



Article

Insight into the Taxonomic Resolution of the Pleosporalean Species Associated with Dead Woody Litter in Natural Forests from Yunnan, China

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Abstract: In the course of investigating the systematics of woody litter micromycete associates in Yunnan Province, China, we found one new species in *Phaeoseptaceae*, one new genus and three new species in *Sulcatissporaceae* from 16 specimens collected (ten collections of ascomycetous teleomorphs, four collections of hyphomycetous and two collections of coelomycetes anamorphs) from Ailaoshan, Chuxiong, Diqing, Honghe, Kunming, Lancang, Mengla and Yuxi in Yunnan Province. These taxonomic novelties were recognized with the aid of morphological comparisons and phylogenetic analyses of multiple gene sequences (non-translated loci and protein-coding regions). *Pleopunctum menglaense* sp. nov. is accommodated in *Phaeoseptaceae* (*Pleosporales*) based on its hyphomycetous anamorph, which is characterized by superficial sporodochia on the host surface, macronematous, mononematous, cylindrical, unbranched, aseptate, hyaline and smooth-walled conidiophores, monoblastic, terminal, hyaline conidiogenous cells, hyaline, muriform α conidia, and brown, muriform β conidia with tri-lobed wing like basal cells. *Kazuakitanaka* gen. nov. (type: *K. yuxiensis*) is introduced in *Sulcatissporaceae* (*Massarineae*, *Pleosporales*) for a saprobic ascomycete with teleomorphic and anamorphic (coelomycetous) features. The teleomorph possesses globose to subglobose ascomata with acentric ostiole, a peridial wall of textura angularis to textura prismatica, cylindrical-clavate, pedicellate asci with an ocular chamber, and 1–2-septate, hyaline, fusiform, guttulate ascospores with a distinct mucilaginous sheath. The anamorph features pycnidial conidiomata, phialidic, ampulliform to cylindrical, hyaline conidiogenous cells and ampulliform to cylindrical, one-to-three-septate, hyaline, guttulate conidia. *Loculosulcatisspora* was known only from its anamorph of *L. thailandica*. We observed the teleomorph of *Loculosulcatisspora hongheensis* sp. nov. and amended the generic description of *Loculosulcatisspora* accordingly. *Loculosulcatisspora hongheensis* is characterized by globose to subglobose ascomata with a central ostiole, a peridial wall of textura angularis to globosa, branched, septate, pseudoparaphyses, clavate asci with a short pedicel and a minute ocular chamber and hyaline, fusiform, 1-septate ascospores with a thick irregular mucilaginous sheath. This study provides some insights into the diversity of fungi on dead woody litter in terrestrial habitats.

Keywords: *Ascomycota*; anamorph; *Dothideomycetes*; Greater Mekong Subregion; teleomorph



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1. Introduction

Despite estimates that ~2.2–3.8 million or more fungal species exist on Earth [1], we know of only 146,150 [2], suggesting that 96% of fungal species remain unknown [3]. Some

fungus groups are well researched because of their impact on human lives, while others remain seriously neglected. This could potentially distort our understanding of fungal diversity [4]. Owing to their abundance across ecosystems, the importance of fungi cannot be discounted in any region [5]. There are numerous understudied habitats that harbor abundant species, and if they were to be thoroughly studied, many new species could be found [6]. Most species of plant-associated fungi can be pathogens, endophytes, saprobes or epiphytes on a wide range of hosts that reside in terrestrial as well as aquatic habitats [7]. Particularly, microfungi play important functional roles in almost all ecosystems, functioning as decomposers that degrade dead organic materials and recycle nutrients for reuse in the ecosystem [8,9]. Given the omnipresent nature of microfungi, additional taxonomic and ecological knowledge is prerequisites to understanding microfungi biology and their environmental significance.

Ascomycota is the most species-rich phylum of *Fungi*, comprising more than 92,700 species [2], and *Dothideomycetes* is the largest and most ecologically diverse class of the phylum [10]. This class comprises saprobes, human and plant pathogens, endophytes, epiphytes, ectomycorrhizal, lichens, lichenicolous, nematode-trapping and rock-inhabiting members [11]. In the *Dothideomycetes*, *Pleosporales* is the most species-rich order, consisting of 10,142 species [2] recognized in more than 90 families and 650 genera [12]. In recent years, molecular studies coupled with morphological evidence have revealed numerous novel families, genera and species within *Pleosporales* [12]. Recently, many new woody litter pleosporalean lineages from terrestrial [5,11,13–19] or freshwater [20–25] environments have been reported in Yunnan Province, China.

At the Center for Mountain Futures (Kunming Institute of Botany), we are investigating the diversity of microfungi on woody litter [3,15–17,26,27]. During this survey of lignicolous ascomycetes in Yunnan, we encountered numerous undescribed pleosporalean fungi. This study aimed to document taxonomic novelties in *Phaeoseptaceae* and *Sulcatisporaceae*. We used sequence data from relevant members of *Pleosporales*, three functional ribosomal RNA genes, the small and large subunit of the nuclear ribosomal RNA (nc18S and nc28S), the internal transcribed spacer region (ITS) and two protein-coding genes, the second largest subunit of RNA polymerase II (*rpb2*) and translation elongation factor 1-alpha gene (*tef1- α*) to perform an in-depth phylogenetic study. We present morphological and molecular phylogenetic evidence that supports the recognition of a new species in *Phaeoseptaceae* and four new species in *Sulcatisporaceae*, including a new genus.

2. Materials and Methods

2.1. Isolates and Specimens

During our fieldwork across the Ailaoshan, Chuxiong, Diqing, Honghe, Kunming, Lancang, Mengla and Yuxi regions of Yunnan Province, China, typical black ascomata/conidiomata appearing on dead twigs were collected during both dry (January, March and April) and wet (May, June and July) seasons. In total, sixteen specimens were included in this study. The collected samples were placed in Ziploc bags and taken to the mycology laboratory of the Kunming Institute of Botany and stored inside paper envelopes. Single spore isolation was conducted, and germinated spores were handled in accordance with the methods described in Wanasinghe et al. [15]. Dried specimens (at room temperature) were preserved in the fungarium of the Cryptogams Kunming Institute of Botany, Academia Sinica (KUN-HKAS). Representative cultures were deposited in the Kunming Institute of Botany Culture Collection (KUMCC), Kunming, China and China General Microbiological Culture Collection Center (CGMCC). Nomenclatural data of fungal novelties were deposited in MycoBank [28].

2.2. DNA Extraction, PCR Amplifications and Sequencing

The extraction of genomic DNA was performed using fresh mycelia in accordance with the methods of Wanasinghe et al. [5], using the Biospin Fungus Genomic DNA Extraction Kit-BSC14S1 (BioFlux, Shanghai, China) and following manufacturer guidelines. When

cultures could not be maintained for certain collected samples, DNA was extracted directly from the fruiting bodies of the fungus as outlined by Wanasinghe et al. [29]. The reference DNA for the polymerase chain reaction (PCR) was stored at 4 °C for regular use and stored at –20 °C for long-term storage.

Primers ITS5/ITS4 [30], LR0R/LR5 [31,32], NS1/NS4 [30], EF1-983F/EF1-2218R [33,34] and fRPB2-5f/fRPB2-7cR [35] were used to amplify the DNA sequences of the internal transcribed spacers (ITS), partial 28S large subunit rDNA (LSU), partial 18S small subunit rDNA (SSU), translation elongation factor 1- α (*tef1*), and RNA polymerase II second largest subunit (*rpb2*). Protocols used for PCR amplification (SSU, LSU, ITS and *tef1*) were used as in Wanasinghe et al. [15]. PCR amplification conditions of *rpb2* were set as initial denaturation at 98 °C for 2 min, followed by 35 cycles of denaturation at 98 °C for 10 s, annealing at 52 °C for 10 s and extension at 72 °C for 20 s, with a final extension step at 72 °C for 5 min. The amplified PCR fragments were then sent to a private company for sequencing (BGI, Ltd., Shenzhen, China).

2.3. Molecular Phylogenetic Analyses

2.3.1. Sequencing and Sequence Alignment

BLAST searches using the BLASTn algorithm were performed to retrieve similar sequences from GenBank (<http://www.ncbi.nlm.nih.gov>, accessed on 29 December 2021) and relevant publications [36,37]. The collection/strain numbers for these sequences (Table 1) are presented in the corresponding phylogenetic trees (Figures 1 and 2). All alignments were produced with the server version of MAFFT v.7 [38], checked and refined using BioEdit v.7.0.5.2 software [39].

Table 1. Taxa used in the phylogenetic analyses and their corresponding GenBank numbers. Isolates/sequences in bold were isolated/sequenced in the present study.

Species	Strain ¹	GenBank Accession Numbers				
		SSU	LSU	ITS	<i>tef1</i>	<i>rpb2</i>
<i>Alfoldia vorosii</i>	CBS 145501 ^T	MK589346	MK589354	JN859336	MK599320	-
<i>Amorocoelophoma cassiae</i>	MFLUCC 17-2283 ^T	NG_065775	NG_066307	NR_163330	MK360041	MK434894
<i>Angustimassarina acerina</i>	MFLUCC 14-0505 ^T	NG_063573	KP888637	NR_138406	KR075168	-
<i>Angustimassarina populi</i>	MFLUCC 13-0034 ^T	NG_061204	KP888642	KP899137	KR075164	-
<i>Angustimassarina quercicola</i>	MFLUCC 14-0506 ^T	NG_063574	KP888638	KP899133	KR075169	-
<i>Anthosulcatispora brunnea</i>	MFLU 18-1393 ^T	-	MH644791	MH644792	-	-
<i>Anthosulcatispora subglobosa</i>	MFLUCC 17-2065 ^T	MT214592	MT214592	MT310636	MT394649	MT394706
<i>Bambusicola bambusae</i>	MFLUCC 11-0614 ^T	JX442039	JX442035	JX442031	KP761722	KP761718
<i>Bambusicola didymospora</i>	MFLUCC 10-0557 ^T	KU872110	KU863105	KU940116	KU940188	KU940163
<i>Bambusicola didymospora</i>	MFLUCC 15-0189	KU872111	KU863106	KU940117	KU940189	KU940164
<i>Bambusicola irregulispota</i>	MFLUCC 11-0437 ^T	JX442040	JX442036	JX442032	KP761723	KP761719
<i>Bambusicola loculata</i>	MFLUCC 13-0856 ^T	KP761735	KP761729	KP761732	KP761724	KP761715
<i>Bambusicola massarinia</i>	MFLUCC 11-0135 ^T	KU872115	KU863111	KU940122	KU940192	KU940169
<i>Bambusicola pustulata</i>	MFLUCC 15-0190 ^T	KU872112	KU863107	KU940118	KU940190	KU940165
<i>Bambusicola sichuanensis</i>	SICAUCC 16-0002 ^T	MK253528	MK253532	MK253473	MK262828	MK262830
<i>Bambusicola splendida</i>	MFLUCC 11-0439 ^T	JX442042	JX442038	JX442034	KP761726	KP761717
<i>Bambusicola subthailandica</i>	SICAU 16-0005 ^T	MK253529	MK253533	MK253474	MK262829	MK262831
<i>Bambusicola thailandica</i>	MFLUCC 11-0147 ^T	KU872113	KU863108	KU940119	KU940191	KU940166
<i>Crassiclypeus aquaticus</i>	CBS 143643 ^T	LC312472	LC312530	LC312501	LC312559	LC312588
<i>Decaisnella formosa</i>	BCC 25616	GQ925833	GQ925846	-	GU479851	-
<i>Decaisnella formosa</i>	BCC 25617	GQ925834	GQ925847	-	GU479850	-
<i>Flabellascoma cycadicola</i>	CBS 143644 ^T	LC312473	LC312531	LC312502	LC312560	LC312589
<i>Forliomyces uniseptata</i>	MFLUCC 15-0765 ^T	NG_061234	NG_059659	NR_154006	KU727897	-
<i>Glontiopsis praelonga</i>	CBS 112415	FJ161134	FJ161173	-	FJ161090	FJ161113
<i>Guttulispora crataegi</i>	MFLUCC 13-0442 ^T	KP899125	KP888639	KP899134	KR075161	-
<i>Halothia posidoniae</i>	BBH 22481	GU479752	GU479786	-	-	-
<i>Hysterium angustatum</i>	MFLUCC 16-0623	GU397359	FJ161180	-	FJ161096	MH535875

Table 1. Cont.

