이학석사 학위논문

# Phylogenetic study of family Bankeraceae in Korea

Bankeraceae과의 계통학적 연구

2017년 8월

서울대학교 대학원

생명과학부

**Li, Vladimir**

# Phylogenetic study of family Bankeraceae in Korea

**Advisor: Professor Young Woon Lim, Ph.D.**

**A thesis submitted in partial satisfaction of the**

**Requirements for the degree Master of Science in**

**Biological Sciences**

**August 2017**

**Graduate School of Biological Sciences**

**Seoul National University**

# Phylogenetic study of family Bankeraceae in Korea

**Vladimir Li**

**School of biological sciences**

**The graduate school**

**Seoul National University**

## Abstract

Bankeraceae is an ectomycorrhizal family of stipitate hydnoid fungi, the unique feature of which is presence of spines under the basidiocarp as a mean of spores distribution, rather than pores and gills. Species of family Bankeraceae are under strong ecological concern in Europe, and included in the Red Data list in several countries, due to the fact that their numbers have reduced dramatically. Six genera and 98 species have been reported worldwide. In Korea, four genera and 11 species have been reported to date. Those scientific names have been taken based on comparisons of macromorphology to European and North American species without detailed descriptions. Recent studies showed, that Asian species of fungi differ greatly from those in Europe and North America when molecular comparison is applied. The purpose of this study is to re-evaluate species of family Bankeraceae in Korea using morphological characteristics and molecular analysis of internal transcribed spacer (ITS), large subunit

rRNA (LSU) region and the second largest subunit of RNA polymerase II (*rpb*2) region. 32 specimens were identified as 17 species, including one *Boletopsis*, six *Phellodon*, four *Hydnellum* and six *Sarcodon* species. Among these, two previously recorded species in Korea, five unrecorded species in Korea, three new species to the family and seven new candidate species to the family were identified. The resulting phylogram showed that *Boletopsis* and *Phellodon* formed monophyletic groups, while genera *Hydnellum* and *Sarcodon* remained to be complicated for most instances.

**Key words**: Bankeraceae, hydnoid fungi, ITS, LSU, *rpb*2, phylogenetic study

**Student number**: 2015-22296

# List of contents

# List of figures

# List of Tables

# 1. Introduction

## 1.1. Background

Family Bankeraceae was first proposed by Donk in 1961 and included genera *Boletopsis* and *Phellodon*, which initially belonged to family Thelephoraceae (order Thelephorales). Currently, family Bankeraceae belongs to order Thelephorales, which share a notable common feature – basidiospore shape, which is usually characterized by the presence of warts or small spines covering the surface; and includes 98 species spread across six genera, including *Bankera*, *Boletopsis*, *Corneroporus*, *Hydnellum*, *Phellodon* and *Sarcodon* (Cannon and Kirk 2007). All species of family Bankeraceae are ectomycorrhizal fungi, which form symbiotic relationships with trees, benefiting them by augmenting uptake of nutrients from soil, such as phosphorous, nitrogen and microelements and thus represent important ecological contributors (Chalot and Bran 1998; Stalpers 1993; Smith and Read 2008). Species of family Bankeraceae are associated with angiosperms and gymnosperms, specifically with species of Fagaceae and Pinaceae (Cannon and Kirk 2007).

## 1.2. Existing morphological and molecular data

The family is characterized by centrally or eccentrically stipitate annual basidiomata, monomitic hyphal system with or without clamp connections. Cystidia are present by occasion. Basidia are clavate, with or without basal clamp connections, with four sterigmata. Basidiospores are small, subgolose to ellipsoidal, thin-walled, do not stain in iodine (Cannon and Kirk 2007; Marren and Dickson 2000; Donk 1961). Genera *Boletopsis* and *Phellodon* share a common feature by having white spore print, *Boletopsis* is differentiated by having poroid hymenophore (Fayod 1899; Stalpers 1993). Also, *Phellodon* possesses spores with smaller and equidistantly located warts and fragrant odor (Karst 1881; Stalpers 1993). *Hydnellum* and *Sarcodon*, on the other hand, also share common characteristics including brown spore print and warted, irregular spores (Stalpers 1993). Species of *Sarcodon* may possess scales on their basidiocarp, which are unique to the genus (Stalpers 1993). *Phellodon* and *Hydnellum* share morphology of their fruiting bodies, making it difficult to differentiate them in nature (Parfitt *et al*. 2007). However, different spore print colors and spores morphology are the key to distinguish them (Stalpers 1993). Genus *Bankera* can be differentiated by the presence of white spore print, however, unlike *Phellodon* it lacks zonation on the basidiocarp and has fleshy fruiting body (Stalpers 1993). Genus *Corneroporus* consists of a single species – *Corneroporus subcitrinus* and was originally described by Corner as *Boletopsis* (Corner 1989; Hatton 2001). *Corneroporus* can be differentiated by its spores being echinulate and hyaline, while

*Boletopsis* species usually have angular spores with indeterminate warts (Stalpers 1993; Hatton 2001).

Molecular analysis more and more accompanies traditional methods of identification of fungal species. Internal transcribed spacer (ITS) is a common molecular marker and it was proposed as a major barcode gene for fungi (Schoch *et al*. 2012). Huge pool of ITS region sequences is available for public use (Schoch *et al*. 2012). ITS region was rendered efficient to separate genera *Sarcodon, Hydnellum* and *Phellodon* (Baird *et al*. 2013). The effectiveness was also demonstrated for differentiating species of *Phellodon* and *Hydnellum* (Parfitt *et al*. 2007). Interspecific variations in ITS 1 and ITS 2 of rDNA allow to determine the degree of relatedness of closely associated taxa (White *et al*. 1990). In addition, two more genes were proven to be effective for distinguishing fungal species, which are *rpb*2 (the second largest subunit of RNA polymerase II) and LSU (large-subunit rRNA) regions. Unlike ITS, *rpb*2 gene has single copy nature, which results in the constant length of the resulting amplified sequences, and thus, it is much less prone to PCR bias, which is observed with ITS (Větrovský *et al*. 2015). Also, taxonomic sensitivity of *rpb*2 is much greater than that of ITS, proving its effectiveness to differentiate basal fungal lineages (Větrovský *et al*. 2015). LSU gene has been used extensively for fungal phylogeny and even became a standard for identification. It was previously used for the creating AFTOL (Assembling the Fungal Tree of Life) (Arnold *et al*. 2009; Blackwell 2011; James *et al*. 2006; Öpik *et al*. 2010). This gene is an essential marker for analysis of basal fungal lineages or assessment of new lineages (James *et al*. 2006;

Öpik *et al*. 2010).

According to Baird *et al*. (2013) genus *Phellodon* is a monophyletic group to *Sarcodon* and *Hydnellum* and resolved with a solid bootstrap value of 100% (Baird *et al*. 2013). The relationship between *Sarcodon* and *Hydnellum* remains to be complicated, since neither genus can be classified as monophyletic (Baird *et al*. 2013).

## 1.3. Korean Bankeraceae

Currently, four genera and 11 species reported in South Korea (Table 4), including one *Boletopsis* (*B. leucomelaena*), four *Hydnellum* (*H. aurantiacum*, *H. caeruleum*, *H. concrescens*, *H. ferrugineum*), three *Phellodon* (*P. melaleucus*, *P. niger*, *P. tomentosus*) and three *Sarcodon* (*S. imbricatus*, *S. scabrosus*, *S. underwoodii*) (Lee *et al*. 2015). Species of genera *Bankera* and *Corneroporus* were not reported (Lee *et al*. 2015). The existing inventory was created based on the reference data obtained from European and North American species.

*Boletopsis leucomelaena* was first reported in 1940 under the name *Polyporus leucomelas* (Kaburagi 1940). *Hydnellum aurantiacum* was reported back in 1978 as *Calodon aurantiacum* (Korean Society of Mycology 1978). *Hydnellum caeruleum* is one of the recent reports in Korea (Lee and Jung 2005). *Hydnellum concrescens* and *Hydnellum ferrugineum* were previously known in Korea as *Hydnellum zonatum* and *Hydnum ferrugineum* correspondingly, and were reported in 1957 (Lee and Lee 1957). Initial report of *Phellodon melaleucus* was dated on 1985

along with complete morphological description (Lee and Hong 1985). *Phellodon niger* was reported on 1990 by Lee. Another recent report is *Phellodon tomentosus* (Lee and Jung 2005); species *Sarcodon aspratus* and *Sarcodon imbricatus* are currently same species designated as *Sarcodon imbricatus*, and were first reported on 1940 (Kaburagi 1940). *Sarcodon scabrosus* was reported in 1985 with complete morphological description (Lee and Hong 1985). Lastly, *Sarcodon underwoodii* was reported in 1978 as *Sarcodon* murillii (Korean Society of Mycology 1978).

All reports were done based on macro- and micromorphological features. However, there is a high chance for misidentified species to take place, as well as new species being mistaken as existing ones. Notable examples from previous European studies of family bankeraceae are species of genus *Sarcodon*: *S. imbricatus* and *S. squamosus*, which are almost indistinguishable in their characters, including both macro- and micromorphology (Johannesson *et al*. 1999). Genera *Hydnellum* and *Phellodon* share multiple macromorphological signs making them complicated for identification in a wild (Parfitt *et al*. 2007). Taking into considerations experience from earlier works dedicated to studying this family, it is reasonable to assume the need to re-evaluate these species based on combined morphological and molecular analyses and establish their phylogenetic relationships.

## 1.4. Objective

Previous identifications of Korean Bankeraceae were solely relied on morphological keys derived from European species with similar morphology (Korean Society of Mycology 1978; Lee and Cho 1975). However, recent trends in fungal identification, involving molecular works, demonstrated, that fungi found in Asia can be different from those from Europe and the United States (Park *et al*. 2013; Lee *et al*. 2015). This is especially the case with ectomycorrhizal fungi, which do not exhibit intercontinental conspecificity (Lee *et al*. 2007; Stubbe *et al*. 2008). The species of the family in Korea have never been evaluated based on the molecular data analysis and their phylogenetic relationships also have never been studied before. Thus, the main objective of this study is to re-evaluate species of family Bankeraceae based on the combined morphological and molecular analyses and study phylogenetic relationships between them.

# 2. Materials and Methods

## 2.1. Collection of Bankeraceae in Korea

A total of 32 specimens of Bankeraceae family were used in this study. Specimens were obtained from Seoul National University Fungal Collection (SFC), Korean National Institute of Biological Resources (NIBRF) and Kangwon National University (KNU) (Table 1). The samples were initially identified based on their morphology and sorted with accordance to the features of their corresponding genera as *Boletopsis* (2 specimens), *Phellodon* (9 specimens), *Hydnellum* (10 speciemens) and *Sarcodon* (11 specimens) (Table 1).

## 2.2. Microscopic observation

Species of Bankeraceae were verified by their microscopic characters: basidia, basidiospores and hypha. Dried samples were prepared on slides using 3% KOH solution with Phloxin or Congo Red staining dyes and were observed using Nikon 80i light microscope (Nikon, Tokyo, Japan) (Kim *et al*. 2016; Min *et al*. 2014). Samples were assorted based on the detailed description of morphological characters presented by Stalpers (1993).

**Table 1**. List of Bankeraceae specimens, identification history, final identification using ITS sequence and distribution information used in this study

| Final ID | Preliminary ID | Specimen No. | Locality | Date |
|---|---|---|---|---|
| *Boletopsis* sp. | *Boletopsis leucomelaena* | SFC20161002-01 | Dogye-eup, Samcheok-si, Gangwon-do, Korea | 2016.10.02 |
| *Boletopsis* sp. | *Boletopsis perplexa* | KA12-1573 | Chubu-myeon, Geumsan-gun, Chungcheongnam-do, Korea | 2012.09.26 |
| *Hydnellum earlianum* | *Hydnellum* sp. | TPML20120808-040 | Daehang-myeon, Gimcheon-si, Gyeongsangbuk-do, Korea | 2012.08.08 |
| *Hydnellum ferrugineum* | *Hydnellum caeruleum* | KA12-1584 | Chubu-myeon, Geumsan-gun, Chungcheongnam-do, Korea | 2012.09.26 |
| *Hydnellum* sp. 2 | *Hydnellum caeruleum* | TPML20120905-063 | Noeun-myeon, Chungju-si, Chungcheongbuk-do, Korea | 2012.09.05 |
| *Hydnellum* sp. 2 | *Phellodon melaleucus* | SFC20150828-40 | Seolcheon-myeon, Muju-gun, Jeollabuk-do, Korea | 2015.08.28 |
| *Hydnellum* sp. 2 | *Hydnellum concrescens* | SFC20140822-13 | Girin-myeon, Inje-gun, Gangwon-do, Korea | 2014.08.22 |
| *Hydnellum* sp. 2 | *Hydnellum caeruleum* | SFC20140725-28 | Donghyang-myeon, Jinan-gun, Jeollabuk-do, Korea | 2014.07.28 |
| *Hydnellum* sp. 2 | *Hydnellum concrescens* | SFC20120926-19 | Jeongan-myeon, Gongju-si, Chungcheongnam-do, Korea | 2012.09.26 |
| *Hydnellum* sp. 2 | *Hydnellum* sp. | TPML20130924-02 | Geumgangsong-myeon, Uljin-gun, Gyeongsangbuk-do, Korea | 2013.09.24 |
| *Hydnellum* sp. 1 | *Hydnellum* sp.1 | SFC20151015-04 | Cheongha-myeon, Buk-gu, Pohang-si, Gyeongsangbuk-do, Korea | 2015.07.25 |
| *Hydnellum* sp. 1 | *Phellodon melaleucus* | SFC20150725-07 | Sokrisan-myeon, Boeun-gun, Chungcheongbuk-do, Korea | 2015.07.25 |
| *Phellodon fuligineoalbus* | *Phellodon fuligineoalbus* | SFC20151015-03 | Cheongha-myeon, Buk-gu, Pohang-si, Gyeongsangbuk-do, Korea | 2015.07.25 |
| *Phellodon koreeanus* | *Phellodon* sp. 1 | SFC20140723-54 | Jeongcheon-myeon, Jinan-gun, Jeollabuk-do, Korea | 2014.07.23 |
| *Phellodon koreeanus* | *Phellodon* sp. | NIBRFG0000138138 | Hogeun-dong, Seogwipo-si, Jeju-do, Korea | 2014.07.15 |
| *Phellodon niger* | *Phellodon niger* | SFC20150701-110 | Jocheon-eup, Jeju-si, Jeju-do, Korea | 2015.07.01 |

**Table 1.** Continued

| Final ID | Preliminary ID | Specimen No. | Locality | Date |
|---|---|---|---|---|
| *Phellodon orientizonatus* | *Hydnellum concrescens* | TPML20140827-115 | Daehang-myeon, Gimcheon-si, Gyeongsangbuk-do, Korea | 2014.08.27 |
| *Phellodon orientizonatus* | *Hydnellum caeruleum* | SFC20140911-03 | Donghyang-myeon, Jinan-gun, Jeollabuk-do, Korea | 2014.09.11 |
| *Phellodon* sp. | *Phellodon niger* | NIBRFG0000138070 | Seohong-dong, Seogwipo-si, Jeju-do, Korea | 2014.07.15 |
| *Phellodon confluens* | *Phellodon confluens* | SFC20150902-93 | Inje-eup, Inje-gun, Gangwon-do, Korea | 2015.09.02 |
| *Phellodon confluens* | *Hydnellum caeruleum* | SFC20140827-15 | Beonam-myeon, Jangsu-gun, Jeollabuk-do, Korea | 2014.08.27 |
| *Sarcodon squamosus* | *Sarcodon imbricatus* | SFC20141001-12 | Girin-myeon, Inje-gun, Gangwon-do, Korea | 2014.10.01 |
| *Sarcodon glabrus* | *Sarcodon* sp. | TPML20130628-34 | Chunyang-myeon, Bonghwa-gun, Gyeongsangbuk-do, Korea | 2013.06.28 |
| *Sarcodon glabrus* | *Sarcodon scabrosus* | SFC20140822-38 | Kangwon-do, Inje-gun, Mt. Jeombong | 2014.08.22 |
| *Sarcodon* sp. 1 | *Sarcodon imbricatus* | KM07-0701 | Buk-myeon, Inje-gun, Gangwon-do, Korea | 2007.09.08 |
| *Sarcodon* sp. 2 | *Sarcodon imbricatus* | SFC20160921-01 | Bukbang-myeon, Hongcheon-gun, Gangwon-do, Korea | 2016.09.21 |
| *Sarcodon* sp. 2 | *Sarcodon scabrosus* | SFC20160923-43 | Dongsan-myeon, Chuncheon-si, Gangwon-do, Korea | 2016.09.23 |
| *Sarcodon* sp. 3 | *Sarcodon imbricatus* | KM04-0159 | Buk-myeon, Inje-gun, Gangwon-do, Korea | 2004.08.25 |
| *Sarcodon* sp. 3 | *Sarcodon imbricatus* | KM04-0137 | Buk-myeon, Inje-gun, Gangwon-do, Korea | 2004.07.21 |
| *Sarcodon* cf. *underwoodii* | *Sarcodon imbricatus* | SFC20160923-44 | Dongsan-myeon, Chuncheon-si, Gangwon-do, Korea | 2016.09.23 |
| *Sarcodon* cf. *underwoodii*. | *Sarcodon underwoodii* | TPML20101008-133 | Yeonpung-myeon, Goesan-gun, Chungcheongbuk-do, Korea | 2010.10.08 |
| *Sarcodon* cf. *underwoodii*. | *Sarcodon scabrosus* | KA12-1410 | Buk-myeon, Ulleung-gun, Gyeongsangbuk-do, Korea | - |

## 2.3. DNA extraction, PCR amplification, and sequencing

A piece of tissue from dried specimens was placed in 2X CTAB buffer solution. DNA was extracted from a tissue samples using modified cetyltrymethyl ammonium bromide (CTAB) extraction protocol (Rogers and Bendich 1994). The ITS region was then amplified using different set of primers, depending on the samples (NSI1, NLB4, ITS1F and ITS4B) (Table 2) (Gardes and Bruns 1993; Martin and Rygiewicz 2005). NSI1 (forward) and NLB4 (reverse) were used for the 1st run of PCR reaction, while ITS1F (forward) and ITS4B (reverse) were used for the 2nd run (nested) of PCR reaction.

