RESEARCH ARTICLE

First Korean Records of *Scutellinia sinosetosa* and *S. subhirtella* (Pezizales, Pyronemataceae) Based on Integrative Taxonomy

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ABSTRACT

Recently added new species and taxonomic re-evaluations of previously recorded ones have warranted a revision of *Scutellinia* specimens deposited in Korea. By applying an integrative taxonomic framework to *Scutellinia* specimens curated at the Korea National Arboretum (KH), we recorded two species previously unreported in Korea, namely *Scutellinia sinosetosa* and *S. subhirtella*. Morphological assessment covered the macroscopic traits of apothecia as well as microscopic characteristics, including asci, paraphyses, marginal hairs, ascospore size, and ornamentation. Molecular evidence was obtained from internal transcribed spacer (ITS) and large subunit (LSU) rDNA sequences; phylogenetic analyses placed the Korean specimens in strongly supported clades alongside authenticated reference material, confirming their identities. *S. sinosetosa* is distinguished by small apothecia and low, reticulate ascospore ornamentation, whereas *S. subhirtella* is characterized by medium-sized, ellipsoid ascospores densely covered with discrete, fine verrucae. These findings represent the first confirmed records of both species in Korea, extend the known distribution of *Scutellinia* on the Korean Peninsula, and provide ITS/LSU barcode data to facilitate reliable molecular identification in future studies.

Keywords: Pezizales, *Scutellinia*, *Scutellinia sinosetosa*, *Scutellinia subhirtella*
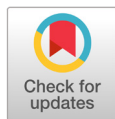
INTRODUCTION

The genus *Scutellinia* (Pezizales, Pyronemataceae) is a group of saprophytic discomycetes that typically produces apothecia on decaying wood or moist soil. Members of this genus are morphologically characterized by disc- to cup-shaped fruiting bodies, dark-colored marginal hairs, and ornamented, ellipsoid ascospores [1]. *Scutellinia* species are distributed worldwide, with numerous reports originating from temperate regions of the Northern Hemisphere [1–3]. As of July 2025, approximately 150 valid species of the genus are listed in Index Fungorum (October 2025).

Many of these species were originally described based solely on morphological features. However, characteristics such as spore ornamentation, marginal hair structure, and coloration or size of the fruiting bodies can vary depending on specimen condition, environmental factors, and the observer's interpretation, making species-level identification unreliable in many cases [1,4]. In response, molecular phylogenetic analyses using ribosomal DNA regions, such as internal transcribed spacer (ITS) and large subunit (LSU), have become increasingly important tools for accurate classification and identification of *Scutellinia* species [1,5,6].

Angelini et al. [1] conducted a broad phylogenetic analysis of numerous specimens and proposed nearly 50 putative phylogenetic species, noting that several morphologically identified taxa were actually polyphyletic. These findings, along with other recent studies, suggest that many previously reported species may not represent true taxonomic units, underscoring the need to reassess species concepts and perform more precise identifications [3,7].

Despite a growing body of sequencing data, more than half of the currently recognized *Scutellinia* species still lack ITS and LSU sequences in public databases, such as GenBank. Furthermore, several registered sequences were not linked to type specimens or voucher materials, and some were positioned unexpectedly in the phylogenetic trees [5,6]. These issues highlight the importance of careful selection and verification of comparative sequences, and they emphasize the need for a critical interpretation of phylogenetic results.

Nevertheless, ITS and LSU remain the most widely used and essential molecular markers for species identification in *Scutellinia*. They have been consistently applied for the discovery and description of new species and taxa worldwide [1,8].

In Korea, studies on *Scutellinia* have been historically limited. However, two new species and eight new records were reported in morphological investigations by Han et al. [8] and Choi et al. [9], significantly expanding the known diversity of the genus in the region. Additional collections have since been reported in areas, such as Mudeungsan National Park [10]. Nonetheless, some previously reported species were later excluded based on morphological reassessments or their identification has been questioned [6,9], warranting a comprehensive taxonomic revision. Morphological and molecular examinations are essential for a more accurate assessment of species diversity [1,5,8].

