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Phlebia nigrodontea sp. nov. in Meruliaceae (Polyporales) with a black hymenial surface

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Abstract

A new wood-inhabiting fungus, *Phlebia nigrodontea*, is proposed based on a combination of morphological features and molecular evidence. The species is characterized by a grandinioid hymenophore with vinaceous brown to black colour, a monomitic hyphal system with clamped generative hyphae and ellipsoid, colourless, thin-walled, smooth basidiospores (3.9–4.9 × 2.3–3.1 µm). Sequences of ITS and LSU nrRNA gene regions of the studied samples were generated, and phylogenetic analyses carried out using maximum likelihood, maximum parsimony and Bayesian inference methods. The phylogenetic analyses based on the molecular data of ITS+nLSU sequences showed that *P. nigrodontea* nested within the phlebioid clade. A further investigation of more representative taxa from *Phlebia*, based on ITS+nLSU sequences, demonstrated that the species *P. nigrodontea* formed a monophyletic lineage with strong support (100% BS, 100% BT, 1.00 BPP) and closely grouped with *P. chrysocreas*.

Keywords: China, phylogeny, taxonomy, wood-rotting fungi

Introduction

Phlebia Fr. (1821: 426) was described by Fries (1821), and *P. radiata* Fr. (1821: 427) was designated the type species. It is a morphologically diverse genus characterized by resupinate or rarely pileate basidiocarps that have a subceraceous to subgelatinous consistency when fresh, and are membranaceous to coriaceous when dry; hymenophores that are smooth, tuberculate, phlebioid, odontioid, merulioid or poroid; a monomitic hyphal structure (rarely dimitic) with clamp connections or simple septa; narrowly clavate basidia and basidiospores that are hyaline, thin-walled, smooth, allantoid to ellipsoid, acyanophilous and negative in Melzer's reagent (Bernicchia & Gorjón 2010). *Phlebia* contains many species with more than 100 taxa accepted worldwide (Fries 1821, Ginns 1969, Nakasone & Burdsall 1984, 1995, Dhingra 1989, Nakasone 1997, 2002, 2003, 2009, Roberts 2000, Gilbertson & Hemmes 2004, Duhem & Michel 2007, Duhem 2009, 2013, Bernicchia & Gorjón 2010, Singh *et al.* 2010, Westphalen *et al.* 2010, Gorjón & Greslebin 2012, Binder *et al*. 2013, Floudas & Hibbett 2015, Kaur *et al*. 2017, Shen *et al*. 2018, Huang *et al*. 2020).

Earlier molecular studies placed *Phlebia* in the polyporoid clade (Hibbett & Thorn 2001), but later studies indicated a high phylogenetic diversity among corticioid homobasidiomycetes and that *Phlebia* should belong in a separate phlebioid clade (Larsson *et al*. 2004). Larsson (2007) presented a phylogenetic classification for corticioid fungi at the family level and demonstrated that *Phlebia* nested within the Meruliaceae. Using a multi-gene dataset, Binder *et al*. (2013) demonstrated that the type species, *P. radiata*, belonged to the phlebioid clade and appeared to group with *Ceraceomyces* Jülich (1972: 146), *Ceriporia* Donk (1933: 170) and *Ceriporiopsis* Domański (1963: 731). Also using the multi-gene datasets, Justo *et al*. (2017) revised the family-level classification of the Polyporales (Basidiomycota), including eighteen families and showed that *P. radiata* belonged to the family Meruliaceae and grouped with *Aurantiporus* Murrill (1905: 487) and *C. gilvescens* (Bres.) Domański (1963: 731).

Recently, we collected an undescribed taxon from Yunnan Province, P.R. China, that could not be assigned to any

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described species. Here we present morphological and molecular phylogenetic evidence that support the recognition of a new species in *Phlebia*, as *P. nigrodontea*.

Materials and methods

Morphological studies

The specimens studied are deposited in the herbarium of Southwest Forestry University (SWFC), Kunming, Yunnan Province, P.R. China. Macromorphological descriptions are based on field notes. Colour terms follow Petersen (1996). Micromorphological data were obtained from the dried specimens and observed under a light microscope following Dai (2012). The following abbreviations are used: KOH = 5% potassium hydroxide, CB = Cotton Blue, CB– = acyanophilous, IKI = Melzer's reagent, IKI– = both inamyloid and non-dextrinoid, L = mean spore length (arithmetic average of all spores), W = mean spore width (arithmetic average of 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.

Molecular techniques and phylogenetic analyses

The EZNA HP Fungal DNA Kit (Omega Biotechnologies Co., Ltd, Kunming) was used to obtain PCR products from dried specimens, according to the manufacturer's instructions with some modifications. The ITS region was amplified with primer pairs ITS5 and ITS4 (White *et al*. 1990). The nLSU region was amplified with primer pairs LR0R and LR7 (http://www.biology.duke.edu/fungi/mycolab/primers.htm). 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 of 72 °C for 10 min. The PCR procedure for nLSU was 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 of 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).

Sequencher 4.6 (GeneCodes, Ann Arbor, MI, USA) was used to edit the DNA sequences. Sequences from the Russulales were used as outgroup for the Polyporales (Polyporales dataset), and *Hydnophlebia* was the outgroup for the *Phlebia* species (*Phlebia* dataset). Sequences were aligned in MAFFT 7 (http://mafft.cbrc.jp/alignment/server/) using the "G-INS-I" strategy and manually adjusted in BioEdit (Hall 1999). Sequence alignments were deposited in TreeBase (submission ID 26629). Sequences of *Heterobasidion annosum* (Fr.) Bref. (1889: 154) and *Stereum hirsutum* (Willd.) Pers. (1800: 90) obtained from GenBank were used as outgroups to root trees following Binder *et al*. (2013) in the ITS+nLSU analysis (Fig. 1 in present study) and *Hydnophlebia chrysorhiza (*Torr.) Parmasto (1967: 384) obtained from GenBank was used as an outgroup to root trees following Floudas & Hibbett (2015) in the ITS+nLSU analyses (Fig. 2).

Maximum parsimony analysis was applied to the ITS+nLSU sequences for two matrices (Figs. 1, 2). Approaches to phylogenetic analysis followed Zhao & 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 1000 random sequence additions. Max-trees were set to 5000, 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 Maximum Parsimonious Tree (MPT) generated. Sequences were also analyzed using Maximum Likelihood (ML) with RAxML-HPC2 on Abe through the Cipres Science Gateway (www.phylo.org; Miller *et al*. 2009). Branch support for ML analysis was determined by 1000 bootstrap replicates.

MrModeltest 2.3 (Nylander 2004) was used to determine the best-fit evolution model for data set for Bayesian Inference (BI). Bayesian inference was calculated with MrBayes_3.1.2 using 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 500,000 generations (Fig. 1), for 3 million generations (Fig. 2) and trees were sampled every 100 generations. The first 25% of the generations were discarded as burn-in. A majority rule consensus tree of all remaining trees was calculated. Branches that received bootstrap support for maximum likelihood (BS) ≥75%, maximum parsimony (BT) ≥75%, and Bayesian posterior probabilities (BPP) ≥0.95 were considered significantly supported.

