

The Genus *Amyloflagellula* in West-Africa (Basidiomycetes, Cyphellaceae)

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Summary. — Two new species of the genus *Amyloflagellula* are described: *A. inflata* and *A. verrucosa*. The similarities and differences to the species *A. pulchra*, *A. pseudoarachnoidea* and "*Cyphella*" *byssacea* are discussed. The two new species are the first find of the genus in Africa. The mycelia are described.

The genus *Amyloflagellula* was proposed by SINGER (1966: 14), comprising two species, the former *Cyphella pulchra* BERK. & BR. (type-species) and *Marasmius pseudoarachnoideus* DENNIS.

Previously *Cyphella pulchra* had been placed in two other genera, *Lachnella* (SINGER, 1961: 60) and *Marasmius* (PETCH, 1924: 19), until SINGER (1966: 14) set up a separate genus for this relationship.

Species of the genus *Amyloflagellula* are only known from Tropical America and Asia (SINGER, 1975: 372) so far.

Amyloflagellula pulchra (BERK. & BR.) SING. is described from Ceylon (type-specimen) and known from some other Asian countries as well: India (PETCH, 1924: 72) and Japan (KOBAYASI, 1971: 78). The other species previously known in the genus *Amyloflagellula* — *A. pseudoarachnoidea* (DENNIS) SING. — was described by DENNIS (1951: 425) from Neotropics (Trinidad).

There are some further species, which probably belong to the genus *Amyloflagellula*, but of these not a single species grows in Africa. All differ markedly from all species characterized in this paper.

Cyphella reniformis PAT. (1897: 115) is probably nearest to *C. byssacea* P. HENN. & NYM. (HENNINGS, 1900: 7) and is perhaps conspecific (AGERER, unpublished); both are described from Java. DONK (1959: 43) already believed that this two species belong to this relationship.

DONK (1959: 43) also suggested that *C. juruensis* P. HENN. (1904:

¹⁾ With collaboration of Miss P. LANQUETIN for the study of cultures.

173), known from the Amazonian-region, is related with *C. pulchra* and therefore it would belong to the genus *Amyloflagellula*, too. According to the original description, the basidia and spores are very small. This species is therefore quite different from all others in question.

Other species probably belonging to the genus *Amyloflagellula* (cf. DONK 1959: "was described among these fungi", i. e. *Cyphella-pulchra*-relationship) are *Marasmius cyphella* DENNIS & REID (1957: 288) and *Marasmius scandens* (MASSEE) DENNIS & REID (1957: 289) — the latter probably a related species, too — possess ventral-lateralstipes and are therefore different from all species described in this paper.

COOKE (1961: 45) described *Glabrocyphella ailanthis* W. B. COOKE from the United States as a new species. It is said to grow "on a common subiculum composed of hyaline, branched hyphae which extend half way up the cups". These hyphae are probably similar to the flagelliform elements of the species of *Amyloflagellula*. SINGER (1975: 330; 1978: 271), however supposed that *G. ailanthis* might belong to *Calyptrella* subg. *Syncyphella* SING. The small spores and basidia distinguish this species from all others discussed in this paper of the genus *Amyloflagellula*.

Key to the Species of *Amyloflagellula*

1. Fruit-bodies solitary on prominent, white rizomorphs. *A. pseudoarachnoidea*
- 1*. Fruit-bodies \pm densely crowded on a \pm dense subiculum
2. Margin of fruit-bodies with warty surface hairs
3. Fruit-bodies symmetric. 2. *A. verrucosa*
- 3*. Fruit-bodies asymmetrical, with a lateral point of attachment. "Cyphella" *byssacea*
- 2*. Margin of fruit-bodies with flagella-bearing surface hairs
4. Basis of fruit-bodies with many thickwalled hyphae, without inflated ones *A. pulchra*
- 4*. Basis of fruit-bodies with few thick-walled and with many inflated hyphae 1. *A. inflata*

1. *Amyloflagellula inflata* AGERER & BOIDIN, spec. nov.²⁾ — Fig. 1, 2

Typus: GABON: Forêt de la Mondah, km 26.5 N. Libreville, GILLES 1439, 11. 2. 1979 (Holotypus LY, 9157; Isotypus herb. R. AGERER, TUB).

