

RESEARCH ARTICLE

Taxonomic Characteristics of Twelve New Yeast Species Isolated in 2023 Not Listed in the National Species List of Korea

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ABSTRACT

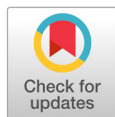
With a history spanning 9,000 years, yeast has played a central role in the traditional production of fermented foods, particularly alcohol and bakery. Recent research has highlighted the versatility of yeast in various industries, including the production of bioactive compounds, bioethanol, pigments, and enzymes, and as a host for molecular biology studies. In Korea, yeast isolation has traditionally focused on traditional fermented foods such as soybean paste. However, there is a growing trend in the isolation and characterization of yeasts from natural environments such as flowers and fruits for industrial applications. In this study, we isolated and identified 12 yeasts from various natural environments in Korea, including botanical gardens and parks that are not listed on the National Species List of Korea (NSLK). These newly discovered species included *Sakaguchia oryzae*, *Cystobasidium raffinophilum*, *Meira argovae*, *Kazachstania humilis*, *Meyerozyma smithsonii*, *Anthracoecystis trispicatae*, *Naganishia brisbanensis*, *Tremella yokohamensis*, *Kwoniella shandongensis*, *Kwoniella newhampshirensis*, *Aureobasidium proteae*, and *Rhodotorula dairenensis*.

KEYWORDS: National Species List of Korea (NSLK), Unrecorded yeast

INTRODUCTION

Yeasts are unicellular fungi that reproduce by budding or fission and are not a taxonomic classification. They possess specific morphological and physiological traits that distinguish them from other fungi [1,2]. Yeast has a rich history in the production of traditional fermented foods (particularly alcohol) that was utilized approximately 9,000 years ago. Recent studies show that yeasts are a versatile group of organisms with the potential for use in various industries. They can produce a range of bioactive substances, bioethanol, pigments, and enzymes, and serve as hosts in molecular biology [3,4].

In the past, yeasts found in Korea was primarily isolated and identified from fermented foods such as traditional soybean paste or *meju*. However, there is a recent increase in the isolation and characterization of yeasts from natural environments (including flowers and fruits) for application in numerous industries



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[5-8]. These studies indicate that wild yeasts in natural environments exhibit species diversity based on their source of isolation, which varies according to the climatic characteristics of the region. Furthermore, the same source of isolation can yield different yeast communities, depending on the collection location [3].

In this study, we isolated and identified wild yeasts from flowers, trees, and fruits collected in the natural environment in Korea, including various botanical gardens and parks in Yongin, Suwon, Daegu, Hongseong, Jeju, and Osan. Finally, we have selected 12 unrecorded species of yeasts that are not included in the National Species List of Korea (NSLK), namely *Sakaguchia oryzae*, *Cystobasidium raffinophilum*, *Meira argovae*, *Kazachstania humilis*, *Meyerozyma smithsonii*, *Anthracoystis trispicatae*, *Naganishia brisbanensis*, *Tremella yokohamensis*, *Kwoniella shandongensis*, *Kwoniella newhampshirensis*, *Aureobasidium proteae*, and *Rhodotorula dairenensis*. The microbiological characteristics of these yeast strains are described in detail.

MATERIALS AND METHODS

Species and strain selection

Four hundred and fifty yeast strains were isolated from various regions and materials such as Zelkova, Quercus oak, Camellia, and tangerine trees in Korea. Among them, twelve selected yeast strains not recorded in the NSLK were selected for listing. Samples were collected in sterilized tubes from different locations, crushed with a mortar and pestle and soaked overnight in 10 mL saline solution (pH 3.3) to obtain a suspension. The suspension was spread on yeast extract peptone dextrose (YPD) agar containing ampicillin (0.1 mL/100 mL), dichloran rose bengal chloramphenicol (DRBC) agar, and dichloran glycerol (DG18) agar, as previously described [9]. Yeast colonies were grown for 48 h at 25°C, then picked and isolated as a single colony for identification [10].

DNA isolation, amplification, and phylogenetic analysis

The isolated yeast strains were identified by analyzing the sequence of the internal transcribed spacer (ITS) region with biological relationships registered in the National Center for Biotechnology Information (NCBI) database. Polymerase chain reaction (PCR) was performed to amplify the ITS region using the ITS-1 (TCCGTAGGTGAACCTGCGG) and ITS-4 (TCCTCCGCTTATTGATATGC) primers. The reaction was performed in the following steps: pre-denaturation at 95°C for 5 min, 30 cycles of denaturation at 95°C for 30 s, annealing at 55°C for 30 s, and extension at 72°C for 30 s. Finally, a post-extension step was performed at 72°C for 10 min. The ITS region sequences of the isolates were deposited in GenBank (<https://www.ncbi.nlm.nih.gov/genbank/>).

Phylogenetic analysis was performed for taxonomic classification using the ITS region nucleotide sequences of the type strains of species within the genus or family from the NCBI database. A phylogenetic tree was constructed using MEGA X software through the neighbor-joining method, which relies on the Tamura-Nei model. Bootstrap analysis was performed 1,000 times to ensure accuracy [11].

Morphology

Cell morphology was observed by culturing cells on yeast malt (YM) agar plates for 3 d at 25°C and observing them using a light microscope (DN-10A, Samwon, Goyang, Korea). Colony formation was observed by inoculating the single colony and growing on YM agar plate for 7 d at 25°C. The formation of hyphae and pseudohyphae was observed using Dalmau plates on cornmeal agar for 14 d at 25°C [12].

RESULTS AND DISCUSSION

The origin of 12 undocumented yeast strains identified in this study are shown in Table 1. These 12 strains consisted of three *Ascomycota* and nine *Basidiomycota* yeasts. These strains were identified by comparison with the ITS region sequence of the type strain. Consequently, the yeast strain *Naganishia brisbanensis* KHU20220630-01 (Genbank: OR791288) obtained from *Liriope muscari* produced a single clade with the *N. brisbanensis* holotype strain BRIP 28244 (Fig. 1A). Similarly, the yeast strain *Sakaguchia oryzae* KHU20220630-02 (Genbank: OR791290) obtained from *Pteridium aquilinum* var. *latiusculum* formed a single clade with the type strain CBS 9745 of *S. oryzae* (Fig. 2A). The yeast strain *Cystobasidium raffinophilum* KHU20220630-03 (GenBank: OR791282) from *Camellia japonica* formed a single clade with the holotype strain CGMCC 2.3822 of *C. raffinophilum* (Fig. 3A), and the yeast strain *Meira argovae* KHU20220630-04 (GenBank: OR791287) from *Olea europaea* formed a single clade with the type strain CBS 110053 of *M. argovae* (Fig. 4A). *Kazachstania humilis* KHU20220630-05 (GenBank: OR791284) obtained from *C. reticulata* × *C. sinensis* formed a single clade with *K. humilis* type strain CBS 5658 (Fig. 5A). *Meyerozyma smithsonii* KHU20220630-06 (GenBank: OR791289) from *Citrus reticulata* confirmed the presence of two sequences within the same species in the database and produced a single clade using *M. smithsonii* strain H1 instead of the type strain (Fig. 6A). *Anthracocystis trispicatae* KHU20220630-07 (GenBank: OR791280), obtained from *Bambusoideae* formed a single clade with isotype strain BRIP 47730 of *A. trispicatae* (Fig. 7A). *Tremella yokohamensis* KHU20230328-01 (GenBank: OR791291) from *Quercus serrata* formed a single clade with CBS 11776, the type strain of *T. yokohamensis* (Fig. 8A), *Kwoniella shandongensis* KHU20230328-02 (GenBank: OR791286) obtained from *Malus sieboldii* formed a single clade with CBS 12478, the type strain of *K. shandongensis* (Fig. 9A). *Kwoniella newhampshirensis* KHU20230809-01 (Genbank: OR791285) from *Ligustrum japonicum* produced a single clade with CBS 13917 the holotype strain of *K. newhampshirensis* (Fig. 10A). *Aureobasidium proteae* KHU20230809-02 (Genbank: OR791281) from *Ipomoea nil* produced a single clade with CBS 111973, an epitype strain of *A. proteae* (Fig. 11A), and *Fellomyces penicillatus* KHU20230809-03 (Genbank: OR791283) from *Zelkova serrata* produced a single clade with *F. penicillatus* CBS 5492 (Fig. 12A). The yeasts were morphologically characterized and 12 yeasts are listed in the NSLK database.