Accordingly, we re-examined *Scutellinia* specimens deposited at the Korea National Arboretum (KH), during which we identified several species previously unrecorded in Korea. These taxa are reported herein.

MATERIALS AND METHODS

Specimen collection

This study used dried specimens of the genus *Scutellinia* deposited at the Forest Biological Specimen Center of the Korea National Arboretum (KH). To clarify the taxonomy of the unrecorded taxa, we examined both the morphological traits and DNA sequences of selected specimens (Table 1).

Table 1. GenBank accession numbers used for the phylogenetic analyses in this study, including origin and specimen number

Species name	Voucher	Country	GenBank accession number	
			ITS	LSU
Melastiza cornubiensis	PM: 513	Italy	MW540956	MW541033
Scutellinia badio-berbis	JAC9770	New Zealand	PX215453	–
Scutellinia barlae	PM: 442	Italy	MW540900	MW540984
Scutellinia barlae f. *aurantiaca*[T]	MPU:JCD 177-78	France	MT273642	MT273655
Scutellinia citrina	LY: NV 2021.08.13	France	OM681398	OL832124
Scutellinia colensoi	MCKU 020906-13	South Korea	AY220828	–
Scutellinia colensoi	MCKU 020913-16	South Korea	AY220829	–
Scutellinia crinita	ACD0154	USA, WI	MZ919161	–
Scutellinia erinaceus	FLAS:F-65564	USA, MN	MT386345	MT375870
Scutellinia hirta	MCVE 12884	Italy	JF908713	–
Scutellinia hrabanovi[T]	PRM: 147265	Czech Rep.	MW540958	–
Scutellinia jejuensis[T]	KUS-F52411	South Korea	–	GU361609
Scutellinia kerguelensis	LY: NV 2018.08.11	Switzerland	–	OL832127
Scutellinia megalosphaera[T]	C: F-86967	Greenland	–	OL832129
Scutellinia minor	PM: 472	Italy	MW540923	MW541006
Scutellinia nigrohirtula	420526MF0257	China	MH142016	MH668011
Scutellinia nigrohirtula	MCKU 020711-25	South Korea	AY220809	–
Scutellinia olivascens	iNat: 136703676	USA, PA	OP749073	–
Scutellinia parvispora[T]	PRM: 724262	Greece	–	OL832131
Scutellinia patagonica	LY: NV 2013.08.13	France	–	OL832132
Scutellinia pseudoumbrarum[T]	PRM: 710003	Bosnia	MW540967	MW541040
Scutellinia scutellata	PM: 456	Italy	MW540908	MW540991
Scutellinia setosa	FLAS:F-65570	USA, IA	MT386365	MT375881
Scutellinia setosissima	PM: 460	Italy	MW540912	MW540995
Scutellinia sinosetosa	HKAS 124373	China	OQ024064	OP291093
Scutellinia sinosetosa	**KA24-0296**	**South Korea**	**PX443499**	**PX443266**
Scutellinia subhirtella	**KA21-0226**	**South Korea**	**PX443497**	**PX443264**
Scutellinia subhirtella	**KA25-0537**	**South Korea**	**PX443500**	**PX443267**
Scutellinia subhirtella	LY:NV 2017.08.12	France	OM681385	OL832133
Scutellinia subhirtella	MCKU 010829-01	South Korea	AY220824	–
Scutellinia subhirtella[T]	PRM: 616842	Czech Rep.	MW540963	MW541038
Scutellinia superba	PM: 467	Italy	MW540919	MW541002
Scutellinia umbrorum	FLAS: F-64198	USA, OH	MT386366	MT375882
Scutellinia verruculosa[T]	HKAS 104667	China	OQ024065	NG_229023

ITS: internal transcribed spacer; LSU: large subunit ribosomal DNA.
[T]type strain.
The sequences from this study are bold.

