



RESEARCH ARTICLE



Seven Unrecorded Indigenous Fungi from Mudeungsan National Park

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ABSTRACT

Fungi act as important decomposers in the forest environment. They recycle essential nutrients, promote plant growth through mycorrhizal relationships, and act as food for small animals. Samples of 265 indigenous fungal species were collected from Mudeungsan National Park in 2020. These species were identified based on morphological, molecular, and phylogenetic analyses using the internal transcribed spacer (ITS), nuclear large subunit rRNA (LSU), and RNA polymerase II second largest subunit (rpb2) regions. Subsequently, seven species were identified as unrecorded species in Korea: Cordyceps cicadae, Dentocorticium bicolor, Hymenochaete nanospora, Physisporinus crataegi, Rigidoporus piceicola, Russula raoultii, and Scutellinia crinita. This study reveals their detailed macro- and microscopic morphological characteristics with phylogenetic trees to report them as unrecorded species in Korea.

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1. Introduction

Fungi play important roles in forest environments. They act as wood decomposers and food for small animals. They also promote or inhibit growth of plants through mycorrhizal formation or parasitism, respectively. Studies have estimated that 2.2-3.8 million fungal species live on Earth [1]. However, about 10% of the fungal species, between 220,000 and 380,000, were assumed as macrofungal species in worldwide [1,2]. Meanwhile, only about 1900 macrofungal species have been recorded in Korea until 2013 [3], and currently, it is estimated that more than 2200 macrofungal species are identified in Korea [4]. This suggests that continuous research is required to discover new and unrecorded indigenous fungal species.

Mudeungsan National Park is located in Gwangju Metropolitan City and the province of Jeollanam-do in the western part of Korea. Mudeungsan was designated as a provincial park in 1972 and redesignated as a national park in 2012 owing to its importance in preserving biodiversity and natural resources in Korea. Flora and fauna in Mudeungsan National Park have been studied extensively to determine their diversity and composition [5-8]. However, studies surveying fungal diversity have not vet been conducted.

As a project to discover indigenous fungi in Korea, fungal specimens were collected Mudeungsan National Park in 2020. The surveys were conducted in the east region of Mudeungsan National Park. Among them, 200 specimens had previously been identified using DNA molecular and morphological analyses, whereas seven were identified as unrecorded in Korea: Cordyceps cicadae, Dentocorticium bicolor, Hymenochaete nanospora, Physisporinus crataegi, Rigidoporus piceicola, Russula raoultii, and Scutellinia crinita. Five species are wood-decaying fungi, except C. cicadae, which has parasitic form, and Ru. raoultii, which grows solitary on soil covered with woody debris. This study describes detailed macroand micro-morphological characteristics of the species and provides phylogenetic trees to report them as newly recorded species in Korea.

2. Materials and methods

2.1. Sampling

Mudeungsan National Park (35°03′06″~35°12′59″ N, 126°53′41"~127°05′01" E) is located across Buk-gu of Gwangju Metropolitan City, and Damyang-gun and Hwasun-gun of Jeollanam-do Province, Korea. Collection of fungal species in Mudeungsan National Park commenced from May to September 2020. Afterwards, they were dried at 60 °C for 72 h and stored in silica gel. The specimens were deposited in the Korea University Collection (KUC) and National Institute of Biological Resources (NIBR).

2.2. Molecular approach

Genomic DNA of the dried specimens was extracted using AccuPrep® Genomic DNA Extraction Kit (Bioneer, Daejeon, Korea). The internal transcribed spacer (ITS) region was amplified with ITS5/ITS4 or ITS1F/ITS4 primer sets [9,10]. The nuclear large subunit rRNA (LSU) region was amplified with the primer sets, LR0R/LR5 or LR0R/LR7 [11,12]. For Rigidoporus piceicola, bRPB2-6F/bRPB2-7.1R primer set was used to amplify RNA polymerase II second largest subunit (rpb2) region [13]. The PCR prodpurified using AccuPrep® Purification Kit and AccuPrep® Gel Purification Kit (Bioneer) according to the manufacturer's instrucsequencing was performed DNA Cosmogenetech (Seoul, Korea), and each sequencing result was edited using SeqMan Lasergene package version 7.0.0 (DNAStar Inc., Madison, WI). Reference sequences were collected from the NCBI GenBank database (www.ncbi.nlm.nih.gov/genbank/), and the edited sequences were compared with the reference sequences using BLAST. Each sequence was assembled and aligned using MEGA version 7 and MAFFT version 7.130 [14,15]. Phylogenetic analyses were performed using the maximum likelihood (ML) method, using RAxML with GTR+G model, and 1000 bootstrap replicates were used for tree inference [16]. All analyses were performed using CIPRES [17]. The obtained trees were edited using FigTree version 1.4.3. [18] and Adobe Illustrator CS6 (Adobe Systems, Inc., San Jose, CA). Bootstrap support values above 70% are shown in the tree. All newly generated sequences of the seven species have been deposited in GenBank (Table 1).

