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PII: S1878-6146(24)00079-5

DOI: https://doi.org/10.1016/j.funbio.2024.05.014

Reference: FUNBIO 1475

To appear in: *Fungal Biology*

Received Date: 14 August 2023

Revised Date: 1 April 2024

Accepted Date: 14 May 2024

Please cite this article as: Lee, H.B., Nguyen, T.T.T., Noh, S.J., Kim, D.H., Kang, K.H., Kim, S.J., Kirk, P.M., Avery, S.V., Medina, A., Hallsworth, J.E., *Aspergillus ullungdoensis* sp. nov., *Penicillium jeongsukae* sp. nov., and other fungi from Korea, *Fungal Biology*, https://doi.org/10.1016/j.funbio.2024.05.014.

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Fungal Biology, Available online 6 June 2024
DOI:10.1016/j.funbio.2024.05.014

Aspergillus ullungdoensis sp. nov., *Penicillium jeongsukae* sp. nov., and other fungi from Korea

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ABSTRACT

Eurotiales fungi are thought to be distributed worldwide but there is a paucity of information about their occurrence on diverse substrates or hosts and at specific localities. Some of the *Eurotiales*, including *Aspergillus* and *Penicillium* species, produce an array of secondary metabolites of use for agricultural, medicinal, and pharmaceutical applications. Here, we carried out a survey of the *Eurotiales* in Korea, focusing on soil, freshwater, and plants (dried persimmon fruits and seeds of *Perilla frutescens*, known commonly as shiso). We obtained 11 species that—based on morphology, physiology, and multi-locus (ITS, *BenA*, *CaM*, and *RPB2*) phylogenetic analyses—include two new species, *Aspergillus ullungdoensis* sp. nov. and *Penicillium jeongsukae* sp. nov., and nine species that were known, but previously not described in Korea, *Aspergillus aculeatinus*, *Aspergillus aurantiacoflavus*, *Aspergillus croceiaffinis*, *Aspergillus pseudoviridinutans*, *Aspergillus uvarum*, *Penicillium ferraniaense*, *Penicillium glaucoroseum*, *Penicillium sajarovii*, and one, *Penicillium charlesii*, that was isolated from previously unknown host, woodlouse (*Porcellio scaber*). We believe that biodiversity surveys and identifying new species can contribute to set a baseline for future changes in the context of humanitarian crises such as climate change.

Keywords: Aspergillus ullungdoensis sp. nov; ascomycete phylogeny, biodiversity surveys, *Eurotiales*, fungal ecology, *Penicillium jeongsukae* sp. nov.

1. Introduction

Aspergillus and *Penicillium* are fungal genera belonging to the ascomycete order *Eurotiales*. This order includes five families:—*Aspergillaceae, Elaphomycetaceae, Penicillaginaceae, Thermoascaceae,* and *Trichocomaceae*—that represent a total of 28 genera—*Acidotalaromyces,*

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The final published version (version of record) is available online at DOI:10.1016/j.funbio.2024.05.014. Please refer to any applicable publisher terms of use.

Ascospirella, Aspergillago, Aspergillus, Dendrosphaera, Elaphomyces, Evansstolkia, Hamigera, Leiothecium, Monascus, Paecilomyces, Penicillago, Penicilliopsis, Penicillium, Phialomyces, Pseudohamigera, Pseudopenicillium, Pseudotulostoma, Rasamsonia, Sagenomella, Sclerocleista, Talaromyces, Thermoascus, Thermomyces, Trichocoma, Warcupiella, Xerochrysium, and *Xeromyces* (Houbraken et al., 2020). These fungi are commonly found in air/bioaeorsols, soil, plants and animals, freshwater and seawater, the subsurface, and the built environment and many can grow at low water activities and low temperatures (Nguyen et al., 2021a; Nguyen et al., 2020; Houbraken et al., 2016; Hubka et al., 2014; Samson et al., 2014, 2010; Frisvad and Samson, 2004; Lee and Magan, 1999; Micheluz et al., 2021). Some of these fungi are economically important in the fields of biotechnology, food, and medicine (Hyde et al., 2019; Houbraken et al., 2014; Frisvad and Samson, 2004) and can make important contributions towards Sustainability Development Goals (Purchase, 2016; Noel et al., 2023).

The genus *Aspergillus* (family *Aspergillaceae*, order *Eurotiales*) comprises a large group of filamentous fungi subdivided into six subgenera, 27 sect. and 75 ser. by a phylogenetic approach, often supported by morphological, physiological, and/or extrolite profile data (Houbraken et al., 2020). Members of this genus are mainly saprophytes that can decompose organic materials that can be found in plants, fruits, foods, indoor environments, water, soil, air, and rock as well as on various other surfaces (Pangging et al., 2022; Sklenář et al., 2017; Chen et al., 2017; Hyde et al., 2016; Hubka et al., 2014; Samson et al., 2014; Perrone et al. 2008; Hallsworth, 2019). Species of *Aspergillus* can have positive or negative impacts on human activities. Indeed, some species of *Aspergillus* have economic importance in industry, agriculture and medicine (Meyer et al., 2011). For example, *A. terreus* is known for producing itaconic acid used in polymer manufacture, the cholesterol-lowering drug lovastatin (Wierckx et al., 2020; Boruta and Bizukojc, 2017), and the phytohormone gibberellin (produced by *A. fumigatus*) which improves plant growth (Khan et al., 2011). Furthermore, penicillixanthone A produced by *A. fumigatus* is reported to have effective anti-HIV-1 activity (Tan et al., 2019). *Aspergillus niger* is known to produce citric acid which is widely used in beverages, foods, detergents, cosmetics and pharmaceutical industries (Behera, 2020).

On the contrary, some species can produce mycotoxins, cause food spoilage, and sometimes cause infections in humans and animals (Kim et al., 2022; Ráduly et al., 2020; Frisvad et al., 2019; Mousavi et al., 2016; Magan, 2006; Paulussen et al., 2017). *Aspergillus ochraceus* belonging to sect. *Circumdati* produces the well-known the mycotoxin ochratoxin A (Visagie et al., 2014; Magan and Aldred, 2005; van der Merwe et al., 1965). Aflatoxins produced by *A. flavus* belonging to sect. *Flavi* are major toxic, carcinogenic secondary metabolites (Frisvad et al., 2019).

Currently, the *Aspergillus* genus consists of 480 species (Index Fungorum 2023; Sung et al., 2022; Wang and Zhuang, 2022; Houbraken et al., 2020), only two of which are registered in Korea according to the Index Fungorum database (http://www.indexfungorum.org). These are *Aspergillus cibarius* S.B. Hong & Samson and *Aspergillus koreanus* Hyang B. Lee, T.T. Duong & T.T. Nguyen.

The genus *Penicillium* (family *Aspergillaceae*, order *Eurotiales*), first described by Link in 1809, is one of the most common fungal genera. Species of *Penicillium* play an important role in diverse natural environments by degrading and cycling the organic matter in microbial necromass. They also have medicinal and industrial uses. For example, *Penicillium* species have been known to produce hydrolytic enzymes (Hamlyn et al., 1987). Important medical compounds, such as penicillin, are produced by *P. chrysogenum* (now reclassified as *P. rubens*) (Houbraken

et al., 2011). *Penicillium compactum* and *P. coprobium* produce pyripyropens that act to inhibit acyl-CoA:cholesterol acyltransferase (ACAT) (Frisvad et al., 2004). However, some species cause food spoilage, produce various mycotoxins, and also cause diseases in animals (including humans).

The genus *Penicillium* is made up of two subgenera, 32 sect., 89 ser. (Houbraken et al. 2020), with a total of more than 500 species (Index Fungorum 2023; Visagie and Yilmaz, 2022; Nguyen et al., 2021a; Houbraken et al., 2020). So far only these 10 species are registered in Korea according to the Index Fungorum database. These are *Penicillium acidum* Hyang B. Lee, T.T. Duong & T.T.T. Nguyen, *Penicillium aquadulcis* Hyang B. Lee & T.T.T. Nguyen, *Penicillium aquaticum* Hyang B. Lee, T.T. Duong & T.T.T. Nguyen, *Penicillium circulare* Hyang B. Lee, P.M. Kirk & T.T.T. Nguyen, *Penicillium daejeonium* S.H. Yu & H.K. Sang, *Penicillium geumsanense* Hyang B. Lee, P.M. Kirk & T.T.T. Nguyen, *Penicillium koreense* S.B. Hong, D.H. Kim & Y.H. You, *Penicillium mali-pumilae* Hyang B. Lee, T.T.T. Nguyen & Houbraken, *Penicillium psychrotrophicum* Hyang B. Lee, H.Y. Mun, J.C. Frisvad & Houbraken, and *Penicillium ulleungdoense* D.H. Choi & J.G. Kim.

Whereas some known *Aspergillus* and *Penicillium* species originate in Korea, there has been little systematic work to understand the diversity of these fungal taxa across environmental niches of this geographical region. Yet Korea has a peculiar climate with dry cold winters and humid hot summers, is coastal and subject to the North Pacific anticyclone, and has extensive natural environments but also much land disturbed by anthropogenic activity, all of which collectively creates a unique combination of niches for fungi. Here, we carried out a survey of *Aspergillus* and *Penicillium* species in different environmental settings in Korea. We discovered 11 species, including two new species, proposed as *Aspergillus ullungdoensis* sp. nov. and *Penicillium jeongsukae* sp. nov.