Morphological observations

Habitat conditions were recorded through photographs taken during the initial collection. The macroscopic and microscopic features were documented using a stereomicroscope (Leica Z16 APO; Leica, Wetzlar, Germany) and a compound light microscope (Leica DM 6000 FS; Leica). Microscopic preparations were mounted in distilled water, and 3% potassium hydroxide (KOH) to facilitate the observations and measurements of ascospores, asci, marginal hairs, and tissue structures.

DNA extraction and PCR amplification

Genomic DNA was isolated from the dried specimens using the DNeasy Plant Mini Kit (Qiagen, Hilden, Germany), following the manufacturer's protocol. The ITS and LSU of ribosomal DNA regions were targeted for amplification. The ITS region was amplified using primers ITS5 and ITS4, whereas the LSU region was amplified using the primers LR0R and LR7 [11]. PCR reactions were prepared using either AccuPower® Taq PCR PreMix or AccuPower® Gold HotStart Taq PCR PreMix & Master Mix (Bioneer, Daejeon, Korea). The 20 μL PCR mixture included 1 μL each of the forward and reverse primers, 1 μL of DNA template, and nuclease-free water to the final volume. Each PreMix contained all necessary PCR reagents, including buffer, dNTPs, Taq polymerase, and tracking dye. Thermal cycling was conducted as follows: initial denaturation at 94℃ for 5 min; 30 cycles of 94℃ for 20 s, 56℃ for 20 s, and 72℃ for 30 s; and a final elongation at 72℃ for 3 min. PCR products were stored at 4℃ until sequencing.

Sequencing and phylogenetic analysis

The amplified products were purified and sent to Macrogen, Inc. (Seoul, Korea) for sequencing. Sequence reads were trimmed, assembled, and edited using Geneious Prime software. BLASTn searches (v. 2.13.0) were conducted against the GenBank database to determine species-level similarity. Sequences were aligned using MAFFT version 7 and manually adjusted as necessary. Phylogenetic relationships were inferred using maximum likelihood (ML) and Bayesian inference (BI). JModelTest v. 2.1.10 (https://github.com/ddarriba/jmodeltest2) was used to identify the best-fitting model of nucleotide substitution, which was GTR + I + G. BI analysis was performed over 1,000,000 generations, with the first 250,000 discarded as burn-in. Consensus trees were visualized using FigTree v. 1.4.4 (https://tree.bio.ed.ac.uk/software/figtree/).

RESULTS

Molecular analysis

To assess the genetic identity of *Scutellinia* specimens curated at the KH, we sequenced the rDNA ITS and LSU regions and queried them against NCBI BLAST. The ITS sequences of KA21-0226 and KA25-0537 showed the highest similarity to *Scutellinia subhirtella* MCKU 010829-01 (GenBank AY220824), with 99.66% (580/582 bp) and 100% (582/582) identity, respectively. Matches were also detected to the unidentified *Scutellinia* sp. 'IN06' OMDL iNat #221953006 (PX118746) at 97.73% (604/618) and 97.69% (591/605) as well as to *S. subhirtella* HMAS 287034 (OQ534227) at 97.68% (547/560) and 97.68% (548/561). LSU sequences were likewise closest to *S. subhirtella* HMAS 287034 (OQ534511) at 99.55% (879/883) and 99.44% (883/888), followed by *S. subhirtella* MYC2105-15 (OL832133) at 99.33% (817/821) and 99.21% (881/888). Specimen KA24-0296 showed an ITS sequence most similar to *S. superba* MCKU 010719-06 (AY220818) at 99.65% (572/574), with the following additional *S.*

superba accessions from MCKU showing 98.78–99.30% identity. A further match to *S. sinosetosa* ZM 137 (OQ024064) was observed at 98.66% (589/597). The LSU sequence was closest to *S. sinosetosa* HKAS 124373 (OP291093) at 99.45% (899/904) and *S. subhirtella* KS-95-059A (C) (DQ220423) at 98.53% (870/883). Based on NCBI BLAST searches of the ITS and LSU sequences from the KH specimens, KA21-0226 and KA25-0537 were most similar to *S. subhirtella*, whereas KA24-0296 showed similarity to *S. superba* and *S. sinosetosa*.