2.3. Morphological observation

For morphological characteristics, microscopic features were observed using Olympus BX51 light microscope (Olympus, Tokyo, Japan), and pictures were taken using DP20 microscope camera (Olympus). The observations were performed using 5% KOH and Congo red solutions. The special color terms follow the Munsell Soil Color Book [19]. The following abbreviations are used: L = mean spore length, W = mean spore width, and Q = L/W ratio.

3. Results and discussion

Seven unrecorded species were identified using phylogenetic analyses of DNA sequences, particularly the ITS or LSU regions (Table 1). Six species were enough to be identified using ITS or LSU regions. However, Ri. piceicola needed multigene analysis (ITS, LSU, and rpb2) [20]. At the molecular level, each of the seven species was compared with its reference sequence and clearly identified with a high percentage of homology (Table 1). The identification results revealed the orders of the species: Polyporales (two spp.), Hymenochaetales (two spp.), Hypocreales (one sp.), Pezizales (one sp.), and Russulales (one sp.) (Table 1).

Seven species were clearly distinguished through ML method using ITS or LSU or rpb2 sequence datasets for phylogenetic analyses. According to the phylogenetic trees (Figures S1-S7, Supporting Information), C. cicadae formed monophyletic groups with high bootstrap values and its sister group was C. tenuipes (Figure S1). D. bicolor was clearly separated from other taxa with high bootstrap values (Figure S2). H. nanospora formed monophyletic groups with high bootstrap values and its sister group was H. cana (Figure S3). P. crataegi formed monophyletic groups with high bootstrap values and its sister group was P. vitreus (Figure S4). Ri. piceicola was not clearly separated from Ri.

Table 1. Molecular identification of seven unrecorded species in Korea.

Identity	Specimen ID	Order	Gen. Acc. No. ^f				Homology	Accordance
			ITS	LSU	rpb2	Ref ^g	(%)	of bp
Cordyceps cicadae	KUC20200924-44	Hypª	OK559813	_	_	KX017277	99	536/537 bp
Dentocorticium bicolor	KUC20200527-18	Pol ^b	OK559816	_	_	MF626355	100	394/394 bp
	KUC20200617-44		OK559817	_	_	MF626355	99	547/549 bp
Hymenochaete nanospora	KUC20200924-48	Hym ^c	OK559811	_	_	NR_160200	99	701/710 bp
Physisporinus crataegi	KUC20200903-15	Pol	OK559818	_	_	KY131845	99	544/547 bp
Rigidoporus piceicola	KUC20200924-53	Hym	OK559812	_	_	KT203301	99	528/535 bp
			_	ON171629	_	KT203322	98	873/889 bp
			_	_	ON204049	KT210376	99	595/598 bp
Russula raoultii	KUC20200616-18	Rus ^d	_	OK576271	_	KT933863	99	773/781 bp
Scutellinia crinita	KUC20200923-07	Pez ^e	OK559814	_	_	MN752431	99	540/543 bp

^aHyp: Hypocreales; ^bPol: Polyporales; ^cHym: Hymenochaetales; ^dRus: Russulales; ^ePez: Pezizales; ^fGen. Acc. Num.: GenBank Accession Number; gRef: reference.

obducens, Ri. populinus, and Ri. subpopulinus using ITS or LSU regions, so combined tree of ITS, LSU, and rpb2 regions is provided for accurate identification [20]. As a result, Ri. piceicola formed monophyletic groups with high bootstrap values (Figure S5). Ru. raoultii was clearly separated from the other taxa with high bootstrap values (Figure S6). S. crinita was clearly separated from the other taxa, but the clade was divided within the S. crinita group (Figure S7). Therefore, further research on the genus Scutellinia is required to determine why the clade was divided.

