

Morphology and phylogeny of *Cytospora* (Cytosporaceae, Diaporthales) species associated with plant cankers in Tibet, China

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Abstract

During our biodiversity investigations in Tibet, China, typical *Cytospora* canker symptoms were observed on branches of hosts *Myricaria paniculate*, *Prunus cerasifera* and *Sibiraea angustata*. Samples were studied, based on morphological features coupled with multigene phylogenetic analyses of ITS, *act*, *rpb2*, *tef1* and *tub2* sequence data, which revealed two new species (*Cytospora myricicola* **sp. nov.** and *C. sibiraeicola* **sp. nov.**) and a known species (*C. populina*). In addition, *Cytospora populina* is newly discovered on the host *Prunus cerasifera* and in Tibet.

Key words: Ascomycota, molecular phylogeny, novel taxa, Sordariomycetes, taxonomy



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Introduction

The genus *Cytospora* (Cytosporaceae, Diaporthales, Sordariomycetes, Ascomycota) was proposed by Ehrenberg (1818) and *C. chrysosperma* was selected as the lectotype later (Donk 1964). *Cytospora* has priority over *Leucocytospora*, *Leucostoma*, *Valsa*, *Valsella* and *Valseutypella* based on the dual-nomenclature criterion (Adams et al. 2005; Rossman et al. 2015). Members of *Cytospora* are characterised by the single or labyrinthine, loculate stromata, filamentous conidiophores or asci and allantoid hyaline conidia or ascospores (Spielman 1985; Adams et al. 2005; Norphanphoun et al. 2017, 2018; Fan et al. 2020; Shang et al. 2020). Species identification in *Cytospora* was previously largely based on the host affiliation and morphological descriptions; however, molecular phylogeny combined with morphology and host affiliation have become the main approaches recently (Fan et al. 2020; Shang et al. 2020; Zhu et al. 2020). Currently, over 690 species epithets of *Cytospora* have been listed in Index Fungorum (<http://www.indexfungorum.org/>; 2023). However, most of these were regarded as synonyms and most descriptions were unable to identify them accurately (Adams et al. 2005; Fan et al. 2020; Pan et al. 2020).

Cytospora is distributed worldwide and often known to be associated with plant diseases (Monkai et al. 2021; Pan et al. 2021; Lin et al. 2022; Travadon et al. 2022).

For example, *C. chrysosperma* is the main canker disease pathogen of polar and willow trees in China (Fan et al. 2014; Wang et al. 2015; Lin et al. 2023); *C. kuanchengensis* and additional five species are associated with Chinese chestnut cankers (Jiang et al. 2020); Fifteen *Cytospora* species were identified from destructive canker and dieback pathogens of woody hosts in the USA (Lawrence et al. 2018).

The Tibet Tibetan Autonomous Region is located on the Qinghai-Tibet Plateau, which is known as the third pole of the earth. During our biodiversity investigations in Tibet, typical fruiting bodies of *Cytospora* were discovered from *Myricaria paniculate* (Tamaricaceae), *Prunus cerasifera* (Rosaceae) and *Sibiraea angustata* (Rosaceae). The aim of the present study was to identify *Cytospora* species from the three hosts based on morphological features and molecular phylogeny of combined sequence data.

Materials and methods

Sample collection, morphology and isolation

Our biodiversity investigations were conducted in Lhasa and Shigatse cities in Tibet Tibetan Autonomous Region, China during 2022 and 2023. Diseased branches of *Myricaria paniculate*, *Prunus cerasifera* and *Sibiraea angustata* were observed and collected, packed in paper bags and returned to the laboratory for morphological study and fungal isolation.

Observation and description of *Cytospora* species was based on fruiting bodies naturally formed on the host barks. Ascostromata and conidiomata from tree barks were sectioned by hand using a double-edged blade and structures were observed under a dissecting microscope. At least 10 conidiomata/ascostromata, 10 asci and 50 conidia/ascospores were measured to calculate the mean size and standard deviation. Measurements are reported as maximum and minimum in parentheses and the range representing the mean plus and minus the standard deviation of the number of measurements is given in parentheses. Microscopy photographs were captured with a Nikon Eclipse 80i compound microscope, equipped with a Nikon digital sight DS-Ri2 high definition colour camera, using differential interference contrast illumination.

Isolates of *Cytospora* were obtained by removing the spore masses from the fruiting bodies on to clean PDA plates and incubating at 25 °C until spores germinated. Single germinated spores were then transferred to the new PDA plates and incubated at 25 °C in the dark. The cultures were deposited in the China Forestry Culture Collection Center (CFCC, <http://cfcc.caf.ac.cn/>) and the specimens in the Herbarium of the Chinese Academy of Forestry (CAF, <http://museum.caf.ac.cn/>).

DNA extraction, PCR amplification and sequencing

Genomic DNA was extracted from fresh fungal mycelia following the method described by Doyle and Doyle (1990). Polymerase chain reactions (PCR) were conducted to amplify the internal transcribed spacer region rDNA (ITS), the partial actin (*act*) region, RNA polymerase II second largest subunit (*rpb2*), translation elongation factor 1-alpha (*tef1*) and the partial beta-tubulin (*tub2*) gene using primers and conditions listed in Table 1. The PCR products were assayed via electrophoresis in 2% agarose gels. DNA sequencing was performed using an ABI PRISM 3730XL

Table 1. Primers and PCR protocols.

Gene Regions	Primers	PCR conditions	References
ITS	ITS5/ITS4	95 °C for 4 min, 35 cycles of 94 °C for 45 s, 48 °C for 1 min, and 72 °C for 2 min, 72 °C for 10 min	White et al. (1990)
<i>act</i>	ACT512F/ACT783R	95 °C for 4 min, 35 cycles of 94 °C for 45 s, 55 °C for 1 min, and 72 °C for 2 min, 72 °C for 10 min	Carbone and Kohn (1999)
<i>rpb2</i>	fRPB2-5f/fRPB2-7cR	95 °C for 5 min, 35 cycles of 95 °C for 1 min, 55 °C, 1.25 min, and 72 °C for 2 min, 72 °C for 10 min	Liu et al. (1999)
<i>tef1</i>	983F/2218R	94 °C for 3 min, 35 cycles of 94 °C for 30 s, 54 °C for 50 s, and 72 °C for 2 min, 72 °C for 10 min	Rehner (2001)
<i>tub2</i>	Bt2a/Bt2b	95 °C for 4 min, 35 cycles of 94 °C for 45 s, 54 °C for 1 min, and 72 °C for 2 min, 72 °C for 10 min	Glass and Donaldson (1995)

DNA Analyser with a BigDye Terminator Kit v.3.1 (Invitrogen, Waltham, MA, USA) at the Shanghai Invitrogen Biological Technology Company Limited (Beijing, China).

Sequence alignment and phylogenetic analyses

The obtained sequences of ITS, *act*, *rpb2*, *tef1* and *tub2* were assembled using SeqMan software version 7.1.0 (DNASTAR Inc., WI) and subjected to BLASTn search against the GenBank nucleotide database at National Center for Biotechnology Information (NCBI) to identify closely-related sequences. Sequences data of related taxa were obtained from previous publications (Fan et al. 2020; Lin et al. 2023) and downloaded from the GenBank database (Table 2). The sequences were aligned using MAFFT v.7 online web server (<http://mafft.cbrc.jp/alignment/server/index.html>, Katoh et al. 2019) under default settings. The Maximum Likelihood (ML) phylogenetic analysis was run in the CIPRES Science Gateway platform (Miller et al. 2010), using RAXMLHPC2 on the XSEDE (v. 8.2.10) tool under the GTR substitution model and 1000 non-parametric bootstrap replicates. Bayesian analysis was performed using MrBayes v. 3.2.6 on XSEDE at the CIPRES Science Gateway with four simultaneous Markov Chain runs for 1,000,000 generations. The resulting trees were visualised in FigTree v. 1.4.0 (Rambaut 2012).

Results

Phylogenetic analyses

The combined ITS, *act*, *rpb2*, *tef1* and *tub2* dataset consisted of 199 strains, with *Diaporthe vaccinii* (CBS 160.32) as the outgroup taxon (Table 2). The final alignment comprised 3,166 characters (ITS: 567, *act*: 323, *rpb2*: 741, *tef1*: 727, *tub2*: 808), including gaps. The final ML optimisation likelihood value of the best RAXML tree was -60353.67 and the matrix had 2069 distinct alignment patterns, with 40.24% undetermined characters or gaps. Estimated base frequencies were as follows: A = 0.244507, C = 0.288246, G = 0.237262, T = 0.229984; substitution rates AC = 1.372283, AG = 2.995828, AT = 1.353835, CG = 0.976452, CT = 5.021434, GT = 1.0; gamma distribution shape parameter α = 0.372885. The RAXML and Bayesian analyses yielded a similar tree topology. The topology of our phylogenetic tree is nearly identical to previous publications. Six isolates from the present study formed two new clades distinct from previously-known species named *Cytospora myricicola* sp. nov. and *C. sibiraeicola* sp. nov. and a known clade named *C. populina* (Fig. 1).

Table 2. GenBank accession numbers used in the phylogenetic analyses.

