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A New Species and Five New Records of Talaromyces (Eurotiales, Aspergillaceae) Belonging to Section Talaromyces in Korea

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ABSTRACT

Talaromyces is a genus within the phylum Ascomycota (class Eurotiomycetes, order Eurotiales, family Trichocomaceae). Many species in this genus are known to produce diverse secondary metabolites with great potential for agricultural, medical, and pharmaceutical applications. During a survey on fungal diversity in the genus Talaromyces in Korea, six strains were isolated from soil, indoor air, and freshwater environments. Based on morphological, physiological, and multi-locus (ITS, BenA, CaM, and RPB2) phylogenetic analyses, we identified five previously unrecorded species in Korea (T. brevis, T. fusiformis, T. muroii, T. ruber, and T. soli) and a new species (T. echinulatus sp. nov.) belonging to section Talaromyces. Herein, detailed descriptions, illustrations, and phylogenetic tree are provided.

ARTICLE HISTORY

Received 28 August 2023 Revised 25 September 2023 Accepted 26 September 2023

KEYWORDS

Eurotiales; freshwater; indoor air; phylogeny; soil; taxonomy

1. Introduction

Talaromyces is one of the largest fungal genera in the family Trichocomaceae. It consists of 171 accepted species [1]. So far, accepted eight sections in Talaromyces include Bacillispori, Helici, Islandici, Purpurei, Subinflati, Talaromyces, Trachyspermi, and *Tenues* [2,3].

Section Talaromyces which was first introduced by Stolk and Samson in 1972 [4] represents the largest section containing more than 87 species [1,5-7]. It contains both asexual and sexual species and is characterized by the production of creamish, yellow, white, pinkish, or reddish ascomata and yellow ascospores [2,8-10]. Species in this section displays great diversity in their morphological characteristics. For example, most species have biverticillate conidiophores, while some have both biverticillate and monoverticillate conidiophores.

Species of section Talaromyces are commonly isolated from soil, indoor environments, food products, and leaf litter; they are also present in association with the ascomata of Pseudocosmospora sp. on rotten twigs of ginkgo (Ginkgo biloba L.) [2,6,11-15]. Some species have clinical, ecological, and industrial importance [14,16]. The dimorphic species, T. marneffei is a pathogen in patients with HIV infection. It is mostly detected in East Asia and causes 50,000 deaths each year [17-20]. Talaromyces pinophilus produces talaromycolides 1-3, 5 and 11 that act to inhibit the growth of the human pathogen methicillin-resistant Staphylococcus aureus [21]. It is also an important cellulose-degrading [21,22]. Talaraculones produced by T. aculeatus had inhibitory effect on α -glucosidase and thus can be used to prevent the progression of type II diabetes [23].

Only six new Talaromyces species have been reported based on materials collected from Korea: T. angelicae S.H. Yu, T.J. An & H.K. Sang [as "angelicus"], T. cnidii S.H. Yu, T.J. An & H.K. Sang, T. gwangjuensis Hyang B. Lee & T.T.T. Nguyen, T. halophytorum Y.H. You & S.B. Hong, T. koreanus Hyang B. Lee [as "koreana"], and T. teleomorphus Hyang B. Lee, Frisvad, P.M. Kirk, H.J. Lim & T.T.T. Nguyen [as "teleomorpha"] [7,24-26]. Only 11 Talaromyces species belonging to section Talaromyces have been well described in Korea [24,27-33]. In a survey of the genus Talaromyces in different environments in Korea, we discovered numerous species. Five of these species, namely T. brevis, T. fusiformis, T. muroii, T. ruber, and T. soli, have been reported for the first time in Korea. Herein, we have also proposed a new species, Talaromyces echinulatus sp. nov.

2. Materials and methods

2.1. Sampling and isolation

Soil samples were collected from Cheongyang and Anmyeondo in Chungnam Province, Gangbuk-gu in

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Seoul, and Dong-gu in Gwangju. The freshwater sample was taken from Hwangnyong river located at Gwangsan-gu, Gwangju, Korea. Fungal isolation was carried out from soil and freshwater samples as previously described [26,27]. For indoor contaminant samples, plates were contaminated with fungi from indoor air. Individual fungal colonies were transferred to potato dextrose agar (PDA) plates and incubated at 25 °C for 3-7 days. The cultures were maintained in 20% glycerol at -80° C at the Environmental Microbiology Laboratory Fungarium (Chonnam National University, Gwangju, Korea). The strains were also deposited at the National Institute of Biological Resources (NIBR, Incheon, Korea) under the numbers NIBRFG0000509602, NIBRFG0000509608, NIBRFGC000508622, NIBRFGC000510103 and the Nakdonggang National Institute of Biological Resources (NNIBR, Sangju, Korea) under the number NNIBRFG25658.