The genus Cordyceps Fr. is characterized by fusiform to clavate fertile heads of the stroma and filiform ascospores [21]. Additionally, this genus is widely used for medicinal applications, especially in Asia. Although 183 species are accepted as Cordyceps worldwide [22], only 46 species are listed in the NIBR database in Korea. Occurrence of Cordyceps is affected by conditions, such as humidity, temperature, elevation, and life cycle of its host species [21]. The host species are mostly insects, but other Cordyceps species, fungi Elaphomyces, spiders, nematodes, and plant tissues have also been reported as the hosts [23,24]. Many synonyms have been listed because of their sexual and asexual morphs [21]. Therefore, additional studies are required to correct sexual and asexual names and to reclassify hidden Cordyceps species.

The genus Dentocorticium (Parmasto) M.J. Larsen & Gilb was separated from the extinct genus Laeticorticium [25]. Currently, six species are listed in the genus, and five previously listed species have been reclassified as other genera [26-29].Dendrodontia and Fuscocerrena have been identified as synonyms of *Dentocorticium* based on phylogenetic analysis [30]. These two genera have macroscopic features similar to Dentocorticium [30]; therefore, accurate observations of microscopic characteristics and molecular analyses are required to identify them correctly. Dentocorticium is characterized by effused basidiocarps, brown skeletal hyphae, and irregularly branched dendrohyphidia [30].

The genus Hymenochaete Lév is characterized by brown effused basidiocarps, hymenial setae, and small basidiospores [31,32]. More than 120 species have been reported globally [33], but only 12 species were reported in Korea. Phylogenetic analysis revealed that in addition to Hymenochaete, H. tabacina (Sowerby) Lév is closely related to the poroid genera [33,34]. Consequently, the new genus Pseudochaete was separated [33,34]. Therefore, molecular approaches are required because of the similar morphological characteristics of these two genera [33].

The genus Physisporinus P. Karst. has eight species worldwide and is new to Korea. Physisporinus species are characterized by soft basidiocarps, and lack of cystidia [20,35-37]. Phylogenetic analysis showed that Physisporinus and several species of Rigidoporus Murrill were closely related in the Polyporales clade [34,38-40]. However, it is difficult to distinguish between the two genera because of their similar morphological characteristics [20]. Therefore, accurate phylogenetic analysis is required to correctly identify the species in these genera [20].

The genus Rigidoporus Murrill has morphological characteristics similar to genera Leucophellinus Bondartsev & Singer, Oxyporus (Bourdot & Galzin) Donk, and Physisporinus P. Karst [35,37,41]. Rigidoporus is distinguished by ochraceous and corky basidiocarps, and hyphoid cystidia [20]. According to phylogenetic analyses, the four genera are closely related [34]. Several species of Oxyporus and Rigidoporus were classified in the order Hymenochaetales, but species of Physisporinus and some species of Rigidoporus were classified in the order Polyporales [34,38,39]. Therefore, additional research is required for accurate classification.

The genus Russula Pers. is the second largest genus in the class Agaricomycetes, with has 1331 recorded species; it is estimated that almost 3000 species exist globally [42,43]. However, only 82 species of this genus were reported in Korea. In some studies, Russula was studied using the LSU region, and it was proved that the LSU region is an effective tool for identifying this species [44,45]. Russula is characterized by a large, bright-colored pileus and spores with echinulate surfaces [46,47]. The species in this genus are difficult to distinguish through morphological characteristics because of their large population [48,49]. Since an accurate description of taxonomy is difficult, molecular and phylogenetic analyses are necessary [47].

The genus Scutellinia (Cooke) Lambotte is a cosmopolitan fungi which have been reported worldwide, usually in the Northern Hemisphere [50]. This genus is characterized by orange to red apothecia and blackish brown hairs around the margin of apothecia [51]. Although over 100 species are listed in the genus, only four species were reported in Korea [22]. Among the species, 10 species that occur in soil have globose to subglobose ascospores, and the others that occur on decayed wood have ellipsoid ascospores [50-54]. Phylogenetic analysis showed that the genus Scutellinia is well divided in the family Pyronemataceae [51].

This study identified several unrecorded species in Mudeungsan National Park. These records are important resources for determining the diversity and composition of fungal species in Korea.

The survey started in 2020; therefore, it is expected that many new and unrecorded species are yet to be discovered. Thus, additional surveys are required at different locations, altitudes, and seasons.

