



Endophytic fungi in green manure crops; friends or foe?

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Abstract

Astragalus sinicus and *Vicia villosa*; are frequently applied green manure plants used in China. However, there is poor knowledge of the fungal endophytic community and the mycobiome of green manure crops. Field surveys were conducted during 2017–2019 in four provinces in China. Asymptomatic plant parts were collected. Using a culture-dependent method, 517 fungal isolates were obtained from *Astragalus sinicus* and *Vicia villosa*. These isolates were further identified using a combination of morphological and multi-loci phylogenetic analyses and were differentiated into 30 species in 15 genera in ten families belonging to only *Ascomycota*. Most isolated strains belonged to *Sordariomycetes*. The most dominant genus was *Fusarium*, with 381 isolates from both crops, while all other taxa were isolated less than 40 times. The similarity search on the *Fusarium* MLST database showed the 370 strains belonged to seven *Fusarium* complexes and one subclade. Eleven strains could not be assigned to any complex. The remaining 136 isolates were identified and assigned to 23 known and seven novel species. A total of 178 Operational Taxonomic Units (OTUs) were obtained from Illumina analysis and mainly classified into five phyla (*Ascomycota*, *Basidiomycota*, *Chytridiomycota*, *Cryptomycota*, and *Mucoromycota*). Overall OTUs were further assigned to 21 classes, 48 orders, 66 families, and 74 genera. Based on overall OTUs, the most abundant species was *Alternaria alternata*, which was also isolated from the culture-dependent method. Most species and genera recorded from the High Throughput Sequencing (HTS) approach were not obtained in the culture-dependent method (*Boeremia*, *Cladosporium*, *Filobasidium*, *Magnoportha*, *Mucor*, *Rhizoctonia*, *Sporidiobolus*). Functional annotation reveals that all *Ascomycetes* genera obtained in both approaches comprised several plant pathogenic species.

Potential beneficial and/or biocontrol strains were also identified. The common green manure crops used in China harbors a hidden, underexplored mycobiome which may comprise potential for application. These results will increase awareness of green manure practices. Precautions need to be in place when incorporating green manure crops in the soil, as these could facilitate inoculum sources for the next disease cycle of the main crop.

Keywords – 7 new taxa – Checklist – Culture dependent – Cover crops – High-throughput sequencing – Taxonomy

Introduction

Utilizing green manure (GM) crops in agriculture is an ancient practice in China. Some records show that 3,000 years ago, green manure was practiced in China by growing legumes and ploughing them into rice fields (Pieters 1927). Early Greek and Roman farmers recognized the value of legumes as green manure to improve soil fertility (Parsons 1984). Adding green manure to the soil will enhance the organic content of soil, maintains, and improve soil structure, provide a source of nitrogen (N) for crops, and reduce the losses of nutrients and soil erosion (Parsons 1984). Incorporation of organic matter into the soil can enhance the number of archaea bacteria and their activity (Yue et al. 2005). Moreover, the amount of active organic material for methane (CH₄) production can be improved (Lauren et al. 1994, Sethunathan et al. 2000). Even though these are presently essential and well-intentioned practices in traditional farming, they are suitable to use in intensive agricultural systems to reduce environmental problems (Parsons 1984).

China applies chemical fertilizers more than most other countries in their fields. This rate is 75% higher than the rest of the world (Peng et al. 2002). This excessive application of chemicals, especially N, leads to the emission of nitrous oxide (N₂O); a greenhouse gas (Shi et al. 2010). According to previous studies, poor soil organic matter content and imbalanced nutrient levels are the main factors that caused yield reduction in rice-based cropping systems (Namniar 1995, Reddy & Krishnaiah 1999). The application of organic materials has been recommended to protect the desired agricultural productivity and sustainability in a particular field (FAO 1993). Chinese milk vetch (*Astragalus sinicus* L. (AS) and hairy vetch (*Vicia villosa* Roth. (VV) (*Fabaceae*) are the major traditional leguminous crops used as N sources in organic crop production in China (Bo et al. 2012, FAO). These plants play an important role in maintaining rice soil fertility, especially in the double rice farming systems in southern China (Rong-shen & Qi-xiao 1981, Bo et al. 2012, Xie et al. 2016, Ntakirutimana et al. 2019). Rice is one of the prominent cereal crops in China with about 65 % of the population relying on rice (Zhang et al. 2005). Rice production has more than tripled in the past five decades and pesticides are misused in rice cultivation (Fang et al. 2004). Currently, most farmers use green manure in their rice fields leading toward sustainable rice production based on agroecology and biodiversity. Over-winter vetch crops: *Astragalus sinicus* and *Vicia villosa* grow in the spring and during the flowering stage of the manure crop, are ploughed. These crops are covered with soil for decomposition before the early season when rice is planted (Rong-shen & Qi-xiao 1981, Ntakirutimana et al. 2019).

In the 1990s, the identification of fungal species was mainly based on traditional approaches such as macroscopic or microscopic observations and culture-dependent analyses. Molecular analyses later provided an improved taxonomic resolution (Hyde et al. 2010, 2017, Cai et al. 2011, Tibpromma et al. 2017). One of the main reasons is that culture-dependent methods hinge on cultivability on specific media and thus exclude uncultivable fungi (Stewart 2012). Morphology-based identifications of fungal cultures are even problematic when strains do not develop any identifiable structures on the growth media (such as conidiomata and/or ascomata). This becomes more difficult when some species show phenotypic plasticity and/or belong to some complexes of cryptic species that cannot be differentiated morphologically (e.g.: species in genera *Colletotrichum*, *Diaporthe* and *Fusarium*). Therefore, applying traditional approaches alone may not provide a complete picture of fungal communities (Kozich et al. 2013). Therefore, High Throughput Sequencing (HTS) technologies have become more accessible recently, allowing in-

depth surveys of microbial diversity and other complex ecological communities (Peršoh 2015, Ampt et al. 2018, Tedersoo et al. 2018, 2020, 2021, Nilsson et al. 2019). HTS allows quick and cost-effective taxonomic assessments of a wide range of microbial groups. Most studies on plant microbes have focused on a single group (e.g., epiphytic fungi or bacteria, pathogenic fungi or bacteria, or mycorrhizal fungi or rhizobacteria) (Pérez-Jaramillo et al. 2018, Xia et al. 2020).

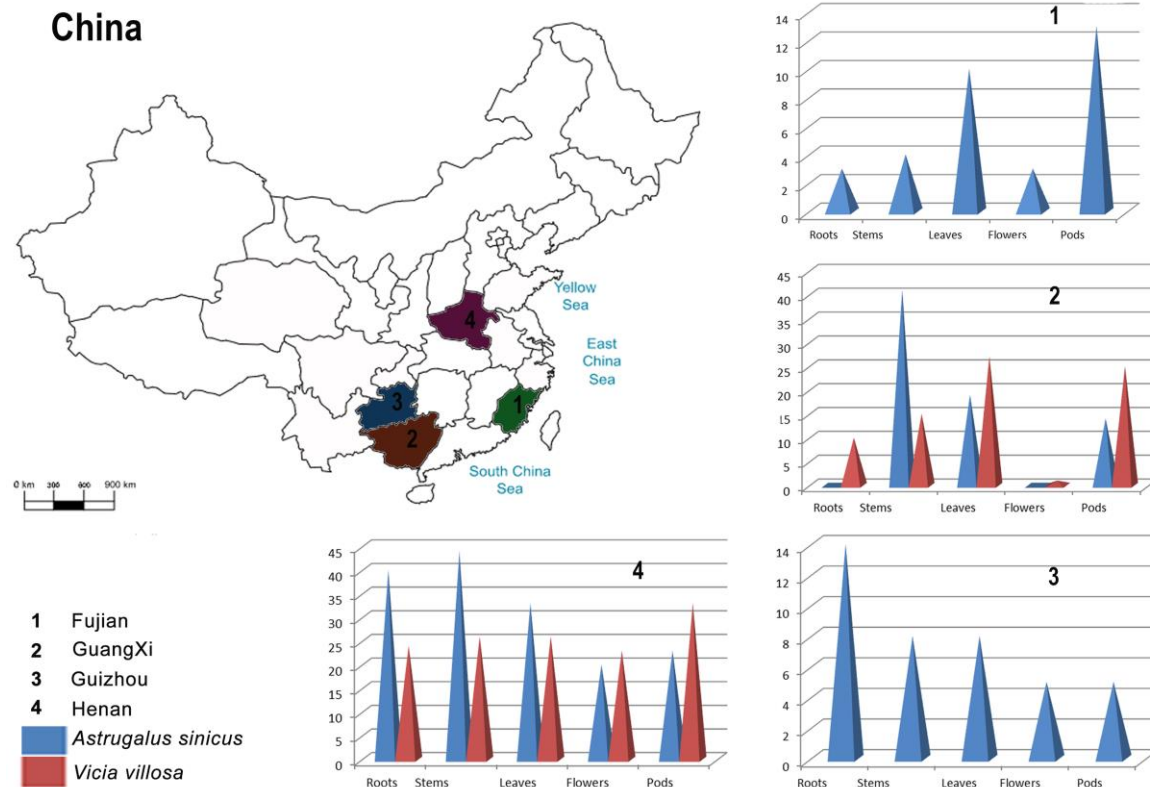


Figure 1 – Collection sites in four provinces in China. Clustered pyramid columns represent the frequency of the obtained fungal strains from the culture-dependent method with the host in each province.

Fungal diversity is an important aspect of crop and soil health in the field of agriculture (Selosse et al. 2006). Soil microbial communities play an important role in enhancing various biogeochemical processes (Basu et al. 2021). They are sensitive to disturbances, positively or negatively, that can lead to long-lasting ecosystem effects (Weller et al. 2002, Garbeva et al. 2004, Berg & Smalla 2009, Kallenbach & Grandy 2011, Lehman et al. 2015). However, the incorporation of green manure into the soil increases numerous benefits to soil, including the addition of organic C and the improvement of soil structure. Furthermore, manure crops protect the land from soil erosion and enhance the soil-water-holding capacity of the ecosystem. Green manure practices can alter the microbial community in the soil (Mendes et al. 1999, Abawi & Widmer 2000, Schutter & Dick 2002, Buyer et al. 2010). Fungal species in agricultural soil have functional traits including decomposing ability, plant infectivity and symbiotic ability (eg: arbuscular mycorrhizal fungi) (Wang & Qiu 2006). Many studies of manure crops or cover crops have focused on the effect of arbuscular mycorrhizal fungi related to their colonization (Marschner & Dell 1994, Lehmann et al. 2014). Furthermore, most previous studies did not fully address the total fungal community on green manure crops such as *Astragalus sinicus* and *Vicia villosa*.

Fungal endophytes are part of the microbial community, which survive inside plant tissues without causing any visible symptoms (Fröhlich et al. 2000, Ghimire & Hyde 2008, Hyde & Soyong 2008, Zabalgoceazcoa 2008, Le Cocq et al. 2016). Endophytes can support plants to obtaining nutrients, enhance the nutritional quality of crops and resist some diseases through

mechanisms such as competition, antibiosis and parasitism (Khidir et al. 2010, Porrás-Alfaro and Bayman 2011). Many endophytic fungi could also act as biocontrol agents (Kumar et al. 2017). However, still it is unclear how most of the endophytes affect plant health and its functions (Porrás-Alfaro & Bayman 2011). Endophytes may not remain as it is throughout their lifecycle (Zabalgogea 2008, Porrás-Alfaro & Bayman 2011). They can be latent pathogens or latent saprotrophs due to stress or any changes in the host or the environment (Porrás-Alfaro & Bayman 2011).

Most studies on green manure crops focused primarily on their effect on the targeted crops and the agricultural systems such as enhancing the soil properties or the emissions of CH₄ and N₂O (Parsons 1984, Bo et al. 2012). There is a possibility that endophytic species are latent or quiescent on crops. They may have pathogenic or saprobic phases upon introduction to a new agricultural field (Saikkonen et al. 1998, Arnold et al. 2000, Rodriguez et al. 2009). Alternatively, allied myco-communities can be a source of decomposers, nutrient cyclers, soil aggregators, and mycorrhizal symbionts, in the context of green manuring. Based on these hypotheses, we aimed to understand the poorly explored fungal diversity associated with *Astragalus sinicus* and *Vicia villosa* in this study, thus addressing major gaps in the understanding of green manuring practices.

Table 1 Sample site details.

Collection Site	Province	Geographical location	Annual Temperature (°C)	Annual Rainfall (mm)
Guilin City	Guangxi	E110.31; N25.07	19.1 °C	1887.6
Nanning City	Guangxi	E116.46; N39.92	21.6 °C	1304.2
Xinyang (including Luoshan City and Shihe District)	Henan	E114.08; N32.11	15.1 °C	1109.11
Fuzhou City	Fujian	E119.36; N26.08	19.6 °C	1342
Tongren City	Guizhou	E108.23; N27.52	17.1 °C	1073.2

In this study, we focused on culture-dependent and culture-independent mycobiome analyses of *Astragalus sinicus* and *Vicia villosa* with their entire habitat including all the fungi in plants and in the surrounding environments. The objectives of the present study are; (i) to investigate the endophytic fungi associated with green manure crops from China with a comprehensive sampling and to identify the cultivable fungi obtained, to genus or species levels using morpho-molecular techniques, (ii) to understand the community composition and diversity of fungal species associated with *Astragalus sinicus* and *Vicia villosa* using HTS (iii) to describe novel species with detailed descriptions and illustrations, (iv) and update host and geographical records in China and (v) to provide a worldwide checklist of fungal species associated with *Astragalus sinicus* and *Vicia villosa* based on previous and current research. Finally, we raise safety concerns about field applications of green manure crops and whether fungal endophytes in green manure crops really matter.

Materials & Methods

Sampling and fungal isolation

Astragalus sinicus and *Vicia villosa* samples were collected from six sites: in Fuzhou, Guilin, Luoshan, Nanning, Tongren, and Xinyang in four provinces (Fujian, Guangxi, Guizhou, and Henan) in China during 2017–2019 (Figure 1, Table 1). Asymptomatic flowers, leaves, pods, roots, and shoots were collected from each site (Figure 2). In total 54 samples were collected and from each sample, five plant pieces (5 mm³ sizes) were excised. Plant pieces were surface sterilized for 30 seconds in NaOCl, washed for 1 minute in sterilized, distilled water, 1 minute in 70% ethanol and washed three times in sterilized, distilled water. Once the plant pieces were dried in aseptic conditions, each of the five pieces was placed on potato dextrose agar (PDA) medium

supplemented with 100 mg/L penicillin. After incubation for several days at 25 °C, the hyphal tips of developing fungi were transferred to the PDA medium. Pure cultures were obtained via single-spore or single-hyphae isolation for further study. The isolated fungal strains are preserved in PDA slants at +4 °C in the culture collection of the Beijing Academy of Agricultural and Forestry Sciences (JZB), Beijing, China. Specimens (dried cultures) were also deposited in the fungarium of the Beijing Academy of Agricultural and Forestry Sciences (JZBH). Taxonomic descriptions for novel species were deposited in faces of fungi database (<https://www.facesoffungi.org/>; Jayasiri et al. 2015).

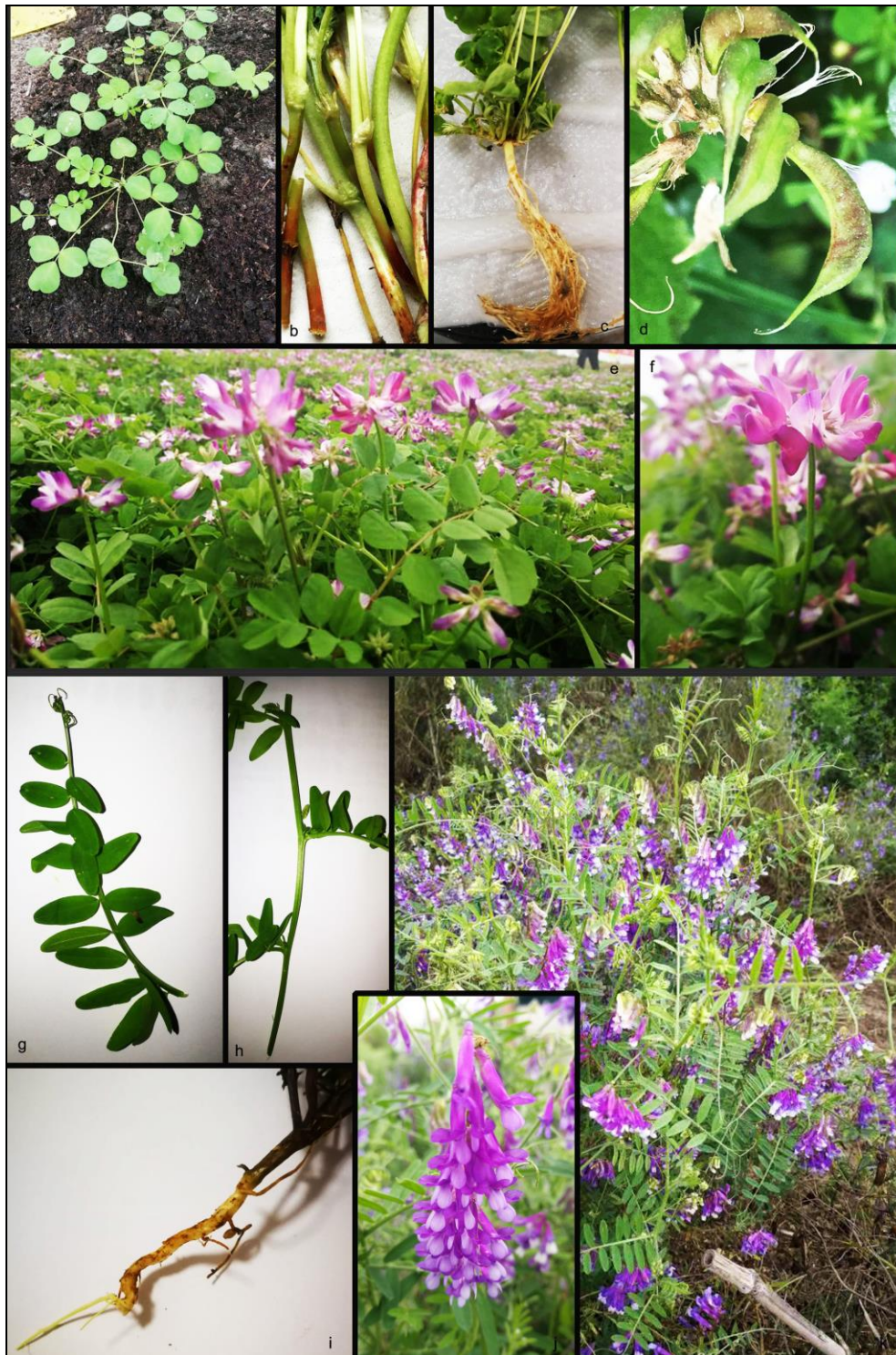


Figure 2 – Green manure crops; *Astragalus sinicus* (a–f) a Leaves. b Stems. c Roots. d Pods. e, f Flowers. *Vicia villosa* (g–k) g Leaves. h Stem. i Root. j, k Flowers.

DNA extraction, polymerase chain reaction and phylogenetic analyses

Total genomic DNA was extracted according to a modified method described below, by using CTAB (cetyltrimethylammonium bromide) extraction buffer. Fresh fungal mycelia were scraped from the colonies grown on the PDA plates, which were incubated at 25 °C for one week. Mycelia were collected into 1.5 ml microtubes and crushed with liquid nitrogen. We then added pre-heated CTAB extraction buffer [(2% CTAB 20 g, 2% PVP-40 20 g, NaCl 81.81g, 1M Tris-HCl 100 ml (PH 8), 0.5 EDTA 40 ml (PH8))] to the microtubes. The content was incubated at 65 °C in a water bath for 1 hour with random mixings. Equal volumes of (300 ml) phenol to chloroform: Isoamyl alcohol (24: 1) were added to the content and centrifuged at 10,000 rpm for 10 min (Eppendorf centrifuge 5424). The upper aqueous phase with no visible cloudy appearance was transferred to a new 1.5 ml microtube and treated with 0.6V ml (V=Total volume of newly taken upper aqueous phase) of isopropyl alcohol. The resulting content was kept precipitating the DNA at -20 °C for 1 hour. The upper layer was discarded after the content was centrifuged at 12,000 rpm for 10 min and precipitated DNA was washed twice with 70% ethanol, dried under vacuum, and re-suspended in 20–30 µl TE buffer (RNase added) (TaKaRa Products Catalog 2014–2015). The extracted DNA was stored at -20 °C until it was used for further analyses.

Polymerase chain reaction (PCR) was carried out for the obtained DNA in a total volume of 25 µl which contained 12.5 µl of 2 × Taq PCR Master-Mix (Biomed Co., China), 1 µl of each primer (forward and reversed), 1 µl genomic DNA, and 9.5 µl of deionized water. Amplified gene regions with respective primer pairs and thermal cycler reactions for each genus/family are given in Table 2. The positive amplicons identified on 1% agarose electrophoresis gels stained with ethidium bromide and visualized under UV light using Gel Doc XR + Molecular Imager Imaging system (BIO-RAD, USA). The amplified PCR fragments from the culture-dependent method were sequenced by Biomed Company, Beijing, China. The forward and reverse sequences were assembled by using Bio Edit Sequence Alignment Editor (v. 7.0.9, Hall 1999).

Sequences generated from different primers were analyzed with other sequences retrieved from GenBank. The related sequences were obtained from a BLASTn search and recently published data (<https://blast.ncbi.nlm.nih.gov/>). The sequences were aligned in the Multiple alignment program for amino acid or nucleotide sequences (MAFFT v. 7) at the webserver (<http://mafft.cbrc.jp/alignment/server>) using default settings (Kuraku et al. 2013, Katoh et al. 2017). The alignments were manually edited where necessary with Bio Edit v 7.0.9 (Hall 1999).

The phylogenetic analyses were conducted using Bayesian inference analyses (BI), performed in MrBayes v. 3.2.7a, (Huelsenbeck & Ronquist 2001, Ronquist & Huelsenbeck 2003) and Maximum Likelihood (ML) in the CIPRES Science Gateway platform. Bayesian posterior probability (BYPP) analyses (Huelsenbeck & Ronquist 2001, Ronquist & Huelsenbeck 2003) were evaluated (Rannala & Yang 1996, Zhaxybayeva & Gogarten 2002) by Markov Chain Monte Carlo sampling (MCMC). Six simultaneous Markov chains were run for at least 1,000,000 generations and trees were sampled every 100th generation. The distribution of log-likelihood scores was examined to determine the stationary phase for each search and to decide if extra runs were required to achieve convergence, using the program Tracer V 1.5 (Rambaut & Drummond 2003). All sampled topologies beneath the asymptote (10%) were discarded as part of the burn-in procedure; the remaining trees were used for calculating posterior probabilities (PP) in the majority rule consensus tree.

Maximum likelihood trees were generated using the RAxML-HPC2 on XSEDE (8.2.8) (Stamatakis 2006, 2014) in the CIPRES Science Gateway platform (Miller et al. 2010) using the GTRGAMMA model with the rapid bootstrapping and search for best-scoring ML tree algorithm including 1,000 bootstrap replicates. Phylograms were visualized with FigTree v1.4.0 (Rambaut 2014) and annotated in Microsoft PowerPoint (2007) or Adobe Illustrator CS5 (Version 15.0.0, Adobe, San Jose, CA). For the taxonomic treatments we follow Wijayawardene et al. (2020, 2022). The DNA sequence data generated in this study are deposited in GenBank (Supplementary Table 1) (<https://www.ncbi.nlm.nih.gov/genbank/>).

Table 2 Respective PCR reaction primers (forward and reverse) for amplification of genetic markers of each fungal genus and references used in the study.

Genus	Gene	Primers	PCR conditions	Reference
<i>Albifimbria</i>	LSU	LROR/LR5	(94 °C: 30 s, 50 °C: 50 s, 72 °C: 30 s) × 35 cycles	Vilgalys & Hester (1990), Rehner & Samuels (1994)
	SSU	NS1/NS4	(94 °C: 30 s, 40 °C: 50 s, 72 °C: 30 s) × 35 cycles	White et al. (1990)
	<i>tefl-α</i> <i>rpb2</i>	EF-983F/EF-2218R RPB2-5F/RPB2-7cR	(94 °C: 30 s, 55 °C: 50 s, 72 °C: 30 s) × 35 cycles (94 °C: 30 s, 56 °C: 30 s, 72 °C: 30 s) × 35 cycles	Rehner et al. (2001) Liu et al. (1999), Sung et al. (2007)
<i>Alternaria</i>	ITS	ITS 1/ITS 4	(94 °C: 30 s, 54 °C: 30 s, 72 °C: 30 s) × 35 cycles	White et al. (1990)
	GAPDH	gpd1/gpd2	(96 °C: 60 s, 58 °C: 30 s, 72 °C: 30 s) × 35cycles	Berbee et al. (1999)
	<i>rpb2</i>	RPB2-5F/RPB2-7cR	(94 °C: 30 s, 56 °C: 50 s, 72 °C: 30 s) × 35 cycles	Liu et al. (1999), Sung et al. (2007)
	<i>tefl-α</i>	EF1-728F/EF1-986R	(94 °C: 30 s, 54 °C: 30 s, 72 °C: 30 s) × 35 cycles	Carbone & Kohn (1999)
	Alt-a1	Alt-F/Alt-R	(94 °C: 60 s, 57 °C: 30 s, 72 °C: 30 s) × 35 cycles	Hong et al. (2005)
<i>Arthrinium</i>	ITS	ITS 1/ITS 4	(94 °C: 30 s, 54 °C: 30 s, 72 °C: 30 s) × 35 cycles	White et al. (1990)
	LSU	LR5/LROR	(94 °C: 30 s, 50 °C: 50 s, 72 °C: 30 s) × 35 cycles	Vilgalys & Hester (1990), Rehner & Samuels (1994)
	<i>tefl-α</i>	EF1-728F/EF2	(94 °C: 30 s, 54 °C: 30 s, 72 °C: 30 s) × 35 cycles	O'Donnell et al. (1998), Carbone & Kohn (1999)
	<i>tub</i>	T1/Bt2b	(94 °C: 30 s, 53 °C: 30 s, 72 °C: 30 s) × 35 cycles	Glass & Donaldson (1995), O'Donnell & Cigelnik (1997)
<i>Botrytis</i>	<i>rpb2</i>	RPB2 F/RPB2 R	(94 °C: 30 s, 54 °C: 50 s, 72 °C: 30 s) × 35 cycles	Staats et al. (2005)
	HSP60	HSP60 F/HSP60 R	(94 °C: 30 s, 59 °C: 50 s, 72 °C: 30 s) × 35 cycles	Staats et al. (2005)
	GAPDH	G3PDH F/G3PDH R	(94 °C: 30 s, 58 °C: 50 s, 72 °C: 30 s) × 35 cycles	Staats et al. (2005)
<i>Clonostachys</i>	ACL	Acl-1230up/Acl-11220low	(94 °C: 30 s, 57 °C: 30 s, 72 °C: 30 s) × 35 cycles	Gräfenhan et al. (2011)
	<i>tub</i>	T1/T2	(94 °C: 30 s, 52 °C: 30 s, 72 °C: 30 s) × 35 cycles	O'Donnell & Cigelnik (1997)
	<i>rpb1</i>	RPB1-Fa/RPB1-R8	(94 °C: 30 s, 57 °C: 50 s, 72 °C: 30 s) × 35 cycles	O'Donnell et al. (2010)
	<i>tefl-α</i>	EF1-728F/EF2	(94 °C: 30 s, 54 °C: 30 s, 72 °C: 30 s) × 35 cycles	O'Donnell et al. (1998), Carbone & Kohn (1999)
<i>Colletotrichum</i>	ITS	ITS 1/ITS 4	(94 °C: 30 s, 54 °C: 30 s, 72 °C: 30 s) × 35 cycles	White et al. (1990)
	GAPDH	GDF/GDR	(94 °C: 30 s, 62 °C: 30 s, 72 °C: 30 s) × 35 cycles	Templeton et al. (1992)
	CHS-1	CHS-79F/CHS-354R	(94 °C: 30 s, 59 °C: 30 s, 72 °C: 30 s) × 35 cycles	Carbone & Kohn (1999)
	ACT	ACT-512F	(94 °C: 30 s, 58 °C: 30 s, 72 °C: 30 s) × 35 cycles	Carbone & Kohn (1999)
	<i>tub</i>	BT 2F/BT 4R	(94 °C: 30 s, 56 °C: 30 s, 72 °C: 30 s) × 35 cycles	O'Donnell & Cigelnik (1997)
<i>Diaporthe</i>	ITS	ITS 1/ITS 4	(94 °C: 30 s, 54 °C: 30 s, 72 °C: 30 s) × 35 cycles	White et al. (1999)
	CAL	CAL228F/CAL737R	(94 °C: 30 s, 57 °C: 30 s, 72 °C: 30 s) × 35 cycles	Carbone & Kohn (1999)
	HIS	CYLH3F/CYLH3R	(94 °C: 30 s, 57 °C: 30 s, 72 °C: 30 s) × 35 cycles	Crous et al. (2004)

Table 2 Continued.

Genus	Gene	Primers	PCR conditions	Reference
	<i>tefl-α</i>	EF1-728F/EF1-986R	(94 °C: 30 s, 54 °C: 30 s, 72 °C: 30 s) × 35 cycles	Carbone & Kohn (1999)
	<i>tub</i>	BT2a/BT2b	(94 °C: 30 s, 52 °C: 30 s, 72 °C: 30 s) × 35 cycles	Glass & Donaldson (1995)
<i>Epicoccum</i>	LSU	LR5/LROR	(94 °C: 30 s, 50 °C: 50 s, 72 °C: 30 s) × 35 cycles	Vilgalys & Hester (1990), Rehner & Samuels (1994)
	ITS	ITS 1/ITS 4	(94 °C: 30 s, 54 °C: 30 s, 72 °C: 30 s) × 35 cycles	White et al. (1990)
	<i>rpb2</i>	RPB2–5F/RPB2–7cR	(94 °C: 30 s, 56 °C: 50 s, 72 °C: 30 s) × 35 cycles	Liu et al. (1999), Sung et al. (2007)
	<i>tub</i>	BT-2F/BT-4R	(94 °C: 30 s, 56 °C: 30 s, 72 °C: 30 s) × 35 cycles	O'Donnell & Cigelnik (1997)
<i>Fusarium</i>	<i>tefl-α</i>	EF1/EF2	(94 °C: 30 s, 52 °C: 30 s, 72 °C: 30 s) × 35 cycles	O'Donnell et al. (1998)
	ITS	ITS 1/ITS 4	(94 °C: 30 s, 54 °C: 30 s, 72 °C: 30 s) × 35 cycles	White et al. (1990)
	<i>rpb2</i>	RPB2–5F/RPB2–7cR	(94 °C: 30 s, 56 °C: 50 s, 72 °C: 30 s) × 35 cycles	Liu et al. (1999), Sung et al. (2007)
<i>Lasiodiplodia</i>	ITS	ITS 1/ITS 4	(94 °C: 30 s, 54 °C: 30 s, 72 °C: 30 s) × 35 cycles	White et al. (1990)
	<i>tefl-α</i>	EF1-728F/EF1-986R	(94 °C: 30 s, 54 °C: 30 s, 72 °C: 30 s) × 35 cycles	Carbone & Kohn (1999)
<i>Leptosphaerulina</i>	LSU	LR5/LROR	(94 °C: 30 s, 50 °C: 50 s, 72 °C: 30 s) × 35 cycles	Vilgalys & Hester (1990), Rehner & Samuels (1994)
	ITS	ITS 1/ITS 4	(94 °C: 30 s, 54 °C: 30 s, 72 °C: 30 s) × 35 cycles	White et al. (1990)
	<i>rpb2</i>	RPB2–5F/RPB2–7cR	(94 °C: 30 s, 56 °C: 50 s, 72 °C: 30 s) × 35 cycles	Liu et al. (1999), Sung et al. (2007)
<i>Neofusicoccum</i>	ITS	ITS 1/ITS 4	(94 °C: 30 s, 54 °C: 30 s, 72 °C: 30 s) × 35 cycles	White et al. (1990)
	<i>tefl-α</i>	EF-728F/EF-986R	(94 °C: 30 s, 54 °C: 30 s, 72 °C: 30 s) × 35 cycles	Carbone & Kohn (1999)
	<i>tub</i>	BT-2b/BT-2a	(94 °C: 30 s, 52 °C: 30 s, 72 °C: 30 s) × 35 cycles	Glass & Donaldson (1995)
<i>Plectosphaerella</i>	LSU	LROR/LR5	(94 °C: 30 s, 50 °C: 50 s, 72 °C: 30 s) × 35 cycles	Vilgalys & Hester (1990), Rehner & Samuels (1994)
	ITS	ITS 1/ITS 4	(94 °C: 30 s, 54 °C: 30 s, 72 °C: 30 s) × 35 cycles	White et al. (1990)
<i>Pseudopithomyces</i>	LSU	LROR/LR5	(94 °C: 30 s, 50 °C: 50 s, 72 °C: 30 s) × 35 cycles	Vilgalys & Hester (1990), Rehner & Samuels (1994)
	SSU	NS1/NS4	(94 °C: 30 s, 54 °C: 50 s, 72 °C: 30 s) × 35 cycles	White et al. (1990)
	ITS	ITS 1/ITS 4	(94 °C: 30 s, 54 °C: 30 s, 72 °C: 30 s) × 35 cycles	White et al. (1990)
	<i>tefl-α</i>	EF1-983F/EF1-2218R	(94 °C: 30 s, 55 °C: 50 s, 72 °C: 30 s) × 35 cycles	Rehner et al. (2001)
<i>Sclerotinia</i>	CAL	CAL-228F/CAL-737R	(94 °C: 30 s, 57 °C: 30 s, 72 °C: 30 s) × 35 cycles	Carbone & Kohn (1999)
	ITS	ITS 1/ITS 4	(94 °C: 30 s, 54 °C: 30 s, 72 °C: 30 s) × 35 cycles	White et al. (1990)
	MCM	Mcm7-709F/Mcm7-1048R	(94 °C: 30 s, 60 °C: 30 s, 72 °C: 30 s) × 35 cycles	Schmitt et al. (2009)
<i>Stemphylium</i>	ITS	ITS 1/ITS 4	(94 °C: 30 s, 54 °C: 30 s, 72 °C: 30 s) × 35 cycles	White et al. (1990)
	GAPDH	gpd1/gpd2	(96 °C: 60 s, 58 °C: 30 s, 72 °C: 30 s) × 35cycles	Berbee et al. (1999)
	CAL	CAL-228F/CAL-737R	(94 °C: 30 s, 57 °C: 30 s, 72 °C: 30 s) × 35 cycles	Carbone & Kohn (1999)

Identification of *Fusarium* isolates

All isolated *Fusarium* strains were identified to genus or species level, based on a comparison of their internal transcribed spacer region (ITS), translation elongation factor 1- α gene (*tef1-a*) and RNA polymerase II gene (*rpb2*) sequences. For generic and species determination of the isolates, BLASTn searches were performed on the NCBI GenBank (<https://blast.ncbi.nlm.nih.gov/>) and *Fusarium* MLST (<https://fusarium.mycobank.org/>) databases. The strains were identified to species, genus, or higher level, depending on the affinity to the available reference sequences (Supplementary Table 2).

The mycobiome analysis

Sampling, library preparation and statistical and diversity analysis

Fresh plant specimens of *Astragalus sinicus* and *Vicia villosa* were collected from four provinces in China (Fujian, Guangxi, Guizhou, and Henan). *Astragalus sinicus* specimens were collected from all four provinces. However, *Vicia villosa* specimens were only collected from the Henan and Guangxi provinces (Figure 1, Table 1). For each crop, six representative plant individuals were sampled and homogenized in each province. Total genomic DNA was extracted using 1g of ground specimens using the 2 \times CTAB method. The extracted DNA was quantified, and quality was checked with the NanoDrop ND-2000C spectrophotometer (Thermo Fisher Scientific, Dreieich, Germany). Extracted DNA was kept at -20°C for further analysis.

For HTS, we used the 18S rRNA V4 region of the ribosomal RNA gene cluster. This region was amplified with the forward primer 528F (GTGCCAGCMGCCGCGGTAA) and reverse primer 706R (GGACTACHVGGGTWTCTAAT) (Cheung et al. 2010). The PCR reaction was performed in a 50 μl volume that contained approximately 10 mg of DNA, Ex Taqbuffer, 0.2mM of dNTPs, 0.2mM of each primer, and 2 units of ExTaq DNA polymerase. The cycling consisted of an initial denaturing step at 94°C for 30 sec., followed by 25 cycles of denaturing at 94°C for 30 sec., annealing at 54°C for 1 min, extension at 72°C for 2 min, and a final extension at 72°C for 8 min. All PCR reactions were carried out with Phusion[®] High-Fidelity PCR Master Mix (New England Bio Labs Inc. Ipswich, MA, USA). The PCR products were mixed with the same volume of 1 \times loading buffer (contained SYB green) and then run on a 2% agarose gel for quality detection. Only samples with a bright main strip between 400–450 bp were chosen for further experiments.

The PCR products were purified using Qiagen Gel Extraction Kit (Qiagen, Germany) following the manufacturer's protocol. Sequencing libraries were generated using Ion plus Fragment Library Kit 48 rxns (Massachusetts, USA) following the manufacturer's recommendations. The library quality was assessed on the Qubit[®] 2.0 Fluorometer (Thermo Scientific) and Agilent Bioanalyzer 2100 system. The library was sequenced on an Ion S5[™] XL platform and single-end reads were generated.

Low-quality reads were assigned to samples based on their unique barcode, truncated by removing the barcode and primer sequence and then quality filtered to obtain the high-quality clean reads using Cutadapt by the parameters of `-overlap 10 -q 17 -m 450 -M 550` (V1.9.1, <http://cutadapt.readthedocs.io/en/stable/>). The raw sequences were aligned to the SILVA 132 reference database (<http://www.arb-silva.de/>, Quast et al. 2013) using the LCA algorithm to detect chimeric sequences (Edgar et al. 2011). The chimeric sequences were removed using VSEARCH 2.8.1 (Torbjörn Rognes et al. 2016) and clean reads were obtained for further analysis. Sequences analysis was performed in Uparse v. 7.0.1001 (<http://drive5.com/uparse/>; Edgar 2013). Sequences with 97% similarity were assigned to the same operational taxonomic units (OTUs). The representative sequence for each OTU was examined for taxonomic affiliation using SILVA 132 reference database based on the RDP classifier v.2.2 (<http://rdp.cme.msu.edu/>; Wang et al. 2007). To study the phylogenetic relationship of different OTUs, and the difference of the dominant species in different samples (groups), multiple sequence alignment was conducted using the MUSCLE software (Version 3.8.31, <http://www.drive5.com/muscle/>) (Edgar 2004).

All OTU abundance information was normalized using a standard of sequence number corresponding to the sample with the least sequences. Subsequent analysis of alpha diversity (observed OTU and Shannon) and beta diversity were all performed based on this output normalized data. All the alpha diversity index values and beta diversity were calculated by QIIME software.

Fungal OTUs shared between different samples were illustrated by the VENNY 2.0 online tool (<https://bioinfogp.cnb.csic.es/tools/venny/index2.0.2.html>). Wilcoxon test was used to determine whether sample classifications (e.g., crops sampling locations) contained statistically significant differences in the alpha diversities. Principal coordinate analysis (PCoA) and NMDS (Non-metric Multidimensional Scaling) were performed to evaluate the distribution patterns of mycobiome based on beta-diversity calculated by the Bray–Curtis distance with the ‘vegan’ and ‘WGCNA’ packages. Permutational multivariate analysis of variance (PERMANOVA) based on Bray-Curtis distance matrices was conducted within each sample category to determine the statistically significant differences by ‘vegan’ package. Significant taxonomic differences of fungi between different habitats were tested using linear discriminant analysis (LDA) and effect size (LEfSe) analysis (Segata et al. 2011) (<https://huttenhower.sph.harvard.edu/galaxy/>). Discriminating species between different groups were obtained by SIMPER analysis based on Bray-Curtis dissimilarities using the ‘vegan’ package. The co-occurrence network was explored using network analysis with ‘igraph’ package. Correlations with a Spearman correlation coefficient $\rho \geq 0.6$ and a $P < 0.05$ were considered statistically robust and displayed in the networks by graphviz-2.38. Functional properties were annotated using both FUNGuild (Nguyen et al. 2016) and Fungaltraits databases (Pölme et al. 2020).

The fungal 18S rDNA gene Illumina sequencing data are deposited in the NCBI under the BioProject number: PRJNA813628.

Compiling the checklist

The checklist is based on articles in referred journals and web-based resources such as the systematic mycology and microbiology laboratory nomenclature database (SMML) (<https://nt.ars-grin.gov/fungalatabases/>) (latest accessed 30-1-2022). The checklist includes fungal species names, families, and localities for both green manure crops. The current name is used according to Index Fungorum (2022) and Wijayawardene et al. (2020) and the classification follows Wijayawardene et al. (2020, 2022). Genera and species are listed in alphabetical order (Supplementary Table 3).

Results

Diversity and abundance of culturable fungi

In total, 517 fungal strains were isolated from *Astragalus sinicus* and *Vicia villosa* plants, which belong to 15 genera. Among them, 381 strains belonged to *Fusarium*. Inferred multi-gene phylogenies identified the remaining 136 strains which were to species level in 14 genera in ten families. The number of strains isolated per host species was as follows: 307 isolates from *Astragalus sinicus* and 210 isolates from *Vicia villosa*.

Species belonging to *Arthrinium*, *Botrytis*, *Leptosphaerulina*, *Pseudopithomyces*, *Myrothecium*, *Stemphylium*, *Sclerotinia*, *Lasiodiplodia*, *Neofusicoccum*, and *Plectosphaerella* were isolated from one of the hosts, while *Alternaria*, *Colletorichum*, *Diaporthe*, *Epicoccum* and *Fusarium* were associated with both plants. *Alternaria* and *Fusarium* species were isolated from both crops collected in Guangxi and Guizhou provinces. We were only able to collect samples from *Astragalus sinicus* from the provinces of Fujian and Henan. However, *Fusarium* species were isolated in both green manure crops and in all sampling areas.

Astragalus sinicus had higher species richness (307 strains) than *Vicia villosa* (210 strains). All obtained cultivable fungi were ascomycetes. From the identified isolates, 78% were Sordariomycetes, 15% were Dothideomycetes, and 7% were Leotiomycetes. The identified

Sordariomycetes belonged to *Nectriaceae* (94.5%), *Glomerellaceae* (3%) and other families (2.5%) (*Apiosporaceae*, *Diaporthaceae*, *Plectosphaerellaceae*, *Bionectriaceae*, and *Stachybotryaceae*). Identified Dothideomycetes belong to *Didymellaceae* (48%), *Pleosporaceae* (45.5%) and other families (6.5 %) (*Botryosphaeriaceae* and *Didymosphaeriaceae*).

Separate multi-loci phylogenetic analyses (based on the genus or the family that they belong to) were performed for the strains isolated from the culture-dependent approach.

Taxonomy

The numbers of taxa in this study are organised following Wijayawardene et al. (2020, 2022) and updated from recent relevant literature. For the delineation of novel ascomycetous fungal species, we follow the guidelines from Jayawardena et al. (2021), Maharachchikumbura et al. (2021) and Manawasinghe et al. (2021). Descriptions and photo plates were provided for the novel species derived from this study.

Ascomycota R.H. Whittaker (1959).

Dothideomycetes O.E. Erikss. & Winka (1997).

Dothideomycetidae P.M. Kirk, P.F. Cannon, J.C. David & Stalpers (2001).

Botryosphaeriales C.L. Schoch, Crous & Shoemaker (2007).

Botryosphaeriaceae Theiss. & Syd. (1918).

Lasiodiplodia Ellis & Everh. (1896).

Lasiodiplodia mediterranea Linald., Deidda & Berraf-Tebbal (2015).

For description, see Linaldeddu et al. (2015).

Material examined – China, Guangxi Province, Nanning City, from *Vicia villosa* leaves, May 2018, Zhao Wensheng and Zhang Guozhen, living cultures = JZB 3130012, JZB 3130013.

Notes – Two isolates (JZB 3130012, JZB 3130013) were recovered from *Vicia villosa* leaves in Guangxi Province. These new isolates share a close phylogenetic affinity to *Lasiodiplodia mediterranea* (BL 1) in our combined ITS, *tef1-α* and *tub* sequence analyses with 80% ML support (Fig. 3). We compared the morphological characters together with phylogenetic placement of the isolates and identified them as *L. mediterranea*. Linaldeddu et al. (2015) introduced pathogenic *L. mediterranea* from the symptomatic grapevine and other few woody hosts; holm oak (*Quercus ilex*) and sweet orange (*Citrus sinensis*) in Algeria and Italy. This species is associated with “Botryosphaeria dieback” of grapevine in Italy (Linaldeddu et al. 2015). According to the Farr and Rossman (2022), *L. mediterranea* have been reported on *Citrus sinensis* (Algeria), *Quercus ilex* (Italy), *Vaccinium corymbosum* (United States), *Vitis* spp. (United States) and *Vitis vinifera* (Italy).

We could not find any records of *Lasiodiplodia* species from *Vicia villosa* in China or in other parts of the world (Farr & Rossman 2022). Therefore, we provided the first host association of *L. mediterranea* with *Vicia villosa* in China, as well as worldwide.

Neofusicoccum Crous, Slippers & A.J.L. Phillips (2006).

Neofusicoccum parvum (Pennycook and Samuels) Crous, Slippers & A.J.L. Phillips (2006).

For description, see Crous et al. (2006).

Material examined – China, Henan Province, Shihe District, from *Vicia villosa* root, May 2018, Zhao Wensheng, and Zhang Guozhen, living culture JZB 3120007.

Notes – An isolate (JZB 3120007) was recovered from a healthy *Vicia villosa* root from Henan Province. This isolate fits well into the species concept of *Neofusicoccum*. Multi-marker analysis for *Neofusicoccum* using ITS region, *tef1-α*, *tub* and *rpb2* genes, showed that our isolate clustered within other *N. parvum* isolates (Fig. 4). Isolate JZB 3120007 had similar sized conidia as *N. parvum*. Further, JZB 3120007 isolate showed 98.24%, 91.08% and 99.01% base-pair similarities with the ex-type of *N. parvum* (CMW 9081) in ITS, *tef1-α* and *tub* genes, respectively. *Neofusicoccum* is considered as one of the most species-rich genera in *Botryosphaeriaceae*, and

most of the species share similar morphological characters (Lopes et al. 2016). Even though species of *Neofusicoccum* have a wide host range in terrestrial habitats, we could not find any *Neofusicoccum* species that have reported as associated with *Vicia villosa* in China or the world (Farr & Rossman 2022). Therefore, we also provide the first host association of *Neofusicoccum* species on *Vicia villosa* from this study.

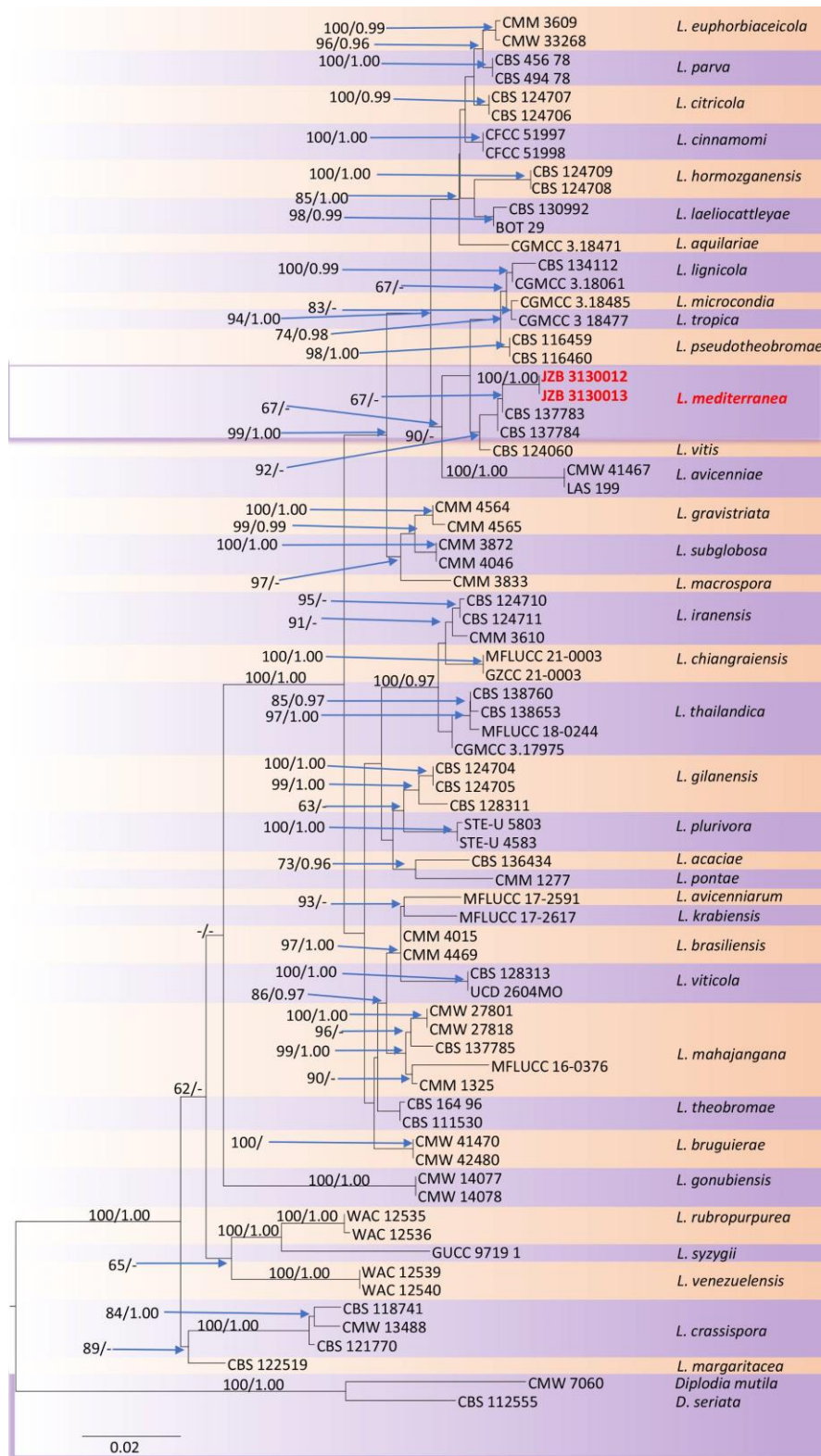


Figure 3 – Phylogram generated from maximum likelihood analysis based on combined ITS, *tef1-α* and *tub* sequence data. The matrix had 411 distinct alignment patterns, with 17.09% undetermined characters or gaps. Estimated base frequencies were as follows: A = 0.207397, C = 0.304033, G =

0.257071, T = 0.231499; substitution rates AC = 0.920212, AG = 3.393256, AT = 1.029814, CG = 0.920111, CT = 4.356229, GT = 1.000000; gamma distribution shape parameter $\alpha = 0.743135$. Bootstrap values for maximum likelihood equal to or greater than 60% and Bayesian posterior probabilities equal or greater than 0.95 are placed above the branches. The newly generated sequences are indicated in red.