Species	Strain ¹	GenBank Accession Numbers				
		SSU	LSU	ITS	<i>tef1</i>	<i>rpb2</i>
<i>Kazuakitanaka lancangensis</i>	HKAS122922 ^T	ON009088	ON009104	ON009120	ON009263	-
<i>Kazuakitanaka lancangensis</i>	HKAS122923	ON009089	ON009105	ON009121	ON009264	-
<i>Kazuakitanaka yuxiensis</i>	HKAS122924	ON009092	ON009108	ON009124	ON009267	ON009290
<i>Kazuakitanaka yuxiensis</i>	HKAS122925 ^T	ON009093	ON009109	ON009125	ON009268	ON009291
<i>Leptoparies palmarum</i>	CBS 143653 ^T	LC312485	LC312543	LC312514	LC312572	LC312601
<i>Leptosphaeria doliolum</i>	CBS 505.75 ^T	GU296159	GU301827	NR_155309	GU349069	KY064035
<i>Leucaenicola aseptata</i>	MFLUCC 17-2423 ^T	NG_065776	MK347963	NR_163332	MK360059	MK434891
<i>Leucaenicola phraeana</i>	MFLUCC 18-0472 ^T	NG_065784	MK348003	MK347785	MK360060	MK434867
<i>Lignosphaeria fusispora</i>	MFLUCC 11-0377 ^T	-	KP888646	NR_164233	-	-
<i>Loculosulcatispora thailandica</i>	MFLUCC 20-0108 ^T	MT383968	MT383964	MT376742	MT380476	MT380475
<i>Loculosulcatispora hongheensis</i>	HKAS122920	ON009090	ON009106	ON009122	ON009265	ON009288
<i>Loculosulcatispora hongheensis</i>	HKAS122921 ^T	ON009091	ON009107	ON009123	ON009266	ON009289
<i>Magnicamarosporium diospyricola</i>	MFLUCC 16-0419 ^T	NG_065102	KY554212	NR_153486	KY554209	KY554208
<i>Magnicamarosporium iriomotense</i>	KT 2822 ^T	AB797219	AB807509	AB809640	AB808485	-
<i>Mauritiana rhizophorae</i>	BCC 28866	GU371832	GU371824	-	GU371817	-
<i>Misturatosphaeria aurantiacinotata</i>	GKM 1238 ^T	-	NG_059927	-	GU327761	-
<i>Neobambusicola strelitzia</i>	CBS 138869 ^T	-	KP004495	NR_137945	MG976037	-
<i>Parasulcatispora clematidis</i>	MFLUCC 17-2082 ^T	-	MT214593	MT310637	MT394650	-
<i>Phaeoseptum aquaticum</i>	CBS 123113 ^T	-	JN644072	KY940803	-	-
<i>Phaeoseptum carolshearerianum</i>	NFCCI-4221 ^T	MK307816	MK307813	MK307810	MK309874	MK309877
<i>Phaeoseptum carolshearerianum</i>	NFCCI-4384	MK307818	MK307815	MK307812	MK309876	MK309879
<i>Phaeoseptum hydei</i>	MFLUCC 17-0801 ^T	MT240624	MT240623	MT240622	MT241506	-
<i>Phaeoseptum mali</i>	HKAS122916	ON009082	ON009098	ON009114	ON009257	ON009282
<i>Phaeoseptum mali</i>	HKAS122917	ON009083	ON009099	ON009115	ON009258	ON009283
<i>Phaeoseptum mali</i>	MFLUCC 17-2108 ^T	-	MK625197	MK659580	MK647990	-
<i>Phaeoseptum manglicola</i>	NFCCI-4666 ^T	MK307817	MK307814	MK307811	MK309875	MK309878
<i>Phaeoseptum terricola</i>	MFLUCC 10-0102 ^T	MH105780	MH105779	MH105778	MH105781	-
<i>Pleopunctum ellipsoideum</i>	MFLUCC 19-0390 ^T	MK804514	MK804517	MK804512	MK828510	-
<i>Pleopunctum pseudoellipsoideum</i>	KUMCC 21-0820	ON009084	ON009100	ON009116	ON009259	ON009284
<i>Pleopunctum pseudoellipsoideum</i>	HKAS122915	ON009085	ON009101	ON009117	ON009260	ON009285
<i>Pleopunctum pseudoellipsoideum</i>	MFLUCC 19-0391 ^T	-	MK804518	MK804513	MK828511	-
<i>Pleopunctum thailandicum</i>	MFLUCC 21-0039 ^T	-	MZ198896	MZ198894	MZ172461	-
<i>Pleopunctum menglaense</i>	KUMCC 21-0025	ON009086	ON009102	ON009118	ON009261	ON009286
<i>Pleopunctum menglaense</i>	KUMCC 21-0026 ^T	ON009087	ON009103	ON009119	ON009262	ON009287
<i>Polyschema congolensis</i>	CBS 542.73 ^T	-	EF204502	MH860770	-	EF204486
<i>Polyschema larviformis</i>	CBS 463.88 ^T	-	EF204503	-	-	-
<i>Polyschema terricola</i>	CBS 301.65 ^T	EF204519	EF204504	NR_160100	-	EF204487
<i>Pseudoaurantiascoma kenyense</i>	GKM 1195 ^T	-	NG_059928	-	GU327767	-
<i>Pseudobambusicola thailandica</i>	BCC 79462	-	MG926560	MG926559	MG926562	MG926561
<i>Pseudolophiostoma cornisporum</i>	CBS 143654 ^T	LC312486	LC312544	LC312515	LC312573	LC312602
<i>Ramusculicola thailandica</i>	MFLUCC 13-0284 ^T	KP899131	KP888647	KP899141	KR075167	-
<i>Sporormurispora atraphaxidis</i>	MFLUCC 17-0742 ^T	NG_061296	NG_059880	NR_157546	-	-
<i>Stemphylium vesicarium</i>	CBS 191.86 ^T	DQ247812	DQ247804	MH861935	DQ471090	KC584471
<i>Sulcatispora acerina</i>	KUMCC 21-0821	ON009096	ON009112	ON009128	ON009271	ON009294
<i>Sulcatispora acerina</i>	KUMCC 21-0822	ON009097	ON009113	ON009129	ON009272	ON009295
<i>Sulcatispora acerina</i>	KT 2982 ^T	LC014605	LC014610	LC014597	LC014615	-
<i>Sulcatispora berchemiae</i>	KT 1607 ^T	AB797244	AB807534	AB809635	AB808509	-
<i>Sulcatispora berchemiae</i>	KUMCC 21-0823	ON009094	ON009110	ON009126	ON009269	ON009292
<i>Sulcatispora berchemiae</i>	KUMCC 21-0824	ON009095	ON009111	ON009127	ON009270	ON009293
<i>Sulcosporium thailandicum</i>	MFLUCC 12-0004 ^T	KT426564	KT426563	MG520958	-	-
<i>Teichospora melanommoides</i>	CBS 140733 ^T	-	KU601585	NR_154632	KU601610	-
<i>Teichospora pusilla</i>	CBS 140731 ^T	-	KU601586	NR_154633	KU601605	-
<i>Teichospora rubriostiolata</i>	CBS 140734 ^T	-	KU601590	NR_154634	KU601609	KU601599
<i>Thyridaria macrostomoides</i>	GKM 1033	-	GU385190	-	GU327776	-
<i>Thyridaria macrostomoides</i>	GKM 1159	-	GU385185	-	GU327778	-
<i>Thyridaria macrostomoides</i>	GKM 224N	-	GU385191	-	GU327777	-
<i>Vaginatispora appendiculata</i>	MFLUCC 16-0314 ^T	KU743219	KU743218	KU743217	KU743220	-
<i>Westerdykella ornata</i>	CBS 379.55	GU296208	GU301880	AY943045	GU349021	-

¹ Ex-type strains are mentioned with superscripted "T".

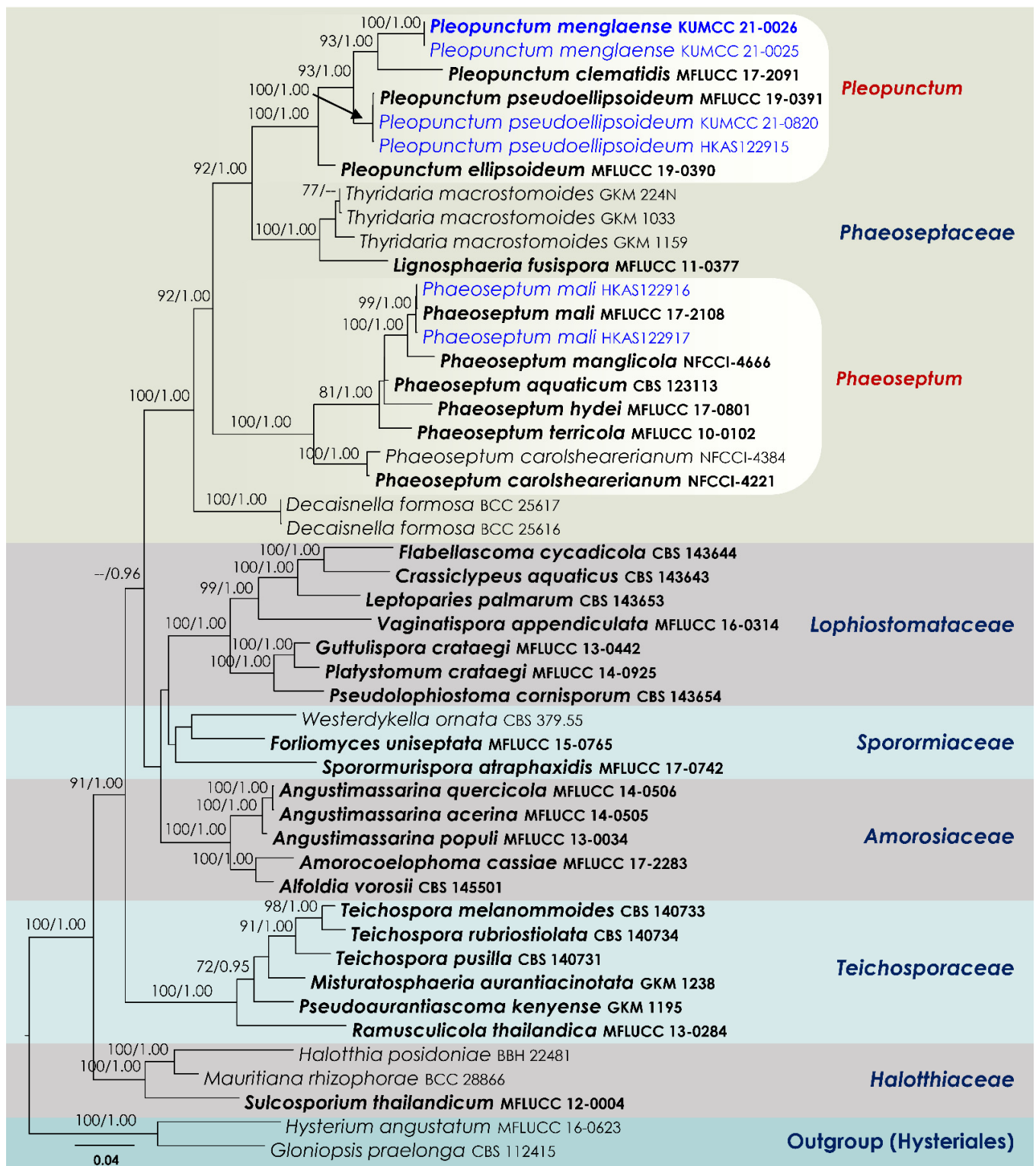


Figure 1. RAxML tree based on a combined dataset of a partial SSU, LSU, ITS, *tef1* and *rpb2* DNA sequence analysis in *Phaeoseptaceae*. Bootstrap support values for ML (MLB) equal to or greater than 70% and Bayesian posterior probabilities (BYPP) equal to or greater than 0.95 are shown as MLB/BYPP above the nodes. The new isolates are in blue. Species names given in bold indicate ex-type and ex-paratype strains. The scale bar represents the expected number of nucleotide substitutions per site.