4. Taxonomy

4.1. Cordyceps cicadae (miq.) Massee, annals of botany 9: 38 (1895) [MB#311793]

4.1.1. Asexual morphs

Synnemata erect, irregularly branched, clavate, with powdery and floccose apex due to the conidial mass, white (2.5Y, 9.5/1) to pale yellow (2.5Y, 8.5/2) mycelium, 30–50 mm in length and 0.5–2.5 mm in diam (Figures 1(A) and 2(A)). **Conidiophores** and Phialides were not observed. **Conidia** narrowly cylindrical with round apex, sometimes slightly curved, smooth, thin-walled, hyaline, containing one or two guttules, $6.0-7.5 \times 2.0-3.5 \, \mu m \, [Q=2.14-3.25, Q=2.7\pm0.5] \, (n=40).$

4.1.2. Specimen examined

Korea. Gwangju Metropolitan City, Mudeungsan National Park, 35°07′51″ N, 126°59′62″ E, mixed hardwood forest, parasitic in cicada, 24 Sep 2020, S. L. Kwon, KUC20200924-44 (NIBRFG0000511371).

4.1.3. Remarks

Cordyceps cicadae KUC20200924-44 matched well with C. cicadae (Miq.) Massee in the phylogenetic analysis. The size of the conidia was measured and found to be similar to the original description [55]. Further microscopic observations are required for accurate identification. According to phylogenetic analysis, C. qingchengensis is the most closely related species. The difference between C. cicadae and C. qingchengensis is the host species. The hosts of C. cicadae are cicada nymphs, while the host of C. qingchengensis has been identified to be the coconed pupa of a large silk moth [21]. However, the asexual morph of C. qingchengensis is unknown; therefore, it was impossible to compare its morphological characteristics.

4.2. Dentocorticium bicolor (P.H.B. Talbot) Nakasone & S.H. He, MycoKeys 32: 42 (2018) [MB#823073]

Basidiocarp annual, resupinate, loosely adnate, effused, little tough, easily separated from substrate when dried, up to 200–300 µm thick (Figures 1(B) and 2(B)). **Hymenial surface** membranaceous, slightly odontoid, hydonoid, tuberculate because of hyphal pegs, margin abrupt, very pale yellow (2.5Y, 9.5/2) to

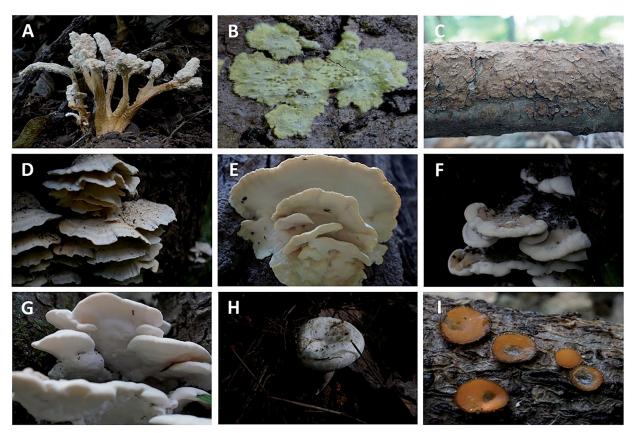


Figure 1. Fruiting bodies of the seven unrecorded species in Korea. (A) *Cordyceps cicadae*; (B) *Dentocorticium bicolor*; (C) *Hymenochaete nanospora*; (D and E) *Physisporinus crataegi*; (F and G) *Rigidoporus piceicola*; (H) *Russula raoultii*; (I) *Scutellinia crinita*.

