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Two new Daedalea species (Polyporales, Basidiomycota) from South China

Hai-Jiao Li, Bao-Kai Cui*

Institute of Microbiology, Beijing Forestry University, Beijing 100083, China

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ABSTRACT

Two new Daedalea species were described from South China based on morphological and molecular evidences. Daedalea circularis sp. nov. was discovered from Guangdong and Yunnan provinces, it is characterized by its bluish gray to peach, glabrous, concentrically sulcate and zonate pileal surface with irregular, white to cream outgrowth and fuscous to black patches spreading from the base, round hymenophore, colorless, thin-walled cystidioles present in the hymenium which sometimes with branched apiculus and occasionally collapsed and septate. Daedalea radiata sp. nov. was discovered from Yunnan Province, it is easily recognizable by its effused-reflexed, grayish-brown to fuscous, hispid pileal surface with angular to daedaleoid pores, tissue darkening and skeletal hyphae slightly swollen in KOH, and a typical catahymenium formed by apices of skeletal hyphae in the hymenium.

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1. Introduction

Daedalea Pers. was founded by Persoon (1801), typified by Daedalea quercina (L.) Pers. The genus is characterized by pileate, mostly perennial basidiocarps, irregular, labyrinthine/ daedaleoid to lamellate or poroid hymenophore, light brown context, trimitic hyphal system with clamped generative hyphae, a catahymenium formed by skeletal hyphae in the hymenium, colorless, smooth, thin-walled basidiospores, and mainly on hardwoods causing a brown rot (Ryvarden and Johansen 1980; Gilbertson and Ryvarden 1986; Ryvarden and Gilbertson 1993; Núñez and Ryvarden 2001; Lindner et al. 2011).

Numerous epithetic names have been placed under *Daedalea*, and 356 and 387 records of them can be found in Mycobank (http://www.mycobank.org/mycotaxo.aspx) and Index Fungorum (http://www.indexfungorum.org/Names/Names.asp, until 07 Dec, 2011), respectively. However, phylogenetic studies on *Daedalea* were meager, only a few sequence data were

available. Recently, Lindner et al. (2011) described a new species, *Daedalea neotropica* D.L. Lindner, Ryvarden & T.J. Baroni, from Maya Mountains of Belize. They also provided a synopsis of *Daedalea sensu stricto* including *Daedalea dickinsii* Yasuda, *D. neotropica*, *Daedalea pseudodochmia* (Corner) T. Hatt., and D. quercina based on morphological and DNA sequence data.

During surveys of polypores in South China, two new species, *Daedalea circularis* and *Daedalea radiata* were described based on both morphological and molecular evidences. Their illustrated descriptions are provided in this paper.

2. Materials and methods

2.1. Morphological studies

The studied specimens were deposited at the herbaria of the Institute of Microbiology, Beijing Forestry University (BJFC)

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^{*} Corresponding author. Tel.: +86 (0) 10 62336309; fax: +86 (0) 10 62336309.

E-mail address: baokaicui@yahoo.com.cn (B.-K. Cui).

and Institute of Applied Ecology, Chinese Academy of Sciences (IFP). The microscopic procedure followed Dai (2010). To present the basidiospore size variation, 5% of measurements were excluded from each end of the range, and were given in parentheses. In the text the following abbreviations were used: IKI = Melzer's reagent, IKI = both inamyloid and indextrinoid, KOH = 5% potassium hydroxide, CB = Cotton Blue, CB = acyanophilous, 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 color codes followed Petersen (1996).

2.2. Molecular study and phylogenetic analysis

Molecular techniques followed Cui et al. (2008) and Dai et al. (2010). Phire[®] Plant Direct PCR Kit (Finnzymes Oy, Finland) was used to obtain the products of polymerase chain reactions (PCR) from dried specimens, according to the manufacturer's instructions. Nuclear internal transcribed spacer (ITS) region was amplified with primer pair ITS5 (GGA AGT AAA AGT CGT AAC AAG G) and ITS4 (TCC TCC GCT TAT TGA TAT GC; White et al. 1990). The PCR procedure for ITS was: initial denaturation at 98 °C for 5 min, followed by 39 cycles at 98 °C for 5 s, 58 °C for 5 s and 72 °C for 5 s, and a final extension of 72 °C for 10 min. The PCR products were directly sequenced in Beijing Genomics Institute, China, with the same primers.

