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A reappraisal of the fungus genus *Phalangispora* with the rediscovery of *P. bharathensis* on leaf litter of *Mangifera indica* from the northern Western Ghats, India

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During July 2010 a survey was conducted to explore the microfungal diversity in the natural forests of Tamhini Ghats situated in the northern part of the Western Ghats, India, at 18^{0}27'N & 73^{0}25'E. The average altitude of the area is 600m, while the surrounding hilltops range from 850–1050 m. The hilly regions show little primary evergreen forest restricted to sacred groves and comparatively more secondary evergreen and moist deciduous forests (Dahanukar & Padhye 2005). Tamhini Ghats and surrounding areas are rich and diverse for their fungal diversity; many new species including a recent new genus *Tamhinispora* Rajeshkumar & Rahul Sharma were identified from Tamhini Ghat valleys (Rajeshkumar et al. 2011a,b; Rajeshkumar & Singh 2012; Rajeshkumar & Sharma 2013). An obscure species of *Phalangispora* Nawawi & Webster form sporodochia with stauroconidia with branches radiating or up (Seifert et al. 2011) and sporodochial setae surrounding the conidiomata was found on fallen leaves of *Mangifera indica*.

The genus *Phalangispora* was described by Nawawi & Webster (1982) for a fungus having branched tetra-radiate conidia collected from the water forms of Ulu Gaombak in Malaysia and sporulating cultures on 2% Malt Extract Agar (IMI 256650). They established the genus *Phalangispora* with key characteristics 'Mycelium primo subhyalinum, postea brunneo olivaceum, septatum, Conidiomata (sporodochia) pustulata aut obconica primo albida postea olivacea, sicca apparentia, laxa, ca. 5mm lata, ad basim cum 1–5 seris, Setare brunneae vel attae, septatae, parietibus crassis, subulatae, Conidia ramose, septate, constricta, brunnea'; with type species *P. constricta* Nawawi & Webster. The present study aims at an addition of new characteristics found in this genus in nature which is unique in the new strain and a rediscovery of *P. bharathensis* Keshava Prasad & Bhat with a varied conidial and conidial chain dimensions found on a new substrate (dead fallen leaves of *Mangifera indica*), collected from Tamhini Ghats, Maharashtra State, India.

Methods: Isolates and morphology: Sporodochia of the fungus were isolated from the lower surface of fallen leaves and observed under a Nikon Binocular stereo microscope (Model SMZ-1500 with Digi-CAM, Japan). For morphotaxonomic studies and photomicrographs an Olympus CX-41 (Japan) microscope was used. Conidia, conidiophores, and setae were measured using an ocular micrometer. The specimens were deposited in Ajrekar Mycological Herbarium (AMH 9580), Agharkar Research Institute, Pune, India.

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Results

Phalangispora bharathensis Keshava Prasad & Bhat 2002 (Image 1 and Fig. 1)

Type description: Terrestrial litter hyphomycete. Colonies effuse, olivaceous brown to dark brown. Mycelium partly superficial, hyphae 2.5–3.5 µm wide. Conidiomata sporodochial, solitary, pulvinate, slightly elevated, with 8–16 setae arising from the margin of the base. Setae subulate, acute at apex, septate, thick walled, dark brown, smooth, 300–400 × 7–10 µm. Conidiophores mononematous, arising in groups, septate, 1–2 times branched, 15–30 µm long, 2–4.5 µm wide. Conidiogenous cells polyblastic, terminal, hyaline, smooth with denticulate scars at the rounded apex. Conidia hyaline, smooth, aseptate, in 2–3 branched chains of 75–85 µm long, 2.5–4 µm wide, connected by narrow isthmi, uniseriate below, bi to tri seriate above, with branches arising from the third or fourth cells of the main axis, of two types; apical or basal cells conical to obclavate, 7–9 × 2.5–3 µm; intermediate cells cylindrical with truncate ends, 8–10 × 2.5–4 µm; in mass initially whitish, later becoming pale brown.

Type specimen: Herb No. IMI 387091, 11.iii.1999, on decaying leaves of *Holigarna arnotiana* (Wt. & Arn.) Hook.f., Cotigao Wildlife Sanctuary, Goa, India, coll. Keshava Prasad.