2.2. Fungal morphology studies

Morphological characteristics was investigated according to the method described by Yilmaz et al. [2]. Five media were used including, Czapek yeast autolysate agar (CYA; [34]), malt extract agar (MEA; [35]), yeast extract sucrose agar (YES; [36]), oatmeal agar (OA; Difco), and creatine sucrose agar (CREA; [36]). Medium preparation, inoculation, and microscopic examinations, were following our previous studies [26].

2.3. DNA extraction, PCR amplification, and sequencing

Total genomic DNA was extracted from fungal mycelia using the SolgTM Genomic DNA Preparation Kit (Solgent Co. Ltd., Daejeon, Korea) instructions. following the manufacturer's Polymerase chain reaction (PCR) was used to amplify four markers: The internal transcribed spacer (ITS) region of the rRNA, β -tubulin (BenA), calmodulin (CaM), and the RNA polymerase II second largest subunit (RPB2) genes. ITS was amplified using the primers ITS1/ITS4 [37], and V9G/LS226 [38,39]. For BenA, primers Bt2a/Bt2b [40], T10/Bt2b [40,41], and Tub2Fd/Tub4Rd [42] were used. CaM was amplified using primers Cmd5/Cmd6 [43], and CF1M/CF4 [44], while RPB2 was amplified using the primers RPB2-5F/RPB2-7cR [45]. PCR amplification and sequencing of ITS, BenA, CaM, and RPB2 gene was conducted using methods previously described [2,27].

2.4. Molecular analysis

The reference sequences of Talaromyces section Talaromyces used in this study were obtained from GenBank (Table 1). The original sequences were checked for their quality and assembled using BioEdit [46] and SeqMan v. 7.1.0. Sequences were aligned using MAFFT online program (http://mafft. cbrc.jp/alignment/server) [47] and then manually edited in MEGA7 [48]. A maximum likelihood analysis was carried out using RAxML-HPC2 on XSEDE 8.2.12 in the CIPRES Science Gateway [49] under the default settings with a GTR + G + I evolutionary model and bootstrap support obtained from 1000 replicates. Bayesian inference (BI) analyses were performed using MrBayes v. 3.2.6 [50]. Four independent MCMC chains were run for five million generations and trees were sampled every 100th generation. The first 25% of trees sampled were discarded as burn-in and the remaining trees were used to calculate Bayesian posterior probability (PP). The resulting tree was visualized using FigTree ver. 1.3.1 [51]. Support values were provided at the branches (ML bootstrap support (BS) and BI posterior probability (PP)). Talaromyces dendriticus CBS 660.80 was used as an outgroup.

3. Results

3.1. Phylogenetic analysis

Phylogenetic relationships within Talaromyces section Talaromyces were studied using the combined sequences of four loci: ITS, BenA, CaM, and RPB2. The alignment of the ITS-BenA-CaM-RPB2 concatenated sequences comprised 61 isolates, including the outgroup taxon, yielding 2486 characters (ITS: 1-592, BenA: 593-1087, CaM: 1088-1641, RPB2: 1642-2486). Strains CNUFC CY2268, CNUFC AS2-6, CNUFC HRGP2, CNUFC U7-11C, and CNUFC S5-T11 clustered with the type species of T. brevis, T. fusiformis, T. muroii, T. ruber, and T. soli, respectively, with strong bootstrap support (Figure 1). Strain CNUFC HB1206 clustered close to but separated from T. mangshanicus HMAS 248733 with 96% MLBS, 1 PP support (Figure 1).

3.2. Taxonomy

Talaromyces echinulatus Hyang B. Lee & T.T.T. Nguyen sp. nov. (Figure 2).

Index Fungorum number: IF900636.

Etymology: Referring to the species producing conidia with echinulate walls.

Type: Republic of Korea, Seoul, Gangbuk-gu, Beondong (37°37'52.3"N 127°02'28.1"E), from a soil

Table 1. Accession numbers for the fungal strains used for the phylogenetic analysis.