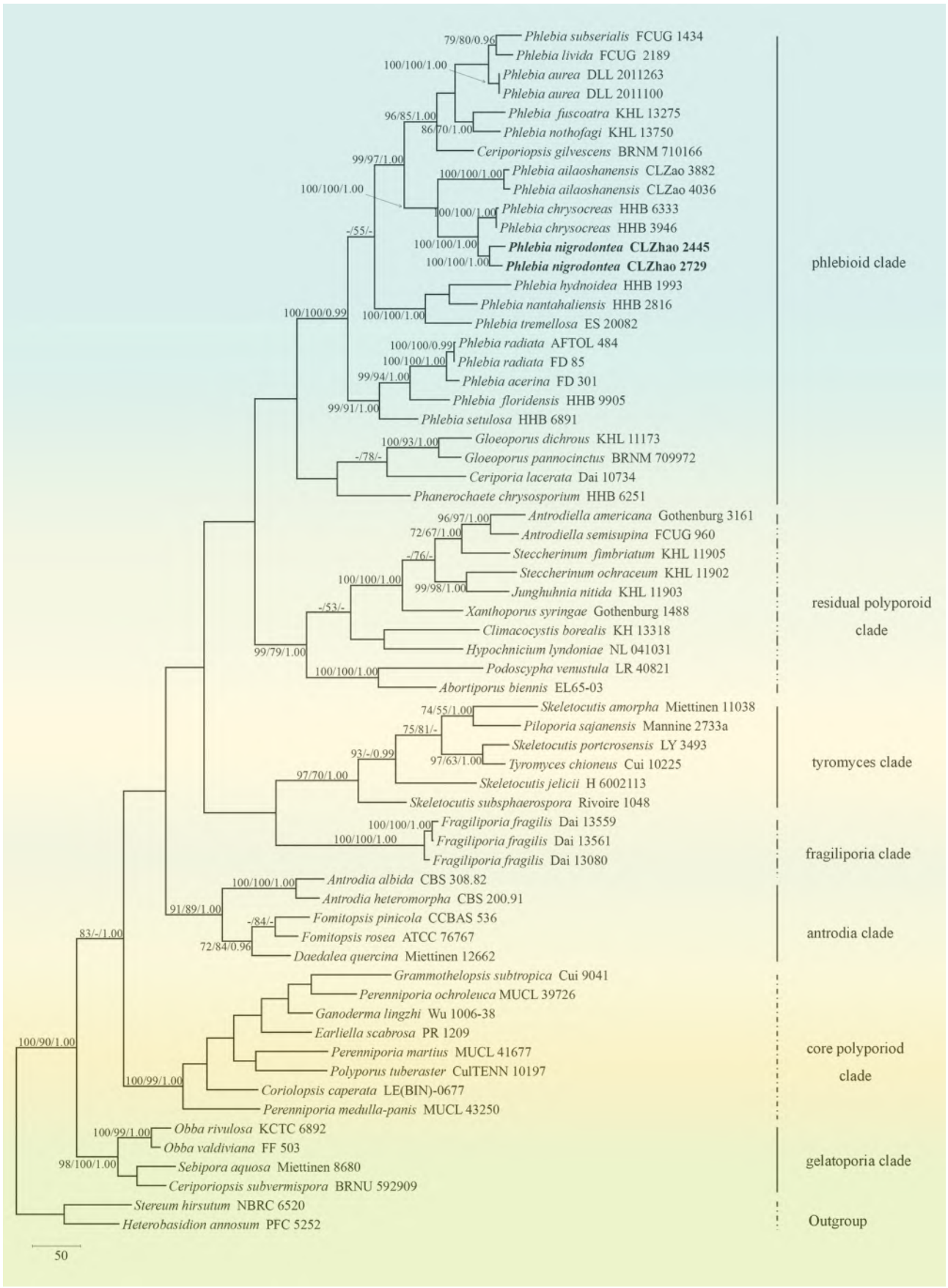

FIGURE 1. Maximum Parsimony strict consensus tree of Polyporales illustrating the phylogeny of *Phlebia nigrodontea* and related species based on ITS+nLSU sequences. Branches are labeled with maximum likelihood bootstrap higher than 70%, parsimony bootstrap proportions higher than 50% and Bayesian posterior probabilities more than 0.95 respectively. Clade names follow Binder *et al*. (2013).

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

Species name	Sample no.	GenBank accession no.		References
		ITS	nLSU	
Abortiporus biennis	EL65-03	JN649325	JN649325	Sjökvist *et al*. 2012
Antrodia albida	CBS 308.82	DQ491414	DQ491414	Kim *et al*. 2007
A. heteromorpha	CBS 200.91	DQ491415	DQ491415	Kim *et al*. 2007
Antrodiella americana	Gothenburg 3161	JN710509	JN710509	Binder *et al*. 2013
A. semisupina	FCUG 960	EU232182	EU232182	Binder *et al*. 2005
Ceriporia lacerata	Dai 10734	JX623916	JX623916	Jia *et al*. 2014
Ceriporiopsis gilvescens	BRNM 710166	FJ496684	FJ496684	Tomšovský *et al*. 2010
Climacocystis borealis	KHL 13318	JQ031126	JQ031126	Binder *et al*. 2013
Coriolopsis caperata	LE(BIN)-0677	AB158316	AB158316	Tomšovský *et al*. 2010
Daedalea quercina	Miettinen 12662	JX109855	JX109855	Binder *et al*. 2013
Earliella scabrosa	PR 1209	JN165009	JN165009	Binder *et al*. 2005
Fomitopsis pinicola	CCBAS 536	FJ608588	—	Homolka *et al*. 2010
F. rosea	ATCC 76767	DQ491410	DQ491410	Kim *et al*. 2007
Fragiliporia fragilis	Dai 13080	KJ734260	KJ734260	Zhao *et al*. 2015
F. fragilis	Dai 13559	KJ734261	KJ734261	Zhao *et al*. 2015
F. fragilis	Dai 13561	KJ734262	KJ734262	Zhao *et al*. 2015
Ganoderma lingzhi Sheng	Wu 1006-38	JQ781858	—	Zhao *et al*. 2015
Gelatoporia subvermispora	BRNU 592909	FJ496694	FJ496694	Tomšovský *et al*. 2010
Gloeoporus pannocinctus	BRNM 709972	EU546099	EU546099	Tomšovský *et al*. 2010
G. dichrous	KHL 11173	EU118627	EU118627	Binder *et al*. 2005
Grammothelopsis subtropica	Cui 9035	JQ845094	JQ845097	Zhao *et al*. 2015
Heterobasidion annosum	PFC 5252	KC492906	KC492906	Binder *et al*. 2013
Hornodermoporus martius	MUCL 41677	FJ411092	FJ411092	Robledo *et al*. 2009
Hydnophlebia chrysorhiza	FD 282	KP135338	KP135216	Floudas & Hibbett 2015
Hypochnicium lyndoniae	NL 041031	JX124704	JX124704	Binder *et al*. 2013
Junghuhnia nitida	KHL 11903	EU118638	EU118638	Binder *et al*. 2005
Obba rivulosa	KCTC 6892	FJ496693	FJ496693	Miettinen & Rajchenberg 2012
O. valdiviana	FF 503	HQ659235	HQ659235	Miettinen & Rajchenberg 2012
Perenniporia medulla-panis	MUCL 43250	FJ411087	FJ411087	Robledo *et al*. 2009
P. ochroleuca	MUCL 39726	FJ411098	FJ411098	Robledo *et al*. 2009
Phanerochaete chrysosporium	HHB 6251	KP135094	KP135094	Justo *et al*. 2017
Phlebia acanthocystis	FP 150571	KY948767	KY948844	Floudas & Hibbett 2015
P. acerina	FD 301	KP135378	KP135378	Justo *et al*. 2017
P. acerina	HHB 11146	KP135372	—	Floudas & Hibbett 2015
P. acerina	FP 135252	KP135371	—	Floudas & Hibbett 2015
P. ailaoshanensis	CLZhao 3882	MH784919	MH784929	Shen *et al*. 2018
P. ailaoshanensis	CLZhao 4036	MH784927	MH784927	Shen *et al*. 2018
P. aurea	DLL 2011100	KJ140614	—	Binder *et al*. 2013
P. aurea	DLL 2011263	KJ140747	KJ140747	Binder *et al*. 2013
P. aurea	FCUG 2767	HQ153409	HQ153409	Binder *et al*. 2013
P. aurea	RLG 5075	KY948759	—	Justo *et al*. 2017
P. brevispora	HHB 7030	KP135387	—	Floudas & Hibbett 2015
P. centrifuga	HHB 9239	KP135380	KP135380	Floudas & Hibbett 2015