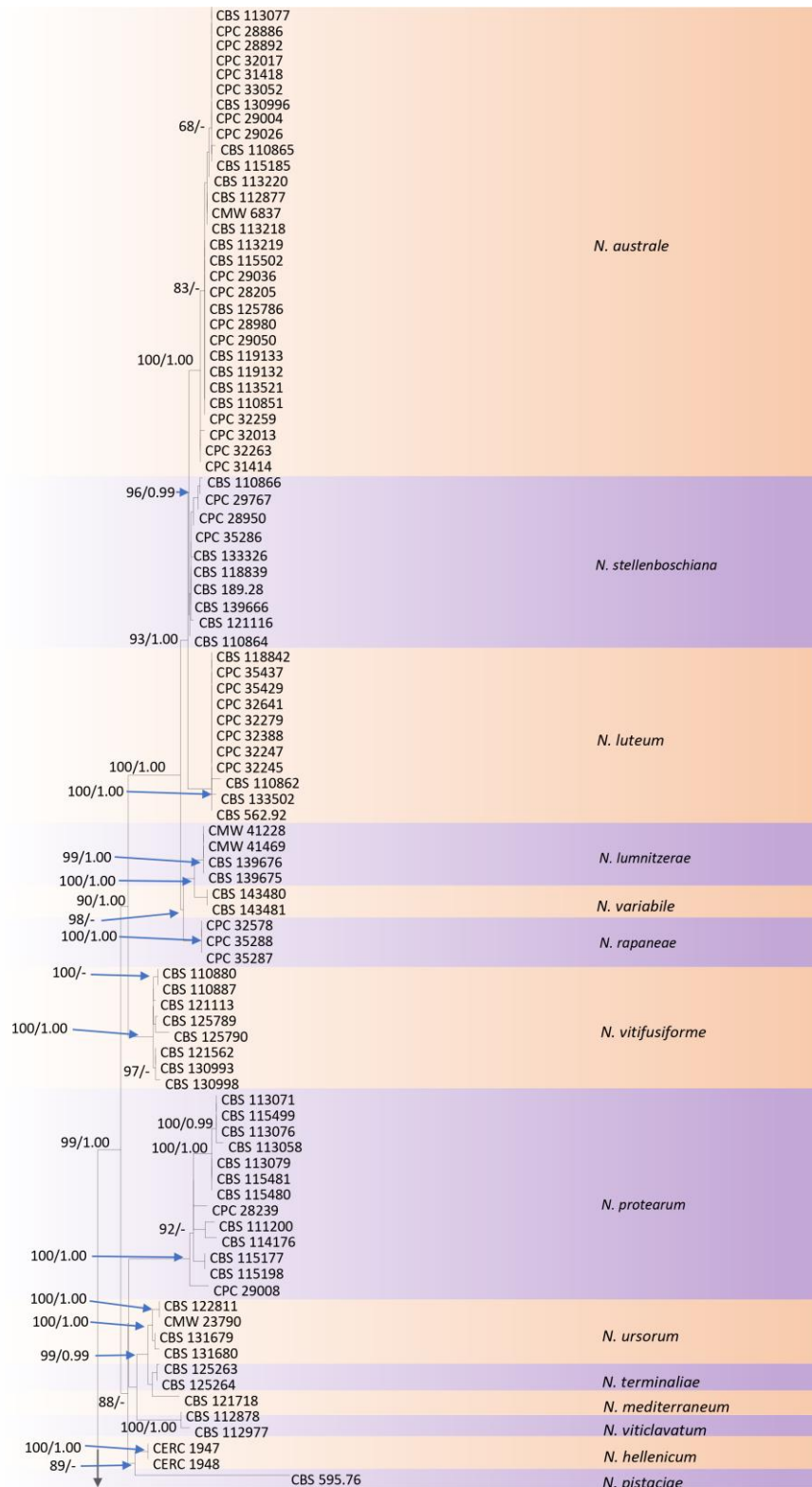


Figure 4 – Phylogram generated from maximum likelihood analysis based on combined ITS, *tefl- α* , *tub* and *rpb2* sequence data. The matrix had 501 distinct alignment patterns, with 15% undetermined characters or gaps. Estimated base frequencies were as follows: A = 0.197, C =

0.325, G = 0.262, T = 0.215; substitution rates AC = 1.00000, AG = 4.02459, AT = 1.00000, CG = 1.00000, CT = 6.97200, GT = 1.000000; gamma distribution shape parameter $\alpha = 0.820$. Bootstrap values for maximum likelihood equal to or greater than 60% and Bayesian posterior probabilities equal or greater than 0.95 are placed above the branches. The newly generated sequences are indicated in red.

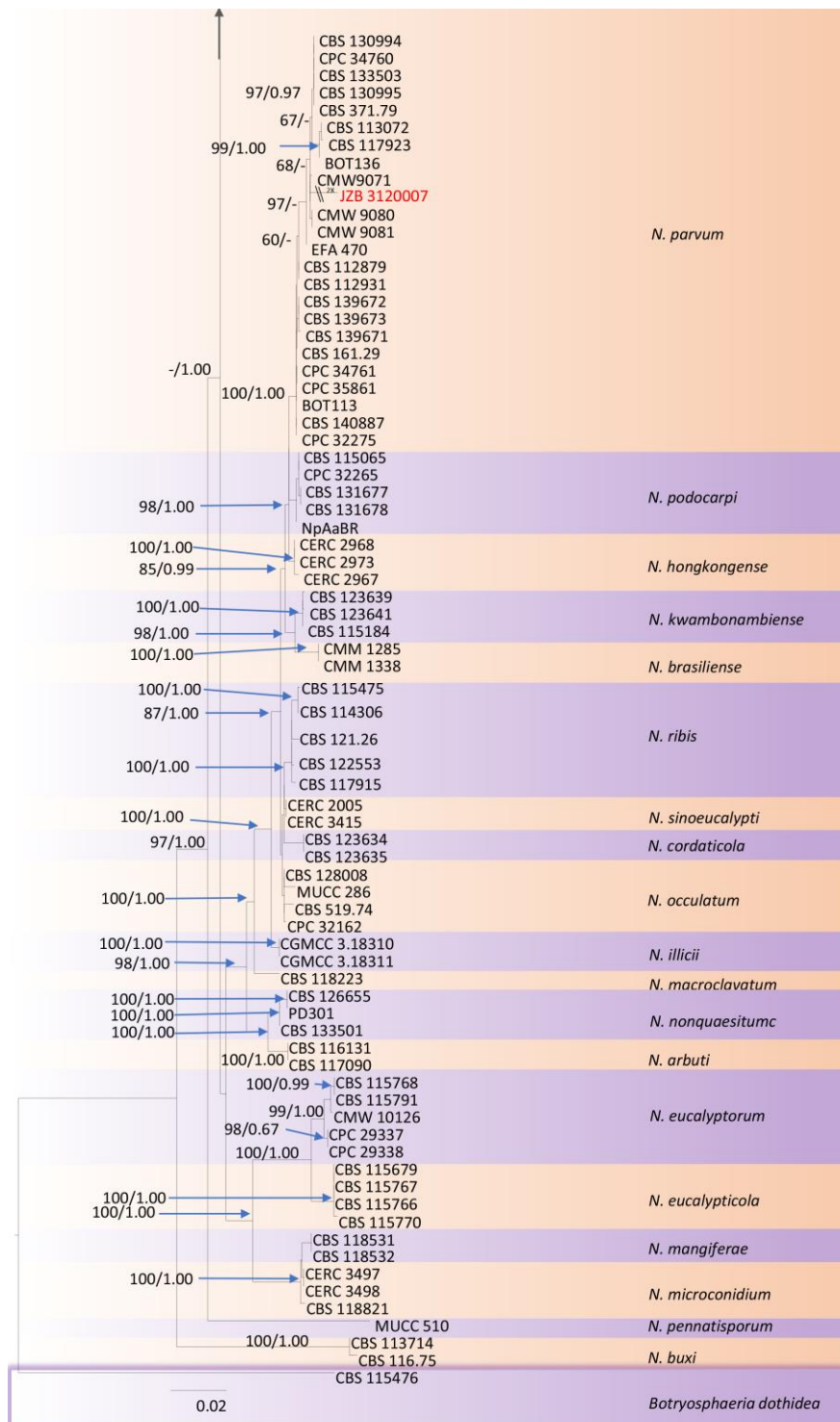


Figure 4 – Continued.

Pleosporales Luttr. ex M.E. Barr (1987).
Didymellaceae Gruyter, Aveskamp & Verkley (2009).

Epicoccum Link (1816).

Epicoccum astragali W. Zhao, Q. Ning, & J.Y. Yan, sp. nov.

Index Fungorum Number: IF 558420; Facesoffungi number: FoF 10792

Etymology – ‘astragali’ refers to the host plant genus *Astragalus* which was isolated.

Ecology – Associated with healthy leaves of *Astragalus sinicus*

Holotype – JZBH 380085

Fig. 6

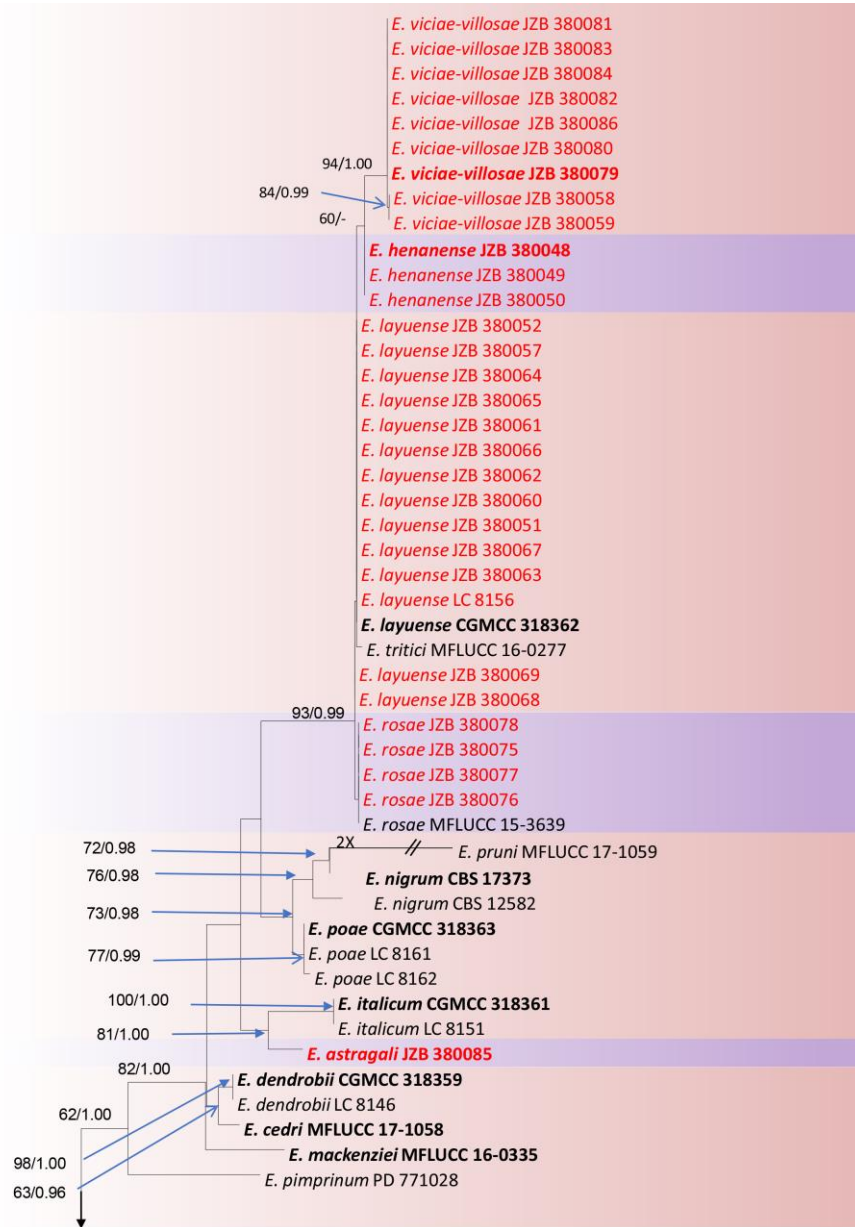


Figure 5 – Phylogram generated from maximum likelihood analysis based on combined LSU, ITS, *rpb2* and *tefl- α* sequence data. The matrix had 483 distinct alignment patterns, with 6.60% undetermined characters or gaps. Estimated base frequencies were as follows: A = 0.235206, C = 0.249893, G = 0.275404, T = 0.239497; substitution rates AC = 1.764991, AG = 5.670728, AT = 1.846180, CG = 1.160676, CT = 13.189586, GT = 1.000000; gamma distribution shape parameter α = 0.518933. Bootstrap values for maximum likelihood and maximum parsimony equal to or greater than 60% and Bayesian posterior probabilities equal or greater than 0.90 are placed above the branches. The newly generated sequences are indicated in blue. Type and ex-type strains are in bold. We obtained 35 *Epicoccum* strains and among them, 15 strains were identified as *E. layuense* that are new records on both *Astragalus sinicus* and *Vicia villosa*, five strains were identified as *E. laticollum*, and four were identified as *E. rosae* that are new records on *Astragalus sinicus* in

China. Further, *Epicoccum astragali* sp. nov., *Epicoccum henanense* sp. nov., and *Epicoccum viciae-villosae* sp. nov. are described herein.

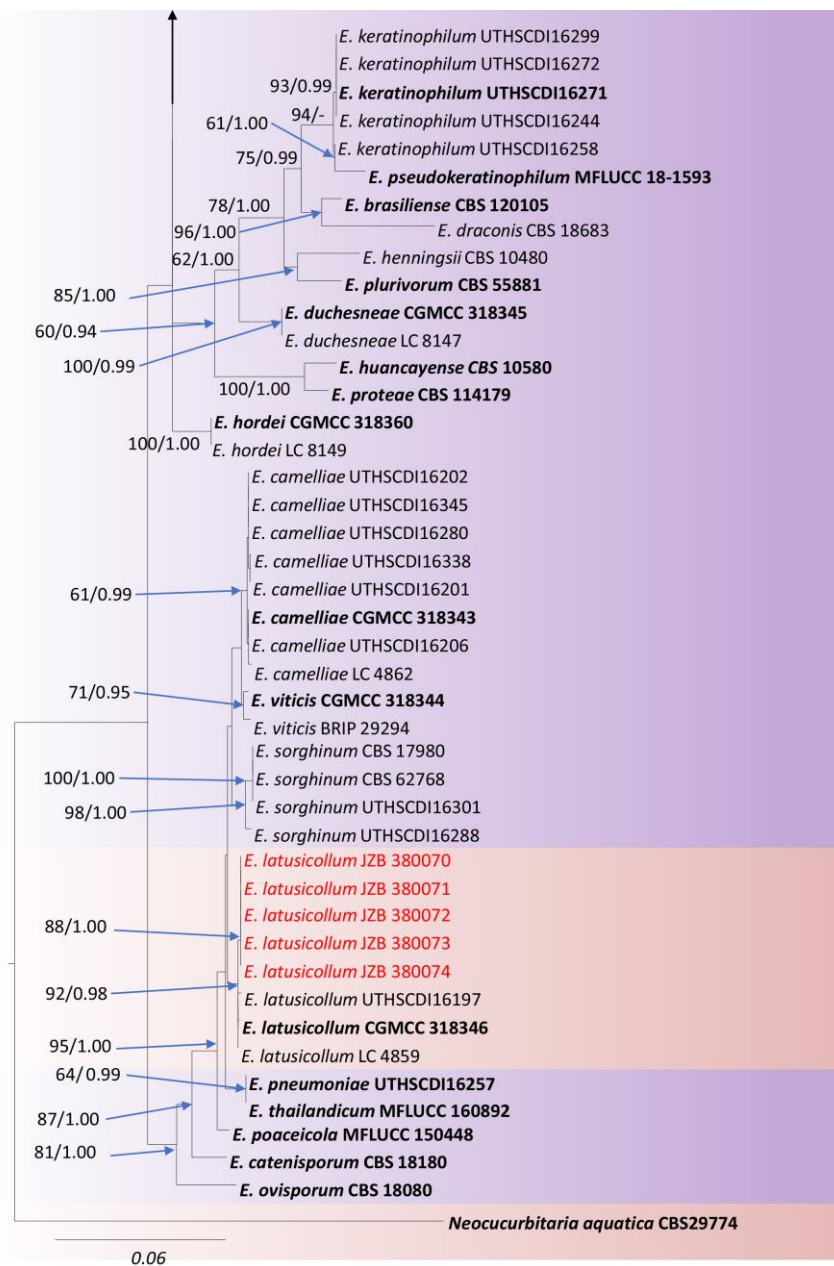


Figure 5 – Continued.

Asexual morph: Conidiomata sporodochial, solitary or aggregated, immersed to semi-immersed, glabrous, covered with hyphal growth, blackish brown. *Hyphae* smooth, branched, septate, hyaline. *Chlamydospores* multicellular, produced in agar, hyaline to pale brown, subglobose or oblong. *Conidia* multicellular, pale brown to dark brown, globose to subglobose, 8–15×7–11 μm (\bar{x} = 11.0×8.6 μm, n = 50). *Sexual morph*: not observed.

Cultural characteristics – Colonies on PDA are slow growing, covering a 30 mm Petri dish in 10 days after incubation at 25 ± 1 °C, white (surface) and yellow brown (reverse), with a dense mat of mycelium, rough, later give yellowish-brown to red colour to the PDA media.

Material examined – China, Henan Province, Luoshan City, from *Astragalus sinicus* leaves, May 2018, Zhao Wensheng and Zhang Guozhen (JZBH 380085, holotype inactive dry culture), ex-type living culture = JZB 380085.

Notes – The new strain fits well into the generic concept of *Epicoccum* in *Didymellaceae*. *Epicoccum astragali* is described herein as a new species based on multi-gene analysis of LSU, ITS, *rpb2*, and *tub* markers. The phylogenetic tree shows a moderately supported sister-clade relationship (Fig. 5) with *E. italicum* (CGMCC 318361, LC8151) (81% ML support). *Epicoccum astragali* differs from *E. italicum* by having smaller conidia ($11 \times 8.6 \mu\text{m}$ in *E. astragali* compared to $12.5\text{--}28$ diam. in *E. italicum*). Furthermore, *E. italicum* produces conidia with a basal cell, which we could not observe in *E. astragali*. *Epicoccum* species have been recorded in many hosts, and Farr and Rossman (2022) indicated that *Epicoccum nigrum* was recorded on *Astragalus sinicus* from China (Tai 1979). The phylogenetic placement of these isolates is shown in Fig. 5.

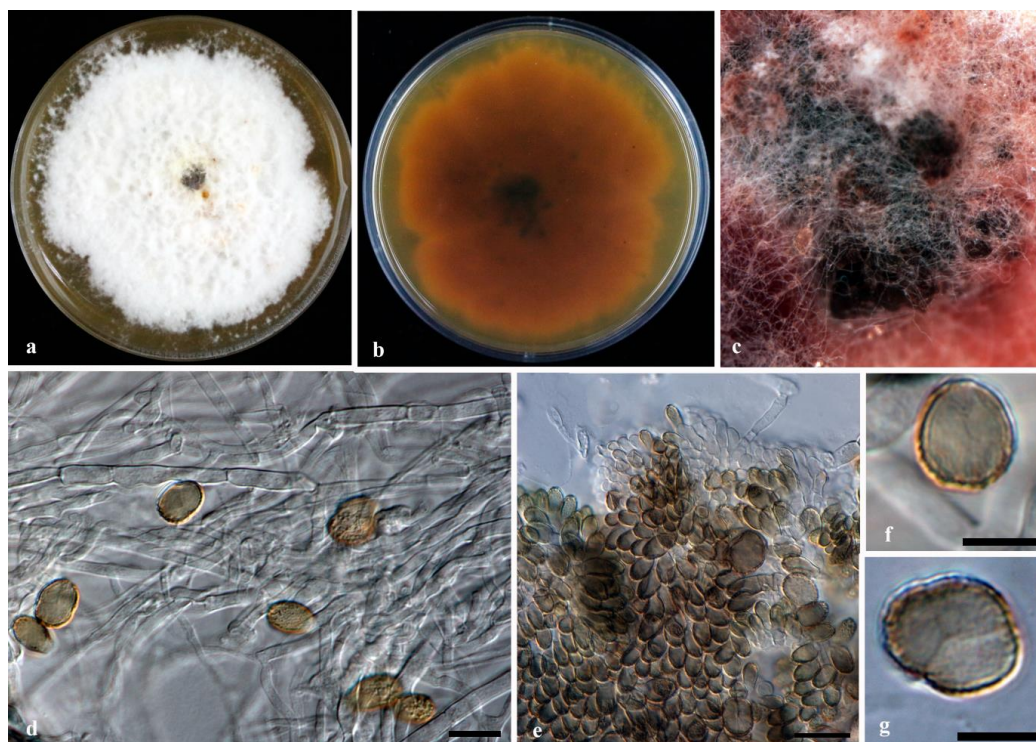


Figure 6 – *Epicoccum astragali*. a, b Colony on PDA, 10 days after incubation at $25 \pm 1 \text{ }^\circ\text{C}$ (a from above, b from below). c Pycnidia on PDA medium. d Sporodochia. e-g Conidia. Scale bars: d-g = $10 \mu\text{m}$.

Epicoccum henanense W. Zhao, Q. Ning, & J.Y. Yan, sp. nov.

Fig. 7

Index Fungorum Number: IF558420; Facesoffungi number: FoF 10793

Etymology – ‘henanense’ refers to the Henan province in China from which it was isolated.

Ecology – Associated with healthy pods of *Astragalus sinicus*

Holotype – JZBH 380048

Asexual morph: *Conidiomata* sporodochial, solitary or aggregated, immersed to semi-immersed, glabrous, covered with hyphal growth, blackish brown. *Hyphae* smooth, branched, septate, hyaline. *Chlamydo-spores* multicellular, produced in agar, hyaline to pale brown, subglobose or oblong, $5\text{--}11 \times 5\text{--}9 \mu\text{m}$ ($\bar{x} = 7.5 \times 7.3 \mu\text{m}$, $n = 20$). *Conidia*, multicellular, pale brown to dark brown, globose to subglobose, $18\text{--}30 \times 13\text{--}26 \mu\text{m}$ ($\bar{x} = 21.8 \times 18.7 \mu\text{m}$, $n = 50$). **Sexual morph:** not observed.

Cultural characteristics – Colonies on PDA are slow-growing, covering a 30 mm Petri dish in 7 days after incubation at $25 \pm 1 \text{ }^\circ\text{C}$, yellowish, red-white (surface) and reddish-black (reverse), with a dense mat of aerial mycelium, rough, entire slightly radiating at the margin; colony from above, rough, white to reddish at the fruiting zone, slimy reddish spore mass at the centre, whitish, pale yellow at productive and yellowish at ageing zone. Later give reddish-brown colour to the PDA media.

Material examined – China, Henan Province, Shihe District, from *Astragalus sinicus* pod, May 2018, Zhao Wensheng and Zhang Guozhen (JZBH 380048, holotype inactive dry culture), ex-type living culture = JZB 380048.

Additional material examined – China, Henan Province, Shihe District, from *Vicia villosa* flower, May 2018, Zhao Wensheng and Zhang Guozhen, living culture = JZB 380049, Henan province, Shihe District, from *Astragalus sinicus* pod, May 2018, Zhao Wensheng and Zhang Guozhen, living culture = JZB 380050.

Notes – The new strains fit well with the concept of *Epicoccum* in *Didymellaceae*. In multi-gene analysis using LSU, ITS, *rpb2*, and *tub* markers *Epicoccum henanense* is sister to *E. layuense* with 60% ML support (Fig. 5). *Epicoccum henanense* differs in having relatively larger conidia than *E. layuense* (18–30×13–26 μm vs 13–19.5 μm diam.). Further *E. henanense* produces globose to subglobose conidia, while *E. layuense* has subglobose-pyriform conidia. In this study, this new species was isolated from both *Astragalus sinicus* and *Vicia villosa*. So far *Epicoccum nigrum*, was recorded on *Vicia villosa* from Oregon (Shaw 1973, Farr & Rossman 2022). In China, there are two records of *Epicoccum nigrum* on *Vicia* species (Farr & Rossman 2022, Zhuang 2005). Therefore, we herein provide the first record of *Epicoccum* sp. on *Vicia villosa* in China. The phylogenetic placement of these isolates is shown in Fig. 5.

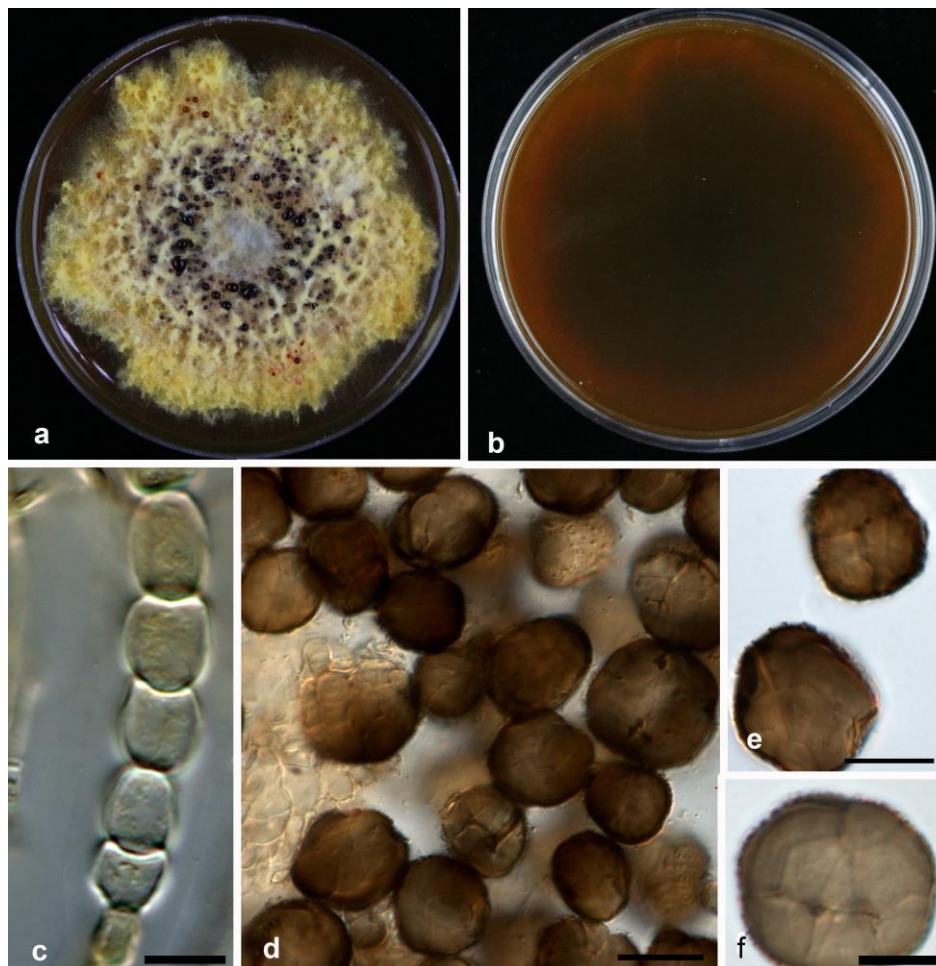


Figure 7 – *Epicoccum henanense*. a, b Colony on PDA, 10 days after incubation at 25 ± 1 °C (a from above, b from below). c Sporodochia d-f Conidia. Scale bars: c-f= 10 μm .

Epicoccum layuense Qian Chen, Crous & L. Cai (2017).

For description see Chen et al. (2017).

Material examined – China, Henan Province, Shihe District, from *Astragalus sinicus* root, May 2018, Zhao Wensheng and Zhang Guozhen, living culture JZB 380051, JZB 380052, Henan

Province, Shihe District, from *Vicia villosa* flower, May 2018, Zhao Wensheng and Zhang Guozhen, living culture JZB 380057, Henan Province, Shihe District, from *Vicia villosa* pod, May 2018, Zhao Wensheng and Zhang Guozhen, living culture JZB 380060, Henan Province Luoshan City, from *Astragalus sinicus* flower, May 2018, Zhao Wensheng and Zhang Guozhen, living culture JZB 380061, from *Astragalus sinicus* leaf, May 2018, Zhao Wensheng and Zhang Guozhen, living culture JZB 380062, Henan Province, Shihe District, from *Astragalus sinicus* pod, May 2018, Zhao Wensheng and Zhang Guozhen, living culture JZB 380063, JZB 380064, JZB 380065, Henan Province, Shihe District, from *Astragalus sinicus* leaf, May 2018, Zhao Wensheng and Zhang Guozhen, living culture JZB 380066, Guizhou Province, from *Astragalus sinicus* pod, May 2018, Zhao Wensheng and Zhang Guozhen, living culture JZB 380067, JZB 380068, JZB 380069.

Notes – Thirteen isolates were recovered from *Astragalus sinicus* (flower, leaf, pod, and roots) and *Vicia villosa* (flower, pod, and stems) plants from Henan and Guizhou provinces. These new isolates share a close phylogenetic affinity to *Epicoccum layuense* (CGMCC 318362 and LC8156) in our combined LSU, ITS, *rpb2*, and *tub* sequence data analyses (Fig. 5). *Epicoccum layuense* has been reported from *Avena sativa*, *Camellia sinensis*, and *Perilla* sp. from China (Chen et al. 2017, Valenzuela-Lopez et al. 2018, Raza et al. 2019, Chen et al. 2020). However, this species has not been reported from *Astragalus sinicus* or the *Vicia villosa*. Therefore, this is the first association of this species with *Astragalus sinicus* and *Vicia villosa* in China and worldwide (Farr & Rossman 2022). The phylogenetic placement of these isolates is shown in Fig. 5.

Epicoccum latusicollum Qian Chen, Crous & L. Cai (2017).

For description see Chen et al. (2017).

Material examined – China, Henan Province, Shihe District, from *Astragalus sinicus* pod, May 2018, Zhao Wensheng and Zhang Guozhen, living culture = JZB 380070, Henan Province, Shihe District, from *Astragalus sinicus* stem, May 2018, Zhao Wensheng, and Zhang Guozhen, living culture = JZB 380071, Fujian Province, from *Astragalus sinicus* stem, May 2018, Zhao Wensheng and Zhang Guozhen, living cultures = JZB 380072, JZB 380073, JZB 380074.

Notes – Five fungal isolates obtained from stems and pods of *Astragalus sinicus*, were identified as *Epicoccum latusicollum*, with the support of both morphology and phylogeny. These isolates formed a clade together with the type isolate of *E. latusicollum* (CGMCC 3.18346) in the combined LSU, ITS, *rpb2*, and *tub* phylogenetic tree (Fig. 5). *Epicoccum latusicollum* has been reported from *Acer palmatum*, *Camellia sinensis*, *Podocarpus macrophyllus*, *Saccharum officinarum*, *Sorghum bicolor*, and *Vitex negundo* from China, Japan, and Pakistan (Farr & Rossman 2022). However, this is the first report of *Epicoccum latusicollum* on *Astragalus sinicus* in China as well as in the world (Farr & Rossman 2022). The phylogenetic placement of these isolates is shown in Fig. 5.

Epicoccum rosae Wanas., Camporesi, E.B.G. Jones & K.D. Hyde (2018).

For description see Wanasinghe et al. (2018).

Material examined – China, Henan Province, Shihe District, from *Astragalus sinicus* flower, May 2018, Zhao Wensheng and Zhang Guozhen, living culture JZB 380075, JZB 380076, Henan Province, Shihe District, from *Astragalus sinicus* root, May 2018, Zhao Wensheng and Zhang Guozhen, living culture JZB 380077, JZB 380078.

Notes – Four new isolates associated with *Astragalus sinicus* are morphologically similar and phylogenetically related to *Epicoccum rosae*. *Epicoccum rosae* (holotype: MFLU 15-3639) was first reported on *Rosa canina* from Italy (Wanasinghe et al. 2018). This study is the first to report *Epicoccum rosae* occurring on *Astragalus sinicus* (Farr & Rossman 2022). The phylogenetic placement of these isolates is shown in Fig. 5.

Epicoccum viciae-villosae W. Zhao, Q. Ning, & J.Y. Yan, sp. nov.

Fig. 8

Index Fungorum Number: IF558424; Facesoffungi number: FoF 10794

Etymology – ‘*viciae-villosae*’ refers to the host plant *Vicia villosa* from which it was isolated.

Ecology – Associated with healthy pods of *Vicia villosa*

Holotype – JZBH 380079

Asexual morph: *Conidiomata* sporodochial, solitary or aggregated, immersed to semi-immersed, glabrous, covered with hyphal growth, blackish brown. *Hyphae* smooth, branched, septate, hyaline. *Chlamydoconidia* multicellular, produced in agar, hyaline to pale brown, subglobose or oblong, 10–20×6–10 μm (\bar{x} = 16.3×8.1 μm, n = 10). *Conidia*, multicellular, yellowish-brown to brown, globose to subglobose, 25–68×20–52 μm (\bar{x} = 37.3×29.7 μm, n = 40). *Sexual morph*: not observed.

Cultural characteristics – Colonies on PDA are slow-growing, covering a 30 mm Petri dish in 10–14 days after incubation at 25 ± 1 °C, yellowish white (surface) and reddish black (reverse), with a dense mat of aerial mycelium, rough, entire slightly radiating at the margin; colony from above rough, white at fruiting zone, slimy reddish spore mass at the centre, and yellowish at ageing zone. Later give reddish-brown colour to the PDA media.

Material examined – China, Henan Province, Shihe District, from *Vicia villosa* pod, May 2018, Zhao Wensheng and Zhang Guozhen (JZBH 380079, holotype inactive dry culture); ex-type living culture = JZB 380079.

Additional material examined – China, Henan Province, Shihe District, from *Vicia villosa* pod, May 2018, Zhao Wensheng and Zhang Guozhen, living culture JZB 380080, JZB 380081, JZB 380082, JZB 380086, Henan Province, Shihe District, from *Vicia villosa* stem, May 2018, Zhao Wensheng and Zhang Guozhen, living culture JZB 380083, Henan Province, Shihe District, from *Vicia villosa* leaf, May 2018, Zhao Wensheng and Zhang Guozhen, living culture JZB 380084, Henan Province, Shihe District, from *Vicia villosa* stem, May 2018, Zhao Wensheng and Zhang Guozhen, living culture JZB 380058, JZB 380059.

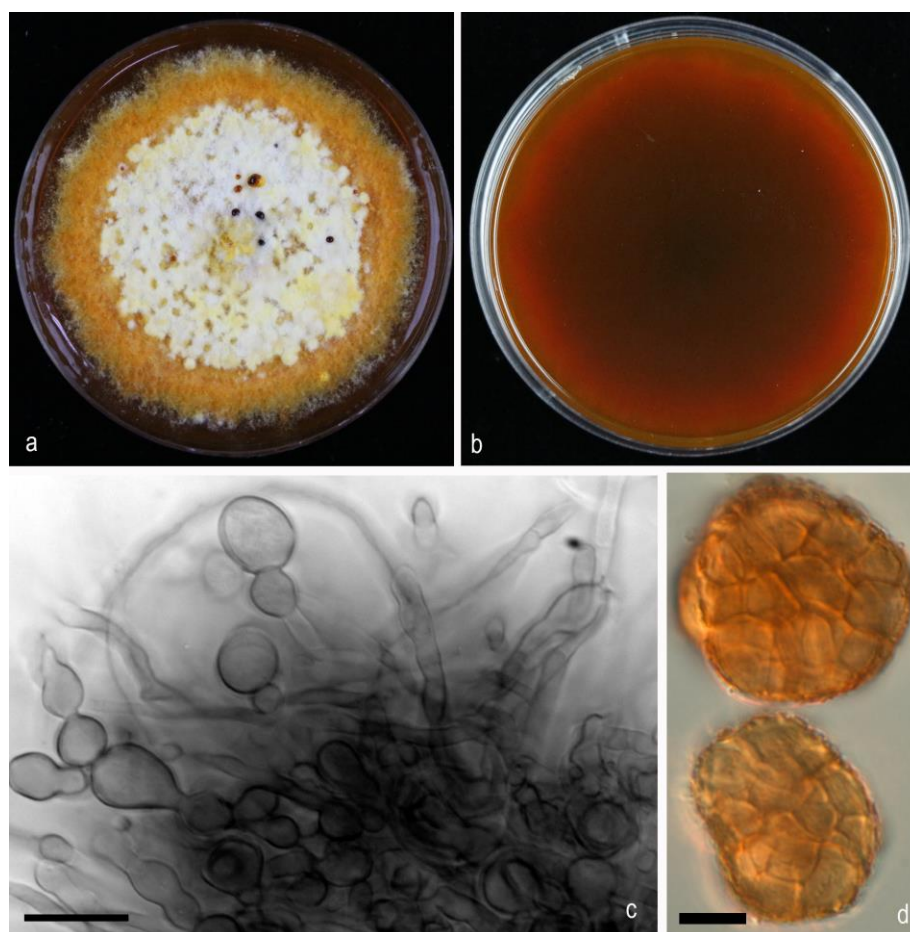


Figure 8 – *Epicoccum viciae-villosae*. a, b Colony on PDA, 10 days after incubation at 25 ± 1 °C (a from above, b from below). c Sporodochia. d Conidia. Scale bars: c, d = 10 μm.

Notes – In multi-loci phylogenetic analysis of *Epicoccum* species using LSU, ITS, *rpb2*, and *tub* markers, *Epicoccum viciae-villosae* developed a monophyletic clade with 84% ML support (Fig. 5) with *E. layuense*. *Epicoccum viciae-villosae* has relatively larger conidia than *E. layuense* (25.4–67.5×20.1–51.5 μm vs 13–19.5 μm). *Epicoccum layuense* produced subglobose -pyriform conidia, with a basal cell, that we could not find in *Epicoccum viciae-villosae*.

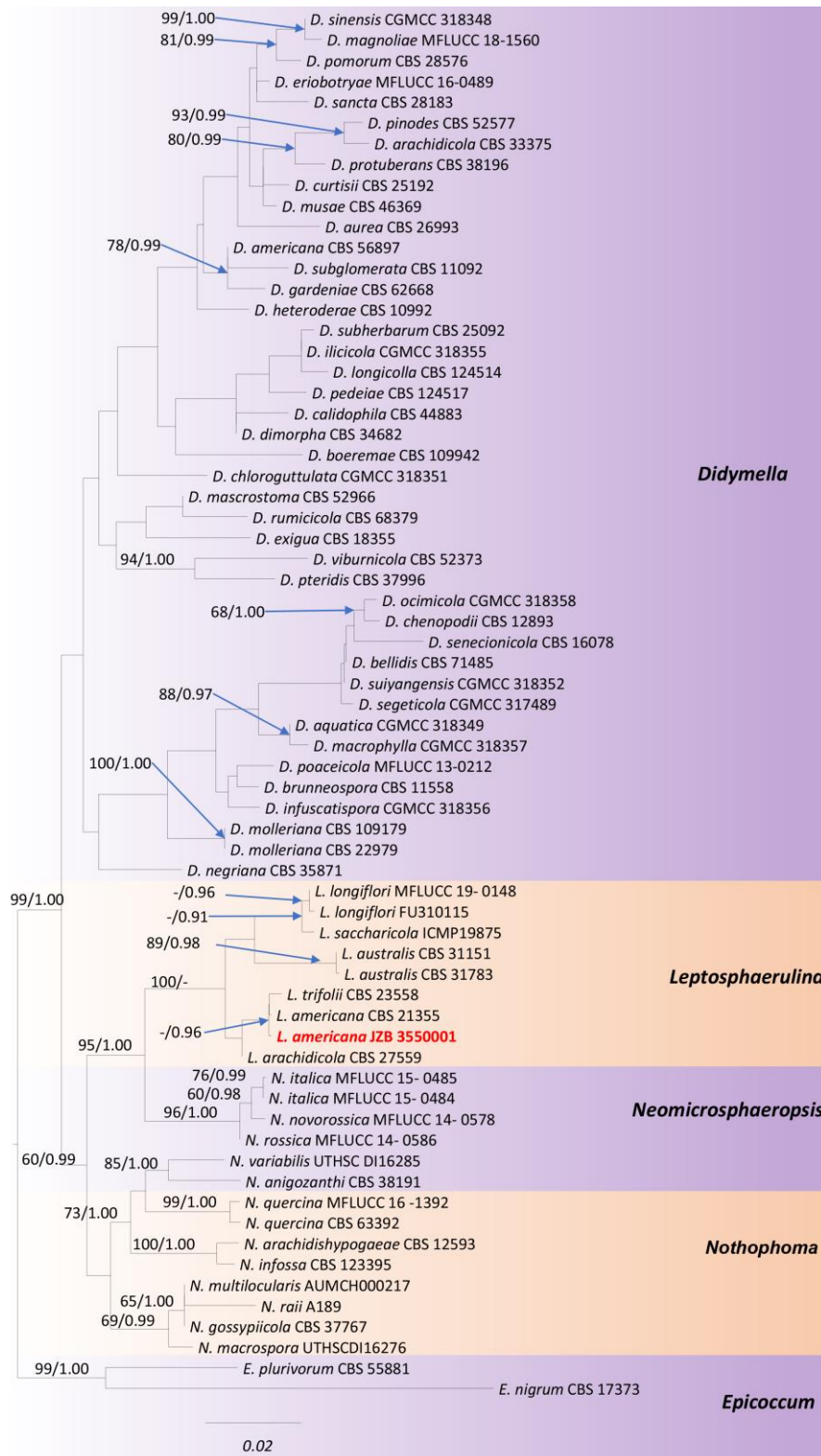


Figure 9 – Phylogram generated from maximum likelihood analysis based on combined LSU, ITS, and *rpb2* sequence data. The matrix had 373 distinct alignment patterns, with 11.94% undetermined characters or gaps. Estimated base frequencies were as follows: A = 0.246401, C = 0.228275, G =

0.280360, T = 0.244965; substitution rates AC = 1.283982, AG = 6.214793, AT = 1.728042, CG = 1.044122, CT = 15.789933, GT = 1.000000; gamma distribution shape parameter α = 0.837651. Bootstrap values for maximum likelihood and maximum parsimony equal to or greater than 60% and Bayesian posterior probabilities equal or greater than 0.90 are placed above the branches. The newly generated sequences are indicated in red. In this study, we recovered a strain of *Leptosphaerulina* which was identified as *Leptosphaerulina americana*, recorded for the first time on *Astragalus sinicus* from China.

Leptosphaerulina McAlpine (1902).

Leptosphaerulina americana (Ellis & Everh.) J.H. Graham & Luttr. (1961)

For description see Graham & Luttrell (1961).

Specimens examined – China, Guangxi Province, Guilin City, from *Astragalus sinicus* leaf, May 2018, Zhao Wensheng and Zhang Guozhen, living culture = JZB 3550001.

Notes – Isolate JZB 3550001 shared a close phylogenetic affinity to *Leptosphaerulina americana* (CBS 21355) in our combined LSU, ITS, and *rpb2* analyses (Fig. 9). Morphological characters and multi-marker analyses revealed and confirmed that JZB 3550001 is another strain for *L. americana*. *Leptosphaerulina americana* has been reported from *Terminalia bellerica* and *Trifolium pratense* from India, Georgia, and the USA (Farr & Rossman 2022). However, this species has not been reported from *Astragalus sinicus* and here we provide the first association of *L. americana* with *Astragalus sinicus* (Farr & Rossman 2022).

Didymosphaeriaceae Munk (1953).

Pseudopithomyces Ariyaw. & K.D. Hyde (2015).

Pseudopithomyces chartarum (Berk. & M.A. Curtis) Jun F. Li, Ariyaw. & K.D. Hyde (2015).

For description see Ariyawansa et al. (2015).

Specimens examined – China, Henan Province, Luoshan City, from *Astragalus sinicus* leaf, May 2018, Zhao Wensheng and Zhang Guozhen, living cultures = JZB 3560001, JZB 3560002.

Notes – Analyses of the concatenated ITS, GAPDH, and *tef1- α* dataset and morphological comparisons supported the isolates from this study as belonging to *Pseudopithomyces*. Even though there are numerous *Pseudopithomyces* species described from different host plants (*Pseudopithomyces chartarum*- *Triticum aestivum*: Argentina, *Pseudopithomyces karoo*- *Gnidia polycephala*: South Africa, *Pseudopithomyces palmicola*- *Chromolaena odorata*: Thailand, and *Pseudopithomyces pandanicola*- *Pandanus amaryllifolius*: Thailand), there is no record of *Pseudopithomyces* on *Astragalus sinicus*. Thus, this is the first report of *Pseudopithomyces chartarum* associated with *Astragalus sinicus*. *Pseudopithomyces kunmingensis* (Karun. & K.D. Hyde 2017) (Holotype: HKAS 97353) was introduced by Hyde et al. (2017) and was collected from the Yunnan Province, China on a dead leaf of an unidentified grass species (*Poaceae*). According to the phylogenetic analysis of Hyde et al. (2017), *P. kunmingensis* formed a clade together with *P. chartarum* with 61% ML support. In our phylogenetic analysis of combined ITS, GAPDH, and *tef1- α* sequence data, *P. kunmingensis* formed a clade with the strains of *P. chartarum* with high support (98% ML value; Fig. 10). There were no significant nucleotide differences among the gene regions. Thus, here we synonymise *Pseudopithomyces kunmingensis* with *P. chartarum*, based on morphological similarities and phylogenetic analysis.

Pleosporaceae Nitschke (1869).

Alternaria Nees (1816).

Alternaria alternata (Fr.) Keissl. (1912).

For description see Domsch et al. (2007).

