# MYCOTAXON

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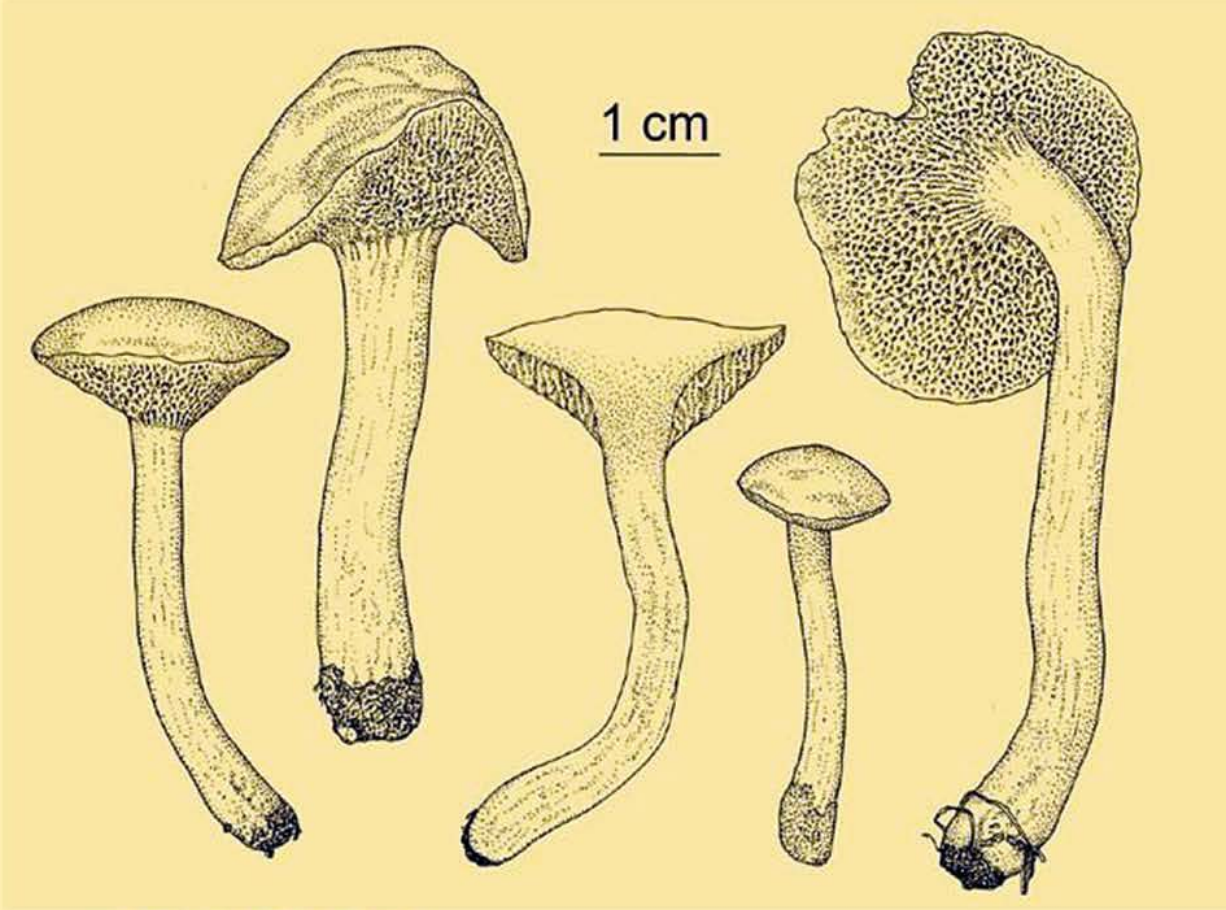


*Omer Van de Kerckhove, artist*

DEGREEF & DE KESEL
Fig 1. *Chalciporus africanus*
(p. 331)


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# MYCOTAXON

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# MYCOTAXON

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MYCOTAXON

*Volume 103, pp. 1–8* *January–March 2008*

# Two new anamorphic fungi from Brazil: *Dictyochaetopsis polysetosa* and *Myrothecium compactum*

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**Abstract** — *Dictyochaetopsis polysetosa* anam. sp. nov. and *Myrothecium compactum* anam sp. nov. found on a decaying leaves of an unidentified plant in "Morro do Corcovado" and "Pista Claudio Coutinho" near Pão de Açucar, Rio de Janeiro, Brazil are described and illustrated. The former is characterized by verticillate, polysetose, branched conidiophores and the latter is distinguished by its compact conidiomata with coarsely verrucose to tuberculate, darkly green-brown marginal hyphae and bacilliform, aseptate, dark green conidia. A description and illustrations of the comparable taxon, *Dictyochaetopsis gonytrichoides*, are provided.

**Key words** — conidial fungi, leaf litter, systematics, tropical rainforest

## Introduction

During an expedition in 2002 through the "Morro do Corcovado" and "Pista Claudio Coutinho" near the "Pão de Açucar" rainforest, Rio de Janeiro, Brazil, two conspicuous anamorphic fungi from the genera *Dictyochaetopsis* and *Myrothecium* were collected. Morphologically the two fungi were distinctly different from previously described species and are therefore described as new.

## Materials and methods

Samples of submerged plant material were placed in separate paper bags and taken to the laboratory, then incubated in Petri dishes, at 25° C, in moist chambers composed of plastic containers (50 L capacity) with 200 ml of sterile water plus 2 ml of glycerol. The plant material was examined at regular intervals for the presence of microfungi. Mounts were prepared in polyvinyl alcohol-glycerol (8 g in 100 ml of water, plus 5 ml of glycerol) and measurements made at a magnification of × 1000. Micrographs were obtained with a Leite Dialux 20 EB microscope and a Jeol JSM-6400 scanning electron microscope using the techniques described previously by Figueras & Guarro (1988).

The fungi were isolated into pure culture by transferring single conidia observed under a stereo microscope onto oatmeal agar in Petri dishes, then incubated at 25° C under 12 h alternating cycles of daylight/dark.

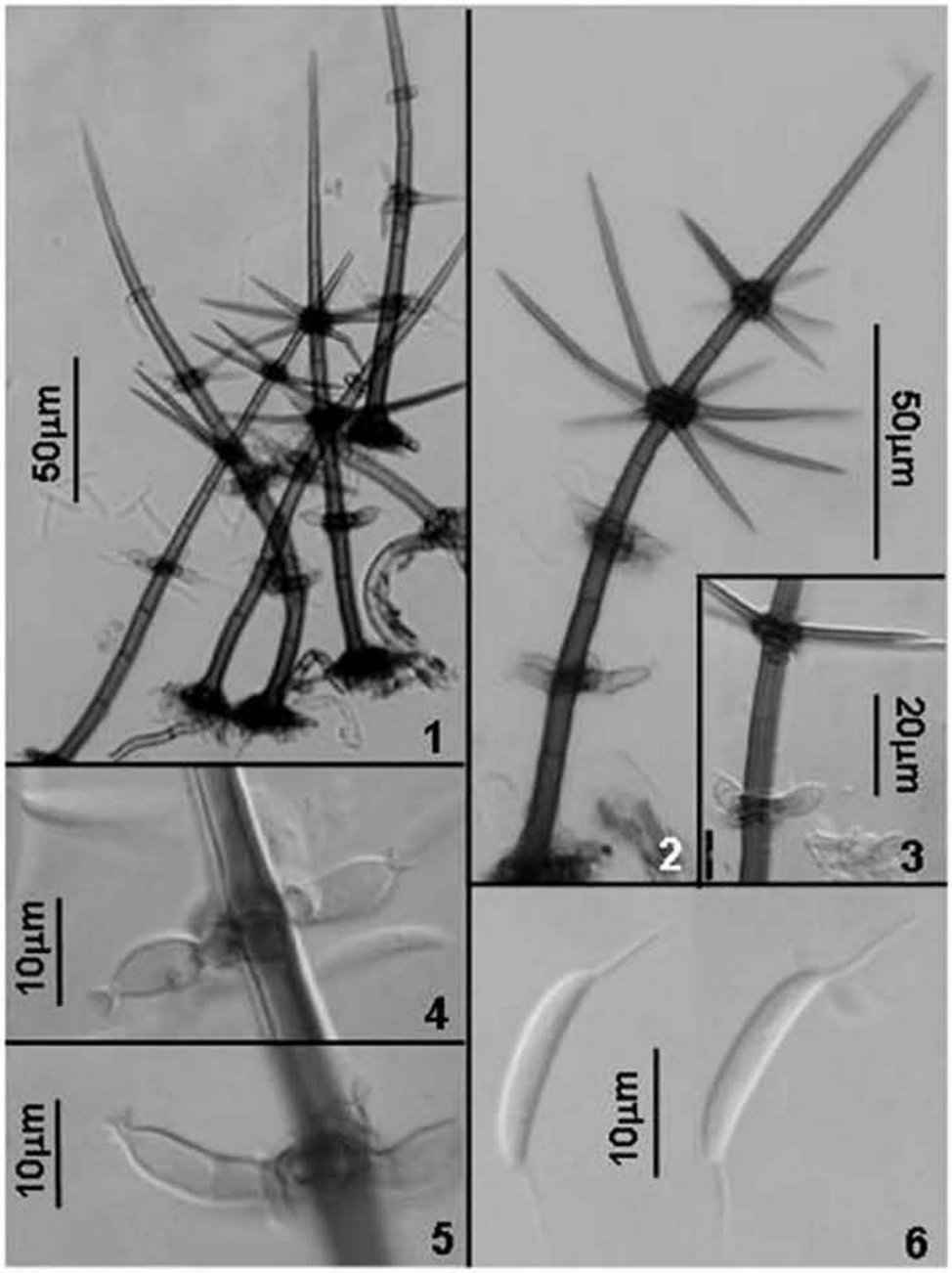
## Taxonomy

***Dictyochaetopsis polysetosa*** R.F. Castañeda, Gusmão, Guarro & Saikawa, **anam. sp. nov.** **Figs. 1-8**

MycoBank #: MB. 510905

*Ad omnibus speciebus Dictyochaetopsis differt in conidiophoris polysetosis, verticillatim ramosis in partibus superioribus, 200–320 µm altis, 8–15 µm crassis ad basim et ramis setosis, atrobrunneis vel nigris, acicularis, 18–50× 8–9 µm. Conidia alantoidea usque ad falcata, 12–15 × 2 µm, hyalina, utrimque setulosa.*

**Etymology:** Latin, *polysetosus*, referring to the setose branches of the conidiophores.




**Figs. 1-6.** *Dictyochaetopsis polysetosa* (From CBS H-6586a). **1-2.** Branched setiform conidiophores. **3-5.** Detail of conidiogenous cells. **6.** Conidia. Scale is indicated by bars.

