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Addition to Microascales (Sordariomycetes, Ascomycota): Synnematotriadelphiaceae fam. nov., *Triadelphia mukdahanensis* sp. nov. (Triadelphiaceae) and the validation of Graphiaceae

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Abstract

A new species, *Triadelphia mukdahanensis* from northeastern Thailand, is introduced based on morphology and multi-gene phylogeny of combined ITS, LSU, SSU, and RPB2 sequence datasets. Phylogenetic analysis indicates that *T. mukdahanensis* is closely related to *T. heterospora* with high bootstrap support. However, *T. mukdahanensis* is distinct from *T. heterospora* and other exist species in conidial characteristics. In addition, the present study clarifies the current taxonomic placement of the genus *Synnematotriadelphia* based on phylogenetic analyses. The result shows that *Synnematotriadelphia* forms a distinct lineage in the clade comprising Triadelphiaceae and Graphiaceae. Therefore, a new family, Synnematotriadelphiaceae, is introduced to accommodate this distinct lineage and Graphiaceae is also validated herein.

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Introduction

Microascales was introduced by Benny & Kimbrough^[1]. Currently, seven families, Ceratocystidaceae, Chadefaudiellaceae, Gondwanamycetaceae, Graphiaceae, Halosphaeriaceae, Microascaceae, and Triadelphiaceae are accepted in the order^[2–5].

Graphiaceae was previously published by de Beer et al.^[6] with nine accepted *Graphium* species. However, the family is currently recorded as 'Nom. inval., Art. F.5.1 (Shenzhen)' with Editorial comment as 'An identifier issued by a recognized repository was not cited in the protologue" based on Index Fungorum^[7].

Triadelphiaceae was introduced to accommodate a monotypic genus *Triadelphia*^[8]. The family is characterized by hyphomycetous, holoblastic, monoblastic, fusiform to cylindrical or clavate, hyaline to pale brown conidiogenous cells, acrogenous with one to five different forms of conidia^[8–10]. Chuaseeharonnachai et al.^[2] introduced *Triadelphia hexaformispora*, a new species from a freshwater habitat in northern Thailand and a new genus *Synnematotriadelphia*, to accommodate two synnematous *Triadelphia* species, *T. stilboidea* and *T. synnematofera* under the new genus name, based on both morphology and molecular data.

Currently, 20 epithets are listed in *Triadelphia*^[7], being mostly reported from terrestrial habitats. *Triadelphia fusiformis, T. heterospora, T. hexaformispora* and *T. morgoensis* have been reported from freshwater habitats^[2,8]. In this study, a new *Triadelphia* species was collected from terrestrial decaying wood and is introduced based on both morphological

characteristics and multi-gene analyses. Morphological descriptions and illustrations are provided. The current taxonomic placement of Microascales is also clarified. In addition, a new family, Synnematotriadelphiaceae is introduced and Graphiaceae is validated herein.

Material and methods

Collection, examination, and isolation of fungi

Fresh samples were collected from decaying wood in a terrestrial habitat in Mukdahan Province, Thailand. The samples were processed following the methodology as described by Boonmee et al.^[11]. A Carl Zeiss GmbH stereo microscope (SteREO Discovery. V12, Germany) fitted with an AxioCam 105 color camera was used for image capture. Fungal micromorphology was examined using a Leica EZ4 Series stereomicroscope and photographed with a Nikon ECLIPSE 80i compound microscope (Nikon, Japan) fitted with a NikonDS-Ri2 digital camera (Nikon, Japan). Measurements of microscopic characters were calculated using the Tarosoft (R) Image Frame Work (IFW) software version 0.97 and a photoplate was made using Adobe Photoshop CS6 version 13.1.2. software (Adobe Systems, USA).

Single spore isolations were obtained following the methodology as described by Senanayake et al.^[12]. Germinating spores were aseptically transferred to fresh malt extract agar plates (MEA: 33.6 g/l sterile distilled water, Difco malt extract) and incubated at 25 °C. Cultures were incubated for 28 d and morphological characters, such as colour, colony shape, and

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texture were recorded. Type materials were deposited in the Herbarium of Mae Fah Luang University (Herb. MFLU), Chiang Rai, Thailand and Ex-type living cultures were deposited at Mae Fah Luang University Culture Collection (MFLUCC). Faces of fungi and Index Fungorum numbers were obtained as described in Jayasiri et al.^[13] and Index Fungorum^[7].

