Adams et al. (2005) (as Valsa eucalypti), Lawrence et al. (2018).

Additional materials examined: **USA**, California, Mount Tamalpais, on fallen cankered branch of *Sequoia sempervirens*, 1995, G.C. Adams (MSC 380713, culture CBS 116815); Palo Alto, campus of Stanford University, on branch canker on *E. paniculata*, 2001, G.C. Adams (culture CBS 116816).

Notes: Adams et al. (2005) reduced Leucostoma sequoiae to a synonym of Valsa eucalypti based on ITS-rDNA sequence and morphology. MSC 11471 is an isotype of Valsa eucalypti (USA California, on dead branches of Eucalyptus globulus, 1880, Cooke & W.H. Harkness) while UC 469596 is the holotype of Leucostoma sequoiae (USA, Marin County, Mill Valley, Lake Lagunitas on Mt. Tamalpais, on twig of Sequoia sempervirens, 1923, L. Bonar). At this type locality, Adams et al. (2005) identified MSC 380713,

MSC 380714, with cultures CBS 116815, CBS 116814 respectively (on a fallen branch of *Sequoia sempervirens*, 1995, G.C. Adams) and MSC 380708, culture CBS 116816 (on fallen cankered branch of *E. paniculata*, 2001, G.C. Adams) as *Valsa eucalypti*. Therefore, CBS 116814 is designated here as ex-epitype. The heterotypic *Cytospora eucalypti* J.K. Sharma *et al.* has been applied in the past (Sharma *et al.* 1985), whereas no type was indicated and it is not validly published according to ICN Art. 40.1. *Valsa eucalypti* can therefore be validly combined in *Cytospora. Cytospora eucalypti* is sister species to *C. californica*, but has nucleotide differences and distinct conidial dimensions (see notes of *C. californica*).

*Cytospora eucalypticola* Van der Westh., South. Afr. For. J. 54: 10. 1965. (Fig. 1: Clade 197)

Synonyms: Valsa fabianae G.C. Adams et al., Stud. Mycol. 52: 85. 2005.

Cytospora granati D.P. Lawr. et al., IMA Fungus 9: 354. 2018.

*Typus*: **South Africa**, Tzaneen, in bark of *Eucalyptus saligna*, 1964, unknown collector (**holotype** PRE 42543); KwaZulu-Natal Province, King Cetshwayo, Kwambonambi, on large fallen branch of *Eucalyptus saligna*, 1999, G.C. Adams (**epitype** designated here MSC 380718, MBT 10017458, ex-epitype culture CBS 116852).

*Description*: See van der Westhuizen (1965), Adams *et al.* (2005) (under *Valsa fabianae* for sexual morph and culture characteristics), Lawrence *et al.* (2018) (under *Cytospora granati*).

Additional materials examined: Australia, Tasmania, on cankers of Eucalyptus nitens, 1987, K.M. Old (holotype of V. fabianae DAR 43948, culture CBS 116840); on leaves of Eucalyptus nitens, 2005, C. Mohammed (culture CBS 120064); unknown specific locality, on Eucalyptus marginata, unknown date and collector (culture CBS 118084 = ATCC 56123); Victoria, Cann River, on Eucalyptus globulus, unknown date and collector (culture CBS 118085 = CMW 6748). South Africa, Mpumalanga, Sabie, Frankfort Nursery, from water, 1989, P.W. Crous (culture CBS 115392 = CPC 210); Newcastle, Normandine plantation, on bark of advancing canker on Eucalyptus dunnii, 22 Jun. 1999, J. Roux & G.C. Adams (MSC380697, culture CBS 116851); Western Cape Province, Gordon's Bay, on twig litter of Protea neriifolia, 26 Jun. 2000, S. Marincowitz (culture CBS 122689 = CMW 20401 = CPC 13258); Malmesbury, on Eucalyptus sp., 2006, P.W. Crous (CBS H-22925 dried culture, culture CBS 141601 = CPC 12700). Uganda, Ntungamo, Ruhaama, Mishenyi-Itojo, on cankered branch of Eucalyptus grandis, Jun. 1999, J. Roux (MSC 380698, culture CBS 118088).

Notes: Cytospora eucalypticola was described by van der Westhuizen (1965) from Eucalyptus saligna in South Africa but without DNA sequence data. The isolates CBS 116840, 116851, 116852, 122689, 118084, 118085 and 118088 were identified as C. eucalypticola in the CBS database. They were sequenced to generate a five gene phylogeny. The specimen MSC 380718 (culture CBS 116852) is designated here as an epitype of this species consistent with the collection locality (South Africa) and host species (Eucalyptus saligna). Cytospora granati was first described by Lawrence et al. (2018) on Punica granatum in the USA with CBS 144237 as ex-holotype culture. The isolate CBS 144237 clusters together with C. eucalypticola in the present study. The ex-epitype culture of C. eucalypticola (CBS 116852) has the following nucleotide similarities with the sequences of the ex-holotype of C. granati (CBS 144237). In ITS, act1 and tef1-a, respectively: 500/504 (99.2 %), 238/243 (97.9 %) and 550/551 (99.8 %) (no rpb2 and tub2 sequences available for CBS 114237). Therefore, it is treated as a synonym of C. eucalypticola.

**Cytospora eugeniae** (Nutman & F.M. Roberts) G.C. Adams & Rossman, IMA Fungus 6: 147. 2015. (Fig. 1: Clade 207)

Basionym: Valsa eugeniae Nutman & F.M. Roberts, Trans. Brit. Mycol. Soc 36: 229. 1953.

*Typus*: **Tanzania**, on *Eugenia aromatica* [**holotype** IMI 44958 (as *Valsa eugeniae*)].

Description: See Adams et al. (2005).

*Materials examined*: **Australia**, Queensland, Brisbane, on *Tibouchina heteromalla*, unknown date, *M.J. Wingfield* (culture CBS 116839 = CMW 7030). **Indonesia**, on *Eucalyptus grandis*, 2003, *M.J. Wingfield* (MSC 380723, culture CBS 116834); Sulawesi, on *Eugenia* sp., 2003, M.J. Wingfield (culture CBS 116835 = CMW 8646, CBS 116836 and CBS 116837. **Malaysia**, on *Eugenia aquea*, unknown date and *collector* (culture CBS 116817 = IMI 062499).

*Notes*: The ascomata of *Cytospora eugeniae* are octosporous monostichous, with unique ostiolar beaks, which distinguishes it from other species in the genus *Cytospora* (Adams *et al.* 2005). It is distributed in Australia, Indonesia, and West Malaysia on hosts such as *Eucalyptus* spp., *Eugenia* spp. (*Myrtaceae*) and *Tibouchina* spp. (*Melastomataceae*) (Adams *et al.* 2005). In the ITS, *rpb2*, *tef1-a* and *tub2* gene trees, the BI and ML phylogenies all place *C. eugeniae* in a distinct clade, with high support (ML/BI = 100/1, 89/0.97, 84/0.999 and 100/1, respectively); however, the *act* phylogeny does not resolve it from its closest sister species, *C. castaneicola*.

*Cytospora fraxinea* X.L. Fan & C.M. Tian, *sp. nov.* MycoBank MB 850182. Fig. 21. (Fig. 1: Clade 87)

*Etymology*: Name refers to *Fraxinus*, the host genus of the holotype.

*Typus*: **China**, Beijing City, Huairou District, Forestry Centre of Beijing University of Agriculture, 40°57'23.44"N, 116°29'43.74"E, on branches of *Fraxinus chinensis* subsp. *rhynchophylla*, 18 Jun. 2021, *X.L. Fan* & *Y.K. Bai* (**holotype** BJFC-S2141, ex-holotype culture CFCC 56036); Yanqing District, Songshan National Nature Reserve, 40°29'47.24"N, 115°46'51.55"E, on twigs and branches of *Corylus heterophylla*, 4 Aug. 2021, *X.L. Fan* & *Y.K. Bai* (**paratype** BJFC-S2142, ex-paratype culture CFCC 56703).

Description: Conidiomata Group AIII (type a11), occasionally Group AII (type a9), immersed in bark, erumpent through the surface of bark when mature, conical. Conceptacle absent. Ectostromatic disc prominent, hazel to dark brown, circular to ovoid, (545–)600–850(–1020) µm diam, with one ostiole per disc. Ostiole in the centre of the disc, conspicuous, grey to black, at the same level as the disc surface, area below disc a lighter entostroma, 130–285(–325) µm diam. Locules numerous, arranged circularly with individual walls, occasionally with common walls, 1 700–2 335 µm diam. Conidiophores borne along the locules, hyaline, branched at base, in the middle or unbranched, thin-walled, 15.5–21 × 1.5–2 µm, embedded in a gelatinous layer. Conidiogenous cells enteroblastic, phialidic, subcylindrical to cylindrical. Conidia hyaline, allantoid, eguttulate, smooth, aseptate, thin-walled, (4.5–)5–5.5(–6.5) × 1–2 (av. =  $5.28 \pm 0.3 \times 1.5 \pm 0.2$ , n = 30) µm.

*Culture characteristics*: Colonies on PDA initially white, growing up to 9-cm-diam after 4 d, becoming buff after 14 d, flat and with a uniform texture. Conidiomata randomly distributed on medium surface.





Fig. 21. Cytospora fraxinea (BJFC-S2141). A, B. Habit of conidiomata on twig. C. Transverse section through conidioma. D. Longitudinal section through conidioma. E. Conidiophores and conidiogenous cells. F. Conidia. Scale bars: A = 2 mm; B–D = 500 µm; E, F = 10 µm.

Additional materials examined: China, Beijing City, Mentougou District, Baihuashan National Nature Reserve, 39°50'35.24"N, 115°26'57.89"E, on twigs and branches of *Fraxinus chinensis*, 24 Aug. 2021, *X.L. Fan* (BJFC-S2143, culture CFCC 56249); *ibid.*, Xiaolongmen Forestry Centre, 39°50'4"N, 115°34'35"E, on twigs and branches of *Fraxinus chinensis*, 25 Aug. 2022, *L. Lin & X.L. Fan* (BJFC-S2011, living culture CFCC 58439); *ibid.*, 39°50'16"N, 115°34'1"E, on twigs and branches of *Fraxinus* sp., 25 Aug. 2022, *L. Lin & X.L. Fan* (BJFC-S2012, living culture CFCC 58442).

Notes: Cytospora fraxinea is described being associated with a canker disease of *Fraxinus* sp. and *Corylus heterophylla* in China. In ITS, *act1*, *rpb2*, *tef1-a* and *tub2* gene trees, the BI and ML phylogenies all place it as a distinct clade, with high support (ML/BI = 96/0.987, 100/1, 100/1, 100/1 and 99/1, respectively). Cytospora pruinosa and C. kuanchengensis are also associated with the same host, *Fraxinus*. Morphologically, *C. fraxinea* is distinguished from *C. pruinosa* by having larger multi-locules (1 700–2 335 µm vs 690–935 µm). However, *C. kuanchengensis* has a black conceptacle and smaller, multiple locules (355–520 µm) with longer conidia (5.5–8 × 1–2 µm) as compared to *C. fraxinea* (5–5.5 × 1–2 µm).

**Cytospora fuckeliana** L. Lin, X.L. Fan & Crous, *nom. nov.* MycoBank MB 853112. (Fig. 1: Clade 108) *Replaced synonym: Valsella salicis* Fuckel, Jb. Nassau. Ver. Naturk. 23–24: 203. 1870, *non Cytospora salicis* (Corda) Rabenh., Deutschl. Krypt.-Fl. (Leipzig) 1: no. 1340. 1844. *Synonyms: Cytospora salicella* Sacc., Syll. Fung. (Abellini) 3: 260. 1884.

*Etymology*: Named in honour of the German mycologist Fuckel, in recognition of his contributions to this species.

*Typus*: **Germany**, on *Salix aurita* (Fuckel, Fungi Rhen. Exs. 2261, **isotype** S F 12679, designated as **lectotype** here, MBT 10017463). **Sweden**, Dalby Par., Jerusalem, on *Salix cinerea*, 10 Apr. 1986, *K. & L. Holm* (**epitype** designated here UPS F-133233, MBT 10017462, ex-epitype culture CBS 113699).

Notes: The type species of Valsella, V. salicis, was described from Salix aurita in Germany. According to Saccardo (1884), C. salicella is the asexual morph of V. salicis; a change of epithet was necessary because the epithet was already occupied by Cytospora salicis (Corda) Rabenh., a different species (see clade 103). However, Cytospora salicella Sacc. is younger than Valsella salicis Fuckel, which means that the name Cytospora salicella Sacc. cannot be used for the species without conservation. Therefore, Cytospora fuckeliana L. Lin, X.L. Fan & Crous is published here as an explicit substitute ("nom. nov.") for the legitimate name Valsella salicis Fuckel (Jb. Nassau. Ver. Naturk. 23-24: 203. 1870). In the present study, the two isolates CBS 109754 and 113699, both bearing the name V. salicis, reside in different clades. However, CBS 109754 differs from CBS 113699 in ITS (10/511 bp difference), act1 (20/240 bp difference), rpb2 (24/650 bp difference), tef1- $\alpha$  (72/539 bp difference) and tub2 (27/403 bp difference) loci. We apply the name C. fuckeliana to CBS 113699 based on its host and European origin. Isolate CBS 113699 was collected from the same host genus and continent as the isotype and is therefore designated as an epitype for C. fuckeliana.

*Cytospora fugax* (Bull.) Fr., Syst. Mycol. (Lundae) 2(2): 542. 1823. Fig. 22. (Fig. 1: Clade 33)

Basionym: Variolaria fugax Bull., Herb. Fr. Champ., Hist. Champ. Fr. (Paris) 1: 187. 1791.

Synonyms: Sphaeria salicina Pers., Observ. Mycol. (Lipsiae) 1: 64. 1796.

Valsa salicina (Pers.) Fr., Summa veg. Scand., Sectio Post. (Stockholm): 412. 1849.

Valsa salicina var. tetraspora Fr., Summa veg. Scand., Sectio Post. (Stockholm): 412. 1849.

Valsa socialis Ellis & Everh., Bull. Torrey Bot. Club 24: 132. 1897.

*Typus*: **France**, on bark (**lectotype** designated here, tab. 432, fig. 5 in Bull. 1789, MBT 10017548). **Switzerland**, Bouveret, on *Salix* sp., unknown date, *G. Défago* (**epitype** designated here CBS H-19181, MBT 10017465, ex-epitype culture CBS 203.42).



**Fig. 22**. *Cytospora fugax* (BJFC-S2192). **A**, **B**. Habit of pseudostromata on twig. **C**. Transverse section through pseudostroma with ascomata. **D**. Longitudinal section through pseudostroma with ascomata. **E**. Degraded asci. **F**. Ascospores. Scale bars: A = 1 mm; B = 250 µm; C, D = 500 µm; E, F = 10 µm.

Description: Pseudostromata Group SIII (type s8), immersed in bark, erumpent slightly through the bark surface, scattered, circular, (1 660–)1 770–1 950(–2 010) µm diam, with 4–8 circularly arranged perithecia. Conceptacle absent. Ectostromatic disc white, circular to ovoid, (460–)570–670(–730) µm diam, with 4–8 ostioles circularly arranged in disc. Ostioles black, (115–)130–180(–230) µm diam. Perithecia flask-shaped to spherical, (380–)480–670(– 820) µm diam. Asci free, clavate to elongate-obovoid, (35.5–)38.5– 47.5(–50) × (6–)6.5–9.5(–10) µm, 4-spored. Ascospores elongateallantoid, thin-walled, hyaline, aseptate, (20.5–)21–25.5(–26) × (4–)4.5–5.5(–6) (av. = 23 ± 0.8 × 5 ± 0.4, n = 30) µm. Asexual morph not observed.

*Culture characteristics*: Colonies white, thick, growing up to 9-cm-diam after 5 d, with felty aerial mycelium and regular edge, becoming grey olivaceous in reverse. Sterile.

Additional materials examined: China, Gansu Province, Gannan Tibetan Autonomous Prefecture, Yeliguan National Forest Park, 34°57'59.42"N, 103°40'9.49"E, on branches of *Salix matsudana*, 21 Oct. 2020, *C. Peng* & & *C.M. Tian* (BJFC-S2192, culture CFCC 56022; BJFC-S2193, culture CFCC 56023).

*Notes*: Isolate CBS 203.42, collected by G. Défago in Switzerland, was accepted as *Valsa salicina* (= *Cytospora fugax*) by Adams *et al.* (2006). The species description of Adams *et al.* (2006) was based on the fungarium specimen PREM 20469 (South Africa) listed as *Valsa salicina* by Doidge (1950), but due to several morphological differences it was questioned whether the South African material represents that species, as no cultures were available for sequencing for confirmation of conspecificity with CBS 203.42. The type is from *Salix* and *Corylus* species in Europe, but the type material could not be traced and therefore we lectotypify the species with fig. 5 of Bulliard (1789) and designate CBS H-19181 as the epitype specimen, with CBS 203.42 becoming an ex-epitype isolate in this study. Wang *et al.* (2015) treated the isolates CXY 1371 and



CXY 1381 as *C. fugax* based on ITS and *tub2* sequences data, but without a morphological description. In the present study they grouped separately in clade 34 with full support (ML/BI = 100/1) based on multigene sequences. Type studies of this clade are necessary. *Cytospora fugax* is sister species to the *Cytospora* sp. CXY 1371/1381 -*C. populina* clade (Fig. 1). However, *Cytospora fugax* (CBS 203.42) differs from *C. populina* (CFCC 89644) in ITS (2/514), *act1* (12/220), *rpb2* (24/650), and *tub2* (19/386) loci; no *tef1-a* sequence is available for CBS 203.42.

*Cytospora globosa* W.J. Li *et al.*, Fungal Diversity 100: 447. 2020. (Fig. 1: Clade 45)

*Typus*: **Italy**, Province of Forlì-Cesena, Monte Fumaiolo, on dead needles of *Abies alba*, 2 Jul. 2016, *E. Camporesi* (**holotype** MFLU 16-2054, exholotype culture MFLUCC 16-1153 = KUMCC 16-0105; *ibid.*, KUM, HKAS 97500, **isotype**).

Description: See Li et al. (2020).

Additional materials examined: **Switzerland**, Albispass, on twig of Abies alba, unknown date, *T. Sieber* (cultures CBS 118976, CBS 118977).

*Notes*: Li *et al.* (2020) described *C. globosa* with globose conidiomata. In the present study, the isolates CBS 118976 and CBS 118977 collected from *Abies alba* cluster together with MFLU 16-2054 with high support in *act1*, *rpb2*, *tef1-α* and *tub2* gene trees (ML/BI = 82/0.75, 100/1, 99/1, and 100/1, respectively). Therefore, these two isolates are identified as *C. globosa* here.

*Cytospora guyuanensis* L. Lin & X.L. Fan, *sp. nov.* MycoBank MB 850165. Fig. 23. (Fig. 1: Clade 68)

*Etymology*: Name refers to Guyuan City, Ningxia Hui Autonomous Region (China), where it was collected.

*Typus*: **China**, Ningxia Hui Autonomous Region, Guyuan City, Jingyuan County, 35°24'43.14"N, 106°23'53.77"E, on branches of *Salix* sp., 20 Oct. 2020, *C. Peng & C.M. Tian* (**holotype** BJFC-S2177, ex-holotype culture CFCC 55855; **paratype** BJFC-S2178, ex-paratype culture CFCC 56037).

Description: Conidiomata Group AI (type a4), pycnidial, immersed in bark, scattered, erumpent, discoid to conical, with a single locule. Conceptacle absent. Ectostromatic disc dark grey to black, ovoid to circular, (475–)585–650(–712) µm diam. Beak slim, spiny, 880–1 060 µm, with one ostiole per beak. Ostiole in the centre of the disc, conspicuous, 125.5–163 µm diam. Locule discoid, invaginated, single-loculed, (1 075–)1 125–1 380(–1 405) µm diam. Conidiophores borne along the locules, hyaline, branched at base, in the middle or unbranched, (11–)12–18.5(–21) × 1.5–2 µm, embedded in a gelatinous layer. Conidiogenous cells enteroblastic, phialidic, subcylindrical to cylindrical. Conidia hyaline, allantoid, eguttulate, smooth, aseptate, thin-walled, (2.5–)3–3.5(–4) × 1–1.5 (av. =  $3.2 \pm 0.2 \times 1.3 \pm 0.1$ , n = 30) µm.

*Culture characteristics*: Colonies initially white, becoming pale buff after 4 d, flat, felt-like, thin with a uniform texture, lacking aerial mycelium after 14 d. Sterile.

Additional material examined: China, Ningxia Hui Autonomous Region, Guyuan City, Jingyuan County, 35°24'43.14"N, 106°23'53.77"E, on branches of Salix sp., 20 Oct. 2020, C. Peng & C.M. Tian (BJFC-S2179, culture CFCC 56038).

*Notes: Cytospora guyuanensis* is associated with canker disease of *Salix* sp., and is phylogenetically close to *C. rostrata*, discovered from the same host in Gansu Province, China. Morphologically, *C.* 

*guyuanensis* is characterised by having a thorn-like ostiolar beak similar to *C. rostrata*, whereas *C. guyuanensis* has smaller conidia  $(3-3.5 \times 1-1.5 \,\mu\text{m})$  than *C. rostrata*  $(3.6-4.8 \times 1-1.6 \,\mu\text{m})$ ; Fan *et al.* 2014b). Phylogenetically, *C. guyuanensis* differs from *C. rostrata* by 1 bp in ITS, 12 bp in *rpb2*, 15 bp in *tef1-a* and 18 bp in *tub2* gene regions. Therefore, this species is treated as new with full support values (ML/ BI = 100/1) in the present study.

*Cytospora hoffmannii* L. Lin, X.L. Fan & Crous, *nom. nov.* MycoBank MB 853113. Fig. 24. (Fig. 1: Clade 98)

Replaced synonym: Sphaeria nivea Hoffm., Veg. Crypt. 1: 28. 1787, non Cytospora nivea (Hoffm.) Sacc., Michelia 2: 264. 1881. Synonyms: Valsa nivea (Hoffm.) Fr., Summa Veg. Scand., Section Post. (Stockholm): 411. 1849.

Cytospora nivea (Hoffm.) Sacc., Michelia 2: 264. 1881.

*Leucostoma niveum* (Hoffm.) Höhn., Mitt. Bot. Inst. Tech. Hochsch. Wien 5: 58. 1928.

*Cytospora paratranslucens* Norph. *et al.*, Mycosphere 8(1): 75. 2017.

*Etymology*: Named in honour of the German mycologist Hoffmann, in recognition of his contributions to this species.

*Typus*: **Germany**, on bark, Hoffmann (**lectotype** designated here plate VI, fig. 3 in Hoffmann (1787), MBT 10017544). **Switzerland**, on *Populus nigra*, unknown date, *G. Défago* (**epitype** designated here CBS H-19169, MBT 10017505, ex-epitype culture CBS 258.34).

Description: Conidiomata Group AII (type a7), semi-immersed in bark, erumpent when mature, discoid to conical,  $450-750 \mu m$  diam, with multi-locule. Conceptacle prominent. Ectostromatic disc white, circular,  $85-160 \mu m$  diam, with single ostiole per disc in the centre.



Fig. 23. Cytospora guyuanensis (BJFC-S2177). A, C. Habit of conidiomata on twig. B. Transverse section through conidioma. D. Longitudinal section through conidioma. E. Conidiophores and conidiogenous cells. F. Conidia. Scale bars: A = 2 mm; B–D = 500 µm; E, F = 10 µm.



**Fig. 24.** *Cytospora hoffmannii* (BJFC-S2049). **A.** Habit of conidiomata on twig. **B.** Transverse section through conidioma. **C.** Longitudinal section through conidioma. **D.** Conidiophores and conidiogenous cells. **E.** Conidia. Scale bars: A–C = 200 μm; D, E = 10 μm.

Ostiole circular to ovoid, black, 35–55 µm diam. Locules multiple, subdivided with common walls. Conidiophores hyaline, unbranched or occasionally branched, 10.5–18.5 × 1–2 µm (av. = 13.8 ± 1.3 × 1.6 ± 0.1 µm, n = 30). Conidiogenous cells enteroblastic, phialidic, subcylindrical to cylindrical, 7.5–11.5 × 1–2 µm (av. = 10.2 ± 1.1 × 1.5 ± 0.1 µm, n = 30). Conidia hyaline, unicellular, eguttulate, elongate-allantoid, 6–8 × 1.5–2 µm (av. = 7.3 ± 0.3 × 1.7 ± 0.1 µm, n = 50).

*Culture characteristics*: Colonies on PDA white, reaching 9-cmdiameter after 6 d at 25 °C, flat, felt-like, with aerial mycelium.

Additional materials examined: China, Beijing City, Yanqing District, Zhangshanying Town, 40°28'33"N, 115°49'58"E, on twigs and branches of *Populus* sp., 5 Sep. 2022, *L. Lin* (BJFC-S2049, culture CFCC 58497). **South Africa**, Western Cape Province, Central Karoo, Beaufort West, on twig of *Malus sylvestris*, unknown date, *G.C. Adams* (MSC 384990, culture CBS 118562); Mpumalanga Province, Ehlanzeni, Mbombela, on *Populus balsamifera*, 2010, unknown collector (MSC 384989, culture CBS 118565 = CMW 5250). **Switzerland**, on *Populus nigra*, unknown date, *G. Défago* (culture CBS 259.34).

Notes: Cytospora hoffmannii L. Lin, X.L. Fan & Crous is published here as an explicit substitute ("nom. nov.") for the legitimate name Sphaeria nivea Hoffm. [Veg. Crypt. 1: 28. 1787]. The heterotypic Cytospora nivea Fuckel (1860), described from Prunus padus without reference to Valsa nivea and being a synonym of Cytospora leucostoma (Fuckel 1870), predates Cytospora nivea (Hoffm.) Sacc. (1881). Therefore, a new replacement name is established in the current study. Cytospora hoffmannii (as Cytospora nivea) was originally described from Populus tremula, P. nigra, Betula alba and Prunus domestica in Europe. In absence of an extant specimen, plate VI, fig. 3 in Hoffmann (1787) provides original material for lectotypification here. Adams et al. (2002,



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2006) identified CBS 258.34 as *C. nivea* (= *Valsa nivea*). We have designated CBS H-19169 as an epitype with CBS 258.34 as ex-epitype culture in this study. *Cytospora paratranslucens* was described from *Populus alba* in Russia with the holotype MFLU 15-1986 (ex-holotype culture MFLUCC 16-0506) (Norphanphoun *et al.* 2017). However, the ex-holotype of *C. paratranslucens* (MFLUCC 16-0506) and CBS 259.34 are highly similar based on ITS (99.8 % identity, with 1 bp difference), *act* (99.5 % identity, with 1 bp differences) and *rpb2* (99.5 %, with 2 bp differences) and they cluster together with high support (ML/BI = 97/1). Therefore, *C. paratranslucens* is treated as a synonym of *C. nivea* in the current study. As *C. paratranslucens*, it displayed strong pathogenicity to *Populus alba* var. *pyramidalis* in the study of Lin *et al.* (2023a).

*Cytospora hippophaës* Thüm., Fungi Austr. Exsicc. 3: no. 282. 1872. (Fig. 1: Clade 37)

*Typus*: **Austria**, on *Hippophae rhamnoides* de Thümen, Fungi Austr. Exsicc. 282 (S-F42221).

Description: See Fan et al. (2015b).

Additional materials examined: **China**, Gansu Province, Lanzhou City, Yongdeng County, 102°49'27"E, 36°44'20"N, on branches of *Hippophaë rhamnoides*, 20 Oct. 2020, *X.L. Fan*, *N. Jiang & C. Peng* (BJFC-S2157, culture CFCC 58942; BJFC-S2158, culture CFCC 58943). **Netherlands**, Zeeland Province, Kamperland, Schotsman, on dead branches of *Hippophaë rhamnoides*, 1987, *H.A. van der Aa* (CBS H-11652, culture CBS 259.88).

*Notes: Cytospora hippophaës* has been recorded from Austria, China and Spain (Saccardo 1884, Gonzalez 1916, Zhuang 2005). In the ITS, *act1*, *rpb2*, *tef1-* $\alpha$  and *tub2* gene trees, the BI and ML phylogenies all place it in a distinct clade, with high support (ML/

BI = 66/1, 100/1, 100/1, 100/1 and 100/1, respectively). Fan *et al.* (2015b) isolated and described a *Cytospora* sp. from twigs and branches of *Hippophae rhamnoides* in China (BJFC-S779), to which they applied this name. The specimen CBS H-11652 was collected from *Hippophae rhamnoides* in the Netherlands, which represents the host and continent of the type. Therefore, CBS 259.88 can be treated as a reference strain of *C. hippophaës*.

*Cytospora italica* Thambugala *et al.*, Fungal Diversity 82: 290. 2017. (Fig. 1: Clade 177)

*Typus*: **Italy**, Province of Forlì-Cesena, Ravaldino in Monte - Forlì, on dead branches of *Tamarix gallica*, 28 Mar. 2014, *E. Camporesi*, IT 918D (**holotype** MFLU 14-0592, ex-holotype culture MFLUCC 14-0440 = ICMP 20691).

Description: See Thambugala et al. (2017).

*Material examined*: **Unknown location**, host, date and *collector* (culture CBS 112156 = CPC 3326).

*Notes*: Isolate CBS 112156 was treated as *C. acaciae* based on morphology. However, *C. acaciae* was described from *Acacia verticillata* in the Netherlands and there is no type or ex-type culture available for this species for comparison purposes. IMI 259790 was treated as *C. magnoliae*, but the type material could not be traced and therefore typification will be needed in the future. Thambugala *et al.* (2017) described isolate MFLUCC 14-0440 as a new species, *C. italica*, based on morphology and ITS and LSU sequences. In the present study, the isolates CBS 112156, IMI 259790 and MFLUCC 14-0440 clustered together with full support (ML/BI = 100/1). The isolate MFLUCC 14-0440 and CBS 112156 share identical ITS sequences. Furthermore, isolates MFLUCC 14-0440 and IMI 259790 are almost identical in ITS (99.6 % identity, with 2 bp difference). In the absence of typification of *C. acaciae* and *C. magnoliae*, this clade is treated as *C. italica*.

*Cytospora jiufengensis* L. Lin & X.L. Fan, *sp. nov.* MycoBank MB 850158. Fig. 25. (Fig. 1: Clade 150)

*Etymology*: Name refers to the Jiufeng National Forest Park where it was collected.

*Typus*: **China**, Beijing City, Haidian District, Jiufeng National Forest Park, 40°3'41.76"N, 116°5'10.62"E, on branches of *Paulownia fortunei*, 5 Jun. 2021, *M. Pan & Y.K. Bai* (**holotype** BJFC-S2180, ex-holotype culture CFCC 55839; **paratype** BJFC-S2181, ex-paratype culture CFCC 55840).

Description: Conidiomata Group AI (type a5), immersed in bark, scattered, erumpent, discoid, with a solitary undivided locule. Conceptacle absent. Ectostromatic disc isabelline to brown, nearly flat, circular to ovoid, (985–)1 010–1 310(–1 415) µm diam, with one ostiole per disc. Ostiole buff to honey, conspicuous, at the same level as the disc, 180–235 µm diam. Locules undivided, circular to ovoid, occasionally wrinkled, (1 385–)1 435–1 890(–2 235) µm diam. Conidiophores borne along the locules, hyaline, branched at base, in the middle or unbranched, or occasionally branched at base, (9.5–)10.5–22(–26.5) × 1.5–2 µm, embedded in a gelatinous layer. Conidiogenous cells enteroblastic, phialidic, subcylindrical to cylindrical. Conidia hyaline, allantoid, eguttulate, smooth, aseptate, thin-walled, 4.5–6(–6.5) × 1–1.5 (av. =  $5.3 \pm 0.5 \times 1.4 \pm 0.1$ , n = 30) µm.

*Culture characteristics*: Colonies initially white, growing radially and covering the 9-cm-diam Petri dish and becoming olivaceous buff after 7 d, becoming grey olivaceous after 14 d. Sterile.

*Notes*: *Cytospora jiufengensis* is associated with canker disease of *Paulownia fortunei* in China, representing the first report of a *Cytospora* from this host. The clear phylogenetic distinction resulted in our decision to describe this species as new based on sequence data and morphology. In the combined analysis, the most closely related species to *C. jiufengensis* are *C. uniloculata* and *C. kuanchengensis*. The two isolates currently available have the following nucleotide differences from the sequences for *C. uniloculata* and *C. kuanchengensis*: in ITS: 5/514 bp and 5/469 bp, respectively; in *act*: 27/248 bp and 27/247 bp, respectively; in *rpb2*: no *rpb2* available for *C. uniloculata* and 29/716, respectively; in *tef1-a*: 78/307 bp and no *tef1-a* available for *C. kuanchengensis*. Morphologically, conidial size ranges distinguish the three species from each other.



Fig. 25. Cytospora jiufengensis (BJFC-S2180). A, B. Habit of conidiomata on twig. C. Transverse section through conidioma. D. Longitudinal section through conidioma. E, F. Conidiophores and conidiogenous cells. G. Conidia. Scale bars: B–D = 500 μm; E–G = 10 μm.

Conidia are  $4.5-6 \times 1-1.5 \mu m$  in *C. jiufengensis*,  $5.5-8 \times 1-2 \mu m$  in *C. kuanchengensis* and  $8-9.5 \times 2-3 \mu m$  in *C. uniloculata* (Jiang *et al.* 2020b). Additionally, this species can be easily distinguished from *C. kuanchengensis* by conidiomata with a solitary undivided locule and the absence of a conceptacle (Jiang *et al.* 2020b).