Besides sequences obtained from this study, other sequences were downloaded from GenBank (Table 1). ClustalX1.83 (Chenna et al. 2003) was used to align sequences. Sequence alignment was deposited at TreeBase (submission ID 12538). Maximum parsimony analysis was performed using PAUP* 4.0b10 (Swofford 2002) with gaps treated as missing data. Trees were generated using 100 replicates of random stepwise addition of sequence and tree-bisection reconnection (TBR) branch-swapping algorithm. All characters were given equal weight. Branch support for all parsimony analysis was estimated by performing 1000 bootstrap replicates (Felsenstein 1985) with a heuristic search of 10 randomaddition replicates for each bootstrap replicate.

3. Results

3.1. Taxonomy

Daedalea circularis B. K. Cui & Hai J. Li, sp. nov. Fig. 1. MycoBank no.: MB 564113

Carpophorum perennium, sessilium, singulum vel imbricatum. Facies pororum creamea vel bubalina; pori rotundi, 4–6 per mm. Systema hypharum trimiticum, hyphae generativae fibulatae, hyphae skeletales in contextu 2.8–4.5 μ m in diam. Sporae cylindricae, hyalinae, IKI–, CB–, 4.1–6 \times 2.1–2.7 μ m.

Type specimens: China, Yunnan Province, Mengla County, Wangtianshu Park, on fallen angiosperm trunk, 2 Nov 2009, B. K. Cui 8488 (Holotype in BJFC); Lvshilin Park, on fallen angiosperm trunk, 1 Nov 2009, B. K. Cui 8389 (BJFC, paratype); Xishuangbanna, Mangao National Nature Reserve, on fallen

Table 1 — Information of ITS sequences used in the phylogenetic analysis.

	GenBank	
Species	no.	Sample no.
Antrodia juniperina (Murrill)	DQ491416	CBS 117.40
Niemela & Ryvarden	FI 1000010	TEDI 240
(Berk & M & Curtis) Donk	EU232213	IFRI 349
A serialis (Fr.) Donk	EU162052	910310.8
A variiformis (Peck) Donk	AY966453	_
A. xantha (Fr.) Ryvarden	EU232210	TFRI 879
Antrodiella americana	EU232185	L 3468
Ryvarden & Gilb.		
A. romellii (Donk) Niemelä	AF126900	Saarenoksa 18594
A. stipitata H.S. Yuan & Y.C. Dai	FJ613651	Yuan 07
Cerrena consors (Berk.)	FJ821529	F20080405CGY02
K.S. Ko & H.S. Jung		
C. unicolor (Bull.) Murrill	FJ810175	dd08084
Coriolopsis polyzona (Pers.)	JN164979	OH272sp
Ryvarden		
Daedalea circularis	JQ314351 ^a	Cui 8488
D. circularis	JQ780411 ^a	Cui 10125
D. circularis	JQ314352 ^a	Cui 10134
D. dickinsii	EU661878	strain 027
D. dickinsii	FJ481049	xsd08139
D. dickinsii	FJ810173	dd08076
D. dickinsii	JQ314353 ^a	Cui 6825
D. dochmia	DQ491401	CBS 426.84
D. neotropica	FJ403217	DLC04-80
D. neotropica	FJ403218	DLC04-100
D. neotropica	FJ403219	DLC04-174
D. pseudodochmia	FJ403210	10533
D. quercina	FJ403213	4//99
D. quercina	FJ403214	HHB8/35
D. radiata	JQ314349 ⁻	Cui 8487
D. radiata	JQ314348	Cui 8024
D. radiata	JQ314350 EM02215	10551
D. stereoliues	IN165007	10551 PI C5624cp
D scutellata (Schwein)	IN165004	RLG30245P RI C9584T
Cilb & Bywarden)1105001	REG99011
Farliella scabrosa (Pers)	FI711056	MUCL 45097
Gilb & Ryvarden	1)/11050	WOGE 1909/
Fomitonsis caianderi	EU232200	BCRC 35447
(P. Karst.) Kotl. & Pouzar	10101100	201000011
F. incarnatus	DO491411	HSI-2006a
F. meliae (Underw.) Gilb.	DQ491421	CBS 179.34
F. ostreiformis (Berk.) T. Hatt.	HQ248222	PCO.43
F. palustris (Berk. & M.A. Curtis)	EU024965	_
Gilb. & Ryvarden		
F. pinicola (Sw.) P. Karst.	EU232197	TFRI 513
Funalia trogii (Berk.)	FJ555688	MS1
Bondartsev & Singer		
Lenzites betulina (L.) Fr.	GU067734	F3
Piptoporus betulinus (Bull.) P. Karst.	DQ491423	CBS 378.51
Pycnoporus coccineus (Fr.)	EU520116	NW580
Bondartsev & Singer		
P. sanguineus (L.) Murrill	JN164981	CR35
Trametes gibbosa (Pers.) Fr.	AY684176	CCBAS 006N
T. suaveolens (L.) Fr.	GU199349	4220
Trichaptum abietinum (Dicks.) Ryvarden	AY781273	olrim286