**Colonies** on the natural substratum effuse, arboreo-pilose, amphigenous, black. Mycelium superficial and immersed composed of septate, branched,

smooth-walled, 2–4 µm diam., brown hyphae. **Conidiophores** macronematous, mononematous, polysetose, acicular at the apex, 8- to 12-septate in the main axis, erect, straight or flexuous, dark brown to brown, smooth, 200–320 µm tall, 8–15 µm wide at the base, verticillate with 3-6 acicular, setose branches, which usually arise close to, and just below, the septa in the middle part of the conidiophore and towards the apical region; setose branches 1- to 3-septate, dark brown to brown, 18–50× 8–9 µm. **Conidiogenous cells** enterogenous, unilocal, discrete, lageniform or cylindrical, smooth-walled, brown, 9–14 × 4–7 µm, arising from superficial hyphae encircling the basal cell of the conidiophores and on the second to fourth septa of the main axis which usually lacks setose branches; or on the setose branches, with a conspicuous, 3.0–3.5 µm wide, 1.0–1.5 µm deep, infundibuliform collarette. **Conidia** accumulating in a white, slimy mass, falcate to allantoid, aseptate, hyaline, smooth, 12–15 × 2 µm, with a filiform appendage at each end, appendages 6–11 µm. **Teleomorph**: unknown.

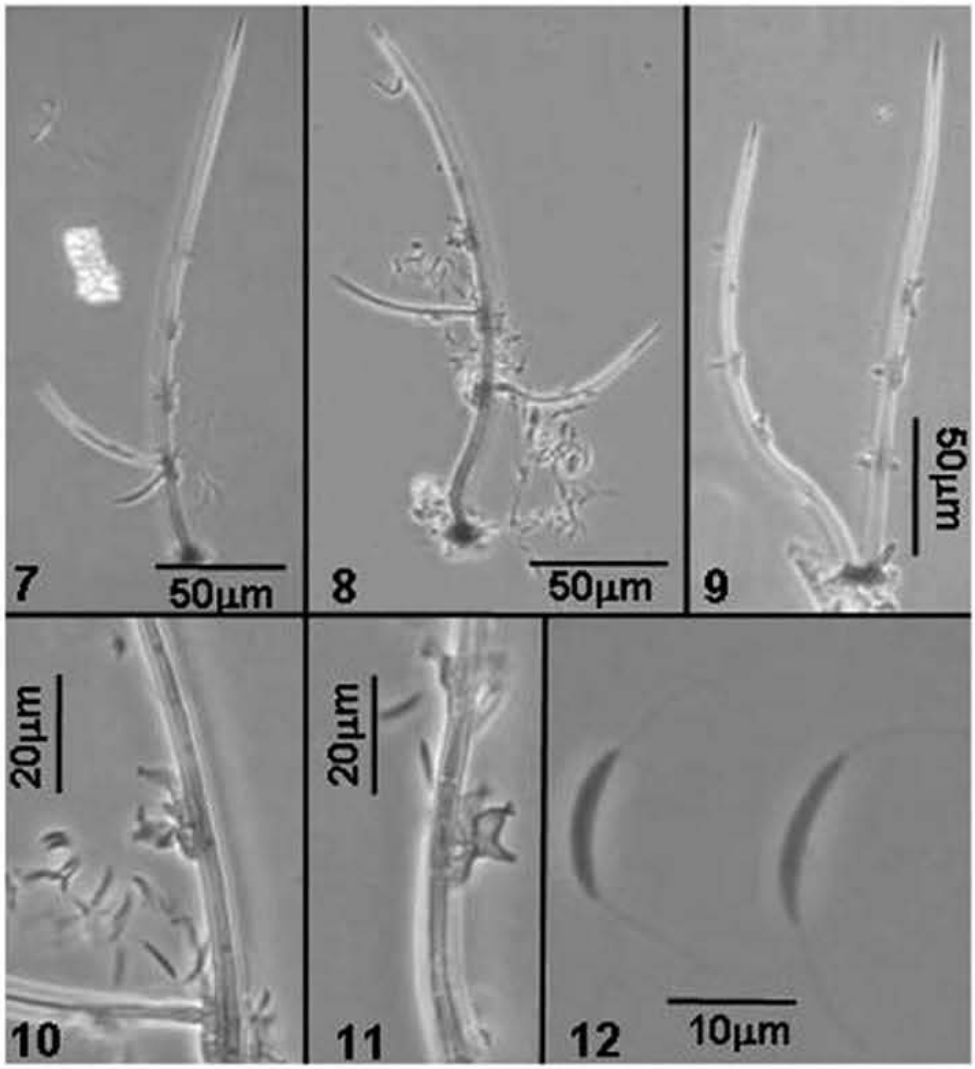
Matrix: **BRAZIL. Rio de Janeiro, Rio de Janeiro**, "Morro do Corcovado", on decaying leaves of unidentified plant. Coll. A.M. Stchigel and J. Guarro, 12.X.2002, (INIFAT C02/57-3). **HOLOTYPE**: CBS H-6586a. **ISOTYPE**: HUEFS120867.

***Dictyochaetopsis gonytrichoides*** (Shearer & J.L. Crane) Whitton, McKenzie & K.D. Hyde, Fungal Diversity 4: 156 (2000). **Figs 7-12**

Colonies on the natural substratum effuse, amphigenous, brown. Conidiophores setose, macronematous, mononematous, unbranched or branched with some sparsely or nodose setose branches and 2-7-fascicles of verticillate conidiogenous cells. Conidia accumulating in white, slimy mass, allantoid, 14–17 × 2.5–3.0 µm, hyaline, smooth, with a filiform appendage at each end, 6–10 µm long.

Specimen examined: **BRAZIL. Piauí, Caracol**, "Serra das Confusões", on decaying leaves of unidentified plant. Coll. J.R. Rocha, 01.IV.2006. HUEFS105759. (Slide preparation).

COMMENTS: The genus *Dictyochaetopsis* was introduced by Arambarri & Cabello (1990) for species of *Dictyochaeta,* which produce enterogenous, lateral, sessile, lageniform, cylindrical to subulate, mostly discrete, sometimes integrated conidiogenous cells on setiform conidiophores. Twelve accepted species were treated in a taxonomical account by Whitton et al. (2000) and subsequently another species was added, *D. brasiliensis* M. Calduch et al. (Calduch et al. 2002). *D. polysetosa* is similar to *D. gonytrichoides* (Whitton et al. 2002) in conidial morphology, but differs in its setose conidiophores; the latter has sometimes setose branches that are pale brown to brown, sparse or nodose around the septa, and develop terminal conidiogenous cells. Conidiogenous cells in *D. gonytrichoides* are polyphialidic, mostly cylindrical or lageniform, mostly recumbent and brown, whereas *D. polysetosa* has acicular, setose branches that




**Figs. 7-12.** *Dictyochaetopsis gonytrichoides* (From HUEFS105759). **7-9.** General aspect of branched setiform conidiophores. **10-11.** Detail of conidiogenous cells. **12.** Conidia.
Scale is indicated by bars.

are dark brown to black, verticillate at the upper part of the conidiophores, and remain sterile.

***Myrothecium compactum*** **R.F. Castañeda, Gusmão, Stchigel & M. Stadler, anam. sp. nov.** **Figs 13-20**

MycoBank #: MB 492172

*Ad omnibus speciebus Myrothecii differt in conidiomatis compactis, mucosis, atroviridis ad usque nigris, 280–450 diam. et hyphis marginalibus coarctatis, grossis rugosis vel tuberculatis, atrobrunneo-viridis, apicibus capitulatis vel clavatis, 5–8 µm crassis. Conidia*

*bacilliformia, brunneo-olivacea, unicellularia, 7–8 × 1.5 µm, in massa nigra vel atroviridis, permucosa congesta.*

**Etymology:** Latin, *compactum*, referring to the compact arrangements of marginal hyphae and conidiogenous structures within the conidiomata.

