Cryptogamie, Mycologie, 2014, 35 (2): 199-207
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Morphological and molecular evidence for a new species of *Postia* (Basidiomycota) from China

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Abstract – A new polypore, *Postia hirsuta* sp. nov., collected in Shaanxi Province, central China, is described and illustrated on the basis of morphological characters and molecular data. This fungus is characterized by an annual growth, pileate basidiocarps with a mouse-grey and hirsute pileal surface, a white to straw-colored pore surface, a monomitic hyphal system with thick-walled generative hyphae, and allantoid to cylindrical basidiospores (4-4.8 × 1-1.2 µm). Phylogenetic inferences based on the internal transcribed spacer (ITS) regions and nuclear large subunit (nLSU) ribosomal RNA gene regions supported *Postia hirsuta* as a distinct species in *Postia*.

Basidiomycota / brown-rot fungi / Fomitopsidaceae / molecular phylogeny / taxonomy

INTRODUCTION

Postia Fr. (Fomitopsidaceae, Basidiomycota) is a large and cosmopolitan genus. According to the current concept, it is characterized by an annual growth, a monomitic or dimitic hyphal structure with clamped generative hyphae, thin-walled, allantoid to cylindrical or ellipsoid basidiospores, and a production of a brown rot. It grows on both living or dead conifers and hardwoods (Hattori *et al.*, 2010; Cui & Li, 2012). Until now, about 60 species have been accepted in the genus worldwide (Jülich, 1982; Larsen & Lombard, 1986; Renvall, 1992; Buchanan & Ryvarden, 2000; Hattori *et al*., 2010; Dai, 2012).

The taxonomy of *Postia* in China has been dealt with in the last twenty years. Thirty species were recorded in the country of which ten were described as new (Wei & Dai, 2006; Dai *et al.*, 2009; Wei & Qin, 2010; Yuan *et al*., 2010; Cui & Li, 2012; Dai, 2012; Shen *et al*., 2014).

During a continuing survey of wood-decaying polypores in China, an additional undescribed species of *Postia* was discovered in Shaanxi Province. To confirm the affinity of the new species and to infer its relationships among similar species of *Postia*, phylogenetic analysis was performed based on the ITS and nLSU rRNA gene sequences.

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doi/10.7872/crym.v35.iss2.2014.199

MATERIAL AND METHODS

Morphological studies. – The voucher specimens of the new species are deposited at the herbarium of Beijing Forestry University (BJFC). The microscopic examinations followed Dai *et al.* (2010). Sections were studied at magnification up to × 1000 using a Nikon Eclipse 80i microscope and phase contrast illumination. Drawings were made with the aid of a drawing tube. Measurements and drawings of microscopic features, were made from slide preparations stained with Cotton Blue and Melzer's reagent. Spores were measured from sections cut from the tubes. In presenting the variation in spore size, 5% of measurements were excluded from each end of the range, and were given in parentheses. In the text the following abbreviations were used: KOH = 5% potassium hydroxide, IKI = Melzer's reagent, IKI– = negative in Melzer's reagent, CB = Cotton Blue, CB– = negative in Cotton Blue, CB+ = cyanophilous, 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 = number of spores measured from given number of specimens. Special colour terms followed Petersen (1996).

Molecular procedures and phylogenetic analysis. – The fungal taxa used in this study are listed in Table 1. A Phire Plant Direct PCR Kit (Finnzymes, Vantaa, Finland) was used to extract total genomic DNA from the dried specimens and performed the polymerase chain reaction (PCR). PCR amplification was confirmed on 1% agarose electrophoresis gels stained with ethidium bromide (Stöger *et al*., 2006). DNA sequencing was performed at Beijing Genomics Institute. All newly generated sequences have been deposited at GenBank and are listed in Table 1.