Differt ab *Amyloflagellula verrucosa* cupulis majoribus interdum asymmetricis, hymenio interdum accliviatis exiguis, hyphis basibus cupularum inflatis, pilis externis marginis cupulae flagelligeribus non verrucosis, prope basem constrictum cupularum cellulis laevis, subicula hyphis paulum crassi-

²⁾ Etymology: inflata (lat.) = inflated; the hyphae of the trama in the stipe like base are inflated.

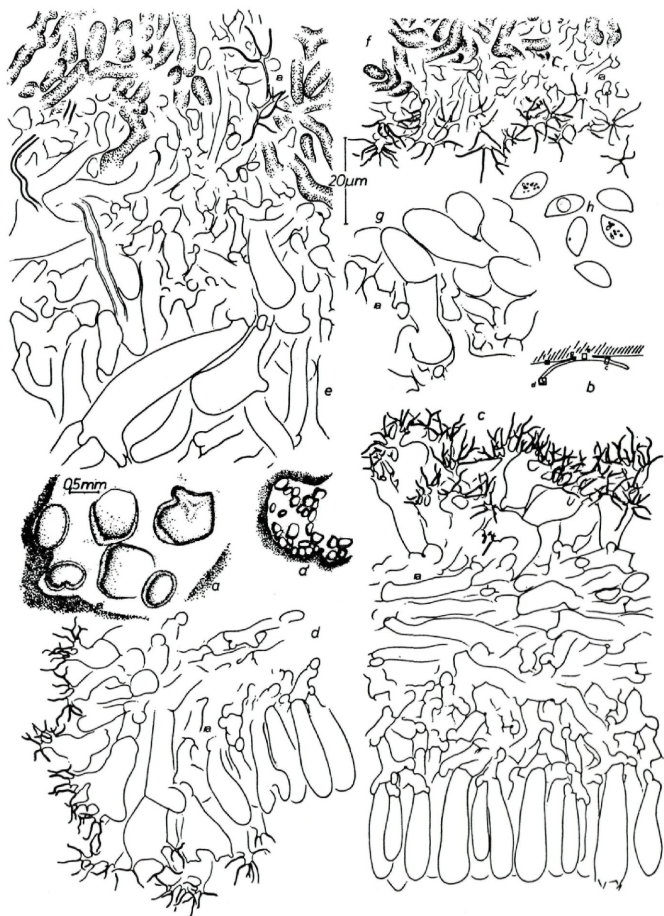


Fig. 1. *Amyloflagellula inflata*: a—a'. Habit of fruit-bodies. — Section through a fruit-body, general view. c—e. Sections through a fruit-body in detail: — c. from the middle; — d. edge; — e. from the stipe-like base. — f. Section through the subiculum. — g. Detail next the stipe-like base. — h. Spores. (All figs. from Holotype, LY, 9157)

tunicatis, hyphis incrustatis absentibus; — differt ab *A. pulchra* cupulis paulum asymmetricis, subiculo hyphis crassitunicatis absentibus; — differt ab *A. pseudoarachnoidea* rhizomorphis absentibus, hymenio non lamelliformibus; — differt ab "*Cyphella*" byssacea cupulis non laterale affixis, pili externi marginis cupulae flagelligeribus non verrucosis.

