

Four Unrecorded Wood Decay Fungi from Seoul in Korea

Yeongseon Jang¹, Sung Wook Lee¹, Seokyeon Jang¹, Young Woon Lim², Jin Sung Lee³ and Jae-Jin Kim^{1*}

¹Division of Environmental Science and Ecological Engineering, College of Life Sciences and Biotechnology, Korea University, Seoul 136-701, Korea

²School of Biological Sciences, Seoul National University, Seoul 151-747, Korea

³National Institute of Biological Resources, Environmental Research Complex, Incheon 404-708, Korea

(Received August 11, 2012. Accepted September 10, 2012)

During the investigation of Korean indigenous fungi from Seoul, three genera-*Fuscoporia*, *Porostereum*, and *Trametopsis*, and four species-*Fuscoporia senex*, *Phlebia acerina*, *Porostereum spadiceum*, and *Trametopsis cervina* were found. Their morphological characteristics were examined and their identification was confirmed by molecular analysis based on internal transcribed spacer (ITS) and nuclear large subunit ribosomal DNA region sequences. These fungi are new to Korea and registered here with descriptions.

KEYWORDS : Basidiomycete, Seoul, Taxonomy, Wood decay fungi

Introduction

Seoul is the capital of Korea. The total area is about 605 km², and around 5% of the area (31 km²) is urban forest according to the Korean statistical information service. There are numerous mountains (27% of the total area) that are covered with coniferous and deciduous trees due to the temperate climate. Thus, many studies have been carried out to investigate indigenous fungi located on mountains in Seoul. As a result, many wood-decaying fungi have been reported in this area [1-8].

To understand fungal diversity by collecting mushrooms, long-term investigation is important. Hawksworth [9] previously found that the fungal diversity of a site increased as more investigations were carried out, even after 25 years in the UK. In this sense, we collected wood-decaying fungi in different locations in Seoul in 2008~2011. In our investigation, we report four unrecorded wood-decaying fungi that are new to Korea.

Materials and Methods

Microscopic observation. Basidiocarps were collected from various places in 2008~2011 in Seoul. Macro- and microscopic features of basidiocarps were observed from the collected specimens. Slide preparations were made from dried specimens mounted in 5% (w/v) KOH, 1%

(w/v) phloxine, and Melzer's reagent (IKI) [10] using an Olympus BX51 light microscope (Olympus, Tokyo, Japan). More than 30 basidiospores were measured. Mean spore length (L) and mean spore width (W) are shown in the description of each species. 'Methuen handbook of color' [11] was used as the color standard. Voucher specimens were deposited at the National Biological Resources Center (KB).

Phylogenetic analysis. The DNAs from specimens were extracted, after which internal transcribed spacer (ITS) and nuclear large subunit ribosomal DNA (nuc-LSU) sequences were obtained using the primer sets ITS1F/ITS4, ITS1F/ITS4B, or ITS5/ITS4 for the ITS region [12, 13] and LR0R/LR3 or LR0R/LR5 for the nuc-LSU region [14], according to Jang *et al.* [15]. The sequences obtained in this study were deposited under GenBank accession Nos. JX463652~JX463663. Each sequence was compared to the reference sequences in GenBank, NCBI using a BLAST search (<http://blast.ncbi.nlm.nih.gov/Blast.cgi>). Each sequence was aligned with those obtained from GenBank using MAFFT 6.884 [16] by following the L-INS-i method and manually edited using MacClade 4.08 [17]. For analysis, each dataset from each species was treated separately. Bayesian analyses were performed with MrBayes 3.2.1 [18]. The best-fit model for each dataset was selected by MrModeltest 2.3 (AIC) [19]. Two independent

*Corresponding author <E-mail : jae-jinkim@korea.ac.kr>

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runs with 1,000,000 generations were performed, with sampling of every 100th generation. The first 25% of the trees was discarded while the last 75% of the trees was used to construct consensus trees. We also checked the graphic representation of the likelihood scores of the sampled trees. Potential scale reduction factors were close to 1.0 for all parameters. A 50% majority-rule consensus tree for each dataset was constructed, and tree reliability was assessed by posterior probability.

Results and Discussion

Taxonomy.