*Cytospora junipericola* Senan. *et al.*, Stud. Mycol. 86: 242. 2017. (Fig. 1: Clade 188)

Synonyms: Cytospora quercicola Senan. et al., Stud. Mycol. 86: 244. 2017.

*Cytospora fraxinigena* Senan. *et al.*, Stud. Mycol. 86: 242. 2017. *Cytospora rosae* Senan. *et al.*, Stud. Mycol. 86: 244. 2017.

*Typus*: **Italy**, Province of Forlì-Cesena, Santa Sofia, near Cabelli, on dead branch of *Juniperus communis*, 13 Jan. 2014, *E. Camporesi*, IT 1643 (**holotype** MFLU 17-0882; **isotype** BBH42444).

Description: See Senanayake et al. (2017).

*Materials examined*: **France**, Pouzols Minervois, La Pyramide, on dead branch of *Cupressus sempervirens*, 1996, *H.A. van der Aa* (culture CBS 654.97). **Italy**, unknown specific locality, *Thuja* sp., unknown date, *R. Ciferri* (culture CBS 196.50); Milano, on trunk of *Castanea sativa*, 1980, *M. Bisiach* (culture CBS 216.81). **Netherlands**, unknown specific locality, on dead wood, 2019, *N. Jiang* (BJFC-S2105, culture CFCC 54017). **Spain**, unknown specific locality, on decaying twig of *Quercus ilex*, 1996, *R.F. Castañeda* (culture CBS 196.97).

Notes: Senanayake et al. (2017) introduced C. guercicola, C. fraxinigena, C. junipericola and C. rosae from Quercus sp. (Fagaceae), Fraxinus ornus (Oleaceae), Juniperus communis (Cupressaceae) and Rosa canina (Rosaceae), respectively. Unfortunately, none of these species have act1, rpb2, tef1- $\alpha$ , or tub2 sequence data available for reference purposes, apart from C. junipericola having a tef1- $\alpha$  sequence. In the present study, they grouped together in the multi-gene phylogenetic tree (Fig. 1). The ex-holotype culture of C. junipericola (MFLU 17-0885) has the following nucleotide similarities with the ITS sequence data: 503/504 bp (99.8 %) with C. quercicola (MFLUCC 14-0867), 503/504 bp (99.8 %) with C. fraxinigena (MFLUCC 14-1868) and 502/504 bp (99.6 %) with C. rosae (MFLU 17-0885). It is recommended to classify them as belonging to the same species, unless further multi-gene data are added and it is determined that they differ significantly. Cytospora junipericola is the best name for this group as it is the species for which the most DNA sequence data are available.

Isolate CBS 196.50 was identified as *C. cedri* by Adams *et al.* (2006) based on ITS sequence data. However, there is no evidence for that identification, as the host and location of CBS 196.50 are highly different from the type of *C. cedri* (India, on *Cedrus libani* var. *deodara*, F46877). The type specimen of *C. cedri* has no sequence data to reveal its phylogenetic position. Therefore, we have treated CBS 196.50 as *C. junipericola*. However, recollection of *C. cedri* (Himachal Pradesh, on branch of *Cedrus libanus* var. *deodara*.) is needed to clearly resolve this question.

*Cytospora kantschavelii* Gvrit., Mikol. Fitopatol. 7: 547. 1973. (Clade 76)

Synonym: Cytospora parakantschavelii Norph. et al., Mycosphere 8(1): 70 (2017).

Typus: Georgia, on Populus nigra. Needs typification.

*Descriptions*: See See Gvritishvili (1973), Norphanphoun *et al.* (2017, as *C. parakantschavelii*).

*Materials examined*: **China**, Yunnan Province, Dali City, Xiaguan District, 25°36'58"N, 100°12'37"E, on branches of *Populus × canadensis*, 4 Aug. 2022, *L. Lin & M. Lin* (BJFC-S2013, culture CFCC 58194); Kunming City, Panlong District, Southwest Forestry University, 102°45'22"E, 25°3'43"N, on branches of *Populus × canadensis*, *L. Lin & Z.Q. Wu*, 11 Aug. 2022 (BJFC-S2014, culture CFCC 58205; BJFC-S2015, culture CFCC 58213; BJFC-S2016, culture CFCC 58219); Beijing City, Daxing District, Gusang National Forest Park, 39°38'1"N, 116°32'40"E, on twigs and branches of *Populus* sp., 4 Aug. 2022, *Y.K. Bai & M. Pan* (BJFC-S2017, culture CFCC 59071). **Italy**, Umbria, Perugia, on twig of *Salix viminalis*, 1963, deposited by *M. Ribaldi* (culture CBS 485.63).

Notes: Norphanphoun et al. (2017) described C. parakantschavelii as a new species and as sister clade to C. kantschavelii (287-2). However, in the current phylogeny it clustered with the C. kantschavelii strain 287-2 isolated from Populus deltodies in Iran (Fotouhifar 2007, 2010) and strains CXY1386 from P. maximowiczii in China (Wang et al. 2015). CBS 485.63 was deposited as V. salicina in the CBS collection based on morphology. However, in the current phylogeny it clusters with the C. kantschavelii strain 287-2. Cytospora kantchavelii was described by Gvritishvili (1973) from branches of Populus nigra in Georgia. Our isolates clusters in a clade with high support (ML/BI=92/1) (Fig. 1). However, in the present study, the ex-holotype strain of C. parakantschavelii (MFLUCC 15-0857) clusters together with C. kantschavelii. The isolate CXY 1386 and MFLUCC 15-0857 (the ex-holotype of C. parakantschavelii) have identical ITS sequences. They are also similar in having multiloculate conidiomata (Norphanphoun et al. 2017). Therefore, C. parakantchavelii is regarded as a synonym of C. kantschavelii.

Cytospora kunzei (Kunze) Sacc., Syll. Fung. (Abellini) 3: 270. 1884. (Fig. 1: Clade 162)

*Basionym: Sphaeria kunzei* Kunze, Mykologische Hefte (Leipzig) 2: 45. 1823.

Synonyms: Leucocytospora kunzei (Sacc.) Z. Urb., Rozpr. Českoslov. Akad. Véd. Ročn. 68 (Ses. 12): 74. 1958.

Sphaeria kunzei Fr., Mykologische Hefte (Leipzig) 2: 45. 1823.

*Valsa kunzei* (Fr.) Fr., Summa veg. Scand., Sectio Post. (Stockholm): 411. 1849.

Leucostoma kunzei (Fr.) Munk, Dansk bot. Ark. 15(no. 2): 80. 1953.

Typus: Europe, on Abies alba.

Description: See Adams et al. (2006).

*Materials examined*: **USA**, Illinois, Champaign, on *Picea pungens*, 1986, *Schoeneweiss* (culture CBS 114651 = UPSC 3149); Michigan, on *Picea pungens*, 1987, *T. Proffer* (cultures CBS 118093 = ATCC 64880, CBS 118094 = ATCC 64881).

*Notes: Cytospora kunzei* is an aggressive pathogen of *Pinus elderica* in Turkey (Kavak 2005) and *Picea pungens* in North America (Proffer & Hart, 1988). It has leucocytosporoid conidiomata (Group AII, type a7) and octosporous leucocircinate ascomata (Group SII, type a5) (Adams *et al.* 2006). There is no type or extype culture available for this species. In ITS, *act1*, *rpb2*, *tef1-α* and *tub2* gene trees, the BI and ML phylogenies all place it as a distinct clade, with high support (ML/BI = 99/1, 100/1, 98/1, 100/1 and 100/1, respectively). We have retained these strains in the present study as *C. kunzei* pending future typification.



*Cytospora lauricola* L. Lin & X.L. Fan, *sp. nov.* MycoBank MB 850159. Fig. 26. (Fig. 1: Clade 73)

*Etymology*: Name refers to the host genus, *Laurus* from which it was collected.

*Typus*: **China**, Yunnan Province, Diqing Tibetan Autonomous Prefecture, Shangri-La City, Sanba Naxi Town, East Ring Road, 27°34'26"N, 100°1'5"E, on branches of *Laurus nobilis*, 9 Aug. 2022, *L. Lin & M. Lin* (**holotype** BJFC-S2020, ex-holotype culture CFCC 58221; **paratype** BJFC-S2018, ex-paratype culture CFCC 58193).

Description: Conidiomata Group AII (type a6), immersed in bark, erumpent when mature, discoid to conical, 900–1 085 µm diam, with multi-locule. Conceptacle absent. Ectostromatic disc buff to black, circular to ovoid, 300–385 µm diam. Ostiole inconspicuous. Locules divided with shared walls. Conidiophores hyaline, unbranched or occasionally branched at the bases, 13–27.5 × 1–2 µm (av. = 18.4 ± 2.9 × 1.3 ± 0.2 µm, n = 30). Conidiogenous cells enteroblastic, phialidic, subcylindrical to cylindrical, 2.5–4 × 0.8–1.5 µm (av. = 3.3 ± 0.4 × 1.0 ± 0.1 µm, n = 30). Conidia hyaline, unicellular, eguttulate, elongate-allantoid, 3.5–5 × 1–1.5 µm (av. = 4.2 ± 0.3 × 1.3 ± 0.1 µm, n = 50).

*Culture characteristics*: Cultures on PDA initially white, growing up to 7 cm diam after 3 d, entirely covering the 9-cm-diam Petri dish after 4 d and becoming buff after 20 d, flat with a uniform texture.

Additional material examined: China, Yunnan Province, Diqing Tibetan Autonomous Prefecture, Shangri-La City, Sanba Naxi Town, East Ring Road, 27°34'26"N, 100°1'5"E, on branches of *Laurus nobilis*, 9 Aug. 2022, *L. Lin & M. Lin* (BJFC-S2019, culture CFCC 58208).

*Notes*: The currently available three isolates are associated with canker disease of *Laurus nobilis* in Yunnan, China. Phylogenetically, *C. lauricola* is closely related to *C. lijiangensis* but differs in ITS (3/512 bp), *act1* (4/201 bp), *rpb2* (5/705 bp), *tef1-* $\alpha$  (30/266 bp) and *tub2* (14/449 bp) loci. Morphologically, conidial size ranges

distinguish them ( $3.5-5 \times 1-1.5 \mu m$  in *C. lauricola* and  $5.5-7.5 \times 1.5-2.5 \mu m$  in *C. lijiangensis*). *Cytospora lauri* is also associated with the same host genus, *Laurus* (Kirk 1981), but without sequence data. *Cytospora lauricola* is distinguished from *C. lauri* by its wider conidia ( $3.5-5 \times 1-1.5 \mu m vs 4-5 \times 0.7-1 \mu m$ ) (Grove 1937). Therefore, it is described here as a new species.

*Cytospora leucosperma* (Pers.) Fr., Syst. Mycol. (Lundae) 2(2): 543. 1823. Fig. 27. (Fig. 1: Clade 11)

Basionym: Naemaspora leucosperma Pers., Observ. mycol. (Lipsiae) 1: 81. (1796).

Valsa ambiens (Pers.) Fr., Summa veg. Scand., Sectio Post. (Stockholm): 412. 1849.

Synonyms: Cytospora galegicola Q.J. Shang et al., Mycosphere 11: 206. 2020.

Cytospora hippophaicola Spetik et al., Persoonia 47: 299. 2021.

*Typus*: **Sweden**, Fries, Scler. Suec. 156 (UPS F-127268, **lectotype** selected by Spielman 1985)

Description: Conidiomata Group AII (type a6, a9), immersed in bark, erumpent when mature, discoid to conical, 670–1 005 µm diam, with multi-locule. Conceptacle absent. Ectostromatic disc isabelline to umber, circular to ovoid, 150–230 µm diam, with single ostiole per disc in the centre. Ostiole circular to ovoid, honey to isabelline, 65–100 µm diam. Locules numerous, divided with shared walls. Conidiophores hyaline, unbranched or branched at the middle, 11–22.5 × 1–2 µm (av. = 14.7 ± 3.0 × 1.5 ± 0.2 µm, n = 30). Conidiogenous cells enteroblastic, phialidic, subcylindrical to cylindrical, 3–5 × 1–1.5 µm (av. = 4.2 ± 0.4 × 1.3 ± 0.18 µm, n = 30). Conidia hyaline, unicellular, eguttulate, elongate-allantoid, 3.5–6 × 1–2 µm (av. = 4.6 ± 0.5 × 1.5 ± 0.15 µm, n = 50). For the description of the sexual morph, see Spielman 1985.

*Materials examined*: **Austria**, Niederösterreich, on branches of *Fagus sylvatica*, 2000, *W. Jaklitsch* (CBS H-22693, culture CBS 109491=WJ1607); *ibid*. (culture CBS 141469). **Germany**, Braunschweig, on branches of *Fagus* 



**Fig. 26.** *Cytospora lauricola* (BJFC-S2020). **A**, **B**. Habit of conidiomata on twig. **C**. Transverse section through conidioma. **D**. Longitudinal section through conidioma. **E**, **F**. Conidiophores and conidiogenous cells. **G**. Conidia. Scale bars: B = 100 μm; C, D = 200 μm; E–G = 10 μm.


Fig. 27. Cytospora leucosperma (BJFC-S2022). A, B. Habit of conidiomata on twig. C. Transverse section through conidioma. D. Longitudinal section through conidioma. E, F. Conidiophores and conidiogenous cells. G. Conidia. Scale bars: A = 1 mm; B–D = 200 µm; E–G = 10 µm.

sylvatica, 1987, A. Wulf (culture CBS 345.87). Switzerland, Monthey, on branches of Fagus sylvatica, G. Défago (culture CBS 202.42). Netherlands, Baarn, Groeneveld, on dead branches of Betula verrucosa, unknown date, W. Loerakker (culture CBS 283.74); ibid., on branches of Rhododendron sp., 1981, H.A. van der Aa (culture CBS 280.81); ibid., on leaf of Rhododendron ponticum, unknown date and collector (culture CBS 117.67); unknown specific locality, on branches of Alnus sp., 2019, N. Jiang (BJFC-S2024, culture CFCC 54018; BJFC-S2021, culture CFCC 54031); on branches of Quercus sp., 2019, N. Jiang (BJFC-S2022, culture CFCC 54029; BJFC-S2026, culture CFCC 54024); on dead wood, 2019, N. Jiang (BJFC-S2025, culture CFCC 54023; BJFC-S2023, culture CFCC 54027, 54028; BJFC-S2027, CFCC 54029). UK, on branches of Fagus sylvatica, unknown date and collector (culture CBS 423.52). USA, New York, Syracuse, Oakwood cemetery, on twigs of Betula sp., unknown date and collector (culture CBS 141617 = CPC 28401 = LCM 167.01); on twigs of Betula sp., unknown date, L.C. Mejía (culture CBS 141642 = CPC 28405 = LCM 169.02).

*Notes: Cytospora leucosperma* (syn. *Valsa ambiens*) is the type species of the sexual genus *Valsa*, which was re-described by Adams *et al.* (2005). Urban (1957) and Spielman (1985) designated PR 163781 from *Tilia* as neotype of *Valsa ambiens*, but without any available culture or DNA sequence data. Spielman (1985) selected UPS F-127268 as lectotype of *Cytospora leucosperma*. Fan *et al.* (2020) provisionally treated CFCC 89622 and 89894 on *Pyrus* sp. from China as *C. leucosperma* as it matched with the host records of *C. leucosperma* in China. However, CFCC 89622 and 89894 clustered apart in the phylogenetic tree.

The isolates CBS 345.87, 423.52, 109491 and 141469 are treated as *C. leucosperma* in the present study. Additionally, CBS 345.87 was collected on *Fagus sylvatica* from Germany, which is located geographically nearby and serves as a closely related host to those recorded by Spielman in 1985 (Saccardo 41, on *Juglans*, France). Thus, clade 11 in the present study is best suited to being treated as *C. leucosperma*. *Cytospora galegicola* and *C. hippophaicola* were published in 2020 and 2021, respectively (Shang *et al.* 2020, Crous *et al.* 2021). Therefore, they are treated as synonyms of *C. leucosperma* in the present study.

Cytospora leucostoma (Pers.) Sacc., Michelia 2(7): 264. 1881. (Fig. 1: Clade 122)

Basionyms: Sphaeria leucostoma Pers., Ann. Bot. 11: 23. 1794. Synonyms: Valsa leucostoma (Pers.) Fr., Summa veg. Scand., Section Post. (Stockholm): 411. 1849. Valsa persoonii Nitschke, Pyrenomyc. Germ. 2: 222. 1870. Leucostoma persoonii (Nitschke) Höhn., Mitt. Bot. Inst. Tech. Hochsch. Wien 5: 78. 1928. Cytospora erumpens Norph. et al., Mycosphere 8: 64. 2017.

Cytospora nivea Fuckel, Jb. nassau. Ver. Naturk. 15: 51 (1860).

Typus: Requires typification.

Description: See Fan et al. (2020) (under C. erumpens).

Additional material examined: **Netherlands**, Abcoude, Botshol, on Sorbus aucuparia, 27 Dec. 1975, *M.J.C. Lips* (CBS H-14029, culture CBS 133.76); on dead wood, 2019, N. Jiang (BJFC-S2101, culture CFCC 54021).

Notes: Cytospora leucostoma is a common species associated with a canker disease on Rosaceae hosts. Specimens residing in clade 122 (BJFC-S1064, culture CFCC 50022) have typical leucostomalike conidiomata (Fan et al. 2020). Isolate CBS 133.76 in clade 116 was identified as V. leucostoma by Adams et al. (2005) and is listed in the CBS database as Leucostoma persoonii. According to Saccardo (1882), C. leucostoma is the asexual morph of V. leucostoma, while V. leucostoma and V. persoonii are synonyms. Sphaeria leucostoma (the basionym of C. leucostoma) was originally described from Prunus cerasus, but no type material could be traced. Isolate CBS 133.76 has same host family (Rosaceae) and continent (Europe) as that of the original specimen. Therefore, CBS 133.76 can be treated as a reference strain of C. leucostoma. *Cytospora erumpens* introduced by Norphanphoun *et al.* (2017) from Salix × fragilis in Russia (ex-holotype MFLUCC 16-0580) is treated as synonym of C. leucostoma, as it groups together with CBS 113.76.

Strains residing in clade 111 in the present study were previously identified as *C. leucostoma* by Fan *et al.* (2020). Although there are many strains residing in clade 111 occurring on *Rosaceae*, they have all been collected in China and do not correspond to the original collection location of *C. leucostoma*.

Cytospora Ihasaensis L. Lin & X.L. Fan, sp. nov. MycoBank MB 850192. Fig. 28. (Fig. 1: Clade 70)

*Etymology*: Name refers to Lhasa City (China) where it was collected.





Fig. 28. Cytospora Ihasaensis (BJFC-S2100). A, B. Habit of conidiomata on twig. C. Transverse section through conidioma. D. Longitudinal section through conidioma. E, F. Conidiophores and conidiogenous cells. G. Conidia. Scale bars: A = 1 mm; B = 100 µm; C, D = 500 µm; E–G = 10 µm.

*Typus*: **China**, Tibet Autonomous Region, Lhasa City, Mozhugongka County, Zen Town, 29°42′23″N, 92°6′40″E, on branches of *Salix wallichiana*, 20 Aug. 2022, Y.Y. *Liu, M. Liu, N. Jiang, & P. Jin* (**holotype** BJFC-S2100, ex-holotype culture CFCC 58706).

Description: Conidiomata Group AIII (type a11), immersed in bark, erumpent when mature, discoid to conical, 610–915 µm diam, with multi-locule. Conceptacle absent. Ectostromatic disc mouse grey to black, circular to ovoid, 200–260 µm diam, with single ostiole per disc in the centre. Ostiole circular to ovoid, black, 75–85 µm diam. Locules independent, sharing one ostiole. Conidiophores hyaline, unbranched or branched, 14–27.5 × 1–2 µm (av. = 18.8  $\pm$  3.0 × 1.5  $\pm$  0.2 µm, n = 30). Conidiogenous cells enteroblastic, phialidic, subcylindrical to cylindrical, 3.5–5 × 1–1.5 µm (av. = 4.2  $\pm$  0.4 × 1.2  $\pm$  0.1 µm, n = 30). Conidia hyaline, unicellular, eguttulate, elongate-allantoid, 3.5–5 × 1–1.5 µm (av. = 4.4  $\pm$  0.3 × 1.4  $\pm$  0.08 µm, n = 50).

Notes: Phylogenetically, *C. Ihasaensis* is closely related to *C. suecica*. However, *C. Ihasaensis* (CFCC 58706) differs from *C. suecica* (CBS 450.51) in ITS (9/512), *act1* (14/253), *rpb2* (13/650), *tef1-a* (35/491) and *tub2* (29/422) loci. Morphologically, they are different in conidial size (3.5–5 × 1–1.5 µm in *C. Ihasaensis* and 0.5–2 × 0.5–1 µm in *C. suecica*). Additionally, *C. suecica* has a flat colony lacking aerial mycelium, whereas *C. Ihasaensis* has aerial mycelium.

Cytospora lijiangensis L. Lin & X.L. Fan, sp. nov. MycoBank MB 850160. Fig. 29. (Fig. 1: Clade 72)

Etymology: Name refers to Lijiang City where it was collected,

*Typus*: **China**, Yunnan Province, Lijiang City, Gucheng District, Yuze East Road, 100°12'2"E, 26°52'57"N, on branches of *Populus* × *canadensis*, 5 Aug. 2022, *L. Lin & M. Lin* (**holotype** BJFC-S2030, ex-holotype culture CFCC 58483); Dali City, Xiaguan District, 25°39'9"N, 100°10'50"E, on branches of *Populus* × *canadensis*, 4 Aug. 2022, *L. Lin & M. Lin* (**paratype** BJFC-S2028, ex-paratype culture CFCC 58209). Description: Pseudostromata Group SII (type a3), immersed in the bark, erumpent slightly through the bark surface, scattered, circular, 910-1 275 µm diam, with 2-4 perithecia arranged circularly. Conceptacle absent. Ectostromatic disc primrose, ovoid to rhomboid, 280-500 µm diam, with ostioles arranged circularly per disc. Ostioles black, 65-80 µm diam. Perithecia flask-shaped to spherical, 255-325 µm diam. Asci free, clavate to elongateobovoid, 52.5–59 ×10.5–13.5 (av. = 55.7  $\pm$  0.7 × 11.6  $\pm$  0.6, n = 30) µm, 8-spored. Ascospores biseriate, elongate-allantoid, thinwalled, hyaline, aseptate,  $14.5-20.5 \times 5-6.5$  (av. =  $16.8 \pm 0.4 \times$ 6.1 ± 0.3, n = 50) μm. Conidiomata Group All (type a6), pycnidial, scattered or serried, immersed in bark, erumpent through the bark surface, discoid to conical, with multiple locules. Conceptacle absent. Ectostromatic disc ochreous to dark mouse grey, circular, 230-450 µm diam, with one ostioles per disc. Ostiole grey to black, 40-55 µm diam. Locules numerous, arranged circularly with common walls. Conidiophores borne along the locules, hyaline, unbranched or occasionally branched at the base,  $13.5-20.5 \times 1-2$  $(av. = 17.4 \pm 2.1 \times 1.6 \pm 0.2, n = 30) \mu m$ , embedded in a gelatinous layer. Conidiogenous cells enteroblastic, phialidic, subcylindrical to cylindrical,  $4.5-7 \times 1-2$  (av. =  $5.2 \pm 0.5 \times 1.4 \pm 0.2$ , n = 30) µm. Conidia hyaline, allantoid, thin-walled, eguttulate, aseptate, smooth,  $5.5-7.5 \times 1.5-2.5$  (av. =  $6.5 \pm 0.5 \times 2.0 \pm 0.1$ , n = 30)  $\mu$ m.

Additional material examined: China, Diqing Tibetan Autonomous Prefecture, Weixi Lisu Autonomous County, Yezhi Town, 27°49'37"N, 99°2'38"E, on branches of *Populus × canadensis*, 10 Aug. 2022, *L. Lin & M. Lin* (BJFC-S2031, culture CFCC 58488); Beijing City, Mentougou District, Xiaolongmen Forestry Centre, 39°50'5"N, 115°34'35"E, on twigs and branches of *Salix matsudana*, 25 Aug. 2022, *L. Lin & X.L. Fan* (BJFC-S2029, culture CFCC 58453); *ibid.*, Fengtai District, Beigong National Forest Park, 39°51'42"N, 116°7'8"E, on twigs and branches of *Acer truncatum*, Aug. 2022, *Y.K. Bai & M. Pan* (BJFC-S2032, culture CFCC 59069); *ibid.*, Changping District, Xinchengbinhe National Forest Park, 40°7'47.741"N, 116°18'43.942"E, on twigs and branches of *Cotoneaster horizontalis*, 3 Jul. 2022, *Y.K. Bai & M. Pan* (BJFC-S2033, culture CFCC 59074).

Notes: Fan et al. (2020) introduced C. euonymina with the isolates CFCC 89993 (as ex-holotype culture) and CFCC 89999. Although



**Fig. 29.** *Cytospora lijiangensis* (BJFC-S2030). **A, B.** Habit of conidiomata on twig. **C.** Transverse section through conidioma. **D.** Longitudinal section through conidioma. **E.** Habit of pseudostroma on twig. **F.** Transverse section through pseudostroma with ascomata. **G.** Longitudinal section through pseudostroma with ascomata. **H.** Conidiophores and conidiogenous cells. **I.** Conidia. **J.** Ascus. **K.** Ascospores. Scale bars: A = 1 mm; B = 100 μm; C, E–G = 200 μm; D = 500 μm; H–K = 10 μm.

these two isolates clustered together in the study of Fan *et al.* (2020) (MP/ML/BI = 76/59/0.98), they differ in in *act1* (4/220 bp), *rpb2* (9/726 bp) and *tef1-a* genes (25/304 bp). In the present study, the isolate CFCC 89999 and an additional six isolates formed a distinct cluster with CFCC 89993 with high support (ML/ BI = 94/1) (Fig. 1). Therefore, a new species *C. lijiangensis* is proposed to represent the clade accommodating CFCC 89999 with BJFC-S2030 as the holotype and CFCC 58483 as the ex-holotype culture. Morphologically, *C. lijiangensis* is distinguished from *C. euonymina* by its shorter conidiogenous cells (4.5–7 × 1–2 µm vs 8.5–12 × 1.5–2 µm).

Cytospora Ivxinensis L. Lin & X.L. Fan, sp. nov. MycoBank MB 850168. Fig. 30. (Fig. 1: Clade 175)

*Etymology*: Name refers to the City Green Heart Forest Park, 'Lvxin (lu xīn)' in Chinese, where the holotype specimen was collected.

*Typus*: **China**, Beijing City, Tongzhou District, the City Green Heart Forest Park, 39°53'2.92"N, 116°43'42.90"E, on branches of *Salix* sp., 8 Sep. 2021, *M. Pan & X.L. Fan* (**holotype** BJFC-S2173, ex-holotype culture CFCC 56780); 39°52'59.87"N, 116°43'12.59"E, on branches of *Ziziphus jujuba*, 8 Sep. 2021, *M. Pan & X.L. Fan* (**paratype** BJFC-S2174, exparatype culture CFCC 56803).

Description: Conidiomata Group AII (type a9), pycnidial, scattered or serried, immersed in bark, erumpent through the bark surface, discoid, with multiple locules. Conceptacle absent. Ectostromatic disc black, circular, (170–)230–370(–750) µm diam, with one ostiole per disc. Ostiole grey to black, (85–)130–215(–260) µm diam. Locules numerous, arranged circularly with common walls, (560–)600–720(–850) µm diam. Conidiophores borne along the locules, hyaline, branched at the base, in the middle or occasionally unbranched, (12–)15.5–22.5(–26) × 1–2 µm, embedded in a gelatinous layer. Conidiogenous cells enteroblastic, phialidic, subcylindrical to cylindrical. Conidia hyaline, allantoid, thin-walled, eguttulate, aseptate, smooth, (4–)4.5–6(–6.5) × 1–1.5 (av. = 5.3 ± 0.5 × 1.3 ± 0.2, n = 30) µm.



*Culture characteristics*: Colonies white, growing up to 8 cm diam at 7 d and producing slightly yellow pigment with a uniform texture after 2 wk, ultimately lacking aerial mycelium, thick with a regular edge. Conidiomata randomly distributed on medium surface.

*Notes*: *Cytospora Ivxinensis* is phylogenetically close to *C. italica* and *C. phitsanulokensis* and can be distinguished from *C. phitsanulokensis*, its closest relative in lacking a conceptacle, locules with common walls, and having smaller conidia (4.5–6 × 1–1.5 µm vs 5.2–6.9 × 1.2–1.7 µm, Monkai *et al.* 2021) and sequence differences (11/440 bp in *tef1-α* and 4/715 bp in *tub2*). *Cytospora Ivxinensis* differs from *C. italica* isolated from *Tamarix gallica* by its longer conidia (4.5–6 × 1–1.5 µm vs 3.7–5.2 × 1–1.3 µm) (Thambugala *et al.* 2016).

*Cytospora mali* Grove, British Stem and Leaf Fungi (Coelomycetes) (Cambridge) 1: 279. 1935. (Fig. 1: Clade 93) *Synonym: Valsa mali* Miyabe & G. Yamada, M. Miura Agr. Exp. Stn Bull. 4: 17. 1915.

*Typus*: **UK**, on twigs of *Malus domestica*.

Description: See Fan et al. (2020).

*Materials examined*: **China**, Beijing City, Yanqing District, Duck Lake Wetland Park, 40°23'41.089"N, 115°50'25.314"E, on twigs and branches of *Malus spectabilis*, 3 Jul. 2022, *Y.K. Bai & X.L. Fan* (BJFC-S2034, culture CFCC 59059). **Japan**, unknown specific locality, on *Eucalyptus grandis*, unknown date, *Y. Koboyashi* (culture CBS 117012 = ATCC 56632). **Russia**, Primorsky Territory, Vladivostok, on *Malus* sp., 2000, *A.Y. Rossman* (culture CBS 109499).

*Notes*: In the ITS gene trees, the ML phylogeny places it as a distinct clade with high support (ML = 96), while the BI phylogeny do not resolve *C. mali* from its closest sister species, *C. eriobotryae*. In *act1, rpb2, tef1-a* and *tub2* gene trees, the BI and ML phylogenies all place it as a distinct clade with high support (ML/BI = 96/1, 100/1, 82/1 and 98/1, respectively) within a larger clade containing *C.* 



Fig. 30. Cytospora lvxinensis (BJFC-S2173). A, B. Habit of conidiomata on twig. C. Transverse section through conidioma. D. Longitudinal section through conidioma. E. Conidiophores and conidiogenous cells. F. Conidia. Scale bars: A = 1 mm; B–D = 500 μm; E–F = 10 μm.

*eriobotryae.* The name *Valsa mali* Miyabe & G. Yamada is older than *Cytospora mali* Grove and they are commonly considered synonymous. As Grove (1935) made no connection with *Valsa mali*, his *Cytospora mali* cannot be treated as a new combination. It is not possible to introduce a new combination, as the epithet is already occupied in *Cytospora*. It is preferable to preserve the name and the synonym, *Cytospora mali* Grove is thus retained here. The use of the name *Cytospora mali* will require conservation following an investigation of the type specimens.

*Cytospora malvicolor* X.L. Fan & C.M. Tian, *sp. nov.* MycoBank MB 850161. Fig. 31. (Fig. 1: Clade 23)

*Etymology*: Name refers to the distinctive colour of the colonies. Latin *malvicolor* (mauve).

*Typus*: China, Beijing City, Huairou District, Forestry Centre of Beijing University of Agriculture, 39°59'12.54"N, 115°26'17.08"E, on branches of *Corylus mandshurica*, 19 Jun. 2021, *X.L. Fan & Y.K. Bai* (holotype

BJFC-S2120, ex-holotype culture CFCC 56577; **paratype** BJFC-S2121, ex-paratype culture CFCC 56567).

Description: Pseudostromata Group SII (type s3), immersed in the bark, erumpent through the bark surface, scattered, circular to ovoid, (700-)890-1 340(-1 500) µm diam, with 3-4 perithecia arranged circularly in well-developed black entostromata. Conceptacle absent. Ectostromatic disc brown to black, circular to ovoid, (320-)380-420(-520) µm diam, with 3-4 ostioles arranged circularly per disc. Ostioles brown to black, (90-)100-135(-150) µm diam. Perithecia flask-shaped to spherical, (260-)290-320(-470) µm diam. Asci free, clavate to elongate-obovoid, (45.5-)52-58(-70.5) × (10.5-)11.5-12(-13.5) µm, 8-spored. Ascospores biseriate, elongate-allantoid, thin-walled, hyaline, aseptate, (14.5-)  $15.5-17.5(-18.5) \times (3.5-)4-4.5(-5)$  (av. =  $16.5 \pm 1.0 \times 4.3 \pm 0.3$ , n = 30) µm. Conidiomata Group All (type a7), pycnidial, scattered, immersed in bark, erumpent through the bark surface, discoid, with multiple locules. Conceptacle black. Ectostromatic disc grey to dark, circular to ovoid, (400-)430-490(-520) µm diam, with 1-3



Fig. 31. Cytospora malvicolor (BJFC-S2120). A, B. Habit of pseudostromata on twig. C. Transverse section through pseudostroma with ascomata. D. Longitudinal section through pseudostroma with ascomata. E. Asci. F. Ascospores. G, H. Habit of conidiomata on twig. I. Transverse section through conidioma. J. Longitudinal section through conidioma. K. Conidiophores and conidiogenous cells. L. Conidia. Scale bars: A, G = 1 mm; B–D, H–J = 500 µm; E, F, K, L = 10 µm.

ostioles per disc. Ostiole grey to black, (60–)80–135(–150)  $\mu$ m diam. *Locules* numerous, arranged circularly with common walls, (720–)870–960(–1 250)  $\mu$ m diam. *Conidiophores* borne along the locules, hyaline, branched at the base, in the middle or occasionally unbranched, (15.5–)16.5–18(–20.5) × 1–2  $\mu$ m, embedded in a gelatinous layer. *Conidiogenous cells* enteroblastic, phialidic, subcylindrical to cylindrical. *Conidia* hyaline, allantoid, thin-walled, eguttulate, aseptate, smooth, (6–)6.5–7.5(–8) × 1.5–2 (av. = 6.8 ± 0.5 × 1.7 ± 0.2, n = 30)  $\mu$ m.