		GenBank accession nos.			
Species	Strain no	ITS	BenA	CaM	RPB2
Talaromyces aculeatus	CBS 289.48 ^T	KF741995	KF741929	KF741975	MH793099
Talaromyces adpressus	CBS 140620 ^T	KU866657	KU866844	KU866741	KU867001
Talaromyces alveolaris	CBS 142379 ^T	LT558969	LT559086	LT795596	LT795597
Talaromyces amestolkiae	CBS 132696 ^T	JX315660	JX315623	KF741937	JX315698
Talaromyces angelicae	KACC 46611	KF183638	KF183640	KJ885259	KX961275
Talaromyces annesophieae	CBS 142939 ^T	MF574592	MF590098	MF590104	MN969199
Talaromyces apiculatus	CBS 312.59 ^T	JN899375	KF741916	KF741950	KM023287
Talaromyces aurantiacus	CBS 314.59	JN899380	KF741917	KF741951	KX961285
Talaromyces australis	CBS 137102	KF741991	KF741922	KF741971	KX961284
Talaromyces beijingensis	CBS 140617	KU866649	KU866837	KU866733	KU866993
Talaromyces brevis	CBS 141833'	MN864269	MN863338	MN863315	MN863328
Talaromyces brevis	CNUFC CY2268	OR462361	OR507570	OR591615	OR608368
Talaromyces cnidii	KACC 46617	KF183639	KF183641	KJ885266	KM023299
Talaromyces derxii	CBS 412.89	JN899327	JX494306	KF/41959	KM023282
Talaromyces dendriticus	CBS 660.80	JN899339	JX091391	KF741965	KM023286
Talaromyces dimorphus	AS3.15692'	KY007095	KY007111	KY007103	KY112593
Talaromyces duclauxii	CBS 322.48	JN899342	JX091384	KF/41955	JN121491
Talaromyces echinulatus	CNUFC HB1206	OR462362	OK507571	OR608367	OR591610
Talaromyces euchlorocarpius	PF 1203	AB1/661/	KJ865733	KJ885271	KM023303
Talaromyces flavovirens	CBS 102801	JN899392	JXU91376	KF741933	KX961283
Talaromyces flavus	CBS 310.38	JN899360	JX494302	KF/41949	JF41/426
Talaromyces fusiformis	CDS TISTS4	KAUTISTU	KAUT 1409	KAUTISUT	NIN909100
Talaromyces fusiformis		00462363	OP507573	02602360	OP501611
Talaromyces aalanaaensis	$CRS 751 74^{T}$	INI800358	12/001388	KE741966	KX961280
Talaromyces indiaoticus	$CBS 100534^{T}$	IN899331	1X494308	KF741931	KX961278
Talaromyces intermedius	$CBS 152 65^{T}$	IN899332	IX091387	K 1885290	KX961282
Talaromyces kendrickii	$CBS 136666^{T}$	KF741987	KF741921	KF741967	MN969158
Talaromyces lentulus	AS3,15689 ^T	KY007088	KY007104	KY007096	KY112586
Talaromyces liani	CBS 225.66 ^T	JN899395	JX091380	KJ885257	KX961277
Talaromyces mae	AS3.15690 ^T	KY007090	KY007106	KY007098	KY112588
Talaromyces mangshanicus	HMAS 248733 ^T	KX447531	KX447530	KX447528	KX447527
Talaromyces marneffei	CBS 388.87 ^T	JN899344	JX091389	KF741958	KM023283
Talaromyces muroii	CBS 756.96 ^T	MN431394	KJ865727	KJ885274	KX961276
Talaromyces muroii	CNUFC HRGP2	OR462364	OR507572	OR591616	OR591612
Talaromyces mycothecae	URM 7622	MF278326	LT855561	LT855564	LT855567
Talaromyces neofusisporus	AS3.15415'	KP765385	KP765381	KP765383	MN969165
Talaromyces oumae-annae	CBS 138208 ⁺	KJ775720	KJ775213	KJ775425	KX961281
Talaromyces panamensis	CBS 128.89	JN899362	HQ156948	KF741936	KM023284
Talaromyces pinophilus	CBS 631.66'	JN899382	JX091381	KF741964	KM023291
Talaromyces pratensis	NRRL 62170	MH793075	MH792948	MH793012	MH793139
Talaromyces purpureogenus	CBS 286.36	JN899372	JX315639	KF741947	JX315709
Talaromyces qii	AS3.15414	KP/65384	KP/65380	KP/65382	MN969164
Talaromyces ruber	CBS 132/04	JX315662	JX315629	KF/41938	JX315/00
Talaromyces ruber		UK462365	UK507574	UK591617	UK591613
Talaromyces ravulitonsis	CDS 542.59 CPS 129204 ^T	JIN099504	JA494309 K1775306	KF741950 K1775455	MN060146
Talaromyces sayuntensis	CDS 138204	NJ773713	IV001270	KJ773422 KE741060	KM032370
Talaromyces soli	NPPL 62165 ^T	MH203024	MH702047	MH703011	MH703138
Talaromyces soli	CNILEC S5-T11	OR462366	OR507575	OR608370	OR591614
Talaromyces stellenhoschensis	CRS 135665 ^T	12091471	1X091605	IX140683	MN969157
Talaromyces stenenoosenensis Talaromyces stinitatus	$CBS 375.48^{T}$	IN899348	KM111288	KF741957	KM023280
Talaromyces stollii	$(BS 408.93^{T})$	IX315674	IX315633	IX315646	IX315712
Talaromyces thailandensis	CBS 133147 ^T	JX898041	JX494294	KF741940	KM023307
Talaromyces verruculosus	DTO 264-18 ^T	KF741994	KF741928	KF741944	KM023306
Talaromyces viridis	CBS 114.72 ^T	AF285782	JX494310	KF741935	JN121430
Talaromyces wushanicus	CS17-05 ^T	MZ356356	MZ361347	MZ361354	MZ361361
Talaromyces xishaensis	CGMCC 3.17995 [™]	KU644580	KU644581	KU644582	