Fuscoporia Murrill, N. Am. Fl. (New York) 9: 3 (1907). Basidiocarp resupinate, effused-reflexed or pileate, annual to perennial; pileus usually brown, hirsute to glabrous, azonate to zonate; pore surface usually brown. Hyphal system monomitic to dimitic; hymenial setae abundant. Basidiospores hyaline, thin-walled, allantoid to cylindrical to ellipsoid. They occur on coniferous and deciduous trees.

Fuscoporia senex (Nees & Mont.) Ghobad-Nejhad, Mycotaxon 101: 208 (2007) (Fig. 1).

Basionym: *Polyporus senex* Nees & Mont., Ann. Sci. Nat. Ser. 2 5: 70 (1836).

Synonym: *Phellinus senex* (Nees & Mont.) Imazeki, Bull. Govt. For. Exp. Stn. Meguro 57: 115 (1952).

Basidiocarp: Pileate, solitary, up to 3.5×8.5 cm and 2 cm thick, broadly attached to substrate, semicircular, convex, consistency coriaceous to woody hard; pileus surface first fulvous then brown (6E8) to raw umber (5F8), finely velvety tomentose, narrow concentric sulcate zones with stiff, erect hairs, margin 1 mm thick, paler, obtuse, entire; context fibrous, glossy, dark blond (5D4) to golden brown (5D7), a darker line above the tubes, 2 mm thick; pore surface fulvous then brownish grey (6F8), pores round and small, 10~11 per mm, invisible to the naked eye, dissepiments thin and entire, tubes stratified two layers, bottom layer concolorous with the pore

surface, top layer more fulvous, layers up to 6 mm thick, margin sterile, fulvous.

Hyphal system: Dimitic; generative hyphae simple-septate, hyaline and thin-walled, $2\sim 2.5$ μ m wide, skeletal hyphae abundant, yellow to bay, thick-walled to almost solid, $2.5\sim 3.6$ μ m wide, unbranched. Hymenial setae straight, thick-walled, acuminate, $20\sim 30 \times 6\sim 7.5$ μ m, often swollen near the base.

Basidia: Only effete ones observed.

Basidiospores: Broadly ellipsoid, hyaline, thin-walled, smooth, $3.8\sim 4.2 \times 2.5\sim 3$ μ m, L = 4.04, W = 2.81.

Specimen examined: Korea, Seoul, Mt. Choansan, on the trunk of dead wood, 22 Sep 2011, Yeongseon Jang (KUC20110922-13, GenBank JX463652 for LSU and JX463658 for ITS).

Distribution: Pantropical, also in China, Taiwan, Japan, Korea, and Vietnam.

Remarks: *Fuscoporia* Murrill represents a rather heterogeneous group both morphologically and anatomically, but phylogenetic analysis of *Phellinus* s.l and *Inonotus* s.l revealed that genus *Fuscoporia* forms a monophyletic group [20]. *Fuscoporia* differs from *Phellinus* in that it occurs in both deciduous and coniferous trees (unlike only in deciduous trees in the case of *Phellinus*), hyphal system is monomitic to dimitic (unlike dimitic system of *Phellinus*), and is annual to perennial (unlike perennial basidiocarps of *Phellinus*) [20]. Phylogenetic analysis of the ITS region confirmed that our specimen of *Fuscoporia senex* is monophyletic to the *F. senex* strain CBS 442.76 with a high posterior probability value (100%), and it resides within the *Fuscoporia* clade (Fig. 2) (LSU region analysis not shown). *F. senex* is recognized based on brownish pileate basidiocarp with tomentose and narrow concentric sulcate zones in the field.

In Korea, 15 *Phellinus* species have been reported to date [21, 22]. Among them, *Phellinus contiguus* (Pers.) Pat., *P. ferruginosus* (Schr.) Pat., *P. gilvus* (Schweinitz) Pat., and *P. viticola* (Schweinitz) Donk have affinity to genus *Fuscoporia* [20] and should be treated in genus



Fig. 1. *Fuscoporia senex*. A, Basidiocarp; B, Microscopic features: a, basidiospores; b, setae; c, generative hyphae; d, skeletal hyphae.