2. Materials and methods

2.1. Sampling and isolation

Soil samples, freshwater, dried persimmon fruits, and seeds of the green shiso plant (*Perilla frutescens*) were collected from different areas in Korea (Fig. 1). The soil was collected from Cheongyang, located in Chungnam Province, and from Ulleung Island, North Gyeongsang Province, South Korea. The freshwater sample was taken from a depth of 0–10 cm in lake located at Chonnam National University, Gwangju and from waterfall freshwater located in Jeju Island, Jeju Province, South Korea. Dried persimmon fruits purchased from a grocery store located at Cheongyang, Chungnam Province, and then stored at the room temperature. Finally, the seeds of *Per. frutescens* were obtained from plants growing on agricultural land in Jeongeup, Jeollabuk-do, South Korea.

Samples were individually stored in paper bags or sterile 50-mL conical tubes and brought back to the laboratory for fungal isolations. For the soil samples, 1 g of soil was added to 9 mL of sterile distilled water. Then, serial dilutions of the mixture (from 10 parts water to 1 part of sample mixture to 10 parts of water to 5 parts of sample mixture, by volume) were prepared. A 100-µL aliquot of the soil solution was dispensed onto potato dextrose agar (PDA; Becton, Dickinson and Co., Sparks, MD, USA) containing the antibiotic neomycin (50 mg/L). For freshwater samples, a 100-µL aliquot of water was dispensed onto PDA containing neomycin (50 mg/L). For dried

persimmon samples, spores were carefully examined under a stereomicroscope and transferred to PDA plates. The Petri plates were incubated at 25 °C for 3–7 days. To isolate fungi from seed samples, 7–10 seeds were directly plated on a moist blotter and incubated at 25 °C in the dark for 7–14 days. Fungal spores on the seeds were transferred to PDA (Difco™, Sparks, MD, USA) using a capillary tube under a stereomicroscope.

Pure isolates were obtained and stored on PDA slants at 4 °C and in 20% (v/v) glycerol at −80 °C at the Environmental Microbiology Laboratory Fungarium (Chonnam National University, Gwangju, Korea). The isolates were also deposited at the National Institute of Biological Resources (NIBR, Incheon, Korea) under the accession numbers NIBRFG0000510049, NIBRFGC000508992, NIBRFGC000508615, NIBRFG0000509597, and NIBRFGC000510099 at the Nakdonggang National Institute of Biological Resources (NNIBR, Sangju, Korea) under the accession number NNIBRFG31720 and NNIBRFG46695.

2.2. Fungal morphology studies

Fungal isolates were three-point inoculated onto Czapek yeast autolysate agar (CYA), malt extract agar (MEA), yeast extract sucrose agar (YES), oatmeal agar (OA), dichloran 18% (w/v) glycerol agar (DG18), CYA agar supplemented with 5% (w/v) NaCl (CYAS), and creatine sucrose agar (CREA). For each medium type and each isolate, Petri plates were inoculated at 20, 25, 30, and 37 °C for 7 days. Medium preparation, inoculation, and incubation were performed according to Samson et al. (2014). Colony characteristics such as colour, margin, diameter and texture were recorded after 7 days of incubation. Slides were prepared using 60% (v/v) lactic acid, and 96% (v/v) ethanol was used to remove excess conidia. An Olympus BX53 light microscope with differential interference contrast optics (Olympus, Tokyo, Japan) was used to obtain digital images of conidiophores, vesicles, metulaes, phialides, and conidia.

2.3. Extraction, PCR amplification, and sequencing of DNA

Total genomic DNA was extracted from fungal mycelia using the SolgTM Genomic DNA Preparation Kit (Solgent Co. Ltd., Daejeon, Korea) following the manufacturer's instructions. Marker genes were selected as follows: the internal transcribed spacer (ITS) region of the rRNA, *β*-tubulin (*benA*), calmodulin (*CaM*), and RNA polymerase II second largest subunit (*RPB2*). Amplification of the ITS region, *BenA*, *CaM* and *RPB2* was performed using the following conditions: initial denaturation 95 °C for 5 min, 30 cycles of denaturation (95 °C for 30 s), annealing [temperature according to the primer for 45 s, (Table S1)], and extension (72 °C for 45 s), final extension 72 °C for 7 min. The PCR products were purified using an Accuprep PCR Purification Kit (Bioneer Corp.). Sequencing was performed using the same primers on an ABI PRISM 3730XL Genetic Analyzer (Applied Biosystems, California, USA).

2.4. Molecular analyses

The raw forward and reverse reads were edited for ambiguous bases at both ends and assembled into contigs using SeqMan Version 7.1.0. The edited sequences were BLAST-searched against the NCBI GenBank nucleotide database (https://blast.ncbi.nlm.nih.gov/Blast.cgi) to identify their closest relatives. Sequences of previously published species of *Aspergillus* and *Penicillium* were

Journal Pre-proof

obtained from the GenBank sequence database (http://www. ncbi.nlm.nih.gov/). The sequences were aligned using MAFFT (http://mafft.cbrc.jp/alignment/server) (Katoh et al. 2019), then confirmed manually in MEGA v. 7 (Kumar et al. 2016). Data were converted from a FASTA format to NEXUS and PHYLIP formats using the online tool Alignment Transformation Environment (https://sing.ei.uvigo.es/ALTER/) (Glez-Peña et al. 2010). Phylogenetic reconstructions based on the maximum likelihood (ML) method were performed using RAxML-HPC2 on XSEDE via the CIPRES web portal (https://www.phylo.org/portal2), with a default GTR substitution matrix and 1,000 rapid bootstraps. Bayesian inference (BI) analyses were performed using MrBayes v. 3.2.6 (Ronquist et al. 2012). Four Markov chain Monte Carlo (MCMC) chains were run from a random starting tree for 5 million generations, and trees were sampled every 100th generation. The first 25% of the trees were removed as burn-in, and the remaining trees were used to calculate posterior probabilities. Final consensus trees were viewed using FigTree v. 1.3.1 (Rambaut 2009). Support values were provided at the branches (ML bootstrap support (BS) and BI posterior probability (PP). The newly obtained sequences were deposited in the GenBank sequence database under the accession numbers provided in Table S2.

3. Results

3.1. Fungal phylogeny

The phylogenetic relationship of the new *Aspergillus* species with accepted species in ser. *Spelaei* (sect. *Flavipedes*) was determined by analysing single and concatenated sequence datasets of four loci (ITS, *BenA*, *CaM*, and *RPB2*). The concatenated alignment consisted of 3084 characters (including alignment gaps): 751 (ITS), 537 (*BenA*), 783 (*CaM*), and 1013 (*RPB2*). Strains CNUFC UD06 and CNUFC UD07 clustered close to but separated from *A. lanuginosus*, *A. alboviridis* in the single (ITS, *BenA*, *CaM*, and *PRB2*) and combined phylogenies (Fig. 2 and Figs. S1–S4). The combined ITS, *BenA*, *CaM*, and *RPB2* phylogeny data indicated that strains CNUFC YB6 and CNUFC UDS2111, placed in ser. *Japonici* (sect. *Nigri*), were closely related to the ex-type strains of *A. uvarum* CBS 121591 and *A. aculeatinus* CBS 121060, respectively (Fig. S5). Strain CNUFC HSW10 grouped with ex-type strain of *A. aurantiacoflavus* CBS 141930 (Fig. S6) in ser. *Aspergillus* (sect. *Aspergillus*). Strain CNUFC 92316 clustered with the ex-type strain of *A. croceiaffinis* CCF 6035 in ser. *Unguium* (sect. *Nidulantes*) (Fig. S7). CNUFC JPS72 clustered with the ex-type strain of *A. pseudoviridinutans* NRRL 62904 in ser. *Viridinutantes* (sect. *Fumigati*) (Fig. S8).

The phylogenetic relationship of the new *Penicillium* species with accepted species in sect. *Charlesia* was determined by analysing single and concatenated sequence datasets of four loci (ITS, *BenA*, *CaM*, and *RPB2*). The concatenated alignment consisted of 2790 characters (including alignment gaps): 732 (ITS), 490 (*BenA*), 550 (*CaM*), and 1018 (*RPB2*). Strains CNUFC JSK815 and CNUFC JSK816 formed a well-supported independent branch (Fig. 3 and Figs. S9-S12). Strain CNUFC CY22014 clustered with the ex-type strain of *P. charlesii* (Fig. 3 and Figs. S9-S12). Strains CNUFC JYS28, CNUFC CY22324, and CNUFC U734 grouped with the ex-type strain of *P. ferraniaense* (sect. *Sclerotiorum*, ser. *Sclerotiorum*), *P. glaucoroseum* (sect. *Lanata-Divaricata*, ser. *Janthinella*), and *P. sajarovii* (sect. *Ramosum*, ser. *Raistrickiorum*), respectively (Figs. S13–15).

3.2. Taxonomy of the Korean isolates

Aspergillus aculeatinus Noonim, Frisvad, Varga, & Samson, Int. J. Syst. Evol. Microbiol. 58: 1733 (2008) (Fig. 4A-D)

Colony diameter, 7 days at 25 °C (mm): CYA 31–36; MEA, 61–65; YES 31–33.