The *rpb*2 region was amplified using primers fRPB2-5F, bRPB2-6F1, bRPB2-6F, bRPB2-8.2R, bRPB2-7.1R and bRPB2-7R (Table 2) (Hibbett lab; Hall lab). This region was amplified using three steps PCR reactions: 1st reaction with fRPB2-5F (forward) and bRPB2-8.2R (reverse), 2nd reaction with bRPB2-6F1 (forward) and bRPB2-7.1R (reverse) and 3rd reaction with bRPB2-6F (forward) and bRPB2-7R (reverse). In addition, in order to amplify species of genus *Phellodon*, a specific set of primers was prepared for two steps PCR reactions including PheF1 (forward) and PheR1 (reverse) for the 2nd reaction and PheF2 (forward) and PheR2 (reverse) (Table 2) for the 3rd reaction. The primers were designed using program Primer3 and were based on the *rpb*2 sequences from the first PCR run (fRPB2-5F and bRPB2-8.2R primers). The LSU region was amplified using both ITS primers and the following primers: LROR, LR7, and LR5 (Table 2) (Vigalys lab). For the 1st

PCR reaction usually NSI1 or ITS1F was used as forward primer, while LR7 or LR5 was used as reverse primer. The second PCR involved usage of either ITS1F (if NSI1 was used for the first run) or LROR as forward primers and LR5 as a reverse primer.

All PCR reactions were performed on thermal cyclers either C1000TM; Bio-Rad (Richmond, CA, USA) or Biometra TProfessional Standard 96 (Göttingen, Germany) using AccuPower PCR Premix (Bioneer Co., Daejeon, Korea) based on the protocol outlined by Min *et al*. (2014). The final volume of the mixture was 20 µl, containing 1 µl of each primer and 1 µl of DNA. The PCR conditions for the amplification of the ITS were as follows: 95°C for 5 min, followed by 35 cycles of 95°C for 40 sec, 55°C for 40 sec and 72°C for 1 min, and a final elongation step at 72°C for 10 min. The settings for the 2nd PCR were 95°C for 5 min, followed by 35 cycles of 95°C for 30 sec, 58°C for 30 sec and 72°C for 40 sec, and a final elongation step at 72°C for 10 min. PCR conditions for the first run of PCR reaction for *rpb*2 were 95°C for 4 in, followed by 35 cycles of 95°C for 1 30 min, 50°C for 1 30 min at a rate 0.3, followed by 72°C for 1 30 min, and a final elongation step at 72°C for 10 min. 2$^{nd}$ and 3$^{rd}$ of *rpb*2 region were same as 1$^{st}$ run and 2$^{nd}$ run for ITS region, correspondingly. PCR conditions sets for the amplification of the LSU region were as follows: 95°C for 5 min, followed by 35 cycles of 95°C for 1 min, 55°C for 1 min and 72°C for 1 30 min, and a final elongation step at 72°C for 10 min. The 2nd PCR conditions remained the same as for the ITS region. The resulting PCR products were imaged on a 1% agarose gel stained with EcoDye DNA staining solution (Solgent, South Korea) and purified using the Expin™ PCR Purification Kit (GeneAll

Biotechnology, Seoul, Korea). Samples were then sequenced by Macrogen (Seoul, Korea) on an automated DNA sequencer (ABI3700; Applied Biosystems, Foster City, CA, USA) using corresponding primers utilized for the PCR reaction.

## 2.4. Sequence analysis

Resulting sequences delivered by Macrogen were edited and aligned using MEGA ver. 7 (Kumar *et al*. 2016). Reference sequences were obtained from the GeneBank. Multiple sequence alignment was performed using Multiple Alignment Fast Fourier Transform (MAFFT) (Katoh and Stanley 2013). Each sequence was manually proofread and adjusted where necessary. Maximum likelihood (ML) analysis was performed for the ITS regions using CIPRES program with 1,000 bootstrap replicates (Miller *et al*. 2010).

In order to define the genetic variation of Bankeraceae species using ITS region, maximum interspecific sequence dissimilarities were calculated for each sister clades appearing in the ITS phylogram. Region ITS1-5.8S-ITS2 was analyzed and compared. Maximum interspecific sequence dissimilarities were calculated using Mega ver. 7 software (Kumar *et al*. 2016).

**Table 2.** Information on the primers used for the PCR in this study

| **Region** | **Primer** | **Sequence** (5' - 3') | **Reference** |
|---|---|---|---|
| **ITS** | NSI1 | GATTGAATGGCTTAGTGAGG | Martin and Rygiewicz (2005) |
| | NLB4 | GGATTCTCACCCTCTATGAC | Martin and Rygiewicz (2006) |
| | ITS1F | CTTGGTCATTTAGAGGAAGTAA | White *et al*. (1990) |
| | ITS4B | CAGGAGACTTGTACACGGTCC | Gardes and Bruns (1993) |
| ***rpb*2** | fRPB2-5F | GAYGAYMGWGATCAYTTYGG | Liu, Whelen, and Hall (1999) |
| | bRPB2-8.2R | CTNCGGAANAGRCCRCGRTC | Matheny *et al*. (2007) |
| | bRPB2-6F1 | CACAAYCANCAYTGGGGWATGGT | Hibbett lab |
| | bRPB2-7.1R | CCCATRGCYTGYTTMCCCATDGC | Matheny (2005) |
| | bRPB2-6F | TGGGGYATGGTNTGYCCYGC | Hall lab |
| | bRPB2-7R | GAYTGRTTRTGRTCRGGGAAVGG | Hall lab |
| | PheF1 | TATYTCYGTYGGTTCTYTATCTGCT | This study |
| | PheR1 | CAAACGRGCRGAAGGATCATACTC | This study |
| | PheF2 | CTGCTCCCGTTATYGAAWTYTTGG | This study |
| | PheR2 | CTCGCCYTCRTKTTCRTGAGTTTC | This study |
| **LSU** | LROR | ACCCGCTGAACTTAAGC | Vilgalys and Hester (1990) |
| | LR7 | TACTACCACCAAGATCT | Vilgalys and Hester (1990) |
| | LR5 | TCCTGAGGGAAACTTCG | Vilgalys and Hester (1990) |

The Hibbett lab:http://www2.clarku.edu/faculty/dhibbett/

Vilgalys Website: http://sites.biology.duke.edu/fungi/mycolab/primers.htm

Hall Lab: http://faculty.washington.edu/benhall/

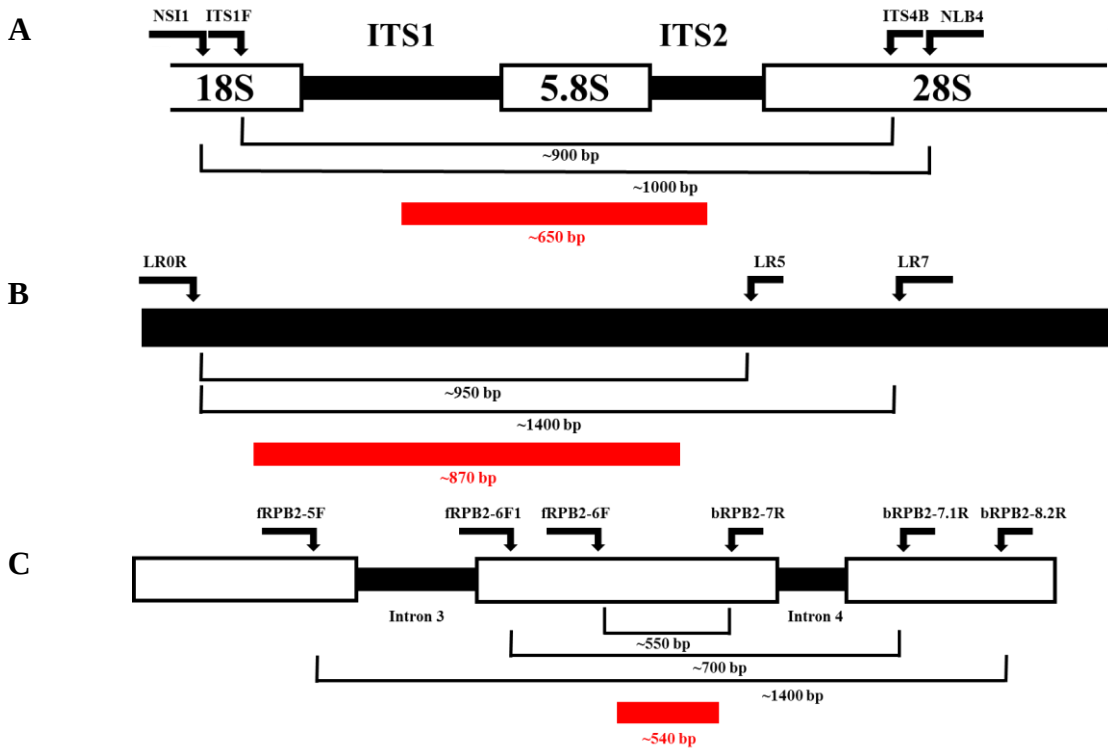


**Figure 1.** Location of primers for PCR amplification. A) ITS region, B) LSU region, C) *rpb*2 region

*regions marker red indicate final sequences used for analysis.

**Table 3.** List of the reference sequences used in this study

| Species | Origin | Accession No. |
|---|---|---|
| *Boletopsis grisea* | USA | JF908784 |
| *Boletopsis grisea* | Europe | DQ408768 |
| *Boletopsis leucomelaena* | USA | DQ484064 |
| *Boletopsis* sp . | Europe | DQ408767 |
| *Boletopsis* sp . | Europe | DQ408766 |
| *Boletopsis subsquamosa* | Canada | FJ845401 |
| *Hydnellum concrescens* | USA | KC571715 |
| *Hydnellum earlianum* | USA | KC571724 |
| *Hydnellum earlianum* | USA | JN135179 |
| *Hydnellum ferrugineum* | USA | JN135185 |
| *Hydnellum ferrugineum* | USA | KC571729 |
| *Hydnellum scrobiculatum* | USA | AY569032 |
| *Hydnellum scrobiculatum* | USA | JN135181 |
| *Hydnellum* sp. 1 | USA | JN135177 |
| *Hydnellum* sp. 2 | USA | JN135187 |
| *Hydnellum* sp. 2 | USA | KC571717 |
| *Hydnellum spongiosipes* | USA | KC571741 |
| *Hydnellum spongiosipes* | USA | JN135184 |
| *Phellodon alboniger* | USA | KC571750 |
| *Phellodon confluens* | USA | KC571756 |
| *Phellodon confluens* | USA | KC571755 |
| *Phellodon ellisianus* | USA | JN135204 |
| *Phellodon ellisianus* | USA | KC571758 |
| *Phellodon melaleucus* | USA | KC571763 |
| *Phellodon melaleucus* | USA | JN135197 |
| *Phellodon niger* | USA | KC571766 |
| *Phellodon niger* | USA | JN135202 |
| *Phellodon* sp. | USA | KC571746 |
| *Phellodon* sp. | USA | KC571747 |
| *Bankera fuligineoalba* | USA | KC571760 |
| *Bankera fuligineoalba* | Europe | EU784181 |

**Table 3.** Continued

| Species | Origin | Accession No. |
|---|---|---|
| *Sarcodon imbricatus* | Europe | AF103888 |
| *Sarcodon imbricatus* | Europe | AF103889 |
| *Sarcodon joeides* | USA | KC571773 |
| *Sarcodon joeides* | USA | JN135193 |
| *Sarcodon scabripes* | S. America | EU293829 |
| *Sarcodon scabripes* | USA | JN135191 |
| *Sarcodon squamosus* | Europe | AF103892 |
| *Sarcodon squamosus* | Europe | AF103890 |
| *Sarcodon underwoodii* | USA | KC571781 |
| *Sarcodon underwoodii* | USA | JN135189 |
| *Tomentella pilosa* (outgroup) | Europe | AF272925 |
| *Tomentella pilosa* (outgroup) | Europe | AJ421252 |
| *Boletopsis grisea* | Europe | AY586636 |
| *Boletopsis grisea* | Canada | EU522836 |
| *Hydnellum aurantiacum* | Europe | AF347113 |
| *Phellodon niger* | Europe | AY586694 |
| *Phellodon niger* | Canada | EU522797 |
| *Phellodon tomentosus* | USA | AF518637 |
| *Sarcodon imbricatus* | Europe | AY586711 |
| *Sarcodon* sp. | USA | EF561639 |
| *Tomentella* sp. (outgroup) | S. America | KT032103 |
| *Tomentella* sp. (outgroup) | S. America | KT032102 |
| *Boletopsis leucomelaena* | USA | GU187820 |
| *Hydnellum geogenium* | USA | DQ408133 |
| *Tomentella* sp. (outgroup) | USA | DQ835999 |

# 3. Results

## 3.1. Morphological and molecular analysis

32 specimens were identified to four genera, including *Boletopsis*, *Phellodon*, *Hydnellum* and *Sarcodon* (Table 1). First, species were assigned to three groups based on their macroscopic characteristics: 1) *Boletopsis*; 2) *Sarcodon*; 3) *Hydnellum* and *Phellodon*. Species of genus *Boletopsis* were identified based on their feature – poroid hymenophore, which is unique to the genus. Also, species that did not qualify for *Boletopsis*, *Hydnellum* or *Phellodon* were also assigned to genus *Sarcodon.* Species of genus *Sarcodon* were identified by the presence of scales breaking from the basidiocarp or having smooth basidiocarp and generally large fruiting body. Species of genera *Hydnellum* and *Phellodon* share general macromorphology and were grouped together due to ambiguity associated with their identification. But overall they were selected based on their zonated basidiocarp, relatively small dimensions and woody and brittle fruiting body.

Second, specimens were assigned to the four corresponding genera based on combined macro- and microscopic analysis data. Species of genera *Boletopsis* and *Sarcodon* featured spores with indeterminate shape and warts. Microscopic analysis of genera *Hydnellum* and *Phellodon* allowed to properly assign species to their corresponding genus. Species of *Hydnellum* featured basidiospores similar to those

of *Sarcodon*, featuring inequidistant warts and indeterminate shape, while species of *Phellodon* had ellipsoid, echinulate basidiospores with fine equidistant spines, covering their surface.

Third, species of the four genera were further sub-grouped within their corresponding genus based on their common morphology. Thus, genera *Boletopsis*, *Hydnellum* and *Phellodon* were assigned as single group genera, while genus *Sarcodon* was divided into two sub-groups. One group had specimens with scaly basidiocarps, while the other group contained samples with smooth basidiocarps.

The ITS region was amplified for all specimens. On the ML tree, 32 specimens were divided into four groups (G1 – G4) (Figure 2). 32 specimens were identified across 17 species including 7 recorded species and 10 new species. The species were distributed across four genera as following: *Boletopsis* (1), *Phellodon* (6), *Hydnellum* (4) and *Sarcodon* (6). Of those species only two were identified from previous Korean records: *Hydnellum ferrugineum* and *Phellodon niger*. Only three species were identified properly based on the primary morphological identification, including species of *Hydnellum* sp. 1, *Phellodon fuligineoalbus* and *Phellodon niger*. The remaining species were misidentified or labeled as "sp.". All species grouped in ITS tree were in correlation with their morphological identification. The data for *rpb*2 and LSU trees were much less efficient: 1) fewer samples were successfully amplified; 2) the existing database did not contain sufficient number of reliable reference sequences to compare our results with. Thus, the data from *rpb*2 and LSU trees were

not used for accounting species of Bankeraceae.

Based on the findings from morphological and molecular analyses, 32 specimens were re-evaluated as 17 species across four genera. Overall, *Hydnellum earlianum* (1), *Hydnellum ferrugineum* (1), *Hydnellum* sp. 1 (2), *Phellodon fuligineoalbus* (1), *Phellodon niger* (1), *Phellodon confluens* (2), *Sarcodon squamosus* (1); as well as new species *Boletopsis* sp. (2), *Hydnellum* sp. 2 (6), *Phellodon koreeanus* (2), *Phellodon orientizonatus* (2), *Phellodon* sp. (1), *Sarcodon glabrus* (2), *Sarcodon* sp. 1 (1), *Sarcodon* sp. 2 (2), *Sarcodon* sp. 3 (2) and *Sarcodon* cf. *underwoodii* (3) were used in this study (Table 1 and Figure 2).