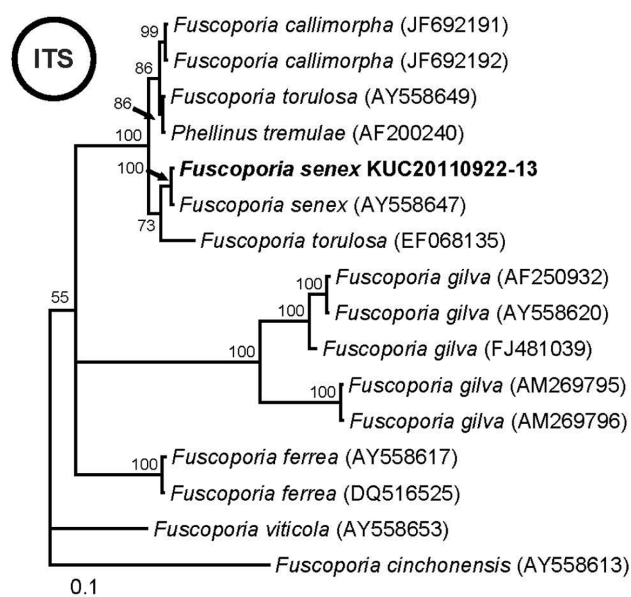


Fig. 2. Internal transcribed spacer (ITS) region bayesian analysis of *Fuscoporia senex* and its allied species. The dataset composed of 16 taxa, 599 characters. GTR + G model was applied. Posterior probability values 50% are shown on the branches. Specimen found in this study is in bold. GenBank accession numbers are in parenthesis.

Fuscoporia with the newly reported *F. senex*. Thus, there are five *Fuscoporia* species in Korea, *Fuscoporia contigua* (Pers.) G. Cunn., *F. ferruginosa* (Schr.) Murrill, *F. gilva* (Schwein.) T. Wagner & M. Fisch., *F. senex* (Nees & Mont.) Ghob.-Nehj., and *F. viticola* (Schwein.) Murrill.

Phlebia acerina Peck, Ann. Rep. N. Y. State Mus. 42: 123 (1889) (Fig. 3).

Synonyms: *Phlebia merulioides* Lloyd, Mycol. Writ. 4: 537 (1915); *Phlebia vassilkovii* Parmasto, Notul. Syst. Sect. Cryptogam. Inst. Bot. Acad. Sci. U. S. S. R. 15: 130 (1962).

Basidiocarp: Annual, resupinate, effused, confluent, waxy, hymenia continuous but frequently interrupted by white, felty subicular mycelia, central area well-developed and composed of raised folds that develop into dissepiments or teeth, single or fused, folds often anastomosing to form irregular reticulations or pits, one to two pits per mm, folds and teeth often translucent, yellowish white (4A2) when fresh, grayish orange (5A4~5A5) when dry, but fertile areas between folds often paler, then toward margins, folds becoming more appressed, slightly broader, and forming shallow pits, irregular reticulations, and warts; margins smooth, white, detached from substrate, sometimes curling away from substrate, entire, occasionally with fibrillose edges; context less than 0.1 mm thick.

Hyphal system: Monomitic; generative hyphae 3~4 µm diam. hyaline, even or irregular due to numerous knobs, thin- to thick-walled. Cystidia of two types: 1) subicular cystidia large, clavate, thin-walled, (45~) 58~88 (~125) × 6~9 µm, 4~6 µm diam at base, 2) hymenial cystidia smaller, clavate to spathulate, thin-walled, smooth, 29~44 (~50) × (4.3) 5~7 (8) µm, 1.5~2 (2.7) µm diam at base.

Basidia: 4-sterigmate, narrowly clavate, hyaline, (23~) 26~29 (~38) × (3.5~) 4~5 µm, with a basal clamp.

Basidiospores: Cylindrical to oblong ellipsoid, smooth, hyaline, (4.2~) 4.4~5.1 (~5.4) × 1.9~2.2 (2.3) µm, L = 4.56, W = 2.04, IKI-.

Specimen examined: Korea, Seoul, Mt. Guryongsan (Mt.), on the branch of dead wood, 8 Sep 2011, Yengseon Jang (KUC20110908-44, GenBank JX463653 for LSU and JX463659 for ITS).