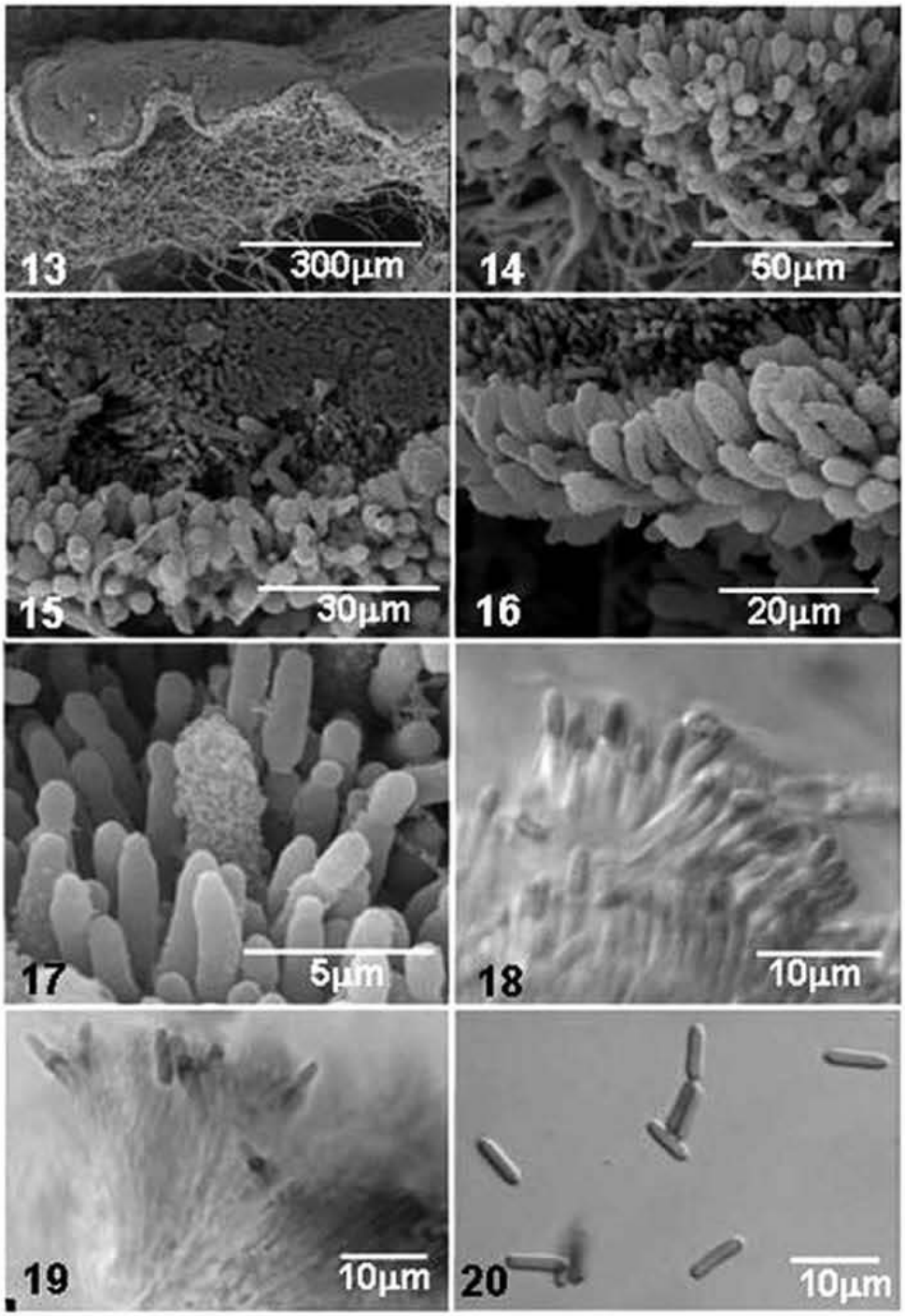
**Conidiomata** on the natural substratum sporodochial, scattered, amphigenous, pulvinate, gelatinous, dark green-brown to greenish black, 280–450 diam. **Mycelium** superficial and immersed. **Hyphae** septate, branched, 1.5–2.5 µm, hyaline to subhyaline, smooth. **Marginal hyphae** septate, unbranched or branched, capitate to clavate at the apex, coarsely rugose or tuberculate, pale brown-green below, dark brown green at the apex, very compactly grouped, encircling the conidiomata, 5–8 µm wide. **Conidiophores** macronematous, numerous, septate, tightly aggregated, 25–33 × 2–3 µm, subhyaline to pale green brown at the apex, smooth. **Conidiogenous cells** enterogenous, discrete, determinate, penicillate, unilocal, cylindrical to slightly subulate, with an inconspicuous opening, slightly brown-green, smooth; forming a very compact turf. **Conidia** accumulating in a black to darkish green, very gelatinous mass, holoblastic, bacilliform, aseptate, dark olivaceous-brown, smooth, 7–8 × 1.5 µm. **Teleomorph:** unknown.

Matrix: BRAZIL, Rio de Janeiro, Rio de Janeiro, "Pista Cláudio Coutinho", near Pão de Açúcar, on decaying leaves of unidentified plant. Coll. A.M. Stchigel and J. Guarro, 12.X.2002, (INIFAT C02/63). HOLOTYPE: CBS H-6584a. ISOTYPE: HUEFS120874 (slide preparation).

Culture from the holotype: **Colonies** on corn meal agar 25–31 mm after 7 days at 25° C, floccose, greenish gray. **Reverse** gray. **Hyphae** thick walled, septate, hyaline, 1.5–2.5 µm diam, smooth. **Sporulation** obtained after five days, conidia similar to those observed *in vivo*.

Cultures deposited: CBS 111739, IMI 390539.

Comments: The genus *Myrothecium* Tode: Fr. was treated in a taxonomic account by Tulloch (1972). The generic concept is broadly applied to taxa with sporodochial and synnematous conidiomata with or without marginal hyphae, mostly hyaline setae, sometimes stained by the pigment present in the mucilaginous mass of conidia. Conidiogenous cells are arranged in most of the species on somewhat penicillate conidiophores that form a turf in the pulvinate conidiomata. *M. compactum* can be compared with species that have ornamented marginal hyphae and non-setose conidiomata, such as *M. longistriatisporum* Matsush. (Matsushima 1971), *M. masonii* M. C. Tulloch (Tulloch 1972) and *M. renaudii* Escalona (Escalona 1997). However, *M. longistriatisporum* has fusiform, longitudinally striate, conidia, 12–16 × 3–4 µm; *M. masonii* has long, synnematal conidiomata and ellipsoid, subhyaline to pale olivaceous conidia, 4.5–9.0 × 1.5–2.5 µm; *M. renaudii* has synnematous conidioma and cylindrical to oblong, subhyaline to pale olivaceous conidia, 7–19 × 3–6 µm.




**Figs. 13-20.** *Myrothecium compactum* (From CBS H-6584a). **13.** General aspect of sporodochia (SEM). **14-16.** Mass of conidia and marginal hyphae (SEM). **17.** Detail of verrucose or tuberculate marginal hyphae (SEM). **18-19.** Conidia production and tightly aggregated conidiogenous cells. **20.** Conidia. Scale is indicated by bars.

## Acknowledgements

We are deeply indebted to Prof. Lori M. Carris (Washington State University) and Dr. Mary Palm (APHIS, United States Department of Agriculture) for kindly reviewing the manuscript and for many suggestions that greatly improved it. We thank Ciencia y Tecnología para el Desarrollo (CYTED RED-XII.J) and the Cuban Ministry of Agriculture for facilities. The author RFCR thanks Drs. David W. Minter, Pedro Crous, Uwe Braun, Ludmila Marvanová, Cony Decock, Jerry A. Cooper, Rosa M. Arias, Antonio Hernández-Gutiérrez and Josep Cano for their generous and valuable assistance with literature not available in our library. The author LFP Gusmão thanks the CNPq (proc. 471619/2004) for financial support.

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# RAPD patterns in three Argentine *Coprotus* species: a test case

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**Abstract** - The genus *Coprotus* consists of coprophilous fungi in the *Pyronemataceae*. 44 monosporic strains were morphologically identified as *Coprotus lacteus*, *C. niveus*, and *C. sexdecimsporus*. Using random amplified polymorphic DNA (RAPD) to analyse two *C. lacteus*, five *C. niveus*, and two *C. sexdecimsporus* geographical groups, the authors obtained diagnostic bands for species recognition, estimated polymorphism levels within species, analysed inter-species phenetic relationships, and compared the findings from the RAPD study to those from a previous isozyme study. The phenogram obtained from Nei's genetic distances matrix was similar to the one previously obtained by isozyme markers. Both matrices show strains grouping into three defined clusters, each representing one of the three species analysed. The difference between the similarity levels among species and those observed in the isozyme report are compared and discussed. The results of this study support the use of RAPD markers as a reliable tool comparable to traditional morphological and isozyme methods.

**Key words** - *Ascomycota*, taxonomy

## Introduction

The genus *Coprotus* Korf ex Korf & Kimbr. includes coprophilous discomycetes fungi, which belong to the *Pyronemataceae* Corda. From an ecological point of view, these fungi are very important since they have the ability to grow saprophytically on herbivore dung transforming it into humus. They also have the ability to perform the biological degradation of cellulose (Wicklow et al. 1980), which contributes to soil fertilization.

Coprophilous fungi have a cosmopolitan distribution due to the dispersion of their spores by animals, wind and water, and also to the fact that spores often adhere to forage and are usually transported over long distances.

Most of these fungi are homothallic, probably originating as an adaptation to the substrate, which determines their sexual isolation (Wicklow 1981). Sexual reproduction enables ascospore dispersal by effective discharge methods.

Traditional characterization of *Coprotus* species is based on cytological and morphological characters (Kimbrough et al. 1972). However, in many cases, the analyses of these characters have led to uncertainties in the identification of some species, because they often overlap and some of them vary according to culture conditions.

