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Glomus insculptum, a new arbuscular mycorrhizal species from Poland

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Abstract–A new ectocarpic arbuscular mycorrhizal fungal species, *Glomus insculptum* (Glomerales, Glomeromycota), was found in inland sand dunes of southern Poland. *Glomus insculptum* produces yellow-colored spores that are globose to subglobose, (50-) 71(-85) μm diam, or ovoid, 55–60 x 80–85 μm, and have a spore wall composed of two layers. The outer layer is permanent and the inner layer is laminate and ornamented with round pits evenly distributed on its inner surface. *Glomus insculptum* forms arbuscular mycorrhizae in single-species pot cultures with *Plantago lanceolata*.

Key Words–Glomeromycota, mycorrhizae, new species

Introduction

The largest inland sandy area of Poland is the Błędowska Desert (Szczypek & Wika 1984). It is located in the eastern part of Silesia Upland (50°22'N, 19°34'E) and occupies 30 km². The unique plant communities of the Błędowska Desert are phytocenosis of inland sandy dunes and xerothermic swards with original psammophytic species. *Corynephorus canescens* (L.) P. B., *Elymus arenarius* L., and *Koeleria glauca* (Schkuhr) DC. are the dominant plant species in the dunes (Mrozik & Wika 1993).

Apart from 20 described species of arbuscular mycorrhizal fungi of the phylum Glomeromycota, examination of rhizosphere soil samples collected under plants of the Błędowska Desert in the years 1995-1997 revealed a new species of *Glomus* forming spores with pitted inner surface of their laminate spore wall layer (Błaszkowski et al. 2002). Subsequent culturing of this fungus in both trap and single-species cultures confirmed the uniqueness of this as a new species. The fungus is described here as *Glomus insculptum*.

Materials and Methods

Collection of soil samples, establishment of trap and single-species pot cultures, as well as growth conditions generally are as those described previously (Błaszkowski & Tadych 1997). Briefly, rhizosphere soils and roots of sampled plants were collected from a depth of 5-30 cm using a small garden shovel. In the laboratory, about 200-g subsamples were taken from each sample to determine the species of arbuscular fungi sporulating in the field. Then, the remaining soil-root mixtures were either air dried for 2 weeks and subsequently refrigerated at 4°C or directly used to establish trap cultures. Trap cultures were established to obtain a great number of living spores of different developmental stages and to initiate sporulation of species that were present but not sporulating in the field collections. The growing substrate of the trap cultures was the field-collected material mixed with an autoclaved coarse-grained sand coming from maritime dunes adjacent to Świnoujście (pH 6.7; 12 and 26 mg L^{-1} P and K, respectively; Błaszkowski 1995). These mixtures were placed in 9x12.5-cm plastic pots (500 cm^3) and thickly seeded with *Plantago lanceolata* L. Plants were grown in a greenhouse at 15-30°C with supplemental 8-16-h lighting provided by one SON-T AGRO sodic lamp (Philips Lighting Poland S. A.) placed 1 m above pots. The maximum light intensity was 180 $\mu E\ m^{-2}s^{-1}$ at pot level. Plants were watered 2-3 times a week. No fertilizer was applied during the growing period. Trap cultures were harvested at approximately 1-month intervals, beginning three months and ending five to seven months after plant emergence. Spores were extracted by wet sieving and decanting (Gerdemann & Nicolson 1963). Presence of mycorrhizae was determined following clearing and staining of roots (Phillips & Hayman 1970) modified as follow: tissue acidification with 20% HCl instead of 1%, and trypan blue concentration 0.1% instead of 0.05% (Koske, pers. comm.).

Single-species pot cultures were established from about 50 to 100 newly formed spores stored before inoculation in water at 4°C for 24 h. After removal of soils debris, spores were collected in a pipette and transferred onto a compact layer of mycorrhizae-free roots of 10-14-day-old seedlings of *P. lanceolata* placed at the bottom of a hole of *ca.* 1 cm wide and 4 cm deep formed in a wetted growing medium filling 8-cm plastic pots (250 cm^3). The growing medium was an autoclaved sand of maritime dunes adjacent to Świnoujście with chemical properties listed above. Subsequently, the spores were covered with another layer of roots attached to 4-6 additional host plants, and the roots and sandwiched spores were buried in the growing medium. Finally, three to six seeds of *P. lanceolata* were placed on the surface of the growing substrate and covered with a thin layer of autoclaved sand. The cultures were harvested after 4-8 months and spores extracted. The

effectiveness of the method of establishment of one-species cultures described above usually exceeded 90% (Błaszkowski et al. 2002).

Morphological properties of spores and their subcellular structures were determined based on at least 100 spores mounted in polyvinyl alcohol/lactic acid/glycerol (PVLG; Koske & Tessier 1983) and a mixture of PVLG and Melzer's reagent (1:1, v/v). Spores in all stages of development were crushed to varying degrees by applying pressure to the coverslip and then stored at 65°C for 24 h to clear their contents of oil droplets. These were examined under an Olympus BX 50 compound microscope equipped with Nomarski differential interference contrast optics. Microphotographs were captured in a Sony 3CDD color video camera coupled to the microscope.

Terminology of spore structure is that suggested by Spain et al. (1989), Stürmer & Morton (1997), and Walker (1983). Spore color was examined under a dissecting microscope on fresh specimens immersed in water. Color names are from Kornerup & Wanscher (1983). Nomenclature of fungi and plants is that of Walker & Trappe (1993) and Mirek et al. (1995), respectively. Specimens were mounted in PVLG on slides and deposited in the Department of Plant Pathology (DPP), University of Agriculture, Szczecin, Poland, and in the herbarium at Oregon State University (OSC) in Corvallis, Oregon, USA.

Color microphotographs of spores and mycorrhizae of *G. insculptum* can be viewed at the URL http://www.agro.ar.szczecin.pl/~jblaszkowski/.

Descriptions of the species

Glomus insculptum J. Błaszkowski, sp. nov. **Figs. 1-10**

Sporocarpia ignota. Sporae singulatim in solo vel in radici, e sporophoris rectic effomatae. Sporophorum nonseptatum vel parce septatum; hyalinum; (3.7-)4.8(-5.6) µm latum; pariete 0.3-0.5 µm crasso; rectum. Sporae pallide luteae vel aureae; globosae vel subglobosae; (50-)71(-85) µm diam; aliquando ovoideae; 55-60 x 80-85 µm; hypha subtenda solitaria. Tunica sporae e startis duobus (strati 1-2); strato "1" rigido, diuturno, glabro, hyalino vel pallide luteo, (0.9-)1.1(-1.5) µm crasso; strato "2" laminato, pallide luteo vel aureo, (3.2-)4.4(-6.1) µm crasso cum superficie interia cum orbicularis cavernis ordinatis, (1.2-)1.6(-2.0) µm diam, 1.2—2.0 µm profundis. Hypha hyalina; recta vel recurva; cylindrica vel infundibuliforma, raro coliga; (2.9-)5.2(-6.9) µm lata ad basim sporae; pariete hyalino; (0.7-)1.3(-2.9) µm crasso, stratis 1-2 sporae continuo. Porus e septo continuo strati 2 sporae effornata. Arbuscular mycorrhizae formans.

Sporocarps unknown. Spores borne singly in the soil (Figs. 1, 2); produced from straight sporophores (Figs. 1-7). *Sporophore* coenocytic to sparsely septate; hyaline; (3.7-)4.8(-5.6) µm wide; with a wall 0.3—0.5 µm thick; bearing spores by swelling at hyphal tips (Fig. 1). *Spores* yellowish white (2A2) to golden yellow (5B8); globose to subglobose; (50-)71(-85) µm

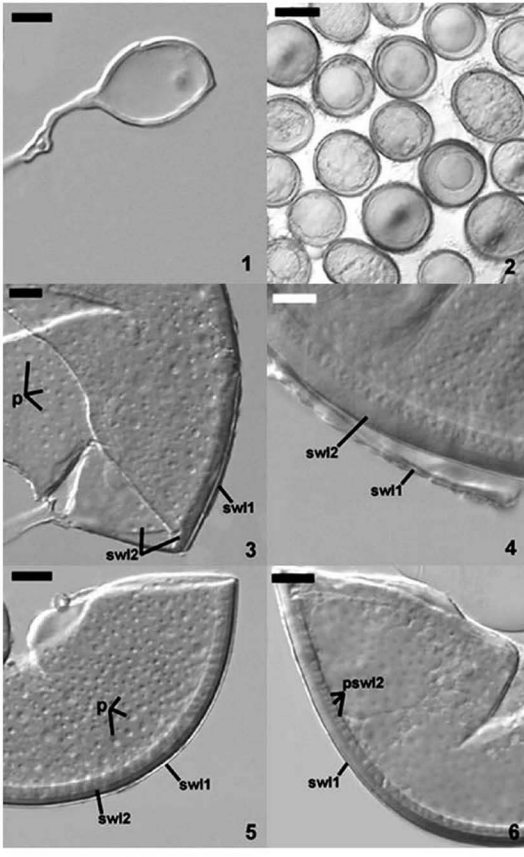
diam; sometimes ovoid; 55-60 x 80-95 μm; with a single subtending hypha (Fig. 1-3, 7). Subcellular structure of spores consists of one wall with two layers (swl 1-2; Figs. 3-7). Outermost layer 1 permanent, smooth or with a slightly roughened outer surface, hyaline, ca. 0.5 μm thick, continuous with a one-layered subtending hypha of the most juvenile spores (Fig. 1), then darkening to pale yellow (3A3) and thickening to (0.9-)1.1(-2.7) μm (Figs. 3-7). Layer 2 laminate, yellowish white (2A2) to golden yellow (5B8), (3.2-)4.4(-6.1) μm thick, with a smooth outer surface and an evenly pitted inner surface (Figs. 3-6); pits round, (1.2-)1.6(-2.0) μm diam (Figs. 3-5), 1.2-2.0 μm deep, separated by ridges, (0.7-)1.1(-1.7) μm wide (Figs. 4, 6); in the most juvenile spores, the inner surface is frequently smooth; in young (immature) spores, it contains shallow pits that may be difficult to see (Figs. 6, 7). No spore wall layers react in Melzer's reagent.

Subtending hypha hyaline to pale yellow (3A3); straight or recurved; cylindrical or slightly flared (Figs. 3, 7), rarely constricted; (2.9-)5.2(-8.5) μm wide at the spore base, consisting of two layers (shwl 1 and 2) continuous with spore wall layers 1 and 2 (Fig. 7); layer 2 of spore wall is highly thinned at the spore base and then either further thins to 0.7 μm or forms a recurved septum, 1.5-2.7 μm wide and 0.5—0.7 μm thick, positioned 1.0—6.0 μm in the hyphal lumen (Fig. 7).

Etymology. Latin, *insculptum*, referring to the pitted inner surface of the laminate spore wall layer 2.

Specimen examined: HOLOTYPE: Poland. Szczecin, associated with roots of pot-cultured *P. lanceolata*, 25 June 1999, *Błaszkowski, J., 2271* (DPP); ISOTYPES: *Błaszkowski, J.*, 2272-2309 (DPP) and two slides at OSC.

Figs. 1-10. Spores and mycorrhizae of *Glomus insculptum* in roots of *Plantago lanceolata* stained in 0.1% trypan blue. **1.** Young spore. **2.** Mature spores. **3.** Spore wall layers 1 (swl1) and 2 (swl2), pits (p) on the inner surface of spore wall layer 1, and subtending hypha (sh). **4.** Spore wall layer 1 (swl1) separated from pitted spore wall layer 2 (swl2). **5.** Spore wall layers 1 (swl1) and 2 (swl2) with pits (p) seen through the spore surface. **6.** Spore wall layer 1 (swl1) and pits of spore wall layer 2 (pswl2) seen in a cross-sectional view. **7.** Spore wall layers 1 (swl1) and 2 (swl2) continuous with subtending hyphal wall layers 1 (shwl1) and 2 (shwl2); septum (s) of the subtending hypha is visible. **8.** Arbuscule (arb) with its poorly visible trunk (tr). **9.** Coils of intraradical hyphae. **10.** Coiled (chyp) and straight (shyp) intraradical hyphae. Figs. 1, 3, 4, 6, 7, spores crushed in PVLG + Melzer's reagent. Fig. 2, spores in water. Fig. 5, spore crushed in PVLG. Figs. 8-10, roots in PVLG. Figs. 1 and 3-10, differential interference contrast; Fig. 2, bright field microscopy. Bars: Figs. 1 and 3-10=10 μm, Fig. 2=50 μm.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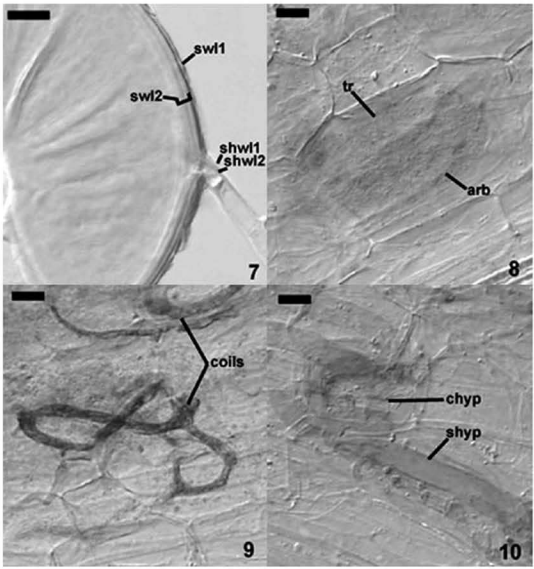

Other materials examined. Poland. The Błędowska Desert (50°22' N, 19°34' E), from the root zone of *Corynephorus canescens* (L.) P. Beauv., *Festuca rubra* L. s. s., *Holcus mollis* L. and *Juniperus communis* L., 26 June 1997, *Błaszkowski, J.*, unnumbered collection (DPP). Spores from trap pot cultures established based on rhizosphere soils of the plant species listed above and from five other cultures with rhizosphere soils of the same plant species and *P. major* L., 2 Oct. 1999, *Błaszkowski, J.*, unnumbered collection (DPP).

Distribution and habitat. Spores of *G. insculptum* were isolated from seven field-collected soil samples and 10 trap pot cultures that were established with rhizosphere soils of five plant species colonizing inland sand dunes of the Błędowska Desert (50°22' N, 19°34' E) in south of Poland. The plant species colonized by *G. insculptum* in the field were *C. canescens*,

F. rubra, *H. mollis*, *J. communis* and *P. major*. Spore abundance of *G. insculptum* in the field-collected samples ranged from 0 to 79 (mean 18) in 100 g dry soil. The arbuscular mycorrhizal fungal species richness in the soil samples containing *G. insculptum* ranged from 2 to 6 (mean 2.5) in 100 g dry soil. The fungi co-occurring with *G. insculptum* in the field were *Acaulospora lacunosa* Morton, *A. mellea* Spain & Schenck, *G. aggregatum* Schenck & Smith emend. Koske, *G. fasciculatum* (Thaxter) Gerd. & Trappe emend. Walker & Koske, *G. mosseae* (Nicol. & Gerd.) Gerd. & Trappe, and *Scutellospora dipurpurescens* Morton & Koske. The fungi accompanying *G. insculptum* in trap cultures were *A. lacunosa*, *Archaeospora trappei* (Ames & Linderman) Morton & Redecker, *G. clarum* Nicol. & Schenck, *G. intraradices* Schenck & Smith, *G. lamellosum* Dalpé, Koske & Tews, *G. pustulatum* Koske, Friese, Walker & Dalpé, two undescribed *Glomus* species., *Paraglomus occultum* (Walker) Morton & Redecker, *S. armeniaca* Błaszk., and *S. dipurpurescens*.

The soil chemical properties of the Błędowska Desert dunes ranged: pH, 5.6-5.9; NO_3 (mg L^{-1}), 9.3-10.8; P, 3-5; K, 5-13; Mg, 7-23; Na, 0-1; Cl, 13.7-18.2; KCl (g L^{-1}), 0.060-0.075; organic C (%), 0.27-0.42.

Mycorrhizal associations. In the field, *Glomus insculptum* occurred among vesicular-arbuscular mycorrhizal roots of *C. canescens*, *F. rubra*, *H. mollis*, *J. communis*, and *P. major*. Mycorrhizae of this fungus formed in one-species cultures with *P. lanceolata* as the plant host consisted of arbuscules, intra- and extraratrical hyphae. Arbuscules appeared as granular structures in cortical cells (Fig. 8). Fine branches were difficult to see. Arbuscules were numerous, but unevenly distributed in roots. The intraratrical hyphae were (4.2-)6.2(-7.4) μm wide and grew parallel to the root axis (Fig. 10). They were straight or slightly curved, sometimes dichotomously branched and frequently coiled (Figs. 9, 10); the coils were 20.8—34.6 x 10.5—22.3 μm. No vesicles were present in roots of plants up to 8-months-old. Extraradical hyphae were (1.7-)3.2(-3.5) μm wide and frequently associated with young and mature spores. In 0.1% trypan blue, arbuscules stained violet white (17A2), intramatrical hyphae violet white (17A2) to pastel violet (17A4), and extraradical hyphae pale violet (17A3).

Discussion

When observed under a dissecting microscope, spores of *G. insculptum* most resemble small-spored isolates of *G. aggregatum*, *G. arenarium* Błaszk. at al., *G. etunicatum* Becker & Gerd., *G. intraradices* Schenck & Smith, *G. pustulatum*, *G. trimurales* Koske & Halvorson, and *G. versiforme* (Karsten) Berch. All eight species form globose to subglobose and yellow-colored

spores, whose size range partly overlaps (Becker & Gerdemann 1977; Berch & Fortin 1983; Błaszkowski 1991; Błaszkowski et al. 2001, 2003; Koske 1985; Koske & Halvorson 1989; Koske et al. 1986; Morton 2000; Schenck & Smith 1982; Stürmer & Morton 1997).

Examination of subcellular structure and phenotypic properties of layers in the spore wall of specimens crushed in Melzer's reagent readily separates these species. Only *G. insculptum* forms spores in which the laminate spore wall layer is regularly pitted (Figs. 3-6). However, in young spores, the inner surface either is smooth or the pits are very shallow and difficult to see (Figs. 6, 7). Even then, *G. insculptum* is distinguishable from other species. Although *G. etunicatum*, *G. insculptum*, and *G. versiforme* have a spore wall composed of two layers, the outer layer of each differs. In *G. etunicatum* the layer sloughs as spores mature (Stürmer & Morton 1997), while in *G. versiforme* it is semi-permanent (Morton 2000) and in *G. insculptum* it is permanent (Figs. 3-7). Additionally, the outer spore wall layer of *G. insculptum* and *G. versiforme* (Morton 2000) is nonreactive in Melzer's reagent but stains dark pinkish red to reddish-purple in *G. etunicatum* (Stürmer & Morton 1997). *Glomus versiforme* also differs from *G. insculptum* in the occasional production of spores arranged in epigeous sporocarps (Berch & Fortin 1983; Morton 2000) vs. only single, hypogeous spores in *G. insculptum* and in that the mean diameter of globose spores of the former fungus is almost twice that of spores of the latter species. In contrast to the two-layered subcellular spore wall structure of *G. insculptum* (Figs. 3-7), that of *G. arenarium*, *G. pustulatum*, and *G. trimurales* consists of three layers.

The only other species of arbuscular fungi forming spores with an ornamented inner surface of their innermost wall layer are *G. kerguelense* Dalpé & Strullu and *G. verruculosum* Błaszk. However, compared with *G. insculptum*, spores of the two species are much larger (mean diameter = 71 μm in *G. insculptum* vs. 186.3 μm and 189.0 μm in *G. kerguelense* and *G. verruculosum*, respectively) and the ornamentation of the innermost layer of their wall consists of fine granules (*G. kerguelense*) or warts (*G. verruculosum*; Błaszkowski & Tadych 1997; Dalpé et al. 2002) vs. pits in *G. insculptum*.

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A new aquatic nematode-trapping hyphomycete

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Abstract—A new *Dactylella* species was isolated during a survey of aquatic fungi on submerged wood in Dianchi Lake, Kunming, Yunnan Province, China. Its simple and branched conidiophores form a few short branches or spurs near apex and produce 2–3 conidia in sympodial arrangement. Conidia are hyaline, spindle or elavate, round at the distal end, truncate at the base, 1–7-septate, mainly 2-5-septate, 37.5–100.0 (70.0) X 10.0–17.5 (14.3) μm.

Keywords—aquatic hyphomycete, *Dactylella dianchiensis*., nematode-trapping fungi

Introduction

Nematode-trapping fungi are usually isolated from rotten plant debris and soil. Their distribution on waterlogged soil or submerged wood is rarely mentioned in research literature. Only seven species of predacious hyphomycetes from an aquatic environment have been recorded (Ingold 1944, Peach 1950 and 1952, Nakagiri 1991). Recently, a survey of the fungi on submerged wood and waterlogged soil in Dianchi Lake was conducted. The methods used for the survey were to place wood samples into zipped plastic bags with waterish filter papers and to sprinkle soil samples onto CMA agar challenged with the free-living nematode, *Paragrellus redivius*. After incubation for 1–4 weeks at 25°C, the samples were examined under a dissecting microscope. A *Dactylella* species was isolated from the submerged wood and several recorded nematode-trapping fungi such as *Arthrobotrys oligospora* Fres, *A. conoides* Drechsler, *Monacrosporium elegans* Oudem, *M. thaumasium* (Drechsler) de Hoog & van Oorschot, *M. sphaeroides* Castaner, *M. cystosporium* Cooke & Dickinson and

Dactylella leptospora Drechsler were isolated from the waterlogged soil. After a study of the morphological characteristics of the *Dactylella* species and a survey of the literature (Ingold 1944, Dowsett et al. 1984, Watanabe 1992, Liu & Qiu 1993, Zhang, Liu & Cao 1994, Rubner 1996, Nakagiri & Tadeyoshi 1996, Miao et al. 1999), we believe that this fungus is an undescribed species and name it *Dactylella dianchiensis*.

We follow the traditional view that the genera *Arthrobotrys, Dactylella* and *Monacrosporium* are distinguished by conidia with or without the largest cell, conidia septa, shape and size and conidiophores knotted or producing solid conidia, which has been widely accepted (Cooke & Dickinson 1965, Castaner 1968, Scheuck et al. 1977, Van Oorschot 1985, Zhang et al. 1994, Rubner 1996, Li & Miao et al. 2003, Liu & Zhang 2003). We therefore place the new fungus into *Dactylella* rather than *Arthrobotrys*, which has been amended by Schaller et al. (1999), who considered that *Arthrobotrys* forms adhesive networks. The detailed morphological characteristics of the new species are described in comparison with those of its similar species.

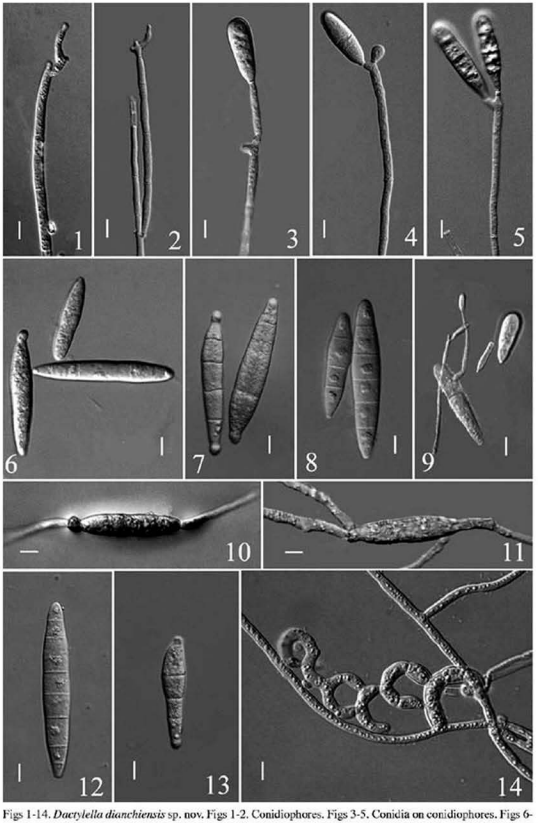
Dactylella dianchiensis Y. E. Hao et K. Q. Zhang sp. nov. (Figs 1-14)

Etym.: The species *Dactylella dianchiensis* was named after Dianchi Lake, Kunming, Yunnan Province, China, where it was first isolated.

Coloniae in agaro CMA albidae, post 6 dies 25°C 6 cm diam. Mycelium sparsum, hyphis septatis, ramosis, 2.5–5 μm latis, trimensionis formantibus. Conidiophora erecta simplex vel ramosa, 2–4 septata, 245–425 μm longa, 5–7.5 μm lata ad basim, 2.5–5 μm lata ad apicem. Conidia hyalina, fusiformis vel clavata, conidia e tuberculis geminate. 1–7 septata, praecipue 2–5 septata. Conidia secundaria clavatia, plerumque 1 septata, vulgo circa 23.9 μm longa et 5 μm lata. Habitat in ligno submerse.

Holotype: YMF1.00571D, Dianchi Lake, Kunming, Province Yunnan, China, 10-II-2003, Jing Luo. The holotype and its culture (YMF1.00571) are deposited in the Key Laboratory of Microbial Fermentation, Yunnan University.

A single spore was inoculated on PDA, a colony reached 7 cm in diameter at 28°, 5 cm at 25°C and did not grow at –4°C or 35°C within 4 days. The colony was initially whitish and turned orange white after 10 days of incubation. At the same time the reverse side of the media became faintly yellow to reddish orange. Colonies on CMA whitish, rapidly- growing and extending a diameter of 6 cm at 25°C within 6 days. Mycelium hyaline, scanty, hyphae septate, branched, 2.5–5 μm wide. Primary conidiophores (Figs 1-5) erect, simple or branched, 2–4-septate, 245–425 μm high, 5–7.5 μm wide at the base, gradually tapering upward to



Figs 1-14. *Dactylella dianchiensis* sp. nov. Figs 1-2. Conidiophores. Figs 3-5. Conidia on conidiophores. Figs 6-8,12-13. Mature conidia. Fig 9. Germinated conidium with secondary conidiophore with a secondary one-septate conidium attached. Figs 10-11. Germinated conidia. Fig 14. Adhesive network. bar=10 μm.

a width of 2.5–5 μm at the tip, initially a width of 2.5–5 μm at the tip, later often producing a few short branches or spurs near the apex, and bearing 2-3 conidia in sympodial arrangement. Primary conidia (Figs 6, 8, 12) hyaline, spindle or clavate, narrowly round at the distal end, truncate at the base, 37.5–100.0 (70.0) X10.0–17.5 (14.3) μm, 1-7-septate, mainly 2-5-septate. The proportion of conidia with 1, 2, 3, 4, 5, 6 and 7 septa was 10.0%, 17.1%, 14.3%, 24.3%, 21.4%, 8.6% and 4.3% respectively. Some primary conidia had small tubercles at both ends (Figs 7, 13) and could germinate from these tubercles (Fig 10).The primary conidia could produce secondary conidiophores and secondary conidia (Fig 9). The secondary conidia spindle-shaped, about 23.9 X 5 μm, with 1 septum. Capturing nematodes by simple adhesive networks (Fig 14).

Table 1. Morphological comparison of *Dactylella dianchiensis, D. iridis (Trinacrium iridis, D. ramiformis)*[1], *D. crassa*, and *D. multiformis*

Species	Size of primary conidia	Number of primary conidial septa
D. dianchiensis	37.5–100.0 (70.0) X 10.0–17.5 (14.3) μm	1-7 (mainly 2-5)
D. iridis	47.5–155.0 X 7.5–16.3 μm	4-10
D. crassa	44.5–60 X 10–13 μm	1-5 (mainly 3-4)
D. multiformis	35–90.3 X 4–7.5 μm	4-12

[1] *Trinacrium iridis* (Watanabe 1992) and *Dactyllela ramiformis* (Liu & Qiu 1993) are synonyms of *Dactyllela iridis* (Nakagiri & Tadeyoshi 1996, Rubner 1996).

Dactylella dianchiensis is distinguished from *D. iridis* (Zhang, Liu & Cao 1994), *D. crassa* (Miao, Lei & Liu 1999) and *D. multiformis* (Dowsett, Reid & Kalkat 1984) by the size and septation of primary conidia (table 1), although it resembles those fungi in primary conidial shape and traps nematodes by means of the three-dimensional adhesive networks. *D. dianchiensis* produces conidia with small tubercles at both ends and without any branches, while *D.iridis* produces some branched conidia. Another similar species, *D. submerse* (Ingold 1944), does not produce any trapping devices.

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MYCOTAXON

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Notes on dictyosporic hyphomycetes from China IV. The genus *Berkleasmium*

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Abstract—Two new species and a new Chinese record of the genus *Berkleasmium* are reported. They are *B. atrovirens* and *B. taishanense*. The new record for China is *B. inflatum*. Latin diagnoses, English descriptions and illustrations of the two new species are presented. The specimens studied were deposited in the Herbarium of Shandong Agricultural University: Plant Pathology (HSAUP).

Key words— *Berkleasmium atrovirens*, *Berkleasmium taishanense*

Introduction

Zobel (1854) erected *Berkleasmium* with *B. cordaeanum* Zobel as its type. However, *B. cordaeanum* is a synonym of *Sporidesmium concinnum* Berkeley (1845). Saccardo (1882) included the fungus as a species of *Sporidesmium* Link ex Fr. (1809). Moore (1958, 1959) reassessed *Sporidesmium*, and the genus *Berkleasmium* was retrieved for certain sporodochial deuteromycetes that had been described in *Sporidesmium* (sensu Saccardo). *Berkleasmium concinnum* (Berk.) Hughes (1958) is the correct name of type species of the genus. Subsequently at least 10 species were assigned to the genus, which produces phaeodictyospores acrogenously on short, simple conidiophores, or else are sessile with sporodochial conidiomata. This genus is reported for the first time from China.

Taxonomic Description

Berkleasmium atrovirens G. Z. Zhao et T. Y. Zhang *sp. nov.* **FIGURE 1**

Coloniae in substrato naturali nigrae, granulosae, inconspicuae, constantes e sporodochiis arcte aggregates, sub lente sporodochia singula sparsa brunnea visibilia. Mycelium sparsum, pro maxima parte submersum. Conidiophora brevia, simplicia, non ramosa, erecta, recta vel flexuosa, pallide brunnea vel brunnea, 13-16µm longa, 1.5-3.5µm lata. Conidia singula in apice conidiophori oriunda, elliptica, ovoidea vel clavata, irregulariter muriformia, viridia vel atrovirentia, levia, 30-37 x 15-17µm; in parte basali obconica, denticulata, pallidiora vel subhyalina.

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Habitat: In cortice emortuo Syzygii samarangensis (Bl.) Merr. & Perry, Nanning, Guangxi Provincia sinica, 9 XI 1999, leg. G. Z. Zhao, HSAUP992154 (=ZGZ990654), hic designatus holotypus. In ramis emortuis Artocarpi heterophylii Lam. Xiamen, Fujian Provincia sinica, 25 VII 2000, leg. X. Sun, HSAUPH5086 (=SX-20086).

Colonies on natural substrate black, granular, inconspicuous, consisting of groups of sporodochia. Under the stereo microscope individually scattered, superficial, brown sporodochia are visible. Mycelium scanty, mostly immersed. Conidiophores short, simple, unbranched, erect, straight or flexuous, pale brown to brown, 13-16 x 1.5-3.5μm. Conidia formed singly at the apex of each conidiophore, ellipsoidal, ovoid to clavate, irregularly muriform, green to dark green, with basal part paler than the upper part or subhyaline, smooth, 30-37 x 15-17μm, basal cell obconical, pale brown to subhyaline, truncate at the base.

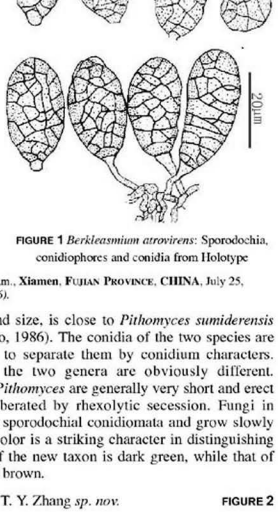

FIGURE 1 *Berkleasmium atrovirens*: Sporodochia, conidiophores and conidia from Holotype

Holotype: On dead bark of *Syzygium samarangense* (Bl.) Merr. & Perry, **Nanning, GUANGXI PROVINCE**, 9 Nov 1999, Coll. G. Z. Zhao, *HSAUP992154 (=ZGZ990654)*. Other specimen examined on dead branch of *Artocarpus heterophyllus* Lam., **Xiamen, FUJIAN PROVINCE, CHINA**, July 25, 2000, Coll. X. Sun, *HSAUPH5086 (=SX-20086)*.

This fungus, in conidium morphology and size, is close to *Pithomyces sumiderensis* Hol.-Jech. (Holubová-Jechová & Mercado, 1986). The conidia of the two species are both clavate or obovoid. It is difficult to separate them by conidium characters. However, the sporulation patterns in the two genera are obviously different. Conidiophores or conidiogenous cells in *Pithomyces* are generally very short and erect on creeping hyphae, the conidia are liberated by rhexolytic secession. Fungi in *Berkleasmium* often form fundamentally sporodochial conidiomata and grow slowly in the substrates. In addition, conidium color is a striking character in distinguishing them from each other. Conidium color of the new taxon is dark green, while that of *Pithomyces sumiderensis* is brown or dark brown.

Berkleasmium taishanense G. Z. Zhao et T. Y. Zhang *sp. nov.* **FIGURE 2**

Sporodochia disseminata, punctiformia, atra, nitentia, saepe inconspicua. Mycelium immersum, ex hyphis pallide brunneis laevibus ramosis septatis 1-2μm latis compositum. Conidiophora seminacronematosa vel micronematosa, fasciculata, simplicia, sensim septata, erecti, recta vel flexuosa, laevia, hyaline vel subhyalina, usque ad 10μm longa, 1.5-2μm lata, vesiculis inflatis 7μm crassis praedita. Cellulae conidiogenae monoblastieae in conidiophoris incorporatae, terminales, clavatae vel pyriformes, laeves. Conidia acrogena, solitaria, irregulariter muriformia, ellipsoidea, clavata, obovata vel pyriformia, brunnea vel atro-brunnea, levia, 22-35 x 15-18.75μm, aliquot conidia separata saepe cellulis conidiogenis persistentibus.

Etym.: L., *taishanense* = referring to Taishan mountain area, the type locality.

Habitat: in ligno putrido rami emortui arborum indeterminatarum in Mounte Taishan, Shandong Provincia sinica, 10 VII 2001, leg. G. Z. Zhao, IISAUPH$_{tr}$0452 (=ZGZII$_{tr}$002), Holotypus.

Sporodochia scattered, punctiform, black, shining, often inconspicuous. Mycelium immersed in the substrate, composed of pale brown, smooth, septate, 1-2μm wide hyphae. Conidiophores semimacronematous or micronematous, fasciculate, simple, unbranched, inconspicuously septate, erect, straight or flexuous, smooth, hyaline to subhyaline, up to 10μm long (including the conidiogenous cell), 1.5-2μm wide, with bladder-like swelling 7μm wide. Conidiogenous cells monoblastic, integrated, terminal, clavate or pyriform, inflated, smooth. Conidia acrogenous, solitary, dry, ellipsoidal, clavate, obovate or pyriform, brown to dark brown, 22-35 x 15-18.75μm, when detached usually carrying away with them the upper part of the conidiophore.

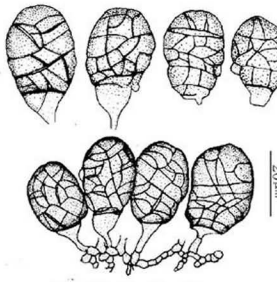

FIGURE 2 *Berkleasmium taishanense*: Sporodochia, conidiophores and conidia

On rotten wood of dead branch of an undetermined tree, Mount Taishan, **SHANDONG PROVINCE, CHINA**, 10 July 2001, Coll. G. Z. Zhao, *IISAUP$_{tr}$0452* (=*ZGZII$_{tr}$002*), Holotype.

Berkleasmium taishanense resembles *B. atrovirens* and *B. inflatum* with respect to conidium morphology. However, it can be separated from *B. atrovirens* by conidium color and basal cell characters. The characteristic green to dark green colored conidia and the small unswollen conidial basal cell of *B. atro virens* provide the useful features in differentiating *B. atrovirens* from *B. taishanense*. Conidia of *B. inflatum* reach a size range of 30–47 x 18.5-21μm, and they are considerably larger than conidia of *B. taishanense*. The much

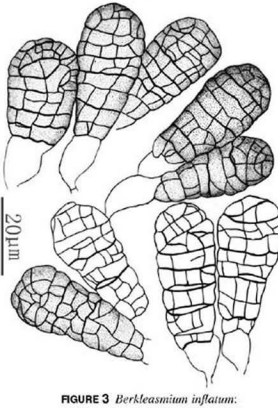

FIGURE 3 *Berkleasmium inflatum*: Conidiogenous cells and conidia

more inflated cells of conidiophore in *B. inflatum* are also helpful in separating *B. inflatum* from *B. taishanense*.

Berkleasmium inflatum Hol.-Jech., 1987, Ceská Mykologie, 41(1):29-31. **FIGURE 3**

Conidiophores clavate to obpyriform, up to 30μm long (including the conidiogenous cells), apical cell inflated, 6-9.5μm wide. Conidia obovoid, dark brown in the apical part, pale brown in the basal cells, 30-47 x 18.5-21μm.

On dead rotten wood, **CHONGQING**, **CHINA**, Aug. 8 2000, Coll. G. Z. Zhao, *HSAUPH 2745 (=ZGZH 0445)*.

This collection differs slightly from the original description (Holubová-Jechová, 1987). In the original description, the conidiophore had 1-3 bladder-like swellings, but in our collection only 1 bladder-like swelling was found. Other characters are consistent with the original description.

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中国砖格孢子丝孢菌研究 IV. 顶丛格孢属*

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摘 要：报道顶丛格孢属的两个新种，暗绿顶丛格孢*Berkleasmium atrovirens* G. Z. Zhao et T. Y. Zhang和泰山顶丛格孢*Berkleasmium taishanense* G. Z. Zhao et T. Y. Zhang，和一个中国新记录种，膨梗顶丛格孢*Berkleasmium inflatum* Hol.-Jech。研究标本保存在山东农业大学植物病理学标本室（HSAUP）。

关键词：新种，暗绿顶丛格孢，泰山顶丛格孢

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Glomus hyderabadensis, a new species: its taxonomy and phylogenetic comparison with related species

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Abstract—A novel *Glomus* species was isolated from rhizosphere soils supporting *Allamanda cathartica* from Hyderabad, India. The spores proliferate and bud out a daughter spore terminally which is connected to the mother spore by a small connective. Sequence analysis of D1D2 variable domain of 26S ribosomal RNA gene of this *Glomus* sp. shows that it is different from other known AM fungi. The morphological and molecular phylogenetic analyses support the creation of *Glomus hyderabadensis* sp. nov.

Key words—morphotaxonomy, molecular sequencing, phylogeny

Introduction

The genus *Glomus* Tulsane & Tulsane represents by far the largest genus within the Glomerales and is placed in the monogeneric family Glomeraceae (Schüßler et al. 2001). Based on rRNA SSU analysis and on the basis of natural relationship of AM fungi, a new fungal phylum Glomeromycota has been erected (Schüßler et al. 2001). Taxonomy and identification of glomalean fungi has traditionally relied on the morphology of their large multinucleate spores. Phylogenetic analysis based on ribosomal RNA gene sequences (Redecker et al. 2000, Schwarzott et al. 2001) indicate that the genus is non-monophyletic in nature.

During a survey of the rhizosphere soils of medicinal plants an interesting arbuscular mycorrhizal (AM) fungus belonging to genus *Glomus* was collected. The spores of this AM fungus are unique, as they proliferate / bud out a daughter spore terminally. The wall layers in the daughter spore are similar to the mother spore. This type of proliferation or bud out spore production is not reported in any of the existing AM fungi (Morton & Benney 1990; Schenck & Perez 1990; Manoharachary et al. 2002). Further the other spore characteristics do not match with any of the described species under genus *Glomus*. Sequence analysis of the D1/D2 variable domain of 26S ribosomal RNA gene shows that the sequence of this fungus is different from all the existing AM fungi reported in the literature, hence it is described here as a new taxon.

Materials and Methods

Collection of AM fungal spores

The AM fungal spores were isolated by wet sieving and decanting method (Gerdemann & Nicolson 1963) from the rhizosphere soil of *Allamanda cathartica* L. (Apocyanaceae).

DNA isolation

Twenty spores were washed thoroughly three times with sterile distilled water. DNA was isolated using GenElute™ Plant Genomic DNA kit, G2N10 (Sigma Chemical Company, St. Louis, USA). The fungal spores were mechanically disrupted in a 1.5 ml microfuge tube using disposable pistel (Sigma Chemical Company, St. Louis, USA). The lysis of spores and purification of DNA was performed as per manufacturer's instructions.

Amplification of ITS region and D1D2 region of 26S ribosomal RNA gene

The entire ITS and D1D2 variable domain of 26S rRNA was amplified with primers pITS1 (5'-TCC GTA GGT GAA CCT GCG G-3') (White et al. 1990) and pNL4 (5'-GGT CCG TGT TTC AAG ACG G-3') (Kurtzman & Robnett 1998). The primers were obtained from Integrated DNA Technologies, Inc., USA. Polymerase Chain Reactions were performed in a final reaction mixture (100μl) containing 50ng genomic DNA, 25pmols each of primers (pITS1 and pNL4R), 200 mM each of dATP, dTTP, dGTP and dCTP (Promega Corporation, USA); 2.5mM $MgCl_2$ and 2.0 units of Taq polymerase (Promega); and 10 μl of 10X reaction buffer (Promega). The amplification reactions were performed in a PTS 100 Mini Cycler (MJ Research, USA) with the following cycling parameters; initial denaturation for 5 minutes at 94°C, followed by 30 cycles of 30 seconds at 94°C, 30 seconds at 55°C and 1.0 min at 72°C, with a final extension for 10 minutes at 72°C and cooled at 4°C. The amplified products were separated on 1.2%

agarose (Sisco Research Laboratories, India) gel by electrophoresis and visualized by staining with ethidium bromide (0.5μg/ml). Gel photographs were taken using VDS Image Master (Pharmacia Biotech, USA).

Sequencing of D1D2 region

Direct sequencing of PCR-amplified ITS and D1/D2 domains was performed by using ABI Big Dye Terminator kit. The D1D2 variable domain of 26S ribosomal RNA was performed with 2 primers, pNL1 (5 -GCA TAT CAA TAA GCG GAG GAA AAG-3) and NL4. Sequencing reactions were purified by ethanol and sodium acetate precipitation, the pellet was washed twice with 70% ethanol, which considerably improved the removal of dye terminators from the reaction. Processing of the samples for loading on to ABI 310 model sequencer was performed as per manufacturer's instructions. The oligonucleotide primers used for sequencing were procured from Integrated DNA Technologies (IDT), USA.

Molecular phylogenetic analysis

The sequences were aligned using Clustal X program (Thompson et al. 1997) and manually corrected. Sequence similarity search was done using GenBank BLASTN search (Altschul et al. 1990), sequences of closely related taxa were retrieved and used for phylogenetic analysis. The GenBank accession numbers of the related species are given in the phylogenetic tree. For the Neighbour-Joining analysis (Saito & Nei 1987), the distances between the sequences were calculated using Kimura's two-parameter model (Kimura 1980). Sites where gaps existed in any of the sequences were excluded.

Nucleotide sequence accession number

The D1D2 domain sequence determined in this study has been deposited in GenBank database with the following number: AY211274

Results and Discussion

Glomus hyderabadensis Swarupa, Kunwar, Prasad et Manohar ***sp. nov.*** **Figs. 1-7**

Sporocarpia ignota. Sporae singulae, globosae, subglobosae vel ellipsoidae, 97-136 μm diametro, melleus vel aurantius-brunneus; sporae tunica e stratis tribus in uno terma (terma A). Hypha sustinens singulus, infundibuliformis. Maturus sporae prolificus terminalis, procreans gemmiformis sporae filiales; sporae filia sessilis, singulae, globosae, vel sub-globosae vel ellipsoideus. Sporae filialis consociatus cum nater sporae ab apertus connectivum.

[***Etymology***: Specific epithet derived from the place of collection (Hyderabad)].

HOLOTYPUS HIC DESIGNATUS ex ralix solum *Allamanda cathartica*, Hyderabad, India. 30. 5. 2001, leg. Swarupa, Herb. No. HCIO 43,918.

Sporocarps unknown. Spores formed singly, globose, subglobose to ellipsoidal, 97-136 μm in diam, honey coloured to orange-brown; spore wall as three walls in a single group (group A). Composite spore wall 3.3-5.9 μm thick, adherent, outer wall smooth or roughened, dull yellow, 1.1-2.4 μm, in older spores walls perforated, with an aperture of 1-1.5 μm diam, middle wall single, non layered, rigid, unit, orange-brown, 1.1-2.7 μm, inner wall rigid, non layered, dull yellow, 1.1-2.1 μm. Subtending hypha single, slightly flared toward the point of attachment, 15-32 μm thick, 136-233 μm long, rarely branched, pore in the subtending hypha occluded by a thick straight septum. Mature spores proliferating terminally, producing bud-like daughter spore; daughter spore sessile, single, globose, subglobose, ellipsoidal, 18-48.4 μm in diam, honey coloured to orange-brown; composite daughter spore wall 3-5.6 μm thick, number of wall groups one (group A), adherent, three wall layers, outer wall smooth or roughened, dull yellow, 1-2.2 μm thick, middle wall single, non layered, rigid, unit, orange-brown, 1-2 μm thick, inner wall rigid, non layered, dull yellow, 1-1.8 μm thick. Daughter spore linked to mother spore by an open connective, 5.6-9.5x8.2-11 μm.

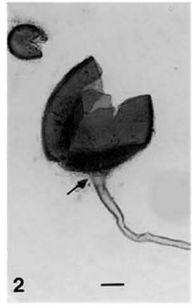
Glomus hyderabadensis is characterized by the production of sessile, single, daughter spores from the mother spore and perforations in the spore wall. The spore measurements and wall characteristics also differ from the known species of *Glomus* (Schenck & Perez 1990). These observations merit its description and erection as a new taxon of species rank.

Glomus pustulatum Koske, Friese, C. Walker & Dalpe produces numerous hyaline, circular to irregular blister like thickenings on the outermost spore wall (Koske et al. 1986). In the present fungus only one daughter spore is produced per mother spore and the colour and wall structure of the daughter spore is like the mother spore. In *G. etunicatum* W.N. Becker & Gerd. the outer wall is smooth or roughened from decomposition of outer wall and adherent debris but has two groups (A and B) of spore walls and the composite wall is thicker (4-13 μm) (Becker & Gerdemann 1977), whereas in the new taxon only one group (A) of spore wall is present and it is thinner (3.3-5.9 μm).

Perforations in the wall are reported in *G. geosporum* (T.H. Nicolson & Gerd.) C.Walker also, but these perforations are sometimes absent from mature specimens and the middle wall is laminated. The inner wall is membranous and forms a septum separating the spore contents from the lumen (Walker 1982). However, in the present fungus perforations are present in the mature spores, outer wall is dull yellow and is always intact even at maturity unlike in *G. geosporum* where it is

Figs. 1-7. *Glomus hyderabadensis* Scale bar = 30 μm. 1. Mother spore with young terminal daughter spore (arrow). 2. Daughter spore detached from mother spore, subtending hypha flared at the point of attachment with the spore (arrow). 3. Ellipsoidal spore. 4. Subglobose spore. 5. Daughter spore connected with the mother spore with a connective (arrow), septum (arrow) at the point of attachment of subtending hypha with the spore. 6. Similar three layered wall in mother and daughter spore (arrows). 7. Perforations (arrows) in the spore wall.


1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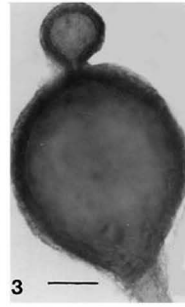
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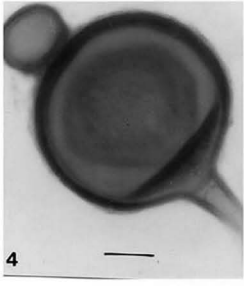
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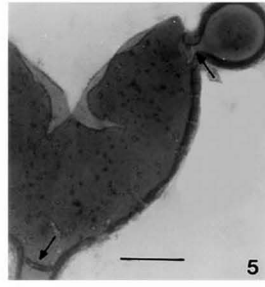
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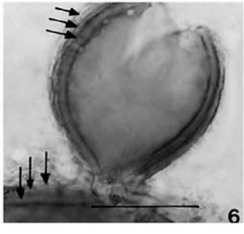
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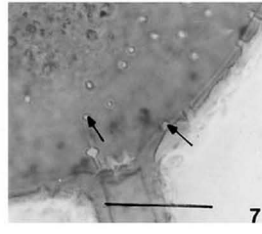
7

hyaline and sometimes absent from mature specimens. In *G. multisubstensum* Mukerji, Bhattacharjee & J.P. Tewari and *Gigaspora rosea* T.H. Nicolson & N.C. Schenck (= *Gi. candida* Bhattacharjee, Mukerji, J.P. Tewari & Skoropod) perforations are reported but they seem to be caused due to hyperparasitism. In *G. multisubstensum* the perforations were seen in surface view while in sectional view transverse fissures were seen (Mukerji et al. 1983). In *Gi. rosea* the perforations in the spore wall appeared to be caused by the parasitic action of soil microorganisms since its inner surface showed numerous hypha like structures passing through the perforations (Bhattacharjee et al. 1982). The perforations were either dispersed or in groups and varied in size indicating that more than one type of parasite may have been involved, while in our study of *G. hyderabadensis* perforations were uniformly distributed and were mostly uniform in size.

In *G. globiferum* Koske & C. Walker the spores are formed singly or in pairs or triplets adhering to each other by common peridial hyphae, bearing numerous terminal or intercalary, globose or ovoid vesiculate swellings (Koske & Walker 1986). Wheras in *G. hyderabadensis* peridium is absent with single daughter spore attached to the mother spore.

Phylogenetic analysis

The ITS and D1/D2 region was amplified with primers ITS1 and NL4, the amplicon was about 1.2 kb. Attempts to sequence the ITS region with primers ITS1, ITS2, ITS3 and ITS4 were unsuccessful for unknown reasons. As an alternative, we have sequenced the D1/D2 domain of 26S rRNA gene. The 26S rRNA domain D1/D2 have been determined for all the known yeast taxa (Kurtzman & Robnett 1998; Fell et al. 2000) and are extensively used for describing new yeast species along with morphological and physiological characters. With few exceptions, the yeast taxa that differ by more than 1% variation in the D1/D2 domain are considered as separate species (Kurtzman & Robnett 1998; Fell et al. 2000). The D1/D2 sequence of PCR amplified ITS-D1D2 fragment was determined using primers NL1 and NL4. Both the strands of the PCR product were sequenced. The determined sequence is 510 bases in length.

GenBank search using BLAST (Altschul et al. 1990) resulted in retrieval of several algal sequences. However, these sequences showed 23.6 to 35.1% variation with *G. hyderabadensis* sequence and appear to be not closely related to *G. hyderabadensis*. The first 10 hits were as follows, GYNRGNP *Gymnodinium catenatum* Graham, AF409122 *Mallomonas asmundae* (Wujek and van der Veer) Nichols, AF409121 *Chrysolepidomonas dendrolepidota* Peters and Anderson, AF318261 *Dinophyceae* gen. sp. *anufer* Cons4, AF409125 *Vacuolaria virescens* Cienkowski, AF210742 *Vacuolaria virescens*, AF417673 *Nitzschia laevis* Hust., AF417670 *Nitzschia alba* (Kütz) W. Smith, AF210743 *Olisthodiscus luteus* Carter and OD28SRRNA *Ochromonas danica* Prings. *Glomus* sp. HClO – 43,918 differs from the algal taxa by 23.6 to 35.1% variation. The sequence divergence among the

algal taxa used in the analysis ranged from 15.5 to 34.3%, suggesting that they are highly divergent sequences. Our attempts to culture algae from the crushed spores of *Glomus* sp. HCIO-43,918 on different media did not yield any algal species.

As the morphotaxonomic features of the new fungus suggest that it may be a new *Glomus* species, we restricted our next BLAST search to Glomeromycota. The first hits in the BLAST search were *Archaeospora gerdemannii* (S.L. Rose, B.A. Daniels & Trappe) J.B. Morton & D. Redecker AJ510234, AJ271712 and AJ510233 followed by *Glomus occultum* C. Walker AJ271713. Phylogenetic position of *G. hyderabadensis* in relation to other AM fungi based on D1/D2 variable domain sequences is shown in Figure-8. Based on the sequence analysis of D1/D2 domain *Ar. gerdemannii* is the nearest relative of *Glomus* sp. HCIO – 43,918, but the

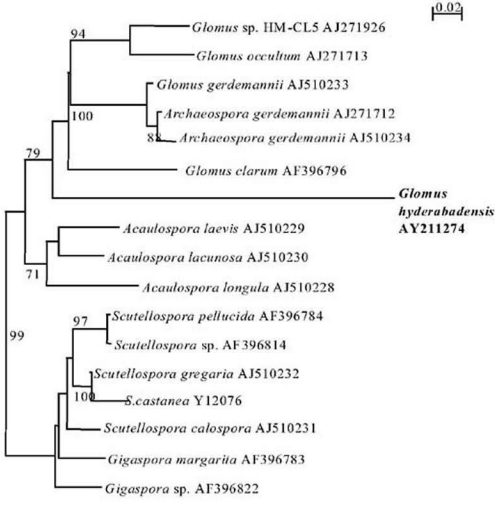

Fig. 8. Phylogenetic tree depicting the relationship between *Glomus hyderabadensis* and other reference taxa. The tree was constructed based on D1/D2 variable domain of 26S rDNA using program N J Plot. Bootstrapping was done in 1000 replications. Values for frequencies less than 70% are not given.

new fungus differs from its nearest relative by more than 29.2% sequence variation. The sequence variation between two isolates of *Ar. gerdemannii* was 0.8%. Variation among the three *Acaulospora* sp. viz. *A. lacunosa* J.B. Morton, *A. longula* Spain & N.C. Schenck and *A. laevis* Gerd. & Trappe ranged from 9.3 to 9.8% indicating that they are distinct species.

Among the *Glomus* species, *Glomus* sp. strain HM-CL5 has more sequence imilarity to *Glomus* sp. HCIO – 43,918, however both of them differ from each other by 30.5% variation. *G. occultum* is the nearest relative of *Glomus* sp. strain HM-CL5 (6.8% variation). Interestingly *Glomus* sp. strain HM-CL5 differs from other *Glomus* species used in the sequence analysis by 16.1 to 22.2% variation. These results indicate that D1/D2 sequences are highly divergent among *Glomus* species.

These results together with the novel morphotaxonomic features of *Glomus* sp. HCIO – 43,918 support the description of a new taxon *G. hyderabadensis*.

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MYCOTAXON

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Changes and additions to the checklist of North American Lichens. - I

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Abstract–*Bactrospora macrospora* has been found to be synonymous with *Gyalecta lamprospora*, resulting in the new combination *Bactrospora lamprospora*. *Lecidea hebescens* is placed in synonymy with *Porpidia albocaerulescens*, and *Arthonia willeyi* is placed in synonymy with *A. diffusa*. The following taxa are lectotypified: *Arthonia diffusa*, *Arthonia willeyi*, *Gyalecta lamprospora*, *Lecanora pleiospora*, *Lecanora pleistospora*, *Lecanora thelococcoides*, and *Lecidea hebescens*.

1. *Arthonia diffusa* Nylander

Arthonia diffusa Nylander, Flora, 68(24): 448. 1885. TYPE: On bark, Illinois, USA. *Hall s.n.* (H-NYL 5046a!, lectotype **[selected here]**; PH!, isolectotype; H-NYL 5044!, H-NYL 5045!, H-NYL 5047! syntypes (see below).

Arthonia diffusa Nylander *nom. nud.*, Mem. Soc. Sci. Nat. Cherbourg, 5: 337. 1858.

Syn. nov. *Arthonia willeyi* Tuckerman *ex* Hedrick, Mycologia, 25(4): 311. 1933. TYPE: On trees near Athens, Illinois, USA. *Wolf s.n.* (PH!, lectotype **[selected here]**)

Arthonia diffusa is a distinctive species, apparently endemic to eastern North America, that is easily recognized by its sessile usually pruinose ascomata, often constricted at the base; large, usually pruinose pycnidia; *Trentepohlia* photobiont and 2-septate ascospores. The measurement of immature ascospores likely accounts for the size difference noted by Hedrick when establishing *A. willeyi*. Thus, Willey's (1890) recognition that the then-unpublished *A. willeyi* was identical with *A. diffusa* is confirmed.

As discussed by Lendemer & Hewitt (2002) there are five syntype specimens of *A. diffusa* in H-NYL, four of which include material from New England. It was two of these specimens collected by Edward Tuckerman that served as the basis for Nylander's invalid publication of the name in 1858. The description of 1885 however, includes three additional specimens; two collected by Henry Willey and one by Elihu

Hall in Illinois. The Hall collection is here selected as the lectotype because both pycnidia and apothecia are present on the specimen.

Though a holotype was clearly designated in the protologue of *A. willeyi*, the specimen could not be found at MICH (P. Rogers, pers. comm.). Thus, the isotype in PH is here selected as the lectotype. Further study of *Arthonia* will likely show that additional names should be placed in synonymy with *A. diffusa*.

2. *Bactrospora lamprospora* (Nylander) Lendemer *comb. nov.*

Gyalecta lamprospora Nylander, Flora, 68(16): 312-313. 1885. TYPE: On bark, near Philadelphia, Pennsylvania, USA. *Eckfeldt 45* (PH!, lectotype **[selected here]**; H-NYL 21968!, isolectotype).

Syn nov. *Bactrospora macrospora* R. C. Harris, Some Florida Lichens, 40. 1990. TYPE: On *Acer*, hardwood swamp, just E of Loften Creek on Fla. Hwy. A1A, 5.5 miles W of Amelia River, Nassau Co., USA. *Harris 21152* (NY!, holotype).

Melampylidium macrosporum (R. C. Harris) Henssen *in* Henssen & Thor *in* Hawksworth, DL (ed.): Ascomycete Systematics. Problems and Perspectives in the Nineties, p. 45.

The generic disposition of *Bactrospora macrospora* has been the subject of some controversy. However, Egea and Torrente (1995) consider *Melampylidium* Stirton a synonym of *Bactrospora* A. Massalongo. Harris (1995) also argued for retaining *B. macrospora* in *Bactrospora*. Refer to Harris (1990) or Egea and Torrente (1993) for a description of *B. macrospora*.

3. *Porpidia albocaerulescens* (Wulfen) Hertel & Knoph

Lichen albo-caerulescens Wulfen *in* Jacquin, Collect. Bot. 2: 184, tab. XV. fig. 1. 1788. TYPE: *Arnold Lich. Exs. 894* (M, neotype (selected by Hertel, 1977). (for synonymy see Hertel, 1984)

Syn. nov. *Lecidea hebescens* Nylander, Sert. Lich. Trop., 41. 1891. TYPE: On rock, Lookout Mountain, Tennessee, USA. *Calkins s.n.* (H-NYL 15497!, lectotype **[selected here]**; PH!, NY! probable isolectotypes).

Fink (1935) included *L. hebescens* as a synonym of *P. albocaerulescens*. Magnusson (1936) however, retained the species on the basis of the brown coloration of the thallus. Other then the brown coloration the type collection is typical *P. albocaerulescens*. See Gowan (1989) for a description of this species.

4. *Acarospora thelococcoides* (Nylander) Zahlbruckner

Lecanora thelococcoides Nylander, Set. Lich. Trop., 37. 1891. TYPE: On soil, southern California, USA. *Orcutt s.n.* (H-NYL 24962!, lectotype **[selected here]**). EPITYPE: On granite derived soil, open spaces in full sun among *Adenostoma fasciculatum* chaparral, Menifee Hills, Wildomar, Riverside Co., California, USA. *Knudsen 529* (hb. Lendemer!, epitype **[selected here]**; herb.

Knudsen, hb. Lendemer, ASU!, BG!, CANB!, CHR!, FH!, H!, MIN!, NDA!, NY!, OSU!, SBBG!, TNS!, UC!, isoepitypes).

Acarospora thelococcoides (Nylander) Zahlbruckner, Lich. Catalogus, 5: 98. 1928

Lecanora pleiospora Nylander *in* Hasse, Bull. Tor. Bot. Club, 24(9): 446. 1897. TYPE: On clay, San Gabriel Mountains, California, USA. *Hasse s.n.* (H-NYL 24865!, lectotype **[selected here]**; H-NYL 24868!, isolectotypes).

Acarospora pleiospora (Nylander *in* Hasse) Zahlbruckner, Beihefte Bot. Centralbl., 13: 163. 1902.

Lecanora pleistospora Nylander *in* Hasse, Bull. Tor. Bot. Club, 24(9): 446. 1897. TYPE: On clayey soil, Soldier's Home, Los Angeles Co., California, USA. *Hasse s.n.* (H-NYL 24866!, lectotype **[selected here]**; PH!, isolectotype).

Acarospora pleistospora (Nylander *in* Hasse) Zahlbruckner, Beihefte Bot. Centralbl., 13: 163. 1902.

The synonymy of *A. thelococcoides* presented here has already been proposed previously by Magnusson (1929). However, recent research (Knudsen, 2003) resulted in the discovery that Hasse (1897) confused *A. pleistospora*, with *A. obpallens*. The later descriptions by Hasse (1913) show that he corrected this confusion. The names *L. pleiospora* and *L. pleistospora* are not currently included in the North American checklist (Esslinger 1997). However, they should be included as synonyms of *A. thelococcoides* because specimens with these obsolete names remain un-annotated in many herbaria. It is important to note that the type material of *L. pleistospora* is in fact a mixture of *A. thelococcoides* (the lectotype) and *A. obpallens* (H-NYL 24867). The specimen representing *A. thelococcoides* is here selected as the lectotype because *L. pleistospora* and *L. obpallens* were simultaneously published and the best course of action seems to be to reduce *L. pleistospora* into synonymy with a name that clearly has priority. Also, all specimens (examined by Knudsen) later distributed by Plitt in *Lichenes Exsiccati ex Herb. Dr. H. E. Hasse Relicti* under the names *A. pleistospora* and *A. pleiospora* are *A. thelococcoides*. See Knudsen (2003) for a discussion of the differences between *A. obpallens* and *A. thelococcoides*. Because the type specimen of *A. thelococcoides* consists of only a few fertile areoles I have chosen to designate (and widely distribute) a recent collection as an epitype to aid future workers in the application of this name.

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Synopsis and systematic reconsideration of *Karlingiomyces* (Chytridiomycota)

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Abstract—The genus *Karlingiomyces* is reviewed and the validity of endo-operculation as a character is questioned in comparison to exo-operculation. *Karlingiomyces* has been considered an operculate member of the *Rhizophlyctis/Karlingia* complex, an assemblage of saprotrophic, interbiotic, mostly monocentric Chytridiomycota. Members of this complex have similar thallus morphology, including often more than one discharge pore and several main rhizoidal axes per sporangium. However, a definite exo-operculum delimits *Karlingiomyces* species from other taxa of the complex. *Karlingiomyces* contains five species, of which three are chitinophilic and two are cellulosic. One of the cellulosic species, *Karlingiomyces exooperculatus*, represents a new combination. A key to species of *Karlingiomyces*, descriptions, illustrations, information on types, and discussions of taxa are provided. Based on probable endo-operculation and on what may be assessed of the zoospore, three other species of *Karlingiomyces* and one variety are excluded from the genus. Recent morphological and molecular data have cast doubt on the continued inclusion of *Karlingiomyces* in the *Rhizophlyctis/Karlingia* complex.

Key words—chytrid, monocentric, nomenclature, operculum, polycentric, zoospore

Introduction

Rhizophlyctoid fungi were conceived as eucarpic, monocentric, interbiotic Chytridiomycota (Rhizidiaceae, *sensu* Sparrow 1943, 1960) possessing subspherical to asymmetric sporangia, commonly with more than one discharge papilla and with several main rhiziodal axes. Characteristically each rhizoidal axis arises from a different point on the sporangium. This rhizophlyctoid assemblage at first included two groups: (1) the inoperculate members of *Rhizophlyctis* that Fischer (1892) established and Minden (1911) emended (cf. Sparrow 1943) and (2) endo-operculate (wall-like material contained within the discharge papillae) species described as *Karlingia* (Johanson 1944). Sparrow (1960) later erected *Karlingiomyces* for exo-operculate (operculum apical on the papilla) *Rhizophlyctis*-like chytrids. However, the exo-operculate condition prompted Sparrow to place *Karlingiomyces* in the Chytridiaceae (*sensu* Sparrow 1960) rather than in the Rhizidiaceae. Regardless of supposed family and order assignments, Dogma (1973) considered that the

"rhizophlyctoid alliance" contained *Rhizophlyctis*, *Karlingia* and *Karlingiomyces*. Barr (1980) later emended *Karlingia* Johanson based on zoospore ultrastructure.

Questions of validity of primary taxonomic characters for members of this "alliance" have generated disagreement. Distinguishing features for classification within this alliance include the development of the discharge papillae, the nature of operculation, and the mode of zoospore discharge. Based on Haskins' (1948, 1950) and Haskins & Weston's (1950) studies of lower Chytridiales, Sparrow (1960) considered endo-operculation an unreliable character. Sparrow believed that virtually any chytrid, especially *Rhizophlyctis*-like chytrids, could produce a so-called endo-operculum, even if typically inoperculate or exo-operculate. Sparrow (1960) thus placed the "endo-operculate genus" *Karlingia* Johanson (1944) in synonymy of *Rhizophlyctis*. On the other hand, Sparrow (1960) considered exo-operculation of major systematic importance and placed exo-operculate species of *Karlingia* in his genus *Karlingiomyces*. Karling (1966) disagreed with Sparrow and re-established *Karlingia* for rhizophlyctoid chytrids that were endo- or exo-operculate (Karling 1977a, 1977b). Karling (1966) regarded *Karlingiomyces* as a synonym of *Karlingia*. Dogma (1973, 1974) recognized all three "rhizophlyctoid" genera: *Rhizophlyctis* (inoperculate), *Karlingia* (endo-operculate), and *Karlingiomyces* (exo-operculate). In an apparent change of mind, Sparrow (1973) also recognized all three genera. Blackwell & Powell (1999) noted that species of *Rhizophlyctis* could be either inoperculate or endo-operculate, obviating the need to separate the genus *Karlingia*. They showed that *Karlingia* was based on *Rhizophlyctis rosea*, the type for the genus, and concluded that *Karlingia* should be rejected on nomenclatural grounds regardless of further morphological considerations. In the "rhizophlyctoid alliance" (Dogma 1973), only the generic names *Rhizophlyctis* (applicable, *sensu lato*, to inoperculate and/or endo-operculate taxa) and *Karlingiomyces* (applicable to exo-operculate taxa) are available to formal nomenclature (Blackwell & Powell 1999). This does not, however, answer the question of how many genera of "rhizophlyctoid chytrids" exist or the extent to which these are related. Recent evidence indicates that *Karlingiomyces* is perhaps more closely related to certain polycentric forms than to *Rhizophlyctis* (James et al. 2000), a concept we expand in the Discussion.

Inasmuch as *Karlingiomyces* Sparrow (1960) is a valid genus, it remains to be determined: which taxa should be included in it (cf. Longcore 1996), which taxa should be transferred to other genera such as *Rhizophlyctis*, and which taxa presently assigned elsewhere such as *Karlingia* should be admitted to *Karlingiomyces?* Such considerations require refinement of the understanding and limits of characters that define the genus. Is exo-operculation, for example, a truly meaningful character in classification; is endo-operculation inconsistent or inappropriate for generic characterization; and, are there other characters correlated with or additional to operculation that may be employed to strengthen the generic concept? Clearly we need to determine whether there is substantive reason to continue with the concept of the "rhizophlyctoid alliance" (sensu Dogma 1973) or if this "alliance" is merely an umbrella for morphologically more or less similar (*Rhizophlyctis*-like) taxa, some related and others not. Generic relationships and general systematic questions relating to *Karlingiomyces* are also considered in the Discussion. Our taxonomic synopsis of *Karlingiomyces* follows Sparrow (1960) with explanations of synonymy and other

taxonomic matters outlined as necessary. Our treatment differs from Sparrow's in precisely which taxa are accepted in the genus. Included taxa are presented first, followed by taxa that we consider too questionable for inclusion. Descriptions are adapted from Karling (1949, 1977b) and Sparrow (1960), but other sources were also consulted (e.g., Willoughby 1957; Dogma 1973, 1974).

Taxonomic Descriptions

KARLINGIOMYCES Sparrow
Aquatic Phycomycetes, 2nd ed., p. 559, 1960

Vegetative: Thallus usually interbiotic; rhizoidal tips penetrating the substrate. **Reproductive:** Usually monocentric, eucarpic; sporangium developed from the encysted zoospore or more rarely from the germ tube of the zoospore, forming one to several discharge papillae, each surmounted by a distinct exo-operculum; endo-opercula sometimes formed within the papilla. **Rhizoidal system:** Typically of several main rhizoidal axes, arising at different points on the sporangium, the rhizoidal axes branching. **Zoospore discharge:** Zoospores released free or as a relatively compact group in a vesicle or a gelatinous matrix, at first motionless but soon becoming active and swarming. **Zoospore microscopic:** Posteriorly uniflagellate, spherical; typically with a single large, hyaline, refractive lipid globule located laterally, toward the posterior of the cell. **Zoospore ultrastructure:** Unknown. **Resting spore:** Formed either asexually or sexually, the wall often somewhat thickened, the wall surface relatively smooth to rugose or significantly ornamented with pegs or spines; resting spore borne like sporangium on the thallus, upon germination functioning as a prosporangium. **Ecology and Distribution:** Apparently widespread in soil or water; saprophytic on chitinous, keratinous and cellulosic debris; genus known from eastern UNITED STATES, ENGLAND, AUSTRALIA, NEW ZEALAND, MAYLASIA, and INDIA; also AFRICA ("Liberia", indet. to species, cf. Dogma 1973).

Latin description of genus provided by Sparrow (1960), p. 559.

Type of genus: Sparrow (1960) designated *Karlingiomyces asterocystis* as the "species typica" of *Karlingiomyces*. This "type species" of *Karlingiomyces* was based on *Karlingia asterocysta*, described by Karling (*Mycologia* 41: p. 509, 1949). Since, according to Article 10.1 of the *International Code of Botanical Nomenclature* (ICBN, 2000), "the type of a name of a genus…is the type of a name of a species", see this further typification under *Karlingiomyces asterocystis* (Karling) Sparrow.

Key to Species of *Karlingiomyces*

1. Chitinophilic or keratinophilic; rhizoids with or without constrictions; sporangial exit papillae one to four, low and dome-shaped or conical and more conspicuous; resting spore wall "spiny" (i.e., with peg-like projections), roughened or smooth in appearance.

    2. Resting spore wall with distinct pegs or spine-like processes; rhizoids relatively thin to relatively thick-walled, but not showing appreciable constrictions; exo-operculum up to 18 μm in diameter, the rim flush on exit papilla orifice (or sporangial surface); zoospores usually less than 5 μm in diameter.

3.Resting spore ornamentations usually straight, blunt, peg-like, relatively sparse; released zoospores at first swarming in an evanescent vesicle outside the sporangium; zoospore flagellum 24-26μm in length; exit papillae often barely rising above sporangial surface; sporangia intercalary 1. *K. asterocystis*

3.Resting spore ornamentations often spiny, acute, hamate, numerous; released zoospores not contained in a vesicle, but may be temporarily immersed in extruded granular matrix material; zoospore flagellum 10-13 μm long; exit papillae usually more elevated; sporangia intercalary or stalked 2. *K. curvispinosus*

2.Resting spore wall smooth to rugose or verrucose; rhizoids thick-walled, distinctly constricted at intervals ("catenulate" appearance); exo-operculum up to 30 μm in diameter, appearing to "sit" on a raised, cushion-like area; zoospores usually more than 5 μm in diameter 3. *K. dubius*

1.Inhabiting cellulosic debris; rhizoids showing constrictions; sporangial exit papilla one or two, often becoming elongated and tube-like; resting spores smooth-walled or absent.

4.Operculum uniform in thickness (relatively thin); sporangium developing by enlargement of encysted zoospore; zoospores sometimes of two distinct sizes in a single sporangium; main rhizoid walls somewhat thickened, rhizoidal inclusions not readily evident; resting spores apparently lacking 4. *K. marylandicus*

4.Operculum distinctly thickened, especially toward the apex; sporangium developing from encysted zoospore or from a swelling of the zoospore germ tube; zoospores more uniform in size (not of two distinct size classes); rhizoidal axes relatively thin-walled, distinct globules and/or "inclusions" evident within; smooth-walled resting spores often produced 5. *K. exooperculatus*

Accepted species of *Karlingiomyces*

1. ***Karlingiomyces asterocystis*** (Karling) Sparrow, Aquatic Phycomycetes, 2nd ed., p. 560, 1960. **FIGURES 1-5**

Karlingia asterocysta Karling, Mycologia 41: 509, Figs. 9-19, 1949.

Vegetative: Thallus epibiotic on chitinous substrates. **Reproductive:** Sporangium hyaline, smooth-walled, spherical, oval, pyriform, oblong, fusiform, elongate or irregular, diameter 12-110 μm (usually toward the larger end of the range). **Rhizoidal system:** Extensive, branching, each of the main axes up to 18 μm in diameter. **Zoospore discharge:** Zoospore discharge papillae 1 to 4, low, often barely rising above the sporangial surface (more distinct, or relatively elevated, as much as 11 μm high on small sporangia), 8-14 μm in diameter; with a conspicuous hyaline area beneath, extending up to 16 μm into the sporangium; exo-operculum, convex though shallow, hyaline, diameter up to 14 μm; zoospores discharged *en masse* through opening left by expressed operculum, swarming briefly in an evanescent vesicle, then dispersing. **Zoospore microscopic:** Zoospore spherical, hyaline, 4.2-4.6 μm in diameter, with a single, relative large (0.7-1.2 μm) hyaline refractive lipid globule; flagellum 24-26 μm long. **Resting spore:** Subspherical, oval, oblong, angular or irregular, 12-30 μm in diameter, the contents hyaline; spore wall becoming dark greenish-brown, becoming more or less covered with blunt to somewhat pointed, straight or slightly curved (at the tip) peg-like "spines"; these pegs 4-8 μm in height, 2-4 μm broad at base, smooth or rarely verrucose, 22-60 per spore; pegs becoming dark greenish-brown (as the spore wall); resting spore germination not observed, though resting spores are sometimes

found in greater numbers than sporangia. **Ecology and Distribution:** Saprophytic on chitinous material, such as insect exuviae; readily baited on shrimp exoskeleton (Murray & Lovett 1966); isolated from soil and water; US, Maryland (cf. Karling 1949, Sparrow 1960), Louisiana (cf. Murray & Lovett 1966); NEW ZEALAND (Karling 1967).

Type: No type specimen, as can be determined, was specifically designated or deposited, either by Karling or Sparrow. In accordance with Articles 8 and 9 of the *International Code of Botanical Nomeclature* (ICBN 2000), an illustration may be utilized as the type. Hence, Karling's illustration (Figs. 9-19) of (the basionym) *Karlingia asterocysta* Karling (Mycologia 41: p. 507, 1949) is accepted as the holotype of *Karlingiomyces asterocystis* (Karling) Sparrow (1960). These figures are also the type of the genus name *Karlingiomyces* (cf. Article 10, *ICBN* 2000). Original material of *Karlingia asterocysta* was isolated from soil and water, Charles County, Maryland.

Discussion: Karling (1967) attempted to return this species to *Karlingia* and used the epithet spelling "*asterocystis*" (applied to *Karlingiomyces* by Sparrow, 1960); if used as a final name element (e.g., "astero-cystis"), "-cystis" is permissible in either case (Stearn 1983). Regardless, the shallow but definite exo-opercula (Fig. 1)—no endo-opercula being evident in this case—support placement of this species in *Karlingiomyces*.