Specimens examined – China, Guizhou Province, from *Astragalus sinicus* flower, May 2018, Zhao Wensheng and Zhang Guozhen, living culture JZB 3180041, Guangxi Province, Guilin city,

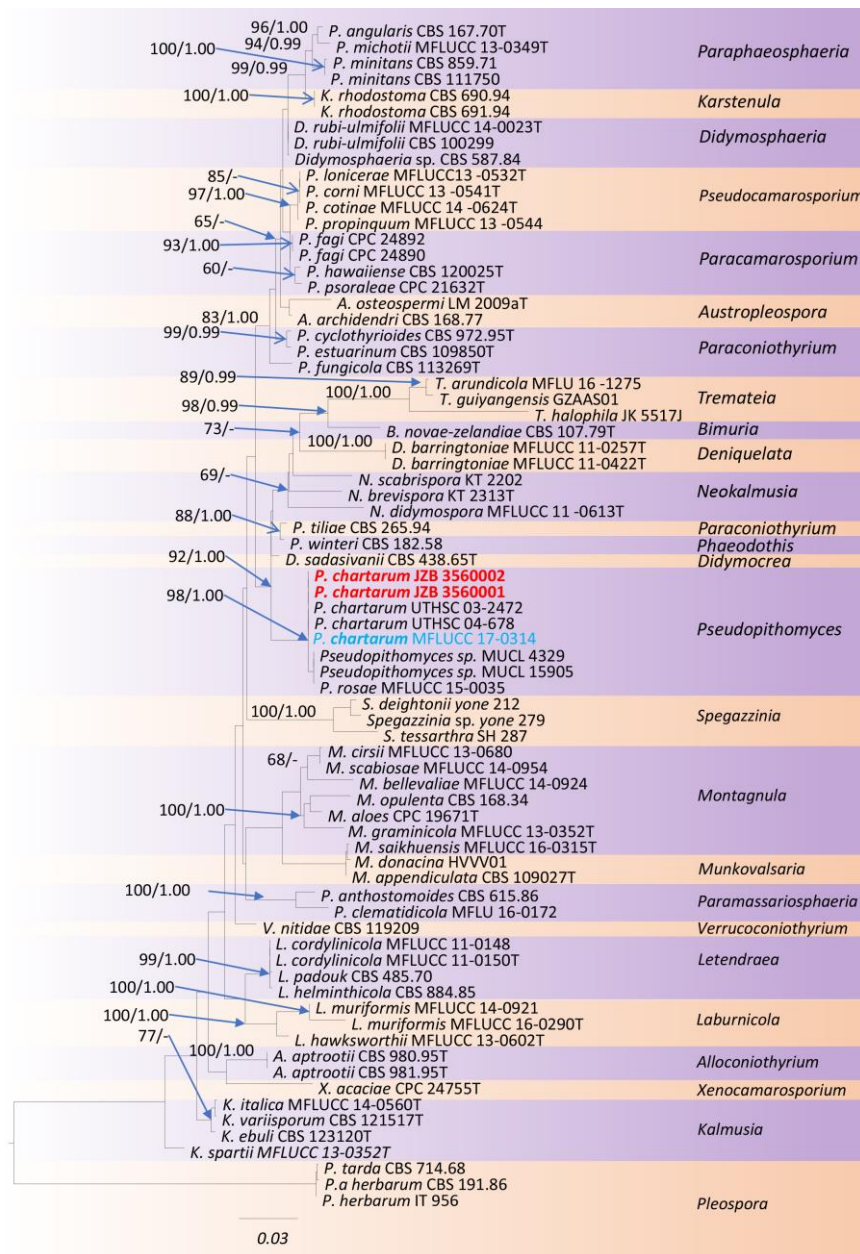


Figure 10 – Phylogram generated from maximum likelihood analysis based on combined LSU, SSU, ITS and *tefl-α* sequence data. The matrix had 901 distinct alignment patterns, with 38.17% undetermined characters or gaps. Estimated base frequencies were as follows: A = 0.240536, C = 0.242362, G = 0.277805, T = 0.239298; substitution rates AC = 1.314304, AG = 2.236188, AT = 1.277777, CG = 0.802011, CT = 7.326785, GT = 1.000000; gamma distribution shape parameter α = 0.802116. Bootstrap values for maximum likelihood and maximum parsimony equal to or greater than 60% and Bayesian posterior probabilities equal or greater than 0.90 are placed above the branches. The newly generated sequences are indicated in red. We obtained two strains of *Pseudopithomyces* which were identified as *Pseudopithomyces chartarum*, recorded for the first time on *Astragalus sinicus* from China.

from *Vicia villosa* stem, May 2018, Zhao Wensheng and Zhang Guozhen, living culture JZB 3180042, Guangxi Province, Guilin City, from *Vicia villosa* pod, May 2018, Zhao Wensheng and Zhang Guozhen, living culture JZB 3180043, Guangxi Province, Guilin City, from *Astragalus sinicus* leaf, May 2018, Zhao Wensheng and Zhang Guozhen, living culture JZB 3180044, Henan Province, Shihe District, from *Astragalus sinicus* stem, May 2018, Zhao Wensheng and Zhang Guozhen, living culture JZB 3180045, Henan Province, Shihe District, from *Vicia villosa* leaf, May

2018, Zhao Wensheng and Zhang Guozhen, living culture JZB 3180046, Henan Province, Luoshan City, from *Astragalus sinicus* leaf, May 2018, Zhao Wensheng and Zhang Guozhen, living culture JZB 3180047, living culture JZB 3180048, living culture JZB 3180049, Guangxi Province, Guilin City, from *Vicia villosa* pod, May 2018, Zhao Wensheng and Zhang Guozhen, living culture JZB 3180050, living culture JZB 3180051, Guizhou Province, from *Astragalus sinicus* leaf, May 2018, Zhao Wensheng and Zhang Guozhen, living culture JZB 3180052, Henan Province, Shihe district, from *Astragalus sinicus* leaf, May 2018, Zhao Wensheng and Zhang Guozhen, living culture JZB 3180053, Guangxi Province, Guilin City, from *Vicia villosa* pod, May 2018, Zhao Wensheng and Zhang Guozhen, living culture JZB 3180054, living culture JZB 3180055, Guangxi Province, Guilin City, from *Astragalus sinicus* leaf, May 2018, Zhao Wensheng and Zhang Guozhen, living culture JZB 3180056, living culture JZB 3180057, Guangxi Province, Guilin City, from *Astragalus sinicus* pod, May 2018, Zhao Wensheng and Zhang Guozhen, living culture JZB 3180058, Guangxi Province, Guilin City, from *Astragalus sinicus* leaf, May 2018, Zhao Wensheng and Zhang Guozhen, living culture JZB 3180059, Henan Province, Shihe District, from *Astragalus sinicus* flower, May 2018, Zhao Wensheng and Zhang Guozhen, living culture JZB 3180060, Guangxi Province, Guilin City, from *Vicia villosa* pod, May 2018, Zhao Wensheng and Zhang Guozhen, living culture JZB 3180061, Guangxi Province, Guilin City, from from *Vicia villosa* pod, May 2018, Zhao Wensheng and Zhang Guozhen, living culture JZB 3180062, Henan Province, Shihe District, from *Astragalus sinicus* stem, May 2018, Zhao Wensheng and Zhang Guozhen, living culture JZB 3180063.

Notes – Twenty-three isolates of *Alternaria alternata* were recovered from *Astragalus sinicus* and *Vicia villosa* in Guangxi, Guizhou, and Henan provinces (Fig. 11). According to Farr & Rossman (2022), *Alternaria alternata* was recorded from *Vicia villosa* in Oregon (Shaw 1973). However, we could not find any records of *Alternaria* species from *Astragalus sinicus* in China or other parts of the world (Farr & Rossman 2022). Therefore, we provide the first host association of *Alternaria alternata* with *Astragalus sinicus*.

Alternaria astragalicola W. Zhao, Q. Ning, & J.Y. Yan, sp. nov.

Fig. 12

Index Fungorum Number: IF558425; Facesoffungi number: FoF 10795

Etymology – ‘astragalicola’ refers to the host plant *Astragalus* from which it was isolated.

Ecology – Associated with healthy pods of *Astragalus sinicus*

Holotype – JZBH 3180064

Asexual morph: *Hyphae* subhyaline to pale olivaceous, branched, smooth, septate. *Conidiophores* 25–119×2.5–4.5 μm (\bar{x} = 44.6×3.9 μm, n = 10), solitary, simple, straight, or flexuous, pale brown, multi-septate, with a single terminal conidiogenous locus. *Conidia* 10.5–30×7–12 μm (\bar{x} = 17.1×9.2 μm, n = 50) solitary or in branched chains of 2, straight, clavate to elongated clavate, light brown to dark brown, with a smooth outer wall, some muriform, usually with 2–3 transverse septa and 1–2 longitudinal septa, rounded apex, stalked or stalkless. *Sexual morph.* not observed.

Culture characteristics – Colonies on PCA attaining 80 mm diam. after 10 days at 25 °C in 12h light and 12h dark, circular, entire-edged, effuse, floccose to woolly, surface pale olivaceous grey near the margin changing to dark green in the centre and reverse olivaceous black.

Material examined – China, Henan Province, Shihe District, from *Astragalus sinicus* pod, May 2018, Zhao Wensheng and Zhang Guozhen, (JZBH 3180064, holotype inactive dry culture), ex-type living culture = JZB 3180064.

Notes – The new strain fits well into the concept of *Alternaria*. In multi-marker analysis, for *Alternaria astragalicola* formed a sister clade to *A. pseudoehorniae* (MFLUCC 18–1589) with a 62% ML support (Fig. 11). Morphological comparison between ex-type strains of *Alternaria pseudoehorniae* and *A. astragalicola* revealed different conidiophores and conidial characters. Compared to our strain, *A. pseudoehorniae* have small conidiophores (24.9– 118.8×2.5– 4.5 μm vs 18– 48.5 × 2.5– 6 μm) and larger conidia (10.5– 29.7×6.7–12.1 μm vs 16–30.2 × 5– 13 μm)

(Chethana et al. 2019). *Alternaria astragalicola* have conidia in branched chains of two, while *A. pseudoeichhorniae* produce conidia in a chain of 2–4 or more.

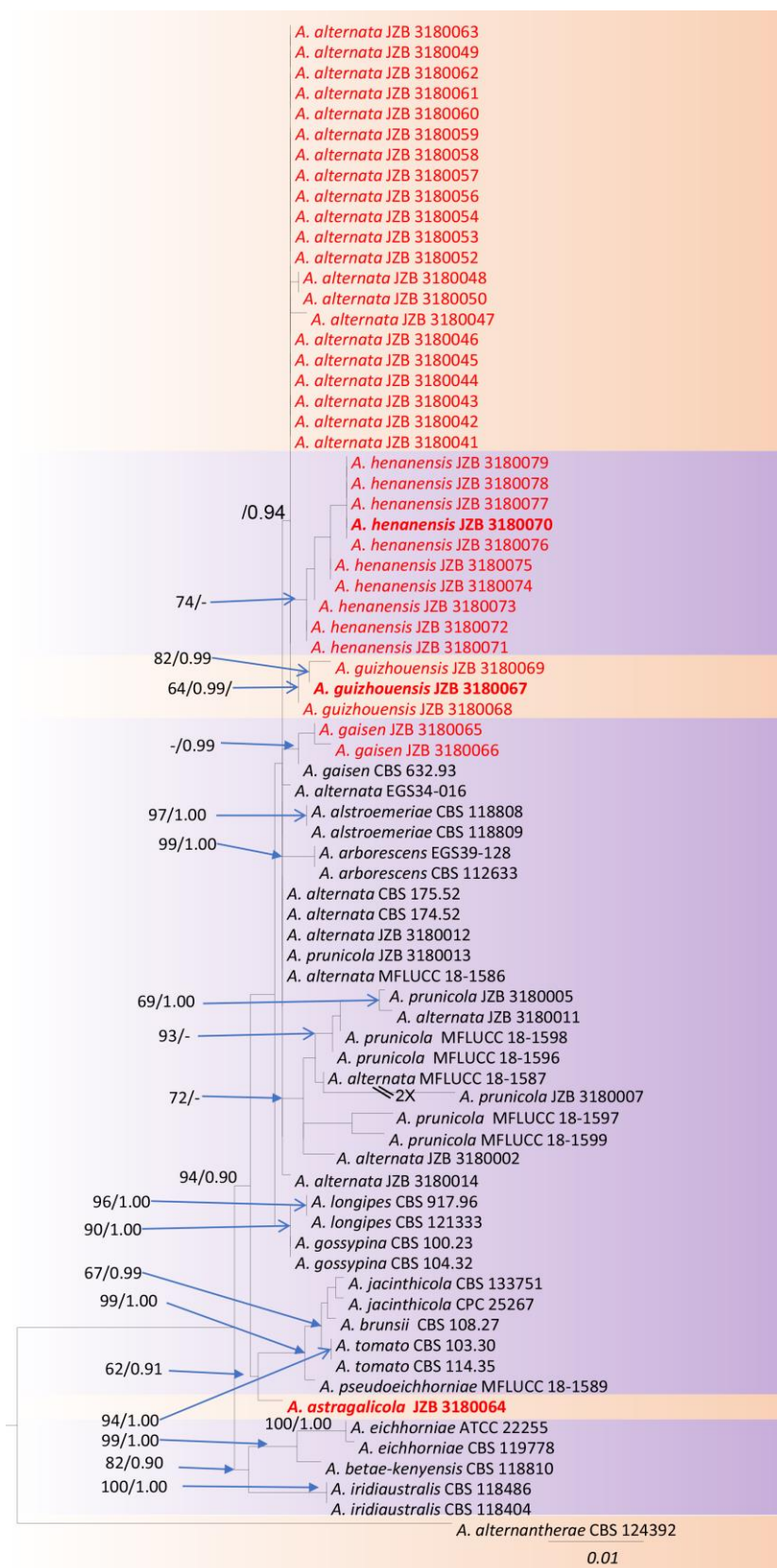


Figure 11 – Phylogram generated from maximum likelihood analysis based on combined ITS, *GAPDH*, and *tef-1a* sequence data. The matrix had 138 distinct alignment patterns, with 2.70% undetermined characters or gaps. Estimated base frequencies were as follows: A = 0.230278, C =

0.283771, G = 0.239952, T = 0.245998; substitution rates AC = 1.696344, AG = 2.226031, AT = 1.247758, CG = 1.341410, CT = 3.973766, GT = 1.000000; gamma distribution shape parameter α = 0.802116. Bootstrap values for maximum likelihood and maximum parsimony equal to or greater than 60% and Bayesian posterior probabilities equal or greater than 0.90 are placed above the branches. The newly generated sequences are indicated in red.



Figure 12 – *Alternaria astragalicola*. a Colony on PCA. b. Conidiophore. c–f Conidia. g Conidial arrangement. Scale bars: b–g = 10 μ m.

Alternaria gaisen Nagano ex Hara (1928).

For description see Simmons (2007).

Material examined – China, Guangxi Province, Nanning City, from *Vicia villosa* pod, May 2018, Zhao Wensheng and Zhang Guozhen, living culture JZB 3180065, Guangxi Province, Guilin city, from *Astragalus sinicus* leaf, living culture JZB 3180066.

Note – Two isolates were recovered from *Astragalus sinicus* and *Vicia villosa* in Guangxi Province. These new isolates shared a close phylogenetic affinity to *Alternaria gaisen* (CBS 632.93) in our sequence analyses (Fig. 11). This relationship is supported by ML analysis. We could not find any records of *Alternaria gaisen* species from *Astragalus sinicus* or *Vicia villosa* in China or other parts of the world (Farr & Rossman 2022). Therefore, we provide the first host association of *Alternaria gaisen* with *Astragalus sinicus* and *Vicia villosa*.

Alternaria guizhouensis W. Zhao, Q. Ning, & J.Y. Yan, sp. nov.

Fig. 13

Index Fungorum Number: IF558426; Facesoffungi number: FoF 10796

Etymology – ‘guizhouensis’ refers to the Guizhou province in China from which it was isolated.

Ecology – Associated with healthy flowers of *Astragalus sinicus*

Holotype – JZBH 3180067

Asexual morph: Hyphae subhyaline to pale olivaceous, branched, smooth, septate. Conidiophores 18–116 \times 4–6 μ m (\bar{x} = 40.1 \times 4.7 μ m, n = 20), solitary, simple, straight, or flexuous, dark brown, multi-septate, with a single or two terminal conidiogenous loci. Conidia 15–38 \times 8–13 μ m (\bar{x} = 27.7 \times 10.6 μ m, n = 50), solitary or in branched chains of 4 or more, straight, clavate to elongated clavate, olivaceous to light brown, with a smooth outer wall, some muriform, usually with 4–6 transverse septa and 0–1 longitudinal septum, rounded apex, stalked or stalkless. *Sexual morph*: not observed.

Culture characteristics – Colonies on PCA attaining 80 mm diam. after 8–9 days at 25 $^{\circ}$ C in 12h light and 12h dark, circular, entire-edged, effuse, floccose to woolly, surface pale olivaceous grey- white near the margin changing to dull green in the centre and reverse olivaceous black in the centre and pale olivaceous grey near the margin.

Material examined – China, Guizhou Province, from *Astragalus sinicus* flower, May 2018, Zhao Wensheng, and Zhang Guozhen (JZBH 3180067, holotype inactive dry culture) ex-type living culture = JZB 3180067.

Additional material examined – China, Henan Province, Shihe District, from *Astragalus sinicus* stem, May 2018, Zhao Wensheng and Zhang Guozhen, living culture = JZB 3180068, Guangxi Province, Guilin City, from *Astragalus sinicus* leaf, May 2018, Zhao Wensheng and Zhang Guozhen, living culture = JZB 3180069.

Notes – We have recovered three isolates from *Astragalus sinicus* flowers, leaves, and stems that fit well with the species concept of *Alternaria*. Multi-marker analysis revealed that *Alternaria guizhouensis* isolates form a sister clade to *A. henanensis*; another novel species recovered in this study (Fig. 11) with 64% ML support. Morphological comparison between the ex-type strain of *Alternaria guizhouensis* and *A. henanensis* revealed different conidial characters. Compared to *Alternaria guizhouensis*, *A. henanensis* has small conidiophores ($24.9\text{--}118.8 \times 2.5\text{--}4.5 \mu\text{m}$ vs $18\text{--}48.5 \times 2.5\text{--}6 \mu\text{m}$), larger conidia ($10.5\text{--}29.7 \times 6.7\text{--}12.1 \mu\text{m}$ vs $16\text{--}30.2 \times 5\text{--}13 \mu\text{m}$), also *A. guizhouensis* produced conidia in branched chains of 4 or more, while *A. henanensis* produced conidia in branched chains of three. Further these two species also can distinguish by the septa in conidia; *A. guizhouensis* have 4–6 transverse septa and 0–1 longitudinal septum while, *A. henanensis* produce conidia with 3–7 transverse septa and 0–1 longitudinal septum.

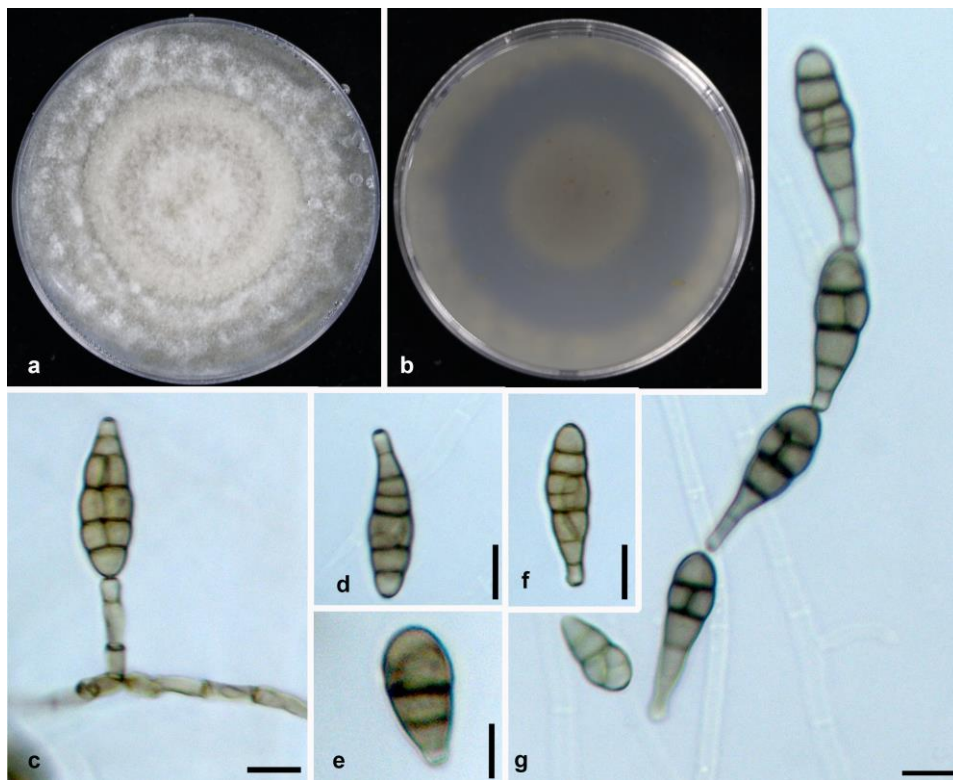


Figure 13 – *Alternaria guizhouensis*. a Upper view of the colony on PCA. b Back view of the colony on PCA. c Conidiophore. d-f Conidia. g Conidial arrangement. Scale bars: c = 20 μm , d-f = 10 μm , g = 20 μm .

Alternaria henanensis W. Zhao, Q. Ning, & J.Y. Yan, sp. nov.

Fig. 14

Index Fungorum Number: IF558421; Facesoffungi number: FoF 10797

Etymology – ‘henanensis’ refers to the Henan province in China from which the holo-type was isolated.

Ecology – Associated with healthy leaves of *Astragalus sinicus*

Holotype – JZBH 3180070

Asexual morph: *Hyphae* subhyaline to pale olivaceous, branched, smooth, septate. *Conidiophores* 25–95×3.5–6 µm (\bar{x} = 50.9×4.6 µm, n = 20), solitary, simple, straight, or flexuous, hyaline to pale brown, septate, with a single terminal conidiogenous locus. *Conidia* 13–54.5×8–18 µm (\bar{x} = 30.9×12.3 µm, n = 50), solitary or in branched chains of 3, 1–3 chains from one conidium, straight, clavate to elongated clavate, olivaceous to light brown, with a smooth outer wall, some muriform, usually with 3–7 transverse septa and 0–1 longitudinal septum, rounded apex, stalked or stalkless. *Sexual morph:* not observed.

Culture characteristics – Colonies on PCA attaining 80 mm diam. after 9–10 days at 25 °C in 12h light and 12h dark, circular, entire-edged, effuse, floccose to woolly, surface pale olivaceous grey near the margin changing to dull green in the centre and reverse olivaceous black in the centre and pale olivaceous grey near the margin.

Material examined – China, Henan Province, Shihe District, from *Astragalus sinicus* leaf, May 2018, Zhao Wensheng and Zhang Guozhen, (JZBH 3180070, holotype inactive dry culture), ex-type living culture = JZB 3180070.

Additional material examined – China, Guizhou Province, from *Astragalus sinicus* pod, May 2018, Zhao Wensheng and Zhang Guozhen, living culture = JZB 3180071, JZB 3180072, Henan Province, Shihe District, from *Vicia villosa* pod, May 2018, Zhao Wensheng and Zhang Guozhen, living culture = JZB 3180073, Henan Province, Shihe District, from *Astragalus sinicus* flower, May 2018, Zhao Wensheng and Zhang Guozhen, living culture = JZB 3180074, Henan Province, Shihe District, from *Vicia villosa* pod, May 2018, Zhao Wensheng and Zhang Guozhen, living culture = JZB 3180075, Guizhou Province, from *Astragalus sinicus* flower, May 2018, Zhao Wensheng and Zhang Guozhen, living culture = JZB 3180076, Guangxi Province, Guilin City, from *Astragalus sinicus* pod, May 2018, Zhao Wensheng and Zhang Guozhen, living culture = JZB 3180077, Henan Province, Shihe District, from *Astragalus sinicus* leaf, May 2018, Zhao Wensheng and Zhang Guozhen, living culture = JZB 3180078, Guangxi Province, Guilin City, from *Astragalus sinicus* pod, May 2018, Zhao Wensheng and Zhang Guozhen, living culture = JZB 3180079.

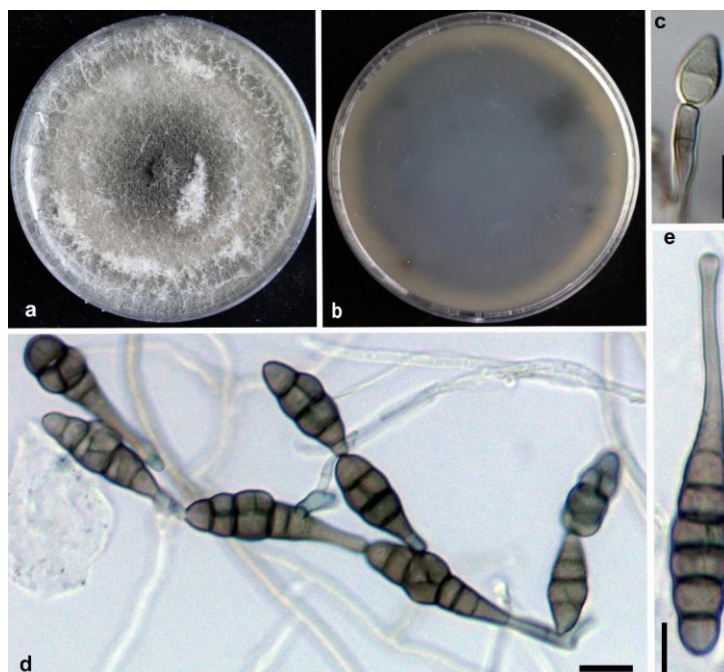


Figure 14 – *Alternaria henanensis*. a Upper view of the colony on PCA. b Back view of the colony on PCA. c Conidiophore. d Conidial arrangement, e Conidia. Scale bars: c = 10 µm, d, e = 20 µm.

Notes – During this study, we recovered ten *Alternaria* isolates from both *Astragalus sinicus* and *Vicia villosa* crops and morphologically and phylogenetically they are new species. Multi-

marker phylogeny showed that *A. henanensis* produced a sister clade to *A. alternata* with a 74% ML support (Fig. 11). Morphological comparison between the ex-type strain of *Alternaria henanensis* and *A. alternata* revealed different conidial characters. *Alternaria henanensis* produced relatively larger conidia (13–55×8–18 µm) than *A. alternata*.

Stemphylium Wallr. (1833).

Stemphylium astragali (Yoshii) W. Yamam. (1960).

For description see Yoshii (1929).

Material examined – China, Henan Province, Shihe District, from *Astragalus sinicus* flower, May 2018, Zhao Wensheng and Zhang Guozhen, living cultures = JZB 3240024, Henan Province, Shihe District, from *Astragalus sinicus* leaf, May 2018, Zhao Wensheng and Zhang Guozhen, living cultures = JZB 3240025.

Notes – Two isolates were recovered from *Astragalus sinicus* in Henan Province. These new isolates shared a close phylogenetic affinity to *Stemphylium astragali* (CBS 116583) in our combined sequence analyses. This relationship was strongly supported (100%) in our ML bootstrap analysis (Fig. 15). Four records of *Stemphylium astragali* species have been recovered from *Astragalus sinicus* in China, Japan, Korea, and South Korea so far (Farr & Rossman 2022).

Leotiomyces O.E. Erikss. & Winka (1997).

Helotiales Nannf. (1932).

Sclerotiniaceae Whetzel (1945).

Botrytis P. Micheli (1729).

Botrytis cinerea Pers. (1801).

For description see Persoon (1794).

Material examined – China, Guangxi Province, Guilin City, from *Astragalus sinicus* leaf, March 2018, Zhao Wensheng and Zhang Guozhen, living cultures = JZB 350044, JZB 350045, JZB 350046, JZB 350047.

Notes – Four isolates were recovered from *Astragalus sinicus* leaves in China. They were morphologically similar and phylogenetically related to *Botrytis cinerea*. According to Farr and Rossman (2022), *Botrytis cinerea* has been recorded on many host plants including *Astragalus sinicus* from China. The phylogenetic placement of these isolates is shown in Fig. 16.

Sclerotinia Fuckel (1870).

Sclerotinia minor Jagger (1920).

For description see Jagger (1920).

Material examined – China, Henan Province Luoshan City, from *Astragalus sinicus* stem, March 2018, Zhao Wensheng and Zhang Guozhen, living cultures = JZB 3570001, JZB 3570002, JZB 3570003, JZB 3570004, JZB 3570005, JZB 3570006, JZB 3570007, JZB 3570008, JZB 3570009, JZB 3570010, JZB 3570020, JZB 3570021, JZB 3570022, JZB 3570023, JZB 3570024, JZB 3570025, JZB 3570026, JZB 3570027, JZB 3570028, JZB 3570029, JZB 3570030, JZB 3570031.

Notes – Twenty-two new isolates were recovered from *Astragalus sinicus* stems in China. These were morphologically similar and phylogenetically related to *Sclerotinia minor*. According to Farr and Rossman (2022), *Sclerotinia minor* has been recorded on a range of hosts (*Brassica rapa* subsp. *pekinensis*, *Capsella bursa-pastoris*, *Conyza canadensis*, *Fragaria gracilis*, *Helianthus annuus*, *Lactuca sativa*, *Oenanthе javanica*, *Orobanche cumana*, *Pisum sativum*, *Plantago* sp., *Ranunculus ternatus*, *Salvia plebeia*, *Trifolium repens*, and *Vicia faba*) from China. However, our collection on *Astragalus sinicus* is the first to report *Sclerotinia minor* associated with *Astragalus sinicus* in China (Farr & Rossman 2022). The phylogenetic placement of these isolates is shown in Fig. 17.

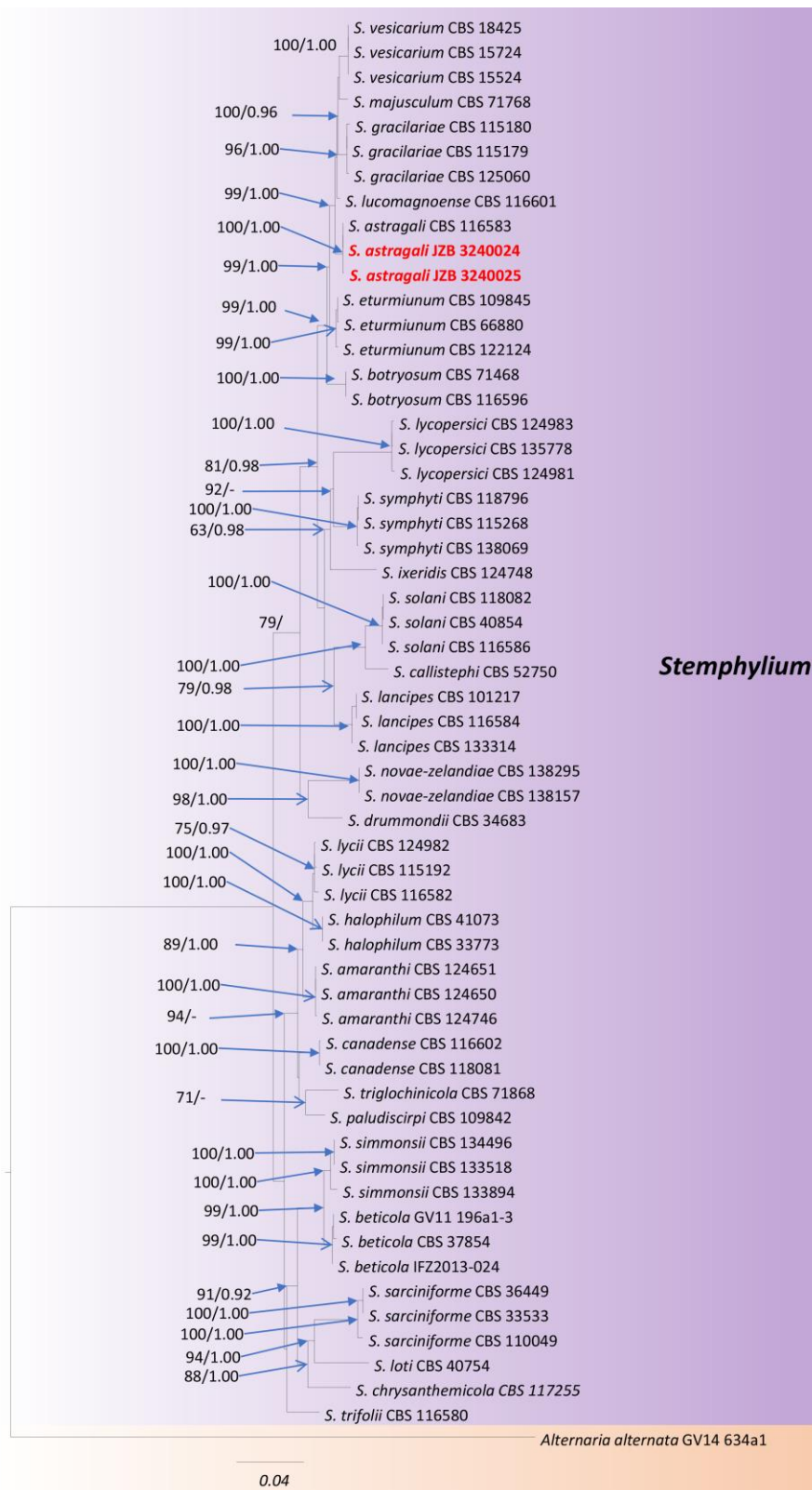


Figure 15 – Phylogram generated from maximum likelihood analysis based on combined ITS, *GAPDH* and *CAL* sequence data. The matrix had 438 distinct alignment patterns, with 0.52% undetermined characters or gaps. Estimated base frequencies were as follows: A = 0.271410, C = 0.237641, G = 0.235589, T = 0.255360; substitution rates AC = 1.485583, AG = 4.028671, AT = 1.106764, CG = 0.545460, CT = 10.621277, GT = 1.000000; gamma distribution shape parameter α = 0.802116. Bootstrap values for maximum likelihood and maximum parsimony equal to or greater than 60% and Bayesian posterior probabilities equal or greater than 0.90 are placed above

the branches. The newly generated sequences are indicated in red. We obtained two strains of *Stemphylium* which were identified as *Stemphylium astragali* on *Astragalus sinicus*.

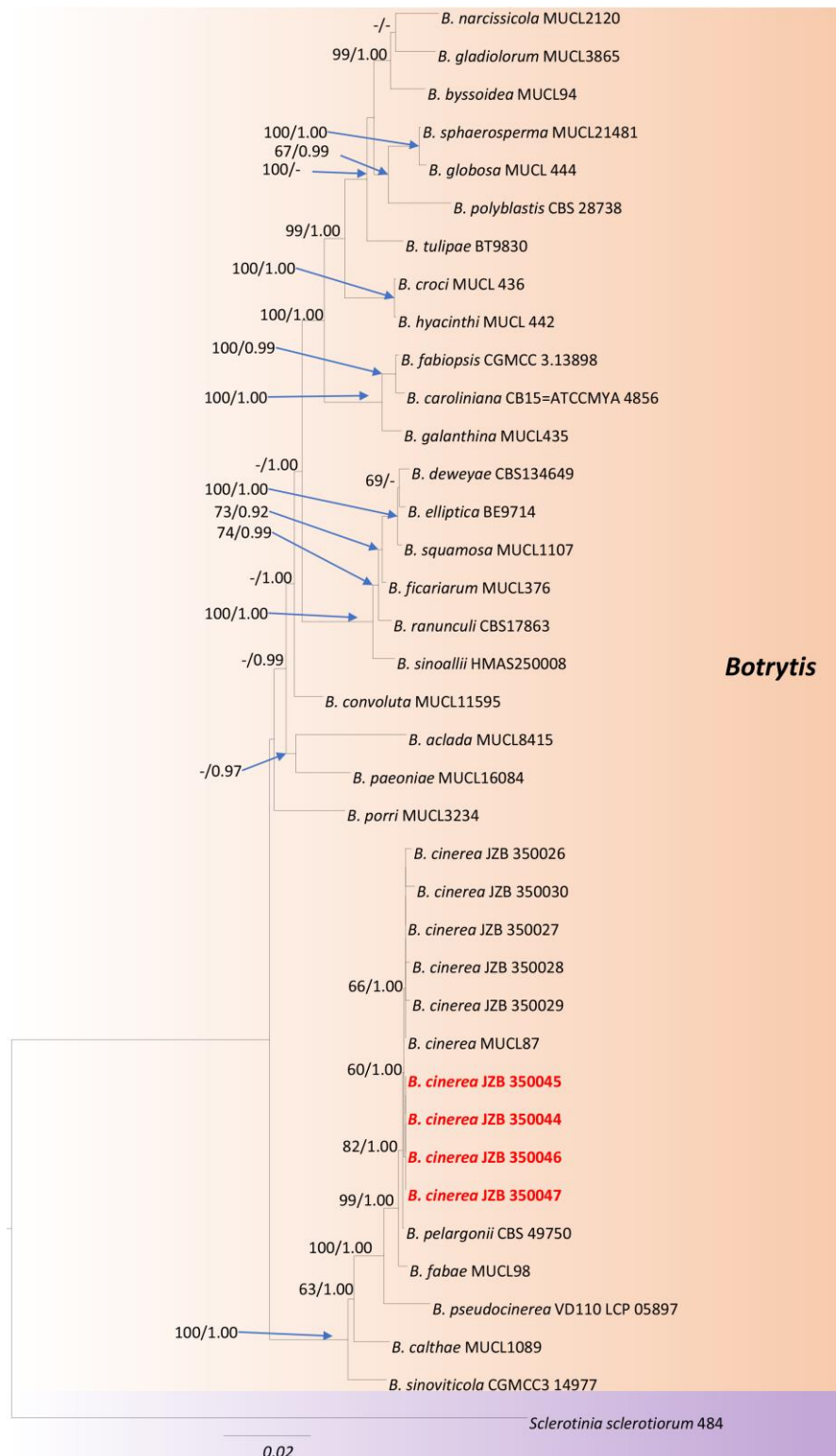


Figure 16 – Phylogram generated for *Botrytis* species from maximum likelihood analysis based on combined *rpb2*, *G3PDH* and *HSP60* sequence data. The matrix had 438 distinct alignment patterns, with 0.52% undetermined characters or gaps. Estimated base frequencies were as follows: A = 0.271410, C = 0.237641, G = 0.235589, T = 0.255360; substitution rates AC = 1.485583, AG = 4.028671, AT = 1.106764, CG = 0.545460, CT = 10.621277, GT = 1.000000; gamma distribution shape parameter α = 0.802116. Bootstrap values for maximum likelihood and maximum parsimony

equal to or greater than 60% and Bayesian posterior probabilities equal or greater than 0.90 are placed above the branches. The newly generated sequences are indicated in red. We obtained four strains of *Botrytis* which were identified as *Botrytis cinerea* on *Astragalus sinicus*.

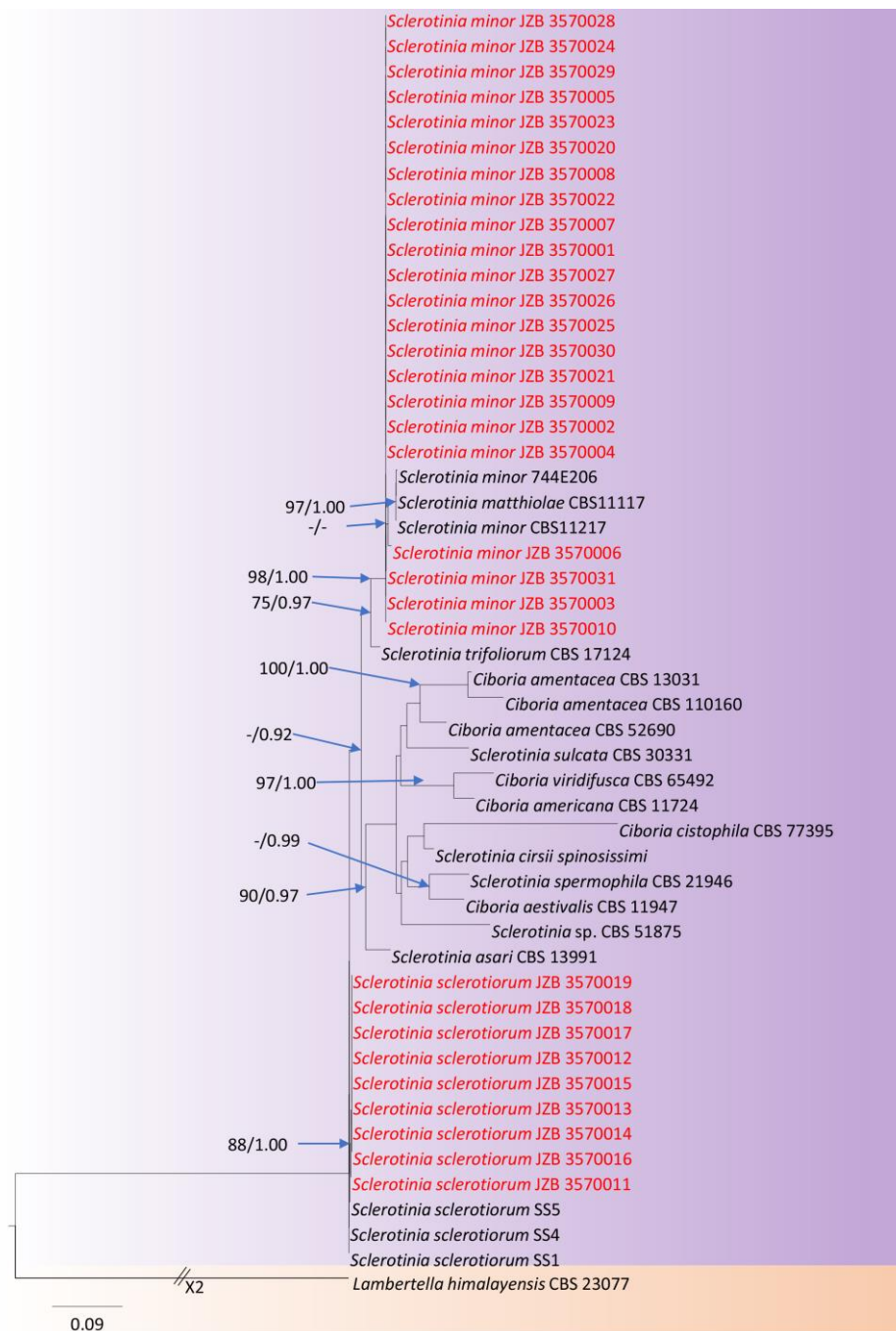


Figure 17 – Phylogram generated for *Sclerotinia* species from maximum likelihood analysis based on combined *CAL*, *ITS* and *MCM* sequence data. The matrix had 592 distinct alignment patterns, with 11.68% undetermined characters or gaps. Estimated base frequencies were as follows: A = 0.273395, C = 0.222565, G = 0.222517, T = 0.281523; substitution rates AC = 2.163924, AG = 5.100866, AT = 1.480784, CG = 1.173353, CT = 9.436685, GT = 1.000000; gamma distribution shape parameter $\alpha = 0.516199$. Bootstrap values for maximum likelihood and maximum parsimony equal to or greater than 60% and Bayesian posterior probabilities equal or greater than 0.90 are placed above the branches. The newly generated sequences are indicated in red. We obtained 31 strains of *Sclerotinia* which were identified as *Sclerotinia minor* and *Sclerotinia sclerotiorum* on *Astragalus sinicus*.

Sclerotinia sclerotiorum (Lib.) de Bary (1884).

For description see Bary (1884).

Material examined – China, Guangxi Province, Guilin City, from *Astragalus sinicus* stem, March 2018, Zhao Wensheng and Zhang Guozhen, living cultures = JZB 3570011, JZB 3570012, JZB 3570013, JZB 3570014, JZB 3570015, JZB 3570016, JZB 3570017, JZB 3570018, JZB 3570019.

Notes – Nine new isolates were recovered from *Astragalus sinicus* stems in China. These were similar and phylogenetically related to *Sclerotinia sclerotiorum*. Previously *Sclerotinia sclerotiorum* has been reported on many host plants in China including *Astragalus sinicus* (Tai 1979). The phylogenetic placement of these isolates is shown in Fig. 17.

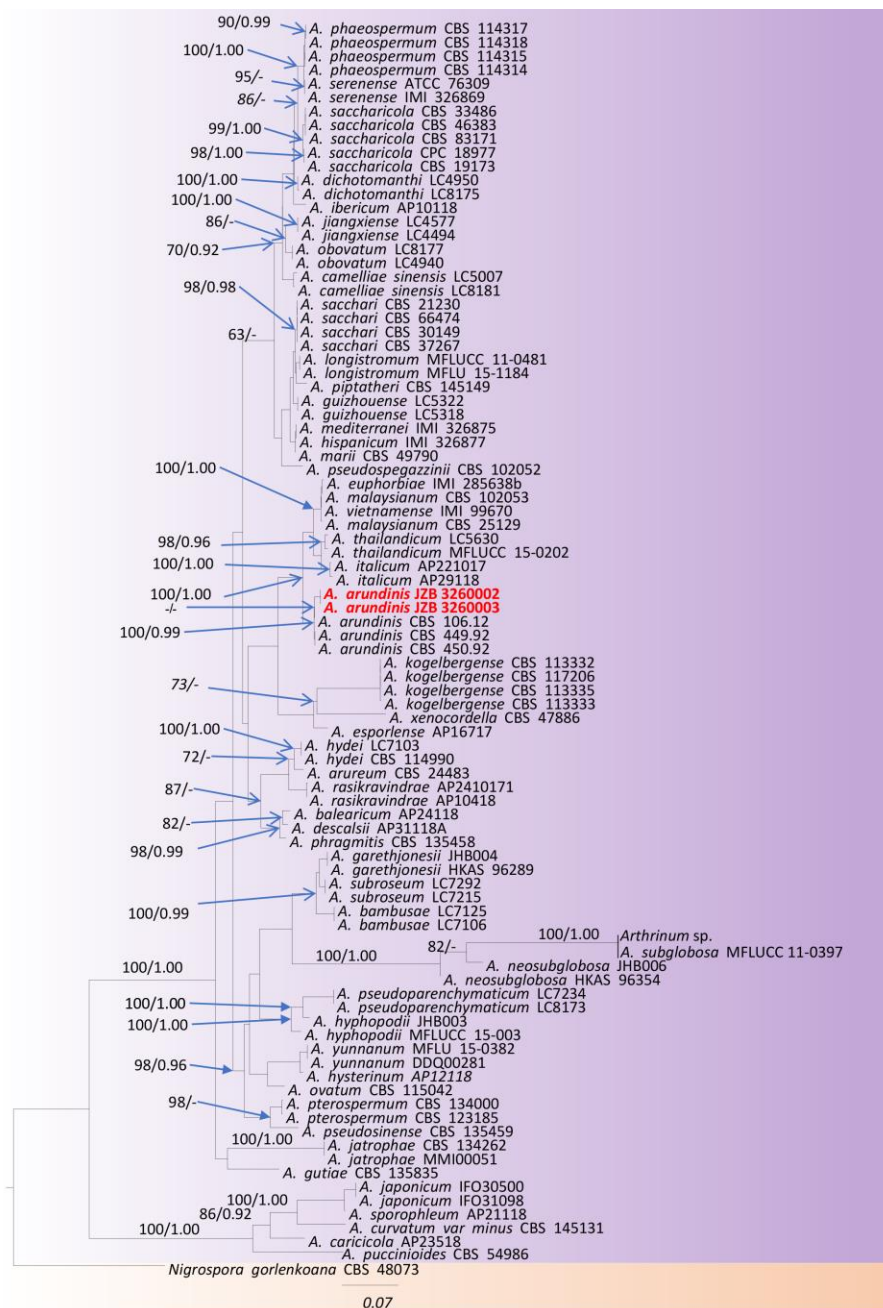


Figure 18 – Phylogram generated for *Arthriniinae* species from maximum likelihood analysis based on combined ITS, LSU, *tef-1a* and *tub* sequence data. The matrix had 1163 distinct alignment patterns, with 28.39% undetermined characters or gaps. Estimated base frequencies were as follows: A = 0.236174, C = 0.249633, G = 0.260384, T = 0.253810; substitution rates AC = 1.097861, AG = 3.110670, AT = 1.177962, CG = 0.973136, CT = 5.327378, GT = 1.000000;

gamma distribution shape parameter $\alpha = 0.757680$. Bootstrap values for maximum likelihood and maximum parsimony equal to or greater than 60% and Bayesian posterior probabilities equal to or greater than 0.90 are placed above the branches. The newly generated sequences are indicated in red. We obtained two strains of *Arthrinium* which were identified as *Arthrinium arundinis* on *Astragalus sinicus* from China.

Sordariomycetes O.E. Erikss. & Winka (1997).

Amphisphaeriales D. Hawksw. & O.E. Erikss. (1986).

Apiosporaceae K.D. Hyde, J. Fröhl., Joanne E. Taylor & M.E. Barr (1998).

Arthrinium Kunze (1817)

Arthrinium arundinis (Corda) Dyko & B. Sutton (1979).

For description see Crous & Groenewald (2013).

Material examined – China, Henan Province, Luoshan City, from *Astragalus sinicus* leaf, May 2018, Zhao Wensheng and Zhang Guozhen, living cultures JZB 3260002, JZB 3260003.

Notes – Two isolates were recovered from *Astragalus sinicus* leaves in China. These were similar and phylogenetically related to *Arthrinium arundinis*. Many *Arthrinium arundinis* species have been recorded from many host plants in China. However, according to our knowledge, this study is the first to report *Arthrinium arundinis* associated with *Astragalus sinicus* (Farr & Rossman 2022). The phylogenetic placement of this isolate is shown in Fig. 19.

Diaporthales Nannf. (1932).

Diaporthaceae Höhn. ex Wehm. (1926).

Diaporthe Nitschke (1870).

Diaporthe longicolla J.M. Santos, Vrandečić & A.J.L. Phillips (2011).

For description see Santos et al. (2011).

Material examined – China, Guizhou Province, from *Astragalus sinicus* stem, May 2018, Zhao Wensheng and Zhang Guozhen, living cultures = JZB 320180.

Notes – One isolate from *Astragalus sinicus* stems is similar and phylogenetically related to *Diaporthe longicolla*. According to Farr and Rossman (2022), many *Diaporthe* species have been recorded from broad host ranges in China. *Diaporthe longicolla* was initially reported from seeds, pods and stems of *Glycine max* cv. Wells from Ohio, USA (Dissanayake et al. 2017). However, according to our knowledge, this study is the first to report *Diaporthe longicolla* associated with *Astragalus sinicus* (Farr & Rossman 2022). The phylogenetic placement of this isolate is shown in Fig. 19.

