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The generic circumscription of *Mrakia* and *Mrakiella*: The proposal of *Thomashallia* gen. nov.

Taweesak Malimas,[1] Huong Thi Lan Vu,[2] Pattaraporn Yukphan,[3]
Somboon Tanasupawat,[4] Yuzo Yamada,[3,5,6]

[1]Microbial Laboratory Biosafety Level-1, 46 M, 9 Nongphus, Muangsuang, Roi-Et 45220, Thailand

[2]Department of Microbiology, Faculty of Biology and Biotechnology, University of Science, Vietnam National University-HCM City, 227 Nguyen Van Cu Street, Ward 4, District 5, Hochiminh City, Vietnam

[3]Microbial Diversity and Utilization Team, Thailand Bioresource Research Center (TBRC), National Center for Genetic Engineering and Biotechnology (BIOTEC), National Science and Technology Development Agency (NSTDA), 113 Thailand Science Park, Phaholyothin Road, Klong 1, Klong Luang, Pathumthani 12120, Thailand

[4]Department of Biochemistry and Microbiology, Faculty of Pharmaceutical Sciences, Chulalongkorn University, 254 Phayathai Road, Wangmai, Pathumwan, Bangkok 10330, Thailand

[5]JICA Senior Overseas Volunteer, Japan International Cooperation Agency, Shibuya-ku, Tokyo 151-8559. Japan

[6]Laboratory of Applied Microbiology (Professor Emeritus), Department of Agricultural Chemistry, Faculty of Agriculture, Shizuoka University, 836 Ohya, Suruga-ku, Shizuoka 422-8529, Japan

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Abstract: **In the family Mrakiaceae, the type genus *Mrakia* senseu stricto included five teleomorphic species with the type species, *Mrakia frigida*. In contrast, the anamorphic genus *Mrakiella* sensu stricto contained nine species with the type species, *Mrakiella cryoconiti*. Between the two genera, the completely separated clusters were shown respectively in the phylogenetic tree based on the 28S rRNA gene D1/D2 domain sequences derived from the neighbour-joining method. Between *Mrakia frigida* and *Mrakiella cryoconiti* as well as *Mrakiella aquatica*, the pair-wise sequence similarities were 98.6% and 97.4% (1.2% width) respectively. However, the similarities between *Mrakia frigida* and the remaining four *Mrakia* species were extremely high (99.4 - 100% with 0.6% width). On the other hand, the similarity was relatively low (98.2%) between *Mrakiella cryoconiti* and *Mrakiella aquatica*, showing the wide range or the diversity of the anamorphic genus phylogenetically. The teleomorphic genus *Thomashallia* was newly introduced based on the formation of basidia and basidiospores with the type species, *Thomashallia stelviica*. The genus *Krasilnikovozyma* contained four species with the type species, *Krasilnikovozyma curviuscula*. Thus, the three teleomorphic genera were respectively taxonomic homogeneous-natured, and the two of the three were surely characteristic of Q-8.**

Keywords: ***Mrakia frigida*; *Mrakiella cryoconiti*; *Thomashallia* gen. nov.; *Thomashallia stelviica* comb. nov.; *Thomashallia montana* comb. nov.**

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Supplementary Abstract

The family Mrakiaceae Liu et al.	
Genus	Species
Mrakia Yamada et Komagata sensu stricto	*Mrakia frigida*^{T} (Fell et al.) Yamada et Komagata (1987)
	Mrakia gelida (Fell et al.) Yamada et Komagata (1987)
	Mrakia psychrophila Xin et Zhou (2007)
	Mrakia robertii Thomas-Hall et Turchetti (2010)
	Mrakia blollopis Thomas-Hall (2010)
Mrakiella Margesin et Fell sensu stricto	*Mrakiella cryoconiti*^{T} Margesin et Fell (2008)
	Mrakiella aquatica (Jones et Slooff) Margesin et Fell (2008)
	Mrakiella niccombsii Thomas-Hall (2010)
	Mrakiella arctica (Tsuji) comb. nov.
	Mrakiella hoshinonis (Tsuji et al.) comb. nov.
	Mrakiella fibulata (Yuekov et Turchetti) comb. nov.
	Mrakiella panshiensis (Jia et Hui) comb. nov.
	Mrakiella terrae (Park et al.) comb. nov.
	Mrakiella soli (Park et al.) comb. nov.
Thomashallia gen. nov.	*Thomashallia stelviica* (Turchetti et Buzzini) comb. nov.
	Thomashallia montana (Turchetti et Buzzini) comb. nov.
Krasilnikovozyma Liu et al.	*Krasilnikovozyma curviuscula*^{T} (Bav'eva et al.) Yurkov et al. (2019)
	Krasilnikovozyma huempii f.a. (Ramirez et Gonzalez) Liu et al. (2015)
	Krasilnikovozyma tahquamenonensis f.a. (Wang et al.) Liu et al. (2015)
	Krasilnikovozyma fibulata f.a. Gushkova et Kachalkin (2019)