2. ***Karlingiomyces curvispinosus*** (Karling) Sparrow, Aquatic Phycomycetes, 2nd ed., p. 560, 1960. **FIGURES 6-9**
Karlingia curvispinosa Karling, Mycologia 41:511, Figs. 20-35, 1949

Vegetative: Thallus epibiotic on chitinous substrates, the substrate sometimes becoming discolored. **Reproductive:** Sporangium quite variable, sessile or occasionally stalked, subspherical, oval, pyriform, obclavate, oblong, or elongate, sometimes apiculate, 8-160 μm diameter, more or less hyaline, relatively thin- and smooth-walled, occasionally with in-growths (plugs) of wall material. **Rhizoidal system:** Much branched; usually developed from several main axes (formed at different points on the sporangium), these relatively coarse in appearance, up to 16 μm in diameter. **Zoospore discharge:** Zoospore discharge papillae 1 to 3, prominent to relatively obscure, conical or low and dome-shaped; exo-operculum 6-18 μm broad, somewhat convex, thin or slightly thickened, hyaline, ephemeral after dehiscence; zoospores not contained in a vesicle upon release from sporangium, but may be temporarily immersed in a granular matrix. **Zoospore microscopic:** Zoospores hyaline, spherical, 3.8-4.2 μm in diameter, with one prominent, hyaline refractive globule 0.6-0.8 μm broad; flagellum 10-13μm long. **Resting spore:** Spherical, oval, or slightly angular to somewhat irregular, 6-21 μm in diameter, contents granular, sometimes developing a small vacuole; young resting spore often surrounded by a hyaline zone of amorphous material; pegs or spines appearing to gradually develop in this hyaline layer; pegs or spines eventually becoming numerous, covering the resting spore surface, curved (somewhat hook-like) and pointed; resting spore more rarely merely echinulate, verrucose or smooth; color of resting spore eventually greenish- or dark-amber or brownish; resting spore, functioning as a prosporangium in germination. **Ecology and Distribution:** Soil or

water; saprophytic on chitin or keratin substrates; often rapid and prolific in growth (readily cultured); resting spores sometimes imparting a distinctive greenish-amber or darkened coloration to the substrate; US, Maryland (cf. Karling 1949; Sparrow 1960), Maine (collections by J. E. Longcore, JEL 93 and JEL 223, are probably this species; however, resting spores, critical to determination, were not observed); NEW ZEALAND (Karling 1967).

Type: Karling's illustration (Figs. 20-35; Mycologia 41: p. 507 and 512, 1949) of *Karlingia curvispinosa* is accepted as the type of *Karlingiomyces curvispinosus* (Karling) Sparrow (1960). Original material isolated from soil and water in a ravine near the Monocacy River at Frederick, Maryland.

Discussion: Karling (1967) noted that New Zealand specimens of *K. curvispinosus* were identical with those from the United States, except for the "lack of a...clear zone" surrounding the developing resting spore—perhaps a "culture" or "artifact"-based observation.

3. *Karlingiomyces dubius* (Karling) Sparrow, Aquatic Phycomycetes, 2nd ed., p. 561, 1960. **FIGURES 10-14**

Karlingia dubia Karling, Mycologia 41:513, Figs. 36-51, 1949.

Vegetative: Thallus epibiotic on chitinous substrates. **Reproductive:** Sporangium subspherical, oval, pyriform or oblong, variable in diameter (15-240 μm), hyaline, smooth-walled. **Rhizoidal system:** Extensive, copiously branched; the several main rhizoidal axes each attaining a diameter of 12 to 16 μm, usually thick-walled, with a number of regular or irregular constrictions which may in places appear to virtually occlude the rhizoidal lumen (pseudo-catenulate). **Zoospore discharge:** Zoospore discharge papillae 1 to 4, low to relatively prominent, 12-34 μm in diameter, subtended by a more or less hemispherical hyaline zone that may extend as much as 16 μm into the sporangium; exo-operculum convex but shallow, 10-30 μm in diameter, relatively uniform in thickness, usually dehiscing prior to zoospore discharge and remaining for a time "perched" on hyaline material of the exit papilla (which appears as a "cushion"); zoospores eventually expressing the operculum, emerging together in a gelatinous "matrix" of material, but soon dispersing.

Zoospore microscopic: Zoospores spherical, hyaline, 6-6.5 μm in diameter, with a single, basal, relatively large (2-2.3 μm) hyaline globule apparent; flagellum long (32-35μm), but at first (upon zoospore discharge) obscure and coiled around body of zoospore, then uncoiling for motility and dispersal. **Resting spore:** Spherical, oval, elongate or somewhat angular, 8-20 μm in diameter, with relatively coarse, granular contents; resting spore wall smooth to rugose or verrucose, becoming dark brown; resting spores formed as result of sexual process (Willoughby 1957), remnants of the two fused gametangial bodies sometimes remaining evident externally on the resting spore (Dogma 1974); resting spore functioning as a prosporangium in germination (Dogma 1974). Ecology and Distribution: Soil and water; saprophytic on chitinous residue; readily baited, e.g., on shrimp exoskeleton; US, Maryland (cf. Karling 1949, Sparrow 1960); ENGLAND (Willoughby 1957, 1964); AUSTRALIA, State of Victoria (Willoughby 1965), New South Wales (recent observation P.M. Letcher); NEW ZEALAND (Karling 1967); MALAYSIA, Singapore (Dogma 1974).

Type: Karling's illustration (Figs. 36-51; Mycologia 41: p. 512, 1949) of *Karlingia dubia* is accepted as the type of *Karlingiomyces dubius* (Karling) Sparrow (1960). Original material isolated from Calvert County, Maryland.

Discussion: Some disagreement has occurred over taxonomic placement of *Karlingiomyces dubius*. Willoughby (1957) recognized Karling's (1949) *Karlingia dubia*, but confirmed the unique ("raised") exo-operculum (Willoughby's Figs. 3C,E). However, Sparrow (1960) transferred this species to his new genus, *Karlingiomyces*, on the basis of definitive exo-operculation. Although Sparrow selected *K. asterocystis* as the type of *Karlingiomyces*, it was only *K. dubius* that he elected to illustrate. Karling (1967) sought to return "*K. dubia*", among several taxa, to *Karlingia*. Dogma (1973), in a delimiting viewpoint of genera (based on differences in operculation), recognized all three genera of the "rhizophlyctoid assemblage" (*Rhizophlyctis*, *Karlingia* and *Karlingiomyces*). Dogma (1974) accepted *K. dubius* as exo-operculate, and as belonging to *Karlingiomyces*. We concur with Sparrow's and Dogma's placement of this species in *Karlingiomyces*.

4. *Karlingiomyces marylandicus* (Karling) Sparrow, Aquatic Phycomycetes, 2nd ed., p. 562, 1960. Spelling of the epithet given as "*marilandicus*" by Sparrow (1960).

FIGURES 15-17

Karlingia marylandica Karling, Mycologia 41:518, Figs. 70-78, 1949.

Vegetative: Extramatrical on cellulose substrates. **Reproductive:** Sporangium spherical, oval, pyriform, oblong to somewhat elongate or irregular, 20-85 μm in diameter, the wall smooth and often somewhat thickened (1.8-2.6 μm thick). **Rhizoidal system:** Rhizoid axes usually emerging from several different points on the sporangium (occasionally monorhizoidal, cf. Karling 1966), relatively coarse in appearance, up to 12 μm broad, irregularly constricted, the walls somewhat thickened, ultimately extensively branched. **Zoospore discharge:** Zoospore discharge papillae 1 or 2, varying from nearly sessile to elongate and tube-like (the "tube" straight, irregular or contorted), 10-15 μm broad, 10-205 μm long; exo-operculum up to 17 μm in diameter, convex, thin or uniformly thickened; endo-opercula sometimes also observed; zoospores emerging temporarily as a globular mass in a slimy matrix. **Zoospore microscopic:** Zoospores hyaline, spherical, either 5.5-6 μm or 2-3.5 μm (apparently of two sizes in the same sporangium), with a single, relatively large, hyaline refractive globule (0.8-2.8 μm broad). **Resting spore:** Not observed. **Ecology and Distribution:** Soil and water. Saprophytic on natural cellulose materials, i.e. various "vegetable debris", cellophane or bleached corn leaves; US, Maryland (cf., Karling 1949; Sparrow 1960); INDIA, Kerala State and Madras State (Karling 1966); NEW ZEALAND (Karling 1967).

Type: Karling's illustration (Figs. 70-78; Mycologia 41: p. 516, 1949) of *Karlingia marylandica* is accepted as the type of *Karlingiomyces marylandicus* (Karling) Sparrow (1960). Original material isolated from a farm near Frederick, Maryland.

Discussion: Karling (1966, 1967) did not accept Sparrow's (1960) placement of this species in *Karlingiomyces*, and continued to recognize it under *Karlingia*. Karling (1966) first accepted Sparrow's altered spelling "marilandica", then (1967) changed

it back to the original "marylandica". According to the ICBN (2000), Article 60.4, the letter "y", foreign to classical Latin, is permissible in Latin scientific names. Since an original spelling (if not demonstrably incorrect) is to be retained (Article 60.1), there is no cogent reason to alter Karling's (1949) spelling, except for a termination appropriate to the generic name *Karlingiomyces*, viz. "*marylandicus*".

Dogma (1973) limited *Karlingiomyces* to only three species: *K. asterocystis*, *K. curvispinosus*, and *K. dubius*. Dogma suggested that *Karlingia marylandica* (=*Karlingiomyces marylandicus*) was among certain *Karlingia* species that, while appearing to be exo-operculate, were actually to be considered as endo-operculate—the operculum being interpreted as slightly sunken in the apex of the discharge papilla or exit tube, as revealed by a slight "collar-like" remnant of the papillar wall remaining above the operculum. However, Karling's (1949) original description and illustration and Sparrow's re-description (1960) of *Karlingiomyces marylandicus* indicate that this species is exo-operculate, although endo-opercula may also form, even in the same exit papilla (Karling 1949) surmounted by an exo-operculum (Fig. 16). Karling (1966) reconfirmed the predominantly exo-operculate character of this taxon.

It is difficult to know whether the lack of resting spores in *K. marylandicus* is real or merely apparent. A species such as *Karlingia rosea* (=*Rhizophlyctis rosea*), though known to occasionally produce resting spores, often does not exhibit these. Sparrow (1973) related this dearth of resting spores to the capacity of ordinary sporangia of *K. rosea* to form dormant, resistant structures, serving a similar perennating function to resting spores. Only further collecting and culture will demonstrate whether a comparable situation pertains to *Karlingiomyces marylandicus*.

5. *Karlingiomyces exooperculatus* (Karling) Blackwell, Letcher & Powell, *comb. nov.* **FIGURES 18-22**

Basionym: *Karlingia exooperculata* Karling, Nova Hedwigia 28:209, Figs. 1-16, 1977b.

Vegetative: Thallus generally extramatrical on celluose debris, eucarpic, monocentric,or occasionally polycentric. **Reproductive:** Sporangia developing from the encysted zoospore or from a swelling in germ tube of zoospore, spherical, subspherical, pyriform, ovoid, obpyriform or even reniform, sometimes irregular and somewhat elongate, highly variable in size (15-252 μm in diameter), hyaline, smooth-walled, the wall somewhat thickened (1.8-2.7 μm thick). **Rhizoidal system:** Rhizoidal axes up to 12 μm in diameter, relatively thin-walled, sometimes containing hyaline and other globules or inclusions, arising at several points on periphery of sporangium, or in some cases from the base of the sporangium, eventually much branched, extending considerable distances (up to 960 μm). **Zoospore discharge:** Zoospore discharge papillae 1 or 2, low in relief to long and tube-like, straight,curved or somewhat contorted, 6-240 μm long, 3-10 μm broad; exo-operculum conspicuous, 6-12 μm broad, usually rather strongly convex, often differentially thickened toward the apex; nature of zoospore discharge not reported. **Zoospore microscopic:** Zoospores spherical, 6-7 μm; a large hyaline refractive globule apparent, 3.5-4.2 μm in diameter; flagellum long (48-55 μm). **Resting spore:** Spherical, ovoid,

oblong, or somewhat elongate, 10-28 μm in diameter, usually containing a large hyaline refractive globule; wall smooth, golden-brown, thickened (up to 4 μm), functioning as a prosporangium in germination. **Ecology and Distribution:** Saprophytic on cellulosic substrates, e.g., may be isolated on cellophane from watered soil cultures; US, Florida (Orlando), cf. Karling (1977b).

Type: Karling's illustration with analysis (Figs. 1-16; Nova Hedwigia 28: pp. 224-225, 1977b) of *Karlingia exoperculata* (the basionym, cf. Article 33.3, ICBN, 2000) may be accepted as the holotype of the new combination, *Karlingiomyces exoperculatus*. This illustration was in fact referred to by Karling as the "iconotypus". Original material of this taxon was isolated from a water culture of soil collected near Orlando, Florida.

Discussion: When Karling (1977b) described *Karlingia exooperculata* (the description we follow above) he made a point to distinguish *K. exooperculata*, from certain other "hyaline" species of *Karlingia* (e.g., *K. hyalina*), by its conspicuous external operculum. Because of the obvious exo-operculation, and the seemingly correlated feature of a spherical zoospore—containing a single, rather large, refractive globule—we believe that *K. exooperculata* is more properly placed in *Karlingiomyces*, than in *Karlingia*. As regards orthography, Longcore (1996), consistent with the citation in *Index of Fungi* (Vol. 4, p. 476), hyphenated the epithet (under *Karlingia*) as *exo-operculata*. However, Karling (1977b) employed the species epithet without a hyphen (viz. *exooperculata*); and, since non-hyphenation is generally preferable in scientific epithets (ICBN 2000, Article 60.9), we use the name without the hyphen. The termination (gender) of the epithet is, however, changed herein to *–us* (i.e., *exooperculatus*), appropriate to the genus name *Karlingiomyces*.

Excluded Taxa

1. *Karlingiomyces granulatus* (Karling) Sparrow, Aquatic Phycomycetes, 2nd ed., p. 563, 1960.
Karlingia granulata Karling, Mycologia 39: 57, Figs. 1-22, 1947.

Karling (1947) described this Brazilian species as *Karlingia* in a paper dealing with "new species with sunken opercula". Yet, Karling considered "the occurrence of exo- and endoopercula" a distinguishing character. Sparrow (1960) transferred *Karlingia granulata* to *Karlingiomyces*, presumably based on the nature of the operculum—which, though "delicate", could be considered "apical *or* submerged in the discharge papilla tube," a direct translation from Karling's (1947) Latin description. The exact position of the operculum is thus equivocal. From Karling's original figures (e.g., Figs. 9, 11), one might infer the origin as an endo-operculum, possibly near the summit of an exit papilla. Willoughby (1965), however, reported a single isolate of *Karlingiomyces granulatus* from Australia as exo-operculate. Subsequently, Dogma (1974) made a detailed study of the position of the operculum in a Colombian isolate of *K. granulata*. Dogma determined that this isolate (and presumably the species) possessed a type 2A endo-operculation (Dogma 1973, Sparrow 1973, Fig. 25) that superficially resembles exo-operculation. When zoospore discharge occurs, the operculum, initially possibly located near the apex of the discharge papilla, is pushed out

by the zoospores (released in mass), much as an exo-operculum would be. However, the development of the operculum is technically that of an endo-operculum. Dogma (1974) noted that small collar-like flaps (papillar wall remnants external to the operculum) can be seen to persist—evidence that the source of the delicate operculum is endo-papillar. Dogma (1974) accepted *Karlingia granulata* Karling, placing *Karlingiomyces granulatus* (Karling) Sparrow in synonymy.

Zoospores of *Karlingia granulata* do not appear comparable to those of the other species in *Karlingiomyces*. Zoospores of the *Karlingiomyces* species accepted are spherical and have one obvious, rather large, hyaline refractive lipid granule, usually at a rather specific position lateral or posterior in the cell. Although there has been no ultrastructural study of *Karlingiomyces* zoospores, based on light microscopy, which permits only a tentative conclusion, this type of zoospore seems consistent with what Barr (1980) called the "chytridialean" type. In contrast, the zoospore of *K. granulata*, though more or less spherical, contains a number of small refractive granules, none dominant in size or position. Sparrow (1960) emphasized the feature of numerous, coequal refractive granules in his key to alleged species of *Karlingiomyces*, as distinguishing *K. granulatus* and one other supposed species of *Karlingiomyces* (discussed below). It is possible that these two species were "distinct" within *Karlingiomyces* because of their misplacement in the genus. Thus, the zoospore of *K. granulatus* may be more consistent with types of zoospores other than chytridealean (Barr 1980, 1990, 2001; Barr & Désaulniers 1986). For the present, the only taxonomic place for *K. granulata* to reside is in the genus *Rhizophlyctis*.

2. *Karlingiomyces lobatus* (Karling) Sparrow, Aquatic Phycomycetes, 2nd ed., p. 563, 1960.
Karlingia lobata Karling, Mycologia 41: 515, Figs. 52-69, 1949.

This is an interesting taxon, often with wrinkled (older) sporangia and usually deeply lobed resting spores. However, the same points apply to *K. lobatus* as to *K. granulatus* in terms of taxonomic disposition. Known first from Maryland as *Karlingia lobata* (Karling 1949), Konno (1972) recognized it in Japan as *Karlingiomyces lobatus*. Dogma (1973), however, considered *K. lobata*, as well as *K. granulata*, to be strictly endo-operculate, in disagreement with Sparrow's (1960) conclusion of possession of both exo- and endo-opercula. As stated by Dogma (1973) the "exo-operculation is only apparent, not real". A careful study of Karling's (1949) original figures (e.g., Figs. 53, 56) suggests that Dogma's interpretation is correct,and hence this species should not be recognized in *Karlingiomyces*.

Zoospores of *K. lobata* have a number of small, variable (but more or less equal), refractive granules, indicative of a zoospore type more similar to *K. granulata* than to species included in *Karlingiomyces*. Karling (1949) noted that swimming zoospores of *K. lobata* were spherical, but that when coming to rest, "creep about", exhibiting a more elongate form with amoeboid properties. If demonstrated to be amoeboid while still in the swimming phase, this (along with the numerous, small lipid globules) would suggest the possibility of a spizellomycetalean type of zoospore (Barr 1980, 1990, 2001). As in the case of *K. granulata*, the placement of *K. lobata* would (temporarily at least) be in *Rhizophlyctis*.

Additionally, Karling (1984, *Nova Hedwigia* 40: 332) described a small-zoospored variety from Colombia, *K. lobata* var. *microspora*, bearing some resemblance to *K. granulata* (cf. Karling 1947). Regardless of its correct taxonomic placement, the zoospores (with multiple minute granules), and the exclusively endo-operculate (as stated by Karling) nature of the sporangial discharge "lid", would mitigate against including this "variety", as well as the above two species, in *Karlingiomyces*.

3. *Karlingiomyces laevis* Konno, Sci. Rept. Tokyo Kyoiku Daigaku, B, 14: 256, Plate 8 (Figs. E-H), 1972.

The presence (uniformly, as indicated by Konno) of only a single exit papilla on the sporangium of *K. laevis* does not necessilarily exclude this Japanese species from *Karlingiomyces*, although it does cast doubt on such a placement (cf. Sparrow 1960, 1973). However, *K. laevis* is described (in the figure legend), and illustrated, by Konno as (strictly) endo-operculate—and this would in fact exclude it from the exo-operculate genus *Karlingiomyces*. Konno (1972) provided a Latin diagnosis of *Karlingiomyces laevis*, disconnected from the English description (p. 256), and without reference to it, 12 pages following (p. 268). Konno (1972) described the zoospore of *K. laevis* as spherical, with a single large refractive globule. Thus, one cannot assume that all "rhizophlyctoid taxa" excluded from *Karlingiomyces* possess zoospores with multiple small globules (as in *K. granulatus* and *K. lobatus*).

In the same publication, Konno (1972) described a chytrid he commonly found in Japan, *Rhizophlyctis willoughbyi*. This species morphologically resembles *Karlingiomyces*, but the exact nature of the operculation is difficult to discern from Konno's description and illustrations. One or rarely two exit papillae are present on sporangia of *R. willoughbyi*. Regardless of its exact placement, mention here of this species, and of *K. laevis* above, addresses the issue that the number of "rhizophlyctoid" taxa (interbiotic, multiple rhizoidal axes) is probably underestimated and/or that these taxa are poorly understood (especially those bearing only a single exit papilla). More collecting, sampling, culture and descriptive work would be useful in obtaining a better appreciation of the diversity of *Rhizophlyctis*-like organisms.

Discussion

Acceptance of *Karlingiomyces* and its species depends on what is meant by "operculation" and its importance as a character in the taxonomy of Chytridiomycota. Sparrow (1933) stated that "the operculum impresses one as a morphological structure of some significance" and used it (1942, 1943) as the primary character separating groups of families in his Order Chytridiales. In emphasizing the systematic importance of operculation, Sparrow (1960) intended exo-operculation (i.e., delimiting his "operculate series" of chytrids from those that were "inoperculate"). Based partly on the work of Haskins & Weston (1950), Sparrow (1960) considered endo-operculation to be taxonomically unstable, "generically invalid" (Sparrow 1958), and possibly found in any chytrid with discharge papillae—although "rhizophlyctoid" chytrids seem prone to exhibit this feature (Sparrow 1960). Haskins

(1950) noted the erratic occurrence of endo-operculation, confusing the distinction made among certain chytrid genera (e.g., *Diplophlyctis* and *Nephrochytrium*).

Whiffen (1944) rejected operculation as a major character in chytridiomycete taxonomy. Instead she considered developmental pattern as a primary character, such as whether the encysted zoospore body or the germ tube gave rise to the sporangium or prosporangium. In *Karlingiomyces* sporangial development is directly from the zoospore; although, in *K. exooperculatus*, the sporangium may also arise from the germ tube. Powell & Koch (1977) showed a similar dual thallus developmental sequence, also within a single species, *Entophlyctis variabilis* (= *Powellomyces variabilis*, Longcore et al. 1995), casting doubt on the broader taxonomic significance of such a developmental character. Whiffen (1944), however, also emphasized other characters, i.e., whether a chytrid was holocarpic or eucarpic, and whether the thallus was monocentric or polycentric. Sparrow (1943, 1960) accorded importance to both of these latter characters. Barr (1980) felt that nuclear migration (whether the nucleus migrates into a germ tube or remains in the zoospore cyst) was a significant feature, supportive of Whiffen's developmental emphasis. But, Barr found a certain taxonomic consistency also in operculation. Barr's Chytridiales (defined by zoospore type) contained both operculate and inoperculate forms; yet, all operculate forms were found to belong to this order, not to the Spizellomycetales. Thus, Barr's findings support in part Sparrow's emphasis on operculation, as well as Whiffen's emphasis on development.

Karling (1937) also considered operculation to be systematically significant in chytrid taxonomy (e.g., *Endochytrium*), but eventually (1966, 1977) adopted a broad, less critically demanding, position—that operculation (exo- or endo-) was important, regardless of its exact nature. Karling recognized *Karlingia* Johanson as embracing both endo-and exo-operculate forms, seeing no need to recognize *Karlingiomyces* Sparrow as a separate "exo-operculate" genus. Dogma (1973) attempted to resolve this issue by recognizing an inoperculate category (e.g., certain *Rhizophlyctis* species), two categories of endo-operculate organisms (*e.g.*, various *Karlingia* species), and an exo-operculate category (*Karlingiomyces*), within the rhizophlyctoid assemblage of chytrids (Figs. 23-26). However, the taxonomic significance of an endo-operculum is suspect. Chambers & Willoughby (1964) indicated that in *Rhizophlyctis* (*Karlingia*) *rosea* endo-opercula sealed off the bases of open exit tubes. Correspondingly, Powell (1976) showed, in an ultrastructural study of *Entophlyctis*, that the endo-operculum is formed as a kind of wound response, after the opening of the exit papilla and release of the apical discharge plug—in other words, it is environmentally induced. Similarly, in *Rhizophlyctis rosea*, Powell (1994) demonstrated that the "endo-operculum" is a late addition of wall material covering exposed protoplast inside the papilla. These observations are consistent with statements by Sparrow (1960), Haskins (1948, 1950), and Haskins & Weston (1950). Endo-operculation is probably at least partly artifactual. Exo-operculation appears to have the sounder morphological and genetic basis. Hence, there is (to the reverse of Karling's opinion) a substantial reason for recognizing the exo-operculate genus *Karlingiomyces*, and distinguishing it from *Rhizophlyctis* (which is variously inoperculate or "endo-operculate"), but not for recognizing *Karlingia* (which is allegedly "endo-operculate", and nomenclaturally defunct in any case, cf. Blackwell

& Powell 1999). Our decision to include species in *Karlingiomyces* (*K. asterocystis, K. curvispinosus, K. dubius, K. marylandicus, K. exooperculatus*) hinges on the fact that a distinct exo-operculum has been demonstrated in each case.

Salkin (1970) noted that a similar exo-operculate genus, *Allochytridium*, resembled *Karlingiomyces* in sporangial morphology and in the often constricted, catenulate, rhizoids. Salkin suggested distinguishing *Allochytridium* from *Karlingiomyces* by a difference in the range of number of discharge papillae and by specific aspects of sporangial development. Sparrow (1973), in apparent agreement, separated the two genera by the key character of "sporangium with one discharge papilla" (*Allochytridium*), and "more than one" (*Karlingiomyces*). However, Salkin's (1970) original statement was that *Karlingiomyces* possessed "one or more discharge papillae", and *Allochytridium* "usually one discharge papilla". It is clear, however, that additional distinctions exist. The sporangium of *Karlingiomyces* typically develops by expansion of the zoospore cyst (in *K. exoperculatus* also by expansion of the germ tube); whereas in *Allochytridium* the sporangium consistently develops from the germ tube and, most significantly, eventually fuses with the zoospore cyst (Salkin 1970, Sparrow 1973, Karling 1977a). Whereas Powell & Koch (1977) pointed out that similar (not necessarily identical) thallus variations could occur within chytrid taxa, we nonetheless believe (as did Karling 1977a) that *Allochytridium* is distinct from other "rhizophlyctoid" genera (including *Karlingiomyces*) when total characters are considered. An additional potential distinguishing character in *Allochytridium* is that although several rhizoid groups arise at different points on the sporangium, as in *Karlingiomyces*, the initial grouping arises "from the endobiotic tip of the germ tube" (Salkin 1970, Karling 1977).

Although our decision to include species of "rhizophlyctoid morphology" in *Karlingiomyces* is based on the presence of an exo-operculum, one should not ignore zoospore type. Barr (1980) determined that two fundamentally different types of zoospores were represented in the former Chytridiales (cf. Sparrow 1960): (1) those characteristic of the Spizellomycetales (Barr 1980), and (2) those characteristic of the taxonomically narrowed Chytridiales (*sensu* Barr 1980). Barr (1990, 2001) extended his coverage to all groups of Chytridiomycota, and included additional zoospore types characteristic of orders such as Blastocladiales and Monoblepharidales. Barr's (1980) distinction of fungal zoospore types (spizellomycetalean and chytridialean), and Barr & Désaulniers' (1986) work on zoospore subtypes within the *Rhizophlyctis/Karlingia* complex, are especially pertinent to our study. Although distinctions of these zoospore types are mainly ultrastructural, certain critical features are, fortunately, also often discernible with the light microscope (Barr 1980). The morphology and structure of the zoospore, as visible with the light microscope, of all *Karlingiomyces* species accepted in our study are of an apparently chytridialean type, or are perhaps referable to subtype "D" of Barr & Désaulniers (1986). In either case, they are spherical and have a single large refractive lipid globule positioned posteriorly or laterally in the cell. Zoospores of certain alleged *Karlingiomyces* species that we do not include in the genus (*K. granulatus, K. lobatus*) appear to have multiple small lipid globules and other features, such as shape or movement, not consistent with included species. Our primary reason for excluding these taxa was, however, not the zoospore, but that they are apparently endo-operculate rather than

exo-operculate. Although our decision for inclusion is based on exo-operculation, a certain zoospore type (chytridialean, a tentative conclusion based light microscopic features) appears to be consistent with included (exo-operculate) species. The zoospores of all alleged species of *Karlingiomyces* should be investigated with TEM. We note Barr's (2001) caveat that distinctive zoospore types occur within the order Chytridiales. As James *et al.* (2000) indicated, chytrids with chytridialean zoospores (*sensu lato*) may eventually be placed in several different, even disparate, clades of Chytridiomycota. Certainty of relationship cannot be asserted on the basis of general zoospore type, especially at the light microscopic level.

Most chytrid systematists (e.g., Sparrow 1943, 1960, Whiffen 1944, Karling 1977a, Barr 1980) have considered the monocentric/polycentric distinction as significant at one taxonomic level or another. The majority of chytrids (including *Karlingiomyces*) are monocentric. Polycentric chytrids have been classified in one (Whiffen 1944) or

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Figs. 1-5. *Karlingiomyces asterocystis*. 1. Multi-operculate zoosporangium. 2. Early stage of vesicle discharge from sporangium with exo-operculum visible at the right of the discharge plug. 3. Mass of zoospores emerging into an exogenous discharge vesicle. 4. Mature resting spore covered with peg-like processes. 5. Spherical zoospore with single lipid globule.

Figs. 6-9. *Karlingiomyces curvispinosus*. 6. Zoosporangium with multiple rhizoidal axes, typical of interbiotic growth habit. 7. Zoosporangia (zoospores evident within), showing exit papillae; sporangium on right is stalked. 8. Release of zoospores into a densely granular matrix. 9. Mature resting spore, and variously shaped pegs or spines that ornament the wall.

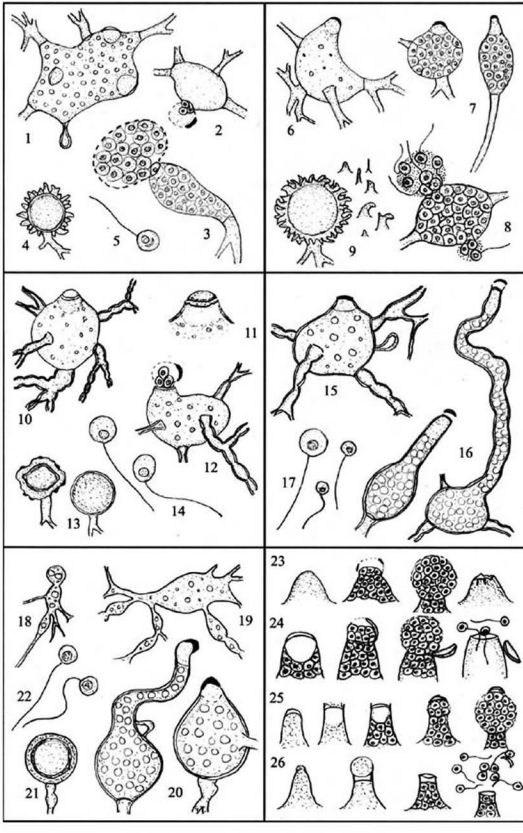
Figs. 10-14. *Karlingiomyces dubius* 10. Zoosporangium bearing thick-walled, irregularly constricted rhizoidal axes. 11. Exit orifice and detached (but still "perched"), relatively broad, exo-operculum. 12. Initial zoospore discharge with exo-operculum pushed to the right of the discharge plug matrix. 13. Mature resting spores; on the one on the left, remnants of both putative gametangial bodies evident, the wall verrucose; the one on right exhibits the more typical smooth wall. 14. Characteristic, relatively large zoospores; conspicuous hyaline globule evident toward posterior of the cell.

Figs. 15-17. *Karlingiomyces marylandicus*. 15. Zoosporangium with thickened wall and irregularly constricted rhizoids. 16. Zoosporangia with elongate exit tubes, each with a shallowly convex, exo-operculum (detached); endo-operculum evident near apex in sporangial exit tube on right. 17. Two sizes of zoospores, each with a single hyaline globule.

Figs. 18-22. *Karlingiomyces exooperculatus*. 18. Germinated zoospore with enlargement evident in germ tube. 19. Stage in sporangium development. 20. Pyriform sporangium (right); sporangium with elongate exit tube (left); note thickened exo-operculum. 21. Smooth, thick-walled resting spore containing a single, large refractive globule. 22. Zoospores, each with a single, posteriorly oriented, refractive globule.

Figs. 23-26. Developmental stages of discharge papillae for a variety of sporangial discharge forms. 23. Zoospore discharge from an inoperculate papilla. 24. Dehiscence and discharge from an exo-operculate papilla. 25. Endo-operculate discharge papilla from which zoospores are released *en masse* and passively. 26. Endo-operculate discharge in which zoospores are explosively released.

*Drawings adapted, with permission of the journal, from Karling (1949, 1977b) and Dogma (1973).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two (Sparrow 1960) families of their own. However, the systematic distinction between monocentric and polycentric chytrids may blur with additional knowledge. Blackwell et al. (2002) noted that a presumed monocentric form such as *Chytridium* (*Diplochytridium*) *lagenaria* may develop endobiotically a polycentric aspect. A monocentric chytrid, in which the nucleus migrates from the zoospore cyst or sporangium (Barr 1980) into the rhizoidal system, is capable of developing additional reproductive bodies and thus becoming to an extent polycentric. Powell & Koch (1977) showed, in *Entophlyctis variabilis* (=*Powellomyces variabilis*), that zoospore cysts with migrating and non-migrating nuclei may occur in the same taxon. The polycentric condition probably arose, independently, a number of times in the Chytridiomycota, the monocentric condition being the putative initial state in each instance. An example of a progression from monocentric to polycentric thalli may be seen in *Catenophlyctis* (Karling 1965). Karling (1977a, 1977b) noted that in certain species of *Karlingia* (=*Rhizophlyctis*) the thallus, usually monocentric, may occasionally become polycentric. *Karlingiomyces* may also prove interesting in this regard, in that at least one species (*K. exooperculatus*) may likewise develop in a polycentric fashion (Karling 1977b). *Karlingiomyces* is especially intriguing because, based on molecular data (James *et al.* 2000), it did not cluster (if the isolate utilized in the study was truly *Karlingiomyces*) with forms of superficially similar morphology, such as *Rhizophlyctis rosea*, but with the "*Lacustromyces* clade". The *Lacustromyces* clade contains polycentric forms such as *Polychytrium*, and the relatively recently described polycentric genus *Lacustromyces* (Longcore 1993). The generally polycentric "*Nowakowskiella* clade" (James *et al.* 2000), also including *Allochytridium*, might also be explored for possible relationships with *Karlingiomyces*. Further developmental, ultrastructural and molecular studies should clarify the relationships of *Karlingiomyces* and its overall systematic position.

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A note on some morphological features of *Chorioactis geaster* (Pezizales, Ascomycota)

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Abstract–A study of *Chorioactis geaster* (Sarcosomataceae) has shown the presence of several unreported or unconfirmed characters for this unusual and rare operculate discomycete. The ascospores are ornamented, they mature more or less simultaneously in all asci of a single ascoma, and asci have a thin hyphal base. The species is compared with species of the genera *Cookeina* and *Microstoma* (Sarcoscyphaceae) that also have this character. SEM shows open asci have a two-layered opercular region confirming TEM reports of differentiated wall layering in this region of the ascus. These features are discussed and the isolated systematic position of *Chorioactis* suggested by previous studies is confirmed.

Key words–Ascus morphology, ascospore maturation, spore ornamentation

Introduction

Recently we showed that *Chorioactis geaster* (Peck) Kupfer ex Eckblad populations in Japan and North America represent distinct but closely related lineages. Molecular clock estimates suggest that they have probably been separate for at least 19 million years (Peterson et al. 2004). In the course of that study we examined a number of collections and determined that morphologically we could not distinguish the North American and Japanese collections. Our detailed studies, however, uncovered morphological features of the species that had not been noted previously. These observations are reported here.

The background and history of the genus *Chorioactis* Kupfer ex Eckblad was reviewed by Peterson et al. (2004). Previously considered to be a member of the Sarcosomataceae (Korf 1973), *C. geaster* was shown by Harrington et al. (1999) to be part of a weakly supported clade including species in the genera

Desmazierella Lib., *Neournula* Paden and Tylutki, and *Wolfina* Seaver ex Eckblad. These species have dark, roughened, superficial hairs on the outer surface of the ascomata (Eckblad 1968, Kupfer 1902). Unlike the central species of the Sarcosomataceae, however, their hymenia are not black but range in color from tan to butterscotch to orange. In this study we report our observations on *C. geaster*, especially those related to ascospore surface ornamentation and ascus morphology.

Materials and Methods

Material was studied using free-hand sections of fresh or dried ascomata. Portions of ascomata were rehydrated in tap water. Measurements and initial observations employed sections mounted in tap water. Subsequently, material was mounted in Congo Red in ammonia and cotton blue in lactic acid following Hansen et al. (2001). An Olympus BH-1 microscope was used for observations and photographs. Scanning Electron Microscopy (SEM) was done on an AM-RAY model 1000 SEM. Portions of hymenia of mature ascomata were mounted on stubs and sputter-coated with gold-palladium alloy.

Observations

Ascospore surface — Ascospores of *C. geaster* consistently have been described as smooth (Heald and Wolf 1910, Imazeki and Otani 1975, Seaver 1928, 1942). Close examination with the light microscope and subsequent SEM studies show that the spores are minutely punctate (Fig. 1 A-D). In a TEM study Bellèmere et al. (1994) indicated that the ascospore surface was marked, "Sa surface externe, irrégulière, forme de nombreuses petites protubérances ornementant." Under the light microscope cyanophilic markings are detected in both fresh and dried material but they are particularly evident in freshly discharged ascospores. The markings are low and are not visible in optical sections. Additionally, they may be overlooked because they are obscured by the refractive, granular contents of the ascospores; these are lipids according to Bellemère et al. (1994).

Ascus maturation — In our study we found that all asci within a single ascoma had ascospores at more or less the same stage of development, that is, the ascospores develop synchronously. Synchronous ascospore maturation also is found in species in the genera *Cookeina* Kuntze and *Microstoma* Bernstein, in the family Sarcoscyphaceae. Further, we noted that the asci of *C. geaster* have a further shared feature with species of these two sarcoscyphaceous genera. Asci are constricted abruptly at the base (Fig. 2 A)

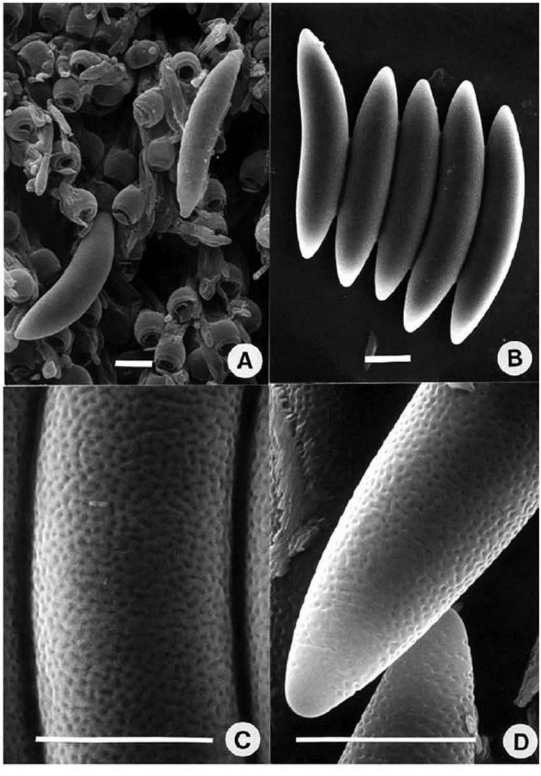

Fig. 1 A – D. SEM photographs of ascospores of *C. geaster*, scale bars in all figures = 10 μm. A. Ascospores lodged on the hymenial surface; note asci with prominent operculа. x 1000. B. Discharged ascospores, x 1200. C and D. View of surfaces of ascospores showing punctate wall ornamentation, x 5000.

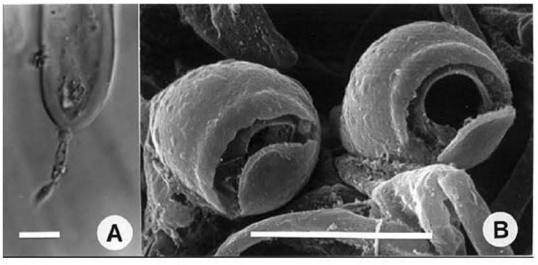

Fig. 2 A – B. Asci of *C. geaster*, scale bars in both figures = 10 μm. A. Light microscopic view of ascus showing hyphal base. B. SEM photograph of open asci, note opercula and the distinct wall layers at the region of dehiscence.

rather than tapering gradually as is the case with most other members of the order Pezizales. Imazeki and Otani (1975) describe and illustrate this feature with no comment. Similar bases are known in the Pezizales only in *Chorioactis geaster* and the previously mentioned members of the Sarcoscyphaceae. In some cases the hyphal base of the asci of *C. geaster* showed a thickened area or a globose intercalary swelling. Phylogenetic studies have placed *Cookeina* and *Microstoma* together in the family Sarcoscyphaceae; *Chorioactis* is only distantly related to these genera (Harrington et al. 1999). Although this ascus character may be useful in characterizing genera, it does not have broad phylogenetic value.

Operculum construction — The ascus tip opens via a thick-walled, somewhat eccentrically placed operculum. Our SEM studies indicated that a distinctive two-layered ring zone develops at dehiscence (Fig. 2 B). This configuration is evident in light microscopic examinations as well as in SEM. In TEM studies Bellemère et al. (1994) showed that the ascus lateral wall layering in *C. geaster* differs from other member of both the Sarcosomataceae and the Sarcoscyphaceae and suggested a somewhat isolated position for *C. geaster*. Additional evidence of such a view has been supported by molecular phylogenetic analyses (Harrington et al. 1999). We continue to work on the resolution of the placement of *Chorioactis* and its close relatives, *Desmazierella, Neournula* and *Wolfina*.

Acknowledgments

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MYCOTAXON

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Hypogeous fungi from the southeastern United States 3. The genus *Macowanites*

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Abstract—*Macowanites arenicola* is proposed and described as a new species. Information is provided on the ecology, morphology, anatomy and comparisons are made with related agaricoid taxa.

Key words—ectomycorrhiza, taxonomy, gasteroid, *Russulales*, Florida

Introduction

The genus *Macowania* was erected by Kalchbrenner (1876a) and emended by Kalchbrenner (1876b) to *Macowanites* to include fungi that have gasteroid basidiocarps resembling *Russula* that lack latex and possess a well-developed stipe-columella. Approximately 30 species have now been described (Singer & Smith 1960; Smith 1963; Beaton, Pegler & Young 1984; Cázares & Trappe 1991; Llistosella & Vidal 1995; Bougher & Syme 1998; Lebel & Trappe 2000; Calonge & Vidal 2001; Vidal, Calonge & Martín 2002) most of them from the western United States, Australia and New Zealand, although many more remain to be formally written up for publication from Argentina, Chile and elsewhere. No species have been recorded previously from the southeastern United States.

As a result of yearly visits to the coastal scrub habitat of northwestern Florida, Donna Mitchell and Bill Roody, two outstanding amateur mycologists from West Virginia, encountered a *Macowanites* species that had not been previously described. Interestingly, three species of *Macowanites* have recently been found in similar coastal dune habitat in Spain (Calonge & Vidal 2001; Vidal et al. 2002).

Materials and Methods

Methods of collection, terminology used, and classification were generally those of Singer and Smith (1960). Basidiospore size and shape were determined from optical sections in side view with the apiculus clearly evident and excluding the ornamentation. Color names and descriptions were taken from the Methuen Handbook of Colour (Kornerup & Wanscher 1981). Herbarium

names are abbreviated according to Holmgren, Holmgren & Barnett (1990). Fresh and dried specimens were examined with standard microchemical reagents useful in the genus (Singer & Smith 1960).

Taxonomic Description

Macowanites arenicola S.L. Mill. & Mitchell **FIGURES 1–2, 3–8**

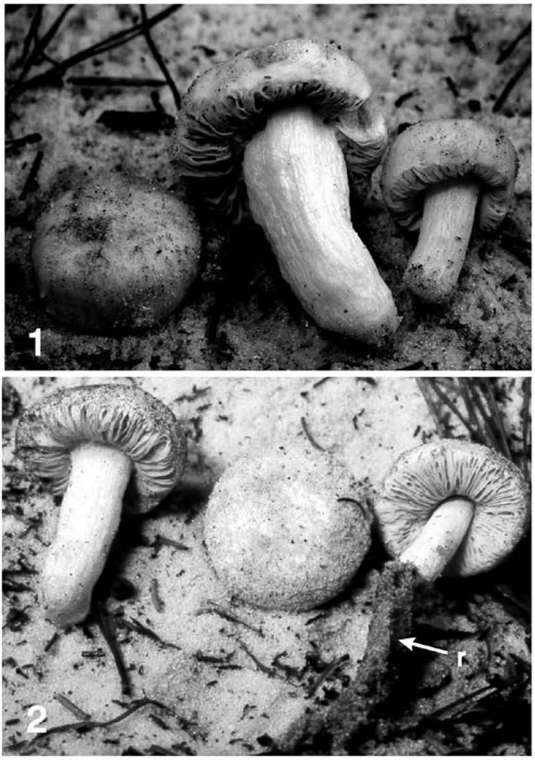
Basidiomata gregaria, hypogaea ad erumpentia, stipitata. Pileus 2–2.5 cm in diam., laevis, viscidus vel siccus, granis arenae adhaerentibus conspersus, albidus ad pallide rosaceo-aurantiacus, maculis luteolis notatus. Stipiti-columella percurrens, 1.5–2.5 x 0.5–1.2 cm, alba, interdum canescens, ubi contusus immutabilis; extensio basis stipitis e mycelio, arena, radicibusque constans, facile disrupta; contextus albus solidus demum loculatus vel cavus. Gleba convoluta vel in parte superiore loculata, sublamellata ubi exposita prope marginem, luteola ad flava. Sporae 8.8–10.4 (10.8) x 6.8–7.2 µm, Q = 1.37, late ellipticae ad ellipticae, heterotropicae, appendiculo hilari, ornatae reticulo tenui fracto e connectivis humillibus saepe cristatis et verrucis humillibus 0.2–0.4 µm altis constanti, plaga parva irregulari, saepe ad collum ad basin appendiculi hilaris redacta vel nulla.

Fructificat in arenis albis quartziticis Floridae a mense Decembri ad Februarium in sylvis apertis pini quercusque cum Pino clausa, P. elliottii, Quercu myrtifolia, Q. geminata et Q. virginiana. Holotypus: Florida, Comitatus Gulf, Caput San Blas, die 8 Februarii 1999, a Donna Mitchell et Bill Roody lectus DMFL 99-3 (RMS).

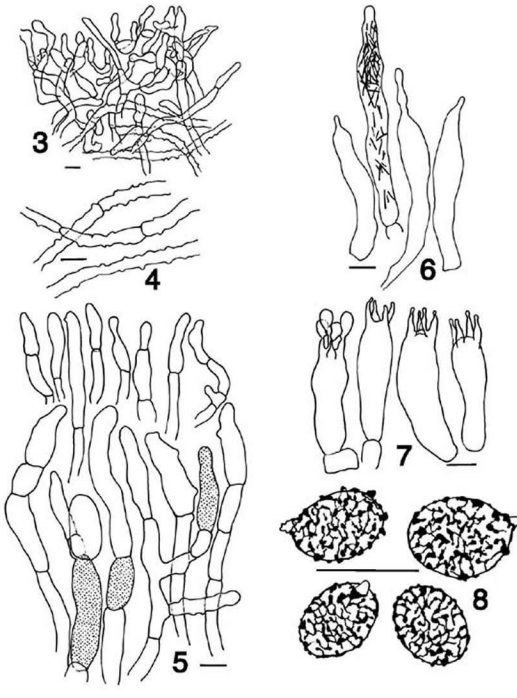
Etymology: Latin (*arenicola*, sand-dwelling), referring to the white quartz sand common in the coastal scrub forests of northwestern Florida were it grows.

Basidiocarps: gregarious, hypogeous to erumpent, stipitate. **Pileus** 2–4.5 cm diam, convex to broadly convex, margin decurved or rarely involute; smooth, tacky or dry, with adherent granules of sand, not touching stipe but occasionally covering gleba in places, peeling one-half to completely; whitish to pale buff (4B–3), occasionally mottled with yellowish white (4A-2) to pale yellow (4A–3) or rarely pinkish (9A–3 to 9B–3). **Stipe-columella:** percurrent, cylindrical or tapering slightly toward the base, 1.5–2.5 x 0.5–1.2 cm, low longitudinal ridges, white, occasionally with greyish cast to the ridges, unchanging when bruised; extension of stipe base consisting of mycelium, sand and ectomycorrhizal roots, easily disarticulated; context white, initially solid, then chambered or hollow. **Gleba:** convoluted to loculate in the upper zone, sublamellate or rarely lamellate where exposed near the margin, pale to light yellow or pale ochraceous (4A3–5). **Odor:** of old yogurt or soy sauce. **Taste:** mild. **Macrochemical reactions:** not noted.

Spores: 8.8–10.4 (10.8) x 6.8–7.2 µm (excl. orn.), Q= 1.37, broadly elliptical to elliptical, mostly heterotropic, with a hilar appendage. Ornamentation a fine broken reticulum of low often cristate connectives and low verrucae 0.2–0.4 µm high, plage small, irregular, often reduced to a collar at base of hilar appendage or absent, hyaline to pale yellow in KOH, ornamentation amyloid in Melzer's reagent, hilar appendix 3–4 x 2.5–4.0 µm, conical, often with a ragged terminal hilar scar. **Hymenial elements**: basidia 42–52 x 7–9 µm, clavate, bearing 4 spores, sterigmata 4–6 µm long, tapering toward the apex, usually curving; macrocystidia scattered but moderately abundant in localized patches, 80–120 x 6–7.5 µm, mucronate to ventricose-rostrate, with short simple



Figs. 1–2. *Macowanites arenicola* (FLDM99–3), x 2. 2. *Macowanites arenicola* (DMFL–2), x 2.5. The arrow at "r" denotes an extension of the stipe base consisting of mycelium, sand and ectomycorrhizal roots (photo by Bill Roody).



Figs. 3–8. Microscopic spore and peridium features of *Macowanites arenicola* from the southeastern United States. Scale bars = 10 μm. 3. Pileipellis *(DMFL–2)* composed of large articulated dermatocystidia and smaller hyphal end cells. 4. Ornamented hyphae frequent in the subpellis *(FLDM 99–13)*. 5. Elements of the pileipellis. 6. Pleuromacrocystidia. Crystals in second cystidium from the left are indicative of sulfovanillin staining. 7. 4-sterigmate basidia. 8. Basidiospores with fine amyloid reticulate ornamentation and plage reduced to amyloid collar on hilar appendage or absent.

or long strangulated rostrum, hyaline in KOH, dark grey-black in sulfovanillin with crystalline contents. **Hymenophoral trama:** regular or nearly so, of interwoven, subparallel hyphae 3–7 μm diam with large inflated, isodiametric, thin-walled cells 15–20 μm scattered throughout, laticiferous hyphae infrequent 7–12 μm diam, cylindrical to swollen and contorted, thin-walled, with yellowish refractive contents in KOH. **Subhymenium:** subcellular to cellular, 1–2 cells thick, cell diam. 12–20 μm. **Pileipellis**: Epicutis 190–250 μm thick, a trichodermial layer of branching interwoven hyphae in a gelatinous matrix, terminal cells clavate, obclavate to ventricose, 2.5–3 μm, mixed with large articulated dermatocystidia, terminal cells cylindrical, claviform to obclavate or ventricose, 5–7 μm, frequently with irregular short pseudobranches; subcutis not well defined, containing periclinally arranged hyphae 3–4 μm, with irregular wall processes, laticiferous hyphae infrequent, 7–12 μm diam. **Context**: of pileus and stipe heteromerous with nested sphaerocytes up to 50 μm.

Habitat and season: Fruiting in the white quartzite sands of the Florida northwestern coastal scrub forests from December to March associated with open *Pinus* and *Quercus* woodlands containing *Pinus clausa, P. elliotii, Quercus myrtifolia, Q. gemata* and *Q. virginiana*.

Material examined—USA. HOLOTYPE: Florida: **Gulf Co.**, Cape San Blas, 8 February 1999, Donna Mitchell and Bill Roody DMFL 99-3 (RMS); other collections: **Gulf Co.**, Cape San Blas, 16 January 2000, Donna Mitchell and Bill Roody 2610, (RMS, DEWV); 20 February 2000, 2612 (DEWV); 25 January 2001, 2611 (DEWV); 3 February 2001, 2613 (RMS, DEWV); **Franklin Co.**, Magnolia Cemetery, Apalachicola, 26 December 2002, Donna Mitchell and Bill Roody 3633, (RMS, DEWV); **Bay Co.**, Crooked Island, 27 January 2003, Donna Mitchell and Bill Roody 3632, (DEWV).

Discussion

Macowanites arenicola is locally common in the early winter months in the coastal scrub forests of northwestern Florida. It can be found below the surface or may be erumpent through the sand. This fungus is undoubtedly ectomycorrhizal, as the extension of the stipe columella often contains ectomycorrhizal rootlets along with masses of mycelia and sand, but it is unknown whether it is associated with both pines and/or the live oaks in the area.

The distinction between *Macowanites* and *Russula* is tenuous at best. The chambered, loculate or "gasteroid" condition of the hymenophore is the single character that is most useful in separating the genera, however there is a great deal of plasticity in this character in both genera. Because of ongoing molecular studies in the *Russulaceae* which suggest that gasteroid taxa do not appear in all infrageneric taxa of *Russula*, we prefer to place this fungus in the genus *Macowanites* for the present time. *Macowanites arenicola* collections observed in the drier growing season of 2003 tended to be much more lamellate than those observed at the same collecting locales in previous years. A similar condition has been observed in *M. messapicoides* Llistosella & Vidal and *Russula messapica* Sarnari (Martín, Högberg & Llistosella 1999). Molecular evidence (Miller *et al.* 2001) indicates that *Macowanites* is polyphyletic, with different species aligning with disparate clades representing well accepted infrageneric taxa within *Russula*.

Based on microscopic characters, *M. arenicola* would be placed in the subsection *Cupreinae* of Bon. The base tissue of the pileipellis is composed of narrow, branching hyphae with tapering or ventricose terminal cells. Mixed through this basic ground tissue are the distinctive large multicelled or articulated dermatocystidia that subtly stain greyish in sulfovanillin and frequently have irregular knobby projections. Microscopically, the pileipellis is similar to *R. cupreola* Sarnari. The relatively thin spore wall with ornamentation composed of scattered verrucae and fine, incomplete and cristulate reticulum is typical for many members of the *Cupreinae* except that the plage is somewhat underdeveloped in *M. arenicola*. Other members of the *Cupreinae* such as *R. cuprea* typically have ornamentation composed of isolated verrucae or warts with a well developed, darkly amyloid plage.

Acknowledgements

Many thanks go to Donna Mitchell and Bill Roody who collected this fungus and brought it to my attention. Thanks to Dr. Scott Redhead and Michael Castellano for valuable suggestions on the manuscript. Portions of this study were supported by grants from the National Science Foundation, DEB-0315607 and DEB-0315613, and USDA Competitive Grants 03-01542. Mark Garland provided valuable corrections for the Latin diagnoses.

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Lactarius in Kumaon Himalaya 2: New and interesting species of subgenus *Plinthogali*

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Abstract—*Lactarius montoyae* proposed as new to science and *L. lignyotus* var. *canadensis* reported for the first time from India are described and illustrated here.

Key words—Macrofungi, *Russulaceae*, taxonomy, ectomycorrhizae, India

Introduction

The subgenus *Plinthogali* (Burl.) Hesler & A.H. Smith, a well defined group in the genus *Lactarius*, is characterized as follows: pileus dry, velvety to unpolished, typically blackish, fuscous, date-brown, alutaceous, dingy buff or dull white; latex milk white, cream or rarely brown, unchanging or changing to reddish cinnamon, vinaceous red, reddish brown, lilac or violaceous or staining injured areas with these colours; true cystidia mostly absent; pileipellis a trichoderm, less frequently a cellular subpellis and with a turf as suprapellis. Hesler & Smith (1979) divided the subgenus into two sections, viz., sect. *Plinthogali* and sect. *Fumosi* Hesler & A.H. Sm. Practically, this classification was rather artificial and quite inapplicable. Further, Verbeken (2000) during the studies on tropical African mycoflora emended the definition of the subgenus and divided it into three sections, viz., sect. *Nigrescentes* Verbeken (so far comprising only African representatives), sect. *Pseudofuliginosi* Verbeken and sect. *Plinthogali*. During the macrofungal investigation in Kumaon, Himalaya, authors collected two interesting taxa of this subgenus under the sect. *Plinthogali*. One appears as an undescribed taxon and is proposed here as *Lactarius montoyae*, whereas, another one, *L. lignyotus* var. *canadensis* is not yet recorded from India.

Materials and Methods

The present communication is based on the repeated surveys undertaken to Pindari and Sunderdhunga Glaciers and surrounding forests of Kumaon Himalaya during September-October in 1999 and 2003 respectively.

Macromorphological characters were noted from the fresh specimens in the field. Micromorphological characters were studied with the dried material mounted in 5% KOH, Melzer's reagent, Lactophenol-cotton blue or carbol fuchsin. Colour terms mentioned are from Kelly & Judd (1955). All the line drawings were made by K. Das. Microscopic line drawings were prepared with the aid of the camera lucida attachment at original magnification of 1500x for basidiospores, 1000x for cystidia and basidia, 500x for other microstructures. Density of lamellae was measured at the margin excluding lamellulae. Basidiospore length excludes the length of ornamentation. Basidium length excludes the length of sterigmata. For measurement of basidiospores, 20 basidiospores were observed. Quotient (Q = L/W) was calculated considering the mean value of length and width. Different terms regarding cystidia are after (Heilmann-Clausen et al., 1998). Herbarium name used is following Holmgren et al. (1990).

Results

Including the two taxa discussed here, subgenus *Plinthogali* is represented by 12 taxa (Atri et al. 1994, Das & Sharma 2002) in India. Among them only five i.e. *L. subisabellinus* var. *murrillianus* (Hesler & A.H. Sm.) Hesler & A.H. Sm., *L. picinus* Fr., *L. gerardii* Peck var. *subrubescens* (A.H. Sm. & Hesler) Hesler & A.H. Sm., *L. montoyae* and *L. lignyotus* var. *canadensis* are hitherto known from Kumaon Himalaya.

Provisional key to the species from Kumaon Himalaya

1a. Pileipellis colour light brown, pileipellis not distinct in two layers; fresh cut lamellae unchanging ..... *L. subisabellinus* var. *murrillianus*

b. Pileipellis colour gray to dark gray yellowish brown, grayish to deep grayish brown, brownish black, dark olive brown, pileipellis of distinctly two layers; fresh cut lamellae changing or unchanging 2

2a. Spore ornamentation 2–2.7 μm high; never reticulate ..... *L. montoyae*

b. Spore ornamentation not more than 2 μm high, forming incomplete to almost complete reticulum 3

3a. Pileus papillate; pleuromacrocystidia present ..... *L. lignyotus* var. *canadensis*

b. Pileus not papillate; pleuromacrocystidia absent 4

4a. Pileus areolate at maturity; lamellae subdistant to distant, medium pink on bruising ..... *L. gerardii* var. *subrubescens*

b. Pileus never areolate; lamellae rather crowded, dark reddish brown on bruising ..... *L. picinus*

Lactarius montoyae K. Das & J.R. Sharma *sp. nov.* **Figure 1, a–h**

Etymology: In recognition to Dr. L. Montoya for her contributions to the genus *Lactarius*.

*Pileus 30–60 mm diam., convexus, planoconvexus ad applanatus, leviter depressus in centro, interdum umbonatus, atrobrunneus, margine leviter decurvato. Lamellae adnatae ad subdecurrentes, distantes, pallide ochraceae ad luteae. Stipes 25–58 x 7–12 mm, cylindratus, pileo concoloratus. Sporae in cumulo pallide ochraceae ad luteae, globosae ad late ellipsoidae, amyloideae, alatae, cristis usque ad 2.7 µm altis ornatae. Pleuromacrocystidia absentia. Pleuropseudocystidia abundantia. Paracystidia 45–56 x 8–11 µm, subclavata. Pileipellis bistrata; elementa suprapellis cylindrata, longa, 18–65 x 4–9 µm; subpellis ex cellulis sphaeris. INDIA, Uttaranchal, Bageswar, Dhakuri, September 29, 1999, leg. K. Das & J.R. Sharma, KD1065 (**holotypus**, BSD).*

Pileus 30–60 mm diam., convex, planoconvex to applanate or slightly planoconcave at maturity; center sometimes depressed and often with an umbo; pileipellis pruinose to velutinous, dry, gray yellowish to dark gray yellowish brown, grayish to deep grayish brown; margin slightly decurved or plane, sometimes broadly wavy, often crenate in older specimens. **Lamellae** adnate or / to subdecurrent, distant (*ca* 4 per cm), edge smooth, concolorous, pale ochre to orange yellow, unchanging when bruised; lamellulae of different sizes. **Stipe** 25–58 x 7–12 mm, central or slightly eccentric, cylindric or slightly tapering downwards, sometimes slightly grooved longitudinally, glabrous, dry, concolorous, often yellowish white at the base. **Context** stuffed to hollow, yellowish white. **Latex** rather sparse, white, unchanging. **Odor** mild. **Spore print** pale yellow ochre to orange yellow.

Basidiospores 7.5–10.0 (10.3) x (7.0) 7.3–9.3 µm, globose to broadly ellipsoid [Q = 1.03–1.15 (1.2), av. 1.06–1.09]; ornamentation amyloid, up to 2.7 µm high, composed of ridges arranged in somewhat parallel and / or in a spiral pattern, rarely branched, never reticulate; often shorter ridges occupying the gap between the main ridges; plage amyloid. **Basidia** 44–60 x 9–12.5 µm, subclavate to clavate, 4-spored; sterigma up to 7.5 µm long. **Pleuromacrocystidia** absent. **Pleuropseudocystidia** up to 8.5 µm broad, abundant, mostly cylindrical with rounded apex. Lamellae edge sterile. **Cheilomacrocystidia** absent. **Paracystidia** 45–56 x 8–11 µm, subclavate to clavate, sometimes narrower towards apex. **Hymenophoral trama** mixed, composed of hyphae, sphaerocytes and laticifers. **Pileipellis** a trichopalisade, up to 110 µm thick; terminal elements of suprapellis, cylindrical to ventricose, long, slender, 18–65 x 4–9 µm, thin walled, with brown intracellular pigmentation; subpellis composed of mostly globose cells, up to 27 µm diam. **Stipitipellis** a trichopalisade; terminal elements of suprapellis, cylindrical, slender, 15–50 x 3–7 µm, thin walled, with brown intracellular pigmentation; subpellis of globose cells. **Stipe trama**

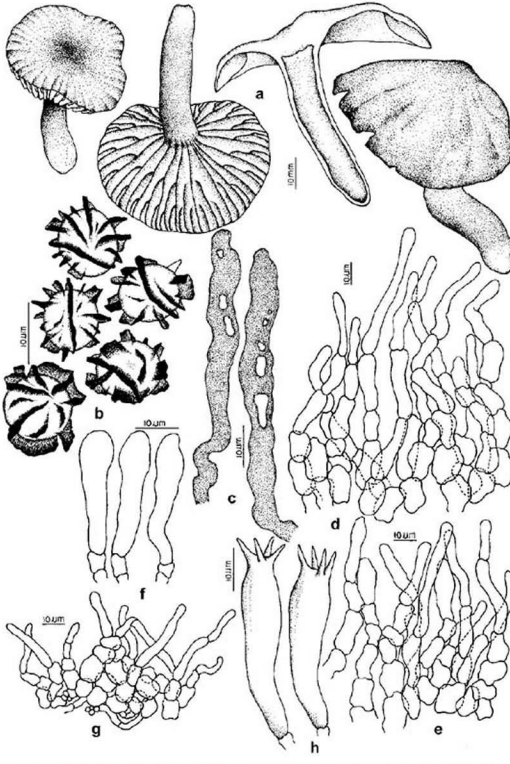

Fig. 1. *Lactarius montoyae* (from holotype). a. Basidiomes. b. Basidiospores. c. Pleuropseudocystidia. d, e. Cross-section of pileipellis. f. Paracystidia. g. Cross-section of stipitipellis. h. Basidia.

with numerous sphaerocytes.

Ecology — *Lactarius montoyae* grows preferably among mosses, forming ectomycorrhizal association with species of *Quercus* and *Rhododendron* in moist deciduous to mixed temperate (2500–3000 m) forests.

Notes — *Lactarius montoyae* is a rare species and was repeatedly collected only from the forests surrounding Dhakuri. *Lactarius romagnesii* Bon. with which the present taxon closely resembles regarding the basidiome morphology and high spore ornamentation, differs in having globose to broadly ellipsoid spores with the ornamentations forming a reticulate pattern (Heilmann-Clausen *et al.*, 1998) and shorter (10–35 x 4–9 μm) terminal elements of suprapellis. Microscopically, *L. montoyae* is also close to *L. pterosporus* Romagn. However, the pileus of *L. pterosporus* is strongly wrinkled in the center at maturity, "olivaceous buff to dark grayish buff or clay-buff" coloured (Heilmann-Clausen et al., 1998), lamellae are crowded and latex is white which turns pale grayish pink on drying on the flesh, not turning colour when isolated from flesh.

MATERIAL EXAMINED — INDIA. Uttaranchal: Bageswar, Dhakuri, September 29, 1999, leg. K. Das & J.R. Sharma, KD1065 (**holotype**, BSD); *ibid.*, September 16, 2003, leg. K. Das, KD7003 (BSD); *ibid.*, Dhakuri top, September 25, 2003, leg. K. Das & J.R. Sharma, KD7080, KD7081 (BSD).

Lactarius lignyotus Fr. var. ***canadensis*** A.H. Sm. & Hesler, Brittonia 14: 398 (1962). **Figure 2, a–i**

Pileus 40–75 mm diam., convex, becoming slightly depressed in the center with a papilla; pileipellis wrinkled, somewhat felty, dark grayish brown to brownish black; margin decurved, sulcate to irregularly loabed at maturity. **Lamellae** subdistant to distant (*ca* 4–5 per cm), sometimes intervenose, cream to pale ochre, pinkish when injured; lamellulae of different sizes; edge brownish black. **Stipe** 34–50 x 7–9 mm, dry felty, cylindric mostly concolorous or slightly paler towards base. **Context** hollow, yellowish white, slowly pinkish at the base. **Latex** white, unchanging. **Odor** indistinct. **Taste** mild. **Spore print** pale ochre or buff.

Basidiospores 8.5–11 x 8.2–9.7 μm, globose to subglobose (Q = 1.00–1.14, av. 1.01–1.04); ornamentation amyloid, up to 2 μm high, composed mostly of irregular ridges with spiny extremes, forming an incomplete reticulation, plage amyloid. **Basidia** 35–52 x 11–14 μm, subclavate, 4-spored; sterigma up to 7 μm long. **Pleuromacrocystidia** 55–64 x 9–11 μm, rare, subfusoid to ventricose. **Pleuropseudocystidia** up to 7 μm broad, cylindrical. Lamellae edge sterile. **Cheilomacrocystidia** 18–54 x 4–11 μm, abundant, clavate,

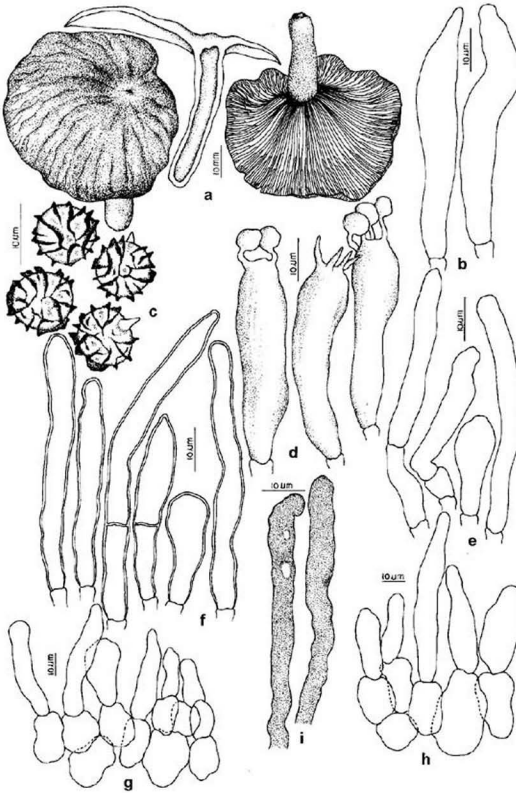

Fig. 2. *Lactarius lignyotus* var. *canadensis* (from KD7006). a. Basidiomes. b. Pleuromacrocystidia. c. Basidiospores. d. Basidia. e. Cheilomacrocystidia. f. Caulocystidia. g, h. Cross-section of pileipellis. i. Pleuropseudocystidia.

ventricose or cylindric, with brown intracellular pigmentation. **Pileipellis** a hymeniderm, composed of short chains of vesiculose cells; terminal cells 19–42 x 9–18 μm, cylindrical to clavate, thin walled, with brown intracellular pigmentation. **Stipitipellis** with numerous cystidia. **Caulocystidia** 16–90 x 6–13.4 μm, cylindrical to clavate with brown intracellular pigmentation; wall up to 1 μm thick. **Stipe trama** with abundant sphaerocytes.

Ecology — *Lactarius lignyotus* var. *canadensis* grows preferably among mosses and ectomycorrhizally with species of *Quercus* and *Rhododendron* in moist deciduous subtropical to temperate (2000–2800 m) Himalayan forests.

Notes — *Lactarius lignyotus* var. *canadensis* is also not common in Kumaon Himalaya and is readily recognized by its papillate pileus, radially wrinkled pileipellis, distinctly margined lamellae and mild taste. Besides, the habitat (among the mosses), pinkish stipe base after cutting, presence of pleurocystidia and wider caulocystidia make the present taxon distinct among the other taxa in this subgenus. It resembles *L. gerardii* Peck var. *subrubescens* (Smith & Hesler) Hesler & Smith closely. But the latter has only 1–2 rows of lamellulae, up to 0.6 μm thick basidiospore ornamentations and no pleuromacrocystidia.

MATERIAL EXAMINED — INDIA. Uttaranchal: Bageswar, Dhakuri, September 16, 2003, leg. K. Das, KD7006 (BSD); *ibid.*, Champawat, Mayawati, October 6, 2002, leg. K. Das & J.R. Sharma, KD5000 (BSD).

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Two new Hyphomycetes from rainforests of México, and *Briansuttonia*, a new genus to accommodate *Corynespora alternarioides*

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Abstract—*Corynespora aquatica* anam. sp. nov. on decaying leaves of an unidentified plant submerged in a stream, and *Solicorynespora sylvatica* anam. sp. nov. on leaf litter, both found in tropical rainforests, are described and illustrated. *Solicorynespora mulanjeensis* comb. nov. is proposed, and a key to *Solicorynespora* species is provided. *Briansuttonia* gen. nov. is established to accommodate *Corynespora alternarioides*.

Key words—submerged leaves, systematics, tropical fungi.

Introduction

Over 40 hyphomycetes were collected during two mycological surveys of conidial fungi from tropical plant material in several undisturbed rainforests of Los Tuxtlas (Veracruz) and Chajul (Chiapas), México. Among the collections were two conspicuous fungi clearly related to the genera *Corynespora* Güssow (Siboe & Kirk, 1999) and *Solicorynespora* R.F. Castañeda & W.B. Kendr. (1990). They appear to be new to Science.

The genus *Corynespora* is characterized by monotretic, terminal conidiogenous cells which undergo percurrent enteroblastic proliferation. Conidiogenous loci are apical, slightly depressed, somewhat melanized around the pore to produce solitary or catenate, multi-distoseptate, obclavate, cylindrical, obovate, oval to irregular, smooth-walled or verruculose, pale brown to pale olivaceous brown conidia, usually with a slightly melanized basal scar.

Corynespora cassiicola (Berk. & M.A. Curtis) C.T. Wei 1950, the type species, shows great variability in conidial morphology and can be found on a wide range on substrata as a plant pathogen or as a saprobe on decaying plant material. The illustrations provided by Wei (1950) of *C. cassiicola* from several collections on different substrata can be compared with many other species described in *Corynespora* such as: *C. heterospora* J.M. Yen 1980, *C. eranthemi* J.M. Yen & Lim 1980, *C. hemigraphidis* J.M. Yen & Lim 1980 and *C. ruelliae* J.M. Yen & Lim 1980, *C. longispora* Sarboy & Saikia 1980, *C. ligustri* Y.L. Guo 1984, *C. merremiae* Y.L. Guo 1984, *C. millettiae* Y.L. Guo 1984 and *C. viticis* Y.L. Guo 1984. All of them show a conidial morphology somewhat similar or strongly related to *C. cassiicola* as was suggested by Morgan-Jones (1988). However, the latter author did not synonymize them, and molecular studies may be needed to clarify their affinities and differences. Castañeda Ruíz and Kendrick (1990) introduced the genus *Solicorynespora* for species with tretic and percurrently proliferating conidiogenous cells analogous to *Corynespora* species, but with euseptate conidia. Similar criteria were adopted for the genera *Ellisembia* Subram. 1992 and *Sporidesmiella* P.M. Kirk 1982 to segregate species with distoseptate conidia and a conidial ontogeny similar to *Sporidesmium* Link 1809 (Ellis 1971, 1976), but with enteroblastic percurrent proliferation marked with annellations during conidiogenesis.

Taxonomy

Corynespora aquatica R.F. Castañeda, Heredia et Arias, anam. sp. nov.

FIGURES 1-2

Ad fungos conidiales, hyphomycetes pertinens. ***Coloniae*** *in substrato naturali effusae pilosae, amphigenae, brunneae; mycelium plerumque in substrato immersum, ex hyphis septatis, ramosis, laevibus, brunneis usque pallide-brunneis, 1.5–2.5 µm diam., compositum.* ***Conidiophora*** *conspicua, mononemata, erecta, recta, raro flexuosa, cylindrica, levier clavata ad apicem, 1- ad 4-septata, laevia, brunnea, 49–68 x 3–4 µm, ex cellula basali radiatim lobata orientes.* ***Cellulae conidiogenae*** *uniloculosae, tretieae, terminales, brunneae, 7–12 x 3–3.5 µm, plerumque determinatae, interdum 1 proliferatione enteroblastica percurrenti praeditae.* ***Loci conidiogeni*** *pori apicales.* ***Secedentia conidiorum*** *schizolytica.* ***Conidia*** *obclavata usque ad* cylindrica, solitaria, *acrogena, (1- ad) 2- (ad 3-) distoseptata, 54–46 x 3.0–4.5 µm, pallide brunnea, laevia, sicca. Teleomorphosis ignota.*

Etymology: Latin, *aquaticus* refers to its growing in water.

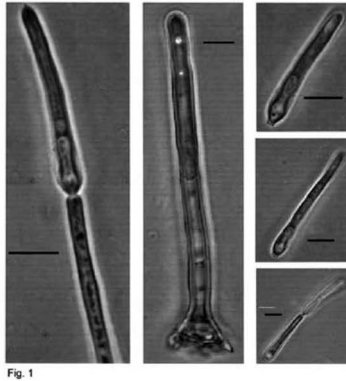

Fig. 1

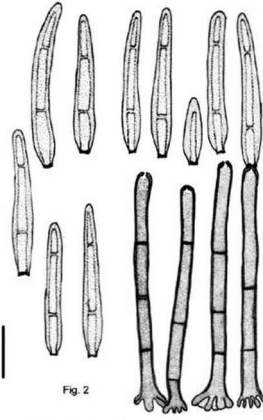

Fig. 2

Figs. 1-2. *Corynespora aquatica*. Conidiophores, conidiogenous cells and conidia. **Fig. 1**: Micrographs. **Fig. 2**: Drawings. Scale is indicated by bar (10 μm).

Conidial fungi, hyphomycetes. **Colonies** on the natural substrate effuse, hairy, amphigenous, brown; mycelium mostly immersed composed of septate, branched, smooth-walled, 1.5–2.5 diam., pale brown to brown hyphae. **Conidiophores** differentiated, mononematous, erect, straight, rarely flexuous, slightly clavate towards the apex, 1- to 4-septate, smooth-walled, brown, 49–68 × 3–4 μm; arising from a radially lobed basal cell. **Conidiogenous cells** unilocal, tretic, terminal, brown, 7–12 × 3–3.5 μm, mostly determinate, but sometimes with an enteroblastic, percurrent proliferation. **Conidiogenous loci** an apical pore. **Conidial secession** schizolytic. **Conidia** obclavate to cylindrical, solitary, acrogenous, (1- to) 2- (to 3-) distoseptate, 34–46 × 3.0–4.5 μm, pale brown, smooth-walled, dry. **Teleomorph**: unknown. **Matrix:** on decaying leaves submerged in a stream, in the rainforest 'Los Tuxtlas', Veracruz, México, leg. R.M. Arias, J. Yadeneiro de la Cruz Elizondo & R.F. Castañeda, 19. V. 2002 (**Holotype**- XAL CB742, Instituto de Ecología, Xalapa, Veracruz, México).