Bold letters indicate strains and accession numbers determined in this study.

CBS: Culture collection of the Westerdijk Fungal Biodiversity Institute, The Netherlands; CGMCC: China General Microbiological Culture Collection Center; CNUFC: Chonnam National University Fungal Collection, Gwangju, Korea; DTO: Internal Culture Collection of the CBS-Fungal Biodiversity Center; FMR: Facultat de Medicina i Ciencies de la Salut, Reus, Spain; HMAS: Institute of Microbiology, Beijing, China; KACC: Korean Agricultural Culture Collection, Republic of Korea; NRRL: Agricultural Research Service Culture Collection, Peoria, IL, USA;^T: ex-type strain.

sample, March 2021, H.B. Lee (holotype CNUFC HT2197; ex-type culture CNUFC HB1206).

Colony diameters (mm), 7 days: CYA 25 °C 20–21; CYA 30 °C 11–12; CYA 37 °C no growth; MEA 25 °C 11–12; OA 25 °C 23–24; YES 25 °C 18–19; CREA 25 °C 10–12. Macromorphology (7 days at $25 \,^{\circ}$ C): On CYA, colonies with irregular margins, raised at the center and surrounded by sunken depression; mycelia white, texture with some floccose areas; sporulation poor; no exudate or soluble pigments; reverse centrally dark reddish brown, fading into light yellow.



Figure 1. Phylogram generated from maximum likelihood analysis based on combined ITS, *BenA, CaM*, and *RPB2* sequences data for species classified in *Talaromyces* section *Talaromyces*. The branches with values \geq 70% ML BS and \geq 0.95 PP indicated above or below branches. Bootstrap values lower than 0.95 and 70% are marked with "-". *Talaromyces dendriticus* CBS 660.80 was used as an outgroup. The newly generated sequence is in blue. ^T = ex type.

On MEA, colonies plane; margins submerged, irregular; sporulation poor; no exudate or soluble pigments; reverse light yellow. On YES, colonies nearly circular; margins low, plane, entire; mycelia white; texture velutinous; sporulation poor; no exudate or soluble pigments; reverse beige. On OA, colonies raised at the center, slightly sulcate; margins low, plane; mycelia white and yellow; texture velvety and floccose; sporulation strong; no exudate or soluble pigments; reverse light yellow. On CREA, acid production absent.

Micromorphology: Conidiophores mostly biverticillate, sometimes monoverticillate. Stipes smooth, $(24-)62-287(-341) \times 2.5-4.0 \,\mu\text{m}$. Metulae 3-5, 7- $10 \times 3-4 \,\mu\text{m}$. Phialides 3-5, flask-shaped, 8- $11.5 \times 2.5-3.5 \,\mu\text{m}$. Conidia subglobose to ovoid, or globose, thick-walled, echinulate-walled, 3.0- $4.5 \times 2.5-3.5 \,\mu\text{m}$.