Table 1. List of 12 yeast species used in this study

Genus	Species	Strains	Origin	Locality ^a
Basidiomycota, Agaricomycotina, Tremellomycetes, Filobasidiales				
<i>Naganishia</i>	<i>Brisbanensis</i>	KHU20220630-01	<i>Liriope muscari</i>	Yongin, GG
Basidiomycota, Agaricomycotina, Tremellomycetes, Tremellales				
<i>Tremella</i>	<i>Yokohamensis</i>	KHU20230328-01	<i>Quercus serrata</i>	Osan, GG
<i>Kwoniella</i>	<i>Shandongensis</i>	KHU20230328-02	<i>Malus sieboldii</i>	Osan, GG
<i>Kwoniella</i>	<i>Newhamphshirensis</i>	KHU20230809-01	<i>Ligustrum japonicum</i>	Daegu, GN
<i>Fellomyces</i>	<i>Penicillatus</i>	KHU20230809-03	<i>Zelkova serrata</i>	Hongseong, CN
Basidiomycota, Pucciniomycotina, Cystobasidiomycetes, Cystobasidiomycetes incertae sedis				
<i>Sakaguchia</i>	<i>Oryzae</i>	KHU20220630-02	<i>Pteridium aquilinum</i> var. <i>latiusculum</i>	Yongin, GG
Basidiomycota, Pucciniomycotina, Cystobasidiomycetes, Cystobasidiales				
<i>Cystobasidium</i>	<i>Raffinophilum</i>	KHU20220630-03	<i>Camellia japonica</i>	Yongin, GG
Basidiomycota, Ustilaginomycotina, Exobasidiomycetes, Exobasidiales				
<i>Meira</i>	<i>Argovae</i>	KHU20220630-04	<i>Olea europaea</i>	Yongin, GG
Basidiomycota, Ustilaginomycotina, Ustilaginomycetes, Ustilaginales				
<i>Anthracoystis</i>	<i>Trispicatae</i>	KHU20220630-07	<i>Bambusoideae</i>	Seogwipo, JJ
Ascomycota, Saccharomyceta, Saccharomycotina, Saccharomycetes, Saccharomycetales				
<i>Kazachstania</i>	<i>Humilis</i>	KHU20220630-05	<i>C. reticulata</i> × <i>C. sinensis</i>	Seogwipo, JJ
<i>Meyerozyma</i>	<i>Smithsonii</i>	KHU20220630-06	<i>Citrus reticulata</i>	Seogwipo, JJ
Ascomycota, Saccharomyceta, Pezizomycotina, leotiomyceta, dothideomyceta, Dothideomycetes, Dothideomycetidae, Dothideales, Saccotheciaceae, Aureobasidium				
<i>Aureobasidium</i>	<i>Protea</i>	KHU20230809-02	<i>Ipomoea nil</i>	Hongseong, CN

^a GG: Gyeonggi-do; GN: Gyeongsangnam-do; CN: Chungcheongnam-do; JJ: Jeju-do.

Species description

Naganishia brisbanensis Tan, Y. P., Marney, T. S., Shivas, R. G., Index Fungorum 495:4, 2021

Naganishia is a fungal genus that belongs to the *Filobasidiales* order. The genus *Naganishia* was initially described by Goto et al. (1963) [13]. *Naganishia brisbanensis* was distinguished from *Naganishia albida* [14].

Colonies were regular in shape with an entire margin and ivory to white color after 3 d on YM agar at 25°C. Cells were ovoid to elongated with a size of $7.84 \times 7.14 \mu\text{m}$, and occurring singly or in pairs after 3 days on YM agar at 25°C. The budding was polar and on a narrow base. A teliospore formed after two weeks of culture on Dalmau plates at 25°C (Fig. 1B-E).

Remarks: *Naganishia brisbanensis* is unstudied and reported. *Naganishia albida* is genetically close to *Naganishia brisbanensis* and can produce lipids using agricultural and industrial wastes [15].

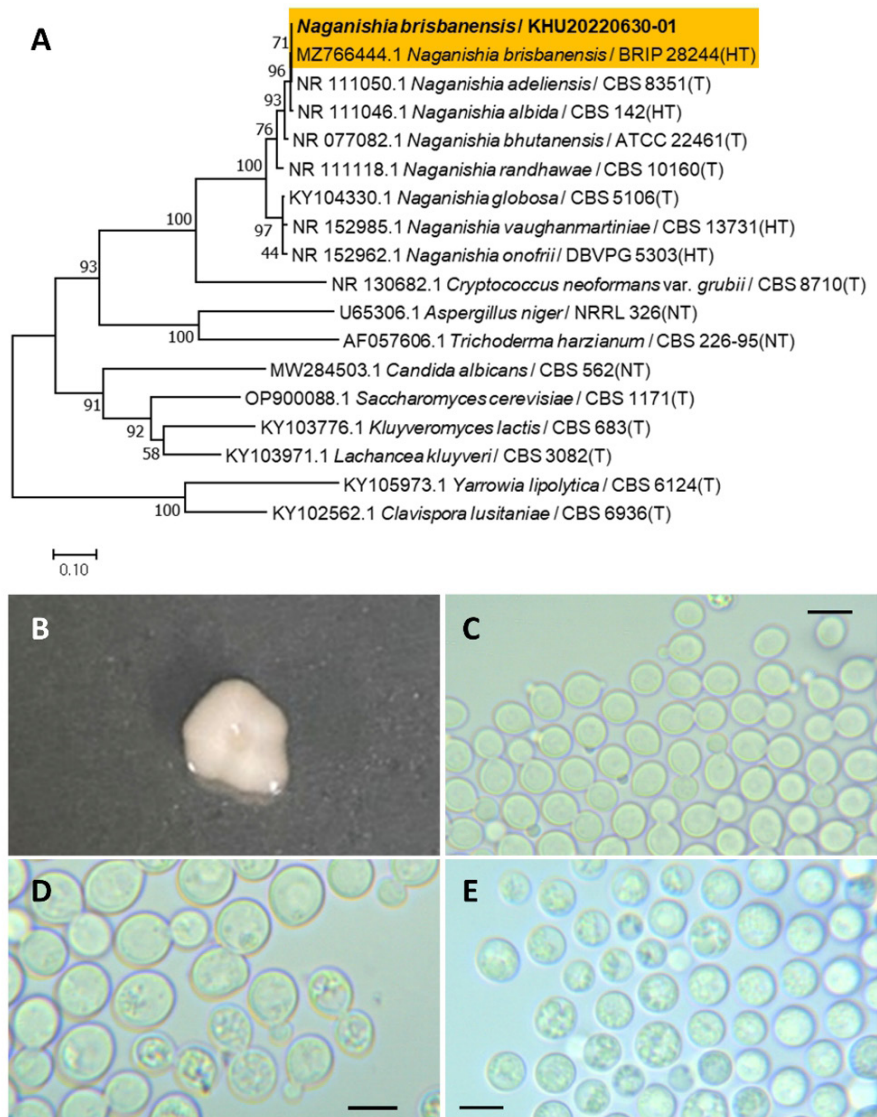


Fig. 1. Phylogenetic tree and morphological characteristics of *Naganishia brisbanensis*. A. Phylogenetic tree drawn from neighbor-joining analysis based on sequence of the internal transcribed spacer region, showing positions of *N. brisbanensis* strains isolated from Korea. Bold means representative strain. B-E. Morphology of *N. brisbanensis* KHU20220630-01. B. Colony on yeast aalt (YM) agar after 7 d at 25°C. C. Budding cells on YM agar after 3 d at 25°C. D. Budding cells occurring in short chains on YM agar after 7 d at 25°C. E. Teliospores are formed on Dalmau plates containing cornmeal agar after 2 weeks. Scale bars, 10 µm. (T, Type strain; HT, Holotype; NT, Neotype.)