COMMENTS: Among described *Corynespora* spp. listed by Siboe & Kirk (1999) the new species resembles only *C. matuszakii* Morgan-Jones 1988, which has 2- to 10-distoseptate conidia, 56–260 × 10.0–12.5 μm in size, formed solitary or in a chain, whereas those of *C. aquatica* are always solitary, mostly 2-distoseptate and smaller.

Solicorynespora sylvatica R. F. Castañeda, Heredia, Arias et Guarro, anam. sp. nov. FIGURES 3-4

*Ad fungos conidiales, hyphomycetes pertinens. **Coloniae** in substrato naturali effusae pilosae, hypophyllae, brunneae; mycelium plerumque in substrato immersum, ex hyphis septatis, ramosis, laevibus, brunneis usque pallide-brunneis, 1–2 μm diam., compositum. **Conidiophora** conspicua, mononemata, erecta, recta vel leviter flexuosa, cylindrica, leviter clavata ad apicem, 2- ad 7-septata, laevia, brunnea, 30–120 x 5–7 μm. **Cellulae conidiogenae** unilocolosae, treticae, terminales, brunneae, 8–15 x 3–5 μm, determinatae vel indeterminatae, interdum 1–2 proliferationibus enteroblasticis percurrentibus proeditae. **Loci conidiogeni** pori apicales. **Secedentia conidiorum** schizolytica. **Conidia** obpyriformia usque ad obclavata, longissima rostrata, solitaria, acrogena, 3- ad 6-euseptata, 35–67 x 5–7 μm (rostro incluso), brunnea, pallide-brunnea vel ad apicem subhyalina; laevia, sed quaeque in cellula basali verruculosa; rostro 30–50 x 1.5–2.0 μm induta. Teleomorphosis ignota.*

Etymology: Latin, *sylvatica*, refers to a forest inhabitant growing wild.

Conidial fungi, hyphomycetes. **Colonies** on the natural substrate, effuse, hairy, hypophyllous, brown; mycelium mostly immersed, composed of septate, branched, smooth-walled, brown to pale brown hyphae, 1–2 μm diam. **Conidiophores** differentiated, mononematous, erect, straight, or slightly flexuous, cylindrical, slightly clavate towards the apex, 2- to 7-septate, smooth-walled, brown, 30–120 × 5–7 μm. **Conidiogenous cells** unilocal, tretic, terminal, brown, 8–15 × 3–5 μm, determinate or indeterminate, sometimes with 1–2 enteroblastic percurrent proliferations. **Conidiogenous loci** an apical pore. **Conidial secession** schizolytic. **Conidia** obpyriform to obclavate, very long rostrate, solitary, acrogenous, 3- to 6-euseptate, 35–67 × 5–7 μm (rostrum included), brown and pale brown to subhyaline towards the apex, smooth-walled, with verruculose basal cell; rostrum 30–50 × 1.5–2.0 μm. Teleomorph unknown. **Matrix**: on decaying leaves of unidentified plant, in a rainforest Chajul, Chiapas, Mexico, leg. R. Guevara, 10. VI. 1999 (**Holotype**- XAL 803-3, Instituto de Ecología, Xalapa, Veracruz, México).

COMMENTS: The genus *Solicorynespora* (Figs 5–6) comprises seven accepted species: *S. aterrima* (Berk. & M.A. Curtis) R.F. Castañeda & W.B. Kendr. 1990, *S. calophylli* (Hol.-Jech. & R.F. Castañeda) R.F. Castañeda & W.B. Kendr.1990, *S. kendrickii* R.F. Castañeda 1996, *S. litchii* (Matsush.) R.F. Castañeda & W.B. Kendr. 1990, *S. obclavata* (Dyko & B. Sutton) R.F. Castañeda & W.B. Kendr. 1990, *S. pseudolmediae* (R.F. Castañeda) R.F. Castañeda & W.B. Kendr.1990, and the type species, *S. zapatensis* R.F. Castañeda & W.B. Kendr. 1990, but only *S. aterrima*, *S. calophylli*, *S. litchii* and *S. obclavata* superficially resemble *S. sylvatica* in their rostrate conidial morphology. Of them, the first has obclavate, conico-truncate, rostrate, 3- to 5-septate conidia, 33–74 × 8–10 μm, with verrucose, dark brown lower cells and pale brown, smooth upper cells; *S. calophylli* has 2-septate conidia, each with a stout, shortly rostrate, 3–6.5 × 2–3 μm, hyaline to subhyaline apical cell; *S. litchii* has conidia 3- (to 4-) septate, each with a subulate-rostrate, 7–17 × 1.5–2.0 μm, hyaline to subhyaline apical cell and another rostrate hyaline to subhyaline, 9–15 × 1.5 μm, lateral, subapical cell (appendix) directed downwards. *S. obclavata* has 4- to 6-euseptate, smooth conidia, 32.0–62.5 × 9.5–11.0 μm, with the upper two cells subhyaline to pale brown and the lower 3–4 cells brown. *Solicorynespora sylvatica* is therefore clearly distinct from these species.

Corynespora mulanjeensis B. Sutton 1993 has solitary, euseptate conidia which arise from an apical pore, making it more closely related to the genus *Solicorynespora*. The following new combination is therefore proposed:

Solicorynespora mulanjeensis (B. Sutton) R.F. Castañeda, M. Stadler et Guarro, comb. nov. FIGURE 6d

BASIONYM: *Corynespora mulanjeensis* B. Sutton, Mycological Papers **167**: 23 (1993).

Key to *Solicorynespora* species

1 *Conidia neither rostrate nor strongly tapered towards the apex* *2*

Conidia rostrate or strongly tapered towards the apex *3*

2 (1) *Conidia cylindrical to ellipsoid, 2- to 4-euseptate, 15–26 × 6–7 µm, smooth-walled, sometimes slightly verruculose, with brown central cells and subhyaline ends* ..... **kendrickii**

Conidia obclavate to oval (2- to) 4- (to 5-) euseptate, 16–29 × 8.5–12.0 µm, smooth-walled, brown, each with a subhyaline to pale brown apical cell ..... **pseudolmediae**

3 (1) *Conidia smooth-walled* *4*

Conidia verruculose or verrucose *5*

4 (3) *Conidia obclavate, rostrate towards the apex, 4- to 6-euseptate, 32.0–62.5 × 9.5–11.0 µm (rostrum included), smooth-walled, with lower cells medium brown and two upper cells subhyaline to pale* ..... **obclavata**

Conidia obclavate to fusiform, gradually tapered to an obtuse apex, 5- to 8-euseptate, 56–71 × 10.0–12.5 µm, medium brown with the lower 4–5 cells slightly darker and dark brown at the base ..... **mulanjeensis**

5 (3) *Conidia with no more than 4 septa* *6*

Conidia sometimes with more than 4 septa *7*

6 (5) *Conidia obpyriform to obclavate, 2-euseptate, 11–16 × 5.0–7.0 µm, brown; with a subhyaline to subhyaline, 3.0–6.5 × 2–3 µm, stoutly and shortly rostrate at apical cell* ..... **calophylli**

Conidia obclavate, 3- (to 4-) euseptate, 21–32 × 6.5–8.5 µm, brown with a rostrate-subulate, apically elongated, 7–17 × 1.5–2.0 µm, hyaline or subhyaline apical cell and very often with a lateral, subulate, 9–15 × 1.5 µm, hyaline to subhyaline appendage ..... **litchii**

7 (5) *Conidia 3- to 5-euseptate, obclavate, rostrate towards the apex, 33–74 ×8–10 µm, with dark brown lower cells and pale brown apical and subapical cells* ..... **aterrima**

Conidia 3- to 6-euseptate, obpyriform to obclavate, very longrostrate, 35–67 ×5–7 µm (rostrum ncluded), brown and pale brown to subhyaline towards the apex. ..... **sylvatica**

Figs 3-4. *Solicorynespora sylvatica*. Conidiophores, conidiogenous cells and conidia. **Fig. 3**. Drawings. **Fig. 4**: Micrographs. Bar = 10 µm.

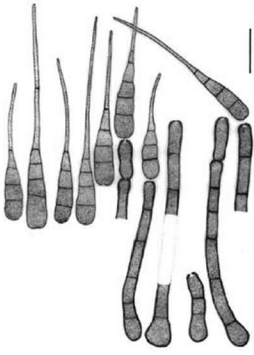

Fig. 3

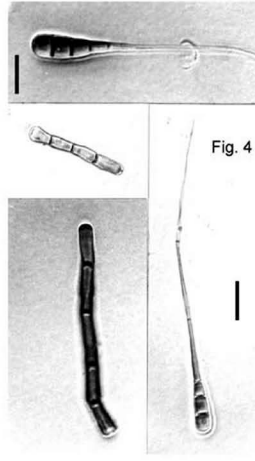

Fig. 4

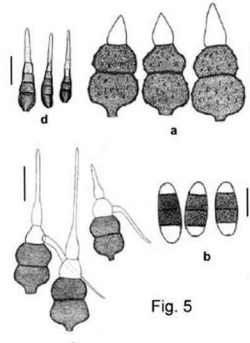

Fig. 5

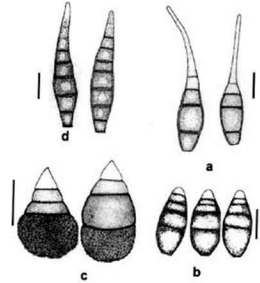

Fig. 6

Figs 5-6. Representative conidia of *Solicorynespora* spp. **Fig. 5a.**: *S. calophylli*. **Fig. 5b**: *S. kendrickii*. **Fig. 5c**: *S. litchii*. **Fig. 5d.** S. aterrima. - **Fig. 6a**:. *S. obclavata*. **Fig. 6b**: *S. pseudolmediae*. **Fig. 6c**: *S. zapatensis*. **Fig. 6d**: *S. mulanjeensis*. Bars = 10 μm.

Briansuttonia R.F. Castañeda, Minter et Saikawa, anam. gen. nov.

Ad fungos conidiales, hyphomycetes pertinens. ***Coloniae*** *effusae, pilosae usque funiculosae, brunneae vel atrobrunnea. Mycelium superficiale et aliquot in substrato immersum, ex hyphis septatis, anastomosantibus, brunneis vel atrobrunneis, laevibus vel verrucosis compositum.* ***Conidiophora*** *conspicua, mononemata, septata, erecta, brunnea vel atrobrunnea, laevia vel verrucosa.* ***Cellulae conidiogenae*** *uniloculosae, tretícae, terminales, determinatae vel indeterminatae, proliferationibus percurrentibus enteroblasticis praeditae.* ***Loci conidiogeni*** *pori apicales.* ***Secedentia conidiorum*** *schyzolytica.* ***Conidia*** *solitaria, brunnea vel atrobrunnea, acrogena, sicca, obclavata, obpyriformia, ellipsoidea vel irregularia, truncata ad basin, laevia vel verrucosa, ex 2- ad 15-distoseptis transversis et pluribus distoseptis longitudinalibus et obliquis praedita.* ***Teleomorphosis*** *ignota.* ***Species typica***: ***Briansuttonia alternarioides*** (B. Sutton & Pascoe), comb. nov.

Etymology: In honour of Dr Brian C. Sutton (United Kingdom) in recognition of his contributions to the study of anamorphic fungi.

Conidial fungi, Hyphomycetes. **Colonies** effuse, hairy to funiculose, brown to dark brown. Mycelium superficial and rather immersed, composed of septate, anastomosing, brown or dark brown, smooth-walled or verrucose hyphae. **Conidiophores** differentiated, mononematous, septate, erect, brown to dark brown, smooth-walled or verrucose. **Conidiogenous cells** unilocal, tretic, terminal, determinate or indeterminate with enteroblastic percurrent proliferations. **Conidiogenous loci** an apical pore. **Conidial secession** schyzolytic. **Conidia** solitary, brown to dark brown, acrogenous, dry, obclavate, obpyriform, ellipsoid or irregular, truncated at the base, smooth-walled or verrucose, with 2- to 15-transverse distosepta and several longitudinal and oblique distosepta. Teleomorph: unknown. **Type Species**: *Briansuttonia alternarioides* comb. nov.

COMMENTS: When Sutton & Pascoe (1988) described *Corynespora alternarioides*, they remarked that, although the conidiophores and tretic conidium ontogeny resemble those of other *Corynespora* species, the resulting conidia are quite different: "no species hitherto described in *Corynespora* has conidia with transverse, longitudinal and oblique distosepta". If species with distoseptate, muriform conidia were to be included in this genus the generic concept would have to be expanded drastically. The differences noted by Sutton & Pascoe (1988) therefore provide good reasons to regard this taxon as different at generic level.

The genera *Alternaria* Nees (Ellis 1971, 1976) and *Stemphylium* Wallr. (Ellis 1971, 1976) resemble the new genus in having unilocal, apical conidiogenous loci and muriform conidia, but the conidiogenous loci in *Alternaria* are flat and melanized, and the conidia are euseptate. In *Stemphylium* the conidial ontogeny is holoblastic and that genus is also clearly distinguished by its euseptate muriform conidia.

Briansuttonia alternarioides (B. Sutton & Pascoe) R.F. Castañeda, Minter & Saikawa, comb. nov.

Basionym: *Corynespora alternarioides* B. Sutton & Pascoe, *Australian Systematic Botany* **1:** 127 (1988).

Acknowledgments

We are deeply indebted to Profs. Lori M. Carris (Washington State University) and Roger Goos (University of Rhode Island) for kindly reviewing the manuscript and for many suggestions that greatly improved it. We thank the Cuban Ministry of Agriculture and CITMA for facilities through projects 2053, 2053 and 2054, and the UK Darwin Initiative for financial support through the project "*Biodiversity Conservation in Cuba*". We also acknowledge the technical assistance of M. Caraballo.

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MYCOTAXON

Volume 89(2), pp. 307-310 *April-June 2004*

Two new species and a new record of *Anthracoidea (Ustilaginales)* from China

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Abstract—Two new species, *Anthracoidea kanasensis* on *Carex ovalis* and *A. striata* on *C. pediformis* are described. A new species for China, *A. rupestris* on *C. pediformis* is reported.

Keywords—Ustilaginomycetes, taxonomy, smut fungi

A smut fungus on *Carex ovalis*, a new record of plant for China, in the section *Planatae* of the subgenus *Vignea*, was collected from the Kanas Lake of Xinjiang Uygur Autonomous Region, in the Northwestern part of China. According to Prof. Liang Songjun's identification, the host plant is very closely related to *Carex maackii* Maxim. Until now no *Anthracoidea* has been reported from the section *Planatae*. Kükenthal (1909) put *Carex ovalis* in the section *Ovales* of the subgenus *Vignea*. On species of *Carex* in the subgenus *Vignea*, section *Ovales* two species of *Anthracoidea* have been recognized: 1) *Anthracoidea uleiana* (Syd. & P. Syd) Vánky (1997) [syn. *A. pannucea* (Liro) Vánky (1985)], with ustilospores measuring 14-17 µm in diameter, and "dark-brown spores with 3-5 light, thin-walled, rounded areas, which when the spores are dried collapse and form mamillate depressions, giving a peculiar aspect to the spores" (Vánky 1987); type on *Carex bonplandii* Kunth, Brazil, and 2) *Anthracoidea verrucosa* (Savile) Nannf., with ustilospores measuring 14-23 x 13-17 µm, wall 1 µm, evenly thickened; type on *Carex ebenea* Rydb., USA.

The fungus on *Carex ovalis* in China is similar to *Anthracoidea verrucosa* from which it differs mainly in having unevenly thickened ustilospore walls. It is described as:

Anthracoidea kanasensis H. C. Zhang & L. Guo, sp. nov. Figs. 1-2

Sori in ovariis, subglobosi, 1.5-2 mm longi, 1-1.5 mm lati, primo membrane cinerascenti, fungali cooperti, deinde exposite. Massa sporarum nigra, semiagglutinata. Ustilosporae a fronte subglobosae, ellipsoideae, ovoideae vel irregulares, (10.5-)15-22.5(-25) x (10.5-)11-20 µm, ab acie 9-12.5 µm latae, rubrobrunneae; pariete inaequaliter incrassato, 1-3 µm crasso, in angulis

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crassissimo, nonnunquam protuberantiis 1-4, gibberis internis 1-4 et areis refractivis 1-2, superficie dense verruculoso.

Sori in ovaries, subglobose, 1.5-2 mm long, 1-1.5 mm wide, at first covered by a grayish, fungal membrane, later becoming exposed. Spore mass black, semi-agglutinated. Ustilospores in plan view subglobose, ellipsoidal, ovoid or irregular, (10.5-)15-22.5(-25) x (10.5-)11-20 µm, in side view 9-12.5 µm wide, reddish-brown; wall unevenly thickened, 1-3 µm, thickest at the angles, sometimes protuberances 1-4, internal swellings 1-4, light reflective areas 1-2, surface densely verruculose, the warts often confluent.

On *Carex ovalis* Good. (Cyperaceae), Xinjiang: Burqin, Kanas Lake, alt. 1400 m, 8 VIII 2003, L. Guo & H. C. Zhang 2182, HMAS 86708 (holotypus); Burqin, Kanas Lake, alt. 1700 m, 8 VIII 2003, L. Guo & H. C. Zhang 2180, HMAS 86709 (paratypus).

There is a species of *Anthracoidea* on *Carex pediformis* in the subgenus *Carex*, section *Digitatae*, which is closely related to *Anthracoidea irregularis* (Liro) Boidol & Poelt. However it differs from *A. irregularis* in having larger ustilospores and striate walls. It is described as:

Anthracoidea striata H. C. Zhang & L. Guo, sp. nov. Figs. 3-4

Sori in ovariis, subglobosi, 1.5-2.5 mm longi, 1.5-2.2 mm lati, primo membrane cinerascenti, fungali cooperti, deinde expositi. Massa sporarum nigra, semiagglutinata. Ustilosporae a fronte subglobosae, ellipsoideae, ovoideae vel irregulares, 15-30(-37.5) x 14-20 µm, ab acie 10-13 µm latae, rubrobrunneae vel flavidobrunneae; pariete inaequaliter incrassato, 1-3.5 µm crasso, in angulis crassissimo, protuberantiis 1-7, gibberis internis 1-5 et areis refractivis 1-3, superficie dense verruculoso, sub SEM verrucoso et striato.

Sori in ovaries, subglobose, 1.5-2.5 mm long, 1.5-2.2 mm wide, at first covered by a grayish, fungal membrane, later becoming exposed. Spore mass black, semi-agglutinated. Ustilospores in plan view subglobose, ellipsoidal, ovoid or irregular, 15-30(-37.5) x 14-20 µm, in side view 10-13 µm wide, reddish-brown or yellowish-brown; wall unevenly thickened, 1-3.5 µm, thickest at the angles, protuberances 1-7, internal swellings 1-5, light reflective areas 1-3, surface densely verruculose, the warts often confluent, in SEM prominently verrucose and partly striate.

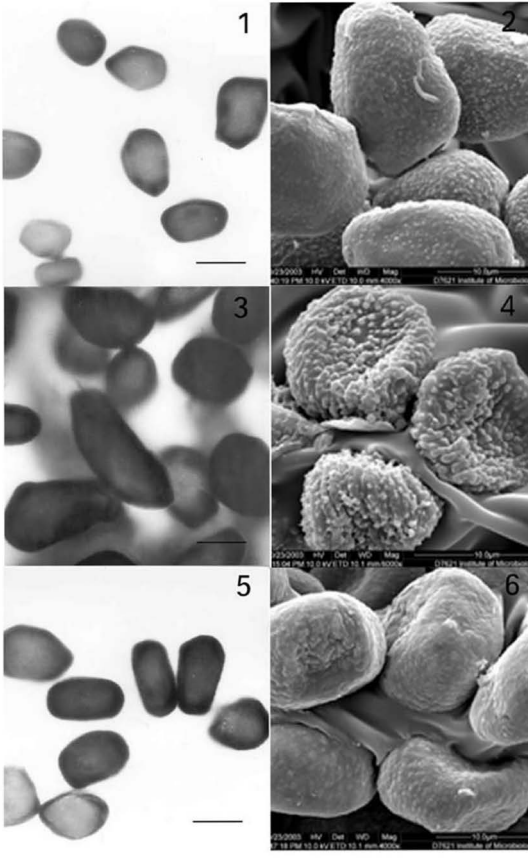
On *Carex pediformis* C.A. Mey (Cyperaceae), Xinjiang: Burqin, Hemuxiang, alt. 1100 m, 6 VIII 2003, L. Guo & H. C. Zhang 2153, HMAS 86710 (holotypus).

On *Carex pediformis* in the subgenus *Carex*, section *Digitatae*, a further species of *Anthracoidea* was collected in the Xinjiang Region. It is identified as *A. rupestris*, which is new to China (Guo 2000, 2002, 2004; Wang & Piepenbring 2002):

Figs. 1-2. Ustilospores of *Anthracoidea kanasensis* on *Carex ovalis* in LM and SEM (HMAS 86708, holotypus).

Figs. 3-4. Ustilospores of *Anthracoidea striata* on *Carex pediformis* in LM and SEM (HMAS 86710 holotypus).

Figs. 5-6. Ustilospores of *Anthracoidea rupestris* on *Carex pediformis* in LM and SEM (HMAS 86711). Bars = 11 µm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Anthracoidea rupestris Kukkonen

Ann. Bot. Soc. Zool.-Bot. Fenn. „Vanamo" 34(3):47, 1963. Figs. 5-6

Sori in ovaries, subglobose, 2.2-3.5 mm long, 2-3 mm wide, at first covered by a grayish membrane. Spore mass semi-agglutinated, black. Ustilospores in plan view subglobose, ellipsoidal, ovoid or irregular, 15-25.5(-30.5) x 14.5-22.5 µm, in side view 8-14 µm wide, reddish-brown or yellowish-brown; wall unevenly thickened, 1-3(-4) µm, thickest at the angles, protuberances 1-4, with light reflective areas, internal swellings commen, densely verruculose, the warts low, often confluent.

On *Carex pediformis* C. A. Mey, Xinjiang: Burqin, Kanas Lake, alt. 1500 m, 8 VIII 2003, L. Guo & H. C. Zhang 2181, HMAS 86711; Burqin, Kanas Lake, alt. 1800 m, 8 VIII 2003, L. Guo & H. C. Zhang 2175, HMAS 86712.

So far 22 species of *Anthracoidea* have been recorded for China, including *A. kanasensis*, *A. striata*, and *A. rupestris*.

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MYCOTAXON

Volume 89(2), pp. 311-320 *April-June 2004*

A revision of the types of *Diderma subcaeruleum* and *D. globosum* var. *europaeum*

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Abstract—The types of *Diderma alpinum*, *D. globosum* var. *europaeum*, *D. niveum* and *D. subcaeruleum* were compared by applying electron microscopy techniques. *Diderma subcaeruleum* was synonymized with *D. niveum* and *D. globosum* var. *europaeum* was synonymized with *D. alpinum*.

Key words—Myxomycetes, *Physarales*, SEM, taxonomy

Introduction

Meylan was the first author who studied exhaustively the nivicolous myxomycetes and who proposed a large number of new taxa for science. Altogether he proposed 14 species, 18 varieties and 13 forms. Unfortunately, he did not cite type material, as this was not mandatory at the time. It was Kowalski, who made an extensive revision of Meylan's material and lectotypified the abundant material stored in the herbarium LAU (Kowalski 1975).

In 2002 we began studying Meylan's type material, putting special emphasis on the genera *Lamproderma* and *Diderma*. Within the so-called *D. niveum* complex (Moreno et al. 2003c), consisting of nivicolous species of *Diderma* with sessile, white sporocarps and coriaceous peridia, the following type material of Meylan's species was studied: *Diderma alpinum*, *D. alpinum* var. *macrosporum*, *D. microcarpum* and *D. niveum* var. *ferrugineum*. Fortunately, we were also able to find and to study *D. niveum* proposed by Rostafinski. We have been able to synonymize *D. niveum* with *D. niveum* var. *ferrugineum* and we have proposed a new species, *D. meyerae*. However, we have not included the variety *D. globosum* var. *europaeum* proposed by Buyck (1988) in our study, that also can be included within the *D. niveum* complex, as the type was not available to us at that time. We have since been able to study the type material proposed by Buyck and to compare it with the *Diderma* already studied by us.

At the same time, we are studying the type material of the nivicolous species of myxomycetes proposed by Kowalski, who described 15 nivicolous

species as new between the years 1966 and 1975. Among these, there are some very common species, such as *Lamproderma maculatum* and *Lepidoderma aggregatum*, but also rare species that have not been collected since and are known only from his collections, such as *Diderma subcaeruleum*, *D. nigrum*, or from a single collection, *Diacheopsis spinosifila* and *Trichia synspora*. In our recent studies, we have been able to demonstrate that various species are synonyms of other, more common species. Thus, we have proposed the synonymy of *Trichia synspora* with *T. varia* (Singer et al. 2003), *Diderma nigrum* with *D. asteroides* (Moreno et al. 2003b) and *Diacheopsis spinosifila* with *Lepidoderma didermoides* (Moreno et al. 2003a).

This shows how important it is to revise the type material of the nivicolous myxomycetes, in order to specify accurately the macroscopic and microscopic characteristics of the species described from this particular habitat.

Materials and Methods

The material was studied with a binocular microscope and, after mounting in Hoyer's medium, with a Nikon (Optiphot) microscope. Scanning electron microscopy (SEM) micrographs were taken in the University of Alcalá de Henares, using a Zeiss DSM-950. SEM-preparation was made as indicated in Moreno et al. (2002).

The type material comes from the herbaria BPI and NY and was compared with the material indicated in Moreno et al. (2003c).

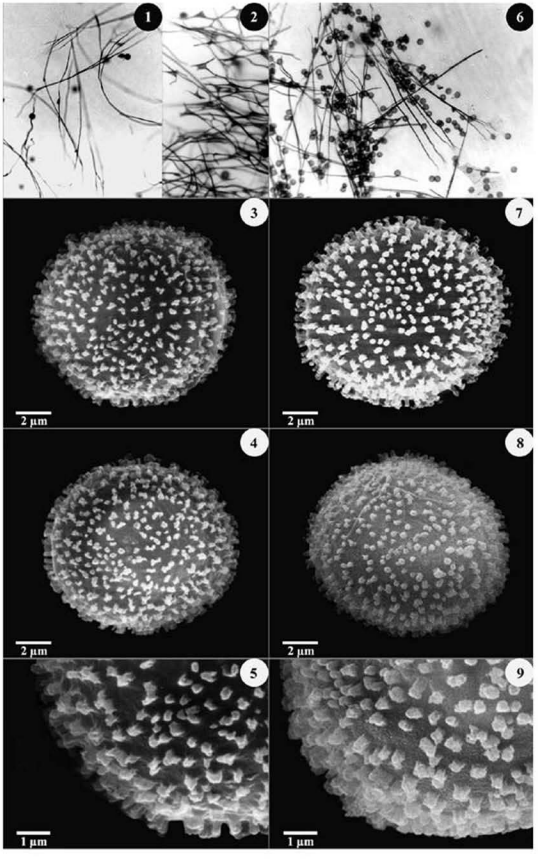
Diderma subcaeruleum Kowalski, *Mycologia* 60(3): 598. 1968

Figs 1-5

Original diagnosis: *Sporangiis gregariis, sessilibus, haemisphaericis vel paulo elongatis, chalybeis, 1-2 mm diam; peridio simplici, tenui, fragili, parce et innate calcareo; hypothallo inconspicuo; columella nulla vel, quandocumque presenti, exigua, alba; capillitio copioso, e filamentis gracilibus, rigidis, atropurpureo-brunneis ad apicem incoloratis, aliquantum parce ramosis anastomosantibusque composito; sporis globosis, atrobrunneis, verrucosis, 10-12 (-16) µm diam; plasmodio ignoto.*

Material studied: On decaying plant debris, near garbage dump, 6400 ft, 7.VIII.1967, Crater Lake National Park, Oregon, BPI 815218 (type) and BPI 815212 (isotype).

Figs 1-5. *Diderma subcaeruleum* (type). 1-2. Capillitium. 3-4. Spores. 5. Detail of spore ornamentation. **Figs 6-9.** *Diderma niveum* (type). 6. Capillitium. 7-8. Spores. 9. Detail of spore ornamentation.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The type material is kept in a box with Kowalski's personal herbarium number D.T.K. 6887 and the number BPI 815218. An isotype is kept in another box bearing the number BPI 815212. The type consists of four twigs with the bark still attached, covered by abundant well preserved sporocarps. The isotype presents one bark bearing twig with abundant fructifications arranged in three portions.

Sporophores sporocarpic to subplasmodiocarpic, 0.5-2.0 x 0.3-1 mm, aggregated, sessile, hemispheric to pulvinate, crushed laterally between them. Hypothallus translucent, generally inconspicuous. Stalk absent. Peridium double, persistent. Exoperidium calcareous, smooth to rugose, white to white with bluish discontinuous tones in the zones of sporocarps where this layer is badly formed, sometimes absent, in this case the endoperidium can be observed; irregular dehiscence. Endoperidium membranous, translucent, iridescent, closely united with the exoperidium. Columella sometimes absent, if present whitish to white with beige tones, very small to subglobose, up to 0.3 mm in diam. Capillitium (Figs 1-2) very dense, formed by straight, rigid filaments, 1-2 μm in diam., sometimes branched and anastomosed, with triangular to polygonal widened areas in the union of the threads, on the surface of the threads rarely appear globose nodules, filaments dark brown, paler towards the extremities with hyaline tips. Spores 10-12 μm in diam., spiny, violaceous brown in LM, dark brown in the mass. By SEM (Figs 3-5) the ornamentation is formed by baculae.

Diderma globosum* var. *europaeum Buyck, Bull. Jard. Bot. Belg. 58(1-2): 199. 1988

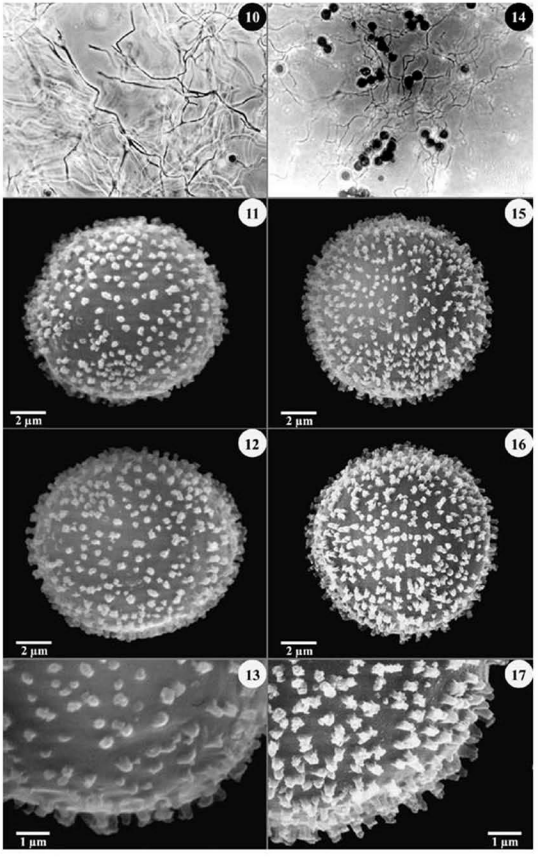
Figs 10-13

Original diagnosis: *A var. globoso recedens plasmodiocystis vel sporocystis irregularibus, columella depressa et fere convexa, capillitio firmo, sporis subglobosis, (10.07) 10.41-11.57 (12.30) µm diam (75 S, 3 C), lucem orientem versus visae egaliter coloratis.*

Material studied: Switzerland, on herbs and twigs, 2.V.1914, *leg.* Jaap, BPI 814106 (holotype) and NY 7830 (isotype).

The type is kept in a matchbox, in which two dry stems are stuck, where strongly aggregated fructifications can be observed, lacking almost completely peridium, capillitium and spores, but showing a columella taking up almost the whole base. In the sides of the sporocarps residues of the peridium remain.

Figs 10-13. *Diderma globosum* var. *europaeum* (type). 10. Capillitium. 11-12. Spores. 13. Detail of spore ornamentation. **Figs 14-17.** *Diderma alpinum* (type). 14. Capillitium. 15-16. Spores. 17. Detail of spore ornamentation.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The isotype is more abundant and consists of four pieces of dry stems stuck on the lid of a box, with abundant well preserved fructifications in three of them; one piece consists only of the bases of the sporotheca, as in the type material.

Sporophores sporocarpic, 1-1.5 mm in diam., to subplasmodiocarpic, 2-3 x 0.8-1.2 mm, some fructifications up to 6 mm long, aggregated, sessile, hemispheric to pulvinate. Hypothallus continuous, whitish, sometimes hardly conspicuous. Stalk absent. Peridium double, normally persistent. Exoperidium calcareous, smooth, white, with irregular dehiscence. Endoperidium membranous, translucent, iridescent, clearly separated from the exoperidium. Columella white, flat to broadly convex, covering almost the entire base of the sporotheca. Capillitium (Fig 10) formed by flexuouse filaments, about 1 μm in diam., rarely attaining 2 μm, branched and anastomosed, light violaceous brown, hyaline towards the extremities. Spores 11-12 μm in diam., spiny, violaceous brown in LM, dark brown in the mass. By SEM (Figs 11-13) the ornamentation is formed by baculae.

Discussion and Conclusions

According to Kowalski (1968) the outstanding characteristic of *Diderma subcaeruleum* is its blue-grey or steel-grey peridium, which is double, but due to the tight union of the thin, membranous, colourless inner layer with the outer layer, appears single. He was able to recognize this taxon in the field because of its colour, its crowded habit and its substrate, always being twigs.

He stresses the similarity with *Diderma niveum* because they both are alpine species having crowded sporocarps of the same diameter and identical spore size and ornamentation. As for differences between these two species he quotes the habitat, *D. niveum* preferring logs, *D. subcaeruleum* being found almost exclusively on decaying coniferous twigs, the colour of the sporocarps being usually white in *D. niveum* and greyish in *D. subcaeruleum*, the columella being large, globose or hemispherical, ochraceous to deep orange in *D. niveum* being a small, white or cream coloured mound at the base of the sporocarp, sometimes lacking in *D. subcaeruleum*, and finally the double peridium of *D. niveum* with its two layers distinctively separated.

Recently, we have made a study on several nivicolous taxa with white sporocarps (Moreno et al. 2003c) and synonymized *Diderma niveum* with *D. niveum* var. *ferrugineum*, proposing a hitherto undescribed species, *D. meyerae*, which was confused with the latter two taxa by early myxomycetologists. Thus, in order to understand Kowalski's observations, we have to know how he interpreted *D. niveum*, from which he separated *D. subcaeruleum* on the basis of the differences mentioned above. Kowalski (1975) distinguished the two species *D. niveum* and *D. alpinum*, considering

D. niveum var. *ferrugineum* a sporangiate form and thus a synonym of *D. alpinum*. He attributed a dark purple-brown capillitium to both *D. niveum* and *D. alpinum*, separating the latter species from *D. niveum* only by its usually plasmodiocarpous habit, or if sporocarpous, with sporocarps less than 1 mm in diam. Obviously, he misinterpreted *D. alpinum*, whose key character is its light-coloured, delicate capillitum, and not the growth habit that can be sporocarpous or plasmodiocarpous, regardless of its size. Thus, he did not know the *D. alpinum* of Meylan, which now can be studied in the new lectotype proposed by Moreno et al. (2003c). His concept of *D. niveum*, *D. niveum* var. *ferrugineum* and *D. alpinum* is, in our opinion, the same and corresponds to *D. niveum*.

We differ in our observations on *Diderma subcaeruleum*, a species that is known only from Kowalski's collections in California, in a few essential points from his original diagnosis and we cannot support his arguments that justify its separation from *D. niveum*. The exoperidium of *D. subcaeruleum* is white to white with bluish tones and is definitely lacking of a predominant greyish colouring, described by Kowalski as bluish grey, grey or steel-grey. Faint bluish tones can be observed in the areas of sporocarps where the exoperidium is badly developed and are due to inner layer showing through and producing this coloration. Furthermore, according to Kowalski, *D. subcaeruleum* prefers decaying coniferous twigs as its substrate, while *D. niveum* prefers logs. Certainly, it is true that some species of myxomycetes prefer a specific substrate, but as *D. subcaeruleum* was not collected again, we cannot be sure if this species was found exclusively on dead coniferous twigs or not. In the course of our studies we have also seen *D. niveum* on different types of substrates including decaying coniferous twigs. Also, we sometimes find a well developed columella in *D. subcaeruleum* which, according to Kowalski, is reduced to a mound at the base of the sporocarp: It should be kept in mind that the morphology of the columella in the nivicolous species of *Diderma* is highly variable. Finally, we consider the close union of the two peridial layers as not being a stable character, as this depends very much on environmental conditions. The spore diameters and spore ornamentation formed by baculae (Figs 3-5, 7-9) are the same in both species. Comparing the capillitium of the two types of *D. niveum* and *D. subcaeruleum* we observe a capillitium formed by straight and rigid threads with a few ramifications both in *D. niveum* and in *D. subcaeruleum* (Figs 1-2, 6), the latter taxon showing furthermore areas with a more dense ramification (Fig 2). As we have been able to study the variability of the species of the *D. niveum* complex (Moreno et al. 2003c), we agree with Neubert et al. (1995) in that the capillitium of *D. niveum* is very variable, and can even form a lax net.

All these reasons lead us to propose the synonymy of *Diderma subcaeruleum* with *D. niveum*.

Diderma globosum var. *europaeum* was described as new by Buyck (1988) after studying species of the *D. spumarioides-globosum* complex, characterized by its white, sessile fruitings seated on a well developed white, limey hypothallus: *Diderma cinereum*, *D. cingulatum*, *D. crustaceum*, *D. globosum* and *D. spumarioides*. Buyck observed differences between European and non-European species of *D. globosum*, the first ones - the new var. *europaeum* - having a capillitium similar to *D. cinereum*, with slender, flexuous threads, and the last one - the var. *globosum* - presenting capillitium features of *D. spumarioides*, i.e. having straight, more robust filaments.

Buyck considered *Diderma globosum* as a species preferring lower temperatures, and therefore relates it to *D. alpinum* and *D. niveum*, due to their same nivicolous preference. He distinguishes *D. globosum* from these two species by its white columella, somewhat smaller sporocarps and more intense iridescence of the peridial inner layer and according to him it shares with *D. niveum* spore features, having spores with one side distinctly darker by transmitted light, due to a better development of the spore wall.

According to Buyck (1982) *Diderma niveum* is not a strictly nivicolous species and has "spores densely and minutely warted, the warts often in lines so that in some specimens the spores appear subreticular." This is a key feature of *D. meyerae* that allows us to distinguish this species from *D. alpinum* and *D. niveum* (Moreno et al., 2003c). *Diderma meyerae*, however, is a strictly nivicolous species that cannot be found at lower elevations. The only thing we know for sure is that he misinterpreted *D. niveum*, which has a distinct type of episporal ornamentation.

Diderma globosum var. *europaeum* is, from our point of view, very similar to *D. alpinum*, mainly due to its flexuous, light violaceous brown to hyaline, rather slender capillitium (Fig 10) and its spores 11-12 μm in diam., with an ornamentation formed by baculae by SEM (Fig 13). The only difference is the presence of a flat and obtuse columella in *D. globosum* var. *europaeum* and a more prominent columella, subglobose to globose in the typical expression of *D. alpinum*. We consider the morphology of the sporocarps, columella and hypothallus neither constant nor reliable characters for the separation of the species of the *D. niveum* complex and think that only the spore ornamentation and the colour and morphology of the capillitium can be useful to do this (Moreno et al. 2003c). The perseverance in the examined material of the absence of the columella makes us consider it a form of *D. alpinum*, in order to facilitate its taxonomic framing.

Unfortunately, we could not study the *Diderma niveum* f. *endoleuca* described by Meylan (1924), as we have not been able to locate it in the herbarium LAU. This species is characterized by its snow-white inner peridial wall and columella. The presence of a well developed columella indicates a similarity with *D. niveum* or *D. meyerae*. Study of the capillitium

and the spore ornamentation would be necessary in order to decide this and can be made only if material determined by Meylan as *D. niveum* f. *endoleuca* can be found.

Neubert et al. (1995) described *Diderma niveum* f. *endoleuca* furthermore as having short plasmodiocarps and a light brownish, delicate capillitium and believed that *D. globosum* var. *europaeum* matches well with Meylan's description of this form.

In conclusion, we recognize three species within the *Diderma niveum* complex: *Diderma meyerae*, *D. niveum* and *D. alpinum*.

Within the species *Diderma alpinum* we recognize the following forms:

1.- *Diderma alpinum* f. *alpinum* (Meyl.) Meyl., Bull. Soc. Vaud. Sci. Nat. 51: 261. 1917

2.- *Diderma alpinum* f. *macrosporum* (Meyl.) *comb. et stat. nov.*, Bull. Soc. Vaud. Sci. Nat. 58: 319. 1935

Basionym: *Diderma alpinum* var. *macrosporum* Meyl., Bull. Soc. Vaud. Sci. Nat. 58: 319. 1935

3.- *Diderma alpinum* f. *europaeum* (Buyck) *comb. et stat. nov.*

Basionym: *Diderma globosum* var. *europaeum* Buyck, Bull. Jard. Bot. Belg. 58(1-2): 199. 1988

4.- *Diderma alpinum* f. *microcarpum* (Meyl.) G. Moreno, H. Singer & Illana, Cryptog. Mycol. 24: 57. 2003

As it is also the case in other genera of nivicolous myxomycetes where specimens with large spore size are commonly collected, we could consider *Diderma alpinum* f. *macrosporum* to be included within the concept of *D. alpinum* by simply extending range of the spore size for this taxon.

Key of the *Diderma alpinum* complex

1. Sporocarps <1 mm in diam ..... *D. alpinum* f. *microcarpum*
1'.Sporocarps >1 mm in diam. 2

2. Spores 15-17 μm in diam..... *D. alpinum* f. *macrosporum*
2'. Spores 11-13 μm in diam. 3

3. Columella subglobose to globose ..... *D. alpinum* f. *alpinum*
3'. Columella flat to broadly convex ..... *D. alpinum* f. *europaeum*

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Additional new species in the lichen family *Graphidaceae* (Lichenised Ascomycota) from the Solomon Islands

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Abstract—*Graphina maritima, Graphis alloafzelii, G. cristobalensis, G. discarpa, G. luluensis* and *Phaeographina celata* from the Solomon Islands are reported as new to science.

Introduction

In 1965 the Royal Society organised an expedition to study the natural history of the Solomon Islands and, *inter alia*, a large number of lichen specimens were collected. The results of the expedition were discussed at a meeting of the Society held in March 1968 and these were published in 1969 (Corner 1969) but no account of the lichens was included in this discussion.

The first lichen to be described from the 1965 collections was the new genus *Melanophloea* and the new species *M. pacifica* P. James & Vezda (James & Vezda 1971). Later, Stenroos (1986) identified *Cladonia didyma* (Fée) Vain., *C. macilenta* Hoffm. and *C. weymouthii* F. Wilson ex A.W. Archer among the specimens collected by Hill.

Subsequently a large number of species in the genera *Myeloconis, Porina, Pyrenula, Strigula, Tricothelium and Trypethelium* were reported by McCarthy (McCarthy 2002, 2003) based on specimens from the 1965 collections. New species in the genera *Graphina, Graphis* and *Phaeographina* have been described (Archer 2002, 2003a) and new species in the genus *Sclerophyton* were also reported (Archer 2003b).

Further examination of the specimens from the Solomon Islands showed the presence of six new species in the *Graphidaceae* which are described here.

The specimens examined were all collected by D.J. Hill between May and November 1965 and are now housed in BM. The locations of the collecting sites are shown in Corner (Corner 1969: 190, Fig. 1). Chemical constituents

were identified by thin-layer chromatography (Culberson 1972) and, where necessary, their identity was confirmed by high-performance liquid chromatography (Elix *et al.* 2003).

Taxonomic Descriptions

Graphina maritima A.W. Archer, *sp. nov* **Figure 1**

Similis *Graphina pseudoanaloga* (Vain.) Zahlbr. sed acidum norstictícum deficiens differt.

Etymology: from the Latin *maritimus*, growing by the sea.

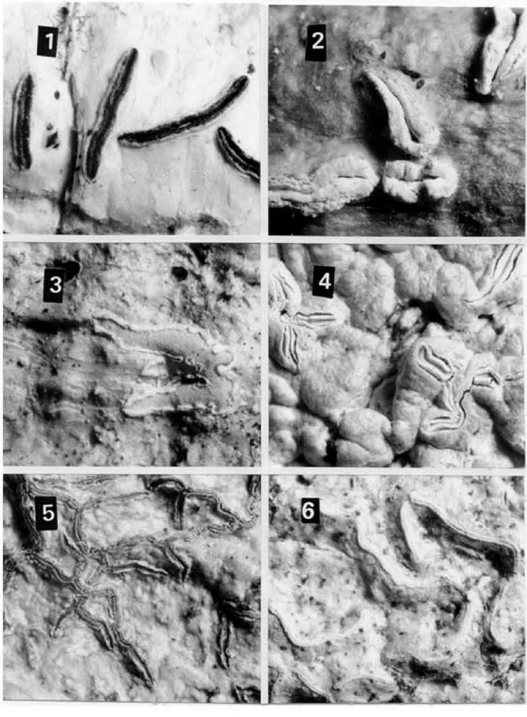
Type: **SOLOMON ISLANDS. Makira Province. San Cristobal Island**, Wainoni Region, between mouth of Huni River and headland along coast, ca. 1/2 mile [0.8 km] to the south, on trees along the beach, *D.J Hill 9821*, 15.viii.1965; holotype BM.

KEY CHARACTERS — Thallus off-white, thin, corticolous, surface smooth and dull. **Apothecia** lirelliform, conspicuous, black, numerous, scattered, simple, straight, curved or sinuous, sessile, 1–3 mm long, 0.15–0.3 mm wide, lips closed, with a conspicuous thalline margin, terminally acute. **Proper exciple** completely carbonised. **Hymenium** 100–120 µm tall, not inspersed, I-ve. **Ascospores** muriform, narrow ellipsoid, hyaline, 8 per ascus, 33–36 µm long, 10–12 µm wide, 8–10 x 2–4-locular, I+ blue.

Chemistry: no compounds found.

Ecology — *Graphina maritima* is a corticolous species found at sea-level on San Cristobal Island.

COMMENTS — *Graphina maritima* is characterised by black lirellae with a conspicuous thalline margin, the completely carbonised proper exciple and the absence of lichen compounds. The new species resemble *G. pseudoanaloga* (Vain.) Zahlbr. (Vainio 1915) from Guadeloupe but that species has an inspersed hymenium, terminally rounded lirellae [cf. Wirth & Hale 1978: 63, Plate 10a] and contains norstictic acid, in contrast to *G. maritima* which has a non-inspersed hymenium, terminally acute lirellae and lacks lichen compounds. *Graphina maritima* also resembles *G. sulcatula* Müll. Arg., but that species has sulcate lirellae and smaller ascospores. The species is so far known only from the type locality.



FIGURES 1–6. New species of Graphidaceae. **1.** *Graphina maritima*; **2.** *Graphis alloafzelii*; **3.** *G. cristobalensis*; **4.** *G. discarpa*; **5.** *G. luluensis*; **6.** *Phaeographina celata*.(Holotypes in BM). Magnification: 14x.

Graphis alloafzelii A.W. Archer, *sp. nov.* **Figure 2**

Similis *Graphis afzelii* Ach. sed acidum psoromicum continens vice acidum lecanoricum differt.

Etymology: from the Greek *allos*, other, and the species resemblance to *Graphis afzelii*.

Type: **SOLOMON ISLANDS. Western Province. Kolombangara Island**, south summit, montane moss forest, 5400 ft [1650 m], *D.J. Hill 10472*, 2.ix.1965; holotype: BM.

KEY CHARACTERS — Thallus thin, pale reddish brown, corticolous, surface smooth and shiny. **Apothecia** lirelliform, white, black where abraded, conspicuous, scattered, sessile, simple, straight, curved or sinuous, 1–3 mm long, 0.25–0.6 mm wide. **Proper exciple** laterally carbonised, covered by a white powdery layer. **Hymenium** 100–120 µm tall, not inspersed. **Ascospores** 8 per ascus, ellipsoid, hyaline, 18–20 µm long, 8–10 µm wide, 4-locular, I-ve.

Chemistry: lirellae: C-ve, Pd+ yellow; psoromic acid (major) and 2'-*O*-demethylpsoromic acid (minor).

Ecology: *Graphis alloafzelii* is a corticolous species found at 1650 m on Kolombangara Island.

COMMENTS — *Graphis alloafzelii* is characterised by simple white lirellae, 4-locular ascospores and the presence of psoromic acid. The new species closely resembles *G. afzelii* Ach., a widely distributed tropical to subtropical species which also occurs in the Solomon Islands [San Cristobal Island, at the mouth of the Huni River at sea level, *D.J. Hill 9019*, BM] but contains psoromic acid in place of the lecanoric acid found in *G. afzelii*. The lecanoric acid in *G. afzelii* is contained in the white outer coating of the lirellae as is the psoromic acid in *G. alloafzelii*. The new species is distinguished from the somewhat similar *Graphina hillii* A.W. Archer (Archer 2002) by the 4-locular ascospores, muriform in *G. hillii* and the chemistry; *G. hillii* contains subpsoromic acid. The new species is so far known only from the type locality.

Graphis cristobalensis A.W. Archer, *sp. nov.* **Figure 3**

Similis *Platythecium acutisporum* Staiger sed ascosporis apicibus rotundis vice acutis et excipulo pallido-badio differt.

Etymology: from the Latin *ensis*, place of origin, and the type locality, San Cristobal Island.

Type: **SOLOMON ISLANDS. Makira Province. San Cristobal Island**, N of Ugi Island, near Pawa School, ca. 1/2 mile [0.8 km] along path inland and into jungle, alt. *ca.* 50ft [15 m], *J.D. Hill 8706*, 12.viii.1965; holotype: BM.

KEY CHARACTERS — Thallus pale fawn, corticolous, surface smooth and shiny. **Apothecia** lirelliform, conspicuous, sessile in irregular clusters *ca.* 3 x 4 mm, individual lirellae 0.25–0.4 mm wide, with a conspicuous, raised, white thalline margin. **Epithecium** pale grey, fine white-pruinose. **Exciple** complete, thin, pale reddish brown, **Hymenium** 100–120 µm tall, pale brown, not inspersed, I-ve. **Ascospores** 8 per ascus, elongate-ellipsoid, hyaline, 13–16 µm long, 5–6 µm wide, 4-locular, I+ pale blue.

Chemistry — testacein.

Ecology — *Graphis cristobalensis* is an uncommon corticolous species found at sea-level at *ca.* 15 m elevation in secondary and disturbed primary rainforest on San Cristobal Island.

COMMENTS— *Graphis cristobalensis* is characterised by conspicuous, sessile, open lirellae, small 4-locular ascospores and the presence of testacein. This compound is uncommon in the Graphidaceae and is found in *Graphina allosporella* (Nyl.) Müll. Arg. (Nylander 1869), *G. sphaerosporella* (Nyl.) Müll. Arg. (Nylander 1869) and *Platythecium acutisporum* Staiger (Staiger 2002). The new species is distinguished from the two *Graphina* species by the 4-locular ascospores, and from the *Platythecium* species by terminally rounded ascospores. In addition, the exciple in *G. cristobalensis* is pale brown, in contrast to that of *P. acutisporum* which is carbonised. The new species is so far known only from two specimens, from the same locality, with the same collector's number.

Graphis discarpa A.W. Archer, *sp. nov.* **Figure 4**

Similis *Graphis descissa* Müll. Arg. sed lirellis interstinctis et ascosporis majoribus.

Etymology: from the Latin *dis*, separate, and the Greek *karpos*, fruit, a reference to the separated lirellae.

Type: **SOLOMON ISLANDS. Western Province. New Georgia Group**, Marovo Lagoon, Paleki Island, sea-level, *D.J. Hill 8400*, 3.viii.1965; holotype: BM.

KEY CHARACTERS —Thallus pale olive-green, corticolous, surface tuberculate and dull. **Apothecia** lirelliform, numerous, scattered, immersed,

visible as a thin black line with white margins separated from the thallus, simple, straight, curved or sinuous, rarely branched, lips closed, 1–3 mm long, 0.15–0.3 mm wide. **Proper exciple** completely carbonised. **Hymenium** 160–180 µm tall, not inspersed, I-ve. **Ascospores** narrowly ellipsoid, hyaline, 8 per ascus, irregularly 2-seriate, 38–48 µm long, 8–10 µm wide, 8–11 locular, I+ blue.

Chemistry — stictic acid.

Ecology — *Graphis discarpa* is an uncommon corticolous species found on tree trunks at sea-level on Paleki Island, New Georgia Group.

COMMENTS — *Graphis discarpa* is characterised by lirellae immersed in, but separated from, the thallus, the completely carbonised exciple and the presence of stictic acid. The new species is distinguished from *G. descissa* Müll. Arg. (Müller 1895) by the sharply separated, immersed lirellae and the larger ascospores, 38-48 µm long in contrast to *G. descissa* which has subsessile lirellae and ascospores 28-38 µm long. *Graphis discarpa* somewhat resembles the chemically similar *G. bougainvillei* Zahlbr. (Zahlbruckner 1912) but that species has immersed lirellae, not separating from the thallus, the exposed exciple is slightly white-pruinose, and it has smaller ascospores, 30-35 µm long. The new species is differentiated from *G. immersella* Müll. Arg. (Müller 1895), which also occurs in the Solomon Islands, by the completely carbonised exciple, in contrast to it being laterally carbonised in *G. immersella*.

The new species is so far known only from two specimens (with the same collector's number) from the same locality.

Graphis luluensis A.W. Archer, *sp. nov.* **Figure 5**

Similis *Graphis centrifuga* Räs. sed ascosporis majoribus et acidum stictícum continens vice acidum norstictícum differt.

Etymology: from the Latin, *ensis*, place of origin, and Lulu Island, the type locality.

Type: **SOLOMON ISLANDS. Western Province. New Georgia Group**, Marovo Lagoon, west side of Lulu Island, sea-level, *D.J. Hill 8667*, 8.viii.1965; holotype: BM.

KEY CHARACTERS — Thallus pale fawn, thin, corticolous, surface smooth and shiny. **Apothecia** lirelliform, conspicuous, numerous, crowded, immersed, curved and sinuous, much-branched, lips closed, sometimes becoming open, 1.5–4 mm long, 0.1–0.2 mm wide. **Proper exciple** completely carbonised. **Hymenium** 100–120 µm tall, inspersed, I-ve;

Ascospores 8 per ascus, hyaline, 25–32 mm long, 6–7 µm wide, 6–8-locular, I+ blue.

Chemistry — stictic acid.

Ecology — *Graphis luluensis* is an uncommon corticolous species found at sea-level on Lulu Island, New Georgia Group.

COMMENTS— *Graphis luluensis* is characterised by the crowded, much branched lirellae, the completely carbonised exciple and the presence of stictic acid. The new species resembles the Australian species *Graphis centrifuga* Räs. (Räsänen 1949; Archer 1999; 288, Fig. 4a) but differs from that species in having larger ascospores and containing stictic acid. *Graphis luluensis* is distinguished from the chemically similar *G. descissa* Müll. Arg. (Müller 1895) and *G. dendrogramma* Nyl. (Nylander 1877) by the smaller ascospores and the completely carbonised exciple, respectively, and from *G. propinqua* Müll. Arg. (Müller 1882) by the presence of stictic acid. *Graphis luluensis* also resembles the chemically similar *G. modesta* Zahlbr. (Zahlbruckner 1912) from Bougainville. Both species possess ascospores of a similar size [20–6 x 6–8 µm, 6–8-locular in *G. modesta*] but the exciple in *G. modesta* is laterally carbonised with open lips revealing a white-pruinose disc, in contrast to the completely carbonised exciple with closed lips of *G. luluensis*.

The new species is so far known only from the type locality from Lulu Island.

Phaeographina celata A.W. Archer, *sp. nov.* **Figure 6**

Similis *Phaeographina quassiicola* (Fée) Müll. Arg. sed lirellis semi-immersis, ascosporis minoribus et acidum norsticticum continens differt.

Etymology: from the Latin, *celatus*, hidden, a reference to the inconspicuous, semi-immersed lirellae.

Type: **SOLOMON ISLANDS. Makira Province. San Cristobal Island**, Wainoni Region, *ca.* 2 miles [3.2 km] SE of Wainoni Mission, alt. 1500–1600 ft [460–490 m], *D.J. Hill 8869*, 14.viii.1965; holotype: BM.

KEY CHARACTERS — Thallus of-white to pale fawn, thin, corticolous, surface smooth and shiny. **Apothecia** lirelliform, inconspicuous, concolorous with the thallus, semi-immersed scattered, curved or sinuous, sometimes branched, 2–5 mm long, 0.3–0.5 mm wide, lips closed. **Proper exciple** black, completely carbonized, thick, covered with a thin thalline coating. **Hymenium** 150–180 µm tall, not inspersed. **Ascospores** 8 per ascus,

muriform, brown, narrowly ellipsoid, 40–48 µm long, 14–18 µm wide, 8–10 x 1–4-locular.

Chemistry: norstictic acid.

Ecology — *Phaeographina celata* is an uncommon corticolous species found on *Casuarina papuana* in disturbed primary rainforest at *ca.* 500 m on San Cristobal Island.

COMMENTS — *Phaeographina celata* is characterised by pale grey, inconspicuous lirellae, the thick black exciple, the pale brown, muriform ascospores and the presence of norstictic acid. The new species resembles *P. quassiicola* (Fée) Müll. Arg. in that both species have have brown, muriform ascospores and a complete carbonised exciple covered by a thin thalline layer. However, *P. celata* has smaller ascospores, 40–48 µm long, compared to those in in *P. quassiicola* which are 55–103 µm long (Staiger 2002: 449) and, in addition, contains norstictic acid; *P. quassiicola* lacks lichen compounds.

The new species is so far known only from the type locality on San Cristobal Island.

A CORRECTION

In a recent paper describing new *Graphidaceae* from the Solomon Islands (*Mycotaxon* **83**: 366 (2002) the holotype of *Phaeographina amnicola* was incorrectly reported to be Hill 8133; the correct specimen number is Hill 8132.

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Hydropus kauffmanii, first records from Europe

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Abstract—*Mycena kauffmanii* is reported for the first time from Europe, where it has been collected under neutrophilic *Salix* spp. from two sites in the Northern French Alps and one on the French littoral of the Channel. The species is compared to the related *Hydropus floccipes*, and its ambiguous systematic position in the genus *Hydropus* is discussed. The European material is shown to be identical to North American collections, including the holotype. The new combination, *Hydropus kauffmanii*, is proposed.

Résumé—*Mycena kauffmanii* est signalé pour la première fois en Europe, à partir de récoltes effectuées dans deux localités des Alpes françaises septentrionales et une localité du littoral de la Manche, sous buissons de saules neutrophiles. Il est comparé à l'espèce voisine *Hydropus floccipes*, et sa position systématique ambiguë au sein du genre *Hydropus* est discutée. Le matériel original d'Amérique du Nord (y compris l'holotype) est identique au matériel européen. Une nouvelle combinaison est proposée: *Hydropus kauffmanii*.

Key-words—Agaricales, Basidiomycota, Tricholomataceae

Introduction

The genus *Hydropus* Kühner ex Singer, which occurs worldwide, is abundant in tropical areas (Singer 1982) but poorly represented in temperate areas. In Europe, about 15 taxa are recognized (Hausknecht *et al.* 1997; Contu & Robich 1998; Bas 1999; Esteve-Raventós et al. 2002). Most are rare and limited to individual ecological habitats: *H. marginellus* (Fr.: Fr.) Singer and *H. atramentosus* (Kalchbr.) Singer occur only on rotten *Abies* stumps, *H. subalpinus* (v. Höhn.) Singer on buried *Fagus* branches, *H. dryadicola* (Kühner) E. Horak in calcareous alpine meadows, *H. paradoxus* var. *xerophyticus* Esteve-Rav. *et al.* in xerophytic grasslands. Some recently described species are known only from a few collections: *H. conicus* Bas & Weholt, *H. moserianus* Bas, *H. nitens* Maas Geest. & Hauskn., *H. paradoxus* M.M. Moser.

Moreover, several taxa initially described in *Hydropus* have been recognized as belonging to other genera. For instance, Robich (2001) treats *Hydropus floccipes* var. *montis-rosae* Jamoni & Bon in *Mycenella*, while Contu (2001) and Eyssartier & Moreau (2001) treat *H. liciosae* Robich & Contu in *Lactocollybia*.

Within the genus *Hydropus*, *H. floccipes* seems to have the widest ecological range and geographical distribution in western Europe, although it clearly shows Mediterranean trends (Malençon & Bertault, 1975; Robich, 1990; Moreau et al., 1999). Several colour variations have been described (Malençon & Bertault, 1975; Ortega-Diaz & Zea, 1991; Perez-de-Gregorio, 2001) that are reported as microscopically identical to the type.

While exploring peat bogs and mires for his PhD fieldwork, the first author (PAM) gathered three mycenoid collections that were first interpreted as representing an unusual form of *H. floccipes* with conspicuous black-edged gills. A thorough study revealed several differences clearly separating our collections from typical *H. floccipes*. Although no detailed European description fit our material [but see below regarding Einhellinger (1976)], a matching description was found in Smith's (1947) North American monograph under the name *Mycena kauffmanii* A.H. Sm. (1936), a name also cited by Kühner (1938). The descriptions given by Smith, as well as earlier notes by Kauffmann (1918: 792) under the name "*Mycena denticulata* Peck", match perfectly our specimens. Our study of the three abundant collections (including the type) from Smith's herbarium (MICH) fully confirms our initial identification.

Below we describe our collections, after which we compare our concept first with Smith's description of *Mycena kauffmanii* and then with *H. floccipes*.

Material and methods

Studied collections of *H. kauffmanii*: Europe -- France. Isère. Saint-Laurent-du-Pont (M. E. N. 3233D), « tourbière de l'Herretang », alt. 400 m, under *Salix cinerea* and *S. aurita*, on naked soil mixed with twigs of *Salix*, close to *Peziza succosa* var. *infuscata*, 01-IX-2000, herb. P.-A. M. n° 00090110 (ZT); same location, on twigs and pieces of wood of *Salix* buried in a deep dry humus, 04-VI-2001, leg. J.-C. Déjana & P.-A. Moreau, herb. P.-A. M. n° 01060401 (ZT) **Fig. 1, 4. Savoie.** Saint-François-de-Sales (M. E. N. 3332B), "Tourbière des Creusates", alt. 1350 m, under a lone *Salix cinerea* on a piece of twig, in a damp neutrophilic mire with *Molinia caerulea*, *Aulacomnium palustre*, *Tomenthypnum nitens* and *Salix repens*, 10-VII-2001, herb. P.-A. M. n° 01071006 (ZT). **Fig. 3-8. Pas-de-Calais.** Merlimont (M. E. N. 2105C), Réserve biologique domaniale, alt. 5 m, in a *Salix* bush (*Salicion auritae*) on alcaline peat, 01-VIII-2003, leg. R. Courtecuisse & P.-A. Moreau, herb. RC/F03.192 (LIP). **North America -- United States. Michigan: Dexter.** Silver Lake, alt. 950 ft, on moss, 11-VI-1934, herb. A. H. Smith n° 13 (NYC 11499, MICH, holotype !). **Fig. 2. Cheboyan Co.** Wolf bog, 29-VI-1941, herb. A. H. Smith n° 23264 (NYC 32364, MICH). **Lakeland.** 24-VI-1935, herb. A. H. Smith n° 1408 (NYC 1408, MICH) [note: collection studied by Kühner, 1937]

Macroscopical and microscopical observations on French material were made on fresh material in H_2O and in Congo red and completed on dry material. Smith's material was rehydrated in 5% KOH before observation. Cyanophily of spore wall was tested in Cotton blue-lactic acid solution, amyloidity and dextrinoidity were tested in Melzer's reagent, and tissue metachromasy was tested in Cresyl blue. Colour reference standard: Séguy (1936). Soil nomenclature following Baize & Girard (1995).

Taxonomy

Hydropus kauffmanii (A. H. Sm.) P.-A. Moreau & Courtec., *comb. nov.*
Basionym: *Mycena kauffmanii* A. H. Sm. 1935, Mycologia 27: 588

Description of European collections

Pileus 0,8-2 cm diam., conical-campanulate, then somewhat flattened or umbonate, often depressed around the umbo, with margin remaining straight a long time; edge exceeding, then revoluted, surface dull, almost velvety, not sulcate or becoming so only at late stage, fibrillose-looking in mature specimens; colour uniform and constant, sepia brown (Seguy 116), umbo often somewhat darker.
Lamellae 26-36 arising from the stipe, 2 series of lamellulae, almost free to adnate-uncinate with age or sometimes arcuate-subdecurrent; the faces markedly venose in mature specimens; colour pure white, with pruinose, obviously black-punctate edge.
Stipe 3-7 x 0,1-0,3 cm; light brownish grey, entirely and densely black-powdered, fistulose, insititious on superficial woody debris or +/- rooting (up to 3 cm deep).
Context white, slightly elastic, not fragile, weakly succulent when cut. Smell fruity-acidic, reminding *Fomitopsis pinicola*. Taste none.
Spores (5,5) 6,5-7,5 x 5,5-6,5 μm (8,5-10 x 7,5-8,2 μm from 2-sp. basidia), Q = 1,1-1,3, globose to subglobose in spore print, but often widely ellipsoid on hymenium, with a large apiculus up to 1-1,5 μm long; inamyloid, non-cyanophilic, with 1-guttulate oily content (fresh material). **Basidia** 22-35 x 6-11 μm, 4-spored in PAM 00090110 and 01060401, equally 2-4-spored with a number of 1-3-spored basidia too in PAM 01071006, clavate to clavate-pedonculate with elongated basis, all clamped; sterigmata prominent, up to 5-6 μm long, but monosterigmatic basidia not infrequent with sterigmata up to 8 μm. Subhymenium 15-20 μm thick, ramose with short dichotomously branched elements 2-3 μm wide; basidioles abundant, some undulate-fusiform and shortly protruding. **Cheilocystidia** numerous, 35-55 x 6-13 μm, rather short, clavate or lageniform (sometimes with elongated neck), to cylindrical, subfusiform or with +/- vermiform neck, the lower part always thick and constant in shape; intracellular content abundant, grey-brownish pigmented, especially in the longest cystidia. **Pleurocystidia** absent.
Pileipellis a trichoderm, with densely erected elements towards the disc, these being more scarce and adpressed towards the margin; suprapellis 70-110 μm thick at disc, with abundant pileocystidia 25-80 x 7-15 μm, often arising from deep layers of subpellis, versiform, lageniform or lageni-fusiform to cylindrical or narrowly clavate, mostly with abundant grey-brown intracellular pigment. Subpellis 80-120 μm thick, clearly differentiated, with globose to ellipsoid, sometimes clavate or utriform elements 30-90 x 18-35 μm, with diluted intracellular pigment, mixed with thin, inconspicuous hyphae x 2-3 μm; no gloeoplerous hyphae seen.
Pileus context with long, fusiform elements up to 300 (500) x 15-25 (35) μm, mixed with thin generative hyphae x 2-3 μm; all elements pigmentless, but with scarcely guttulate content.
Lamellar trama parallel, with the same structure than cap context, the fusiform elements up to 300-1200 μm long.
Stipitipellis a cutis of smooth hyphae x 3-5 μm, without differentiated cortex, with numerous fascicles of 10-25 caulocystidia, 40-65 x 5-12 μm, mostly clavate, smooth,

with dark grey-brown, intracellular pigment, mixed with short and scarce filiform diverticules up to 25 μm long. Stipe context with parallel, poorly differentiated hyphae x 3-5 μm, mixed with gloeoplerous hyphae showing refractive content, x 3-4 μm, conspicuous but not abundant. No part of basidiomata metachromatic or dextrinoid on fresh material; basal part of the stipe weakly dextrinoid in *exsiccata*.

Clamps abundant in all tissues, sometimes scarce in context and among pileipellis hyphae, often very large.

Ecology: found twice (Saint-Laurent-du-Pont, Isère, F) in a mesotrophic *Salix cinerea-S. aurita* stand, with neutral pH (6,5-7); the site is an alkaline peat bog with 40-60-year-old willow bushes developed after drainage; the initial hydromull humus has become a deep dysmull with abundant litter, rich in twigs and small woody pieces of *Salix*. Basidiomata were abundant on the woody debris, with 12-18 gregarious specimens discovered at 2 different sites each time). Found once (Les Creusates, St-François-de-Sales, F) in the neutrophilic part of a mixed peat bog under a lone *Salix cinerea* surrounded by a *Molinia caerulea-Aulacomnium palustre* dominated association with pH about 5,0 (related to *Caricion canescenti-fuscae*); the only specimen was linked to a small woody remnant of *Salix*. The last record was collected from an alkaline *Salix* stand in a coastal dune system with *Populus tremula* and *Betula pubescens* also present. The single specimen grew in a rather thick litter of leaves and woody twiglets of *Salix aurita*. Although quite distant from the first locality, the ecological conditions so resembled Saint-Laurent, that the first author (PAM) jokingly predicted finding the species just a few minutes before the second author (RC) actually found the small specimen. The species is assumed to be lignicolous and associated with *Salix*, growing on small pieces of wood buried in neutrophilic to basic peaty humus. It may fructify from June to September.

Observations on type specimens

Three Smith collections preserved in NYC were studied; all were found to conform perfectly to Smith's (1936, 1947) descriptions. The holotype (of which only one½ specimen was examined) matches exactly all microscopical features found in the European material; 2-spored basidia are scarce, but large spores up to 10 x 8 μm are not rare on the hymenium and due to age, the suprapellis of the type specimen has thinned so that a dense trichoderm is present only in patches. Collection AHS1418, was examined and well described by Kühner (1938: 546). Collection AHS32364 is beautiful (more than 30 specimens in perfect condition) and none of the 5 specimens studied showed divergences with the previously observed ones. It should be noted that all A. H. Smith exsiccati have butter-yellow lamellae and red-brownish pilei, somewhat lighter than European material.

Discussion

Mycena kauffmanii was described by Smith (1936, 1947) as occurring "around elm and ash stumps or on humus in elm and ash swamps", after Kauffman (1918: 792) described it erroneously as "*Mycena denticulata* Peck". Kühner (1938: 546) refined the description and provided spores and cheilocystidia drawings after examining Smith's material. We confirmed Kühner's observation of a weak amyloidity and purplish-violaceous coloration on walls of stipe hyphae in Cresyl blue in the American material, but these reactions were very weak (also on dry material, only cortex ochre-reddish in

Melzer) in our French material. This last character can be discussed in comparison with the closely related *H. floccipes*, where amyloidity and metachromasy can be present or not (Malençon & Bertault, 1975: 261).

Undoubtedly, our collections of *Mycena kauffmanii* are strongly reminiscent of *Hydropus floccipes* (Fr.: Fr.) Singer. Although we are well acquainted with *H. floccipes* (Moreau et al., 1999), we first thought we had found it in an unusual habitat. But our attention was caught by the black-serrulate gill edge, no mention of which was found in any of the usual descriptions of *H. floccipes* (Maire, 1928: 41 – as *Mycena maura*; Kühner, 1938: 540; Smith, 1947: 380; Singer, 1982: 118; Bon & Chevassut, 1989: 33; Robich, 1990: 317; Hausknecht et al., 1997: 186; Pérez-de-Gregorio in Societat Catalana de Micologia, 1998: 823; Watling & Turnbull, 1998: 132). Microscopical differences are sufficient to exclude identifying our collections as any of the described varieties or forms of *H. floccipes*; the densely erected pileal trichodermium, the lack of pleurocystidia and gloeoplerous hyphae in gill trama, and the non strictly globose spores all exclude *H. floccipes*. The two species are compared below in **Table 1**.

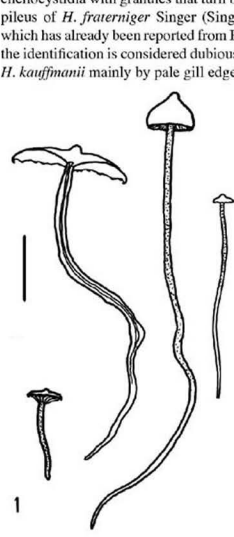
Smith (1947: 379), in his key to sect. *Floccipedes*, emphasizes the black gill edge of *H. kauffmanii* as the determinant character for segregating it within the section. In fact, Smith does not write many lines to compare *M. kauffmanii* with *M. floccipes*, since the microscopical descriptions are obviously different for him. The pileipellis structure looks very distinctive, and the numerous "cystidium-like projections" from hypodermium explain the "subvelvety" surface. This is actually a major difference with *H. floccipes*, which shows a typical cutis-structure; in fact, we observed in our material of *H. kauffmanii* that the typical *Hydropus* structure with parallel inflate hyphae is present, but widely dominated by emergent pileocystidia arising from different layers of subpellis as described by Smith. The remarkable development of pseudoparenchymatous subpellis, which reminds of a typical *Mycena*-structure, is a unique feature as far as European *Hydropus* are concerned.

The deeply rooting stipe is also emphasized for *M. kauffmanii* in the key and discussion by Smith (loc. cit.), but this feature can be influenced by the buried substrate. In the hereby described collections, only some of the specimens were linked with deeply buried *Salix* twigs (up to 3 cm deep), and then the stipe was rooting as described and illustrated by Smith; but other specimens fixed on only slightly buried twigs do not show conspicuous pseudorrhiza. The same substrate-dependent polymorphy can be observed in *H. floccipes*, usually growing on bark but sometimes arising from buried pieces of wood and then with pseudorrhiza (Malençon & Bertault, 1975: 261; Bas, 1999: 167; see Moreau et al., 1999, pl. 1A).

Smith (1947) does not mention explicitly the absence of pleurocystidia in *M. kauffmanii* (but Kauffman, 1918: 793, writes "Cystidia none" in his description of *M. denticulata*) although he mentions them abundant in *H. floccipes*; he does not mention either gloeoplerous hyphae for both species. This difference in pleurocystidia is nevertheless conspicuous, but should be derived from the absence of gloeoplerous hyphae in the subhymenium of *H. kauffmanii*, when the pleurocystidia (pseudocystidia) of *H. floccipes* are issued from the numerous gloeoplerous hyphae running into its subhymenium (Moreau et al., loc. cit.).

In recent times, no mycenoid specialist seems to have reported on *Mycena kauffmanii*, and the combination in *Hydropus* Singer seems still unpublished. *H. kauffmanii* undoubtedly belongs in that genus because of its typical sarcodimitic trama, intracellular pigmentation, and presence of cheilocystidia. Subglobose, non-amyloid spores place it in sec. *Floccipedes* Kühner ex Singer, subsec. *Floccipedes* according to the European classification (Contu & Robich, 1998, Bas, 1999); but Singer's classification (1982: 53) based on a hymenodermic pileipellis and absence of pleurocystidia would place it insect. *Mycenoides* Singer, subsect. *Anthidepades* Singer, which has not yet been reported from temperate areas. Because the subsect. *Anthidepas* looks highly artificial and contains species that share only negative characters (inamyloid spores and lack of pleurocystidia; Singer, 1982), we prefer to follow the conservative path by retaining *H. kauffmanii* in sect. *Floccipedes* sensu Smith (1947).

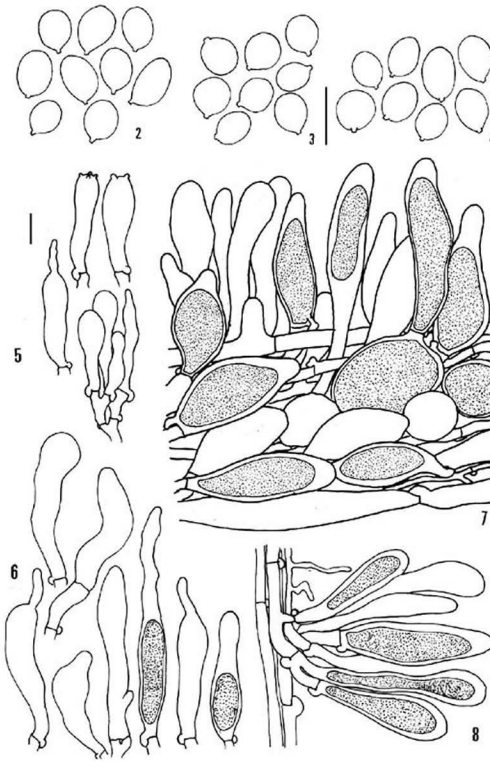
In Singer's (1982) monograph of South American species of *Hydropus*, *H. kauffmanii* would key out closely to another species sharing dark colours and black edges: *H. nigromarginatus* Singer, which differs in producing decurrent pinkish gills and cheilocystidia with granules that turn blackish to lilac in iodine. The dark and granulose pileus of *H. fraterniger* Singer (Singer, 1946) also resembles that of *H. kauffmanii*, which has already been reported from Europe (Austria: Hausknecht et al. 1997, although the identification is considered dubious by Esteve-Raventós et al., 2002); it differs from *H. kauffmanii* mainly by pale gill edge, 2-spored basidia and amyloid spores.