A maximum-likelihood tree based on the concatenated ITS+LSU alignment, together with the reference accessions (Table 1), placed KA21-0226 and KA25-0537 in a single clade with *S. subhirtella* (including the type) with strong support (Fig. 1). KA24-0296 clustered in a well-supported clade with *S. sinosetosa* HKAS 124373, clearly separated from neighboring lineages (Fig. 1). These phylogenetic positions were similar to the BLAST results.
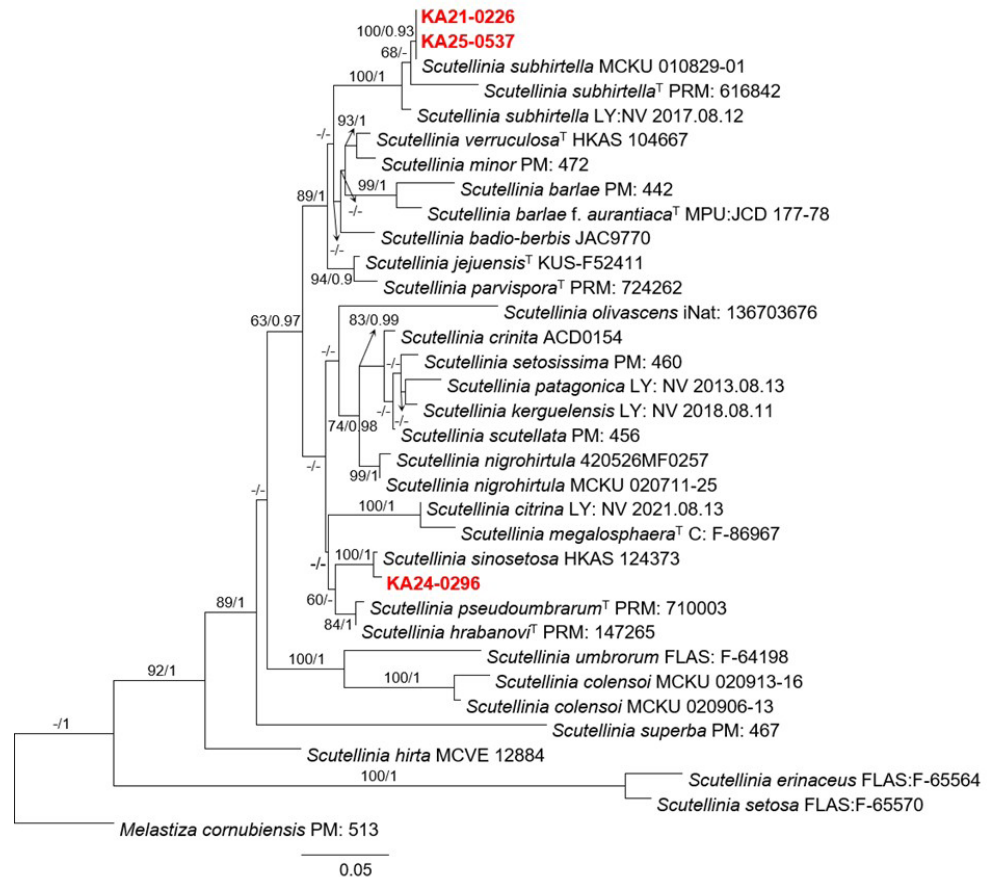

Fig. 1. RAxML phylogenetic tree inferred from a concatenated dataset of the internal transcribed spacer (ITS) and large subunit ribosomal DNA (LSU) regions. Maximum-likelihood bootstrap support (BS) values ≥ 50% and Bayesian posterior probabilities (BPP) ≥ 0.90 are shown at the nodes (BS/BPP). The newly described species in this study are indicated in red.

