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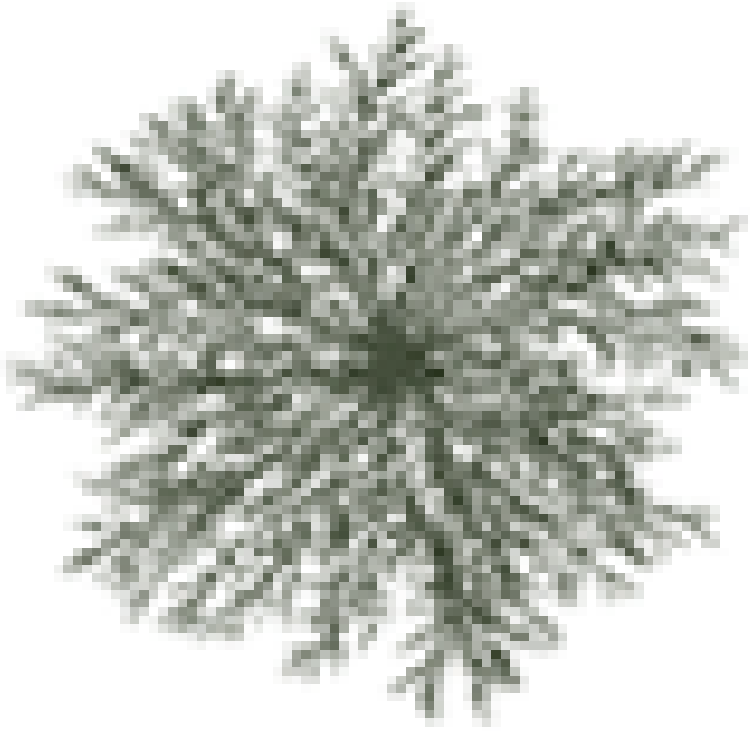

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Re-examination of a rare protosteloid amoeba *Schizoplasmodiopsis micropunctata*, and the revision of *Tychosporium* (*Cavosteliida, Variosea, Amoebozoa*)

Yoshiaki Iwamoto, Yousuke Degawa, Takeshi Nakayama

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Rhodocybe subasyae, a new species of *Rhodocybe* sect. *Rufobrunnea* (*Entolomataceae, Agaricales*) from northeast China

Ya-Li Sun, Tolgor Bau

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Molecular phylogeny and morphology reveal a new wood-rotting fungal species, *Sistotrema yunnanense* sp. nov. from the Yunnan-Guizhou Plateau

Li-Qiong Cai, Chang-Lin Zhao

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Wood-rotting fungi are important components of woody plant ecosystems and play an active role in the decomposition and turnover of nutrients from wood, and are among the major groups of *Basidiomycota*. In this study, a new species of wood-rotting fungus, *Sistotrema yunnanense*, was proposed based on morphological characteristics and molecular evidence. It is characterized by resupinate basidiomata, a monomitic hyphal system having generative hyphae with clamp connections, suburniform to urniform basidia, and short-cylindrical to oblong ellipsoid basidiospores (4.5-6.5 × 3-4 µm). Phylogenetic analyses performed using the large subunit nuc rDNA indicated that *S. yunnanense* was nested within the genus *Sistotrema s.l.* of the family *Hydnaceae*, within the order *Cantharellales*.

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Short communication

Molecular phylogeny and morphology reveal a new wood-rotting fungal species, *Sistotrema yunnanense* sp. nov. from the Yunnan-Guizhou Plateau

Li-Qiong Cai[a], Chang-Lin Zhao[a, b, c*]

[a] *Key Laboratory of Forest Disaster Warning and Control in Universities of Yunnan Province, Southwest Forestry University, Kunming 650224, P.R. China*
[b] *Yunnan Key Laboratory for Fungal Diversity and Green Development, Kunming Institute of Botany, Chinese Academy of Science, Kunming 650201, P.R. China*
[c] *Yunnan Key Laboratory of Gastrodia and Fungal Symbiotic Biology, Zhaotong University, 657000, P.R. China*

ABSTRACT

Wood-rotting fungi are important components of woody plant ecosystems and play an active role in the decomposition and turnover of nutrients from wood, and are among the major groups of *Basidiomycota*. In this study, a new species of wood-rotting fungus, *Sistotrema yunnanense*, was proposed based on morphological characteristics and molecular evidence. It is characterized by resupinate basidiomata, a monomitic hyphal system having generative hyphae with clamp connections, suburniform to urniform basidia, and short-cylindrical to oblong ellipsoid basidiospores (4.5–6.5 × 3–4 µm). Phylogenetic analyses performed using the large subunit nuc rDNA indicated that *S. yunnanense* was nested within the genus *Sistotrema s.l.* of the family *Hydnaceae*, within the order *Cantharellales*.