Another member of subsect. *Floccipes* should be *Mycena ulmicola* A.H. Sm. (Smith, 1939: 281; 1947: 382), which seems to be as neglected as *M. kauffmanii* in current literature. According to Smith's discussion, its characters are closer to those of *H. floccipes*, except small spores (3,5-4 μm) and rarefaction of pleurocystidias from the edge. A collection from Switzerland (Favre, 1957) is the only report of *M. ulmicola* from Europe, with some minor divergences in microscopical features.

Our collections of *H. kauffmanii* should not be the first ones from Europe: Einhellinger (1976) briefly reports under the name "*Hydropus floccipes*" a collection from alkaline bog in Bavaria, with black gill edge, which could fit with *H. kauffmanii*. Unfortunately no description is published of this collection, and no material could be found in Einhellinger's herbarium in Munich (D. Triebel, pers. comm.).

Fig. 1: *Hydropus kauffmanii*. Sporophores (PAM 01060401). Scale = 10 mm.



Figs. 2-8: *Hydropus kauffmanii*. 2: spores (holotype, NYC 11499). 3: spores (PAM 01060401). 4: spores (PAM 01071001). 5: basidia and basidioles; 6: cheilocystidia; 7: pileipellis (radial cut); 8: stipitipellis (PAM 01071001). Scale = 10 μm.

Table 1. Comparison of distinctive characters in *Hydropus floccipes* and *H. kauffmanii*

	H. floccipes	*H. kauffmanii*
Cap	Smooth, slightly fibrillose.	Matte, almost velvety at least at centre, later fibrillose-radiate.
Gills	Pure white, edge pruinose, white.	Pure white, edge pruinose, but with black punctuations along the edge.
Spores	5,5-7,5 μm, globose.	6,5-7,5 x 5,5-6,5 μm, globose to widely ellipsoid.
Cheilocystidia	40-60 x 6-8 μm, with hyaline content.	35-55 x 6-13 μm, most with brownish content on fresh and dried material.
Pleurocystidia	60-100 x 12-22 μm, very numerous.	Absent.
Pileipellis structure	10-15 μm thick, a cutis with inflated cystidia-like protuberances up to 25 μm long. Subpellis filamentous, with long hyphae 60-150 x 8-20 μm.	70-110 μm thick, a trichoderm with cystidia arising from subpellis, mixed with scarce thin, radial hyphae. Subpellis pseudoparenchymatous, with globose to shortly ellipsoid hyphae 30-90 x 18-35 μm.
Caulocystidia	30-40 x 5-10 μm.	40-65 x 5-12 μm.
Gloeoperous hyphae	Very abundant in subpellis, gill and stipe trama.	Abundant only in stipe trama, rare and scarce in subpellis and gill trama.
Habitat	On bark or rotten wood, usually of *Quercus spp.*, rather thermophilous.	On buried twigs of *Salix, Ulmus, Fraxinus*, in wet habitats.

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Revision and nomenclature of several boletes in China

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Abstract— Revision of several boletes in China is reported with proposals of a new name and two new combinations. *Boletus atratus* is proposed as *nomen novum* to replace *B. nigricans* M. Zang, M. S. Yuan & M. Q. Gong, which is a later homonym of *B. nigricans* Pat. & Baker. This is considered as a recognizable species based on examination of the type material from China in comparison with specimens of *B. nigerrimus* and *Tylopilus alboater*. *Boletinus cavipoides* and *B. kunmingensis* are transferred to *Suillus* as the new combinations *S. cavipoides* and *S. kunmingensis* respectively, based on the characters of viscid pileus and the absence of clamp connexions in tube trama hyphae. A revision of the type material of *S. pinetorum* is also presented to extend the species description and to clarify the nomenclature of the species.

Key words—*Boletales*, *Boletus* sect. *Nigrex*, type revision

中文摘要

本文报道对中国产的4种牛肝菌的研究结果，提出了1个新名称和2个新组合。建议以新名称*Boletus atratus*取代*B. nigricans* M. Zang, M. S. Yuan & M. Q. Gong（黑牛肝菌，汉语学名保留不变），因为后者是*B. nigricans* Pat. & Baker的晚出同名。经与近缘种*B. nigerrimus*（黑紫牛肝菌）和*Tylopilus alboater*（黑盖粉孢牛肝菌）等进行比较，确认*Boletus atratus*是一个独立的种。根据菌盖胶粘及菌管菌丝不具锁状联合等特征，将*Boletinus cavipoides*（美小牛肝菌）和*B. kunmingensis*（昆明小牛肝菌）归并到*Suillus*（铆盖牛肝菌属）中，构成*Suillus cavipoides*（美小粘盖牛肝菌）和*S. kunmingensis*（昆明粘盖牛肝菌）两个新组合。基于模式标本的观察，本文也对*S. pinetorum*（松林粘盖牛肝菌）进行了重新描述，并对中国文献中出现的命名法问题作了讨论。

Introduction

The boletes, or fleshy pore fungi, are a group of fungi mostly living symbiotically with plants, forming ectomycorrhizal associations (Smith & Thiers 1971, Corner 1972,

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Singer 1986, Bessette et al. 2000). Ectomycorrhizal basidiomycetes, including most boletes, constitute an important component of forests fungal communities (Dahlberg et al. 1997, Smith & Read 1997, Baxter & Dighton 2001) and count greatly in the ecosystem of forest. Many species are edible and delicious (Singer 1986, Smith & Thiers 1971, Bessette et al. 2000), although some others are poisonous. Boletes are the main wild fungi exported in large quantity from China to Japan, Italy, France, Switzerland, etc. The exported boletes (including their products) from a province of China, Yunnan, reached 6053.2 tons in 2000, generating US$ 19.3 million of income for the province (www.unn.com.cn/GB/channel23/176/).

Boletes are usually placed in *Boletales* E.-J. Gilbert 1931, with about 1025 species worldwide (Kirk et al. 2001), with more than 350 species distributed in China (Li & Song 2000). Investigation by Chinese mycologists of species of *Boletus* Fr. 1821, *Xerocomus* Quél. 1888 and *Boletinus* Kalchbr. 1867 in China has revealed several new species in *Boletinus* (e.g. Chiu 1948, Zang 1980, Bi et al. 1982) and one in *Boletus* (Zang et al. 1993). Among the new species described from China, *B. nigricans* M. Zang, M. S. Yuan & M. Q. Gong 1993 is found to be a later homonym of *B. nigricans* Pat. & Baker 1918. Re-examination of holotypes of these taxa and other relevant specimens preserved in Herbarium, Kunming Institute of Botany, Chinese Academy of Sciences (HKAS), Mycological Herbarium, Institute of Microbiology, Chinese Academy of Sciences (HMAS) and Herbarium, Guandong Institute of Microbiology (HMIGD) has provided some noteworthy information on several boletes. The results are presented in this paper.

Taxonomy

Boletus atratus Q. B. Wang & Y. J. Yao, **nom. nov.**
≡*Boletus nigricans* M. Zang, M. S. Yuan & M. Q. Gong in *Acta Mycol. Sin.* 12: 278 (1993). non *Boletus nigricans* Pat. & Baker in *J. Straits Branch R. A. Soc.* 78: 70 (1918).

Etym.: atratus, black; referring to the color of the fruitbody.

Pileus 6.0–9.0 cm in diam., subconvex to plane, velvety-tomentose, aerolate, dark brown to black. *Context* 1.0 cm thick at stipe, dark gray brown, taste and odor mild. *Tubes* 0.6–1.0 cm long, dark brown, depressed around the stipe apex; pores 9–11 pores/cm, multi-angular, concolorous with the tubes. *Stipe* 6.5–8.5 cm long, 0.7–1.2 cm thick, enlarged downward, concolorous with the pileus, reticulate at least on the upper half, solid. *Basidiospores* 9.0–11.0 x 4.0–5.0 μm, broad ellipsoid, thin-walled, olive brown to brown. *Basidia* 26–36 x 8.0–11.0 μm, clavate, 2- and 4-spored. *Pleurocystidia* 42–63 x 9.0–19.0 μm, thin-walled, fusiform. *Tube trama* poorly rehydrating, hyphae brownish in KOH. *Clamp connexions* absent.

Specimen examined: **China: Sichuan Province,** Pujiang County, Datang, alt. 650 m, 10 June 1991, M. S. Yuan No. 1334, HKAS 23843 (HOLOTYPE).
Habit and habitat: Scattered to solitary in the forest under *Pinus massoniana* and *Camellia oleifera*.
Distribution: China, Sichuan Province (type locality).

The above description is based on the holotype deposited in HKAS. For detailed description of field characters, please see Zang et al. (1993). However, the stipe color was not mentioned by Zang et al. (1993). According to the type material and the color photographs in Yuan & Sun (1995), the stipe is concolorous with the pileus. Based on this species, Zang et al. (1993) proposed a new section within the genus *Boletus*, *B.* sect. *Nigres* M. Zang, which is mainly characterized by the dark context, unchanging tube color when bruised and the reticulate stipe. Six species with diverse characters, i.e. *B. badius* Fr. 1828, *B. brunneissimus* W. F. Chiu 1948, *B. nigerrimus* Heim 1963, *B. tomentulosus* Zang et al. 1991, *B. umbrinellus* Pat. & Baker 1918 and *B. umbrinus* Pers. 1825, were subsequently added to the section (Zang 1999).

There are different treatments of the taxonomic position of *B. nigricans*. By studying the type material, Singer (1947) placed this species in *Tylopilus* P. Karst 1881 as *T. nigricans* (Pat. & Baker) Singer and pointed out that it was very close to *T. alboater* (Schwein.) Murrill 1909, but different only in smaller size of carpophores, the shape of cystidia and spores. After re-examination of the type of *B. alboater* Schwein. 1822, Corner (1972) regarded *B. nigricans* as a synonym of *B. alboater* on the basis of the description of and notes on the type of the former by Singer. We agree with Corner (1972) that *B. nigricans* and *B. alboater* (= *T. alboater*) are conspecific.

Zang et al. (1993) noticed that *B. atratus* (as *Boletus nigricans* M. Zang et al.) was similar to *B. nigerrimus*. According to Corner (1972), the latter has a sooty pileus often with a purple tint, and a fuliginous olive or greenish olive to olivaceous buff stipe with wholly black scurfy pruinose reticulation and white mycelium at the base. In contrast, *B. atratus* has sooty black to black pileus without purple tint, sooty to black stipe with fine reticulation at least on the upper half and black mycelium at the base.

In fact, *B. atratus* is closer to *T. alboater*, which has velvety black to dark grayish brown pileus and concolorous stipe sometimes with reticulation at the apex. The black hymenophore, black and unchanging context of the pileus when bruised, and a stipe with reticulation sometimes on the lower half and with black mycelium at the base in *B. atratus* (see Zang et al. 1993) make it distinguishable from *T. alboater*, which has a pinkish or flesh colored hymenophore, pallid white or yellowish white context eventually blackening, and a stipe without reticulation on the lower half and with white mycelium at the base in the latter. However, the black hymenophore and the black context seldom appear in the genus *Boletus*. Whether *B. atratus* is a member of *Boletus* or *Tylopilus* may be determined when the two genera are better circumscribed.

Suillus cavipoides (Z. S. Bi & G. Y. Zheng) Q. B. Wang & Y. J. Yao, **comb. nov.**
≡*Boletinus cavipoides* Z. S. Bi & G. Y. Zheng in *Acta Bot. Yunnan.* 4: 58 (1982).

Pileus 1.8–5.5 cm in diam., subconvex to plane, tomentose, very viscid when moist, pale yellow to pale yellow brown. *Context* 3–8 mm thick, pale yellow, concolorous with the pileus, unchanging when bruised, odor and taste mild. *Tubes* 3 mm long, yellow, unchanging when bruised, decurrent around the stipe; pores 1–2 pores/mm, nearly circular to sub-multiangular, compound, radically arranged, yellow, unchanging when bruised. *Stipe* 2.2–4.5 cm long, 0.4–0.7 cm thick, cylindric, pale

brown or concolorous with the pileus, hollow when mature. *Basidiospores* 5.0–9.0 x 3.0–4.0 μm, mostly 7.0–8.0 x 3.0 μm, narrowly elliptical, smooth, thin-walled, olive brown. *Basidia* 13.0–27 x 5.0–7.0 μm, clavate, 2- and 4-spored. *Pleurocystidia* 37–48 x 5.5–8.0 μm, clavate. *Tube trama Boletus*-type, divergent from a central strand. *Clamp connexions* absent.

Specimens examined: **China: Guangdong Province**, Dinghu, Caotang, 19 April 1981, Z. S. Bi, HMIGD 4819; Dinghu Mountains, in mixed woods, 12 May 1981, Z. S. Bi et al., HMIGD 4901; Dinghu Mountains, 9 May 1982, Y. Z. Wang & J. Q. Liang, HMIGD 5769.
Habit and Habitat: Gregarious in mixed forest.
Distribution: China, Guangdong Province (type locality).

The type material of this species cannot be located in HMIGD where it was housed. The above description is based on other specimens (HMIGD 4901, 5769) determined by Z. S. Bi. Re-examination of these specimens shows spores of 5.0–9.0 x 3.0–4.0 μm, mostly 7.0–8.0 x 3.0 μm, basically in accord with those by Bi et al. (1982, 7–10.5 x 3–3.5μm). This is an easily recognized species and is close to *S. cavipes* (Opat.) A. H. Sm. & Thiers 1964, which has a dull yellow to dark reddish brown pileus (Smith & Thiers 1964, 1971). However, the viscid and tomentose pileus distinguishes *Suillus cavipoides* from *S. cavipes*. The absence of clamp connexions in tube trama hyphae and the viscid pileus strongly suggest it to be a member of *Suillus* S. F. Gray 1821.

Boletinus and *Psiloboletinus* Singer 1945 have traditionally been classified with *Suillus* (e.g. Singer 1945, 1975), largely on the boletinoid hymenophore and the elongate spores (Pegler & Young 1981). Characters used to differentiate *Boletinus* from *Suillus* are the presence of clamp connexions, a sterile stipe surface without granular dots, the absence of a viscid or glutinous layer on the pileus and fasciculate encrusted pleurocystidia (Kretzer et al. 1996, Singer 1975, 1986). However, these characters are not consistent (Singer 1962). It is well known that species of *Suillus sensu stricto* are also able to develop clamp connexions in mycelial cultures (Hübsch 1961, Pantidou & Groves 1966). Smith & Thiers (1964) reduced *Boletinus* to a section of the genus *Suillus*. Singer (1975, 1986) restricted the genus to three species (*B. asiaticus* Singer 1938, *B. cavipes* (Opat.) Kalchbr. 1867 and *B. paluster* (Peck) Peck 1889), which are characterized by the presence of clamp connexions in their fruitbodies (Kretzer et al. 1996). Kretzer et al. (1996) proved neither *Boletinus* as defined by Singer (1975, 1986) nor *Fuscoboletinus* Pomerleau & A. H. Sm. 1962 to be a monophyletic group. Molecular phylogenetic analysis (Kretzer et al. 1996) based on the internal transcribed spacer sequences (ITS) strongly supports the concept of a genus *Suillus sensu lato*, which comprises both *Boletinus* and *Fuscoboletinus*, as introduced by Bruns & Palmer (1989).

Suillus kunmingensis (W. F. Chiu) Q. B. Wang & Y. J. Yao, **comb. nov.**
≡*Boletinus kunmingensis* W. F. Chiu in *Mycologia* 40: 199 (1948).

Pileus 3.0–5.0 cm in diam., hemispherical then becoming broad convex to plane, flesh, ochre, cinnamon-brown or orange-cinnamon, very viscid when moist, glabrous,

margin wavy. *Context* pale yellow, deeper at the center, unchanging when bruised. *Tubes* 3–4 mm long, pale peach yellow to buff yellow, unchanging when bruised; pores about 8–10 pores/cm, multi-angular, radically arranged, pale yellow to brown, unchanging when bruised, short decurrent down to the stipe. *Stipe* cylindric or tapering downward, 3.0–4.0 cm long, 5–7 mm thick, pale yellow, dotted with dark-colored granules overall, solid; context yellowish, paler than that of the cap. *Basidiospores* 9.0–12.0 x 4.0–5.0 μm (mostly 10.0–11.0 x 5.0 μm), narrowly ellipsoid, smooth, thin-walled, pale olive brown to pale brown. *Basidia* 24–32 x 6.0–10.0 μm, clavate, 2- and 4-spored. *Pleurocystidia* 36–53 x 6.0–11.0 μm, clavate. *Tube trama* poorly rehydrating. *Clamp connexion* absent.

Specimen examined: **China: Yunnan Province**, Kunming, Tiefeng'an, 12 Nov. 1930, W. F. Chiu, HMAS 3695 (Tsinghua 7695, HOLOTYPE).
Habit and Habitat: Solitary on the ground in a mixed forest (Yuan & Sun 1995).
Distribution: China, Sichuan (Yuan & Sun 1995) and Yunnan (type locality) Provinces.

The description of microscopic characters is based on examination of the holotype. Chiu (1948, 1957) did not measure the sizes of basidia and pleurocystidia, but provided a good description of the macro-features which are followed here.

Suillus kunmingensis is close to *S. pinetorum* (W. F. Chiu) H. Engel & Klofac 1996 and *S. punctatipes* (Snell & E. A. Dick) Snell & E. A. Dick 1961, which all have a brown or cinnamon brown and viscid pileus, and a stipe glandular-dotted at the upper half (see below and also Snell & Dick 1941, Bessette et al. 2000). However, the spores of *S. kunmingensis* (9.0–12.0 x 4.0–5.0 μm) are larger than those of the latter two species, 7.0–9.0 (–10.0) x 3.0–4.0 μm in *S. pinetorum* and 7–10 x 3–3.5μm in *S. punctatipes* (Snell & Dick 1941, 1961). Further, the stipe of *S. kunmingensis* has an entirely granular stipe, compared with granular dots on the upper part of the stipe in both *S. pinetorum* and *S. punctatipes*. As discussed above, species of *Boletinus* can not be separated from that of *Suillus* (Kretzer et al. 1996). The very viscid pileus, the granular stipe and the absence of clamp connexion in tube trama hyphae also show *Boletinus kunmingensis* to be a species within the genus of *Suillus*.

Suillus pinetorum (W. F. Chiu) H. Engel & Klofac in Engel et al., *Schmier- und Filzrohrlinge s.l. in Europa. Die Gattungen* Boletellus, Boletinus, Phylloporus, Suillus, Xerocomus: 12 (1996).
≡*Boletinus punctatipes* Snell & E. A. Dick var. *pinetorum* W. F. Chiu in *Mycologia* 40: 200 (1948).
≡*Boletinus pinetorm* (W. F. Chiu) Teng, *Fungi of China*: 346 (1962).
≡*Suillus pinetorum* (W. F. Chiu) T. H. Li in Z. S. Bi et al., *Prelim. Agaric Flora Hainan Prov.*: 278 (1997), superfluous comb.

Pileus 4.5–5.0 cm in diam., hemispherical, becoming subconvex to plane, glabrous, very viscid when moist, cinnamon brown to brown, often with a red tone. *Context* 1.0–1.5 cm thick at the stipe, pale red orange, unchanging when bruised, taste and odor mild. *Tubes* 0.5 cm long, dark yellow-brown, unchanging when bruised, decurrent down the stipe; pores 5–8 pores/cm, multi-angular, compound, radiately

arranged, concolorous with tubes, unchanging when bruised. *Stipe* cylindric, 2.0–3.0 cm long, 0.5–1.5 cm thick, concolorous with the cap or paler, with brown dots on the upper part, reticulation absent, solid. *Basidiospores* 7.0–9.0 (–10.0) x 3.0–4.0 μm, mostly 8.0 x 3.0 μm, narrowly ellipsoid, smooth, thin-walled, pale olive brown to pale brown. *Basidia* 21–32 x 5.0–7.0 μm, clavate, sterigmata 3.0–8.0 μm long, 2-, 3- and 4-spored. *Pleurocystidia* 43–53 x 5.0–8.0 μm, long clavate. *Tube trama* subparallel, somewhat divergent toward the subhymenium. *Clamp connexions* absent.

Specimens examined: **China: Yunnan Province**, Nanhua Market, 31 Aug. 2002, Q. B. Wang No. 80, HMAS 83652; and 12 Aug. 2003, Q. B. Wang No. 215, HMAS 85234; Songming, Baiyi Wild Mushroom Market, 25 Aug. 2002, F. Q. Yu No. 930, HKAS 41746; Kunming, 11 July 1941, W. F. Chiu, HMAS 3717 (Tsinghua 7717, HOLOTYPE). **Fujian Province**, Shaowu, Baoji, in mixed forest, 8 Sept. 1957, B. N. Jiang No. 3, HMAS 23853; Nanjing, Nankeng, Daji, on the ground, 14 June 1958, S. C. Teng No. 5869, HMAS 22797. **Hunan Province**, Longshan, on the ground, 30 Oct. 1958, Q. T. Chen & L. S. Liang, No. 1368, HMAS 26073.
Habit and Habitat: Scattered to solitary in the forest of *Keteleeria* spp. and *Pinus* spp.
Distribution: China, Fujian, Hunan and Yunnan Provinces.

The newly obtained specimens cited above from Yunnan from 2002–2003, match the description by Chiu (1948, 1957), except for the size of the basidia and pleurocystidia not mentioned originally. The type and the early collections also exhibit abundant pleurocystidia.

Accepting only *Boletus* and *Boletinus* in the family *Boletaceae*, Chiu (1948, 1957) described the above taxon as a variety under *Boletinus punctatipes*, noting that it differs from the species in the pale red cinnamon to pale red brown stipe compared with the white stipe with brown tints in the latter (Snell & Dick 1941). Teng (1963) elevated it to the species level on the basis of the difference mentioned above. Because of the viscid pileus and the absence of clamp connexions in tube trama hyphae, both *B. pinetorum* and *B. punctatipes* have been transferred to *Suillus* as *S. punctatipes* and *S. pinetorum* respectively. Phylogenetic analysis based on ITS sequenced performed in this laboratory (data unpublished) also supports this result.

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Pluteus thomsonii (Pluteaceae): A northern agaric found in South America

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Abstract—*Pluteus thomsonii (Pluteaceae)*, a widespread agaric in the Northern Hemisphere (Europe, North America and Asia), is reported for the first time in South America, in southern Brazil. Full description and line drawings of this species are presented.

Key words—*Agaricales*, *Pluteus* Sect. *Celluloderma*, Brazilian mycobiota

Introduction

The genus *Pluteus* Fr. *(Pluteaceae, Agaricales)* is macroscopically characterized by basidiomata with free lamellae, a pinkish spore print and the absence of both annulus and volva; microscopically, it is defined by the presence of an inverse hymenophoral trama, stramineous spores, and the presence of hymenial cystidia (metuloidal or not); furthermore, it has a lignicolous habit (Singer 1986). Currently the genus comprises 300 species (Kirk et al. 2001) but only 35 *Pluteus* taxa are recognized from Brazil (Wartchow et al., ined.), most of them recorded by Singer (1953, 1956, 1958, 1961, 1989), and recently by Stijve & de Meijer (1993) and Pegler (1997). In several papers by Rick, summarized in Rick (1961), he reported and described several *Pluteus* species, but many were revised by Singer (1953, 1956) and synonymized with other taxa or considered *nomen dubium* because the lack of type material. This paper reports a *Pluteus* species previously unrecorded from Brazil. Our goal is to obtain a better understanding of the diversity within this genus in the state of Rio Grande do Sul, in southern Brazil.

Materials and methods

The species was collected at the "Morro do Elefante", in Santa Maria, state of Rio Grande do Sul, in southern Brazil. The locality of "Morro do Elefante" comprises subtropical

rain forests, with the following species being most abundant: *Allophylus edulis* (St. Hill.) Radlk., *Cupania vernalis* Camb., *Enterolobium contortisiliquum* (Vell.) Morong, *Nectandra megapotamica* (Spreng.) Mez, and *Trichilia claussenii* C.DC. (Machado & Longhi 1990). The methodology of Singer (1986) was followed, and the microscopic line drawings were prepared using a camera lucida. Generic and infra-generic concepts follow Singer (1986). Material is preserved at the Herbarium of the Department of Biology of the Universidade Federal de Santa Maria (SMDB).

Taxonomic Description

Pluteus thomsonii (Berk. & Broome) Dennis, Trans. Br. Mycol. Soc. 31: 206, 1948. FIGURES 1-6

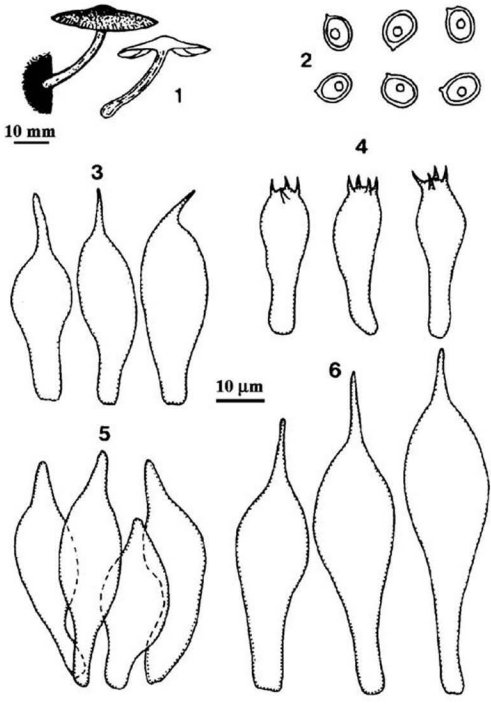
- = *Agaricus thomsonii* Berk. & Broome, Ann. Mag. Nat. Hist., Ser. IV, 17: 131, 1876.
  - = *P. cinereus* Quél., Ann. Sci. Nat. Bord. (Suppl.) 14: 3, 1884.
- = *Entoloma thomsonii* (Berk. & Broome) Sacc., Syll. Fung. 5: 693, 1887.
  - = *P. reisneri* Velen., Ceské Houby: 610, 1921.
  - = *P. pilatii* Velen., Mykologia 6: 25, 1929 (as *P. pilati*).
  - = *P. cinereus* Quél. f. *evenosus* Kühner, Bull. Soc. Mycol. Fr. 72: 181, 1956.
  - = *P. cinereus* Quél. f. *typicus* Kühner, Bull. Soc. Mycol. Fr. 72: 199, 1956.
- = *P. thomsonii* f. *evenosus* (Kühner) Wuilb., Misc. Mycol. 15: 16, 1986.

Pileus 36 mm in diam., umbonate to applanate, blackish-grey at the umbo, castaneous-whitish toward the margin, surface radially rugose to slightly venulose on disc, but without a strong venation. **Lamellae** free, pink-colored, abundant. **Stipe** 32 x 3 mm, central, cylindrical, with a slightly expanded base, whitish, longitudinally striate, fibrous, without any kind of velar remnants. **Context** thin, membranous, whitish. **Spore print** pink. **Spores** 6-8 x 4-6.5 □m, subglobose to short ellipsoid in shape, stramineous under light microscope (in KOH), smooth and thick-walled, apiculus evident. **Basidia** 24-33 x 7.5-10.5 □m, thin-walled, clavate, with four sterigmata. **Pleurocystidia** 41.5-76 x 12-23 □m, fusiform, with a long and acute apex, hyaline, abundant. **Cheilocystidia** 32-46 x 9-17.5 □m, similar to the pleurocystidia in shape (fusiform) but smaller in size. **Pileipellis** hymeniform, with cells 29.5-57 x 8-18.5 □m, comprising vesiculose to clavate elements mixed with fusiform to ventricose cystidioid hyphae with an acute apex; brownish pigments uniformly distributed on the cell wall. **Caulocystidia** not seen. **Clamp connections** not observed. **Hymenophoral trama** inverse, with hyaline, thin-walled hyphae.

Habit and habitat: solitary, on decayed wood of unidentified angiosperm, in a subtropical rain forest.

> **Material examined – BRAZIL. Rio Grande do Sul: Santa Maria**, Camobi, Morro do Elefante, 15.X.2001. *Wartchow* (SMDB 9194).

Distribution: Widespread in Europe (Breitenbach & Kränzlin 1995; Citérin & Eyssartier 1998; Consiglio 2000; Dennis 1948; Enderle 1986; Kühner & Romagnesi 1956; Moser 1978; Orton 1960, 1986; Vellinga 1990; Vellinga & Schreurs 1985; Wuilbaut 1986) and North America (Homola 1972; Rodríguez & Guzmán-Dávalos 1999, 2000, 2001), with sporadic records from Asia (Imazeki et al. 1988) and North Africa (Vellinga 1990).



Figures 1-6. *Pluteus thomsonii*: 1) Basidiome and section. 2) Spores. 3) Cheilocystidia. 4) Basidia. 5) Pileipellis elements. 6) Pleurocystidia.

Remarks: *Pluteus thomsonii* belongs to Section *Celluloderma* Fayod, Subsection *Mixtini* Singer, due to the presence of dimorphic (fusiform and vesiculose) elements on the hymeniform pileipellis (Singer 1986). This species, in its typical form, is characterized by the strong venation of the pileus surface, the size of the spores, and especially by the shape of the cystidia (Consiglio 2000).

Some specimens without a distinctly venose pileus have been called *P. thomsonii* f. *evenosus* (Kühner) Wuilb., as indicated by Wuilbaut (1986), Citérin & Eyssartier (1998) and Consiglio (2000). Our collection agrees very well with the descriptions given by these authors because of the absence of conspicuous veins on the pileus surface. Nevertheless, Orton (1986) established that the presence or absence of veins on the pileus lacks taxonomic value because young basidiomata present a reduced or absent venation, which is very conspicuous in the mature ones. Thus, the above-cited forms are currently considered synonyms by these and other modern authors (e.g., Vellinga 1990; Rodríguez & Guzmán-Dávalos 1999), and this taxonomic treatment is herein applied.

This species is somewhat similar to other taxa of Section *Celluloderma*, such as *P. insidiosus* Vellinga & Schreurs, a European species which has only vesiculose elements on pileipellis (Subsection *Eucellulodermini* Singer) instead of *P. thomsonii* which has dimorphic elements on it (Rodríguez & Guzmán-Dávalos 1999).

Singer (1953, 1956, 1958, 1961, 1969, 1989) studied many *Pluteus* collections from South America, including Brazil, but does not refer to *P. thomsonii* or its synonyms. Other authors as like Courtecuisse (1991), Dennis (1953, 1970), Horak (1964, 1979), Pegler (1997), Singer & Digilio (1951), and Stijve & de Meijer (1993), in their studies of *Pluteus* in South America, also do not cited this species. This is the first record of *P. thomsonii* from South America and Brazil.

Acknowledgements

The authors thank all mycologists who have kindly sent us literature on *Pluteus*. We thank to Dr. Iuri Baseia (Universidade Federal de Pernambuco, Brazil) for suggestions. Dr. Else Vellinga (University of California, USA) and Dr. Laura Guzmán-Dávalos (University of Guadalajara, Mexico) are acknowledged for critically reviewing the manuscript and improvements of the English.

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The *Strophariodeae (Strophariaceae, Agaricales)* from Santa Maria, Rio Grande do Sul, Brazil

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Abstract–Collections of *Hypholoma subviride, Melanotus proteus, Psilocybe caeruleoannulata, P. coprophila, P. cubensis, P. moellerii, P. pegleriana, Stropharia coronilla, S. rugosoannulata,* and *S. semiglobata,* all placed in subfamily *Strophariodeae (Strophariaceae, Agaricales)* are discussed and illustrated from southern Brazil. *Melanotus proteus, P. moellerii* and *P. pegleriana* are recorded for the first time from the country, and *S. rugosoannulata* and *H. subviride* collections represent new records for the state of Rio Grande do Sul.

Key words – Basidiomycetes, tropical agarics, Brazilian mycobiota

Introduction

Little is known about members of the family Strophariaceae Singer & A.H. Sm. in Brazil. Guzmán (1978) and Guzmán et al. (1984), who described new *Psilocybe* taxa, and Pegler (1997) have contributed the most significant modern records of these fungi in Brazil. Stijve & de Meijer (1993), who studied psychoactive agarics from the state of Paraná, recorded several *Psilocybe* and some *Stropharia* species. Guzmán et al. (2000) included 18 neurotropic species of *Psilocybe* from Brazil on their checklist of world psychotropic fungi, and regional checklists by Bononi et al. (1984), Sotão et al. (1991), and Vinha (1988) cited a few species of *Strophariaceae (Strophariodeae)* from some Brazilian states.

Rick (1907, 1930, 1939, 1961), who provided the earliest and possibly only records of Strophariaceae from Rio Grande do Sul, cited 9 *Stropharia*,

10 *Hypholoma*, and 5 *Psilocybe* species, including the new taxa, *S. subcyanescens* Rick and *S. siccipes* P. Karst. var. *lugubris* Rick. Singer (1953) revised several Rick's types and collections, and reported other taxa.

The goal of our current taxonomic survey is to expand what is known about the Strophariaceae from the Rio Grande do Sul state, in southern Brazil. The current paper focuses on the subfamily Stropharioideae (Singer) Singer in the sense of Singer (1986), although several taxonomic concepts about the family Strophariaceae have been recently arisen (e.g., Kühner 1984; Noordeloos 1999; Smith 1979; Watling & Gregory 1987).

Materials and methods

Collections were made from March 2001 to March from within Santa Maria between 53°30'22"- 54°19'32" W, and 29°20'28" - 30°00'16" S, within the Depressão Central of Rio Grande do Sul state. The area encompasses two main vegetation types (Pereira et al. 1989); subtropical forests to the northeastern and meadows to the southwestern. The subtropical forests contain trees belonging to the Fabaceae, Lauraceae, Myrtaceae, Meliaceae, and Sapindaceae (Longhi et al. 2000; Machado & Longhi 1990). The meadows are dominated by species of Poaceae, Fabaceae, Asteraceae, Rubiaceae, and Cyperaceae (Quadros et al. 2003).

Tissues of collected specimens were mounted in 5% KOH and 10 % NH_4OH aqueous solutions. Basidiospore colors in descriptions are indicated under KOH observations. Vouchers of all studied collections are deposited at the Herbarium of the Department of Biology of the Universidade Federal de Santa Maria (SMDB). Exsiccati from PACA (Herbarium Anchieta, UNISINOS) and ICN (Department of Botany, Universidade Federal do Rio Grande do Sul) were also examined.

Results

Ten species representing the genera *Hypholoma* (1), *Melanotus* (1), *Psilocybe* (5), and *Stropharia* (3) were identified.

Key to the species of subfamily *Strophorioideae* of Santa Maria

1a. Chrysocystidia present. Spore print dark vinaceous brown **2**
1b. Chrysocystidia absent. Spore print violaceous black..... **5**

2a. Lignicolous, cespitose. Pileus sulfur yellow, ≤35 mm diam. Annulus absent. Basidiospores 5-9 μm long (genus *Hypholoma*) ..... **1. *H. subviride***
2b. Coprophilous or terrestrial, solitary to gregarious (genus *Stropharia*) **3**

3a. Coprophilous. Basidiospores ≥16 μm long ..... **10. *S. semiglobata***
3b. Terrestrial. Basidiospores ≤16 μm long **4**

4a. In meadows. Pileus ≤ 60 mm diam., yellowish. Basidiospores 6-10 μm long. Cheilocystidia clavate ..... **8. *S. coronilla***
4b. In forests. Pileus ≥ 60 mm diam., vinaceous. Basidiospores 9-14μm long. Cheilocystidia fusoid-ventricose ..... **9. *S. rugosoannulata***

5a. Habit pleurotoid, on coniferous wood. Stipe reduced. Basidiospores 5-7 μm long, subhexagonal in face view (genus *Melanotus*) ..... **2. *M. proteus***
5b. Habit never pleurotoid. Stipe well developed (genus *Psilocybe*) **6**

6a. Annulus absent. Pileus reddish brown, strongly hygrophanous. Basidiospores 10-12 μm. On dung ..... **4. *P. coprophila***
6b. Annulus or velar remnants present on stipe. Pileus yellowish to brownish **7**

7a. Specimen not bluing on stipe **8**
7b. Specimen bluing on stipe **9**

8a. Annulus submembranous. Basidiospores 9-11 μm long ... **7. *P. pegleriana***
8b. Annulus fibrillose, not membranous. Basidiospores 12-14 μm long ..... **6. *P. moellerii***

9. Basidiomata ≤ 50 mm. Basidiospores 9-12 μm long, ovoid in face view ..... **3. *P. caeruleoannulata***
9b. Basidiomata usually ≥ 50 mm. Basidiospores 12-17μm long, subhexagonal in face view ..... **5. *P. cubensis***

Hypholoma (Fr.) P. Kumm.

1. *Hypholoma subviride* (Berk. & M.A. Curtis) Dennis, Kew Bull. 15: 134, 1961. FIGURE 1 A-D

Pileus 8-35 mm diam., conic, becoming plano-convex; surface dry, smooth; color sulfur-yellow, darker on central disc; margin non-striate, brownish, without velar remnants; context thin, fleshy, yellow. **Lamellae** adnate, initially sulfur-yellow colored, becoming vinaceous brown in age. **Stipe** 25-66 x 1.5-4 mm, central, cylindrical, sometimes sinuous, concolorous with the pileus. **Veil** never forming an annulus.

Basidiospores dark vinaceous brown in mass; 5.5-9 x 3-5 μm, ellipsoid both in face and side view, apically truncated by a broad germ pore; smooth, thick-walled, yellowish brown. **Basidia** 16-22.5 x 4.5-7 μm, clavate, tetrasporic. **Pleurocystidia** 19-38.5 x 5.5-9.5 μm, as true chrysocystidia; fusoid, mucronate, with amorphous content. **Cheilocystidia** not observed. **Pileipellis** composed of prostrate, thick walled hyphae. **Hypodermium** subcellular, with inflated hyphae. **Hymenophoral trama** regular. **Clamp connections** present.

Ecology: Lignicolous on decomposed wood, particularly *Eucalyptus*.

Specimens examined: BRAZIL. Rio Grande do Sul: **Santa Maria**, Campus UFSM, 20.VIII.2000 *Cortez* 041/00 (SMDB 9226); Morro do Elefante, 9.V.2001 *Cortez* 024/01 (SMDB 9244), 21.VII.2001 *Wartchow* 007 (SMDB 9190).

Distribution: North America (Smith 1951); Central America (Pegler 1983, 1988); South America (Dennis 1970; Pulido 1983); Africa (Pegler 1977; Reid & Eicker 1999); Europe (Krieglsteiner & Enderle 1986); Asia (Pegler 1986).

Remarks: Previously reported from Brazilian states of Amapá (Sotão et al. 1991) and São Paulo (Bononi et al. 1984; Pegler 1997), this is the first record of *H. subviride* from the Rio Grande do Sul state. Dennis (1970) interpreted this species as a reduced tropical form of the temperate *H. fasciculare* (Huds.: Fr.) P. Kumm., reported from Rio Grande do Sul by Rick (1961). Krieglsteiner & Enderle (1986) considered *H. subviride* as a variety of that species. Smith (1951) separated the two species based on differences in geographic distribution and in some secondary characters (e.g., for *H. fasciculare* a pileus size > 30 mm and convex shape).

Other *Hypholoma* species – Rick (1961) reported *Hypholoma ericaeum* (Pers.: Fr.) Kühner (*Rick 14.554*, PACA, as *Psilocybe ericacea* Pers.) from Santa Maria. We have not examined Rick's collection or gathered any fresh specimens during our study.

Melanotus Pat.

2. *Melanotus proteus* (Kalchbr.) Singer, Lloydia 9: 130, 1946. FIGURE 2 A-E

Pileus 2-12 mm diam., reniform, convex; surface dry to moist, glabrous; pale brown, opaque; margin incurved, non-striate; velar remnants absent; context thin, whitish. **Lamellae** adnate to subdecurrent; pale fuscous brown. **Stipe** 1-2 x –1 mm, lateral, cylindrical, very reduced; brown with abundant white mycelium at the base. **Veil** absent.

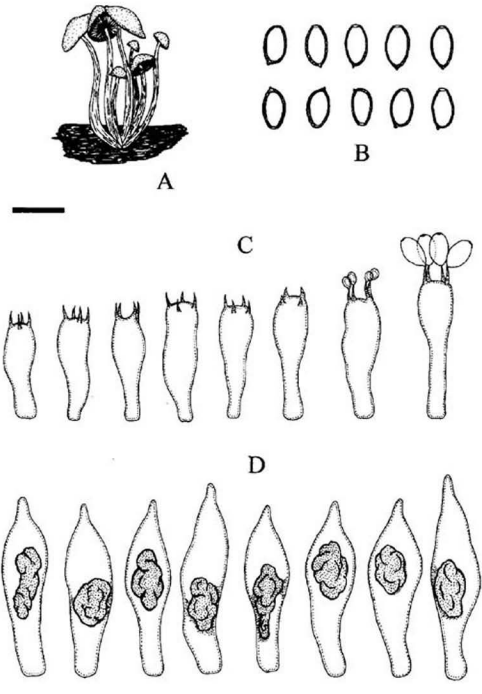

Figure 1. ***Hypholoma subviride*:** A. Basidiomata. B. Basidiospores. C. Basidia. D. Chrysocystidia. Scale bar: A = 20 mm; B-D = 10μm.

Basidiospores violaceous brown as noted on stipe and pileus surfaces; 4.5-7 x 3-5 μm, broad ellipsoid to subhexagonal both in face and side view, apically truncated by a broad germ pore, smooth and thick-walled, brown. **Basidia** 13-21.5 x 5-7 μm, clavate, tetrasporic. **Pleurocystidia** absent. **Cheilocystidia** 14.5-25 x 4-6.5 μm, lageniform to subcapitate, long-necked, hyaline, thin-walled. **Pileipellis** an ixocutis composed by prostrate, clamped, 5-8 μm wide hyphae, walls somewhat thickened and incrusted. **Hymenophoral trama** regular. **Clamp connections** present.

Ecology: Lignicolous on fallen branches of *Araucaria angustifolia* (Bertol.) O. Ktze.

Specimens examined: BRAZIL. Rio Grande do Sul: **Itaara,** Parque Pinhal, 23.II.2002 *Cortez* 008/02 (SMDB).

Distribution: Europe (Bon & Pacaud 2002; Kits Van Waveren 1979; Redfern 1991; Vesterholt 1993; Watling & Gregory 1987); Asia (Pegler 1986); Africa (Horak 1977; Pegler 1977).

Remarks: Mating studies by Sime & Petersen (1999) have suggested that *M. proteus* and *M. textilis* Redhead & Kroeger are both synonyms of *M. horizontalis* (Bull.: Fr.) P.D. Orton. Watling & Gregory (1987) separated *M. proteus* by the coniferous substrate, pale velutinate pileus, and ellipsoid-amygdaliform spores, and their concept is here followed. *Melanotus proteus* is known to be a potential competitor of *Heterobasidium annosum* (Fr.) Bref., a polypore which causes root and heart-rot in coniferous trees in Europe (Redfern 1991), and has been studied as a biological controller of this wood-pathogen (Pratt & Quill 1996). This *Melanotus* is recorded for the first time in Brazil.

Psilocybe (Fr.) P. Kumm.

3. *Psilocybe caeruleoannulata* Singer ex Guzmán, Mycotaxon 7: 235, 1978.

Figure 2 F-I

Pileus 10-50 mm diam., conic to convex, surface subviscid, smooth; brownish with striate margin; context fleshy, pale brownish, probably bluing. **Lamellae** adnexed, blackish brown, rather close. **Stipe** 30-40 x 5 mm, central, cylindrical, flexuous, annulate. **Veil** present as a membranous annulus, bluing.

Basidiospores violaceous brown in mass; 9.5-12 x 6.5-7 μm, subellipsoid in side view and ovoid in frontal view, smooth and thick-walled, germ-pore broad, brownish. **Basidia** 17.5-21 x 5.5-7 μm, clavate, tetrasporic.

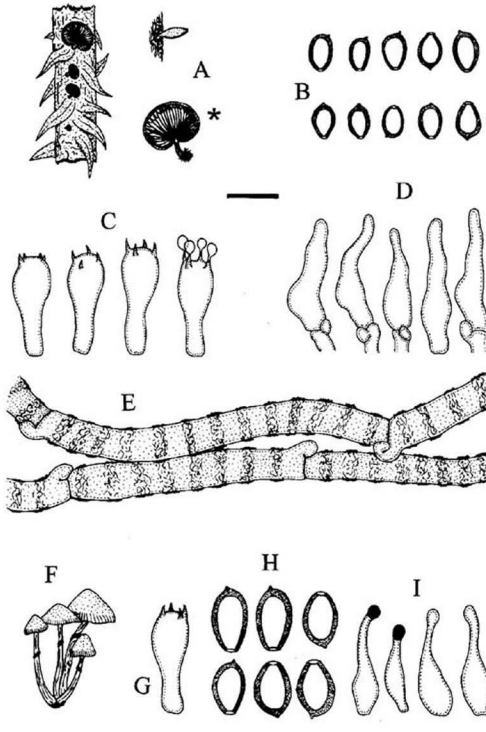

Figure 2. A-E: ***Melanotus proteus*****:** A. Basidiomata. B. Basidiospores. C. Basidia. D. Cheilocystidia. E. Pileipellis. **F-I:** ***Psilocybe caeruleoannulata*****:** F. Basidiomata. G. Basidium. H. Basidiospores. I. Cheilocystidia. Scale bar: A*= 40 mm; A, F = 20 mm; B-E, G-I = 10 μm.

Pleurocystidia absent. **Cheilocystidia** 15.5-21.5 x 3-6.5 μm, ventricose, with a long neck, subcapitate, thin-walled, sometimes with an oleous incrustation at the apex. **Pileipellis** an ixocutis, with hyaline to slightly incrusted hyphae. **Hymenophoral trama** regular. **Clamp connections** present.

Ecology: Lignicolous, cespitose on rotting woodchips of *Araucaria angustifolia* or *Podocarpus*.

Specimens **examined:** BRAZIL. Rio Grande do Sul: **Santa Maria,** 1936, *Rick* 15.234 (PACA 9370, holotype of *Stropharia siccipes* var. *lugubris*), *Rick* 15.237 (PACA 9371), *Rick* 15.238 (PACA 9372).

Distribution: South America (southern Brazil and Uruguay - Guzmán 1983).

Remarks: No fresh specimens of this species were collected in our study, and the collections studied are not in good condition. The macroscopic description above is based on observations by Rick (1961), who first described the species as *Stropharia siccipes* P. Karst. var. *lugubris* Rick (Rick 1939). Guzmán (1978) studied the type and considered it as a synonym of *P. caeruleoannulata*. This is a bluing species, probably with psychotropic effects, as supported by the chemical analysis performed by Stijve & de Meijer (1993). It belongs to the Section *Stuntzae* Guzmán and is most closely related to *P. uruguayensis* Singer ex Guzmán and *P. stuntzii* Guzmán & Ott, two closest species. Stijve & de Meijer (1993) also suggested that *P. caeruleoannulata* and *P. uruguayensis* represent a single South American endemic species. *Psilocybe caeruleoannulata* has been also reported from the states of Paraná (Stijve & de Meijer 1993) and São Paulo (Pegler 1997).

4. *Psilocybe coprophila* (Bull.: Fr.) P. Kumm., Führ. Pilzk.: 71, 1871.

Figure 3 A-E

Pileus 4-14 mm diam., convex to campanulate or applanate-umbonate; surface subviscid, glabrous, smooth; margin striate, frequently with appendiculate whitish velar remnants; color dark reddish brown to orange brown, finally pale yellowish, strongly hygrophanous; context thin, whitish. **Lamellae** adnate to subdecurrent, brownish to violaceous in age, with a whitish edge. **Stipe** (6) 11-36 x 1-2.5 mm, central, cylindrical, hollow, basal surface with little squamules; pale to dark brown, never bluing. **Veil** fugacious, present as fibrillose remnants on pileus margin, not forming an annulus.

Basidiospores violaceous brown to violaceous black in mass; 11-14.5 x 7-9.5 μm, subellipsoid in side view and subhexagonal in frontal view; smooth and thick-walled, with a well developed germ pore, yellowish brown.

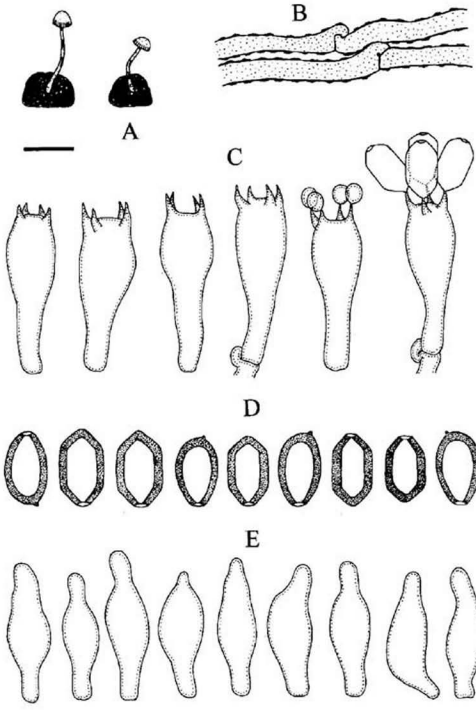

Figure 3. ***Psilocybe coprophila*****:** A. Basidiomata. B. Hyphae of the hypodermium. C. Basidia. D. Basidiospores. E. Cheilocystidia. Scale bar: A = 20 mm; B-E = 10 □m.

Basidia 25-36 x 9-13.5 μm, hyaline, subclavate, tetrasporic. **Pleurocystidia** absent or if present as like the cheilocystidia. **Cheilocystidia** 19-37 x 6.5-9.5 μm, lageniform, with a short to long neck, hyaline, thin-walled, forming a sterile zone in the gill margin. **Pileipellis** an ixocutis composed by prostrate hyphae. **Hypodermium** formed by hyphae encrusted with golden yellowish pigment. **Hymenophoral trama** regular. **Clamp connections** present.

Ecology: Coprophilous, gregarious on cow and horse dung, in meadows.

Specimens examined: BRAZIL. Rio Grande do Sul: **Santa Maria,** UFSM Expositions Park, 7.XI.2000 *Cortez* 051/00 (SMDB 9227); 21.VI.2001 *Cortez* 036/01 (SMDB); **Nova Petrópolis,** V.1992 *RT Guerrero* (ICN 95509, as *Stropharia semiglobata*).

Distribution: Worldwide (Guzmán 1983).

Remarks: *Psilocybe coprophila* is a very common, widespread and non-hallucinogenic species (Guzmán 1983; Stijve & de Meijer 1993). Basidiospore morphology separates it from the macroscopically similar *P. subcoprophila* (Britz.) Sacc. (unknown in Brazil), which is characterized by larger (15-19 μm long) ellipsoid in both side and frontal view (Noordeloos, 1998). In Brazil, *P. coprophila* was previously recorded from the states of São Paulo (Guzmán 1983), Paraná (Stijve & de Meijer 1993), and Rio Grande do Sul (Rick 1961).

5. *Psilocybe cubensis* (Earle) Singer, Sydowia 2: 37, 1948. FIGURE 4 A-E

Pileus 26-85 mm diam., conic to campanulate, becoming convex or applanate but sub-umbonate with the age; surface subviscid, smooth or with small whitish or yellowish scales; color orange brown on the center and pale yellow toward the margin, bluing with the age; margin slightly striate, sometimes with membranous velar remnants appendiculate; context fleshy, whitish, but becoming blue when cut. **Lamellae** adnexed to adnate, grayish to violaceous brown with the maturity of the spores; edges whitish. **Stipe** 47-112 x 5-14 mm, central, cylindrical to sub-bulbous; white to cream color, changing to blue when touched or cut, surface striate near the pileus and with white to bluish squamules bellow the annulus. **Veil** forming a membranous, well developed white annulus.

Basidiospores deep violaceous brown in mass; 12-17.5 x 7-11 μm, subellipsoid in side view and subhexagonal in face view; smooth and thick-walled, truncated, with a wide germ-pore; yellowish brown. **Basidia** 21.5-27 x 8-13 μm, tetrasporic, subclavate to ventricose, with a medial constriction.

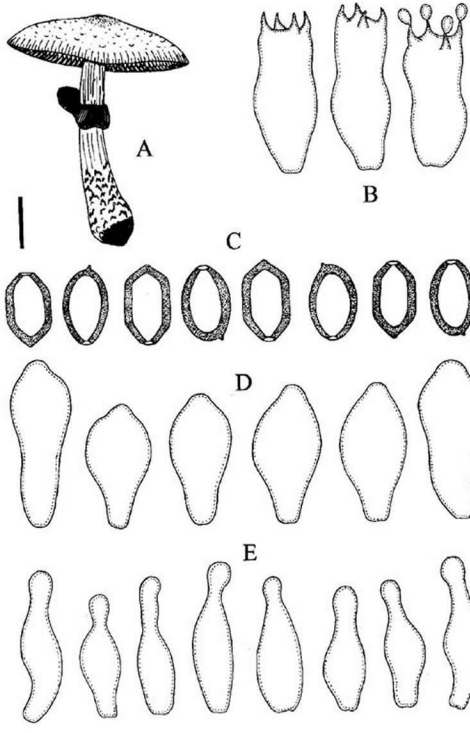

Figure 4. ***Psilocybe cubensis*:** A. Basidiome. B. Basidia. C. Basidiospores. D. Pleurocystidia. E. Cheilocystidia. Scale bar: A = 20 mm; B-E = 10 μm.

Pleurocystidia 21.5-32 x 7-13μm, utriform to subpiriform, hyaline, rare. **Cheilocystidia** 20-32 x 5.5-10.5 μm, lageniform to subcapitate, hyaline, forming a sterile band on the gill edge. **Pileipellis** an ixocutis formed by radially arranged hyphae, gelatinized. **Hymenophoral trama** regular. **Clamp connections** present.

Ecology: Coprophilous, solitary to gregarious on horse and cow dung, in meadows.

Specimens examined: BRAZIL. Rio Grande do Sul: **Santa Maria,** 1936 *Rick* 15227 (PACA 9376, neotype of *Stropharia subcyanescens* Rick); V.1981 *Flavio* (ICN 6676); Arroio Grande District, 5.II.2001 *Cortez* 003/01 (SMDB 9233); UFSM Expositions Park, 8.XI.2000 *Cortez* 052/00 (SMDB 9228), 10.IV.2001 *Cortez* 015/01 (SMDB 9240), 10.X.2001 *Cortez* 046/01 (SMDB 9259). **Restinga Seca,** 11.V.2001 *Cortez* 029/01 (SMDB 9249). **Vera Cruz,** Linha Um, 1.XII.2001 *Cortez & J Putzke* (SMDB 9265). **Guaíba,** Centro Agronômico, 10.IV.1972 *MH Homrich* (ICN 6351).

Distribution: Pantropical (Guzmán 1983).

Remarks: *Psilocybe cubensis*, one of the most common hallucinogenic mushrooms, is widespread in the tropics (Guzmán 1983). *Psilocybe subcubensis* Guzmán, another species in Section *Cubensae* is very close, but has shorter spores (11-13 μm long – Guzmán 1978). Since the two species are recorded from the same habitats, the occurrence of *P. subcubensis* in Brazil should be suspected. *Psilocybe cubensis* is known from the states of Espírito Santo (Vinha 1988), São Paulo (Guzmán 1978; Pegler 1997), Paraná (Stijve & de Meijer 1993), and Rio Grande do Sul (Guerrero & Homrich 1983, 1999; Rick 1930, 1939, 1961 – as *Stropharia subcyanescens* Rick; Singer 1953). *Agaricus rhytopilus* Mont., described from an undetermined locality in Brazil (possibly central western), corresponds to *P. cubensis* according to Pegler (1989), who studied only the Montagne's illustrations and based his identification on the habit, annulus, and habitat on horse dung.

6. *Psilocybe moellerii* Guzmán, Mycotaxon 7: 245, 1978. FIGURE 5 A-D

Pileus 10 mm diam., convex to hemispheric, surface apparently dry, but possibly subviscid when moist, smooth, color brownish to ochraceous; margin striate, non hygrophanous; context thin, whitish. **Lamellae** adnexed to slightly subdecurrent, violaceous grey, with a whitish edge. **Stipe** 35 x 3 mm, central, cylindrical to slightly expanded at the base, pale brownish; surface striate above the annular zone, but somewhat smooth toward the base. **Veil** forming an annular zone in the upper quarter of the stipe, violaceous due the deposition of the spores.

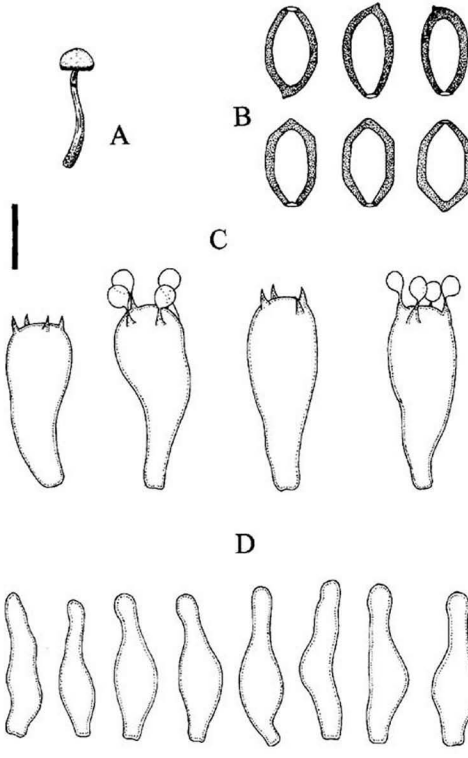

Figure 3. ***Psuocype moeuera*:** A. Basidiome. B. Basidiospores. C. Basidia. D. Cheilocystidia. Scale bar: A = 20 mm; B-E = 10 μm.

Basidiospores violaceous in mass, as seen on stipe surface; 10.5-15 x 7-9.5 μm, slightly subhexagonal in frontal view and subellipsoid in side view, smooth and thick-walled, truncated due the presence of a conspicuous germ-pore, yellowish brown. **Basidia** 22.5-31 x 10.5-13.5 μm, hyaline, clavate, tetrasporic. **Pleurocystidia** absent. **Cheilocystidia** 16-26.5 x 4-7 μm, ventricose, lageniform or subcapitate, thin-walled, hyaline. **Pileipellis** an ixocutis formed by thin-walled, parallel hyphae. **Hymenophoral trama** regular, with thin-walled hyphae. **Clamp connections** present.

Ecology: Coprophilous, solitary on cow dung, in pastures.

Specimens examined: BRAZIL. Rio Grande do Sul: **Santa Maria,** Camobi, near the Morro do Elefante, 24.XII.2001 *Cortez* 052/01 (SMDB).

Distribution: Temperate to subtropical Europe (Breitenbach & Kränzlin 1995; Noordeloos 1998, 1999; Watling & Gregory 1987), North and South America (Guzmán 1983; Yokoyama 1987).

Remarks: This uncommon and non-bluing representative of *Psilocybe* Section *Merdariae* (Fr.) Singer, occurs in both temperate and subtropical regions of the world (Guzmán 1983). *Psilocybe moellerii* is close to *P. merdaria* (Fr.) Ricken and *P. merdicola* Huijsman; the former has slightly shorter spores (10-14μm long), and the second has larger (12-18 μm long) oblong spores (Guzmán 1983; Noordeloos 1999). The studied specimen was collected unfortunately in dry conditions, and the macroscopic features are limited. It is the first record of this species from Brazil.

7. *Psilocybe pegleriana* Guzmán, Doc. Mycol. 29: 43, 2000. FIGURE 6 A-E

Pileus 4.5-10 mm diam., hemispheric, umbonate; surface moist, glabrous, hygrophanous; color grayish brown at margin and orange brown at the disc; margin striate; context fleshy, pale brownish, unchanging. **Lamellae** adnate to subdecurrent; brown to violaceous brown. **Stipe** 38-42 x 1.5-3 mm, cylindrical, central, slightly expanded at base; softly grayish, longitudinally striate. **Veil** forming a well developed, membranous, white and persistent annulus, but sometimes vestigial; violaceous at the upper surface due to spore deposition.

Basidiospores violaceous black in mass; 9.5-11 x 7-9.5 μm, subhexagonal in frontal view, ellipsoid in side view; smooth and thick-walled, with a conspicuous germ pore. **Basidia** 19-28 x 7-9.5 μm, clavate, tetrasporic. **Pleurocystidia** absent. **Cheilocystidia** 20-27.5 x 4-8 μm, mainly lageniform, hyaline. **Pileipellis** an ixocutis of hyaline, prostrate hyphae. **Hymenophoral trama** regular. **Clamp connections** present.

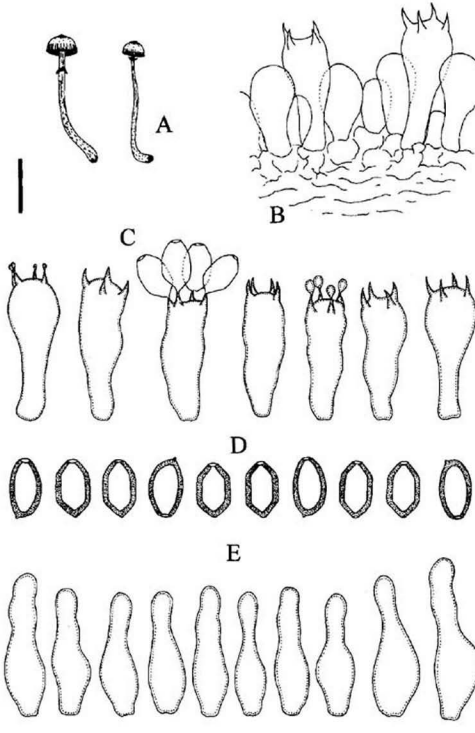

Figure 6. ***Psilocybe pegleriana*:** A. Basidiomata. B. Detail of the hymenium. C. Basidia. D. Basidiospores. E. Cheilocystidia. Scale bar: A = 20 mm; B-E = 10 μm.

Ecology: Coprophilous, gregarious on cow dung, in pastures.

Specimens examined: BRAZIL. Rio Grande do Sul: **Santa Maria,** UFSM Expositions Park, 13.XII.2000 *Cortez* 054/00 (SMDB 9229).

Distribution: North America (Guzmán 2000), South America, Africa (Guzmán 1983, 2000), and Asia (Thomas et al. 2002).

Remarks: *Psilocybe pegleriana* also belongs to Section *Merdariae*. Guzmán (2000) described the species based on collections from Mexico, Venezuela and Africa that he (Guzmán 1983) first considered as *P. pseudobullacea* (Petch) Pegler, following Pegler (1977). However, Pegler (1986) re-described the species after noting the presence of pleurocystidia in the type of *P. pseudobullacea* from Sri Lanka. Our collection of *P. pegleriana* represents a new addition to Brazilian mycobiota.

Stropharia (Fr.) Quél.

8. *Stropharia coronilla* (Bull.: Fr.) Quél., Mem. Soc. d'Emul. Mont. Ser. II, 5: 237, 1872. Figure 7 A-E

Pileus 20-55 mm diam., hemispheric to plano-convex; surface moist, glabrous; pale yellow in color, margin slightly involute in young basidiomata; context fleshy, whitish, unchanging. **Lamellae** adnexed to adnate, grayish, becoming violaceous to vinaceous brown with whitish edges. **Stipe** 22-47 x 4-8 mm, central, cylindrical to sub-bulbous, solid; somewhat longitudinally striate. **Veil** well developed, forming a membranous to thick and fleshy annulus; white but becoming vinaceous due the deposition of the spores, grooved on upper surface.

Basidiospores dark vinaceous brown in mass; 6.5-10.5 x 5-6.5 μm, ellipsoid both in frontal and side view; smooth and thick-walled, with an absent or inconspicuous germ-pore, brown. **Basidia** 20-29 x 6.5-10 μm, clavate, tetrasporic or occasionally bisporic. **Pleurocystidia** 28-48 x 7-13 μm, clavate to mucronate, as true chrysocystidia, with amorphous contents. **Cheilocystidia** 21.5-56 x 8-13 μm, chrysocistidiod or not, mainly clavate or fusiform, similar to pleurocystidia. **Pileipellis** an ixocutis, composed by filamentous, prostrate, somewhat gelatinized hyphae. **Hymenophoral trama** regular. **Clamp connections** present.

Ecology: Terrestrial, solitary or gregarious in soil of gardens and fields between grasses.

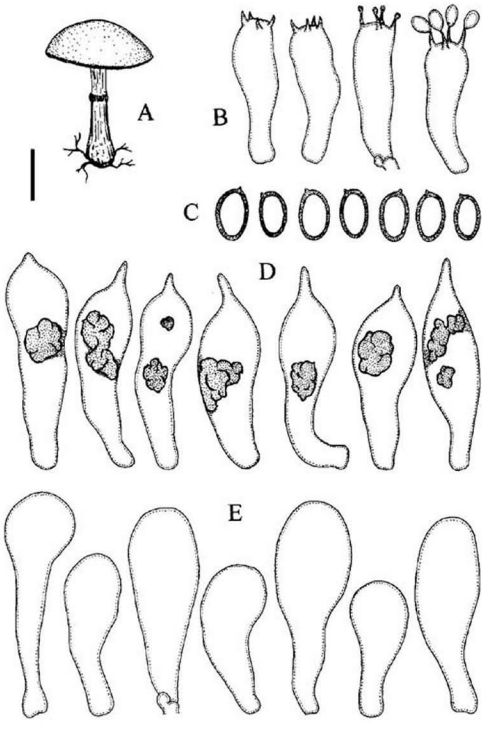

Figure 7. ***Stropharia coronilla*****:** A. Basidiome. B. Basidia. C. Basidiospores. D. Chrysocystidia. E. Cheilocystidia. Scale bar: Fig. A = 20 mm; Figs. B-E = 10 μm.

Material examined: BRAZIL. Rio Grande do Sul: **Santa Maria,** Vila Caramelo, 30.IV.2001 *Cortez* 020/01 (SMDB 9242); Passo dos Ferreiros, 2.V.2001 *Cortez* 021/01 (SMDB 9243).

Distribution: South America (Singer & Digilio 1951; Wright & Albertó 2002), North America (Lincoff 1988; Murrill 1922), Europe (Breitenbach & Kränzlin 1995; Moser 1978; Noordeloos 1999; Watling & Gregory 1987), Africa (Pegler, 1977).

Remarks: *Stropharia coronilla* was recorded by Watling & Gregory (1987) with conspicuously germ-pored spores, but our collections agree with those recorded by Breitenbach & Kränzlin (1995) and Noordeloos (1999), whose collections were without a conspicuous germ-pore, from Switzerland and Netherlands respectively. *Stropharia coronilla* belongs to Section *Mundae* (Fr.) Konr. & Maubl. due the subviscid to dry pileus, the grooved annulus and the presence of true chrysocystidia (Noordeloos 1999). Some authors cite *S. coronilla* as a psilocin containing agaric (Sticht & Käferstein 2000), but Stijve & de Meijer (1993), who chemically analyzed southern Brazilian collections, and Ballero & Contu (1998) found neither psilocin nor psilocybin. In Brazil, *S. coronilla* has been previously recorded from the states of Pernambuco (Batista & Bezerra 1960), Paraná (Stijve & de Meijer 1993), and Rio Grande do Sul (Rick 1907, 1939, 1961; Singer 1953).

9. *Stropharia rugosoannulata* Farl. ex Murrill, Mycologia 14: 139, 1922.
Figure 8 A-E

Pileus 55-77(-150) mm diam., convex to applanate, robust; surface humid, smooth to slightly squamulose at the margin; color dark vinaceous brown, margin encurved, ondulate, with abundant appendiculate velar remnants; context fleshy and thick, white. **Lamellae** adnate, membranous; grayish to vinaceous brown; edges crenulated, whitish. **Stipe** 61-117 x 9-12 mm, central; cylindrical to sub-bulbous at base, with conspicuous basal mycelium; solid, striate above the annulus; white to cream. **Veil** well developed, forming a persistent, white, grooved, fleshy, thick, and striate annulus, and velar remnants at the pileus margin.

Basidiospores dark vinaceous brown in mass; 9.5-14.5 x 6.5-7 μm, subhexagonal in frontal view and ellipsoid in side view; smooth and thick-walled, with a wide germ-pore, brown. **Basidia** 21.5-38 x 8-11 μm, ventricose, with a medial constriction, tetrasporic. **Pleurocystidia** 24-53 x 6.5-13.5 μm, clavate to fusiform, as true chrysocystidia, with colored amorphous content. **Cheilocystidia** 22.5-45 x 6.5-17.5 μm, fusoid to mucronate, also as chrysocystidia, somewhat smaller than the pleurocystidia.

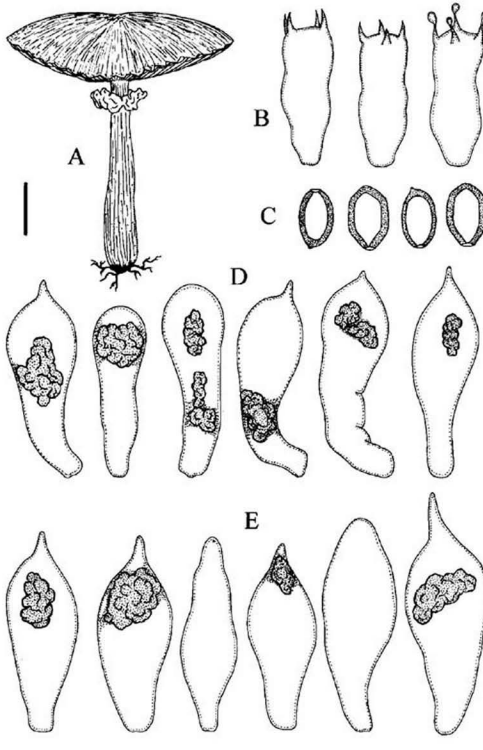

Figure 8. ***Stropharia rugosoannulata*****:** A. Basidiome. B. Basidia. C. Basidiospores. D. Chrysocystidia. E. Cheilocystidia. Scale bar: A = 20 mm; B-E = 10 μm.

Pileipellis is an ixocutis, composed of prostrate hyphae, incrusted with brownish pigment. **Hymenophoral trama** regular. **Clamp connections** present.

Ecology: Terrestrial, solitary or gregarious on soil and litter, in deciduous forests.

Material examined: BRAZIL. Rio Grande do Sul: **Santa Maria**, Passo dos Ferreiros, 17.IV.2001 *Cortez* 018/01 (SMDB 9241); Morro do Elefante, 9.V.2001 *Cortez* 027/1 (SMDB 9247), 9.V.2001 *Cortez* 028/01 (SMDB 9248).

Distribution: North America (Murrill 1922), South America (Wright & Albertó 2002), Europe (Guinberteau 1978; Moser 1978; Wasser & Grodzinskaya 1996; Watling & Gregory 1987).

Remarks: *Stropharia rugosoannulata* is a good edible species, well studied and cultivated in many European countries (Singer 1986). Medicinal properties have been investigated by Grodzinskaya et al. (1999). There are only sporadic records of this species from South America. Wright & Albertó (2002) recorded it from Argentina, where seems to be used as food. In Brazil, *S. rugosoannulata* is not well documented and not consumed as food; previous records are from the states of São Paulo (Pegler 1997; Singer 1950) and Paraná (Stijve & de Meijer 1993). This is the first record of *S. rugosoannulata* from Rio Grande do Sul.

10. *Stropharia semiglobata* (Batsch: Fr.) Quél., Mem. Soc. d'Émul. Mont. Ser. II, 5: 112, 1872. FIGURE 9 A-E

Pileus 35 mm diam., hemispheric, somewhat umbonate; surface subviscid, smooth, pale yellow colored, margin striate, context fleshy, unchanging. **Lamellae** adnate, grayish to dark vinaceous brown, edges whitish. **Stipe** 104 x 4 mm, central, cylindrical, slightly expanded at base, elongated; color similar to pileus, subviscid. **Veil** forming a yellowish, glutinous annulus, at the upper part of the stipe.

Basidiospores dark vinaceous brown in mass; 17.5-21 x 9.5-12 μm, ellipsoid both in frontal and side view; brown, smooth and thick-walled, truncate by a broad germ-pore. **Basidia** 35-48 x 13.5-17 μm, clavate, hyaline, tetrasporic. **Pleurocystidia** 29.5-50.5 x 13-19 μm, mucronate to clavate, as true chrysocystidia, with amorphous refringent contents. **Cheilocystidia** 25.5-32 x 8-9.5 μm, hyaline, lageniform, abundant in the gill edge. **Pileipellis** an ixocutis of parallel and prostrate hyphae. **Hymenophoral trama** regular. **Clamp connections** present.

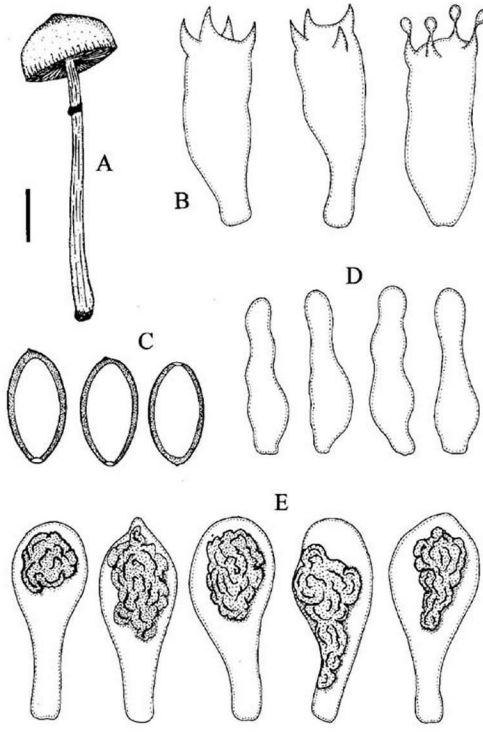

Figure 9. ***Stropharia semiglobata*:** A. Basidiome. B. Basidia. C. Basidiospores. D. Chrysocystidia. E. Cheilocystidia. Scale bar: A = 20 mm; B-E = 10μm.

Ecology: Coprophilous, solitary on horse dung, in pastures.

Specimens examined: BRAZIL. Rio Grande do Sul: **Santa Maria,** Camobi, margin of the Vacacaí-Mirim river, near the Morro do Elefante, 23.IX.2000 *Cortez* 048/00 (SMDB, material lost).

Distribution: Europe (Breitenbach & Kränzlin 1995; Cacialli et al. 1995; Esteve-Raventós & Barrasa 1995; Kytövuory 1999; Moser 1978; Noordeloos 1999; Watling & Gregory 1987), North America (Lincoff 1988; Murrill 1922; Stamets 1996), South America (Dennis 1970; Pulido 1983; Singer & Moser 1964; Wright & Albertó 2002; Yokoyama 1984).

Remarks: *Stropharia semiglobata* is cited in the literature (e.g., Wright & Albertó, 2002) as a very common species on dung, but we have collected it only once during our study. It is very similar to *S. dorsipora* Esteve-Rav. & Barrasa, a European taxon that produces basidiospores with an eccentric germ pore, in contrast to the central germ pore found in *S. semiglobata* (Esteve-Raventós & Barrasa 1995; Noordeloos 1999). *Stropharia semiglobata* is known from the states of São Paulo (Bononi et al. 1984; Pegler 1997) and Rio Grande do Sul (Rick 1907, 1939, 1961).

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Megasporoporia (Aphyllophorales, Basidiomycota*)* in China

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Abstract—The knowledge on the species of *Megasporoporia (Aphyllophorales,* Basidiomycota*)* from China is summarised in this paper, and four species in the genus are recorded from the country. An identification key, including statistical variations of the spore dimensions of each species, is supplied. Two new species, *Megasporoporia quercina* and *M. subcavernulosa*, are described and illustrated. *M. quercina* is characterized by its perennial habit, extensive basidiocarps, the abundance of hyphal pegs at dissepiment edges and tube walls, smaller basidiospores, and by its growth on fallen trunks of *Quercus*. *M. subcavernulosa* resembles *M. cavernulosa*, but differs by having smaller basidiospores, and by the presence of both hyphal pegs and dendrohyphidia. Moreover, *M. cavernulosa* is widely distributed in tropical America and Africa, while *M. subcavernulosa* is found in temperate and warm temperate areas of East Asia. Specimens of *M. setulosa* collected from China were studied, and their basidiospores are smaller than in specimens from tropical America.