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TABLE 1. (Continued)

Species name	Sample no.	GenBank accession no. ITS	nLSU	References
P. centrifuga	GB 1013	KP135379	—	Floudas & Hibbett 2015
P. chrysocreas	HHB 6333	KP135358	KP135358	Floudas & Hibbett 2015
P. chrysocreas	HHB 3946	KP135357	KP135357	Floudas & Hibbett 2015
P. chrysocreas	FP 102161	AY219367	—	Binder *et al*. 2005
P. floridensis	HHB 9905	KP135383	KP135383	Justo *et al*. 2017
P. floridensis	HHB 6466	KP135385	KP135385	Floudas & Hibbett 2015
P. floridensis	HHB 7175	KP135384	KP135384	Floudas & Hibbett 2015
P. floridensis	FP 102562T	KP135386	KP135386	Floudas & Hibbett 2015
P. fuscoatra	HHB 15354	KP135367	KP135367	Floudas & Hibbett 2015
P. fuscoatra	HHB 18642	KP135366	—	Floudas & Hibbett 2015
P. fuscoatra	FP 102173	KP135364	—	Floudas & Hibbett 2015
P. fuscoatra	KHL 13275	JN649352	JN649352	Tomšovský *et al*. 2010
P. hydnoidea	HHB 1993	KY948778	KY948778	Justo *et al*. 2017
P. lindtneri	GB 501	KY948772	KY948772	Justo *et al*. 2017
P. livida	FCUG 2189	AF141624	AF141624	Tomšovský *et al*. 2010
P. livida	FCUG 1290	HQ153414	—	Ghobad-Nejhad & Hallenberg 2012
P. ludoviciana	FD 427	KP135342	—	Floudas & Hibbett 2015
P. nantahaliensis	HHB 2816	KY948777	KY948777	Justo *et al*. 2017
P. nigrodontea	CLZhao 2445	MT896821	MT896818	Present study
P. nigrodontea	CLZhao 2548	MT896822	—	Present study
P. nigrodontea	CLZhao 2729	MT896823	MT896819	Present study
P. nigrodontea	CLZhao 2758	MT896824	—	Present study
P. nothofagi	HHB 4273	KP135369	KP135369	Floudas & Hibbett 2015
P. nothofagi	HHB 6906	KP135368	—	Floudas & Hibbett 2015
P. nothofagi	HHB 12067	KP135370	—	Floudas & Hibbett 2015
P. nothofagi	KHL 13750	GU480000	GU480000	Tomšovský *et al*. 2010
P. radiata	AFTOL 484	AY854087	AY854087	Binder *et al*. 2005
P. radiata	FD 85	KP135377	KP135377	Justo *et al*. 2017
P. radiata	CBS 285.56	MH857642	MH869187	Vu *et al*. 2019
P. rufa	HHB 14924	KP135374	KP135374	Floudas & Hibbett 2015
P. setulosa	HHB 6891	KP135382	KP135382	Justo *et al*. 2017
P. setulosa	PH 11749	GU461312	—	Binder *et al*. 2005
P. serialis	FCUG 2868	HQ153429	—	Ghobad-Nejhad & Hallenberg 2012
P. subochracea	HHB 8715	KY948770	KY948846	Floudas & Hibbett 2015
P. subserialis	FCUG 1434	AF141631	AF141631	Tomšovský *et al*. 2010
P. tuberculata	FCUG3157	HQ153427	—	Ghobad-Nejhad & Hallenberg 2012
P. tuberculata	MG127	HQ153424		Ghobad-Nejhad & Hallenberg 2012
P. tremellosa	ES 20082	JX109859	JX109859	Binder *et al*. 2013
P. tremellosa	CBS 217.56	MH857589	MH869138	Vu *et al*. 2019
P. uda	FP 101544	KP135361	KP135361	Floudas & Hibbett 2015
P. uda	FCUG 2452	AF141614	—	Parmasto & Hallenberg 2000
P. wuliangshanensis	CLZhao 3475	MK881897	MK881787	Huang *et al*. 2020
P. wuliangshanensis	CLZhao 3639	MK881898	MK881788	Huang *et al*. 2020
Piloporia sajanensis	Mannine 2733a	HQ659239	HQ659239	Tomšovský *et al*. 2010

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TABLE 1. (Continued)