Species	Strain	Host	Origin	GenBank accession numbers				
				ITS	act	rpb2	tef1	tub2
<i>Cytospora ailanthicola</i>	CFCC 89970	<i>Ailanthus altissima</i>	China	MH933618	MH933526	MH933592	MH933494	MH933565
<i>Cytospora albobdisca</i>	CFCC 53161	<i>Platycladus orientalis</i>	China	MW418406	MW422899	MW422909	MW422921	MW422933
<i>Cytospora albobdisca</i>	CFCC 54373	<i>Platycladus orientalis</i>	China	MW418407	MW422900	MW422910	MW422922	MW422934
<i>Cytospora alba</i>	CFCC 55462 ^T	<i>Salix matsudana</i>	China	NR182387	OK303457	OK303516	OK303577	OK303644
<i>Cytospora alba</i>	CFCC 55463	<i>Salix matsudana</i>	China	MZ702596	OK303458	OK303517	OK303578	OK303645
<i>Cytospora ampulliformis</i>	MFLUCC 16-0583 ^T	<i>Sorbus intermedia</i>	Russia	KY417726	KY417692	KY417794	NA	NA
<i>Cytospora ampulliformis</i>	MFLUCC 16-0629	<i>Acer platanoides</i>	Russia	KY417727	KY417693	KY417795	NA	NA
<i>Cytospora amygdali</i>	CBS 144233 ^T	<i>Prunus dulcis</i>	USA	MG971853	MG972002	NA	MG971659	NA
<i>Cytospora atrocirrhatta</i>	CFCC 89615	<i>Juglans regia</i>	China	KR045618	KF498673	KU710946	KP310858	KR045659
<i>Cytospora atrocirrhatta</i>	CFCC 89616	<i>Juglans regia</i>	China	KR045619	KF498674	KU710947	KP310859	KR045660
<i>Cytospora beilininensis</i>	CFCC 50493 ^T	<i>Pinus armandii</i>	China	MH933619	MH933527	NA	MH933495	MH933561
<i>Cytospora beilininensis</i>	CFCC 50494	<i>Pinus armandii</i>	China	MH933620	MH933528	NA	MH933496	MH933562
<i>Cytospora berberidis</i>	CFCC 89927 ^T	<i>Berberis dasystachya</i>	China	KR045620	KU710990	KU710948	KU710913	KR045661
<i>Cytospora berberidis</i>	CFCC 89933	<i>Berberis dasystachya</i>	China	KR045621	KU710991	KU710949	KU710914	KR045662
<i>Cytospora bungeana</i>	CFCC 50495 ^T	<i>Pinus bungeana</i>	China	MH933621	MH933529	MH933593	MH933497	MH933563
<i>Cytospora bungeana</i>	CFCC 50496	<i>Pinus bungeana</i>	China	MH933622	MH933530	MH933594	MH933498	MH933564
<i>Cytospora californica</i>	CBS 144234 ^T	<i>Juglans regia</i>	USA	MG971935	MG972083	NA	MG971645	NA
<i>Cytospora carbonacea</i>	CFCC 89947	<i>Ulmus pumila</i>	China	KR045622	KP310842	KU710950	KP310855	KP310825
<i>Cytospora carpobroti</i>	CMW48981 ^T	<i>Carpobrotus edulis</i>	South Africa	MH382812	NA	NA	MH411212	MH411207
<i>Cytospora celtidicola</i>	CFCC 50497 ^T	<i>Celtis sinensis</i>	China	MH933623	MH933531	MH933595	MH933499	MH933566
<i>Cytospora celtidicola</i>	CFCC 50498	<i>Celtis sinensis</i>	Anhui, China	MH933624	MH933532	MH933596	MH933500	MH933567
<i>Cytospora centrivillosa</i>	MFLUCC 16-1206 ^T	<i>Sorbus domestica</i>	Italy	MF190122	NA	MF377600	NA	NA
<i>Cytospora centrivillosa</i>	MFLUCC 17-1660	<i>Sorbus domestica</i>	Italy	MF190123	NA	MF377601	NA	NA
<i>Cytospora ceratosperma</i>	CFCC 89624	<i>Juglans regia</i>	China	KR045645	NA	KU710976	KP310860	KR045686
<i>Cytospora ceratosperma</i>	CFCC 89625	<i>Juglans regia</i>	China	KR045646	NA	KU710977	KP310861	KR045687
<i>Cytospora ceratospermopsis</i>	CFCC 89626 ^T	<i>Juglans regia</i>	China	KR045647	KU711011	KU710978	KU710934	KR045688
<i>Cytospora ceratospermopsis</i>	CFCC 89627	<i>Juglans regia</i>	China	KR045648	KU711012	KU710979	KU710935	KR045689
<i>Cytospora chrysosperma</i>	CFCC 89629	<i>Salix psammophila</i>	China	KF765673	NA	KF765705	NA	NA
<i>Cytospora chrysosperma</i>	CFCC 89981	<i>Populus alba</i>	China	MH933625	MH933533	MH933597	MH933501	MH933568
<i>Cytospora chrysosperma</i>	CFCC 89982	<i>Ulmus pumila</i>	China	KP281261	KP310835	NA	KP310848	KP310818
<i>Cytospora cinnamomea</i>	CFCC 53178 ^T	<i>Prunus armeniaca</i>	China	MK673054	MK673024	NA	NA	MK672970
<i>Cytospora coryli</i>	CFCC 53162 ^T	<i>Corylus mandshurica</i>	China	MN854450	NA	MN850751	MN850758	MN861120
<i>Cytospora corylina</i>	CFCC 54684 ^T	<i>Corylus heterophylla</i>	China	MW839861	MW815937	MW815951	MW815886	MW883969
<i>Cytospora corylina</i>	CFCC 54685	<i>Corylus heterophylla</i>	China	MW839862	MW815938	MW815952	MW815887	MW883970
<i>Cytospora cotini</i>	MFLUCC 14-1050 ^T	<i>Cotinus coggygria</i>	Russia	KX430142	NA	KX430144	NA	NA
<i>Cytospora cotoneastricola</i>	CF 20197027	<i>Cotoneaster</i> sp.	China	MK673072	MK673042	MK673012	MK672958	MK672988
<i>Cytospora cotoneastricola</i>	CF 20197028	<i>Cotoneaster</i> sp.	China	MK673073	MK673043	MK673013	MK672959	MK672989
<i>Cytospora curvata</i>	MFLUCC 15-0865 ^T	<i>Salix alba</i>	Russia	KY417728	KY417694	NA	NA	NA
<i>Cytospora curvispora</i>	CFCC 54000 ^T	<i>Corylus heterophylla</i>	China	MW839851	MW815931	MW815945	MW815880	MW883963
<i>Cytospora curvispora</i>	CFCC 54001	<i>Corylus heterophylla</i>	China	MW839853	MW815932	MW815946	MW815881	MW883964
<i>Cytospora davidiana</i>	CXY 1350 ^T	<i>Populus davidiana</i>	China	KM034870	NA	NA	NA	NA
<i>Cytospora diopuiensis</i>	CFCC 55479	<i>Euonymus japonicus</i>	China	OQ344753	OQ410625	OQ398735	OQ398762	OQ398791