The family Mrakiaceae Liu et al.	
Genus	Species
Mrakia Yamada et Komagata emend. Liu et al.	*Mrakia frigida*^{T} (Fell et al.) Yamada et Komagata (1987)
	Mrakia gelida (Fell et al.) Yamada et Komagata (1987)
	Mrakia psychrophila Xin et Zhou (2007)
	Mrakia robertii Thomas-Hall et Turchetti (2010)
	Mrakia blollopis Thomas-Hall et Turchetti (2010)
	Mrakia cryoconiti (Margesin et Fell) Liu et al. (2015)
	Mrakia aquatica (Jones et Slooff) Liu et al. (2015)
	Mrakia niccombsii (Thomas-Hall) Liu et al. (2015)
	Mrakia arctica Tsuji (2018)
	Mrakia hoshinonis Tsuji et al. (2019)
	Mrakia fibulata Yurkov et Turchetti (2020)
	Mrakia panshiensis Jia et Hui (2020)
	Mrakia stelviica Turchetti et Buzzini (2020)
	Mrakia montana Turchetti et Buzzini (2020)
	Mrakia terrae Park et al. (2021)
	Mrakia soli Park et al. (2021)
Krasilnikovozyma Liu et al.	*Krasilnikovozyma huempii*^{T} (Ramirez et Gonzalez) Liu et al. (2015)
	Krasilnikovozyma tahquamenonensis (Wang et al.) Liu et al. (2015)

The teleomorphic genus *Mrakia* Yamada et Komagata was separated from the genus *Leucosporidium* Fell et al. and introduced with *Mrakia frigida* as the type species on the basis of its characteristic isoprenoid quinone-8 (Q-8) (Yamada and Komagata 1987). Up to now, five species have been described; *Mrakia frigida*, *Mrakia gelida*, *Mrakia psychrophila* (Xin and Zhou 2007), *Mrakia robertii* (Thomas-Hall et al. 2010) and *Mrakia blollopis* (Thomas-Hall et al. 2010). In contrast, the anamorphic genus *Mrakiella* Margesin et Fell was introduced with the type species, *Mrakiella cryoconiti* (Margesin and Fell 2008), and the 11 species have been included.

Later, the genus *Mrakiella* was transferred taxonomically to the teleomorphic genus *Mrakia* with emendation (Liu et al. 2015). The genus *Mrakia* Yamada et Komgata emend. Liu et al. formed the monophyletic group along with *Krasilnikovozym*a, *Phaffia*, *Udeniomyces*, *Itersonilia* and *Tausonia* (Liu et al. 2015).

This paper is concerned with the revival of the anamorphic genus *Mrakiella* on the basis of the phylogenetic separation from the genus *Mrakia* emend. i.e., the teleomorphic and the anamorphic groups, the former of which corresponded to the genus *Mrakia* sensu stricto that was especially taxonomic homogeneous-natured and the latter of which did to the genus *Mrakiella* sensu stricto, from which the new teleomorphic genus *Thomashallia* was introduced with *Thomashallia stelviica*, the type species and *Thomashallia montana* by phylogenetic analyses (Yamada et al. 2022a).

The family Mrakiaceae Liu, Bai, Groenew et Boekhout, the order Cystofilobasidiales Fell, Roeijman et Boekhout:

Genus I. *Mrakia* Yamada et Komagata sensu stricto (MB25264)

One to three-celled metabasidium with basidiospores is shown (Fell 2011), extremely short phylogenetic branches are produced within the genus in the phylogenetic tree based on the 28S rRNA gene D1/D2 domain sequences and Q-8.

The type species is *Mrakia frigida*.

1. *Mrakia frigida* (Fell, Statzell, Hunter et Phaff) Yamada et Komagata (1987) (MB135389) Basionym: *Leucosporidium frigidum* Fell, Statzell, Hunter et Phaff (1969) The type strain is CBS 9136^{T}.