Key words—polypores, taxonomy, wood-rotting fungi

Introduction

Megasporoporia Ryvarden & J.E. Wright was established with *Poria setulosa* Henn. as the type species (Ryvarden et al. 1982). The genus is characterized by large pores, long and cylindric basidiospores, clamp connections on generative hyphae, and by dextrinoid skeletal hyphae with arboriform branches (the so-called skeleto-binding hyphae). Because the skeletal hyphae are dentritically branched in *Megasporoporia*, a hyphal structure similar to that in the genera *Dichomitus* D.A. Reid and *Polyporus* P. Micheli ex Adams.:Fr. *sensu str.*, *Megasporoporia* was recently merged in *Dichomitus* (Masuka & Ryvarden 1999). However, *Megasporoporia* usually has shallow pores, plenty of hyphal pegs, dextrinoid skeletal hyphae, dendrohyphidia, and polyhedric crystals in most species. Taxa sharing these characters constitute in

our opinion a natural group, and in the present study *Megasporoporia* is treated as an independent genus.

Before this study, two species of *Megasporoporia* were recorded from China (Hattori 1995, Dai & Li 2002). During the survey of wood-rotting fungi in China, more specimens belonging to this genus were studied, and four species, including two new ones, were found. In this paper we describe the new taxa, and give notes on some other species.

Materials and methods

This study is mainly based on the following materials: the collections of the authors from the Yunnan and Hainan provinces of China; specimens from other parts of China collected by Tsutomu Hattori (Japan, TFM-F), Guo-Yang Zheng (China, HMIGD) and Emilio Licent (the Czech Republic, PRM). The herbaria of the studied materials are mentioned in the text. For comparison, some American collections were studied to support our results.

The microscopic routine used in the study is as presented by Dai and Niemelä (1997). In the text the following abbreviations are used: L = mean spore length (arithmetical mean of all spores), W = mean spore width (arithmetical mean of all spores), Q = variation in the L/W ratios between the specimens studied (quotient of the mean spore length and the mean spore width of each specimen), n=(x/y) means the number (x) of spores (or other structures) measured from given number (y) of specimens. In presenting the variation in the size of spores, 5% of the measurements were excluded from each end of the range, and are given in parentheses; IKI stands for Melzer's reagent and KOH for 5% potassium hydroxide, and CB is the abbreviation of Cotton Blue. CB+ means cyanophilous and CB– acyanophilous; IKI+ means amyloid and IKI– means both inamyloid and indextrinoid. The authors of the scientific names were abbreviated mostly according to Brummitt and Powell (1992).

Results

Descriptions

Megasporoporia quercina Y.C. Dai, *sp. nov.* (Figs. 1-2)

Carpophorum perenne, resupinatum. Facies pororum cremea vel pallide ravido-cremea; pori 1–2 per mm. Systema hypharum dimiticum, hyphae generatoriae fibulatae, hyphae skeletales subiculi 2.2 –3.5 µm in diam. Sporae hyalinae, cylindricae, 5.6–8 × 2.3–3 µm.

Type: China. Yunnan Province, Lijiang County, Heishuihe, mixed broad-leaved forest, on fallen decorticated trunk of *Quercus*, 15.VI.1999 *Dai 3054* (holotype in IFP; isotype in H).

Etymology. — *quercina* (Lat.): referring to the host tree genus *Quercus*.

Fruitbody. — Basidiocarps perennial, resupinate, difficult to separate from substrate, leathery when fresh, becoming hard upon drying, up to 150 cm long, 15 cm wide, and 4 mm thick. Pore surface cream to pale greyish cream when fresh, becoming greyish cream to pale straw coloured upon drying; pores mostly round, sometimes slightly irpicoid, freely arranged, 1–2 per mm; dissepiments thin, lacerate, bearing abundant hyphal pegs (easily seen by lens). Subiculum pale straw coloured, hard corky, up to 0.5 mm thick. Tubes concolorous with poroid surface, tubes hard, up to 3 mm long, tube walls bearing crowded hyphal pegs, tubes stratified, tube layers usually distinct.

Hyphal structure. — Hyphal system dimitic, generative hyphae with clamp connections, hyaline, thin-walled; skeletal hyphae thick-walled with a narrow lumen to subsolid, unchanged in KOH, dextrinoid, CB+.

Subiculum. — Hyphae of the context tightly interwoven, agglutinated; generative hyphae infrequent, occasionally branched, 2–4 µm in diam; skeletal hyphae dominant, thick-walled, flexuous, dentritically branched, 2.2–3.5 µm in diam.

Tubes. — Tramal hyphae tightly interwoven, strongly agglutinated; generative hyphae infrequent, thin-walled, occasionally branched, 2–3.5 µm in diam; skeletal hyphae dominant, mostly subsolid, dentritically branched, 2–3.3 µm in diam. Cystidia and cystidioles absent, hyphal pegs frequent, some of them submerged in trama, mostly penetrating into hymenium. Hyphae of hyphal pegs hyaline, thick-walled, straight, strongly agglutinated, parallel along the peg, strongly dextrinoid, CB+, 2–3 µm in diam. Basidia narrowly clavate, with a basal clamp and four sterigmata, 20–26 × 4–6 µm; basidioles in shape similar to basidia, but slightly smaller. Polyhedric crystals frequent among subhymenium and hymenium. Dendrohyphidia frequent in hymenium and the edges of dissepiments.

Spores. — Basidiospores cylindrical, hyaline, thin-walled, smooth, glued in tetrads, CB–, IKI–, (5.1–)5.6–8(–8.2) × (2.1–)2.3–3(–3.5) µm, L = 6.56 µm, W = 2.71 µm, Q = 2.42 (n=60/1).

Five species of *Megasporoporia* were detected: *M. cavernulosa* (Berk.) Ryvarden, *M. setulosa* (Henn.) Rajchenb., *M. hexagonoides* (Speg.) J.E. Wright & Rajchenb., *M. mexicana* Ryvarden and *M. major* (G.Y. Zheng & Z.S. Bi) Y.C. Dai & T.H. Li. Among them the first two species have shorter spores (10–16 × 5–7 µm in *M. cavernulosa* (Ryvarden et al. 1982) and 8–11 × 3.5–4.2 µm in *M. setulosa*), and the three last-mentioned species have long spores (16.6–21.8 × 5.2–6.8 µm in *M. hexagonoides* and 20–26 x 6–9 µm in *M. mexicana*, Ryvarden et al. 1982; 15.2–20 × 5.5–7.1 µm in *M. major*).

Megasporoporia quercina differs mainly from the other species of *Megasporoporia* by its distinctly smaller basidiospores, and it is perennial, in contrast to the annual habit in the other species of the genus. In addition, *M. quercina* produces extensive basidiocarps (up to 150 cm long, 15 cm wide) on large decorticated trunks of *Quercus*, but the other species of the genus usually form just small patches on fallen branches of angiosperm trees. Another distinct character of the new species is the abundance of its hyphal pegs in both dissepiments and tube walls; the hyphae of the pegs are strongly dextrinoid.

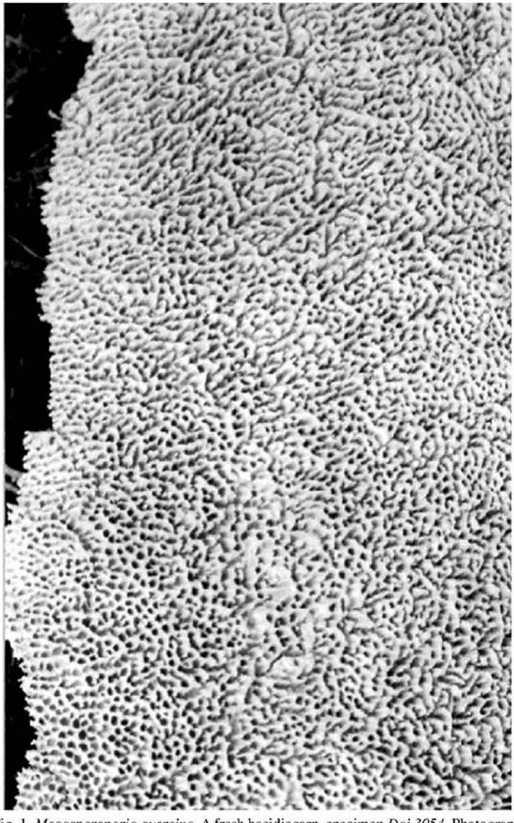

Fig. 1. *Megasporoporia quercina*. A fresh basidiocarp, specimen *Dai 3054*. Photograph Y.C. Dai *in situ*, × 1.1 .

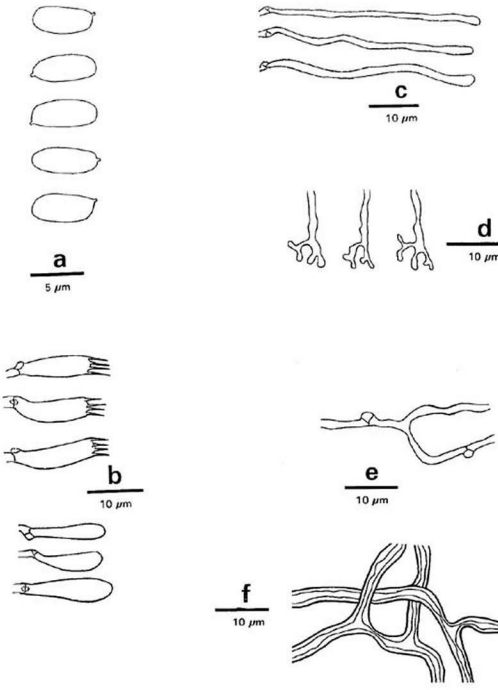

Fig. 2. Anatomical details of *Megasporoporia quercina* (drawn from the holotype). — a: Basidiospores. — b: Basidia and basidioles. — c: Cystidioles. — d: Dendrohyphidia from dissepiment edge. — e: Generative hyphae from subiculum. — f: Skeletal hyphae from subiculum.

Dichomitus eucalypti Ryvarden was described on *Eucalyptus* from Australia (Ryvarden 1985). Its basidiospores are rather similar to those in *Megasporoporia quercina*, but the former species has smaller pores (2–3 per mm), and it lacks dendrohyphidia and hyphal pegs. Its basidiospores are wider, 7–8.5 × 3–4 µm, and its skeletal hyphae are negative in Melzer's reagent.

Grammothele Berk. & M.A. Curtis is closely related to *Megasporoporia*, and species of the latter genus were for some time included in the former one (Ryvarden & Johansen 1980). Species having large pores, big basidiospores, and a distinctly poroid hymenophore were later separated from *Grammothele*, and they were treated in *Megasporoporia* by Ryvarden et al. (1982). Basidiospores of *M. quercina* are not as large as in most species of *Megasporoporia*, and they are closer to the spores of *Grammothele*, and so *M. quercina* seems to be an intermediate species between *Grammothele* and *Megasporoporia*. However, the new species has distinct pores, cylindrical basidiospores, and its skeletal hyphae are distinctly branched and dextrinoid. These characters indicate it to be closely related to *M. setulosa* and the other species of the genus, and therefore we treat it accordingly.

Additional specimen examined (paratype). — China. Yunnan Prov., Lijiang County, Heishuihe, mixed broad-leaved forest, on fallen decorticated trunk of *Quercus*, 15.VI.1999 *Dai 3052* (IFP).

Megasporoporia subcavernulosa Y.C. Dai & Sheng H. Wu, *sp. nov.* (Fig. 3)

Carpophorum annuum, resupinatum. Facies pororum cremea vel pallide ravida; pori 2–4 per mm. Systema hypharum dimiticum, hyphae generatoriae fibulatae, hyphae skeletales subiculi 2–3.8 µm in diam. Sporae hyalinae, cylindricae, 9–12.1 x 4.2–5.2 µm.

Type: China. Yunnan Prov., Kunming, Xishan Park, on fallen twig of *Cyclobalanopsis*, 25.VIII.1995 *Wu 9508–328* (holotype in TNM).

Etymology. — *subcavernulosa* (Lat.): somewhat resembling *M. cavernulosa*.

Fruitbody. — Basidiocarps annual, resupinate, difficult to separate from substrate, leathery when fresh, becoming hard corky upon drying, up to 7 cm long, 1 cm wide, and 1.5 mm thick. Pore surface cream when fresh, becoming pale greyish upon drying; pores round, freely arranged, 2–4 per mm; dissepiments thin, entire. Subiculum cream, corky, up to 0.5 mm thick. Tube layer concolorous with pore surface, tubes hard corky, up to 1 mm long.

Hyphal structure. — Hyphal system dimitic, generative hyphae with clamp connections, hyaline, thin-walled; skeletal hyphae thick-walled with a narrow lumen or subsolid, unchanged in KOH, dextrinoid, CB+.

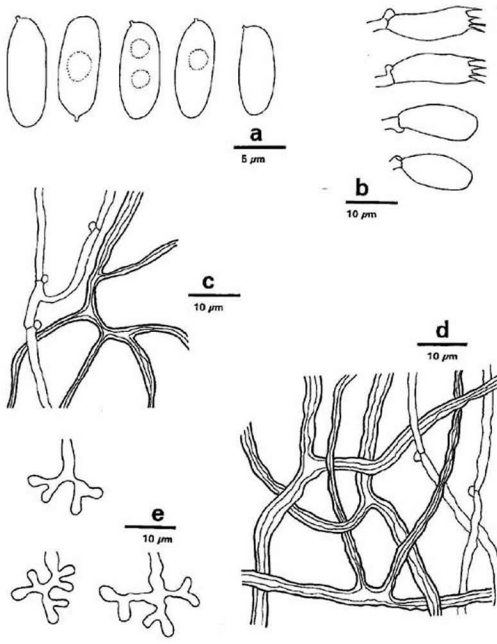

Fig. 3. Anatomical details of *Megasporoporia subcavernulosa* (drawn from the holotype). — a: Basidiospores. —b: Basidia and basidioles. —c: Hyphae from trama. —d: Hyphae from subiculum. —e: Dendrohyphidia from dissepiment edge.

Subiculum. — Hyphae in subiculum tightly interwoven, agglutinated; generative hyphae infrequent, hyaline, thin-walled, 2–3.5 µm in diam; skeletal hyphae dominant, thick-walled, occasionally branched in the apical parts, flexuous, some encrusted, 2–3.8 µm in diam.

Tubes. — Tramal hyphae tightly interwoven, strongly agglutinated; generative hyphae infrequent, thin-walled, 1.8–3.3 µm in diam; skeletal hyphae dominant, mostly subsolid, branched in an arboriform fashion, 2–3.5 µm in diam. Cystidia and cystidioles absent, hyphal pegs frequent; hyphae of pegs hyaline, thin-walled, frequently septate with clamp connections, weakly dextrinoid, slightly CB+. Basidia clavate, with a basal clamp and four sterigmata, 18–24 × 8–11 µm; basidioles in shape similar to basidia, but distinctly smaller. Polyhedric crystals frequent among subhymenium and hymenial elements. Dendrohyphidia frequent in hymenium and the edges of dissepiments.

Spores. — Basidiospores cylindrical, hyaline, thin-walled, smooth, CB–, IKI–, (8.5–)9–12.1(–13.2) × (4–)4.2–5.2(–6) µm, L = 10.27 µm, W = 4.77 µm, Q = 2.09–2.42 (n=120/4).

Megasporoporia subcavernulosa was first recorded as *Poria ambigua* Bres. (*Oxyporus latemarginatus* (Dur. & Mont.) Donk) from the Shanxi Province in northern China (Pilát 1940). However, after examining its voucher specimen, it turned out to be a distinct species of *Megasporoporia*. *M. cavernulosa* (Berk.) Ryvarden was reported from Zhejiang Province, eastern China (Hattori 1995). Its cited material was studied, and it has both hyphal pegs and dendrohyphidia. In addition, basidiospores of the specimen are distinctly smaller than in *M. cavernulosa* (9.2 –11 × 4.3–5.1 µm vs. 10–16 × 5–7 µm, Ryvarden et al. 1982). Furthermore, specimens from Yunnan (southwestern China) and Primorye (the Russian Far East) were studied, they are identical to the material collected from Zhejiang and Shanxi. The new species, *M. subcavernulosa*, is described here on the basis of these materials. *M. subcavernulosa* is closely related to *M. cavernulosa*, but differs by having smaller basidiospores, and by having both hyphal pegs and dendrohyphidia. Furthermore, *M. cavernulosa* is widely distributed in tropical America and Africa, while *M. subcavernulosa* is found in the temperate and warm temperate areas of East Asia only.

Additional specimens examined (paratypes). — China. Shanxi Prov., Yaochan, on angiosperm, 2.IX.1935 *Licent 4925* (PRM 806786). Zhejiang Prov., Baishanzu Mts., VII.1994 *Hattori* (TFM-F). Russia. Primorye Terr., Hasan, 23.VII.1985 *Parmasto* (TAA 106222).

Key to species of *Megasporoporia* from China

(statistical variations of spore dimensions of each species are included)

1. Basidiospores < 8.5 µm in length . . . . . *M. quercina*
   (5.1–)5.6–8(–8.2) × (2.1–)2.3–3(–3.5) µm,
   L = 6.56 µm, W = 2.71 µm, Q = 2.42 (n=60/1)
1. Basidiospores > 8.5 µm in length 2

2. Basidiospores > 14 µm in length . . . . . *M. major*
(14.7–)15.2–20(–22) × (5–)5.5–7.1(–7.5) µm,
L = 17.82 µm, W = 6.57 µm, Q = 2.72 (n=60/1)
2. Basidiospores < 14 µm in length 3

3. Pores 2–4 per mm; dendrohyphidia present along hymenium and in dissepiment edges . . . . . *M. subcavernulosa*
(8.5–)9–12.1(–13.2) × (4–)4.2–5.2(–6) µm,
L = 10.27 µm, W = 4.77 µm, Q = 2.09–2.42 (n=120/4)
3. Pores 1–2 per mm; dendrohyphidia absent from hymenium and in dissepiment edges . . . . . *M. setulosa*
(7.2–)8–11(–11.2) × (3.1–)3.5–4.2(–4.5) µm,
L = 9.24 µm, W = 3.77 µm, Q = 2.45 (n=30/1)

Notes on other species

Two specimens of *Megasporoporia setulosa* from Taiwan and Hainan were studied; basidiospores of the Chinese materials are smaller than those from tropical America (8–11 x 3.5–4.2 µm vs. 10–14 x 4–6 µm, Gilbertson and Ryvarden, 1986–1987), but all the other characters fit the species well. *M. setulosa* is similar to *M. cavernulosa*, but differs from the latter one by having larger pores, and by lacking dendrohyphidia.

Megasporoporia *major* was originally described from the Guangdong Province, subtropical China, as *Pachykytospora major* G.Y. Zheng & Z.S. Bi. Dai and Li (2002) merged it in *Megasporoporia* because it has smooth basidiospores, and its hyphal structure is very similar to that in the other *Megasporoporia* species.

Other *specimens examined*. — *Megasporoporia major*. China. Guangdong Prov., Huidong County, Gutian, on branch of angiosperm, 26.XI.1986 *Zheng* (HMIGD). *Megasporoporia setulosa*. China. Hainan Prov., Ledong County, Jianfengling Nat. Res., on fallen branch of angiosperm, 21.IX.2002 *Dai 4373* (IFP). Taiwan, Gaoxiong, Liukuei, Shanping, on branch of angiosperm, 22.12.1993 *Wu 9312-54* (TNM). Jamaica, Trewlany Crowlands, on dead wood, 10.VI.1999 *Ryvarden 41574* (O).

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Notes on the genus *Antrodiella* (Basidiomycota, *Aphyllophorales*) in China

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Abstract—Antrodiella micra (Basidiomycota, Aphyllophorales) from northeastern China is described and illustrated. It has resupinate or effused-reflexed basidiocarps, snow-white hymenophore when fresh, small pores (7–9 per mm), small and broadly ellipsoid basidiospores, and it grows on wood of angiosperms decayed by some species of the Hymenochaetaceae. The new species resembles A. pallasii, which has pale yellowish pore surface when fresh, and somewhat larger pores (6–7 per mm). In addition, A. pallasii usually grows on spruce trees decayed by Trichaptum abietinum. An identification key to the species of Antrodiella from China is provided along with a synoptic description of each species.

Key words—polypores, taxonomy, wood-rotting fungi

Introduction

The genus *Antrodiella* Ryvarden & I. Johans. was established by Ryvarden and Johansen (1980) to embrace the *Polyporus semisupinus* Berk. & M.A. Curtis complex. The species of *Antrodiella* usually have small pores, light colours, resupinate to pileate basidiocarps with a dimitic or trimitic hyphal system, small basidiospores, but no cystidia, and they cause a white rot of dead wood. The genus was originally defined mostly by European and North American species. *Antrodiella* has been intensively studied during the past 20 years, and more species have been subsequently described in or transferred into the genus (Vampola 1991, Hattori & Ryvarden 1993, Ryvarden & Gilbertson 1993, Vampola & Pouzar 1996, Dai & Niemelä 1997, Johannesson, Renvall & Stenlid 2000). The added species have greatly expanded the original definition of the genus by including species with irpicoid to dentate hymenophore, e.g. *A. foliaceodentata* (Nikol.) Gilb. & Ryvarden and *A. zonata* (Berk.) Ryvarden; species with large pores, e.g. *A. americana* Ryvarden & Gilb. and *A. aurantilaeta* (Corner) T. Hattori & Ryvarden; species with yellow colouration, e.g. *A. albocinnamomea* Y.C. Dai & Niemelä and *A. citrinella* Niemelä & Ryvarden; species having hymenial cystidia, e.g. *A. albocinnamomea* and *A.*

parasitica Vampola; and species with a stipe, e.g. *A. liebmannii* (Fr.) Ryvarden. However, an important and consistent character in all species of the genus is the cyanophilous colour reaction of the skeletal hyphae.

Species of *Antrodiella* from Northeast China were reported by Dai and Niemelä (1997). Subsequently more material from other parts of China was studied, and 12 species, including one previously undescribed, are now recognized. In this paper I give an illustrated description of the new species, and compile a key for the 12 species of my research area. A synoptic description of each species is given in the key, along with data on spore size variation measured mostly from the Chinese material. Other species of *Antrodiella* from East Asia were previously summarized by Núñez and Ryvarden (2001).

Materials and methods

Most of the material in the present study was collected by the author from Northeast, Southwest and Central China during field trips in 1993–2002; some specimens were collected by Marja Härkönen (Helsinki, Finland) from Hunan Province, subtropical China. The specimens studied are preserved in the Herbarium of the Institute of Applied Ecology, Academia Sinica (IFP), and duplicates are deposited in the Botanical Museum of the University of Helsinki (H). Some specimens were obtained as loans from HMAS (China), TFM (Japan), BPI (USA) and PRM (Czech Republic). For comparison some European and North American specimens were studied. The microscopic routine used was that described by Dai and Niemelä (1997). In the text the following abbreviations are used: Frb. = Fruitbody, L = mean spore length (arithmetical mean of all spores), W = mean spore width (arithmetical mean of all spores), Q = variation in the L/W ratios between the specimens studied (quotient of the mean spore length and the mean spore width of each specimen), n=x/y means the number (x) of spores (or other structures) measured from given number (y) of specimens. In presenting variation in the size measurements of spores, hyphae, pores and spines, 5% of the measurements were excluded from each end of the range and are given in parentheses. IKI stands for Melzer's reagent, KOH for 5% potassium hydroxide, and CB for Cotton Blue. CB+ means cyanophilous and CB– acyanophilous; IKI– means both inamyloid and indextrinoid. The authors of scientific names have been abbreviated mostly according to Brummitt and Powell (1992).

Results

Key to species of *Antrodiella* from China

(synoptic description of each species is provided)

1. Hymenophore irpicoid to dentate . . . . . *A. zonata*
   Frb. annual, mostly pileate, sometimes effused-reflexed, upper surface zonate; hymenophore pale yellowish, becoming yellowish brown with age, irpicoid or toothed, spines 2–4 per mm; tramal hyphae parallel along tubes, basidiospores oblong-ellipsoid, IKI–, CB–,
   (3.8–)4.4–6(–6.5) × (2.7–)3–4(–4.3) µm,
   L = 5.14 µm, W = 3.39 µm, Q = 1.44–1.59 (n=90/3).

1. Hymenophore poroid 2

2. Pores 1–3 per mm 3
2. Pores > 3 per mm 4

3. Basidiocarps resupinate, pores cream coloured . . . . . *A. americana*
Frb. annual, resupinate; pore surface cream coloured, pores 1–2 per mm; tramal hyphae interwoven, gloeocystidia present, basidiospores oblong-ellipsoid, IKI–, CB–,
(2.9–)3–4(–4.5) × (1.2–)1.4–2(–2.2) µm,
L = 3.50 µm, W = 1.82 µm, Q = 1.77–2.18 (n=120/4).
3. Basidiocarps effused-reflexed, pores orange coloured . . . . . *A. aurantilaeta*
Frb. annual, effused-reflexed; pore surface orange yellow, pores 1–2 per mm; tramal hyphae interwoven, basidiospores oblong-ellipsoid, IKI–, CB–,
(2.7–)2.8–4.3(–5.2) × (1.4–)1.7–2.2(–2.3) µm,
L = 3.38 µm, W = 1.95 µm, Q = 1.57–1.91 (n=66/2).

4. Pileus brown to deep bay, context brown and darker than tubes . . . *A. liebmannii*
Frb. annual to perennial, sessile or stipitate, upper surface brown to purplish black, zonate; pore surface tan to dull straw coloured, pores 14–16 per mm; tramal hyphae loosely parallel, basidiospores ellipsoid, IKI–, CB–,
(2.7–)2.8–3.8(–3.9) × (1.4–)1.5–2(–2.1) µm,
L = 3.10 µm, W = 1.73 µm, Q = 1.79 (n=30/1).
4. Pileus cream or straw coloured, tan or yellowish, context concolorous with the tubes 5

5. Pore surface yellowish 6
5. Pore surface cream coloured, never yellowish 9

6. On gymnosperms; basidiospores broadly ellipsoid to subglobose 7
6. On angiosperms; basidiospores oblong-ellipsoid to subcylindrical 8

7. On white rot wood decayed by *Trichaptum*; basidiocarps mostly > 5 cm in longest dimension, pores 6–7 per mm; basidiospores usually bearing a guttule, < 2.1 µm in width . . . . . *A. pallasii* Renvall, Johannesson & Stenlid
Frb. annual, resupinate or effused-reflexed; pore surface cream coloured to pale yellowish, pores 6–7 per mm; tramal hyphae interwoven, basidiospores broadly ellipsoid, IKI–, CB–,
(2.7–)2.9–3.3(–3.7) × (1.8–)1.9–2.1(–2.2) µm,
L = 2.97 µm, W = 1.90 µm, Q = 1.56 (n=30/1).
7. On brown rot wood decayed by *Fomitopsis pinicola;* basidiocarps mostly < 5 cm in longest dimension, pores 4–5 per mm; basidiospores usually without guttules, > 2.1 µm in width . . . . . *A. citrinella*
Frb. annual, resupinate or effused-reflexed; pore surface cream coloured to citric yellow, pores 4–5 per mm; tramal hyphae interwoven, basidiospores broadly ellipsoid, IKI–, CB–,
(2.9–)3–3.9(–4) × (2–)2.1–2.9(–3) µm,
L = 3.55 µm, W = 2.38 µm, Q = 1.49 (n=30/1).

8. Basidiocarps resupinate; cystidia present, basidiospores > 2 µm in width . . . . . *A. albocinnamomea*

Frb. annual, resupinate; pore surface cream to cinnamon coloured, pores 3–5 per mm; tramal hyphae interwoven, pyriform cystidia present, basidiospores oblong-ellipsoid, IKI–, CB–,

(3–)3.7–5(–5.5) × (2–)2.1–2.9(–3.3) µm,

L = 4.18 µm, W = 2.38 µm, Q = 1.61–1.84 (n=183/6).

8. Basidiocarps resupinate or effused-reflexed; cystidia absent, basidiospores < 2 µm in width . . . . . A. *ussurii* Y.C. Dai & Niemelä

Frb. annual, pileate or effused-reflexed, upper surface pale tan to brownish; pore surface cream coloured to pale buff, pores 6–8 per mm; tramal hyphae interwoven, basidiospores oblong-ellipsoid, IKI–, CB–,

(2.9–)3–4(–4.7) × (1.1–)1.2–2(–2.1) µm,

L = 3.50 µm, W = 1.54 µm, Q = 2.17–2.54 (n=260/8).

9. On gymnosperms; subulate cystidia present . . . . . *A. gypsea* (Yasuda) T. Hattori & Ryvarden

Frb. annual to perennial, resupinate to effused-reflexed or pileate, upper surface cream coloured to pale greyish; pore surface white to cream coloured, pores 6–8 per mm; tramal hyphae interwoven, subulate cystidia present, basidiospores oblong-ellipsoid, IKI–, CB–,

(2.5–)2.6–3(–3.1) × 1.2–1.7(–1.8) µm,

L = 2.90 µm, W = 1.37 µm, Q = 2.11 (n=30/1).

9. On angiosperms; cystidia absent 10

10. Basidiocarps usually pileate, occasionally effused-reflexed . . . . . *A.* cf. *semisupina* (Berk. & M.A. Curtis) Ryvarden

Frb. annual, pileate, upper surface and pore surface cream coloured, pores 6–8 per mm; tramal hyphae interwoven, basidiospores ellipsoid, IKI–, CB–,

(2.5–)2.8–3.8(–4) × (1.7–)1.9–2.5 µm,

L = 3.13 µm, W = 2.10 µm, Q = 1.49–1.50 (n=60/2).

10. Basidiocarps usually resupinate, occasionally effused-reflexed 11

11. Basidiocarps not associated with *Hymenochaetaceae*, pores 4–5 per mm . . . . . *A. romellii* (Donk) Niemelä

Frb. annual, resupinate; pore surface cream to pale straw coloured, pores 4–5 per mm; tramal hyphae interwoven, basidiospores broadly ellipsoid, IKI–, CB–,

(3.3–)3.5–4(–4.1) × (2–)2.1–2.7(–2.9) µm,

L = 3.85 µm, W = 2.35 µm, Q = 1.64 (n=30/1).

11. Basidiocarps usually associated with species of *Hymenochaetaceae*, pores 7–9 per mm . . . . . *A.micra*

Frb. annual, resupinate or effused-reflexed; pore surface snow white when fresh, ochraceous when dry, pores 7–9 per mm; tramal hyphae interwoven, basidiospores broadly ellipsoid, IKI–, CB–,

(2.7–)2.9–3.5 (–3.8) × (1.7–)1.9–2.2(–2.3) µm,

L = 3.12 µm, W = 2.05 µm, Q=1.48–1.57 (n=60/2).

Description

Antrodiella micra Y.C. Dai, *sp. nov.* (Figs.1-2)

Carpophorum annuum, resupinatum vel pileatum, contextum cremeum. Facies pororum cremea; pori 7–9 per mm. Systema hypharum dimiticum, hyphae generatoriae fibulatae, hyphae skeletales subiculi 3.1–4.4 µm in diam. Sporae perlato-ellipsoideae, 2.9–3.5 x 1.9–2.2 µm.

Type: China. Jilin Prov., Antu County, Changbaishan Nat. Res., rotten wood of *Populus*, associated with *Phellinus gilvus* (Schwein.:Fr.) Pat., 19.IX.1998 *Dai 2998, Niemelä & Qin* (holotype in IFP, isotype in H).

Etymology. — *micra* (Lat.): referring to small pores.

Fruitbody. — Basidiocarps annual, mostly resupinate, easily detachable, at first emerging as small patches which fuse together with age, resupinate part up to 25 cm long or more in longest dimension and 6 cm wide, when fresh elastic tough, without odour or taste, when dry becoming corky. Pileus projecting up to 3 mm, 2 cm wide, and 0.5 mm thick. Pileal surface cream, azonate, smooth; margin acute, curving down upon drying. Pore surface snow white when fresh, becoming pale buff when bruised, and pale ochraceous when dry; margin cottony, cream coloured to white, up to 1 mm wide; pores round, (6–)7–9 per mm (n=50/2), dissepiments thin, even to slightly lacerate. Context cream, corky, ca. 0.2 mm thick. Tubes cream, ca. 0.3 mm long.

Hyphal structure. — Hyphal system dimitic, generative hyphae bearing clamp connections, skeletal hyphae dominant, all the hyphae IKI–, CB+, and unchanged in KOH.

Context. — Generative hyphae hyaline, thin-walled, frequently with clamp connections and branched, (1.8–)2–3.6(–4.2) µm in diam (n=30/1); skeletal hyphae thick-walled with a medium or narrow lumen, flexuous, unbranched, agglutinated, (2.7–)3.1–4.4(–4.7) µm in diam (n=30/1).

Tubes. — Tramal hyphae similar to those in context but slightly thinner, skeletal hyphae usually with a lumen, interwoven, more or less agglutinated. Cystidia and cystidioles absent; basidia barrel-shaped, bearing four sterigmata and a basal clamp, (8–)9–11(–12) × (3.5–)4–5(–5.5) µm (n=28/1); basidioles in shape similar to basidia, but slightly smaller. Rhomboid crystals present.

Spores. — Basidiospores broadly ellipsoid, hyaline, thin-walled, smooth, IKI–, CB–, (2.7–)2.9–3.5(–3.8) × (1.7–)1.9–2.2(–2.3) µm, L = 3.12 µm, W = 2.05 µm, Q=1.48–1.57 (n=60/2).

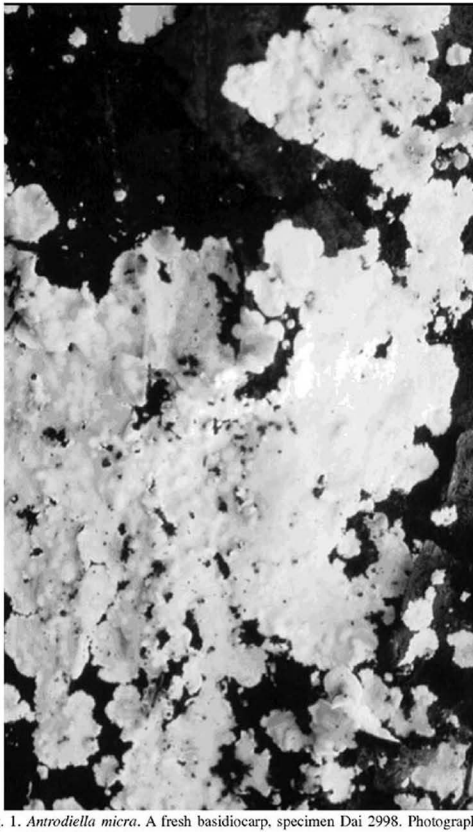

Fig. 1. *Antrodiella micra*. A fresh basidiocarp, specimen Dai 2998. Photograph Y.C. Dai in situ, × 0.6.

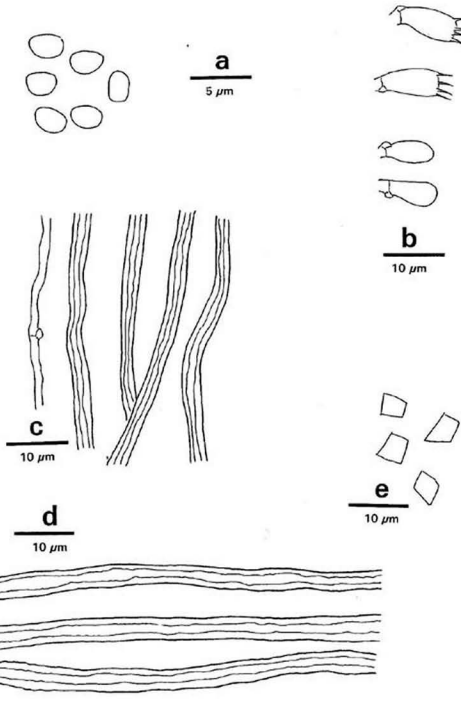

Fig. 2. Anatomical details of *Antrodiella micra* (drawn from the holotype). — a: Basidiospores. — b: Basidia and basidioles. — c: Generative hyphae and skeletal hyphae from trama. — d: Skeletal hyphae from subiculum. — e: Rhomboid crystals

Additional specimen (paratype) examined. — China. Jilin Prov., Fusong County, Shuguang, rotten wood of *Acer* decayed by *Hymenochaete* sp., 21.VII.1993 *Dai 632* (IFP).

The resupinate or effused-reflexed basidiocarps of *Antrodiella micra* have a snow white pore surface, and small pores and spores. It resembles *A. pallasii*, but that species has pale yellowish or sordid cream coloured pores when fresh, and its pores are somewhat larger (6–7 per mm). In addition, *A. pallasii* grows mostly on rotten spruce wood decayed by *Trichaptum abietinum* (Pers.:Fr.) Ryvarden, while the new species grows on wood of angiosperm trees decayed by species of the *Hymenochaetaceae*.

Antrodiella micra is closely related to the species of the *A. romellii* complex, which however have larger pores (4–5 per mm) and bigger basidiospores (3.5–4 × 2.1–2.7 µm), and are not associated with the *Hymenochaetaceae*. Four taxa related to *A. semisupina* have recently been published from Europe and North America (Vampola & Pouzar 1996): *A. faginea* Vampola & Pouzar, *A. thompsonii* Vampola & Pouzar, *A. beschidica* Vampola & Pouzar, and *A. farinacea* Vampola & Pouzar. The first two differ from *A. micra* by having gloeocystidia. *A. beschidica* distinguishes from *A. micra* by its imbricate basidiocarps, somewhat larger pores (5–7 per mm), and oblong-ellipsoid basidiospores. In addition, it grows on *Abies*. Pore size and dimensions of basidiospores of *A. farinacea* are similar to those of *A. micra*, but the spores of *A. farinacea* are curved, dinstinctly tapering at apiculus.

Notes on some other species

Antrodiella liebmannii

A. liebmannii has stipitate basidiocarps, and its upper surface is chestnut to deep bay coloured, making it unusual in the genus of *Antrodiella*. However, it has a dimitic hyphal system, with the generative hyphae bearing clamp connections. Hyphae are strongly agglutinated and cyanophilous. In addition, its basidiospores are small and ellipsoid, hyaline, thin-walled, and negative in Melzer's reagent and Cotton Blue, making it a characteristic species of *Antrodiella*. The spores in the Chinese specimens are longer and narrower than those reported from East Africa (2.8–3.8 × 1.5–2 µm vs. 2–3 × 2–2.5 µm, Ryvarden & Johansen 1980).

Antrodiella zonata

There is a confusion concerning the identity of *A. zonata*, because at least five names (*Irpex cingulatus* Lloyd 1918, *I. consors* Berk. 1877, *I. decurrens* Berk. ex Cooke 1891, *Daedalea gollanii* Maas Geest. 1908 and *I. zonatus* Berk. 1854) are associated with the complex, and most collections are sterile. Cunningham (1965) believed that all the names refer to a single species, while Maas Geesteranus (1974) thought they represented different taxa. Ryvarden (1992) merged all the five species together, and proposed the new combination *A. zonata* based on the priority of *Irpex zonatus*. *I. zonatus* was described on material from Nepal and Sri Lanka, which are

geographically close to my material from Hainan, Yunnan, Guizhou, Sichuan and Hunan. The Chinese specimens are fertile, and the basidiospores are ellipsoid to oblong-ellipsoid, (3.8–)4.2–6(–6.5) × (2.7–)3–4(–4.3) µm, L = 5.10 µm, W = 3.52 µm, Q = 1.44–1.46 (n=60/2). *A. zonata* was also reported from Zhejiang Province, East China (Hattori & Zang 1995), and that material is identical to my specimens. Therefore I believe that all the Chinese collections belong to *A. zonata*. When fresh *A. zonata* has pale yellow to yellowish brown basidiocarps, which become brownish when bruised.

Specimens examined. — *A. aurantilaeta*. China. Guangxi-zhuangzu Auto. Reg., Longlin County, rotten bamboo, 21.X.1957 *Xu 175* (HMAS 28367). Jundong County, rotten wood, 13.VIII.1957 *Xu 1205* (HMAS 28365). Hunan Prov., Sanzhi County, Badagongshan Nat. Res., rotten wood of *Davidia*, 26.IX.1999 *Härkönen 357* (H). Japan. Ibaraki, Kitaibaraki, Ogawa, 25.IX.1989 *Hattori* (TFM-F 15154). — *A. liebmannii*. China. Hainan Prov., Tingan, 31.VIII.1934 *Teng 4001* (BPI 221375), 22.XII.1934 *Teng 8081* (BPI 221590). — *A. pallasii*. China. Heilongjiang Prov., Yichun, Fenglin Nat. Res., rotten *Picea* decayed by *Trichaptum*, 9.IX.2002 *Dai 3717* (IFP). Jilin Prov., Huinan County, Hongqi, rotten wood of *Picea*, 12.X.1993 *Dai 1555b* (IFP). Russia, Bashkortostan, Uchaly Dist., Ikhsanovo, Verkhne-Belskoe, fallen trunk of *Picea*, 23.VIII.2001 *Dai 3323* (H). — *A. romellii*. China. Jilin Prov., Antu County, Changbaishan Nat. Res., fallen branch of *Acer*, 20.IX.2002 *Dai 3867* (IFP). Russia. Siberia, *Betula*, IX.1934 *Krawtzew 115* (PRM 811661). — *A. zonata*. China. Guizhou Province, Shuiyang County, Wangcao, Kuankuoshui Nat. Res., fallen trunk of *Quercus*, 17.VI.2000 *Dai 3220* (IFP). Hainan Prov., Lingshui County, Diaoluoshan Nat. Res., fallen branch of angiosperm, 23.XI.2002 *Dai 4481* (IFP). Hunan Province, Changsha, Yuelushan Park, on dead tree of *Liquidambar*, 20.XII.2000 *Dai 3252* (IFP, H). Sichuan Prov., Emei County, Emei Mts., dead angiosperm tree, 20.X.2002 *Dai 4317* (IFP). Zhejiang Prov., Baishazushan, 23.VII.1994 *Abe* (TNS).

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New species of marasmioid genera (Basidiomycetes, *Tricholomataceae*) from tropical Africa III. *Marasmius* sect. *Sicci*

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Abstract– Twelve new tropical African taxa of *Marasmius* sect. *Sicci* (*M. confertus* var. *tenuicystidiatus*, *M. confertus* var. *parvisporus*, *M. ferruginacies*, *M. ferruginoides*, *M. longistipitatus*, *M. pseudotorquescens*, *M. rammelooi*, *M. robertsii*, *M. rubro-stipitatus*, *M. striaepileus*, *M. tanougouensis*, *M. xestocephaloides*) are described and briefly discussed.

Key words– *Agaricales*, new taxa, taxonomy, nomenclature

Introduction

This paper represents the third part of a series of descriptions of new taxa of marasmioid genera from the tropical Africa (Antonín 2003a, b). All of the taxa described here belong to the richest and most complicated section, sect. *Sicci*. The complete study of these genera will be published in a series of monographs titled „Flore illustrée des champignons d´Afrique centrale" published by the National Botanical Garden in Meise (Belgium) in 2004. For material and methods see Antonín (2003a).

Species descriptions

Marasmius confertus Berk. & Broome var. *tenuicystidiatus* Antonín var. nov. Figure 1

A varietate typica pileo solum marginem leviter striato, pleurocystidiis raris, angustis (25–45(–55) x 7,0–10 µm) et projectionibus crassitunicatis differt.
Holotypus: Cameroon, Provincia Sud, Somalomo, area biosphaerica protecta Dja, 8. IV. 2001 leg. V. Antonín Cm 01.41 (holotypus in herbario BRNM 666109 asservatur).

Macroscopically almost identical with the type variety, but differing by having an only slightly at margin striate pileus, scattered, shorter and narrower pleurocystidia (25–45(–55) x 7.0–10 μm) and by the absence of distinctly thick-walled pileipellis broom cells with less numerous and longer projections.

Ecology – Saprophytic, growing on dead leaves.

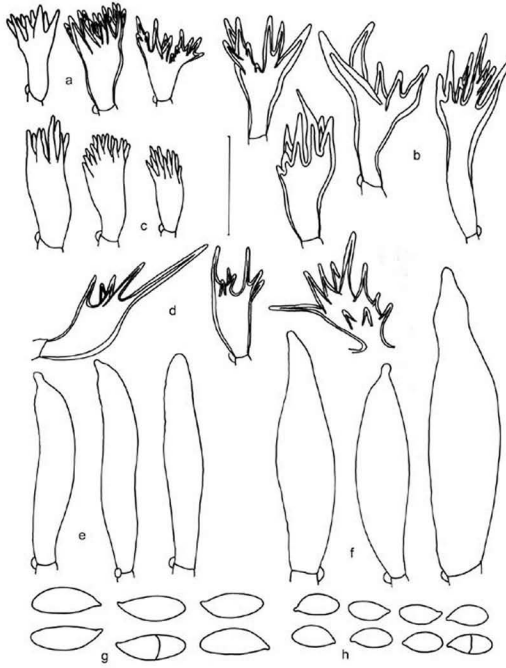

Figure 1. *Marasmius confertus*: a. and b. pileipellis cells, c. cheilocystidia, d. caulocystidia, e. pleurocystidia of var. *tenuicystidiatus* (holotype), f. pleurocystidia of var. *confertus*, g. basidiospores of var. *confertus*, h. basidiospores of var. *parvisporus* (holotype). Scale bar = 20 μm.

Distribution – So far known only from Burundi, Cameroon and the Democratic Republic of Congo.

REVISED SPECIMENS – BURUNDI: Bururi Province, Kigwena, Forêt de Kigwena, 22 Feb. 1979 leg. J. Rammeloo 6722 (BR 11951-20). – **CAMEROON:** Sud Province, Somalomo, Dja Biosphere Reserve, 8 Apr. 2001 leg. V. Antonín Cm 01.41 (holotype, BRNM 666109). – Ditto, 12 Apr. 2001 leg. V. Antonín Cm 01.103 (BRNM 666151). – **DEMOCRATIC REPUBLIC OF CONGO:** Tshopo Province, Kisangani, 2 May 1984 leg. B. Buyck 1614 (BR 11732-92). – Katanga Province, Plateau de Biano, 29 July 1986 leg. J. Rammeloo 8779 (BR 1255-91). – Katanga Province, Plateau de Biano, 12 Jan. 1987 leg. J. Rammeloo 8781 (BR 1261-00).

Marasmius confertus Berk. & Broome var. *parvisporus* Antonín var. nov. Figure 1

A varietate typica basidiosporis brevibus (7,3–10 x 3,6–4,6 μm) differt.
HOLOTYPUS: Kenya, Provincia Central, Districtus Nairobi, Thika, cataracta Thika, 16. III. 1968 leg. D.N. Pegler K101 (holotypus in herbario K asservatur).

Differs from the type variety by having distinctly smaller, 7.3–10 x 3.6–4.6 μm large basidiospores (E = 1.7–2.3, Q = 2.0–2.1).

Ecology – Saprophytic, growing on dead fallen twigs.

Distribution – So far known from the Democratic Republic of Congo, Kenya and Uganda.

REVISED SPECIMENS – DEMOCRATIC REPUBLIC OF CONGO: Katanga Province, Kakombwe, 27 March 1986 leg. J. Schreurs 1525 (BR 8287-42). – **KENYA:** Central Province, Nairobi District, Thika, Thika Fall, 16 March 1968 leg. D.N. Pegler K101 (holotype, K, as *M. corrugatiformis*). – **UGANDA:** Buganda Province, Mengo District, Muwacota County, Mpanga Research Forest, 7 June 1968 leg. D.N. Pegler U1252 (K, as *M. corrugatiformis*).

Marasmius ferruginacies Antonín sp. nov. Figure 2

Pileo 30 mm lato, convexo, umbilicato et papillato, sulcato-striato, brunneo-rubro vel rubro-brunneo. Lamellis distatibus, L = 22, intervenosis, luteolo-albidis, acie ferrugineis. Stipite 75 x 0,75 mm, filiforme, glabro, apicem luteolo-albido, ad basim obscure brunneo. Basidiosporis 9,0–12 x 5,0–6,0 µm, ellipsoideis, fusiformis-ellipsoideis vel subamygdaliformibus, hyalinis. Basidiis tetrasporis. Cheilocystidiis e cellulis similibus cellulis typo Marasmii sicci, 17–22 x 8,0–10,0 µm, clavatis vel subcylindraceis, tenuitunicatis vel leviter crassitunicatis. Pileipellis hymeniformis, e cellulis similibus cellulis hymenidermatis Marasmii sicci, 15–29 x 6,0–15 µm, clavatis vel subcylindraceis, tenuitunicatis vel leviter crassitunicatis. Caulocystidiis absentibus. Hyphis fibulatis, in stipite et trama dextrinoideis. Ad ramulos putridos.

HOLOTYPUS: Cameroon, area biosphaerica protecta Dja, prope Somalomo, 9. IV. 2001 leg. V. Antonín Cm01.59 (holotypus in herbario BRNM 666127 asservatur).

Carpophores single. **Pileus** 30 mm broad, convex, with slightly depressed centre and small broad papilla in depression, crenulate at margin, sulcate-striate, slightly tomentose, entirely brownish red to reddish brown (9C–D7), slightly darker at centre. **Lamellae** distant, L = 22, l = 0–1, forming a pseudocollarium, intervenose, broad (up to 4 mm), yellowish white (3–4A2), with ferrugineous, pubescent edge. **Stipe** 75 x 0.75 mm, filiform, hollow, slightly broadened at apex, non-insititious, smooth, glabrous, yellowish

white (± concolorous with lamellae at apex), dark brown (8F8) towards base; rhizomorphs well-developed.

Basidiospores 9.0–12 x 5.0–6.0 μm, E = 1.3–2.2, Q = 1.9, ellipsoid, fusoid-ellipsoid to subamygdaliform, thin-walled, hyaline, nondextrinoid. **Basidia** 4-spored, clavate. **Basidioles** 15–32 x 4.0–9.0 μm, cylindrical, clavate or subfusoid. **Cheilocystidia** in the form of broom cells of the Siccus-type, 17–22 x 8.0–10 μm, clavate to subcylindrical, thin- to slightly thick-walled, nondextrinoid, similar to pileipellis cells. **Pleurocystidia** absent. **Trama hyphae** cylindrical to subinflated, ± thin-walled, hyaline, dextrinoid, up to 15 μm wide. **Pileipellis** a hymeniderm made up of broom cells of the Siccus-type, 15–29 x 6.0–15 μm, clavate or subcylindrical, thin- or less frequently slightly thick-walled below, slightly thick-walled above, nondextrinoid, with 7–15 short and wide, obtuse, thick-walled, up to 9.0 x 3.0 μm projections; thick-walled parts ochraceous yellow in KOH. **Stipitipellis** a cutis consisting of cylindrical, parallel, slightly thick-walled (up to 1.0 μm), nondextrinoid hyphae with ochraceous brown walls in KOH. **Caulocystidia** absent. **Clamp connections** present in all tissues.

Ecology – Saprophytic, growing on fallen twigs.

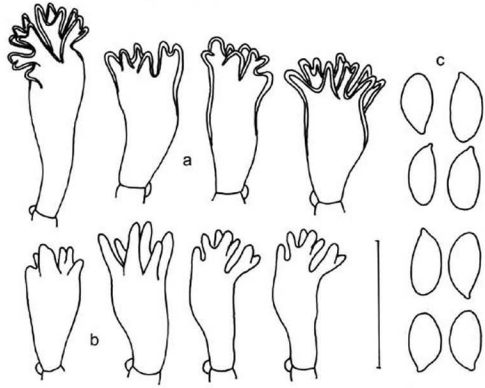

Figure 2. *Marasmius ferruginacies* (holotype): a. pileipellis cells, b. cheilocystidia, c. basidiospores. Scale bar = 20 μm.

REVISED SPECIMENS – CAMEROON: Dja Biosphere Reserve, c. 14 km ESE of Somalomo, 9 Apr. 2001 leg. V. Antonín Cm01.59 (holotype, BRNM 666127).

Comments – *Marasmius ferruginacies* is characterised by having a moderately large, ± uniformly brownish red to reddish brown coloured pileus, distant lamellae with coloured edge, a thin, dark brown stipe, small, ellipsoid, fusoid-ellipsoid to subamygdaliform basidiospores, and cheilocystidia and pileipellis broom cells with short and wide projections.

A very close species, both macro- and microscopically, is *M. nodulocystis* Pegler. It differs in having well-developed rhizomorphs, a ferrugineous lamellar edge, and slightly smaller and distinctly narrower basidiospores (7.5–10.5 x 3.5–4.4 μm). Compared with it, *Marasmius floriceps* Berk. & M.A. Curtis has a smaller pileus (10–17 mm), more distant lamellae (14–18), a shorter (25–35 mm), red-brown stipe and smaller basidiospores (7–10 x 3.5–4 μm) (Desjardin & Horak 1997). *Marasmius pusio* Berk. & M.A. Curtis is a small fungus with a non-striate, only 4–10 mm broad, ochre-, apricot- or orangish brown pileus, more distant lamellae (10–13), well-developed lamellulae, a small (7–18 x 0.5–0.8 mm), pale orangish brown stipe, smaller basidiospores (8–10.5 x 3.5–4.5 μm) and different (narrow and long) projections of pileipellis broom cells (Desjardin & Horak 1997, Singer 1976). *Marasmius corrugatus* (Pat.) Sacc. & P. Syd. has a reddish cinnamon to chestnut brown pileus, concolorous lamellar edges, a more robust stipe (25–35 x 1–2.5 mm), narrower basidiospores (9–13 x 2.7–4 μm) and narrow and acute projections of cheilocystidia and pileipellis broom cells (Pegler 1983); *M. floridens* Berk. & Broome has a smaller pileus (10–20 mm), a more robust (30–55 x 1–2 mm), reddish brown stipe, smaller basidiospores (8–10 x 3–3.5 μm), smaller basidia (14–18 x 4–5 μm) and smaller cheilocystidia and pileipellis broom cells with only 3–5 acute projections (Pegler 1986); *M. bezerrae* Singer has a smaller pileus (7–17 mm), a shorter stipe (35–50 x 0.6–1 mm), smaller basidiospores ((8–)9–11 x 3.2–5.4 μm) and spinulose projections of pileipellis broom cells (Singer (1976)).

***Marasmius ferruginoides* Antonín sp. nov.** **Figure 3**

Pileo 5–20 mm lato, campanulato vel late campanulato, leviter striato, obscure luteo, luteolo-aurantiaco vel aurantiaco. Lamellis, L = 18–21, liberis, luteolo-albidis. Stipite 30–65 x ± 1 mm, cylindraceo, glabro, apicem pallido, ad basim nigro-brunneo. Basidiosporis 11,5–14 x 3,8–5,2 µm, fusiformibus, clavato-fusiformibus, hyalinis. Basidiis tetrasporis. Cheilocystidiis e cellulis similibus cellulis typo Marasmii sicci, (10–)13–19 x 6,2–8,1 µm, clavatis vel subcylindraceis, tenuitunicatis. Pleurocystidiis 27–42 x 6,2–9,0 µm, subcylindraceis, subfusiformibus, rostratis, tenuitunicatis. Pileipellis hymeniformis, e cellulis similibus cellulis hymenidermatis Marasmii sicci, 8,5–14 x 3,8–7,7 µm, clavatis vel subcylindraceis, tenuitunicatis. Caulocystidiis absentibus. Hyphis fibulatis, in stipite et trama dextrinoideis. Ad folia putrida.

Holotypus: Democratic Republic of Congo, Provincia Tshopo, Kisangani, silva prope hortum animalium, 2. V. 1984 leg. B. Buyck 1615 (holotypus in herbario BR 11731-91 asservatur).

Misapplied name: *Marasmius gardneri* Singer s. Pegler, Kew Bull. Addit. Ser. 6: 194. 1977.

Pileus 5–20 mm broad, campanulate, then broadly campanulate with reflexed margin, smooth, finely tomentose, slightly striate almost to centre, deep yellow, yellowish orange (4A7–8, 4B7–8) or orange. **Lamellae** close,

L = 18–21, l = 2–3, free, with concolorous edge. **Stipe** 30–65 x ± 1 mm, cylindrical, laterally compressed, smooth, glabrous, lustrous, black-brown, with paler apex, with rusty brown, woolly basal mycelium.

Basidiospores 11.5–14 x 3.8–5.2 μm, E = 2.4–2.9, Q = 2.6, (sub)fusoid, clavate-fusoid, thin-walled, hyaline, nondextrinoid. **Basidia** 4-spored, clavate. **Basidioles** 10–29 x 4.5–8.5 μm, cylindrical, clavate, fusoid. **Cheilocystidia** in the form of broom cells of the Siccus-type, (10–)13–19 x 6.2–8.1 μm, clavate to subcylindrical, thin-walled, nondextrinoid, with obtuse to subacute, slightly thick-walled projections. **Pleurocystidia** 27–42 x 6.2–9.0 μm, subcylindrical, subfusoid, rostrate, thin-walled, nondextrinoid, with slightly refractive contents. **Trama hyphae** cylindrical to subinflated, thin- to slightly thick-walled, smooth to finely incrusted (subpileipellis), hyaline, dextrinoid, up to 18 μm wide. **Pileipellis** a hymeniderm made up of broom cells of the Siccus-type, 8.5–14 x 3.8–7.7 μm, clavate to subcylindrical, entirely thin-walled or with slightly thick-walled apex, nondextrinoid, with 15–30 digitate, obtuse to subacute, sometimes acute, slightly nodulose, up to 6.5 x 0.8(–1.5) μm projections; thick-walled parts pale yellow-brown in KOH. **Stipitipellis** a cutis consisting of cylindrical, parallel, slightly thick-walled, dextrinoid, up to 5.0 μm wide hyphae, with brownish yellow walls. **Caulocystidia** absent. **Clamp connections** present in all tissues.

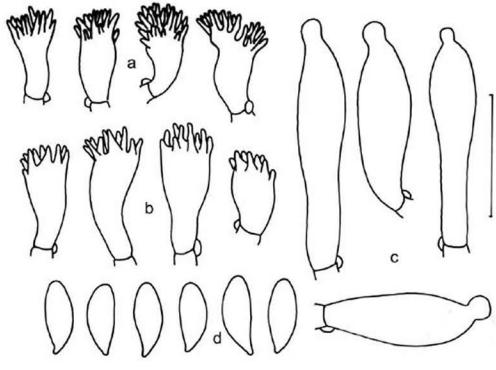

Figure 3. *Marasmius ferruginoides* (holotype): a. pileipellis cells, b. cheilocystidia, c. pleurocystidia, d. basidiospores. Scale bar = 20 μm.

Ecology – Saprophytic, growing on fallen leaves, rarely on strongly decayed wood.

Distribution – So far known from Cameroon, Democratic Republic of Congo, Ghana, Kenya and Nigeria.

REVISED SPECIMENS – CAMEROON: South West Province, Korup National Park, trail to Erat, 2 May 1996 leg. P.J. Roberts K360 (K(M) 39230, as *M. gardneri*). – Ditto, 25 Apr. 1996 leg. P.J. Roberts K51 (K(M) 39484, as *M. gardneri*). – **DEMOCRATIC REPUBLIC OF CONGO:** Tshopo Province, Kisangani, forest near ZOO, 2 May 1984 leg. B. Buyck 1615 (holotype, BR 11731-91). – **GHANA:** Cape Coast, University College, 12 June 1971 leg. A.C. Rose CC7131 (K, as *M. gardneri*). – **KENYA:** Central Province, Nairobi District, Nairobi, City Park, 12 March 1968 leg. D.N. Pegler K11 (K, as *M. gardneri*). – **NIGERIA:** Akwa Ibom State, Ekpere Obam, 12 May 1990 leg. R.A. Nicholson 438 (K(M) 16693, as *M. ferrugineus*). – Cross River State, Aking, Oban Forest, 19 June 1990 leg. R.A. Nicholson (K(M) 16707, as *M. ferrugineus*). – Cross River State, Anua Ravine, 4 June 1990 leg. R.A. Nicholson 486 (K(M) 16860, as *M. ferrugineus*). – Ditto, 13 May 1989 leg. R.A. Nicholson 199 (K(M) 7591, as *M. gardneri*). – Cross River State, Obudu Ranch, 24 Apr. 1989 leg. R.A. Nicholson 174 (K(M) 5388, as *M. gardneri*). – Cross River State, Uyo, Anua Ravine, 6 June 1990 leg. R.A. Nicholson 508 (K(M) 17070, as *M. sierraleonis*).

Comments – *Marasmius ferruginoides* is characterised by having a campanulate to broadly campanulate, deep yellow to (yellowish) orange pileus, close lamellae, a black-brown stipe, rather small, (sub)fusoid, clavate-fusoid basidiospores, well-developed narrow pleurocystidia and lacking caulocystidia.

This species was published from Africa as *Marasmius gardneri* Singer by Pegler (1977). However, the revision of the type specimen (Brazil, Minas Gerais, leg. Gardner, K(M) 92652) showed that the true *Marasmius ferrugineus* (Berk.) Berk. & M.A. Curtis (= *M. gardneri*) has a smaller, 2–10 mm broad, fulvous-ferrugineous, light orange to greyish orange pileus and distinctly longer basidiospores (18–22 x 4.0–5.5–6.0) μm).

Among African species, *Marasmius confertus* Berk. & Broome has a brown, orange or brownish orange pileus and larger pleurocystidia (25–60(–75) x 10–16(–20) μm).

Marasmius dennisii Singer has a distinctly larger, 15–50 mm broad, orange-chrome to ochraceous orange pileus, and larger basidiospores (12.5–17 x 3–4 μm and 14–17 x 3–3.5 μm, respectively, Dennis 1951, 1970, Pegler 1983, Singer 1976). *Marasmius nocturnus* Har. Takahashi has a light yellow, reddish yellow, light orange to orange, later brown pileus, smaller basidiospores (9–10.5 x 4–4.5 μm), very narrow pleurocystidia (25–45 x 4–6 μm) and large pileipellis broom cells (20–28.5 x 5–12 μm) (Takahashi 2000).

***Marasmius longistipitatus* Antonín sp. nov.** **Figure 4**

Pileo 1–10 mm lato, junio subhaemisphaerico, papillato, dein convexo, leviter umbonato et papillato, striato, centro pallide griseo-aurantiaco, marginem auratiaco-albido. Lamellis, L = 9–12, adnatis, albido-luteolis. Stipite 20–150 x 0,3–0,6 mm, cylindraceo, apicem pallide luteo, ad basim rubro-auratiaco vel brunneo-aurantiaco. Basidiosporis (17–)20–28 x 4,5–6,0 µm, clavatis, clavato-

fusiformibus, cylindrice-clavatis, hyalinis. Basidiis tetrasporis. Cheilocystidiis e cellulis similibus cellulis typo Marasmii sicci, 12–18 x 6,0–9,0 µm, clavatis vel subcylindraceis, tenuitunicatis. Pleurocystidiis 35–68 x 6,0–9,0 µm, cylindraceis, anguste clavatis, anguste fusiformibus, tenuitunicatis. Pileipellis hymeniformis, e cellulis similibus cellulis hymenodermatis Marasmii sicci, 11–18 x (4,0–)6,0–9,0 µm, clavatis vel subcylindraceis, leviter crassitunicatis vel ad basim tenuitunicatis. Caulocystidiis absentibus. Hyphis fibulatis, in stipite et trama dextrinoideis. Ad folia putrida.

HOLOTYPUS: Benin, Provincia Atacora, Tanougou, cataracta Tanougou, 8. IX. 1997 leg. V. Antonín B97.172 (holotypus in herbario BR 101210-39 asservatur).

Pileus 1–10 mm broad, almost hemispherical when young, then convex, with a pronounced conical papilla when young, with rugulose and slightly papillate centre in small central depression when old, translucently striate, with crenulate and slightly inflexed margin, sulcate, slightly tomentose, pale greyish orange (6B6) at centre, pallescent up to orange-white (5A2, sometimes even more whitish) towards margin. **Lamellae** distant, L = 9–12, l = 0–1, adnate to a pseudocollarium, rather thick, not intervenose, white-yellowish (2-3A2), then pale yellow (up to 4A3), with concolorous, entire, finely pubescent edge. **Stipe** very long and thin, 20–150 x 0.3–0.6 mm, cylindrical, slightly broadened at apex or not, smooth, slightly pruinose at apex, lustrous, often curved, except for cream or pale yellow (2–3A2 to 4A3) apex, entirely reddish orange to brownish orange (through 7B–C7) towards base; forming small abortive pilei on a long stipe.

Basidiospores (17–)20–28 x 4.5–6.0 µm, E = 4.0–5.4, Q = 4.5, clavate, fusoid-clavate, cylindrical-clavate, hyaline, nondextrinoid, thin-walled. **Basidia** 35 x 12 µm (only one found), 4-spored, (broadly) clavate. **Basidioles** 16–35 x 4.0–13 µm, (broadly) clavate, subfusoid, cylindrical. **Cheilocystidia** in the form of broom cells of the Siccus-type, 12–18 x 6.0–9.0 µm, clavate to subcylindrical, thin-walled, hyaline, nondextrinoid, with nodulose, obtuse to subacute, slightly thick-walled, up to 5.0 x 1.0 µm large projections. **Pleurocystidia** 35–68 x 6.0–9.0 µm, cylindrical, narrowly clavate, narrowly fusoid, often moniliform, often subcapitate, often branched above, thin-walled, nondextrinoid, with refractive contents. **Trama hyphae** cylindrical, thin- to slightly thick-walled, smooth or minutely incrusted, hyaline, dextrinoid, up to 8.0 µm wide. **Pileipellis** a hymeniderm made up of broom cells of the Siccus-type, 11–18 x (4.0-)6.0–9.0 µm, (sub)cylindrical to clavate, entirely slightly thick-walled or with thin-walled base, with 8–25(–30) nodulose, ± digitate, subacute to acute, slightly thick-walled, up to 5.0 x 1.0 µm large projections; thick-walled parts ochraceous yellow in KOH. **Stipitipellis** a cutis consisting of cylindrical, parallel, slightly thick-walled, dextrinoid, up to 6.0 µm wide hyphae, with ochraceous walls in KOH. **Caulocystidia** absent. **Clamp connections** present in all tissues.

Ecology – Saprophytic on dead leaves, mostly on or near their veins.

Distribution – So far known only from the type locality in Benin.

REVISED SPECIMENS: BENIN: Atacora Province, Tanougou, Chutes de Tanougou, 8 Sept. 1997 leg. V. Antonín B97.172 (holotype, BR 101210-39).

Comments – *Marasmius longistipitatus* is a very distinct species characterised by having a small, papillate, pale greyish orange to orange-white pileus, distant lamellae, a very long and thin, reddish orange to brownish orange stipe, very long basidiospores, long and narrow pleurocystidia, and lacking caulocystidia.

The combination of a very long stipe with very large basidiospores is rather rare among *Marasmius* species. Only *Marasmius megistosporus* Singer has similar features. However, it has a larger, 35 mm broad, deep brown pileus at centre which turns through deep ferrugineous to gold brown towards margin, and even larger basidiospores (28–37.5 x 3.5–5.5 μm) (Singer 1976).

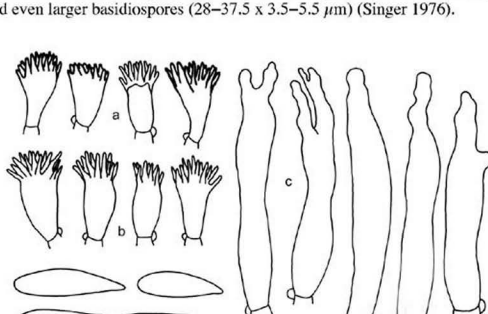

Figure 4. *Marasmius longistipitatus* (holotype): a. pileipellis cells, b. cheilocystidia, c. pleurocystidia, d. basidiospores. Scale bar = 20 μm.

Marasmius pseudotorquescens Antonín sp. nov. **Figure 5**

Pileo 17–29 mm lato, convexo, subumbonato, leviter striato, luteolo-brunneo, centro obscure brunneo. Lamellis, L = 18–20, liberis, luteolo-albidis. Stipite 60–70 x 1 mm, cylindraceo, subtiliter pruinoso vel pubescente, apicem griseo-aurantiaco, ad basim obscure brunneo. Basidiosporis (14–) 15–16(–17) x (4,0–)4,5–5,5(–6,0) μm, fusiformibus vel clavatis, hyalinis. Basidiis tetrasporis. Cheilocystidiis 25–55 x 8,0–12 μm, fusiformibus, (sub)cylindraceis, lageniformibus, rostratis, tenuitunicatis. Pleurocystidiis 41–82 x 12–18 μm, lageniformibus, fusiformibus, cylindraceis, clavatis, subutriformibus, rostratis, tenuitunicatis. Pileipellis hymeniformis, e cellulis similibus cellulis hymenidermatis Marasmii sicci, 8,0–12 x 6,5–9,0 μm, clavatis vel subcylindraceis, crassitunicatis vel ad basim tenuitunicatis. Pileoseris 32–70 x 6,0–8,0 μm, subuliformibus, lageniformibus, obtusis vel acutis, crassitunicatis, dextrinoideis. Caulocystidiis e cellulis similibus cellulis hymenodermatis Marasmii sicci et seriformibus, 35–60 x 7,0–12 μm, subuliformibus, fusiformibus, lageniformibus, subcylindraceis crassitunicatis. Hyphis fibulatis,

in stipite et trama dextrinoidels. Ad ramulos putridos.

Holotypus: Democratic Republic of Congo, Irangi, Kivu, 17. IV. 1972 leg. J. Rammeloo JR Z316 (holotypus in herbario GENT asservatur).

Pileus 17–29 mm broad, convex, with small umbo, slightly radially striate, finely velvety, yellowish brown (5D5) with dark brown (7E–F6) centre. **Lamellae** rather close, L = 18–20, with lamellulae, free, not intervenose, yellowish white (4A2), with concolorous edge. **Stipe** 60–70 x 1 mm, cylindrical, finely pruinose to finely pubescent (lens), hollow, greyish orange (5B4) at apex, dark brown (7F5) towards base. **Context** without any distinct smell or taste.

Basidiospores (14–)15–16(–17) x (4.0–)4.5–5.5–6.0) μm, E = 2.5–3.7, Q = 2.9–3.1, fusoid to clavate, thin-walled, hyaline, nondextrinoid. **Basidia** 4-spored, clavate. **Basidioles** up to 30 x 9.0 μm, clavate, cylindrical, fusoid. **Cheilocystidia** 25–55 x 8.0–12 μm, versiform, fusoid, (sub)cylindrical, lageniform, often rostrate, often moniliform, thin-walled, never in the form of broom cells, nondextrinoid. **Pleurocystidia** 41–82 x 12–18 μm, versiform, lageniform, fusoid, cylindrical, clavate, subutriform, often rostrate, rostrum often moniliform, thin-walled, hyaline, nondextrinoid, with refractive contents. **Trama hyphae** cylindrical, subfusoid, ± thin-walled, hyaline, dextrinoid, up to 20 μm wide. **Pileipellis** a hymeniderm made up of broom cells of the Siccus-type, 8.0–12 x 6.5–9.0 μm, subcylindrical to clavate, entirely thick-walled or thin-walled at base, with 6–20(–25) nodulose, digitate or conical, obtuse to subacute, up to 10 x 1.5 μm large projections. **Pileosetae** 32–70 x 6.0–8.0 μm, subulate, lageniform, obtuse to subacute, thick-walled, dextrinoid. **Stipitipellis** a cutis made up of cylindrical, parallel, slightly thick-walled, dextrinoid, up to 5.0 μm wide hyphae with ochraceous-olivaceous walls in KOH. **Caulocystidia** in the form of setoid broom cells or setae, 35–60 x 7.0–12 μm, awl-form, fusoid, lageniform, subcylindrical, thick-walled (up to 1.5 μm) at least in upper part, concolorous with stipitipellis hyphae, nondextrinoid. **Clamp connections** present in all tissues.

Ecology – Saprophytic, growing on dead twigs.

Distribution – Known only from the Democratic Republic of Congo.

REVISED SPECIMENS – DEMOCRATIC REPUBLIC OF CONGO: Irangi, Kivu, 17 Apr. 1972 leg. J. Rammeloo JR Z316 (holotype, GENT). – Kisantu, 1970, leg. H. Vanderyst s.n. (BR A768, as *M. castaneovelutinus*).

Comments – *Marasmius pseudotorquescens* is macroscopically similar to the European *M. torquescens* Quél., and is characterised by having a yellowish brown pileus with dark brown centre, fusoid, (sub)cylindrical, lageniform cheilocystidia (not in the form of broom cells), large, refractive pleurocystidia with a similar shape as the cheilocystidia and by the presence of pileo- and caulosetae.

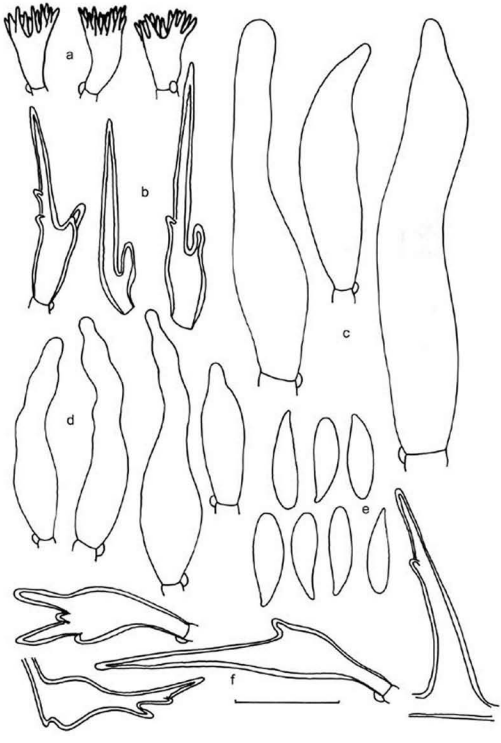

Figure 5. *Marasmius pseudotorquescens* (holotype): a. pileipellis cells, b. pileosetae, c. pleurocystidia, d. cheilocystidia, e. basidiospores, f. caulocystidia. Scale bar = 20 μm.

The presence of fusoid, (sub)cylindrical, lageniform, often rostrate cheilocystidia represents an unique character in sect. *Sicci*. Only *Marasmius mengoënsis* Pegler has similar cheilocystidia. However, it has a small (5–8 mm broad), reddish brown pileus, white lamellae, a smaller stipe (25–35 x 0.3–0.7 mm), distinctly smaller basidiospores (7.5–10 x 4–5 μm), smaller pleurocystidia (25–45 x 8–11 μm), and no pileosetae.

***Marasmius rammelooi* Antonín sp. nov.** **Figure 6**

Pileo 4 mm lato, convexo, sulcato, pallide aurantiaco-ochraceo, centro obscuriore. Lamellis, L = cca 12, acie irregulariter brunneis. Stipite 3–4 mm longo, cylindraceo, pruinoso, junio albido, dein brunneo. Basidiosporis 13,5–16,9 x 3,8–4,6 µm, clavatis vel clavato-lacrimoideis, hyalinis. Basidiis tetrasporis. Cheilocystidiis 20–37 x 5,0–6,9 µm, cylindraceis vel clavatis, irregularibus vel coralloideis, crassitunicatis. Pileipellis hymeniformis, e cellulis similibus cellulis hymenidermatis Marasmii sicci, 10,8–21 x 7,7–11,5(–17) µm, clavatis, tenuitunicatis vel apicem levier crassitunicatis. Caulocystidiis e cellulis similibus cellulis hymenodermatis Marasmii sicci, 7,7–14 x 5,4–6,6 µm, tenuitunicatis. Hyphis fibulatis, in stipite et trama dextrinoideis. Ad ramulos putridos.

Holotypus: Mauritius, 10. VI. 1990 leg. J. Rammeloo 9251 (holotype, BR 6902-15).

Pileus 4 mm broad, regularly convex, membranaceous, sulcate, finely tomentose, pale orangish ochraceous (paler than 7.5YR 8/4), slightly darker at centre. **Lamellae** distant, L = c. 12, l = 0, adnate, narrow, with irregularly dark brown edge. **Stipe** 3–4 mm long, cylindrical, finely pruinose, ± white when young, then brown, non-insititious.

