

Annulohypoxyton spougei



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***Annulohypoxyton spougei* Suwannasai, M.P. Martín, Phosri & Whalley, sp. nov.**

Etymology. Named after the American bioinformatician John L. Spouge who contributed to the discovery of this species, and for his efforts to implement tools for DNA barcoding analyses within the genus *Annulohypoxyton*.

Classification — *Hypoxyloaceae*, *Xylariales*, *Sordariomycetes*.

Stromata glomerate to hemispherical, effused-pulvinate, with perithecial mounds 1/4 to 2/3 exposed and not covered by the outermost stromatal layer, 0.3–6 cm long × 0.3–3 cm broad and 1–1.6 mm thick; surface dark brown vinaceous, becoming black with reddish brown hues, finally black and shiny; black granules immediately below the surface, KOH pigments green olivaceous. **Perithecia** spherical, 0.5–0.7 mm diam. **Ostioles** conical papillate, surrounded by a flattened *bovei*-type disc, 0.2–0.5 mm diam. **Asci** 62–114 × 4–4.5 µm, the spore bearing parts 64–73 µm long with stipes 20–32(–46) µm long, with apical ring bluing in Melzer's iodine reagent, discoid 0.7 µm high × 1.5–2 µm broad. **Ascospores** pale brown, unicellular, ellipsoid-inequilateral with narrowly rounded ends, 6–10.5 × 3–4.5(–5.5) µm with straight germ slit along the full length of the spore; perispore dehiscent in 10 % KOH, epispore smooth.

Culture characteristics — Colonies on potato dextrose agar (PDA) covering Petri dish in 2 wk, at first white, becoming hazel to dull green, azonate, with diffuse margins, with scattered black patches; reverse dull green to dark brown. **Conidiogenous structure** nodulisporium-like, brown. **Conidia** hyaline, smooth, ellipsoid, 3.5–4.5 × 2–3 µm.

Typus. THAILAND, Phitsanulok, Khao KraYang Forest Planation, on corticated wood, Sept. 2006. *C. Phosri & N. Suwannasai* H099 (holotype SWUF-H099; ITS, α -actin and β -tubulin sequences GenBank FN252419, FR875158 and KP134519, MycoBank MB811164).

Additional materials examined. Herbarium number is indicated, as well as the ITS, α -actin, β -tubulin and *EF1- α* GenBank sequences between brackets, absent sequences are indicated with '–'. ***Annulohypoxyton spougei*:** THAILAND, Phitsanulok Province, *Dipterocarpaceae* forest, Sept. 2006, *C. Phosri & N. Suwannasai* SWUF-H087 (FN252418, KP134506, FR875164, –); SWUF-H181 (FN252420, KP134507, KP134520, –); SWUF-H203 (FN252421, FR875159, FR875165, –); SWUF-H215 (FN252422, KP134508, KP134521, –); SWUF-H254 (FN252423, FR875160, FR875166, –); Nakhon Ratchasima Province, *Dipterocarpaceae* forest, July 2003, *N. Suwannasai* SUT081 (DQ322105, –, –, –); Trad Ratchasima Province, *Dipterocarpaceae* forest, Aug. 2003, *C. Phosri & N. Suwannasai* SUT236 (DQ322106, –, –, –); SUT242 (DQ322107, –, –, –); SUT244 (DQ322108, –, –, –); SUT251 (DQ322109, –, –, –); Kanchanaburi Province, *Dipterocarpaceae* forest, Aug. 2003, *N. Suwannasai* SUT285 (DQ322110, –, –, –); Chaiyaphum Province, *Dipterocarpaceae* forest, June 2009, *C. Phosri & N. Suwannasai* PK09007 (KP134526, KP134509, KP134522, KP134499); PK09026 (KP134527, KP134510, –, KP134500); PK09027 (KP134528, KP134511, –, KP134500);

Colour illustrations. Thailand, Chaiyaphum Province, Phu Khiao Wildlife Sanctuary, where the specimens were collected. From top to bottom: stromata with ostiolar discs (SWUF-H099); ascospores under SEM (SWUF-H099); fungal culture on PDA (SWUF-H099); nodulisporium-like anamorph (SWUF-H099); ascospores with apical apparatus (SWUF-H099). Scale bars = 0.5 mm (stromata), 5 µm (ascospores SEM), 1 cm (fungal culture), 15 µm (asexual morph), 5 µm (ascospores)

PK09029 (KP134529, KP134512, –, –). ***Annulohypoxyton nitens*:** THAILAND, Chiang Rai Province, *Dipterocarpaceae* forest, Sept. 2006, *C. Phosri & N. Suwannasai* SWUF-H154 (FM209453, FR875161, KP134513, –); SWUF-H157 (FM209455, FR875162, KP134514, –); Phitsanulok Province, *Dipterocarpaceae* forest, Sept. 2006, *C. Phosri & N. Suwannasai* SWUF-H189 (FM209459, KP134502, FR875167, –); SWUF-H197 (FM209461, FR875163, KP134515, –); Chaiyaphum Province, *Dipterocarpaceae* forest, June 2009, *C. Phosri & N. Suwannasai* PK121044 (KP134523, KP134503, KP134516, KP134496); PK121063 (KP134524, KP134504, KP134517, KP134498); PK121086 (KP134525, KP134505, KP134518, KP134497).

Notes — During extensive studies of the *Hypoxyloaceae* in Thailand over a period of almost 20 yr, problems were encountered in the identification of several taxa, especially *A. nitens*. A previous study on species of *Hypoxyton* and *Annulohypoxyton* using morphology and ITS nrDNA sequences (Suwannasai et al. 2013) indicated that this taxon was not monophyletic but could be separated into *A. nitens* and another species. Twenty-eight fungal specimens of *A. nitens* and a cryptic species collected from Thailand, previously named '*A. nitens*' in our study (Suwannasai et al. 2013), were carefully re-analysed based on morphological and asexual morph characters. The comparison of morphological characters between *A. nitens* and a cryptic species showed unclear distinction of these species. The cryptic species, here named as *A. spougei* possesses spherical perithecia (0.5–0.7 mm diam), which are slightly narrower than those of *A. nitens* described by Ju & Rogers (1996) ((0.4–)0.5–1(–1.2) mm). The ostiolar discs of both species groups are *bovei*-type and have the same dimensions of 0.2–0.5 mm. Ascospore sizes of *A. nitens* and the cryptic species are 7.5–9 × 2.8–4.2 µm and 6–10.5 × 3–4.5(–5.5) µm, respectively. These are similar to the species description for *A. nitens* (as *H. nitens*) (6.5–10(–11) × 3–4.5 µm) from Ju & Rogers (1996). The cultural and asexual morph characters were observed from both PDA and oatmeal agar. Colonies of *A. spougei* are white at first becoming hazel and dull green with scattered black patches. The asexual morph is nodulisporium-like and conidial size (3.5–4.5 × 2–3 µm) is similar to *A. nitens* (4–5 × 2.5–3 µm). With those similar features, it is very difficult to separate the *A. spougei* from *A. nitens* by using only morphological and asexual morph characters. However, although morphological data for all of the collections initially identified as *A. nitens* failed to provide clear separation of the two entities, there are clear supporting DNA data for their separation. In the present study based on α -actin, β -tubulin and elongation factor 1- α sequences, we confirm the separation of two taxa mentioned in Suwannasai et al. (2013).

Supplementary material

FP1060 UPGMA reconstruction based on K2P distances of α -actin, β -tubulin and *EF1- α* sequences of *Annulohypoxyton nitens* and *A. spougei* specimens using PAUP* v. 4.0b10 (Swofford 2003).

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