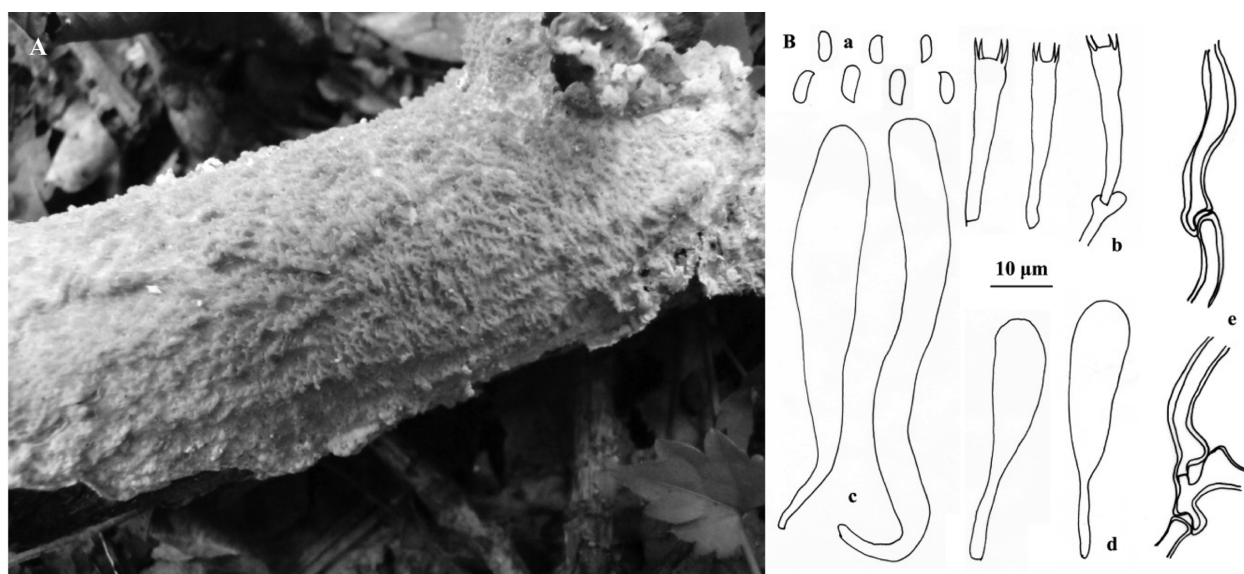


Fig. 3. *Phlebia acerina*. A, Basidiocarp; B, Microscopic features: a, basidiospores; b, basidia; c, subicular cystidia; d, hymenial cystidia; e, generative hyphae.

Distribution: Austria, Yugoslavia, Iran, Tanzania, Sri Lanka, Pakistan, Taiwan, China, Korea, USSR, Australia, New Zealand, Argentina, Canada, USA.

Remarks: *Phlebia acerina* is very similar to *P. rufa*, but *P. acerina* has yellowish brown hymenia (unlike reddish brown hymenia of *P. rufa*), often detached margins (unlike appressed margins of *P. rufa*), and hymenial cystidia (unlike

rare or absent in *P. rufa*) [23]. Phylogenetic analysis of the ITS region confirmed that our specimen is within the *P. acerina* clade with a high posterior probability value (100%) (nuc-LSU region analysis not shown) (Fig. 4).

Porostereum Pilát, Bull. Soc. Mycol. Fr. 52: 330 (1936). Basidiocarp resupinate, effuse-reflexed, upper sterile surface tomentose; hymenophore smooth to tuberculate, usually brown. Hyphal system dimitic; generative hyphae with clamps, skeletal hyphae thick-walled, brown. Cystidia brown, thick-walled, encrusted. Basidia clavate, with 4-sterigmata, with a basal clamp. Basidiospores ellipsoid, smooth, IKI-, acyanophilous.

Porostereum spadiceum (Pers.) Hjortstam & Ryvarden, Syn. Fung. (Oslo) 4: 51 (1990) (Fig. 5).

Basionym: *Thelephora spadicea* Pers., Syn. Meth. Fung. (Göttingen) 2: 568 (1801).

Synonym: *Lopharia spadicea* (Pers.) Boidin, Bull. Mens. Soc. Linn. Lyon 28: 211 (1959).

Basidiocarp: Resupinate or reflexed, grow as small patches then confluent, less than 1 mm thick, reflexed part tomentose, hymenium smooth or with radial ridges, brownish and turning grey with age; cracked when dried; margin whitish; consistency leathery, brittle when dried.

