The genus *Annulohypoxylon* (*Xylariaceae*) in Guadeloupe and Martinique (French West Indies)

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Abstract: This survey deals with the *Annulohypoxylon* spp. collected in the French West Indies in the course of an ongoing inventorial work on the mycobiota of these islands initiated in 2003. Based on the evaluation and comparison of their morphological characters, eleven species are described, illustrated and discussed, including three new species, *A. caravellense*, *A. ramulorum* and *A. splendens* and a taxonomically unsettled species provisionally referred to *A. moriforme*. Other recorded taxa include *A. atroroseum*, *A. leptascum*, *A. moriforme* var. *microdiscum*, *A. nitens*, *A. purpureopigmentum*, *A. purpureonitens* and *A. stygium*, of which only the latter was already known from Guadeloupe. A dichotomous identification key is provided.

Keywords: Ascomycota, *Hypoxyloideae*, pyrenomycetes, saproxylic fungi, taxonomy, tropical mycology, *Xylariales*.

Résumé : cette étude porte sur les espèces d'*Annulohypoxylon* récoltées lors de missions d'inventaire de la fonge des Antilles françaises commencées en 2003. En se fondant sur l'évaluation et la comparaison de leurs caractères morphologiques, onze espèces sont décrites, illustrées et commentées, comprenant trois espèces nouvelles, *A. caravellense*, *A. ramulorum* et *A. splendens*, ainsi qu'une espèce de position taxinomique incertaine provisoirement rattachée à *A. moriforme*. Les autres taxons recensés comprennent *A. atroroseum*, *A. leptascum*, *A. moriforme* var. *microdiscum*, *A. nitens*, *A. purpureopigmentum*, *A. purpureonitens* et *A. stygium*, dont ce dernier seulement était déjà connu de Guadeloupe. Une clé dichotomique de ces espèces est proposée. **Mots-clés :** Ascomycota, champignons saproxyliques, *Hypoxyloideae*, mycologie tropicale, pyrénomycètes, taxinomie, *Xylariales*.

Introduction

This survey of *Annulohypoxylon* Y.-M. Ju, J.D. Rogers & H.-M. Hsieh in the French Caribbean islands of Guadeloupe and Martinique comes after similar studies carried out on *Hypoxylon* Bull. in the same region (Fournier *et al.*, 2015) and on *Annulohypoxylon* in French Guiana (Fournier & Lechat, 2016). The reader is referred to the former publication for details on the framework within which this work was carried out and for the most salient ecological features characterizing these two islands. In the latter publication an updated historical and taxonomic review of the genus was provided, which is not repeated here.

Annulohypoxylon belongs to the Xylariaceae Tul. & C. Tul. based on its stromatic ascomata, unitunicate asci with an amyloid apical apparatus, brown, one-celled ascospores with a germ slit and hyphomycetous asexual morph with holoblastic conidiogenesis. It is accommodated in the subfamily Hypoxyloideae because of its nodulisporium-like asexual morphs and is most closely related to Daldinia Ces. & De Not. and Hypoxylon because of its secondary stromatal metabolites yielding pigments in 10% KOH and ascospores with a perispore frequently dehiscent in 10% KOH. It is primarily distinguished from Daldinia and Hypoxylon by more carbonaceous stromata and conic-papillate ostioles encircled with a disc resulting from the dehiscence of stromatal tissue (Læssøe, 1994; Ju & Rogers, 1996). The morphology-based segregation of Annulohypoxylon from Hypoxylon was confirmed by phylogenetic analyses of α-actin and β-tubulin sequences (HSIEH et al., 2005).

The limited available information on the presence of supposed *Annulohypoxylon* spp. in the French West Indies dates back to the check-list published by Duss (1903). At that time the name *Hypoxylon* was applied in a wide sense to many xylariaceous fungi that are currently accommodated within more narrowly delimited genera segregated from *Hypoxylon* (Ju & Rogers, 1996). However, the presence of ostiolar discs was already noticed and used as a differential character by earlier mycologists, reflected by suggestive epithets like *annulatum*, *circumscribum*, *marginatum*, *platystomum*, *truncatum* which leave little doubt these fungi pertain to *Annulohypoxylon*.

Duss (1903) recorded *Hypoxylon annulatum* (Schwein.: Fr.) Mont., *H. marginatum* (Schwein.) Berk. and *H. stygium* (Lév.) Sacc. from both Guadeloupe and Martinique. In their taxonomic and nomenclatural reinterpretation of the two former taxa, Ju & ROGERS (1996) restricted

H. annulatum to a taxon with hemispherical stromata with barely exposed perithecial contours, brown to dark brown ascospores, host affinity to Quercus and temperate to subtropical distribution; H. marginatum was considered as a synonym. Subsequently, H. annulatum was moved to Annulohypoxylon by HSIEH et al. (2005). Although the epithet annulatum in its former wide sense could have been applied to most of the material we collected, we did not encounter material matching A. annulatum sensu Ju & Rogers during this survey. Therefore H. stygium, widespread in both islands and easily set apart by its very small ostiolar discs and small ascospores, is the only species recorded by Duss that can be reliably retained in Annulohypoxylon, as A. stygium (Lév.) Y.-M. Ju, J.D. Rogers & H.-M. Hsieh.

The present work summarizes the observations made on the material of *Annulohypoxylon* collected during an ongoing inventorial survey initiated in 2003 (COURTECUISSE, 2006).

As noted in FOURNIER & LECHAT (2016), identifying *Annulohypoxylon* is most often a challenging task owing to the paucity of reliable morphological differential characters and the confusing intraspecific variations of stromatal morphology.

Ostiolar disc morphology appears a good and reliable differential character. The diameter of ostiolar discs is fairly consistent within a given species, provided it is recorded on well- developed, mature stromata. The type of dehiscence of the disc, either *bovei*-type or *truncatum*-type as defined by Ju & Rogers (1996), is likewise consistent for a given species but must be inferred from the direct observation of the dehiscence on immature stromata more than from the appearance of the disc itself resulting from the dehiscence.

Stromatal metabolites aggregated in waxy granules, forming a thick crust or mixed with carbonaceous tissue around the perithecia yield coloured reactions with 10% KOH. In most species of *Annulohypoxylon* the KOH-extractable pigments are olivaceous to green and therefore of limited differential value. This colour reaction proved in some cases more informative when coupled with 1) the observation under the microscope of stromatal granules in water in order to evaluate their colour prior to the KOH reaction and 2) with recording how the colour of KOH-extractable pigments evolve after 20–30 min incubation.

The colour of the outermost stromatal coating is often informative at species level but this character is usually absent from mature and overmature stromata. In some species the outermost coating flakes off early to reveal a shiny black surface, which is a taxonomi-

cally relevant character, but in such species the coating may persist at maturity, making this character sometimes difficult to interpret.

Stromatal thickness and perithecial shape and dimensions may be informative but most often are not diagnostic.

Ascospores may offer good differential characters when they are pale or dark brown, when they are strongly inequilateral, when they average less than 6 μm long or more than 10 μm long, when they have a short germ slit or have a perispore not dehiscent in 10% KOH. Unfortunately, many species, especially those related to A. moriforme (Henn.) Y.-M. Ju, J.D. Rogers & H.-M. Hsieh and A. nitens (Ces.) Y.-M. Ju, J.D. Rogers & H.-M. Hsieh feature medium brown, slightly inequilateral ascospores in the size range of 7–10 \times 3–5 μm , with a long germ slit and a perispore dehiscent in KOH, which does not help separating them.

When the asexual morph is present on surface or at margins of young or overmature stromata, it usually features short to long dark brown conidiophores apically ending in a ramified conidiogenous structure varying from nodulisporium-like to periconiella-like as defined by Ju & ROGERS (1996). As the influence of environmental factors on the morphology of the asexual morph on natural substrate is likely to be non-negligible, this character is difficult to be regarded as diagnostic when compared with the asexual morphs obtained by *in vitro* cultures.

Based on the combination of the aforementioned morphological characters, the material collected in Guadeloupe and Martinique since 2003 revealed the presence of eleven different species of *Annulohypoxylon*, including three new species that could not be equated to known species and an unsettled species possibly related to *A. moriforme. Annulohypoxylon atroroseum* (J.D. Rogers) Y.-M. Ju, J.D. Rogers & H.-M. Hsieh and *A. stygium* were diagnosed because of

their small ostiolar discs and small ascospores with a germ slit and perisporial thickening located on the flattened side. *Annulohypoxylon leptascum* (Speg.) Y.-M. Ju, J.D. Rogers & H.-M. Hsieh was readily recognized based on its pale brown fusiform ascospores with a short germ slit originating from one end. *Annulohypoxylon moriforme* var. *microdiscum* (Y.-M. Ju & J.D. Rogers) Y.-M. Ju, J.D. Rogers & H.-M. Hsieh, *A. purpureopigmentum* Jad. Pereira, J.D. Rogers & J.L. Bezerra and *A. purpureonitens* (Y.-M. Ju & J.D. Rogers) Y.-M. Ju, J.D. Rogers & H.-M. Hsieh were identified through their different KOH-extractable pigments respectively ochreous, dense livid purple and pale vinaceous purple.

Three different species featuring shiny black stromata were encountered, of which three collections matching well *A. nitens* are regarded as typical for this taxon. The new species *A. caravellense* and *A. splendens* are segregated from typical *A. nitens* by smaller ostiolar discs and denser green KOH-extractable pigments for the former and by *truncatum*-type ostiolar discs and larger ascospores for the latter. Several collections of an *Annulohypoxylon* forming small glomerate stromata with a woolly reddish brown outermost coating, small *truncatum*-type ostiolar discs and olivaceous KOH-extractable pigments represent a new species. Because of its consistent occurrence on small branches and twigs this species is given the name *A. ramulorum*.

These species are described and illustrated in the taxonomic part and their affinities and the differential characters supporting their taxonomic delimitation are discussed. A dichotomous identification key to the species known from the French West Indies is proposed.

Aside from the species cited by Ju & Rogers (1996) and the species described by Pereira *et al.* (2010), the only report of neotropical *Annulohypoxylon* spp. available in literature is that of SAN MARTÍN *et al.*

Dichotomous key to <i>Annulohypoxylon</i> spp. known from French West Indies				
1	KOH-extractable pigments olivaceous or green			
	2 Ostiolar discs 0.15–0.2 mm diam; ascospores 5–6 × 2.2–2.5 µm with long inconspicuous germ slit on the ventral side			
3	Stromatal surface pink to vinaceous grey, eventually blackish			
	Ascospores fusiform, pale brown, with short germ slit originating from one end; perispore indehiscent in 10% KOH <i>A. leptascum</i> Ascospores ellipsoid-inequilateral, brown, with central germ slit spore-length or nearly so; perispore dehiscent in 10% KOH 5			
5	Stromatal surface becoming shiny black at maturity by flaking off of the outermost layer			
6	Ascospores $8.8-10.1\times3.9-4.5~\mu m$; ostiolar discs <i>truncatum</i> -type			
7	Ostiolar discs 0.4–0.5 mm diam; KOH-extractable pigments greenish-olivaceous			
	Stromata glomerate, few-peritheciate, reddish brown, becoming dull black; ostiolar discs 0.2–0.3 mm diam; ascospores 6.6–8.4 × 3.1–3.8 µm			
9	,			
	O Stromata with persistent, umber to blackish brown, matt outermost coating; ostiolar discs 0.2–0.25 mm diam; KOH-extractable pigments dense livid purple			

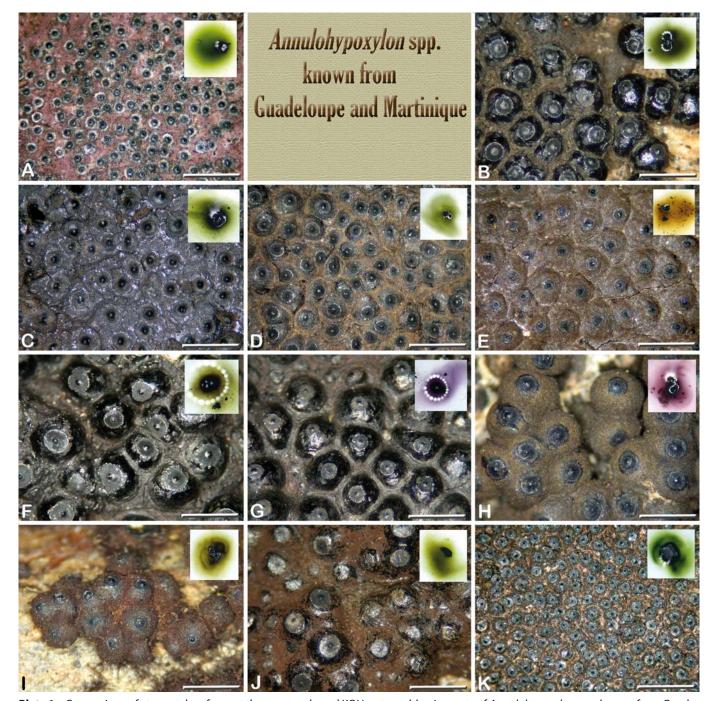


Plate 1 – Comparison of stromatal surfaces at the same scale and KOH-extractable pigments of *Annulohypoxylon* spp. known from Guadeloupe and Martinique. A: *A. atroroseum* MJF 15191; B: *A. caravellense* MJF 10235; C: *A. leptascum* CLL 5340; D: *A. cf. moriforme* MJF 07127; E: *A. moriforme* var. *microdiscum* MJF 07228; F: *A. nitens* MJF 07040; G: *A. purpureonitens* MJF 10124; H: *A. purpureopigmentum* MJF 10309; I: *A. ramulorum* MJF 10312; J: *A. splendens* CLL 2304; K: *A. stygium* MJF 07115.

(1999) from Mexico, in which ten species are listed (as *Hypoxylon*), of which only *A. nitens*, *A. purpureonitens* and *A. stygium* were also recorded in FWI during our survey.

In terms of frequency, two species appear prevailing in Guade-loupe and Martinique: A. stygium, with a strong preference for hygrophilic rainforests and A. caravellense exclusively present in xerophilic to mesophilic forests. Other species are known from only one or a few collections and can therefore be regarded as rarer. Among them only A. atroroseum, A. purpureonitens and A. ramulorum are known from more than three collections.

Materials and methods

Materials and methods, including colour charts, follow Fournier & Lechat (2015).