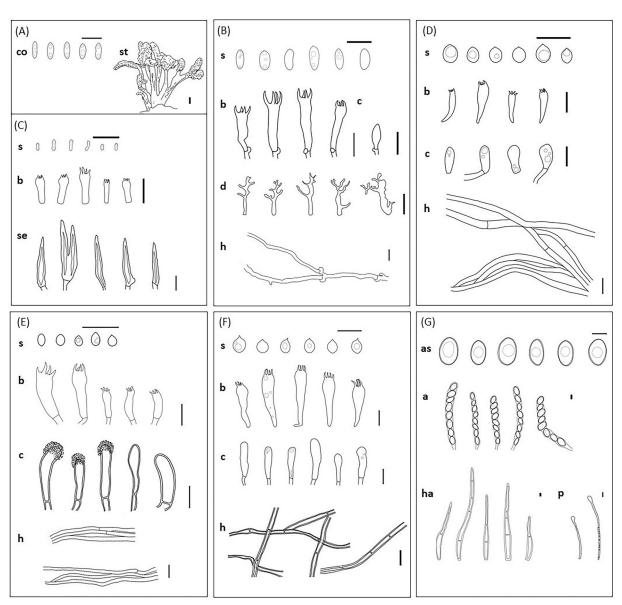


Figure 2. Microscopic view of morphological characteristics of the seven unrecorded species in Korea. (A) Cordyceps cicadae; (B) Dentocorticium bicolor; (C) Hymenochaete nanospora; (D) Physisporinus crataegi; (E) Rigidoporus piceicola; (F) Russula raoultii; (G) Scutellinia crinita. (a: asci, as: ascospores, b: basidia, c: cystidia, co: conidia, d: dendrohyphidia, h: hyphae, ha: hair, p: paraphyses, s: basidiospores, se: setae, st: stroma; a scale bar = $10 \,\mu m$, for stroma, a scale bar = $1 \,cm$).

yellow (5Y, 8/8) when fresh, pale yellow (5Y, 8/2) to yellow (5Y, 8/6) when dry. Hyphal system dimitic, generative hyphae located in subhymenium, moderately septate with clamp connections, branched, thinwalled, hyaline, 2.0-3.5 µm in diam. Skeletal hyphae easily separated from generative hyphae, long, straight, aseptate with clamp connections, unbranched, thickwalled up to 1 µm with thin-walled round apex, brownish yellow (10YR, 6/6), 2.7-4.1 µm in diam. Dendrohyphidia arising from subhymenial hyphae, irregularly and strongly branched, tortuous and with bulges, thin-walled, hyaline, 1.0–2.5 μm in diam. Basidia clavate, 4-spored, smooth, hyaline, containing one or two guttules (17–)18.5–21(–21.5) \times 4.0–5.0 μm (n = 31). **Basidiospores** ellipsoid to cylindrical, smooth, thin-walled, hyaline, containing over two guttules, $7.5-8.5 \times 3.0-4.5 \,\mu\text{m}$ [Q = 1.9-2.5, Q = 2.2 ± 0.3] (n = 34). **Cystidia** lacks, cylindrical to subclavate, smooth, thin-walled, hyaline, $17-27 \times 3.7-6.2 \,\mu\text{m}$ (n = 8).

4.2.1. Specimen examined

Korea. Gwangju Metropolitan City, Mudeungsan National Park, 35°08′80″ N, 126°58′59″ E, mixed hardwood forest, occurring on dead trees, 27 May 2020, S. Kwon, KUC20200527-18 (NIBRFG0000507813), 17 June 2020, S. L. Kwon, KUC20200617-44 (NIBRFG0000508874).

4.2.2. Remarks

Dentocorticium bicolor KUC20200527-18 and KUC20200617-44 matched well with D. bicolor (P.H.B. Talbot) Hjortstam & Ryvarden in the phyloanalysis, with high bootstrap

The morphological characteristics of D. bicolor KUC20200527-18 and KUC20200617-44 are similar to the original description. According to the description of D. bicolor (Talbot) Hjortstam & Ryvarden, the size of the basidia was determined to $30-35 \times 6-7 \,\mu\text{m}$. However, according Grandinia bicolor (P.H.B. Talbot), a synonym of D. bicolor, the basidia size is smaller than that of D. bicolor, KUC20200527-18 and KUC20200617-44. In addition, the context color was reported as brown; however, brown-colored samples were not observed. Further studies are required to obtain more accurate results. Phylogenetic analysis shows that D. sulphurellum is the most closely related species. D. sulphurellum has white to yellow hymenial surface and basidiospores $(7-9.5 \times 2.5-3 \,\mu\text{m})$, similar to *D*. bicolor. However, D. sulphurellum has only been reported in North America [30].

4.3. Hymenochaete nanospora J.C. Léger, cryptogamie mycologie 4 (3): 235 (1983) [MB#108064]

Basidiocarps annual, resupinate, effused, adherent, margin thin, woody hard when dry, cortex present (Figures 1(C) and 2(C)). Hymenial surface crustaceous, smooth, densely cracked, light brown (7.5YR, 6/4) when fresh, brown (7.5YR, 4/4) when dry. Hyphal system monomitic, usually septate without clamp connections, occasionally branched, thin-walled, hyaline, 2.1–3.3 µm in diam. Setae obclavate, fusiform, with acute apex, partition absent, smooth, thick-walled, reddish brown (7.5YR, 6/8), $20-45 \times 4.0-5.0 \,\mu\text{m}$ (n = 30). **Basidia** clavate, subcylindrical, 4-spored, smooth, hyaline (8.5-)9.0- $11.5(-12.5) \times 2.0-3.5 \,\mu\text{m}$ (*n* = 48). **Basidiospores** narrowly cylindrical to allantoid, smooth, thinwalled, hyaline, containing two guttules, $2.5-3.5 \times$ $0.8-1.2 \,\mu\text{m} \, [Q=2.92-3.125, \, Q=3.02\pm0.1] \, (n=23).$ Cystidia absent.