Keywords: *Cantharellales*, *Hydnaceae*, molecular systematics, taxonomy, Yunnan Province

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In forest ecosystems, fungi play essential ecological roles by driving carbon cycling in forest soils, mediating mineral uptake by plants, and alleviating carbon limitations (Tedersoo et al., 2014). Wood-rotting fungi are a highly diverse cosmopolitan group that are associated with a range of plants growing in boreal, temperate, subtropical, and tropical regions (Gilbertson & Ryvarden, 1987; Núñez & Ryvarden, 2001; Bernicchia & Gorjón, 2010; Dai, 2012; Ryvarden & Melo, 2014; Dai et al., 2015; Wu et al., 2020; Dai et al., 2021). The wood-rotting fungal genus, *Sistotrema* Fr. (*Hydnaceae*, *Cantharellales*), typified by *S. confluens* Pers., is a comparatively large genus belonging to the phylum *Basidiomycota*, and is morphologically characterized by resupinate or pileate-stipitate, soft basidiomes, smooth, grandinioid, hydnoid, or poroid hymenophore with various characteristic textures (pellicular, membranaceous, or ceraceous), a monomitic hyphal system with oily inclusions, urniform basidia, and smooth, thin-walled, basidiospores containing cytoplasmic oil droplets (Eriksson, Hjortstam, & Ryvarden, 1984; Bernicchia & Gorjón, 2010). Based on the MycoBank database (http://www.mycobank.org, accessed Jun 20, 2022) and the Index Fungorum (http://www.indexfungorum.org, accessed Jun 20, 2022), the genus *Sistotrema* has 204 registered species and intraspecies names, however the actual number of the species is 60 (Eriksson et al., 1984; Bernicchia & Gorjón, 2010; Sugawara et al., 2022).

These pioneering phylogenetic studies reveal that the genus *Sistotrema* is highly polyphyletic (Nilsson, Larsson, Larsson, & Kõljalg, 2006; Larsson, 2007, Hibbett et al., 2014), and were conducted before the advent of molecular systematics (Kotiranta & Larsson, 2013; Cao, Hu, Yu, Wei, & Yuan, 2021; Sugawara et al., 2022). Kotiranta and Larsson (2013) conducted preliminary phylogenetic research on *Sistotrema* and proposed a new species, *S. luteoviride* Kotir. & K.H. Larss., which clustered with *S. citriforme* (M.P. Christ) K.H. Larss. & Hjortsam with high bootstrap support (98%), and was grouped together with *S. pistilliferum* Hauerslev, *Membranomyces spurius* (Bourdot) Jülich, and two *Clavulina* J. Schröt. species in a moderately supported clade (79%). The nuclear rDNA sequence analysis of the phylogenetic diversity of bulbil-forming lichenicolous fungi in *Cantharellales* by Lawrey et al. (2016) revealed that the type species, *S. confluens*, grouped closely with the genus *Cantharellus* Adans. ex Fr. A comprehensive phylogenetic analysis based on a multiple-marker dataset for the entire *Hydnaceae* sensu stricto indicated that *Sistotrema* along with its sister genus *Hydnum* L. forms a fully supported lineage that is closely related to the genera *Craterellus* Pers. and *Cantharellus* (Cao et al., 2021). Phylogenetic trees obtained using the fungal nuc rDNA ITS and LSU and *rpb2* sequences showed that *Sistotrema* grouped with *Hydnum*; however, the generic boundary was unclear (Sugawara et al., 2022).

An undescribed fungal taxon was identified during investigations on wood-rotting fungi in southern China. The unknown taxon was placed in the genus *Sistotrema* based on analyses of the morphology and sequences of the large subunit (LSU) nuclear ribosomal RNA gene, and is proposed here as a new species, *S. yun-*

* Corresponding author. College of Biodiversity Conservation, Southwest Forestry University, Kunming 650224, China
Email address: fungi@swfu.edu.cn (Chang-Lin Zhao)
Tel: 13330492815

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nanense.

The specimens studied were deposited at the herbarium of Southwest Forestry University (SWFC), Kunming, Yunnan Province, P.R. China. Macroscopic descriptions are based on field notes. Color terms followed Petersen (1996). All the materials were examined under a Nikon 80i microscope. Drawings were made with the aid of a drawing tube. The measurements and drawings were made from slide preparations stained with cotton blue (0.1 mg aniline blue dissolved in 60 g pure lactic acid), Melzer's reagent (1.5 g potassium iodide, 0.5 g crystalline iodine, 22 g chloral hydrate, aq. dest. 20) and 5% (5 g potassiumhydroxide, 100 mL water) potassiumhydroxide. Spores were measured from sections cut from the hymenial layer, in presenting spore size data, 5% of the measurements excluded from each end of the range are shown in parentheses, and spore measurements were made in Cotton Blue. The following abbreviations were used: KOH = 5% potassium hydroxide, CB = Cotton Blue, CB– = acyanophilous, IKI = Melzer's reagent, IKI– = both inamyloid and indextrinoid, L = mean spore length (arithmetic average for all spores), W = mean spore width (arithmetic average for all spores), Q = variation in the L/W ratios between the specimens studied, n (a/b) = number of spores (a) measured from given number (b) of specimens.