**Table 4.** Microscopic characteristics of previously recorded Korean species of family Bankeraceae

| **Species** | **Reference** | **Basidiocarp** (cm) | **Spines/Pores** (mm) | **Stipe** (cm) | **Basidia*** (μm) | **Basidiospore*** (μm) |
|---|---|---|---|---|---|---|
| *Boletopsis leucomelaena* | Karst 1961 | 4~5 x 8~10 | 1~3 per mm (pores) | 3~5 | 17~30 x 5.5~8.5 | 4.5~5 x 6.5~8 |
| *Boletopsis* sp. | Korean species | up to 6.5 | 1~3 per mm (pores) | up to 7 x 1 | 17~27 x 5~8.5 | 4~5.5 x 4~5 |
| *Phellodon confluens* | Korean species | up to 3.5 | up to 2 | up to 3 x 1 | 24~38 x 5.5~7 | 4~5 x 4~4.5 |
| *Phellodon fuligineoalbus* | Korean species | up to 7.5 | up to 4 | up to 4 x 2 | 24~31 x 5~6 | 4~5 x 3.5~4 |
| *Phellodon niger* | Karst 1961 | up to 6 | up to 3 | | 22~35 x 4~7 | 3.5~5 x 3~4.5 |
| | Korean species | up to 4 | up to 3 | up to 3 x 1 | 21.0 – 30.0 x 5 – 8 | 5~6 x 4~5.5 |
| *Phellodon melaleucus* | Karst 1961 | up to 7 | up to 2 | | 20~30 x 4~6 | 3~5 x 3~5 |
| *Phellodon tomentosus* | Banker 1906 | up to 7 | up to 2 | | 20~30 x 4~6 | 3~4.2 x 2.7~4 |
| *Phellodon koreeanus* | Korean species | up to 3 | up to 1 | up to 1 x 1.5 | 27~35 x 5~7 | 4~6 x 4~5.5 |
| *Phellodon orientizonatus* | Korean species | up to 3.5 | up to 4 | up to 0.5 x 1.5 | 25~34 x 5.5~7.5 | 4~5.5 x 4~5 |
| *Phellodon* sp. | Korean species | up to 2.5 | up to 1 | up to 2 x 1 | 25~38 x 5~8 | 4.5~6 x 4~5.5 |
| *Hydnellum aurantiacum* | Karst 1961 | up to 7 | up to 5 | | 25~40 x 5~8 | 5.5~8 x 4.5~6.5 |
| *Hydnellum caeruleum* | Karst 1961 | up to 7 | up to 4 | | 25~40 x 5~7 | 4.2~6.5 x 3.5~5 |
| *Hydnellum concrescens* | Banker 1906 | up to 7~10 | up to 3 | | 25~40 x 5.5~7 | 4.5~6.5 x 3.5~5 |
| *Hydnellum ferrugineum* | Karst 1961 | up to 7~10 | up to 6 | | 25~40 x 5.5~8 | 5~6.3 x 3.5~4.5 |
| | Korean species | up to 4 | up to 5 | up to 3.5 x 1.2 | 28~35 x 5~7 | 5~6 x 4~6 |
| *Hydnellum earlianum* | Korean species | up to 3 | up to 2 | up to 2 x 1.5 | 22~35 x 5~8.5 | 4~7 x 4~6 |
| *Hydnellum* sp. 1 | Korean species | up to 3.5 | up to 2 | up to 4 x 2 | 21~34 x 6~8 | 4.3~6 x 3~5 |
| *Hydnellum* sp. 2 | Korean species | up to 3.5 | up to 2 | up to 3.5 x 0.5 | 28~40 x 6~9 | 4~6 x 4~6 |

**Table 4.** Continued

| **Species** | **Reference** | **Basidiocarp** (cm) | **Spines/Pores** (mm) | **Stipe** (cm) | **Basidia*** (μm) | **Basidiospore*** (μm) |
|---|---|---|---|---|---|---|
| *Sarcodon imbricatus* | Karst 1961 | up to 11~15 | up to 10 | | 25~45 x 5~8 | 7~10 x 6.5~8.5 |
| *Sarcodon scabrosus* | Karst 1961 | up to 10~14 | up to 10 | | 35~50 x 5~9 | 5~6.5 x 3.5~5 |
| *Sarcodon squamosus* | Korean species | up to 6 | up to 2 | up to 5 x 1.5 | 35~42 x 6.5~10 | 7~8.5 x 6~8 |
| *Sarcodon underwoodii* | Banker 1906 | up to 10 | up to 6 | | 30~50 x 6~10 | 5~6.5 |
| *Sarcodon* cf. *underwoodii* | Korean species | up to 3 | up to 4 | up to 4.5 x 3 | 30~45 x 6~10 | 6~7 x 5.5~6.5 |
| *Sarcodon* sp. 1 | Korean species | up to 3 | up to 3 | | 35~48 x 6.5~10 | 7.5~8.5 x 6.5~8 |
| *Sarcodon* sp. 2 | Korean species | up to 8 | up to 2 | up to 8 x 2 | 31~45 x 6.5~9.5 | 6~7.5 x 5~6 |
| *Sarcodon* sp. 3 | Korean species | up to 8 | up to 5 | up to 5 x 1.5 | 30~45.5 x 7~8.5 | 6~7.5 x 5~6.5 |
| *Sarcodon glabrus* | Korean species | up to 3 | up to 2 | up to 0.5 x 2 | 31~48 x 6~10 | 5~7 x 4~5.5 |

*Data obtained from up to 20 basidiospores and 10 basidia. Measurements are given as length range x width range

## 3.2. Phylogenetic analysis and interspecific variation

Based on the resulting phylogram of the ITS region group G1 was monophyletic to the other groups with a resolution of 74% (Figure 2). The group consisted of species of genera *Hydnellum* and *Sarcodon*, which were intermixed with each other (Figure 3). Although, the groups were supported by moderate bootstrap values, neither of them was monophyletic. The maximum interspecific dissimilarity between sister clades of *Hydnellum ferrugineum* and *Hydnellum spongiosipes* was accounted to be 12%. And the maximum interspecific dissimilarity between reference *Hydnellum* sp. and Korean *Hydnellum* sp. 2 clades was accounted to be no greater than 6%. For the species of *Sarcodon* (G1), the maximum dissimilarity between sister clades of reference *Sarcodon joeides* and Korean *Sarcodon glabrus* was accounted for 7%; while the value for *Sarcodon underwoodii* and Korean *Sarcodon* cf. *underwoodii* was accounted to range from 4% to the maximum of 9%. Group G2 solely included species of genus *Sarcodon* and had bootstrap value of 83% (Figure 4). The group is monophyletic and also contained species of *Sarcodon imbricatus*, which is the type species for the genus and can possibly be the group of true *Sarcodon*. The maximum interspecific variation between sister clades of *Sarcodon imbricatus* and *Sarcodon squamosus* was between 4.2 – 4.7%. And the dissimilarity between Korean *Sarcodon* sp. 2 and *Sarcodon* sp. 3 was accounted for being around 7%. Group G3 is the clade

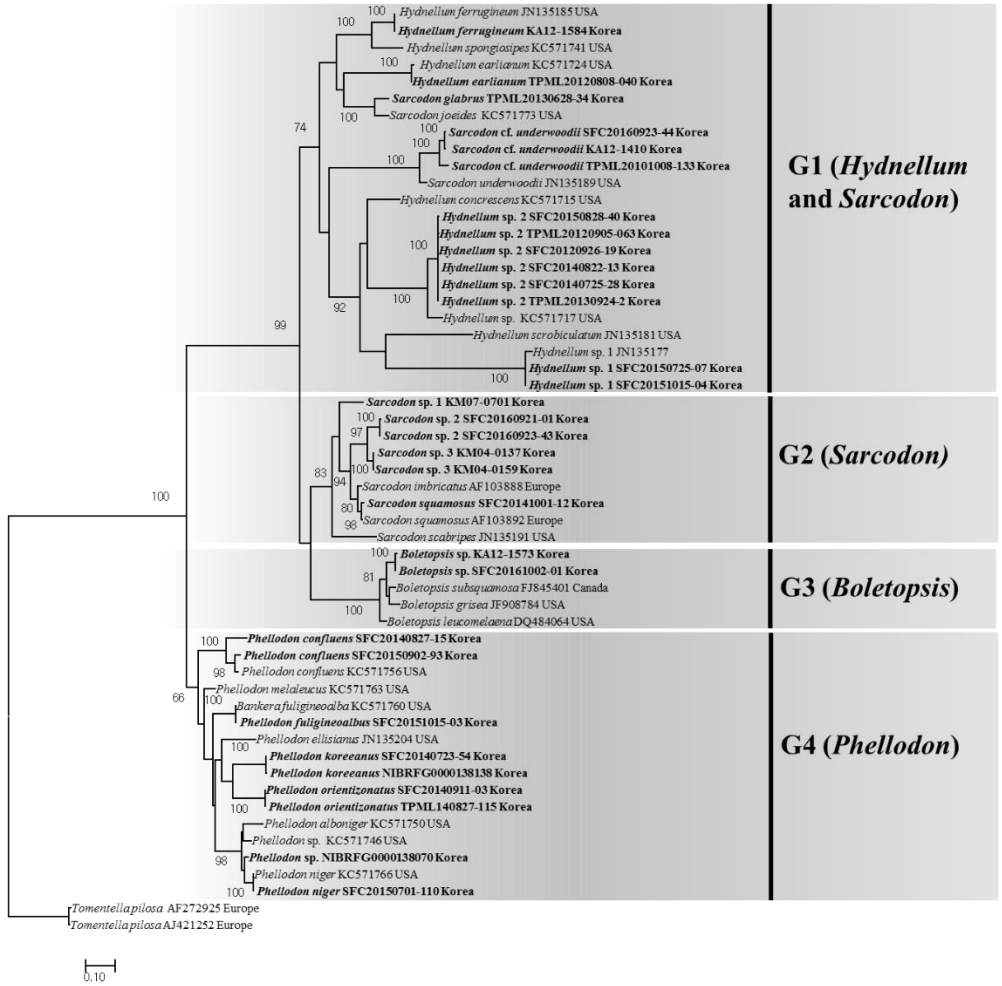


**Figure 2**. Phylogram generated from Maximum Likelihood (RAxML) analysis based on ITS sequence data of four genera of family Bankeraceae**.** ML bootstrap support values are indicated above or below the nodes and the Korean species are in bold.

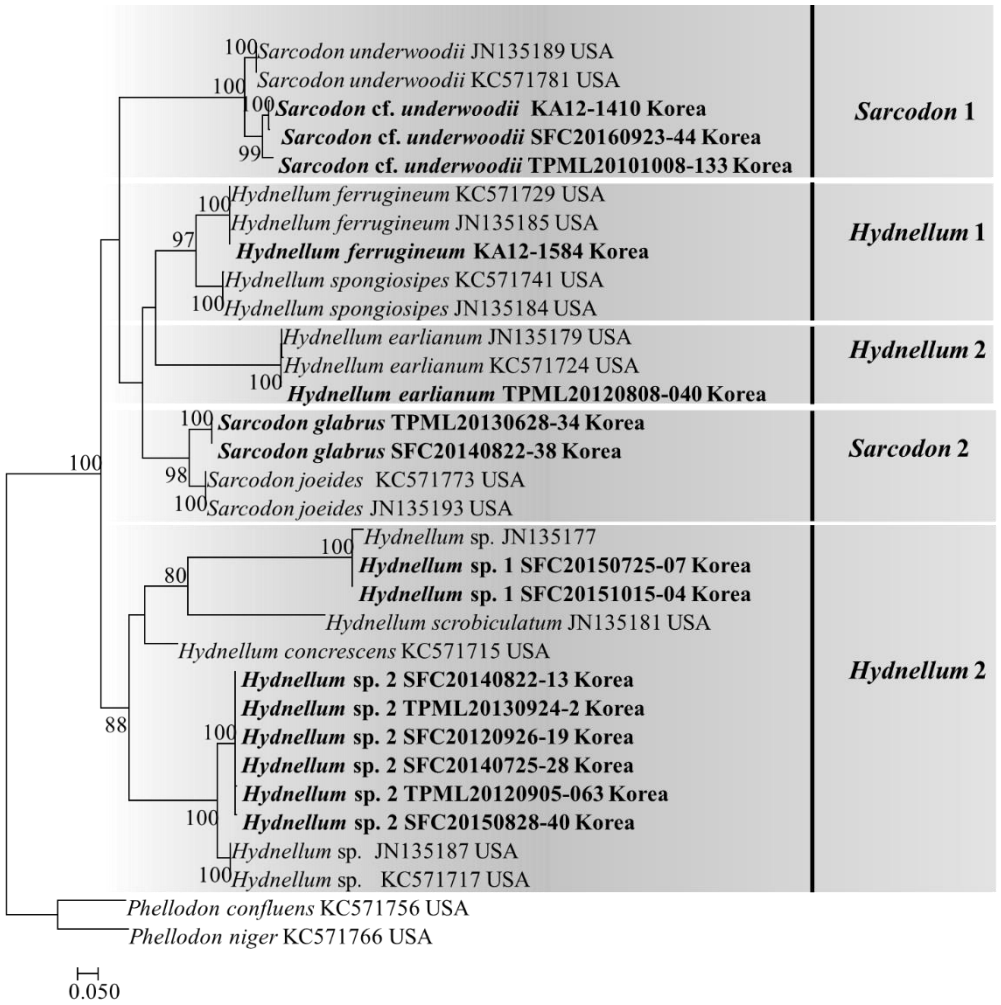


**Figure 3.** Phylogram generated from Maximum Likelihood (RAxML) analysis based on ITS sequence data of group G1 (*Hydnellum* and *Sarcodon*). ML bootstrap support values greater than 70% are indicated above or below the nodes and the Korean species are in bold.

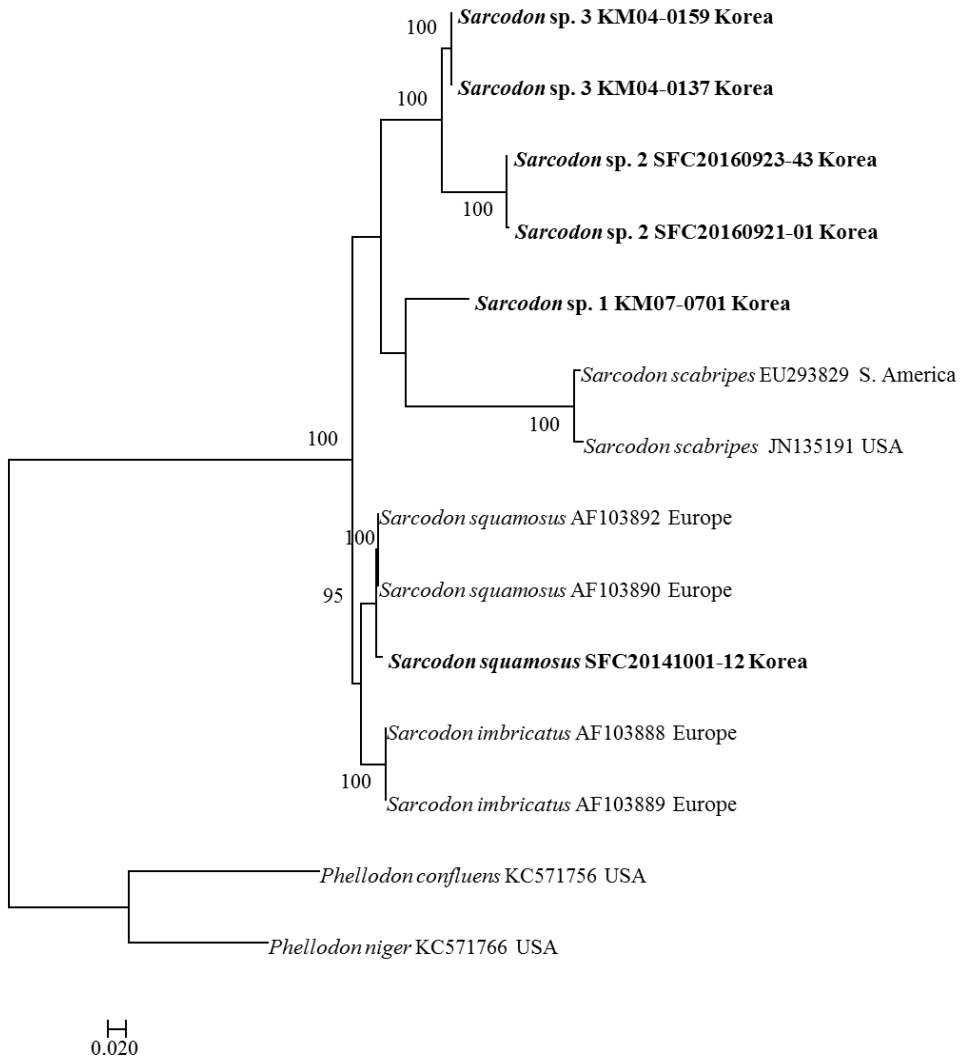


**Figure 4.** Phylogram generated from Maximum Likelihood (RAxML) analysis based on ITS sequence data of group G2 (*Sarcodon*). ML bootstrap support values greater than 70% are indicated above or below the nodes and the Korean species are in bold.

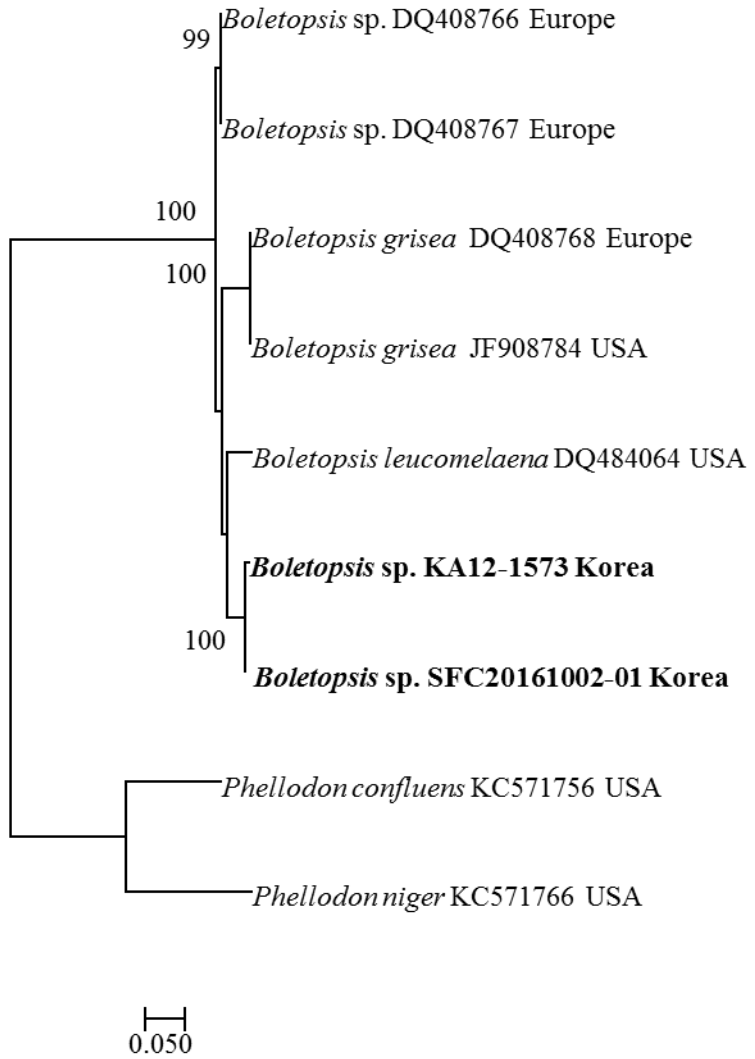


**Figure 5.** Phylogram generated from Maximum Likelihood (RAxML) analysis based on ITS sequence data of group G3 (*Boletopsis*). ML bootstrap support values greater than 70% are indicated above or below the nodes and the Korean species are in bold.