4.3.1. Specimen examined

Korea. Gwangju Metropolitan City, Mudeungsan National Park, 35°07′52" N, 126°59′62" E, mixed hardwood forest, occurring on hardwood branches, 24 Sep 2020, S. L. Kwon, KUC20200924-48 (NIBR FG0000511372).

4.3.2. Remarks

Hymenochaete nanospora KUC20200924-48 was well-matched with H. nanospora J.C. Léger in the phylogenetic analysis. This species is morphologically characterized by small and narrow spores, and its width rarely exceeds 1 µm. According to phylogenetic analysis, H. cana was identified as the most closely related species. H. cana has reddish brown setae $(20-40 \times 5.0-8.0 \,\mu\text{m})$ like *H. nanospora*. However, H. cana differs from H. nanospora in that it has mouse-gray to ash-gray basidiocarps, without a cortex, and larger basidiospores (2.8-)3-3.4 $(-3.6) \times (1.5-)1.6-1.9(-2) \mu m$ [56].

4.4. Physisporinus crataegi F. Wu, jia J. Chen & Y.C. Dai, mycologia 109 (5): 751 (2017) [MB#819191]

Basidiocarps annual, effused-reflexed, pileate, mostly imbricate, soft when fresh, fragile when dry (Figures 1(D,E) and 2(D)). Pileus elongnated, up to 4cm long, 8 cm wide, and 4.5 cm thick at the base. Pileus surface zonate, margin crenate, incurved when dry, pale orange yellow (10YR, 9.5/2) to very pale brown (10YR, 8.5/2). Pores angular to round, 6-7 per mm, dissepiments thin, lacerate. Pore surface white (2.5Y, 8/1) to yellow (2.5Y, 8/6) when fresh, becoming dark yellowish brown (18YR, 4/6) when dry. Context corky, azonate, 1.2 mm thick. Tubes concolorous with surface of the pores, tube layers distinct, each layer up to 5 mm long. Hyphal system monomitic, hyphae usually septate and occasionally with clamp connections, sometimes branched, thin-walled, hyaline, 3.5-5.5 µm in diam. Basidia broadly clavate, 4spored, smooth, hyaline $(8.0-)10.5-13.5 \times 4.5-5.5 \,\mu m$ (n = 17). **Basidiospores** ellipsoid, smooth, thinwalled, hyaline, sometimes containing a guttle, $4.5-5.0(-5.5) \times 3.0-4.0 \,\mu\text{m}$ [Q = 1.25-1.5, Q = 1.38 ± 0.13] (n = 44). Hymenial cystidia absent. Cystidioles present, clavate, ovoid, fusoid, smooth, thin-walled, hyaline, $10-14.5 \times 4.5-6.5 \,\mu\text{m}$ (n = 39).

4.4.1. Specimen examined

Korea. Gwangju Metropolitan City, Mudeungsan National Park, 35°08′59" N, 126°58′97" E, mixed hardwood forest, clustered or mesic in the lower part of a broad-leaved tree stump, 3 Sep 2020, S. L. Kwon, KUC20200903-15 (NIBRFG0000511374).

4.4.2. Remarks

Physisporinus crataegi KUC20200903-15 is matched well with P. crataegi F. Wu, Jia. J. Chen & Y.C. Dai, Wu, Chen, Ji, Vlasak & Dai in the phylogenetic analysis. Besides the presence of various cystidioles observed on the new species, microscopic characteristics of P. crataegi KUC20200903-15 match the original description [20]. P. crataegi is characterized by effused-reflexed and soft basidiocarps, cystidia absent, and ellipsoid basidiospores, and grows on broad-leaved trees in temperate forests. The resupinate region of P. crataegi is similar to that of P. vitreus in having soft basidiocarps, but P. vitreus has larger basidiospores $(5-6 \times 4-5 \,\mu\text{m})$ and pores (4-6)per mm) [20]. Phylogenetically, P. cinereus was identified as the most closely related species (Núñez & Ryvarden) F. Wu et al. However, P. cinereus has fibrillose and grayish pileus, large pores (5-6 per mm), and large globose-shaped basidiospores, 5-6 µm in diam [57], whereas P. crataegi has glabrous and cream-colored pileus with distinct pellicle, smaller pores (6-8 per mm), and broadly ellipsoid to subglobose basidiospores $(4.5-5.0(-5.5) \times 3.0-$ 4.0 μm). In addition, the basidiocarp of *P. crataegi* is fragile when dry, whereas that of P. cinereus is woody hard [57].