Cupulae non profunde patinatae, symmetricae vel leviter asymmetricae, non stipitatae, dense aggregatae, in subiculo cretaceopulveraceo sitae, usque ad 2 mm in diametro, albae, hymenio laeve vel acclivatis exiguis luteoloque instructae, margines cupularum et extrinsecus pruinosa. Hyphae subiculi plus minusve tenuitunicatae, flagellis ramosis tenuissimis usque ad 0.5 μ m in diametro exorientibus, dextrinoideis, hyphae incrustatae deficientes. Pili externi hyalini, clavati, pro parte lobati, (4) 5–10 (12) \times 10–20 μ m, dextrinoidei, flagellis ramosis tenuissimis dextrinoidisque, pili externi plus minusve tenuitunicati, prope bases cupularum pili externi plus minusve clavati flagella deficientes. Hyphae tramae 3–5 (7) μ m in diametro, fibuligera, in subhymenio 1.5–3.5 μ m; bases cupularum hyphis inflatis tenuitunicatis et paucis hyphis tunicis ad 1 μ m crassis. Hymenium basidiolis subfusiformibus; basidia 26–30 (32) \times 6.5–8 μ m, leviter suburniformia, 4-sterigmatica, fibuligera. Sporae asymmetrici-ellipticae vel asymmetrici-ovoideae vel subnaviculares, tenuitunicatae, laeves, hyalinae, 8–9 (9.5) \times 4–5 μ m, ca. 8.5 μ m longae, proportio sporarum 1.9–2.0, nec amyloideae nec dextrinoideae.

Fruit-bodies densely crowded, shallow, symmetrically or sometimes asymmetrically cupshaped, stipeless, white, sitting in a greyish white powdery subiculum, up to 3 mm in diam., hymenium smooth or seldom with very few elevations, yellowish, margin of the fruit-body pruinose. Subiculum composed of more or less thin-walled hyphae with very thin flagelliform ramified appendices up to 0.5 μ m in diam., appendices strongly dextrinoid; without encrusted hyphae. Surface hairs of the cups hyaline, clavate, partly lobed, (4) 5–10 (12) \times 10–20 μ m, with distinctly dextrinoid, flagelliform, ramified appendices, surface hairs with more or less thin walls. Just beside the constricted base of the fruit-body surface with more or less clavate cells without appendices. Hyphae of trama 3–5 (7) μ m in diam., clamped; in the subhymenium 1.5–3.5 μ m; base of fruit-body with few thick-walled hyphae with walls up to 1 μ m thick. Hymenium with slightly fusiform basidiols; basidia (22) 26–30 (32) \times (5) 6.5–8 μ m, slightly suburniform, with four sterigmata, clamped at the base. Spores asymmetrically-ellipsoid to asymmetrically-ovoid, or slightly naviculate, thin-walled, smooth, hyaline, 8–9 (9.5) \times 4–5 (5.5) μ m (on the average 8.0–8.8 μ m long, with spore factor 1.8–2.0), neither amyloid nor dextrinoid.

Substrate. — Twigs and leaves.

Further specimens examined: Guadeloupe: Route de grosse montagne Parc National, sur branchettes et feuilles mortes suspendues, BORDIN, 5. 10. 1976 (LY, 8143). — Gabon: Makokou, sur liane en l'air, BORDIN, 10. 5. 1976 (LY, 7793).

Discussion: *Amyloflagellula inflata* is very conspicuous because of the inflated hyphae in the base of the fruit-body; all other known

species lack them. There are only a few thin-walled hyphae in the very base of *A. inflata*, whereas the fruit-bodies of *A. pulchra* are seated in a more or less dense layer of very thick-walled hyphae (AGERER, unpublished). Another difference between these two species is the shape of the fruit-bodies. Whereas the fruit-bodies in *A. pulchra* are asymmetric and look like an ear, they are in *A. inflata* usually symmetrical and only sometimes of slightly asymmetrical shape, and other specimens of *A. pulchra* are said to have lamellate fruit-bodies (PETCH, 1924: 21).

Amyloflagellula pseudoarachnoidea possesses distinct white rhizomorphs bearing solitary fruit-bodies with true gills and asymmetric stipes (AGERER, unpublished); it is a typical agaricaceous fungus, whereas *A. inflata* and *A. verrucosa* are cyphellaceous, seated in a white downwards facing subiculum.