TAXONOMY

Scutellinia sinosetosa W.Y. Zhuang & Zheng Wang, Mycotaxon 69:352 (1998) [MB#447085] (Fig. 2)

Korean name: 동양털접시버섯

Description: *Apothecia* 3–7.9 mm in diameter, discoid, scattered, bright orange-yellow, margin with long and short hairs. *Hairs* 537–1,429 μm long, 29–55 μm thick, root-like branched at base, thick-walled, densely septate, yellow-brown. *Asci* cylindric, 193.2–269.38 × 14.5–20.25 μm, eight-spored, with uniseriate ascospores. *Ascospores* 18.8–23.3 × 11.73–15.79 μm, broadly ellipsoid to ellipsoid, hyaline, equilateral, rounded at the ends, verruculose. *Paraphyses* 233–290 μm long, 3.04–5.01 μm wide, filiform, slightly enlarged at apex, hyaline.

Specimen examined: Korea, Gyeonggi province, Pochun city, Gwangneung forest, 37°45'45″N, 127°09'25″E, elevation 183 m, on soil, June 11, 2024 (KA24-0296).

Remark: *Scutellinia sinosetosa* is morphologically similar to *S. setosa*, *S. crinita*, and *S. olivascens* in having ellipsoid, warted ascospores and long marginal hairs [12]. However, it can be distinguished from *S. setosa* by its longer marginal hairs (700–1200 μm vs. 600–900 μm) with less frequent branching and more densely warted spore surfaces. Compared to *S. crinita*, *S. sinosetosa* has thicker marginal hairs and rougher, more extensively ornamented spores. It differs from *S. olivascens* by its thicker, longer marginal hairs, and more coarsely warted spores, while *S. olivascens* exhibits finer ornamentation and paler, narrower hairs [3,8,12,13].
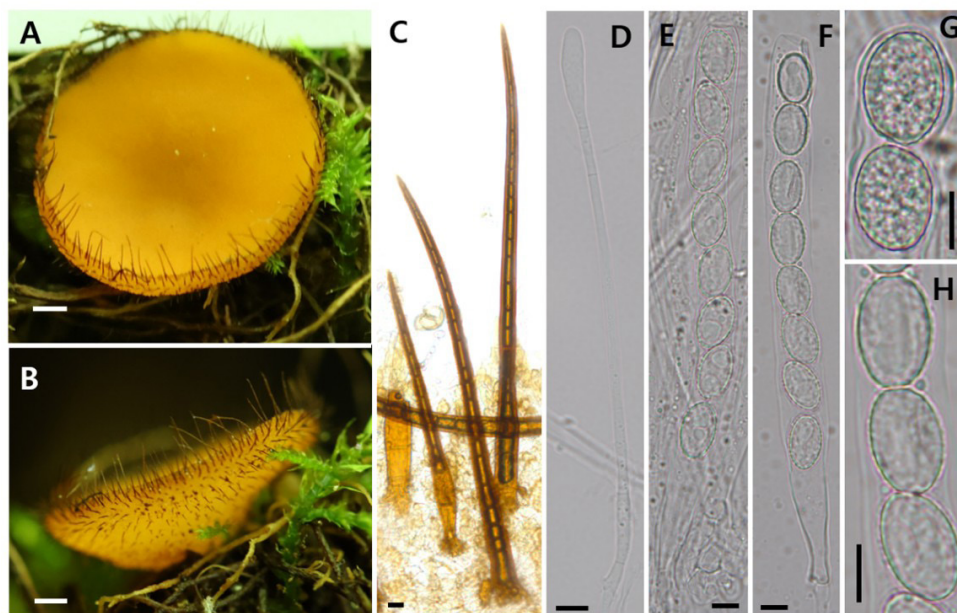

Fig. 2. *Scutellinia sinosetosa* KA24-0296. (A, B) Apothecia; (C) Marginal hairs; (D) Paraphyses; (E, F) Ascus; (G, H) Ascospores. Scale bars = 1 mm (A, B), 10 μm (C–H).