Sequences used in the phylogenetic inference (cf. Table 1) were aligned using BioEdit (Hall, 1999) and ClustalX (Thompson *et al*., 1997). Alignment was manually adjusted and deposited at TreeBase (http://purl.org/phylo/treebase; submission ID 15711). In the study, nuclear ribosomal RNA genes were used to determine the phylogenetic position of the new species. The ITS regions were amplified with the primers ITS5 and ITS4 (White *et al*., 1990; Gardes & Bruns, 1993), and nLSU with the primers LR0R and LR7 (Hopple & Vilgalys, 1999).

Phylogenetic analysis was done as described in Li & Cui (2013). Maximum Parsimony and Bayesian analysis were applied to the combined ITS and nLSU dataset. *Antrodia albida* (Fr.) Donk was used as outgroup to root trees. 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 and branches of zero length were collapsed and all parsimonious trees were saved. Clade stability was assessed using a bootstrap (BT) analysis with 1000 replicates (Felsenstein, 1985). Descriptive tree statistics tree length (TL), consistency index (CI), retention index (RI), rescaled consistency index (RC), homoplasy index (HI) were calculated for all trees generated under different optimality criteria. Bayesian analysis with MrBayes3.1.2 (Ronquist & Huelsenbeck, 2003) implementing the Markov Chain Monto Carlo (MCMC) technique and parameters predetermined with MrMODELTEST2.3 (Posada & Crandall, 1998; Nylander, 2004) was performed and the parameters in MrBayes were set as follows: lset nst = 6, and rates = gamma. Four simultaneous Markov chains were

Table 1. A list of species, specimens and GenBank accession number of sequences used in this study

Species name	*Sample no.*	*GenBank No.*	
		ITS	*nLSU*
Antrodia albida (Fr.) Donk	FP 105979	EU232189	EU232272
Postia alni Niemelä & Vampola	Cui 10094	KF699116[a]	KJ684972[a]
P. alni	Dai 10854	KF699117[a]	KJ684973[a]
P. balsamea (Peck) Jülich	JV 8609-9	JF950570	–
P. balsamea	Cui 8207	KF699118[a]	KJ684974[a]
P. carbophila Rajchenb.	MR 10758	JX090114	JX090132
P. carbophila	MR 12281	JX090115	–
P. caesia (Schrad.) P. Karst.	MR12276	JX090109	JX090129
P. caesia	MR12421	JX090110	JX090130
P. dissecta (Cooke) Rajchenb.	CIEFAP 328	JX090106	JX090134
P. dissecta	MR 12423	JX090107	JX090135
P. duplicata L.L. Shen, B.K. Cui & Y.C. Dai	Cui 10366	KF699124[a]	KJ684975[a]
P. duplicata	Dai 13411	KF699125[a]	KJ684976[a]
P. fragilis (Fr.) Jülich	JV 0610-8	JF950573	–
P. fragilis	Cui 10088	KF699120[a]	KJ684977[a]
P. guttulata (Peck ex Sacc.) Jülich	Cui 10018	KF727432[a]	KJ684978[a]
P. guttulata	Cui 10028	KF727433[a]	KJ684979[a]
P. hibernica (Berk. & Broome) Jülich	K(M) 17352	AJ006665	–
P. hibernica	Cui 8248	KF699126[a]	KJ684980[a]
P. hirsuta L.L. Shen & B.K. Cui	Cui 11237	KJ684970[a]	KJ684984[a]
P. hirsuta	Cui 11180	KJ684971[a]	KJ684985[a]
P. lactea (Fr.) P. Karst.	K(M) 31289	AJ006664	–
P. lactea	Dai 12643	KF699121[a]	KJ684981[a]
P. lateritia Renvall	KUO 020197	JF950566	–
P. lateritia	KUO 021153-1	JF950567	–
P. leucomallella (Murrill) Jülich	Cui 9599	KF699122[a]	KJ684982[a]
P. leucomallella	Cui 9577	KF699123[a]	KJ684983[a]
P. lowei (Pilát ex Pilát) Jülich	X1373	KC595941	–
P. lowei	X1417	KC595942	–
P. pelliculosa (Berk.) Rajchenb.	MR 10671	JX090101	JX090123
P. pelliculosa	MR 10592	JX090102	JX090124
P. pileata (Parmasto) Y.C. Dai & Renvall	Cui 5721	KF699127[a]	–
P. pileata	Cui 5715	KF699128[a]	–
P. placenta (Fr.) M.J. Larsen & Lombard	JV 0108/98	JN592501	–
P. placenta	Wei 1406	KF699129[a]	–
P. punctata Rajchenb. & P.K. Buchanan	MR 11100	JX090112	JX090128
P. punctata	MR 12398	JX090111	JX090127
P. rennyi (Berk. & Broome) Rajchenb.	MR 10497	JX090117	–
P. rennyi	KEW 57	AY218416	AF287876
P. sericeomollis (Romell) Jülich	MJL-3788-Sp	KC585366	KC585195
P. sericeomollis	L-15571-Sp	KC585363	KC585192
P. stiptica (Pers.) Jülich	BRFM 1151	JX082382	–
P. stiptica	Cui 9268	KF727431[a]	–
P. venata (Rajchenb. & J.E. Wright) Rajchenb.	MR 12368	JX090113	JX090133
P. zebra Y.L. Wei & W.M. Qin	Dai 7131	KF727430[a]	–