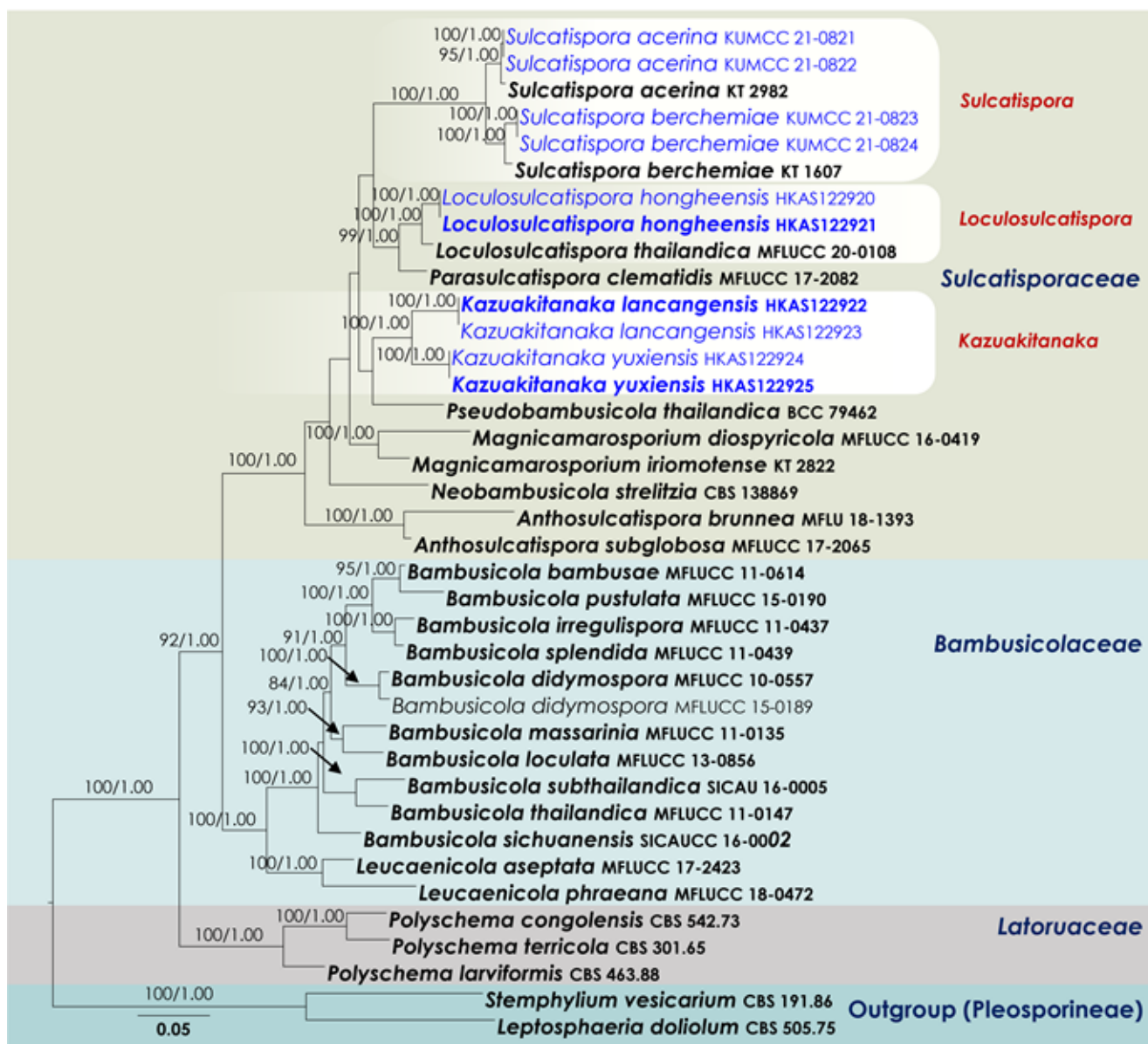


Figure 2. RAxML tree based on a combined dataset of a partial SSU, LSU, ITS, *tef1* and *rpb2* DNA sequence analysis in *Sulcatissporaceae*. Bootstrap support values for ML (MLB) equal to or greater than 70% and Bayesian posterior probabilities (BYPP) equal to or greater than 0.95 are shown as MLB/BYPP above the nodes. The new isolates are in blue. Species names given in bold indicate ex-type and ex-paratype strains. The scale bar represents the expected number of nucleotide substitutions per site.

2.3.2. Phylogenetic Analyses

The single-gene datasets were examined for topological incongruence among loci and the conflict-free alignments were concatenated into a multi-locus alignment that was subjected to maximum-likelihood (ML) and Bayesian (BI) phylogenetic analyses. The evolutionary models for Bayesian analysis and maximum-likelihood were selected independently for each locus using MrModeltest v.2.3 [40] under the Akaike Information Criterion (AIC) implemented in PAUP v.4.0b10.

The CIPRES Science Gateway platform [41] was used to perform RAxML and Bayesian analyses. ML analyses were made with RAxML-HPC2 on XSEDE v.8.2.10 [42] using the GTR+GAMMA swap model with 1000 bootstrap repetitions.

MrBayes analyses were performed setting GTR+I+G for two million generations, sampling every 1000 generations, ending the run automatically when the standard deviation of split frequencies dropped below 0.01, with a burnin fraction of 0.25. ML bootstrap values (MLB) equal to or greater than 70% and posterior probability in Bayesian statistics (BYPP) greater than 0.95 are given above each node of every tree.

Phylograms were visualized with the FigTree v1.4.0 program [43] and reorganized in Microsoft PowerPoint (2007) and Adobe Illustrator® CS5 (Version 15.0.0, Adobe®, San Jose, CA, USA). The finalized alignments and trees were deposited in TreeBASE, submission ID:29570 (<http://purl.org/phylo/treebase/phyloids/study/TB2:S29570>, accessed on 19 March 2022).

2.4. Morphological Observations

Ascomata, conidiophores and conidia from the natural substrates were rehydrated with tap water and examined with a Motic SMZ 168 series stereo-microscope (Motic Asia, Kowloon, Hong Kong). Morphological characteristics were examined via hand sectioning of sporocarps placed on water-mounted glass slides. The following characteristics were evaluated: ascomata/conidiomata diameter, height, color and shape; width of peridium; and height and diameter of ostioles. Microscopic photography was conducted using a Nikon ECLIPSE Ni (Nikon Instruments Inc., Melville, NY, USA) compound microscope with differential interference contrast (DIC) and phase contrast (PC) illumination. Images of microscopic structures were captured with a Canon EOS 600D (Canon Inc., Ōta, Tokyo, Japan) camera. Macroscopic images of colonies were documented using an iPhone XS Max (Apple Inc., Cupertino, CA, USA) with daylight. Measurements were made with the Tarosoft (R) Image Frame Work program and images used for figures were processed with Adobe Photoshop CS6 (Adobe Systems, San Jose, CA, USA).

3. Results

3.1. Phylogenetic Analyses

The final concatenated SSU–LSU–ITS–*tef1*–*rpb2* alignment (Figure 1) of *Phaeoseptaceae* comprised 48 strains, including the outgroup taxa *Gloniopsis praelonga* (CBS 112415) and *Hysterium angustatum* (MFLUCC 16-0623). The final alignment contained a total of 4478 characters used for the phylogenetic analyses, including alignment gaps (available in TreeBASE). The RAxML analysis of the combined dataset yielded a best scoring tree with a final ML optimization likelihood value of -30565.564394 . The matrix had 1930 distinct alignment patterns, with 31.15% undetermined characters or gaps. Parameters for the GTR + I + G model of the combined amplicons were as follows: estimated base frequencies, A = 0.242181, C = 0.257106, G = 0.272525, T = 0.228188; substitution rates, AC = 1.247807, AG = 3.156565, AT = 1.478256, CG = 1.137163, CT = 7.093906, GT = 1.000; proportion of invariable sites, I = 0.397831; gamma distribution shape parameter, α = 0.56366. Bayesian analyses generated 3201 trees (average standard deviation of split frequencies: 0.009446) from which 2401 were sampled after 25% of the trees were discarded as burn-in. The alignment contained a total of 1932 unique site patterns.

The family *Phaeoseptaceae* (SSU, LSU, ITS, *tef1* and *rpb2* phylogeny) was resolved into four distinct clades with 100 MLB and 1.00 BYPP support values. Four strains of *Pleopunctum* (KUMCC 21-0820, KUMCC 21-0025, KUMCC 21-0026 and HKAS122915) isolated in the study formed a well-supported clade with *P. clematidis* (MFLUCC 17-2091), *P. ellipsoideum* (MFLUCC 19-0390) and *P. pseudoellipsoideum* (MFLUCC 19-0391). Three strains of *Thyridaria macrostomoides* (GKM 224N, GKM 1033, GKM 1159) constituted a strongly supported monophyly with the ex-type strain of *Lignosphaeria fusispora* (MFLUCC 11-0377). Two of our new strains, HKAS122916 and HKAS122917, nested with the ex-type strain of *Phaeoseptum mali* MFLUCC 17-2108 with 100 MLB and 1.00 BYPP statistical support. Strains of *Phaeoseptum aquaticum*, *P. carolshearerianum*, *P. hydei*, *P. mali*, *P. manglicola* and *P. terricola* grouped as a monophyletic clade with strong support values in both maximum likelihood and Bayesian analyses. Finally, the two strains of *Decaisnella formosa* (BCC 25616,

BCC 25617) formed a basal terminal clade in *Phaeoseptaceae* with 100 MLB and 1.00 BYPP support values.

The *Sulcatisporaceae* (SSU, LSU, ITS, *tef1* and *rpb2* phylogeny) alignment contained 38 isolates (Figure 2), and the tree was rooted to *Leptosphaeria doliolum* (CBS 505.75) and *Stemphylium vesicarium* (CBS 191.86). The final alignment contained a total of 4931 characters used for the phylogenetic analyses, including alignment gaps (available in TreeBASE). The RAxML analysis of the combined dataset yielded a best scoring tree with a final ML optimization likelihood value of -27594.247903 . The matrix had 1770 distinct alignment patterns, with 23.64% undetermined characters or gaps. Parameters for the GTR + I + G model of the combined amplicons were as follows: estimated base frequencies, A = 0.241395, C = 0.257298, G = 0.268694, T = 0.232613; substitution rates, AC = 1.256046, AG = 2.926191, AT = 1.154561, CG = 0.858049, CT = 6.686514, GT = 1.000; proportion of invariable sites, I = 0.499455; gamma distribution shape parameter, $\alpha = 0.532594$. The Bayesian analyses generated 401 trees (average standard deviation of split frequencies: 0.009088) from which 301 were sampled after 25% of the trees were discarded as burn-in. The alignment contained a total of 1771 unique site patterns.

The family Sulcatisporaceae was composed of distinct lineages that correspond to the genera Anthosulcatispora, Loculosulcatispora, Magnicamarosporium, Neobambusicola, Parasulcatispora, Pseudobambusicola and Sulcatispora. Four strains (HKAS122922, HKAS122923, HKAS122924, HKAS122925) obtained in the present study formed a well-separated clade in Sulcatisporaceae, featuring both a single locus and concatenated datasets. Thus, this new lineage is presented here as the new genus *Kazuakitanaka* gen. nov. Two of our new isolates, KUMCC 21-0821 and KUMCC 21-0822, were grouped with the ex-type strain of *Sulcatispora acerina* (KT 2982) with 95 MLB and 1.00 BYPP values. Another two new strains (KUMCC 21-0823 and KUMCC 21-0824) grouped with the ex-type strain of *Sulcatispora berchemiae* (KT 1607), with 100 MLB and 1.00 BYPP support values. *Loculosulcatispora thailandica* (MFLUCC 20-0108), a type of *Loculosulcatispora*, constituted a monophyletic clade with two of our new isolates (HKAS122920 and HKAS122921). *Neobambusicola strelitzia*, *Parasulcatispora clematidis* and *Pseudobambusicola thailandica* were affiliated as monotypic genera. Two species of *Magnicamarosporium*, *M. diospyricola* (MFLUCC 16-0419) and *M. iriomotense* (KT 2822), grouped with 100 MLB and 1.00 BYPP support values. *Anthosulcatispora brunnea* (MFLU 18-1393) and *A. subglobosa* (MFLUCC 17-2065) formed a basal terminal clade in Sulcatisporaceae, with 100 MLB and 1.00 BYPP statistical support.

3.2. Taxonomy

Pleosporales Luttr. ex M.E. Barr, Prodr. Mycol. 67 (1987).

Phaeoseptaceae Boonmee, Thambug, and K.D. Hyde, Mycosphere 9 (2): 323 (2018).

Phaeoseptum Y. Zhang, J. Fourn. and K.D. Hyde, Mycologia 105 (3): 606 (2013).

Phaeoseptum mali Phukhams. and K.D. Hyde, Asian Journal of Mycology 2 (1): 120 (2019).

Material examined: China, Yunnan, Honghe Hani and Yi Autonomous Prefecture, Kaiyuan, 23.863601° N, 103.407975° E, 1053 m, on dead woody litter, 17 March 2019, D.N. Wanasinghe (HKAS122916), *ibid.* Chuxiong Yi Autonomous Prefecture, Shuangbai County, 24.80527° N, 101.933887° E, 1736 m, on dead woody litter under the *Fagaceae* species, 25 June 2019 (HKAS122917).

Notes: Phukhamsakda et al. [44] introduced *Phaeoseptum mali* from the bark of fallen twigs of *Malus halliana* from the botanical garden of the Kunming Institute of Botany, Yunnan, China. *Phaeoseptum mali* fits well within the generic descriptions of *Phaeoseptum* based on its globose and immersed ascomata, cellular pseudoparaphyses, cylindrical-clavate, pedicellate asci, allantoid and brown muriform ascospores. In this study, we collected another two specimens of *Phaeoseptum mali* from Chuxiong and Honghe Prefectures in Yunnan, China on dead woody litter. Multi-gene phylogenetic analyses of combined SSU, LSU, ITS, *tef1* and *rpb2* DNA sequence showed that the ex-type strain of *Phaeoseptum mali* (MFLUCC 17-2108) and our two strains (HKAS122916 and HKAS122917) are monophyletic

with 100% MLB and 1.00 BYPP support values. Morphologically, these new collections are not different from the holotype of *Phaeoseptum mali*.