Hyphal system: Dimitic due to basal part of tramal cystidia; hyphal texture dense, hyaline to brownish, clamp connections present, thin- to moderately thick-walled, 2.7~3.9 µm wide; basal part of tramal cystidia, brown, thick-walled, 4.1~5.1 µm wide. Sterile hymenial elements present, both as hymenial cystidia and as tramal cystidia, cylindrical to clavate or fusiform, brown, thick-walled,

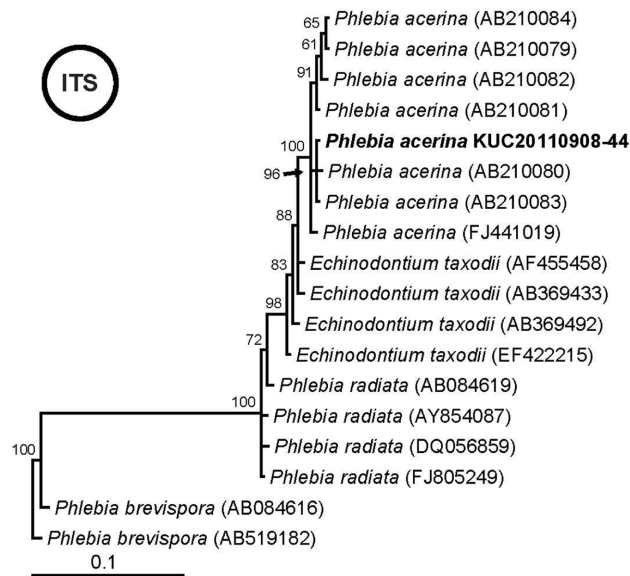


Fig. 4. Internal transcribed spacer (ITS) region bayesian analysis of *Phlebia acerina* and its allied species. The dataset composed of 18 taxa, 513 characters. GTR + I model was applied. Posterior probability values 50% are shown on the branches. Specimen found in this study is in bold. GenBank accession numbers are in parenthesis.

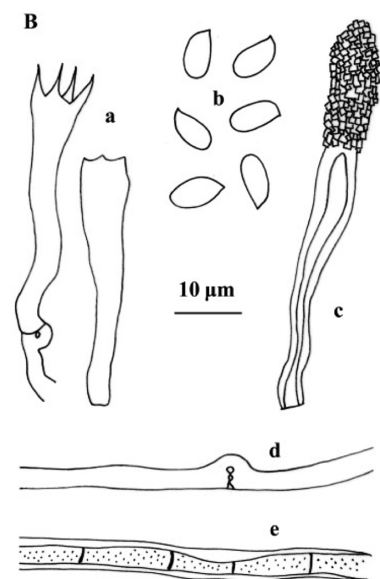
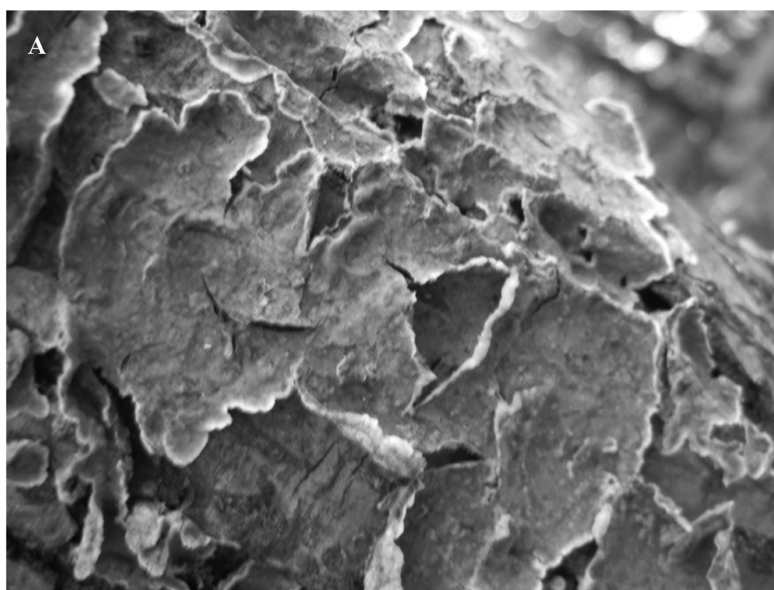


Fig. 5. *Porostereum spadiceum*. A, Basidiocarp; B, Microscopic features: a, basidia; b, basidiospores; c, cystidia; d, generative hyphae; e, basal part of tramal cystidia.

with crystalline encrustation; hymenial cystidia more or less sinuous $25\sim55 \times 3.8\sim6 \mu\text{m}$.