Colony characteristics: CYA 25 °C, 7 days: Colonies moderately deep, sulcate; margins low, plane; mycelium white; texture velvety; sporulation absent to sparse; soluble pigments absent; exudates absent; reverse pale yellow. MEA 25 °C, 7 days: Colonies low, plane; mycelia white to light brown; texture floccose; sporulation moderately dense; soluble pigments absent; exudates absent; reverse light brown. YES 25 °C, 7 days: Colonies moderately deep, sulcate; margins low, plane; mycelium white; texture velvety; sporulation absent to sparse; soluble pigments absent; exudates absent; reverse pale yellow.

Micromorphology: Conidiophores uniseriate, smooth-walled, 197–476 × 7–10.5 µm. Vesicles globose, 31–48 µm. Phialides flask-shaped, covering the entire surface of the vesicle, 5.5–8 × 3–4 µm. Conidia subglobose to ellipsoidal, 3–4.5 × 2–3 µm.

Material examined: Republic of Korea, 2889-5 Saekdal-dong, Seogwipo-si, Jeju-do (33°14′52.7″N 126°24′48.3″E), from soil, April 2021, H.B. Lee (culture CNUFC UDS2111).

Notes: The morphological characteristics of the isolate were consistent with those of *A. aculeatinus*, as described by Noonim et al. (2008). However, CNUFC UDS2111 showed a slower growth in colony diameter on CYA compared to *A. aculeatinus* (CYA >85 mm; MEA 47–50 mm; YES >85 mm) (Noonim et al., 2008). *Aspergillus aculeatinus* was recorded from different samples of Arabica coffee beans, indoor air, vineyard soil, bark of Chinese yew (*Taxus chinensis* var. *mairei*), and sediments from the South China Sea in previous studies (Wu et al., 2021; Zhao et al., 2021; Qiao et al., 2017; Noonim et al., 2008), and from soil in the present study.

Aspergillus aurantiacoflavus Hubka, A.J. Chen, Jurjević & Samson, Studies in Mycology 88: 82 (2017) (Fig. 4E-I)

Colony diameter, 7 days at 25 °C (mm): CYA 8–10; MEA 16–20; YES 27–30

Colony characteristics: CYA 25 °C, 7 days: Colonies grew slowly, were pale yellow to light brown, flocculent, irregular, no soluble pigments, reverse pale to dark orange. On MEA 25 °C, 7 days: Colonies grew with moderate speed, were white to pale yellow, dense sporulation, no soluble pigments, reverse pale yellow. YES 25 °C, 7 days: Colonies grew slowly, texture was floccose, white to bright yellow at the center, sporulation moderate, no soluble pigments, reverse pale yellow.

Micromorphology: Conidiophores smooth stipes, hyaline, smooth-walled, 55–200 × 4–11 µm; Vesicles globose to subglobose, 9.5–25 µm in diameter; Phialides flask-shaped, 5.8–10.8 × 2.8–5.3 µm; Conidia tuberculate, globose, subglobose to ellipsoidal, 5.5–8.1 × 4–7.6 µm; Ascomata eurotium-like, cleistothecial, superficial, globose to subglobose, 60–78 µm.

Material examined: REPUBLIC OF KOREA, Jisan 2-dong, Dong-gu, Gwangju, Korea (35.152111°N, 126.950056°E), from freshwater (culture CNUFC HSW10).

Notes: CNUFC HSW10 has similar morphological characteristics with the previously described species of *A. aurantiacoflavus*. However, there were differences observed in the colony diameter in comparison to previously described by Chen et al. (2017) (CYA 8–9 mm vs 2–3 mm; MEA 16–20 mm vs 2–3 mm). *Aspergillus aurantiacoflavus* was isolated from a baby carrier backpack, rubber toys, and cake in previous works (Chen et al., 2016), and from freshwater in the present study.

Journal Pre-proof

Aspergillus croceiaffinis F. Sklenar, Jurjević, & Hubka, Mycologia 112(2): 359 (2020) (Fig. 4J-M)

Colony diameter, 7 days at 25 °C (mm): CYA 14–16; MEA, 10–13; YES 17–19.

Colony characteristics: CYA 25 °C, 7 days: Colonies moderately deep, plane; margins entire to slightly irregular; texture floccose; sporulation moderately dense; soluble pigments very weak; exudates absent; reverse brownish red. MEA 25 °C, 7 days: Colonies slightly raised at the center; margins low, plane; texture velvety, floccose at the center; sporulation moderately dense; soluble pigments absent; exudates absent; reverse deep orange. YES 25 °C, 7 days: Colonies slightly raised at the center, sulcate; margins low, plane; texture floccose; sporulation moderately dense; soluble pigments absent; exudates absent; reverse dark reddish brown.

Micromorphology: Conidiophores with smooth stipes, 96–147 × 2–3 µm. Vesicles pyriform, 6.5–11 µm in diameter. Metulae cylindrical, 7–8 × 2–2.5 µm. Phialides flask-shaped, 7–8(–10) × 2–2.5 µm. Conidia globose to subglobose, 2.5–3.5 µm in diameter.

Material examined: Republic of Korea, Nari, Buk-myeon, Ulleung-gun, Gyeongsangbuk-do (37°31′28.2″N 130°52′29.4″E), from soil (culture CNUFC 92316).

Notes: The morphological characteristics of the isolate were consistent with those of *A. croceiaffinis*, as described by Sklenář et al. (2020). However, the conidiophore of the isolate CNUFC 92316 was shorter than that of previously reported species [100–200 × 3–4 µm; Sklenář et al. (2020)]. In addition, differences in colony diameter were observed on CYA (10–12 mm) (Sklenář et al., 2020). *Aspergillus croceiaffinis* was isolated from a swab in a previous study (Sklenář et al., 2020), and from soil sample in the present study.

Aspergillus pseudoviridinutans Sugui, S.W. Peterson & Kwon-Chung, in Sugui, Peterson, Figat, Hansen, Samson, Mellado, Cuenca-Estrella & Kwon-Chung, *J. Clin. Microbiol.* 52(10): 3709 (2014) (Fig. 4N-Q)

Colony diameter, 7 days at 25 °C (mm): CYA agar, 36–40; MEA, 66–70; YES agar, 38–42.

Colony characteristics: CYA agar 25 °C, 7 days: Colonies centrally raised, surrounded by sunken depression; margins low, plane; texture floccose; mycelia white; sporulation absent to sparse; soluble pigments absent; exudates absent; reverse light yellow. MEA 25 °C, 7 days: Colonies moderately deep; margins low; mycelium white; texture floccose; sporulation absent to sparse; soluble pigments absent; exudates absent; reverse pale greenish yellow. YES agar 25 °C, 7 days: Colonies raised at the center, slightly sulcate; margins low; mycelia white; texture floccose; sporulation sparse; soluble pigments absent; exudates absent; reverse vivid yellow.

Micromorphology: Conidiophores with smooth stipes, 30–83.5 × 3–6 µm. Vesicles subglobose to flask-shaped, 7–14 µm in diameter. Phialides ampuliform, 3–5 × 1.5–2 µm. Conidia in long chains, globose, 2–3.5 µm in diameter.

Material examined: Republic of Korea, Jeongeup (35°31′34.0″N 126°49′03.7″E), Jeollabuk-do, from *Perilla frutescens* seed, October 2013 (culture CNUFC JPS72).

Notes: CNUFC JPS72 shared similar morphological characters with *A. pseudoviridinutans* (Sugui et al., 2014). However, conidia are bigger than those of *A. pseudoviridinutans* (1.75–2 µm) (Sugui et al., 2014). *Aspergillus pseudoviridinutans* was isolated from the mediastinal lymph node of a 14-year-old boy with chronic granulomatous disease, soil, a plant (*Pin. caribaea*), human lungs and eyes, and nostrils of parrots (*Amazona amazonica*) in

previous studies (Aneke et al., 2022; Langlois et al., 2021; Watanabe et al., 2021; Hubka et al., 2018; Talbot and Barrs, 2018), and from *Per. frutescens* seed in the current study.

Aspergillus ullungdoensis Hyang B. Lee **sp. nov.**

Index Fungorum: IF 900871; Fig. 4R-V and Table 1

Etymology: Refers to the isolation location, Ulleungdo (Korea) from where the species was first isolated.

Type specimen: Republic of Korea, Gitdaebong Peak, Ulleung Island, Korea (37°30'04" N 130°51'23"E), from rhizosphere soil, 30 April 2019 (holotype CNUFC HT19207; ex-type culture CNUFC UD06).

Colony diam, 7 days (mm): CYA25°C 23–26 mm, CYA20°C 21–23; CYA30°C 34–37; CYA37°C No growth; MEA25°C 27–29; YES25°C 32–35; OA25°C 23–27; CREA25°C 20–23.

Colony characteristics. CYA 25°C, 7 days: Colonies raised at center; texture velutinous; mycelium white to beige; soluble pigments absent; exudate absent; sporulation moderate; reverse pale yellow. YES 25°C, 7 days: Colonies radial sulcate; mycelium white; sporulation moderate; exudate absent; soluble pigments absent; reverse moderate yellowish brown. MEA 25°C, 7 days: Colonies low, plain; mycelium white; margins low; exudate absent; soluble pigments absent; reverse pale yellow. OA 25°C, 7 days: Colonies nearly circular, slightly raised at center; texture velutinous; sporulation strong; mycelia white then grayish pink; soluble pigments absent; exudates absent; reverse moderate yellowish brown. CREA 25 °C, 7 days: Colonies slow growth, acid not produced.