Diaporthe viciae W. Zhao, Q. Ning & J.Y. Yan, sp. nov.

Fig. 20

Index Fungorum Number: IF558423; Facesoffungi number: FoF 10798

Etymology – ‘*viciae*’ refers to the host plant genus *Vicia* from which it was isolated.

Ecology – Associated with healthy stems of *Vicia villosa*

Holotype – JZBH 320179

Asexual morph: Coelomycetous, *Conidiomata* visible as black aggregates up to 150–200 μm high, 150–250 μm diam., superficial, oval to round, black. *Peridium* thick, an inner layer composed of light brown to black *textura angularis*, outer layer composed of dark brown to black *textura angularis*. *Conidiophores* 15–32.5 μm long, cylindrical, aseptate, densely aggregated, apex. *Conidiogenous cells* phialidic, cylindrical, terminal, and lateral. *Alpha conidia* with 2–5 guttules per cell, 7–10 \times 2–4 μm (\bar{x} = 8.3 \times 3.0 μm , $n = 50$), hyaline, fusiform or oval. *Beta conidia* not observed. *Sexual morph*: not observed.

Cultural characteristics – Colonies on PDA white (surface) and yellowish white (reverse), reaching the edge of the plate, with a dense mat of aerial mycelium, covering a 30 mm Petri dish in 5–7 days.

Material examined – China, Guangxi Province, Guilin City, from *Vicia villosa* stem, Zhao Wensheng and Zhang Guozhen, May 2018, (JZB 320179, holotype inactive dry culture), ex-type living culture = JZB 320179.

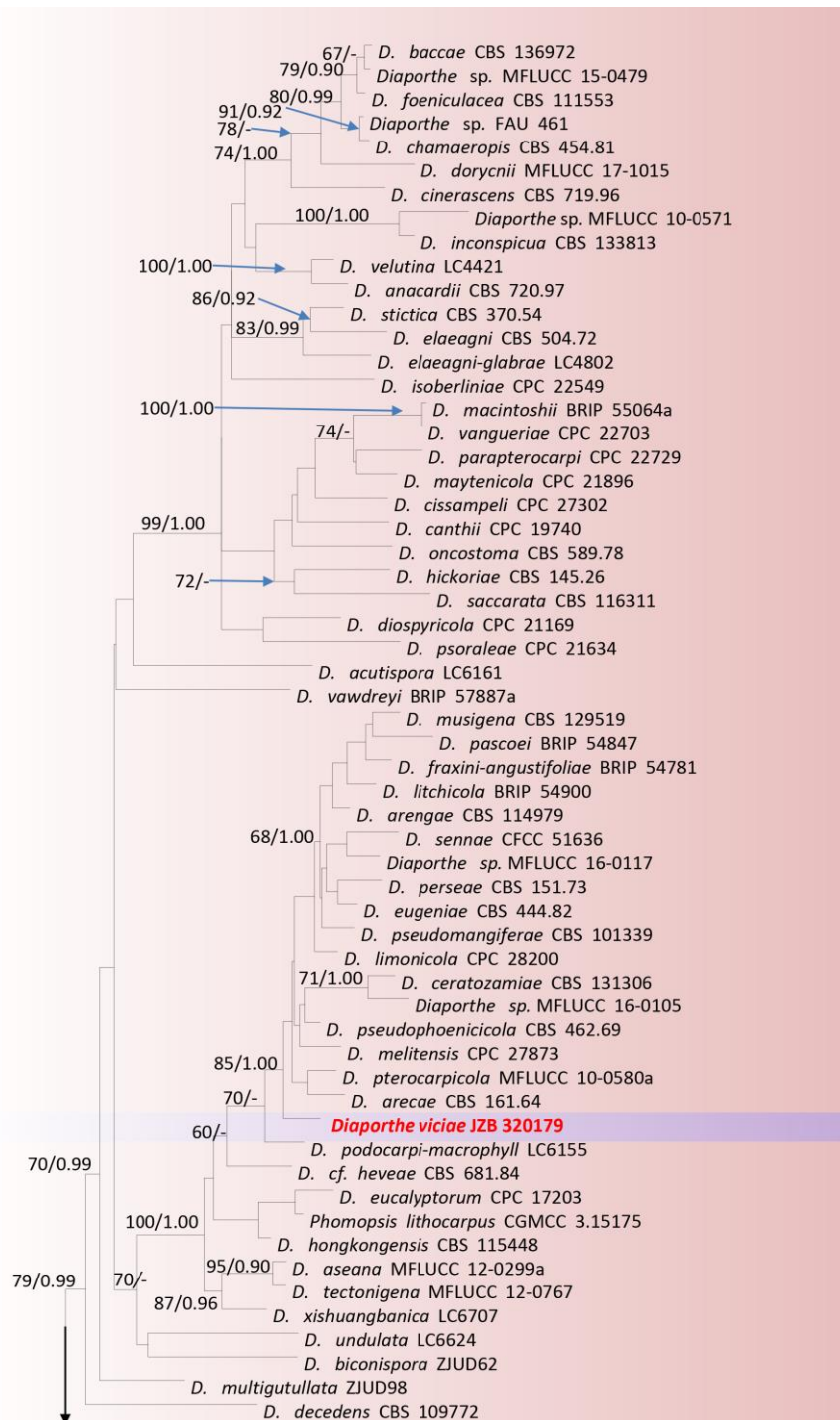


Figure 19 – Phylogram generated from maximum likelihood analysis based on combined ITS, *his*, *tub*, *cal* and *tef1- α* sequence data. The matrix had 1704 distinct alignment patterns, with 26.81% undetermined characters or gaps. Estimated base frequencies were as follows: A = 0.213108, C = 0.327951, G = 0.235405, T = 0.223536; substitution rates AC = 1.156812, AG = 3.661220, AT = 1.256657, CG = 0.910792, CT = 4.756686, GT = 1.000000; gamma distribution shape parameter α = 0.868055. Bootstrap values for maximum likelihood and maximum parsimony equal to or greater than 60% and Bayesian posterior probabilities equal or greater than 0.90 are placed above the branches. The newly generated sequences are indicated in red. We obtained two strains of

Diaporthe. Among them one strain was identified as *Diaporthe longicolla* on *Astragalus sinicus* and the other strain *Diaporthe viciae* sp. nov., from *Vicia villosa* is described herein.

Notes – Isolate (JZB 320179) was recovered from *Vicia villosa* stems from Guangxi Province. This fits the concept of *Diaporthe*. Multi-marker analysis for *Diaporthe viciae* using ITS, *his*, *tub*, *cal*, and *tef-1 α* markers produced a sister clade to *Diaporthe podocarpi-macrophylli* (LC 6155) with 85% ML support (Fig. 19). Comparison between the ex-type strain of *D. podocarpi-macrophylli* and *D. viciae* revealed different conidiomatal and conidial characters. *Diaporthe podocarpi-macrophylli* produced relatively larger conidiomata than *D. viciae* (222–699 μm diam. vs 150–250 μm diam.) and smaller alpha conidia (3.5–8.5 × 1–3 μm vs 7–10 × 2–4 μm). *Diaporthe* is considered as one of the most species-rich genera in *Diaporthaceae*, and most of the species share similar morphological characters (Norphanphoun et al. 2022). Even though species of *Diaporthe* have a wide host range in terrestrial habitats, we could not find any *Diaporthe* species that have been associated with *Vicia villosa* (Farr & Rossman 2022). There were three records in Farr and Rossman database where *Diaporthe* has been recorded from other *Vicia* spp. (*V. fabae* and *V. sativa*) in Australia and Italy. Therefore, from this study, we provide the first host association of *Diaporthe* species on *Vicia villosa*.

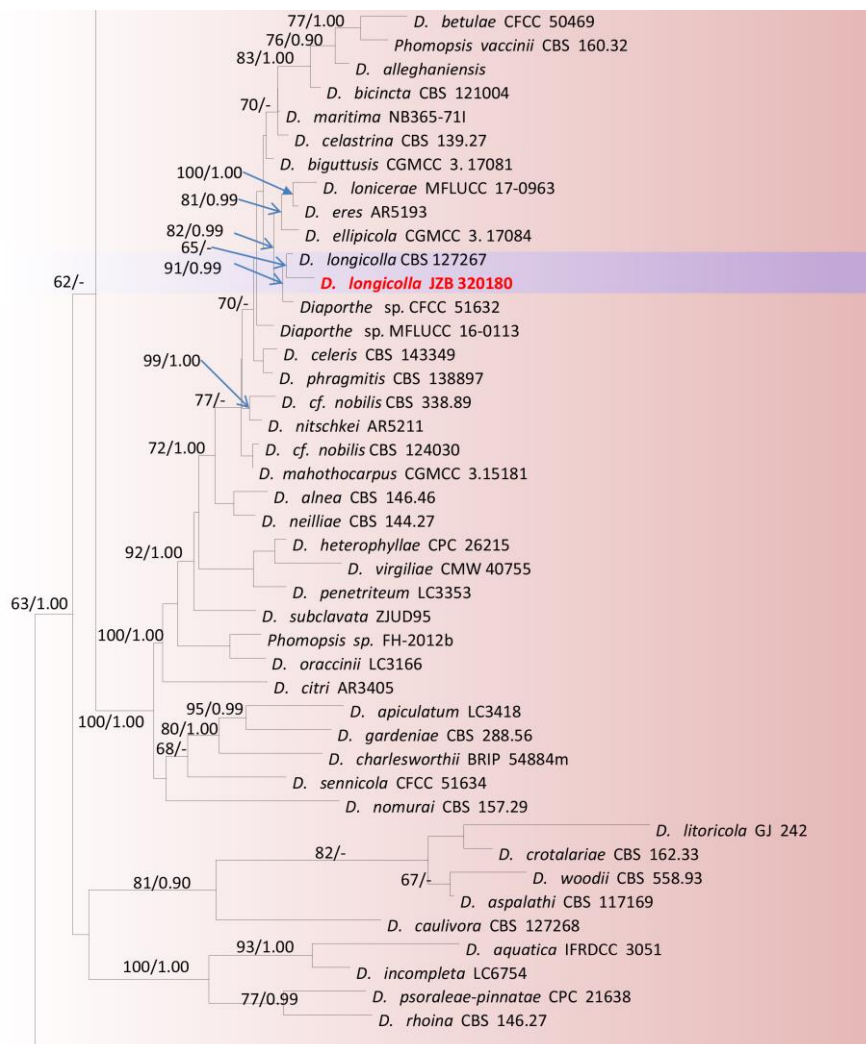


Figure 19 – Continued.

Glomerellales Chadeff. ex Réblová, W. Gams & Seifert (2011).

Glomerellaceae Locq. ex Seifert & W. Gams (2007).

Colletotrichum Corda (1831).

Colletotrichum destructivum O'Gara (1915).

For description see Damm et al. (2014).

Material examined – China, Henan Province, Shihe District, from *Vicia villosa* flower, May 2018, Zhao Wensheng and Zhang Guozhen, living culture = JZB 330198. Henan Province, Shihe District, from *Vicia villosa* leaf, May 2018, Zhao Wensheng and Zhang Guozhen, living culture = JZB 330199.

Notes – Two isolates were recovered from *Astragalus sinicus* and *Vicia villosa* and they were similar and phylogenetically related to *Colletotrichum destructivum* (Bhunjun et al. 2021, Jayawardena et al. 2021). According to Farr & Rossman (2022), *C. destructivum* has been reported from several host plants (*Aster tataricus*, *Bletilla ochracea*, *Cynanchum atratum*, *Echeveria* sp., *Glycine max*, *Helianthus annuus*, *Medicago sativa*, *Nicotiana tabacum*, *Phaseolus limensis*, *Rumex crispus*, *Trifolium repens*) in China. However, according to our knowledge, this study is the first to report *Colletotrichum destructivum* associated with *Astragalus sinicus* and *Vicia villosa* (Farr & Rossman 2022). The phylogenetic placement of these isolates is shown in Fig. 21a.

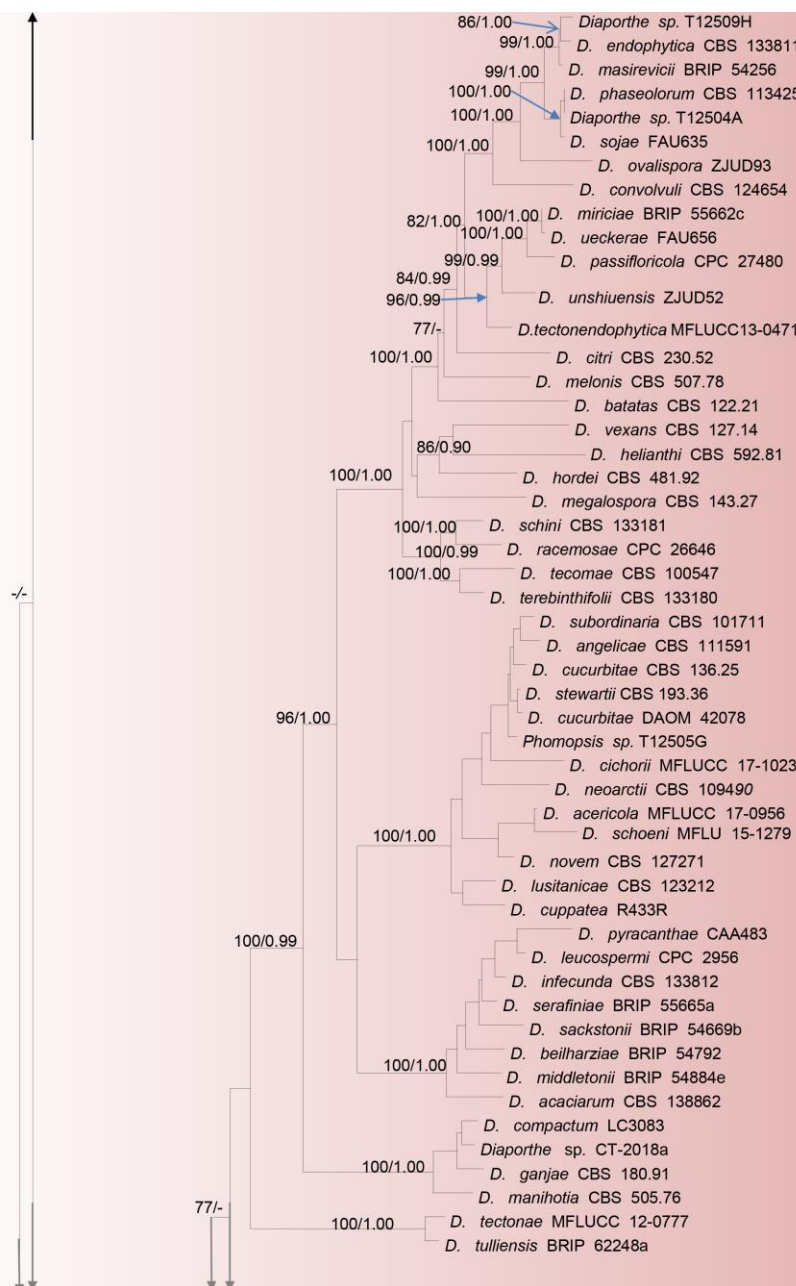


Figure 19 – Continued.

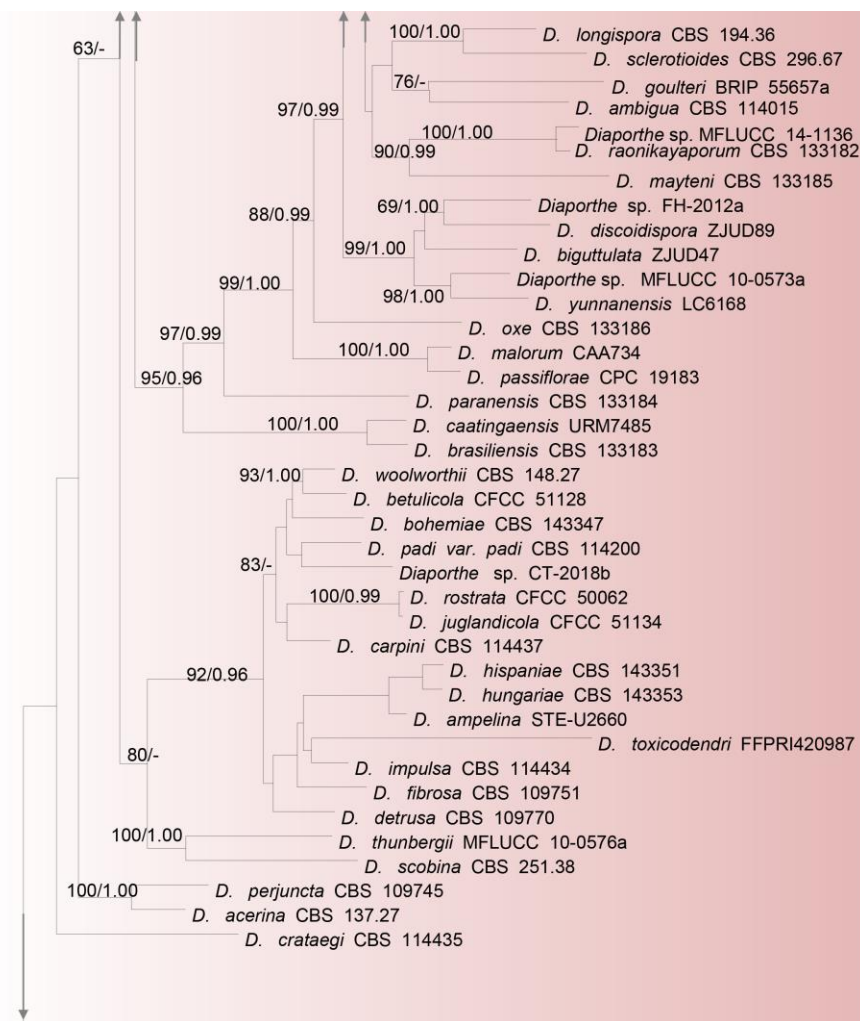


Figure 19 – Continued.

Colletotrichum fructicola Prihastuti, L. Cai & K.D. Hyde (2009).

For description see Prihastuti et al. (2009).

Material examined – China, Guangxi Province, Guilin City, from *Astragalus sinicus* leaf, May 2018, Zhao Wensheng and Zhang Guozhen, living cultures = JZB 330206, JZB 330207, JZB 330208, JZB 330209.

Notes – Four isolates were recovered from *Astragalus sinicus* leaves and they were similar and phylogenetically related to *Colletotrichum fructicola*. *Colletotrichum fructicola* has been clade within the gloeosporioides species complex (Bhunjun et al 2021, Jayawardena et al. 2021). This species has been recorded on many host plants from China (Farr & Rossman 2022), but this study is the first to report *Colletotrichum fructicola* associated with *Astragalus sinicus* (Farr & Rossman 2022). The phylogenetic placement of these isolates is shown in Fig. 21b.

Colletotrichum truncatum (Schwein.) Andrus & W.D. Moore (1935).

For description see Damm et al. (2009).

Material examined – China, Guangxi Province, Nanning City, from *Vicia villosa* leaf, May 2018, Zhao Wensheng and Zhang Guozhen, living cultures = JZB 330200, JZB 330201. Guangxi Province, Nanning City, from *Vicia villosa* root, May 2018, Zhao Wensheng and Zhang Guozhen, living cultures = JZB 330202, JZB 330203. Guangxi Province, Guilin City, from *Astragalus sinicus* stem, May 2018, Zhao Wensheng and Zhang Guozhen, living cultures = JZB 330204, JZB 330205.

Notes – Six isolates were recovered from *Vicia villosa* leaves and roots, as well as *Astragalus sinicus* stems. They are similar and phylogenetically related to *Colletotrichum truncatum*.

Colletotrichum truncatum has a broad host range (Cannon et al. 2012, Damm et al. 2012, Bhunjun et al 2021, Jayawardena et al. 2021, Farr & Rossman 2022) and this species was identified as the causative organism of anthracnose in chili throughout Asia, Australia, and South America (Sharma et al. 2014, Diao et al. 2017, Mongkolporn & Taylor 2018). Our collection of *C. truncatum* was found on *Astragalus sinicus* and *Vicia villosa*. According to our knowledge, this study is the first to report *Colletotrichum truncatum* associated with *Astragalus sinicus* and *Vicia villosa* (Farr & Rossman 2022). The phylogenetic placement of these isolates is shown in Fig. 21c.

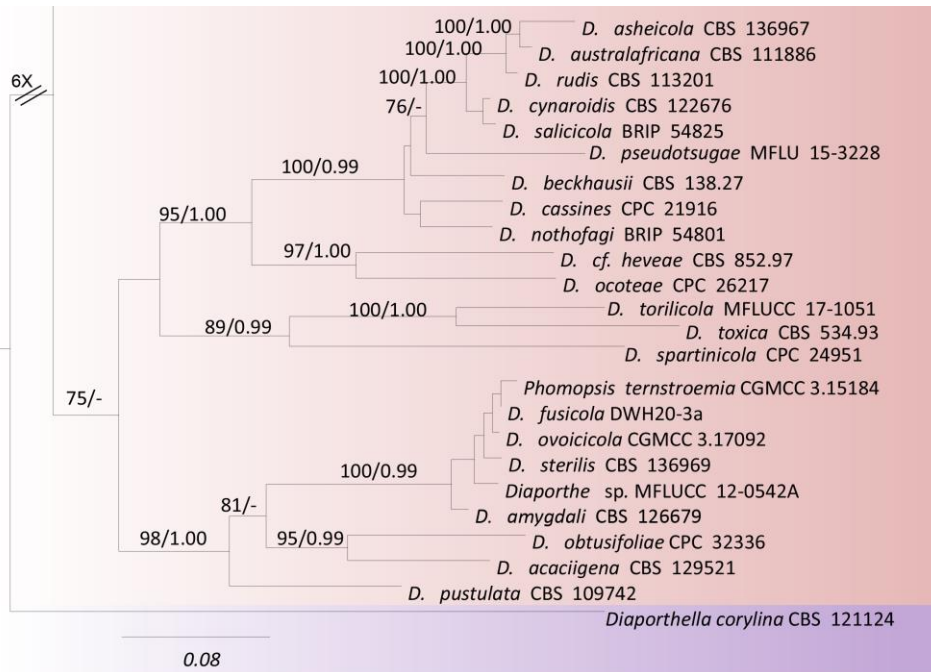


Figure 19 – Continued.

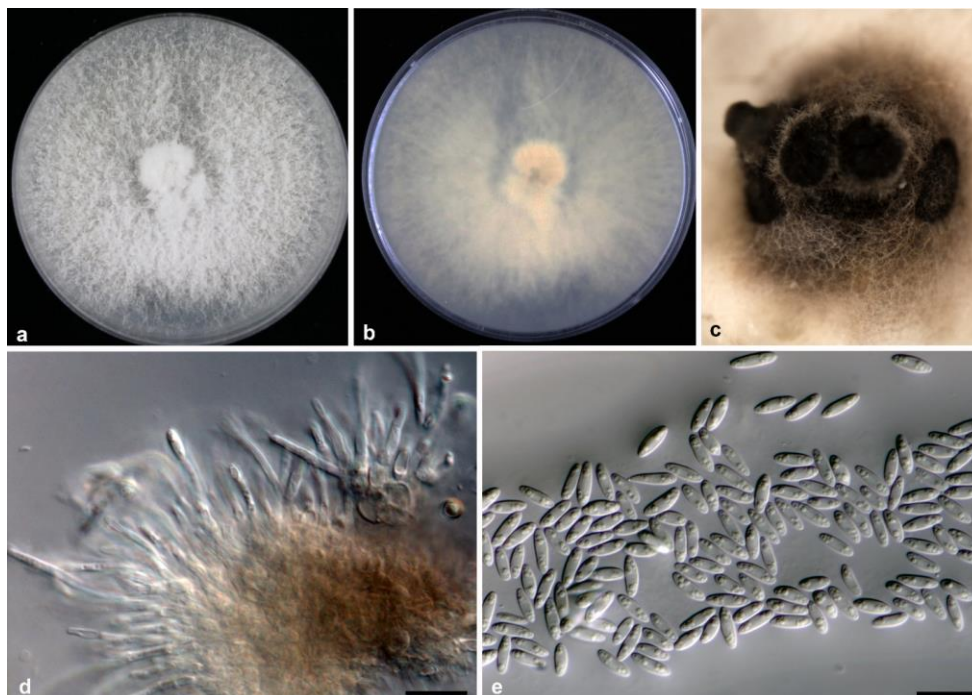


Figure 20 – *Diaporthe viciae*. a Upper view of the colony on PDA. b Back view of the colony on PDA. c Conidiamata on PDA. d Conidia attached to the conidiophores. e Conidia. Scale bars: d, e = 10 μ m.

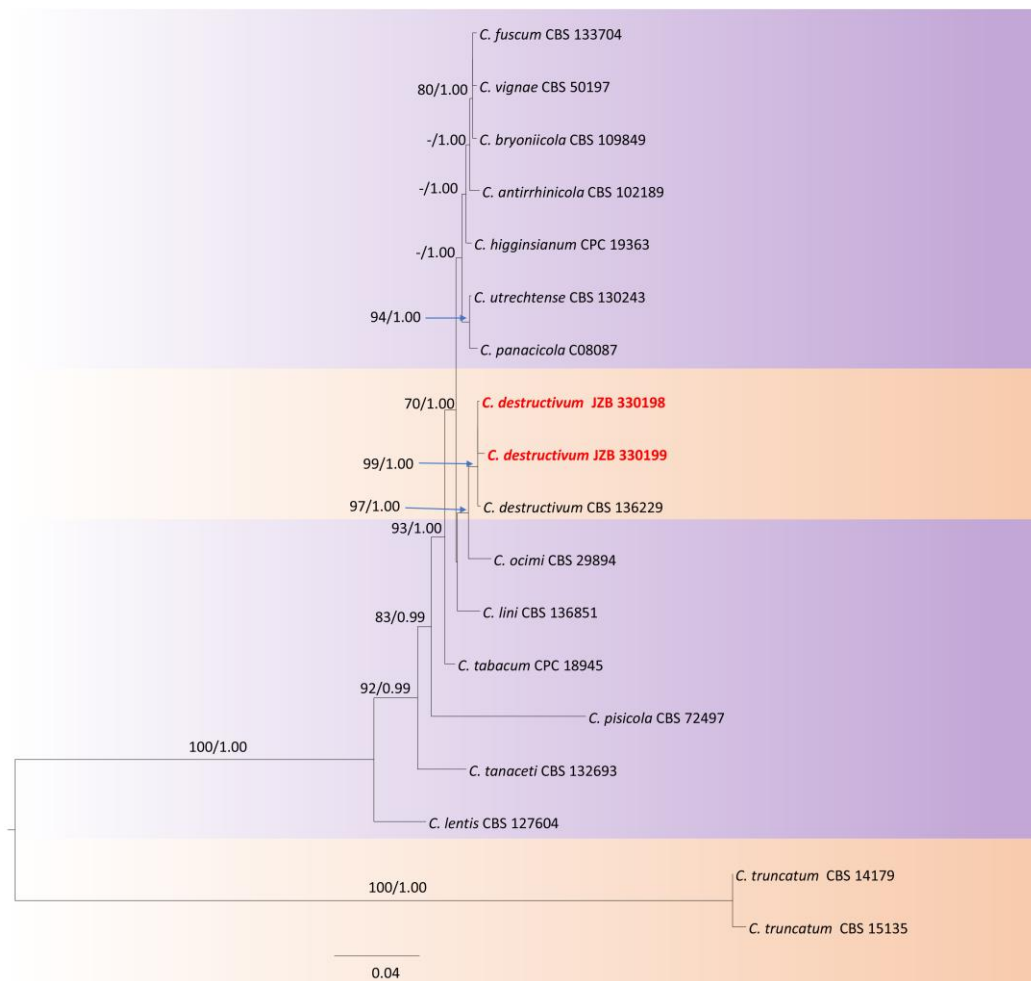


Figure 21a – Phylogram generated for the *Destructivum* species complex from maximum likelihood analysis based on combined ITS, *GAPDH*, *CHS*, *ACT* and *tub* sequence data. The matrix had 290 distinct alignment patterns. Estimated base frequencies were as follows: A = 0.250, C = 0.250, G = 0.250, T = 0.250; substitution rates AC = 1.000000, AG = 2.97679, AT = 1.000000, CG = 1.000000, CT = 5.14921, GT = 1.000000; gamma distribution shape parameter $\alpha = 0.333$. Bootstrap values for maximum likelihood and maximum parsimony equal to or greater than 60% and Bayesian posterior probabilities equal or greater than 0.90 are placed above the branches. The newly generated sequences are indicated in red. We obtained two strains of *Colletotrichum* and identified them as *C. destructivum*. These are new records on *Astragalus sinicus*.

Plectosphaerellaceae W. Gams, Summerb. & Zare (2007).

Plectosphaerella Kleb. (1929).

Plectosphaerella cucumerina (Lindf.) W. Gams, Persoonia 5 (2): 179 (1968).

For description see Domsch et al. (2007) and Carlucci et al. (2012).

Material examined – China, Henan Province, Shihe District, from *Vicia villosa* stem, May 2018, Zhao Wensheng and Zhang Guozhen, living culture JZB = 3540001.

Notes – The isolate recovered from *Vicia villosa* is similar and phylogenetically related to *Plectosphaerella cucumerina*. According to Farr and Rossman (2022), *Plectosphaerella cucumerina* has been recorded on many hosts worldwide and this species was reported from *Brassica oleracea*, *Cucumis sativus*, *Helianthus annuus*, *Lagenaria siceraria*, *Lycopersicon esculentum*, *Phaseolus vulgaris*, *Sedum* sp., *Solanum lycopersicum*, and *Solanum tuberosum* in China (Farr & Rossman 2022). However, according to our knowledge, this study is the first to report *Plectosphaerella cucumerina* associated with *Vicia villosa* (Farr & Rossman 2022). The phylogenetic placement of this isolate is shown in Fig. 22.

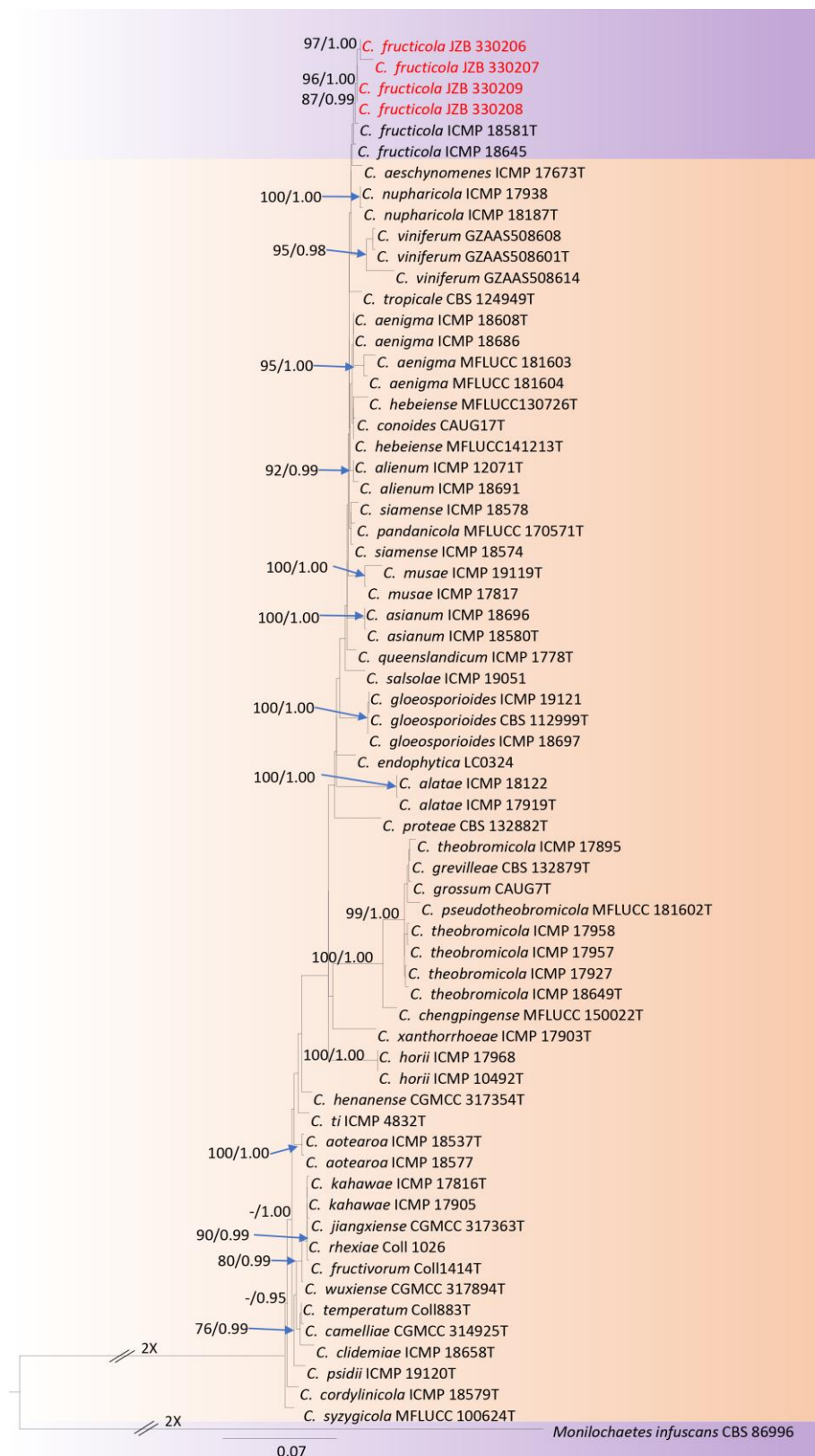


Figure 21b – Phylogram generated for the Gloeosporioides species complex from maximum likelihood analysis based on combined ITS, *GAPDH*, *CHS*, *ACT* and *tub* sequence data. The matrix had 628 distinct alignment patterns, with 13.66% undetermined characters or gaps. Estimated base frequencies were as follows: A = 0.230712, C = 0.302046, G = 0.243533, T = 0.223708; substitution rates AC = 1.114082, AG = 2.699988, AT = 0.747762, CG = 0.697623, CT = 4.725125, GT = 1.000000; gamma distribution shape parameter $\alpha = 0.711937$. Bootstrap values for maximum likelihood and maximum parsimony equal to or greater than 60% and Bayesian posterior

probabilities equal or greater than 0.90 are placed above the branches. The newly generated sequences are indicated in red. We obtained four strains of *Colletotrichum*, which were identified as *Colletotrichum fructicola*. These are new records on *Astragalus sinicus* in China.

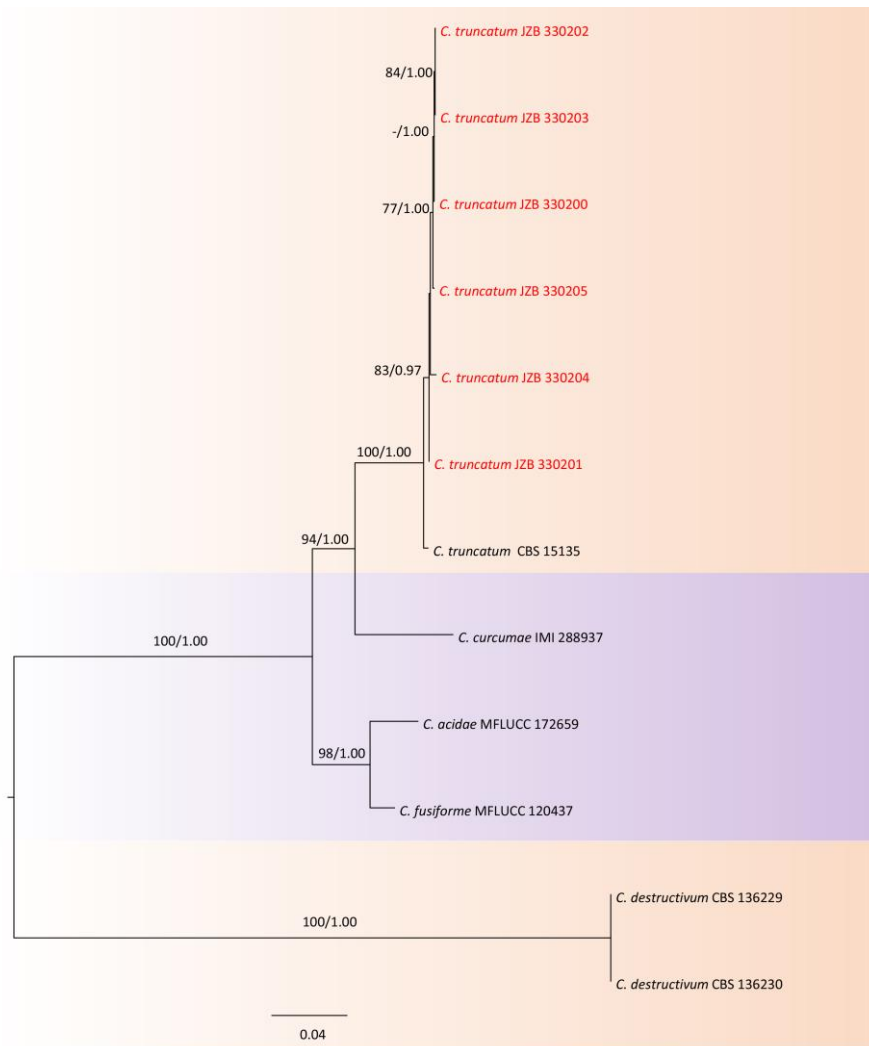


Figure 21c – Phylogram generated for the Truncatum species complex from maximum likelihood analysis based on combined ITS, *GAPDH*, *CHS*, *ACT* and *tub* sequence data. The matrix had 252 distinct alignment patterns. Estimated base frequencies were as follows: A = 0.250, C = 0.250, G = 0.250, T = 0.250; substitution rates AC = 1.00000, AG = 3.71746, AT = 1.00000, CG = 1.00000, CT = 3.71746, GT = 1.000000; gamma distribution shape parameter $\alpha = 0.274$. Bootstrap values for maximum likelihood and maximum parsimony equal to or greater than 60% and Bayesian posterior probabilities equal or greater than 0.90 are placed above the branches. The newly generated sequences are indicated in red. We obtained six strains of *Colletotrichum*, and they were identified as *C. truncatum*. These are new records on *Astragalus sinicus* and *Vicia villosa* in China.

Hypocreales Lindau (1897).

Bionectriaceae Samuels & Rossman (1999).

Clonostachys Corda (1839).

Clonostachys eriocamporesii R.H. Perera & K.D. Hyde (2020).

For description see Hyde et al. (2020).

Material examined – China, Henan Province, Shihe District, from *Vicia villosa* root, May 2018, Zhao Wensheng and Zhang Guozhen, living culture = JZB 3530004.

Notes – The isolate was recovered from *Vicia villosa* roots and is similar and phylogenetically related to *Clonostachys eriocamporesii*. The recently introduced *Clonostachys eriocamporesii* was recorded on *Pennisetum polystachion* in Thailand (Hyde et al. 2020) and our collection was found on *Vicia villosa*. According to our knowledge, this study is the first to report *Clonostachys eriocamporesii* associated with *Vicia villosa* (Farr & Rossman 2022). The phylogenetic placement of these isolates is shown in Fig. 23.

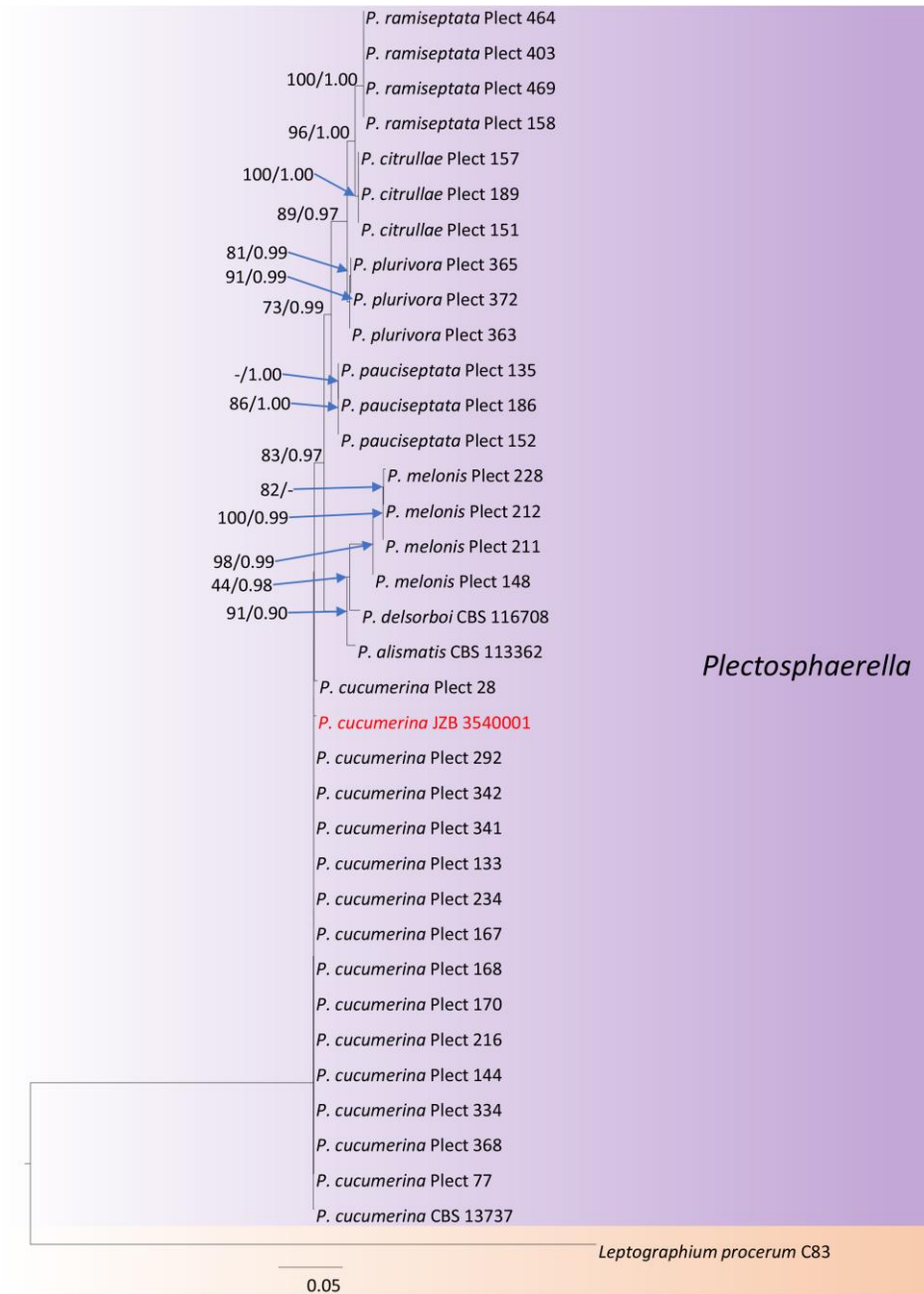


Figure 22 – Phylogram generated for *Plectosphaerella* species from maximum likelihood analysis based on combined LSU and ITS sequence data. The matrix had 125 distinct alignment patterns, with 16.50% undetermined characters or gaps. Estimated base frequencies were as follows: A = 0.219652, C = 0.278630, G = 0.282925, T = 0.218793; substitution rates AC = 1.092661, AG = 3.061387, AT = 0.968115, CG = 0.222991, CT = 8.313793, GT = 1.000000; gamma distribution shape parameter $\alpha = 0.527914$. Bootstrap values for maximum likelihood and maximum parsimony equal to or greater than 60% and Bayesian posterior probabilities equal or greater than 0.90 are placed above the branches. The newly generated sequences are indicated in red. We obtained a

strain of *Plectosphaerella*, which was identified as *Plectosphaerella cucumerina* which is a new record on *Vicia villosa* in China.

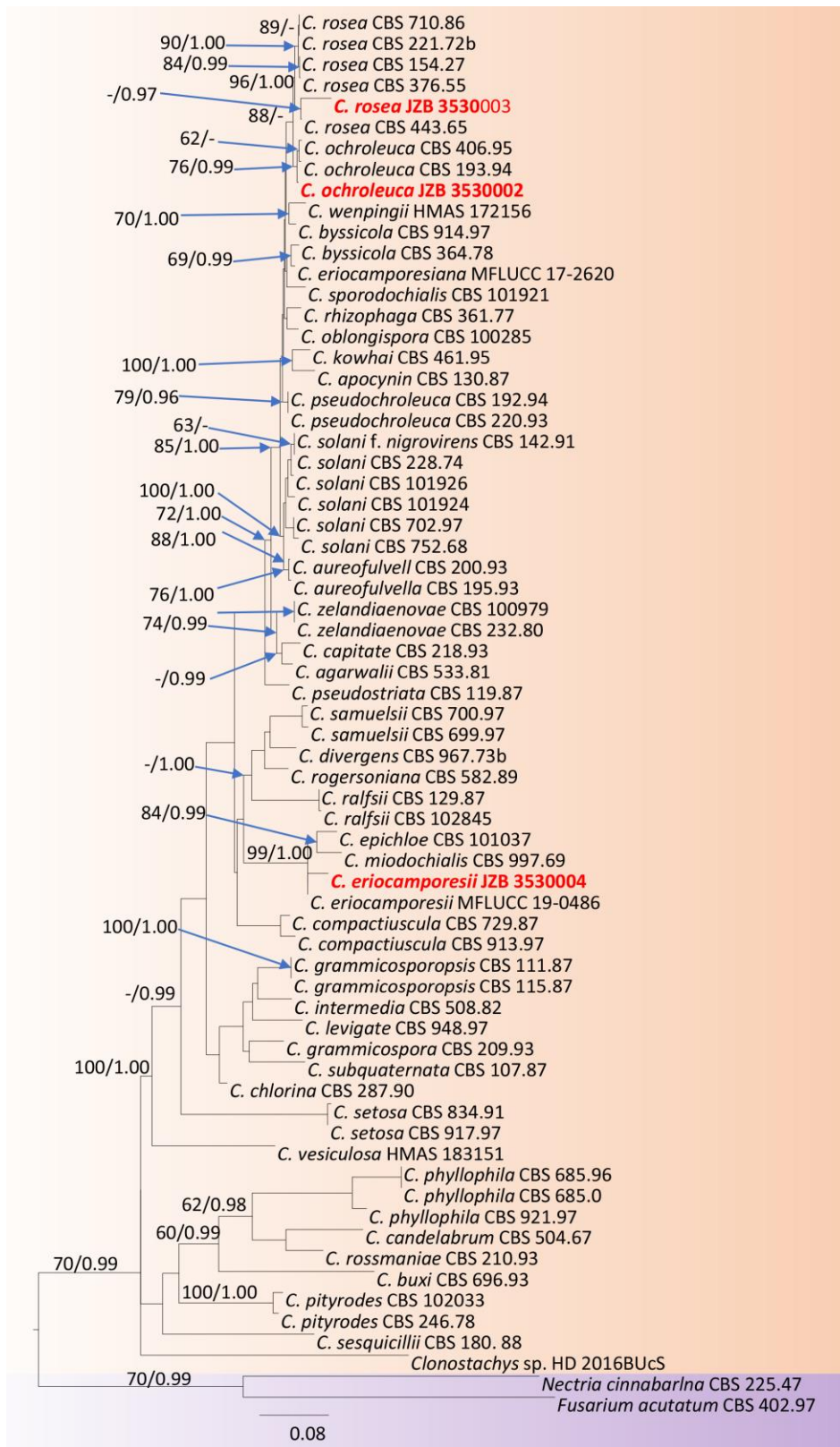


Figure 23 – Phylogram generated for *Clonostachys* species from maximum likelihood analysis based on combined ITS and *tub* sequence data. The matrix had 464 distinct alignment patterns, with 15.79% undetermined characters or gaps. Estimated base frequencies were as follows: A = 0.211586, C = 0.281640, G = 0.252623, T = 0.254151; substitution rates AC = 1.242565, AG =

3.469354, AT = 1.462245, CG = 0.573693, CT = 4.537269, GT = 1.000000; gamma distribution shape parameter $\alpha = 0.699805$. Bootstrap values for maximum likelihood and maximum parsimony equal to or greater than 60% and Bayesian posterior probabilities equal or greater than 0.90 are placed above the branches. The newly generated sequences are indicated in red. We obtained three strains of *Clonostachys*, which were identified as new records; *C. eriocamporesii*, *C. rosea*, and *C. ochroleuca*.

Clonostachys ochroleuca (Schwein.) Schroers & Samuels (1997).

For description see Schroers and Samuels (1997).

Material examined – China, Henan Province, Shihe District, from *Vicia villosa* pods, May 2018, Zhao Wensheng and Zhang Guozhen, living culture = JZB 3530002.

Notes – The isolate was recovered from *Vicia villosa* pods and is similar and phylogenetically related to *Clonostachys ochroleuca*. *Clonostachys ochroleuca* is previously known as *Bionectria ochroleuca* and it has been recorded on many hosts worldwide (Farr & Rossman 2022). This is the first to report *C. ochroleuca* associated with *Vicia villosa* (Farr & Rossman 2022). The phylogenetic placement of these isolates is shown in Fig. 23.

Clonostachys rosea (Link) Schroers, Samuels, Seifert, & W. Gams (1999).

For description see Schroers et al. (1999).