Basidia: Cylindrical to clavate, $30.5\sim38.5 \times 4.3\sim6.3 \mu\text{m}$, with four sterigmata and with basal clamp.

Basidiopores: Narrowly ellipsoid, smooth, thin-walled, $6.2\sim8.1 \times 3.3\sim4.2 \mu\text{m}$, $L = 7.03$, $W = 3.66$, IKI-, acyanophilous.

Specimens examined: Korea, Seoul, Mt. Gwanaksan, on deciduous wood, 28 Jul 2008, Yeongseon Jang (KUC20080728-31, GenBank JX463654 and JX463660 for ITS); Korea, Seoul, Mt. Gaeunsan, on bark of dead wood, 28 Jul 2010, Yeongseon Jang (KUC20100728-24, GenBank JX463655 for LSU and JX463661 for ITS).

Distribution: Africa, America, Asia, and Europe.

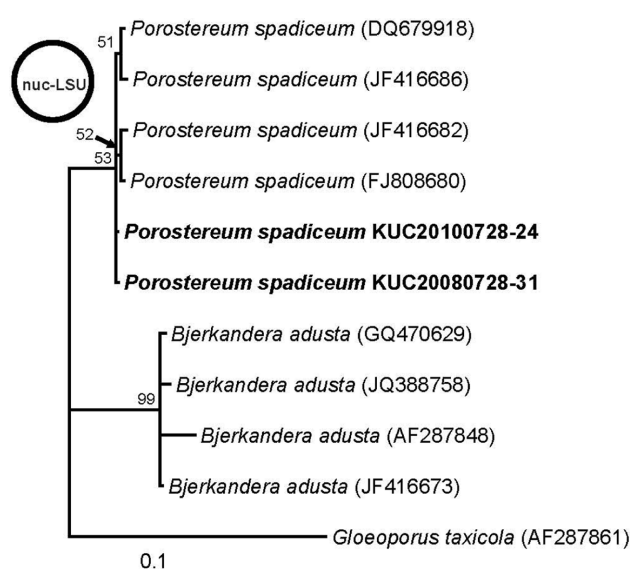


Fig. 6. Nuclear large subunit (nuc-LSU) region bayesian analysis of *Porostereum spadiceum* and its allied species. The dataset composed of 11 taxa, 558 characters. GTR + I model was applied. Posterior probability values 50% are shown on the branches. Specimens found in this study are in bold. GenBank accession numbers are in parenthesis.

Remarks: Hjortstam and Ryvarden [24] separated the genus from *Lopharia* based on colored cystidia. Binder *et al.* [25] showed that *P. spadiceum* is in the phlebioid clade, whereas *Lopharia cinerascens* and *L. mirabilis*, which is the type species of genus *Lopharia*, are in the core polyporoid clade. *P. spadiceum* is rather close to *Bjerkandera adusta*, which has no cystidia. Our specimens are monophyletic with other *Porostereum spadiceum* sequences with weak support (53% posterior probability value) in the phylogenetic analysis of the nuc-LSU region (ITS region analysis not shown) (Fig. 6).

Trametopsis Tomšovský, Czech Mycol. 60: 8 (2008).

Basidiocarps annual, sessile to effused-reflexed, usually imbricate. Pores irregular, daedaloid to irpicoid. Hyphal system dimitic; generative hyphae thin-walled with clamps, hyaline, skeletal hyphae thick-walled, hyaline, nonseptate. Basidia clavate, 4-sterigmate. Basidiospores cylindric, slightly curved, hyaline, IKI-.

Trametopsis cervina (Schwein.) Tomšovský, Czech Mycol. 60: 8 (2008) (Fig. 7).

Basionym: *Boletus cervinus* Schwein., Schr. Naturfr. Ges. Leipzig 1: 96 (1822).