Species	Strain	Host	Origin	GenBank accession numbers				
				ITS	act	rpb2	tef1	tub2
<i>Cytospora diopuiensis</i>	CFCC 55527	<i>Euonymus japonicus</i>	China	OQ344754	OQ410626	OQ398736	OQ398763	OQ398792
<i>Cytospora discotoma</i>	CFCC 53137 ^T	<i>Platyclusus orientalis</i>	China	MW418404	MW422897	MW422907	MW422919	MW422931
<i>Cytospora discotoma</i>	CFCC 54368	<i>Platyclusus orientalis</i>	China	MW418405	MW422898	MW422908	MW422920	MW422932
<i>Cytospora donetzica</i>	MFLUCC 15-0864	<i>Crataegus monogyna</i>	Russia	KY417729	KY417695	KY417797	NA	NA
<i>Cytospora donetzica</i>	MFLUCC 16-0574 ^T	<i>Crataegus monogyna</i>	Russia	KY417731	KY417697	KY417799	NA	NA
<i>Cytospora donglingensis</i>	CFCC 53159 ^T	<i>Platyclusus orientalis</i>	China	MW418412	MW422903	MW422915	MW422927	MW422939
<i>Cytospora donglingensis</i>	CFCC 53160	<i>Platyclusus orientalis</i>	China	MW418414	MW422905	MW422917	MW422929	MW422941
<i>Cytospora elaeagni</i>	CFCC 89632	<i>Elaeagnus angustifolia</i>	China	KR045626	KU710995	KU710955	KU710918	KR045667
<i>Cytospora elaeagni</i>	CFCC 89633	<i>Elaeagnus angustifolia</i>	China	KF765677	KU710996	KU710956	KU710919	KR045668
<i>Cytospora elaeagnicola</i>	CFCC 52882 ^T	<i>Elaeagnus angustifolia</i>	China	MK732341	MK732344	MK732347	NA	NA
<i>Cytospora elaeagnicola</i>	CFCC 52883	<i>Elaeagnus angustifolia</i>	China	MK732342	MK732345	MK732348	NA	NA
<i>Cytospora erumpens</i>	CFCC 50022	<i>Prunus padus</i>	China	MH933627	MH933534	NA	MH933502	MH933569
<i>Cytospora erumpens</i>	CFCC 53163	<i>Prunus padus</i>	China	MK673059	MK673029	MK673000	MK672948	MK672975
<i>Cytospora eucalypti</i>	CBS 144241	<i>Eucalyptus globulus</i>	USA	MG971907	MG972056	NA	MG971617	NA
<i>Cytospora euonymicola</i>	CFCC 50499 ^T	<i>Euonymus kiautschovicus</i>	China	MH933628	MH933535	MH933598	MH933503	MH933570
<i>Cytospora euonymicola</i>	CFCC 50500	<i>Euonymus kiautschovicus</i>	China	MH933629	MH933536	MH933599	MH933504	MH933571
<i>Cytospora euonymina</i>	CFCC 89993 ^T	<i>Euonymus kiautschovicus</i>	China	MH933630	MH933537	MH933600	MH933505	MH933590
<i>Cytospora euonymina</i>	CFCC 89999	<i>Euonymus kiautschovicus</i>	China	MH933631	MH933538	MH933601	MH933506	MH933591
<i>Cytospora fraxinigena</i>	MFLU 17-0880 ^T	<i>Fraxinus ornus</i>	Italy	NR154859	NA	NA	NA	NA
<i>Cytospora fugax</i>	CXY 1371	<i>Populus simonii</i>	China	KM034852	NA	NA	NA	KM034891
<i>Cytospora fugax</i>	CXY 1381	<i>Populus ussuriensis</i>	China	KM034853	NA	NA	NA	KM034890
<i>Cytospora fusispora</i>	NFCCI 4372	NA	India	MN227694	NA	NA	NA	NA
<i>Cytospora galegicola</i>	MFLUCC 18-1199 ^T	<i>Galega officinalis</i>	Italy	MK912128	MN685810	MN685820	NA	NA
<i>Cytospora gyalocus</i>	CFCC 89620 ^T	<i>Juglans regia</i>	China	KR045628	KU710997	KU710957	KU710920	KR045669
<i>Cytospora gyalocus</i>	CFCC 89621	<i>Juglans regia</i>	China	KR045629	KU710998	KU710958	KU710921	KR045670
<i>Cytospora gigaspora</i>	CFCC 50014	<i>Juniperus procumbens</i>	China	KR045630	KU710999	KU710959	KU710922	KR045671
<i>Cytospora gigaspora</i>	CFCC 89634 ^T	<i>Salix psammophila</i>	China	KF765671	KU711000	KU710960	KU710923	KR045672
<i>Cytospora globosa</i>	MFLU 16-2054 ^T	<i>Abies alba</i>	Italy	MT177935	NA	MT432212	MT454016	NA
<i>Cytospora granati</i>	CBS 144237 ^T	<i>Punica granatum</i>	USA	MG971799	MG971949	NA	MG971514	NA
<i>Cytospora haidianensis</i>	CFCC 54056	<i>Euonymus alatus</i>	China	MT360041	MT363978	MT363987	MT363997	MT364007
<i>Cytospora haidianensis</i>	CFCC 54057 ^T	<i>Euonymus alatus</i>	China	MT360042	MT363979	MT363988	MT363998	MT364008
<i>Cytospora hippophaës</i>	CFCC 89639	<i>Hippophaë rhamnoides</i>	China	KR045632	KU711001	KU710961	KU710924	KR045673
<i>Cytospora hippophaës</i>	CFCC 89640	<i>Hippophaë rhamnoides</i>	China	KF765682	KF765730	KU710962	KP310865	KR045674
<i>Cytospora japonica</i>	CFCC 89956	<i>Prunus cerasifera</i>	China	KR045624	KU710993	KU710953	KU710916	KR045665
<i>Cytospora japonica</i>	CFCC 89960	<i>Prunus cerasifera</i>	China	KR045625	KU710994	KU710954	KU710917	KR045666
<i>Cytospora joaquinensis</i>	CBS 144235	<i>Populus deltoides</i>	USA	MG971895	MG972044	NA	MG971605	NA
<i>Cytospora junipericola</i>	MFLU 17-0882 ^T	<i>Juniperus communis</i>	Italy	MF190125	NA	NA	MF377580	NA
<i>Cytospora juniperina</i>	CFCC 50501 ^T	<i>Juniperus przewalskii</i>	China	MH933632	MH933539	MH933602	MH933507	NA

Species	Strain	Host	Origin	GenBank accession numbers				
				ITS	act	rpb2	tef1	tub2
<i>Cytospora juniperina</i>	CFCC 50502	<i>Juniperus przewalskii</i>	China	MH933633	MH933540	MH933603	MH933508	MH933572
<i>Cytospora kantschavelii</i>	CXY 1383	<i>Populus maximowiczii</i>	China	KM034867	NA	NA	NA	NA
<i>Cytospora kuanchengensis</i>	CFCC 52464 ^T	<i>Castanea mollissima</i>	China	MK432616	MK442940	MK578076	NA	NA
<i>Cytospora kuanchengensis</i>	CFCC 52465	<i>Castanea mollissima</i>	China	MK432617	MK442941	MK578077	NA	NA
<i>Cytospora leucosperma</i>	CFCC 89622	<i>Pyrus bretschneideri</i>	China	KR045616	KU710988	KU710944	KU710911	KR045657
<i>Cytospora leucosperma</i>	CFCC 89894	<i>Pyrus bretschneideri</i>	China	KR045617	KU710989	KU710945	KU710912	KR045658
<i>Cytospora longispora</i>	CBS 144236 ^T	<i>Prunus domestica</i>	USA	MG971905	MG972054	NA	MG971615	NA
<i>Cytospora longistiolata</i>	MFLUCC 16-0628	<i>Salix × fragilis</i>	Russia	KY417734	KY417700	KY417802	NA	NA
<i>Cytospora lumnitzericola</i>	MFLUCC 17-0508 ^T	<i>Lumnitzera racemosa</i>	Tailand	MG975778	MH253457	MH253461	NA	NA
<i>Cytospora mali</i>	CFCC 50028	<i>Malus pumila</i>	China	MH933641	MH933548	MH933606	MH933513	MH933577
<i>Cytospora mali</i>	CFCC 50029	<i>Malus pumila</i>	China	MH933642	MH933549	MH933607	MH933514	MH933578
<i>Cytospora mali-spectabilis</i>	CFCC 53181 ^T	<i>Malus spectabilis</i>	China	MK673066	MK673036	MK673006	MK672953	MK672982
<i>Cytospora melnikii</i>	CFCC 89984	<i>Rhus typhina</i>	China	MH933644	MH933551	MH933609	MH933515	MH933580
<i>Cytospora myricicola</i>	CFCC 59323^T	<i>Myricaria paniculate</i>	China	OR769868	OR767324	OR767338	OR767364	OR767351
<i>Cytospora myricicola</i>	CFCC 59324	<i>Myricaria paniculate</i>	China	OR769869	OR767325	OR767339	OR767365	OR767352
<i>Cytospora myricicola</i>	CFCC 59325	<i>Myricaria paniculate</i>	China	OR769870	OR767326	OR767340	OR767366	OR767353
<i>Cytospora myrtagena</i>	CFCC 52454	<i>Castanea mollissima</i>	China	MK432614	MK442938	MK578074	NA	NA
<i>Cytospora myrtagena</i>	CFCC 52455	<i>Castanea mollissima</i>	China	MK432615	MK442939	MK578075	NA	NA
<i>Cytospora nivea</i>	MFLUCC 15-0860	<i>Salix acutifolia</i>	Russia	KY417737	KY417703	KY417805	NA	NA
<i>Cytospora nivea</i>	CFCC 89641	<i>Elaeagnus angustifolia</i>	China	KF765683	KU711006	KU710967	KU710929	KR045679
<i>Cytospora notastroma</i>	NE_TFR5	<i>Populus tremuloides</i>	USA	JX438632	NA	NA	JX438543	NA
<i>Cytospora notastroma</i>	NE_TFR8	<i>Populus tremuloides</i>	USA	JX438633	NA	NA	JX438542	NA
<i>Cytospora ochracea</i>	CFCC 53164 ^T	<i>Cotoneaster</i> sp.	China	MK673060	MK673030	MK673001	MK672949	MK672976
<i>Cytospora oleicola</i>	CBS 144248 ^T	<i>Olea europaea</i>	USA	MG971944	MG972098	NA	MG971660	NA
<i>Cytospora olivacea</i>	CFCC 53174	<i>Prunus cerasifera</i>	China	MK673058	MK673028	MK672999	NA	MK672974
<i>Cytospora olivacea</i>	CFCC 53175	<i>Prunus dulcis</i>	China	MK673062	MK673032	MK673003	NA	MK672978
<i>Cytospora palm</i>	CXY 1276	<i>Cotinus coggygria</i>	China	JN402990	NA	NA	KJ781296	NA
<i>Cytospora palm</i>	CXY 1280 ^T	<i>Cotinus coggygria</i>	China	JN411939	NA	NA	KJ781297	NA
<i>Cytospora paracinnamomea</i>	CFCC 55453 ^T	<i>Salix matsudana</i>	China	MZ702594	OK303456	OK303515	OK303576	OK303643
<i>Cytospora paracinnamomea</i>	CFCC 55455 ^T	<i>Salix matsudana</i>	China	MZ702598	OK303460	OK303519	OK303580	OK303647
<i>Cytospora parakantschavelii</i>	MFLUCC 15-0857 ^T	<i>Populus × sibirica</i>	Russia	KY417738	KY417704	KY417806	NA	NA
<i>Cytospora parapistaciae</i>	CBS 144506 ^T	<i>Pistacia vera</i>	USA	MG971804	MG971954	NA	MG971519	NA
<i>Cytospora paraplurivora</i>	FDS-439	<i>Prunus armeniaca</i>	Canada	OL640182	OL631586	NA	OL631589	NA
<i>Cytospora paraplurivora</i>	FDS-564 ^T	<i>Prunus persica</i> var. <i>nucipersica</i>	Canada	OL640183	OL631587	NA	OL631590	NA
<i>Cytospora parasitica</i>	CFCC 53173	<i>Berberis</i> sp.	China	MK673070	MK673040	MK673010	MK672957	MK672986
<i>Cytospora paratranslucens</i>	MFLUCC 15-0506 ^T	<i>Populus alba</i> var. <i>bolleana</i>	Russia	KY417741	KY417707	KY417809	NA	NA
<i>Cytospora paratranslucens</i>	MFLUCC 16-0627	<i>Populus alba</i>	Russia	KY417742	KY417708	KY417810	NA	NA
<i>Cytospora phialidica</i>	MFLUCC 17-2498	<i>Alnus glutinosa</i>	Italy	MT177932	NA	MT432209	MT454014	NA
<i>Cytospora piceae</i>	CFCC 52841 ^T	<i>Picea crassifolia</i>	China	MH820398	MH820406	MH820395	MH820402	MH820387
<i>Cytospora piceae</i>	CFCC 52842	<i>Picea crassifolia</i>	China	MH820399	MH820407	MH820396	MH820403	MH820388
<i>Cytospora pingbianensis</i>	MFLUCC 18-1204 ^T	Undefined wood	China	MK912135	MN685817	MN685826	NA	NA
<i>Cytospora pistaciae</i>	CBS 144238 ^T	<i>Pistacia vera</i>	USA	MG971802	MG971952	NA	MG971517	NA