[a] Sequences newly generated in this study

ran with 5 million generations, starting from random trees, and keeping one tree every 1000 generation. Branches that received bootstrap support for maximum parsimony (MP) and Bayesian posterior probabilities (BPP) greater than or equal to 75% (MP) and 0.95 (BPP) respectively were considered as significantly supported.

RESULTS

The combined [ITS-nLSU] dataset included sequences from 45 fungal specimens representing 24 species. The dataset had an aligned length of 1879 characters, of which 1378 characters are constant, 58 are variable and parsimony-uninformative, and 443 are parsimony-informative. Maximum parsimony analysis yielded two equally parsimonious trees (TL = 1422, CI = 0.539, RI = 0.761, RC = 0.410, HI = 0.461), and one of the maximum parsimonious trees was shown in Fig. 1. Best model for ITS + nLSU estimated and applied in the Bayesian analysis: 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 = 0.005226.

The ITS + nLSU strict consensus tree (Fig. 1) generated by Bayesian analysis and maximum parsimony showed that two samples of *Postia hirsuta* were grouped together with other species of *Postia* as a monophyletic lineage with strong support (100% MP, 1.00 BPP).

TAXONOMY

Postia hirsuta L.L. Shen & B.K. Cui, **sp. nov.** **Figs 2, 3**

MycoBank: MB 808784

Original diagnosis: Differs from other *Postia* species by pileate basidiocarps with mouse-grey and hirsute pileal surface, cream to buff yellow pores, a monomitic hyphal system with thick-walled generative hyphae and allantoid to cylindrical basidiospores (4-4.8 × 1-1.2 μm).

Etymology. hirsuta (Lat.) refers to the hirsute pileal surface.

Basidiocarps annual, pileate, solitary, soft corky, watery, and without odour or taste when fresh, becoming hard corky upon drying. **Pileus** flabelliform, projecting up to 4.5 cm long, 5.5 cm wide and 2 cm thick at base. **Pileal surface** white to pale mouse-grey when fresh, becoming pale mouse-grey to mouse-grey upon drying, hirsute; margin obtuse. **Pore surface** white when fresh, cream to buff yellow upon drying; sterile **margin** narrow, up to 1 mm wide, white when fresh, becoming cream buff upon drying. **Pores** round to angular, 3-4 per mm; **dissepiments** thin, entire. **Context** white, soft corky when fresh, becoming corky upon drying, up to 1.7 cm thick. **Tubes** white and soft corky when fresh, becoming cream buff to buff yellow and hard corky upon drying, up to 3 mm long.

