

ASCOMYCETES FROM KARST LANDSCAPE OF GUIZHOU PROVINCE IN CHINA

JINFENG ZHANG

DOCTOR OF PHILOSOPHY
IN
BIOSCIENCES

SCHOOL OF SCIENCE
MAE FAH LUANG UNIVERSITY
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Jinfeng Zhang

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ABSTRACT

Karst landscape has become a research hotspot as it play important roles in ecosystem and provide a unique ecological environment for animals, plants and microorganisms. Although ascomycetes is the largest group within the kingdom of fungi, and numerous novel taxa were discovered from karst in recent years, a systematic taxonomy on ascomycetes from this special habitat has never been reported. In order to improve the understanding of ascomycetes from karst landscape, lots of dead woody or herbaceous plant materials were collected from Guizhou Province, China for isolation and identification by employing molecular analysis and morphological characteristics. And a total of 76 species were identified, and which are distributed into 3 classes, 15 orders, 36 families and 59 genera. Among these taxa, Dothideomycetes is the dominant group that accounts for 61.8%, and the proportion of Sordariomycetes and Leotiomycetes are 32.9% and 5.3%, respectively.

Within Dothideomycetes collected in this study, two families (Paraleptosphaeriaceae and Profundusphaeriaceae), six genera (*Hydeopsis*, *Karstiomyces*, *Neodictyospora*, *Paraleptosphaeria*, *Profundusphaeria* and *Pseudoophiosphaerella*) with 27 species were established as new to science, based on morphological characteristics and phylogenetic affinities. In addition, the natural classification placement of Eriomycetaceae and an overlooked genus *Phragmogibbera* was also resolved in this study.

The family Eriomycetaceae was initially established to accommodate five non-lichenized genera, but synonymized under Monoblastiaceae based mainly on molecular analysis. In the present study, it was resurrected as a separate family to accommodate 9 non-lichenized genera (*Eriomyces*, *Funbolia*, *Haudseptoria*, *Heleiosa*, *Italiofungus*, *Karstiomyces*, *Neoheleiosa*, *Phellinocrescentia* and *Pseudopassalora*) and placed within Monoblastiales, based on morphological and multi-gene phylogenetic evidence. While the genus *Phragmogibbera* has always been overlooked in previous studies and accommodated within Dothideomycetes genera *incertae sedis*. In this study, two phragmogibbera-like samples were collected and included in our phylogenetic analysis. And the results showed that *Phragmogibbera* was placed within Leptosphaeriaceae, and the genus *Praeclarispora* should be a synonym of this genus.

Among the taxa of Sordariomycetes collected in this study, one order Planisphaeriales, two families (Neoleptosporellaceae and Planisphaeriaceae), 6 genera (*Conicumisphaeria*, *Neoceratosphaeria*, *Neodiaporthe*, *Planisphaeria*, *Stellatus* and *Truncatascus*), and 13 species were introduced, based on morphological traits conjunction with phylogenetic analysis. The order Planisphaeriales was introduced to accommodate its monotypific family Planisphaeriaceae and included within the subclass Sordariomycetidae. While the family Neoleptosporellaceae was established to accommodate *Neoleptosporella* species and placed within Chaetosphaeriales, based on the present phylogenetic affinities and morphological distinctions.

Another group that discovered during this study is Leotiomycetes. Although Rhytismataceae is the largest family within Leotiomycetes, there are only four species were collected, and all of these taxa were identified as new to science. The genetic relationship within Rhytismataceae has been problematic and challenged due to lack a systematic genus-level taxonomic revision in the present and previous studies. Hence, more fresh collections and molecular data are needed for revision of this family in the future study.

A brief notes for each species, genus, family and order that mentioned in this study were updated and provided, based on the current classification system and relative publications. In addition, 44 new species, 9 geographical records and 1 combination introduced in this study are as follows: Novel species: *Atractospora terrestrica*, *Conicumisphaeria vaginasporum*, *Corylicola hydei*, *Diaporthe cylindriciospora*, *Dictyosporium karstium*, *Dothiorella sexualis*, *Dyfrolomyces maolanensis*, *Hydeomyces pinicola*, *Hydeopsis verrucispora*, *Hypoderma paralinderae*, *Hysterobrevium karstium*, *Karstiomyces guihzouensis*, *Lophiotrema hydei*, *Leptospora hydei*, *Lophiotrema karstii*, *Neoaquastroma guizhouense*, *Neoceratosphaeria karstiea*, *Neodiaporthe reniformesporum*, *Neodictyospora karstia*, *Neoheleiosa guihzouensis*, *Neoleptosporella fusiformispora*, *Neoophiobolus filiformesporum*, *Occultibambusa jonesii*, *Occultibambusa maolanensis*, *Ophioceras guizhouense*, *Ophiosphaerella karstii*, *Paraleptosphaeria guizhouensis*, *Paraeutypella elongiascus*, *Paraeutypella karstia*, *Phragmogibbera karstia*, *Planisphaeria karstium*, *Planisphaeria reniformisporum*, *Plenodomus karsti*, *Plenodomus macrosporium*, *Poaceascoma herbacum*, *Profundusphaeria fusiformispora*, *Pseudocoleophoma guizhouensis*, *Pseudoophiosphaerella huishuiensis*, *Stellatus guizhouensis*, *Terriera karsti*, *Terriera meitanensis*, *Terriera sigmoideospora*, *Tremateia guiyangensis*, *Truncatascus microsporum*; New geographical records: *Acrocalymma medicaginis*, *Annulohypoxylon thailandicum*, *Astrosphaeriella bambusae*, *Lophiotrema neohysterioides*, *Ophiosphaerella agrostidis*, *Ophiosphaerella chiangraiensis*, *Psiloglonium macrosporum*, *Thyridaria broussonetiae*, *Tremateia chromolaenae*; New combination: *Phragmogibbera artemisiae.*

Keywords: Ascomycota, Dothideomycetes, Karst, Leotiomycetes, Phylogeny, Sordariomycetes, Taxonomy

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ABBREVIATION AND SYMBOL

cm	centimeter
com.	combination
diam.	diameter
et al.	and others
eg.	for example
gen.	Genus (singular) and genera (plural)
g/l	gram per liter
i.e.	that is
mg	milligram
mm	millimeter
nov.	novum (Latin for new thing)
rpm	revolutions per minute
sp.	species
spp.	species (plural)
$\bar{x}$	average
=	heterotypic (taxonomic, facultative) a synonym
µg/ml	microgram per milliliter
µm	micrometer
µl	microliter
viz.	videlicet
vs.	versus
100%	percentage

CHAPTER 1

INTRODUCTION

1.1 Introduction of Karst Landscapes

Karst landscape is recognized as a special ecological environment, and generally characterized by sinking streams, caves, enclosed depressions, fluted rock outcrops and large springs (Ford & Williams, 2007). The latest World Karst Aquifer Map (WOKAM) showed that karst is widely distributed worldwide and encompass about 15% of land surface, which are usually occur on plains, hills and mountains (Goldscheider et al., 2020). Based on the geological characteristics, elevation, expansion, hydrology and the morphology of surface, there are 81 karst types were categorized, and the main characteristics of all these types were also described (Veress, 2020).

The karst in Southwest China is one of the largest karst areas around the world, which are particularly concentrated in the provinces of Guangxi, Guizhou and Yunnan (Huang & Cai, 2009). Of which Guizhou is known as the “Karst province” of China since 73.6% of the total area of this province is made up of karst landscapes (Zhang et al., 2006). The type of karst in Guizhou Province is Plateau-canyon type, which has initially developed as Peak-forest type before the Pliocene, and evolved to Plateau-canyon type after Pliocene because of the Neotectonic movement and climate influence (Zhang et al., 1980). Due to irrational and intensive land use, the ecological environment of Guizhou Province is extremely fragile, and which led to soil erosion, extensive exposure of basement rocks and drastic decrease in soil productivity (LaMoreaux et al., 1997; Wang et al., 2004). Nevertheless, the biodiversity in this Province is richness, with 264 families, 1667 genera and 7505 vascular plants species were inventoried from Guizhou Province (Liu et al., 2018).

1.2 Introduction of Ascomycetes

Ascomycetes are the largest group of true fungi, and which are significantly different from other taxa by production of sexual spores in a sac-like ascus (Whittaker, 1959; Taylor et al., 1999; Hibbett et al., 2007). The oldest fossil ascomycetes was found from the Early Devonian that associated with the vascular plant *Asteroxylon*, and then identified as *Paleopyrenomycites devonicus* (Taylor et al., 1999, 2005). And this species has been widely used as a calibration to estimate the divergence time of Ascomycetes, especially the higher ranking taxa (Taylor & Berbee, 2006; Hongsanan et al., 2017; Liu et al., 2017; Samarakoon et al., 2019, 2022). Members of Ascomycetes are widely distributed in terrestrial, marine and freshwater habitats worldwide, and which are associated with variety of plants, animals and green algae (Taylor et al., 2005; Hibbett et al., 2007; Kirk et al., 2008). With the rapid development of molecular biology, the phylogenetic analysis was universally used in the taxonomy of Ascomycetes, and yield a series of monograph to update the classification system, especially on the two largest of classes Dothideomycetes and Sordariomycetes (Zhang et al., 2012; Hyde et al., 2013, 2020c; Maharachchikumbura et al., 2015; Hongsanan et al., 2020a, 2020b).

Ascomycetes play an important role in forest ecosystem as they can decompose the plant litter through soil nutrient recycle (Osono & Takeda, 2002, 2006; Osono et al., 2003). Moreover, they are also important to human beings as members of this group are usually reported as pathogens of economic plants, humans and animals (Bowen et al., 2011; Ahmed et al., 2017). In addition, some ascomycetous fungal species are widely used in industry for production of food, vitamins and natural pigments (Spatafora, 1992; Gmoser et al., 2017).

The low correlation between morphological characteristics and phylogenetic information resulted in several conflicted classification schemes were proposed by mycologists (Barr, 1983, 1990; Eriksson & Hawksworth, 1993; Hawksworth, 1995). Hence, Liu et al. (1999) employed the Protein-coding gene (Rpb2) to explore the phylogenetic relationships among Ascomycetes, and indicated that it is comprised of three major lineages *viz*. Archiascomycetes, Saccharomycetales, and Euascomycetes.

With the rapid development of fungal taxonomy in the past two decades, the classification system is constantly updated (Eriksson et al., 2003, 2004; Lumbsch & Huhndorf, 2007; Wijayawardene et al., 2018, 2020, 2022). Until now, 22 classes, 160 orders with more than 650 families were accepted in Ascomycota (Wijayawardene et al., 2022).

1.3 Why this Study is Carried Out?

Karst has become a research hotspot in recent years. This may be attributed that it plays important roles in ecosystem (Liu et al., 2018). Falkowski et al. (2000) indicated that more than 99% of carbon were stored on sedimentary carbonates, and it is therefore essential in global carbon cycle (Cao et al., 2016). Moreover, there are around 9.2% of the global population consume the groundwater from karst as it is a tremendous storage (Stevanović, 2019). In addition, karst also develops a unique geological and biological composition, resulting in lots of new species of animals, plants and microorganisms were continuously reported from this habitats (Beruete et al., 2002; Quang et al., 2007; Grismer et al., 2008, 2016; Grismer & Chan, 2009; Schneider et al., 2011; Xu et al., 2012; Mo et al., 2015; Zhang et al., 2017b, c, e, 2018, 2019, 2020a, 2021b; Cai et al., 2020a; Liu & Rao, 2021).

The earliest study that mentioned the ascomycetes from karst landscape of Guizhou Province was published by Wu et al. (1993), however, they mainly focused on the species composition and ecological distribution instead of a systematic classification. Since then, there has almost no related reports on the taxanomy of Ascomycetes in this special habitat. With the increasing attention on karst landforms from different fields of researchers, an ongoing investigation of fungal diversity from this special ecological environment in Guizhou Province have been carried out in the past few years, and yield numerous novel taxa based on morphological and phylogenetic analysis (Ariyawansa et al., 2016a, b, c; Chen et al., 2020, 2021; Zhang et al., 2017b, c, 2018, 2019, 2020a; Dissanayake et al., 2020, 2021).

The numbers of fungi were estimated and ranged from 500,000 to 13.2 million in past decades (Hawksworth, 1991, 2001, 2012; Arnold et al., 2000; O'Brien et al.,

2005; Schmit & Mueller, 2007; Blackwell, 2011; Hawksworth & Lücking, 2017; Wu et al., 2019), and the estimation (2.2 to 3.8 M) from Hawksworth and Lücking (2017) is generally accepted by mycologists. Currently, approximately 120,000 species of described fungi, only representing 3% to 8% of the estimation of global fungal diversity (Hawksworth & Lücking, 2017). That is to say, a large number of undescribed fungi are waiting for discovery. Hyde (2001) indicated that most of these missing fungal taxa might occur in poorly studied countries, hosts, habitats, niches or tissues, and this have been gradually confirmed in recent studies (Hyde et al., 2018b; Mapook et al., 2020a; Phukhamsakda et al., 2020; Zhang et al., 2017e, 2021b).

Consideration for the special ecological environment of karst formations in Guizhou Province, and the poorly studied on the taxonomic of ascomycetes from this area, a hypothesis is therefore proposed that it would be interesting to investigate the fungal diversity from this habitat. In addition, Ascomycetes are the largest group in the kingdom of fungi, and the natural placement of lots of taxa are confused. Hence, a systematic taxonomic on ascomycetes from karst landscapes in Guizhou Province is necessary to carry out for contribution to modern classification system, and providing a better understanding on fungal species diversity of ascomycetes.

1.4 Research Objectives

1.4.1 To investigate the diversity of ascomycetes from karst landscapes in Guizhou Province, China.

1.4.2 To contribute the taxa for modern classification system with new fresh collections and molecular sequence data.

1.4.3 To resolve the natural classification placement of confused taxa of Ascomycetes based on morpho-phylogenetic analysis.

1.5 Methods to Study of Ascomycetes in this Study

1.5.1 Specimen Collection

Dead and decaying woody/herbaceous plant materials were collected from hills or forest of karst landscapes in Guizhou Province, China (Guizhou Province, China), and then taken back to laboratory for examination.

1.5.2 Morphological Examination

Materials were examined under a stereomicroscope (Nikon SMZ168) for checking the situation of samples. Macro-morphological characteristics of fruiting bodies were captured using a stereomicroscope (Nikon SMZ800N) with a Cannon EOS 70D digital camera. Vertical sections for each specimens were made by hand using a razor blade and mounted in water for microscopy (Chomnunti et al., 2014). And the microscopic structures were observed by using a Nikon ECLIPSE 80*i* compound microscope and photographed by a Canon 600D digital camera. Measurements were processed in a Tarosoft (R) Image Frame Work version 0.9.7 program, and photographic plates were edit in Adobe Photoshop CS6 (Adobe Systems Inc., USA).

1.5.3 Single Spore Isolation

The process of single spore isolation was performed under a stereomicroscope (Nikon SMZ168) in a sterile environment. Fruiting bodies were picked up to a drop of sterile water on a small glass container with a sterilized fine forceps (Chomnunti et al., 2014). Then, the hymenium contents were broken up by using the sterile needles until the single spores suspended in the sterile water, and then sucked into a sterilized pipette or pasteur syringe, and dropped on 2 % water agar (WA) or potato dextrose agar (PDA) and incubated at an incubator (25 °C). Subsequently, the germinated ascospore was transferred to the new PDA/MEA plates for recording the growth rate and colony characteristics.

1.5.4 Herbarium and Culture Deposition

Herbarium material were naturally air-dried and then cut to a small piece associated with the target fruiting bodies. Subsequently, these materials were put in

herbarium packets individually, and labeled and deposited at the herbarium of Mae Fah Luang University (MFLU) or Guizhou Academy of Agricultural Sciences (GZAAS). And the pure cultures were cut to small agar pieces and transferred to 2 ml cryovials that containing medium slope and 15% glycerin, respectively, and then deposited in Mae Fah Luang University Culture Collection (MFLUCC) or Guizhou Culture Collection (GZCC).

1.5.5 DNA Extraction, PCR Amplification and Sequencing

In this study, following the manufacturer's instructions, the total genomic DNA was extracted from cultures using a Biospin Fungus Genomic DNA Extraction Kit (BioFlux, Hangzhou, P. R. China) or extracted from the fruiting bodies using an E.Z.N.A. Forensic DNA kit (Omega Bio-Tek, Doraville, Georgia, USA) for those fungi which could not be isolated.

Polymerase chain reactions (PCR) were performed in 25 μl reaction volumes, which contained 9.5 μl distilled-deionised-water, 12.5 μl of 2 × Power Taq PCR Master Mix (TIANGEN Co., China), 1 μl of DNA template and 1 μl of each forward and reverse primers. Eight different partial gene regions are used for phylogenetic analyses for various groups in this study. They are the large subunit of the nuclear ribosomal DNA (LSU), small subunit of the nuclear ribosomal DNA (SSU), internal transcribed spacers (ITS), translation elongation factor (TEF1-α), RNA polymerase II subunit 2 (RPB2), mitochondrial small subunit (mtSSU), α-actin (ACT) and β-tubulin (TUB2), which are amplified using the primer pairs LROR/LR5 (Vilgalys & Hester, 1990), NS1/NS4 (White et al., 1990), ITS5/ITS4 (White et al., 1990), EF1-728F/EF1-986R or EF1-983F/EF1-2218R (Carbone & Kohn, 1999; Rehner, 2001), fRPB2-5F/fRPB2-7cR (Liu et al., 1999), mrSSU1/mrSSU3R (Zoller et al., 1999), ACT512F/ACT783R (Carbone & Kohn, 1999), and T1/Bt2b (O'Donnell & Cigelnik, 1997), respectively. The PCR thermal cycle program was performed for amplification these different loci according to related publications. Quality of PCR products was checked by 1.2% agarose gel electrophoresis stained with ethidium bromide and then sent to sequence at Invitrogen Biotechnology Co., Ltd (Shanghai, P. R. China).

1.5.6 Phylogenetic Analyses

Newly generated sequence data were used to carry out the BLAST search in NCBI for preliminary identification. Additional related sequences were downloaded from GenBank based on the blast results and related publications.

Single gene sequence alignments were performed in MAFFT v. 7 (http://mafft.cbrc.jp/alignment/server/), and edited manually where necessary in the program BioEdit v. 7.2 (Hall, 1999) or MEGA v. 6.0 (Tamura et al., 2013).

Maximum-likelihood (ML) analyses including 1000 bootstrap replicates were performed in the program of RAxMLGUI v. 1.5b1 (Silvestro & Michalak, 2012). Alignments in PHYLIP format were transferred and loaded from the website (http://sing.ei.uvigo.es/ALTER). The final tree was selected among suboptimal trees from each replicate by comparing likelihood scores under the GTR+GAMMA substitution model.

Bayesian analyses were carried out by using MrBayes v. 3.2 (Ronquist et al., 2012). The best-fit model of evolution was estimated in MrModeltest 2.3 (Nylander, 2008). Posterior Probabilities (PP) (Rannala & Yang, 1996; Zhaxybayeva & Gogarten, 2002) were determined by Markov Chain Monte Carlo sampling (MCMC) in MrBayes v. 3.2. Six simultaneous Markov chains were run for 100,000,000 generations and trees were sampled every 100th generation. The temperature values were lowered to 0.15, burn-in was set to 0.25 and the run was automatically stopped as soon as the average standard deviation of split frequencies reached below 0.01.

Trees were visualized with TreeView (Page, 1996), and additionally layouts were done in the program of Adobe Illustrator CS v. 5 (Adobe Systems Inc., USA).

1.6 Research Contents

The thesis is divided into five chapters.

Chapter 1 is the general introduction consists of a brief introduction for karst landscapes and the karst in Guizhou Province, the classification studies on Ascomycetes, main reasons that drove us to carry out this research, as well as the objectives and the outline of the thesis are provided.

Chapter 2 is to deal with the taxonomic and phylogenetic studies of Dothideomycetes from karst landscapes of Guizhou Province, China. The description, illustration and notes for each newly collected species are provided. And phylogenetic analyses for these taxa are individually carried out in different rank levels.

Chapter 3 is to deal with the taxonomic and phylogenetic studies of Sordariomycetes from karst landscapes of Guizhou Province, China. The description, illustration and notes for each newly collected species are provided. And phylogenetic analyses for these taxa are individually carried out in different rank levels.

Chapter 4 is to deal with the taxonomic and phylogenetic studies of Leotiomycetes from karst landscapes of Guizhou Province, China. The description, illustration and notes for each newly collected species are provided.

Chapter 5 provides the overall conclusion of this study and suggestions for future work and a list of publications.

CHAPTER 2

TAXONOMIC AND PHYLOGENETIC OF DOTHIDEOMYCETES FROM KARST LANDSCAPE OF GUIZHOU PROVINCE

2.1 Introduction

Dothideomycetes is considered as the largest and most diverse class of ascomycota. (Kirk et al., 2008; Hyde et al., 2013, Wijayawardene et al., 2018, 2021, 2022). The notable characteristics used to define Dothideomycetes are ascolocular ascomata and bitunicate, usually fissitunicate asci (Nannfeldt, 1932; Luttrell, 1955; Barr & Huhndorf, 2001; Schoch et al., 2009). However, these characters especially the bitunicate or fissitunicate asci are also founded in other classes, such as Arthoniomycetes and Eurotiomycetes. Major members of the Dothideomycetes are pathogens, endophytes or epiphytes on living plants and lichens, and saprobes on dead or partially decaying plants or plant leaf and woody litter or on herbivore dung, as well distributed in worldwide (Schoch et al., 2006; Hyde et al., 2013). Recently, Liu et al. (2017) used molecular clock methods and establish the divergence times for the orders and families of Dothideomycetes, and recommended that subclasses of Dothideomycetes should have evolved between 235 and 250 MYA (crown age) and 260–322 MYA (stem age), orders should have evolved between 100 and 220 MYA (crown age) and 130 and 310 MYA (stem age), and families should have evolved between 20 and 100 MYA (crown age). They also suggested that some genera should be raised to family level based on divergence estimates (Liu et al., 2017). Based on the latest outline of fungi, there are 49 orders, 201 families with 1956 genera were comprised of Dothideomycetes (Wijayawardene et al., 2022) and more than 19,000 species were estimated to include within this class (Kirk et al., 2008).

Recently, a series of studies have been carried out to investigate the microscopic fungi from karst formations, and numerous novel taxa and known species were described, however, the dothideomycetous taxa occupy only a small proportion of all described taxa that collected from this habitats (Nováková, 2009; Ariyawansa et al., 2016a, b, c; Tang et al., 2016; Chen et al., 2020, 2021; Zhang et al., 2017b, c, e, 2018, 2019, 2020a, 2021b; Dissanayake et al., 2020, 2021; Feng et al., 2019; Long et al., 2021). While, Dothideomycetes is known as the largest and most diverse class of ascomycota. (Kirk et al., 2008; Hyde et al., 2013, Wijayawardene et al., 2018, 2021, 2022), therefore a hypothesis is raised that lots of members in this class are ignored. Hence, a large-scale taxonomic study of Dothideomycetes from karst formations is necessary.

2.2 Materials and Methods

2.2.1 Collection, Examination, Isolation and Specimen Deposition

See 1.5.1–1.5.4.

2.2.2 DNA Extraction, PCR Amplification and Sequencing

See 1.5.5.

2.2.3 Sequence Alignment and Phylogenetic Analysis

See 1.5.6.

2.3 Results

2.3.1 Phylogenetic Analyses

A backbone tree of Dothideomycetes is initially constructed, but without ideal topology because of the high diversity of the newly collected taxa in this study. Therefore, the individual phylogenetic trees at the order, family or genus levels were carried out for clarifying the natural placement of our new collections. All newly generated/sequenced sequence data are deposited in GenBank.

2.3.2 Taxonomy

Class Dothideomycetes

Hysteriales Lindau

Hysteriales was introduced by Lindau (1897) to accommodate the monotypic family Hysteriaceae, and stably placed in Dothideomycetes based on phylogenetic analysis since 2009 (Boehm et al., 2009a, b; Hyde et al., 2013; Jayasiri et al., 2018). In this study, we follow the latest classification treatment of Hysteriales in Jayasiri et al. (2018) and Wijayawardene et al. (2022), with updated accounts of the novel species from recent publications (Kumar et al., 2019; Valenzuela-Lopez et al., 2019; Dayarathne et al., 2020; Mapook et al., 2020).

Hysteriaceae Chevall

Hysteriaceae was established by Chevallier (1826) with *Hysterium* as the type genus, then the placement of this family has been argued because of depending only on its morphological features (Nannfeldt, 1932; Von Arx & Müller, 1954, 1975). Boehm et al. (2009a) first presented a phylogenetic analysis of Hysteriaceae based on four nuclear genes, and a reappraisal of the phylogenetic integrity and a key to the genera and allied genera of Hysteriaceae were also provided in the same year (Boehm et al., 2009b). Currently, there are 13 genera accepted in this family (Jayasiri et al., 2018; Wijayawardene et al., 2022), of which nine genera were classified on the basis of both morphology and molecular evidence (*Gloniopsis*, *Graphyllium*, *Hysterium*, *Hysterobrevium*, *Hysterodifractum*, *Oedohysterium*, *Ostreichnion*, *Psiloglonium* and *Rhytidhysteron*), while other four genera (*Actidiographium*, *Gloniella*, *Hysterocarina* and *Hysteropycnis*) based only on morphological characteristics.

Hysterium Pers., Tent. disp. meth. fung. (Lipsiae): 5 (1797)

A historical overview of the nomenclature of *Hysterium* was presented in Boehm et al. (2009a). Member of this genus are characterized by pigmented versicolorous or concolorous asymmetric phragmospores, three- or more transversely-septate, with cells often constricted (Boehm et al., 2009a). More than 400 epithets of *Hysterium* were listed in Index Fungorum (2022), however, most of these taxa were transferred to other groups and 14 species were accepted in the latest outline of fungi (Wijayawardene et al., 2022). Nevertheless, the fresh samples are needed to obtain more sequence data to delimit the genus concept of *Hysterium* as it

was showed to be polyphyletic in previous studies (Schoch et al., 2006; Boehm et al., 2009a, b; Mugambi & Huhndorf, 2009) and the present study.

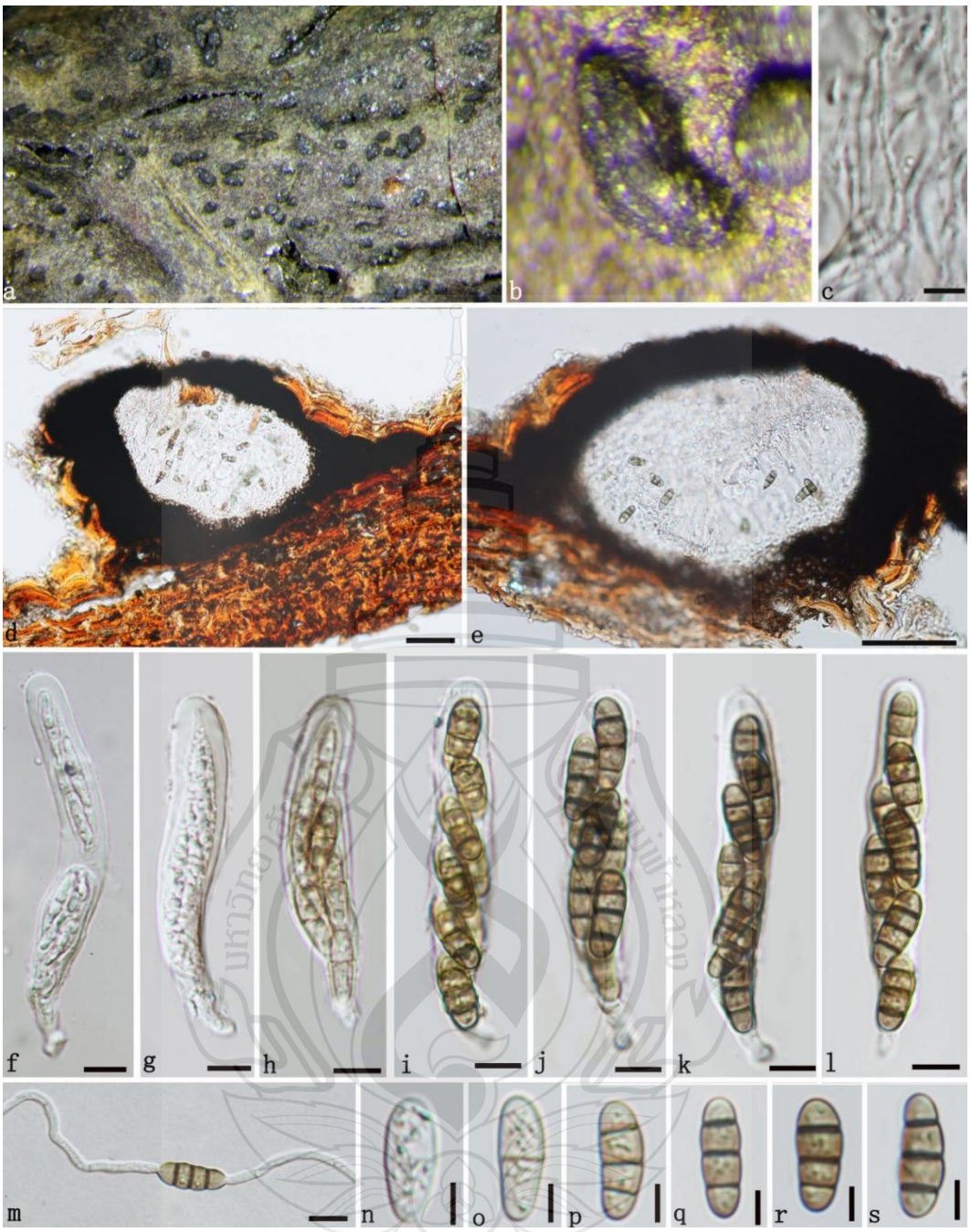
Hysterium angustatum Alb. & Schwein., Consp. fung. (Leipzig): 55 (1805), Figure 2.1

Saprobic on dead wood. Sexual morph: *Hysterothecia* 208–232 µm high, 256–284 µm wide, superficial with base immersed, carbonaceous, black, subglobose to elongate, straight or flexuous, smooth laterally, with a longitudinal slit. *Peridium* 45–65 µm wide, carbonaceous, brittle, of heavily pigmented, small prosenchymatous cells. Hamathecium comprising numerous 1.5–2 µm wide, hyaline, non-trabeculate pseudoparaphyses, anastomosing between and above the asci. *Asci* 59–86 × 10.5–13 µm ($\bar{x}$ = 68 × 11.5 µm, n = 30), 8-spored, bitunicate, cylindrical to clavate, short pedicillate, apically round with a distinct ocular chamber. *Ascospores* 13–17 × 4.5–5.5 µm ($\bar{x}$ = 15 × 5 µm, n = 30), ellipsoidal to fusiform, slightly curved or straight, hyline and two-celled when young, becoming brown and 3-septate at maturity, not constricted at the septa. Asexual morph: Undetermined.

Material examined: CHINA, Guizhou Province, Libo County, Maolan Town, Maolan National Nature Reserve, on dead wood, 24 June 2018, Y.Y. Chen 18-96 (GZAAS 19-1836); Guiyang City, Yunyan District, Guizhou Botanical Garden, on dead wood, 08 August 2017, J.F. Zhang ZWY-15 (MFLU 18-2248); Huaxi District, Guiyang City, Guizhou Academy of Agricultural Sciences, on dead wood, 28 April 2019, J.F. Zhang GZAAS-2019-5 (GZAAS 21-2007).

Notes: Present phylogenetic analysis shows that our three new collections cluster with other three isolates of *Hysterium angustatum* (GKM5211, CMW20409 and MFLUCC 16-0623), and where they form a full bootstrap supported clade (Figure 2.5). And the topology in this study is similar to Boehm et al. (2009a) and Jayasiri et al. (2018). Based only on the phylogeny, accessions of *H. angustatum* that collected various geographical locations including our new collections maybe identified as several separate species. However, this has been discussed by Boehm et al. (2009a) that the little morphological or sequence variation was detected among geographically disparate accessions of *H. angustatum*. Examination of these newly collected specimens, we found that they are resemble to *H. angustastum* and also with the overlap sized asci and ascospores, based on the description from Jayasiri et al. (2018)

and the key to the species *Hysterium* that provided by Boehm et al. (2009a). Hence, we identify these collections as *H. angustatum*.



Note a, b Appearance of hysterothecia on the host surface. c Pseudoparaphyses. d, e Vertical section through hysterothecium. f–l Asci. m Germinated ascospore. n–s Ascospores. Scale bars: c = 10 μm, d, e = 50 μm, f–m = 10 μm, n–s = 5 μm.

Figure 2.1 *Hysterium angustatum* (GZAAS 19-1836)

Hysterobrevium E. Boehm & C.L. Schoch, Stud. Mycol. 64: 62 (2009)

Based on morphological and molecular analysis, Boehm et al. (2009b) introduced *Hysterobrevium* to accommodate *H. constrictum*, *H. mori* and *H. smilacis*, with *H. mori* as the type species. Subsequently, other three species *viz. H. curvatum*, *H. hakeae*, *H. rosae* and *H. walvisbayicola* were comprised of this genus (Jayasiri et al., 2018; Crous et al., 2021a), and 7 species were currently accommodated in *Hysterobrevium*. In this study, a novel species *H. karstium* is introduced hereby on the basis of morphological examination and molecular data.

Hysterobrevium karstium J.F. Zhang, J.K. Liu & Z.Y. Liu *sp. nov.*, Figure 2.2

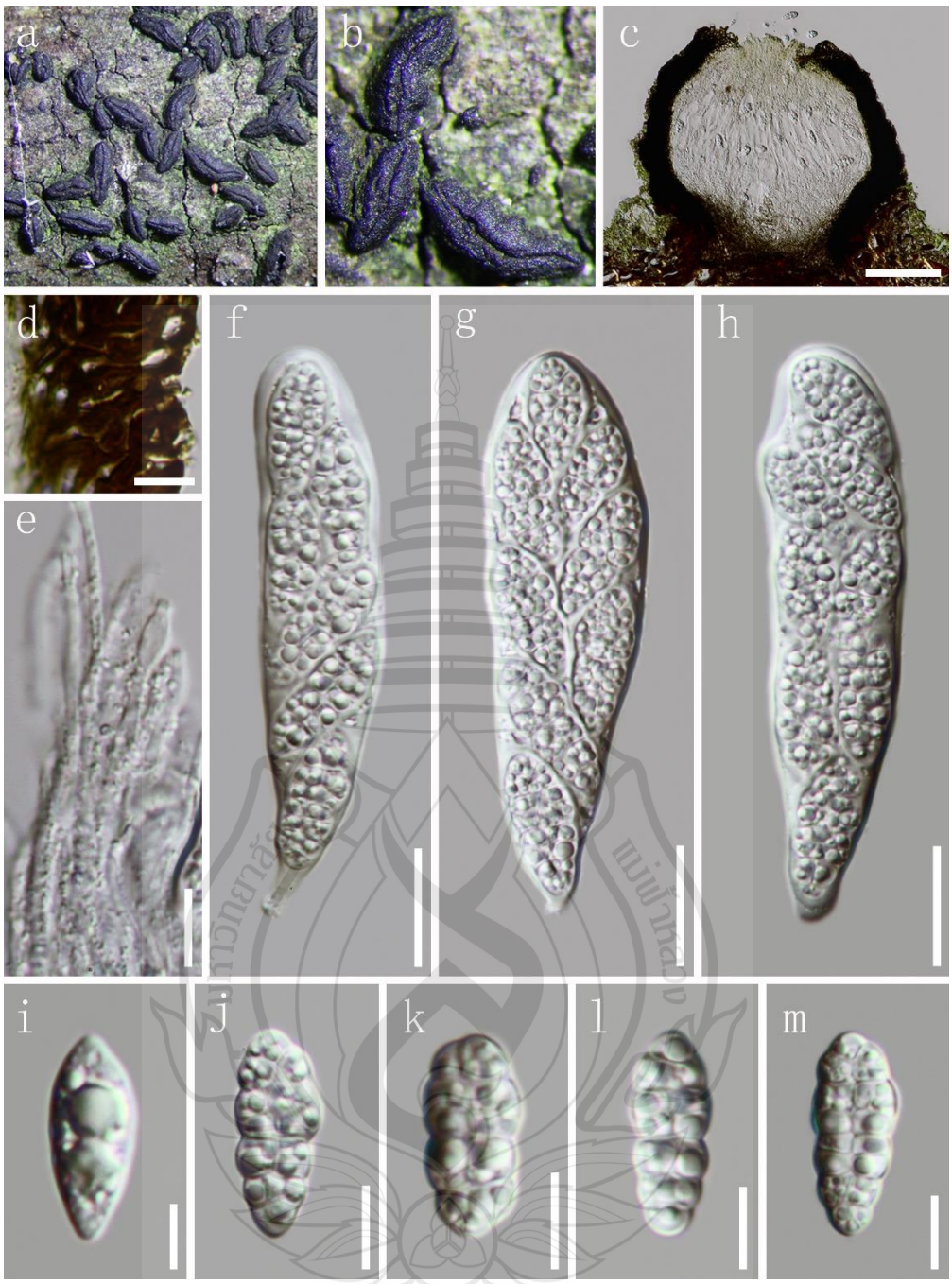
Etymology: The name "*karstium*" refers to the holotype was collected from karst formations.

Holotype: MFLU 18-2251.

Saprobic on dead twigs of woody plants. Sexual morph: *Hysterothecia* erumpent, ellipsoidal, oblong, usually surrounded at the base by ruptured epidermis or periderm cells, longitudinally striated, navicular with tapering ends. *Peridium* 25–45 μm wide, narrower at base within the substrate, widest at mid-point, carbonaceous and brittle when dry. *Hamathecium* consisting of 1–2 μm wide, cellular, septate, persistent pseudoparaphyses, hyaline to pale yellow in mass, branched above, forming an epithecium, but not darkly pigmented, exposed surface yellow-brown. *Asci* 89–100(–106) × 20–23 μm ($\bar{x}$ = 96 × 21.5 μm, n = 20), 8-spored, cylindrical to clavate, bitunicate, short-stipitate. *Ascospores* 22–28(–35) × 8.5–11(–14) μm ($\bar{x}$ = 27 × 10.5 μm, n = 35), asymmetric, hyaline, dictyospores, tapering at both ends, with 5–8 transverse and 1–2 longitudinal septa, constricted along multiple transverse septa, and a gelatinous sheath that usually dissipates at maturity. Asexual morph**:** Undetermined.

Material examined: CHINA, Guizhou Province, Guiyang City, Yunyan District, Guizhou Botanical Garden, on dead branch of woody palnt, 8 August 2017, J.F. Zhang ZWY-20 (MFLU 18-2251, holotype); Leishan County, on dead twigs of woody plants, 2 November 2017, J.F. Zhang LS-11 (GZAAS 19-1759); Guiyang City, Yunyan District, Shunhai Park, on dead branch of woody plants, 6 May 2015, H.Y. Ran SH-04 (MFLU 18-2287); Guizhou Province, Shibing County, on dead branch of woody plant, 19 June 2020, J.F. Zhang YTS-2020-9 (GZAAS 21-2008).

Notes: In the present phylogenetic study (Figure 2.5), *Hysterobrevium karstium* sisters to *H. constrictum* and *H. hakeae* with well support. Morphologically, they are similar in having ellipsoidal, oblong hysterothecia and hyaline ascospores, nevertheless, the new collection is obviously distinguished from *H. constrictum* and *H. hakeae* by its comparatively larger ascospores (*H. karstium* 22–28(–35) × 8.5–11(–14) μm vs. *H. constrictum* (11–)13–20(–23) × 5–12 μm vs. *H. hakeae* 18–22 × (5.4–)8.5–11.5 μm) (Boehm et al., 2009a; Jayasiri et al., 2018). The measurements of asci and ascspores of *H. karstium* corresponds to *H. smilacis*, however, the latter was described by having pigmented ascospores at maturation, while this feature was not observed in our new specimen (Boehm et al., 2009b). In view of the above evidences, *Hysterobrevium karstium* was identified as a new species and introduced herein.



Note a, b Hysterothecia on host. c Vertical section through hysterothecium. d Section of peridium. e Pseudoparaphyses. f–h Asci. i–m Ascospores. Scale bar: c = 100 µm, d, e, j–m = 10 µm, g, h = 20 µm, i = 5 µm.

Figure 2.2 *Hysterobrevium karstium* (MFLU 18-2251, holotype)

Psiloglonium Höhn., Annls mycol. 16(1/2): 149 (1918)

Psiloglonium was introduced by von Höhnel (1918) and subsequently typified with *P. lineare*. Then, there has always been debated on the classification between *Glonium* and *Psiloglonium* until Boehm et al. (2009a) reinstated *Psiloglonium* within Hysteriaceae based on morphological and molecular analyses (von Höhnel, 1918; Petrak, 1923a, b; Zogg, 1962; von Arx & Müller, 1975). So far, there are 16 species listed in Index Fungorum (2022), and 7 of these taxa (*P. araucanum*, *P. clavisporum*, *P. colihuae*, *P. macrosporum*, *P. multiseptatum*, *P. sasicola*, *P. simulans*) with available sequence data in GenBank.

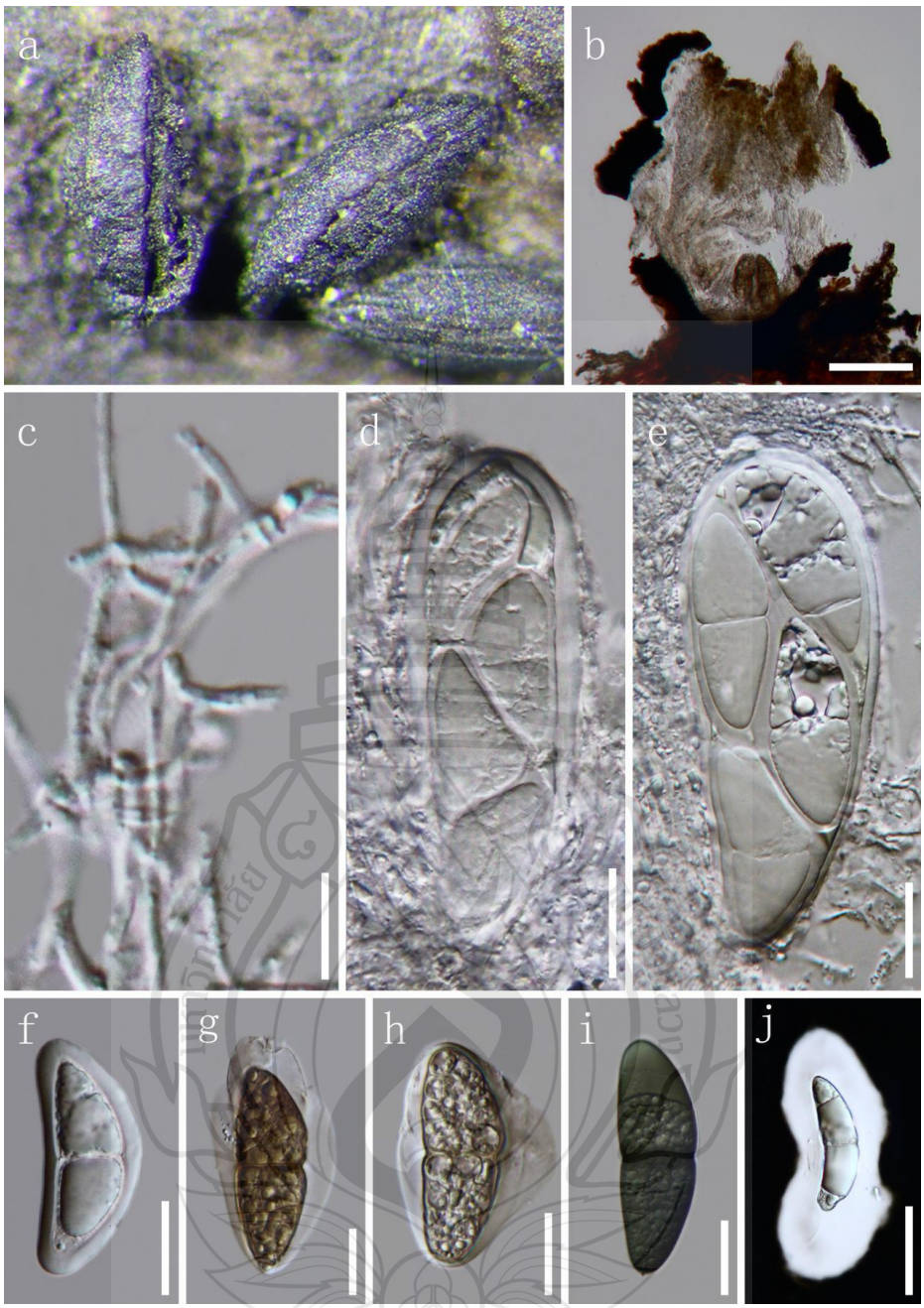
Psiloglonium macrosporum Thambug., Senan. & Hyde, in Li et al., Fungal Diverse. 78: 26 (2016) *new record*, Figure 2.3

Saprobic on dead twigs of woody plants. Sexual morph: *Hysterothecia* scattered, superficial, base immersed in the substrate, elongate and depressed conchate, globose, surface black, shiny, longitudinally striate, apex compressed, opening by a longitudinal slit. *Peridium* 25–55 μm wide, carbonaceous, brittle, comprising heavily pigmented, small, prosenchymatous cells. *Hamathecium* comprising 1.5–2.5 μm wide, hyaline, aseptate, branched, trabeculate pseudoparaphyses, in a gelatinous matrix. *Asci* 141–157.5 × 46.5–57.5 μm ($\bar{x}$ = 151.5 × 53 μm, n = 10), bitunicate, 4-spored, oblong to clavate, with a very short pedicel or apedicellate, apically thickened, with a distinct ocular chamber. *Ascospores* (73–)85– 95.5(–105.5) × 27–32.5 μm ($\bar{x}$ = 91 × 30 μm, n = 25), crowded to biseriate, fusiform, two-celled, constricted at the septum, hyaline when young, become brown and verrucose at maturity, surrounded by a thick mucilaginous sheath. Asexual morph: Undetermined.

Material examined: CHINA, Guizhou Province, Libo County, Maolan Town, on dead twigs of woody plants, 20 July 2016, H.Y. Ran MLCC-7 (MFLU 18-2218).

Notes: The multi locus phylogenetic analysis (Figure 2.5) shows that our new collection (MFLU 18-2218) clusters with reported taxon of *Psiloglonium macrosporum* with strong statistical support. Observation of the gross morphological characteristics of the new specimen, we found it also well fits to *P. macrosporum* that described by Li et al. (2016) and Jayasiri et al. (2018). However, the measurements of asci and ascospores are differ between them. Nevertheless, this is not enough to

warrant them as separate species as the specimens may be in different states of maturity, and we choose here to simply expand the asci and ascospores measurements to (110–)145–168(–215) × (28–)46.5–60 μm and (50–)80–100(–115) × (17–)25–32.5 μm respectively. Based on combination of morphological and molecular evidence, the new collection is therefore identified as *P. macrosporum* and reported as a new record for China since it has only discovered from Thailand before (Li et al., 2016; Jayasiri et al., 2018).



Note a Appearance of apothecia on substrate. b Vertical section through apothecium. c Pseudoparaphyses. d, e Asci. f–j Immature and mature ascospores. Note j is stained with India ink. Scale bar: b = 200 µm, c = 20 µm, d–i = 50 µm, j = 100 µm.

Figure 2.3 *Psiloglonium macrosporum* (MFLU 18-2218, new record)

Rhytidhysteron Speg. [as '*Rhytidhysterion*'], Anal. Soc. cient. argent. 12(4): 188 (1881)

Rhytidhysteron was introduced by Spegazzini (1881) to accommodate *R. brasiliense* and *R. viride*, but none of these two were designated as the type. Clements and Shear (1931) designated *R. brasiliense* as the type species of *Rhytidhysteron*, but this genus was subsequently placed within Patellariaceae by various authors (Kutorga & Hawksworth, 1997; Eriksson et al., 2001; Lumbsch & Huhndorf, 2010). However, recent multi-gene phylogenetic analyses clearly indicated that *Rhytidhysteron* was nested in Hysteriaceae (Boehm et al., 2009a, b; de Almeida et al., 2014; Thambugala et al., 2016; Jayasiri et al., 2018; Wijayawardene et al., 2018; Kumar et al., 2019), and around 30 species were accepted in this genus until now.

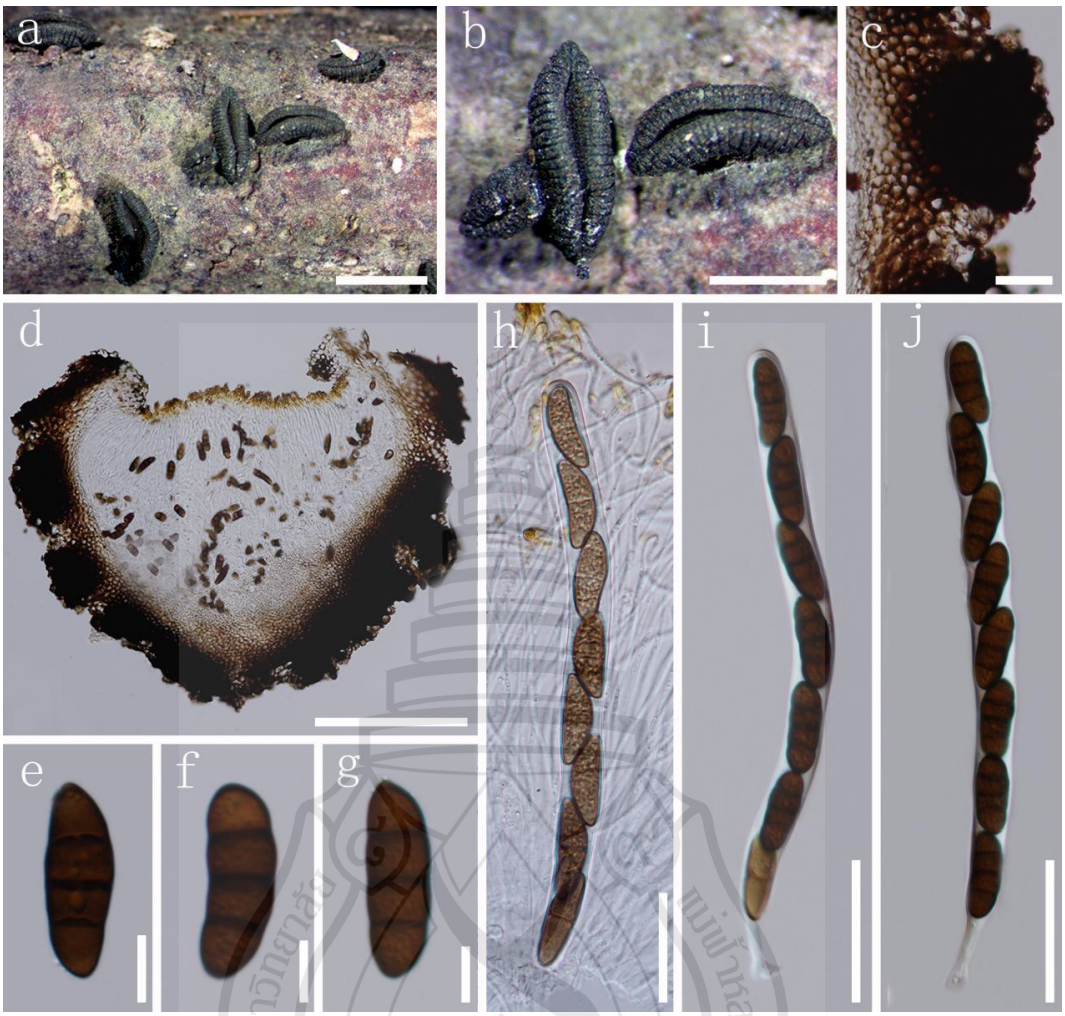
Rhytidhysteron rufulum (Spreng.) Speg., Anal. Soc. cient. argent. 90(1-6): 177 (1921) [1920]. Figure 2.4

Saprobic on dead twigs of woody plants. Sexual morph: *Hysterothecia* 1.5–2.2 mm long × 0.5–0.9 mm wide, hysterothecial, scattered to gregarious, superficial, black, carbonaceous to coriaceous, elliptic or irregular in shape, with lenticular or irregular opening when wet, perpendicularly striate along the long axis, black or red at the center, when dry folded at the margin, forming an elongate slit. *Exciple* 26–47 μm wide, composed of two stratum, outer layer comprising dark brown to black, cells of *textura angularis* to *globosa*, inner layer of hyaline to lightly pigmented cells of *textura angularis* to *prismatica*. *Hamathecium* comprising 1.5–2.5 μm wide, dense, septate, pseudoparaphyses, branched and forming a dark epithecium above the asci, fused and slightly swollen at the apex and enclosed in a gelatinous matrix. *Asci* (217–)230–237(–245) × 13–16 μm ($\bar{x}$ = 234 × 14.5 μm, n = 15), 8-spored, bitunicate, cylindrical, with short furcate pedicel, rounded at the apex, with a distinct ocular chamber. *Ascospores* (25–)28–31(–34) × 10–12 μm ($\bar{x}$ = 30 × 11 μm, n = 30), uni-seriate, slightly overlapping, ellipsoidal to fusiform, slightly rounded or pointed at both ends, hyaline to pale brown when young, become reddish-brown to brown at maturity, 3-septate, slightly constricted at the septa. Asexual morph: Undetermined.

Material examined: CHINA, Guizhou Province, Libo County, Maolan Town, Maolan Nationla Nature Reserve, on dead branch of woody plant, 8 July 2015, H.Y. Ran MLC-16 (GZAAS 19-4015); Guiding County, on dead branch of woody plant, 4

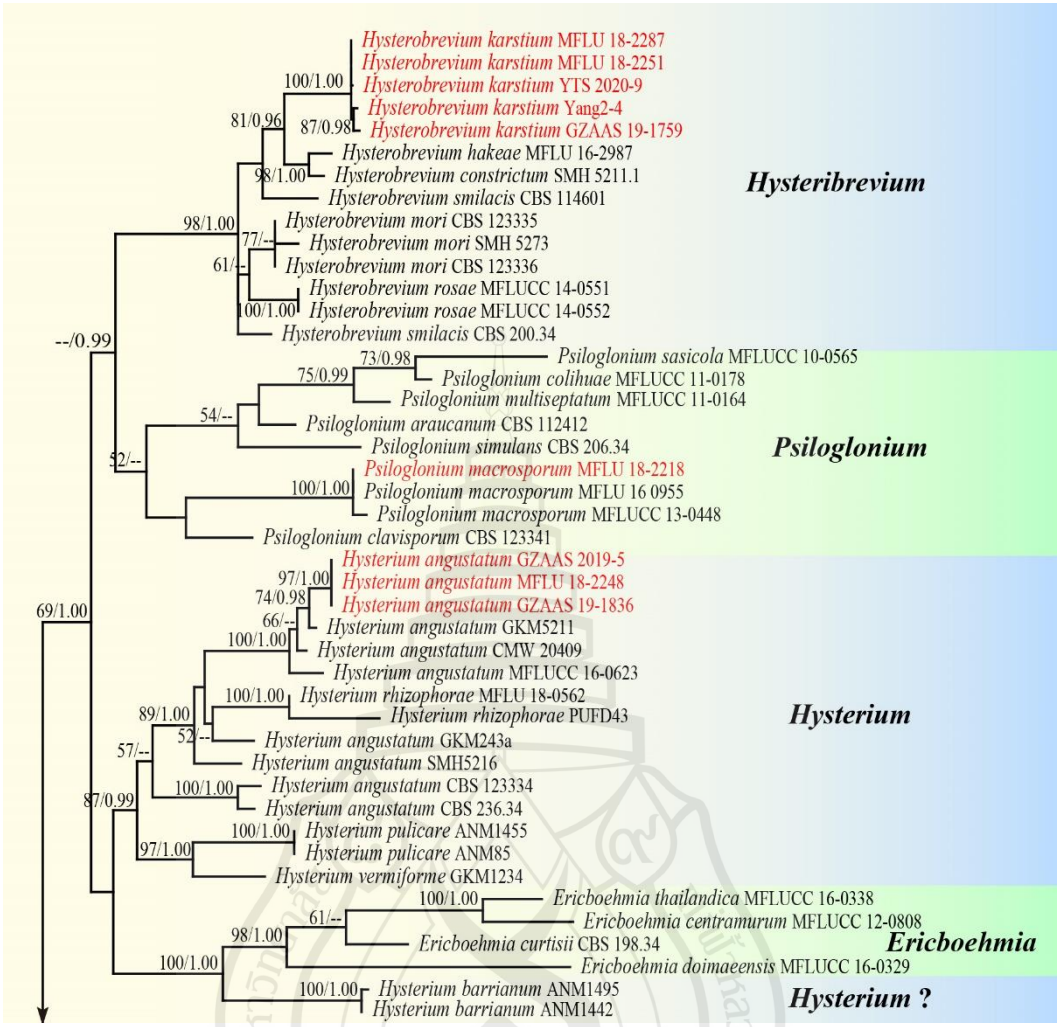
August 2017, J.F. Zhang GD-01 (MFLU 18-2260); Guiyang City, Huaxi District, on dead branch of woody plant, 2 September 2016, H.Y. Ran SD-2016-10 (GZAAS 19-4024).

Notes: Present phylogenetic analysis in this study shows that three newly collected specimens are phylogenetically related to *R. rufulum*, and where they form a robust clade (Figure 2.5). Moreover, the gross of morphological characteristics of these specimens (including the measurements of hysterothecia, asci and ascospores) are also correspond to the reference specimen of *R. rufulum* that designated by Thambugala et al. (2016). Hence, we identify these new collections as *R. rufulum*.



Note a, b Appearance of hysterothecia on substrate. c Section of peridium. d Vertical section through ascoma. e-g ascospores. h Pseudoparaphyses anastomosing between ascus. i, j Asci. Scale bars: a, b = 1 mm, c = 30 µm, d = 200 µm, e–g = 10 µm, h–j = 50 µm.

Figure 2.4 *Rhytidhysteron rufulum* (GZAAS 19-4015)



Note Maximum likelihood bootstrap values equal or greater than 50% and Bayesian posterior probabilities equal or greater than 0.95 are indicated near the nodes. New sequence generated in this study are given in red. The tree is generated from maximum likelihood (ML) analysis based on LSU, ITS, SSU and TEF1-α sequence data, and rooted with *Mytilinidion scolecosporum* CBS 305.34 and *Lophium mytilinum* CBS 269.34.

Figure 2.5 Phylogenetic tree of the family Hysteriaceae

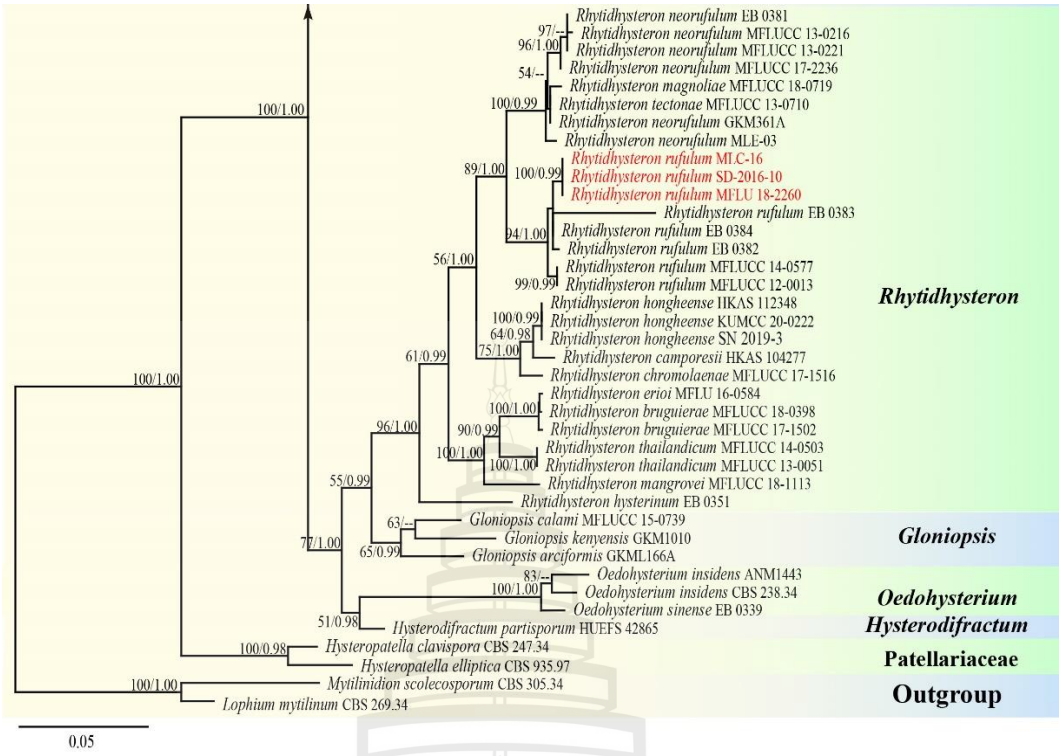
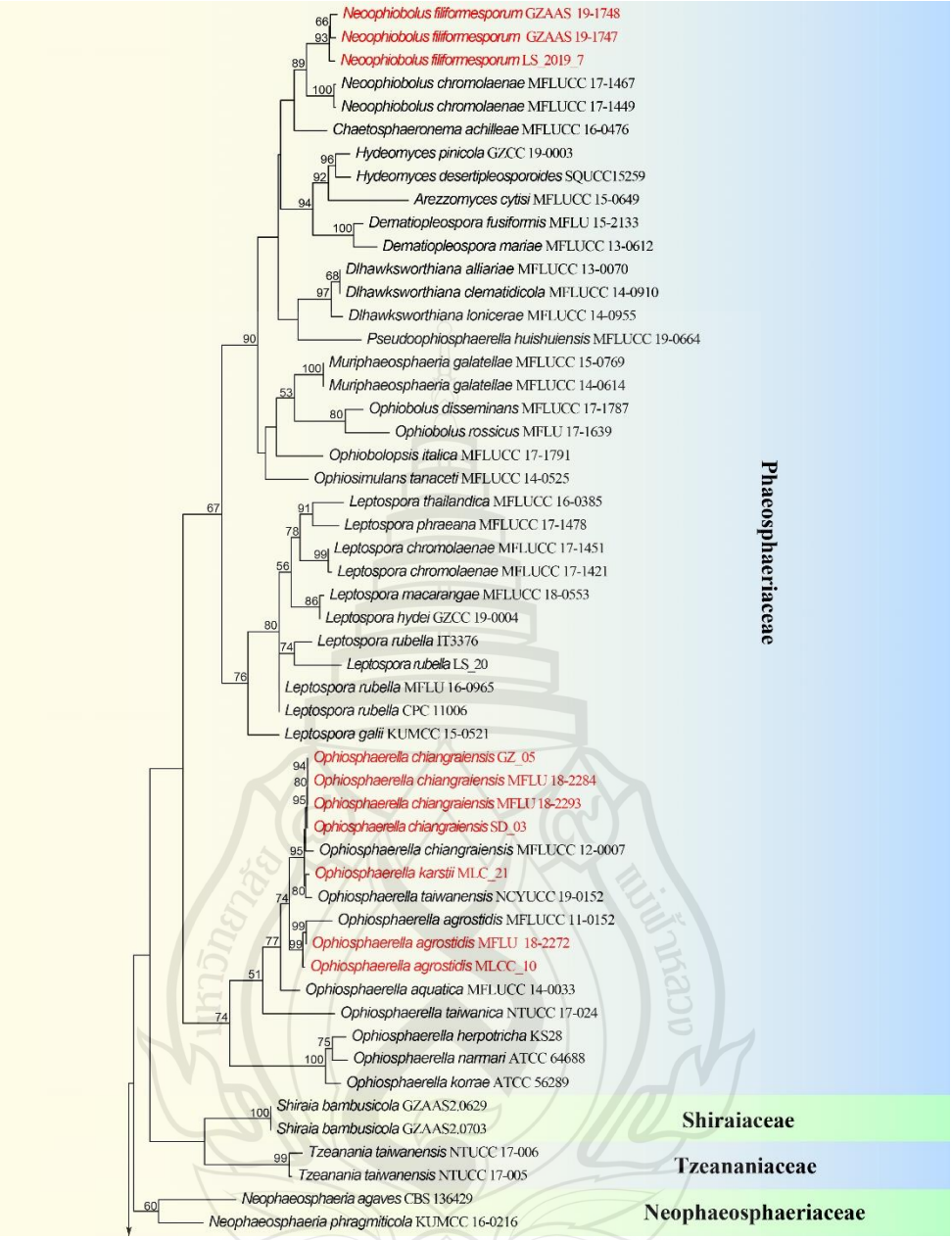


Figure 2.5 (continued)

Pleosporales Luttr. ex M.E. Barr

Pleosporales, the largest order in Dothideomycetes, was introduced by Luttrell (1955) and comprised of more than 90 families with highly diverse species distributed around the world (Zhang et al., 2012; Hyde et al., 2013; Wijayawardene et al., 2022). Members of Pleosporales are normally characterized by perithecioid ascomata typically with a papillate apex, and bitunicate, generally fissitunicate asci bearing mostly septate ascospores of various pigmentation and shapes, with or without a gelatinous sheath (Zhang et al., 2012; Hyde et al., 2013; Jaklitsch & Voglmayr, 2016; Jaklitsch et al., 2018).



Note Maximum likelihood bootstrap values equal or greater than 50% are indicated near the nodes. New sequence generated in this study are given in red. The tree is generated from maximum likelihood (ML) analysis based on LSU, Rpb2, SSU and TEF1-α sequence data, and rooted with Mytilinidion acicola EB0349 and M. californicum EB0385

Figure 2.6 Phylogenetic tree of the order Pleosporales

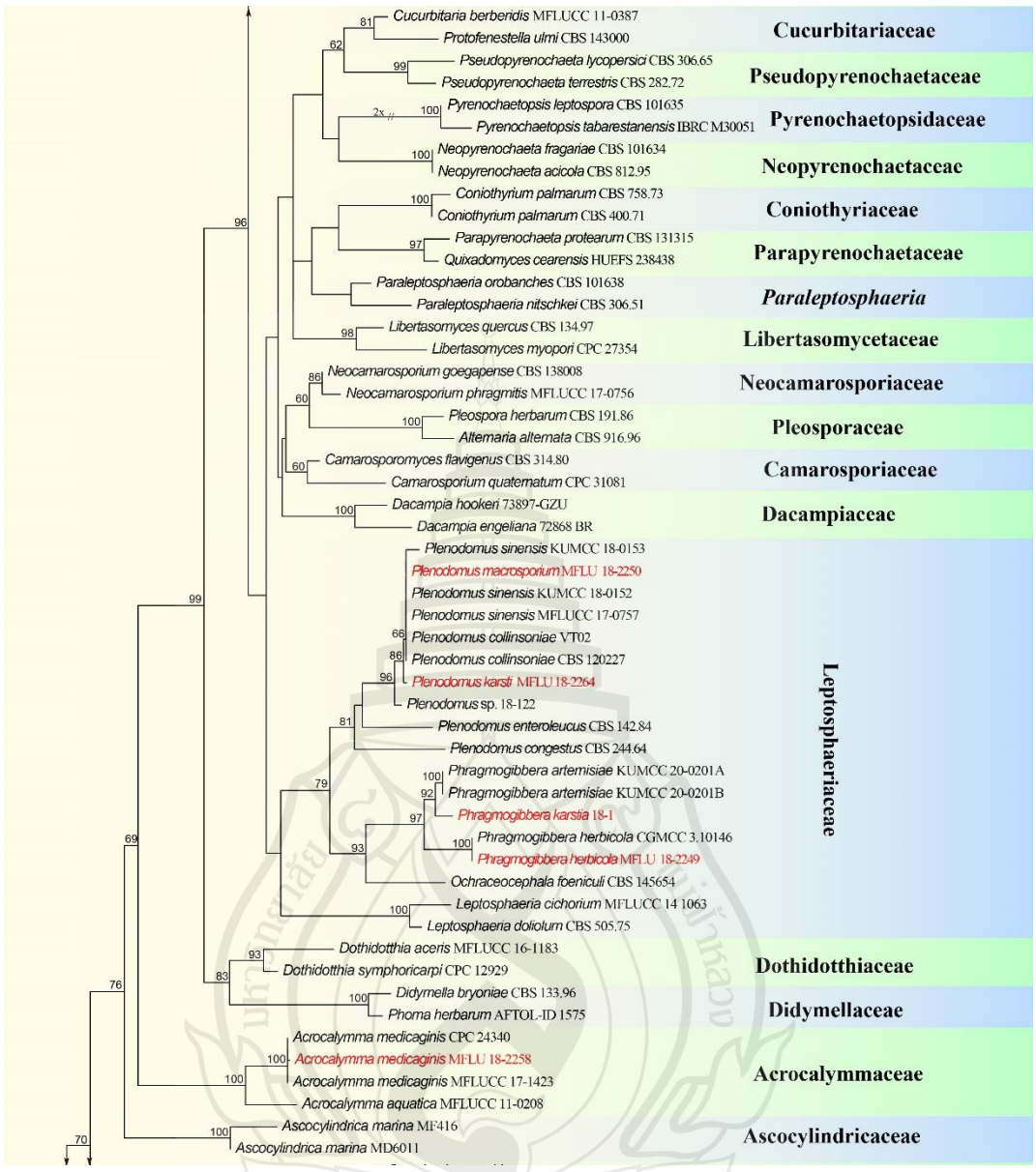


Figure 2.6 (continued)

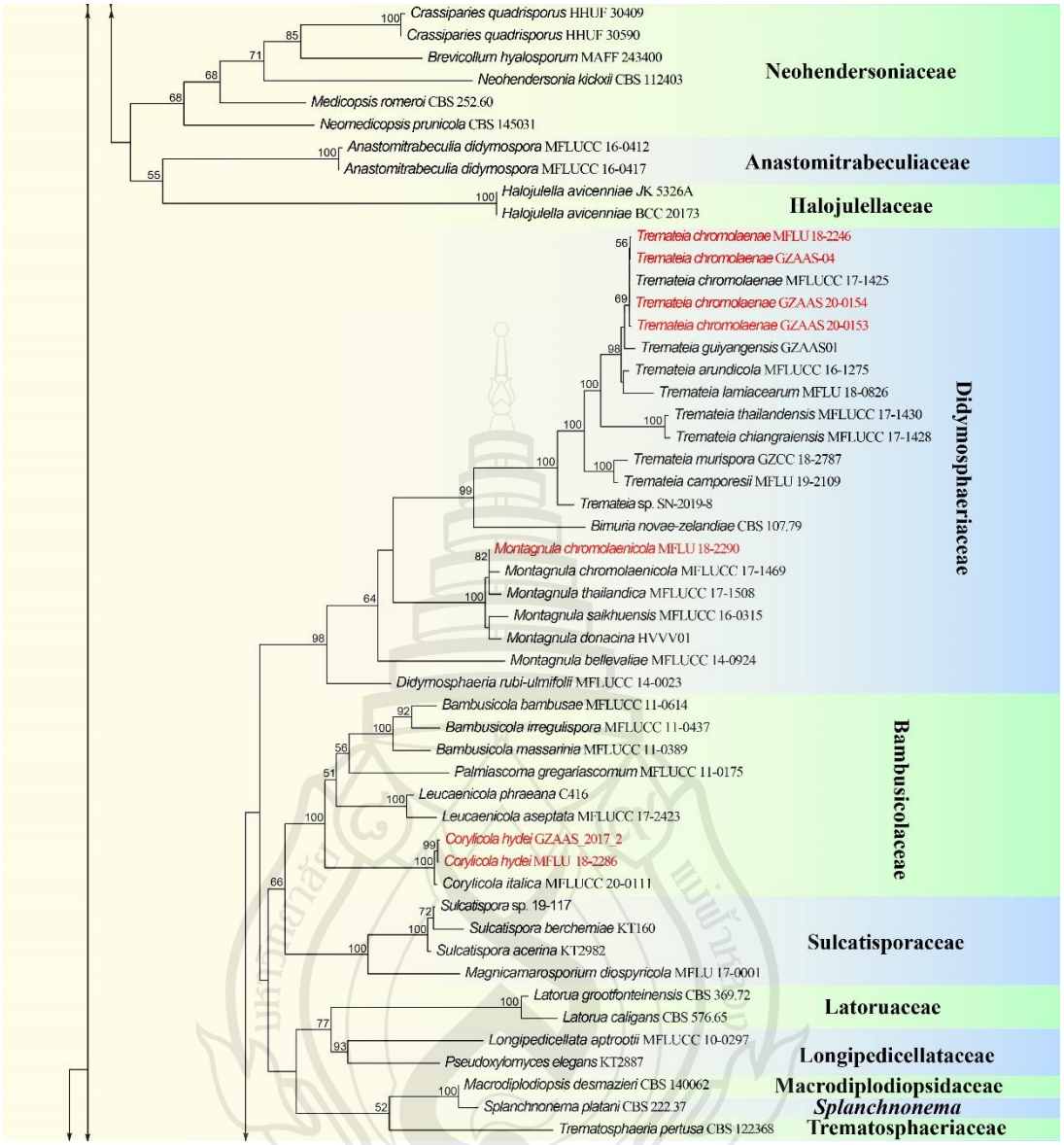


Figure 2.6 (continued)

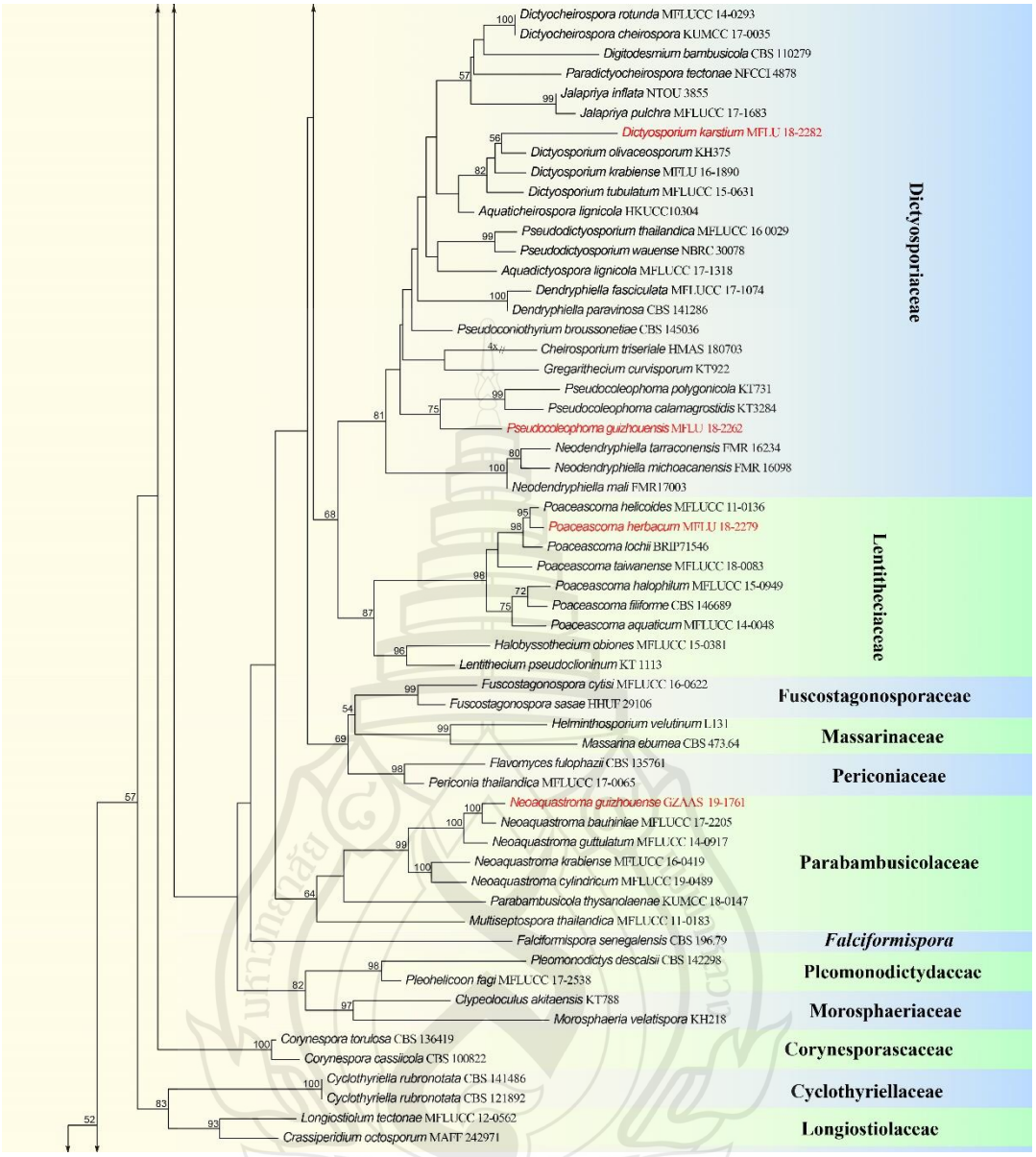


Figure 2.6 (continued)

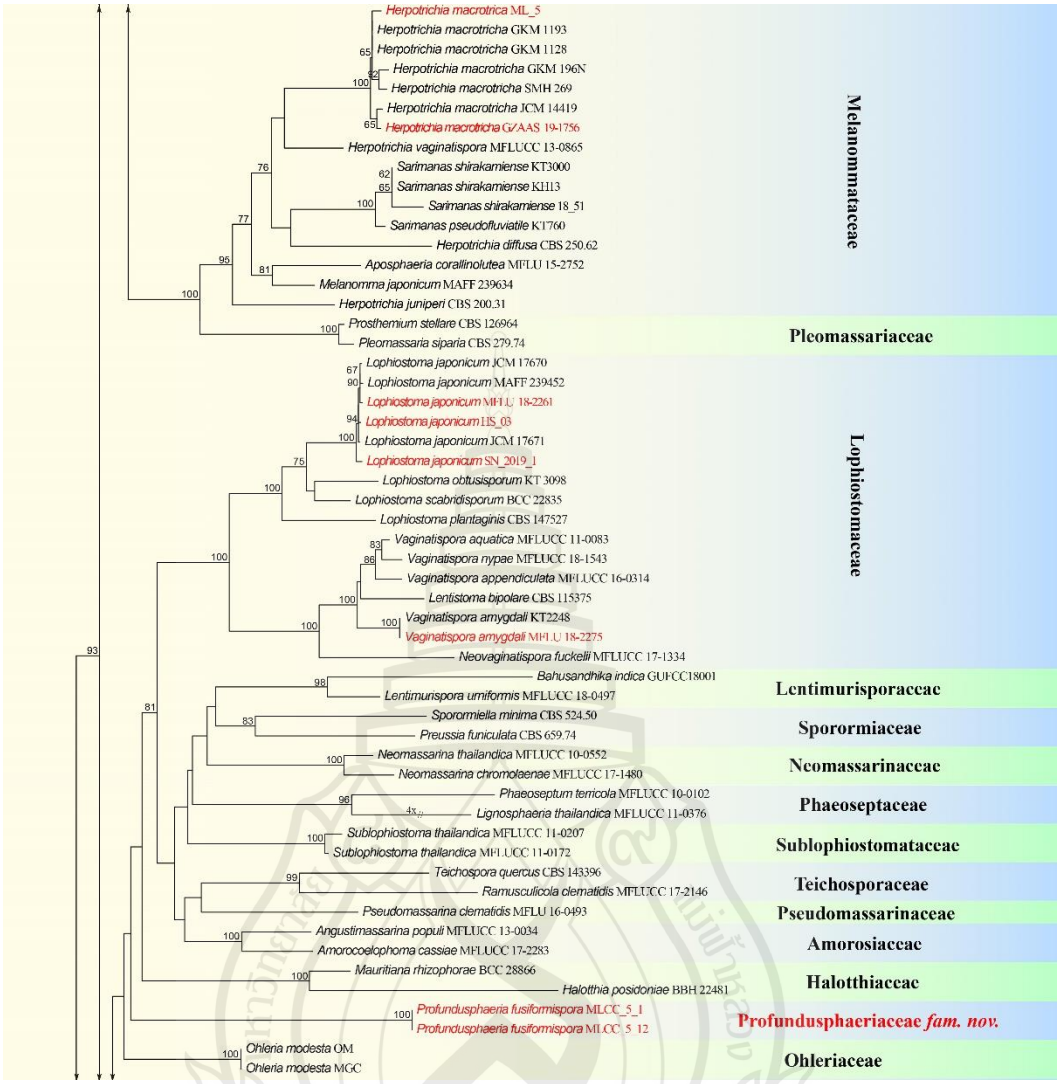


Figure 2.6 (continued)

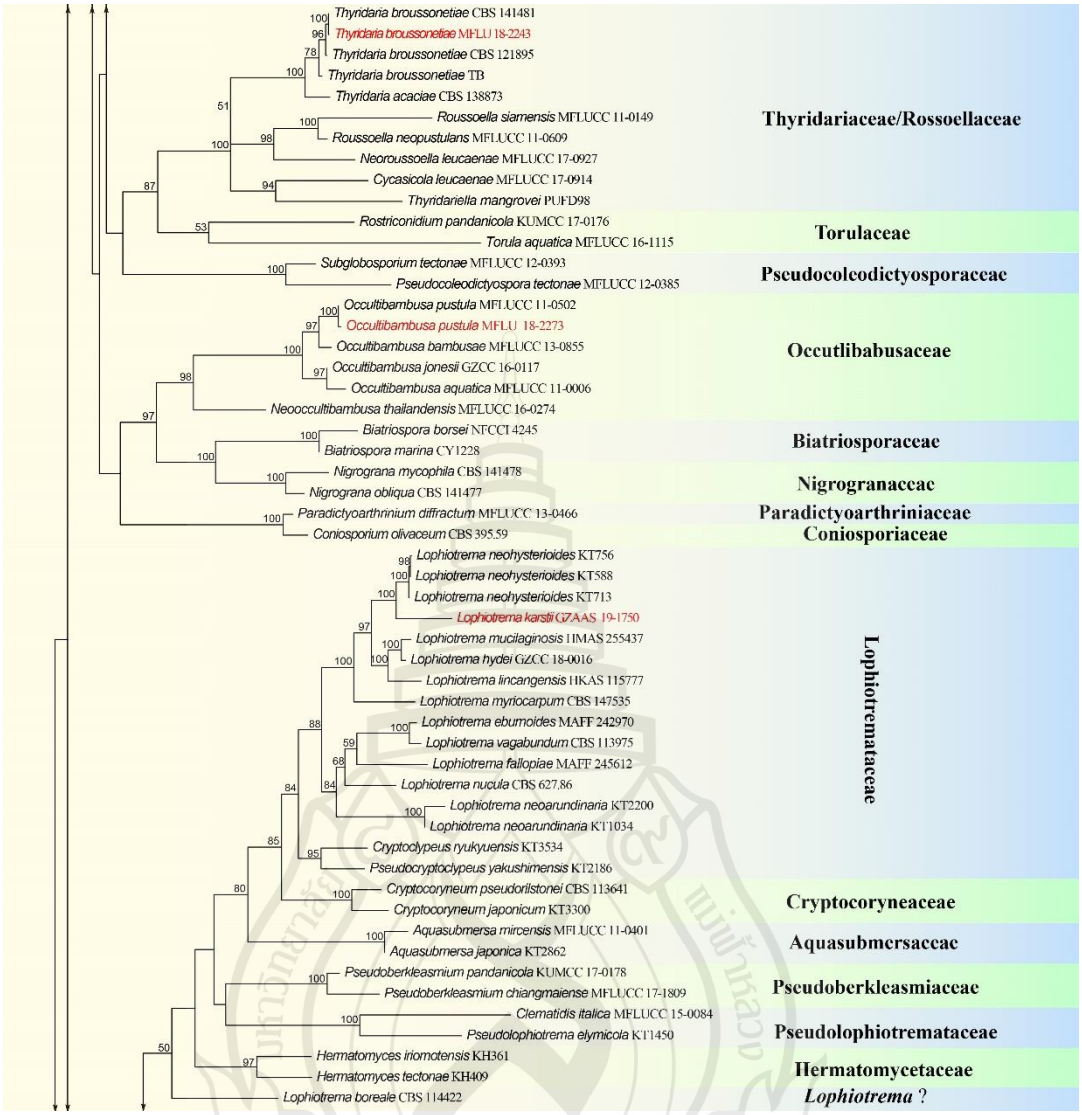


Figure 2.6 (continued)

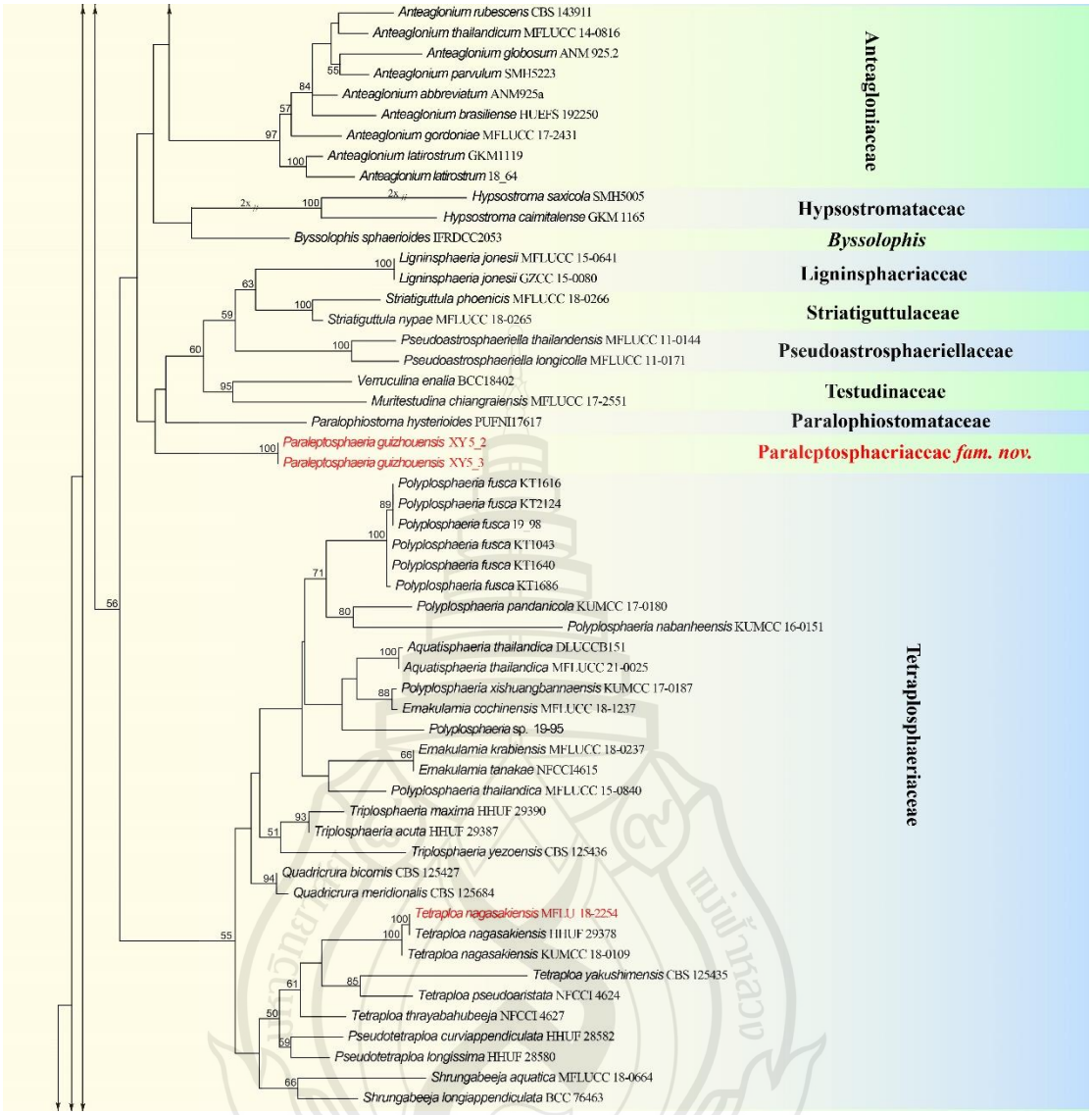


Figure 2.6 (continued)

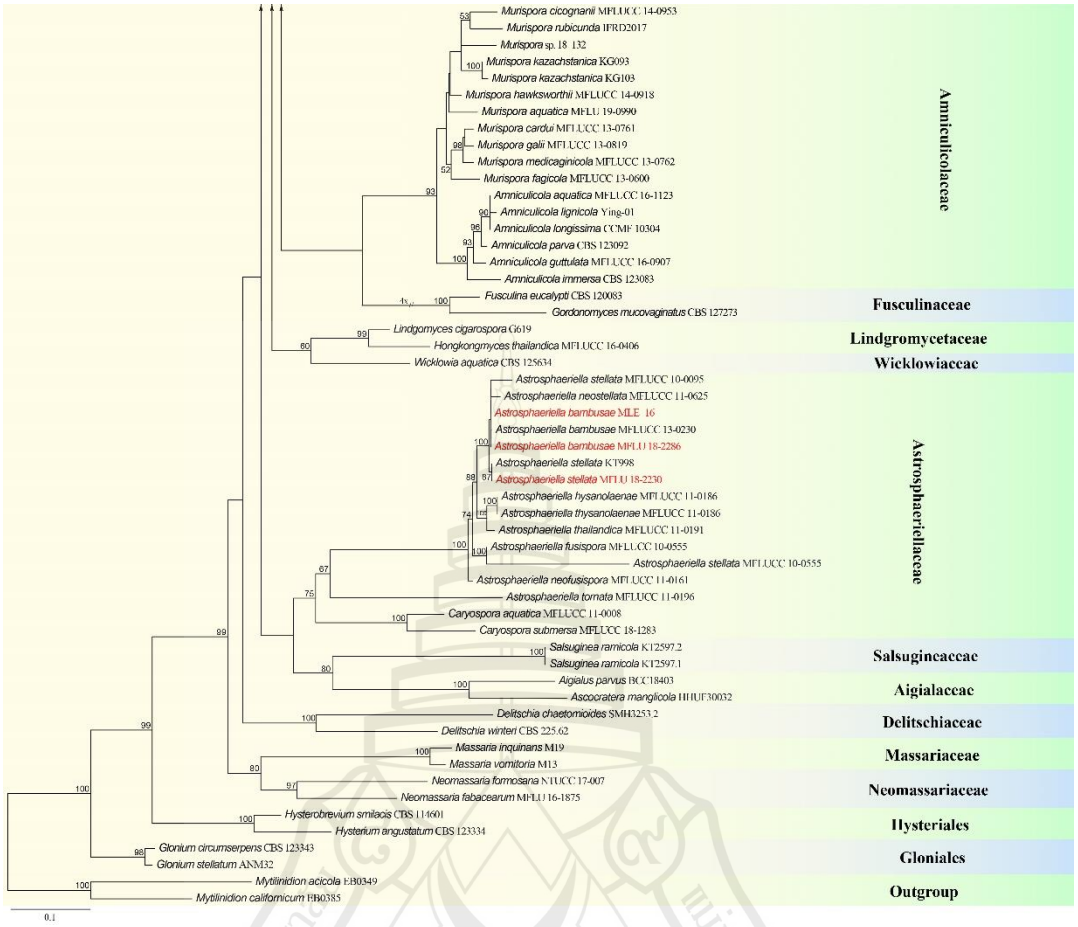


Figure 2.6 (continued)

Acrocalymmaceae Crous & Trakun

Acrocalymmaceae was introduced by Trakunyingcharoen et al. (2014) to accommodate *Acrocalymma* based on phylogenetic analysis. And the members of this family share the massarina-like morphological characteristics in sexual morph. It is a monotypic family as only the type genus was included.

Acrocalymma Alcorn & J.A.G. Irwin, Trans. Br. mycol. Soc. 88(2): 163 (1987)

Acrocalymma was introduced by Alcorn and Irwin (1987) with *A. medicaginis* as the type species. It was not until more than twenty years later that the species within this genus were updated, and these members are ecologically diverse as they were found as plant pathogens, endophytes, and saprobes from terrestrial or

freshwater as well as human superficial tissue (Alcorn & Irwin, 1987; Zhang et al., 2012; Trakunyingcharoen et al., 2014; Valenzuela-Lopez et al., 2017; Jin et al., 2018; Jayasiri et al., 2019; Dong et al., 2020; Mapook et al., 2020; Mortimer et al., 2021; Tennakoon et al., 2021). So far, eleven species were accommodated in this genus.

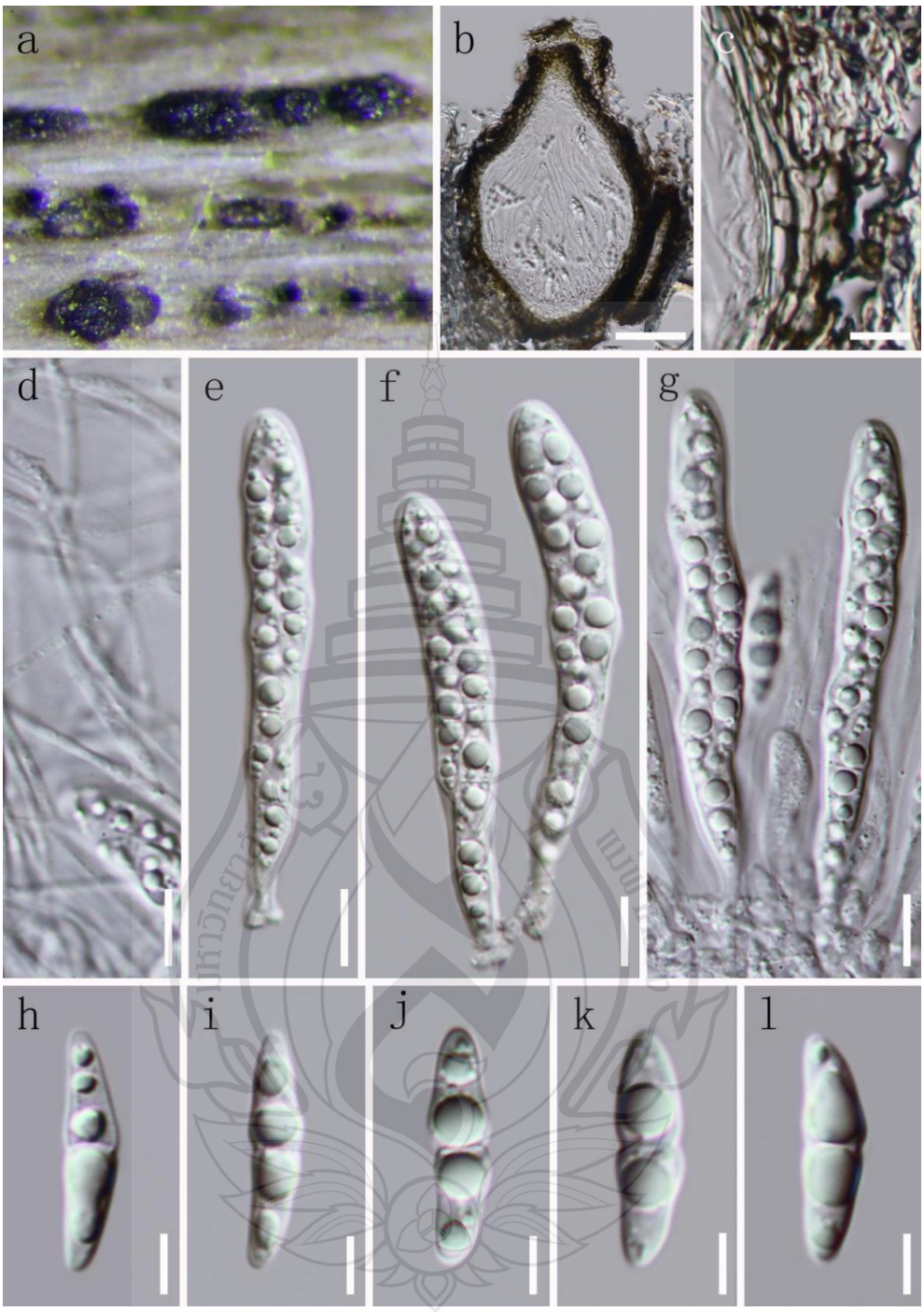
Acrocalymma medicaginis Alcorn & J.A.G. Irwin, Trans. Br. Mycol. Soc. 88(2): 163 (1987), *new record*. Figure 2.7

Saprobic on dead culm of unidentified herbaceous plant. Sexual morph: *Ascomata* 170–230 µm high × 105–140 µm diam., immersed, appearing as black spots, coriaceous, solitary or scattered, subglobose to obpyriform, brown to dark brown, ostiolate. *Ostiole* central, 39–55 µm high × 38–55 µm wide, with paraphysis lined with in ostiolar canal. *Peridium* 22–30 µm wide, comprising 5–8 layers, pale brown to brown cells of *textura angularis*. *Hamathecium* comprising numerous, 1.5–2 µm wide, filiform, trabeculate, branching pseudoparaphyses. *Asci* (66–)73–79.5(–85) × 8.5–11 µm ($\bar{x}$ = 75 × 9.5 µm, n = 25), 8-spored, bitunicate, fssitunicate, cylindric-clavate, straight or slightly curved, with a short pedicel, apically rounded, with an ocular chamber. *Ascospores* 18–21 × 4–6 µm ($\bar{x}$ = 20 × 5 µm, n = 30), overlapping, 1–2-seriate, hyaline, oblong to broadly fusiform with obtuse to slightly obtuse ends, uniseptate, straight or slightly curved, smooth, constricted at the septum, surrounded by hyaline gelatinous sheath observed clearly when mounted in Indian ink. Asexual morph: Undetermined.

Material examined: CHINA, Guizhou Province, Danzhai County, on dead culm of unidentified herbaceous plant, 29 July 2017, J.F. Zhang DZ-06 (MFLU 18-2258).

Notes: Current phylogenetic analysis (Figure 2.6) shows that our newly collected specimen (MFLU 18-2258) clusters with two strains (CPC24340 and MFLUCC 17-1423) of *A. medicaginis* with strong statistical support. The recommendations of species delineation from Jeewon and Hyde (2016) were followed, and the comparison of each gene region between these three strains was processed, and the results showed that they are identical with 100% similarity. Additionally, the morphological traits of this new specimen also match with the sexual morph of *A. medicaginis* that described by Mapook et al. (2020a), and it is therefore identified as *A. medicaginis* based on the above analysis. *Acrocalymma medicaginis* has only been

found in Australia and Thailand in previous studies (Alcorn & Irwin, 1987; Trakunyingcharoen et al., 2014; Mapook et al., 2020), hence, we hereby report it as a new geographical record in China.



Note a Appearance of ascomata on substrate. b Vertical section through ascoma. c Section of peridium. d Pseudoparaphyses. e–g asci. h–l Ascospores. Scale bars: b = 50 µm, c–g = 10 µm, h–l = 5 µm.

Figure 2.7 *Acrocalymma medicaginis* (MFLU 18-2258, new record)

Astrosphaeriellaceae Phook. & K.D. Hyde

Astrosphaeriellaceae was esablished to accommodate two genera *Astrosphaeriella* and *Pteridiospora* with *Astrosphaeriella* as the type genus, based on morphological and molecular studies (Phookamsak et al., 2015b). In their study, the genus *Astrosphaeriellopsis* was also introduced but not included in Astrosphaeriellaceae. However, Wanasinghe et al. (2018) assigned *Astrosphaeriellopsis* within Astrosphaeriellaceae on the basis of multi-gene analysis, and accepted four genera *viz*. *Astrosphaeriella*, *Astrosphaeriellopsis*, *Pithomyces* and *Pteridiospora* within this family. Subsequently, other four genera were comprised of Astrosphaeriellaceae (Wijayawardene et al., 2018; Jayasiri et al., 2019). And the confusion of placement between Astrosphaeriellaceae and Caryosporaceae was pointed out and suggested to resolve in further study (Jayasiri et al., 2019).

Astrosphaeriella Syd. & P. Syd., Annls mycol. 11(3): 260 (1913)

Astrosphaeriella was introduced by Sydow and Sydow (1913) with *A. fusispora* as the type species. Then, this genus has been morphologically well studied but lacking of the phylogenetic investigation (Hawksworth, 1981; Hawksworth & Boise, 1985; Hyde, 1992, 1994a, b, 1998; Aptroot, 1995a, b; Fröhlich & Hyde, 1995; Hyde & Fröhlich, 1998; Zhou et al., 2003; Chen & Hsieh, 2004). Tanaka et al. (2009) first included the strains of *Astrosphaeriella* species in their molecular analysis, and indicated that it is not monophyletic and removed it from Melanommataceae. Subsequently, Liu et al. (2011) re-circumscribed this genus based on morphology and phylogeny and confirmed *Astrosphaeriella* is polyphyletic, as well as suggested that a suitable family should be established to accommodate it. Phookamsak et al. (2015b) provided a natural classification of astrosphaeriella-like species and placed *Astrosphaeriella* in the family Astrosphaeriallaceae that introduced in their study. Currently, more than 60 epithets were listed in Index Fungorum (2022), while only about 10 species were accepted in *Astrosphaeriella* by Wijayawardene et al. (2022).

Astrosphaeriella bambusae Phook. & K.D. Hyde, Fungal Diversity 74: 165 (2015), *new record*. Figure 2.8

Saprobic on dead stem of bamboo. Sexual morph: *Ascostromata* erumpent to superficial, dark opaque, solitary to gregarious, mammiform to conical, with ruptured, reflexed, stellate, host remnants, around the base, uni-loculate, glabrous, brittle,

carbonaceous, with indistinct ostioles. *Peridium* 53–75 μm in sides, of unequal thickness, poorly developed at the base, composed of thick, dark brown to black cells, arranged in a *textura angularis* to *textura prismatica*, with palisade-like cells at rim. *Hamathecium* composed of dense, 1–1.5 μm wide, filiform, branching pseudoparaphyses, anastomosing at the apex, embedded in a hyaline gelatinous matrix. *Asci* (150–)177–188(–198) × 9–12 μm ($\bar{x}$ = 183 × 11 μm, n = 20), 8-spored, bitunicate, fissitunicate, cylindrical, with furcate pedicel, apically rounded, with narrow or wedge-shaped ocular chamber. *Ascospores* (35.5–)41–47.5(–50.5) × 7–8.5 μm ($\bar{x}$ = 44 × 7.5 μm, n = 25), overlapping bi-seriate, pale yellowish or pale brown, fusiform with acute ends, 1-septate, constricted at the septum, smooth-walled with a thin, distinct, mucilaginous sheath. Asexual morph: Undetermined.

Material examined: CHINA, Guizhou Province, Libo County, Maolan Town, Maolan National Nature Reserve, on dead clum of bamboo, 20 July 2016, H.Y. Ran MLCC-14 (MFLU 18-2286); Libo County, Maolan Town, Maolan National Nature Reserve, on dead stem of bamboo, 9 July 2015, J.F. Zhang MLE-16 (GZAAS 19-4016).

Notes: The newly collected specimens are phylogenetically placed within in *Astrosphaeriella* and related to *A. bambusae* (MFLUCC 13-0230 and MFLUCC 10-0095) and *A. neostellata* (MFLUCC 11-0625), but without a strong statistical support (Figure 2.6). Each gene region between these taxa is compared follow the recommendations for delimitation species from Jeewon and Hyde (2016), and the results showed that there are 100% similarity in LSU and TEF1-α between our specimens and *A. bambusae*. Additionally, the morphological characteristics of the new specimens were observed and found that they are well fits the sexual morph of *A. bambusae* that described by Phookamsak et al. (2015b), including the measurements of asci and ascospores. On the basis of the above evidence, these newly collected specimens are identified as *A. bambusae* and reported as a new geographical record for China as it has only been found in Thailand (Phookamsak et al., 2015b).



Note a, b Appearance of ascomata on substrate. c Pseudoparaphases anastomosing between the asci. d, e Asci. f–h Ascospores. Scale bars: c–e = 50 µm, f–h = 20 µm.