Scutellinia subhirtella Svrček, Česká Mykol. 25(2): 85 (1971) [MB#323350] (Fig. 3)

Korean name: 잔털접시버섯

Description: *Apothecia* 1.3–7.9 mm in diameter, scattered but occasionally gregarious, patellarioid to disciform, orange to orange-red, margin shortly but not densely hairy, with yellow-brown hairs. *Hairs* 251–443 μm long, 16–26 μm thick, root-like branched at base, thick-walled, densely septate. *Asci* cylindric, 197.59–280.59 × 14.45–17.98 μm, eight-spored, with uniseriate ascospores. *Ascospores* 17.73–22.68 × 10.97–14.72 μm, broadly ellipsoid to ellipsoid, rarely slightly narrowly ellipsoid, hyaline, equilateral, rounded at the ends, densely verrucose. *Paraphyses* 258–290 μm long, 3.06–4.17 μm wide, filiform, slightly enlarged at apex, hyaline.

Specimen examined: Korea, Gangwon province, Pyongchang-gun, Jinbu-myeon, Dongsan-ri, 37°44'20″N, 128°35'33″E, on decayed wood, elevation 789 m, May 30, 2021 (KA21-0226); Korea, Gangwon province, Inje-gun, Girin-myeon, Gombaeryeong, 38°02'23″N, 128°28'5″E, elevation 757 m, on decayed wood, July 02, 2025 (KA25-0537).

Remark: *Scutellinia subhirtella* is morphologically comparable to *S. setosa*, *S. umbrorum*, and *S. olivascens*, sharing ellipsoid, warted ascospores and marginal hairs. However, it can be readily distinguished from *S. setosa* by its shorter, thick-walled marginal hairs with bifurcate bases and more uniformly ornamented ascospores. Compared to *S. umbrorum*, *S. subhirtella* has less reticulate ornamentation and shorter, broader hairs. It differs from *S. olivascens* by possessing more conspicuous, evenly distributed warts on the spore surface and generally darker, thicker marginal hairs. These morphological differences support the recognition of *S. subhirtella* as a distinct species [8,13–15]. This species resembles *S. patagonica*
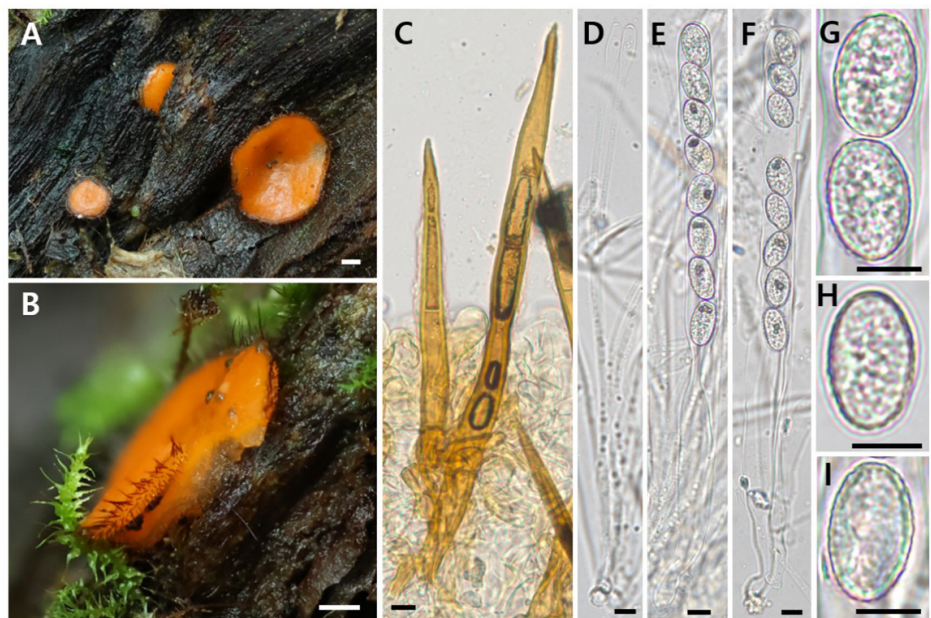