Species name	Sample no.	GenBank accession no. ITS	nLSU	References
Polyporus tuberaster	CulTENN 10197	AF516596	AF516596	Binder *et al*. 2013
Sebipora aquosa	Miettinen 8680	HQ659240	HQ659240	Miettinen & Rajchenberg 2012
Skeletocutis amorpha	Miettinen 11038	FN907913	FN907913	Tomšovský *et al*. 2010
S. jelicii	H 6002113	FJ496690	FJ496690	Tomšovský *et al*. 2010
S. portcrosensis	LY 3493	FJ496689	FJ496689	Tomšovský *et al*. 2010
S. subsphaerospora	Rivoire 1048	FJ496688	FJ496688	Tomšovský *et al*. 2010
Steccherinum fimbriatum	KHL 11905	EU118668	EU118668	Tomšovský *et al*. 2010
S. ochraceum	KHL 11902	JQ031130	JQ031130	Tomšovský *et al*. 2010
Stereum hirsutum	NBRC 6520	AB733150	AB733325	Tomšovský *et al*. 2010
Tyromyces chioneus	Cui 10225	KF698745	KF698745	Zhao *et al*. 2015
Xanthoporus syringae	Gothenburg 1488	JN710607	JN710607	Tomšovský *et al*. 2010

Results

Molecular phylogeny

The ITS+nLSU dataset (Fig. 1) included sequences from 63 fungal specimens representing 56 species of the Polyporales. The dataset had an aligned length of 2219 characters, of which 1262 characters are constant, 269 are variable and parsimony-uninformative, and 688 are parsimony-informative. Maximum parsimony analysis yielded 2 equally parsimonious trees (TL = 5156, CI = 0.314, HI = 0.686, RI = 0.540, RC = 0.170). Best model for the ITS+nLSU dataset estimated and applied in the Bayesian analysis: GTR+I+G, lset nst = 6, rates = invgamma; prset statefreqpr = dirichlet (1,1,1,1). Bayesian analysis and ML analysis resulted in a similar topology as the MP analysis, with an average standard deviation of split frequencies = 0.008638 (BI).

The phylogenetic tree (Fig. 1) inferred from ITS+nLSU sequences, demonstrated seven major clades for 56 sampled species of the Polyporales, in which the new species *Phlebia nigrodontea* fell into the 'phlebioid' clade.

The ITS+nLSU dataset (Fig. 2) included sequences from 53 fungal specimens representing 27 species in *Phlebia*. The dataset had an aligned length of 2059 characters, of which 1577 characters are constant, 108 are variable and parsimony-uninformative, and 374 are parsimony-informative. Maximum parsimony analysis yielded 100 equally parsimonious trees (TL = 1368, CI = 0.527, HI = 0.473, RI = 0.779, RC = 0.411). Best model for the ITS+nLSU dataset estimated and applied in the Bayesian analysis: GTR+I+G, lset nst = 6, rates = invgamma; prset statefreqpr = dirichlet (1,1,1,1). Bayesian analysis and ML analysis resulted in a similar topology as the MP analysis, with an average standard deviation of split frequencies = 0.009431 (BI).

The phylogeny (Fig. 2) inferred from the combined ITS+nLSU sequences within the genus *Phlebia* was obtained for 53 fungal specimens representing 27 taxa, in which it revealed that the new species formed a monophyletic entity with a high 100% BS, 100% BT and 1.00 BPP and grouped with *P. chrysocreas* (Berk. & M.A. Curtis) Burds. (1975: 497).

Taxonomy

Phlebia nigrodontea C.L. Zhao & R.X. Huang, *sp. nov.* Figs. 3, 4
MycoBank no.: MB 836685

Holotype:—CHINA. Yunnan Province, Yuxi, Xinping County, Shimenxia Forestry Park, on angiosperm trunk, 21 August 2017, *CLZhao 2758* (SWFC!).

Etymology:—*nigrodontea* (Lat.): refers to the black hymenial surface.

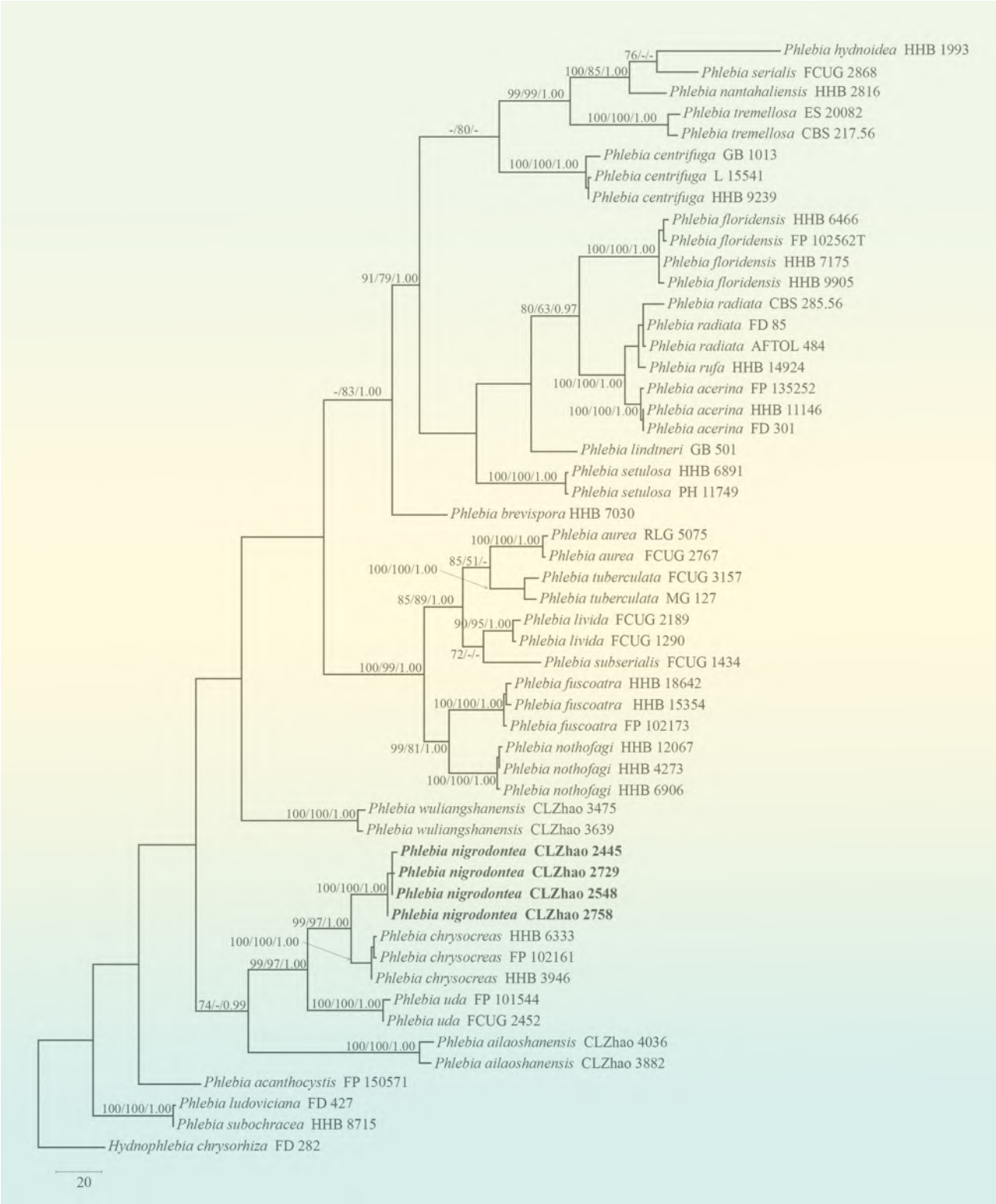

FIGURE 2. Maximum Parsimony strict consensus tree illustrating the phylogeny of *Phlebia nigrodontea* and related species in *Phlebia* based on ITS+nLSU sequences. Branches are labeled with maximum likelihood bootstrap higher than 70%, parsimony bootstrap proportions higher than 50% and Bayesian posterior probabilities more than 0.95, respectively.