Pleopunctum N.G. Liu, K.D. Hyde and J.K. Liu, Mycosphere 10 (1): 767 (2019).

Pleopunctum menglaense Wanas. sp. nov. (Figure 3).

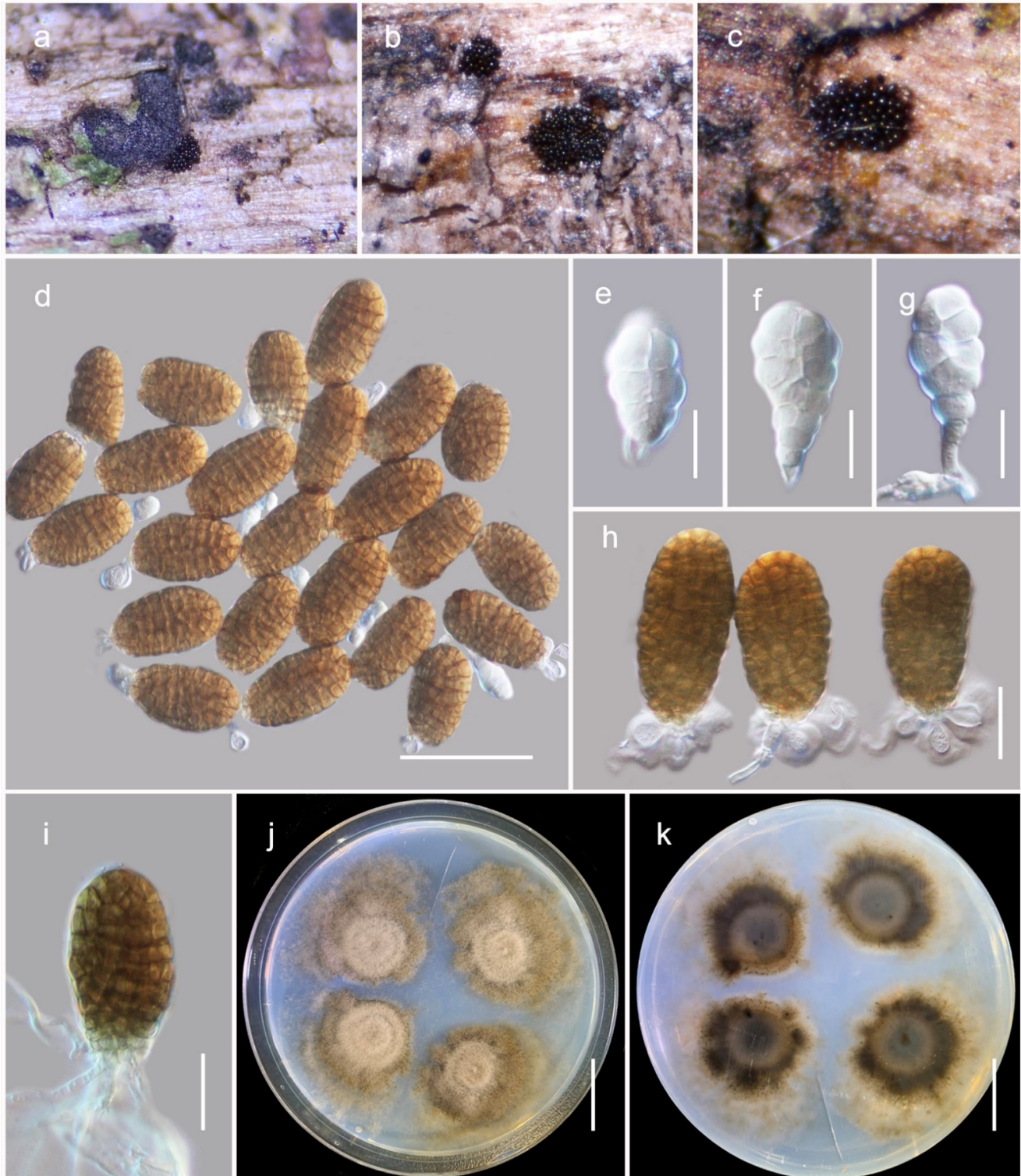


Figure 3. The anamorph of *Pleopunctum menglaense* (HKAS122683, holotype). (a–c) Colonies on host surface; (d) β conidia with basal cells; (e,f) α conidia showing remnant of conidiogenous cells at base; (g) α conidia with the conidiophore;(h) β conidia with distinct basal cells; (i) germinating conidium; (j,k) colonies on PDA after 21 days. Scale bars: (d) 50 μ m; (e–g) 10 μ m; (h,i) 20 μ m; (j,k) 2 cm.

Mycobank: MB843430.

Etymology: The specific epithet is derived from Mengla County, Yunnan Province, China.

Holotype: HKAS122683.

The species is a saprobe on dead twigs of forest litter in terrestrial habitats. Teleomorph: undetermined. Anamorph: hyphomycetous. Colonies on host, sporodochial, superficial, black, scattered and punctiform. The conidiophores are arising from hyaline unbranched hyphae. They are $10\text{--}20 \times 2.5\text{--}3.5 \mu\text{m}$ ($M = 16.7 \times 3.1$, $n = 15$), macronematous, mononematous, cylindrical, unbranched, aseptate, hyaline and smooth-walled. The conidiogenous cells $5\text{--}8 \times 2.8\text{--}3.5 \mu\text{m}$ ($M = 6.9 \times 3.2 \mu\text{m}$, $n = 30$) are monoblastic, terminal, hyaline. The conidia are dimorphic, acrogenous and solitary. The α conidia are $18\text{--}25 \times 10\text{--}14 \mu\text{m}$ ($M = 23.1 \times 11.7 \mu\text{m}$, $n = 30$), hyaline, multi-septate, muriform, spatulate to obovate, notably constricted at septa, slightly obtuse to rounded at apex, notably narrow at base, often carrying remnants of conidiogenous cells at base. The β conidia are $38\text{--}55 \times 20\text{--}26 \mu\text{m}$ ($M = 45.6 \times 24.4 \mu\text{m}$, $n = 30$), brown, muriform, ellipsoidal to oblong shaped, moderately rough-walled, and slightly constricted at septa often with a hyaline, elliptical to globose, 1-3 basal cells, $7.5\text{--}12 \times 4\text{--}6 \mu\text{m}$ ($M = 9 \times 5.1 \mu\text{m}$, $n = 25$) with a tri-lobed wing-like appearance.

Culture characteristics: the conidia germinated on PDA within 12 h and germ tubes were produced from the basal cells of conidia. The colonies on PDA reached a 5 cm diameter after 2 weeks at 25 °C. They were effused, circular to lobed, with incised margin, flat or slightly hairy, pinkish brown in the center with concentric rings and pale brown at the periphery and reverse; three distinct color zones were present, namely greyish brown at the center, pinkish brown at the middle, golden brown at the periphery.

Material examined: China, Yunnan, Xishuangbanna Dai Autonomous Prefecture, Mengla County, Wangtianshu, 21.61852° N, 101.58171° E, 747 m, on dead woody litter, 12 January 2021, D.N. Wanasinghe (HKAS122683, holotype), ex-type living culture, KUMCC 21-0826. *ibid.* 21.618839° N, 101.581098° E, 774 m (HKAS122684), living culture, KUMCC 21-0827.

Pleopunctum pseudoellipsoideum N.G. Liu, K.D. Hyde and J.K. Liu, Mycosphere 10 (1): 768 (2019)

Material examined: China, Yunnan, Diqing Tibetan Autonomous Prefecture, Weixi Lisu Autonomous County, Zili Village, 27.803791° N, 99.059136° E, 2250 m, on dead woody litter, 25 June 2019, L.Q. Xian (HKAS122914), living culture, KUMCC 21-0820, *ibid.* Kunming, Luquan Yi and Miao Autonomous County, 26.052826° N, 102.670548° E, 2629 m, on dead woody litter under the *Fagaceae* species, 2 July 2019, D.N. Wanasinghe (HKAS122915).

Notes: Liu et al. [45] introduced *Pleopunctum pseudoellipticum* from decaying wood in Guizhou Province, China as a hyphomycetous muriform spored anamorph taxon in *Phaeoseptaceae*. During our investigation on the diversity of woody litter microfungi in Yunnan, China, two collections were made from the Diqing and Kunming areas. Morphological characteristics of our new collections, such as conidiophores and conidia, fit well within the anamorph of *Pleopunctum pseudoellipticum*. In our phylogenetic study (Figure 1), the new strains (KUMCC 21-0820, HKAS122915) cluster with *Pleopunctum pseudoellipticum* (MFLUCC 19-0391, ex-type strain) as a monophyletic clade with 100% MLB and 1.00 BYPP support values. Comparison of ITS, LSU and *tef1* sequence data reveals there is no significant difference between our new isolates and *Pleopunctum pseudoellipticum*. Therefore, we recognize our new isolates as additional collections of *Pleopunctum pseudoellipticum*, and we provide SSU and *rpb2* sequence data that were not provided by Liu et al. [45].

Sulcatisporaceae Kaz. Tanaka and K. Hiray., Studies in Mycology 82: 119 (2015).

Kazuakitanaka Wanas. gen. nov.

Mycobank: MB843431.

Etymology: The generic epithet stems from the combined two words “kazuaki” and “tanaka”, referring to Kazuaki Tanaka, who introduced the family Sulcatisporaceae.

The genus comprises saprobic fungi on woody substrates in terrestrial habitats. Teleomorph: the ascomata is a solitary or gregarious, semi-immersed, erumpent through the host

surface, coriaceous, dark brown to black, globose to subglobose, ostiolate. The ostiole is central, with hyaline to pale-brown pseudoparenchymatous cells. The peridium is broader at the apex and thinner at the base, comprising two strata with several layers of brown or lightly pigmented to hyaline cells building *textura angularis* to *textura prismatica*, indistinguishable from the host tissues. The hamathecium comprises many branched, septate, cellular pseudoparaphyses, located between and above the asci, embedded in a gelatinous matrix. The asci are eight-spored, bitunicate, fissitunicate, cylindric-clavate, pedicellate, and apically rounded, with an ocular chamber. The ascospores are uni- to bi-seriate, partially overlapping in lateral view, and are hyaline. They are fusiform, 1–2-septate, slightly curved, deeply constricted at the central septum, conically rounded at the ends, and smooth-walled, with a distinct mucilaginous sheath, comprising a large guttule in each cell. Anamorph: Coelomycetous. The conidiomata are pycnidial, solitary, gregarious, dark brown to black, immersed or slightly erumpent, coriaceous to carbonaceous, papillate or apapillate. The conidiomatal wall is multi-layered; the outer layers are pseudoparenchymatous, with brown-walled cells, with the innermost layer thin and hyaline. The conidiophores are reduced to conidiogenous cells. The conidiogenous cells are phialidic, ampulliform to cylindrical, determinate, hyaline, smooth-walled and formed from the inner layer of the pycnidium wall. The conidia are hyaline, one-to-three-septate, straight to curved, fusiform, with conical ends, thick-walled, smooth, with large guttules in each cell.

Type species: *Kazuakitanaka yuxiensis*

Kazuakitanaka yuxiensis Wanas. sp. nov. (Figure 4).

Mycobank: MB843432.

Etymology: The specific epithet is derived from Yuxi, Yunnan Province, China.

Holotype: HKAS122924.

The species is a saprobe on dead twigs of forest litter in terrestrial habitats. Teleomorph: the ascomata is 350–400 μm high, 220–260 μm diam. ($M = 386.6 \times 249.9 \mu\text{m}$, $n = 5$), scattered to gregarious, semi-immersed to erumpent, coriaceous, dark brown to black, globose to subglobose ostiolate. The ostiole is 110–140 μm long, 90–115 μm diam. ($M = 125.6 \times 98.7 \mu\text{m}$, $n = 5$), centrally papillate, comprising hyaline cells. The peridium is 10–15 μm wide at the base, 12–20 μm wide at the sides, broad at the apex (30–40 μm), comprising two strata, with the outer stratum composed of small, pale brown to brown, slightly flattened, thick-walled cells of *textura angularis*, fusing and indistinguishable from the host tissues. The inner stratum is composed of several layers with lightly pigmented to hyaline cells centrally *textura angularis* to *textura prismatica*. The hamathecium is 1–2 μm wide, branched, septate, cellular pseudoparaphyses, situated between and above the asci, embedded in a gelatinous matrix. The asci are 80–120 \times 17–21 μm ($M = 102.2 \times 18.5 \mu\text{m}$, $n = 20$), eight-spored, bitunicate, fissitunicate, cylindric-clavate, with a pedicel, and is rounded at the apex, with an ocular chamber. The ascospores are 25–32 \times 7–9 μm ($M = 29.1 \times 7.9 \mu\text{m}$, $n = 30$), uni- to bi-seriate, overlapping, and hyaline. They are fusiform, 1–2-septate. They are slightly curved, deeply constricted at the central septum, conically rounded at the ends, and smooth-walled, surrounded by a distinct mucilaginous sheath, each cell with a large guttule. Anamorph: undetermined.