Hyphal system monomitic; generative hyphae with clamp connections, IKI–, CB–; tissues unchanged in KOH. **Context** of generative hyphae that are hyaline, thick-walled with a wide lumen, occasionally branched, loosely

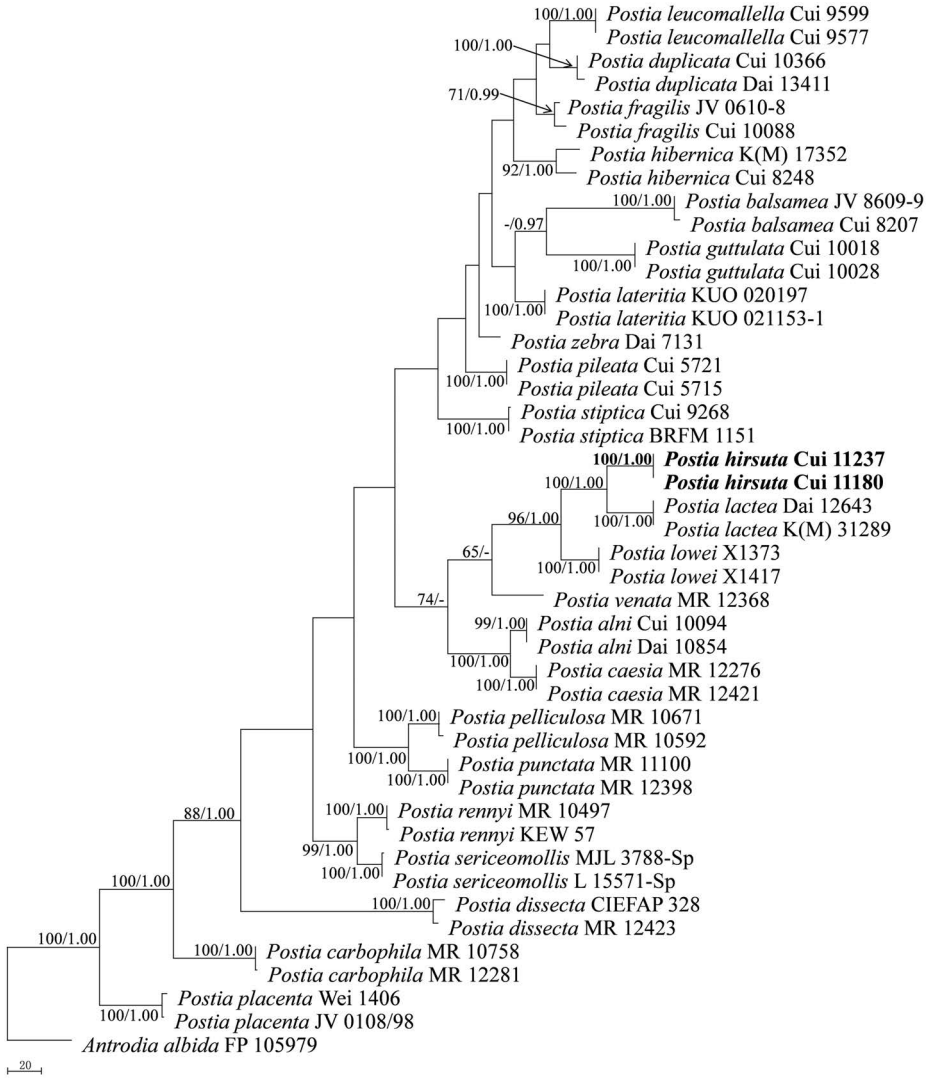

Fig. 1. Strict consensus tree illustrating the phylogeny of *Postia hirsuta* and related species generated by Maximum parsimony based on combined ITS + nLSU sequences. Parsimony bootstrap proportions (before the slash markers) higher than 50% and Bayesian posterior probabilities (after the slash markers) more than 0.95 are indicated along branches.

interwoven, 3-6 µm in diam. **Tubes**: generative hyphae hyaline, thick-walled with a wide lumen, frequently branched, interwoven, 2.5-4 µm in diam.; cystidia and cystidioles absent; basidia short clavate to barrel-shaped, with four sterigmata and a basal clamp connection, 14.5-18 × 5-7.5 µm; basidioles dominant, in shape similar to basidia or pear-shaped, but slightly smaller. **Basidiospores** allantoid to cylindrical, hyaline, thin-walled, smooth, IKI–, CB–, (3.6-)4-4.8(-5.2) × (0.8-)1-1.2 µm, L = 4.35 µm, W = 1.1 µm, Q = 4.33-4.35 (n = 60/2).