Taxonomy

Annulohypoxylon atroroseum (J.D. Rogers) Y.-M. Ju, J.D. Rogers & H.-M. Hsieh, Mycologia, 97 (4): 857 (2005). Plates 2–3

Stromata effused-pulvinate, 8–38 mm long \times 4–25 mm wide \times 0.5–0.75 mm thick, with inconspicuous to faintly exposed perithecial contours, rarely strongly exposed; surface with a livid purple (61, oac449), pale vinaceous grey (115, oac569) or vinaceous grey (116, oac527) pruinose outermost coating, blackish with age; texture weakly carbonaceous, with subsurface composed of olivaceous granules yielding dark herbage green (69, oac54) KOH-extractable pigments within 1 min incubation, turning olivaceous (48, oac869) upon prolonged incubation; subperithecial tissue inconspicuous. **Perithecia** obovoid to tubular,

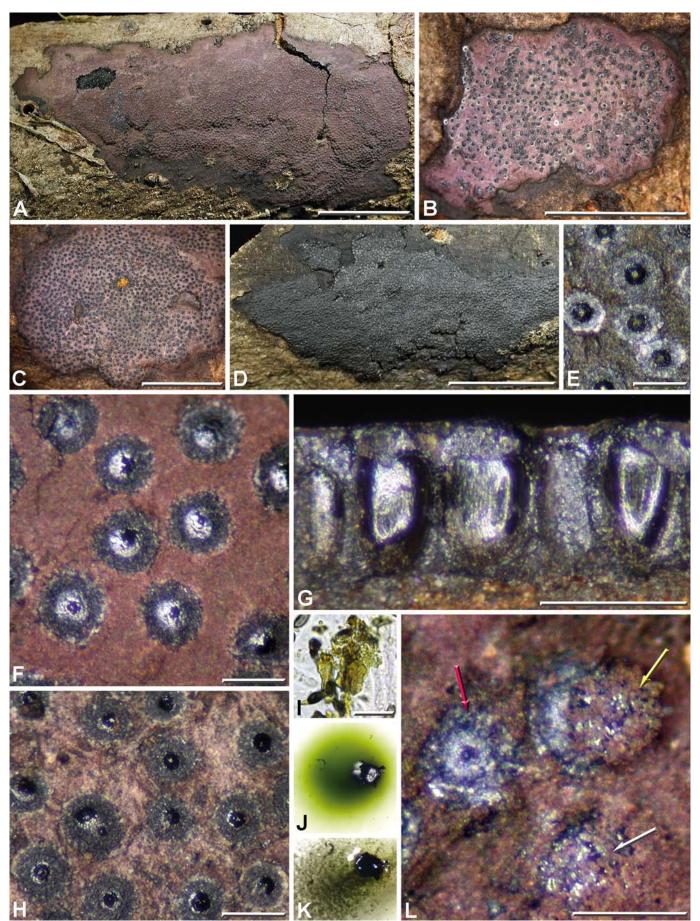


Plate 2 – Annulohypoxylon atroroseum. MJF 15191. A-C: Young stromata with pinkish surface; D: Mature stroma with dull black surface; E, F, H: Stromatal surface in close-up showing the ostiolar discs and the variously coloured outermost coating (F, H), absent in E; G: Stroma in vertical section (broken) showing the perithecia encased by carbonaceous tissue; I: Stromatal waxy granules from subsurface mixed with carbonaceous elements, in water; J, K: KOH-extractable pigments after 1 min and 30 min incubation respectively; L: Perithecial mound prior to the dehiscence of the ostiolar disc (white arrow), dehiscing ostiolar disc (yellow arrow) and freshly exposed ostiolar disc (red arrow). Scale bars: A, D = 10 mm; B, C = 5 mm; E, F, H, L = 0.2 mm; G = 0.5 mm; I: 10 μ m.

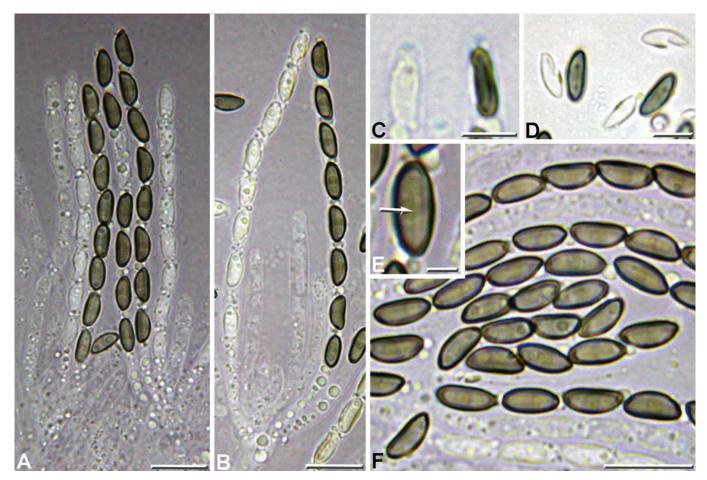


Plate 3 – **Annulohypoxylon atroroseum.** MJF 15191. A, B: Mature and immature asci in black Pelikan ink, showing the sort sipes; C: Ascal apical apparati in Melzer's reagent; D: Ascospores in 10% KOH showing the dehiscent perispore with a thickening on the ventral side; E: Ascospore in ventral view showing the faint germ slit (arrow), in black Pelikan ink; F: Ascospores in black Pelikan ink. Scale bars: A, B, F = 10 μ m; C, D = 5 μ m; E = 2 μ m.

 $0.38-0.45 \times 0.2-0.3$ mm. **Ostioles** conic-papillate, encircled with a shiny black, slightly convex *bovei*-type disc 0.15–0.2 mm diam with most often irregular rims.

Asci cylindrical, with eight uniseriate ascospores, 58–70 µm total length, the spore-bearing parts 47–56 \times 3–3.5 µm, the stipes 9–16 µm long, with a minute discoid apical apparatus faintly bluing or not bluing in Melzer's reagent. **Paraphyses** sparse, simple to furcate, 4–5.5 µm wide at base, tapering to 2–3 µm above asci. **Ascospores** (5.5–)5.8–6.4(–6.6) \times (2.2–)2.4–2.7(–2.8) µm, Q = (2.1–)2.2–2.6(–2.7); N = 60 (Me = 6.1 \times 2.5 µm; Qe = 2.4), ellipsoid-inequilateral with narrowly to broadly rounded ends, pale brown, with an inconspicuous straight germ slit almost spore-length on the ventral side; perispore dehiscent in 10% KOH, smooth, with a thickening on the ventral side at ca. ½ spore length; epispore smooth.

Asexual morph on the natural substrate not seen. Asexual morph in culture on OA nodulisporium-like, based on material from Taiwan (Ju & ROGERS, 1996).

Specimens examined: FRENCH WEST INDIES: MARTINIQUE: Case-Pilote, Crête Jean-Louis, hygrophilic rainforest, dead corticated branch of *Ficus* sp., 3 Sept. 2003, *leg*. C. Lechat, CLL 0801 (LIP); Morne-Rouge, La Propreté, forest trail, hygrophilic rainforest, dead corticated branch of Mahogany (*Swietenia macrophylla* King, *Meliaceae*), 24 Aug. 2007, *leg*. J. Fournier, MJF 07057 (LIP); *ibid.*, same date and host, *leg*. C. Lechat, MJF 07087 (LIP); Morne-Rouge, domaine d'Émeraude, hygrophilic rainforest, on a dead corticated branchlet, 9 Aug. 2013, *leg* J. Fournier, MJF 13095 (LIP); Morne-Rouge, La Propreté, forest trail, hygrophilic rainforest, dead corticated branch of Mahogany

(*Swietenia macrophylla* King, *Meliaceae*), 23 Jun. 2015, *leg.* J. Fournier, MJF 15191 (LIP).

Distribution: Pantropical.

Discussion: Annulohypoxylon atroroseum, known from Gabon and Taiwan, was segregated from A. stygium based on a vinaceous grey stromatal surface and a nodulisporium-like asexual morph in culture on OA (Rogers, 1981; Ju & Rogers, 1996). Both are very similar and set apart from other Annulohypoxylon spp. by the combination of very small ostiolar discs and small perithecia, green KOH-extractable pigments and small ascospores with an inconspicuous germ slit on the ventral side. Their ostiolar discs are said to be of the truncatum-type (Ju & Rogers, 1996) but our observations of dehiscing discs on young stromata showed they are of the bovei-type, both in A. atroroseum (the present study) and in A. stygium (FOURNIER & LECHAT, 2016).

In absence of data on the asexual morph in culture, the distinction between *A. atroroseum* and *A. stygium* relies only on the stromatal surface colour. The stromata illustrated above clearly display a typical pinkish to vinaceous surface, while typical stromata of *A. stygium* are brown to reddish brown. Unfortunately many collections that display a vague vinaceous brown surface are difficult to unequivocally assign to either species. Fully mature and overmature stromata of both species become black by losing their outermost pigmented pruina and therefore cannot be distinguished.

Moreover, based on our observations, both species share a similar preference for hygrophilic rainforest.

Annulohypoxylon caravellense J. Fourn. & Lechat, *sp. nov.* – MycoBank MB 816975. Plates 4–5. Table 1.

Diagnosis: Differs mainly from the most resembling species *Annulohypoxylon* nitens by darker green KOH-extractable pigments, smaller perithecia, smaller ostiolar discs and slightly larger ascospores averaging $8 \times 3.5 \, \mu m$.

Holotype: French West Indies: Martinique: Trinité (Caravelle peninsula), Pointe-Rouge, xerophilic coastal forest, dead corticated branch, 29 Aug. 2010, *leg.* J. Fournier, MJF 10235 (LIP).

Etymology: For the Caravelle peninsula, where this fungus is widespread and where most collections come from.

Stromata effused to slightly pulvinate in places, with conspicuously $\frac{1}{2}$ - $\frac{2}{3}$ exposed perithecial contours, 1.5–30 mm long × 1.5– 15 mm wide \times 0.6–0.85 mm thick, separate to irregularly coalescent into larger compound stromata, perithecioid in places; outermost coating thick, dark olivaceous grey to grey brown, eventually blackish grey, long-persistent, squamulose, gradually worn off to more or less reveal the shiny black and smooth subsurface of exposed perithecial contours at maturity; texture carbonaceous, with pale olivaceous yellow waxy granules mixed with carbonaceous tissue forming a thick crust encasing each perithecium, releasing dark greenish olivaceous (90, oac880) KOH-extractable pigments within 1 min, becoming dark grey olivaceous (107, oac866) upon prolonged incubation; subperithecial tissue inconspicuous to 0.2 mm thick, olivaceous brown to blackish, woody. Perithecia subspherical, 0.5–0.55 mm diam. Ostioles finely conic-papillate, encircled with a flattened bovei-type disc 0.3-0.34 mm diam with sharp to slightly notched edges.

Asci cylindrical, with eight uniseriate ascospores, 95–132 μm total length, the spore-bearing parts $58-70 \times 4.2-4.5$ μm, the stipes 34-65 μm long originating from swollen contorted ascogenous hyphae, with a discoid apical apparatus $0.6-0.8 \times 1.3-1.6$ μm, bluing in Melzer's reagent. **Hamathecium** of thin-walled paraphyses 5-7.5 μm wide at base, slightly constricted at septa, tapering above asci. **Ascospores** $(7.4-)7.6-8.6(-8.8) \times (2.9-)3.2-3.7(-4.2)$ μm, Q=(2-)2.2-2.5(-3); N=60 (Me $=8.1 \times 3.5$ μm; Q=2.3), ellipsoid-inequilateral with narrowly to broadly rounded ends, brown, with a conspicuous straight germ slit almost spore-length; perispore dehiscent in 10% KOH, smooth, with a thickening on the dorsal side at ca. $\frac{1}{3}$ spore length; epispore smooth.

Asexual morph on the natural substrate: Conidiophores scattered or arising in loose tufts, originating from olivaceous yellow tissue present at margins of young stromata or from olivaceous sterile plaques between stromata; conidiophores upright, with a stout brown to dark brown septate axis 1–2.2 mm high \times 6–8.5 μ m wide, roughened, apically pale olivaceous and branching; conidiogenous structure nodulisporium-like to periconiella-like, with conidiogenous cells $10-15\times3.5-4.5~\mu$ m, finely roughened, pale olivaceous; conidia subhyaline, smooth, ellipsoid, 5–5.5 \times 2.5–3 μ m.

Other specimens examined (paratypes): French West Indies: Gua-DELOUPE: Les Saintes, Terre-de-Bas, Étangs, meso-xerophilic coastal forest, dead corticated and decorticated branch, 15 Jan. 1994, leg. J. Vivant, communicated by F. Candoussau, JF 01190 (LIP); Sainte-Anne, Douville, Liard, mesophilic to xerophilic forest, on corticated wood, 13 Sept. 2003, leg. C. Lechat, CLL 1075 (LIP). MARTINIQUE: Trinité (Caravelle peninsula), Pointe-Rouge, xerophilic coastal forest, dead corticated branch, 27 Aug. 2005, leg. C. Lechat, CLL 5233 (LIP); ibid., dead corticated branch, 21 Aug. 2007, leg. J. Fournier, MJF 07006 (LIP); ibid., dead corticated branch, 21 Aug. 2007, leg. J. Fournier, MJF 07007 (LIP); ibid., dead corticated branch, 21 Aug. 2007, leg. J. Fournier, MJF 07008 (LIP); ibid., dead corticated branch, 31 Aug. 2007, leg. J. Fournier, MJF 07230 (LIP); ibid., dead corticated branch, 31 Aug. 2007, leg. J. Fournier, MJF 07236 (LIP); ibid., dead corticated branch, 31 Aug. 2007, leg. J. Fournier, MJF 07239 (LIP); ibid., dead corticated branch, 22 Aug. 2010, leg. J. Fournier, MJF 10039 (LIP); Le Robert, Pointe-Bateau, xerophilic coastal forest, dead corticated branch, 26 Aug. 2010, *leg.* J. Fournier, MJF 10134 (LIP); *ibid.*, dead corticated branch, 26 Aug. 2010, *leg.* J. Fournier, MJF 10135 (LIP); Trinité (Caravelle peninsula), Pointe-Rouge, xerophilic coastal forest, dead decorticated branch, 29 Aug. 2010, *leg.* J. Fournier, MJF 10211 (LIP); *ibid.*, dead decorticated branch, 29 Aug. 2010, *leg.* J. Fournier, MJF 10226 (LIP); *ibid.*, dead decorticated branch, 29 Aug. 2010, *leg.* J. Fournier, MJF 10231 (LIP); *ibid.*, dead corticated branch, 29 Aug. 2010, *leg.* J. Fournier, MJF 10238 (LIP); Trinité (Caravelle peninsula), Balata, xerophilic coastal forest, dead corticated branch, 11 Aug. 2013, *leg.* J. Fournier, MJF 13128 (LIP); Case-Pilote, Fond Boucher, mesophilic to xerophilic forest, dead corticated branch, 17 Aug. 2013, *leg.* J. Fournier, MJF 13287 (LIP); *ibid.*, dead corticated branch, 17 Aug. 2013, *leg.* J. Fournier, MJF 13304 (LIP).

Known distribution: Guadeloupe, Martinique.

Discussion: This Annulohypoxylon, repeatedly collected in relatively dry coastal forests of eastern Martinique was long given the provisional name A. cf. moriforme owing to the lack of distinctive characters and its resemblances with A. moriforme and A. nitens, both ill-defined taxa. A careful observation of stromata at all stages of development showed that the thick outermost coating is gradually worn off at maturity, revealing a shiny black stromatal surface typical of A. nitens and A. purpureonitens. The latter was easily distinguished because of its purplish KOH-extractable pigments but the differences with A. nitens as currently conceived in a broad sense by Ju & Rogers (1996) were more difficult to appraise. The most salient difference of this Annulohypoxylon with A. nitens is that its stromata do not become entirely shiny black but instead keep more or less conspicuous remnants of the grey brown outermost coating. In this paper we tentatively redefine A. nitens based on typical collections from French West Indies and from tropical China (Hainan), in order to facilitate its comparison with related but deviating collections that otherwise would have fallen in the current wide concept of this species. Besides the long-persistent outermost coating, this Annulohypoxylon species consistently differs from our restricted concept of A. nitens by thinly effused stromata 0.6-0.85 mm vs. 1-1.8 mm thick, smaller ostiolar discs 0.3-0.34 mm vs. 0.4-0.5 mm diam, smaller perithecia 0.5–0.55 mm vs. 0.6–0.7 mm diam and darker green KOH-extractable pigments. Ascospore dimensions largely overlap but they average slightly larger in the new Annulohypoxylon: $8\times3.5~\mu m$ vs. 7.6 \times 3.3 μm . Based on these differences with A. nitens we feel justified in proposing the new taxon A. caravellense to accommodate the numerous collections made in Guadeloupe and especially in Martinique.

We also noticed in *A. caravellense* a stronger reaction of ascal apical apparati to Melzer's reagent and shorter ascal stipes than in *A. nitens* but it is assumed that these differences are less significant than the previous ones.