Material examined – China, Henan Province, Shihe District, from *Astragalus sinicus* roots, May 2018, Zhao Wensheng and Zhang Guozhen, living culture = JZB 3530003.

Notes – The isolate was recovered from *Astragalus sinicus* and is similar and phylogenetically related to *Clonostachys rosea*. According to Farr and Rossman (2022), *Clonostachys rosea* has been recorded on many host plants worldwide. Previously, *Clonostachys rosea* was recorded from *Vitis* sp. in China (Jayawardena et al 2018). This is the first to report *Clonostachys rosea* associated with *Astragalus sinicus* (Farr & Rossman 2022). The phylogenetic placement of these isolates is shown in Fig. 23.

Stachybotryaceae L. Lombard & Crous (2014).

Albifimbria L. Lombard & Crous (2016).

Albifimbria verrucaria (Alb. and Schwein.) L. Lombard & Crous (2016).

For description see Lombard et al. (2016)

Material examined – China, Guangxi Province, Guilin City, from *Astragalus sinicus* pods, May 2018, Zhao Wensheng and Zhang Guozhen, living culture = JZB 3510001, JZB 3510002.

Notes – Two isolates were recovered from *Astragalus sinicus* pods and they were similar and phylogenetically related to *Albifimbria verrucaria*. According to Farr and Rossman (2022), *Albifimbria verrucaria* has been recorded on few host plants (*Cucurbita* sp., *Diplotaxis* sp., *Solanum* sp., *Spinacia* sp., *Valerianella* sp., and *Vitis* sp.) from China, Cyprus, Italy, and Tunisia. Our collection was found on *Astragalus sinicus* and this is the first to report *Albifimbria verrucaria* associated with *Astragalus sinicus* (Farr & Rossman 2022). The phylogenetic placement of these isolates is shown in Fig. 24.

Identification of *Fusarium* isolates

In total, 381 isolates were assigned to *Fusarium* species with high certainty. A further 370 isolates were determined to have seven complexes (fujikuroi species complex; 54 strains, incarnatum-equiseti species complex; 34 strains, nisikadoi complex; 30 strains, oxysporum species complex; 16 strains, sambucinum species complex; 44 strains, solani species complex; 5 strains, tricinctum species complex; 132 strains) and one subclade (Asian subclade; 54 strains). Eleven isolates could not be assigned to any level (Supplementary Table 2). In this study, *Fusarium* isolates could not be confirmed by multi-markers phylogeny to species level because of the high plasticity of species boundaries (Leslie et al. 2007, O'Donnell et al. 2009, 2013). The genetic

diversity of *Fusarium* is complex, and taxa have high genetic variability within morphologically defined species.

To resolve the phylogenetic relationship among *Fusarium* species, it is recommended that the need of construct a reliable taxonomic system based on the combination of morphological, molecular, toxicological, and biological data (Leslie & Bowden 2008, Watanabe 2013, Walder et al. 2017).

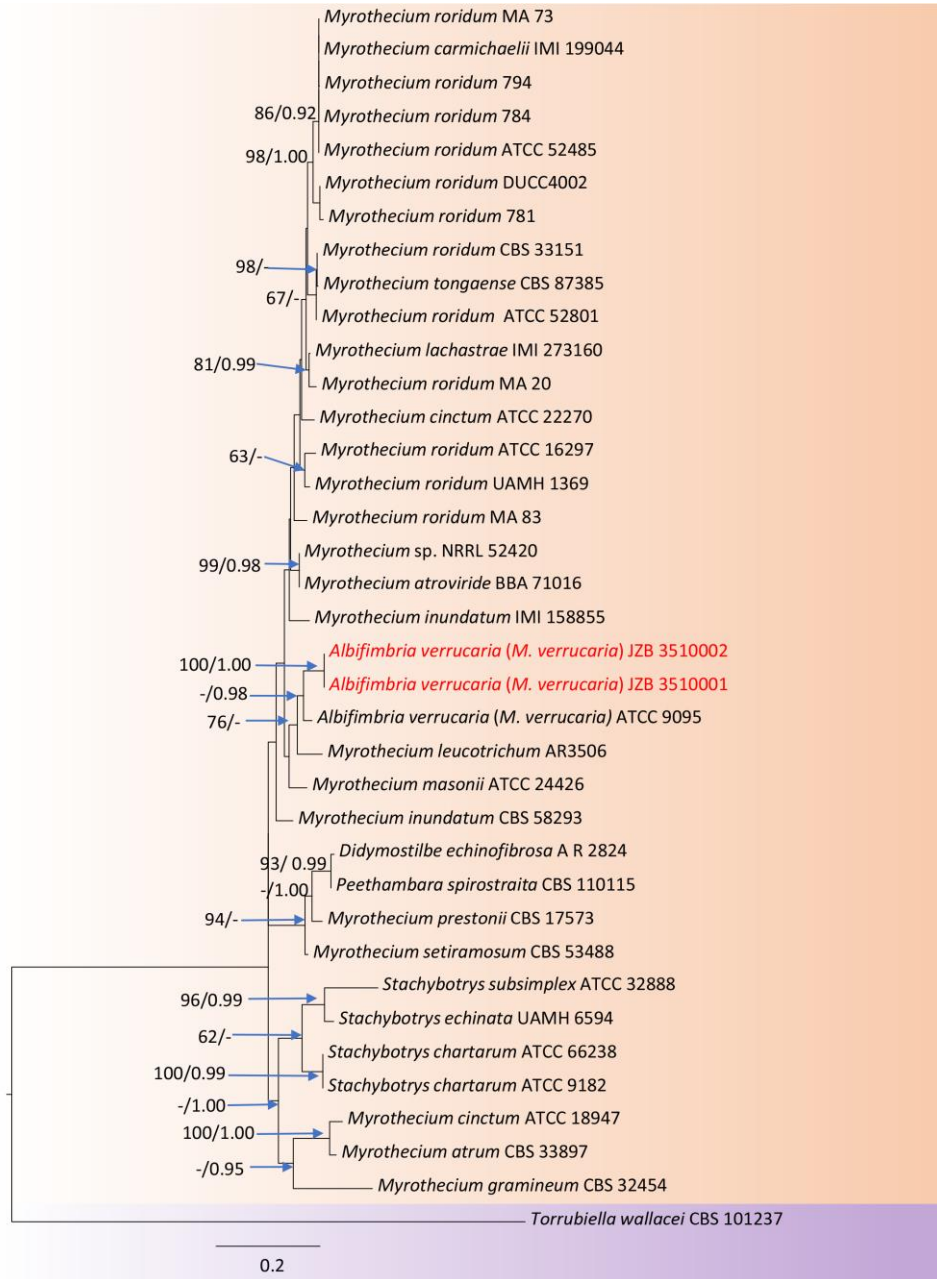


Figure 24 – Phylogram generated from maximum likelihood analysis based on combined ITS and *tefl-a* sequence data. The matrix had 471 distinct alignment patterns, with 39.34% undetermined characters or gaps. Estimated base frequencies were as follows: A = 0.213797, C = 0.325466, G = 0.251434, T = 0.209303; substitution rates AC = 1.182710, AG = 1.437422, AT = 1.210215, CG = 1.026837, CT = 5.250391, GT = 1.000000; gamma distribution shape parameter α = 0.396388. Bootstrap values for maximum likelihood equal to or greater than 60% and Bayesian posterior probabilities equal or greater than 0.90 are placed above the branches. We obtained two strains of *Albifimbria* which were identified as *Albifimbria verrucaria*, which is a new record on *Astragalus sinicus* in China.

Diversity of fungal communities from High-Throughput Sequencing

Table 3 Summary of OTU assigned

Phylum*	Class	Order	Family	Genus	Species
5 known	21 known	48 known	66 known	74 known	61 known
1 others	11 others	22 others	40 others	52 others	74 others
1 _	6 _	8 _	9 _	11 _	38 _

* The word “others” represents the OTUs that were not assigned to certain taxa. The symbol “_” stands for the OTUs that were not well classified.

Fungal abundance, community composition and diversity

A total of 2,260,349 clean reads of 18S rRNA V4 amplicon were generated from 30 samples covering two manure crops of *Astragalus sinicus* and *Vicia villosa* collected from Central (Henan Province) and Southern China (Fujian and Guangxi Provinces) respectively. Many of these sequences were annotated as Eukaryota and Metazoa. Unclassified sequences and the sequences that did not belong to the fungi were removed and obtained 178 fungal OTUs (Table 3 and Supplementary Table 4). Even though the fungal sequence reads per sample obtained in this study were relatively low, the rare-fraction curve showed that the number of species basically reached saturation for all samples. This indicates that the data is representative (Fig. 25).

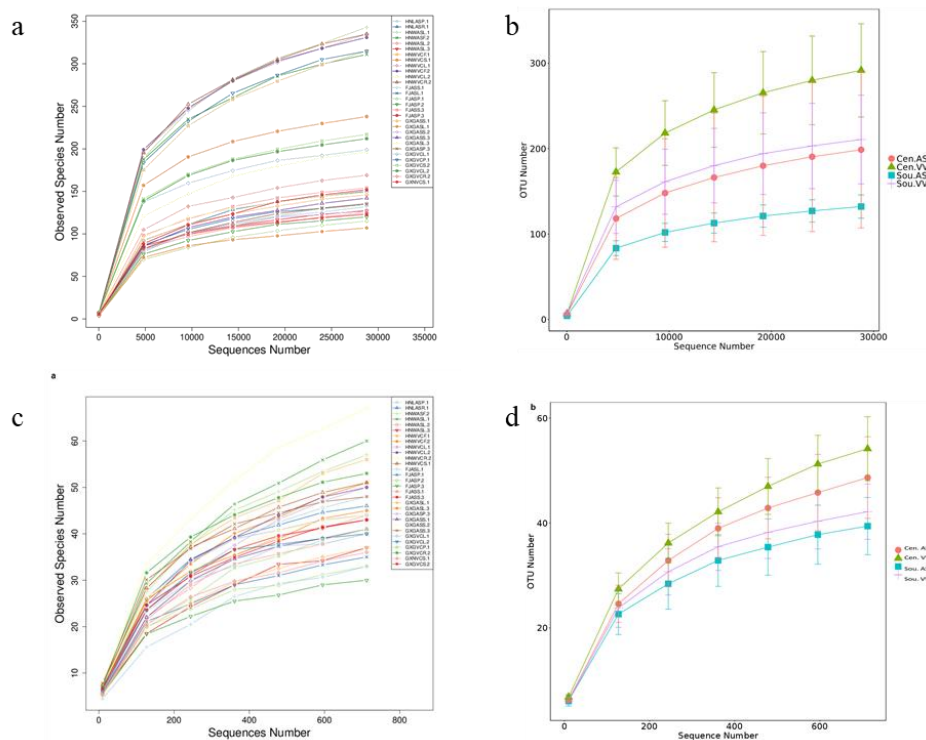


Figure 25 – (a) and (b) Rarefaction curves of each sample and group based on all the 18S rRNA sequences. (c) and (d) Rarefaction curves of each sample and group based on the fungal sequences (Cen. AS- *Astragalus sinicus* samples collected from Central China, Cen. VV- *Vicia villosa* samples collected from Central China, Sou. AS- *Astragalus sinicus* samples collected from South China, Sou.VV- *Vicia villosa* samples collected from South China).

In this study, we analyzed OTU richness and Shannon diversity. Fungal OTU richness was not significantly different between the two crops tested in this study, ranging from 26–52 (38.78 ± 7.29 (mean \pm SD; *Astragalus sinicus*) and 33–60 (42.92 ± 7.10 (mean \pm SD; *Vicia villosa*) ($P = 0.143$, Wilcoxon test). Shannon diversity also showed a similar trend ranging from 2.81–4.47

(3.63 ± 0.49 (mean \pm SD; *Astragalus sinicus*) and $3.02\text{--}4.53$ (3.77 ± 0.48 (mean \pm SD; *Vicia villosa*) ($P = 0.440$, Wilcoxon test; Fig. 26). Significant differences were showed in observed OTU richness between *Vicia villosa* samples collected from the South or Central China ($P = 0.026$, Wilcoxon test; Fig. 26).

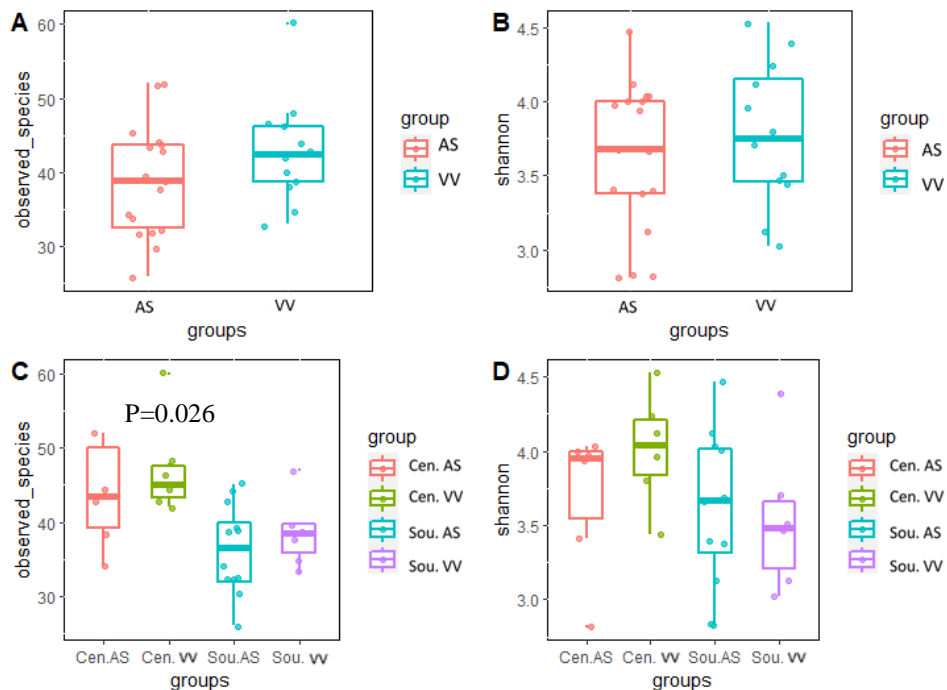


Figure 26 – The barplot showing the comparison of OTU richness and Shannon index between each group (AS- *Astragalus sinicus*, VV-*Vicia villosa*, Cen. AS- *Astragalus sinicus* samples collected from Central China, Cen. VV- *Vicia villosa* samples collected from Central China, Sou. AS- *Astragalus sinicus* samples collected from South China, Sou. VV- *Vicia villosa* samples collected from South China).

Alpha diversity indices are given in Supplementary Table 5. In total, we detected 178 fungal OTUs (Supplementary Table 4). There were 103 fungal OTUs shared between the two crops while 25 and 18 OTUs were specific to *Astragalus sinicus* and *Vicia villosa*, respectively. Twelve, 7, 12 and 10 OTUs were specific to Cen. AS, Cen. VV, Sou. AS and Sou. VV, respectively (Fig. 27).

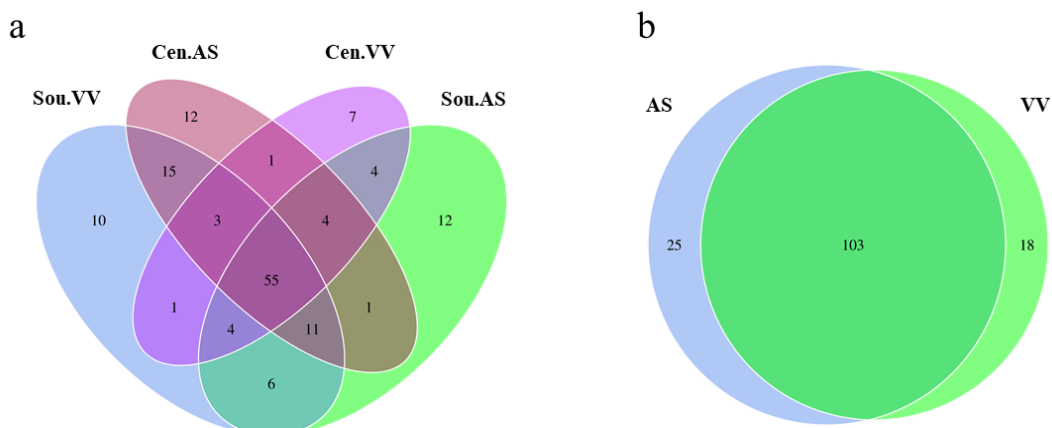


Figure 27 – Venn diagrams show the distribution of OTUs across different groups (AS- *Astragalus sinicus*, VV-*Vicia villosa*, Cen. AS- *Astragalus sinicus* samples collected from Central China, Cen. VV- *Vicia villosa* samples collected from Central China, Sou. AS- *Astragalus sinicus* samples

collected from South China, Sou. VV- *Vicia villosa* samples collected from South China).

In *Astragalus sinicus*, members of *Ascomycota* were commonly detected accounting for 85% of total sequences, and *Basidiomycota* were accounting for 12.5%. In *Vicia villosa*, most of the sequences were also assigned to *Ascomycota* (70%) and followed by *Basidiomycota* (28%). The top five abundant orders are *Pleosporales* (34%), *Capnodiales* (23%), *Magnaporthales* (9%), *Glomerellales* (8%) and *Helotiales* (4.5%) for *Astragalus sinicus*. *Pleosporales* (20%), *Capnodiales* (19%), *Helotiales* (15%), *Cantharellales* (11%) and *Filobasidiales* (8%) for *Vicia villosa*. The most common species of these green manure crops based on OTU data is *Cladosporium herbarum*. The relative abundance of the top ten phyla, classes, order, families, genera, and species from different samples of the two crops are shown in Supplementary Fig. 1-6.

Table 4 Results of Adonis Bray- Curtis analysis.

Vs_group*	F. Model	R ²	Pr(>F)
AS-VV	2.4144	0.07938	0.007
Sou.AS- Sou.VV	1.7651	0.09936	0.082
Sou.AS-Cen.AS	2.6015	0.13986	0.008
Cen.VV-Sou. VV	2.8306	0.22061	0.003
Cen.VV-Cen.AS	1.6935	0.14482	0.117

* Cen. AS- *Astragalus sinicus* samples collected from Central China, Cen. VV- *Vicia villosa* samples collected from Central China, Sou. AS- *Astragalus sinicus* samples collected from South China, Sou. VV- *Vicia villosa* samples collected from South China.

To explore the differences in fungal community structure and composition correlated with sampling location and crop types, we computed the beta-diversity analysis based on the Bray-Curtis distance. Samples of *Astragalus sinicus* and *Vicia villosa* clustered respectively and showed a clear distinction in the PCoA and NMDS ($P = 0.007$, PERMANOVA test) (Fig. 28a, b, Table 4). The samples of *Astragalus sinicus* and *Vicia villosa* obtained from south China were closely clustered (Fig. 28c, d) and showed no significant difference (Fig. 28c, d, $P = 0.082$ PERMANOVA test). Similar results were observed for the samples obtained from central China as well (Fig. 28c, d, $P = 0.117$, PERMANOVA test).

Samples of *Astragalus sinicus* collected from south and central China clustered significantly far apart (Fig. 28c, d, $P = 0.008$, PERMANOVA test) and also it same for the *Vicia villosa* samples as well (Fig. 28c, d, $P = 0.003$, PERMANOVA test). The R^2 of PERMANOVA results indicates the degree of interpretation of the difference between the different groups. The R^2 of Sou. AS-Cen. AS group pair (0.14) is greater than that of Sou. AS- Sou. VV (0.10). And the R^2 of Cen. VV-Sou. VV group pair (0.22) is greater than that of Cen. VV-Cen. AS (0.14) (Table 4). A higher R^2 indicates a higher degree of explanation for the difference between the groups. Hence, these results suggested that the sampling location has a greater influence on the fungal community structures of *Astragalus sinicus* and *Vicia villosa*.

Linear discriminant analysis (LDA) and effect size (LEfSe) analysis was used to further investigate the fungal biomarkers with distinct relative abundances between *Astragalus sinicus* and *Vicia villosa* sampled in the south and central China (Fig. 29). *Basidiomycota* and *Hypocreales* sp. were enriched in Sou. VV samples. Ten taxa including Dothideomycetes and *Ascomycota* were enriched in Sou. AS samples. Twelve taxa including *Leotiomycetes* and *Helotiales* were enriched in Cen. VV samples. Five taxa including *Magnaporthe* and *Glomeromycetes* were enriched in Cen. AS samples (Fig. 29).

In addition, a T-test between groups was performed to find the taxa with significant differences (p -value < 0.05). *Alternaria* and *Sporisorium* had a significant relative abundance in *Astragalus sinicus* and *Sclerotinia* was more abundant in *Vicia villosa* ($p < 0.05$) (Fig. 30).

Simper analysis showed the top ten taxa with the highest contribution to the differences in fungal community structure between *Astragalus sinicus* and *Vicia villosa* (Fig. 31). *Rhizoctonia* had

the highest contribution with 14%, followed by *Cladosporium* (13%), *Alternaria* (10%), *Magnaporthe* (9%), *Sclerotinia* (8%), *Boeremia* (7%), *Plectosphaerella* (7%), *Filobasidium* (7%), *Fusarium* (4%) and *Cystofilobasidium* (3%) (Fig. 31).

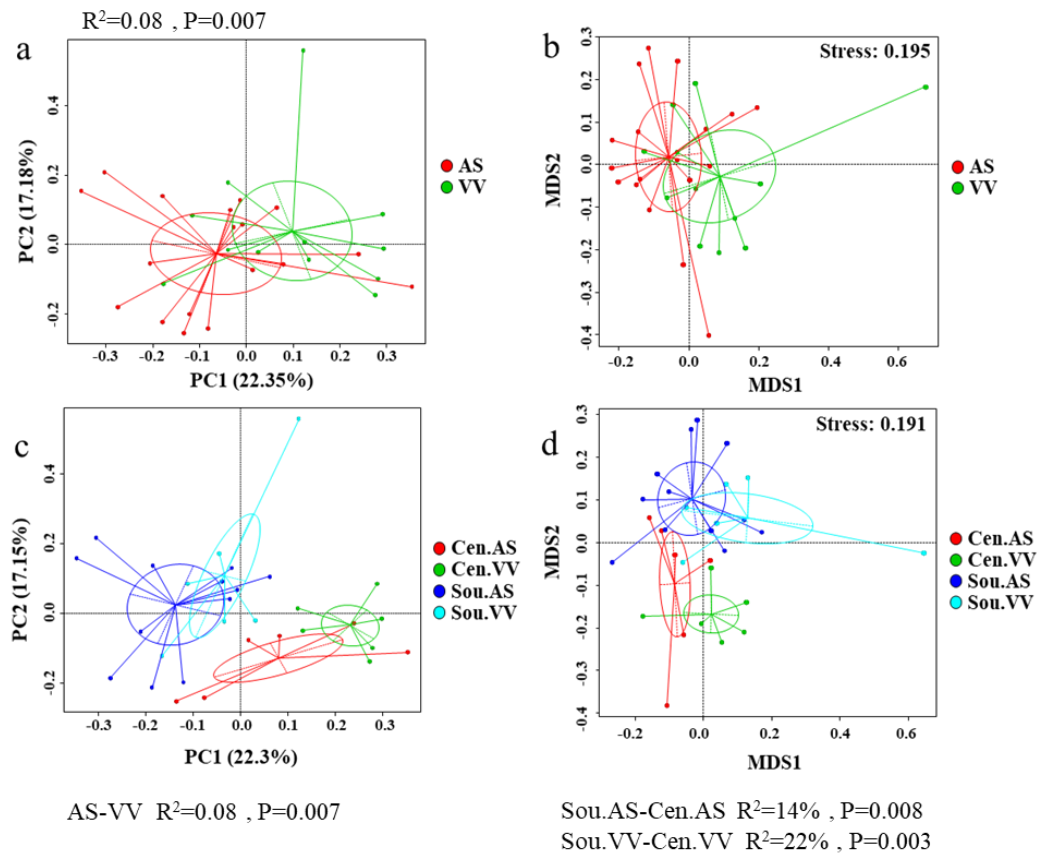


Figure 28 – (a) and (b) are the PCoA and NMDS plots showing the clustering of samples of *Astragalus sinicus* (AS) and *Vicia villosa* (VV), respectively. (c) and (d) are the PCoA and NMDS showing the clustering of samples of *Astragalus sinicus* collected from central (Cen. AS) and south China (Sou. AS), samples of *Vicia villosa* collected from central (Cen.VV) and south China (Sou. VV). (All the plots were plotted based on the Bray–Curtis distance. The PERMANOVA test was used to do the statistical analysis. And the R^2 and P values of group pairs with significant differences were shown below).

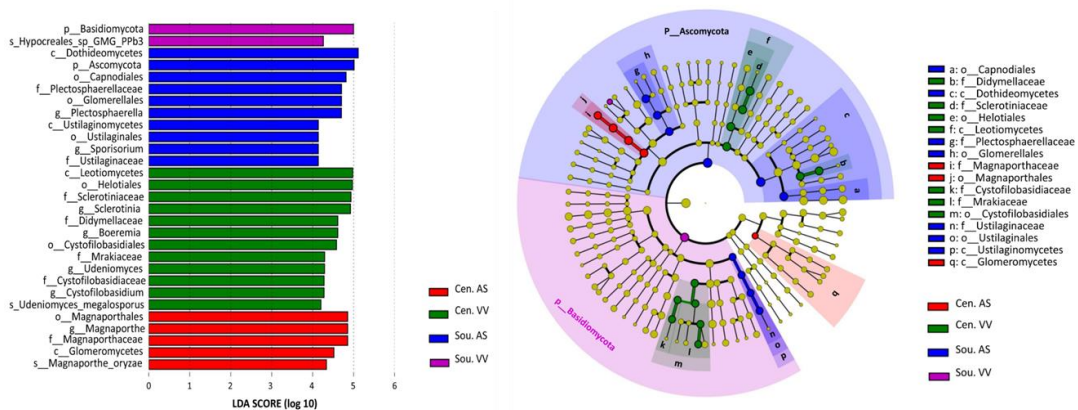


Figure 29 – LefSe analysis of fungal enrichment at different classification levels (p: Phylum, c: Class, o: Order, f: Family, g: Genus, s: Species) among different sample groups. Fungal biomarker

enrichment among groups with LDA value >4 is shown in histogram (a) and evolutionary branching diagram (b).

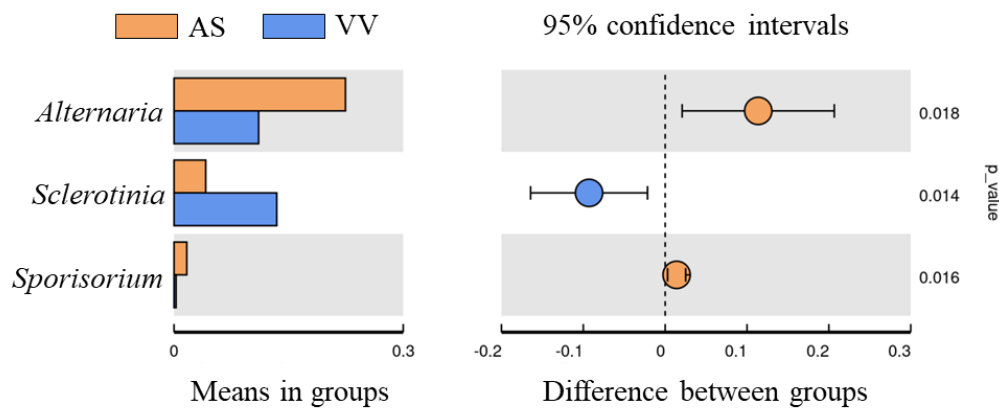


Figure 30 – Genera with significant differences between *Astragalus sinicus* and *Vicia villosa* tested by T-test.



Figure 31 – Top 10 genera with the highest contribution to the differences in fungal community structure between *Astragalus sinicus* and *Vicia villosa* based on Simper analysis.

Functional annotation of fungi

The co-occurrence relationship of microorganisms in different environments is completely different and a network map of genera here is used to visually understand the networks present in the environment (Supplementary Figs 7, 8). The genera with high relative abundance or dominance often played unique or important roles in maintaining the stability of microbial community structure and the functions of the environment. *Eremothecium*, *Chaetospermum*, *Acaulospora*, *Torula*, *Funneliformis*, *Leucosporidium*, *Claroideoglossum*, *Doassansia*, *Lectera*, *Cunninghamella*, *Tausonia*, *Hanseniaspora*, *Pachylepyrium* and *Athelia* were specific to *Astragalus sinicus* and *Malassezia*, *Acremonium*, *Buckleyzyma* and *Ochroconis* were specific to *Vicia villosa*.

Potential pathogens and beneficial fungi

Fungal species classification and abundance information present in the environment can be obtained through the analysis of ribosomal DNA amplicons. Knowledge of the role of fungal species in their natural environment is important, to understanding their life cycle. The fungal ecological function of each DNA amplicon sequence was determined and compared using both

FUNGuild and FungalTraits annotation tools (Table 5, Supplementary Table 8, Fig. 32). Further, the functional annotation for the fungal species obtained from the culture-dependent approach was also summarized (Table 5, Supplementary Table 8).

Table 5 Number of OTUs assigned to functions by FUNGuild and FungalTraits.

Functions	Total number of OTUs assigned to functions by both annotation tools	Total number of OTUs assigned to functions by FUNGuild	Total number of OTUs assigned to functions by FungalTraits	Shared OTUs	OTUs specific to FUNGuild	OTUs specific to Fungaltraits
All functions	117	62	110	55	7	55
Algal parasite	2	0	2	0	0	2
Animal parasite	2	0	2	0	0	2
Arbuscular Mycorrhizal	4	4	4	4	0	0
Ectomycorrhizal	1	1	0	0	1	0
Endophyte	1	0	1	0	0	1
Epiphyte	4	1	3	0	1	3
Lichenized	1	1	1	1	0	0
Lichenized parasite	1	0	1	0	0	1
multifunction	21	21	0	0	21	0
Mycoparasite	8	0	8	0	0	8
Plant-Pathogen	38	15	35	12	3	23
Saprotroph	57	19	53	15	4	38
Unassigned	104	89	68	54	35	14
Uncertain (FUNGuild with a confidence level of “possible”)	27	27	0	0	27	0

According to their trophic modes, the annotations from the FUNGuild database resulted in nine groups of fungal OTUs (Fig. 32). These DNA amplicons in the 30 samples were mostly involved in the pathotroph, followed by pathotroph-symbiotroph and pathotroph-saprotroph-symbiotroph (Fig. 32). Similarly, when analyzing the ecological function of OTUs with FungalTraits, it also showed the most abundant functional groups were plant pathogens followed by litter saprotrophs (Fig. 32).

The annotations from both tools, showed in total, 38 potentially pathogenic OTUs that belong to 21 genera (Table 5). These genera were *Alternaria*, *Boeremia*, *Chytridium*, *Colletotrichum*, *Diaporthe*, *Doassansia*, *Eremothecium*, *Erysiphe*, *Fusarium*, *Itersonilia*, *Lectera*, *Limonomyces*, *Magnaporthe*, *Olpidium*, *Plectosphaerella*, *Protomyces*, *Rhizoctonia*, *Sarocladium*, *Sclerotinia*, *Sporisorium* and *Tilletiopsis*.

Further, four arbuscular mycorrhizal OTUs belong to three genera (*Acaulospora*, *Claroideoglomus*, and *Funneliformis*), four epiphytic OTUs belong to two genera (*Buckleyzyma* and *Symmetrospora*), one lichenized OTU belongs to *Arthopyrenia*, and 57 Saprotroph OTUs belong to 37 genera (*Acremonium*, *Aspergillus*, *Buckleyzyma*, *Chaetomium*, *Chaetospermum*, *Cladosporium*, *Cunninghamella*, *Cyphellophora*, *Cystofilobasidium*, *Dactylella*, *Dissoconium*, *Endogone*, *Filobasidium*, *Gongronella*, *Hanseniaspora*, *Holtermanniella*, *Infundibulomyces*, *Knufia*, *Kondoa*, *Leucosporidium*, *Malassezia*, *Metschnikowia*, *Mucor*, *Naganishia*, *Ochroconis*, *Pachylepyrium*, *Pichia*, *Pyxidiophora*, *Rhizopus*, *Rhodotorula*, *Saccharomyces*, *Sarocladium*, *Sistotrema*, *Tausonia*, *Tetracladium*, *Udeniomyces*, and *Vishniacozyma*) were identified.

In this study, 15 fungal genera were obtained by culturomics, and most of them were previously reported as devastating plant pathogens for many crops (Supplementary Table 7).

Interestingly, several fungal species have been identified as potentially beneficial fungi or/and biocontrol fungi (Supplementary Table 7). However, several species that have been reported as both potentially pathogenic and beneficial were also identified (eg. *Albifimbria*, *Arthrinium*, *Epicoccum*, *Clonostachys*, and *Plectosphaerella*).

Discussion

In this study, we sought to determine the diversity and identification of fungi colonizing two green manure crops (*Astragalus sinicus* and *Vicia villosa*) in different geographical locations in China. To characterize the fungal community structure of these crops, we used both culture-dependent and culture-independent techniques. Here we provide the first comprehensive work comparing fungal communities on *Astragalus sinicus* and *Vicia villosa* using both approaches with well-resolved taxonomic identifications based on multi-marker phylogenies. Furthermore, a worldwide checklist of fungi on *Astragalus sinicus* and *Vicia villosa* is also provided which is an important resource for research that focuses on fungal diversity in green manure crops.

Fungal diversity of green manure crops

Every plant species has its own hidden, large community of endophytes which is a component of fungal diversity (Porrás-Alfaro & Bayman 2011, Du et al. 2020). This undescribed biodiversity and its lifestyle have received the attention of taxonomists, mycologists, ecologists, chemists, and evolutionary biologists (Song et al. 2016, Carbungco et al. 2017, Khiralla et al. 2017, Kumar et al. 2017, An et al. 2020, de Silva et al. 2021). The fungal endophytic community in many hosts are dominated by various classes including Dothideomycetes, Eurotiomycetes, Leotiomycetes, Pezizomycetes and Sordariomycetes (Qadri et al. 2014, Yu et al. 2018, Dong et al. 2021). Many endophytic Basidiomycetes and Zygomycetes are also common in grasses (Sánchez Márquez et al. 2007). Additionally, it is stated that because of the high plant diversity in the tropics, endophyte diversity might also be highest in the tropics rather than in temperate regions (Cannon & Simmons 2002, Banerjee 2011). However, this needs to be confirmed with more extensive studies on plant species to estimate the distribution patterns and diversity of endophytic fungi across wide geographical ranges.

In this study, the fungal diversity isolated far exceeds the number of strains usually reported from hosts. In many cases, no more than 50-100 strains were reported (Anita et al. 2009, Dissanayake et al. 2018, Choosa-Nga et al. 2019, de Silva et al. 2021). Only in a few studies were up to or more than 100 strains isolated (Hilarino et al. 2011, Smith et al. 2011, de Pádua et al. 2019). The high number of detected strains in our study is because different target host species were investigated over a wider geographical area, in contrast to most studies that display less diversity. However, the isolated strains only include those present at the time of sampling and cultivable, as most of the species cannot be cultured on media.

Culture-independent techniques usually yield a high number of species (more than 50 OTUs) as compared to using traditional approaches (Dissanayake et al. 2018, Jayawardena et al. 2018). The fungal community obtained from the culture-dependent approach appeared to be dominated by members of Sordariomycetes with species of *Fusarium* isolated 381 times. *Fusarium* is a ubiquitous fungal group and the world's most economically destructive and species-rich genus found in many environments including soil and litter (Aoki et al. 2014, O'Donnell et al. 2015). *Fusarium* has also been found as asymptomatic endophytes, plant pathogens and/or associated with lignocellulosic wastes due to their saprobic lifestyle (Márquez et al. 2008, 2012, Orgiazzi et al. 2013, Demers et al. 2015, Benitez et al. 2016). Several studies have shown a high relative abundance of *Fusarium* species from cover crops, such as hairy vetch (*Vicia villosa*) and they were often regarded as host-generalists (Benitez et al. 2016). Walder et al. (2017) have revealed that the hairy vetch acts as a potential alternative host for *Fusarium*, and it showed higher relative abundance compared to other cover crop treatments in their study. However, we could not find any previous report of *Fusarium* spp. on milk-vetch (*Astragalus sinicus*), and this may be due to a lack of studies on green manure crops. According to the *Fusarium* MLST database

(<http://www.cbs.knaw.nl/fusarium/>; O'Donnell et al. 2010), our strains belong to several of the most important plant pathogenic lineages (eg. *F. fujikuroi*, *F. oxysporum*, *F. solani* and *F. sambucinum* species complexes, Supplementary Tables 3, 5). Taxa of the *Fusarium* complexes can cause devastating diseases, such as rice bakanae, maize ear rot and soybean root rot (O'Donnell et al. 2015, Qiu et al. 2020). Species of *Fusarium* produce mycotoxins (eg. Beauvericin, Enniatins, Fumonisin, Fusaric acid, Fusaproliferin, Gibberellic acids, and Moniliformin) which cause chronic and acute toxicity to humans and livestock (Bottalico 1998, Desjardins et al. 2000, Qiu et al. 2020). Metabolites such as Fumonisin are found in relatively high concentrations, especially in rice and maize (Ferrigo et al. 2016, Qiu et al. 2020).

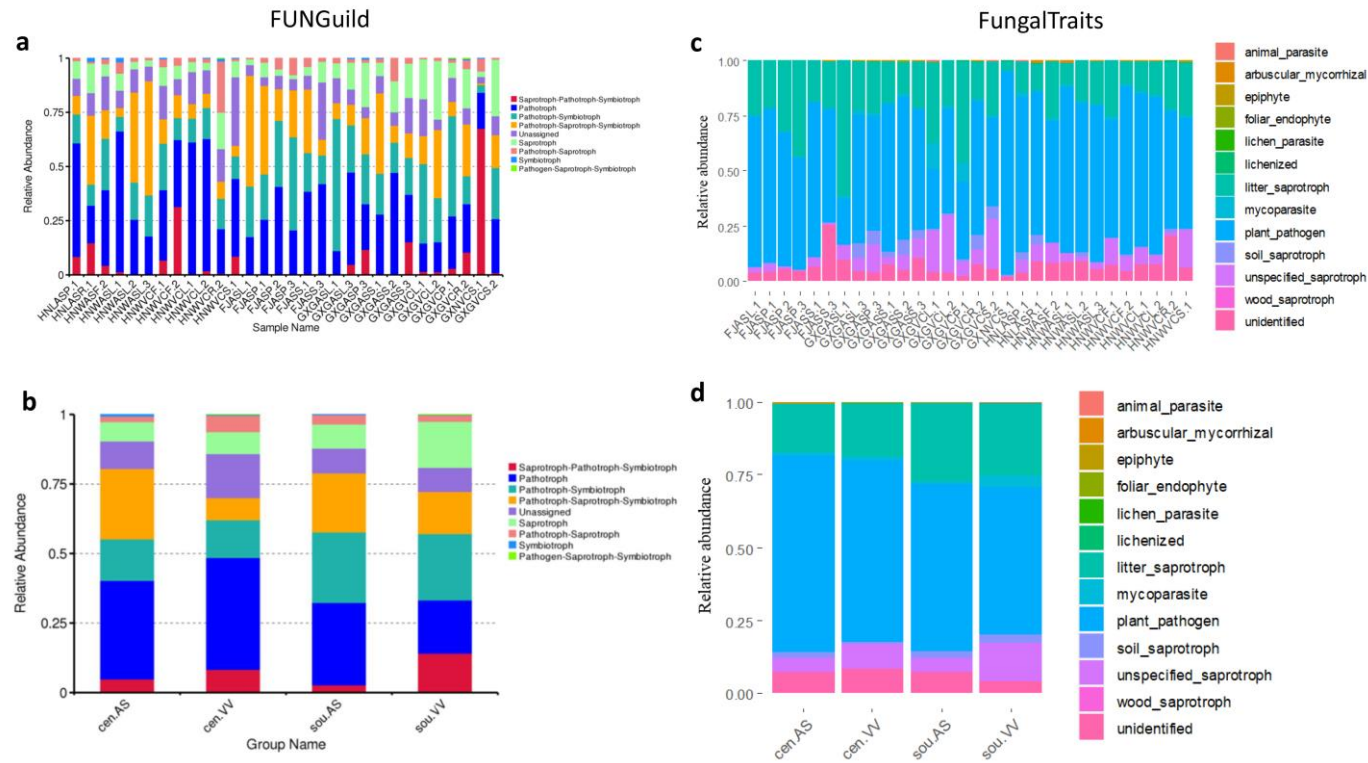


Figure 32 – a Relative abundance of all samples (FUNGuild). b Summary of functional annotation of two crops (FUNGuild). c Relative abundance of all samples (FungalTraits). d Summary of functional annotation of two crops (FungalTraits).

The second and third most abundant species from the culture-dependent approach were *Alternaria alternata* and *Epicoccum layuense*. These taxa have not been frequently reported on *Astragalus sinicus* and *Vicia villosa*. *Alternaria* has previously been isolated from roots and aerial parts of standing milk-vetch (*Astragalus adsurgens*); a perennial native legume pasture plant in China (Li et al. 2007). However, *Alternaria* spp. were most common from aerial tissues of diseased plants of *Astragalus adsurgens*, while *Fusarium chlamydosporum* and *F. solani* were isolated from roots. Root rot caused by *Embellisia* sp. (\equiv *Alternaria* spp.), together with *Fusarium* spp. and *Conostachys rosea*, appears to be the main fungal disease contributor to the decline of standing milk-vetch pasture in northern China (Li et al. 2007). *Hypocreales* spp. were also abundant in vetch roots and *Ilyonectria* species have commonly been described as pathogens on vetch roots and stems (Lombard et al. 2014, Benitez et al. 2016).

Few *Glomeromycetes* OTUs (OTU 287, OTU 391, OTU 447 and OTU 537) were found in this study (Supplementary Tables 4, 7, 8), and they were identified as *Claroideoglossum etunicatum*, *Acaulospora laevis*, *Glomus* sp. and *Glomeromycotina* species. These arbuscular mycorrhizal (AM) fungi are known to be dominant in soils that are treated with vetch crops (Benitez et al. 2016). Similar observations were obtained in many studies, whereas *Diversisporales* (*Acaulosporaceae*, *Acaulospora*) and *Glomerales* (*Glomus* and *Funneliformis*) like sequences were significantly more abundant in vetch (Benitez et al. 2016). It might be that the 18S rRNA primer (528F/706R) used in this study is biased towards preferential amplification of *Ascomycota* and exhibits low amplicon recovery of taxa within the *Glomeromycetes*. To target arbuscular mycorrhizal fungi, AM-specific small subunits of the ribosomal gene region were used (AM-specific AML1/AML2), resulting in 91% of the *Glomerales* sequences and 9% of *Diversisporales* in prairie soils (Lee et al. 2008, Benitez et al. 2016).

Variability of fungi

The endophytic fungal communities within a single host may differ depending on internal and external factors. External factors are different sites, climates, seasons, nutrient availability and environmental conditions whereas internal factors are plant species, plant density, and interactions with other microbes. We observed variations in endophytic communities in above-ground plant parts and below-ground plant parts within the same plant species and also between *Astragalus sinicus* and *Vicia villosa*. This difference may be due to the external environmental variations of exposure to air and sunlight. Previous studies have shown that the plant species is a major driver in shaping the microbial communities that inhabit the phyllosphere (Redford et al. 2010, Rastogi et al. 2012, Kembel & Mueller 2014). Furthermore, temporal effects have also been shown to significantly affect microbial community structures in agroecosystems such as conventional, agricultural plots and early successional grasslands (Lauber et al. 2013). Herein, we show that the differences in fungal communities between two crops may be due to the impact of a temporal component, as the samples have not been harvested at the same time.

Comparison of ecological functions; FUNGuild vs FungalTraits

To obtain insight into the role of endophytic fungi–fungi and/or host–endophytic fungi interactions in shaping the mycobiome and plant health, it is necessary to establish their functional characterization. However, most of the community studies rely only on previous literature to assign a potential function to the taxa detected by cultivation-independent approaches (Manzotti et al. 2020). Here, we used both FungalTraits and FUNGuild to interpret the functional annotations for the obtained mycobiome members (Table 5, Fig. 32). These two tools have been widely used in many mycobiome studies on different ecosystems and biomes including terrestrial and aquatic environments (Tanunchai et al. 2022). FUNGuild is an open annotation tool based on Python script that can be used to analyze fungal OTUs taxonomically and provide their ecological guild (Nguyen et al. 2016, Tanunchai et al. 2022). FungalTraits has also used a similar Python script, and it is stated that this tool is more user-friendly, and it offers an Excel-based database and a web-based interface for users without Python expertise (Pölme et al. 2020, Tanunchai et al. 2022).

Several authors have suggested that in order to obtain a better scientific interpretation of a particular mycobiome study, it is necessary to compare the performance and the ecological explanation provided by these two annotation tools (Lepinay et al. 2021, Wang et al. 2021, Tanunchai et al. 2022). Therefore, we also compared the performance of both annotation tools, and our results confirmed that FungalTraits provide better performance than FUNGuild (Table 5). We found that the total number of OTUs assigned to FungalTraits (110 OTUs) is higher than FUNGuild (62 OTUs) (Table 5). FungalTraits has assigned several OTUs for each ecological function as Algal parasite, Animal parasite, Lichenized parasite, Mycoparasite, and Endophyte category while FUNGuild did not assign any of the OTUs for these (Table 5, Fig. 32). This may be due to the high number of fungal genera in the FungalTraits database than FUNGuild (Nguyen et al. 2016, Pölme et al. 2020, Tanunchai et al. 2022). There are some similarities also found in the interpretations derived from FUNGuild and FungalTraits; the OTU number assigned for Arbuscular Mycorrhizal and Lichenized fungi are the same in both tools (Table 5, Fig. 32).

Shifts in potential pathogens and beneficial/biocontrol fungi in response to green manure applications

This study revealed a significantly higher relative abundance of pathogenic genera such as *Alternaria*, *Cladosporium*, *Fusarium* and *Rhizoctonia* associated with green manure crops in China. *Alternaria* and *Sporisorium* showed a significant relative abundance in *Astragalus sinicus* and *Sclerotinia* was more abundant in *Vicia villosa* (Fig. 30).

A major question is whether the endophytic communities in green manure *Astragalus sinicus* and *Vicia villosa* are latent pathogens of the main crop. Several major rice pathogens are among the most abundant species detected in the mycobiome of these manure crops. The functional characterization of these isolates showed that they most likely were present as latent pathogens for the rice as well as other hosts (Supplementary Table 7). External factors, such as changes in plant gene expression, habitat, nutrient status, or stress, may trigger the shift of endophytes to a pathogenic state (Schulz et al. 1999, Baayen et al. 2002, Schulz & Boyle 2005, Rojas et al. 2010, Hardoim et al. 2015). Several endophytes are known to be vertically transmitted and complete their whole life cycle within one host. However, the vast majority of horizontally transmitted endophytes are known to have a part of their life cycle on the other host and /or in soil (Peršoh 2015). Some studies showed that endophytes become saprotrophic decomposers after leaves fall and/or inhabit living leaves as dormant saprobes (Peršoh 2015).

Assessing the latent pathogenicity of endophytes has been problematic as all endophytes are not cultivable in culture media (Porrás-Alfaro & Bayman 2011). The information concerning lifestyles of endophytic fungi obtained in this study, based on previous studies are summarized in Supplementary Table 7. Most endophytic fungi associated with green manure crops have previously been recorded as plant pathogens on different hosts. Surprisingly, we observed that major rice pathogens such as; *Alternaria*- (Stackburn, seedling blight and *Alternaria* leaf spot), *Athelia*- (Seedling blight), *Fusarium*- (Pecky rice (kernel spotting), Root rots and Seedling blight), *Pyricularia (Magnaporthe)*- (Blast (leaf, neck, nodal and collar) and Stem rot), *Rhizoctonia*- (Aggregate sheath, Sheath blight, Sheath spot, Seedling blight), *Sarocladium*- (Sheath rot, Pecky rice (kernel spotting) (Groth 1991, Naeimi et al. 2003, Akhtar et al. 2014, Saichuk et al. 2014, Karthikeyan et al. 2015, Premi et al. 2019) have also been associated with these green manure crops (Supplementary Table 7). It seems that besides their beneficial traits, cover crops can also entail phytopathological risks by acting as alternative hosts for *Fusarium* and other noxious plant pathogens.

Eight known fungal human pathogens were identified in this study and previously most of them have been reported from composts and/or soil (*Alternaria alternata*, *Aspergillus lentulus*, *Chaetomium* sp., *Filobasidium* sp., *Fusarium* sp., *Ochroconis* sp., *Pichia kudriavzevii* (teleomorph of *Candida krusei*) and *Saccharomyces* sp.) (De Gannes et al. 2013). Fungal pathogens are a threat to human health, and those above-listed species are known to cause several types of mycoses and immune-compromised diseases. Prior studies have shown that bio-monitoring efforts needed to be

expanded because of the presence of opportunistic pathogens in composting systems, such as *Alternaria alternata*, *Aspergillus fumigatus*, *Candida tropicalis*, *C. krusei* and *Scytalidium lignicola* (Bonito et al. 2010, Dehghani et al. 2012, De Gannes et al. 2013). According to the American Biological Safety Association, several above-mentioned pathogens (eg. *Alternaria alternata* and *Fusarium oxysporum*) identified in this study are categorized as Biosafety Level 2 (Boutati & Anaissie 1997, Halonen et al., 1997). Because of the presence of main types of potential pathogens (including *Alternaria alternata* and *Fusarium oxysporum*) in composting systems, De Gannes et al. (2013) recommended that personal protective equipment be worn when handling plant-based composed materials (De Gannes et al. 2013). Even though this present study provides evidence of a potential health threat, it needs to be noted that these species can show substantial intra-specific variation in virulence; thus, additional bioassays of isolates are required to assess the virulence (Ben-Ami et al. 2010, De Gannes et al. 2013).