*Culture characteristics*: Colonies initially pale vinaceous to vinaceous buff, flat with a uniform texture, aerial mycelium absent, growing up to 9 mm diam after 5 d, becoming dark at the centre

and spreading outwards. Conidiomata are randomly distributed on the medium surface.

Additional materials examined: China, Beijing City, Huairou District, Forestry Centre of Beijing University of Agriculture, 39°59'12.54"N, 115°26'17.08"E, on branches of *Corylus mandshurica*, 19 Jun. 2021, *X.L. Fan & Y.K. Bai* (BJFC-S2122, culture CFCC 56565; BJFC-S2123, culture CFCC 56578).

Notes: Cytospora malvicolor from Corylus mandshurica is closely related to *C. crataegina* from *Crataegus pinnatifida* var. *major* (Fig. 1) but differs from that species based on the nucleotide differences (see notes of *C. crataegina*). Although the sexual morph of *C. crataegina* has not been observed, *C. malvicolor* can be easily

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distinguished from *C. crataegina* by the larger conidia (6.5–7.5 × 1.5–2  $\mu$ m in *C. malvicolor vs* 3.5–4.5 × 1–1.5  $\mu$ m in *C. crataegina* and a distinct black conceptacle surrounding by its conidiomata.

*Cytospora massarii* L. Lin, X.L. Fan & Crous, *nom. nov.* MycoBank MB 853114. Fig. 32. (Fig. 1: Clade 141)

Replaced synonym: Valsa massariana De Not., Sfer. Ital.: 34. 1863, non Cytospora massariana Sacc., Syll. fung. (Abellini) 3: 253. 1884. Synonym: Cytospora massariana Sacc., Syll. fung. (Abellini) 3: 253. 1884.

Leucocytospora massariana (Sacc.) Z. Urb., Rozpr. Českoslov. Akad. Véd. Ročn. 68(Ses. 12): 72. 1958.

*Etymology*: Named after an older valid name of the taxon, *Valsa massariana* De Not.

*Typus*: On branches of *Sorbus aucuparia*: France, Germany, Italy. Requires lectotypification.

Description: Pseudostromata immersed in bark, erumpent, lenticular, 1 × 2 mm, leucostomoid circinate, 8–12 perithecia, circinately arranged, delimited by distinct brown conceptacle, tissues demarcating with a black line the ascoma from the host tissues. Discs below bark surface, medium brown, circular to ovoid, convex, 70–150 µm diam, 12–20 obscured ostioles. Ostioles many per disc, periphysate; walls of perithecia bilayered, narrow, above the discs, 40–100 µm diam. Perithecia inclined to upright, in valsoid or diatrypelloid configurations, immersed, usually embedded in entostroma, with beaks converging at disc or surface, 100 × 200 µm diam. Asci not observed. Ascospores hyaline, allantoid, aseptate, thin-walled, smooth, biseriate, multiseriate, (12-)13-16.5(-17.5) × (1-)1.5-3.5(-4) µm. Conidiomata produced on MEA in 2-4 wk, solitary, stromata up to 500 µm wide, multilocular, convoluted, semi-immersed, globose, exuding a pale luteous conidial cirrhus or globoid conidial mass. Conidiophores hyaline, smooth, branched, 0-3-septate, 10-50 × 1-2.5  $\mu$ m, embedded in a gelatinous layer. Conidiogenous cells phialidic, with periclinal thickening, subcylindrical, collarettes minute, 5-40 × 1.5-2 µm; arranged in rosettes of up to four, or lateral along branching conidiophores with terminal stipe extensions that resemble paraphyses, but at times also fertile. Paraphyses intermingled among conidiophores, hyaline, smooth, branched, septate, up to 50 µm long, 1-2.5 µm diam, at times developing fertile lateral cells. Conidia hyaline, smooth, guttulate, elongated allantoid, apex rounded, tapering to a subtruncate base, aseptate,  $4.5-6.5 \times 1-1.5 \mu m$ .

*Culture characteristics*: Colonies covering the 9-cm-diam Petri dish after 10 d at 24 °C, with aerial mycelium flat, moderately sparse. On MEA buff coloured on the surface and brown in reverse.

Material examined: Austria, Steiermark, Pack, former motorway picnic area close to motorway tunnel Kalcherkogel, 1 000 m s. m, on Sorbus aucuparia, 17 Apr. 2009, W. Jaklitsch (CBS H-22691, culture CBS 141473). Notes: Cytospora massarii L. Lin, X.L. Fan & Crous is published here as an explicit substitute ("nom. nov.") for the legitimate name Valsa massariana De Not. (Sfer. Ital.: 34. 1863) because



Fig. 32. *Cytospora massarii* (CBS 141473). A. Conidiomata on MEA. B–D. Conidiophores and conidiogenous cells. E. Conidia. F, H. Ascomata on dead branches of *Sorbus aucuparia*. G. Longitudinal section of pseudostromata with ascomata. I. Tangential section of pseudostromata with ascomata. J. Ascospores. Scale bars: A, F–I = 100  $\mu$ m; B–D = 10  $\mu$ m; E, J = 5  $\mu$ m.

the epithet massariana was unavailable in Cytospora as the name Cytospora massariana Sacc. [Syll. fung. (Abellini) 3: 253. 1884]. Cytospora massariana Sacc. is a younger (heterotypic) synonym of the valid name, Valsa massariana De Not. However, no type specimen of Valsa massariana De Not. was mentioned (De Not. 1863). The original material (from Sorbus aucuparia in Europe) mentioned by Saccardo (1884) is preserved in the PAD which can be chosen for lectotypification. There is no accurate sequence data or precise morphological characteristics for this species. Isolate CBS 141473 obtained from Sorbus aucuparia was identified as Leucostoma massarianum in the CBS database. The ITS, act1, *rpb2*, *tef1-\alpha* and *tub2* gene trees, the BI and ML phylogenies all place it in a distinct clade. That identification is retained here and we have added a morphological description and DNA sequence data for C. massarii. Consistent with its origin on the same host species and continent, the specimen CBS H-22691 with isolate CBS 141473 represents a potential neotype for this species, if no holotype can be located.

*Cytospora mougeotii* Lév., Bot. Gall., 2: 1272. 1830. (Fig. 1: Clade 49)

Typus: Europe, on Abies alba, Stirp. Crypt. Vog.-Rhen. nr. 1272 (F 42801).

Description: See Robak (1956), Roll-Hansen & Roll-Hansen (1980).

*Material examined*: **Norway**, on dead top of *Picea abies*, unknown date and *collector* (culture CBS 198.50).

*Notes*: Roll-Hansen & Roll-Hansen (1980) reported that *C. mougeotii* infects *Picea abies* in seasonal stem wounds. CBS 198.50 was identified as *C. mougeotii* by Adams *et al.* (2005). However, the type host is a different species, *Abies alba.* We retain the name for this taxon pending future epitypification. In the ITS, *act1, rpb2, tef1-α* and *tub2* gene trees, the BI and ML phylogenies all place it in a distinct clade.

Cytospora multicollis (Checa et al.) L. Lin, X.L. Fan & Crous, comb. nov. MycoBank MB 850162. (Fig. 1: Clade 145)

*Basionym*: Valseutypella multicollis Checa et al., Mycotaxon 25: 524. 1986.

*Typus*: **Spain**, Revenga, Carretera de La Granja a Riofrío, *Quercus ilex* subsp. *rotundifolia*, 10 Jun. 1984, *P. Yebes & J. Checa* (**holotype** AH 3854, **isotypes** AH 9306, AH 9388, NY 00936730, ex-holotype culture CBS 105.89).

Description: See Checa et al. (1986), Checa & Martinez (1989).

*Notes*: Checa & Martinez (1989) described *Cytospora* as asexual morph for *Valseutypella multicollis* and Adams *et al.* (2005) mentioned that *Valseutypella multicollis* is a typical member of *Valsa* based on ITS sequences. As *Valseutypella* and *Valsa* are synonyms of *Cytospora*, we introduce a new combination in *Cytospora* for this species. In the ITS, *act1*, *rpb2*, *tef1-α* and *tub2* gene trees, the BI and ML phylogenies all reveal *Cytospora multicollis* in a distinct clade.

Cytospora multiseriata L. Lin & X.L. Fan, sp. nov. MycoBank MB 850163. Fig. 33. (Fig. 1: Clade 110)

Etymology: Name refers to the multiseriate ascospores in the asci.

*Typus*: **China**, Tibet Autonomous Region, Lhasa City, Mozhugongka County, Zen Town, 29°42'20"N, 92°6'32"E, on branches of *Salix wallichiana*, 20 Aug. 2022, *Y.Y. Liu, M. Liu, N. Jiang, & P. Jin* (**holotype** BJFC-S2036, ex-holotype culture CFCC 58707).

Description: Pseudostromata Group SII (type s4), immersed in the bark, scattered, with 2–4 irregularly arranged perithecia. *Conceptacle* prominent. *Ectostromatic disc* white, with single ostiole per disc in the centre, triangular to circular, 200–350 µm diam. *Ostiole* single, umber to black when mature, arranged irregularly in a disc, 85–140 µm diam. *Perithecia* primrose to black when mature, flask-shaped to spherical, arranged irregularly, 340–



**Fig. 33.** *Cytospora multiseriata* (BJFC-S2036). **A**, **B**. Habit of pseudostromata on twig. **C**. Transverse section through pseudostroma with ascomata. **D**. Longitudinal section through pseudostroma with ascomata. **E**, **F**. Asci. **G**. Ascospores. Scale bars: A = 1 mm; B–D = 200 μm; E–G = 10 μm.



365 µm diam. *Asci* hyaline, with chitinoid, refractive apical ring, clavate to elongate-obovoid,  $34.5-62 \times 6.5-14$  µm (av. = 57.3 ± 4.3 × 11.7 ± 2.1 µm, n = 30), 8-spored. *Ascospores* hyaline, multiseriate, elongate-allantoid, aseptate, 9–17.5 × 2–4 µm (av. = 13.0 ± 2.0 × 2.8 ± 0.4 µm, n = 50).

*Notes*: Based on the ITS, *act1*, *rpb2*, *tef1-a* and *tub2* gene trees, *Cytospora multiseriata* forms a clade distinct from other *Cytospora* species (Fig. 1). Morphologically, it can be characterised by forming a black area on the bark in combination with white disc. Phylogenetic data and morphology justify describing *C. multiseriata* as a new species.

*Cytospora myrtagena* (G.C. Adams & M.J. Wingf.) G.C. Adams & Rossman, IMA Fungus 6: 147. 2015. (Fig. 1: Clade 204) *Basionym: Valsa myrtagena* G.C. Adams & M.J. Wingf., Stud. Mycol. 52: 97. 2005.

*Typus*: **USA**, Hawaii, on dead cankered branch of *Tibouchina urvilleana*, 2001, *M.J. Wingfield* (**holotype** of *V. myrtagena* MSC 380715, ex-holotype culture CBS 116843).

Description: See Adams et al. (2005).

Notes: Cytospora myrtagena was described from Eucalyptus and Tibouchina in Hawaii and Indonesia (Adams et al. 2005). It has a rosette cytosporoid type of conidiomata (Group All, type a7) and octosporous circinate type of pseudostromata (Group SII, type s3) (Adams et al. 2005). Jiang et al. (2020b) identified CFCC 52454 and 52455 as *C. myrtagena* provisionally and indicated that further research was needed to confirm their classification. These two isolates are introduced as a new species *C. castaneicola* in this study (see notes of *C. castaneicola*). The isolate CBS 117013 was identified as *C. myrtagena* (Adams et al. 2005). However, there are 14 bp differences between the ITS sequences of CBS 117013 (AY347380) and the ex-type strain CBS 116843 (AY347363). In this study, four isolates CBS 117013–117016 form a distinct and well-supported clade from CBS 116843 (ML/BI = 100/1). The morphological characteristics of specimens in this clade need to

be further studied. They are here retained as *Cytospora* sp. for the time being.

*Cytospora nanyangensis* X.L. Fan & C.M. Tian, *sp. nov.* MycoBank MB 850164. Fig. 34. (Fig. 1: Clade 38)

Etymology: Name refers to Nanyang City where it was collected.

*Typus*: **China**, Henan Province, Nanyang City, Baotianman National Nature Reserve, 33°26'36.33"N, 111°58'37.72"E, on twigs and branches of *Lindera glauca*, 5 Aug. 2020, *C. Peng & C.M. Tian* (**holotype** BJFC-S2167, ex-holotype culture CFCC 56024; **paratype** BJFC-S2168, ex-paratype culture CFCC 55846).

Description: Conidiomata Group AII (type a6), pycnidial, immersed in bark, scattered, erumpent through the bark surface in a large circular area, discoid, with large multiple locules. Conceptacle absent. Ectostromatic disc light yellow to brown, circular to ovoid, 245–325(–390) µm diam, with one ostiole per disc. Ostiole in the centre of the disc, inconspicuous, dark grey to black, at the level of the disc surface, (130–)150–195(–215) µm diam. Locules numerous, arranged irregularly with common walls, 860–1 125 µm diam. Conidiophores borne along the locules, hyaline, branched at base or unbranched, thin-walled, (10.5–)11.5–21.5(–24.5) × 1–2 µm, embedded in a gelatinous layer. Conidiogenous cells enteroblastic, phialidic, subcylindrical to cylindrical. Conidia hyaline, allantoid, eguttulate, smooth, aseptate, thin-walled, (5–)5.5–6(–6.5) × 1–2 (av. = 5.7 ± 0.3 × 1.6 ± 0.2, n = 30) µm.

*Culture characteristics*: Colonies fast growing, entirely covering the 9-cm-diam Petri dish and turning buff from middle to all sides after 6 d, after 14 d flat with a uniform texture. Sterile.

*Notes*: This species is characterised by its erumpent pycnidia through the bark, multiple locules with common walls and conidia  $5.5-6 \times 1-2 \mu m$ . Furthermore, in the ITS, *act1*, *rpb2*, *tef1-α* and *tub2* gene trees, the two isolates are phylogenetically distinct with high support (ML/BI = 100/1, 94/0.96, 100/1, 100/1 and 100/1, respectively).



**Fig. 34.** *Cytospora nanyangensis* (BJFC-S2167). **A, B.** Habit of conidiomata on twig. **C.** Transverse section through conidioma. **D.** Longitudinal section through conidioma. **E, F.** Conidiophores and conidiogenous cells. **G.** Conidia. Scale bars: A = 1 mm; B–D = 500 μm; E–G = 10 μm.

Cytospora nitschkeana L. Lin, X.L. Fan & Crous, nom. nov. MycoBank MB 853115. Fig. 35. (Fig. 1: Clade 77)

Replaced synonym: Valsa germanica Nitschke ex G.H. Otth, Mitt. naturf. Ges. Bern 654-683: 45 (1869) [1868]., non Cytospora germanica Sacc., Syll. fung. (Abellini) 3: 262 (1884) Synonym: Valsa germanica Nitschke, Pyrenomyc. Germ. 2: 215. 1870.

*Etymology*: Named in honour of the German mycologist Nitschke, in recognition of his contributions to this species.

## Typus: Requires typification.

Description: Conidiomata produced on MEA after 6–8 wk, solitary, globose, stromata black, up to 1 mm diam, immersed, convoluted multilocular, with one ostiole, exuding orange conidial cirrhus or globoid conidial mass. Conidiophores hyaline, smooth, acropleurogenous, branched, 0–2-septate, with terminal stipe extensions that resemble paraphyses, but at times also fertile cells, embedded in a gelatinous layer. Conidiogenous cells with periclinal thickening, subcylindrical, tapering towards apices, collarettes minute,  $3-7 \times 0.5-1.5 \ \mu$ m. Conidia hyaline, eguttulate, allantoid or straight, aseptate, apex subobtusely rounded,  $5.5-8 \times 0.5-2 \ \mu$ m. For the description of the sexual morph, see Nitschke (1870).

*Culture characteristics*: Colonies covering the Petri dish after 10 d at 24 °C, spreading, flat, olivaceous black on both surface and reverse.

*Materials examined*: **Germany**, on rotten branches, 1 Sep. 1864, *Nitschke* (Fuckel, Fung. rhen. n. 605, **lectotype** of *Valsa germanica* Nitschke designated here BGBM 132267, MBT 10017547). **Netherlands**, on *Salix alba* var. *vitellina-pendula*, unknown date and *collector* (**epitype** of *Valsa germanica* Nitschke designated here CBS H-22970, MBT 10017467, exepitype culture CBS 118.22). **Switzerland**, Bouveret, unknown host, 1942, *G. Défago* (CBS H-19163, culture CBS 195.42); Chippis, unknown host and date, *G. Défago* (culture CBS 196.42).

Notes: Cytospora nitschkeana L. Lin, X.L. Fan & Crous is published here as an explicit substitute ("nom. nov.") for the legitimate name Valsa germanica Nitschke ex G.H. Otth [Mitt. naturf. Ges. Bern 654–683: 45. 1869 (1868)] because the epithet germanica was unavailable in Cytospora as the name Cytospora germanica (Sacc., Syll. Fung. (Abellini) 3: 262. 1884) is a younger (heterotypic) synonym of the valid name, Valsa germanica Nitschke ex G.H. Otth [Mitt. naturf. Ges. Bern 654–683: 45. 1869 (1868)]. Several specimens are cited in the publication of Nitschke on page 217, from Salix fragilis, Betula and Populus tremula (Nitschke 1870). The original material (Fuckel, Fung. rhen. n. 605) mentioned by Nitschke (1870) is preserved in the Lichen Herbarium Berlin (BGBM 132263-132267). We choose the catalogue number 132267 as a lectotype of Valsa germanica Nitschke. Due to the collecting location and the same host family, CBS H-22970 is designated as an epitype isolate with CBS 118.22 as the ex-epitype of Valsa germanica Nitschke. Although they cannot be chosen as the type of Cytospora nitschkeana, as the legitimate name Valsa germanica Nitschke ex Otth has priority with all consequences concerning typification, CBS 118.22 can be a reference isolate of this clade. The main collection of Otth is now maintained in Zurich where it may contain a type specimen but further typification is needed.

CBS 195.42 and CBS 196.42 were identified as *V. germanica* (Adams *et al.* 2005). CBS 118.22 was deposited as *C. fugax* (= *V. salicina*) in the CBS collection based on morphological features. However, the present study has shown that it clusters distant from *C. fugax* but is contained within the *C. nitschkeana* clade (Fig. 1). *Cytospora nitschkeana* is sister to *C. azerbaijanica*. However, *Cytospora nitschkeana* (CBS 118.22) differs from *C. azerbaijanica* (Iran 4201C) in the ITS (1/512), *act* (7/241), *rpb2* (4/650), and *tef1-α* (11/270) loci (no available *tub2* sequence data are available for *C. azerbaijanica*).

*Cytospora nitschkei* G.C. Adams *et al.*, Stud. Mycol. 52: 105. 2005. (Fig. 1: Clade 181)

*Typus*: **Ethiopia**, Sidama Region, Wondo Genet, Forestry College Compound, on dead twigs of *Eucalyptus saligna*, 2002, *A. Gezahgne* (**holotype** MSC 380701, ex-holotype culture CBS 116854 = CMW 10180).

Description: Adams et al. (2005).

*Notes: Cytospora nitschkei* was described from *Eucalyptus saligna* having hemispherical conidiomata with upright diatrypelloid locules with ITS sequence data (Adams *et al.* 2005). The geographic distribution of this species includes only Ethiopia. In the current study, *act1*, *rpb2*, *tef1-a* and *tub2* gene sequences of ex-holotype culture CBS 116854 are provided.



Fig. 35. Cytospora nitschkeana (CBS 118.22). A. Conidiomata on MEA. B, C. Conidiophores and conidiogenous cells. D. Conidia. Scale bars: A = 100 μm; B, C = 10 μm; D = 5 μm.



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*Cytospora nobilis* Traverso, Boll. Soc. Bot. Ital.: 211. 1904. Fig. 36. (Fig. 1: Clade 61)

Typus: Italy, Villa Stroppa near Tradate (Como), on Laurus nobilis.

Description: Conidiomata Group AII (type a6), immersed in bark, erumpent when mature, discoid to conical, 330–495 µm diam, with multi-locules. Conceptacle absent. Ectostromatic disc buff to black, circular to ovoid, 110–245 µm diam with single ostiole per disc in the centre. Ostiole circular to ovoid, citrine to black, 35–60 µm diam. Locules divided with shared walls. Conidiophores hyaline, unbranched or occasionally branched at the bases, 11–23 × 1–2 µm (av. =  $16.4 \pm 3.0 \times 1.4 \pm 0.2 \mu m$ , n = 30). Conidiogenous cells enteroblastic, phialidic, subcylindrical to cylindrical, 2.5–4.5 × 1–1.5 µm (av. =  $3.6 \pm 0.4 \times 1.1 \pm 0.14 \mu m$ , n = 30). Conidia hyaline, unicellular, eguttulate, elongate-allantoid, 3.5–4.5 × 1.3–1.6 µm (av. =  $4.0 \pm 0.2 \times 1.43 \pm 0.1 \mu m$ , n = 50).

*Culture characteristics*: Cultures on PDA initially white, growing up to 7.5 cm diam after 3 d, entirely covering the 9-cm-diam Petri dish after 4 d and becoming light buff after 20 d, flat with a uniform texture.

*Materials examined*: **China**, Yunnan Province, Diqing Tibetan Autonomous Prefecture, Shangri-La City, Sanba Naxi Town, East Ring Road, 27°34'26"N, 100°1'5"E, on branches of *Laurus nobilis*, 9 Aug. 2022, *L. Lin & M. Lin* (BJFC-S2058, culture CFCC 58228; BJFC-S2057, culture CFCC 58227).

*Notes*: The two specimens examined in this study were collected from *Laurus nobilis* in Yunnan, China. In the ITS, *act1*, *rpb2*, *tef1-a* and *tub2* gene trees, the BI and ML phylogenies all place them in a distinct clade (ML/BI = 62/0.76, 99/1, 100/1, 99/0.97 and 98/1 respectively). *Cytospora lauri, C. nobilis, C. sacculus, C. leucosperma* (as *Valsa ambiens*), and *C. ceratosperma* (as *Valsa ceratosperma*) have been reported to infect *Laurus nobilis* (Kirk 1981, Kirk & Spooner 1989, Dudka *et al.* 2004). Of these four species, *C. lauri* and *C. nobilis* have no available DNA sequence data. These two species and the specimens studied here all have cytosporoid conidiomata. However, they can be distinguished from *C. lauri* by their wider conidia (3.5–4.5 × 1.3–1.6 µm vs 4–5 × 0.7–1 µm) (Grove 1937). Therefore, the name *C. nobilis* is tentatively applied to this clade pending further typification.

Cytospora oleicola D.P. Lawr. et al., IMA Fungus 9: 357. 2018. (Fig. 1: Clade 152)

*Typus*: **USA**, California, San Joaquin County, from twig canker of *Olea europaea* (**holotype** BPI 910657, ex-holotype culture CBS 144248).

Descriptions: See Adams et al. (2006), Lawrence et al. (2018).

Additional material examined: **South Africa**, Gauteng Province, Sterkfontein Cave, on dead twigs of *Olea europaea* at locations of insect feeding injury, 1998, *G.C. Adams & J. Roux* (MSC 384995, culture CBS 118555).

*Notes*: Adams *et al.* (2006) placed CBS 118555 within the *C. pruinosa* (= *V. cypri*) complex. However, this isolate clusters separately from *C. pruinosa* but together with *C. oleicola* based on the multigene tree and each of the single gene trees (Fig. 1). Isolate CBS 118555 has the same host species, unilocular conidiomata and similar conidial size as CBS 144248 (the ex-holotype culture of *C. oleicola*) (Lawrence *et al.* 2018). It is thus identified as *C. oleicola* here.

*Cytospora palmoides* Qing T. Zhang & X.Y. Zhang [as '*palm*'], Cryptog. Mycol. 35: 216. 2014. (Fig. 1: Clade 184)

*Typus*: **China**, Beijing City, Haidian District, Xiangshan, on twig of *Cotinus coggygria*, 1 May 2006, X.Y. *Zhang* (**holotype** culture CXY 1280).

Description: See Zhang et al. (2014).

Materials examined: China, Beijing City, Mentougou District, Xiaolongmen Forestry Centre, 39°50'54"N, 115°33'17"E, on twigs and branches of Rhus typhina, 25 Aug. 2022, L. Lin & X.L. Fan (BJFC-S2039, culture CFCC 58443; BJFC-S2040, culture CFCC 58445); 39°50'57"N, 115°33'39"E, on twigs and branches of Cotinus coggygria var. cinereus, 23 Aug. 2022, L. Lin & X.L. Fan (BJFC-S2041, culture CFCC 58451; BJFC-S2042, culture CFCC 58452); 39°50'48"N, 115°33'11"E, on twigs and branches of Cotinus coggygria var. cinereus, 23 Aug. 2022, L. Lin & X.L. Fan (BJFC-S2043, culture CFCC 58454); 39°50'39"N, 115°34'18"E, on twigs and branches of Cotinus coggygria var. cinereus, 23 Aug. 2022, L. Lin & X.L. Fan (BJFC-S2044, culture CFCC 58455); 39°51'45"N, 115°34'5"E, on twigs and branches of Rhus typhina, 25 Aug. 2022, L. Lin & X.L. Fan (BJFC-S2045, culture CFCC 58585); 39°54'37"N, 115°33'41"E, on twigs and branches of Cotinus coggygria var. cinereus, 23 Aug. 2022, L. Lin & X.L. Fan (BJFC-S2046, culture CFCC 58592; BJFC-S2047, culture CFCC 58593); Yunnan Province, Diqing Tibetan Autonomous Prefecture,



Fig. 36. Cytospora nobilis (BJFC-S2058). A, B. Habit of conidiomata on twig. C. Transverse section through conidioma. D. Longitudinal section through conidioma. E, F. Conidiophores and conidiogenous cells. G. Conidia. Scale bars: A = 1 mm; B = 100 µm; C, D = 200 µm; E–G = 10 µm.

Weixi Lisu Autonomous County, Yezhi Town, 27°49'37"N, 99°2'38"E, on branches of *Populus* × *canadensis*, 10 Aug. 2022, *L. Lin & M. Lin* (BJFC-S2048, culture CFCC 58489).

*Notes*: *Cytospora palmoides* can be recognised by its obvious morphological characteristics in having beaks protruding 1–3 mm long above the disks. It was characterised by having conidiomata with multiple locules sharing common walls, 8–15 × 1–1.5 µm long conidiophores and conidia of size 4–4.7 × 1–1.5 µm (Zhang *et al.* 2014). In the ITS, *act1, rpb2, tef1-a* and *tub2* gene trees, the currently available 10 isolates all cluster together with the exholotype strain of *C. palmoides* (CXY 1280), with high support (ML/BI = 88/0.997, 99/1, 100/1, 100/1 and 100/1, respectively). *Cytospora palmoides* was reported as a unique *Cytospora* species from *Cotinus coggygria* in Beijing, China (Zhang *et al.* 2014). The present study expands its host range to include *Rhus typhina*.

*Cytospora personata* (Fr.) Sacc., Syll. Fung. (Abellini) 1: 138. 1882. (Fig. 1: Clade 113)

*Basionym: Sphaeria personata* Fr., K. Sven. Vetensk. Akad. Handl. 40: 105. 1818.

Synonyms: Leucocytospora personata (Fr.) Höhn., Ber. Dt. Bot. Ges. 35: 352. 1917.

Valsa auerswaldii Nitschke, Pyrenomyc. Germ. 2: 225 (1870)

Typus: Sweden, on bark of Betula. Requires typification.

Description: See Hayova & Minter (2012a).

*Material examined*: **Austria**, Kärnten, St. Margareten im Rosental, Tumpfi, on *Frangula alnus*, 18 Mar. 2000, *W. Jaklitsch* (WJ 1424, BPI 748456, culture CBS 109774 = AR 3428).

Notes: CBS 109774 was identified as *Valsa auerswaldii*. According to Nitschke (1870) and Saccardo (1882), *C. personata* is the asexual morph of *V. auerswaldii*. However, in the original species description of *Sphaeria personata* no type material is mentioned and in the sanctioning publication (Fries 1823) the host is given as *Betula*. Nitschke (1870) also mentions collections on this host. Therefore, uncertainty remains regarding the synonymy of both taxa. Because there is no ex-type isolate for neither *C. personata* nor *V. auerswaldii*, we provisionally retain the name *C. personata* for the material examined, pending future typification. In the ITS, *act1, rpb2, tef1-α* and *tub2* gene trees, the BI and ML phylogenies all place CBS 109774 in a distinct clade.

*Cytospora pinastri* Fr. [as '*Cytispora*'], Syst. Mycol. (Lundae) 2(2): 544. 1823. (Fig. 1: Clade 44)

Synonyms: Cytospora friesii (Duby) Sacc., Syll. Fung. (Abellini) 3: 269. 1884.

Valsa friesii (Duby) Fuckel, Jahrb. Nassau. Ver. Naturkd. 23–24: 198. 1870.

Sphaeria friesii Duby, Bot. Gall., Edn 2 (Paris) 2: 118. 1830.

*Typus*: **Sweden**, on dead needles of *Pinus*, anon. s.n. (Fries, Scler. Suec. no. 247). Requires lectotypification.

Description: See Fries (1823), Hayova & Minter (2012b) (under Valsa friesii).

Additional materials examined: France, Villeurbanne, on Abies alba, 1972, F. Gourbière (CBS H-19158, culture CBS 505.72). Germany, Freiburg, on Abies alba, 1980, H. Courtois (CBS H-19160, culture CBS 113.81).



Switzerland, Vionnaz, on *Abies alba*, unknown date, *G. Défago* (culture CBS 194.42).

*Notes*: The isolates CBS 113.81, CBS 192.42 and CBS 505.72 were identified as *V. friesii* (sexual morph of *Cytospora pinastri*) (Adams *et al.* 2005). In the ITS, *act1*, *rpb2*, *tef1-α* and *tub2* gene trees, the BI and ML phylogenies all place these isolates in a distinct clade, with high support (ML/BI = 70/1, 100/1, 100/1, 100/1 and 100/1, respectively). Although there is no ex-type isolate available, CBS 113.81 occurs on the same host family and location as the type and is appropriate as a reference strain.

*Cytospora polyspora* X.L. Fan & C.M. Tian, *sp. nov.* MycoBank MB 850170. Fig. 37. (Fig. 1: Clade 106)

*Etymology*: Name refers to the distinctive polysporous asci in this species.

*Typus*: **China**, Ningxia Hui Autonomous Region, Guyuan City, Jingyuan County, 35°25'68.29"N, 106°22'68.17"E, on branches of *Salix* sp., 18 Oct. 2020, *C. Peng & C.M. Tian* (**holotype** BJFC-S2194, ex-holotype culture CFCC 55834); Gansu Province, Gannan City, Yeliguan National Forest Park, 34°56'61.40"N, 103°35'73.29"E, on branches of *Salix* sp., 24 Oct. 2020, *C. Peng & C.M. Tian* (**paratype** BJFC-S2196, ex-paratype culture CFCC 56012).

Description: Pseudostromata Group SI (type s2), immersed in the bark, erumpent through the bark surface, scattered, circular, (1 130–)1 270–1 470(–1 530) µm diam, with 5–7 perithecia arranged circularly in well-developed black entostromata. *Conceptacle* black. *Ectostromatic disc* brown to black, circular to ovoid, (550–)570–690(–800) µm diam, with ostioles arranged circularly per disc. *Ostioles* white, (100–)120–165(–240) µm diam. *Perithecia* flask-shaped to spherical, (220–)290–370(–390) µm diam. *Asci* free, clavate to elongate-obovoid, polysporous, (45.5– )47.5–53.5(–55) × (10–)10.5–12(–13) µm. Ascospores elongateallantoid, thin-walled, hyaline, aseptate, (12.5–)13–16(–18) × (2.9– )3.1–3.8(–4) (av. = 14.5  $\pm$  1.0 × 3.5  $\pm$  0.2, n = 30) µm. *Asexual morph* not observed.

*Culture characteristics*: Colonies initially white and becoming smoke grey to grey olivaceous after 14 d, lacking aerial mycelium, with a regular margin. Sterile.

Additional material examined: China, Ningxia Hui Autonomous Region, Guyuan City, Jingyuan County, 35°25'68.29"N, 106°22'68.17"E, on branches of *Salix* sp., 18 Oct. 2020, *C. Peng & C.M. Tian* (BJFC-S2195, culture CFCC 56014).

