



***Kamalomyces polyseptatus* sp. nov. from an unidentified bamboo twig in Andaman Islands, India**

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Abstract

The examination of decaying twig samples fallen on the forest floor in the Andaman Islands, India yielded a new fungal species in the genus *Kamalomyces* (Tubeufiaceae). The new species, *Kamalomyces polyseptatus* is described in this paper supported by photomicrographs. The novel species is characterized by superficial, scattered, globose to sub-globose, dark brown to black ascomata covered by dense black mycelium, clavate asci, vermiform, elongate, transverse septate, hyaline ascospores. The new species *K. polyseptatus* is easily distinguishable from other species of the genus by the presence of higher number of septa in the ascospores. A synopsis of salient features of different species of *Kamalomyces* provided along with a dichotomous key to the known species of *Kamalomyces*.

Key words – Bamboo twigs – Dothideomycetes – Taxonomy – *Tubeufiaceae*

Introduction

The order Tubeufiales was introduced by Boonmee et al. (2014) based on morphology and molecular phylogeny to accommodate the family *Tubeufiaceae*. This family was established by Barr (1979) based on the type genus *Tubeufia* to accommodate bitunicate ascomycetes, with superficial ascomata surrounded by hyphae or setae, growing as saprobes on decaying wood and producing phragmosporous ascospores (Barr 1979, Phookamsak et al. 2017). Currently 25 genera have been accepted in this family (Wijayawardene et al. 2018) including *Acanthohelicospora*, *Acanthophiobolus*, *Acanthostigma*, *Acanthostigmina*, *Aquaphila*, *Berkleasium*, *Bifrontia*, *Boerlagiomyces*, *Chaetosphaerulina*, *Chlamydotubeufia*, *Dictyospora*, *Helicangiospora*, *Helicoma*, *Helicomycetes*, *Helicosporium*, *Kamalomyces*, *Muripulchra*, *Neoacanthostigma*, *Neohelicomyces*, *Neotubeufia*, *Podonectria*, *Tamhinispora*, *Thaxteriella*, *Thaxteriellopsis* and *Tubeufia*. Most of the genera have uniloculate, superficial, pigmented ascomata (Boonmee et al. 2014). Earlier *Tubeufiaceae* was placed in Pleosporales (Barr 1979) but was later transferred to Dothideales (Eriksson & Winka 1998). Subsequently, based on phylogenetic analysis of 28S rDNA, Kodsueb et al. (2006) placed the family in Pleosporales once again. Boonmee et al. (2011) revisited the family and examined the type and rDNA phylogeny, which showed that it has close relationship with Botryosphaerales. Finally, Boonmee et al. (2014) raised a new order Tubeufiales to accommodate *Tubeufiaceae* and added three new genera *Acanthohelicospora*, *Helicangiospora* and *Neoacanthostigma* with sexual and asexual morphs connections.

Kamalomyces was introduced by Verma et al. (2008) with *Kamalomyces indicus* as the type species recorded on bamboo. Recently three new species were added in *Kamalomyces*: *K.*

mahabaleshwarensis, *K. thailandicus* and *K. bambusicola* (Dubey & Neelima 2013, Phookamsak et al. 2017). *Kamalomyces* is distinct from the other genera of *Tubeufiaceae* (Phookamsak et al. 2017) in the occurrence of sub-globose to lemoniform ascomata with solitary-clusters surrounded by hyphae, broadly cylindrical to clavate asci and hyaline, vermiform, septate ascospores. *Kamalomyces indicus* and *K. thailandicus* have vermiform spores (Phookamsek et al. 2017) while the remaining two species: *K. bambusicola* and *K. mahabaleshwarensis* have non-vermiform ascospores (Phookamsak et al. 2017). Boonmee et al. (2014) proposed a key to the genera of *Tubeufiaceae*.

We are investigating the diversity of wood degrading, saprophytic, filamentous ascomycetous fungi from Andaman Islands, India. In our recent collection we found a specimen that has characters similar to the genus *Kamalomyces*. Upon closer examination it has been found that the new collection does not fit in any of the existing species in the genus *Kamalomyces* and hence it is reported as a new species in this paper.