Notes: In the multigene (ITS/*BenA/CaM/RPB2*) phylogeny, *T. echinulatus* clustered as a sister taxon to *T. mangshanicus* with high statistical support (96% MLBS, 1 PP) (Figure 1). *Talaromyces echinulatus* differs from *T. mangshanicus* by bigger colonies on CYA at 25 °C, smaller conidia $(3.0-4.5 \times 2.5-3.5 \,\mu\text{m} \text{ vs. } 4.5-5.5 \times 4-5 \,\mu\text{m})$ and fewer phialides per metula and metulae [52]. Based on its morphology and phylogenetic placement, herein we introduce *T. echinulatus* as a new species.

Talaromyces brevis B.D. Sun, A.J. Chen, Houbraken & Samson, MycoKeys 68: 92 (2020) [MB#833132] (Figure 3).

Colony diameters (mm), 7 days: CYA 25 °C 25–28; CYA 30 °C 25–30; CYA 37 °C 13–16; MEA 25 °C 45–47; OA 25 °C 37–39; YES 25 °C 41–42; CREA 25 °C 29–30.

Macromorphology (7 days at 25 °C): On CYA, sporulation moderate; texture velvety; mycelium white and yellow; reverse yellowish white. On MEA, sporulation poor; texture velvety; mycelium white; reverse pale yellow. On YES, colonies raised at the center; sporulation moderate; mycelium white and yellow; texture velvety; reverse yellowish white. On OA, colonies plane, not sulcate; mycelium white and yellow; sporulation moderate; reverse yellowish white; ascomata yellow to orange. On CREA, strong acid produced.

Micromorphology: Conidiophores monoverticillate and biverticillate. Stipes smooth, $8.5-65 \times 2-3 \mu m$.



Figure 2. Morphology of *Talaromyces echinulatus* sp. nov. (A,E) Colonies on Czapeck yeast autolysate agar (CYA). (B,F) Malt extract agar (MEA). (C,G) Yeast extract sucrose agar (YES). (D) Oatmeal agar (OA). (H) Creatine sucrose agar (CREA). (A–D, H: Obverse view, E–G: reverse view). (I–K) Conidiophores. (L) Conidia. Scale bars = $10 \,\mu$ m.



Figure 3. Morphology of *Talaromyces brevis* CNUFC CY2268. (A,E) Colonies on Czapeck yeast autolysate agar (CYA). (B,F) Malt extract agar (MEA). (C,G) Yeast extract sucrose agar (YES). (D) Oatmeal agar (OA). (H) Creatine sucrose agar (CREA). (A–D, H: Obverse view, E–G: reverse view). (I,J) Conidiophores. (K) Conidia. Scale bars: I, $J = 10 \,\mu$ m, $K = 5 \,\mu$ m.

Metulae 1–3, divergent, $8.5-13.5 \times 2.5-3 \mu m$. Phialides 3–5, flask-shaped, $8-13 \times 2-3 \mu m$. Conidia smooth, mostly subglobose, some fusiform, 2.5– $3.5 \times 2-3 \mu m$. Ascomata maturing within 2 weeks on OA and MEA, yellow, globose to subglobose.

Material examined: Republic of Korea, Chungnam Province, Cheongyang, Cheongyang-eup, Kunryangri (36°26'16.2" N 126°46'04.6" E), from a soil sample, May 15, 2022, H.B. Lee and J.S. Kim (culture CNUFC CY2268).

Notes: Our strain CNUFC CY2268 and the type strain of *T. brevis* CBS 141833 clustered together with 100% MLBS and 1 PP support (Figure 1). However, our strain has smaller conidia from the type species $[2.5-3.5 \times 2-3 \,\mu\text{m} \text{ vs. } 3-4(-5) \times 2.5-3.5(-4.5) \,\mu\text{m}]$ [3].

Talaromyces fusiformis A.J. Chen, Frisvad & Samson, Studies in Mycology 84: 139 (2016) [MB#817396] (Figure 4).

Colony diameters (mm), 7 days: CYA 25 °C 23–28; CYA 30 °C 36–42; CYA 37 °C 39–41; MEA 25 °C 36–40; OA 25 °C 37–42; YES 25 °C 23–28; CREA 25 °C 18–20.

Macromorphology (7 days at $25 \,^{\circ}$ C): On CYA, colonies slightly raised at the center; sporulation moderate; mycelium white and grayish green; texture floccose; reverse yellowish brown at the center, pale yellow to white at margins. On MEA, colonies plane; sporulation strong; mycelium white; reverse orange at the center, yellowish brown at the edge. On YES, colonies slightly sulcate; texture floccose; sporulation poor; mycelium white; reverse yellowish brown. On OA, sporulation moderate; mycelium white and greenish grey; texture funiculose; reverse yellowish green. On CREA, acid not produced.