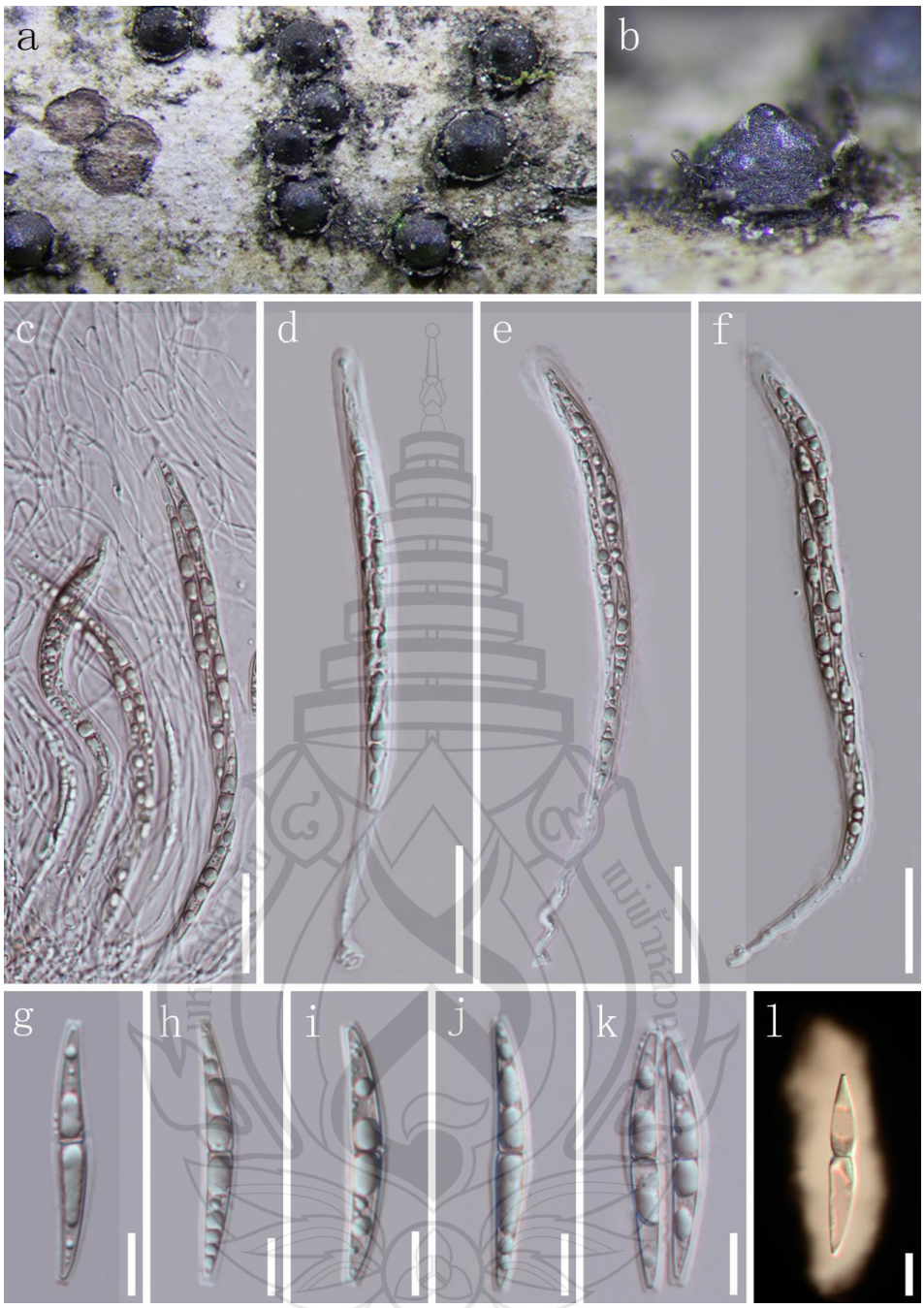
Figure 2.8 *Astrosphaeriella bambusae* (MFLU 19-2286, new record)

Astrosphaeriella stellata (Pat.) Sacc., Syll. fung. (Abellini) 24(2): 938 (1928) Figure 2.9

Saprobic on dead stems of bamboo. Sexual morph: *Ascosmata* dark opaque, solitary to gregarious, erumpent to superficial, mammiform to conical, with ruptured, reflexed, stellate, host remnants, around the base, uni-loculate, glabrous, brittle, carbonaceous, with indistinct ostioles. *Peridium* of unequal thickness, poorly developed at the base, composed of thick, dark brown to black cells, arranged in a *textura angularis* to *prismatica*, with palisade-like cells at rim. *Hamathecium* composed of dense, 1–1.8 μm wide, filiform, trabeculate pseudoparaphyses, anastomosing at the apex, embedded in a hyaline gelatinous matrix. *Asci* (164.5–)181–192 × 9–11.5 μm ($\bar{x}$ = 182 × 10.5 μm, n = 20), 8-spored, bitunicate, fissitunicate, cylindrical, with furcate pedicel, apically rounded, with narrow or wedge-shaped ocular chamber. *Ascospores* 37–43.5 × 4–5 μm ($\bar{x}$ = 40 × 4.5 μm, n = 25), overlapping bi-seriate, pale yellowish or pale brown, fusiform with acute ends, 1-septate, constricted at the septum, smooth-walled with a thin, distinct, mucilaginous sheath. Asexual morph: Undetermined.

Material examined: CHINA, Guizhou Province, Meitan County, on dead culm of bamboo, 28 August 2017, J.F. Zhang MT-2 (MFLU 18-2030).

Notes: The newly collected specimen (MFLU 18-2030) clusters together with *A. stellata* (MAFF 239487) with full bootstrap support in the present multi-gene phylogenetic analysis (Figure 2.6), and this topology is somewhat similar with Phookamsak et al. (2015b). Morphologically, they are also resemble including the measurements of pseudoparaphyses, asci and ascospores based on the description in Phookamsak et al. (2015b). Thus, we identify it as *A. stellata*, and this taxon has been known distributed in some temperate and tropical regions (Hawksworth, 1981; Hyde & Fröhlich, 1998; Chen & Hsieh, 2004; Liu et al., 2011; Phookamsak et al., 2015b).



Note a, b Appearance of ascomata on substrate. c Pseudoparaphases anastomosing between the asci. d–f Asci in different maturation stage. g–l Ascospores. Note: l is stained with India ink. Scale bars: c–f = 30 µm, g–l = 10 µm.

Figure 2.9 *Astrosphaeriella stellata* (MFLU 18-2030)

Bambusicolaceae D.Q. Dai & K.D. Hyde

Bambusicolaceae was established by Hyde et al. (2013) to accommodate the type genus *Bambusicola.* Subsequently, four genera (*Leucaenicola, Longipedicellata, Neobambusicola* and *Palmiasicola*) were introduced and comprised of this family by various authors (Crous et al., 2014; Liu et al., 2015; Zhang et al., 2016; Jayasiri et al., 2019). However, *Longipedicellata* and *Neobambusicola* were transferred to Longipedicellataceae and Sulcatisporaceae, respectively, based on morphological and molecular evidence (Tanaka et al., 2015; Phukhamsakda et al., 2016). And this treatment was followed by Hongsanan et al. (2020a) and Wijayawardene et al. (2020). Recently, Wijesinghe et al. (2020) reported a new genus *Corylicola* from Italy, and accommodated four genera *viz. Bamusicola*, *Corylicola*, *Leucaenicola* and *Palmiasicola* in Bambusicolaceae.

Corylicola Wijesinghe, Camporesi, Yong Wang bis & K.D. Hyde, Biodiversity Data Journal 8(e55957): 7 (2020)

Corylicola was introduced by Wijesinghe et al. (2020) to accommodate the type species *C. italica*. *Corylicola* has similar morphological characteristics with other genera in Bamusicolaceae by having globose to subglobose, ostiolate ascomata, cylindrical asci and one-septate ascospores. However, it is easily differentiated from these genera based on its independently phylogenetic placement and yellowish-brown ascospores. Until now, only the type species was included in this genus in Index Fungorum (2022).

Corylicola hydei J.F. Zhang & Z.Y. Liu *sp. Nov*., Figure 2.10

Etymology: "*hydei"* named for in honour of Prof. Kevin D. Hyde for his contribution to ascomycetes taxonomy.

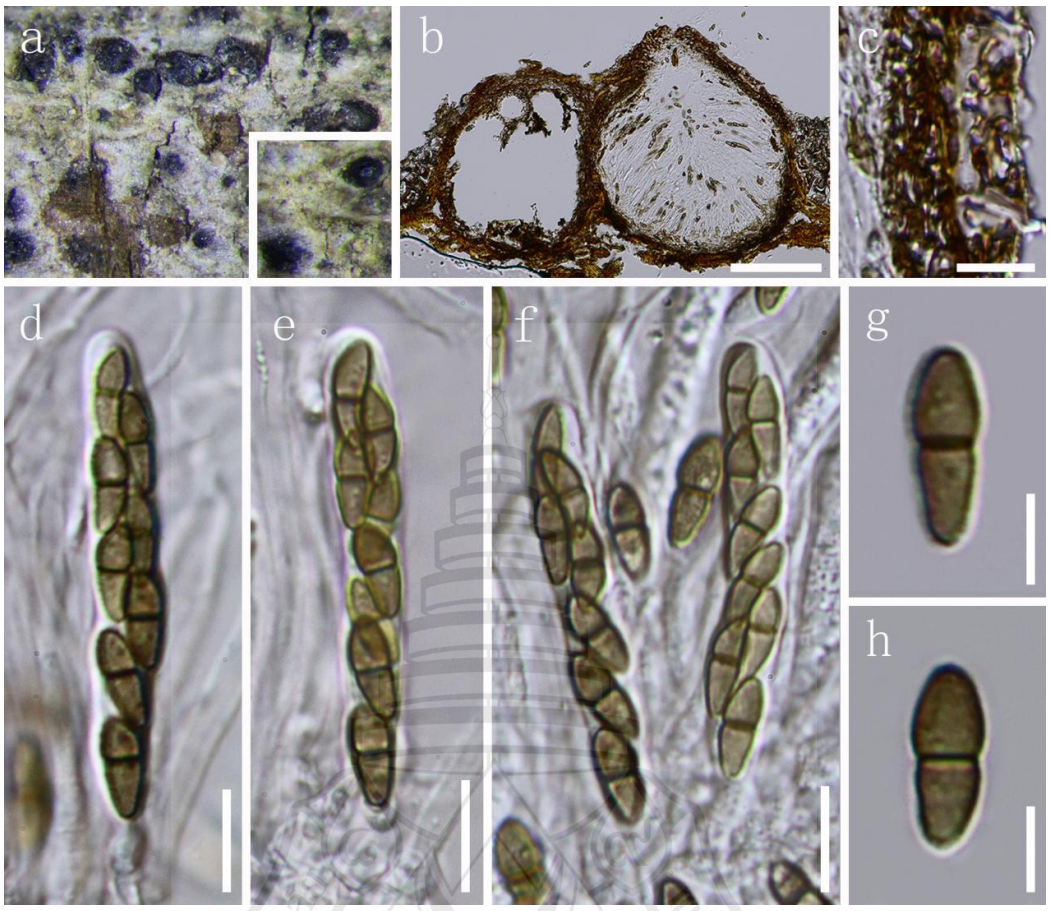
Holotype: MFLU 18-2286.

Saprobic on dead branches of woody plant, visible as raising, black dots in host surface. Sexual morph: *Ascomata* 195–250 μm high, 162–232 μm diam., gregarious, scattered to clusterred, semi-immersed to erumpent throughout the host tissue, uni-loculate, conical to subglobose, glabrous, ostiole central, with minute papilla. *Peridium* 15–25 μm thick, composed several layers thick-walled and brwon cells outwardly, and become thin-walled and hyaline towards inwardly, organized in a *textura angularis*, and the outermost layer consisted of brown to dark brown,

pseudoparenchymatous cells, confusing with host tissue. *Hamathecium* comprising 1–1.8 μm wide, dense, branched, trabeculate pseudoparaphyses, anastomosing between and above the asci, embedded in a gelatinous matrix. *Asci* 47–51.5 (–59) × 5–7 μm ($\bar{x}$ = 50.5 × 6 μm, n = 25), 8-spored, bitunicate, fissitunicate, cylindric-clavate, short pedicellate, apically rounded, with a well-developed ocular chamber. *Ascospores* 8–11.5 × 3–4.5 μm ($\bar{x}$ = 10.3 × 3.7 μm, n = 35), overlapping uni to bi-seriate, ellipsoidal to fusiform, pale brown, didymosporous, obviously constricted at the septum, rounded at the apices, upper cell is wider than the lower cell, occasionally guttulate. Asexual morph: Undetermined.

Material examined: CHINA, Guizhou Province, Guiyang City, Huaxi District, Guizhou Academy of Agricultural Sciences, on dead branches of woody plant, 22 September 2017, J.F. Zhang GZAAS-2017-7 (MFLU 18-2286, holotype); ibid. GZAAS-2017-2 (GZAAS 21-2001).

Notes: *Corylicola hydei* sisters to the type species of *Corylicola* (*C. italica*) with strong statistical support (Figure 2.6), and it is also typical of *Corylicola* that described in Wijesinghe et al. (2020). However, *C. hydei* differs from *C. italica* in having comparatively smaller asci (47–51.5 (–59) × 5–7 μm vs. 55–65 × 6–8 μm) and ascospores (8–11.5 × 3–4.5 μm vs. 10–15 × 3–4 μm). And the ascospores of *C. italica* are echinulate, while this was not observed in *C. hydei* (Wijesinghe et al., 2020).



Note a Appearance of ascomata in the host surface. b Vertical section through Ascomata. c Section of Peridium. d-f Asci with pseudoparaphyses. g, h Ascospores. Scale bars: b = 100 µm, c–f = 10 µm, g–h = 5 µm.

Figure 2.10 *Corylicola hydei* (MFLU 18-2286, holotype)

Dictyosporiaceae Boonmee & K.D. Hyde

Dictyosporiaceae was mentioned in Liu et al. (2015) and Tanaka et al. (2015), but formally established by Boonmee et al. (2016) to accommodate 10 genera with *Dictyosporium* as the typus. Subsequently, several genera were included in this family by later studies (Li et al., 2017; Iturrieta-González et al., 2018; Wijayawardene et al., 2018, 2020; Crous et al., 2019b; Hongsanan et al., 2020a; Atienza et al., 2021; Rajeshkumar et al., 2021). And currently, 17 genera were comprised of Dictyosporiaceae, which are *Aquadictyospora*, *Aquaticheirospora*, *Cheirosporium*, *Dendryphiella*, *Dictyocheirospora*, *Dictyopalmispora*, *Dictyosporium*, *Digitodesmium*,

Gregarithecium, *Jalapriya*, *Neodendryphiella*, *Paradictyocheirospora*, *Pseudocoleophoma*, *Pseudoconiothyrium*, *Pseudodictyosporium*, *Verrucoccum* and *Vikalpa* (Wijayawardene et al., 2022).

Dictyosporium Corda, Weitenweber's Beitr. Nat.: 87 (1836)

Dictyosporium was established and typified with *D. elegans*. The genus is characterized by subglobose superficial ascomata, bitunicate cylindrical asci and hyaline, fusiform uniseptate ascospores with or without a sheath in sexual morph; micronematous to macronematous conidiophores and cheiroid, digitate complanate conidia with several parallel rows of cells in asexual morph (Boonmee et al., 2016; Yang et al., 2018a). Members of this genus are distributed worldwide on dead wood and plant litter in terrestrial and aquatic habitats (Hyde & Goh, 1998; Pinnoi et al., 2006; Pinruan et al., 2007; Yang et al., 2018).

Dictyosporium karstium J.F. Zhang & K.D. Hyde, *sp. nov.,* Figure 2.11

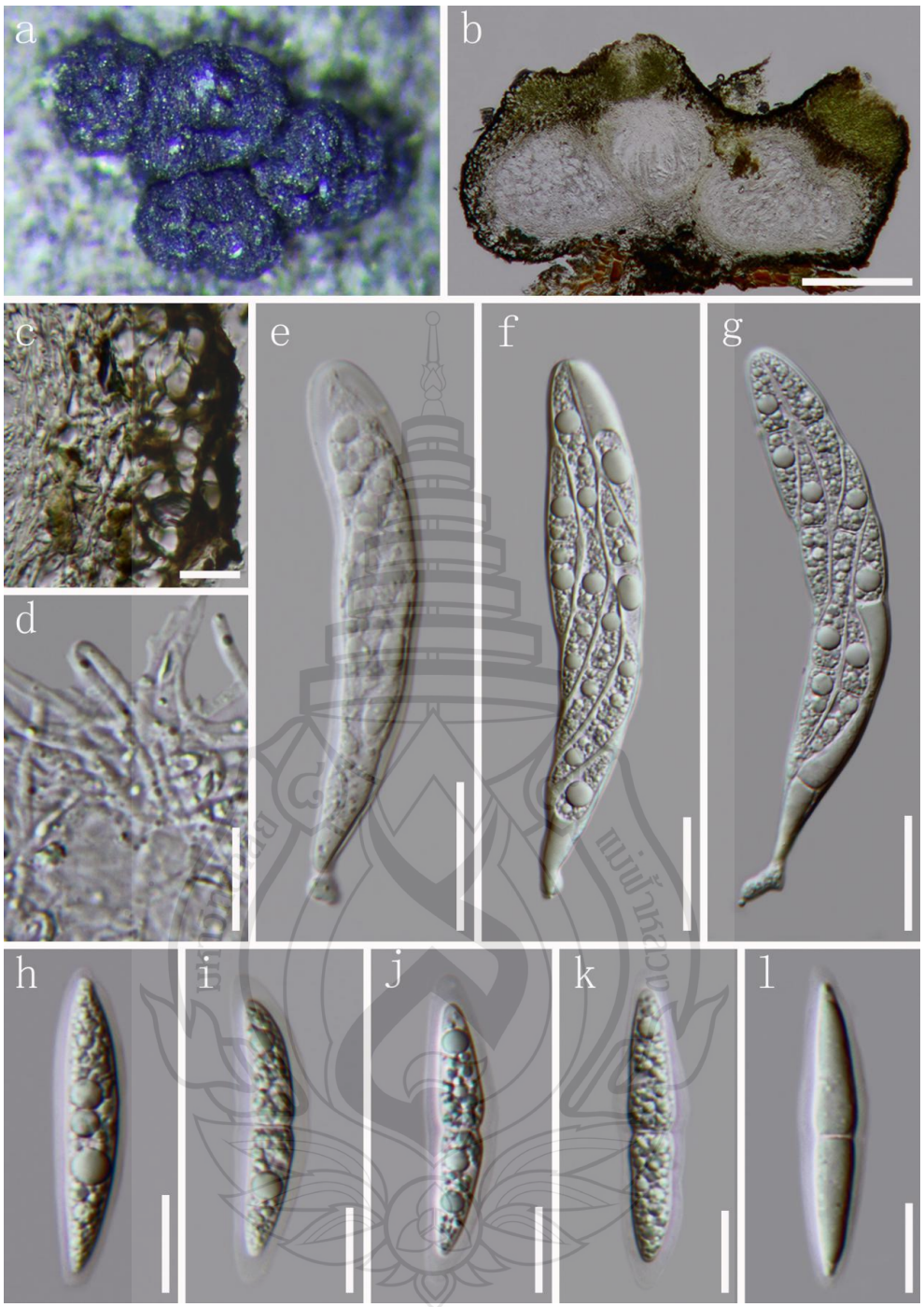
Etymology: "*karstium"* named refer to the holotype was collected from karst formations.

Holotype: MFLU 18-2282.

Saprobic on dead branch of woody plant. Sexual morph: *Ascomata* superficial, black, wrinkled, normally groped with 3 or 4, rarely scattered, globose to subglobose, leathered. *Peridium* laterally 65–85 μm wide, composed of three strata, with the innermost stratum comprising several layers of hyaline, thin-walled cells of *texura angularius*, an internal stratum of several layers of thick-walled, brown cells of *texura angularius*, and an outermost layer of black, thick-walled cells, and also production several layers of light green cells around the apex. *Hamathecium* comprising 1.5–2 μm wide, filamentous, trabeculate pseudoparaphyses, anastomosing between and above the asci, embedded in a gelatinous matrix. *Asci* (90.5–) 95–101 (–106.5) × 12–15 (–16.5) μm ($\overline{x}$ = 97 × 13.8 μm, n=20), 8-spored, bitunicate, fissitunicate, broadly cylindric-clavate, with a short, furcate pedicel, apically rounded, ocular chamber distinct. *Ascospores* 27–33 (–36.5) × 4–5.5 (–6.5) μm ($\overline{x}$ = 30.8×5 μm, n=30), overlapping bi-seriate to tri-seriate, hyaline, guttulate, verrucose, fusiform, aseptate when young, and become two-celled at maturity, constricted at the septum, the upper cell slightly wider, surrounded by a entire gelatinous sheath, extend to 1 μm at both ends. Asexual morph: Undetermined.

Material examined: CHINA, Guizhou Province, Libo County, Maolan Town, Maolan National Nature Reserve, on dead branch of unidentified woody plant, 20 July 2016, H.Y. Ran, MLCC-9 (MFLU 18-2282).

Notes: *Dictyosporium karstium* is phylogenetically placed within in the mid of *Dictyosporium*, but without a high support (Figure 2.13). There are only two sexually reproduction species (*D. meiosporum* and *D. sexualis*) have been reported amongst all dictyosporium-like genera (Liu et al., 2015; Boonmee et al., 2016), and *D. karstium* is the third species of this genus that observed with sexual state from nature. These three taxa share similar morphological characteristics of superficial ascomata, cylindric-clavate asci and hyaline, one-septate ascospores that surrounded by a mucilaginous sheath. However, they are distinguished from each other by the different measurements of asci (*D. karstium*, (90.5–) 95–101 (–106.5) × 12–15 (–16.5) μm vs. *D. meiosporum*, 83–135.5 × 13–17 μm vs. *D. sexualis*, 100–145 × 10–14 μm) and ascospores (*D. karstium*, 27–33 (–36.5) × 4–5.5 (–6.5) μm vs. *D. meiosporum*, 31–39 × 6–8.5 μm vs. *D. sexualis*, 36–48 × 6–8 μm). Moreover, the ascospores of *D. meiosporum* are surrounded by a thin gelatinous sheath, while it is obviously found that the ascospores of other two taxa are surrounded by a thick gelatinous sheath. Additionally, both *D. meiosporum* and *D. sexualis* were collected from the tropics, while *D. karstium* was found in the subtropics (Liu et al., 2015; Boonmee et al., 2016).



Note a Appearance of ascomata in host surface. b Vertical section through ascomata. c Section of peridium. d Pseudoparaphyses. e–g Asci. h–l Ascospores. Scale bars: b = 300 μm, c, d, h–l = 10 μm, e–g = 20 μm.

Figure 2.11 *Dictyosporium karstium* (MFLU 18-2282, holotype)

Pseudocoleophoma Kaz. Tanaka & K. Hiray., Stud. Mycol. 82: 89 (2015)

Pseudocoleophoma was established by Tanaka et al. (2015) to accommodate *P. calamagrostidis* and *P. polygonicola* with the former as the type species. This genus is characterized by immersed to erumpent, globose to subglobose, ostiolate ascomata, cylindrical to clavate, fissitunicate asci and fusiform, two-celled ascospores with a conspicuous mucilaginous sheath (Tanaka et al., 2015). Recently, other 6 species were added to *Pseudocoleophoma* (Hyde et al., 2016; Jayasiri et al., 2019; Tennakoon et al., 2019; Li et al., 2020b; Phukhamsakda et al., 2020), and a novel species *Pseudocoleophoma guizhouensis* is identified and introduced in this study, based on morphological and phylogenetic analysis. In addition, the genus *Verrucoccum* is nested within in *Pseudocoleophoma* with high bootstrap support, while the difference in morphological characteristics between these two genera are quite obvious (Atienza et al., 2021). Therefore, the nature placement of *Verrucoccum* is expected to clarify in the further study.

Pseudocoleophoma guizhouensis J.F. Zhang & Z.Y. Liu, *sp. nov*., Figure 2.12

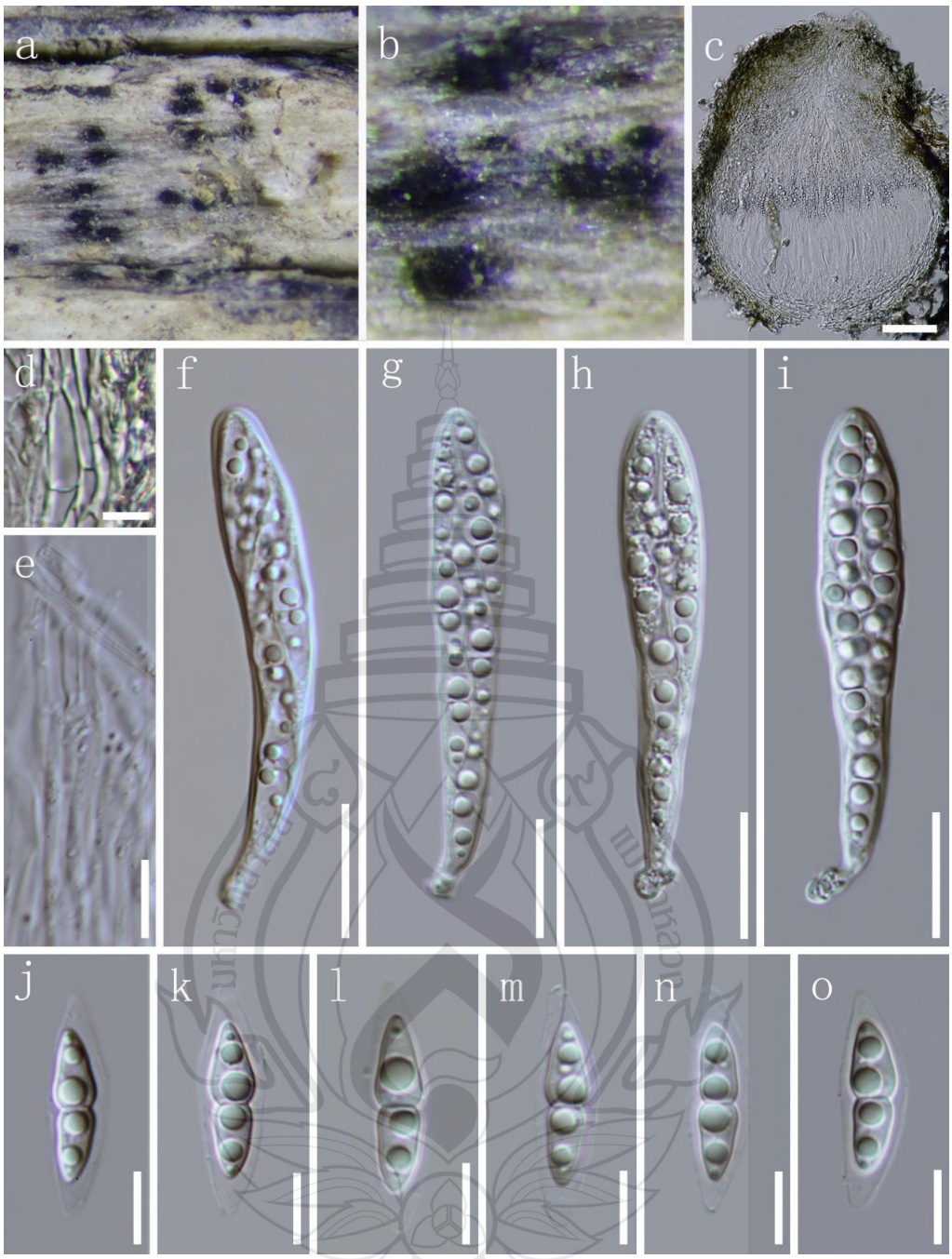
Etymology: named refers to where the holotype was collected.

Holotype: MFLU 18-2262.

Saprobic on decaying woody plant, visible as black dots in host surface. Sexual morph: *Ascomata* immersed to erumpent through the substrate, gregarious, scattered, coriaceous, subglobose to globose, ostiolate. *Peridium* laterally 18–25 μm wide, composed of two strata, with the inner stratum of several layers of hyaline, thin-walled cells of *texura angularius*, and outer stratum of yellowish to brown pseudoparenchymatous cells confusing with host tissue, and with thick-walled, brown cells of *textura angularius* around the ostiole. *Hamathecium* consisting of dense, 1.5–3 μm wide, filamentous, septate pseudoparaphyses, anastomosing between and above the asci. *Asci* (74–) 77–88 (–92) × 10.5–13 (–14) μm ($\overline{x}$ = 81.3 ×12 μm, n = 20), 8-spored, bitunicate, fissitunicate, clavate, with a short, simple or furcate pedicel, apically rounded, with a developed ocular chamber. *Ascospores* 19.5–23 (–25) × 5–6.5 μm ($\overline{x}$ = 21×5.8 μm, n = 35), overlapping bi-seriate to tri-seriate, hyaline, guttulate, fusiform, narrowly towards the ends, two-celled, distinctly constricted at the septum, surrounded by an entire mucilaginous sheath, 1–1.5 μm wide at sides, and extension to 3–5 μm long at both ends. Asexual morph: Undetermined.

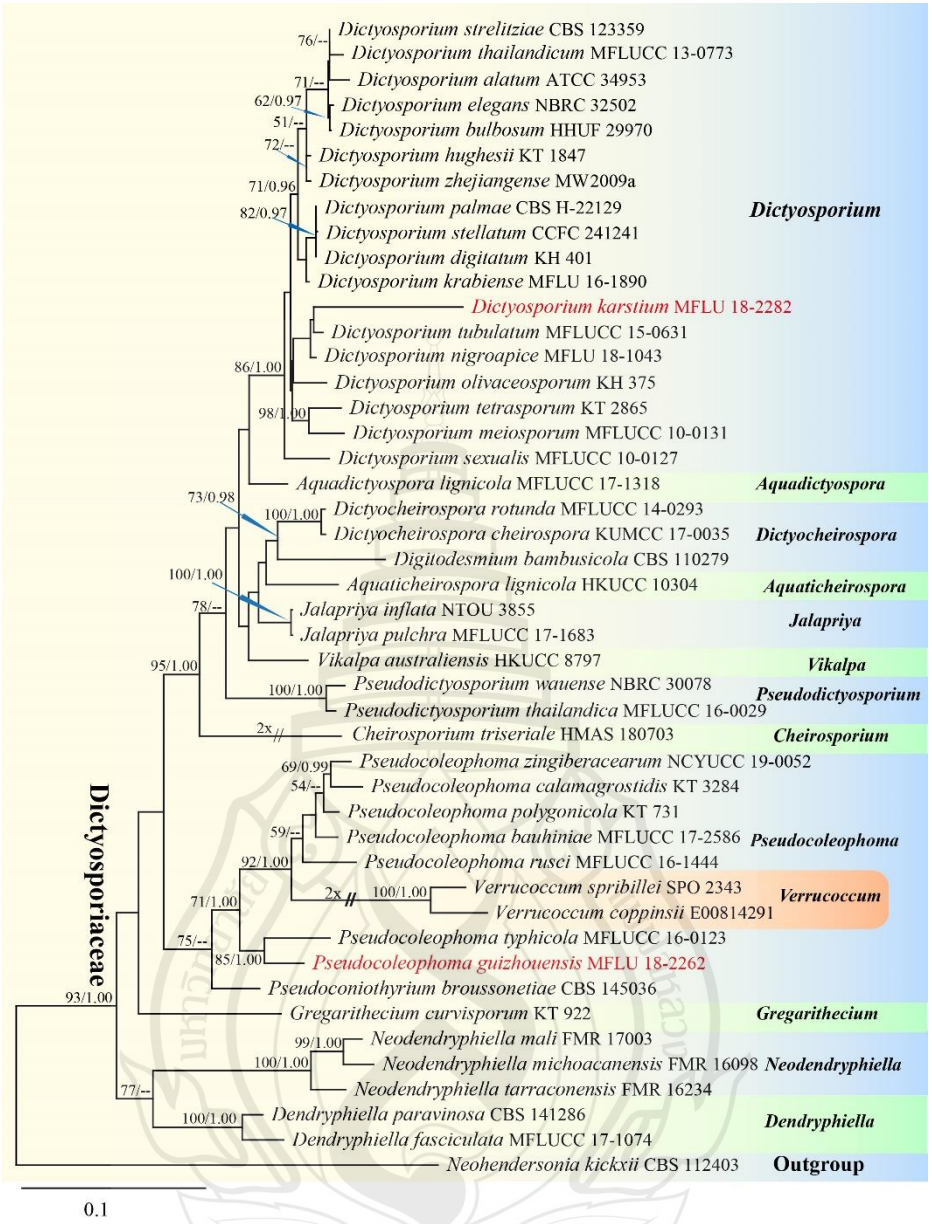
Material examined: CHINA, Guizhou Province, Huishui County, on unidentified decaying woody plants, 21 July 2017, J.F. Zhang HS-09 (MFLU 18-2262, holotype).

Notes: Present phylogenetic analysis shows that *Pseudocoleophoma guizhouensis* sisters to *P. typhicola* with well bootstrap support (Figure 2.13). Morphologically, *P. guizhouensis* is typical of *Pseudocoleophoma* by having globose to subglobose, ostiolate ascomata, cylindrical to clavate, short pedicellate asci that bearing fusiform, one septate ascospores surround by a conspicuous mucilaginous sheath (Tanaka et al., 2015). Because of its closed relative species *P. typhicola* was introduced as asexual morph, while our new specimen is observed as sexual morph. Therefore, each gene region between these two taxa is compared based on the recommendations of delimitation of species from Jeewon and Hyde (2016). And the results shows that there are 11 base pair differences in LSU across 669 nucleotides, and 50 base pair differences in ITS across 549 nucleotides, respectively. And this is enough to warrant that they are separate species. There is no significant difference in measurements of asci and ascospores between *P. guizhouensis* and other reported *Pseudocoleophoma* taxa, but *P. guizhouensis* is phylogenetically distant from them in the present study.



Note a, b Appearance of ascomata in the host surface. c Vertical section through ascomata. d Section of peridium. e Pseudoparaphyses. f–i Asci. j–o Ascospores. Scale bars: c = 50 μm, d, e, j–o = 10 μm, f–i = 20 μm.

Figure 2.12 *Pseudocoleophoma guizhouensis* (MFLU 18-2262, holotype)



Note Maximum likelihood bootstrap values equal or greater than 50% and Bayesian posterior probabilities equal or greater than 0.95 are indicated near the nodes. New sequence generated in this study are given in red. The tree is generated from maximum likelihood (ML) analysis based on LSU, Rpb2, SSU and TEF1-α sequence data, and rooted with *Neohendersonia kickxii* CBS 112403.

Figure 2.13 Phylogenetic tree of the family Dictyosporiaceae

Didymosphaeriaceae Munk

Didymosphaeriaceae was introduced by Munk (1953) to accommodate the genus *Didymosphaeria* with *D. epidermidis* as the type species. Ariyawansa et al. (2014) reappraised this family and accepted sixteen genera. Then, the accounts of this family were continually updated in subsequent studies (Ariyawansa et al., 2014; Wijayawardene et al., 2014; Crous et al., 2015a, b; Wanasinghe et al., 2016; Thambugala et al., 2017; Jayasiri et al., 2019; Phookamsak et al., 2019; Mapook et al., 2020), and 33 genera were included within Didymosphaeriaceae until now (Wijayawardene et al., 2022).

Montagnula Berl., Icon. fung. (Abellini) 2(2-3): 68 (1896)

Montagnula was introduced and typified by *M. infernalis* (Berlese 1896). Then the classification placement of this genus has been changed several times until Ariyawansa et al. (2014) placed it in Didymosphaeriaceae based on morphological and phylogenetic studies. Subsequently, members of this genus have been discovered and included (Liu et al., 2015; Hongsanan et al., 2015; Wanasinghe et al., 2016; Hyde et al., 2016; Tennakoon et al., 2016b; Niranjan & Sarma, 2018; Tibpromma et al., 2018; Mapook et al., 2020). Presently, 44 species were listed in Index Fungorum (2022), with 18 of these taxa have available sequence data in GenBank.

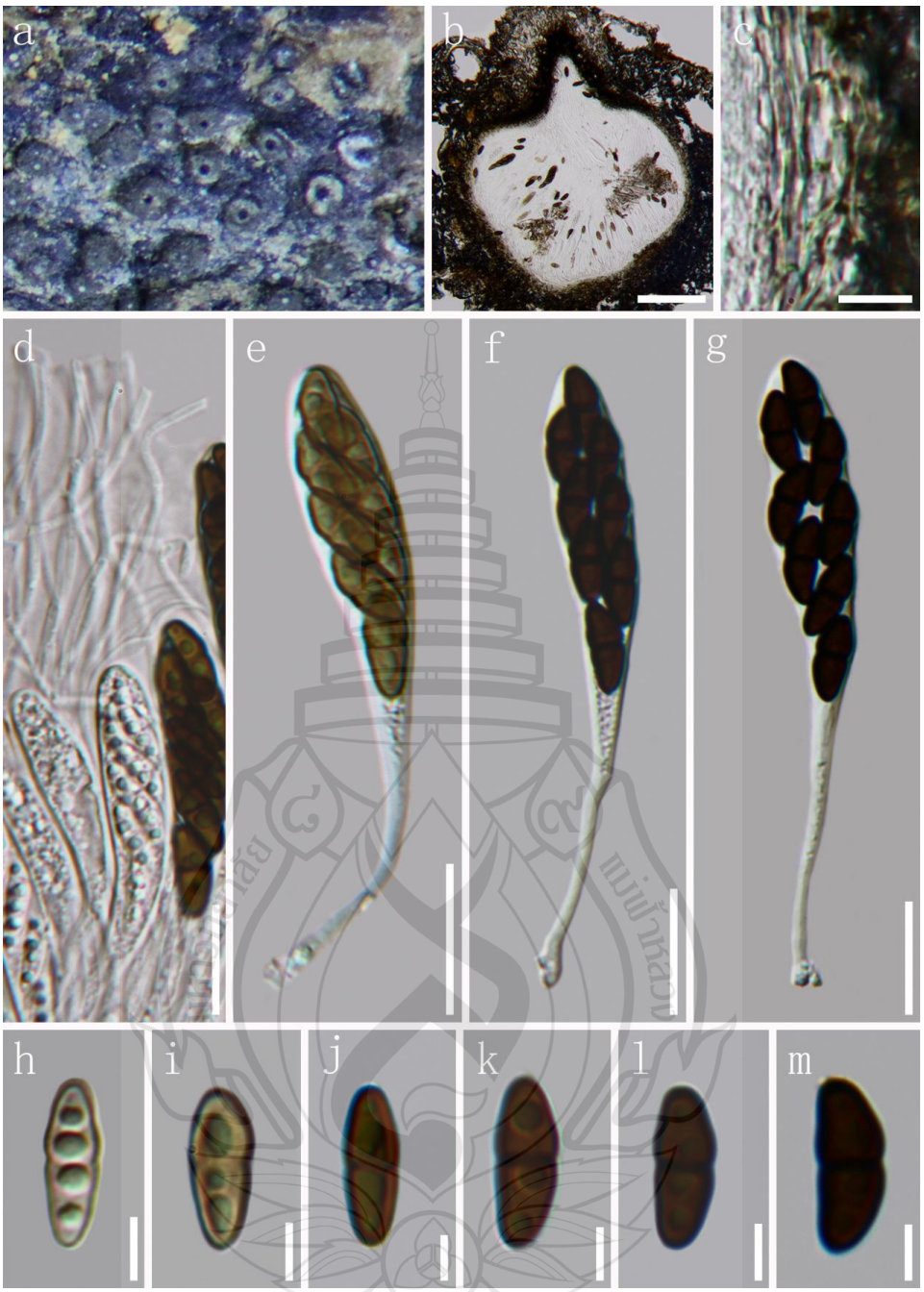
Montagnula chromolaenicola Mapook & K.D. Hyde, Fungal Divers 101: 35 (2020), *new record*, Figure 2.14

Saprobic on dead branch of unidentified woody plants. Sexual morph: *Ascomata* semi-immersed to erumpent, solitary or scattered, globose to obpyriform, coriaceous, brown to dark brown. *Ostiole* papillate, protruding from substratum. *Peridium* 25–40 μm wide, comprising several layers of thin-walled, pale brown to brown cells of *textura angularis*. *Hamathecium* comprising 1.5–4 μm wide, cylindrical to filiform, septate, branching pseudoparaphyses, swollen at the base. *Asci* (83–)86–97(–111) × 12–14 μm ($\bar{x}$ = 95 × 12.5 μm, n = 20), 8-spored, bitunicate, elongate-clavate, slightly curved, long pedicel. *Ascospores* 14–17 × 5.5–6.5 μm ($\bar{x}$ = 15 × 6 μm, n = 25), overlapping 1–2-seriate, pale brown to yellowish brown when immature, becoming brown to dark brown when mature, broadly fusiform to ellipsoid, 1-septate, constricted at the septum, slightly wider upper cell and tapering towards

ends, straight to slightly curved, without terminal appendages. Asexual morph: Undetermined.

Material examined: CHINA, Guizhou Province, Guiyang City, on dead branch of unidentified woody plants, 27 September 2016, J.F. Zhang GZ-07 (MFLU 18-2290).

Notes: The newly collected specimen (MFLU 18-2290) clusters with *M. chromolaenicola* (MFLUCC 17-1469) and *M. thailandica* (MFLUCC 17-1508) in the present molecular analysis (Figure 2.6). According to the recommendations of delimitation species from Jeewon and Hyde (2016), each gene region between these above taxa was compared, and the results showed that there are 100% similarity in LSU, SSU and ITS between our new collection and *M. chromolaenicola*. Besides, the morphological features of this specimen also match with the descriptions of *M. chromolaenicola* that introduced by Mapook et al. (2020a). Therefore, we identify this specimen as *M. chromolaenicola* and report it as a new geographical record for China (Mapook et al., 2020).



Note a Appearance of ascomata in the host surface. b Vertical section through ascoma. c Section of peridium. d Pseudoparaphyses. e–g Asci in different stage. h-m Ascospores in different maturation stage. Scale bars: b = 100 µm, c = 10 µm, d–g = 20 µm, h–m = 5 µm.

Figure 2.14 *Montagnula chromolaenicola* (MFLU 18-2290)

*Trematei*a Kohlm., Volkm.-Kohlm. & O.E. Erikss., Bot. Mar. 38(2): 165 (1995)

*Trematei*a was introduced by Kohlmeyer et al. (1995) with *T. halophila* as the type species. Until twenty years later, two species *T. arunicola* and *T. guiyangensis* were discovered and accepted in this genus based on morphological and molecular data (Hyde et al., 2016). With the latest update of accounts in *Trematei*a (Feng et al., 2019; Hyde et al., 2020a; Mapook et al., 2020), nine species were comprised of this genus.

*Trematei*a *chromolaenae* Mapook & K.D. Hyde, in Mapook et al., Fungal Diversity 101: 40 (2020), *new record*. Figure 2.15

Saprobic on dead culm of herbaceous plant. Sexual morph: *Ascomata* immersed, solitary or scattered, appearing as small dark spots, coriaceous, globose, dark brown to brown. *Ostiolar* neck protruding. *Peridium* 17–24 µm wide, comprising 4–6 layers, brown to dark brown cells of *textura angularis*. *Hamathecium* comprising 1.5–3.5 µm wide, broadly cylindrical, septate, branching pseudoparaphyses. *Asci* 77–102 × 13–15.5 µm ($\bar{x}$ = 92.5 × 14.5 µm, n = 15), 8-spored, bitunicate, fssitunicate, clavate to cylindric-clavate, straight or slightly curved, pedicellate, apically rounded. *Ascospores* (14–)17–21 × 7–8.5 µm ($\bar{x}$ = 18.5 × 7.5 µm, n = 25), overlapping, 1–2-seriate, initially hyaline, 1-septate when immature, becoming golden-brown at maturity, oval to ellipsoidal, muriform, 3–7-transversely septate, with 1–3 longitudinal septa, upper part wider, slightly constricted at the central septum, straight or slightly curved, guttulate, surrounded by hyaline gelatinous sheath observed clearly when mounted in Indian ink. Asexual morph: Undetermined.

Material examined: CHINA, Guizhou Province, Guiyang City, Yunyan District, Guizhou Botanical Garden, on dead culm of herbaceous plant, 8 August 2017, J.F. Zhang ZWY-13 (MFLU 18-2246); Huaxi District, Guizhou Academy of Agricultural Sciences, on dead culm of herbaceous plant, 23 September 2016, J.F. Zhang GZ-04 (GZAAS 21-2003); Zunyi City, on dead culm of herbaceous plant, 16 May 2019, Y.Y. Chen 20-036 (GZAAS 20-0153); Zunyi City, on dead culm of herbaceous plant, 16 May 2019, Y.Y. Chen 20-037 (GZAAS 20-0154).

Notes: Current phylogenetic analysis shows that four newly collected specimens cluster with *T. chromolaenae* and form a subclade where it is sisters to *T. guiyangensis* (Figure 2.6). Observation of the gross morphological characteristis of our new collections, we found that they are also resembles to *T. chromolaenae* including the overlap sized asci, ascospores and pseudoparaphysis (Mapook et al., 2020). Therefore, these four specimens are identified as *T. chromolaenae* and reported as a new geographical record for China.



Note a Appearance of ascomata in the host surface. b Vertical section through ascoma. c Section of peridium. d Pseudoparaphyses. e–g Asci in different maturation stage. h–l Ascospores in different maturation stage. Scale bars: b = 100 μm, c, h–l = 5 μm, d–g = 20 μm.

Figure 2.15 *Trematei chromolaenae* (MFLU 18-2246, new record)

Trematеia guiyangensis J.F. Zhang, J.K. Liu, K.D. Hyde & Z.Y. Liu, *sp. nov.*

Index Fungorum number: IF552160; Facesoffungi number: FoF02235, Figure 2.16

Etymology: Name reflects the place Guiyang, where the holotype was collected.

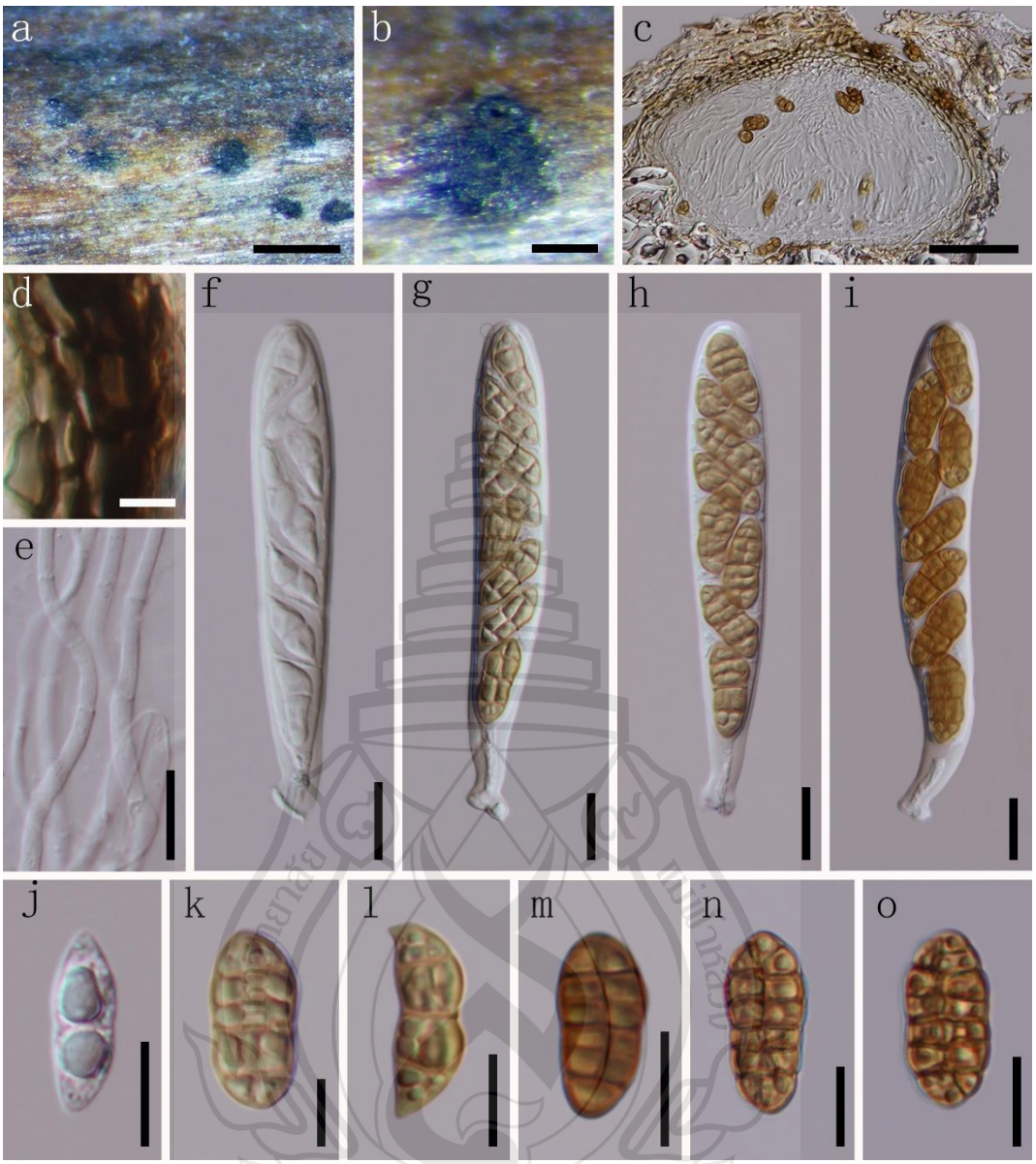
Holotype: MFLU 16-1299.

Saprobic on dead herbaceous stems. Sexual morph: *Ascomata* 130–280 μm high, 190–400 μm diam., scattered to clustered, subglobose to ovoid, with flattened base, coriaceous, immersed to semi-immersed or breaking through the host epidermis, dark brown to black, ostiolate, minutely papillate. *Peridium* up to 9–16 μm, composed of several layers of light to brown, thick-walled cells of *textura angularis*, becoming thin-walled and hyaline towards the centrum. *Hamathecium* comprising numerous, 2.3–5.1 μm wide, hypha-like, septate pseudoparaphyses, tapering towards the terminal cells, and intermingled among asci, embedded in a gelatinous matrix. *Asci* 152–160 × 21–27 μm ($\bar{x}$ = 149 × 24 μm, n = 20), 8-spored, bitunicate, fissitunicate, cylindrical-clavate, with a furcate pedicel, apically rounded, with a minute ocular chamber. *Ascospores* 20–28 × 9–12 μm ($\bar{x}$ = 25 × 11 μm, n = 30), muriform, normally overlapping 1–2-seriate, ellipsoid to broad fusiform, both parts of the spore ±equal in size, with 3–5 transverse septa, and 1 longitudinal septum in each row, obviously constricted in the central septum and slightly constricted at other septa, initially hyaline, becoming golden-brownish to brown when mature, rounded at both ends and lacking any gelatinous sheath or appendages. Asexual morph: Undetermined.

Material examined: CHINA, Guizhou, Guiyang, around Guizhou Academy of Agriculture Sciences, on dead herbaceous stems, 17 August 2015, J.F. Zhang, GZ-01 (MFLU 16-1299, holotype), ex-type living culture.

Notes: *Trematеia guiyangensis* is phylogenetically close to *T. arundicola* and *T. halophila*, and also morphologically similar in having globose to subglobose or flattened, immersed to semi-immersed ascomata, with clavate asci and muriform ascospores. However, *T. guiyangensis* was collected from terrestrial habitat, while the other two species were collected from aquatic habitats. In addition, the asci of *T. guiyangensis* are shorter than *T. arundicola* ($\bar{x}$ = 149 vs. 185 μm) and the ascospores

are smaller than *T. halophila* ($\bar{x}$ = 26 × 6.5 μm vs. 30 × 16 μm). There are more transverse and vertical septa in *T. guiyangensis* than in *T. arundicola*, and the ascospores of *T. arundicola* are paler than the other two species.



Note a, b Appearance of ascomata immersed on host surface. c Vertical section of ascoma. d Section of peridium. e Pseudoparaphyses. f–i Cylindrical-clavate asci. j–o Ascospores. Scale bars: a = 500 μm, b = 100 μm, c = 50 μm, d, j–o = 10 μm, e–i = 20 μm.

Figure 2.16 *Trematеia guiyangensis* (MFLU 16-1299, holotype)

Lentitheciaceae Y. Zhang ter et al.

Lentitheciaceae was introduced by Zhang et al. (2008b) with *Lentithecium* as the typus. In their study, they definitely accepted the genus *Katumotoa*, and demonstrated that *Keissleriella* may also be a member of this family. Subsequently, Hyde et al. (2013) tentatively assigned *Keissleriella* within Lentitheciaceae based on phylogenetic analysis, and listed 4 genera (*Katumotoa*, *Keissleriella*, *Lentithecium* and *Tingoldiago*) in this family. With the update of accounts within this family in recent studies (Wanasinghe et al., 2014; Knapp et al., 2015; Phookamsak et al., 2015a; Tanaka et al., 2015; Li et al., 2016; Dayarathne et al., 2018; Wijayawardene et al., 2018; Hyde et al., 2020a), and 14 genera are currently included in Lentitheciaceae (Hongsanan et al., 2020a; Wijayawardene et al., 2022).

Poaceascoma Phook. & K.D. Hyde, Cryptog. Mycol. 36(2): 231 (2015)

Poaceascoma was introduced by Phookamsak et al. (2015a) with *P. helicoides* as the type species, based on morphological and molecular data. Then, five species viz. *P. aquaticum*, *P. filiforme*, *P. halophium*, *P. lochii* and *P. taiwanense* were subsequently discovered and comprised of this genus in later studies (Luo et al., 2016; Hyde et al., 2017, 2018a; Crous et al., 2020a; Tan et al., 2021). Members of *Poaceascoma* are generally characterized by uni-loculate, globose to subglobose ascomata, 8-spored, bitunicate, fissitunicate, elongate-cylindrical asci, as well as fasciculate, filiform, hyaline ascospores that spirally arranged within the ascus (Phookamsak et al., 2015a). In this study, a novel species *Poaceascoma herbacum* is introduced based on its morphological characteristics and phylogenetic analysis.

Poaceascoma herbacum J.F. Zhang & K.D. Hyde, *sp. nov*., Figure 2.17

Etymology: "herbacum" named refers to the holotype associated with the herbaceous plant.

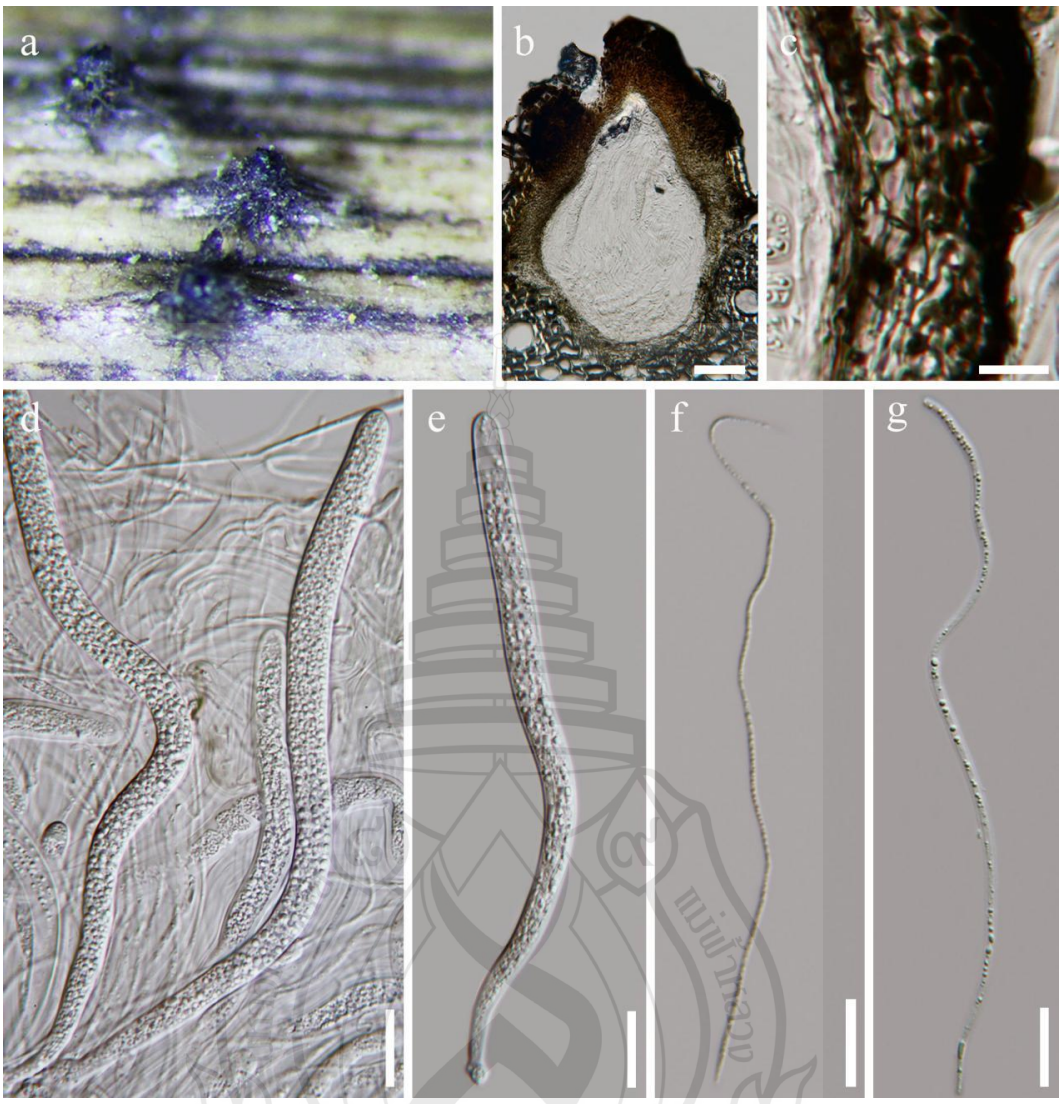
Holotype: MFLU 18-2279.

Saprobic on dead culm of unidentified herbaceous plant. Sexual morph: *Ascomata* solitary to gregarious, semi-immersed to erumpent, visible as raised, dark spots on host surface, uni-loculate, globose to subglobose, ostiolate, papillate. *Papilla* conical to cylindrical, erumpent, exposed parts covered with raised brown tufts of hyphae. *Peridium* (26–)35–84 μm wide, with equal thickness at the sides, thin at the base and become broadly near the ostiole, composed of several layers of dark brown

to black, pseudoparenchymatous cells arranged in a *textura angularis* to *textura prismatica*. *Hamathecium* composed of dense, 1.5–2.5 μm wide, trabeculate, filamentous pseudoparaphyses, anastomosing between the asci, embedded in a gelatinous matrix. *Asci* (190–)207–216(–226) × 9–11 μm ($\bar{x}$ = 210 × 10 μm, n = 25), 8-spored, bitunicate, fissitunicate, elongate-cylindrical, short pedicellate, with rounded to obtuse pedicel, apically rounded, with an indistinct ocular chamber. *Ascospores* (239–)252–298(–328) × 2–3 μm ($\bar{x}$ = 282 × 2.5 μm, n = 30), spirally arranged in the ascus, filiform, wider in the upper part, narrow towards the ends, hyaline, multi-septate, but not constricted at the septa, guttulate, smooth-walled. Asexual morph: Undetermined.

Material examined: CHINA, Guizhou Province, Libo County, Maolan Town, Maolan National Nature Reserve, on dead culm of unidentified herbaceous plant, 20 July 2016, H.Y. Ran MLCC-3 (MFLU 18-2279, holotype).

Notes: Present phylogenetic analysis shows that *Poaceascoma herbacum* closely related to the type species *P. helicoides* with high support (Figure 2.6). Morphologically, *Poaceascoma herbacum* is similar to *P. helicoides* by producing ascomata that covered with brown tufts of hyphae, short-pedicellate, elongate-cylindrical asci with filiform ascospores. Nevertheless, it differs from the latter by its remarkably large asci (*P. herbacum* (190–)207–216(–226) × 9–11 μm vs. *P. helicoides* (150–)160–185(–190) × 8.5–10(–11) μm) and ascospores (*P. herbacum* (239–)252–298(–328) × 2–3 μm vs. *P. helicoides* (148–)150–185(–215) × 2–2.5 μm) (Phookamsak et al., 2015a).



Note a Appearance of ascomata throughout the host surface. b Vertical section through ascoma. c Section of peridium. d Pseudoparaphyses anastomosing between the mature asci. e Immature asci. f Mature ascospore. g Immature ascospore. Scale bars: b = 50 µm, c = 10 µm, d, e, g = 20 µm, f = 30 µm.

Figure 2.17 *Poaceascoma herbacum* (MFLU 18-2279, holotype)

Leptosphaeriaceae M.E. Barr

Leptosphaeriaceae was established by Barr (1987) and is typified with *Leptosphaeria*. Ariyawansa et al. (2015b) revised Leptosphaeriaceae based on morphology in conjunction with multi-gene phylogeny, and accepted ten genera within this family. This is in agreement with the subsequent study by Wijayawardene

et al. (2018). In this study, an overlooked genus *Phragmogibbera* is included and analyzed, and the results showed that it is well nested within in Leptosphaeriaceae and sisters to *Praeclarispora*, which was introduced by Doilom et al. (2021). Moreover, the morphological features of *Praeclarispora* are quite similar to *Phragmogibbera*. In view of these above evidence, the genus *Praeclarispora* is synonymized under *Phragmogibbera* as the latter is the older available name. And *Phragmogibbera* is also transferred to Leptosphaeriaceae from Dothideomycetes genera *incertae sedis* (Wijayawardene et al., 2020). And 15 genera *viz*. *Alloleptosphaeria*, *Alternariaster*, *Chaetoplea*, *Heterosporicola*, *Leptosphaeria*, *Neoleptosphaeria*, *Ochraceocephala*, *Paraleptosphaeria*, *Phragmogibbera*, *Plenodomus*, *Pseudoleptosphaeria*, *Querciphoma*, *Sclerenchymomyces*, *Sphaerellopsis* and *Subplenodomus* are accepted in Leptosphaeriaceae based on recent publications (Aiello et al., 2020; Hongsanan et al., 2020a; Phukhamsakda et al., 2020; Doilom et al., 2021; Wijayawardene et al., 2022).

Phragmogibbera Samuels & Rogerson, Mem. N. Y. bot. Gdn 64: 178 (1990)

Phragmogibbera was established to accommodate the type species *P. xylariicola*, which was collected from Guayana Highlands and characterized by uni-loculate, nonpapillate ascomata, bitunicate asci as well as septate ascospores that with the middle cell dark brown, and apically attached (Samuels & Rogerson, 1990). It has been monotypic until Zhuang and Li (2009) introduced a second species *P. herbicola* that discovered from Yunnan Province, China. In this study, two *Phragmogibbera*-like specimens are collected and identified as a reported species *P. herbicola* and a new species *P. karstia* based on morphological and phylogenetic analysis. And a new combination *P. artemisiae* (synonym of *Praeclarispora artemisiae*) is also included in this genus.

Phragmogibbera artemisiae (Doilom, W. Dong, K.D. Hyde & C.F. Liao) J.F. Zhang, J.K. Liu & K.D. Hyde *comb. nov.*

Synonym: *Praeclarispora artemisiae* Doilom, W. Dong, K.D. Hyde & C.F. Liao., Frontiers in Microbiology 12(no. 660261): 8 (2021)

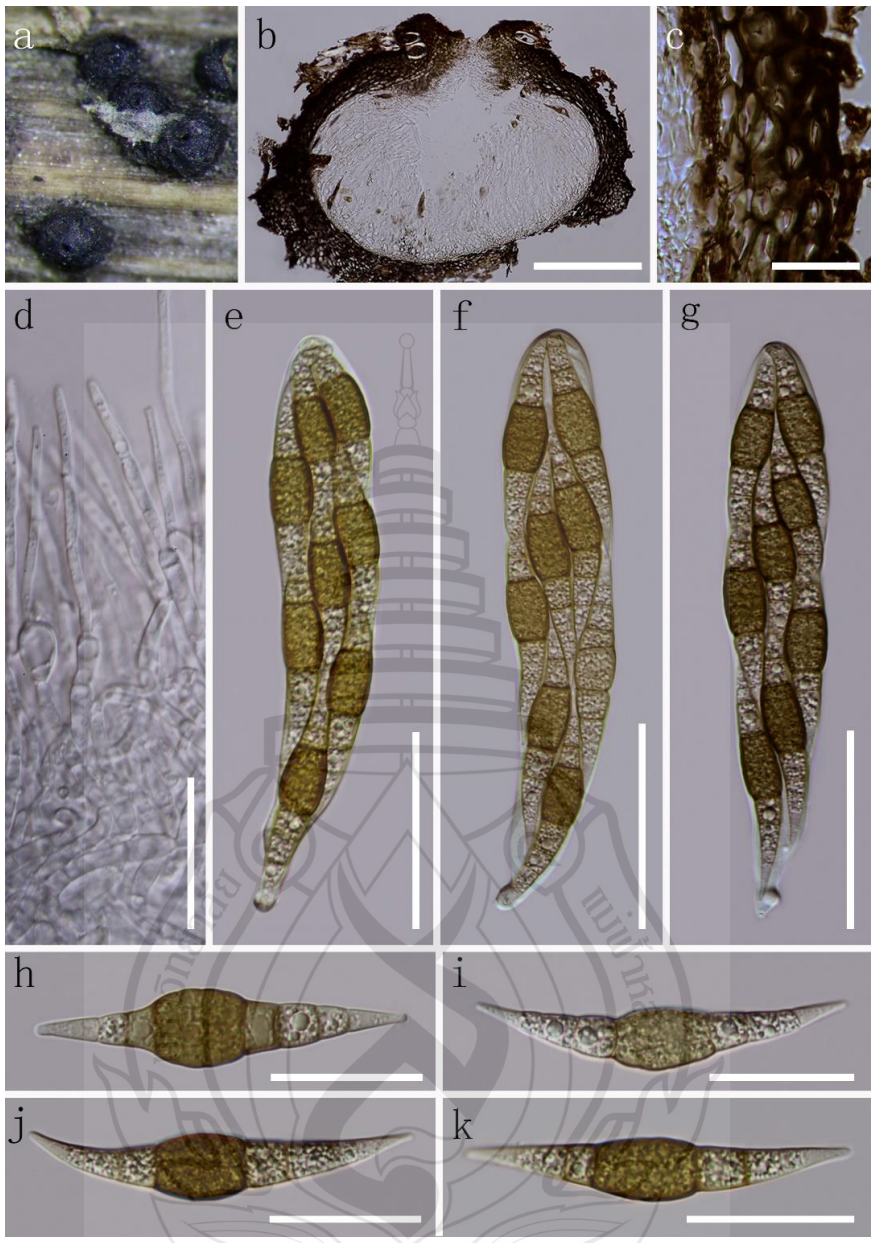
Notes: For the description of *Phragmogibbera artemisiae*, please see the Doilom et al. (2021).

Phragmogibbera herbicola W.Y. Zhuang & W.Y. Li in Zhuang and Li, Macotaxon 107: 340 (2009), Figure 2.18

Saprobic on dead culm of unidentified herbaceous plant. Sexual morph: *Ascomata* semi-immersed to erumpent through epidermis, mostly solitary, occasionally gregarious, uni-loculate, subglobose, nonpapillate or with a slightly protruding apex, dark brown to nearly black, surface slightly roughened, opening through a well-developed ostiole. *Peridium* 36–69 µm wide, composed of several layers of thick-walled, dark brown cells of *textura angularis*, narrower in sides, become broader towards ostiole. *Hamathecium* consisting of 2–3 µm wide (up to 7 µm in somewhere), branched, cellular, hyaline, septate pseudoparaphyses, anastomosing between and above the asci. *Asci* (115–)137–154(–164) × 21–26 µm ($\bar{x}$ = 147 × 23.5 µm, n = 25), bitunicate, clavate to cylindrical-clavate, 8-spored, short pedicellate, with a minute ocular chamber in the apex. *Ascospores* (56.5–)67.5–77 × 9.5–13(–15) µm ($\bar{x}$ = 69 × 11 µm, n = 30), fusiform, tapering above and below to very narrower nearly pointed ends, 3–5-septate when young, 7–9-septate at maturity, with central cell brown to dark brown and swollen, other cells much paler to subhyaline, not constricted at septum, irregularly biseriate. Asexual morph: undetermined.

Material examined: CHINA, Guizhou Province, Guiyang City, Yunyan District, Guizhou Botanical Garden, on dead culm of unidentified herbaceous plant, 8 August 2017, J.F. Zhang ZWY-17 (MFLU 18-2249).

Notes: This newly collected specimen is phylogenetically clusters to *P. herbicola* with strong statistical support (Figure 2.6). Comparison of the morphological characteristics between it and *P. herbicola*, we found that they are almost identical including the size of asci and ascospores. (Zhuang & Li, 2009). Therefore, it is identified as *Phragmogibbera herbicola*, and this taxon has been discovered from Yunnan Province, China.



Note a Appearance of ascomata on the host surface. b Vertical section through ascoma. c Section of peridium. d Pseudoparaphyses. e, f Asci. h–k Ascospores. Scale bars: b = 200 µm, c = 20 µm, d, h–k = 30 µm, e–g = 50 µm.

Figure 2.18 *Phragmogibbera herbicola* (MFLU 18-2249)

Phragmogibbera karstia J.F. Zhang, Y.Y. Chen & J.K. Liu, *sp. nov.*, Figure 2.19

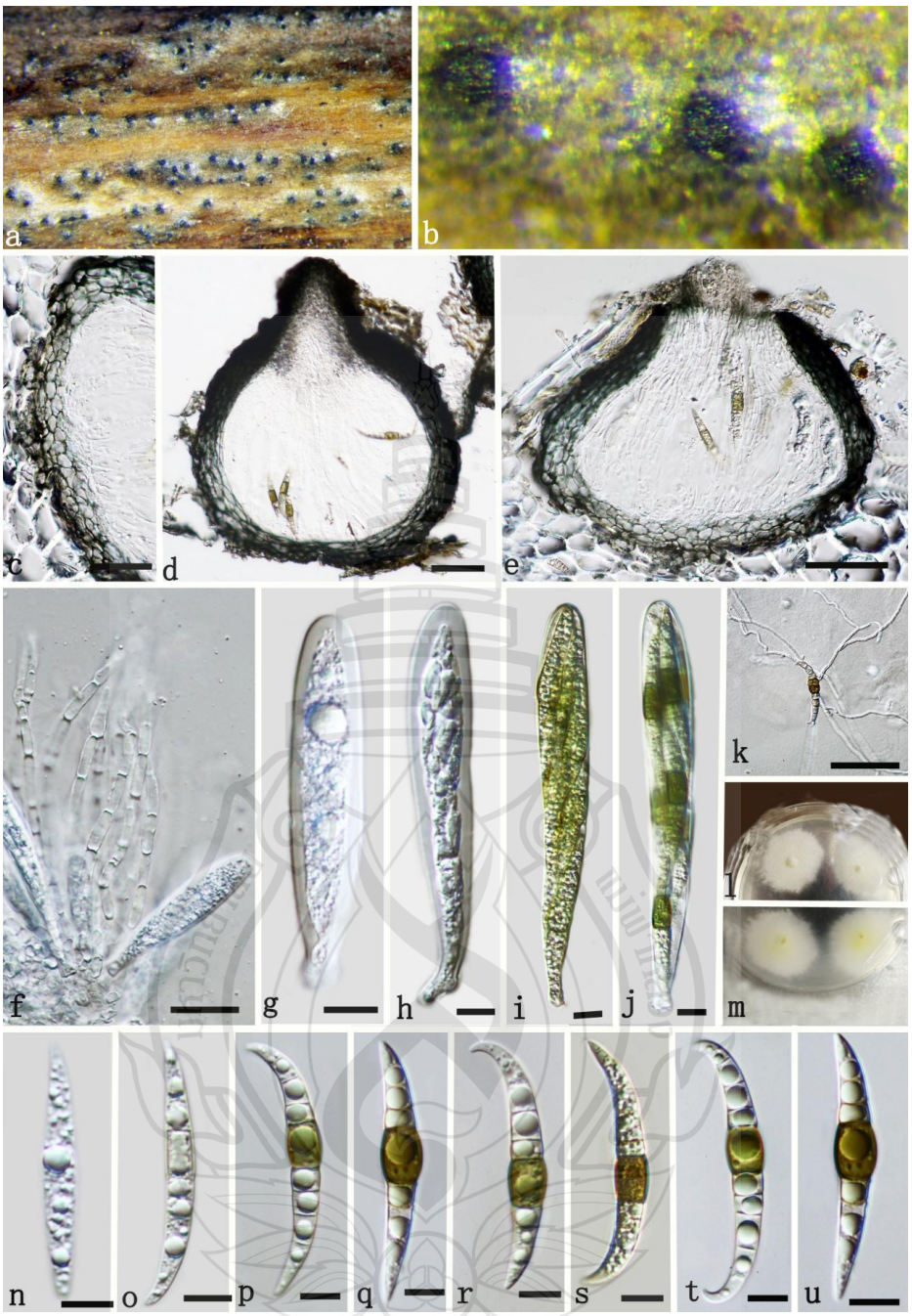
Etymology: "*karstia*" named refers to the holotype was collected from karst habitat.

Holotype: GZAAS 19-1772.

Saprobic on dead culm of unidentified herbaceous plant. Sexual morph: *Ascomata* 140–280 µm high, 190–280 µm diam., black, scattered to gregarious, breaking the epidermis in linear fissures, semi-immersed, becoming erumpent to superficial, subglobose, uni-loculate, coriaceous, ostiolate. *Ostiole* central, brown, ostiolar canal filled with periphyses. *Peridium* 35–45 µm wide, unevenly thick, composed of scleroplectenchymatous cells, arrange in *textura angularis*, outer layer black, inner layer brown. *Hamathecium* comprising 1.5–2 µm wide, numerous, filiform, septate, cellular pseudoparaphyses, embedded in a gelatinous matrix. *Asci* 105–145 × 11–16 µm ($\bar{x}$ = 118 × 13 µm, n = 20), 8-spored, bitunicate, fissitunicate, cylindric-clavate, short pedicellate, apically rounded, with a minute ocular chamber. *Ascospores* 42–55 × 5–7.5 µm ($\bar{x}$ = 47 × 6 µm, n = 30), overlapping bi- to tri-seriate, fusiform, curved and tapered to acute ends, versicolor, becoming brown in median cell, normally hyaline to pale brown in other cells, middle cell is longer and wider than the other cells, 7–9 euseptate at maturity, scarcely constricted at the septa, guttulate, smooth-walled. Asexual morph: Undetermined.

Material examined: CHINA, Guizhou Province, Guiyang City, Huaxi District, on dead culm of unidentified herbaceous plant, 11 May 2018, Y.Y. Chen 18-3 (GZAAS 19-1772, holotype).

Notes: *Phragmogibbera karstia* sisters to *P. artemisiae* and where they are phylogenetically related to *P. herbacola* in the present study (Figure 2.6). Morphologically, *P. karstia* is typical of *Phragmogibbera* in having non-papillate, uni-loculate ascomata, cellular, septate pseudoparaphyses, cylingrical to clavate asci that producing multi-septate fusiform ascospores that with the middle cell pigmented (Samuels & Rogerson, 1990; Zhuang & Li, 2009). However, it is easily differentiated from *P. artemisiae* and *P. herbacola* by its smaller sized ascospores (*P. karstia* 42–55 × 5–7.5 µm vs. *P. artemisiae* 55–70 × 6–11 µm vs. *P. herbacola* (56.5–)67.5–77 × 9.5–13(–15) µm) (Doilom et al., 2021).



Note a, b Appearance of ascomata on the host surface. c Section of peridium. d, e Vertical section through ascoma. f Pseudoparaphyses. g-j Asci. k Germinated ascospore. l, m Colonies on PDA (forward and reverse). n–u Ascospores. Scale bars: c–e = 50 µm, f = 20 µm, g–j, n–u = 10 µm.

Figure 2.19 *Phragmogibbera karstia* (GZAAS 19-1772, holotype)

Plenodomus Preuss, Linnaea 24: 145 (1851)

Plenodomus was introduced by Preuss (1851), and typified with *P. rabenhorstii*. However, Boerema and Kesteren (1964) designated *P. lingam* as the type species as the type material of *P. rabenhorstii* was lost (de Gruyter et al., 2013). Subsequently, Ariyawansa et al. (2015b) re-circumscribed the genus *Plenodomus* and followed their treatment based on examination of type and representative specimens coupled with molecular data. Presently, the accounts of *Plenodomus* were updated by various authors (Marin-Felix et al., 2017; Tennakoon et al., 2017; Phookamsak et al., 2019; Hyde et al., 2020a; Safi et al., 2021) and around 100 epithets listed in Index Fungorum (2022), but accepted with 18 species by Wijayawardene et al. (2022) in the latest outline of fungi.

Plenodomus karsti J.F. Zhang, J.K. Liu & K.D. Hyde, *sp. nov*., Figure 2.20

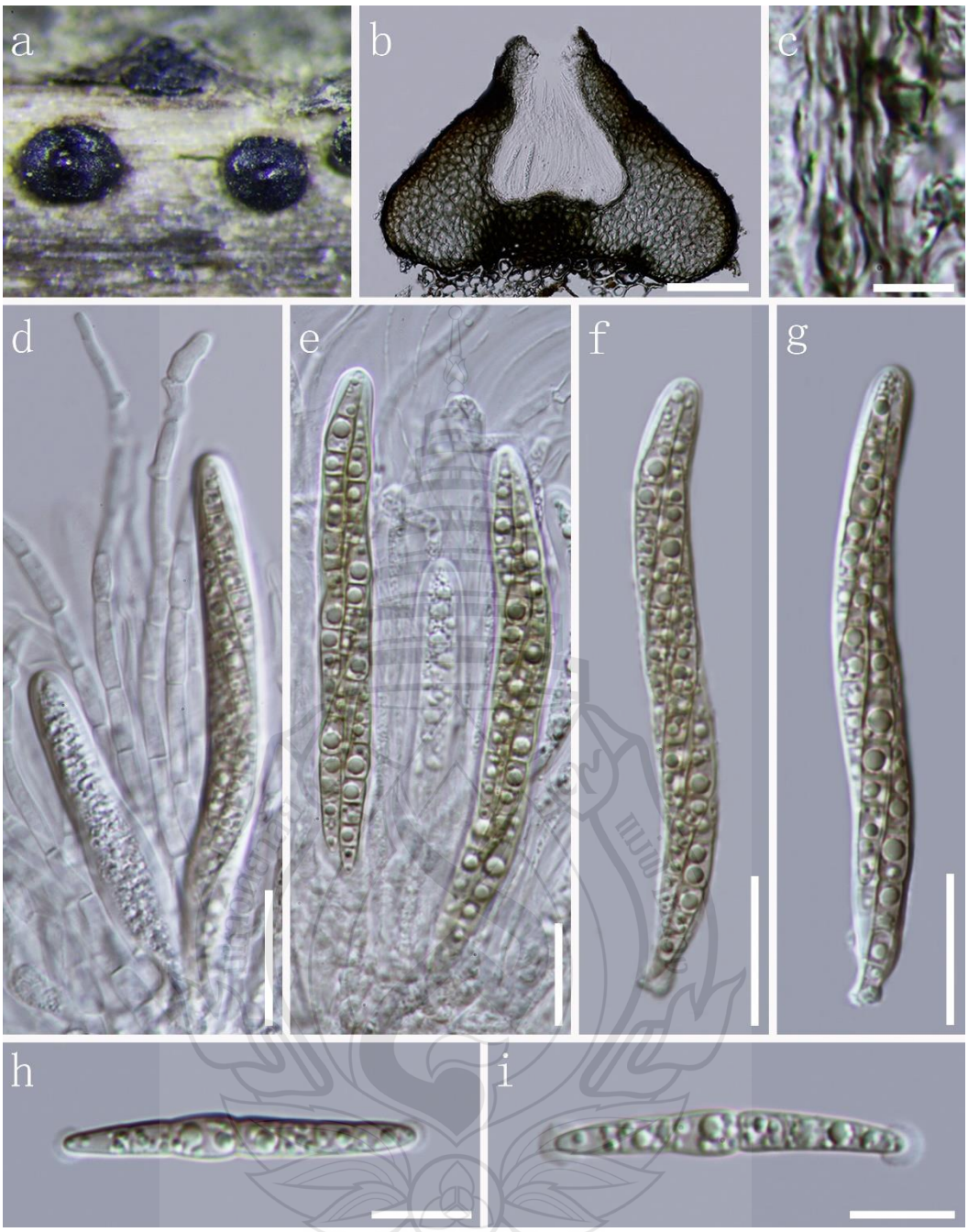
Etymology: "*karstia*" named refers to the holotype was collected form karst formations.

Holotype: MFLU 18-2264.

Saprobic on dead stem of unidentified herbaceous plant. Sexual morph: *Ascomata* solitary, scattered, appearing as small, raised black dots on the host surface, superficial, depressed globose to subglobose, smooth, easily removed from the substrate, with a flattened base, black, ostiolate, short papillate. *Peridium* 80–198 μm at sides, 44–85 μm at base, composed of three layers of scleroplectenchymatous cells, inner layer comprising 3–5 layers of flattened, light brown cells, arranged in a *textura angularis*, middle layer comprising several layers of sub-hyaline cells arranged in a *textura globulosa*, outer layer composed of dark pigmented, thick-walled, dark brown cells arranged in irregular organization. *Hamathecium* comprising 2–3 μm wide, septate, cellular pseudoparaphyses, branching between the asci, embedded in a gelatinous matrix. *Asci* (77–)80–88(–93) × (7–)8–10 μm ($\bar{x}$ = 86 × 8.5 μm, n = 20), 8-spored, bitunicate, fissitunicate, cylindrical, short pedicel, apically rounded, with a distinct ocular chamber. *Ascospores* 29–35 × 3.5–4.5 μm ($\bar{x}$ = 32.5 × 4 μm, n = 25), overlapping uni- or bi-seriate, olivaceous to yellowish, cylindrical to subfusoid, with obtuse ends, 3–6 septate, not or slightly constricted at each septum, cell above central septum slightly wider, guttulate, thick and smooth-walled, with mucilaginous globoid-shaped apical and basal appendages. Asexual morph: Undetermined.

Material examined: CHINA, Guizhou Province, Huishui County, on dead stem of unidentified herbaceous plant, 21 July 2017, J.F. Zhang, HS-11 (MFLU 18-2264, holotype).

Notes: *Plenodomus karsti* clusters with *P. macrosporium* and where they form an independent clade that sister to other two reported taxa *P. collinsoniae* and *P. sinensis* by strong statistical support (Figure 2.22). *Plenodomus karsti* resembles to *P. macrosporium* and *P. sinensis* in having peridium with scleroplectenchymatous cells, cylindrical, fissitunicate asci with yellowish brown, multi-septate ascospores bearing globoid-shaped mucilaginous appendages at both ends. However, it is distinguished from the latter two taxa by its comparatively smaller sized asci (*P. karsti*, (77–)80–88(–93) × (7–)8–10 μm vs. *P. macrosporium*, (108.5–)116–126(–134) × 14–17 μm vs. *P. sinensis*, (78–)80–100(–108) × (9.5–)10–12(–15) μm) and ascospores (*P. karsti*, 29–35 × 3.5–4.5 μm vs. *P. macrosporium*, (45–)47–53(–58.5) × 6–8 μm vs. *P. sinensis*, (25–)27–40(–42) × 3.8–4.4 μm) (Tennakoon et al., 2017). While *P. karsti* differs from *P. collinsoniae* in having 3–6 septate ascospores with mucilaginous globoid-shaped appendages at both ends, but the ascospores of *Plen. collinsoniae* are 5–8 septate and without any gelatinous appendages (de Gruyter et al., 2013).



Note a Appearance of ascomata in the host surface. b Vertical section through ascoma. c Section of inner layer of peridium. d, e Pseudoparaphyses anastomosed between asci. f, g Asci. g, h Ascospores. Scale bars: b = 100 μm, c = 10 μm, d–g = 20 μm, h, i = 10 μm.

Figure 2.20 *Plenodomus karsti* (MFLU 18-2264, holotype)

Plenodomus macroaporium J.F. Zhang, J.K. Liu & Z.Y. Liu, *sp. nov.*, Figure 2.21

Etymology: "*macrosporium*" named refers to the large sized ascospores of the holotype.

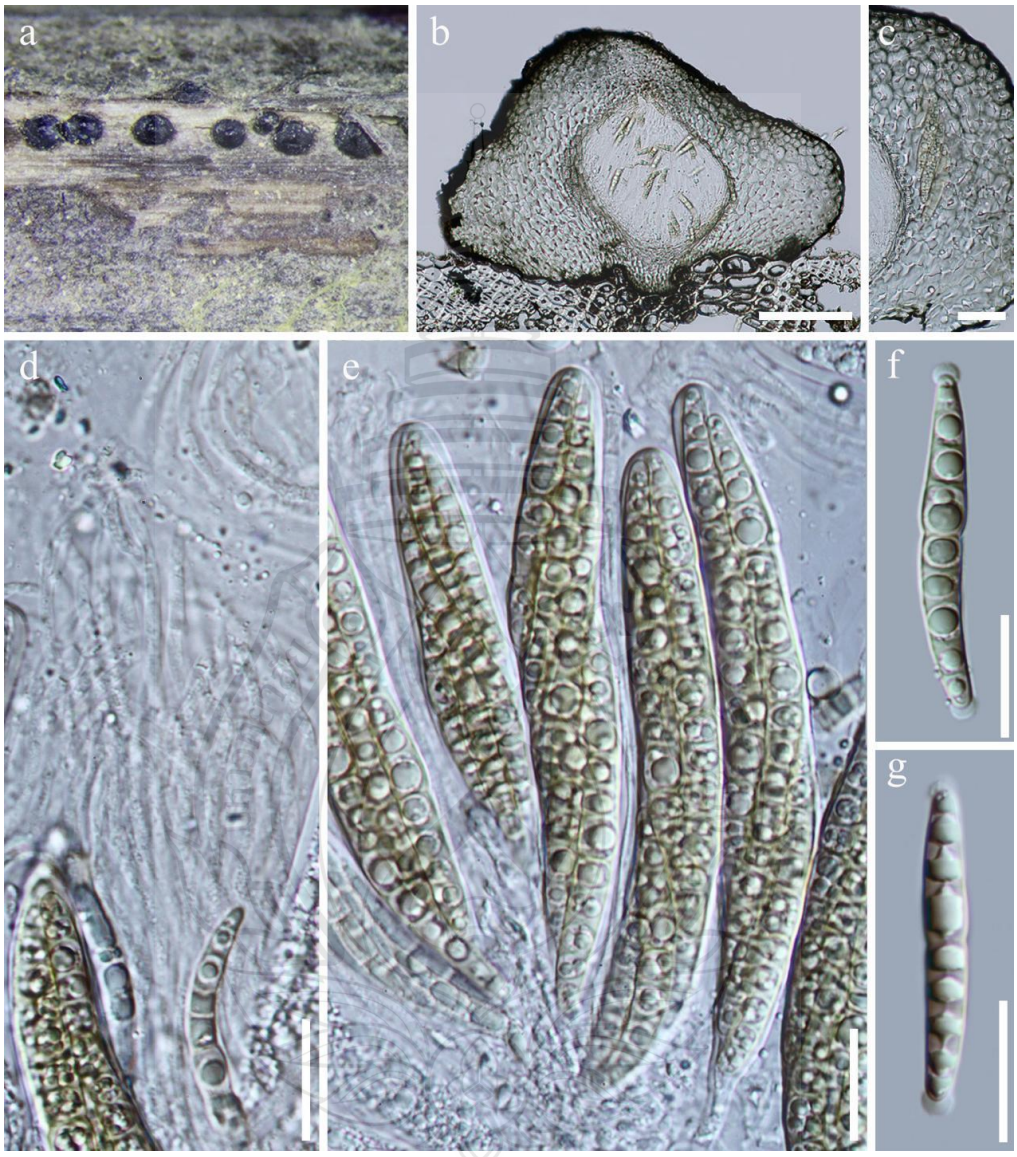
Holotype: MFLU 18-2250.

Saprobic on dead culm of unidentified herbaceous plant. Sexual morph: *Ascomata* solitary, scattered, appearing as small, raised black dots on the host surface, superficial, globose to subglobose, smooth, easily removed from the host substrate, with a flattened base, black, ostiolate, papillate. *Peridium* broadly at the sides, up to 90–134 μm, thin at the base of 27–55 μm, composed of three layers of scleroplectenchymatous cells, inner layer comprising 3–5 layers of flattened, pale brown cells, arranged in a *textura angularis*, middle layer comprising several layers of sub-hyaline cells arranged in a *textura globulosa*, outer layer composed of dark pigmented, thick-walled, dark brown cells arranged in irregular organization. *Hamathecium* comprising 2–3 μm wide, septate, cellular pseudoparaphyses, anastomosing between the asci, embedded in a gelatinous matrix. *Asci* (108.5–)116–126(–134) × 14–17 μm ($\bar{x}$ = 121.5 × 16 μm, n = 25), 8-spored, bitunicate, fissitunicate, cylindrical, non-pedicellate, apically rounded, with a distinct ocular chamber. *Ascospores* (45–)47–53(–58.5) × 6–8 μm ($\bar{x}$ = 51.5 × 7 μm, n = 35), overlapping bi- to tri-seriate, cylindrical to subfusoid, with obtuse ends, straight or lightly curved, hyaline and one-celled when young, and become 3–6 septate, olivaceous to yellowish at maturity, not or slightly constricted at each septum, cell above central septum slightly wider, guttulate, thick and smooth-walled, with a globoid-shaped mucilaginous appendages at both ends. Asexual morph: Undetermined.

Material examined: CHINA, Guizhou Province, Guiyang City, Yunyan District, Guizhou Botanical Garden, on dead culm of unidentified herbaceous plant, 8 August 2017, J.F. Zhang, ZWY-18 (MFLU 18-2250, holotype).

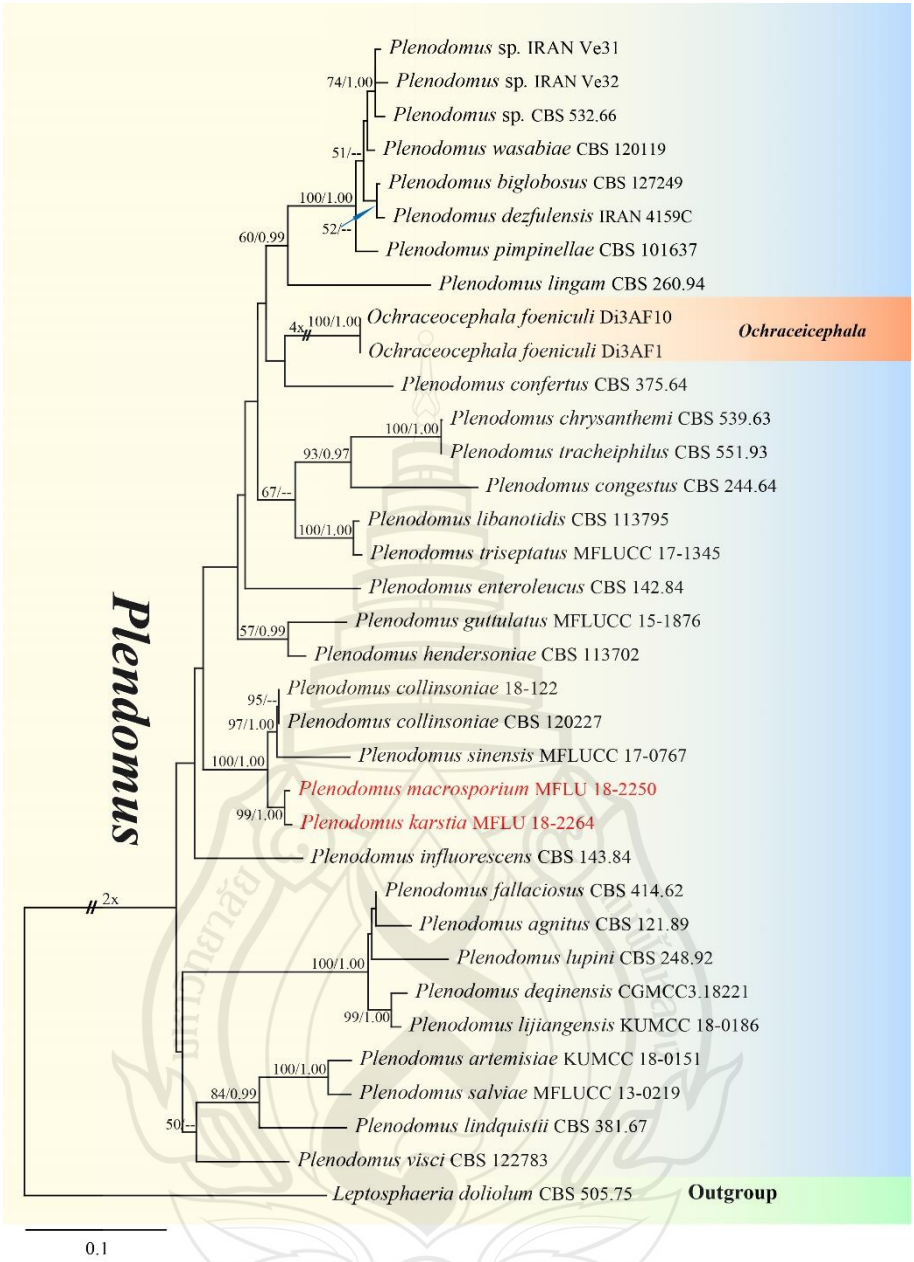
Notes: Present phylogenetic analysis shows that *Plenodomus macroaporium* is phylogenetically related to *P. collinsoniae*, *P. karsti* and *P. sinensis* (Figure 2.22). However, it differs from these taxa by having obviously large sized asci and ascospores. Moreover, the measurements of the asci and ascospores of *P.*

macroaporium correspond to the type species of *Plenodomus*, *P. lingam*, nevertheless, the ascospores of the latter lacking the mucilaginous appendages at both ends, and they are also phylogenetically distant in the present analysis (Figure 2.22).



Note a Appearance of ascomata in the host surface. b Vertical section through ascoma. c Section of peridium. d Pseudoparaphyses. e Asci. f, g Ascospores. Scale bars: b = 100 μm, c = 30 μm, d–g = 20 μm.

Figure 2.21 *Plenodomus macroaporium* (MFLU 18-2250, holotype)



Note Maximum likelihood bootstrap values equal or greater than 50% and Bayesian posterior probabilities equal or greater than 0.95 are indicated near the nodes. New sequence generated in this study are given in red. The tree is generated from maximum likelihood (ML) analysis based on ITS, Tub and Rpb2 sequence data, and rooted with Leptosphaeria doliolum CBS 505.75.

Figure 2.22 Phylogenetic tree of the genus *Plendomus*

Lophiostomataceae Sacc.

Lophiostomataceae was established by Saccardo (1883) with *Lophiostoma* as the typus. Then, the placement of this family was challenged and placed in different orders (Dothideales and Pleosporales) based on morphology (Luttrell, 1973; Von Arx & Müller, 1975; Hawksworth et al., 1995). However, Schoch et al., (2006, 2009) demonstrated that Lophiostomataceae belongs to Pleosporales in their multi-gene phylogenetic analysis. Subsequently, several new genera were introduced and different account were included in Lophiostomataceae by various studies (Lumbsch & Huhndorf, 2010; Hyde et al., 2013; Thambugala et al., 2015; Hashimoto et al., 2018; Wanasinghe et al., 2018b, 2021; Wijayawardene et al. 2018, 2020; Crous et al., 2019a; Hongsanan et al., 2020a; Mapook et al., 2020). However, Andreasen et al. (2021) synonymized 14 genera under *Lophiostoma* based mainly on multi-gene (ITS, LSU, TEF1-α and RPB2) phylogenetic analysis, and broadened the concept of *Lophiostoma*.

Lophiostoma Ces. & De Not., Comm. Soc. crittog. Ital. 1(fasc. 4): 219 (1863)

Lophiostoma was introduced by Cesati and De Notaris (1863) with *L. macrostomum* as the type species. This genus is morphologically similar to *Lophiotrema*, and it has been therefore treated as a synonym of *Lophiostoma* (Chesters & Bell, 1970). Nevertheless, Holm and Holm (1988) distinguished these two genera based on the difference type of peridium and asci, and Hirayama and Tanaka (2011) also confirmed that they are separate genus on the basis of both molecular and morphological studies. Recently, the lophiostomatoid fungi was revisited and the concept of *Lophiostoma* was broadened by comprising of 14 genera that accepted in previous studies (Andreasen et al., 2021).

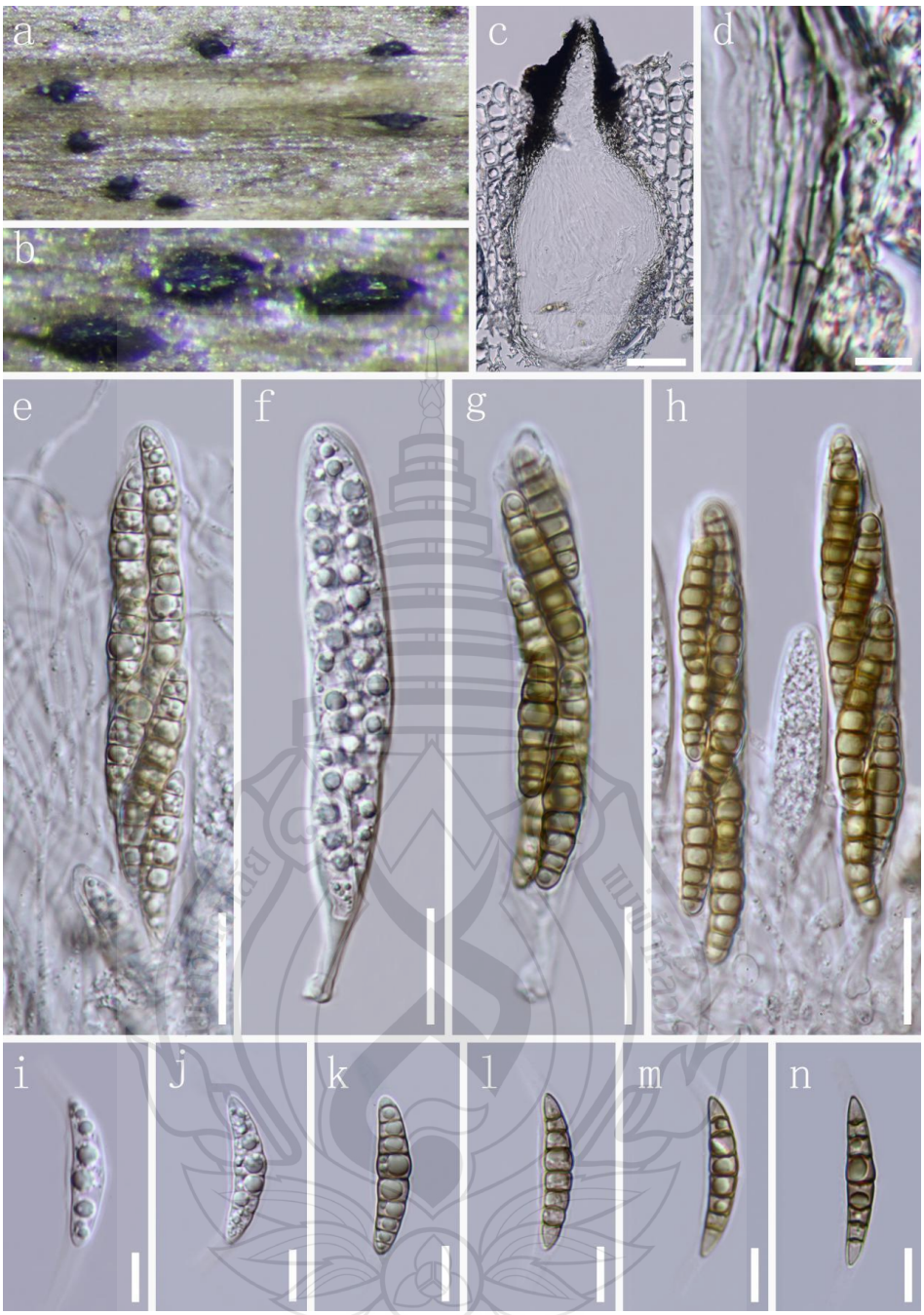
Lophiostoma japonicum (Thambug., Wanas., Kaz. Tanaka & K.D. Hyde) Andreasen, Jaklitsch & Voglmayr, Persoonia 46: 256 (2021). Figure 2.23

Saprobic on dead culm of unidentified herbaceous plant. Sexual morph: *Ascomata* immersed to erumpent through host surface with papilla, solitary, gregarious, coriaceous, dark brown to black, subglobose, ostiolate. *Ostiole* central, rounded or slit-like, with a pore-like opening. *Peridium* 14–22 μm wide, composed of several layers of lightly pigmented to brown cells of *textura angularis*, cells towards the inside lighter, at the outside, darker, fusing and indistinguishable from the host

tissues, and becoming thick, and black near the apex. *Hamathecium* comprising numerous, 1.5–2.5 μm wide, branched, septate pseudoparaphyses, situated between and above the asci, embedded in a gelatinous matrix. *Asci* 96.5–103 × 13–14.5 (–16) μm ($\bar{x}$ = 100×14.2 μm, n=20), 8-spored, bitunicate, fissitunicate, cylindric-clavate, with short, bulbous pedicel, rounded at the apex, with an ocular chamber. *Ascospores* 30–32.5 (–35.5) × 5–7 (–8) μm ($\bar{x}$ = 32.4 × 6.3 μm, n=25), overlapping biseriate to 3-seriate, fusiform with acute ends, sometimes curved, hyaline and non-septate when young, and becoming 7–9-septate, brown at maturity, constricted at the septa, cell above central septum widest, guttulate, smooth-walled, surrounded by a gelatinous appendage, and extended up to 7–11 μm at both ends. Asexual morph: Undetermined.

Material examined: CHINA, Guizhou Province, Huishui County, on dead culm of unidentified herbaceous plant, 21 July 2017, J.F. Zhang HS-07 (MFLU 18-2261); Huishui County, on dead culm of unidentified herbaceous plant, 21 July 2017, J.F. Zhang HS-03 (GZAAS 21-2004); Sinan County, on dead culm of unidentified herbaceous plant, 22 May 2019, J.F. Zhang SN-2019-1 (GZAAS 21-2015).

Notes: Current phylogenetic analysis shows that three newly collected specimens cluster to *L. japonicum* by high bootstrap support (Figure 2.6). Moreover, these fresh specimens are typical of *Lophiostoma*, and similar to *L. japonicum* including the overlapped sized of pseudoparaphyses, asci and ascospores (Thambugala et al., 2015; Bao et al., 2019). In view of these above evidence, these new collections in this study are identified as *L. japonicum*.



Note a, b Appearance of ascomata in the host surface. c Vertical section through ascomata. d Section of peridium. e Pseudoparaphyses anostomosing with the asci. f–h Young and mature asci. i–n Young and mature ascospores. Scale bars: c = 50 µm, d = 5 µm, e–h = 20 µm, i–n = 10 µm.

Figure 2.23 *Lophiostoma japonicum* (MFLU 18-2261)

Vaginatispora K. D. Hyde, Nova Hedwigia 61(1-2): 234 (1995)

Vaginatispora was established to accommodate the type species *V. aquatica* (Hyde, 1995), and synonymized under *Lophiostoma* by Zhang et al. (2014). However, Thambugala et al. (2015) retained *Vaginatispora* as a separate genus based on phylogenetic and morphological evidence. Then, *Vaginatispora* was recircumscribed by Hashimoto et al. (2018), and 8 species are currently included in this genus (Devadatha et al., 2017; Hashimoto et al., 2018; Hyde et al., 2019a; Jayasira et al., 2019).