DNA extraction, PCR amplification and sequencing

Genomic DNA was extracted from fungal mycelium using PureLink[™] Quick Gel Extraction and PCR Purification Kit (Invitrogen by Thermo Fisher Scientific) following the manufacturer's instructions. Five gene regions were amplified with primer pairs as following: the internal transcribed spacer (ITS) region with primer pairs ITS5 and ITS4^[14], the large subunit nuclear ribosomal DNA (LSU) with primer pairs LROR and LR5^[15], the small subunit ribosomal DNA (SSU) with primer pairs NS1 and NS4^[14] and the partial RNA polymerase II subunit (RPB2) with primer pairs fRPB2-5F and fRPB2-7cR^[16].

The polymerase chain reaction (PCR) thermal cycle program for ITS, LSU and SSU amplification were initially 94 °C for 3 min, followed by 35 cycles of denaturation at 94 °C for 30 s, annealing at 55 °C for 50 s, elongation at 72 °C for 1.30 min and final extension at 72 °C for 10 min. The PCR thermal cycle program for RPB2 was initially 95 °C for 5 min, followed by 40 cycles of denaturation at 95 °C for 1 min, annealing at 52 °C for 1 min, elongation at 72 °C for 1.30 min and final extension 72 °C for 10 min. The quality of amplified PCR products was checked on 0.85% agarose gel electrophoresis. The PCR products were sent for sequencing at Macrogen Inc. (Geumcheon-gu, Seoul, Korea).

Phylogenetic analysis

Phylogenetic analysis of the combined aligned dataset of ITS, LSU, SSU, and RPB2 sequence data was analyzed based on maximum likelihood (ML) and Bayesian inference (BI) following the methodology as described by Mapook et al.^[17]. The sequence data of closest relative strains were selected and downloaded from GenBank based on the latest treatment and updated accounts of Microascales with updated accounts of the genus *Triadelphia* from recent relevant literature^[2,8,10]. The sequences used for analyses with GenBank accession numbers are given in Table 1. Phylogenetic trees were drawn using FigTree 1.4.2^[18] and edited by Microsoft Office PowerPoint 2019 and Adobe Photoshop CS6 (Adobe Systems, USA). The new nucleotide sequence data is deposited in GenBank.

Results

Phylogenetic analysis

The combined dataset of ITS, LSU, SSU, and RPB2 sequence data including our new strain were analyzed by maximum likelihood (ML) and Bayesian analyses. The combined sequence alignment is comprised of 37 taxa (3,520 characters with gaps) with *Falcocladium sphaeropedunculatum* (CBS 111292) and *F. multivesiculatum* (CBS 120386) as outgroup taxa. A best scoring RAxML tree with a final likelihood value of -20,343.554289 is presented in Fig. 1. The matrix had 1,454 distinct alignment patterns, with 40.85% of undetermined characters. Estimated base frequencies were as follows: A = 0.249759, C = 0.237431, G = 0.277058, T = 0.235751; substitution rates: AC = 1.533582, AG = 3.298266, AT = 1.550122, CG = 1.111961, CT = 6.988888, GT = 1.000000; gamma distribution shape parameter α = 0.206975.

In a BLASTn search of NCBI GenBank, the closest matches of the closest matches of the LSU, SSU and RPB2 sequence of *Triadelphia mukdahanensis* (MFLUCC 23-0049, ex-holotype) is *T. heterospora* (strain CBS 222.83) with 98.15% (MF434789), 99.85% (NG_067662), and 92.02% (MF434798) similarity, in respectively, while the closest matches of the ITS sequence with 91.86% similarity, was *T. loudetiae* (strain CBS 589.77, MF434776). Based on the phylogram generated from maximum likelihood analysis (Fig. 1) shows that our strain is closely related to *T. heterospora* with high bootstrap support (97% ML and 1.00 BYPP) and clustered with *T. loudetiae*. Interestingly, two *Synnematotriadelphia stilboidea* strains which introduced by Chuaseeharonnachai et al.^[2] form a well-supported separate clade with Triadelphiaceae and Graphiaceae, with high bootstrap support (99% ML and 1.00 BYPP).

Taxonomy

Graphiaceae Z.W. de Beer, Seifert & M.J. Wingf. ex Mapook, Boonmee & K.D. Hyde, *fam. nov*.