*Notes*: *Cytospora polyspora* was discovered on *Salix* sp. in northwest China. Phylogenetically, *C. polyspora* is a sister group to *C. tongzhouensis* and has the following nucleotide differences from the sequences for the ex-type of *C. tongzhouensis*: in ITS 0/510 bp, *act1* 2/241 bp, *rpb2* 6/726 bp and *tub2* 11/397 bp; no *tef1-a* available for *C. tongzhouensis*. Morphologically, *C. polyspora* differs from *C. tongzhouensis* by having pseudostromata with more perithecia (5–7 vs 4–5), larger asci (47.5–53.5 × 10.5–12 vs 38.5–47.5 × 8–9.5 µm) and larger ascospores (13–16 × 3.1–3.8 vs 7.5–10 × 1.3–2.2 µm). This species can be easily identified by its obvious white ectostromatic disc on infected branches, which is similar to *C. albodisca* (Pan *et al.* 2021). Additionally, the multigene phylogram reveals it as a distinct clade with high support (ML/BI = 99/1). Furthermore, this species has typical polysporus



**Fig. 37.** *Cytospora polyspora* (BJFC-S2194). **A, B.** Habit of pseudostromata on twig. **C.** Transverse section through pseudostroma with ascomata. **D.** Longitudinal section through pseudostroma with ascomata. **E.** Asci. **F.** Ascospores. Scale bars: A = 1 mm; B–D = 250 μm; E, F = 10 μm.

leucostomoid ascomata (Group SI, type s2), which is consistent with the morphology of *Valsella*.

*Cytospora populi* L. Lin & X.L. Fan, Plant Dis. 107: 87. 2023. Fig. 38. (Fig. 1: Clade 124)

*Typus*: **China**, Gansu Province, Lanzhou City, Yongdeng County, on dead branches of *Populus* sp., 20 Oct. 2020, *X.L. Fan, N. Jiang & C. Peng* (holotype BJFC CF20201004, ex-holotype culture CFCC 55473; paratype BJFC CF20201020, ex-paratype culture CFCC 55472).

Description: Pseudostromata Group SII (type s5), immersed in the bark, scattered, with 5-10 perithecia arranged irregularly. Conceptacle prominent. Ectostromatic disc buff to salmon, usually surrounded by ostiolar necks, triangular to circular, 295–465 µm diam, with 6-14 irregularly arranged ostioles. Ostioles brown to black when mature, arranged irregularly in a disc, 50-70 µm diam. Perithecia primrose to black when mature, flask-shaped to spherical, arranged irregularly, 150–210 µm diam. Asci hyaline, with chitinoid, refractive ring, clavate to elongate-obovoid, 38.5-53 × 10–14.5 μm (av. = 45.4 ± 3.9 × 11.3 ± 0.8 μm, n = 30), 8-spored. Ascospores hyaline, biseriate to multiseriate, elongate-allantoid, aseptate, 9–14.5 × 2.5–4  $\mu$ m (av. = 12.5 ± 1.3 × 3.2 ± 0.3  $\mu$ m, n = 50). Conidiomata Group AIII (type a12), immersed in bark, erumpent when mature, discoid to conical, 525-710 µm diam, with multi-locule. Conceptacle prominent. Ectostromatic disc buff to salmon, circular to ovoid, 245-290 µm diam, with single ostiole per disc in the centre. Ostiole circular to ovoid, black, 60-80 µm diam. Locules numerous, irregular arrangement with individual walls. Conidiophores hyaline, unbranched or occasionally branched,  $26.5-45.5 \times 2-3.5 \ \mu m$  (av. =  $40.0 \pm 5.5 \times 2.9 \pm 0.4 \ \mu m$ , n = 30). Conidiogenous cells enteroblastic, phialidic, subcylindrical to cylindrical, 5.5–8.5 × 1.5–3  $\mu$ m (av. = 7.9 ± 0.4 × 2.2 ± 0.4  $\mu$ m, n = 30). Conidia hyaline, unicellular, eguttulate, elongate-allantoid,  $4.5-6 \times 1-2 \ \mu m$  (av. =  $5.3 \pm 0.4 \times 1.6 \pm 0.2 \ \mu m$ , n = 50).

*Culture characteristics*: Cultures on PDA are initially white, growing up to 6 cm diam after 3 d, becoming isabelline after 10 d. The colonies are olivaceous on the surface and dark mouse grey in reverse after 20 d, flat with a uniform texture.

*Material examined*: **China**, Yunnan Province, Diqing Tibetan Autonomous Prefecture, Shangri-La City, Sanba Naxi Town, East Ring Road, 27°34'26"N, 100°1'5"E, on branches of *Populus adenopoda*, 9 Aug. 2022, *L. Lin & M. Lin* (BJFC-S2050, culture CFCC 58250).

*Notes*: Lin *et al.* (2023a) described *C. populi* from *Populus* sp. in Gansu, China. The isolates CFCC 58250 and CFCC 55473 (the ex-holotype culture of *C. populi*) have no clear differences based in ITS (100 % identity, with 0 bp difference), *act1* (99.2 % identity, with 2 bp differences), *tef1-a* (97.9 %, with 6 bp differences), *tub2* (99.2 %, with 3 bp differences) sequences and cluster together with high support (ML/BI = 100/1). Both collections have torsellioid conidiomata, with a black conceptacle (Lin *et al.* 2023a). Therefore, isolate CFCC 58250 is identified as *C. populi* based on morphology and phylogenetic placement and this study adds a sexual morph description for *C. populi*.

*Cytospora pruinopsis* C.M. Tian & X.L. Fan, Mycol. Progr. 14: 74. 2015. (Fig. 1: Clade 155)

*Typus*: **China**, Shaanxi Province, Yulin City, Yuyang District, Red Stone Gorge, 38°19'32.43"N, 109°42'00.63"E, on twigs and branches of *Ulmus pumila*, 29 Jul. 2013, *X.L. Fan* (**holotype** BJFC-S1073, ex-holotype culture CFCC 50034).

Description: See Yang et al. (2015), Fan et al. (2020).

Additional material examined: **China**, Beijing City, Mentougou District, Mount Dongling, Xiaolongmen Forestry Centre, 39°51'45"N, 115°34'5"E, on twigs and branches of *Ulmus pumila*, 25 Aug. 2022, *L. Lin & X.L. Fan* (BJFC-S2051, culture CFCC 58463).



Fig. 38. Cytospora populi (BJFC-S2193). A. Habit of pseudostromata and conidiomata on branch. B. Disc and ostiole of conidioma. C. Transverse section through conidiomata. D. Longitudinal section through conidioma. E. Ectostromatic disc and ostioles of pseudostroma. F. Transverse sections through pseudostroma with ascomata. G, H. Longitudinal section through pseudostroma with ascomata. I–J. Conidiophore and conidiogenous cell. K. Conidia. L, M. Ascus. N. Ascospores. Scale bars: A = 250 µm; B–H = 200 µm; I–N = 10 µm.

*Notes*: *Cytospora pruinopsis* was described being associated with a canker disease on *Ulmus pumila*, and can be distinguished by having single locules with one ostiole. However, it has similar characteristics to *C. pruinosa*, but this species differs from *C. pruinosa* by having smaller conidia (2.5–3.5 × 1 vs 5.5–7 × 1.5  $\mu$ m) (Yang *et al.* 2015). In the ITS, *act1*, *rpb2*, *tef1-α* and *tub2* gene trees, the BI and ML phylogenies all place *C. pruinopsis* in a distinct clade, with high support (ML/BI = 100/1 in all five genes).

*Cytospora pruinosa* (Fr.) Sacc., Michelia 1(5): 519. 1879. Fig. 39. (Fig. 1: Clade 146)

*Basionym: Sphaeria pruinosa* Fr., K. Sven. Vetensk. Akad. Handl. 39: 104. 1817.

Synonyms: Dendrophoma pruinosa (Fr.) Sacc., Syll. Fung. (Abellini) 3: 179. 1884.

Cytophoma pruinosa (Fr.) Höhn., Sber. Akad. Wiss. Wien, Math.naturw. Kl., Abt. 1 123: 133. 1914.

Valsa pruinosa (Fr.) Défago, Phytopath. Z. 14: 123. 1942.

Sphaeria cypri Tul., Forstwiss. Zbl.: 706. 1856.

Cytospora cypri (Tul.) Défago, Phytopath. Z. 14: 120. 1942.

Valsa cypri (Tul.) Tul. & C. Tul., Select. Fung. Carpol. (Paris) 2: 194. 1863.

*Typus*: **Sweden**, on bark of branches of *Fraxinus excelsior*, *Fries*, Scler. Suec. Exs. n. 82. Requires lectotypification.

Description: Conidiomata Group AI (type a5), immersed in bark, scattered, erumpent, conical, with a single locule. Conceptacle absent. Ectostromatic disc brown to grey, circular to ovoid, 420–600(–695)  $\mu$ m diam, with single ostiole per disc. Ostiole in the centre of the disc, conspicuous, dark grey to black, at the same level as

the disc surface, (165–)200–260(–280) µm diam. *Locule* undivided, circular to ovoid, occasionally wrinkled, (625–)690–935(–1020) µm diam. *Conidiophores* borne along the locules, hyaline, branched at base or unbranched, thin-walled, (8.5–)10.5–18.5 × 1.5–2 µm, embedded in a gelatinous layer. *Conidiogenous cells* enteroblastic, phialidic, subcylindrical to cylindrical. *Conidia* hyaline, allantoid, eguttulate, smooth, aseptate, thin-walled, (4.5–)5–5.5(–6) × 1.5–2 (av. =  $5.3 \pm 0.2 \times 1.6 \pm 0.1$ , n = 30) µm. For the description of sexual morph, see Saccardo (1884) (as *Dendrophoma pruinosa*).

*Culture characteristics*: Colonies on PDA initially white, growing up to 4.5 cm diam and becoming buff after 7 d, ceasing growth at a colony diameter of 7 cm, with an irregular edge. Sterile.

Additional materials examined: China, Liaoning Province, Fuxin City, Zhonghua Road, 42°1'54.35"N, 121°41'31.85"E, on branches of *Fraxinus chinensis*, 28 Jul. 2021, *X.L. Fan & C.M. Tian* (BJFC-S2139, culture CFCC 55838; BJFC-S2140, culture CFCC 56019). Netherlands, on dead wood, 2019, *N. Jiang* (BJFC-S2104, culture CFCC 54025). Switzerland, on *Fraxinus excelsior*, 1942, *Gasser* (CBS H-19176, culture CBS 200.42); on *Syringa* sp., 1942, G. Défago (CBS H-19178, culture CBS 201.42); unknown host and date, *G. Défago* (culture CBS 199.42). Unknown location, on *Fraxinus americana*, 1925, unknown *collector* (CBS H-22923, culture CBS 133.25).

*Notes: Cytospora pruinosa* is distributed worldwide on *Fraxinus*, *Syringa* and *Olea* (Adams *et al.* 2006, Fan *et al.* 2020). In the ITS, *act1*, *rpb2*, *tef1-α* and *tub2* gene trees, the BI and ML phylogenies all placed it in a distinct clade, with high support (ML/ BI = 99/1, 100/1, 100/1, 100/1 and 100/1, respectively). No type material could be located for this species, but a lectotype can be chosen from Fries, namely Scleromycetes Sueciae 82, which





Fig. 39. Cytospora pruinosa (BJFC-S2139). A–B. Habit of conidiomata on twig. C. Transverse section through conidioma. D. Longitudinal section through conidioma. E, F. Conidiophores and conidiogenous cells. G. Conidia. Scale bars: A = 1 mm; B–D = 250 µm; E–G = 10 µm.

is mentioned in the sanctioning publication (Fries 1823). In the present study, CBS 200.42 is treated as a reference isolate by the same host species and collected continent as type. *Valsa cypri* which commonly considered a synonym of *Cytospora pruinosa* is described from *Ligustrum vulgare* (Défago 1942, Hayova & Minter 1998b), but this needs to be confirmed with sequence data for isolates from the type host.

Cytospora pseudochrysosperma L. Lin & X.L. Fan, sp. nov. MycoBank MB 850167. Fig. 40. (Fig. 1: Clade 60)

Etymology: Name reflects a connection to C. chrysosperma.

*Typus*: **China**, Gansu Province, Gannan Tibetan Autonomous Prefecture, Lintan County, Ligang Village, 103°27'14.78"E, 34°39'07.89"N, on branches of *Populus alba* var. *pyramidalis*, *X.L. Fan*, 8 Aug. 2012 (**holotype** BJFC-S750, ex-holotype culture CFCC 89981); 103°41'44.77"E, 34°30'4.4"N, on branches of *Populus* sp., *X.L. Fan*, 7 Aug. 2012 (**paratype** BJFC-S2223, ex-paratype culture CFCC 54081).

Description: Pseudostromata immersed in the bark, erumpent through the bark surface in a large area, scattered, with 12-18 irregularly arranged perithecia. Conceptacle absent. Ectostromatic disc hazel to dark mouse grey, usually surrounded by ostiolar necks, triangular to circular, 600-1260 µm in diam., with 3-10 irregularly arranged ostioles. Ostioles umber to black when mature, at the same or level protruding above the disc, arranged irregularly in a disc, 170-390 µm in diam. Perithecia dark mouse grey to black when mature, flask-shaped to spherical, arranged irregularly, 360-575 µm in diam. Asci hyaline, with chitinoid, refractive ring, clavate to elongate-obovoid, 40.0-50.0 × 8.0-11.0 µm (av. = 46.4  $\pm$  1.9 × 9.4  $\pm$  0.7 µm, n = 30), 8-spored. Ascospores hyaline, biseriate to multiseriate, elongate-allantoid, thin-walled, aseptate,  $9.0-11.0 \times 2.0-2.5 \ \mu m (av. = 9.9 \pm 0.4 \times 2.2 \pm 0.1 \ \mu m, n = 50).$ Conidiomata Group All (type a6), immersed in bark, erumpent when mature, discoid, flask-shaped to conical, 540-865  $\mu m$  in diam, with large multi-locule. *Conceptacle* absent. *Ectostromatic disc* conspicuous, umber to isabelline, circular to ovoid, 290–465 µm in diam. *Ostiole* conspicuous, circular to ovoid, isabelline to black at the same level as the disc surface, 135–190 µm in diam. *Locules* subdivided frequently by invaginations with common walls. *Conidiophores* hyaline, unbranched or occasionally branched at the bases, 13–25 × 1–2.5 µm (av. = 17.3 ± 2.0 × 1.6 ± 0.2 µm, n = 30). *Conidiogenous cells* enteroblastic, phialidic, subcylindrical to cylindrical, 4.5–9 × 1–2.5 µm (av. = 7.0 ± 0.8 × 1.6 ± 0.2 µm, n = 30). *Conidia* hyaline, unicellular, eguttulate, elongate-allantoid, 5–6 × 1–2 µm (av. = 5.6 ± 0.3 × 1.6 ± 0.1 µm, n = 50).

Notes: Cytospora pseudochrysosperma, reported as a pathogen of poplar and willow in China, was initially identified as *C. chrysosperma* based on morphology (Deng 1963, Tai 1979, Wei 1979, Zhuang 2005). Fan *et al.* (2014b) first provided ITS sequence data for *C. pseudochrysosperma* (as *C. chrysosperma*) and subsequently described it using multi-locus DNA sequence data (Fan *et al.* 2015a). In this study, we describe it as a new species *C. pseudochrysosperma*. It has a similar asexual morph type to that of *C. chrysosperma* (Group AII, type a6), but they can be distinguished from that species based on phylogenetic inference (Fig. 1). The exholotype of *C. pseudochrysosperma* (CFCC 89981) differs from the ex-epitype of *C. chrysosperma* (CBS 197.50) in ITS (2/514 bp difference), *act1* (10/255 bp difference), *rpb2* (26/650 bp difference), *tef1-a* (27/492 bp difference) and *tub2* (13/411 bp difference) loci.

*Cytospora qinghaiensis* L. Lin & X.L. Fan, *sp. nov.* MycoBank MB 850171. Fig. 41. (Fig. 1: Clade 14)

*Etymology*: Name refers to, Qinghai Province where it was collected.

*Typus*: **China**, Qinghai Province, Xining City, 36°38'32.51"N, 101°44'42.89"E, on twigs and branches of *Ulmus pumila*, 16 Aug. 2012, *X.L. Fan* (**holotype** BJFC-S671, ex-holotype culture CFCC 50026).



Fig. 40. Cytospora pseudochrysosperma (BJFC-S750). A, B. Habit of conidiomata on twig. C. Transverse section through conidioma. D. Longitudinal section through conidioma. E. Conidiophores and conidiogenous cells. F. Conidia. G, H. Habit of pseudostromata on twig. I. Transverse section of pseudostroma with ascomata. J. Longitudinal section through pseudostroma with ascomata. K, L. Asci. M. Ascospores. Scale bars: A, G = 5 mm; B–D, G, H = 1 mm; E, F, K–M = 10 µm.

Description: Conidiomata Group AII (type a6, a9), immersed in bark, erumpent when mature, discoid, flask-shaped to conical, with large multi-locule. Conceptacle absent. Ectostromatic disc grey to black, circular to ovoid, 360–465 µm in diam. Ostiole circular to ovoid, isabelline to black at the same level as the disc surface, 135–155 µm in diam. Locules subdivided frequently by invaginations with common walls. Conidiophores reduced to conidiogenous cells, hyaline, unbranched or occasionally branched at the bases, 17–19 × 1–2 µm (av. =  $18.3 \pm 0.6 \times 1.6 \pm 0.2 \mu$ m, n = 30). Conidia hyaline, unicellular, eguttulate, elongate-allantoid, 7–10.5 × 1–2 µm (av. =  $8.2 \pm 1.3 \times 1.6 \pm 0.1 \mu$ m, n = 50).

*Additional material examined:* **China**, Qinghai Province, Xining City, 36°38'32.51"N, 101°44'42.89"E, on twigs and branches of *Ulmus pumila*, 16 Aug. 2012, *X.L. Fan* (culture CFCC 50027).

*Notes*: Yang *et al.* (2015) treated CFCC 50026 and 50027 as *C. ribis* based on ITS sequence data, but in the multigene phylogenies of the present study it groups separately from the clade 1, and it is placed as a distinct highly supported clade (ML/ BI = 100/0.96) inside a larger clade containing *C. auerswaldii* and *C. tanaitica*. However, *C. qinghaiensis* (ex-holotype CFCC 50026) has the following nucleotide differences from the sequences of *C. auerswaldii* (CBS 153.29) and *C. tanaitica* (ex-holotype MFLUCC 14-1057): In ITS: 1/506 bp and 1/493 bp, respectively; in *act1*:



**Fig. 41.** *Cytospora qinghaiensis* (BJFC-S671). **A.** Habit of conidioma on twig. **B.** Transverse section through conidioma. **C.** Longitudinal section through conidioma. **D.** Conidiophores and conidiogenous cells. **E.** Conidia. Scale bars: A–C = 200 μm; D, E = 10 μm.

4/241 bp and 13/226 bp, respectively; in *rpb2*: 25/650 bp and no *rpb2* available for *C. tanaitica*, respectively; in *tef1-* $\alpha$ : 3/481 bp and no *tef1-* $\alpha$  available for *C. tanaitica*, respectively; in *tub2*: 0/412 bp and no *tub2* available for *C. tanaitica*, respectively. Therefore, it should be treated as a new species.

*Cytospora qingshuiensis* L. Lin & X.L. Fan, *sp. nov.* MycoBank MB 850172. Fig. 42. (Fig. 1: Clade 39)

Etymology: Name refers to Qingshui Town where it was collected.

*Typus*: **China**, Beijing City, Mentougou District, Qingshui Town, 39°51'56.38"N, 115°26'54.10"E, on branches of *Platycladus orientalis*, 21 Aug. 2021, *X.L. Fan & M. Pan* (**holotype** BJFC-S2111, ex-holotype culture CFCC 56268; **paratype** BJFC-S2112, ex-paratype culture CFCC 56349).

Description: Conidiomata Group AII (type a6), scattered or serried, immersed in bark, erumpent through the bark surface, discoid to conical, with multiple locules. Conceptacle absent. Ectostromatic disc black to grey, circular, (210–)320–380(–690) µm diam, with one ostiole per disc. Ostiole grey to black, (150–)240–320(–340) µm diam. Locules numerous, arranged circularly with common walls, (690–)800–970(–1 150) µm diam. Conidiophores borne along the locules, hyaline, unbranched or occasionally branched at the base, (20–)22–26.5(–29) × 1–2 µm, embedded in a gelatinous layer. Conidiogenous cells enteroblastic, phialidic, subcylindrical to cylindrical. Conidia hyaline, allantoid, thin-walled, eguttulate, aseptate, smooth, (5–)6–7.5(–8.5) × 1–2 (av. = 6.6 ± 0.7 × 1.5 ± 0.2, n = 30) µm.

*Culture characteristics*: Colonies initially white and entirely covering the 9-cm-diam Petri dish after 6 d, flat with a uniform texture and deepened in later stage gradually after 14 d, ultimately lacking aerial mycelium, thin with an irregular edge and becoming honey. Conidiomata randomly distributed on medium surface.

Additional materials examined: China, Changping District, Baihujian Natural Scenic Area, 40°7'21.79"N, 116°5'30.44"E, on twigs and branches of *Platycladus orientalis*, 4 Aug. 2021, *X.L. Fan & Y.K. Bai* (BJFC-S1990, culture CFCC 59053); *ibid.*, Yanqing District, Songshan National Nature Reserve, 40°29'48"N, 115°46'51"E, on twigs and branches of *Platycladus orientalis*, 4 Aug. 2021, *X.L. Fan & Y.K. Bai* (BJFC-S1991, culture CFCC 59054).

*Notes: Cytospora qingshuiensis* was discovered on the stems of *Platycladus orientalis.* It has multiple locules without conceptacles, similar to most *Cytospora* species, but a clear phylogenetic position with maximum support (ML/BI = 100/1), making it distinct from other isolates included in this study. In the ITS, *act1*, *rpb2*, *tef1-α* and *tub2* gene trees, the BI and ML phylogenies all place it in a distinct clade, with high support (ML/BI = 89/0.93, 100/1, 100/1, 100/1, 100/1, and 100/1, respectively).

*Cytospora ribis* Ehrenb., Sylv. mycol. berol. (Berlin): 28. 1818. Fig. 43. (Fig. 1: Clade 1)

Synonyms: Cytospora ampulliformis Norph. et al. Mycosphere 8: 56. 2017.

Cytospora cotini Norph. et al., Fungal Diversity 80: 176. 2016.

Typus: Requires typification.

Description: Conidiomata Group AII (type a9), scattered, immersed in bark, erumpent slightly through the bark surface, discoid to conical, with multiple locules, 480–740 µm diam. Conceptacle absent. Ectostromatic disc straw to pale luteous, circular, 190–340 µm diam, with one ostiole per disc. Ostiole grey to black, 70–120 µm diam. Locules numerous, arranged circularly with common walls. Conidiophores borne along the locules, hyaline, branched at base, in the middle or unbranched, 11.5–16.5 × 1.5–2 (av. = 13.6 ± 0.5 × 1.7 ± 0.1, n = 30) µm, embedded in a gelatinous layer. Conidiogenous cells enteroblastic, phialidic, subcylindrical



Fig. 42. Cytospora qingshuiensis (BJFC-S2111). A, B. Habit of conidiomata on twig. C. Transverse section through conidioma. D. Longitudinal section through conidioma. E. Conidiophores and conidiogenous cells. F. Conidia. Scale bars: A = 1 mm; B–D = 500 µm; E, F = 10 µm.

to cylindrical,  $2.5-5 \times 1.5-2$  (av. =  $4.3 \pm 0.2 \times 1.7 \pm 0.1$ , n = 30). *Conidia* hyaline, allantoid, thin-walled, eguttulate, aseptate, smooth,  $5-6 \times 1.5-2$  (av. =  $5.6 \pm 0.2 \times 1.7 \pm 0.1$ , n = 50) µm.

*Materials examined*: **Netherlands**, on dead wood, 2019, *N. Jiang* (BJFC-S2059, culture CFCC 54030; BJFC-S2060, culture CFCC 54014; BJFC-S2061, culture CFCC 54022, 54026; BJFC-S2062, culture CFCC 54032); on *Alnus* sp., 2019, *N. Jiang* (BJFC-S2063, culture CFCC 54015; BJFC-S2064, culture CFCC 54016). **Switzerland**, on *Acer campestre*, unknown date and *collector* (culture CBS 186.42); on *Ribes rubrum*, unknown date and *collector* (culture CBS 187.36); on *Syringa* sp., unknown date and *collector* (sp. 490.42); on *Taxus baccata*, unknown date and *collector* (sp. 490.42); on *Taxus ba* 

*collector* (culture CBS 191.42); unknown host, 1942, *Gasser* (culture CBS 198.42); on dead twig of *Fraxinus excelsior*, unknown date and *collector* (culture CBS 349.69); **USA**, on twig of *Clethra alnifolia*, unknown date and *collector* (culture CBS 141609 = CPC 28369 = LCM 171.01); on twigs of *Salix lucida*, unknown date and *collector* (culture CBS 141610 = CPC 28370 = LCM 437.01).

*Notes: Cytospora ribis* was first collected in Europe, but the type specimen could not be traced. The isolate CBS 187.36 has been treated as *C. ribis* in the CBS database based on morphology and Adams *et al.* (2006) accepted this name and provided rDNA-ITS



Fig. 43. Cytospora ribis (BJFC-S2059). A, B. Habit of conidiomata on twig. C. Transverse section through conidioma. D. Longitudinal section through conidioma. E. Conidiophores and conidiogenous cells. F. Conidia. Scale bars: A = 2 mm; B–D = 200 μm; E, F = 10 μm.



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sequences. We recommend treating this isolate as reference for C. ribis consistent with the fact that it was collected in Europe and from the same host (Ribes) on which it was first described. Hyde et al. (2016) and Norphanphoun et al. (2017) described C. cotini (MFLUCC 14-1050) and C. ampulliformis (MFLUCC 16-0629) that group in the same clade as C. ribis and they are therefore considered to be conspecific. The isolate CFCC 54030 has nucleotide similarities with the sequences of the ex-holotype of C. cotini (MFLUCC 14-1050) and C. ampulliformis (MFLUCC 16-0629): in ITS: identical in both species; in act1: no act1 available for C. cotini (MFLUCC 14-1050) and 214/214 bp (100 %), respectively; in rpb2: 703/703 bp (100 %) and bp 701/703 (99.7 %), respectively. Cytospora cotini was collected in Russia, on Cotinus coggygria (Onocleaceae) and C. ampulliformis in Russia, on Sorbus intermedia (Rosaceae), are more recent names for this species. The isolate CBS 198.42 was deposited as Valsa platani in the CBS database. However, the type of Valsa platani is from Platanus in USA, but CBS 198.42 is from Switzerland from an unknown host. Isolate CBS 349.69 was treated as C. pruinosa in the CBS database. However, the conidiomata of C. pruinosa (BJFC-S2139, see the description of clade 81) have undivided locules, while the conidiomata of BJFC-S2059 in this clade have numerous locules. Therefore, prior to future epitypification, the names C. platani or C. pruinosa cannot be used for this clade.

*Cytospora saccardoi* L. Lin, X.L. Fan & Crous, *nom. nov.* MycoBank MB 853116. Fig. 44. (Fig. 1: Clade 47) *Replaced synonym: Valsa cenisia* De Not., Hedwigia 2: 178 (1863), *non Cytospora cenisia* Sacc., Syll. Fung. (Abellini) 3: 271. 1884. *Synonym: Cytospora cenisia* Sacc., Syll. fung. (Abellini) 3: 271. 1884.

*Etymology*: Named in honour of the Italian mycologist Saccardo, in recognition of his contributions to this species.

## Typus: Requires typification.

Description: Conidiomata produced on MEA after 6–8 wk, solitary, stromata black, up to 300  $\mu$ m diam, semi-immersed, unilocular, not exuding conidia. Conidiophores hyaline, smooth, branched, 0–4-septate, 3–10 × 1.5–3  $\mu$ m, embedded in a gelatinous layer. Conidiogenous cells embedded in a continuous gelatinous matrix, phialidic, subcylindrical, tapering towards apices, collarettes minute, 3–8 × 1.5–3  $\mu$ m arranged in rosettes of up to three. Conidia hyaline, eguttulate, cylindrical to ellipsoidal, variable in shape,

aseptate, apex sub-obtusely rounded, tapering to a subtruncate base,  $4-6 \times 1.5-2.5 \ \mu\text{m}$ .

*Culture characteristics*: Colonies covering the Petri dish after 10 d at 24 °C, with aerial mycelium flat, moderately sparse. On MEA surface honey and brown on the reverse.

*Material examined*: **Germany**, Hesse, Johannisberg, on dry branches of *Juniperus communis*, spring, without date, *Fuckel*, in Fuckel, Fungi Rhenani 2139 (**lectotype** of *Cytospora cenisia* Sacc. **designated here** G 00585193, MBT 10019182; G 00585194, G 00585195 isolectotypes). **Austria**, Steiermark, Obdach, Zirbitzkogel at the Sabathyhütte, on *Juniperus communis*, 3 Sep. 2000, *W. Jaklitsch*, WJ 1583 (**epitype** of *C. cenisia* Sacc. designated here BPI 748457, MBT 10017478, WU-MYC 0052107 isoepitype, ex-epitype culture CBS 109752 = AR 3522). **USA**, Colorado, on dead branch of *Cercocarpus montanus*, 2001, unknown *collector* (CBS H-22982, culture CBS 141615 = CPC 28393 = AR 3721).

Notes: Cytospora saccardoi L. Lin, X.L. Fan & Crous is published here as an explicit substitute ("nom. nov.") for the legitimate name Valsa cenisia De Not. (Hedwigia 2: 178, 1863). Saccardo (1884) recorded C. cenisia from Juniperus communis and J. virginiana in Finland, Germany, Italy and USA but did not designate a type. In the protologue of his Cytospora cenisia, Saccardo referred to Nitschke's description of an asexual morph, described by the latter under Valsa cenisia De Not. The description provided by Saccardo for "stat. sperm. of Valsa cenisia" mirrors that of Nitschke. It is evident that Saccardo introduced the name Cytospora cenisia for this anamorphic state not a new combination. Therefore, the earlier Valsa cenisia, which has priority, cannot be combined in Cytospora as the epithet is occupied. Nitschke, in his examination, referred to material collected by Fuckel (page 186 "auf dickeren Zweigen von Juniperus communis L. bei Oestrich in Nassau von Fuckel gesammelt"), that belongs to the protologue and is viable for lectotypification. Fuckel (1870) cited an asexual morph under Valsa cenisia with a brief description, referring to his exsiccatae Fuckel, Fungi Rhen. 2139. A copy of this exsiccatae deposited in G (G 00585193) is therefore here selected as lectotype of Cytospora cenisia.

Isolate CBS 109752 was identified as *Valsa cenisia* (Castlebury *et al.* 2002). It was isolated from the same host (*Juniperus communis*) and continent (Europe) as Saccardo's records. Its conidial size corresponds with the original description (4–6 × 1.5–2.5  $\mu$ m *vs* 5–7 × 1.5  $\mu$ m). Therefore, we designate the specimen BPI 748457 as the epitype for *Cytospora cenisia*, with the isolate CBS 109752



Fig. 44. Cytospora saccardoi (CBS 141615). A. Conidioma on twig in MEA. B–E. Conidiophores and conidiogenous cells. F. Conidia. Scale bars: A = 100 μm; B–E = 10 μm; F = 5 μm.

designated as an ex-epitype which can be a reference isolate of this clade. However, there is still no type of *Cytospora saccardoi* as the legitimate name *Valsa cenisia* De Not. has priority, with all consequences concerning typification, that needs further study.

*Cytospora sacchari* E.J. Butler, Agric. India, Bot. Ser. 1: 31. 1906. (Fig. 1: Clade 168)

*Typus*: India, Tamil Nadu, Madras, Coimbatore, Shinga Nellore, on stem of *Saccharum officinarum*, 13 Sep. 1904, *E.J. Butler* (possible type 1764).

Description: See Sivanesan (1983).

Material examined: India, Bihar, Pusa, on Saccharum officinarum, unknown date, W. McRae (culture CBS 160.33).

*Notes: Cytospora sacchari* is mostly isolated from sheaths, also cuttings, young shoots, stems and stubble of *Saccharum officinarum*, *S. spontaneum* and *Holcus sorghum* (*Sorghum vulgare*). In the ITS, *act* and *rpb2* gene trees, the BI and ML phylogenies all place isolate CBS 160.33 in a distinct clade. There is a possible type specimen of *C. sacchari* in Herbarium Catalogue (Stockholm), but this could not be obtained. CBS 160.33 which was isolated from the same host and location as the original records is treated a reference isolate and a potential epitype for *C. sacchari*.

*Cytospora salicina* Norph. *et al.*, Mycosphere 8: 80. 2017. (Fig. 1: Clade 67)

*Typus*: **Russia**, Rostov Region, Krasnosulinsky District, Donskoye forestry, riparian forest, on dead and dying branches of *Salix alba*, 18 Jun. 2015, *T. Bulgakov* (**holotype** MFLU 15-2212, ex-holotype culture MFLUCC 15-0862).

Description: See Norphanphoun et al. (2017).