Description:—*Basidiomata* annual, resupinate, subceraceous to ceraceous, without odour or taste when fresh, becoming coriaceous to rigid upon drying, up to 12 cm long, 1 mm thick. *Hymenial surface* grandinioid, vinaceous brown to black when fresh, having a uniform surface, becoming black and retracting into a netlike appearance upon drying, in which the hymenophore shows a netlike appearance with the lighter coloured subiculum showing through. *Hyphal system* monomitic; generative hyphae with clamp connections, colourless, sclerified, slightly thick-walled to thick-

walled, frequently branched, 1.5–4 µm in diameter, IKI–, CB–; tissues unchanged in KOH. *Cystidia* capitate, rare, 8.5–15.5 × 3.6–6.5 µm, apex capped with a dark gold-coloured resinous-like substance; presence of abundant resinous substances among hyphae and subhymenium; *basidia* clavate, 10–23.5 × 3–6 µm, four-spored and with a basal clamp connection; basidioles dominant, in shape similar to basidia, but slightly smaller. *Basidiospores* ellipsoidal, colourless, thin-walled, smooth, IKI–, CB–, 3.9–4.9 × 2.3–3.1(–3.8) µm, L = 4.42 µm, W = 2.61 µm, Q = 1.60–1.73 (n = 120/4).

Ecology and distribution:—Lignicolous, causing a white rot. Found in China.

Additional specimens (paratypes) examined:—CHINA. Yunnan Province, Yuxi, Xinping County, Mopanshan National Forestry Park, on angiosperm trunk, 19 August 2017, *CLZhao 2445*, 20 August 2017, *CLZhao 2548*, 21 August 2017, *CLZhao 2729* (SWFC!).

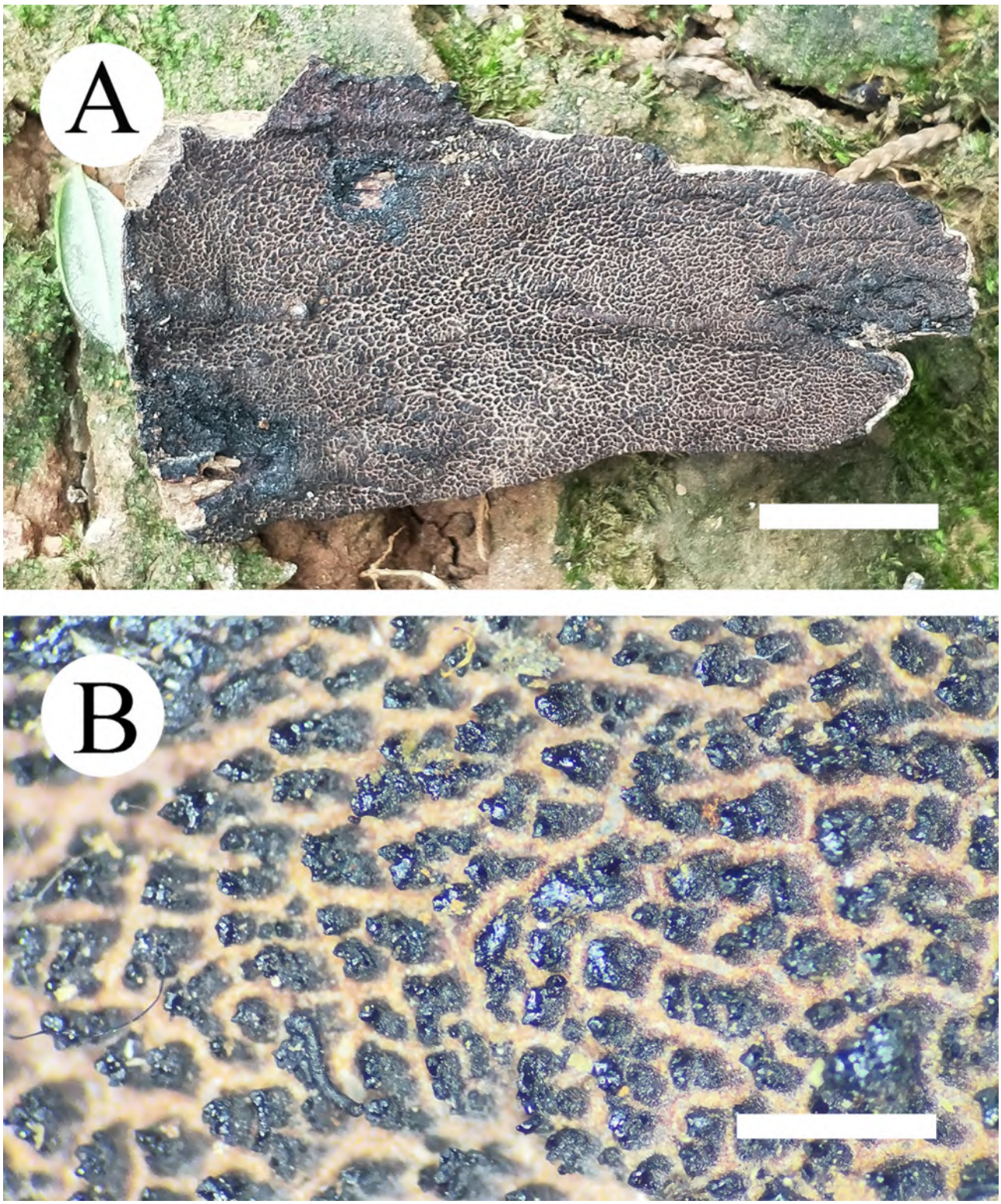

FIGURE 3. Basidiomata of *Phlebia nigrodontea.* Bars: A–1 cm; B–1 mm (holotype). Photos by: Chang-Lin Zhao