Index Fungorum number: IF 559717, Faces of fungi number: FoF 01099

Type genus: **Graphium** Corda, Icon. fung. (Prague) 1: 18 (1837)

Saprobic on plant and wood substrates, occasionally isolated from soil, manure, polluted water, sometimes causing wounds on tree bark or associated with beetles, and fungemia in an immunosuppressed child post stem-cell transplant. Sexual morph: Undetermined. Asexual morph: Hyphomycetous. Colonies on the natural substrate superficial, Conidiomata synnematous, determinate with pigmented stipes. Conidiophores macronematous, penicillately branches in two to three levels, compact, pigmented, septate with metulae present at the apex. Conidiogenous cells in whorls of two to six, enteroblastic, phialidic, with annellidic extensions. Conidia cylindrical to obovoid, hyaline, aseptate, sometimes slightly curved with age, truncate at base, often with a distinct basal frill, conidial mass produced in a transparent and slimy droplet. Rarely synanamorphic forming obovoid, monoblastic, with pigmented chlamydospore-like conidia (adapted from de Beer et al.^[6], Maharachchikumbura et al.^[19], Hyde et al.^[3]).

Type species: **Graphium penicillioides** Corda, Icon. fung. (Prague) 1: 18 (1837)

Holotype: CZECH REPUBLIC, Prague, on *Populus nigra* cv. *ilalica*, 14 December 1988, PR 155518; a slide from the holotype (DAOM 51800, isotype)

Epitype: CZECH REPUBLIC, České Budějovice, isolated from wood core of *Populus nigra* cv. *ilalica*, 3 September 1998, T. Kirisits, PRM 842988 (JCM 10498 = CBS J02632 = T. Kirisits No.3)

Morphological description and illustration: see Okada et al.^[20] and Hyde et al.^[3]

Notes: Graphiaceae was published by de Beer et al.^[6] with nine accepted *Graphium* species based on morphological description of a type species, *G. penicillioides* and available DNA sequence data from other know species^[20–22]. However, because no registration identifier was cited in the protologue, the family name is invalid (Nom. inval., Art. F.5.1)^[7]. In this study, Graphiaceae form a distinct family in the clade comprising Triadelphiaceae and Synnematotriadelphiaceae within Microascales (Fig. 1). Therefore, we validate Graphiaceae as a separate family in the order Microascales to accommodate the genus *Graphium* by providing a registration identifier and presenting the corrected type citations along with references to the original descriptions herein.

Addition to Microascales

Table 1. Taxa used in this study and their GenBank accession numbers. New sequences are in bold.