Type of rot: **Brown rot**.

Holotype: **CHINA**, Shaanxi Province, Zhashui County, Niubeiliang Forest Park, on fallen angiosperm trunk, 16 Sep 2013, B.K. Cui, Cui 11237 (BJFC holotypus).

Additional specimen examined: *Postia hirsuta* – CHINA. Shaanxi Province, Taibai Mountains, Honghegu Forest Park, on fallen angiosperm trunk, 10 Sep 2013, B.K. Cui, Cui 11180 (BJFC).

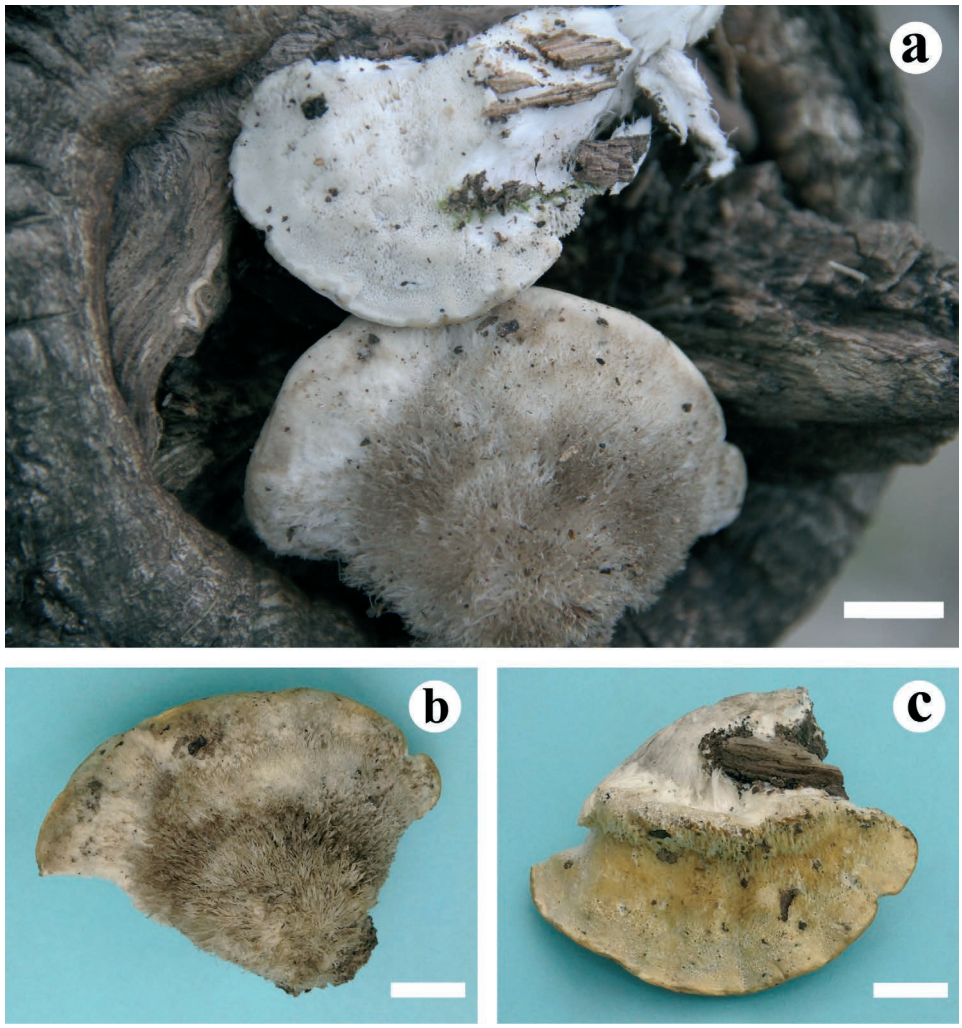

Fig. 2. Basidiocarps of *Postia hirsuta* (holotype). **a.** Fresh basidiocarps. **b, c.** Dried basidiocarp. *Bars* a, b, c = 1 cm.