2. *Mrakia gelida* (Fell, Statzell, Hunter et Phaff) Yamada et Komagata (1987) (MB135390) Basionym: *Leucosporidium gelidum* Fell, Statzell, Hunter et Phaff (1969)

3. *Mrakia psychrophila* Xin et Zhou (2007) (MB508500)

4. *Mrakia robertii* Thomas-Hall et Turchetti (2010) (MB514690)

5. *Mrakia blollopis* Thomas-Hall (2010) (MB514691)

In the phylogenetic tree based on the 28S rRNA gene D1/D2 domain sequences (LSU D1/D2) derived from the neighbour-joining method, the clusters of the teleomorphic and the anamorphic species were completely separated from each other (Fig. 1). In addition, the phylogenetic branches of the five *Mrakia* species were extremely short, when compared with those of the anamorphic representative species, *Mrakiella cryyoconiti* and *Mrakiella aquatica*. It is suspected that the appearance of *Mrakia* species on the earth was relatively new from the viewpoint of evolutionary aspects.

The calculated pair-wise sequence similarities between the type species, *Mrakia frigida* and the remaining four *Mrakia* species were extremely high (99.4 - 100%; 0.6% width) (Table 1). In contrast, the sequence similarities were low and diverse (98.6 and 97.4%, respectively, 1.2% width) between *Mrakia frigida* and *Mrakiella cryyoconiti* as well as *Mrakiella aquatica*. The similarity was also very low (90.4%) between *Mrakia frigida* and *Krasilnikovozyma curviuscula*.

To introduce the taxonomic homogeneous-natured genus, the calculated pair-wise sequence similarities were 98% or more between *Kockiozyma suominensis* and *Myxozyma geophila* (= *Kockiozyma geophila* f.a.; Lipomycetaceae) (Yamada et al. 2022a) and between *Octosporomyces octosporus* (= *Schizosaccharomyces octosporus*) and *Octosporomyces osmophilus* (= *Schizosaccharomyces osmophilus*; Schizosaccharomycetaceae) (Vu et al. 2022a) in the 26S rRNA gene D1/D2 domain sequences. In the 18S rRNA gene sequences, 98% or more sequence similarities were also calculated to accomodate seven *Myxozyma* species to the teleomorphic genus *Kockiozyma* (Lipomycetaceae) (Vu et al. 2022b).

On the other hand, the calculated sequence similarities were 97.4 - 100% (2.6% width) in the genus *Mrakia* emend. Liu et al., indicating that the emended genus appeared to be taxonomic heterogeneous-natured.

From the data obtained above, the teleomorphic genus *Mrakia* sensu stricto was preferablly accepted but not discarded, since the sequence similarities were very high (99.4 - 100%, 0.6% width) in the family Mrakiaceae.

Genus II. *Mrakiella* Margesin et Fell sensu stricto (MB536881)

No metabasidium is shown (Fell and Margesin 2011), long phylogenetic branches are produced within the genus in a phylogenetic tree (LSU D1/D2) and Q-8

The type species is *Mrakiella cryoconiti*

1. *Mrakiella cryoconiti* Margesin et Fell (2008) (MB537002) The type strain is CBS 5443^{T}.

2. *Mrakiella aquatica* (Jones et Slooff) Margesin et Fell (2008) (MB514705) Basionym: *Candida aquatica* Jones et Slooff (1966)

3. *Mrakiella niccombsii* Thomas-Hall (2010) (MB514692)

4. *Mrakiella arctica* (Tsuji) comb. nov. Basionym: *Mrakia arctica* Tsuji, Mycoscience, 59: 57 (2018) (MB821502) The type strain is JCM 32070^{T}. MycoBank number is 848791.

5. *Mrakiella hoshinonis* (Tsuji, Tanabe, Vincent et Uchida) comb. nov. Basionym: *Mrakia hoshinonis* Tsuji, Tanabe, Vincent et Uchida, Int. J. Syst. Evol. Microbiol., DOI 10.1099/ijsem.0.003216: 4 (2019) (MB825484) The type strain is JCM 32575 ^{T}. MycoBank number is 848792.

6. *Mrakiella fibulata* (Yurkov et Turchetti) comb. nov. Basionym: *Mrakia fibulata* Yurkov et Turchetti, Antonie van Leeuwenhoek, 113: 506 (2020) (MB 830398) The type strain is DSM 103931^{T}. MycoBank number is 848793.

7. *Mrakiella panshiensis* (Jia et Hui) comb. nov. Basionym: *Mrakia panshiensis* Jia et Hui, Mycokeys, 74: 82 (2020) (MB834813) The type strain is NYNU 18562^{T}. MycoBank number is 848794.