Micromorphology: Conidiophores biverticillate. Stipes 76–142 × 2.5–3.0 μ m. Metulae 3–4, appressed, 9–14 × 2.5–3.0 μ m. Phialides acerose, 3–4 per metula, 8.5–13.5 × 2.0–3.0 μ m. Conidia smooth, ellipsoidal to fusiform, 2.5–4.0 × 2.0–2.5 μ m.

Material examined: Republic of Korea, Chungnam Province, Anmyeon-eup, Taean-gun (36°34'46.0"N,



Figure 4. Morphology of *Talaromyces fusiformis* CNUFC AS2-6. (A,E) Colonies on Czapeck yeast autolysate agar (CYA). (B,F) Malt extract agar (MEA). (C,G) Yeast extract sucrose agar (YES). (D) Oatmeal agar (OA). (H) Creatine sucrose agar (CREA). (A–D, H: Obverse view, E–G: reverse view). (I–K) Conidiophores. (L) Conidia. Scale bars = 10 μm.

126°19'42.7"E), from a soil sample, January 29, 2019, H.B. Lee (culture CNUFC AS2-6).

Notes: Phylogenetic analysis placed our strain CNUFC AS2-6 close to the type strain of *T. fusiformis* CBS 140637 with 100% MLBS and 1 PP support (Figure 1). Our strain can grow on CREA, while *T. fusiformis* CBS 140637 is unable to grow on CREA [13].

Talaromyces muroii Yaguchi, Someya & Udagawa, Mycoscience 35 (3): 252 (1994) [MB#362930] (Figure 5).

Colony diameters (mm), 7 days: CYA 25 °C 22– 25; CYA 30 °C 40–42; CYA 37 °C 38–40; MEA 25 °C 35–38; OA 25 °C 33–36; YES 25 °C 34–36; CREA 25 °C 24–26.

Macromorphology (7 days at 25 °C): On CYA, sporulation moderate; texture velvety to slightly floccose; mycelia white to pastel yellow; reverse brownish yellow. On MEA, sporulation poor; texture velvety mycelia white; reverse uncolored. On

YES, colonies raised in the center; texture floccose; mycelia white to yellow; sporulation moderate; reverse brownish orange. On OA, colonies texture velutinous and granular; sporulation moderate; mycelium white and yellow; reverse light greenish yellow. On CREA, strong acid produced.

Micromorphology: Conidiophores biverticillate and monoverticillate. Stipes $(35-)57-145(-169) \times 2.5-4 \mu m$, Metulae 3–6, 9–13.5 × 2–3 μm . Phialides acerose, 3–5 per metula, 7–13 × 2–2.5 μm . Conidia smooth, globose to subglobose, 2–3.5 × 2–3 μm .

Material examined: Republic of Korea, Gwangju, Gwangsan-gu, Dosan-dong, Hwangnyong River (35.12031°N, 126.7934°E), from a freshwater sample, January 02, 2019 (culture CNUFC HRGP2).

Notes: Our strain CNUFC HRGP2 clustered with the type strain of *T. muroii* CBS 756.96 with a high statistical support (100% MLBS and 1 PP support) (Figure 1). However, our strain has a faster growth than *T. muroii* on CYA (22-25 mm vs. 16–18 mm), and OA (33–36 mm vs. 16–18 mm) at 25 $^{\circ}$ C [53].



Figure 5. Morphology *Talaromyces muroii* CNUFC HRGP2. (A,E) Colonies on Czapeck yeast autolysate agar (CYA). (B,F) Malt extract agar (MEA). (C,G) Yeast extract sucrose agar (YES). (D) Oatmeal agar (OA). (H) Creatine sucrose agar (CREA). (A–D, H: Obverse view, E–G: reverse view). (I–K) Conidiophores. (L) Conidia. Scale bars $= 20 \,\mu$ m.

Talaromyces ruber (Stoll) Yilmaz, Houbraken, Frisvad, & Samson, Persoonia 29: 48 (2012) [MB#801360] (Figure 6).

≡Penicillium rubrum Stoll, Beiträge zur Morphologischen und Biologischen Charakteristik von *Penicillium*-Arten: 35 (1904).