Material and methods

Dead and decaying twig samples fallen on the forest floor in the reserved forests of North Andaman Islands, India were collected in January 2017 and transferred into zip lock plastic bags, air dried overnight, and packed into new plastic bags for shipment to the laboratory for further processing. Before undertaking the microscopic examination, the twigs were placed individually into plastic bread boxes lined with sterile tissue paper, rehydrated by sprinkling sterile water and incubated for one week to three months. The samples are then examined under a Stereo Zoom microscope (Optika SZM-LED, Italy) to locate the fungal fruiting structures. Hand sections were taken wherever necessary. The fruit bodies were cut with a razor and the spore constituents were transferred to a micro slide mounted with stains like Lacto phenol, Lacto phenol cotton blue, Lougal's reagent. These slides were then examined under the Nikon ECLIPSE TiU upright microscope with DIC objectives fitted with Nikon DS-Fi2 digital camera, Japan to take photomicrographs. Measurements were taken with Nikon NIS-Elements-Imaging Software version 4.4 program. Photo plates were prepared and edited with Microsoft power point, and Adobe Photoshop version 7.0. Morphological identification was carried out by referring to various individual publications (Verma et al. 2008, Dubey & Neelima 2013, Phookamsak et al. 2017, Kirk et al. 2008). The herbarium materials were deposited at Ajrekar Mycological Herbarium (AMH), Agharkar Research Institute (ARI), Pune, India.

Kamalomyces polyseptatus M. Niranjan and V.V. Sarma sp. nov.

Mycobank: MB824916

Etymology – With reference to the multiple septate ascospores

Saprobic on an un-identified bamboo twig. Sexual morph – *Colonies* 8–20×10–15 cm, superficial, surrounded by dense black, hyphal mat with swollen brown cells, apically raised, loosely connected, brown, septate, thick-walled, hyphae 4.6 µm wide. *Ascomata* 230–270 × 200–260 µm, perithecial, globose to sub-globose, basal dark brown, lateral and apical parts brown, immersed in the hyphae, coriaceous, scattered. *Peridium* 22.1 µm wider, consists two layers, outer rough, brown layer of *textura angularis* cells with munk pores and an inner layer of hyaline *textura angularis* cells. *Hamathecium*: *Pseudoparaphyses* 1.7µm wide, filamentous, anastomosing, longer than asci, interspersed within gelatinous matrix. *Asci* 154–224 (247.7) × 25.2–33.5 (\bar{X} = 186.4 × 29.5) µm (n=25), bitunicate, fissitunicate, broad clavate to clavate, with thick ocular chamber covered by smooth wall layer, pedicellate, pedicel long when young, becomes short at maturity. *Ascospores* 83–99.5 (104.2) × 6.8–8.4 (8.8) (\bar{X} = 91.3 × 7.7) µm (n=25), multi-seriate, vermiform, hyaline to pale brown, fusiform-clavate, apically obtuse, basally with narrow ends, mostly curved to straight, smooth-walled, two layered, phragmosporous, distoseptate, pleomorphic spores with 3 recognizable types, in first type apical 4-9 cells are wider, second type only 6th cell is wider, third type mostly 5th cell enlarged along with 11 and 12 or 16 and 18th also enlarged. Swollen cells are 8–9.9 µm (\bar{X} = 8.9) (n=25) wide. Asexual morph–Undetermined.

Known distribution – India

Material examined – India, Andaman and Nicobar Islands, North Andaman, Khalighat, (13°13'43.9"N 92°56'35.3"E). Isolated from an unidentified bamboo twig (Holotype AMH 9968), 6 January, 2017, Niranjana M & Sarma VV.

Notes – *Kamalomyces polyseptatus* is different from *K. indicus* in having hyaline ascospores with supramedian swollen cells (Figs 1, 2). Spores are similar to *K. bambusicola* excepting the presence of a supramedian swollen cell in *K. polyseptatus*. *Kamalomyces polyseptatus* has more septa than any other existing species (Table 1). *Kamalomyces polyseptatus* having the 5th cell swollen is similar to *K. bambusicola*, but differs from the latter in having two more cells that are wider such as 11th and 12th or 16 and 18th cells. *Kamalomyces polyseptatus* is different from *K. thailandicus* in having hyphal subiculum and more than one swollen cell and from *K. mahabaleshwarensis* in having larger ascospores and more septa (Table 1).