In an earlier study, Suárez et al. (2006) performed an isozyme analysis in order to confirm previous morphological identification of *Coprotus* species and to detect intraspecific variability. In that study, the isozymes showed scarce intraspecific variability and confirmed the previous identification of the strains using morphological characters, although only few species-specific bands were found.

In order to obtain a greater number of diagnostic bands, which could allow a better differentiation of the *Coprotus* species, new molecular markers seem to be necessary.

During the last decade, molecular techniques have been widely used as alternative methods for taxonomic studies, providing important tools for species characterization (Maclean et al. 1993). Among the wide variety of techniques, the random amplified polymorphic DNA (RAPD) (Williams et al. 1990) is a powerful tool for rapid detection of DNA polymorphisms among species.

This method has also been successfully used in taxonomic and evolutionary studies of fungi (Crowhurst et al. 1991, Aufauvre-Brown et al. 1992, Khush et al. 1992, Mills et al. 1992, Smith et al. 1992, Vaillancourt & Hanau 1992, Jacobson et al. 1993, Yoon & Glawe 1993, Cooke et al. 1996, Glienke-Blanco et al. 2002, Frazzon et al. 2002, Doherty et al. 2003).

Individuals of the genus *Coprotus* are homothallic and produce clonal offspring (Rayner 1994), which leads to low morphological and isoenzymatic variability.

Taking into account that a large number of amplicons can be screened in a relatively short period, RAPD is especially useful for the characterization of clonal lineages and for establishing phenetic relationships among species with scarce morphological differentiation (McDonald 1997).

This study represents the first report on RAPD markers able to differentiate three species of the genus *Coprotus*. We applied RAPD technique with the purpose of i) estimating levels of polymorphism within species, ii) obtaining diagnostic bands for species recognition, iii) analyzing the phenetic relationships among species and iv) comparing and contrasting the findings of the RAPD study to that of a previous isozyme study.

## Materials and methods

### Isolation and maintenance of monosporic strains

Mature apothecia of *Coprotus lacteus* (Cooke & W. Phillips) Kimbr. et al. and *Coprotus niveus* (Fuckel) Kimbr. et al. (Kimbrough et al. 1972), and *Coprotus sexdecimsporus* (P. Crouan & H. Crouan) Kimbr. & Korf (Kimbrough & Korf 1967) were obtained by placing cow and horse dung from different geographical locations in Petri dishes with a layer of filter paper. The isolation and maintenance of monosporic strains followed the procedure indicated by Suárez et al. (2006). Forty-four monosporic strains, from each geographical location were obtained (Table 1). All strains were deposited in the Herbarium and Culture Collection of the Department of Biodiversity, Faculty of Natural & Exact Sciences, University of Buenos Aires (BAFC).

Table 1. List of strains with their geographical location*, substrate, and BAFC number.

| STRAIN | LOCATION | SUBSTRATE | BAFC |
|---|---|---|---|
| | *COPROTUS LACTEUS* | | |
| lacA1 | Agronomía | cow dung | 874 |
| lacA2 | Agronomía | cow dung | 1936 |
| lacA3 | Agronomía | cow dung | 1937 |
| lacA4 | Agronomía | cow dung | 1938 |
| lacA5 | Agronomía | cow dung | 1939 |
| lacA6 | Agronomía | cow dung | 1940 |
| lacA10 | Agronomía | cow dung | 1941 |
| lacA13 | Agronomía | cow dung | 1942 |
| lacA14 | Agronomía | cow dung | 1943 |
| lacL1 | Villa Lugano | cow dung | 1944 |
| lacL3 | Villa Lugano | cow dung | 1945 |
| lacL4 | Villa Lugano | cow dung | 1946 |
| lacL6 | Villa Lugano | cow dung | 1947 |
| | *COPROTUS SEXDECIMSPORUS* | | |
| sexG1 | Los Gigantes | cow dung | 1948 |
| sexG2 | Los Gigantes | cow dung | 873 |
| sexG3 | Los Gigantes | cow dung | 1949 |
| sexG4 | Los Gigantes | cow dung | 1950 |
| sexG7 | Los Gigantes | cow dung | 1951 |
| sexU1 | Ciudad Universitaria | horse dung | 1952 |
| sexU2 | Ciudad Universitaria | horse dung | 1953 |
| sexU4 | Ciudad Universitaria | horse dung | 1954 |
| sexU5 | Ciudad Universitaria | horse dung | 1955 |
| | *COPROTUS NIVEUS* | | |
| nivE1 | Bahía Ensenada | cow dung | 1956 |
| nivE2 | Bahía Ensenada | cow dung | 982 |
| nivE3 | Bahía Ensenada | cow dung | 1957 |
| nivE4 | Bahía Ensenada | cow dung | 1958 |
| nivE5 | Bahía Ensenada | cow dung | 1959 |
| nivC2 | Campana | cow dung | 1960 |
| nivC3 | Campana | cow dung | 1961 |
| nivC4 | Campana | cow dung | 1962 |
| nivC5 | Campana | cow dung | 1963 |
| nivU1 | Ciudad Universitaria | horse dung | 1964 |
| nivU3 | Ciudad Universitaria | horse dung | 1965 |
| nivU6 | Ciudad Universitaria | horse dung | 1966 |
| nivU7 | Ciudad Universitaria | horse dung | 1967 |
| nivU8 | Ciudad Universitaria | horse dung | 1968 |
| nivBC1 | Bahía Craft | cow dung | 1969 |
| nivBC2 | Bahía Craft | cow dung | 1970 |
| nivBC3 | Bahía Craft | cow dung | 1971 |
| nivBC4 | Bahía Craft | cow dung | 1972 |
| nivL1 | Villa Lugano | cow dung | 1973 |
| nivL3 | Villa Lugano | cow dung | 1974 |
| nivL4 | Villa Lugano | cow dung | 1975 |
| nivL5 | Villa Lugano | cow dung | 1976 |

*BAHÍA CRAFT: Villa La Angostura, Neuquén province; BAHÍA ENSENADA: Tierra del Fuego province; CAMPANA: Buenos Aires province; LOS GIGANTES: Córdoba province, Agronomía, Ciudad Universitaria and Villa Lugano are different locations in Buenos Aires city

### Identification of species

The characters used to identify the *Coprotus* species were the same used by Kimbrough et al. (1972). Other characters like the distribution and appearance of apothecia in PF medium (yeast extract, 3 g; agar 18 g; distilled water, 1000 ml; a slice of filter paper) (Ranalli & Forchiassin 1974) and the germination of ascospores in AA medium (Bacto agar, 20g; distilled water, 1000 ml) were also used. Studies of spore germination and ascocarp development were performed on AA plates and in PF medium plates respectively. The latter were incubated at 23 °C under continuous light.

### DNA extraction

The mycelium was obtained using the Suárez et al. (2006) procedure and ground to powder in liquid nitrogen using a sterile pestle. Genomic DNA was extracted from the mycelium according to the method of Dellaporta et al. (1983).

DNA concentration was estimated by comparing electrophoretic patterns on 0.8 % agarose (in 1x TAE buffer) gels with standard DNA marker sets (phage λ) double digested with EcoRI and HindIII.

### Polymerase chain reaction

PCR was carried out in 50 µl final volume using 40-60 ng of genomic DNA, 5 µl dNTP mix (100 µM), 6 µl $MgCl_2$ (25 mM), 10 µl primer (3 ng/µl), 5 µl 10x Taq DNA polymerase buffer (10mM tris-HCl, pH 9 at 25 °C, 50 mM KCl, 0.1% Triton X-100) and 0.5 µl of Taq DNA polymerase (Promega) (0.5 units/µl). The mixture was amplified in a thermal cycler (Techne Gene E). The thermal cycler was programmed for one cycle of 94 °C for 6 min., 45 cycles of 94 °C for 1 min., 36 °C for 1 min. and 72 °C for 2 min., followed by a final extension step of 72 °C for 6 min.

Two PCR amplifications per individual were carried out to ensure repeatability of banding patterns. The usual caution needed to prevent contamination of PCR experiments with previously amplified fragments was considered. In particular, pre and post-amplification procedures were separated and fresh aliquots of reagents were used for each experiment wherever possible. To test the reliability of PCR products, several controls were routinely used: one without primer, a second one without Taq DNA polymerase, and the third with no genomic DNA. No amplification occurred in any of these controls.

Twenty arbitrary primers (Promega, series A and B) were screened for suitability in a small number of individuals. Only those primers that produced clear RAPD bands for all of the fungal isolates and in addition resulted the most informative to detect polymorphisms were chosen. These primers were: A01 (5′CCC AAG GTC C3′), A02 (5′GGT GCG GGA A3′), A10 (5′ACG GCG TAT G3′), B08 (5′TCA CCA CGG T3′), B09 (5′ATG GCT CAG C3′) and B10 (5′CAG GCA CTA G3′). The 85 bands that appeared consistently in all experiments were used in the study.

### DNA fragment analysis

The amplification products were resolved on 1.4% agarose gels stained with EtBr (0.5µg/ml). Fragments were observed and photographed on a UV transilluminator (312 nm).

### Statistical methods

The nine geographical groups of strains of *C. lacteus* (lacA and lacL), *C. niveus* (nivU, nivC, nivBC, nivL and nivE) and *C. sexdecimsporus* (sexG and sexU) were considered as operative taxonomic units (OTUs), because no RAPD pattern variation was detected among monosporic strains of the same geographic location.