Knowledge of the pathogenic potential of a fungal strain (or isolate) is particularly important for the species that can act either as a pathogen or as a biological control agent (Taguiam et al. 2021). Even though the abundance of the potentially beneficial fungi detected in this study is relatively low, we have identified several species that can be used as potential biological control, bioherbicidal, antifungal or antagonistic agents.

Leguminous green manure crops, such as vetch crops (*Vicia villosa*, *V. sativa* and *Astragalus sinicus*), have the ability to fix air N by their nodules and activate potential nutrient components in soil (Wang et al. 2022). Kataoka et al. (2017) have also shown that incorporating green manure especially, hairy vetch (*Vicia villosa*) into the soils can stimulate fungal activity in soils (Kataoka et al. 2017). They have stated this increases the fungal biomass and certain fungal species in the soil (eg. *Cladosporium* sp.). However, these authors did not find that *Cladosporium* sp. was associated with hairy vetch plants but its presence in the soil and biomass of the *Cladosporium* sp. increased after the incorporation of hairy vetch (Kataoka et al. 2017). They have concluded that *Cladosporium* sp. was derived from the soil and hairy vetch incorporation stimulated its proliferation (Kataoka et al. 2017).

We have recovered endophytic *Epicoccum layuense* from culturomics. This species is known to cause plant diseases in several hosts (*Camellia sinensis*, cowpea, maize, oat), and some reported it as a biological control agent against plant pathogens (Supplementary Table 7, Taguiam et al. 2021). *Epicoccum layuense*, E 24 isolate showed antifungal activity against esca disease complex of grapevine pathogens; *Phaeoconiella chlamydospora* and *Phaeoacremonium minimum* in both in-vitro and in-vivo conditions (Del Frari et al. 2019, Taguiam et al. 2021). However, during this interaction, it is stated that there is no direct evidence of chemical inhibition, therefore the role of the *E. layuense* metabolites produced remains to be investigated (Taguiam et al. 2021).

During this study, we were also able to isolate a nematophagous fungus; *Plectosphaerella cucumerina* and recent works have demonstrated that *P. cucumerina* has potential as a biological control agent against potato cyst nematodes (Atkins et al. 2003). Also, this species is known to have the potential as a selective bioherbicide for controlling *Galium aparine* (false cleavers) in *Brassica napus* (canola), *Sagittaria trifolia* (arrowhead) in *Oryza sativa* (rice) grown in paddies, the water weed; *Hydrilla verticillate*, and *Cirsium arvense* (Bailey et al. 2017).

Another excellent mycoparasite, *Clonostachys rosea* is also able to isolate as an endophyte during this study. *Clonostachys rosea* demonstrates effective biological control ability against numerous fungal plant pathogens (*Alternaria dauci*, *A. radicina*, *Botrytis cinerea*, *B. aclada*, *Bipolaris sorokiniana*, *Drechslera teres*, *Fusarium graminearum*, *F. verticillioides*, *F. crookwellense*, *F. culmorum*, *F. solani*, *Moniliophthora roreri*, *Phytophthora palmivora*, *Rhizoctonia solani*, *Rhynchosporium commune* and *Sclerotinia sclerotiorum*), nematodes (*Bursaphelenchus xylophilus*, *Caenorhabditis elegans*, *Haemonchus contortus*, *Meloidogyne* sp., *Oncometopia tucumana*, *Panagrellus redivivus*) and insects (*Myzus persicae*, *Rhopalosiphum padi*, *Thrips tabaci* and *Varroa destructor*) (Sun et al. 2020). Overall, these findings indicate that green manure crops like *Astragalus sinicus* and *Vicia villosa* provide the habitat for both pathogenic and

beneficial fungi and this may lead to an antagonistic development to shaping the fungal community structure.

How do findings from HTS studies contribute to the global number of fungi?

The recent estimation of fungal numbers was 2.2-3.8 million (Hawksworth & Lücking 2017, Hyde et al. 2020, Hyde 2022). However, up to 150,000 species have been identified and this is 2.6-4.5% of the estimated species (Hyde et al. 2020, Hyde 2022). Therefore, there is much research needed to quantify the actual number of fungi (Hyde 2022). Many authors have suggested that traditional approaches do not really demonstrate or quantify the exact number of fungi or the global fungal diversity or elucidate the fungal community composition (Fadrosh et al. 2014, Talbot et al. 2014, Tedersoo et al. 2020, Baldrian et al. 2022). Next-generation sequencing provides novel information on fungal numbers and HTS has been widely utilized in several areas of biodiversity research including mycology (Hongsanan et al. 2018, Thines et al. 2018, Baldrian et al. 2022). It is stated that from the studies concerning natural habitats in terrestrial ecosystems, over 250 million ITS2 sequences have been generated (Baldrian et al. 2022). However, Baldrian et al. (2022) have analyzed these OTUs and the total richness of non-singleton fungal taxa across the studies published so far is 1.08% million. Among them, the majority were Ascomycota (56.8%) (Baldrian et al. 2022). According to this analysis, soil and litter showed the highest alpha diversity of fungi (Baldrian et al. 2022). Samples of lichen and plant tissues showed the highest proportion of unknown fungal species (Baldrian et al. 2022). Most of the OTUs in our study belong to Ascomycota (77) followed by Basidiomycota (55), Mucoromycota (19), Chytridiomycota (11), Cryptomycota (8) and fungal organisms with unclear phylum level classification (8) (Table 3, Supplementary Table 4). Most of the metabarcoding studies also showed similar observations (Baldrian et al. 2022). While this study also provides the evidence for high potential of HTS studies to uncover global fungal diversity. However, when considering the use of HTS for estimates of fungal numbers or global fungal diversity, care is needed concerning the limitations of metabarcoding approaches (Hongsanan et al. 2018, Thines et al. 2018, Baldrian et al. 2022). Therefore, the use of a combination of HTS, fungal taxonomy and ecological analyses has been recommended (Dissanayake et al. 2018, Jayawardena et al. 2018, Baldrian et al. 2022).

Checklist of fungi on *Astragalus sinicus* and *Vicia villosa*

Eighty-seven micro-fungi have been reported on green manure crops (*Astragalus sinicus*-20 and *Vicia villosa*-67) are listed in this study. This is an updated worldwide checklist of fungi on *Astragalus sinicus* and *Vicia villosa*. These taxa are distributed in 25 families and 41 genera. For each species, family, and known locality, as well as references, are provided (Supplementary Table 4).

Conclusion

In this study, variation and the diversity of the fungal community of green manure crops (*Astragalus sinicus* and *Vicia villosa*) were explored. More fungal genera were obtained by metagenomics than by culturomics. Apparently, there were some differences in the fungal community between crops and the location. The abundance of Ascomycota was higher in both crops. Functional prediction analyses visualized the fungal community structure in these green manure crops and pathogens and/or pathotroph-saprotroph-symbiotroph were the dominant trophic mode in these fungal communities. Potential beneficial/biocontrol strains were also detected. Taken together, our findings suggest that assessing the relationship between fungal communities in green manure crops to the main crop is challenging. However, to clarify and confirm the functional roles of fungal endophytes in green manure crops such as *Astragalus sinicus* and *Vicia villosa*, additional studies would be required using taxa that have been isolated and cultured. Even though the pathogenic lifestyle of isolates belonging to these species was not confirmed by using a planta assay, we recommend taking precautions before incorporating green manure crops into the soil.

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Declarations

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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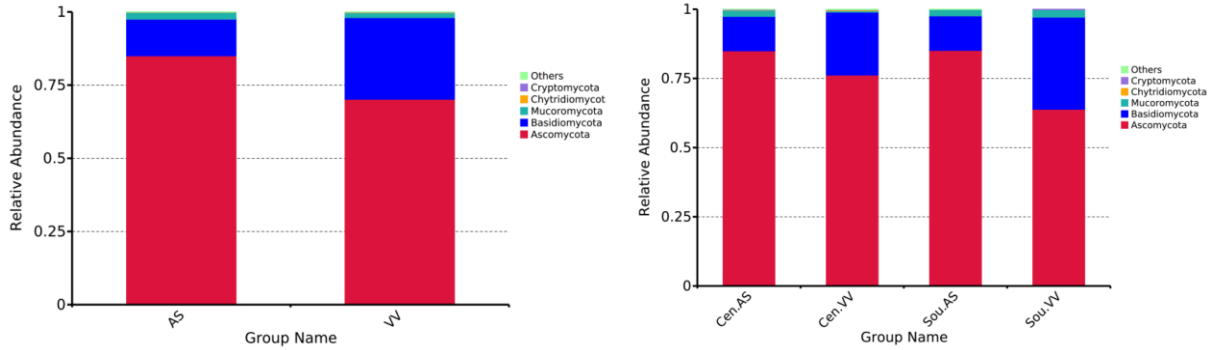
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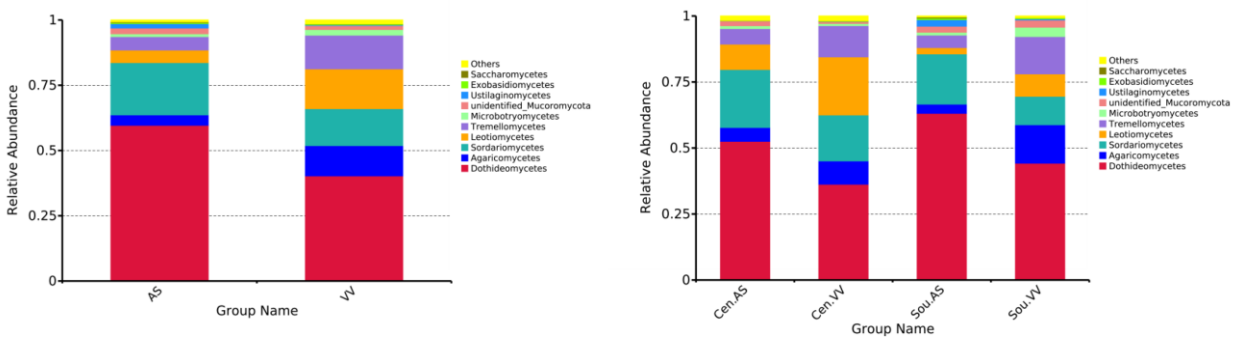
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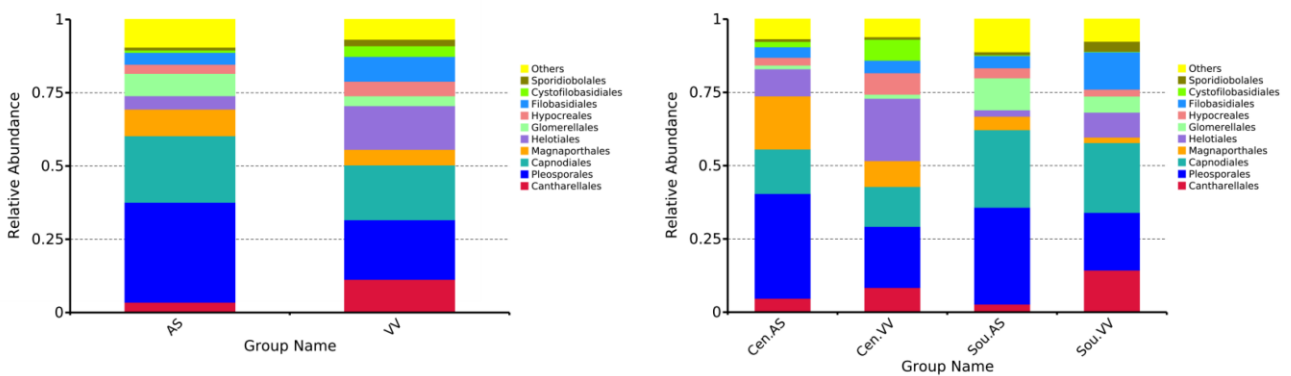
Supplementary materials



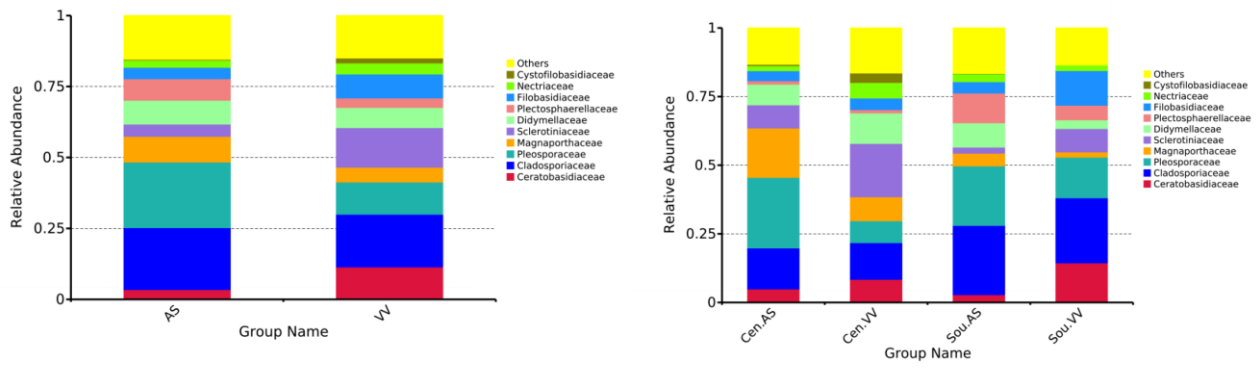
Supplementary Figure 1 – Phylum-level distribution of *Astragalus sinicus* and *Vicia villosa* mycobiota in different locations.



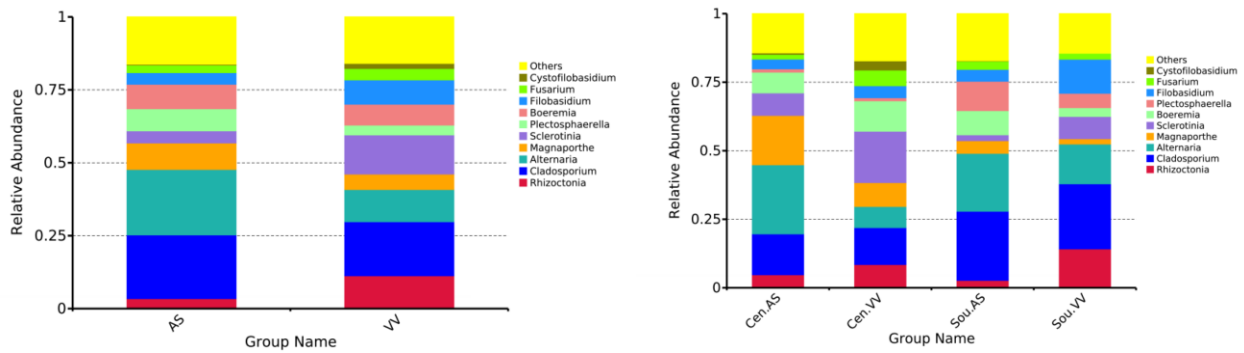
Supplementary Figure 2 – Class-level distribution of *Astragalus sinicus* and *Vicia villosa* mycobiota in different locations.



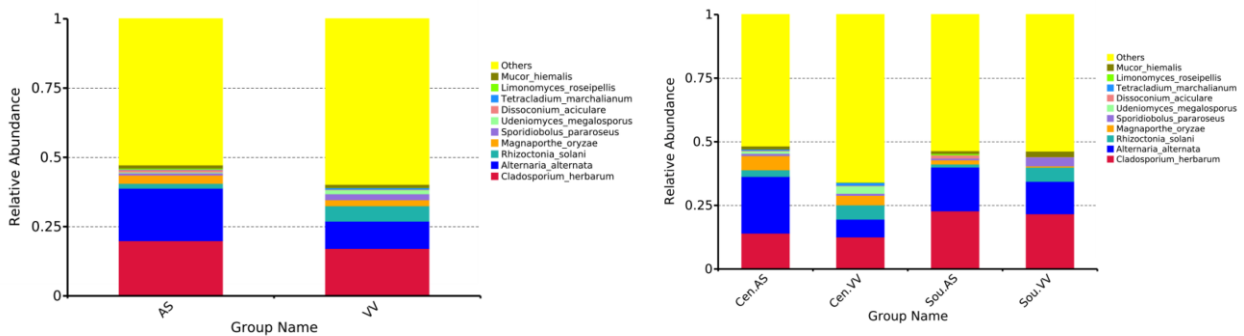
Supplementary Figure 3 – Order-level distribution of *Astragalus sinicus* and *Vicia villosa* mycobiota in different locations.



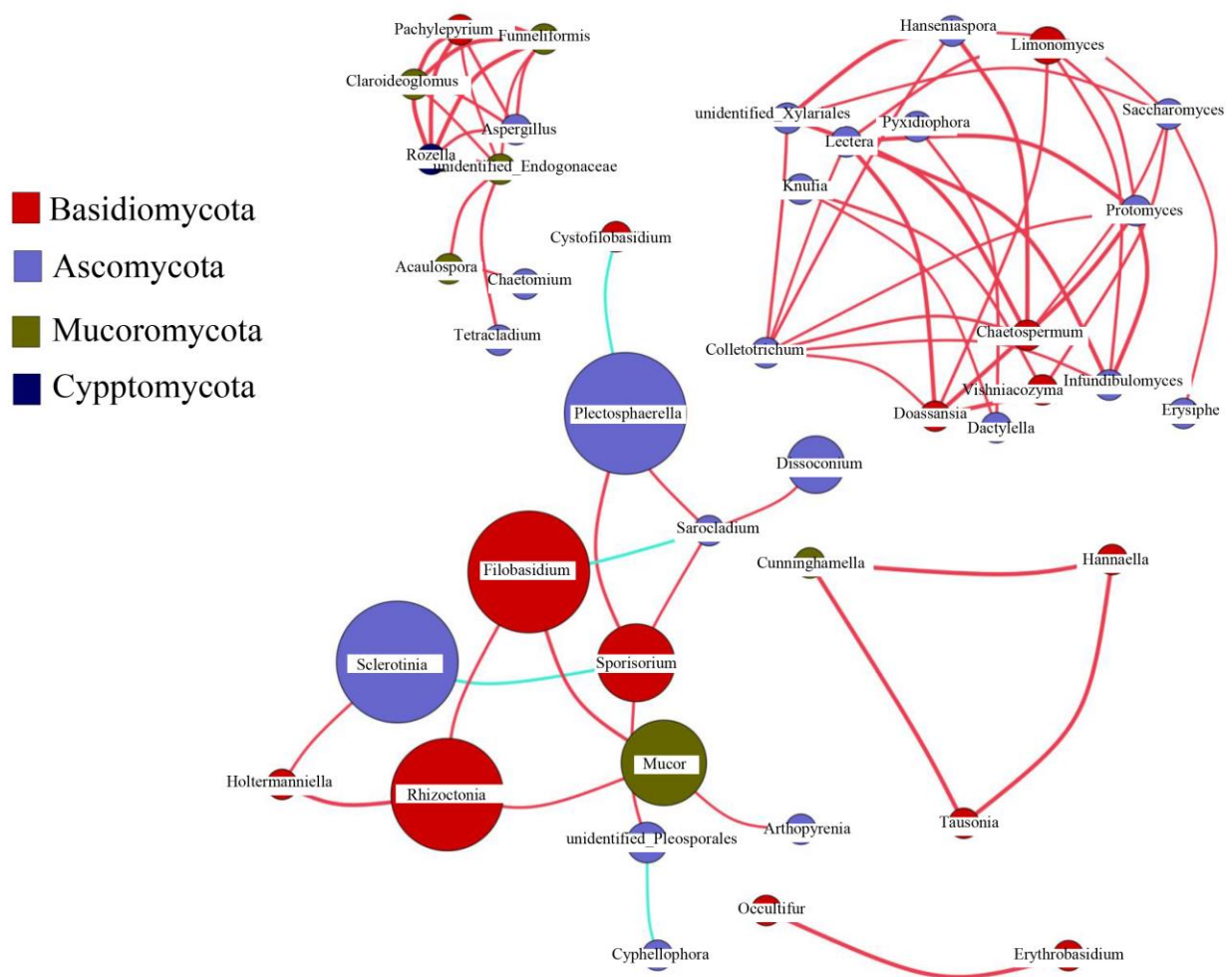
Supplementary Figure 4 – Family-level distribution of *Astragalus sinicus* and *Vicia villosa* mycobiota in different locations.



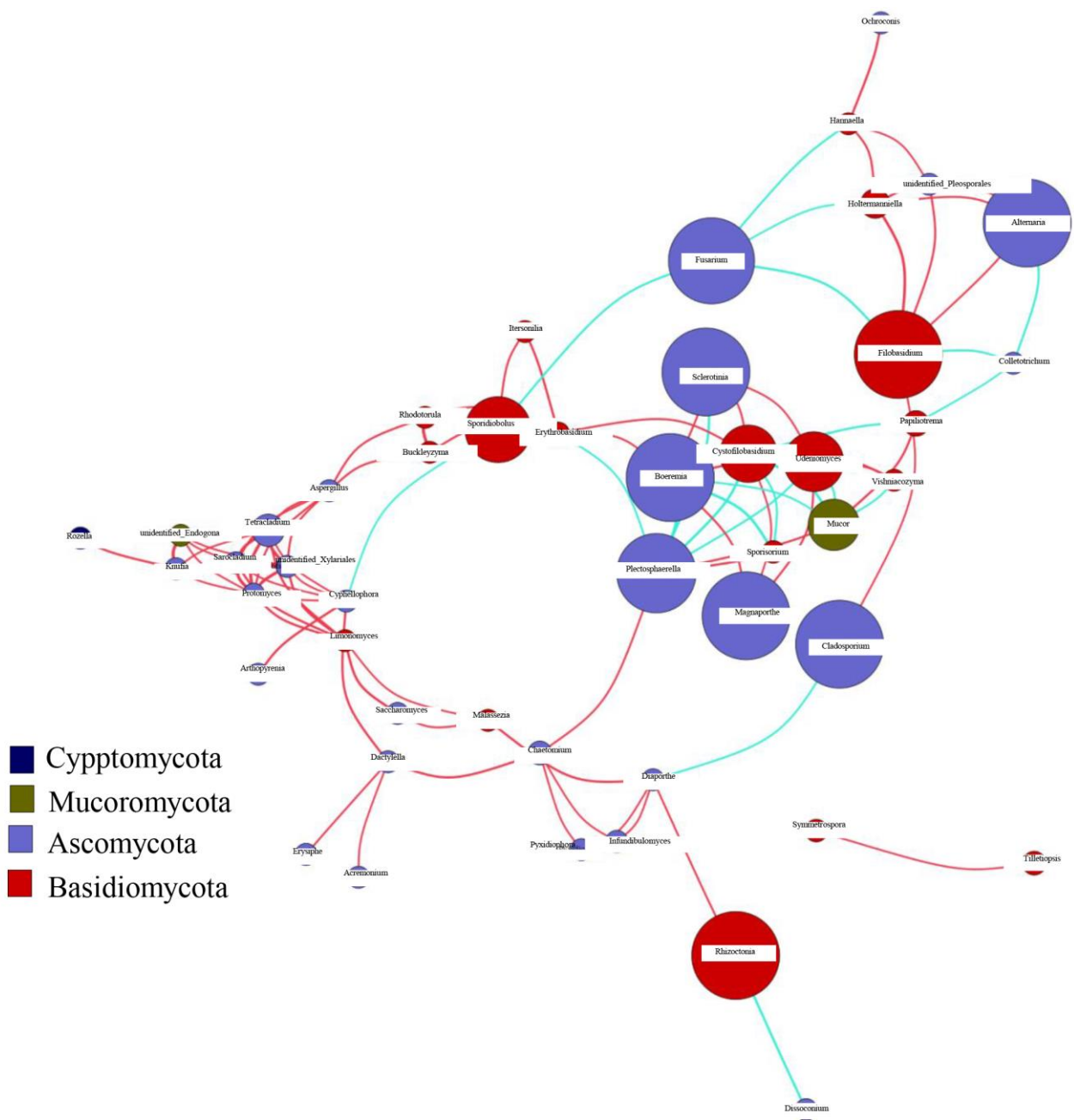
Supplementary Figure 5 – Genus-level distribution of *Astragalus sinicus* and *Vicia villosa* mycobiota in different locations.



Supplementary Figure 6 – Species-level distribution of *Astragalus sinicus* and *Vicia villosa* mycobiota in different locations



Supplementary Figure 7 – Co-occurrence network analysis of fungal taxa in *Astragalus sinicus* (Edges are connected between nodes that were significantly ($P < 0.05$; Pearson correlation test) and highly correlated (Pearson's $r > 0.6$). The size of the node is proportional to the abundance of the genus. Node colour corresponds to the taxonomic classification of the genus. Positive or negative correlations are shown in red or cyan, while edges thickness is related to correlation magnitude).



Supplementary Figure 8 – Co-occurrence network analysis of fungal taxa in *Vicia villosa* (Edges are connected between nodes that were significantly ($P < 0.05$; Pearson correlation test) and highly correlated (Pearson's $r > 0.6$). The size of the node is proportional to the abundance of the genus. Node colour corresponds to the taxonomic classification of the genus. Positive or negative correlations are shown in red or cyan, while edges thickness is related to correlation magnitude).

Supplementary Table 1 GenBank accession numbers for the isolates recovered from this study.

Species	Culture collection number	GenBank accession number			
		ITS	GAPDH	<i>tef-1a</i>	<i>Alt-a</i>
<i>Alternaria alternata</i>	JZB3180041	MW793870	MW817982	MW818056	MW818019
<i>Alternaria alternata</i>	JZB3180042	MW793871	MW817983	MW818057	MW818020
<i>Alternaria alternata</i>	JZB3180043	MW793872	MW817984	MW818058	MW818021
<i>Alternaria alternata</i>	JZB3180044	MW793873	MW817985	MW818059	MW818022
<i>Alternaria alternata</i>	JZB 3180045	MW793874	MW817986	MW818060	MW818023
<i>Alternaria alternata</i>	JZB3180046	MW793875	MW817987	MW818061	MW818024
<i>Alternaria alternata</i>	JZB3180047	MW793876	MW817988	MW818062	MW818025
<i>Alternaria alternata</i>	JZB3180048	MW793877	MW817989	MW818063	MW818026
<i>Alternaria alternata</i>	JZB3180049	MW793878	MW817990	MW818064	MW818027
<i>Alternaria alternata</i>	JZB3180050	MW793879	MW817991	MW818065	MW818028
<i>Alternaria alternata</i>	JZB3180051	MW793880	MW817992	MW818066	MW818029
<i>Alternaria alternata</i>	JZB3180052	MW793881	MW817993	MW818067	MW818030
<i>Alternaria alternata</i>	JZB3180053	MW793882	MW817994	MW818068	MW818031
<i>Alternaria alternata</i>	JZB3180054	MW793883	MW817995	MW818069	MW818032
<i>Alternaria alternata</i>	JZB3180055	MW793884	MW817996	MW818070	MW818033
<i>Alternaria alternata</i>	JZB3180056	MW793885	MW817997	MW818071	MW818034
<i>Alternaria alternata</i>	JZB3180057	MW793886	MW817998	MW818072	MW818035
<i>Alternaria alternata</i>	JZB3180058	MW793887	MW817999	MW818073	MW818036
<i>Alternaria alternata</i>	JZB3180059	MW793888	MW818000	MW818074	MW818037
<i>Alternaria alternata</i>	JZB3180060	MW793889	MW818001	MW818075	MW818038
<i>Alternaria alternata</i>	JZB3180063	MW793890	MW818002	MW818076	MW818039
<i>Alternaria astragalicola</i>	JZB3180064	MW793891	MW818003	MW818077	MW818040
<i>Alternaria gaisen</i>	JZB3180065	MW793892	MW818004	MW818078	MW818041
<i>Alternaria gaisen</i>	JZB3180066	MW793893	MW818005	MW818079	MW818042
<i>Alternaria guizhouensis</i>	JZB3180067	MW793894	MW818006	MW818080	MW818043
<i>Alternaria guizhouensis</i>	JZB3180068	MW793895	MW818007	MW818081	MW818044
<i>Alternaria guizhouensis</i>	JZB3180069	MW793896	MW818008	MW818082	MW818045
<i>Alternaria henanensis</i>	JZB3180070	MW793897	MW818009	MW818083	MW818046
<i>Alternaria henanensis</i>	JZB3180071	MW793898	MW818010	MW818084	MW818047
<i>Alternaria henanensis</i>	JZB3180072	MW793899	MW818011	MW818085	MW818048
<i>Alternaria henanensis</i>	JZB3180073	MW793900	MW818012	MW818086	MW818049
<i>Alternaria henanensis</i>	JZB3180074	MW793901	MW818013	MW818087	MW818050
<i>Alternaria henanensis</i>	JZB3180075	MW793902	MW818014	MW818088	MW818051
<i>Alternaria henanensis</i>	JZB3180076	MW793903	MW818015	MW818089	MW818052
<i>Alternaria henanensis</i>	JZB3180077	MW793904	MW818016	MW818090	MW818053

Supplementary Table 1 Continued.

Species	Culture collection number	GenBank accession number				
		ITS	GAPDH	<i>tef-1a</i>	Alt-a	
<i>Alternaria henanensis</i>	JZB3180078	MW793905	MW818017	MW818091	MW818054	
<i>Alternaria henanensis</i>	JZB3180079	MW793906	MW818018	MW818092	MW818055	
		ITS	LSU	<i>tef-1a</i>	<i>tub</i>	
<i>Arthrinium</i>	JZB 3260002	MT664206	MT666065	MW768811	MW768813	
<i>Arthrinium</i>	JZB 3260003	MT664207	MT666066	MW768812	MW768814	
		<i>rpb2</i>	G3PDH	HSP60		
<i>Botrytis cinerea</i>	JZB 350044	MW768737	MN953418	MW768741		
<i>Botrytis cinerea</i>	JZB 350045	MW768738	MN953419	MW768742		
<i>Botrytis cinerea</i>	JZB 350046	MW768739	MN953420	MW768743		
<i>Botrytis cinerea</i>	JZB 350047	MW768740	MN953421	MW768744		
		ITS	GAPDH	CHS	ACT	<i>tub</i>
<i>Colletotrichum</i>	JZB 330198	MW487987	MW768839	MW768827	MW768815	MW768851
	JZB 330199	MW487988	MW768840	MW768828	MW768816	MW768852
	JZB 330206	MW488046	MW768841	MW768829	MW768817	MW768853
	JZB 330207	MW488047	MW768842	MW768830	MW768818	MW768854
	JZB 330208	MW488048	MW768843	MW768831	MW768819	MW768855
	JZB 330209	MW488049	MW768844	MW768832	MW768820	MW768856
	JZB 330200	MW488050	MW768845	MW768833	MW768821	MW768857
	JZB 330201	MW488051	MW768846	MW768834	MW768822	MW768858
	JZB 330202	MW488052	MW768847	MW768835	MW768823	MW768859
	JZB 330203	MW488053	MW768848	MW768836	MW768824	MW768860
	JZB 330204	MW488054	MW768849	MW768837	MW768825	MW768861
	JZB 330205	MW488055	MW768850	MW768838	MW768826	MW768862
		ITS	LSU			
<i>Plectosphaerella</i>	JZB 3540001	MT679247	MW757269			
		CAL	ITS	MCM		
<i>Sclerotinia minor</i>	JZB 3570001	MW768749	MW757287	MW768780		
<i>Sclerotinia minor</i>	JZB 3570002	MW768750	MW757288	MW768781		
<i>Sclerotinia minor</i>	JZB 3570003	MW768751	MW757289	MW768782		
<i>Sclerotinia minor</i>	JZB 3570004	MW768752	MW757290	MW768783		
<i>Sclerotinia minor</i>	JZB 3570005	MW768753	MW757291	MW768784		
<i>Sclerotinia minor</i>	JZB 3570006	MW768754	MW757292	MW768785		
<i>Sclerotinia minor</i>	JZB 3570007	MW768755	MW757293	MW768786		
<i>Sclerotinia minor</i>	JZB 3570008	MW768756	MW757294	MW768787		

Supplementary Table 1 Continued.

Species	Culture collection number	GenBank accession number			
		ITS	GAPDH	<i>tef-1a</i>	Alt- <i>a</i>
<i>Sclerotinia minor</i>	JZB 3570009	MW768757	MW757295	MW768788	
<i>Sclerotinia minor</i>	JZB 3570010	MW768758	MW757296	MW768789	
<i>Sclerotinia minor</i>	JZB 3570020	MW768759	MW757297	MW768790	
<i>Sclerotinia minor</i>	JZB 3570021	MW768760	MW757298	MW768791	
<i>Sclerotinia minor</i>	JZB 3570022	MW768761	MW757299	MW768792	
<i>Sclerotinia minor</i>	JZB 3570023	MW768762	MW757300	MW768793	
<i>Sclerotinia minor</i>	JZB 3570024	MW768763	MW757301	MW768794	
<i>Sclerotinia minor</i>	JZB 3570025	MW768764	MW757302	MW768795	
<i>Sclerotinia minor</i>	JZB 3570026	MW768765	MW757303	MW768796	
<i>Sclerotinia minor</i>	JZB 3570027	MW768766	MW757304	MW768797	
<i>Sclerotinia minor</i>	JZB 3570028	MW768767	MW757305	MW768798	
<i>Sclerotinia minor</i>	JZB 3570029	MW768768	MW757306	MW768799	
<i>Sclerotinia minor</i>	JZB 3570030	MW768769	MW757307	MW768800	
<i>Sclerotinia minor</i>	JZB 3570031	MW768770	MW757308	MW768801	
<i>Sclerotinia sclerotiorum</i>	JZB 3570011	MW768771	MW757309	MW768802	
<i>Sclerotinia sclerotiorum</i>	JZB 3570012	MW768772	MW757310	MW768803	
<i>Sclerotinia sclerotiorum</i>	JZB 3570013	MW768773	MW757311	MW768804	
<i>Sclerotinia sclerotiorum</i>	JZB 3570014	MW768774	MW757312	MW768805	
<i>Sclerotinia sclerotiorum</i>	JZB 3570015	MW768775	MW757313	MW768806	
<i>Sclerotinia sclerotiorum</i>	JZB 3570016	MW768776	MW757314	MW768807	
<i>Sclerotinia sclerotiorum</i>	JZB 3570017	MW768777	MW757315	MW768808	
<i>Sclerotinia sclerotiorum</i>	JZB 3570018	MW768778	MW757316	MW768809	
<i>Sclerotinia sclerotiorum</i>	JZB 3570019	MW768779	MW757317	MW768810	
		<i>tef-1a</i>	ITS		
<i>Lasiodiplodia mediterranea</i>	JZB 3130012	MW790280	MW774349		
<i>Lasiodiplodia mediterranea</i>	JZB 3130013	MW790281	MW774350		
		LSU	ITS	<i>rpb2</i>	
<i>Leptosphaerulina americana</i>	JZB 3550001	MW774414	MW774396	MW790282	
		ITS	<i>tub</i>		
<i>Clonostachys eriocamporesii</i>	JZB 3530004	MW774568	MW790285		
<i>Clonostachys rosea</i>	JZB 3530003	MW774569	MW790286		
<i>Bionectria ochroleuca</i>	JZB 3530002	MW774570	MW790287		
		<i>tef-1a</i>	ITS		
<i>Albifimbria verrucaria</i>	JZB 3510001	MW790283	MW774438		

Supplementary Table 1 Continued.

Species	Culture collection number	GenBank accession number				
		ITS	GAPDH	<i>tef-1a</i>	Alt- <i>a</i>	
<i>Albifimbria verrucaria</i>	JZB 3510002	MW790284	MW774439			
		ITS	CAL	HIS	<i>tef-1a</i>	<i>tub</i>
<i>Diaporthe longicolla</i>	JZB 320180	OP603019	OP627275	OP627276	OP627277	OP627278
<i>Diaporthe viciae</i>	JZB 320179	OP626092	-	OP627279	OP627280	OP627281
		<i>tef-1a</i>	ITS	<i>tub</i>		
<i>Neofusicoccum parvum</i>	JZB 3120007	MW790288	MW783674	MW790289		
		LSU	ITS	<i>rpb2</i>	<i>tub</i>	
<i>Epicoccum astragalina</i>	JZB 380085	MW861422	MW861392	MW861496	MW861466	
<i>Epicoccum henanensis</i>	JZB 380048	MW861423	MW861393	MW861497	MW861467	
<i>Epicoccum henanensis</i>	JZB 380049	MW861424	MW861394	MW861498	MW861468	
<i>Epicoccum henanensis</i>	JZB 380050	MW861425	MW861395	MW861499	MW861469	
<i>Epicoccum laticollum</i>	JZB 380072	MW850468	MW850445	MW861456	MW861461	
<i>Epicoccum laticollum</i>	JZB 380071	MW850469	MW850446	MW861457	MW861462	
<i>Epicoccum laticollum</i>	JZB 380070	MW850470	MW850447	MW861458	MW861463	
<i>Epicoccum laticollum</i>	JZB 380074	MW850471	MW850448	MW861459	MW861464	
<i>Epicoccum laticollum</i>	JZB 380073	MW850472	MW850449	MW861460	MW861465	
<i>Epicoccum layuense</i>	JZB 380067	MW861426	MW861396	MW861500	MW861470	
<i>Epicoccum layuense</i>	JZB 380068	MW861427	MW861397	MW861501	MW861471	
<i>Epicoccum layuense</i>	JZB 380069	MW861428	MW861398	MW861502	MW861472	
<i>Epicoccum layuense</i>	JZB 380051	MW861429	MW861399	MW861503	MW861473	
<i>Epicoccum layuense</i>	JZB 380052	MW861430	MW861400	MW861504	MW861474	
<i>Epicoccum layuense</i>	JZB 380061	MW861431	MW861401	MW861505	MW861475	
<i>Epicoccum layuense</i>	JZB 380062	MW861432	MW861402	MW861506	MW861476	
<i>Epicoccum layuense</i>	JZB 380063	MW861433	MW861403	MW861507	MW861477	
<i>Epicoccum layuense</i>	JZB 380064	MW861434	MW861404	MW861508	MW861478	
<i>Epicoccum layuense</i>	JZB 380065	MW861435	MW861405	MW861509	MW861479	
<i>Epicoccum layuense</i>	JZB 380066	MW861436	MW861406	MW861510	MW861480	
<i>Epicoccum layuense</i>	JZB 380057	MW861437	MW861407	MW861511	MW861481	
<i>Epicoccum layuense</i>	JZB 380060	MW861438	MW861408	MW861512	MW861482	
<i>Epicoccum layuense</i>	JZB 380058	MW861439	MW861409	MW861513	MW861483	
<i>Epicoccum layuense</i>	JZB 380059	MW861440	MW861410	MW861514	MW861484	
<i>Epicoccum rosae</i>	JZB 380075	MW861441	MW861411	MW861515	MW861485	
<i>Epicoccum rosae</i>	JZB 380076	MW861442	MW861412	MW861516	MW861486	
<i>Epicoccum rosae</i>	JZB 380077	MW861443	MW861413	MW861517	MW861487	
<i>Epicoccum rosae</i>	JZB 380078	MW861444	MW861414	MW861518	MW861488	

Supplementary Table 1 Continued.

Species	Culture collection number	GenBank accession number			
		ITS	GAPDH	<i>tef-1a</i>	Alt- <i>a</i>
<i>Epicoccum viciae-villosae</i>	JZB 380080	MW861445	MW861415	MW861519	MW861489
<i>Epicoccum viciae-villosae</i>	JZB 380081	MW861446	MW861416	MW861520	MW861490
<i>Epicoccum viciae-villosae</i>	JZB 380082	MW861447	MW861417	MW861521	MW861491
<i>Epicoccum viciae-villosae</i>	JZB 380079	MW861448	MW861418	MW861522	MW861492
<i>Epicoccum viciae-villosae</i>	JZB 380083	MW861449	MW861419	MW861523	MW861493
<i>Epicoccum viciae-villosae</i>	JZB 380086	MW861450	MW861420	MW861524	MW861494
<i>Epicoccum viciae-villosae</i>	JZB 380084	MW861451	MW861421	MW861525	MW861495
		ITS	GAPDH	CAL	
<i>Stemphylium astragali</i>	JZB 3240024	MT672523	MW768745	MW768747	
<i>Stemphylium astragali</i>	JZB 3240025	MT672524	MW768746	MW768748	
		ITS	LSU	SSU	<i>tef-1a</i>
<i>Pseudopithomyces chartarum</i>	JZB 3560001	MW768090	MW774265	MW774277	MW790278
<i>Pseudopithomyces chartarum</i>	JZB 3560002	MW768091	MW774266	MW774278	MW790279

Supplementary Table 2 Identification Result from *Fusarium* MLST database.

Isolate no.	Identification Result from <i>Fusarium</i> MLST database*	Similarity
GZASS_FU1	<i>F. sambucinum</i> complex; NRRL 13818	100%
GZASS_FU4	<i>F. fujikuroi</i> , Asian subclade; NRRL 13566	99.29%
GZASS_FU2	<i>F. sambucinum</i> complex; NRRL 13818	98%
GZASF_FU1	<i>F. graminearum</i> (<i>F. sambucinum</i> complex)	98.59%
FJASR_FU1	<i>F. incarnatum-equiseti</i> complex; NRRL 26417	99.39%
FJASL_FU1	<i>F. graminearum</i> (<i>F. sambucinum</i> complex); MRC 2580	98.74%
HNLASL_FU1	<i>F. sambucinum</i> complex; NRRL 13818	100%
HNLASL_FU2	<i>F. sambucinum</i> complex; NRRL 13818	99.69%
HNLASR_FU1	<i>F. sambucinum</i> complex; NRRL 13818	98.99%
HNLASL_FU3	<i>F. graminearum</i> (<i>F. sambucinum</i> complex); MRC 1785	98.59%
HNWASP_FU1	<i>F. sambucinum</i> complex; NRRL 13818	99.69%
HNWASS_FU1	<i>F. sambucinum</i> complex; NRRL 13818	99.59%
HNLASR_FU2	<i>F. sambucinum</i> complex; NRRL 13818	99.90%
HNWASS_FU2	<i>F. sambucinum</i> complex; NRRL 13818	99.59%
FJASP_FU1	<i>F. sambucinum</i> complex; NRRL 13818	100%
HNWVCP_FU1	<i>F. sambucinum</i> complex; NRRL 13818	100%
GXGVCS_FU1	<i>F. sambucinum</i> complex; NRRL 13818	100%
HNWVCR_FU1	<i>F. avenaceum</i> (<i>F. tricinctum</i> complex)	95.78%
GZASR_FU1	<i>F. sambucinum</i> complex; NRRL 13818	99.30%
GZASL_FU2	<i>F. graminearum</i> (<i>F. sambucinum</i> complex)	98.13%
HNWVCR_FU2	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex)	99.02%
HNWASL_FU1	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex)	98.83%
HNWVCP_FU2	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex); NRRL 45994	99.08%
GZASL_FU1	<i>F. proliferatum</i> (<i>Fusarium fujikuroi</i> complex)	98.63%
HNWASS_FU3	<i>F. fujikuroi</i> , Asian subclade; NRRL 22944	99.89%
HNWASF_FU1	<i>F. oxysporum</i> complex NRRL 36408	100.00%
HNWVCL_FU1	<i>F. incarnatum-equiseti</i> complex; NRRL 26417	99.16%
HNWVCF_FU1	<i>F. tricinctum</i> complex; NRRL 22748	96.78%
HNWVCF_FU2	<i>F. sambucinum</i> complex; NRRL 13374	83.95%
GXGASP_FU1	<i>F. nisikadoi</i> complex; NRRL 28387	99.69%
GZASS_FU3	<i>F. sambucinum</i> complex; NRRL 13818	100.00%
HNWVCS_FU2	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex)	98.70%
HNWVCP_FU3	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex)	99.17%
HNWVCL_FU2	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex)	98.51%
GXNVCP_FU1	<i>Gibberella fujikuroi</i> complex NRRL 13602	98.91%
GZASR_FU2	<i>F. sambucinum</i> complex; NRRL 13818	100.00%
GXGASP_FU2	<i>F. acuminatum</i> (<i>F. tricinctum</i> complex)	98.05%
HNWASF_FU2	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex); NRRL 45994	99.08%
HNWASR_FU1	<i>F. avenaceum</i> (<i>F. tricinctum</i> complex)	96.69%
HNWVCF_FU3	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex); NRRL 36147	98.75%
HNWVCF_FU4	<i>F. acuminatum</i> (<i>F. tricinctum</i> complex)	97.75%
HNWVCP_FU4	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex); NRRL 45994	99.01%
HNWVCP_FU5	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex); NRRL 45994	98.93%
HNWVCF_FU5	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex); NRRL 36147	97.49%
HNLASR_FU3	<i>F. sambucinum</i> complex; NRRL 13818	100.00%
HNLASL_FU4	<i>F. sambucinum</i> complex; NRRL 13818	99.90%
GXGVCL_FU1	<i>F. graminearum</i> (<i>F. sambucinum</i> complex)	98.30%
GZASR_FU3	<i>F. sambucinum</i> complex; NRRL 13818	99.59%
HNWASP_FU2	<i>F. sambucinum</i> complex; NRRL 13818	100.00%
HNWASF_FU3	<i>F. sambucinum</i> complex; NRRL 13818	100.00%
HNWVCP_FU6	<i>F. sambucinum</i> complex; NRRL 13818	99.69%
HNWVCP_FU7	<i>F. sambucinum</i> complex; NRRL 13818	100.00%
GXGVCP_FU1	<i>F. sambucinum</i> complex; NRRL 13818	99.59%
GXGVCP_FU2	<i>F. sambucinum</i> complex; NRRL 13818	100.00%
GXGVCP_FU3	<i>F. sambucinum</i> complex; NRRL 13818	100.00%
FJASP_FU2	<i>F. sambucinum</i> complex; NRRL 13818	99.69%
GXGVCP_FU4	<i>F. sambucinum</i> complex; NRRL 13818	99.90%
GXGVCP_FU5	<i>F. sambucinum</i> complex; NRRL 13818	100.00%
HNWASS_FU5	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex)	96.96%

Supplementary Table 2 Continued.

Isolate no.	Identification Result from <i>Fusarium</i> MLST database*	Similarity
HNWASL_FU2	<i>F. avenaceum</i> (<i>F. tricinctum</i> complex)	98.33%
HNWASL_FU3	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex)	98.49%
HNWASL_FU4	<i>F. avenaceum</i> (<i>F. tricinctum</i> complex)	98.32%
HNWASL_FU5	<i>F. avenaceum</i> (<i>F. tricinctum</i> complex)	98.33%
GZASL_FU3	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex)	98.94%
GZASF_FU2	<i>F. avenaceum</i> (<i>F. tricinctum</i> complex)	98.13%
GZASF_FU3	<i>F. avenaceum</i> (<i>F. tricinctum</i> complex)	98.10%
HNWASS_FU6	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex)	98.77%
HNWVCR_FU3	<i>F. avenaceum</i> (<i>F. tricinctum</i> complex)	98.25%
HNWASS_FU4	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex)	98.79%
HNWASS_FU10	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex)	99.41%
HNWASF_FU4	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex)	98.54%
HNWASF_FU5	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex)	98.96%
GZASR_FU4	<i>F. avenaceum</i> (<i>F. tricinctum</i> complex)	98.64%
HNWASS_FU11	<i>F. avenaceum</i> (<i>F. tricinctum</i> complex)	98.43%
HNWVCR_FU4	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex)	99.17%
HNWVCS_FU3	<i>F. avenaceum</i> (<i>F. tricinctum</i> complex)	98.32%
HNWASS_FU13	<i>F. avenaceum</i> (<i>F. tricinctum</i> complex)	98.33%
HNWASL_FU6	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex)	98.54%
HNLASP_FU1	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex)	98.71%
HNWVCP_FU8	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex)	98.72%
HNWVCL_FU3	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex)	98.50%
HNWVCR_FU5	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex)	98.51%
HNWVCS_FU4	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex)	98.65%
HNWVCS_FU5	<i>F. avenaceum</i> (<i>F. tricinctum</i> complex)	97.90%
HNWVCR_FU6	<i>F. avenaceum</i> (<i>F. tricinctum</i> complex)	97.92%
HNWASS_FU14	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex)	99.25%
HNWVCF_FU6	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex)	98.71%
HNWASS_FU7	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex)	98.73%
GZASR_FU5	<i>F. avenaceum</i> (<i>F. tricinctum</i> complex)	98.74%
GZASR_FU6	<i>F. avenaceum</i> (<i>F. tricinctum</i> complex)	98.32%
GZASR_FU7	<i>F. avenaceum</i> (<i>F. tricinctum</i> complex)	98.74%
HNWVCR_FU7	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex)	98.79%
HNWVCR_FU8	<i>F. avenaceum</i> (<i>F. tricinctum</i> complex)	98.21%
HNWVCR_FU9	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex)	98.70%
HNWVCF_FU7	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex)	98.64%
HNWVCF_FU8	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex)	97.63%
HNWVCF_FU9	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex)	99.48%
HNWASR_FU2	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex)	98.49%
HNWASR_FU3	<i>F. avenaceum</i> (<i>F. tricinctum</i> complex)	98.33%
HNWASR_FU4	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex)	98.87%
HNWASR_FU5	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex)	98.71%
HNWVCL_FU4	<i>F. avenaceum</i> (<i>F. tricinctum</i> complex)	98.32%
HNWVCL_FU5	<i>F. avenaceum</i> (<i>F. tricinctum</i> complex)	98.32%
HNWVCL_FU6	<i>F. avenaceum</i> (<i>F. tricinctum</i> complex)	98.12%
HNWVCL_FU7	<i>F. avenaceum</i> (<i>F. tricinctum</i> complex)	98.31%
HNWVCL_FU8	<i>F. avenaceum</i> (<i>F. tricinctum</i> complex)	98.36%
HNWVCL_FU9	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex)	98.54%
HNWVCL_FU10	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex)	98.58%
HNWVCL_FU11	<i>F. avenaceum</i> (<i>F. tricinctum</i> complex)	98.21%
HNWVCS_FU6	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex)	99.04%
HNWVCS_FU7	<i>F. avenaceum</i> (<i>F. tricinctum</i> complex)	98.21%
HNWVCS_FU8	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex)	98.73%
HNWVCS_FU9	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex)	97.20%
HNWVCS_FU10	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex)	98.43%
HNWVCS_FU11	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex)	98.87%
HNWVCS_FU12	<i>F. avenaceum</i> (<i>F. tricinctum</i> complex)	98.33%
HNWVCS_FU13	<i>F. avenaceum</i> (<i>F. tricinctum</i> complex)	97.48%
HNWVCS_FU14	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex)	98.88%

Supplementary Table 2 Continued.