We also considered A. squamulosum (Y.-M. Ju, J.D. Rogers & H.-M. Hsieh) Y.-M. Ju, J.D. Rogers & H.-M. Hsieh, known from Taiwan, which like A. caravellense features an outermost coating flaking off into persistent scales and revealing a shiny black stromatal surface and ascospore size in the same range 7–8.5 \times 3–4 μm (Ju et al., 2004). In this study A. squamulosum was segregated from A. nitens based on its much smaller ostiolar discs 0.1-0.2 mm diam and its smaller perithecia 0.4-0.5 mm diam. The significantly larger ostiolar discs of A. caravellense clearly set it apart from A. squamulosum to which, however, it might be closely related. Other species segregated from the A. nitens complex are A. splendens J. Fourn. & Lechat (this paper) and A. subnitens J. Fourn. & Lechat (FOURNIER & LECHAT, 2016). The former is distinguished from A. caravellense by a reddish brown outermost coating, truncatum-type ostiolar discs and significantly larger ascospores averaging $9.5 \times 4.2 \mu m$. Annulohypoxylon subnitens, known from only one collection from primary rainforest in French Guiana, is set apart from other members of the A. nitens complex, including A. caravellense, by inconspicuous pale olivaceous grey



Plate 4 – Annulohypoxylon caravellense. A-H, J: MJF 10135, holotype; I: MJF 10238, paratype; K, L: MJF 13304, paratype. A: Widely effused stroma on bark, shiny black in places; B, C: Close-up on stromatal surface showing the grey brown outermost tissue and the ostiolar discs; D: Stroma in vertical section showing the shiny black crust and the thin subperithecial tissue; E: Close-up on perithecioid stromata with grey brown scales on a shiny black subsurface; F: Olivaceous yellow stromatal granules mixed with carbonaceous tissue, in water; G, H: KOH-extractable pigments after 1 min and 30 min incubation respectively; I: Asexual morph present at the margin of a young stroma; J: Ostiolar disc in surface view; K, L: Dehiscing ostiolar discs *bovei*-type (arrows). Scale bars: A = 10 mm; B = 5 mm; C, E, I = 1 mm; D, J-L = 0.5 mm; F = 10 µm.

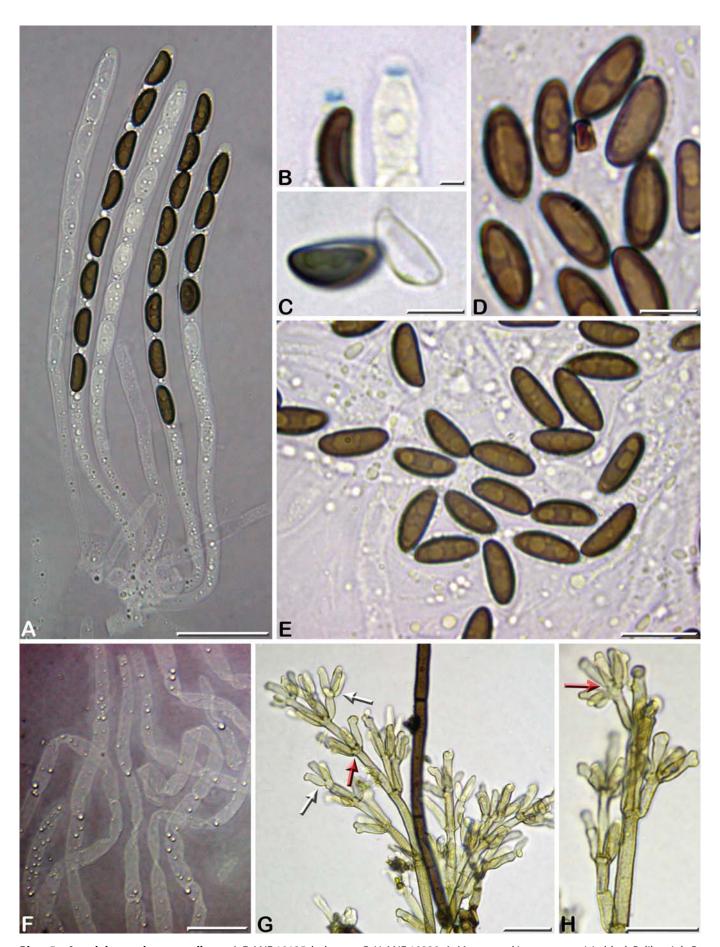


Plate 5 – Annulohypoxylon caravellense. A-F: MJF 10135, holotype; G, H: MJF 10238. A: Mature and immature asci, in black Pelikan ink; B: Ascal apical apparati, in Melzer's reagent; C: Ascospore with perispore dehiscent in 10% KOH; D: Ascospores in dorsal view showing the germ slit, in black Pelikan ink; E: Ascospores in 1% SDS; F: Paraphyses at their base, in black Pelikan ink; G, H: Asexual morph showing a dark brown conidiophore and pale olivaceous conidiogenous structure varying from nodulisporium-like (white arrows) to periconiella-like (red arrows), in 1% SDS. Scale bars: A, F-H = 20 μ m; B = 2 μ m; C, D = 5 μ m; E= 10 μ m.

Table 1 – Ascospore dimensions in seven collections of A. caravellense showing the intraspecific variations

Measurements Collection numbers	Ascospore measurements with extreme values in parentheses	Q = quotient I/w N = number of measurements	Mean values
MJF 07007	$(7.6-)7.9-8.6(-8.9) \times (3.2-)3.3-3.7(-4.0) \ \mu m$	Q = (2.1–)2.2–2.5(–2.6); N = 36	Me = $8.2 \times 3.5 \mu m$; Qe = 2.4
MJF 10039	$(7.1-)7.5-8.3(-8.8) \times (3.2-)3.3-3.7(-3.8) \mu m$	Q = (2.0-)2.1-2.4(-2.6); N = 36	Me = $8.0 \times 3.5 \mu m$; Qe = 2.3
MJF 10135	$(6.9-)7.6-8.4(-8.5) \times (3.2-)3.3-3.6(-3.7) \ \mu m$	Q = (2.1–)2.2–2.4(–2.6); N = 36	Me = $8.0 \times 3.5 \mu m$; Qe = 2.3
MJF 10211	$(6.9-)7.0-8.0(-8.7) \times (3.2-)3.4-3.9(-4.1) \ \mu m$	Q = (1.8–)2.0–2.3(–2.6); N = 36	Me = $7.6 \times 3.6 \mu\text{m}$; Qe = 2.1
MJF 10226	$(7.2-)7.3-8.3(-8.8) \times (3.2-)3.4-3.9(-4.0) \ \mu m$	Q = (1.9–)2.0–2.3(–2.4); N = 36	Me = $7.9 \times 3.7 \mu\text{m}$; Qe = 2.1
MJF 10235	$(7.4-)7.6-8.6(-8.8) \times (2.9-)3.2-3.7(-4.2) \ \mu m$	Q = (2.0-)2.2-2.5(-3.0); N = 60	Me = $8.1 \times 3.5 \mu m$; Qe = 2.3
MJF 13304	$(6.8-)7.5-8.4(-9.1) \times (2.9-)3.2-3.9(-4.2) \ \mu m$	Q = (1.9–)2.0–2.5(–2.7); N = 60	Me = $8.0 \times 3.5 \mu m$; Qe = 2.3
Cumulated values from above	$(6.8-)7.0-8.6(-8.9) \times (2.9-)3.2-3.9(-4.2) \mu m$	Q = (1.9–)2.0–2.5(–3.0); N = 300	Me = $8.0 \times 3.5 \mu m$; Qe = 2.3

KOH-extractable pigments in contrast to the olivaceous or green pigments recorded in other species.

The numerous collections of *A. caravellense* made during this inventorial survey of the mycobiota of the French West Indies over more than 10 years all come from mesophilic to xerophilic forests, predominantly on the east coast of Martinique. The collections from Guadeloupe come from a similar environment, especially in the small islands Les Saintes, also characterized by dry coastal forests, while the other location (Sainte-Anne, in Grande-Terre) is a bit more mesophilic. As such dry coastal forests exist in lowlands of most islands of the Caribbean, we assume that a targeted search in this specific environment would significantly expand the known distribution of *A. caravellense*.

Annulohypoxylon leptascum (Speg.) Y.-M. Ju, J.D. Rogers & H.-M. Hsieh, *Mycologia*, 97 (4): 859 (2005). Plate 6

Stroma fragmentary, effused-pulvinate, with inconspicuous perithecial contours, 40 mm long \times 22 mm wide \times 1.1–1.3 mm thick; surface black, matt, slightly roughened by the ostioles and the ostiolar discs; subsurface carbonaceous, brittle, extending downwards as a crust completely encasing the perithecia, composed of abundant olivaceous brown waxy granules in a carbonaceous matrix, releasing dark green (21, oac82) KOH-extractable pigments, becoming blackish green to olivaceous grey (121) after 30 min incubation; subperithecial tissue 0.25–0.45 mm thick, dark greyish brown; underlying host tissue strongly blackened. **Perithecia** obovoid to tubular, 0.75–1 \times 0.45–0.6 mm. **Ostioles** coarsely conic-papillate, black, encircled with a flattened, slightly immersed disc 0.3–0.35 mm diam *truncatum*-type with strongly notched edges.

Asci absent; minute amyloid bodies representing remnants of apical apparati detected by mounting perithecial contents in Melzer's reagent. **Ascospores** (7.9–)8.6–9.9(–10.4) \times (2.8–)3–3.7(–4) µm, Q = (2.3–)2.5–3(–3.3); N = 60 (Me = 9.1 \times 3.4 µm; Qe = 2.7), fusoid almost equilateral with narrowly rounded ends, pale yellowish brown, with a short straight germ slit originating from one end, frequently at the centre of a more pigmented zone, inconspicuous in water, visible in PVA-lactophenol; perispore indehiscent in 10% KOH; epispore smooth.

Asexual morph on the natural substrate not observed.

Specimen examined: French West Indies: Guadeloupe: Saint-Claude, Matouba, Victor Hugues trail, hygrophilic rainforest, on bark, 4 Sept. 2005, *leg*. C. Lechat, CLL 5340 (LIP).

Known distribution: Brazil, Guadeloupe, ?Thailand, USA (Florida), Venezuela.

Discussion: Our specimen consists of only one stroma in fairly poor condition and lacking asci. However, the combination of effused-pulvinate carbonaceous stroma with inconspicuous perithecial contours, small ostiolar discs with conic-papillate ostioles, green

KOH-extractable pigments and pale brown fusoid-equilateral ascospores with a short germ slit originating from one end and a perispore indehiscent in 10% KOH unambiguously leads to *A. leptascum* as defined by Ju & ROGERS (1996). Due to the overmature state of this stroma we could not observe the typical brown to dark brown outer coating reported by these authors; unlike them who reported the ostiolar discs as slightly convex we observed that the ostiolar discs of our specimen are not convex but slightly lower than their raised margins; it should be noted that they are likewise slightly larger than those reported by Ju & ROGERS (1996), 0.3–0.35 vs. 0.2–0.3 mm diam.

Annulohypoxylon leptascum is primarily characterized by its pale brown fusoid ascospores and their peculiar germ slit morphology. The two other species sharing the same features are

A. leptascum var. macrosporum (Y.-M. Ju & J.D. Rogers) Y.-M. Ju, J.D. Rogers & H.-M. Hsieh and A. urceolatum (Rehm) Y.-M. Ju, J.D. Rogers & H.-M. Hsieh. The former is diagnosed by significantly larger ascospores $19.5-23.5\times4.5-6.5~\mu m$ and the latter by vinaceous purple KOH-extractable pigments (Ju & ROGERS, 1996; FOURNIER & LECHAT, 2016). The germ slit morphology is essential to segregate these three species but the observation of the germ slit on such lightly pigmented ascospores is often inconclusive when they are mounted in water. Mounting the ascospores in PVA-lactophenol and observation after at least 48~h makes the germ slit of ascospores much easier to make out.

Annulohypoxylon palmicola J.K. Liu & K.D. Hyde, recently described from Thailand on palm material (ARIYAWANSA et al., 2016) shares most of characteristic features of A. leptascum, including ostiolar morphology, green KOH-extractable pigments and pale brown fusoid ascospores. Its status is unclear since the protologue deviates in many respects from our observations on the isotype kindly sent by Dr. Liu prior to his publication. The main difference with A. leptascum, regarded as diagnostic by the authors, is the smaller diameter of ostiolar discs 0.1–0.2 mm. However, we recorded the ostiolar discs averaging 0.25 mm diam, thus exactly in the size range 0.2-0.3 mm diam given by Ju & Rogers (1996). Moreover, the ascospores of A. palmicola are described as brown to dark brown, 1-septate and possessing a curved germ slit ¾ of the spore length while, according to our observations, they are typically pale brown, non-septate and with a short straight germ slit originating from one end or without visible germ slit. The dimensions $12-14.5 \times 3.5-5 \mu m$ fit more or less those of A. leptascum in the upper size range (7.5–13 \times 3–4 μ m) but deviate in being more inequilateral. It cannot be ruled out that cryptic species are currently lumped under the name A. leptascum but their segregation should be based on more accurate observations than in the case of A. palmicola.

Annulohypoxylon leptascum is a rarely recorded species. As we collected it only once during our numerous field trips over 12 years in Guadeloupe and Martinique, we are inclined to regard it as a rare species, possibly with narrow ecological requirements.

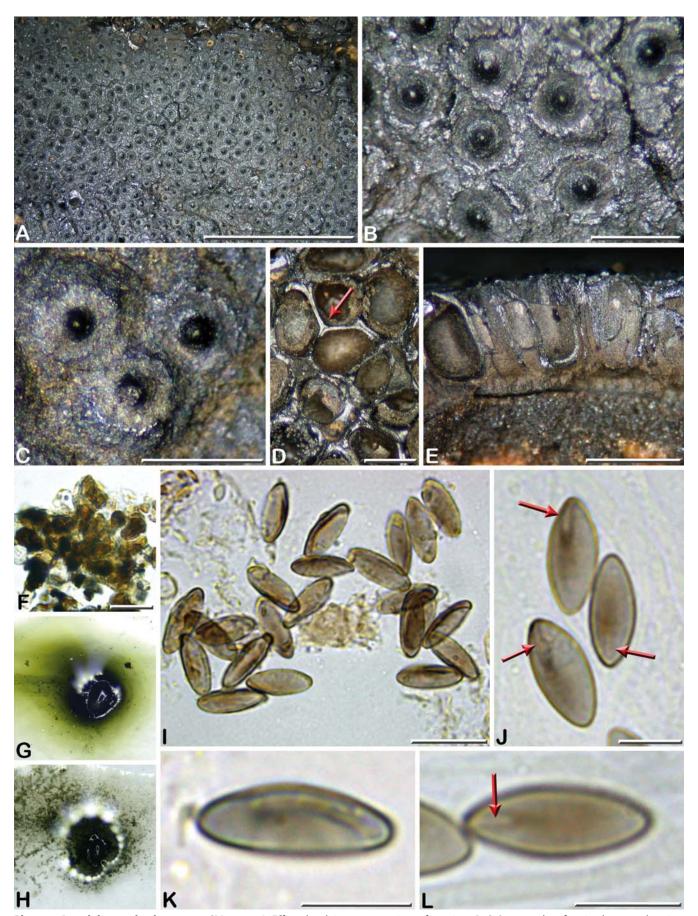


Plate 6 – Annulohypoxylon leptascum CLL 5340. A: Effused-pulvinate stroma in surface view; B, C: Stromatal surface in close-up showing the ostiolar discs and the coarsely papillate ostioles; D: Stroma in horizontal section showing the carbonaceous walls (arrow) delimiting the perithecia; E: Stroma in vertical section (broken) showing the perithecia, the carbonaceous tissue above and between the perithecia, the brown subperithecial tissue and the blackened underlying host tissue; F: Olivaceous brown stromatal waxy granules and blackish carbonaceous fragments from the subsurface, in water; G, H: KOH-extractable pigments after 1 min and 30 min incubation respectively; I: Free ascospores in 1% SDS; J, L: Ascospores in PVA-lactophenol showing the short germ slit next to one end (arrows); K: Ascospore in 10% KOH showing the absence of dehiscing perispore. Scale bars: A = 5 mm; B = 0.5 mm; C = 0.5 mm;

Annulohypoxylon cf. moriforme (Henn.) Y.-M. Ju, J.D. Rogers & H.-M. Hsieh, *Mycologia*, 97(4): 859 (2005). Plates 7–8

Stromata effused-pulvinate to widely effused, coalescent, with inconspicuous to strongly exposed perithecial contours, 3–100 mm $long \times 3-40 \text{ mm}$ wide $\times 0.75-1 \text{ mm}$ thick, associated with widely effused dark brown sterile areas bearing scattered, isolated perithecial contours; outermost coating dark cinnamon (62, oac748), gradually fading, leaving a dull black and roughened squamulose surface; texture carbonaceous, with subsurface composed of dull orange brown granules in a carbonaceous matrix forming a thick layer above and around the perithecia, releasing yellow green (71, oac874) KOH-extractable pigments within 1 min, becoming pale grey olivaceous (107, oac881) upon prolonged incubation; subperithecial tissue 0.25-0.5 mm thick, blackish brown, yellow brown with black streaks in places. **Perithecia** subspherical, 0.45–0.6 mm diam. Ostioles conic-papillate, encircled with a flattened, often slightly sunken, truncatum-type disc 0.25-0.30(-0.35) mm diam with sharp to notched edges.