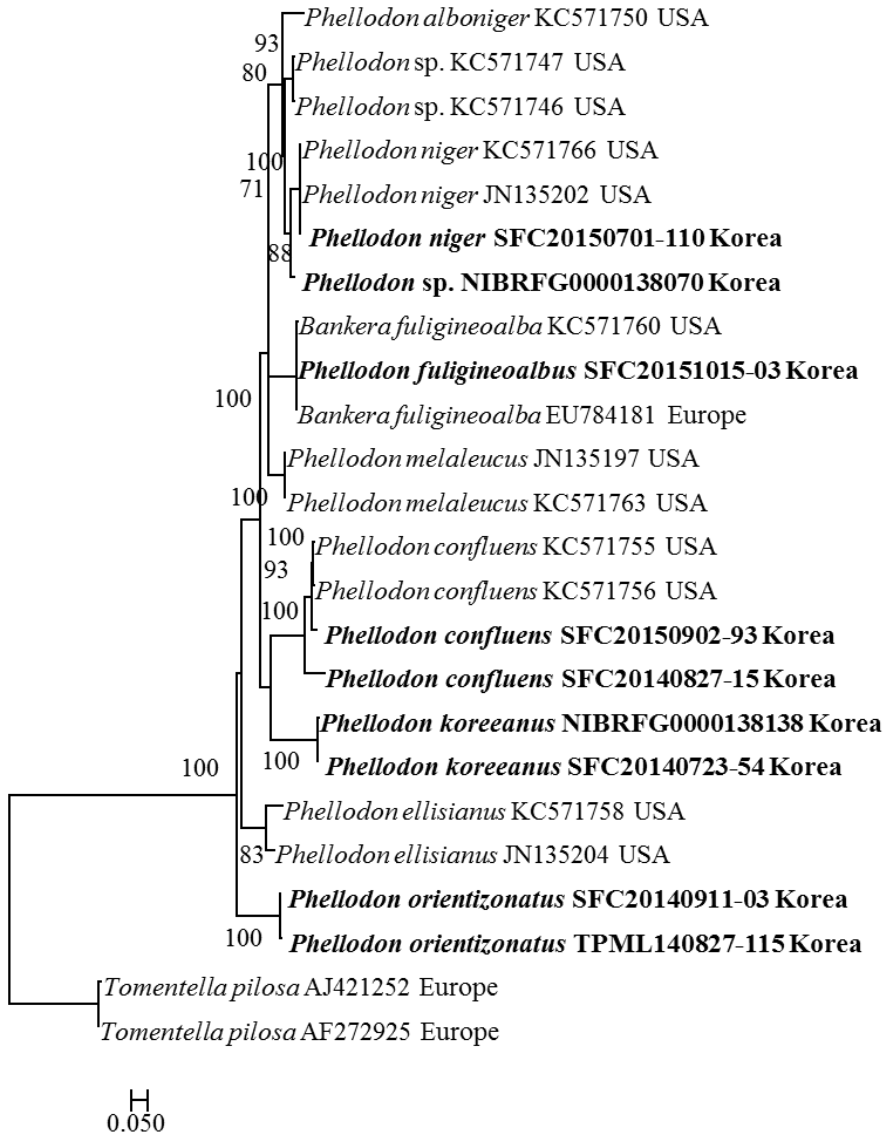


**Figure 6.** Phylogram generated from Maximum Likelihood (RAxML) analysis based on ITS sequence data of group G4 *(Phellodon*). ML bootstrap support values greater than 70% are indicated above or below the nodes and the Korean species are in bold.

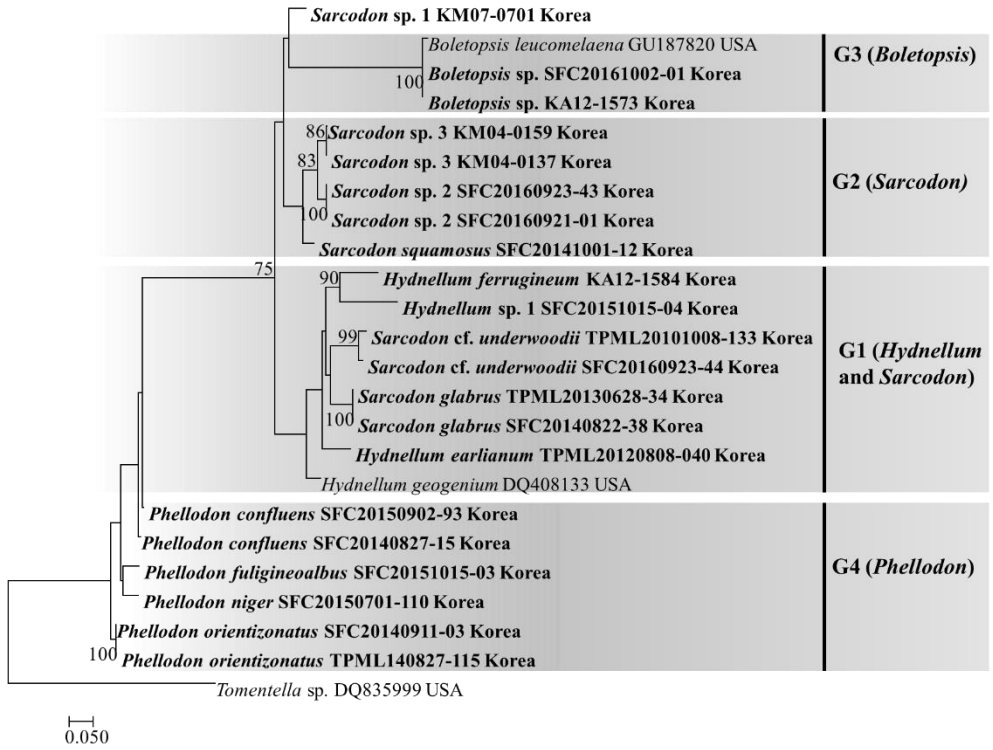


**Figure 7.** Phylogram generated from Maximum Likelihood (RAxML) analysis based on *rpb*2 sequences data of four genera of family Bankeraceae. ML bootstrap support values greater than 70% are indicated above or below the nodes and the Korean species are in bold.

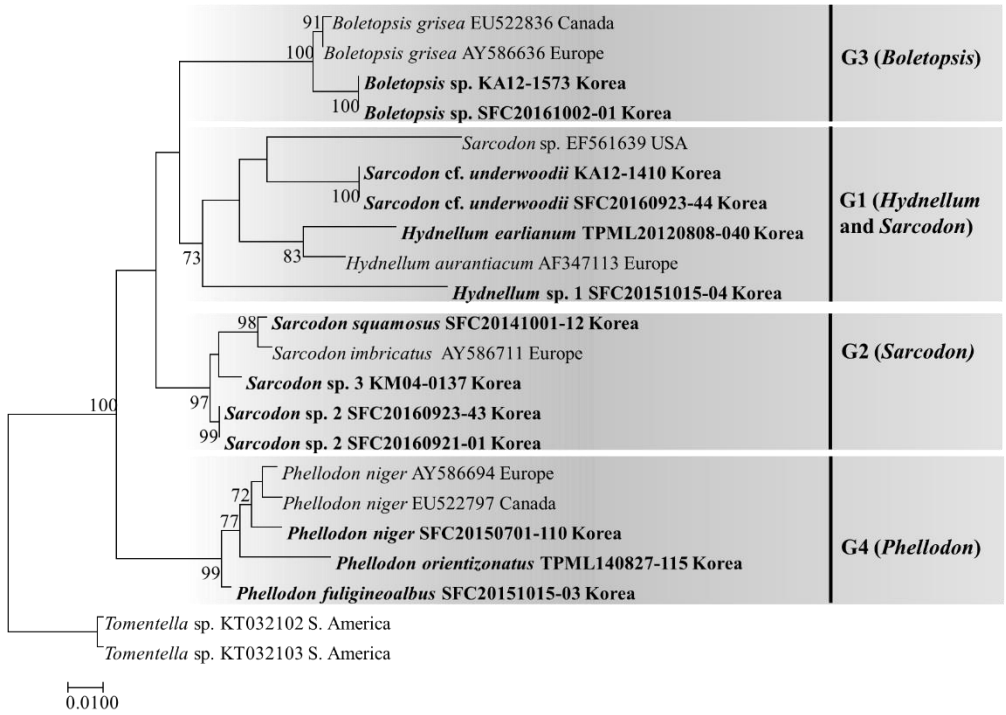


**Figure 8.** Phylogram generated from Maximum Likelihood (RAxML) analysis based on LSU sequence data of four genera of family Bankeraceae. ML bootstrap support values greater than 70% are indicated above or below the nodes and the Korean species are in bold.

of genus *Boletopsis*, which was monophyletic with solid bootstrap value of 100% (Fig. 5). *Boletopsis* appeared to be closely related to *Sarcodon* (G2). The maximum interspecific dissimilarity was calculated between Korean *Boletopsis* sp. and reference *Boletopsis leucomelaena* and the resulting value was approximately 5%. Group (G4) of genus *Phellodon* was monophyletic and appeared as an outgroup to the other genera with a moderate bootstrap value of 66% (Fig. 6). The maximum interspecific dissimilarity between group of *Phellodon* sp. and group of *Phellodon niger* including *Phellodon* sp. from Korea was in the range of 3.7 – 6.5%. The dissimilarity between sister clades of *Phellodon koreeanus* and a group of *Phellodon confluens* was accounted for being around 20%.

Although, phylogenetic trees of *rpb*2 and LSU regions contained only a fraction of all specimens, the resulting groups resolved with similar pattern as ITS tree (Fig. 7, Fig. 8). Genera *Boletopsis*, *Sarcodon* (G2) and *Phellodon* appeared as monophyletic groups, while group G4 again consisted of intermixed species of both *Hydnellum* and *Sarcodon*. However, there were certain deviations not observed in the ITS tree: in *rpb*2, Korean specimen *Sarcodon* sp. 1 appeared as a taxa related to genus *Boletopsis*, rather than *Sarcodon* (G2) (Fig. 7); in LSU tree, genus *Boletopsis* formed appeared to be the most distant to other genera rather than *Phellodon* (Fig. 8). The maximum interspecific dissimilarity was not calculated for these regions.

## 3.3. Taxonomy

### ***Boletopsis*** Fayod

Pileus annual, tissue fleshy to hard and fibrous, basidiomata is usually central, stipitate, round can be concrescent (Stalpers 1993). Color ranges from grey, brown to black-brown. Hymenophore is poroid, covers the beneath of the pileus. Pores color ranges from cream to different shades of grey. Spore print is white to cream colored. Hyphae is monomitic, clamp connections present. Cystidia absent. Basidia clavate, with basal clamps, four sterigmata present. Spores warted, lobed (Stalpers 1993). Phylogenetic analysis of ITS region positioned genus *Boletopsis* to be close to genus to be close to genus *Hydnellum* (Larsson *et al.* 2004).

***Boletopsis* sp.**

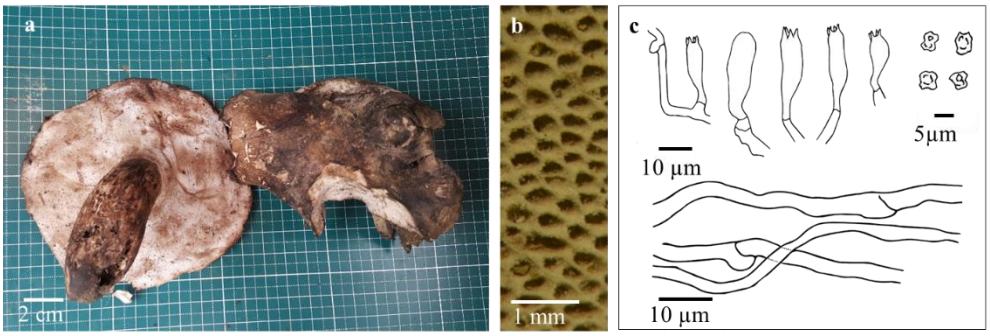


**Figure 9.** *Boletopsis* sp. a fruting bodies. b pores of fruiting body. c. microscopic structures; basidia, basidiospores, generative hyphae.

*Basidiocarp* single, fleshy to tough, positioned centrally, divided into pileus and stipe;

pileus is round, up to 6.5 cm broad, light brown to brown when fresh, brown to dark brown when dry. Context is up to 0.4 cm, brittle when dry. *Hymenophore* is covered with pores, 1 – 3 per mm, white to cream, grey colored. *Stipe* up to 7 x 1 cm, individual, bulbous base, brown to dark brown when fresh, dark brown to black-brown upon drying. *Hyphae* uninflated and simply septated, clamps connections present; pileus trama hyphae up to 9 µm wide; trama hyphae up to 3.5 µm wide; stipe hyphae up to 13 µm wide. *Basidia* 17.0 – 27.0 × 5.0 – 8.5 µm, clavate, 4 spored, sterigmata up to 5 µm long. *Basidiospores* ovoid, ornamentated echinulate, small spinules, 4.0–5.5 × 4.0–5.0 µm. L = 5.1 µm, W = 4.3 µm, Q = 1.19 (n=20/1). *Chemical reaction* is absent in KOH, water and Melzer's reagent.

*Habitat*: On soil in broadleaved forest, near *Quercus* species.

*Material examined*: KOREA, Chubu-myeon, Geumsan-gun, Chungcheongnam-do, on soil, 29 August 2012, S. K. Han (KA12-1573, holotype; GeneBank ITS: XXXX). KOREA, Dogye-eup, Samcheok-si, Gangwon-do, on soil, 2 October 2016, N. K. Kim, Y. W. Lim (SFC201610021-01, paratype; GeneBank ITS: XXXX).

*Notes*: The examined specimen has a resemblance of *Boletopsis leucomelaena* (Pers.) Fayod and shares morphological characters. However, ITS analysis positioned this species quite distant from *B. leucomelaena* sequence available on GeneBank. Our sample closely matched the sequence of *Boletopsis* sp. uploaded earlier by Watling and Milne (Unpublished).

## ***Hydnellum*** Karst

Basidiocarp annual, from soft to woody, centrally or excentrically stipitate, can be concrescent, tomentose; possible branching from a common base (Stalpers 1993). The color variation includes different shades of brown, yellow, orange, blue, red and orange. Stipe can be tomentose, usually of the same color as pileus. Spines color varies from brown to slightly purple. Spores create brown print. Hypha uninflated; clamps may or may not be present. Cystidia are not present. Basidia with four sterigmata, basal clamps may or may not be present. Spores have irregular shape, subglobose to ellipsoid, ornamentation is present (Stalpers 1993). The analysis of the ITS region in the latest work dedicated to studying family Bankeraceae showed that species of genus *Hydnellum* are intermixed with species of genus *Sarcodon*, and form a paraphyletic group (Baird *et al.* 2013).

**KEY TO KOREAN SPECIES OF *HYDNELLUM***

1. Basidiomata is brightly colored, has shades of orange·····*H. earlianum*

1. Basidiomata is brown to dark brown·····2

2. Basidiomata has pronounced margins·····*Hydnellum* sp. 2

2. Basidiomata does not have pronounced margins·····3

3. Spines are up to 0.2 cm long·····*Hydnellum* sp. 1

3. Spines are up to 0.5 cm long·····*H. ferrugineum*

**1. *Hydnellum earlianum*** Banker

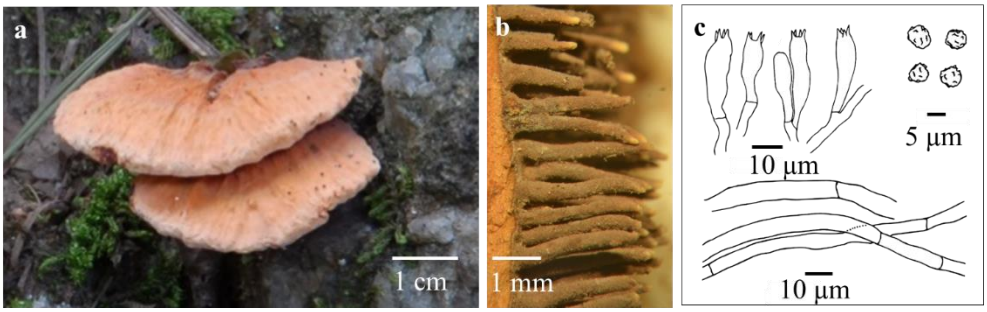


**Figure 10.** *Hydnellum earlianum*. a fruting bodies in the field. b spines of fruiting body. c microscopic structures; basidia, basidiospores, generative hyphae.