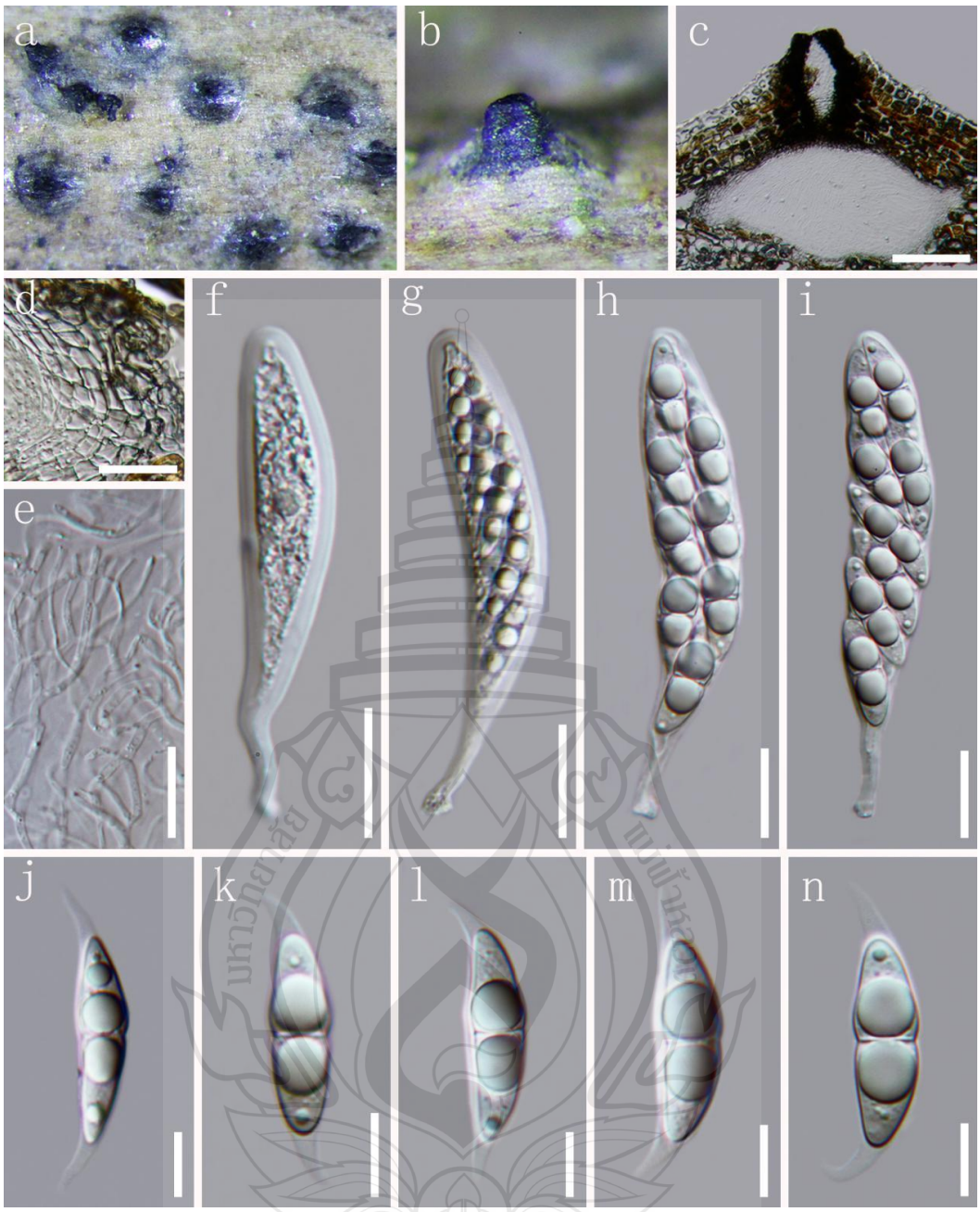
Vaginatispora amygdali A. Hashim et al., in Hashimoto et al., Stud. Mycol 90: 179 (2018), Figure 2.24

Saprobic on dead branch of unidentified woody plant. Sexual morph: *Ascomata* subglobose, scattered, immersed, dark brown to black, 281–312 μm high, 352–433 μm diam. *Ostiolar* neck crest-like, elongated, laterally compressed, 124–159 μm high, composed of 110–188 μm diam, globose to elongated, brown to black cells, with hyaline periphyses. *Peridium* 15–42 μm wide, composed of several layers of rectangular, thin-walled, hyaline to pale brown cells, arrange in *textura angularis*. *Hamathecium* comprising of numerous, 1–3 μm wide, septate, branched *pseudoparaphyses*, anastomosed between and above the asci. *Asci* (87.5–)95–110(–116.5) × 16–20 μm ($\bar{x}$ = 101 × 18 μm, n = 20), 8-spored, bitunicate, fissitunicate, cylindric-clavate, with a short stipe, apically rounded with a broad ocular chamber. *Ascospores* (23.5–)25.5–30(–32.5) × 7–9 μm ($\bar{x}$ = 27.5 × 8.5 μm, n = 40), fusiform with obtuse ends, hyaline, slightly constricted at the septum, smooth, with a narrow sheath. Sheath drawn out 6–8.5 μm long at both ends, with a lateral pad-like structure within the sheath, with an internal chamber at both ends of ascospores. Asexual morph: Undetermined.

Material examined: CHINA, Guizhou Province, Libo County, Maolan Town, Maolan National Nature Reserve, on dead branch of unidentified woody plant, 18 July 2016, J.F. Zhang MLAA-04 (MFLU 18-2275).

Notes: The new specimen (MFLU 18-2275) is phylogenetically placed in *Vaginatispora* and clusters to *V. amygdali* by strong statistical support in the present study (Figure 2.6). Additionally, the morphological features of this specimen are typical of *Vaginatispora*, and especially similar to *V. amygdali* by having almost the

same sized asci and asocpores (Hashimoto et al., 2018). Hence, it is identified as *V. amygdali*, and which has been found from China and Japan (Hashimoto et al., 2018; Yang et al., 2022).



Note a, b Appearance of ascomata in the host surface. c Vertical section through ascoma. d Section of peridium. e Pseudoparaphyses. f–i Asci in different maturation stage. j–n Ascospores in different maturation stage. Scale bars: c = 100 μm, d–i = 20 μm, j–n = 10 μm.

Figure 2.24 *Vaginatispora amygdali* (MFLU 18-2275)

Lophiotremataceae K. Hiray. & Kaz. Tanaka

Lophiotremataceae was established as a monotypic family to accommodate the type genus *Lophiotrema* (Hirayama & Tanaka, 2011). Subsequently, other two genera *Aquasubmersa* and *Hermatomyces* were respectively assigned in this family based on phylogenetic studies (Ariyawansa et al., 2015a; Doilom et al., 2017). However, Hashimoto et al. (2017) removed *Aquasubmersa* and *Hermatomyces* from this family, and included other five genera (*Atrocalyx*, *Crassimassarina*, *Cryptoclypeus*, *Galeaticarpa* and *Pseudocryptoclypeus*) by revision of Lophiotremataceae based on morphological characteristics and multi-gene analysis. And this treatment was followed by Wijayawardene et al. (2018). Currently, the genus *Koordersiella* was also comprised of this family in the updated document on families of Dothideomycetes and outline of fungi (Hongsanan et al., 2020a; Wijayawardene et al., 2022).

Lophiotrema Sacc., Michelia 1(no. 3): 338 (1878)

Saccardo (1878) established the genus *Lophiotrema* and defined it as a group that comprised lophiostomataceous fungi, which are characterized by having hyaline, multi-septate ascospores. However, Chesters and Bell (1970) synonymized it under *Lophiostoma* and until Holm and Holm (1988) distinguished these two genera on the basis of morphology. Hirayama and Tanaka (2011) separated *Lophiotrema* from *Lophiostoma* based on morphological and phylogenetic analysis, and established the family Lophiotremataceae to accommodate *Lophiotrema.* In the past few years, *Lophiotrema* taxa were continually being discovered by mycologists (Liu et al., 2015; Hyde et al., 2016; Hashimoto et al., 2017; Mathiassen et al., 2017, 2018; Zhang et al., 2018; Phookamsak et al., 2019) Nevertheless, Hashimoto et al. (2017) suggested that the boundary of *Lophiotrema* should be narrowed, and the ambiguous taxa included in this genus well be examined in further study.

Lophiotrema hydei J.F. Zhang, J.K. Liu & Z.Y. Liu, *sp. nov*., Figure 2.25

Etymology: Named in honour of Prof. K.D. Hyde for his contributions in mycology.

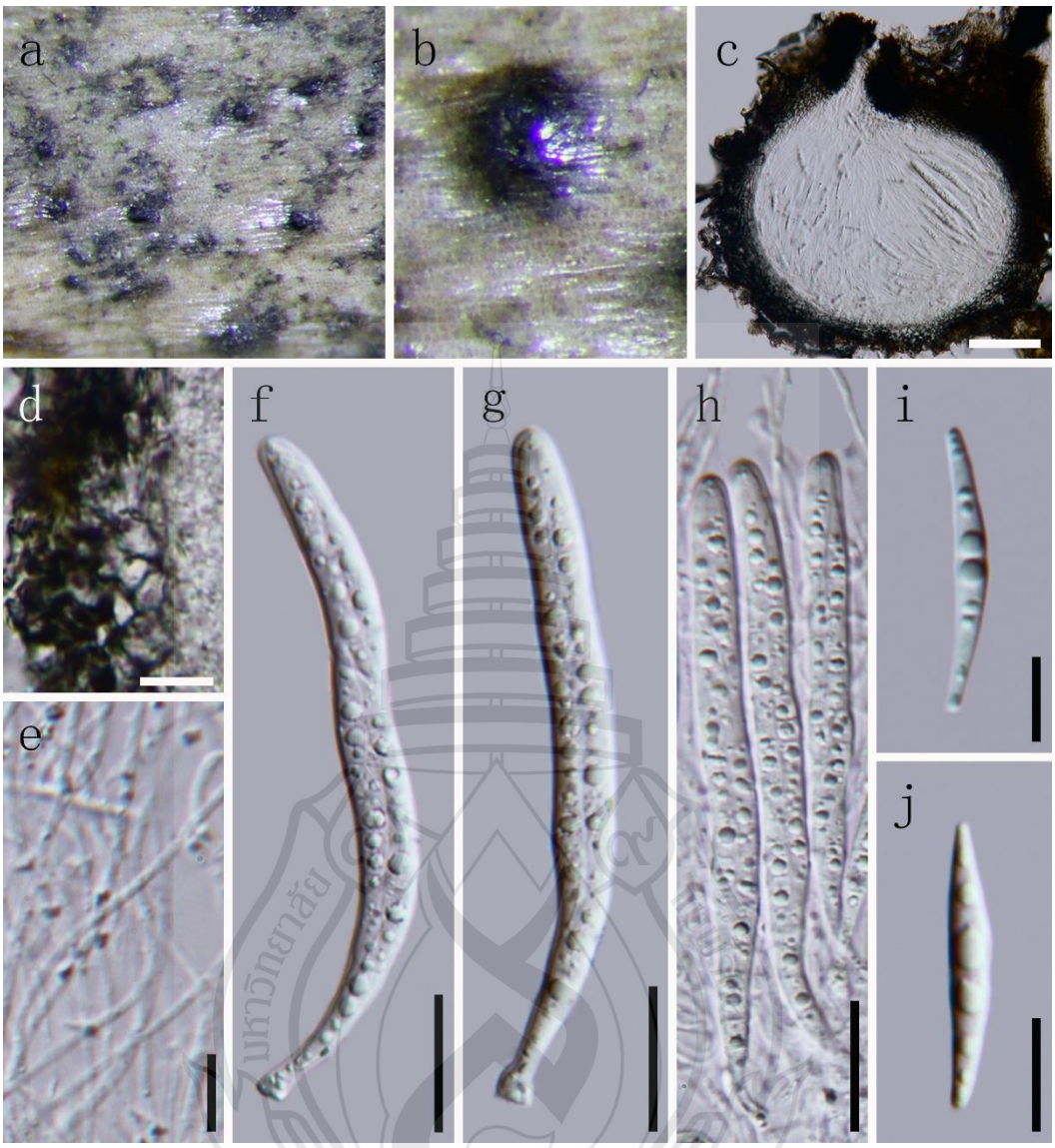
Holotype: MFLU 18-2075.

Saprobic on dead branch of unidentified herbaceous plant. Sexual morph: *Ascomata* dark brown to black, scattered, gregarious, immersed under the host

epidermis, globose to subglobose, uni-loculate, glabrous, coriaceous, ostiolate. *Ostioles* central or lateral, carbonaceous, normally with a pore-like opening. *Peridium* up to 26–36 μm thick, composed of several layers of dark-brown to black, thick-walled pseudoparenchymatous cells, arranged in a *textura angularis*, fusing with host tissue at the most outside layer and pale inwardly. *Hamathecium* comprising 1.2–1.7 μm wide, branched, cellular pseudoparaphyses, anastomosing among and between the asci, embedded in a gelatinous matrix. *Asci* 78–89(–99) × 6.9–8.8 μm ($\bar{x}$ = 88.5 × 7.7 μm, n = 20), 8-spored, bitunicate, fissitunicate, cylindrical, pedicellate, apically rounded with a minute ocular chamber. *Ascospores* 23–28 × 3–4 μm ($\bar{x}$ = 25.8 × 3.7 μm, n = 25), overlapping bi-seriate, hyaline, fusiform, straight or slightly curved, one-celled when young and become inconspicuously 1-septate at maturation, the upper cell is longer than the lower cell, slightly constricted at the septum, narrower towards both ends, smooth-walled, guttulate. Asexual morph: Undetermined.

Material examined: CHINA, Guizhou Province, Guiyang City, Yunyan District, on dead branch of unidentified herbaceous plant, 6 May 2016, J.F. Zhang, SH-08 (MFLU 18-2075, holotype; GZAAS 18-0005, isotype), ex-type living culture GZCC 18-0016.

Notes: Phylogeny (Figure 2.28) indicates that *Lophiotrema hydei* is phylogenetically close to *L. neohysterioides*, however it differs morphologically from the latter in several respects. The appearance of ascomata in *L. neohysterioides* is strongly erumpent throughout the substrate with ostiole, but does not stain the host tissue, whereas the ascomata of our new species is immersed, and the host epidermis is become dark pigmented around the ostiole. Moreover, the ascospores of our new species are inconspicuously 1-septate and narrower towards both ends, nevertheless, the ascospores of *L. neohysterioides* are observed as in having three equidistant septa, and both of the ends are rounded or obtuse. In addition, there are 9 and 38 nucleotide differences in ITS and TEF1-α gene regions respectively between these two taxa, therefore *Lophiotrema hydei* and *L. neohysterioides* can be clearly identified as different species based on the recommendations of Jeewon and Hyde (2016).



Note a, b Ascomata immersed under the host surface. c Vertical section through ascoma. d Section of peridium. e Pseudoparaphyses. f–h Long cylindrical asci. i, j Ascospores. Scale bars: c = 50 μm, d, e, i, j = 10 μm, f–h = 20 μm.

Figure 2.25 *Lophiotrema hydei* (MFLU 18-2075, holotype)

Lophiotrema karstii J.F. Zhang & J.K. Liu, *sp. nov.*, Figure 2.26

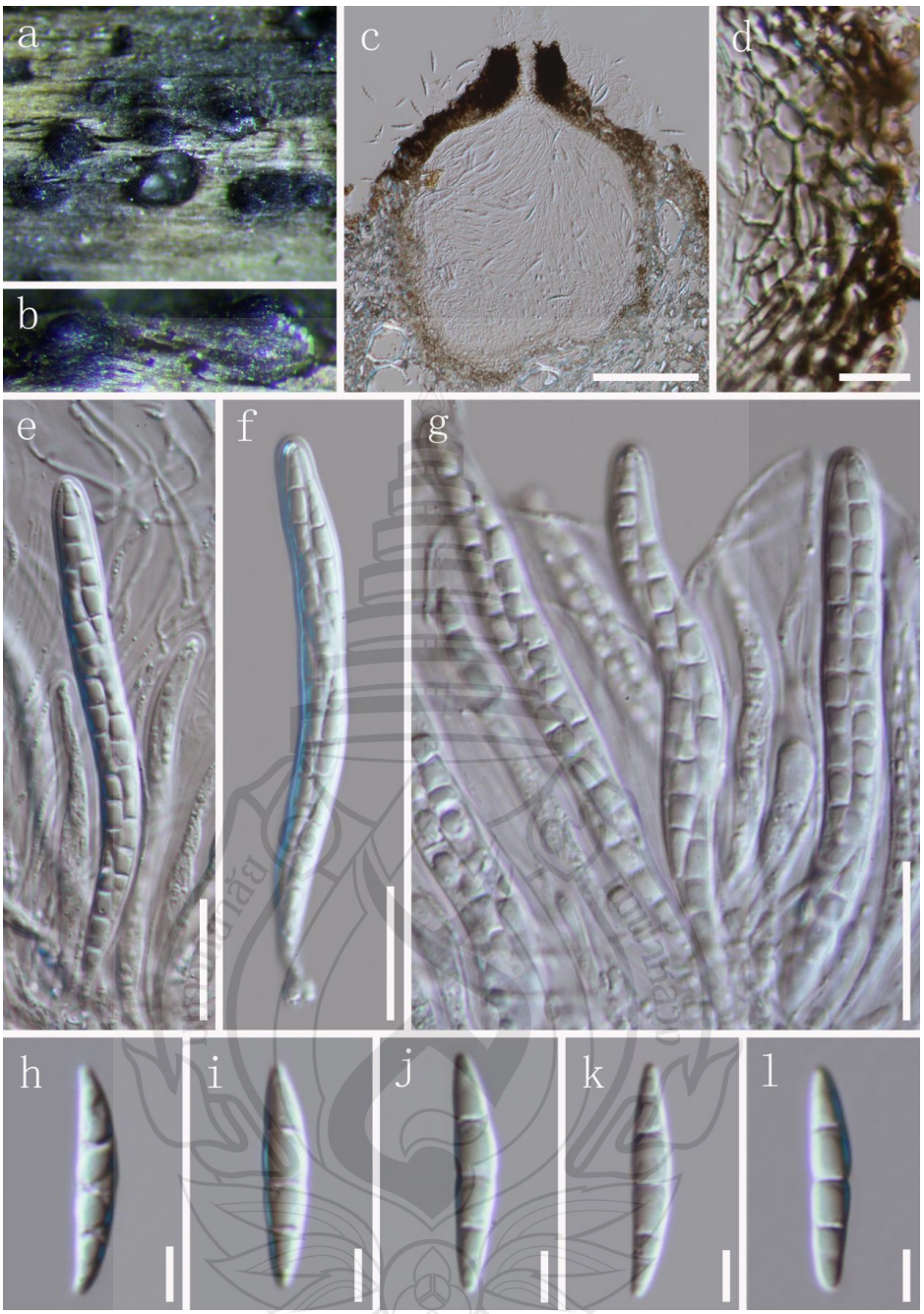
Etymology: "*karstii*" named refers to the holotype is collected form karst formations.

Holotype: GZAAS 19-1750.

Saprobic on dead culm of unidentified herbaceous plant. Sexual morph: *Ascomata* dark brown to black, scattered, gregarious, immersed under the host epidermis, globose to subglobose, uni-loculate, glabrous, coriaceous, ostiolate. *Ostioles* central or lateral, carbonaceous, normally with a pore-like opening. *Peridium* 21–46 μm thick, composed of several layers of dark-brown to black, thick-walled pseudoparenchymatous cells, arranged in a *textura angularis*, fusing with host tissue at the most outside layer and pale inwardly. *Hamathecium* comprising 1.5–2 μm wide, branched, hypha-like pseudoparaphyses, anastomosing among and between the asci, embedded in a gelatinous matrix. *Asci* (76.5–)80.5–90(–93) × 6.5–7.5 μm ($\bar{x}$ = 86 × 7 μm, n = 25), 8-spored, bitunicate, fissitunicate, cylindrical, pedicellate, apically rounded with a minute ocular chamber. *Ascospores* 19.5–23.5 × 3.5–4.5 μm ($\bar{x}$ = 22 × 3.5 μm, n = 25), overlapping bi-seriate, hyaline, fusiform, straight or slightly curved, one-celled when young and become 3-euseptate at maturation, the upper cell is longer than the lower cell, slightly constricted at the septum, narrower towards both ends, smooth-walled, guttulate. Asexual morph: Undetermined.

Material examined: CHINA, Guizhou Province, Sinan County, on dead culm of unidentified herbaceous plant, 22 May 2019, H.Y. Ran SN-2019-2 (GZAAS 19-1750, holotype).

Notes: *Lophiotrema karstii* is phylogenetically close to *L. neohysterioides* by high bootstrap support in this study (Figure 2.6). This species is typical of *Lophiotrema* in having subglobose ascomata with crest-like papilla and cylindrical, short pedicellate asci as well as fusiform, hyaline, multi-septate ascospores (Saccardo, 1878; Hirayama & Tanaka, 2011; Andreasen et al., 2021). Morphologically, our new specimen resembles to *Lop. neohysterioides* by having overlap sized of asci and ascospores (Tanaka & Harada, 2002; Zhang et al., 2018), however, it can be distinguished from the latter by the comparison of each partial gene regions (there are 2.5% and 2.76% differences in TEF1-a and Rpb2 respectively) based on the recommendations from Jeewon and Hyde (2016). Therefore, *Lophiotrema karstii* is identifies as a new species and described herein.



Note a, b Appearance of ascomata in the host surface. c Vertical section through ascoma. d Section of peridium. e Pseudoparaphyses anastomosing between asci. f, g Asci. h–l Ascospores in different maturation stage. Scale bars: c = 100 μm, d = 10 μm, e–g = 20 μm, h–l = 5 μm.

Figure 2.26 *Lophiotrema karstii* (GZAAS 19-1750, holotype)

Lophiotrema neohysterioides M.E. Barr, Mycotaxon 45: 208, 1992. Figure 2.27

Facesoffungi number: FoF 05217

≡ *Sphaeria hysterioides* Schwein., Trans. Am. Philos. Soc. 4: 216, 1832.

≡ *Lophiostoma hysterioides* (Schwein.) Sacc., Syll. Fung. 2: 709, 1883.

≡ *Lophiostoma hysterioides* Ellis & Langl., J. Mycol. 4: 76, 1888.

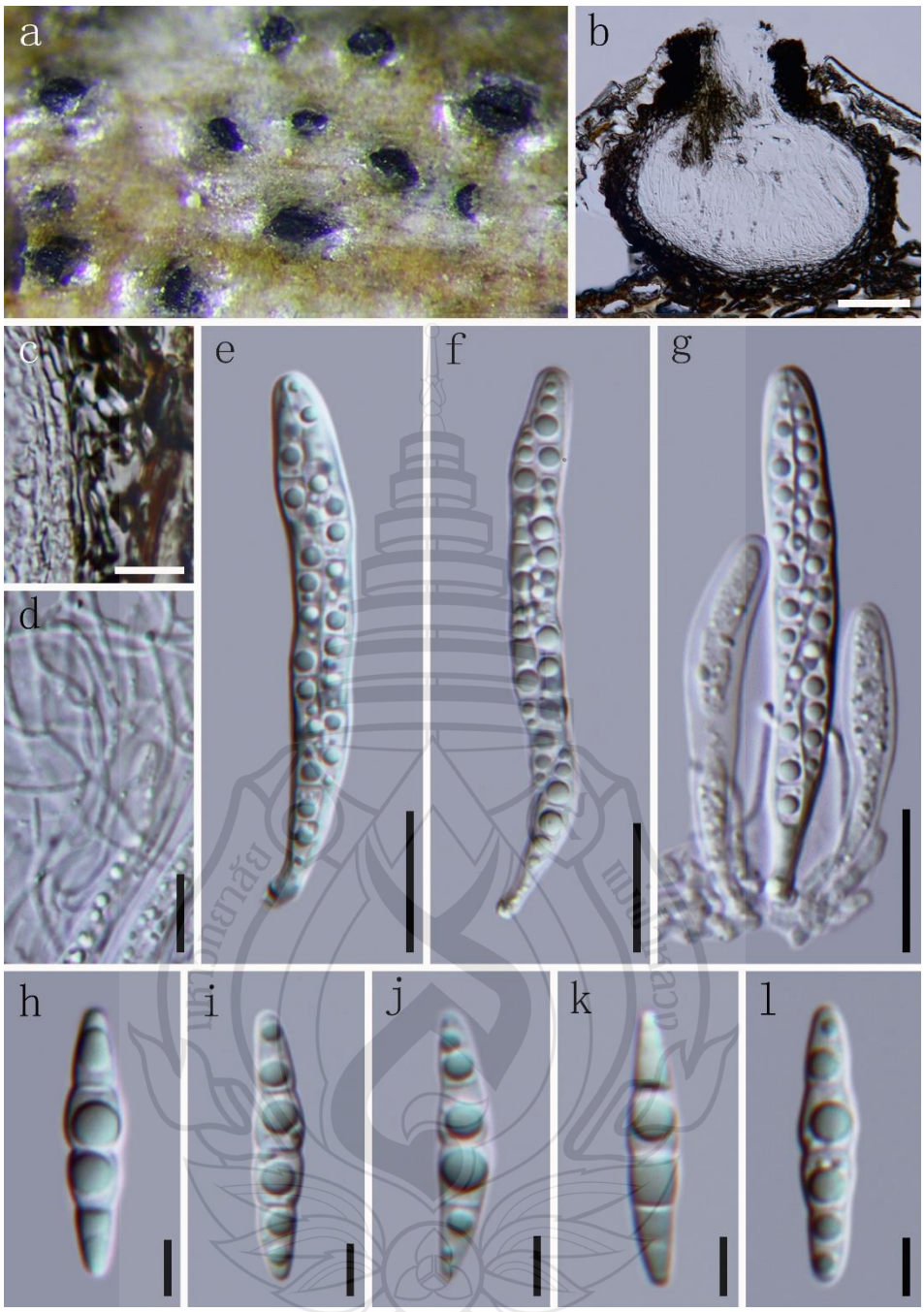
≡ *Lophiotrema hysterioides* (Ellis & Everhart.) Berl., Icon. Pyrenomyc. 1: 4, 1890.

Saprobic on dead branch of unidentified woody plant. Sexual morph: *Ascomata* 182–232 μm high, 205–274 μm diam., dark brown to black, scattered, gregarious, immersed to erumpent throughout the host tissue, globose to subglobose, uni-loculate, glabrous, coriaceous, ostiolate. *Ostioles* central, carbonaceous, with a crest-like opening. *Peridium* 15–39 μm thick, composed of several layers of dark brown to black, thin to thick-walled pseudoparenchymatous cells, arranged in a *textura angularis*, slightly thick at the sides and near the apex, and pale inwardly. *Hamathecium* comprising dense, 1–2 μm wide, branched, cellular pseudoparaphyses, anastomosing among and between the asci, embedded in a gelatinous matrix. *Asci* 73–90(–94) × 7.5–9.5 μm ($\bar{x}$ = 84.5 × 8.5 μm, n = 20), 8-spored, bitunicate, fissitunicate, cylindric-clavate, short pedicellate, apically rounded with a minute ocular chamber. *Ascospores* 22.5–24.5(–29) × 4–5 μm ($\bar{x}$ = 24 × 4.7 μm, n = 25), overlapping bi-seriate, hyaline, fusiform, straight or slightly curved, 3-septate, with the primary septum median, normally constricted at median septum, smooth-walled, guttulate. Asexual morph: Undetermined.

Material examined: CHINA, Guizhou Province, Guiyang City, on dead branch of unidentified woody plant, 2 September 2016, H.Y. Ran, SD-2016-8 (MFLU 18-2074, GZAAS 18-0005), living culture GZCC 18-0015.

Notes: This fungus is observed and described as in having clavate asci and hyaline, fusiform, three equidistant septate ascospores, which are similar to *L. neohysterioides* described in Tanaka & Harada (2003), as well as the sizes of asci and ascospores. Although a short branch is shown between these two strains (Figure 2.28), however, the comparison of each gene region between them was carried out, and the results showed that there are only 5 and 4 base differences in ITS and TEF1-α gene

regions respectively, which cannot demarcate them as different species based on the recommendations of Jeewon and Hyde (2016). Therefore, the newly collected taxon is identified as *L. neohysterioides*, and reported as a new record from China.



Note a Ascomata semi-immersed to erumpent throughout the host surface. b Vertical section through ascoma. c Section of peridium. d Pseudoparaphyses. e–g Long cylindrical asci. h–l Ascospores. Scale bars: b= 50 μm, c, d =10 μm, e–g =20 μm, h–l =5 μm.

Figure 2.27 *Lophiotrema neohysterioides* (MFLU 18-2074, new record)

Table 2.1 GenBank accession numbers of sequences used in phylogenetic analysis. New sequences from this study are in bold

Species	Specimen/Strain No.	ITS	LSU	SSU	Rpb2	TEF1-α
Antealophiotrema brunneosporum	CBS 123095	LC194474	LC194340	LC194298	LC194419	LC194382
Aquasubmersa japonica	MAFF 245218	LC061591	LC061586	LC061581	LC194420	LC194383
Aquasubmersa mircensis	MFLUCC 11-0401	JX276954	JX276955	JX276956	---	---
Atrocalyx acutisporus	MAFF 245613	LC194475	LC194341	LC194299	LC194423	LC194386
Atrocalyx asturiensis	CBS 143912	MG912912	MG912912	---	MG912920	MG912916
Atrocalyx bambusae	MFLUCC 10-0558	KX672149	KX672154	KX672159	KX672161	KX672162
Atrocalyx lignicola	CBS 122364	LC194476	LC194342	LC194300	LC194424	LC19438
Crassimassarina macrospora	MAFF 239606	LC194478	LC194344	LC194302	LC194426	LC194389
Cryptoclypeus oxysporus	MAFF 245614	LC194479	LC194345	LC194303	LC194427	LC194390
Cryptoclypeus ryukyuensis	MAFF 245615	LC194481	LC194347	LC194305	LC194429	LC194392
Cryptocoryneum condensatum	MAFF 245367	LC096159	LC194353	LC194311	LC194435	LC096141
Cryptocoryneum japonicum	MAFF 245370	LC096162	LC194356	LC194314	LC194438	LC096144
Cryptocoryneum longicondensatum	MAFF 245374	LC096166	LC194360	LC194318	LC194442	LC096148

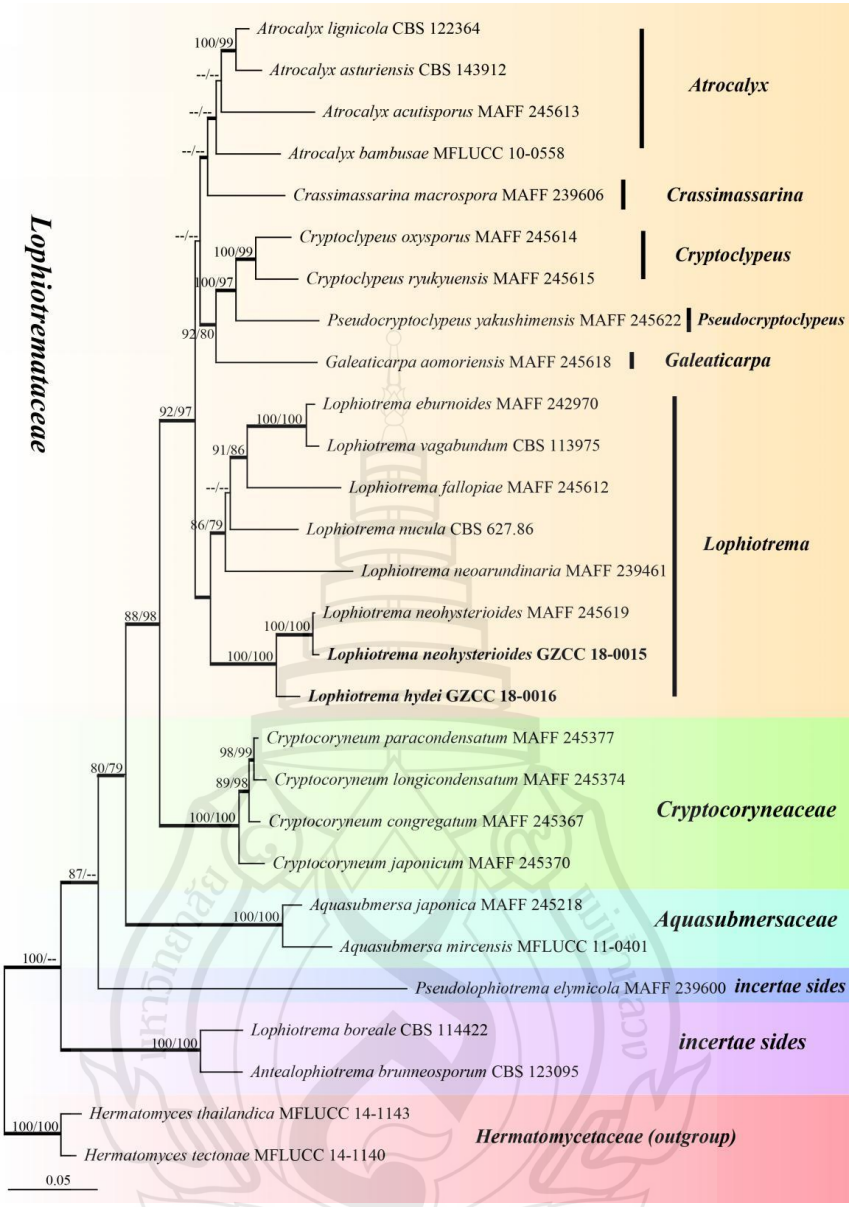
Table 2.1 (continued)

Species	Specimen/Strain No.	ITS	LSU	SSU	Rpb2	TEF1-α
Cryptocoryneum paracondensatum	MAFF 245377	LC096169	LC194363	LC194321	LC194445	LC096151
Galeaticarpa aomori	MAFF 245618	LC194482	LC194366	LC194324	LC194448	LC194393
Hermatomyces tectonae	MFLUCC 14-1140	KU144917	KU764695	KU712465	KU712486	KU872757
Hermatomyces thailandicus	MFLUCC 14-1143	KU144920	KU764692	KU712468	KU712488	KU872754
'*Lophiotrema*' *boreale*	CBS 114422	LC194491	LC194375	LC194333	LC194457	LC194402
Lophiotrema eburneoides	MAFF 242970	LC001709	LC001707	LC001706	LC194458	LC194403
Lophiotrema fallopiae	MAFF 245612	LC149913	LC149915	LC149911	LC194459	LC194404
Lophiotrema hydei	**GZCC 18-0016**	**MK108982**	**MK108978**	**MK108980**	**---**	**MK108984**
Lophiotrema neoarundinaria	MAFF 239461	AB524786	AB524596	AB524455	AB539096	AB539109
Lophiotrema neohysterioides	**GZCC 18-0015**	**MK108983**	**MK108979**	**MK108981**	**---**	**MK108985**
Lophiotrema neohysterioides	MAFF 245619	LC194493	LC194376	LC194334	LC194461	LC194406
Lophiotrema nucula	CBS 627.86	LC194497	AB619021	AB618703	LC194465	LC194410
Lophiotrema vagabundum	CBS 113975	LC194502	AB619025	AB618707	LC194470	LC194415
Pseudocryptoclypeus yakushimensis	MAFF 245622	LC194504	LC194380	LC194338	LC194472	LC194417

Table 2.1 (continued)

Species	Specimen/Strain No.	ITS	LSU	SSU	Rpb2	TEF1-α
Pseudolophiotrema elymicola	MAFF 239600	LC194505	LC194381	LC194339	LC194473	LC194418
Pseudolophiotrema elymicola	MAFF 239600	LC194505	LC194381	LC194339	LC194473	LC194418

Note CBS: CBS-KNAW Fungal Biodiversity Centre, Utrecht, The Netherlands; **GZCC**: Guizhou Culture Collection, Guiyang, China; **MAFF**: Ministry of Agriculture, Forestry and Fisheries, Japan; **MFLUCC:** Mae Fah Luang University Culture Collection, ChiangRai, Thailand.



Note Maximum likelihood bootstrap values equal or greater than 75% are presented above the nodes. The branch with Bayesian posterior probabilities equal or greater than 0.95 are thickened. The tree is generated from maximum likelihood (ML) analysis based on LSU, TEF1-α, SSU, ITS and Rpb2 sequence data, and rooted with *Hermatomyces thailandica* MFLUCC 14-1143 and *H. tectonae* MFLUCC 14-1140.

Figure 2.28 Phylogenetic tree of the family Lophiotremataceae

Melanommataceae G. Winter

Melanommataceae was established by Winter (1885) and is typified by *Melanomma*. Then, a series of mycologists reviewed this family with additions and exclusions of genera in subsequent studies (Barr, 1990a; Lumbsch & Huhndorf, 2007, 2010; Kirk et al., 2008; Hyde et al., 2011, 2013; Wijayawardene et al., 2012; Zhang et al., 2012; Wijayawardene et al., 2014). Tian et al. (2015) reappraised Melanommataceae and accepted 13 genera based on combination of morphological and phylogenetic evidence. Recently, several new genera were introduced and accommodated in this family (Hyde et al., 2018a; Wanasinghe et al., 2018b; Phukhamsakda et al., 2020), and 35 genera are currently recognized in Melanommataceae (Hongsanan et al., 2020a; Wijayawardene et al., 2020).

Herpotrichia Fuckel, Fungi rhenani exsic., suppl., fasc. 7(nos 2101-2200): no. 2171 (1868)

Herpotrichia was established by to accommodate two species *H. rhenana* and *H. rubi* but without designation the type. Subsequently, the argument of generic type was discussed in Bose (1961), and until Cannon (1982) introduced and assigned *H. herpotrichoides* as the type species. There are around 110 epithets listed in Index Fungorum (2022), but with only a few species having available sequence data in GenBank. Tian et al. (2015) reported the latest novel species *H. vaginatispora*, and suggested that the sequences data for the type species *H. herpotrichoidesis* is needed to resolve the natural placement of *Herpotrichia* in future study.

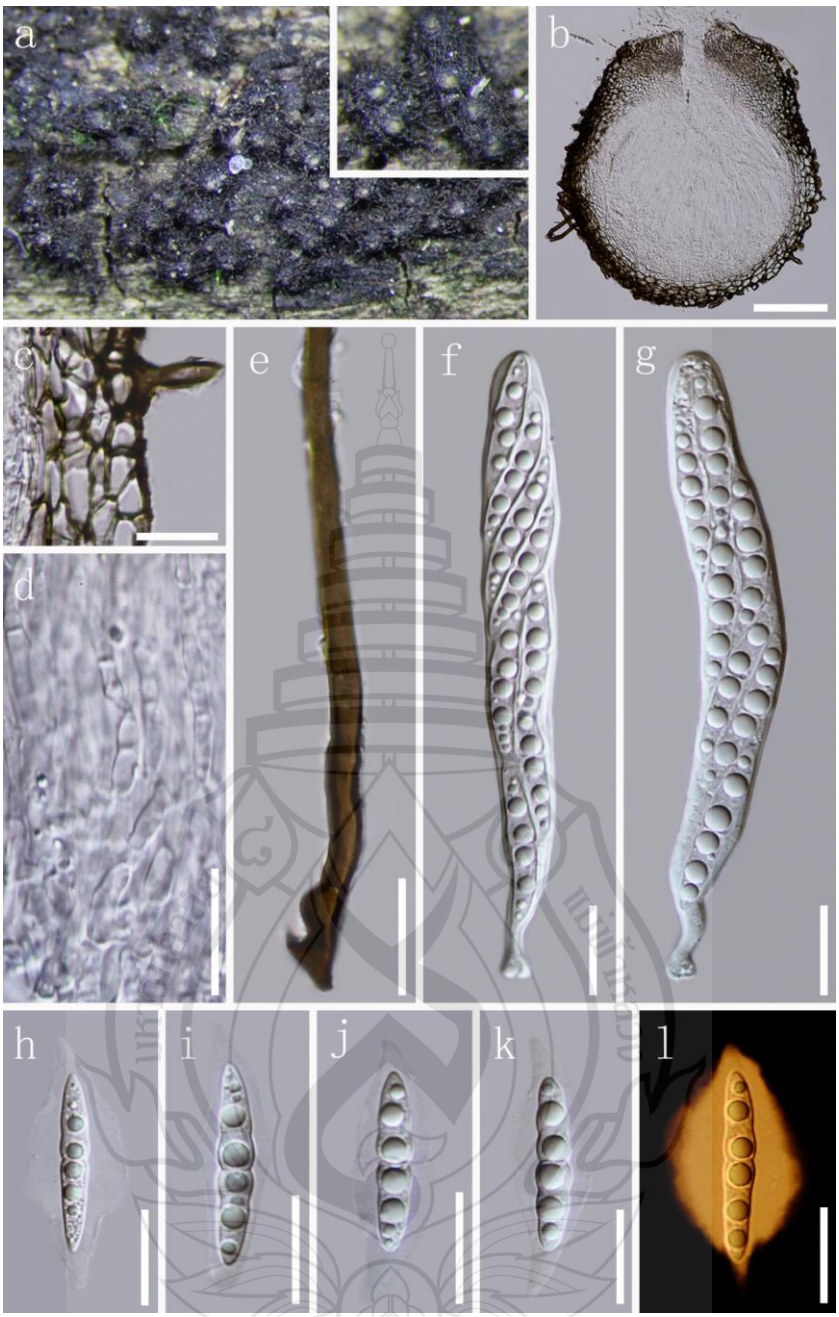
Herpotrichia macrotricha (Berk. & Broome) Sacc., in Saccardo Syll. fung. 2: 213, 1883 Figure 2.29

Saprobic on dead branch of unidentified woody plant. Sexual morph: *Ascomata* solitary or gregarious, globose, semi-immersed to superficial, dark brown to black, coriaceous, ostiolate, associated with numerous, black seta. *Ostiole* single, central, papillate, without periphyses. *Peridium* 32–91 μm, becoming thick towards the apex, comprising 2 strata, outer strata composed of heavily pigmented, thick-walled cells of textura angularis, inner strata comprising light brown, thin-walled cells of *textura angularis*. *Hamathecium* of dense, 1.5–2 μm wide, septate, pseudoparaphyses, branching and anastomosing above the asci, embedded in a gelatinous matrix. *Asci* (116.5–)130.5–140(–153.5) × 14–15.5(–18) μm ($\bar{x}$ = 136 ×

15.5 μm, n = 20), 8-spored, bitunicate, fissitunicate, cylindric-clavate, bulbous pedicellate, apically rounded with a minute ocular chamber. *Ascospores* 34.5–39.5 × 6.5–8 μm ($\bar{x}$ = 37 × 7 μm, n = 20), uni-seriate, partly overlapping or bi-seriate, hyaline, fusoid, ellipsoid or oblong with narrowly rounded ends, 1-septate, deeply constricted at the septum, the upper cell often broader than the lower one, guttulate, smooth-walled, with a structured mucilaginous sheath. Asexual morph: Undetermined.

Material examined: CHINA, Guizhou Province, Leishan County, on dead branch of unidentified woody plant, 2 November 2017, J.F. Zhang LS-12 (GZAAS 19-1756); Maolan County, on dead branch of unidentified woody plant, 8 May 2017, J.F. Zhang ML-5 (GZAAS 19-4019).

Notes: The newly collected specimens are phylogenetically placed within in *Herpotricha*, and clustered to all of the availabe sequences of *H. macrotrhicha* (GKM1128, GKM1193, GKM196N, JCM14419 and SMH269) obtained from GenBank by strong statistical support (Figure 2.6). However, several separate clades appear between this group, and this topology agree with previous studies (Mugambi & Huhndorf, 2009; Tian et al., 2015). Based on the morpholigcal and phylogenetic studies, Mugambi and Huhndorf (2009) indicated that there may be separate species are hidden in *H. macrotrhicha*, but all of the reported specimens should be checked to confirm this decision. The new specimens examined in this study share the most striking features of *H. macrotrhicha* that with cap-like apex ascomata, and the shape as well as the measurements of asci and asospores also fit with the latter that described by different mycologists (Chen & Hsieh, 2004; Tanaka & Hosoya, 2006). Therefore, these two specimens are tentatively identified as *H. macrotrhicha*, and which has been reported widely distributed in temperate regions (Barr, 1984).



Note a Appearance of ascomata in the host surface. b Vertical section through ascoma. c Section of peridium. e Pseudoparaphyses. e Seta. f, g Asci. h–l Ascospores in different maturation stage. Note l is stained with India ink. Scale bars: b = 100 μm, c–j = 20 μm.

Figure 2.29 *Herpotrichia macrotricha* (GZAAS 19-1756)

Occultibambusaceae D.Q. Dai & K.D. Hyde

Occultibambusaceae was established by Dai et al. (2017) to accommodate 4 genera (*Neooccultibambusa*, *Occultibambusa*, *Seriascoma* and *Versicolorisporium*) with *Occultibambusa bambusae* as the type species. And this treatment was also followed by Wijayawardene et al. (2018). Phookamsak et al. (2019) added another genus *Brunneofusispora* within this family, and all of the above five genera were included in Occultibambusaceae in the latest outline of fungi (Wijayawardene et al., 2022).

Occultibambusa D.Q. Dai & K.D. Hyde, Fungal Diversity: 10.1007/s13225-016-0367-8, [26] (2016)

Occultibambusa was introduced by Dai et al. (2017) to accommodate 3 species viz. *O. bambusae, O. fusispora* and *O. pustula* with the first as the type species. With the updates accounts of this genus in recent studies (Hyde et al., 2016; Zhang et al., 2017b; Jiang et al., 2021b), nine species were accepted in *Occultibambusa*.

Occultibambusa jonesii J.F. Zhang, J.K. Liu, K.D. Hyde & Z.Y. Liu, *sp. nov.*

Index Fungorum number: IF 552743; Faces of fungi number: FoF 02873; Figure 2.30

Etymology: Named in honor of E.B. Gareth Jones for his contributions to tropical mycology.

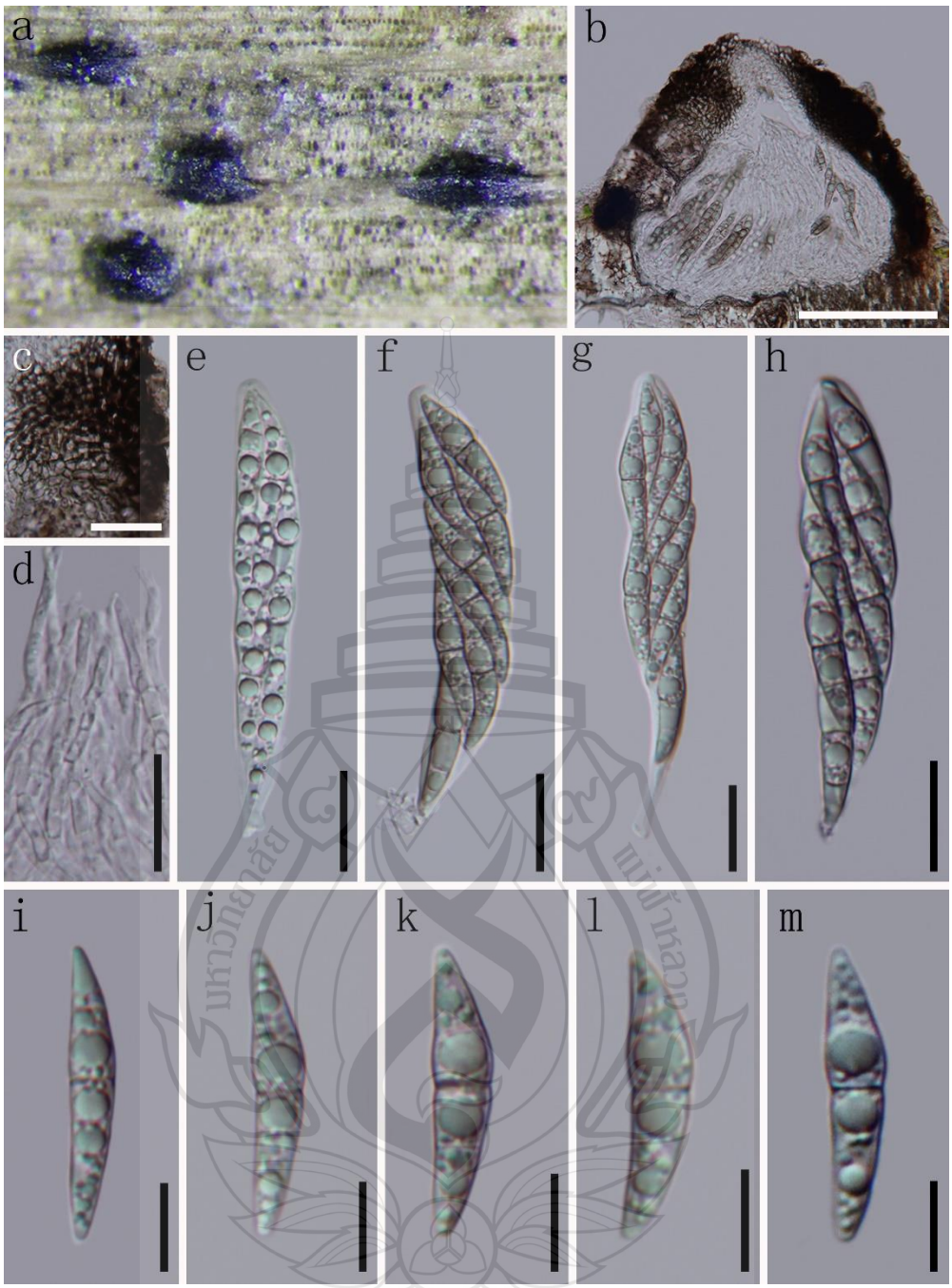
Holotype: GZAAS 16-0162.

Saprobic on dead bamboo culms, forming dark, raised spots on the host surface. Sexual morph: *Ascostromata* 196–236 µm high, 200–260 µm diam., immersed to erumpent, solitary to gregarious, subglobose, ostiolate, papillate, coriaceous, flattened at the base. *Peridium* up to 10–52 µm wide, thin at the base and becoming wider laterally, composed of several layers of dark brown cells, arranged in a *textura angularis*, and the outermost layer intermingled with host tissue. *Hamathecium* comprising dense, 2–3 µm wide, pseudoparaphyses, which anastomose above and between the asci, embedded in a gelatinous matrix. *Asci* (65–)75–89(–105) × 13.5–19 µm ($\bar{x}$ = 85 × 16.5 µm, n = 20), 8-spored, bitunicate, fissitunicate, broadly cylindrical to clavate, short pedicellate, apically rounded to truncate, with an ocular chamber. *Ascospores* 27–33.5 × 5.5–6.5 µm ($\bar{x}$ = 29.5 × 6 µm, n = 20), 1–3-seriate,

2-celled, constricted at the septum, and the upper cell swollen near the septum, inequilateral-fusiform, slightly curved, hyaline and guttulate when young and becoming brown to grayish when mature, wall smooth, without any mucilaginous sheath and appendages. Asexual morph: Undetermined.

Material examined: CHINA, Guizhou Province, Maolan Town, on dead bamboo culms, 20 July 2016, J.F. Zhang, MLC'-12, (GZAAS 16-0162, holotype); ex-type living culture, GZCC 16-0117; *Ibid*., 10 November 2016, J.F Zhang (HKAS96379); living culture KUMCC 17-0136.

Notes: *Occultibambusa jonesii* is phylogenetically close to *O. aquatica* (Figure 2.33), but can be distinguished from it by its larger asci (65– 105 × 13.5– 19 µm vs. 73–86 × 9– 13 µm), longer ascospores (27–33.5 µm vs. 19–25 µm), and the new taxa also lacks a mucilage sheath surrounding the ascospores.



Note a Appearance of ascostromata on dead bamboo culms. b Vertical section through ascostroma. c Section through peridium. d Pseudoparaphyses. e–h Asci with ascospores. i–m Ascospores. Scale bars: b = 100 µm, c = 30 µm, d– h = 20 µm, i–m = 10 µm.

Figure 2.30 *Occultibambusa jonesii* (GZAAS 16-0162, holotype)

Occultibambusa maolanensis J.F. Zhang, J.K. Liu, K.D. Hyde & Z.Y. Liu, *sp. nov.*

Index Fungorum number: IF 552744; Faces of fungi number: FoF 02874; Figure 2.31

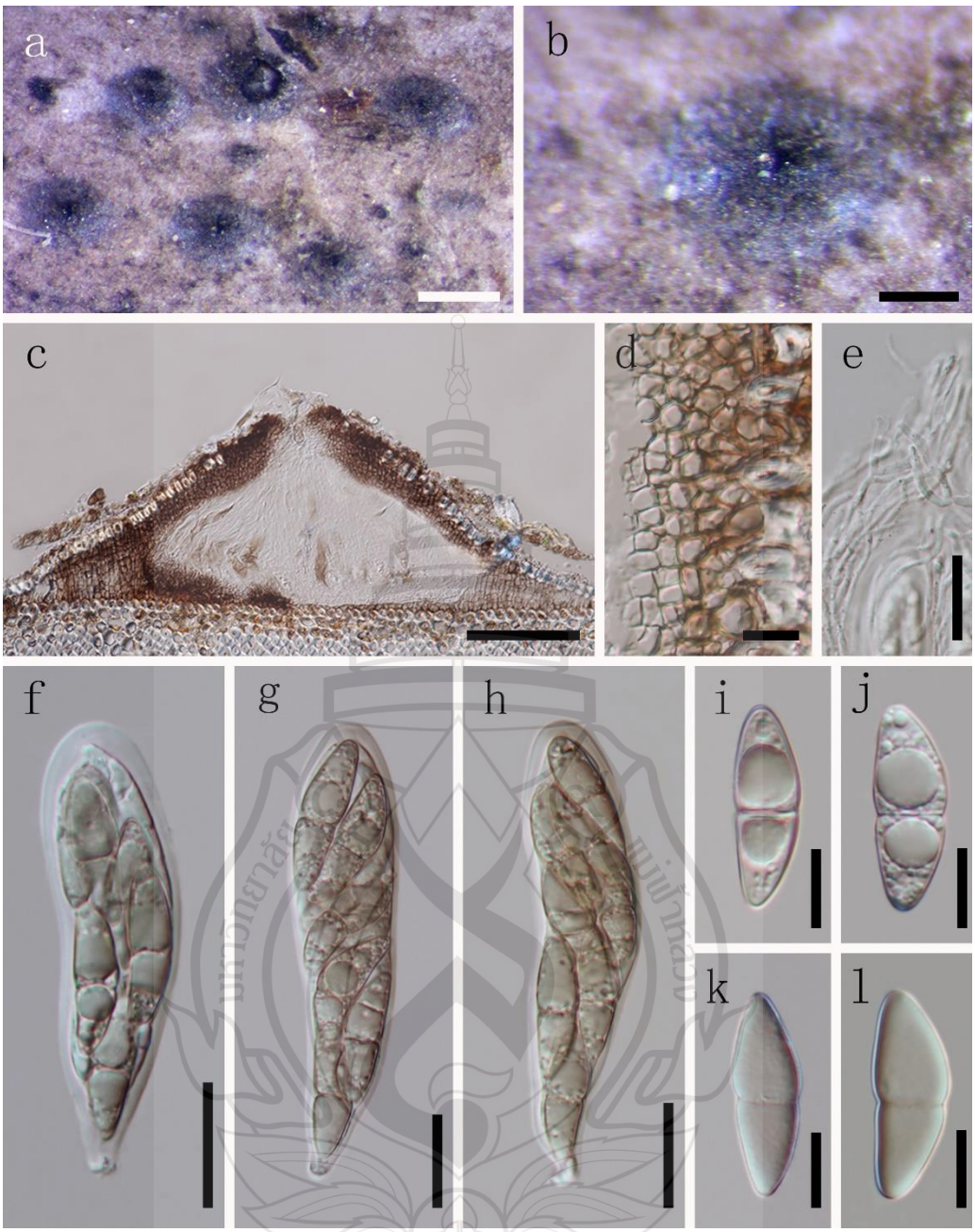
Etymology: Refers to the holotype was collected from Maolan Town.

Holotype: GZAAS 16-0161.

Saprobic on dead bamboo culms, forming dark, rounded spots on the host surface. Sexual morph: *Ascostromata* 544–600 µm diameter, solitary to gregarious, immersed under the epidermis, subglobose, coriaceous, slightly conical in vertical section, and flattened at the base, ostiolate, with a short, rounded, shiny, black papilla. *Peridium* up to 20–42 µm laterally composed of several layers of brown cells, becoming thin-walled and hyaline towards the centrum, arranged in a *textura angularis*, thick and darkly pigmented around ostiole, intermingled with host tissue. *Hamathecium* comprising dense, 1.5–2.4 µm wide, hypha-like pseudoparaphyses, branched and swollen towards the terminal cells, anastomosing above and between the asci, embedded in a gelatinous matrix. *Asci* (66–)77–85(–94) × 17–20(–24) µm ($\bar{x}$ = 81 × 20 µm, n = 20), 8-spored, bitunicate, fissitunicate, broadly cylindrical to clavate, short pedicellate, apically rounded to truncated with a visible ocular chamber (2.5–3.5 µm wide). *Ascospores* 25–31 × 8–10 µm ($\bar{x}$ = 28 × 9 µm, n = 30), 2–4-seriate, 2- celled, and moderately constricted at the septum, inequality-fusiform, apical cells 14–18 µm, basal cells 11–15 µm, slightly curved, hyaline and guttulate when young and become light brown when mature, wall smooth, without any mucilaginous sheath and appendages. Asexual morph: Undetermined.

Material examined: CHINA, Guizhou Province, Maolan Town, on dead bamboo culms, 8 July 2015, J.F. Zhang, MLC-29, (GZAAS 16-0161, holotype); ex-type living culture, GZCC 16- 0116; *Ibid*., 10 November 2016, J.F Zhang (HKAS96380); living culture KUMCC 17-0137.

Notes: *Occultibambusa maolanensis* clusters to *O. fusispora* in a well-supported clade in the phylogenetic analysis (Figure 2.33), but they can be distinguished readily by the difference in appearance of ascostromata, the wider asci (17–24 µm vs. 11–16 µm) and larger ascospores (25–31 × 8–10 µm vs. 20–26 × 5–6.5 µm).



Note a Appearance of ascostromata on dead bamboo culms. b Rounded, shiny black papilla. c Vertical section through ascostroma. d Section through peridium. e Pseudoparaphyses. f–h Asci with ascospores. i–l Ascospores. Scale bars: a = 500 µm, b = 200 µm, c = 100 µm, d = 10 µm, e-n = 20 µm.

Figure 2.31 *Occultibambusa maolanensis* (GZAAS 16-0161, holotype)

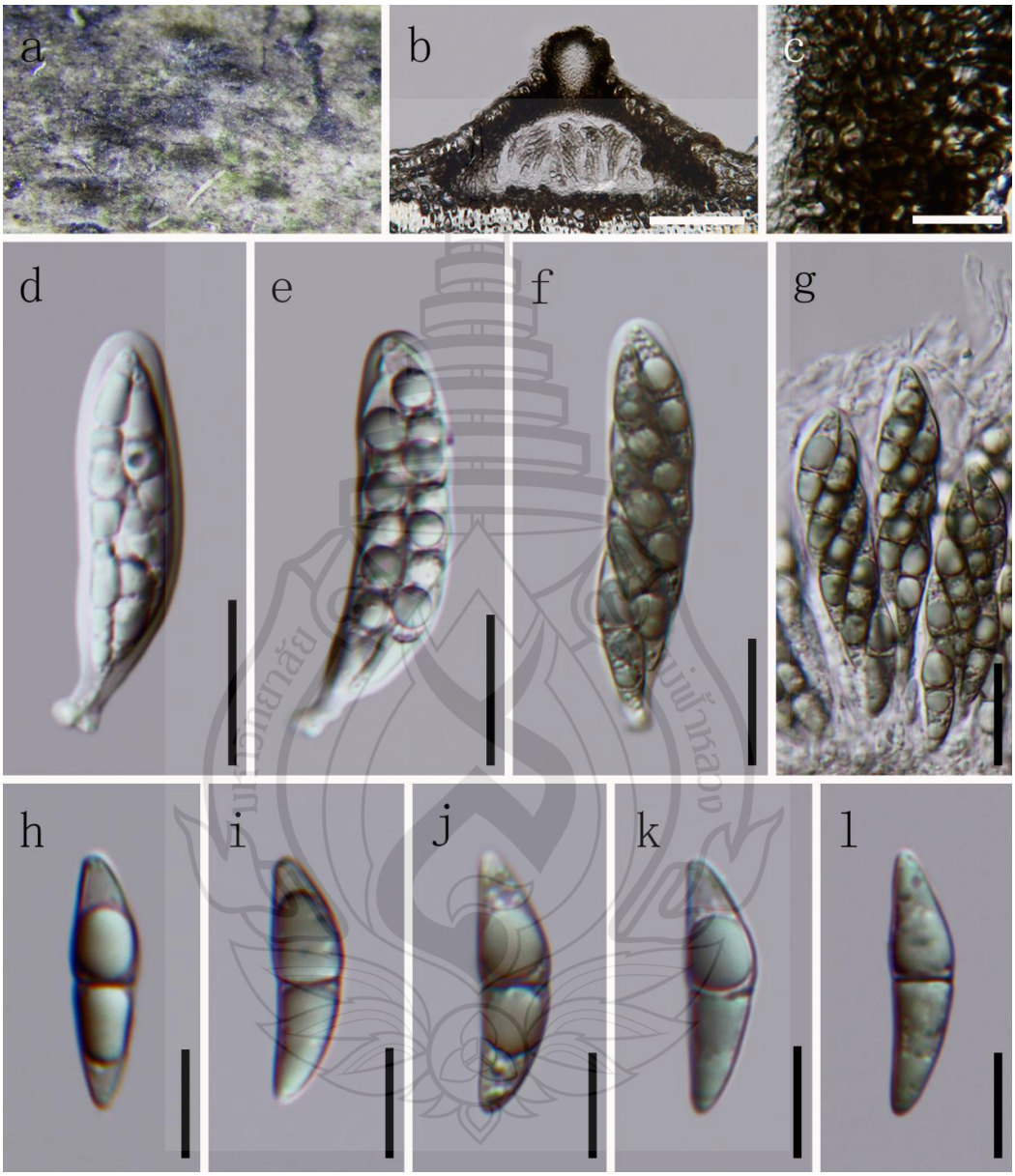
Occultibambusa pustula D.Q. Dai & K.D. Hyde, Fungal Diversity 82: 30 (2017) *new record*. Figure 2.32

Saprobic on dead bamboo culms, forming black, rounded, crest-like spots on host surface. Sexual morph: *Ascostromata* immersed, subepidermal, slightly conical, black, with a centrally located ostiole. *Peridium* 22–45 μm, composed of inner layer of hyaline to brown, thick-walled cells of *textura angularis* to *primastica*, and outer layer of black, irregular cells that confused with host epidermis. *Hamathecium* compring of 2–2.5 μm wide, septate, cellular pseudoparaphyses, anastomosing between and above the asci. *Asci* (50.5–)57–62.5(–68) × 13–15.5 μm ($\bar{x}$ = 60.5 × 13.5 μm, n = 30), 8-spored, bitunicate, cylindrical, with a short furcate pedicel, apically round, with a distinct ocular chamber. *Ascospores* 22–25.5 × 6–7 μm ($\bar{x}$ = 24 × 6.5 μm, n = 35), bi- to tri-seriate, slightly broad-fusiform, hyaline and non-septate when young, become olive-greenish and 1-septate at maturity, slightly constricted at the septum, upper cell is wider than the lower, wall smooth. Asexual morph: Undetermined.

Material examined: CHINA, Guizhou Province, Libo County, Maolan Town, Maonlan National Nature Reserve, on dead bamboo culms, 18 July 2016, J.F. Zhang MLAA-1 (MFLU 18-2273).

Notes: Present phylogenetic analysis shows that our new collection clusters with *O. pustula* by strong bootstrap support (Figure 2.6). However, the morphological characteristics of this specimen are not particularly similar to the holotype of *O. pustula*, this is attribute to that the holotype is immature (Dai et al., 2016). Dong et al. (2020) recollected a mature specimen of *O. pustula* from Thailand and emended its descriptions, but it differs from our collection in having larger sized asci and more septate ascospores. Nevertheless, this could not enough to warrant a new species as they are collected from different habitat, which may slightly affect the morphological features of the same species. In addition, comparison of each gene region between our collection and two reported collections (MFLUCC 11-0502 and MFLUCC 18-1028) of *O. pustula*, and the results showed that there are only 4 bp and 1 bp base pair difference in ITS between MFLU 18-2273 and MFLUCC 11-0502, as well as MFLU 18-2273 and MFLUCC 18-1028, respectively. Thus, the new collection in this study is identified as *O. pustula*, and the measurement of asci is expanded to

(50.5–)57–105(–125) 8–15.5 µm, based on our observation and previous studies (Dai et al., 2016; Dong et al., 2020).



Note a Appearance of ascomata in the host surface. b Vertical section through ascoma. c Section of peridium. d–f Asci in different maturation stage. g Pseudoparaphyses anastomosed between asci. h–l Ascospores in different maturation stage. Scale bars: b = 100 µm, c–g = 20 µm, h–l = 10 µm.

Figure 2.32 *Occultibambusa pustula* (MFLU 18-2273, new record)

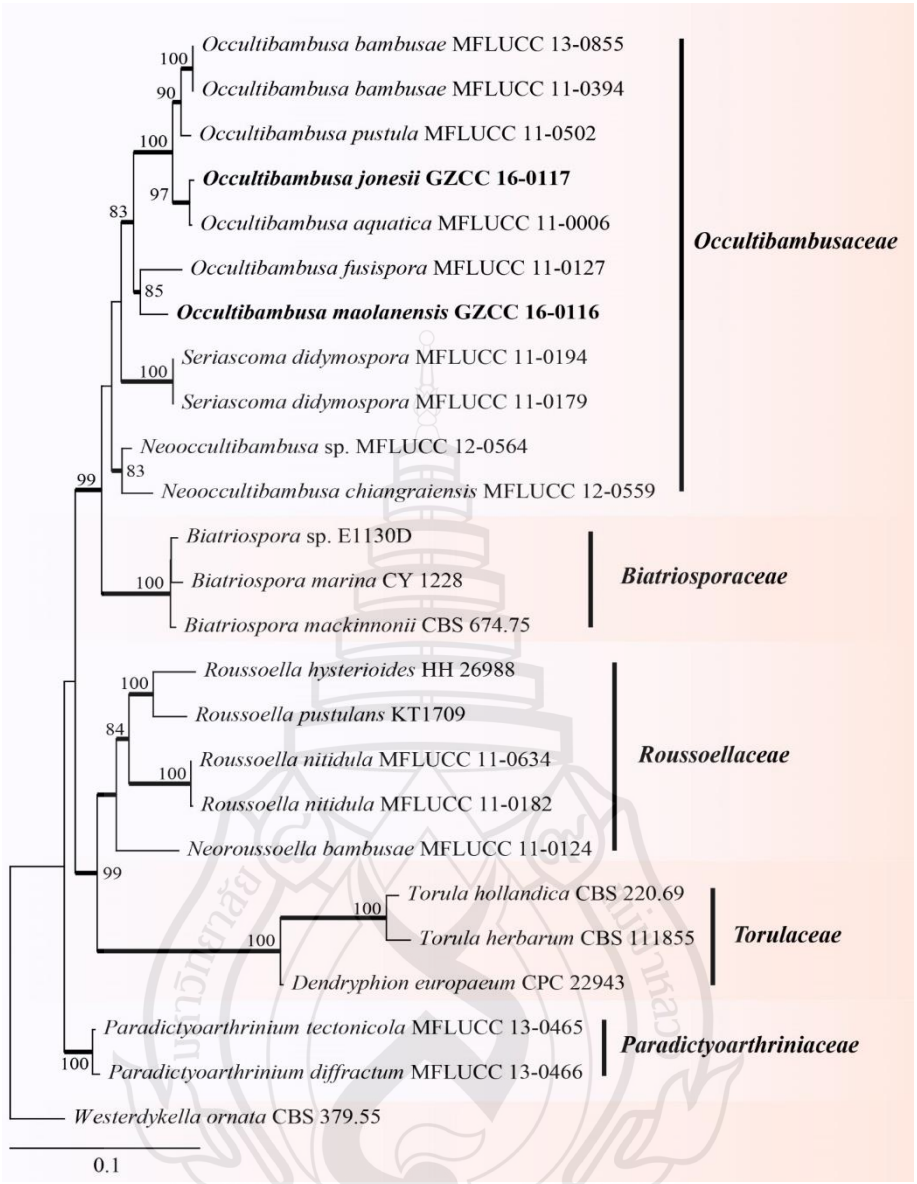
Table 2.2 GenBank accession numbers of sequences used in phylogenetic analysis. New sequences from this study are in bold

Species	Specimen/Strain No.	LSU	SSU	TEF1-α	Rpb2
Biatriospora mackinnonii	CBS 674.75	KF015612	GQ387552	KF407986	KF015703
Biatriospora marina	CY 1228	GQ925848	GQ925835	GU479848	GU479823
Biatriospora sp.	CCF 4485	LN626683	LN626677	LN626671	LN626663
Dendryphion europaeum	CPC 22943	KG869203	---	---	---
Neooccultibambusa chiangraiensis	MFLUCC 12-0559	KU764699	KU712458	---	---
Neooccultibambusa sp.	MFLUCC 12-0564	---	---	KU872761	---
Neoroussoella bambusae	MFLUCC 11-0124	KJ474839	---	KJ474848	KJ474856
Occultibambusa aquatica	MFLUCC 11-0006	KX698110	KX698112	---	---
Occultibambusa bambusae	MFLUCC 11-0394	KU863113	KU872117	KU940194	KU940171
Occultibambusa bambusae	MFLUCC 13-0855	KU863112	KU872116	KU940193	KU940170
Occultibambusa fusispora	MFLUCC 11-0127	KU863114	---	KU940195	KU940172
Occultibambusa jonesii	GZCC 16-0117	KY628322	KY628324	KY814756	KY814758
Occultibambusa maolanensis	GZCC 16-0116	KY628323	KY628325	KY814757	KY814759

Table 2.2 (continued)

Species	Specimen/Strain No.	LSU	SSU	TEF1-α	Rpb2
Occltibambusa pustulata	MFLUCC 11-0502	KU863115	KU872118	---	---
Paradictyoarthrinium diffractum	MFLUCC 13-0466	KP744498	KP753960	---	---
Paradictyoarthrinium tectonicola	MFLUCC 13-0465	KP744500	KP753961	---	---
Roussoella hysterioides	HH 26988	AB524622	AB524481	AB539115	AB539102
Roussoella nitidula	MFLUCC 11-0182	KJ474843	---	KJ474852	KJ474859
Roussoella nitidula	MFLUCC 11-0634	KJ474842	---	KJ474851	KJ474858
Roussoella pustulans	KT 1709	AB524623	AB524482	AB539116	AB539103
Seriascoma didymospora	MFLUCC 11-0179	KU863116	---	KU940196	KU940173
Seriascoma didymospora	MFLUCC 11-0194	KU863117	---	KU940197	KU940174
Torula herbarum	CBS 111855	KF443386	KF443391	KF443403	KF443396
Torula hollandica	CBS 220.69	KF443384	---	---	---
Westerdykella ornata	CBS 379.55	GU301880	GU296208	GU349021	GU371803

Note CBS: Centraalbureau voor Schimmelcultures, The Netherlands; **CPC:** Collection of Pedro Crous housed at CBS; **GZCC:** Guizhou culture collection, Guizhou, China; **MFLUCC:** Mae Fah Luang University Culture Collection, Chiang Rai, Thailand; **KT**: K. Tana



Note Maximum likelihood bootstrap values equal or greater than 75% are presented above the nodes. The branch with Bayesian posterior probabilities equal or greater than 0.95 are thickened. New sequence generated in this study are given in bold. The tree is generated from maximum likelihood (ML) analysis based on LSU, SSU, TEF1-α and Rpb2 sequence data, and rooted with *Westerdykella ornata* CBS 379.55.

Figure 2.33 Phylogenetic tree of the family Occultibambusaceae

Parabambusicolaceae Kaz. Tanaka & K. Hiray

Parabambusicolaceae was established by Tanaka et al. (2015) to accommodate *Aquastroma*, *Multiseptospora* and *Parabambusicola* with the latter as the typus. Subsequently, other 6 genera, viz. *Lonicericola*, *Multilocularia*, *Neoaquastroma*, *Paramonodictys*, *Paratrimmatostroma* and *Pseudomonodictys* were introduced and placed within this family based on morphological and molecular studies (Ariyawansa et al., 2015a; Li et al., 2016; Wanasinghe et al., 2017; Phookamsak et al., 2019; Hyde et al., 2020a).

Neoaquastroma Wanas., E.B.G. Jones & K.D. Hyde, Phytotaxa 302(2): 139 (2017)

Neoaquastroma was introduced by Wanasinghe et al. (2017) to accommodate the type species *N. guttulatum* based on morphological and multi-gene analysis. Then, other 3 species (*N. bauhiniae*, *N. cylindricum* and *N. krabiense*) were subsequently accepted in later studies (Phukhamsakda et al., 2018; Samarakoon et al., 2019). In this study, a new species *N. guizhouense* is introduces hereby based on its morphological and molecular data.

Neoaquastroma guizhouense J.F. Zhang & J.K. Liu, *sp. nov*., Figure 2.34

Etymology: Name refers to the location where the holotype was collected.

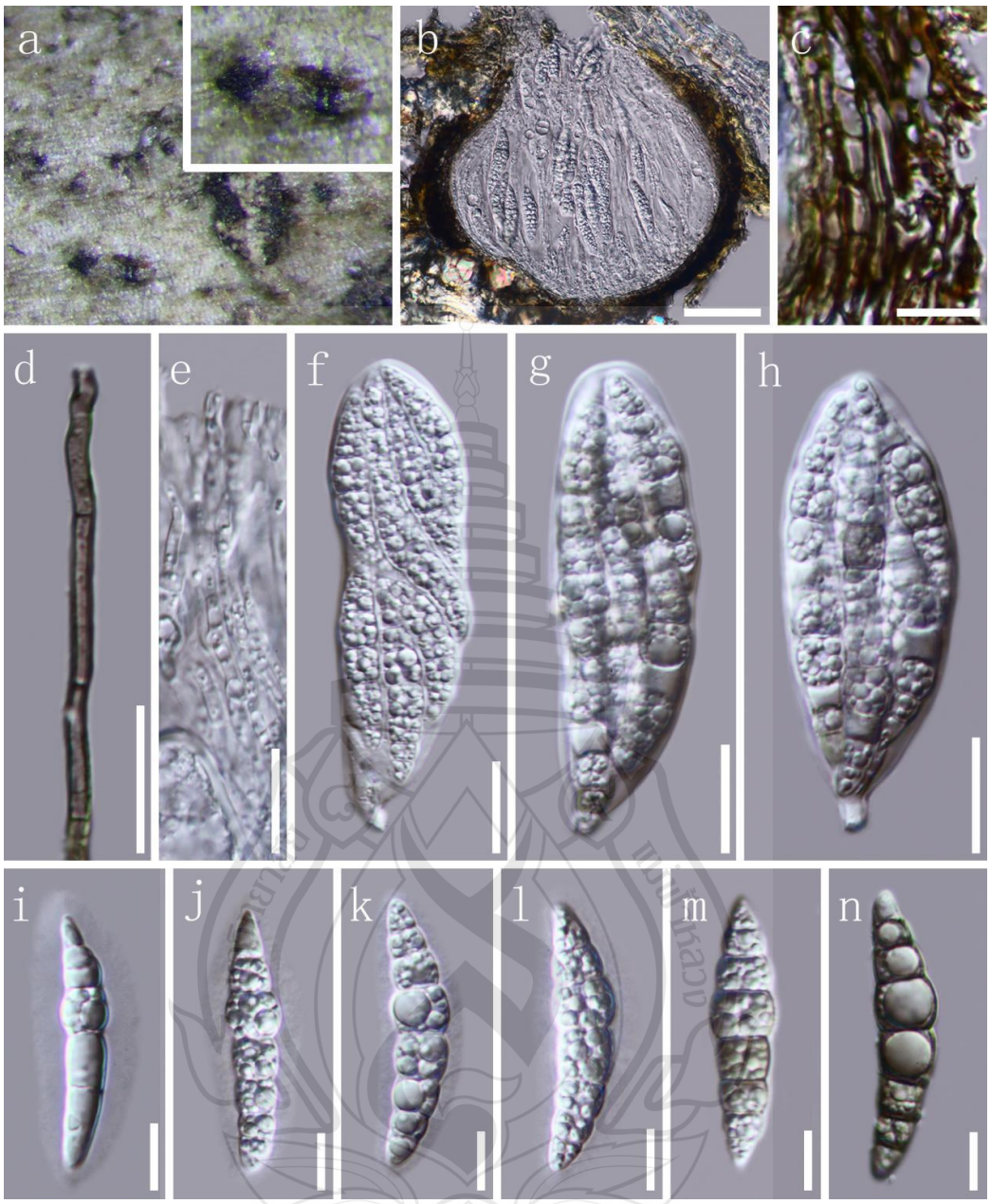
Holotype: GZAAS 19-1761.

Saprobic on dead culm of unidentified herbaceous plant. Sexual morph: *Ascomata* 167–188 μm high × 208–247 μm diam., semi-immersed to immersed, solitary, scattered, subglobose to globose, brown to dark brown, coriaceous, associated with numerous, black, septate seta. *Ostiole* central, short papillate. *Peridium* 15–31 μm wide, composed of two strata, of the outermost stratum with dark brown pseudoparenchymatous cells confusing with host tissue, and an inner stratum consisted of several layers of hyaline to pale brown cells of *textura angularis*. *Hamathecium* composed of dense, 1.5–2.5 μm wide, filamentous, trabeculate psedoparaphyses, anastomosing between and above the asci, embedded in a gelatinous matrix. *Asci* (74–) 91–95 (–101.5) × 25.5–30 (–32.5) μm ($\bar{x}$ = 86 × 29 μm, n = 20), 8-spored, bitunicate, fissitunicate, oboviod to oblong, with a very short pedicel, apically rounded, and a minute ocular chamber is presented. *Ascospores* 38–46 (–51) × 9–11.5 μm ($\bar{x}$ = 43.5 × 10.2 μm, n = 35), 2–4-seriate, broadly

fusiform, narrow towards the ends, initially hyaline, 1–3-septate, and surrounded by a 2.5–5 μm wide mucilaginous sheath, become brown to dark brown, 5–7-septate, and the mucilaginous sheath disappeared at maturation, slightly constricted at the septa, with cell above central septum wider, guttulate, verrucose. Asexual morph: Undetermined.

Material examined: CHINA, Guizhou Province, Meitan County, dead culm of unidentified herbaceous plant, 8 August 2017, J.F. Zhang MT-3 (GZAAS 19-1761, holotype).

Notes: *Neoaquastroma guizhouense* is phylogenetically assigned into *Neoaquastroma* and closed to *N. bauhiniae* with strong statistical support (Figure 2.6). Morphologically, *N. guizhouense* is somewhat typical of *Neoaquastroma* in having obvoid to cylindrical, short pedicellate asci and fusiform, multi-septate ascospores that surrounded by a thick mucilaginous sheath. However, one of the striking features of *N. guizhouense* is that numerous black, septate seta associated with ascomata, while this trait has never been described in any of the known *Neoaquastroma* taxa in previous studies (Wanasinghe et al., 2017; Phukhamsakda et al., 2018; Samarakoon et al., 2019). *Neoaquastroma guizhouense* resembles to *N. bauhiniae* by having overlap sized asci and ascospores (Phukhamsakda et al., 2018), nevertheless, the comparison results of each gene region (1/837, 11/530 and 27/973 base pair differences in LSU, ITS and TEF, respectively) between them indicate that they are separate species, based on the recommendations to delineate species boundaries from Jeewon and Hyde (2016).



Note a Appearance of ascomata in the host surface. b Vertical section through ascoma. c Section of Peridium. d Seta projecting from peridium. e Pseudoparaphyses. f–h Young and mature asci. i–d Young and mature ascospores. Scale bars: b = 50 μm, c, i–n = 10 μm, d–h = 20 μm.

Figure 2.34 *Neoaquastroma guizhouense* (GZAAS 19-1761, holotype)

Paraleptosphaeriaceae J.F. Zhang & K.D. Hyde, *fam. nov.*

Etymology: Referring to the name of the type genus.

Type genus: *Paraleptosphaeria* J.F. Zhang & K.D. Hyde.

Saprobic on dead twigs of unidentified woody plant. Sexual morph: *Ascomata* semi-immersed to superficial, black, scattered, or gregarious, globose to subglobose. *Ostiole* central, broadly papillate. *Peridium* composed of multi-layered scleroplectenchymatous cells, normally arranged in a *textura angularius*. *Hamathecium* comprising numerous, non-trabeculate, branched pseudoparaphyses anastomosing between and above the asci, embedded in a gelatinous matrix. *Asci* bitunicate, fissitunicate, broad cylindrical to clavate, with a furcate pedicel, apically rounded to truncate, with an ocular chamber. *Ascospores* fusiform, hyaline, bi- to tri-seriate, septate, with or without mucilaginous sheath. Asexual morph: Undetermined.

Notes: Paraleptosphaeriaceae is typical of Pleosporales in having perithecioid ascomata with bitunicate, fissitunicate asci bearing mostly septate ascospores. This family resembles to Leptosphaeriaceae by its semi-immersed to superficial, globose to subglobose ascomata with broadly papilla in face view, cylindrical to clavate asci with a furcate asci and oucular chamber, as well as fusoid, septate ascospores. However, the ascospores of most taxa in Leptosphaeriaceae are multi-septate and pigmented, while the new taxon of Paraleptosphaeriaceae is observed with asymmetrical, one-septate, hyaline ascospores. Phylogenetically, Paraleptosphaeriaceae forms an independent clade and distant from Leptosphaeriaceae. Based on the above evidence, Paraleptosphaeriaceae is introduced to accommodate the type genus *Paraleptosphaeria* in the present study.

Paraleptosphaeria J.F. Zhang & K.D. Hyde, *gen. nov.*

Etymology: The generic epithet "*Paraleptosphaeria*" refers to the resemblance of *Leptosphaeria.*

Type species: *Paraleptosphaeria guizhouensis* J.F. Zhang & K.D. Hyde.

Saprobic on dead twigs of unidentified woody plant. Sexual morph: *Ascomata* semi-immersed to superficial, black, scattered, solitary or gregarious, globose to subglobose, glabrous. *Ostiole* central, broadly papillate. *Peridium* comprised of several layers of pale brown to hyaline cells of *textura angularius*. *Hamathecium*

comprising numerous, non-trabeculate, branched pseudoparaphyses anastomosing between and above the asci, embedded in a gelatinous matrix. *Asci* 8-spored, bitunicate, fissitunicate, broad cylindrical to clavate, with a furcate pedicel, apically rounded to truncate, with a developed ocular chamber. *Ascospores* fusiform, hyaline, one septate, guttulate and surrounded by an indistinctly mucilaginous sheath when young, verrucose at maturity. Asexual morph: Undetermined.

Notes: *Paraleptosphaeria* is established as a monotypic genus with *P. guihzouensis* as the type species. Morphologically, it resembles to *Leptosphaeria* by its semi-immersed to superficial, subglobose ascomata with broadly conical papilla, cylindrical to clavate asci with a furcate pedicel (Ariyawansa et al., 2014). However, it obviously differs from *Leptosphaeria* in having one-septate, hyaline, fusoid ascospores, whereas the taxa in *Leptosphaeria* reproduce 3-septate, narrowly fusoid, pigmented ascospores (Ariyawansa et al., 2014). Moreover, these two genera are phylogenetically distant in the present study.

Paraleptosphaeria guizhouensis J.F. Zhang & K.D. Hyde, *sp. nov*., Figure 2.35

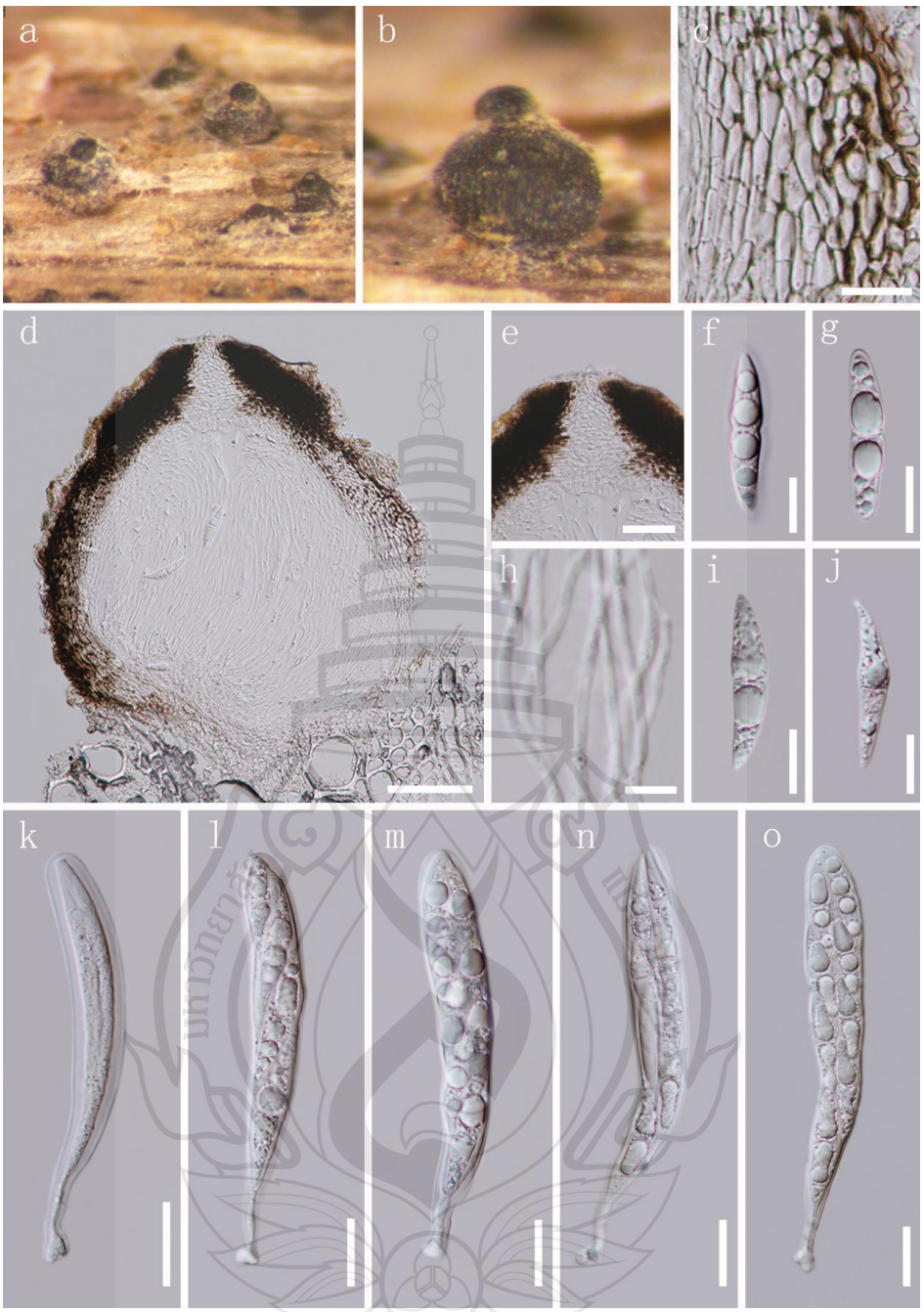
Etymology: Named "*guizhouensis*" refers to where the holotype was collected.

Holotype: GZAAS 19-4017.

Saprobic on dead branch of unidentified woody plant. Sexual morph: *Ascomata* 210–436 µm high (including neck), 186–433 µm diam ($\bar{x}$ = 306 × 329 µm, n = 10), semi-immersed to superficial, black, scattered, solitary, globose to subglobose, glabrous, ostiole central, broadly papillate, normally some host fragments adherently. *Peridium* up to 18–45(–67) µm wide, thin and hyaline on the base, becoming thick and dark pigment around ostiole, composed of two strata, an outer stratum of thick-walled, dark brown cells confusing with substrate, and an inner stratum of thin-walled, pale brown to hyaline cells arranged in a *textura angularius*. *Hamathecium* comprising numerous, non-trabeculate, branched pseudoparaphyses, 2–3 µm wide, anastomosing between and above the asci, embedded in a gelatinous matrix. *Asci* (180–) 188–208 (–228) × 23–28 (–31) µm ($\bar{x}$ = 198 × 26 µm, n = 20), 8-spored, bitunicate, fissitunicate, clavate, pedicellate with a furcate pedicel, apically rounded to truncate, with a developed ocular chamber. *Ascospores* (52–) 56–60 × 11–13 µm ($\bar{x}$ = 58 × 12 µm, n = 40), fusiform, guttulate, one septate and slightly

constricted at the septum, asymmetrical, the upper cell is wider than the basal one, guttulate and surrounded by an indistinctly mucilaginous sheath when young, verrucose at maturity. Asexual morph: Undetermined.

Material examined: CHINA, Guizhou Province, Xiyi City, on dead branch of unidentified woody plant, 8 July 2015, J.F. Zhang XY 5-2 (GZAAS 19-4017); Xiyi City, on dead branch of unidentified woody plant, 8 July 2015, J.F. Zhang XY 5-3 (GZAAS 19-4018).



Note a, b Appearance of ascomata in host surface. c Section of peridium. d Vertical section through ascoma. e Ostile with paraphysis. f, g, i, j Yong and mature ascospores. h Pseudoparaphyses. k–o Young and mature asci. Scale bars: c = 20 µm, d = 100 µm, e = 10 µm, f–j = 20 µm, k–o = 30 µm.

Figure 2.35 *Paraleptosphaeria guizhouensis* (GZAAS 19-4017, holotype)

Phaeosphaeriaceae M.E. Barr

Phaeosphaeriaceae was introduced and typified by *Phaeosphaeria* with *P. oryzae* as the type species (Barr, 1979a). Then, this family was reviewed by a series of mycologists in the next few decades, and different accounts were included (Barr, 1992; Eriksson & Hawksworth, 1993; Kirk et al., 2001, 2008; Lumbsch & Huhndorf, 2007, 2010; Hyde et al., 2011, 2013; Zhang et al., 2012; Wijayawardene et al., 2014b). Phookamsak et al. (2014) provided a systematic revision of Phaeosphaeriaceae based on morphological examination and molecular analysis, and accepted 28 genera within this family. Recently, lots of new and reported genera were accommodated in this family (Ariyawansa et al., 2015a; Crous et al., 2015a, 2017, 2019a; Li et al., 2015c; Liu et al., 2015; Phukhamsakda et al., 2015c, 2020; Abd-Elsalam et al., 2016; Hyde et al., 2016, 2017, 2020a; Tennakoon et al., 2016a, 2020; Tibpromma et al., 2016; Wijayawardene et al., 2016; 2018; Karunarathna et al., 2017; Phookamsak et al., 2017, 2019; Thambugala et al., 2017; Bakhshi et al., 2018; Wanasinghe et al., 2018b; Devadatha et al., 2019; Maharachchikumbura et al., 2019; Marin-Felix et al., 2019; Yang et al., 2019; Zhang et al., 2019; Karunarathna et al., 2020; Mapook et al., 2020), and 83 genera are currently listed under Phaeosphaeriaceae.

Hydeomyces Maharachch., H.A. Ariyaw., Wanas. and Al-Sadi., Phytotaxa 391(1): 33 (2019)

Hydeomyces was established by Maharachchikumbura et al. (2019) to accommodate the type species *Hydeomyces desertipleosporoides*. This genus is morphologically characterized by having globose-depressed, brown to black ascomata, cylindrical, fissitunicate asci, ellipsoidal to subfusiform, muriform ascospores, and phoma-like asexual morphs.

Hydeomyces pinicola J.F. Zhang, J.K. Liu & Z.Y. Liu, *sp. nov.*

Index Fungorum number: IF 556249; Facesoffungi Number: FoF05839; Figure 2.36

Etymology: Refers to the fungus being isolated from the dead wood of *Pinus* sp.

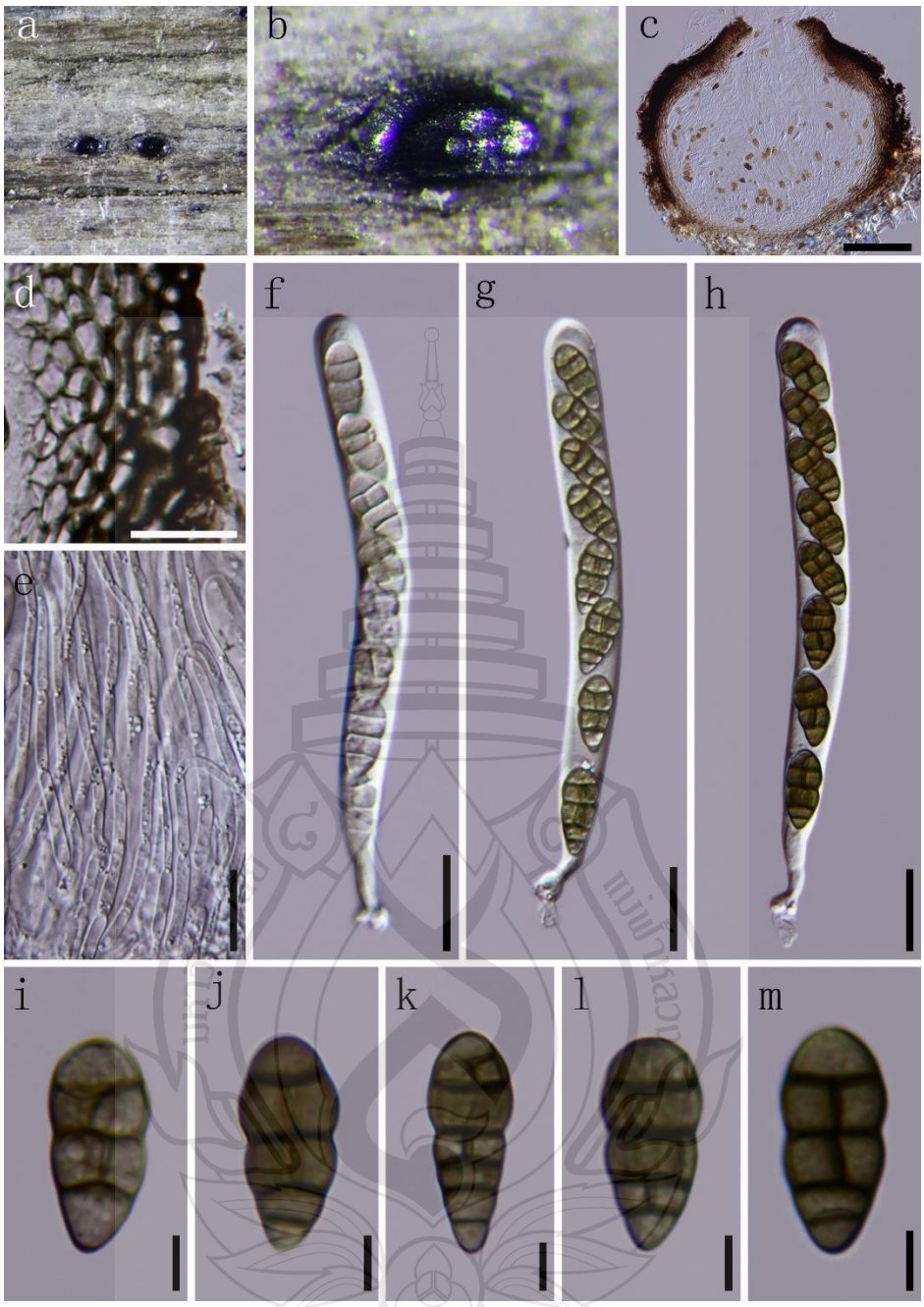
Holotype: MFLU 18-2289.

Saprobic on dead wood of *Pinus* sp. Sexual morph: *Ascomata* 185–417 µm high, 210–341 µm wide, scattered, solitary to gregarious, immersed, semi-immersed

to erumpent throughout the host tissue, subglobose, dark brown to black, coriaceous, ostiolate. *Ostiole* central, with a minute, broadly, flattened papilla, and a pore-like opening. *Peridium* up to 16–24 µm at the base, 42–59 µm at the sides, especially thick near the ostiole, comprising two strata, the outer stratum composed of black, thick-walled cells of *textura angularis*, fusing with the host tissues, inner stratum composed of several layers of brown to slightly pigmented, thin-walled cells of *textura angularis*. *Hamathecium* comprising numerous, 2–3.5 µm wide, hypha-like, septate pseudoparaphyses, branched at the apex, anastomosing between and among asci. *Asci* 103–111(–119) × 9–11.5 µm ($\bar{x}$ = 110 × 10 µm, n = 20), 8-spored, bitunicate, fissitunicate, cylindrical, short pedicellate with a furcate pedicel, rounded at the apex, with a minute ocular chamber. *Ascospores* 15.5–19.5 × 7–10 µm ($\bar{x}$ = 17.5 × 8 µm, n = 35), pyriform to subfusiform, muriform, asymmetrical, broader apically and narrower basally, round to truncate at the upper cell, uniseriate, partially overlapping, 3–5- transversely septate, with 0–3 vertical septa, initially hyaline, become greenish-brown at maturation, deeply constricted at the central septum, rounded at the ends, smooth-walled. Asexual morph: Undetermined.

Material examined: CHINA, Guizhou Province, Guiyang City, Huaxi District, Guizhou academy of agricultural sciences, on the dead wood of *Pinus* sp., 25 Septermber 2016, J.F. Zhang, GZ-06 (MFLU 18-2289, GZAAS 18-0009); ex-type living culture (GZCC 19-0003).

Notes: Phylogenetic analyses place the new species *H. pinicola* in the genus *Hydeomyces*, where it is close to the type species *H. desertipleosporoides* with high support (Figure 2.44). Although *H. pinicola* is morphologically similar to *H. desertipleosporoides* in having globose, brown to black ascomata, cylindrical, fissitunicate asci, and subfusiform, muriform ascospores, it can be distinguished from the latter by its larger asci (103–119 µm vs. 70–85 µm) and ascospores (15.5–19.5 × 7–10 µm vs. 10–15 × 5–6.5 µm) (Maharachchikumbura et al., 2019). They are phylogenetically distinct species.



Note a Appearance of ascomata on host surface. b Ascomata with a central, pore-like opening ostiole. c Vertical sections through ascoma. d Section of peridium. e Pseudoparaphyses. f–h Asci. i–m Ascospores. Scale bars: c = 100 µm, d–h = 20 µm, i-m = 5 µm.

Figure 2.36 *Hydeomyces pinicola* (MFLU 18-2289, holotype)

Hydeopsis J.F. Zhang, J.K. Liu & Z.Y. Liu, *gen. nov.*

Index Fungorum number: IF 556247; Facesoffungi Number: FoF 05837

Etymology: Named in honour of Prof. Kevin D. Hyde for his immense contribution to mycology.

Type species: *Hydeopsis verrucispora.*

Saprobic on dead culms of herbaceous plant. Sexual morph: *Ascomata* immersed, scattered, solitary, subglobose, dark brown to black, coriaceous, ostiolate. *Hamathecium* comprising numerous, hypha-like, cellular pseudoparaphyses, anastomosing between and among the asci. *Asci* clavate, 8-spored, bitunicate, fissitunicate, pedicellate, with a furcate pedicel at the base, rounded to truncate at the apex, with a developed ocular chamber. *Ascospores* ellipsoidal to fusiform, hyaline to pale pigmented, aseptate when young, and becoming septate at maturation, constricted at the median septum, surrounded by a mucilaginous sheath. Asexual morph: Undetermined.

Notes: Phylogeny indicates that *Hydeopsis* can be recognized as member of Phaeosphaeriaceae (Figure 2.44), where it is phylogenetically close to *Dactylidina* and *Phaeopoacea.* However, it can be differentiated from these genera as it is characterized by ellipsoidal to fusiform, hyaline to pale pigmented, phragmosporous ascospores, while the members of *Dactylidina* and *Phaeopoacea* are described as having muriform, dark pigmented ascospores (Liu et al., 2015; Hyde et al., 2017; Wanasinghe et al., 2018b). Although *Hydeopsis* is morphologically similar to the genera *Acericola*, *Bricckea*, *Juncaceicola*, *Loratospora*, *Phaeosphaeria*, *Pseudophaeosphaeria*, *Setomelanomma* and *Setophoma* in having 3-septate, fusiform ascospores (Phookamsak et al., 2014; Ariyawansa et al., 2015; Tennakoon et al., 2016a; Hyde et al., 2016, 2017), our multigene phylogeny presented herein indicates that they are phylogenetically distinct genera.

Hydeopsis verrucispora J.F. Zhang, J.K. Liu & Z.Y. Liu, *sp. nov*.

Index Fungorum number: IF 556248; Facesoffungi Number: FoF 05838; Figure 2.37

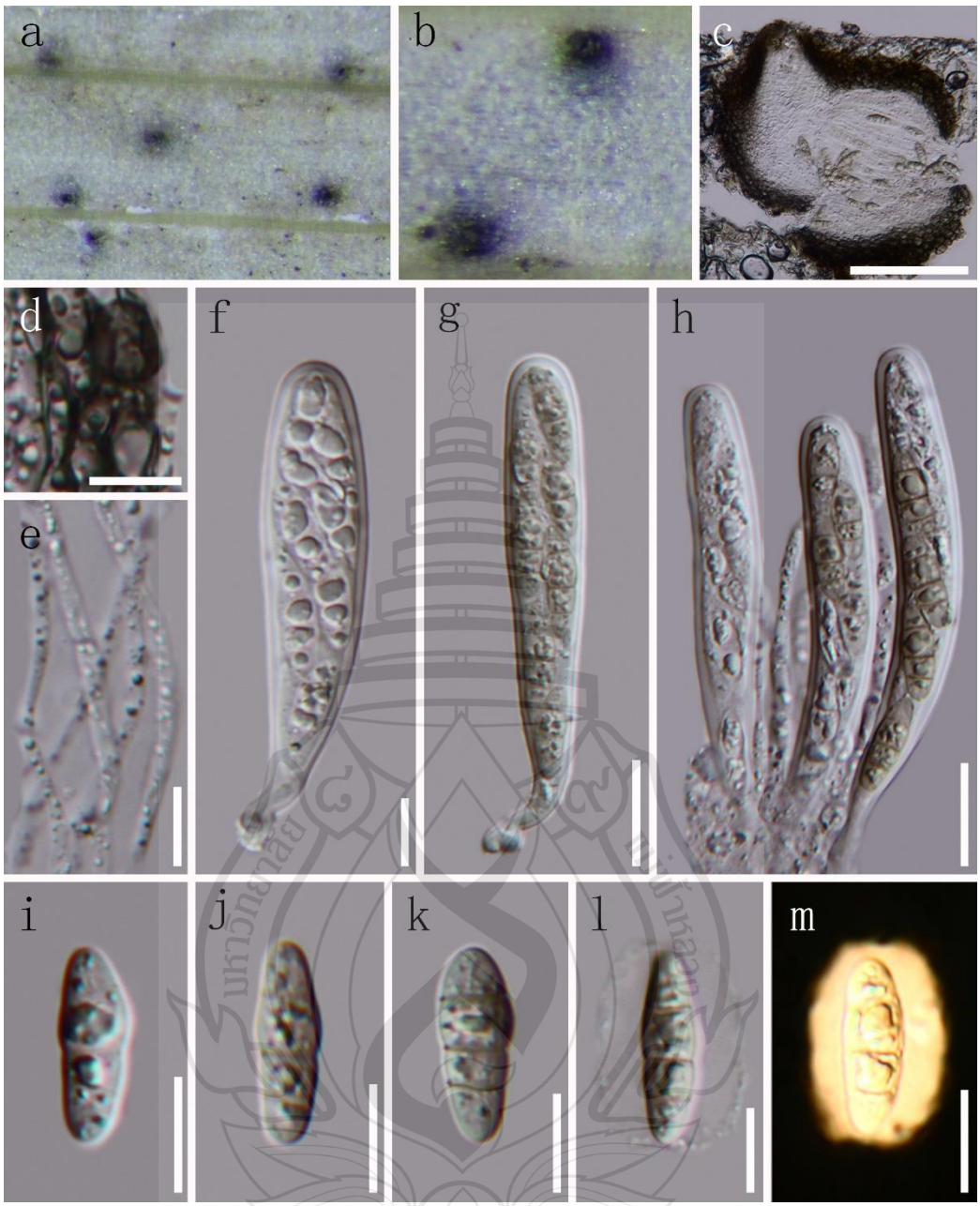
Etymology: In reference to the fungus having verrucose ascospores.

Holotype: MFLU 18-2269.

Saprobic on dead culms of unidentified herbaceous plant. Sexual morph: *Ascomata* 191–235 µm high, 186–215 µm wide, immersed, scattered, solitary, subglobose, dark brown to black, coriaceous, ostiolate. *Ostiole* central, with a flat, pore-like opening. *Peridium* up to 19–37 µm thick, comprising several layers of dark brown to black, thick-walled cells of textura angularis, outwardly dark pigmented, and become pale brown to hyaline towards inwardly. *Hamathecium* comprising numerous, 2–3.5 µm wide, hypha-like, cellular pseudoparaphyses, anastomosing between and among asci. *Asci* (69–) 84.5–93.5 × 11– 16.5 µm ($\bar{x}$ = 85 × 14.5 µm, n = 20), 8-spored, bitunicate, fissitunicate, clavate, pedicellate, with a furcate stalk at the base, rounded and truncate at the apex, with a developed ocular chamber. *Ascospores* 18.5–20.5(–23.5) × 6–7.5 µm ($\bar{x}$ = 20 × 6.5 µm, n = 25), ellipsoidal to fusiform, hyaline to pale brown, verrucose, aseptate when young, and become 3- septate upon maturation, constricted at the median septum, surrounded by a 4.5–7 µm wide, mucilaginous sheath. Asexual morph: Undetermined.

Material examined: CHINA, Guizhou Province, Guiyang City, Huaxi District, dead culms of herbaceous plant, 03 April 2016, J.F. Zhang, SD-2016-5 (MFLU 18-2269; GZAAS 18-0007); ex- type living culture (MFLUCC 19-0163, GZCC 19-0001).