Material examined: China, Yunnan, Yuxi, Xinping Yi and Dai Autonomous County, 24.09083° N, 101.935124° E, 2091 m, on dead woody litter, 25 May 2019, D.N. Wanasinghe (HKAS122924, holotype). *ibid.* 24.09083° N, 101.935124° E, 2095 m (HKAS122925).

Kazuakitanaka lancangensis Wanas. sp. nov. (Figure 5).

Mycobank: MB843433.

Etymology: The specific epithet is derived from Lancang, Yunnan Province, China.

Holotype: HKAS122922.

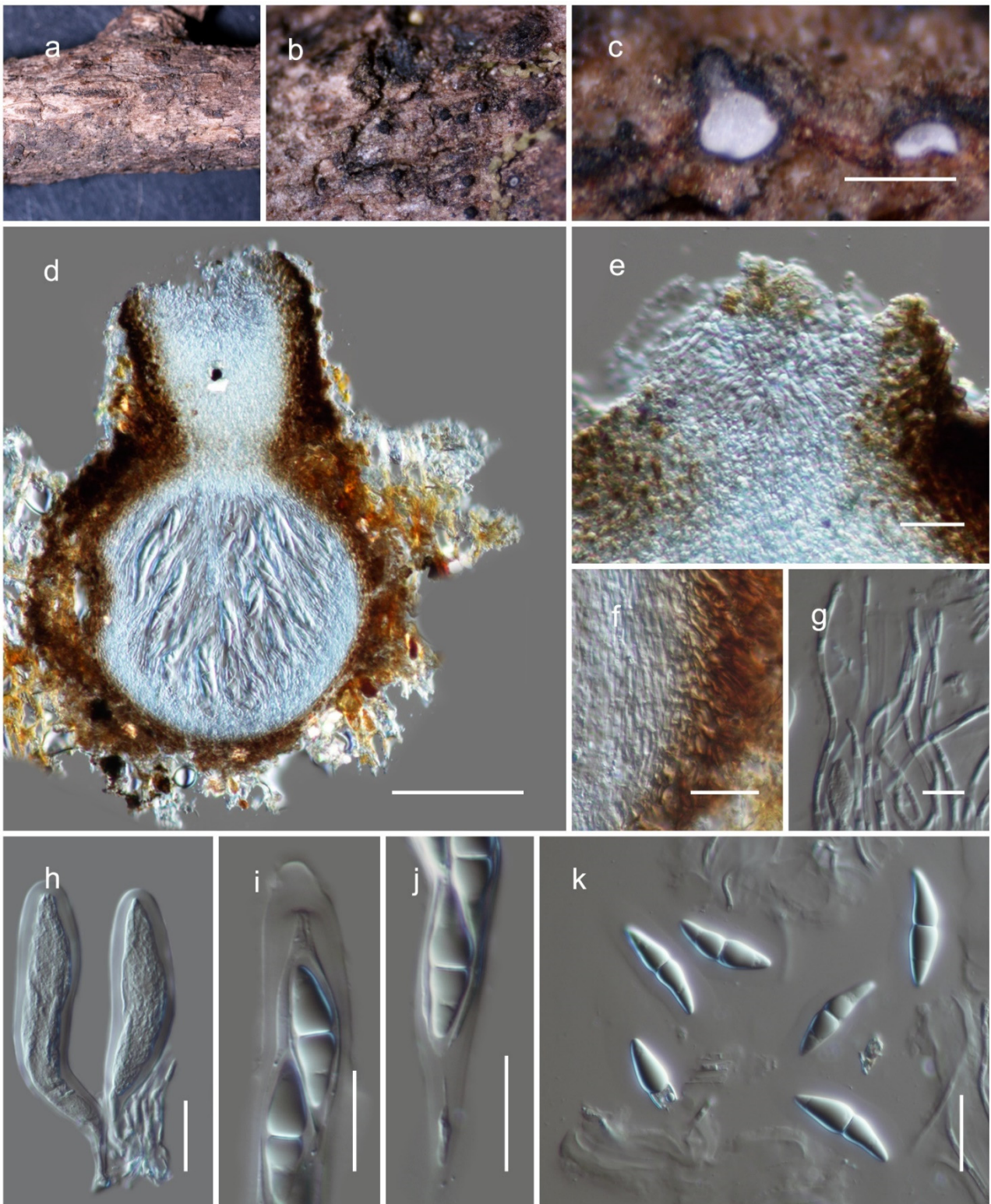


Figure 4. The teleomorph of *Kazuakitataka yuxiensis* (HKAS122924, holotype). (a,b) Ascomata on dead woody twigs; (c,d) vertical section of ascomata; (e) closeup of ostiole; (f) peridium; (g) pseudoparaphyses; (h–j) asci; (k) ascospores. Scale bars: (c) 500 μm ; (d) 100 μm ; (e,f,h–k) 20 μm ; (g) 10 μm .

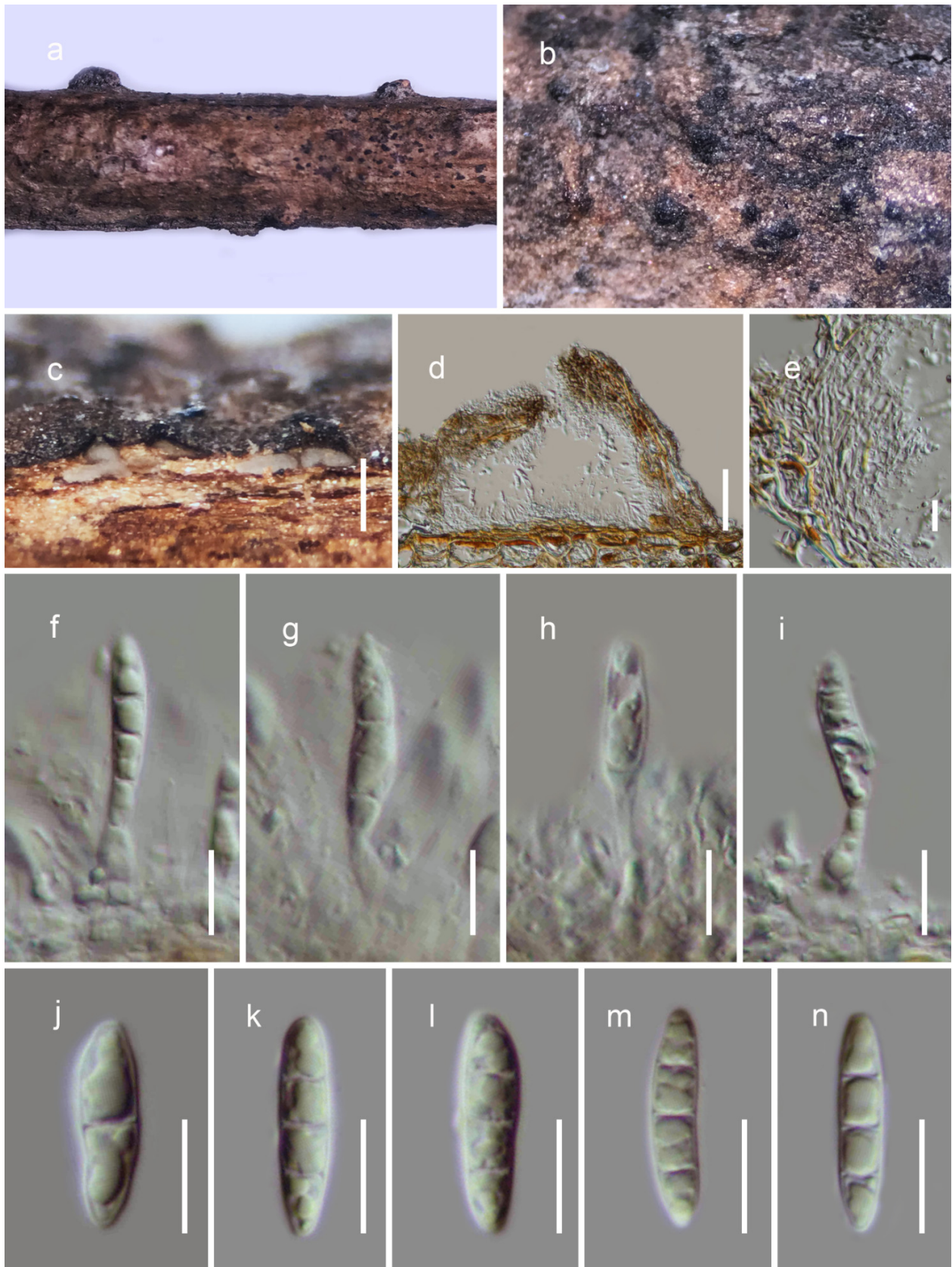


Figure 5. *Kazuakitanaka lancangensis* (HKAS122922, holotype). (a,b) Conidiomata on host surface; (c,d) sections through conidiomata; (e) conidioma wall; (f–i) conidiogenous cells and developing conidia; (j–n) conidia. Scale bars: (c) 500 μm ; (d) 50 μm ; (e) 15 μm ; (f–n) 10 μm .

It is saprobic on dead twigs of forest litter in terrestrial habitats. Teleomorph: undetermined. Anamorph: coelomycetous. The conidiomata is 130–150 µm high, 160–190 µm diam. ($M = 140 \times 170 \mu\text{m}$, $n = 5$), pycnidial, solitary, gregarious, globose to subglobose, coriaceous, uni-loculate, dark brown to black, and immersed, with a central ostiole. The conidiomata wall is 10–20 µm wide, multi-layered, with brown-walled pseudoparenchymatous cells, with a hyaline inner most layer. The conidiophores are reduced to conidiogenous cells. The conidiogenous cells are 7–10 × 2–3.5 µm ($M = 8.3 \times 2.7 \mu\text{m}$, $n = 15$), phialidic, ampulliform to cylindrical, determinate, hyaline, smooth-walled and formed from the inner layer of the pycnidium wall. The conidia are 17–23 × 3.7–4.7 µm ($M = 20 \times 4.2 \mu\text{m}$, $n = 30$), fusiform, straight, hyaline, one-to-three-septate, not constricted at septa, tip and base rounded, thick-walled, and smooth, with numerous large guttules in each cell.

Material examined: China, Yunnan, Pure, Lancang Lahu Autonomous County, 23.164681° N, 99.969248° E, 1990 m, on dead woody litter, 11 April 2019, D.N. Wanasinghe (HKAS122922, holotype). *ibid.* 23.164686° N, 99.969492° E, 1975 m (HKAS122923).

Loculosulcatispora G.C. Ren and K.D. Hyde, *Phytotaxa* 475 (2): 70 (2020) *amended*

MycoBank: MB557580

The genus comprises saprobic species on dead wood or twigs. Teleomorph: the ascomata are scattered, immersed to semi-immersed, globose to subglobose, brown to dark-brown, with a central ostiole. The peridium is composed of small, pale brown to brown, thick-walled cells forming *textura angularis* to *globosa*. The hamathecium comprises branched, septate, cellular pseudoparaphyses. The asci are eight-spored, bitunicate, clavate, straight to curved, with a short pedicel. They are apically rounded, with or without an ocular chamber. The ascospores are overlapping uni- to bi-seriate, hyaline, fusiform with acute ends, 1-septate, smooth-walled, surrounded by a thick irregular mucilaginous sheath. Anamorph: see Ren et al. [36].

Type species: *Loculosulcatispora thailandica* G.C. Ren and K.D. Hyde, *Phytotaxa* 475 (2): 73 (2020).

Loculosulcatispora hongheensis Wanas. sp. nov. (Figure 6).

MycoBank: MB843434.

Etymology: The specific epithet is derived from Honghe, Yunnan Province, China.

Holotype: HKAS122920.

The species is a saprobe on dead twigs of forest litter in terrestrial habitats. Teleomorph: the ascomata is 130–490 × 120–560 ($M = 260 \times 280$, $n = 5$) µm, scattered, immersed, coriaceous, globose to subglobose, brown to dark-brown, with a central ostiole. The peridium is 10–15 µm wide; the outer stratum composed of small, pale brown to brown, thick-walled cells forming *textura angularis* to *globosa* indistinguishable from the host tissues. The inner stratum is composed of hyaline cells forming *textura angularis*. The hamathecium is 1–2.5 µm wide, branched, septate, cellular pseudoparaphyses, situated between and above the asci, embedded in a gelatinous matrix. The asci are 60–100 × 10–18 µm ($M = 84 \times 13.7 \mu\text{m}$; $n = 15$) µm, eight-spored, bitunicate, fission-tunicate, clavate, straight to curved, with a short pedicel. They are apically rounded, with a minute ocular chamber. The ascospores are 20–30 × 4–6.5 µm ($M = 25 \times 5 \mu\text{m}$, $n = 30$), overlapping uni- to bi-seriate, hyaline, fusiform with acute ends, 1-septate, constricted at the septum, slightly longer upper cell than lower cell, slightly curved, smooth-walled, and surrounded by a thick irregular mucilaginous sheath. Anamorph: undetermined.