*Materials examined*: **Italy**, on *Vitis vinifera*, 1977, deposited by *G.B. Mascher* (culture CBS 507.77). **Iran**, West Azerbaijan Province, Miandoab, on *Vitis vinifera*, 2010, *S. Moshari* (culture CBS 141626 = CPC 19921; CBS 141629 = CPC 19925). **Turkey**, Besevler, Ankara, Gazi University Hospital, from human sinus, 2004, unknown *collector* (culture CBS 115107).

*Notes*: *Cytospora salicina* was described from *Salix alba* (Norphanphoun *et al.* 2017). Although CBS 115107 was identified as *Valsa sordida* (= *C. chrysosperma*) based on ITS sequence data by Kalkanci *et al.* (2006), isolates CBS 115107 and MFLUCC 15-0862 (the ex-holotype of *C. salicina*) cluster together with high support (ML/BI = 100/1) and differ from *C. chrysosperma* (CBS 197.50) in the ITS (4/514 bp difference), *act1* (16/255 bp difference), *rpb2* (15/650 bp difference), *tef1-a* (23/492 bp difference) and *tub2* (17/411 bp difference) loci in the current study. Therefore, it was reclassified as *C. salicina* in the present study.

*Cytospora salicis* (Corda) Rabenh., Deutschl. Krypt. Fl. (Leipzig) 1: 1340. 1844. (Fig. 1: Clade 103)

*Basionym: Naemaspora salicis* Corda, Icon. Fung. (Prague) 3: 26. 1839.

Typus: Czech Republic, unknown host (holotype PRM 155584).

*Material examined*: **Italy**, Südtirol, Sexten, near Moos, Porzenwald, *Salix fragilis*, 1 Sep. 2000, *W. Jaklitsch & H. Voglmayr*, WJ 1580 (BPI 748461, culture CBS 109754).

*Notes*: Use of the epithet "salicis" presents a number of problems. Despite its use in the protologue only Betula (in particular Betula carpathica) and not Salix is mentioned as a host (Rabenhorst 1844). The holotype is lodged at PRM consisting of a single twig fragment, which from the bark characteristics could be Salix, but the specimen has not been investigated in detail in the present study. Pending further investigation, CBS 109754 on Salix fragilis is tentatively identified as Cytospora salicis. See note for Cytospora fuckeliana (Fig. 1: Clade 108). Cytospora salicis clusters sister to C. polyspora and C. tongzhouensis but has the following nucleotide differences from the sequences for C. polyspora (CFCC 55834) and C. tongzhouensis (CFCC 58947). In ITS: 0/510 bp and 0/510 bp, respectively; in act1: 5/241 bp and 7/241 bp, respectively; in *rpb2*: 12/650 bp and 7/650 bp, respectively; in *tef1-a*: 22/504 bp and no *tef1-\alpha* available for *C. tongzhouensis*, respectively; in *tub2*: 11/396 bp and 11/397 bp, respectively.

*Cytospora sanbaensis* L. Lin & X.L. Fan, *sp. nov.* MycoBank MB 850174. Fig. 45. (Fig. 1: Clade 50)

*Etymology*: Name refers to Sanba Naxi Town where it was collected.

*Typus*: **China**, Yunnan Province, Diqing Tibetan Autonomous Prefecture, Shangri-La City, Sanba Naxi Town, East Ring Road, 27°34'26"N, 100°1'5"E, on branches of *Populus adenopoda*, 9 Aug. 2022, *L. Lin* & *M. Lin* (**holotype** BJFC-S2066, ex-holotype culture CFCC 58242); 27°34'27"N, 100°1'5"E, on branches of *Populus adenopoda*, 9 Aug. 2022, *L. Lin* & *M. Lin* (**paratype** BJFC-S2067, ex-paratype culture CFCC 58243).

Description: Pseudostromata Group SIII (type s8), immersed in the bark, scattered, with 4–7 perithecia arranged irregularly. *Conceptacle* absent. *Ectostromatic disc* dark brick to sepia, usually surrounded by ostiolar necks, rhombic to circular, 790–950 µm diam, with 3–8 irregularly arranged ostioles. *Ostioles* black when mature, 50–80 µm diam. *Perithecia* buff to black when mature, flask-shaped to spherical, arranged in a circle in pseudostroma, 195–295 µm diam. *Asci* hyaline, with chitinoid, refractive ring, clavate to elongate-obovoid, 64–81.5 × 13–20.5 µm (av. = 71.8  $\pm$  4.0 × 15.0  $\pm$  1.7 µm, n = 30), 4-spored. *Ascospores* hyaline, biseriate to multiseriate, elongate-allantoid, aseptate, 19–28.5 × 5–8.5 µm (av. = 23.4  $\pm$  2.6 × 6.9  $\pm$  1.0 µm, n = 50).

*Culture characteristics*: Cultures on PDA initially white, growing up to 1.5 cm diam after 3 d, entirely covering the 9-cm-diam Petri dish after 7 d, fawn (surface) and cinnamon (reverse) after 20 d, flat with a uniform texture.

*Notes*: *Cytospora sanbaensis* was associated with canker disease on *Populus adenopoda* in China. It is characterised by having asci with four ascospores, which is similar to *C. populina* and *C. populinopsis* (Fan *et al.* 2015b). However, *Cytospora sanbaensis* can be distinguished from them by its wider ascospores (5–8.5 µm vs 4.4–5.8 µm in *C. populina* and 3–4.5 µm in *C. populinopsis*) (Fan *et al.* 2015b). In the ITS, *act1*, *rpb2*, *tef1-a* and *tub2* gene trees, isolates CFCC 58242 and 58243 all form a distinct clade with maximum support (ML/BI = 100/1 in all five genes). Therefore, they are consequently described as a new species here.

**Cytospora schulzeri** Sacc. & P. Syd., Syll. fung. (Abellini) 14: 918. 1899. (Fig. 1: Clade 51)

Synonyms: Valsa malicola Z. Urb., Česká Mykol. 10: 209. 1956. Cytospora parasitica Norph. et al., Fungal Diversity 75: 172. 2015.





**Fig. 45.** *Cytospora sanbaensis* (BJFC-S2066). **A**, **B**. Habit of pseudostroma on twig. **C**. Transverse section through pseudostroma with ascomata. **D**. Longitudinal section through pseudostroma with ascomata. **E**, **F**. Asci. **G**. Ascospores. Scale bars: A = 1 mm; B = 200 μm; C, D = 500 μm; E–G = 10 μm.

Typus: Requires typification.

*Description*: See Hayova & Minter (1998c) (as *Valsa malicola*), Ariyawansa *et al.* (2015).

*Materials examined*: **USA**, Michigan, on *Malus sylvestris*, unknown date, *T. Proffer* (culture CBS 118570). **Switzerland**, on *Phragmites australis*, unknown date and *collector* (culture CBS 188.42).

Notes: Cytospora schulzeri (= Valsa malicola), a common species causing apple canker disease, was first described as Cytospora capitata (Saccardo 1884). Cytospora schulzeri is a replacement name for Cytospora capitata Schulzer & Sacc. 1884, which is a later homonym of Cytospora capitata Fuckel 1874, and therefore is an illegitimate name (ICN Art. 53.1). Schulzer types are difficult to locate, and may have been lost in the case of Cytospora capitata Schulzer & Sacc. 1884 (= Cytospora schulzeri). A type may also be present in the Saccardo herbarium in PAD. The type specimen of Valsa malicola (=Cytospora schulzeri) was collected on Malus sylvestris subsp. mitis from Europe (PR, Urban 4246). However, as this is a younger heterotypic synonym, it is not relevant for the typification of Cytospora schulzeri. Therefore, a typification of Cytospora schulzeri is needed. Adams et al. (2006) treated CBS 118570 as C. schulzeri (= Valsa malicola) which can be treated as a reference isolate before further typification.

Fan et al. (2020) identified the isolates CFCC 50040 and 50042 as *C. schulzeri* based on the presence of multiple ostioles and the host *Malus pumila*. However, the isolates CFCC 50040 and 50042 formed a distinct lineage with CBS 118570 (Fig. 1). These isolates clustered with CBS 118570 and were collected from the Northern Hemisphere (China, Russia, Switzerland, USA), including Europe, and the USA where the holotype of *V. malicola* was collected. This clade corresponds with the characteristics of *C. schulzeri* that is known to be widely distributed and causes apple disease in many different parts of the world. It is therefore more likely to accurately represent *C. schulzeri*. Ariyawansa *et al.* (2015) introduced *C. parasitica* as a new species isolated in Russia from dying branches of *Malus domestica*. However, *C. parasitica* (MFLUCC 15-0507) and *C. schulzeri* (CBS 118570) clustered together in the ITS, *act1* and *rpb2* trees (no *tef1-\alpha* and *tub2* available for MFLUCC 15-0507). *Cytospora parasitica* is thus treated as a synonym of *C. schulzeri*.

*Cytospora shaanxiensis* L. Lin & X.L. Fan, *sp. nov.* MycoBank MB 850175. Fig. 46. (Fig. 1: Clade 19)

*Etymology*: Name refers to the location where it was collected, Shaanxi Province (China).

*Typus*: **China**, Shaanxi Province, Xi'an City, Chang'an District, Mount Wutai, 34°2'41.55"N, 108°53'56.08"E, on branches of *Lindera obtusiloba*, 5 Aug. 2020, *C. Peng & C.M. Tian* (holotype BJFC-S2169, ex-holotype culture CFCC 56032; **paratype** BJFC-S2170, ex-paratype culture CFCC 56033).

Description: Conidiomata Group AIII (type a11), pycnidial, immersed in bark, scattered, erumpent through the surface, with multiple locules. Conceptacle absent. Ectostromatic disc cinnamon to smoke grey, ovoid to circular, 172–220(–285) µm diam, with one ostiole per disc. Ostiole in the centre of the disc, conspicuous, mouse grey to black, (37.5-)43-57(-73) µm diam. Locule numerous, subdivided frequently by invaginations with independent walls, (1 268.5–)1 330–1 600(–1 686) µm diam. Conidiophores borne along the locules, hyaline, unbranched or occasionally branched at the base or in the middle,  $(8.5-)10-16.5(-19.5) \times 1-1.5$  µm, embedded in a gelatinous layer. Conidiogenous cells enteroblastic, phialidic, subcylindrical to cylindrical. Conidia hyaline, allantoid, eguttulate, smooth, aseptate, thin-walled,  $4-5(-5.5) \times 1-1.5$  (av. =  $4.6 \pm 0.3 \times 1.3 \pm 0.1$ , n = 30) µm.

*Culture characteristics*: Colonies flat, felt-like, thin with a uniform texture, lacking aerial mycelium, growing up to 8 cm diam and becoming honey after 6 d, after 2 wk deepened in later stage gradually and becoming hazel after 14 d. Conidiomata are randomly distributed on the medium surface.



**Fig. 46.** *Cytospora shaanxiensis* (BJFC-S2169). **A**, **B**. Habit of conidiomata on twig. **C**. Transverse section through conidioma. **D**. Longitudinal section through conidioma. **E**. Conidiophores and conidiogenous cells. **F**. Conidia.Scale bars: A = 1 mm; B–D = 500 μm; E, F = 10 μm.

Notes: Cytospora shaanxiensis can be characterised by the multiple locules with independent walls and conidia of size  $4-5 \times 1-1.5 \,\mu\text{m}$ . It occurs on the same host genus, Lindera, as C. henanensis and C. nanyangensis. Morphologically, C. shaanxiensis has smaller conidia (4–5 × 1–1.5  $\mu$ m), compared to the longer conidia of C. henanensis and C. nanyangensis (5-5.5 × 1-1.5, 5.5-6 × 1-2 µm). In addition, molecular phylogenies show them to be distinct. Cytospora shaanxiensis clusters together with C. tetraspora. The ITS gene phylogenies do not resolve C. shaanxiensis from its closest sister species, C. tetraspora. However, the act1, rpb2, tef1-a and tub2 gene phylogenies place it as a lineage distinct from the other species with high support (ML/BI = 100/0.76, 98/0.8, 100/1 and 100/1, respectively). Furthermore, it can be distinguished from C. tetraspora by smaller locules subdivided by invaginations (1 330-1 600 vs 2 050-2 435 µm) and smaller conidia (4-5 × 1-1.5 vs 5.5-6.5 × 1.5-2 µm).

*Cytospora shangrilaensis* L. Lin & X.L. Fan, *sp. nov.* MycoBank MB 850176. Fig. 47. (Fig. 1: Clade 125)

*Etymology*: Name refers to Diqing Tibetan Autonomous Prefecture, Shangri-La City (China), where it was collected.

*Typus*: **China**, Yunnan Province, Diqing Tibetan Autonomous Prefecture, Shangri-La City, Sanba Naxi Town, East Ring Road, 27°34'26"N, 100°1'5"E, on branches of *Populus adenopoda*, 9 Aug. 2022, *L. Lin & M. Lin* (**holotype** BJFC-S1965, ex-holotype culture CFCC 58247); *ibid.*, 27°34'28"N, 100°1'25"E, on branches of *Populus adenopoda*, 9 Aug. 2022, *L. Lin & M. Lin* (**paratype** BJFC-S1966, ex-paratype culture CFCC 58248).

Description: Conidiomata Group AIII (type a12), immersed in bark, erumpent when mature, discoid to conical, 390–650 µm diam, with multi-locule. Conceptacle prominent. Ectostromatic disc buff to isabelline, circular to ovoid, 255–285 µm diam, with single ostiole

per disc in the centre. *Ostiole* circular to ovoid, buff to black, 75–95 µm diam. *Locules* numerous, irregularly arranged with individual walls. *Conidiophores* hyaline, unbranched or occasionally branched, 11–20.5 × 1.5–2.5 µm (av. =  $16.0 \pm 2.5 \times 1.7 \pm 0.18$  µm, n = 30). *Conidiogenous cells* enteroblastic, phialidic, subcylindrical to cylindrical, 3.5–4.5 × 1–1.5 µm (av. =  $4.0 \pm 0.3 \times 1.3 \pm 0.1$  µm, n = 30). *Conidia* hyaline, unicellular, eguttulate, elongate-allantoid,  $4-6 \times 1-2$  µm (av. =  $5.3 \pm 0.3 \times 1.6 \pm 0.2$  µm, n = 50).

*Culture characteristics*: Cultures on PDA initially white, growing up to 3 cm diam after 3 d, entirely covering the 9-cm-diam Petri dish after 11 d and becoming honey to isabelline after 14 d with irregular edge, isabelline to black after 20 d, flat with a uniform texture.

Additional material examined: **China**, Yunnan Province, Diqing Tibetan Autonomous Prefecture, Shangri-La City, Sanba Naxi Town, East Ring Road, 27°34'26"N, 100°1'5"E, on branches of *Populus adenopoda*, 9 Aug. 2022, *L. Lin & M. Lin* (BJFC-S1967, culture CFCC 59055).

Notes: Cytospora shangrilaensis is phylogenetically closely related to *C. davidiana* and *C. notastroma* (Fig. 1). However, *C. shangrilaensis* (CFCC 58247) differs from *C. davidiana* (CXY 1350) in the ITS (6/503) and *tub2* (31/407) gene regions (no *act1, rpb2* and *tef1-α* available for *C. davidiana*), and differs from *C. notastroma* (NE TFR8) in ITS (16/503) and *tef1-α* (41/255) gene regions (no *act1, rpb2* and *tub2* available for *C. notastroma*). Morphologically, it differs from *C. davidiana* by having a black conceptacle and from *C. notastroma* by its larger conidia, (4–6 × 1–2 vs 2 × 0.5 µm) (Kepley *et al.* 2015, Wang *et al.* 2015). Additionally, the conidiomata of *C. shangrilaensis* are leucotoselloid, but in *C. notastroma* they are leucocytosporoid (Kepley *et al.* 2015). Therefore, *C. shangrilaensisi* is identified as a new species.

Cytospora sidaohensis L. Lin & X.L. Fan, sp. nov. MycoBank MB 850169. Fig. 48. (Fig. 1: Clade 8)





Fig. 47. Cytospora shangrilaensis (BJFC-S1965). A, B. Habit of conidiomata on twig. C. Transverse section through conidioma. D. Longitudinal section through conidioma. E. Conidiophores and conidiogenous cells. F. Conidia. Scale bars: B–D = 200 µm; E, F = 10 µm.

*Etymology*: Name refers to Sidaohe County where the holotype was collected.

*Typus*: **China**, Beijing City, Huairou District, Baoshan Town, Sidaohe County, Forestry Centre of Beijing University of Agriculture, 40°57'37.47"N, 116°28'38.12"E, on branches of *Corylus heterophylla*, 18 Jun. 2021, *X.L. Fan & Y.K. Bai* (holotype BJFC-S2115, ex-holotype culture CFCC 56042; paratype BJFC-S2116, ex-paratype culture CFCC 56043).

Description: Pseudostromata immersed in the bark, erumpent slightly through the bark surface, scattered, circular, (1 980–) 2 090–2 320(–2 510)  $\mu$ m diam, with 8–10 perithecia arranged circularly. Conceptacle absent. Ectostromatic disc hazel, circular to ovoid, (470–)520–890(–970)  $\mu$ m diam, with ostioles arranged circularly arranged in the disc. Ostioles black, (155–)170–270(–410)  $\mu$ m diam. Perithecia flask-shaped to spherical, (850–)920– 1 060(–1 100)  $\mu$ m diam. Asci free, clavate to elongate-obovoid, (50–)53.5–64.5(–70) × (10.5–)11.5–13.5(–15.5)  $\mu$ m, 8-spored. Ascospores biseriate, elongate-allantoid, thin-walled, hyaline,

aseptate,  $(15-)16.5-18(-19) \times (3-)3.5-5.5$  (av. =  $16.8 \pm 0.7 \times 4.2 \pm 0.6$ , n = 30) µm. Asexual morph not observed.

*Culture characteristics*: Colonies white, entirely covering the 9-cmdiam Petri dish after 4 d and becoming pale straw-yellow, thick with a regular edge. Sterile.

*Notes: Cytospora sidaohensis* is associated with canker disease of *Corylus heterophylla*. This species is characterised by having 8–10 perithecia in a pseudostroma without a conceptacle, 8-spored asci and biseriate ascospores. In the phylogenetic analyses, *C. sidaohensis* forms a sister group with *C. platycladicola* with full support (ML/BI = 100/1), but differs from that species in its smaller ascospores (16.5–18 × 3.5–5.5 vs 12–12.5 × 3–4 µm) (Fan *et al.* 2020). In the *act1*, *rpb2*, *tef1-α* and *tub2* gene trees, the BI and ML phylogenies all separate it from its closest sister species, *C. platycladicola* (ML/BI = 82/0.998, 97/0.87, 100/0.999 and 100/1, respectively).



Fig. 48. Cytospora sidaohensis (BJFC-S2115). A, B. Habit of pseudostromata on twig. C. Transverse section through pseudostroma with ascomata. D. Longitudinal section through pseudostroma with ascomata. E. Asci. F. Ascospores. Scale bars: A = 2 mm; B–D = 500 μm; E, F = 10 μm.

Cytospora sinensis L. Lin & X.L. Fan, sp. nov. MycoBank MB 850178. Fig. 49. (Fig. 1: Clade 65)

Etymology: Named after the country where it was collected, China.

*Typus*: **China**, Gansu Province, Lanzhou City, Yuzhong County, Guantangou, 35°53'19"N, 103°56'21"E, on branches of *Populus simonii*, 10 Jul. 2022, *L. Lin & X. H. Wang* (**holotype** BJFC-S2071, ex-holotype culture CFCC 58235); *ibid.*, 35°53'15"N, 103°56'22"E (**paratype** BJFC-S2068, ex-paratype culture CFCC 58224).

Description: Conidiomata Group AII (type a6), immersed in bark, erumpent when mature, discoid to conical, 420–560 µm diam, with multi-locule. Conceptacle absent. Ectostromatic disc honey to isabelline, circular to ovoid, 95–250 µm diam, with single ostiole per disc in the centre. Ostiole circular to ovoid, black, 35–110 µm diam. Locules numerous, divided with shared walls. Conidiophores hyaline, branched at the base, in the middle, or unbranched, 10–20.5 × 1–2 µm (av. =  $14.9 \pm 3.0 \times 1.6 \pm 0.3$  µm, n = 30). Conidiogenous cells enteroblastic, phialidic, subcylindrical to cylindrical,  $3.5-5.5 \times 1-1.5$  µm (av. =  $3.9 \pm 0.7 \times 1.3 \pm 0.1$  µm, n = 30). Conidia hyaline, unicellular, eguttulate, elongate-allantoid, 4–6 × 1–2 µm (av. =  $4.7 \pm 0.4 \times 1.6 \pm 0.14$  µm, n = 50).

*Culture characteristics*: Cultures on PDA initially white, growing up to 7 cm diam after 3 d, entirely covering the 9-cm-diam Petri dish after 4 d and becoming olivaceous after 10 d.

Additional materials examined: China, Gansu Province, Lanzhou City, Yuzhong County, Guantangou, 35°53'15"N, 103°56'28"E (BJFC-S2069, culture CFCC 58231); *ibid.*, 35°53'16"N, 103°56'21"E (BJFC-S2070, culture CFCC 58234); *ibid.*, 35°53'14"N, 103°56'22"E (BJFC-S2072, culture CFCC 58236); *ibid.*, 35°53'11"N, 103°56'22"E (BJFC-S2078, culture CFCC 59058); *ibid.*, 35°53'11"N, 103°56'26"E (BJFC-S2079, culture CFCC 59063); Yunnan Province, Diqing Tibetan Autonomous Prefecture, Deqin County, Shengping Town, 28°28'30"N, 98°55'5"E, on branches of *Populus haoana*, 9 Aug. 2022, *L. Lin & M. Lin* (BJFC-S2074, culture CFCC 58466); *ibid.*, Yunling County, S233 Highway, 28°12'22"N, 98°51'37"E, on branches of *Populus yunnanensis*, 7 Aug. 2022, *L. Lin & M. Lin* (BJFC-S2075, culture CFCC 58470; BJFC-S2076, culture CFCC 58471); *ibid.*, Weixi Lisu Autonomous County, Badi Town, 27°49'37"N, 99°1'58"E, on branches of *Populus canadensis*, 9 Aug. 2022, *L. Lin &*  *M. Lin* (BJFC-S2077, culture CFCC 58493); Dali City, Xiaguan District, 25°39'9"N, 100°10'50"E, on branches of *Populus canadensis*, 4 Aug. 2022, *L. Lin & M. Lin* (BJFC-S2073, culture CFCC 58239).

*Notes*: Phylogenetic analyses suggest that *C. sinensis* resides in the *C. chrysosperma* complex and is closely related to *C. chrysosperma* and *C. sophoriopsis* (Fig. 1). However, *C. sinensis* has the following nucleotide differences from the sequences for *C. chrysosperma* and *C. sophoriopsis*: in ITS: 1/513 bp and 0/513 bp, respectively; in *act*: 7/255 bp and 10/255 bp, respectively; in *rpb2*: 6/650 bp and 8/726 bp, respectively; in *tef1-a*: 11/294 bp and 5/294 bp, respectively; in *tub2*: 17/408 bp and 14/408 bp, respectively. Morphologically, *C. sinensis* differs from *C. chrysosperma* in having smaller conidiogenous cells (3.5–5.5 × 1–1.5 vs 6.1–8.1 × 1.5–1.9 µm) and from *C. sophoriopsis* in having longer conidiophores (10– 20.5 × 1–2 vs 8–15 × 1–1.5 µm; Lawrence *et al.* 2018, Fan *et al.* 2020).

*Cytospora songshanensis* L. Lin & X.L. Fan, *sp. nov.* MycoBank MB 850179. Fig. 50. (Fig. 1: Clade 9)

*Etymology*: Name refers to the Songshan National Nature Reserve (China) where it was collected.

*Typus*: **China**, Beijing City, Yanqing District, Songshan National Nature Reserve, 40°30'5.21"N, 115°50'3.35"E, on twigs and branches of *Quercus mongolica*, 3 Aug. 2021, *Y.K. Bai & X.L. Fan* (**holotype** BJFC-S2220, exholotype culture CFCC 56685); *ibid.*, Huairou District, Forestry Centre of Beijing University of Agriculture, 39°58'58.21"N, 115°26'52.27"E, on branches of *Platycladus orientalis*, 20 Jun. 2021, *X.L. Fan & Y.K. Bai* (**paratype** BJFC-S2221, ex-paratype culture CFCC 56351).

Description: Conidiomata Group AII (type a9), pycnidial, scattered, immersed in bark, erumpent slightly through the bark surface, discoid to conical, with multiple locules. Conceptacle absent. Ectostromatic disc buff to dark, circular, (420–)500–630(–900)  $\mu$ m diam, with one ostioles per disc. Ostiole grey to black, (200–)270–360(–440)  $\mu$ m diam. Locules numerous, arranged circularly with common walls, (700–)850–980(–1 110)  $\mu$ m diam. Conidiophores borne along the locules, hyaline, branched at base, in the middle or unbranched, (14.5–)16–18.5(–22) × 1–2  $\mu$ m, embedded in a



**Fig. 49.** *Cytospora sinensis* (BJFC-S2071). **A.** Habit of conidiomata on twig. **B.** Transverse section through conidioma. **C.** Longitudinal section through conidioma. **D, E.** Conidiophores and conidiogenous cells. **F.** Conidia. Scale bars: A = 1 mm; B, C = 200 μm; D–F = 10 μm.





Fig. 50. Cytospora songshanensis (BJFC-S2220). A, B. Habit of conidiomata on twig. C. Transverse section through conidioma. D. Longitudinal section through conidioma. E. Conidiophores and conidiogenous cells. F. Conidia. Scale bars: A = 2 mm; B–D = 1 mm; E, F = 10 µm.

gelatinous layer. Conidiogenous cells enteroblastic, phialidic, subcylindrical to cylindrical. Conidia hyaline, allantoid, thin-walled, eguttulate, aseptate, smooth, (4–)4.5–5.5(–6) × 1–2 (av. = 5.1  $\pm$  0.5 × 1.4  $\pm$  0.2, n = 30) µm.

*Culture characteristics*: Colonies initially white, growing up to 8 cm diam after 7 d, flat with a uniform texture and thicken gradually, ultimately lacking aerial mycelium, thin with a regular edge. Conidiomata randomly distributed on medium surface.

Additional material examined: China, Beijing City, Huairou District, Forestry Centre of Beijing University of Agriculture, 39°58'58.21"N, 115°26'52.27"E, on branches of *Platycladus orientalis*, 20 Jun. 2021, *X.L. Fan & Y.K. Bai* (BJFC-S2222, culture CFCC 56352).

Notes: Cytospora songshanensis and C. platycladicola were described from Platycladus orientalis. Cytospora songshanensis has conidia with similar dimensions, ( $4.5-5.5 \times 1-2 \ \mu m \ vs \ 4.5-5$ 

× 1.5–2 µm) to the latter species (Fan *et al.* 2020). However, *C.* songshanensis differs from *C. platycladicola* in the ITS (11/651), *act1* (3/392), *rpb2* (8/732), *tef1-* $\alpha$  (46/770) and *tub2* (18/707) gene regions (Fan *et al.* 2020).

*Cytospora sophoriopsis* X.L. Fan & C.M. Tian, Persoonia 45: 39. 2020. (Fig. 1: Clade 64)

*Typus*: **China**, Gansu Province, Gannan City, Diebu County, 34°04'05.76"N, 103°11'33.63"E, on dead branches of *Styphnolobium japonicum*, 10 Aug. 2012, *X.L. Fan* (**holotype** BJFC-S713 = BJFC-CGHs10, ex-holotype culture CFCC 89600).

Description: See Fan et al. (2020).

*Materials examined*: **China**, Beijing City, Daxing District, 116°27'19.44"E, 39°46'34.68"N, on branches of *Populus* sp., 18 Jul. 2017, *H.Y. Zhu* (BJFC CF20191210, culture CFCC 54061); *ibid.*, Mentougou District, Baihuashan

National Nature Reserve, 39°50'49"N, 115°33'12"E, on twigs and branches of *Populus beijingensis*, 23 Aug. 2022, *L. Lin & X.L. Fan* (BJFC-S2081, culture CFCC 59072); Inner Mongolia Autonomous Region, Huh hot City, 111°9'48.89"E, 40°43'46.45"N, on branches of *Populus* sp., 30 May 2012, *X.L. Fan* (BJFC CF20191235, culture CFCC 54070); Yunnan Province, Diqing Tibetan Autonomous Prefecture, Deqin County, Shengping Town, 28°28'2"N, 98°55'1"E, on branches of *Populus szechuanica*, 10 Jul. 2022, *L. Lin & M. Lin* (BJFC-S2080, culture CFCC 58464).

Notes: Cytospora sophoriopsis was described on Styphnolobium japonicum in China (Fan *et al.* 2020) and clusters as sister species to *C. chrysosperma*, but differs in DNA data (see notes of *C. chrysosperma*). It has a labyrinthine cytosporoid conidiomata without a black conceptacle (Group AII, type a6) (Fan *et al.* 2020). Lin *et al.* (2023a) extended its host range to include *Populus* spp., and confirmed that *C. sophoriopsis* was able to cause serious canker disease of *Populus alba* var. *pyramidalis*.

*Cytospora sorbariae* A.L. Jia & X.L. Fan, Mycokeys 101: 184. 2024. Fig. 51. (Fig. 1: Clade 28)

*Typus*: **China**, Beijing City, Fengtai District, Beijing Garden Expo, 39°52'35.65"N, 116°11'4.02"E, from branches of *Sorbaria sorbifolia*, 7 Apr. 2023, *A.L. Jia* & *X.L. Fan* (**holotype** BJFC CF20230417, ex-holotype living culture CFCC 59445); *ibid.*, 39°52'35.43"N, 116°11'4.62"E, from branches of *Sorbaria sorbifolia*, 7 Apr. 2023, *A.L. Jia* & *X.L. Fan* (**paratype** BJFC CF20230419, ex-paratype living culture CFCC 59529).

Description: Conidiomata Group AII (type a9), pycnidial, immersed in bark, scattered, erumpent through the bark surface, discoid, with multiple locules. Conceptacle absent. Ectostromatic disc prominent, buff to grey, ovoid,  $(375-)480-560(-650) \mu m$  diam, with 1–2 ostiole(s) per disc. Ostiole in the centre of the disc, conspicuous, grey to black, at the same level as the disc surface, 140–195(–225) µm diam. Locule numerous, subdivided frequently by invaginations with common walls, (1 610–)1 820–2 365(–2 700) µm diam. Conidiophores and conidiogenous cells inconspicuous. Conidia hyaline, allantoid, eguttulate, smooth, aseptate, thinwalled, 5.5–6.5 × 1.5–2 (av. = 6 ± 0.2 × 1.8 ± 0.1, n = 30) µm. *Culture characteristics*: Colonies on PDA are initially white, producing mycelium covering the 9-cm-diam Petri dish after 6 d with a uniform texture, lacking aerial mycelium, with a regular margin. Sterile.

*Materials examined*: **China**, Beijing City, Huairou District, Forestry Centre of Beijing University of Agriculture, 40°52′48.54″N, 116°26′24.68″E, on branches of *Corylus heterophylla*, 14 Jun. 2021, *X.L. Fan & Y.K. Bai* (BJFC-S2124, culture CFCC 56025, CFCC 56026); *ibid.*, Mentougou District, Xiaolongmen Forest Centre, 115°34′35″E, 39°50′4″N, on branches of *Betula* sp., 23 Aug. 2022, *X.L. Fan & L. Lin* (BJFC-S2096, culture CFCC 58456).

*Notes: Cytospora sorbariae* was described from *Sorbaria sorbifolia* in Beijing, China (Jia *et al.* 2024). In the current study, three isolates are shown to be *C. sorbariae* based on fully supported phylogenetic topology (Fig. 1), same asexual morphological type (Group AII, type a9) and overlapped conidia size from previous descriptions (Jia *et al.* 2024). A new record of *C. sorbariae* from *Corylus heterophylla* and *Betula* sp. are included.

*Cytospora suecica* Jami, Crous & M.J. Wingf., *sp. nov.* MycoBank MB 850177. Fig. 52. (Fig. 1: Clade 71)

Etymology: Name refers to Sweden where it was collected

*Typus*: **Sweden**, red rot in sapwood of green log of *Populus tremula*, 1950, *U. Bärlund* (**holotype** CBS H-22924, ex-holotype culture CBS 450.51).

Description: Conidiomata produced on twigs on MEA after 6–8 wk, stromata up to 500 µm diam, multilocular, exuding a creamy conidial cirrhus or globoid conidial mass. Conidiophores scarce, hyaline, smooth, branched, aseptate,  $3-6 \times 0.5-1.3$  µm, embedded in a gelatinous layer. Conidiogenous cells phialidic, with periclinal thickening, subcylindrical, tapering towards apices, collarettes minute,  $2.5-12 \times 1.2-2$  µm, lateral along branching conidiophores. Conidia hyaline, smooth, guttulate, ellipsoidal, apex subobtusely rounded, tapering to a subtruncate base, aseptate,  $0.5-2 \times 0.5-1$  µm.



**Fig. 51.** *Cytospora sorbariae* (BJFC-S2124). **A–C.** Habit of conidiomata on twig. **D.** Transverse section through conidioma. **E.** Longitudinal section through conidioma. **F.** Conidia. Scale bars: A = 1 mm; B–E = 500 μm; F = 5 μm.





Fig. 52. Cytospora suecica (CBS 450.51). A. Conidiomata on MEA. B. Conidiophores and conidiogenous cells. C. Conidia. Scale bars: A = 100 µm; B, C = 5 µm.