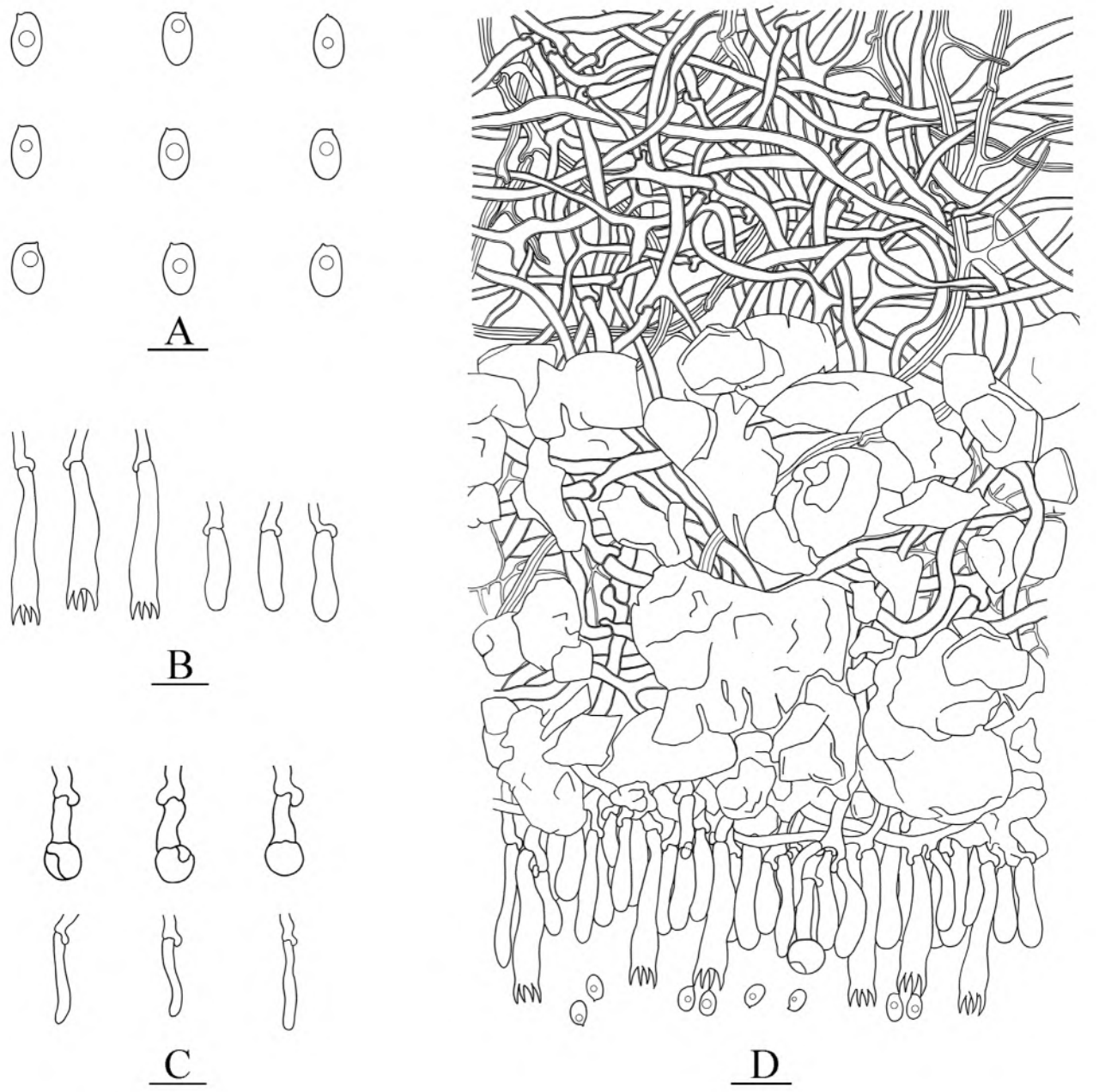

FIGURE 4. Microscopic structures of *Phlebia nigrodontea* (drawn from the holotype). A. Basidiospores. B. Basidia and basidioles. C. Cystidia and cystidioles. D. A section of hymenium. Bars: A–5 µm; B, C, D–10 µm. Drawings by: Ruo-Xia Huang

Discussion

Previously, seven clades were found in the Polyporales: the 'antrodia' clade, the 'core polyporoid' clade, the 'fragiliporia' clade, the 'gelatoporia' clade, the 'phlebioid' clade, the 'residual polyporoid' clade and the 'tyromyces' clade (Binder *et al*. 2013, Zhao *et al*. 2015). According to our result based on the combined ITS+nLSU sequence data (Fig. 1), *Phlebia nigrodontea* is nested in the phlebioid clade with strong support (100% BS, 100% BT, 1.00 BPP).

Phlebia nigrodontea appears closely related to *P. ailaoshanensis* C. L. Zhao (2018: 190), *P. chrysocreas* and *P. uda* (Fr.) Nakasone (Nakasone 1997) in the analyses of the phlebioid clade (Fig. 2). However, *Phlebia ailaoshanensis* differs from *P. nigrodontea* by having generative hyphae with simple septa and larger basidiospores (5.7–8.5 × 3–4.3 µm; Shen *et al.* 2018). *Phlebia chrysocreas* differs in its ochraceous buff to yellow ochre hymenial surface, and

cylindrical to ventricose-rostrate cystidia (Lombard *et al*. 1975). *Phlebia uda* differs from *P. nigrodontea* by having larger basidiospores (5–6 × 2.5–3 µm); in addition, the hymenial surface turns purple, red or brown in KOH (Nakasone 1997).

Morphologically, *Phlebia nigrodontea* is similar to *P. badia* (Pat.) Nakasone (2002: 478), *P. bispora* (Stalpers) Nakasone (2002: 481), and *P. hinnulea* (Bres.) Nakasone (2002: 484) in sharing thick-walled generative hyphae. However, *P. badia* differs from *P. nigrodontea* by having effused-reflexed basidiomata and simple septate generative hyphae (Nakasone 2002). *Phlebia bispora* differs in its greyish orange to brown hymenial surface and larger basidiospores (5–6.5 × 2.5–3 µm; Nakasone 2002). *Phlebia hinnulea* is separated from *P. nigrodontea* by its larger basidia (25–35 × 5–6 µm) and basidiospores (5.5–7 × 3–4 µm; Nakasone & Gilbertson 1998).

Phlebia is one of the most intensively studied genera among the Meruliaceae (Bernicchia & Gorjón 2010, Dai 2012, Binder *et al*. 2013, Justo *et al*. 2017), and has great morphological diversity and complexity. However, it is still not well understood in the subtropical and tropical regions of China and there are likely to be more species to be discovered in China and other parts of the world, particularly the Southern Hemisphere.

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References

Bernicchia, A. & Gorjón, S.P. (2010) *Fungi Europaei 12: Corticiaceae s.l.* Edizioni Candusso, Lomazzo pp. 1–1007.