Cetyltrimethylammonium bromide (CTAB) rapid plant genome extraction kit-DN14 (Aidlab Biotechnologies Co., Ltd, Beijing) was used to obtain genomic DNA from dried specimens, according to the manufacturer's instructions with some modifications that a small piece of dried fungal specimen (about 30 mg) was ground to powder with liquid nitrogen. The powder was transferred to a 1.5 mL centrifuge tube, suspended in 0.4 mL of lysis buffer, and incubated at 65 °C in a water bath for 60 min. After that, 0.4 mL phenol-chloroform (24:1) was added to each tube and the suspension was shaken vigorously. After centrifugation at 13,000 rpm for 5 min, 0.3 mL of supernatant was transferred to a new tube and mixed with 0.45 mL of binding buffer. The mixture was then transferred to an adsorbing column (AC) for centrifugation at 13,000 rpm for 0.5 min. Then, 0.5 mL of inhibitor removal fluid was added in AC for a centrifugation at 12,000 rpm for 0.5 min. After washing twice with 0.5 mL of washing buffer, the AC was transferred to a clean centrifuge tube, and 100 mL elution buffer was added to the middle of adsorbed film to elute the genome DNA. The nuc rDNA ITS region was amplified with primer pair ITS5 and ITS4 (White, Bruns, Lee, & Taylor, 1990). The nuc rDNA LSU region was amplified with primer pair LR0R and LR7 (Rehner & Samuels, 1994; Vilgalys & Hester, 1990). The PCR procedure for ITS was as follows: initial denaturation at 95 °C for 3 min, followed by 35 cycles at 94 °C for 40 s, 58 °C for 45 s and 72 °C for 1 min, and a final extension at 72 °C for 10 min. The PCR procedure for LSU marked as follows: initial denaturation at 94 °C for 1 min, followed by 35 cycles at 94 °C for 30 s, 48 °C 1 min and 72 °C for 1.5 min, and a final extension at 72 °C for 10 min. The PCR products were purified and directly sequenced at Kunming Tsingke Biological Technology Limited Company. All newly generated sequences were deposited in GenBank (Table 1).

DNA sequences were aligned in MAFFT 7 (https://mafft.cbrc.jp/alignment/server/) using the "G-INS-I" strategy for LSU, and manually adjusted in BioEdit (Hall, 1999). *Tilletiaria anomala* Bandoni & B.N. Johri and *Platygloea disciformis* (Fr.) Neuhoff were selected as an outgroup for LSU analysis based on previous study (Sugawara et al., 2022) (Fig. 1). The sequence alignment was deposited in TreeBASE (submission ID 29714).

Maximum parsimony (MP) analyses were applied to the LSU dataset sequences. Approaches to phylogenetic analysis followed Zhao and Wu (2017), and the tree construction procedure was performed in PAUP* version 4.0b10 (Swofford, 2002). All characters were equally weighted and gaps were treated as missing data. Trees were inferred using the heuristic search option with TBR branch swapping and 1,000 random sequence additions. Max-trees were set to 5,000, branches of zero length were collapsed and all parsimonious trees were saved. Clade robustness was assessed using a bootstrap (BT) analysis with 1,000 replicates (Felsenstein, 1985). Descriptive tree statistics tree length (TL), consistency index (CI), retention index (RI), rescaled consistency index (RC), and homoplasy index (HI) were calculated for each MP tree generated. Datamatrix was also analyzed using Maximum Likelihood (ML) approach with RAxML-HPC2 through the Cipres Science Gateway (www.phylo.org; Miller et al., 2009). Branch support (BS) for ML analysis was determined by 1,000 bootstrap replicates.

MrModeltest 2.3 (Nylander, 2004) was used to determine the best-fit evolution model for each data set for Bayesian inference (BI). BI was calculated with MrBayes 3.1.2 with a general time reversible (GTR+I+G) model of DNA substitution and a gamma distribution rate variation across sites (Ronquist & Huelsenbeck, 2003). Four Markov chains were run for 2 runs from random starting trees for 640 thousand generations for LSU (Fig. 1), and trees were sampled every 100 generations. The first one-fourth generations were discarded as burn-in. A majority rule consensus tree of all remaining trees was calculated. Branches were considered as significantly supported if they received the maximum likelihood bootstrap (BS) >70%, the maximum parsimony bootstrap (BT) >50%, or Bayesian posterior probabilities (BPP) >0.95.

The LSU dataset included sequences from 75 fungal specimens representing 59 taxa. The dataset had an aligned length of 1,622 characters in the dataset, of which 689 characters were constant, 463 were variable and parsimony-uninformative, and 470 were parsimony-informative. MP analysis yielded one equally parsimonious tree (TL = 2,903, CI = 0.471, HI = 0.528, RI = 0.611, RC = 0.288). The best model for LSU estimated and applied in Bayesian analysis had the following characteristics: GTR+I+G, lset nst = 6, rates = invgamma; prset statefreqpr = dirichlet (1,1,1,1). Bayesian analysis resulted in a similar topology with an average standard deviation of split frequencies (BI). The effective sample size (ESS) across the two runs was twice that of the average ESS (avg ESS) = 255. The phylogeny (Fig. 1) inferred from the LSU sequences showed that *S. yunnanense* nested in the family *Hydnaceae*.