Notes: The type species of *Dactylidina* (*D. dactylidis*) and *Phaeopoacea* (*P. festucae*) share close phylogenetic affinities with *Hydeopsis verrucispora* in both ML and BI analyses (Figure 2.44). *Hydeopsis verrucispora* also shares some similar morphological characteristics to those species in *Dactylidina* and *Phaeopoacea* by having globose or subglobose, glabrous ascomata, cylindrical to clavate asci. However, it can be easily distinguished from members of these two genera as the new taxon is observed to have 3-septate, fusiform ascospores, while the members of *Dactylidina* are characterized by muriform ascospores with 5–6-transverse septate and 1-longitudinal septum (Wanasinghe et al., 2018), and *Phaeopoacea* species are described with narrowly fusiform ascospores with 5-transverse septate and 1-longitudinal septum in the mid cells (Thambugala et al., 2017).



Note a Appearance of ascostromata on host surface. b Ascomata with a central, pore-like opening ostiole. c Vertical sections through ascomata. d Section of peridium. e Cellular pseudoparaphyses. f–h Asci. i–m Ascospores. Note: the m is stained with India ink to present the mucilaginous sheath. Scale bars: c = 100 µm, d–f, i–m = 10 µm, g, h = 20 µm.

Figure 2.37 *Hydeopsis verrucispora* (MFLU 18-2269, holotype)

Leptospora Rabenh., Hedwigia 1: 116 (1857)

Leptospora is typified by *L. rubella* and characterized by large, flask shaped ascomata with long cylindrical asci and thin, filiform, multi-septate ascospores, and members of this genus had also been reported to produce a reddish-purple pigmentation to stain the host tissue around the ostiole (Walker, 1980; Hyde et al., 2016).

Leptospora hydei J.F. Zhang, J.K. Liu & Z.Y. Liu, *sp. nov*.

Index Fungorum number: IF 556264; Facesoffungi Number: FoF 05840; Figure 2.38

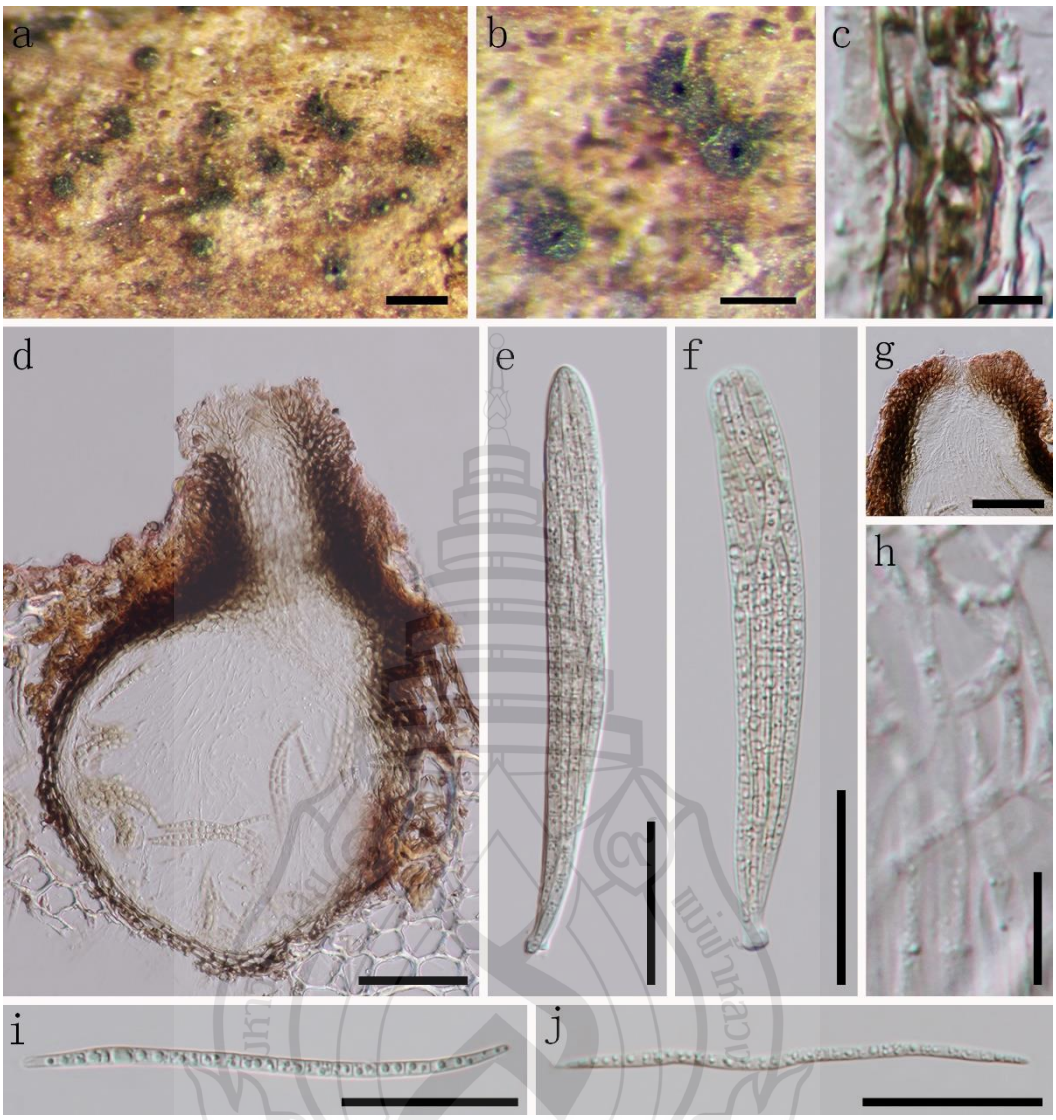
Etymology: In honor of Prof. Kevin D. Hyde for his contribution to ascomycete taxonomy.

Holotype: MFLU 19-0564.

Saprobic on decaying culms of unidentified herbaceous plant. Sexual morph: *Ascomata* 196– 267 µm high, 168–187 µm diam, immersed to erumpent through host tissue with papilla, solitary, scattered, depressed globose to subglobose, brown to dark brown, ostiolate. *Ostiole* central, papillate, cylindrical, truncate at the apex, dark brown to light brown, ostiloar canal filled with periphyses, and produces a reddish-purple discoloration in host tissue around ostiole. *Peridium* up to 11–24 µm wide, coriaceous, composed of several layers of thick-walled, brown to dark brown, pseudoparenchymatous cells, arrange in a *textura angularis* to *prismatica*, thin at the base, becoming thick and dark-pigmented towards ostiole. *Hamathecium* comprising numerous, 1.5–3 µm wide, filamentous, branched, trabeculate pseudoparaphyses anastomosing above and between the asci. *Asci* 147–165(–180) × 13–17(–19.5) µm ($\bar{x}$ = 161 × 15.2 µm, n = 25), 8-spored, bitunicate, cylindrical to cylindrical-clavate, with very short swollen pedicel, apically rounded to truncate with a minute ocular chamber. *Ascospores* (104–)124–138(–146) × 4–4.8 µm ($\bar{x}$ = 133.5 × 4.4 µm, n = 30), fasciculate, tri- to multiseriate, scolecosporous, filiform, tapering towards the ends, minute guttules in each cell, hyaline when young, and become pale brown at maturation, normally 11-septate, not constricted at the septa. Asexual morph: Undetermined.

Material examined: CHINA, Guizhou Province, Guiyang City, Huaxi District, on decaying branch, 20 September 2016, J.F. Zhang, SD-02 (MFLU 19-0564, GZAAS 18-0010); ex-type living culture (GZCC 19-0004).

Notes: *Leptospora hydei* shares key morphological characters with existing *Leptospora* species in staining the host tissue with reddish-purple pigmentation around ostiole canal, and filiform, multi-septate ascospores. The phylogenetic result also assigns *L. hydei* to the genus *Leptospora*, as it clusters together with *L. thailandica* with strong statistical support (Figure 2.44). However, it differs from *L. thailandica* in having larger asci (147–80 × 13–19.5 µm vs. 68–114 × 7–13 µm) and ascospores (104–146 × 4–4.8 µm vs. 63–89 × 1.8–3.8 µm), with less number of septum in ascospores (11-septate vs. 14–22-septate) (Hyde et al., 2016).



Note a Appearance of ascostromata on host surface. b The ostiole on face view. c Peridium. d Vertical section through ascomata. e, f Asci. g Ostiolar canal filled with numerous periphyses. h Pseudoparaphyses. i, j Ascospores. Scale bars: a = 200 µm, b = 100 µm, c = 5 µm, d-g, i–j = 50 µm, h = 10 µm.

Figure 2.38 *Leptospora hydei* (MFLU 19-0564, holotype)

Neoophiobolus Mapook & K.D. Hyde, Fungal Diversity 101: 75 (2020)

Neoophiobolus was introduced by Mapook et al. (2020a) to accommodate the type species *N. chromolaenae* based mainly on phylogenetic analysis and its ophiobolus-like morphological characteristics. Combination the morphological and phylogenetic evidence, the newly collected specimens are identified as separate species, and introduced herein as two novel species *N. karstii* and *N. filiformesporum*. Moreover, the discovery of these taxa also stabilized the placement of *Neoophiobolus* within in Phaeosphaeriaceae. Nevertheless, a morphological feature of the ascospores separate into two parts near at the constricted central septum that described by Mapook et al. (2020a) was not shown in their photoplate, and also not observed in our new collections. Therefore, the holotype of *N. chromolaenae* should be re-examined to emend the conception of *Neoophiobolus*.

Neoophiobolus filiformesporum J.F. Zhang & J.K. Liu *sp. nov.*, Figure 2.39

Etymology: "*filiformesporum"* named refers to the ascospore of the holotype is filiform.

Holotype: GZAAS 19-1748.

Saprobic on dead culm of unidentified herbaceous plant. Sexual morph: *Ascomata* 334–438 µm high, 357–493 µm diam., dark brown to black, scattered or gregarious, semi-immersed to erumpent through the host tissue with papilla, visible as raised, small, black dots on the host surface, uni-loculate, subglobose to ampulliform, ostiolate, papillate. *Ostiole* central, rounded to truncate at the apex, with periphyses inside. *Peridium* 15–30 µm wide, becomeing thickness towards ostile, composed of several layers of dark brown to black, thick pseudoparenchymatous cells in outermost, pale brown to hyaline towards inwardlly, arranged in *textura angularis*. *Hamathecium* comprised of numerous, 1–2 µm wide, filamentous, branched, septate pseudoparaphyses, anastomosing between and above the asci, embedded in a mucilaginous matrix. *Asci* (95–)105–111.5(–120) × 7.5–10 µm ($\bar{x}$ = 109 × 9 µm, n = 20), 8-spored, bitunicate, cylindrical to clavate, short pedicellate, apically rounded with an indistinct ocular chamber. *Ascospores* (92–)97.5–108.5(–113) × 2.5–3 µm ($\bar{x}$ = 100.5 × 2.6 µm, n = 25), fasciculate in parallel, scolecosporous, filiform, curved, tapered towards the lower cells, swollen near the 1/4 to 1/3 from the upper, guttulate, lacking bipolar appendages. Asexual morph: Undetermined.

Material examined: CHINA, Guizhou Province, Leishan County, on dead culm of unidentified herbaceous plant, 24 May 2019, J.F. Zhang LS-2019-26 (GZAAS 19-1748, holotype); Ibid: LS-2019-4 (GZAAS 19-1747).

Notes: *Neoophiobolus filiformesporum* sisters to *N. karstii*, and where they cluster with *N. chromolaenae* with high bootstrap support (Figure 2.6). Morphologically, *N. filiformesporum* is typical of *Neoophiobolus*, and also producing the overlap sized of asci and ascospores with the type species *N. chromolaenae* (Mapook et al., 2020). However, the results of comparison each gene region between our new collection and the ex-type of *N. chromolaenae* (3/818, 21/504, 25/869 base pair differences in LSU, ITS and TEF1-a, respectively) can fully prove that they are separate species.



Note a Appearance of ascomata in the host surface. b Vertical section through ascoma. c Section of Peridium. d Immature ascus. e Pseudoparaphyses anastomsed between asci. f, g Asci. h, i Ascospores. Scale bars: b = 100 μm, c = 5 μm, d–g = 20 μm, h, i = 30 μm.

Figure 2.39 *Neoophiobolus filiformesporum* (GZAAS 19-1748, holotype)

Ophiosphaerella Speg., Anal. Mus. nac. B. Aires, Ser. 3 12: 401 (1909)

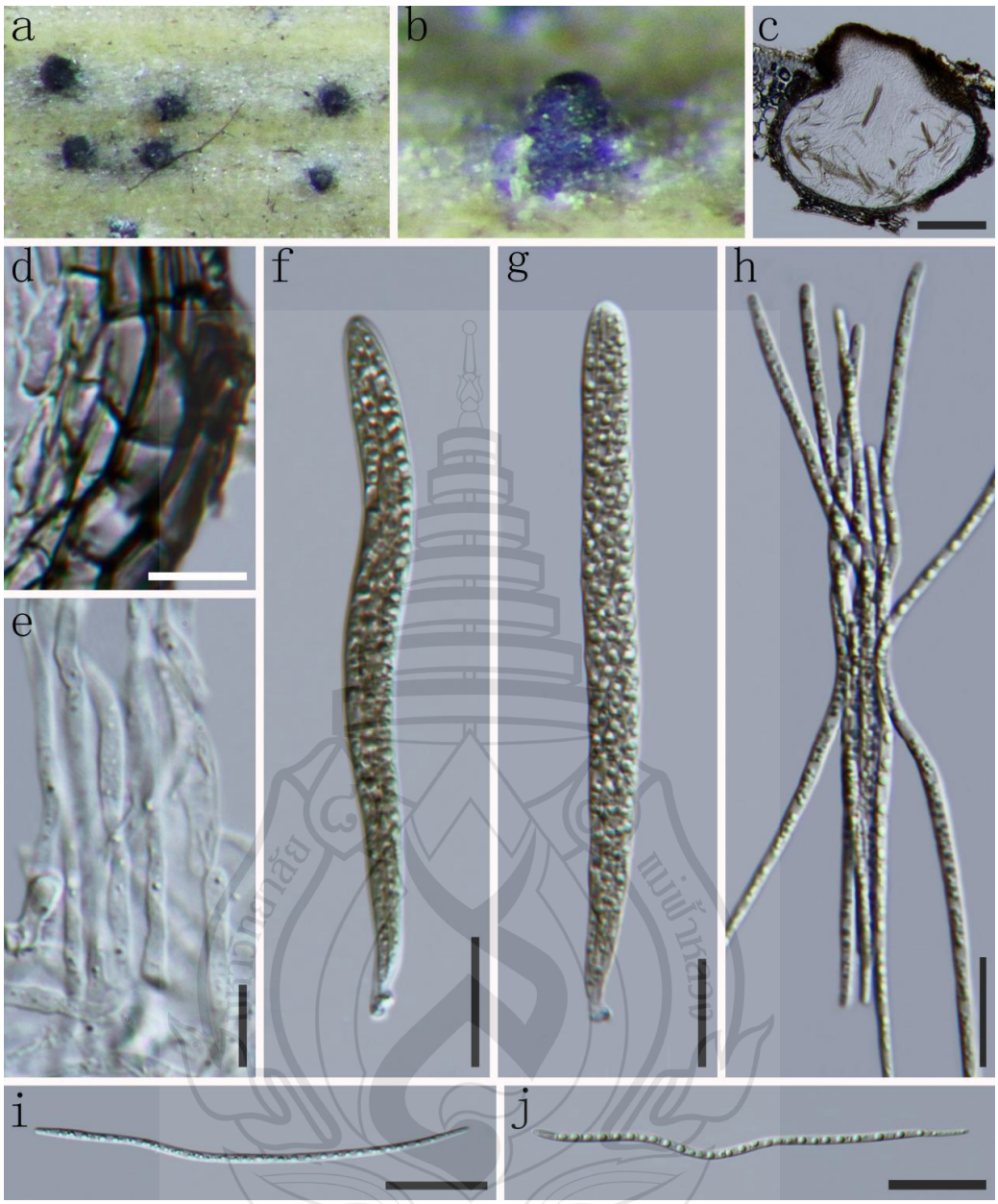
Ophiosphaerella, first appeared in 1909 with the type species *O. graminicola*. Petrak and Sydow (1936) synonymized *O. graminicola* with *Ophiobolus graminicolus*, and transferred *Ophiosphaerella* under *Ophiobolus*. However, Walker (1980) denied this result, and confirmed that *Ophiosphaerella* is a seperate genus that distinguished from *Ophiobolus* by its ascospores without swollen cells or separating into partspores. Then, a series of phylogenetic analysi were carried out to investigate the placement of *Ophiosphaerella* (Wetzel et al., 1999; Schoch et al., 2006, 2009a; Hyde et al., 2013). Phookamsak et al. (2014) revised the family Phaeosphaeriaceae and indicated that *Ophiosphaerella* seem to be polyphyletic, however, the available sequence data of this genus are still limited in GenBank. especially. Currently, 12 species were accepted in *Ophiosphaerella*.

Ophiosphaerella agrostidis Dern. et al., in Camara et al., Mycologia 92(2): 320 (2000), *new record.* Figure 2.40

Saprobic on dead culm of unidentified herbaceous plant. Sexual morph: *Ascomata* subglobose to pyriform, scattered, solitary, immersed to semi-immersed or erumpent throughout the host tissue with papilla,visible as raised, small black dots on host surface, uni-loculate, dark brown to black. *Ostiole* central, with periphyses inside the ostiolar canal. *Peridium* 18–44 μm wide, of unequal thickness, outer layer comprising dark brown, occluded thick-walled cells, fusing with the host, inner layer comprising several layers of thin-walled, flattened, pale brown to hyaline cells of *textura angularis*. *Hamathecium* comprised of numerous, 2–3.5 μm wide, filamentous, septate pseudoparaphyses, embedded in a mucilaginous matrix, anastomosing at the apex. *Asci* (109–)120–138(–145.5) × 7–9 μm ($\bar{x}$ = 127 × 8 μm, n = 25), 8-spored, bitunicate, cylindrical to cylindric-clavate, short pedicellate, apically rounded, with a minute ocular chamber. *Ascospores* (119–)130–139(–144) × 2–3 μm ($\bar{x}$ = 133 × 2.5 μm, n = 20), fasciculate, scolecosporous, spiral, filiform, hyaline and aseptate when young, becoming pale brown to brown, 12–15-septate at maturity, not constricted at the septa, smooth-walled, guttulate. Asexual morph: Undertermined.

Material examined: CHINA, Guizhou Province, Libo County, Maolan Town, on dead culm of unidentified herbaceous plant, 18 July 2016, H.Y. Ran MLBB-7 (MFLU 18-2272).

Notes: The new collection clusters with *O. agrostidis* by strong statistical support in the present study (Figure 2.6). Morphologically, it is also similar to *O. agrostidis* by having cylingrical, short pedicellate asci, and multi-septate filiform ascospores, and including the measurements of these above features correspond to the descriptions from Câmara et al. (2000) and Phookamsak et al. (2014). Besides, each gene region between our new specimens and *O. agrostidis* (MFLUCC 11-0152) was compared, and the results showed that there are no differences in LSU, SSU and ITS respectively, except 3/878 bp difference in TEF1-a. In view of these above evidences, this new specimen is identified as *O. agrostidis*, and reported as a new geographical record for China as it has only been known from Thailand and USA (Câmara et al., 2000; Phookamsak et al., 2014).



Note a, b Appearance of ascomata in the host surface. c Vertical section through ascoma. d Section of peridium. e Pseudoparaphyses. f, g Asci. h The arrangement of ascospores. i, j Ascospores. Scale bars: c = 200 μm, d, f, g = 20 μm, e = 5 μm, h, i, j = 30 μm.

Figure 2.40 *Ophiosphaerella agrostidis* (MFLU 18-2272, new record)

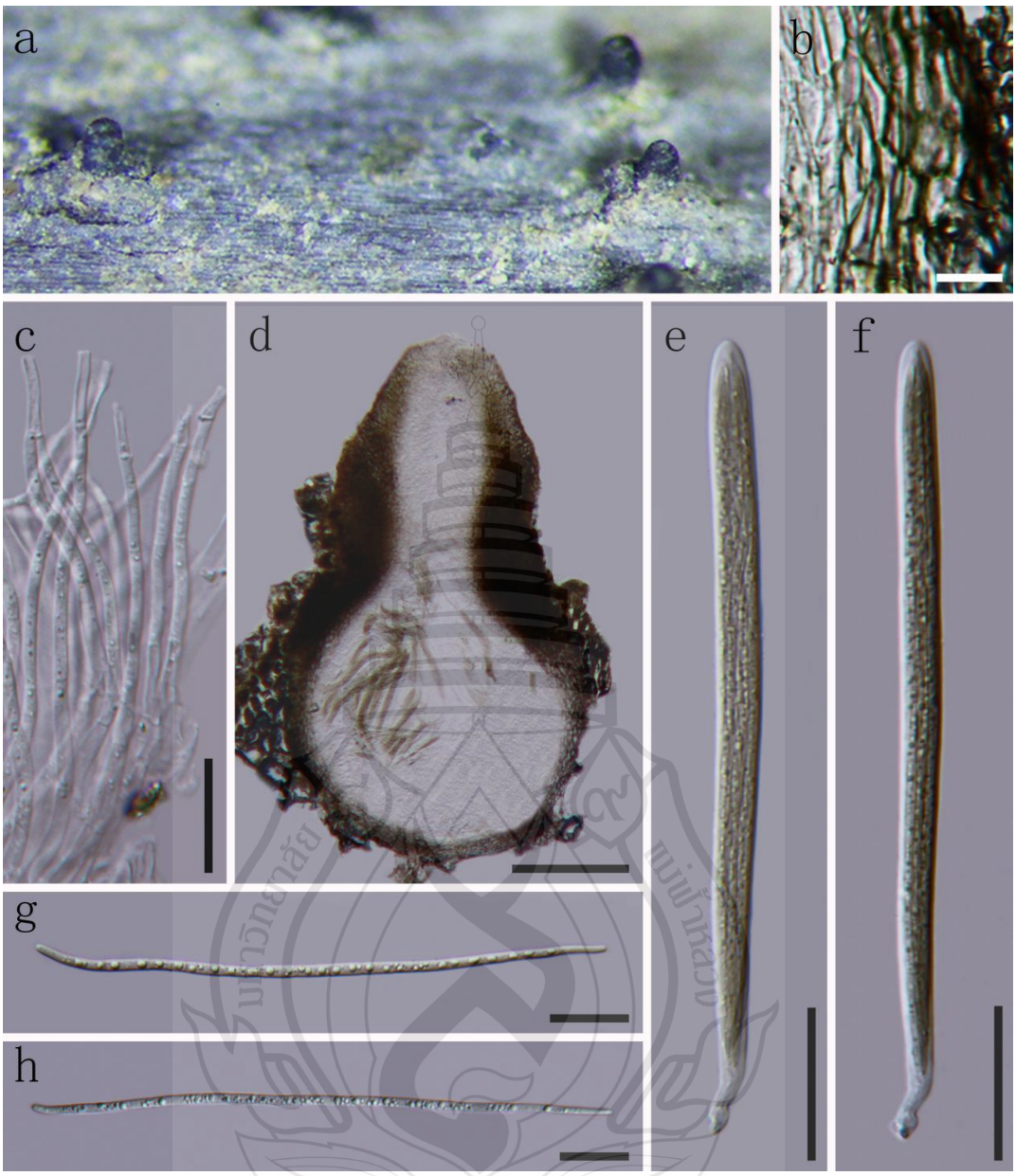
Ophiosphaerella chiangraiensis Phookamsak & K.D. Hyde, Fungal Diversity: 104, [29] (2020), *new record* Figure 2.41

Saprobic on dead culm of unidentified herbaceous plant. Sexual morph: *Ascomata* 352–453 μm high, 189–323 μm diam., subglobose to pyriform, scattered, solitary, immersed to semi-immersed or erumpent throughout the host tissue, with minute papilla, visible as raised, small black dots on host surface, uni-loculate, dark brown to black. *Ostiole* central, with periphyses lined in the ostiolar canal. *Peridium* of unequal thickness, 17–25 μm at the base, 25–46 μm in sides, and 52–78 μm around the ostiole, composed of several layers of *pseudoparenchymatous* cells, outer layer comprising dark brown, occluded thick-walled cells, fusing with the host, inner layer comprising several layers of thin-walled, flattened, pale brown cells of *textura angularis*. *Hamathecium* composed of numerous, 2–4.5 μm wide, filamentous, septate pseudoparaphyses, swollen near the base, embedded in a mucilaginous matrix, anastomosing between the asci. *Asci* (136–)143–155(–170) × 7–8.5 μm ($\bar{x}$ = 151 × 7.5 μm, n = 25), 8-spored, bitunicate, cylindrical to cylindric-clavate, short pedicellate, apically rounded, with a distinct ocular chamber. *Ascospores* (109–)129–151(–164) × 2–2.7 μm ($\bar{x}$ = 140 × 2.5 μm, n = 30), fasciculate, scolecosporous, parallel or spiral, filiform, pale brown to brown, multi-septate, not constricted at the septa, taper towards one end, smooth-walled, guttulate. Asexual morph: Undertermined.

Material examined: CHINA, Guizhou Province, Guiyang City, Huaxi District, Guizhou Academy of Agricultural Sciences, on dead culm of unidentified herbaceous plant, 5 Octomber 2016, J.F. Zhang GZ-10 (MFLU 18-2293); Guizhou Academy of Agricultural Science, on dead culm of unidentified herbaceous plant, 25 September 2016, J.F. Zhang GZ-05 (GZAAS 21-2005); Huaxi District, dead culm of unidentified herbaceous plant, 20 September 2016, J.F. Zhang SD-03 (GZAAS 21-2006); Libo County, Maolan Town, Maolan National Nature Reserve, on dead dead culm of unidentified herbaceous plant, 20 July 2016, J.F. Zhang MLCC-11 (MFLU 18-2284);

Notes: Four newly collected specimens are phylogenetically clustered together and closed to the known species *O. chiangraiensis* in the present study (Figure 2.6). Morphologically, these specimens resemble with *O. chiangraiensis* by having the overlap sized ascomata, asci and ascospores (Yuan et al., 2020). Moreover, each gene region between these collections and *O. chiangraiensis* was compared, and the results

showed that there are 100% similarity in LSU and SSU, and 2/494, 3/878 and 10/769 base pair differences in ITS, TEF1-a, and Rpb2, respecitively. Combine these above analysis and recommendations from Jeewon and Hyde (2016), these four new collections are identified as *O. chiangraiensis*, and reported it as a new geographical record since this species was discovered for the first time in China.



Note a Appearance of ascomata in the host surface. b Section of peridium. c Pseudoparaphyses. d Vertical section through ascoma. e, f Asci. g, h Ascospores. Scale bars: b = 10 μm, c, g, h = 20 μm, d = 100 μm, e, f = 30 μm.

Figure 2.41 *Ophiosphaerella chiangraiensis* (MFLU 18-2293, new record)

Ophiosphaerella karstii J.F. Zhang, H.Y. Ran & J.K. Liu, *sp. nov*., Figure 2.42

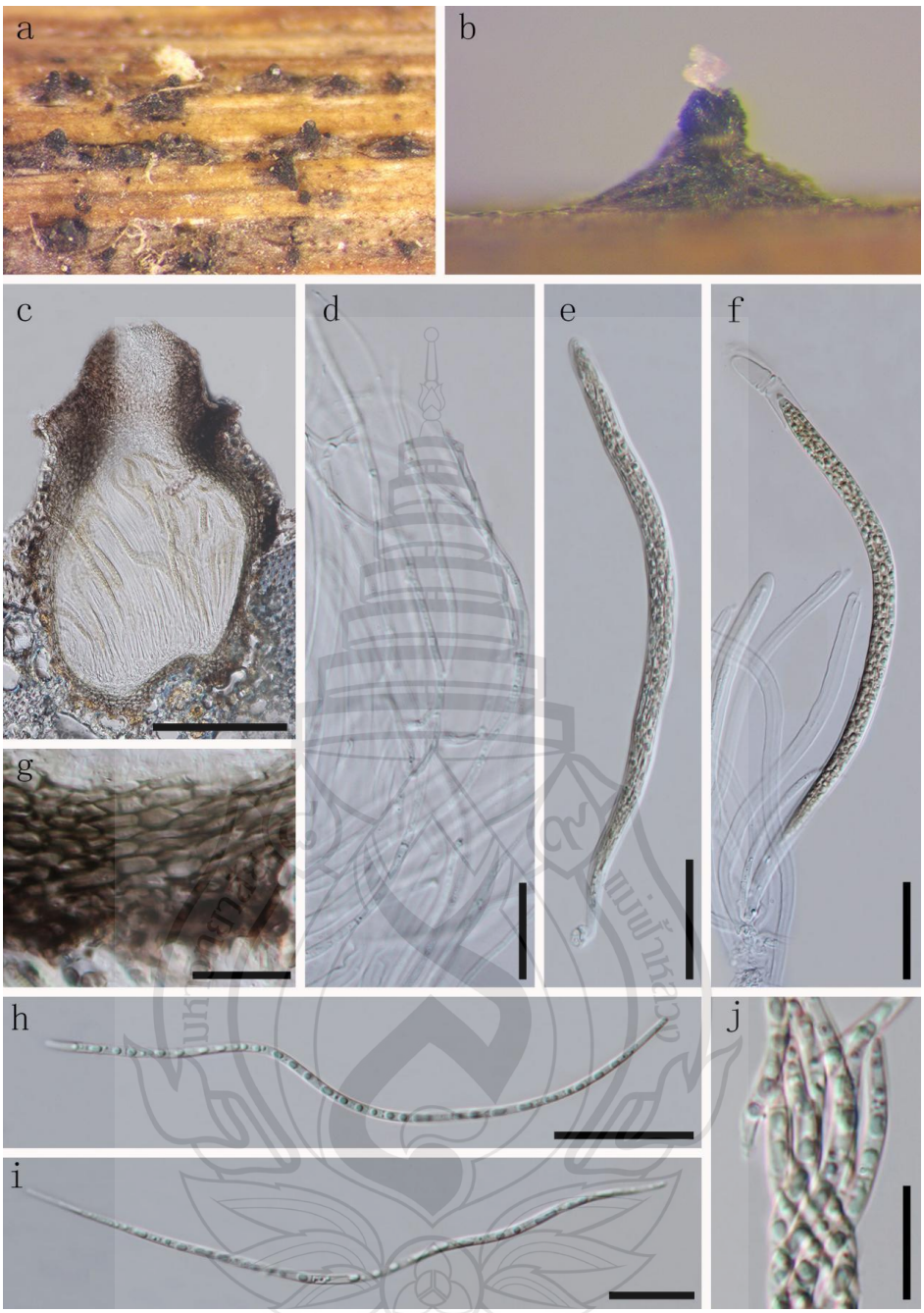
Etymology: "*karstii"* named refers to the holotype was discovered in karst formations.

Holotype: GZAAS 19-4021.

Saprobic on dead culm of unidentified herbaceous plant. Sexual morph: *Ascomata* subglobose to pyriform or irregular, scattered or gregarious, immersed to semi-immersed or erumpent throughout the host tissue with papilla, ostiolate. *Ostiole* central, with periphyses lined inside. *Peridium* 19–48 μm, composed of several layers of pseudoparenchymatous cells of textura angularis, brown to dark brown in the outermost, and fusing with the host, pale brown to hyaline towards inwardlly. *Hamathecium* compring of numerous, 1.5–2.5 μm wide, filamentous, septate, branched pseudoparaphyses, anastomosing between the asci. *Asci* (142.5–)165–176(–188) × 6.5–8 μm ($\bar{x}$ = 169 × 7.5 μm, n = 25), 8-spored, bitunicate, long cylindrical, short pedicellate, apically rounded, with a distinct ocular chamber. *Ascospores* (132–)156–160 × 2–2.5 μm ($\bar{x}$ = 151 × 2.4 μm, n = 30), fasciculate, scolecosporous, spiral, filiform, pale brown to brown, multi-septate, not constricted at the septa, taper towards one end, smooth-walled, guttulate. Asexual morph: Undertermined.

Material examined: CHINA, Guizhou Province, Libo County, Maolan Town, on dead culm of unidentified herbaceous plant, 8 July 2015, H.Y. Ran MLC-21 (GZAAS 19-4021, holotype).

Notes: *Ophiosphaerella karstii* is phylogenetically placed within in *Ophiosphaerella*, and close related to *O. taiwanensis* in current phylogenetic analysis (Figure 2.6). However, the new species is obviously different from *O. taiwanensis* by its longer asci (165–176 × 6.5–8 μm vs. 115–140 × 8.5–10 μm) and ascospores (156–160 × 2–2.5 μm vs. 110–132 × 2.2–2.7 μm) (Tennakoon et al., 2020).



Note a Appearance of ascomata in the host surface. b Section of peridium. c Pseudoparaphyses. d Vertical section through ascoma. e, f Asci. g, h Ascospores. Scale bars: c = 100 μm, d, g = 20 μm, e, f, h, i = 30 μm, h, i = 50 μm, j = 10 μm.

Figure 2.42 *Ophiosphaerella karstii* (GZAAS 19-4021, holotype)

Pseudoophiosphaerella J.F. Zhang, J.K. Liu & Z.Y. Liu, *gen. nov.*

Index Fungorum number: IF 556244; Facesoffungi Number:FoF 05835

Etymology: Refers to the morphological resemblance to *Ophiosphaerella.*

Type species: *Pseudoophiosphaerella huishuiensis.*

Saprobic on dead culms of herbaceous plant. Sexual morph: *Ascomata* scattered, solitary to gregarious, globose to subglobose, dark brown to black, immersed to erumpent, ostiolate. *Hamathecium* comprising numerous, hyphae-like, trabeculate, branched pseudoparaphyses. *Asci* cylindrical, 8-spored, bitunicate, fissitunicate, short pedicellate, apically rounded. *Ascospores* filiform, fasciculate, hyaline and aseptate when young, become pale-yellowish and multi-septate at maturation, lying parallel or spirally coiled, obtuse in one end and towards acute in another end, smooth-walled, guttulate. Asexual morph: Undetermined.

Notes: Our phylogeny indicates that *Pseudoophiosphaerella* shares a close relationship to *Dlhawksworthia* with good support (Figure 2.44). However, it is morphologically different from the latter in having filiform ascospores, while the ascospores in *Dlhawksworthia* were observed as ellipsoidal or muriform. Although *Pseudoophiosphaerella* is morphologically similar to *Ophiosphaerella* (in having long cylindrical asci with septate, filiform ascospores), our multi-gene analyses indicate that they are phylogenetically distant. Phookamsak et al. (2014) indicated that many sexual genera within Phaeosphaeriaceae share similar morphological characters, and it is difficult to distinguish these genera based only on morphological examination. *Pseudoophiosphaerella* is introduced herein as a new genus to accommodate the new species *Pseudoophiosphaerella huishuiensis*.

Pseudoophiosphaerella huishuiensis J.F. Zhang, J.K. Liu & Z.Y. Liu sp. nov.

Index Fungorum number: IF 556246; Facesoffungi number: FoF05836; Figure 2.43

Etymology: Refers to the place Huishui County, where the fungus was collected.

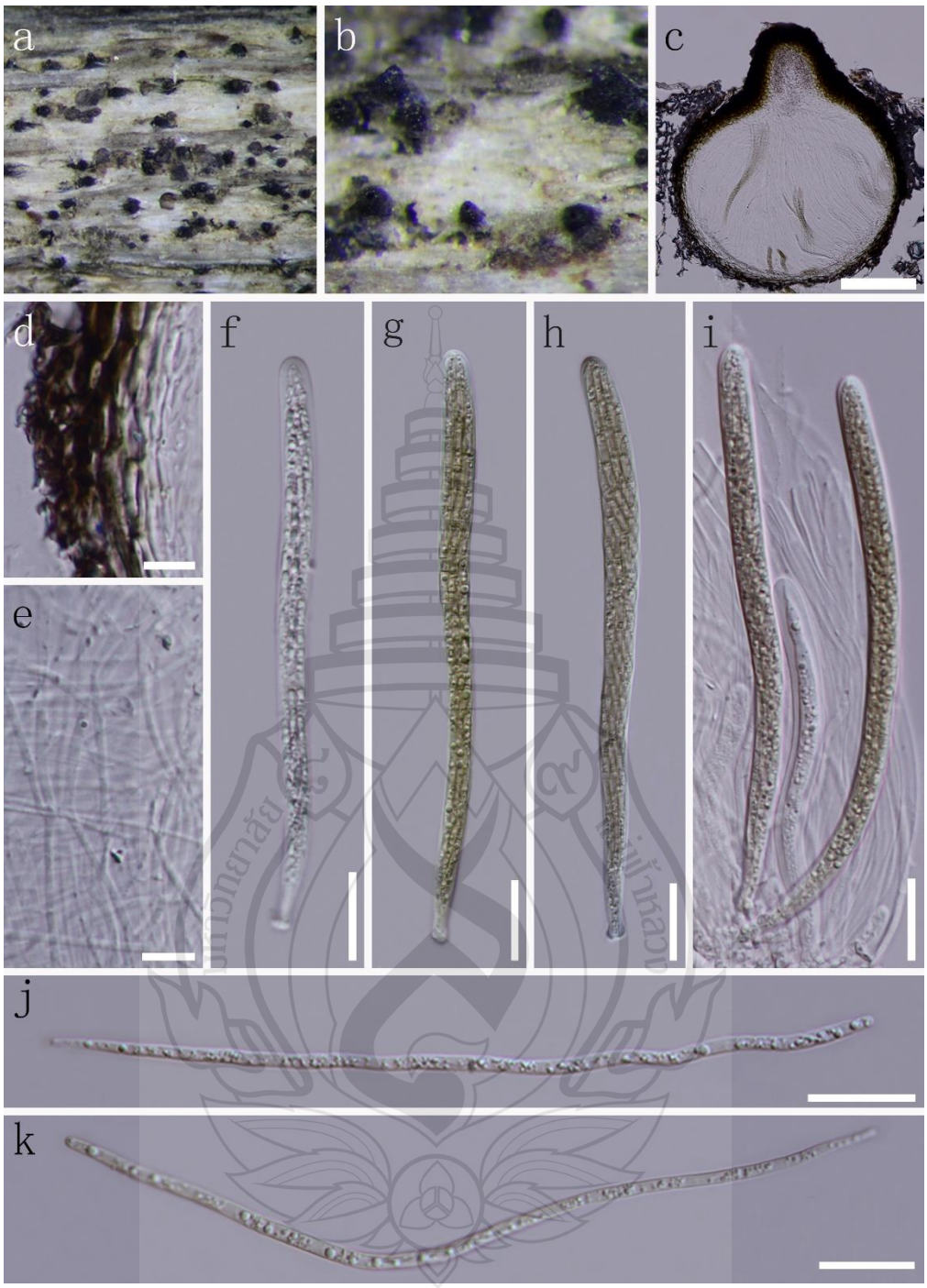
Holotype: MFLU 18-2266.

Saprobic on dead culms of unidentified herbaceous plant. Sexual morph: *Ascomata* scattered, solitary to gregarious, globose to subglobose, dark brown to black, immersed to erumpent, ostiolate. *Ostiole* central, papillate, conical, with a pore-like opening. *Peridium* 25–34 μm thick, coriaceous, composed of several layers of

thick-walled, brown to dark brown, pseudoparenchymatous cells of *textura angularis*, become pale brown to hyaline towards inner layers. *Hamathecium* comprising numerous, 1.5–3 μm wide, hyphae-like, pseudoparaphyses, trabeculate at the base and branched at the apex, anastomosing between and above asci, embedded in a gelatinous matrix. *Asci* 143–151.5 × 8–9.5 μm ($\bar{x}$ = 148 × 8.7 μm, n=25), 8-spored, bitunicate, fissitunicate, long cylindrical, short pedicellate, apically rounded, with a minute ocular chamber. *Ascospores* 130.5–151.5 × 2.5–3.5 μm ($\bar{x}$ = 142.5 × 3 μm, n=30), filiform, fasciculate, hyaline and aseptate when young, become pale-yellowish and multi-septate at maturation, multiseriate, lying parallel or spirally coiled, obtuse in one end and acute in another end, smooth-walled, guttulate. Asexual morph: Undetermined.

Material examined: CHINA, Guizhou Province, Huishui County, on dead twigs of unidentified host, 21 July 2017, J.F. Zhang, HS- 13 (MFLU 18-2266; GZAAS 18-0008); ex-type living culture (MFLUCC 19-0164, GZCC 19-0002).

Notes: The new species *Pseudoophiosphaerella huishuiensis* is phylogenetically related to *Dlhawksworthia* species with good support (Figure 2.44). However, they can be distinguished on the basis of their morphological characteristics as the new taxon is characterized by long cylindrical asci with filiform, fasciculate, multiseriate ascospores, while the members of *Dlhawksworthia* are characterized by cylindric-clavate asci with ellipsoidal, fusiform, overlapping uniseriate ascospores (Wanasinghe et al., 2018a).



Note a, b Appearance of ascostromata on host surface. c Vertical sections through ascomata. d Section of peridium. e Pseudoparaphyses. f–i Asci. j, k Ascospores. Scale bars: c = 100 µm, d, e = 10 µm, f–k = 20 µm.

Figure 2.43 *Pseudoophiosphaerella huishuiensis* (MFLU 18-2266, holotype)

Table 2.3 GenBank accession numbers of sequences used in phylogenetic analysis. New sequences from this study are in bold

Species	Specimen/Strain No.	LSU	ITS	SSU	TEF1-α
Acericola italica	MFLUCC 13-0609	MF167429	MF167428	MF167430	---
Allophaeosphaeria muriformia	MFLUCC 13-0349	KP765681	KP765680	KP765682	---
Allophaeosphaeria subcylindrospora	MFLUCC 13-0380	KT314183	KT314184	KT314185	---
Amarenographium ammophilae	MFLUCC 16-0296	KU848197	KU848196	KU848198	MG520894
Amarenomyces dactylidis	MFLUCC 14-0207	KY775575	KY775577	---	---
Ampelomyces quisqualis	CBS 129.79	EU754128	HQ108038	EU754029	---
Bhatiellae rosae	MFLUCC 17-0664	MG828989	MG828873	MG829101	---
Camarosporioides phragmitis	MFLUCC 13-0365	KX572345	KX572340	KX572350	KX572354
Chaetosphaeronema achilleae	MFLUCC 16-0476	KX765266	KX765265	---	---
Chaetosphaeronema hispidulum	CBS 216.75	KF251652	KF251148	EU754045	---
Dactylidina dactylidis	MFLUCC 13-0618	KP744473	KP744432	KP753946	---
Dactylidina dactylidis	MFLUCC 14-0966	MG829002	MG828886	MG829113	MG829199
Dematiopleospora donetzica	MFLU 15-2199	MG829005	---	MG829116	---
Dematiopleospora mariae	MFLUCC 13-0612	KJ749653	KJ749654	KJ749652	KJ749655
Diederichomyces ficuzzae	CBS 128019	JQ238616	KP170647	---	---
Diederichomyces xanthomendozae	CBS 129666	JQ238634	KP170651	---	---
Dlhawksworthia clematidicola	MFLUCC 14-0910	MG829011	MG828901	MG829120	MG829202

Table 2.3 (continued)

Species	Specimen/Strain No.	LSU	ITS	SSU	TEF1-α
Dlhawksworthia lonicerae	MFLUCC 14-0955	MG829012	MG828902	MG829121	MG829203
Embarria clematidis	MFLUCC 14-0652	KT306593	KT306949	KT306956	---
Embarria clematidis	MFLUCC 14-0976	MG828987	MG828871	MG829099	MG829194
Equiseticola fusispora	MFLUCC 14-0522	KU987669	KU987668	KU987670	MG520895
Galliicola pseudophaeosphaeria	MFLUCC 14-0524	KT326693	KT326692	---	MG520896
Hydeomyces desertipleosporoides	SQUCC 15259	MK290839	MK290841	MK290843	MK290848
Hydeomyces desertipleosporoides	SQUCC 15260	MK290840	MK290842	MK290844	MK290849
Hydeomyces pinicola	**GZCC 19-0003**	**MK522496**	**MK522506**	**MK522502**	**MK523386**
Hydeopsis verrucispora	**MFLUCC 19-0163**	**MK522498**	**MK522508**	**MK522504**	**MK523388**
Italica achilleae	MFLUCC 14-0959	MG829013	MG828903	MG829122	MG829204
Juncaceicola italica	MFLUCC 13-0750	KX500107	KX500110	KX500108	MG520897
Juncaceicola luzulae	MFLUCC 16-0780	KX449530	KX449529	KX449531	MG520898
Juncaceicola typharum	CBS 296.54	KF251695	KF251192	---	GU456287
Leptosphaeria doliolum	CBS 505.75	GU301827	JF740205	GU296159	GU349069
Leptospora galii	KUMCC 15-0521	KX599548	KX599547	KX599549	MG520899
Leptospora rubella	CPC 11006	DQ195792	DQ195780	DQ195803	---
Leptospora rubella	MFLU 16-0965	KX757834	KX757835	---	---
Leptospora thailandica	MFLUCC 16-0385	KX655549	KX655559	KX655554	KX655564

Table 2.3 (continued)

Species	Specimen/Strain No.	LSU	ITS	SSU	TEF1-α
Leptospora hydei	**GZCC 19-0004**	**MK522497**	**MK522507**	**MK522503**	**MK523387**
Loratospora luzulae	MFLUCC 14-0826	KT328495	KT328497	KT328496	---
Melnikia anthoxanthii	MFLUCC 14-1010	KU848204	---	KU848205	---
Muriphaeosphaeria galatellae	MFLUCC 14-0614	KT438329	KT438333	KT438331	MG520900
Muriphaeosphaeria galatellae	MFLUCC 15-0769	KT438330	---	KT438332	---
Neosetophoma garethjonesii	MFLUCC 14-0528	KY496738	KY496758	KY501126	KY514402
Neosetophoma rosigena	MFLUCC 17-0768	MG829037	MG828928	MG829143	---
Neostagonospora arrhenather	MFLUCC 15-0464	KX910091	KX926417	KX950402	MG520901
Neostagonospora caricis	CBS 135092	KF251667	KF251163	---	---
Neostagonospora phragmitis	MFLUCC 16-0493	KX910090	KX926416	KX950401	MG520902
Neostagonosporella sichuanensis	MFLU 18-1223	MH394690	MK296469	MH394687	MK313854
Neostagonosporella sichuanensis	MFLUCC 18-1228	MH368073	MH368088	MH368079	MK313851
Neosulcatispora agaves	CPC 26407	KT950867	KT950853	---	---
Neosulcatispora strelitziae	CPC 25657	KX228305	KX228253	---	---
Nodulosphaeria guttulatum	MFLUCC 15-0069	KY496726	KY496746	KY501115	KY514394
Nodulosphaeria multiseptata	MFLUCC 15-0078	KY496728	KY496748	KY501116	KY514396
Nodulosphaeria scabiosae	MFLUCC 14-1111	KU708846	KU708850	KU708842	KU708854

Table 2.3 (continued)

Species	Specimen/Strain No.	LSU	ITS	SSU	TEF1-α
Ophiobolopsis italica	MFLUCC 17-1791	MG520959	MG520939	MG520977	MG520903
Ophiobolus artemisiae	MFLUCC 14-1156	KT315509	KT315508	MG520979	MG520905
Ophiobolus disseminans	MFLUCC 17-1787	MG520961	MG520941	MG520980	MG520906
Ophiobolus ponticus	MFLUCC 17-2273	MG520963	MG520943	MG520982	MG520908
Ophiosimulans tanaceti	MFLUCC 14-0525	KU738891	KU738890	KU738892	MG520910
Ophiosphaerella agrostidis	MFLUCC 11-0152	KM434281	KM434271	KM434290	KM434299
Ophiosphaerella aquaticus	MFLUCC 14-0033	KX767089	KX767088	KX767090	MG520911
Ophiosphaerella herpotricha	KY423	KP690989	KP690989	KP690989	KP691011
Ophiosphaerella korrae	ATCC 56289	KC848509	KC848509	KC848509	KC848515
Ophiosphaerella narmari	ATCC 64688	KC848510	KC848510	KC848510	KC848516
Paraleptosphaeria dryadis	CBS 643.86	GU301828	JF740213	KC584632	GU349009
Paraophiobolus arundinis	MFLUCC 17-1789	MG520965	MG520945	MG520984	MG520912
Paraophiobolus plantaginis	MFLUCC 17-0245	KY815010	KY797641	KY815012	MG520913
Paraphoma chrysanthemicola	CBS 522.66	KF251670	FJ426985	GQ387521	---
Paraphoma radicina	CBS 111.79	KF251676	KF251172	EU754092	---
Parastagonospora italica	MFLUCC 13-0377	KU058724	KU058714	MG520985	MG520915
Parastagonospora minima	MFLUCC 13-0376	KU058723	KU058713	MG520986	MG520916
Parastagonospora nodorum	CBS 110109	KF251681	KF251177	---	---
Parastagonosporella fallopiae	CBS 135981	MH460545	MH460543	---	---

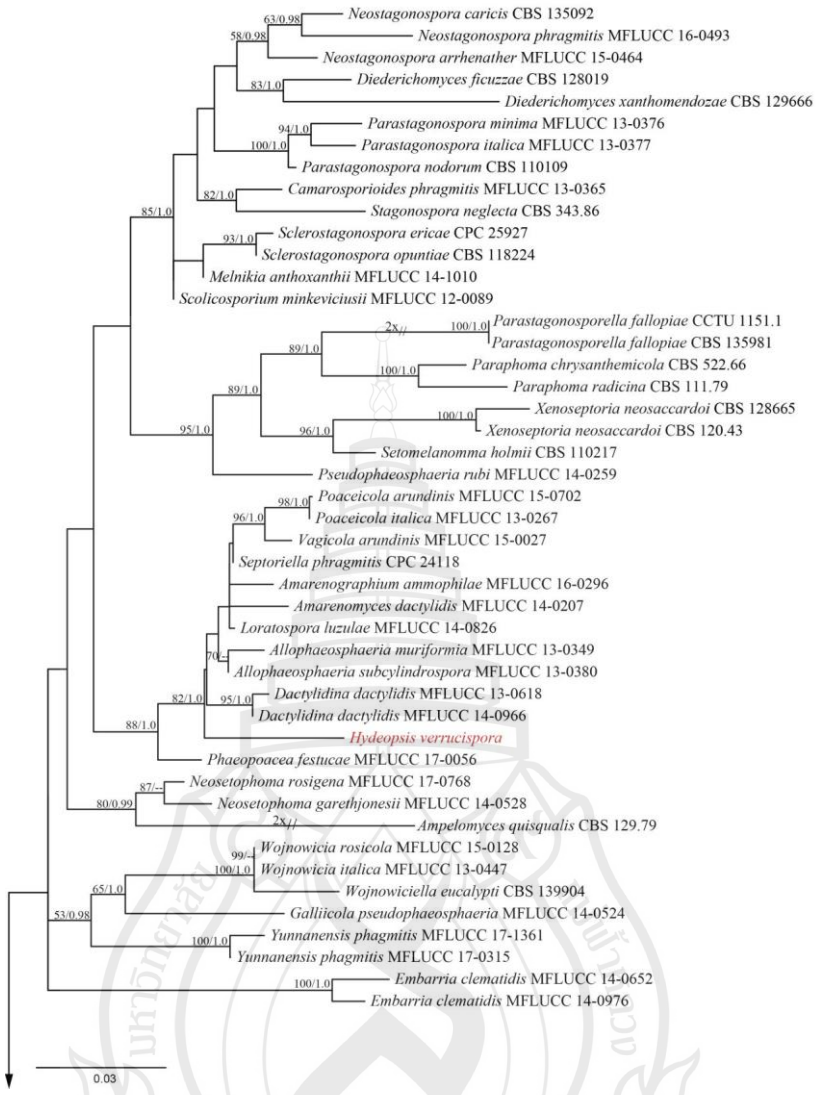
Table 2.3 (continued)

Species	Specimen/Strain No.	LSU	ITS	SSU	TEF1-α
Parastagonosporella fallopiae	CCTU 1151.1	MH460546	MH460544	---	---
Phaeopoacea festucae	MFLUCC 17-0056	KY824767	KY824766	KY824769	---
Phaeosphaeria chiangraina	MFLUCC 13-0231	KM434280	KM434270	KM434289	KM434298
Phaeosphaeria oryzae	CBS 110110	KF251689	KF251186	GQ387530	---
Phaeosphaeriopsis glaucopunctata	MFLUCC 13-0265	KJ522477	KJ522473	KJ522481	MG520918
Phaeosphaeriopsis triseptata	MFLUCC 13-0271	KJ522479	KJ522475	KJ522484	MG520919
Phaeosphaeriopsis yuccae	MFLUCC 16-0558	KY554481	KY554482	KY554480	MG520920
Poaceicola arundinis	MFLUCC 15-0702	KU058726	KU058716	MG520988	MG520921
Poaceicola italica	MFLUCC 13-0267	KX910094	KX926421	KX950409	MG520924
Populocrescentia ammophilae	MFLUCC 17-0665	MG829059	MG828949	MG829164	MG829231
Populocrescentia forlicesenensis	MFLUCC 14-0651	KT306952	KT306948	KT306955	MG520925
Populocrescentia rosae	MFLU 17-0128	MG829060	---	MG829165	MG829232
Pseudoophiobolus mathieui	MFLUCC 17-1784	MG520969	MG520949	MG520991	MG520928
Pseudoophiobolus rosae	MFLUCC 17-1786	MG520972	MG520952	MG520993	MG520930
Pseudoophiobolus urticicola	KUMCC 17-0168	MG520975	MG520955	MG520996	MG520933
Pseudoophiosphaerella huishuiensis	**MLUCC 19-0164**	**MK522499**	**MK522509**	**MK522505**	**MK523389**
Pseudophaeosphaeria rubi	MFLUCC 14-0259	KX765299	KX765298	KX765300	MG520934

Table 2.3 (continued)

Species	Specimen/Strain No.	LSU	ITS	SSU	TEF1-α
Sclerostagonospora ericae	CPC 25927	KX228319	KX228268	---	
Sclerostagonospora opuntiae	CBS 118224	DQ286772	DQ286768	---	---
Scolicosporium minkeviciusii	MFLUCC 12-0089	KF366382	---	KF366383	---
Septoriella phragmitis	CPC 24118	KR873279	KR873251	---	---
Setomelanomma holmii	CBS 110217	GQ387633	KT389542	GQ387572	GU349028
Setophoma terrestris	CBS 335.29	KF251749	KF251246	GQ387526	---
Stagonospora neglecta	CBS 343.86	EU754218	AJ496630	EU754119	---
Tintelnotia destructans	CBS 127737	KY090664	KY090652	KY090698	---
Wojnowicia italica	MFLUCC 13-0447	KX430001	KX342923	KX430002	KX430003
Wojnowicia rosicola	MFLUCC 15-0128	MG829091	MG828979	MG829191	---
Wojnowiciella eucalypti	CBS 139904	KR476774	KR476741	---	---
Xenoseptoria neosaccardoi	CBS 128665	KF251784	KF251281	---	---
Xenoseptoria neosaccardoi	CBS 120.43	KF251783	KF251280	---	---
Yunnanensis phagmitis	MFLUCC 17-0315	MF684863	MF684862	MF684867	MF683624
Yunnanensis phagmitis	MFLUCC 17-1361	MF684865	MF684869	MF684864	MF683625

Note CBS: CBS-KNAW Fungal Biodiversity Centre, Utrecht, The Netherlands; **CPC** Collection of Pedro Crous housed at CBS; **CCTU** Culture Collection of Tabriz University, Tabriz, Iran; **GZCC**: Guizhou Culture Collection, Guiyang, China; **MFLUCC:** Mae Fah Luang University Culture Collection, ChiangRai, Thailand.



Note Maximum likelihood bootstrap values equal or greater than 50 and Bayesian posterior probabilities equal or greater than 0.95 are indicated near the nodes. New sequence generated in this study are given in red. The tree is generated from maximum likelihood (ML) analysis based on LSU, ITS, SSU and TEF1-α sequence data, and rooted with *Leptosphaeria doliolum* CBS 505.75 and *Paraleptosphaeria dryadis* CBS 643.86.

Figure 2.44 Phylogenetic tree of the family Phaeosphaeriaceae

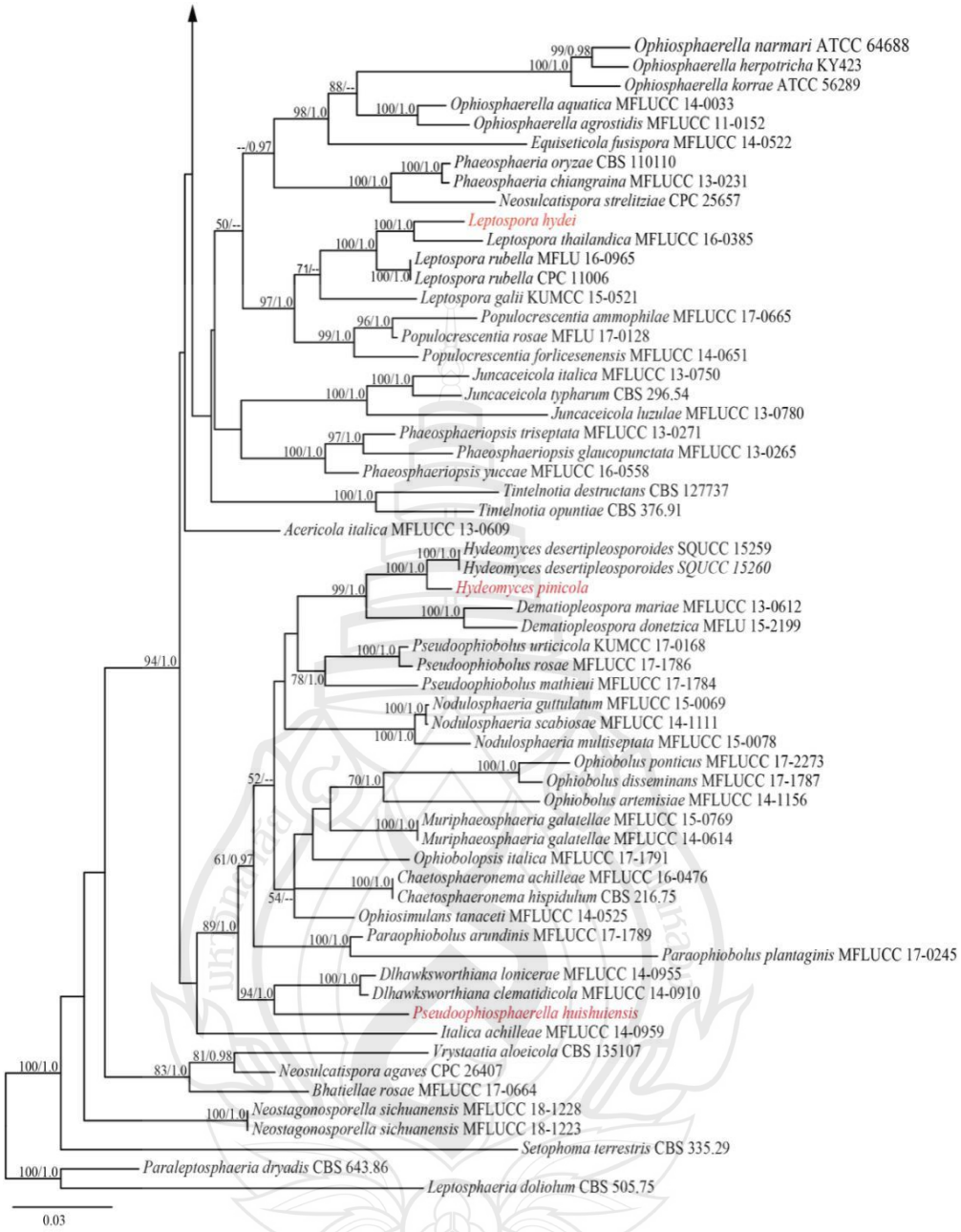


Figure 2.44 (continued)

Profundusphaeriaceae J.F. Zhang & K.D. Hyde, *fam. nov.*

Etymology: Referring to the name of the type genus.

Typus: *Profundusphaeria* J.F. Zhang, J.K. Liu & Z.Y. Liu.

Saprobic on dead culm of unidentified herbaceous plant. Sexual morph: deeply immersed to throughout the surface with a broadly pore-like ostiole, globose to subglobose, dark brown to black, scattered, solitary, apapillate. *Peridium* carbonaceous to coriaceous, composed of several layers of dark pigmented cells or pale pigmented to hyaline cells of *textura angularius*. *Hamathecium* comprising branched, trabeculate, filamentous pseudoparaphyses, anastomosing between and above the asci, intermixed with a gelatinous matrix. *Asci* bitunicate, fissitunicate, cylindrical to clavate, short pedicellate, apically round with a developed ocular chamber. *Ascospores* narrow fusoid, septate. Asexual morph: Undetermined.

Notes: Morphologically, Profundusphaeriaceae is typical of Pleosporales in having perithecioid ascomata with bitunicate, fissitunicate asci bearing mostly septate ascospores. However, it differs from other families of Pleosporales by its ostiole with broadly pore-like opening. Moreover, Profundusphaeriaceae forms an independent clade within Pleosporales in the present phylogenetic analysis (Figure 2.6). Therefore, Profundusphaeriaceae is established as a new family to accommodate the type genus *Profundusphaeria* in this study.

Profundusphaeria J.F. Zhang, J.K. Liu & Z.Y. Liu, gen. nov.

Etymology: From the Latin Profundus refers to deep and sphaeria meaning fruiting body, in reference to the fruiting bodies deeply immersed in the host.

Type species: Profundusphaeria fusiformispora J.F. Zhang, J.K. Liu & Z.Y. Liu.

Saprobic on dead culm of unidentified herbaceous plant. Sexual morph: Ascomata deeply immersed, globose to subglobose, scattered, solitary, apapillate, ostiolate. Ostiole central, with a broadly pore-like opening, and lined with numerous paraphysis in the ostiole canal. Peridium carbonaceous to coriaceous, composed of two strata, with the outer stratum consisting dark pigmented cells of unclear texture, and the inner stratum of several layers of pale pigmented to hyaline, thin-walled cells of *textura angularius*. *Hamathecium* comprising numerous, branched, trabeculate, filamentous pseudoparaphyses, anastomosing between and above the asci, embedded

in a gelatinous matrix. *Asci* 8-spored, bitunicate, fissitunicate, cylindrical to clavate, short pedicellate, apically round with a developed ocular chamber. *Ascospores* overlapping bi-seriate to 3-seriate, narrow fusoid, one-septate, constricted at the septum, asymmetrical, guttulate. Asexual morph: Undetermined.

Notes: *Profundusphaeria* is introduced herein as a monotypic genus to accommodate the type species *P. fusiformispora* based on morphological examination and molecular analysis. And more fresh collections of *Profundusphaeria* are needed to stabilize the placement of this genus and even the family Profundusphaeriaceae in the further study.

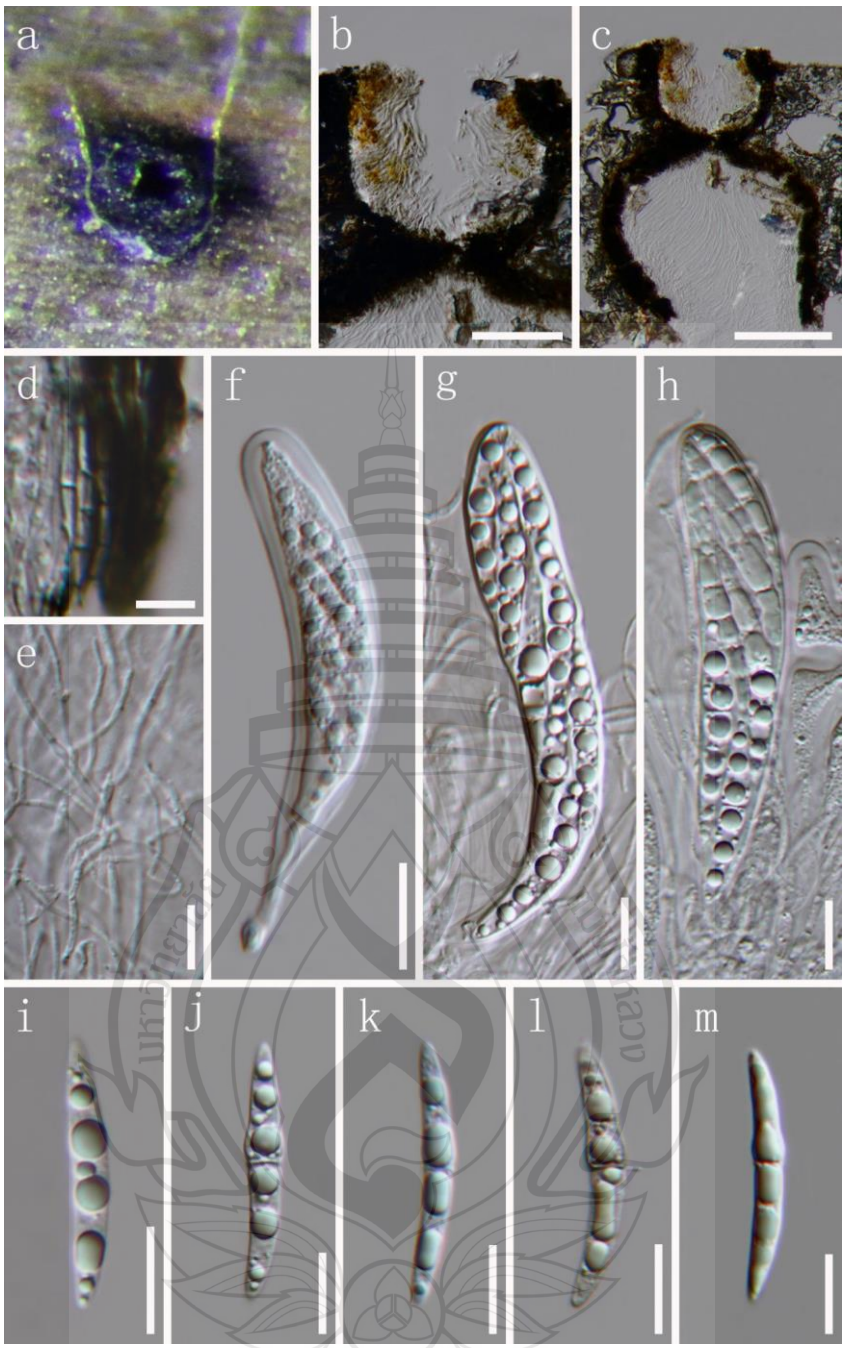
Profundusphaeria fusiformispora J.F. Zhang, J.K. Liu & Z.Y. Liu, *sp. nov.*, Figure 2.45

Etymology: From the Latin *Profundus* refers to deep and *sphaeria* meaning fruiting body, in reference to the fruiting bodies deeply immersed in the host.

Holotype: GZAAS 20-4010.

Saprobic on dead culm of unidentified herbaceous plant, appearing with small, black dots in substrate. Sexual morph: *Ascomata* deeply immersed to throughout the host surface with ostiole, dark brown to black, scattered, solitary, apapillate, ostiolate. *Ostiole* central, with a broadly pore-like opening, and lined with numerous paraphysis in the ostiole canal. *Peridium* up to 50–61 µm wide, composed of two strata, with the outer stratum consisting dark pigmented cells of unclear texture, and the inner stratum of several layers of pale pigmented to hyaline, thin-walled cells of *textura angularius*. *Hamathecium* comprising 1–1.5 µm wide, dense, branched, trabeculate, filamentous pseudoparaphyses, anastomosing between and above the asci, intermixed with a gelatinous matrix. *Asci* (65–) 71–81 (–85) × 11–13 µm ($\bar{x}$ = 77 × 11.5 µm, n = 20), 8-spored, bitunicate, fissitunicate, cylindrical to clavate, short pedicellate, apically round, with a developed ocular chamber. *Ascospores* 27–30.5 (–32) × (3–) 3.5–4.5 (–5) µm ($\bar{x}$ = 30 × 4.2 µm, n = 25), overlapping bi-seriate to 3-seriate, narrow fusoid, straight or curved, hyaline and one celled when young, becoming pale grayish and one septate at maturity, constricted at the septum, asymmetrical, the upper cell is slightly wider than the basal cell, guttulate, without mucilaginous appendages. Asexual morph: Undetermined.

Material examined: CHINA, Guizhou Province, Libo City, Maolan Town, Maolan National Nature Reserve, on dead culm of unidentified herbaceous plant, 20 July 2016, J.F. Zhang MLCC-5 (GZAAS 20-4010, holotype).



Note a Appearance of ascomata deeply immersed in the host. b Ostiole with numerous paraphysis. c Vertical section through ascoma. d Section of peridium. e Pseudoparaphyses. f–h Young and mature asci. i–m Ascospores. Scale bars: b = 100 µm, c = 200 µm, d = 5 µm, e–m = 10 µm.

Figure 2.45 *Profundusphaeria fusiformispora* (GZAAS 20-4010, holotype)

Tetraplosphaeriaceae Kaz. Tanaka & K. Hiray.

Tetraplosphaeriaceae was established by Tanaka et al. (2009) to accommodate five genera (*Polyplosphaeria*, *Pseudotetraploa*, *Quadricrura*, *Tetraplosphaeria* and *Triplosphaeria*) with *Tetraplosphaeria* as the type. However, Hyde et al. (2013) synonymized *Tetraplosphaeria* with *Tetraploa* according to nomenclatural priority. Then, other three genera *viz*. *Byssolophis*, *Ernakulamia* and *Shrungabeeja* were respectively assigned into this family based on morphological and phylogenetic analysis (Delgado et al., 2017; Pem et al., 2019; Hyde et al., 2020a). And this treatment was followed by Hongsanan et al. (2020a) on their monograph for refining the families of Dothideomycetes. Subsequently, Li et al. (2021) established a new hyphomycetous genus *Aquatisphaeria* and placed in Tetraplosphaeriaceae.

Tetraploa Berk. & Broome, Ann. Mag. nat. Hist., Ser. 2 5: 459 (1850)

Tetraploa was erected by Berkeley and Broome (1850) to accommodate *T. aristata*. But Tanaka et al. (2009) redefined *Tetraploa* as *Tetraploasphaeria* based on its massarina-like sexual morph and *Tetraploa sensu stricto* asexual morphs. However, Hyde et al. (2013) resurrected and validated the genus name as *Tetraploa* based on ICN (Art. 59). Until now, twenty-one valid species are recorded in the Index Fungorum (2022).

Tetraploa nagasakiensis (Kaz. Tanaka & K. Hiray.) Kaz. Tanaka & K. Hiray., Fungal Diversity 63(1): 253 (2013) *new record*. Figure 2.46

Saprobic on dead culm of unidentified herbaceous plant, appearing as black spots in host surface. Sexual morph: *Ascomata* immersed under the epidermis, subglobose to globose, dark brown to black, scattered, gregarious, ostiolate, papillate. *Ostiole* central, with periphyses lined within the ostiolar canal. *Peridium* 18–30 µm, composed several layers of thick-walled, brown cells of *texutura angularis*, and become thick and black around the ostiole. *Hamathecium* comprising dense, 1.5–4.5 µm wide, trabeculate, branched pseudoparaphyses, swollen at the base, intermixed with the asci, and embedded in a gelatinous matrix. *Asci* 76–87.5 (–94) × (10.5–) 12–14 (–15.6) µm ($\bar{x}$ = 83.3 × 13 µm, n = 20), 8-spored, bitunicate, fissitunicate, cylindric-clavate, with a moderate, furcate pedicel, apically rounded, ocular chamber distinct. *Ascospores* (26.5–) 29–31.2 (–33.5) × 4.5–5.5 µm ($\bar{x}$ = 30.5 × 4.7 µm, n = 25), fusiform, straight or slightly curved, guttulate, one-celled when young and

production one septum upon maturation, slightly constricted at the septum, wall smooth. Asexual morph: Undetermined.

Material examined: CHINA, Guizhou Province, Kaiyang County, on dead culm of unidentified herbaceous plant, 26 July 2017, H.Y. Ran KY-07 (MFLU 18-2254).

Notes: Present phylogenetic analysis shows that our new collection cluster to the holotype of *T. nagasakiensis* (HHUF 29378) by full bootstrap support (Figure 2.6). Morphologically, this specimen is also particularly similar to *T. nagasakiensis* by having overlap sized asci and ascospores (Tanaka et al., 2009; Hyde et al., 2013). However, the feature of ascospores with bipolar elongated sheath at both ends was now found in our specimen, but clearly presented at the photoplate of the holotype (Tanaka et al., 2009). We thought that this difference may be attributed to the unequal maturation state of these two specimens, and not enough to warrant them as separate species. Hence, the newly collected specimen was identified as *T. nagasakiensis*, and reported as a new geographical record herein as it has only been found from Japan.



Note a Appearance of ascomata in the host surface. b Vertical section through ascomata. c Section of peridium. d Pseudoparaphyses. e–g Asci. h–l Ascospores. Scale bars: b = 100 µm, c = 10 µm, d–g = 20 µm, h–l = 10 µm.

Figure 2.46 *Tetraploa nagasakiensis* (MFLU 18-2254, new record)

Thyridariaceae Q. Tian & K.D. Hyde

Thyridariaceae was introduced as a monotypic family to accommodate the type genus *Thyridaria* based on morphology and phylogeney (Hyde et al., 2013). Subsequently, Jaklitsch and Voglmayr (2016) emended this family and synonymized Roussoellaceae under Thyridariaceae, as well as placed a genus *Parathyridaria* into this family. However, Tibpromma et al. (2017b) reinstated Roussoellaceae and distinguished from Thyridariaceae based on the significant morphological differences and sufficient phylogenetic analysis. Then, other five genera *viz*. *Chromolaenomyces*, *Cycasicola*, *Liua*, *Pseudothyridariella*, *Thyridariella* were separately included in this family by various authors (Devadatha et al., 2018; Wanasinghe et al., 2018a; Phookamsak et al., 2019; Mapook et al., 2020). And this treatment was followed by the latest outline of fungi (Wijayawardene et al., 2022) and the updated document on families of Dothiodeomycetes (Hongsanan et al., 2020a).

Thyridaria Sacc., Grevillea 4(no. 29): 21 (1875)

Thyridaria was introduced with *T. incrustans* as the type species. Then, the classification placement of this genus has always been argumented by various authors (Munk, 1957; Luttrell, 1973; Müller & Arx, 1973; Barr, 1979a, b). Barr (2003) summarized the varied description of *Thyridaria* and concluded that *Thyridaria sensu stricto* should be assigned in Didymosphaeriaceae. However, this was not followed by Lumbsch & Huhndorf (2010) but placed it in Dothideomycetes, genera *incertae sedis*. Even recent phylogenetic studies showed that *Thyridaria* is a distinct group within Pleosporales (Damm et al., 2008; Schoch et al., 2009a, b; de Gruyter et al., 2013), however no family is available to accommodate this genus until Hyde et al. (2013) introduced Thyridariaceae. Currently, more than 50 *Thyridaria* species included in Index Fungorum (2022).

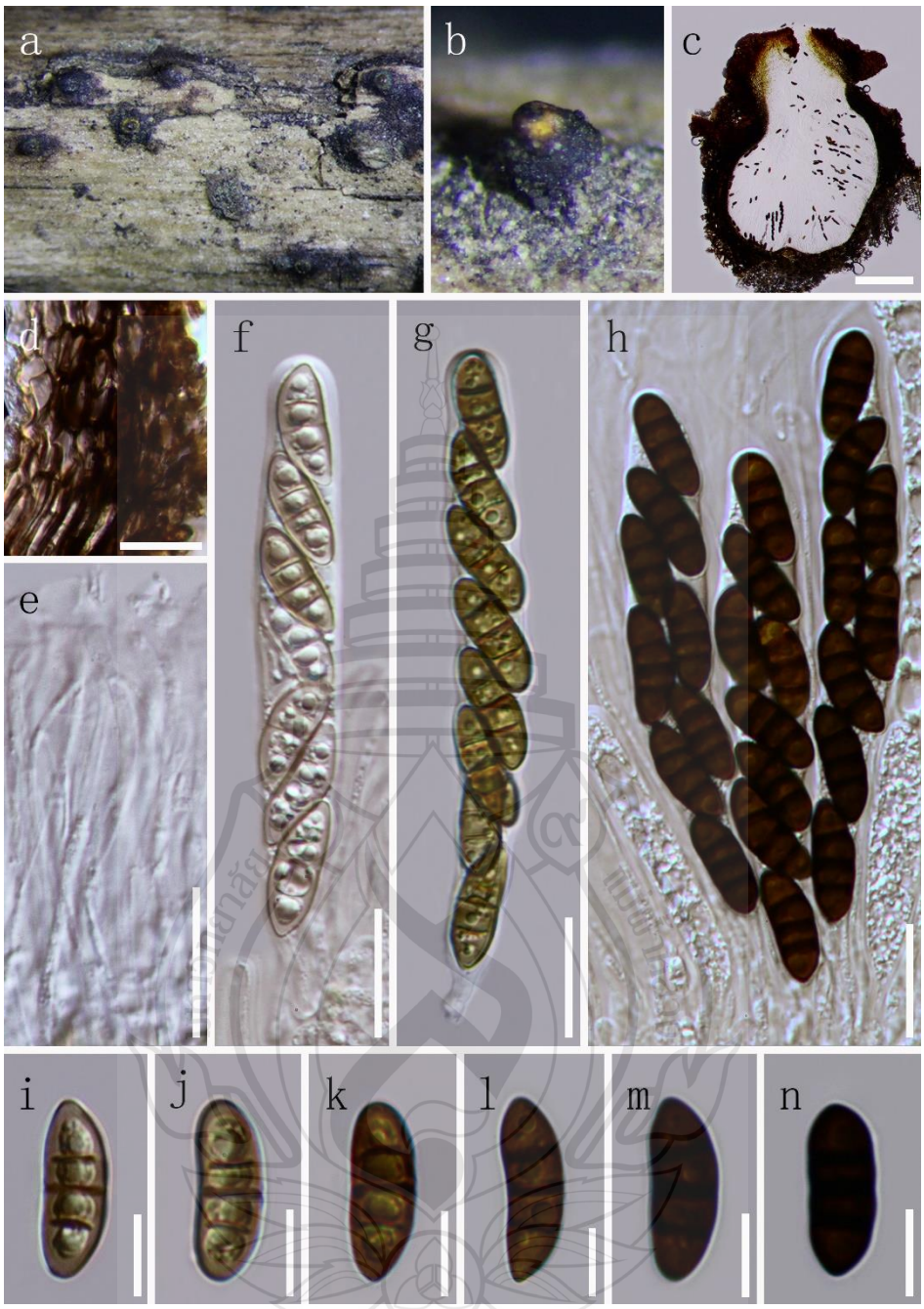
Thyridaria broussonetiae (Sacc.) Traverso, in Traverso, Fl. ital. crypt., Pars 1: Pyrenomycetae. Xylariaceae, Valsaceae, Ceratostomataceae (Florence) 1(2): 301 (1906). *new record*. Figure 2.47

Saprobic on dead branch of unidentified woody plant. Sexual morph: *Ascomata* immersed to erumpent throughout the host, aggregated, clustered, globose to subglobose, uni-loculate, dark brown to black, ostiolate. *Ostiole* central, with a pore-like opening, and usually a pruinose layer of yellow to orange-brown material

appearing around the apices of the ostiolar. *Peridium* 35–65 µm wide, comprising several layers of thick-walled, pseudoparenchymatous cells of *textura angularis*, pale pigmented to hyaline towards inwardly. *Hamathecium* comprising 1–2 µm wide, filamentous pseudoparaphyses, anastomosing between and above the asci, embedded in a gelatinous matrix. *Asci* 109–119 × 13–16.5 µm ($\bar{x}$ = 112 × 14.5 µm, n = 15), 8-spored, bitunicate, cylindrical to cylindric-clavate, short pedicellate, apically rounded, with a distinct ocular chamber. *Ascospores* 19–22(–26.5) × 7.5–9 µm ($\bar{x}$ = 21.5 × 8 µm, n=25), overlapping uni- to bi-seriate, ellipsoidal to fusiform, straight or slightly curved, pale or yellowish brown when young, dark to blackish brown at maturity, 3-septate with one median euseptum and 2 additional incomplete septa, not constricted at the septa, end cells often slightly longer, verruculose, without mucilaginous appendages. Asexual morph: Undetermined.

Material examined: CHINA, Guizhou Province, Guiyang City, Yunyan District, Guizhou Botanical Garder, on dead branch of unidentified woody plant, 8 August 2017, J.F. Zhang ZWY-08 (MFLU 18-2243).

Notes: Our new collection clusters with the type species *T. broussonetiae* (CBS 141481) by strong statistical support in this study (Figure 2.6). Observation of the morphological characteristics of this new specimen, we found that it is typical of *Thyridaria*, and well fits to *T. broussonetiae* in having overlap sized asci and ascospors (Jaklitsch & Voglmayr, 2016). Therefore, this new collection is identified as *Thyridaria broussonetiae*, based on the above morphological and phylogenetic analysis, and reported as a new geographical record as it has only been discovered from Southern European.



Note a, b Appearance of ascomata in the host surface. c Vertical section through ascomata. d Section of peridium. e Pseudoparaphyses. f–h Asci in different maturation stage. i–n Ascospores in different maturation stage. Scale bars: c = 200 µm, d–h = 20 µm, i–n = 10 µm.

Figure 2.47 *Thyridaria broussonetiae* (MFLU 18-2243, new record)

Dothideomycetes orders *incertae sedis*

Botryosphaeriales C.L. Schoch et al.

Botryosphaeriales was introduced by Schoch et al. (2006) to accommodate Botryosphaeriaceae because of the multi-gene analysis did not place this family within any reported orders within Dothideomycetes. Minnis et al. (2012) and Wikee et al. (2013) subsequently assigned two families Planistromellaceae and Phyllostictaceae into Botryosphaeriales based on morphology and phylogeny, and this agreement with Liu et al. (2012) who indicated that more than one family (Botryosphaeriaceae) should be comprised of this order. Then, other six families *viz.* Aplosporellaceae, Endomelanconiopsisaceae, Melanopsaceae, Pseudofusicoccumaceae, Saccharataceae and Septorioideaceae were accepted in this order based solely on phylogenetic analyses by various authors (Slippers et al., 2013; Wyka & Broders, 2016; Yang et al., 2017b). However, Phillips et al. (2019) re-assessed the families of Botryosphaeriales in terms of morphology, phylogeny and evolution analysis, and reduced three families (Endomelanconiopsisaceae, Pseudofusicoccumaceae and Septorioideaceae) to synonym under existing families. This classification placement was followed by Hongsanan et al. (2020b) in their refined updated document on orders and families *incertae sedis* of Dothideomycetes.

Botryosphaeriaceae Theiss. & Syd.

Botryosphaeriaceae was introduced by Theissen and Sydow (1918) with *Botryosphaeria* as the typus. Since then, the members of genera in this family were increased and reduced by various authors (Von Arx & Müller, 1954; Barr, 1987; Hawksworth et al., 1995; Lumbsch & Huhndorf, 2010; Liu et al., 2012; Linaldeddu et al., 2016; Wijayawardene et al. (2018) accepted 28 genera within this family. However, Phillips et al. (2019) restricted to 22 genera on the basis of morphology and phylogenetic relationships. This treatment was followed by Hongsanan et al. (2020b) and the latest outline of fungi (Wijayawardene et al., 2022).

Botryosphaeria Ces. & De Not., Comm. Soc. crittog. Ital. 1(fasc. 4): 211 (1863)

Botryosphaeria was introduced by De Cesati & De Notaris (1863) to accommodate 12 species, but without providing the detail morphological descriptions and designation the type species. Over a hundred years later, Barr (1972) designated *B.*

dothidea as the lectotype of this genus. Subsequently, Slippers et al. (2004) designated the only remaining sample of *Sphaeria dothidea* from Fries herbarium as the neotype representing *B. dothidea*, however, this specimen is not definitive as it is immature. Therefore, an epitype was designated in their study to complement the neotype and other authentic specimens of *B. dothidea*, based on morphological characteristics, in combination with molecular data. Recently, there are more than 200 peithets listed in Index Fungorum (2022), however, only 8 species were accepted in the latest publication (Zhang et al., 2021a).

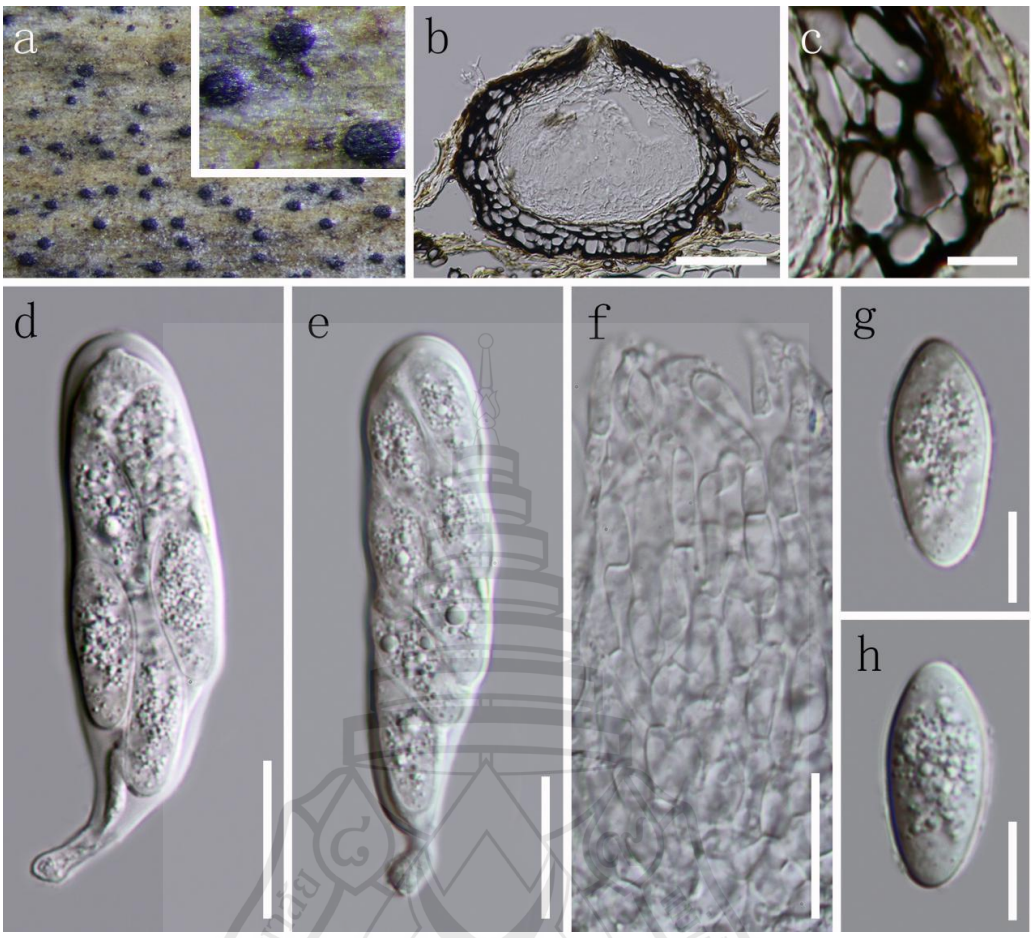
Botryosphaeria dothidea (Moug.) Ces. & De Not., Comm. Soc. crittog. Ital: 212 (1863) Figure 2.48

Saprobic on dead culm or branch of herbaceous and woody plant. Sexual morph: *Ascomata* semi-immersed to erumpent, globose, dark brown to black, forming a botryose aggregate, sometimes solitary, with a central ostiole, emergent or embedded, papillate. *Peridium* 20–30 μm, comprised of 4–7 layers of *textura angularis*, outer layer of dark brown, thick-walled cells, and inner layer of hyaline cells lining the locule. *Hamathecium* comprising numerous, 3–5 μm wide, septate, cellular pseudoparaphyses, swollen at the base, anastomosing between the asci. *Asci* 68.5−80(−97.5) × 19–22.5 μm ($\bar{x}$ = 80 × 20 μm, n = 20), 8-spored, bitunicate, clavate, rounded at the apex and with a distinct ocular chamber, short pedicellate. *Ascospores* 21–27 × 8.5–11 μm ($\bar{x}$ = 23.5 × 9.5 μm, n = 25), fusoid to ovoid, hyaline, bi- to tri-seriate smooth walled, guttulate, without mucilaginous sheath. Asexual morph: Undetermined.

Material examined: CHINA, Guizhou Province, Danzhai County, on dead culm of unidentified herbaceous plant, 29 July 2017, J.F. Zhang DZ-08 (MFLU 18-2259); Kaiyang County, on dead branch of unidentified woody plant, 26 July 2017, H.Y. Ran KY-05 (MFLU 18-2253); on dead branch of unidentified woody plant, 2 July 2019, H.Y. Ran KY-2019-5 (GZAAS 21-2010); on dead branch of unidentified woody plant, 2 July 2019, H.Y. Ran KY-2019-8 (GZAAS 21-2012); Leishan County, on dead branch of unidentified woody plant, 2 November 2017, J.F. Zhang LS-2 (GZAAS 21-2002); on dead branch of unidentified woody plant, 2 November 2017, J.F. Zhang LS-18 (GZAAS 21-2011); on dead branch of unidentified woody plant, 24 May 2019, J.F. Zhang LS-2019-13 (GZAAS 19-1743); Sinan County, on dead branch

of unidentified woody plant, 22 May 2019, J.F. Zhang SN-2019-10 (GZAAS 21-2013); on dead branch of unidentified woody plant, 22 May 2019, J.F. Zhang SN-2019-11 (GZAAS 21-2014).

Notes: Present phylogenetic analysis shows that our new collections cluster together with *Botryosphaeria dothidea* and *B. guttulata* by high bootstrap support (Figure 2.50). Morphologically, these new specimens resemble with the sexual morph of *B. dothidea* (PREM57372, epitype) that described by Liu et al. (2012), and all of the measurements of microscopic structures also fit with the latter. Therefore, these collections are identified as *B. dothidea*. Additionally, Zhang et al. (2021a) had evaluated several species in Botryosphaeriales, and the topology in our study (Figure 2.50) is quite similar with their analysis. Based on their new insights for the boundaries of *B. dothidea*, the species *B. guttulata* maybe a new synonym, and this will be confirmed in the further study.



Note a Appearance of ascomata in host surface. b Vertical section through ascoma. c Section of peridium. d, e Asci. f Pseudoparaphyses. g, h Ascospores. Scale bars: b = 50 μm, c, g, h = 10 μm, d–f = 20 μm.

Figure 2.48 *Botryosphaeria dothidea* (MFLU 18-2259)

Neofusicoccum Crous, Slippers & A.J.L. Phillips, Stud. Mycol. 55: 247 (2006)

Neofusicoccum was introduced by Crous et al. (2006) to accommodate the species that differs from *Fusicoccum* species and designated *N. parvum* as the type species. Then, the accounts of this genus were continually being discovered by various authors (Pavlic et al., 2009; Begoude et al., 2010; Crous et al., 2011a; Marques et al., 2013; Berraf-Tebbal et al., 2014; Chen et al., 2015; Marin-Felix et al., 2017; Yang et al., 2017b; Zhang et al., 2017d; Li et al., 2018, 2020a; Tibpromma et al., 2018; Tennakoon et al., 2021). Recently, Zhang et al. (2021a) evaluated the

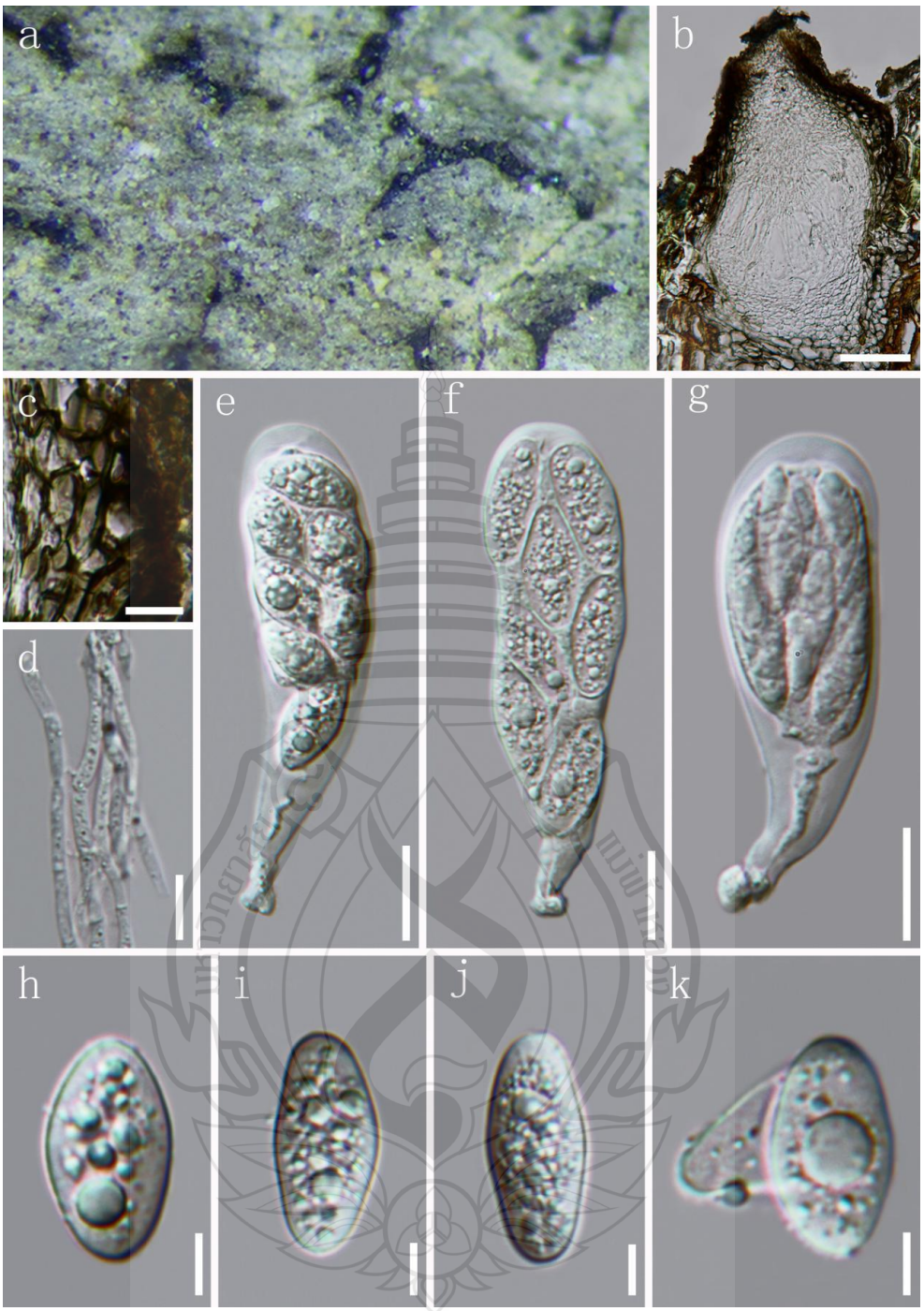
species of Botryosphaeriales based on morphological and multi-gene analysis, and restricted to 36 species within *Neofusicoccum.*

Neofusicoccum parvum (Pennycook & Samuels) Crous, Slippers & A.J.L. Phillips, in Crous et al., Stud. Mycol. 55: 248 (2006) Figure 2.49

Saprobic on decaying wood. Sexual morph: *Ascostromata* semi-immersed to erumpent under epidermis, individually globose to subglobose, solitary or clustered, longitudinal axis vertical to the host surface, visible as minute black dots or papilla on host tissue. *Locules* 171–283 μm diam, 214–302 μm high. *Ostiole* central, circular, papillate. *Peridium* of locules up to 18–44 μm wide, comprised of two stratum, outer layer of dark brown to black, thick-walled cells of pseudoparenchymatous, inner layer of pale brown to hyaline, thin-walled cells of *textura angularis*. *Hamathecium* comprising numerous, 2–3.5 μm wide, trabeculate, hyphae-like pseudoparaphyses, anastomosing between the asci. *Asci* (74−)79.5−87.5 × 22.5−24.5 μm ($\bar{x}$ = 82 × 23.5 μm, n = 15), 8–spored, bitunicate, fissitunicate, broadly clavate, with a short, furcate pedicel, apically rounded with a distinct ocular chamber. *Ascospores* (17−)19−23.5(−25) × 8−11(−13) μm ($\bar{x}$ = 20.5 × 10 μm, n = 25), uni-seriate at the base, 2–3-seriate at the apex, hyaline, aseptate, ellipsoidal to fusiform, guttulate. Asexual morph: Undetermined.

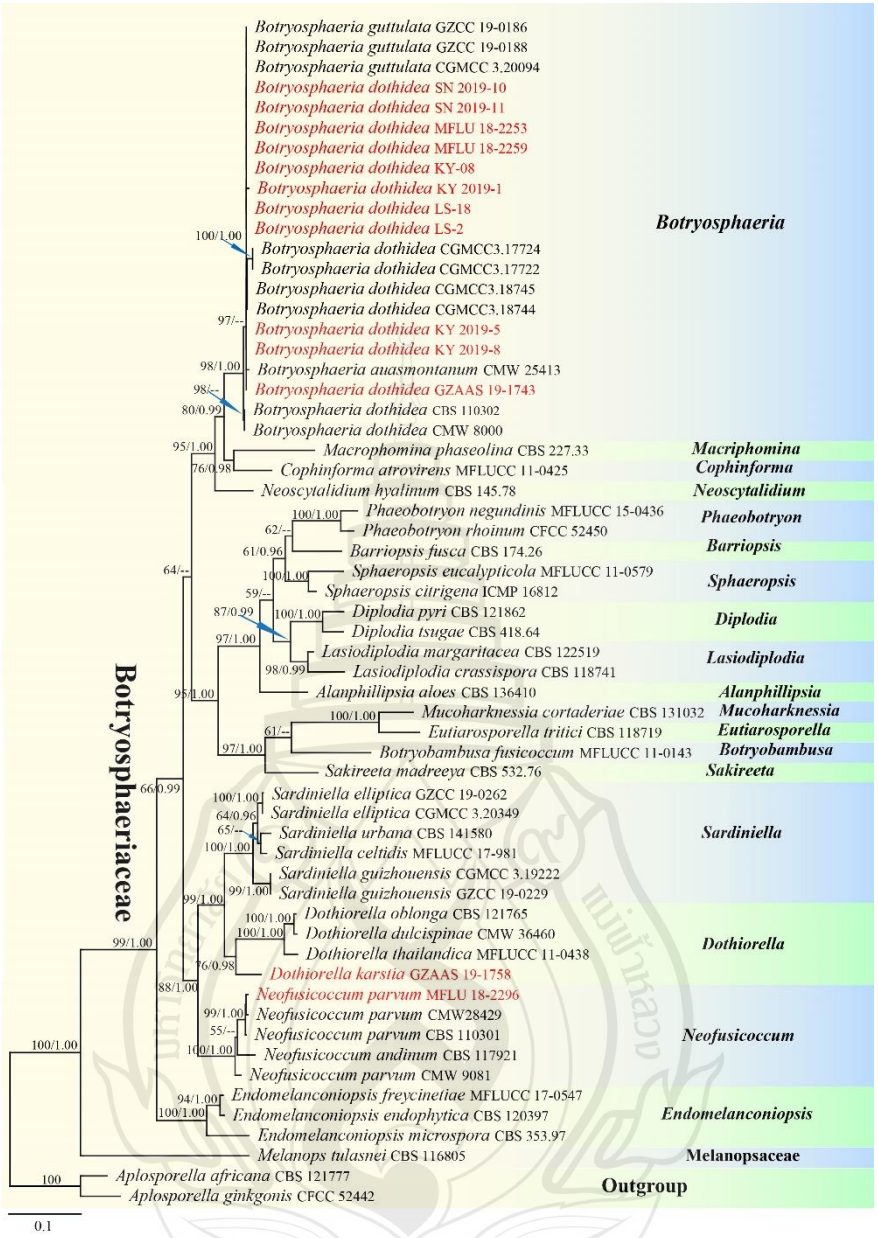
Material examined: CHINA, Guizhou Province, Guiyang City, Huaxi District, Guizhou Academy of Agricultural Sciences, on decaying wood, 5 October 2016, J.F. Zhang GZ-14 (MFLU 18-2296).

Notes: The new collection is assigned within Botryosphaeriaceae, and clustered together with *N. parvum* by strong statistical support in the present study (Figure 2.50). Moreover, our new specimen is morphologically similar to the sexual morph of *N. parvum* that described by Liu et al. (2012) in having similar shaped and overlap sized asci and ascospores. Therefore, the new collection in this study is identified as *N. parvum* based on morphological examination and molecular analysis.



Note a Appearance of ascostromata in host surface. b Vertical section through ascoma. c Section of peridium. d Pseudoparaphyses. e–g Asci. h–k Ascospores. Scale bars: b = 100 µm, c, d = 10 µm, e–g = 20 µm, h–k = 5 µm.

Figure 2.49 *Neofusicoccum parvum* (MFLU 18-2296)



Note Maximum likelihood bootstrap values equal or greater than 50 and Bayesian posterior probabilities equal or greater than 0.95 are indicated near the nodes. New sequence generated in this study are given in red. The tree is generated from maximum likelihood (ML) analysis based on ITS and TEF1-α sequence data, and rooted with *Aplosporella africana* CBS 121777 and *A. ginkgonis* CFCC 52442.

Figure 2.50 Phylogenetic tree of the family Botryosphaeriaceae

Dothiorella Sacc., Michelia 2 (no. 6): 5 (1880).

The genus *Dothiorella* has always been confused with *Diplodia* until Phillips et al. (2005) distinguished these two genera by examination the type species, and resurrected *Dothiorella* to accommodate species with conidia that turn brown while still attached to their conidiogenous cells in asexual state. And rarely sexual morphs (characterized by immersed to erumpent, ostiolate, papillate ascomata, with cylindric-clavate, bitunicate, fissitunicate, pedicellate asci that bearing brown, one-septate, oblong to ovoid ascospores) of this genus are reported from nature (Phillips et al., 2005). Recently, Zhang et al. (2021a) evaluated the *Dothiorella* species by employing multi-gene analysis, and recognized 27 species with several possible synonyms.

Dothiorella sexualis J.F, Zhang & J.K. Liu, *sp. nov*., Figure 2.51

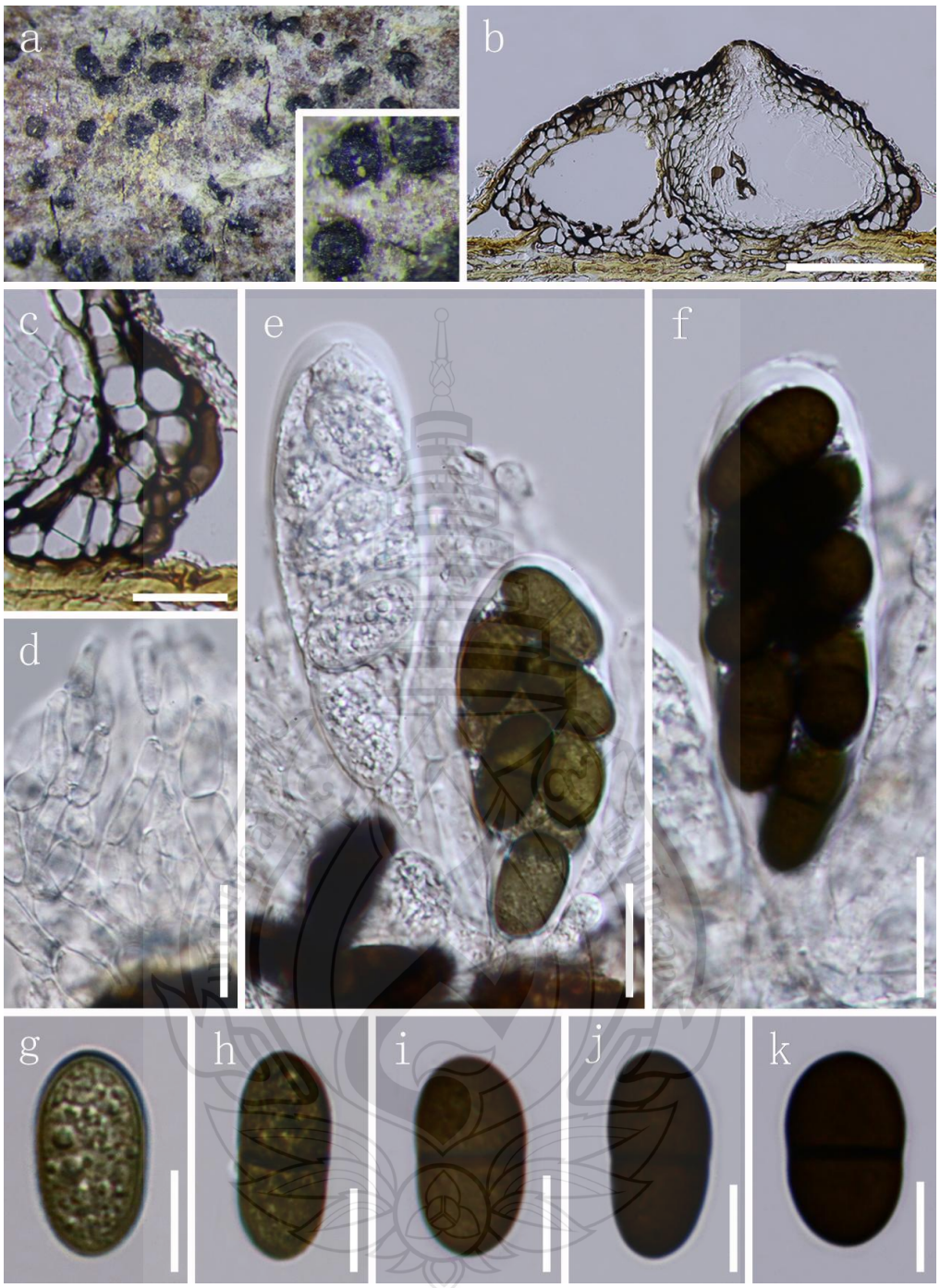
Etymology: Named refers to the holotype is found in sexual morph from nature.