4.5. Rigidoporus piceicola (B.K. Cui & Y.C. Dai) F. Wu, jia J. Chen & Y.C. Dai, mycologia 109 (5): 761 (2017) [MB#819208]

Basidiocarps annual, effused-reflexed, pileate, mostly imbricate, soft when fresh, becoming woody hard when dry (Figures 1(F,G) and 2(E)). Pileus broadly attached, up to 5 cm wide, 4 cm long, and 3.5 cm thick at the base. Pileus surface very pale brown (10YR, 8/3) to reddish yellow (7.5YR, 8/6) when fresh, very pale yellow (2.5Y, 9.5/2) to margin yellow (2.5Y, 7/8) when dry, azonate, margin sinuate. Pores angular to round, 4-5 per mm, dissepiments thin, entire. Pore surface white (2.5Y, 8/1) to yellow (2.5Y, 8/6). Context pale brown (2.5Y, 8/2), corky, azonate, up to 5 mm thick. Tubes concolorous with pore surface, each layer up to 1.5 mm long, tube layers distinct. Hyphal system monomitic, hyphae usually septate and without clamp connections, occasionally branched, thin-walled, with round apex, hyaline, 2.5-4.0 µm in diam. Basidia clavate, 4-spored, smooth, hyaline, $11.5-13 \times 3.5-4.0 \,\mu m \, (n=30)$. Basidiospores ellipsoid, smooth, thin-walled, hyaline, sometimes containing a guttule $(2.5-)3.0-4.5 \times 2.0-3.0(-3.5)$ µm [Q = (1.25-)1.3-1.5, $Q = 1.4 \pm 0.15$ (n = 38). Cystidia arising from the trama, clavate, apically encrusted with coarse crystals, thick-walled, hyaline $(19-)20.5 - 25.5(-28) \times 4.0-5.0 \,\mu\text{m} \,(n=42).$

4.5.1. Specimen examined

Korea. Gwangju Metropolitan City, Mudeungsan National Park, 35°07′52" N, 126°59′62" E, mixed hardwood forest, clustered or mesic in the lower part of a broad-leaved tree stump, 24 Sep 2020, S. L. Kwon, KUC20200924-53 (NIBRFG0000511373).

4.5.2. Remarks

Rigidoporus piceicola is characterized by thick-walled and apically encrusted cystidia with coarse crystals, and ellipsoid thin-walled basidiospores. The microscopic characteristics of Ri. piceicola KUC20200924-53 are similar to Ri. piceicola (B.K. Cui & Y.C. Dai) F. Wu, Jia J. Chen & Y.C. Dai (basionym: Oxyporus piceicola) [20,58]. However, the basidiospore size of KUC20200924-53 is smaller than previously reported [58]. Although Ri. piceicola is parasitic only on gymnosperm trees mostly on the genus Picea in the original description, it was found on a broad-leaved tree stump in this study. Based on phylogenetic analysis, Rigidoporus populinus (Fr.) Donk is the most closely related species. Ri. populinus has clavate basidia and apically encrusted cystidia, like Ri. piceicola. However, the former could be distinguished by subglobose and thick-walled basidiospores, and shorter cystidia $(10-15 \times 4.0-5.0 \,\mu\text{m})$ [59]. In addition, Ri. subpopulinus is similar with Ri. piceicola by having imbricate basidiocarps and ellipsoid basidiospores, but the former has shorter basidia $(7.6-12 \times 5-6 \,\mu\text{m})$ [60]. Because of the structural similarities in Rigidoporus, it is highly recommended that the DNA-based identification with combined regions, ITS, LSU, and rpb2.