a Sequences generated in this study. All other sequences were derived from GenBank.



Fig. 1 – Microscopic structures of Daedalea circularis (drawn from holotype). a: Basidiospores. b: Basidia and basidioles. c: Cystidioles. d: Hyphae from trama. e: Hyphae from context.

angiosperm trunk, 11 Aug 2005, H. S. Yuan 1449 (IFP, paratype); Guangdong Province, Heyuan, Daguishan Forest Park, on angiosperm stump, 18 Aug 2011, B. K. Cui 10125, 10134 (BJFC, paratype).

ITS sequence ex holotype: JQ314351.

Etymology: circularis (Lat.): referring to the circular pores. Basidiocarps perennial, sessile, single or imbricate, without odor or taste when fresh, hard corky to woody hard and light in weight upon drying. Pilei applanate, up to 11.5 cm long, 17.5 cm wide and 3 cm thick at base. Pileal surface bluish gray to peach, irregular, white to cream outgrowth and fuscous to black patches spreading from the base with age, glabrous and smooth when young, finely tuberculate near the base when old, concentrically sulcate and zonate. Pileal margin cream to buff, distinctly paler than the pileus, obtuse. Pore surface cream to buff; sterile margin distinct, white to

65

cream, up to 3 mm wide; pores round, 4-6 per mm; dissepiments moderate to thick, entire. Context buff-yellow to honey-yellow, hard corky, zonate, up to 1 cm thick. Tube layer near pore surface white to cream, older layers buff-yellow to grayish blue, and single layer up to 3 mm long. Tissue unchanged in KOH. Hyphal system trimitic; generative hyphae bearing clamp connections; all hyphae IKI-, CB-. Generative hyphae in context infrequent, colorless, thinwalled, moderately branched, 1.4-3.3 µm in diam; skeletal hyphae in context dominant, colorless to pale yellowish brown, thick-walled, unbranched, straight, 2.8-4.5 μm in diam, interwoven; binding hyphae colorless to pale yellowish brown, thick-walled with a narrow lumen to subsolid, flexuous, frequently branched, 1.8-3.8 µm in diam. Tramal generative hyphae infrequent, colorless, thin-walled, moderately branched, 1.8–2.5 μm in diam; skeletal hyphae in trama dominant, colorless to pale yellowish brown, thick-walled, unbranched, more or less straight, interwoven, 2.4-3.5 µm in diam; binding hyphae colorless to pale yellowish brown, thick-walled with a narrow lumen to subsolid, flexuous, frequently branched, 1.4–3 µm in diam. Cystidia absent, but sometimes skeletal hyphae penetrated into the hymenium, but not from typical catahymenium; cystidioles present, fusoid to tubular, sometimes with branched apiculus and occasionally collapsed and septate, colorless, thin-walled, $13-30 \times 2.5-4 \mu m$; basidia clavate with constricted base, bearing four sterigmata and a basal clamp connection, 12–22 \times 4–6 μ m; basidioles dominant, in shape similar to basidia, but smaller. Basidiospores cylindrical, colorless, thinwalled, smooth, IKI-, CB-, $(4-)4.1-6(-7.2) \times (2-)$ $2.1-2.7(-2.8) \mu m$, L = 5.05 μm , W = 2.36 μm , Q = 2.14 (n = 30/1).

Type of rot: Brown rot.