Basidiomata single to gregarious, can be concrescent. Pileus up to 3 cm broad, planar to depressed, tomentose, radially rugulose to rugose. Colors are brown to orange when fresh, brown to dark brown, and orange when dry. Context up to 0.2 cm thick, zonate. Spines up to 0.2 cm long, brown when fresh, dark brown when dry. Stipe up to 2.0 x 1.5 cm, bulbous at base, eccentric, shares color scheme with the pileus. Hyphae uninflated and simple septated; pileus trama hyphae up to 5.7 µm wide; spine trama hyphae up to 4.5 µm wide; stipe hyphae up to 5 µm wide. Basidia 22.0 – 35.0 × 5.0 – 8.5 µm, clavate, unclamped, 4 spored, sterigmata up to 4.5 µm long. Basidiospores subglobose, ovoid, ornamentation tuberculate, 4.0 – 7.0 × 4.0 – 6.0 µm.

L = 5.6 µm, W = 6 µm, Q = 1.12 (n=20/1). Chemical reaction of tissue is green in KOH, no reaction with water or Meltzer's reagent.

*Habitat*: On soil in broadleaved forest, near *Quercus* species.

*Material examined*: KOREA, Daehang-myeon, Gimcheon-si, Gyeongsangbuk-do, on soil, 8 August 2012, N. K. Kim, J. K. Lee (TPML20120808-040, holotype; GeneBank ITS: XXXX).

*Notes*: The color scheme is a feature allowing to differentiate this species from others of the genus. Pileus is yellow, when young, turning orange upon maturation.

**2. *Hydnellum ferrugineum*** Karst, Baird *et al*.

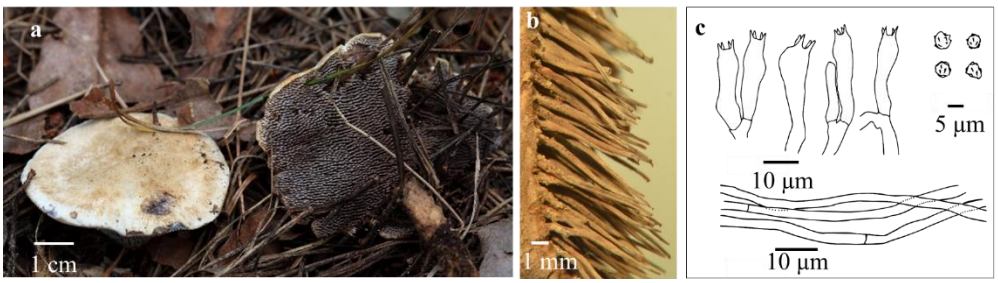


**Figure 11.** *Hydnellum ferrugineum.* a fruting bodies in the field. b spines of fruiting body. c microscopic structures; basidia, basidiospores, generative hyphae.

*Basidiomata* single to gregarious, not concrescent. *Pileus* up to 4 cm broad, planar to depressed, surface is clearly zonate. Colors are shades of grey and brown when fresh, and grey to dark brown when dry. Context up to 0.5 cm thick. *Spines* up to 0.5 cm

long, brown when fresh, dark brown to black when dry. *Stipe* up to 3.5 x 1.2 cm, central to eccentric, not bulbous base. *Hyphae* uninflated and simple septated; pileus trama hyphae up to 5 µm wide; spine trama hyphae up to 4 µm wide; stipe hyphae up to 6 µm wide. *Basidia* 28.0 – 35.0 × 5.0 – 7.0 µm, clavate, unclamped, 4 spored, sterigmata up to 4 µm long. *Basidiospores* subglobose, ovoid, ornamentation tuberculate, 5.0 – 6.0 × 4.0 – 6.0 µm. L = 5.6 µm, W = 4.7 µm, Q = 1.17 (n=20/1). *Chemical reaction* of tissue is olive in KOH, no reaction with water or Meltzer's reagent.

*Habitat*: On soil in broadleaved forest, near *Quercus* species.

*Material examined*: KOREA, Chubu-myeon, Geumsan-gun, Chungcheongnam-do, on soil, 26 September 2012, S. K. Han (KA12-1584, holotype; GeneBank ITS: XXXX).

*Notes*: According to the latest study, it is assumed, that *H. ferrugineum* appears only in conifer forests (Baird *et al.* 2013). The examined sample also featured a distinct zonation on the pileus with brown colored center, and light grey colored margins.

**3. *Hydnellum* sp. 1**

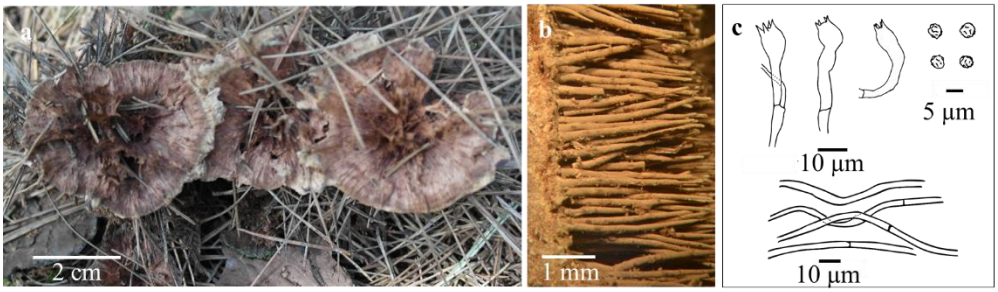


**Figure 12.** *Hydnellum* sp. 1. a fruting bodies in the field. b spines of fruiting body. c microscopic structures; basidia, basidiospores, generative hyphae.

*Basidiomata* single to gregarious, can be concrescent. *Pileus* up to 3.5 cm broad, planar to depressed, tomentose, radially rugulose to rugose. Colors are cream, brown-red brown to dark brown when fresh, brown to dark brown when dry. Context up to 0.15 cm thick, zonate. *Spines* up to 0.2 cm long, cream, brown to red-brown when fresh, brown when dry. *Stipe* up to 4 x 2 cm, usually central, concolorous with pileus,can be fused. *Hyphae* uninflated, pileus trama hyphae up to 5.7 µm wide; spine trama hyphae up to 4 µm wide; stipe hyphae up to 5 µm wide. *Basidia* 21 – 34 × 6 – 8 µm, clavate, unclamped, 4 spored, sterigmata up to 4.5 µm long. up *Basidiospores* subglobose, ovoid, ornamentation tuberculate, 4.3 – 6.0 × 3.0 – 5.0 µm. L = 5 µm, W = 4 µm, Q = 1.16 (n=20/1). *Chemical reaction* of tissue is olive-brown in KOH, no reaction with water or Meltzer's reagent.

*Habitat*: On soil in broadleaved forest, near *Quercus* species.

*Material examined*: KOREA, Cheongha-myeon, Buk-gu, Pohang-si, Gyeongsangbuk-do, on soil, 15 October 2015, N. K. Kim, Y. W. Lim (SFC20151015-

04, holotype; GeneBank ITS: XXXX). KOREA, Sokrisan-myeon, Boeun-gun, Chungcheongbuk-do, on soil, 25 July 2015, H. Lee, Y. W. Lim (SFC20150725-07, paratype; GeneBank ITS: XXXX).

*Notes*: As indicated by Baird *et al*., the species shares morphology with *Hydnellum concrescens* (Pers.) and extends from its clade (Baird *et al*. 2013). However, based on analysis of the ITS region our species appeared in the clade of *Hydnellum* sp. 1 with bootstrap value of 100. Morphological characters are ambiguous and molecular analysis is required for precise identification.

**4. *Hydnellum* sp. 2**

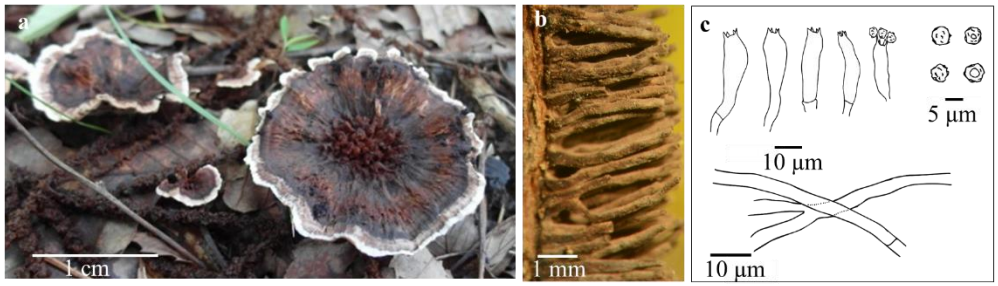


**Figure 13**. *Hydnellum* sp. 2. a fruiting bodies in the field. b spines of fruiting body. c microscopic structures; basidia, basidiospores, generative hyphae.

*Basidiomata* single to gregarious, can be concrescent. *Pileus* up to 3.5 cm broad, planar to depressed at disc, tomentose, zonate, the center and margins can be easily separated. Colors are cream, brown-red brown to dark brown when fresh, dark beige, brown to dark brown when dry. Context up to 0.4 cm thick, zonate. *Spines* up to 0.2 cm long, cream, brown to red-brown when fresh, brown when dry. *Stipe* up to 3.5 x

0.5 cm, usually central to sub-centric, concolorous with pileus, can be fused. *Hyphae* uninflated, pileus trama hyphae up to 6 µm wide; spine trama hyphae up to 4 µm wide; stipe hyphae up to 6 µm wide. *Basidia* 28 – 40 × 6 – 9 µm, clavate, unclamped, 4 spored, sterigmata up to 3.3 µm long. *Basidiospores* subglobose, ovoid, ornamentation tuberculate, 4.0 – 6.0 × 4.0 – 6.0 µm. L = 5.0 µm, W = 4.6 µm, Q = 1.1 (n=20/1). *Chemical reaction* of tissue is slightly brown in KOH, no reaction with water or Meltzer's reagent.

*Habitat*: On soil in broadleaved forest, near *Quercus* species.

*Material examined*: KOREA, Seolcheon-myeon, Muju-gun, Jeollabuk-do, on soil, 5 September 2012, N. K. Kim, J. K. Lee (TPML20120905-063, holotype; GeneBank ITS: XXXX). KOREA, Seolcheon-myeon, Muju-gun, Jeollabuk-do, on soil, 28 August 2015, H. Lee, Y. W. Lim (SFC20150828-40, paratype; GeneBank ITS: XXXX). KOREA, Girin-myeon, Inje-gun, Gangwon-do, on soil, 22 August 2015, H. J. Cho, Y. W. Lim (SFC20140822-13, paratype; GeneBank ITS: XXXX). KOREA, Donghyang-myeon, Jinan-gun, Jeollabuk-do, on soil, 25 July 2014, J. Y. Park, Y. W. Lim (SFC20140725-28, paratype; GeneBank ITS: XXXX). KOREA, Jeongan-myeon, Gongju-si, Chungcheongnam-do, on soil, 26 September 2012, J. Y. Park, Y. W. Lim (SFC20120926-19, paratype; GeneBank ITS: XXXX). KOREA, Geumgangsong-myeon, Uljin-gun, Gyeongsangbuk-do, on soil, 24 September 2013, N. K. Kim, J. K. Lee (TPML20130924-02, paratype; GeneBank ITS: XXXX).

*Notes*: This species together with *Hydnellum* sp. 1 have morphological

characters shared with *Hydnellum concrescens* as pointed out by Baird *et al*., and *Hydnellum* sp. 2 also appears in the group of *Hydnellum concrescens* in the phylogenetic tree. Morphological characters are ambiguous and molecular analysis is required for precise identification of the species.

## ***Phellodon*** P. Karst.

Genus *Phellodon* belongs to hydnoid fungi of family Bankeraceae. Pileus of the species is usually tomentose, zonated, rugulose to pitted or scrobiculate, contains shades of brown or black colors (Baird *et al*. 2013, Karst 1881). It is extremely similar in macro-morphology to genus *Hydnellum*. The unique feature of this genus is spores having fine spines, which are not easily seen under low power microscope, and can be used as a feature to differentiate from *Hydnellum* species; clamp connections are usually absent (Karst 1881). Based on the analysis of the ITS region genus *Phellodon* is a monophyletic group within family Bankeraceae (Baird *et al*. 2013).

**KEY TO KOREAN SPECIES OF *PHELLODON***

1. Basidiocarp glabrous, not zonated·····*P. fuligineoalbus*

1. Basidiocarp is zonated·····2

2. Basidiocarp is light brown to brown·····3

2. Basidiocarp is brown to dark brown to black·····4

3. Basidiocarp has pronounced margins·····4

3. Basidiocarp does not have pronounced margins·····5

4. Hymenophoral surface is white to grey·····*P. confluens*

4. Hymenophoral surface is tan to light brown·····5

5. Basidiocarps can be fused together·····*P. koreeanus*

5. Basidiocarps can be separated on individual·····6

6. Spines are up to 0.1 cm·····*P. orientizonatus*

6. Spines are up to 0.3 cm·····*P. niger*

**1. *Phellodon confluens*** (Pers.) Pouzar

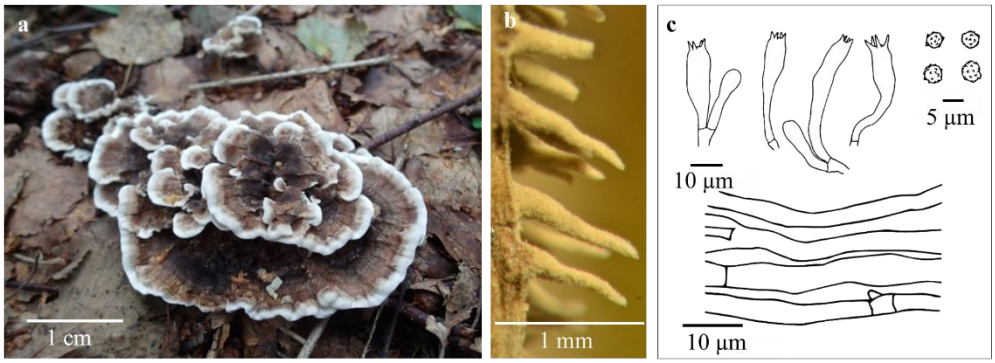


**Figure 14.** *Phellodon confluens*. a fruting bodies in the field. b spines of fruiting body. c microscopic structures; basidia, basidiospores, generative hyphae.

*Basidiocarps* single to gregarious, concrescent; pileus up to 3.5 cm broad, can be fused, depressed at disc, with planar margins; zonate, brown to dark brown, black, margins are white to grey when fresh, brown to dark brown, black upon drying; context is thin up to 0.3 cm, brittle when dry. *Spines* up to 0.2 cm long, decurrent, white to grey when fresh, grey to dark grey upon drying. *Stipe* up to 3 × 1 cm, individual sometimes fused, brown to dark brown when fresh, brown to dark brown upon drying. *Hyphae* uninflated and simply septated; pileus trama hyphae up to 5.6 µm wide; spine trama hyphae up to 4.5 µm wide; stipe hyphae up to 6.5 µm wide. *Basidia* 24.0 – 38.0 × 5.5 – 7.0 µm, clavate, unclamped, 4 spored, sterigmata up to 4.5 µm long. *Basidiospores* ovoid, ornamentated echinulate, small spinules, 4.0 – 5.0 × 4.0 – 4.5 µm. L = 4.4 µm, W = 4.2 µm, Q = 1.04 (n=20/1). *Chemical reaction* of tissues is brown in KOH, no reaction with water and Melzer's reagent.

*Habitat*: On soil in broadleaved forest, near *Quercus* species.

*Material examined*: KOREA, Inje-eup, Inje-gun, Gangwon-do, on soil, 2 September 2015, H. Lee, Y. W. Lim (SFC20150902-93, holotype; GeneBank ITS: XXXX). KOREA, Beonam-myeon, Jangsu-gun, Jeollabuk-do, on soil, 27 August 2014, J. Y. Park, Y. W. Lim (SFC20140827-15, holotype; GeneBank ITS: XXXX).

*Notes*: Morphologically similar to *P. niger* (Fries) Karst, *Phellodon confluens* can be distinguished by the absence of black context, which is present in *P. niger* (Baird *et al*. 2013). Basidiocap is lightly colored (light brown, tan). Usually multiple pilei are fused together, making it difficult to separate on individual parts. Phylogenetic analysis showed, that our samples belong to *P. confluens* when compared to a sequence from GeneBank, with supportive value of 100.

**2. *Phellodon fuligineoalbus*** (Schmidt) Baird

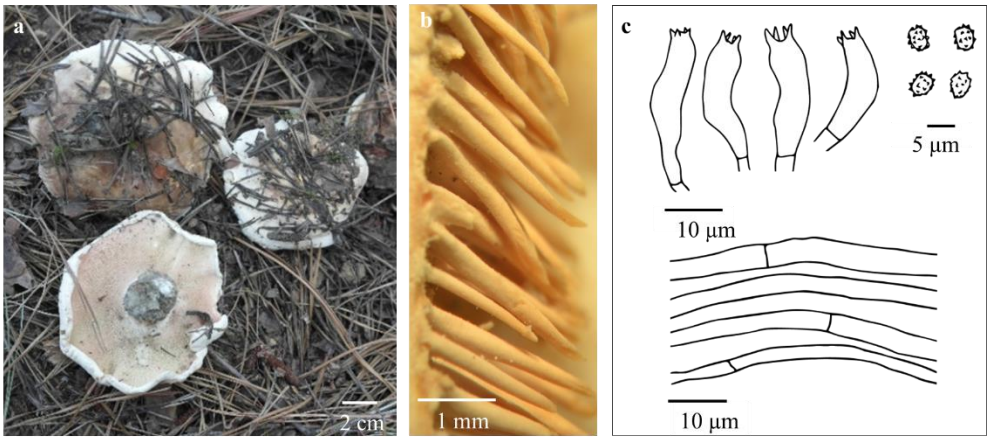


**Figure 15.** *Phellodon fuligineoalbus*. a fruting bodies in the field. b spines of fruiting body. c microscopic structures; basidia, basidiospores, generative hyphae.