Discussion

The family Tubeufiaceae was established by Barr (1979) who placed it in Pleosporales and included 5 genera. Recently Boonmee et al. (2011) carried out a revision on this family and included 20 genera based on morphological and molecular criteria. Many of the genera of this family have uniloculate, superficial, pigmented ascomata e.g. pale brown, brown, and dark brown to black; multi-celled, hyaline ascospores, and produce, helicosporous asexual morph. Most species in this family are saprobic on terrestrial woody substrata (Boonmee et al. 2011, 2014). Based on their distinct morphology as well as combined LSU, SSU and TEF1 sequence analyses Boonmee et al. (2014) established a new order Tubeufiales to accommodate Tubeufiaceae. A dichotomous key has been provided by Boonmee et al. (2014) to delineate different genera in Tubeufiaceae. Our present collection fits very well into the genus *Kamalomyces* based on the morphological characters. Upon closer examination it was found that it does not fit into any of the existing species of the genus *Kamalomyces* and hence a new species *Kamalomyces polyseptatus* has been proposed to be accommodated in this genus. The differences among different species of *Kamalomyces* including *K. polyseptatus* have already been discussed in the 'notes' section above in addition to enlisting them in Table 1. There were slight overlapping characters found between *K. polyseptatus* and *K. indicus* in ascospore dimensions. However, when 'average length and width ratio' of ascospores was taken into consideration there were sharp differences found between these two species which are shown in Table 1 and also in the dichotomous key provided at the end. These were 99 x 9 µm in *K. indicus* vs. 91.3 x 7.7 µm in *K. polyseptatus*. This difference, in addition to the higher number of septa found in *K. polyseptatus* clearly distinguish *K. polyseptatus* from *K. indicus*. Hence a new species *K. polyseptatus* has been proposed to be accommodated in *Kamalomyces*, which is justified based on the above mentioned characters.

Table 1 Synopsis of important characteristics of different species belonging to *Kamalomyces*.

	Fungi	Ascomata	Asci	Ascospores	Septa no.
1	<i>Kamalomyces indicus</i>	(169–) 216.5–253 × (184–) 225.5 – 295.5 (–331) µm	(140–) 157–214 (–223) × (27.5) 29–36 (–38) µm	(92–) 95.5 – 104.5 (–107) × 8– 10.5µm (X̄ = 99 × 9µm)	38–42
2	<i>Kamalomyces mahabaleshwarensis</i>	320-460 × 360-450 µm	170–290 × 24–34 µm	55-86 × 8-12 µm	20
3	<i>Kamalomyces bambusicola</i>	250 – 300 × 220–300 µm	(150–) 180–190 (–215) × (21–) 23–28 (–30) µm	(57–) 75 – 90 (–98) × 4–7 (X̄ = 83.8 × 5.5) µm	(20–) 27–30
4	<i>Kamalomyces thailandicus</i>	180 – 250 × 250–280 µm	(174–) 185–220 (–243) × (32–) 35–42 µm	(100–) 120 – 135 (–155) × 8–10 (X̄ = 129 × 8.8 µm)	(19–) 33–36
5	<i>Kamalomyces polyseptatus</i>	230–270 × 200 – 260 µm	154–224 (247.7) × 25.2–33.5 µm	83 – 99.5 (104.2) × 6.8–8.4 (8.8) (X̄ = 91.3 × 7.7) µm	42–46



Fig. 1 – *Kamalomyces polyseptatus* (AMH-9968 Holotype). a Colony on host. b Vertical section of subiculum. c Hypha d Peridium. e Pseudoparaphyses. f, g Ascomata. h Transverse section of asci. i Peridium *textura angularis*. j peridium layer with monk pores. k Ascus with apical chamber. l-m Asci. Bars – b = 200 μm , f = 100 μm , l, m = 50 μm , c-e, h, k = 20 μm , g, i, j = 10 μm .