Holotype: GZAAS 19-1758.

Saprobic on dead wood. Sexual morph: *Ascomata* semi-immersed to erumpent the epidermis, individually globose to subglobose, solitary or clustered, visible as minute black dots. *Ostiole* central, circular, papillate. *Peridium* of locules 29–43 μm wide, comprised of two stratum, outer stratum of dark brown to black, thick-walled cells of *textura angularis*, inner stratum of hyaline, thin-walled cells of *textura angularis*. *Hamathecium* comprising numerous, 3.5–8 μm wide, septate, cellular pseudoparaphyses, constricted at the septum, swollen at the base, anastomosing between the asci. *Asci* 78−85(−95) × 25−28 μm ($\overline{x}$ = 84.5 × 26.5 μm, n = 15), 8–spored, bitunicate, fissitunicate, clavate to cylindric-clavate, short pedicellate, apically rounded with developed ocular chamber. *Ascospores* 21−25 × 11−13 μm ($\overline{x}$ = 22.5 × 12 μm, n = 25), uni-seriate at the base, 2–3-seriate at the apex, hyaline to pale brown, aseptate when young, become dark brown to black and two-celled at maturation, broadly fusiform to ellipsoidal, smooth-walled. Asexual morph: Undetermined.

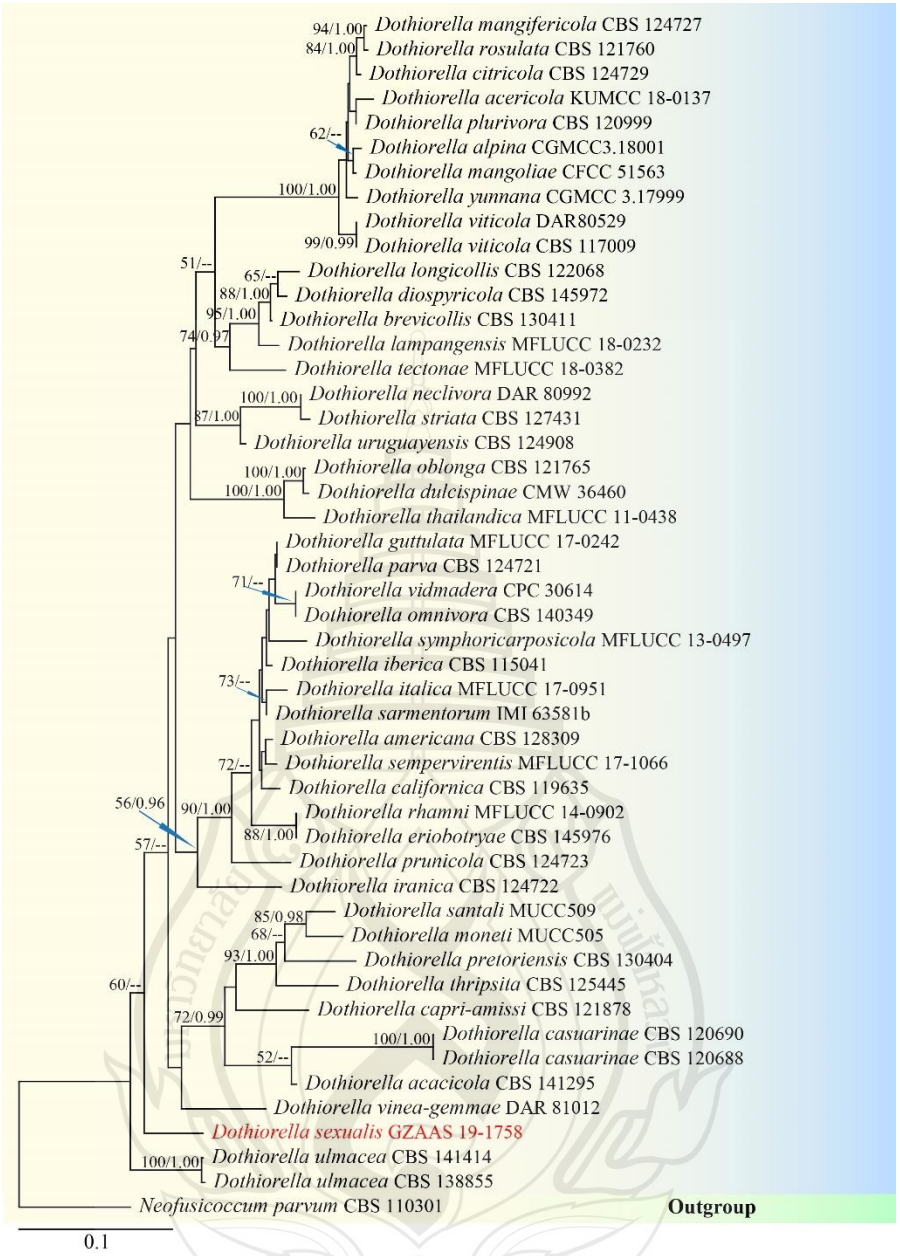
Material examined: CHINA, Guizhou Province, Guiyang City, Huaxi District, Guizhou Academy of Agricultural Sciences, on dead wood of unidentified host, 5 November 2017, J.F. Zhang GZAAS-2017-6 (GZAAS 19-1758, holotype).

Notes: *Dothiorella sexualis* is placed in the basal of *Dothiorella* with a separate subclade in the present study (Figure 2.52), and the topology of our analysis is somewhat similar to the latest study from Zhang et al. (2021). Morphologically, *D. sexualis* resembles to *D. iberica* and *D. sarmentorum* in having cylindric-clavate asci and one-septate, dark pigmented ascospores. However, these three taxa can be distinguished from one another based on the measurements of asci and the shape of ascospores. Asci of *D. sexualis* are obvious smaller than *D. iberica* and *D. sarmentorum* (*D. sexualis* 78−85(−95) × 25−28 μm vs. *D. iberica* 100−125 × 18−25 μm vs. *D. sarmentorum* 140−210 × 17−24 μm), and the ascospores of *D. iberica* are observed tapering towards the basal cell, while this is not found in *D. sexualis* (Phillips et al., 2005).



Note a Appearance of ascostromata in host surface. b Vertical section through ascoma. c Section of peridium. d Pseudoparaphyses. e, f Asci. g–k Ascospores. Scale bars: b = 100 μm, c–f = 20 μm, g–k = 10 μm.

Figure 2.51 *Dothiorella sexualis* (GZAAS 19-1758, holotype)



Note Maximum likelihood bootstrap values equal or greater than 50 and Bayesian posterior probabilities equal or greater than 0.95 are indicated near the nodes. New sequence generated in this study are given in red. The tree is generated from maximum likelihood (ML) analysis based on ITS and TEF1-α sequence data, and rooted with *Neofusicoccum parvum* CBS 110301.

Figure 2.52 Phylogenetic tree of the genus *Dothiorella*

Dyfrolomycetales K. L. Pang et al.

Dyfrolomycetales was established as a marine order to accommodate Dyfrolomycetaceae with *Dyfrolomyces* as the typus. However, Maharachchikumbura et al. (2016) synonymized Dyfrolomycetaceae under Pleurotremataceae, and removed Pleurotremataceae from Sodariomycetes to be a single family of Dyfrolomycetales, based on the examination of isotype of *Pleurotrema polysemum*. So far, there are only the single family Pleurotremataceae with three genera *viz*. *Dyfrolomyces*, *Melomastia* and *Pleurotrema* accepted in Dyfrolomycetales (Hongsanan et al., 2020b; Wijayawardene et al., 2022).

Pleurotremataceae Walt. Watson

The modern classification placement of Pleurotremataceae was revised by Maharachchikumbura et al. (2016), based on examination the isotype of *P. polysemum*. In their study, the family Dyfrolomycetaceae (Dyfrolomycetales) was synonymized under Pleurotremataceae, and removed this family from Sordariomycetes to be a single family of Dyfrolomycetales. Pleurotremataceae comprises three genera *viz*. *Dyfrolomyces*, *Melomastia* and *Pleurotrema*.

Dyfrolomyces K.D. Hyde, K.L. Pang, Alias, Suetrong & E.B.G. Jones, Cryptog. Mycol. 34(3): 227 (2013).

Dyfrolomyces was introduced by Pang et al. (2013) to accommodate the type species *D. tiomanensis* with three new combinations *D. mangrovei, D. marinospora* and *D. rhizophorae*, based on the morphological and phylogenetic analysis. *Dyfrolomyces* species are normally characterized by relatively large, immersed, globose or subglobose, clypeate, papillate ascomata, cylindrical, 8-spored, bitunicate, short pedicellate asci with a ring-like subapical apparatus, and broadly fusiform, symmetrical, hyaline, overlapping uniseriate, multi-septate ascospores (Pang et al., 2013).

Dyfrolomyces maolanensis J.F. Zhang, J.K. Liu, K.D. Hyde & Z.Y. Liu, *sp. nov*.

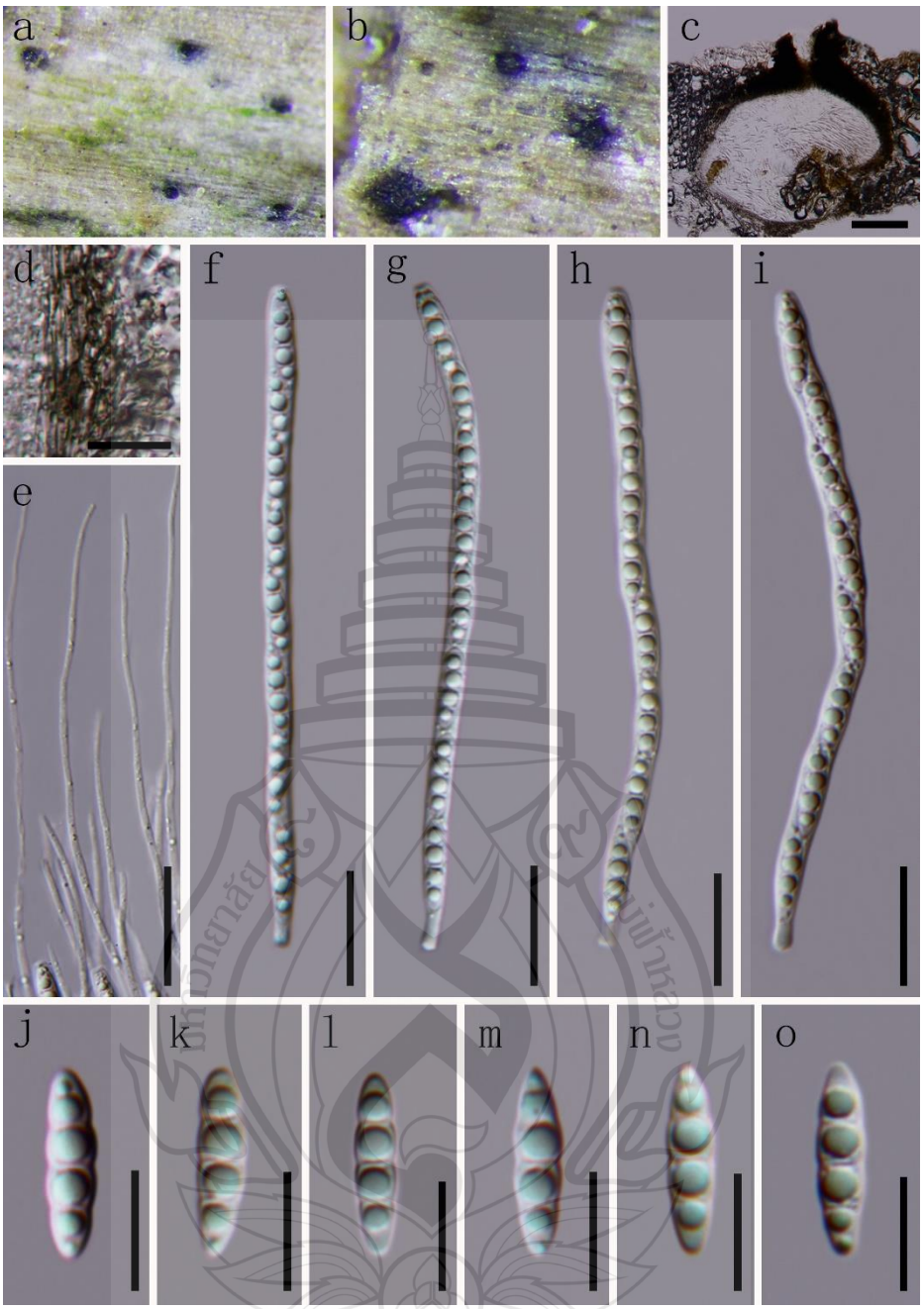
Index Fungorum number: IF 552554; Facesoffungi number: Fof 02695 Figure 2.53

Etymology: In reference to the collection site from where this fungus was collected, Maolan Town, China.

Holotype: GZAAS 16-0114.

Saprobic on dead branch. Sexual morph: *Ascomata* scattered or gregarious, visible on the host surface as dark, raised spots, black, immersed, clypeate, subglobose to obpyriform, coriaceous to carbonaceous, ostiolate, papillate. *Clypeus* 33–56 µm thick, extending outwards around the ascomata, thicker around the papilla, composed of dense, melanized cells. *Papilla* blunt conical or opening-like, periphyses not seen. *Peridium* composed of several layers of pale brown cells of *textura angularis*, becoming hyaline towards inner side and fusing with tissue on outer side. *Hamathecium* comprising long, sparse, 1–2 µm wide, hypha-like pseudoparaphyses, attached at the base and between the asci, embedded in a matrix. *Asci* (95–) 103–118 (–122) × 4–5.5 µm ($\bar{x}$ = 110.9 × 4.9 µm, n = 20), 8-spored, long cylindrical, short-pedicellate, apically rounded to truncate with a ring-like apparatus. *Ascospores* 13.5–18 × 3.5–4.5 µm ($\bar{x}$ = 16.2 × 3.9 µm, n = 30), overlapping-uniseriate, hyaline, fusiform with round ends, normally 3-septate, constricted at the septum, smooth-walled, containing several guttules when young. Asexual morph: Undetermined.

Material examined: CHINA, Guizhou Province, Maolan Town, on dead branch, 20 July 2016, J.F. Zhang, MLA'-2, (GZAAS 16-0114, holotype); ex-type living culture, GZCC 16-0102; Ibid., 10 November 2016, J.F Zhang (MFLU 16-2666); living culture MFLUCC 16-1375.



Note a, b Appearance of ascomata immersed in the host. c Vertical section through ascoma. d Section of peridium. e Pseudoparaphyses. f–i Long, cylindrical asci with ascospores. j–o Ascospores. Scale bars: c = 100 µm, d, f–i = 20 µm, e = 30 µm, j–o = 10 µm.

Figure 2.53 *Dyfrolomyces maolanensis* (GZAAS 16-0114; holotype)

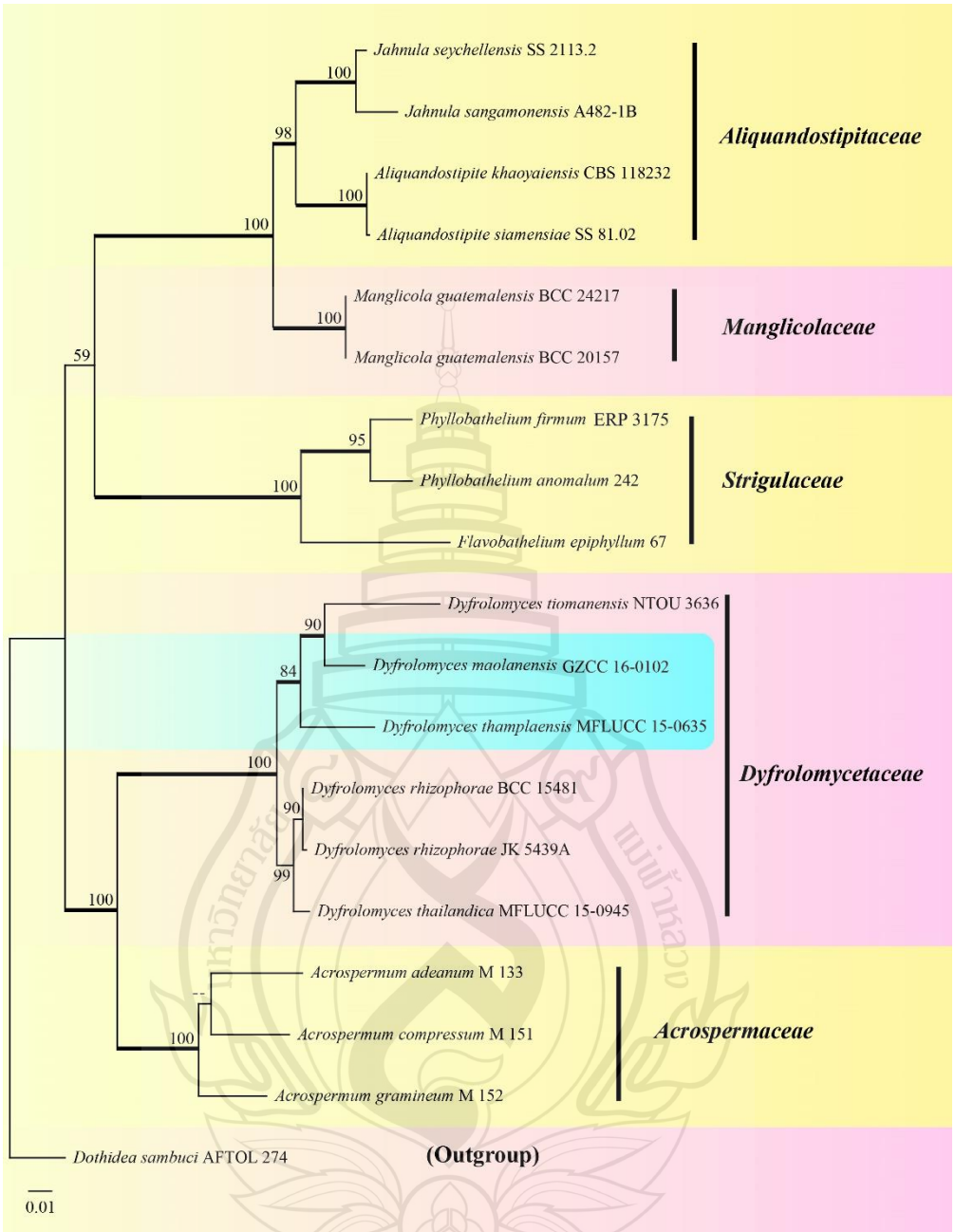
Table 2.4 Taxa used in this study. Strains generated/sequenced in this study are given in bold

Species	Specimen/Strain No.	LSU	SSU	TEF1-α
Acrospermum adeanum	M 133	EU940104	EU940031	---
Acrospermum compressum	M 151	EU940084	EU940012	---
Acrospermum graminum	M 152	EU940085	EU940013	---
Aliquandostipite siamensiae	SS 81.02	EF175666	EF175645	---
Aliquandostipite khaoyaiensis	CBS 118232	GU301796	AF201453	GU349048
Dothidea sambuci	AFTOL 274	AY544681	AY544722	DQ497606
Dyfrolomyces maolanensis	**GZCC 16-0102**	**KY111905**	**KY111906**	**KY814762**
Dyfrolomyces rhizophorae	BCC 15481	---	KF160009	---
Dyfrolomyces rhizophorae	JK 5439A	GU479799	GU479766	GU479860
Dyfrolomyces thailandica	MFLUCC 15-0945	KX611366	KX611367	---
Dyfrolomyces thamplaensis	MFLUCC 15-0635	KX925435	KX925436	KY814763
Dyfrolomyces tiomanensis	MFLUCC 13-0440	KC692156	KC692155	KC692157
Flavobathelium epiphyllum	67	GU327717	---	---
Jahnula sangamonensis	A 482 -1B	EF175662	EF175640	---
Jahnula seychellensis	SS 2113.2	EF175664	EF175643	---

Table 2.4 (continued)

Species	Specimen/Strain No.	LSU	SSU	TEF1-α
Manglicola guatemalensis	BCC 20157	FJ743450	FJ743444	---
Manglicola guatemalensis	BCC 24217	FJ743447	J743441	---
Phyllobathelium anomalum	242	GU327722	GU327698	---
Phyllobatheliumfirmum	ERP 3175	GU327723	---	---

Note AFTOL-ID: Assembling the Fungal Tree of Life: **BCC**: Belgian Coordinated Collections of Microorganisms: **CBS**: CBS-KNAW Fungal Biodiversity Centre, Utrecht, The Netherlands; **DAOM**: Canadian Collection of Fungal Cultures, Ottawa, Canada; **GZCC**: Guizhou Culture Collection, Guiyang, China; **MAFF**: Ministry of Agriculture, Forestry and Fisheries, Japan; **MFLUCC:** Mae Fah Luang University Culture Collection, ChiangRai, Thailand; **JK**: J. Kohlmeyer.



Note Maximum likelihood bootstrap values equal or greater than 50 are shown near the nodes and branches with Bayesian posterior probabilities equal or greater than 0.95 are given in bold. The tree is generated from maximum likelihood (ML) analysis based on LSU, SSU and TEF1-α sequence data, and rooted with *Dothidea sambuci* AFTOL 274.

Figure 2.54 Phylogenetic tree of the family Dyfrolomycetaceae

Monoblastiales Lücking et al.

Monoblastiales was established as a lichenized fungi group to accommodate Monoblastiaceae within class Dothideomycetes based on morphological and phylogenetic analysis (Hyde et al., 2013). It is morphologically differentiated from other two lichenized orders Strigulales and Trypetheliales in the net-like physes, euseptate ascospores and macroconidia arranged in a gelatinous matrix (Hyde et al., 2013). Monoblastiales has always been a monotypific order as it only contains the single family Monoblastiaceae (Hyde et al., 2013; Hongsanan et al., 2020b; Wijayawardene et al., 2020). In this study, the family Eriomycetaceae is resurrected and accommodated within Monoblastiales based on morphological characteristics and phylogenetic analysis.

Eriomycetaceae Huanraluek & K.D. Hyde.

Eriomycetaceae was established to accommodate five non-lichenized genera that include the typus *Eriomyces*, as well as *Funbolia*, *Heleiosa*, *Phellinocrescentia* and *Pseudopassalora* which have been placed in Dothideomycetes genera *incertae sedis* (Crous et al., 2011b, 2014; Hyde et al., 2020a). However, a refined updated document on orders and families *incertae sedis* of Dothideomycetes was provided by Hongsanan et al. (2020b), and Eriomycetaceae was synonymized under Monoblastiaceae. In the present phylogenetic study (Figure 2.63), all of the genera (with available sequence data) that included within Monoblastiaceae in previous studies (Crous et al., 2020b, 2021b; Hongsanan et al., 2020b; Hyde et al., 2020a; Mortimer et al., 2021) are separated into two clades. Of which one is comprised of 8 known non-lichenzied genera (*Eriomyces*, *Funbolia*, *Haudseptoria*, *Heleiosa*, *Italiofungus*, *Neoheleiosa*, *Phellinocrescentia* and *Pseudopassalora*) and the new genus *Karstiomyces* introduced in this study, while other lichenzied genera located in another clade. And this topology is somewhat similar to Hyde et al. (2020a) and Hongsanan et al. (2020b). Therefore, we resurrect the family Eriomycetaceae to accommodate the above 8 non-lichenzied genera and assign it within Monoblastiales,

and reduce the accounts of Monoblastiaceae to 6 lichenzied genera, which are *Acrocordia*, *Anisomeridium*, *Caprettia*, *Megalotremis*, *Monoblastia* and *Trypetheliopsis*.

Karstiomyces J.F. Zhang & K.D. Hyde, *gen. nov.*

Etymology: Named refers to karst, the habitat of this genus.

Type species: *Karstiomyces guihzouensis*

Saprobic on dead culm of herbaceous plant. Sexual morph: *Ascomata* immersed, subglobose to basal laterally flatten, dark brown to black, solitary to gregarious, epapillate. *Peridium* composed of several layers of hyaline to brown, thick-walled pseudoparenchymatous cells, arranged in *textura globulosa*. *Hamathecium* comprising branched, non-trabeculate, filamentous pseudoparaphyses, anastomosing between and above the asci. *Asci* 8-spored, bitunicate, fissitunicate, cylindrical, furcate pedicellate, apically rounded, with an ocular chamber. *Ascospores* overlapping to bi-seriate, oval to broadly fusiform, pale brown, septate, and constricted at the septa, verrucose. Asexual morph: Undetermined.

Notes: *Karstiomyces* is sister to *Pseudopassalora* with full bootstrap support in the present study (Figure 2.57). Out of 8 reported genera in Eriomycetaceae, 3 genera (*Eriomyces*, *Heleiosa*, *Neoheleiosa*) are described with sexual morph. However, *Karstiomyces* is morphologically differs from these genera by its multi-septate, brown ascospores and the unique texture shape of peridium (Kohlmeyer et al., 1996; Hyde et al., 2020a; Mortimer et al., 2021). Moreover, they are also phylogenetically distant in the molecular studies. Based on a megablast search of NCBI's GenBank nucleotide database, the closest hits using the LSU sequence had highest similarity to *Haudseptoria typhae* (strain CBS 146790, GenBank NG_076711; Identities = 782/805 (97 %), no gaps), *Heleiosa barbatula* (strain JK 5548I, GenBank GU479787; Identities = 819/852 (96 %), 3 gaps (0%)), *Italiofungus phillyreae* (strain CPC 35566, GenBank MT223804; Identities = 807/843 (96 %), 2 gaps (0%)) and *Neoheleiosa lincangensis* (strain HKAS 111914, GenBank NG_075423; Identities =

815/849 (96 %), 1 gaps (0%)). Closest hits using the SSU sequence had highest similarity to *Heleiosa barbatula* (strain JK 5548I, GenBank GU479787; Identities = 872/904 (96 %), 2 gaps (0%)) and *Neoheleiosa lincangensis* (strain HKAS 111914, GenBank NG_075423; Identities = 860/893 (96 %), 3 gaps (0%)). Closest hits using the ITS sequence had highest similarity to *Pseudopassalora gouriqua* (strain CBS 101954, GenBank NR_160207; Identities = 470/507 (93 %), 5 gaps (0%)) and *Italiofungus phillyreae* (strain CPC 35566, GenBank MT223804; Identities = 407/447 (90 %), 10 gaps (2%)). While no significant hits were obtained when the TEF1 and Rpb2 sequences was used in blastn and megablast searches. Based on the present study and megablast search results, *Karstiomyces* is introduced here as a new genus to accommodate the type species *Karstiomyces guizhouenbsis*.

Karstiomyces guihzouensis J.F. Zhang & K.D. Hyde, *sp. nov.*, Figure 2.55

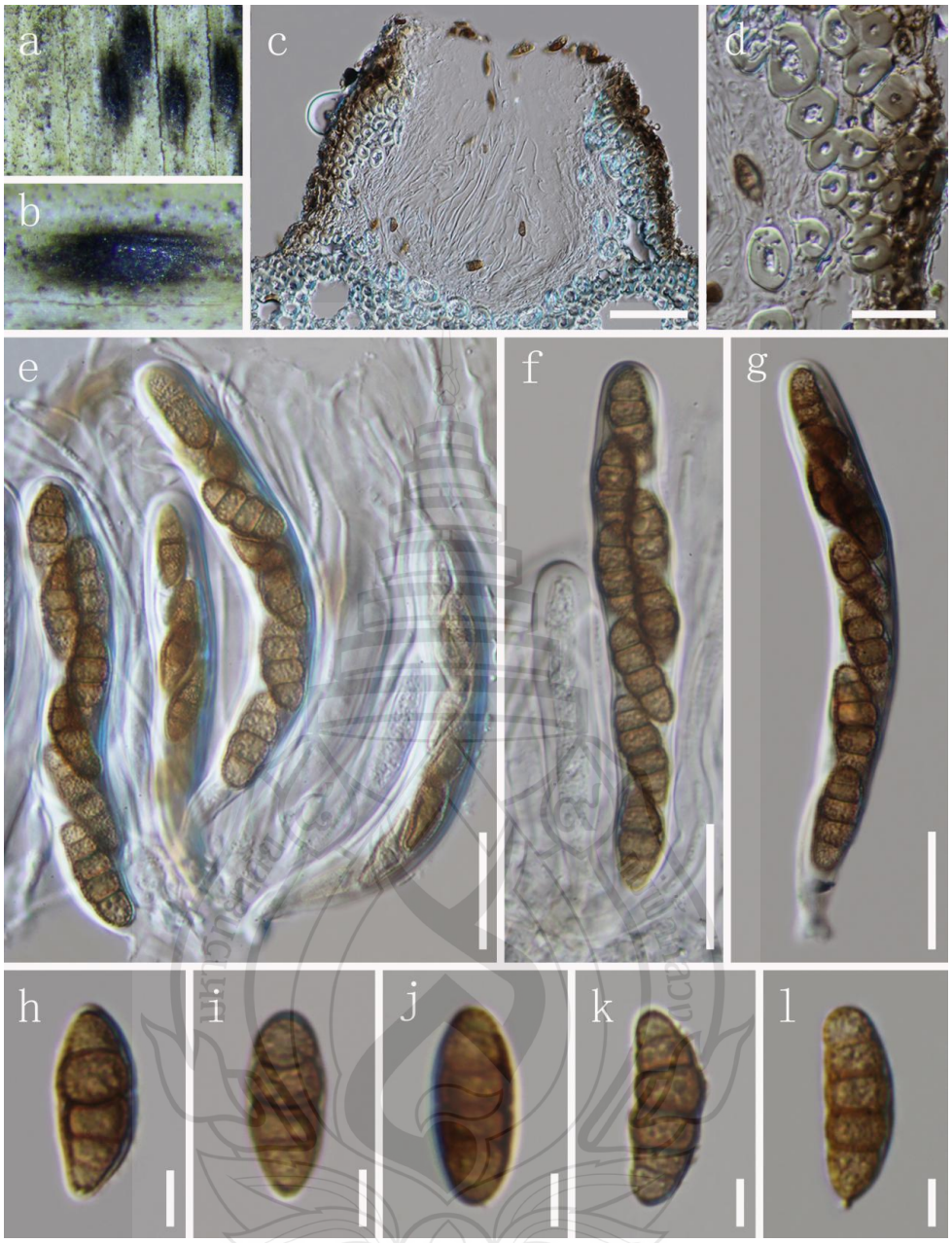
Etymology: Named refers to Guizhou Province where the holotype was collected.

Holotype: GZAAS 19-1754.

Saprobic on dead culm of herbaceous plant, appearing with significant, black spots in substrate. Sexual morph: *Ascomata* 180–190 µm high × 239–323 µm wide, semi-immersed to superficial, under epidermis, dark brown to black, scattered, solitary to gregarious, subglobose to hemispherical, epapillate. *Peridium* thin on the base, 15–20 µm wide, with hyaline, thin-walled cells of *textura angularius*, and becoming thick laterally, 48–59 µm wide, with the outermost stratum of several layers of dark brown cells of *textura angularis*, and confuse with numerous, hyaline, thick-walled, pseudoparenchymatous cells in central stratum, and become hyaline and thin towards inwardly with several layer cells of *textura angularis*. *Hamathecium* comprising 1.5–2 µm wide, branched, non-trabeculate, filamentous pseudoparaphyses, anastomosing between and above the asci, embedded in a gelatinous matrix. *Asci* (78–) 80–90 (–95) × 10–13.5 µm ($\bar{x}$ = 87.5 × 11.5 µm, n = 20), 8-spored, bitunicate, fissitunicate, cylindrical, with a furcate pedicel, apically rounded, with a developed

ocular chamber. *Ascospores* 16.5–19 (–21) × 6–8 µm ($\bar{x}$ = 18.5 × 7 µm, n = 35), overlapping to bi-seriate, oval to broadly fusiform, 3–5 septate, and constricted at the septa, straight or slightly curved, verrucose, without mucilaginous appendages. Asexual morph: Undetermined.

Material examined: CHINA, Guizhou Province, Sinan County, on dead culm of unidentified herbaceous plant, 22 May 2019, H.Y. Ran SN-2019-15 (GZAAS 19-1754, holotype).



Note a, b Appearance of ascomata in host surface. c Vertical section through ascomata. d Section of peridium. e Pseudoparaphyses intermixed with asci. f, g Asci. h–l Ascospores. Scale bars: c = 50 µm, d–g = 20 µm, h–l = 5 µm.

Figure 2.55 *Karstiomyces guihzouensis* (GZAAS 19-1754, holotype)

Neoheleiosa Mortimer, Frontiers in Microbiology: 6 (2021)

Neoheleiosa was introduced and assigned within *Monoblastiaceae* based on morphological and molecular analysis (Mortimer et al., 2021). It is phylogenetically close to *Heleiosa*, however, differing from the latter by the ornamentation of ascospores. *Heleiosa* was characterized by ellipsoidal to fusiform, two-celled, brown ascospores with numerous cilia-like subapical appendages at each end. Nevertheless, *Neoheleiosa* was observed by having narrowly ovoid to clavate, one-septate, dark brown, guttulate ascospores with conically rounded ends and faintly longitudinally striation, as well lacking any appendages (Kohlmeyer et al., 1996; Mortimer et al., 2021).

Neoheleiosa guihzouensis J.F. Zhang & K.D. Hyde, *sp. nov*., Figure 2.56

Etymology: Named refers to Guizhou where the holotype was collected.

Holotype: MFLU 18-2268.

Saprobic on dead culm of unidentified herbaceous plant, visible as black dots in face view. Sexual morph: *Ascomata* immersed to erumpent, dark brown to black, scattered, solitary, irregular subglobose, ostiolate, with a minute papilla. *Peridium* 7–15 µm wide, coriaceous, composed of several layers of thin to thick-walled cells of *texutura angularis*, hyaline and pale brown towards inwardly, and becoming dark towards the apex. *Hamathecium* comprising of sparse, 1–2 µm wide, non-trabeculate, branched pseudoparaphyses, intermixed with the asci, and embedded in a gelatinous matrix. *Asci* (72–) 79–85 (–88) × 8–9.5 µm ($\bar{x}$ = 81.5 × 8.9 µm, n = 20), 8-spored, bitunicate, fissitunicate, cylindrical, with a short, furcate pedicel, apically rounded, with a distinct ocular chamber. *Ascospores* 12–15 × 5.5–7.5 µm ($\bar{x}$ = 13.2 × 6 µm, n = 30), overlapping uniseriate, narrowly ovoid to clavate, verrucose, one-septete and slightly constricted at the septum, wall smooth, without any appendages. Asexual morph: Undetermined.

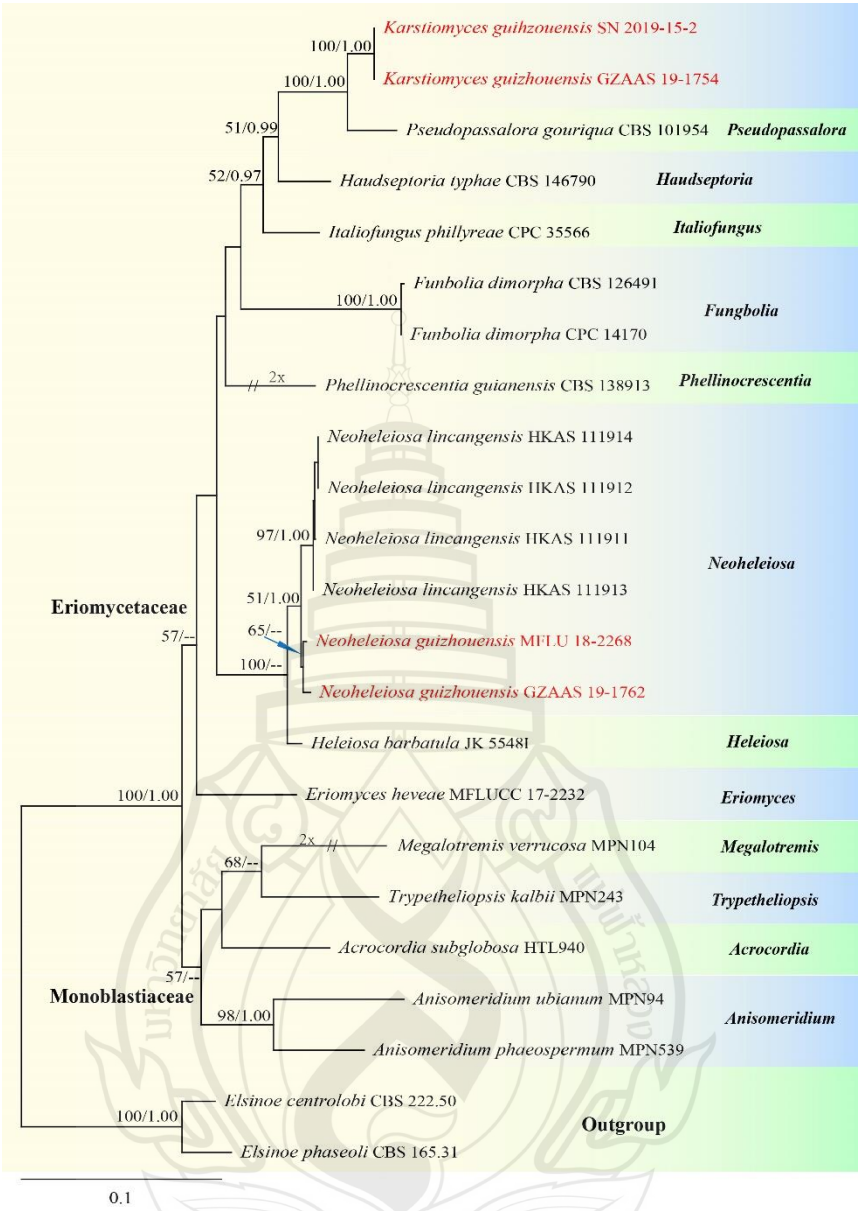
Material examined: CHINA, Guizhou Province, Guiyang City, Huaxi District, on dead culm of unidentified herbaceous plant, 3 April 2016, J.F. Zhang SD-2016-2 (MFLU 18-2268, holotype).

Notes: *Neoheleiosa guihzouensis* is phylogenetically close to *N. lincangensis* in the present study (Figure 2.57). And it is also typical of *Neoheleiosa* in having immersed to erupment, subglobose ascomata, cylindrical asci with producing narrowly ovoid to clavete, two-celled, brown ascospores. Nevertheless, *Neoheleiosa guihzouensis* is obviously differs from *N. lincangensis* by its smaller asci ((72–) 79–85 (–88) × 8–9.5 µm vs. 115–135 × 10–12 µm) and ascospores (12–15 × 5.5–7.5 µm vs. 16.5–17.5 × 7–9 µm) (Mortimer et al., 2021).



Note a, b Appearance of ascomata in the host surface. c Vertical section through ascomata. d Section of peridium. e Pseudoparaphyses. f–i Asci. j–o Ascospores. Scale bars: c = 100 µm, d, e = 10 µm, f–i = 20 µm, j–o = 5 µm.

Figure 2.56 *Neoheleiosa guihzouensis* (MFLU 18-2268, holotype)



Note Maximum likelihood bootstrap values equal or greater than 50 and Bayesian posterior probabilities equal or greater than 0.95 are presented near the nodes. Newly generated sequences in this study are given in red. The tree is generated from maximum likelihood (ML) analysis based on LSU, SSU, ITS and TEF1-α sequence data, and rooted with *Elsinoe centrolobi* CBS 222.50 and *E. phaseoli* CBS 165.31.

Figure 2.57 Phylogenetic tree of the order Monoblastiales

Tubeufiales Boonmee & K.D. Hyde

Tubeufiales was originally mentioned in Hyde et al. (2013) and formally introduced by Boonmee et al. (2014) to accommodate the single family Tubeufiaceae. In the same year, Suetrong et al. (2014) established and placed a second family Wiesneriomycetaceae within Tubeufiales on the basis of morphology and phylogeny. However, Bezerra et al. (2017) excluded Wiesneriomycetaceae from this order and introduced two new orders Bezerromycetales and Wiesneriomycetales to accommodate Bezerromycetaceae and Wiesneriomycetaceae, which are phylogenetically close to Tubeufiales. Soon after, Liu et al. (2017) processed a study of ranking higher taxa in Dothideomycetes by using divergence times, and included these above three families in Tubeufiales as well as treated Bezerromycetales and Wiesneriomycetales as synonyms of Tubeufiales. And this treatment was followed in recent studies (Lu et al., 2018b; Hongsanan et al., 2020b; Wijayawardene et al., 2020).

Tubeufiaceae M.E. Barr

Tubeufiaceae was established by Barr (1979a) and is typified with *Tubeufia*. Since then, the various classification treatments for this family were proposed in later studies (Barr, 1980; Rossman, 1987; Kirk, et al. 2001; Lumbsch & Huhndorf, 2010; Boonmee et al., 2011, 2014; Rajeshkumar & Sharma, 2013; Brahamanage et al., 2017; Chaiwan et al., 2017; Doilom et al., 2017; Luo et al., 2017; Liu et al., 2018; Lu et al., 2018a; Wijayawardene et al., 2018). Subsequently, a reassessment of Tubeufiales was carried out by Lu et al. (2018b) based on multi-locus phylogeny and morphology, and 42 genera were accepted within Tubeufiaceae in their study. Presently, Liu et al. (2019) and Hyde et al. (2020a) added two genera *Kevinhydea* and *Camporesiomyces* in this family respectively, and herein we follow this latest treatment.

Neodictyospora J.F. Zhang & K.D. Hyde, *gen. nov.*

Etymology: Named refers to the fruiting body of *Neodictyospora* resembles to the sexual morph of *Dictyospora* in nature.

Type species: *Neodictyospora karstia*

Saprobic on dead culm of unidentified herbaceous plant. Sexual morph: *Ascomata* superficial, solitary or scattered, globose to subglobose, black, setaceous, coriaceous, with a central ostiole. *Setae* covering the whole ascoma, dark brown to black, tapering towards the apex. *Peridium* composed of several layers of cells of *textura angularis*. *Hamathecium* comprising branched pseudoparaphyses, anastomosing between and above the asci. *Asci* 8-spored, bitunicate, fissitunicate, cylindrical to clavate, short pedicellate. *Ascospores* fasciculate, filiform to narrowly fusiform, hyaline, tapered toward the ends, slightly wider at the central, guttulate, 9-septate, not constricted at the septa. Asexual morph: Undetermined.

Notes: *Neodictyospora* is placed within Tubeufiaceae with an independent clade that sister to *Camporesiomyces* in the present study (Fig. 2.59). Both of these two genera are characterized by setaceous, globose to subglobose ascomata, and clavate asci. However, they can be differentiated from one another by the genus *Camporesiomyces* are described in having multi-loculate ascomata that produce cylindrical ascospores. Nevertheless, *Neodictyospora* is clearly presented by having uni-loculate ascomata with narrowly fusiform ascosproes (Hyde et al., 2020a). *Neodictyospora* is morphologically close to the sexula morph of *Dictyospora* by uni-loculate, globose to subglobose ascomata that surrounded by short, dark brown to black setae, whereas, they can be distinguished by the shape of ascospores (Brahmanage et al., 2017; Lu et al., 2018b) and phylogenetic distance in this study. Based on a megablast search of NCBI's GenBank nucleotide database, the closest hits using the LSU sequence had highest similarity to *Helicosporium luteosporum* (strain MFLUCC 16-0266, GenBank KY321327; Identities = 830/843 (98%), 1 gaps (0%)), *Tubeufia cerea* (strain IF)0914, GenBank AY849964; Identities = 831/849 (98%), 1 gaps (0%)) and *Camporesiomyces mali* (strain KUMCC 19-0126, GenBank MN792811; Identities = 828/849 (98 %), 1 gaps (0%)). Closest hits using the ITS sequence had highest similarity to *Camporesiomyces vaccinii* (strain CBS 216.90,

GenBank NR_156202; Identities = 288/319 (90%), 11 gaps (3%)), *Helicosporium luteosporum* (strain MFLUCC 16-0266, GenBank KY321324; Identities = 283/321 (88 %), 17 gaps (5%)) and *Zaanenomyces quadripartis* (strain CBS 148310, GenBank OK664721; Identities = 279/320 (87 %), 11 gaps (3%)). Closest hits using the *TEF1* sequence had highest similarity to *Camporesiomyces mali* (strain KUMCC 19-0126, GenBank MN794018; Identities = 822/890 (92%), 2 gaps (0%)), *Tubeufia hyalospora* (strain MFLUCC 15-1250, GenBank MH550979; Identities = 820/890 (92%), 6 gaps (0%)) and *Neohelicosporium aquaticum* (strain MFLUCC 17-1519, GenBank MF535242; Identities = 817/889 (92%), 4 gaps (0%)). Closest hits using the Rpb2 sequence had highest similarity to *Dematiohelicosporum guttulatum* (strain MFLUCC 17-2011, GenBank MH551021; Identities = 800/1012 (79%), 6 gaps (0%)). Based on the above evidence, *Neodictyospora* is introduced herein as a new genus to accommodate the type species *Neodictyospora karstia.*

Neodictyospora karstia J.F. Zhang & K.D. Hyde, *sp. nov*., Figure 2.58

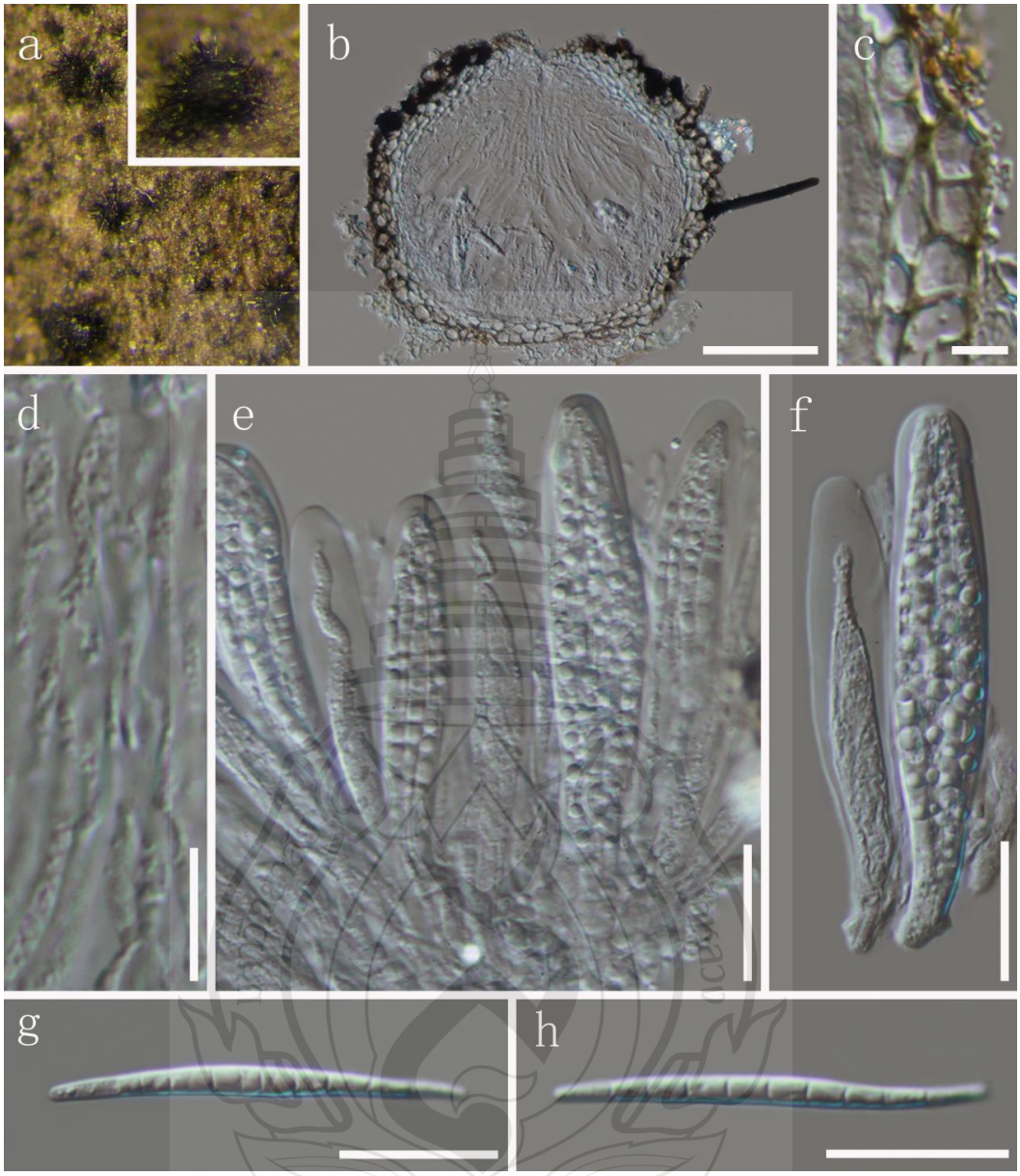
Etymology: Named refers to the holotype was collected from karst.

Holotype: GZAAS 19-1751.

Saprobic on dead culm of unidentified herbaceous plant. Sexual morph: *Ascomata* semi-immersed to superficial, solitary or scattered, globose to subglobose, black, setaceous, coriaceous, with a central ostiole. *Setae* 48–57 × 4–5 µm, covering the whole ascoma, dark brown to black, tapering towards the apex. *Peridium* 12–23 µm thick, composed of several layers of cells of *textura angularis*, hyaline to pale brown in sides, and become thick and dark brown to black around the ostiole. *Hamathecium* comprising of 1–1.5 µm wide, branched pseudoparaphyses, anastomosing between and above the asci, embedded in a gelatinous matrix. *Asci* (60.5–) 69.5–78.5 × 12–15 µm ($\bar{x}$ = 72.5 × 13.5 µm, n = 20), 8-spored, bitunicate, fifissitunicate, cylindric-clavate, apically rounded to truncate, short pedicellate. *Ascospores* 48–52 × 3.5–4 µm ($\bar{x}$ = 50 × 3.5 µm, n = 30), fasciculate, fusiform,

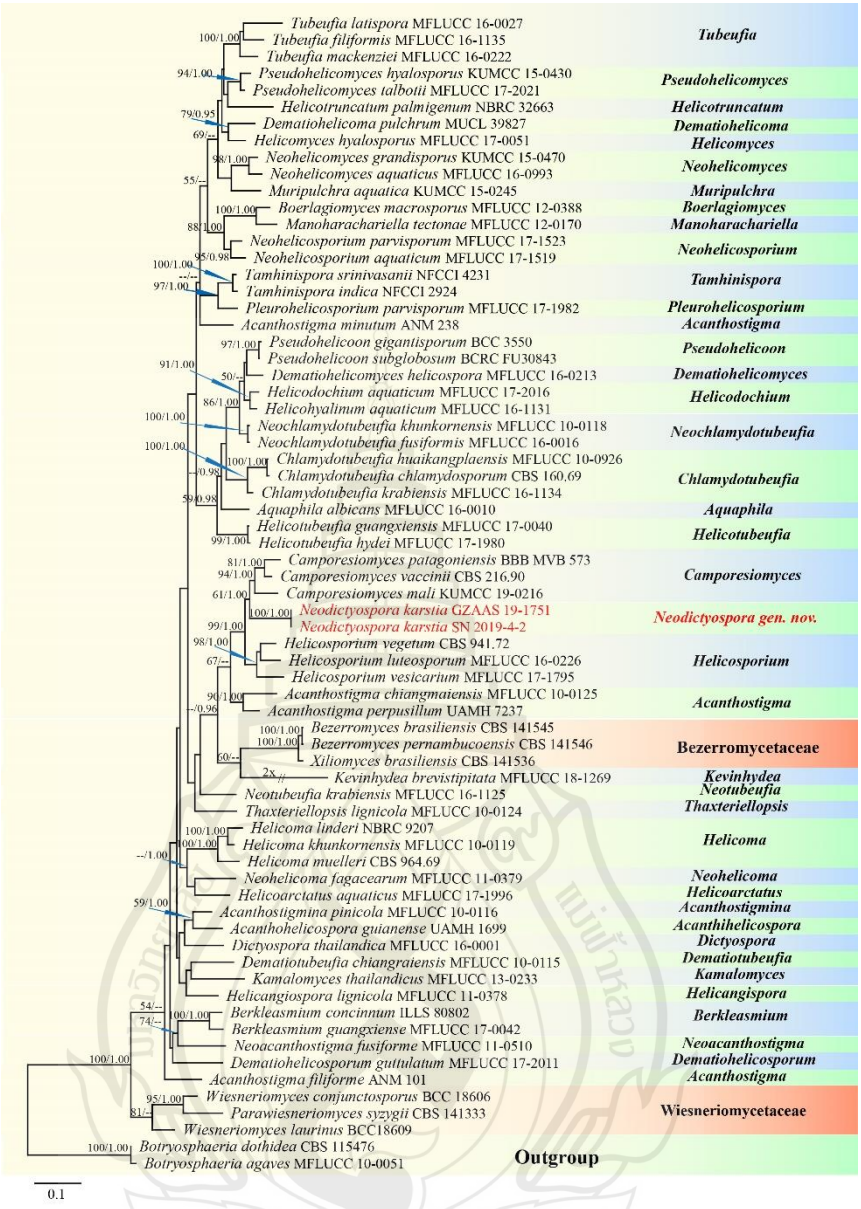
hyaline, straight or slightly curved, tapering toward the ends, guttulate, normally 9-septate, not constricted at the septa. Asexual morph: Undetermined.

Material examined: CHINA, Guizhou Province, Sinan County, on dead culm of unidentified herbaceous plant, 22 May 2019, H.Y. Ran SN-2019-4 (GZAAS 19-1751, holotype).



Note a Appearance of ascomata in host surface. b Vertical section through ascoma. c Section of peridium. d Pseudoparaphyses. e, f Asci. g, h Ascospores. Scale bars: b = 50 µm, c = 5 µm, d = 10 µm, e–h = 20 µm.

Figure 2.58 *Neodictyospora karstia* (GZAAS 19-1751, holotype)



Note Maximum likelihood bootstrap values equal or greater than 50 and Bayesian posterior probabilities equal or greater than 0.95 are presented near the nodes. Newly generated sequences in this study are given in red. The tree is generated from maximum likelihood (ML) analysis based on ITS, LSU, Rpb2 and TEF1-α sequence data, and rooted with *Botryosphaeria agaves* MFLUCC 10-0051 and *B. dothodea* CBS 115476.

Figure 2.59 Phylogenetic tree of the order Tubeufiales

CHAPTER 3

TAXONOMIC AND PHYLOGENETIC OF SORDARIOMYCETES FROM KARST LANDSCAPE OF GUIZHOU PROVINCE

3.1 Introduction

Sordariomycetes, as the second largest class of Ascomycota, was introduced by Eriksson and Winka (1997) and comprised of highly diverse range of fungi which are characterized mainly by perithecial ascomata and inoperculate or unitunicate asci or fissitunicate asci (Kirk et al., 2008). In addition, the species in this class have a cosmopolitan distribution, and usually isolated from the host as pathogens, endophytes, saprobes, epiphytes, coprophilous and fungicolous, lichenized or lichenicolous taxa (Maharachchikumbura et al., 2016). Maharachchikumbura et al. (2016) proposed a classification scheme of Sordariomycetes of using multi-gene phylogenetic analysis, and accepted 6 subclasses, 32 orders, 105 families and 1331 genera within this class. Subsequently, Hongsanan et al. (2017) provided an update of phylogeny of Sordariomycetes based on phylogenetic and molecular clock evidence. In their study, the subclass Meliolomycetidae that accepted in previous studies was transferred to Sordariomycetidae (Maharachchikumbura et al., 2016; Hyde et al., 2017) and a novel subclass Savoryellomycetidae was introduced. They recommended that the the divergence for Sordariomycetes should have evolved at 341 MYA (crown age), and the six subclasses should have evolved between 250–289 MYA (stem age). While the divergence of orders and families within Sordariomycetes were estimated between 250–289 MYA (stem age) and 50–130 MYA (stem age), respectively (Hyde et al.,

2017). In the latest outline of fungi, 46 orders, 184 families and 1692 genera were listed under Sordariomycetes.

Several novel sordariomycetous fungal species have been reported from karst formations in resent studies (Chen et al., 2017, 2021, Zhang et al., 2017c, 2020c; Dissanayake et al., 2020; Feng et al., 2021; Long et al., 2021; Wijayawardene et al., 2021). However, a hypothesis is proposed that lots of members of Sordariomycetes in this unique eco-habitat are waiting to be found as Sordariomycetes is a largest and most diverse class of ascomycota (Kirk et al., 2008; Hyde et al., 2013, Wijayawardene et al., 2018, 2021, 2022).

3.2 Materials and Methods

3.2.1 Collection, Examination, Isolation and Specimen Deposition

See 1.5.1–1.5.4.

3.2.2 DNA Extraction, PCR Amplification and Sequencing

See 1.5.5.

3.2.3 Sequence Alignment and Phylogenetic Analysis

3.3 Results

3.3.1 Phylogenetic Analyses

Because of high diversity of the newly collected taxa, the individual phylogenetic trees at the order, family or genus levels were carried out for clarifying the natural placement of our new collections in this study. All newly generated/sequenced sequence data are deposited in GenBank. Illustrations and descriptions are provided in below.

3.3.2 Taxonomy

Class Sordariomycetes O.E. Erikss. & Winka

Subclass Diaporthomycetidae Senan. et al.

Atractosporales H. Zhang et al.

Atractosporales was established by Zhang et al. (2017) to accommodate three new families viz. Atractosporaceae, Conlariaceae and Pseudoproboscisporaceae, which were placed in Sordariomycetes genera *incertae sedis* and Annulatascaceae in previous studies (Maharachchikumbura et al., 2016). However, the monophyly of this order was argued by Luo et al. (2019) and Hyde et al. (2020) based on their phylogenetic analysis. Even the status of Atractosporales members was suggest to revise in further study (Hyde et al., 2020b), these above three family were still placed in Atractosporales (Hyde et al., 2020b; Wijayawardene et al., 2022).

Atractosporaceae H. Zhang et al.

Atractosporaceae was introduced by Zhang et al. (2017) to accommodate the genera *Atractospora* and *Rubellisphaeria* based on morphological and phylogenetic analysis. Both of these two genera share freshwater habitats and are morphologically similar in having immersed to semi-immersed, usually light brown to dark brown ascomata, with a relatively thin-walled peridium of *textura angularis*, long cylindrical asci and hyaline, uni-seriate, fusiform ascospores. And this classification treatment was also followed by Hyde et al. (2020b) and Wijayawardene et al. (2022).

Atractospora Réblová & J. Fourn., Mycol. Progr. 15(no. 21): 8 (2016)

Atractospora was introduced by Réblová et al. (2016) as an aquatic genus to accommodate three new species *A. decumbens*, *A. reticulata*, *A. verruculosa* and a new combination *A. ellipsoidea* with *Atractospora reticulata* as the type species. Subsequently, other two species *A. thailandensis* and *A. aquatica* were respectively assigned within this genus (Zhang et al., 2017; Luo et al., 2019), and totally six species were accepted in *Atractospora* until now.

Atractospora terrestrica J.F. Zhang, Y.Y. Chen & Z.Y. Liu *sp. nov*., Figure 3.1

Etymology: Named '*terrestrica*' refers to the habitat of the holotype was collected.

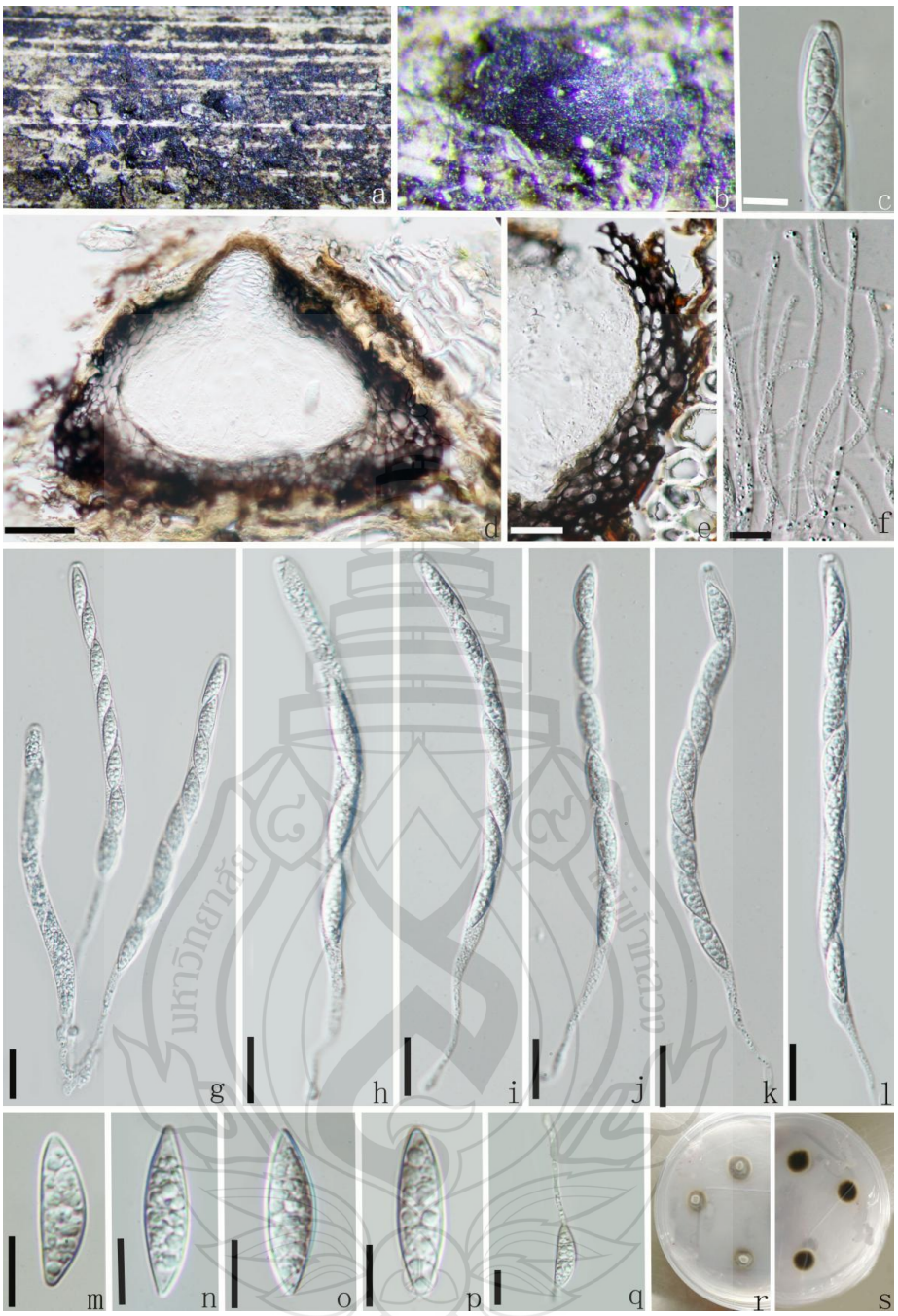
Holotype: GZAAS 20-0223.

Saprobic on dead culm of herbaceous plant. Sexual morph: *Ascomata* 313−410 μm high, 323−440 μm diam., semi-immersed to superficial, solitary, scattered, venter subglobose to conical, dark brown to black, lying horizontally on the substrate, ostiolate. *Ostiole* central, paraphysate, with a pore-like opening. *Peridium*

41−67 μm wide, corioceous, composed of several layers of cells of *textura prismatica*, thick-walled and dark pigmented outwardlly, becoming subhyaline and thin-walled towards the interior. *Hamathecium* comprising numerous, 2−2.5 μm wide, aseptate, cellular paraphyses, anastomosing between and above the asci. *Asci* 174−218 × 12−16 μm ($\bar{x}$ = 192.5 × 14.5 μm, n = 30), 8-spored, cylindrical with a slender, long pedicellate, apex broadly rounded with a conspicuous, non-amyloid apical annulus. *Ascospores* 21−26 × 7−10 μm ($\bar{x}$ = 23.5 × 8.5 μm, n = 30), fusiform, one-celled, uni- to overlapping biseriate, slightly inequilateral, hyaline, verrucose. Asexual morph: Undetermined.

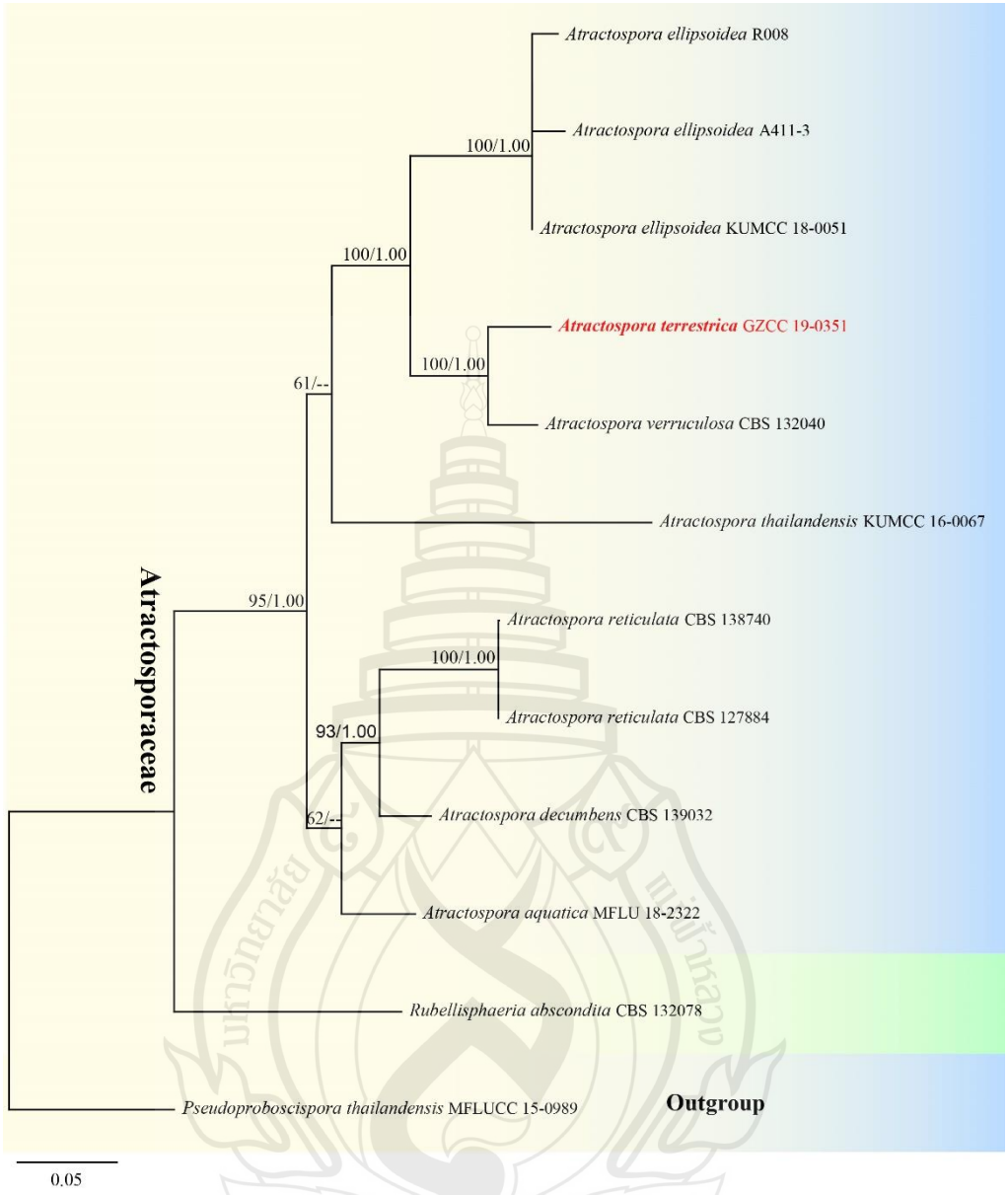
Material examined: CHINA, Guizhou Province, Tongren City, on dead culm of unidentified herbaceous plant, 16 April 2018, Y.Y. Chen F19-5 (GZAAS 20-0223, holotype); ex-type living culture: GZCC 19-0351.

Notes: *Atractospora terrestrica* is phylogenetically sister to *A. verruculosa* with full bootstrap support in the present study (Figure 3.2). Morphologically, *A. terrestrica* is typical of *Atractospora* in having subglobose to conical, perithecial ascomata, unitunicate asci bearing a non-amyloid apical annual, as well as fusiform, hyaline, ascospores. However, it differs from closet relative taxon *A. verruculosa* by its larger asci (174−218 × 12−16 μm vs. (155−)165−190 × 9.5−12 μm) and non-septate ascospores (Réblová et al., 2016). *Atractospora terrestrica* resembles to *A. reticulata* by having overlap sized asci and ascospores, nevertheless, these two taxa can be easily distinguished by the ascospores of *A. reticulata* are well presented with a conspicuous reticulate ornamentation, while *A. terrestrica* is characterized by verrucose ascospores (Réblová et al., 2016). Hence, the new specimen collected in this study is introduced as a new species based on the above morpho-phylogenetic evidence. Additionally, *Atractospora* was established as an aquatic genus and all of the accounts (*A. aquatica*, *A. decumbens*, *A. ellipsoidea*, *A. reticulata*, *A. thailandensis* and *A. verruculosa*) reported in previous studies are collected from aquatic habitats (Ho et al., 1999; Réblová et al., 2016; Zhang et al., 2017; Luo et al., 2019), while the new species in this study was discovered in terrestrial, and therefore the habitat of *Atractospora* is extend to aquatic or terrestrial in the present study.



Note a, b ascomata on host surface. c Apex of ascus. d vertical section through ascoma. e Section of peridium. f Paraphyses. g–l Asci. m–p Ascospores. q Germinated ascospore. r, s Colony on PDA. Scale bars: c = 10 µm, d = 100 µm, e = 50 µm, f = 10 µm, g–l = 20 µm, m–q = 10 µm.

Figure 3.1 *Atractospora terrestrica* (GZAAS 20-0223, holotype)



Note Maximum likelihood bootstrap values equal or greater than 50% and Bayesian posterior probabilities equal or greater than 0.95 are indicated near the nodes. New sequence generated in this study is given in red. The tree is generated from maximum likelihood (ML) analysis based on LSU, ITS, Rpb2 and SSU sequence data, and rooted with *Pseudoproboscispora thailandensis* MFLUCC 15-0989.

Figure 3.2 Phylogenetic tree of the family Atractosporaceae

Diaporthales Nannf.

Diaporthales was introduced by Nannfeldt (1932) to accommodate Höhnel's Eu-Diaportheen and Valseen taxa. Then, a series of families were recognized and placed within this order based on morphological characteristics (Luttrell, 1951; Chadefaud, 1960; Wehmeyer, 1975; Barr, 1978; Eriksson, 2001). Castlebury et al. (2002) first employed the LSU sequence data, and included four family viz. Diaporthaceae, Gnomoniaceae, Melanconidaceae and Cytosporaceae within Diaporthales but excluded Vialaeaceae from it. Subsequently, additions and exclusions of families in Diaporthales were proposed in past decades (Réblová et al., 2004; Gryzenhout et al., 2006; Rossman et al., 2007; Cheewangkoon et al., 2010; Crous et al., 2012, 2015; Voglmayr & Jaklitsch, 2014; Jones et al., 2015; Maharachchikumbura et al., 2015; Suetrong et al., 2015; Alvarez et al., 2016; Norphanphoun et al., 2016; Senanayake et al., 2017a; Voglmayr et al., 2017; Yang et al., 2017; Braun et al., 2018; Fan et al., 2018). Senanayake et al. (2018) circumscribed Diaporthales and accepted 27 families based on multi-gene phylogeney and morphology evidence. Soon after, other four families (Dwiroopaceae, Foliocryphiaceae, Mastigosporellaceae and Phaeochorellaceae) were placed within this order in recent studies (Guterres et al., 2019; Xavier et al., 2019; Jiang et al., 2020).

Cytosporaceae Fr.

Cytosporaceae was introduced by Fries (1825) and placed into Diaporthales with *Cytospora* as the typus. Maharachchikumbura et al. (2015, 2016) monographed the families of Sordariomycetes, and assigned 13 genera within Cytosporaceae. Then, Senanayake et al. (2017a, 2018) provided an outline and circumsciption of Diaporthales based on morphological and phylogenetic evidence, and accpted 7 genera under Cytosporaceae. However, Hyde et al. (2020) refined families of Sordariomycetes, and transfered the genus *Hypophloeda* to Diaporthales genera *incertae sedis* as it is atypical of Cytosporaceae. Therefore, six genera *viz*. *Cryptascoma*, *Cytospora*, *Pachytrype*, *Paravalsa*, *Waydora* and *Xenotypa* were currently included in Cytosporaceae, and this treatment was followed by Wijayawardene et al. (2020) in the updated outline of Fungi and fungus-like taxa.

Cytospora Ehrenb., Sylv. mycol. berol. (Berlin): 28 (1818)

Cytospora was introduced by Ehrenberg (1818) with *Cytospora chrysosperma* as the type species. Members of this genus are distributed worldwide and reported as important pathogens that causing canker and dieback disease of a wide range of plants, and accounts of new taxa were discovered in recent studies (Adams et al., 2005, 2006, Fan et al., 2014; 2015a, 2015b; Wang et al., 2015; Lawrence et al., 2017; Norphanphoun et al., 2018; Jiang et al., 2020; Pan et al., 2020).

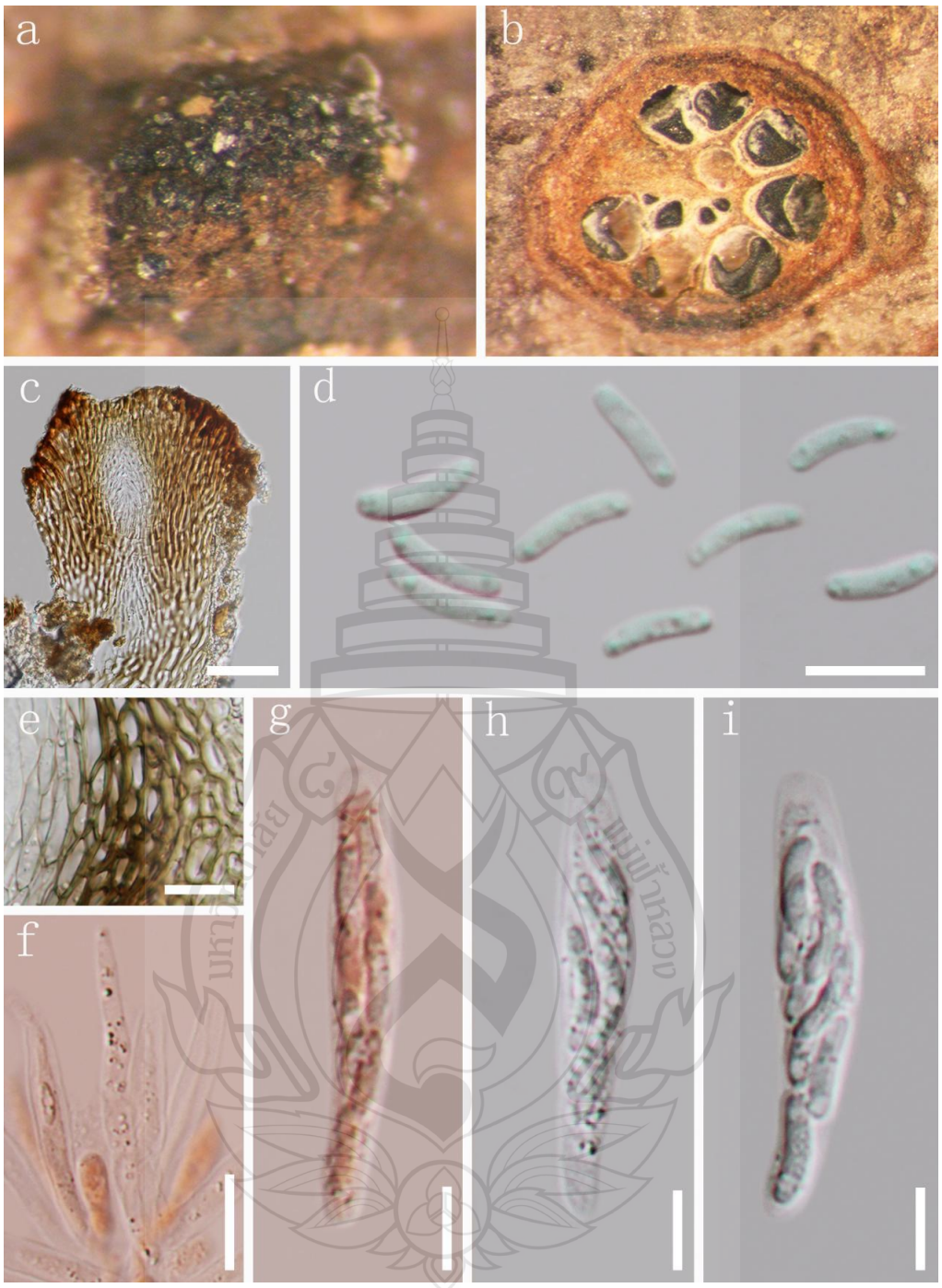
Cytospora predappioensis Q.J. Shang, Norph., Camporesi & K.D. Hyde, Mycosphere 9(2): 376 (2018). Figure 3.3

Saprobic on dead branch of unidentified woody plant. Sexual morph: *Stromata* solitary to gregarious, with the poorly developed interior, immersed to erumpent throughout the bark by the ostiolar canal, dark brown to black, glabrous, circular to irregular in shape, arranged with conspicuous, clustered, roundish to cylindrical prominent ostioles. *Ascomata* perithecial, immersed in a stroma, dark brown to brown, globose to subglobose, glabrous, individual ostiole with the neck. *Peridium* 15–20 μm wide, composed of two stratum, outer stratum comprising several layers of brown to dark brown, thick–walled cells of pseudoparenchymatous, the inner stratum comprising 3–5 layers of hyaline to pale brown cells of *textura angularis*. *Hamathecium* comprising of sparse, 2–4 μm wide, septate, cullular paraphyses, anastomosing between the asci. *Asci* 27–35 × 5–7 μm ($\bar{x}$ = 31 × 6 μm, n = 30), 8-spored, unitunicate, clavate, sessile, apically rounded to truncate, with a J- apical apparatus. *Ascospores* 6–8 × 1.5–2.5 μm ($\bar{x}$ = 7 × 2 μm, n = 30), overlapping 1–3-seriate, hyaline, oblong to allantoid, aseptate, smooth-walled. Asexual morph: undetermined.

Material examined: CHINA, Guizhou Province, Guiyang City, on dead branch of unidentified woody plant, 14 May 2017, J.F. Zhang XCH-04 (GZAAS 20-4009); Kaiyang County, on dead branch of unidentified woody plant, 26 July 2017, H.Y. Ran KY-09 (MFLU 18-2256).

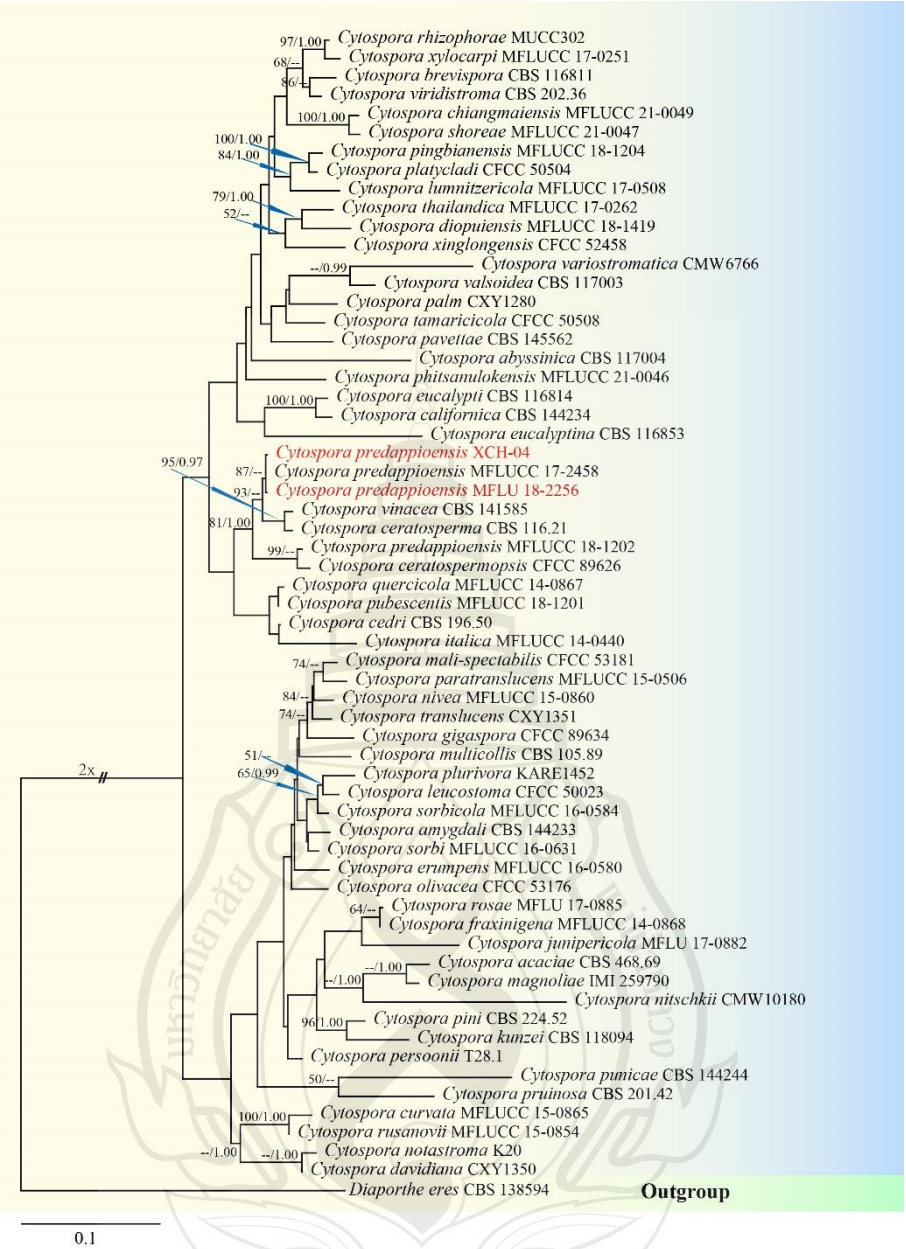
Notes: Present phylogenetic analysis shows that out new collections cluster to the ex-type strain of *Cytospora predappioensis* with moderate bootstrap support (Figure 3.4). Observation of the morphological characteristis of these newly specimen, we found that they are well fits with *C. predappioensis* including the measurements of asci and ascospores (Hyde et al., 2018). Therefore, these new collections are

identified as *C. predappioensis*, and which has been reported that distributed in China and Italy (Hyde et al., 2018; Shang et al., 2020).



Note a ascostromata on host surface. b Cross section through ascostroma. c Ostiole. d Ascospores. e Section of peridium. f Paraphyses. g–i Asci. Scale bars: c = 30 μm, d, e, g, i = 10 μm, e = 20 μm.

Figure 3.3 *Cytospora predappioensis* (GZAAS 20-4009)



Note Maximum likelihood bootstrap values equal or greater than 50% and Bayesian posterior probabilities equal or greater than 0.95 are indicated near the nodes. New sequence generated in this study are given in red. The tree is generated from maximum likelihood (ML) analysis based on ITS, LSU, ACT, Rpb2, TEF1-α, and Tub sequence data, and rooted with *Diaporthe eres* CBS 138594.

Figure 3.4 Phylogenetic tree of the genus *Cytospora*

Diaporthaceae Höhn. ex Wehm.

Diaporthaceae was introduced and accommodated in Diaporthales by Höhnel (1917), and it comprise numerous endophytic and phytopathogenic fungal species (Udayanga et al., 2011). The classification of Diaporthaceae has been argued and processed by various authors based on morphological and/or phylogenetic analysis (Wehmeyer, 1975; Castlebury et al., 2002; Sogonov et al., 2008; Lumbsch & Huhndorf, 2010; Lamprecht et al., 2011; Gomes et al., 2013; Dai et al., 2014; Voglmayr & Jaklitsch, 2014; Maharachchikumbura et al., 2015, 2016; Senanayake et al., 2017a, 2018), and the members of this family have increased to fifteen (*Apioporthella*, *Apiosphaeria*, *Chaetoconis*, *Chiangraiomyces*, *Diaporthe*, *Hyaliappendispora*, *Leucodiaporthe*, *Massariothea*, *Mazzantia*, *Ophiodiaporthe*, *Paradiaporthe*, *Phaeocytostroma*, *Phaeodiaporthe*, *Pustulomyces* and *Stenocarpella*) in the latest updated document of families of sordariomycetes and outline of fungi (Hyde et al., 2020b; Wijawawardene et al., 2020).

Diaporthe Nitschke

Diaporthe was introduced by Nitschke (1867) to accommodate the taxa with stromata which are often present with blackened zones in the substrate, ellipsoid to fusiform ascospores and enclosed, unilocular pycnidia that contain spermatia, stylospores and conidia (Wehmeyer, 1933). Members of this genus are not only known as endophytic, saprobic and plant pathogenetic fungi (Gomes et al., 2013), they are also morphologically diverse based on their ecology, host or substrate and geography (Senanayake et al., 2017b). In addition, *Diaporthe* was confirmed as a paraphyletic group based on phylogenetic evidence (Gao et al., 2017), and numerous of novel taxa in this genus were discovered in recent studies (Dissanayake et al., 2017, 2020; Gao et al., 2017; Yang et al., 2018, 2021; Guo et al., 2020; Li et al., 2020; Tennakoon et al., 2021) So far, more than 1100 epithets listed in Index Fungorum (Index Fungorum, 2022).

Diaporthe cylindriciospora J.F. Zhang & J.K. Liu *sp. nov*., Figure 3.5

Etymology: Named refers to its cylindrical ascospores.

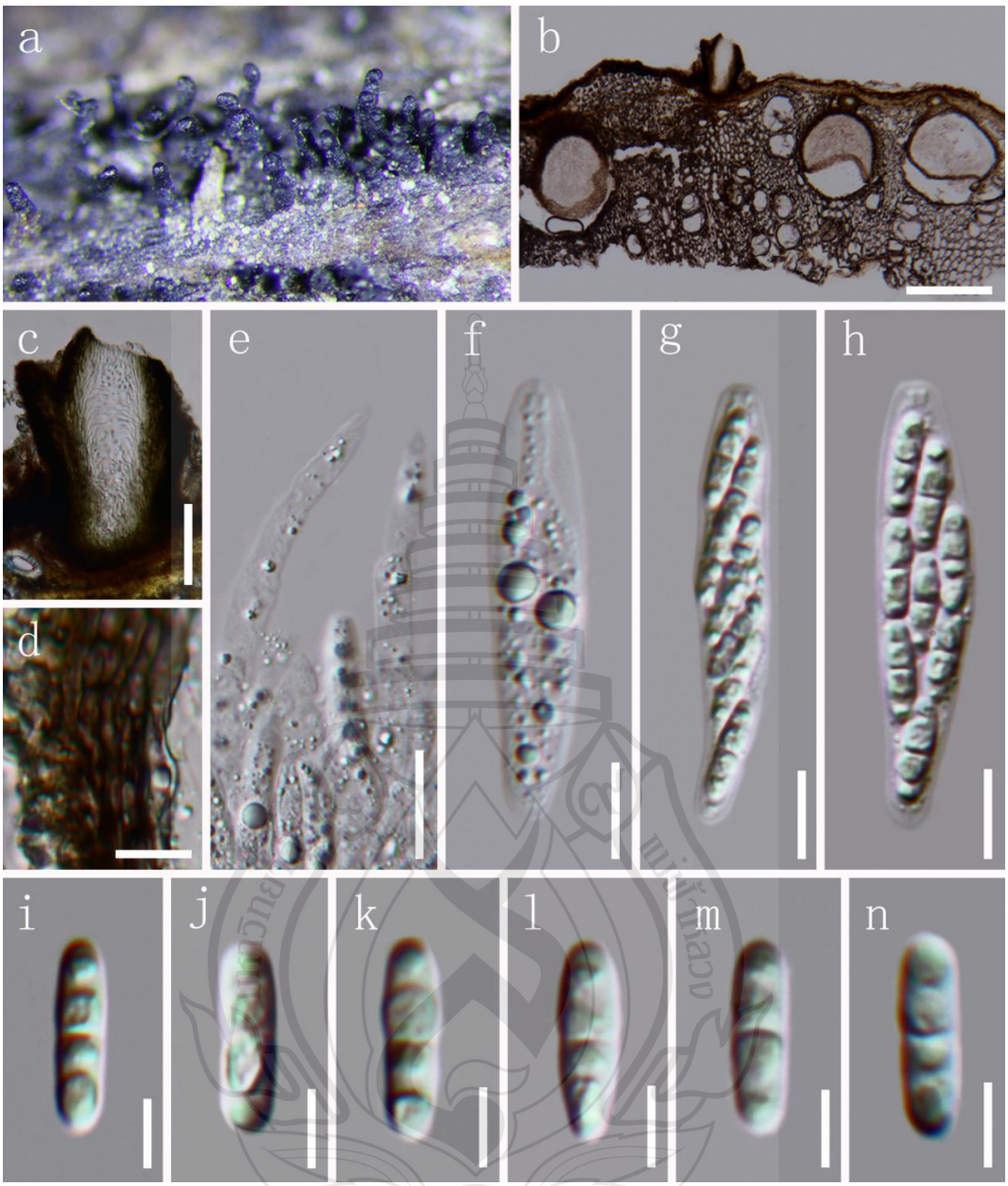
Hylotype: MFLU 18-2292.

Saprobic on dead culms of unidentified herbaceous plant. Sexual morph: *Ascomata* immersed, aggregated, globose to subglobose, dark brown, coriaceous,

ostiolate. *Ostiole* central or asymmetrically located, thick-walled, internally covered by numerous periphyses. *Peridium* 11–20 μm wide, composed of several layers of brown to dark brown, thick-walled cells of *textura angularis* outwardly, and become thin and hyaline towards inwardly. *Hamathecium* comprising sparse, trabeculate, cellular paraphyses, 2.5–7.5 μm wide, tapering towards the apex. *Asci* (41–)43.5–48.5(–50.5) × 8.5–11 μm ($\bar{x}$ = 46 × 9 μm, n = 25), 8-spored, unitunicate, cylindric-clavate, sessile, apically rounded to truncaate, with a visible refractive apical ring. *Ascospores* 11–13 × 2.5–4 μm ($\bar{x}$ = 12 × 3 μm, n = 30), randomly arranged, cylindrical, rounded at both ends, hyaline, one-celled when young and become one-septate at age, wall smooth, without any gelatinous sheath. Asexual morph: Undetermined.

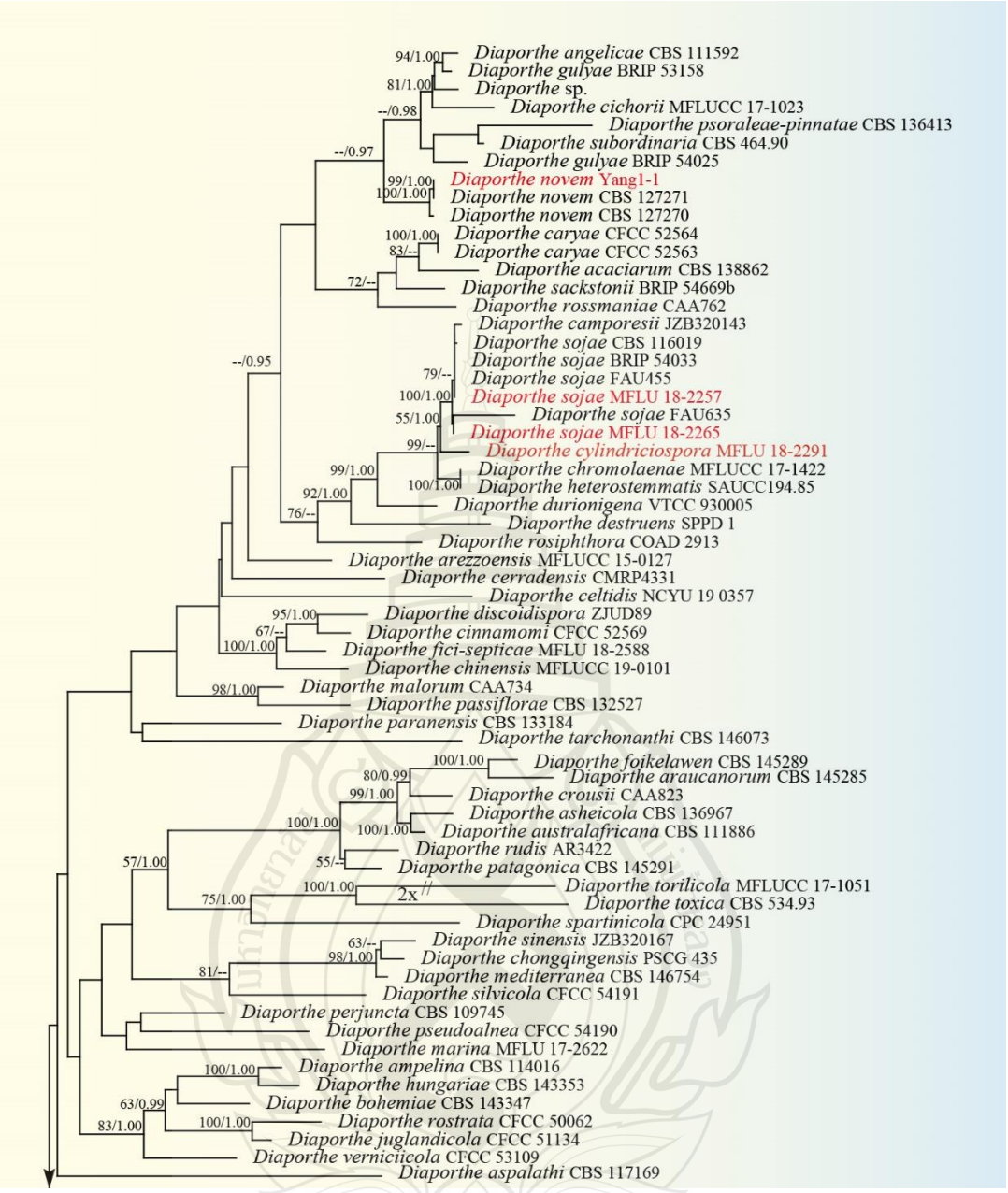
Material examined: CHINA, Guizhou Province, Guiyang City, Huaxi District, Guizhou Academy of Agricultural Sciences, on dead culm of herbaceous plant, 27 September 2016, J.F. Zhang GZ-09 (MFLU 18-2292).

Notes: *Diaporthe cylindriciospora* is closely related to *D. camporesii* and *D. sojae* in the present phylogenetic analysis (Figure 3.6). *Diaporthe cylindriciospora* resembles to *D. sojae* in having subglobose to globose ascomata with long ostiolar neck, unitunicate, cylindric-clavate asci with a visible refractive apical ring, nevertheless, they can be easily differentiated by the ascospores of *D. sojae* are fusoid, 1-septate and constricted at the septum, while *D. cylindriciospora* is presented with cylindrical ascospores that not constricted at the septum (Udayanga et al., 2015; Yang et al., 2021). Since *D. camporesii* was introduced as an asexual morph species (Hyde et al., 2020b), comparison of each gene region between it and our new collection is carried out based on the recommendations of Jeewon and Hyde (2016). And the results shows that there are 29.6% base pair differences for ITS across 506 nucleotides, and 13.4% base pair differences for Tub across 522 nucleotides, respectively. While the TEF1-α gene region (MN984254) of *D. camporesii* was not included in our phylogenetic analysis as the BLASTn search results in NCBI shows that it is closely related to *Alternaria* species, and we therefore suspect that it may be wrongly sequenced. Combination of these above analysis, *Diaporthe cylindriciospora* is introduced as a new species in this study.



Note Appearance of stroma in the host surface. b Vertical section through stroma. c Ostiole with paraphysis. d Section of peridium. e Paraphyses. f–h Young and mature asci. i–n Ascospores. Scale bars: b = 200 μm, c = 50 μm, d, f–h = 10 μm, e = 20 μm, i–n = 5 μm.

Figure 3.5 *Diaporthe cylindriciospora* (MFLU 18-2292, holotype)



Note Maximum likelihood bootstrap values equal or greater than 50% and Bayesian posterior probabilities equal or greater than 0.95 are indicated near the nodes. New sequence generated in this study are given in red. The tree is generated from maximum likelihood (ML) analysis based on ITS, TEF1-α, Tub, Cal and His sequence data, and rooted with *Diaporthella corylina* CBS 121124.

Figure 3.6 Phylogenetic tree of the genus *Diaporthe*



Figure 3.6 (continued)

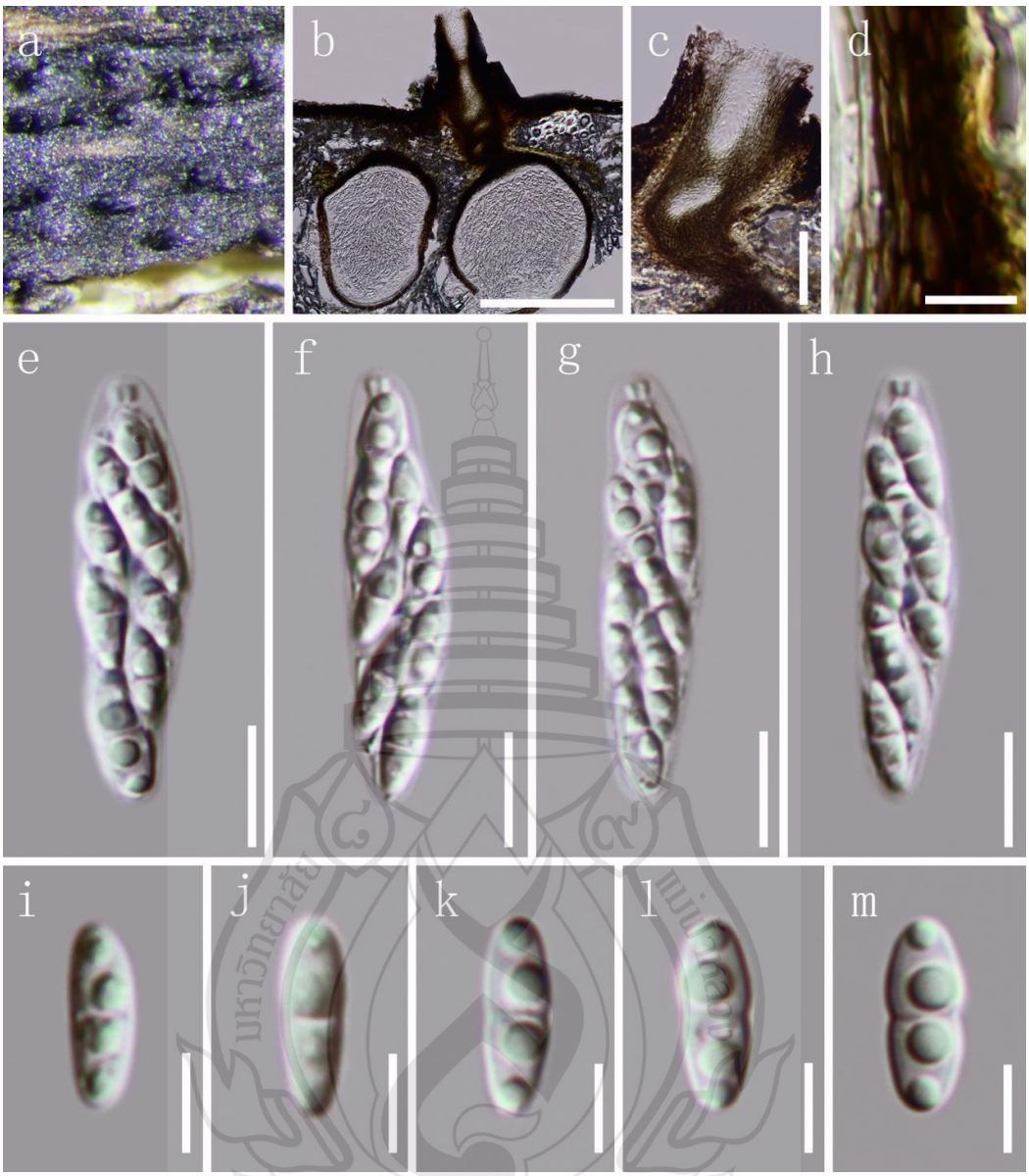
Diaporthe sojae Lehman, Ann. Mo. bot. Gdn 10: 128 (1923) Figure 3.7

Saprobic on dead culm of unidentified herbaceous plant. Sexual morph: *Ascomata* 234–300 µm high × 179–250 µm diam., perithecial, immersed, single to grouped, globose, coriaceous, black, ostiolate, papillate. *Neck* 162–230 µm high × 78–108 µm diam., periphysate. *Peridium* 11–28 µm wide, composed of two stratum, outer stratum of brown, thick-walled cells of *textura epidermoidea*, inner stratum of pale brown, thin-walled cellos of *textura angularis*. *Hamathecium* comprising numerous asci, paraphyses not observed. *Asci* 33–39 × 7.5–10.5 µm ($\bar{x}$ = 36 × 9 µm,

n = 35), unitunicate, 8-spored, cylindrical to clavate, sessile, apically flat and blunt, with a visible refractive apical ring. *Ascospores* 10–11 × 3–4.5 µm ($\bar{x}$ = 10.5 × 3.5 µm, n = 20), hyaline, randomly arranged, fusoid, 1-septate with a constriction, acute ends, smooth-walled, guttulate. Asexual morph: undetermined.

Material examined: CHINA, Guizhou Province, Danzhai County, on dead culm of herbaceous plant, 29 July 2017, J.F. Zhang DZ-05 (MFLU 18-2257); Huishui County, on dead culm of herbaceous plant, 21 July 2017, J.F. Zhang HS-12 (MFLU 18-2265).

Notes: In the present study, two newly collected specimens are closely related to *D. sojae* in the combined phylogenetic tree (Figure 3.6). Compared with the descriptions of *D. sojae* in Udayanga et al. (2015) and Yang et al. (2021), the gross morphological characteristics of our new specimens including the measurements of micro structures are well fits this species. Hence, we identify them as *D. sojae*. And this species has been reported as important plant pathogen on a wide range host (Udayanga et al., 2015; Guo et al., 2020; Yang et al., 2021).



Note a Appearance of ascomata in the host surface. b Vertical section through ascomata. c Ostiole with paraphysis. d Section of peridium. e–h Asci. i–m Ascospores. Scale bars: b = 200 μm, c = 50 μm, d–h = 10 μm, i–m = 5 μm.