RAPD bands were scored as presence (1) or absence (0). The matrix data was used to estimate Nei's (1978) genetic distance. Calculations were made using the RAPD programs by Black IV (1996). A phenogram was obtained from Nei's distance matrix by the UPGMA method using the program STATISTICA (ver. 6.0) (Statsoft, Inc. 2001). The reliability of its branches was evaluated by the analysis of consensus of 100 trees obtained from 100 bootstrapped pseudoreplicates of the original data matrix. Bootstrapping was performed using Black IV (1996) programs, and the consensus analysis was done with the PHYLIP program (Felsenstein 1993). The approach used to estimate the levels of polymorphism for each species was the percentage of polymorphic loci (P = N° of polymorphic loci/ N° of loci analyzed).

## Results

### RAPD genetic variability

The analysis of six primers yielded a total of 91 bands. Only 85 of these bands were variable and useful for comparison among species and were used in this study. Figures 1 and 2 show RAPD patterns obtained with primers A02 and B09.

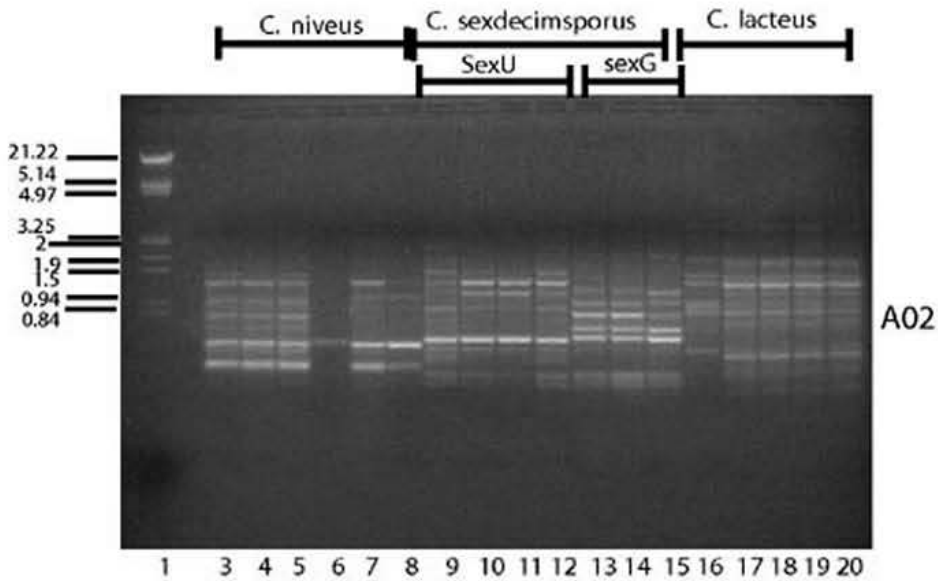


Figure 1. Amplification products generated with primer A02 in *Coprotus* species. Lane 1. Lambda DNA digested with EcoRI /HindIII used as molecular weight marker. Lanes 3-8: Strains of *C. niveus*; Lanes 9-15: Strains of *C. sexdecimsporus*; Lanes 16-20: Strains of *C. lacteus*.

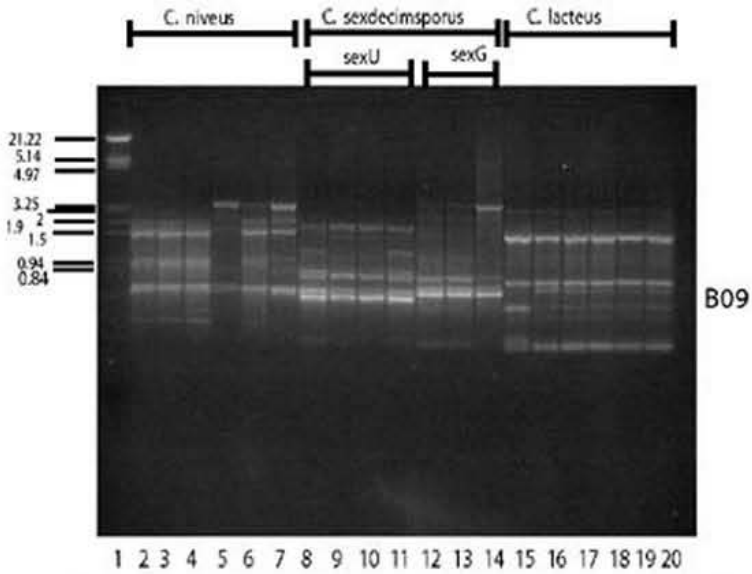


Figure 2. Amplification products generated with primer B09 in *Coprotus* species. Lane 1: Lambda DNA digested with EcoRI/HindIII used as molecular weight marker; Lanes 2-7: Strains of *C. niveus*; Lanes 8-14: Strains of *C. sexdecimsporus*; Lanes 15-20: Strains of *C. lacteus*.

All the species were characterised by band combinations. Twenty-six bands provided a reliable tool for the identification of the species studied in this paper (Table 2). Bands A02-2, A02-4, A02-11, B08-1, B09-2, B09-6′, B09-11, B10-11, B10-12 and B10-14 were present in all the strains of *C. lacteus* and absent in *C. niveus* and *C. sexdecimsporus*. Bands A02-14, A02-17, A10-2, A10-9, A10-10. B09-4, B09-6, B10-4, B10-9 and B10-10 were observed in strains of *C. niveus* and absent in *C. lacteus* and *C. sexdecimsporus*. Bands A2-13, A02-18, A02-19, A10-5, A10-11 and B08-3 were present in *C. sexdecimsporus* and absent in *C. lacteus* and *C. niveus*.

Genetic variability within species, measured in terms of percentage of polymorphic loci was low, being *C. sexdecimsporus* (P = 25%) and *C. niveus* (P = 17 %) the most variable species, and *C. lacteus* the least variable (P = 14%).

### Genetic distance and cluster analysis

RAPD patterns of strains of each geographical group within species were almost identical for *C. lacteus* and *C. niveus*. This was reflected in the low genetic distances among geographical groups within each species (D = 0.03 between *C. lacteus* geographic groups and D = 0.02-0.03 between *C. niveus* geographic groups). In contrast, *C. sexdecimsporus* showed the highest genetic distance (D = 0.4) among geographical groups (Table 3).

Genetic distances among species were high (D = 0.68 between *C. lacteus* and *C. sexdecimsporus* and D= 0.75 between *C. niveus* and the cluster formed by *C. lacteus* and *C. niveus*). The phenogram obtained by means of UPGMA

Table 2. Distribution of RAPD species-specific bands in *Coprotus* using primers (A02, A10, B8, B9, B10) across nine geographical groups (OTUs).

| Bands | *C. LACTEUS* | | *C. SEXDEC.* | | *C. NIVEUS* | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | lac A | lac L | sex G | sex U | niv U | niv C | niv BC | niv L | niv E |
| A02-2 | + | + | - | - | - | - | - | - | - |
| A02-4 | + | + | - | - | - | - | - | - | - |
| A02-11 | + | + | - | - | - | - | - | - | - |
| A02-13 | - | - | + | + | - | - | - | - | - |
| A02-14 | - | - | - | - | + | + | + | + | + |
| A02-17 | - | - | - | - | + | + | + | + | + |
| A02-18 | - | - | + | + | - | - | - | - | - |
| A02-19 | - | - | + | + | - | - | - | - | - |
| A10-2 | - | - | - | - | + | + | + | + | + |
| A10-5 | - | - | + | + | - | - | - | - | - |
| A10-9 | - | - | - | - | + | + | + | + | + |
| A10-10 | - | - | - | - | + | + | + | + | + |
| A10-11 | - | - | + | + | - | - | - | - | - |
| B08-1 | + | + | - | - | - | - | - | - | - |
| B08-3 | - | - | + | + | - | - | - | - | - |
| B09-2 | + | + | - | - | - | - | - | - | - |
| B09-4 | - | - | - | - | + | + | + | + | + |
| B09-6 | - | - | - | - | + | + | + | + | + |
| B09-6' | + | + | - | - | - | - | - | - | - |
| B09-11 | + | + | - | - | - | - | - | - | - |
| B10-4 | - | - | - | - | + | + | + | + | + |
| B10-9 | - | - | - | - | + | + | + | + | + |
| B10-10 | - | - | - | - | + | + | + | + | + |
| B10-11 | + | + | - | - | - | - | - | - | - |
| B10-12 | + | + | - | - | - | - | - | - | - |
| B10-14 | + | + | - | - | - | - | - | - | - |

* + = presence - = absence

Table 3. Nei's genetic distances between geographical groups.

| | lacL | lacA | sexG | sexU | nivU | nivE | nivC | nivL | nivBC |
|---|---|---|---|---|---|---|---|---|---|
| lacL | 0.00 | | | | | | | | |
| lacA | 0.03 | 0.00 | | | | | | | |
| sexG | 0.63 | 0.62 | 0.00 | | | | | | |
| sexU | 0.74 | 0.73 | 0.40 | 0.00 | | | | | |
| nivU | 0.73 | 0.72 | 0.64 | 0.78 | 0.00 | | | | |
| nivE | 0.77 | 0.74 | 0.62 | 0.77 | 0.02 | 0.00 | | | |
| nivC | 0.82 | 0.80 | 0.67 | 0.80 | 0.02 | 0.02 | 0.00 | | |
| nivL | 0.78 | 0.75 | 0.63 | 0.78 | 0.02 | 0.01 | 0.01 | 0.00 | |
| nivBC | 0.79 | 0.77 | 0.65 | 0.79 | 0.02 | 0.01 | 0.01 | 0.00 | 0.00 |

method (Figure 3) showed that all the strains were grouped into three defined clusters, which represented the three species analysed.