Binder, M., Hibbett, D.S., Larsson, K.H., Larsson, E., Langer, E. & Langer, G. (2005) The phylogenetic distribution of resupinate forms across the major clades of mushroom-forming fungi (Homobasidiomycetes). *Systematics & Biodiversity* 3: 113–157. https://doi.org/10.1017/S1477200005001623

Binder, M., Justo, A., Riley, R., Salamov, A., López-Giráldez, F., Sjökvist, E., Copeland, A., Foster, B., Sun, H., Larsson, E., Larsson, K.H., Townsend, J., Grigoriev, I.V. & Hibbett, D.S. (2013) Phylogenetic and phylogenomic overview of the Polyporales. *Mycologia* 105: 1350–1373. https://doi.org/10.3852/13-003

Brefeld, O. (1889) Basidiomyceten III. Autobasidiomyceten und die Begründung des natürlichen Systemes der Pilze. *Untersuchungen aus dem Gesammtgebiete der Mykologie* 8: 1–305.

Dai, Y.C. (2012) Polypore diversity in China with an annotated checklist of Chinese polypores. *Mycoscience* 53: 49–80. https://doi.org/10.1007/s10267-011-0134-3

Dhingra, G.S. (1989) Genus *Phlebia* Fr. in the eastern Himalaya. *Journal of the Indian Botanical Society* 84: 111–117.

Domański, S. (1963) Dwa nowe rodzaje grzybów z grupy "Poria Pers. ex S.F. Gray". *Acta Societatis Botanicorum Poloniae* 32: 731–739. https://doi.org/10.5586/asbp.1963.044

Donk, M.A. (1933) Revisie van de Nederlandse Heterobasidiomyceteae (uitgez. Uredinales en Ustilaginales) en Homobasidiomyceteae-Aphyllophraceae: II. *Mededelingen van het botanisch Museum en Herbarium van de Rijksuniversiteit Utrecht* 9: 1–278.

Duhem, B. (2009) *Phlebia pyrenaica* sp. nov., une nouvelle espèce méditerranéenne. *Cryptogamie Mycologie* 30: 319–328.

Duhem, B. (2013) *Phlebia rhodana* sp. nov. et *Phlebia jurassica* sp. nov. (Agaricomycotina). *Cryptogamie Mycologie* 34: 291–301. https://doi.org/10.7872/crym.v34.iss4.2013.291

Duhem, B. & Michel, H. (2007) Une espèce nouvelle de *Phlebia* possédant des fibres arboriformes. *Cryptogamie Mycologie* 28: 29–38.

Felsenstein, J. (1985) Confidence intervals on phylogenetics: an approach using bootstrap. *Evolution* 39: 783–791. https://doi.org/10.1111/j.1558-5646.1985.tb00420.x

Floudas, D. & Hibbett, D.S. (2015) Revisiting the taxonomy of *Phanerochaete* (Polyporales, Basidiomycota) using a four gene dataset and extensive ITS sampling. *Fungal Biology* 119: 679–719. https://doi.org/10.1016/j.funbio.2015.04.003

Fries, E.M. (1821) Systema mycologicum. *Greifswald* 1: 1–520.

Ghobad-Nejhad, M. & Hallenberg, N. (2012) Multiple evidence for recognition of *Phlebia tuberculata*, a more widespread segregate of *Phlebia livida* (polyporales, basidiomycota). *Mycological Progress* 11: 27–35.
https://doi.org/10.1007/s11557-010-0722-1

Gilbertson, R.L. & Hemmes, D.E. (2004) New species of lignicolous basidiomycetes from Hawaii. *Memoirs of the New York Botanical Garden* 89: 81–92.

Ginns, J.H. (1969) The genus *Merulius* 2. Species of *Merulius* and *Phlebia* proposed by Lloyd. *Mycologia* 61: 357–372.
https://doi.org/10.1080/00275514.1969.12018737

Gorjón, S.P. & Greslebin, A.G. (2012) Type studies of the species of *Odontia* described by G.H. Cunningham. *New Zealand Journal of Botany* 50: 289–301.
https://doi.org/10.1080/0028825X.2012.681385

Hall, T.A. (1999) Bioedit: a user-friendly biological sequence alignment editor and analysis program for Windows 95/98/NT. *Nucleic Acids Symposium Series* 41: 95–98.

Hibbett, D.S. & Thorn, R.G. (2001) Basidiomycota: homobasidiomycetes. *In Systematics and Evolution* pp. 121–168. Springer, Berlin, Heidelberg.
https://doi.org/10.1007/978-3-662-10189-6_5

Homolka, L., Lisa, L., Eichlerova, I., Valaskova, V. & Baldrian, P. (2010) Effect of long-term preservation of basidiomycetes on perlite in liquid nitrogen on their growth, morphological, enzymatic and genetic characteristics. *Fungal Biology* 114: 0–935.
https://doi.org/10.1016/j.funbio.2010.08.009

Huang, R.X., Luo, K.Y., Ma, R.X. & Zhao, C.L. (2020) Morphological and molecular identification of a new species of *Phlebia* (Polyporales, Basidiomycota) in China. *Mycotaxon* 135: 103–117.
https://doi.org/10.5248/135.103

Jia, B.S., Zhou, L.W., Cui, B.K., Rivoire, B. & Dai, Y.C. (2014) Taxonomy and phylogeny of *Ceriporia* (Polyporales, Basidiomycota) with an emphasis of Chinese collections. *Mycological Progress* 13: 81–93.
https://doi.org/10.1007/s11557-013-0895-5

Jülich, W. (1972) Monographie der Athelieae (Corticiaceae, Basidiomycetes). *Willdenowia Beiheft* 7: 1–283.

Justo, A., Miettinen, O., Floudas, D., Ortiz-Santana, B., Sjökvist, E., Lindner, D., Nakasone, K., Niemelä, T., Larsson, K.H., Ryvarden, L. & Hibbett, D.S. (2017) A revised family-level classification of the Polyporales (Basidiomycota). *Fungal Biology* 121: 798–824.
https://doi.org/10.1016/j.funbio.2017.05.010

Kaur, G., Singh, A.P. & Dhingra, G.S. (2017) *Phlebia brevibasidia* sp. nov. from India. *Mycotaxon* 132: 95–97.
https://doi.org/10.5248/132.95

Kim, K.M., Lee, J.S. & Jung, H.S. (2007) *Fomitopsis incarnatus* sp. nov. based on generic evaluation of *Fomitopsis* and *Rhodofomes*. *Mycologia* 99: 833–841.
https://doi.org/10.1080/15572536.2007.11832515

Larsson, K.H. (2007) Re-thinking the classification of corticioid fungi. *Mycological research* 111: 1040–1063.
https://doi.org/10.1016/j.mycres.2007.08.001

Larsson, K.H., Larsson, E. & Kõljalg, U. (2004) High phylogenetic diversity among corticioid homobasidiomycetes. *Mycological Research* 108: 983–1002.
https://doi.org/10.1017/S0953756204000851

Lombard, F.F., Burdsall, H.H. & Gilbertson, R.L. (1975) Taxonomy of *Corticium chrysocreas* and *Phlebia livida*. *Mycologia* 67: 495–510.
https://doi.org/10.1080/00275514.1975.12019775