There are similarly clear differences between *A. inflata* and *A. verrucosa*. In contrast to *A. inflata*, which produces flagella-bearing surface hairs on the margin of the cupshaped fruit-body, *A. verrucosa* possesses warty surface hairs like "*Cyphella*" *byssacea*. Besides the differences in the stipelike base of the fruit-body — inflated hyphae occur only in *A. inflata* —, the subicula of the two species differ. In *A. inflata* the subiculum is built up by more or less thin-walled hyphae with flagella-like appendices; in *A. verrucosa* the subiculum is composed of three types of hyphae: (1) more or less thin-walled with flagella-like appendices and (2) thick-walled hyphae which are often followed by the thin-walled ones, and (3) those bearing hyaline, very small crystals. Next to the constricted bases of the fruit-bodies of *A. verrucosa* there are warty cells too, in *A. inflata* the cells lack any exrescences.

There are marked differences between *A. inflata* and "*Cyphella*" *byssacea* too; see above.

Cultural characters of *Amyloflagellula inflata* (LY, 9157):

Spores. — Uninucleate.

Monosporous cultures. — With spores received at Lyon 5 days after collection, a dispersion allowed to obtain only two germinations after 12 days. These monosporous cultures show thin-walled, hyphae without clamps and flagelliform dextrinoid very thin hair-like elements. Hyphae are composed of regularly uninucleate articles.

Polysporous cultures. — Obtained by means of cutting.

Growth: very slow (Petri dishes are not covered after six weeks).

Aspect: Remarkably constant. Margin: Irregular, lobed.

After six weeks, aerial whitish mycelium is scarce, it shows delicate nerves more or less radiating on the medium tinged pale orange brown. Slightly more abundant on inoculum and its surroundings, it makes a continuous surface tinged alutaceous (Munsell³⁾

³⁾ Munsel Color Company (1954). Munsell book of color. Baltimore.

2,5 Y 8/4), jaune de Naples (M 2, 5 Y 8, 5) to (M 10 YR 8/4 8/6) on inoculum. Weak fruity odour (Peach-Apricot). Reverse seems pale orange brown, actually this coloration is limited to a thin layer of medium, 0.5 μm thick, located at 1 mm of the surface and there is no diffusion in the whole of the medium.

Microscopical characters:

Aerial mycelium: 1) It is constituted by aerial regular hyphae, $\times 1-2.5$ (3) μm , thin walled, homogeneous contents and constant

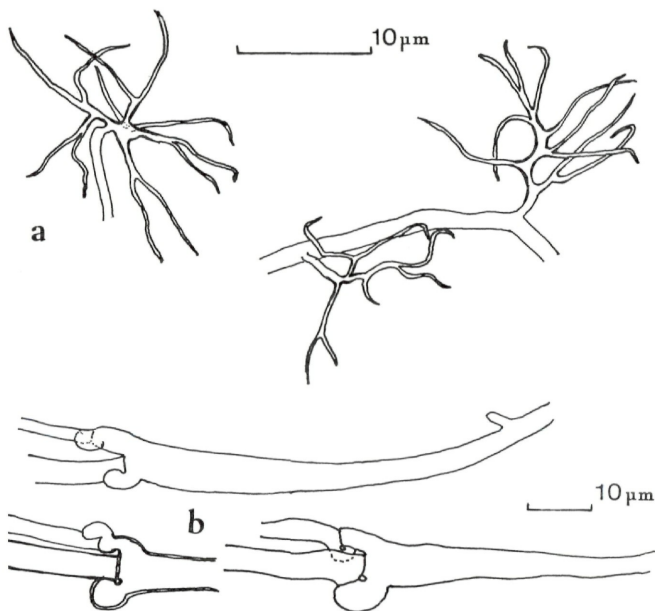


Fig. 2. *Amyloflagellula inflata* (LY, 9157, in culture): a. Flagelliform, dextrinoid elements. — b. Inflated articles in submerged mycelium

clamp-connections. Just underneath, wider hyphae, $\times 4-5$ μm , rather irregular, often with small clamp-connections reminding hyphae of *Hyphoderma*. Some of these hyphae have orange yellowish contents that appear sometimes granular, more or less divided or crystallized. — 2) Numerous congophilous, dextrinoid, flagelliform elements with capillary hair-like branches measuring at most $5-10 \times 0.5$ μm (cf.