Asci cylindrical, with eight uniseriate ascospores, $80-88~\mu m$ total length, the spore-bearing parts $55-60\times4.5-5.5~\mu m$, the stipes $20-29~\mu m$ long, with a discoid apical apparatus $0.5-0.6\times1.5~\mu m$, faintly bluing in Melzer's reagent. **Hamathecium** of sparse paraphyses $4-4.5~\mu m$ wide at base, tapering above asci, slightly constricted at septa. **Ascospores** $(6.2-)6.6-7.7(-8.4)\times(2.7-)3.1-3.6(-3.7)~\mu m$, Q=(1.8-)1.9-2.3(-2.5); $N=60~(Me=7.1\times3.3~\mu m$; Q=2.1), ellipsoidinequilateral with most often broadly rounded ends, brown, with a conspicuous straight germ slit spore-length; perispore dehiscent in 10%~KOH, smooth, with a thickening on the dorsal side at ca. $\frac{1}{3}$ spore length; epispore smooth.

Asexual morph on the natural substrate not observed.

Specimen examined: FRENCH WEST INDIES: MARTINIQUE: Prêcheur, Anse-Couleuvre, mesophilic coastal rainforest, dead corticated branch, 27 Aug. 2007, *leg.* J. Fournier & C. Lechat, MJF 07127 (LIP).

Known distribution: Martinique.

Discussion: This *Annulohypoxylon* fits in the concept of *A. moriforme* as defined by Ju & Rogers (1996) by having effused-pulvinate blackish stromata with green KOH-extractable pigments, *truncatum*-type ostiolar discs 0.2–0.4 mm diam and ascospores with a germ slit spore-length and a perispore dehiscent in 10% KOH, within the size range of $6-9 \times 2.5-4 \ \mu m$.

However, it slightly deviates from typical *A. moriforme* by widely effused vs. glomerate stromata, by having paler green KOH-extractable pigments and a dark orange brown outermost coating vs. olivaceous in *A. moriforme*. As it lacks distinctive features, it is unknown whether these differences are incidental or reflect a different taxonomic status. A similar situation was encountered in French Guiana (Fournier & Lechat, 2016) where five collections of *Annulohypoxylon* were tentatively referred to *A. moriforme* although they deviated from one another. The material from Martinique illustrated here likewise deviates from the material from French Guiana, which suggests that *A. moriforme* as currently conceived is likely a wide complex of species which should be revised in the future.

Annulohypoxylon moriforme var. microdiscum (Y.-M. Ju & J.D. Rogers) Y.-M. Ju, J.D. Rogers & H.-M. Hsieh, *Mycologia*, 97(4): 859 (2005). Plate 9

Stromata effused-pulvinate to effused, coalescent, with inconspicuous to strongly exposed perithecial contours, $3-43 \text{ mm long} \times 3-12 \text{ mm wide} \times 0.6-0.85 \text{ mm thick}$, associated with widely effused blackish sterile areas bearing scattered, isolated perithecial contours; outermost coating dull brown to dull olivaceous brown, gradually fading, leaving a dull black and roughened surface; tex-

ture carbonaceous, with subsurface composed of orange brown granules in a carbonaceous matrix forming a thick layer above and around the perithecia, releasing fugacious amber (47, oac852) to ochreous (44, oac756) KOH-extractable pigments within 1 min, becoming fawn (87, oac647) upon prolonged incubation; subperithecial tissue 0.2–0.5 mm thick, blackish brown. **Perithecia** obovoid to subspherical, 0.3–0.4 mm diam. **Ostioles** conic-papillate, encircled with a flattened, often slightly sunken, *truncatum*-type disc 0.15–0.20 mm diam with notched edges.

Asci cylindrical, with eight uniseriate ascospores, 120–128 µm total length, the spore-bearing parts $69-74\times5-5.5$ µm, the stipes 50-55 µm long, with a discoid apical apparatus $0.6-0.8\times1.5-1.8$ µm, bluing in Melzer's reagent. **Hamathecium** of dense, indistinct paraphyses. **Ascospores** (8–)8.8–9.7(–10.3) \times (3.4–)3.7–4.6(–4.9) µm, Q = (1.9–)2–2.5(–2.6); N = 60 (Me = 9.2×4.1 µm; Qe = 2.2), ellipsoid-inequilateral with most often narrowly rounded ends, brown, with a conspicuous straight germ slit spore-length; perispore dehiscent in 10% KOH, smooth, with a thickening on the dorsal side at ca. $\frac{1}{3}$ spore length; epispore smooth.

Asexual morph on the natural substrate not observed. Asexual morph in culture on OA nodulisporium-like, based on material from Taiwan (Ju & ROGERS, 1996).

Specimens examined: French West Indies: Martinique: Trinité (Caravelle peninsula), Pointe-Rouge, meso-xerophilic forest, corticated branch, 31 Aug. 2007, *leg.* J. Fournier, MJF 07228 (LIP). Taiwan: Taipei City, Nankang, campus of Academia Sinica, corticated branch, 17 Jan. 2002, *leg.* Y.-M. Ju, Ju Y.-M.91011701. Thailand: Chiang Mai Prov., Mae Teang district, Bahn Pha Deng, Mushroom Research Centre, N 19° 01′ 6.15″ E 98° 41′ 8.84″, ca. 900 m, corticated branch, 26 May 2005, *leg.* J. Fournier, JF-TH 26–02.

Known distribution: Martinique, Taiwan, Thailand.

Discussion: This *Annulohypoxylon* is characterized by blackish brown stromata with often conspicuous perithecial contours, small ostiolar discs truncatum-type less than 0.2 mm diam, amber to fawn KOH-extractable pigments and ascospores 9–10 \times 3.7–4.6 μ m. It conforms to A. moriforme var. microdiscum as briefly described by Ju & Rogers (1996) except in the colour of KOH-extractable pigments that are not olivaceous green as in A. moriforme var. moriforme. However, examination of material from Taiwan kindly provided by Dr. Y.-M. Ju showed that pigments appeared more brownish than olivaceous, a feature also observed in material collected in Thailand from which the presence of cohaerin-type azaphilones was reported by FOURNIER et al. (2010). Cohaerins were so far only known from the north temperate A. cohaerens (Pers.) Y.M. Ju, J.D. Rogers & H.M. Hsieh (QUANG et al., 2005; 2006) and their presence in a tropical species was unexpected, making it a precious differential character. The HPLC profile of the material from Martinique revealed the presence of cohaerins (STADLER, unpublished results), supporting the morphological similarities between the Asian and the Caribbean material.

Annulohypoxylon nitens (Ces.) Y.-M. Ju, J.D. Rogers & H.-M. Hsieh, *Mycologia*, 97 (4): 861 (2005). Plates 10–11

Stromata effused-pulvinate to glomerate, at times perithecioid, with usually strongly exposed perithecial contours, 2–70 mm long \times 2–26 mm wide \times 1–1.8 mm thick; surface typically becoming shiny black by flaking off of the reddish brown outermost coating which is continuous with a sterile layer extending on bark surrounding young stromata; mature stromata, especially those with slightly exposed perithecial contours, occasionally dull black, with dark grey tissue and persistent scales roughening the shiny black subsurface; texture carbonaceous, with subsurface predominantly composed of olivaceous yellow granules mixed with black carbonaceous tissue, forming a thick layer above and around the perithecia, releasing

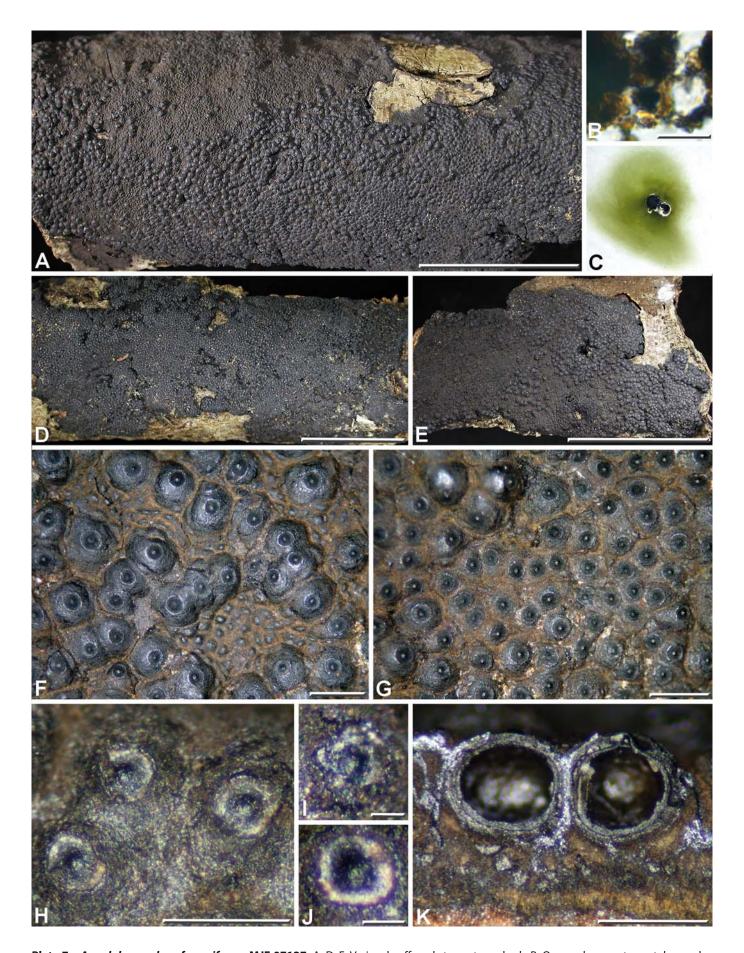


Plate 7 – Annulohypoxylon cf. moriforme MJF 07127. A, D, E: Variously effused stromata on bark; B: Orange brown stromatal granules mixed with carbonaceous tissue, in water; C: KOH-extractable pigments after 1 min incubation; F, G: Stromatal surface showing the orange brown outermost coating, the ostiolar discs and the variously exposed perithecial contours; H-J: Close-up on dehiscing truncatum-type ostiolar discs; K: Stroma in vertical section showing the perithecia and the yellow brown and black subperithecial tissue. Scale bars: A, D, E = 20 mm; B = 10 \mu m; F, G = 1 mm; H, K = 0.5 mm; I, J = 100 \mu m.

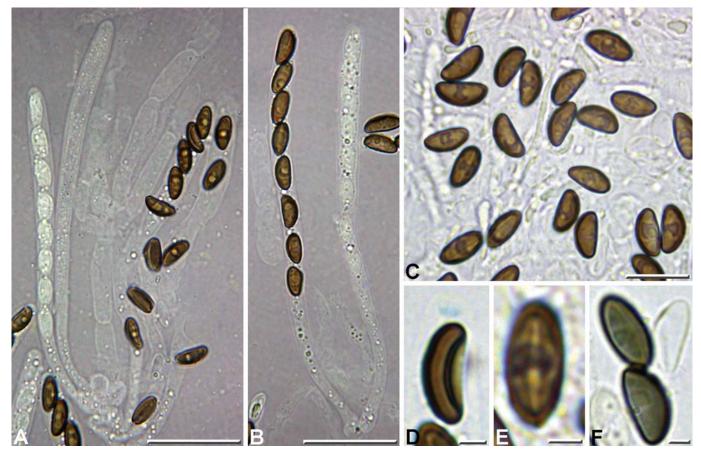


Plate 8 – **Annulohypoxylon cf. moriforme MJF 07127.** A, B: Immature and mature asci with paraphyses, in black Pelikan ink; C: Ascospores in 1% SDS; D: Ascal apical apparatus in Melzer's reagent; E: Ascospore in dorsal view showing the germ slit; F: Ascospores in 10% KOH with a dehiscent perispore. Scale bars: A, B = $20 \mu m$; C = $10 \mu m$; D-F = $2 \mu m$.

greenish-olivaceous (90, oac860) KOH-extractable pigments within 1 min incubation, evolving to pale olivaceous grey (120, oac884) upon prolonged incubation; subperithecial tissue 0.4–1.2 mm thick, dark brownish grey, woody-carbonaceous. **Perithecia** subspherical, 0.6–0.7 mm diam. **Ostioles** conic-papillate, encircled with a flattened *bovei*-type disc 0.4–0.5 mm diam with sharp to notched rims.

Asci cylindrical, with eight uniseriate ascospores, 120–140 µm total length, the spore-bearing parts $56-63 \times 4.5-5$ µm, the stipes 65-82 µm long, with a discoid apical apparatus $0.5 \times 1.5-1.7$ µm, faintly bluing in Melzer's reagent, often very inconspicuous or not bluing. **Paraphyses** 4-5 µm wide at base, septate, thin-walled, tapering above asci and embedded in mucilage. **Ascospores** $(6.9-)7.1-8.2(-8.8) \times (2.7-)3-3.6(-3.7)$ µm, Q=(2-)2.1-2.5(-2.8); N=60 (Me = 7.6×3.3 µm; Q=2.3), ellipsoid slightly inequilateral with narrowly to broadly rounded ends, brown, with a straight germ slit almost spore-length to spore-length; perispore dehiscent in 10% KOH, smooth, with a thickening on the dorsal side at ca. $\frac{1}{12}$ spore length; epispore smooth. (**Ascospores** of CLL 1088, CLL 2328 and ZY 07046 average 7.6×3.3 µm, 7.2×3.0 µm and 7.6×3.1 µm respectively)

Asexual morph on the natural substrate not seen. Asexual morph in culture on OA nodulisporium-like, based on material from Mexico and Taiwan (Ju & ROGERS, 1996).

Specimens examined: French West Indies: Guadeloupe: Sainte-Anne, Douville, Liard, xerophilic to mesophilic forest, on corticated wood, 13 Sept. 2003, *leg*. C. Lechat, CLL 1088 (LIP); Le Gosier, Grand-Bois, Montête, xerophilic to mesophilic forest, dead corticated branch, 4 Sept. 2004, *leg*. C. Lechat, CLL 2327 (LIP); *ibid.*, CLL 2328 (LIP). MARTINIQUE: Prêcheur, Anse-Couleuvre, coastal mesophilic forest, dead corticated branch, 23 Aug. 2007, *leg*. J. Fournier, MJF 07040 (LIP); *ibid.*, 2 Sept. 2007, leg. J. Fournier, MJF 07260 (LIP) (barely

mature). P. R. China: Hainan, Wuhzi mountain, ca. 700 m elevation, rainforest, dead decorticated wood, Aug. 2007, *leg*. Y. Zhang, ZY 07046 (JF).

Known distribution: Pantropical.