Species	Strain	Host	Origin	GenBank accession numbers				
				ITS	act	rpb2	tef1	tub2
<i>Cytospora platycladi</i>	CFCC 50504 ^T	<i>Platycladus orientalis</i>	China	MH933645	MH933552	MH933610	MH933516	MH933581
<i>Cytospora platycladi</i>	CFCC 50505	<i>Platycladus orientalis</i>	China	MH933646	MH933553	MH933611	MH933517	MH933582
<i>Cytospora platycladicola</i>	CFCC 50038 ^T	<i>Platycladus orientalis</i>	China	KT222840	MH933555	MH933613	MH933519	MH933584
<i>Cytospora platycladicola</i>	CFCC 50039	<i>Platycladus orientalis</i>	China	KR045642	KU711008	KU710973	KU710931	KR045683
<i>Cytospora plurivora</i>	CBS 144239 ^T	<i>Olea europaea</i>	USA	MG971861	MG972010	NA	MG971572	NA
<i>Cytospora populi</i>	CFCC 55472 ^T	<i>Populus</i> sp.	China	MZ702609	OK303471	OK303530	OK303591	OK303658
<i>Cytospora populi</i>	CFCC 55473	<i>Populus</i> sp.	China	MZ702610	OK303472	OK303531	OK303592	OK303659
<i>Cytospora populicola</i>	CBS 144240	<i>Populus deltoides</i>	USA	MG971891	MG972040	NA	MG971601	NA
<i>Cytospora populina</i>	CFCC 89644 ^T	<i>Salix psammophila</i>	China	KF765686	KU711007	KU710969	KU710930	KR045681
<i>Cytospora populina</i>	CFCC 58856	<i>Prunus cerasifera</i>	China	OR769873	OR767329	OR767343	OR767369	NA
<i>Cytospora populinopsis</i>	CFCC 50032 ^T	<i>Sorbus aucuparia</i>	China	MH933648	MH933556	MH933614	MH933520	MH933585
<i>Cytospora populinopsis</i>	CFCC 50033	<i>Sorbus aucuparia</i>	China	MH933649	MH933557	MH933615	MH933521	MH933586
<i>Cytospora predappioensis</i>	MFLUCC 17-2458 ^T	<i>Platanus hybrida</i>	Italy	MG873484	NA	NA	NA	NA
<i>Cytospora predappioensis</i>	MFLU 17-0327	<i>Platanus hybrida</i>	Italy	MH253451	MH253449	MH253450	NA	NA
<i>Cytospora prunicola</i>	MFLU 17-0995 ^T	<i>Prunus</i> sp.	Italy	MG742350	MG742353	MG742352	NA	NA
<i>Cytospora pruni-mume</i>	CFCC 53179	<i>Prunus armeniaca</i>	China	MK673057	MK673027	NA	MK672947	MK672973
<i>Cytospora pruni-mume</i>	CFCC 53180 ^T	<i>Prunus mume</i>	China	MK673067	MK673037	MK673007	MK672954	MK672983
<i>Cytospora pruinopsis</i>	CFCC 50034 ^T	<i>Ulmus pumila</i>	China	KP281259	KP310836	KU710970	KP310849	KP310819
<i>Cytospora pruinopsis</i>	CFCC 53153	<i>Ulmus pumila</i>	China	MN854451	MN850763	MN850752	MN850759	MN861121
<i>Cytospora pruinosa</i>	CFCC 50036	<i>Syringa oblata</i>	China	KP310800	KP310832	NA	KP310845	KP310815
<i>Cytospora pruinosa</i>	CFCC 50037	<i>Syringa oblata</i>	China	MH933650	MH933558	NA	MH933522	MH933589
<i>Cytospora pubescentis</i>	MFLUCC 18-1201 ^T	<i>Quercus pubescens</i>	Italy	MK912130	MN685812	MN685821	NA	NA
<i>Cytospora punicae</i>	CBS 144244	<i>Punica granatum</i>	USA	MG971943	MG972091	NA	MG971654	NA
<i>Cytospora quercicola</i>	MFLU 17-0881	<i>Quercus</i> sp.	Italy	MF190128	NA	NA	NA	NA
<i>Cytospora ribis</i>	CFCC 50026	<i>Ulmus pumila</i>	China	KP281267	KP310843	KU710972	KP310856	KP310826
<i>Cytospora ribis</i>	CFCC 50027	<i>Ulmus pumila</i>	China	KP281268	KP310844	NA	KP310857	KP310827
<i>Cytospora rosae</i>	MFLU 17-0885	<i>Rosa canina</i>	Italy	MF190131	NA	NA	NA	NA
<i>Cytospora rosicola</i>	CF 20197024 ^T	<i>Rosa</i> sp.	China	MK673079	MK673049	MK673019	MK672965	MK672995
<i>Cytospora rosigena</i>	MFLUCC 18-0921 ^T	<i>Rosa</i> sp.	Russia	MN879872	NA	NA	NA	NA
<i>Cytospora rostrata</i>	CFCC 89909	<i>Salix cupularis</i>	China	KR045643	KU711009	KU710974	KU710932	KR045684
<i>Cytospora rostrata</i>	CFCC 89910	<i>Salix cupularis</i>	China	KR045644	KU711010	KU710975	KU710933	NA
<i>Cytospora rusanovii</i>	MFLUCC 15-0853	<i>Populus × sibirica</i>	Russia	KY417743	KY417709	KY417811	NA	NA
<i>Cytospora rusanovii</i>	MFLUCC 15-0854 ^T	<i>Salix babylonica</i>	Russia	KY417744	KY417710	KY417812	NA	NA
<i>Cytospora salicacearum</i>	MFLUCC 15-0509	<i>Salix alba</i>	Russia	KY417746	KY417712	KY417814	NA	NA
<i>Cytospora salicacearum</i>	MFLUCC 15-0861	<i>Salix × fragilis</i>	Russia	KY417745	KY417711	KY417813	NA	NA
<i>Cytospora salicicola</i>	MFLUCC 14-1052 ^T	<i>Salix alba</i>	Russia	KU982636	KU982637	NA	NA	NA
<i>Cytospora salicicola</i>	MFLUCC 15-0866	<i>Salix</i> sp.	Thailand	KY417749	KY417715	KY417817	NA	NA
<i>Cytospora salicina</i>	MFLUCC 15-0862	<i>Salix alba</i>	Russia	KY417750	KY417716	KY417818	NA	NA
<i>Cytospora salicina</i>	MFLUCC 16-0637	<i>Salix × fragilis</i>	Russia	KY417751	KY417717	KY417819	NA	NA
<i>Cytospora schulzeri</i>	CFCC 50042	<i>Malus pumila</i>	China	KR045650	KU711014	KU710981	KU710937	KR045691
<i>Cytospora sibiraeae</i>	CFCC 50045 ^T	<i>Sibiraea angustata</i>	China	KR045651	KU711015	KU710982	KU710938	KR045692
<i>Cytospora sibiraeae</i>	CFCC 50046	<i>Sibiraea angustata</i>	China	KR045652	KU711016	KU710983	KU710939	KR045693
<i>Cytospora sibiraeicola</i>	CFCC 59100^T	<i>Sibiraea angustata</i>	China	OR769871	OR767327	OR767341	OR767367	OR767354
<i>Cytospora sibiraeicola</i>	CFCC 59101	<i>Sibiraea angustata</i>	China	OR769872	OR767328	OR767342	OR767368	OR767355
<i>Cytospora sophorae</i>	CFCC 50047	<i>Styphnolobium japonicum</i>	China	KR045653	KU711017	KU710984	KU710940	KR045694