flagelligeribus, subiculo hyphis crassitunicatis et hyphis incrustatis instructo; — differt ab *A. pulchra* cupulis symmetricis, pilis externis marginis cupulae verrucosis non flagelligeribus; — differt ab *A. pseudoarachnoidea* rhizomorphis absentibus, hymenio laeve; — differt ab “*Cyphella*” byssaceae cupulis minoribus, symmetricis, non lamelligeribus.

Cupulae non profunde patinaceae, symmetricae, non stipitatae, dense aggregatae, in subiculo albo-floccoso sitae, usque ad 0.3 mm in diametro, albae hymenio laeve luteoloque, margines cupularum et extrinsecus pruinosa. Hyphae subiculi tribus formis: forma una plus minusve tenuitunicata, flagellis ramosis tenuissimis dextrinoideis, usque ad 0.5 μ m in diametro ornata — forma alia 2.5–4 μ m in diametro, tunicis usque ad 1.5 μ m crassis, dextrinoideis, in 10% KOH subturgescentibus, secundarie septata (forma prima exoriens formis secundis) — forma tertia 1.5–3 μ m in diametro, tunicis 0.5–1 μ m crassis, non dextrinoideis, incrustata hyalinis, minute granularibus vel acicularibus crystallis in HCl rapide solventibus. Pili externi hyalini, subdextrinoidei, clavati, usque ad 10–15 \times 7–10 μ m, brevibus verrucis dextrinoideis; pili externi marginis tenuitunicati alii crassitunicati, tunicis usque ad 1 μ m crassis. Hyphae tramae (1.5) 2–3.5 (4) μ m in diametro, fibuligerae; bases cupularum hyphis 2.5–5 (6) μ m in diametro, crassitunicatis formatae. Hymenium basidiolis subfusiformibus; basidia 20–24 (28) \times (6.5) 7.5–8.5 (9) μ m, leviter suburniformia, 4-sterigmatica, fibuligera. Sporae asymmetrici-ellipticae vel subnaviculares, tenuitunicatae, laeves, hyalinae, (8.5) 9–10 \times 4–4.5 (5) μ m, ca. 9.3 μ m longae, proportio sporarum 2.2, nec amyloideae nec dextrinoideae nec cyanophilae.

Fruit-bodies densely crowded, shallow, symmetrically cupshaped, stipeless, sitting in a white, tufted subiculum, up to 0.3 mm in diam., white, hymenium smooth and yellowish, margin of the fruit-body pruinose. Subiculum existing of “three types” of hyphae: (1) with more or less thin walls, with very thin, flagelliform, ramified appendices up to 0.5 μ m in diam., appendices strongly dextrinoid; (2) dextrinoid, 2.5–4 μ m in diam., with walls up to 1.5 μ m thick, slightly swelling in 10% KOH, secondarily septate, (“first type” originates probably mostly from the “second type”); (3) not dextrinoid, 1.5–3 μ m in diam., with walls 0.5–1 μ m thick, encrusted with small, hyaline, granular or acicular crystals, which dissolve rapidly in hydrochloric acid. Surface hairs hyaline, slightly dextrinoid, clavate, up to 10–15 \times 7–10 μ m, with short, wart-like, distinctly dextrinoid appendices; with thin walls near the margin of the fruit-body otherwise slightly thick-walled with walls up to 1 μ m thick. Hyphae of trama (1.5) 2–3.5 (4) μ m in diam., clamped, base of fruit-bodies with hyphae 2.5–5 (6) μ m in diam., with thick walls. Hymenium with slightly fusiform basidiols; basidia (18) 20–24 (28) \times (6.5) 7.5–8.5 (9) μ m, slightly suburniform, with four sterigmata, clamped at base. Spores asymmetrically ellipsoid to slightly naviculate, thin-walled, smooth, hyaline, (8.5) 9–10 (10.5) \times (3.5) 4–4.5 (5) μ m (on the average 8.6–9.4 μ m long, with sporefactor 2.1–2.4), neither amyloid nor dextrinoid nor cyanophil.