Sakaguchia oryzae (F. Y. Bai & Y. M. Cai) Wang, Q. M., Bai, F. Y., Groenewald, M., Boekhout, T., Stud. Mycol., 81(1), 149-189, 2015

Sakaguchia is a fungal genus belonging to the order *Cystobasidiomycetes* incertae [16]. Colonies were irregular in shape with an entire margin and orange to pink in color after 3 days on YM agar at 25°C. Cells were ovoid to elongated with a size of 9.96×4.40 µm, and occur singly or in pairs after 3 d on YM agar at 25°C. The budding was polar and on Fa narrow base. The teliospore was formed after two weeks of culture on Dalmau plates at 25°C (Fig. 2B-E). This species has teliospores lateral or terminal to the hyphae, and

pseudohyphae or true hyphae present or absent [17].

Remarks: The *Sakaguchia* genus is unstudied, although the *Rhodotorula* genus related to *Sakaguchia* can produce carotenoids including β -carotene, and lipid can be used for biodiesel [18].

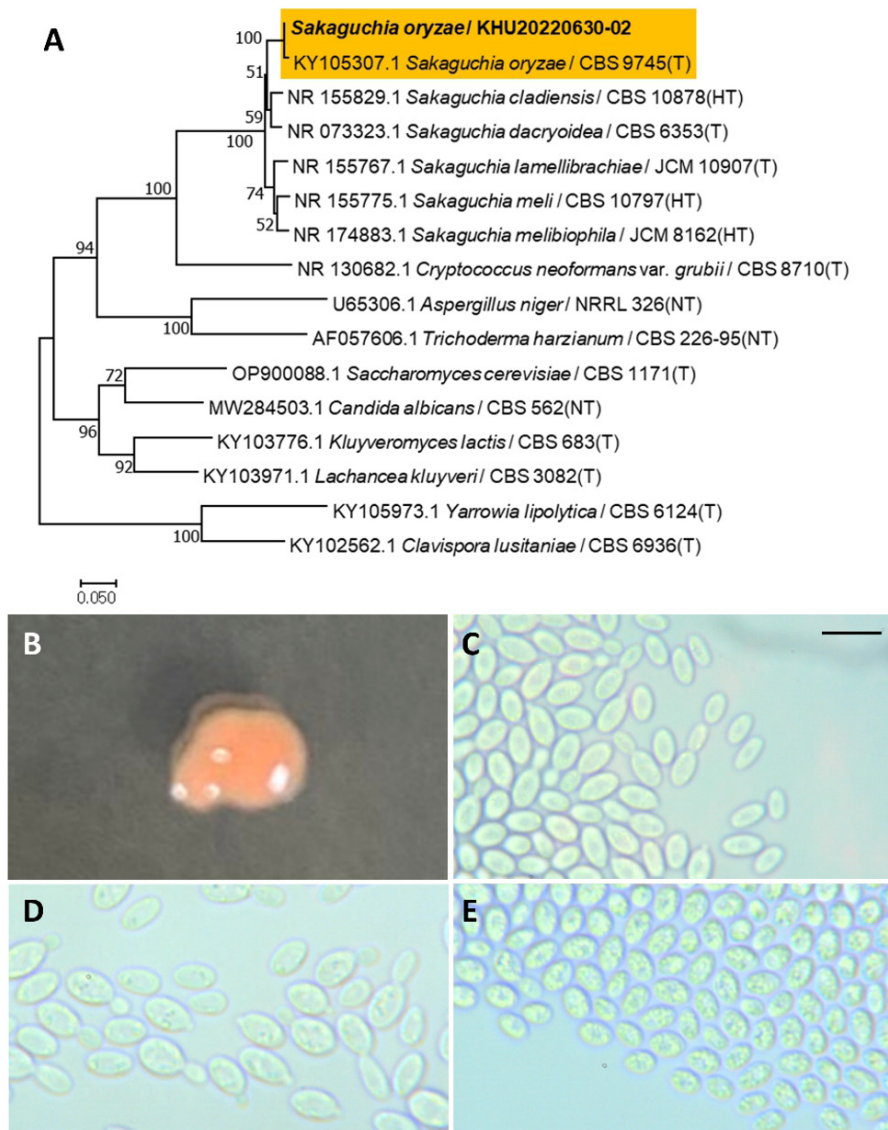


Fig. 2. Phylogenetic tree and morphological characteristics of *Sakaguchia oryzae*. A. Phylogenetic tree drawn from neighbor-joining analysis based on sequence of the internal transcribed spacer region, showing positions of *S. oryzae* strains isolated from Korea. Bold means representative strain. B-E. Morphology of *S. oryzae* KHU20220630-02. B. Colony on yeast malt (YM) agar after 7 d at 25°C. C. Budding cells on YM agar after 3 d at 25°C. D. Budding cells occurring in short chains on YM agar after 7 d at 25°C. E. Teliospores are formed on Dalmau plates containing commel agar after 2 weeks. Scale bars, 10 μ m. (T, Type strain; HT, Holotype; NT, Neotype.)

Cystobasidium raffinophilum Wang, Q. M. Bai, F. Y. Li, A. H. Stud. Mycol., 96(1), 17-140, 2020

Cystobasidium is a fungal genus belonging to the *Cystobasidiales* order. The genus *Cystobasidium* was originally established by Lagerheim in 1898 and described by Neuhoﬀ in 1924 [19].

Colonies were round with entire margins and pink to white in color after 3 days on YM agar at 25°C. Cells were ovoid to elongated with a size of 8.85 × 4.20 µm, and occurred singly or in pairs after 3 d on YM agar at 25°C. The budding was polar and on a narrow base. Pseudohyphae were formed after two weeks of culture on Dalmau plates at 25°C. Ascospores were also observed (Fig. 3B-E). This species has pseudohyphae, but sexual structures were not observed [20].

Remarks: *Cystobasidium raffinophilum* is unstudied, although the genus *Cystobasidium* produces carotenoids including β-carotene, γ-carotene, torulene, and torularhodin [21]. The *Cystobasidium* genus also produces lipids [22].