Micromorphology: Conidiophores biseriate. Stipes long, hyaline or light yellowish brown, smooth, not septate, 150–1200 × 4–6 µm. Vesicles hyaline, globose, subglobose to pyriform, 9–14 × 7.5–13 µm. Metulae 5.5–8(–9.5) × 3–3.5 µm. Phialides ampulliform, 5.5–9(–10) × 2.5–3.0 µm. Conidia globose, smooth, 2.5–3(–3.5) µm. Hülle cells present.

Additional material examined: Republic of Korea, Gitdaebong Peak, Ulleung Island, (37°30'04" N 130°51'23"E), from rhizosphere soil, 30 April 2019 (culture CNUFC UD07).

Notes: In a NCBI BLASTn search based on ITS, *BenA, CaM* and *RPB2* sequences, the closest match of *A. ullungdoensis* was 99.87% with *A. lanuginosus* NRRL 4610 (EF669604) (for ITS), 98.16% with *A. alboviridis* FMR 15175 (LT798936) (for *BenA*), 97.67% with *A. lanuginosus* NRRL 4610 (EF669562) (for *CaM*), and 99.21% with *A. movilensis* CCF 4410 (HG916718) (for *RPB2*).

A concatenated phylogenetic tree (ITS, *BenA*, *CaM*, and *RPB2*) revealed that *Aspergillus ullungdoensis* forms distinct lineages sister to *A. lanuginosus* but with low statistical support. However, *A. ullungdoensis* differs from *A. lanuginosus* by production Hülle cells. *Aspergillus ullungdoensis* grows faster on MEA at 25 °C than *A. lanuginosus* on MEA at 25 °C (20–21 mm). In additional, *A. lanuginosus* can grow at 37 °C, whereas *A. ullungdoensis* cannot grow at 37 °C.

Aspergillus uvarum G. Perrone, Varga, & Kozak, Int. J. Syst. Evol. Microbiol. 58(4): 1036 (2008) (Fig. 4W-Z)

Colony diameter, 7 days at 25 °C (mm): CYA 30–35; MEA, 30–48; YES 22–33.

Colony characteristics: CYA 25 °C, 7 days: Colonies raised at the center, sulcate; margins low, plane; mycelium white to pale olive; texture velvety; sporulation moderately dense; soluble pigments absent; exudates absent; reverse light yellow. MEA 25 °C, 7 days: Colonies low, sulcate; margins low, plane; mycelia white; texture velvety; sporulation absent to sparse; soluble

Journal Pre-proof

pigments absent; exudates absent; reverse pale yellow. YES 25 °C, 7 days: Colonies raised at the center, strongly sulcate; mycelium white to brown; texture velvety; sporulation dense; soluble pigments absent; exudates absent; reverse moderate yellow.

Micromorphology: Conidiophores with smooth stipes, 314–1480 × 5–12 μm. Vesicles globose to subglobose or elliptical, 35–70 μm in diameter. Phialides ampulliform, 6–10 × 2.5–5 μm. Conidia globose to subglobose, conspicuously spinose at maturity, 3.5–5 μm in diameter.

Material examined: Republic of Korea, Yongbong Reservoir, Yongbong-dong, Buk-gu, Gwangju (35°11′00.9″N 126°53′27.4″E), from freshwater, March 2022 (culture CNUFC YB6).

Notes: The morphological characteristics of the isolate CNUFC YB6 were similar to those of the previously described *A. uvarum* by Perrone et al. (2008). CNUFC YB6 showed slower growth on CYA and MEA in comparison to *A. uvarum* (Perrone et al., 2008) (CYA>90 mm; MEA>90 mm). *Aspergillus uvarum* was isolated from grapes, soil, food, indoor air, and a patient with onychomycosis in previous studies (Perrone et al., 2017; Qi et al., 2016; Zarei et al., 2015; Jurjević et al., 2012; Perrone et al., 2008), and from freshwater in the current study.

Penicillium charlesii G. Sm., Trans. Brit. Mycol. Soc. 18 (1): 90 (1933)

Descriptions & Illustrations: Raper and Thom (1949)

Material examined: Republic of Korea, in a home garden located on a hill in Kunryang-ri (36°26′16.2″N 126°46′04.6″E), Cheongyang-eup, Cheongyang, Chungnam Province, from a woodlouse (*Poc. scaber*), July 2022, H.B. Lee & J.S. Kim (culture CNUFC CY22014).

Notes: *Penicillium charlesii* has been isolated from mouldy corn (Zea mays L.), soil, surface water, greenhouses used for shiitake cultivation, cream, pasteurised milk, tomato, sputum, underground sites of Brestovská Cave, body surface and gastrointestinal tract of oribatid mites, corn kernels (Sun et al.,2021; Ahn et al., 2016; Raghuwanshi et al., 2013; Devi et al., 2012; Houbraken and Samson, 2011), and from *Porcellio scaber* in the current study. This species has been reported in UK, Italy, Japan, Portugal, Brazil, and Korea (Sun et al., 2021; Ahn et al., 2016).

Penicillium ferraniaense Houbraken & Di Piazza, Persoonia 46: 491 (2021) (Fig. 5A-E)

Colony diameter, 7 days at 25 °C (mm): CYA 19–24; MEA, 25–27; YES 19–25.

Colony characteristics: CYA 25 °C, 7 days: Colonies raised at the center, radially sulcate; mycelia white; sporulation strong; soluble pigments absent; exudates absent; reverse yellowish white to yellowish gray. MEA 25 °C, 7 days: Colonies radially sulcate, concentric rings at the center; mycelia white; texture velvety; sporulation moderate; soluble pigments absent; exudates absent; reverse strong orange at the center, pale yellow at the edge. YES 25 °C, 7 days: Colonies sulcate; texture velvety; sporulation strong; soluble pigments absent; exudates absent; reverse pale yellow.

Micromorphology: Conidiophores monoverticillate, minor portion biverticillate; stipes smooth-walled, 55–80 × 2.5–3.5 μm. Phialides 5–7 per metula, ampulliform, 7–11 × 2–3 μm. Conidia smooth-walled, globose to subglobose, 2.5–3.5 μm.

Material examined: Republic of Korea, Chonnam National University, Yongbong-dong, Buk-gu, Gwangju (35°10′28.8″N 126°54′32.3″E), from the soil, February 2022 (culture CNUFC JYS28).

Notes: The morphological descriptions of CNUFC JYS28 matched well with the previously described species of *P. ferraniaense* (Crous et al., 2021). CNUFC JYS28 differed from *P. ferraniaense* in colony diameter on CYA (19–24 mm vs 25–28 mm). *Penicillium ferraniaense*

was isolated from anaerobic digestate (compost) in a previous study (Crous et al., 2021), and from soil in the present study.

Penicillium glaucoroseum Demelius, Verhandlungen der Zoologisch-Botanischen Gesellschaft Wien 72: 2 (1923) (Fig. 5F-I)

Colony diameter, 7 days at 25 °C (mm): CYA 21–23; MEA 32–37; YES 19–23.

Colony characteristics: CYA 25 °C, 7 days: Colonies slightly radially sulcate; margins low, narrow; mycelia white; texture floccose; sporulation sparse; soluble pigments absent; exudates absent; reverse yellowish white. MEA 25 °C, 7 days: Colonies raised at the center; margins moderately wide; mycelia white; texture floccose; sporulation sparse; reverse pale greenish yellow. YES 25 °C, 7 days: Colonies radially sulcate, sunken centrally; margins low, wide; mycelia white; texture velvety; soluble pigments absent; exudates absent; reverse pale yellow.

Micromorphology: Conidiophores biverticillate, few terverticillate; stipes rough walled, 105–170 × 2.0–3.0 µm. Phialides 3–4 per metula, ampulliform, 7–12 × 2–2.5 µm. Conidia smooth-walled, globose, 2.5–3.5 µm. Ascomata not observed.

Material examined: Republic of Korea, Janggok-ri (36°25′00.6″N 126°51′31.8″E), Daechi-myeon, Cheongyang, Chungnam Province, from soil, February 2022, H.B. Lee (culture CNUFC CY22324).

Notes: The observed characteristics were mostly similar to those previously described for *P. glaucoroseum* (Visagie et al. 2015). However, isolate CNUFC CY22324 grew slow than *P. glaucoroseum* (CYA 47–49 mm; MEA 54–56 mm; YES 50–53 mm) (Visagie et al. 2015). *Penicillium glaucoroseum* was isolated from live roots of the coniferous tree kauri (*Agathis australis*), soil, worm excreta, farm sludge, and activated human sludge in previous studies (Taghavi et al., 2021; Demelius et al., 1922), and from soil in the current study.

Penicillium jeongsukae Hyang B. Lee & T.T.T. Nguyen **sp. nov.**

Index Fungorum: IF 558124; Fig. 5J-N and Table 2

Etymology: jeongsukii, named after Jeong Suk Kim. This species was isolated from dry persimmons collected by her.

Type specimen: Republic of Korea, Kunryang-ri (36°26′16.2″N 126°46′04.6″E), Cheongyang-eup, Cheongyang, Chungnam Province, from Korean dried persimmon, October 25, 2021, J.S. Kim (holotype CNUFC HT2102; ex-type culture CNUFC JSK815).

Colony diameter, 7 days (mm): CYA 25 °C 35–37, CYA20 °C 22–24, CYA30 °C 40–43, CYA 37 °C 30–34, CYA39 °C no growth; MEA25 °C, 40–42; YES25 °C 31–33; OA25 °C 36–38; CREA 25 °C 24–26; CYAS 25 °C 15–18; DG18 25 °C 22–25.