*Culture characteristics*: Colonies covering the Petri dish after 20 d at 24 °C, with aerial mycelium flat, sparse to moderate. On MEA surface olivaceous, brown in reverse. On OA surface olivaceous.

*Notes*: Isolate CBS 450.51 was deposited as *V. ambiens* in the CBS collection, but DNA analysis revealed it to be a new species. *Cytospora suecica* is phylogenetically close to *C. Ihasaensis* (Fig. 1), but differs in smaller conidia  $(0.5-2 \times 0.5-1 \mu m \text{ in } C. suecica vs 3.5-5 \times 1-1.5 \mu m \text{ in } C. Ihasaensis$ ) and in their DNA data (see notes of *C. Ihasaensis*). *Cytospora suecica* is the second *Cytospora species* isolated from *Populus tremula* in Europe, the first species being *C. chrysosperma* from the UK.

*Cytospora syringina* L. Lin & X.L. Fan, *sp. nov.* MycoBank MB 850180. (Fig. 1: Clade 147)

Etymology: Name refers to Syringa, the host genus of the holotype.

*Typus*: **China**, Qinghai Province, Haidong City, Pinan County, Pinan Town, 36°29'25.92"N, 102°08'34.1"E, from twigs and branches of *Syringa oblata*, 15 Aug. 2012, *X.L. Fan* (**holotype** BJFC-S636, ex-holotype culture CFCC 50036).

Description: See Fan et al. (2020) (as C. pruinosa).

Additional material examined: China, Qinghai Province, Haidong City, Pinan County, Pinan Town, 36°29'25.92"N, 102°08'34.11"E, from twigs and branches of *Syringa oblata*, 15 Aug. 2012, *X.L. Fan* (BJFC-S640, culture CFCC 50037).

Notes: Cytospora syringina was originally identified as *C. pruinosa* (Fan *et al.* 2020). However, it formed a distinct clade differing from *C. pruinosa* in the present study (Fig. 1). Phylogenetically, *C. syringina* (CFCC 50036) differs from *C. pruinosa* (CBS 200.42) in ITS (3/513), *act1* (12/215), *tef1-α* (61/502) and *tub2* (33/411) loci (no *rpb2* available for *C. syringina*). Morphologically, *C. syringina* can be distinguished from *C. pruinosa* by its smaller locules (430–550 vs 690–935 µm) and larger conidia (5.5–7 × 1.5 vs 5–5.5 × 1.5–2 µm) (Fan *et al.* 2020).

*Cytospora tamaricicola* X.L. Fan & C.M. Tian, Persoonia, 45: 41. 2020. (Fig. 1: Clade 180)

*Typus*: **China**, Yunnan Province, Kunming City, Kunming World Expo, on branches of *Tamarix chinensis*, 18 Mar. 2015, *B. Cao*, *Q. Yang & Z. Du* (**holotype** CF-2015510, ex-holotype culture CFCC 50508).

Description: See Fan et al. (2020).

Materials examined: China, Yunnan Province, Dali City, Xiaguan District, Cangshan, 25°38'49"N, 100°10'8"E, on Rosaceae sp., 4 Aug. 2022, L. Lin & M. Lin (BJFC-S2085, culture CFCC 58268); Kunming City, Panlong District, Southwest Forestry University, 102°45'22"E, 25°3'43"N, on branches of Ligustrum lucidum, 11 Aug. 2022, L. Lin & Z.Q. Wu (BJFC-S2086, culture CFCC 58482; BJFC-S2087, culture CFCC 59052).

*Notes*: Fan *et al.* (2020) introduced *C. tamaricicola* and described its asexual and sexual morph. In the ITS, *act1, rpb2, tef1-* $\alpha$  and *tub2* gene trees, the BI and ML phylogenies all place it in a distinct clade, with high support (ML/BI = 95/1, 71/0.96, 100/1, 100/1 and 100/1, respectively). It is associated with canker disease of *Rosa multiflora* and *Tamarix chinensis* (Fan *et al.* 2020). The host range of this species is expanded in the present study to include *Ligustrum lucidum*. This species is known only for Yunnan, China.

*Cytospora tenebrica* L. Lin & X.L. Fan, *sp. nov.* MycoBank MB 850173. Fig. 53. (Fig. 1: Clade 158)

*Etymology*: Name refers to producing the darkness on the surface of host epidermis.

*Typus*: **China**, Beijing City, Haidian District, the National Botanical Garden, 40°0'27.21"N, 116°12'58.26"E, on branches of *Salix babylonica* 'Tortuosa', 8 May. 2021, *M. Pan & X. Zhou* (**holotype** BJFC-S2197, ex-holotype culture CFCC 55841; **paratype** BJFC-S2198, ex-paratype culture CFCC 56269).

Description: Conidiomata Group AI (type a2), immersed in bark, erumpent through the surface of bark when mature, discoid to conical. Conceptacle conspicuous. Ectostromatic disc dark, circular to ovoid, (495–)690–1 050(–1 250)  $\mu$ m diam, with one ostiole per disc. Ostiole conspicuous, one ostiole per disc, circular to ovoid, dark brown at the same level as the disc surface, (135–) 225–300  $\mu$ m. Locule undivided, circular to ovoid, 1 035–1 285  $\mu$ m diam. Conidiophores hyaline, branched at base or occasionally unbranched, (8–)13–20.5(–21.5) × 1.5–2  $\mu$ m. Conidiogenous cells enteroblastic, phialidic, subcylindrical to cylindrical. Conidia



Fig. 53. Cytospora tenebrica (BJFC-S2197). A, B. Habit of conidiomata on twig. C. Transverse section through conidioma. D. Longitudinal section through conidioma. E. Conidiophores and conidiogenous cells. F. Conidia. Scale bars: A = 2 mm; B–D = 500 μm; E, F = 10 μm.

hyaline, eguttulate, elongate-allantoid, aseptate, (5.5–)6.5–8(–9) × 1.5–2 (av. = 7.3  $\pm$  0.6 × 1.8  $\pm$  0.2, n = 30) µm.

*Culture characteristics*: Colonies white, becoming salmon to buff and growing up to 5.5 cm diam after 7 d, flat, felt-like, with thin texture in centre, ultimately hazel and flat, with a uniform texture and concentric circles. Conidiomata randomly distributed on medium surface.

Notes: Cytospora tenebrica was associated with canker disease of Salix babylonica. Phylogenetically, it has a close affinity to *C. yuduensis*, but with clearly different ITS (3/516 bp), act (6/243 bp), rpb2 (5/712 bp), tef1- $\alpha$  (37/273 bp) and tub2 (7/382 bp) sequences. Morphologically, it can be distinguished from *C. yuduensis* based on a black area formed on the bark surface, larger ectostromatic discs (690–1 050 vs 150–210 µm) and larger ostioles (225–300 vs 50–70 µm).

*Cytospora tetraspora* L. Lin & X.L. Fan, *sp. nov.* MycoBank MB 850181. Fig. 54. (Fig. 1: Clade 18)

Etymology: Name refers to its 4-spored asci, Latin tetraspora.

*Typus*: **China**, Beijing City, Huairou District, Forestry Centre of Beijing University of Agriculture, 40°53'23.63"N, 116°26'54.87"E, on branches of *Tilia mongolica*, 10 Jun. 2021, *X.L. Fan & Y.K. Bai* (**holotype** BJFC-S2207, ex-holotype culture CFCC 56279); *ibid.*, 40°53'45.25"N, 116°25'16.44"E, on branches of *Quercus aliena*, 10 Jun. 2021, *X.L. Fan & Y.K. Bai* (**paratype** BJFC-S2208, ex-paratype culture CFCC 55847).

Description: Pseudostromata Group SIII (type s8), immersed in the bark, erumpent through the bark surface, scattered, circular to ovoid, (1 030–)1 315–1 490(–1 580) µm diam, with 4–8 perithecia arranged circularly in well-developed black entostromata. *Conceptacle* absent. *Ectostromatic disc* brown to black, circular to ovoid, (365–)450–560(–665) µm diam, with 4–8 ostioles arranged circularly in the disc. *Ostioles* brown to black, (90–)100–135(–150)



µm diam. Perithecia flask-shaped to spherical, (395-)420-510(-565) µm diam. Asci free, clavate to elongate-obovoid, (60-)64-72(-77) × (13-)14.5-16(-17) µm, 4-spored. Ascospores biseriate, elongate-allantoid, thin-walled, hyaline, aseptate, (19-)20.5-24.5(-25) × (5–)6–7 (av. = 22.2 ± 1.7 × 6.4 ± 0.4, n = 30) µm. Conidiomata Group All (type a6), pycnidial, immersed in bark, erumpent through the bark surface in a large area, discoid, with large multiple locules. Conceptacle absent. Ectostromatic disc grey to dark, nearly flat, circular to ovoid, (730–)750–790(–810) µm diam, with one ostioles per disc. Ostiole grey to black, inconspicuous, (345)-360-385(-395) µm diam. Locules numerous, arranged circularly with common walls, (1 890-)2 050-2 435(-2 490) µm diam. Conidiophores borne along the locules, hyaline, branched at base or occasionally not branched, (11.5-)13.5-22(-25.5) × 2-2.5 µm, embedded in a gelatnous layer. Conidiogenous cells enteroblastic, phialidic, subcylindrical to cylindrical. Conidia hyaline, allantoid, eguttulate, smooth, aseptate, thin-walled, (5-)5.5-6.5 × 1.5-2 (av. = 5.9 ± 0.5 × 1.8 ± 0.2, n = 30) µm.

*Culture characteristics*: Colonies initially white, growing up to 5 cm diam at 7 d and producing slightly yellow pigment with a uniform texture after 14 d, ultimately lacking aerial mycelium, thin with an irregular edge. Conidiomata randomly distributed on medium surface.

Additional materials examined: China, Beijing City, Huairou District, 40°52'32.14"N, 116°26'36.65"E, on branches of *Quercus mongolica*, 9 Jun. 2021, *X.L. Fan* & Y.K. Bai (BJFC-S2209, culture CFCC 55848); Yanqing District, Songshan National Nature Reserve, 40°29'35.16"N, 115°45'28.72"E, on twigs and branches of *Quercus aliena*, 5 Aug. 2021, *Y.K. Bai* & *X.L. Fan* (BJFC-S2212, culture CFCC 56674); *ibid.*, Mentougou District, Baihuashan National Nature Reserve, 39°50'37.44"N, 115°26'56.22"E, on twigs and branches of *Koelreuteria paniculata*, 22 Aug. 2021, *Y.K. Bai* & *X.L. Fan* (BJFC-S2210, culture CFCC 56316); *ibid.*, Mount Dongling, Xiaolongmen Forestry Centre, 39°59'23.58"N, 115°27'05.00"E, on twigs and branches of *Quercus mongolica*, 24 Aug. 2021, *Y.K. Bai* & *X.L. Fan* (BJFC-S2211, culture CFCC 56553); *ibid.*, 39°50'6"N, 115°34'35"E,



Fig. 54. Cytospora tetraspora (BJFC-S2207). A, B. Habit of pseudostromata on twig. C. Transverse section through pseudostroma with ascomata. D. Longitudinal section through pseudostroma with ascomata. E, F. Habit of conidiomata on twig. G. Transverse section through conidioma. H. Longitudinal section through conidioma. I. Asci. J. Ascospores. K. Conidiophores and conidiogenous cells. L. Conidia. Scale bars: A, E = 2 mm; B–H = 500 µm; I–L = 10 µm.

on twigs and branches of *Quercus mongolica*, 25 Aug. 2022, *L. Lin & X.L. Fan* (BJFC-S2088, culture CFCC 58457; BJFC-S2089, culture CFCC 58458; BJFC-S2090, culture CFCC 58604).

Notes: Cytospora tetraspora was isolated from Koelreuteria paniculata, Platycladus orientalis, Quercus aliena, Quercus mongolica and Tilia mongolica in China. Morphologically, it differs from most Cytospora species in having ascomata with 4-spored

asci. Three other species in this study, namely *C. populina*, *C. populinopsis* and *C. tetraspora*, also have four ascospores per ascus. However, it is easy to distinguish between them by the different sizes of their ascospores (18.9–24.3 × 4.4–5.8 vs 14–20 × 3–4.5 vs 20.5–24.5 × 6–7  $\mu$ m) (Fan *et al.* 2020). Moreover, *C. tetraspora* differs from *C. populina* in various gene regions: ITS (6/633), *act1* (38/382), *rpb2* (44/731), *tef1-a* (119/796) and *tub2* (64/723) and from *C. populinopsis*: ITS (9/633), *act1* (27/382), *rpb2* (46/731), *tef1-a* (125/796) and *tub2* (53/723).

*Cytospora tongzhouensis* X.L. Fan & C.M. Tian, *sp. nov.* MycoBank MB 850183. Fig. 55. (Fig. 1: Clade 107)

*Etymology*: Name refers to Tongzhou where it was collected.

*Typus*: **China**, Beijing City, Tongzhou District, the City Green Heart Forest Park, 39°52'59.87"N, 116°43'12.59"E, on branches of *Salix* sp., 10 Sep. 2021, *M. Pan & X.L. Fan* (**holotype** BJFC-S2213, ex-holotype culture CFCC 56779; **paratype** BJFC-S2214, ex-paratype culture CFCC 58947).

Description: Pseudostromata Group SI (type s2) immersed in the bark, erumpent slightly through the bark surface, scattered or serried, circular, (660–)740–820(–860) µm diam, with 4–6 perithecia circularly arranged in well-developed black entostromata. *Conceptacle* black. *Ectostromatic disc* brown to black, circular to ovoid, (140–)190–260(–270) µm diam. *Ostioles* black, (40–) 50–75(–90) µm diam. *Perithecia* flask-shaped to spherical, (120–)180–250(–270) µm diam. *Asci* free, clavate to elongateobovoid, polysporous, (35.5–)38.5–47.5(–50) × (7–)8–9.5(–10) µm. *Ascospores* elongate-allantoid, thin-walled, hyaline, aseptate,



**Fig. 55.** *Cytospora tongzhouensis* (BJFC-S2213). **A**, **B**. Habit of pseudostromata on twig. **C**. Transverse section through pseudostroma with ascomata. **D**. Longitudinal section through pseudostroma with ascomata. **E**. Asci. **F**. Ascospores. Scale bars: A = 1 mm; B–D = 500 μm; E–F = 10 μm.

(7–)7.5–10(–10.5) × (1–)1.3–2.2(–2.5) (av. = 8.7  $\pm$  0.9 × 1.8  $\pm$  0.3, n = 30) µm. As exual morph not observed.

*Culture characteristics*: Colonies on PDA smoke grey to grey olivaceous, growing fast and entirely covering the 9-cm-diam Petri dish after 3 d, flat with a uniform texture lacking aerial mycelium. Sterile.

*Notes: Cytospora tongzhouensis* was collected from *Salix* sp. in Beijing, China. Phylogenetically, it clusters as sister species to *C. polyspora*, but differs in sequence loci (see notes of *C. polyspora*). Morphologically, *C. tongzhouensis* is distinct from *C. polyspora* in having smaller asci,  $(38.5-47.5 \times 8-9.5 vs 47.5-53.5 \times 10.5-12 \mu m)$  and ascospores  $(7.5-10 \times 1.3-2.2 vs 13-16 \times 3.1-3.8 \mu m)$ . *Cytospora tongzhouensis* also shares morphological characteristics with the sexual morph of *C. yulinensis*. However, it can be easily distinguished from that species by its larger asci  $(38.5-47.5 \times 8-9.5 vs 33.6-42.1 \times 6-7.1 \mu m)$ , and smaller ascospores  $(7.5-10 \times 1.3-2.2 vs 10.7-14.8 \times 2.4-2.9 \mu m)$  (Fan *et al.* 2014b, as *C. nivea*). In the phylogenetic analysis (Fig. 1), it resides in a clearly defined clade.

*Cytospora translucens* Sacc., Syll. Fung. (Abellini) 3: 261. 1884. Fig. 56. (Fig. 1: Clade 100)

Synonym: Valsa translucens (De Not.) De Not., Comm. Soc. crittog. Ital. 1(fasc. 4): 208. 1863.

## Typus: Requires typification.

Description: Conidiomata Group AII (type a7), immersed in bark, erumpent when mature, discoid, flask-shaped to conical, with large multi-locule. Conceptacle prominent, and uniquely visible as a dark circle surrounding the disc on the plant epidermis or bark exterior in young branches of *Populus* and *Salix. Ectostromatic disc* conspicuous, umber to straw, circular to ovoid, 230–370 µm in diam. Ostiole conspicuous, circular to ovoid, orange to black at the same level as the disc surface, 30–45 µm in diam. Locules subdivided frequently by invaginations with common walls. Conidiospores hyaline, unbranched or occasionally branched at the bases, 11–17 × 1–2.5 µm (av. = 15.3 ± 1.6 × 1.7 ± 0.2 µm, n

= 30). Conidiogenous cells enteroblastic, phialidic, subcylindrical to cylindrical,  $3.5-6 \times 1-2 \mu m$  (av. =  $5.0 \pm 0.8 \times 1.6 \pm 0.1 \mu m$ , n = 30). Conidia hyaline, unicellular, eguttulate, elongate-allantoid,  $4.5-6 \times 1-2 \mu m$  (av. =  $5.0 \pm 0.2 \times 1.7 \pm 0.1 \mu m$ , n = 50).

*Materials examined*: **China**, Beijing City, Yanqing District, Zhangshanying Town, 40°28'33"N, 115°49'58"E, on twigs and branches of *Populus* sp., 5 Sep. 2022, *L. Lin* (BJFC-S2095, culture CFCC 58498); Yunnan Province, Diqing Tibetan Autonomous Prefecture, Shangri-La City, Sanba Naxi Town, East Ring Road, 27°34'26"N, 100°1'5"E, on branches of *Populus adenopoda*, 9 Aug. 2022, *L. Lin & M. Lin* (BJFC-S2091, culture CFCC 58255; BJFC-S2092, culture CFCC 58256; BJFC-S2093, culture CFCC 58257; BJFC-S2094, culture CFCC 58258).

Notes: A wide host range has been reported for Cytospora translucens, including Rosaceae, Salicaceae and Vitaceae (Esfandiari 1946, Scharif & Ershad 1966, Ashkan & Hedjaroude 1981, Fotouhifar et al. 2010). The ITS gene phylogenies do not resolve C. translucens from a larger clade contains C. alba, C. hoffmannii, C. mali-spectabilis and C. yulinensis. In the act1, *rpb2*, *tef1-\alpha* and *tub2* gene trees, the BI and ML phylogenies all place C. translucens in a distinct clade, with high support (ML/BI = 91/0.999, 88/0.97, 100/1 and 100/1, respectively). Until recently, this species has been mainly reported from stems of Populus spp. in Inner Mongolia and Heilongjiang Provinces, China (Wang et al. 2015) and in eastern and western North America (Adams et al. 2005; Keply et al. 2015). In the present study, it was also found on Populus in Beijing City and Yunnan Province, China. As Cytospora translucens Sacc. is a younger synonym of Valsa translucens (De Not.) De Not., use of the epithet translucens in Cytospora requires conservation. However, detailed studies including verified strains from Europe need to be included before typification.

*Cytospora tristicha* (De Not.) L. Lin, X.L. Fan & Crous, *comb. nov.* MycoBank MB 850184. (Fig. 1: Clade 143)

Basionym: Diatrype tristicha De Not., Comm. Soc. Crittog. Ital. 2(fasc. 3): 481. 1867.

Synonym: Valseutypella tristicha (De Not.) Höhn., Ann. Mycol. 18(1/3): 72. 1920.



Fig. 56. Cytospora translucens (BJFC-S2093). A, B. Habit of conidiomata on twig. C. Transverse section through conidioma. D. Longitudinal section through conidioma. E. Conidiophores and conidiogenous cells. F. Conidia. Scale bars: A = 2 mm; B–D = 200 µm; E–G = 10 µm.

Typus: Requires typification.

Description: See Hubbes (1960).

Material examined: Switzerland, Kt. Graubünden, Castiel, Schanfigg, on Rosa sp., 15 Jul. 1959, E. Müller (culture CBS 465.59).

*Notes:* Valseutypella tristicha is the type species of Valseutypella. This species was described from *Rosa sylvatica* in Italy, but no extype isolate is available for comparison. Hubbes (1960) considered the morphology of the specimen from which CBS 465.59 was isolated and confirmed its identity. In addition, he used this strain for detailed axenic culture studies. Unfortunately, no specimen of this collection is extant at Z and ZT, where the collection of E. Müller has been deposited (R. Berndt, pers. comm.). The isolate CBS 465.59 is thus treated as reference strain in the present study, as it has a same host genus and continent as the original records. *Cytospora tristicha* (CBS 465.59) differs from *C. rosicola*. However, *C. tristicha* (CBS 465.59) differs from *C. rosicola* (CF 20197024) in ITS (1/510), *act1* (11/234), *rpb2* (21/650), *tef1-a* (35/510) and *tub2* (25/338) loci.

*Cytospora tritici* Punith., Nova Hedwigia 32: 586. 1980. (Fig. 1: Clade 54)

Synonym: Cytospora ershadii Zafari & Hanifeh, Fungal Biol. 126(11–12): 715. 2022.

Description: See Hanifeh et al. (2022) (as C. ershadii).

Typus: Australia, on wheat grains. Requires typification.

*Materials examined*: **Germany**, Monheim, on leaf spot of *Triticum aestivum*, 1984, *Gasser* (CBS H-11545, culture CBS 827.84). **Iran**, East Azerbaijan Province, Afil, on *Prunus* sp., 2010, *M. Arzanlou* (culture CBS 141625 = CPC 19905). **South Africa**, KwaZulu-Natal Province, Newcastle, on *Populus nigra* cv. *italica*, unknown date and *collector* (culture MSC 384988 = CBS 118563); Free State Province, Bloemfontein, on *Populus simonii*, unknown date and *collector* (culture CBS 118561 = CMW 5273).

*Notes*: Adams *et al.* (2006) treated CBS 827.84 as *C. tritici.* The isolates CBS 118563 and CBS 118561 were deposited as *C. chrysosperma* and *C. nivea* in the CBS collection, respectively, but they group in clade 54 along with an isolate of *C. tritici. Cytospora ershadii* (IRAN 4197C and IRAN 4198C) was described from apple trees in Iran (Hanifeh *et al.* 2022). It clusters together with *C. tritici* and has the following nucleotide similarities with the sequences of CBS 827.84: In ITS, *act1, rpb2* and *tef1-a*: 514/514 (100 %), 241/243 (99.2 %), 650/650 (100 %) and 276/279 (98.9 %), respectively. Therefore, *C. ershadii* is treated as a synonym of *C. tritici*.

*Cytospora ulmi* Norph. *et al.*, Mycosphere 8: 85. 2017. (Fig. 1: Clade 17)

*Typus*: **Russia**, Rostov Region, Krasnosulinsky District, Donskoye forestry, ravine forest, on branches of *Ulmus minor*, 28 Jun. 2015, *T. Bulgakov*, T-521 (**holotype** MFLU 15-1910, ex-holotype culture MFLUCC 15-0863).

Description: See Norphanphoun et al. (2017).

Material examined: Australia, on twig of Ulmus procera, 2000, M.J. Wingfield (CBS H-22975 dried culture, culture CBS 116856 = CMW 6758).

Notes: Cytospora ulmi was described on dead and dying branches of Ulmus minor by Norphanphoun et al. (2017). Adams et al. (2005)

identified CBS 116856 as C. subclypeata based on rDNA-ITS phylogenies, which revealed a close relationship to the isolate of C. subclypeata (CBS 117.67) using rDNA-ITS sequences. However, in the present study CBS 116856 groups with the ex-holotype culture of C. ulmi (MFLUCC 15-0863), based on ITS, act1, rpb2, tef1-α and tub2 sequences. Isolate CBS 116856 differs from C. subclypeata (CBS 117.67) in ITS (8/506), act1 (13/212), rpb2 (37/650), tef1-a (33/487) and tub2 (27/411) loci, while it does not differ in ITS (100.0 % identity, act1 (100 % identity) and only insignificantly in rpb2 (99.4%, with 4 bp differences) from C. ulmi (MFLUCC 15-0863) (no *tef1-\alpha* and *tub2* available for MFLUCC 15-0863). Additionally, the host genus of CBS 116856 is the same as that for the holotype of C. ulmi. The sequence dataset of Norphanphoun et al. (2017) included CBS 117.67, but not CBS 116856. Isolate MFLUCC 15-0863 does not cluster together with CBS 117.67 in their phylogram, but forms an independent branch, and they subsequently named MFLUCC 15-0863 as C. ulmi. Cytospora ulmi is the best isolate to define clade 17, as CBS 116856 is not an ex-type strain of C. subclypeata.

*Cytospora uniloculata* L. Lin & X.L. Fan, *sp. nov.* MycoBank MB 850186. Fig. 57. (Fig. 1: Clade 149)

*Etymology*: Name refers to the unilocular conidiomata in this species.

*Typus*: **China**, Beijing City, Mentougou District, Xiaolongmen Forestry Centre, 39°50'32"N, 115°33'55"E, on twigs and branches of *Malus baccata*, 25 Aug. 2022, *L. Lin & X.L. Fan* (**holotype** BJFC-S2035, exholotype culture CFCC 58460).

Description: Conidiomata Group AI (type a1), immersed in bark, erumpent when mature, discoid to conical, 345–450 µm diam, with an undivided locule. *Conceptacle* absent. *Locule* single, without wing-like ectostroma around the ostiole. *Ectostromatic disc* honey to umber, circular to ovoid, 140–200 µm diam, with single ostiole per disc in the centre. *Ostiole* circular to ovoid, black, 55–75 µm diam. *Conidiophores* hyaline, unbranched or branched, 17.5–36.5 × 1.5–3.5 µm (av. =  $25.1 \pm 5.0 \times 2.5 \pm 0.5$  µm, n = 30). *Conidiogenous cells* enteroblastic, phialidic, subcylindrical to cylindrical, 3.5–7.5 × 1.5–2.5 µm (av. =  $5.4 \pm 0.9 \times 1.8 \pm 0.2$  µm, n = 30). *Conidia* hyaline, unicellular, eguttulate, elongate-allantoid, 8–9.5 × 2–3 µm (av. =  $8.9 \pm 0.35 \times 2.6 \pm 0.3$  µm, n = 50).

*Culture characteristics*: Cultures on PDA white, growing up to 6 cm diam after 3 d, flat with a uniform texture, without aerial mycelium.

*Notes*: Phylogenetically, *C. uniloculata* is closely related to *C. kuanchengensis*. However, *C. uniloculata* can be distinguished from *C. kuanchengensis* by its unilocular conidiomata lacking a conceptacle, and larger conidia [8–9.5 × 2–3 µm in *C. uniloculata* compared with (5.5–)6–7.5(–8) × 1–2 µm in *C. kuanchengensis*] (Jiang *et al.* 2020b). Additionally, *C. uniloculata* was collected from *Malus baccata*, while *C. kuanchengensis* was collected from *Castanea mollissima* (Jiang *et al.* 2020b).

*Cytospora valsoidea* G.C. Adams & M.J. Wingf., Stud. Mycol. 52: 101. 2005. (Fig. 1: Clade 210)

*Typus*: Indonesia, North Sumatra, Sibisa, on dead cankered branch of *Eucalyptus grandis*, 1992, *M.J. Wingfield* (holotype MSC 380717, exholotype culture CBS 117003 = CMW 4309).





Fig. 57. Cytospora uniloculata (BJFC-S2035). A, B. Habit of conidiomata on twig. C. Transverse section through conidioma. D. Longitudinal section through conidioma. E, F. Conidiophores and conidiogenous cells. G. Conidia. Scale bars: A = 1 mm; B–D = 200 μm; E–G = 10 μm.

Description: See Adams et al. (2005).

*Additional material examined:* **Indonesia**, North Sumatra, Sibisa, on dead cankered branch of *Eucalyptus grandis*, 1992, *M.J. Wingfield* (MSC 380707, culture CBS 116857 = CMW 4310).

*Notes: Cytospora valsoidea* was described from *Eucalyptus grandis* as having a valsoid arrangement of the conidiomata with ostiolar necks that converge laterally into a disc (Adams *et al.* 2005). This species may be removed from the genus *Cytospora* after further collections due to its distant position in phylogram (Fig. 1) and unique, straight conidia.

*Cytospora variostromatica* G.C. Adams & M.J. Wingf., Stud. Mycol. 52: 108. 2005. (Fig. 1: Clade 208)

*Typus*: **Australia**, Victoria, Orbost, Toslaree, on dead branches of *Eucalyptus globulus*, 2000, *M.J. Wingfield* (**holotype** MSC38069, exholotype culture CBS 116858 = CMW 6766).

Description: See Adams et al. (2005).

Additional materials examined: **Australia**, on dead branches of *Eucalyptus* grandis × *E. camaldulensis*, unknown date, *M.J. Wingfield* (CBS H-22977 dried culture, culture CBS 116859 = CMW 6746). **South Africa**, KwaZulu-Natal Province, King Cetshwayo District, KwaMbonambi, on twig of *Eucalyptus grandis*, unknown date, *M.J. Wingfield* (CBS H-22919 dried culture, culture CBS 116860 = CMW 1240); Free State Province, Allemanskraal Dam, on twig of *Populus deltoides*, unknown date, *G.C. Adams* (CBS H-22980 dried culture, culture CBS 118564 = CMW 5270); Mpumalanga Province, Amsterdam, on *Eucalyptus nitens*, unknown date and *collector* (culture CBS 118087 = CMW 1514).

*Notes: Cytospora variostromatica* was described from *Eucalyptus globulus* having variable conidiomatal stromata, which included unilocular (Group AI, type a1), cytosporoid (Group AII, type a6) and lamyelloid type with ostioles converging to a shared disc (Group AIII, type a11) (Adams *et al.* 2005). Isolate CBS 118564 represents the only isolate from *Populus deltoides*. Phylogenetic analysis (Fig. 1) shows that this species is distinct from all others in the genus within this species clade. In the ITS, *act1, rpb2, tef1-a* and *tub2* gene trees, the BI and ML phylogenies all place it in a

distinct clade, with high support (ML/BI = 100/1, 100/1, 100/1, 74/1 and 99/1, respectively). Further morphological examination of fungarium specimens will be needed to clarify the relation between the phylogenetic differentiation and variation in conidiomatal types in this species.

*Cytospora viridistroma* (Wehm.) C.M. Tian *et al.*, Mycologia 112: 281. 2020. (Fig. 1: Clade 173)

*Basionym: Endothia viridistroma* Wehm., Mycologia 28: 35. 1936. *Synonyms: Valsa brevispora* G.C. Adams & Jol. Roux, Stud. Mycol. 52: 91. 2005.

Valsa viridistroma (Wehm.) G.C. Adams et al., Australas. Pl. Pathol. 35: 526. 2006.

*Cytospora brevispora* G.C. Adams & Rossman, IMA Fungus 6: 147. 2015.

*Typus*: **USA**, Georgia, University of Georgia, on *Cercis canadensis*, 30 Apr. 1934, *J.H. Miller* (ex-holotype culture CBS 202.36).

Description: See Wehmeyer (1936), Adams et al. (2005) (as Valsa brevispora).

Additional materials examined: **Colombia**, Cali, on dead branches of *Eucalyptus grandis*, 2000, *M.J. Wingfield* (MSC 375217, culture CBS 116853); on *Eucalyptus grandis*, unknown date, *M.J. Wingfield* (CBS H-22920 dried culture, culture CBS 117011). **Mexico**, on twig of *Eucalyptus grandis*, 1996, *M.J. Wingfield* (CBS H-22921, culture CBS 117080 = CMW 516). **Republic of Congo**, Tchittanga, *Eucalyptus grandis* × *E. tereticornis*, 1998, *J. Roux* (cultures CBS 116811 = CMW 5260 and CBS 116812 = CMW 5261). **Venezuela**, Acarigua, on bark of *E. camaldulensis*, 1997, *M.J. Wingfield* (cultures CBS 116813 and CBS 116829).

*Notes*: Wehmeyer (1936) described *Cytospora viridistroma* with slightly allantoid ascospores (5–6 × 1–1.5 µm) and cylindrical to allantoid conidia (2.5–3.5 × 0.8–1 µm). Jiang *et al.* (2020a) sequenced the ex-holotype strain of *Endothia viridistroma* (CBS 202.36) and showed that it was a *Cytospora* sp. *Cytospora brevispora* was introduced as *Valsa brevispora* from *Eucalyptus* spp. with isolate CBS 116811 as holotype (Adams *et al.* 2005). Rossman *et al.* (2015) combined *V. brevispora* in *Cytospora*. In

the present study, the isolates CBS 116811 and 202.36 cluster in a strongly supported clade (ML/BI = 99/1) (Fig. 1). *Cytospora viridistroma* has to be applied to this clade based on priority of the basionym. Adams *et al.* (2005) treated the sister clade (CBS 116853, 117011 and 117080) as *C. eucalyptina* after examining the holotype of *C. eucalyptina* (Argentina, from *Eucalyptus globulus*, 1888, C. Spegazzini, LPS 11656). However, the culture and DNA sequence data based on the type material are necessary due to the confused history and morphological limitation of *C. eucalyptina* (Adams *et al.* 2005). The present study adds additional strains (CBS 116853, 117011 and 117080) and treats the clade as *Cytospora viridistroma* based on both multigene and single gene phylogenies (Fig. 1). The choice of name should be seen as tentative as *C. eucalyptina* requires typification before a stable species concept is found.