Species	Strain	Host	Origin	GenBank accession numbers				
				ITS	act	rpb2	tef1	tub2
<i>Cytospora sophorae</i>	CFCC 89598	<i>Styphnolobium japonicum</i>	China	KR045654	KU711018	KU710985	KU710941	KR045695
<i>Cytospora sophoricola</i>	CFCC 89596	<i>Styphnolobium japonicum</i>	China	KR045656	KU711020	KU710987	KU710943	KR045697
<i>Cytospora sophoricola</i>	CFCC 89595 ^T	<i>Styphnolobium japonicum</i>	China	KR045655	KU711019	KU710986	KU710942	KR045696
<i>Cytospora sophoriopsis</i>	CFCC 55469	<i>Salix matsudana</i>	China	MZ702583	OK303445	OK303504	OK303565	OK303632
<i>Cytospora sophoriopsis</i>	CFCC 89600	<i>Styphnolobium japonicum</i>	China	KR045623	KU710992	KU710951	KU710915	KP310817
<i>Cytospora sorbi</i>	MFLUCC 16-0631 ^T	<i>Sorbus aucuparia</i>	Russia	KY417752	KY417718	KY417820	NA	NA
<i>Cytospora sorbicola</i>	MFLUCC 16-0584 ^T	<i>Acer pseudoplatanus</i>	Russia	KY417755	KY417721	KY417823	NA	NA
<i>Cytospora sorbicola</i>	MFLUCC 16-0633	<i>Cotoneaster melanocarpus</i>	Russia	KY417758	KY417724	KY417826	NA	NA
<i>Cytospora sorbina</i>	CF 20197660 ^T	<i>Sorbus tianschanica</i>	China	MK673052	MK673022	NA	MK672943	MK672968
<i>Cytospora spiraeae</i>	CFCC 50049 ^T	<i>Spiraea salicifolia</i>	China	MG707859	MG708196	MG708199	NA	NA
<i>Cytospora spiraeae</i>	CFCC 50050	<i>Spiraea salicifolia</i>	China	MG707860	MG708197	MG708200	NA	NA
<i>Cytospora spiraeicola</i>	CFCC 53138 ^T	<i>Spiraea salicifolia</i>	China	MN854448	NA	MN850749	MN850756	MN861118
<i>Cytospora spiraeicola</i>	CFCC 53139	<i>Tilia nobilis</i>	China	MN854449	NA	MN850750	MN850757	MN861119
<i>Cytospora tamaricicola</i>	CFCC 50507	<i>Rosa multiflora</i>	China	MH933651	MH933559	MH933616	MH933525	MH933587
<i>Cytospora tamaricicola</i>	CFCC 50508 ^T	<i>Tamarix chinensis</i>	China	MH933652	MH933560	MH933617	MH933523	MH933588
<i>Cytospora tanaitica</i>	MFLUCC 14-1057 ^T	<i>Betula pubescens</i>	Russia	KT459411	KT459413	NA	NA	NA
<i>Cytospora thailandica</i>	MFLUCC 17-0262 ^T	<i>Xylocarpus moluccensis</i>	Thailand	MG975776	MH253459	MH253463	NA	NA
<i>Cytospora thailandica</i>	MFLUCC 17-0263 ^T	<i>Xylocarpus moluccensis</i>	Thailand	MG975777	MH253460	MH253464	NA	NA
<i>Cytospora tibetensis</i>	CF 20197026	<i>Cotoneaster</i> sp.	China	MK673076	MK673046	MK673016	MK672962	MK672992
<i>Cytospora tibetensis</i>	CF 20197029	<i>Cotoneaster</i> sp.	China	MK673077	MK673047	MK673017	MK672963	MK672993
<i>Cytospora tibouchinae</i>	CPC 26333 ^T	<i>Tibouchina semidecandra</i>	France	KX228284	NA	NA	NA	NA
<i>Cytospora translucens</i>	CXY 1351	<i>Populus davidiana</i>	China	KM034874	NA	NA	NA	KM034895
<i>Cytospora translucens</i>	CXY 1359	<i>Populus</i> × <i>beijingensis</i>	China	KM034871	NA	NA	NA	KM034894
<i>Cytospora ulmi</i>	MFLUCC 15-0863 ^T	<i>Ulmus minor</i>	Russia	KY417759	NA	NA	NA	NA
<i>Cytospora verrucosa</i>	CFCC 53157 ^T	<i>Platyclusus orientalis</i>	China	MW418408	NA	MW422911	MW422923	MW422935
<i>Cytospora verrucosa</i>	CFCC 53158	<i>Platyclusus orientalis</i>	China	MW418410	MW422901	MW422913	MW422925	MW422937
<i>Cytospora vinacea</i>	CBS 141585 ^T	<i>Vitis interspecific</i>	USA	KX256256	NA	NA	KX256277	KX256235
<i>Cytospora viridistroma</i>	CBS 202.36 ^T	<i>Cercis canadensis</i>	USA	MN172408	NA	NA	MN271853	NA
<i>Cytospora viticola</i>	Cyt2	<i>Vitis interspecific</i>	USA	KX256238	NA	NA	KX256259	KX256217
<i>Cytospora viticola</i>	CBS 141586 ^T	<i>Vitis vinifera</i>	USA	KX256239	NA	NA	KX256260	KX256218
<i>Cytospora xinjiangensis</i>	CFCC 53182	<i>Rosa</i> sp.	China	MK673064	MK673034	MK673004	MK672951	MK672980
<i>Cytospora xinjiangensis</i>	CFCC 53183 ^T	<i>Rosa</i> sp.	China	MK673065	MK673035	MK673005	MK672952	MK672981
<i>Cytospora xinglongensis</i>	CFCC 52458	<i>Castanea mollissima</i>	China	MK432622	MK442946	MK578082	NA	NA
<i>Cytospora xinglongensis</i>	CFCC 52459	<i>Castanea mollissima</i>	China	MK432623	MK442947	MK578083	NA	NA
<i>Cytospora xylocarpi</i>	MFLUCC 17-0251 ^T	<i>Xylocarpus granatum</i>	Thailand	MG975775	MH253458	MH253462	NA	NA
<i>Cytospora zhaitangensis</i>	CFCC 56227 ^T	<i>Euonymus japonicus</i>	China	OQ344750	OQ410623	OQ398733	OQ398760	OQ398789
<i>Cytospora zhaitangensis</i>	CFCC 57537	<i>Euonymus japonicus</i>	China	OQ344751	OQ410624	OQ398734	OQ398761	OQ398790
<i>Diaporthe vaccinii</i>	CBS 160.32	<i>Vaccinium macrocarpon</i>	USA	KC343228	JQ807297	NA	KC343954	KC344196

Ex-type strains are indicated with (°) after the collection number; "NA" indicates unavailable sequences; sequences produced in the current study are in bold.

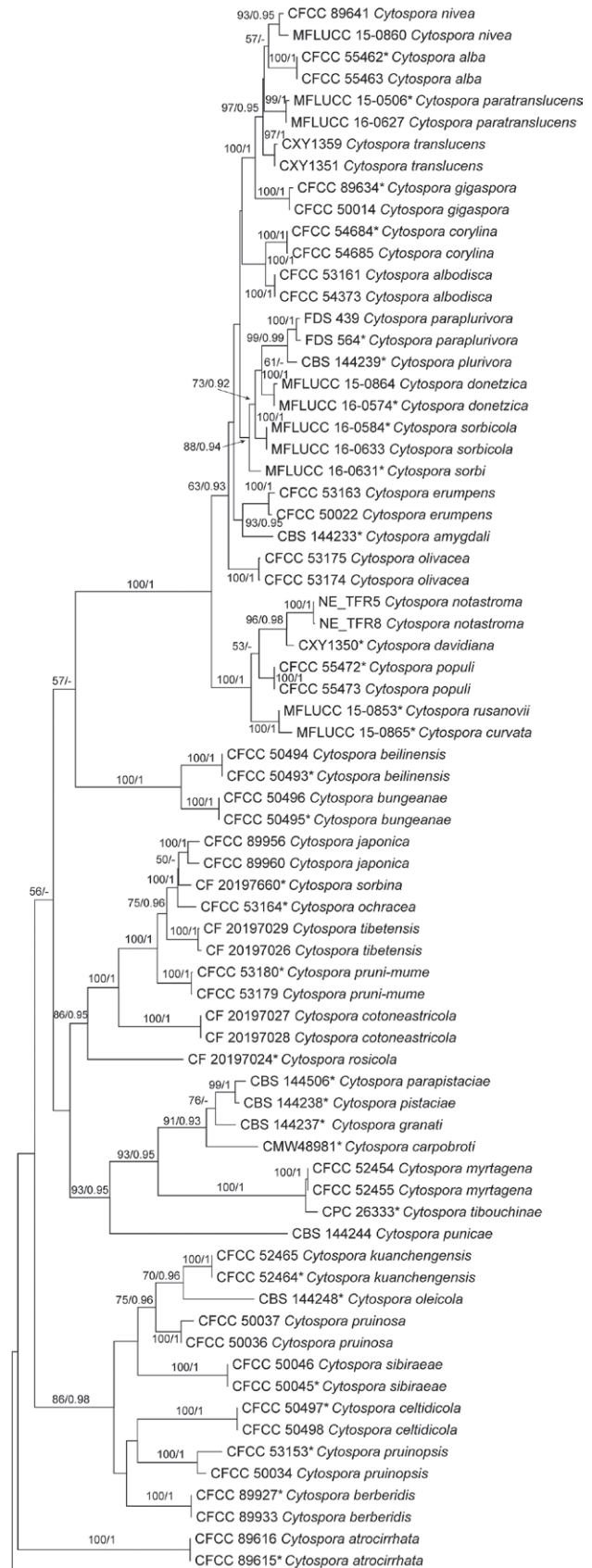


Figure 1. Maximum Likelihood tree generated from combined ITS, *act*, *rpb2*, *tef1* and *tub2* sequence data. Bootstrap support values $\geq 50\%$ and Bayesian posterior probabilities ≥ 0.90 are demonstrated at the branches. Ex-type cultures are marked with (*).