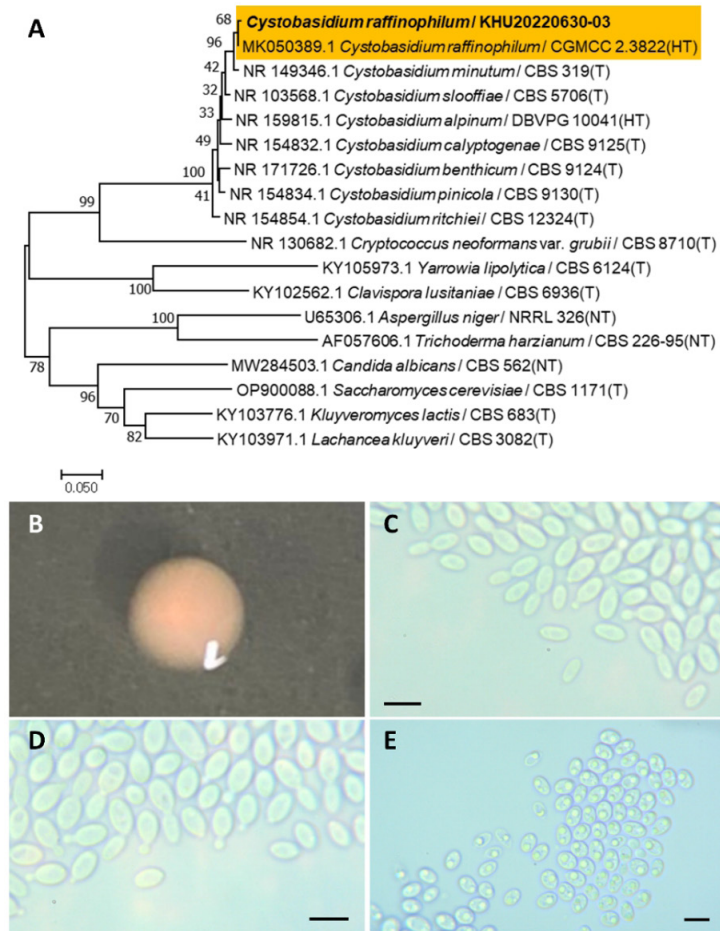


Fig. 3. Phylogenetic tree and morphological characteristics of *Cystobasidium raffinophilum*. A. Phylogenetic tree drawn from neighbor-joining analysis based on sequence of the internal transcribed spacer region, showing positions of *C. raffinophilum* strains isolated from Korea. Bold means representative strain. B-E. Morphology of *C. raffinophilum* KHU20220630-03. B. Colony on yeast malt (YM) agar after 7 d at 25°C. C. Budding cells on YM agar after 3 d at 25°C. D. Budding cells occurring in short chains on YM agar after 7 d at 25°C. E. Pseudohyphae and ascospores formed on Dalmau plates containing cornmeal agar after 2 weeks. Scale bars, 10 µm. (T, Type strain; HT, Holotype; NT, Neotype.)

Meira argovae Boekhout, T., Scorzetti, G., Gerson, U., Sztejnberg, A., Denchev, C. M., Denchev, T. T. Mycobiota, 11, 1-10, 2021

Meira is a fungal genus that belongs to the *Exobasidiales* order. *Meira argovae* was originally described by Boekhout et al. (2003) [23] and validated by Denchev et al. (2021) owing to the invalid published name of the holotype [24].

Colonies were wrinkled with irregular margins and white in color after 3 d on YM agar at 25°C. Cells were ovoid to elongated with a size of $10.37 \times 2.40 \mu\text{m}$, and occurred singly or in pairs after 3 d on YM agar at 25°C. The budding was polar and on a narrow base. Hyphae were formed after two weeks of culture on Dalmau plates at 25°C (Fig. 4B-E). This species is sympodially branched and is usually a true hyphae [23].

Remarks: *Meira argovae* produces a chemical compound called argovin. There are no reports on the biological activity of argovin in organisms, although it can inhibit mites or *Agrobacterium tumefaciens* and has the potential for use in agricultural applications [25].

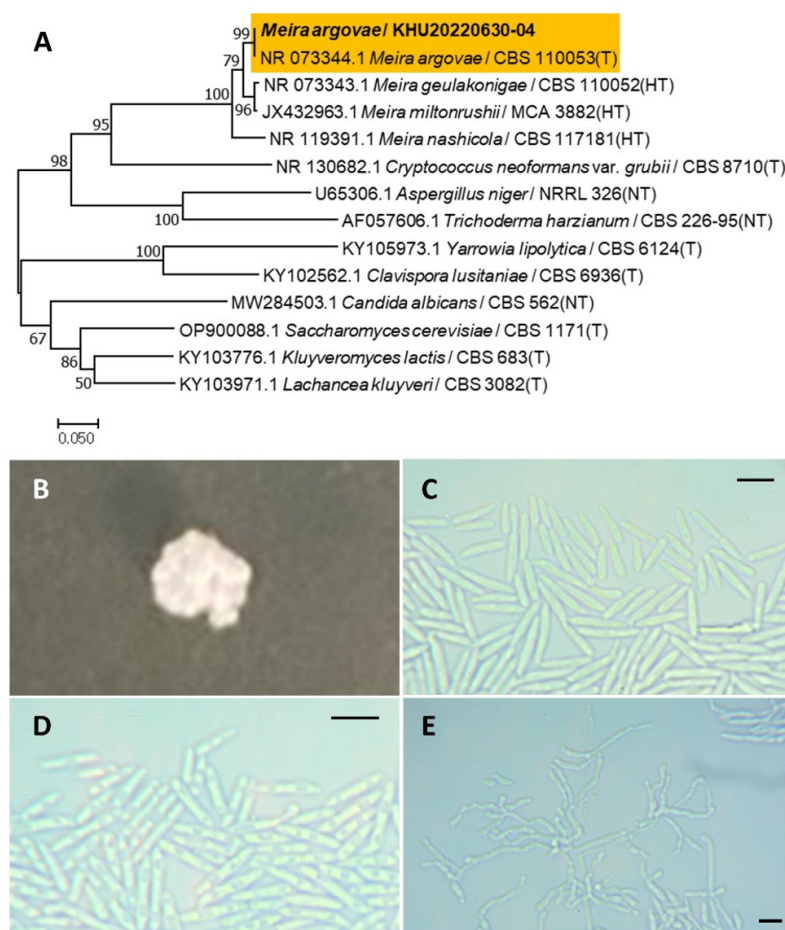


Fig. 4. Phylogenetic tree and morphological characteristics of *Meira argovae*. A. Phylogenetic tree drawn from neighbor-joining analysis based on sequence of the internal transcribed spacer region, showing positions of *M. argovae* strains isolated from Korea. Bold means representative strain. B-E. Morphology of *M. argovae* KHU20220630-04. B. Colony on yeast malt (YM) agar after 7 d at 25°C. C. Budding cells on YM agar after 3 d at 25°C. D. Budding cells occurring in short chains on YM agar after 7 d at 25°C. E. Hyphae formed on Dalmau plates containing cornmeal agar after 2 weeks. Scale bars, 10 μm. (T, Type strain; HT, Holotype; NT, Neotype.)

Kazachstania humilis (E. E. Nel & Van der Walt) Jacques N., Sarilar, V., Casaregola, S. Int. J. Syst. Evol. Microbiol., 66(12), 5192-5200, 2016

Kazachstania is a fungal genus belonging to the *Saccharomycetales* order. *Kazachstania humilis* has homotypic synonyms of *Candida humilis*. *Candida humilis* and *Candida milleri* are synonymous based on their similar nucleotide sequences [26]. However, it was later identified that *Candida humilis* and *Candida milleri* are not synonymous owing to their differences in the assimilation of sucrose and raffinose [27]. *Candida humilis* was reassigned as *Kazachstania humilis* based on sequence analysis [28].

Colonies were round with entire margins and ivory in color after 3 d on YM agar at 25°C. Cells were ovoid to elongated with a size of $6.98 \times 4.53 \mu\text{m}$, and occurred singly or in pairs after 3 d on YM agar at 25°C. The budding was polar and on a narrow base. Pseudohyphae were formed after two weeks of culture on Dalmau plates at 25°C. Ascospores were also observed (Fig. 5B-E). This species lacks pseudohyphae [27].