Synonym: *Trametes cervina* (Schwein.) Bres., Ann. Mycol. 1: 81 (1903).

Basidiocarps: Annual, sessile, effused-reflexed, usually imbricate; upper surface hirsute to strigose, pale yellow (4A3) to brownish yellow (5C7), azonate to zonate; pore surface concolorous with the upper surface or darker brown in some dried specimens, pores irregular becoming daedaloid to irpicoid, dissepiments becoming thin and lacerate; tube layer concolorous with the context, up to 4 mm thick; context pale yellow (4A3) to light yellow (4A5), azonate, up to 1 mm thick.

Hyphal system: Dimitic; tramal generative hyphae thin-walled, with clamps, rarely branched, $1.9\sim2.8 \mu\text{m}$ in diameter; tramal skeletal hyphae thick-walled, rarely



Fig. 7. *Trametopsis cervina*. A, Basidiocarp; B, Microscopic features: a, basidiospores; b, basidia; c, generative hyphae; d, skeletal hyphae.

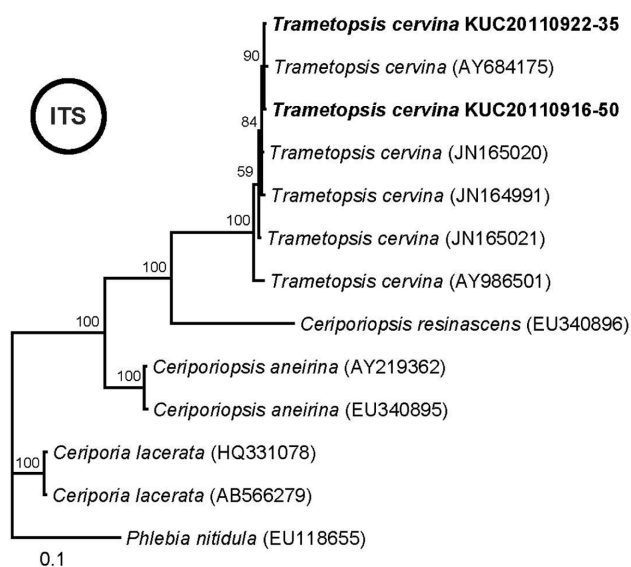


Fig. 8. Internal transcribed spacer (ITS) region bayesian analysis of *Trametopsis cervina* and its allied species. The dataset composed of 13 taxa, 566 characters. GTR + G model was applied. Posterior probability values 50% are shown on the branches. Specimens found in this study are in bold. GenBank accession numbers are in parenthesis.

branched, nonseptate, sometimes curved, flexuous, 2.5~4 μm in diameter, interwoven; contextual generative hyphae thin-walled, often sclerified, with clamps, rarely branched, 2.4~4.3 μm in diameter; contextual skeletal hyphae thick-walled, rarely branched, nonseptate, 2.8~4.5 μm in diam, interwoven. Cystidia or other sterile hymenial elements absent.

Basidia: Clavate, 4-sterigmate, 23~25 $\times$ 5~6 μm .

Basidiospores: Cylindric, slightly curved, hyaline, smooth, 4.7~7.1 $\times$ 1.9~2.7 μm , L = 5.72 μm , W = 2.13 μm , IKI-.

Specimens examined: Korea, Seoul, Heonilleung, on hardwood, 16 Sep 2011, Yeongseon Jang (KUC20110916-50, GenBank JX463656 for LSU and JX463662 for ITS); Korea, Seoul, Mt. Dobong, on dead wood, 22 Sep 2011, Yeongseon Jang (KUC20110922-35, GenBank JX463657 for LSU and JX463663 for ITS).

Distribution: Asia, Europe, and North America.

Remarks: This species was recently transferred from genus *Trametes* by Tomšovský [26]. Our specimens are monophyletic with other *Trametopsis cervina* sequences with a high posterior probability value in the ITS region consensus tree (100%) (nuc-LSU region analysis not shown) (Fig. 8). *T. cervina* is easily recognized by yellowish basidiocarp and irregular hymenophore becoming daedaloid to ipicoid.

Acknowledgements

This work was supported by the Korean indigenous

species research project from the National Institute of Biological Resources (NIBR), Ministry of Environment, Republic of Korea.

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