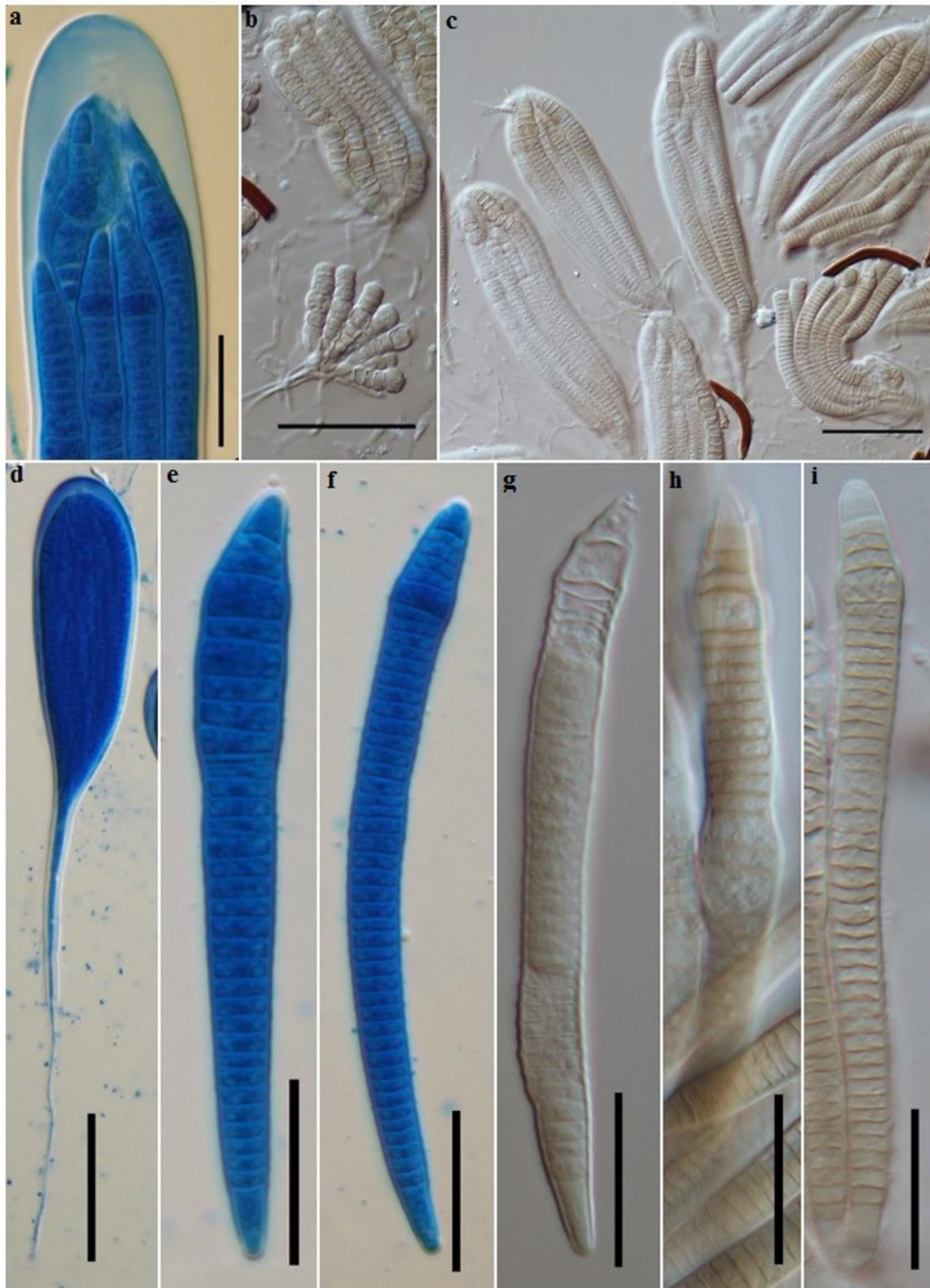


Fig. 2 – *Kamalomyces polyseptatus* a-d Asci. e-i Ascospores. (e=type 1, f,g type 2, a,g,h type 3)
 Bars – b–d = 50µm, a, e–i = 20 µm.

Dichotomous key to different species of *Kamalomyces*

- 1. Ascospores with less than 30 septa2
- 1' Ascospores with more than 30 septa3
- 2. Ascospores dia more than 8 µm*K. mahabaleshwarensis*
- 2' Ascospores dia more than 8 µm*K. bambusicola*

3. Ascospores mostly with more than 42 septa, average dia is 7.7µm*K. polyseptatus*
 3' Ascospores mostly with less than 42 septa, average dia is above 8.8 µm4
 4. Ascospores with less than 36 septa, average length of ascospores is 129µm*K. thailandicus*
 4' Ascospores with more than 36 septa, average length of ascospores is 99 µm*K. indicus*

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