These clusters were highly supported by the analysis of consensus of 100 trees obtained from 100 bootstrapped pseudoreplicates of the original data

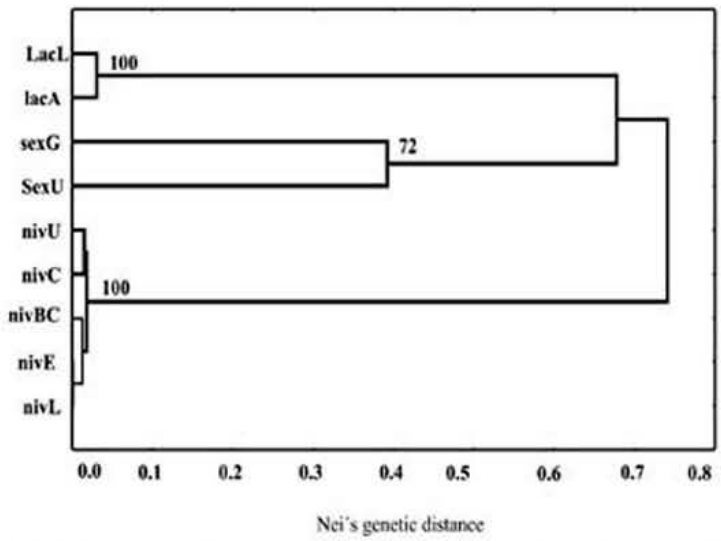


Figure. 3. UPGMA tree showing relationships of *Coprotus* species on the basis of Nei's genetic distance estimated from RAPD loci. Number over branches represents bootstrap for each node.

matrix. The two clusters involving *C. lacteus* and *C. niveus* geographical groups showed 100% bootstrap support and the third cluster of *C. sexdecimsporus* geographical groups showed a value of 72 % (Figure 3).

## Discussion

In a previous isozyme study, Suárez et al. (2006) observed low isozyme variability within species of the genus *Coprotus* by studying eleven isozyme systems. In this paper a high number of RAPD bands were analysed, which confirmed earlier results. The high intraspecific homogeneity detected by biochemical (Suárez et al. 2006) and RAPDs markers could be related to the fact that *Coprotus* is homothallic and produces clonal offspring (Rayner 1994). Anderson et al. (1992) stated that fungi have a wide array of reproductive strategies, which have an impact on their genetic variability. Therefore, measure of genetic diversity could be a potential indicator of the relative abundance of sexual and asexual reproduction (Chen & McDonald 1996). While high genetic variability was described for fungal species with sexual reproduction (Wang et al. 1997), low genetic polymorphism was found in species with asexual reproduction (Zeigler et al. 1995).

In non-out crossing type (homothallic) of a haploid organism, polymorphism could be the consequence of different evolutionary forces. Mutation and

natural selection could lead to genetic divergence and therefore increase genetic variability levels in the homothallic (non-out crossing populations). In *Coprotus* species, RAPD patterns of strains of the same geographical groups were almost identical, which could support the hypothesis that time was not enough to allow differentiation.

Another interesting result is that RAPD technique provided twenty-six RAPD bands that were useful to recognise the species. This result contrasts with previous isozyme studies (Suárez et al. 2006), where ALP was the only system that showed a diagnostic electromorph for each species. Moreover, RAPD have a better ability to differentiate species than isozymes because they allow an extensive random sampling of the genoma providing a great number of DNA markers and therefore a higher number of species specific markers.

The difference between isozyme and RAPD markers was also reflected in the phenograms that represent phenetic relationships among species. Although in both phenograms all strains of the same species are grouped into three defined clusters, some disagreements can be observed. While the phenogram obtained from isozymes datasets (Suárez et al. 2006) showed a higher association between *C. niveus* and *C. lacteus* species, which is consistent with the morphologic similarity, in the present RAPD tree *C. lacteus* and *C. sexdecimsporus* are joined in a cluster, which is linked to *C. niveus*. This disagreement might be related to the different nature of these markers. RAPDs are usually considered neutral markers without known physiological function, whereas an adaptative significance of isozyme and morphological characters can not be ruled out because they are exposed to selective pressure.

Previous isoenzymatic studies (Suárez et al. 2006) showed a low intraspecific variability and characterized the *Coprotus* species by some isoenzymatic bands. In this paper, RAPD technique both effectively differentiated three *Coprotus* species by providing a greater number of species specific bands and confirmed the low isoenzymatic intraspecific variability.

This technique may therefore be considered a powerful and reliable taxonomic tool to add to traditional morphological and isozyme analytical methods.

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# *Alternaria roseogrisea*, a new species from achenes of *Helianthus annuus* (sunflower)

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**Abstract** — *Alternaria roseogrisea* was isolated from the achenes of *Helianthus annuus* during studies conducted in 1983-85 to document the diversity of fungi occurring in sunflower seeds and the possible role these fungi play in degradation of oilseed quality. *A. roseogrisea* was reported as "*Alternaria* sp. 2" in this early study because there were no published descriptions in *Alternaria* to accommodate it, a deficiency that has persisted. More recently, cluster analyses of combined RAPD, RAMS (microsatellite) and AFLP fingerprints support the placement of *A. roseogrisea* within the *A. infectoria* species-group sensu Simmons and its segregation from other members of this group. *A. roseogrisea* is known only from high-quality oil-type sunflower seed, and is recognized by the grayish-pink color of mature colonies on 6% NaCl-tomato juice agar and the striking appearance of the very small, often one or two-celled conidia separated by secondary conidiophores that can be many times the length of and nearly as wide as the conidia themselves.

**Key words** — hyphomycetes, sporulation pattern, *Alternaria infectoria*

## Introduction

During studies of fungi occurring in the achenes of *Helianthus annuus* L. in 1983-85, an unusual *Alternaria* was regularly isolated from samples of high-quality oilseed-type sunflower achenes that originated from the upper Midwestern U.S.A. An expert opinion on the identity of this fungus was sought from Dr. E. G. Simmons, who indicated it was representative of an undescribed taxon among a group of undescribed taxa that were *Alternaria* anamorphs of *Pleospora* (personal communication). Barr & Simmons subsequently described *Lewia* M.E. Barr & E.G. Simmons to accommodate those *Pleospora* species with small ascomata and ascospores and (where known) *Alternaria* anamorphs (Simmons, 1986b). Now, some 21 years later, description of this interesting and unusual fungus resolves a lingering deficiency in our knowledge of *Alternaria* species from *Helianthus*.

## Materials and methods

### Conditions of isolation, culture and observation

During 1983-85, isolates of *A. roseogrisea* were obtained with ease and regularity from samples of Grades No. 1 and No. 2 sunflower seeds that originated in the upper Midwestern U.S., which were provided by the U.S. Department of Agriculture, Federal Grain Inspection Service. Sunflower achenes were surface disinfested and plated onto 6% sodium chloride-tomato juice agar (TJ) as described previously (Roberts et al. 1986). Representative isolates were preserved as lyophilized conidial suspensions held at -20 C. The media, methods and conditions for growth and observation of *Alternaria* follow those described in pages 136-137 in Simmons & Roberts (1993). Three isolates, RGR 85.0034, RGR 85.0035 and RGR 85.0036, were revived onto potato carrot agar (PCA, Simmons & Roberts, 1993) and incubated under standardized conditions for *Alternaria* (25 C under cool white fluorescent lighting, 8 hr/16 hr of light/dark, Simmons & Roberts, 1993) until growth reached the edges of the plates and sporulation was evident. Cultures were examined at 50x using a stereoscopic microscope to assess the pattern of sporulation, then double-stick tape was pressed onto the colony surface and then mounted spore side up in several drops of lactic acid on a microscope slide for observation at higher magnifications. Microscopic morphology was recorded digitally using a Zeiss Axiocam Mr5a camera and a Zeiss Axioplan microscope. Color references in the taxonomic descriptions are to the corresponding color plates in the Methuen Handbook of Colour (Kornerup & Wanscher, 1989).

### Molecular analysis

Cultures were grown and DNA prepared for analysis by the methods reported previously (Roberts et al. 2000 and Roberts, 2007). Methods and conditions for analysis of RAPD, RAMS and AFLP fingerprints followed Roberts et al. (2000) and Roberts (2007), to which the reader is referred for details.