Isolate no.	Identification Result from <i>Fusarium</i> MLST database*	Similarity
HNWVCP_FU9	<i>F. avenaceum</i> (<i>F. tricinctum</i> complex)	98.32%
HNWVCP_FU10	<i>F. avenaceum</i> (<i>F. tricinctum</i> complex)	98.00%
HNWASP_FU3	<i>F. avenaceum</i> (<i>F. tricinctum</i> complex)	98.13%
HNWASR_FU6	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex)	98.64%
HNWASR_FU7	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex)	98.56%
HNWVCL_FU12	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex)	98.81%
HNWASS_FU8	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex)	98.59%
HNWASS_FU9	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex)	98.65%
HNWASP_FU4	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex)	98.68%
HNWASP_FU5	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex)	98.68%
HNWASP_FU6	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex)	98.41%
HNWASP_FU7	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex)	98.49%
HNWASP_FU8	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex)	98.22%
HNWASS_FU12	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex)	98.94%
HNWASP_FU9	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex)	98.71%
HNWVCP_FU11	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex)	98.50%
HNWASR_FU8	<i>F. avenaceum</i> (<i>F. tricinctum</i> complex)	98.33%
HNWVCR_FU10	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex)	98.31%
HNWVCR_FU11	<i>F. avenaceum</i> (<i>F. tricinctum</i> complex)	98.32%
HNWVCR_FU12	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex)	98.87%
HNWVCR_FU13	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex)	98.65%
HNWVCR_FU14	<i>F. avenaceum</i> (<i>F. tricinctum</i> complex)	97.99%
HNWVCR_FU15	<i>F. avenaceum</i> (<i>F. tricinctum</i> complex)	96.23%
HNWASF_FU6	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex)	98.87%
HNWASL_FU7	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex)	98.11%
HNWVCS_FU15	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex)	99.02%
HNWVCF_FU10	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex)	98.45%
HNWVCF_FU11	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex)	98.86%
HNWVCF_FU12	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex)	98.79%
HNWVCL_FU13	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex)	98.80%
HNWVCL_FU14	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex)	98.54%
HNWVCF_FU13	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex)	98.39%
HNWVCF_FU14	<i>F. avenaceum</i> (<i>F. tricinctum</i> complex)	98%
HNWVCF_FU15	<i>F. avenaceum</i> (<i>F. tricinctum</i> complex)	91.97%
HNWVCF_FU16	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex)	98.94%
HNWVCF_FU17	<i>F. avenaceum</i> (<i>F. tricinctum</i> complex)	98.33%
HNWVCL_FU15	<i>F. avenaceum</i> (<i>F. tricinctum</i> complex)	96.62%
HNWVCL_FU16	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex)	99.02%
HNWVCS_FU16	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex)	99.10%
HNWVCL_FU17	<i>F. avenaceum</i> (<i>F. tricinctum</i> complex)	98.41%
HNWVCF_FU18	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex)	98.77%
HNWVCL_FU18	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex)	98.37%
HNLASR_FU4	<i>F. nisikadoi</i> complex; NRRL 54252	97.58%
HNWASR_FU9	<i>F. nisikadoi</i> complex; NRRL 28387	100.00%
HNWASR_FU10	<i>F. oxysporum</i> complex; NRRL 36408	100.00%
HNLASR_FU5	<i>F. nisikadoi</i> complex; NRRL 28387	100.00%
HNLASR_FU6	<i>F. nisikadoi</i> complex; NRRL 28387	99.60%
HNLASR_FU7	<i>F. nisikadoi</i> complex; NRRL 28387	99.38%
GZASR_FU8	<i>F. nisikadoi</i> complex; NRRL 28387	99.90%
HNWASR_FU12	<i>F. nisikadoi</i> complex; NRRL 28387	99.90%
HNWASR_FU13	<i>F. oxysporum</i> complex (NRRL 36408)	100.00%
HNWASR_FU14	<i>F. nisikadoi</i> complex; NRRL 28387	99.90%
HNWASR_FU15	<i>F. oxysporum</i> complex (NRRL 36408)	100.00%
HNWASR_FU18	<i>F. oxysporum</i> complex (NRRL 36408)	99.84%
HNWASR_FU16	<i>F. nisikadoi</i> complex; NRRL 28387	100.00%
HNWASR_FU11	<i>F. oxysporum</i> complex (NRRL 36408)	100%
HNWVCR_FU16	<i>F. nisikadoi</i> complex; NRRL 28387	100.00%
HNWASF_FU10	<i>F. nisikadoi</i> complex; NRRL 28387	100.00%
HNWASF_FU7	<i>F. nisikadoi</i> complex; NRRL 28387	100.00%

Supplementary Table 2 Continued.

Isolate no.	Identification Result from <i>Fusarium</i> MLST database*	Similarity
HNWASF_FU8	<i>F. nisikadoi</i> complex; NRRL 28387	100.00%
HNWASF_FU9	<i>F. nisikadoi</i> complex; NRRL 28387	100%
HNWASR_FU17	<i>F. nisikadoi</i> complex; NRRL 28387	99.30%
HNWVCP_FU12	<i>F. incarnatum-equiseti</i> complex; NRRL 26417	98.75%
GXGASS_FU1	<i>F. incarnatum-equiseti</i> complex; NRRL 20697	95.01%
GXGASS_FU2	<i>F. incarnatum-equiseti</i> complex; NRRL 20697	95.06%
GXGVCL_FU2	<i>F. incarnatum-equiseti</i> complex; NRRL 13402	97.13%
GXGVCL_FU3	N/A	
GXNVCR_FU1	N/A	
HNLASP_FU2	<i>F. incarnatum-equiseti</i> complex; NRRL 26417	94.74%
GXNVCP_FU2	N/A	
FJASL_FU2	<i>Fusarium</i> sp. (<i>F. incarnatum-equiseti</i> complex)	98.03%
FJASL_FU3	<i>F. sambucinum</i> complex; NRRL 3299	81.10%
FJASP_FU3	<i>F. incarnatum-equiseti</i> complex (NRRL 28029)	92.98%
HNWVCP_FU13	<i>F. incarnatum-equiseti</i> complex; NRRL 20697	94.25%
HNWVCP_FU14	<i>F. incarnatum-equiseti</i> complex; NRRL 20697	96.65%
GZASR_FU9	<i>F. incarnatum-equiseti</i> complex; NRRL 20423	98.33%
HNLASS_FU1	<i>F. nisikadoi</i> complex; NRRL 28387	96.36%
GZASS_FU5	<i>Gibberella fujikuroi</i> complex (NRRL 13602)	98.44%
HNLASS_FU2	<i>Gibberella fujikuroi</i> complex (NRRL 13602)	98.59%
GZASL_FU4	<i>F. fujikuroi</i> , Asian subclade; NRRL 13566	99.80%
FJASP_FU4	<i>Gibberella fujikuroi</i> complex (NRRL 47473)	98.28%
FJASL_FU4	<i>F. nisikadoi</i> complex; NRRL 28387	96.01%
GXGVCL_FU4	<i>F. fujikuroi</i> , Asian subclade; NRRL 13566	99.80%
GXNVCP_FU3	<i>F. fujikuroi</i> , Asian subclade; NRRL 13566	99.80%
GZASL_FU5	<i>F. fujikuroi</i> , Asian subclade; NRRL 13566	99.80%
GXGVCS_FU2	<i>F. fujikuroi</i> , Asian subclade; NRRL 13566	99.90%
FJASL_FU5	<i>Gibberella fujikuroi</i> complex (NRRL 47473)	99.53%
FJASL_FU6	<i>F. fujikuroi</i> , Asian subclade; NRRL 13566	99.80%
FJASL_FU7	<i>F. fujikuroi</i> , Asian subclade; NRRL 13566	99.69%
HNWVCP_FU15	<i>Gibberella fujikuroi</i> species complex (NRRL 13602)	98.28%
GXGVCS_FU3	<i>F. fujikuroi</i> , Asian subclade; NRRL 22944	92.21%
GXGASS_FU3	<i>F. fujikuroi</i> , Asian subclade; NRRL 13566	100.00%
GXGASL_FU1	<i>F. fujikuroi</i> , Asian subclade; NRRL 13566	99.80%
GXGASS_FU4	<i>F. fujikuroi</i> , Asian subclade; NRRL 13566	99.90%
GXGASS_FU5	<i>F. fujikuroi</i> , Asian subclade; NRRL 13566	99.90%
GXGASS_FU6	<i>F. fujikuroi</i> , Asian subclade; NRRL 13566	99.80%
GXGASS_FU7	<i>F. fujikuroi</i> , Asian subclade; NRRL 13566	99.90%
GXGVCS_FU4	<i>Gibberella fujikuroi</i> complex (NRRL 13602)	97.97%
HNLASS_FU3	<i>Gibberella fujikuroi</i> complex (NRRL 13602)	98.59%
HNLASS_FU4	<i>Gibberella fujikuroi</i> complex (NRRL 13602)	98.13%
HNLASS_FU5	<i>Gibberella fujikuroi</i> complex (NRRL 13602)	98.13%
GXGVCS_FU5	<i>F. fujikuroi</i> , Asian subclade; NRRL 13566	99.59%
FJASF_FU1	<i>F. fujikuroi</i> , Asian subclade; NRRL 13566	99.90%
GXGASL_FU2	<i>F. fujikuroi</i> , Asian subclade; NRRL 13566	99.90%
FJASP_FU5	<i>Gibberella fujikuroi</i> complex (NRRL 13602)	97.97%
GXGASS_FU8	<i>F. fujikuroi</i> , Asian subclade; NRRL 13566	99.69%
GXGVCL_FU5	<i>F. fujikuroi</i> , Asian subclade; NRRL 13566	98.58%
GXGASS_FU9	<i>F. fujikuroi</i> , Asian subclade; NRRL 13566	99.60%
FJASP_FU6	<i>Gibberella fujikuroi</i> complex (NRRL 47473)	99.22%
FJASP_FU7	<i>Gibberella fujikuroi</i> complex (NRRL 47473)	98.59%
FJASP_FU8	<i>Gibberella fujikuroi</i> complex (NRRL 47473)	98.28%
FJASF_FU2	<i>F. fujikuroi</i> , Asian subclade; NRRL 13566	99.90%
GXGVCF_FU1	<i>F. fujikuroi</i> , Asian subclade; NRRL 13566	99.29%
GXGVCP_FU6	<i>Gibberella fujikuroi</i> complex (NRRL 13602)	98.59%
GXGVCP_FU7	<i>F. fujikuroi</i> , Asian subclade; NRRL 13566	99.90%
GXGVCL_FU6	<i>F. fujikuroi</i> , Asian subclade; NRRL 13566	99.90%
GXGVCL_FU7	<i>Gibberella fujikuroi</i> complex (NRRL 13602)	98.28%
GXGVCL_FU8	<i>F. fujikuroi</i> , Asian subclade; NRRL 13566	100.00%

Supplementary Table 2 Continued.

Isolate no.	Identification Result from <i>Fusarium</i> MLST database*	Similarity
GXGASS_FU10	<i>F. fujikuroi</i> , Asian subclade; NRRL 13566	100.00%
GXGASS_FU11	<i>F. sambucinum</i> complex; NRRL 31084;	99.80%
GXGASS_FU12	<i>F. fujikuroi</i> , Asian subclade; NRRL 13566	99.80%
GXGASS_FU13	<i>F. fujikuroi</i> , Asian subclade; NRRL 13566	99.80%
GXGASS_FU14	<i>F. fujikuroi</i> , Asian subclade; NRRL 13566	99.90%
GXGASS_FU15	<i>F. fujikuroi</i> , Asian subclade; NRRL 13566	99.90%
GXGASP_FU3	<i>Gibberella fujikuroi</i> complex (NRRL 13602)	98.44%
GXGVCP_FU8	<i>F. fujikuroi</i> , Asian subclade; NRRL 13566	99.69%
GXGASP_FU4	<i>Gibberella fujikuroi</i> complex (NRRL 13602)	99.06%
HNWASP_FU10	<i>Gibberella fujikuroi</i> complex (NRRL 13602)	97.97%
GZASS_FU6	<i>F. fujikuroi</i> , Asian subclade; NRRL 13566	99.69%
HNWASR_FU19	<i>F. sambucinum</i> complex; NRRL 31084	100.00%
HNLASF_FU1	<i>F. sambucinum</i> complex; NRRL 31084	99.69%
HNLASL_FU5	<i>F. sambucinum</i> complex; NRRL 13818	100.00%
HNWASS_FU15	<i>F. sambucinum</i> complex; NRRL 31084	99.49%
HNWASS_FU17	<i>F. graminearum</i> (<i>F. sambucinum</i> complex)	99.88%
HNWASS_FU16	<i>F. graminearum</i> (<i>F. sambucinum</i> complex)	99.89%
GXNVCR_FU2	<i>F. incarnatum-equiseti</i> complex; NRRL 26417	92.32%
HNLASP_FU3	<i>F. incarnatum-equiseti</i> complex; NRRL 26417	97.47%
GXGVCL_FU9	<i>F. incarnatum-equiseti</i> complex; NRRL 32175	99.28%
FJASL_FU8	<i>F. incarnatum-equiseti</i> complex; NRRL 26417	98.96%
FJASL_FU9	<i>F. incarnatum-equiseti</i> complex; NRRL 26417	99.38%
FJASP_FU9	<i>F. incarnatum-equiseti</i> complex; NRRL 26417	99.38%
FJASP_FU10	N/A	
HNWVCL_FU19	<i>F. fujikuroi</i> , Asian subclade; NRRL 22944	100.00%
HNWVCL_FU20	<i>F. incarnatum-equiseti</i> complex; NRRL 26417	99.05%
FJASS_FU1	<i>F. sambucinum</i> complex; NRRL 3299	81.47%
GXGVCP_FU9	<i>F. incarnatum-equiseti</i> complex; NRRL 26417	99.39%
GXNVCL_FU1	<i>F. incarnatum-equiseti</i> complex; NRRL 26417	99.80%
GXNVCP_FU4	N/A	
HNWVCF_FU19	<i>F. incarnatum-equiseti</i> complex; NRRL 26417	99.27%
HNWVCP_FU16	<i>F. incarnatum-equiseti</i> complex; NRRL 26417	99.07%
GXGVCS_FU6	<i>F. incarnatum-equiseti</i> complex; NRRL 26417	99.28%
HNWVCS_FU17	<i>F. incarnatum-equiseti</i> complex; NRRL 26417	98.64%
GXGVCL_FU10	<i>F. incarnatum-equiseti</i> complex; NRRL 26417	99.27%
FJASP_FU11	<i>F. incarnatum-equiseti</i> complex; NRRL 26417	98.96%
FJASS_FU2	<i>F. incarnatum-equiseti</i> complex; NRRL 26417	99.37%
GXGASP_FU5	N/A	
HNWASS_FU18	<i>F. incarnatum-equiseti</i> complex; NRRL 26417	98.98%
GXGVCL_FU11	<i>F. incarnatum-equiseti</i> complex; NRRL 26417	99.27%
GXGVCL_FU12	<i>F. incarnatum-equiseti</i> complex; NRRL 26417	86.79%
GXGVCL_FU13	<i>F. incarnatum-equiseti</i> complex; NRRL 26417	87.34%
GXGASP_FU6	<i>F. incarnatum-equiseti</i> complex; NRRL 26417	99.37%
HNWVCP_FU17	<i>F. incarnatum-equiseti</i> complex; NRRL 26417	99.16%
FJASF_FU3	<i>F. kyushuense</i> (<i>F. sambucinum</i> complex)	98.69%
GZASR_FU10	<i>F. nisikadoi</i> complex; NRRL 28387	100.00%
GZASR_FU11	<i>F. oxysporum</i> complex; NRRL 36408	100.00%
HNLASR_FU8	<i>F. nisikadoi</i> complex; NRRL 28387	99.90%
HNLASR_FU9	<i>F. nisikadoi</i> complex; NRRL 28387	99.69%
FJASR_FU2	<i>F. oxysporum</i> complex; NRRL 20433	99.39%
GXGASP_FU7	<i>F. fujikuroi</i> , Asian subclade; NRRL 13566	99.90%
FJASR_FU3	<i>F. oxysporum</i> complex; NRRL 22518	99.84%
FJASL_FU10	<i>F. oxysporum</i> complex; NRRL 22518	99.84%
HNWVCP_FU18	<i>F. oxysporum</i> complex; NRRL 31495	99.84%
HNWASR_FU20	<i>F. nisikadoi</i> complex; NRRL 28387	100.00%
GXGASP_FU8	<i>F. oxysporum</i> complex; NRRL 32881	100.00%
GXGASP_FU9	<i>F. oxysporum</i> complex; NRRL 20433	99.80%
HNLASR_FU10	<i>F. nisikadoi</i> complex; NRRL 28387	99.90%
GXNVCR_FU3	<i>F. oxysporum</i> complex; NRRL 25387	99.28%

Supplementary Table 2 Continued.

Isolate no.	Identification Result from <i>Fusarium</i> MLST database*	Similarity
GXNVCR_FU4	N/A	
GXNVCR_FU5	N/A	
GXNVCL_FU2	<i>F. oxysporum</i> complex; NRRL 32881	99.84%
GXNVCR_FU6	N/A	
GXGVCS_FU7	<i>F. nisikadoi</i> complex; NRRL 28387	100.00%
GXNVCR_FU7	<i>F. oxysporum</i> complex; NRRL 20433	99.80%
GXGVCP_FU10	<i>F. fujikuroi</i> , Asian subclade; NRRL 13566	99.80%
GXGVCL_FU14	<i>F. nisikadoi</i> complex; NRRL 28387	96.42%
HNWASF_FU11	<i>F. nisikadoi</i> complex; NRRL 28387	96.12%
GZASL_FU6	<i>Gibberella fujikuroi</i> complex; NRRL 13308	97.62%
GZASL_FU7	<i>Gibberella fujikuroi</i> complex; NRRL 13308	98.01%
GZASR_FU12	<i>F. fujikuroi</i> , Asian subclade; NRRL 13566	100.00%
GZASR_FU13	<i>F. proliferatum</i> (<i>F. fujikuroi</i> complex)	98.75%
GZASS_FU7	<i>F. proliferatum</i> (<i>F. fujikuroi</i> complex)	98.78%
GZASR_FU14	<i>Fusarium nisikadoi</i> complex; NRRL 28387	99.50%
HNWVCF_FU20	<i>F. proliferatum</i> (<i>F. fujikuroi</i> complex)	98.27%
HNWVCR_FU17	<i>F. proliferatum</i> (<i>F. fujikuroi</i> complex)	99.05%
HNWVCP_FU19	<i>F. fujikuroi</i> , Asian subclade; NRRL 22944	99.59%
HNWVCP_FU20	<i>F. nisikadoi</i> complex; NRRL 28387	96.42%
HNWVCS_FU18	<i>F. fujikuroi</i> , Asian subclade; NRRL 22944	99.69%
HNWASS_FU19	<i>F. fujikuroi</i> , Asian subclade; NRRL 22944	99.59%
HNWASS_FU20	<i>F. proliferatum</i> (<i>F. fujikuroi</i> complex)	98.50%
HNWASL_FU8	<i>F. proliferatum</i> (<i>F. fujikuroi</i> complex)	98.17%
HNWASL_FU9	<i>F. proliferatum</i> (<i>F. fujikuroi</i> complex)	98.42%
GXGVCP_FU11	<i>F. proliferatum</i> (<i>F. fujikuroi</i> complex)	98.90%
GXGVCP_FU12	<i>F. proliferatum</i> (<i>F. fujikuroi</i> complex)	98.68%
GXGVCP_FU13	<i>F. proliferatum</i> (<i>F. fujikuroi</i> complex)	98.76%
GXGVCL_FU15	<i>F. proliferatum</i> (<i>F. fujikuroi</i> complex)	98.75%
GXGVCL_FU16	<i>F. fujikuroi</i> , Asian subclade; NRRL 22944	99.70%
GXGASS_FU16	<i>F. proliferatum</i> (<i>F. fujikuroi</i> complex)	97.55%
GXGASL_FU3	<i>F. proliferatum</i> (<i>F. fujikuroi</i> complex)	98.68%
GXGVCL_FU17	<i>F. proliferatum</i> (<i>F. fujikuroi</i> complex)	98.75%
GXGVCS_FU8	<i>F. proliferatum</i> (<i>F. fujikuroi</i> complex)	98.62%
GXGVCS_FU9	<i>F. proliferatum</i> (<i>F. fujikuroi</i> complex)	98.80%
GXGVCS_FU10	<i>F. proliferatum</i> (<i>F. fujikuroi</i> complex)	98.55%
HNLASL_FU6	<i>F. proliferatum</i> (<i>F. fujikuroi</i> complex)	98.90%
HNLASL_FU7	<i>F. fujikuroi</i> , Asian subclade; NRRL 22944	99.59%
HNLASR_FU11	<i>F. nisikadoi</i> complex; NRRL 28387	100.00%
HNLASF_FU2	<i>F. proliferatum</i> (<i>F. fujikuroi</i> complex)	97.59%
HNLASF_FU3	<i>Gibberella fujikuroi</i> complex; NRRL 13308	97.79%
HNWVCR_FU18	<i>F. nisikadoi</i> complex; NRRL 28387	96.12%
GXGVCS_FU11	<i>Gibberella fujikuroi</i> complex; NRRL 25195	98.76%
GXGVCS_FU12	<i>F. fujikuroi</i> , Asian subclade; NRRL 13566	99.69%
GXGASS_FU17	<i>F. fujikuroi</i> , Asian subclade; NRRL 22944	99.59%
HNLASL_FU8	<i>F. proliferatum</i> (<i>F. fujikuroi</i> complex)	98.55%
HNWASL_FU11	<i>F. proliferatum</i> (<i>F. fujikuroi</i> complex)	97.70%
GXNVCS_FU1	<i>F. proliferatum</i> (<i>F. fujikuroi</i> complex)	98.41%
GXNVCL_FU3	N/A	
GXNVCL_FU4	N/A	
GXNVCP_FU5	<i>Gibberella fujikuroi</i> complex; NRRL 13308	97.67%
GXNVCP_FU6	<i>Gibberella fujikuroi</i> complex; NRRL 13602	93.51%
GXNVCL_FU5	<i>Gibberella fujikuroi</i> complex; NRRL 13602	94.13%
HNWVCP_FU21	<i>F. fujikuroi</i> , Asian subclade; NRRL 22944	99.90%
HNWASS_FU21	<i>F. proliferatum</i> (<i>F. fujikuroi</i> complex)	98.82%
HNWASS_FU22	<i>F. proliferatum</i> (<i>F. fujikuroi</i> complex)	98.69%
HNWASS_FU23	<i>F. fujikuroi</i> , Asian subclade; NRRL 22944	99.90%
HNWASS_FU24	<i>F. proliferatum</i> (<i>F. fujikuroi</i> complex)	98.38%
HNWASL_FU10	<i>F. fujikuroi</i> , Asian subclade; NRRL 22944	98.24%
HNWASS_FU25	<i>F. fujikuroi</i> , Asian subclade; NRRL 22944	99.69%

Supplementary Table 2 Continued.

Isolate no.	Identification Result from <i>Fusarium</i> MLST database*	Similarity
HNWVCL_FU21	<i>F. fujikuroi</i> , Asian subclade; NRRL 22944	99.90%
HNWVCS_FU19	<i>F. proliferatum</i> (<i>F. fujikuroi</i> complex)	98.15%
HNWVCS_FU20	<i>F. fujikuroi</i> , Asian subclade; NRRL 22944	99.90%
HNWVCS_FU21	<i>F. fujikuroi</i> , Asian subclade; NRRL 22944	99.58%
GXNVCL_FU6	<i>Gibberella fujikuroi</i> complex; NRRL 13308	97.00%
HNWVCS_FU22	<i>F. fujikuroi</i> , Asian subclade; NRRL 22944	99.90%
HNWVCP_FU22	<i>F. solani</i> complex NRRL 32542	98.95%
GXNVCR_FU8	<i>F. solani</i> complex; NRRL 43529	99.79%
FJASP_FU12	<i>Fusarium solani</i> complex; NRRL 32791	98.68%
FJASP_FU13	<i>F. solani</i> complex; NRRL 32791	99.26%
HNWVCP_FU23	<i>F. solani</i> complex; NRRL 43529	100.00%
HNWVCR_FU19	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex); NRRL 36147	98.97%
HNWVCR_FU20	<i>F. avenaceum</i> (<i>F. tricinctum</i> complex)	96.61%
HNWVCP_FU24	<i>F. avenaceum</i> (<i>F. tricinctum</i> complex)	95.90%
HNWASP_FU12	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex); NRRL 36147	96.57%
HNWASR_FU21	<i>F. avenaceum</i> (<i>F. tricinctum</i> complex)	96.54%
HNWASR_FU22	<i>F. avenaceum</i> (<i>F. tricinctum</i> complex)	96.56%
HNWASR_FU23	<i>F. avenaceum</i> (<i>F. tricinctum</i> complex)	96.60%
HNWASP_FU13	<i>F. avenaceum</i> (<i>F. tricinctum</i> complex)	96.54%
HNWVCL_FU22	<i>F. avenaceum</i> (<i>F. tricinctum</i> complex)	96.21%
HNWVCL_FU23	<i>F. avenaceum</i> (<i>F. tricinctum</i> complex)	96.53%
HNWVCR_FU21	<i>F. avenaceum</i> (<i>F. tricinctum</i> complex)	96.54%
GXGASS_FU18	<i>F. avenaceum</i> (<i>F. tricinctum</i> complex)	97.05%
HNWASP_FU11	<i>F. tricinctum</i> complex; NRRL 25481	94.80%
HNWASR_FU24	<i>Fusarium</i> sp. (<i>F. tricinctum</i> complex); NRRL 45994	98.69%
HNWVCS_FU1	<i>F. fujikuroi</i> , Asian subclade; NRRL 13566	99.40%
HNWVCS_FU2	N/A	

*N/A -Could not be assigned to any level.

Supplementary Table 3 Checklist.

Fungal species*	Family	Locality	Reference
<i>Astragalus sinicus</i>			
<i>Albifimbria verrucaria</i>	Stachybotryaceae	China	This study
<i>Alternaria alternata</i>	Pleosporaceae	China	This study
<i>Alternaria astragalicola</i>	Pleosporaceae	China	This study
<i>Alternaria gaisen</i>	Pleosporaceae	China	This study
<i>Alternaria guizhouensis</i>	Pleosporaceae	China	This study
<i>Alternaria henanensis</i>	Pleosporaceae	China	This study
<i>Arthrimum arundinis</i>	Apiosporaceae	China	This study
<i>Botrytis cinerea</i>	Sclerotiniaceae	China	Zhang (2006), This study
<i>Cercospora astragali</i>	Mycosphaerellaceae	China, Taiwan	Tai (1979), Hsieh & Goh (1990)
<i>Cladosporium astragali</i>	Cladosporiaceae	Japan	
<i>Cladosporium nigrellum</i>	Cladosporiaceae	China	Zhang (2003)
<i>Colletotrichum fructicola</i>	Glomerellaceae	China	This study
<i>Colletotrichum truncatum</i>	Glomerellaceae	China	This study
<i>Clonostachys eriocamporesii</i>	Bionectriaceae	China	This study
<i>Clonostachys ochroleuca</i>	Bionectriaceae	China	This study
<i>Clonostachys rosae</i>	Bionectriaceae	China	This study
<i>Diaporthe longicolla</i>	Diaporthaceae	China	This study
<i>Epicoccum astragali</i>	Didymellaceae	China	This study
<i>Epicoccum henanense</i>	Didymellaceae	China	This study
<i>Epicoccum laticollum</i>	Didymellaceae	China	This study
<i>Epicoccum layuense</i>	Didymellaceae	China	This study
<i>Epicoccum nigrum</i>	Didymellaceae	China	Tai (1979)
<i>Epicoccum rosae</i>	Didymellaceae	China	This study
<i>Erysiphe astragali</i>	Erysiphaceae	China, Japan, Taiwan	Amano (1986), Tai (1979), Peregrine & Siddiqi (1972)
<i>Erysiphe pisi</i>	Erysiphaceae	China, Japan, Taiwan, Korea	Amano (1986), Shin (2000), Cho & Shin (2004), Peregrine & Siddiqi (1972), Sawada (1959)
<i>Erysiphe polygoni</i>	Erysiphaceae	China	Tai (1979)
<i>Fusarium graminearum</i>	Nectriaceae	China	Tai (1979)
<i>Leptosphaerulina americana</i>	Didymellaceae	China	This study
<i>Neovularia nomuriana</i>	Incertae sedis	China, Japan	Tai (1979), Braun (1998), Videira et al. (2016, 2017)
<i>Oidium</i> sp.	Erysiphaceae	Australia	Amano (1986)
<i>Peronospora aestivalis</i>	Peronosporaceae	Japan	
<i>Physoderma trifolii</i>	Physodermataceae	China	Tai (1979)
<i>Pseudophthomyces chartarum</i>	Didymosphaeriaceae	China	This study
<i>Sclerotinia sclerotiorum</i>	Sclerotiniaceae	China Taiwan	Tai (1979), Peregrine & Siddiqi (1972)
<i>Sclerotinia trifoliorum</i>	Sclerotiniaceae	Japan, China	Richardson (1990), Tai (1979)

Supplementary Table 3 Continued.

Fungal species*	Family	Locality	Reference
<i>Stemphylium astragali</i>	Pleosporaceae	China, Japan, Korea	Tianyu (2009), Camara et al. (2002), Cho & Shin (2004), Yu (2001), This study
<i>Uromyces pisi-sativi</i>	Pucciniaceae	China, Japan	Tai (1979), Guo & Wang (1986), Guyot (1957), Zhuang (2005a), Zhuang (2005b)
<i>Vicia villosa</i>			
<i>Acremoniella atra</i>	Incertae sedis	Oregon	Shaw (1973)
<i>Alternaria alternata</i>	Pleosporaceae	Oregon, China	Shaw (1973), This study
<i>Alternaria gaisen</i>	Pleosporaceae	China	This study
<i>Alternaria henanensis</i>	Pleosporaceae	China	This study
<i>Ascochyta</i> sp	Didymellaceae	Bulgaria, Mississippi, Washington Kentucky	Peever et al. (2007), Parris (1959), Valleau (1950a, b)
<i>Septoria viciae</i>	Mycosphaerellaceae	Poland	Mulenko et al. (2008)
<i>Ascochyta viciae-pannonicae</i>	Didymellaceae	Poland	Mulenko et al. (2008)
<i>Ascochyta viciae-villosae</i>	Didymellaceae	Czechoslovakia, Poland	Watson (1971), Fatehi & Bridge (1998), Mulenko et al. (2008)
<i>Aspergillus</i> sp	Aspergillaceae	Oregon	Shaw (1973)
<i>Aureobasidium pullulans</i>	Sacotheciaceae	Oregon	Shaw (1973)
<i>Botrytis cinerea</i>	Sclerotiniaceae	China, Oregon	Zhuang (2005), Shaw (1973)
<i>Botrytis fabae</i>	Sclerotiniaceae	Norway, Poland, United Kingdom, USSR	Richardson (1990)
<i>Botrytis</i> sp.	Sclerotiniaceae	Wisconsin	Greene (1964)
<i>Cercospora</i> sp.	Mycosphaerellaceae	Mississippi	Hare (1954)
<i>Chaetomium</i> sp.	Chaetomiaceae	Oregon	Shaw (1973)
<i>Cladosporium cladosporioides</i>	Cladosporiaceae	Oregon	Morgan-Jones & McKemy (1992)
<i>Clonostachys rosea</i>	Bionectriaceae	China	This study
<i>Colletotrichum destructivum</i>	Glomerellaceae	China	This study
<i>Colletotrichum trifolii</i>	Glomerellaceae	North Carolina	Grand (1985)
<i>Colletotrichum truncatum</i>	Glomerellaceae	China	This study
<i>Colletotrichum viciae</i>	Glomerellaceae	China, Louisiana, Maryland, Mississippi, Oklahoma, Pennsylvania, Wisconsin	Tai (1979), Anonymous 1960, Parris (1959), Preston (1945),
<i>Colletotrichum villosum</i>	Glomerellaceae	Florida, Georgia, Illinois, Louisiana, Mississippi, Oklahoma, Tennessee	Anonymous 1960, Boewe (1964), Richardson (1990), Hare (1954), Preston (1947), Allison et al. (1950)
<i>Colletotrichum villosum</i>	Glomerellaceae	Wisconsin	Greene (1949)
<i>Diaporthe viciae</i>	Diaporthaceae	China	This study
<i>Dictyochoaeta fertilis</i>	Chaetosphaeriaceae	North Carolina	Grand (1985)
<i>Didymella pinodes</i>	Didymellaceae	China, Georgia, New York, South Carolina, Washington	Tai (1979), Zhuang (2005), Anonymous (1960)

Supplementary Table 3 Continued.

Fungal species*	Family	Locality	Reference
<i>Didymella pisi</i>	Didymellaceae	China, Idaho, Illinois, Mississippi, Oklahoma, Oregon, Sweden, Tennessee, Washington	Tai (1979), Shaw (1973), Boewe (1964), Parris (1959), Preston (1945), Buchanan (1987), Allison et al. (1950)
<i>Didymella</i> sp.	Didymellaceae	Washington	Peever et al. (2007)
<i>Epicoccum henanense</i>	Didymellaceae	China	This study
<i>Epicoccum layuense</i>	Didymellaceae	China	This study
<i>Epicoccum nigrum</i>	Didymellaceae	Oregon	Shaw (1973)
<i>Epicoccum viciae-villosae</i>	Didymellaceae	China	This study
<i>Erysiphe baeumleri</i>	Erysiphaceae	Poland, Romania, Ukraine	Ruszkiewicz (2000), Ruszkiewicz-Michalska & Michalski (2005), Mulenko (2008), Braun (1995) Amano (Hirata) (1986)
<i>Erysiphe pisi</i>	Erysiphaceae	China	Zhuang (2005)
<i>Erysiphe pisi</i> var. <i>pisi</i>	Erysiphaceae	France, Germany, Hungary, Romania, Sweden, Switzerland, USSR	Braun (1995), Bolay (2005)
<i>Erysiphe pisi</i> var. <i>pisi</i>	Erysiphaceae	Switzerland	Braun (1995)
<i>Erysiphe polygoni</i>	Erysiphaceae	China, Mississippi, Texas	Tai (1979), Parris (1959), Anonymous (1960)
<i>Erysiphe viciae-unijugae</i>	Erysiphaceae	Korea	Shin (2000), Cho & Shin (2004),
<i>Fusarium oxysporum</i>	Nectriaceae	China	Zhuang (2005)
<i>Fusarium roseum</i>	Nectriaceae	Oregon	Shaw (1973)
<i>Fusarium graminearum</i>	Nectriaceae	Czechoslovakia	Richardson (1990)
<i>Gloeosporium americanum</i>	Drepanopezizaceae	Oklahoma	Preston (1945)
<i>Kabatiella nigricans</i>	Sacotheciaceae	Georgia, Louisiana, Mississippi, Montana, North Carolina, New York, Ohio Oklahoma, Oregon South Carolina, Tennessee Wisconsin	Anonymous (1960), Parris (1959), Preston (1947), Shaw (1973)
<i>Lasiodiplodia mediterranea</i>	Botryosphaeriaceae	China	This study
<i>Leveillula taurica</i>	Erysiphaceae	USSR	Amano (Hirata) (1986)
<i>Neofusicoccum parvum</i>	Botryosphaeriaceae	China	This study
<i>Ochrocladosporium elatum</i>	Pleosporales	Oregon	Shaw (1973)
<i>Oidium</i> sp.	Erysiphaceae	China, Portugal, Greece, Spain, England	Zheng & Yu (1987), Amano (Hirata) (1986)
<i>Peyronellaea lethalis</i>	Didymellaceae	Italy	Sisic et al. (2018)
<i>Peronospora viciae</i>	Peronosporaceae	Bulgaria, China, Czech Republic, Illinois, Kansas, Mississippi, North Carolina, Poland, South Carolina, Australia, Central Asia, Germany	Vanev et al. (1993), Tai (1979), Yu (1998), Zhuang (2005), Muller & Kokes (2008), Boewe (1964), Rogerson (1958), Anonymous (1960), Parris (1959), Ruszkiewicz-Michalska & Michalski (2005), Mulenko et al. (2008), Cook & Dubé (1989), Gaponenko (1972), Constantinescu (1991)

Supplementary Table 3 Continued.

Fungal species*	Family	Locality	Reference
<i>Phyllosticta phaseolina</i>	Phyllostictaceae	China	Zhuang (2005)
<i>Plectosperella cucumerina</i>	Plectosphaerellaceae	China	This study
<i>Pseudoidium</i> sp.	Erysiphaceae	Russia	Rusanov & Bulgakov (2008)
<i>Pseudopeziza medicaginis</i>	Ploettnerulaceae	Mississippi	Anonymous (1960), Parris (1959)
<i>Ramularia schwarzhiana</i>	Mycosphaerellaceae	Portugal, Sweden, California, Idaho, Oregon	de Sousa Dias & Lucas (1980), Anonymous (1960)
<i>Ramularia sphaeroidea</i>	Mycosphaerellaceae	Austria, California, Czech Republic, Denmark, Estonia, Europe, France, Germany, Hungary, Idaho, Italy, New Zealand, Oregon, Poland, Portugal, Russia, Sweden, Turkmenistan, Ukraine, Washington, California, Wisconsin	Braun (1998), McKenzie & Dingley (1996), Mulenko et al. (2008), French (1989), Greene (1953), Koike et al. (2004), Videira et al. (2016)
<i>Sclerotinia</i> sp.	Sclerotiniaceae	California, Maryland	Anonymous (1960), French (1989), Morgan (1964)
<i>Septoria pisi</i>	Mycosphaerellaceae	Georgia	Anonymous (1960)
<i>Stemphylium vesicarium</i>	Pleosporaceae	China	Yan et al. (2019)
<i>Uromyces viciae-fabae</i>	Pucciniaceae	China, Hungary, Poland, Romania, Ukraine, Greece, Morocco, Bulgaria, China, Germany, Poland, Romania, Turkey	Tai (1979), Guyot (1957), Pantidou (1973), Denchev (1995), Guo & Wang (1986), Cao et al. (1999, 2000), Zhuang (2005), Braun (1982), Mulenko & Ruskiewicz-Michalska (2008), Savulescu (1953), Bahcecioglu & Kabaktepe (2012)
<i>Uromyces fischeri-eduardi</i>	Pucciniaceae	Bulgaria, Turkey	Denchev (1995), Bahcecioglu & Kabaktepe (2012)
<i>Uromyces heimerlianus</i>	Pucciniaceae	Poland	Guyot (1957)

* Records obtain from this study are **bold**

Supplementary Table 4 OTU Table.

OTU_ID	Phylum	Class	Order	Family	Genus	Species
OTU_49	Others	Others	Others	Others	Others	Others
OTU_28	Others	Others	Others	Others	Others	Others
OTU_475	Others	Others	Others	Others	Others	Others
OTU_267	Others	Others	Others	Others	Others	Others
OTU_497	Others	Others	Others	Others	Others	Others
OTU_29	Others	Others	Others	Others	Others	Others
OTU_521	Others	Others	Others	Others	Others	Others
OTU_509	p__	c__	o__	f__	g__	s__
OTU_526	p__Ascomycota	c__Leotiomyces	o__Helotiales	Others	Others	Others
OTU_173	p__Ascomycota	c__Sordariomyces	o__Hypocreales	f__Nectriaceae	g__Fusarium	Others
OTU_126	p__Ascomycota	c__Laboulbeniomyces	Others	Others	Others	Others
OTU_365	p__Ascomycota	c__Dothideomyces	o__Capnodiales	f__Cladosporiaceae	g__Cladosporium	Others
OTU_381	p__Ascomycota	c__Sordariomyces	o__Magnaporthales	f__Magnaporthaceae	g__Magnaporthe	Others
OTU_241	p__Ascomycota	c__Sordariomyces	o__Glomerellales	f__Glomerellaceae	g__Colletotrichum	s__Colletotrichum tofieldiae
OTU_496	p__Ascomycota	c__Dothideomyces	o__Pleosporales	Others	Others	Others
OTU_263	p__Ascomycota	c__Sordariomyces	o__Sordariales	Others	Others	Others
OTU_589	p__Ascomycota	c__Saccharomyces	o__Saccharomycetales	f__Debaryomycetaceae	g__unidentified_Debaryomycetaceae	Others
OTU_73	p__Ascomycota	c__Sordariomyces	o__Sordariales	f__Chaetomiaceae	g__Chaetomium	s__Chaetomium globosum
OTU_83	p__Ascomycota	c__Eurotiomyces	o__Chaetothyriales	f__Trichomeriaceae	Others	Others
OTU_260	p__Ascomycota	c__Sordariomyces	o__Hypocreales	Others	Others	Others
OTU_399	p__Ascomycota	c__Dothideomyces	o__Venturiales	f__Symptoventuriaceae	g__Ochroconis	Others
OTU_276	p__Ascomycota	c__Saccharomyces	o__Saccharomycetales	f__Saccharomycetaceae	g__Saccharomyces	s__
OTU_3	p__Ascomycota	c__Dothideomyces	o__Pleosporales	f__Pleosporaceae	g__Alternaria	s__Alternaria alternata
OTU_224	p__Ascomycota	c__Sordariomyces	o__Glomerellales	f__Plectosphaerellaceae	g__Plectosphaerella	s__
OTU_417	p__Ascomycota	c__Dothideomyces	o__Capnodiales	Others	Others	Others
OTU_154	p__Ascomycota	c__Leotiomyces	o__Helotiales	Others	Others	Others
OTU_84	p__Ascomycota	c__Sordariomyces	o__Hypocreales	f__Nectriaceae	g__Fusarium	Others
OTU_262	p__Ascomycota	Others	Others	Others	Others	Others
OTU_197	p__Ascomycota	c__Taphrinomyces	o__Taphrinales	f__Protomycetaceae	g__Protomyces	s__Protomyces inouyei
OTU_492	p__Ascomycota	c__Sordariomyces	o__Sordariales	f__Chaetomiaceae	g__Chaetomium	Others
OTU_483	p__Ascomycota	Others	Others	Others	Others	Others
OTU_182	p__Ascomycota	c__Pezizomyces	o__Pezizales	f__Ascodesmidaceae	Others	Others
OTU_481	p__Ascomycota	c__Sordariomyces	o__Magnaporthales	f__Magnaporthaceae	g__Magnaporthe	Others
OTU_471	p__Ascomycota	c__Dothideomyces	o__Pleosporales	Others	Others	Others
OTU_411	p__Ascomycota	c__Saccharomyces	o__Saccharomycetales	f__Pichiaceae	g__Pichia	s__Pichia kudriavzevii
OTU_5	p__Ascomycota	c__Leotiomyces	o__Helotiales	f__Sclerotiniaceae	g__Sclerotinia	s__
OTU_74	p__Ascomycota	c__Laboulbeniomyces	o__Pyxidiophorales	f__Pyxidiophoraceae	g__Pyxidiophora	s__Pyxidiophora arvernensis

Supplementary Table 4 Continued.

OTU_ID	Phylum	Class	Order	Family	Genus	Species
OTU_164	p__Ascomycota	c__Sordariomycetes	o__Glomerellales	f__Plectosphaerellaceae	g__ <i>Plectosphaerella</i>	s__
OTU_119	p__Ascomycota	c__Saccharomycetes	o__Saccharomycetales	f__Saccharomycetaceae	g__ <i>Eremothecium</i>	Others
OTU_568	p__Ascomycota	c__Sordariomycetes	o__Glomerellales	f__Plectosphaerellaceae	g__ <i>Lectera</i>	s__ <i>Lectera colletotrichoides</i>
OTU_311	p__Ascomycota	c__Sordariomycetes	o__Diaporthales	f__Diaporthaceae	g__ <i>Diaporthe</i>	s__ <i>Diaporthe amygdali</i>
OTU_13	p__Ascomycota	c__Sordariomycetes	o__Hypocreales	f__Nectriaceae	g__ <i>Fusarium</i>	Others
OTU_513	p__Ascomycota	c__Saccharomycetes	o__Saccharomycetales	Others	Others	Others
OTU_68	p__Ascomycota	c__Sordariomycetes	o__Magnaporthales	f__Magnaporthaceae	g__ <i>Magnaporthe</i>	s__ <i>Magnaporthe oryzae</i>
OTU_527	p__Ascomycota	c__Eurotiomycetes	o__Chaetothyriales	f__Trichomeriaceae	g__ <i>Knufia</i>	s__ <i>Knufia petricola</i>
OTU_206	p__Ascomycota	c__Dothideomycetes	Others	Others	Others	Others
OTU_533	p__Ascomycota	c__Sordariomycetes	o__	f__	g__	s__
OTU_495	p__Ascomycota	c__Leotiomycetes	o__Helotiales	f__Sclerotiniaceae	Others	Others
OTU_9	p__Ascomycota	c__Sordariomycetes	o__Glomerellales	f__Plectosphaerellaceae	g__ <i>Plectosphaerella</i>	s__
OTU_66	p__Ascomycota	c__Sordariomycetes	o__Hypocreales	f__unidentified_ Hypocreales	g__ <i>Sarocladium</i>	s__
OTU_538	p__Ascomycota	c__Sordariomycetes	o__Xylariales	f__unidentified_Xylariales	g__unidentified_Xylariales	s__ <i>Bartalinia</i> _sp._SYP-F-7162
OTU_252	p__Ascomycota	c__Dothideomycetes	o__Pleosporales	f__Torulaceae	g__ <i>Torula</i>	s__ <i>Torula herbarum</i>
OTU_307	p__Ascomycota	c__Leotiomycetes	o__Helotiales	f__unidentified_Helotiales	g__ <i>Tetracladium</i>	s__ <i>Tetracladium marchalianum</i>
OTU_364	p__Ascomycota	c__Dothideomycetes	Others	Others	Others	Others
OTU_506	p__Ascomycota	c__Sordariomycetes	o__Hypocreales	f__unidentified_Hypocreales	g__ <i>Acremonium</i>	s__ <i>Hypocreales</i> _sp._GMG_P Pb3
OTU_321	p__Ascomycota	c__Orbiliomycetes	o__Orbiliales	f__Orbiliaceae	g__ <i>Dactylella</i>	s__ <i>Dactylella oxyspora</i>
OTU_146	p__Ascomycota	c__Dothideomycetes	o__Pleosporales	Others	Others	Others
OTU_268	p__Ascomycota	c__Leotiomycetes	Others	Others	Others	Others
OTU_176	p__Ascomycota	c__Sordariomycetes	o__Hypocreales	Others	Others	Others
OTU_405	p__Ascomycota	c__Saccharomycetes	o__Saccharomycetales	f__Saccharomycodaceae	g__ <i>Hanseniaspora</i>	s__
OTU_350	p__Ascomycota	c__Dothideomycetes	o__Pleosporales	f__Pleosporaceae	Others	Others
OTU_567	p__Ascomycota	c__Sordariomycetes	o__Hypocreales	f__Nectriaceae	g__ <i>Fusarium</i>	Others
OTU_2	p__Ascomycota	c__Dothideomycetes	o__Capnodiales	f__Cladosporiaceae	g__ <i>Cladosporium</i>	s__ <i>Cladosporium herbarum</i>
OTU_329	p__Ascomycota	c__Saccharomycetes	o__Saccharomycetales	f__Metschnikowiaceae	g__ <i>Metschnikowia</i>	s__ <i>Metschnikowia reukaufii</i>
OTU_186	p__Ascomycota	c__Dothideomycetes	o__Pleosporales	f__Pleosporaceae	g__ <i>Alternaria</i>	s__ <i>Alternaria</i> _sp._PMK1
OTU_334	p__Ascomycota	c__Eurotiomycetes	o__Chaetothyriales	f__Cyphellophoraceae	g__ <i>Cyphellophora</i>	s__ <i>Cyphellophora laciniata</i>
OTU_110	p__Ascomycota	c__Sordariomycetes	o__Glomerellales	f__Plectosphaerellaceae	g__ <i>Plectosphaerella</i>	s__
OTU_424	p__Ascomycota	c__Laboulbeniomycetes	Others	Others	Others	Others
OTU_99	p__Ascomycota	c__Dothideomycetes	o__Pleosporales	f__Arthopyreniaceae	g__ <i>Arthopyrenia</i>	s__ <i>Arthopyreniaceae</i> _sp._GM G_P1
OTU_205	p__Ascomycota	c__Sordariomycetes	o__Sordariales	f__Chaetomiaceae	g__ <i>Chaetomium</i>	s__ <i>Podospora</i> _sp._7GJ-4

Supplementary Table 4 Continued.