Remarks: *Kazachstania humilis* is a common dough-leaving agent for sourdough. This yeast is thought to be associated with other lactic acid bacteria in sourdough and cleaves dough [29].

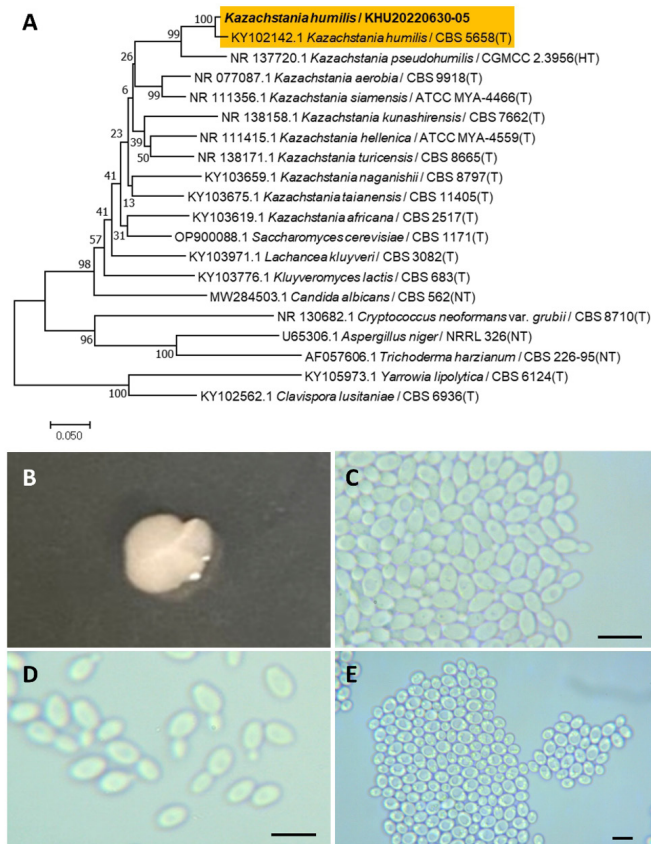


Fig. 5. Phylogenetic tree and morphological characteristics of *Kazachstania humilis*. A. Phylogenetic tree drawn from neighbor-joining analysis based on sequence of the internal transcribed spacer region, showing positions of *K. humilis* strains isolated from Korea. Bold means representative strain. B-E. Morphology of *K. humilis* KHU20220630-05. B. Colony on yeast malt (YM) agar after 7 d at 25°C. C. Budding cells on YM agar after 3 d at 25°C. D. Budding cells occurring in short chains on YM agar after 7 d at 25°C. E. Pseudohyphae and ascospores formed on Dalmau plates containing commmeal agar after 2 weeks. Scale bars, 10 μm . (T, Type strain; HT, Holotype; NT, Neotype.)

Meyerozyma smithsonii (S. O. Suh and Blackw.) A. M. Yurkov and G. Peter, Int. J. Syst. Evol. Microbiol., 67(10), 3977-3981, 2017

Meyerozyma is a fungal genus belonging to the *Saccharomycetales* order. *Meyerozyma* was originally described by Kurtzman et al., 2010 [30]. *Candida smithsonii* was identified and named by Suh et al. 2004 [31] and reassigned to *Meyerozyma smithsonii* by Yurkov et al. (2017) [32].

Colonies were round with entire margins and ivory to white in color after 3 d on YM agar at 25°C. Cells were ovoid to elongated with a size of $6.31 \times 3.00 \mu\text{m}$, and occurred singly or in pairs after 3 d on YM agar at 25°C. The budding was polar and on a narrow base. After two weeks of culture on Dalmat plates at 25°C (Fig. 6B-E), pseudohyphae and blastoconidia were formed. This species possesses pseudohyphae with blastoconidia [31].

Remarks: *Meyerozyma smithsonii* is unstudied, although the *Meyerozyma* genus can degrade the mycotoxin patulin or use crude oil at contaminated sites [32,33].

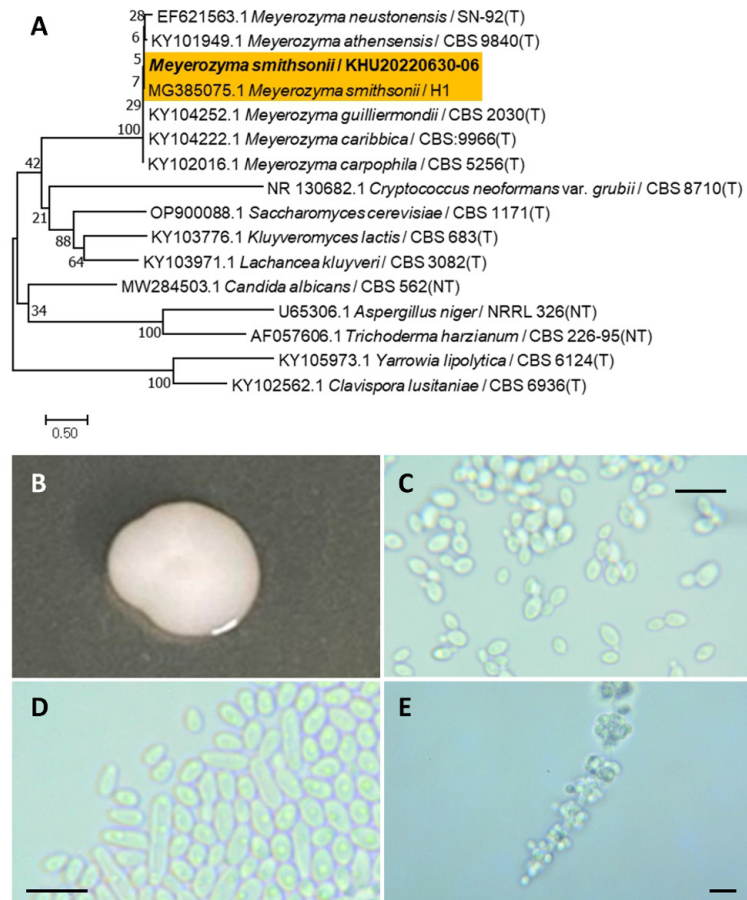


Fig. 6. Phylogenetic tree and morphological characteristics of *Meyerozyma smithsonii*. A. Phylogenetic tree drawn from neighbor-joining analysis based on sequence of the internal transcribed spacer region, showing positions of *M. smithsonii* strains isolated from Korea. Bold means representative strain. B-E. Morphology of *M. smithsonii* KHU20220630-06. B. Colony on yeast malt (YM) agar after 7 d at 25°C. C. Budding cells on YM agar after 3 d at 25°C. D. Budding cells occurring in short chains on YM agar after 7 d at 25°C. E. Pseudohyphae and blastoconidia formed on Dalmat plates containing commel agar after 2 weeks. Scale bars, 10 μm. (T, Type strain; NT, Neotype.)

Anthracocystis trispicatae (R. G. Shivas, Vanky & Athipunyakom) McTaggart, A., Shivas, R., Persoonia, 29(1), 116-132, 2012

Anthracocystis is a fungal genus belonging to the *Ustilaginales* order. *Anthracocystis trispicatae* was reassigned by McTaggart et al., 2012 [34], and has a basionym for *Sporisorium trispicatae* [35] and a homotypic synonym for *Sporisorium vermiculum* [36].

Colonies were wrinkled and ivory to white in color after 3 d on YM agar at 25°C. Cells were ovoid to elongated with a size of $13.38 \times 3.40 \mu\text{m}$, and occurred singly or in pairs after 3 d on YM agar at 25°C. The budding was polar and on a narrow base. Hyphae were formed after two weeks of culture on Dalmau plates at 25°C. Furthermore, ascospores were observed (Fig. 7B-E). This species has subglobose outer spores and globoid or slightly irregular inner spores [35].