Material examined: China, Yunnan, Honghe Hani and Yi Autonomous Prefecture, Mengzi, 23.18604° N, 103.413838° E, 1854 m, on dead woody litter, 16 March 2019, D.N. Wanasinghe (HKAS122920, holotype). *ibid.* 23.186552° N, 103.414063° E, 1866 m (HKAS122921).

Sulcatispora Kaz. Tanaka and K. Hiray., *Studies in Mycology* 82: 120 (2015).

Sulcatispora acerina Kaz. Tanaka and K. Hiray., *Studies in Mycology* 82: 120 (2015).

Material examined: China, Yunnan, Pure, Jingdong Yi Autonomous County, 24.549205° N, 101.027521° E, 2560 m, on dead woody litter, 8 June 2021, D.N. Wanasinghe (HKAS122926), living culture, KUMCC 21-0821. *ibid.* 24.548691° N, 101.027802° E, 2546 m (HKAS122927), living culture, KUMCC 21-0822.

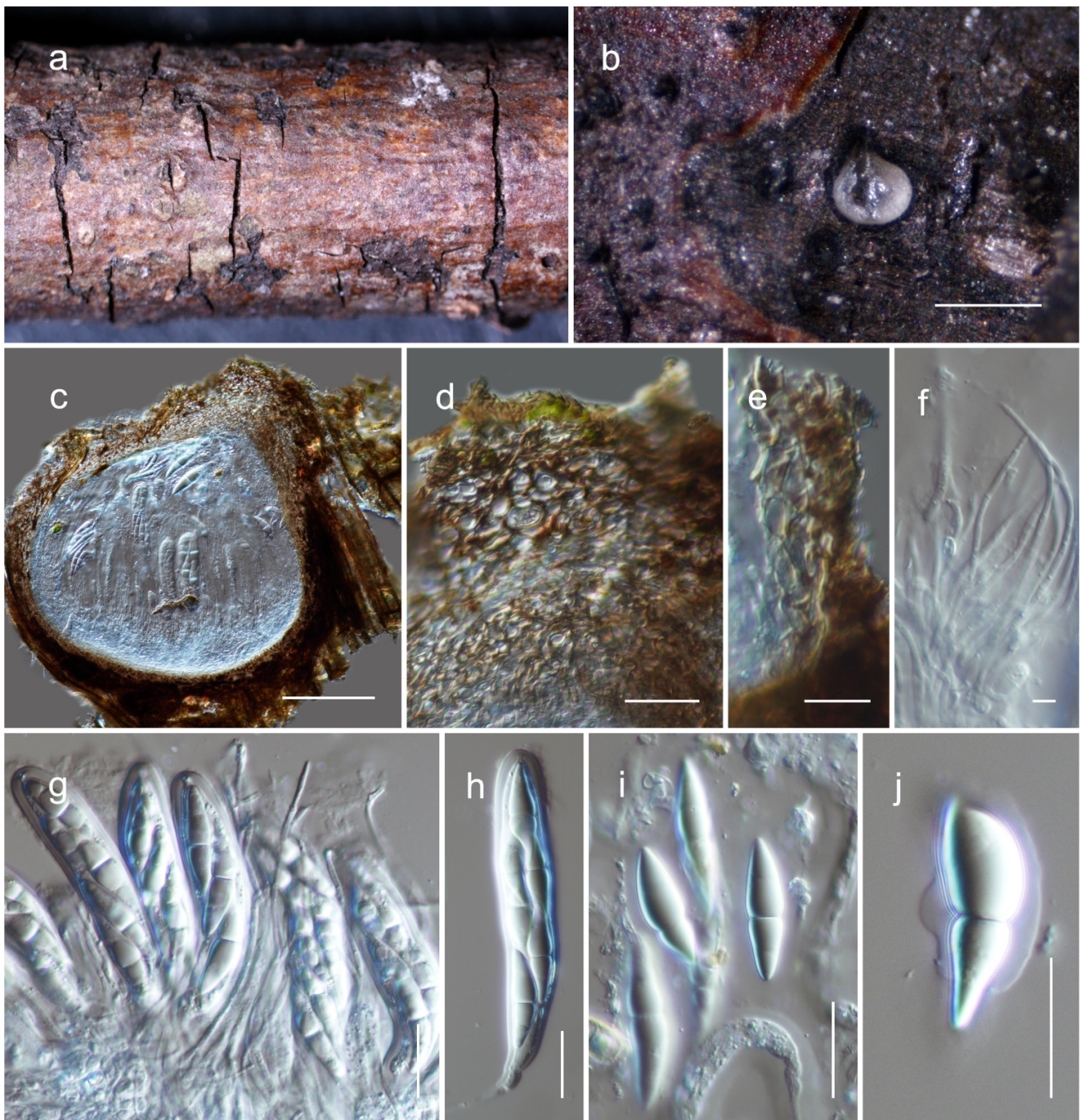


Figure 6. *Loculosulcatispora hongheensis* (HKAS122920, holotype). (a) Dead wood host substrate; (b,c) section of ascoma; (d); close-up of ostiole; (e) peridium; (f) pseudoparaphyses; (g,h) asci; (i,j) ascospores. Scale bars: (b) 500 μ m; (c) 100 μ m; (d,g,h) 20 μ m; (e,i,j) 10 μ m; (f) 5 μ m.

Notes: *Sulcatispora acerina*, introduced by Tanaka et al. [46], was collected from *Acer palmatum* in a terrestrial habitat. Based on phylogenetic analysis of combined SSU, LSU, ITS, *tef1* and *rpb2* sequence data, two of our isolates, KUMCC 21-0821 and KUMCC 21-0822, were clustered with the ex-type strain of *Sulcatispora acerina* (KT 2982) with 95% MLB and 1.00 BYPP support (Figure 2). Our isolate resembles *Sulcatispora acerina* in shape and size of the ascomata, asci and ascospores. Moreover, there are no base pair differences of the SSU and LSU nucleotides and only four and seven nucleotide differences in the ITS and *tef1* regions, respectively. Therefore, we recognize that our new isolates belong to *Sulcatispora*

acerina as new records from China, providing *rpb2* sequence data that were not provided by Tanaka et al. [46].

Sulcatispora berchemiae Kaz. Tanaka and K. Hiray., Studies in Mycology 82: 121 (2015).

Material examined: China, Yunnan, Pure, Jingdong Yi Autonomous County, 24.548237° N, 101.026484° E, 2516 m, on dead woody litter, 8 June 2021, D.N. Wanasinghe (HKAS122918), living culture, KUMCC 21-0823. *ibid.* 24.548142° N, 101.026572° E, 2510 m (HKAS122919), living culture, KUMCC 21-0824.

Notes: In the phylogenetic analysis (Figure 2), two of our new isolates (KUMCC 21-0823, KUMCC 21-0824) clustered with the type strain of *Sulcatispora berchemiae* (KT 1607) with high bootstrap support (100% MLB, 1.00 BYPP). These three strains share similar morphological features in ascomata, asci and ascospores. KT 1607 was collected from Japan on *Berchemia racemosa*, whereas KUMCC 21-0823 and KUMCC 21-0824 were collected from China on dead woody litter. Based on both morphology and molecular data, we consider our new isolates and *Sulcatispora berchemiae* to be conspecific. While extending the biogeography of *Sulcatispora berchemiae*, we also provide the *rpb2* gene region for the species, which was not accounted for earlier.

4. Discussion

Hyde et al. [47] established *Phaeoseptaceae* in *Pleosporales* to accommodate *Phaeoseptum*. However, this family is now recognized as a group of heterogenous taxa that have diversified habitats with different types of anamorphs and diverged morphological characteristics [37]. In the recent outlines of Hongsanan et al. [10] and Wijayawardane et al. [12], only *Phaeoseptum* and *Pleopunctum* are accepted in *Phaeoseptaceae*. In the combined multi-gene phylogenetic analyses, the ex-type strains of *Lignosphaeria diospyrosa*, *L. fuispora* and *L. thailandica* also clustered in *Phaeoseptaceae* [48]. Therefore, *Lignosphaeria* should be an accepted genus within *Phaeoseptaceae*. Even though the putative strains of *Decaisnella formosa* (BCC 25616, BCC 25617) and *Thyridaria macrostomoides* (GKM 1033, GKM 1159, GKM 224N) are phylogenetically affiliated in *Phaeoseptaceae* [37,47], they are not related to any type specimens. Therefore, it is necessary to recollect and epitypify them with DNA sequence data in order to ensure correct generic placement of these two species [49,50].

Phaeoseptum was introduced by Zhang et al. [51] to accommodate *P. aquaticum*, and it was initially described as an accepted species in *Halotthiaceae* based on phylogenetic analysis of LSU sequence data. Currently, six species are accepted in *Phaeoseptum* viz.: *P. aquaticum*, *P. carolshearerianum*, *P. hydei*, *P. mali*, *P. manglicola* and *P. terricola* [10]. *Phaeoseptum aquaticum* was reported from freshwater habitats, and later Hyde et al. [47] added *P. terricola* from a terrestrial environment. In recent studies, Phukhamsakda et al. [44] and Wanasinghe et al. [37] collected *Phaeoseptum mali* and *P. hydei* from terrestrial habitats, respectively. Dayarathne et al. [52] showed that these species can be found in tropical coastlines, while introducing *Phaeoseptum carolshearerianum* and *P. manglicola* from mangroves. In this study, we accounted for another two records of *Phaeoseptum mali* from Chuxiong and Honghe Prefectures in Yunnan (China) from terrestrial habitats.

Pleopunctum, typified by *P. ellipsoideum*, is a phylumycetous genus established in *Phaeoseptaceae* by Liu et al. [45]. Species in the genus are characterized by macronematous, mononematous conidiophores with monoblastic conidiogenous cells and oval to ellipsoidal, muriform conidia mostly with a hyaline, elliptical to globose basal cell [45]. Currently, five *Pleopunctum* species (viz. *P. bauhinae*, *P. clematidis*, *P. ellipsoideum*, *P. pseudoellipsoideum*, *P. thailandicum*) are accepted in Species Fungorum [53]. However, Koukol and Delgado [54] argued that *Pleopunctum clematidis* should be a synonym of *P. bauhinae* (= *Hermatomyces bauhinae*). Members in the genus have appeared to be saprobic on dead twigs of deciduous hosts in terrestrial habitats [45,55–57]. In this study, morphological characteristics and multi-gene phylogenetic analysis of combined SSU, LSU, ITS, *tef1* and *rpb2* DNA sequence data reveals a new species of *Pleopunctum* from the dead woody litter collected in Yunnan, China. In a multi-gene (concatenated LSU, SSU, ITS, *tef1* and *rpb2*) phylogenetic analysis, two of our strains (KUMCC 21-0025, KUMCC 21-0026) clustered with *Pleopunctum*

species as a monophyletic clade with 100 MBP and 1.00 BYPP bootstrap support (Figure 1). Morphological features of KUMCC 21-0025 and KUMCC 21-0026 are similar to extant species of *Pleopunctum* in having sporodochial colonies, holoblastic, monoblastic conidiogenous cells and brown muriform conidia [45]. We introduce these two isolates as a novel species belonging to this genus, *Pleopunctum menglaense*. Within the *Pleopunctum* clade, *P. menglaense* strains constitute a sister lineage to *P. pseudoellipsoideum* (MFLUCC 19-0391, KUMCC 21-0820, HKAS122915). *Pleopunctum menglaense* has contrasting morphological features to *P. pseudoellipsoideum* by its distinct, hyaline, 1–3 basal cells in β conidia, whereas *P. pseudoellipsoideum* has a single basal cell in β conidia [45].

Sulcatissporaceae currently accommodates seven genera: viz. *Anthosulcatisspora*, *Loculosulcatisspora*, *Magnicamarosporium*, *Neobambusicola*, *Parasulcatisspora*, *Pseudobambusicola* and *Sulcatisspora* [12]. The species in this family are mostly restricted to terrestrial habitats excluding *Neobambusicola strelitziae*, which was found from the coastal region (Eastern Cape Province) of South Africa [58]. The conidia of anamorphs of *Sulcatissporaceae* can vary from hyaline, aseptate or septate (*Anthosulcatisspora*, *Loculosulcatisspora*, *Neobambusicola*, *Pseudobambusicola*), to pigmented phragmo-conidia (*Sulcatisspora*) or muriform conidia (*Magnicamarosporium*), with or without striation [36,46,56,58–60]. The anamorph of *Parasulcatisspora* is yet to be discovered. Shared characteristics within the family consist of immersed ascomata, peridium comprising pseudoparenchymatous cells with a thickened apex, cellular or trabeculate pseudoparaphyses and having cylindrical to cylindrical-clavate asci with a long pedicel. However, the ascospore arrangement inside an ascus and the features of ascospores can differ between species belonging to this group. The spore arrangement can be uniseriate (*Anthosulcatisspora*) and 2–3-seriate in others. Spores are hyaline and fusiform with conical ends that are completely surrounded by a sheath excluding *Anthosulcatisspora brunnea*, which is brown to dark brown, oblong to ellipsoidal and lacking a sheath.