Daedalea radiata B. K. Cui & Hai J. Li, sp. nov. Fig. 2.

MycoBank no.: MB 564114

Carpophorum annuum vel biennium, pileatum vel effusoreflexum, imbricatum. Facies pororum bubalinus vel melleus; pori angulati vel sinulis, 2–4 per mm. Systema hypharum trimiticum, hyphae generativae fibulatae, hyphae skeletales in contextu 2.2–3.8 μ m in diam. Sporae cylindricae, hyalinae, IKI–, CB–, 4.5–5 × 2.4–2.9 μ m.

Type specimens: China. Yunnan Province, Mengla County, Wangtianshu Park, on fallen angiosperm trunk, 16 Sep 2007, H. S. Yuan 3629 (Holotype in IFP, isotype in BJFC); H. S. Yuan 3580 (BJFC and IFP, paratype); 2 Nov 2009, B. K. Cui 8487, 8540, 8575 (BJFC, paratype); 3 Nov 2009, B. K. Cui 8624 (BJFC, paratype).

ITS sequence ex holotype: JQ314350.

Etymology: radiata (Lat.): referring to the radiated fibrils on the pileal surface.

Basidiocarps annual to biennial, sessile or effusedreflexed, imbricate, easily separable from the substrate, usually several pilei fused laterally, soft corky to corky upon drying; pileus semicircular or laterally elongated, convex, projecting up to 3 cm long, 6 cm wide, 1 cm thick at base. Pileal surface grayish-brown to fuscous at base and buff to cinnamon-buff toward the margin, hispid with adpressed fibrils, scales or striae, azonate, slightly sulcate or not; margin acute. Pore surface buff to cinnamon-buff when dry, shining; pores mostly angular, in parts irregular and daedaleoid, 2–4 per mm, dissepiments thin, entire. Context buff to cinnamon-buff, corky, up to 3 mm thick. Tubes concolorous with pore surface, soft corky to corky, up to 7 mm long. Tissue turns to black in KOH. Hyphal system trimitic; generative hyphae bearing clamp connections, all hyphae IKI-, CB-, skeletal hyphae slightly swollen in KOH. Generative hyphae in context colorless to pale yellowish brown, thin- to thickwalled, moderately branched, 2-3.5 µm in diam; skeletal hyphae in context dominant, colorless to pale yellowish brown, thick-walled to subsolid, rarely branched, more or less straight, 2.2–3.8 μ m in diam, interwoven; binding hyphae colorless to pale yellowish brown, thick-walled with a narrow lumen to subsolid, flexuous, moderately branched, 1.2–2.5 µm in diam. Tramal generative hyphae colorless to pale yellowish brown, thin- to thick-walled, moderately branched, 1.7-4 µm in diam; skeletal hyphae in trama dominant, colorless to pale yellowish brown, thick-walled to subsolid, rarely branched, interwoven, 2.2–4.2 µm in diam; binding hyphae colorless to pale yellowish brown, thick-walled to subsolid, flexuous, frequently branched, 1.3–2.8 μm in diam. Cystidia lacking, but skeletal hyphae penetrate into the hymenium and form a catahymenium with cystidia-like, thinning or toruloid and thick-walled apices; cystidioles present, fusoid with a long constricted base, colorless, thin-walled, $24-30 \times 3.5-4.5 \ \mu m$; basidia clavate with a long constricted base, bearing four sterigmata and a basal clamp connection, $28-42 \times 4.5-5.5 \mu m$; basidioles dominant, in shape similar to basidia, but distinctly smaller. Basidiospores cylindrical, colorless, thin-walled, smooth, IKI–, CB–, (4.3–)4.5–5(–5.1) \times (2.3–)2.4–2.9 μ m, $L=4.75~\mu m,\,W=2.66~\mu m,\,Q=1.79$ (n = 15/1).

Type of rot: Brown rot.

3.2. Molecular phylogeny

The alignment of ITS sequences included 657 base pairs with 330 parsimony-informative characters. Ten most parsimonious tree was yielded (tree length = 1484, CI = 0.477, RI = 0.722, RC = 0.344; Fig. 3). In this phylogenetic tree, D. circularis formed a well-supported clade clustered with D. quercina, D. neotropica, D. dickinsii and D. pseudodochmia which was recognized by Lindner et al. (2011) as D. sensu stricto. D. radiata, Daedalea dochmia (Berk. & Broome) T. Hatt. and Daedalea stereoides Fr. formed a moderately supported clade (bootstrap value = 75) which then grouped with the former five Daedalea species.