Sistotrema yunnanense L.Q. Cai & C.L. Zhao, sp. nov. Figs. 2, 3. MycoBank no.: MB 844701.

Holotype: CHINA, Yunnan Province, Chuxiong, Zixishan National Forestry Park, E 101°24′, N 25°0′, alt. 1,950 m, on fallen branch of angiosperm, 2 Jul 2018, CLZhao 7357 (SWFC 007357). GenBank: LSU = ON810362, ITS = ON817194.

Etymology: *Yunnanense* (Lat.): referring to the province name of the type locality.

Basidiomata annual, resupinate, farinaceous to pruinose when fresh, becoming membranaceous upon drying, up to 11 cm long and 2 cm wide, 100–200 μm thick. Hymenial surface smooth, white when fresh, turning to pale cream upon drying, cracking with age. Margin narrow, slightly cream, fragile. Hyphal system monomitic; generative hyphae with clamp connections, colorless, thin- to slight thick-walled, branched, 2.5–5 μm in diam, frequently with oily contents, IKI–, CB–, tissues unchanged in KOH. Cystidia and cystidioles absent. Basidia suburniform to urniform, thin-walled, with four sterigmata and a basal clamp connection, with oily contents, 13.5–24 × 2.5–5.5 μm; basidioles abundant, in shape similar to basidia, but slightly smaller. Basidiospores subcylindrical to oblong ellipsoid, colorless, thin-walled, smooth, with oily contents,

Table 1. List of species, specimens, and GenBank accession numbers of sequences used in this study.