OTU_ID	Phylum	Class	Order	Family	Genus	Species
OTU_582	p__Ascomycota	c__Dothideomycetes	o__Capnodiales	f__Dissoconiaceae	g__ <i>Dissoconium</i>	s__ <i>Dissoconium aciculare</i>
OTU_273	p__Ascomycota	c__Sordariomycetes	Others	Others	Others	Others
OTU_175	p__Ascomycota	c__Sordariomycetes	o__Hypocreales	f__unidentified_Hypocreales	g__ <i>Acremonium</i>	s__ <i>Acremonium curvulum</i>
OTU_163	p__Ascomycota	c__Eurotiomycetes	o__Eurotiales	f__Aspergillaceae	g__ <i>Aspergillus</i>	s__ <i>Aspergillus lentulus</i>
OTU_204	p__Ascomycota	c__Sordariomycetes	o__Hypocreales	Others	Others	Others
OTU_275	p__Ascomycota	c__Sordariomycetes	o__unidentified_Sordariomycetes	f__unidentified_Sordariomycetes	g__ <i>Infundibulomyces</i>	s__ <i>Infundibulomyces</i> _sp._NR-2006a
OTU_363	p__Ascomycota	c__Leotiomycetes	o__Erysiphales	f__Erysiphaceae	g__ <i>Erysiphe</i>	s__ <i>Erysiphe pisi</i>
OTU_385	p__Ascomycota	c__Sordariomycetes	o__Hypocreales	Others	Others	Others
OTU_6	p__Ascomycota	c__Dothideomycetes	o__Pleosporales	f__Didymellaceae	g__ <i>Boeremia</i>	s__
OTU_501	p__Ascomycota	c__Sordariomycetes	o__unidentified_Sordariomycetes	f__unidentified_Sordariomycetes	Others	Others
OTU_522	p__Ascomycota	c__Sordariomycetes	o__Glomerellales	f__Glomerellaceae	g__ <i>Colletotrichum</i>	Others
OTU_348	p__Ascomycota	Others	Others	Others	Others	Others
OTU_75	p__Ascomycota	c__Dothideomycetes	o__Pleosporales	f__unidentified_Pleosporales	g__unidentified_Pleosporales	s__fungal_sp.
OTU_220	p__Ascomycota	c__Sordariomycetes	o__Xylariales	f__Xylariaceae	Others	Others
OTU_338	p__Ascomycota	c__Dothideomycetes	o__Pleosporales	f__Pleosporaceae	g__ <i>Alternaria</i>	Others
OTU_30	p__Basidiomycota	c__Exobasidiomycetes	o__Entylomatales	f__unidentified_Entylomatales	g__ <i>Tilletiopsis</i>	s__ <i>Golubevia pallescens</i>
OTU_458	p__Basidiomycota	c__Tremellomycetes	o__Cystofilobasidiales	f__Cystofilobasidiaceae	g__ <i>Cystofilobasidium</i>	Others
OTU_373	p__Basidiomycota	c__Tremellomycetes	o__Cystofilobasidiales	f__Mrakiaceae	g__ <i>Udeniomyces</i>	s__ <i>Udeniomyces megalosporus</i>
OTU_37	p__Basidiomycota	c__Agaricomycetes	o__Cantharellales	f__Ceratobasidiaceae	Others	Others
OTU_437	p__Basidiomycota	c__Agaricomycetes	o__Sebacinales	f__unidentified_Sebacinales	g__ <i>Chaetospermum</i>	s__ <i>Chaetospermum artocarpi</i>
OTU_570	p__Basidiomycota	c__Tremellomycetes	o__Filobasidiales	f__Filobasidiaceae	Others	Others
OTU_7	p__Basidiomycota	c__Agaricomycetes	o__Cantharellales	f__Ceratobasidiaceae	g__ <i>Rhizoctonia</i>	s__
OTU_189	p__Basidiomycota	c__Exobasidiomycetes	o__Doassansiales	f__Doassansiaceae	g__ <i>Doassansia</i>	s__ <i>Doassansia hygrophilae</i>
OTU_469	p__Basidiomycota	c__Agaricomycetes	o__Corticiales	f__Corticaceae	g__ <i>Limonomyces</i>	s__ <i>Limonomyces roseipellis</i>
OTU_152	p__Basidiomycota	c__Microbotryomycetes	o__Leucosporidiales	f__Leucosporidiaceae	g__ <i>Leucosporidium</i>	Others
OTU_12	p__Basidiomycota	c__Microbotryomycetes	o__Sporidiobolales	f__Sporidiobolaceae	g__ <i>Sporidiobolus</i>	s__ <i>Sporidiobolus pararoseus</i>
OTU_132	p__Basidiomycota	c__Microbotryomycetes	o__Sporidiobolales	f__Sporidiobolaceae	g__ <i>Rhodotorula</i>	s__ <i>Rhodotorula glutinis</i>
OTU_429	p__Basidiomycota	c__Cystobasidiomycetes	o__unidentified_Cystobasidiomycetes	f__unidentified_Cystobasidiomycetes	g__ <i>Symmetrospora</i>	s__ <i>Symmetrospora symmetrica</i>
OTU_335	p__Basidiomycota	c__Agaricomycetes	Others	Others	Others	Others
OTU_106	p__Basidiomycota	c__Tremellomycetes	o__Cystofilobasidiales	f__Mrakiaceae	g__ <i>Udeniomyces</i>	s__ <i>Udeniomyces pyricola</i>
OTU_239	p__Basidiomycota	c__Agaricomycetes	o__Cantharellales	f__Ceratobasidiaceae	g__ <i>Rhizoctonia</i>	s__ <i>Rhizoctonia solani</i>

Supplementary Table 4 Continued.

OTU_ID	Phylum	Class	Order	Family	Genus	Species
OTU_375	p__Basidiomycota	c__Agaricomycetes	o__Cantharellales	f__Ceratobasidiaceae	g__ <i>Rhizoctonia</i>	Others
OTU_249	p__Basidiomycota	c__Agaricomycetes	Others	Others	Others	Others
OTU_16	p__Basidiomycota	c__Tremellomycetes	o__Holtermanniales	f__unidentified_ Holtermanniales	g__ <i>Holtermanniella</i>	s__ <i>Holtermanniella</i> <i>takashimae</i>
OTU_124	p__Basidiomycota	c__Tremellomycetes	o__Tremellales	f__Bulleribasidiaceae	g__ <i>Vishniacozyma</i>	s__
OTU_591	p__Basidiomycota	c__Agaricomycetes	o__Atheliales	f__Atheliaceae	g__ <i>Athelia</i>	s__ <i>Athelia rolfsii</i>
OTU_36	p__Basidiomycota	c__Tremellomycetes	o__Tremellales	f__Bulleribasidiaceae	g__ <i>Hannaella</i>	Others
OTU_512	p__Basidiomycota	c__Agaricomycetes	o__Corticiales	f__Corticiaceae	g__ <i>Sistotrema</i>	Others
OTU_298	p__Basidiomycota	c__Tremellomycetes	o__Filobasidiales	f__Filobasidiaceae	g__ <i>Filobasidium</i>	s__
OTU_374	p__Basidiomycota	c__Malasseziomycetes	o__Malasseziales	f__Malasseziaceae	g__ <i>Malassezia</i>	s__
OTU_121	p__Basidiomycota	c__Agaricomycetes	o__Agaricales	f__Strophariaceae	g__ <i>Pachylepyrium</i>	s__ <i>Pachylepyrium</i> <i>carbonicola</i>
OTU_380	p__Basidiomycota	c__Tremellomycetes	o__Cystofilobasidiales	f__Mrakiaceae	g__ <i>Tausonia</i>	s__ <i>Tausonia pullulans</i>
OTU_8	p__Basidiomycota	c__Ustilaginomycetes	o__Ustilaginales	f__Ustilaginaceae	g__ <i>Sporisorium</i>	s__
OTU_588	p__Basidiomycota	c__Agaricomycetes	o__Agaricales	f__Tricholomataceae	Others	Others
OTU_18	p__Basidiomycota	c__Tremellomycetes	o__Cystofilobasidiales	f__Mrakiaceae	g__ <i>Udeniomyces</i>	s__ <i>Udeniomyces</i> <i>megalosporus</i>
OTU_408	p__Basidiomycota	c__Tremellomycetes	o__Tremellales	f__Rhynchogastremataceae	g__ <i>Papiliotrema</i>	s__ <i>Papiliotrema flavescens</i>
OTU_427	p__Basidiomycota	c__Cystobasidiomycetes	o__Cystobasidiales	f__unidentified_ Cystobasidiales	g__ <i>Occultifur</i>	s__ <i>Occultifur externus</i>
OTU_162	p__Basidiomycota	c__Tremellomycetes	o__Tremellales	Others	Others	Others
OTU_493	p__Basidiomycota	c__Tremellomycetes	o__Filobasidiales	f__Filobasidiaceae	g__ <i>Filobasidium</i>	s__
OTU_55	p__Basidiomycota	c__Tremellomycetes	o__Cystofilobasidiales	f__Mrakiaceae	g__ <i>Itersonilia</i>	s__
OTU_242	p__Basidiomycota	c__Agaricomycetes	o__Agaricales	Others	Others	Others
OTU_64	p__Basidiomycota	c__Agaricomycetes	o__Corticiales	f__Corticiaceae	g__ <i>Limonomyces</i>	s__ <i>Limonomyces roseipellis</i>
OTU_43	p__Basidiomycota	c__Cystobasidiomycetes	o__unidentified_ Cystobasidiomycetes	f__unidentified_ Cystobasidiomycetes	g__ <i>Symmetrospora</i>	s__ <i>Symmetrospora coprosmae</i>
OTU_502	p__Basidiomycota	c__Tremellomycetes	o__Filobasidiales	f__Filobasidiaceae	g__ <i>Naganishia</i>	s__ <i>Naganishia vishniacii</i>
OTU_562	p__Basidiomycota	c__Agaricomycetes	o__Agaricales	Others	Others	Others
OTU_266	p__Basidiomycota	c__Agaricomycetes	o__Cantharellales	f__Ceratobasidiaceae	g__ <i>Rhizoctonia</i>	s__ <i>Rhizoctonia solani</i>
OTU_33	p__Basidiomycota	c__Tremellomycetes	o__Cystofilobasidiales	f__Cystofilobasidiaceae	g__ <i>Cystofilobasidium</i>	s__
OTU_586	p__Basidiomycota	c__Agaricomycetes	o__Cantharellales	f__Ceratobasidiaceae	Others	Others
OTU_443	p__Basidiomycota	c__Cystobasidiomycetes	o__unidentified_ Cystobasidiomycetes	f__unidentified_ Cystobasidiomycetes	g__ <i>Buckleyzyma</i>	s__ <i>Buckleyzyma aurantiaca</i>
OTU_294	p__Basidiomycota	c__Tremellomycetes	o__Filobasidiales	f__Filobasidiaceae	g__ <i>Filobasidium</i>	s__
OTU_398	p__Basidiomycota	c__Tremellomycetes	o__Filobasidiales	f__Filobasidiaceae	g__ <i>Filobasidium</i>	s__
OTU_149	p__Basidiomycota	c__Tremellomycetes	o__Filobasidiales	f__Filobasidiaceae	g__ <i>Filobasidium</i>	s__

Supplementary Table 4 Continued.

OTU_ID	Phylum	Class	Order	Family	Genus	Species
OTU_35	p__Basidiomycota	c__Exobasidiomycetes	o__Entylomatales	f__unidentified_ Entylomatales	g__ <i>Tilletiopsis</i>	Others
OTU_228	p__Basidiomycota	c__Agaricostilbomycetes	o__Agaricostilbales	f__Kondoaceae	g__ <i>Kondoa</i>	s__ <i>Kondoa sorbi</i>
OTU_24	p__Basidiomycota	c__Tremellomycetes	o__Tremellales	f__Rhynchogastremataceae	g__ <i>Papiliotrema</i>	s__ <i>Papiliotrema flavescens</i>
OTU_46	p__Basidiomycota	Others	Others	Others	Others	Others
OTU_518	p__Basidiomycota	c__Agaricomycetes	o__Cantharellales	f__Ceratobasidiaceae	Others	Others
OTU_183	p__Basidiomycota	c__Cystobasidiomycetes	o__Erythrobasidiales	f__Erythrobasidiaceae	g__ <i>Erythrobasidium</i>	Others
OTU_402	p__Basidiomycota	c__Agaricomycetes	o__Agaricales	Others	Others	Others
OTU_4	p__Basidiomycota	c__Tremellomycetes	o__Filobasidiales	f__Filobasidiaceae	g__ <i>Filobasidium</i>	s__
OTU_245	p__Chytridiomycota	c__Chytridiomycetes	o__Lobulomycetales	f__Lobulomycetaceae	g__	s__
OTU_284	p__Chytridiomycota	c__Chytridiomycetes	o__Chytridiales	f__Chytridiaceae	g__ <i>Chytridium</i>	s__ <i>Chytridium polysiphoniae</i>
OTU_201	p__Chytridiomycota	c__Chytridiomycetes	o__Rhizophydiales	Others	Others	Others
OTU_214	p__Chytridiomycota	c__Chytridiomycetes	Others	Others	Others	Others
OTU_184	p__Chytridiomycota	c__Chytridiomycetes	o__Spizellomycetales	f__Olpidiaceae	g__ <i>Olpidium</i>	s__ <i>Olpidium brassicae</i>
OTU_494	p__Chytridiomycota	c__Chytridiomycetes	Others	Others	Others	Others
OTU_91	p__Chytridiomycota	c__Chytridiomycetes	o__	f__	g__	s__
OTU_470	p__Chytridiomycota	c__Chytridiomycetes	Others	Others	Others	Others
OTU_304	p__Chytridiomycota	c__Chytridiomycetes	o__Rhizophydiales	f__	g__	s__
OTU_330	p__Chytridiomycota	c__Chytridiomycetes	o__Chytridiales	f__Chytridiaceae	g__	s__
OTU_118	p__Chytridiomycota	c__Chytridiomycetes	o__Lobulomycetales	f__Lobulomycetaceae	Others	Others
OTU_129	p__Cryptomycota	c__unidentified_ Cryptomycota	o__unidentified_ Cryptomycota	f__unidentified_ Cryptomycota	g__ <i>Rozella</i>	s__
OTU_561	p__Cryptomycota	c__	o__	f__	g__	s__
OTU_305	p__Cryptomycota	c__	o__	f__	g__	s__
OTU_378	p__Cryptomycota	c__	o__	f__	g__	s__
OTU_86	p__Cryptomycota	c__unidentified_Crypto mycota	o__unidentified_ Cryptomycota	f__unidentified_ Cryptomycota	g__ <i>Rozella</i>	s__
OTU_477	p__Cryptomycota	c__	o__	f__	g__	s__
OTU_112	p__Cryptomycota	c__unidentified_Crypto mycota	o__unidentified_ Cryptomycota	f__unidentified_ Cryptomycota	g__ <i>Rozella</i>	s__
OTU_387	p__Cryptomycota	c__	o__	f__	g__	s__
OTU_287	p__Mucoromycota	c__Glomeromycetes	o__Glomerales	f__Claroideoglomeraceae	g__ <i>Claroideoglomerus</i>	s__ <i>Claroideoglomerus etunicatum</i>
OTU_87	p__Mucoromycota	c__unidentified_ Mucoromycota	o__Mucorales	f__Mucoraceae	g__ <i>Mucor</i>	s__ <i>Mucor amphibiorum</i>
OTU_333	p__Mucoromycota	c__unidentified_ Mucoromycota	o__Mucorales	f__Mucoraceae	g__ <i>Mucor</i>	s__

Supplementary Table 4 Continued.

OTU_ID	Phylum	Class	Order	Family	Genus	Species
OTU_240	p__Mucoromycota	c__unidentified_ Mucoromycota	o__Mucorales	f__Cunninghamellaceae	g__ <i>Gongronella</i>	s__ <i>Gongronella</i> _sp._w5
OTU_145	p__Mucoromycota	c__unidentified_ Mucoromycota	o__Mortierellales	Others	Others	Others
OTU_156	p__Mucoromycota	c__unidentified_ Mucoromycota	o__Mucorales	f__Cunninghamellaceae	g__ <i>Cunninghamella</i>	s__ <i>Cunninghamella</i> <i>bertholletiae</i>
OTU_516	p__Mucoromycota	c__unidentified_ Mucoromycota	o__Mucorales	f__Mucoraceae	g__ <i>Mucor</i>	s__ <i>Mucor hiemalis</i>
OTU_15	p__Mucoromycota	c__unidentified_ Mucoromycota	o__Mucorales	f__Mucoraceae	g__ <i>Mucor</i>	s__ <i>Mucor hiemalis</i>
OTU_391	p__Mucoromycota	c__Glomeromycetes	o__Diversisporales	f__Acaulosporaceae	g__ <i>Acaulospora</i>	s__ <i>Acaulospora laevis</i>
OTU_537	p__Mucoromycota	c__Glomeromycetes	o__Glomerales	f__Claroideoglomeraceae	g__ <i>Claroideoglomerus</i>	s__ <i>Glomus</i> _sp._NBR_PP1
OTU_274	p__Mucoromycota	c__unidentified_ Mucoromycota	o__Endogonales	f__Endogonaceae	g__unidentified_ Endogonaceae	s__ <i>Mucoromycotina</i> _sp._MIB_ 8846
OTU_447	p__Mucoromycota	c__Glomeromycetes	o__Glomerales	f__Glomeraceae	g__ <i>Funneliformis</i>	s__ <i>Glomeromycotina</i> _sp._WR8 56-B
OTU_361	p__Mucoromycota	c__unidentified_ Mucoromycota	o__Mucorales	f__Mucoraceae	g__ <i>Mucor</i>	s__
OTU_32	p__Mucoromycota	c__unidentified_ Mucoromycota	o__Mucorales	f__Mucoraceae	g__ <i>Mucor</i>	s__
OTU_317	p__Mucoromycota	c__unidentified_ Mucoromycota	o__Mucorales	f__Rhizopodaceae	g__ <i>Rhizopus</i>	s__ <i>Rhizopus oryzae</i>
OTU_508	p__Mucoromycota	c__unidentified_ Mucoromycota	o__Mucorales	f__Mucoraceae	g__ <i>Mucor</i>	s__ <i>Mucor mucedo</i>
OTU_107	p__Mucoromycota	c__unidentified_ Mucoromycota	o__Mucorales	f__Mucoraceae	g__ <i>Mucor</i>	Others
OTU_448	p__Mucoromycota	c__unidentified_ Mucoromycota	o__Endogonales	f__Endogonaceae	g__ <i>Endogone</i>	s__ <i>Mucoromycotina</i> _sp._MIB_ 8447
OTU_221	p__Mucoromycota	c__unidentified_ Mucoromycota	o__Endogonales	f__Endogonaceae	g__unidentified_ Endogonaceae	Others

Supplementary Table 5 The Alpha diversity indexes are based on fungal sequences. Data are shown as mean \pm sd.

Group	sample	observed_species	Shannon	Simpson	chao1	ace	goods_coverage	PD_whole_tree
AS	–	38.78 \pm 7.29	3.63 \pm 0.49	0.85 \pm 0.07	42.07 \pm 8.22	42.77 \pm 8.61	0.99 \pm 0.00	2.24 \pm 0.45
VV	–	42.92 \pm 7.10	3.77 \pm 0.48	0.86 \pm 0.06	47.37 \pm 9.23	48.35 \pm 9.97	0.99 \pm 0.00	2.45 \pm 0.52
Cen.AS	–	43.83 \pm 7.28	3.69 \pm 0.49	0.85 \pm 0.06	48.45 \pm 8.26	49.15 \pm 9.12	0.99 \pm 0.01	2.60 \pm 0.35
Cen.VV	–	47.17 \pm 6.65	4.01 \pm 0.38	0.89 \pm 0.04	53.85 \pm 7.37	55.18 \pm 8.61	0.99 \pm 0.00	2.64 \pm 0.61
Sou.AS	–	36.25 \pm 6.09	3.60 \pm 0.51	0.85 \pm 0.07	38.88 \pm 6.33	39.57 \pm 6.59	0.99 \pm 0.00	2.07 \pm 0.39
Sou.VV	–	38.67 \pm 4.84	3.54 \pm 0.49	0.84 \pm 0.06	40.89 \pm 5.70	41.53 \pm 5.74	0.99 \pm 0.00	2.26 \pm 0.35
Cen.AS	HNLASP.1	44	4.00	0.90	46.14	46.21	0.99	2.59
	HNLASR.1	43	3.97	0.88	43.20	43.60	1.00	2.39
	HNWASF.2	52	3.94	0.88	62.11	64.54	0.98	3.17
	HNWASL.1	52	4.04	0.89	54.33	55.92	0.99	2.86
	HNWASL.2	38	3.41	0.83	39.67	41.61	0.99	2.25
	HNWASL.3	34	2.81	0.74	45.25	43.04	0.99	2.36
Cen.VV	HNWVVF.1	48	4.12	0.90	52.50	56.32	0.99	2.60
	HNWVVF.2	44	3.96	0.90	53.17	51.58	0.98	2.44
	HNWVVL.1	43	3.44	0.80	50.50	51.00	0.99	2.72
	HNWVVL.2	42	3.80	0.88	45.50	45.35	0.99	2.55
	HNWVVR.2	60	4.53	0.93	67.58	70.73	0.98	3.71
	HNWVVS.1	46	4.24	0.92	53.86	56.08	0.98	1.83
Sou.AS	FJASL.1	32	2.83	0.77	41.00	41.07	0.99	1.48
	FJASP.1	32	3.40	0.84	35.75	35.82	0.99	2.50
	FJASP.2	30	3.38	0.85	31.00	31.67	0.99	1.89
	FJASP.3	26	3.13	0.80	26.33	26.68	1.00	1.28
	FJASS.1	34	3.67	0.87	37.33	36.67	0.99	2.25
	FJASS.3	39	4.12	0.91	39.86	41.20	0.99	1.80
	GXGASL.1	32	2.82	0.69	33.67	35.45	0.99	2.11
	GXGASL.3	44	3.68	0.85	46.55	49.42	0.99	2.16
	GXGASP.3	43	4.03	0.90	43.75	44.86	0.99	2.33
	GXGASS.1	39	3.66	0.86	44.60	43.53	0.99	2.13
	GXGASS.2	39	4.01	0.90	40.00	41.09	0.99	2.29
	GXGASS.3	45	4.47	0.94	46.67	47.39	0.99	2.57
Sou.VV	GXGVVL.1	38	3.51	0.84	41.00	41.22	0.99	2.23
	GXGVVL.2	40	3.47	0.84	42.00	41.79	0.99	1.98
	GXGVVP.1	35	3.12	0.78	36.67	38.90	0.99	2.26
	GXGVVR.2	47	4.39	0.92	51.00	52.03	0.99	2.92
	GXNVVS.1	33	3.02	0.77	34.50	34.73	0.99	2.17
	GXGVVS.2	39	3.70	0.88	40.20	40.51	0.99	1.97

Supplementary Table 6 The Alpha diversity indexes are based on all the 18S rRNA reads. Data are shown as mean \pm sd.

Group	sample	observed_species	Shannon	Simpson	chao1	ace	goods_coverage	PD_whole_tree
AS	-	42 \pm 8.07	3.63 \pm 0.50	0.85 \pm 0.07	53.59 \pm 16.73	53.12 \pm 12.88	0.98 \pm 0.00	2.34 \pm 0.55
VV	-	42.92 \pm 7.72	3.82 \pm 0.49	0.86 \pm 0.06	63.67 \pm 17.24	66.74 \pm 15.29	0.98 \pm 0.00	2.79 \pm 0.53
Cen.AS	-	47.67 \pm 9.81	3.70 \pm 0.49	0.85 \pm 0.06	59.01 \pm 19.43	62.40 \pm 19.60	0.98 \pm 0.01	2.71 \pm 0.64
Cen.VV	-	55.67 \pm 5.43	4.07 \pm 0.37	0.89 \pm 0.04	74.44 \pm 18.42	77.88 \pm 12.58	0.97 \pm 0.01	3.11 \pm 0.52
Sou.AS	-	39.17 \pm 5.52	3.60 \pm 0.52	0.85 \pm 0.07	50.87 \pm 15.40	48.47 \pm 4.53	0.99 \pm 0.00	2.16 \pm 0.40
Sou.VV	-	44.17 \pm 4.71	3.57 \pm 0.50	0.84 \pm 0.06	52.90 \pm 6.02	55.60 \pm 7.62	0.98 \pm 0.00	2.47 \pm 0.32
Cen.AS	HNLASP.1	44	3.95	0.90	47.60	49.63	0.99	2.38
	HNLASR.1	48	4.01	0.88	49.88	50.92	0.99	2.91
	HNWASL.1	64	4.08	0.89	87.00	95.38	0.97	3.75
	HNWASF.2	52	3.91	0.88	80.50	75.91	0.97	2.88
	HNWASL.2	43	3.44	0.83	46.75	49.95	0.99	2.49
	HNWASL.3	35	2.81	0.73	42.33	52.61	0.98	1.86
Cen.VV	HNWVCF.1	61	4.21	0.90	103.86	96.79	0.96	3.60
	HNWVCS.1	52	4.30	0.92	60.27	66.96	0.98	2.33
	HNWVCL.1	49	3.48	0.80	57.08	66.94	0.98	2.73
	HNWVCF.2	56	4.05	0.90	87.63	87.62	0.97	3.16
	HNWVCL.2	53	3.85	0.88	62.07	68.28	0.98	3.11
	HNWVCR.2	63	4.52	0.92	75.75	80.68	0.97	3.72
Sou.AS	FJASS.1	41	3.71	0.87	46.00	51.90	0.99	1.85
	FJASL.1	32	2.80	0.77	45.20	48.11	0.98	1.45
	FJASP.1	34	3.36	0.84	43.00	45.51	0.99	2.48
	FJASP.2	33	3.37	0.85	99.00	47.47	0.98	2.09
	FJASS.3	41	4.09	0.91	48.20	49.21	0.99	2.47
	FJASP.3	31	3.15	0.80	40.33	37.19	0.99	1.42
	GXGASS.1	38	3.63	0.86	49.25	46.08	0.99	2.09
	GXGASL.1	40	2.87	0.69	48.67	55.82	0.98	2.40
	GXGASS.2	45	4.03	0.90	47.33	51.42	0.99	2.47
	GXGASS.3	47	4.49	0.94	50.00	51.18	0.99	2.66
	GXGASL.3	43	3.66	0.85	47.00	49.50	0.99	2.17
	GXGASP.3	45	4.06	0.90	46.50	48.29	0.99	2.34
Sou.VV	GXGVCL.1	42	3.55	0.84	51.00	49.24	0.99	2.30
	GXGVCP.1	43	3.18	0.78	55.00	66.53	0.98	2.68
	GXGVCS.2	46	3.72	0.88	61.00	61.52	0.98	2.43
	GXGVCL.2	46	3.49	0.84	54.25	54.27	0.98	2.08
	GXGVCR.2	51	4.45	0.92	53.55	56.07	0.99	2.98
	GXNVCS.1	37	3.04	0.77	42.63	45.96	0.99	2.31

Supplementary Table 7 Functional annotation of the genera and species recorded in culture-independent and dependent methods (Based on the literature).

Genus	Species	Life mode or Function (and/or potential pathogenic/beneficial/biocontrol)	References
Culture-independent			
<i>g__Fusarium</i>	-	Endophyte/Epiphytes/Pathogen	Inácio et al. (2002), Leslie et al. (1990), Kuldau & Yates (2000), Bacon & Yates (2006), Imazaki & Kadota (2015), Gonzalez & Tello (2011)
<i>g__Cladosporium</i>	-	Endophyte/Pathogen/Saprotroph	Swett et al. (2016)
<i>g__Magnaporthe</i>	-	Pathogen	Ou (1985), Prabhu et al. (2009)
<i>g__Colletotrichum</i>	<i>s__Colletotrichum_tofieldiae</i>	Endophyte, Beneficial fungus, plant growth promotion	García et al. (2013)
<i>g__Chaetomium</i>	<i>s__Chaetomium_globosum</i>	Endophyte/Saprophytic, growth and mycotoxin, bioactive metabolites, antifungal activity, a biocontrol agent	Reissinger et al. (2003), Shi et al. (2016), Thongkantha et al. (2008)
<i>g__Ochroconis</i>	<i>Others</i>	Human pathogen	Cardeau-Desangles et al. (2013)
<i>g__Saccharomyces</i>	<i>s__</i>	Human pathogen	Yamamoto et al. (2002)
<i>g__Alternaria</i>	<i>s__Alternaria alternata</i>	Endophyte/Pathogen/Saprotroph	Meena et al. (2017)
<i>g__Plectosphaerella</i>	<i>s__</i>	Pathogen	Carlucci et al. (2012)
<i>g__Fusarium</i>	<i>Others</i>	Endophyte/Epiphytes/Pathogen	Inácio et al. (2002), Leslie et al. (1990), Kuldau & Yates (2000), Bacon & Yates 2006, Imazaki & Kadota (2015), Gonzalez & Tello (2011)
<i>g__Protomyces</i>	<i>s__Protomyces_inouyei</i>	Pathogen	Wang et al. (2019)
<i>g__Chaetomium</i>	<i>Others</i>	Endophyte/Saprophytic	Reissinger et al. (2003), Shi et al. (2016), Thongkantha et al. (2008)
<i>g__Magnaporthe</i>	<i>Others</i>	Pathogen	Ou (1985), Prabhu et al. (2009)
<i>g__Pichia</i>	<i>s__Pichia_kudriavzevii</i>	Human pathogen	Kurtzman et al. (1904)
<i>g__Sclerotinia</i>	<i>s__</i>	Pathogen	Abawi & Grogan (1979)
<i>g__Pyxidiophora</i>	<i>s__Pyxidiophora_arvernensis</i>	Saprophytic	Blackwell & Malloch (1989)
<i>g__Plectosphaerella</i>	<i>s__</i>	Pathogen	Carlucci et al. (2012)
<i>g__Eremothecium</i>	<i>Others</i>	Pathogen	Ashby & Nowell (1926)
<i>g__Lectera</i>	<i>s__Lectera_colletotrichoides</i>	Pathogen	Cannon et al. (2012)
<i>g__Diaporthe</i>	<i>s__Diaporthe_amygdali</i>	Pathogen	Meng et al. (2018)
<i>g__Fusarium</i>	<i>Others</i>	Endophyte/Epiphytes/Pathogen	Inácio et al. (2002), Leslie et al. (1990), Kuldau & Yates (2000), Bacon & Yates (2006), Imazaki & Kadota (2015), Gonzalez & Tello (2011)
<i>g__Magnaporthe</i>	<i>s__Magnaporthe_oryzae</i>	Pathogen	Ou (1985), Prabhu et al. (2009)
<i>g__Knufia</i>	<i>s__Knufia_petricola</i>	inhabiting on insects	
<i>g__Plectosphaerella</i>	<i>s__</i>	Pathogen	Carlucci et al. (2012)
<i>g__Sarocladium</i>	<i>s__</i>	Pathogen	Ayyadurai et al. (2005)
<i>g__unidentified_Xylariales</i>	<i>s__Bartalinia_sp._SYP-F-7162</i>	Saprophytic	Nguyen et al. (2019)

Supplementary Table 7 Continued.

Genus	Species	Life mode or Function (and/or potential pathogenic/beneficial/biocontrol)	References
<i>g__Torula</i>	<i>s__Torula_herbarum</i>	Saprophytic	Crous et al. (2020), Tibpromma et al. (2017)
<i>g__Tetracladium</i>	<i>s__Tetracladium_marchalianum</i>	Saprophytic	Anderson & Shearer (2011)
<i>g__Acremonium</i>	<i>s__Hypocreales_sp._GMG_PPb3</i>	saprophytic, opportunistic pathogens	Glenn et al. (1996)
<i>g__Dactylella</i>	<i>s__Dactylella_oxyspora</i>	saprotrophic, oospore or nematode-egg parasite	Chen et al. (2007)
<i>g__Hanseniaspora</i>	<i>s__</i>	Yeast	Albertin et al. (2016)
<i>g__Fusarium</i>	Others	Endophyte/Epiphytes/Pathogen	Inácio et al. (2002), Leslie et al. (1990), Kulda & Yates (2000), Bacon & Yates (2006), Imazaki & Kadota (2015), Gonzalez & Tello (2011)
<i>g__Cladosporium</i>	<i>s__Cladosporium_herbarum</i>	Endophyte/Pathogen/Saprotroph	Swett et al. (2016)
<i>g__Metschnikowia</i>	<i>s__Metschnikowia_reukaufii</i>	Natural contaminant	Carlos (2014)
<i>g__Alternaria</i>	<i>s__Alternaria_sp._PMK1</i>	Endophyte/Pathogen/Saprotroph	Meena et al. (2017)
<i>g__Cyphellophora</i>	<i>s__Cyphellophora_laciniata</i>	Human pathogen/ Endophyte/plant pathogen/Saprotroph	Feng et al. (2012), Decock et al (2003), de Hoog (1999), Gams & Holubová-Jechová (1976), Grabowski (2007)
<i>g__Plectosphaerella</i>	<i>s__</i>	Pathogen	Carlucci et al. (2012)
<i>g__Arthopyrenia</i>	<i>s__Arthopyreniaceae_sp._GMG_PI</i>	corticolous lichenized or non-lichenized fungi	Coppins (1988)
<i>g__Chaetomium</i>	<i>s__Podospora_sp._7GJ-4</i>	Coprophilous fungi	Hu et al. (2006)
<i>g__Dissoconium</i>	<i>s__Dissoconium_aciculare</i>	hyper parasitic fungi	Crous et al. (2007), Li et al. (2012)
<i>g__Acremonium</i>	<i>s__Acremonium_curvulum</i>	saprophytic/opportunistic pathogens	Kiwan (2019)
<i>g__Aspergillus</i>	<i>s__Aspergillus_lentulus</i>	saprophytes/ human pathogen	Thom and Church (1926)
<i>g__Infundibulomyces</i>	<i>s__Infundibulomyces_sp._NR-2006a</i>	saprophytes	Paingam et al. (2003)
<i>g__Erysiphe</i>	<i>s__Erysiphe_pisi</i>	plant pathogens	Abasova et al. (2018)
<i>g__Boeremia</i>	<i>s__</i>	plant pathogens	Chen et al. (2015)
<i>g__Colletotrichum</i>	Others	Endophyte/Pathogen/Saprotroph	Jayawardena et al. (2016)
<i>g__Alternaria</i>	Others	Endophyte/Pathogen/Saprotroph	Meena et al. (2017)
<i>g__Tilletiopsis</i>	<i>s__Golubevia_pallescens</i>	plant pathogens (Sumts)	Wang et al. (2015)
<i>g__Cystofilobasidium</i>	Others	yeast	Sampaio et al. (2001)
<i>g__Udeniomyces</i>	<i>s__Udeniomyces_megalosporus</i>	yeast	Nakase & Takematsu (1992)
<i>g__Chaetospermum</i>	<i>s__Chaetospermum_artocarp</i>	saprophyte	Tangthirasunun et al. (2014)
<i>g__Rhizoctonia</i>	<i>s__</i>	saprotrophic, facultative plant pathogens, endomycorrhizal	Wu et al. (2010)
<i>g__Doassansia</i>	<i>s__Doassansia_hygrophilae</i>	pathogen (Sumt)	Thirumalachar (1946)
<i>g__Limonomyces</i>	<i>s__Limonomyces_roseipellis</i>	pathogen	Zhang et al. (2013)
<i>g__Leucosporidium</i>	Others	yeast	Watson et al. (1976)
<i>g__Sporidiobolus</i>	<i>s__Sporidiobolus_pararoseus</i>	yeast	Michael et al. (2009)
<i>g__Rhodotorula</i>	<i>s__Rhodotorula_glutinis</i>	yeast/post-harvest pathogen	Zhang et al. (2009)

Supplementary Table 7 Continued.

Genus	Species	Life mode or Function (and/or potential pathogenic/beneficial/biocontrol)	References
<i>g__Symmetrospora</i>	<i>s__Symmetrospora_symmetrica</i>	yeast	Wang et al. (2015)
<i>g__Udeniomyces</i>	<i>s__Udeniomyces_pyricola</i>	yeast	Nakase & Takematsu (1992)
<i>g__Rhizoctonia</i>	<i>s__Rhizoctonia_solani</i>	saprotrophic, facultative plant pathogens, endomycorrhizal	Wu et al. (2010)
<i>g__Rhizoctonia</i>	Others	saprotrophic, facultative plant pathogens, endomycorrhizal	Wu et al. (2010)
<i>g__Holtermanniella</i>	<i>s__Holtermanniella_takashimae</i>	yeast	Wuczkowski et al. (2011)
<i>g__Vishniacozyma</i>	<i>s__</i>	psychrophilic basidiomycetous yeast	Tsuji et al. (2019)
<i>g__Athelia</i>	<i>s__Athelia_rolfsii</i>	facultative parasites of plants and lichens	Esslinger (2009)
<i>g__Hannaella</i>	Others	basidiomycetous yeast	Surussawadee et al. (2015)
<i>g__Sistotrema</i>	Others	Basidiomycota fungi	Kirk et al. (2008)
<i>g__Filobasidium</i>	<i>s__</i>	Yeast	Fell et al. (2000)
<i>g__Malassezia</i>	<i>s__</i>	inhabiting on the skin of humans and animals	Yuping (2016)
<i>g__Pachylepyrium</i>	<i>s__Pachylepyrium_carbonicola</i>	Basidiomycota fungi	Singer (1957)
<i>g__Tausonia</i>	<i>s__Tausonia_pullulans</i>	Yeast	Sampaio (2011)
<i>g__Sporisorium</i>	<i>s__</i>	plant pathogen	Maya et al. (2020)
<i>g__Udeniomyces</i>	<i>s__Udeniomyces_megalosporus</i>	yeast	Nakase & Takematsu (1992)
<i>g__Papiliotrema</i>	<i>s__Papiliotrema_flavescens</i>	Yeast	Into et al. (2018)
<i>g__Occultifur</i>	<i>s__Occultifur_externus</i>	Yeast	Šibanc et al. (2018)
<i>g__Filobasidium</i>	<i>s__</i>	Yeast	Fell et al. (2000)
<i>g__Itersonilia</i>	<i>s__</i>	pathogen	Palacioğlu et al. (2019)
<i>g__Limonomyces</i>	<i>s__Limonomyces_roseipellis</i>	pathogen	Zhang et al. (2014)
<i>g__Symmetrospora</i>	<i>s__Symmetrospora_coprosmiae</i>	Yeast	Wang et al. (2015)
<i>g__Naganishia</i>	<i>s__Naganishia_vishniacii</i>	psychrophilic yeast	Rossi et al. (2009)
	<i>s__Rhizoctonia_solani</i>	saprotrophic, facultative plant pathogens, endomycorrhizal	Wu et al. (2010)
<i>g__Cystofilobasidium</i>	<i>s__</i>	yeast	Sampaio et al. (2001)
<i>g__Buckleyzyma</i>	<i>s__Buckleyzyma_aurantiaca</i>	Yeast	Wang et al. (2015)
<i>g__Filobasidium</i>	<i>s__</i>	Yeast	Fell et al. (2000)
<i>g__Filobasidium</i>	<i>s__</i>	Yeast	Fell et al. (2000)
<i>g__Filobasidium</i>	<i>s__</i>	Yeast	Fell et al. (2000)
<i>g__Tilletiopsis</i>	Others	saprotrophic yeast-like	Richter et al. (2019)
<i>g__Kondoa</i>	<i>s__Kondoa_sorbi</i>	Yeast	Wang et al. (2015)
<i>g__Papiliotrema</i>	<i>s__Papiliotrema_flavescens</i>	Yeast	Into et al. (2018)
<i>g__Erythrobasidium</i>	Others	Yeast	Aime (2006), Hamamoto (2011), Yamada & Komagata (1983)
<i>g__Filobasidium</i>	<i>s__</i>	Yeast	Fell et al. (2000)

Supplementary Table 7 Continued.

Genus	Species	Life mode or Function (and/or potential pathogenic/beneficial/biocontrol)	References
<i>g__Chytridium</i>	<i>s__Chytridium_polysiphoniae</i>	potential algal parasite/pathogen	Raghukumar (1985)
	<i>s__Olpidium_brassicae</i>	Plant-pathogen/fungal obligate parasite	Tewari & Bains (2010)
<i>g__Rozella</i>	<i>s__</i>	endoparasites	Lara et al. (2010), Letcher et al. (2017), Cornu (1872)
<i>g__Rozella</i>	<i>s__</i>	endoparasites	Lara et al. (2010), Letcher et al. (2017), Cornu (1872)
<i>g__Rozella</i>	<i>s__</i>	endoparasites	Lara et al. (2010), Letcher et al. (2017), Cornu (1872)
<i>g__Claroideoglomus</i>	<i>s__Claroideoglomus_etunicatum</i>	arbuscular mycorrhizal fungi	Błaszowski et al. (2015)
<i>g__Mucor</i>	<i>s__Mucor_amphibiorum</i>	saprotrophs	Lebreton et al. (2020)
<i>g__Mucor</i>	<i>s__</i>	saprotrophs	Lebreton et al. (2020)
<i>g__Gongronella</i>	<i>s__Gongronella_sp._w5</i>	inhabit in soil	Zhang et al. (2019)
<i>g__Cunninghamella</i>	<i>s__Cunninghamella_bertholletiae</i>	saprotroph/ opportunistic human pathogen	Reiss et al. (2011), Chung et al. (1992)
<i>g__Mucor</i>	<i>s__Mucor_hiemalis</i>	saprotrophs	Lebreton et al. (2020)
<i>g__Mucor</i>	<i>s__Mucor_hiemalis</i>	saprotrophs	Lebreton et al. (2020)
<i>g__Acaulospora</i>	<i>s__Acaulospora_laevis</i>	arbuscular mycorrhizal fungi	Abdelmoneim et al. (2014)
<i>g__Claroideoglomus</i>	<i>s__Glomus_sp._NBR_PP1</i>	arbuscular mycorrhizal fungi	Błaszowski et al. (2015)
<i>g__unidentified_Endogonaceae</i>	<i>s__Mucoromycotina_sp._MIB_8846</i>	symbioses	Chang et al. (2019)
<i>g__Funneliformis</i>	<i>s__Glomeromycotina_sp._WR856-B</i>	arbuscular mycorrhizal fungi	Schüßler & Walker (2010)
<i>g__Mucor</i>	<i>s__</i>	saprotrophs	Lebreton et al. (2020)
<i>g__Mucor</i>	<i>s__</i>	saprotrophs	Lebreton et al. (2020)
<i>g__Rhizopus</i>	<i>s__Rhizopus_oryzae</i>	post-harvest pathogen	Kwon et al. (2012)
<i>g__Mucor</i>	<i>s__Mucor_mucedo</i>	saprotrophs	Lebreton et al. (2020)
<i>g__Mucor</i>	Others	saprotrophs	Lebreton et al. (2020)
<i>g__Endogone</i>	<i>s__Mucoromycotina_sp._MIB_8447</i>	symbioses	Chang et al. (2019)
<i>g__unidentified_Endogonaceae</i>	Others	symbioses	Chang et al. (2019)
Culture-dependent			
<i>Lasiodiplodia</i>	<i>Lasiodiplodia_mediterranea</i>	Pathogenic, Endophytic, Saprobic	Linaldeddu et al. (2015), Reis et al. (2022), Wiseman et al. (2022), Berraf-Tebbal et al. (2020)
<i>Neofusicoccum</i>	<i>Neofusicoccum_parvum</i>	Pathogenic, Saprobic, Endophytic	Mohammadi et al. (2013), Baskarathevan et al. (2012), Carlucci et al. (2013), Golzar & Burgess (2011), Iturrutxa et al. (2011), Thomidis et al. (2011)
<i>Epicoccum</i>	<i>Epicoccum_astragali</i>	Endophytic	This study

Supplementary Table 7 Continued.

Genus	Species	Life mode or Function (and/or potential pathogenic/beneficial/biocontrol)	References
	<i>Epicoccum henanense</i>	Endophytic	This study
	<i>Epicoccum layuense</i>	potential biological control agent, pathogenic, Endophytic	Del Frari et al. (2019), Chen et al. (2020), Chen et al. (2020), Sanhueza et al. (2022), Del Frari (2022)
	<i>Epicoccum latusicollum</i>	Pathogenic, Endophytic, beneficial	
	<i>Epicoccum rosae</i>	Endophytic	This study
	<i>Epicoccum viciae-villosae</i>	Endophytic	This study
<i>Leptosphaerulina</i>	<i>Leptosphaerulina americana</i>	Pathogenic, Endophytic	Irwin et al. (1985), Zhang and Li (2022), Abler (2003), Liang et al. (2021)
<i>Pseudopithomyces</i>	<i>Pseudopithomyces chartarum</i>	Pathogenic, Endophytic	Perelló et al. (2017)
<i>Alternaria</i>	<i>Alternaria alternata</i>	Pathogenic, Endophytic, Saprobic	
	<i>Alternaria astragalicola</i>	Endophytic	This study
	<i>Alternaria gaisen</i>	Pathogenic, Endophytic	Akhtar et al. (2014), Perveen et al. (2018), Tian et al. (2020),
	<i>Alternaria guizhouensis</i>	Endophytic	This study
	<i>Alternaria henanensis</i>	Endophytic	This study
<i>Stemphylium</i>	<i>Stemphylium astragali</i>	Pathogenic, Endophytic	Brahamanage et al. (2018), Uchino et al. (1986)
<i>Botrytis</i>	<i>Botrytis cinerea</i>	Pathogenic, Endophytic	Williamson et al. (2007)
<i>Sclerotinia</i>	<i>Sclerotinia minor</i>	Pathogenic, Endophytic	Melzer et al. (1997), Hao et al. (2003)
	<i>Sclerotinia sclerotiorum</i>	Pathogenic, Endophytic	Hao et al. (2003), Hegedus and Rimmer (2005)
<i>Arthrinium</i>	<i>Arthrinium arundinis</i>	Pathogenic, cytotoxic and antifungal potential	Chen et al. (2014), Zhang et al. (2018), Shu et al. (2022), Ji et al. (2020), Jiang et al. (2018)
<i>Diaporthe</i>	<i>Diaporthe longicolla</i>	Pathogen, bioactive potential	Zhang et al. (1999), Zhang et al. (1997), Nishad et al. (2021),
	<i>Diaporthe viciae</i>	Endophytic	This study
<i>Colletotrichum</i>	<i>Colletotrichum destructivum</i>	Pathogen, Endophytic	
	<i>Colletotrichum fructicola</i>	Pathogen, Endophytic	
<i>Plectosphaerella</i>	<i>Plectosphaerella cucumerina</i>	Pathogen, nematode-biocontrol, bioherbicide potential, Endophytic	Pétriacq et al. (2016), Atkins et al. (2003), Bailey et al. (2017),
<i>Clonostachys</i>	<i>Clonostachys eriocamporesii</i>	Pathogen, insect-biocontrol, Endophytic	Rodrigues et al. (2022)
	<i>Clonostachys ochroleuca</i>	Cytotoxic activity, Endophytic	Han et al. (2020)
	<i>Clonostachys rosea</i>	Mycoparasitic fungus, potential biological control for <i>Rhizoctonia solani</i> , Cytotoxic activity, Endophytic	Karlsson et al. (2015), Salamone et al. (2018), Han et al. (2020)
<i>Albifimbria</i>	<i>Albifimbria verrucaria</i>	Bioherbicidal, Antagonist on <i>Botrytis cinerea</i> , Pathogenic, antifungal activity against plant pathogenic fungi, Insecticidal activity, Endophytic	Weaver et al. (2021), Li et al. (2020), Gilardi et al. (2020), Nguyen et al. (2022), Assaf et al. (2020)
<i>Fusarium</i>	<i>Fusarium</i> spp.	Pathogenic, Saprobic, Endophytic	

Supplementary Table 8 Fungaltraits_vs_FUNGuild.

Below is the link to the electronic supplementary material.

[Supplementary Table 8](#)