Discussion: The definition of A. nitens according to Ju & ROGERS (1996) is based on the combination of stromata with conspicuous perithecial contours and a shiny black surface, greenish-olivaceous KOH-extractable pigments and bovei-type ostiolar discs. This definition is problematic since it encompasses species with ostiolar discs varying from 0.2 to 0.5 mm diam, with perithecia 0.4–1.2 mm diam and ascospores $6.5-10 \times 3-4.5 \mu m$. From this wide concept, these authors segregated only A. purpureonitens based on purple KOHextractable pigments (Ju & Rogers, 1996). Field observations on Annulohypoxylon spp. with a somewhat shiny black surface strongly suggest that, as currently conceived, A. nitens is a complex of related species. Segregating a species from this complex implies to give A. nitens a narrower definition based on the type material. We unfortunately could not revise the type material from Malaysia (Borneo, Sarawak) and soundly re-defining A. nitens would require the designation of an epitype based on material collected in the same region from which cultural and molecular data would be obtained. In the meantime, it appears practical to restrict A. nitens to species with stromata becoming conspicuously shiny black, greenish-olivaceous KOH-extractable pigments and bovei-type ostiolar discs. Since the three above specimens from Guadeloupe and Martinique and the specimen from Hainan match well this concept, they are assumed to represent typical A. nitens. The only difference of the material from Hainan from the Caribbean material is the more strongly amyloid reaction of the ascal apical apparati. As they share perithecia 0.6-0.7 mm diam, ostiolar discs 0.4-0.5 mm diam and ascospores

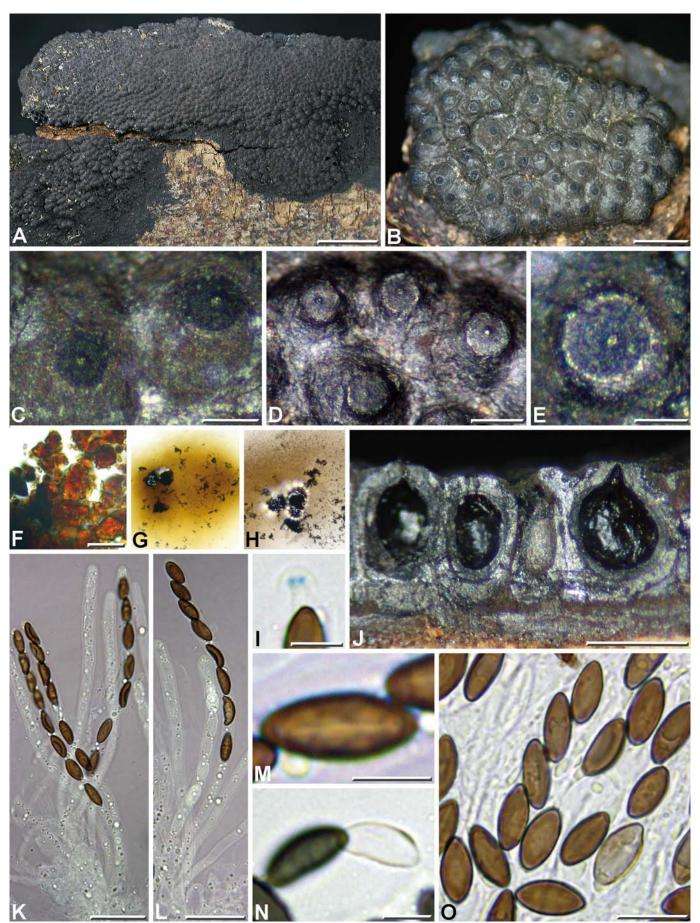


Plate 9 – Annulohypoxylon moriforme var. microdiscum MJF 07228. A, B: Effused-pulvinate stromata on bark; C-E: Stromatal surface in close-up showing the olivaceous to dull black surface colour and the *truncatum*-type ostiolar discs; F: Orange brown stromatal granules mixed with carbonaceous tissue, in water; G, H: KOH-extractable pigments after 1 min and 30 min incubation respectively; I: Ascal apical apparatus in Melzer's reagent; J: Stroma in vertical section showing the perithecia and the black subperithecial tissue; K, L: Mature and immature asci in black Pelikan ink; M: Ascospore in dorsal view showing the germ slit; N: Ascospore in 10% KOH with a dehiscent perispore; O: Ascospores in 1% SDS. Scale bars: A = 5 mm; B = 1 mm; C = 0.2 mm; C = 0.1 mm; $C = 0.1 \text{ m$

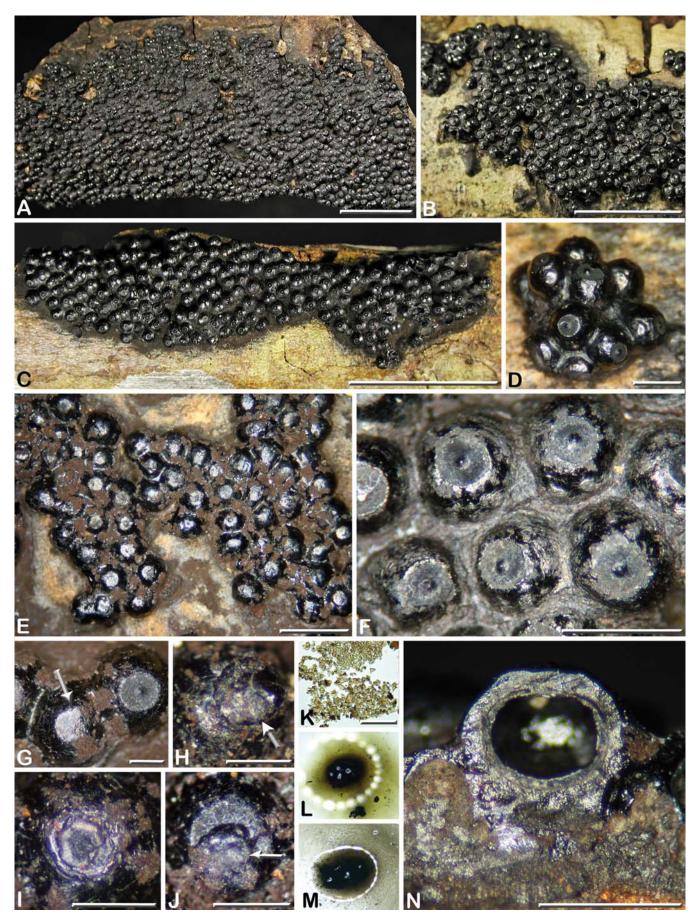


Plate10 – **Annulohypoxylon nitens.** A, E-N: MJF 0740; B, D: CLL 2328; C: CLL 1088. A-C: Mature effused-pulvinate stromata on bark; D: Glomerate stroma entirely shiny black; E: Immature stroma showing the reddish brown outermost coating and the shiny black perithecioid stromata prior to the dehiscence of ostiolar discs; F: Mature stroma in close-up showing the partly shiny black surface and the boveitype ostiolar discs; G: Stromatal apex prior to the dehiscence of the disc (arrow); H, J: Ostiolar discs detached in one piece from the stroma (arrows); I: Atypical dehiscence of a disc by crumbling from the centre; K: Olivaceous yellow stromatal granules mixed with carbonaceous particles, in water; L, M: KOH-extractable pigments after 1 min and 30 min incubation respectively; N: Stroma in vertical section. Scale bars: A-C = 10 mm; D, F, N = 1 mm; E = 2 mm; G-J = 0.5 mm; K = 10 μm.

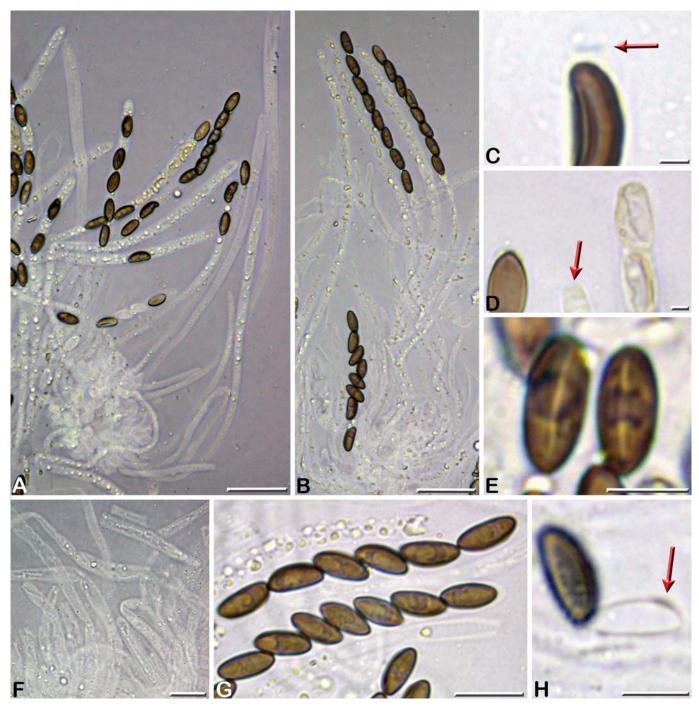


Plate 11 – Annulohypoxylon nitens MJF 07040. A: Immature and few-spored mature asci in black Pelikan ink showing the swollen contorted ascogenous hyphae; B: Mature and immature asci in in black Pelikan ink showing the long stipes; C: Faintly amyloid apical apparatus (arrow), in Melzer's reagent; D: Three ascal apices in Melzer's reagent, showing the apparent absence of apical apparatus or very faint amyloid reaction (arrow); E: Ascospore in dorsal view showing the germ slit, in 1% SDS; F: Paraphyses in black Pelikan ink; G: Ascospores in 1% SDS; H: Ascospore in 10% KOH with a dehiscent perispore showing the dorsal thickening (arrow). Scale bars: A, B = 20 μ m; C, D = 2 μ m; E, H = 5 μ m; F, G = 10 μ m.

7–8 × 3–3.5 μm, these characters contribute to define *A. nitens* more accurately. This somewhat arbitrary but provisional interpretation of *A. nitens* is justified by the necessity to segregate several species with more or less shiny black surface substantially deviating from this concept and from each other, that would have otherwise been lumped under the name *A. nitens*. In this way, *A. subnitens* J. Fourn. & Lechat was segregated from *A. nitens* based on inconspicuous KOH-extractable pigments and smaller ostiolar discs (FOURNIER & LECHAT, 2016). In the present paper, *A. caravellense* is recognized as a new taxon based on darker green KOH-extractable pigments, smaller ostiolar discs and slightly larger ascospores; *A. splendens* is likewise segregated from *A. nitens* by smaller *truncatum*-type ostiolar

discs and larger ascospores. These new species will be interestingly re-evaluated when the species complex around *A. nitens* is revised.

Annulohypoxylon purpureonitens (Y.-M. Ju & J.D. Rogers) Y.-M. Ju, J.D. Rogers & H.-M. Hsieh, Mycologia, 97 (4): 861 (2005). Plates 12–13

Stromata effused-pulvinate with $\frac{1}{3}$ to $\frac{3}{4}$ exposed perithecial contours, perithecioid in places, 3–72 mm long \times 2–30 mm wide \times 0.85–1.1 mm thick, frequently associated with dark vinaceous brown to dull black sterile primordial areas; outermost coating dark



Plate 12 – **Annulohypoxylon purpureonitens.** A-F, H-J: MJF 10124; G: MJF 13312; K-N: MJF 13133. A, C, D, G: Effused to perithecioid stromata in surface view; note the vinaceous brown tissue at margins in A, C and D; B: Waxy stromatal granules in water; E, F: KOH-extractable pigments after 1 and 30 min incubation respectively; H, J: Stromatal surface in close-up showing the vinaceous brown coating, the shiny subsurface and the ostiolar discs; I: Stroma in vertical section showing the thick subperithecial tissue with vinaceous brown inclusions; K-N: Ostiolar discs at various stages of *truncatum*-type dehiscence. Scale bars: A, C = 10 mm; B = 10 μ m; D, G = 2 mm; H-J, L, N = 0.5 mm; K, M = 0.1 mm.

vinaceous (82, oac523) to blackish brown, persistent or cracking and flaking off into large scales and gradually revealing a smooth and shiny black surface; shiny crust 40–70 μ m thick, brittle, completely encasing the perithecia and spreading around the base of perithecioid stromata and uniting them, primarily composed of reddish brown waxy granules in a loose carbonaceous matrix, releasing pale

vinaceous purple (101, oac401) KOH-extractable pigments, eventually pale purplish grey (128, oac528) after 30 min incubation; subperithecial tissue 0.2–0.5 mm thick, dark brown, frequently with vinaceous brown inclusions. **Perithecia** subspherical, 0.5–0.6 mm diam. **Ostioles** finely papillate, encircled with a flattened disc 0.3–0.35 mm diam *truncatum*-type with slightly notched edges.

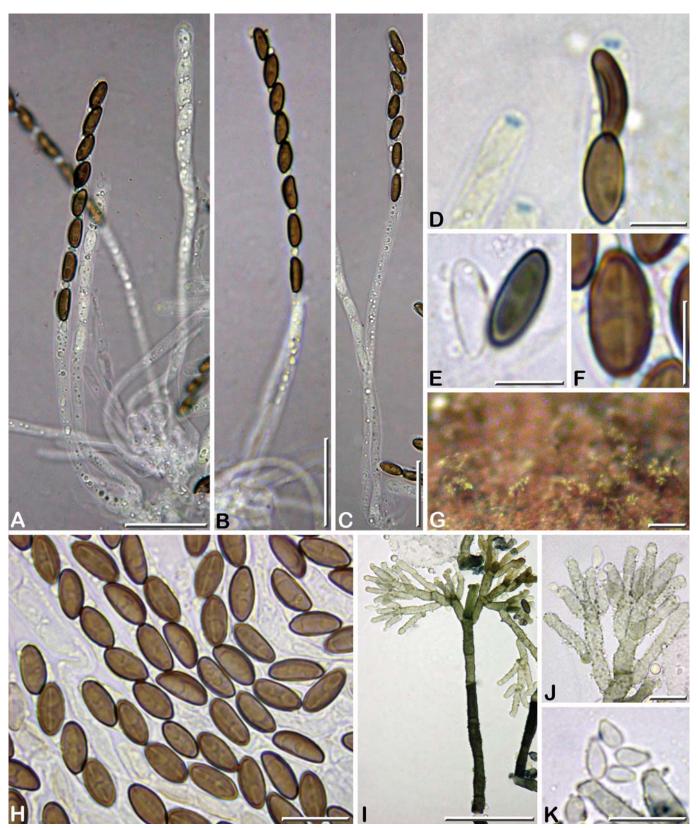


Plate 13 – **Annulohypoxylon purpureonitens MJF 10124.** A-C: Short-stipitate and long-stipitate asci, in black Pelikan ink; D: Ascal apical apparati, in Melzer's reagent; E: Ascospore in 10% KOH showing the dehiscent perispore with a thickening on the dorsal side; F: Ascospore in dorsal view in water, showing the germ slit; G: Asexual morph on the natural substrate; H: Ascospores in water, some showing the germ slit; I: Conidiophore and nodulisporium-like conidiogenous structure, in 10% KOH; J, K: Conidiogenous cells and conidia, in 10% KOH. Scale bars: A, C = 20 μ m; D-F = 5 μ m; G = 100 μ m; H, J, K = 10 μ m; I = 50 μ m.

Asci cylindrical, with eight uniseriate ascospores, originating from short contorted swollen ascogenous hyphae, $100-160~\mu m$ total length, the spore-bearing parts $54-63\times4~\mu m$, the stipes $38-70(-100)~\mu m$ long, with a discoid apical apparatus $0.5-0.8\times1.4-1.7~\mu m$, bluing in Melzer's reagent. **Paraphyses** filiform, slightly embedded in mucilage. **Ascospores** $(6.9-)7.2-8.1(-8.6)\times(2.6-)2.8-3.3(-3.6)~\mu m$, Q=(2.1-)2.3-2.8(-3.1); $N=60~M=7.6\times3~\mu m$; Q=2.6, ellipsoid slightly inequilateral with narrowly to broadly rounded ends, medium brown, with a straight germ slit spore-length; perispore dehiscent in 10%~KOH at a small percentage, smooth, with a thickening on the dorsal side; epispore smooth.