Colony characteristics: CYA 25 °C, 7 days: Colonies slightly radially sulcate; margins entire; texture floccose; mycelium white; soluble pigments absent; exudates absent; sporulation moderate; conidia green; reverse grayish. YES 25 °C, 7 days: Colonies radially sulcate; mycelium white; sporulation poor, exudates absent; soluble pigments absent; reverse pale yellow. MEA 25 °C, 7 days: Sporulation strong; colonies low, plane; mycelium white; margins low; exudates absent; soluble pigments absent; reverse yellowish white. CYA 25 °C, 7 days: Colonies raised centrally; margins entire; texture floccose; sporulation moderate; mycelium white; soluble pigments absent; exudates absent; reverse light yellow. OA 25 °C, 7 days: Colonies low, plane;

margins entire; mycelia white; sporulation moderate; soluble pigments absent; exudates absent; reverse light grayish. CREA 25 °C, 7 days: Colonies showing good growth; acid produced.

Micromorphology: Sclerotia absent. Conidiophores 17.5–40(–45) × 2–3.5 µm, with smooth-walled stipes, monoverticillate. Vesicles (2.5–)3.0–6.0 µm. Phialides ampulliform, 3–15 per vesicle, 6.5–10 × 2.0–3.0 µm. Conidia in long chains, smooth-walled, predominantly broadly ellipsoidal or cylindrical, 2.5–3.5 × (1.5–)2–3 µm.

Additional material examined: Republic of Korea, Kunryang-ri (36°26′16.2″N126°46′04.6″E), Cheongyang-eup, Cheongyang, Chungnam Province, from Korean dried persimmon, October 25, 2021, J.S. Kim (culture CNUFC JSK816).

Notes: In a NCBI BLASTn search based on ITS, *BenA, CaM* and *RPB2* sequences, the closest match of *P. jeongsukae* was 99.15% with *P. lunae* CMV006E6 (MK450725) (for ITS), 96.20% with *P. cuddlyae* CMV016A6 (MK951835) (for *BenA*), 96.43% with *P. cuddlyae* CMV016A6 (MK951908) (for *CaM*), and 98.06% with *P. chermesinum* CBS 231.81 (MN969111) (for *RPB2*).

Phylogenetic analysis revealed that the new species was distinct and formed a different clade in sect. *Charlesia*. Good growth on Czapek yeast autolysate (CYA) agar at 37 °C, fewer phialides per vesicle, shorter conidiophores, and predominantly broadly ellipsoidal or cylindrical conidia distinguished the new species from other known species.

Penicillium sajarovii Quintan., Av. Aliment. Majora Anim.: 539 (1981) (Fig. 5O-R)

Colony diameter, 7 days at 25 °C (mm): CYA 26–28; MEA, 26–29; YES 22–24.

Colony characteristics: CYA 25 °C, 7 days: Colonies plane to slightly sulcate; texture velvety; texture floccose; mycelium white; sporulation dense; soluble pigments absent; exudates absent; reverse light yellowish brown at the center. MEA 25 °C, 7 days: Colonies plane; texture floccose; mycelium white; sporulation moderate; soluble pigments absent; exudates absent; reverse light yellow. MEA 25 °C, 7 days: Colonies sulcate, slightly raised at the center; mycelium white; sporulation sparse; soluble pigments absent; exudates absent; reverse vivid yellow.

Micromorphology: Conidiophores biverticillate, 130–181 × 2.0–3.5 µm. Phialides ampulliform, 5.5–8.5 × 2.0–3 µm. Conidia globose to subglobse, 2.5–3.0 µm.

Material examined: Republic of Korea, Gitdaebong Peak, Ulleung Island, Korea (37°30′04″N 130°51′23″E), from soil, May 01, 2019 (culture CNUFC U734).

Penicillium sajarovii was isolated from rye (*Secale cereale*) as endophyte, the Himalayan herb *Arnebia euchroma*, and the rhizosphere of diseased white lupin (*Lupinus albus*) in previous studies (Jain et al. 2021; Quintanilla 1981), and from soil in the current study.

4. Discussion

Saprotrophic fungi would be expected in humic soils, but perhaps more surprisingly were also present in freshwater and on seeds. Dried fruits represent a high-sugar, low water-activity habitat for fungi that favours the growth of only xerotolerant and xerophilic species (Lievens et al., 2015). The *Eurotiales* is known to contain a considerable number of very xerotolerant species (see Stevenson et al., 2015; 2017) so it is unsurprising that we isolated such fungi from dried persimmon fruits in the current study. Fungal saprotrophs not only degrade dead plant matter, but in some cases degrade insect exoskeletons, fish scales, and animal hair (Christian et al., 2011).

Interesting, freshwater and seawater can also be a source of saprotrophic fungi and can even act to preserve them long-term in an inactive condition (Hallsworth, 2022; Jones et al., 2014). So far, a total of 3,870 species were reported from freshwater, with only 76 species belonging to the order *Eurotiales* (Calabon et al., 2022; Nguyen et al., 2021b). Recently, Torres-Garcia et al. (2022) reported eight *Penicillium* species from Spanish freshwater sediments so the number of known *Eurotiales* species from freshwater has increased to at least 84. Likely, a large number of species are yet to be discovered in freshwater habitats. Whereas we had expected to identify diverse *Eurotiales* fungi in the current study, the discovery of two new species was unexpected. Given the various crises currently facing humanity and the biosphere (habitat destruction including deforestation, wildfires, climate change, biodiversity loss, etc), we believe that it is as important as ever to continue to document species not only for the scientific value, but also for their potential worth in biotechnological solutions to the various problems humanity is facing (e.g., Purchase, 2016; Timmis and Ramos 2021; Anand et al., 2023).

The new *Aspergillus* species identified in this study was located in sect. *Flavipedes* and ser. *Spelaei* and was found to be related to *A. alboviridis*, *A. lanuginosus* and *A. movilensis* in individual and combined phylogenetic analyses with ITS, *BenA*, *CaM* and *RPB2* sequences. Production of Hülle cells, and the inability to grow on CYA at 37 ◦C can be used to distinguish between *A. ullungdoensis* and *A. lanuginosus* (that does not produce Hülle cells and can grow on CYA at 37 ◦C). In addition, *A. ullungdoensis* grows faster than *A. lanuginosus* on MEA at 25 °C (20–21 mm). Like *A. ullungdoensis*, *A. alboviridis*, and *A. movilensis* also produce Hülle cells, but conidiophores of *A. ullungdoensis* are longer than *A. alboviridis* [100–200(–550) μm] and *A. movilensis* (usually < 400 μm) (Sklenář et al., 2021). *Aspergillus ullungdoensis* grows faster than *A. alboviridis* on MEA (18–23 mm) and OA (18–19 mm) at 25°C (Sklenář et al., 2021). *Aspergillus alboviridis* and *A. movilensis* grow at 37 °C, but *A. ullungdoensis* cannot grow at 37 °C. Sklenář et al. (2021) revealed that length of stipes can be considered taxonomically important and may be used to distinguish *A. alboviridis*, *A. movilensis* and *A. lanuginosus*. Our data demonstrate that *A. ullungdoensis* is morphologically and genetically different from *A. lanuginosus*, *A. alboviridis*, and *A. movilensis*. Therefore, it is a newly described species.

Species in sect. *Flavipedes* occurs globally in soil, indoor or outdoor environment, as endophytes, food contaminants, dung of herbivores, cave environments including bat guano-contaminated soils, and clinical samples (Sklenář et al., 2021; Visagie et al., 2021). Sect. *Flavipedes* also contains species that are widely used in biotechnology industries (Sklenář et al., 2021; Frisvad and Larsen, 2015). *Aspergillus iizukae* is known to produce oxidative enzymes with an industrial application (Noman et al., 2020) and the potential antiviral activity compounds against influenza virus (H1N1), herpes simplex virus types 1 (HSV-1) and 2 (HSV-2) (Kang et al., 2018). Metabolites 3-hydroxy-2′,4,4′,6′-tetramethoxychalocone, 7,3,4,5′-tetramethoxyflavanone, isovitexin, and amodiaquine produced by *A. flavipes* were found effective antimicrobial against *Phytophthora parasitica* (El-Sayed and Ali, 2020). Therefore, new *Aspergillus* species could be promising candidate for product discovery.