Remarks: *Anthracocystis trispicatae* is unstudied, although *Anyhracocystis fluoculossa* is genetically close to *Anthracocystis trispicatae* and is known for its potential against *Fusarium* head blight in wheat [37].

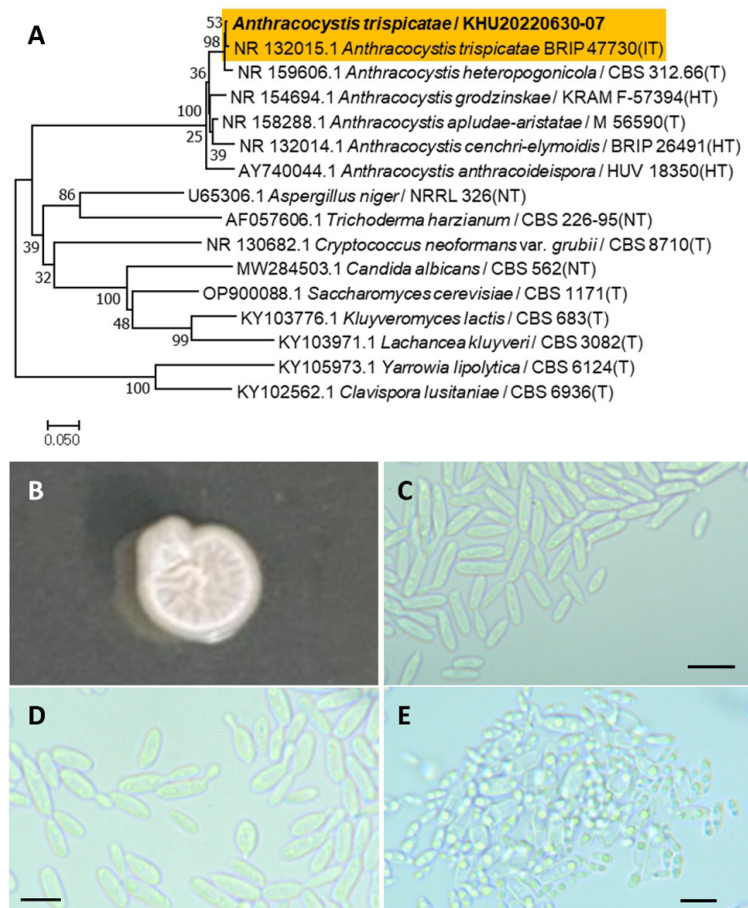


Fig. 7. Phylogenetic tree and morphological characteristics of *Anthracocystis trispicatae*. A. Phylogenetic tree drawn from neighbor-joining analysis based on sequence of the internal transcribed spacer region, showing positions of *A. trispicatae* strains isolated from Korea. Bold means representative strain. B-E. Morphology of *A. trispicatae* KHU20220630-07. B. Colony on yeast malt (YM) agar after 7 d at 25°C. C. Budding cells on YM agar after 3 d at 25°C. D. Budding cells occurring in short chains on YM agar after 7 d at 25°C. E. Hyphae and ascospores are formed on Dalmau plates containing cornmeal agar after 2 weeks. Scale bars, 10 μm . (T, Type strain; HT, Holotype; NT, Neotype; IT: Isotype.)

Tremella yokohamensis (Alshahni, Satoh & Makimura) Malysheva, V. F., Malysheva, E. F., Bulakh, E. M., Phytotaxa, 238(1), 40, 2015

Tremella is a fungal genus belonging to the *Tremellales* order. *Cryptococcus yokohamensis* was originally identified by Alshahni et al. (2011) [38] and reassigned by Malysheva et al. (2015) [39] based on micromorphological features.

Colonies were round with entire margins and ivory to white in color after 3 d on YM agar at 25°C. Cells were ovoid to elongated with a size of $7.72 \times 5.21 \mu\text{m}$, and occurred singly or in pairs after 3 d on YM agar at 25°C. The budding was polar and on a narrow base. After two weeks of culture on Dalmu plates at 25°C, teliospores were formed (Fig. 8B-E). This species has internal hyphae and irregular septa [39].

Remarks: *Tremella yokohamensis* is unstudied, although the *Tremella* genus produces polysaccharides. *Tremella* polysaccharides have a unique structure and activity with applications in the food, chemical, and medicinal industries [40].

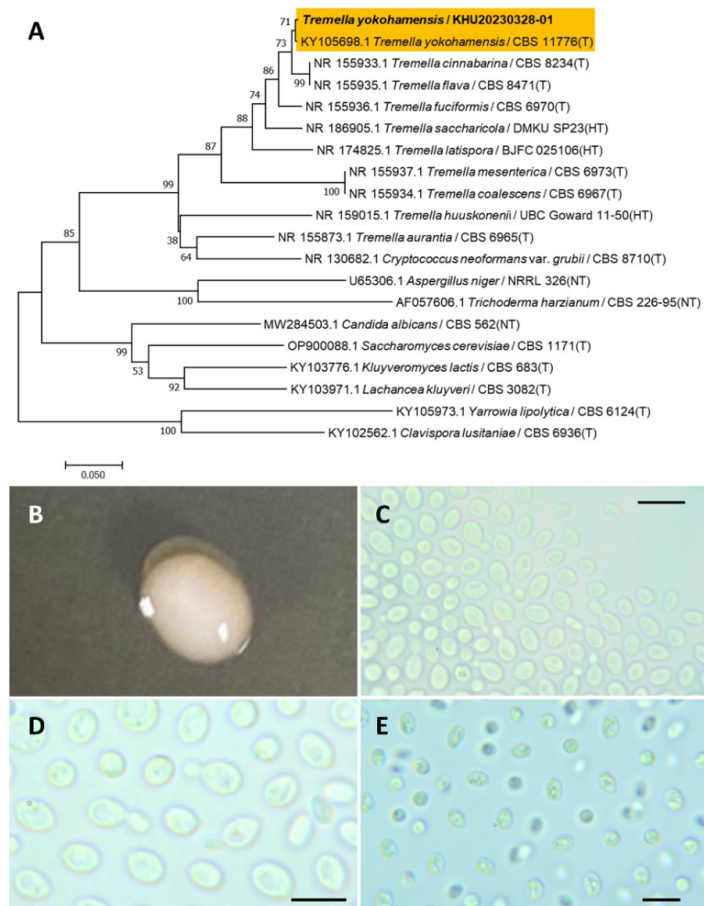


Fig. 8. Phylogenetic tree and morphological characteristics of *Tremella yokohamensis*. A. Phylogenetic tree drawn from neighbor-joining analysis based on sequence of the internal transcribed spacer region, showing positions of *T. yokohamensis* strains isolated from Korea. Bold means representative strain. B-E. Morphology of *T. yokohamensis* KHU20230328-01. B. Colony on yeast malt (YM) agar after 7 d at 25°C. C. Budding cells on YM agar after 3 d at 25°C. D. Budding cells occurring in short chains on YM agar after 7 d at 25°C. E. Teliospores are formed on Dalmu plates containing cornmeal agar after 2 weeks. Scale bars, 10 μm. (T, Type strain; HT, Holotype; NT, Neotype.)

Kwoniella shandongensis Chen, R., Yuan, F. X., Jiang, M., Wei, S. C., Groenewald, M., Wang, Q. M., Stud. Mycol., 96(1), 17-140, 2020

Kwoniella is a fungal genus belonging to the *Tremellales* order. *Kwoniella shandongensis* was first named by Chen et al. (2012) [41] by phylogenetic analysis. Li et al. (2020) validated *Kwoniella shandongensis* [20].