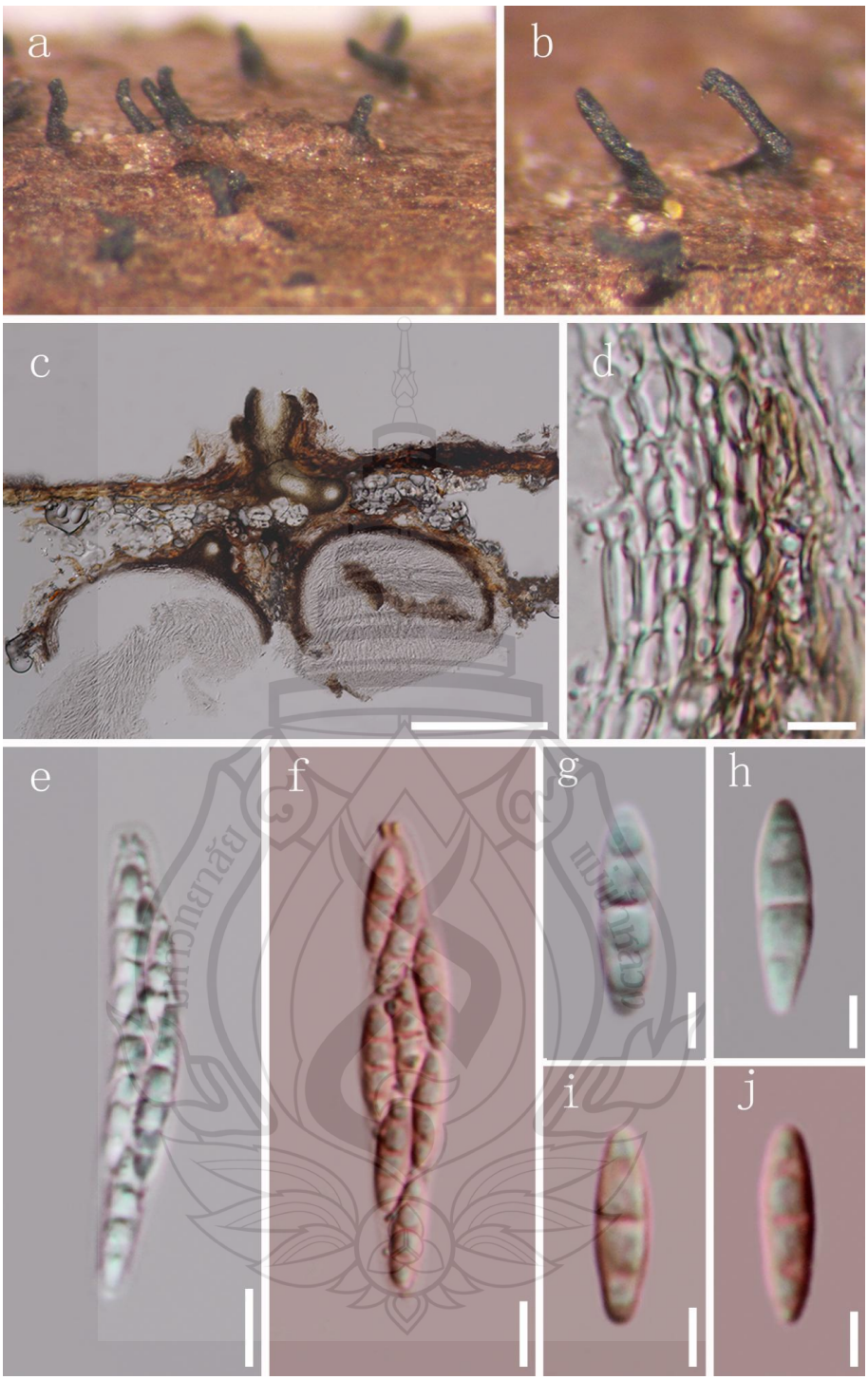
Figure 3.7 *Diaporthe sojae* (MFLU 18-2257)

Diaporthe novem J.M. Santos, Vrandečić & A.J.L. Phillips, Persoonia 27: 14 (2011) Figure 3.8

Saprobic on decaying woody stems. Sexual morph: *Ascomata* immersed in the host, scattered or gregarious, with black, hairy, long necks, bursting through the bark of host, coriaceous to carbonaceous. *Peridium* 20–38 μm, composed of several layers of hyaline to brown cells of *textura prismatica* to *angularis*, inwardly hyaline and thin-walled, and outwardly fusing with the host tissue. *Hamathecium* comprising abundant of asci, paraphyses not observed. *Asci* 39–45 (–48) × 6–9 μm, ($\overline{x}$ = 44 × 7 μm, n = 20), 8-spored, unitunicate, clavate, sessile, with visible refractive apical ring. *Ascospores* 11–13.5 × 2.5–3.5 μm, ($\overline{x}$ = 12 × 3 μm, n = 30), cylindrical, slightly tapered and rounded towards the ends, hyaline, guttulate when young, and medianly septate at maturity, randomly arranged, smooth walled. Asexual morph: Undetermined.

Material examined: CHINA, Guizhou Province, Xingyi City, on decaying woody stems, 9 July 2015, J.F. Zhang XY-1-1 (GZAAS 19-4020).

Notes: Present phylogenetic analysis in this study shows that our new collection clusters with the ex-type strains of *Diaporthe novem* (CBS 127270 and CBS 127271) and where they form a subclade with full bootstrap support values (Figure 3.6). Morphologically, they are similar in having subglobose ascomata with a long, hairly neck, sessile unitunicate asci with visible refractive apical ring, and cylindrical, medianly septate ascospores, including the overlapped measurements of asci and ascospores (Santos et al., 2011). Therefore, this new specimen is identified as *D. novem* in this study, and reported as a new geographical record for China as it has been reported widely distributed (Santos et al., 2011; Díaz et al., 2014, 2017; Petrović et al., 2016; Guarnaccia & Crous, 2017; Thompson et al., 2018; Lesuthu et al., 2019), but is first time found in China.



Note a, b Appearance of ascomata in the host surface. c Vertical section through ascomata. d Section of peridium. e, f Asci. g–j Ascospores. Scale bars: c = 200 µm, d–f = 10 µm, g–j = 5 µm.

Figure 3.8 *Diaporthe novem* (GZAAS 19-4020)

Sydowiellaceae Lar.N. Vassiljeva

Sydowiellaceae was established to accommodate the genus *Sydowiella* with *S. fenestrans* as the type species (Vasilyeva, 1987). Based on molecular analysis, Kruys and Castlebury (2012) confirmed that Sydowiellaceae is a monophyletic family within Diaporthales, and resolved the natural placement of *Cainiella* within this family. A series of phylogenetic studies have been processed by various authors (Rossman et al., 2007; Kruys & Castlebury, 2012; Maharachchikumbura et al., 2015), however, a comprehensive revision for Sydowiellaceae was still not carried out until Senanayake et al. (2017, 2018) provided an updated outline and key to families of Diaporthales, and they accepeted 15 genera in this family. Voglmayr and Mehrabi (2018) accommodated *Caudospora* in Sydowiellaceae based on multi-gene analysis, therefore, this family currently encompassed 16 genera, and this treatment was followed by the subsequent studies (Hyde et al., 2020b; Wijayawardene et al., 2020).

Neodiaporthe J.F. Zhang & K.D. Hyde, *gen. nov.*

Etymology: Named refers to it resembles to *Diaporthe* in having long necked ascomata and sessile asci.

Type species: *Neodiaporthe reniformesporum*

Saprobic on dead branch of unidentified woody plant. Sexual morph: *Ascomata* immersed to erumpent through the host surface, scattered, solitary or gregarious, depressed globose to subglobose, dark brown to black, coriaceous, ostiolate. *Ostiole* central, with a long neck that emerging through cracks in the substrate. *Peridium* composed of several layers of cells of *textura angularis*. *Hamathecium* comprising sparse, trabeculate, cellular paraphyses, intermixed with the asci. *Asci* unitunicate, cylindric-clavate, sessile, apex blunt, with J- apical ring. *Ascospores* reniform, hyaline, overlapping uniseriate to biseriate, tapering to the ends, one-septate, constricted at the septum, guttulate. Asexual morph: Undetermined.

Notes: *Neodiaporthe* is phylogenetically placed within Sydowiellaceae, and where it sisters to *Breviappendix* and *Sydowiella* in the present study (Figure 3.10). Morphologically, *Neodiaporthe* resembles to *Breviappendix* in having long-necked ascomata and sessile asci, whereas, they are distinguished by *Breviappendix* producing ellipsoid ascospores with setose appendages at both ends, and the paraphyses was also not appeared (Senanayake et al., 2017b). While *Neodiaporthe*

differs from *Sydowiella* by the latter was characterized by short papillated ascomata, cylindrical, short pedicellata asci that bearing oval to ellipsoid ascospores (Maharachchikumbura et al., 2016). Based on the present phylogenetic analysis and morphological characteristics, *Neodiaporthe* is established as a new genus to accommodate the type species *Neodiaporthe reniformesporum* that collected from karst formation in Guizhou Province, China.

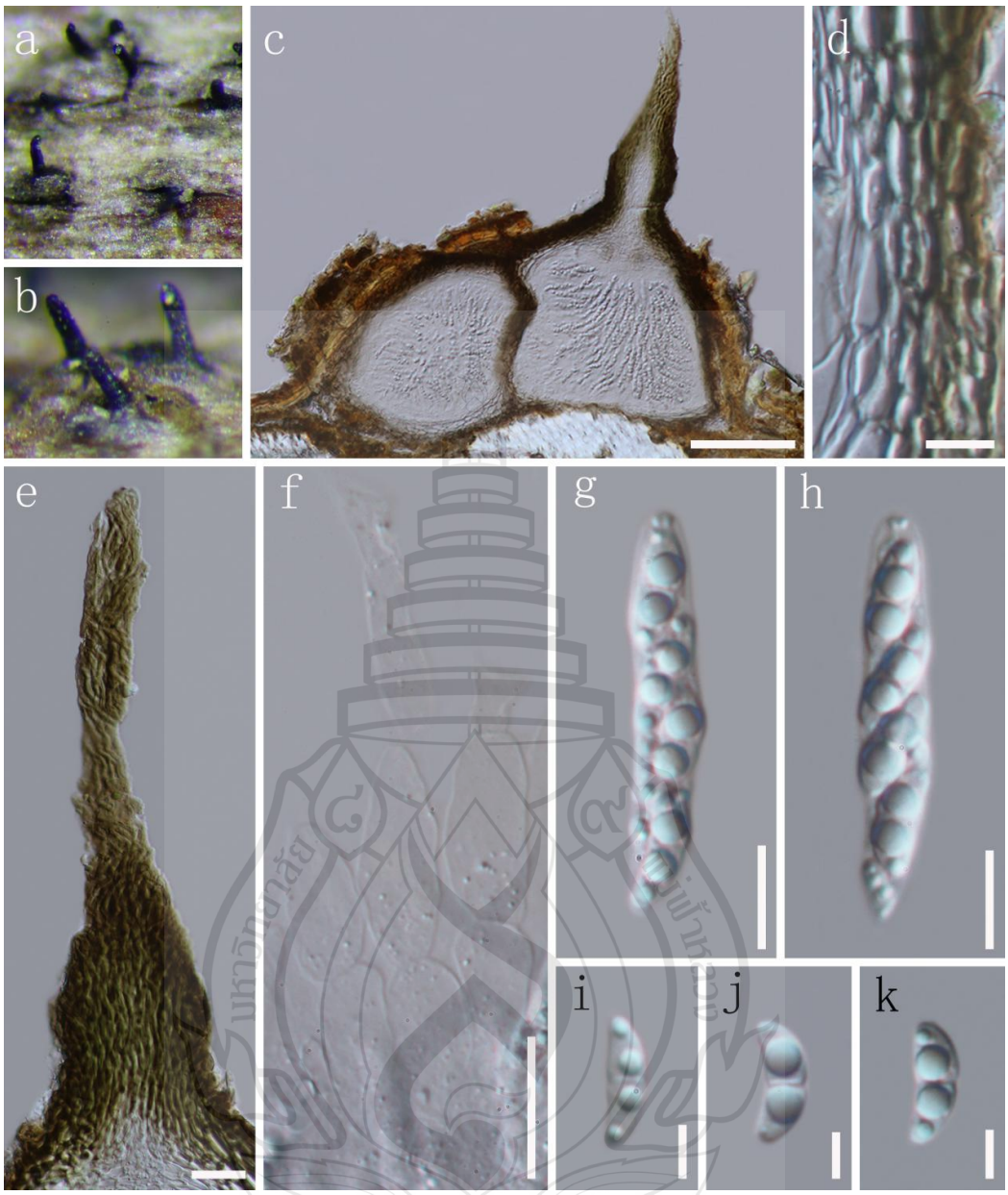
Neodiaporthe reniformesporum J.F. Zhang & K.D. Hyde, *sp. nov*., Figure 3.9

Etymology: Named refers to its reniform ascospores.

Holotype: GZAAS 19-1742.

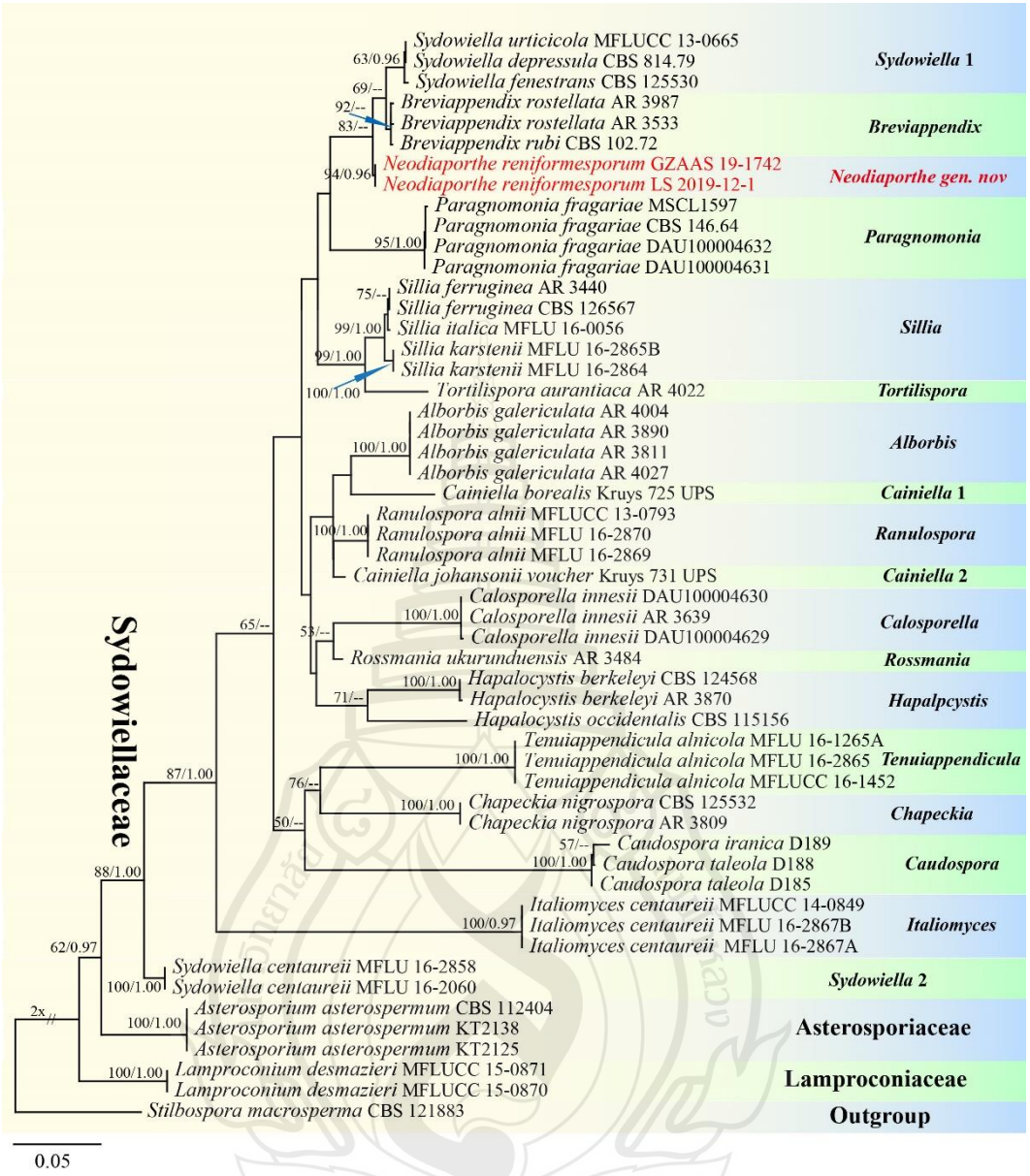
Saprobic on dead branch of unidentified woody plant. Sexual morph: *Ascomata* 338–441 μm high, 170–219 μm diam., immersed to erumpent through the host surface, scattered, solitary to gregarious, depressed globose to subglobose, dark brown to black, coriaceous, ostiolate. *Ostiole* central, with a long neck that emerging through cracks in the substrate, comprising dense, brown, thick-walled pseudoparenchymatous cells. *Peridium* 19–31 μm wide, composed of several layers of cells of *textura angularis*, hayline and thin inwardly, becoming brown and thick-walled outwardly. *Hamathecium* comprising 7–11 μm wide, sparse, trabeculate, cellular paraphyses, swollen at the base, intermixed with the asci. *Asci* 37–42(–45.5) × 6–9(–11) μm ($\bar{x}$ = 40.5 × 8 μm, n = 25), 4–6-spored, unitunicate, cylindric-clavate, sessile, apex blunt with a J- apical ring. *Ascospores* 11–14 × 4–5 μm ($\bar{x}$ = 12.5 × 4.5 μm, n = 40), reniform, hyaline, overlapping uniseriate to biseriate, tapering to the ends, 1-septate, constricted at the septum, normally four guttulates, smaller near the ends, and larger around the septum, without any gelatinous/mucilaginous appendages. Asexual morph: Undetermined.

Material examined: CHINA, Guizhou Province, Leishan City, on dead branch of unidentified woody plant, 24 May 2019, J.F. Zhang LS-2019-12 (GZAAS 19-1742, holotype).



Note a, b Ascomata erumpent through the host with a long neck. c Vertical section through ascomata. d Section of peridium. e Long neck. f Paraphyses. g, h Asci i–k Young and mature ascospores. Scale bars: c = 100 μm, d, g, h = 10 μm, e, f = 20 μm, i–k=5 μm.

Figure 3.9 *Neodiaporthe reniformesporum* (GZAAS 19-1742, holotype)



Note Maximum likelihood bootstrap values equal or greater than 50% and Bayesian posterior probabilities equal or greater than 0.95 are indicated near the nodes. New sequence generated in this study is given in red. The tree is generated from maximum likelihood (ML) analysis based on LSU, ITS and TEF1-α sequence data, and rooted with *Stilbospora macrosperma* CBS 121883.

Figure 3.10 Phylogenetic tree of the family Sydowiellaceae

Magnaporthales Thongk. et al.

Magnaporthales was established by Thongkantha et al. (2009) to accommodate the family Magnaporthaceae based on phylogeny. Klaubauf et al. (2014) resolved the nature polyphyletic placement of *Pyricularia*, and defined two families Ophioceraceae and Pyriculariaceae which were comprised of Magnaporthales on the basis of morphological and phylogenetic analysis. Hongsanan et al. (2017) provided an update phylogeny of Sordariomycetes based on morphology and molecular clock evidence, and accommodated Pseudohalonectriaceae under Magnaporthales. Subsequently, Luo et al. (2019) introduced the fifth family Ceratosphaeriaceae to Magnaporthales, and this classification treatment was followed by Hyde et al. (2020b) and Wijayawardene et al. (2020) in their studies.

Magnaporthaceae P.F. Cannon

Magnaporthaceae was introduced by Cannon (1994) to accommodate the genera *Buergenerula*, *Clasterosphaeria*, *Gaeumannomyces*, *Herbampulla*, *Magnaporthe* and *Omnidemptus*. The genus within this family has been continued discovered and accepted in recent years (Luo & Zhang, 2013; Klaubauf et al., 2014; Luo et al., 2014, 2015; Liu et al., 2015; Crous et al., 2015b; Hernández-Restrepo et al., 2016). According to the the latest update of accounts of this family and the outline of Ascomycota (Silva et al., 2019; Custódio et al., 2020; Hyde et al., 2020b; Wijayawardene et al., 2020), twenty-three genera were accepted in Magnaporthaceae.

Neoceratosphaeria J.F. Zhang & Z.Y. Liu, *gen. nov.*

Etymology: Named refers to it resembles to *Ceratosphaeria* in having long-beaked ascomata associated in substrate.

Type species: *Neoceratosphaeria karstiea*

Saprobic on dead culm of unidentified herbaceous plant. Sexual morph: *Ascomata* immersed to erumpent, globose to subglobose, long-beaked, black to dark brown, ostiolate. *Ostiole* central, with periphyses lined within the ostiolar canal. *Peridium* composed of several layers of cells of *textura angularis*. *Hamathecium* comprising of septate, cellular paraphyses, taping towards the apex. *Asci* 8-spored, unitunicate, broadly cylindrical to clavate, apedicellate, apically rounded to truncate, with an inconspicuous apical ring. *Ascospores* faciculate, hyaline, filiform, curved to sigmoid, 2–5-euseptate at maturation. Asexual morph: Undetermined.

Notes: Phylogenetic analysis in the present study shows that *Neoceratosphaeria* forms an independent clade and closes to *Muraeriata* by strong statistical support (Figure. 3.13). However, these two genera can be morphologically distinguished from one another by the shape and seriation of ascospores. Ascospore in *Muraeriata* is narrowly fusiform with curved ends, that arranged in ascus with tri–tetra seriation (Huhndorf et al., 2008), while *Neoceratosphaeria* is characterized by broadly cylindrical asci that bearing faciculate, filiform, sigmoid ascospores. Additionally, *Neoceratosphaeria* is somewhat similar to *Aquafiliformis* in having long-necked ascomata, sepetate, cellular paraphyses and cylindric-clavate asci, nevertheless, they are differentiated by the species in *Aquafiliformis* was collected from aquatic habitat, and having comparatively larger sized asci and ascospores, and the ascospores are non-septate, whereas, *Neoceratosphaeria* is found in terrestrial, and the ascospores of this genus are 2–5-euseptate at maturation (Luo et al., 2019). According the morphological and phylogenetic analysis, *Neoceratosphaeria* is therefore introduced as a new genus to accommodate the type species *Neoceratosphaeria karstiea.*

Neoceratosphaeria karstiea J.F. Zhang & Z.Y. Liu, *sp. nov.*, Figure 3.11

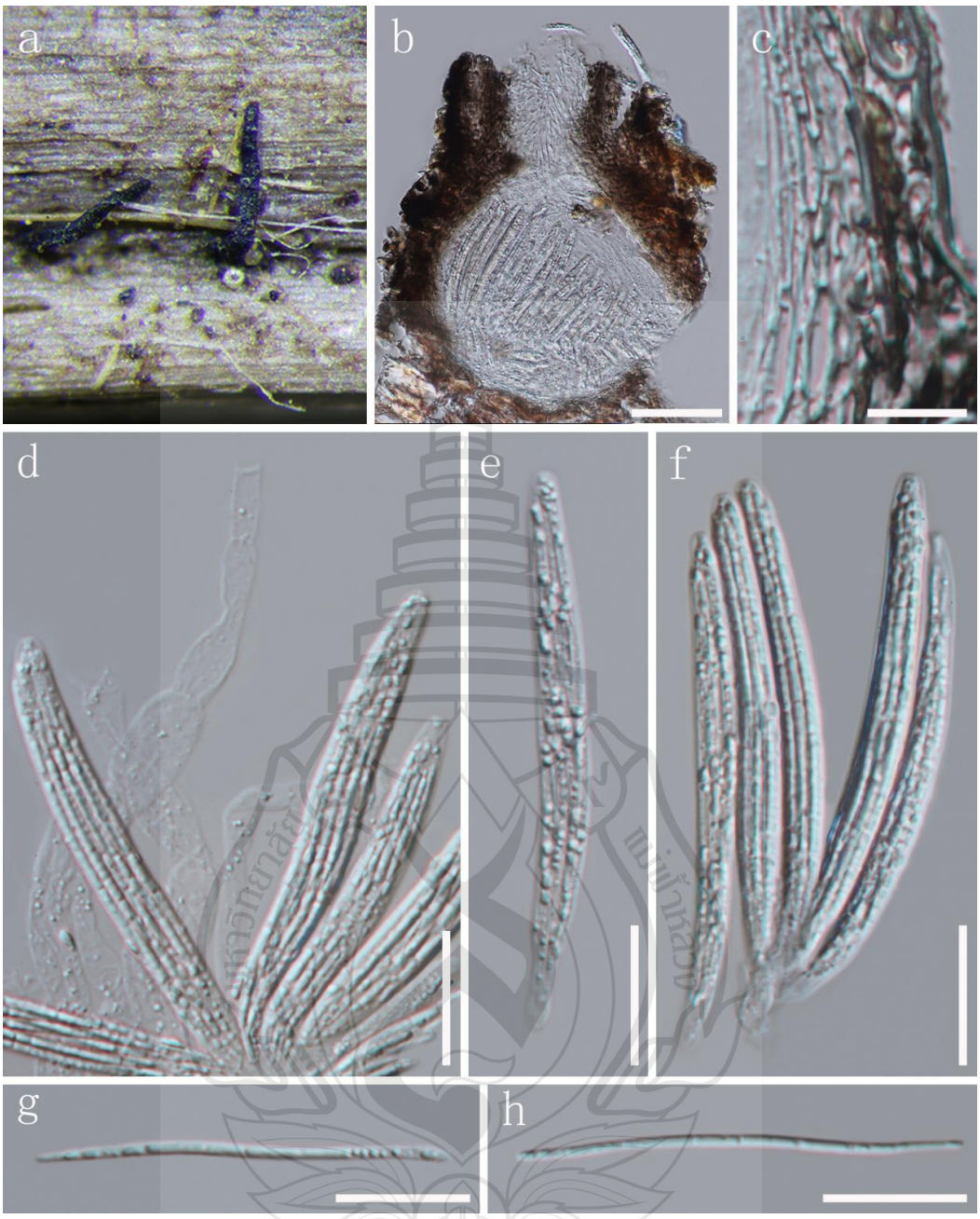
Etymology: Named refers to the habitat where the holotype was collected.

Holotype: GZAAS 19-1767.

Saprobic on dead culm of unidentified herbaceous plant. Sexual morph: *Ascomata* immersed to erumpent through host surface with a long, carbonaceous neck, solitary or gregarious, globose to subglobose, black to dark brown, ostiolate. *Ostiole* central, with periphyses lined within the ostiolar canal. *Peridium* laterally 15–25 μm wide, composed of several layers of hyaline to pale pigmented, thin-walled cells of *textura angularis*, and with outer layer composed of dark brown and thick-walled cells confusing with host tissue. *Hamathecium* consisting of abundant, 3–10 μm wide, septate, cellular paraphyses, swollen at the base, and taping towards the apex, anastomosing between and above the asci. *Asci* (67.5–) 72.5–79.5 (–81.5) × 6–8.5 μm ($\bar{x}$ = 75 × 7 μm, n=25), 8-spored, unitunicate, cylindrical, apedicellate, apically rounded to truncate, with an inconspicuous apical ring. *Ascospores* (62.5–) 65–69 (–71) × 1.5–2 μm ($\bar{x}$ = 67 × 1.75 μm, n=30), faciculate, hyaline, filiform, curved to

sigmoid, towards narrowly in one end and round in another end, 2–5-euseptate at maturation, smooth-walled. Asexual morph: Undetermined.

Material examined: CHINA, Guizhou Province, Libo County, Maolan Town, on dead culm of unidentified herbaceous plant, 17 July 2017, J.F. Zhang ML-1 (GZAAS 19-1767, holotype).



Note a Appearance of ascomata erumpent through host surface with long neck. b Vertical section through ascomata. c Section of peridium. d Paraphyses anostomosing with the asci. e, f Asci. g, h Ascospores. Scale bars: b = 50 μm, c = 10 μm, d–h = 20 μm.

Figure 3.11 *Neoceratosphaeria karstiea* (GZAAS 19-1767, holotype)

Ophioceraceae Klaubauf et al.

Ophioceraceae was introduced by Klaubauf et al. (2014) to accommodate the type genus *Ophioceras* based on its separate phylogenetic placement within Magnoporthales. So far, only the type genus was comprised of this family (Hyde et al., 2020b; Wijayawardene et al., 2020).

Ophioceras Sacc., Syll. fung. (Abellini) 2: 358 (1883)

Ophioceras was established by Saccardo (1883) with the lectotype *O. dolichostomum*. It resembles to *Gaeumannomyces*, Conway & Barr (1978) however distinguished these two genera depending on different nutritional type. Chen et al. (1999) indicated that these two genera are phylogenetically closely related, and placed them in Magnaporthaceae. Nevertheless, Klaubauf et al. (2014) established Ophioceraceae to accommodate *Ophioceras* based on multi-gene phylogeny. Recently, Jiang et al. (2021) appraised *Ophioceras* based on morphological and phylogenetic evidence, and treated *Ceratosphaerella* as the synonym under *Ophioceras*.

Ophioceras guizhouense J.F. Zhang & J.K. Liu, *sp. nov*., Figure 3.12

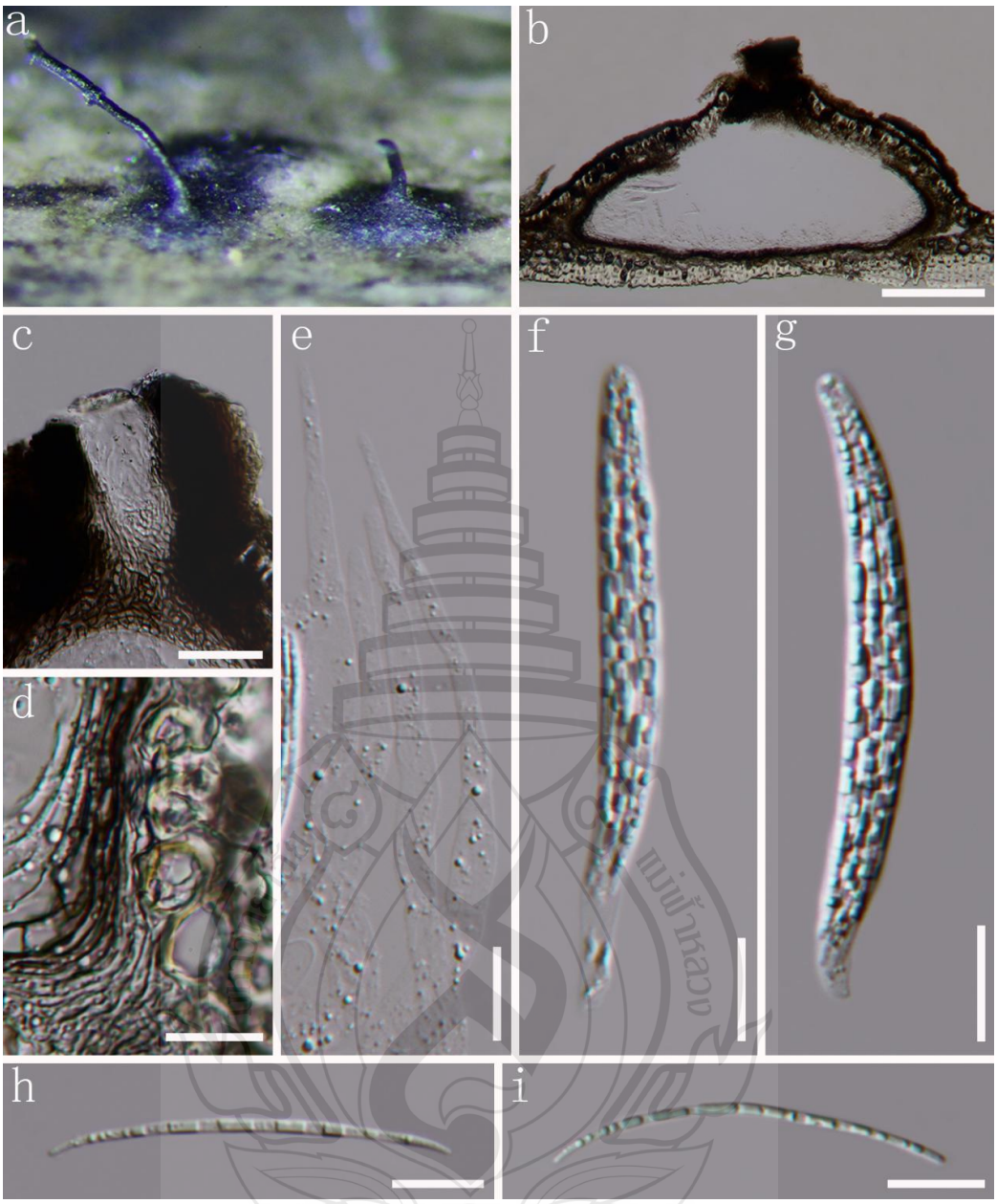
Etymology: Named refers to Guizhou where the holotype was collected.

Holotype: MFLU 18-2277.

Saprobic on dead bamboo culms, visible as a dome-shaped sports with a long, hair-like neck in substrate. Sexual morph: *Ascomata* semi-immersed to superficial, depressed hemispherical, solitary or gregarious, shiny black, coriaceous, bearing a very long, cylindrical, black, fragile neck. *Ostiole* central, with sparse periphyses lined with the ostiolar canal. *Peridium* 15–42 μm wide, composed of several layers of large cells of *textura angularis*, pale brown to hyaline towards inside, and become brown to balck outwardly. *Hamathecium* comprising numerous, 2.5–6.5 μm wide, trabeculate, cellular paraphyses, swollen at the base, tapering distally, anastomosing between and above the asci. *Asci* (48.5–) 51–56 (–59) × 4.5–6 μm ($\overline{x}$ = 53.5 × 5 μm, n=30), 8-spored, subcylindrical, apedicellate, unitunicate, with a J-, thimble-shaped, subapical apparatus. *Ascospores* (41–) 45.5–49 (–53) × 1.2–1.7 μm ($\overline{x}$ = 47 × 1.4 μm, n=30), fasciculate, filiform, slightly acute at each end, falcate or sigmoid, 3–6-septate at maturation, not constricted at the septa, hyaline, smooth-walled, guttulate. Asexual morph: Undetermined.

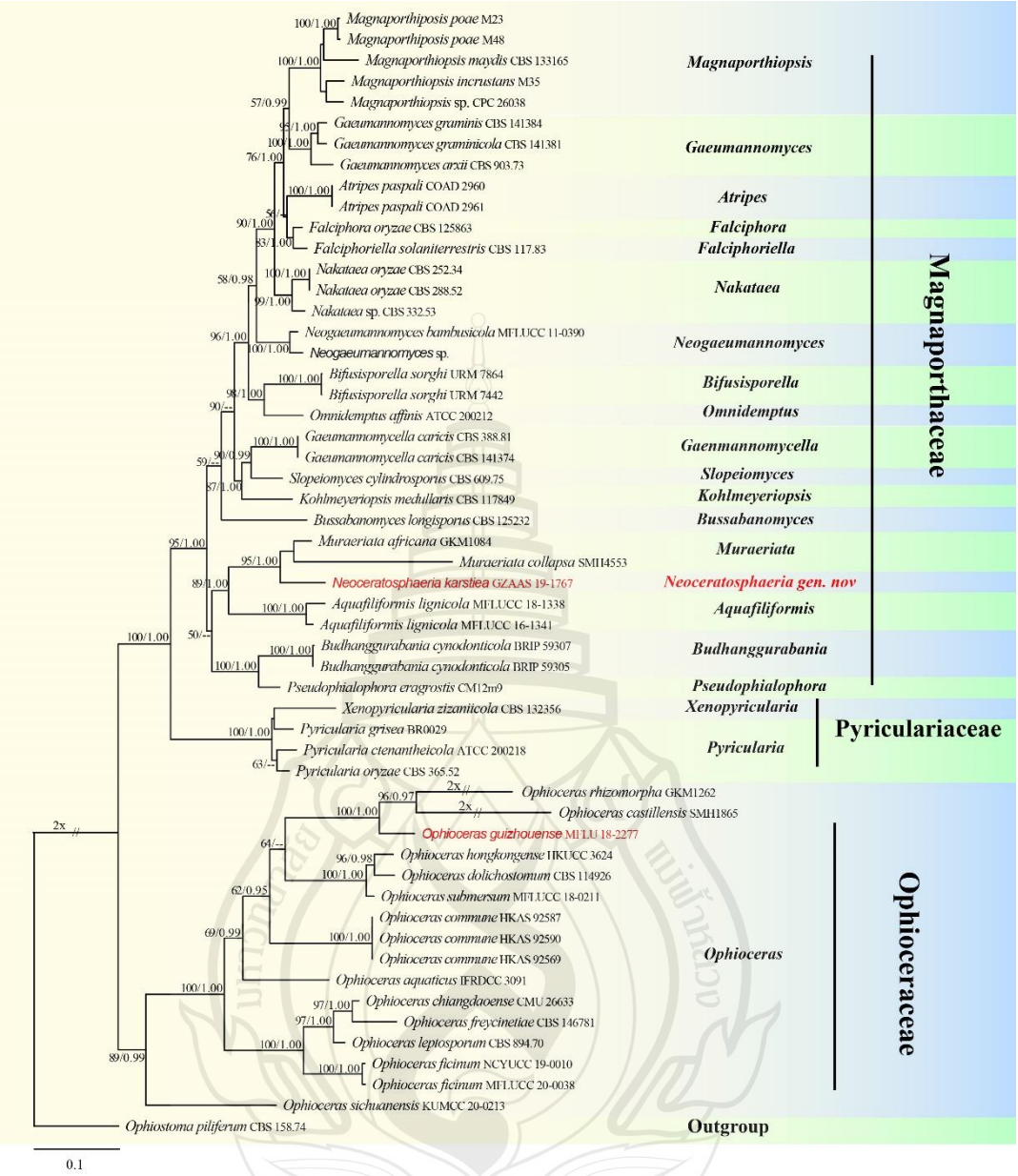
Material examined: CHINA, Guizhou Province, Libo County, Maolan Town, Maolan National Nature Reserve, on dead bamboo culms, 18 July 2016, J.F. Zhang MLAA-7 (MFLU 18-2277, holotype).

Notes: *Ophioceras guizhouense* is typical of *Ophioceras* in having long-necked ascomata, subcylindrical, sessile asci that producing hyaline, filiform spetate ascospores. However, it differs from other *Ophioceras* taxa by having smaller asci and ascospores, except for the ascospores of *O. castillensis* are smaller than our new collection (Jiang et al., 2021). Phylogenetically, *O. guizhouense* is placed within *Ophioceras* and formed an independent clade that sister to *O. castillensis* and *O. rhizophoma* with high bootstrap support (Figure 3.13). Based on the above analysis, *Ophioceras guizhouense* is introduced as a new species.



Note a Appearance of ascomata in host surface with long neck. b Vertical section through ascomata. c Ostiole with periphyses. d Section of peridium. e Paraphyses. f, g Asci. h, i Ascospores. Scale bars: b = 200 μm, c = 50 μm, d–i = 10 μm.

Figure 3.12 *Ophioceras guizhouense* (MFLU 18-2277, holotype)



Note Maximum likelihood bootstrap values equal or greater than 50% and Bayesian posterior probabilities equal or greater than 0.95 are indicated near the nodes. New sequence generated in this study are given in red. The tree is generated from maximum likelihood (ML) analysis based on ITS, LSU and Rpb1 sequence data, and rooted with *Ophiostoma piliferum* CBS 158.74.

Figure 3.13 Phylogenetic tree of the order Magnaporthales

Subclass Hypocreomycetidae O.E. Erikss. & Winka

Glomerellales Chadef. ex Réblová et al.

The original name of Glomerellales was first proposed by Chadefaud (1960) with "Glomérellales" to accommodate a group of endophytic fungi and parasites of living plants with ascomata that varying from endostromatal to apostromatal and ascospores that are often unicellular and hyaline, however, a Latin diagnosis was not provided. Around half a century, Zhang et al. (2006) provided systematic overview of Sordariomycetes based on multi-gene phylogeny, and validated the family Glomerellaceae. Then, the order Glomerellales was formally introduced and validated by Réblová et al. (2011) with detail descriptions and diagnosis, as well as phylogeny affinities. And in their study, three families viz. Australiascaceae, Glomerellaceae and Reticulascaceae were accpeted in Glomerellales (Réblová et al., 2011). Maharachchikumbura et al. (2015) provided a backbone tree for Sordariomycetes and assigned another family Plectosphaerellaceae within this order. Subsequently, Tibpromma et al. (2018) introduced a monotypic family Malaysiascaceae to this order based on morphology and phylogeny. However, the divergence time of Malaysiascaceae was estimated by Hyde et al. (2020c) and indicated that it falls in the genus status. Therefore, the above family were currently accepted in Glomerellales, but the status of Malaysiascaceae may need revision following further study (Hyde et al., 2020c; Wijayawardene et al., 2020).

Plectosphaerellaceae W. Gams et al.

Plectosphaerellaceae was introduced by Zare et al. (2007) to acoommodate the genera *Acrostalagmus*, *Gibellulopsis*, *Musicillium* and the type genus *Plectosphaerella*. It has been listed as families *incertae sedis* in the Hypocreomycetidae (Zhang et al., 2007), and until Maharachchikumbura et al. (2015) presented the multi-gene analysis of Sordariomycetes, and indicated that Plectosphaerellaceae is referred to the order Glomerellales. Accounts of genera within this family were continually increased by a serious of studies (Kirk et al., 2008; Réblová et al., 2011; Cannon et al., 2012; Grum-Grzhimaylo et al., 2013, 2016; Hirooka et al., 2014; Maharachchikumbura et al., 2016; Giraldo et al., 2017b; Hyde et al., 2017; Tibpromma et al., 2018; Giraldo & Crous, 2019), and there are currently 25

genera accepted in Plectosphaerellaceae (Hyde et al., 2020c; Phukhamsakda et al., 2020; Wijayawardene et al., 2020).

Truncatascus J.F. Zhang & J.K. Liu, *gen. nov.*

Etymology: Named based on Latin '*truncatum*' and '*ascus*' refers to its ascus with a truncate apex.

Type species: *Truncatascus microsporum*

Saprobic on dead branch of unidentified woody plant. Sexual morph: *Ascomata* superficial, aggregated, globose to subglobose scattered, black, short papillate, ostiolate, coriaceous. *Ostiole* central, paraphysate. *Peridium* composed of several layers of pseudoparenchymatous cells of *textura angularis*. *Hamathecium* comprising abundant, hypha-like paraphyses, intermixed with and above the asci. *Asci* 8-spored, unitunicate, cylindrical, short pedicellate, apically truncate, without apical apparatus. *Ascospores* oval, one-celled, overlapping uniseriate, guttulate. Asexual morph: Undetermined.

Notes: Multi-gene phylogenetic analysis in the present study placed *Truncatascus* within Plectosphaerellaceae, and where it forms a seperate clade that close to the genus *Fuscohypha* (Figure 3.15). However, these two genera can be distinguished by the sexual morph of *Fuscohypha* was characterized by having obclavate to obpyriform asci that bearing one-septate, ellipsoidal to subfusoid ascospores with numerous guttulates (Phookamsak et al., 2019; Phukhamsakda et al., 2020), while *Truncatascus* is presented in having cylindrical asci and non-septate, ovoid ascospores that only with two guttulates near the both ends. Observation of the morphological characteristics of this new collection, we found that it is not fit any know genera within Plectosphaerellaceae. Hence, we introduce it as a new genus to accommodate the type species *Truncatascus microsporum*.

Truncatascus microsporum J.F. Zhang & J.K. Liu, *sp. nov*., Figure 3.14

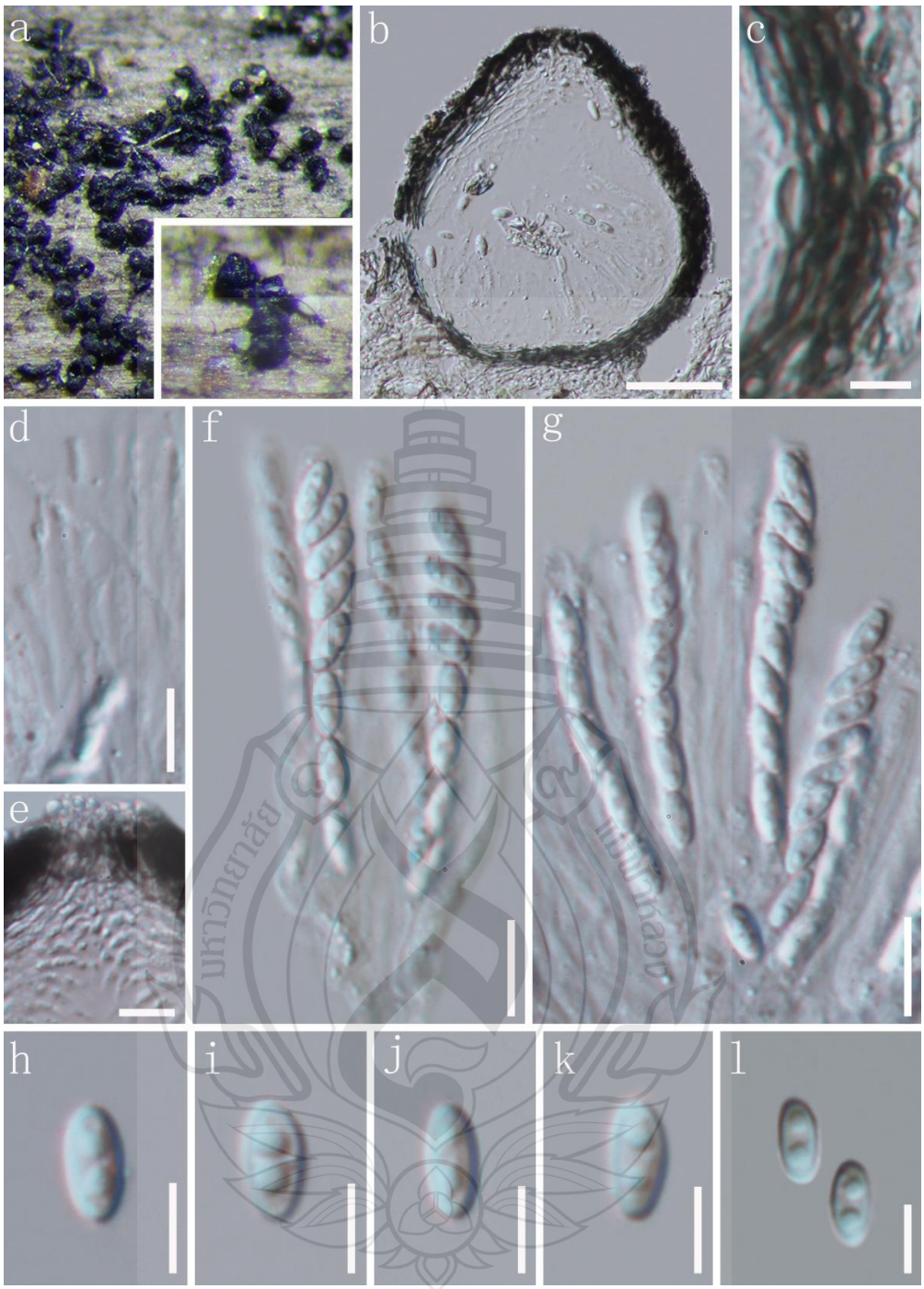
Etymology: Named refers to its smaller sized ascospores.

Holotype: GZAAS 19-1744.

Saprobic on dead branch of woody plant, visible as black spots in the substrate surface. Sexual morph: *Ascomata* superficial, black, uni-loculate, gregarious, glaborus, coriaceous, short papillate, ostiolate, *Ostiole* central, internally covered with numerous paraphysis. *Peridium* 10–12 µm wide, composed of several layers of

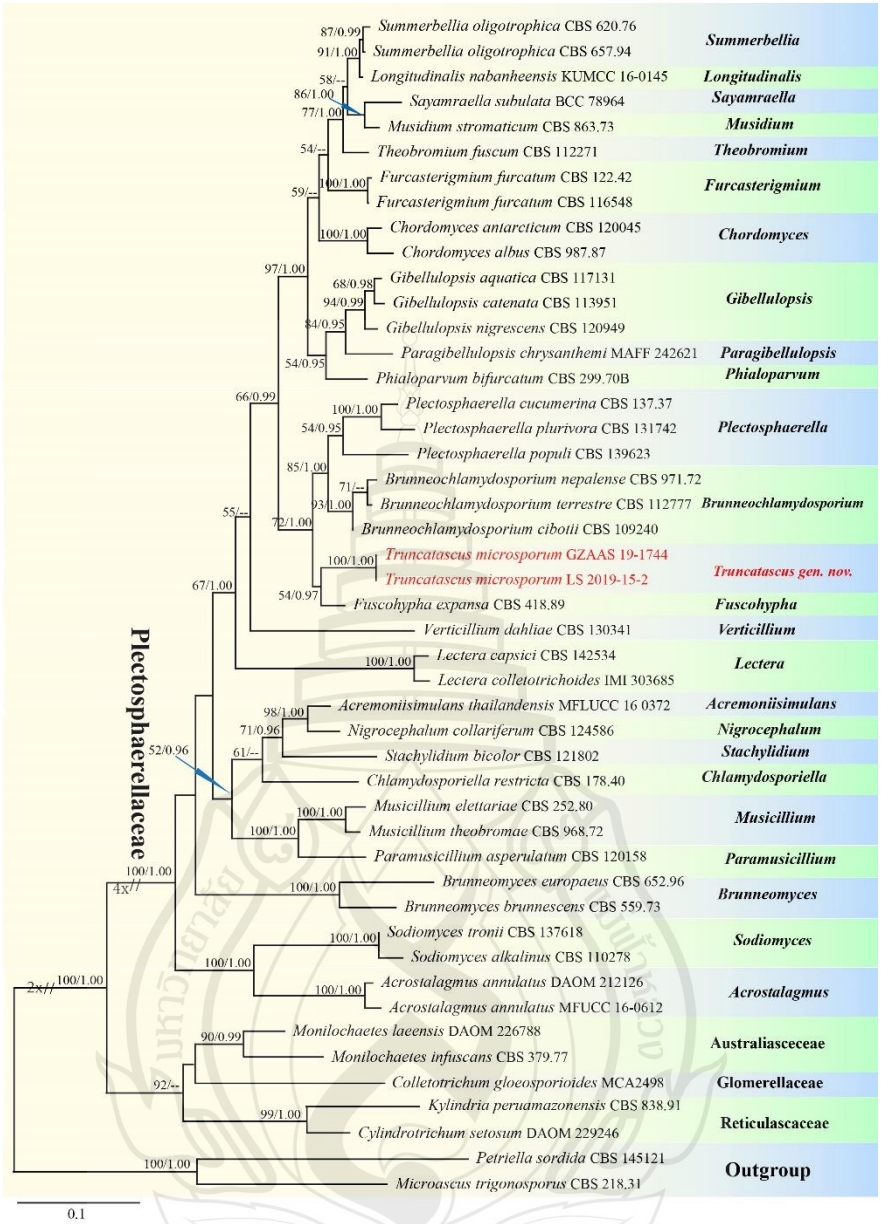
pseudoparenchymatous cells of *textura angularis*, thick-walled, and dark brown outwardly, becoming thin-walled and subhyaline inwardly. *Hamathecium* comprising 1–1.5 µm wide, non-septate, hypha-like paraphyses, intermixed with and above the asci. *Asci* (38–)41–43.5(–46) × (3–)4–5.5 µm ($\bar{x}$ = 42.5 × 4.5 µm, n = 30), 8-spored, unitunicate, cylindrical, short pedicellate, apically truncate, without apical apparatus. *Ascospores* 6–8 × 2.5–3.5 µm ($\bar{x}$ = 7 × 3 µm, n = 35), oval, one-celled, overlapping uni-seriate, normally two guttulates near the both ends, wall smooth. Asexual morph: Undetermined.

Material examined: CHINA, Guizhou Province, Leishan County, on dead branch of woody plant, 24 May 2019, J.F. Zhang LS-2019-15 (GZAAS 19-1744, holotype).



Note a Appearance of ascomata in host surface. b Vertical section through ascoma. c Section of peridium. d Paraphyses. e Ostiole with paraphysis. f, g Asci. h–l Ascospores. Scale bars: b = 30 µm, c, h–l = 5 µm, d–g = 10 µm.

Figure 3.14 *Truncatascus microsporum* (GZAAS 19-1744, holotype)



Note Maximum likelihood bootstrap values equal or greater than 50% and Bayesian posterior probabilities equal or greater than 0.95 are indicated near the nodes. New sequence generated in this study are given in red. The tree is generated from Maximum likelihood (ML) analysis based on LSU ITS, Rpb2 and SSU sequence data, and rooted with *Petriella sordaria* CBS 145121 and *Microascus trigonosporus* CBS 218.31.

Figure 3.15 Phylogenetic tree of the order Glomerellales

Hypocreales Lindau

Hypocreales was established by Lindau (1897) to accommodate the single family Hypocreaceae with 6 subfamilies. Later, a series of taxonomic studies for this order were carried out by various authors in the past more than a century (Earle, 1901; Seaver, 1909a, b, 1910a, b, 1911; Nannfeldt, 1932; Luttrell, 1951, 1955; Ainsworth, 1961; Alexopoulos, 1962; Rogerson, 1970; Rehner & Samuels, 1995; Rossman, 1996; Kirk et al., 2008; Crous et al., 2015b, 2018a, b; Maharachchikumbura et al., 2016; Hongsanan et al., 2017; Sun et al., 2017; Wijayawardene et al., 2018), and currently 14 families, which namely: Bionectriaceae, Calcarisporiaceae, Clavicipitaceae, Cocoonihabitaceae, Cordycipitaceae, Flammocladiellaceae, Hypocreaceae, Myrotheciomycetaceae, Nectriaceae, Niessliaceae, Ophiocordycipitaceae, Sarocladiaceae, Stachybotryaceae, and Tilachlidiaceae were accepted in Hypocreales by Hyde et al. (2020c) based on molecular evidence.

Nectriaceae Tul. & C. Tul.

Nectriaceae was originally reported as '*Nectriei*', and first spelt correctly at family rank by Nitschke (1869). Members of this family are normally characterized by uni-loculate ascomata that are brightly pigmented with fusiform to allantoid ascospores and globose to fusiform phialidic conidia, and occur worldwide (Rossman et al., 1999, 2000, 2013; Chaverri et al., 2011; Schroers et al., 2011; Lombard et al., 2015). The majority of these species were reported as saprobes, plant pathogens, fungicolous, insecticolous, and even human pathogens (Rossman et al., 1999, 2000, 2013; Chang et al., 2006; Chaverri et al., 2011; de Hoog et al., 2011; Gräfenhan et al., 2011; Schroers et al., 2011; Guarro, 2013; Lombard et al., 2015). Hyde et al. (2020c) accepted 69 genera within Nectriaceae in their monograph of refinement families of sordariomycetes. And subsequently, another genus *Longinectria* was found in cheese and accommodated in this family by Savary et al. (2021).

Nectria (Fr.) Fr., Summa veg. Scand., Sectio Post. (Stockholm): 387 (1849)

The name of *Nectira* was first proposed by Fries (1825) as a section of the genus *Hypocrea*. De Notaris (1844) changed the name *Hypocrea* to *Hypocreacei* and suggested that each section of *Hypocreacei* should be seperated as an independant genera. Then, Fries (1849) formally erected the genus *Nectria* with diagnosis, but without designating a type species. Rossman et al. (1999) recognized the legitimate

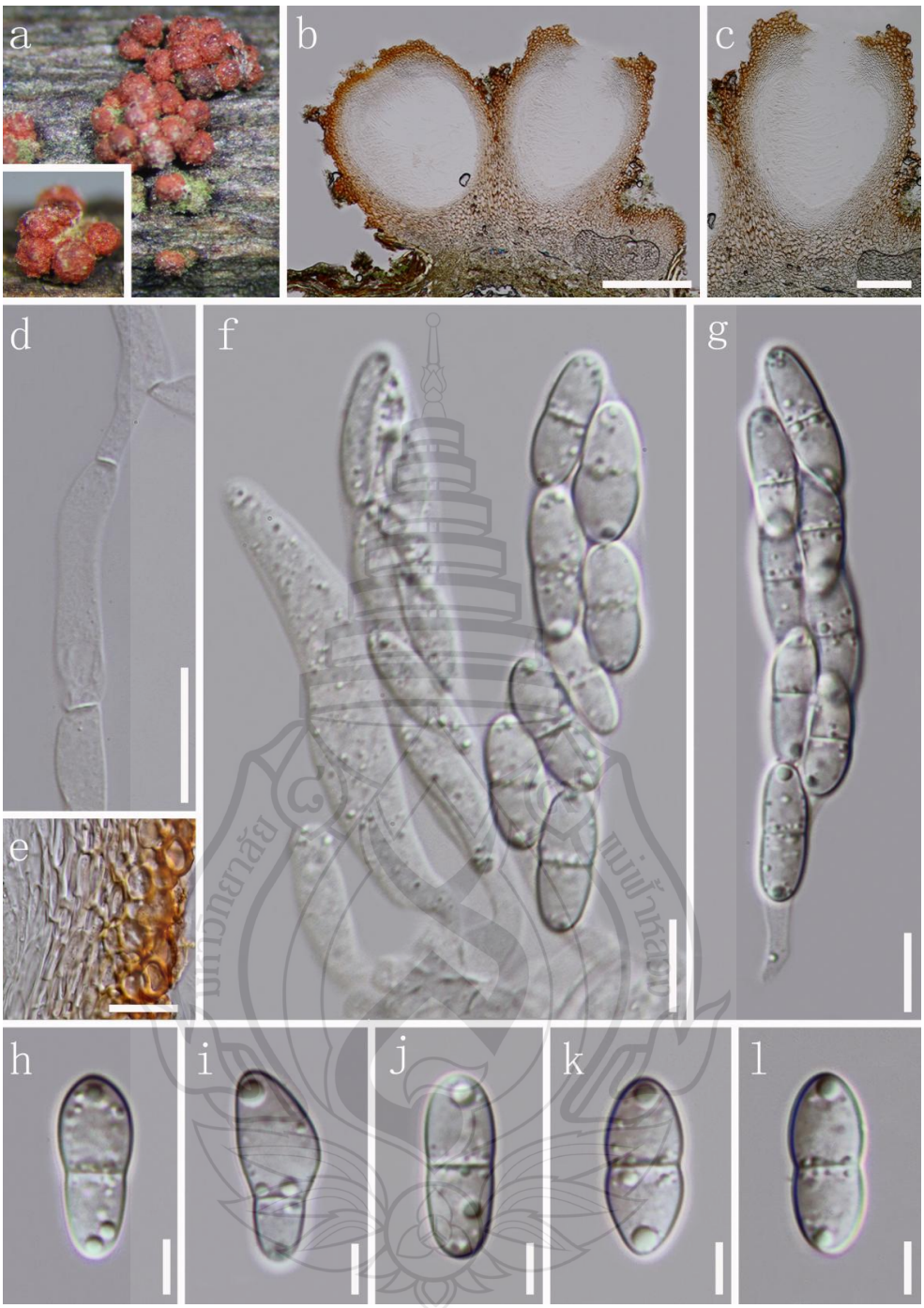
typification by Clements and Shear (1931) who designated *N. cinnabarina* as the lectotype of *Nectria*, and indicated that the type *N. peziza* desiganated by Seaver (1909) was illegitimate. Members of accounts in this genus was restricted to 27 by Rossman et al. (1999), and 14 novel species were discoverd in later studies (Döbbeler, 2005; Hirooka et al., 2011, 2012; Jaklitsch & Voglmayr, 2011; Rossman et al., 2013; Vasilyeva et al., 2013; Zeng et al., 2017; Yang et al., 2018; Dayarathne et al., 2020; Ma et al., 2020).

Nectria cinnabarina (Tode: Fr.) Fr., Summa Veg. Scand. 2: 388. 1849. Figure 3.16

Saprobic on dead branch of unidentified woody plant. Sexual morph: *Ascomata* 303–524 µm high, 335–451 µm diam., superficial, gregarious, subglobose to globose, red to reddish brown, rarely with mycelium around the surface, apappilate. *Peridium* 46–123 µm, composed of two strata, of outer layer comprising thick-walled, pigmented, pseudoparenchymatous cells of *textura globulosa* or *angularis*, inner layer of elongated, thin-walled, subhyaline cells of *textura prismatica*. *Hamathecium* comprising 3–8 µm, septate, branched paraphyses. *Asci* 70–81(–87) × 11–14 µm ($\bar{x}$ = 78 × 12.5 µm, n = 25), unitunicate, 8-spored, cylindrical to narrowly clavate, tapping towards the pedicel. *Ascospores* (13.5–)16–19 × 6–7 µm ($\bar{x}$ = 17 × 6.5 µm, n = 45), uniseriate below and biseriate above, hyaline, ellipsoidal to fusiform, straight, one-septate, constricted at the septum, sometimes guttulate, smooth-walled. Asexual morph: Undetermined.

Material examined: CHINA, Guizhou Province, Leishan County, on dead branch of woody plant, 2 November 2017, J.F. Zhang LS-22 (GZAAS 19-1763).

Notes: Our new collection clusters together with *N. cinnabarina* with high bootstrap value (Figure 3.18). Observation of the gross morphological characteristics of this new specimen, we found that it well fits with *N. cinnabarina* by having subglobose to globose, red to reddish brown ascomata with rarely mycelium around the surface, same textura of peridium, cylindrical to narrowly clavate asci, and ellipsoidal to fusiform, one-septate ascospores. Moreover, the measurements of asci and ascospores between the new collection and *N. cinnabarina* are also overlapped (Hirooka et al., 2011). Therefore, we identify the new specimen as *Nectria cinnabarina*.



Note a Appearance of ascomata in host surface. b, c Vertical section through ascoma. d Paraphyses. e Section of peridium. f, g Asci. h–l Ascospores. Scale bars: b = 200 μm, c = 100 μm, d, e = 20 μm, f, g = 10 μm, h–l = 5 μm.

Figure 3.16 *Nectria cinnabarina* (GZAAS 19-1763)

Neonectria Wollenw., Annls mycol. 15(1/2): 52 (1917)

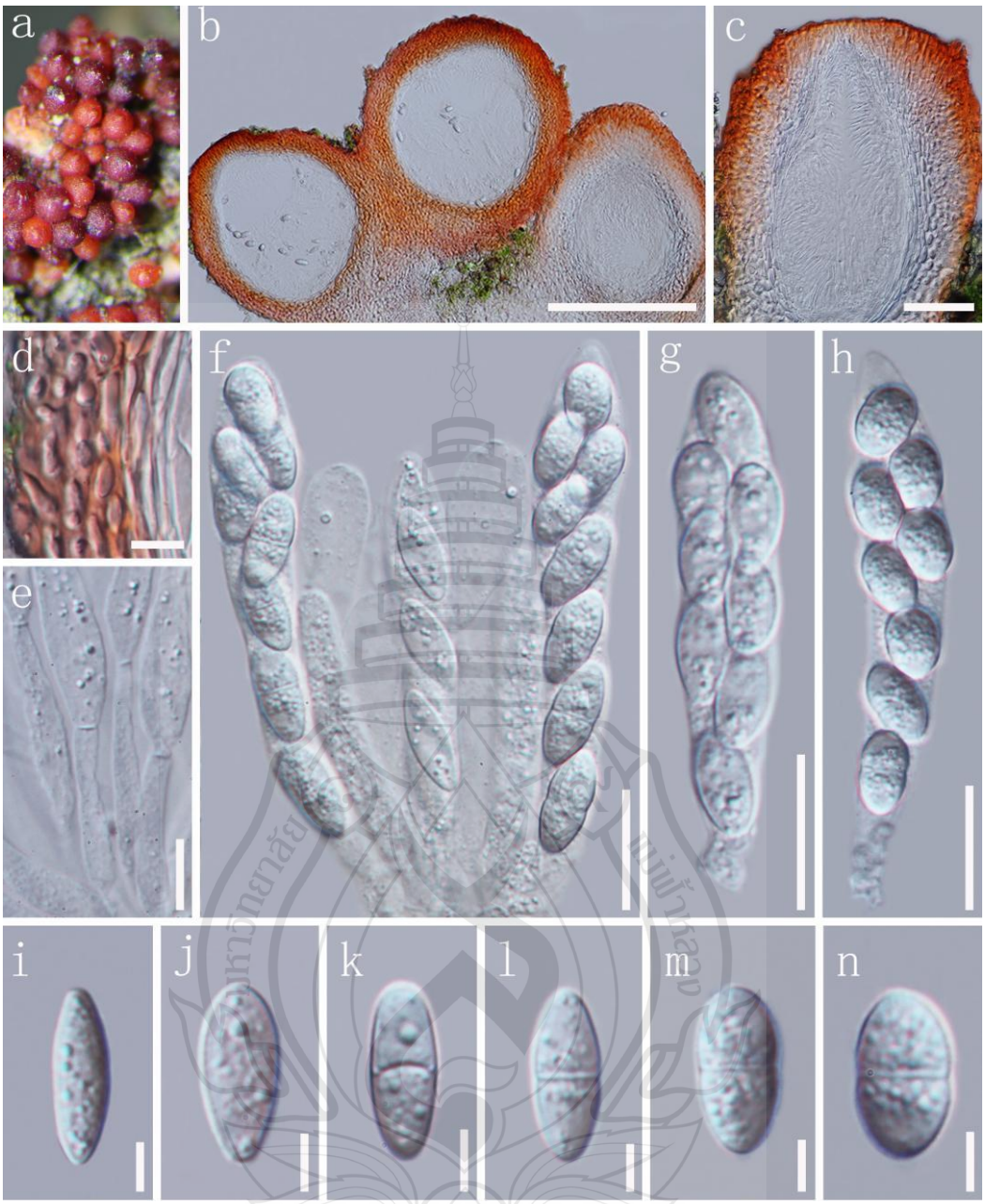
Neonectria was introduced by Wollenweber (1917) with *N. ramulariae* as the type species. Members of this genus are usually occurred on tropical and temperate regions, and some species are reported as plant pathogens that causing cankers, root rots, and other diseases on hardwood and coniferous trees (Samuels & Brayford, 1994; Rossman et al., 1999; Hirooka et al., 2005; Kobayashi et al., 2005; Castlebury et al., 2006; Halleen et al., 2006; Chaverri et al., 2011). Because of the morphological variation, *Neonectria* has been subdivided into five informal groups (Booth, 1959; Brayford & Samuels, 1993; Samuels & Brayford, 1994), and these groups were recognized as five independent genera based on morphological features and multi-gene analysis (Chaverri, 2011). In the latest outline of fungi, thirty species were included in *Neonectria*. Nevertheless, these numbers are underestimated because several species of Nectria-like fungi with *Cylindrocarpon* anamorphs have not been transferred to *Neonectria* (Chaverri, 2011), and a systematic revision work for this genus is need proposed.

Neonectria ditissima (Tul. & C. Tul.) Samuels & Rossman, CBS Diversity Ser. (Utrecht) 4: 134 (2006) Figure 3.17

Saprobic on hard wood. Sexual morph: Ascomata forming superficially on well-developed stroma, aggregated in groups, rarely solitary, 260–387 μm high, 187–259 μm diam., subglobose to globose, glabrous, shiny to slightly roughened, red to dark red, non-papillate but with a slightly darkened region at the apex. Peridium 35–72 μm, composed of two strata, of outlayer comprising thick-walled, pigmented, pseudoparenchymatous cells of *textura globulosa* to *intricta*, inner layer of elongated, thin-walled, subhyaline cells of *textura prismatica*. *Hamathecium* comprising 5–13 μm, septate, branched paraphyses, anastomosing between and above the asci. *Asci* (72–)82–96(–101) × 12.5–16 μm ($\bar{x}$ = 86 × 14.5 μm, n = 20), unitunicate, 8-spored, narrowly clavate, pedicellate, without apical ring. *Ascospores* 13.5–16 × 6.5–7.5 μm ($\bar{x}$ = 15 × 7 μm, n = 35), uniseriate below and biseriate above, ellipsoidal to fusiform with narrowly rounded ends, hyaline, one-septate, slightly constricted at the septum, smooth-walled. Asexual morph: Undetermined.

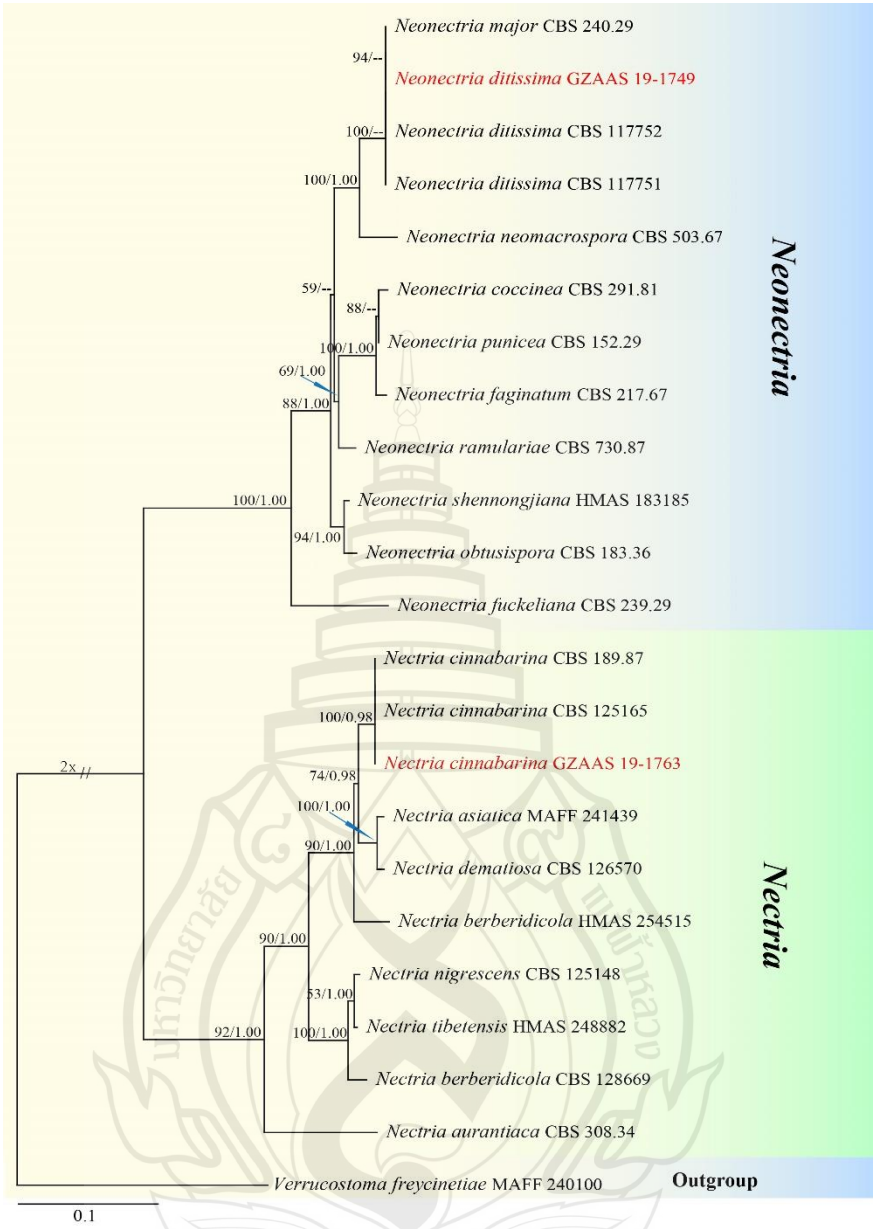
Material examined: CHINA, Guizhou Province, Leishan County, on dead branch of hard wood, 24 May 2019, J.F. Zhang LS-2019-27 (GZAAS 19-1749).

Notes: Phylogenetic analysis in the present study shows that the newly collected specimen clusters with *N. ditissima* and *N. major* by high bootstrap surpport (Figure 3.18). Comparison of the gross morphological characteristics between the new collection and the above related taxa, *N. ditissima* is sutible to fit our new specimen based on the synopsis that provided by Castlebury et al. (2006), and the new collection in this study is therefore identified as *N. ditissima* on the basis of morphological and phylogenetic evidence.



Note a Appearance of ascomata in host surface. b, c Vertical section through ascomata. d Section of peridium. e Paraphyses. f–h Asci. i–n Ascospores. Scale bars: b = 200 µm, c = 50 µm, d, e = 10 µm, f–h = 20 µm, i–n = 5 µm.

Figure 3.17 *Neonectria ditissima* (GZAAS 19-1749)



Note Maximum likelihood bootstrap values equal or greater than 50% and Bayesian posterior probabilities equal or greater than 0.95 are indicated near the nodes. New sequence generated in this study are given in red. The tree is generated from maximum likelihood (ML) analysis based on Act, ITS, LSU, Rpb1, TEF1-α and Tub sequence data, and rooted with *Verrucostoma freycinetiae* MAFF 240100.

Figure 3.18 Phylogenetic tree of the genera *Nectria* and *Neonectira*

Subclass Sordariomycetidae O.E. Erikss & Winka

Chaetosphaeriales Huhndorf et al.

Chaetosphaeriales was established by Huhndorf et al. (2004) to accommodate the family Chaetosphaeriaceae with *Chaetosphaeria* as the typus. Recently, other three families viz. Helminthosphaeriaceae, Leptosporellaceae and Linocarpaceae were recognized and placed within Chaetosphaeriales based on morpho-phylogenetic evidence (Maharachchikumbura et al., 2015; Konta et al., 2017). And this treatment was followed by Wijayawardene et al. (2020) and Hyde et al. (2020c).

Chaetosphaeriaceae Réblová, M.E. Barr & Samuels, Sydowia 51(1): 56 (1999)

Chaetosphaeriaceae was first established to accommodate five genera (*Chaetosphaeria*, *Loramyces*, *Niesslia*, *Rhagadostoma*, and *Zignöella*) but without valid description (Locquin, 1984). And it was therefore considered as a invalid family until Réblová et al. (1999) re-described it and included seven genera viz. *Ascocodinaea*, *Chaetosphaeria*, *Melanochaeta*, *Melanopsammella*, *Porosphaerella*, *Porosphaerellopsis*, and *Striatosphaeria*. Based only on the morphological characteristics, Chaetosphaeriaceae has been assigned within Sordariales by Eriksson et al. (2001, 2003) and Kirk et al. (2001), however, Huhndorf (2004) introduced the order Chaetosphaeriales to accommodate this family based on the phylogenetic evidence. Subsequently, lots of genera were comprised of this family in a series of studies (Maharachchikumbura et al., 2016; Crous et al., 2016, 2018d; Ma et al., 2016; Wijayawardene et al., 2018a; Yang et al., 2018a; Lin et al., 2019b; Zheng et al., 2020; Hyde et al., 2020c; Réblová et al., 2021a, 2021b) and 47 genera are currently included in Chaetosphaeriaceae. In this study, a new genus *Conicumisohaeria* is introduced and accepted in this family based on morpho-phylogenetic analysis.

Conicumisphaeria J.F. Zhang & K.D. Hyde, *gen. nov.*

Etymology: Named based on Latin '*Conicum*' and '*sphaeria*' refers to its conical ascoma in face view.

Type species: *Conicumisphaeria vaginasporum*

Saprobic on decaying wood. Sexual morph: *Ascomata* semi-immersed to erumpent, subglobose to globose, gregarious or scattered, black, ostiolate, non-papillate, fragile in the apex. *Ostiole* central, with a pore-like opening. *Peridium* composed of several layers of pseudoparenchymatous cells of *textura angularis* to

prismatica. *Hamathecium* comprising septate, unbranched, cellular paraphyses, intermixed with the asci. *Asci* 8-spored, unitunicate, cylindric-clavate, pedicellate, apically round with a J- apical apparatus. *Ascospores* overlapping bi- to 4-seriate, hyaline, fusiform, septate, straight, guttulate, surrounded by a thin gelatinous sheath. Asexual morph: Undetermined.

Notes: *Conicumisphaeria* is placed within Chaetosphaeriaceae and formed an independent clade in the present study (Figure 3.21). Morphologically, it corresponds to Chaetosphaeriaceae in having subglobose to globose ascomata, with peridium comprising of pseudoparenchymatous cells of *textura angularis* to *prismatica*, and cylindric-clavate, pedicellate asci that bearing septate, fusiform ascospores (Réblová et al., 1999; Hyde et al., 2020c). However, the strict feature of *Conicumisphaeria* differs from other sexual genera of Chaetosphaeriaceae by its sepetate, fusiform ascospores surrounded by a gelatinous sheath. Based on a megablast search of NCBI's GenBank nucleotide database, the closest hits using the LSU sequence had highest similarity to *Dictyochaeta stratosa* (strain CBS 138739, GenBank MT454505; Identities = 853/894 (95 %), 2 gaps (0%)), *Zanclospora xylophila* (strain ICMP 22737, GenBank MW144417; Identities = 848/895 (95 %), 4 gaps (0%)), *Chaetosphaeria luquillensis* (strain SMH 2973, GenBank AF466074; Identities = 850/893 (95 %), 3 gaps (0%)), *Kionochaeta ivoriensis* (strain CBS 374.76, GenBank MH872758; Identities = 848/895 (95 %), 4 gaps (0%)) and *Nawawia filiformis* (strain MFLU 18-1501, GenBank MH758206; Identities = 845/892 (95 %), 4 gaps (0%)). Closest hits using the ITS sequence had highest similarity to *Dictyochaeta callimorpha* (strain CBS 525.88, GenBank AF178555; Identities = 435/506 (86 %), 27 gaps (5%)), *Pseudolachnea hispidula* (strain MFLUCC 17-1238, GenBank MG764003; Identities = 422/493 (86 %), 23 gaps (4%)) and *Stilbochaeta cangshanensis* (strain GZCC 19-0525, GenBank MW133883; Identities = 412/486 (85 %), 26 gaps (5%)). Hence, *Conicumisphaeria* is introduced hereby as a new genus to accommodate its type species *Conicumisphaeria vaginasporum*.

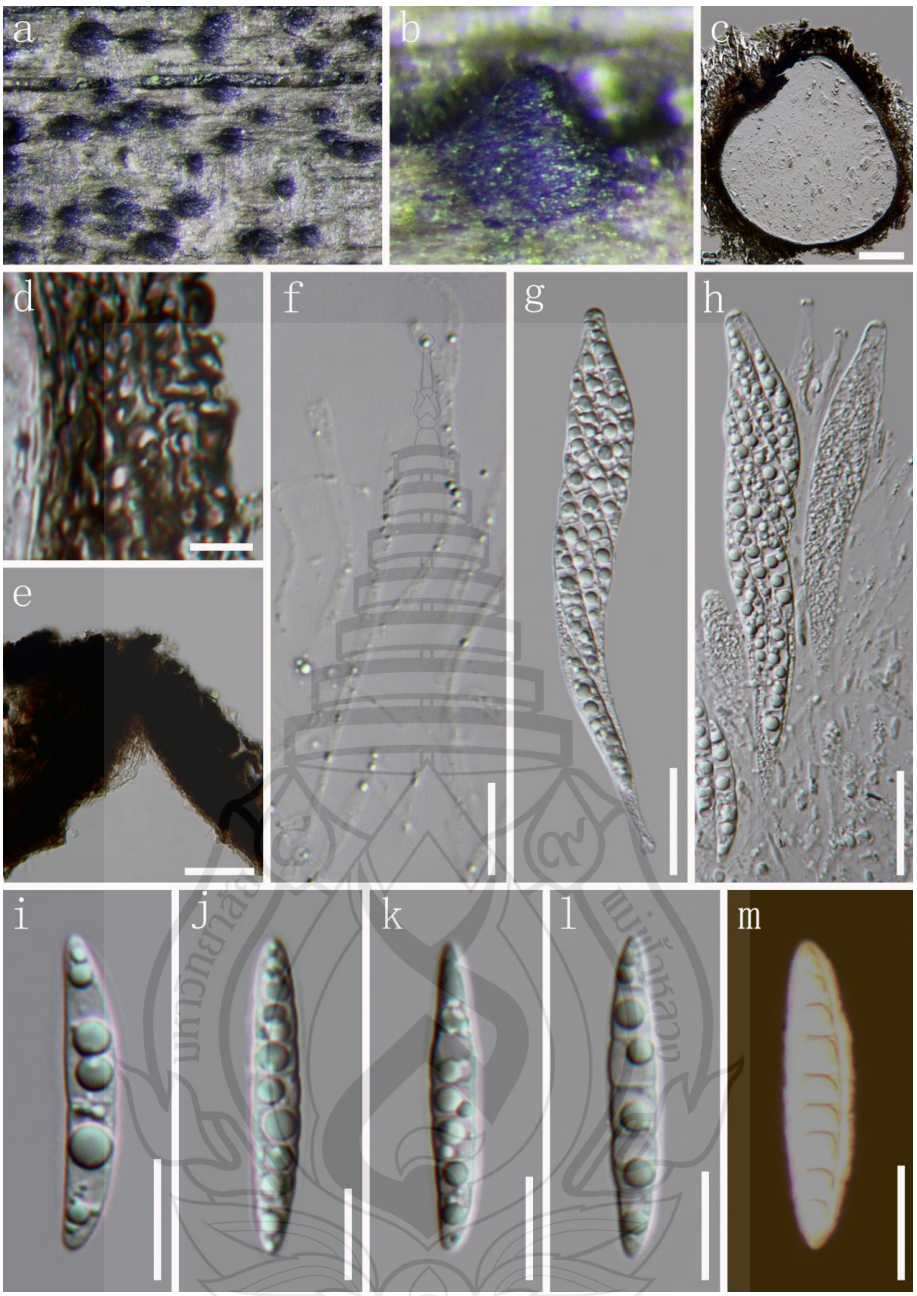
Conicumisphaeria vaginasporum J.F. Zhang & K.D. Hyde, *sp. nov*., Figure 3.19

Etymology: Named based on Latin '*Vagina*' and '*sporum*' refers to its ascospores surround by a gelatinous sheath.

Holotype: MFLU 18-2271.

Saprobic on dead branch of wood plant, forming black, conical spots in substrate. Sexual morph: *Ascomata* semi-immersed to erumpent throughout the epidermis, gregarious or scattered, subglobose to globose, black, ostiolate. *Ostiole* central, periphyses absent. *Peridium* 23–32 µm, composed of two strata, of outer layer with dark brown, thick-walled cells of *textura angularis* to *globosa*, inner layer of subhyaline to hyaline, thin-walled, elongated cells of *textura prismatica*. *Hamathecium* compering 1–2 µm wide, septate, unbranched, cellular paraphyses, intermixed with the asci. *Asci* (93–) 100–105 × 12–14 ($\bar{x}$ = 100 × 13 µm, n = 20) µm, 8-spored, unitunicate, cylindrical-clavate, with a moderate, furcate pedicel, apically round with a J- apical apparatus. *Ascospores* (26–) 28–32 × 3–5 ($\bar{x}$ = 30 × 4 µm, n = 30) µm, overlapping bi-seriate to 4-seriate, hyaline, fusiform, 7-septate at maturity, straight, guttulate, surrounded by a thin gelatinous sheath. Asexual morph: Undetermined.

Material examined: CHINA, Guizhou Province, Libo County, Maolan Town, Maolan National Nature Reserve, on dead branch of woody plant, 18 July 2016, H.Y. Ran MLBB-1 (MFLU 18-2271, holotype).



Note a, b Appearance of ascomata in host surface. c Vertical section through ascoma. d Section of peridium. e Ostiole. f Paraphyses. g, h Asci. i–m Ascospores. Note: m is stained with India ink. Scale bars: c = 100 µm, d, f, i–m = 10 µm, e = 50 µm, g, h = 20 µm.

Figure 3.19 *Conicumisphaeria vaginasporum* (MFLU 18-2271, holotype)

Neoleptosporellaceae J.F. Zhang, Y.Y. Chen & J.K. Liu, fam. nov.

Etymology: Refers to its type genus *Neoleptosporella*.

Type genus: *Neoleptosporella*

Saprobic on stem of herbaceous plants that collected from terrestrial habitat. Sexual morph: *Ascomata* solitary, immersed, brown to black, coriaceous, subglobose to depressed globose, ostiolate, clypeate. *Peridium* composed of dark brown to black cells of *textura angularis* that merging with the host epidermal cells outwardly. *Hamathecium* of numerous, branched, septate, cellular paraphyses. *Asci* unitunicate, broad cylindrical, pedicellate, with a J-, wedge-shaped, subapical ring. *Ascospores* fasciculate, fusiform, C-shaped or sigmoid, aseptate, ends acute, guttulate. Asexual morph: Undetermined.

Notes: Neoleptosporellaceae was established herein to accommodate *Neolepotosporella* species. In the present study, Neoleptosporellaceae is phylogenetically closed to Helminthosphaeriaceae with moderate bootstrap support (Figure 3.21), however, it is easily differentiated from Helminthosphaeriaceae as the latter is characterized by superficial or immersed ascomata that rough, tuberculate, smooth or with setae, and peridium of carbonaceous or membranaceous cells of *textura angularis* or *prismatica*, cylindrical to clavate, pedicellate asci with a truncate apex and J-, apical ring, as well as hyaline or pigmented, allantoid, clavate, cylindrical to ellipsoid ascospores with 0–3-septa, arranged in ascus as 2-seriate.

Neoleptosporella Phukhams. & K.D. Hyde, Fungal Diversity 102: 147 (2020)

Neoleptosporella, typified with *N. clematidis*, was introduced and placed in Chaetosphaeriales genera *incertae sedis* based on morphological and phylogenetic analysis (Phukhamsakda et al; 2020). Then, another species *N. camporesiana* was accommodated in this genus by Hyde et al. (2020a). Members of *Neoleptosporella* are normally characterized by having immersed, subglobose to depressed globose ascomata that beneath a small clypeus, with the peridium of dark brown to black cells of *textura angularis*, hyaline, branched, septate paraphyses, and unitunicate, pedicellate, broadly cylindrical asci that producing fasicculate, aseptate, guttulate, fusiform to filiform ascospores, without any appendages (Hyde et al., 2020a; Phukhamsakda et al., 2020. In this study, a new species *Neoleptosporella fusiformispora* is introduced on the basis of morpho-phylogenetic evidence.

Neoleptosporella fusiformispora J.F. Zhang, Y.Y. Chen & J.K. Liu *sp. nov*., Figure 3.20

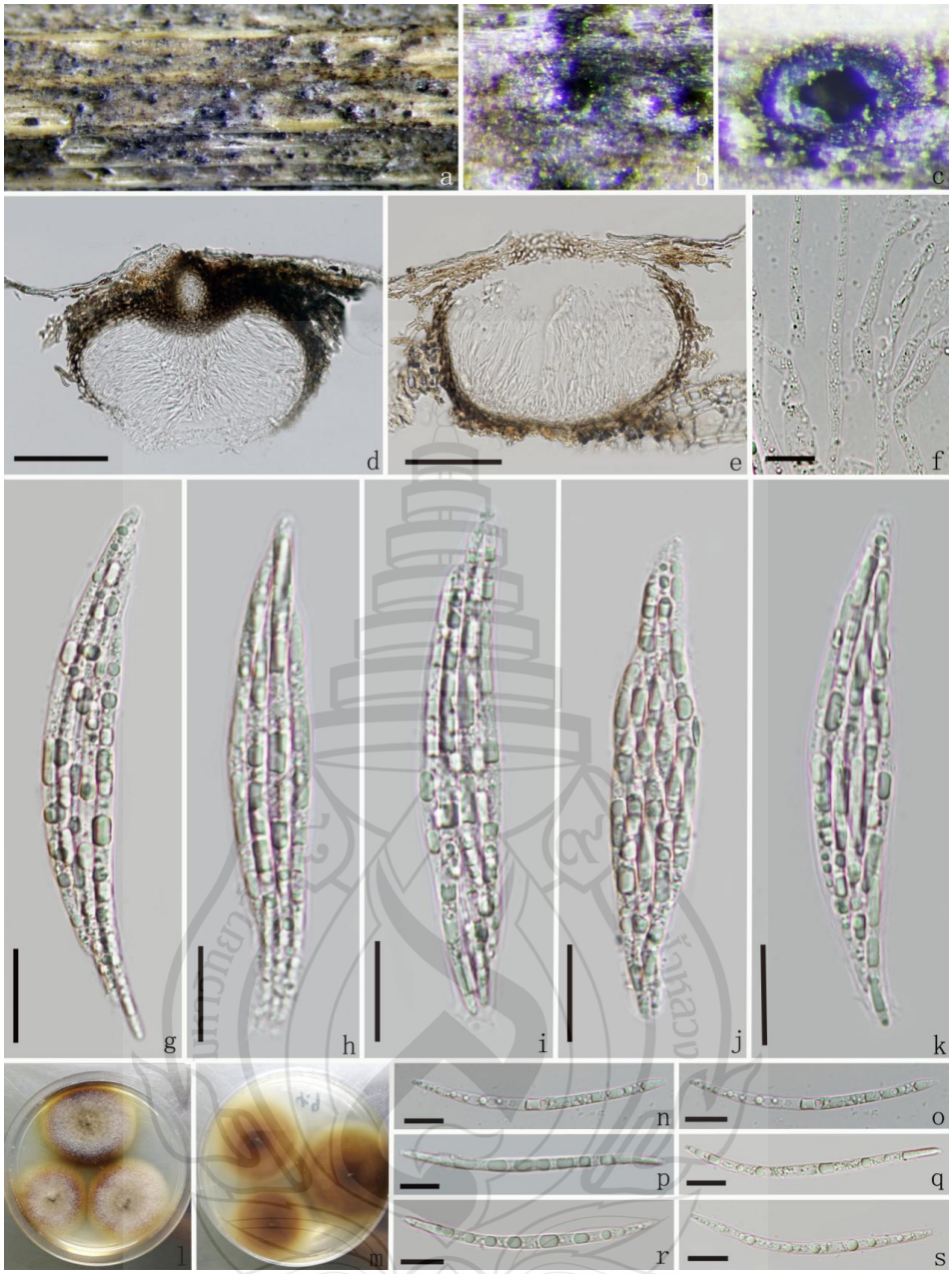
Etymology: Named refers to its fusiform ascospores.

Holotype: GZAAS 20-0253.

Saprobic on dead culm of unidentified herbaceous plant. Sexual morph: *Ascomata* 180–260 μm high, 210–280 μm diam., immersed, subglobose to depressed globose, solitary or gregarious, coriaceous, ostiolate, short papillate. *Ostiole* central, lined with periphyses in the ostiolar canal. *Peridium* 21–32 μm wide, composed of several layes of thick-walled, brwon to dark brown, pseudoparenchymatous cells of *textura angularis* that merging with the host tissues outwardly, becoming subhyaline to hyaline of elongated, thin-walled cells of *textura angularis* inwardly. *Hamathecium* of abundant, 3–4 μm wide, branched, septate paraphyses, intermixed between and above the asci. *Asci* 89–116 × 17–21.5 μm ($\bar{x}$ = 101.5 × 19.5 μm, n = 20), 8-spored, unitunicate, clavate, pedicellate, with a J-, wedge-shaped, subapical ring. *Ascospores* 62–85 × 2.5–3.5 μm ($\bar{x}$ = 71.5 × 3.5 μm, n = 30), fasciculate, narrowly fusiform, C-shaped or sigmoid, aseptate, guttulate, without any appendages or sheath. Asexual morph: Undetermined.

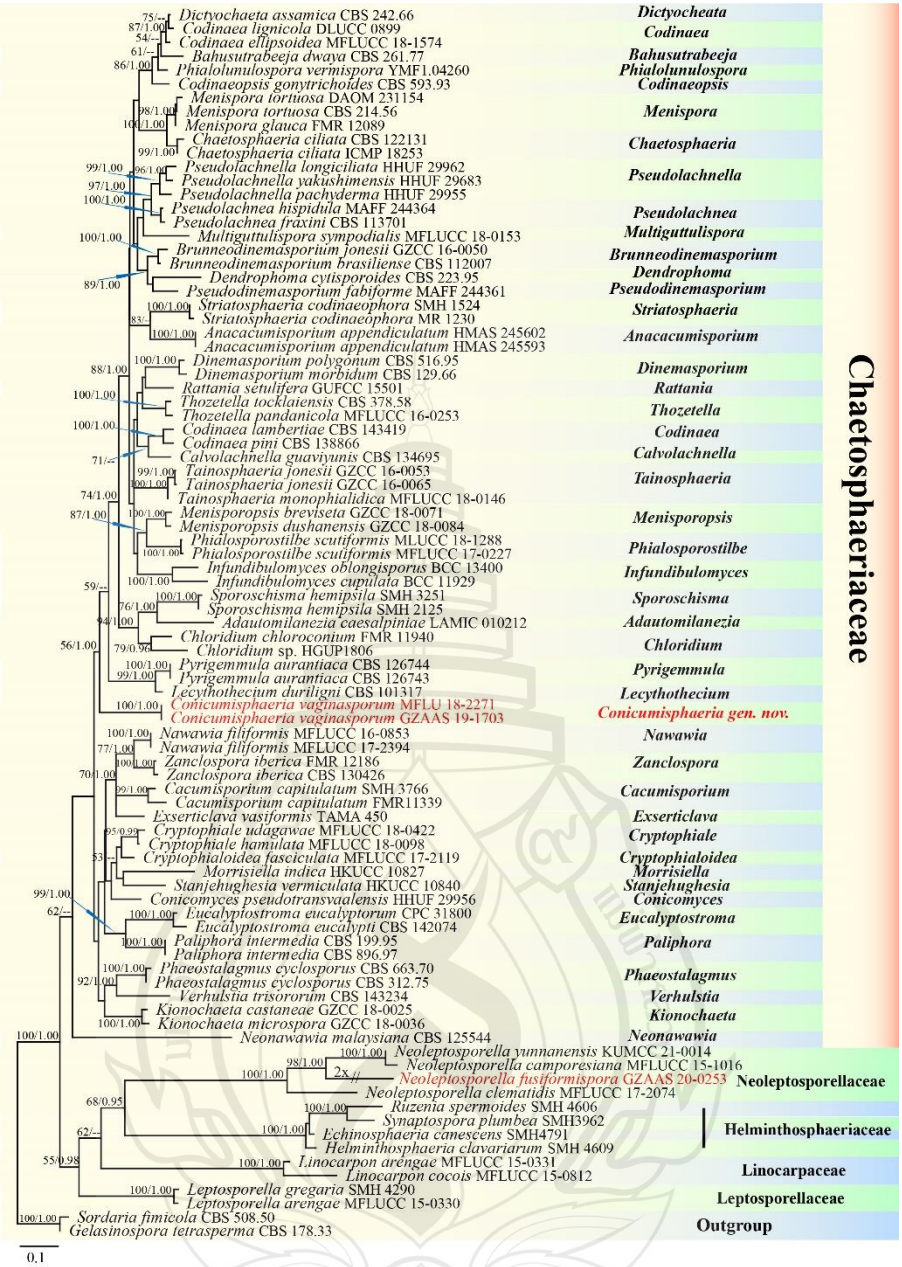
Material examined: CHINA, Guizhou Province, Zunyi City, Suiyang County, on dead culm of unidentified herbaceous plant, 19 May 2018, Y.Y. Chen 20-041 (GZAAS 20-0253, holotype).

Notes: *Neoleptosporella fusiformispora* is phylogenetically placed within Neoleptosporellaceae with high bootstrap support in this study (Figure 3.21). However, it can be distinguished from other reported *Neoleptosporella* species by the measurements of asci and ascospores. The asci and ascospores of *N. fusiformispora* are smaller than *N. camporesiana* but larger than the type species *N. clematidis* (Hyde et al., 2020a; Phukhamsakda et al., 2020).



Note a, b Ascomata on host surface. c Cross section through ascoma. d, e Vertical section through ascoma. f Paraphyses. g–k Asci. l, m Colony on PDA. n–s Ascospores. Scale bars: d, e = 100 µm, f = 10 µm, g–k = 20 µm, n–s = 10 µm.

Figure 3.20 *Neoleptosporella fusiformispora* (GZAAS 20-0253, holotype)



Note Maximum likelihood bootstrap values equal or greater than 50% and Bayesian posterior probabilities equal or greater than 0.95 are indicated near the nodes. New sequence generated in this study are given in red. The tree is generated from Maximum likelihood (ML) analysis based on ITS and LSU sequence data, and rooted with *Sordaria fimicola* CBS 508.50 and *Gelasinospora tetrasperma* CBS 178.33.

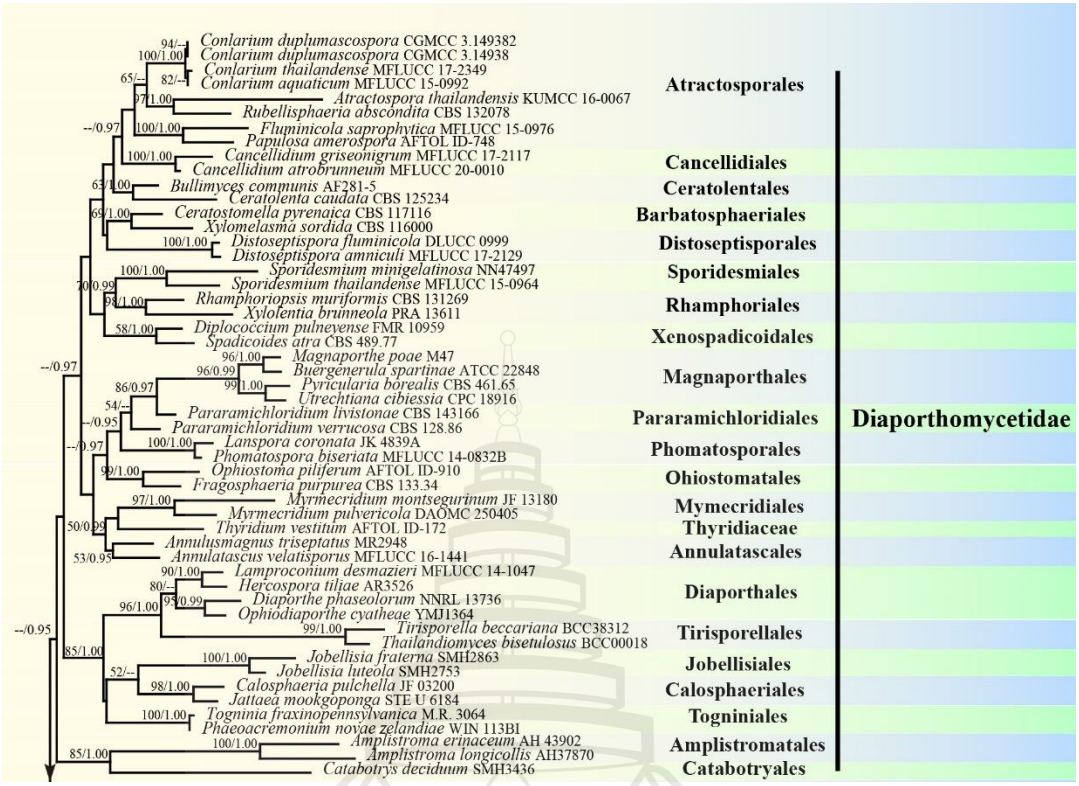
Figure 3.21 Phylogenetic tree of the order Chaetosphaeriales

Planisphaeriales J.F. Zhang, J.K. Liu & K.D. Hyde, *ord. nov.*

Type family: Planisphaeriaceae J.F. Zhang, J.K. Liu & K.D. Hyde.

Saprobic on dead woody plant from terrestrical habitats. Sexual morph: *Ascomata* perithecial, immersed to erumpent through the host tissue, with broadly crack at the apex, gregarious or scattered, depressed subglobose to obpyriform, with a flattened base, coriaceous, brown to black, clypeate, paraphysate. *Peridium* composed of several layers of subhyaline to hyaline, thin-walled, elongated cells of *textura angularis* merging with red-brown pseudoparenchymatous cells. *Paraphyses* dissolved or persistent at age. *Asci* unitunicate, clavate, pedicellate, with or without apical rings. *Ascospores* hyaline, aseptate, reniform or narrowly ovoid, rarely in irregular shapes, straight or curved, with or without mucilaginous sheath. Asexual morph: Undetermined.

Notes: Multi-gene phylogenetic analysis in the present study shows that our new collections form a distinct clade that sister to Phyllachorales, and where they are closely related to Boliniales (Figure 3.22). This topology is somewhat similar to recent studies (Maharachchikumbura et al., 2016; Hongsanan et al., 2017; Hyde et al., 2020c). Morphologically, Phyllachorales resembles to some members of Phyllachorales and Boliniales in having clavate, pedicellate asci and hyaline, aseptate ascospores, nevertheless, it is significantly differentiated from these above orders by its non-stromatic ascomata with a broadly crack at the apex, and the reniform or narrowly ovoid, hyaline, aspetate ascospores (Maharachchikumbura et al., 2016; Hyde et al., 2020c; Huang et al., 2021). Moreover, the results of megablast search in NCBI's GenBank nucleotide database for closest hits of LSU, SSU, TEF and Rpb2 sequences show that these two newly collected specimens related to different orders in the subclass Sordariomycetidae. Therefore, Planisphaeriales is introduced as a new order to accommodate its type family Planisphaeriaceae.



Note Maximum likelihood bootstrap values equal or greater than 50% and Bayesian posterior probabilities equal or greater than 0.95 are indicated near the nodes. New sequence generated in this study are given in red. The tree is generated from maximum likelihood (ML) analysis based on LSU, SSU, TEF1-α and Rpb2 sequence data, and rooted with *Dothidea sambuci* DAOM 231303 and *Botyrotinia fuckeliana* AFTOL ID-59.

Figure 3.22 Phylogenetic tree of the class Sordariomycetes

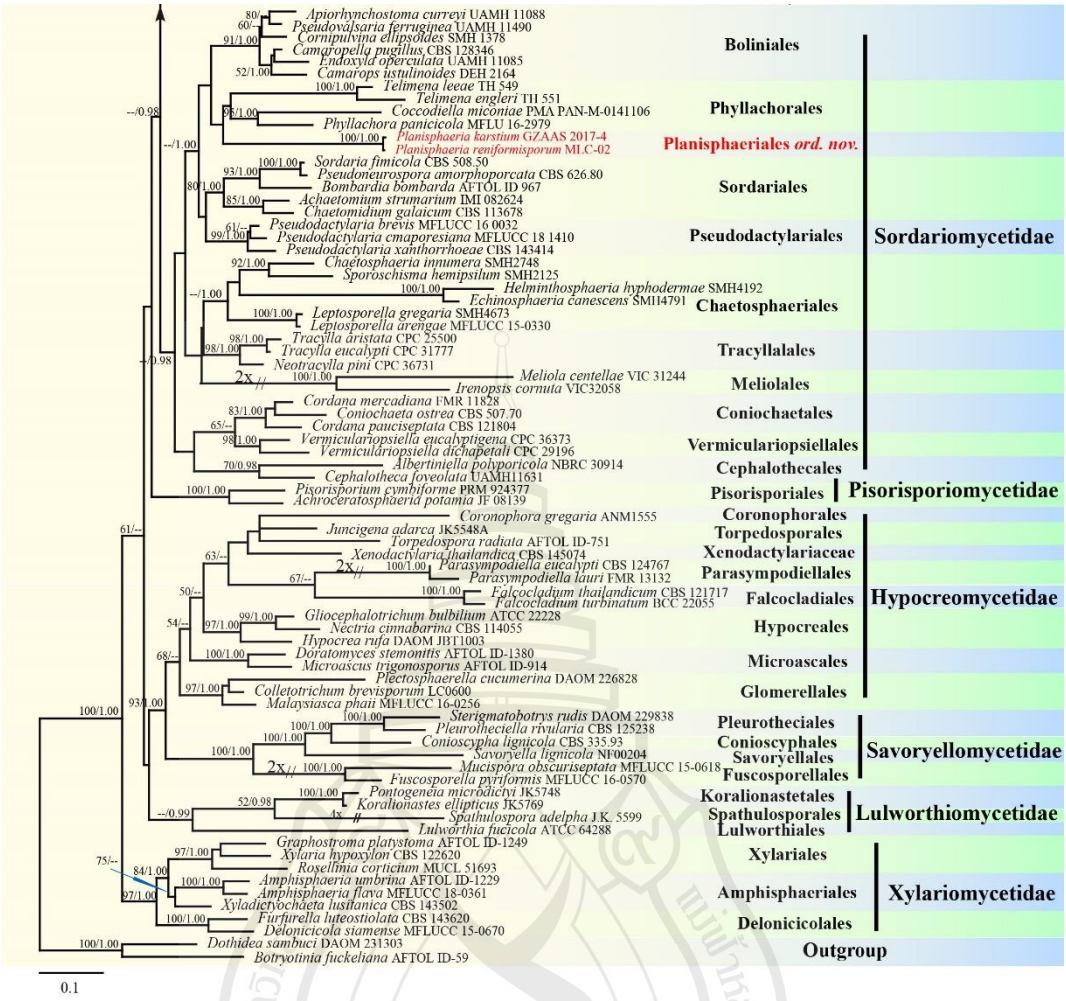


Figure 3.22 (continued)

Planisphaeriaceae J.F. Zhang J.K. Liu & K.D. Hyde, *fam. nov.*

Type genus: *Planisphaeria* J.F. Zhang, J.K. Liu & K.D. Hyde.

Saprobic on dead woody plant. Sexual morph: *Ascomata* perithecial, immersed to erumpent through the host tissue, broadly crack at the apex, gregarious or scattered, depressed subglobose to obpyriform, flattened at the base, coriaceous, dark brown to black, ostiolate, clypeate. *Ostiole* central, paraphysate. *Peridium* composed of two strata, of outlayer comprising thick-walled, red-brown pseudoparenchymatous cells merging with host tissue, towards subhyaline to hyaline, thin-walled, elongated cells of *textura angularis* inwardly. *Paraphyses* hypha-like, persistant or absent at age, slightly longer than asci. *Asci* unitunicate, cylindric-clavate,

short pedicellate, with or without apical ring. *Ascospores* hyaline, aseptate, reniform, narrowly ovoid, rarely irregular shapes, thick-walled, straight or curved, with or without mucilaginous sheath. Asexual morph: Undetermined.