*Basidiocarps* glabrous, single to gregarious up to 7.5 cm broad, convex to planar, depressed at disc; cream, white, pink, light brown when fresh, cream, white, light brown upon drying; context up to 0.5 cm, brittle when dry. *Spines* up to 0.4 cm long, cream, white to pink when fresh, cream or pink upon drying. *Stipe* up to 4 × 2 cm, central to sub-central, same color scheme with that of pileus. *Pileus trama hyphae* up to 4 µm wide; spine trama hyphae up to 3.5 µm wide; stipe hyphae up to 7 µm wide. *Basidia* 24.0 – 31.0 × 5.0 – 6.0 µm, clavate, unclamped, 4 spored, sterigmata up to 3.6 µm long. *Basidiospores* ovoid, ornamentated echinulate, small spinules, 4.0 – 5.0 × 3.5 – 4.0 µm. L = 4.4 µm, W = 3.6 µm, Q = 1.22 (n=20/1). *Chemical reaction* is absent in KOH, water and Melzer's reagent.

*Habitat*: On soil in broadleaved forest, near *Quercus* species.

*Material examined*: KOREA, Cheongha-myeon, Buk-gu, Pohang-si, Gyeongsangbuk-do, on soil, 25 July 2015, N. K. Kim, Y. W. Lim (SFC20151015-03, holotype; GeneBank ITS: XXXX).

*Notes*: The species was initially named as *Bankera fuligineoalba* (Schmidt) and its morphology resembles that of species of genus Bankera (Baird *et al*. 2013). Analyzed sample is covered in conifer needles. However, analysis of ITS region showed that this species belongs to *Phellodon*, rather than Bankera. The results of molecular analysis of this study matches with earlier work performed by Baird *et al*.

**3. *Phellodon* sp.**

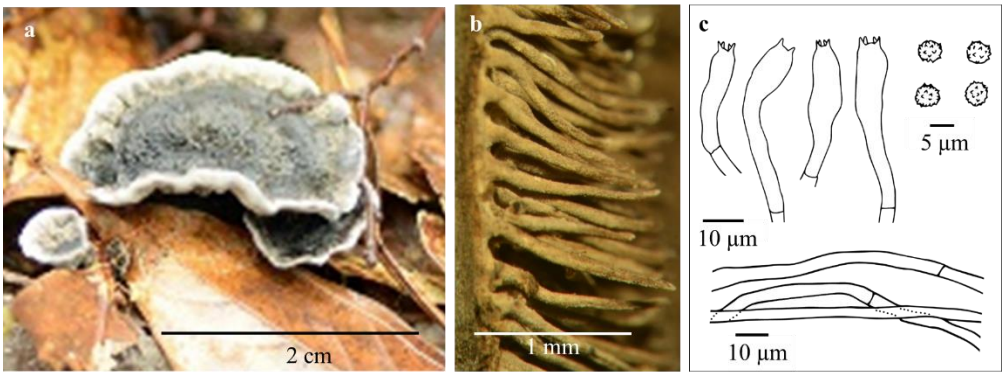


**Figure 16.** *Phellodon* sp. a fruting bodies. b spines of fruiting body. c microscopic structures; basidia, basidiospores, generative hyphae.

*Basidiocarps* single to gregarious, can be concrescent up to 2.5 cm broad, planar to depressed, becoming tomentose; brown to dark brown, black when fresh, dark brown to black upon drying; context up to 0.3 cm. *Spines* up to 0.1 m long, brown when fresh, brown to dark brown upon drying. *Stipe* up to 2 × 1 cm, central to sub-central, same color scheme with that of pileus. *Pileus trama hyphae* up to 6.0 µm wide; spine trama hyphae up to 4.5 µm wide; stipe hyphae up to 8 µm wide. *Basidia* 25.0 – 38.0 × 5.0 – 8.0 µm, clavate, unclamped, 4 spored, sterigmata up to 4.0 µm long. *Basidiospores* ovoid, ornamentated echinulate, small spinules, 4.5 – 6.0 × 4.0 – 5.5 µm. L = 5.0 µm, W = 4.8 µm, Q = 1.05 (n=20/1). *Chemical reaction* of tissue is olive in KOH, no reaction with water and Melzer's reagent.

*Habitat*: On soil in broadleaved forest, near *Quercus* species.

*Material examined*: KOREA, Seohong-dong, Seogwipo-si, Jeju-do, on soil,

1 July 2015, P. Y. Ko. (NIBRFG0000138070, holotype; GeneBank ITS: XXXX).

*Notes*: Morphology is very close to *P. niger*, probably it is a sister species. Results of ITS analysis showed that our sample is positioned close to the *Phellodon niger* group.

**4. *Phellodon niger*** Karst

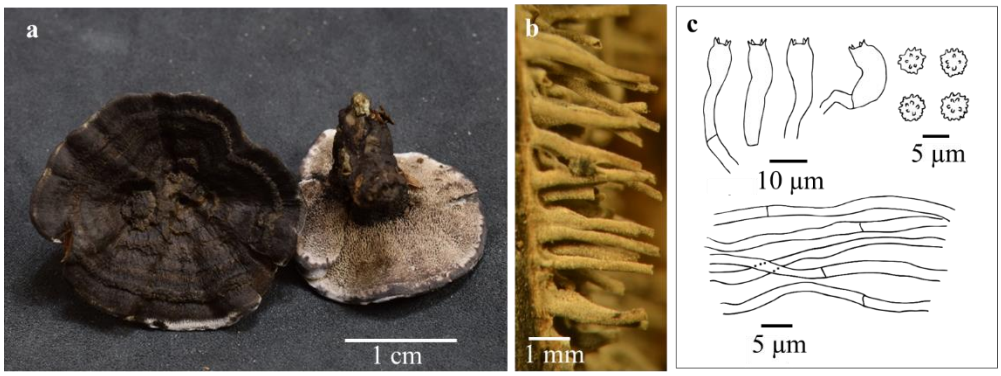


**Figure 17.** *Phellodon niger*. a fruting bodies. b spines of fruiting body. c microscopic structures; basidia, basidiospores, generative hyphae.

*Basidiocarps* single to gregarious, can be concrescent up to 4 cm broad, planar to depressed, becoming tomentose; brown to dark brown, black when fresh, dark brown to black upon drying; context up to 0.2 cm. *Spines* up to 0.3 cm long, brown when fresh, brown to dark brown upon drying. *Stipe* up to 3 × 1 cm, central to sub-central, color is brown. *Pileus trama hyphae* up to 5.5 µm wide; spine trama hyphae up to 5 µm wide; stipe hyphae up to 7 µm wide. *Basidia* 21.0 – 30.0 x 5 – 8 µm, clavate, unclamped, 4 spored, sterigmata up to 2.5 µm long. *Basidiospores* ovoid,

ornamentated echinulate, small spinules, 5.0 – 6.0 × 4.0 – 5.5 µm. L = 5.6 µm, W = 5.5 µm, Q = 1.01 (n=20/1). *Chemical reaction* of tissue is olive in KOH, no reaction with water and Melzer's reagent.

*Habitat*: On soil in broadleaved forest, near *Quercus* species.

*Material examined*: KOREA, Jocheon-eup, Jeju-si, Jeju-do, on soil, 15 July 2014, N. K. Kim, H. Lee, Y. W. Lim (SFC20150701-110, holotype; GeneBank ITS: XXXX).

*Notes*: In terms of morphology, our sample has a unique feature of *Phellodon niger* by having dark, almost black basidiocarp and brown coloured hymenophoral surface. Results of ITS analysis showed that our sample is positioned close to the sample from GeneBank (USA reference) with supporting bootstrap value of 100%

**5. *Phellodon koreeanus*** V. Li, N. K. Kim & Y. W. Lim, **sp. nov.**

*Index Fungorum number*: XXXX, *Faces of fungi number*: XXXX.

*Etymology*: referring to a more slender spore relative to other *Phellodon* species.

*Holotype*: SFC20140723-54.

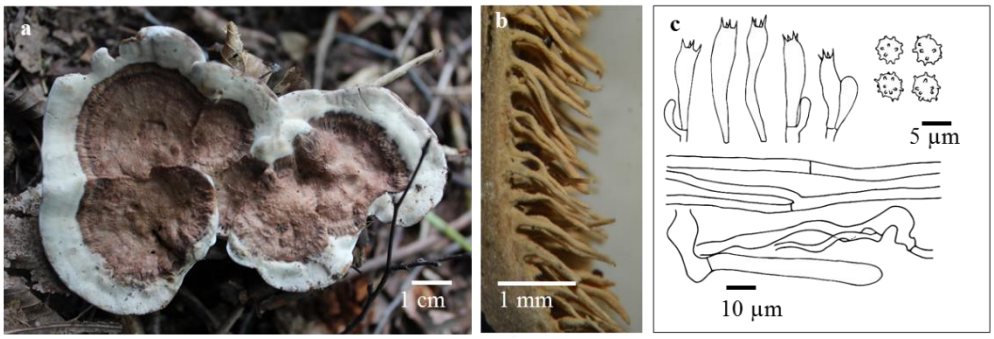


**Figure 18.** *Phellodon koreeanus*. a fruting bodies in the field. b spines of fruiting body. c microscopic structures; basidia, basidiospores, generative hyphae.

*Basidiocarps* single to gregarious or fused, divided into pileus and stipe; pileus up to 3 cm broad, larger when fused, depressed at disc, with planar margins; pileal surface zonate, clearly separated from the middle of the cap, light brown on the edges to brown in the center upon drying; contexts is thin up to 0.4 cm, brittle when dry. *Hymenophore* spines up to 0.1 cm long, decurrent, ivory to light brown when fresh, beige to brown upon drying. *Stipe* up to 1.0 × 1.5 cm, individual but sometimes fused, brown to dark brown when fresh, brown to dark brown upon drying. *Hyphae* uninflated and simply septated; pileus trama hyphae up to 6.5 µm wide; spine trama hyphae up to 4.4 µm wide; stipe hyphae up to 6 µm wide. *Basidia* 27 – 35 × 5 – 7

μm, clavate, no clamps, 4 spored, sterigmata up to 4 μm long. *Basidiospores* ovoid, ornamentated echinulate, small spinules, 4.0–6.0 × 4.0–5.5 μm. L = 5 μm, W = 4.7 μm, Q = 1.05 (n=20/1). *Chemical reaction* of tissues is slightly brown in KOH, no reaction with water and Melzer's reagent.

*Habitat*: On soil in broadleaved forest, near *Quercus* species.

*Material examined*: KOREA, Jeollabuk-do, Jinan-gun, Mt. Unjang, on soil, 23 July 2014, J. Y. Park, Y. W. Lim (SFC20140723-54, holotype; GeneBank ITS: XXXX). KOREA, Jeju-do, Seogwipo-si, Hogeun-dong, on soil, P. Y. Ko (NIBRFG0000138138, paratype; GeneBank ITS: XXXX).

*Notes*: This species shares similar morphology with *P. tomentosus*, such as being able to fuse several basidiocarps and grow together, and also having a bicolor nature of basidiocarp. The species does not possess unique features allowing it to be identified as a separate species based on morphology. However, molecular analysis using ITS sequences showed, that *Phellodon koreensis* is clearly separated with *P. tomentosus* and formed distinct clade.

**6. *Phellodon orientizonatus*** V. Li, N. K. Kim & Y. W. Lim, **sp. nov.**

*Index Fungorum number*: XXXX, *Faces of fungi number*: XXXX.

*Etymology*: referring to a more slender spore relative to other *Phellodon* species.

*Holotype*: SFC20140911-03.

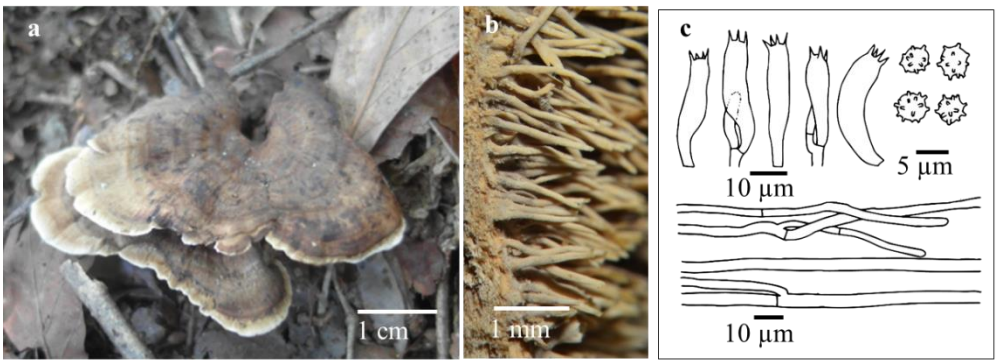


**Figure 19.** *Phellodon orientizonatus*. a fruting bodies in the field. b spines of fruiting body. c microscopic structures; basidia, basidiospores, generative hyphae.

*Basidiocarps* single to gregarious, divided into pileus and stipe; pileus up to 3.5 cm broad, depressed at disc, with planar margins; zonate, light brown to brown when fresh, light brown, brown and dark brown upon drying; context is thin up to 0.4 cm, brittle when dry. *Hymenophore* up to 0.1 cm long, decurrent, cream to brown when fresh, orange to light brown upon drying. *Stipe* up to 0.5 × 1.5 cm, individual sometimes fused, brown to dark brown when fresh, brown to dark brown upon drying. *Hyphae* uninflated and simply septated; pileus trama hyphae up to 6.5 µm wide; spine trama hyphae up to 4.5 µm wide; stipe hyphae up to 6 µm wide. *Basidia* 25 – 34 ×

5.5 – 7.5 µm, clavate, 4 spored, sterigmata up to 4 µm long. *Basidiospores* ovoid, ornamentated echinulate, small spinules, 4.0–5.5 × 4.0–5.0 µm. L = 4.6 µm, W = 4.3 µm, Q = 1.05 (n=20/1). *Chemical reaction* of tissues is slightly brown in KOH, no reaction with water and Melzer's reagent.

*Habitat*: On soil in broadleaved forest, near *Quercus* species.

*Material examined*: KOREA, Jeollabuk-do, Jinan-gun, Mt. Cheonban, on soil, 11 September 2014, J. Y. Park, Y. W. Lim (SFC20140911-03, holotype; GeneBank ITS: XXXX). KOREA, Gyeongsangbuk-do, Gimcheon-si, Mt. Hwangak, on soil, 27 August 2014, N. K. Kim (TPML20140827-115, paratype; GeneBank ITS: XXXX).

*Notes*: This species is similar in morphology to *Hydnellum caeruleum*, a species from other genus of family Bankeraceae, however, it does not have a bluish tint on the basidiocarps. Also, *Phellodon orientizonatus* is generally smaller in its dimensions, having pileus twice as smaller than *H. caeruleum*, in addition, basidia of *Phellodon orientizonatus* are also smaller in size. In terms of molecular similarity, *Phellodon orientizonatus* is relatively close to *P. melaleucus*. Morphological characters are ambiguous and insufficient to draw a solid conclusion regarding species identity, and molecular conformation is required to determine its position.

## ***Sarcodon*** Quélet ex P. Karst

Genus *Sarcodon* is one of four genera of stipitate hydnoid fungi belonging to family Bankeraceae. Basidiocarp of the species may have scales, centrally or eccentrically stipitate. Spines are usually of brown to purple colors. Spores produce brown print, shape is tuberculate (Karst 1881). Hypha may or may not have clamp connections, usually inflated. Based on earlier work species of genus *Sarcodon* are intermixed with species of genus *Hydnellum*, and represented as a paraphyletic group in ITS tree (Baird *et al*. 2013).

**KEY TO KOREA SPECIES OF *SARCODON***

1. Basidiocarp has no scales·····2

1. Basidiocarp has scales·····3

2. Basidiocarp has single color scheme·····*Sarcodon* sp. 1

2. Basidiocarp has bicolor scheme·····*S. glabrus*

3. Hymenophoral spines are up to 0.2 cm long·····4

3. Hymenophoral spines are up to 0.5 cm long·····6

4. Clamp connections are present occasionally·····*Sarcodon* sp. 3

4. Clamp connections are absent·····5

5. Basidiocarp is white to light brown colored·····*Sarcodon* sp. 2

5. Basidiocarp is brown to dark brown colored·····6

6. Chemical reaction of tissue is absent in KOH·····*S. squamosus*

6. Chemical reaction of tissue is brownish to green, olive in KOH·····

·····*S.* cf. *underwoodii*

**1. *Sarcodon* sp. 1**

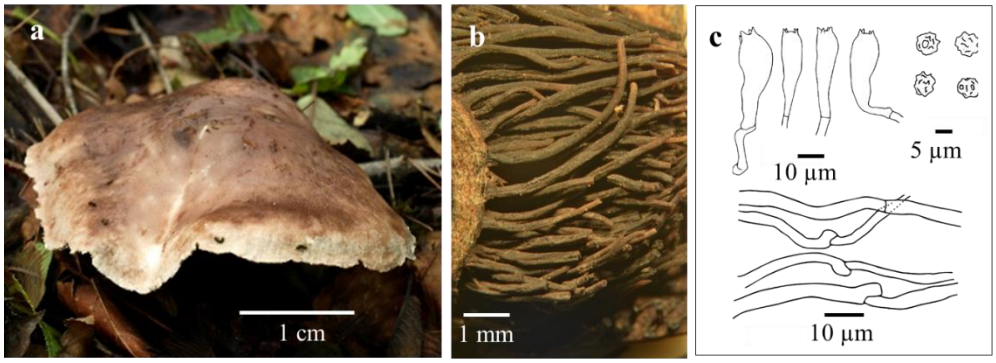


**Figure 20.** *Sarcodon* sp. 1. a fruting bodies in the field. b spines of fruiting body. c microscopic structures; basidia, basidiospores, generative hyphae.