8. *Mrakiella terrae* (Park, Maeng et Sathiyaraj) comb. nov. Basionym: *Mrakia terrae* Park, Maeng et Sathiyaraj, Mycobiology, 49: 470 (2021) (MB836844) The type strain is YP416^{T}. MycoBank number is 848795.

9. *Mrakiella soli* (Park, Maeng et Sathiyaraj) comb. nov. Basionym: *Mrakia soli* Park, Maeng et Sathiyaraj, Mycobiology, 49: 472 (2021) (MB836847) The type strain is YP421^{T}. MycoBank number is 848801.

In contrast to the teleomorphic species of the genus *Mrakia* sensu stricto, the anamorphic *Mrakiella* species represented relatively long phylognetic branches (Fig. 1), indicating that the evolutionary stages might be different from each other. Within the genus *Mrakiella*, it was noticiable that there were two subclusters; one was comprised of *Mrakiella cryoconniti* and *Mrakiella arctica*, designated as Group a, and the other was of *Mrakiella aquatica*, *Mrakiella panshiensis*, *Mrakiella terrae*, *Mrakiella hoshinonis*, *Mrakiella nicombsii*, *Mrakiella fibulata* and *Mrakiella soli*, designated as Group b.

The calculated pair-wise sequence similarities within the genus *Mrakiella* were obviously diverse (98.2 - 99.6%, 1.4% width) (Table 1) in contrast to the teleomorphic genus *Mrakia* sensu stricto (99.4 - 100%; 0.6% width).

In Group a of *Mrakiella*, *Mrakia stelviica* and *Mrakia montana* were reported to produce basidiospores from germinating teliospores (Turchetti et al. 2020). In addition, Zhang et al. (2020) also showed that in Group b of *Mrakiella Mrakia panschiensis* represented the teleomorphic stage, i.e., teliospores were produced and might germinate by a bud-like projection. For the former two species, the new genus was able to be introduced (Fig. 1).

Genus III. *Thomashallia* gen. nov.

MycoBank number is 848796.

Thomashallia (Tho.mas.hal'li.a, N.L. fem. n. *Thomashallia* Thomas-Hall, in honour of Dr. Skye Robin Thomas-Hall, University of New England, Armidale, Australia, who contributed largely to the systematics of yeasts, especially of psychrophilic yeasts).

The cells are elongate and budding is bipolar (Turchetti et al. 2020). Pseudohyphae are shown. After a long incubation, septate hyphae are observed with clamp connections. Teliospores are produced after long incubation, and germinating teliospores produce sessile bacilliform basidiospores (Turchetti et al. 2020). Glucose, sucrose and trehalose are fermented. Good growth is shown at 10ºC and 15ºC, but no growth is done at 25ºC (Turchetti et al. 2020).

The type species is *Thomashallia stelviica*.

1. *Thomashallia stelviica* (Turchetti et Buzzini) comb. nov. Basionym: *Mrakia stelviica* Turchetti et Buzzini, Int. J. Syst. Evol. Microbiol. 70: 4707 (2020) (MB835624). The type strain is DBVPG 10734^{T}. MycoBank number is 848797.

2. *Thomashallia montana* (Turchetti et Buzzini) comb. nov. Basionym: *Mrakia montana* Turchetti et Buzzini, Int. J. Syst. Evol. Microbiol. 70: 4709 (2020) (MB835626). The type strain is CBS 16462^{T}. MycoBank number is 848798.

The differentiation of the new genus *Thomashallia* from the genus *Mrakia* sensu stricto was able to be phenotypically done in the presence or absence of clamp connection and of fermentation of trehalose (Table 2).

In the phylogenetic tree based on the ITS sequences derived from the neighbour-joining method (Fig. 2), the cluster of the genus *Mrakiella* sensu stricto was completely divided into two. Of the two, one, i.e., Group a of *Mrakiella* including *Thomashallia stelviica*, *Thomashallia montana* and *Mrakiella cryoconiti* and *Mrakiella arctica* was connected to all the species of the genus *Mrakia* sensu stricto, however, the other, i.e., Group b of *Mrakiella* including *Mrakiella aquatica Mrakia panshiensis* and so on was not. Additionally, it is of interest that the phylogenetic branches of *Mrakiella aquatica* and its related species were shorter than those of the *Mrakia* species.

In the phylogenetic tree based on the concatenated sequences of the ITS and 28S rRNA gene D1/D2 domain derived from the neighbour-joining method, the similar topology was given to that of ITS only (the present authors' data not shown).