Notes: Planisphaeriaceae is introduced hereby as a monotypic family to accommodate its type genus *Planisphaeria.*

Planisphaeria J.F. Zhang J.K. Liu & K.D. Hyde, *gen. nov.*

Etymology: Named refers to its ascomata with a flattened base.

Type species: *Planisphaeria reniformisporum*

Saprobic on dead woody plant. Sexual morph: *Ascomata* perithecial, immersed to erumpent through the host tissue, broadly crack at the apex, gregarious or scattered, depressed subglobose to obpyriform, with a flattened base, coriaceous, dark brown to black, sometimes stained the substrate in brown to dark brown, ostiolate, clypeate. *Ostiole* central, lined with abundant, hypha-like, non-septate paraphysis in ostiolar canal. *Peridium* composed of two strata, of outer stratum comprising thick-walled, red-brown to brown pseudoparenchymatous cells merging with host tissue, inner stratum of hyaline, thin-walled, elongated cells of *textura angularis* to *prismatica*. *Hamathecium* of filiform paraphyses, slightly longer than asci, dissovling or persistant at age. *Asci* unitunicate, 8-spored, broadly clavate, short pedicellate, apically round with or without apical rings. *Ascospores* overlaping bi- to tri-seriate, hyaline, aseptate, reniform or narrowly ovoid, rarely in irregular shapes, thick-walled, with or without mucilaginous sheath. Asexual morph: Undetermined.

Notes: *Planisphaeria* is introduced to accommodate two novel species *P. karstium* and *P. reniformisporum* with the latter as the type species. Even these two taxa form an independent clade within subclass Sordariomycetidae and warrant the family Planisphaeriaceae and order Planisphaeriales, more fresh collections and molecular sequence data are needed to stabilize their placement.

Planisphaeria reniformisporum J.F. Zhang & K.D. Hyde, *sp. nov.*, Figure 3.23

Etymology: Named refers to its reniform ascospores.

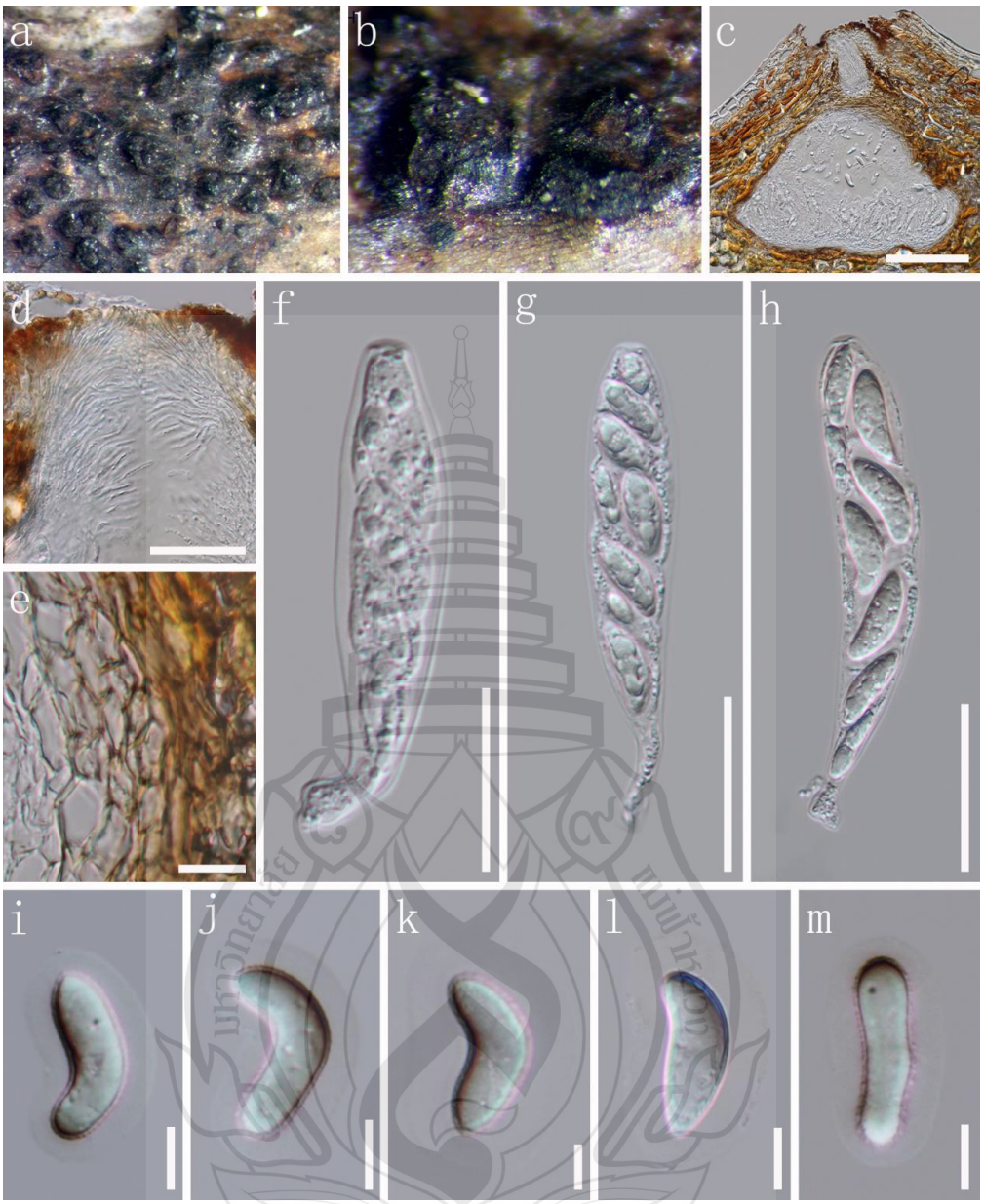
Holotype: GZAAS 20-4004

Saprobic on dead branch of unidentified woody plant, appearing as raising, rough, black spots. Sexual morph: *Ascomata* 281–312 μm high, 293–342 μm diam., immersed to erumpent through the host tissue, stain the host in reddish brown to dark

brown, gregarious or scattered, uni-loculate, depressed subglobose to obpyriform, with a flattened base, dark brown to black, coriaceous, ostiolate, broadly crack at the apex. *Ostiole* central, with abundant, hypha-like paraphysis lined in ostiolar canal. *Peridium* 11–28 μm wide, composed of two stratum, inner stratum of subhyaline to hyaline cells of *textura angularis*, outer stratum of thick-walled, red-brown to brown cells of Pseudoparenchymatous, merging with host tissue, and become thick towards the apex. *Hamathecium* only comprising asci, paraphyses not observed. *Asci* (89–)93.5–111(–123) × (15–)17–22 μm ($\bar{x}$ = 101 × 18.5 μm, n = 20), 8-spored, unitunicate, clavate, with a short, bulbose pedicel, rounded to truncate at the apex, with an inconspicuous apical ring. *Ascospores* 19–23 × 6–8 μm ($\bar{x}$ = 20 × 7.2 μm, n = 30), overlapping bi-seriate, hyaline, mostly reniform, rarely irregular shapes, tapering towards the ends, one-celled, thick-walled, surrounded by a hyaline, 3–6 μm wide, mucilaginous sheath. Asexual morph: Undetermined.

Material examined: CHINA, Guizhou Province, Libo County, Maolan Town, on dead branch of unidentified woody plant, 8 July 2015, J.F. Zhang MLC-02 (GZAAS 20-4004, holotype).

Notes: *Planisphaeria reniformisporum* clusters with *P. karstium* with full bootstrap support in this study (Figure 3.22). Morphologically, they are similar in having uni-loculate ascomata with a flattened base and broadly crack at the apex, similar texture of peridium, unitunicate, 8-spored asci and hyaline, aseptate ascospores. However, they can be easily distinguished by the shape and ornamentation of ascospores. Ascospores of *P. karstium* are normally in irregular ellipsoid, acute towards one end and rounded at another end, guttulate, verrucose, while *P. reniformisporum* is observed in having reniform asocspores that surround a thick mucilaginous sheath.



Note a, b Appearance of ascomata on host surface in face view. c Vertical section through ascomata. d Ostiole with periphyses. e Section of peridium. f–h Asci. i–m Ascospores. Scale bars: c = 100 µm, d, f–h = 20 µm, e = 10 µm, i–m = 5 µm.

Figure 3.23 *Planisphaeria reniformisporum* (GZAAS 20-4004, holotype)

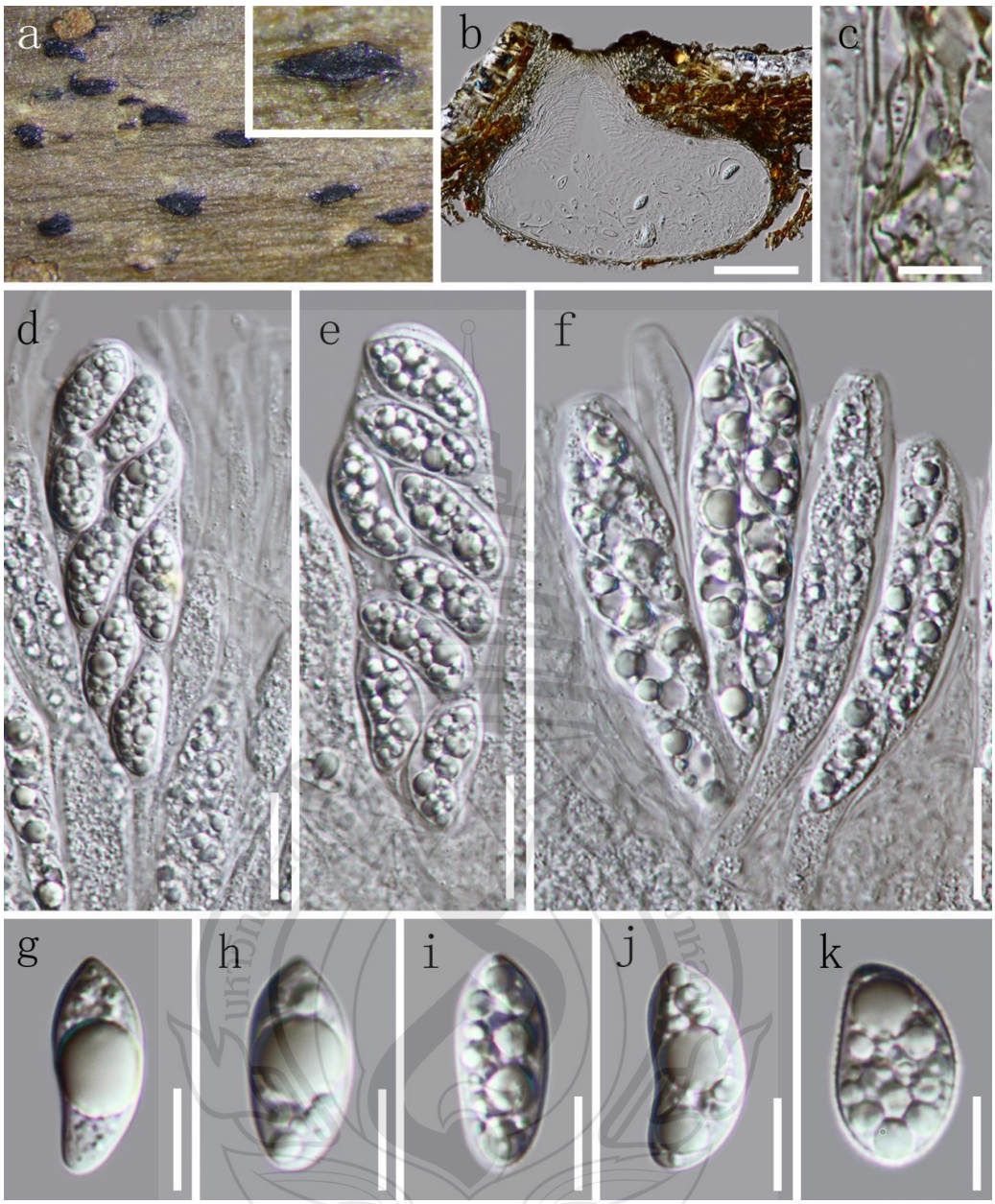
Planisphaeria karstium J.F. Zhang & K.D. Hyde, *sp. nov*., Figure 3.24

Etymology: Named refers to karst formations where the holotype was collected.

Holotype: GZAAS 20-4008.

Saprobic on dead branch of woody plant Sexual morph: *Ascomata* immersed to erumpent through the host tissue, solitary, scattered, depressed subglobose to globose, flattened as the base, black, coriaceous, broadly crack and leathered to carbonaceous as the apex. *Ostiole* paraphysate. *Peridium* 13–20 μm wide, composed of two strata, of inner stratum of several layers of thin-walled, hyaline cells of *textura prismatica*, outer stratum of reddish brown to brown pseudoparenchymatous that merging with host tissue. *Hamathecium* comprising abundant, 2–4.5 μm wide, hyphae-like paraphyses, anastomosed, slightly longer than asci. *Asci* (80–)87.5–95(–103) × 18–24(–27) μm ($\overline{x}$ = 92 × 22 μm, n = 20), 8-spored, unitunicate, broadly clavate, short pedicellate, rounded to truncate at the apex, with an inconspicuous apical ring. *Ascospores* 20.5–24 × 8.5–12 μm ($\overline{x}$ = 22.5 × 10.5 μm, n = 30), overlapping bi- to tri-seriate, hyaline, irregular ellipsoid, normally tapered towards one end and rounded at another end, guttulate, verrucose, aseptate, without mucilaginous sheath or appendages. Asexual morph: Undetermined.

Material examined: CHINA, Guizhou Province, Guiyang City, Huaxi District, Guizhou Academy of Agricultural Sciences, on dead branch of woody plant, 4 November 2017, J.F. Zhang GZAAS-2017-4 (GZAAS 20-4008, holotype).



Note a Appearance of ascomata on host surface in face view. b Vertical section through ascoma. c Section of peridium. d Paraphyses anastomosing between the asci. e, f Asci. g–k Ascospores. Scale bars: b = 100 µm, c, g–k = 10 µm, d–f = 30 µm.

Figure 3.24 *Planisphaeria karstium* (GZAAS 20-4008, holotype)

Subclass Xylariomycetidae O.E. Erikss. & Winka

Xylariales Nannf.

Xylariales is a large order, which was established by Nannfeldt (1932) with *Xylaria* as the type genus. Depending on morphological characteristics, the number of families within Xylariales has been increased to 11 in various studies (Munk, 1953; Müller & von Arx, 1962, 1973; Wehmeyer, 1975; Barr, 1990a; Hawksworth et al., 1995; Eriksson et al., 2001, 2003) Recently, Maharachchikumbura et al. (2016) assigned 22 families within Xylariales, and indicated that it is the only order in the subclass Xylariomycetidae. However, Wijayawardene et al. (2018) reduced these to 12 families in this order. Hyde et al. (2020c) accepted 15 families in Xylariales, but did not include the family Leptosilliaceae that introduced by Voglmayr et al. (2019). Then, other two families Fasciatisporaceae and Vamsapriyaceae were introduced and accommodated in Xylariales (Hyde et al., 2020c; Sun et al., 2021).

Cainiaceae J.C. Krug

Cainiaceae was introduced by Krug (1978) to accommodate *Cainia*, which was segregated from Amphisphaeriaceae, based on its unique apical apparatures and ascospores characteristics. Kang et al. (1999) reviewed Cainiaceae and accpeted 6 genera (*Arecophila*, *Atrotorquata*, *Cainia*, *Ceriophora*, *Reticulosphaeria* and *Ommatomyces*) with available descriptions and illustrations. And subsequently, a different number of genera were accommodated within this family by a series of studies (Jeewon et al. 2003; Smith et al. 2003; Liu et al. 2015; Maharachchikumbura et al. 2015, 2016; Senanayake et al. 2015; Mapook et al. 2020a). In the latest outline of fungi, there are 10 genera *viz*. *Alishanica*, *Amphibambusa*, *Arecophila*, *Atrotorquata*, *Burrowsia*, *Cainia*, *Endocalyx*, *Longiappendispora*, *Seynesia* and *Vesiculozygosporium* were accepted within in this family (Wijayawardene et al. 2022), however, the genus *Alishanica* was synonymized under *Arecophila* that introduced by Hyde (1996), based on morphological characteristics and phylogenetic affinity (Li et al. 2022).

Arecophila K.D. Hyde

Arecophila was introduced by Hyde (1996) with five species (*A. calamicola*, *A. egueissonae*, *A. gulubiicola*, *A. notabilis* and *A. nypae*) and two combinations (*A. chamaeropis* and *A. serrulata*), and accommodated in Amphisphaeriaceae. However,

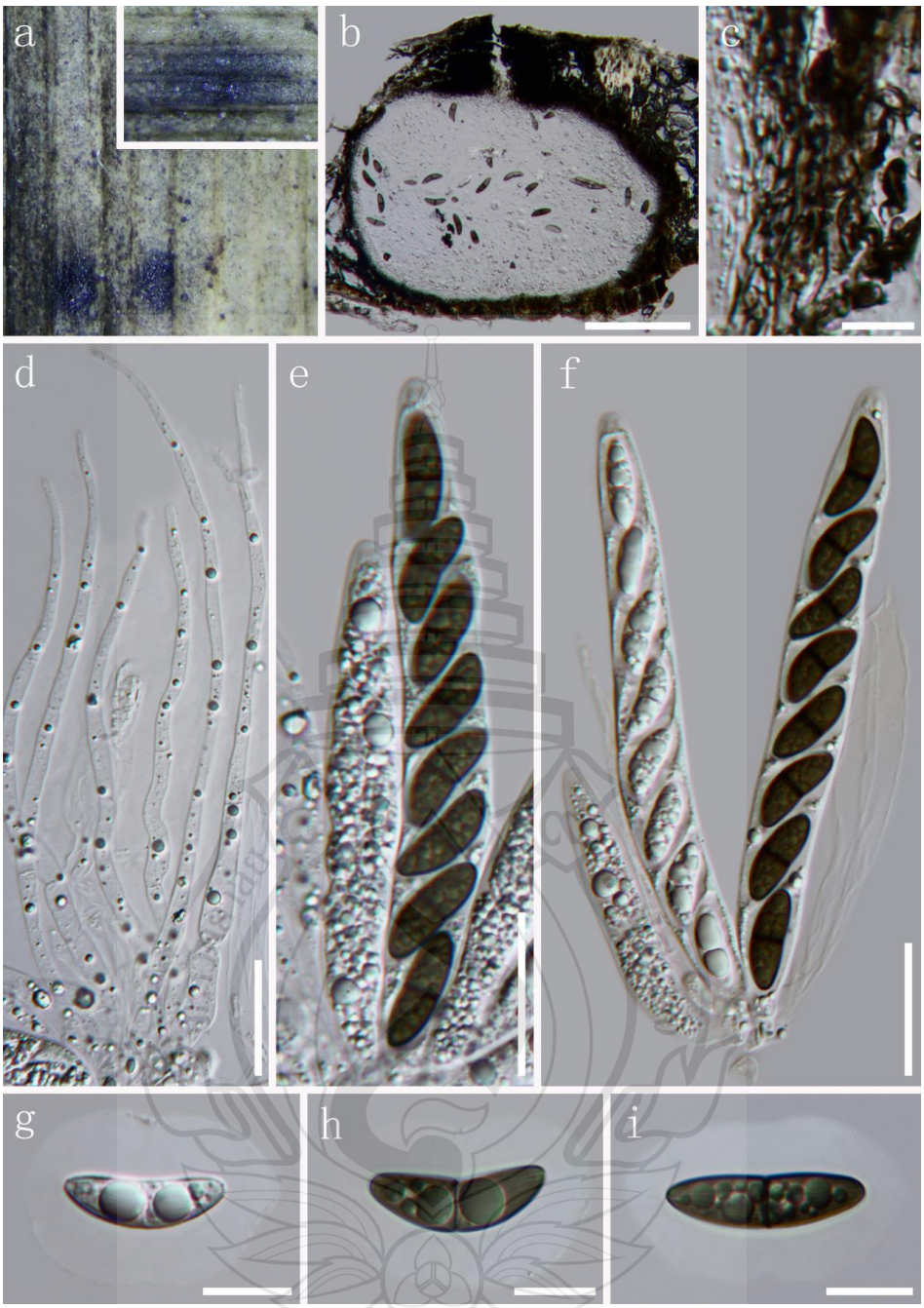
Kang et al. (1999) revived the family Cainiaceae and included *Arecophila* based on typical morphological characteristics. Members of *Arecophila* are characterized by immersed ascomata under a non- or poorly-developed clypeus, long cylindrical to cylindric-clavate asci with a wedge-shaped, J+ apical ring, and one septate, brown ascospores with longitudinal striations, and surrounded by a mucilaginous sheath (Hyde 1996; Kang et al. 1999). Until now, eighteen epithets were listed in Index Fungorum (July 2022), but only five species with available sequence data in GenBank. And the synopsis of *Arecophila* was provided by Li et al. (2022). In this study, two arecophila-like samples were collected and identified as two reported species *A. clypeata* and *A. miscanthi*, based on morpho-molecular analysis.

Arecophila clypeata Q.R. Li, J.C. Kang & K.D. Hyde, in Li et al., MycoKeys 88: 135 (2022), Figure 3.25

Saprobic on dead herbaceous culms. Sexual morph: *Ascomata* immersed beneath blackened aggregated clypeus of the host surface, solitary, depressed globose to subglobose, black, coriaceous, short papillate, ostiolate. *Ostiole* central, fragile, surrounded by obvious shiny black flanges the tissue spreads down along the papilla. *Peridium* laterally 15–35 μm wide, becoming thick around the ostiole, comprising several layers of subhyaline to hyaline, thin-walled, cells of *textura angularis* inwardly, and dark brown, thick-walled cells of pseudoparenchymatous confusing with host tissue. *Hamathecium* of numerous, 2–5.5 μm wide, filamentous, trabeculate paraphyses, tapered towards the end, intermixed between and above the asci. *Asci* 127.5–150 × 13–16 μm ($\overline{x}$ = 141 × 14.5 μm, n = 15), 8-spored, unitunicate cylindrical to subclavate, short pedicellate, rounded at the apex, apical apparatus distinct. *Ascospores* 21.5–24 × 6.5–8.5 μm ($\overline{x}$ = 23.5 × 7 μm, n = 25), overlapping bi-seriate, ellipsoidal, tapering towards both ends, slightly cureved, 1-septate, not constricted at the septum, guttulate, hyaline when young, becoming dark brown at maturity, with 8–11 longitudinal striations superficially resembling ridges, surrounded by a 8–14 μm wide gelatinous sheath. Asexual morph: Undetermined.

Material examined: CHINA, Guizhou Province, Libo County, Maolan Town, Maolan National Nature Reserve, on dead herbaceous culms, 20 July 2016, J.F. Zhang MLCC-6 (MFLU 18-2280).

Notes: Our new collection (MFLU 18-2280) was placed within Cainiaceae in the present study, and where it clusters with the specimens of *Arecophila clypeata* with full bootstrap support (Figure 3.27). Morphologically, the new specimen is similar to *A. clypeata* in having subglobose to globose ascomata under a clypus, similar wide and textura of peridium, cylindrical, short pedicellate asci, and same dimensions of brown, two-celled, equilateral ascospores that surround by a thick mucilaginous sheath (Li et al. 2022). However, the length of asci between these two specimens are different, and this maybe attributed that the asci of *A. clypeata* presented by Li et al. (2022) are immature and deformed. In addition, each gene regions between our new sample and *A. clypeata* were compared based on the recommendations from Jeewon and Hyde (2016), and the results showed that there are no difference in LSU and ITS, respectively. Therefore, the new sample collected in this study is identified as *A. clypeata*, and which has been found in Guizhou and Yunnan Province of China (Li et al. 2022).



Note a Appearance of ascomata on host surface. b Vertical section through ascoma. c Section of peridium. d Paraphyses. e, f Young and mature asci. g–i Immature and mature ascospores. Scale bars: b, c = 200 μm, d–f = 50 μm, g–i = 20 μm.

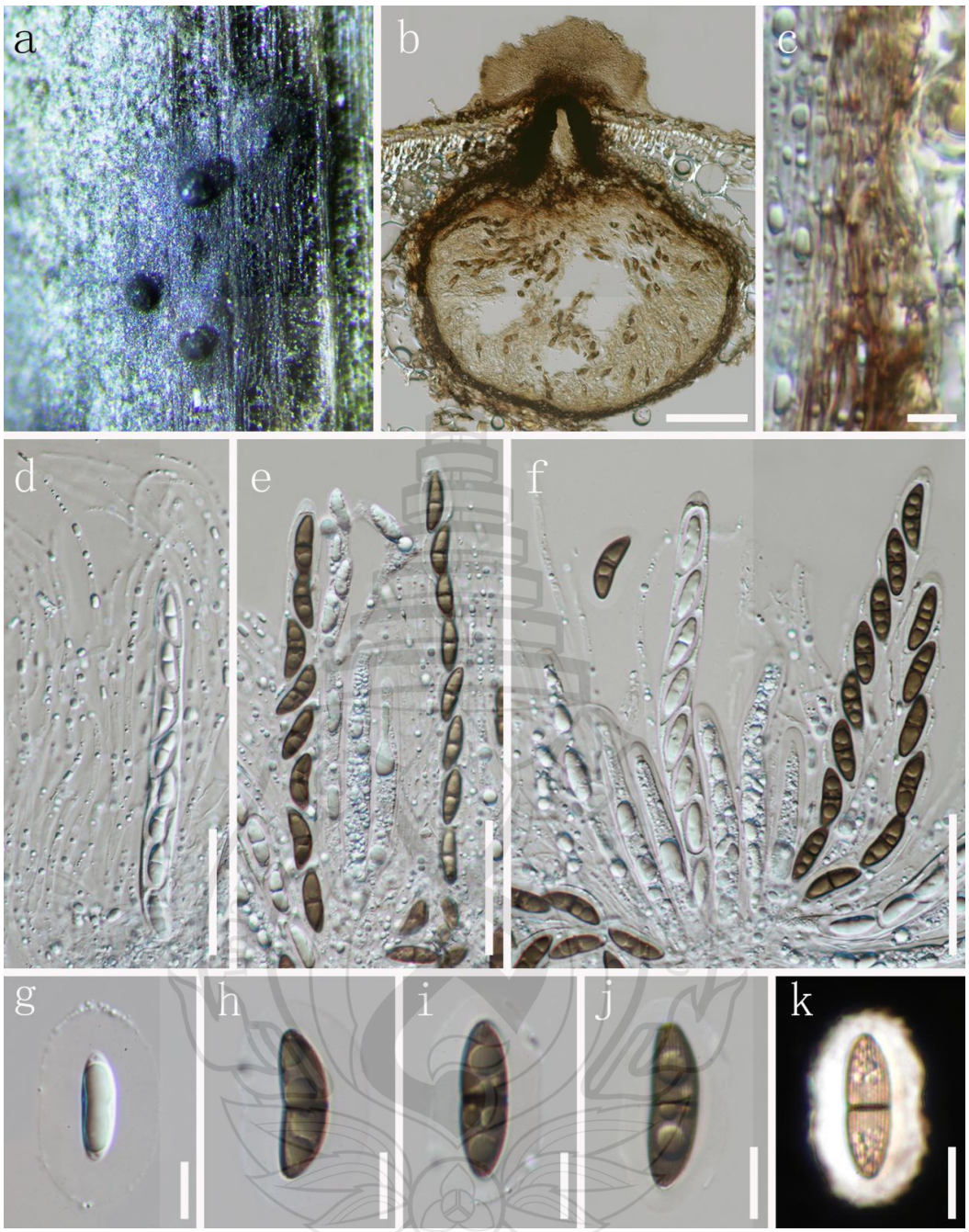
Figure 3.25 *Arecophila clypeata* (MFLU 18-2280)

Arecophila miscanthi (Karun., C.H. Kuo & K.D. Hyde) Q.R Li & J.C. Kang, in Li et al., MycoKeys 88: 139 (2022), Figure 3.26

Saprobic on dead culm of unidentified herbaceous plant. Sexual morph: *Ascomata* immersed beneath blackened aggregated clypeus of the host tissue, loosely aggregated or sometimes solitary, dark brown to black, subglobose to globose, uniloculate, coriaceous, glabrous, ostiolate, papillate. *Ostiole* central, with periphysate, surrounded by obvious shiny black flanges the tissue spreads down along the papilla. *Peridium* 13–33 μm wide, comprising several layers of thin-walled cells of *textura angularis* to *prismatica*, hyaline to subhyaline inwardly, towards brown to dark brown outwardly, and becoming thick and darker around the ostiole. *Hamathecium* of abundant, 2–4 μm wide, filamentous, non-septate, paraphyses, intermixed between and above the asci. *Asci* (133–)160–172(–175) × (10.5–)12–13.5(–14) μm ($\overline{x}$ = 165 × 12.5 μm, n = 20), 8-spored, unitunicate, cylindrical, short pedicellate, narrowly rounded at the apex, apical apparatus distinct. *Ascospores* 20–23(–25) × 6–7(–8) μm ($\overline{x}$ = 22.3 × 6.8 μm, n = 35), overlapping uni-seriate, ellipsoidal, tapering towards the ends, equally 2-celled and guttulate at both cells, slightly constricted at the septum, hyaline and aseptate when young, becoming brown at maturity, often with 10 longitudinal striations superficially, surrounded by a 4–7 μm thick gelatinous sheath. Asexual morph: Undetermined.

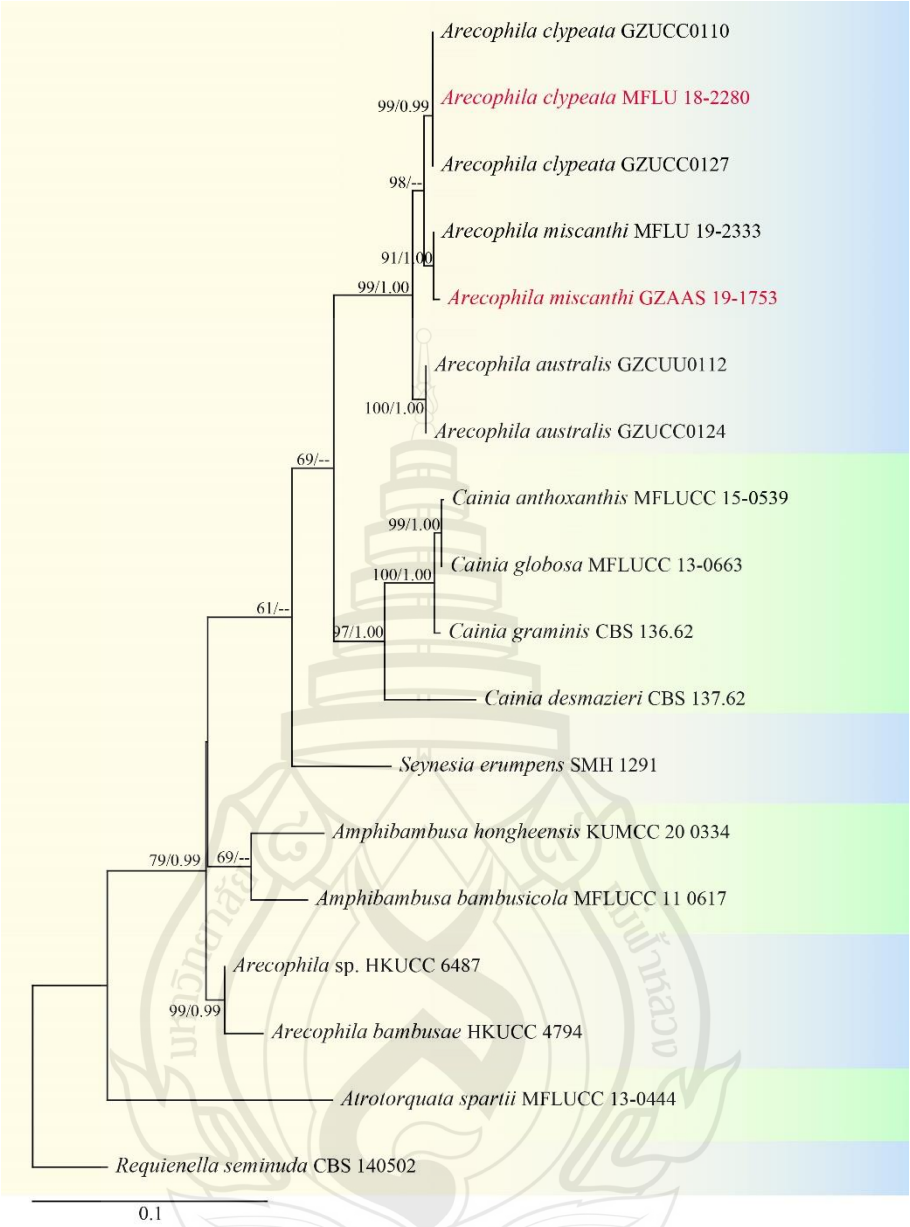
Material examined: CHINA, Guizhou Province, Sinan County, on dead culm of herbaceous plant, 8 July 2019, H.Y. Ran SN-2019-12 (GZAAS 19-1753).

Notes: Present phylogenetic analysis shows that our new collection (GZAAS 19-1753) clusters to *Arecophila miscanthi* in the present study (Fig. 3.27). Examination of the gross morphological characteristics of this specimen, we found it well fits with *A. miscanthi* that described by Hyde et al. (2020) and Li et al. (2022), including the overlap sized of asci and ascospores. Hence, this new collection was identified as A. miscanthi, and this species have been reported from Taiwan and Yunnan Province of China (Hyde et al. 2020; Li et al. 2022).



Note a Appearance of ascomata in substrate. b Vertical section through ascoma. c Section of peridium. d Paraphyses. e, f Young and mature asci. g–k Immature and mature ascospores. Note: the k is stained with India ink. Scale bars: b = 100 μm, c = 5 μm, d–f = 20 μm, g–k = 10 μm.

Figure 3.26 *Arecophila miscanthi* (GZAAS 19-1753)



Note Maximum likelihood bootstrap values equal or greater than 50% and Bayesian posterior probabilities equal or greater than 0.95 are indicated near the nodes. New sequence generated in this study are given in red. The tree is generated from maximum likelihood (ML) analysis based on LSU and ITS sequence data, and rooted with Requienella seminuda CBS 140502.

Figure 3.27 Phylogenetic tree for the family Cainiaceae

Diatrypaceae Nitschke

Diatrypaceae was introduced by Nitschke (1869) and is typified by *Diatrype*. Members of this family were well-studied, especially in recent phylogenetic studies. Such as Senwanna et al. (2017) and Phookamsak et al. (2019) provided backbone trees with sequence data, as well as the detailed taxonomic reviews for the family were proposed by Trouillas et al. (2011) and Mehrabi et al. (2015). Maharachchikumbura et al. (2015) included 15 genera within Diatrypaceae, and subsequently, several genera such as *Allocryptovalsa*, *Halodiatrype* and *Neoeutypella* were discovered (Dayarathne et al., 2016; Senwanna et al., 2017; Phookamsak et al., 2019), and 20 genera were accepted in this family (Hyde et al. 2020). Subsequently, other three genera viz. *Allodiatrype*, *Paraeutypella* and *Pseudodiatrype* were added within Diatrypaceae (Konta et al., 2020; Dissanayake et al., 2021; Long et al., 2021), and these above 23 genera were recognized by Zhu et al. (2021).

Diatrypella (Ces. & De Not.) De Not., Sfer. Ital.: 29 (1863)

Diatrypella was erected to accommodate the fungi which were characterized by ovoid and unlimited number of ascospores (Cesati & De Notaris, 1863). It differs from the close relative genus *Diatrype* by having polysporous asci (Croxall, 1950). There are more than 150 epithets listed in Index Fungorum (2022), and approximately 115 species accepted by Wijayawardene et al. (2022) in the latest outline of fungi. However, only a few taxa of these above species with available sequence data in GenBank, and this will definitely bring great challenges to those mycologists who study this group. Therefore, fresh samples are needed to delimit the species boundaries of *Diatrypella* in the further study.

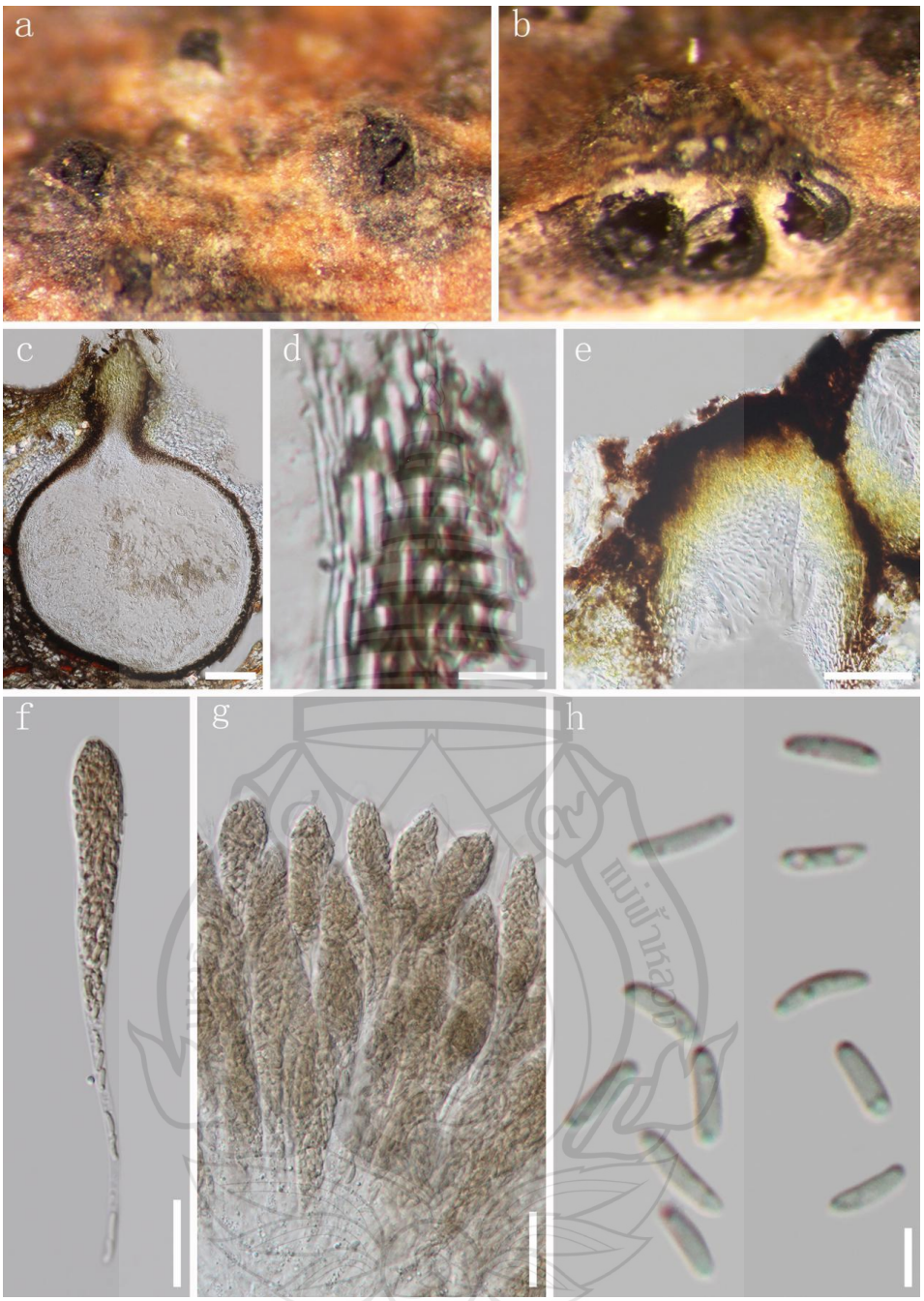
Diatrypella vulgaris Trouillas, W.M. Pitt & Gubler, Fungal Diversity 49: 212 (2011). Figure 3.28

Saprobic on decaying wood. Sexual morph: *Stromata* well developed, in pustules, solitary or gregarious on host, semi-immersed, erumpent through host epidermis, with 3–6 ascomata immersed in a single stroma. *Ascomata* perithecial, immersed in stromatic tissues, aggregated, globose to subglobose, coriaceous, pale to dark brown, ostioles sulcate, slightly prominent, paraphysate, carbonaceous or fragile at the apex. *Peridium* 15–33 μm, composed of two stratum, inner stratum of hyaline, thin-walled, elongated cells of *textura prismatica*, outer stratum of brown, thick-

walled cells of *textura angularis*. *Hamathecium* comprising abundant asci, paraphyses not observed. *Asci* 130–143 × 14–15 µm ($\bar{x}$ = 137 × 14.5 µm, n = 25), polysporous, unitunicate, clavate, pedicellate, with swollen upper portion, apex rounded, without an apical apparatus. *Ascospores* (7–)8–9(–10.5) × 2–2.6 *µm* ($\bar{x}$ = 8.6 × 2.2 µm, n = 50), overlapping, allantoid, one-celled, yellowish in mass. Asexual morph: Undetermined.

Material examined: CHINA, Guizhou Province, Libo County, Maolan Town, Maolan National Nature Reserve, on decaying wood, 8 July 2015, J.F. Zhang MLE-02 (GZAAS 20-4005); on decaying wood, 8 July 2015, J.F. Zhang MLE-12 (GZAAS 20-4006); on dead culm of herbaceous plant, 8 July 2015, J.F. Zhang MLC-30 (GZAAS 20-4007); on dead culm of herbaceous plant, 20 July 2016, J.F. Zhang MLC'-2 (MFLU 18-2287); Guiyang City, Yunyan District, Guizhou Botanical Garden, on dead branch of woody plant, 8 August 2017, J.F. Zhang ZWY-03 (MFLU 18-2240); Huaxi District, Guizhou Academy of Agricultural Sciences, on dead branch of woody plant, 28 April 2019, J.F. Zhang GZAAS-2019-7 (GZAAS 21-2009).

Notes: Six newly collected diatrypella-like specimen cluster with *D. vulgaris* in the present phylogenetic analysis (Figure 3.33). Morphologically, these specimens are typical of fungi in the genus *Diatrypella*, and especially resembles to *D. vulgaris* in many aspects that described by Trouillas et al. (2011), and including share the overlap sized of asci and ascospores. Therefore, these newly collections are identified as *Diatrypella vulgaris* based on the above evidence. And this species has been reported in Australia, China and Thailand (Trouillas et al., 2011; Hyde et al., 2017; Long et al., 2020).



Note a Appearance of stromata on host substrate. b Vertical section through stroma. c Vertical section through ascoma. d Section of peridium. e Ostiole with pariphysis. f, g Asci. h Ascospores. Scale bars: c = 100 µm, d = 10 µm, e, f = 30 µm, g, h = 5 µm.

Figure 3.28 *Diatrypella vulgaris* (GZAAS 20-4005)

Eutypa Tul. & C. Tul., Select. fung. carpol. (Paris) 2: 52 (1863)

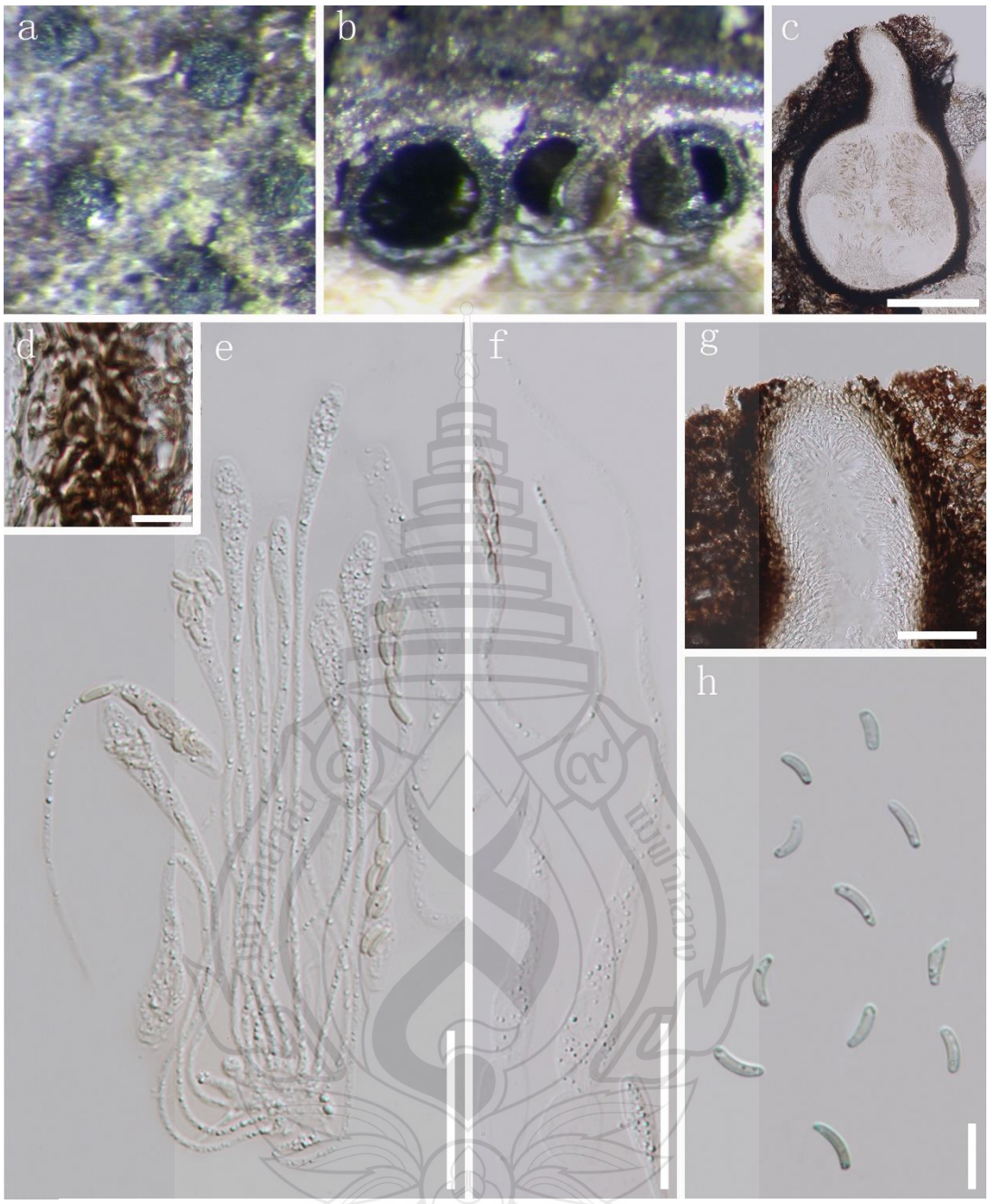
Eutypa was introduced by Tulasne & Tulasne (1863) with *Eutypa lata* as the type species. characteristics of this genus include irregular-shaped, solitary to gregarious stromata that erumpent through host epidermis, and often with entostromatic region, which comprised of white pseudoparenchymatous cells and thin black pseudoparenchymatous tissue around the white entostroma, 8-spored, spindle-shaped, long pedicellate asci, as well as oblong to allantoid ascospores (Rappaz, 1987; Moyo et al., 2018). *Eutypa* taxa have a broad geographic distribution (Carmarán et al., 2009; de Almeida et al., 2016; Moyo et al., 2018; Long et al., 2021), and are well known as the causal agents of Eutypa dieback of grapevine, apricots and cherries (Trouillas & Gubler, 2004, 2010; Baumgartner et al., 2010; Blanco-Ulate et al., 2013; Camps et al., 2014). Recently, around 131 species were accepted in *Eutypa* (Wijayawardene et al., 2020), while, the available sequence data of this genus is still limited in GenBank.

Eutypa cerasi S.H. Long & Q.R. Li, Journal of Fungi 83:23 (2021) Figure 3.29

Saprobic on decaying woody plant. Sexual morph: *Stromata* immersed, covering surface of host, surface dark brown to black, with 2–5 ascomata immersed in a single stroma. *Ascomata* semi-immersed in stroma, 362–602 μm high, 304–390 μm diam., globose to subglobose, with cylindrical neck, ostiolate. *Ostiole* central, opening separately, lined with abundant paraphysis in the ostiolar canal. *Peridium* 30–39, composed of several layers of thick-walled cells of *textura angularis*, becoming pale brown to subhyaline inwardly. *Hamathecium* of spase, septate, cellular paraphyses, 8–11 μm at the base, tapered towards the apex. *Asci* (91.5–)101.5–118(–150) × 5.5–7.5 μm ($\overline{x}$ = 111 × 6.5 μm n = 30) 8-spored, clavate, unitunicate, long pedicellate, rounded to truncate apex, apical rings inamyloid. *Ascospores* (5–)6–8.5 × 1.5–2 μm ($\overline{x}$ = 7 × 1.6 μm, n = 45), overlapping uni- to biseriate, allantoid, slightly curved, subhyaline to pale yellowish, smooth, aseptate, usually with oil droplets. Asexual morph: undetermined.

Material examined: CHINA, Guizhou Province, Guiyang City, Nanming District, on decaying woody plant, 14 May 2017, J.F. Zhang XCH-08 (GZAAS 20-4003).

Notes: Present phylogenetic analysis shows that our new specimen clusters with the strains of *Eutypa cerasi* with high bootstrap support (Figure 3.33). Observation of the morphological traits of the new collection, we found that it well fits *E. cerasi* that described by Long et al. (2021), including the similar texture of peridium and overlap sized asci and ascospores, except for the slightly larger ascomata and the different appearance of stromata in substrate. Each gene region between the strains of *E. cerasi* (GMBC0048 and GMBC0049) and our new collection was compared, and the results showed that there are only 1 base pair difference in ITS and Tub, respectively. Combining the morphological and phylogenetic analysis, the new collection in this study is identified as *Eutypa cerasi*. And which has been reported from karst formations, in Guizhou Province, China (Long et al., 2021).



Note a Appearance of stromata on host substrate. b Vertical section through stroma. c Vertical section through ascoma. d Section of peridium. e Immature and mature asci. f, Paraphyses. h Ascospores. Scale bars c = 200 μm, d, h = 10 μm, e, f = 30 μm, g = 50 μm.

Figure 3.29 *Eutypa cerasi* (GZAAS 20-4003)

Neoeutypella M. Raza, Q.J. Shang, Phookamsak & L. Cai, Fungal Diversity Notes: 10.1007/s13225-019-00421-w, [167] (2019)

Neoeutypella was introduced to accommodate its type species *N. baoshanensis* based on morphological and phylogenetic evidence (Phookamsak et al., 2019). So far, there is still only the single species accommodated in this genus.

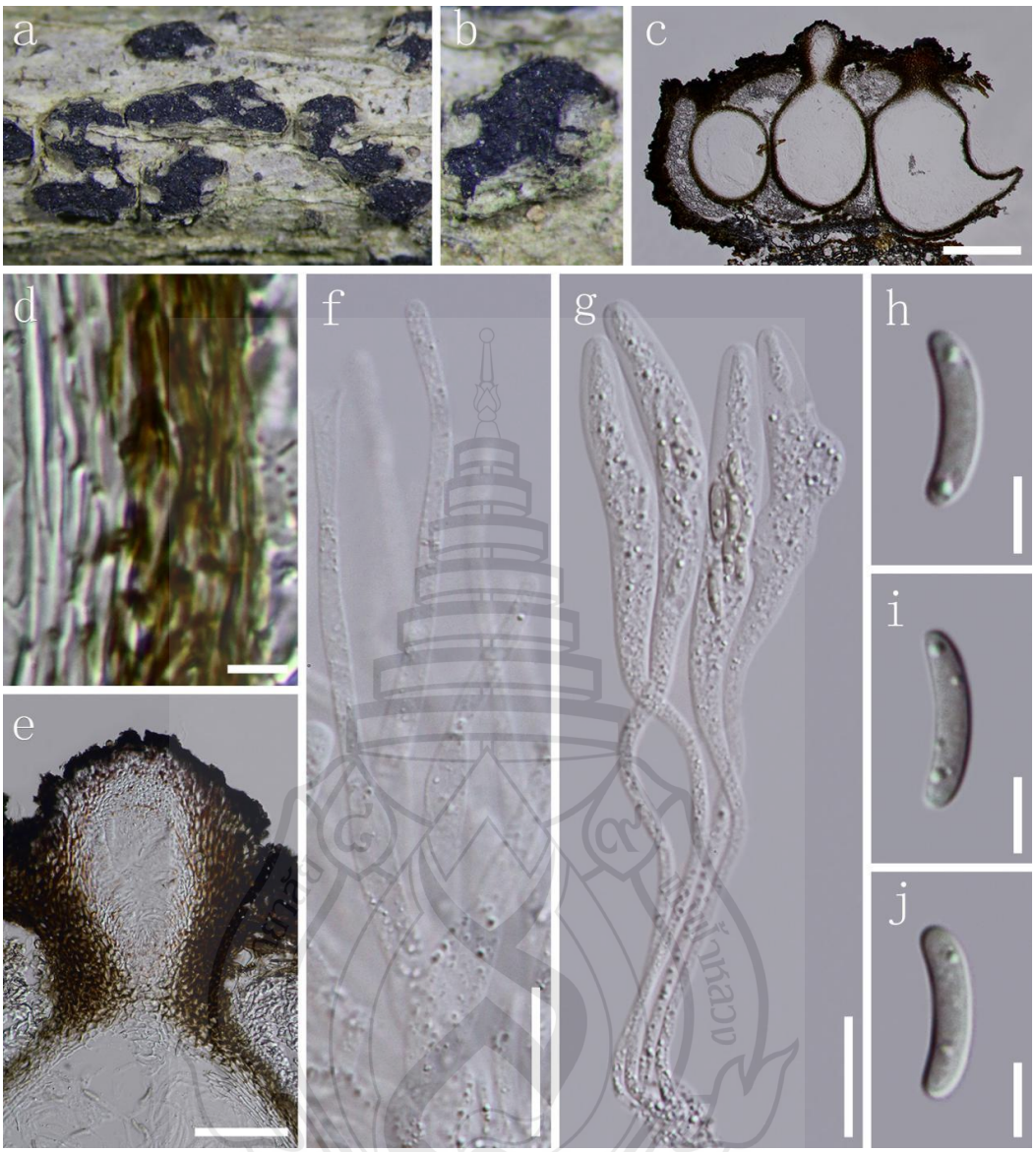
Neoeutypella baoshanensis M. Raza, Q.J. Shang, Phookamsak & L. Cai, Fungal Diversity 95: 168 (2019) Figure 3.30

Saprobic on dead woody or herbaceous plant. Sexual morph: *Stromata* 1602–2859 × 480–1078 μm, entostromatic, carbonaceous, black, solitary to gregarious, globose to long irregular in shape on host surface, erumpent through host epidermis, producing yellow pigments surrounding stroma. *Ascomata* perithecial, 489–670 μm high, 329–460 μm diam., black, immersed to semi-immersed in stromatic tissues, aggregated, globose or subglobose, papillate, ostiolate, ostiolar canal lined with numerous periphyses. *Peridium* 16.5–30 μm wide, two stratum, outer stratum comprising thick-walled, brown cells of *textura angularis*, inner stratum comprising 3–6 layers of thin-walled, hyaline cells of *textura prismatica*. *Hamathecium* comprising 3–9 μm wide, septate, filamentous paraphyses, tapering towards the apex, embedded in hyaline gelatinous matrix. *Asci* 112–131(–146) × 6–8(–9) μm ($\bar{x}$ = 128 × 7 μm, n = 20), 8-spored, unitunicate, spindle-shaped, long pedicellate, apically rounded with refractive cytoplasmic strands, amyloid, with a J-, subapical ring. *Ascospores* (8–)10–12.5(–14) × 2–3 μm ($\bar{x}$ = 10.8 × 2.3 μm, n = 40), overlapping 1–3-seriate, initially hyaline, becoming pale brown at maturity, allantoid, slightly or moderately curved, aseptate, mostly with 1–2 small guttules. Asexual morph: Undetermined.

Material examined: CHINA, Guizhou Province, Guiyang City, Huaxi District, Guizhou Academy of Agricultural Sciences, on dead woody plant, 5 November 2017, J.F. Zhang GZAAS-2017-8 (GZAAS 19-1764); Libo County, Maolan Town, Maolan National Nature Reserve, on dead culm of herbaceous plant, 8 July2015, J.F. Zhang MLC-12 (GZAAS 19-4022); on dead branch of woody plant, 5 November 2017, J.F. Zhang MLC-01 (GZAAS 19-4023).

Notes: Three new collections are phylogenetically placed within Diatrypaceae and clustered with *Neoeutypella baoshanensis* with full bootstrap support (Figure

3.33). Morphologically, these specimens resemble to *N. baoshanensis* in having perithecial ascomata that immersed in entostroma, unitunicate, spindle-shaped, long pedicellate asci, and similar sized, allantoid, aseptate ascospores, nevertheless, a few differences between them were also presented. The paraphyses of *N. baoshanensis* was described as aseptate and filamentous (Phookamsak et al., 2019), but it is clearly showed as septate, and swollen at the base in our new collections. Moreover, the measurements of asci in *N. baoshanensis* are comparatively smaller. However, these are not enough to warrant them as the separate species because of the paraphyses that presented in photo plate of *N. baoshanensis* looks like immature asci, and the asci are broken and seems to be missing a part of pedicel (Phookamsak et al., 2019). Based on the recommendations to delimit the species boundaries, each gene region between these newly collected specimens and *N. baoshanensis*, the results shows that there are only 1 and 2 base pair differences in ITS and Tub, respectively. Hence, we identify these three new collections as *N. baoshanensis*, and this species has been reported from Yunnan Province, China.



Note a, b Blackish ascostromata surrounded by yellow pigments on substrate. c Vertical section through ascostroma. d Section of peridium. e Ostiole. f Paraphyses. g Asci. h–j Ascospores. Scale bars: c = 200 μm, d, h–j = 5 μm, e = 50 μm, f, g = 20 μm.

Figure 3.30 *Neoeutypella baoshanensis* (GZAAS 19-1764)

Paraeutypella L.S. Dissan., J.C. Kang, Wijayaw. & K.D. Hyde

Paraeutypella was established by Dissanayake et al. (2021) to accommodate the type species *P. guizhouensis* and two combinations *P. citricola* and *P. vitis*. *Paraeutypella* is morphologically resembles to *Eutypella*, however, it is distinguished from the latter by comprising different number of perithecia in stroma, and phylogenetically distant from *Eutypella* in molecular analysis (Dissanayake et al., 2021). In this study, two novel species *P. elongiascus* and *P. karstia* are introduced based on morphological examination and molecular data. In addition, two species of *Allocryptovalsa* (including the type species *A. polyspora*) are placed within the genus *Paraeutypella* with high bootstrap support, however, *Allocryptovalsa* is clearly differs from *Paraeutypella* by its polysporous asci (Senwanna et al., 2017). Therefore, the phylogenetic relationship between these two genera is expected to document in the further study.

Paraeutypella elongiascus J.F. Zhang, J.K. Liu & Z.Y. Liu, *sp. nov*., Figure 3.31

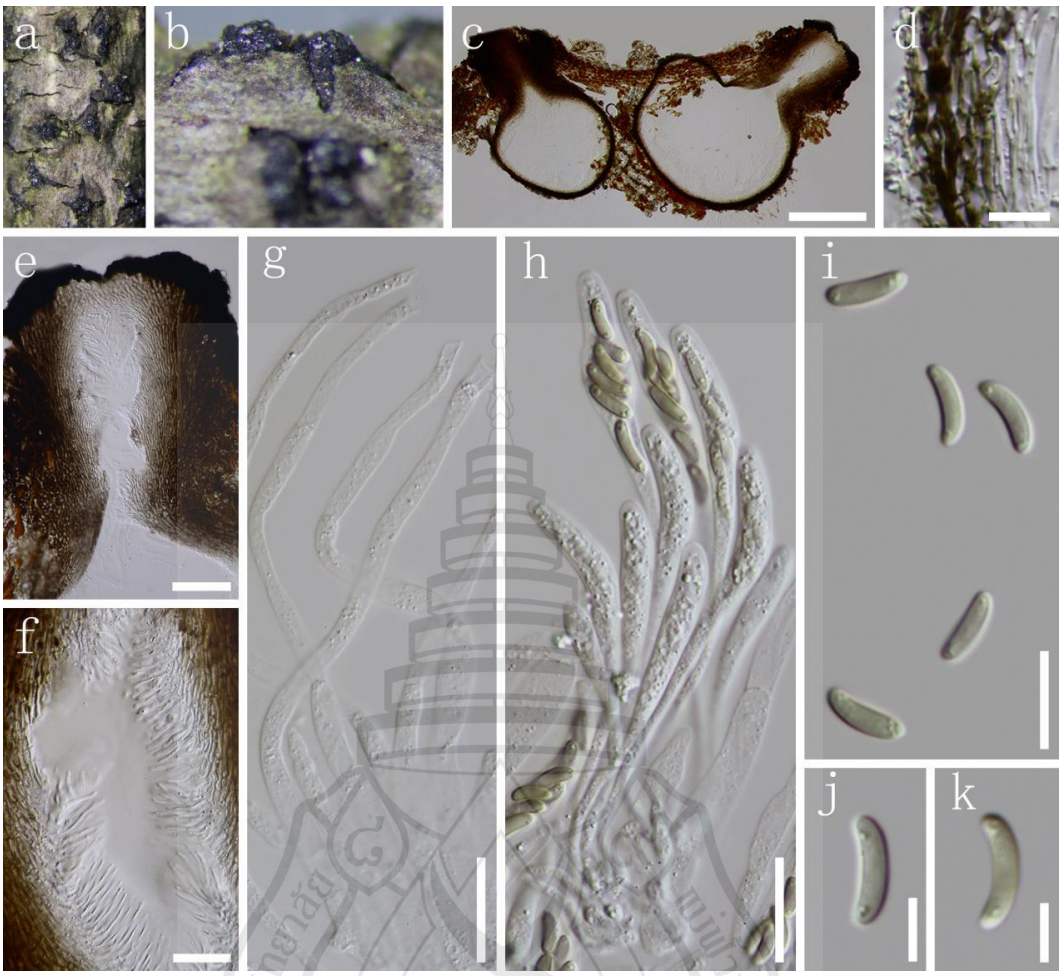
Etymology: named refers to its longer asci.

Holotype: GZAAS 19-1765

Saprobic on dead wood. Sexual morph: *Stromata* immersed in bark, erumpent, solitary or aggregated, circular to irregular in shape, superficial, carbonaceous. *Ascomata* 329–460 × 489–670 μm, perithecial, with groups of 4–6 ascomata arranged in a valsoid configuration, black, subglobose, clustered, immersed in ascostroma with ostiolar neck. *Ostiole* central, lined with numerous pariphysis in the ostiolar canal. *Peridium* 21–48 μm wide, composed of two stratum, inner stratum comprising 3–5 layers of hyaline, thin-walled cells arranged in *textura prismatica*, outer stratum of dark brown to black, thick-walled cells of *textura angularis*. *Hamathecium* comprising sparse, hyaline, unbranched, septate paraphyses, 5–9 μm wide at the base, tapering towards the apex, arising from base of perithecia. *Asci* (82–)93–104(–109) × 6–8.5 μm ($\overline{x}$ = 97.5 × 7 μm, n = 20), 8-spored, unitunicate, thin-walled, clavate to cylindric-clavate, long pedicellate, apically round with a J- apical ring. *Ascospores* 7.5–9.5 × 2–3 μm ($\overline{x}$ = 8.5 × 2.5 μm, n = 35), overlapping biseriate, allantoid, hyaline to light brown, smooth, aseptate, usually with two guttulates near the ends. Asexual morph: Undetermined.

Material examined: CHINA, Guizhou Province, Guiyang City, Huaxi District, Guizhou Academy of Agricultural Sciences, on dead wood, 30 Octomber 2017, J.F. Zhang GZAAS-2017-10 (GZAAS 19-1765, holotype); Yunan District, on dead branch of woody plant, 8 August 2017, J.F. Zhang ZWY-05 (MFLU 18-2241).

Notes: *Paraeutypella elongiascus* sisters to *P. vitis* with strong statistical support in the present study (Figure 3.33). Morphologically, *P. elongiascus* resembles to *P. guizhouensis* and *P. vitis* in having stromata that erumpent throughout the host, with groups of several ascomata arranged in a valsoid configuration, 8-spored, unitunicate, clavate to cylindric-clavate, long pedicellate asci and allantoid ascospores. However, it can be easily differentiated from three known *Paraeutypella species*, *P. citricola*, *P. guizhouensis* and *P. vitis* by its longer asci (Dissanayake et al., 2021). Hence, *Paraeutypella elongiascus* is introduced here as a new species on the basis of these above analysis.



Note a, b Stromata on substrate. c. vertical section through stroma. d Section of peridium. e Ostiolar canal. f Periphyses. g Paraphyses h Asci. i–k ascospores. Scale bars: c = 200 µm, d = 10 µm, e = 50 µm, f–i = 20 µm, j, k = 5 µm.

Figure 3.31 *Paraeutypella elongiascus* (GZAAS 19-1765, holotype)

Paraeutypella karstia J.F. Zhang & K.D. Hyde, *sp. nov.*, Figure 3.32

Etymology: named refers to the holotype was collected from karst formations.

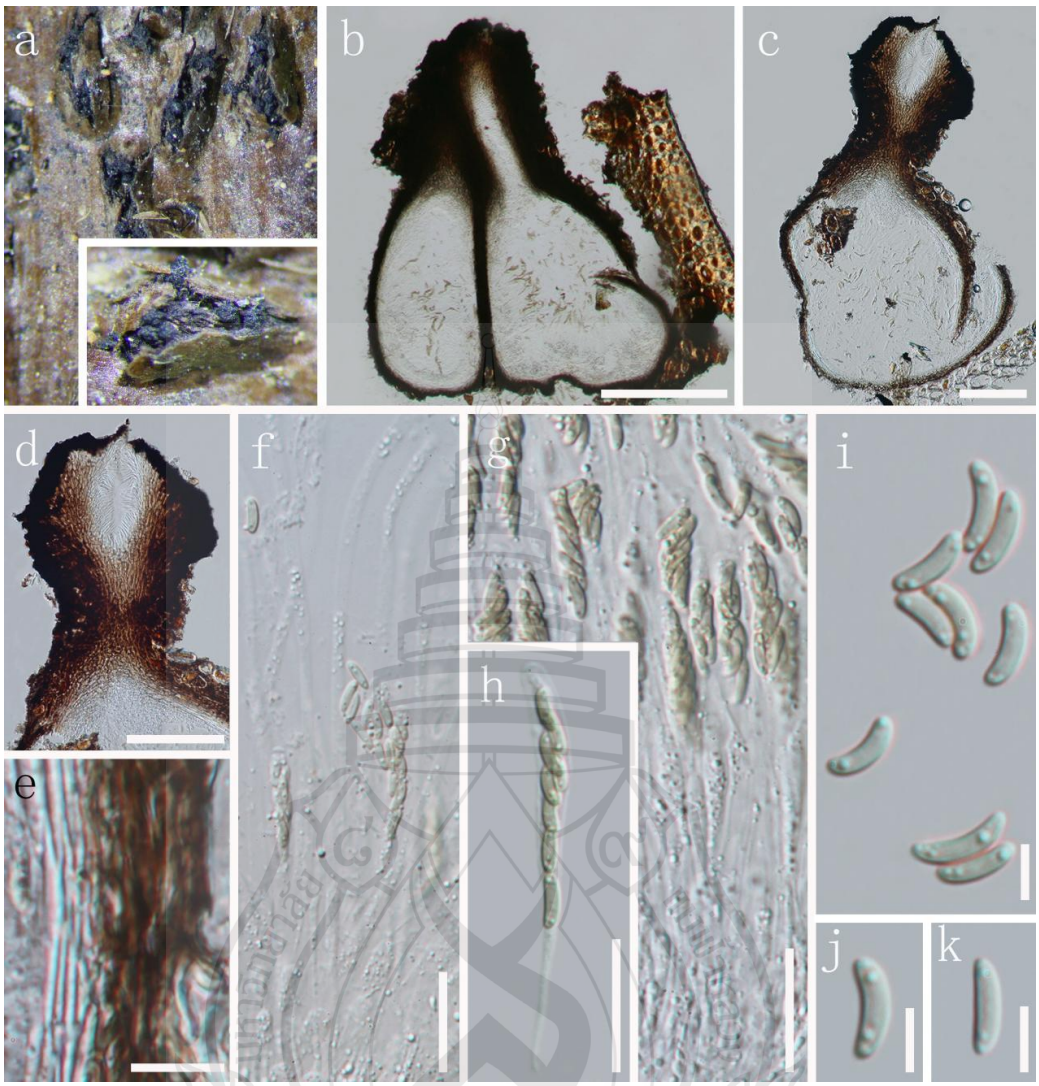
Holotype: GZAAS 20-4001.

Saprobic on dead wood. Sexual morph: *Stromata* immersed in bark, erumpent, solitary or aggregated, irregular in shape, superficial, carbonaceous. *Ascomata* immersed in ascostroma with ostiolar neck, 545–613 µm high, 262–392 µm wide., perithecial, with groups of several ascomata arranged in a valsoid configuration, black, subglobose, clustered. *Ostiole* central, 215–290 µm high × 145–194 µm wide., lined

with numerous pariphysis in the ostiolar canal. *Peridium* 18–51 μm wide, becoming thick towards the ostiole, composed of two stratum, inner stratum comprising several layers of hyaline, thin-walled cells arranged in *textura prismatica*, outer stratum of dark brown to dark brown, thick-walled cells of *textura angularis*. *Hamathecium* comprising sparse, 2.5–8 μm wide hyaline, unbranched, septate paraphyses, swollen at the base, tapering towards the apex, arising from base of perithecia. *Asci* 93–104(–114) × 4–5.5 μm ($\overline{x}$ = 100.5 × 5 μm, n = 25), 8-spored, unitunicate, thin-walled, clavate to cylindric-clavate, long pedicellate, apically round with a J- apical ring. *Ascospores* 7–9 × 1.5–2.5 μm ($\overline{x}$ = 8 × 2 μm, n = 40), overlapping uni- to biseriate, allantoid, hyaline to pale yellowish, smooth, aseptate, usually with two guttules near the ends. Asexual morph: Undetermined.

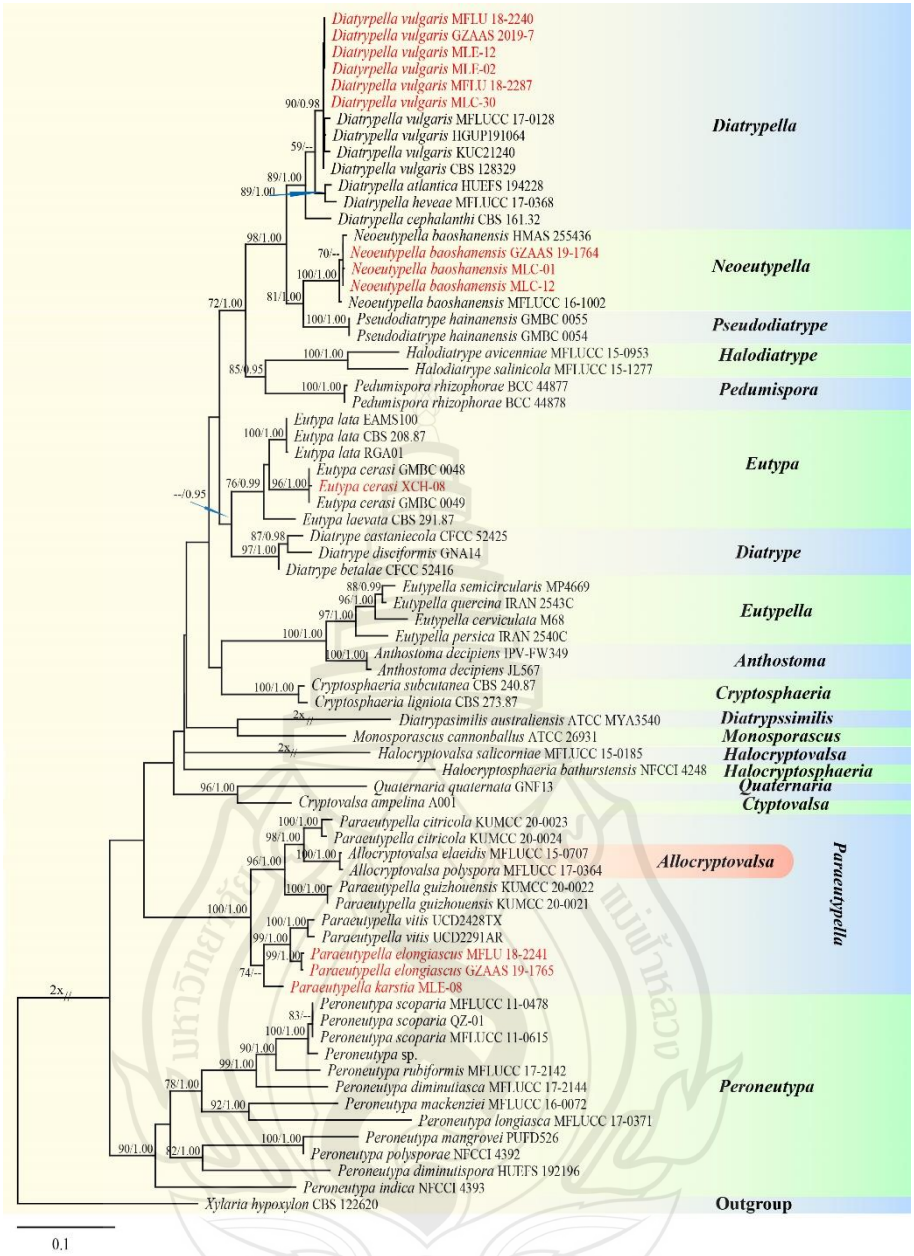
Material examined: CHINA, Guizhou Province, Libo County, Maolan Town, Maolan National Nature Reserve, on dead wood, 8 July 2015, J.F. Zhang MLE-08 (GZAAS 20-4001, holotype).

Notes: Phylogenetic analysis in the presnet study shows that *Paraeutypella karstia* forms an independent clade and where it is sister to *P. elongiascus* and *P. vitis* (Figure 3.33). Observation of the gross morphological features, we found that they are resemble in having stromatic ascomata that clustered and arranged in a valsoid configuration, and with a long ostiolar neck; 8-spored, unitunicate, clavate to cylindric-clavate, long pedicellate asci and allantoid ascospores. However, *P. karstia* is obviously distinguished from *P. vitis* by its longer asci (93–104(–114) × 4–5.5 μm vs. 55–80 × 5–9 μm), but smaller ascoapores (7–9 × 1.5–2.5 μm vs. 12–14 μm) (Ellis & Everhart, 1892; Dissanayake et al., 2021). As *P. karstia* and *P. elongiascus* share the similar measurements of peridium, paraphyses, asci and ascospores, comparison of each gene region between these two taxa is processed, and the results shows that there are 32/500 and 52/681 base pair differences (including the gaps) in ITS and Tub, respectively. Based on the above evidence, *Paraeutypella karstia* is therefore introduced as a new species.



Note a Stromata on substrate. b. vertical section through stroma. c Vertical section through ascomata. d Ostiolar canal with pariphysis. e Section of peridium. f Paraphyses. g, h Asci. i–k ascospores. Scale bars: b = 200 μm, c, d = 100 μm, e = 10 μm, f–h = 20 μm, i–k = 5 μm.

Figure 3.32 *Paraeutypella karstia* (GZAAS 20-4001, holotype)



Note Maximum likelihood bootstrap values equal or greater than 50% and Bayesian posterior probabilities equal or greater than 0.95 are indicated near the nodes. New sequence generated in this study are given in red. The tree is generated from maximum likelihood (ML) analysis based on ITS and Tub sequence data, and rooted with *Xylaria hypoxylon* CBS 122620.

Figure 3.33 Phylogenetic tree of the family Diatrypaceae

Hypoxylaceae DC.

The term Hypoxylaceae was originally mentioned in Lamarck and De Candolle (1805), but generally regarded as a synonym of Xylariaceae since they are similar in morphology. Both Daranagama et al. (2018) and Wendt et al. (2018) provided phylogenetic evidences for revising Hypoxylaceae and allied families at the family level, and resurrected Hypoxylaceae as a separate family in Xylariales. Wibberg et al. (2021) strengthened the phylogenetic placement of this family based on high quality genome sequences. Various numbers of genera were accepted within Hypoxylaceae by a series of studies (Daranagama et al., 2018; Wendt et al., 2018; Wijayawardene et al., 2018; Lambert et al., 2019; Phookamsak et al., 2019), but herein we follow Hyde et al. (2020c) who comprised 19 genera in this family.

Annulohypoxylon Y.M. Ju, J.D. Rogers & H.M. Hsieh, Mycologia 97(4): 855 (2005)

Based on morphological and molecular evidence, *Annulohypoxylon* was introduced by Hsieh et al. (2005) to accommodate *Hypoxylon* sect. *Annulata* with *Annulohypoxylon truncatum* as the type species. In their study, thirty-three *Hypoxylon* taxa were synonymized under *Annulohypoxylon*, and accounts of this genus were subsequently discovered by means of morphological, phylogenetic and chemotaxonomic analysis (Hsieh et al., 2005; Pereira et al., 2010; Liu et al., 2015; Fournier & Lechat, 2016; Li et al., 2016; Vasilyeva et al., 2016; Kuhnert et al., 2017). Currently, eighty-six epithets were listed in Index Fungorum (2022), however, several species were transferred to the genus *Jackrogersella* (Kuhnert et al., 2017), and around 60 species were included in *Annulohypoxylon* in the latest updated outline of fungi (Wijayawardene et al., 2022).

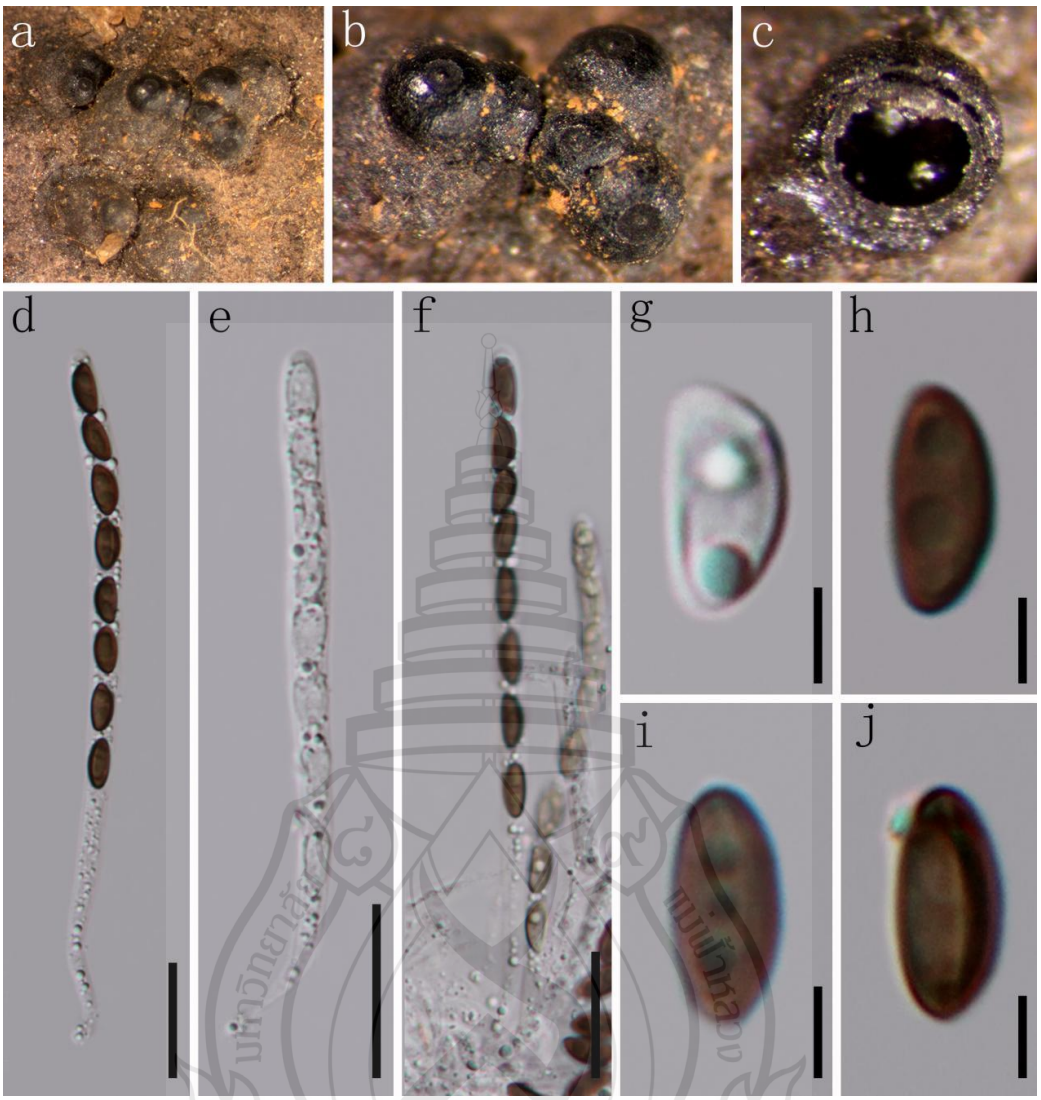
Annulohypoxylon thailandicum Daranag. & K.D. Hyde, Fungal Diversity: 10.1007/s13225-015-0324-y, [53] (2015) Figure 3.34

Saprobic on decaying wood. Sexual morph: *Ascostromata* glomerate, with conspicuous perithecial mounds, surface shiny black, blackish granules immediately beneath surface and between perithecia. *Perithecia* 0.45–0.55 × 0.6–0.9 mm, spherical, carbonaceous, stable attached with the host. *Ostioles* central, short papillate, encircled with a truncate-type disc, 0.2–0.35 μm diam. *Asci* 85.5–101.5(–110) × 4.2–5.2 μm ($\bar{x}$

= 93.5 × 4.5 µm, n = 20), 8-spored, unitunicate, cylindrical, long pedicellate, with a wedge-shaped, J+, subapical apparatus. *Ascospores* 7.5–9.5 × 3–4.5 µm ($\bar{x}$ = 8.5 × 3.5 µm, n = 35), uniseriate, one-celled, inequilaterally ellipsoidal with narrowly to rounded ends, hyaline when young, becoming brown at maturation, guttulate, with a straight germ slit, running along the entire spore-length on flattened side. Asexual morph: Undetermined.

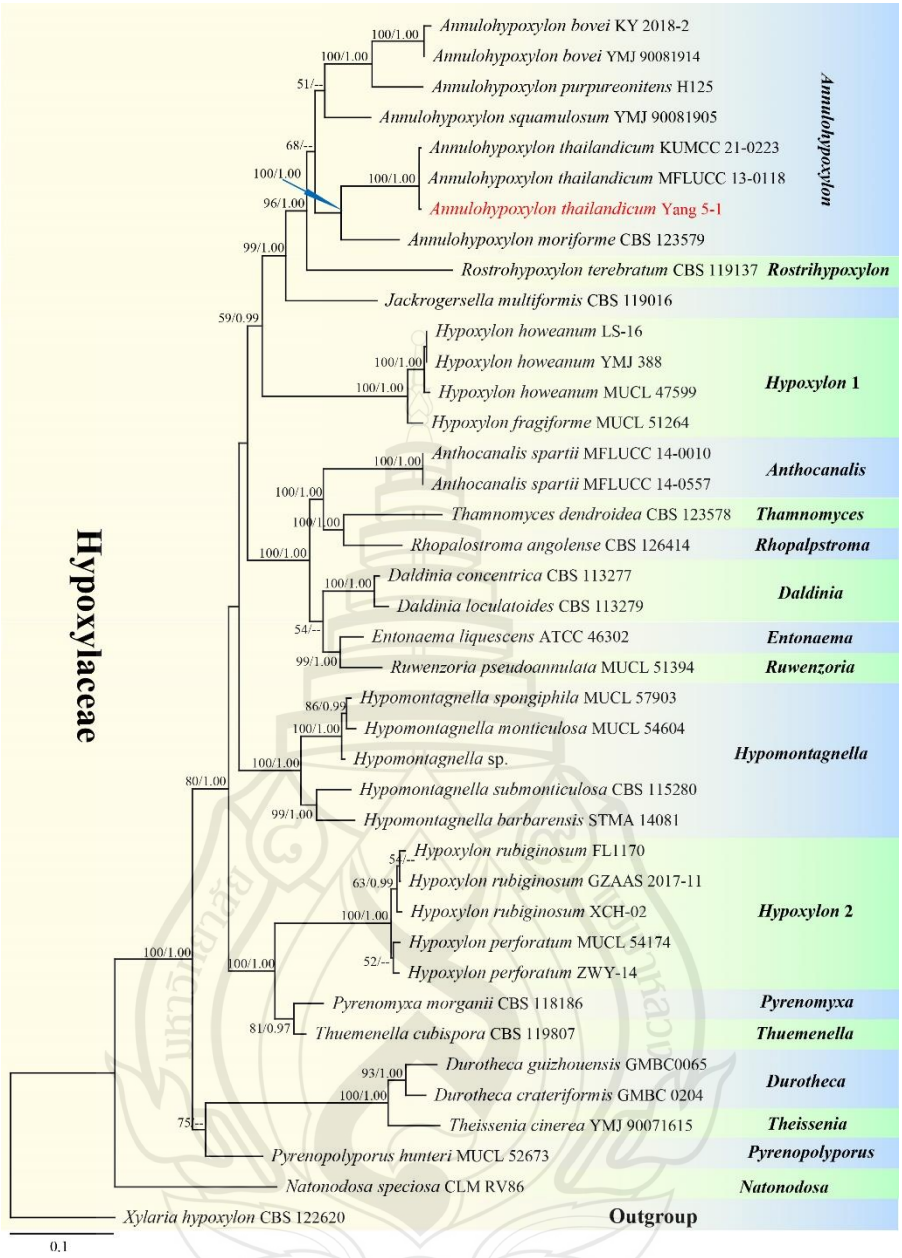
Material examined: CHINA, Guizhou Province, Xiyi City, on decaying wood, 8 June 2015, J.F. Zhang XY-5-1 (GZAAS 19-4025).

Notes: Our new collection clusters with the strains of *A. thailandicum* with high bootstrap support in the present study (Figure 3.35). Morphologically, it also well fits to *A. thailandicum* in having spherical ascomata with papillate ostiole that encircled with a truncate disk, long pedicellate asci, and brown, aseptate, inequilaterally ellipsoidal ascospores with a germ slit running full length of the spore. And even the measurements of the truncate disc at the apex, asci and ascospores between them are overlapped (Liu et al., 2015). Therefore, this new collection is identified as *A. thailandicum* and reported as a new geographical record as it has only discovered in Thailand (Liu et al., 2015).



Note a Apperance of stromata in host surface. b Stromata with a visible, flattened disc. c Vertical section through the perithecia. d–f Long, cylindrical asci with ascospores. g–j Ascospores. Scale bars: d–f= 30 µm, g–i = 5 µm.

Figure 3.34 *Annulohypoxylon thailandicum* (GZAAS 19-4025, new record)



Note Maximum likelihood bootstrap values equal or greater than 50% and Bayesian posterior probabilities equal or greater than 0.95 are indicated near the nodes. New sequence generated in this study is given in red. The tree is generated from maximum likelihood (ML) analysis based on ITS, LSU, Rpb2 and Tub sequence data, and rooted with *Xylaria hypoxylon* CBS 122620.

Figure 3.35 Phylogenetic tree of the family Hypoxylaceae

Xylariaceae Tul. & C. Tul.

Xylariaceae was originally introduced with an uncertain ranked term "*Xylariei*", and until it was taken up as a family by Miler (1928). It is generally considered as one of the largest and most diverse ascomycete families with 87 genera and more than 1300 species in previous studies (Tang et al., 2009; Daranagama et al., 2015; Maharachchikumbura et al., 2015; Johnston et al., 2016; Koyani et al., 2016; U'Ren et al., 2016; Kuhnert et al., 2017; Tibpromma et al., 2017a). However, there are only 32 genera were accommodated within this family by Hyde et al. (2020c) and Wijayawardene et al. (2020). Recently, other two genera *Linosporopsis* and *Neoxylaria* were added to Xylariaceae by Voglmayr and Beenken (2020) and Konta et al. (2020) based on morphological and phylogenetic studies. Wittstein et al. (2020) resurrected the genus *Dematophora* in accordance with polythetic taxonomy.

Stellatus J.F. Zhang & K.D. Hyde, *gen. nov.*

Etymology: named refers to the ascomata of type species appeared as stellate in host surface.

Type species: *Stellatus guiuzhouensis*

Saprobic on dead leaves of woody plant. Sexual morph: *Ascomata* scattered, solitary to gregarious, dark brown to black, globose to subglobose, immersed to erumpent, coriaceous, ostiolate, appeared as irregular stellate in substrate surface. *Peridium* composed of several layers of brown, thick-walled cells of *textura angularis* to *prismatica*. *Hamathecium* of numerous, septate paraphyses that anastomosing between and above the asci. *Asci* 8-spored, cylindric-clavate, unitunicate, short-pedicellate. *Ascospores* overlapping uni- to bi-seriate, ellipsoidal inequilaterally with narrowly round ends, aseptate, chocolate or dark brown, with germ slit straight that running along with the entire spore-length, associated with a mucilaginous appendage at both ends. Asexual morph: Undetermined.

Notes: *Stellatus* is placed within Xylariaceae as an independent clade with moderate bootstrap support in the present study (Figure 3.38). The gross morphological characteristics of *Stellatus* are corresponds to Xylariaceae, but significantly differs from the known genera by its uni-loculate, non-stromatica ascomata appeared as stellate in substrate surface, and the ascospores associated with a mucilaginous appendage at both ends.

Stellatus guizhouensis J.F. Zhang & K.D. Hyde, *sp. nov*., Figure 3.36

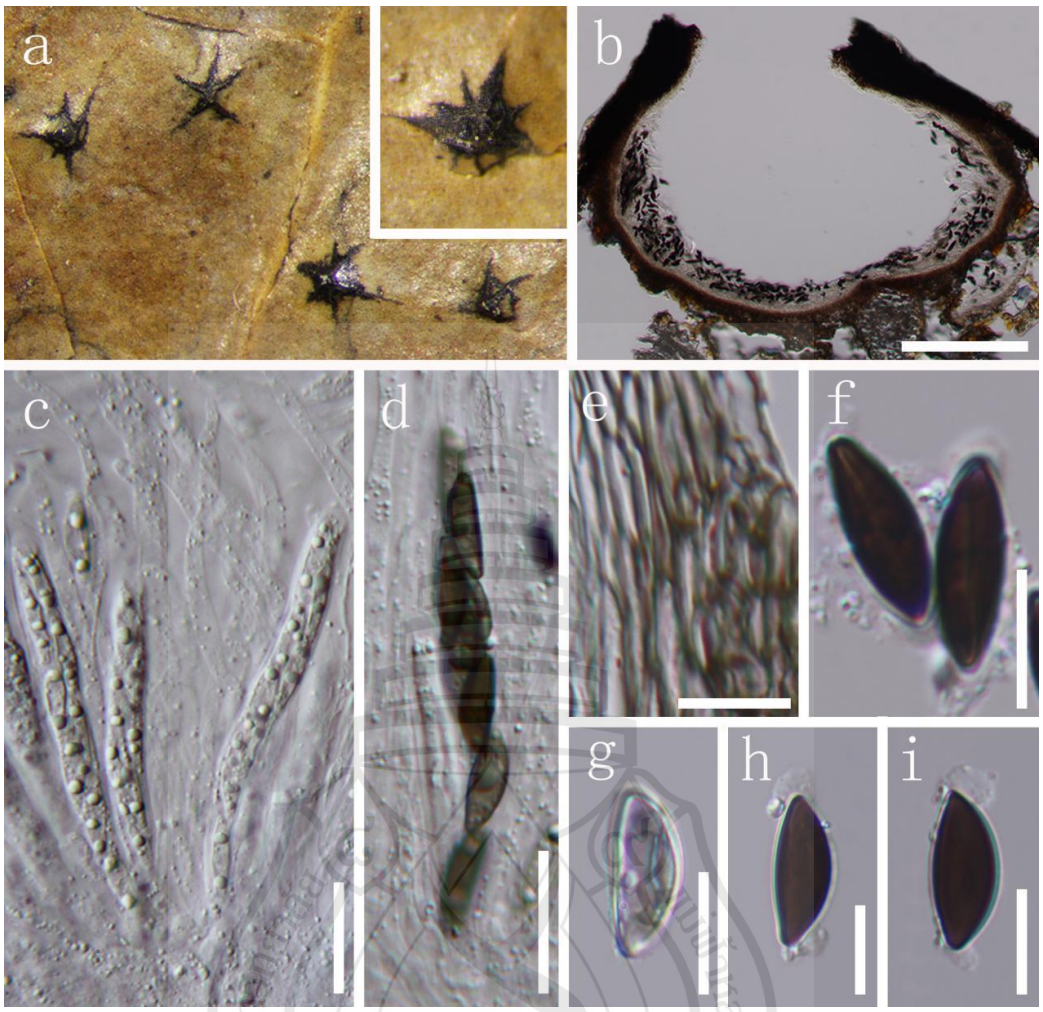
Etymology: named refers to Guizhou Province where the holotype was collected.

Holotype: GZAAS 20-4002.

Saprobic on dead leaves of woody plant. Sexual morph: *Ascomata* scattered, solitary to gregarious, dark brown to black, globose to subglobose, immersed to erumpent, coriaceous, ostiolate, visible as irregular stellate in substrate surface. *Peridium* 19–48 µm diam, thin at the base and sides, become thick around the ostiole, composed of several layers of brown, thick-walled cells of *textura angularis* to *prismatica*. *Hamathecium* of numerous, septate paraphyses, 3–5.5 µm at the base, and tapering towards the apex, anastomosing between and above the asci. *Asci* 82–92 × 7.5–9.5(–11) µm ($\overline{x}$ = 86 × 8 µm, n = 15), 8-spored, cylindric-clavate, unitunicate, short-pedicellate. *Ascospores* 14–17.5 × 5–6 µm ($\overline{x}$ = 16 × 5.5 µm, n = 35), overlapping uni- to bi-seriate, ellipsoidal inequilaterally with narrowly round ends, aseptate, initially hyaline and becoming chocolate or dark brown at maturation, with germ slit straight that running along with the entire spore-length, wall smooth and thick, associated with a mucilaginous appendage at both ends. Asexual morph: Undetermined.

Material examined: CHINA, Guizhou Province, Libo County, Maolan Town, Maolan National Nature Reserve, on dead leaves of woody plant, 18 July 2016, J.F. Zhang MLBB-6 (GZAAS 20-4002, holotype).

Notes: *Stellatus guizhouensis* forms an independent clade that closes to the taxa of *Brunneiperidium*, *Kretzschmaria* and *Xylaria* (Figure 3.38). However, *Stellatus* is easily differs from *Brunneiperidium* by its immersed ascomata, clavate asci and ellipsoidal inequilaterally ascospores associated with a mucilaginous appendage at both ends. While the species in *Brunneiperidium* are characterized by having superficial, conical ascomata, slender cylindrical asci and ellipsoidal ascospores that surround by an entire mucilaginous sheath (Daranagama et al., 2015). And *Stellatus* can be distinguished from *Kretzschmaria* and *Xylaria* by the taxa in the latter two genera are characterized in forming variable shaped, obviously stromata in host surface, cylindrical asci and the ascosproes also lacking any mucilaginous appendages (Pourmoghaddam et al., 2018; Hyde et al., 2020c).



Note a Appearance of ascomata immersed in the host. b Vertical section through ascomata. c, d Immature and mature asci with paraphyses. e Section of Peridium. f Ascospores with germ slit. g–i Immature and mature ascospores. Scale bars b = 200 µm, c, d = 20 µm, e–i = 10 µm.

Figure 3.36 *Stellatus guizhouensis* (GZAAS 20-4002, holotype)

Rosellinia De Not., G. bot. ital. 1(1): 334 (1844)

Rosellinia was introduced by De Notaris (1844) to accommodate the species with uniperitheciate, gregarious, stromata. Petrini (2013) revised this genus and accepted 142 species which are characterized by superficial, conspicuous, carbonaceous, ostiolate stromata with subicula. *Dematophora* has been synonymized under *Rosellinia* because of its sexual morph resembles to *Rosellinia*, but these two

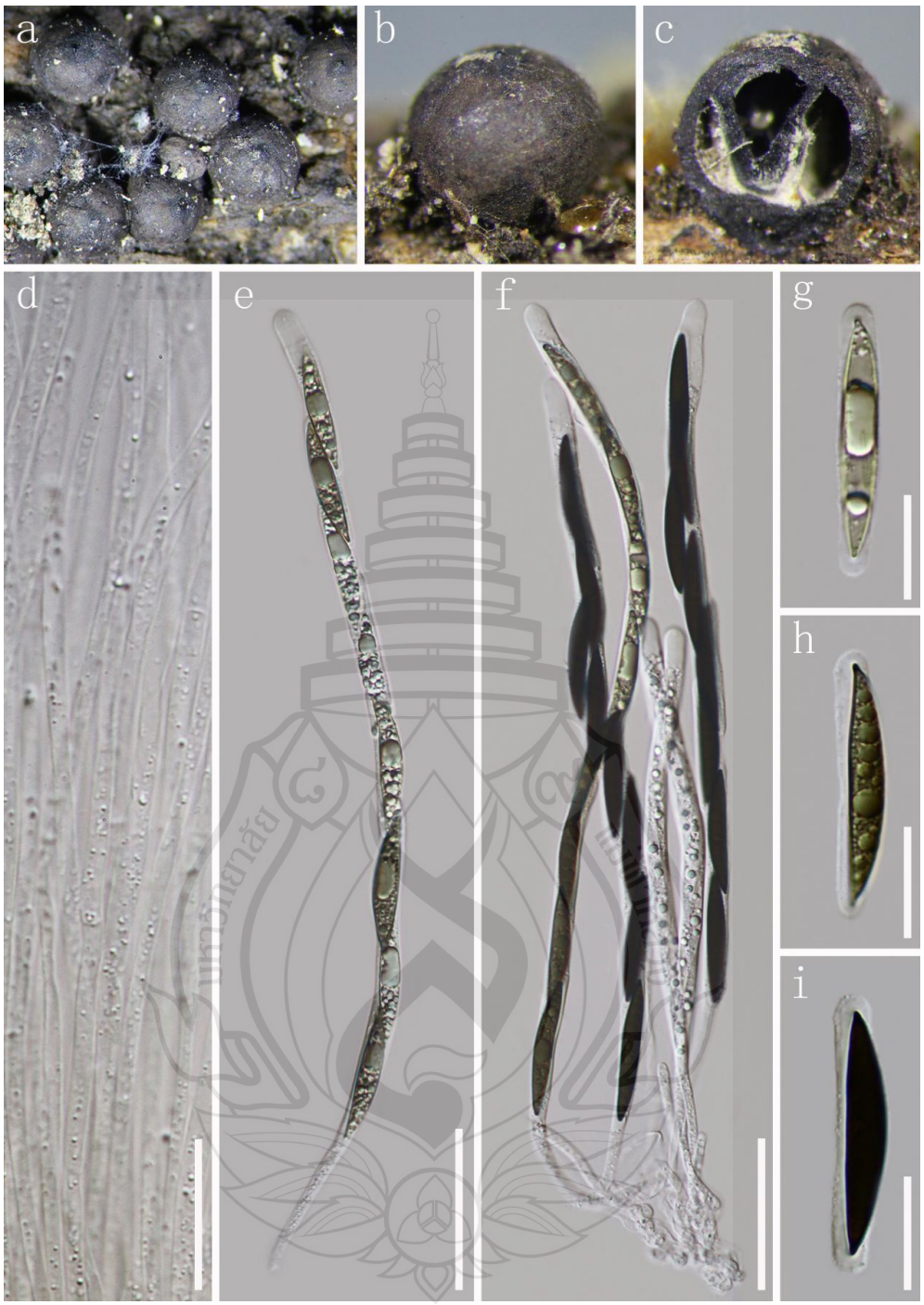
genera were differentiated and resurrected based on phylogenetic and secondary metabolite data (Wittstein et al., 2020). There are more than 500 epithets listed in Index Fungorum (2022), only 158 species were accepted (Hyde et al., 2020c) and 27 taxa with available sequence data.

Rosellinia necatrix R. Hartig in Untersuch. Forstbot. Inst. München 3: 126 (1883). Figure 3.37

Saprobic on decaying wood. Sexual morph: *Ascomata* semi-immersed to superficial, spherical with a short papilla, scattered or clustered in dense. *Peridium* composed of dark pigmented, carbonaceous cells. *Hamathecium* comprising dense, 2–4 µm wide, trabeculate paraphyses, anastomosing between and above the asci, embedded in a gelatinous matrix. *Asci* (286–)304.5–336(–356.5) × 7–10(–12) µm ($\overline{x}$ = 324 × 9 µm, n = 25), 8-spored, long-cylindric, apical thick, rounded with an apical apparatus, long pedicellate with a bulbous pedicel. *Ascospores* 42.5–54.5 × 7.5–10.5 µm ($\overline{x}$ = 49 × 8.5 µm, n = 40), fusiform, one-celled, uniseriate, straight or lightly curved, tapering towards both ends, associated with gelatinous sheath in both ends and one side, hyaline to pale brown, guttulate, verrucose when young, becoming dark brown to black at maturation. Asexual morph: Undetermined.

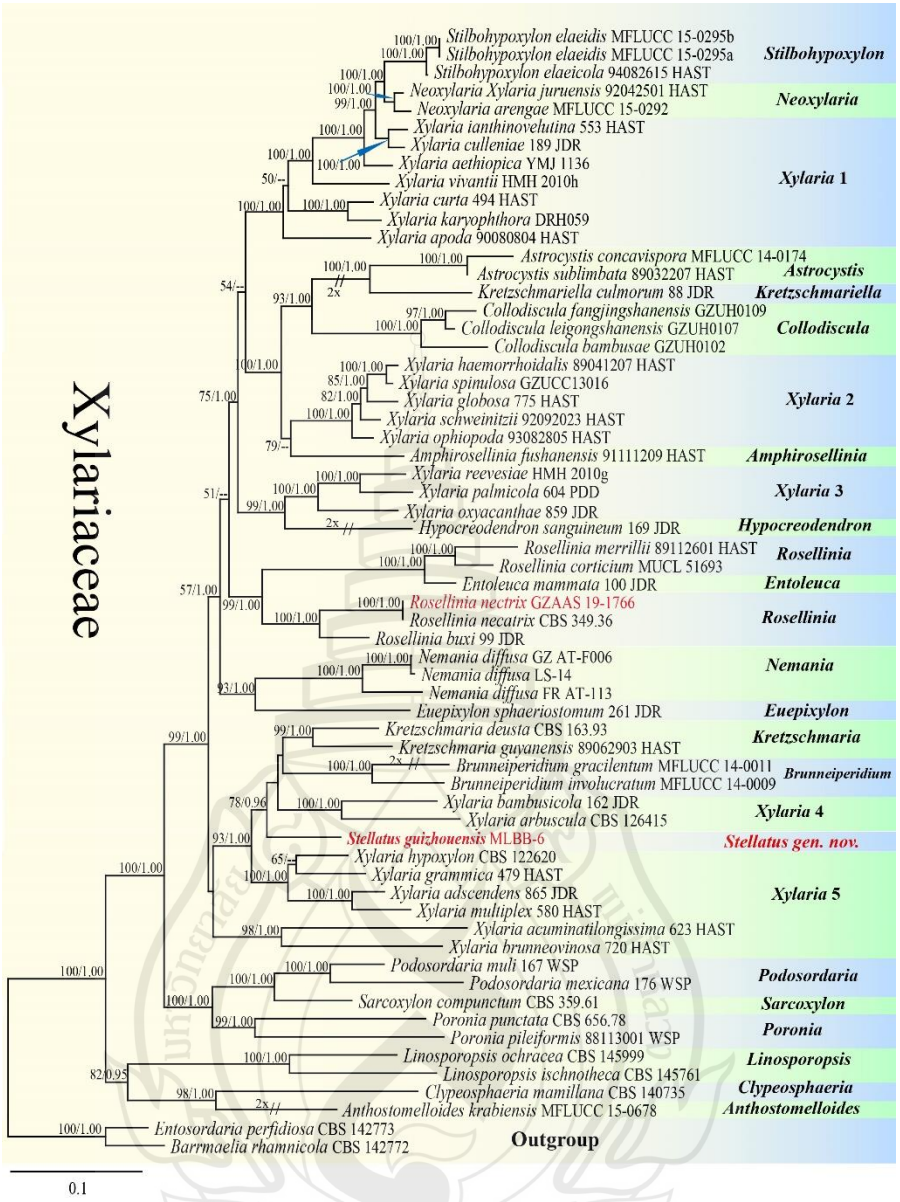
Material examined: CHINA, Guizhou Province, Leishan County, on decaying wood, 2 November 2017, J.F. Zhang LS-23 (GZAAS 19-1766).