4. Discussion

The two *Daedalea* species were described based on morphological features and molecular data. The phylogenetic analysis (Fig. 3) inferred from ITS sequences provides strong support for the placement of *D. circularis* in *Daedalea* s. s., and *D. radiata* forms a highly supported clade grouped with *D. dochmia* and *D.* stereoides. The results also provide further confirmation that *Daedalea* is clustered with other brown-rot genera, such as *Antrodia* P. Karst., *Fomitopsis* P. Karst. and *Piptoporus* P. Karst. in the *Antrodia* clade (Binder et al. 2005), though a better phylogenetic research of this group is needed based on more samples and multi-locus.



Fig. 2 — Microscopic structures of Daedalea radiata (drawn from holotype). a: Basidiospores. b: Basidia and basidioles. c: Cystidioles. d: Section of hymenium. e: Hyphae from trama. f: Hyphae from context.

D. circularis is characterized by its bluish gray to peach pileal surface, which is glabrous, concentrically sulcate and zonate, irregular, white to cream outgrowth and fuscous to black patches spreading from the base, round pores rather than daedaleoid or lamellate hymenophore, and presence of sometimes branched and occasionally collapsed and septate cystidioles in the hymenium. Phylogenetically, the three sampled specimens of *D. circularis* formed a distinct lineage by 100% bootstrap value in the ITS MP tree, and clustered with the core *Daedalea* species by 99% bootstrap value. However,



Fig. 3 — The MP tree (species plus accession number) inferred from ITS sequences. Sequence from Trichaptum abietinum was used as outgroup. Bootstrap values more than 50% from 1000 replications are shown at nodes.

pores are partly sinuous, daedaleoid or lamellate in D. quercina and D. neotropica, and larger in D. dickinsii (1-2 per mm), which can easily distinguish from D. circularis (Hattori and Ryvarden 1994; Lindner et al. 2011). Morphologically, D. dochmia (Berk. & Broome) T. Hatt. also has pileate basidiocarps with similar pores (4-6 per mm, Hattori 2005) and more or less buff context, but its basidiospores are longer and narrower (6–7 \times 1.5–2 μm , Ryvarden and Johansen 1980) than D. circularis. Daedalea fulvirubida (Corner) T. Hatt. shares similar basidiospores (4.5–6.5 \times 2–2.8 μ m, Corner 1989; Hattori 2005), but its almost glabrous to matted, light brownto brown pileal surface, and distinctly larger and angular to daedaleoid pores (1-3 per mm, Corner 1989; Hattori 2005) can easily differentiate from D. circularis. Fomitopsis incarnatus K.M. Kim, J.S. Lee & H.S. Jung also has similar basidiospores (4.5–6.3 \times 2.2–2.9 μ m), but its brownish gray to grayish black pileal surface with broad concentric bulges which frequently fissured, small (6-8 per mm, Kim et al. 2007) pinkish white pores distinctly differs from D. circularis.

D. radiata is easy to recognize by its frequently effusedreflexed basidiocarps, grayish-brown to fuscous, hispid pileal surface with angular to daedaleoid pores. Based on ITS sequences, the three sampled specimens of D. radiata formed a highly supported lineage (bootstrap value 100%) grouped with *D. dochmia* and *D. stereoides*, but the effused-reflexed, grayish-brown to fuscous basidiocarps, hispid pileal surface of *D. radiata* is easy to differentiate from the latter two species. Macroscopically, *D. radiata* resembles *Funalia* species which have tomentose to hispid pileal surface, while the latter have regular hymenophore with euhymenium, cyanophilous skeletal hyphae and cause a white rot (Niemelä et al. 1992; Dai 1996). *D. neotropica* has similar pores (mostly poroid, in parts irregular and daedaleoid, 3–5 per mm) with *D. radiata*, but bears glabrous and finely tuberculate basidiocarps with irregular violaceous patches on the pileus and pore surfaces, and having slightly larger basidiospores (5–5.5 × 2–3 µm, Lindner et al. 2011).

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