Asexual morph on the natural substrate: Present in places on the reddish brown margins of young stromata, forming honey (64, oac847) scattered tufts; conidiophores upright, septate, 45–75 μm high, 4.5–6 μm wide, dark brown, coarsely roughened; conidiogenous structure nodulisporium-like to periconiella-like in places, conidiogenous cells 7–18 \times 3.2–4.5 μm , roughened, turning pale green in 10% KOH; conidia ellipsoid, 3.5–5.5 \times 1.8–3 μm , subhyaline, smooth.

Specimens examined: Brazil: Serra Araca, corticated wood, 10-13 Mar. 1984, leg. G.J. Samuels, G.J. 808 (WSP 69635, isotype of A. purpureonitens). Guadeloupe: Sainte-Anne, Douville, Liard, mesophilic to xerophilic forest, on corticated wood, 13 Sept. 2003, leg. C. Lechat, CLL 1067 (LIP). MARTINIQUE: Le Robert, Pointe-Bateau, xerophilic coastal forest, dead corticated branch, 26 Aug. 2010, leg. J. Fournier, MJF 10124 (LIP); Trinité (Caravelle peninsula), Balata, ZNIEFF 0001, xerophilic coastal forest, dead corticated branch, 11 Aug. 2013, leg. J. Fournier, MJF 13121 (LIP); ibid., MJF 13129 (LIP); ibid., MJF 13133 (LIP); Schoelcher, Fond-Lahaye, mesophilic rainforest, dead corticated branch, 12 Aug. 2013, leg. J. Fournier, MJF 13167 (LIP); Macouba, Trou-Navet, hygrophilic rainforest, decorticated dead wood, 13 Aug. 2013, leg. J. Fournier, MJF 13185 (LIP); Le Diamant, Morne-Blanc, trail from Ancinel to Morne-du-Riz through Morne-Fournirey, meso- to xerophilic forest, dead corticated branch, 18 Aug. 2013, leg. J. Fournier, MJF 13312 (LIP). MEXICO: Chiapas state, Ocosingo municipality, Ejido Boca de Chajul, wood, 29 May 1988, leg. F. San Martín, San Martin-862 (JDR, A. purpureonitens).

Known distribution: Brazil, Guadeloupe, Martinique, Mexico, ?Thailand.

Discussion: Annulohypoxylon purpureonitens was segregated from its look-alike A. nitens by different purplish KOH-extractable pigments vs. greenish-olivaceous in A. nitens (Ju & Rogers, 1996, as Hypoxylon purpureonitens). Such an Annulohypoxylon species with a shiny black stromatal surface and purplish KOH-extractable pigments was repeatedly collected during this survey, exhibiting strong variations in stromatal habit but consistently with pale purplish KOH-extractable pigments turning grey over time and ascospores averaging 7–7.6 \times 3–3.6 µm, that we assigned to *A. purpureonitens*. Annulohypoxylon purpureonitens is a rarely reported species, so far known from Brazil and Mexico (Ju & Rogers, 1996) and possibly from Thailand (Makornwattana et al., 2013). The isotype from Brazil and the collection from Mexico were studied for comparison with A. nouraguense J. Fourn. & Lechat, a new species from French Guiana (Four-NIER & LECHAT, 2016). The specimens from Guadeloupe and Martinique deviate from the isotype of A. purpureonitens in having thinner stromata 0.85-1.1 mm thick vs. 2 mm thick, paler KOH-extractable pigments and smaller ascospores 7–7.6 \times 3–3.6 μ m vs. 7.9– 9×3 –3.6 µm. Interestingly, they resemble in these respects the specimen San Martin-862 from Mexico, which however deviates by significantly larger ostiolar discs 0.5–0.6 mm diam. Annulohypoxylon nouraguense was segregated from A. purpureonitens by KOH-extractable pigments turning hazel upon prolonged incubation, a distinctive feature which sets it apart from all taxa closely related to A. purpureonitens.

It is noteworthy that the material studied above features *trunca-tum*-type ostiolar discs, unlike *A. nitens* in which they are *bovei*-type. Unfortunately, the type of dehiscence of ostiolar discs of the isotype material could not be assessed for comparison.

These somewhat discordant observations suggest that *A. purpureonitens* might encompass several cryptic species that more extensive sampling could help resolve. For the time being, the material collected in Guadeloupe and Martinique illustrated above is assigned to *A. purpureonitens sensu lato*.

Annulohypoxylon purpureopigmentum Jad. Pereira, J.D. Rogers & J.L. Bezerra, *Mycologia*, 102 (1): 250 (2010). Plates 14–15

Stromata glomerate and few-peritheciate or effused-pulvinate, 0.8–2 mm diam, confluent into irregularly effused-pulvinate stromata on bark or linear rows on decorticated wood, 5–20 mm long × 1.3–5.2 mm wide × 0.7–1.2 mm thick, with ½ exposed to fully exposed perithecial contours; surface even, matt, with persistent pruinose outermost coating rust (39, oac644) to umber with olivaceous tone (9, oac736), turning blackish brown at maturity; texture carbonaceous around the perithecia, with subsurface composed of pale orange brown granules yielding dense livid purple (81, oac439) KOH-extractable pigments within 1 min incubation, fading to pale vinaceous grey (115, oac569) upon prolonged incubation; subperithecial tissue 0.2–0.5 mm thick, brown to blackish. **Perithecia** subspherical, 0.4–0.5 mm diam. **Ostioles** conic-papillate, encircled with a black, flattened *truncatum*-type disc 0.20–0.25 mm diam with sharp to irregular rims.

Asci cylindrical, with eight ascospores arranged uniseriately, 85–120 µm total length, the spore-bearing parts $46-52\times3.5-4$ µm, the stipes 38-60(-72) µm long, with a discoid apical apparatus $0.6-0.7\times1.4-1.6$ µm, bluing in Melzer's reagent; asci arising from swollen, moniliform, contorted hyphae 3-5 µm wide. **Paraphyses** filiform 1-2.5 µm wide, with refractive contents, embedded in mucilage. **Ascospores** $(6-)6.2-7(-7.4)\times(2.4-)2.6-3(-3.3)$ µm, Q=(2.1-)2.2-2.5(-2.7); N=120 (Me $=6.6\times2.8$ µm; Q=2.4), ellipsoid slightly inequilateral with narrowly to broadly rounded ends, brown, with a straight germ slit almost spore-length; perispore dehiscent in 10% KOH, smooth, with a thickening on the dorsal side at ca. $\frac{1}{3}$ spore length; epispore smooth.

Asexual morph on the natural substrate not seen.

Specimens examined: FRENCH WEST INDIES: MARTINIQUE: Case-Pilote, Morne-Rose, mesophilic rainforest, dead corticated and decorticated branch, associated with overmature stromata of *Annulohypoxylon* sp., 1 Sept. Aug. 2010, *leg.* J. Fournier, MJF 10309 (LIP); Saint-Joseph, Coeur-Bouliki forest, hygrophilic rainforest, on dead corticated branchlets in a Mahogany (*Swietenia macrophylla* King, *Meliaceae*) plantation, 26 Aug. 2007, *leg.* J. Fournier, MJF 07109 (LIP).

Known distribution: Brazil, French Guiana, Martinique.

Discussion: Annulohypoxylon purpureopigmentum is a distinctive species, readily recognized by its small, often glomerate stromata with conspicuous perithecial contours, with livid purple KOH-extractable pigments and small black ostiolar discs contrasting with the dull brown persistent pruinose surface. It was recently described from Brazil and its morphological differences from other Annulohypoxylon spp. with purplish KOH-extractable pigments were discussed in detail by Pereira et al. (2010). The material from Martinique studied here conforms well to the collection from French Guiana illustrated and discussed by Fournier & Lechat (2016) and to the original description. Our observations confirm that ostiolar discs are of the truncatum-type, unlike the original description of bovei-type ostiolar discs.

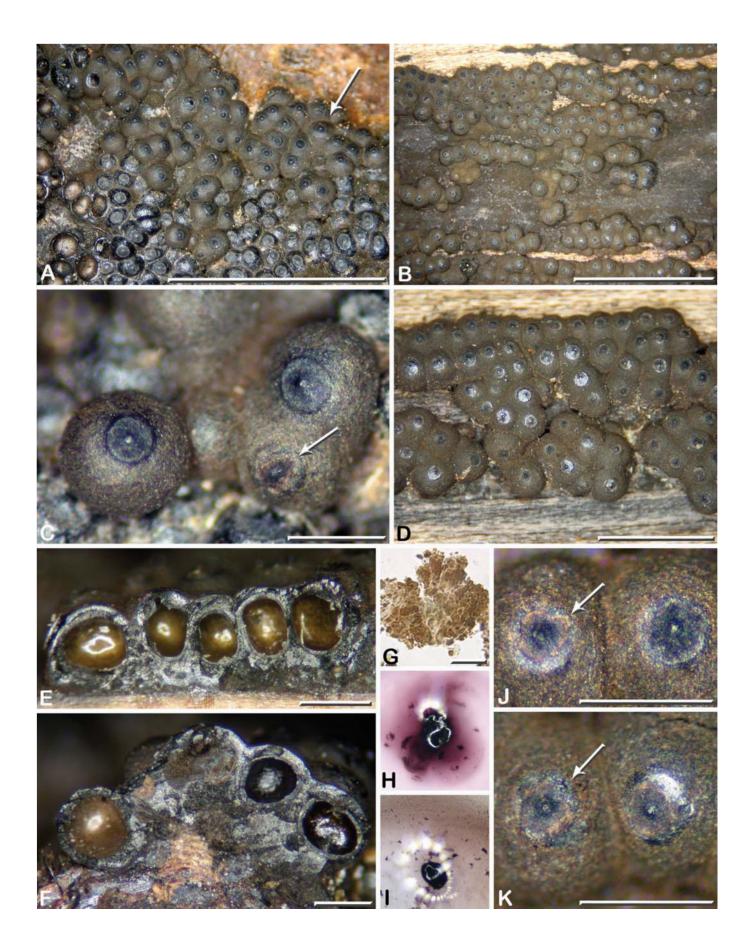


Plate 14 – *Annulohypoxylon purpureopigmentum* **MJF 10309.** A: Effused-pulvinate and perithecioid stromata (arrow) over an overmature shiny Annulohypoxylon sp.; B: Immature stromata on decorticated wood; C: Close-up on perithecioid stromata showing the black *trunca-tum*-type ostiolar discs with one in formation (arrow); D: Mature confluent stromata on decorticated wood; E: Effused immature stroma in vertical section; F: Mature glomerate stroma in vertical section; G: Stromatal waxy granules from subsurface, in water; H, I: KOH-extractable pigments after 1 min and 30 min incubation respectively; J, K: Close-ups on two adjacent ostiolar discs of immature stromata, showing the outermost stromatal tissue flaking off outwardly from the ostiole (arrows). Scale bars: A, B = 5 mm; C, E, F, J, K = 0.5 mm; D = 2 mm; G: 10 μ m.

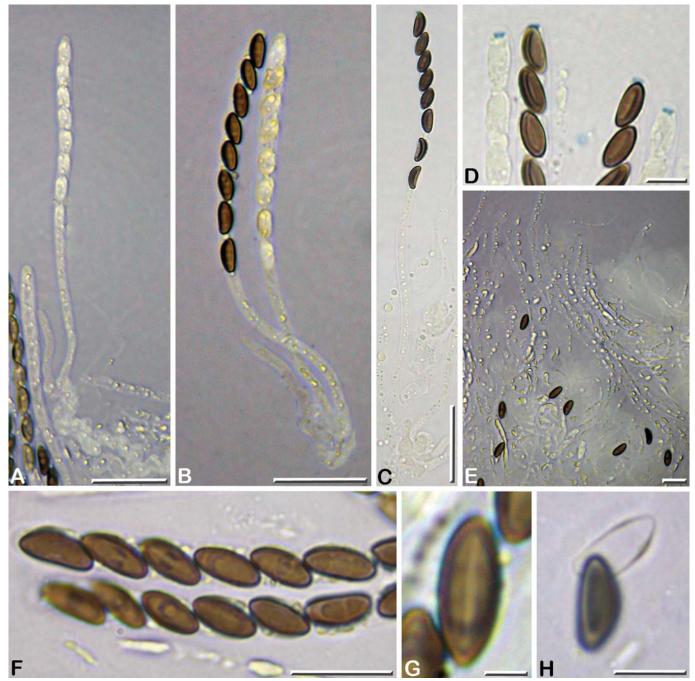


Plate 15 – **Annulohypoxylon purpureopigmentum MJF 10309.** A: Immature asci arising from a swollen hypha, in black Pelikan ink; B: Mature and immature asci, in black Pelikan ink; C: Long-stipitate ascus, in Melzer's reagent; D: Ascal apical apparati in Melzer's reagent; E: Paraphyses with refractive contents and embedded in mucilage, in black Pelikan ink; F: Ascospores in 1% SDS, some showing a faint germ slit; G: Ascospore in dorsal view showing a germ slit, in 1% SDS; H: Ascospore in 10% KOH showing the dehiscent perispore with a thickening on the dorsal side. Scale bars: $A-C = 20 \mu m$; $D, H = 5 \mu m$; $E, F = 10 \mu m$; $C = 2 \mu m$.

Annulohypoxylon ramulorum J. Fourn. & Lechat, *sp. nov.* – MycoBank MB 816976. Plates 16–17

Diagnosis: Differs from the most resembling species *Annulohypoxylon moriforme* by a red-brown woolly stromatal surface of young stromata, a periconiella-like asexual morph on natural substrate and consistent occurrence on twigs and branchlets.

Holotype: French West Indies: Martinique: Case-Pilote, Morne-Rose, mesophilic rainforest, on corticated twigs and branchlets 5–12 mm diam, 1 Sept. 2010, *leg.* J. Fournier, MJF 10312 (LIP).

Etymology: From Latin *ramulus* = branchlet, twig, for the stromata unusually and consistently occurring on small branches and twigs.

Stromata glomerate, rarely slightly pulvinate, few-peritheciate, with strongly exposed perithecial contours to often perithecioid, 0.6-3 mm diam \times 0.6-0.9 mm thick, coalescent into clusters 5-18 mm in their greatest dimension; outermost coating dark brick (60, oac635), woolly, bearing the associated asexual morph, gradually vanishing, revealing a dull black and roughened surface at maturity; texture weakly carbonaceous, with subsurface composed of olivaceous yellow waxy granules appearing dull orange brown in mass, interspersed in a carbonaceous matrix forming a thick layer above and around the perithecia, releasing greenish olivaceous (90, oac859) KOH-extractable pigments within 1 min, becoming grey olivaceous (107, oac882) upon prolonged incubation; subperithecial tissue inconspicuous to 0.3 mm thick, blackish. **Perithecia** subsphe-

rical, 0.4–0.5 mm diam. **Ostioles** conic-papillate, encircled with a flattened, *truncatum*-type disc 0.20–0.3 mm diam with sharp to notched edges.

Asci cylindrical, with eight uniseriate ascospores, 75–105 μm total length, the spore-bearing parts $53–59 \times 4–5$ μm, the stipes 19–48 μm long, with a discoid apical apparatus $0.5–0.6 \times 1.5–1.8$ μm, faintly bluing in Melzer's reagent. **Hamathecium** of sparse paraphyses 3-4.5 μm wide at base, tapering above asci, not constricted at septa. **Ascospores** $(6.2–)6.6–8.4(–9.2) \times (2.9–)3.1–3.8(–3.9)$ μm, Q=(1.8–)1.9–2.4(–2.5); N=190 (Me = 7.5×3.4 μm; $N=1.5 \times 3.4$ μ

Asexual morph on the natural substrate: Present at margins of young stromata, forming dense pale brown tufts emerging from a dense red-brown hyphal mat; conidiophores upright, with a stout brown to dark brown septate axis 280–325 μ m high \times 5–6 μ m wide, apically branching, coarsely roughened; conidiogenous structure nodulisporium-like to periconiella-like, with conidiogenous cells 11– 13 \times 2.5–3 μ m, finely roughened, pale brown to subhyaline, turning greenish in 3% KOH; conidia pale brown, smooth, ellipsoid, 3.5–4.5 \times 1.8–2.2 μ m.