Таха	Strain no. ¹ —	GenBank accession numbers ²			
		ITS	LSU	SSU	RPB2
Acaulium albonigrescens	CBS 109.69	KY852469	KY852480	_	-
Ambrosiella xylebori	CBS 110.61	-	EU984294	AY858659	-
Ceratocystis fagacearum	CMW 2656	-	KM495341	-	-
Corollospora marina	AFTOL-ID 5008	-	AY762985	FJ176843	-
Corollospora ramulosa	PP1232	-	AF491276	-	-
Falcocladium multivesiculatum	CBS 120386 ^T	EU040217	JF831932	JF831928	-
Falcocladium sphaeropedunculatum	CBS 111292 [™]	JF831938	JF831933	JF831929	-
Faurelina elongata	CBS 126.78	GU291802	DQ368625	DQ368657	DQ368639
Faurelina fimigena	CBS 352.78 ^T	MH861149	-	-	-
Graphium fimbriasporum	CMW 5605 ^T	NR_155108	NG_067529	NG_064882	-
Graphium fimbriisporum	CMW 5606	AY148180	-	AY148172	-
Graphium laricis	CMW 5601 ^T	AY148183	NG_064370	AY148162	-
Graphium penicillioides	CBS 102632 ^T	KY852474	NG_067324	-	-
Graphium pseudormiticum	CMW 503 ^T	NR_136962	NG_064371	NG_064881	_
Knoxdaviesia capensis	AFTOL-ID 1907	-	FJ176888	FJ176834	-
Knoxdaviesia proteae	CBS 486.88 ^T	AY372072	AF221011	AY271804	-
Knoxdaviesia serotecta	CBS 129738 ^T	MH865536	MH876970	-	-
Knoxdaviesia ubusi	CBS 129742 ^T	MH865540	KM495395	-	-
Knoxdaviesia wingfieldii	CBS 132470 ^T	NR_111723	NG_042656	_	_
Microascus longirostris	AFTOL-ID 1237	-	-	DQ471026	-
Nimbospora effusa	JKI 5104A	-	U46892	U46877	DQ836887
Parascedosporium tectonae	CBS 127.84 ^T	MH861707	EF151332	U43907	-
Pseudoscopulariopsis schumacheri	CBS 435.86 ^T	LM652455	AF400874	-	-
Synnematotriadelphia stilboidea	CBS 101294	MF434783	MF434793	MF434811	MF434802
Synnematotriadelphia stilboidea	CBS 221.85 [™]	MF434781	MF434792	MF434810	MF434801
Triadelphia disseminata	CBS 138592 [™]	MF434784	MF434788	MF434806	MF434797
Triadelphia diversa	CBS 113.90 ^T	MF434782	MF434790	MF434808	MF434799
Triadelphia fusiformis	MFLUCC 16-0231 [™]	MH777097	MH777098	-	-
Triadelphia heterospora	CBS 222.83 ^T	MF434779	MF434789	MF434807	MF434798
Triadelphia hexaformispora	TBRC 9288 ^T	MK588842	MK588850	MK588840	MK578528
Triadelphia loudetiae	CBS 589.77 ^T	MF434776	MF434785	MF434803	MF434794
Triadelphia moubasheri	AUMC 10746 ^T	KY611849	-	_	_
Triadelphia mukdahanensis	MFLUCC 23-0049 ^T	OQ871494	OQ871495	OQ883648	OQ871561
Triadelphia pulvinata	CBS 590.77 ^T	MF434777	MF434786	MF434804	MF434795
Triadelphia pulvinata	CBS 744.84	MF434780	MF434787	MF434805	MF434796
Triadelphia romanica	CBS 162.79 ^T	MF434778	MF434791	MF434809	MF434800
Wardomyces anomalus	CBS 299.61	LN850992	LN851044	_	-

¹ AFTOL-ID: Assembling the Fungal Tree of Life; AUMC: Assiut University Mycological Centre; CBS: Westerdijk Fungal Biodiversity Institute, Utrecht, The Netherlands; CMW: Collection of the Forestry and Agricultural Biotechnology Institute (FABI), University of Pretoria, South Africa; JKI: Culture collection of the Julius Kühn-Institute, Siebeldingen, Germany; MFLUCC: Mae Fah Luang University Culture Collection, Chiang Rai, Thailand; TBRC: Thailand Bioresource Research Center; T: ex-type isolates. ² ITS: internal transcribed spacer regions 1 and 2 including 5.85 nrRNA gene; LSU: 28S large subunit of the nrRNA gene; SSU: 18S small subunit of the nrRNA gene; RPB2: partial RNA polymerase II second largest subunit gene.

Synnematotriadelphiaceae Mapook & K.D. Hyde, fam. nov.

Index Fungorum number: IF 559716, Faces of fungi number: FoF 14113

Type genus: **Synnematotriadelphia** Chuaseehar., Somrith., Nuankaew & Boonyuen, in Chuaseeharonnachai et al., Mycol. Progr. 19(2): 132 (2020)

Saprobic on a decayed petiole of a palm. **Sexual morph**: Undetermined. **Asexual morph**: Hyphomycetous. Colonies on natural substrata effuse, hairy, developing synnemata or hairylike structures in synthetic media. Mycelium partly superficial, hyaline to subhyaline, or olivaceous, septate, branched hyphae. Conidiomata synnematous, erect, simple or branched, solitary to gregarious, determinate, cylindrical or clavate, brown to blackish brown; parallel hyphae of synnemata septate, pale brown, with terminal short branches bearing single or fasciculate conidiogenous cells. *Conidiogenous cells* arising from metulae on the side of synnemata, holoblastic, monoblastic, solitary or aggregated, oblong to clavate, or spherical to subspherical, subhyaline to olivaceous. *Conidia* pleomorphic, solitary, acrogenous, produced in two or three different conidial forms on synnemata [as conidial type (a), (c) and (e) in *Triadelphia*], septate with transverse septum or septa, hyaline to pigmented, thin- to thick-walled (adapted from Chuaseeha-ronnachai et al.^[2]).