According to Tsuji et al. (2019), the calculated pair-wise ITS sequence similarities were also high (97.4 - 98.3%, 0.9% width) between *Mrakia frigida* and other four *Mrakia* species. Between *Mrakia frigida* and *Mrakiella cryoconiti* as well as *Mrakiella aquatica*, 94.9 and 92.3% sequence similarities were also calculated with 2.6% width. On the other hand, *Thomashallia stelviica* represented 94.4, 97.4, 90.3 and 99.6% sequence similarities respectively to *Mrakia frigida*, *Mrakiella cryoconiti*, *Mrakiella aquatic*a and *Thomashallia montana* (the present authors' unpublished data). From the results obtained above, the genus *Mrakia* and the genus *Thomashallia* were also obviously differentiated phylogenetically and phenotypically from each other (Table 2).

Thus, it is reasonable that a new genus was introduced for the two teleomorphic species, *Mrakia stelviica* and *Mrakia montana* (Turchetii et al. 2020) and for the one species, *Mrakia panshiensis* (Zhang et al. 2020) another new genus will be additionally done.

Genus IV. *Krasilnikovozyma* Liu et al. (2015) (MB812178)

Non-septate tubular metabasidium with sporidia is shown (Fell 2011), not so short phylogenetic branches are produced within the genus in a phylogenetic tree (LSU D1/D2) and Q-8

The type species is *Krasilnikovozyma curviuscula.*

1. *Krasilnikovozyma curviuscula* (Bav'eva, Lisichkina, Reshetova et Danilevitch) Yurkov, Kachalkin et Sampaio (2019) (MycoBank829125) Basionym: *Mrakia curviuscula* Bav'eva, Lisichkina, Reshetova et Danilevitc (2002) (MB529873) The type strain is CBS 9136^{T}.

2. *Krasilnikovozyma huempii* f.a. (Ramirez et Gonzalez) Liu et al. (2015) (MB812179)

3. *Krasilnikovozyma tahquamenonensis* f.a. (Wang et al.) Liu et al. (2015) (MB813656)

4. *Krasilnikovozyma fibulata* f.a. Glushakova et Kachalkin (2019) (MB829124)

According to Fell (2011), *Mrakia curviuscula* (= *Krasilnikovozyma curviuscula*) produced a non-septate tubular metabasidium with one to two sporidia, which appeared to differ morphologically from those of *Mrakia frigida* and *Mrakia gelida*.

Liu et al. (2015) introduced the genus *Krasilnikovozyma* as an anamorphic taxon, since the type species was designated as *Krasilnikovozyma huempii* (= *Cryptococcus huempii*). From the view-point of the traditional yeast systematics, it appeared to be problematic. Namely, the basic characteristics of living things on the earth are based on their reproduction, especially their sexual reproduction. Therefore, it is general that the teleomorphic genus has precedence over the anamorphic genus in the yeast systematics, and the name of the teleomorphic genus *Krasilnikovozyma* is able to be given to the corresponding anamorphic species (Lachance 2012).

In the four *Krasilnikovozyma* species, the calculated pair-wise 28S rRNA gene D1/D2 domain sequences were somewhat diverse (97.8 - 100% with 2.2% width) (Table 1).

As described above, the branches of the five species within the genus *Mrakia* were abnormally short in the phylogenetic tree based on the LSU D1/D2 sequences (Fig. 1). In addition, the sequence similarities of the five species were extremely high (99.4 - 100%, 0.6% width, Table 1), when compared with the combination of *Kockiozyma suomiensis* and *Myxozyma geophila* (= *Kockiozyma geophila* f.a.) in the family Lipomycetaceae (98.0%, Yamada et al. 2022a) and with the combination of *Octosporomyces osmophilus* (= *Schizosaccharomyces osmohilus*) and *Octosporomyces octosporus* (= *Schizosaccharomyces octosporus*) in the family Schizosaccharomycetaceae (98.1%, Vu et al. 2022a). In the SSU (small subunit) sequences, the calculated similarities were 99.2 - 99.8% (0.6% width) in *Kockiozyma suomienesis* and its related seven *Myxozyma* species in the family Lipomycetaceae (Vu et al. 2022b).

On the other hand, *Mrakiella cryoconiti* and *Mrakiella aquatica* constituted a single cluster, which differed from that of *Mrakia frigida*, the type species in the phylogenetic tree based on the LSU D1/D2 sequences (Fig.1). The calculated sequence similarities between *Mrakia frigida* (the type species) and *Mrakiella cryoconiti* (the type species) as well as *Mrakiella aquatica* were not so high (98.6% and 97.4%, respectively). From the phylogenetic point of view, the teleomorphic genus *Mrakia* and the anamorphic genus *Mrakiella* were not able to be combined to produce the genus *Mrakia* emend., since a taxonomic heterogeneous-natured taxon will be born.