Miettinen, O. & Rajchenberg, M. (2012) *Obba* and *Sebipora*, new polypore genera related to *Cinereomyces* and *Gelatoporia* (Polyporales, Basidiomycota). *Mycological Progress* 11: 131–147.
https://doi.org/10.1007/s11557-010-0736-8

Miller, M.A., Holder, M.T., Vos, R., Midford, P.E., Liebowitz, T., Chan, L., Hoover, P. & Warnow, T. (2009) The CIPRES Portals. CIPRES. Available from: http://www.phylo.org/sub_sections/portal (accessed 4 August 2009) [Archived by WebCite(r) at http://www.webcitation.org/5imQlJeQa]

Murrill, W.A. (1905) The Polyporaceae of North America: XII. A synopsis of the white and bright-colored pileate species. *Bulletin of the Torrey Botanical Club* 32: 469–493.
https://doi.org/10.2307/2478463

Nakasone, K.K. (1997) Studies in *Phlebia*. Six species with teeth. *Sydowia* 49: 49–79.
https://doi.org/10.2307/2807699

Nakasone, K.K. (2002) *Mycoaciella*, a synonym of *Phlebia*. *Mycotaxon* 81: 477–490.

Nakasone, K.K. (2003) Type studies of resupinate hydnaceous Hymenomycetes described by Patouillard. *Cryptogamie Mycologie* 24:

131–145.
Nakasone, K.K. (2009) Type studies of corticioid Hymenomycetes (Basidiomycota) with aculei. *Sydowia* 61: 273–285.
Nakasone, K.K. & Burdsall Jr, H.H. (1984) *Merulius*, a synonym of *Phlebia*. *Mycotaxon* 21: 241–246.
Nakasone, K.K. & Burdsall Jr, H.H. (1995) *Phlebia* species from eastern and southeastern United States. *Mycotaxon* 54: 335–359.
Nakasone, K.K. & Gilbertson, R.L. (1998) Three resupinate hydnaceous basidiomycetes from Hawaii. *Folia Cryptogamica Estonica* 33: 85–92.
Nylander, J.A.A. (2004) *MrModeltest v2. Program distributed by the author.* Evolutionary Biology Centre, Uppsala University.
Parmasto, E. (1967) Corticiaceae U.R.S.S. IV. Descriptiones taxorum novarum. Combinationes novae. *Eesti NSV Teaduste Akadeemia Toimetised* 16: 377–394.
Parmasto, E. & Hallenberg, N. (2000) A taxonomic study of phlebioid fungi (Basidiomycota). *Nordic Journal of Botany* 20: 105–118. https://doi.org/10.1111/j.1756-1051.2000.tb00740.x
Petersen, J.H. (1996) Farvekort. *The Danish Mycological Society's colour-chart*. Foreningen til Svampekundskabens Fremme, Greve. 6 pp.
Persoon, C.H. (1800) *Observationes Mycologicae* 2. Germany, Leipzig; Gesnerus, Usterius & Wolfius, pp. i–xii, 1–106, 6 plates.
Roberts, P. (2000) Corticioid fungi from Korup National Park, Cameroon. *Kew Bulletin* 55: 803–842. https://doi.org/10.2307/4113628
Robledo, G.L., Amalfi, M., Castillo, G., Rajchenberg, M. & Decock, C. (2009) *Perenniporiella chaquenia* sp. nov. and further notes on *Perenniporiella* and its relationships with *Perenniporia* (Poriales, Basidiomycota). *Mycologia* 101: 657–673. https://doi.org/10.3852/08-040
Ronquist, F. & Huelsenbeck, J.P. (2003) MrBayes 3: bayesian phylogenetic inference under mixed models. *Bioinformatics* 19: 1572–1574. https://doi.org/10.1093/bioinformatics/btg180
Shen, S., Ma, X., Xu, T.M. & Zhao, C.L. (2018) *Phlebia ailaoshanensis* sp. nov. (Polyporales, Basidiomycota) evidenced by morphological characters and phylogenetic analyses. *Phytotaxa* 373: 184–196. https://doi.org/10.11646/phytotaxa.373.3.2
Singh, A.P., Dhingra, G.S. & Singla, N. (2010) A new species of *Phlebia* (Basidiomycetes) from India. *Mycotaxon* 112: 21–24. https://doi.org/10.5248/112.21
Sjökvist, E., Larsson, E., Eberhardt, U., Ryvarden, L. & Larsson, K.H. (2012) Stipitate stereoid basidiocarps have evolved multiple times. *Mycologia* 104: 1046–1055. https://doi.org/10.3852/11-174
Swofford, D.L. (2002) *PAUP*: phylogenetic analysis using parsimony (*and other methods). Version 4.0b10.* Sinauer Associates, Massachusetts.
Tomšovský, M., Menkis, A. & Vasaitis, R. (2010) Phylogenetic relationships in European *Ceriporiopsis* species inferred from nuclear and mitochondrial ribosomal DNA sequences. *Fungal Biology* 114: 350–358. https://doi.org/10.1016/j.funbio.2010.02.004
Vu, D., Groenewald, M., de Vries, M., Gehrmann, T., Stielow, B., Eberhardt, U., Al-Hatmi, A., Groenewald, J.Z., Cardinali, G., Houbraken, J., Boekhout, T., Crous, P.W., Robert, V. & Verkley, G.J.M. (2019) Large-scale generation and analysis of filamentous fungal DNA barcodes boosts coverage for kingdom fungi and reveals thresholds for fungal species and higher taxon delimitation. *Studies in Mycology* 92: 135–154. https://doi.org/10.1016/j.simyco.2018.05.001
Westphalen, M.C., Reck, M.A. & Silveira, R.M. (2010) First record of *Phlebia incarnata* from the southern hemisphere. *Mycotaxon* 114: 305–310. https://doi.org/10.5248/114.305
White, T.J., Bruns, T., Lee, S. & Taylor, J. (1990) Amplification and direct sequencing of fungal ribosomal RNA genes for phylogenetics. *In:* Innis, M.A., Gelfand, D.H., Sninsky, J.J. & White, T.J. (Eds.) *PCR protocols: a guide to methods and applications.* Academic Press, San Diego, pp. 315–322. https://doi.org/10.1016/B978-0-12-372180-8.50042-1
Zhao, C.L., Cui, B.K., Song, J. & Dai, Y.C. (2015) Fragiliporiaceae, a new family of Polyporales (Basidiomycota). *Fungal Diversity* 70: 115–126. https://doi.org/10.1007/s13225-014-0299-0
Zhao, C.L. & Wu, Z.Q. (2017) *Ceriporiopsis kunmingensis* sp. nov. (Polyporales, Basidiomycota) evidenced by morphological characters and phylogenetic analysis. *Mycological Progress* 16: 93–100. https://doi.org/10.1007/s11557-016-1259-8