## Taxonomic Description

***Alternaria roseogrisea*** R.G. Roberts **sp. nov.** Fig. 1

MycoBank MB 511161

Etym.: *rosea* - pink; + *grisea* - gray

*Ex cultura in agaro PCA descripta. Colonia tempore diei septem ca 60 mm diam, inconspicue concentrice zonata, Centrum coloniae atrogriseum, granulosum, sporulatione densissima; area peripherica coloniae griseoviridis. Hyphae aeriae funiculosae in strato minus quam 1 mm crasso. Conidiophora primaria plerumque lateralia ex hyphis aeriis, solitaria, 25-210 x 4-4.8 µm, plerumque ramosa, geniculata. Conidia globosa vel subcylindrica, solitaria vel catenulata, 4.8-38.4 x 3.2-18.7 µm, in catenis pseudorostrata. Conidia minima globosa, eseptata vel 1-2 transverse septata; conidia subcylindrica plerumque transeptata. Conidia maxima ellipsoidea vel obpyriformibus, 5(-7) transverse septata et septis longitudinalibus*

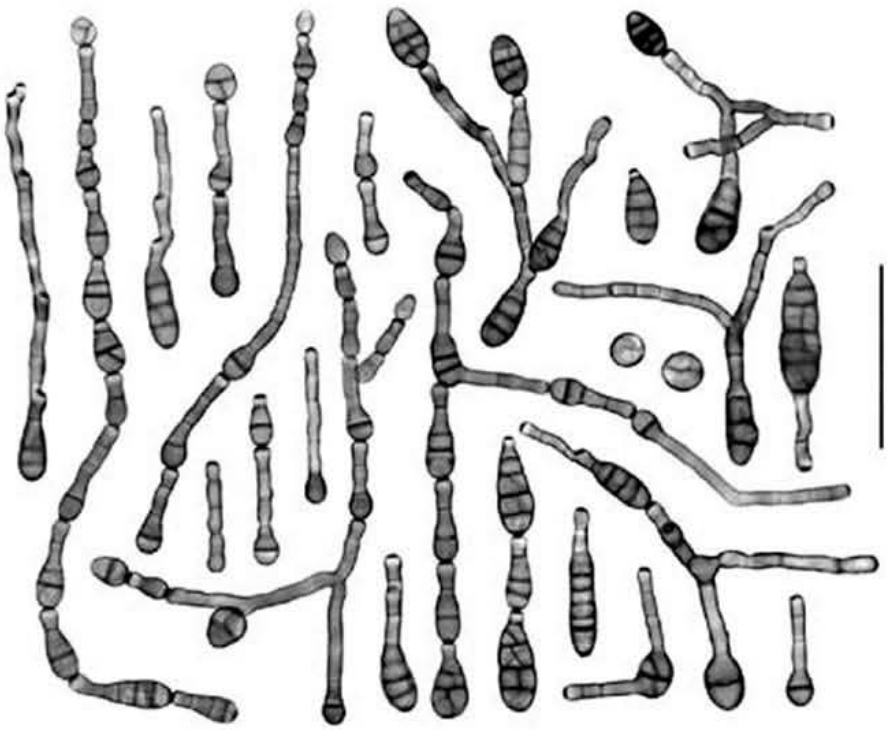

Fig.1. Conidia of *A. roseogrisea* from PCA. *Bar = 50* µm

*vel obliquis numeris varialibus. Conidiophora secundaria ex corpore conidiorum apicalibus vel lateralibus, 3.2-160 x 3.6-9.8 µm, saepe apice amplificato. Corpus conidiorum laeve vel punctatum, saepe verrucosum.*

Description from cultures grown on PCA for 7 days at 25 C and 21% relative humidity. Colonies about 60 mm diam., faintly and concentrically zonate. Sporulation most dense in center of colony, which is granular in appearance, centrally dark gray (1F1) to grayish green (1D4) at the outer portions of the colony. Primary conidiophores are 25-210 x 4-4.8 µm, mononematous, frequently branched, geniculate, developing predominantly as lateral branches from funiculose aerial hyphal elements that form a low turf less than 1 mm tall. Conidia globose to subcylindrical, solitary to catenulate, pseudorostrate when catenulate, 4.8-38.4 x 3.2-18.7 µm. Spore bodies are smooth to punctate, frequently verrucose. The smallest conidia are globose and aseptate or with 1-2 transverse septa. Subcylindrical conidia are predominantly transversely septate. The largest conidia are ellipsoid to obpyriform, with up to 5(-7) transverse septa and variable numbers of longitudinal septa and oblique septa. Secondary conidiophores develop apically or laterally from conidium bodies, 3.2-160 x

3.6-9.8 µm, frequently with an enlarged apex, especially when produced from globose conidia. When grown on TJ for 7 days, colonies are low, plane to faintly sulcate, with indistinct concentric zonation present in older portions but absent from areas of recent growth. Colonies of *A. roseogrisea* RGR 85.0036 on TJ are ca 59 mm diam, and the dull red (10B3, outer colony) to grayish brown (10F3, inner colony) color of the colony is the character for which the species is named and the reason for prior reference to *A. roseogrisea* as the "pink" *Alternaria* (Roberts et al. 1986).

Type (holotype): BPI 878240; (dried culture preparation ex RGR 85.0036, isol. Roberts from an achene of oil-type *Helianthus annuus*, Athens, GA, 1984; origin of achenes from U.S. Department of Agriculture, Federal Grain Inspection Service, North Dakota, USA. Ex-type culture deposited at Centraalbureau voor Schimmelcultures, Utrecht, The Netherlands as CBS 121921.

## Discussion

Roberts et al. (1986) reported a relative frequency of isolation of 0.012 for *A. roseogrisea* (as *Alternaria* sp. 2, "pink") from more than 28,000 oilseed-type sunflower achenes assayed. Assays of confectionery-type sunflower achenes during and after development in experimental plots in Georgia did not yield any isolates of *A. roseogrisea*. It may be inferred from this admittedly limited sampling that the distribution of *A. roseogrisea* is limited to sunflower production areas in the upper Midwestern U.S., but it would likely also be encountered in other temperate sunflower production areas (mostly between 45° and 50° N latitudes). The occurrence of *A. helianthinficiens* E.G. Simmons et al. in sunflower achenes from production areas in the U.S. and Europe supports this conjecture (Simmons, 1986a). *A. roseogrisea* is not known to be associated with any disease symptom of sunflower, and was associated by Roberts et al. (1986) with higher-quality seed samples (low free fatty acids and high germination percentages).

In Roberts (2007, Fig. 4), the dendrogram representation of a combined cluster analysis of RAPD, RAMS and AFLP fingerprints of *Alternaria* strains belonging in the *A. infectoria* species-group (Simmons & Roberts, 1993) showed the ex-type isolate of *A. incomplexa* E.G. Simmons clustered with isolate RGR 85.0034 of *A. roseogrisea* (as *Alternaria* sp. from *Helianthus annuus*). The central cluster in Fig. 4 contains three isolates of *A. arbusti* E.G. Simmons including the ex-type isolate, three isolates of *A. roseogrisea* including the ex-type isolate, and the *A. incomplexa* isolate. All three species are resolved within this central cluster save *A. incomplexa*, although inclusion of additional isolates of *incomplexa* in the analysis might reasonably be expected to pull RGR 96.0013 into an 'incomplexa' cluster. A similar situation was noted and discussed by Roberts et al. (2000), who presented a cluster analysis of a subset (28 isolates,

Fig. 4) of the 260 isolates used (Fig. 3). In the analysis of the 28-isolate subset, *A. arbusti* clustered with *A. gaisen* but when analyzed among all 260 isolates, which contained additional representative isolates, *A. arbusti* and *A. gaisen* Nagano were fully resolved. Roberts et al. (2000) therefore cautioned against "... inferring relationships between strains from RAPD fragment patterns because the topologies of the dendrograms from cluster analyses are influenced by the number of isolates and the representation within the group". Given that the present analysis is also a cluster analysis of a relatively small data set (e.g., a single isolate for *A. incomplexa*), a similar caution applies here. It is unfortunate that additional isolates of *A. incomplexa* were not available for analysis.

The author considers the inclusion of *A. incomplexa* in a cluster of *A. roseogrisea* isolates to be an artifact of sample size for reasons stated. As such, the current results do not qualify the conclusion that *A. roseogrisea* is recognizably distinct from other members of the *A. infectoria* species-group or the hundreds of isolates from all species-groups that have been studied over the years, and no practitioner with access to authenticated cultures of *A. roseogrisea* and *A. incomplexa* would have difficulty distinguishing between them. That *A. arbusti*, *A. roseogrisea* and *A. incomplexa* would cluster together is neither illogical nor problematic, as they are all representatives of the *A. infectoria* species-group. *A. roseogrisea* and *A. incomplexa* both produce a high percentage of spores that are relatively small for the genus and other members of the *A. infectoria* species-group; the smallest of the conidia typically exhibit 0-3 transverse septa and typically lack longitudinal septa. Similarly, although conidia of *A. arbusti* are larger and more robust than conidia of either *A. incomplexa* or *A. roseogrisea*, the pattern of relatively unbranched sporulation is among the simplest encountered in the *A. infectoria* species-group. *A. incomplexa* differs from *A. roseogrisea* in several characters (Simmons 1996), including larger and more ellipsoidal conidia and development of primary conidiophores from surface or subsurface hyphal elements versus aerial elements in *A. roseogrisea*.

## Acknowledgments

The constructive comments by Frank Dugan are greatly appreciated. Efforts by Emory Simmons since 1984 to help the author with this taxon have long been appreciated, and now even more so as he also provided the Latin description for which the author is especially grateful. Steve Reymond conducted the referenced molecular analyses and has provided ongoing techincal support, for which the author is indebted.

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# A new bluing species of *Psilocybe* (*Basidiomycota, Agaricales, Strophariaceae*) — the first record of section *Stuntzii* for Mexico

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**Abstract** — *Psilocybe meridionalis* is described as a new species from an oak-pine subtropical forest in the western mountains of the state of Jalisco, Mexico. Because of its bluing feature, annulus and subrhomboid basidiospores, this species belongs to section *Stuntzii*. The smaller spores and presence of pleurocystidia distinguish this species from the others in the section. This is the first record of a member of section *Stuntzii* from Mexico.

**Keywords** — new edition, *Psilocybe* monograph

## Introduction

The genus *Psilocybe* has been studied in Mexico since the 1950's (e.g., Heim & Wasson 1958, Singer & Smith 1958) when hallucinogenic mushrooms were first revealed. It is likely that there are still undescribed species of *Psilocybe* as indicated by Guzmán (1998) and Guzmán et al. (2005). Moreover, in the state of Jalisco there have been few mycological explorations despite on-going mushroom research carried out by Guzmán-Dávalos for more than 20 years (e.g., Guzmán-Dávalos et al. 1983). In this paper, we describe a new species of *Psilocybe* made by Sánchez-Jácome in an oak-pine subtropical forest in the western mountains of Jalisco.