Concerning the two teleomorphic species, *Thomashallia stelviica* and *Thomashallia montana* derived from the genus *Mrakiella*, there was not any drastic differences from the *Mrakia* species phenotypically (Table 2). The difference was found in the presence or absence of clamp connections and trehalose fermentation. It was probably due to the short-period evolution found in the psychrophilic yeasts, e.g., the calculated sequence similarities were 99.0% between *Mrakia firigida* and *Thomashallia stelviica* (Table 1), which basically differed from the fission yeasts that represented the long phylogenetic branches and the very low sequence similarities (Vu et al. 2022a); e.g., the calculated pair-wise sequence similarities were 84.9 - 90.5% among the genera *Schizosaccharomyces*, *Octosporomyces* and *Hasegawaea* in the family Schizosaccharomycetaceae, and which was also different from the Lipomycetaceous yeasts that showed the same phylogenetic phenomena (Yamada et al. 2022a); e.g., the calculated pair-wise sequence similarities were 81.6 - 97.5% among the 10 genera in the family Lipomycetaceae.

In the psychrophilic yeasts including *Mrakia* and *Mrakiella* species, the optimal growth temperature appeared to be almost the same (Table 2), i.e., 15 - 17ºC, the maximum temperature for growth was 20ºC and no growth was found at 25ºC. According to Tsuji et al. (2018), *Mrakiella arctica* (= *Mrakia arctica*) was able to grow at -3ºC. And at this temperature, *Mrakiella arctica* produced extracellular enzymes such as lipase, cellulase and protease. From the industrial point of view, the psychrophilic yeasts will be expected to be utilized for producing important materials at a low temperature.

Epilogue

In the classification of the yeasts, the present authors adopted the traditional method, i.e., the generic names of teleomorphs have precedence over those of anamorphs, since a large number of anamorphic species are isolated and described at the present time but the teleomorphic species are very few and hard to be discriminated from the anamorphs that have identical generic names. For example, the teleomorphic stage-equipped *Mrakia sterviica* has the common generic name *Mrakia* in spite of being phylogenetically distinct and outside the cluster of the type species, *Mrakia frigida* in the genus *Mrakia* emend. Liu et al. In this case, it is adequate that a different generic name is appropriately given to the species concerned, e.g., as *Thomashallia sterviica*.

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Conflicts of interest

The authors declare that there are no conflicts of interest.

Author contributions

Y.Y., T.M., H.T.L.V., P.Y. and S.T. designed the study. T.M. performed the main experiments. P.Y. instructed how to make the experiments. Y.Y. prepared the manuscript. The detailed discussions were made among Y.Y., T.M.,H.T.L.V., P.Y., and S.T.

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This work was preliminarily opened [5, 15].