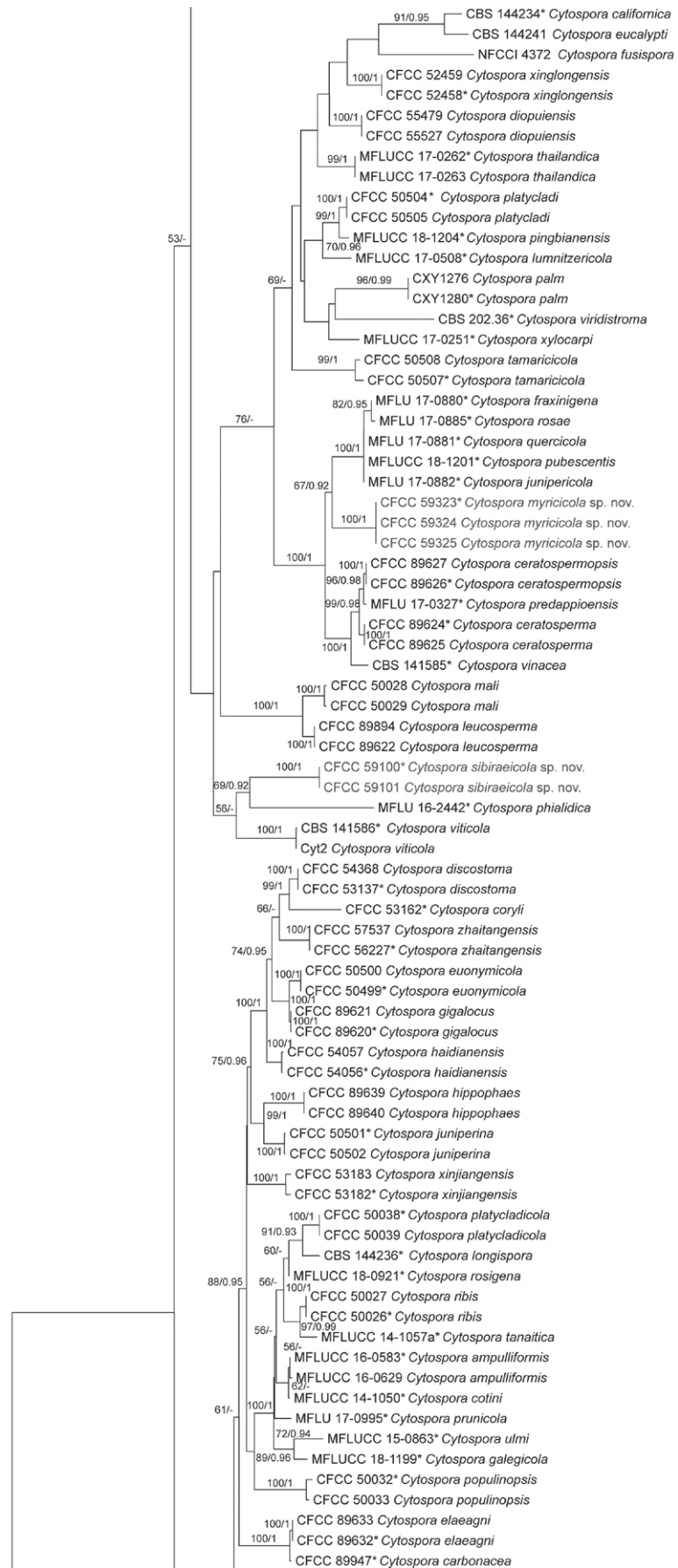


Figure 1. Continued.

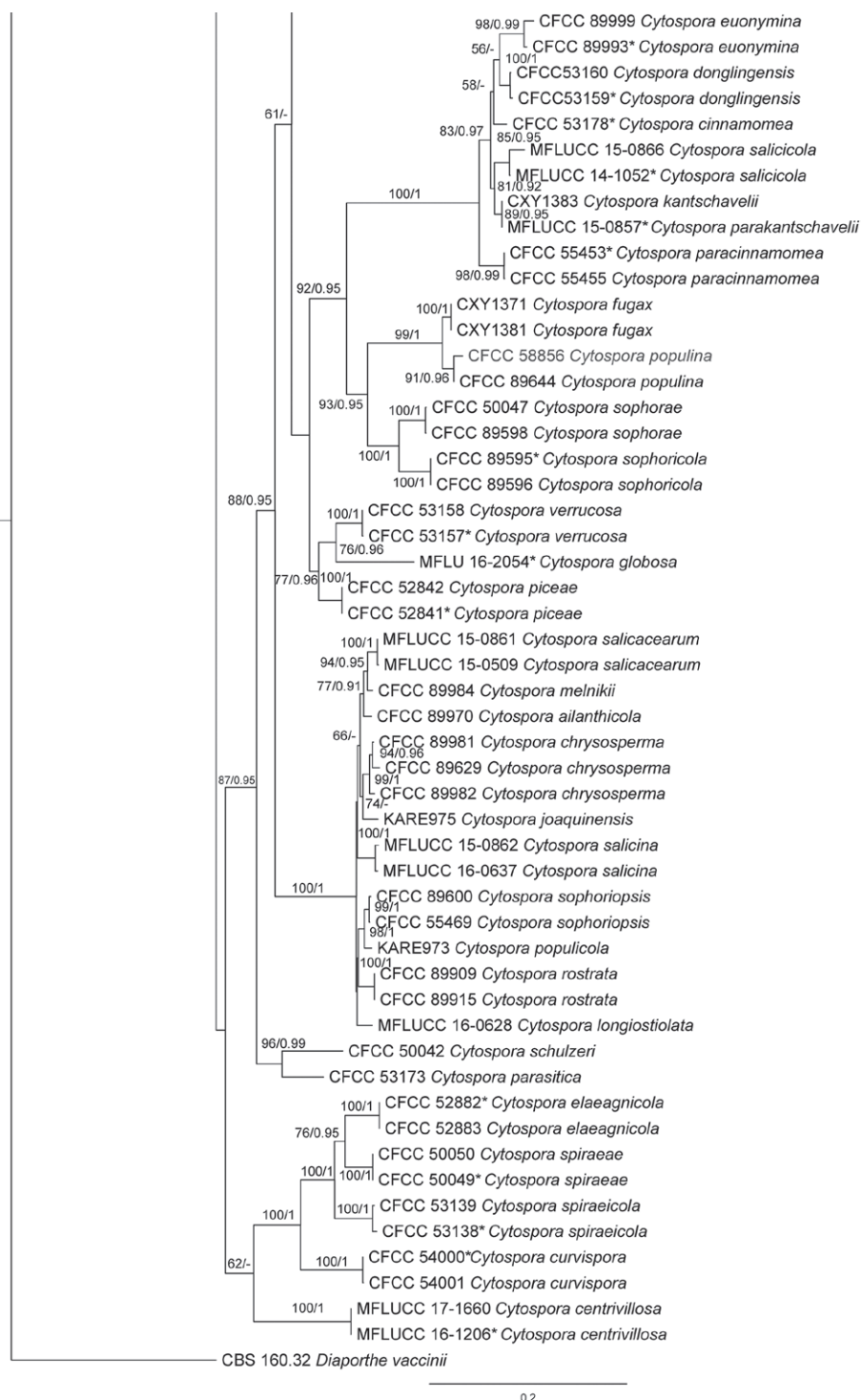


Figure 1. Continued.

Taxonomy

Cytospora myricicola Ning Jiang, sp. nov.

MycoBank No: 850240

Fig. 2

Etymology. “*myrici*” refers to the host genus *Myricaria* and “-cola” means inhabiting.

Holotype. CAF800083.

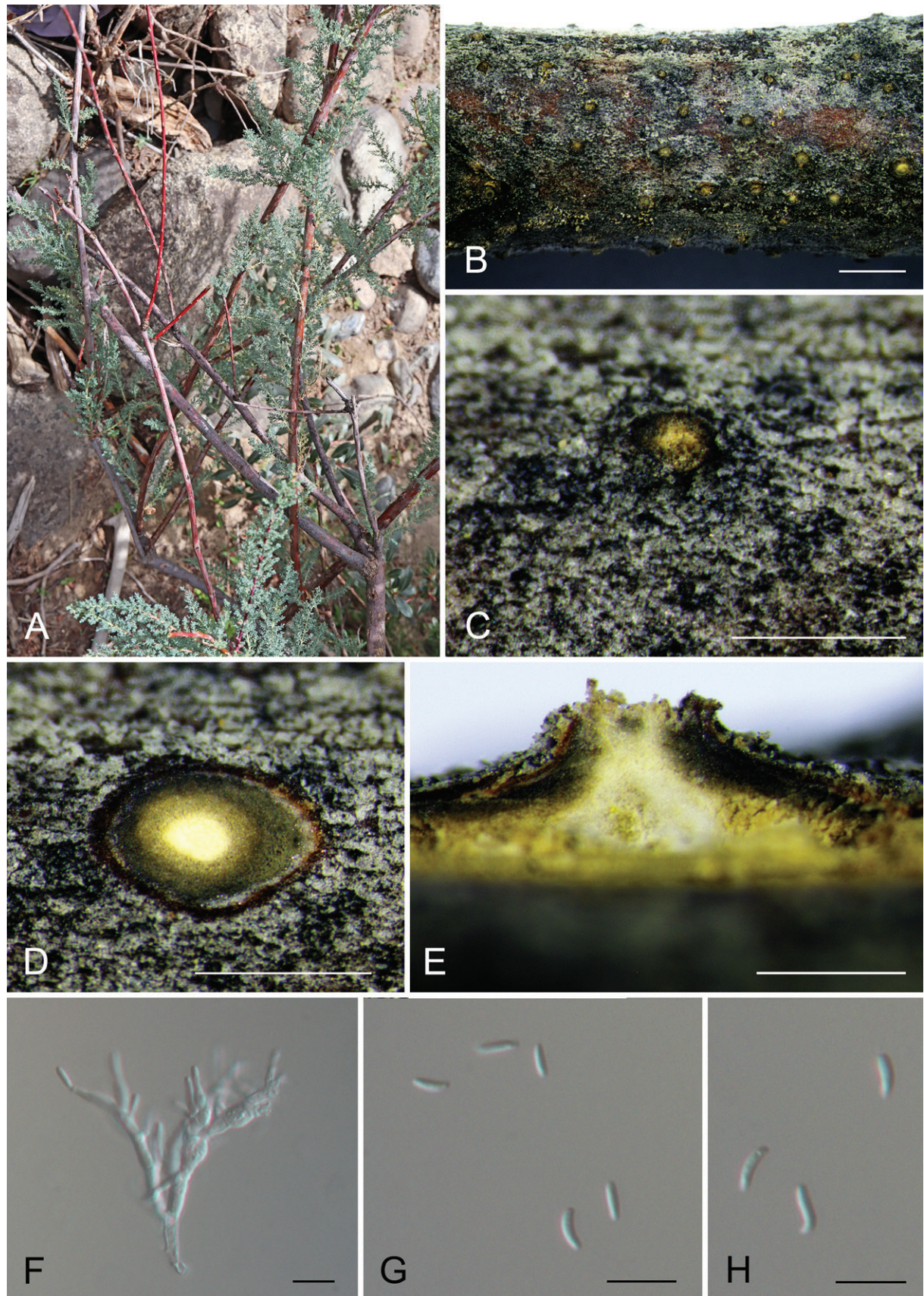


Figure 2. *Cytospora myricicola* (CAF800083, holotype) **A, B** canker disease symptom **C** conidioma **D** transverse section through a conidioma **E** longitudinal section through a conidioma **F** conidiophores and conidia **G, H** conidia. Scale bars: 2000 μm (**B**); 1000 μm (**C, D**); 500 μm (**E**); 10 μm (**F–H**).