*Cytospora viticola* D.P. Lawr. *et al.*, Pl. Pathol. 66: 718. 2016. (Fig. 1: Clade 88)

*Typus*: **USA**, Connecticut, Litchfield County, from wood canker of *Vitis vinifera* 'Cabernet Franc', 20 Mar. 2008, *P. Rolshausen* (holotype BPI 910161, ex-holotype culture CBS 141586).

Description: Lawrence et al. (2017).

Materials examined: Iran, East Azerbaijan Province, Malakan, on Vitis vinifera, 2010, S. Moshari (cultures CBS 141605 = CPC 19916, CBS 141606 = CPC 19926, CBS 141627 = CPC 19922, CBS 141628 = CPC 19923, CBS 141631 = CPC 19935). USA, Pennsylvania, on Vitis sp., 2002, unknown collector (culture CBS 141614 = CPC 28388); on twig of Vitis sp., 2003, E. Stewart (culture CBS 141623 = CPC 28413).

*Notes: Cytospora viticola* was described from *Vitis vinifera* in the USA (Lawrence *et al.* 2017). In the ITS, *act1*, *rpb2*, *tef1-a* and *tub2* gene trees, the BI and ML phylogenies all placed it in a distinct clade, with high support (ML/BI = 100/1, 98/1, 100/1, 100/1 and 100/1, respectively). It has conidiomata in Group AII (type a9) and is characterised by colonies with filiform margins with aerial mycelial tufts throughout (Lawrence *et al.* 2017). This species has been isolated from *Vitis vinifera* and *Castanea* sp. in Canada, USA and Iran (Lawrence *et al.* 2017).

*Cytospora washingtonensis* Jami, Crous & M.J. Wingf., *sp. nov.* MycoBank MB 850151. Fig. 58. (Fig. 1: Clade 12)

*Etymology*: Name refers to federal state where it was collected, Washington (USA).

*Typus*: **USA**, Washington, Kitsap, Poulsbo, Kitsap Memorial State Park, on *Crataegus* sp., unknown date, *D. Walker* (**holotype** CBS H-22935 dried culture, ex-holotype culture CBS 141619 = CPC 28403 = DMW 163.1).

Description: Conidiomata produced on MEA in 2–4 wk, solitary, globose, black, up to 1 mm wide, immersed, unilocular, exuding a creamy conidial mass. Conidiophores hyaline, smooth, 0–2-septate, acropleurogenous. Conidiogenous cells hyaline, smooth, embedded in a continuous gelatinous matrix, 4–9 × 1–1.5 µm, phialidic, subcylindrical, tapering towards the apex, minute collarette. Paraphyses intermingled among conidiophores, hyaline, unbranched, septate, up to 10 µm long. Conidia hyaline, eguttulate, allantoid, apex subobtuse, base truncate, aseptate, 4–7 × 1–2 µm.

*Culture characteristics*: Colonies covering the Petri dish after 10 d at 24 °C, with aerial mycelium flat, moderately sparse. On MEA surface buff colour on both surface and reverse with white and yellow tufts and patches on the surface. On OA surface olivaceous.

Additional materials examined: **USA**, Washington, Kitsap, Poulsbo, Kitsap Memorial State Park, *Crataegus* sp., unknown date, *D. Walker* (cultures CBS 141620 = CPC 28404, CBS 141616 = CPC 28398).

Notes: Cytospora washingtonensis and C. crataegina occur on the same host genus Crataegus. However, they form different lineages in the phylogenetic tree (Fig. 1). Additionally, C. washingtonensis differs from C. crataegina by its larger conidia ( $4-7 \times 1-2 \times 3.5-4.5 \times 1-1.5 \mu$ m). Cytospora washingtonensis clusters in a phylogenetic clade sister to C. betulae (Fig. 1). However, C. washingtonensis (CBS 141619) differs from C. betulae (CBS 141622) in ITS (2/506 bp), act1 (7/246 bp), rpb2 (6/657 bp), tef1-a (16/495 bp) and tub2 (3/340 bp) loci. Morphologically, they can be identified by their different conidial sizes (see notes of C. betulae).

*Cytospora weiriana* (Petr.) X.L. Fan & Crous, *comb. nov.* MycoBank MB 850187. (Fig. 1: Clade 189) *Basionym: Valsa weiriana* Petr., Ann. Mycol. 22(3/6): 387. 1924.

Typus: USA, on Thuja plicata. Requires typification.

*Material examined*: **Canada**, British Columbia, on *Pseudotsuga menziesii*, unknown date, *G.C. Adams* (MSC 384998, culture CBS 118567).

Notes: Adams et al. (2005) noted that isolates identified as V. abietis from the Pacific north-western USA are not related to isolates of



**Fig. 58**. *Cytospora washingtonensis* (CBS 141619). **A.** Conidiomata on MEA. **B–E.** Conidiophores and conidiogenous cells. **F.** Conidia. Scale bars: A = 100 μm; B–E = 10 μm; F = 5 μm.



*V. abietis* from eastern North America based on ITS-rDNA data. Therefore, they tentatively referred to the isolates (including culture CBS 118567) on *Pseudotsuga* and *Chamaecyparis* from the Pacific Northwest as *Valsa weiriana*. Isolate CBS 118567 clusters as a sister clade of *C. junipericola*. However, it differs from *C. junipericola* (CBS 196.97) in its ITS (8/510), *act1* (8/198), *rpb2* (13/650), *tef1-a* (55/521) and *tub2* (33/441) loci. This taxon is retained as *Cytospora weiriana* in the current treatment until further collections can facilitate a definitive typification.

*Cytospora xiaolongmenensis* L. Lin & X.L. Fan, *sp. nov.* MycoBank MB 850152. Fig. 59. (Fig. 1: Clade 151)

*Etymology*: Name refers to Xiaolongmen Forestry Centre (China) where it was collected.

*Typus*: **China**, Beijing City, Mentougou District, Xiaolongmen Forestry Centre, 39°50'11"N, 115°34'27"E, on twigs and branches of *Malus baccata*, 25 Aug. 2022, *L. Lin & X.L. Fan* (**holotype** BJFC-S2037, exholotype culture CFCC 58459).

Description: Pseudostroma Group SII (type s7), immersed in the bark, scattered, with 15–23 perithecia arranged irregularly. *Conceptacle* absent. *Ectostromatic disc* pale luteous to luteous, usually surrounded by ostiolar necks, triangular to circular, 350– 600 µm diam, with 3–8 irregularly arranged ostioles in the disc. *Ostioles* black when mature, 65–95 µm diam. *Perithecia* buff to black when mature, flask-shaped to spherical, arranged irregularly, 130–240 µm diam. *Asci* hyaline, with chitinoid, refractive ring, clavate to elongate-obovoid, 38–51 × 8.5–13.5 µm (av. = 44.5  $\pm$  3.7 × 10.2  $\pm$  1.2 µm, n = 30), 8-spored. *Ascospores* hyaline, biseriate to multiseriate, elongate-allantoid, aseptate, 9.5–16.5 × 2–3.5 µm (av. = 14.0  $\pm$  1.4 × 2.6  $\pm$  0.3 µm, n = 50).

*Culture characteristics*: Cultures on PDA are initially white, growing up to 3 cm diam after 3 d, entirely covering the 9-cm-diam Petri dish after 7 d and becoming milky after 20 d.

Notes: Cytospora xiaolongmenensis collected from Malus baccata forms a distinct lineage in the Cytospora phylogeny (Fig.1) Morphologically, it can be distinguished from other species by its numerous perithecia in a pseudostroma. Cytospora mali and C. uniloculata were also collected from Malus baccata (Tai 1979, Pan *et al.* 2018, this study). *Cytospora xiaolongmenensis* is clearly distinct from these two species in the phylogram (Fig. 1).

*Cytospora xylocarpi* Norph. *et al.*, MycoKeys 38: 108. 2018. Fig. 60. (Fig. 1: Clade 174)

*Typus*: **Thailand**, Ranong Province, Ngao Mangrove Forest, on branches of *Xylocarpus granatum*, 6 Dec. 2016, *C. Norphanphoun*, NG09b (**holotype** MFLU 17-0708, ex-holotype culture MFLUCC 17-0251).

Description: Conidiomata produced on MEA in 5–7 wk, solitary, stromata, black, up to 200  $\mu$ m diam, semi-immersed, unilocular, yellow exuding conidia. Conidiophores hyaline, smooth, branched, 0–3-septate, 3–11 × 1.5–2.5  $\mu$ m, embedded in a gelatinous layer. Conidiogenous cells embedded in a continuous gelatinous matrix, phialidic, subcylindrical, tapering towards apices, collarettes minute, 3–9 × 1.5–2.5  $\mu$ m arranged in rosettes of up to three. Conidia hyaline, eguttulate, cylindrical to ellipsoidal, variable in shape, aseptate, apex subobtusely rounded, tapering to a subtruncate base, 2.5–4 × 0.8–1.5  $\mu$ m.

*Culture characteristics*: Colonies covering the Petri dish after 10 d at 24 °C, with aerial mycelium flat, with zonal growth. On MEA surface greyish sepia and black on the reverse.

*Material examined*: **Thailand**, on dead branches of *Eucalyptus grandis*, 1996, *M.J. Wingfield* (culture CBS 116861 = CMW 464).

Notes: Cytospora xylocarpi was described from Xylocarpus granatum in Thailand (Norphanphoun *et al.* 2018). Adams *et al.* (2005) identified isolate CBS 116861 as a putative Cytospora species. The present study treats it as Cytospora xylocarpi, based on the similar greyish sepia culture on MEA and overlapping conidial dimensions (2.5–4 × 0.8–1.5 vs 3–3.1 × 0.8–1 µm). Isolate MUCC 302 (Australia, *Eucalyptus grandis*) was deposited in GenBank by Burgess & Wingfield (2001) as C. rhizophorae. Phylogenetically, CBS 116861 and MUCC 302 group together with the ex-holotype culture of C. xylocarpi (MFLUCC 17-0251) (in ITS, act1, and rpb2 gene trees, with high support of ML/BI = 95/0.98, 99/1 and 94/0.99, respectively, no *tef1-α* and *tub2* sequences available for MFLUCC 17-0251). Isolate MUCC 302 is consequently treated as C. xylocarpi, until the paratype of C. rhizophorae (Guatemala, from *Rhizophora mangle*, 1970, J. J. Kohlmeyer NY Barcode 01389553) is re-examined.



**Fig. 59.** *Cytospora xiaolongmenensis* (BJFC-S2037). **A**, **B**. Habit of pseudostromata on twig. **C**. Transverse section through pseudostroma with ascomata. **D**. Longitudinal section through pseudostroma with ascomata. **E**, **F**. Asci. **G**. Ascospores. Scale bars: A = 1 mm; B–D = 200 μm; E–G = 10 μm.



Fig. 60. Cytospora xylocarpi (CBS 116861). A. Conidiomata on MEA. B, C. Conidiophores and conidiogenous cells. D. Conidia. Scale bars: A = 100 μm; B–D = 10 μm.

*Cytospora yinchuanensis* L. Lin & X.L. Fan, *sp. nov.* MycoBank MB 850190. Fig. 61. (Fig. 1: Clade 52)

*Etymology*: Name refers to Yinchuan City (China) where it was collected

*Typus*: **China**, Ningxia Hui Autonomous Region, Yinchuan City, 106°01'00" E, 38°27'40"N, on branches of *Malus pumila*, 2 Jun. 2012, *X.L. Fan* (**holotype** BJFC-S538, ex-holotype culture CFCC 50040); Gansu Province, Gannan City, Diebu County, Zhouqu Town, 104°20'12.65" E, 33°46'56.55" N, on branches of *Malus pumila*, 10 Aug. 2012, *X.L. Fan* (**paratype** BJFC-S773, ex-paratype culture CFCC 50042).

Description: Conidiomata Group AII (type a6), immersed in bark, erumpent when mature, flat, discoid, flask-shaped to conical, with large multi-locule. Conceptacle absent. Ectostromatic disc light brown, circular to ovoid, 290–415  $\mu$ m in diam. Ostiole 1–7, circular to ovoid, isabelline to black, 35–45  $\mu$ m in diam. Locules subdivided frequently by invaginations with common walls. Conidiospores hyaline, unbranched or branched at the bases, 13–20 × 1.5–2  $\mu$ m (av. = 17.3 ± 1.6 × 1.7 ± 0.1  $\mu$ m, n = 30). Conidiogenous cells

enteroblastic, phialidic, subcylindrical to cylindrical,  $6.5-11 \times 1.5-2.5 \mu m$  (av. =  $8.0 \pm 1.8 \times 2.0 \pm 0.2 \mu m$ , n = 30). *Conidia* hyaline, unicellular, eguttulate, elongate-allantoid,  $5-6 \times 1-1.5 \mu m$  (av. =  $5.7 \pm 0.2 \times 1.3 \pm 0.1 \mu m$ , n = 50).

*Notes*: Specimens BJFC-S538 and BJFC-S773 were collected from *Malus pumila* and previously identified as *C. schulzeri* (Fan *et al.* 2020). However, they are phylogenetically distinct from *C. schulzeri* in the present study (Fig. 1). *Cytospora yinchuanensis* (CFCC 50040) differs from *C. schulzeri* (CBS 118570) in ITS (3/512 bp), *act1* (20/250 bp), *rpb2* (30/650 bp), *tef1-a* (51/488 bp) and *tub2* (35/419 bp) loci. Morphologically, *C. yinchuanensis* differs from *C. schulzeri* based on its smaller conidia (4.5–6.5 × 1–1.5 µm vs 6.5–8 × 1.3–1.5 µm) (Fan *et al.* 2020).

*Cytospora yuduensis* L. Lin & X.L. Fan, *sp. nov.* MycoBank MB 850191. Fig. 62. (Fig. 1: Clade 157)

*Etymology*: Name refers to Yudu Mountain (China) where it was collected.



**Fig. 61.** *Cytospora yinchuanensis* (BJFC-S538). **A**, **B**. Habit of conidiomata on twig. **C**. Transverse section through conidioma. **D**. Longitudinal section through conidioma. **E**. Conidiophores and conidiogenous cells. **F**. Conidia. Scale bars: A = 2 mm; B–D = 200 µm; E, F = 10 µm.





Fig. 62. Cytospora yuduensis (BJFC-S2097). A, B. Habit of conidiomata on twig. C. Transverse section through conidioma. D. Longitudinal section through conidioma. E. Conidiophores and conidiogenous cells. F. Conidia. Scale bars: A = 500 μm; B–D = 100 μm; E, F = 10 μm.

*Typus*: **China**, Beijing City, Yanqing District, Yudu Mountain, 40°32'39"N, 115°54'15"E, on twigs of *Populus × beijingensis*, 23 Aug. 2021, *L. Lin & Z. Du* (**holotype** BJFC-S2097, ex-holotype CFCC 57539; **isotype** BJFC-S2098, ex-isotype culture CFCC 57540); Mentougou District, Xiaolongmen Forestry Centre, 39°57'51"N, 115°25'42"E, on twigs and branches of *Populus × beijingensis*, 25 Aug. 2022, *L. Lin & X.L. Fan* (**paratype** BJFC-S2099, ex-paratype culture CFCC 58269).

Description: Conidiomata Group AI (type a2), immersed in bark, erumpent when mature, discoid to conical, 240–360 µm diam, with an undivided locule. Locules single, delimited by black conceptacle. Ectostromatic disc honey to isabelline, circular to ovoid, 150–210 µm diam, with single ostiole per disc in the centre. Ostiole circular to ovoid, isabelline to black, 50–70 µm diam. Conidiophores hyaline, unbranched or occasionally branched, 12.5–26.5 × 1.5–2.5 µm (av. = 19.0 ± 3.1 × 1.8 ± 0.2 µm, n = 30). Conidiogenous cells enteroblastic, phialidic, subcylindrical to cylindrical, 3.5–6 × 1–1.5 µm (av. = 4.8 ± 0.7 × 1.3 ± 0.1 µm, n = 30). Conidia hyaline, unicellular, eguttulate, elongate-allantoid, 5.5–8 × 1.5–2 µm (av. = 7.0 ± 0.5 × 1.8 ± 0.17 µm, n = 50).

*Culture characteristics*: Cultures on PDA initially white, growing up to 5 cm diam after 3 d, entirely covering the 9-cm-diam Petri dish after 5 d and becoming olivaceous after 10 d greenish on the surface and dark mouse grey in reverse after 20 d, flat with a uniform texture. Conidioma produced after 15 d.

*Notes: Cytospora yuduensis* was collected from *Populus* twigs with canker disease. Its pathogenicity to poplar remains to be confirmed. It is distinguished from other species of *Cytospora* by undivided single locules surrounded by a black conceptacle. *Cytospora yuduensis* clusters as sister species to *C. tenebrica*, but differs in sequence loci (see notes of *C. tenebrica*).

*Cytospora yulinensis* L. Lin & X.L. Fan, *sp. nov.* MycoBank MB 851590. Fig. 63. (Fig. 1: Clade 102)

*Etymology*: Name refers to Yulin City (China) where the holotype specimen was collected.

*Typus*: **China**, Shaanxi Province, Yulin City, Hongshi Gorge, 38°19'32.43"N, 109°42'00.69"E, on twigs and branches of *Salix psammophila*, 29 Jul. 2013, *X.L. Fan* (**holotype** BJFC-S979, ex-holotype culture CFCC 89643); Ningxia Ningxia Hui Autonomous Region, Guyuan City, Changchengliang, 36°03'01.78"N, 106°16'18.09"E, on twigs and branches of *Elaeagnus angustifolia*, 24 Jul. 2013, *X.L. Fan* (**paratype** BJFC-S964, ex-paratype culture CFCC 89641).

Description: Conidiomata Group AII (type a7), semi-immersed in bark, erumpent when mature, discoid to conical, 550–850 µm diam, with multi-locule. Conceptacle prominent. Ectostromatic disc buff to brown, circular, 150–230 µm diam, with single ostiole per disc in the centre. Ostiole circular to ovoid, black, 50–75 µm diam. Locules multiple, subdivided with common walls. Conidiophores hyaline, unbranched or occasionally branched, 15–23.5 × 1–2 µm (av. = 17.8 ± 1.7 × 1.5 ± 0.1 µm, n = 30). Conidiogenous cells enteroblastic, phialidic, subcylindrical to cylindrical, 4.5–8.5 × 1–2 µm (av. = 6.2 ± 1.1 × 1.5 ± 0.1 µm, n = 30). Conidia hyaline, unicellular, eguttulate, elongate-allantoid, 4.5–7.5 × 1.5–2 µm (av. = 6.2 ± 0.2 × 1.7 ± 0.1 µm, n = 50).

Notes: Cytospora yulinensis was identified as *C. nivea* (Hoffm.) Sacc. ( $\equiv$ Sphaeria nivea Hoffm.) based on its host and morphology (Fan *et al.* 2015b). However, it differs phylogenetically from the clade 98 (*Cytospora hoffmannii*) but clusters in a sister clade with *C. alba* in the current study. It can be distinguished from *C. alba* by its disc colour (Lin *et al.* 2022). Phylogenetically, *Cytospora yulinensis* (CFCC 89643) differs from *C. alba* (CFCC 55462) in ITS (4/508), *act1* (6/241), *rpb2* (12/726), *tef1-α* (43/540) and *tub2* (15/400) loci.

*Cytospora zhaitangensis* L. Lin & X.L. Fan, J. Fungi 9: 20. 2023. Fig. 64. (Fig. 1: Clade 30)

*Typus*: China, Beijing City, Mentougou District, next to the Zhaitang Reservoir, on branches of *Euonymus japonicus*, 21 Aug. 2021, *H. Gao & X.L. Fan* (holotype BJFC CF20220144, ex-holotype culture CFCC 56227).

Description: Pseudostromata Group SII (type s3), immersed in the bark, scattered, with 7-15 perithecia irregularly. Conceptacle


Fig. 63. Cytospora yulinensis (BJFC-S979). A. Habit of conidiomata on twig. B. Transverse section through conidioma. C. Longitudinal section through conidioma. D. Conidiophores and conidiogenous cells. E. Conidia. Scale bars: A–C = 200 μm; D, E = 10 μm.

absent. *Ectostromatic disc* fawn to umber, usually surrounded by ostiolar necks, circular, 160–390  $\mu$ m diam, with 7–18 irregularly arranged in the disk. *Ostioles* black when mature, 35–55  $\mu$ m diam. *Perithecia* buff to black when mature, flask-shaped to spherical, circularly arranged, 210–300  $\mu$ m diam. *Asci* hyaline, with chitinoid, refractive ring, clavate to elongate-obovoid, 64.5–72 × 12.5–15  $\mu$ m (av. = 69.5 ± 1.3 × 13.9 ± 0.9  $\mu$ m, n = 30), 8-spored. *Ascospores* hyaline, biseriate, elongate allantoid, aseptate, 13.5–18.5 ×4–6  $\mu$ m (av. = 16.4 ± 1.1 × 4.8 ± 0.5  $\mu$ m, n = 50).

*Culture characteristics*: Cultures on PDA are initially white, growing up to 7 cm diam after 3 d, entirely covering the 9-cm-diam Petri dish after 4 d and becoming pale vinaceous to vinaceous buff after 10 d.

*Material examined*: **China**, Beijing City, Mentougou District, Xiaolongmen Forest Centre, 115°34'35"E, 39°50'4"N, on branches of *Corylus mandshurica*, 23 Aug. 2022, *L. Lin & X.L. Fan* (BJFC-S2001, culture CFCC 58608).

*Notes*: *Cytospora zhaitangensis* was described by Lin *et al.* (2023b) from *Euonymus japonicus* based on its asexual morph. On *act1*, *tef1-α* and *tub2* gene trees, isolate CFCC 58608 (isolated from an ascoma) and isolate CFCC 56227 (the ex-holotype of *C. zhaitangensis*) group together with high support (ML/BI = 99/1, 100/1 and 94/1, respectively). In ITS and *rpb2* gene trees, the BI and ML phylogenies do not resolve CFCC 58608 from the clade including CFCC 56227, with maximum support (ML/BI = 100/1). It



**Fig. 64.** *Cytospora zhaitangensis* (BJFC-S2001). **A–C.** Habit of pseudostromata on twig. **D.** Transverse section through pseudostroma with ascomata. **E.** Longitudinal section through pseudostroma with ascomata. **F.** Ascus. **G.** Ascospores. Scale bars: A = 1 mm; B, C = 200 µm; D, E = 500 µm; F, G = 10 µm.



is suggested to classify isolate CFCC 58608 as *C. zhaitangensis*. The distinction between the asexual and sexual morphs and the different hosts could account for its currently uncertain placement. The host range of *C. zhaitangensis* is expanded to include *Corylus mandshurica* in this study.

## DISCUSSION

In this revision of *Cytospora*, sequence data for five gene regions (ITS, *act1*, *rpb2*, *tef1-a* and *tub2*) were utilised in the phylogenetic analyses. Cultures were collected from Africa, Asia, Europe, North America, Oceania and South America. Three asexual morphological groups (13 asexual morphological types) and three sexual morphological groups (eight sexual morphological types) were designated and descriptive words supplied. Forty-four species were described as new and four new combinations were introduced. In addition, lectotypes and epitypes were provided for three species. Replacement names were proposed for six species. Overall, the results supported treating *Cytospora* as a single genus in the *Cytosporaceae*. Importantly, this study provides the most comprehensive analysis of *Cytospora* species to date based on DNA sequence data linked to a large collection of isolates.

# Cytosporaceae includes a single genus, Cytospora

The family name *Cytosporaceae* Fr. (1825) has priority over *Valsaceae* Tul. & C. Tul. (1861) according to the *International Code* of *Nomenclature for algae, fungi and plants* (McNeill *et al.* 2012). The *Cytosporaceae* accommodates all species of *Cytospora* investigated here, which cluster in a highly supported monophyletic clade.

The number of genera previously accommodated in the Cytosporaceae is unclear because DNA sequence data were available for only a few generic types. Based on published literature, Maharachchikumbura et al. (2015) listed 13 genera in this family, including Amphicytostroma, Chadefaudiomyces, Cryptascoma, Cytospora, Ditopellina, Durispora, Harpostroma, Hypospilina, Kapooria, Leptosillia, Maculatipalma, Pachytrype and Paravalsa. However, Amphiporthe (= Amphicytostroma) belongs in Gnomoniaceae (Sogonov et al. 2008, Rossman et al. 2015). Senanayake et al. (2017) excluded several genera and recognized only Cytospora, Pachytrype, Paravalsa, Xenotypa, and Waydora. Among these, only Cytospora, Pachytrype (specifically, P. princeps and P. rimosa with only LSU data), and Waydora (W. typica with only ITS and LSU data) had available DNA data for phylogenetic analyses. However, Pachytrype princeps (isolate Rogers s.n.) and Cytospora diatrypelloidea (culture CBS 120062) were phylogenetically similar based on LSU (98.4 % identity, with 17 bp difference); Pachytrype rimosa (isolate FF1066) and Cytospora brabeji (culture CBS 119207) were similar on LSU (98.9 % identity, with 10 bp difference); Waydora typica (isolate PDD 103894) and Cytospora variostromatica (culture CBS 116860) had no clear differences based on ITS (99.2 % identity, with 4 bp difference). We consequently reduced Pachytrype and Waydora to synonymy with Cytospora. It was not possible to include all previously mentioned genera in this study, as there were no DNA data or cultures available for them. Based on available DNA sequence data, the Cytosporaceae includes a single genus, Cytospora.

Few epi- or neotypes had previously been provided for genera in the *Cytosporaceae* thus complicating the placement of older names based on phylogenetic inference. Adams *et al.* (2005) reduced Vasella, Valseutypella and Leucostoma to synonymy under Valsa/Cytospora. Fan et al. (2015a, b) and Rossman et al. (2015) proposed the name Cytospora over Valsa based on priority and the fact that this name was more familiar to plant pathologists (Wingfield et al. 2012). However, the phylogenetic placement of verified isolates based on types for these five genera confirmed that they should reside in Cytospora.

The type species of Valsa, V. ambiens (syn.: Sphaeria ambiens), was described from Crataegus and Fagus in Europe. Due to the lack of a suitable type in the Persoon herbarium, Urban (1957) selected a lectotype from Tilia from Scleromycetes Sueciae 8, which was mentioned in the sanctioning publication by Fries (1823). This species was reduced to synonymy with C. *leucosperma* (Farr & Rossman 2023), for which no host was given in the protologue. However, Fagus was mentioned as the first host in the sanctioning publication by Fries (1823), and was lectotypified with a Fries specimen by Spielman (1985). Because there is no extype isolate for V. ambiens and C. *leucosperma*, a new collection of V. ambiens was made from Fagus sylvatica in Austria. This collection is represented by isolate CBS 141469. Our analyses do not support CBS 141469 residing in a separate genus and this supports our treatment of Valsa as a synonym of Cytospora.

The type species of *Leucostoma*, *L. massarianum*, was described from *Sorbus aucuparia* in Europe. As part of the present study, this species was recollected from the same host in Austria (CBS 141473) and shown to cluster close to *C. avicennae*, *C. conceptaculata*, *C. japonica*, *C. ochracea*, *C. pruni-mume*, *C. sorbina* and *C. tibetensis*. *Leucostoma massarianum* was classified as *C. massariana* by Rossman *et al.* (2015) and our phylogenetic analyses confirmed that this fungus resides in *Cytospora*.

The type species of Valsella, V. salicis, was described from Salix aurita in Germany. There is some confusion regarding the names Valsella salicis and C. salicella, C. fugax and C. fertilis. According to Saccardo (1884), C. salicella was the asexual morph of V. salicis, C. fertilis the asexual morph of V. fertilis and C. salicis the asexual morph of Valsa salicina. However, Spielman (1985) introduced C. fugax as the asexual morph of V. salicis, but did not clarify the status of C. salicis. Cytospora fertilis was introduced as a synonym of V. fertilis, C. salicella and V. salicis (Farr & Rossman 2023). In the present study, the two isolates CBS 109754 and CBS 113699, both bearing the name V. salicis resided in different phylogenetic clades. These two species were clearly not conspecific and because there was no type material for C. fertilis, the name C. fuckeliana (a replacement name based on Valsella salicis) was applied to CBS 113699 (Clade 108) and C. salicis to CBS 109754 (Clade 103).

The type species of *Valseutypella*, *V. tristicha* (syn.: *Diatrype tristicha*), was introduced by von Höhnel (1918) collected from *Rosa sylvatica* in Italy. We designated isolate CBS 465.59 as a reference isolate for this species, based on its morphology, host and country of origin. Furthermore, a new combination in *Cytospora* as *C. tristicha* was provided.

The type species of *Cytospora* is *C. chrysosperma*. This fungus was described from *Populus* in Europe (Persoon 1794) and in the sanctioning publication (Fries 1823), only *Populus* was mentioned as a host. Material from Fries Scler. Suec. 154 cited in the sanctioning publication (Fries 1823) was available for lectotypification. *Valsa sordida* Nitschke (1870) is the sexual morph of *C. chrysosperma* (Spielman 1985). Maharachchikumbura *et al.* (2016) erroneously chose a specimen of *V. ambiens* from *Tilia* as neotype for *C. chrysosperma*. Because there was no extype isolate available for *C. chrysosperma*, we chose Herb. Univ. Upsaliensis (F-117599) 289268 (a copy of Scleromycetes Sueciae

154) as a lectotype and CBS 197.50 isolated from *Populus tremula* in the United Kingdom as an epitype of *C. chrysosperma*.

DNA sequence data and phylogenetic inference made it possible to resolve the sub-species and varieties that were introduced for the *Cytosporaceae* based on morphology. The subspecies and variety ranks were equivalent to species rank in this study. Three subspecies were recognised including *V. ambiens* subsp. *ambiens* (= *C. leucosperma*), *V. ambiens* subsp. *leucostomoides* and *V. ambiens* subsp. *dolosella*. Spielman (1985) introduced *C. annulata* as the asexual morph of *V. ambiens* subsp. *leucostomoides* and we consequently used *C. annulata* for both CBS 116810 and 118089 (Clade 24) that were previously identified as *V. ambiens* subsp. *leucostomoides*. There were no DNA sequence data available for *V. ambiens* subsp. *dolosella*. *Valsa mali* var. *pyri* also represented a distinct species. Based on sequence data generated in this study, a new name was provided for it.

## The C. chrysosperma complex

The taxonomic concept for the *Cytospora chrysosperma* complex was introduced by Fan *et al.* (2020) based on ITS and multigene analyses. Previously, the *C. chrysosperma* complex accommodated 12 species including *C. ailanthicola*, *C. chrysosperma*, *C. joaquinensis*, *C. longiostiolata*, *C. melnikii*, *C. populicola*, *C. rostrata*, *C. salicacearum*, *C. salicina*, *C. sophoriopsis*, *C. tritici* and *C. yakimana* (Fan *et al.* 2020, Travadon *et al.* 2022). In the present study, *Cytospora populicola* was regarded as a synonym of *C. chrysosperma*. DNA sequence data for *C. nobilis* was also provided. The strains originally regarded as *C. chrysosperma* (CFCC 89629, 89981 and 89982) were treated as a new species, *C. pseudochrysosperma*. Three other new species (*C. guyuanensis*, *C. eastringensis* and *C. sinensis*) were also described. Together with these additions, there are now 16 species included in the *C. chrysosperma* complex.

Species in the *C. chrysosperma* complex were found to be consistently associated with canker diseases of trees in the *Salicaceae* (Norphanphoun *et al.* 2017, Lawrence *et al.* 2018, Fan *et al.* 2020, Lin *et al.* 2023a). Most of these species had similar asexual morphs including conidial size and the presence of cytosporoid conidiomata (Group All, type a6). They were however easily separated based on DNA sequences and phylogenetic inference.

*Cytospora chrysosperma* was once believed to be the cause of poplar canker disease, a species with a global distribution (Biggs *et al.* 1983, Su *et al.* 2018). Although the biology of this species was extensively studied in China (Su *et al.* 2018), this should be interpreted with care, as taxonomic boundaries for species in the *C. chrysosperma* complex were unclear. For example, while the strain of *C. chrysosperma* (G-YS-11-C1) used by Su *et al.* (2018) belongs to the *C. chrysosperma* complex, it is not *C. chrysosperma*, but rather *C. pseudochrysosperma*, which is closely related to *C. chrysosperma* occurs only in Europe, and consequently results of biological studies conducted in China may not be applicable to the fungus found in Europe.

# Identification of Cytospora species

In 2024, a total of 677 *Cytospora* spp., 857 *Valsa* spp., 45 *Valsella* spp., three *Valseutypella* spp., and 26 *Leucostoma* spp. were regarded as having legitimate names in MycoBank. However, many species published prior to 2000 lack DNA sequence data,



and some taxa published prior to this date cannot be validated due to a lack of original material, which poses a serious limitation for their identification.

Species of *Cytospora* display a great diversity and complexity of morphological characteristics and ecologies. The asexual morph of the same species can have either a single or multiple ostioles (Lin et al. 2023). Spore size can fluctuate depending on the culture medium or the specific host plant (Fan et al. 2020, Jiang et al. 2020b, Lin et al. 2023). The asexual morph is recognized as part of the ectostroma when it forms within an ascostroma, a rare condition found in species like Valsa cincta Fr. and V. massariana (Adams et al. 2005). Despite such variations, certain morphological traits remain consistent within a species. These include the colour of the apical disc, the presence of single or multiple locules, whether these locules share a common wall, the presence of a central column, and the existence of a black conceptacle in the asexual morph. In the sexual morph, consistent features are also observed, such as the number of ascospores per ascus, the presence of single or multiple ostioles, and the presence or absence of a black conceptacle. These characteristics serve as reliable criteria to distinguish between species within the genus Cytospora, providing a valuable framework for mycologists and researchers in fungal taxonomy and evolution.