Type species: **Synnematotriadelphia stilboidea** (Mercado & R.F. Castañeda) Chuaseehar., Somrith., Nuankaew & Boonyuen, in Chuaseeharonnachai et al., Mycol. Progr. 19(2): 133 (2020)

Holotype: CUBA, Havana, Botanical Garden, on dead leaf petiole of *Roystonea regia*, 20 April 1982, A. Mercado, No. 6865, ex-holotype living culture CBS 221.85.

Morphological description and illustration: see Chuaseeha-ronnachai et al.^[2]

Addition to Microascales

97/1.00 Triadelphia heterospora CBS 222.83 Triadelphia mukdahanensis MFLUCC 23-0049 Triadelphia loudetiae CBS 589.77 Triadelphia hexaformispora TBRC 9288 Triadelphia diversa CBS 113.90 Triadelphia romanica CBS 162.79 Triadelphia moubasheri AUMC 10746 Triadelphia pulvinata CBS 744.84 Triadelphia disseminata CBS 138592 Triadelphia pulvinata CBS 590.77 Triadelphia fusiformis MFLUCC 16-0231	
97/1.00 89/0.99 98/1.00 99/1.00 99/1.00 Graphium fimbriisporum CMW 5606 Graphium laricis CMW 5601 Graphium pseudormiticum CMW 503 Graphium penicillioides CBS 102632	ales
100/1.00 Synnematotriadelphia stilboidea CBS 101294 Synnematotriadelphia stilboidea CBS 221.85	asc
97/- Knoxdaviesia capensis AFTOL ID 1907 94/0.96 Knoxdaviesia wingfieldii CBS 132470 100/1.00 Knoxdaviesia proteae CBS 486.88 Gondwanamycetaceae 100/1.00 Knoxdaviesia ubusi CBS 129742 Knoxdaviesia serotecta CBS 129738	Micre
100/1.00 Ambrosiella xylebori CBS110.61 100/1.00 Ceratocystis fagacearum CMW2656 Ceratocystidaceae	
98/0.92 Faurelina fimigena CBS 352.78 Faurelina elongata CBS 126.78 Chadefaudiellaceae	
Pseudoscopulariopsis schumacheri CBS 435.86 100/- Wardomyces anomalus CBS 299.61 87/1.00 Acaulium albonigrescens CBS 109.69 Microascaceae Microascus longirostris AFTOL 1237 Parascedosporium tectonae CBS 127.84	
100 1.00 Corollospora marina AFTOL 5008 96/1.00 Corollospora ramulosa PP1232 Halosphaeriaceae Nimbospora effusa IKI 5104A	
100/1.00 Falcocladium sphaeropedunculatum CBS 111292 Falcocladium multivesiculatum CBS 120386 Outgr	oups

0.08

Fig. 1 Phylogram generated from maximum likelihood analysis based on combined dataset of ITS, LSU, SSU and RPB2 sequence data of Microascales group. Bootstrap support values for maximum likelihood (ML) equal to or greater than 75% and Bayesian posterior probabilities (PP) equal to or greater than 0.90 are given above the nodes. A newly generated sequence and a newly introduced family are in blue bold. Type species are in bold. The tree is rooted with *Falcocladium sphaeropedunculatum* (CBS 111292) and *F. multivesiculatum* (CBS 120386).

Notes: Chuaseeharonnachai et al.^[2] introduced a genus *Synnematotriadelphia* to accommodate two synnematous *Triadelphia* species (*T. stilboidea* and *T. synnematofera*). Two *Synnematotriadelphia* formed a basal taxa to *Triadelphia* and were positioned in Triadelphiaceae^[2]. In our analyses, Synnematotriadelphiaceae forms a distinct family within Microascales in the clade comprising Triadelphiaceae and Graphiaceae (Fig. 1), with 99% ML and 1.00 BYPP bootstrap support. The three families differ as follows: Triadelphiaceae (type *Triadelphia*) has sporodochium-like structures, lack of conidiophores, conidia with one to five different forms, while Graphiaceae has synnematous, penicillately branched conidiophores, with cylindrical to obovoid, aseptate, hyaline conidia and Synnematotriadel-

phiaceae (type *Synnematotriadelphia*) has synnematous conidiophores with two or three different conidial forms on synnemata [as conidial type (a), (c) and (e) in *Triadelphia*].