Substrate: dead or living stems and twigs.

Further specimens examined: Gabon: Makokou, Com-

bretum mort en l'air, BOLDIN, 10. 5. 1976 (LY, 7795). — Makokou, Station CNRS, sur branchettes, BOLDIN, 4. 5. 1976 (LY, 7733). — Republic of Central Africa: La Maboké, petites branches à nameaux opposés, BOLDIN, 1. 5. 1965 (LY, 5288). — La Maboké, province de la Lobaye pres de Mbaiki, BOLDIN, 6. 5. 1965 (LY, 5331). — La Maboké (Sud), sur branchette en l'air, BOLDIN, 13. 5. 1965 (LY,

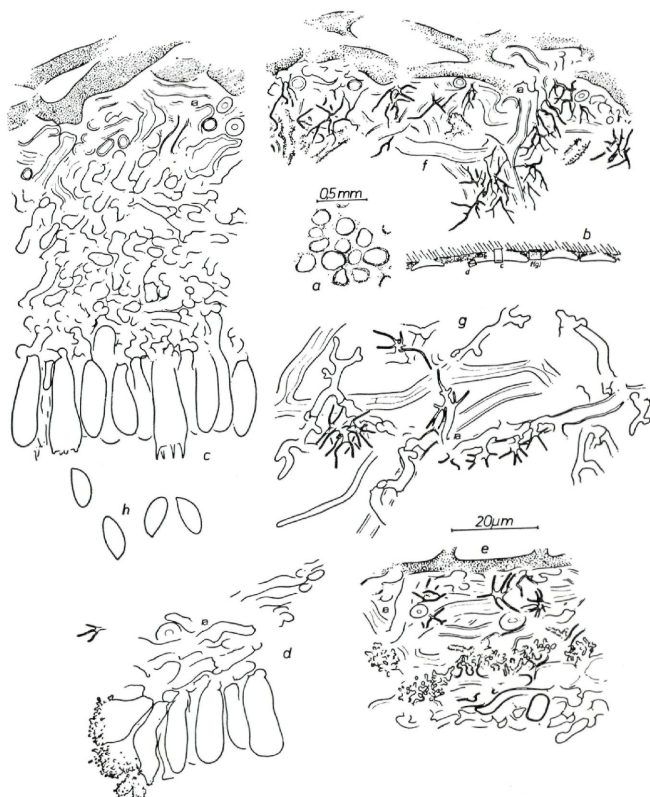


Fig. 3. *Amyloflagellula verrucosa*: a. Habit of fruit-bodies. — b. Section through fruit-bodies, general view. — c — e. Sections through fruit-bodies in detail: — c. from the centre; — d. edge; — e. next the stipelike base. — f. Section through the subiculum. — g. view of the subiculum. — h. Spores. (All figs. from Holotype, LY, 7911 bis)

5424). — La Maboké, sur liane, *Cissus* spec., BOLDIN, 10. 5. 1969 (LY, 5384).

Discussion: The differences to *Amyloflagellula inflata* are discussed in the description of *A. inflata*. *A. verrucosa* is devoid of rhizomorphs and is therefore quite different from *A. pseudoarachnoidea*. Furthermore, *A. pseudoarachnoidea* possesses true gills, and the fruit-bodies are solitary. *A. verrucosa* is in contrast to *A. pulchra* of symmetrical shape and the surface hairs on the margin of the fruit-bodies are warty in *A. verrucosa* but they bear flagella-like appendices in *A. pulchra*. Thick-walled hyphae exist only in the very base of *A. verrucosa*, whereas *A. pulchra* possesses a more or less dense layer of thick-walled hyphae. "*Cyphella*" *byssacea* produces fruit-bodies, which are laterally attached and are gilled at least in the final stages of development.