Description. Associated with branch canker disease of *Myricaria paniculate*. **Sexual morph:** Undetermined. **Asexual morph:** Pycnidial stromata ostiolated, semi-immersed in the host bark, scattered, discoid, with multiple locules. Conceptacle dark brown to black, circular surrounded stromata. Ectostromatic disc dark yellow, circular to ovoid, (250–)350–450(–550) µm diam., with one ostiole per disc. Ostioles dark, at the same level as the disc, (35–)55–85(–100) µm diam. Locule numerous, arranged circularly or elliptically with independent walls (245–)300–450(–550) µm diam. Peridium comprising a few layers of cells of textura angularis, with innermost layer brown, outer layer brown to dark brown. Conidiophores hyaline, branched, thin-walled, filamentous. Conidigenous cells enteroblastic polyphialidic, 6.5–35.5 × 1.5–2.5 µm. Conidia hyaline, allantoid, smooth, aseptate, thin-walled, (4.4–)4.7–5.6(–5.8) × 1.4–1.7 µm ($\bar{x}$ = 5.2 × 1.6 µm).

Culture characteristics. Colonies on PDA flat, with flocculent aerial mycelium and entire edge, initially white, becoming dark and reaching 90 mm diameter after 10 days at 25 °C, sterile.

Materials examined. CHINA, Tibet Tibetan Autonomous Region, Lhasa City, Mozhugongka County, Riduo Township, Zen Village, on cankered branches of *Myricaria paniculate*, 28 July 2022, Jin Peng, Liu Yuanyuan, Jiang Ning and Liu Min (CAF800083, **holotype**); ex-type culture CFCC 59323. China, Tibet Tibetan Autonomous Region, Lhasa City, Mozhugongka County, Riduo Township, Renqinglin Village, on cankered branches of *Myricaria paniculate*, 28 July 2022, Jin Peng, Liu Yuanyuan, Jiang Ning and Liu Min (XZ010B); cultures CFCC 59324 and CFCC 59325.

Notes. Phylogenetically, *Cytospora myricicola* is close to *C. fraxinigena*, *C. junipericola*, *C. pubescentis*, *C. quercicola* and *C. rosae* (Fig. 1). Of these six species, only *C. myricicola*, *C. pubescentis* and *C. rosae* have asexual morph descriptions; *C. myricicola* (4.7–5.6 × 1.4–1.7 µm) is different from *C. pubescentis* (5.8–7.5 × 1.3–1.6 µm) by shorter conidia and from *C. rosae* (3–5 × 0.5–1 µm) by larger conidia (Senanayake et al. 2017; Shang et al. 2020). In addition, *C. myricicola* can be distinguished from the other five species by host and distribution (*C. myricicola* from *Myricaria paniculate* in China vs. *C. fraxinigena* from *Fraxinus ornus* in Italy vs. *C. junipericola* from *Juniperus communis* in Italy vs. *C. pubescentis* from *Quercus pubescens* in Italy vs. *C. quercicola* from *Quercus* sp. in Italy vs. *C. rosae* from *Rosa canina* in Italy) (Senanayake et al. 2017; Shang et al. 2020).

***Cytospora populina* (Pers.) Rabenh., Deutschl. Krypt.-Fl. (Leipzig) 1: 148. 1844**
Fig. 3

Description. Associated with branch canker disease of *Prunus cerasifera*. **Sexual morph:** Stromata immersed in bark. Ascstromata, erumpent through the surface of bark, lenticular, extending to a large circular area, (750–)900–1200(–1350) µm diam. Disc grey to black, circular to ovoid, (85–)100–150(–195) µm in diameter. Ostioles numerous, dark brown to black, at the same level as the disc, (25–)31–46(–52) µm diam. Locules dark brown, arranged circularly, flask-shaped to spherical, (180–)195–285(–340) µm diam. Asci clavate to elongate obovoid, (45–)55.5–62.5(–67) × (6.5–)8–12(–16) µm, 4-spored. Ascospores

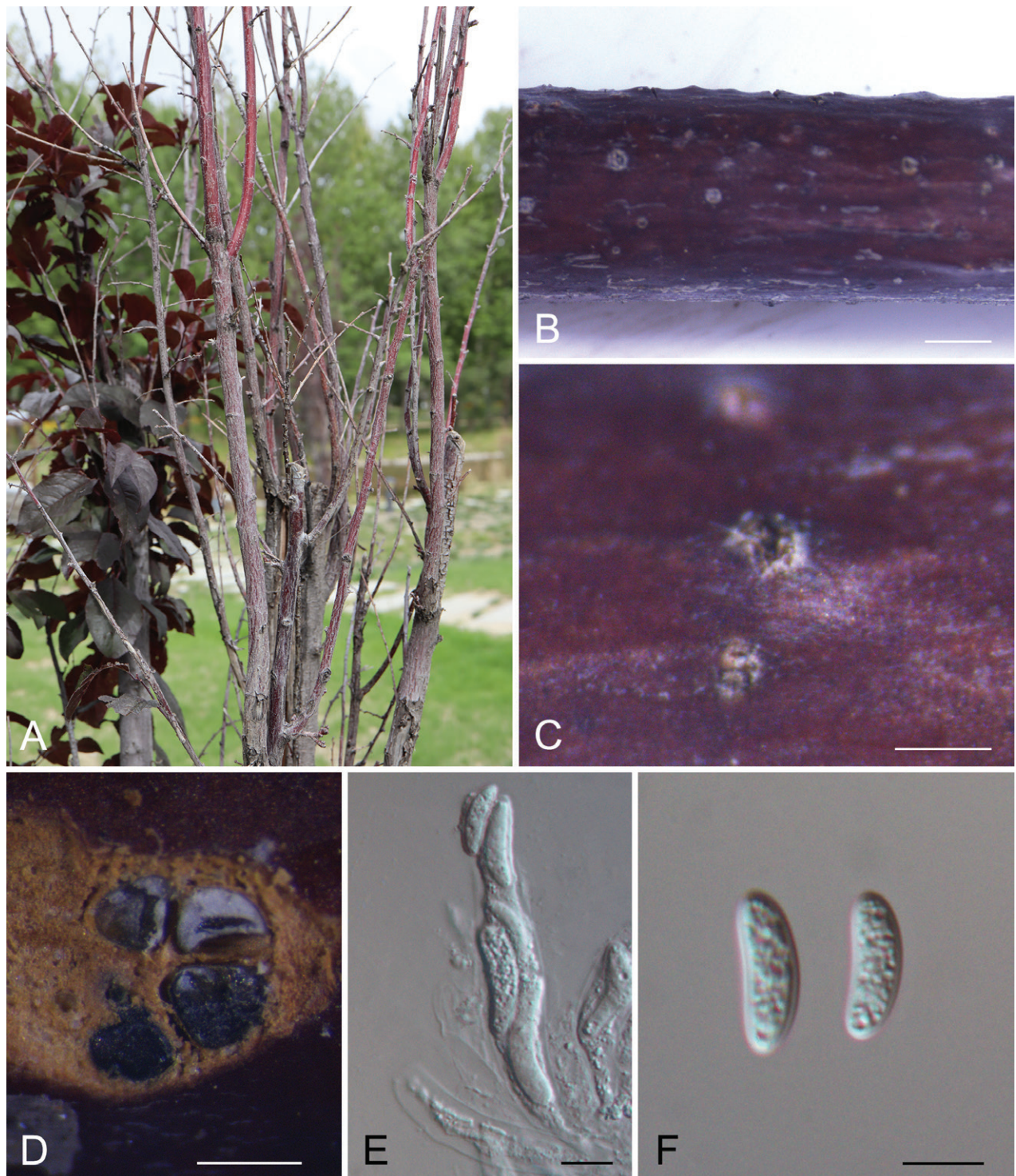


Figure 3. *Cytospora populina* (CAF800085) **A, B** canker disease symptom **C** ascostromata **D** transverse section through an ascostroma **E** asci and Ascospores **F** ascospores. Scale bars: 2000 μm (**B**); 500 μm (**C**); 200 μm (**D**); = 10 μm (**E–F**).

biseriate, elongate-allantoid, thin-walled, hyaline, aseptate, (15–)18.5–23.5(–25.5) $\times$ (4–)4.5–5.5(–6.5) μm ($\bar{x}$ = 20.4 $\times$ 5.1 μm). **Asexual morph:** Undetermined.

Culture characteristics. Colonies on PDA flat, with flocculent aerial mycelium and entire edge, initially white, becoming luteous and reaching 80 mm diameter after 10 days at 25 °C, sterile.

Materials examined. CHINA, Tibet Tibetan Autonomous Region, Shigatse City, Sangzhu District, Gongjuelinka Park, on cankered branches of *Prunus cerasifera*, 2 August 2022, Jin Peng, Jiang Ning and Liu Min (XZ063); culture CFCC 58856.