Kazuakitanaka is introduced in *Sulcatissporaceae* as a new genus, based on LSU, ITS, *tef1* and *rpb2* sequence data. *Kazuakitanaka* morphologically resembles *Parasulcatisspora* and *Sulcatisspora* with its cylindrical-clavate asci and fusiform, 1-septate hyaline ascospores. However, these genera were revealed as phylogenetically distant in multi-gene phylogenetic analysis (Figure 2). The coelomycetous anamorph of *Kazuakitanaka* is similar to species of *Neobambusicola* and *Pseudobambusicola* by its pycnidial conidiomata, phialidic conidiogenous cells and hyaline, fusiform, multi-septate conidia. The new genus has a close phylogenetic proximity to *Pseudobambusicola* even though this relationship is not statistically significant, and *Neobambusicola* was placed in a distinct branch apart from the *Kazuakitanaka* clade (Figure 2). The teleomorph for these genera is still undetermined and therefore they cannot be morphologically compared with the teleomorph of the new genus.

Ren et al. [36] introduced *Loculosulcatisspora* as a monotypic genus to accommodate *L. thailandica*, and it was known only from its anamorph. *Loculosulcatisspora thailandica* was introduced as a decaying woody-based saprobe from Thailand (Chiang Rai). The genus was characterized by multilocular conidiomata, phialidic, discrete, determinate, doliiform to cylindrical, hyaline conidiogenous cells, unbranched and aseptate paraphyses, and 1-celled, oblong conidia with guttules. During our sample collections, we found a teleomorphic species characterized by globose to subglobose, immersed ascomata with a central ostiole, peridium composed of textura angularis, hamathecium comprises branched, septate, cellular pseudoparaphyses, clavate, short pedicellate asci with a minute ocular chamber, hyaline, fusiform, and 1-septate ascospores with a mucilaginous sheath. In the multi-gene phylogenetic analyses, two of the isolates of this teleomorphic species (HKAS122920, HKAS122921) constituted a monophyletic clade (100% MLB and 1.00 BYPP, Figure 2) with the type strain of *Loculosulcatisspora thailandica* (MFLUCC 20-0108). Therefore, we introduce our new collections as *Loculosulcatisspora hongheensis* sp. nov. and amended the generic descriptions herein to accommodate its teleomorphic characteristics. The *Loculosulcatisspora* clade has a sister affiliation to *Parasulcatisspora clematidis*, which is known only from its teleomorph [56]. It could also be a species in *Loculosulcatisspora*. For us to

combine these two genera here would require an extensive taxonomic reassessment and describing anamorphic morphology, which is beyond the scope of the current study.

Tanaka et al. [46] introduced *Sulcatispora* to accommodate *S. acerina* and *S. berchemiae*, which were collected as saprobes on *Sapindaceae* (*Acer palmatum*) and *Rhamnaceae* (*Berchemia racemosa*) hosts. The teleomorph of the genus is characterized by globose, subglobose to hemispherical and immersed to erumpent ascomata, short papillate, central ostioles with periphyses, peridium composed of several layers of compressed angular cells, trabeculate pseudoparaphyses, clavate asci with a short pedicel and fusiform, 1-septate, hyaline ascospores with a large sheath. The anamorph is characteristic with globose, pycnidial conidiomata, cylindrical and annellidic conidiogenous cells, ellipsoid, yellowish brown, multi-septate conidia with striate ornamentation. These teleomorphic characteristics are somewhat similar to those in *Massarina*. The most distinctive feature of *Sulcatispora* is the longitudinal striae on the surface of conidia. Some species in *Barriopsis* (e.g., *B. iraniana*), *Dwiroopa* (e.g., *D. ramya*), *Endomelanconium* (e.g., *E. pini*), *Lasiodiplodia* (e.g., *L. theobromae*), *Mucoharknessia* (e.g., *M. cortaderiae*), *Neodeightonia* (e.g., *N. phoenicum*) and *Phaeophleospora* (e.g., *P. striae*) are known to have such conidia [61].

In the present study, we identified five genera and eight species associated with dead woody litter in Yunnan, China. Among them, one genus and four species are new to science. Our results emphasize that Yunnan Province has not yet been properly studied and is an open field for new fungal discoveries.

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References

1. Hawksworth, D.L.; Lücking, R. Fungal Diversity Revisited: 2.2 to 3.8 Million Species. *Microbiol. Spectr.* **2017**, *5*, 1–17. [[CrossRef](#)] [[PubMed](#)]
2. Bánki, O.; Roskov, Y.; Döring, M.; Ower, G.; Vandepitte, L.; Hobern, D.; Remsen, D.; Schalk, P.; DeWalt, R.E.; Keping, M.; et al. *Catalogue of Life Checklist (Version 2022-01-14)*; Catalogue of Life: Leiden, NL, USA, 2022. [[CrossRef](#)]
3. Wanasinghe, D.N.; Mortimer, P.E.; Bezerra, J. Editorial: Fungal Systematics and Biogeography. *Front. Microbiol.* **2022**, *12*, 827725. [[CrossRef](#)] [[PubMed](#)]
4. Hawksworth, D.L. The magnitude of fungal diversity: The 1.5 million species estimate revisited. *Mycol. Res.* **2001**, *105*, 1422–1432. [[CrossRef](#)]
5. Wanasinghe, D.N.; Wijayawardene, N.N.; Xu, J.; Cheewangkoon, R.; Mortimer, P.E. Taxonomic novelties in *Magnolia*-associated pleosporalean fungi in the Kunming Botanical Gardens (Yunnan, China). *PLoS ONE* **2020**, *15*, e0235855. [[CrossRef](#)] [[PubMed](#)]

6. Hyde, K.D.; Norphanphoun, C.; Chen, J.; Dissanayake, A.J.; Doilom, M.; Hongsanan, S.; Jayawardena, R.S.; Jeewon, R.; Perera, R.H.; Thongbai, B.; et al. Thailand's amazing diversity: Up to 96% of fungi in northern Thailand may be novel. *Fungal Divers.* **2018**, *93*, 215–239. [[CrossRef](#)]
7. Pölme, S.; Abarenkov, K.; Nilsson, R.H.; Lindahl, B.D.; Clemmensen, K.E.; Kauserud, H.; Nguyen, N.; Kjøller, R.; Bates, S.T.; Baldrian, P.; et al. FungalTraits: A user-friendly traits database of fungi and fungus-like stramenopiles. *Fungal Divers.* **2020**, *105*, 1–16. [[CrossRef](#)]
8. Schmit, J.P.; Mueller, G.M. An estimate of the lower limit of global fungal diversity. *Biodivers. Conserv.* **2007**, *16*, 99–111. [[CrossRef](#)]
9. Dighton, J. *Fungi in Ecosystem Processes*, 2nd ed.; CRC Press: Boca Raton, FL, USA, 2016; 434p. [[CrossRef](#)]
10. Hongsanan, S.; Hyde, K.D.; Phookamsak, R.; Wanasinghe, D.N.; McKenzie, E.H.C.; Sarma, V.V.; Boonmee, S.; Lücking, R.; Bhat, D.J.; Liu, N.G. Refined families of *Dothideomycetes*: *Dothideomycetidae* and *Pleosporomycetidae*. *Mycosphere* **2020**, *11*, 1553–2107. [[CrossRef](#)]
11. Mortimer, P.E.; Jeewon, R.; Xu, J.C.; Lumyong, S.; Wanasinghe, D.N. Morpho-phylo taxonomy of novel dothideomycetous fungi associated with dead woody twigs in Yunnan Province, China. *Front. Microbiol.* **2021**, *12*, e654683. [[CrossRef](#)]
12. Wijayawardene, N.N.; Hyde, K.D.; Dai, D.Q.; Sánchez-García, M.; Goto, B.T.; Saxena, R.K.; Erdoğan, M.; Selçuk, F.; Rajeshkumar, K.C.; Aptroot, A.; et al. Outline of Fungi and fungus-like taxa-2021. *Mycosphere* **2022**, *13*, 53–453. [[CrossRef](#)]
13. Jiang, H.B.; Hyde, K.D.; Jayawardena, R.S.; Doilom, M.; Xu, J.; Phookamsak, R. Taxonomic and phylogenetic characterizations reveal two new species and two new records of *Roussioella* (*Roussioellaceae*, *Pleosporales*) from Yunnan, China. *Mycol. Prog.* **2019**, *18*, 577–591. [[CrossRef](#)]
14. Thiagaraja, V.; Hyde, K.D.; Wanasinghe, D.N.; Worthy, F.R.; Karunarathna, S.C. Addition to *Melanommataceae*: A new geographical record of *Alpinaria rhododendri* from Shangri La, China. *Asian J. Mycol.* **2020**, *3*, 335–344. [[CrossRef](#)]
15. Wanasinghe, D.N.; Mortimer, P.E.; Xu, J. Insight into the systematics of microfungi colonizing dead woody twigs of *Dodonaea viscosa* in Honghe (China). *J. Fungi* **2021**, *7*, 180. [[CrossRef](#)] [[PubMed](#)]
16. Dissanayake, L.S.; Maharachchikumbura, S.S.N.; Mortimer, P.E.; Hyde, K.D.; Kang, J.C. *Acrocordiella yunnanensis* sp. nov. (*Requienellaceae*, *Xylariales*) from Yunnan, China. *Phytotaxa* **2021**, *487*, 103–113. [[CrossRef](#)]
17. Ren, G.C.; Wanasinghe, D.N.; Monkai, J.; Mortimer, P.E.; Hyde, K.D.; Xu, J.C.; Pang, A.; Gui, H. Novel saprobic *Hermatomyces* species (*Hermatomycetaceae*, *Pleosporales*) from China (Yunnan Province) and Thailand. *MycoKeys* **2021**, *82*, 57–79. [[CrossRef](#)]
18. Yang, E.; Tibpromma, S.; Karunarathna, S.C.; Phookamsak, R.; Xu, J.; Zhao, Z.; Karunanayake, C.; Promputtha, I. Taxonomy and phylogeny of novel and extant taxa in *Pleosporales* associated with *Mangifera indica* from Yunnan, China (Series I). *J. Fungi* **2022**, *8*, 152. [[CrossRef](#)]
19. Xie, N.; Phookamsak, R.; Jiang, H.; Zeng, Y.; Zhang, H.; Xu, F.; Lumyong, S.; Xu, J.; Hongsanan, S. Morpho-molecular characterization of five novel taxa in *Parabambusicolaceae* (*Massarinaceae*, *Pleosporales*) from Yunnan, China. *J. Fungi* **2022**, *8*, 108. [[CrossRef](#)]
20. Su, H.; Hyde, K.D.; Maharachchikumbura, S.S.; Ariyawansa, H.A.; Luo, Z.; Promputtha, I.; Tian, Q.; Lin, C.; Shang, Q.; Zhao, Y. The families *Distoseptisporaceae* fam. nov., *Kirschsteinioteliaceae*, *Sporormiaceae* and *Torulaceae*, with new species from freshwater in Yunnan Province, China. *Fungal Divers.* **2016**, *80*, 375–409. [[CrossRef](#)]
21. Su, H.; Luo, Z.; Liu, X.; Su, X.; Hu, D.; Zhou, D.; Bahkali, A.H.; Hyde, K.D. *Lentithecium cangshanense* sp. nov. (*Lentitheciaceae*) from freshwater habitats in Yunnan Province, China. *Phytotaxa* **2016**, *267*, 61–69. [[CrossRef](#)]
22. Su, X.; Luo, Z.; Jeewon, R.; Bhat, D.J.; Bao, D.; Li, W.; Hao, Y.; Su, H.; Hyde, K.D. Morphology and multigene phylogeny reveal new genus and species of *Torulaceae* from freshwater habitats in northwestern Yunnan, China. *Mycol. Prog.* **2018**, *17*, 531–545. [[CrossRef](#)]
23. Luo, Z.; Bhat, D.J.; Jeewon, R.; Boonmee, S.; Bao, D.; Zhao, Y.; Chai, H.; Su, H.; Su, X.; Hyde, K.D. Molecular phylogeny and morphological characterization of asexual fungi (*Tubeufiaceae*) from freshwater habitats in Yunnan, China. *Cryptog. Mycol.* **2017**, *38*, 27–53. [[CrossRef](#)]
24. Luo, Z.L.; Hyde, K.D.; Liu, J.K.; Bhat, D.J.; Bao, D.F.; Li, W.L.; Su, H.Y. Lignicolous freshwater fungi from China II: Novel *Distoseptispora* (*Distoseptisporaceae*) species from northwestern Yunnan Province and a suggested unified method for studying lignicolous freshwater fungi. *Mycosphere* **2018**, *9*, 444–461. [[CrossRef](#)]
25. Huang, S.; Maharachchikumbura, S.S.; Jeewon, R.; Bhat, D.J.; Chomnunti, P.; Hyde, K.D.; Lumyong, S. Morphological and molecular taxonomy of *Jahnula dianchia* sp. nov. (*Jahnulales*) from submerged wood in Dianchi Lake, Yunnan China. *Mycol. Prog.* **2018**, *17*, 547–555. [[CrossRef](#)]
26. Wanasinghe, D.N.; Jeewon, R.; Gareth Jones, E.; Boonmee, S.; Kaewchai, S.; Manawasinghe, I.S.; Lumyong, S.; Hyde, K.D. Novel palmicolous taxa within *Pleosporales*: Multigene phylogeny and taxonomic circumscription. *Mycol. Prog.* **2018**, *17*, 571–590. [[CrossRef](#)]
27. Wanasinghe, D.N.; Bezerra, J.D.P.; Mortimer, P.E. (Eds.) *Fungal Systematics and Biogeography*; Frontiers Media SA: Lausanne, Switzerland, 2022; 446p. [[CrossRef](#)]
28. Crous, P.W.; Gams, W.; Stalpers, J.A.; Robert, V.; Stegehuis, G. MycoBank: An online initiative to launch mycology into the 21st century. *Stud. Mycol.* **2004**, *50*, 19–22.
29. Wanasinghe, D.N.; Phukhamsakda, C.; Hyde, K.D.; Jeewon, R.; Lee, H.B.; Jones, E.B.G.; Tibpromma, S.; Tennakoon, D.S.; Dissanayake, A.J.; Jayasiri, S.C.; et al. Fungal diversity notes 709–839: Taxonomic and phylogenetic contributions to fungal taxa with an emphasis on fungi on *Rosaceae*. *Fungal Divers.* **2018**, *89*, 1–236. [[CrossRef](#)]