Present collection: AMH 9580. On fallen decaying leaves of *Mangifera indica*, Mycelium semi-immersed, 2–3 µm wide, pale brown, septate, branched, thin walled, smooth. Conidiomata sporodochial, flattened, yellowish central part with white to off white marginal area, spherical or hemispherical, 100–240 µm diam. Setose, setae 155–250 × 6–7.25 µm, 9–28 in numbers per sporodochia, arranged around the sporodochium, pale to dark brown or blackish, tapering towards the acute apex, paler towards tip, septate, septa thick walled, base wider 12–15 µm, swollen with rhizoid like structures, wall smooth and thick. Conidiophores present, highly branched, with primary and secondary branches, cylindrical, hyaline smooth, primary branches 16–25 × 3–3.5 µm, secondary branches 8–12 × 2.5–3.5 µm, Phialides cylindrical, smooth, hyaline 6–8 × 2.5–3

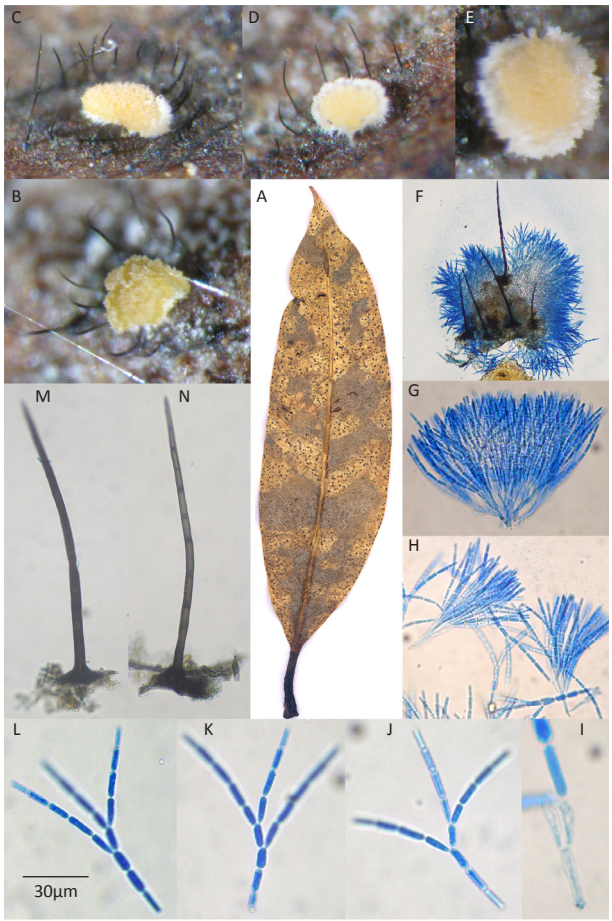

Image 1. *Phalangispora bharathensis,* AMH 9580.
A - Habit; B–E - Sporodochia with surrounding setae; F - Sporodochia top view; G–H - Conidiophores bunches with setae and conidia; I - Conidial attachment to condiogenous cells; J–L - Conidia; M–N - Setae. © K.C. Rajeshkumar

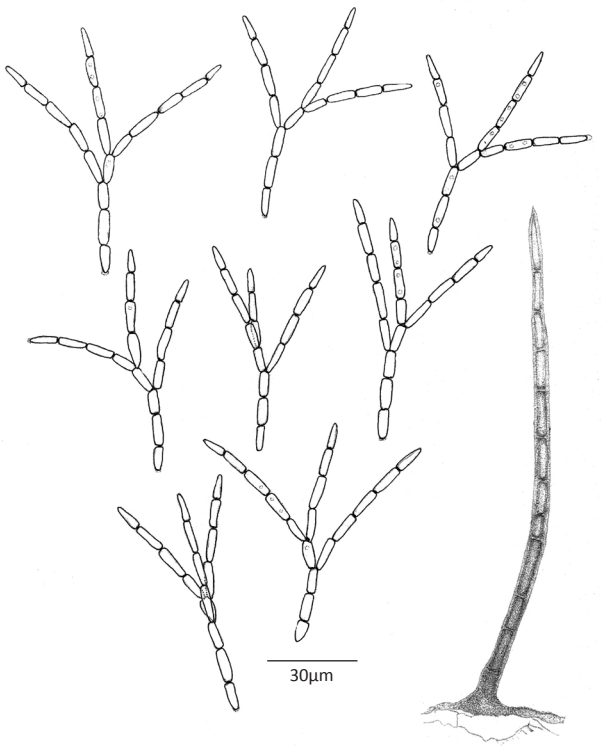

Figure 1. *Phalangispora bharathensis,* AMH 9580.
A - Variation in conidial shape; B - Setae.

µm, tip slightly bulged with two apical conidiogenous loci. Conidia holoblastic, hyaline, connected in chain through isthmi, branched, constricted at septa, overall conidia measure up to 74–87.5 × 2–4.5 µm, uniseriate below, bi to tri seriate above, with branches arising from the third and/or fourth cells of the main axis, conidia are two types; apical or basal cells conical, 8–11.3 × 2–3.3 µm; intermediate cells cylindrical with truncate ends, 9.5–12.5 × 3–4.5 µm. Y shaped conidia were also occasionally observed.