Species name	Sample no.	GenBank accession no.		References
		ITS	LSU	
Bergerella atrofusca	Berger 34240		MN902070	Lawrey et al. (2020)
Botryobasidium candicans	GEL3083		AJ406440	unpublished
B. conspersum	KHL11063		AY586657	Larsson et al. (2004)
B. subcoronatum	FCUG1286 SWE		AY647212	unpublished
Bryoclavula phycophila (Type)	TNS-F-79667		LC508118	Masumoto & Degawa (2020)
Burgella lutea	Etayo 27623		KC336075	Diederich et al. (2014)
Burgellopsis nivea (Type)	ATCC MYA-4209		KC336077	Diederich et al. (2014)
Burgoa verzuoliana (Type)	ATCC 24040		DQ915475	Lawrey et al. (2007)
Cantharellus alborufescens	JLS880b		KR677531	Olariaga et al. (2015)
C. ambohitantelyi (type)	BB 08.336		KF294656	Buyck et al. (2014)
Ceratobasidium bulbillifaciens	CBS 129339		KC336073	Diederich et al. (2014)
Clavulina cristata	EL95_97		AY586648	Larsson et al. (2004)
C. livida	MCCNNU140159		KT946799	He et al. (2016)
Hydnum albidum	MB11-6024/2		AY293186	Binder et al. (2005)
H. crocidens	PERTH08095981		KU612684	Feng et al. (2016)
H. elatum	FRI62309		KU612691	Feng et al. (2016)
H. repandum	KHL 8552		AF347095	Larsson et al. (2004)
Membranomyces delectabile	KHL11147		AY586688	Larsson et al. (2004)
M. spurius	Hjm 19169		KF218966	Kotiranta & Larsson (2013)
Minimedusa obcoronata	F-082, 316		AY004068	Platas et al. (2001)
M. polyspora (Type)	ATCC 24041		DQ915476	Lawrey et al. (2007)
Multiclavula corynoides	Lutzoni 930804-2, DUKE		U66440	Lutzoni (1997)
M. mucida	AFTOL-ID 1130		AY885163	unpublished
Neoburgoa freyi isolate	JL596-16		KX423755	Lawrey et al. (2016)
Platygloea disciformis	AFTOL-ID 710		AY629314	unpublished
Rogersiomyces malaysianus	LE-BIN 3507-10		KU820986	Psurtseva et al. (2016)
Sistotrema adnatum (Type)	FCUG 700		DQ898699	Moncalvo et al. (2006)
S. alboluteum	TAA167982		AY586713	Larsson et al. (2004)
S. alboluteum	TAA 180259		AJ606042	Nilsson et al. (2006)
S. alboluteum	MB6		KX358055	Stephenson et al. (2017)
S. albopallescens	KHL11070		AM259210	Nilsson et al. (2006)
S. athelioides (Type)	FCUG 701		DQ898700	Moncalvo et al. (2006)
S. biggsiae (Type)	FCUG 782		DQ898697	Moncalvo et al. (2006)
S. brinkmannii	FCUG 2198		DQ898705	Moncalvo et al. (2006)
S. brinkmannii	FCUG 2055		DQ898706	Moncalvo et al. (2006)
S. brinkmannii	FCUG 2217		DQ898709	Moncalvo et al. (2006)
S. brinkmannii	aurim1111		JQ912675	Menkis et al. (2006)
S. chloroporum	TUMH:64400		LC642058	Sugawara et al. (2022)
S. chloroporum	TUMH:64396		LC642054	Sugawara et al. (2022)
S. citriforme	KHL15898		KF218962	Kotiranta & Larsson (2013)
S. confluens	PV174		AY586712	Larsson et al. (2004)
S. confluens	FCUG 298		DQ898711	Moncalvo et al. (2006)
S. coroniferum	KH Larsson s.n.		KF218968	Kotiranta & Larsson (2013)
S. coroniferum	Herbarium GB-BN-2		AM259215	Nilsson et al. (2006)
S. coronilla	NH7598		AF506475	Larsson et al. (2004)
S. coronilla	AFTOL-ID 618		DQ457641	Matheny et al. (2006)
S. farinaceum (Type)	FCUG 659		DQ898707	Moncalvo et al. (2006)
S. flavorhizomorphae	TUMH:64401		LC642059	Sugawara et al. (2022)
S. flavorhizomorphae	TUMH:64402		LC642060	Sugawara et al. (2022)
S. hypogaeum	CBS:393.63		MH869925	Vu et al. (2019)
S. hypogaeum (Type)	CBS:394.63		MH869926	Vu et al. (2019)
S. luteoviride (Type)	HK23176		KF218963	Kotiranta & Larsson (2013)
S. muscicola	KHL8791		AF506474	Larsson & Larsson (2003)
S. muscicola	KHL 11721		AJ606040	Nilsson et al. (2006)
S. oblongisporum	KHL 14077		KF218970	Kotiranta & Larsson (2013)
S. oblongisporum	KHL 11189		GQ162819	Kotiranta et al. (2011)
S. oblongisporum	GEL2125		DQ898728	Moncalvo et al. (2006)
S. oblongisporum	FCUG 1490		DQ898702	Moncalvo et al. (2006)
S. oblongisporum	KHL 14077		KF218970	Kotiranta & Larsson (2013)
S. octosporum	FCUG 2822		DQ898698	Moncalvo et al. (2006)
S. octosporum	CBS:126038		MH875510	Vu et al. (2019)
S. pistilliferum	EL 28/10		KF218964	Kotiranta & Larsson (2013)
S. raduloides	LR 44004		KF218969	Kotiranta & Larsson (2013)
S. raduloides	FCUG 1695		DQ898710	Moncalvo et al. (2006)
S. resinicystidium	FCUG 2188		DQ898708	Moncalvo et al. (2006)
S. sernanderi	CBS 926.70		AF518650	Hibbett & Binder (2002)
S. sernanderi	FCUG1049 SWE		AY647215	unpublished
S. subconfluens	Dai 12578		JX076811	Zhou & Qin (2013)
S. subconfluens (Type)	Dai 12577		JX076810	Zhou & Qin (2013)
S. yunnanense	CLZhao 7341 (SWFC007341)	ON817192	ON810360	Present study
S. yunnanense	CLZhao 7355 (SWFC007355)	ON817193	ON810361	Present study
S. yunnanense	CLZhao 7395 (SWFC007395)	ON817195	ON810363	Present study
S. yunnanense (Type)	CLZhao 7357 (SWFC007357)	ON817194	ON810362	Present study
Sistotremella perpusilla	CBS:126048		MH875516	Vu et al. (2019)
Thanatephorus cucumeris	AG8		AF354068	Gonzalez et al. (2001)
T. theobromae	Sulawesi-10		HQ424242	Samuels et al. (2012)
Tilletiaria anomala	AFTOL-ID 865		AY745715	unpublished
Tulasnella cystidiophora	KW 2871		AY585831	Shefferson et al. (2005)
T. eremophila	13062MD		KJ701189	Crous et al. (2015)