*Basidiocarps* single to gregarious; pileus up to 3 cm broad, convex to planar, can be depressed, scales are present on pileus; brown, dark brown when fresh; brown to dark brown, black when when dry; context up to 0.15 cm thick. *Spines* up to 0.3 cm long, crowded, light brown when fresh, dark brown when dry. *Hyphae* inflated; pileus trama hyphae up to 13.0 µm wide; spine trama hyphae up to 5.0 µm wide, clamps are present. *Basidia* 35.0 – 48.0 × 6.5 – 10.0 µm, clavate, basal clamps are present, 4 spored, sterigmata up to 4.0 µm long. *Basidiospores* subglobose, ovoid, ornamentation tuberculate, 7.5 – 8.5 × 6.5 – 8.0 µm. L = 8.0 µm, W = 7.2 µm, Q = 1.10 (n=20/1). *Chemical reaction* of tissue is slightly brown in water, brown in KOH, absent in Meltzer's reagent.

*Habitat*: On soil in broadleaved forest, near *Quercus* species.

*Material examined*: KOREA, Buk-myeon, Inje-gun, Gangwon-do, on soil, 8 September 2007, S. K. Han (KM07-0701, holotype; GeneBank ITS: XXXX).

*Notes*: The sample falls in the group of *Sarcodon imbricatus*, although it lacks unique feature of scales on the basidiocarp. However, there is a possibility that our sample is immature, and scales would appear at later life stage. Regarding its position in ITS tree: it does not match closely with existing species, and it is probably a new species. The data are limited due to only a single specimen being present.

**2. *Sarcodon* sp. 2**

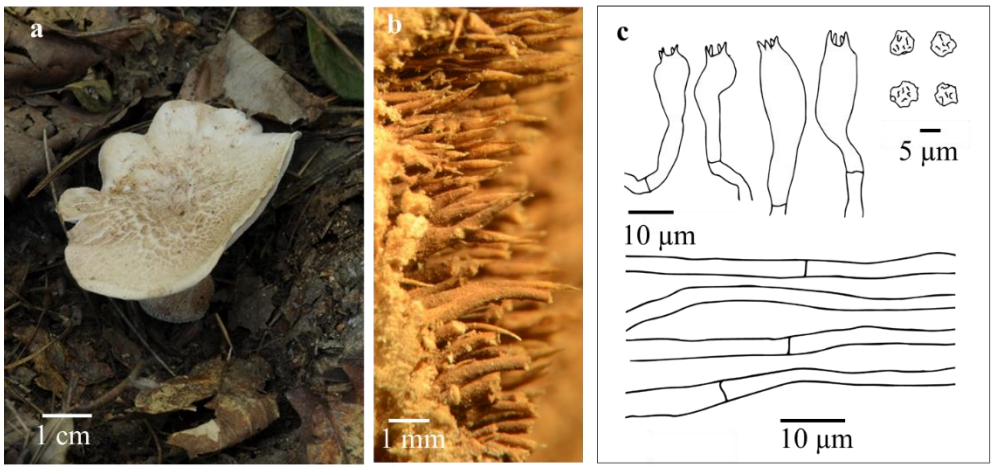


**Figure 21.** *Sarcodon* sp. 2. a fruting bodies. b spines of fruiting body. c microscopic structures; basidia, basidiospores, generative hyphae.

*Basidiocarps* single to gregarious; pileus up to 8 cm broad, convex to planar, glabrous, scales are present on the pileus; cream, light brown, beige when fresh; tan, light brown, brown when dry; context up to 3 cm thick, brittle when dry. *Spines* up to 0.2 cm long, crowded, beige to cream when fresh, cream, light brown when dry. *Stipe* up to 8 × 2 cm, central to sub-eccentric, same color scheme as spines. *Hyphae* inflated and simple

septated; pileus trama hyphae up to 6.0 µm wide; spine trama hyphae up to 4.0 µm wide; stipe hyphae up to 12.0 µm wide. *Basidia* 31.0 – 45.0 × 6.5 – 9.5 µm, clavate, unclamped, 4 spored, sterigmata up to 5 µm long. *Basidiospores* subglobose, ovoid, ornamentation tuberculate, 6.0 – 7.5 × 5.0– 6.0 µm. L = 6.7 µm, W = 6.0 µm, Q = 1.11 (n=20/1). *Chemical reaction* of tissue turns to olive in KOH, no reaction with water or Meltzer's reagent.

*Habitat*: On soil in broadleaved forest, near *Quercus* species.

*Material examined*: KOREA, Dongsan-myeon, Chuncheon-si, Gangwon-do, on soil, 23 September 2016, N. K. Kim, Y. W. Lim (SFC20160923-43, holotype; GeneBank ITS: XXXX). KOREA, Bukbang-myeon, Hongcheon-gun, Gangwon-do, on soil, 21 September 2016 N. K. Kim, Y. W. Lim (SFC20160921-01, holotype; GeneBank ITS: XXXX).

*Notes*: *Sarcodon* sp. 2 shares morphological features of species from *Sarcodon* group (*Sarcodon imbricatus*, *Sarcodon squamosus*), such as presence of scales on the basidiocarp and relatively large fruiting body The results of molecular analysis of ITS region showed, that our specimens appeared to be within group of *Sarcodon imbricatus*.

**3. *Sarcodon* sp. 3**


**Figure 22.** *Sarcodon* sp. 3. a fruting bodies in the field. b spines of fruiting body. c microscopic structures; basidia, basidiospores, generative hyphae.

*Basidiocarps* single to gregarious; pileus up to 8 cm broad, convex to planar, glabrous, small scales are present; white, tan, cream, light brown, beige when fresh; cream, tan, light brown, brown when dry; context up to 0.4 cm thick. *Spines* up to 0.5 cm long, crowded, beige to cream when fresh, cream, light brown when dry. *Stipe* up to 5.0 × 1.5 cm, central to sub-eccentric, concolorous with spines. *Hyphae* inflated and simple septated; pileus trama hyphae up to 6.0 µm wide; spine trama hyphae up to 4.0 µm wide; stipe hyphae up to 12.0 µm wide. *Basidia* 30.0 – 45.5 × 7.0 – 8.5 µm, clavate, clamp connections are present occasionally, 4 spored, sterigmata up to 4 µm long. *Basidiospores* subglobose, ovoid, ornamentation tuberculate, 6.0 – 7.5 × 5.0– 6.5 µm. L = 6.8 µm, W = 5.9 µm, Q = 1.14 (n=20/1). *Chemical reaction* of tissue is absent in KOH, water and Meltzer's reagent.

*Habitat*: On soil in broadleaved forest, near *Quercus* species.

*Material examined*: KOREA, Buk-myeon, Inje-gun, Gangwon-do, Korea, on soil, 25 August 2004, S. K. Han (KM04-0159, holotype; GeneBank ITS: XXXX). KOREA, Buk-myeon, Inje-gun, Gangwon-do, on soil, 21 July 2004, S. K. Han (KM04-0137, holotype; GeneBank ITS: XXXX).

*Notes*: Only photos of dried specimens were available for examination. Although, the fruiting body deformed after drying, a similarity to *Sarcodon imbricatus* can be easily traced: scales breaking from the top of basidiocarp and large spines can be seen.

**4. *Sarcodon squamosus*** (Schaeff) Karst

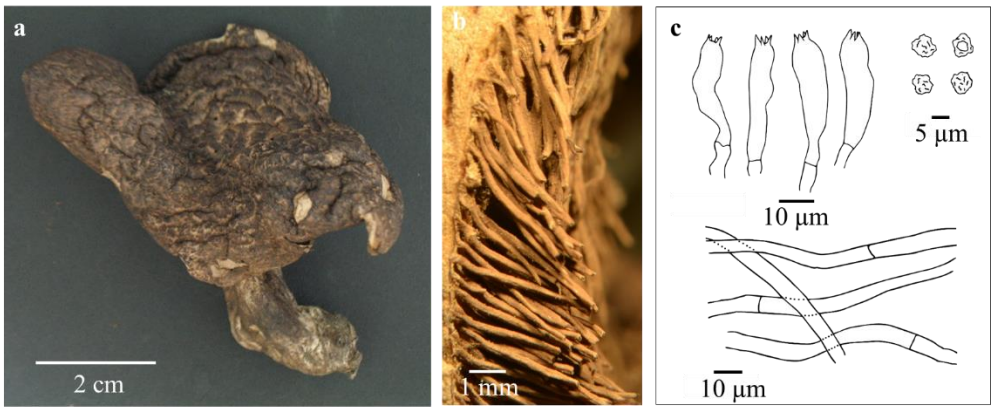


**Figure 23.** *Sarcodon squamosus*. a fruting bodies. b spines of fruiting body. c microscopic structures; basidia, basidiospores, generative hyphae.

*Basidiocarps* single to gregarious; pileus up to 6 cm broad, convex to planar, glabrous, scales are present on pileus; brown to dark brown, black when fresh; dark brown to black when dry; context up to 0.15 cm thick. *Spines* up to 0.2 cm long, crowded,

cream to brown when fresh, dark brown when dry. *Stipe* up to 5.0 × 1.5 cm, central to sub-eccentric, cream to light brown when fresh, brown to dark brown when dry. *Hyphae* uninflated; pileus trama hyphae up to 16.0 µm wide; spine trama hyphae up to 8.0 µm wide; stipe hyphae up to 18.0 µm wide. *Basidia* 35.0 – 42.0 × 6.5 – 10.0 µm, clavate, basal clamps are absent, 4 spored, sterigmata up to 5.5 µm long. *Basidiospores* subglobose, ovoid, ornamentation tuberculate, 7.0 – 8.5 × 6.0 – 8.0 µm. L = 7.6 µm, W = 7.0 µm, Q = 1.08 (n=20/1). *Chemical reaction* of tissue is absent in KOH, water and Meltzer's reagent.

*Habitat*: On soil in broadleaved forest, near *Quercus* species.

*Material examined*: KOREA, Girin-myeon, Inje-gun, Gangwon-do, on soil, 1 October 2014, H. J. Cho, Y. W. Lim (SFC20141001-12, holotype; GeneBank ITS: XXXX).

*Notes*: The examined species matches to morphological characteristics of *Sarcodon squamosus*. The dimensions are also that of *Sarcodon* species. In addition, phylogenetic analysis of ITS region showed that the examined species is close to reference sequences from GeneBank, the differences are due to geographical variations.

**5. *Sarcodon glabrus*** V. Li, N. K. Kim and Y. W. Lim, **sp. nov.**

*Index Fungorum number*: XXXX, *Faces of fungi number*: XXXX.

*Etymology*: referring to a more slender spore relative to other *Sarcodon* species.

*Holotype*: SFC20140822-38

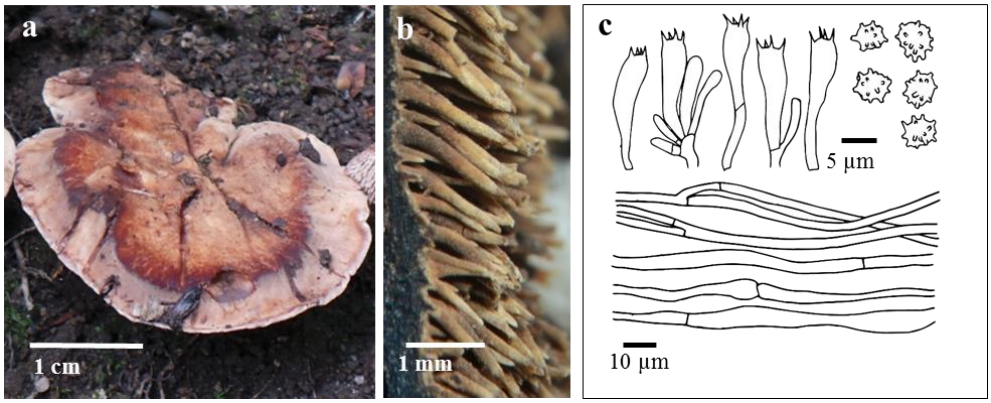


**Figure 24**. *Sarcodon glabrus*. a fruting bodies in the field. b spines of fruiting body. c microscopic structures; basidia, basidiospores, generative hyphae.

*Basidiocarps* single to gregarious; pileus up to 3 cm broad, convex to planar, glabrous, with smooth surface, orange, light brown, reddish brown, beige, or black when fresh; tan, light brown, brown or black when dry; context up to 0.3 cm thick, brittle. *Hymenophore* up to 0.2 cm long, crowded, beige to light brown when fresh, dark brown or black when dry. *Stipe* up to 0.5 × 2 cm, central to sub-eccentric, individual, smooth, white at the bottom, brown to black when fresh, light brown, brown or black when dry. *Hyphae* uninflated and simple septated; pileus trama hyphae up to 7.5 µm wide; spine trama hyphae up to 5 µm wide; stipe hyphae up to 5.5 µm wide.

*Basidia* 31 - 48 × 6–10 µm, clavate, unclamped, 4 spored, sterigmata up to 5 µm long. *Basidiospores* subglobose, ovoid, ornamentation tuberculate, 5–7 × 4–5.5 µm. L = 5.5 µm, W = 4.8 µm, Q = 1.14 (n=20/1). *Chemical reaction* of tissue turns to brownish to green in KOH, no reaction with water or Meltzer's reagent.

*Habitat*: On soil in broadleaved forest, near *Quercus* species.

*Material examined*: KOREA, Kangwon-do, Inje-gun, Mt. Jeombong, on soil, 22 August 2014, H. J. Cho, Y. W. Lim (SFC20140822-38, holotype; GeneBank ITS: XXXX, *rpb*2: KY00000). KOREA, Gyeongsangbuk-do, Bonghwa-gun, Mt. Munsu, on soil, 28 June 2013, N. K. Kim (TPML20130628-34, paratype; GeneBank ITS: XXXX, *rpb*2: KY00000).

*Notes*: The results from ITS identified this species as a relative of *Sarcodon joeides* and *Sarcodon fuligineoviolaceus* with strong supportive value of 97%. In terms of morphology, the studied sample has smooth basidiocarp and its dimensions are rather small, the pileus being only up to 3 cm broad. This species also features a unique color scheme: it is tan to light brown on the margin and then orange to brown in the middle. The border separating these two areas is clearly observable. The color scheme reminds that of *Phellodon tomentosus*, a species from other genus of family Bankeraceae, and can be used to preliminary identify this sample.

**6. *Sarcodon* cf. *underwoodii***

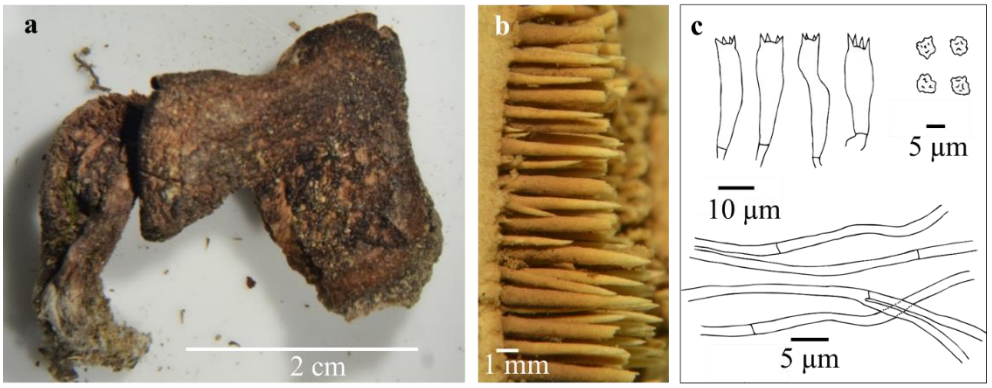


**Figure 25.** *Sarcodon* cf. *underwoodii***.** a fruting bodies in the field. b spines of fruiting body. c microscopic structures; basidia, basidiospores, generative hyphae.

*Basidiocarps* single to gregarious; pileus up to 8 cm broad, convex to planar, small scales are breaking from the basidiocarp; light brown to brown, black when fresh; tan, brown to dark brown or black when dry; context up to 1 cm thick, brittle. *Hymenophore* up to 0.4 cm long, crowded, tan, beige to light brown when fresh, brown to dark brown when dry. *Stipe* up to 4.5 × 3 cm, central to sub-eccentric, same color scheme with basidiocarp. *Hyphae* uninflated and simple septated; pileus trama hyphae up to 5 µm wide; spine trama hyphae up to 4 µm wide; stipe hyphae up to 10 µm wide. *Basidia* 30 – 45 × 6–10 µm, clavate, unclamped, 4 spored, sterigmata up to 5 µm long. *Basidiospores* subglobose, ovoid, ornamentation tuberculate, 6–7 × 5.5–6.5 µm. L = 6.3 µm, W = 5.8 µm, Q = 1.08 (n=20/1). *Chemical reaction* of tissue is absent in KOH, water or Meltzer's reagent.

*Habitat*: On soil in broadleaved forest, near *Quercus* species.

*Material* examined: KOREA, Buk-myeon, Ulleung-gun, Gyeongsangbuk-do, on soil, S. K. Han (KA12-1410, holotype; GeneBank ITS: XXXX). KOREA, Yeonpung-myeon, Goesan-gun, Chungcheongbuk-do, on soil, 8 October 2010, N. K. Kim, J. K. Lee (TPML20101008-133, holotype; GeneBank ITS: XXXX). KOREA, Dongsan-myeon, Chuncheon-si, Gangwon-do, on soil, 23 September 2016, N. K. Kim, Y. W. Lim (SFC20160923-44, holotype; GeneBank ITS: XXXX).

*Notes*: Specimens have all features of genus *Sarcodon*, such as presence of scales on basidiocarp, relatively large fruiting body. Our species resemble *Sarcodon underwoodii*, however ITS tree showed that it is probably a sister species of *Sarcodon underwoodii*.

# 4. Discussion

## 4.1. Morphological and molecular analysis

This work combined morphological and molecular means of identification in order to re-evaluate species of family Bankeraceae in Korea. Although, morphological studies of the family exist in Korean records (Lee and Cho 1975; Lee and Hong 1985), no molecular analysis data were collected previously.