Corresponding: Yuzo Yamada [0000-0002-2799-7045], e-mail: yamada-yuzo@nifty.com

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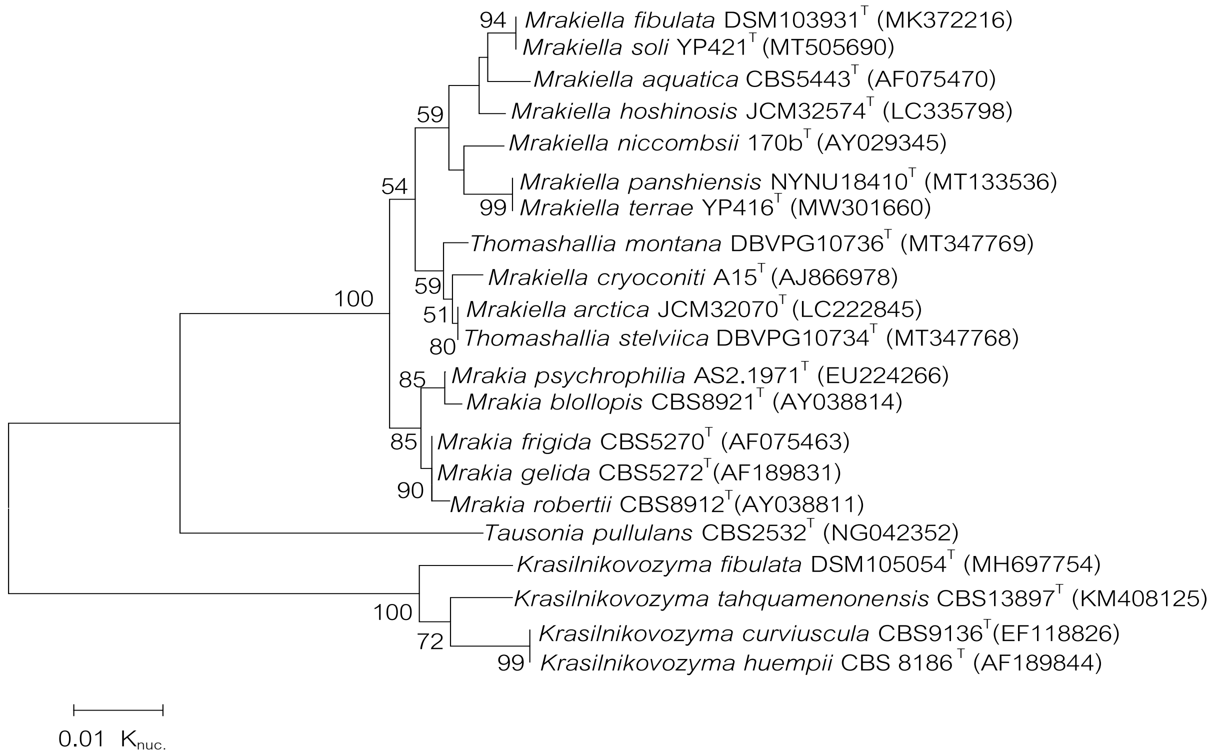

Fig. 1. The phylogenetic tree based on the 28S rRNA gene D1/D2 domain sequences (511 bases) derived from the neighbour-joining method for *Mrakia* and *Mrakiella* species. The numerals at the nodes of respective branches indicate bootstrap values (%) deduced from 1000 replications. For the construction of the phylogenetic tree, refer to Yamada et al. (2022a).