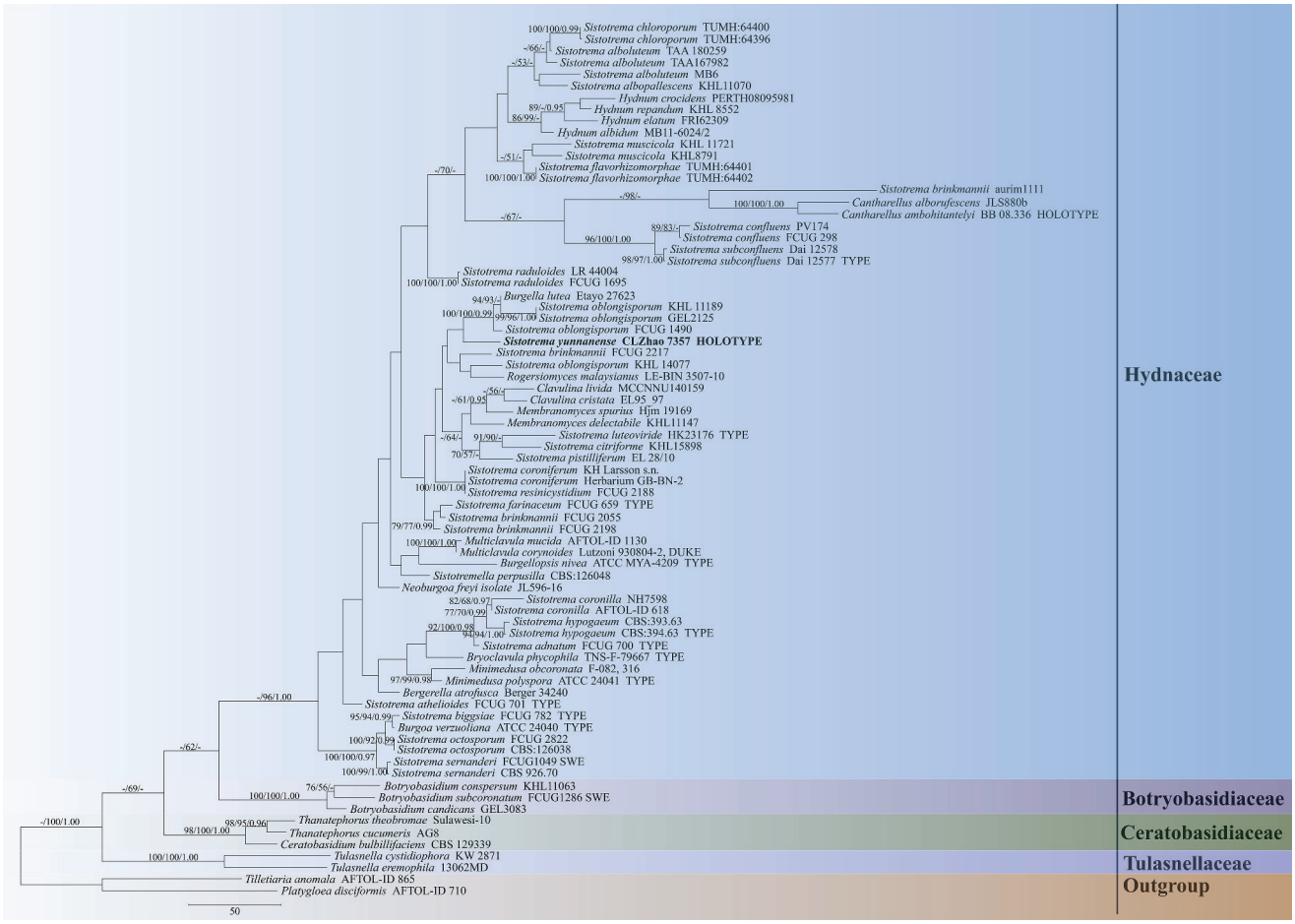

Fig. 1 – Maximum Parsimony strict consensus tree showing the phylogeny of a new *Sistotrema* species and related species in *Hydnaceae s.l.* based on nuc rDNA LSU sequences. Branches are labelled with maximum likelihood bootstrap > 70%, parsimony bootstrap proportions > 50% and Bayesian posterior probabilities > 0.95, respectively.

IKI–, CB–, (4–)4.5–6.5(–7) × (2.5–)3–4(–4.5) µm, L = 5.45 µm, W = 3.67 µm, Q = 1.43–1.51 (n = 120/4).

Type of rot: White rot.

Additional specimens examined (paratypes): CHINA, Yunnan Province, Chuxiong, Zixishan Forestry Park, E 101°24′, N 25°0′, alt. 1,950 m, on fallen branch of angiosperm, 2 Jul 2018, CLZhao 7355 (SWFC 007355), CLZhao 7341 (SWFC 007341), CLZhao 7395 (SWFC 007395).

In this study, a new species, *Sistotrema yunnanense* was described based on phylogenetic analyses and morphological characteristics.

The nuc rDNA LSU analysis showed that *S. yunnanense* belongs to *Hydnaceae* (Fig. 1) but not to *Sistotrema* s.s. clade (Sugawara et al., 2022), i.e., mycorrhizal lineage.

Morphologically, *Sistotrema brinkmannii* (Bres.) J. Erikss. differs from *S. yunnanense* by its rough, white to whitish gray hymenial surface having small aculei, and smaller basidiospores (3.4–4.6 × 1.9–3.0 µm; Dhingra, Priyanka, & Singh, 2009).

Sistotrema yunnanense is similar to *S. diademiferum* (Bourdot & Galzin) Donk and *S. porulosum* Hallenb in having farinaceous to pruinose hymenophores. However, *S. diademiferum* differs from *S. yunnanense* by its brownish-gray hymenial surface, thin-walled generative hyphae, and ellipsoid to ovoid, smaller basidiospores (3–5 × 2–3 µm; Kaur, Kaur, Singh, & Dhingra, 2018); *S. porulosum* differs in its smooth to porulose hymenophore with the grayish-white hymenial surface, basidia with 6–8 sterigmata, and narrowly ellipsoid to allantoid, smaller basidiospores (4–5 × 1.9–2.4 µm; Kaur et al., 2018).