Particularly since DNA sequence data have become more commonly available, the number of recognized cryptic species have increased in *Cytospora* (Adams *et al.* 2005, Fan *et al.* 2020). However, the application of phylogenetic inference alone to identify species is not advised. When identifying and describing species of *Cytospora*, a polyphasic approach is recommended where DNA sequence data, morphological characteristics, host range and geographical distribution are all taken into consideration (Jayawardena *et al.* 2021, Koukol & Delgado 2021). While the present study provides a strong background for future studies, substantial research will be required before a stable phylogenetic backbone and reliable morphological traits become clear for all *Cytospora* species.

#### Host range

Many species of *Cytospora* appeared to have broad host ranges. *Cytospora chrysosperma* and *C. leucosperma* (= *Valsa ambiens*) were reported from 150 and 167 different hosts respectively (http:// nt.ars-grin.gov/fungaldatabases). However, these host ranges need to be treated with caution because many species were described based only on their morphological characteristics. As found for other fungi (Slippers *et al.* 2014, Liu *et al.* 2022), morphological characteristics commonly overlap, can fail to accurately define species boundaries, and often mask boundaries between cryptic species.

The multigene phylogenetic data provided in the present study illustrated how plant hosts commonly provide an inaccurate reflection of taxonomic boundaries in *Cytospora*. For example, the isolates of *C. ceratosperma* used in this study were collected from 10 different host plants in seven countries. In contrast, *C. eucalypti, C. berkeleyi* and *C. kunzei* were reported from single hosts and they could represent host-specific species. However, this might also reflect a sampling effect. It is likely that there are varying levels of host specificity in different species of *Cytospora,* as different species can co-occur on same host plant (Adams *et al.* 2005, Crous *et al.* 2019, Fan *et al.* 2020). This would be like the situation for species in the *Botryosphaeriaceae* (Phillips *et al.* 2013, Slippers *et al.* 2014) that represent fungi with a very similar biology

to *Cytospora*. More extensive sampling of these fungi as well as biological studies will be needed to better understand the biology and taxonomic boundaries in this genus.

### CONCLUSIONS

*Cytospora* species are commonly isolated fungi, many of which are implicated in studies of plant and particularly tree diseases. Very little is known regarding the biology of these fungi, many of which are apparently opportunistic pathogens that might contribute to disease when their host plants are under stress. They are in this regard very similar to the *Botryosphaeriaceae* that have been much more intensively studied, both in terms of their taxonomy and biology. We believe that much can be learned regarding *Cytospora* species by considering the results of studies on the *Botryosphaeriaceae*.

There has been a great need for a taxonomic revision of *Cytospora* for many years and many problems relating to the biology of these fungi relate to a poor understanding of their species boundaries. The present revision has provided detailed phylogenetic inference and morphological comparisons for several problematic taxa and introduced 44 new species of *Cytospora*. Some previously accepted species have been typified, but many well-known species are still in need of in-depth typification, for example *C. ribis*, *C. schulzeri* and *C. translucens*. To further resolve the evolutionary relationships and pathogenicity of *Cytospora* spp., intensive field sampling and biological studies are required. The results of the present taxonomic treatment should provide a foundation that will add future value to those studies.

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# DECLARATION ON CONFLICT OF INTEREST

The authors declare that there is no conflict of interest.

#### REFERENCES

- Abebe G, Hart JH, Adams RD (1990). The relationship of site factors to the incidence of *Cytospora* and *Septoria* cankers and poplar and willow borer in hybrid poplar plantations. *General Technical Report-North Central Forest Experiment Station, USDA Forest Service*: 163–171.
- 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.
- Adams GC, Wingfield WJ, Common R, et al. (2005). Phylogenetic relationships and morphology of Cytospora species and related teleomorphs (Ascomycota, Diaporthales, Valsaceae) from Eucalyptus. Studies in Mycology 52: 1–144.
- Adanson M (1763). Familles des Plantes. Vincent, Paris, France.
- Ariyawansa HA, Hyde KD, Jayasiri SC, et al. (2015). Fungal diversity notes 111–252 taxonomic and phylogenetic contributions to fungal taxa. Fungal Diversity 75: 27–274.
- Ashkan SM, Hedjaroude GHA (1981). Taxonomic and pathologic studies on form-genus Cytospora Ehrb. on friut trees in Iran, 1- Taxonomy. Iranian Journal of Plant Pathology 17: 14–28.
- Castlebury ME (1978). *Diaporthales* in North America: with emphasis on *Gnomonia* and its segregates. *Mycologia Memoirs* **7**: 1–232.
- Barr ME (1990). Prodromus to nonlichenized, pyrenomycetous members of class Hymenoascomycetes. *Mycotaxon* **39**: 43–184.
- Bertrand PF, English H (1976). Release and dispersal of conidia and ascospores of Valsa leucostoma. Phytopathology 66: 987–991.
- Bettucci L, Saravay M (1993). Endophytic fungi of *Eucalyptus globulus*: a preliminary study. *Mycological Research* 97: 679–682.
- Bills GF, Redlin SC, Carris LM (1996). Isolation and analysis of endophytic fungal communities from woody plants. In: *Endophytic fungi in grasses* and woody plants: systematics, ecology, and evolution (Redlin SC, Carris LM, eds). APS Press, USA: 31–65.
- Biggs AR, Davis DD, Merrill W (1983). Histopathology of cankers on Populus caused by Cytospora chrysosperma. Canadian Journal of Botany 61: 563–574.
- Bloomberg WJ (1962a). Cytospora canker of poplars: factors influencing the development of the disease. *Canadian Journal of Botany* **40**: 1271–1280.
- Bloomberg WJ (1962b). Cytospora canker of poplars: the moisture relations and anatomy of the host. *Canadian Journal of Botany* **40**: 1281–1292.
- Burgess T, Wingfield MJ (2001). Impact of fungal pathogens in natural forests ecosystems: A focus on *Eucalyptus*. In: *Microorganisms in plant conservation and biodiversity* (Sivasithamparam K, Dixon KW, eds). Kluwer Academic Press, The Netherlands: 285–306.
- Burks S, Jacobi WR, McIntyre GA (1998). Cytospora canker development on aspen in response to nitrogen fertilization. *Journal of Arboriculture* 24: 28–34.
- Castlebury LA, Rossman AY, Jaklitsch WJ, et al. (2002). A preliminary overview of the Diaporthales based on large subunit nuclear ribosomal DNA sequences. Mycologia 94: 1017–1031.
- Chapela IH (1989). Fungi in healthy stems and branches of American beech and aspen: a comparative study. *New Phytologist* **113**: 65–75.
- Checa J, Martinez AT (1989). Description of the anamorph of Valseutypella multicollis in culture. Mycotaxon 34: 43–45.
- Checa J, Moreno G, Barr ME (1986). Valseutypella multicollis sp. nov. Mycotaxon 25: 523–526.
- Chen SF, Pavlic D, Roux J, et al. (2011), Characterization of Botryosphaeriaceae from plantation-grown Eucalyptus species in South China. Plant Pathology 60: 739–751.
- Chen XF, Chen L, Wang L (2016). First report of 'Jun jujube' stem rot caused by *Cytospora* sp. in south of Xinjiang, China. *Journal of Plant Pathology* **98**: 369–377.
- Christensen CM (1940). Studies on the biology of Valsa sordida and Cytospora chrysosperma. Phytopathology **30**: 459–475.
- Crous PW, Hawksworth DL, Wingfield MJ (2015). Identifying and naming plant pathogenic fungi: Past, Present, and Future. *Annual Review of Phytopathology* 53: 247–267.
- Crous PW, Wingfield MJ, Cheewangkoon R, et al. (2019). Foliar pathogens of eucalypts. Studies in Mycology 94: 125–298.

- Dearness J, Hansbrough JR (1934). Cytospora infection following fire injury in western British Columbia. Canadian Journal of Research 10: 125–128.
- Défago G (1942). Seconde contribution à le connaissance des Valsées v.H. *Phytopathologische Zeitschrift* **14**: 103–147.
- Dennis RWG (1978). British Ascomycetes. 3rd edn. J. Cramer, Vaduz.
- Doidge EM (1950). The South African fungi and lichens to the end of 1945. Bothalia 5: 1–1094.
- Donk MA (1964). Nomina conservanda proposita I. Proposals in fungi. Deuteromycetes. Regnum Vegetabile 34: 7–15.
- Du Q, Zhao SF, Wu CL, et al. (2013). Root rot of Chinese jujube (Ziziphus jujuba) caused by Cytospora sacculus in China. Plant Disease 97: 1661.
- Dudka IO, Heluta VP, Tykhonenko YY, et al. (2004). Fungi of the Crimean Peninsula. M.G. Kholodny Institute of Botany, National Academy of Sciences of Ukraine.
- Ehrenberg CG (1818). *Sylvae mycologicae berolinenses.* Formis Theophili Bruschcke. Berlin, Germany.
- Ellis JB, Everhart BM (1892). The North American Pyrenomycetes. Ellis & Everhart, USA.
- Ellis MB, Ellis JP (1997). *Microfungi on land plant: an identification handbook.* Slough, Berkshire: Richmond Publishing Co. Ltd.
- Esfandiari E (1946). A second list of fungi collected in Iran. *Entomologie et Phytopathologie Appliquees* **2**: 10–16.
- Fan XL, Bezerra JDP, Tian CM, *et al.* (2020). *Cytospora* (*Diaporthales*) in China. *Persoonia* **45**: 1–45.
- Fan XL, Hyde KD, Liu M, et al. (2015a). Cytospora species associated with walnut canker disease in China, with description of a new species C. gigalocus. Fungal Biology 119: 310–319.
- Fan XL, Hyde KD, Yang Q, *et al.* (2015b). *Cytospora* species associated with canker disease of three anti-desertification plants in northwestern China. *Phytotaxa* **197**: 227–244.
- Fan XL, Liang YM, Ma R, et al. (2014a). Morphological and phylogenetic studies of Cytospora (Valsaceae, Diaporthales) isolates from Chinese scholar tree, with description of a new species. Mycoscience 55: 252–259.
- Fan XL, Tian CM, Yang Q, *et al.* (2014b). *Cytospora* from *Salix* in northern China. *Mycotaxon* **129**: 303–315.
- Farr DF, Bills GF, Chamuris GP, et al. (1989). Fungi on plants and plant products in the United States. APS Press, USA.
- Farr DF, Rossman AY (2024). Fungal Databases, Systematic Mycology and Microbiology Laboratory, ARS, USDA. http://nt.ars-grin.gov/ fungaldatabases/.
- Fisher PJ, Petrini O, Sutton BC (1993). A comparative study of fungal endophytes in leaves, xylem and bark of *Eucalyptus* in Australia and England. *Sydowia* **45**: 338–345.
- Fotouhifar KB, Hedjaroude GA, Leuchtmann A (2010). ITS rDNA phylogeny of Iranian strains of *Cytospora* and associated teleomorphs. *Mycologia* **102**: 1369–1382.
- Fries EM (1823). Systema Mycologicum 2(2). Lund, Sweden.
- Fries EM (1825). Systema orbis vegetabilis. Pars 1. Plantae homonemeae. Typographia Academica, Lund, Sweden.
- Fries EM (1849). Summa Vegetabilium Scandinaveae. Sectio posterior. Typographia Academica, Uppsala, Sweden.
- Fuckel KWGL (1870). Symbolae mycologicae. Beiträge zur Kenntniss der Rheinischen Pilze. Jahrbücher des Nassauischen Vereins für Naturkunde 23–24: 1–459, 6 pls.
- Gibbs RD (1957). Patterns in the seasonal water content of trees. In: *The physiology of forest trees* (Thimann KV, ed). Ronald Press Corporation, USA: 43–69.
- Gilman JC, Tiffany LH, Lewis RM (1957). Iowa Ascomycetes II. Diaporthaceae: Valsaceae. Iowa State College Journal of Science 31: 623–647.
- Gonzalez FR (1916) Bosquejo de una Florula Hispalense de Micromicetos. Trabajos del Museo Nacional de Ciencias Naturales. Serie Botanica **10**: 1–221.
- Grove WB (1935). British stem- and leaf-fungi (Coelomycetes) I. Cambridge University Press, Cambridge, UK.
- Grove WB (1937). British stem- and leaf-fungi (Coelomycetes) II. Cambridge University Press, Cambridge, UK.

Guindon S, Dufayard JF, Lefort V, et al. (2010). New algorithms and

methods to estimate maximum-likelihood phylogenies: assessing the performance of PhyML 3.0. *Systematic Biology* **59**: 307–321.

- Guyon JC, Jacobi WR, McIntyre GA (1996). Effects of environmental stress on the development of Cytospora canker of aspen. *Plant Disease* **80**: 1320–1326.
- Gvritishvili MN (1973). Cytospora kantschavelii Gvrit. Mikologia Fitopatologia 7: 547.
- Gvritishvili MN (1982). The fungal genus Cytospora in the USSR. Izdatelstve Sabchota Sakarstvelo, Tbilici, Russia.
- Hanifeh S, Zafari D, Soleimani MJ, et al. (2022). Multigene phylogeny, morphology, and pathogenicity trials reveal novel Cytospora species involved in perennial canker disease of apple trees in Iran. Fungal Biology **126**: 707–726.
- Hayova VP, Minter DW (1998a). Valsa ceratosperma. IMI Descriptions of Fungi and Bacteria. Set 137 No. 1366.
- Hayova VP, Minter DW (1998b). *Valsa cypri*. IMI Descriptions of Fungi and Bacteria. Set 137 No. 1367.
- Hayova VP, Minter DW (1998c). Valsa malicola. IMI Descriptions of Fungi and Bacteria. Set 137 No. 1368.
- Hayova VP, Minter DW (1998d). Valsa sordida. IMI Descriptions of Fungi and Bacteria. Set 137 No. 1370.
- Hayova VP, Minter DW (2012a). *Leucostoma auerswaldii*. IMI Descriptions of Fungi and Bacteria. Set 193 No. 1922.
- Hayova VP, Minter DW (2012b). Valsa friesii. IMI Descriptions of Fungi and Bacteria. Set 193 No. 1924.
- Hillis DM, Bull JJ (1993). An empirical test of bootstrapping as a method for assessing confidence in phylogenetic analysis. *Systematic Biology* 42: 182–192.
- Höhnel von F (1917). System der Diaporthen. Berichte der Deutschen Botanischen Gesellschaft **35**: 631–638.
- Höhnel von F (1918). Mykologische Fragmente. Annales Mycologici Editi in Notitiam Scientiae Mycologicae Universalis 16: 35–174.
- Hubbes M (1960). Studies on the Valsaceae genus Valseutypella. Phytopathologische Zeitschrift **39**: 389–400.
- Hyde KD, Hongsanan S, Jeewon R, *et al.* (2016). Fungal diversity notes 367–490: taxonomic and phylogenetic contributions to fungal taxa. *Fungal Diversity* **80**: 1–270.
- Jami F, Marincowitz S, Crous PW, et al. (2018). A new Cytospora species pathogenic on Carpobrotus edulis in its native habitat. Fungal Systematics and Evolution 2: 37–43.
- Jayawardena RS, Hyde KD, de Farias ARG, et al. (2021). What is a species in fungal plant pathogens? Fungal Diversity **109**: 239–266.
- Jia AL, Chen BY, Lu HY, et al. (2024) Multigene phylogeny and morphology reveal three new species of Cytospora isolated from diseased plant branches in Fengtai District, Beijing, China. MycoKeys 101: 163–189.
- Jiang N, Fan XL, Tian CM, *et al.* (2020a). Reevaluating *Cryphonectriaceae* and allied families in *Diaporthales*. *Mycologia* **112**: 267–292.
- Jiang N, Yang Q, Fan XL, et al. (2020b). Identification of six Cytospora species on Chinese chestnut in China. MycoKeys 62: 1–25.
- Kalkanci A, Kustimur S, Turkoz Sucak G, et al. (2006). Fulminating fungal sinusitis caused by Valsa sordida, a plant pathogen, in a patient immunocompromised by acute myeloid leukemia. *Medical Mycology* 44: 501–509.
- Katoh K, Standley DM (2013). MAFFT multiple sequence alignment software version 7: improvements in performance and usability. *Molecular Biology and Evolution* **30**: 772–780.
- Kavak H (2005). Cytospora kunzei on plantation-grown Pinus elderica in Turkey. Australasian Plant Pathology **34**: 151–156.
- Kepley JB, Reeves FB, Jacobi WR, et al. (2015). Species associated with Cytospora canker on Populus tremuloides. Mycotaxon **130**: 783–805.
- Kobayashi T (1970). *Taxonomic studies of Japanese Diaporthaceae with special reference to their life-histories*. Bulletin of the Government Forest Experiment Station, Tokyo, Japan.
- Kirk PM (1981). New or interesting microfungi: III. A preliminary account of microfungi colonizing *Laurus nobilis* leaf litter. *Transactions of the British Mycological Society* **77**: 457–473.
- Kirk PM, Spooner BM (1989). Ascomycetes on leaf litter of Laurus nobilis and Hedera helix. Mycological Research 92: 335–346.
- Koukol O, Delgado G (2021). Why morphology matters: The negative consequences of hasty descriptions of putative novelties in asexual

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ascomycetes. IMA Fungus 12: 1-8.

- Lawrence DP, Travadon R, Pouzoulet J, *et al.* (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.
- Lawrence DP, Holland LA, Nouri MT, *et al.* (2018). Molecular phylogeny of *Cytospora* species associated with canker diseases of fruit and nut crops in California, with the descriptions of ten new species and one new combination. *IMA Fungus* **9**: 333–370.
- Li GJ, Hyde KD, Zhao RL, *et al.* (2016). Fungal diversity notes 253–366: taxonomic and phylogenetic contributions to fungal taxa. *Fungal Diversity* **78**: 1–237.
- Li WJ, McKenzie EHC, Liu JK, et al. (2020). Taxonomy and phylogeny of hyaline-spored coelomycetes. Fungal Diversity **100**: 279–801.
- Lin L, Pan M, Tian CM, et al. (2022). Fungal richness of Cytospora species associated with willow canker disease in China. Journal of Fungi 8: 377.
- Lin L, Pan M, Bezerra JDP, et al. (2023a). Re-evaluation of the fungal diversity and pathogenicity of *Cytospora* species from *Populus* in China. *Plant Disease* **107**: 83–96.
- Lin L, Pan M, Gao H, *et al.* (2023b). The potential fungal pathogens of *Euonymus japonicus* in Beijing, China. *Journal of Fungi* **9**: 271.
- Liu F, Ma ZY, Hou LW, et al. (2022) Updating species diversity of Collectotrichum, with a phylogenomic overview. Studies in Mycology 101: 1–56.
- Maharachchikumbura SS, Hyde KD, Jones EG, et al. (2015). Towards a natural classification and backbone tree for Sordariomycetes. Fungal Diversity 72: 199–301.
- Maharachchikumbura SS, Hyde KD, Jones EG, et al. (2016). Families of Sordariomycetes. Fungal Diversity 79: 1–317.
- Marincowitz S, Crous PW, Groenewald JZ, et al. (2008). Microfungi occurring on Proteaceae in the fynbos. Westerdijk Fungal Biodiversity Institute, Utrecht, The Netherlands.
- Marincowitz S, Pham NQ, Wingfield BD, et al. (2023). Microfungi associated with dying *Euphorbia mauritanica* in South Africa and their relative pathogenicity. *Fungal Systematics and Evolution* **12**: 59–71.
- McIntyre GA, Jacobi WR, Ramaley AW (1996). Factors affecting Cytospora canker occurrence on aspen. *Journal of Arboriculture* 22: 229.
- Mcneill J, Barrie FR, Buck WR, et al. (2012). International Code of Nomenclature for algae, fungi, and plants (Melbourne Code) adopted by the Eighteenth International Botanical Congress Melbourne, Australia, July 2011. In: *Regnum vegetabile* no. **154**. Koeltz Scientific Books, Germany.
- Mehrabi M, Mohammadi GE, Fotouhifar KB (2011). Studies on Cytospora canker disease of apple trees in Semirom region of Iran. *Journal of Agricultural Technology* **7**: 967–982.
- Monkai J, Tibpromma S, Manowong A, *et al.* (2021). Discovery of three novel *Cytospora* species in Thailand and their antagonistic potential. *Diversity* **13**: 488.
- Munk A (1953). The system of the Pyrenomycetes. Additions and corrections. *Svensk Botanisk Tidskrift* **50**: 68–90.
- Nannfeldt JA (1932). Studien über die Morphologie und Systematik der nichtlichenisierten inoperculaten Discomyceten. Nova Acta Regiae Societatis Scientiarum Upsaliensis 8: 1–368.
- Nitschke TRJ (1870). Pyrenomycetes Germanici. Die Kernpilze Deutschlands. Eduard Trewent, Breslau.
- Norphanphoun C, Doilom M, Daranagama DA, et al. (2017). Revisiting the genus Cytospora and allied species. Mycosphere 8: 51–97.
- Norphanphoun C, Raspé O, Jeewon R, *et al.* (2018). Morphological and phylogenetic characterisation of novel *Cytospora* species associated with mangroves. *Mycokeys* **38**: 93–120.
- Palavouzis SC, Tzamos S, Paplomatas E, et al. (2015). First report of Cytospora punicae isolated from pomegranate plants with symptom of collar rot in northern Greece. Journal of Plant Pathology 97: 216.
- Pan M, Zhu HY, Tian CM, et al. (2018). Cytospora piceae sp. nov. associated with canker disease of Picea crassifolia in China. Phytotaxa 383: 181–196.
- Pan M, Zhu HY, Tian CM, *et al.* (2021). Assessment of *Cytospora* isolates from conifer cankers in China, with the descriptions of four new *Cytospora* species. *Frontiers in Plant Science* **12**: 636460.

- Persoon CH (1794). Dispositio methodica fungorum. Neues Magazin für die Botanik 1: 81–128.
- Phillips AJL, Alves A, Abdollahzadeh J, et al. (2013). The Botryosphaeriaceae: genera and species known from culture. Studies in Mycology 76: 51–167.
- Posada D, Crandall KA (1998). Modeltest: testing the model of DNA substitution. *Bioinformatics* **14**: 817–818.
- Proffer TJ, Hart JH (1988). Vegetative compatibility groups in *Leucocytospora kunzei. Phytopathology* **78**: 256–260.
- Rabenhorst GL (1844). *Deutschlands Kryptogamen-Flora*. Erster Band, Pilze, Leipzig, Germany.
- Rambaut A (2010). *FigTree v. 1.3.1*. Institute of Evolutionary Biology, University of Edinburgh, Edinburgh.
- Rannala B, Yang Z (1996). Probability distribution of molecular evolutionary trees: a new method of phylogenetic inference. *Journal of Molecular Evolution* 43: 304–311.
- Rayner RW (1970). A mycological colour chart. Commonwealth Mycological Institute and British Mycological Society, Kew, Surrey, UK.
- Reich RW, van der Kamp BJ (1993). Frost, canker, and dieback of Douglasfir in the central interior of British Columbia. *Canadian Journal of Forest Research* 23: 373–379.
- Robak H (1956). Some fungi occurring on died-back tops and branches of *Picea abies* and *Abies* spp. in western Norway. *Friesia* 5: 366–389.
- Roll-Hansen F, Roll-Hansen H (1980). Microorganisms which invade *Piceaabies* in seasonal stem wounds: II. Ascomycetes, Fungi imperfecti, and Bacteria. General discussion, *Hymenomycetes* included. *European Journal of Forest Pathology* **10**: 396–410.
- Ronquist F, Huelsenbeck JP (2003). MrBayes 3, Bayesian phylogenetic inference under mixed models. *Bioinformatics* **19**: 1572–1574.
- Rossman AY, Adams GC, Cannon PF, et al. (2015). Recommendations of generic names in *Diaporthales* competing for protection or use. *IMA Fungus* 6: 145–154.
- Rossman AY, Aime MC, Farr DF, et al. (2005). The coelomycetous genera Chaetomella and Pilidium represent a newly discovered lineage of inoperculate discomycetes. Mycological Progress 3: 275–290.
- Saccardo PA (1882). Sylloge Fungorum 1: i–xviii, 1–768. Typis Seminarii, Padua, Italy.
- Saccardo PA (1884). Sylloge Fungorum 3: i–ii, 1–860. Typis Seminarii, Padua, Italy.
- Scharif G, Ershad D (1966). A list of fungi on cultivated plants, shrubs and trees of Iran. Ministry of Information Press, Tehran, Iran.
- Schoeneweiss DF (1981). The role of environmental stress in diseases of woody plants. *Plant Disease* 65: 308–314.
- Schoeneweiss DF (1983). Drought predisposition to *Cytospora* canker in blue spruce. *Plant Disease* 67: 383–385.
- Senanayake IC, Crous PW, Groenewald JZ, et al. (2017). Families of Diaporthales based on morphological and phylogenetic evidence. Studies in Mycology 86: 217–296.
- Senanayake IC, Jeewon R, Chomnunti P, et al. (2018). Taxonomic circumscription of *Diaporthales* based on multigene phylogeny and morphology. *Fungal Diversity* **93**: 241–443.
- Sharma JK, Mohanan C, Florence EJM (1985) Disease survey in nurseries and plantations of forest tree species grown in Kerala. *Kerala Forest Research Institute Research Report* **36**: 258–262.
- Sinclair WA, Lyon HH, Johnson WT (1987). *Diseases of Trees and Shrubs*. Coinstock Publishing Associates, Cornell University Press, USA.
- Slippers B, Roux J, Wingfield MJ, *et al.* (2014) Confronting the constraints of morphological taxonomy in the *Botryosphaeriales*. *Persoonia* **33**: 155–168.
- Sivanesan A (1983) *Cytospora sacchari*. CMI Descriptions of Pathogenic Fungi and Bacteria No. 777.
- Sogonov MV, Castlebury LA, Rossman AY, *et al.* (2008). Leaf-inhabiting genera of the *Gnomoniaceae*, *Diaporthales*. *Studies in Mycology* **62**: 1–77.
- Spielman LJ (1980). Cytospora vs. Cytispora: which is correct? Mycotaxon 10: 473–478.
- Spielman LJ (1985). A monograph of Valsa on hardwoods in North America. Canadian Journal of Botany 63: 1355–1378.
- Su Y, Li HG, Wang Y, et al. (2018). Poplar miR472a targeting NBS-LRRs

is involved in effective defence against the necrotrophic fungus *Cytospora chrysosperma. Journal of Experimental Botany* **69**: 5519–5530.

- Sydow P (1897). Beiträge zur Kenntnis der Pilzflora der Mark Brandenburg. I. Hedwigia Beiblatt **36**: 157–164.
- Tai FL (1979). Sylloge Fungorum Sinicorum. Science Press, Beijing, China.
- Tamura K, Stecher G, Peterson D, et al. (2013). MEGA6: Molecular Evolutionary Genetics Analysis version 6.0. Molecular Biology and Evolution 30: 2725–2729.
- Teng SC (1963). Fungi of China. Science Press, Beijing, China.
- Thambugala KM, Daranagama DA, Phillips AJL, et al. (2017). Microfungi on Tamarix. Fungal Diversity 82: 239–306.
- Thiers B (2017) Index Herbariorum: a global directory of public herbaria and associated staff. New York Botanical Garden's Virtual Herbarium. http://sweetgum.nybg.org/science/ih/
- Travadon R, Lawrence DP, Moyer MM, et al. (2022). Fungal species associated with grapevine trunk diseases in Washington wine grapes and California table grapes, with novelties in the genera Cadophora, Cytospora, and Sporocadus. Frontiers in Fungal Biology 3: 1018140.
- Triki MA, Gharbi Y, Cheffi M, *et al.* (2015). First report of *Cytospora punicae* associated with wood canker and branch dieback disease of pomegranate in Tunisia. *Journal of Plant Pathology* **97**: 550.

Tode HJ (1791). Fungi Mecklenburgenses Selecti. 2: 1-64.

- Tulasne LR, Tulasne C (1863). Selecta Fungoram Carpologia, Vol. 2. Oxford University Press, England.
- Urban Z (1957). Vorläufige Mitteilung der Ergebnisse einer Revision der Gattungen *Valsa* und *Valsella*. *Preslia* **29**: 394–395.
- Urban Z (1958). Revise ceskosl-ovenskych zástapcú rodů Valsa, Leucostoma a Valsella. Rozpravy Ceskoslovenské Akademie Ved 68: 1–250.
- Van Der Westhuizen GCA (1965). *Cytospora eucalypticola sp. nov.* on *Eucalypus saligna* from Northern Transvaal. *South African Forestry Journal* **54**: 8–11.
- Vasilyeva LN (1988). A new treatment of the family Valsaceae. Systema Ascomycetum 7: 13–21.
- Vasilyeva LN (1994). Pyrenomycetes of the Russian Far East, 2. Valsaceae. In: Institute of Biology and Pedology, Far East Branch of the Russian Academy of Sciences: Vladivostok, Russia.
- Videira SI, Groenewald JZ, Braun U, et al. (2016). All that glitters is not Ramularia. Studies in Mycology 83: 49–163.
- Wang X, Shi CM, Gleason ML, *et al.* (2020). Fungal species associated with apple Valsa canker in East Asia. *Phytopathology Research* **2**: 1–14.
- Wang YL, Lu Q, Decock C, et al. (2015). Cytospora species from Populus and Salix in China with C. davidiana sp. nov. Fungal Biology **119**: 420–432.
- Wehmeyer LE (1936). Cultural studies of three new Pyrenomycetes. Mycologia 28: 35–46.
- Wehmeyer LE (1975). The Pyrenomycetous Fungi. *Mycologia Memoirs* 6: 1–250.
- Wei JC (1979). *Identification of fungus handbook.* Shanghai Science and Technology Press, China.

- Wingfield MJ, De Beer ZW, Slippers B, *et al.* (2012). One fungus, one name promotes progressive plant pathology. *Molecular Plant Pathology* **13**: 604–613.
- Worrall JJ, Adams GC, Tharp SC (2010). Summer heat and an epidemic of Cytospora canker of *Alnus*. *Canadian Journal of Plant Pathology* 32: 376–386.
- Yang Q, Fan XL, Crous PW, et al. (2015). Cytospora from Ulmus pumila in Northern China. Mycological Progress 14: 1–12.
- Zhang QT, Lu Q, He M, et al. (2014). Cytospora palm sp. nov. (Diaporthales, Ascomycota), a canker agent on Cotinus coggygria (Anacardiaceae) in Northern China. Cryptogamie, Mycologie **35**: 211–220.

Zhuang WY (2005). Fungi of northwestern China. Ithaca, NewYork, USA.

- Zhang ZX, Deng DF, Qi WJ, et al. (2013). Botryosphaeria dothidea, the causal agent of a new stem canker disease of Tatarian dogwood (*Cornus alba*) in China. Australasian Plant Pathology **42**: 113–119.
- Zhang L, Alvarez LV, Bonthond G, et al. (2019). Cytospora elaeagnicola sp. nov. associated with Narrow-leaved oleaster canker disease in China. Mycobiology 47: 319–328.

#### Supplementary Material: https://studiesinmycology.org/

Fig. S1. Phylogram of *Cytospora* resulting from a maximum likelihood analysis based on ITS gene. Numbers above the branches indicate ML bootstrap values (ML-BS  $\geq$  70 %) and Bayesian Posterior Probabilities (BPP  $\geq$  0.8). Ex-type isolates are in bold. Reference strains are marked with\*. Isolates from the present study are marked in blue.

Fig. S2. Phylogram of *Cytospora* resulting from a maximum likelihood analysis based on *act1* gene. Numbers above the branches indicate ML bootstrap values (ML-BS  $\geq$  70 %) and Bayesian Posterior Probabilities (BPP  $\geq$  0.8). Ex-type isolates are in bold. Reference strains are marked with\*. Isolates from the present study are marked in blue.

**Fig. S3.** Phylogram of *Cytospora* resulting from a maximum likelihood analysis based on *rpb2* gene. Numbers above the branches indicate ML bootstrap values (ML-BS  $\geq$  70 %) and Bayesian Posterior Probabilities (BPP  $\geq$  0.8). Ex-type isolates are in bold. Reference strains are marked with\*. Isolates from the present study are marked in blue.

**Fig. S4.** Phylogram of *Cytospora* resulting from a maximum likelihood analysis based on *tef1-a* gene. Numbers above the branches indicate ML bootstrap values (ML-BS  $\geq$  70 %) and Bayesian Posterior Probabilities (BPP  $\geq$  0.8). Ex-type isolates are in bold. Reference strains are marked with\*. Isolates from the present study are marked in blue.

Fig. S5. Phylogram of *Cytospora* resulting from a maximum likelihood analysis based on *tub2* gene. Numbers above the branches indicate ML bootstrap values (ML-BS  $\geq$  70 %) and Bayesian Posterior Probabilities (BPP  $\geq$  0.8). Ex-type isolates are in bold. Reference strains are marked with\*. Isolates from the present study are marked in blue.

**Table S1.** Isolates of this study used in the phylogenetic analyses. All the new isolates used in this study are in bold, the type materials are marked with \* and the reference isolates are marked with <sup>R</sup>.