 \equiv *P. crateriforme* J.C. Gilman & E.V. Abbott, Iowa State College Journal of Science 1 (3): 293 (1927).

Colony diameters (mm), 7 days: CYA 25 °C 23– 29; CYA 30 °C 18–21; CYA 37 °C 11–12; MEA 25 °C 47–50; OA 25 °C 28–34; YES 25 °C 19–22; CREA 25 °C 16–17.

Macromorphology (7 days at $25 \,^{\circ}$ C): On CYA, colonies sulcate, sunken in the center; texture floccose, mycelia white to yellow green or light orange yellow; sporulation moderate; reverse center grayish purplish red to moderate yellowish pink. On MEA, sporulation strong; texture velvety; mycelia white to greenish yellow; soluble pigments moderate orange; reverse

reddish orange at the center and yellowish pink at margins. On YES, colonies raised at the center, sunken in the center, sulcate; sporulation moderate; texture velvety and floccose; mycelia white; reverse moderate yellow green. On OA, colonies plane; mycelium white and greenish; colony texture velvety; sporulation strong; exudates and soluble pigments absent; reverse grayish red. On CREA, acid production absent.

Micromorphology: Conidiophores biverticillate. Stipes $51-174 \times 2.5-3.5 \,\mu\text{m}$. Metulae 3-5, $9-12.5 \times 2.5-3.5 \,\mu\text{m}$. Phialides acerose, $7-9.5 \times 2-3 \,\mu\text{m}$. Conidia smooth, ellipsoidal, $2.5-4.0 \times 2-3 \,\mu\text{m}$.

Material examined: Republic of Korea, Gwangju, Buk-gu, Chonnam National University (35.17611°N, 126.90582°E), from an indoor air sample, April 26, 2020 (culture CNUFC U7-11C).

Notes: Phylogenetic analysis of a combined ITS, *BenA*, *CaM* and *RPB2* sequences data showed that our strain CNUFC U7-11C is related to *T. ruber* CBS 132704 (type strain) with 100% MLBS and 1



Figure 6. Morphology of *Talaromyces ruber* CNUFC U7-11C. (A,E) Colonies on Czapeck yeast autolysate agar (CYA). (B,F) Malt extract agar (MEA). (C,G) Yeast extract sucrose agar (YES). (D) Oatmeal agar (OA). (H) Creatine sucrose agar (CREA). (A–D, H: Obverse view, E–G: reverse view). (I–K) Conidiophores. (L) Conidia. Scale bars = $10 \mu m$.

PP support (Figure 1). Furthermore, our strain resembles *T. ruber* in shape and size of metulae, phialides and conidia [54].

Talaromyces soli Jurjević & S.W. Peterson, Fungal Biology 123 (10): 757 (2019) [MB#827832] (Figure 7). Colony diameters (mm), 7 days: CYA 25 °C 21–22; CYA 30 °C 35–36; CYA 37 °C 11–12; MEA 25 °C 23–24; OA 25 °C 28–29; YES 25 °C 22–23; CREA 25 °C 18–19.

Macromorphology (7 days at 25 °C): On CYA, colonies slightly sulcate; sporulation poor; mycelia white; texture velvety; yellowish pink exudate; reverse moderate orange yellow. On MEA, sporulation moderate; mycelia white and yellowish green; texture floccose; reverse pale greenish yellow. On YES, colonies raised at center; sporulation poor; texture floccose; mycelia white; reverse light yellow. On OA, sporulation moderate; mycelia white; reverse light yellow. On OA, sporulation moderate; mycelia white and pastel yellow green; texture funiculose and floccose; reverse brownish yellow. On CREA, strong acid production. Micromorphology: Conidiophores biverticillate. Stipes smooth, $(48-)73-157 \times 2.5-3.5 \,\mu\text{m}$. Metulae

3–4, $8.5-10 \times 2.5-3 \mu m$. Phialides 3–4, flask-shaped, $9-12 \times 2-3 \mu m$. Conidia smooth, subglobose to ellipsoidal, $2.5-3 \times 2-2.5 \mu m$.

Material examined: Republic of Korea, Gwangju, Dong-gu (35°07'23.5"N 126°58'20.1"E), from a soil sample, December 2020 (culture CNUFC S5-T11).

Notes: In the phylogenetic analysis, our strain clustered with the type strain of *T. soli* NRRL 62165 with 100% MLBS and 1 PP support (Figure 1). Compared with *T. soli*, our strain has smaller conidia size $[2.5-3 \times 2-2.5 \,\mu\text{m} \text{ vs. } 5-3.5(-5.5) \times 2.5-3.5(-4.5) \,\mu\text{m}]$ [55].