Sistotrema yunnanense is similar to *S. confluens* Pers. and *S. subconfluens* L.W. Zhou in having similar morphological characteristics, namely, subcylindrical to oblong ellipsoid basidiospores. However, *S. confluens* differs from *S. yunnanense* by having brittle, tomentose hymenophore with the whitish to cream hymenial surface and narrower basidiospores (5–6 × 2–3 µm; Piątek & Cabała, 2002); *S. subconfluens* differs from *S. yunnanense* by having pileate basidiomata with buff to cinnamon buff hymenial surface and smaller basidiospores (3.9–4.2 × 2–2.3 µm; Zhou & Qin, 2013). Both species belongs to other linage "*Sistotrema* s.s.".

Wood-rotting fungi are an extensively studied group of *Basidiomycota* (Núñez & Ryvarden, 2001; Dai, 2012; Ryvarden & Melo, 2014; Dai et al., 2015; Wu et al., 2020; Cao et al., 2021; Luo, Chen, & Zhao, 2022; Qu, Wang, & Zhao, 2022); however, the diversity of wood-rotting fungi in East Asia is remains poorly understood, especially in subtropical and tropical regions. Recently, many described taxa in this ecological group were recorded from these areas (Dai, 2012; Chen, Korhonen, Li, & Dai, 2014; Bian & Dai, 2015; Cui et al., 2019; Shen et al., 2019; Zhu, Song, Zhou, Si, & Cui, 2019; Sugawara et al., 2022), and our newly described taxon increases the number of corticioid fungal species in East Asia.

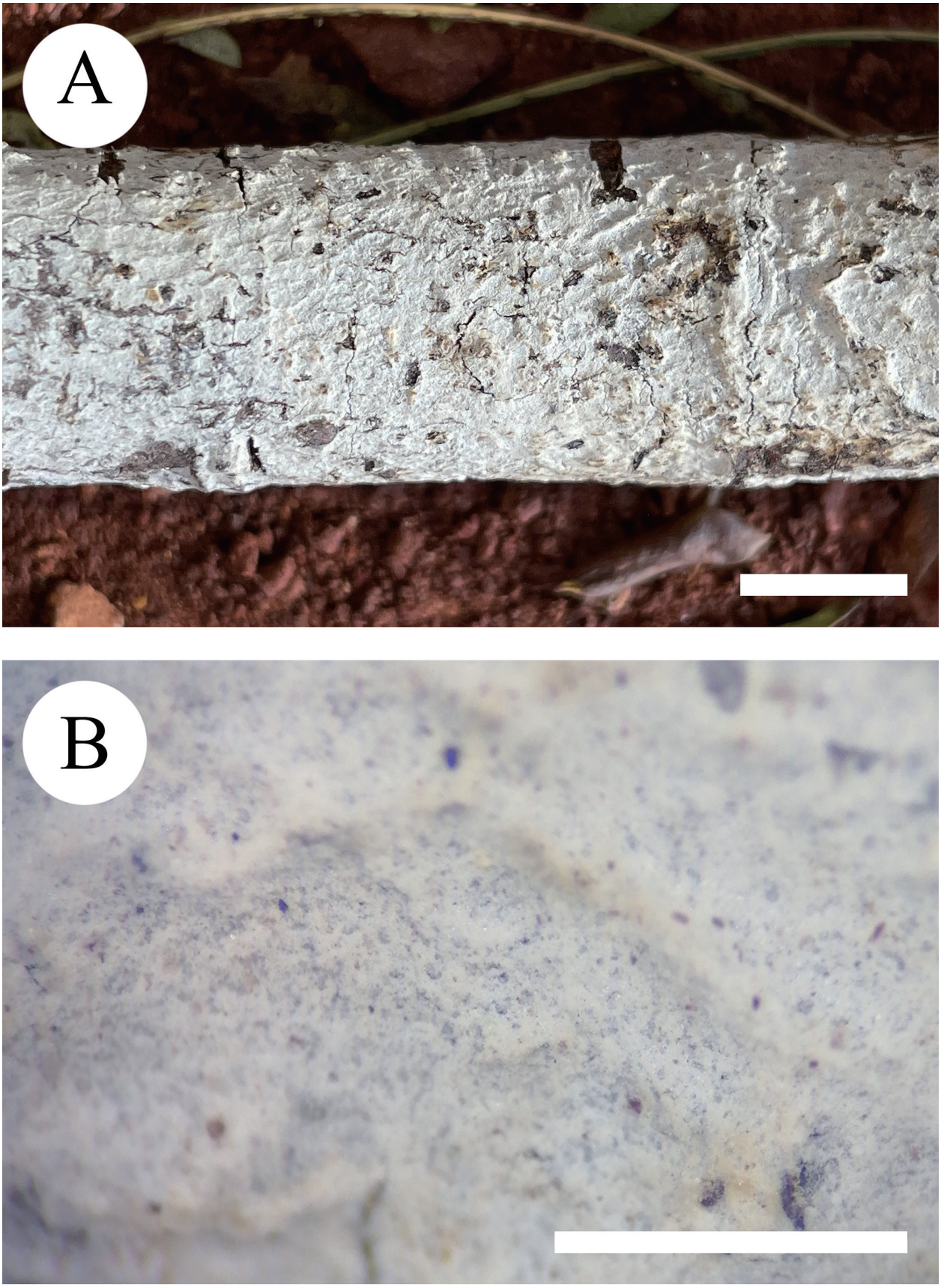

Fig. 2 – Macroscopic features of *Sistotrema yunnanense* (SWFC 007357, holotype): basidiomata. *Bars*: A 1 cm; B 1 mm.