Colonies were round with entire margins and ivory in color after 3 d on YM agar at 25°C. Cells were ovoid to elongated with a size of $9.44 \times 6.96 \mu\text{m}$, and occurred singly or in pairs after 3 d on YM agar at 25°C. The budding was polar and on a narrow base. Teliospores were formed after two weeks of culture on Dalmau plates at 25°C (Fig. 9B-E). This species has no hyphae or sexual structures [41].

Remarks: *Kwoniella shandongensis* is poorly studied much, although different sequences of DNA polymerase A-related species are reported [42].

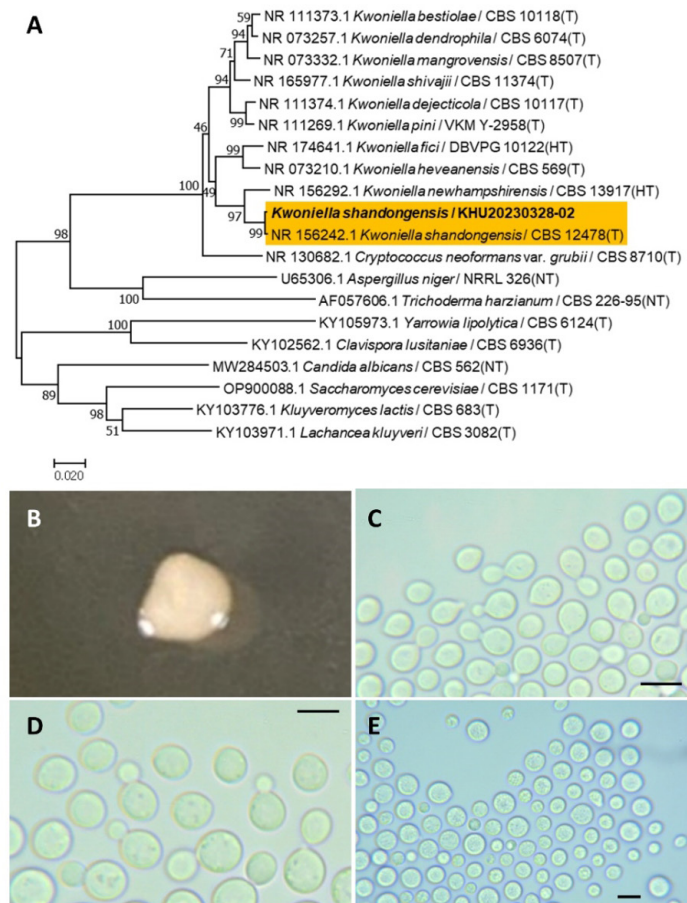


Fig. 9. Phylogenetic tree and morphological characteristics of *Kwoniella shandongensis*. A. Phylogenetic tree drawn from neighbor-joining analysis based on sequence of the internal transcribed spacer region, showing positions of *K. shandongensis* strains isolated from Korea. Bold means representative strain. B-E. Morphology of *K. shandongensis* KHU20230328-02. B. Colony on yeast malt (YM) agar after 7 d at 25°C. C. Budding cells on YM agar after 3 d at 25°C. D. Budding cells occurring in short chains on YM agar after 7 d at 25°C. E. Teliospores are formed on Dalmau plates containing cornmeal agar after 2 weeks. Scale bars, 10 μm. (T, Type strain; HT, Holotype; NT, Neotype.)

Kwoniella newhampshirensis Sylvester, K., Wang, Q. M., Hittinger, C. T., FEMS Yeast Res., 15(3), 7, 2015

Kwoniella is a fungal genus belonging to the *Tremellales* order. Sylvester et al. (2015) discovered *Kwoniella newhampshirensis* using phylogenetic analysis [43].

Colonies were round with entire margins and ivory in color after 3 d on YM agar at 25°C. Cells were ovoid to elongated with a size of $9.84 \times 6.40 \mu\text{m}$, and occurred singly or in pairs after 3 d on YM agar at 25°C. The budding was polar and on a narrow base. Ascospores were formed after two weeks of culture on Dalmau plates at 25°C (Fig. 10B-E). This species was reported as ballistoconidia and is not closely related to *Kwoniella shandongensis* [43].

Remarks: *Kwoniella newhampshirensis* is unstudied. However, *Kwoniella* produces quorum sensing inhibitors that can benefit from hybrid bioprocess optimization [44].

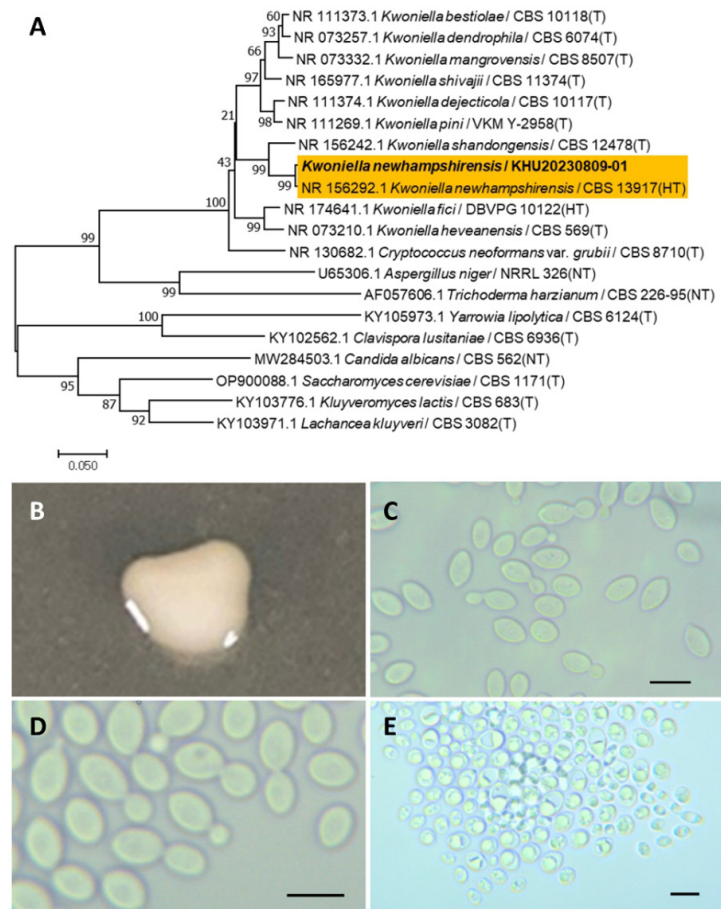


Fig. 10. Phylogenetic tree and morphological characteristics of *Kwoniella newhampshirensis*. A. Phylogenetic tree drawn from neighbor-joining analysis based on sequence of the internal transcribed spacer region, showing positions of *K. newhampshirensis* strains isolated from Korea. Bold means representative strain. B-E. Morphology of *K. newhampshirensis* KHU20230809-01. B. Colony on yeast malt (YM) agar after 7 d at 25°C. C. Budding cells on YM agar after 3 d at 25°C. D. Budding cells occurring in short chains on YM agar after 7 d at 25°C. E. Ascospores are formed on Dalmau plates containing cornmeal agar after 2 weeks. Scale bars, 10 μm. (T, Type strain; HT, Holotype; NT, Neotype.)

Aureobasidium proteae (Joanne E. Taylor & Crous) Taylor, J. E., Crous, P. W., Persoonia, 27(1), 20-45, 2011

Aureobasidium is a fungal genus belonging to the Dothideales order. *Aureobasidium proteae* was originally identified as a *Kabatiella proteae* by Taylor et al. (2000) [45]; however, the older generic name *Aureobasidium* is preferred over the younger and less known *Kabatiella* [46].