Other specimens examined (paratypes): French West Indies: Gua-DELOUPE: State Forest of Basse-Terre, hygrophilic rainforest, on corticated branchlet 1.3 cm diam, 3 Sept. 2005, leg. C. Lechat, CLL 5331 (LIP). MARTINIQUE: Saint-Esprit, Morne-David, mesophilic rainforest, on corticated branchlet 1 cm diam, 23 Aug. 2004, leg. C. Lechat, CLL 2027 (LIP); ibid., leg. C. Lechat, CLL 2032 (LIP); ibid., leg. C. Lechat, CLL 2033 (LIP); ibid., leg. C. Lechat, CLL 2038 (LIP); Saint-Esprit, Bois la Charles, mesophilic rainforest, on corticated branchlet 2 cm diam, Aug. 2004, leg. C. Lechat, CLL 2055 (LIP); ibid., leg. C. Lechat, CLL 2067 (LIP); Case-Pilote, Crête Jean-Louis, mesophilic rainforest, on corticated branchlet 2 cm diam, 21 Aug. 2005, leg. C. Lechat, CLL 5083 (LIP); Case-Pilote, Fond-Bourlet, Prise d'Eau, on corticated branchlets 8-12 mm diam, 21 Aug. 2005, leg. C. Lechat, CLL 5102 (LIP); Fort-de-France, Absalon, trail to Plateau-Michel, hygrophilic rainforest, on corticated branchlet 2-3 cm diam, 7 Aug. 2013, leg. J. Fournier, MJF 13044 (LIP); Case-Pilote, Morne-Rose, mesophilic rainforest, on corticated twigs and branchlets 5-12 mm diam, 14 Jun. 2015, leg. C. Lechat & J. Fournier, MJF 15064 (LIP).

Known distribution: Guadeloupe, Martinique.

Discussion: This *Annulohypoxylon* is readily set apart from other *Annulohypoxylon* spp. occurring in Guadeloupe and Martinique by its small glomerate stromata with small ostiolar discs 0.2–0.25 mm diam and coated with a red-brown woolly outermost layer when young. This red-brown coating is conspicuous in the type specimen MJF 10312 and in CLL 5331, along with dull black more mature stromata.

It fits in the wide concept of *A. moriforme* defined by Ju & ROGERS (1996) by having glomerate stromata with greenish olivaceous KOH-extractable pigments, *truncatum*-type ostiolar discs 0.2–0.4 mm diam and ascospores within the size range of 6–9 \times 2.5–4 μ m, with a germ slit spore-length and a perispore dehiscent in 10% KOH.

However, it deviates from typical *A. moriforme* by small glomerate stromata that do not become widely effused-pulvinate and a woolly red-brown outermost coating, well developed when young, in contrast to an olivaceous one in *A. moriforme*. As these characters are associated with small ostiolar discs in the lower range given for *A. moriforme*, a periconiella-like asexual morph on natural substrate and an apparent strong host preference for twigs and branchlets based on seven collections, we propose to segregate it from the *A. moriforme* complex. Typical *A. moriforme* was not encountered during this survey in Guadeloupe and Martinique.

Annulohypoxylon splendens J. Fourn. & Lechat, sp. nov. – Myco-Bank MB 816977. Plates 18–19

Diagnosis: Differs mainly from the most resembling species *Annulohypoxylon nitens* by smaller *truncatum*-type ostiolar discs and larger ascospores averaging $9.5 \times 4.2 \mu m$.

Holotype: French West Indies: Guadeloupe: Sainte-Rose, Sofaïa, path to Saut des Trois Cornes, mesophilic rainforest, on corticated branch, 3 Sept. 2004, *leg*. C. Lechat, CLL 2304 (LIP).

Etymology: From Latin *splendens* = shiny, for the shiny surface of mature stromata.

Stromata effused-pulvinate to glomerate, with inconspicuous to most often strongly exposed perithecial contours, 1.5–5 mm long \times 1.5–2.6 mm wide \times 0.75–1.2 mm thick, irregularly coalescent into larger clusters; outermost coating dark brick (60, oac635), squamulose, fugacious, revealing a shiny black and smooth subsurface at maturity; texture carbonaceous, with subsurface composed of pale olivaceous yellow waxy granules appearing dull orange brown in mass, distinct from the underlying carbonaceous layer, releasing greenish olivaceous (90, oac859) KOH-extractable pigments within 1 min, becoming grey olivaceous (107, oac882) upon prolonged incubation; subperithecial tissue 0.2–0.6 mm thick, blackish. **Perithecia** subspherical, 0.5–0.6 mm diam. **Ostioles** conic-papillate, encircled with a flattened, *truncatum*-type disc 0.35–0.42 mm diam with sharp to notched edges.

Asci cylindrical, with eight uniseriate ascospores, fragmentary, originating from a thick, amorphous, yellowish subhymenium, 140–170 µm total length, the spore-bearing parts $70-81 \times 4.5-5.5$ µm, the stipes 55-97 µm long, without an apical apparatus, not bluing in Melzer's reagent nor in Lugol's solution. **Hamathecium** of dense paraphyses 3-4.5 µm wide, slightly tapering above asci, collapsed, ribbon-like. **Ascospores** $(8.4-)8.8-10.1(-10.6) \times (3.7-)3.9-4.5(-5)$ µm, Q=(1.9-)2.1-2.5(-2.6); N=60 (Me $=9.5\times4.2$ µm; Q=2.3), ellipsoid-inequilateral with most often narrowly rounded ends, brown, with a conspicuous straight germ slit almost spore-length; perispore dehiscent in 10% KOH, smooth, with a thickening on the dorsal side at ca. $\frac{1}{3}$ spore length; epispore smooth.

Asexual morph on the natural substrate: Scattered conidiophores present on young stromata and at their margins, originating from the red-brown outermost tissue; conidiophores upright, with a stout brown to dark brown septate axis 200–430 μ m high \times 7–9 μ m wide, apically branching, roughened; conidiogenous structure nodulisporium-like to periconiella-like, with conidiogenous cells 9–25 \times 3.5–4.5 μ m, finely roughened, brown to subhyaline, turning greenish in 3% KOH; conidia brown to pale brown, smooth, ellipsoid, 5–6 \times 2.7–3.2 μ m.

Known distribution: Guadeloupe.

Discussion: Any Annulohypoxylon collection featuring more or less shiny black stromata with olivaceous-green KOH-extractable pigments must be compared with A. nitens and its relatives. The taxon described above roughly resembles A. nitens in having stromata with exposed perithecial contours, a reddish brown outermost coating that gradually flakes off to reveal a shiny black subsurface and ostiolar discs 0.35-0.4 mm diam. However, a careful examination of the ostiolar discs formation shows that their dehiscence is of the truncatum-type, by crumbling of the disc from the centre outwardly, unlike that of typical A. nitens in which the dehiscence is of the bovei-type, lifted in one piece. Moreover, its ascospores are 8.8- 10×3.9 –4.5 µm, which is within the wide size range 6.5– 10×3 – 4.5 μm given by Ju & Rogers (1996) but above the narrower size range of 7–8 \times 3–3.5 µm retained in a narrower delimitation of A. nitens proposed in this paper. Based on these differences, the new taxon A. splendens is proposed. The apparent absence of an ascal apical apparatus bluing in Melzer's reagent is not considered diagnostic as it is reported by Ju & Rogers (1996) to occasionally

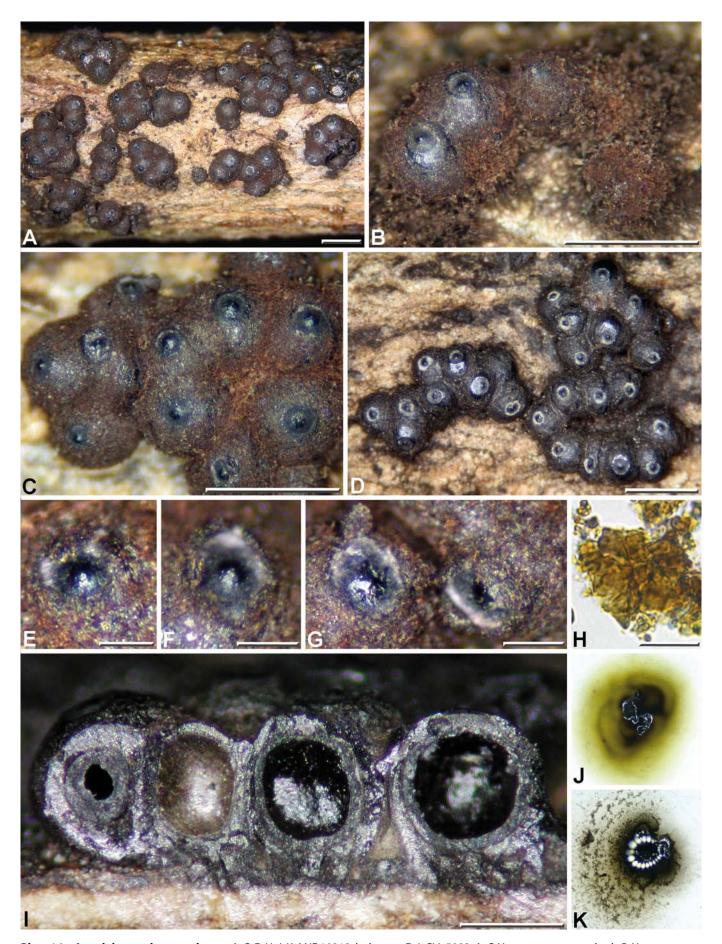


Plate 16 – **Annulohypoxylon ramulorum.** A-C, E, H, J, K: MJF 10312, holotype; D, I: CLL 5083. A, C: Young stromat on bark; B: Young stroma with red-brown woolly coating and tufts of asexual morph; D: Mature black stromata; E-G: Close up on ostioles with *truncatum*-type dehiscing discs; H: Olivaceous yellow to dull orange brown stromatal granules from subsurface, in water; I: Stroma in vertical section showing the perithecia and the weakly developed subperithecial tissue; J, K: KOH-extractable pigments after 1 min and 30 min incubation respectively. Scale bars: A-D=1 mm; E-G=0.2 mm; H=10 µm; I=0.5 mm.

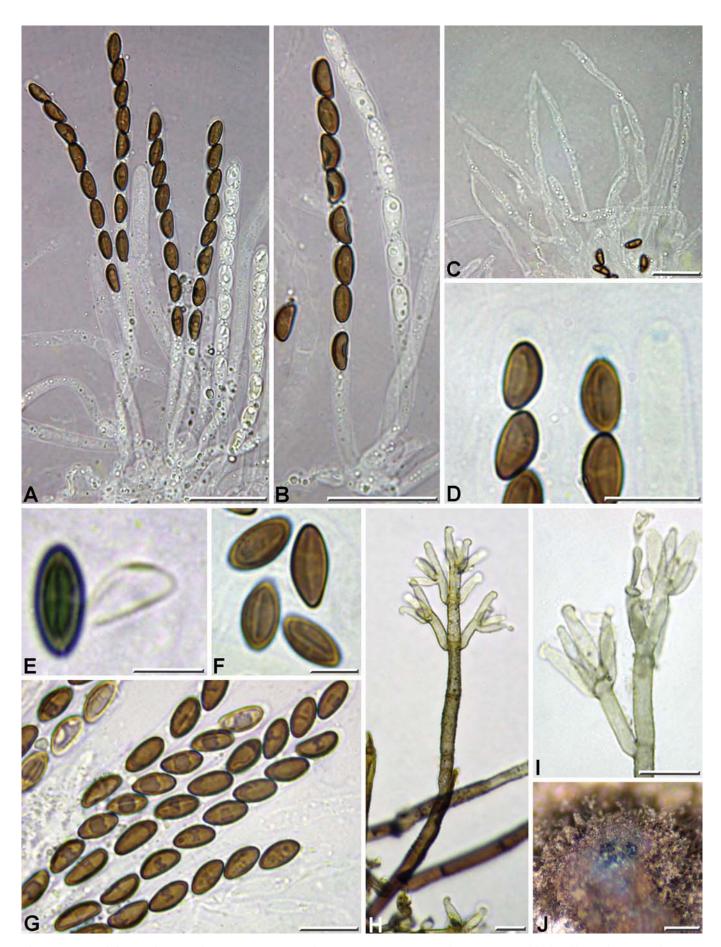


Plate 17 – Annulohypoxylon ramulorum MJF 10312, Holotype. A, B: Immature and mature asci in black Pelikan ink; C: Paraphyses, in black Pelikan ink; D: Ascal apical apparati faintly bluing in Melzer's reagent; E: Ascospore in 10% KOH with a dehiscent perispore; F: Ascospores in dorsal view showing the germ slit, in Melzer's reagent; G: Ascospores in 1% SDS; H: Conidiophore of the asexual morph, with nodulisporium-like branching pattern, in 3% KOH; I: Conidiogenous structure of the asexual morph, with periconiella-like branching pattern, in 3% KOH; J: Tufts of the asexual morph at the base of a primordium. Scale bars: $A-C=20~\mu m$; D, $G-I=10~\mu m$; E, $F=5~\mu m$; J=0.2~m m.

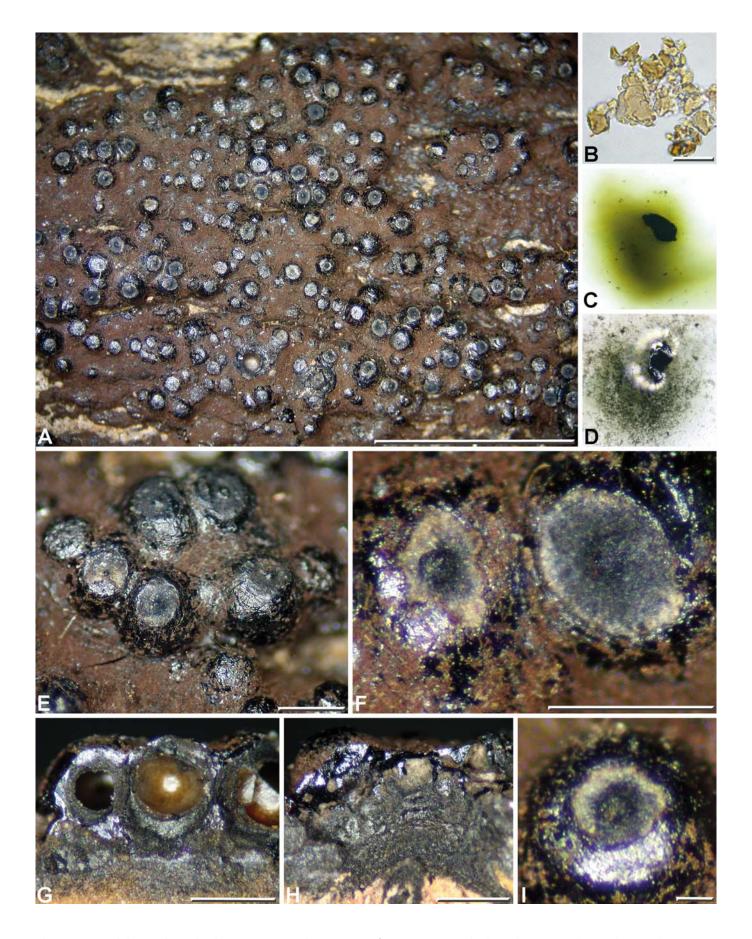


Plate 18 – **Annulohypoxylon splendens CLL 2304, Holotype.** A: Confluent stromata on bark; B: Olivaceous yellow and orange brown stromatal granules from subsurface, in water; C, D: KOH-extractable pigments after 1 min and 30 min incubation respectively; E: Small glomerate stroma showing the reddish brown coating and the shiny black subsurface; F: Two adjacent *truncatum*-type ostioles in close-up, one with ostiolar disc in the process of dehiscence (left) next to a fully dehisced ostiolar disc (right); G: Effused stroma in section; H: Pulvinate stroma in section (broken) showing the shiny crust and the thick subperithecial tissue; I: Dehiscing ostiolar disc in close-up. Scale bars: A = 5 mm; $B = 10 \mu m$; E - H = 0.5 mm; $I = 100 \mu m$.