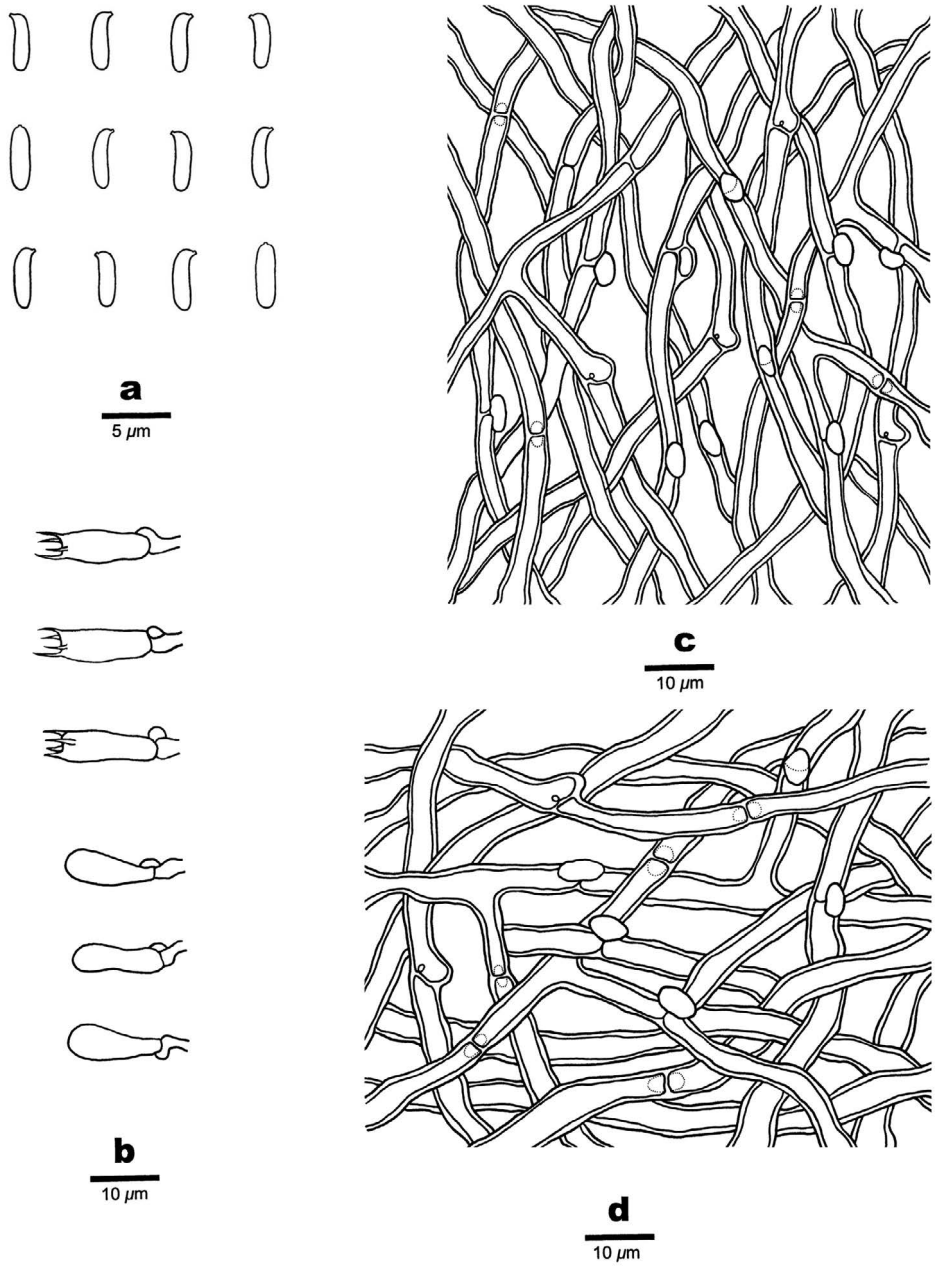

Fig. 3. Microscopic structures of *Postia hirsuta* (drawn from the holotype). **a.** Basidiospores. **b.** Basidia and basidioles. **c.** Hyphae from trama. **d.** Hyphae from subiculum. *Bars*: a 5 μm; b-d 10 μm.