Colonies were round with radiating margins and ivory in color after 3 d on YM agar at 25°C. Cells are ovoid to elongated with a size of $15.92 \times 5.46 \mu\text{m}$ and occurred singly or in pairs after 3 d on YM agar at 25°C. Budding was bipolar with a narrow base. Pseudohyphae and blastoconidia formed after two weeks of culture on Dalmau plates at 25°C (Fig. 11B-E). This species is prominently constricted at the median septum with globose mucoid caps and ascospores observed [46].

Remarks: *Aureobasidium proteae* are pathogens responsible for chronic fungal meningitis in humans [47] and leaf blight in flowers [46].

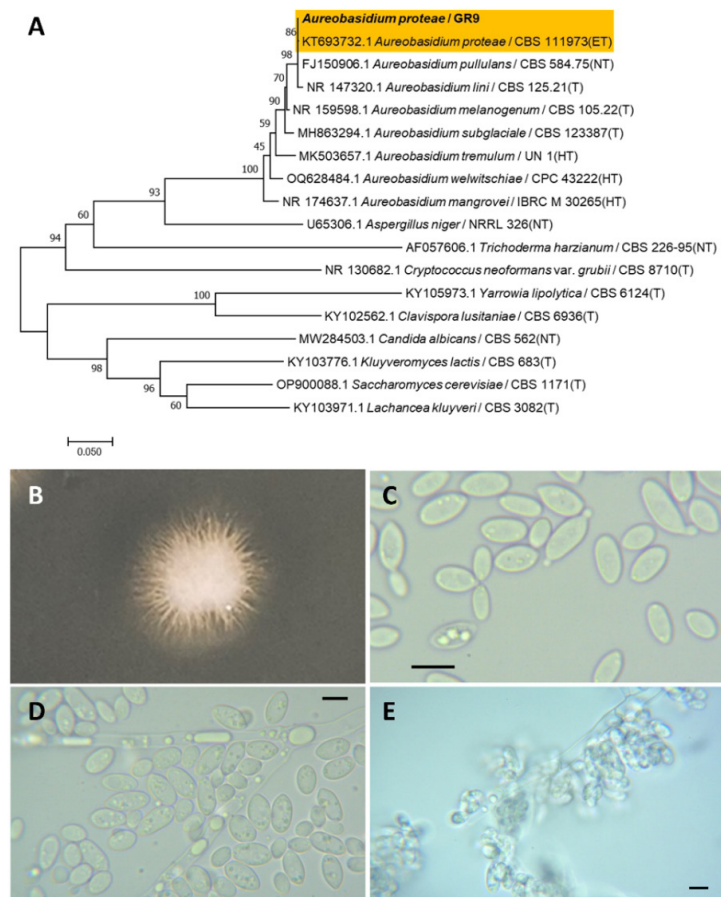


Fig. 11. Phylogenetic tree and morphological characteristics of *Aureobasidium proteae*. A. Phylogenetic tree drawn from neighbor-joining analysis based on sequence of the internal transcribed spacer region, showing positions of *A. proteae* strains isolated from Korea. Bold means representative strain. B-E. Morphology of *A. proteae* KHU20230809-02. B. Colony on yeast malt (YM) agar after 7 d at 25°C. C. Budding cells on YM agar after 3 d at 25°C. D. Budding cells occurring in short chains on YM agar after 7 d at 25°C. E. Pseudohyphae and blastoconidia are formed on Dalmau plates containing cornmeal agar after 2 weeks. Scale bars, 10 μm . (T, Type strain; ET, Epitype; HT, Holotype; NT, Neotype.)

Fellomyces penicillatus (Rodr. Mir.) Yamada, Y., Banno, I., J. gen. appl. Microbiol., 30(6), 524, 1984

Fellomyces is a fungal genus belonging to the *Tremellales* order. *Fellomyces penicillatus* was first reported by Yamada et al. (1984) [48]. Before this, it was named *Sterigmatomyces penicillatus* by Miranda et al. (1975) [49].

Colonies were wrinkled with entire margins and ivory to white in color after 3 d on YM agar at 25°C. Cells were ovoid to elongated with a size of $5.83 \times 4.23 \mu\text{m}$, and occurred singly or in pairs after 3 d on YM agar at 25°C. The bud type is hypha. Hyphae were formed after two weeks of culture on Dalmau plates at 25°C (Fig. 12B-E). A previous study did not observe hyphae and the cellular globose is ovoid [48].

Remarks: *Fellomyces penicillatus* is unstudied, although the *Fellomyces* genus is known for its long neck yeast and was used to inhibit conidiogenesis because it is rich in actin and cytoskeleton [50].

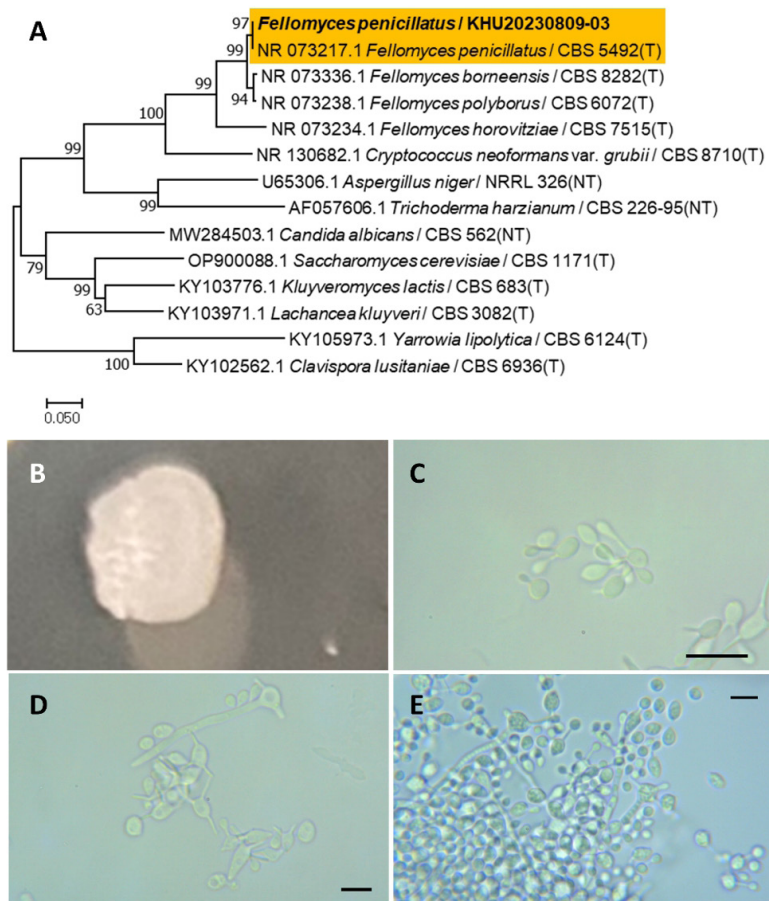


Fig. 12. Phylogenetic tree and morphological characteristics of *Fellomyces penicillatus*. A. Phylogenetic tree drawn from neighbor-joining analysis based on sequence of the internal transcribed spacer region, showing positions of *F. penicillatus* strains isolated from Korea. Bold means representative strain. B-E. Morphology of *F. penicillatus* KHU20230809-03. B. Colony on yeast malt (YM) agar after 7 d at 25°C. C. Budding cells on YM agar after 3 d at 25°C. D. Budding cells occurring in short chains on YM agar after 7 d at 25°C. E. Hyphae is formed on Dalmau plates containing commel agar after 2 weeks. Scale bars, 10 μm (T, Type strain; NT, Neotype.)

CONFLICT OF INTERESTS

The authors declare no competing interests.

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