30. White, T.J.; Bruns, T.; Lee, S.J.W.T.; Taylor, J.W. Amplification and direct sequencing of fungal ribosomal RNA genes for phylogenetics. *PCR Protoc. Appl.* **1990**, *18*, 315–322. [[CrossRef](#)]
31. Rehner, S.A.; Samuels, G.J. Taxonomy and phylogeny of *Gliocladium* analysed from nuclear large subunit ribosomal DNA sequences. *Mycol. Res.* **1994**, *98*, 625–634. [[CrossRef](#)]
32. Vilgalys, R.; Hester, M. Rapid genetic identification and mapping of enzymatically amplified ribosomal DNA from several *Cryptococcus* species. *J. Bacteriol.* **1990**, *172*, 4238–4246. [[CrossRef](#)]
33. Rehner, S.A.; Buckley, E.A. Beauveria phylogeny inferred from nuclear ITS and EF1- α sequences: Evidence for cryptic diversification and links to *Cordyceps* teleomorphs. *Mycologia* **2005**, *97*, 84–98. [[CrossRef](#)]
34. Liu, Y.J.; Whelen, S.; Hall, B.D. Phylogenetic relationships among ascomycetes evidence from an RNA polymerase II subunit. *Mol. Biol. Evol.* **1999**, *16*, 1799–1808. [[CrossRef](#)] [[PubMed](#)]
35. Sung, G.H.; Sung, J.M.; Hywel-Jones, N.L.; Spatafora, J.W. A multi-gene phylogeny of *Clavicipitaceae* (Ascomycota, Fungi): Identification of localized incongruence using a combinational bootstrap approach. *Mol. Phylogenet. Evol.* **2007**, *44*, 1204–1223. [[CrossRef](#)] [[PubMed](#)]
36. Ren, G.; Wanasinghe, D.N.; Wei, D.; Monkai, J.; Yasanthika, E.; Gui, H.; Mortimer, P.E.; Xu, J.; Hyde, K.D. *Loculosulcatispora thailandica* gen. et sp. nov. (*Sulcatisporaceae*), saprobic on woody litter in Thailand. *Phytotaxa* **2020**, *475*, 67–78. [[CrossRef](#)]
37. Wanasinghe, D.N.; Mortimer, P.E.; Senwana, C.; Cheewangkoon, R. Saprobian *Dothideomycetes* in Thailand: *Phaeoseptum hydei* sp. nov., a new terrestrial ascomycete in *Phaeoseptaceae*. *Phytotaxa* **2020**, *449*, 149–163. [[CrossRef](#)]
38. Katoh, K.; Rozewicki, J.; Yamada, K.D. MAFFT online service: Multiple sequence alignment, interactive sequence choice and visualization. *Brief. Bioinform.* **2019**, *20*, 1160–1166. [[CrossRef](#)]
39. Hall, T.A. BioEdit: A user-friendly biological sequence alignment editor and analysis program for Windows 95/98/NT. *Nucl. Acids Symp. Ser.* **1999**, *41*, 95–98.
40. Nylander, J.A.A.; Wilgenbusch, J.C.; Warren, D.L.; Swofford, D.L. AWTY (are we there yet?): A system for graphical exploration of MCMC convergence in Bayesian phylogenetics. *Bioinformatics* **2008**, *24*, 581–583. [[CrossRef](#)]
41. Miller, M.A.; Pfeiffer, W.; Schwartz, T. Creating the CIPRES Science Gateway for inference of large phylogenetic trees. In Proceedings of the 2010 Gateway Computing Environments Workshop (GCE), New Orleans, LA, USA, 14 November 2010; pp. 1–8. [[CrossRef](#)]
42. Stamatakis, A. RAxML version 8: A tool for phylogenetic analysis and post-analysis of large phylogenies. *Bioinformatics* **2014**, *30*, 1312–1313. [[CrossRef](#)]
43. Rambaut, A. FigTree Version 1.4.0. Available online: <http://tree.bio.ed.ac.uk/software/figtree/> (accessed on 14 March 2022).
44. Phukhamsakda, C.; Jeewon, R.; McKenzie, E.H.C.; Xu, J.C. Morphology and phylogeny of *Phaeoseptum mali* sp. nov. (*Phaeoseptaceae*, *Pleosporales*) on bark of *Malus halliana*. *Asian J. Mycol.* **2019**, *2*, 118–128. [[CrossRef](#)]
45. Liu, N.G.; Hyde, K.D.; Bhat, D.J.; Jumpathong, J.; Liu, J.K. Morphological and phylogenetic studies of *Pleopunctum* gen. nov. (*Phaeoseptaceae*, *Pleosporales*) from China. *Mycosphere* **2019**, *10*, 757–775. [[CrossRef](#)]
46. Tanaka, K.; Hirayama, K.; Yonezawa, H.; Sato, G.; Toriyabe, A.; Kudo, H.; Hashimoto, A.; Matsumura, M.; Harada, Y.; Kurihara, Y.; et al. Revision of the *Massarineae* (*Pleosporales*, *Dothideomycetes*). *Stud. Mycol.* **2015**, *82*, 75–136. [[CrossRef](#)] [[PubMed](#)]
47. Hyde, K.D.; Chaiwan, N.; Norphanphoun, C.; Boonmee, S.; Camporesi, E.; Chethana, K.W.T.; Dayarathne, M.C.; De Silva, N.I.; Dissanayake, A.J.; Ekanayaka, A.H.; et al. *Mycosphere* notes 169–224. *Mycosphere* **2018**, *9*, 271–430. [[CrossRef](#)]
48. Hyde, K.D.; Jeewon, R.; Chen, Y.J.; Bhunjun, C.S.; Calabon, M.S.; Jiang, H.B.; Lin, C.G.; Norphanphoun, C.; Sysouphanthong, P.; Pem, D.; et al. The numbers of fungi: Is the descriptive curve flattening? *Fungal Divers.* **2020**, *103*, 219–271. [[CrossRef](#)]
49. Abdel-Wahab, M.A.; Jones, E.B.G. *Decaisnella formosa* sp. nov. (*Ascomycota*, *Massariaceae*) from an Australian sandy beach. *Can. J. Bot.* **2003**, *81*, 598–600. [[CrossRef](#)]
50. Mugambi, G.K.; Huhndorf, S.M. Molecular phylogenetics of *Pleosporales*: *Melanommataceae* and *Lophiostomataceae* recircumscribed (*Plesporomycetidae*, *Dothideomycetes*, *Ascomycota*). *Stud. Mycol.* **2009**, *64*, 103–121. [[CrossRef](#)] [[PubMed](#)]
51. Zhang, Y.; Fournier, J.; Phookamsak, R.; Bahkali, A.H.; Hyde, K.D. *Halotthiaceae* fam. nov. (*Pleosporales*) accommodates the new genus *Phaeoseptum* and several other aquatic genera. *Mycologia* **2013**, *105*, 603–609. [[CrossRef](#)] [[PubMed](#)]
52. Dayarathne, M.C.; Jones, E.B.G.; Maharachchikumbura, S.S.N.; Devadatha, B.; Sarma, V.V.; Khongphinitbunjong, K.; Chomnunti, P.; Hyde, K.D. Morpho-molecular characterization of microfungi associated with marine based habitats. *Mycosphere* **2020**, *11*, 1–188. [[CrossRef](#)]
53. Species Fungorum. Available online: <http://www.speciesfungorum.org/Names/Names.asp> (accessed on 8 March 2022).
54. Koukol, O.; Delgado, G. Why morphology matters: The negative consequences of hasty descriptions of putative novelties in asexual ascomycetes. *IMA Fungus* **2021**, *12*, 26. [[CrossRef](#)]
55. Hyde, K.D.; Tennakoon, D.S.; Jeewon, R.; Bhat, D.J.; Maharachchikumbura, S.S.; Rossi, W.; Leonardi, M.; Lee, H.B.; Mun, H.Y.; Houbraken, J.; et al. Fungal diversity notes 1036–1150: Taxonomic and phylogenetic contributions on genera and species of fungal taxa. *Fungal Divers.* **2019**, *96*, 1–242. [[CrossRef](#)]
56. Phukhamsakda, C.; McKenzie, E.H.C.; Phillips, A.J.L.; Jones, E.B.G.; Bhat, D.J.; Stadler, M.; Bhunjun, C.S.; Wanasinghe, D.N.; Thongbai, B.; Camporesi, E.; et al. Microfungi associated with *Clematis* (*Ranunculaceae*) with an integrated approach to delimiting species boundaries. *Fungal Divers.* **2020**, *102*, 1–203. [[CrossRef](#)]

57. Boonmee, S.; Wanasinghe, D.N.; Calabon, M.S.; Huanraluek, N.; Chandrasiri, S.K.U.; Jones, E.B.G.; Rossi, W.; Leonardi, M.; Singh, S.K.; Rana, S.; et al. Fungal diversity notes 1387–1511: Taxonomic and phylogenetic contributions on genera and species of fungal taxa. *Fungal Divers.* **2021**, *111*, 1–335. [[CrossRef](#)] [[PubMed](#)]
58. Crous, P.W.; Wingfield, M.J.; Schumacher, R.K.; Summerell, B.A.; Giraldo, A.; Gené, J.; Guarro, J.; Wanasinghe, D.N.; Hyde, K.D.; Camporesi, E.; et al. Fungal planet description sheets: 281–319. *Persoonia* **2014**, *33*, 212–289. [[CrossRef](#)] [[PubMed](#)]
59. Phukhamsakda, C.; Bhat, D.J.; Hongsanan, S.; Tibpromma, S.; Yang, J.B.; Promputtha, I. *Magnicamarosporium diospyricola* sp. nov. (*Sulcatisporaceae*) from Thailand. *Mycosphere* **2017**, *8*, 512–520. [[CrossRef](#)]
60. Rupcic, Z.; Chepkirui, C.; Hernández-Restrepo, M.; Crous, P.W.; Luangsa-Ard, J.J.; Stadler, M. New nematocidal and antimicrobial secondary metabolites from a new species in the new genus, *Pseudobambusicola thailandica*. *MycKeys* **2018**, *33*, 1–23. [[CrossRef](#)] [[PubMed](#)]
61. Wijayawardene, N.N.; Hyde, K.D.; Wanasinghe, D.N.; Papizadeh, M.; Goonasekara, I.D.; Camporesi, E.; Bhat, D.J.; McKenzie, E.H.C.; Phillips, A.J.L.; Diederich, P.; et al. Taxonomy and phylogeny of dematiaceous coelomycetes. *Fungal Divers.* **2016**, *77*, 1–316. [[CrossRef](#)]