Triadelphiaceae Y.Z. Lu, J.K. Liu, Z.L. Luo & K.D. Hyde, in Luo et al., Fungal Diversity: 10.1007/s13225-019-00438-1, [105] (2019)

Index Fungorum number: IF 555668, Facesoffungi number: FoF 05449

Morphological description and illustration: See Luo et al. (2019)

Type genus: *Triadelphia* Shearer & J.L. Crane, Mycologia 63(2): 247 (1971)

Triadelphia mukdahanensis Mapook, Huanraluek, Boonmee & K.D. Hyde, *sp. nov.* Fig. 2

Index Fungorum number: IF 900159, Faces of fungi number: FoF 14114

Etymology: Referring to the location where the specimen was collected, Mukdahan Province, Thailand.

Holotype: MFLU 23-0071

Saprobic on decaying wood. **Sexual morph**: Undetermined. **Asexual morph**: Hyphomycetous. *Colonies* on the natural substrate superficial, sporodochium-like structures, scattered, thinly diffuse, black. *Mycelium* partly superficial and partly immersed in the substratum, smooth-walled, with septate hyaline hyphae. *Conidiogenous cells* holoblastic, monoblastic, integrated, borne directly on the mycelium, globose to ampulliform, hyaline to light brown. *Conidia* solitary, smooth-walled, pleomorphic, forming dark brown to pale olivaceous when mounted in 5% KOH, three different forms (a, b, & f); form (a): (9.5–)18–25 × (3.5–)5–7(–8.5) µm ($\overline{\chi}$ = 17.5 × 5.5 µm, n = 25), oblong to cylindrical, (1–)2–septate, not constricted at the septum, thick-walled, straight or slightly curved, guttulate, septa covered with dark 1–2 µm bands, the apical and central cells brown to reddish brown, the basal cell light brown to brown; form (b): 20–30 × 7–12 µm ($\overline{\chi}$ = 26 × 9 µm, n = 25), pyriform to broadly clavate, (1–)2–septate, not constricted at



Fig. 2 *Triadelphia mukdahanensis* (holotype). (a)–(d) Appearance of colonies on substrate. (e)–(g) Conidiogenous cells and conidia (e and f mounted in 5% KOH). (f)–(k) conidial type (a). (l)–(n) Conidial type (b). (o)–(p) Conidial type (f). (q) Conidial type (a), (b) and (f). Scale bars: $a-c = 500 \mu m$, $d = 20 \mu m$, e-i, k-n, $q = 10 \mu m$. j, o, $p = 5 \mu m$.

the septum, thick-walled, straight or slightly curved, guttulate, the apical septum covered with dark (4–)5–6(–8.5) µm band, the basal septum covered with dark 1–3 µm band, the apical and central cells brown to dark reddish brown, the basal cell light brown to brown; form (f): 5–9 × 3.5–5 µm ($\bar{\chi} = 7 \times 4$ µm, n = 10), globose to subglobose, or oval, pale brown to brown, aseptate, guttulate.

Culture characteristics: Conidia germinating on MEA within 24 h at 25 °C and germ tubes produced from the ends. Colonies on MEA irregular, mycelium slightly raised, undulate, greyish to olivaceous-brown at the surface, dark olivaceous in reverse.

Material examined: THAILAND, Mukdahan Province, Wat Pa Sanam Thong, Phang Daeng, Dong Luang District, on decaying wood, 24 July 2019, S. Boonmee, (MFLU 23-0071, **holotype**), extype culture MFLUCC 23-0049.

Notes: *Triadelphia mukdahanensis* (MFLUCC 23-0049) was collected from a terrestrial habitat. Based on phylogenetic analysis, *T. mukdahanensis* is closely related to *T. heterospora* with 97% ML and 1.00 BYPP bootstrap support. *Triadelphia mukdahanensis* have overlapping size of conidial forms type (a) and (f) with *T. heterospora* and both lack conidial forms type (c) (Table 2). However, *T. mukdahanensis* is different from *T. heterospora* in having three different conidial forms type (a), (b) and (f), lacking of conidial forms type (d) and (e), while *T. heterospora* has four different conidial forms type (a), (d), (e) and (f) but lacks conidial forms type (b) (Table 2). In addition, a comparison of the ITS (+ 5.8S) gene region of *T. mukdahanensis* with *T. heterospora* reveals 33 base pair differences (5.6%) across 591 nucleotides, while RPB2 gene reveals 71 base pair differences (8%) across 890 nucleotides (not include gaps).