4.6. Russula raoultii quél., comptes rendus de l'association française pour l'avancement des sciences 14 (2): 449 (1886) [MB#208070]

Pileus largely depressed center, 20 mm in diam, soft when fresh, becoming woody hard when dry; Pileus surface smooth, white (2.5Y, 9.5/1) to cream (2.5Y, 9/2), pale yellow (2.5Y, 8/2) in the center; Margin entire, rounded-obtuse, incurved. Lamellae adnate, subclose to close, pale orange yellow (10YR, 9.5/2) to very pale brown (10YR, 8/4) (Figures 1(H) and 2(F)). Stipe cylindrical, soft-cottony, white (2.5Y, 9.5/1), 25×10 mm. **Hyphal system** monomitic, hyphae occasionally septate and without clamp connections, mostly branched, thin-walled, becoming narrower toward the end with round apex, hyaline, 1.8-4.3 µm in diam. Basidia clavate, 4-spored, smooth, hyaline, $30-36.5 \times 8.0-8.5 \,\mu\text{m}$ (n = 25). Basidiospores broadly ellipsoid, napiform, striate with variable ridges, thin-walled, hyaline, sometimes bearing a big guttle $(5.0-)5.5-6.0(-7.0) \times 4.0-5.0$ $(-5.5) \mu m [Q = (1.2-)1.25-1.27(-1.38), Q = 1.26 \pm$ 0.1] (n = 44). Cystidia mainly fusiform, sometimes capitate, clavate, hyphoid, smooth, thin-walled, hyaline, $25-67 \times 3.3-12 \,\mu\text{m} \, (n=39)$.

4.6.1. Specimen examined

Korea. Gwangju Metropolitan City, Mudeungsan National Park, mixed hardwood forest, 35°08′26″ N, 126°58′00" E, solitary on soil covered with woody debris, 16 Jun 2020, S. L. Kwon, KUC20200616-18 (NIBRFG0000511375).

4.6.2. Remarks

Russula raoultii KUC20200616-18 was well-matched (99%) with Ru. raoultii Quél in the phylogenetic



analysis using the LSU region. For an accurate analysis, additional regions must be identified. Microscopic characteristics of Ru. raoultii KUC2020 0616-18 matched the original description [61]. However, the basidiospore of KUC20200616-18 is smaller than the size described in the original description $(7.0-8.0 \times 5.7-7.0 \,\mu\text{m})$.

4.7. Scutellinia crinita (bull.) Lambotte, Mémoires de la société royale des sciences de liège, sér. 2 14 (7): 299 (1887) [MB#433534]

Apothecia sessile, scattered to gregarious, discoid, undulate; Disk round, 2.5-4 mm in diam; Margin distinct, covered by blackish brown (7.5YR, 2.5/2) hairs which are stretched inward and outward (Figures 1(I) and 2(G)). Hymenial surface smooth, ceraceous, orange (7.5YR, 7/8) when fresh, pale yellow (5Y, 8/4) when dry. Marginal hair acicular, stiff, with pointed apex, 10-12 septate, thick-walled, brown (7.5YR, 4/4) to dark brown (7.5YR, 3/2), $1000-2300 \times 24-80 \,\mu m$. Internal hair similar with marginal hair, but much shorter, < 300 µm. Asci cylindrical, with round apex, 8-spored, smooth, thin-walled, hyaline, $98-140 \times 12.5-14 \,\mu\text{m}$ (n = 28). Ascospores ellipsoid to cylindrical, pustule-cristate, uniseriate, thin-walled, hyaline, containing two guttules, $17-19.5(-21) \times (9.0-)10-13(-13.5) \ \mu m \ [Q =$ (1.5-)1.6-1.7(-1.9), $Q = 1.65 \pm 0.15$] (n = 40).

4.7.1. Specimen examined

Korea. Gwangju Metropolitan City, Mudeungsan National Park, 35°07′84" N, 126°59′26" E, mixed hardwood forest, occurring on rotten branches, 23 Sep 2020, S. L. Kwon, KUC20200923-07 (NIBRFG0 000511370).

4.7.2. Remarks

Scutellinia crinita is similar to S. scutellata in terms of morphological characteristics, geographic distribution, and substrate [51]. Scutellinia crinita KUC20200923-07 has shorter asci and longer marginal hairs than S. scutellata [62]. Notably, Scutellinia species with globose to subglobose ascospores were found in soil. However, other Scutellinia species with ellipsoid ascospores have been found on decayed wood. This suggests that substrates may be crucial for understanding the diversity of the genus Scutellinia [50].

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