Samples were initially grouped based on their macro- and microscopic characteristics across four genera: *Boletopsis*, *Phellodon*, *Hydnellum* and *Sarcodon.* Grouping based on macromorphology formed three major groups: 1) *Boletopsis*; 2) *Sarcodon*; 3) *Hydnellum* and *Phellodon*. *Boletopsis* was differentiated based on poroid hymenophore and *Sarcodon* was selected on the basis of having scaly basidiocarp or included species that did not fit the features of other genera. As a result, *Sarcodon* was split into two sub-categories: 1) Specimens with scales on the basidiocarp; 2) Specimens with smooth basidiocarp. Other genera were not divided and remained as single groups. Species of *Hydnellum* and *Phellodon* share multiple morphological characters (Parfitt *et al*. 2007), making it difficult to differentiate them in wild. Further addition of microscopic analysis allowed to separate genera *Hydnellum* and *Phellodon*. *Hydnellum* features basidiospores with indeterminate shape and large warts, while *Phellodon* has ovoid echinulate basidiospores with fine

spines (Donk 1961). These characteristics are sufficient to differentiate between two genera. Thus, all specimens were presumably assigned to their corresponding genera.

Molecular analysis involved three regions: ITS, *rpb*2 and LSU. All specimens were successfully amplified using ITS region, while only fraction of them was amplified with *rpb*2 and LSU gene markers. Nevertheless, the data of molecular analysis matched the results of morphological analysis to a great extent. Overall, four major groups could be identified from the tree: 1) *Sarcodon* and *Hydnellum*; 2) *Sarcodon*; 3) *Boletopsis* and 4) *Phellodon*. Group G1 included species of both genera (*Hydnellum* and *Sarcodon*), which are intermixed with each other. Correlating morphology with molecular works showed little success, since the species exhibit features unique to individual genus. There is a possibility that this group may be re-evaluated as *Hydnellum*, since species of this genus only appear in this section. Group G2, on the other hand contained solely species of *Sarcodon* and the type species of the genus – *Sarcodon imbricatus* (Karst 1881). All species included in this section possess scaly basidiocarp with except for *Sarcodon* sp. 1. We believe, that this group of *Sarcodon* is possibly a true *Sarcodon* clade. Groups G3 and G4 included only species of *Boletopsis* and *Phellodon* correspondingly.

Taxonomic positions based on *rpb*2 and LSU trees were neglected due to insufficient reference sequences available on the GeneBank. Nevertheless, both *rpb*2 and LSU trees complement the results seen on ITS by forming groups (G1 – G4), meaning that the pattern of phylogenetic relatedness can be traced.

## 4.2. Phylogenetic relationships

The results of this study correlate with earlier work performed by Baird *et al*. (2013) to a great extent. Particularly, genus *Phellodon* was resolved as a monophyletic group and appeared as an outgroup to the family. Genera *Hydnellum* and *Sarcodon*, although, had relatively moderate bootstrap values, were intermixed groups. Also, none of the genera in group G1 was monophyletic and their positions remain to be complicated. Group, labeled G2 in this study, was resolved as monophyletic with strong bootstrap value of 83%. This group of *Sarcodon* species appeared to be segregated from species of *Sarcodon* in group G1. Parfitt *et al*. (2007) showed that molecular analysis based on ITS, allows to easily distinguish between species of *Hydnellum* and *Phellodon*. We were able to confirm that there are no doubts, that these two genera are phylogenetically distant enough from each, thus supporting earlier works. In addition, there are no published data dedicated to studying genus *Boletopsis* with other genera of the family. However, from the result of this work, we can conclude with relative confidence, that *Boletopsis* is probably monophyletic group, since it was resolved with 100% bootstrap value. *Boletopsis* appears to be phylogenetically close to genus *Sarcodon* (G2) despite their quite different morphology.

In *rpb*2 and LSU trees similar pattern of phylogenetic relationships between Korean specimens can be observed. Four groups (G1 – G4) appeared in the tree. However, species *Sarcodon* sp. 1 (Korea) appeared in the group of *Boletopsis* rather

than group *Sarcodon* (G2) like in ITS tree. Groups of *Boletopsis* (G3) and *Sarcodon* (G2) were phylogenetically close to each other according to the results of the ITS tree, and it is possible than some regions can be more conserved among certain species, than other regions.

Korean species of *Sarcodon* sp. 2 and *Sarcodon* sp. 3, identified during this study, deserve special attention. Upon preliminary analysis, all specimens of these species were identified as *Sarcodon imbricatus*, which is currently considered to be present in Korea. Their morphological features are indistinguishable from that of *Sarcodon imbricatus*, and the arisen confusion is understandable. However, after completing molecular analysis, it became clear, that these species are different from *Sarcodon imbricatus*. The maximum sequence dissimilarity between reference *Sarcodon imbricatus* and Korean *Sarcodon* sp. 2 and *Sarcodon* sp. 3 was in a range of 10 – 11%, indicating significant sequence variation. In addition, no specimens of Korean *Sarcodon imbricatus* were identified during this study. This could possibly mean, that the species has never been previously found in Korea, and all species designated as *Sarcodon imbricatus* were misidentified as it.

# 5. Conclusions

Concluding from comprehensive results of morphological and molecular analyses 17 species of family Bankeraceae were identified in this study, including three new species, seven potentially new species to the family and five previously unrecorded species in Korea. Only two species from previous records in Korea were identified in this study, probably due to limitation of the specimens available for this study. Some species can be easily mistaken for the others, so molecular analysis can significantly amplify the efficiency of identification. Through comparative analysis of ITS, LSU and *rpb*2 molecular markers, the relationships between species and genera was established. Molecular analysis showed clear distinction between four groups (G1 – G4). In summary, species in group G2 could be established as true *Sarcodon*. *Boletopsis* and *Phellodon* formed individual groups and thus remain as they are. Group G1 may be considered to be renamed as genus *Hydnellum*, due to the genus species being only present in this group; and as of now, taxonomic positions of species in G1 remain to be complicated.

# 6. References

Ahn D. (1992). Medicinal Fungi in Korea. Kor J Mycol; 20(2): 154-66.

Arnold AE, Miadlikowska J, Higgins KL, Sarvate SD, Gugger P, Way A, Hofstetter V, Kauff F and Lutzoni F. (2009). A phylogenetic estimation of trophic transition networks for ascomycetous fungi: are lichens cradles of symbiotrophic fungal diversification? Syst Biol; 58: 283-97.

Arnolds E. (1991). Decline of ectomycorrhizal fungi in Europe. Agriculture, Ecosystems and Environment; 35: 209-44.

Baird R, Wallace LE, Baker G and Scruggs M. (2013). Stipitate hydnoid fungi of the temperate southeastern United States. Fungal Divers; 62(1): 41-114.

Binder M, Hibbett DS, Larsson KH, Larsson E, Langer E and Langer G. (2004). The phylogenetic distribution of resupinate forms across the major clades of mushroom-forming fungi (Homobasidiomycetes). Systematics and Biodiversity; 3(2): 113-57.

Blackwell M. (2011). The fungi: 1, 2, 3 . . . 5.1 million species? Am J Bot; 98: 426 – 38.

Cannon PF and Kirk PM. (2007). Fungal Families of the World. England: CAB International.

Chalot M and Bran A. (1998). Physiology of organic nitrogen acquisition by

ectomycorrhizal fungi and ectomycorrhizas. *FEMS Microbiology Reviews* 22; 22-44.

Cooper J and Leonard P. (2012). *Boletopsis nothofagi* sp. nov. associated with Nothofagus in the Southern Hemisphere. MycoKeys; 3: 13-22.

Corner EJH. (1989). Ad Polyporaceas V. Beihefte zur Nova Hedwigia; 96: 17.

Donk MA. (1961). Four new families of Hymenomycetes. Persoonia; 1(4): 405-7.

Fayod V. (1889). Sopra un nuovo genere di Imenomiceti. Malpighia (in Italian); 3: 69-73.

Gardes M and Bruns D. (1993). ITS primers with enhanced specificity for basidiomycetes-application to the identification of mycorrhizae and rusts. Mol Ecol; 2: 113-8.

Hattori T. (2001). Type studies of the polypores described by E.J.H. Corner from Asia and West Pacific Areas III. Species described in Trichaptum, Albatrellus, Boletopsis, Diacanthodes, Elmerina, Fomitopsis and Gloeoporus. Mycoscience; 42(5): 423-31.

Hirota MA, Morimura K and Shibata H. (2002). Anti-inflammatory Compounds from the Bitter Mushroom, *Sarcodon scabrosus*. Biosci Biotechnol Biochem; 66(1): 179-84.

James TY, Letcher PM, Longcore JE, Mozley-Standridge SE, Porter D, Powell MJ,

Griffith GW and Vilgalys R. (2006). A molecular phylogeny of the flagellated fungi (Chytridiomycota) and description of a new phylum (Blastocladiomycota). Mycologia; 98: 860-71.

Johannesson H, Ryman S, Lundmark H and Danell E. (1999). *Sarcodon imbricatus* and *S. squamosus* — two confused species. Mycol Res; 103(11): 1447-52.

Jülich W. (1981). Higher taxa of Basidiomycetes. Bibl Mycol; 85: p.485.

Kaburagi Y. (1940). Korean and Manchurian practical manual of forest. Korea Forest Experiment Station. Tokyo: Yokendo; 339-67.

Karst P. (1879). *Hydnellum*. Meddeland. Soc Fauna F1 Fenn; 5: 41.

Karst P. (1881). *Phellodon*. Rev Mycol; 3(9): 19.

Karst P. (1881). *Sarcodon*. Rev Mycol; 3(9): 20.

Katoh K and Standley DM. (2013). MAFFT multiple sequence alignment software version 7: improvements in performance and usability. Mol Biol Evol; 30: 772-80.

Kim NK, Park JY, Park MS, Lee H, Cho HJ, Eimes JA, Kim C and Lim YW. (2016). Five New Wood Decay Fungi (Polyporales and Hymenochaetales) in Korea. Mycobiology; 44(3): 146-154.

Ko P, Seok S and Jeun Y. (2010). Ecological Study on Wild Mushrooms at the Dongbaekdongsan of Seunheulgot in Jeju Island. Kor J Mycol; 38(1): 8-15.

Korean Society of Mycology. (1978). Suggestion on “Standard Korean names of mushrooms in Korea”. Kor J Mycol; 6(2): 43-55.

Kumar S, Stecher G and Tamura KH. (2016). MEGA7: Molecular Evolutionary Genetics Analysis Version 70 for Bigger Datasets. Mol Biol Evol; 33(7): 1870-4.

Larsson K, Larsson E and Kõljalg U. (2004). High phylogenetic diversity among corticioid homobasidiomycetes. Mycol Res; 108(9): 983-1002.

Le HT, Stubbe D, Verbeken A, Nuytinck J, Lumyong S and Desjardin DE. (2007). *Lactarius* in Northern Thailand: 2 *Lactarius* subgenus *Plinthogali*. Fungal Divers; 27: 61-94.

Lee D, Boo KH, Lee JM, Viet CD, Quyen N, Unno T, Cho M, Riu KZ and Lee DS. (2012). Anti-viral activity of *Hydnellum concrescens*, a medicinal mushroom. African Journal of Biotechnology; 11(86): 15241-5.

Lee DS and Lee YW. (1957). List of Korean fungi (1). Korea Forest Research Institute, Seoul.

Lee TS. (1990). The Full List of Recorded Mushrooms in Korea. Kor J Mycol; 18(4): 233-259

Lee H, Park MS, Jung PE, Fong JJ, Oh SY, Verbeken A and Lim YW. (2015). *Lactarius cucurbitoides* (Russulales, Basidiomycota), a new species from South Korea supported by molecular and morphological data. Phytotaxa;

205(3): 168-76.

Lee JS and Jung HS. (2005). List of Recorded Korean Aphyllophorales. Kor J Mycol; 33(1): 38-53.

Lee JY and Cho DH. (1975). Notes on Korean Higher Fungi. Kor J Mycol; 3(2): 13-8.

Lee JY and Hong SW. (1985). Illustrated Flora, Fauna of Korea. Mushrooms; 28.

Lee YS, Lim YW, Kim JJ, Yun HY, Kim C and Park JY, Korean Society of Mycology. (2015). National List of Species of Korea (Basidiomycota). Republic of Korea: National Institute of Biological Resources Ministry of Environment.

Linnaeus C. (1753). Species Plantarum.

Liu YL, Whelen S and Hall BD. (1999). Phylogenetic relationships among ascomycetes: evidence from an RNA polymerase II subunit. Mol Biol Evol; 16: 1799-808.

Marren P and Dickson G. (2000). British tooth-fungi and their conservation. British Wildlife; 11(6): 401-9.

Martin KJ and Rygiewicz PT. (2005). Fungal-specific PCR primers developed for analysis of the ITS region of environmental DNA extracts. BMC Microbiology; 5(28): 1-11.

Miller MA, Pfeiffer W and Schwartz T. (2010). "Creating the CIPRES Science

Gateway for inference of large phylogenetic trees" in Proceedings of the Gateway Computing Environments Workshop (GCE); New Orleans, LA; p.1-8.

Min YJ, Park MS, Fong JJ, Seok SJ, Han S and Lim YW. (2014). Molecular Taxonomical Re-classification of the Genus *Suillus* Micheli ex S. F. Gray in South Korea. Mycobiology; 42(3): 221.

Öpik M, Vanatoa A, Vanatoa E, Moora M, Davison J, Kalwij JM, Reier U and Zobel M. (2010). The online database MaarjAM reveals global and ecosystemic distribution patterns in arbuscular mycorrhizal fungi (Glomeromycota). New Phytol; 188: 223-41.

Parfitt D, Ainsworth M, Simpson D, Rogers HJ and Boddy L. (2007). Molecular and morphological discrimination of stipitate hydnoids in the genera *Hydnellum* and *Phellodon*. Mycol Res III; 761-77.

Park MS, Fong JJ, Lee H, Oh SY, Jung PE, Min YJ, Seok SJ and Lim YW. (2013). Delimitation of *Russula* subgenus *Amoenula* in Korea using three molecular markers. Mycobiology; 41: 191-201.

Schoch CL, Seifert KA, Huhndorf S, Robert V, Spouge JL, Levesque CA and Chen W. (2012). Fungal Barcoding Consortium. Nuclear ribosomal internal transcribed spacer (ITS) region as a universal DNA barcode marker for fungi. Proc Natl Acad Sci U S A; 109: 6241-6.

Senn-Irlet B, Heilmann-Clausen J, Genney DR and Dahlberg A. (2007). Guidance for Conservation of Macrofungi in Europe. ECCF, Strasbourg.

Smith SE and Red D. (2008). Mycorrhizal symbiosis. New York: Academic Press.

Stalpers JA. (1993). The aphyllophoraceous fungi I. Keys to the species of the Thelephorales. Baarn: Centraalbureau voor Schimmelculture.

Stubbe D, Nuytinck J and Verbeken A. (2008). *Lactarius* subgenus *Plinthogalus* of Malaysia. Fungal Divers; 32: 125-56.

Větrovský T, Kolařík M, Žifčáková L, Zelenka T and Baldrian P. (2015). The *rpb*2 gene represents a viable alternative molecular marker for the analysis of environmental fungal communities. Mol Ecol Resour; 16(2): 388-401.

Vilgalys R and Hester M. (1990). Rapid genetic identification and mapping of enzymatically amplified ribosomal DNA from several *Cryptococcus* species. Journal of Bacteriology; 172: 4238-46.

White TJ, Bruns T, Lee S and Taylor J. (1990). Amplification and direct sequencing of fungal ribosomal RNA genes for phylogenetics. In: PCR Protocols: a guide to methods and applications. (Innis MA, Gelfand DH, Sninsky JJ, White TJ, eds). Academic Press, New York, USA: 315-22.

## 7. Abstract in Korean

# Bankeraceae과의 계통학적 연구

Vladimir Li

생명과학부 대학원

서울대학교

## 초 록

Bankeracerae과는 소나무나 참나무와 외생균근형성 형성하고 자실층은 침형이며 대를갖는 특징을 지니고 있다. 현재 유럽에서는 Bankeraceae 과에 속하는 종들의 수가 급격히 감소하고 있어 생태적으로 위기종으로 분류되고 있으며, 일부 종은 적생목록에 등재되어있다. 현재 전세계적으로 6속 98종이 보고되어있으며, 한국에서는 4속 11종이 보고되어있다. 그러나 한국의 종들은 유럽이나 북미의 종과 외형적 특징 비교를 통해 그들의 학명이 도입되었고 정확한 자실체 형태와 현미경적 기재가 없는 경우가 많다. 최근 연구에서 아시아의 종들이 유럽이나 북미의 종들과 상

당히 다르다는 것이 DNA 염기서열 분석을 통해 밝혀지고 있다. 본 연구에서는 한국에 자생하는 32개 Bankeraceae속의 표본을 이용하여 형태적 동정과 함께 ITS, LSU, 그리고 rpb2를 기반으로 한 분자동정으로 Bankerace내의 종의 분류학적 재평가를 하였다. 그 결과 확보된 32개 표본은 *Boletopsis* 에서 1종, *Phellodon*에서 6종, *Hydnellum*에서 4종, 그리고 *Sarcodon*에서 6종으로 총 17종으로 확인되었다. 그 중, 2종만이 기록종과 일치하였고 5종은 미기록종, 3종은 신종, 그리고 7종은 신종 후보 종으로 확인되었다. 본 연구를 통해 계통학적으로 Boletopsis속과 Phellodon속이 단일 계통에 속하는 것을 확인한 반면, Hydnellum속과 Sarcodon속은 여전히 불분명하게 섞이므로 이들이 단계통이고 아님을 재차 확인하게 되었다.

**주요어**: 방패버섯과, 침형버섯, ITS, LSU, *rpb*2, 계통학

**학번:** 2015-22296