DISCUSSION

In the present study, a new *Postia* species, *P. hirsuta*, is described on the basis of both morphological characters and molecular (DNA sequence) data. Phylogenetic analysis carried out from ITS and nLSU DNA sequences showed that *P. hirsuta* forms a strongly supported, distinct lineage (100% MP, 1.00 BPP) within the *Postia* lineage.

Phylogenetically, *P. hirsuta* is closely related to *P. lactea* (Fr.) P. Karst. (Fig. 1). Morphologically, *P. hirsuta* and *P. lactea* share pileate basidiocarps with white pore surface when fresh, and similar pore size. However, *P. lactea* differs from *P. hirsuta* in having a glabrous pileal surface, thin-walled contextual hyphae, and wider basidiospores (3.8-5.1 × 1.2-1.6 μm, Niemelä, 2005).

Postia amurensis Y.C. Dai & Penttilä may be mistaken for *P. hirsuta*; both species have a pale cream coloured to yellowish pore surface when dry and similar pores (3-4 per mm) and basidiospores. *Postia amurensis* differs from *P. hirsuta* in having glabrous pileal surface and distinctly thin-walled tramal hyphae (Dai & Penttilä, 2006).

Postia lowei (Pilát ex Pilát) Jülich also resembles *P. hirsuta* in having annual and pileate basidiocarps with hirsute pileal surface when juvenile and similar sized pores (3-4 per mm), but its pileal surface becoming glabrous with age, and it produces a denser darker zone within the very thin context just above the tubes and wider basidiospores (4.8-5.2 × 1.8-2.2 μm, Ryvarden & Gilbertson, 1994). Moreover, the two species are different in the ITS + nLSU rDNA-based phylogenetic analysis (Fig. 1).

Postia gloeocystidiata Y.L. Wei & Y.C. Dai resembles *P. hirsuta* in having annual and pileate basidiocarps with hirsute pileal surface and similar sized pores (3-4 per mm), but it has abundant gloeocystidia in the hymenium, and shorter basidiospores (3.7-4.5 × 1-1.1 μm, Wei & Dai, 2006).

Postia cana H.S. Yuan & Y.C. Dai also has pileate basidiocarps with hirsute pileal surface and its basidiospores are similar to *P. hirsuta*. However, *P. cana* is distinguished from *P. hirsuta* by having clay pink to fawn and indistinctly zoned, longitudinally grooved pileal surface, and its hyphal wall distinctly swollen in KOH (Yuan *et al.*, 2010).

Postia caesia (Schrad.) P. Karst. and *P. alni* Niemelä & Vampola have annual and pileate basidiocarps, similar pores (3-4 per mm) and basidiospores with *P. hirsuta*, but they are separated from *P. hirsuta* by its greyish to bluish pileal surface, and bluish pore surface (Ryvarden & Gilbertson, 1994). In addition, *P. caesia* and *P. alni* are quite different from *P. hirsuta* in the phylogenetic analysis (Fig. 1).

Acknowledgements. Special thanks are due to Drs. Shuang-Hui He and Chang-Lin Zhao (BJFC, China) for companion in field trips. We express our gratitude to Prof. Yu-Cheng Dai (BJFC, China) for improving the manuscript. The research is supported by the Fundamental Research Funds for the Central Universities (Project No. JC2013-1) and the National Natural Science Foundation of China (Project No. 31170018).

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Mise en page et impression
bialec, nancy (France)
Dépôt légal n° 82991 - 30 juin 2014

Imprimé sur papier de forêts gérées durablement