Material examined: AMH 9580, 30.vii.2010, on fallen decaying leaves of *Mangifera indica* L. (Anacardiaceae), Tamhini Ghats, Maharashtra State, India, coll. K.C. Rajeshkumar.

Teleomorph: Unknown.

Known distribution: Validly published from Malaysia and India

Notes: The arrangement of dark brown, septate sporodochial setae surrounding the sporodochia is a unique character in this strain that differentiates it from all the species so far recorded in this genus.

Discussion: Genus *Phalangispora* is a unique setose sporodochial hyphomycete producing hyaline smooth, unicellular conidia connected by narrow isthmi in branched chains on polyblastic, discrete conidiogenous cells and short, thin walled, hyaline, septate conidiophores. While authenticating the type culture of *Phalangispora* in IMI (IMI 256650), BC Sutton (Nawawi & Webster 1982) had an opinion that the isolate made from the forms in Malaysia belongs to a group of genera which include *Wiesneriomyces* Koord., *Speiropsis* Tubaki, *Gliophragma* Subram. & Lodha and *Pseudogliophragma* Phadke & V.G. Rao. However, *Phalangispora* was related to *Speiropsis*, having branched conidial morphology, but differs from it in conidiomatal structure. He also mentioned the difference found in the presence or absence of setae in these genera; *Speiropsis* and *Pseudogliophragma* lack setae; however, *Wiesneriomyces* have sporodochial setae with simple chained conidia. Prior to this inventory, the conidia of this fungus have been reported from many different countries including Nigeria (Ingold 1959), Ghana (Dixon 1959), Sierra Leone (Le-John 1965) Japan (Miura 1974) and Indonesia (Nayo 1975) but a valid recognition was not made.

Based on the sporodochial development of *P. constricta* on dry Agar, Nawawi & Webster (1982) predicted the possibility of this fungus distribution in nature out of water. Later on, Nawawi (1985) reported that the fungus is semi-aquatic that readily grows on decaying leaves in a moist chamber. While describing a new species of *Phalangispora*, *P. nawawi* Kuthub. (holotype, IMI 312357, Lepar Forest Reserve, Pahang, Malaysia July 1986), Kuthubutheen (1987) confirmed the occurrence of the sporodochia of the type species in natural habitats in Malaysia. *P. nawawi* was erected based on the difference in size of the individual conidia, conidial chain and number of individual cell in the main axis and lateral branches. In *P. nawawi* the conidial chains were smaller and number of cells was fewer than *P. constricta*. The third species of *Phalangispora, P. bharathensis* was added from the Western Ghats of India as a terrestrial litter hyphomycete on *Holigarna arnotiana* (Anacardiaceae) from Cotigao Wildlife Sanctuary, Goa, India (IMI 387091). It was the first published species added to the genus *Phalangispora* outside Malaysia and first record of this genus from India (Prasad & Bhat 2002). The major differences in conidial characteristics of these species are given in Table 1.

In the present study the length of setae in the new strain of *P. bharathensis* was found to be shorter though the arrangement of setae was unique, surrounding the sporodochia. Also, the number of setae is much higher at 9–28 per sporodochia. All the species so far recorded under *Phalangiospora* have been described with sporodochia with basal setae; especially in *P. bharathensis* 8–16 setae arise from the margin of the sporodochial base. The illustrations provided in *P. nawawi* and *P. bharathensis* stated the same. Similar characteristics were validly considered even for differentiation of stauroconidia forming genus like *Fumagopsis* Speg. and *Tridentaria* Preuss in the past. Spegazzini (1910) established the genus *Fumagopsis* with a monotypic species under this genus *F. triglifioides* Speg. Later Kendrick & Carmichael (1973) incorrectly placed this genus as a synonym of *Tridentaria*. But van der Aa & van Oorschot (1985) and van der Aa & von Arx (1986) maintained *Fumagopsis* as an acceptable genus with its unique charaterestics, setose sporodochial conidiomata surrounding the sporodochia. *Tridentaria* produces mononematous conidiophores and colonies

Table 1. Difference of conidia and conidial chain in genus *Phalangispora*.

Species	Conidia size (µm)	Conidial chain size (µm)
P. constricta	11–20×2.5–4	120–140×3–4
P. nawawi	10–12×2 (6 to 8 cells)	65–90×2
P. bharathensis	8–10×2.5–4 (8 to 11 cells mostly 9)	75–85×2.5–4
Present study	8–12.5×2–4.5 (8 to 9 cells mostly 8)	74–87.5×2–4.5

are without setae. However, in the present study considering the conidial shape, size (extend beyond type strain) and conidial chain length in AMH 9580; the new collection is placed under the species *P. bharathensis.*

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