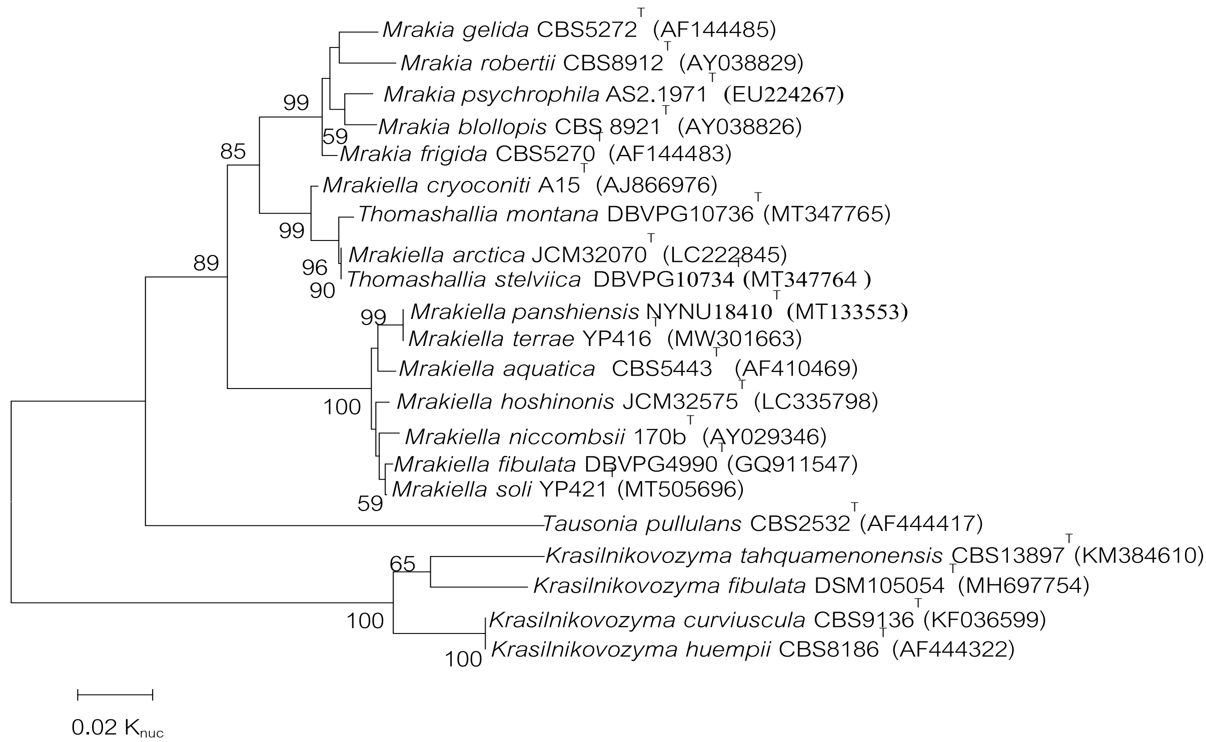

Fig. 2. The phylogenetic trees based on the ITS sequences (442 bases) derived from the neighbour-joining method for *Mrakia* and *Mrakiella* species. The numerals at the nodes of respective branches indicate bootstrap values (%) deduced from 1000 replications. For the construction of the phylogenetic tree, refer to Yamada et al. (2022a).

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Table 1. The pair-wise sequence similarity in *Mrakia*, *Mrakiella*, *Thomashallia* and *Krasilnikovozyma* species.

Species	*Mf*	*Mg*	*Mp*	*Mr*	*Mb*	*Mrc*	*Mrq*	*Ts*	*Tm*	*Mrp*	*Kc*
Sequence similarity (%)	100	100	99.6	99.8	99.4	98.6	97.4	99.0	98.6	97.6	90.4
Species	*Mrc*	*Ts*	*Tm*	*Mra*	*Mrp*	*Mrq*		*Kc*	*Kh*	*Kt*	*Kf*
Sequence similarity (%)	100	99.6	99.0	99.4	98.2	98.2		100	100	98.0	97.8
Species	*Mrq*	*Mrp*	*Mrn*	*Mrh*	*Mrf*	*Mrt*	*Mrs*		*Ts*	*Tm*	*Mra*
Sequence similarity (%)	100	98.4	99.0	98.8	99.2	98.4	99.2		100	99.4	99.8

The pair-wise 28S rRNA gene D1/D2 domain sequence similarities were calculated for 516 - 518 bases with the program BioEdit (version 7-2-5) (Hall, *GERF Biosci* **2**: 60, 2011). All strains examined were the type strains. *Mf*, *Mrakia frigida*; *Mg*, *Mrakia gelida*; *Mp*, *Mrakia psychrophila*; *Mr*, *Mrakia robertii*; *Mb*, *Mrakia blollopsis*; *Mrc*, *Mrakiella cryoconiti*; *Mrq*, *Mrakiella aquatica*; *Mrn*, *Mrakiella niccombsii*; *Mra*, *Mrakiella arctica*, *Mrh*, *Mrakiella hoshinonis*; *Mrf*, *Mrakiella fibulata*; *Mrp*, *Mrakiella panshiensis*, *Ts*, *Thomashallia stelviica*; *Tm*, *Thomashallia montana*; *Mrt*, *Mrakiella terrae*; *Mrsl, Mrakiella soli*; *Kc*, *Krasilnikovozyma curviuscula*; *Kh*, *Krasilnikovozyma huempii* f.a.; *Kt*, *Krasilnikovozyma tahquamenonensis* f.a; *Kf*, *Krasilnikovozyma fibulata* f.a.

Table 2. The phenotypic characteristics of *Mrakia*, *Thomashallia* and *Mrakiella* species.

Species	*Mfrig*	*Mgel*	*Mpsych*	*Mrober*	*Mbloll*	*Tstelv*	*Tmon*	*Melar*
Teliospore	+	+	+	+	+	+	+	-
Metabasidium (-celled)	1-3	1-3	1	1	1	1?	1?	-
Clamp connection	-	-	-	-?	-?	+	+	-
Fermentation of:								
Glucose	+	+	+	+	+	+	+	+
Sucrose	+	+	+	+	+	+	+	+
Trehalose	-	n	n	-	-	+	+	n
Growth at 17°C	+	+	+	+	+	+	+	+
at 20°C	w/-?	w?	+?	+	+	d	v	+
at 25°C	-	-	-	-	-	-	-	-

Abbreviations and data cited from: *Mfrig*, *Mrakia frigida* (Fell 2011); *Mgel*, *Mrakia gelida* (Fell 2011); *Mpsych*, *Mrakia psychrophila* (Xin and Zhou 2007); *Mrober*, *Mrakia robertii* (Thomas-Hall et al. 2010); *Mbloll*. *Mrakia blollopsis* (Thomas-Hall et al. 2010); *Tstelv*, *Thomashallia stelviica* (Turchetti et al. 2020); *Tmon*, *Thomashallia montana* (Turchetti et al. 2020); *Melar*, *Mrakiella arctica* (Tsuji et al. 2018).