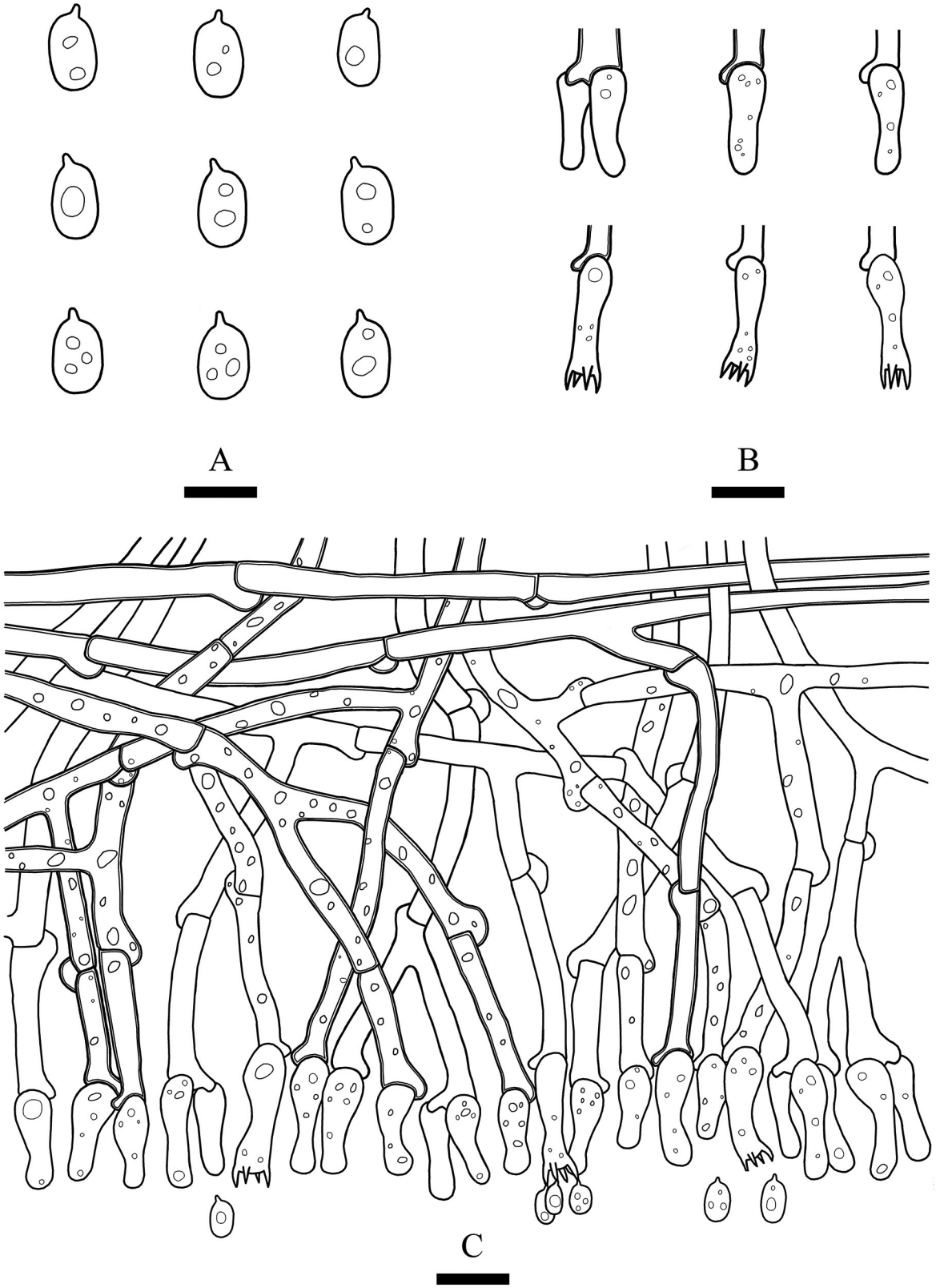

Fig. 3 – Microscopic features of *Sistotrema yunnanense* (SWFC 007357, holotype). A: Basidiospores. B: Basidia and basidioles. C: A part of the hymenial layer and subiculum. *Bars*: A 5 μm; B, C 10 μm.

Disclosure

The authors declare no conflict of interest. All the experiments undertaken in this study comply with the current laws of the People's Republic of China.

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