Basidiospores 13.5–16.9 x 3.8–4.6 μm, E = 3.2–4.5, Q = 3.8, clavate, clavate-lacrimoid, thin-walled, hyaline, nondextrinoid. **Basidia** 23–30 x 6.9–9.6 μm, 4-spored, clavate. **Basidioles** 15–29 x 2.5–8.0 μm, cylindrical, clavate. **Cheilocystidia** scattered among basidioles, 20–37 x 5.0–6.9 μm, cylindrical to clavate, irregular to subcoralloid, thin-walled, nondextrinoid, hyaline. **Pleurocystidia** absent. **Trama hyphae** cylindrical, thin-walled, hyaline, dextrinoid, up to 8.0 μm wide. **Pileipellis** a hymeniderm made up of broom cells of the Siccus-type, 10.8–21 x 7.7–11.5(–17) μm, clavate, entirely thin-walled or slightly thick-walled at apex. **Stipitipellis** a cutis consisting of cylindrical, parallel, thick-walled (up to 1.7 μm), nondextrinoid, up to 7.5 μm wide hyphae. **Caulocystidia** 7.7–14 x 5.4–6.6 μm, in the form of clavate to subcylindrical, thin-walled, hyaline broom cells, with subacute to acute, nodulose, thin-walled, up to 7.0 x 1.5 μm large projections, with transitional forms to simple, ± cylindrical, irregular to subcoralloid, thin-walled cells. **Clamp connections** present in all tissues.

Ecology – Saprophytic, growing on a dead twig.

Distribution – So far known only from the type locality in Mauritius.

REVISED SPECIMENS – MAURITIUS: 10 June 1990 leg. J. Rammeloo 9251 (holotype, BR 6902-15).

Comments – *Marasmius rammelooi* is characterised by having small carpophores with a very pale coloured pileus, distant lamellae with coloured edges, a short stipe, moderately large basidiospores, scattered, irregular to subcoralloid cheilocystidia which are cylindrical to clavate (not in the form of broom cells), a pileipellis consisting of broom cells of the Siccus-type, well-developed caulocystidia in the form of broom cells and lacking pleurocystidia.

A very close species seems to be *M. luteomarginatus* Desjardin, Retnowati & E. Horak. It has slightly larger carpophores (pileus 5–17 mm broad) with an only at margin striate, brightly orange, then orangish white to white pileus, less numerous lamellae (7–9) with brightly yellow edges and an orangish brown stipe at apex (Desjardin & al. 2000). *Marasmius tenuissimus* (Jungh.) Singer has a distinctly larger, 15–40 mm broad, reniform to suborbicular, buff to ochraceous, pale to grey-brown pileus, more distant lamellae (L = 3–6), an absent or only very short eccentric stipe which is concolorous with the pileus, distinctly smaller basidiospores (6.5–10.5 x 4–5 μm), and cheilocystidia in the form of ± poorly developed broom cells (Pegler 1986).

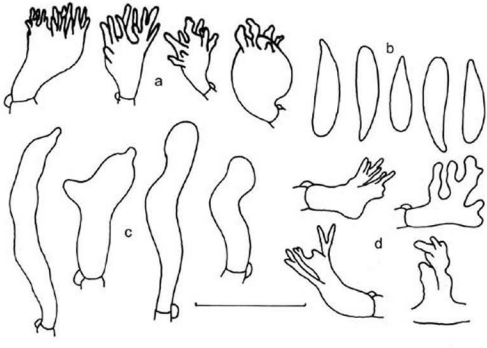

Figure 6. *Marasmius rammelooi* (holotype): a. pileipellis cells, b. basidiospores, c. cheilocystidia, d. caulocystidia. Scale bar = 20 μm.

Marasmius robertsii Antonín sp. nov. **Figure 7**

Pileo usque 5 mm lato, albido. Lamellis sparsis, L = 8–9, non-collariatis. Stipite c. 3–4 mm longo, tenui, curvato, albido. Basidiosporis 11–15(–16) x 3,5–5,5 µm, fusiformibus, hyalinis. Basidiis tetrasporis. Cheilocystidiis e cellulis similibus cellulis typo Marasmii sicci vel Marasmii rotalis transientibus, 15–25 x 5,5–7,0 µm, clavatis, tenuitunicatis. Pleurocystidiis 26–35 x 9,0–11 µm, ± fusiformibus, clavatis, rostratis, tenuitunicatis. Pileipellis hymeniformis, e cellulis similibus cellulis hymenodermatis Marasmii sicci vel Marasmii rotalis transientibus, 12–25 x 7,0–12 µm, clavatis, subcylindraceis, tenuitunicatis. Caulocystidiis absentibus. Mycelio basale cum cellulis similibus cellulis hymenodermatis Marasmii sicci, dextrinoideis. Hyphis fibulatis, in stipite et trama dextrinoideis. Ad detritum.

Holotypus: Cameroon, Provincia South West, area protecta Korup, 26. IV. 1996 leg. P.J. Roberts K129 (holotypus in herbario K(M) 42926 asservatur).

Macroscopic description not available, only collector´s notes completed with notes about herbarium specimen. **Pileus** small to minute, up to 5 mm broad, white. **Lamellae** very few, L = 8–9, l = 0–1, sometimes branched, not collariate. **Stipe** c. 3–4 mm long, thin, curved, central, non-insititious, white.

Basidiospores 11–15(–16) x 3.5–5.5 µm, E = 2.6–3.4, Q = 3.0, fusoid, thin-walled, hyaline, nondextrinoid. **Basidia** not found. **Basidioles** 12–23 x up to 10 µm, clavate, fusoid. **Cheilocystidia** in the form of broom cells of the Siccus-type with transient forms to the Rotalis-type, 15–25 x 5.5–7.0 µm, clavate, thin-walled, nondextrinoid. **Pleurocystidia** 26–35 x 9.0–11 µm, ± fusoid, clavate, rostrate, obtuse, thin-walled, nondextrinoid, with slightly refractive contents. **Trama hyphae** cylindrical to subinflated, thin-walled, dextrinoid, smooth or minutely incrusted, up to 14 µm wide. **Pileipellis** a hymeniderm made up of broom cells of the Siccus-type with transient forms to the Rotalis-type, 12–25 x 7.0–12 µm, clavate, subcylindrical, thin-walled, with ± fine, digitate, obtuse to subacute, nodulose, thin- to slightly thick-walled, up to 3.0 x 1.0 µm large projections. **Stipitipellis** a cutis consisting of cylindrical, parallel, thick-walled, dextrinoid, up to 5.0 µm wide hyphae. **Caulocystidia** absent; at base with long, narrow (up to 4.0 µm), thick-walled, acute to subacute, dextrinoid hairs. **Basal mycelium** with minute, slightly thick-walled, dextrinoid broom cells with long projections ("*Amyloflagellula*-type"). **Clamp connections** present in all tissues.

Ecology – Saprophytic, growing on trapped aerial litter and on a liana.

Distribution – So far known only from Cameroon.

Revised specimens: Cameroon: South West Province, Korup National Park, 26 Apr. 1996 leg. P.J. Roberts K129 (holotype, K(M) 42926). – Ditto, Mundemba, 8 Apr. 1990 leg. R. Watling (E).

Comments – *Marasmius robertsii* is characterised by having very small carpophores with an up to 5 mm broad, white pileus, very few, sometimes branched, not collariate lamellae, a thin, curved, central, non-insititious, white stipe, moderately large, fusoid basidiospores, cheilocystidia in the

form of broom cells of the Siccus-type with transient forms to the Rotalis-type, fusoid or clavate, rostrate pleurocystidia, a pileipellis made up of broom cells of the Siccus-type with transient forms to the Rotalis-type with ± fine, digitate and short projections; caulocystidia are lacking and it has minute, slightly thick-walled broom cells with long projections (of the *Amyloflagellula*-type) in the basal mycelium.

This species is apparently similar to *Marasmius pseudoarachnoideus* Dennis (= *Amyloflagellula pseudoarachnoidea* (Dennis) Singer) (Dennis 1951, 1970), which has less numerous lamellae (4–6), larger basidiospores (18–19 x 4 μm) and longer projections of the pileipellis broom cells. *Marasmius robertsii*, having well-developed dextrinoid broom cells with long projections on its basal mycelium, resembles an *Amyloflagellula* species. However, it possesses cheilocystidia in the form of broom cells and flagelliform projections of pileipellis cells are not developed. Therefore, it is described within the genus *Marasmius*.

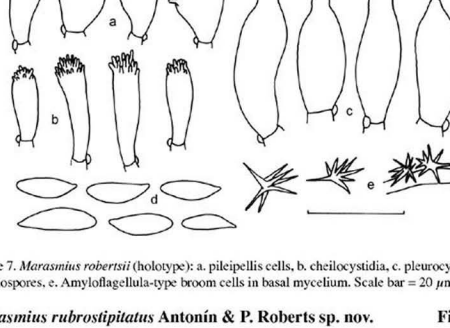

Figure 7. *Marasmius robertsii* (holotype): a. pileipellis cells, b. cheilocystidia, c. pleurocystidia, d. basidiospores, e. Amyloflagellula-type broom cells in basal mycelium. Scale bar = 20 μm.

Marasmius rubrostipitatus Antonín & P. Roberts sp. nov. Figure 8

Pileo 6–15 mm lato, conico vel plano-conico, leviter papillato, subtiliter striato, griseo-brunneo. Lamellis, L = 16–18, angustis, pallide luteis vel luteolo-rubris, acie brunneo maculatis. Stipite 25–40 x usque 0,5 mm, cylindraceo, glabro, apicem pallide luteo, ad basim aurantiaco vel obscure rubro. Basidiosporis 8,2–10,5 x 3,7–4,8 µm, ellipsoideo-fusiformibus vel fusiformibus, hyalinis. Basidiis tetrasporis. Cheilocystidiis e cellulis similibus cellulis typo Marasmii sicci, 9,0–15 x 4,0–8,0 µm, cylindraceis vel subclavatis. Pleurocystidiis 26–45 x 7,7–11 µm, fusiformibus, subcylindraceis, tenuitunicatis. Pileipellis hymeniformis, e cellulis similibus cellulis hymenidermatis Marasmii sicci, 9,0–16 x 6,0–8,5 µm, clavatis, pyriformibus vel subcylindraceis, tenuitunicatis, ad basim tenuitunicatis. Caulocystidiis absentibus. Hyphis fibulatis, in stipite et trama dextrinoideis. Ad folia putrida.

Holotypus: Cameroon, Provincia South West, area protecta Korup, 4. V. 1996 leg. P. J. Roberts K493 (holotypus in herbario K(M) 39594 asservatur).

Pileus c. 6–15 mm broad, conical to plano-conical, sometimes slightly papillate, slightly striate, matt, grey-brown. **Lamellae** moderately close, L = 16–18, l = 2–3, ± narrow, cream to pale buff, with brown spotted edge. **Stipe** 25–40 x up to 0.5 mm, cylindrical, smooth, glabrous, lustrous, cream at apex, orange to dark red towards base.

Basidiospores 8.2–10.5 x 3.7–4.8 µm, E = 1.9–2.6, Q = 2.2, ellipsoid-fusoid or fusoid, thin-walled, hyaline, nondextrinoid. **Basidia** 24 x 7.5 µm (only one found), 4-spored, clavate. **Basidioles** 14–26 x 4.5–7.5 µm, fusoid, clavate, cylindrical. **Cheilocystidia** in the form of broom cells of the Siccus-type, 9.0–15 x 4.0–8.0 µm, cylindrical to subclavate, thin-walled, nondextrinoid, with thin-walled, nodulose, obtuse to subacute, up to 7.5 x 1.2 µm projections. **Pleurocystidia** 26–45 x 7.7–11 µm, fusoid, subcylindrical, often with an obtuse rostrum, often originating in the subhymenium, thin-walled, nondextrinoid, with refractive contents. **Trama hyphae** cylindrical to subinflated, thin-walled, hyaline, dextrinoid, up to 12 µm wide. **Pileipellis** a hymeniderm made up of broom cells of the Siccus-type, 9.0–16 x 6.0–8.5 µm, clavate, pyriform or subcylindrical, thick-walled with ± thin-walled base or entirely slightly thick-walled, with 10–25 nodulose, obtuse to subacute, slightly thick-walled, up to 10 x 1.2 µm projections; thick-walled parts ochraceous brown in KOH. **Stipitipellis** a cutis consisting of cylindrical, parallel, slightly thick-walled, dextrinoid, up to 6.0 µm wide hyphae, with ochraceous (reddish) brown walls in KOH. **Caulocystidia** absent. **Clamp connections** present in all tissues.

Ecology – Saprophytic, growing on fallen leaves.

Distribution – So far known only from the type locality in Cameroon.

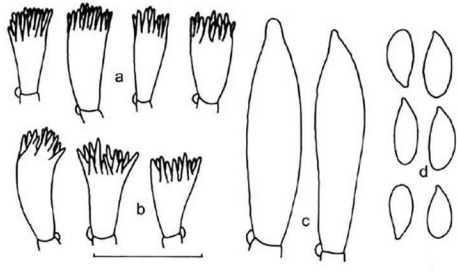

Figure 8. *Marasmius rubrostipitatus* (holotype): a. pileipellis cells, b. cheilocystidia, c. pleurocystidia, d. basidiospores. Scale bar = 20 µm.

Revised specimens: Cameroon: South West Province, Korup National Park, trail from Rengo Camp to Ekunde-Kunde, 4 May 1996 leg. P. J. Roberts K493 (holotype, K(M) 39594, as *M. spegazzinii*).

Comments – *Marasmius rubrostipitatus* is characterised by having a conical to plano-conical, grey-brown pileus, close lamellae, an orange to dark red stipe at base, rather small basidiospores, rather short pleurocystidia, rather small pileipellis broom cells and cheilocystidia; caulocystidia are lacking.

A very close species seems to be *M. spegazzinii* Sacc. & P. Syd., which especially differs in having an orange-ferrugineous, at margin paler pileus, an umbrinous to black stipe, narrower basidiospores (8.0–10 x (2.5–)3.0–3.5 (–4.0) μm), larger cheilocystidia (15–22 x 7.5–9.0 μm), broader pleuro-cystidia (35–55 x 10–21 μm) and a pileipellis of two types of broom cells.

Among other small-spored *Marasmius* species, *Marasmius nocturnus* Har. Takahashi has a light yellow, reddish yellow, light orange to orange, later brown pileus, very narrow pleurocystidia (25–45 x 4–6 μm) and large pileipellis broom cells (20–28.5 x 5–12 μm) (Takahashi 2000); *M. hylaeae* Singer has a rusty spadiceous pileus with lighter brown margin and smaller pleurocystidia (18–35 x 5.5–7 μm); *M. pseudocorrugatus* Singer has a larger (20–62 mm), dull cinnamomeous, sometimes olivaceous tinged pileus; *M. aztecus* Singer has a brightly ochraceous brown to rusty brown or orange pileus, a longer stipe (40–106 x (0.8–)1–2.5 mm) and larger basidiospores ((7.7–)8–13 x (2.5–)3–4 μm) (Singer 1976); moreover, none of them has a similar stipe colour.

***Marasmius striaepileus* Antonín sp. nov.** **Figure 9**

Pileo 15–30 mm lato, campanulato vel applanato, aurantiaco-brunneo, centro brunneo, pallide striato. Lamellis, L = 17–20, adnatis, albidis. Stipite 40–60 x 1 mm, cylindraceo, glabro, junio pallide, dein obscure brunneo. Basidiosporis 15,5–18,5(–19,2) x 3,5–5,0 µm, clavatis, anguste lacrimoideis vel fusiformibus, hyalinis. Cheilocystidiis e cellulis similibus cellulis typo Marasmii sicci, 10,8–16,2 x 5,4–8,5 µm, clavatis vel subcylindraceis, tenuitunicatis. Pileipellis hymeniformis, e cellulis similibus cellulis hymenidermatis Marasmii sicci, 10–15,5 x 6,6–9,2 µm, clavatis vel cylindraceis, tenuitunicatis, apicem subtiliter crassitunicatis. Caulocystidiis absentibus. Hyphis fibulatis, in stipite et trama dextrinoideis. In ligno putrido et ad corticem arboris.

Holotypus: Burundi, Provincia Bururi, Kigwena, silva Kigwena, 22. II. 1979 leg. J. Rammeloo 6724 (holotypus in herbario BR 11953-22 asservatur).

Pileus 15–30 mm broad, campanulate to applanate, membranaceous, apparently glabrous, slightly mealy under lens, sulcate ± up to centre, orange-brown (6C8), darker, brown (6D5) at centre, with paler stripes. **Lamellae** distant, L = 17–20, l = 0–1(–2), adnate, moderately large (2.5–3 mm), thin, white, then slightly darker, with concolorous, finely pubescent edge. **Stipe** 40–60 x 1 mm, cylindrical, smooth, glabrous, hollow, pale when young, then dark brown. **Smell** indistinct. **Taste** fungoid.

Basidiospores 15.5–18.5(–19.2) x 3.5–5.0 μm, E = 3.4–4.6, Q = 4.0, clavate to narrowly lacrimoid or fusoid, thin-walled, hyaline, nondextrinoid. **Basidia** not found. **Basidioles** 11.5–26 x 4.5–7.7 μm, cylindrical, clavate, fusoid.

Cheilocystidia in the form of broom cells of the Siccus-type, 10.8–16.2 x 5.4–8.5 μm, clavate to subcylindrical, entirely thin-, rarely slightly thick-walled below, slightly to distinctly thick-walled above, with nodulose, digitate, up to 12 μm long projections. **Pleurocystidia** absent. **Trama hyphae** cylindrical to subinflated, thin- to slightly thick-walled, dextrinoid, up to 12 μm wide. **Pileipellis** a hymeniderm made up of broom cells of the Siccus-type, 10–15.5 x 6.6–9.2 μm, clavate or cylindrical, thin-walled with slightly thick-walled apex, with 3–15 conical to subulate, mostly subacute, thick-walled, up to 20 x 2.1 μm projections; thick-walled parts ochraceous in KOH. **Stipitipellis** a cutis consisting of cylindrical, parallel, slightly thick-walled, dextrinoid, up to 6.0 μm wide hyphae with ochraceous yellowish walls in KOH. **Caulocystidia** absent. **Clamp connections** present in all tissues.

Ecology – Saprophytic, growing on dead and decaying wood as well as on bark of a living tree.

Distribution – Known only from the type locality in Burundi.

REVISED SPECIMENS: BURUNDI: Bururi Province, Kigwena, Kigwena Forest, 22 Feb. 1979 leg. J. Rammeloo 6724 (holotype, BR 11953-22).

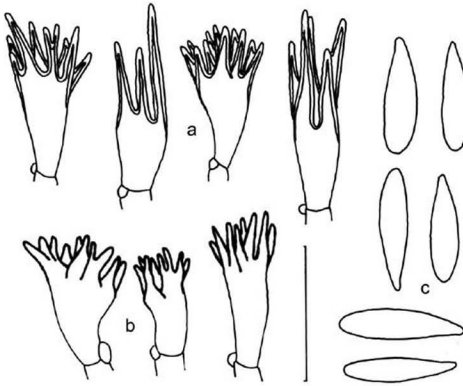

Figure 9. *Marasmius striaepileus* (holotype): a. pileipellis cells, b. cheilocystidia, c. basidiospores. Scale bar = 20 μm.

Comments – *Marasmius striaepileus* is characterised by having a rather large, striped, orange-brown to brown pileus, distant lamellae, a dark brown stipe, moderately large basidiospores and large projections of cheilocystidia and pileipellis broom cells. It lacks pleuro- and caulocystidia.

Marasmius corrugatus (Pat.) Sacc. & P. Syd. has an only at margin striate, otherwise rugulose pileus, a shorter stipe (20–35 x 1–2.5 mm) and smaller basidiospores (9–13 x 2.3–4 μm) (Pegler 1983); *M. trinitatis* Dennis differs especially in having only 9–11.5 x 3.5–4 μm (Singer 1976: 8.3–12.5 x 2.7–4 μm) basidiospores (Dennis 1951, Desjardin & Horak 1997); *M. fulviceps* Berk. has a smaller (10–15 mm), non-striate pileus, smaller basidiospores (10–12.5 x 4–5 μm) and smaller cheilocystidia (7–9 x 3–4 μm) and pileipellis broom cells (7–10 x 4–7 μm) (Pegler 1986).

***Marasmius tanougouensis* Antonín sp. nov.** **Figure 10**

Pileo 9–20 mm lato, late conico vel convexo, junio papillato, dein umbilicato cum vel sine papilla, striato-sulcato, centro rugulosо, obscure brunneo-aurantiaco, dein pallide griseo- vel aurantiaco-brunneo, marginem aurantiaco-ochraceo. Lamellis, L = (8–)10–16, adnatis, pallide ochraceo-luteis. Stipite 11–33 x 0,2–0,8 mm, cylindraceo, glabro, junio apicem subtiliter pruinoso, albido, dein pallide ochraceo vel luteo, ad basim aurantiaco-brunneo vel obscure brunneo. Basidiosporis (11,5–) 12,5–15,5 x 3,5–5,0(–5,5) µm, clavatis vel fusiformibus, hyalinis. Cheilocystidiis e cellulis similibus cellulis typo Marasmii sicci, (10–)12–20(–24) x 4,5–10 µm, anguste clavatis vel subcylindraceis, tenuiparietalibus. Pileipellis hymeniformis, e cellulis similibus cellulis hymenidermais Marasmii sicci, 11–20(–25) x 6,0–11 µm, clavatis vel cylindraceis, tenuitunicatis et apicem subtiliter crassitunicatis vel crassitunicatis. Caulocystidiis absentibus. Hyphis fibulatis, in stipite et trama dextrinoideis. Ad lignum et ramulos putridos et ad corticem arboris.

Holotypus: Benin, Provincia Atacora, Tanougou, cataracta Tanougou, 9. IX. 1997 leg. V. Antonín B97.190 (holotypus in herbario BR101225-54 asservatur).

Carpophores single. **Pileus** 9–20 mm broad, broadly conical to convex with a small conical central papilla when young, then ± conical to almost applanate with central depression with small papilla or without it, with involute, then straight to slightly uplifted, crenulate and slightly translucently striate margin, striate-sulcate, finely tomentose, slightly rugulose at centre, brownish orange (6B5–6, 6B–C8, 7C7–8) at centre, and slightly paler (5–6A5) towards margin when young, pallescent to pale greyish to orange brown (6B5–7, 6C6) at centre and orange-ochraceous (5–6A5) towards margin when old (rarely almost whitish at margin when old). **Lamellae** distant, L = (8–)10–16, l = 0–1, broadly adnate to an adpressed pseudo-collarium, narrow (up to 2 mm), slightly intervenose at base when old, pale ochraceous-cream (3–5A2–3) when young, slightly paler (3–4A2) when old, with concolorous, finely pubescent edge. **Stipe** 11–33 x 0.2–0.8 mm, cylindrical, slightly broadened at apex, slightly broadened to subbulbillose, mostly slightly curved, lustrous, smooth, glabrous, very slightly pruinose at apex when young, translucently whitish when very young, then pale ochraceous to cream-ochraceous (3–5A2–3, ± concolorous with lamellae) at apex, through an orange-brown zone up to dark

brown (7–8E–F7–8) towards base; with well-developed dirty yellowish pad of basal mycelium on substrate around stipe base.

Basidiospores (11.5–)12.5–15.5 x 3.5–5.0(–5.5) μm, E = 2.6–4.0, Q = 2.9–3.4, clavate to fusoid, thin-walled, hyaline, nondextrinoid. **Basidia** 27–32 x 6.5–9.0 μm, 4-spored, clavate. **Basidioles** 13–38(–43) x 3.7–10 μm, cylindrical, clavate, fusoid. **Cheilocystidia** in the form of broom cells of the Siccus-type, (10–)12–20(–24) x 4.5–10 μm, (narrowly) clavate to subcylindrical, thin-walled, nondextrinoid, with nodulose, thin- to slightly thick-walled, short, up to 3.0 x 1.0 μm projections; mixed with rare smooth elements. **Pleurocystidia** absent. **Trama hyphae** cylindrical to subinflated, thin- to sligthly thick-walled, dextrinoid, up to 20 μm wide. **Pileipellis** a hymeniderm made up of broom cells of the Siccus-type, 11–20(–25) x 6.0–11 μm, clavate or cylindrical, thin-walled with slightly thick-walled apex, mixed with entirely thick-walled ones, with 10–25(–35) digitate, nodulose, obtuse to subacute, slightly thick-walled, up to 4.0 x 1.0(–1.5) μm projections; some cells transient to subcoralloid broom cells; thick-walled parts ochraceous in KOH. **Stipitipellis** a cutis consisting of cylindrical, parallel, slightly thick-walled, dextrinoid, up to 5.0 μm wide hyphae with hyaline (above) to ochraceous (below) walls in KOH. **Caulocystidia** absent. **Clamp connections** present in all tissues.

Ecology – Saprophytic, growing on dead and decaying wood and twigs as well as on bark of a living tree.

Distribution – Known only from several localities in Benin and the Democratic Republic of Congo.

REVISED SPECIMENS – BENIN: Atacora Province, Tanougou, Choutes de Tanougou, 9 Sept. 1997 leg. V. Antonín B97.190 (holotype, BR101225-54). – Ditto, 8 Sept. 1997 leg. V. Antonín B97.176 (BR 101214-43). – Ditto, 26 Aug. 1997 leg. V. Antonín B97.108 (BR). – Atacora Province, Kounagnigou, 3 Sept. 1997 leg. V. Antonín B97.145 (BR 101188-17). – Atacora Province, Kota, 29 Aug. 1997 leg. V. Antonín B97.122 (BR 101166-92). – Borgou province, Wari Maro, 21 Aug. 1997 leg. V. Antonín B 97.74 (BR 101122-48). – **DEMOCRATIC REPUBLIC OF CONGO:** Katanga Province, Plateau de Biano, Mengé, 4 Apr. 1986 leg. J. Schreurs 1594 (BR 8343-01).

Comments – *Marasmius tanougouensis* is characterised by having a rather small, brownish orange to pale orange-brown pileus, distant lamellae, a filiform, orange-brown to dark brown stipe, moderately large basidiospores and very short projections of cheilocystidia and pileipellis broom cells. It lacks pleuro- and caulocystidia.

Collection J. Schreurs 1594 differs only in having longer projections of the cheilocystidia (1.5–5.5 μm long) and pileipellis broom cells (3.5–7.5 μm long); in other features, it agrees with the other mentioned collections.

Among species of series *Leonini* without caulocystidia and with cheilo-cystidia in the form of broom cells, *Marasmius subconiatus* Petch has a paler, light orange, 4–7 mm broad pileus, lamellae with an orange edge, a smaller (7–11 x 0.05–0.1 mm), dark brown to black stipe and smaller basidiospores (6.5 x

2.5 μm) (Desjardin & al. 2000); *M. berteroi* (Lév.) Murrill has an only 2–11 mm broad pileus (8–15 mm broad according to Singer 1976), and an entirely brown to dark brown stipe (Desjardin & al. 2000); *M. corrugatus* (Pat.) Sacc. & P. Syd. has a 10–47 mm broad pileus, a more robust, 20–33 x 1–2.5 mm stipe, smaller, 7.5–11 x 3–4.5 μm basidiospores and smaller, up to 20 μm long basidia (Pegler 1983, Singer 1976). *Marasmius bambusiniformis* Singer has an only 3–12 mm broad, orange buff to pale reddish brown pileus, lamellae with an orange-brown edge, and larger basidiospores (15–18 x 3–4.5(–5) μm) (Desjardin & Horak 1997, Singer 1976); *M. onoticus* Singer has an only 5–6 mm broad pileus, longer, 13.8–18 x 3.5–4.8 μm basidiospores and longer (5.5–8.5 x 1.2–1.8 μm) projections of pileipellis broom cells (Singer 1976).

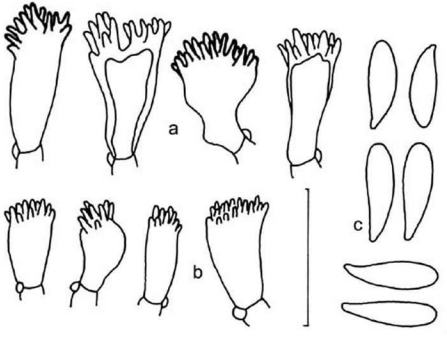

Figure 10. *Marasmius tanougouensis* (holotype): a. pileipellis cells, b. cheilocystidia, c. basidiospores. Scale bar = 20 μm.

Marasmius xestocephaloides Antonín sp. nov. Figure 11

Pileo 10–30 mm lato, convexo, subumbonato, non-striato, ochraceo vel pallide brunneo, centro obscuriore. Lamellis conferris, liberis vel adnexis, angustis, pallide alutaceis vel ochraceis. Stipite 15–40 x 1–1,5 mm, cylindraceo, saepe curvato, subtiliter pruinoso, apicem albido, ad basim castaneo-brunneo. Basidiosporis 8,0–9,5 x 3,7–4,2 µm, ellipsoideis-fusiformibus, fusiformibus, hyalinis. Basidiis tetrasporis. Cheilocystidiis 18–35 x 5,0–8,5 µm, lageniformibus, fusiformibus, plerumque rostratis, irregularibus, tenuitunicatis. Pileipellis hymeniformis, 1) e cellulis similibus cellulis hymenodermatis Marasmii sicci vel Marasmii rotalis transitoris vel subtaevibus, 12–22 x 7,0–16 µm, elavatis, pyriformibus, cylindraceis vel vesiculosis, tenui- vel subtiliter crassitunicatis; 2) e cellulis similibus cellulis hymenodermatis Marasmii sicci vel setas transitoriis, usque 65 x 8,0 µm. Caulocystidiis 25–40 x 7,0–11 µm, cylindraceis, subfusiformibus, subclavatis, ± crassitunicatis. Hyphis fibulatis, in stipite et trama dextrinoideis. In ligno putrido et ad detritum.

Holotypus: Kenya, Provincia Central, Districtus Nairobi, Thika, cataracta Thika, 16. III. 1968 leg. D.N. Pegler 94 (holotypus in herbario K(M) 116841 asservatur).

Misapplied name: *Marasmius xestocephalus* Singer s. Pegler (1977).

Pileus 10–30 mm broad, convex, subumbonate, smooth, glabrous, ochraceous to light brown, darker at centre. **Lamellae** very crowded, l = 3, free to adnexed, narrow (up to 1 mm), pale alutaceous to ochraceous. **Stipe** 15–40 x 1–1.5 mm, cylindrical, often curved, hollow, finely pruinose, whitish above, chestnut brown below; with whitish, strigose mycelium at base. **Context** thin, whitish. (According to Pegler 1977).

Basidiospores 8.0–9.5 x 3.7–4.2 μm, E = 2.0–2.6, Q = 2.2, ellipsoid-fusoid, fusoid, thin-walled, smooth, hyaline, nondextrinoid. **Basidia** 20–24 x 5.0–7.5 μm, 4-spored, clavate. **Basidioles** 15–21 x 4.0–7.0 μm, clavate, cylindrical, fusoid. **Cheilocystidia** 18–35 x 5.0–8.5 μm, lageniform, (sub) fusoid, mostly rostrate, irregular, thin-walled, hyaline, nondextrinoid. **Pleurocystidia** absent. **Trama hyphae** cylindrical to subinflated, thin-walled, hyaline, up to 15 μm wide, mixed with slightly thick-walled, dextrinoid, up to 8.0 μm wide ones. **Pileipellis** a hymeniderm made up of (1) broom cells of the Siccus-type, sometimes transient to the Rotalis-type, sometimes (almost) smooth, 12–22 x 7.0–16 μm, clavate, pyriform, cylindrical or vesiculose, thin- to slightly thick-walled, with up to 10 x 1.5 μm, ± slightly thick-walled, obtuse and smooth projections, and (2) up to 65 x 8.0 μm distinctly thick-walled broom cells transient to setoid cells (however, true setae absent!); subpileipellis made up of ± globose cells. **Stipitipellis** a cutis consisting of cylindrical, parallel, slightly thick-walled, dextrinoid, up to 8.0 μm wide hyphae, with subhyaline to pale yellowish walls in KOH. **Caulocystidia** numerous, 25–40 x 7.0–11 μm, cylindrical, subfusoid, subclavate, ± thick-walled, with subhyaline to pale yellowish walls in KOH. **Clamp connections** present in all tissues.

Ecology – Saprophytic, growing in ± dense groups on dead wood and forest debris (e.g. *Sysygium* sp.).

Distribution – So far known only from Kenya, Uganda and probably from Zambia.

REVISED SPECIMENS – KENYA: Central Province, Nairobi District, Thika, Thika Falls, 16 March 1968 leg. D.N. Pegler 94 (holotype, K(M) 116841, as *M. xestocephalus*). – **UGANDA:** Buganda Province, Mengo District, Mawakota County, Mpanga Reserve Forest, 8 June 1968 leg. D.N. Pegler U1330 (K, as *M. xestocephalus*). – **ZAMBIA:** ? Chowo Forest, 12 Dec. 1981 leg. J. Rammeloo 7843 (BR 12027-96).

Comments – *Marasmius xestocephaloides* is characterised by having an ochraceous to light brown pileus, very crowded lamellae, a chestnut brown stipe base, small basidiospores, short basidia and basidioles, cheilocystidia never in the form of broom cells but lageniform, (sub)fusoid and mostly rostrate and irregular, a pileipellis made up of both ± thin-walled broom cells and (almost) smooth cells, mixed with distinctly thick-walled broom cells transient to setoid cells, and in having cylindrical, subfusoid or subclavate, ± thick-walled caulocystidia. Pleurocystidia as well as true setae are not developed.

Collection J. Rammeloo 7843 from Zambia is very similar, both macroscopically (± same colour of carpophores, very close narrow lamellae and growth in dense groups) and microscopically (pileipellis a mixture of broom- and smooth cells, similar caulocystidia). However, its cheilocystidia are in the form of broom cells mixed with smooth, regular, lobate to subcoralloid cells. Therefore, it is included with a question-mark here.

Marasmius xestocephalus Singer represents a very close species. It differs in having smaller carpophores, larger basidiospores (11.5–14 x 4.5–5.5 μm), longer basidia and basidioles (27–30 x 8.0–10 μm), cheilocystidia in the form of broom cells, and different pileipellis broom cells. *Marasmius subarborescens* Singer has a white pileus, different cheilocystidia, and even smaller basidiospores (6.0–8.0 x 3.0–3.2 μm).

The absence of cheilocystidia in the form of broom cells represents a unique character in this series. Only *Marasmius heterocheilus* Singer is described as having mostly clavate and simple, rarely cylindrical or clavate cheilocystidia with one to four apical projections, which are often irregularly contorted. However, it differs in having a cinnamomeous coloured, up to 50 mm broad pileus, a longer and more robust stipe (up to 80 x 4 mm), smaller basidiospores (6–6.3 x 3.5–4 μm), and differently shaped caulocystidia with the same form as the cheilocystidia (Singer 1976).

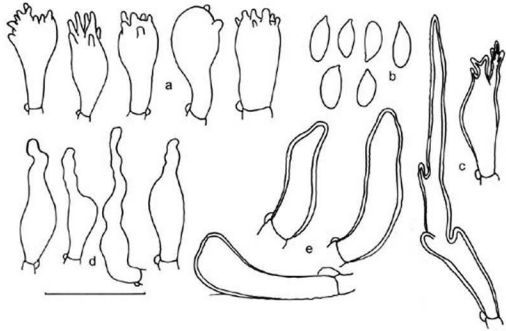

Figure 11. *Marasmius xestocephaloides* (holotype): a. pileipellis cells, b. basidiospores, c. pileosetae, d. cheilocystidia, e. caulocystidia. Scale bar = 20 μm.

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New species of marasmioid genera (Basidiomycetes, *Tricholomataceae*) from tropical Africa IV. Four new taxa of the genus *Marasmius* and one new combination

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Abstract—Four new African tropical taxa of the genus *Marasmius*, *M. aurantiostipitatus*, *M. curreyi* var. *distantifolius*, *M. rotalis* var. *latisporus* (sect. *Marasmius*) and *M. subalbidulus* (sect. *Hygrometrici*) are described and one new combination, *Setulipes congolensis*, is proposed.

Key words—*Agaricales*, new taxa, taxonomy, nomenclature

Introduction

This paper represents the fourth part of a series of descriptions of new taxa of the genera *Gloiocephala*, *Marasmius* and *Setulipes* from tropical Africa. The complete study of these genera will be published in a series of monographs under the name „Flore illustrée des champignons d´Afrique centrale" edited by the National Botanical Garden in Meise (Belgium) in 2004. For material and methods see Antonín (2003a).

Species descriptions

Marasmius aurantiostipitatus Antonín & P. Roberts spec. nov. Figure 1

Pileo usque 10 mm lato, campanulato, umbilicato, brunneo roseo vel roseo rubro. Lamellis collariatis, L = 10–12, albidis. Stipite usque 45 x 0,5 mm, filiforme, insititio, glabro, aurantiaco vel obscuriore aurantiaco. Basidiosporis 13,5–18 x 4,5–7,5 µm, lacrimiformibus vel fusiformibus, hyalinis. Basidiis tetrasporis. Cheilocystidiis e cellulis typo Marasmii sicci similibus, 7,5-21 x 5,5-11 µm, clavatis, subcylindraceis. Pleurocystidiis absentibus. Pileipellis hymeniformis, e cellulis similibus cellulis hymenidermatis Marasmii sicci, 10–20 x 4,5–11 µm, clavatis, sub-cylindraceis vel pyriformibus, (subtiliter) crassiunicatis. Caulocystidiis absentibus. Hyphis fibulatis, in stipite dextrinoideis. Ad folia and ramulos putridos.

Holotypus: Cameroon, South West Province, Korup National Park, trail from Rengo Camp to Erat, 2 May 1996, leg. P.J. Roberts K356 (holotypus in herbario K(M) 39176 asservatur).

Pileus up to 10 mm broad, campanulate, umbilicate, with a very small central papilla or without it, sulcate, smooth, brownish pink to dark pinkish red (maroon). **Lamellae** distant, L = 10–12, l = 0(–1), rather indistinctly collariate, broad, white, with concolorous edge. **Stipe** up to 45 x 0.5 mm, filiform, smooth, glabrous, lustrous, orange to deep orange; sterile stipes and rhizomorphs present.

Basidiospores 13.5–18 x 4.5–7.5 μm, E = 2.4–3.5, Q = 3.0, lacrimoid to fusoid, thin-walled, hyaline, nondextrinoid, smooth. **Basidia** 30–33 x 7.5–10 μm, 4-spored, clavate. **Basidioles** 18–38 x 4.0–8.0 μm, clavate, sub-cylindrical, fusoid. **Cheilocystidia** in the form of broom cells of the Siccus-type, 7.5–21 x 5.5–11 μm, clavate, subcylindrical, thin-walled, with thin- to slightly thick-walled, obtuse, up to 7.0 x 1.5 μm large projections. **Pleuro-cystidia** absent. **Tramal hyphae** cylindrical or subinflated, smooth, hyaline, dextrinoid, up to 15 μm wide. **Pileipellis** a hymeniderm made up of broom cells of the Siccus-type, 10–20 x 4.5–11 μm, clavate, pyriform or subcylindrical, slightly to distinctly thick-walled (up to 1.5 μm), with conical or cylindrical, thick-walled, obtuse, up to 8.0 x 1.5 μm large projections; mixed with thick-walled broom cells or coralloid cells; thick-walled parts with pale ochraceous-brown walls in KOH. **Stipitipellis** a cutis made up of cylindrical, parallel, slightly thick-walled, smooth, up to 5.0 μm wide hyphae with dark brown walls in KOH. **Caulocystidia** absent. **Clamp connections** present in all tissues.

Ecology – Saprophytic, growing gregariously on dead leaves and twigs.

Distribution – So far known only from Cameroon.

REVISED SPECIMENS – CAMEROON: South West Province, Korup National Park, trail from Rengo Camp to Erat, 2 May 1996 leg. P.J. Roberts K356 (holotype, K(M) 39176). – Ditto, trail from Rengo Camp to Ekunde – Kunde, 4 May 1996 leg. P.J. Roberts K494 (K(M) 39177).

Comments – *Marasmius aurantiostipitatus* is characterised by having a brownish pink to dark pinkish red pileus, distant, rather indistinctly collariate lamellae with concolorous edge, an orange to deep orange stipe, rather large basidiospores, cheilocystidia with obtuse projections, and a pileipellis of slightly to distinctly thick-walled broom cells with obtuse projections, mixed with thick-walled broom cells or coralloid cells. It belongs to sect. *Marasmius*.

Among species growing in tropical Africa, *Marasmius guyanensis* Mont. has a yellow orange, orange, brownish red or ferrugineous pileus, less numerous lamellae (L = 6–10), a black stipe and smaller basidiospores ((9.0–) 10–13.5(–14) x (3.0–)3.5–4.5(–5.0) μm); *M. lovedalensis* Antonín & Verbeken has a smaller, 2–5 mm broad, orange pileus, black coloured stipe and smaller basidiospores,

11–13(–14.5) x 4.5–6.0 μm large (Antonín 2003). *Marasmius marthae* Singer, with a similarly coloured pileus, has more numerous lamellae (L = 16–17) with a deep purple edge and narrower basidiospores ((8.3–)14.5–16 x 4(–4.3) μm); *M. xerampelinus* Singer has a purple red or purple pileus, a shorter (10–19 x 0.3–0.4 mm) black stipe and smaller, (6–)8.5–11 x (4–)5.5–7.5 μm large basidiospores (Singer 1976).

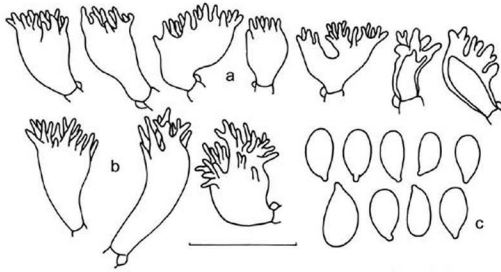

Figure 1. *Marasmius aurantiostipitatus* (holotype): a. pileipellis cells, b. cheilocystidia, c. basidiospores. Scale bar = 20 μm.

***Marasmius curreyi* Berk. & Broome var. *distantifolius* Antonín var. nov. Figure 2**

A varietate typica lamellis distantis et acie ferrugineis differt.

Holotypus: Benin, Oueme Province, Agongo, 17 VIII. 1997 leg. V. Antonín B 97.40 (BR 101094-20).

Pileus 1.5–8 mm broad, hemispherical to convex with central umbilicus with a (distinctly projecting) papilla when young, then campanulate-convex to broadly convex, rarely almost applanate with papilla when old, sulcate, crenulate at margin, finely pubescent, slightly cracking around umbilicus when old, cinnamomeous brown to reddish (up to 8C7–8) when young, pallescent with age up to pinkish ochraceous (6A4–7A4), sometimes washed-up up to whitish at margin when old, always dark brown at centre. **Lamellae** distant, L = (6–)7–9, l = 0(–1), broad, sometimes almost ventricose when old, slightly intervenose at base when old, collariate, collarium often funnel-shaped when old, pale cream when young (paler than 5A2), sometimes with an ochraceous tinge when old, with (irregularly) red-brown, finely pubescent edge. **Stipe** 4–12 x 0.1–0.2 mm, filiform, lustrous, insititious, smooth and glabrous, concolorous with lamellae at apex, through brown to black-brown towards base.

Basidiospores (8.0–)9.0–10.5(–12) x (4.5–)5.0–6.0 µm, E = 1.6–2.3, Q = 1.8, (broadly) ellipsoid, ellipsoid-fusoid, thin-walled, smooth, hyaline, indextrinoid. **Basidia** 24–28 x 8.0–10 µm, 4-spored, clavate. **Basidioles** 18–30 x 4.0–11 µm, cylindrical, clavate, fusoid. **Cheilocystidia** shaped as broom cells of the Siccus-type, 10–23 x 6.5–12 µm, clavate, subcylindrical, thin-walled with slightly thick-walled, nodulose, obtuse, up to 10 x 2.0 µm projections; projections with slightly greyish yellowish walls in KOH. **Pleurocystidia** absent. **Trama hyphae** cylindrical to subinflated, branched, thin-walled, smooth to minutely incrusted, nondextrinoid, up to 12 µm wide. **Pileipellis** a hymeniderm made up of broom cells of the Siccus-type, (5.0–)12–18 x 5.0–11(–13) µm, (broadly) clavate, subcylindrical, ± thin-walled, with slightly thick-walled, nodulose, digitate, obtuse projections, mixed with entirely or at least in upper part thick-walled, ± coralloid cells or broom cells; thick-walled parts greyish ochraceous in KOH. **Pileocystidia** absent. **Stipitipellis** a cutis made up of cylindrical, parallel, slightly thick-walled, smooth, dextrinoid, up to 6.0 µm wide hyphae with brown walls in KOH. **Caulocystidia** absent. **Clamp connections** present in all tissues.

Ecology – Saprophytic, growing on grass remnants in open grassy place.

Distribution – Known only from Benin.

REVISED SPECIMENS – BENIN: Oueme Province, Agongo, 17 Aug. 1997 leg. V. Antonín B 97.40 (holotype, BR 101094-20).

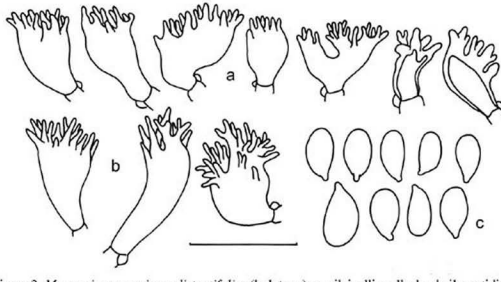

Figure 2. *Marasmius curreyi* var. *distantifolius* (holotype): a. pileipellis cells, b. cheilocystidia, c. basidiospores. Scale bar = 20 µm.

Comments – *Marasmius curreyi* var. *distantifolius* is especially characterised by coloured lamellar edge and distant lamellae. The closest var. *culmisedus* Singer differs by slightly longer and narrower basidiospores ((8–)9–12.3 x (4–)4.2-5.3(–7) µm) and closer lamellae (L = 9–18) (Singer 1976, as *M.*

graminum var. *culmisedus*). Other varieties differ by white coloured lamellar edge, differently numerous lamellae and/or different size of basidiospores (Singer 1976). For relation of *M. curreyi* and *M. graminum*, see Antonín & Noordeloos (1993).

Marasmius rotalis var. *latisporus* Antonín var. nov. **Figure 3**

A varietate typica basidiosporis latis (7.7–10.1(–10.8) x 4.6–5.4 μm) et stipitibus longis (usque 140 mm) differt.

Holotypus: Burundi, prov. Bururi, Bururi, 7. II. 1979 leg. J. Rammeloo 6577 (holotypus in herbario BR 11930-95 asservatur).

Pileus up to 4 mm broad, campanulate, umbilicate, with less distinct to distinct central papilla, distinctly sulcate, glabrous to minutely granulose, margin crenulate, pale beige, with or without black centre. **Lamellae** moderately distant, L = 14–17, l = 0(–1), collariate, white to whitish, with concolorous edge. **Stipe** very long, up to 140 x 1 mm, filiform, glabrous, smooth, lustrous, insititious, dark brown to black, apex concolorous with lamellae.

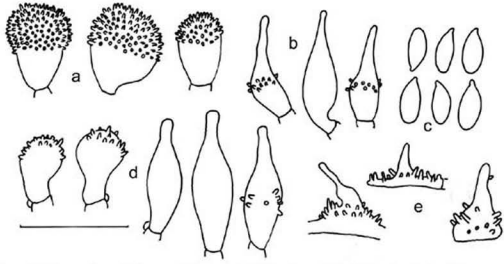

Figure 3. *Marasmius rotalis* var. *latisporus* (holotype): a. pileipellis cells, b. basidiospores, c. cheilocystidia. Scale bar = 20 μm.

Basidiospores 7.7–10.1(–10.8) x 4.6–5.4 μm, E = 1.6–1.9, Q = 1.7, ellipsoid, ellipsoid-fusoid, thin-walled, hyaline, nondextrinoid. **Basidia** not found. **Basidioles** 15–27 x 5.0–10.0 μm, cylindrical, clavate, fusoid. **Cheilocystidia** similar to pileipellis cells, 15–29(–38) x 6.0–17 μm, clavate, thin-walled. **Pleurocystidia** absent. **Trama hyphae** cylindrical, ± thin-walled, nondextrinoid, up to 8 μm wide. **Pileipellis** a hymeniderm made up of broom cells of the Rotalis-type, 11–29 x 7.0–19 μm, thin- or slightly thick-walled, (broadly) clavate, nondextrinoid; projections digitate, obtuse, slightly thick-walled, up to 4.0 x 1.0 μm. **Stipitipellis** a cutis made up of cylindrical,

parallel, smooth, dextrinoid, up to 5.0 μm wide hyphae. Stipe medulla hyphae dextrinoid. **Caulocystidia** absent. **Clamp connections** present in all tissues.

Ecology – On dead, fallen leaves.

Distribution – Known only from the type locality in Burundi.

REVISED SPECIMEN – BURUNDI: Bururi Province, Bururi, 7 Feb. 1979 leg. J. Rammeloo 6577 (BR 11930-96).

Comments – *Marasmius rotalis* var. *latisporus* belongs to sect. *Marasmius* and differs from the type variety by having a very long stipe and larger, especially broader basidiospores.

***Marasmius subalbidulus* Antonín spec. nov.** **Figure 4**

Pileo 0,5–1,5 mm lato, convexo vel applanato, centro obtuso, subtiliter sulcato, albo, centro luteolo. Lamellis distantibus, L = 7–9, l = 0–1, breviter adnatis, non-collariatis, albidis, acie concolore. Stipite usque 15 mm longo, insititio, apice albido, basim brunneo. Basidiosporis 8,0–9,5 x 3,5–4,5 µm, ellipsoideis-fusiformibus, hyalinis, inamyloideis. Basidiis tetrasporis. Cheilocystidiis e cellulis similibus cellulis cheilocystidiorum Marasmii rotalis, 12,5–14 x 8,0–9,5 µm, clavatis vel pyriformibus et e cellulis lageniformibus vel fusiformibus, 18–27 x 6,0–9,0 µm. Pleurocystidiis similibus cheilocystidiis typi 2. Pileipellis hymeniformis e cellulis elavatis, ellipsoideis, pyriformibus vel vesiculosis, similibus cellulis hymenidermatis Marasmii rotalis, 13–21 x 9,0–17 µm constructa. Pileocystidiis 18–25 x 6,0–8,0 µm, lageniformibus vel fusiformibus, basin versus cum diverticulis infrequentibus, nonnumquam laevibus. Caulocystidiis 8,0–16 x 3,0–10 µm, lageniformibus vel conicis, basin versus cum diverticulis. Hyphis indextrinoideis, fibulatis.

Holotypus: Uganda, prov. Buganda, reg. Mengo, silva Mabira, 9. VI. 1968 leg. D.N. Pegler U1376 (holotypus in herbario K(M) 115020 asservatur).

Neither macroscopic description nor slide available; macrodescription made according to dry carpophores. **Pileus** 0.5–1.5 mm broad, convex to applanate, without central papilla, slightly sulcate; white, with yellowish centre. **Lamellae** distant, L = 7–9, l = 0–1, well-developed, shortly adnate, without collarium; white, edge entire, concolorous. **Stipe** up to 15 mm long, ± filiform, insititious, whitish above, brown towards base. Forming sterile stipes and rhizomorphs.

Basidiospores 8.0–9.5 x 3.5–4.5 μm, E = 1.9–2.3, Q = 2.1, pip-shaped, ellipsoid-fusoid, thin-walled, smooth, hyaline, nondextrinoid. **Basidia** (one found) 16 x 8.0 μm, 4-spored, broadly clavate. **Basidioles** 10–18 x 4.0–8.0 μm, clavate, cylindrical, often fusoid. **Cheilocystidia** of two types: (1) broom cells of the Rotalis-type, 12.5–14 x 8.0–9.5 μm, clavate, pyriform, thin-walled, ± hyaline, nondextrinoid, and (2) lageniform to fusoid, often rostrate, obtuse to subcapitate, 18–27 x 6.0–9.0 μm, ± thin-walled cells, sometimes with scattered diverticula at basal part, nondextrinoid. **Pleurocystidia** in the form of cheilocystidia of type 2. **Hyphae** ± cylindrical, thin-walled, branched, smooth, hyaline, nondextrinoid, up to 8.0 μm wide. **Pileipellis** a hymeniderm made up of clavate, ellipsoid, pyriform to vesiculose, nondextrinoid broom cells of the Rotalis-type, 13–21 x 9.0–17 μm, thin-walled at base, thin- to slightly

thick-walled (walls up to 1.0 μm) above, warts ± narrowly conical, subacute to acute, up to 1.5 μm long; thick-walled cells subhyaline to pale yellowish-brownish in KOH. **Pileocystidia** 18–25 x 6.0–8.0 μm, lageniform to fusoid, often rostrate, obtuse to subcapitate, thin- to mostly slightly thick-walled, mostly with some projections in basal parts, sometimes smooth, nondextrinoid, subhyaline or with pale yellow-brown walls in KOH. **Stipitipellis** a cutis made up of parallel, cylindrical, slightly thick-walled, diverticulate, nondextrinoid, up to 5.0 μm wide hyphae, with brown walls in KOH; diverticula up to 2.0 μm long, cylindrical to conical, obtuse to subacute, pale brown to dark brown in KOH. **Caulocystidia** scattered, 8.0–16 x 3.0–10 μm, lageniform or conical, at base with diverticula, slightly thick-walled, nondextrinoid, concolorous with stipitipellis. **Clamp connections** rare, but present in all tissues.

Ecology – Growing on dead leaves.

Distribution – Hitherto known only from Uganda.

REVISED SPECIMEN – UGANDA: Buganda Province, Mengo District, Mabira Forest, 9 June 1968 leg. D.N. Pegler U1376 (holotype, K(M) 115020, as *M. rotalis*).

Comments – *Marasmius subalbidulus* is characterised by having a white pileus, distant lamellae, well-developed, ± lageniform pileo-, cheilo- and pleurocystidia, two types of cheilocystidia and small, lageniform caulocystidia. It belongs to sect. *Hygrometrici*. Although neither a macroscopic description nor slides are available, its features are so distinct that it clearly represents a new species.

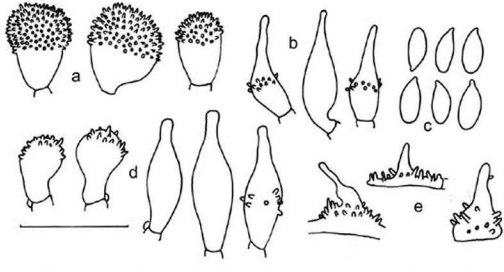

Figure 4. *Marasmius subalbidulus* (holotype): a. pileipellis cells, b. pileocystidia, c. basidiospores, d. cheilocystidia (two types), e. caulocystidia. Scale bar = 20 μm.

The only species from this section with a white pileus becoming pale yellow when dry is *M. dicandidus* Desjardin, Retnowati et E. Horak described from

Bali (Indonesia). It differs in having larger basidiospores (8–10 x 4–5 μm) and lacks cheilo-, pleuro-, pileo- and caulocystidia (Desjardin & al. 2000). Also *M. paucilamellatus* Desjardin & E. Horak, known from Papua New Guinea, is pale coloured. However, it has a pale yellow, then cream-tan pileus, lacking lamellae when young, later distant (L = 3–5), longer and narrower basidiospores ((8.0–)8.5–10(–12) x 3.5–4.0 μm), cheilocystidia in the form of fusoid-mucronate cells, and lacks developed pleuro-, pileo- and caulocystidia (Desjardin & Horak 1997).

New combination

***Setulipes congolensis* (Beeli) Antonín comb. nov.**

Basionym: *Marasmius subsplachnoides* Britzelm. ("Fr.") var. *congolensis* Beeli, Bull. Soc. Roy. Bot. Belg 60: 159. 1928.

Singer (1964, 1965) included this species in sect. *Sicci*. A revision of the type specimen (Democratic Republic of Congo, Equateur Province, Eala, June 1923 leg. M. Goossens-Fontana 108, BR 11421-72) showed, that the only carpophore of the type specimen does not have a hymeniform pileipellis, and undoubtedly belongs to the genus *Setulipes*. However, two other specimens cited in the protologue by Singer (Eala, leg. M. Goossens-Fontana 24, BR A650 and Kipushi, Kipopo, leg. M.C. Schmitz-Levecq 51, BR K243) really belong to sect. *Sicci* and represent *Marasmius sierraleonis* Beeli.

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Sebacinoid species from the Pakaraima Mountains of Guyana

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Abstract—Two sebacinoid basidiomycetes, possibly ectomycorrhizal with *Dicymbe corymbosa (Caesalpiniaceae)*, are reported from the Pakaraima Mountains of western Guyana. One is a *Tremellodendron* species, for which the new combination *T. ocreatum* is proposed. The other is a *Sebacina* species indistinguishable from the cosmopolitan *S. incrustans*.

Key Words—Neotropics, biodiversity, fungi, Guiana Shield

Introduction

As part of a project to survey macromycetes of Guyana's Pakaraima Mountains, collections were made of two sebacinoid basidiomycetes, possibly ectomycorrhizal with *Dicymbe corymbosa* Spruce ex. Benth. *(Caesalpiniaceae)*.

The area surveyed consists of extensive, primary seasonal evergreen rain forest in the Upper Potaro River Basin near the common border with Venezuela and Brazil (general area 5°N 59°W). Research site descriptions and survey methods have been published previously concerning the *Boletaceae* (Henkel 1999), *Amanitaceae* (Simmons et al. 2001), *Clavulinaceae* (Thacker & Henkel 2004), *Cortinariaceae* (Matheny et al. 2003), *Russulaceae* (Henkel et al. 2000; S.L. Miller et al. 2001), *Aphyllophorales* (Aime et al. 2003), and *Elaphomycetaceae* (O.K. Miller et al. 2001).

One of the two sebacinoid taxa collected is a species of *Tremellodendron* Atk., a small genus of clavarioid or ramarioid form in the family *Sebacinaceae*. *Tremellodendron* is confined to the New World, with the majority of species and collections from continental North America, and a minority from the Caribbean, Central and South America. The genus was monographed by Burt (1915) and Bodman (1942), but lacks a modern treatment. Species of *Tremellodendron* are currently distinguished on field characters, but these are "extremely variable" (Bodman 1942) depending on age and conditions of growth. Microscopic characters are readily distinguished for the genus, but vary little between species. Despite this, the collection from Guyana appears distinct.

The second Guyanese taxon is an effused *Sebacina* species, morphologically indistinguishable from the cosmopolitan *Sebacina incrustans* (Pers.:Fr.) Tul. This is the first report of *S. incrustans* in Guyana.

Herbaria designations are according to Holmgren et al. (1990): BRG – University of Guyana, Georgetown; K – Royal Botanic Gardens, Kew, U.K; BPI – U.S. National Fungus Collections. Color designations, noted in parentheses, are from Kornerup and Wanscher (1981).

In the following descriptions, color and habitat details for each species were taken from the collectors' field notes. Dried specimens were mounted in 5% ammonia solution for examination by light microscopy. Illustrations of microscopic details were made using a Leitz Wetzlar drawing tube.

Taxonomy

Tremellodendron ocreatum (Berk.) P. Roberts, comb. nov. Figs. 1, 2

Thelephora ocreata Berk. in Hooker's J. Bot. 8: 238 (1856)

Basidiomata terrestrial, solitary, 40–80 mm tall, branching from the base; branches cylindrical, ascending, non-anastomosing, dividing 4–5 times, the tips acute; entirely white when fresh, infertile surfaces matted or downy, the hymenium more polished; pale ochraceous buff when dried, the hymenium pale tan, greying with age. *Hyphae* hyaline to yellow-brown, 2–3 μm wide, sparsely branched, thick-walled, lacking clamp-connections. *Hyphidia* not seen. *Cystidia* absent. *Basidia* tremelloid, subglobose to ellipsoid, occasionally short-stalked and broadly clavate, 10–13 x 7.5–9 μm. *Sterigmata* four, not clearly seen. *Basidiospores* oblong (Q = 1.5–2.1), sometimes ventrally depressed, 7–10 x 4–5.5 μm.

Specimens examined. BRAZIL: Amazonas, São Jeronimo (formerly Panuré), amongst dead leaves, 1853, *R. Spruce* 11 (ex herb. M.J. Berkeley), holotype, K(M) 8527; same details (ex herb. J. Hooker), isotype, K(M) 8528; GUYANA: Region 8 Potaro-Siparuni, Pakaraima Mountains, Upper Potaro River Basin, under *Dicymbe corymbosa*, 2 July 2002, *T. Henkel* TH 8518 (also K(M) 116793 and BRG); same details, *Dicymbe*-2 plots, frequent, scattered under *D. corymbosa*, 19 June 2002, *M.C. Aime* MCA 2069 (BPI 843411).

Commentary. Corner (1968) noted that *Thelephora ocreata* was a *Tremellodendron* species, but did not recombine the name. We concur with Corner's earlier observation. Basidium and spore morphologies, as well as coralloid macromorphology, clearly align *T. ocreata* with *Tremellodendron* of the *Sebacinaceae*. The description above is taken from the type collections and Berkeley's original account. Based on examination of the types and our Guyanese collections, the salient features of this species within *Tremellodendron* are the lack of anastomosis in the stipe or branches, the more or less cylindrical cross-section of the branches, and their acute, not flattened, tips.

The most common North American species, *Tremellodendron candidum* (Schwein.) Atk. and *T. schweinitzii* (Peck) Atk. (= *T. pallidum* nom. illeg.), are of similar colour and stature but differ inter al. in being highly anastomosed and having flattened, non-acute branch apices. This was noted by Berkeley (1856), who characterized his new species as "resembling *Thelephora candida* Schwein., but without any tendency to be compressed. Much divided forms of *T. pallida* Schwein. also approach it, but there is little doubt that it is distinct."

Of the remaining branched species, *Tremellodendron cladonia* (Schwein.) Burt and *T. tenax* (Schwein.) Burt both have variously flattened branches, *T. tenue* Burt is very sparsely branched, and *T. merismatoides* (Schwein.) Burt has acute but fine, hair-like branches. All are said to be much smaller (under 60 mm high fide Bodman, 1942) than *Tremellodendron ocreatum*.

The Guyanese material differs from the type specimens of *T. ocreatum* primarily in that the basidiomata may reach up to 110 mm tall. All were white to cream (4A2-4A3) when fresh, drying pale buff (4A4). In some basidiomata, however, the hymenium became thickened and dark grey (5C3) when fresh, possibly a factor of age or maturation. Microscopically, branched hyphidia and mature basidia with four flexuose sterigmata are readily apparent (both collapsed in the type specimens); basidiospores measured 7.5–10 x 5–6 μm. It is possible that the Guyanese collection represents a distinct taxon with a graying hymenium, but for the moment it seems best referred to *Tremellodendron ocreatum* which agrees both macroscopically and microscopically in every other respect.

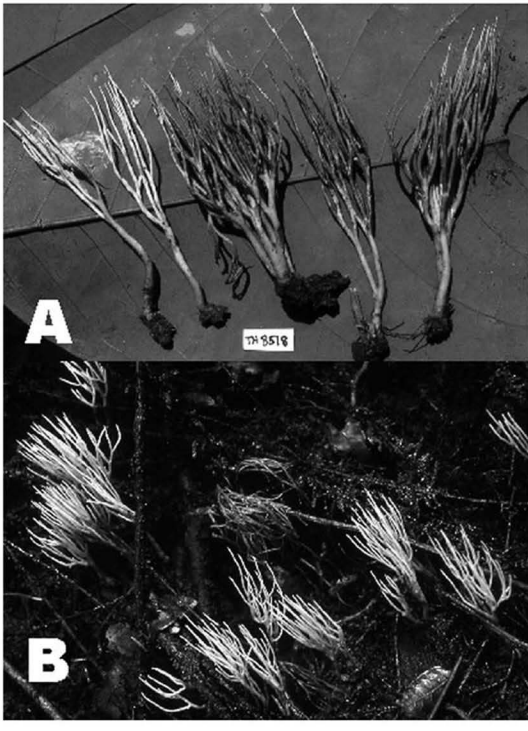

Fig. 1. Basidiomata of *Tremellodendron ocreatum*. A. *Henkel 8518* (× 0.65) B. Field habit under *Dicymbe corymbosa* (× 0.4)

Recent studies have shown that members of the *Sebacinaceae* are important components of the ectomycorrhizal mycota in temperate regions (Selosse et al. 2002, Urban et al. 2003). An ectomycorrhizal habit is also presumed for *Tremellodendron ocreatum* which, in the Pakaraima Mountains, fruits abundantly and exclusively under the ectomycorrhizal *Dicymbe corymbosa* (Henkel et al. 2002). A Nuclear-LSU sequence for *T. ocreatum* has been obtained and deposited in GenBank (MCA 2069; AY393696) that confirms its affinities with members of the *Sebacinaceae*.

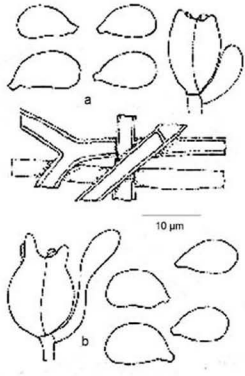

Fig. 2. *Tremellodendron ocreatum*. a) basidiospores, immature basidium, and subhymenial hyphae (isotype); b) basidium and basidiospores (*Henkel 8518*)

Sebacina incrustans (Pers.: Fr.) Tul. in *J. Linn. Soc. Bot.* 13: 36 (1871)Fig. 3

Basidiomata effused, incrusting, on fallen twigs and leaves, and living rootlets, saplings, and stems, smooth, tough, sordid greyish (5D3-5C2) when fresh, drying pale ochraceous. *Hyphae* hyaline, 2.5 - 3.5 µm wide, sparsely branched, thick-walled, lacking clamp-connexions. *Hyphidia* weakly branched. *Cystidia* absent. *Basidia* tremelloid, ellipsoid, occasionally short-stalked and broadly clavate, 15–18 x 10–12.5 µm. *Sterigmata* four, flexuose. *Basidiospores* oblong (Q = 1.4–1.9), 10–12.5 (- 13.5) x 6–8.5 µm.

Specimens examined. ENGLAND: Devon, Dunsford Woods, encrusting litter and twigs, 30 Aug. 1992, *P. Roberts* 424, K(M) 116587; GUYANA: Region 8 Potaro-Siparuni, Pakaraima Mountains, Upper Potaro River Basin, under *Dicymbe corymbosa*, 23 June 2002, *T. Henkel* 8484 (K(M) 116794 and BRG); same details, *Dicymbe*-2 plots, covering rootlets, stems and lower portion of saplings, 12 June 2002, *M.C. Aime* 1975 (BPI 843410).

Commentary. The Guyanese material, described above, is, for the most part, indistinguishable from temperate collections of *Sebacina incrustans*. This is a species originally described from Europe and possibly cosmopolitan, having been reported from Central America and Trinidad by Lowy (1971). A typical collection from England (selected for comparison and cited above) was "sordid greyish-white" when fresh and had basidiospores (from a print) measuring 10–15 x 6.5 - 8.5 μm (Q = 1.4–1.9). Some temperate collections of *S. incrustans* (e.g. McGuire, 1941) have been described as having more acutely branched, tortuous hyphidia than those observed in the Guyanese material.

Sebacina incrustans is known to be ectomycorrhizal with *Picea abies* (Urbanet al. 2003) and it is probable that, like *Tremellodendron ocreatum*, *S. incrustans* is also ectomycorrhizal with *Dicymbe corymbosa* in Guyana, where it is a commonly encountered in *D. corymbosa*-dominated forests.

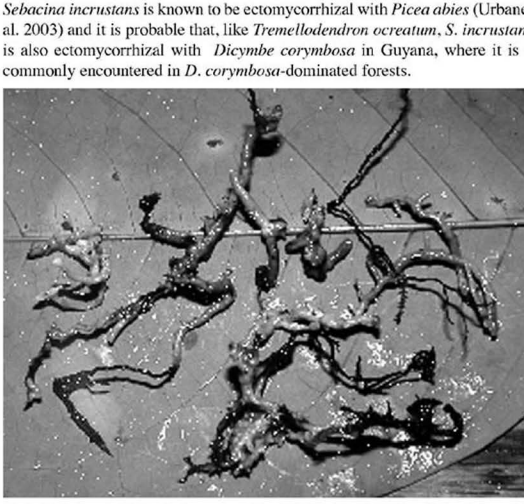

Fig. 3. Basidiomata of *Sebacina incrustans, Aime 1975*, showing field habit of effused, incrusting growth over living and dead plant rootlets and stems

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Comparative analysis of common indoor *Cladosporium* species based on molecular data and conidial characters

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Abstract—ITSs and D1/D2 regions of the LSU rRNA genes of 57 ATCC strains of *Cladosporium* representing common airborne species, *C. herbarum*, *C. cladosporioides* and *C. sphaerospermum*, were PCR-amplified and sequenced in both directions. Sequence alignments and subsequent maximum parsimony analyses on both of the datasets indicated that *C. herbarum* and *C. cladosporioides* were more closely related to each other than either was to *C. sphaerospermum*. Most strains were clustered into one of the three individual groups according to the species identification by the depositors. However, four strains deposited as *C. herbarum* clustered with *C. clasdosporioides*. Conidial shapes of these four strains resembled *C. herbarum* although their size was smaller. Another strain deposited as *C. cladosporioides* clustered with *C. herbarum*. The conidia of this strain were longer than the typical conidia of *C. cladosporioides*, but resembled *C. cladosporioides*. ATCC 64726 (deposited as *C. cladosporioides*) and ATCC 26362 (deposited as *C. herbarum*) were placed close to *C. sphaerospermum*, but formed a distinct subgroup with strong bootstrap supports. The strains of these three *Cladosporium* species collected over a wide range of areas displayed minute intra-species sequence variations and varying degrees of conidial shapes and sizes.

Key words—Airborne *Cladosporium*, rDNA

Introduction

Conidia of *Cladosporium herbarum* (Pers.:Fr.) Link, *C. cladosporioides* (Fresen.) G. A. de Vries and *C. sphaerospermum* Penz. are some of the most ubiquitous bioaerosols found in indoor and outdoor samples (Al-Doory 1984, Domsch et al. 1980, Mullins 2001, Shelton et al. 2002). *C. herbarum* has been implicated as a potential cause of human health problems such as allergy, irritation and infection (Horner et al. 1995). *C. sphaerospermum* and *C. cladosporioides* are two of the most reactive species against patients with allergy, rhinitis and asthma (Tarlo et al. 1988). It also has been shown that *C. sphaerospermum* contained toxigenic compounds to humans (David et al. 1975). Despite the abundance of *Cladosporium* spores in the environment and their medical implications, molecular and morphological protocols for

identification of *Cladosporium* isolates to species level still require substantial improvement. Frequently, published taxonomic profiles of airborne fungal samples have been determined largely at genus level due to the lack of easily identifiable morphological characters and limited DNA sequence information at species level. Although keys were proposed to identify species based on morphology, host and substrate utilization (deVries 1952; de Hoog et al. 2000; David 1997; Domsch et al. 1980; Samson et al. 2000), frequently morphological descriptions were not clear-cut, sizes of reproductive structures tend to overlap among the different taxa and many descriptive terms are often vague. An expanded key recently proposed by Ho et al. (1999) differentiated the three species according to shape, size and ornamentation of conidia, and characters of conidiophores. They did use characters of chlamydospores to identify some species, but chlamydospores were not important for *C. herbarum*, *C. cladosporioides*, or *C. sphaerospermum*.

In 2002, Wirsel et al. estimated the phylogenetic relationship with a few ITS sequences from GenBank among strains of *C. cladosporioides*, *C. herbarum* and *C. sphaerospermum* and found that these strains were almost indistinguishable. In 2003, Braun et al. proposed to erect the teleomorphic genus *Davidiella* and to confine *Cladosporium* to anamorphs of *Davidiella* with coronate conidiogenous loci and conidial hila and to assign many *Cladosporium*-like hyphomyces to other genera. The anamorphs of proposed *Davidiella* include three major airborne *Cladosporium* species, *C. herbarum*, *C. cladosporioides* and *C. sphaerospermum*. It has been known that conidia of *C. herbarum* are bigger than *C. cladosporioides* and the conidia of *C. sphaerospermum* are predominantly globose and often enveloped by a membrane (de Vries 1952).

In this study, the sequences of the D1/D2 regions of the LSU rRNA genes and the ITS regions of the rDNA were employed in order to establish molecular standards for the demarcation of the three species: *C. herbarum*, *C. cladosporioides* and *C. sphaerospermum*. At the same time, comparative assessment of the shape and size of conidia was also performed for these species.

Materials and Methods

Cultivation of strains

Fifty-seven strains of *Cladosporium* were obtained from the cryopreserved seed stocks at ATCC (Table 1). For the study, the strains were cultivated using six solid agars and broth media at 25°C or 30°C for approximately seven days. The media formulations included Blakeslee's formula (ATCC # 325): malt extract 20g, glucose 20g, peptone 1g and agar 20g per liter; PDA (ATCC # 336): diced potatoes 300g, glucose 20g and agar 15g per liter; Emmon's modification of Sabouraud's agar (ATCC # 28): Sabouraud's glucose broth 30g and agar 20g per liter; Harrold's M40Y (ATCC # 319): malt extract 20g, yeast extract 5g, sucrose 400g and agar 20g per liter; malt agar (ATCC # 323): Difco 0024; and malt extract agar (ATCC # 324): malt extract 20g, peptone 5g and agar 15g per liter.

Isolation of genomic DNA

Genomic DNAs from 57 strains of *C. herbarum, C. cladosporioides, C. sphaerospermum* and other species were isolated according to the method of Cenis (1992). Mycelia were pelleted by centrifugation at 13,800×g for 5 minutes, placed in yeast lysis matrix tubes (Bio101, Vista CA) and subjected to vigorous agitation in a FastPrep FP120 shaker (Bio101, Vista, CA) for two 40-second intervals at a speed setting 4.0 m/s. RNase (0.25µl in concentration of 0.5µg/µl, Boehringer Mannheim, Indianapolis IN) was added to 50µl final volume of DNA and the mixture was incubated for 30 minutes at 30°C. Prior to PCR, the concentration of genomic DNA was determined by comparing band intensity with molecular weight standard on agarose gel and UV absorbency at 260nm measured by a GeneQuant Pro RNA/DNA calculator (Biochrom, Cambridge, UK). Isolated genomic DNA was stored in a –80°C freezer.

PCR

The D1/D2 regions of the LSU rRNA genes, putatively located along 63 through 633 relative to the 5'-end of *Saccharomyces cerevisiae* LSU rRNA gene (Georgiev et al. 1981), were PCR-amplified with two primers, F63: gctgaacttaagcatatcaataagcggaggaaa and R635: tagactcctggtcctt tcaagacggge (O'Donnell 1993). ITS regions of rDNA were amplified with NS7 and LR3 primers, NS7: gaggcaataaacaggtctgtgatgc and LR3: cegtgtttcaagacggg (Vilgalys and Gonzalez 1990). Each of the 50µl PCR reaction mixtures consisted of two "Ready-to-Go" PCR beads (Amersham Pharmacia Biotech, Piscataway, NJ), 4µl of template genomic DNA (20ng), 1µl of each primer (10 pmol) and 44µl of deionized H_2O. The amplifications were carried out using a GeneAmp PCR system 9700 thermal cycler (Applied Biosystems, Foster City, CA) with the following parameters: Initial denaturation at 94°C for 5 min, 30 cycles of 94°C for 30 sec, 55°C for 2 min, 72°C for 2 min and an additional 72°C for 5min prior to maintaining the mixture at 4°C (O'Donnell 1993). DNA molecules of about 600 nucleotides were amplified and the PCR products were cleaned with Qiagen gel extraction kit by following the manufacturer's protocol (Qiagen Inc., Chatsworth, CA).