The new *Penicillium* species belongs to sect. *Charlesia* and ser. *Indica* based on multigene phylogeny data using ITS, *BenA*, *CaM*, and *RPB2* sequences. ITS, *BenA*, *CaM*, and *RPB2* data revealed that the novel species was distinct from previously described *Penicillium* species. The data of independent and combined sequence analyses of the four loci revealed a close relationship among *P. chermesinum*, *P. indicum*, *P. lunae*, and *P. cuddlyae*. *Penicillium jeongsukae* shares several similarities with *P. chermesinum*, *P. cuddlyae*, *P. indicum*, and *P. lunae*, such as the

production of monoverticillate sporangiophores and growth on CYA agar at 37 °C (except for *P. lunae*) (Crous et al., 2019a, 2019b). However, *P. jeongsukae* differs from *P. lunae* (MEA: 25–26 mm) and *P. cuddlyae* (21–23 mm) by its capacity for rapid growth on CYA agar, MEA, and OA (Crous et al. 2019a, 2019b). Species in sect. *Charlesia* have been isolated worldwide from various habitats, such as indoor air, banana fruit, tomato, garlic, rice, beans, dried blue peas, spoiled cereals, other dry foods, rhizosphere of peanut plants, stem and root of mangrove tree, soil, greenhouses used for shiitake cultivation, cream, pasteurised milk, sputum, body surface and gastrointestinal tract of mites(*Heritiera littoralis*), and inner tissues of the marine macroalga *Pterocladiella tenuis* (Crous et al., 2019a, 2019b; Su et al., 2018; Peterson et al., 2017; Yilmaz et al., 2016). The current study is the first report of the isolation of *P. charlesii* from the woodlouse *Por. scaber*. Members in sect. *Charlesia* are known to. Their ability to produce enzymes that are efficient and potentially useful for anthropogenic purposes make them ideal candidates for biotechnological applications (Raghuwanshi et al., 2013; Batra and Saxena, 2005; Clutterbuck et al., 1935). *Penicillium charlesii* exhibits exo-α-mannosidase, β-glucosidase, protease, and xylanase activities (Yoon et al., 2007; Abbas et al., 1989; Preston et al., 1973). It also exhibits antinematodal and antiparasitic activities (Ondeyka et al., 1990). Moreover, it produces various organic acids, such as carolic acid, itaconic acid, I-γ-methyltetronic acid, ramigenic acid, tartaric acid, tetronic acid, and verticillic acid. Citreoviridin is another known mycotoxin produced by this strain (Preston et al., 1973; Klatt et al., 1969; Bentley et al., 1962; Lybing et al., 1958; Clutterbuck et al., 1935). *Penicillium costaricense* produces extrolites, such as andrastin A and rastin C (Vega et al., 2006) and *Penicillium coffeae* produces δ-lactone (penicoffeazine A) and isocoumarin derivatives (penicoffrazins B and C) (Cao et al., 2019; Peterson et al., 2005). *Penicillium coffeae* was found to tolerate up to 500 mM of arsenate (Bhargavi and Savitha, 2014) which could be a potential for bioremediation applications. Therefore, the new *Penicillium* species isolated in this study could be a potentially useful source for producing secondary metabolites.

Fungi of the genera *Aspergillus* and *Penicillium* are involved in humanitarian needs in both positive and negative ways given their various roles in relation to agricultural economics, the food sector, and biotechnology. Some species are plant pathogens, and others can cause food spoilage, so have implications for food security. Some species are pathogenic to humans, and their pathogenicity may become worse as the human population increases, malnutrition is commonplace, and modern medical practices result in immuno-compromised patients. Understanding fungal ecology (including mycotoxin production) is also key to keeping water safe and preventing the proliferation of fungi harmful to human health in the built environment, and fungi are key to soil- and plant health (Timmis and Ramos, 2021). In relation to climate change, within the food-supply chain mycotoxins may become prevalent as weather patterns and conditions for the transport and storage of foods and feeds become increasingly difficult to predict and control (Medina et al., 2015) but microbes can be utilised for the common good including for bioprospecting and biotechnological developments to mitigate the various humanitarian crises (see also Timmis et al., 2024; Timmis and Hallsworth, 2024;). We are in favour of a systematic and coordinated international effort to isolate fungi from local environments within regions world-wide to discover more as-yet-undescribed fungi, bring them into accessible culture collections, and assess them for their biotechnological potential.

Acknowledgments

This work was supported by the Survey and Discovery of Indigenous Fungal Species of Korea Project (NIBR202203204) funded by National Institute of Biological Resources (NIBR) of the Ministry of Environment (MOE), and by the Discovery of Fungi from Freshwater and their Collection for Fungaria Project (NNIBR202201206) funded by Nakdonggang National Institute of Biological Resources (NNIBR) of the Ministry of Environment (Korea). This study was also supported by the Ministry of Science and ICT (Korea) (2022M3H9A1082984). The paper is written in memory of Prof. Naresh Magan at Cranfield University (England, UK) who passed away on April 20, 2023 (Hallsworth et al., 2023).

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Table 1. Morphological characteristics of *Aspergillus ullungdoensis* sp. nov. compared with those of the reference strains *A. lanuginosus*, *A. alboviridis*, and *A. movilensis*

Characteristics	***A. ullungdoensis*** [(a)]	***A. lanuginosus*** [(b)]	***A. alboviridis*** [(b)]	***A. movilensis*** [(b,c)]
Size after 7 days at 25 °C (diameter)	23–26 mm on CYA 27–29 mm on MEA 23–27 mm on OA	26–27 mm on CYA 20–21 mm on MEA 25–26 mm on OA	19–21 mm on CYA 18–23 mm on MEA 18–19 mm on OA	22–25 mm on CYA 25–30 mm on MEA NI
Size after 7 days at 37 °C	No growth	Growth at 37 °C	Growth at 37 °C	10-17 mm
Stipes	Hyaline or light yellowish brown, smooth, not septate, 150–1,200 × 4–6 µm	Smooth, hyaline or light brown, 600–1,100 µm × 3.5–4.5 µm	Smooth, hyaline or brown, 120–200(–550) × 4.5–6(–8) µm	Usually < 400 µm length, 3.5–6 µm width

Vesicle	Globose, subglobose to pyriform, hyaline, 9–14 × 7.5–13 µm	Subglobose to pyriform, hyaline, 10–12 µm	Globose to subglobose, hyaline, 12–16(–20) µm	Pyriform, 5-16 µm
Metulae	5.5–8(–9.5) × 3–3.5 µm	Cylindrical, hyaline, 5–5.5 µm	Cylindrical, hyaline, 6–7(–9) µm	4–8(–10) µm length
Phialides	Ampulliform, 5.5–9(–10) × 2.5–3.0 µm l	Flask-shaped, hyaline, 5.5–6.5 µm	Flask-shaped, hyaline, 6–7.5(–8.5) µm	4–7 µm length
Conidia	Globose, smooth, 2.5–3(–3.5) µm	Globose to subglobose, smooth, hyaline, 2.5–3 × 2–2.5 µm	Globose to subglobose, smooth, hyaline, 2.5–3.5 × 2–3 µm	Globose, smooth, 2.5–3.3(–3.5) µm
Hülle cells	Present	Present	Absent	Present

[(a)] The present study; [(b)] From the description by Sklenář et al. (2021); [(c)] From the description by Hubka et al. (2015)

Table 2. Morphological characteristics of *Penicillium jeongsukae* sp. nov. compared with those of the reference strains *P. cuddlyae*, *P. chermesinum*, *P. lunae*, and *P. indicum*

Characteristics	***P. jeongsukae*** [(a)]	***P. cuddlyae*** [(b)]	***P. chermesinum*** [(c)]	***P. lunae*** [(d)]	***P. indicum*** [(e)]
Size after 7 days at 25 °C (diameter)	35–37 mm on CYA 31–33 mm on YES 24–26 mm on CREA 40–42 mm on MEA 36–38 mm on OA 21–23 mm on DG18 15–18 mm on CYAS	24–26 mm on CYA 30–32 mm on YES 12–14 mm on CREA 21–23 mm on MEA 28–30 mm on OA 24–25 mm on DG18 19–20 mm on CYAS	40–50 mm on CYA NI on YES NI on CREA 50–55 mm on MEA NI on OA	34–36 mm on CYA 34–35 mm on YES NI on CREA 25–26 mm on MEA 28 mm on OA 24–25 mm on DG18 33–35 mm on CYAS	50 mm on CYA [(*)] NI on YES NI on CREA 55 mm on MEA [(*)] 50 mm on OA [(*)]
Size after 7 days at 37 °C	30–34 mm on CYA	19–21 mm on CYA	Growth at 37 °C	No growth	Growth at 37 °C
Conidiophores	Monoverticillate, stipes smooth, 17.5–40(–45) × 2–3.5 µm	Monoverticillate, stipes smooth-walled, 20–45 × 2–3 µm	Strictly monoverticillate, stipes smooth-walled, mostly 20–40(–50) × 2.0–2.5 µm	Monoverticillate, minor proportion biverticillate; stipes smooth-walled, 13–60 × 2–3(–3.5) µm	Monoverticillate, stipes smooth or finely roughened, 50–100 × 2.0–2.5 µm
Phialides	Ampulliform, 3–15 per vesicle, 6.5–10 × 2–3 µm.	Ampulliform, 10–20 per vesicle, 8–10 × 2–3 µm	Sterigmata crowded, often incurved, 10–15 per vesicle, 6–8 × 2.0–2.5 µm	Ampulliform, 10–20 per vesicle, (7.5–)8–10 × 2–3 µm	Sterigmata mostly in compact clusters, parallel, up to 12 or 15 per vesicle, 6.8–8.4 × 2.0–2.5 µm
Vesicle	(2.5–)3.0–5.5 µm wide	5–6 µm wide	4.0–4.5 µm wide	5–7 µm wide	3.4–6.5 µm wide

Metulae	NA	NA	NA	Two when present, 18–30 × 2–3(–3.5) µm	NA
Conidia	Smooth-walled, predominantly broadly ellipsoidal or cylindrical, 2.5–3.5 × (1.5–)2–3 µm	Smooth-walled, ellipsoid or cylindrical, 2–3 × 2.5–2 µm	Smooth, elliptical, 2.0–2.5 × 1.5–2.0 µm	Smooth-walled, subglobose to broadly ellipsoid, 2–3(–3.5) × 1.5–2(–2.5) µm	Smooth, elliptical, globose to subglobose, 1.7–2.5(–3.0) µm

[(a)] The present study. [(b)] From the description by Crous et al. (2019a); [(c)] From the description by Raper and Thom (1949); [(d)] From the description by Crous et al. (2019b). [(e)] From the description by Sandhu and Sandhu (1963). NA: not available. [(*)] 10 days.