Notes. *Cytospora populina* has been reported from *Populus canadensis* in Argentina, *Salix psammophila* in Shaanxi Province of China and *Acer pubescens* *Rubus* sp. in Uzbekistan (Farr 1973; Fan et al. 2015; Gafforov 2017). This fungus is distinguished from the other *Cytospora* species by its 4-ascospored asci and undiscovered asexual state (Fan et al. 2015). In the present study, we firstly found this fungus causing cankered branches of *Prunus cerasifera* in Tibet, China.

***Cytospora sibiraeicola* Ning Jiang, sp. nov.**

MycoBank No: 850241

Fig. 4

Etymology. “*sibiraei*” refers to the host genus *Sibiraea* and “-cola” means inhabiting.

Holotype. CAF800084.

Description. Associated with branch canker disease of *Sibiraea angustata*.

Sexual morph: Undetermined. **Asexual morph:** Pycnidial stromata ostiolated, immersed or semi-immersed in the host bark, scattered, discoid, with multiple locules. Conceptacle black, circular surrounded stromata. Ectostromatic disc black, circular to ovoid, (200–)300–450(–500) μm diam., with one ostiole per disc. Ostioles dark, at the same level as the disc, (30–)60–80(–95) μm diam. Locule numerous, arranged circularly or elliptically with independent walls, (200–)250–380(–500) μm diam. Peridium comprising few layers of cells of textura angularis, with innermost layer brown, outer layer brown to dark brown. Conidiophores hyaline, unbranched, thin-walled, filamentous. Conidiogenous cells enteroblastic polyphialidic, $12.5\text{--}32.5 \times 2\text{--}3.5 \mu\text{m}$. Conidia hyaline, allantoid, smooth, aseptate, thin-walled, $(3.3\text{--})3.4\text{--}4.3(4.5) \times 1.2\text{--}1.6 \mu\text{m}$ ($\bar{x} = 3.9 \times 1.5 \mu\text{m}$).

Culture characteristics. Colonies on PDA flat, with flocculent aerial mycelium and undulate margin, initially white, becoming olivaceous grey and reaching 90 mm diameter after 10 days at 25 °C, sterile.

Materials examined. CHINA, Tibet Tibetan Autonomous Region, Lhasa City, Mozhugongka County, Riduo Township, Zen Village, on cankered branches of *Sibiraea angustata*, 28 July 2022, Jin Peng, Liu Yuanyuan, Jiang Ning and Liu Min (CAF800083, holotype); ex-type cultures CFCC 59100 and CFCC 59101.

Notes. *Cytospora sibiraeicola* is phylogenetically close to *C. phialidica* and *C. viticola* (Fig. 1). Morphologically, *C. sibiraeicola* ($3.4\text{--}4.3 \times 1.2\text{--}1.6 \mu\text{m}$) and *C. phialidica* ($3.5\text{--}5 \times 1\text{--}2 \mu\text{m}$) have much shorter conidia than *C. viticola* ($5.2\text{--}7 \times 0.9\text{--}1.6 \mu\text{m}$) (Lawrence et al. 2017; Li et al. 2020). In addition, these three species can be distinguished by the host and distribution (*C. sibiraeicola* from *Sibiraea angustata* in China vs. *C. phialidica* from *Alnus glutinosa* in Italy vs. *C. viticola* from *Vitis vinifera* in the USA) (Lawrence et al. 2017; Li et al. 2020).

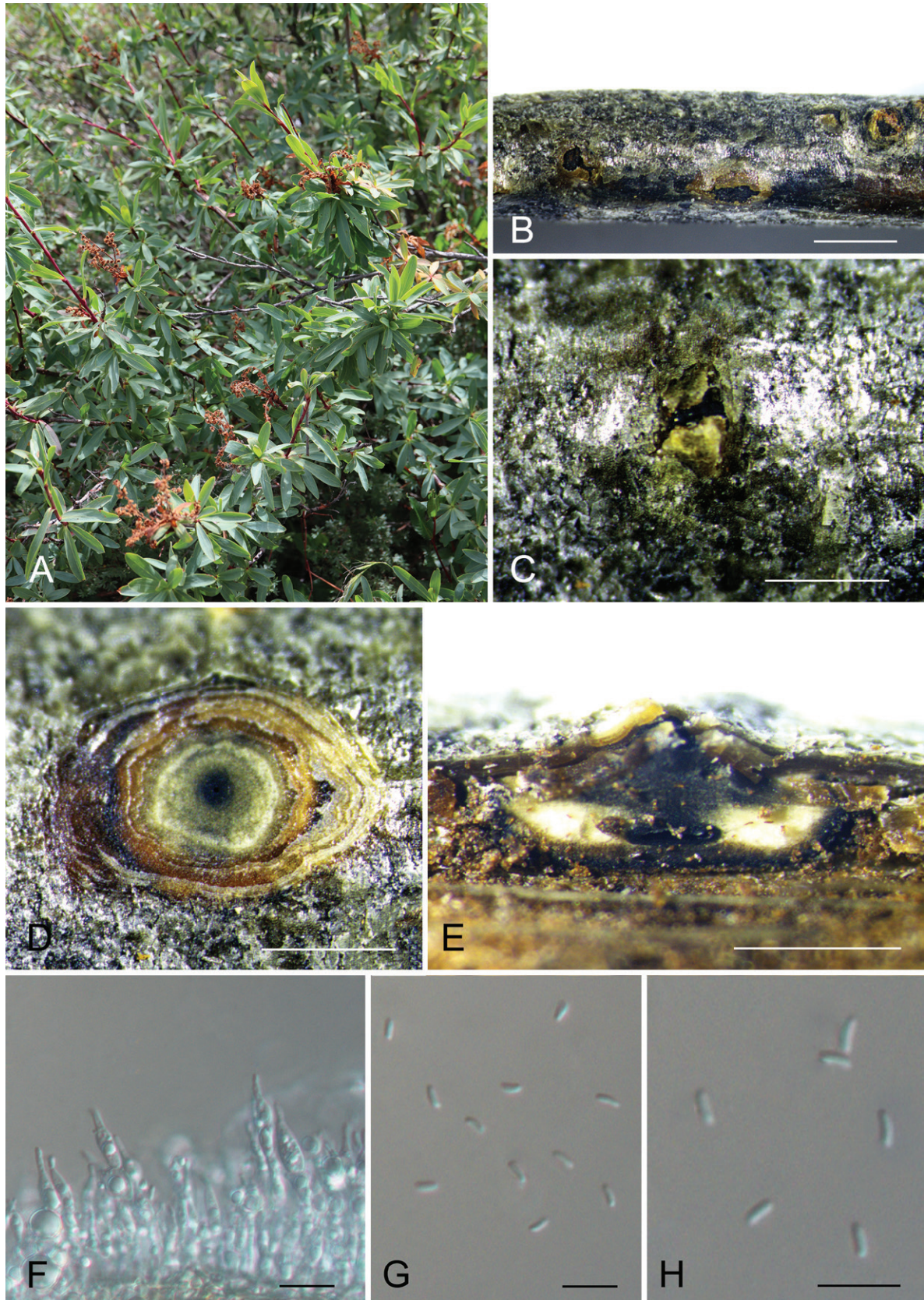


Figure 4. *Cytospora sibiraeicola* (CAF800084, holotype) **A, B** canker disease symptom **C** conidioma **D** transverse section through a conidioma **E** longitudinal section through a conidioma **F** conidiophores and conidia **G, H** conidia. Scale bars: 2000 μm (**B**); 1000 μm (**C–E**); 10 μm (**F–H**).

Discussion

Cytospora is a species-rich genus occurring on various plant hosts (Fotouhifar et al. 2010; Aiello et al. 2019; Jayawardena et al. 2019; Úrbez-Torres et al. 2020; Hanifeh et al. 2022). However, in the third pole of the Earth named Qinghai-Tibet Plateau, canker pathogens, such as *Cytospora*, have been seldom surveyed previously. In the comprehensive study on the genus *Cytospora* in China, only one species *C. chrysosperma* was recorded from *Ulmus pumila* in Tibet (Fan et al. 2020). Subsequently, *Cytospora cotoneastricola* and *C. tibetensis* from *Cotoneaster* sp. and *Cytospora rosicola* from *Rosa* sp. were discovered in Tibet (Pan et al. 2020). The current study introduces two new species named *C. myricicola* from *Myricaria paniculate* and *C. sibiraeicola* from *Sibiraea angustata* in Tibet, China. In addition, a new host record of *C. populina* on *Prunus cerasifera* was discovered.

To our knowledge, *Cytospora myricicola* is the first species of *Cytospora* discovered on the host genus *Myricaria* (Fan et al. 2020). *Cytospora sibiraeicola* and *C. sibiraeae* have been recorded from the host species *Sibiraea angustata* (Liu et al. 2015). *Cytospora sibiraeae* was described, based only on the sexual morph and is currently impossible to be distinguished from *C. sibiraeicola* morphologically (Liu et al. 2015). However, these two species occurring on *Sibiraea angustata* are phylogenetically obviously distinct (Fig. 1).

Species of *Cytospora* are known as opportunistic pathogens mainly infecting woody hosts and some of the species occur on a wide host range (Adams et al. 2005; Fan et al. 2020). The *Cytospora* species and their host association have been revealed in this study; however, further studies are required to confirm the fungal pathogenicity.

Additional information

Conflict of interest

The authors have declared that no competing interests exist.

Ethical statement

No ethical statement was reported.

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Author contributions

Conceptualization: JRL, JTL, NJ. Methodology: NJ. Formal analysis: JRL. Investigation: JRL, JTL, NJ. Resources: JRL, JTL, NJ. Data Curation: JRL, JTL. Writing - Original draft: JRL. Writing - Review and Editing: NJ. Visualization: JTL, NJ.

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Data availability

All of the data that support the findings of this study are available in the main text.

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