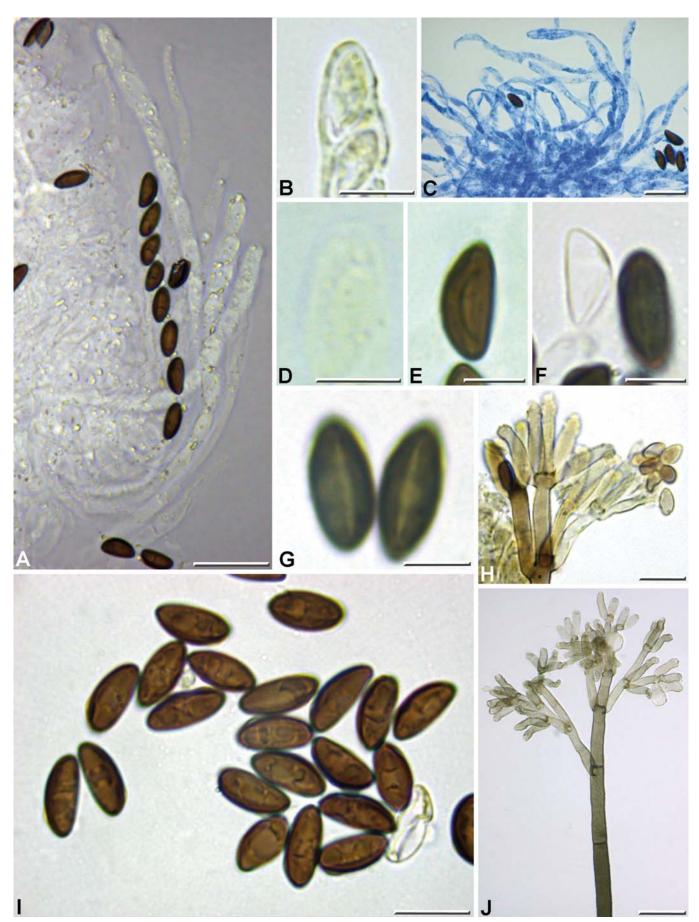


Plate 19 – Annulohypoxylon splendens CLL 2304, Holotype. A: Immature and mature asci in black Pelikan ink; B: Ascal apex showing the lack of apical apparatus and the lack of amyloid reaction, in Lugol's solution; C: Collapsed, ribbon-like paraphyses, in blue Waterman ink diluted in 1% SDS; D, E: Ascal apices showing the lack of apical apparatus and the lack of amyloid reaction, in Melzer's reagent; F: Ascospore in 10% KOH with a dehiscent perispore; G: Ascospores in dorsal view showing the germ slit, in 10% KOH; H: Conidiogenous structure of the asexual morph, with periconiella-like branching pattern and conidia, in 3% KOH; I: Ascospores in 1 % SDS; J: Conidiophore of the asexual morph, with nodulisporium-like branching pattern, in 3% KOH. Scale bars: A, C, J = 20 μ m; B, D-G = 5 μ m; H, I = 10 μ m.

occur in *A. nitens* and likewise observed in some of the collections regarded here as *A. nitens*.

Annulohypoxylon splendens is primarily segregated from A. caravellenese (this paper) by ostiolar discs of the bovei-type and smaller ascospores $7.6-8.6\times3.2-3.7~\mu m$ for the latter.

Annulohypoxylon subnitens, known from French Guiana, was segregated from A. nitens by inconspicuous pale olivaceous grey KOH-extractable pigments and slightly smaller ostiolar discs (FOURNIER & LECHAT, 2016), which also makes it different from A. splendens. Ascospores of A. subnitens likewise average clearly smaller than those of A. splendens respectively 7–7.7 \times 2.6–3.2 μm vs. 8.8–10 \times 3.9–4.5 μm .

Annulohypoxylon splendens is unfortunately known from only one collection from Guadeloupe, thus its possible intraspecific variations are unknown.

Annulohypoxylon stygium (Lév.) Y.-M. Ju, J.D. Rogers & H.-M. Hsieh, *Mycologia*, 97 (4): 861 (2005). Plates 20–21

Stromata effused, effused-pulvinate or perithecioid, 3–62 mm long \times 2–18 mm wide \times 0.5–0.75 mm thick, with most often inconspicuous to faintly exposed perithecial contours, rarely strongly exposed to perithecioid and then often more or less shiny black; surface with a fugacious dark brick (60, oac637) to dark vinaceous (82, oac523), vinaceous buff (86, oac662) or greenish-olivaceous (90, oac860) pruinose outermost coating, turning blackish with age; texture weakly carbonaceous, with subsurface composed of olivaceous buff (89, oac891) granules yielding dark herbage green (69, oac54) KOH-extractable pigments within 1 min incubation, darkening upon prolonged incubation; subperithecial tissue inconspicuous to 0.2 mm thick, blackish brown. **Perithecia** subspherical 0.3–0.35 mm diam to tubular, $0.5-0.6 \times 0.2-0.3$ mm. **Ostioles** broadly conic-papillate, encircled with a shiny black, flattened bovei-type disc 0.17-0.2(- 0.22) mm diam with most often irregular rims, at times conspicuously overlain by white granular tissue.

Asci cylindrical, with eight uniseriate ascospores, $60-80~\mu m$ total length, the spore-bearing parts $45-48\times3.5~\mu m$, the stipes $15-36~\mu m$ long, with a minute discoid apical apparatus bluing in Melzer's reagent. **Paraphyses** evanescent. **Ascospores** $(4.8-)5.2-5.8(-6)\times(2.1-)2.3-2.7(-2.8)~\mu m$, Q=(1.9-)2-2.3(-2.7); $N=60~(Me=5.5\times2.5~\mu m$; Q=2.1), ellipsoid-inequilateral with narrowly to broadly rounded ends, pale brown to medium brown, with an inconspicuous straight germ slit almost spore-length on the ventral side; perispore dehiscent in 10%~KOH, smooth, with a thickening on the ventral side at ca. $\frac{1}{3}$ spore length; epispore smooth.

Asexual morph on the natural substrate not seen. Asexual morph in culture on OA periconiella-like, based on material from Taiwan (Ju & ROGERS, 1996).

Specimens examined: French West Indies: Guadeloupe: Basse-Terre, Petit-Bourg, forest track of Jules, hygrophilic rainforest, on dead corticated twig, 1 Sept. 2004, leg. C. Lechat, CLL 2240 (LIP). FRENCH WEST INDIES: MARTINIQUE: Saint-Esprit, Morne David, mesophilic rainforest, on corticated branch, 23 Aug. 2004, leg. C. Lechat, CLL 2029 (LIP); Saint-Esprit, Bois La Charles, mesophilic rainforest, on dead corticated branch, soc. Hypoxylon monticulosum, 24 Aug. 2004, leg. C. Lechat, CLL 2064 (LIP); ibid., 25 Aug. 2004, leg. C. Lechat, CLL 2095 (LIP); Case-Pilote, Crête Jean-Louis, mesophilic rainforest, on corticated branch, 27 Aug. 2004, leg. C. Lechat, CLL 2162 (LIP); ibid., on corticated branch, 27 Aug. 2004, leg. C. Lechat, CLL 2175 (LIP); ibid., leg. C. Lechat, CLL 2177; Prêcheur, Anse-Couleuvre, mesophilic rainforest, on corticated branch, 23 Aug. 2007, leg. J. Fournier, MJF 07051 (LIP); Morne-Rouge, forest track of La Propreté, hygrophilic rainforest, on corticated branch, 24 Aug. 2007, leg. C. Lechat, MJF 07062 (LIP); Saint-Joseph, Coeur-Bouliki forest, hygrophilic rainforest, on dead corticated branchlets of Mahogany (Swietenia macrophylla King, Meliaceae), 26 Aug. 2007, leg. J. Fournier, MJF 07114 (LIP); ibid., MJF 07115 (LIP); Case-Pilote, trail to Plateau Concorde, hygrophilic rainforest, on dead decorticated branch, soc. Cosmospora sp., 27 Aug. 2010, leg. C. Lechat, CLLMAR 073 (LIP); Fort-de-France, Absalon, trail to Plateau-Michel, hygrophilic rainforest, on dead decorticated branchlet, 7 Aug. 2013, leg. J. Fournier, MJF 13053 (LIP); Saint-Esprit, Bois-La-Charles, mesophilic rainforest, on corticated branch, 20 Aug. 2013, leg. J. Fournier, MJF 13340 (LIP); Morne-Rouge, forest track of La Propreté, hygrophilic rainforest, on corticated branch of Mahogany (Swietenia macrophylla King, Meliaceae), 6 Jun. 2014, leg. J. Fournier, MJF 14061 (LIP); ibid., MJF 14062 (LIP); Fort-de-France, forest track of Fond-Baron, hygrophilic rainforest, on dead corticated branch of Mahogany (Swietenia macrophylla King, Meliaceae), 14 Jun. 2014, leg. J. Fournier, MJF 14152 (LIP); ibid., MJF 14159 (LIP); Schoelcher, Rivière Case-Navire, mesophilic rainforest, on corticated branchlet, 12 Jun. 2015, leg. J. Fournier, MJF 15025 (LIP); Morne-Rouge, forest track of La Propreté, hygrophilic rainforest, on corticated branch of Mahogany (Swietenia macrophylla King, Meliaceae), 23 Jun. 2015, leg. J. Fournier, MJF 15188 (LIP); Fort-de-France, Absalon, trail to Plateau-Michel, hygrophilic rainforest, dead corticated branch of Mahogany (Swietenia macrophylla King, Meliaceae), 15 Jun. 2015, leg. J. Fournier, MJF 15090 (LIP).

Known distribution: Pantropical.

Discussion: Tropical Annulohypoxylon spp. with small ostiolar discs < 0.2 mm diam and small perithecia, dark green KOH-extractable pigments and small ascospores $5-6 \times 2-3 \mu m$ with an inconspicuous germ slit on the ventral side and a perispore dehiscent in 10% KOH are readily identified as A. stygium. This species is very common in the tropics in various habitats and exhibits a wide variation range as to stromatal surface colour, varying from reddish brown to vinaceous, pale brown or olivaceous. Annulohypoxylon atroroseum was segregated from A. stygium by stromatal surface with pink, vinaceous grey to vinaceous brown tones and a different asexual morph (Rogers, 1981, as Hypoxylon atroroseum). Annulohypoxylon stygium is regarded as having typically reddish brown stromata and it is still unknown whether stromata with vinaceous buff or greenish-olivaceous tones like those we encountered in Martinique and illustrate here represent cryptic species or are just the result from environmental conditions. In absence of clear correlation between stromatal colour and other deviating characters, the further splitting of A. stygium does not appear justified so far.

The *bovei*-type dehiscence of the ostiolar discs of *A. stygium* was reported for the first time by FOURNIER & LECHAT (2016) based on material collected in French Guiana. Despite the important number of material collected during this survey and studied subsequently, we only observed twice the *bovei*-type dehiscence of the ostiolar discs in the barely mature collections MJF 14061 and MJF 14062. We frequently observed wider ostiolar discs up to 0.3 mm diam when stromata are perithecioid and separated, while they do not usually exceed 0.2 mm diam in typically effused stromata. This should be taken into account when identifying *A. stygium* with such a deviating morphology.

The observations on the ecology of *A. stygium* made during this survey are consistent with those reported by FOURNIER & LECHAT (2016) for French Guiana.

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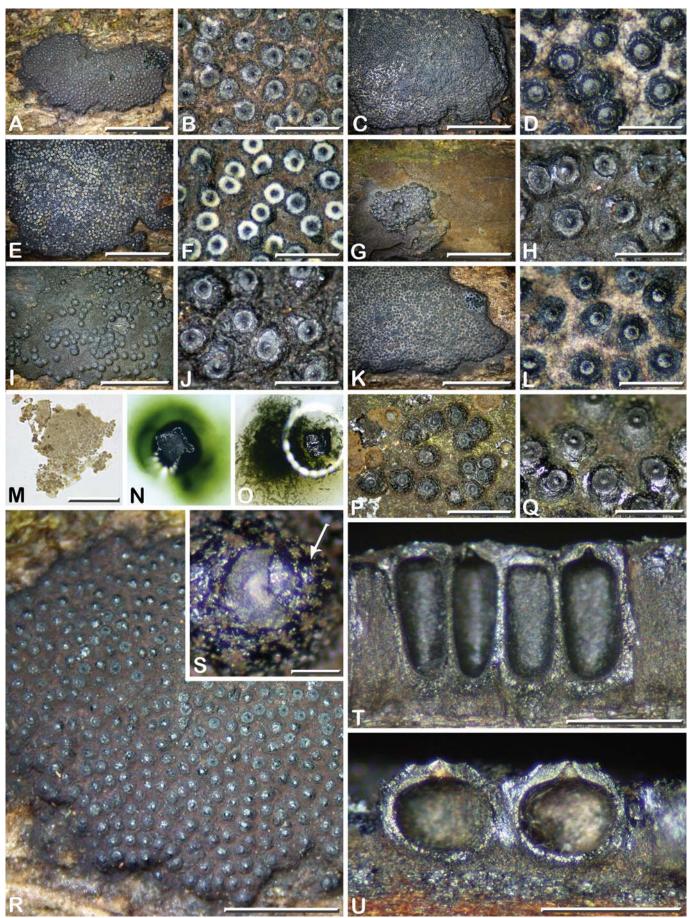


Plate 20 – Annulohypoxylon stygium. A, B, R: MJF 07115; C, D: CLL 2177; E, F, T: MJF 07051; G, H, M-Q, U: MJF 15188; I, J: MJF 14152; K, L: CLL 2029; S: MJF 14062. A, C, E, G, I, K, P, R: Effused, effused-pulvinate and perithecioid stromata in surface view; B, D, F, H, J, L, Q: Close-ups on stromatal surface showing the outermost coating, the subsurface and the ostiolar discs; M: Stromatal waxy granules from subsurface, in water; N, O: KOH-extractable pigments after 1 min and 30 min incubation respectively; S: *bovei*-type dehiscence of an ostiolar disc (arrow); T, U: Stromata in vertical section showing the tubular or subspherical perithecia encased by carbonaceous tissue. Scale bars: A, C, E, G, I, K, P, T, U = 5 mm; B, D, F, H, J, L, Q = 0.5 mm; M = 10 μ m; R = 2 mm; S = 100 μ m.

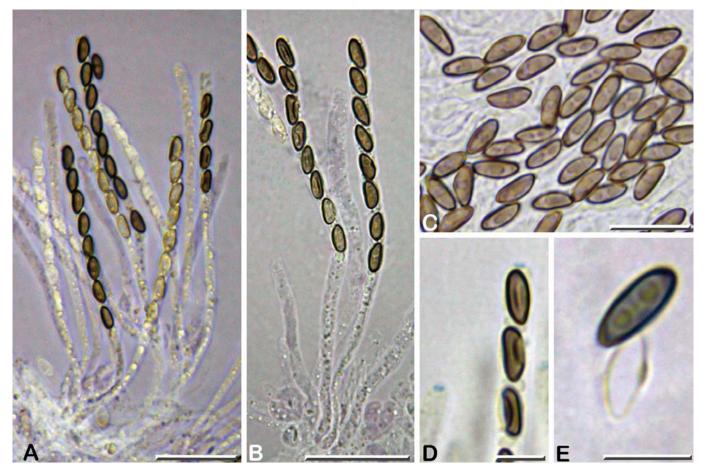


Plate 21 – **Annulohypoxylon stygium MJF 15188.** A, B: Short- and long-stipitate asci in black Pelikan ink; C: Ascospores in 1% SDS; D: Apical apparati in Melzer's reagent; E: Ascospore in 10% KOH showing the dehiscent perispore with a thickening on the ventral side. Scale bars: A, B = 20 μ m; C = 10 μ m; D, E = 5 μ m.

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