4. Discussion

Talaromyces is an important genus in biotechnology and in medical and food mycology [2]. It has a huge impact, both positive and negative, on daily life. The impact of *Talaromyces* species has generated great interest in the taxonomy of this group of fungi. Consequently, the number of new species



Figure 7. Morphology of *Talaromyces soli* CNUFC S5-T11. (A,E) Colonies on Czapeck yeast autolysate agar (CYA). (B,F) Malt extract agar (MEA). (C,G) Yeast extract sucrose agar (YES). (D) Oatmeal agar (OA). (H) Creatine sucrose agar (CREA). (A–D, H: Obverse view, E–G: reverse view). (I–K) Conidiophores. (L) Conidia. Scale bars = $10 \mu m$.

descriptions of *Talaromyces* has dramatically increased in recent years [1–3,7].

In this study, one new species and five new records of *Talaromyces* species belonging to section *Talaromyces* were described based on current classification system [3].

The new Talaromyces species identified in this study is phylogenetically related to T. mangshanicus in combined phylogenetic analyses with ITS, BenA, CaM and RPB2 sequences. However, T. echinulatus differs from T. mangshanicus by having bigger colonies on CYA (6.5-7 mm) and smaller colonies on MEA (15-18 mm) incubated at 25 °C [52]. Talaromyces mangshanicus produces soluble pigments purplish red on YES, while T. echinulatus does not produce. The number of phialides per metula and metulae of T. echinulatus are less than T. mangshanicus [52]. Furthermore, T. echinulatus produces globose, subglobose to ovoid conidia and biverticillate, or monoverticillate, in contrast to the subglobose to ellipsoidal conidia and biverticillate sporangiophores of *T. mangshanicus*. Therefore, it is a newly described species.

Species belonging to section Talaromyces share various extrolites. For example, T. purpurogenus produces a well-known hepatocarcinogenic toxin, rubratoxin [56,57], while T. amazonensis produces the potential anticancer agents duclauxin, berkelic acid, and vermicillin [11]. Talaromyces fusiformis produces purpactin A, secalonic acid D, and chrodrimanin A (= thailandolide) [13], while T. muroii effective as an antifungal agent against is Alternaria brassicicola, which causes leaf spot in kale and anthracnose in coffee [58,59]. Moreover, T. ruber produces several extrolites, including austin, berkelic acid, mitorubrin, mitorubrinic acid, Monascus red azaphilone pigments, pestalacin A, purpactin A, and vermicellin [2,54]. Talaromyces brevis and T. soli produce acid on CREA [3,55]. Therefore, the Talaromyces species isolated in this study are potentially useful sources of secondary metabolites.

Species in section Talaromyces have been isolated worldwide from various habitats. Talaromyces brevis is commonly isolated from soil [3], as in the case of our isolate, and seems to be predominant in soil habitats. Talaromyces fusiformis has been isolated from indoor air samples, respiratory specimens of patients with pulmonary disorders [13,60], and from soil in the current study. Talaromyces muroii has been isolated from soil, sheep dung, chicken crops, jute potato bags treated with copper oxide ammonia, clematis [2,8,53,61], and from freshwater in the current study. Talaromyces ruber has been isolated from a tropical dry forest, soil, an aircraft fuel tank, currency paper, clinical specimens, dung [2,14,54,62,63], and from indoor air in the current study. Talaromyces soli has been isolated from an ant nest in Texas [63], aquatic macrophytes in Arizona [64], soil [55], an endophyte of beans in Columbia [65], and from soil in the current study.

Our findings include the first reports of *T. muroii* in freshwater niche, also reveal a wider distribution of *Talaromyces* species within section *Talaromyces*. Thus this study also provides a better understanding species diversity in the genus *Talaromyces* belonging to section *Talaromyces* in Korea.

Disclosure statement

No potential conflict of interest was reported by the author(s).

Funding

This work was supported by National Research Foundation of Korea (NRF) funded by the Ministry of Science and ICT [2022M3H9A1082984], and also by the Survey and Discovery of Indigenous Fungal Species of Korea Project [NIBR202203204] funded by the NIBR, and by the Discovery of Fungi from Freshwater and their Collection for Fungaria Project [NNIBR202201206] funded by the NNIBR of the Ministry of Environment, Republic of Korea.

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