Figure legends

Fig. 1. Location of sample collection sites within Korea (A). Samples were collected from different sites including soil at Cheongyang (B), water from waterfall freshwater at Jeju Island (C), water from a freshwater lake at Gwangju (D) and agricultural soil Ulleung Island (E).

Fig. 2. Phylogram generated from maximum likelihood analysis based on combined ITS, *BenA*, *CaM*, and *RPB2* sequences data of *Aspergillus* in ser. *Spelaei*. The numbers above or below branches represent maximum likelihood bootstrap percentages (left) and Bayesian posterior probabilities (right). Bootstrap values ≥ 70% and Bayesian posterior probabilities ≥ 0.95 are indicated above or below branches. Bootstrap values lower than 0.95 and 70% are marked with "-". *Aspergillus urmiensis* CBS 139558 was used as an outgroup. The newly generated sequence is indicated in blue. [T] = ex-type.

Fig. 3. Phylogram generated from maximum likelihood analysis based on combined ITS, *BenA*, *CaM*, and *RPB2* sequences data of *Penicillium* in sect. *Charlesia*. The numbers above or below branches represent maximum likelihood bootstrap percentages (left) and Bayesian posterior probabilities (right). Bootstrap values ≥ 70% and Bayesian posterior probabilities ≥ 0.95 are indicated above or below branches. Bootstrap values lower than 0.95 and 70% are marked with "-". *Penicillium chrysogenum* CBS 306.48 was used as an outgroup. The newly generated sequence is indicated in blue. [T] = ex-type.

Fig. 4. Morphology of species in this study. (A, E, J, N, R, W) Colony morphology (top row, left to right: CYA, MEA, YES); (bottom row, left to right: reverse colonies on CYA, MEA, YES). (B, C, F, G, K, L, O, P, S, T, X, Y) Conidiophores. (D, H, M, Q, U, Z) Conidia. (I) Ascomata. (V) Hülle cells. Scale bars = 10 µm.

Fig. 5. Morphology of species in this study. (A, F, J, O) Colony morphology (top row, left to right: CYA, MEA, YES); (bottom row, left to right: reverse colonies on CYA, MEA, YES). (B-D, G, H, K-M, P, Q) Conidiophores. (E, I, N, R) Conidia. Scale bars = 10 µm.

Fig. 1

Journal Pre-proof

Fig. 2

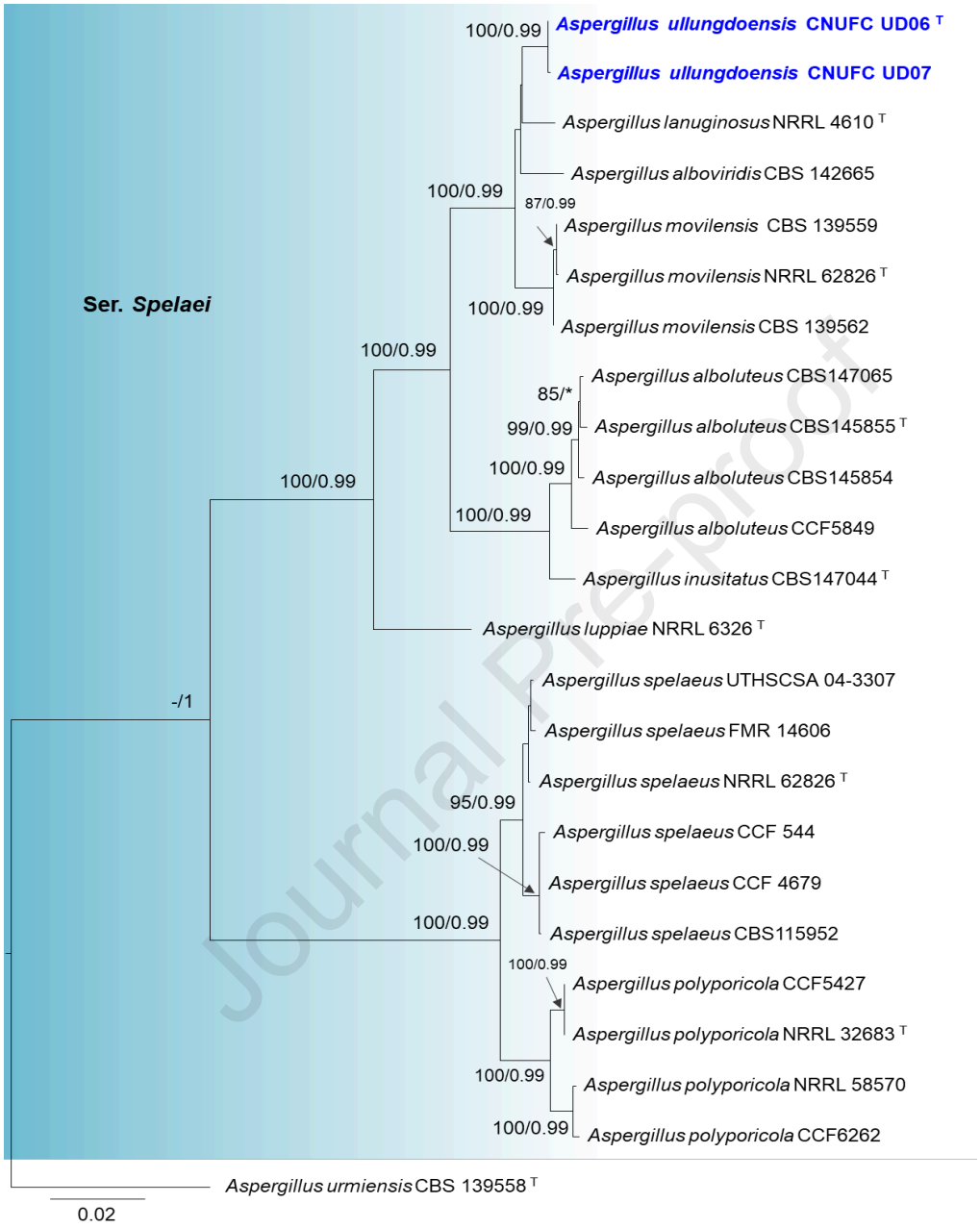

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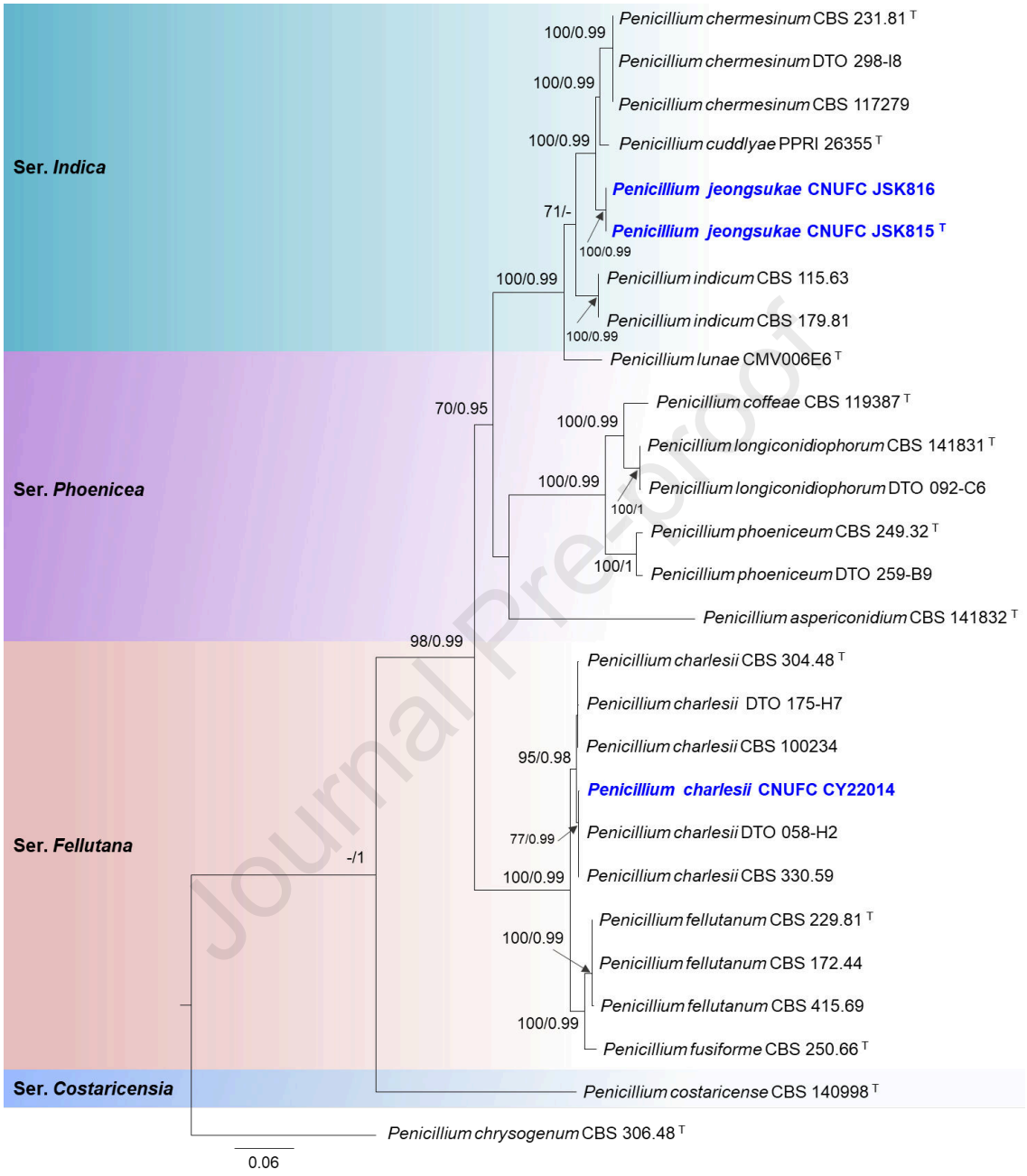
Fig. 3