Fig. 3. *Scutellinia subhirtella* KA25-0537. (A, B) Apothecia; (C) Marginal hairs; (D) Paraphyses; (E, F) Ascus; (G-I) Ascospores. Scale bars = 1 mm (A, B), 10 μm (C–I).

Gamundí in spore size and general apothecial appearance. However, *S. subhirtella* is distinguished by the presence of subcylindrical, thin-walled, hyaline hairs intermixed with marginal hairs, which are absent in *S. patagonica*. In addition, the ascospores of *S. subhirtella* (18–22 $\times$ 12–14 μm) are slightly smaller and more densely verrucose than those of *S. patagonica* (18.5–23.5 $\times$ 14.5–21 μm), and the ornamentation is typically more evenly distributed [13–15].

DISCUSSION

Using an integrative taxonomic approach to KH vouchers, we confirmed two species previously unrecorded in Korea, *Scutellinia sinosetosa* and *S. subhirtella*. BLAST searches of the ITS and LSU sequences and a concatenated (ITS+LSU) ML phylogeny yielded similar outcomes: KA21-0226 and KA25-0537 were consistently supported as *S. subhirtella*, whereas KA24-0296 was supported as *S. sinosetosa*. Morphologically, *S. sinosetosa* is characterized by long, thick-walled marginal hairs and densely, coarsely ornamented ascospores, while *S. subhirtella* shows short, thick-walled marginal hairs that are often bifurcate at the base and ellipsoid ascospores (18–22 $\times$ 12–14 μm) with uniformly distributed fine verrucae.

In NCBI BLAST searches, *Scutellinia sinosetosa* KA24-0296 showed the highest similarity to MCKU accessions labeled *S. superba*, but these sequences lacked publicly verifiable linked morphology; therefore, a species-level assignment based on the labels alone is not possible. Consistent with this, Angelini et al. [1] withheld species-level confirmation, and Carbone and Rubini [2] treated the same clade as *S. sinosetosa*. Morphologically, *S. sinosetosa* has long, thick-walled marginal hairs and spores with dense, coarse warts and only weakly reticulate elements, whereas *S. superba* has been described with relatively shorter hairs and a more evident reticulate component with less densely developed warts [12,15]. Considering the diagnostic morphology of our vouchers, together with their phylogenetic position, the Korean material is best interpreted as *S. sinosetosa*.

In NCBI BLAST searches and ITS+LSU phylogenetic analyses, the Korean spcimens KA21-0226 and KA25-0537 showed the highest similarity to, and clustered with, reference sequences of *S. subhirtella*. Morphologically, they agree with *S. subhirtella* in having short, stout, thick-walled marginal hairs often bifurcate at the base, ellipsoid ascospores with dense fine verrucae lacking a true reticulum, and filiform paraphyses with orange pigment and slightly clavate apices [8,13–15]. Therefore, we regarded the Korean specimens as *S. subhirtella*.

Taken together, the concordance of morphological and molecular evidence enhances the confidence in our determinations. This study provides the first Korean records of *S. sinosetosa* and *S. subhirtella*, extends the known distribution of *Scutellinia* on the Korean Peninsula, and improves reproducibility and routine identification by linking KH-curated vouchers to publicly available ITS and LSU sequences. In *Scutellinia*, morphological characteristics alone are insufficient for identification, and an integrative taxonomic approach coupled with molecular markers is essential.

CONFLICT OF INTEREST

No potential conflict of interest was reported by the authors.

ACKNOWLEDGEMENTS

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