DNA sequencing

The cycle sequencing reactions were carried out using a Big Dye terminator cycle sequencing ready reaction kit (Applied Biosystems, Foster City CA). The reaction mixtures consisted of 4 µl (50 ng) of DNA template, 0.5 µl of primer (5 pmol), 8 µl of Big Dye terminator and 7.5 µl of deionized H_2O for a total volume of 20 µl. The cycle sequencing program was as follows: Initial denaturation at 95°C for 5min, 25 cycles at 95°C for 30 sec, 50°C for 30 sec, 60°C for 2 min and an additional 60°C for 7 min prior to storing the sample at 4°C. The extension products were purified with Centri-Sep spin columns (Princeton Separations, Adelphia, NJ) prior to being loaded onto an ABI 377 automated sequencer (Applied Biosystems, Foster City, CA). Sequencing gel (5% acrylamide) was cast with LongRanger Singel packs (BioWhittaker Molecular Applications, Rockland, ME). The sequences were tracked and extracted with the ABI Prism 377-96 data collection software. Primers F63, R635, NL2 (ctctctttttcaaagttcttttcatct) and NL3 (agataaaaagaacttt gaaaagagag) were used for sequencing the D1/D2 regions of LSU rRNA genes (O'Donnell 1993). Primers ITS 1 (tccgtaggtgaacctgcgg) and ITS 4 (tcctccgcttattgatatgc) were used for sequencing the ITS region (White et al. 1990; Gardes and Bruns 1993). Two sequences in different direction for each strain were compared and any mismatching nucleotides were corrected by comparing them with the electropherograms. Sequence information was submitted to GenBank, National Center for Biotechnology Information (NCBI, http://www.ncbi.nlm. nih.gov/).

Sequence alignment and phylogenetic analysis

The sequences were aligned with CLUSTALX (Thompson et al. 1997). Phylogenetic relationships were estimated with PAUP 4.0b4a (Swofford 2000). For alignments with CLUSTALX, the gap opening cost and the gap extension cost were set between 2 and 16 in increments of 2. All

nucleotides were unordered and given equal weights. Gaps were considered as missing bases and heuristic searches were carried out with the branch swapping option using the tree-bisection-reconnection algorithm. Starting trees were obtained via stepwise addition and branches of maximum length zero were allowed to collapse yielding polytomies.

The sequences of *Mycosphaerella juvenis* (AF309586), *M. sutoniae* (AF309587) and *M. heimioides* (AF309577) from GenBank were included in the data set of the D1/D2 regions of the LSU rRNA genes. The sequence of *M. juvenis* was chosen as outgroup. Four sequences of *C. cladosporioides* (AF455525, AF455535, AF455472 and AF455517), two sequences of *C. herbarum* (AF455404 and AF455517) and one sequence of *C. sphaerospermum* (AF455481) from GenBank were added to the ITS data set. The sequences of *Cladophialophora bantiana* (Saccardo) de Hoog et al. (AF397182), *M. graminicola* (Fuckel) Sanderson (AJ300330) and *Septoria passerinii* Saccardo (AF181697) from GenBank were also added to the ITS data set to estimate relative phylogenetic relationship to the three *Cladosporium* species.

Results

Sequence alignment and maximum parsimony analysis on the 56 D1/D2 sequences of 575 characters clustered the *Cladosporium* strains into three major clades. The first clade included 24 *C. cladosporioides* strains and three strains deposited as *C. herbarum* (ATCC 58927, ATCC 60569 and ATCC 6506). The second clade included 16 *C. herbarum* strains and ATCC 201105. The third group consisted of four *C. sphaerospermum* and one subgroup of ATCC 26362 and ATCC 64726. Out of 575 total characters, 397 were constant, 63 were parsimony-uninformative and 115 were parsimony-informative. The sequences were mostly consistent within each of the three species. *C. herbarum* and *C. cladosporioides* differed by 3 nucleotides. *C. herbarum* and *C. cladosporioides* differed from *C. sphaerospermum* by 13 and 14 nucleotides respectively. These three species formed one lineage (Figs. 2 and 3). Six strains were aberrant in their placement with regard to other strains of the same name (Fig. 2). The sequences of *C. herbarum* and *C. sphaerospermum* were almost identical among the strains of each group in ITS and D1/D2 region of the genes. In contrast, the ITS sequences of six strains identified as *C. cladosporioides* varied by as many as three nucleotides from the other strains of the species. As before, ATCC 26362, ATCC 6506, ATCC 58927, ATCC 28987, ATCC 201105, ATCC 64726 and ATCC 60569 failed to cluster with the majority of the strains of the species (Fig. 3). ATCC 6506, ATCC 58927, ATCC 28987 and ATCC 60569 clustered with *C. cladosporioides* while ATCC 26362 and ATCC 64726 formed a subgroup closely located to the group of *C. sphaerospermum*. ATCC 201105 was clustered with *C. herbarum* (Fig. 3). Out of 631 total characters, 324 were constant, 110 variables were parsimony-uninformative and 197 variables were parsimony-informative. Morphologically, the shapes and sizes of the conidia of the most strains conformed to published species descriptions (Fig. 1).

Table 1 Strains of *Cladosporium*, their GenBank accession numbers and places of isolation

Species Name	ATCC Number	GenBank Accession Number		Place of Isolation
		D1/D2 region	ITS region	
C. cladosporioides	11275	AY 342060	AY 361960	
	26688	AY 342062	AY 361962	Florida,USA
	38494	AY 342064	AY 351963	
	42198	AY 342065	AY 361964	WestIndies
	58227	AY 342066	AY 361964	Florida,USA
	58991	AY 342067	AY 361966	Maryland,USA
	60549	AY 342068	AY 361967	New Zealand
	64726	AY 342069	AY 361968	Georgia,USA
	66468	AY 342070	AY 361969	Maryland, USA
	201092	AY 342071	AY 463363	Washington, USA
	201095	AY 342072	AY 361970	Washington,USA
	201105	AY 342073	AY 361971	Washington,USA
	201140	AY 342074	AY 361972	Brazil
	201141	AY 342075	AY 463364	Brazil
	201142	AY 342076	AY 361973	Brazil
	6721	AY 342077	AY 361995	Canada
	66669	AY 342110	AF393689	New York, USA
	34668	AY 342079	AY 463365	Idaho, USA
	38810	AY 342080	AY 361997	Denmark
	62295	AY 342081		Illinois,USA
	62732	AY 342082	AY 361996	NorthDakota,USA
	201103	AY 342083	AY 361957	Washington,USA
	201102	AY 342107	AF393687	Washington,USA
	201098	AY 342108	AF393688	Washington,USA
	11277	AY 342113	AF393690	UnitedKingdom
	200941	AY 342114	AF393691	theNetherlands
C. herbarum	6506	AY 342084	AY 361974	
	26361	AY 342086	AY 361981	NewJersey,USA
	26362	AY 342087	AY 361982	NewJersey,USA
	58927	AY 342088	AY 361984	Maryland,USA
	60569	AY 342090	AY 361985	Israel
	62720	AY 342091	AY 361986	NorthDakota,USA
	66670	AY 342092	AY 361959	New York, USA
	66671	AY 342093	AY 463366	New York, USA
	76226	AY 342094	AY 361987	California,USA
	201090	AY 342095	AF393705	Washington,USA
	201096	AY 342096	AY 361975	Washington,USA
	201100	AY 342097	AY 361976	Washington,USA
	201101	AY 342098	AY 361977	Washington,USA
	201852	AY 342099	AY 361978	Idaho,USA
	201853	AY 342100	AY 361979	Idaho,USA
	201855	AY 342101	AY 361980	Washington,USA
	11281	AY 342106	AF393707	theNetherlands
	201091	AY 342109	AF393706	Washington,USA
	201093	AY 342111	AF393703	Washington,USA
	201094	AY 342112	AF393704	Washington,USA

Table 1 Continued

Species Name	ATCC Number	GenBank Accession Number		Place of Isolation
		D1/D2 region	ITS region	
	201854		AY 361993	
	28987		AY 361983	
C. sphaerospermum	12092	AY 342102	AY 361988	Canada
	11288	AY 342103	AY 361989	
	11289	AY 342104	AY 361958	
	11290	AY 342105	AY 361990	
	11293		AY 361992	
	200384		AY 361991	
C. breviramosum	64696	AY 345902	AF393684	
Arthrinium sacchari	76303	AY 345898	AF393679	
Amorphotheca resinae	200942	AY 352592	AF393726	

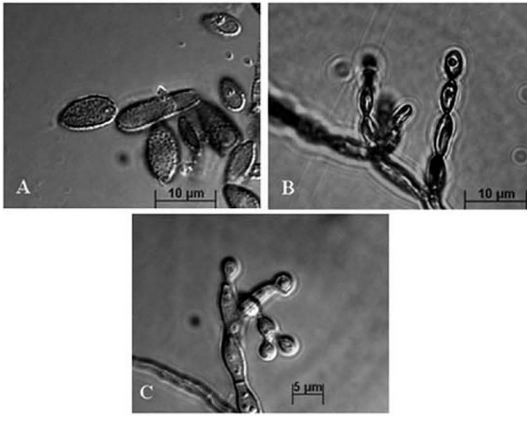

Fig. 1. Conidia of (a) *C. herbarum* ATCC 201094, (b) *C. cladosporioides* ATCC 11275 and (c) *C. sphaerospermum* ATCC 200384 observed with 1000 x magnification

Discussion

The most common airborne *Cladosporium* species, *C. herbarum*, *C. cladosporioides* and *C. sphaerospermum*, have been identified according to size and ornamentation of conidia, shape of conidiophore and degree of branching of conidial chains. However, even with pure cultures, observing naturally branched conidia or mycelium is relatively difficult and laborious. Furthermore, descriptions of morphological characters such as ornamentation and color of conidia and degree of branching of conidial chains are not clear-cut in many cases. In this study, the great majority of the strains within *C. cladosporioides*, *C. herbarum* and *C. sphaerospermum* were tightly grouped with the strains of the same name in trees derived from the D1/D2 and ITS sequence analyses. However, strain ATCC 201105, which was deposited as *C. cladosporioides*, was clustered with *C. herbarum*. The strains ATCC 58927, ATCC 6506 and ATCC 60569, which were deposited as *C. herbarum* were clustered with *C. cladosporioides*. Morphological observations of the conidia showed that the conidial shapes of these strains were congruent with the species designations provided by the depositors, although molecular information classified them as *C. cladosporioides*. The conidia of these three strains were smaller, 4.0-7.7x 3.0-3.5 μm, than *C. herbarum* (Fig. 1a). The conidia of ATCC 201105, which were longer (11.5-14.5 μm) but more ellipsoidal than *C. herbarum*, resemble *C. cladosporioides* (Fig. 1b) in shape while the molecular analysis clustered this strain as *C. herbarum*. The sequence analyses for the strains ATCC 64726 and 26362 also demonstrated that these two strains are related to *C. sphaerospermum* but separate from *C. sphaerospermum*. The conidia of ATCC 64726 were globose and similar to *C. sphaerosermum* (Fig. 1c). The strain ATCC 26362 did not produce any spores on malt extract agar. These two strains were distinct from *C. sphaerospermum* in terms of nucleotide variation and could be a separate taxon. The molecular methods described here certainly aid in resolving some questions pertaining to the identification of certain isolates that are difficult to differentiate on the basis of morphology. For example, David (1997) was of the opinion that ATCC 201100 (CV10-92a) was misidentified by Dugan and Roberts (1994), who assigned this strain to *C. herbarum*. This study shows that the duo's initial assignment was correct based upon the sequence analyses (Figs. 2 and 3). However, Roberts and Dugan were also the source of ATCC 201105, identified as *C. cladosporioides* and here clustering with *C. herbarum* (Figs. 2 and 3). ATCC 201100 (Dugan and Roberts 1994) is representative of *Cladosporium* isolates with conidia somewhat larger and more roughened than is typical of *C. cladosporioides*, but whose conidiophores are not conspicuously nodose and/or geniculate as is typical of *C. herbarum*. Accurate identification of such strains on the basis of

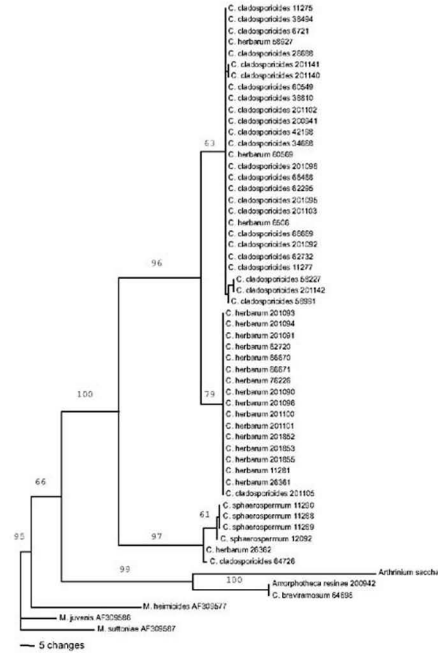

Fig. 2. Maximum parsimony tree generated with the dataset of D1/D2 region of LSU rRNA genes of *C. herbarum, C. cladosporioides, C. sphaerospermum* species and several other related species. *M. juvenis* (AF309586) was chosen as outgroup. Ingroup included 55 taxa. For the strains that were sequenced by this lab, ATCC number of each strain is specified after the species name. Three sequences of *Mycosphaerella* species were obtained from GenBank and are specified after species name in the tree. Bootstrap values larger than 50 percent at 1000 replicates are shown at each node. Bootstrap values less than 50 percent are not shown.

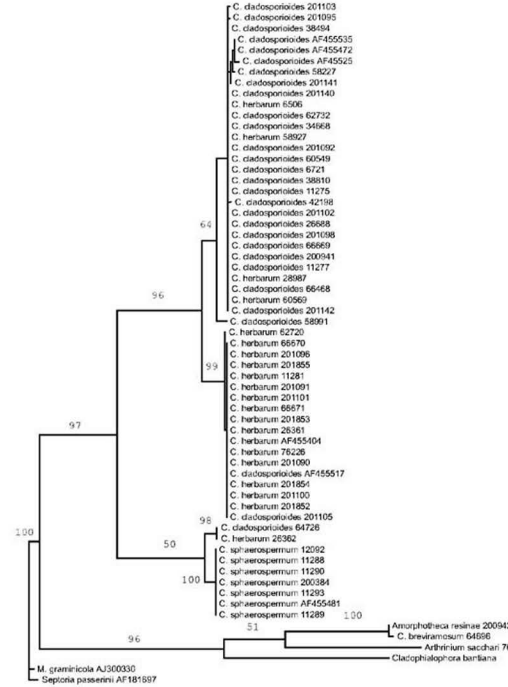

Fig. 3. Most parsimonious tree inferred with PAUP on the dataset of ITS sequences of rDNA of strains of the three airborne *Cladosporium* species. Maximum parsimony tree was constructed with the data set of 63 ITS sequences including nine sequences from GenBank. GenBank accession numbers for the sequences are given after the species name except *Cladophialophora bantiana*. *M. graminicola* was chosen as outgroup. The 62 ITS sequences were the ingroup. Numbers after species names are ATCC strain numbers. The number shown at each node is the Bootstrap value at 1000 replicates. Bootstrap values less than 50 percent are not shown.

morphology is highly problematic as the above situations illustrate. Discovery of analogous misidentifications is commonplace whenever large numbers of strains are closely examined by molecular-genetic methods (e.g. Kuhls et al. 1997).

By utilizing molecular data concurrent with information regarding conidial shapes and sizes, it was concluded that the *Cladosporium* strains could easily be differentiated at the species level. Employment of molecular and classical methods proved to be essential toward resolution of identification issues where morphological or cultural characters alone seemed insufficient. Interestingly, it would appear that the most reliable morphology-based method to differentiate these three species is by comparing the sizes of their conidia. As demonstrated in the phylogenetic trees and sequence alignments, *C. herbarum* and *C. cladosporioides* are more closely related to each other than *C. sphaerospermum* is to either one of them. The molecular data also demonstrated that strains of the three species collected over a broad range of area manifested very small nucleotide sequence variations.

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MYCOTAXON

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The genus *Astraeus* in Thailand

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Abstract—A new species of *Astraeus*, *A. odoratus*, is described from Thailand. *Astraeus odoratus* differs from *A. hygrometricus* in its strong odour when fresh, smooth outer peridium and globose spores with, long, narrow, coalescent, less densely arranged spines.

Key words—Gasteromycete, ectomycorrhizal fungi, taxonomy, ecology

Introduction

Astraeus is one of the most common gasteromycete genera in temperate and tropical ecosystems (Lloyd 1902). It is recognised in the field by the exoperidium splitting radially into segments to form a star-like pattern at maturity and by a strongly hygroscopic nature (Miller & Miller 1988). Morgan (1889) was the first to recognise *Astraeus* as a distinct genus with *A. hygrometricus* (Pers.) Morgan as the type and only species. However, this species was considered to belong to the genus *Geastrum* by many subsequent authors (e.g. Coker & Couch 1928, as *Geaster*). The genus *Astraeus* was based on the following characters; 1) no defined peristome, 2) no columella, 3) spores larger than those of any species of *Geastrum,* and 4) long, much branched, capillitial hyphae. Currently, two species are recognised in the genus, *Astraeus hygrometricus* (Pers.) Morgan, and *Astraeus pteridis* (Shear) Zeller, which differs particularly from the first in its considerably larger basidiomes. It has been reported that *Astraeus* forms ectomycorrhizal associations with several host tree species including *Pseudotsuga*, *Alnus*, *Eucalyptus* and *Castanea*

(Trappe 1967, Molina 1979, Malajczuk et al. 1982, Nouhra & Toledo 1998). *Astraeus* has been reported mainly from North America, Europe, Australia and Africa (Coker & Couch 1928, Cunningham 1944, Dring 1964) with fewer reports from Asia. This may be a reflection of the lack of collectors, although where studies have been carried out, the genus appears to be widespread (Teng 1996). Within Thailand, *Astraeus* is predominantly found in dry lowland dipterocarp forests particularly in the North and North Eastern parts of the country. It is considered to be a culinary delicacy and has a high market value. The lack of a current revision of Thai *Astraeus* species, coupled with their ability to form ectomycorrhizal associations and their importance as a source of food for local people, initiated the present study.

Materials and Methods

Astraeus basidiomes were collected during the rainy season, (May-August) 2001 and 2002, from localities in Thailand. Sources of 26 collections examined are given in Table 1. Conventional methods were used to study the basidiomes and characteristics including thickness of the peridium, glebal colour and features of external peridium whether smooth or encrusted with soil particles were recorded. Basidiospores mounted in Melzer's reagent (Largent et al. 1977) were examined and photographed by light microscopy. Spore size was determined by measuring the diameter of at least twenty spores and calculating their size ranges. Any ornamentation present was recorded and later critically analysed using scanning electron microscopy (SEM). Samples were air-dried and sputter-coated with gold and examined with a JEOL JSM-840 scanning electron microscope. The studies of fresh collections were supplemented with the examination of a selection of material of *A. hygrometricus* from the herbaria E and K (Tables 2 & 3).

Results

Basidiomes of 26 collections were examined (Fig.1). These varied in size (unexpanded specimens ranged from 18.5-39.6 mm. in diameter), shape (ranged from globose to depressed globose), colouring (ranged from dingy white to buff) and surface of the exoperidium (ranged from smooth to encrusted with soil debris). Basidiospores from all collections were studied and although varying in size, in other aspects they appeared very similar under the light microscope (Fig.2). Although examination of spores by light microscopy demonstrated that the basidiospores from the specimens were ornamented with well-developed spines, more detailed examination with SEM has enabled the collections to be divided into two distinct entities

Table 1. Specimens collected from different localities in Thailand during the present study.

No.	Code	Source
1	ASTO_01	Ban Nongpan, Dongkanyai, Yasothon province
2	ASTO_02	Ban Dongkanyai, Yasothon province
3	ASTO_03	Kudchum District, Yasothon Province
4	ASTO_04	Ban Dutoong, Muang District, Yasothon Province
5	ASTO_05	Ban Nampho, Muang District, Yasothon Province
6	ASTO_06	Ban Dutoong, Muang District, Yasothon Province
7	ASTO_07	Ban Dutoong, Muang District, Yasothon Province
8	ASTO_08	Ban Khuangkam, Muang District, Yasothon Province
9	ASTO_09	Ban Warisawadh, Pranomprai District, Roi Et Province
10	ASTO_10	Ban Nampho, Muang District, Yasothon Province
11	ASTO_11	Ban Panus, Pranomprai District, Roi Et Province
12	ASTO_12	Ban Tayiam, Muang District, Yasothon Province
13	ASTO_13	Ban Nongpan, Kamkhuankaew District, Yasothon Province
14	ASTO_14	Ban Warisawadh, Pranomprai District, Roi Et Province
15	ASTO_15	Ban Maesart, Muang District, Chiang Rai Province
16	ASTO_16	Ban Nampho, Muang District, Yasothon Province
17	ASTO_17	Ban Kammuang, Yasothon Province
18	ASTO_18	Ban Tayiam, Muang District, Yasothon Province
19	ASTO_19	Ban Bongkheelek, Khemaraj District, Ubolrajathani Province
20	ASTO_20	Ban Maesart, Muang District, Chiang Rai Province
21	ASTO_21	Ban Dutoong, Muang District, Yasothon Province
22	ASTO_22	Ban Kamnamsarng, Muang District, Yasothon Province
23	ASTO_27	Ban Srisuk, Pranomprai District, Roi Et Province
24	ASTO_28	Huathapan District, Amnajcharoen Province
25	ASTO_29	Ban Srisuk, Pranomprai District, Roi Et Province
26	ASTO_30	Ban Phosri, Muang District, Yasothon Province

referred to as spore type 1 and spore type 2 (Fig.3). In the two groups spore size varied from 8.75-15.2 μm and 7.5-15.2 μm respectively (Table 4). Type 1 was characterized by very dense ornamentation consisting of rounded, narrow, tapered, occasionally coalesced, spines 0.90-1.45 μm long, whilst Type 2 was characterized by moderately dense, rounded, long, narrower, coalesced, spines, 1.04-1.66 μm long. Our observations based on basidiome and spore morphology, show that Thai collections of *Astraeus* can be separated into 2 distinct groups. One can be recognised as *A. hygrometricus* but the other is an undescribed species. Full descriptions of the two entities are given below.

Table 2. Material examined from Royal Botanic Garden at Edinburgh (Scotland)

No	Source	Exsiccata/Collector	Date
1	Florence, Italy	F. Amatt	1821
2	Prenczow, Czechoslovakia	A. Kmet	July 1891
3	Hermosa, Southern Colorado, USA	C.F. Baker 13	29/03/1899
4	Ventuna, Greece	M. Willson 167	Nov 1908
5	Western China	E.E.Maire 1238	July 1913
6	Nagasaki, Japan	No. 574	Apr 1962
7	Zambia (Northern Rhodesia)	A. Angus M2615	08/04/64
8	Jackson Co., Michigan, USA	R. Watling 648/875A	18/04/65
9	Upper Peninsular Michigan, USA	R. Watling 709/1137A	06/08/65
10	Upper Peninsular Michigan, USA	R. Watling 900/A2067	06/08/65
11	Whitefish Pt., Michigan, USA	R. Watling 874/A2032	27/12/65
12	Lower Peninsular Michigan, USA	R. Watling 6982	04/10/69
13	Velem, Hungary	M. Seaward Wat. 26311	04/08/83
14	Lucca Prov. North East of Barga: 1/3 way to Renfio, Italy	L. Cram & M.Potts Coppins 12452	30/04/85
15	Yifeng temple, Lijiang Co., Yunnan, China	D.F.Chamberlain	May 1985
16	Near Dali to Zhongdian, Yunnan, China	R. McBeath	09/06/94
17	Sichuan Rd. from Wengsui to Xang chen, China	R. McBeath	Oct 1994
18	Tsukuba, Japan	C. Walker Wat. 26075	14/11/94
19	Inashik-gun, Ibaraki, Japan	C.Walker Wat. 26076	14/11/94
20	Spain	D.Fieldhouse Wat. 9159	Mar 1955
21	Aiken, South Carolina, USA	H.W.Ravenel 471, Fungi American Exsiccati	(1878-1887)
22	England	M.C. Cooke Fungi Britannici Exsiccati	N/A
23	France	Herb. M.C. Cooke (1885)	N/A
24	A- Brunel, France	Herb. M.C. Cooke (1885)	N/A
25	Paris	N/A	N/A
26	France	Mougeot & Nestler 1193 Crypt.Vogeso-Rhenanae Stirps	N/A
27	Paris	N/A	N/A
28	N/A	Herb. Hookerianum (1867)	N/A
29	West Australia	A. Morrison	N/A

Table 3. Material examined from Royal Botanic Garden at Kew

No.	Herb. No.	Source	Collector	Date
1	K(M) 104975	Mataru district, Sri Lanka	J.Wambeek, H. & H. E. Wanntorp	07/01/74
2	K(M) 104973	West Bengal, India	Sri Sutini Ray	1974
3	K(M) 104971	Los Limones, Honduras	M.H.Ivory	29/09/76
4	K(M) 104968	Zambia	G.D.Pearce	01/02/77
5	K(M) 104969	Mollans sur Ouveze, France	E.A.Ellis	Aug 1977
6	K(M) 104966	Honshu, Japan	D.N.Pegler	06/09/83
7	K(M) 104970	Tenerife, Canary Island	C.L.Champion	16/04/84
8	K(M) 104967	Uzbekistan	B.Mathew & C.D.Brickell	19/04/84
9	K(M) 29029	Devon, England	P.J. Roberts	Oct 1986
10	K(M) 26954	Sao Paulo, Brazil	D.N.Pegler, K.Hjortstam & L.Ryvarden	17/01/87
11	K(M) 18466	Kent, England	J. Weightman	20/10/91
12	K(M) 19550	Mexico	W.C.Weightman	10/03/92
13	K(M) 50616	Gard, France	B. W. Brown	23/03/97
14	K(M) 81503	Cornwall, England	D. Farley	09/11/2000

1. *Astraeus hygrometricus* (Pers.) Morgan

Basidiomes: globose or depressed globose, subepigeous, sessile 18.7-29.7 mm. in diam., splitting to become star-shaped, covered with thin, white mycelial layer when unexpanded which tears away at maturity, often partly encrusted with soil debris; with slight odour when fresh.

Outer peridium: whitish, thick, composed of several layers (≥1 mm.when dry), rigid, surface granulate, at maturity splitting into 5-12 acute rays which expand or recurve when moist and roll inwards again on drying; inner layer whitish becoming smoke-grey, cigar-brown or sepia, thick, extensively longitudinally cracked in mature specimens.

Endoperidum: sessile, subglobose, 13-24 mm. in diam., whitish to smoke-grey, opening by an apical, irregular mouth lacking defined peristome.

Gleba: pure white in early stages, purplish chestnut when mature, columella none; capillitium of long, branched, interwoven, hyaline, slightly encrusted, aseptate threads, 2.5-7.5 μm in diam.; wall thin or thickened with a narrow, sometimes discontinuous lumen; clamp-connections present.

Basidiospores: globose, ranging from 8.75-15.2 μm diam. including ornamentation, purplish chestnut, very densely ornamented with rounded, narrow, tapered, occasionally coalescent, spines 0.90-1.45 μm long.

Habitat: sandy or lateritic soil in dry lowland dipterocarp forests, fruiting frequently in rainy season between May-August.

Distribution: North and North Eastern areas of Thailand

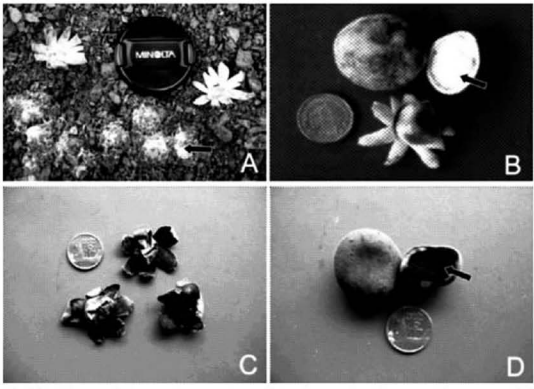

Fig. 1 Basidiomes of *Astraeus hygrometricus* and *A. odoratus*. A: *A. hygrometricus*; unexpanded specimens develop under the ground (arrow) and become strongly hygroscopic when mature. B: pure white gleba (arrow) at early stage of *A. hygrometricus*. (Thai coin *ca.*2.3 cm. in diam.) C: *A. odoratus* at maturity (Thai coin *ca.*2.0 cm. in diam.). D: unexpanded specimens of *A. odoratus* showing purplish chestnut gleba (arrow) and little soil debris on surface (Thai coin *ca.*2.0 cm. in diam.).

2. *Astraeus odoratus* C. Phosri, R. Watling, M. P. Martín, & A. J. S. Whalley, *sp. nov.*

Carpophorum: Subhypogaeum, ≤ 65 mm. diam., globosum ad depresso-globosum, sublaeve, cum applicitis, baso-rhizomorphis circumcinctum. **Exoperidium:** siccum solum crassum ≤ 1 mm., externe bubalinum vel brunneum, interne bubalinum ad olivaceo-brunneum vel fusco-brunneum, stellato-scissorum; **Endoperidium:** subglobosum, albidum vel griseum c. 13-24 mm. diam. **Gleba:** matura purpureo-brunnea, compacta. **Odor:** validus gratus. **Sporae:** globosae 7.5-15.2 μm. diam., brunneae, verrucosae cum verruculis conicus, solis vel aggregatis.1.04-1.66 μm. altis. **Fibulatae:** rarae.

Holotypus: Pranomprai district, Roi Et province, Thailand, 6 June 2002, E159386.

Basidiomes: globose or depressed globose at first, submerged, sessile, becoming superficial, expanding to become ≤ 65 mm. in diam., hard, smooth surface with few soil adhering particles, finally splitting to become star-shaped; basal mycelium consisting of brown vinaceous or bay rhizomorphs; in fresh condition specimens emitting strong odour of moist soil.

Outer peridium: buff or snuff brown, consisting of several layers, ≤1 mm. when dry, at maturity splitting into 3-9 broad rays which expand stellately when wet and become involute on drying; inner layer varying from buff to

brownish, sepia, smoke-grey, hazel, olivaceous or fuscous black, thin, scaly cracked, occasionally smooth.

Endoperidium: sessile, subglobose, ca. 13-25 mm. broad, buff to brownish or violaceous black, opening by irregular apical mouth, lacking defined peristome.

Gleba: purplish chestnut occasionally cigar-brown, date-brown, brown vinaceous or violaceous black; columella none; capillitium of long, branched, interwoven, hyaline, aseptate, slightly encrusted, threads 2.5-6.25 μm in diam.; wall thin with a narrow continuous lumen; clamp-connections occasional, usually absent

Basidospores: globose, 7.5-15.2 μm in diam. including ornamentation, purplish chestnut occasionally cigar- or date-brown, brown vinaceous or violaceous black, ornamented with moderately dense, rounded, long, narrow, coalescent spines, 1.04-1.66 μm long.

Habitat: Sandy or lateritic soil in dry lowland dipterocarp forests, fruiting during rainy season between May-June.

Distribution: North and North Eastern areas of Thailand.

Table 4. Basidiospore characteristics of Thai *Astraeus* collections

Type	Specimen code	Spore diam. (μm)	Mean spore diam. (μm)
1	ASTO_02	8.75-12.50	10.63±1.11
	ASTO_03	8.75-12.50	10.56±1.37
	ASTO_04	8.75-12.50	10.63±1.31
	ASTO_05	8.75-13.30	11.08±1.47
	ASTO_07	9.50-15.20	11.50±1.69
	ASTO_15	9.50-13.30	10.93±1.21
	ASTO_27	9.50-13.30	11.12±0.93
	ASTO_29	9.50-13.30	10.64±1.29
2	ASTO_01	7.50-12.50	10.94±1.21
	ASTO_06	9.50-13.30	11.28±1.20
	ASTO_08	9.50-13.30	10.64±1.29
	ASTO_09	9.50-15.20	12.26±1.44
	ASTO_10	9.50-13.30	11.78±1.17
	ASTO_11	9.50-13.30	11.12±1.12
	ASTO_12	9.50-13.30	11.69±1.66
	ASTO_13	9.50-13.30	10.93±1.05
	ASTO_14	11.40-15.20	12.54±1.14
	ASTO_16	7.60-13.30	10.55±1.44
	ASTO_17	9.50-15.20	11.12±1.54
	ASTO_18	9.50-15.20	11.21±1.62
	ASTO_19	9.50-13.30	11.40±1.51
	ASTO_20	9.50-13.30	10.93±1.21
	ASTO_21	7.60-11.40	10.17±1.12
	ASTO_22	9.50-13.30	11.31±1.30
	ASTO_28	9.50-13.30	10.45±1.31
	ASTO_30	7.60-13.30	10.64±1.43

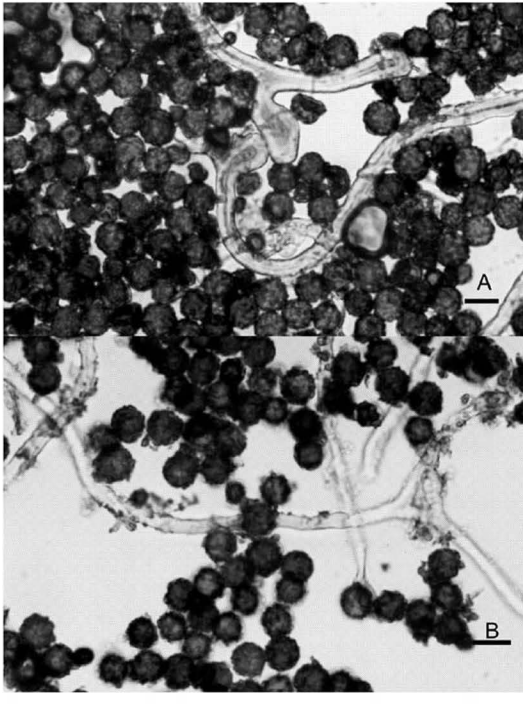

Fig. 2 Light microscopy of mature basidiospores. A: *Astraeus hygrometricus*. B: *Astraeus odoratus*. Scale bars = 10 μm.

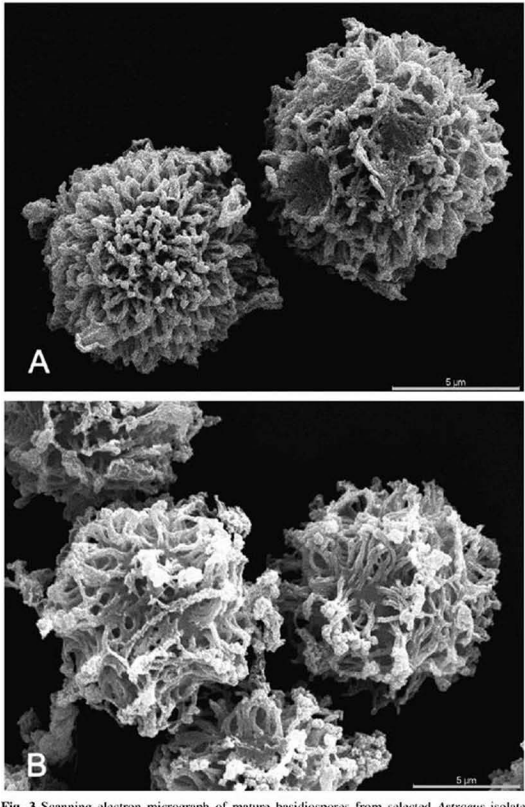

Fig. 3 Scanning electron micrograph of mature basidiospores from selected *Astraeus* isolates displaying different spine morphology A: very dense with rounded, narrow, tapered spines, occasionally coalesced of *Astraeus hygrometricus* .B: less dense, long, narrow, coalesced spines of *Astraeus odoratus*.

Table 5. Structural differences between *A. hygrometricus* and *A. odoratus*

Feature	*A. hygrometricus*	*A. odoratus*
Outer peridium	Granulate	Smooth with few soil particles
Number of rays	5-12 acute rays	3-9 broad rays
Fresh odour	Slight	Strongly reminiscent of moist soil
Spore size	8.75-15.2	7.5-15.2
Fruiting season	May-August	May-June

Discussion

The first description of *A. hygrometricus* was made by Persoon (1801 as *Geastrum*) and it was confirmed as a distinct taxon by Morgan (1889) with further descriptions by Lloyd (1902), Coker & Couch (1928) and Cunningham (1944). It is apparently worldwide in distribution and usually occurs in sandy soil in coniferous or mixed woods as previously reported from North America, Argentina and Mexico, Europe, Australia, China and India (Lloyd 1902, Cunningham 1944, Nouhra & Toledo 1998). All authorities stress the strongly hygroscopic traits and the globose, densely spiny spores ranging from 7.5-12.0 μm in diameter. However, in the current study, the spore size was found to be greater than generally given for this species, varying between 8.75-15.20 μm. in diameter. It was also noticed that the Thai specimens have a slight odour when fresh. The newly proposed taxon, *A. odoratus*, can be distinguished from *A. hygrometricus* by the strong odour when fresh, smooth surface with few adhering soil particles and the number of rays. In addition, this species occurs frequently during May-June whereas *A. hygrometricus* can be found in Thailand over a more prolonged period from May-August (Table 5).

Kope & Fortin (1990) suggested that differences in basidiospore types revealed under SEM could be used to segregate *Pisolithus arhizus* (Pers.) Rausch. into biological species. Although the basidiospores of the newly proposed taxon were identical in morphology with those of *A. hygrometricus* when viewed under the light microscope, observations using SEM showed that basidiospores of *A. odoratus* can be distinguished from those of *A. hygrometricus*. The spore ornamentation of the former is characterized by moderately dense, longer, narrower, coalescent spines.

There is still the possibility that *A. hygrometricus* as currently understood is a complex consisting of species and several approaches similar to those used for analysing *Pisolithus* may allow separation of further distinct taxa within the *A. hygrometricus* group. Thus *A. hygrometricus* var. *koreanus* Stanek, described from a mountainous area near Chon-Dzin, Korea and differing in its smaller size and pale colour may prove to be synonymous.

The differences, apart from size between *A. hygrometricus* and *A. pteridis* (= *Geastrum hygrometricum* var. *giganteus* Lloyd) the only other species in the genus need to be quantified. Further, much work needs to be done before the number and separation of the species are completely understood.

Acknowledgements

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Taxonomic reconsideration of *Epicoccum nigrum* and *Phoma epicoccina* based on DNA sequences and morphological observations

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Abstract—The species *Epicoccum nigrum* and *Phoma epicoccina* were compared morphologically and molecularly. Some isolates originally identified as *E. nigrum* developed a "*Phoma*-like" pycnidial state. A phylogenetic tree derived from the sequences of the ITS region of a group of isolates from both species and other *Phoma* related species revealed that all the strains form a monophyletic group, and no further segregation was found among the isolates analyzed. Based on this data, together with morphological observations, we propose formally the synonymization of both species.

Key Words—ITS, phylogeny, rDNA, synanamorph, taxonomy

Introduction

Epicoccum nigrum Link is a dematiaceous hyphomycete characterized by pulvinate stromata, black sporodochia, which produce granular-verrucose conidia globose to pyriform (Schöl-Schwarz 1959). It is a very polymorphic fungus, showing large differences across isolates in a range of characters, including pigmentation, conidial size, presence of sectors in pure culture, secondary metabolite production and others. This large variability has been evidenced using diverse techniques available for the genotypic and phenotypic characterization of fungal strains (Arenal et al. 1999; 2002; Kilpatrick & Chilvers 1981). *E. nigrum* is a common saprophyte that can be isolated from a broad diversity of substrates (Arenal et al. 1999; Domsch et al. 1993; Ellis 1971).

Phoma epicoccina Punith., Tulloch & Leach was described from seeds of *Dactylis glomerata* (Punithalingam et al. 1972). It was characterized by the presence of an associated "*Epicoccum* state", morphologically

indistinguishable from a typical *E. nigrum*. *Phoma*-like species are a wide spread coelomycetes (Boerema et al. 2004) and sometimes show hyphomycetous synanamorphs assigned to different genera (Grondona et al. 1997). *Phoma epicoccina* was included by Boerema (1997) within section Peyronaella (Goid. ex Togl.) Boerema, together with other *Phoma* species developing multicellular chlamydospores.

Previous molecular data, derived from the sequences of the ITS region of nine strains initially characterized as either *E. nigrum* or *P. epicoccina*, seggested that both were the same biological species (Arenal et al. 2000). However, the fact that no isolate of *E. nigrum* developed the *Phoma* state prevented us to reduce both species as synonyms. More recent morphological observations have revealed that some of the strains initially characterized as *E. nigrum* may, under some conditions, develop a *Phoma* state as well. Based on these observations, firstly reported in this study, and on the molecular data described above, wich are subjected to some reanalyses, we propose formally in this work the synonymization of both species.

Materials and Methods

The *P. epicoccina* and *E. nigrum* strains used in this study and the GenBank accession numbers of their ITS sequences are shown in Table 1.

Table 1. Strains of *Epicoccum nigrum* and *Phoma epicoccina* studied

Strain code*	GenBank accession number	Geographic origin	Substrate
EN22	AF149927	Spain	Fruiting body of *Resupinatus applicatus*
EN27	AF149928	Spain	Twigs of *Thymus mastichina*
EN33	AF149939	Ecuador	Leaves of undetermined bush
EN34	AF149930	Ecuador	Leaves of undetermined bush
EN5	AF149926	Colombia	Bark of *Vismia* sp.
PE20002	AF149931	United Kingdom	Seedlings of *Picea sitchensis*
PE20003	AF149932	United Kingdom	Seedlings of *Picea sitchensis*
PE20028	AF149933	United Kingdom	Emulsion paint of PVA
PE20044	AF149934	The Netherlands	Human toe nail

EN= *Epicoccum nigrum*. Strains isolated by the authors.
PE= *Phoma epicoccina*. Strains from the CECT (Colección Española de Cultivos Tipo, Valencia, Spain), replicated from cultures of the International Mycological Institute (Egham, Surrey, UK). PE20002 and PE20003 are monosporic isolates derived from the same parental strain, IMI276465; PE20028 and PE20044 are monosporic isolates from strains IMI178513 and IMI331914, respectively. All the strains are preserved in the collection of fungal cultures at CIBE-MSD.

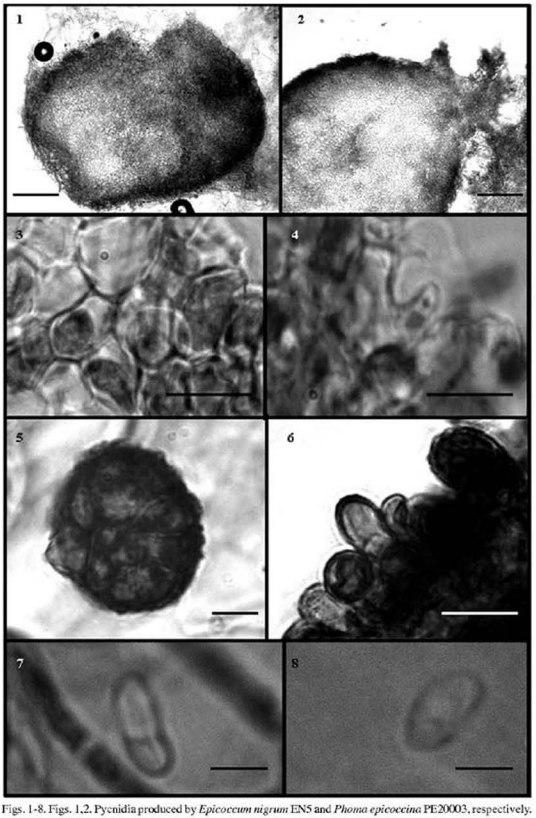
All the isolates were grown at 22ºC under 12 hour day light/dark cycle, on potato dextrose agar (PDA). Two replicates of each *E. nigrum* strain were grown on the same culture media and temperature/humidity

conditions, but with alternate cycles of 12 hour UV irradiation and darkness. After 14 days their morphological features were studied and, after sporulation, slides were prepared on lactophenol cotton blue and observed under a Leitz Diaplan microscope. Photographs were made with an Olympus DP-12 digital camera system incorporated to the Leitz microscope.

Phylogenetic analysis of the aligned sequences was performed by the maximum parsimony method using the heuristic search algorithm of the Phylogenetic Analysis Using Parsimony (PAUP4.0) software (Swofford 1998). Heuristic search was performed with simple addition of sequences and TBR branch swapping, with MaxTrees set to 100. All characters were unordered and equally weighted, with gaps treated as missing data. The trees were rooted with the ITS sequence of *Alternaria alternata* (AF314569) as outgroup. The data were resampled with 1000 bootstrap replicates (Felsenstein 1985).

Taxonomy and Discussion

All the *E. nigrum* and *P. epicoccina* strains were sporulated after 14 days of incubation. All of the isolates showed the macroscopic phenotypic characteristics (mycelium growth and pigmentation) described for both species, being white to pinkish tints and velvety in *P. epicoccina* and yellow to orange, orange to red or pink in color, greenish-brown and woolly to cottony, in *E. nigrum*. The conidiogenesis and conidia showed the morphological features described for both species (Schöl-Schwarz 1959; Punithalingam et al. 1972). Three of the strains initially labeled as *E. nigrum* (EN5, EN33 and EN34), when grown under UV light, developed also a pycnidial *Phoma*-like state, together with the *Epicoccum* state. (Figs. 1-8). The same test had been previously carried out, but at that time these *Epicoccum* strains failed to produce any *Phoma* state at all (Arenal et al. 2000). There is not any obvious explanation for the different results obtained now, although it looks that most of the studies carried out before were done under a *Phoma* point of view. Thus, in former studies (Monte et al. 1990), monosporic cultures from *Epicoccum*-like conidia from *P. epicoccina* (PE isolates) were able to develop mycelium with pycnidia and *Phoma*-like conidia. However, little attention was given to the fact that the *Epicoccum*-like conidia from the PE isolates could originate *Phoma* states, since it was considered as obvious. All the *Epicoccum* states produced by *P. epicoccina* strains were indistinguishable from those produced by *E. nigrum* isolates, as described by Punithalingam et al. (1972), and all the *Phoma*-like states produced by the three *E. nigrum* isolates were indistinguishable from those produced by the *P. epicoccina* strains. The *Epicoccum* mature conidia were globose to pyriform, 10-45 μm in diameter, with reticulated surface, rough and verrucose to warty, and light to dark brown (Fig. 5).



Figs. 1-8. Figs. 1,2. Pycnidia produced by *Epicoccum nigrum* EN5 and *Phoma epicoccina* PE20003, respectively. Fig. 3. Pycnidial wall cells of EN5. Fig. 4. Inner pycnidial line of EN5. Fig. 5. Conidium produced by EN5. Fig. 6. Immature sporodochium in EN5. Figs. 7,8. Pycnidial conidia produced by EN5 and by PE20003, respectively. Bars 1,2= 100 μm; bars 3, 4, 6-8= 5 μm; bar 5= 10 μm.

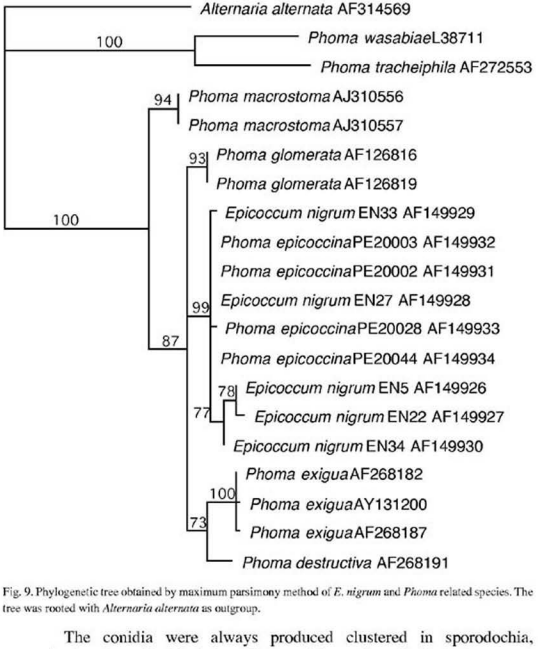


Fig. 9. Phylogenetic tree obtained by maximum parsimony method of *E. nigrum* and *Phoma* related species. The tree was rooted with *Alternaria alternata* as outgroup.

The conidia were always produced clustered in sporodochia, appearing in culture like black dots. The pycnidia produced by both *Epicoccum* and *Phoma* strains were scattered, globose to pyriform shaped, 90-150 μm in diameter, hyaline, reddish to light brown and ostiolate, sometimes with conspicuous neck (Figs. 1 and 2). The conidia produced in the inner line of the pycnidia were unicellular, 5-10 x 2-4 μm, hyaline and cylindrical to ellipsoid shaped (Figs. 7 and 8). To our knowledge, this is the first report of pycnidial development in isolates originally identified as *E. nigrum*.

As previously reported, the sequencing of the ITS region supports that *P. epicoccina* and *E. nigrum* could represent the same biological species (Arenal et al. 2000). The maximum nucleotide divergence found between

isolates of both species collected from very different geographical locations was very low; 1.5% for the entire ITS region, 2.8% for the ITS1 and 3% for the whole region sequenced. Furthermore, two strains identified as *P. epicoccina* (PE20003, PE20044) and two others as *E. nigrum* (EN27, EN33) showed identical sequences.

The closest matches of these sequences found in GenBank were with sequences from other *Phoma* species (Arenal et al. 2000). Some of those ITS sequences were retrieved and aligned together with our *E. nigrum* /*P. epicoccina* strains, and the alignment was subjected to parsimony analyses. One of the most parsimonious trees derived from this analysis is shown in Fig. 9. The complete alignment was 517 characters, with 353 constant characters, 89 parsimony-informative positions and 75 parsimony-uninformative positions. The tree length was 236 steps, with CI = 0.846, HI = 0.136, RI = 0.853, and RC = 0.738. The phylogenetic analysis revealed that all the strains of *E. nigrum* and *P. epicoccina* fall into a monophyletic clade supported by high bootstrap values (99%). The strains labeled as *E. nigrum* or *P. epicoccina* appeared intermingled within this branch, without any further segregation. The only subgroup that was observed in this clade, which is supported by modest bootstrap (77%) contains three *E. nigrum* isolates, two of wich developed a *Phoma* state (EN5 and EN34) and one that did not (EN22). *Phoma glomerata*, *P. exigua* and *P. destructiva* appeared as the closest species to the *E. nigrum*/*P. epicoccina* cluster, all of them grouped together in a single clade with 87% bootstrap support.

In summary, we conclude that the molecular and morphological data presented in this work clearly evidence that *E. nigrum* and *P. epicoccina* are synonyms. Therefore, according the International Code of Botanical Nomenclature (Greuter et al. 2000), the name *E. nigrum* should be used to name all the isolates showing an *E. nigrum* state, regardless of the presence of a *Phoma* state.

Epicoccum nigrum Link, in *Mag. Ges. Naturf. Fr., Berl.* 7: 32. 1816
= *Epicoccum purpurascens* Ehrenb., in *Flora Berol.* 136. 1818
= *Epicoccum purpurascens* Ehrenb. ex Schlecht, in *Flora Berol.* 2: 136. 1824
= *Phoma epicoccina* Punith., Tulloch & Leach, in *Trans. Br. mycol. Soc.* 59 (2): 341. 1972

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Notes on indoor fungi I: New records and noteworthy fungi from indoor environments

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Abstract—Seven fungal species isolated from indoor environments, including four new records for the United States and three other noteworthy taxa, are described and illustrated. The new U.S. records are: *Ascotricha erinacea*, *Sporoschisma saccardoi*, *Stachybotrys microspora*, and *Stachybotrys nephrospora*. The noteworthy fungi are *Ascotricha chartarum*, *Memnoniella echinata*, and *Zygosporium masonii*.

Key words—ascomycetes, hyphomycetes

Introduction

Indoor fungal contamination has recently become a major issue for homeowners, building owners, business owners and the insurance industry, because public awareness of potential detrimental effects of indoor fungi has increased dramatically in the last decade. Morgan-Jones and Jacobsen (1988) described several darkly pigmented moulds, including two new species of *Cladosporium*, associated with biodeterioration of carpet, plaster and wallpaper from hotels in the southern U.S. Samson et al. (2000) described many common airborne fungi from indoors. Flannigan et al. (2001) provided a list of common and important indoor fungi, with brief descriptions. We note that fungi identified from indoor environments are rather diverse and over 600 species have been identified in our laboratory. Over the last fifteen years, we have isolated many unusual fungi from air and from water-damaged building materials. This paper reports four species new to the USA. *Ascotricha chartarum*, *Zygosporium masonii*, and an isolate of *Memnoniella echinata* producing dimorphic conidia are also described.

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Materials and methods

The isolates used in this study were isolated and purified from samples submitted to P & K Microbiology Services for fungal identification and enumeration. The isolates were grown on 2% Malt Extract, 1.5% Agar (MEA), Corn Meal Agar (CMA), or dicholran glycerol agar (DG18) for 1 to 2 weeks at 25 C. Dimension ranges given are based on 30 measurements. If fewer than 30 structures were available, the actual number measured is noted. United States postal abbreviations of State names are used in the distributional records of fungi reported from the United States.

Taxonomy

Ascotricha chartarum Berk. (Figure 1)

Anamorph: *Dicyma ampullifera* Boulanger

Description: Colonies slow-growing, 25-35 mm diam in 36 days on MEA at 25C, dark blue green with a yellowish edge; aerial mycelium yellow. Perithecia dark brown to black, pear-shaped, ostiolate, 94-150 × 60-105 µm (mean = 123 × 87 µm), developing olivaceous to black, geniculate, rigid, erect setae with thin-walled vesicles at geniculate nodes. Asci 8-spored, cylindrical, thin-walled, deliquescing after ascospores mature. Ascospores uniseriate, dark brown to black when mature, smooth, discoid with a distinct equatorial slit, 5.9-8.4 × 4.2-6.4 µm (mean = 7.5 × 5.5 µm).

Conidiophores straight, stiff, profusely branched, up to 1 mm long, 3.5-5.5 µm wide with pale, thin-walled vesicles at the bends. Conidiogenous cells lateral and terminal, cylindrical, sympodial, developing conidia on denticles. Conidia lightly rough, subspherical to ellipsoidal, colorless when young, becoming light brown when mature, 4.5-7.4 × 2.4-3.7 µm (mean = 5.4 × 3.5 µm).

Habitat: plant materials, paper, linoleum, plaster, cardboard, cloth, cork, skin, seeds, soil, and lignum (Hawksworth 1971, Hanlin 1990). Culture 030130-012 was isolated from water-damaged drywall and wood structures from FL.

Distribution: Brazil, China, Denmark, England, France, Germany, India, Italy, New Zealand, Tanzania and USA (FL, NH., MA) (Hawksworth 1971). It was found in indoor environments in AZ, NY and SC.

Remarks: the fungus is a cellulolytic saprobe. It also causes maxillary sinusitis (de Hoog et al, 2000). *Ascotricha chartarum* var. *orientalis* Castell.

& Jacon. was considered to cause dermatoid infection (paracladiosis) (Hawksworth 1971). A dried specimen derived from the culture 030130-012 is deposited in BPI and CBS. Living cultures are deposited in ATCC.

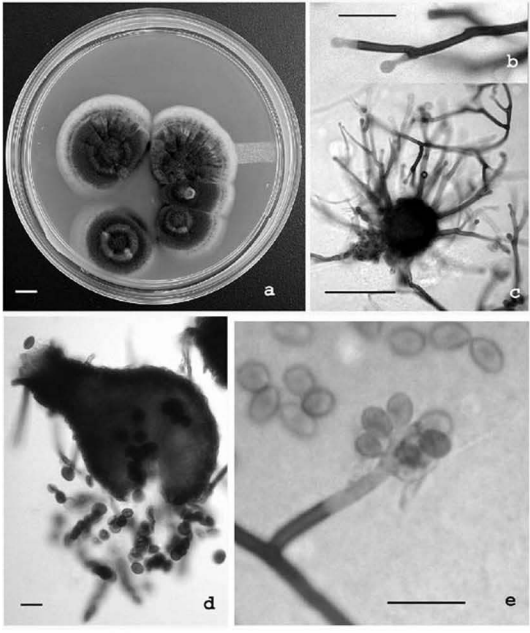

Figure 1. *Ascotricha chartarum* (030130-012). a. Colonies on MEA after 36 days. b. Appendages. c. Perithecia and appendages. d. Perithecia and ascopsores. e. Conidiophores and conidia. Bars: a = 10 mm, b, d, e = 10 μm, c = 100 μm, respectively.

Ascotricha erinacea Zambett. (Figure 2)

Description: Colonies 17-22 mm diam in 7 days on MEA at 25C, green with a raised white center and wavy light brown edge. Conidiophores 44-92 × 2.2-4.8 µm (mean = 60 × 2.9 µm). Conidia 3.7-5.9 × 2.4-3.7 µm (mean = 5.1 × 3.5 µm). Perithecia black, developing among the conidiophores, ostiolate, globose to subglobose, 99-165 × 66-115 µm (mean = 135 × 88 µm). Terminal hairs erect, brown to black, geniculate, often dichotomously and trichotomously branched, remotely septate, narrowing to pointed apices, 4.5-8 µm wide at the base. Lateral hairs sparse or absent, similar to the terminal hairs. Asci 33-72 × 7.8-11 µm (mean = 57 × 8.6 µm). Ascospores 9.4-13 × 5.6-7.4 µm (mean = 12 × 6.5 µm).

Habitat: on paper of water-damaged drywall and wood structures. Culture 021219-026 isolated from indoors in MN.

Distribution: France (Hawksworth 1971) and USA (AZ MD, MN, and NY). It is a new record for the U.S.A.

Remarks: this species was isolated from paper by Zambettakis in 1955. Hawksworth described it in his revision of the genus *Ascotricha*. It appears to be a rare species. We have occasionally encountered it on water damaged paper products or on wood. A dried specimen derived from culture 021219-026 is deposited in BPI and CBS. Living cultures are deposited in ATCC.

Memnoniella echinata (Riv.) Galloway (Figure 3)

Description: Colonies 34 – 37 mm in 21 days on MEA at 25C, gray to dark gray, granular, radially sulcate with irregular edge, reverse light brown. Conidiophores unbranched, colorless at first, olivaceous later, 42-103 µm (mean = 78 µm) long and 2.8-5.6 µm (4.5 µm) wide, branched portion 22-53 µm (mean = 39 µm) smooth to minutely rough. Phialides unicellular, obovoid or ellipsoidal, pale olivaceous, smooth, of two kinds: (1) phialides producing globose conidia 5.6-9.3 × 2.2-3.9 µm (mean = 8 × 3.4 µm), in groups of 3 to 8 (mostly 5); (2) phialides developing oblong conidia 7-9.3 × 3.2-4.3 µm (7.8 × 3.7 µm) in groups of 3 to 4 (mostly 3); terminal, or intercalary because of extension of phialide. Conidia of two kinds: (1) globose, in dry basipetal chains, colorless and smooth-walled at first, later dark olivaceous, coarsely warted, subglobose to globose, 3.2-4.4 × 2.4-4.3 µm (mean = 3.8 × 3.6 µm); (2) oblong to pyriform, smooth to rough, dark olivaceous 5.7-9.3 × 3.3-4.7 µm (mean = 7.9 × 3.7 µm).

Habitat: Indoor air, paper products, textiles, wood (*Hevea brasiliensis*) (Ellis 1971, Florence *et al.* 1998), ginger (Srivastava *et al.* 1998). Culture 021220-067 was isolated from MI.

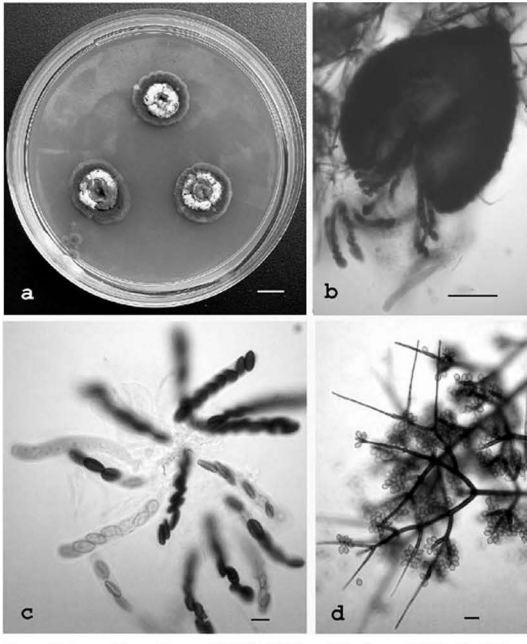

Figure 2. *Ascotricha erinacea* (021219-026). a. Colonies on MEA after 7 days. b. Perithecia. c. Asci and ascospores. d. conidiophores and conidia. Bars: a = 10 mm, b = 40 μm, c, d = 10 μm, respectively.

Distribution: cosmopolitan, but mainly from tropical areas (Domsch *et al.* 1993, Ellis 1971, Jong and Davis 1976). We have identified this fungus from

indoor environments in U.S.A. (AZ, CA, FL, HI, IL, LA, MN, NJ, NY, OH, PA, SC, TN, TX, VA, WA, and WI). It was found more often in NJ, NY, and PA.

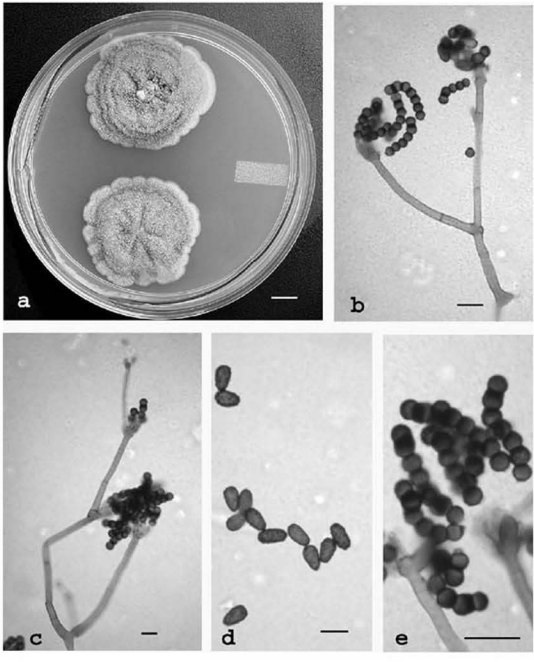

Figure 3. *Memnoniella echinata* (021220-067). a. Colonies on MEA after 7 days. b. Conidiophore with 2 kinds of conidia. c. Proliferation of conidiophore through phialides. d. *Stachybotrys*-type conidia. e. *Memnoniella*-type conidia. Bars: a = 10 mm, b-e = 10 μm, respectively.

Remarks: the fungus produces several mycotoxins: trichodermol, trichodermin, dechlorogriseofulvins, memnobotrins A and B, memenoconol, memnoconone (Jarvis *et al.* 1996). A dried specimen derived from the culture 021220-067 is deposited in BPI and CBS. Living cultures are deposited in ATCC.

Sporoschisma saccardoi Mason & Hughes apud Hughes (Figure 4)

Holomorph: *Melanochaeta hemipsila* (Berk. & Broome) Müller, Harr et Sulmont, Revue de Mycologie 33: 377 (1969).

Description: Colonies restricted, 5 - 8 mm diam in 7 days on MEA at 25C, dark brown to black, covered with grayish aerial mycelium, velvety; reverse dark brown. Conidiophores with terminal phialides, dark brown, smooth, erect, straight, solitary or in groups of 2 to 4, developing very sparsely at center, more profusely at the edge of colonies, 130 – 200 µm (mean = 170 µm) long, 1-septate. Venter of phialides somewhat inflated, 14-20 µm wide (mean =15.6 µm) at inflated portion, apices 11-14 µm wide (mean = 13.5 µm), base 8.4-14 µm wide (mean = 10.8 µm). Conidia in long chains, cylindrical with truncate ends, smooth, dark brown, very dark at the septa, majority 5-septate, the two end cells much shorter and lighter, 32-53 × 11-13 µm (mean = 43 × 11.5 µm), mean conidium length/width ratio = 3.78:1. Developing conidia can be observed through the phialide walls. Capitate hyphae 92-170 × 4.2-5.9 µm (mean = 135 × 5.5 µm), erect, straight, or slightly bent, 2-4-septate, solitary or in pairs, developed in mucilaginous envelopes, mixed with conidiophores; the tips of capitate hyphae are inflated, 7.4-13 × 7.4-13 µm (mean = 10 × 9.5 µm). This fungus sporulates poorly on MEA, but very well on CMA.

Habitat: on wood, especially wet or submerged (Goh *et al.* 1997). Culture 020703-043 was isolated from water-damaged wood structures indoors.

Distribution: Australia, Brunei, Canada, Italy, Hong Kong, Indonesia, Japan, Malaysia, South Africa, South America, Taiwan (Goh *et al.* 1997, Nag Rag and Kendrick 1975, Watanabe 2002), and USA (FL, IL). It is a new record for the U.S.A.

A dried specimen derived from the culture 020703-043 is deposited in BPI and CBS. Living cultures are deposited in ATCC.

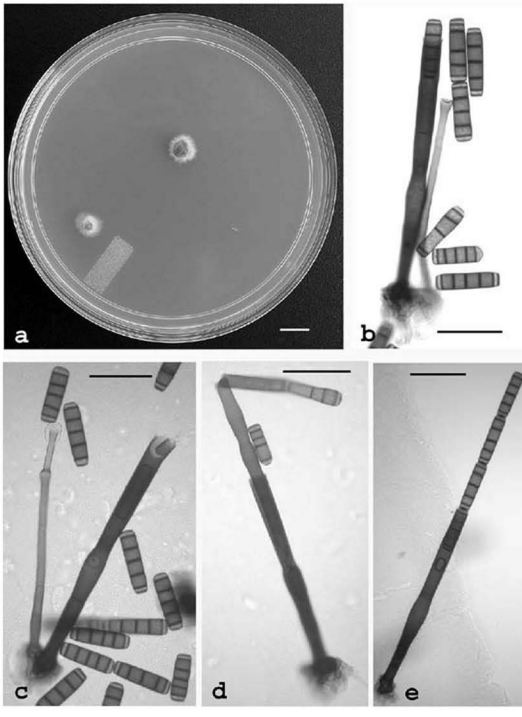

Figure 4. *Sporoschisma saccardoi* (020703-043). a. Colonies on MEA after 17 days. b. Conidiophore, conidia and capitate hypha. c. Percurrent extension of capitate hypha with conidiophore and conidia. d. Extension of conidiophore. e. Conidophores and conidia without vesicles. Bars: a = 10 mm, b-e = 40 μm, respectively.

Stachybotrys microspora (Mathur & Sankhla) Jong & Davis (Figure 5)

Description: Colonies slow growing 16.5 – 19 mm diam in 14 days on MEA at 25C, white with a pink tint; reverse brown; aerial mycelium white; the edges of the colonies irregular. In three weeks the colonies become light gray, still with a light pink tint; clear amber exudates start to develop. After four weeks, the colonies become dark gray to black. Conidiophores differentiated, single, determinate, simple, colorless and smooth, erect, straight, colorless to light brown, smooth, 44-92 × 2.2-4.8 µm (mean = 60 × 2.9 µm). Phialides 7-11 × 3.3-7.4 µm (mean = 8.8 × 4.6 µm) with conspicuous collarettes, in groups of 5-6. Conidia ellipsoidal, olivaceous, smooth to rough, 4.3 - 6.1 × 3.7 – 4.6 µm (mean = 5.4 × 4 µm).

Habitat: paper, soil, seeds, textiles and dead plants (Ellis 1971). It was isolated from paper of water-damaged drywall, and wallpaper, from WA.

Distribution: Canada, Cuba, India, Nigeria, and Pakistan (Ellis 1971). Culture 021020-084 is the first record for the U.S.A.

A dried specimen derived from the culture 021020-084 is deposited in BPI and CBS. Living cultures are deposited in ATCC.

Stachybotrys nephrospora Hansford (Figure 6)

Description: Colonies olivaceous to black, 37 – 42 mm diam in 22 days on MEA at 25C; reverse brown; aerial mycelium white; the edge of the colonies slightly irregular. Conidiophores differentiated, single, determinate, simple, occasionally branched, colorless and smooth, erect, straight or curved (flexuous), 60-150 × 2.5-3.3 µm (mostly 93.6 × 3.2 µm). Phialides obovoid or ellipsoidal, 8-10 × 4.2-6.6 µm (mean = 9 × 5 µm) in group of 2-4 (mostly 3) with conspicuous collarettes. Conidia reniform, colorless at first, becoming olivaceous to nearly black at maturity, smooth to rough, 8.3 – 13 × 5 – 8.3 µm (mean = 10.4 × 6.5 µm).

Habitat: on wood. Culture 021213-012 was isolated from water-damaged wallboard in Florida.

Distribution: Solomon Islands, India, Jamaica, Sierra Leone, Uganda (Ellis 1971). This species is newly recorded for North America. We have seen this fungus three times, from FL and AZ.

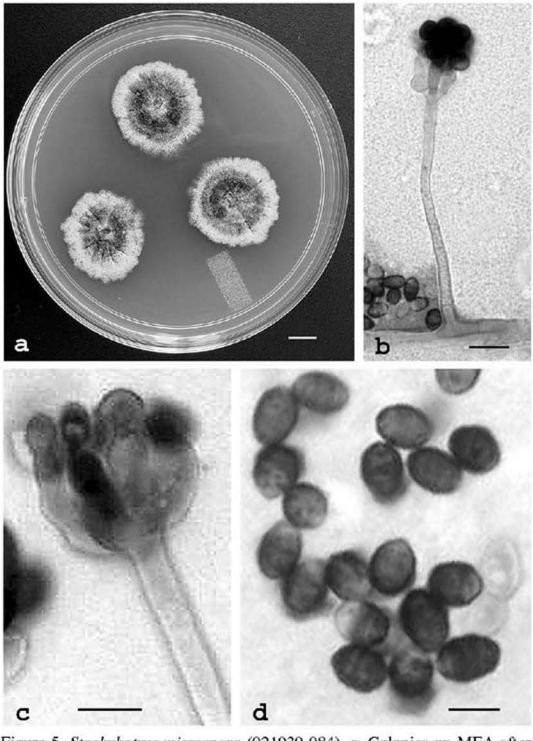

Figure 5. *Stachybotrys microspora* (021030-084). a. Colonies on MEA after 3 weeks. b. Conidiophore and conidia. c. Phialides with immature conidia. d. Conidia. Bars: a = 10 mm, b = 10 μm, c, d = 5 μm, respectively.

A dried specimen derived from the culture 021213-012 is deposited in BPI and CBS. Living cultures are deposited in ATCC.

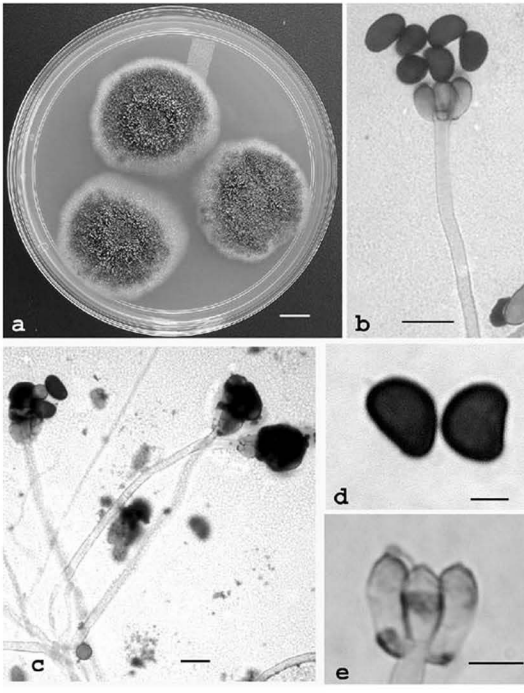

Figure 6. *Stachybotrys nephrospora* (021213-012). a. Colonies on MEA after 7 days. b Conidiophore and conidia. c. Conidia in slimy masses on conidiophores. d. Conidia. e. Phialides. Bars: a = 10 mm, b, c = 10 μm, d, e = 5 μm, respectively.

Zygosporium masonii Hughes (Figure 7)

Description: Colonies slow growing, 4 – 6 mm diam in 7 days on MEA at 25C, raised; aerial mycelium white; reverse brown. Conidiophores differentiated, single, 59-89µm long (mean = 71µm), erect, brown, smooth, with chains of up to 6 integrated vesicles (falces) and a colorless, sterile apical region, which is 15- 39 × 1.1-2.2 µm (mean = 23 × 1.8 µm) and terminates in a small knob. Vesicles (falces) thick-walled, dark brown, reflexed, 7.4-14 × 3.7-7.4 µm (mean = 9.8 × 5.6 µm), each developing 1-3 short, colorless distal conidiogenous cells. Conidiogenous cells monoblastic, discrete, determinate, ellipsoidal, curved, colorless, 3.7- 7.4 × 1.9-3.7 µm (mean = 5.9 × 3.0 µm). Conidia solitary, acrogenous, non-septate, ellipsoidal, colorless, smooth to rough, 3.9- 7.4 × 1.9-3.7 µm (mean = 6.5 × 2.9 µm). Conidiophores develop sparsely on MEA, but more profusely on DG18 medium.

Habitat: dry-wall, wall-paper indoors, air, soil, and dead plant materials. Culture 030201-011 was isolated from AZ.

Distribution: U.S.A. (AZ, southern CA, LA, TX), Assam, Australia, Ghana, Guinea, Hong Kong, India, Jamaica, Japan, Sierra Leone, Tanzania, Venezuela (Barron 1968, Ellis 1971, Matsushima 1975).

Remarks: *Zygosporium* is known to produce cytochalasin, which has cytotoxic effects on membrane function and the contractile mechanisms of cell movement and division (Griffin 1994). It prefers low water activity. A dried specimen derived from the culture 030201-011 is deposited in BPI and CBS. Living cultures are deposited in ATCC.

Discussion

Our observations suggest that *Ascotricha erinacea* is more common than *A. chartarum* in indoor environments, although *A. erinacea* was previously known from France (Hawksworth 1971). Anamorphs of both *Ascotricha chartarum* and *A. erinacea* develop well on MEA and DG18, but their teleomorphs usually fail to develop, or are produced sparsely on the same media. Freshly isolated cultures may develop the teleomorph, but subcultures normally do not produce ascomata.

A number of isolates of *Memnoniella echinata* (021220-067), *M. subsimplex* and *M. longistipitata* were observed to develop both *Stachybotrys*-type and *Memnoniella*-type conidia on the same colonies. *Stachybotrys*-type conidia are more often developed at the edges of colonies. Conidiophores of *Stachybotrys proliferata* Karandikar, Lulkarni & Patwardhan proliferate like those of *M. echinata* (isolate 021220-067) (Karandikar, Lulkarni, and Patwardhan 1992). *Stachybotrys*-type conidia in those isolates of *Memnoniella* spp. developing dimorphic conidia are all more or less oblong.

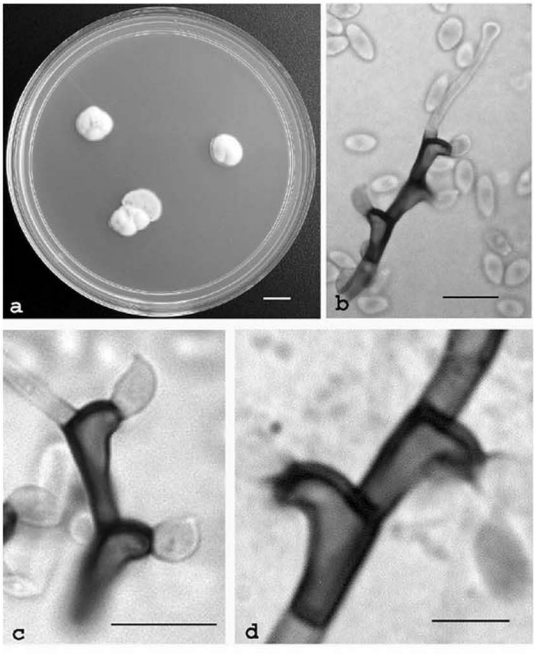

Figure 7. *Zygosporium masonii* (030201-011). a. Colonies on DG 18 after 7 days. b. Conidiophore and conidia. c. Vesicles, conidiogenous cells and conidia. d. Vesicle with collapsed conidiogenous cells. Bars: a = 10 mm, b, c = 10 μm, d = 5 μm, respectively.

But in *M. echinata* (021220-067) while some conidia were oblong or ovoid, a significant portion of the *Stachybotrys*-type conidia were pyriform. This

isolate could be a new taxon, but further studies are needed to determine its disposition.

Sporoschisma saccardoi (020703-043) grown on CMA produced a few conidiophores and capitate hyphae showing percurrent extension. Since the percurrent extension of conidiophores is one of important characters differentiating *Sporoschismopsis* from *Sporoschisma*, *Sporoschisma saccardoi* (020703-043) appears to be intermediate between *Sporoschisma* and *Sporoschismopsis*. This phenomenon raises two questions: 1) whether *Sporoschisma* and *Sporoschismopsis* should be treated as two separate genera; 2) whether the culture we examined should be a new species. To answer above-mentioned questions, further research is necessary. Watanabe (2002) suggested that the capitate hyphae of *S. saccardoi* are possibly in the process of developing into mature conidiophores, but we were unable to confirm this. During the percurrent extension of the capitate hyphae, they remained non-sporulating.