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1 **Fig. 4**

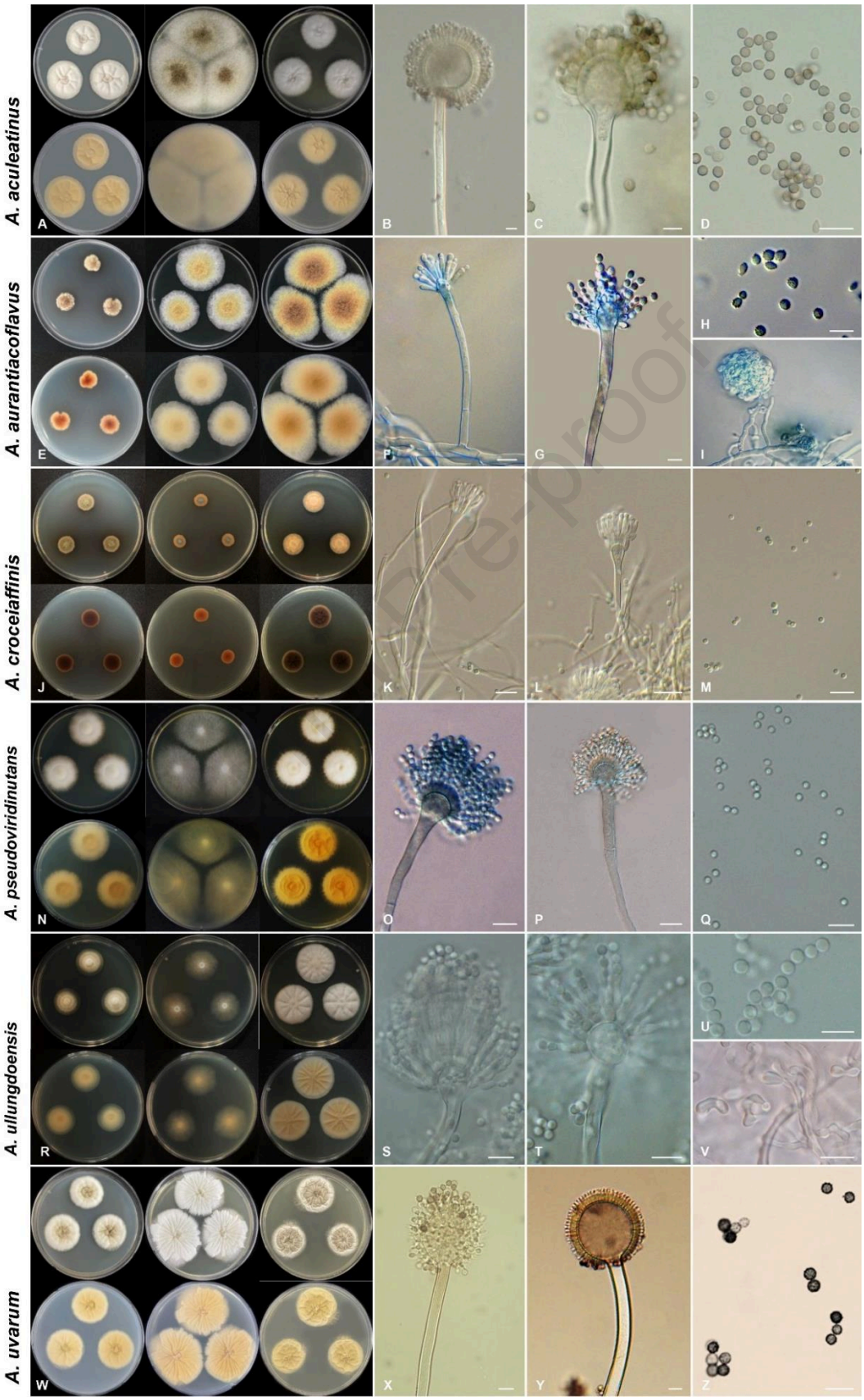
A. aculeatinus
A. aurantiacoflavus
A. croceiaffinis
A. pseudoviridinutans
A. ullungdoensis
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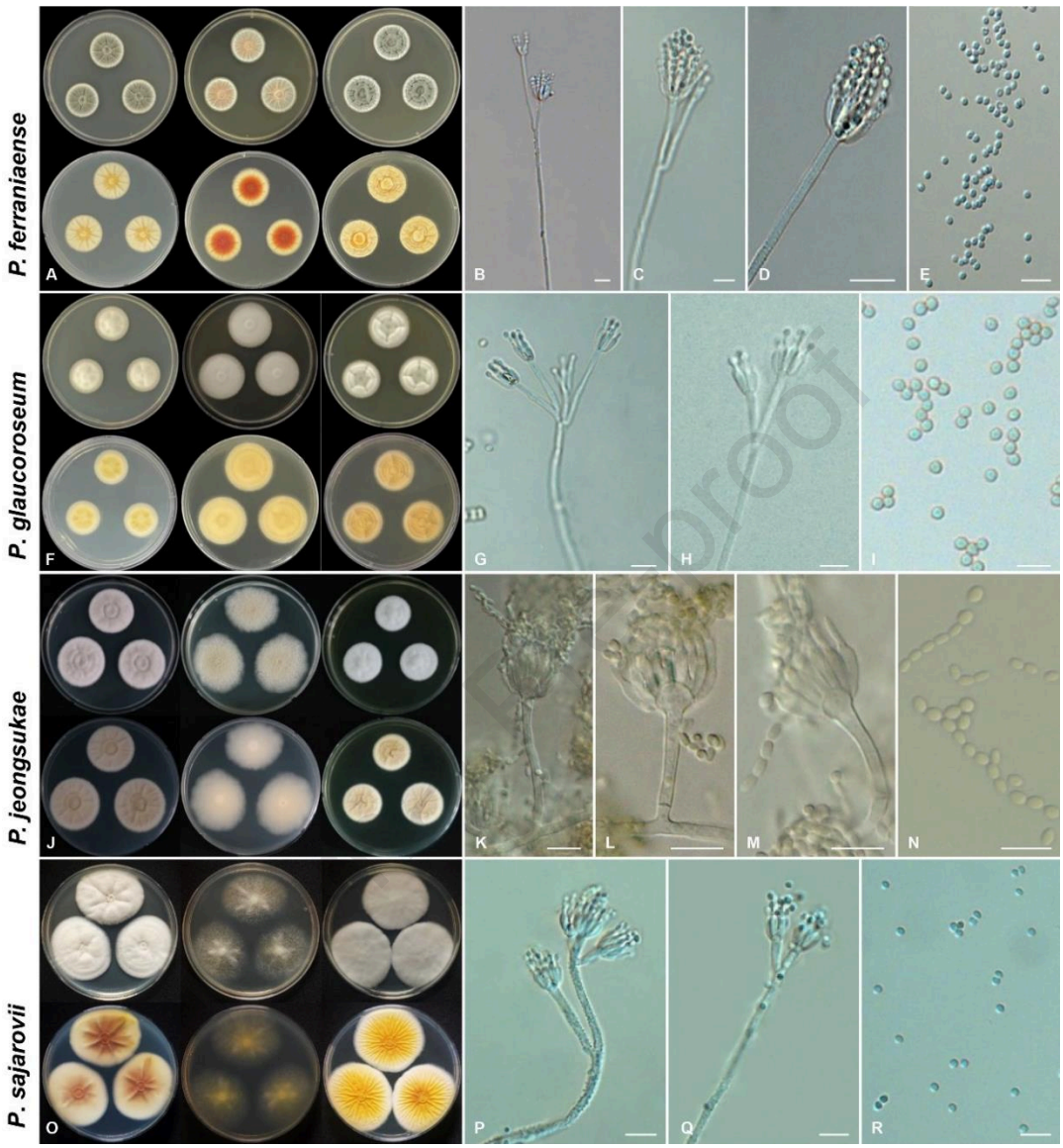
Fig. 5


P. ferraniaense
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Dear Chief-Editor,

I am pleased to submit the manuscript to renowned Fungal Biology as special issue. This manuscript has not been published elsewhere. All the authors have approved the manuscript and agree with this submission. There were no any interest conflicts among all authors.

The authors have no known competing financial interests or personal relationships that could affect the work reported in this paper.

Best regards,

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Aspergillus ullungdoensis sp. nov., Penicillium jeongsukae sp. nov., and other fungi from Korea

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2024-12-01
Elsevier
https://doi.org/10.1016/j.funbio.2024.05.014
Lee HB, Nguyen TT, Noh SJ, et al., (2024) Aspergillus ullungdoensis sp. nov., Penicillium jeongsukae sp. nov., and other fungi from Korea. Fungal Biology, Volume 128, Issue 8, Part B, December 2024, pp. 2479-2492

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