Cultural characters of *Amyloflagellula verrucosa* (LY, 7911 bis):

Spores. — Uninucleate. Though we have tried to get germinations with six different specimens, we obtained no results. But it should be noticed that spore prints were received at Lyon after 6 or 9 days of travel. Among cultures made immediately in Africa one cutting and one polysporous culture (LY, 7911 bis) grew.

Polysporous cultures.

Growth: Moderately rapid (Petri dishes covered in 4 to 5 weeks).

Aspect: Regular margin. After six weeks, white aerial mycelium does not cover entirely medium; it forms a scant mycelium tinged honey (M 2.5 Y 7/6) or very small whitish granulations. Mycelium is denser around inoculum and its surroundings and tinged alutaceous (M 2.5 Y 8/4, 8/6). Odour: none. Reverse regularly tinged honey (M 2.5 Y 7/6).

Microscopical characters:

Aerial mycelium is composed of regular hyphae, 1.5—3 (4) μm wide, thin-walled, with constant clamps and swelling hyphae with restricted septa and rather small clamps. Swellings, 6—9 (12) μm wide, show often thick walls.

All hyphae have guttulate or granular contents. Only in a few cultures, incrustated hyphae with slightly bi-refringent crystals have been observed. It should be noticed that we never saw dextrinoid flagelliform hyphae in the cultures.

Submerged mycelium: Irregular, swelling hyphae, 2—5 (6) μm wide, recalling those of *Hyphoderma* with guttulate contents, restricted septa and small, constant clamps. Their wall is thin or locally thickened, Swellings, 8—10 (12) μm wide, have also a thin wall or sometimes a conspicuously thickened wall (up to 1 μm). Besides we

observed locally small, very dense regions composed of narrow ramifications, $\times 1-1.2 \mu\text{m}$, thinwalled with homogeneous contents, very intertwined making a sort of dense tissue.

Cytology: Hyphae are composed of regularly binucleate clamped articles.

Oxidases: gallic ac.: —; O

guaiacol: +++ tr.

p. cresol: —

tyrosine: —, tr.

CODE: 2a—3c—(9)—26—32—36—39—44 or 45—54—64.

Remarks: The mycelium of *A. verrucosa* differs from that of *A. inflata* by its more rapid growth. Moreover it does not show flagelliform, dextrinoid hyphae which we observed in all cultures of *A. inflata*.

Discussion

Under microscope (in Melzer reagent) the subiculum of *Amyloflagellula verrucosa* shows thick-walled, dextrinoid hyphae and numerous small dichophyses with not differentiated stipe but with slender, dichotomic, dextrinoid ramifications; *A. inflata* possesses in its subiculum the same small dichophyses and besides the marginal cells of the cups bear dextrinoid dichophytic ramifications.

These two types of dextrinoid elements are found in the Lachnoladiaceae: Skeletal dextrinoid hyphae of *Scytinostroma* and dextrinoid dishophysis of *Vararia*, therefore the observation of the subiculum (or what is the same thing, the observation of a young immature specimen) recalls very much a representative of the genus *Vararia* with small capillary dichophyses (see BOLDIN et al. 1980, fig. 2c) such as *V. minidichophysa*, *V. microphysa* or *V. tropica*. However, many differences are to be noticed in *Amyloflagellula*: basidia are short and not at all utriform; gloeocystidia are absent; the hymenium is not a cataphymenium where coexist basidia, dichophyses and gloeocystidia. Therefore we can think that the dichophytic dextrinoid elements whose aspect is very similar to smaller capillary dichophyses of *Vararia* are the result of an astonishing convergence and not of a true relationship.

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