Discussion

Triadelphia is mostly found in terrestrial and rarely in freshwater habitats^[2,8]. Based on our phylogenetic results, our new strain grouped within *Triadelphia* and is closely related to *T. heterospora*. Simultaneously, we compared our new strain with the 20 epithets listed in Index Fungorum^[7], and found that

our new strain has the most morphological similarity to *T. heterospora. Triadelphia heterospora* was originally reported with two different conidial forms (a) and (d)^[23]. Subsequently, Constantinescu & Samson^[24] reported two additional conidial forms (e) and (f). Although our new strain has overlapping size of conidial forms type (a) and (f) with *T. heterospora* and both are lacking conidial forms type (c), our new strain is different in having three conidial forms type (a), (b) and (f), lacking conidial forms type (d) and (e) while *T. heterospora* have lacking reported of conidial forms type (b), which is appeared in our new strain. Therefore, we introduced a new species, *T. mukdahanensis* based on both morphology and phylogeny.

Interestingly, Synnematotriadelphia strains, which were positioned in Triadelphiaceae in Chuaseeharonnachai et al.^[2], form a well-supported separate clade to Triadelphiaceae and Graphiaceae within Microascales, based on combined dataset of ITS, LSU, SSU and RPB2 sequence data. Therefore, we introduce a new family, Synnematotriadelphiaceae to accommodate two Synnematotriadelphia species (S. stilboidea and S. synnematofera), based on both morphology and phylogeny. Synnematotriadelphiaceae (type Synnematotriadelphia) has synnematous conidiophores with two or three different conidial forms on the synnemata as conidial type (a), (c) and (e) *Triadelphia*^[2], while Triadelphiaceae (generic type in Triadelphia) has sporodochium-like structures, lack of conidiophores with one to five different types of conidial forms^[8–10], and Graphiaceae has synnematous with penicillately branched conidiophores, with cylindrical to obovoid, aseptate, hvaline conidia^[3,6,19]. This can be indicated that Synnematotriadelphiaceae have shared some characteristics from both Triadelphiaceae and Graphiaceae. However, Graphiaceae, which was introduced by de Beer et al.^[6], is recorded as an invalid name in Index Fungorum^[7] due to an identifier issued by a recognized repository was not cited in the protologue. In compliance with Article F.5.1^[25], Graphiaceae, is registered in Index Fungorum^[7] and obtained with a unique number. The family is validated by providing a registration identifier and presenting the corrected type citations along with references to the original descriptions herein.

Table 2. Conidial forms comparison of Triadelphia mukdahanensis and T. heterospora discussed in this study.

Conidial forms	T. mukdahanensis (this study)	<i>T. heterospora</i> (Shearer & Crane ^[23])	<i>T. heterospora</i> (Constantinescu & Samson ^[24])
Type (a): oblong to cylindrical, brown to dark brown, (1–)2–septate, septum covered with dark band	(9.5–)18–25 × (3.5–)5–7(–8.5) μm	(14.7–)16.2–19(–21.2) × 3.5–6 μm	15–23 × 5–8.5 μm
Type (b): pyriform to broadly clavate, or club-shaped, brown to dark brown, (1–)2–septate, the apical septum covered with broad dark bands, the basal cell subhyaline or light brown to brown	20–30 × 7–12 μm	-	-
Type (c): obclavate to acicular with a narrow long tip, hyaline or yellowish brown, multiseptate	-	_	-
Type (d): fusiform, obclavate with acicular tips, or rounded at the tip, multi-septate, end cells pale brown, central cells brown to dark brown, dark band covering over the central septa	-	17.6–23.5 × 8.2–10.6 μm	17.6–23.5 × 8.2–10.6 μm
Type (e): allantoid or reniform, hyaline or pale yellowish, 0–3- septate	-	-	8.5–15 × 3–5 μm
Type (f): obovate to broadly ellipsoidal, pale brown, unicellular	5–9 × 3.5–5 μm	-	6-8 × 4-5 μm

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Conflict of interest

The authors declare that they have no conflict of interest.

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