Both *S. microspora* and *S. nephrospora* are reported for the first time in the U.S.A. Without careful examination, both can be confused with *S. chartarum*. Conidia of *S. microspora* are similar to immature or smaller ones of *S. chartarum*. Ellis agreed with Mathur & Sankhla (1966) and treated it as a variety of *S. chartarum* (*Stachybotrys atra* var. *microspora*) (Ellis 1971). Jong and Davis (1976) elevated it to species rank. The comparative sequence analysis conducted by Haugland *et al.* (2001) showed that Jong and Davis were correct. Since the delimitation of *S. microspora* and *S. chartarum* is mainly based on conidial size, it is crucial to measure conidia precisely. Some conidia of *S. nephrospora* may not be reniform due to orientation or stage of development, which can make them appear similar to those of *S. chartarum*. The ecological significance of these closely related species in indoor environments and their effects on human health need more study.

Zygosporium masonii was commonly reported from Europe. In the U.S., it was a laboratory air contaminant in West Virginia, and was isolated from coastal sands of Oahu, Hawaii (Wang and Baker 1967). Recently it has been isolated from indoor environments in southwestern U.S.A. (AZ, CA, TX). Wang and Baker (1967) interpreted conidiogenesis in *Z. masonii* as being phialidic, with a *Sporothrix*-like synanamorph. However, Wang now considers the collarettes observed were probably collapsed conidiogenous cells and that the mode of conidium ontogeny is blastic (personal communication). Hughes (1951) did not consider the conidiogenous cells of this species to be phialides. Ellis (1971) treated them as monoblastic. In our isolates, monoblastic conidiogenous cells predominated. However one conidiophore was found with collarette-bearing conidiogenous cells where conidia can develop directly on vesicles. Each vesicle usually develops 2 conidiogenous cells (Wang and Baker 1967, Ellis

1971), but vesicles may bear 1-3 conidiogenous cells. This species was found in AZ, CA, TX where there is no previous published record. Conidiophores with conidia still attached were observed in an air sample. Meredith (1962) found that the conidia of *Z. oscheoides* Mont. are violently discharged. This may explain why species of *Zygosporium*, an unusual genus, are found in air samples from time to time.

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Notes on dictyostelid cellular slime molds from Taiwan (2): *Dictyostelium exiguum* and its ITS-5.8S rDNA sequences

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Abstract—During a survey of dictyostelid cellular slime molds in 2002, *Dictyostelium exiguum* was isolated from forest soils and was identified as new record for Taiwan. This isolate was morphologically examined and is illustrated in this text. The nucleotide sequences of internal transcribed spacer (ITS) regions and 5.8S of ribosomal DNA were also analyzed. The *D. exiguum* sequence lengths of ITS 1, 5.8S, and ITS 2 were 250 bp, 170 bp and 461bp, respectively.

Key words—dictyostelids, ribosomal RNA gene

Introduction

The taxonomic studies of dictyostelid cellular slime molds in Taiwan have been briefly reviewed in previous papers (Yeh & Chien 1983; Hagiwara et al. 1992; Lin & Yeh 1999; Fan & Yeh, 2001; Hsu et al. 2001; Fan et al. 2002; Yeh 2003). During a 2002 survey of forest soils in Nantou County, central Taiwan, *Dictyostelium exiguum*, was isolated. This was a new record for Taiwan. This isolate was examined and is illustrated here. Moreover, internal transcribed spacer (ITS) and 5.8S ribosomal DNA (rDNA) sequences were also determined. The examined specimens and pure cultures were deposited in the Mycology Laboratory, Department of Life Science, National Taiwan Normal University, Taipei, Taiwan, R. O. C.

Materials and Methods

Morphology

Soil samples were collected from forest floor from Nantou County, central Taiwan, in 2002. Five to ten grams of each soil sample were suspended in 50 ml of distilled water. A small amount of the suspension was spread over the surface of 0.1% lactose yeast-extract agar plates (Norberg 1971). The agar plates were then incubated in darkness at 25°C. When the fruiting structures developed, a sterilized needle was used to remove the spores from the sorocarps and transfer them to a fresh agar plate with a suspension of pre-grown *Escherichia coli*. The plate was examined for one or two weeks to follow the life cycle. Methods used for observations and measurements of this species follow Hsu et al. (2001). The taxonomic system of Hagiwara (1983) was followed for identification.

Genomic DNA extraction

Genomic DNA was extracted from spores using a variation of CTAB method described by Wang & White (1997) as adapted from Doyle & Doyle (1990). Seventy spores were collected from pure cultures of *D. exiguum* in a microfuge tube. After adding a small amount acid-washed sea sand, the spores were homogenized with a "microfuge pestle". Then 500 µl pre-warmed 2% CTAB extraction buffer (1.4 M NaCl, 100 mM Tris-HCl, pH 8.0, 20 mM EDTA, 2% PVP-40 w/v, 20 mM cetyl trimethyl ammonium bromide (CTAB), and 0.5%2-mercaptoethanol v/v) was added into the mixture before grinding more. The mixture was incubated for 10 min at 65°C to break the cell membranes. The tissue/buffer mixture was then emulsified with 500 µl dichloromethane:isoamyl alcohol (24:1) to separate protein from DNA before spinning in a microfuge for 2 min at room temperature. The top phase was transferred to a fresh microfuge tube and 300 µl of isopropanol was added to precipitate the DNA. After spinning for 2 min at room temperature, the supernatant was carefully poured off. Then 500 µl wash buffer was added and was left for 2 min. After spinning for 2-3 min, the supernatant was discarded. The pellet was resuspended in 35 µl~ 40 µl double distilled water. Two to 5 µl DNA solution was used for the PCR reaction.

PCR amplification

The oligonucleotide primers 18S-F (GAGAAGTCGTAACAAGGTATC) and 5.8S-R (GAATTATCGCAGTTTGCTACG) were used to amplify fragments containing ITS1. Another pair of oligonucleotide primers 5.8S-F (CGTAGCAAAC-TGCGATAATTC) and ITS4 (TCCTCCGCTTATTGATATGC) were used to amplify fragments containing ITS 2. A 100 l reaction mixture contained 1.0 mM of each primer, 2.5 units Taq DNA polymerase, 100 M each of the four deoxynucleotides, dATP, dCTP, dGTP, and dTTP, in a PCR reaction buffer (10 mM Tris-HCl, 1.5 mM $MgCl_2$, 50 mM KCl, 0.1 mg gelatin ml-1, pH 9.0), and 2 to 5 µl DNA. PCR products were amplified in a Perkin Elmer DNA Thermal Cycler using the following programme: 94°C for 5 min for denaturation of DNA, followed by 40 cycles at 94°C for 1 min, and 50°C for 1 min for primer annealing, then at 72°C for 2 min extension. PCR amplification templates containing no DNA, were carried out in every experiment as controls to test for contamination of reagents and reaction mixtures by non-sample DNA. Efficiency of amplification was monitored by running 7 µl of each reaction through a 2% agarose gel at 125 V for 3 h in Tris-borate-EDTA buffer (0.44 M boric acid, 0.44 M Trizma base, and 10 mM EDTA), a 100-bp molecular weight ladder (Gene RulerTM) was used as the size standard, stained with ethidium bromide, and visualized and photographed under ultraviolet light.

Sequencing of amplified DNA

Fragments containing ITS 1-5.8S and 5.8S-ITS 2 were amplified from the test cultures of *D. exiguum*. The PCR-amplified products were sequenced using an automated DNA sequencer following the manufacturer's protocols (ABI 3730 DNA Analyzer Applied Biosystems, USA).

Results

Taxonomic Description

Dictyostelium exiguum Hagiwara, Bull. Natn. Sci. Mus., Tokyo, Ser. B, 9(4):149, 1983. Fig. 1.

Sorocarps, solitary or gregarious, unbranched or sparsely branched. Sorophores colorless, sinuous, tapering from bases to tips, ranging from 1.8 to 6.8 mm; bases clavate, 25-36 μm in diam at a level 50 μm above the bottom; tip capitate or somewhat widened, 9.7-16 μm in diam at a level 50 μm below the tip. Sori white, globose, 50-200 μm in diam. Spores hyaline, ellipsoid, with conspicuous consolidated polar granules, mostly 5.5-7.7×2.2-3.3 μm. Pseudoplasmodia with radiate streams, ranging from 0.08-2.6 mm in diam, converging at single center or breaking up into several small aggregation centers.

Distribution: In forest soils of central Taiwan. Known from Nepal and Taiwan.

SPECIMEN EXAMINED: TAIWAN. Nantou County. Yeh *ZY. Tai 2002-1.*

Comment: This species is characterized by the small sorocarps, the thick clavate sorophore bases and the small spores. Taiwanese isolates examined in this study had somewhat larger spore size and longer sorophore than the type specimen (Hagiwara 1983).

Amplification and sequencing of ribosomal RNA gene

The total nucleotide sequence lengths of the products were 1042 bp containing partial 18S and 28S ribosomal RNA gene. They are listed in Table 1.

Discussion

Hagiwara (1983) first reported this species from Nepal. There, this species is found in the surface soil and leaf mold of open forests in the Gosainkund region and the Gokarna Forest (1,350 m). Hagiwara (1990) also obtained some isolates from open oak-laurel forest (1,910-2,150 m) on Mt. Phulchoki, Nepal. In Taiwan, *D. exiguum* was isolated only from Nantou County (ca. 1,000 m), located in central Taiwan. It was also found in the surface soil of a mixed forest of oak-pine and other broad-leaved trees. Nepal extends along the Himalayan range between the latitudes of 26°22'N and 30°27'N. The country has an area of 147,000 sq km. Two-thirds of the area is occupied by hills and mountains, which arise at about 60 m in the south to the crest of the Himalayas reaching over 8,000 m in the north. Average temperature ranges in Kathmandu (the capital) are from 2-20°C in January to 26-32°C in June. Dobremez (1972) divided the vegetation of Nepal into six bio-climate belts, namely, (1) tropical belt (below 1,000 m alt.): dominated by *Shorea*, *Terminalia*, *Eugenia* and *Cassia* spp. etc, (2) subtropical belt (1,000-2,000 m alt.): *Schima-Castanopsis* forest in the east and *Pinus* forest in the west, (3) temperate belt (2,000-3,000 m alt.): characterized by evergreen oaks, rhododendrons, conifers

etc with deciduous maples, (4) sub-alpine belt (3,000-4,000 m alt.): represented by *Abies*, *Betula*, *Acer* and *Sorbus* spp. etc. (5) alpine belt (4,000-5,000 m alt.) and (6) Nival belt (above 5,000 m alt.): dominated by rhododendron bushes, *Berberis* shrub and primroses herbs etc.

Taiwan is an island along the western edge of the Pacific Ocean. It is located across the Tropic of Cancer. Extending 394 km along its longest axis (25°20'N to 21°55'N) and stretching 140 km at its broadest transection, this island measures about 35,800 sq km. In the lowlands, the average temperature is about 26-30°C in summer and 15-23°C in winter. According to Hsieh et al. (1994), five principal forest vegetation belts can be distinguished along the altitudinal distribution, namely, (1) costal vegetation: represented by mangroves, sand-dune plants, and littoral forests, (2) lowland vegetation (below 500 m alt.): including *Ficus-Machilus* forests, secondary forests, and grasslands, (3) montane vegetation (500-2,500 m alt.): dominated by *Machilus-Castanopsis* forests, evergreen oak forests, beech forests, mixed coniferous forests, pine forests, and *Tsuga-Picea* forest, (4) subalpine vegetation (2,500-3,700 m alt.): characterized by *Abies* pure stands, *Berberis* shrub, some *Quercus* trees, and *Yushania* dwarf bamboo thickets, (5) alpine vegetation (3,500-3,900 m alt.): including *Rhododendron* and *Lycopodium* communities etc.

To date, there are two species of dictyostelids found only in Nepal and Taiwan: *D. clavatum* Hagiwara (Hagiwara 1992; Fan et al. 2002) and *D. exiguum*. As Taiwan is close to Nepal geographically and *D. exiguum* was isolated from similar habitats in these two sites, we suggest that *D. exiguum* as well as *D. clavatum* ultimately will be classified as a subtropical species after further investigation.

Current molecular approaches, especially sequences of ribosomal RNA gene (rDNA), are used to assist in studying taxonomic problems and in understanding the phylogenetic relationships among fungal groups (Zambino & Szabo 1993; Berbee et al. 1995; Moncalvo et al. 1995; Mitchell & Bresinsky 1999; Lohtander et al. 2000; Tuthill et al. 2001; Chou & Wu 2002; González et al. 2002; Hofstetter et al. 2002; Legrève et al. 2002; Liu et al. 2002). Ribosomal DNA sequences of *D. exiguum* based on ITS-5.8S gene totalled 881 bp and were registered in GenBank (accession number: AY170307).

Compared to the nucleotide sequences of the other dictyostelids registered in GenBank, *D. exiguum* was similar to *D. aureo-stipes* Cavender, Raper et Norberg var. *aureo-stipes*. There was 93% (286/306) identity between them after making sequence alignment in National Center for Biotechnology Information (NCBI) resources.

According to Ozaki et al. (1984), the ITS-5.8S rDNA sequences of *D. discoideum* Raper are 1078 bp and are longer than that of *D. exiguum*. The A+T contents revealed minor differences between these two species. In particular, the ITS 1- 5.8S- ITS 2 for *D. discoideum* is 74%-57%-57% (Ozaki et al. 1984), for *D. exiguum* was 64%-50%-63% (Table 1). With the development of additional dictyostelid rDNA sequences, it will be possible to construct the phylogenetic relationships within this group.

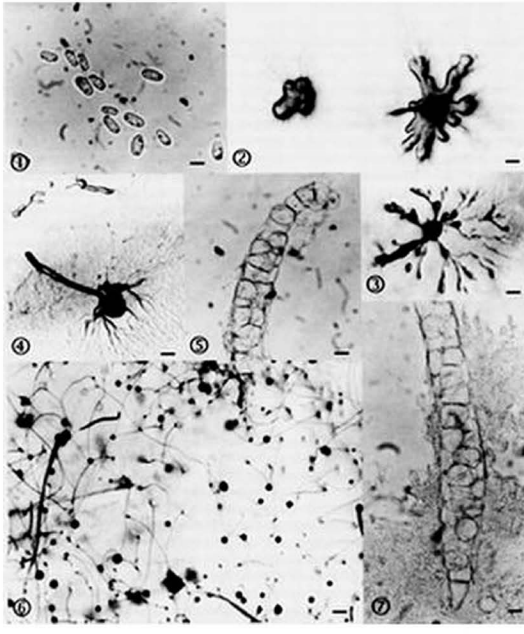

Fig. 1. *Dictyostelium exiguum* found in 2002 in forest soils of Nantou County, central Taiwan. 1. Spore, 2~3. Aggregation of myxoamoebae, 4. Pseudoplasmodia, 5. Sorophore tip, 6. Mature sorocarps, and 7. Sorophore base. Scale bars: 1 = 5 μm, 2 = 150 μm, 3 = 125 μm, 4 =150 μm, 5 = 5 μm, 6 = 180 μm, 7 =10 μm

Table 1. rDNA sequences of *Dictyostelium exiguum*

rDNA regions	Sequences
Partial 18S (44 bp)	GTCGTAACAA GGTATCGGTA GGTGAACCTG CCGATGGATC ATTA
ITS1 (250 bp) (G+C content =35.60%)	ACACAATGTC CATCCATTCA GTTTTGTAAA AGAGGAAAGA ATGAGCAATC ATTACTAGAA TCTTTATCAA TTCTTTATGG GTATACCCAT GTATACCTCT CAATTTTTTT ACGTTTATTC AACTATATTC TATGCCGCAC AACGCCAAAG ACATTGACTT AGGTCTTGTC TAAGGCTAGG GGTGAGCATT TTCAATGACT CCAGTGTTTC TCTTTCATAG AGATCCTACT GAAATTGTCA AAGAAAGCAT
5.8S (170 bp) (G+C content =50%)	AGGCACATTT GGTCGATACC TCGGCTCTCG AATCGACGAA GACCGTAGCA AACTGCGATA ATTCACTTGA ATTGTAGCGT TTACCGCGAT AGTCGAACTG TTGAACGCAC ATGATGACCG ACTGTCCCTT GCAAAAGGGG CAGCTAGGCC ACTCTCGTTT GAGAGACCTT
ITS2 (461 bp) (G+C content =36.88%)	TTCTCTATCT CTCATACTAA TTTATTATTA GTTTAGGGGC GTTGTGAATG ATGATGAACT AAGTCTGCAT ATATACGGGT CATCATTCAT TAAATATTAT AGTCCAATGC AGTCCTTGAC TTACTACCAA TTGGTAAGAG GTCTTTGCTT AGACACGTTG ATCGCTGTCA CTTTAACTTG GTGGGACTAA CATAGTCTTT TGGAATTTTT CCATTTACCA TTCTTAAACT TTGAAAAGAG ACTCGCGAGC TTTGGCTTGC TTGAGACCTT GATTTGGTTT TGAAAGGTTT TGGGAGTATA GTTTCATTAA GGCCTTCGAG GTTTTCGTCA TTTATAAGGT TTGGATCTGT TGAGCCGTGT CTTTTGAGC AAGTCTCTTG GCTTTGGTTT TAGTATTTTC TATGGTTCGA CTAATCAAAG ACTAATTCAA AAGTATCAAG TCCCCTGTGG ACTTTGATCA T
Partial 28S (117 bp)	GTAACTCTTG AAGTCGTTAA AGGCTTTCTC AAGATACATC TTTTTGAAGG CTTTCTCAAG ATACATCTTT TTGAATTGGC AAAACTTAAA ACTTAATTAT AGGTCATCAG ACGAGAG

Acknowledgments

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A new and unusual species of *Inocybe* (Inosperma clade) from tropical Africa

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Abstract—A new African agaric, *Inocybe misakaensis*, is described from tropical dry Miombo woodlands in Zambia. *Inocybe misakaensis* lacks pleurocystidia and has smooth yellowish brown basidiospores, fusiform to fusiform-rostrate cheilocystidia, and necropigmented basidia. Phylogenetically, *I. misakaensis* is sister to section *Cervicolores* in the Inosperma clade with strong bootstrap support based on RPB2 nucleotide sequence data. Despite the possession of necropigmented basidia, *I. misakaensis* is related to *I. calamistrata* and allies. A taxonomic description, illustrations, and the phylogeny of this new paleotropical species are presented.

Keywords—*Agaricales, Cortinariaceae*, phylogeny, RNA polymerase II, taxonomy

Introduction

Several new species of *Inocybe* (Fr.) Fr. have been described recently from Zambia and Cameroon (Buyck & Eyssartier 1999; Watling 2001). Here we propose as new, *I. misakaensis sp. nov.*, a species collected in native Miombo woodlands, a seasonally burnt tropical habitat that includes ectomycorrhizal caesalpinioid legumes of the *Fabaceae* and *Uapaca* of the *Uapacaeae* in the Copperbelt Province of Zambia (Högberg & Piearce 1986).

Anatomically, *I. misakaensis* exhibits basidia that fill with ochraceous contents and collapse after basidiospore discharge, similar to members of subgenus *Mallocybe* Kuyp. (Kuyper 1986). Kuyper referred to this character state as necropigmented basidia. For short, we use the term necrobasidia. Species of subgenus *Mallocybe* are distributed in temperate climates of the northern hemisphere and Australasia (Matheny, unpublished). Despite the anatomical similarity to subgenus *Mallocybe*, *I. misakaensis* would appear

to represent an undescribed species because of its peculiar cheilocystidia morphology and paleotropical distribution. To test this hypothesis, we sequenced a portion of RPB2 (Liu, Whelen, & Hall 1999; Matheny 2004) to determine the phylogenetic status of *I. misakaensis*. RPB2 encodes the second largest subunit of RNA polymerase II and is appropriate to investigate the phylogeny of the genus *Inocybe* and elements within it (Matheny 2004).

Methods and materials

Collections were made fresh by the second author. Color notations follow that of Ridgway (1912) and Munsell Soil Color Charts (1954). The holotype and paratype are deposited at E; the isotype is deposited at WTU. Herbarium abbreviations follow that of Holmgren, Holmgren, and Barnett (1990).

Sections of dried material were rehydrated in 3% KOH. Dimensions of basidiospore length, width and Q values (ratio of length to width) are presented in a range with outliers enclosed in parentheses, following that of Matheny, Aime, & Henkel (2003). Means of spore length, width, and Q values are italicized. Drawings were made with the assistance of a drawing tube. Spores were drawn at 2000x and basidia and cheilocystidia at 800x.

DNA extraction, polymerase chain reation (PCR) amplification, direct sequencing, and cloning follow that of Matheny et al. (2002) and Matheny (2004). Primers 6F and 7.1R (Liu, Whelen, & Hall 1999; Matheny 2004) were used for PCR amplification and direct sequencing of conserved domains 6 to 7 in RPB2. Clones of the undescribed *I. "serpentinocystis"* from Western Australia were sequenced according to the manufacture's protocol (TOPO TA Cloning and TOPO One Shot kits, Invitrogen, Carlsbad, California, USA). This was done because of a faint PCR band for this product. RPB2 accession numbers for the taxa used in this paper are available on GenBank. These include AY333309, AY333311, and AY333763-AY333778. Collections from which the sequences were obtained, and their herbaria, are provided in the appendix.

DNA sequences were aligned using Clustal X (Thompson et al. 1997). The alignment is available online at TreeBASE (M1620). The maximum likelihood (ML) criterion was used in PAUP* (Swofford 2003) to analyze the phylogenetic placement of *I. misakaensis* in relation to samples from other major clades of *Inocybe* (Matheny, 2004). The ML tree was recovered using a heuristic search with the "as-is" addition sequence. The underlying model of evolution used in the ML analysis was estimated by MODELTEST 3.0 (Posada & Crandall 1998, 2001). Bootstrap values (Felsenstein 1985) greater than 50% derived from a maximum parsimony (MP) analysis using a heuristic search are placed above branches to gauge support for branches in the topology. *Crepidotus (Crepidotaceae)* was used to root the tree because a more inclusive analaysis (Matheny 2004) places *Crepidotus* as sister to *Inocybe*.

Taxonomy

Inocybe misakaensis sp. nov. Figs. 1 a-c.

Metuloidae nullae. Basidia plena necropigmenti. Cheilocystidia 26-61 x 7-10 μm, fusiformes-rostellatae vel fusiformes-rostriformes vel clavatae, membrana tenui praedita. Sporae laeves, 8.0-10.0 x 5.0-6.0 μm. Pileus fibrillosus grosse, haud rimosus, alutaceus interdem cum disco fulvo. Lamellae adnatae, confertae. "Cinnamon-

Buff" vel "Clay Color", pallidae-fimbriatae. Stipe 3.0 cm x 3-4 mm, aequalis, pallidus vel subalutaceus, sericeus vel furfuraceus-fibrillosus, haud pruinosus. Dispersa, humi, cum arboribus leguminosis et *Uapaceae*. Holotypus hic designatus in E (Wat. 24837), isotypus in WTU.

Pileus 1.5-2.5 cm, plano-convex with low umbo, surface dry, coarsely fibrillose with scattered appressed clusters of fibrils radiating around the disc, not rimose, in age becoming diffracted-scaly around the center, lacerate; "Cinnamon-Buff" (light yellowish brown or 2.5Y 6/4) to "Clay Color" (light brown or near 10YR 5/3-5/4), at times with fulvous center. Lamellae narrowly adnate to sinuate, seceding; close, about 40 L, with several tiers of lamellulae; about 2-3 mm broad; "Cinnamon-Buff" to "Snuff Brown"; edges pallid, distinctly fimbriate. Stipe 3.0 cm X 3-4 mm, often longer than pileus diam, even, not bulbous, white to light brown, silky-fibrillose, at times scurfy, not pruinose, veil status not confirmed; odor and taste not recorded. No pigments dissolving in alkali mounts.

Basidiospores (7.5-) 8.0-*9.1*-10.0 (-10.5) x (4.5-) 5.0-*5.2*-6.0 μm; Q=(1.50-) 1.60-*1.71*-1.91 (-2.00) (n=41/2), smooth, subamygdaliform, elliptic, or occasionally subphaseoliform, apices bluntly pointed or obtuse; "Ochraceous-Tawny" or yellowish brown to strong brown (10YR 5/6-7.5YR 5/8); wall slightly thickened; germ pore absent, nondextrinoid; apiculus small and indistinct (Fig. 1a).

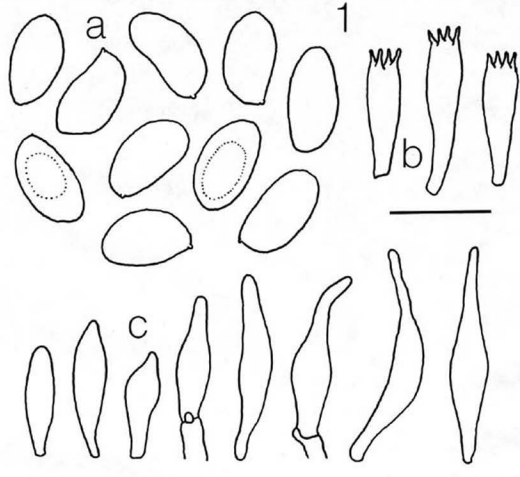

Fig. 1. Basidiospores (a), basidia (b), and cheilocystidia (c) of *Inocybe misakaensis* (holotype). The scale bar is equal to 10 μm for spores and 25 μm for basidia and cheilocystidia.

Basidia 27-35 x 8-10 μm (n=15/2), 4-stergimate, clavate to cylindrico-clavate, hyaline to necropigmented (Fig. 1b). Pleurocystidia absent. Cheilocystidia 26-61 x 7-10 μm (n=20/2), many fusiform with tapered subacute apices, at times rostrate, mixed with shorter clavate cells; thin-walled, hyaline to ochraceous or rusty colored, at times wall wrinkled or incrusted with pigment or cells with granular contents; lamellae edges sterile (Fig. 1c). Caulocystidia not observed; apex with sparse or thin superficial layer of hyphae, these cylindric, "Ochraceous-Tawny" in mass like the pileipellis, faintly incrusted if at all; tramal hyphae cylindric, very pale brown to yellowish with scattered golden refractive hyphae. Lamellar trama parallel, hyaline to very pale yellow, mostly cylindric but occasionally inflated, up to 16 μm diam, smooth, thin-walled; refractive hyphae present; cells unreactive in Melzer's solution. Pileipellis a cutis of interwoven or tangled to parallel cylindric hyphae, "Ochraceous-Tawny" in mass, hyphae mostly 3-10 μm diam, appearing smooth or at least not distinctly incrusted, at times filled with ochraceous-tawny refractive contents, thin-walled; tramal hyphae very pale brown to subhyaline in mass, cylindric; refractive hyphae present. Clamps present.

Scattered singly on soil among humus in seasonally burnt woodland (Miombo), including *Brachystegia, Gilbertiodendron, Isoberlinia*, and *Julbernardia* (caesalpinioid genera of *Fabaceae*) and *Uapaca* (*Uapacaceae*), Copperbelt Province, Ndola to Kitwe, Zambia (~15° S, 30° E), April.

Material examined: ZAMBIA: COPPERBELT PROVINCE: Misaka Forest Reserve, near Kitwe, 3 April 1991, leg. R. Watling, **Wat 24830** (E); same locality as above, on burnt patch, 3 April 1991, leg. R. Watling, **Wat 26674** (E); Ndola burn plots, seasonally burnt woodland, 2 April 1991, leg. R. Watling, **Wat 24837** (E, holotype; WTU, isotype).

Commentary: This species is characterized in part by the cinnamon buff to clay buff color, fibrillose texture, and presence of necrobasidia. However, the fusiform and rostrate cheilocystidia in combination with the paleotropical distribution in seasonally burnt woodlands represent unique characters. *Inocybe misakaensis* is probably ectomycorrhizal with caesalpinioid legumes such as *Brachystegia, Gilbertiodendron, Isoberlinia*, and *Julbernardia*, and/or *Uapaca* of the *Uapacaceae*. Indeed, a number of common Zambian trees in the *Fabaceae, Dipterocarpacae, Uapacaceae*, and *Proteaeae* have been reported as ectomycorrhizal (Högberg & Piearce 1986).

Inocybe misakaensis represents one of few depauperate (i. e., lacking pleurocystidia) species of *Inocybe* known from tropical Africa. *Inocybe aureoplumosa*, recently described by Watling (2001) from Cameroon, also possesses necrobasidia like *I. misakaensis* but morphologically is very different because of the brilliant reddish orange squamules on the pileus and stipe and subglobose spores. De Seynes (1897) described *I. erythroxa* from Gabon, which is colored like *I. aureoplumosa*, but lacks any ornamentation on the stipe and has elliptic spores. *Inocybe fastigiata* var. *brevispora* Heim described from Madagascar (Heim 1931) has a distinctly rimose pileus, short clavate cheilocystidia, and broadly elliptic to subglobose spores. Buyck & Eyssartier (1999) mention the resemblance of some endemic African species of *Inocybe* to *I. dulcamara*, but none of these have yet been described.

Phylogenetic results

A total of 18 accessions were used in this study. 730 positions were aligned in the data set that were sequenced from the 6 to 7 conserved domains of RPB2. The data set contains 242 parsimony-informative sites and 42 autapomorphic sites. The model best-fit to the RPB2 data is a Tamura-Nei model with equal base frequencies; rates of 10.07 and 5.73, respectively, for cytosine-thymine (CT) and adenine-guanine (AG) transitions and 1.00 for transversions; the proportion of invariable sites was estimated at 0.56 in addition to a gamma distribution of 3.38.

The ML analysis of RPB2 DNA sequences of *I. misakaensis* places it sister to section *Cervicolores* of the Inosperma clade (Matheny 2004) with moderately high (79%) bootstrap support (Fig. 2). The ML tree has a -ln L score of 4022.210. A single MP tree of 705 steps was recovered that differs from the ML tree only by the placement of a single branch, *I. adaequata* (Britz.) Sacc. (data not shown). In the MP tree *I. adaequata* is basal to *I. misakaensis* with poor (< 50%) bootstrap support.

The five major clades inferred in *Inocybe* by Matheny (2004) are represented in Fig. 2. *Inocybe misakaensis* is placed with strong bootstrap support (100%) in the Inosperma clade, which contains representatives of sections *Rimosae* (Fr.) Sacc. and *Cervicolores* Singer (Kuyper 1986) sampled from Europe, North America, and Australasia.

Necrobasidia is a plastic morphological trait

Inocybe misakaensis is more closely related to members of section *Cervicolores* that includes, for example, the widely distributed species *I. calamistrata* (Fr.: Fr.) Gillet. This relationship is demonstrated despite the presence of basidia that become ochraceous and collapse (necrobasidia) in *I. misakaensis*. The presence of necrobasidia is a trait that occurs in all samples of the Mallocybe and Auritella clades (Fig. 2). However, a collection of *I. calamistratoides* Horak from New Zealand also features necrobasidia and is strongly placed (100% bootstrap support) in section *Cervicolores* of the Inosperma clade. Thus, it appears that either necrobasidia have evolved more than once or have been repeatedly lost. In any case, the possession of necrobasidia cannot be emphasized as a synapomorphic trait in *Inocybe* (Kuyper 1986) although all accessions sampled to date in the Mallocybe and Auritella clades do have necrobasidia (Matheny 2004).

The unique cheilocystidia morphology and ecology of *I. misakaensis* are supported as distinct characters by the RPB2 phylogeny. Like *I. misakaensis*, only *I. aureoplumosa* from Cameroon (Watling 2001) has both necrobasidia and an association with African caesalpinioid trees. *Inocybe aureoplumosa*, however, is sister to several undescribed Australian taxa in the Auritella clade (Matheny and Bougher, unpublished).

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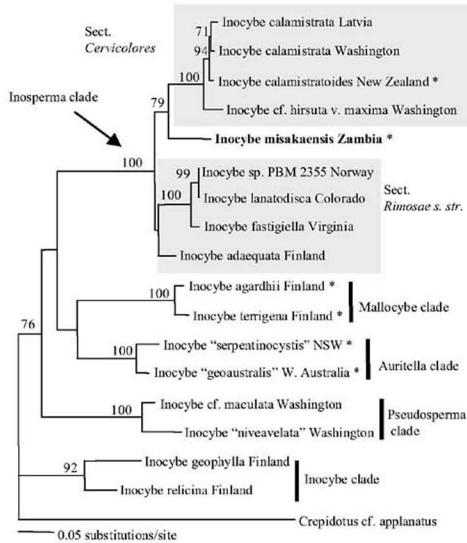

Fig. 2. The ML tree distinguishes *I. misakaensis* (in bold type) from other depauperate taxa of the Mallocybe, Auritella, and Pseudosperma clades and the pleurocystidiate group, the Inocybe clade. The asterisk after a name indicates a necrobasidiate species. Sections *Cervicolores* and *Rimosae s. str.* are boxed in gray. This tree is inferred from nucleotide sequences of RPB2 conserved domains 6 to 7. Branch lengths are proportional to the number of substitutions per site. Bootstrap values greater than 50% derived from a maximum parsimony analysis are indicated above branches. Geographic origins for taxa are also indicated. *Crepidotus* is used to root the tree. NSW is an abbreviation for New South Wales.

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Appendix

Collections used for RPB2 nucleotide sequencing include: Inosperma clade: Section *Cervicolores: I. calamistrata* (JV 11950, TURA, WTU); *I. calamistrata* (PBM 2351, WTU); *I. calamistratoides* (ZT 96/30, ZT, WTU); *I.* cf. *hirsuta* var. *maxima* (PBM 1066, WTU); *I. misakaensis sp. nov.* (Wat. 24830, E, WTU). Section *Rimosae s. str.: Inocybe* sp. PBM 2355 (PBM 2355, WTU); *I. lanatodisca* (ST 99-229-01, WTU); *I. fastigiella* (JRH 408, WTU); *I. adaequata* (JV 16501E, TURA, WTU). Mallocybe clade: *I. agardhii* (JV 7485F, TURA, WTU), *I. terrigena* (JV 16431, TURA, WTU). Auritella clade: *I. "serpentinocystis"* (T25080, WTU, OSC); *I. "geoaustralis"* (H7344, CSIRO-PERTH, WTU). Pseudosperma clade: *I.* cf. *maculata* (PBM 525, WTU), *I. "niveavelata"* (PBM 2337, WTU). Inocybe clade: *I. geophylla* (JV 6374, TURA, WTU), *I. relicina* (JV 10258, TURA, WTU).

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A new species of *Parmotrema* (Ascomycota: *Parmeliaceae*) from Portugal

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Abstract—*Parmotrema sampaioi* is described as new to science. This species is characterized by revolute lobes, the lack of cilia and vegetative propagules, and the presence of medullary protocetraric acid.

Key words—Europe, herbarium, lichen, Sampaio

Introduction

The Herbarium of the University of Porto (PO, Portugal) is housed in the Botanical Institute 'Dr. Gonçalo Sampaio', and is one of the more important herbaria in the Iberian Peninsula as it houses 122,000 herbarium sheets belonging to all plant and fungal groups. The lichen herbarium is represented by c. 9000 specimens and comprises a general herbarium with specimens from Europe, a Portuguese herbarium with local specimens, and several special collections, the most important being Gonçalo Sampaio´s collection with c. 4000 specimens, mostly from Portugal and collected in the first decades of the twentieth century. Lichen taxa described by Sampaio have recently been typified (Paz-Bermúdez et al. 2002), the identity of some critical collections re-determined and new records reported for Portugal (Paz-Bermúdez 2003).

Among the earliest collections from Portugal are some specimens collected at the end of the nineteenth century. These were sent to William Nylander for identification, and one such specimen forms the subject of this paper.

Materials and Methods

The morphology of the lichen specimen was examined using a Zeiss Stemi 2000C stereo microscope, and a Zeiss Axiolab compound microscope. Chemical constituents were identified by thin layer chromatography (Culberson 1972; Culberson et al. 1981; Culberson & Johnson 1982; Elix & Ernst-Russell 1993), high performance liquid chromatography (Elix et al. 2003) and comparison with authentic samples.

Taxonomic Description

Parmotrema sampaioi Paz-Bermúdez & Elix, *sp. nov.* **FIGURE 1**

Thallus *ut in Parmotrema saccatilobum sed superfice superiore exisidiatus differt.*

Etymology: in honor of the Portuguese lichenologist Gonçalo Sampaio.

Type: **PORTUGAL**. Póvoa de Lanhoso: S. Gens, on granite rock, *Isaac Newton*, 25 Sept. 1880; holo: PO-2997L.

KEY CHARACTERS — Thallus saxicolous, foliose, loosely adnate, coriaceous, 9-12 cm wide. **Lobes** imbricate, subirregular, 4-12 mm wide, marginal and submarginal lobes becoming markedly saccate and revolute; margins crenate or irregularly incised-dentate, ascending or revolute; cilia absent; lobules present along the lobe margins, 0.5-1.5 mm wide, monophyllous or rarely branched, minutely ciliate, cilia 0.2-0.5 mm long. **Upper surface** gray, becoming gray-brown in the herbarium, flat to convex, emaculate, smooth to rugulose on older lobes; isidia and soredia absent. **Medulla** white. **Lower surface** black, with a brown erhizinate marginal zone; rhizines very rare or absent, simple, slender, to 1 mm long. **Apothecia** and pycnidia not seen. **Chemistry** — Cortex K+ yellow; medulla K+ pale yellow-brown, C-, KC-, P+ brick-red; containing atranorin (minor), chloroatranorin (minor), protocetraric acid (major), convirensic acid (minor), virensic acid (trace), subvirensic acid (trace).

Distribution — At present this species is only known from the type collection.

COMMENTS — This new species is characterized by the saccate-revolute, eciliate lobes, the lack of vegetative propagules, the marginal, minutely ciliate lobules, very sparse rhizines and the presence of medullary protocetraric acid. Morphologically *P. sampaioi* closely resembles *P. saccatilobum* (Nyl.) Hale and, in fact, W. Nylander identified this specimen as *Parmelia saccatiloba* Nyl. These two species have similar loosely adnate thalli which lack cilia and have saccate-revolute lobes, lack soredia and contain protocetraric acid in the medulla. However, *P. saccatilobum* can clearly be separated by the presence of cylindrical isidia on the upper surface (*P. sampaioi* is non-isidiate), and the absence of minutely ciliate lobules along the lobe margins.

The type locality, Póvoa de Lanhoso, has a rather unique lichen flora. It is one of the two Portuguese locations where *Erioderma mollisimum* (Samp.) DuRietz is known in continental Europe (Paz-Bermúdez et al. 2002; Jørgensen 2000).

Figure 1. *Parmotrema sampaioi* (holotype in PO). Scale bar = 5.0 mm.

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MYCOTAXON

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Book reviews and notices

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General

Mycologia Balcanica. Editor in Chief Cvetomir M. Denchev. Vol. 1 (1). February 2004. Bulgarian Mycological Society, c/o Institute of Botany, Bulgarian Academy of Sciences, Acad. G. Bonchev Street, BG-1113 Sofia, Bulgaria. Pp. 73 + vi. ISSN not indicated. Price: € 43 (individuals) or € 58 (institutions) [Balkan Peninsular countries], € 58 (individual) or € 85 (institutions) [other countries].

It a pleasure to draw attention to a new "international mycological journal", especially one so well-produced. *Mycologia Balcanica* is the official English language journal of the Bulgarian Mycological Society; it is entirely in English. The first issue contains 18 original papers and one book review. Most papers are floristic or systematic, but cover a wide range of fungal groups (e.g. agarics, deuteromycetes, discomycetes, powdery mildews, pyrenomycetes, rusts, smuts) and countries (notably Austria, Bulgaria, Greece, Italy, Romania, Serbia, and Turkey). It is planned to publish one volume consisting of three parts each year.

The journal is large-format (slightly shorter than A4) and printed on coated (glossy) paper. The layout is first-rate, as is the reproduction of line drawings, but the few half-tones in this part are all very dark. Papers are refereed, and the 16 reviewers acknowledged as helping with contributions in this part are mainly specialists with international reputations. The standard of the papers included is consequently high, though I was sad to see the name of the fine colour photograph of *Clathrus ruber* on the front cover attributed to "P. Micheli : Pers." [Micheli cannot be credited as having validly published a species name as he is a pre-starting point author].

I have always been a strong advocate of publishing papers of regional interest in journals issued in the pertinent region; that is the best way of ensuring that the data are available to those working there. That English has been selected as the language to be used, also means that the results appearing here are also easily digested by those not fluent in languages of the region.

Further information is available on the Society's web page (www.mycobalcan.com), which also has information on the contents of future issues. I was personally interested to see that 1(3) is advertised as containing a catalogue of the lichens and lichenicolous fungi of Bulgaria.

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[1] Books for consideration for coverage in this column should be mailed to the Book Review Editor (address above) in the first instance. Fax (+34) 91 857 3640; e-mail: myconova@terra.es.

The cost is very reasonable for a publication of this quality, no doubt a result of the support received from the Bulgarian Ministry of Environment and Water, Ministry of Education and Science, and Foundation 'Biodiversity'. The foresight in such government bodies supporting a journal of this type is to be commended, and hopefully this model of funding can be emulated elsewhere. The journal should be added to the subscription lists of all European mycological libraries, and those of major mycological centres world-wide.

Katalog over Makro- og Mikrosopp angitt for Norge og Svalbard. By Jon-Otto Aarnæs. July 2002. FungiFlora, P. O. Box 95, Blinderen, N-0313 Oslo, Norway. [Synopsis Fungorum Vol. 16.] Pp. viii + 412. ISBN 82 907724 27 6. Price: NOK 450.

This checklist of the macro- and microfungi of Norway (including Spitzbergen) deals with 14 286 names, of which 12 390 are accepted and 4191 treated as synonyms (with about 1000 more old and doubtful). The checklist includes chytrids and slime moulds, but not lichen-forming species, although lichenicolous fungi are covered. This is unfortunate as while the lichens are covered elsewhere as are the lichenicolous fungi (Santesson *et al.*, 2004) their inclusion would have made the whole so much more complete. In all, 4046 basidiomycetes, 2318 ascomycetes, 1270 'deuteromycetes', 211 chromists, 68 zygomycetes, and 286 myxomycetes are accepted.

For each accepted species, the entries include literature references to Norwegian records, and Norwegian common names are given where they exist, but no details of hosts or distribution in the country are provided. Dates of publication of genera are included, but inconsistently not those of species names.

Nevertheless, there is no question that this is a landmark publication for Norwegian mycology which by virtue of the comprehensive an painstaking indexing of the old as well as the new literature will be the starting point for all wishing to locate details of the reports of particular species in the country.

Santesson, R., Moberg, R., Nordin, A., Tønsberg, T. & Vitikainen, O. (2004) *Lichen-forming and lichenicolous fungi of Fennoscandia*.— Uppsala: Museum of Evolution, University of Uppsala.

Microfungi of Tropical and Temperate Palms. By Joanne E. Taylor & Kevin D. Hyde. September 2003. Fungal Diversity Press, Department of Ecology and Biodiversity, University of Hong Kong, Pokfulam Road, Hong Kong S. A. R. [Fungal Diversity Research Series Vol. 12.] Pp. ix + 459. ISBN 962 86765 1 2. Price US$ 80.

The prospect of a single book covering all that is known of the microfungi on palms is attractive and much needed, but that would be a Herculean task. This book is not that, but an approach to the problem. Following a well-referenced overview of the current state of knowledge, the rationale of this study is explained. Three species of palms were selected (*Archontophoenix alexandriae, Cocos nucifera,* and *Trachycarpus fortunei*) and sampled in Australia, China (Hong Kong, Hubi, and Hunnan), Malaysia, Singapore, Switzerland, and the UK. The bulk of the work (344 pages) is devoted to keys and accounts of the 288 species in 153 genera encountered. One genus (*Tribulatia*) and 34 species are described as new. The species accounts include bibliographic citations, detailed descriptions, information on cultural features (where isolated), ecology and distribution, and in many cases first-rate photomicrographs. Tables comparing microscopic features of different collections and also with those already published are an especial feature that is rarely emulated. This section makes this book a 'must have' for those working on the identification of palm fungi, but caution needs to be applied in following some of the citations of older names; there is even a 'Link ex Fr.' (p. 369). The work concludes with an overview and analysis of the data, much of which has appeared in previous papers but had not been brought together in this way. Interestingly, only 26 of the species appeared to be host

specific, and the species found in particular sites appeared to be limited by climate, specific site conditions, and the status of the host palm in that locality. Attention should also be drawn to the Appendix detailing fungi previously descibed from these particular palms. In summary, a significant and useful contribution to our knowledge of palm fungi on which others can build.

Basidiomycetes

Contribution to the knowledge of genus *Agaricus*. By Luis A. P. Sánchez. 2003. Candusso, Alassio, Italy. [Fungi non delineati Pars XXIV.] Pp. 108, coloured photographs 48, figs 24. ISSN 1128 6008. Price: Unknown.

This is another volume in the series *Fungi non delineati*, published by Candusso from Italy which started in 1997. Twenty-four volumes in only seven years is an amazing result. The lay-out is the same as in the other 23 volumes: a number of rare or unknown species are described, the original diagnosis is given, and all species are depicted with colour photographs of the fruit bodies, and also line-drawings of microscopical structures and fruit bodies. Furthermore, the basionym and synonyms and some comments on the taxonomy are given, and the material examined is cited.

In this case this has resulted in a booklet of 108 pages, bilingual since everything is given in Spanish as well as in English. An Introduction of *ca* 1.5 pages gives a list of the characters which are considered constant (taxonomically useful?) by the author. Twelve species are described and depicted, all rare or uncommon in Spain. The order of the species is strictly alphabetical, but there is no reference to the numbered colour plates. The 48 colour plates are of good quality and seem to reproduce the natural colours, although not always typical or fresh specimens are shown. The picture of *Agaricus bohusii* for instance looks a bit dried out. One picture is given of *A. pseudolutosus*, which is not described. The line-drawings are somewhat roughly inked, but in general give a good impression of the features.

Unfortunately the English text has many spelling-mistakes and grammatical errors, and strangely enough the English and Spanish text do not always match.

In general the synonymy is the same as used in *Flora Agaricina Neerlandica* (Nauta, 2001), synonymizing for instance *A. benesii* and *A. squamuliferus*, *A. bernardii* and *A. maleolens*, *A. bresadolanus* and *A. romagnesii*, *A. lanipes* and *A. luteolorufescens*, and *A. subperonatus* and *A. pseudovillaticus*.

I have, however, a few critical remarks. It is a pity that the author did not always compare his collections with material from abroad. *A. lanipes* is only based on two collections, and the colour photographs do not look so very typical. *Agaricus moellerianus*, for instance, is only based on one collection. Although a rare species in most countries in Europe it can be observed regularly, and the difference from *A. campestris* is not always that clear. The strong yellow discoloration which is mentioned is not always that strong or clear, and also occurs in *A. campestris* var. *equestris*.

The described and depicted *A. lutosus* looks remarkably similar to what was formerly called *A. semotus* (and should be called *A. dulcidulus* in my opinion) which is in northern Europe very common and morphologically rather variable. *Agaricus lutosus*, as Møller defined it, differs from "*A. semotus*" only in the stipe tapering at the base and the yellowish pileus with only a few vinaceous fibrils when young.

But these remarks do not detract much from the value of this booklet. It is very useful for those interested in the subject, and a valuable addition to other existing books because of the combination of good coloured photographs and elaborate descriptions.

Nauta, M. M. (2001) *Agaricus*. In *Flora Agaricina Neerlandica* (M. E. Noordeloos, T. W. Kuyper & E. C. Vellinga, eds) 5: 23-61. — Lisse: Balkema Publishers.

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Il Genere *Cortinarius* in Italia. Parte prima. By Giovanni Consiglio, Daniele Antonini & Massimo Antonini. 2003. Associazione Micologica Bresadola (AMB), Via Alessandro Volta 46, I-38100 Trento, Italy. Ring binder, pp. 64 + 50 loose-leaf profiles each of 4 perforated pages. Price: € 50.

The book is the first volume of a monograph of *Cortinarius*. It is being prepared by three specialists of the Associazione Micologica Bresadola, which has already published several other very interesting works on different genera. In the present volume, the 50 first species are presented, as the commencement of a book on a genus not very frequently studied. The authors hopefully announce an annual publication of new chapters. The book has a prologue, written in French by André Bidaud, a cortinariologist co-author of the already classical *Atlas des Cortinaires*.

The description of species is preceded by an introduction of two chapters. The first provides a comparative study of the two classification systems proposed for the genus *Cortinarius* genus, the Nordic *Flora Photografica*, which divides genus into four subgenera, and the French *Atlas des Cortinaires*, where the species are classified in six subgenus. They also cite the historic concept of Moser in *Die Gattung Phlegmacium*, which is specific for that subgenus. The authors of the present book do not make a choice, but give instead in each profile the classification according to the different systems. An extensive bibliography of around 200 references concludes the first chapter. The second part of the introduction, by Alfredo Vizzini, is entitled 'Secondary metabolites of *Cortinarius* s.l. species'. This is a study that goes deeply into the toxicological and pharmaceutical aspects of the genus, and includes also a specific bibliography on this subject. The authors announce that in future issues other general subjects related to the genus will be treated, including history, ecology, chorology, macrochemical reactions, etc.

As regards the descriptive section of the work, there are 50 species profiles, each of four pages, with an emphasis on the subgenus *Phlegmacium*, represented here by 28 species. Each profile includes an exhaustive taxonomic analysis, which places the species in the genus according to the alternative systematic proposals mentioned, and also incorporates the original Latin diagnosis (translated also into Italian), information on the etymology of the name, and a discussion of its taxonomic classification. The descriptive part itself is quite detailed, and comprises macroscopic and microscopic elements, reactions with KOH, and references to the habitat. However, a more extensive reference to macrochemical reactions, which are often the determinants of identifications in the genus, would have been desirable and welcomed. With respect to the images, each profile includes two colour photographs of excellent quality, an SEM micrograph, and a black and white sporogram giving maximum and minimum sizes of length, width and the Q ratio. A regional distribution map for the species in Italy is also presented in each profile.

This is an excellent book, and a very good complement to the existing works on *Cortinarius* in the context of the mediterranean area. I hope that the annual issuing rate will be realized, and expect that the work will help promote the interest of the mycological community to this large and passion-generating genus.

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A Preliminary Monograph of *Lentinellus* (*Russulales*). By Ronald H. Petersen & Karen W. Hughes. March 2004. J. Cramer in der Gebrüder Borntraeger Verlagsbuchhandlung, D-14129 Berlin. [Bibliotheca Mycologica No. 198.] Pp. 268, figs 51, tables 18, col. plates 13. ISBN 3 443 59100 0. Price € 80.

As leader of the London Fungi Recording Group, in the autumn of 2003 I collected a species of *Lentinellus* from Epping Forest, to the north of London. Upon examining the collection both macro- and microscopically, it soon became apparent to me how difficult it was to assign this particular specimen to a species utilising personal and available literature in the library at the Royal Botanic Gardens Kew.

The species I collected was eventually named as *L. ursinus*, but to my mind and others, certain macroscopic features didn't seem to fit with *L. ursinus*, so a nagging uncertainty surrounded my collection.

Upon a recent visit to David Hawksworth, who knew of my problem, I was presented with this new preliminary monograph of the genus *Lentinellus* to utilise and then review. The monograph accepts 24 species, based on a study of worldwide collections.

The introduction to the monograph includes an engrossing section on the historical lineage of *Lentinellus*, leading to the molecular discovery of an evolutionary connection between *Lentinellus* and the *Russulaceae* — fascinating stuff, but the amateur mycologist will need a copy of *Ainsworth & Bisby's Dictionary of the Fungi* by their side to decipher some of the terms. The taxonomic characters of *Lentinellus* are dealt with fully in descriptions and line drawings, making the inherent characteristics of particular species clear and easy to follow. Materials and Methods, along with microscopic techniques are given in this section, also giving a good explanation on how the authors reached their conclusions. *Hemicybe* continues to be treated as a synonym. The key is a simple dichotomous one that is easy to follow.

The descriptions of the accepted species, including cultural morphology and sexual mating compatibilities, are presented in depth, complete with diagrams of microscopic features and colour plates which cover 16 species. There is an excellent commentary section accompanying each species, outlining important information. Many species are lecto-, neo-, or epitypified for the first time. The inclusion of a table of spore statistics for each species as given by different authors and from different collections is also a most useful feature. I also welcomed the reproduction of copies of original illustrations of the species. The copy of Sowerby's illustration of the species now called *L. vulpinus* (originally described from north London) and the detailed notes on smell leave us in no doubt that this was the species we had from Epping Forest.

Going by own experience with the genus *Lentinellus*, this monograph gives an excellent insight into the genus and will prove to be a valuable addition to the growing number of modern generic revisions. Its depth and clarity make it a model to be emulated by others undertaking revisions of macromycete genera.

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Some Neotropical Wood-Inhabiting Fungi. Edited by Leif Ryvarden. 2002. FungiFlora, P. O. Box 95, Blindern, N-0313 Oslo, Norway. [Synopsis Fungorum Vol. 15.] Pp. 80. ISBN 82 907724 26 8. Price: NOK 120.

This is a collection of seven papers by various authors focussing on neotropical wood-inhabiting basidiomycetes. In all, four new genera, 12 new species and 10 new combinations are included for fungi which are generally perceived as relatively well-known in the region. There is a key to *Kneiffiella* (7 spp.) but without a discussion of its separation from *Hyphodontia*. *H.* sect.

Alutaceodontia is raised to generic rank, the new genus *Leptocorticium* is introduced for *Corticium cyatheae, Hydnodon* (based on *Hydnum thelephorum*) is synonymized with *Trechispora*, seven new *Inonotus* species are described (and a key to the 24 neotropical species now recognized provided). In addition, 155 species are reported from Paraná State in Brazil (five new to science, with keys to the neotropical *Antrodiella,* and *Coltrichia* species, and the world *Grammothelopsis* species), and seven new to Venezuela. In summary, something of a *potpourri*, but together making a significant contribution to our basic knowledge of neotropical corticioid and polyporaceous fungi.

A Critical Checklist of Corticioid and Poroid Fungi of Norway. By Leif Ryvarden, Jogeir Stokland & Karl-Henrik Larsson. February 2003. FungiFlora, P. O. Box 95, Blindern, N-0313 Oslo, Norway. [Synopsis Fungorum Vol. 17.] Pp. 109. ISBN 82 907724 28 4. Price: NOK 120.

This checklist accepts 204 poroid and 439 corticioid fungi, 36 species of which are new records for Norway post-Aarnæs (see above) and two are new to science (*Oligoporus hydnoidea* on *Picea,* and *Tyromyces vivii* on *Populus*) and one is a new combination. Keys to the European species of *Antrodiella* (14 spp.) and *Skeletocutis* (21 spp.) incorporated mean that the work will be of widespread interest to workers on these fungi. Much of the new work is a result of studies in 1995-97 of 160 selected sites, mainly in south-eastern Norway, which generated a massive 26 000 collections of fungi in these groups. For each species listed, there are notes of the number of records, indicated by province for less frequent species, and with more precise details for the rarest. No synonyms are provided or cross-referenced, but that information is mostly available from Aarnæs' catalogue. As so many additions were generated from one regional study in the country, it is clear that much more remains to be found in these groups even in Norway where they have probably been more intensively investigated than almost any region in the world.

Annotated list of Heterobasidiomycetous Fungi for the Iberian Peninsula and Balearic Islands. By Margarita Dueñas. 2002. J. Cramer in der Gebrüder Borntraeger Verlagsbuchhandlung, D-14129 Berlin, Germany. [Bibliotheca Mycologica No. 196.] Pp. 90, figs 1, tables 3. ISBN 3 443 59098 5. Price: € 32.

This comprehensive checklist, published in English, is part of the *Flora* (sic!) *Micológia Ibérica* project and is intended as a precursor to a future regional monograph of the Portuguese and Spanish heterobasidiomycetes. Rusts and smuts are excluded, as are the heterobasidiomycetous yeasts, but included are all the teleomorphic species in the *Atractiellales, Auriculariales, Dacrymycetales, Exobasidiales, Platygloeales, Septobasidiales, Tremellales,* and *Tulasnellales* (all *sensu* the 1995 edition of *Ainsworth & Bisby's Dictionary of the Fungi*). The checklist is arranged alphabetically by order, genus, and species, and provides full literature references for each taxon and for the synonyms used in the region, details of type specimens or type localities, distribution by province, details of reported substrata, and a record of herbaria containing voucher collections. Appendices include a list of taxa by province, and a bibliography of the literature surveyed to compile the list.

The checklist will clearly be of value to anyone interested in heterobasidiomycete distribution, not only in the Iberian Peninsula but throughout Europe and further afield. Though some critical work has been undertaken (as evidenced in the author's other papers and in footnotes in the text), the checklist relies substantially on unconfirmed reports and includes a number of highly dubious taxa which require further research. This, presumably, will be the next step in preparing a more thorough, regional mycota. Inevitably, there are also quite a few errors in author citations (the dubious old name *Tremella intumescens,* for example, should be credited,

if that is the right word, to the Englishman J. E. Smith rather than the Norwegian Sommerfelt), and in literature references (particularly old ones), both of which need further attention before a mycota is prepared.

There is certainly quite a lot of work still to do. According to the appendix, the best recorded province (Madrid) has 51 heterobasidiomycete species listed, but more than 25 other Iberian provinces have five or fewer. At least this volume should make the task easier, and as such is much to be welcomed.

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The Rust Fungi of the British Isles: A guide to identification by their host plants. By D[ouglas] M. Henderson. 2004. British Mycological Society, Joseph Banks Building, Royal Botanic Gardens, Kew, Richmond-upon-Thames, Surrey TW9 3AE, UK. Pp. 35. ISBN 0 95227704 9 0. Price: Not indicated.

This is a complement to the checklist published by the same author in 2000 (see *Mycotaxon* **77**: 505, 2001), providing notes on the rusts reported arranged by host families and genera, with indications of the states to be expected and in some cases other hints to help in their separation. An Appendix provides updates, additions and corrections to the 2000 list. There is no doubt that this is a valuable adjunct to the checklist, but together they are still a supplement rather than a replacement for the seminal *British Rust Fungi* (Wilson & Henderson, 1966).

Wilson, M. & Henderson, D. M. (1966) *British Rust Fungi*. — Cambridge: Cambridge University Press.

Lichen-forming Fungi

The Macrolichens of Ohio. By Ray E. Showman & Don G. Flenniken. April 2004. Ohio Biological Survey, P. O. Box 21370, Columbus, Ohio 43221-0370, USA. [Ohio Biological Survey Bulletin, new series Vol. 14(3).] Pp. iv + 279. ISBN 0 86727 149 3. Price: US$ 35.

The Ohio Biological Survey has long been aware of the importance of lichens in its biota, having previously published accounts of the foliose and fruticose species (Taylor, 1967, 1968). However, those works are now long out of print and inevitably somewhat dated. Ray Showman has been working on the lichens of the state since the early 1970s, and the two authors produced a checklist of its macrolichens back in 1990 (Flenniken & Showman, 1990). Since then, Don produced a colour-illustrated treatment of the lichens of West Virginia state (see *Mycotaxon* **74**: 511-512, 2000). Now they have combined to produce an update of Taylor's work in a single new book, which includes all 223 macrolichens currently known from the state, with a checklist to facilitate cross-linking with names used by Taylor. After a basic and sound introduction, there is a key to genera, which are treated alphabetically, and then keys to species within each genus. Each species has a description, a summary of diagnostic features (helpfully in bold type), notes on ecology and chemistry, a distribution map (by county) and cross-references to colour photographs in *Lichens of North America* (see *Mycotaxon* **82**: 481-482, 2002) or Flenniken's earlier book. The only photographs included, apart from six on the front cover, are of the authors. Common names are given for some species, a practice I always think suspect when these are hardly known -- for example, how many lichenologists would know what the Powdery Sunburst Lichen was (actually *Xanthoria ulophyllodes*)? The book is well-presented and the ring-binding facilitates easy use. Now with this and the two recommended books to hand, there

should be no excuse for passing Ohio macrolichens by. Now we await a companion volume on the crustose species in the state . . .

Flenniken, D. G. & Showman, R. E. (1990) The macrolichens of Ohio: a revised checklist. *Ohio Journal of Science* 90: 130-132.

Taylor, C. J. (1967) *Lichens of Ohio*. Part 1. *Foliose Lichens*. [Ohio Biological Survey Bulletin, Biological Notes No. 3.] — Ohio State University, Columbus.

Taylor, C. J. (1968) *Lichens of Ohio*. Part 2. *Fruticose and Cladoniform Lichens*. [Ohio Biological Survey Bulletin, Biological Notes No. 4.] — Ohio State University, Columbus.

Líquenes de la Reserva Natural Integral de Muniellos, Asturias. By Eva Barreno Rodríguez & Sergio Pérez-Ortega. December 2003. Consejería de Medio Ambiente, Ordenación del Territorio e Infraestructuras del Principado de Asurias y KRK Ediciones, Álvarez Lorenzana 27, ES-33006 Oviedo, Spain. [Cuadernos de Medio Ambiente, Serie Naturaleza No. 5.] Pp. 512, col. figs 133. ISBN 84 96119 36 X. Price: € 30.

This book, based on the PhD thesis of the second author, examines the lichens and lichen communities in the Reserva Natural Integral de Muniellos in Asturias, northern Spain. The site is one the six UNESCO MAB [Man and the Biosphere] reserves in Spain, covers 542 ha, and has peaks rising to 1678 m (Pico zos Redondo). The rocks are quartzite, sandstones, shales and slates. In all, 430 species are treated, 45 % occurring on trees, and 60 % of which are crustose. The lichens in the area are clearly very lush, with *Lobaria amplissima* attaining 45 cm diam, and its *'Dendriscocaulon'* up to 2.5 cm tall. Identification problems were sometimes encountered, populations having 'deviant traits'. Nitrophilous communities are hardly developed, suggesting it is relatively free of the nitrogen pollution which is now so widespread in Europe, and making this an exceptional site for Atlantic lichens. The overall species list, while commendable, does seem to have some surprising absences (e.g. *Bryoria smithii* is included but not the usually commoner *B. bicolor*) and could surely be extended by further work. In executing the study 70 sites were concentrated on for sampling and this focussing, while pragmatic for a PhD, might have led to species being missed by more widespread searching. I also found it frustrating that precise localities were not given; perhaps this was deliberate to discourage collecting in the richest sites in the interests of conservation!

But this is much more than a floristic account, as in includes detailed descriptions and keys, and also a very full introduction (112 pp.), an extensive glossary illustrated by numerous line drawings, and an impressive literature list (with 21 web site sources also given on p. 492). Further, the 133 fine colour photographs include 35 of communities and habitats, and 98 of particular species. It is these elements that will ensure that this work will be widely used in Spanish-speaking countries. For those that are not familiar with Spanish, there is an extensive summary in English (pp. 35-43).

The authors are to be congratulated on this major contribution to our knowledge of lichens in the region, and by the back door publishing a general introduction to lichenology in Spanish at a very modest price well suited to a student's pocket.

The Epiphytic Lichens on *Quercus* in Sardinia (Italy) and their value as ecological indicators. By Luciana Zedda. 2002. Botanischen Garten und Botanischen Museum Berlin-Dahlem, Königin-Luise-Straße 6-8, D-4191 Berlin, Germany. [Englera Vol. 15.] Pp. 457, figs 38, tables 38. ISBN 3 9211800 49 8. Price: € 46.

This work is the product of a PhD thesis of which the lichenological supervisor was Harrie J. M. Sipman. The lichens on four *Quercus* species in 92 plots in 62 localities in Sardinia were

examined, mainly in 1996-98. Over 3500 specimens were collected, representing 331 taxa, including the new species *Lecanora leuckertiana* (published elsewhere) and *Ramalina farinacea* var. *variolarica* (*ined.*). An amazing 75 species are first reports for Sardinia, and 13 of those species are new for Italy as a whole; this clearly shows what new information can be generated from closely focussed studies even on such a relatively well-explored island. Half (51 %) of the species were crustose, 62 % reproduced sexually, and 31 % by soredia. The richest host was *Q. ilex*, with 256 species, and more species were found on trunks (287 spp.) than twigs (159 spp.). For each taxon treated, earlier literature reports and details of all collections are cited, followed by discussions of the occurrences, ecology, distributions (especially in Europe), and comments on their taxonomic status.

While a phytosociological classification is not attempted, three of the systems of analysis applied are of especial interest. Two sites score 100 in the Revised Index of Ecological Continuity (RIEC), both in the Marghine-Goceano area; these are clearly old-growth forests of major conservation importance. Disturbance is also scored by an 'hemerobic' seven-zone scale indicative of the extent of human activity, in which H6 is a 'lichen desert' and H0 represents no human impact. New here is an 'Index of Microclimatic Dryness' (IMD), defined as the number of widespread temperate species dived by the number of suboceanic species; the results were well-correlated with forest-type. For those wishing to make other analyses, extensive appendices include data by host and plot.

The whole is presented in the context of other studies of lichens on oak, though I would have expected more comparison in the case of *Q. suber* with the substantial analysis by Fos (1998) of those on cork oak throughout the Iberian Peninsula. That studied recorded 304 species on this oak, compared with 167 reported here on it in Sardinia.

In summary, this is not only an excellent example a study in the systematic-ecological Mediterranean lichenological tradition, but also a work of wider interest because of the taxonomic notes and approaches to the distributional and ecological analyses.

Fos, S. (1998) Líquenes epífitos de los alcornocales Ibéricos: correlaciones bioclimáticas, anatómicas y densimétricas con el corcho de reproducción. *Guineana* 4: 1-507.

Conidial Fungi

Alternaria. By Tian-Yu Zhang. 2003. Science Press, Beijing. [Flora Fungorum Sinicorum Vol. 16.] Pp. xvii + 283, tables 1, figs 132, coloured plates 4. ISBN 7 03 011095 1. Price: 58 Yuan.

This volume is in the tradition of the series, endeavouring to fully describe and document all the species of the pertinent group reported from the People's Republic of China. It is, however, unusual in the focus on a particular 'genus', although this is not the first time this has has been done (e.g. *Aspergillus* in vol. 5, 1997). After a detailed introduction, the species are treated by host family, and alphabetically within each family. Keys are provided to species within each family, but there is no overall key which would have been most welcome in this 'genus'. Synonyms, descriptions, first-rate line drawings, hosts, collections studied, etc., are given for a staggering 123 taxa (including several newly described species and also an unnamed one on *Zingiber officinale*). Teleomorphs where known are indicated, but are not described or discussed. In addition, fourteen reported species are excluded for various reasons. Especially welcome are the coloured plates, each depicting the symptoms caused by 5-6 taxa.

Of especial value are the line drawings. These are mostly full-page, and in them the author has been at pains to pack in numerous conidia in order to fully show the variation within the collection from which they were made (which I was pleased to see was given).

Dr Zhang has extensive experience of these fungi, and I recall meeting him when he worked with John C. David at the International Mycological Institute in the early-mid 1990s.

Although there is a need to re-assess species concepts in such 'genera' by molecular methods and cross-infection experiments, such traditional monographs are necessary as the basis of such newer approaches and to enable identifications to be made in the interim. As such the author is to be commended on this major and painstaking publication, and it is a 'must' for the shelves of all who wrestle with identifying or researching these fungi.

***Mycosphaerella* and its Anamorphs: 1. Names published in *Cercospora* and *Passalora*.** By Pedro W. Crous & Uwe Braun. September 2003. Centraalbureau voor Schimmelcultures, P. O. Box 85167, 3508 AD Utrecht, The Netherlands. [CBS Biodiversity Series Vol. 1.] Pp. [viii] + 571, figs 30. ISBN 90 70351 49 8. Price: € 75.

The thought of wrestling with the names of all described cercosporoid fungi is enough to make most mycologists go weak at the knees, but not for this duo. This work treats an amazing 5720 names, more than 3000 of which were proposed in *Cercospora*, 659 of which are accepted, and about 550 in *Passalora* which is emended here to include *Mycovellosiella, Phaeoisariopsis p. p., Phaeoramularia,* and *Tandonella.*

The work starts with an historical account and notes on all pertinent generic names, including a key to all accepted cercosporoid and morphologically similar genera which will be of wide utility. However, only four genera are accepted as being 'true cercosporoid' ones: *Cercospora, Passalora, Pseudocercospora,* and *Stenella.*

The species are treated alphabetically by species epithet, with the genus name provided afterwards (but with no ','). Original places of publication are provided, together with those for listed synonyms, and also an indication of the herbarium in which the type is found (but without details of that collection). Most types have actually been studied by the authors, but this is not made explicit by each cited herbarium. Also provided is information on reported hosts and the countries from which the species are reported. References to key literature are also included, and synonyms are cross-referenced. There is also a host-index by plant genus.

A staggering 455 new scientific names are introduced, mainly new combinations but including some new names and newly described species. Hopefully the new taxa will be accepted as validly published by nomenclatural purists despite having the species epithet printed before that of the generic name . . . . Discussions and illustrations are minimal, which is understandable in the light of the scale of the work, but I am sure that specialists would have welcomed more information on the basis for the decisions taken.

Especially intriguing is the treatment of 281 names as synonyms of *C. apii s. lat.* This problem is discussed in some detail, and the cautionary note about describing further taxa within it as distinct merits heed, pending a more critical molecular phylogenetic analysis.

While this will not be the last word on the cercosporoid fungi, it will now be the key reference work and starting point for all future studies on them. The mycological community will now look forward to the promised companion volumes on other anamorphs of *Mycosphaerella.*

Myxomycetes

Myxomycetes of New Zealand. By Steven L. Stephenson. 2003. Fungal Diversity Press, Department of Ecology and Biodiversity, University of Hong Kong, Pokfulam Road, Hong Kong S. A. R. [The Fungi of New Zealand Vol. 3; Fungal Diversity Research Series Vol. 11.] Pp. xiv + 238, figs 44, col. plates 4. ISBN 962 86765 4 7. Price: US$ 50.

This is a model for regional accounts of any group of organisms. It is attractively produced, well presented with clear type and good line drawings, and is provided with four good colour plates,

depicting 24 species. The text is authoritative but never wordy, and the descriptions are clear and reliable. Some information is given on distribution within New Zealand but as this is the first complete account of the myxomycetes from that country in book form it is not surprising that no overall data are available. After a concise introduction to biology and structure, keys to orders, families, genera and species are provided. Within each species account comments allow comparison with related or similar species and with other taxonomic literature. The nomenclature used is completely in line with current thinking and where there are differences of opinion the author clearly indicates which opinion he follows.

The only regret I have is that the author, a leading researcher in myxomycete ecology, has not had the space to develop this area of study in the book. One assumes that the richness of New Zealand biodiversity is due not only to its geological history but also to its current geographical position, offering a very wide range of habitats, and this would have been a topic of considerable interest.

With 180 species of myxomycetes so far recorded, New Zealand still offers great scope for the myxomycologist – this modest total will surely be doubled in time. This book provides a firm basis for such further study and the author and publishers are to be congratulated for producing such an attractive and useful book at a fairly moderate price.

BRUCE ING
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Miscellaneous

Codex Botanicus Linnaeanus. By Hermann E. Richter. [Edited by John Edmondson.] 2003. A. R. Gantner Verlag, Ruggell, Liechtenstein. [Distributed by Koeltz Scientific Books, P. O. Box 1360, D-61453 Koenigstein, German.] Vol. I (with a biographical sketch by H. Walter Lack and a translation of the introductory text by Sten Hedberg). Pp. 53 + xxxii +1102. Vol. II (Index alphabiticus by William Ludwig Peterman). Pp. iv + 202. [Regnum Vegetabile Vol. 140.] ISBN 3 906166 03 1. Price: € 360.

Although first published in 1840, this is a remarkable work, which I never heard any mycologist or (lichenologist) mention, and never saw until this reissue. Richter (1808-1876) was a medical doctor in what is now Germany who determined to produce a synthetic account of Linnaeus' works. He combined the different generic and species accounts, clearly indicating which work and edition they had come from, and presents them as a single text including descriptions, synonyms, and notes. He envisaged this as a work that could be used instead of Linnaeus' own books. The details of how he worked and his philosophy are given in an English translation of Richter's own introduction by Sten Hedberg. Walter Lack also provides background on Richter and his struggling career.

However, Richter did not produce an index to the names, that task being taken on by Petermann. What is most valuable is that Petermann's index includes polynomials, so that one can quickly find, for example, what polynomials starting with *Agaricus* or *Lichenoides* have been treated in Linnaeus' works and under what Linnaean names. This means that one can take classic texts such as those of Micheli and Dillenius and find if and how Linnaeus handled the polynomials these authors used in any of his books – not only *Species Plantarum*. I have already found this of value in recent months while investigating some mid-eighteenth century names, and only wish I had known about it 30 plus years ago when I first had to wrestle with pre-Linnaean names.

This is clearly a reference work bibliophiles and nomenclaturalists should be aware of, and one that all major taxonomic libraries have on their shelves – ideally filed next not only

to Linnaeus' works but along with the also little known index to names in the dissertations of Linnaeus' students (Kiger *et al.* 1999).

Kiger, R. W., Tancin, C. A. & Bridson, G. D. R. (1999) *Index to Scientific Names of Organisms cited in the Linnean Dissertations together with a Synoptic Bibliography of the Dissertations and a Concordance for selected editions.* — Pittsburgh: Hunt Institute for Botanical Documentation.

MYCOTAXON

Volume 89(2), p. 521 *April-June 2004*

Mycotaxon changes redux: Online index & new submission protocols

LORELEI L. NORVELL

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New online index—Although *Mycotaxon* will continue to be rigorously indexed, we will no longer print an index at the end of each volume. Instead, the index will be posted on the *Mycotaxon* website (www.mycotaxon.com), from which it can be downloaded and printed. Plans are now underway to combine past and future indices into a robust *Mycotaxon* online search engine.

New submission protocols—This issue is the first to be prepared entirely (except for 6 photographic halftones) from electronic files. Although during the past 3 months your over-zealous Editor-in-Chief devoted too much time to text 'nit-picking,' last week's arrival of Adobe Creative Suite® has shed much-needed light on how to streamline *Mycotaxon* electronic submission. As most future Mycotaxon papers will also be generated from high-resolution PDF files, authors must have the Editor-in-Chief preview complete manuscript text using e-mail attachments before final submission. Text and graphics (TIF or EPS formats) files should remain separate during the pre-submission period.

Text. Electronic—Two types of text files are needed: *text without graphics* but with space for graphics insertion (_txt.doc), and *text with graphics* (_pix.doc). The Editor-in-Chief uses the former to generate PDFs and the latter to guide graphics placement. **Hard copy**—Authors need send only *one* manuscript copy (with graphics included) for editorial use with their cover letter, electronics files, and *two* peer review cover letters & checklists. The EIC will generate composite PDF files for press and index use.

Graphics. Electronic—Smaller JPGs are useful during pre-submission, but for final submission authors *must* submit a scaled-to-fit, high resolution TIF (line drawing, halftone) or EPS (gene tree, graph, diagram) file for *each* separate plate. Submit files up to 4,000k by Email or snail-mail files on CD (preferred), Macintosh zip disk (acceptable), or PC floppy (least desired). **Hard copy**—*Mycotaxon* still sends halftone plates prepared from photographic negatives to the press for high-resolution scanning and digital conversion. Halftones scanned from digital plates degrade, so authors *must never* submit digital halftones on photographic glossy paper for scanning.

Kudos—Thanks to Karen Gettelman (Index), David Hawksworth (Book Reviews), Grégoire Hennenbert (French Language), and Noni Vidal (Web) for continuing to fill their *Mycotaxon* editorial roles so efficiently. Special thanks are also due to Mycotaxon Editorial Board members & Else Vellinga for help with *Mycotaxon* 2004 Guidelines 1.0, and to Dick Korf (Business Manager) for comforting advice when most needed.

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ERRATA

VOLUME EIGHTY-THREE

Page 366, line 6	for	Hill 8133	read	Hill 8132

VOLUME EIGHTY-NINE

Page 1, line 13	for:	*latisporus*	read	*latifolius*

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The Editors express their appreciation to the following individuals who have, prior to acceptance for publication, reviewed one or more of the papers appearing in this volume.

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Index to Fungous and Lichen Taxa, Volume 89

Mycotaxon has always prided itself on providing full indices to the authors and to the taxa that appear in its pages. Beginning with this volume, those lists of taxa (typically 8 to 14 pages) will no longer appear in the printed copy. (We are saving trees.) By going to the website page where this table of contents appears,

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you can click on the taxon index link to examine (and download and print should you so desire) the index to fungous and lichen taxa for this volume (similarly for future ones). Also the taxa in this volume will be found included in our *cumulative index* to volumes 81-90 soon after publication at

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