Notes: Present phylogenetic analysis shows that our new collection clusters to *Rosellinia necatrix* with full bootstrap support (Figure 3.38). It is typical of *Rosellinia* in having superficial, conspicuous, carbonaceous, ostiolate stromata with subicula, long cylindrical asci that bearing long ascospores with thread-like ends and germ slits (Petrini, 2013; Hyde et al., 2020c). Moreover, the measurements of asci and ascospores, and the strict feature of ascospores associated with conspicuous gelatinous sheath in both ends and one side all correspond to *R. necatrix* that described and summarized in previous studies (Petrini & Petrini, 2005; Petrini, 2013). Hence, the new specimen is identified as *R. necatrix* herein based on morphological and phylogenetic evidence. And this species has been reported as an important pathogenetic fungi that caused white root rot in varies plant.



Note a, b Appearance of ascomata in the host. c Veritical section through ascoma. d Paraphyses. e, f Asci in different maturation stage. g–i Ascospores in different maturation stage. Scale bars d, g–i = 20 µm, e, f = 50 µm.

Figure 3.37 *Rosellinia necatrix* (GZAAS 19-1766)



Note Maximum likelihood bootstrap values equal or greater than 50% and Bayesian posterior probabilities equal or greater than 0.95 are indicated near the nodes. New sequence generated in this study are given in red. The tree is generated from maximum likelihood (ML) analysis based on ITS, LSU, Rpb2 and Tub sequence data, and rooted with *Barrmaelia rhamnicola* CBS 142772 and *Entosordaria perfidiosa* CBS 142773.

Figure 3.38 Phylogenetic tree of the family Xylariaceae

CHAPTER 4

TAXONOMIC AND PHYLOGENETIC OF LEOTIOMYCETES FROM KARST LANDSCAPE OF GUIZHOU PROVINCE

4.1 Introduction

Rhytismataceae (Rhytismatales) was established by Chevallier (1826), typified by *Rhytisma* with *R. acerinum* (Pers.) Fr. as the type species and belongs in Rhytismatales, Leotiomycetes, Ascomycota (Wijayawardene et al., 2020). Members of this family produce variously shaped apothecia that may be sessile, circular, navicular or hysteriform and that typically open by a longitudinal split or radial fissures. Asci are cylindrical, saccate to clavate. Ascospores are one-celled or multi-septate and vary from bacilliform to fusiform or filiform, with or without a sheath (Darker, 1967; Ekanayaka et al., 2019). Species of Rhytismataceae occur on a wide range of hosts with a worldwide distribution (Cannon & Minter, 1986; Johnston, 1986; Hou & Piepenbring, 2009; Hernández et al., 2014; Li et al., 2014; Tanney & Seifert, 2017; Cai et al., 2020b).

Darker (1967) proposed the generic delimitation for Rhytismataceae, based on ascoma and ascospore shapes, although this has been challenged in later studies (Cannon & Minter, 1986; Johnston, 1990, 2001; Hou et al., 2005). However, Darker (1967) and Cannon & Minter (1986) were followed due to lack of an alternative scheme. Molecular studies (Gernandt et al., 2001; Johnston & Park, 2007; Lantz et al., 2011; Tian et al., 2013; Zhang et al., 2015) had revealed the phylogenetic relationships amongst members of Rhytismatales, but the available sequence data for this group remains limited and a phylogenetic classification of some members is unresolved. There are around 50 genera with 1000 species presently accepted in

Rhytismataceae (Lumbsch & Huhndorf, 2007; Wijayawardene et al., 2018); however, a systematic genus-level taxonomic revision is needed to provide a clear, natural generic delimitation within this family and the relationship between Rhytismataceae and allied families within Rhytismatales needs to be resolved (Johnston et al., 2019).

Karst formations are generally characterized by sinking streams, caves, enclosed depressions, fluted rock outcrops and large springs (Ford & Williams, 2007). Guizhou, as the eastern portion of the Yunnan-Guizhou Plateau, has the largest proportion of rocky desertification and karst landforms in China (Huang & Cai, 2006). The flora in this area, comprising of 264 families with 1667 genera and 7505 vascular plants species, were inventoried from Guizhou Province (Liu et al., 2018). Therefore, it would be interesting to study the fungi in this area because of its unique ecological environment and rich plant resources. A series of studies have already been carried out and yielded several new species (Zhang et al., 2016, 2017b, c, 2018, 2019). The objectives of this study are to introduce four novel species of Rhytismataceae, based on phylogenetic and morphological evidence and elucidate their affinities with related species.

4.2 Materials and Methods

4.2.1 Collection, Examination, Isolation and Specimen Deposition

Specimens were collected from Guizhou Province from 2016 to 2017 and examined in the laboratory with a Motic SMZ 168 stereomicroscope. Vertical sections of fruiting bodies were made by hand and mounted in water for microscopy. Macro-morphological characters were captured using a stereomicroscope (Nikon SMZ800N) with a Cannon EOS 70D digital camera. Micro-morphological characters were observed by differential interference contrast (DIC) using a Nikon ECLIPSE 80*i* compound microscope and captured by a Cannon EOS 600D digital camera. Measurements were processed in a Tarosoft (R) Image Frame Work version 0.9.7 programme and photographic plates were edited in Adobe Photoshop CS6 (Adobe Systems Inc., USA).

The single spore isolation technique described in Chomnunti et al. (2014) was followed to obtain the pure cultures of these specimens. Single germinated ascospore was picked up and transferred to PDA for recording growth rates and culture characteristics.

The holotypes are deposited at the Herbarium of Mae Fah Luang University (MFLU), Chiang Rai, Thailand or Guizhou Academy of Agricultural Sciences (GZAAS), Guizhou, China. Ex-type living culture is deposited at Guizhou Culture Collection (GZCC), Guiyang, China. Index Fungorum and Facesoffungi numbers are provided according to Jayasiri et al. (2015) and Index Fungorum (2020). New species were established, based on the recommendations from Jeewon & Hyde (2016).

4.2.2 DNA Extraction, PCR Amplification and Sequencing

Following the manufacturer's instructions, the total genomic DNA was extracted from cultures using a Biospin Fungus Genomic DNA Extraction Kit (BioFlux, Hangzhou, P. R. China) or extracted from the fruiting bodies using an E.Z.N.A. Forensic DNA kit (Omega Bio-Tek, Doraville, Georgia, USA).

Polymerase chain reactions (PCR) were performed in 25 µl reaction volumes, which contained 9.5 µl distilled-deionised-water, 12.5 µl of 2 × Power Taq PCR Master Mix (TIANGEN Co., China), 1 µl of DNA template and 1 µl of each forward and reverse primers. Three different loci were used in this study. The internal transcribed spacer (ITS) and 28S large subunit of the nuclear ribosomal DNA (LSU) regions were amplified by using the primers ITS4/ITS5 and LR0R/LR5, respectively (White et al., 1990; Gardes & Bruns, 1993). The primers mrSSU1 and mrSSU3R were used for amplification of the mitochondrial small subunit (mtSSU) partial regions (Zoller et al., 1999). The PCR thermal cycle programme was performed according to White et al. (1990), Gardes & Bruns (1993) and Zoller et al. (1999).

Amplicon size and concentration were assessed by gel electrophoresis with 1.2% agarose stained with ethidium bromide. PCR products were purified and sequenced at Sangon Biotechnology Co. Ltd (Shanghai, P. R. China).

4.2.3 Sequence Alignment and Phylogenetic Analysis

For phylogenetic reconstruction, newly-generated sequences were initially subjected to BLAST search (BLASTn) in NCBI (https://www.ncbi.nlm.nih.gov) and

additional related sequences were selected and downloaded from GenBank (https://www. ncbi.nlm.nih.gov/genbank/), based on BLASTn results and recent publications (Tian et al., 2013; Wang et al., 2013; Zhang et al., 2015; Johnston et al., 2019; Cai et al., 2020b). The sequences used in this study for phylogenetic analysis are listed in Table 1. All of these sequences were aligned and manually improved with BioEdit v. 7.2 (Hall, 1999) and then assembled as a dataset of LSU-ITS-mtSSU to infer the phylogenetic placement of newly identified taxa.

Phylogenetic analyses were performed using the algorithm of Maximum-Parsimony (MP) and Bayesian Inference (BI). MP analyses were run using PAUP v. 4.0b10 (Swofford, 2002) with 1000 replications and inferred using the heuristic search option with 1000 random taxa. All characters were unordered and of equal weight and gaps were treated as missing data. Maxtrees was set as 1000, zero-length branches were collapsed and all equally parsimonious trees were saved. Clade stability was accessed using a bootstrap (BT) analysis with 1000 replicates, each with ten replicates of random stepwise addition of taxa (Hillis & Bull, 1993).

BI analyses were carried out by using MrBayes v. 3.2 (Ronquist et al., 2012). The best-fit model (GTR+I+G for LSU, ITS and mtSSU) of evolution was estimated in MrModeltest 2.3 (Nylander, 2008). Posterior Probabilities (PP) (Rannala & Yang, 1996; Zhaxybayeva & Gogarten, 2002) were determined by Markov Chain Monte Carlo sampling (MCMC) in MrBayes v. 3.2. Six simultaneous Markov chains were run for 10,000,000 generations and trees were sampled every 100th generation. The temperature values were lowered to 0.15, burn-in was set to 0.25 and the run was automatically stopped as soon as the average standard deviation of split frequencies reached below 0.01. The phylogram was visualised in TreeView (Page, 1996) and edited in Adobe Illustrator CS v. 5 (Adobe Systems Inc., USA).

Table 4.1 Taxa used in this study. Strains generated/sequenced in this study are given in bold

Species	Specimen/Strain No.	LSU	ITS	mtSSU
Bifusella camelliae	HOU1094	KF797447	KF797435	KF797458
Bifusella camelliae	HOU701B	KF797448	KF797436	KF797459
Coccomyces anhuiensis	HOU 1265	MK371314	MK371313	MK371315
Coccomyces dentatus	AFTOL ID-147	AY544657	DQ491499	AY544736
Colpoma ledi	Lantz 379 (UPS)	HM140512	---	HM143788
Colpoma quercinum	Lantz 368 (UPS)	HM140513	---	HM143789
Cryptomyces maximus	Lantz & Minter 424 (UPS)	HM140514	---	HM143790
Discocainia nervalis	1045	KJ513473	KJ507206	---
Duplicariella phyllodoces	Lantz 389 (UPS)	HM140516	---	---
Hypoderma berberidis	HOU892	JX232420	JX232414	KF813010
Hypoderma berberidis	HOU942	JX232421	JX232415	KF813009
Hypoderma campanulatum	ICMP 17383	HM140517	---	HM143792
Hypoderma carinatum	ICMP 18322	HM140518	---	HM143793
Hypoderma commune	Hanson 2006-451 (UPS)	HM140519	JF690769	HM143794
Hypoderma cordylines	ICMP 17344	HM140521	JF683421	HM143796
Hypoderma cordylines	ICMP 17396	HM140520	---	HM143795
Hypoderma hederae	Lantz & Minter 421 (UPS)	HM140522	JF690770	HM143797
Hypoderma liliense	ICMP 18323	HM140523	MH921859	HM143798
Hypoderma liliense	ICMP 18324	HM140524	---	HM143799
Hypoderma minterii	275B	JX232418	JX232416	---
Hypoderma obtectum	ICMP 17365	HM140525	---	HM143800
Hypoderma paralinderae	GZAAS 19-1769	MN638878	MN638873	MN638868
Hypoderma rubi	ICMP 17339	HM140526	JF683419	HM143801
Hypoderma rubi	ICMP 18325	HM140527	JF683418	HM143802
Hypoderma sticheri	ICMP 17353	HM140529	MK039702	HM143804
Hypoderma vincetoxici	Lantz 405 (UPS)	HM140530	JF690772	HM143805
Hypohelion anhuiense	HOU 1078	KF797443	KF797431	KF797455
Lirula macrospora	Hou et al. 13 (BJTC)	HQ902159	HQ902152	---
Lirula yunnanensis	Hou et al. 464A (BJTC)	HQ902149	HQ902156	---
Lophodermium arundinaceum	Lantz 323 (UPS)	HM140535	---	HM143811
Lophodermium culmigenum	ICMP 18328	HM140538	---	HM143814
Marthamyces emarginatus	ICMP 22854	MK599203	MH921869	MK598751

Table 4.1 (continued)

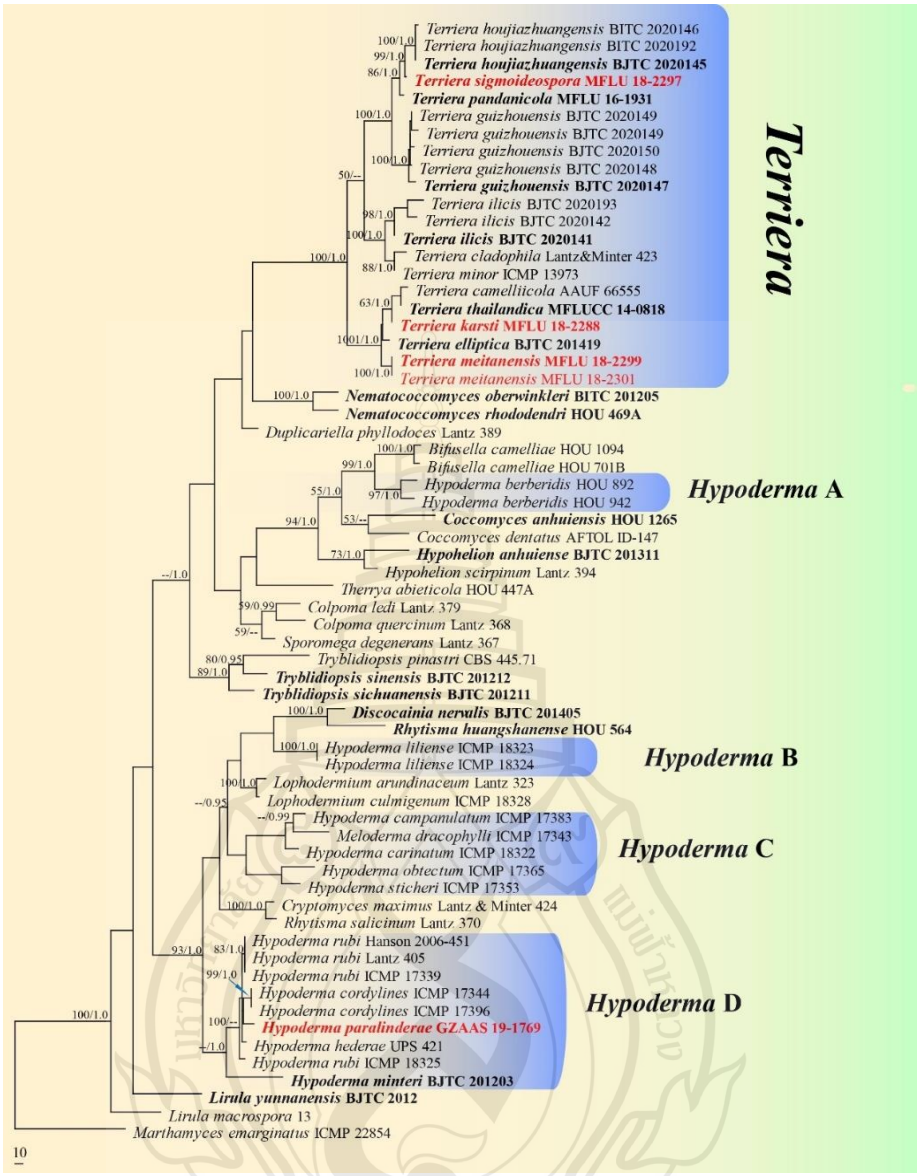
Species	Specimen/Strain No.	LSU	ITS	mtSSU
Meloderma dracophylli	ICMP 17343	HM140561	MH921871	HM143833
Nematococcomyces oberwinkleri	HOU 482B	KC312686	---	KC312689
Nematococcomyces rhododendri	HOU 469A	KC312687	KU213975	KC312691
Rhytisma huangshanense	Hou 564	FJ495192	GQ253101	---
Rhytisma salicinum	Lantz 370 (UPS)	HM140566	---	---
Rhytisma salicinum	Lantz 370 (UPS)	HM140566	---	---
Sporomega degenerans	Lantz 367 (UPS)	HM140567	---	HM143839
Sporomega degenerans	Lantz 367 (UPS)	HM140567	---	HM143839
Terriera camelliicola	L0447	KP878552	---	KP878553
Terriera camelliicola	L0447	KP878552	---	KP878553
Terriera cladophila	Lantz & Minter 423 (UPS)	HM140568	---	HM143840
Terriera elliptica	HOU327	KP878550	KP878549	KP878551
Terriera karsti	MFLU 18-2288	MN638881	MN638876	MN638871
Terriera meitanensis	MFLU 18-2299	MN638879	MN638874	MN638869
Terriera meitanensis	MFLU 18-2301	MN638880	MN638875	MN638870
Terriera minor	ICMP 13973	HM140570	---	HM143842
Terriera pandanicola	MFLU 16-1931	MH260320	MH275086	---
Terriera sigmoideospora	MFLU 18-2297	MN638882	MN638877	MN638872
Terriera thailandica	MFLUCC 14-0818	KX765301	---	---
Therrya abieticola	HOU447A	KP322580	KP322574	KP322587
Tryblidiopsis pinastri	CBS 445.71	MH871979	JF793678	AF431963
Tryblidiopsis sichuanensis	BJTC 201211	KC312683	KC312676	KC312692
Tryblidiopsis sinensis	BJTC 201212	KC312681	KC312674	KC312694

Note CBS: CBS-KNAW Fungal Biodiversity Centre, Utrecht, The Netherlands; GZAAS: Herbarium of Guizhou Academy of Agricultural Sciences, Guiyang, China; ICMP: International Collection of Microorganisms from Plants, New Zealand; MFLU: Hebarium of Mae Fah Luang University, ChiangRai, Thailand; MFLUCC: Mae Fah Luang University Culture Collection, ChiangRai, Thailand.

4.3 Results

4.3.1 Phylogenetic Analyses

The dataset for phylogenetic analysis comprised 64 strains, with *Marthamyces emarginatus* (Cooke & Massee) Minter selected as the outgroup taxon. This dataset consists of 2078 characters (including the gaps), of which 1205 are constant, 236 are variable parsimony-uninformative, while 637 characters are parsimony informative. The most parsimonious tree showed with length of 2843 steps (CI = 0.480, RI = 0.759, RC = 0.364 and HI = 0.520). The best tree revealed by the MP analysis was selected to represent relationships amongst taxa (Figure 4.1). The tree generated from Bayesian in-ference analyses had similar topology. The phylogram (Figure 4.1) shows that *Hypoderma* is non-monophyletic (Clade A, B, C and D), with *H. paralinderae* clusters with three existing species viz. *H. cordylines* P.R. Johnst., *H. hederae* (T. Nees ex Mart.) De Not. and *H. rubi* (Pers.) DC. In contrast, all of the *Terriera* species with available sequences (including the newly generated sequences) form a monophyletic clade with strong statistical support (MPBP 100% and BYPP 1.00). This corresponds to the phylogeny in Zhang et al. (2015). *Terriera meitanensis* and *T. karsti* group together with three reported species viz. *T. camelliicola* (Minter) Y.R. Lin & C.L. Hou, *T. elliptica* T.T. Zhang & C.L. Hou and *T. thailandica* Jayasiri & K.D. Hyde, while *T. sigmoideospora* is placed within another clade that comprises *T. houjiazhuangensis* C.L. Hou & S.R. Cai and *T. pandanicola* Tibpromma & K.D. Hyde.



Note Phylogenetic tree generated from a Maximum parsimony (MP) analysis based on LSU, ITS, and mtSSU sequences data of the family Rhytismataceae. And the tree is rooted with *Marthamyces emarginatus* ICMP 22854. Maximum parsimony bootstrap values equal or greater than 50% and Bayesian posterior probabilities equal or greater than 0.95 are indicated near the nodes. Type strains are indicated in bold and new sequences, generated in this study, are given in red.

Figure 4.1 Phylogenetic tree for the family Rhytismataceae

4.3.2 Taxonomy

Hypoderma De Not., G. bot. ital. 2(2): 13 (1847)

De Candolle (1805) introduced *Hypoderma* to accommodate taxa resembling *Hysterium* Pers., but with apothecia that are immersed in host-plant tissue and the hymenia are exposed via a longitudinal split in the substratum. Subsequently, the nomenclature of *Hypoderma* was challenged by various authors (Chevallier, 1822, 1826; Fries, 1823; Wallroth, 1833). De Notaris (1847) recognised the distinction between *Hypoderma* and *Lophodermium* Chevall. and separated them, based on the ascospore shapes. So far, there are 214 epithets included in Index Fungorum (2020), but around half of these species are synonymized under other genera, such as *Lophodermium*, *Meloderma* Darker and *Terriera*.

Hypoderma paralinderae J.F. Zhang & Z.Y. Liu, *sp. nov*.

Index Fungorum number: IF556909; Facesoffungi number: FoF06797; Figure 4.2

Etymology: Referring to the morphological similarity with *Hypoderma linderae*.

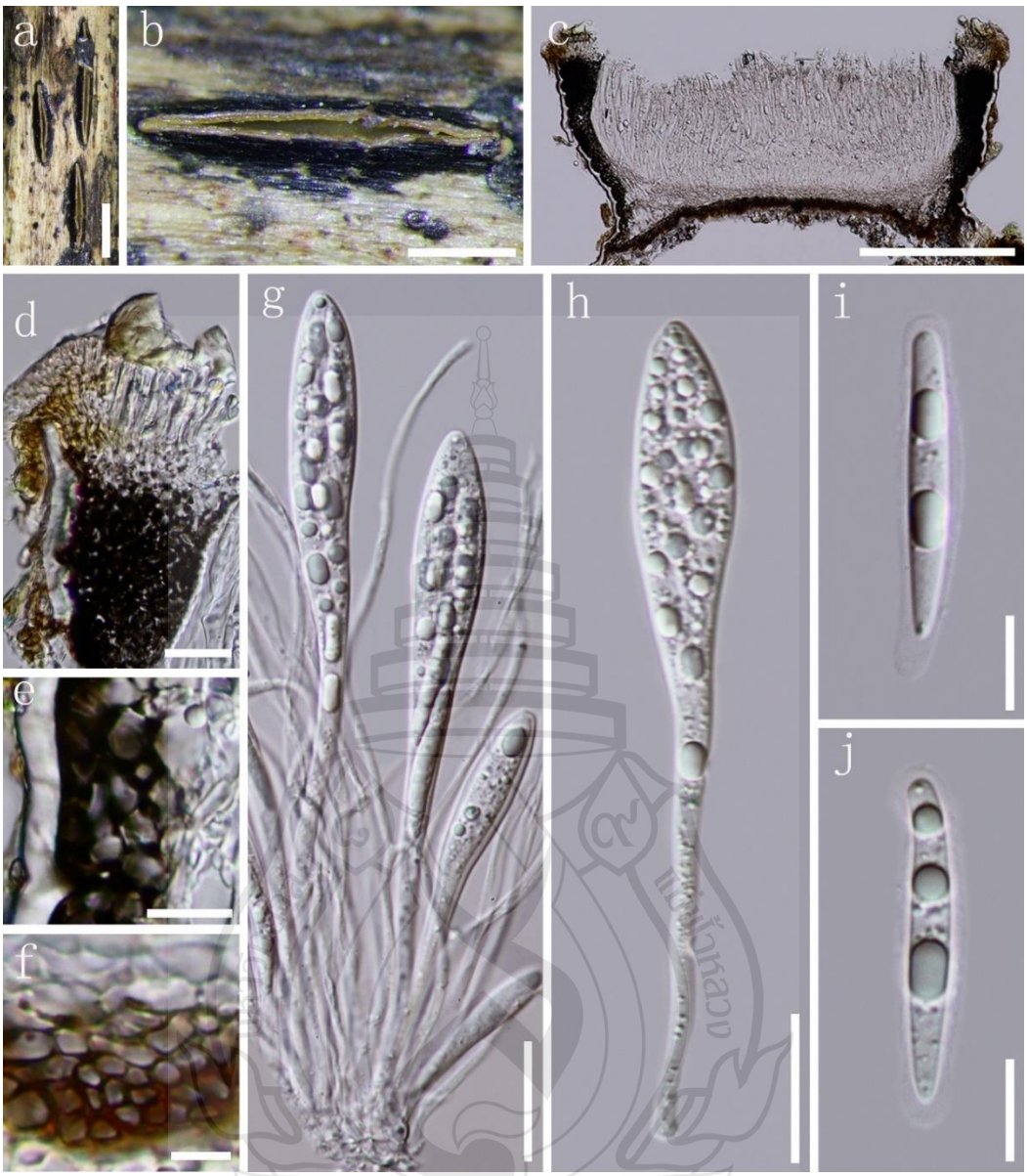
Holotype: GZAAS 19-1769.

Saprobic on dead stems of unidentified of herbaceous plants. Sexual morph: *Apothecia* developing on dead stems, scattered, dark brown to black, shiny, long elliptical to slightly fusiform, straight or somewhat curved, ends rounded or obtuse, rising above the surface of the substrate, opening by a single longitudinal split. *Lips* moderately developed, pale brown. In median vertical section, apothecia subcuticular, 200–280 µm deep. *Covering stroma* up to 38–45 µm thick near the opening, becoming to 12–18 µm thick towards the edges, extending to the basal stroma, consisting of an outer layer of host cuticle and several layers of dark brown, thick-walled cells of *textura angularis*. *Lip cells* clavate to cylindrical, 11–23 × 2–3 µm, thin-walled, hyaline to pale brown, 0–1-septate. *Basal stroma* 10–16 µm thick, consisting of several layers of brown, thick-walled cells, arranged in *textura angularis*, becoming colourless, thin-walled cells of *textura prismatica* towards the subhymenium. *Subhymenium* 19–27 µm thick, composed of several layers of hyaline, thin-walled cells of *textura angularis*. *Paraphyses* 1.5–2 µm, filiform, aseptate,

unbranched, often curved, but not swollen at the apex, anastomosing at the base. *Asci* (81.5–)110–120(–129) × 10–14 µm ($\overline{x}$ = 108 × 12 µm, n = 25), 8-spored, unitunicate, cylindrical-clavate, round to subtruncate at the apex, with a 38–49 µm long stalk, thin-walled, J-, apical ring, without circumapical thickening. *Ascospores* 26–32.5 × 2.5–4.5 µm ($\overline{x}$ = 30.5 × 3.5 µm, n = 35, measured without the gelatinous sheath), multi-seriate and mostly arranged in the upper half of ascus, fusiform to slightly cylindrical, straight or lightly curved, apex rounded and tapering slightly to an acute base, aseptate, hyaline, guttulate, surrounded by a 0.5–1.5 µm thick gelatinous sheath (extending to 2.5 µm at the poles). Asexual morph: Undetermined.

Material examined: CHINA, Guizhou Province, Leishan County, dead stems of unidentified herbaceous plants, 2 November 2017, J.F. Zhang, LS-21 (GZAAS 19-1769, holotype).

Notes: Our phylogenetic analysis shows that *Hypoderma paralinderae* is placed in *Hypoderma* D clade (Figure 4.1), and clustered with *H. cordylines*, *H. hederae* and *H. rubi*. Both *H. paralinderae* and *H. codylines* have similar sized asci (110−122.5 × 5.5−7 µm vs. 90−140 × 11−16 µm), however, they can be distinguished by the different shape and size of ascospores (fusiform to slightly cylindric, 26−32.5 × 2.5−4.5 µm in *H. paralinderae* vs. elliptic, 14−21 × 4.5−6 µm in *H. cordylines*) (Johnston, 1990). *Hypoderma paralinderae* shares similar sized asci with *H. hederae*, however, it is differentiated from the letter by larger ascospores (26−32.5 × 2.5−4.5 µm vs. 18−22 × 3.5−4 µm) (Powell, 1974). Moreover, *H. hederae* was described with oblong-cylindric ascospores that bluntly round on both ends, however, the ascospores in *H. paralinderae* are fusiform to cylindric, but rounded at the apex and tapering slightly to an acute base (Powell, 1974). While, *H. paralinderae* differs from *H. rubi* by having obviously larger asci (110−122.5 × 5.5−7 µm vs. 60−100 × 10−12.5 µm) and ascospores (26−32.5 × 2.5−4.5 µm vs. 14−18 × 3.5−4.5 µm) (Hou et al., 2007). Besides, the recommendations of delineation taxa from Jeewon & Hyde (2016) are followed, and comparisons of ITS gene region between *H. paralinderae* and *H. cordylines* (ICMP 17344), as well as *H. paralinderae* and *H. rubi* (ICMP 17339) are processed. The results showed that there are 9/468 bp (1.9%) and 9/467 (1.9%) bp differences (including gaps) between their, respectively. According to the above evidences, *H. paralinderae* is introduced herein as new to science.



Note a, b Apothecia observed under a dissecting microscope in face view. c Vertical section through an apothecium. d Lips adjacent to the top of covering stroma. e Section of covering stroma. f Section of basal stroma. g Paraphyses and asci in various states of maturity. h Immature ascus. i, j Ascospores. Note: c–j Mounted in water. Scale bar: a = 1 mm, b = 500 µm, c = 200 µm, d, g, h = 20 µm, e, i, j = 10 µm, f = 5 µm.

Figure 4.2 *Hypoderma paralinderae* (GZAAS 19-1769, holotype)

Terriera B. Erikss., Symb. bot. upsal. 19(no. 4): 58 (1970)

Terriera was segregated from *Lophodermium* by Eriksson (1970) with *T. cladophila* as its type species. Johnston (2001) elucidated some distinctive morphological features (described as oblong to sublinear ascomata with single longitudinal opening slit, narrow-cylindric asci and 1-septate ascospores that tapering slightly at both ends, and often becoming gently sigmoid on release and lacking a gelatinous sheath) for this genus and justified its monophyletic classification. There are 38 species accepted in *Terriera* (Index Fungorum 2020) and around half of these species were discovered recently from China (Chen et al., 2011, 2013; Yang et al., 2011; Zheng et al., 2011; Gao et al., 2012; Song et al., 2012; Zhou et al., 2012; Li et al., 2015a, b; Lu et al., 2015; Wu et al., 2015; Cai et al., 2020b). Here, we introduce three novel species. These three species share morphological characters typical of *Terriera* and cluster together with existing *Terriera* species in LSU-ITS-mtSSU phylogenetic analyses. In addition, a synopsis for *Terriera* species is also provided and listed in Table 4.2.

Terriera karsti J.F. Zhang & J.K. Liu, *sp. nov*.

Index Fungorum number: IF556901; Facesoffungi number: FoF06799; Figure 4.3

Holotype: MFLU 18-2288.

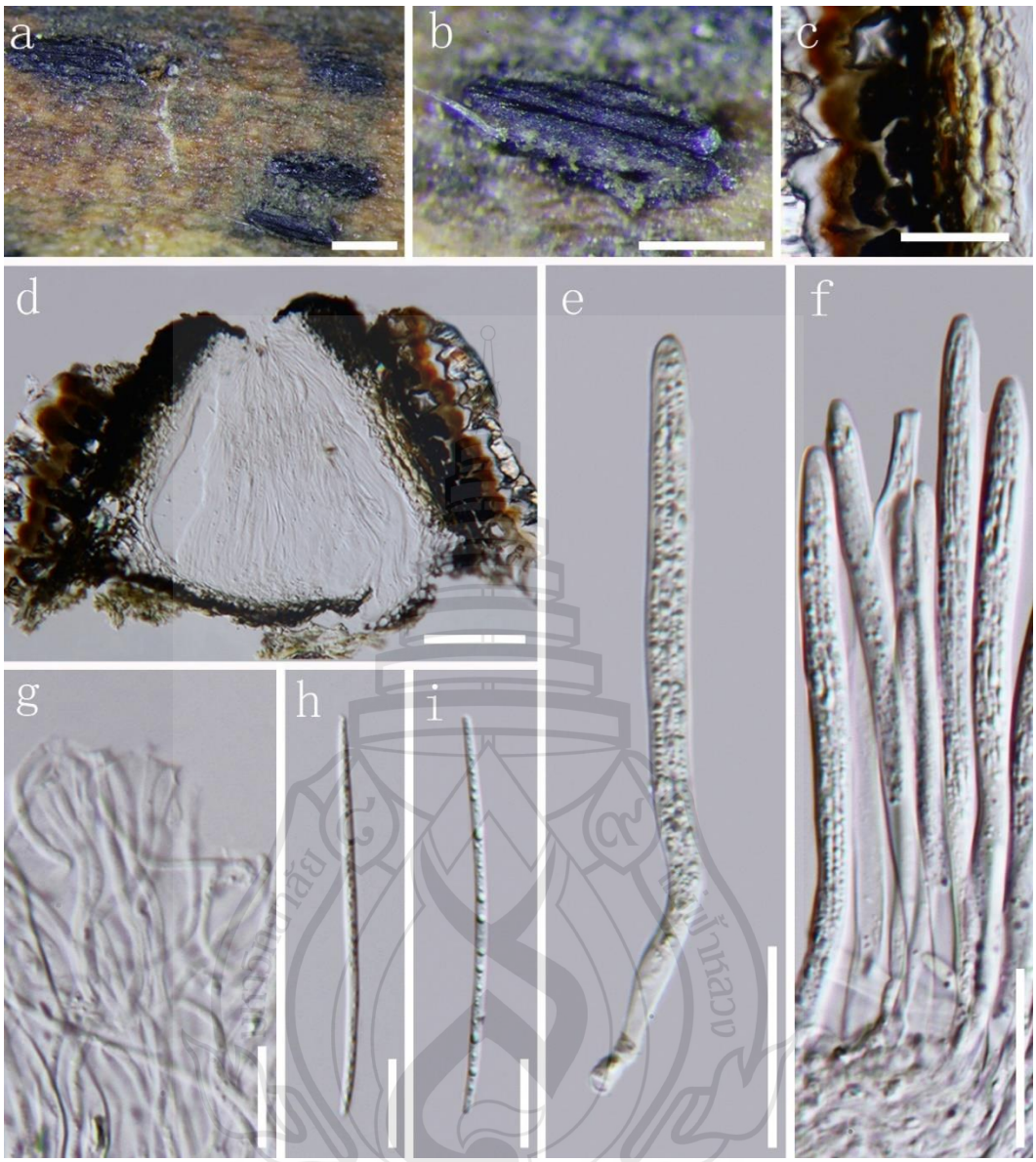
Etymology: Refers to the karst landscape where the holotype was collected.

Saprobic on dead branch of unidentified ligneous plants. Sexual morph: *Apothecia* developing on dead branch, elliptical or oblong-elliptical in outline, ends slightly acute to obtuse. *Apothecia* surface black, matt or slightly glossy, moderately raising the substratum surface, opening by a single longitudinal split that extends to the ends of the apothecium. *Lips* absent. In median vertical section, apothecia deeply embedded in host tissue, with host cells becoming filled with fungal tissue as the apothecium develops. *Covering stroma* 30–45 µm thick, composed of blackish brown to black, thick-walled cells of *textura angularis* towards the exterior, and several layers of pale to nearly hyaline, thin-walled cells towards the interior. Along the edge of the apothecial opening, there is a flattened, 12–20 µm thick extension adjacent to the covering stroma that is composed of strongly melanized tissue with no obvious cellular structure. *Basal stroma* 8–18 µm thick, dark brown or blackish brown

composed of angular to globose, thick-walled cells, 2.5−4 µm diam. A triangular space between the covering stroma and basal stroma consists of thin-walled, nearly hyaline to grey-brown cells arranged in *textura prismatica*. *Paraphyses* 1−2 µm, filiform, hyaline, septate, gradually swollen or branching once at the apex, embedded in gelatinous sheaths. *Asci* (103−)110−122.5 × 5.5−7 µm ($\overline{x}$ = 113 × 6 µm, n = 20), 8-spored, unitunicate, cylindric, long stalk, thin-walled, apex truncate to somewhat round, J-, without circumapical thickening. *Ascospores* 55−66 × 1.5−2.0 µm ($\overline{x}$ = 61 × 1.8 µm, n = 25), fascicle but not coiled, filiform, gradually tapering toward the ends, hyaline, aseptate, smooth-walled, straight or slightly curved, with lacking gelatinous sheath. Asexual morph: Undetermined.

Material examined: CHINA, Guizhou Province, Guiyang, Yunyan District, dead branch of unidentified ligneous plants, 6 May 2016, J.F. Zhang, SH-06 (MFLU 18-2288, holotype); *ibid*. (GZAAS 19-1720, isotype); ex-type living culture, GZCC 19-0047.

Notes: In the present study (Figure 4.1), *Terriera karsti* is phylogenetically close to *T. camelliicola* and *T. thailandica* with moderate support (MPBP 63% and BYPP 1.00). *Terriera karsti* is not significantly distinguished from *T. camelliicola* based only on morphological characters as they share similar sized asci (110−122.5 × 5.5−7 µm vs. 85−120 × 5.5−6.5 µm) and ascospores (55−66 × 1.5−2 µm vs. 50−70 × 1 µm) (Johnston, 2001). However, the ascospores of *T. camelliicola* are covered by a 0.5 µm wide gelatinous sheath, while this is not observed in *T. karsti* (Sharma, 1982). In order to clarify their affinity, the recommendations of species delineation from Jeewon & Hyde (2016) were followed, and the comparison of each gene region between these two taxa is processed and showed that there are 9/840 bp (1%) and 10/694 bp (14.4%) differences in LSU and mtSSU regions respectively. While, *T. karsti* can be easily differentiated from *T. thailandica* by its larger asci (110−122.5 × 5.5−7 µm vs. 80−105 × 3.4−6.6 µm) and ascospores (55−66 × 1.5−2 µm vs. 38−60 × 1−1.5 µm) (Hyde et al. 2016). A comparison of LSU gene region between these two taxa is also processed, and the result showed that there are 3/838 bp (base pair) difference. Based on phylogenetic analyses coupled with morphological distinctiveness, *Terriera karsti* is introduced herein as a new species.



Note a, b Apothecia observed under the dissecting microscope. c Detail of covering stroma in vertical section. d Vertical section through an apothecium. e, f Asci in various states of maturity. g Apices of paraphyses. h, i Ascospores. Note: c–i mounted in water. Scale bar: a = 1 mm, b = 500 µm, c, e, f = 20 µm, d = 100 µm, g, i = 10 µm.

Figure 4.3 *Terriera karsti* (MFLU 18-2288, holotype)

Terriera meitanensis J.F. Zhang & Z.Y. Liu, *sp. nov.*

Index Fungorum number: IF556900; Facesoffungi number: FoF06798; Figure 4.4

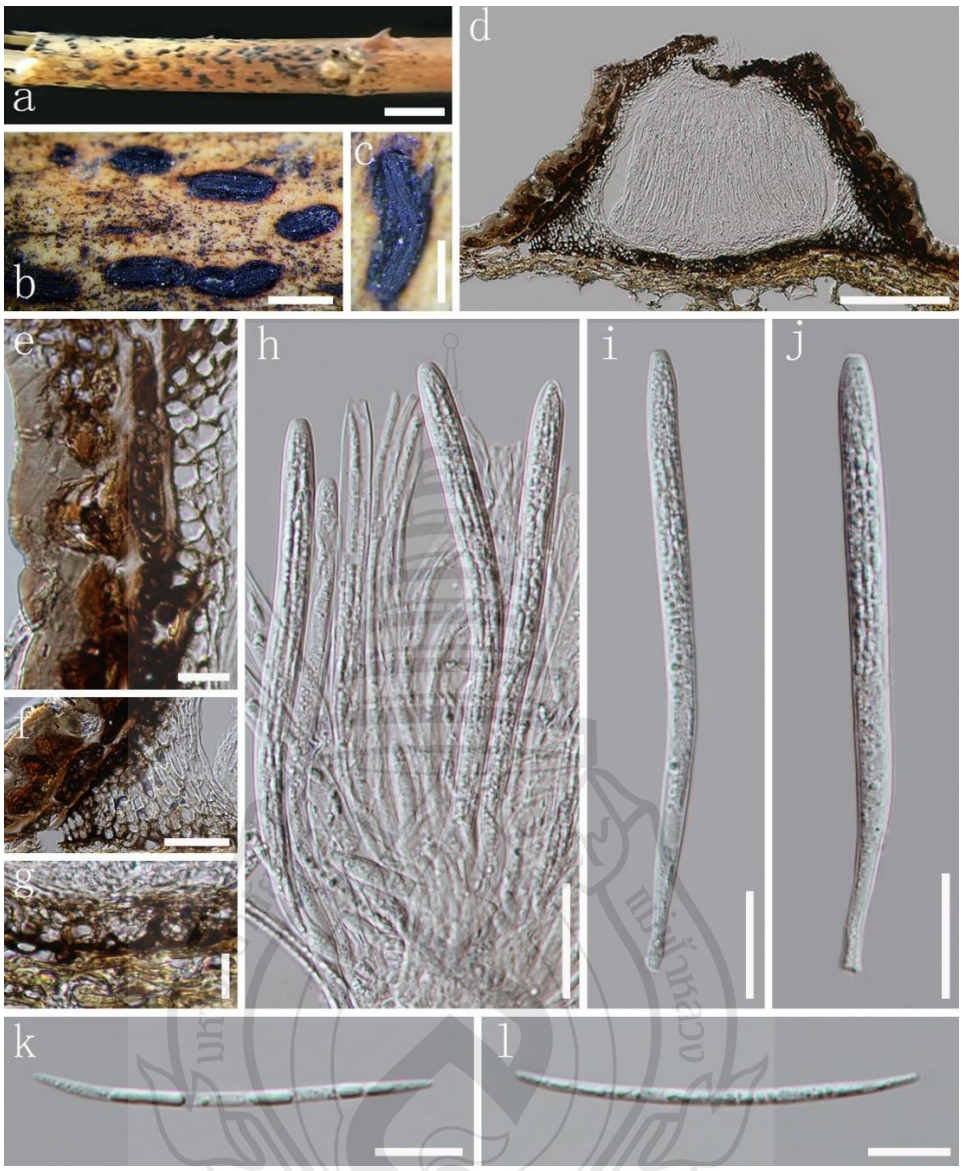
Holotype: MFLU 18-2299.

Etymology: Referring to the locality of the holotype, Meitan County, Guizhou Province, China.

Saprobic on dead stems of unidentified host. Sexual morph: *Apothecia* developing on dead stems, semi-immersed to superficial, elliptical or oblong-elliptical, ends slightly acute to obtuse, surface black, matt, raising the substratum surface, opening by a single longitudinal split that extends nearly the entire length. In median vertical section, apothecia deeply embedded in host tissue, with host cells becoming filled with fungal tissue as the apothecium develops. *Covering stroma* 33–42 µm thick, composed of blackish brown, thick-walled cells that are fused with host tissue in the outermost layers, becoming pale pigmented or nearly colorless towards the hymenium, thin-walled cells, arranged in *textura angularis* or *textura globulosa.* Along the upper edge of the apothecial opening, there is a flattened, 19–34 µm thick extension adjacent to the covering stroma that is composed of strongly melanized tissue with no obvious cellular structure. *Basal stroma* (Fig. 4g) 8–18 µm thick, dark-brown or blackish brown, composed of angular to globose, thick-walled cells, 2.5−4 µm diam. Where the covering stroma meets the basal stroma, there is a triangular-shaped, 35−60 µm thick, tissue composed of thin-walled, hyaline to pale brown cells forming a *textura prismatica*. *Subhymenium* 12–16 µm thick, consisting of hyaline *textura angularis* to *textura intricata*. *Paraphyses* 1−2 µm, filiform, hyaline, septate, gradually swollen or branching once at the apex, embedded in gelatinous matrix, anastomosing at the base. *Asci* (98.5−)113−125.5(−131.5) × 6−7.5 µm ($\bar{x}$ = 117 × 6.5 µm, n = 20), 8-spored, unitunicate, cylindric, somewhat long-stalked, thin-walled, apex generally truncate, J-, without circumapical thickening. *Ascospores* 47−54.5 × 1.5−2.5 µm ($\bar{x}$ = 50.5 × 2 µm, n = 35), fascicle, filiform, gradually tapering towards the ends, hyaline, aseptate, smooth-walled, straight or slightly curved, lacking a gelatinous sheath. Asexual morph: Undetermined.

Material examined: CHINA, Guizhou Province, Zunyi, Meitan County, dead stems of unidentified host, 28 August 2017, J.F. Zhang, MT-1 (MFLU 18-2299, holotype); *ibid*. (GZAAS 19-1731, isotype).

Notes: In our phylogenetic analysis (Figure 4.1), *Terriera meitanensis* is placed in a robust clade with *T. camelliicola*, *T. elliptica*, *T. karsti* and *T. thailandica* by strong statistical support (MPBP 100% and BYPP 1.00). *Terriera meitanensis* has larger asci than *T. camelliicola* and *T. thailandica*, while the ascospores of *T. meitanensis* are smaller (Johnston, 2001; Hyde et al., 2016). Both *T. meitanensis* and *T. karsti* share the similar sized asci, but *T. karsti* has larger ascospores (47−54.5 × 1.5−2.5 μm vs. 55−66 × 1.5−2.0 μm). *Terriera meitanensis* differs from *T. elliptica* by its obviously smaller asci (113−122.5 × 6−7.5 μm vs. 135−175 × 7−9 μm) and ascospores (47−54.5 × 1.5−2.5 μm vs. 60−85 × 1.5−2 μm) (Zhang et al. 2015). Moreover, the ascospores of *T. camelliicola* and *T. elliptica* are enveloped by a gelatinous sheath respectively, while this is not observed in *T. meitanensis*. In addition, the comparison of ITS gene region is processed between *T. meitanensis* and its closest species *T. elliptica* based on the recommendations from Jeewon & Hyde (2016), and the results showed that there are 15/489 bp (3%) differences. Therefore, we introduce *T. meitanensis* herein as a new species based on morphological and molecular evidences.



Note a Habit of apothecia on substrate. b, c Apothecia observed under the dissecting microscope in face view. d Vertical section through an apothecium. e Covering stroma. f Triangular space in section between the covering stroma and basal stroma. g Basal stroma. h Paraphyses with anastomoses among asci in various states of maturity. i, j Immature asci. k, l Ascospores. Note: d–l mounted in water. Scale bar: a = 1 cm, b = 1 mm, c = 500 µm, d = 100 µm, e, g, k, l = 10 µm, f = 30 µm, h–j = 20 µm.

Figure 4.4 *Terriera meitanensis* (MFLU 18-2299, holotype)

Terriera sigmoideospora J.F. Zhang & K.D. Hyde, *sp. nov.*

Index Fungorum number: IF556902; Facesoffungi number: FoF06800; Figure 4.5

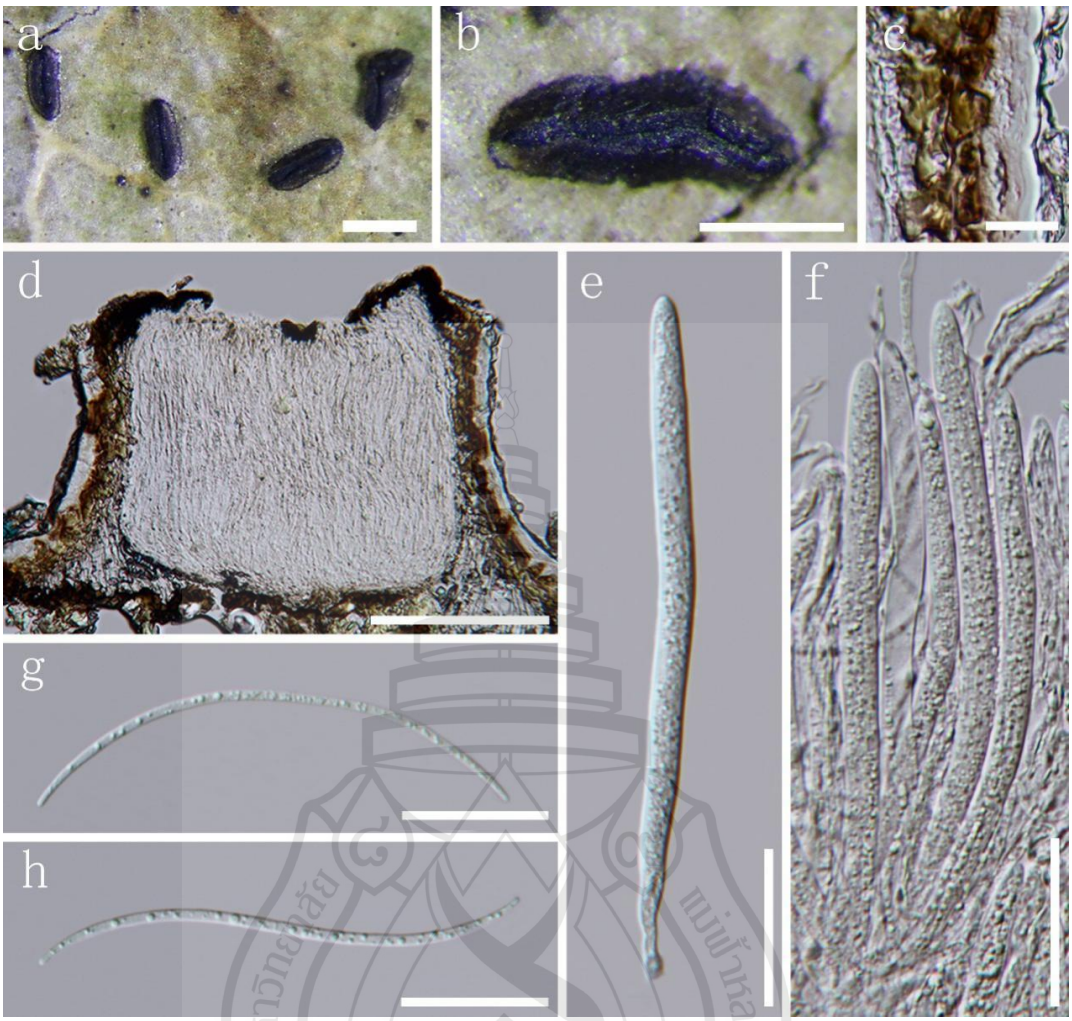
Holotype: MFLU 18-2297.

Etymology: Refers to its sigmoidal ascospores.

Saprobic on dead leaves of unidentified host. Sexual morph: *Apothecia* developing on fallen leaves, scattered, dark brown to black, matt, elliptical, sometimes 3-lobed or triangular, straight or slightly curved, ends rounded to subacute, strongly raising the surface of the substrate at maturity, opening by a single longitudinal split that extends almost the whole length of the apothecium. Immature apothecia appearing as a single dark brown protrusion, circular to slightly elongated. In median vertical section, apothecia 185–220 μm deep. *Covering stroma* 20–25 μm thick near the center of the apothecium, consisting of an outer layer of host cuticle, remains of epidermal and hypodermal cells filled with thick-walled, angular fungal cells, and an inner layer of *textura angularis* to *textura globulosa* with 4–7 μm diam., dark brown, thick-walled cells, slightly thinner towards the edges, extending to the basal stroma, but conspicuously thicker towards the apothecial opening, with a 15–27 μm thick extension comprising highly melanized tissue with no obvious cellular structure. *Excipulum* moderately developed, closely adhering to the covering stroma and the extension, arising from the marginal paraphyses, becoming thinner towards the base. *Basal stroma* concave, 12–15 μm thick, composed of dark brown, thick-walled, angular cells. A triangular space between the covering stroma and basal stroma is composed of thin-walled, colorless cells that vertically arranged in rows. *Subhymenium* 6–9 μm thick, flat, consisting of hyaline cells of *textura intricata*. *Paraphyses* filiform, hyaline, septate, gradually or suddenly swollen to 2.5 μm near the apex, covered by a thin gelatinous sheath, forming a 4–8 μm thick epithecium. *Asci* (93.5–)102–121 × 5–6 μm ($\bar{x}$ = 108.5 × 5.5 μm, n = 20), 8-spored, unitunicate, cylindric, apex tapering to round, thin-walled, J-, without circumapical thickening. *Ascospores* 79–95 × 1.5–2 μm ($\bar{x}$ = 89.5 × 1.9 μm, n = 30), fascicle, filiform, sigmoid, tapering slightly towards the ends, hyaline, aseptate, guttulate, gelatinous sheath not observed. Asexual morph: Undetermined.

Material examined: CHINA, Guizhou Province, Guiyang, dead leaves of unidentified host, 5 October 2016, J.F. Zhang, GZ-28 (MFLU 18-2297, holotype); *ibid*. (GZAAS 19-1729, isotype).

Notes: In the present phylogenetic analysis (Figure 4.1), *Terriera sigmoideospora* is placed within in *Terriera* and related to *T. houjiazhuangensis* C.L. Hou & S.R. Hou by strong statistical support (MPBP 99% and BYPP 1.00). *Terriera sigmoideospora* shares similar sized asci with *T. houjiazhuangensis* (102–121 × 5–6 μm vs. 103–128 × 4–6 μm) but has larger ascospores (79–95 × 1.5–2 μm vs. 73–82 × 0.6–0.9 μm) (Cai et al., 2020b). Besides, the ascospores of *T. houjiazhuangensis* are enveloped by an inconspicuous gelatinous sheath, while this is not observed in *T. sigmoideospora*. In addition, the comparisons of ITS gene region between these two taxa are processed and showed that there are 19/815 (2.3%) bp differences. *Terriera pandanicola* is sister to the above two taxa, however, it is significantly distinguished from *T. sigmoideospora* as its obviously smaller asci (50–66 × 4–5 μm vs. 102–121 × 5–6 μm) and ascospores (55–78 × 1–2 μm vs. 79–95 × 1.5–2 μm) (Tibpromma et al., 2018).



Note a, b Apothecia observed under the dissecting microscope. c Section of covering stroma. d Median vertical section through an apothecium. e Immature ascus. f Paraphyses and asci in various states of maturity. g, h Ascospores. Note: c–h mounted in water. Scale bar: a = 1 mm, b = 500 µm, c = 100 µm, d–h = 20 µm.

Figure 4.5 *Terriera sigmoideospora* (MFLU 18-2297, holotype)

Table 4.2 Synopsis of *Terriera* species. The new species described in this study are indicated in bold

Species	Host	Appearance of apothecia	Asci	Ascospores	Origin	References
Terriera aequabilis	On dead leaves of *Photinia villosa*	Elliptical to sub-circular, straight or slightly curved to one side, ends rounded, and opening by a single longitudinal split	75–105 × 4.5–5.5 µm	55–78 × 0.8–1 µm, filiform, aseptate, ends rounded, covered by a 0.3–0.5 µm wide gelatinous sheath	Jiangxi, China	Li et al. 2015b
T. angularis	On leaves of *Illicium simonsii*	Triangular to quadrangular, rarely elliptical, and opening by 3–4 radial splits or a longitudinal split	105−130 × 5.5−6.5 µm	70−90 × 1−1.2 µm, filiform, aseptate, slightly tapering towards the round base, covered by a 0.8−1 µm wide gelatinous sheath	Hubei, China	Zhou et al. 2013
T. arundinacea	On decomposed leaves of *Bambusa* sp.	Oblong to sublinear, and opening by a single longitudinal split	130−160 × 8−9 µm	90−100 × 2−2.5 µm, slightly tapering towards the base, lacking gelatinous sheath	Java, Indonesia	Johnston 2001

Table 4.2 (continued)

Species	Host	Appearance of apothecia	Asci	Ascospores	Origin	References
T. asteliae	On dead leaves of *Asterlia* sp.	Elliptical to oblong, ends rounded, opening by a single longitudinal slit	75−105 × 8−10.5 µm	45−70 × 2−2.5 µm, slightly tapering towards both ends and slightly constricted near the center, aseptate or 1-septate, gently curved, lacking gelatinous sheath	Northland, New Zealand	Johnston 2001
T. breve	On dead leaves of *Carex*, *Unicinia* and *Gahnia* spp.	Oblong-elliptical, ends rounded, often sublinear, with a single longitudinal opening slit	110−135(−160) × 6−7 µm	(55−)60−75 × 1.5−2 µm, slightly tapering towards both ends, aseptate or 1-septate, gently curved or sigmoid, lacking gelatinous sheath	Campbell I, New Zealand	Johnston 2001
T. camelliae	On fallen leaves of *Camellia* sp.	Subcircular to irregular bleached spots, elliptical or occasionally 3-lobed, and opening by a longitudinal split	85−120 × 5.5−6.5 µm	52−80 × 1−1.2 µm, filiform, aseptate, covered by a ca 0.5 µm wide gelatinous sheath.	Fuzhou, China	Chen et al. 2011

Table 4.2 (continued)

Species	Host	Appearance of apothecia	Asci	Ascospores	Origin	References
T. camelliicola	On twigs of *Camellia sinensis*	Elliptical, occasionally fusing to form elongated elliptical, opening by a single longitudinal split	80−110 × 5−7 µm	50−70 × 1 µm, filiform, aseptate, covered by a 0.5 µm wide gelatinous sheath.	Assam, India	Minter and Sharma 1982
T. breve	On dead leaves of *Carex*, *Unicinia* and *Gahnia* spp.	Oblong-elliptical, ends rounded, often sublinear, with a single longitudinal opening slit	110−135(−160) × 6−7 µm	(55−)60−75 × 1.5−2 µm, slightly tapering towards both ends, aseptate or 1-septate, gently curved or sigmoid, lacking gelatinous sheath	Campbell I, New Zealand	Johnston 2001
T. cladophila	On dead twigs of *Vaccinium myrtillus*	Elliptical, rounded at the ends, with a longitudinal opening split	75−100 × 5.5−8 µm	60−70 × 1 µm, filiform, aseptate, lacking gelatinous sheath	Norway	Terrier 1942; Eriksson 1970

Table 4.2 (continued)

Species	Host	Appearance of apothecia	Asci	Ascospores	Origin	References
T. clithris	On dead leaves of unidentified monocotyledon	Cylindric to linear, with longitudinal opening slit	110−120 × 6.5−7.0 µm	60−80 × 1−1.5 µm, slightly tapering towards both ends, lacking gelatinous sheath	Rio Grande Do Sul, Brazil	Johnston 2001
T. coacervata	On leaves of *Lithocarpus cleistocarpus*	Elliptical, sometimes branching into lobed or polygonal shapes, opening by a longitudinal split, or by more than 3 lobes	90−130 × 6.0−7.0 µm	60−110 × 1.5−1.8 µm, filiform, aseptate, covered by a 1.0−1.5 µm wide gelatinous sheath	Anhui, China	Zheng et al. 2012
T. dracaenae	On dead leaves or stems of *Dracaena* sp.	Oblong to oblong-elliptical, ends rounded, opening by a single longitudinal split	130−140 (−160) × 6−7 µm	100 × 2 µm, 1-septate, lacking gelatinous sheath	California, USA	Johnston 2001
T. elliptica	On living twigs of *Rhododendron* sp.	Elliptical, ends rounded to subacute, opening by a longitudinal split	135–175 × 7–9 µm	60–85 × 1.5–2 µm, filiform, slightly tapering towards both ends, aseptate, covered by a 1–1.5 µm wide gelatinous sheath	Yunnan, China	Zhang et al. 2015

Table 4.2 (continued)

Species	Host	Appearance of apothecia	Asci	Ascospores	Origin	References
T. fici	On dead leaves of *Ficus vasculosa*	Rounded or subrounded, with conspicuous edge, and opening by a single longitudinal split	90−115 × 4−5.5 µm	65−80 × 0.8−1 µm, filiform, aseptate, rounded to obtuse at the apex, slightly tapering towards the rounded or subacute base, covered by a 0.5 µm wide gelatinous sheath	Hainan, China	Wu et al. 2016
T. fuegiana	On dead leaves of *Rostkovia grandiflora*	Oblong elliptical to broad-elliptical, ends rounded, opening by a single, longitudinal slit	75–95 × 7–10 µm	60–65 × 1.5–2.5 µm, slightly tapering towards both ends, 1-septate, lacking gelatinous sheath	Tierra del Fuego, Argentina	Johnston 2001
T. fourcroyae	On dead leaves of *Furcraea* sp.	Oblong-elliptical, ends rounded, with a single longitudinal opening slit	95−110 × 5−6.5 µm	60–70 × 1.5–2.5 µm, slightly tapering towards both ends, gently coiled or sigmoid, 1-septate, lacking gelatinous sheath.	Sri Lanka	Johnston 2001

Table 4.2 (continued)

Species	Host	Appearance of apothecia	Asci	Ascospores	Origin	References
T. guizhouensis	On dead leaves of *Eriobotrya japonica*	Elliptical, occasionally curved, opening by a longitudinal split	88−10 7 × 4−6 µm	50−80 × 1−1.2 µm, filiform, slightly tapering towards both ends, aseptate, pluriguttulate, covered by a thin gelatinous sheath	Guizhou, China	Cai et al. 2020b
T. houjiashanensis	On dead leaves of *Ilex cornuta*	Elliptical, often curved, occasionally confluent, opening by a longitudinal split	103−1 28 × 4−6 µm	73−82 × 0.6−0.9 µm, filiform, slightly tapering towards both ends, aseptate, pluriguttulate, covered by an inconspicuous gelatinous sheath	Anhui, China	Cai et al. 2020b
T. huangshanensis	On leaves of *Eurya muricata* var. *huiana*	Elliptical, fusiform or subfusiform, straight or curved (lunate), sometimes 3-lobed or triangular, ends rounded to subacute, opening by a single longitudinal split	100−1 20 × 5−7 µm	58−90 × 1.5−2 µm, filiform, slightly tapering towards the base, aseptate, covered by a 1−1.5 µm thick gelatinous sheath	Anhui, China	Yang et al. 2011

Table 4.2 (continued)

Species	Host	Appearance of apothecia	Asci	Ascospores	Origin	References
T. ilicis	On dead leaves of *Ilex pernyi*	Elliptical, occasionally curved, triangular, or confluent, opening by a longitudinal split	117−139 × 4−7 µm	52−84 × ca. 1 µm, filiform, slightly tapering towards both ends, aseptate, pluriguttulate, covered by a thin gelatinous sheath	Hubei, China	Cai et al. 2020b
T. illiciicola	On dead leaves of *Lithocarpus cleistocarpus*	Subcircular to broad-elliptical, opening by a longitudinal split	90−135 × 4.0−5.0 µm	65−95 × 1 µm, filiform, aseptate, covered by an inconspicuous gelatinous sheath	Anhui, China	Zheng et al. 2011
T. intraepidermalis	On fallen leaves of *Photinia prunifolia*	Widely elliptical, sometimes elliptical or subcircular, occasionally triangular, straight or curved to one side slightly, ends round to obtuse, opening by a single longitudinal split or by three radial splits	90–135 × 5.5–7.5 µm	70–105 × 1–1.5 µm, with upper end rounded to obtuse, slightly tapering towards the rounded base, covered by a 0.5 µm wide gelatinous sheath	Hunan, China	Lu et al. 2015

Table 4.2 (continued)

Species	Host	Appearance of apothecia	Asci	Ascospores	Origin	References
T. javanica	On dead leaves of *Elettaria* sp.	Oblong-elliptical to sublinear, ends acute, opening by a single longitudinal slit	85–95 × 5.5–7 µm	50–60 × 1.5 µm, but the detail morphological characters were not seen	Java, Indonesia	Johnston 2001
T. karsti	**On dead branch of unidentified host**	**Elliptical or oblong-elliptical, ends slightly acute to obtuse, with a single longitudinal opening split**	**(103−)110−122 .5 × 5.5−7 µm**	**55−66 × 1.5−2.0 µm, filiform, gradually tapering towards both ends, aseptate, lacking gelatinous sheath**	**Guihzou, China**	**In this study**
T. latiascus	On dead leaves of *Euterpe* and *Heliconia* spp.	Oblong-elliptical, with a single longitudinal opening slit	80–95 × 7–8.5 µm	40–50 × 2–2.5 µm, with 1(–3)-septate, slightly tapering to both ends	Amazonas, Brazil	Johnston 2001
T. longissima	On dead leaves of Bambusaceae sp.	Oblong to sublinear, ends rounded, opening by a single, longitudinal slit	175–210 × 6–6.5 µm	Approximately 120–130 µm long, but the detail morphological characters were not seen	Potaro-Siparuni region VII, Guyana	Johnston 2001

Table 4.2 (continued)

Species	Host	Appearance of apothecia	Asci	Ascospores	Origin	References
T. nitens	On leaves of *Cyclobalanopsis myrsinifolia*	Suborbicular or broadly elliptical, straight or slightly curved, opening by a single longitudinal split	95–150 × 1–1.2 µm	68–115 × 0.8–1.2 µm, filiform, aseptate, round at the apex, slightly tapering towards the acute base, covered by a thin gelatinous sheath	Anhui, China	Chen et al. 2013
T. pandani	On dead leaves of *Pandanus* sp.	Oblong to oblong-elliptical, ends rounded, opening by a single longitudinal slit	100–120 × 5–6 µm	50–70 × 1–1.5 µm, lacking gelatinous sheath	San Juan, Puerto Rico	Johnston 2001
T. pandanicola	On dead leaves of *Pandanus* sp.	Elliptical, with rounded to subacute ends, opening by a longitudinal split	50–66 × 4–5 µm	55–78 × 1–2 µm, filiform, slightly tapering towards both ends, aseptate, lacking gelatinous sheath	Prachuap Khiri Khan, Thailand	Tibpromma et al. 2018

Table 4.2 (continued)

Species	Host	Appearance of apothecia	Asci	Ascospores	Origin	References
T. petrakii	On fallen leaves of *Smilax bracteata*	Elongate-elliptical, strongly curved or triangular, often coalesced, opening by a longitudinal split	85−110 × 4−5 µm	(60−)70−85 × 0.8 µm, filiform, aseptate, covered by a thin gelatinous sheath	Yunnan, China	Song et al. 2012
T. rotundata	On fallen leaves of *Quercus* sp.	Elliptical, occasionally triangular, ends rounded, opening by a longitudinal split or occasionally by teeth	90−120 × 4−5.5 µm	70−90(−95) × 0.8−1 µm, filiform, aseptate, lacking gelatinous sheath	Yunnan, China	Song et al. 2012
T. sacchari	On dead leaves and leaf bases of *Saccharum officinarum*	Narrow-oblong to sublinear, with a single longitudinal opening split	90−100 × 5−7 µm	50−60 × 1.5 µm, lacking gelatinous sheath	Hawaii, USA	Johnston 2001

Table 4.2 (continued)

Species	Host	Appearance of apothecia	Asci	Ascospores	Origin	References
T. samuelsii	On dead leaves of unidentified monocotyledon	Oblong to sublinear, ends rounded, opening by a single longitudinal slit	125−140 × 7−8 µm	(65−)75−90 × 2 µm, slightly tapering towards both ends, 1-septate, lacking gelatinous sheath	Amazonas, Brazil	Johnston 2001; 2003
T. sigmoideospora	**On dead fallen leaves of unidentified host**	**Elliptical, ends rounded to subacute, opening by a single longitudinal split**	**(93.5–)102–121 × 5–6 µm**	**79–95 × 5–2 µm, filiform, slightly tapering towards both ends, aseptate, lacking gelatinous sheath**	**Guizhou, China**	**In this study**
T. simplex	On fallen leaves of *Trachelospermum jasminoides*	Elliptical to ovate, ends obtuse, rounded or slightly acute, opening by a single longitudinal split which is sometimes branched in the triangular ascomata	72−95(−105) × 4.8−5.2 µm	(45−)56−82 × 1−1.2 µm, filiform, slightly tapering towards the rounded base, covered by a 0.8−1 µm wide gelatinous sheath	Anhui, China	Gao et al. 2012
T. stevensii	On dead leaves of *Vincentia* sp.	Oblong, ends rounded, opening by a single longitudinal slit	100−125 × 5−6 µm	60−80 × 1.5−2 µm, lacking gelatinous sheath	Hawaii, USA	Johnston 2001

Table 4.2 (continued)

Species	Host	Appearance of apothecia	Asci	Ascospores	Origin	References
T. thailandica	On dead branch of unidentified host	Elliptical, ends rounded to subacute, opening by a longitudinal split	80−105 × 3.4−6.6 µm	38−60 × 1−1.5 µm, filiform, slightly tapering towards both ends, aseptate, lacking gelatinous sheath	Chiang Rai, Thailand	Hyde et al. 2016
T. transversa	On dead leaves of *Pandanus* sp.	Elliptical or oblong-elliptical, ends slightly acute to obtuse, opening by a single longitudinal split	70−86 × 5−6 µm	45−68 × 1−1.2 µm, filiform, slightly tapering towards both ends, aseptate, covered by a 0.5 µm wide gelatinous sheath	Hainan, China	Li et al. 2015a

4.4 Discussion

The diversity of microfungi in many parts of the world is understudied. This is evident from the numerous new species being described from Asia and South America (Hyde et al., 2018, 2019a, 2020b). With this in mind, we are studying the fungi of the Karst regions in China and Thailand, where we are also finding numerous new species (Zhang et al., 2016, 2017b, c, 2018, 2019). Our study is contributing to the knowledge of fungal diversity in the region, where species may also have biotechnological potential (Hyde et al., 2019b). Additionally, as Rhytismataceae is a relatively poorly studied group, we report on one new species from *Hypoderma* and three new *Terriera* species illustrating the diversity and potential for new discoveries of these fungi in Asia.

Hypoderma, a large genus in Rhytismataceae, is a complicated group. There are only a few species in this genus with sequence data, but these have shown the group to be polyphyletic (Lantieri et al., 2011; Wang et al., 2013). This is also true of the phylogenies in this study (Figure 4.1). *Hypoderma* is morphologically similar to *Lophodermium*, and they mainly differ on the basis of ascospore shape as the former have elliptical to cylindric-fusiform ascospores, while the latter has filiform ascospores (Powell, 1974). However, there are no molecular studies that provide a natural classification for these two genera, even though more than 35 species have been synonymized under *Lophodermium* (Index Fungorum 2020). Fresh collections and molecular sequences are required to move toward a revision of these genera.

Terriera is one of the few genera in Rhytismataceae that can be considered a monophyletic group based on distinctive morphology and phylogenetic characterization (Zhang et al., 2015). Our molecular analyses corroborate this. However, there are only nine taxa with available sequences in GenBank, and most of *Terriera* species were established based only on morphological features (Yang et al., 2011; Gao et al., 2012; Song et al., 2012; Zhou et al., 2012; Chen et al., 2013; Li et al., 2015b; Lu et al., 2015; Zhang et al., 2015; Cai et al., 2020b). In the latest study (Cai et al., 2020b), *T. pandanicola* was distant from *Terriera* in ITS analysis, but included in this group on the basis of concatenated LSU-mtSSU sequence data. Cai et al. (2020b)

indicated that this taxon should be revised in a future study. Based on their suggestion, we checked the sequence data of *T. pandanicola*, and found that the ITS sequence of this species is misidentified as it not related *Terriera* even Rhytismataceae species in BLASTn results. However, the newly generated available sequences (ITS and mtSSU) of *T. pandanicola* have been uploaded in GenBank and included in our phylogenetic analysis, and the results indicated that it is a unique species in *Terriera* in the present study (Figure 4.1).

CHAPTER 5

OVERALL CONCLUSIONS

5.1 Ascomycetes from Karst landscapes of Guizhou Province, China

A systematic classification of ascomycetes from karst landscapes in Guizhou Province, China has never been reported. In this study, a large-scale collection of ascomycetes specimens from this area were identified and documented with morphological descriptions and illustrations.

A total of 76 ascomyctous fungal species were identified based on morphological characteristics and phylogenetic affinities. Among these taxa, 44 species were introduced as new to science, and one new order, 4 new families, as well as 12 new genera were proposed. The results from this study indicated that ascomycetes from karst formations of Guizhou Province are distributed in 3 classes Dothideomycetes, Sordariomycetes and Leotiomycetes. Dothideomycetes is the best representative group with 47 species, and the next common group is Sordariomycetes with 25 species, while the least group is Leotiomycetes with only 4 species. In addition, a high diversity of ascomycetes from this special habitat was presented in this study as 76 species were distributed in 15 orders, 36 families and 59 genera. And Pleosporales is the most diversity order among this groups with 36 species.

In the light of high ratio of new species were introduced in this study, it can be stated that there are still abundant undescribed species are hidden in the karst landscaps of Guizhou Province, China.

5.2 Resolve the Natural Classification Placement of Confused taxa

With the additional of fresh collections and sequence data, the natural classification of family Eriomycetaceae, and the genera *Neoleptosporella* and *Phragmogibbera* was resolved. Eriomycetaceae was resurrected based on morpho-phylogentic evidence to represent a non-

lichenized lineage and placed within Monoblastiales. The genus *Neoleptosporella* was taken up to a new family Neoleptosporellaceae and placed within Chaetosphaeriales, and *Phragmogibbera* were transferred to Leptosphaeriaceae and from Dothideomycetes genera *incertae sedis*.

5.3 Research Advantages

Prior to this study, a systematic classification of ascomycetes from karst landscapes of Guizhou Province has never been reported. This research documents the account of ascomycetes from this special habitat based on morphology and phylogeny. The results would provides a better understanding on diversity of ascomycetes from karst landscapes of Guizhou Province in China. Moreover, one new order, 4 new families, 12 new genera with 44 new species are introduced and contributed to the modern classification system. In addition, the natural of several confused taxa are resolved through this study.

5.4 Future Work

In this study, we only collected the specimens from Guizhou Province, China. However, there are lots of karst regions that widely distributed in different climate types around the world. Hence, extensive collection of ascomycetes from different karst landscapes is proposed for a better understanding on diversity of ascomycetes in this special habitat.

The samples that collected in this study are normally dead or decaying plant materials, and it is difficult to identify the host plant. And this led to some host-specific fungal species maybe overlooked and could not clearly recognized during this period. Hence, the ascomycetous fungal diversity on different hosts in this habitat are expected to explore.

There are one new order, 4 new families and 13 new genera are introduced in this study. However, most of these groups are monotypic, and more fresh collections and molecular data are needed to stabilize their placement in the future study.

5.5 Publications

Seven papers as first author, nine other SCI publications have been published as co-author.

List of publications

First author

1. **Zhang, J. F.,** Liu, J. K., Hyde, K. D., Ekanayaka, A. H., Liu, Z. Y. (2020). Morpho-phylogenetic evidence reveals new species in Rhytismataceae (Rhytismatales, Leotiomycetes, Ascomycota) from Guizhou Province, China. *Mycokeys*, *76*, 81–106. https://doi.org/10.3897/mycokeys.76.58465

2. **Zhang, J. F.,** Liu, J. K., Thambugala, K. M., Yang, J., Meng, Z. H., Liu, Z. Y. (2020). Two new species and a new record of *Nigrograna* (Nigrograna ceae, Pleosporales) from China and Thailand. *Mycological Progress*, *19*, 1365–1375. https://doi.org/10.1007/s11557-020-01633-0

3. **Zhang, J. F.,** Liu, J. K., Jeewon, R., Wanasinghe, D. N., Liu, Z. Y. (2019). Fungi from Asian Karst formations III. Molecular and morphological characterization reveal new taxa in Phaeosphaeriaceae. *Mycosphere*, *10*(1), 202–220. https://doi.org/10.5943/mycosphere/10/1/3

4. **Zhang, J. F.,** Liu, J. K., Ran, H. Y., Khongphinitbunjong, K., Liu, Z. Y. (2018). A new species and new record of *Lophiotrema* (Lophiotremataceae, Dothideomycetes) from karst landforms in southwest China. *Phytotaxa*, *379*(2), 169–179. https://doi.org/10.11646/phytotaxa.379.2.5

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APPENDICES

APPENDIX A

PUBLICATIONS

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DGfM

ORIGINAL ARTICLE

Two new species and a new record of *Nigrograna* (Nigrograna ceae, Pleosporales) from China and Thailand

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Abstract
Based on the morphological and phylogenetic analyses, four specimens, which were collected from China and Thailand respectively, are identified as two novel species *Nigrograna hydei* and *N. obtusispora* and a new record *N. magnoliae* in the present study. These share morphologic characters typical of *Nigrograna*, and cluster together with existing *Nigrograna* species in the phylogenetic analyses of a combined ITS-*TEF1-α* sequence dataset. Illustrations and comprehensive descriptions for these taxa are provided.

Keywords Biatriospora · Dothideomycetes · Multi-gene · Phylogeny · Taxonomy

Introduction

The genus *Nigrograna* was initially erected by De Gruyter et al. (2012) to accommodate an important human pathogenic species *Nigrograna mackinnonii* based on morphology and phylogeny. However, Ahmed et al. (2014) transferred *N. mackinnonii* to *Biatriospora* as it is phylogenetically close to the type species of *Biatriospora* (*B. marina*), and *Nigrograna* was therefore considered a synonym of *Biatriospora*. Jaklitsch and Voglmayr (2016) found that three newly collected taxa clustered with the above two *Biatriospora* species in their molecular study, but were morphologically and ecologically different from the type species of *Biatriospora*. Therefore, they established the family Nigrograna ceae to accommodate *Nigrograna*, with *N. mackinnonii* as the type species.

Members of *Nigrograna* are characterized by black ascomata, clavate, short pedicellate asci and pale to chocolate brown, asymmetric, fusoid to narrowly ellipsoid, septate ascospores. Currently, 16 species were reported according to Index Fungorum (September, 2020) and latest publication (Wanasinghe et al. 2020). Moreover, *Nigrograna*, the only genus in Nigrograna ceae, is also considered a geographically and ecologically diverse group as the species in this genus are reported as endophytes, saprobes, or pathogens, and widely distributed around the world (Jaklitsch and Voglmayr 2016; Kolařík et al. 2017; Tibpromma et al. 2017; Kolařík 2018; Zhao et al. 2018; Dayarathne et al. 2020; Mapook et al. 2020; Wanasinghe et al. 2020).

In this study, two novel species and a new record of *Nigrograna* were collected from China and Thailand. In order to explore their taxonomic placement and affinities with related species, the morphological characters along with genetic analyses based on combined ITS and *TEF1-α* sequences are provided.

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RESEARCH ARTICLE

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Morpho-phylogenetic evidence reveals new species in Rhytismataceae (Rhytismatales, Leotiomycetes, Ascomycota) from Guizhou Province, China

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Abstract

Karst formations represent a unique eco-environment. Research in the microfungi inhabiting this area is limited. During an ongoing survey of ascomycetous microfungi from karst terrains in Guizhou Province, China, we discovered four new species, which are introduced here as *Hypoderma paralinderae*, *Terriera karsti*, *T. meitanensis* and *T. sigmoideospora* placed in Rhytismataceae, based on phylogenetic analyses and morphological characters. Molecular analyses, based on concatenated LSU-ITS-mtSSU sequence data, were used to infer phylogenetic affinities. Detail descriptions and comprehensive illustrations of these new taxa are provided and relationships with the allied species are discussed, based on comparative morphology and molecular data.

Keywords

four new taxa, *Hypoderma*, karst formations, taxonomy, *Terriera*

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Fungi from Asian Karst formations III. Molecular and morphological characterization reveal new taxa in Phaeosphaeriaceae

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Abstract

An ongoing study on ascomycetes from Karst landforms in Guizhou Province, China yielded 6 new taxa that belong to the Phaeosphaeriaceae. These include 2 novel monotypic genera: *Hydeopsis* (with *H. verrucispora* sp. nov.) and *Pseudoophiosphaerella* (with *P. huishuiensis* sp. nov.), and two new species: *Hydeomyces pinicola* and *Leptospora hydei*. Multi-gene phylogenetic analyses of a concatenated LSU-ITS-SSU-TEF1-α sequence data were performed to infer their phylogenetic relationships. *Hydeopsis* is morphologically characterized by subglobose, papillate, dark brown to black ascomata, cellular pseudoparaphyses, clavate, bitunicate, fissitunicate asci and ellipsoidal to fusiform, hyaline to pale pigmented, 3-septate ascospores with a mucilaginous sheath. *Hydeopsis* is phylogenetically related to *Dactylidina* and *Phaeopoacea*, but can be recognized as a distinct lineage and these genera can also be distinguished based on morphology. *Pseudoophiosphaerella* is morphologically similar to *Ophiosphaerella* in having cylindric-clavate asci and scolecospores, but is phylogenetically distant from the latter and more closely related to *Dlhawksworthia*. Comprehensive illustrations and detailed descriptions of these taxa are provided, and relationships with allied genera or species are discussed based on comparative morphology and phylogenetic evidence.

Key words – 6 new taxa – Dothideomycetes – Multi-gene – Sexual morph – Phylogeny – Taxonomy

Introduction

Pleosporales is the largest order of the Dothideomycetes and comprise around 80 families with a quarter of all described dothideomycetous fungi (Kodsueb et al. 2006, Kirk 2008, Zhang et al. 2008, 2012, Hyde et al. 2013, Tanaka et al. 2015, Jaklitsch & Voglmayr 2016, Dai et al. 2017,

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A new species and new record of *Lophiotrema* (Lophiotremataceae, Dothideomycetes) from karst landforms in southwest China

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Abstract

During fungal diversity studies on ascomycetes from karst landforms in southwest China, two taxa were collected and morphologically identified as *Lophiotrema* species based on their immersed ascomata, crest-like ostioles, cylindrical, short pedicellate asci and fusiform ascospores. Multi-gene phylogenetic analyses based on a combined LSU-TEF1-α-SSU-ITS-RPB2 sequence data also confirmed their phylogenetic placement within the family Lophiotremataceae, where they clustered with reported *Lophiotrema* species with strong statistical support. *Lophiotrema hydei* sp. nov. is introduced and the species forms a distinct lineage which is sister to *L. neohysterioides*. Morphologically, *Lophiotrema hydei* differs from *L. neohysterioides* by its unique appearance of ascomata and different ascospore ornamentation. Another species is identified as *L. neohysterioides* based on morphological characteristics and phylogenetic data, and it is reported as the new record from China.

Keywords: 1 new taxon, Dothideomycetes, phylogeny, sexual morph, taxonomy

Introduction

We are carrying out surveys of ascomycetous fungi from karst landforms (Zhang *et al*. 2017a,b), and two *Lophiotrema* species were discovered from Guizhou Province, China. The family Lophiotremataceae was established as a monotypic family to accommodate the type genus *Lophiotrema* Sacc. by Hirayama & Tanaka (2011) based on the reevaluation of morphological features and DNA sequence based phylogenetic analyses of genera *Lophiotrema* and *Lophiostoma*. Members of Lophiotremataceae are normally characterized by subglobose to globose ascomata, beak-compressed, slit-like ostioles, cylindrical, short pedicellate asci, and fusiform to cylindrical, 1- to multi-septate, hyaline to brown ascospores, with or without an entire gelatinous sheath (Hirayama & Tanaka 2011). Only *Lophiotrema* was accepted in this family before 2016, subsequently Hashimoto *et al*. (2017) revised this group and introduced another five genera: *Atrocalyx* A. Hashim. & Kaz. Tanaka, *Crassimassarina* A. Hashim. & Kaz. Tanaka, *Cryptoclypeus* A. Hashim. & Kaz. Tanaka, *Galeaticarpa* A. Hashim. & Kaz Tanaka and *Pseudocrptoclypeus* A. Hashim. & Kaz Tanaka on the basis of morphological observations and phylogenetic analyses of multi-gene sequences data. This arrangement is followed by Wijayawardene *et al*. (2018).

Saccardo introduced the genus *Lophiotrema* and defined it as a group that comprised lophiostomataceous fungi, which have hyaline, multi-septate ascospores (Saccardo 1883). However, Chesters & Bell (1970) treated it as a synonym of *Lophiostoma* as they pointed out that both ascospore colour and number of transverse septa are not suitable for generic distinctions. Holm & Holm (1988) distinguished these two genera based on the differences in the type of peridium and asci. Hirayama & Tanaka (2011) separated *Lophiotrema* from *Lophiostoma* based on morphological traits and phylogenetic analyses, and established the family Lophiotremataceae to accommodate *Lophiotrema*. New taxa of *Lophiotrema* have since been reported as saprobes occurring on various plant substrates worldwide (Tanaka

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Two new species of *Dyfrolomyces* (*Dyfrolomycetaceae*, Dothideomycetes) from karst landforms

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Abstract

A survey of saprobic fungi yielded two new ascomycetes, *Dyfrolomyces maolanensis* and *D. thamplaensis*, which were isolated from dead branches collected from karst landforms of China and Thailand respectively. Both of them are characterized by immersed, clypeate, papillate ascomata, long cylindrical, short-pedicellate asci and fusiform, guttulate, 3-septate ascospores. Molecular analysis of combined LSU, SSU and *TEF1-a* sequence data placed them in the family *Dyfrolomycetaceae* with good support. Morphologically, they differ from other species of *Dyfrolomyces* by their ascospores, which are normally 3-septate, fusiform with acute or angular ends. And those reported *Dyfrolomyces* species were collected aquatic habitats, while our new species in this study were collected from terrestrial habitats. *Saccardoella aquatica* shares the similar morphology with these two new taxa, but can be distinguished in having its smaller asci and ascospores. Therefore, we introduce *D thamplaensis* and *D. maolanensis* here as new species and also transfer *S. aquatica* to *Dyfrolomyces* based on its similar morphological characters, as well as the molecular evidence of above two new taxa.

Key words: Phylogeny, *Saccardoella*, Taxonomy, Terrestrial habitats

Introduction

Saccardoella Speg. was introduced by Spegazzini (1879) and is characterized by large ascomata with erumpent papilla, long, cylindrical, unitunicate asci and uniseriate ascospores with transverse septa and usually with sheaths or polar appendages (Petrak 1962, Hyde 1992). There are 22 species epithets listed for the genus in Index Fungorum (March 2017) and most are from aquatic environments (Hyde 1992, Tsui *et al.* 1998, Fallah & Shearer 2001, Cai *et al.* 2002, Luo *et al.* 2004, Suetrong *et al.* 2009, Pang *et al.* 2013). The nature of the asci, which are neither clearly unitunicate nor bitunicate, resulted in confusion in the placement of the genus (Mathiassen 1989, Hyde 1992). Therefore, *Saccardoella* was placed in different families: *Clypeosphaeriaceae*, *Xylariales* (Barr 1989), *Pleurotremataceae* (Barr 1994, Maharachchikumbura *et al.* 2016), and 'Unitunicate *Ascomycota* genera *incertae sedis*' (Jones *et al.* 2009, 2015). However, Maharachchikumbura *et al.* (2016) revised the isotype of *Pleurotrema polysemum* and confirmed that the nature of its asci is unitunicate, but without any molecular evidence. Here, we follow the results of Pang *et al.* (2013) until the sequence data of *P. polysemum* is analysized.

Suetrong *et al.* (2009) investigated the phylogeny of marine Dothideomycetes and found that the molecular data placed *Saccardoella rhizophorae* in Dothideomycetes, but was not related to any existing genus or even families. Pang *et al.* (2013) introduced the family *Dyfrolomycetaceae* with *Dyfrolomyces tiomanensis* as the type species, and transferred three *Saccardoella* species (*S. mangrovei*, *S. marinospora*, *S. rhizophorae*) to *Dyfrolomyces* based on the phylogenetic analysis and morphological traits.

During collections of microfungi from karst landforms (Chen *et al.* 2017, Zhang *et al.* 2017), two *Dyfrolomyces* species were isolated from dead branches of an unidentified host, which were respectively obtained from the Maolan

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Fungi from Asian Karst formations II. Two new species of *Occultibambusa* (*Occultibambusaceae*, Dothideomycetes) from karst landforms of China

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Abstract

During an investigation of saprobic ascomycetes from karst landforms in southwest China, two new species were isolated from dead bamboo culms collected from Maolan Town in Guizhou Province. The new taxa share similar morphological characters as known *Occultibambusa* species in having immersed, papillate ascomata, broadly-cylindrical to clavate asci and fusiform, hyaline to brown ascospores. Phylogenetic analysis of combined LSU, SSU, TEF1-α and RPB2 sequence data also placed the new taxa within the genus *Occultibambusa* in the family *Occultibambusaceae* with good support. The new taxa can be distinguished from other species by septation and different-sized ascospores and the present or absence of sheaths. The new species, *Occultibambusa jonesii* and *O. maolanensis* are introduced here, with descriptions, illustrations and molecular data.

Key words—Dothideomycetes—phylogeny—Pleosporales—taxonomy

Introduction

We are carrying out the fungal diversity survey in the Karst formations of the Asian region and this is the second in a series of papers (Chen et al. 2017). The family of *Occultibambusaceae* D.Q. Dai & K.D. Hyde was introduced by Dai et al. (2017) and assigned to the order Pleosporales. The family is typified by *Occultibambusa* and characterized by immersed, solitary to gregarious ascomata, cylindrical to clavate, bitunicate asci and fusiform, hyaline to brown, septate ascospores and rather diverse asexual morphs. The family presently comprises four genera: *Neoocculitibambusa* Doilom & K.D. Hyde (Doilom et al. 2017), *Occultibambusa* D.Q. Dai, *Seriascoma* Phookamsak., D.Q. Dai & K.D. Hyde (Dai et al. 2017) and *Versicolorisporium* Sat. Hatak., Kaz. Tanaka & Y. Harada (Hatakeyama et al. 2008).

Species in this family occur on monocotyledons and hardwood trees, and share similar morphology with species of the genera *Bambusicola*, *Lophiostoma* and *Massarina* in having clavate asci and fusiform ascospores, however they can be distinguished readily via phylogenetic analysis

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Ligninsphaeria jonesii gen. et. sp. nov., a remarkable bamboo inhabiting ascomycete

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Abstract

A new ascomycete species, inhabiting decaying bamboo culms, is introduced in this paper based on morphology and molecular evidence. Analyses of partial LSU sequence data place the taxon in a monophyletic clade within the order *Pleosporales*. Morphological characters of the taxon are also distinct from other members of *Pleosporales*. *Ligninsphaeria jonesii* gen. et sp. nov. is therefore introduced to accommodate the new taxon, which is characterized by its clypeate, scattered, deeply immersed ascomata, with a slit-like opening, long ostioles, cellular pseudoparaphyses, clavate asci containing broad-fusiform and 1-septate ascospores surrounded by a gelatinous cap at both ends. The phylogenetic analyses show that the new taxon is phylogenetically close but distinct from the families *Amniculicolaceae* and *Testudinaceae*, and forms a distinct clade from other taxa in *Pleosporales*. The new taxon is described and compared with similar taxa. A new genus *Ligninsphaeria* (Pleosporales, *incertae sedis*) is introduced to accommodate this distinct lineage.

Key words: Dothideomycetes, New genus, Phylogeny, Pleosporales, Taxonomy

Introduction

The *Dothideomycetes* is the largest class of Ascomycota with approximately 23 orders, 105 families and 19000 species (Kirk *et al*. 2008, Ruibal *et al*. 2009, Hyde *et al*. 2013, Liu *et al*. 2014, McKenzie *et al*. 2014). This group is ecologically highly diverse, comprising saprobes, pathogens, endophytes, epiphytes, lichenized and rock-inhabiting fungi (Sterflinger *et al*. 1999, Selbmann *et al*. 2008, Schoch *et al*. 2009, Hyde *et al*. 2013). These fungi are characterized mainly by bitunicate asci with fissitunicate dehiscence and occur in terrestrial, freshwater and marine habitats worldwide (Kirk *et al*. 2008, Lumbsch & Huhndorf 2010, Hyde *et al*. 2013, McKenzie *et al*. 2014, Wijayawardene *et al*. 2014).

The *Pleosporales* is the largest order in the class *Dothideomycetes*, comprising 43 families, 332 genera and more than 4700 species (Kirk *et al*. 2008, Zhang *et al*. 2012, Hyde *et al*. 2013, Ariyawansa *et al*. 2013, Liu *et al*. 2013, 2014). Previous classification has posed major challenges in this complicated group, because of lack of molecular sequence data. In recent years, a great amount of molecular data has become available, with many molecular studies revising and classifying taxa (Mugambi & Huhndorf 2009, Zhang *et al*. 2009, Phookamsak *et al*. 2014, Liu *et al* 2015). However, some families lack sequence data and collections are needed to resolve these groups (Liu *et al*. 2014).

Studies on the taxonomy and biodiversity of microfungi inhabiting bamboo have revealed several new taxa over the past 15 years (Hyde *et al*. 2002, Cai *et al*. 2003, Zhou *et al*. 2003, Tanaka *et al*. 2009, Dai *et al*. 2012, 2014, 2015, Liu *et al*. 2013). Collection of fungi from decaying wood in Thailand, yielded a new taxon from bamboo culms collected on limestone outcrops near the Mae Chang Hot Spring in Chiang Rai Province, in northern Thailand. The morphological identification could not place the species in any existing genus or even family. Molecular analysis of LSU sequence data suggested this taxon as a monophyletic clade in the order *Pleosporales*. In this study we introduce *Ligninsphaeria jonesii* gen. et sp. nov. to accommodate this new taxon, which is placed in the order *Pleosporales*, genera *incertae sedis*.

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Didymella eriobotryae sp. nov. (Didymellaceae) and *Arthrinium arundinis* (Apiosporaceae) from fruit of *Eriobotrya japonica* (loquat) in China

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Abstract

Arthrinium (Apiosporaceae) and *Didymella* (Didymellaceae) are species-rich genera and most of the members in these genera are plant pathogenic, saprobic or endophytic species that have been recorded from a wide range of hosts. The present study introduces *Didymella eriobotryae sp. nov.* and a new record of *Arthrinium arundinis* which are associated with decaying fruit of *Eriobotrya japonica* in China. Morphology and DNA sequence data are employed to determine the taxonomic placement of these species.

Key words: new species, asexual morph, morphology, multi-gene phylogeny

Introduction

The family Apiosporaceae was established by Hyde *et al.* (1998) to accommodate *Apiospora* Sacc. *Appendicospora* K.D. Hyde, *Dictyoarthrinium* S. Hughes, *Endocalyx* Berk. and Broome, *Scyphospora* L.A. Kantsch. and *Spegazzinia* Sacc. However, Hyde *et al.* (1998) did not confirm the placement of *Appendicospora* in Apiosporaceae due to the absence of *Arthrinium* asexual morph in their cultures. Subsequently, Crous & Groenewald (2013) confirmed *Arthrinium* Kunze as the asexual morph of *Apiospora* based on morphology and DNA sequence data and *Apiospora* was reduced to synonymy with *Arthrinium*. Currently, Apiosporaceae is established as a well-defined family in the order Xylariales (Crous & Groenewald 2013, Senanayake *et al.* 2015, Maharachchikumbura *et al.* 2016), with six genera viz. *Appendicospora*, *Arthrinium*, *Dictyoarthrinium*, *Endocalyx*, *Scyphospora* and *Spegazzinia* (Maharachchikumbura *et al.* 2016, Wijayawardene *et al.* 2017, 2018). The genus *Arthrinium* was described by Schmidt & Kunze (1817) and currently the genus comprises an estimated number of 64 species (Wijayawardene *et al.* 2017). *Arthrinium* is a cosmopolitan fungal genus consists of endophytes, pathogens or saprobes associated with a wide range of substrates (Crous & Groenewald 2013, Sharma *et al.* 2014, Senanayake *et al.* 2015, Dai *et al.* 2016).

The family Didymellaceae is considered to be one of the largest families in Pleosporales, Dothideomycetes which includes plant pathogenic, saprobic and endophytic species associated with a wide range of hosts and substrates worldwide (Aveskamp *et al.* 2010, Hyde *et al.* 2013, Chen *et al.* 2015, 2017, Thambugala *et al.* 2017a, b). Didymellaceae was revised by Chen *et al.* (2015) based on multi-gene (ITS, LSU, RPB2 and β-tubulin) phylogenetic analyses combined with detailed morphological treatments and introduced nine new genera. Chen *et al.* (2015) accepted 17 genera in Didymellaceae, while Wijayawardene *et al.* (2018) treated the family with 27 genera. *Didymella* was established by Saccardo (1880) with *D. exigua* (Niessl) Sacc. as the type species and is one of the most species-rich genus in Didymellaceae. Chen *et al.* (2015) emended *Didymella* to accommodate *Peyronellaea* and several other phoma-like species that grouped together with the type species of *Didymella*. A number of *Didymella* species have been reported worldwide on a wide range of hosts and substrates (Aveskamp *et al.* 2010, Chen *et al.* 2015, 2017, Thambugala *et al.* 2017b).

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Refined families of Dothideomycetes: Dothideomycetidae and Pleosporomycetidae

Hongsanan S[1,2,6], Hyde KD[2,3,7], Phookamsak R[2,3,4,5,8,35], Wanasinghe DN[3,4,5,35], McKenzie EHC[9], Sarma VV[10], Boonmee S[2,11], Lücking R[12], Bhat DJ[13], Liu NG[2,14,15], Tennakoon DS[2,11], Pem D[2,11], Karunarathna A[2,3,4,6,51], Jiang SH[17], Jones EBG[18,19], Phillips AJL[20], Manawasinghe IS[2,7,21], Tibpromma S[3,4,5,35], Jayasiri SC[2], Sandamali DS[2,11,51], Jayawardena RS[2,11], Wijayawardene NN[22], Ekanayaka AH[2], Jeewon R[23], Lu YZ[24], Dissanayake AJ[16], Zeng XY[2,25,26,33], Luo ZL[27], Tian Q[2], Phukhamsakda C[2], Thambugala KM[28], Dai DQ[22], Chethana KWT[2,11], Samarakoon MC[2,48], Ertz D[29,30], Bao DF[2,6,27], Doilom M[3,4,5,35], Liu JK[16], Pérez-Ortega S[31], Suija A[32], Senwanna C[2,6], Wijesinghe SN[2,11,33], Konta S[2,11], Niranjan M[10,36], Zhang SN[2,6], Ariyawansa HA[34], Jiang HB[2,3,4,11], Zhang JF[2,37], Norphanphoun C[2], de Silva NI[3,6], Thiyagaraja V[2,3,4,6], Zhang H[38], Bezerra JDP[39], Miranda-González R[40], Aptroot A[41], Kashiwadani H[42], Harishchandra D[2,11,21], Sérusiaux E[43,] Aluthmuhandiram JVS[2,11,21], Abeywickrama PD[2,11,21], Devadatha B[2,10], Wu HX[44], Moon KH[45], Gueidan C[46], Schumm F[47], Bundhun D[2,6], Mapook A[2], Monkai J[2], Chomnunti P[11], Suetrong S[49,50], Chaiwan N[2,3], Dayarathne MC[33], Yang J[2,15], Rathnayaka AR[2,11,51], Bhunjun CS[2,11], Xu JC[3,4,5,35,] Zheng JS[1], Liu G[1], Feng Y[52], Xie N[1*]

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Refined families of Dothideomycetes: orders and families incertae sedis in Dothideomycetes

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Abstract

Numerous new taxa and classifications of Dothideomycetes have been published following the last monograph of families of Dothideomycetes in 2013. A recent publication by Honsanan et al. in 2020 expanded information of families in Dothideomycetidae and Pleosporomycetidae with modern classifications. In this paper, we provide a refined updated document on orders and families *incertae sedis* of Dothideomycetes. Each family is provided with an updated description, notes, including figures to represent the morphology, a list of accepted genera, and economic and ecological significances. We also provide phylogenetic trees for each order. In this study, 31 orders which consist 50 families are assigned as orders *incertae sedis* in Dothideomycetes, and 41 families are treated as families *incertae sedis* due to lack of molecular or morphological evidence. The new order, Catinellales, and four new families, *Catinellaceae*, *Morenoinaceae Neobuelliellaceae* and *Thyrinulaceae* are introduced. Seven genera (*Neobuelliella*, *Pseudomicrothyrium*, *Flagellostrigula*, *Swinscowia*, *Macroconstrictolumina*, *Pseudobogoriella*, and *Schummia*) are introduced. Seven new species (*Acrospermum urticae*, *Bogoriella complexoluminata*, *Dothiorella ostryae*, *Dyfrolomyces distoseptatus*, *Macroconstrictolumina megalateralis*, *Patellaria microspora*, and *Pseudomicrothyrium thailandicum*) are introduced base on morphology and phylogeny, together with two new records/reports and five new collections from different families. Ninety new combinations are also provided in this paper.

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Taxonomy and phylogeny of hyaline-spored coelomycetes

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Abstract
Coelomycete is a general term used for asexual fungi which produce conidia in fruiting bodies: pycnidial, acervular, cupulate, pycnothyria or stromatic conidiomata. The group contains numerous plant pathogenic, saprobic and endophytic species associated with a wide range of hosts. Traditionally, morphological characters and host associations have been used as criteria to identify and classify coelomycetes, and this has resulted in a poor understanding of their generic and species boundaries. DNA based taxonomic studies have provided a better outlook of the phylogenetic and evolutionary trends in coelomycetes. However, the present outcomes represent only a preliminary step towards the understanding of coelomycetes. Many genera have not been revisited since they were first described. The present study revises the classification of the hyaline-spored coelomycetes and provides a modern taxonomic framework based on both morphology and phylogeny. In total, 248 genera were investigated, of which less than 100 are known to have sequence data. Multi-locus sequence data analyses of 28S nrDNA, 18S nrDNA, ITS, RNA polymerase II second largest subunit (*rpb2*), and part of the translation elongation factor 1-alpha gene (*tef1*) and β-tubulin (*tub2*) gene regions were analysed. As a result, three new genera and 23 new species are introduced. In addition, three new links between sexual and asexual genera are provided. There are 138 genera that lack sequence data, and these are treated as Ascomycota, genera *incertae sedis*. Line drawings and descriptions are provided based on the examination of types and fresh collections and on the literature.

Keywords 26 new taxa · Ascomycota · Asexual morphs · Basidiomycota

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Fungal diversity notes 1036–1150: taxonomic and phylogenetic contributions on genera and species of fungal taxa

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Abstract
This article is the tenth series of the Fungal Diversity Notes, where 114 taxa distributed in three phyla, ten classes, 30 orders and 53 families are described and illustrated. Taxa described in the present study include one new family (viz. Pseudoberkleasmiaceae in Dothideomycetes), five new genera (*Caatingomyces*, *Cryptoschizotrema*, *Neoacladium*, *Paramassaria* and *Trochilispora*) and 71 new species, (viz. *Acrogenospora thailandica*, *Amniculicola aquatica*, *A. guttulata*, *Angustimassarina sylvatica*, *Blackwellomyces lateris*, *Boubovia gelatinosa*, *Buellia viridula*, *Caatingomyces brasiliensis*, *Calophoma humuli*, *Camarosporidiella mori*, *Canalisporium dehongense*, *Cantharellus brunneopallidus*, *C. griseotinctus*, *Castanediella meliponae*, *Coprinopsis psammophila*, *Cordyceps succavus*, *Cortinarius minusculus*, *C. subscotoides*, *Diaporthe italiana*, *D. rumicicola*, *Diatrypella delonicis*, *Dictyocheirospora aquadulcis*, *D. taiwanense*, *Digitodesmium chiangmaiense*, *Distoseptispora dehongensis*, *D. palmarum*, *Dothiorella styphnolobii*, *Ellisembia aurea*,

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APPENDIX B

CHEMICAL REAGENTS AND MEDIA

1. India ink used for observing mucilaginous sheath or appendage of ascospores

2. Lactoglycerol used for maintaining semipermanent slides

Lactic acid	10 ml
Glycerol	10 ml
Distilled water	10 ml

Mix 10 ml lactic acid, 10 ml glycerol, and add 10 ml distilled water.

3. Lactophenol-Cotton Blue highlight fungal structures for viewing with the compound light microscope. Cotton blue is the most popular stain for observe pseudoparaphyses, septate or ascus walled. This is gives excellent clarity and is also suitable for most fungal groups

Phenol (crystals)	20 g
Lactic acid	16 ml
Glycerol	31 ml
Cotton Blue	0.05 g
Distilled water	20 ml

Dissolve phenol in distilled water, add lactic acid, glycerol and 0.05 g of Poirrier's (cotton) blue or acid fuchsin.

4. Melzer' s Reagent is a good general mounting medium that clears the material somewhat and allows particularly brilliant resolution with a microscope, and used for identification of ascomycete fungi. Amyloid reaction of asci changed to blue or heavily purple colours.

Chloral hydrate	100 g
Potassium iodide	5 g
Iodine	1.5 g
Distilled water	100 ml

5. Malt Extract Agar (MEA) used for fungal cultivation

Agar	20 g
Malt extract	20 g
D-glucose	20 g
Peptone	6 g
Distilled water	1000 ml

Suspend 66 g of Malt extract agar in 1000 ml of distilled water and mix thoroughly. Heat with frequent agitation and boil to completely dissolve the power. Autoclave at 121 ℃ for 15 minutes.

6. Potassium hydroxide (KOH) used in the rehydration of dried specimens. 5% aqueous solution

7. Potato Dextrose Agar (PDA) used for fungal cultivation

Potato Infusion	4 g
Dextrose	20 g
Agar	15 g
Distilled water	1000 ml

Suspend 39 g of Potato dextrose agar in 1000 ml of distilled water and mix thoroughly. Heat with frequent agitation and boil to completely dissolve the power. Autoclave at 121 ℃ for 15 minutes.