## Materials and methods

Microscopic observations were made obtaining thin sections of the tissues of dried basidiomata. These sections were mounted in 5% $NH_4OH$, and 5%

KOH or with 1% Congo red solution previously treated with 96% alcohol for rehydrating. At least 25 measurements were taken of each microstructure, which was drawn with the aid of a light tube. Basidiospore sizes provided are length, width in face-view, and thickness in side-view.

## Results

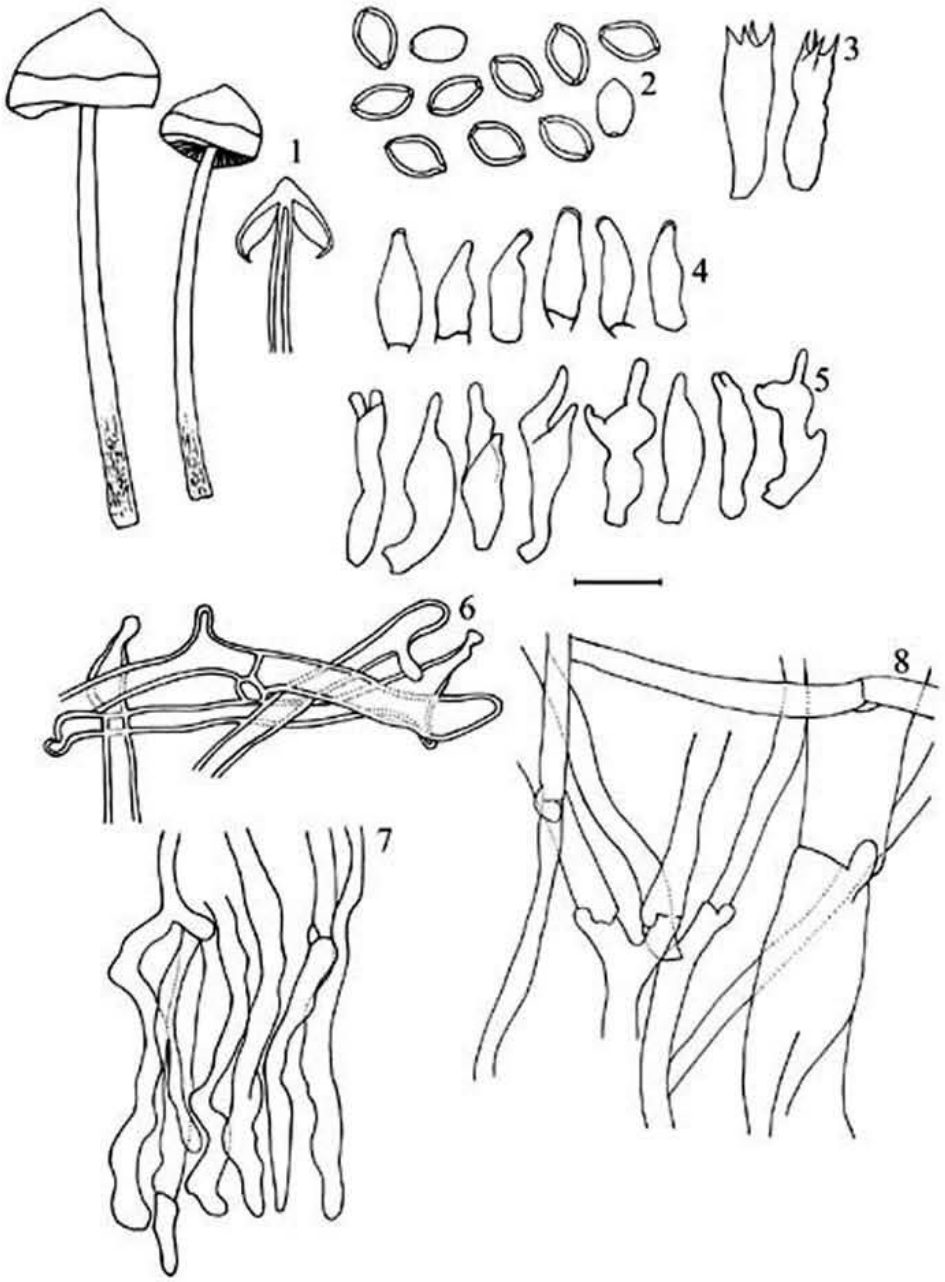
***Psilocybe meridionalis*** Guzmán, Ram.-Guill. & Guzm.-Dáv., **sp. nov.** **Figs. 1-8**
MycoBank MB 511238

*Etymology.* From southern region, related to the austral distribution in the Tropic of Cancer, as opposed to the more northern (United States) distribution for other species in section *Stuntzii*.

*Pileus 20-30(-40) mm latus, conicus vel campanulatus, glaber, rufobrunneus, hygrophanous, caerulescente. Lamellae adnate vel annexa, brunneus violaceous, marginis albidus, caerulescente. Stipes 85-100(-120) × 3-5 mm, concolor pileus, caerulescente. Annulus membranaceous, caerulescente. Sporae 6-7 (-8) × 4-5 × 4-5 µm, subrhomboideus vel subellipsoideus, pariete 1 µm crasis, brunneolus, poro germinali lato. Pleurocystidia 11-14 × 4-6 µm, hyaline, communis, ventricosus rostratus. Cheilocystidia 13-26 × 4-5.5 µm, hyaline, ventricosus rostratus, frequens ramus irregulariter. Pileipelles nullus gelatinous. Fibulae communis. Mexico, Jalisco, mountis Cacoma, prope Autlan, mensis september, Sánchez-Jácome 1163 (holotype IBUG).*

Pileus 20–30(–40) mm diam., conical to campanulate, with a slight papilla, smooth, light to dark reddish-brown, hygrophanous, changing to dark beige, dry specimens chocolate-brown, highly caerulescent to blackish, margin entire, appendiculate. Lamellae adnate to adnexed, pinkish-brown to dark violaceous-brown, with whitish and finely floccose margin (observed with a lens), caerulescent. Stipe 85-100(-120) × 3-5 mm, smooth but slightly scaly toward the base, concolorous with the pileus, hollow, with subbulbous base. Veil well developed, as a thick membrane covering the lamellae, which leaves a membranous annulus on the stipe and/or remains on the margin of the pileus, caerulescent. Context whitish to brownish, caerulescent. Odor and taste not checked.

Basidiospores 6-7 (-8) × 4-5 × 4-5 µm, subrhomboid in face-view, subellipsoid in side-view, thick-walled, wall up to 1 µm thick, yellowish-brown, with a broad germ pore in one end and a short hilar appendage on the other. Basidia 16-22 × 4-5.5 µm, 4-spored, hyaline, variable from oblong to ventricose-clavate, with a median constriction. Pleurocystidia 11-14 × 4-6 µm, hyaline, common, but difficult to find, it is necessary to observe them with Congo red, ventricose-rostrate, apex prolongation short, frequently with a dark apex by the reaction of the Congo red. Cheilocystidia 13-26 × 4-5.5 µm, hyaline, forming a sterile edge, ventricose-rostrate, with a short or long apical prolongation, frequently irregularly branched or irregular in form. Subhymenium subcellular, elements




**Figs. 1-8.** *Psilocybe meridionalis*. **1:** basidiomata, **2:** basidiospores, **3:** basidia, **4:** pleurocystidia, **5:** cheilocystidia, **6:** setoid hyphae of the base of stipe, **7:** hyphae in the stipe context, **8:** hyphae of the veil (all from the holotype). Scale bar = 17 mm in 1, 10 µm in 2-8.

4-8 µm wide, hyaline. HYMENOPHORAL TRAMA regular, hyaline to yellowish in mass, hyphae 5-20 µm wide. PILEIPELLIS a cutis, with postrate, hyaline hyphae, 2-3 µm wide, not gelatinous. SUBPELLIS with hyaline to yellowish hyphae, 2-8 µm wide. CONTEXT OF STIPE with hyaline to pale yellowish hyphae, 3-14 µm wide. SETACEOUS HYPHAE in the base of stipe 4-5.5 µm wide, thick-walled, yellowish-brown. CLAMP CONNECTIONS present. VEIL formed by radially disposed, hyaline to grayish, thin-walled, branched hyphae, 1.5-4 µm wide.

HABITAT AND DISTRIBUTION—Gregarious on soil, in a subtropical *Quercus-Pinus* forest, close to tropical vegetation, at 2200 m alt. Known only from the type locality.

SPECIMEN STUDIED. **MEXICO, Jalisco**, Sierra Cacoma, Municipio Autlán, Neverías, Sept. 27, 2005, *Sánchez-Jácome 1163* (holotype IBUG, isotype XAL).

DISCUSSION—The combination of the bluing basidioma, well developed veil and annulus, and subrhomboid, thick-walled basidiospores places *Psilocybe meridionalis* in section *Stuntzii* Guzmán, following the classification of Guzmán (1983, 1995). The smaller spores and the presence of pleurocystidia distinguish this species from other species in the section. It is probable that *P. meridionalis* has a pseudorhiza because the basidiomata studied had incomplete bases and the remaining ends may have buried in the clay soil. It is interesting to observe the highly caerulescent discoloring of the basidiomata on all parts. Also, it is important to note the setaceous hyphae in the base of the stipe. Although rare, these hyphae, have not been reported in any species of section *Stuntzii* until now, but such hyphae are present in several species of *Psilocybe* section *Cordisporae* (e.g., *P. guilartensis* Guzmán et al., *P. mesophylla* Guzmán et al., *P. oaxacana* Guzmán et al., *P. fagicola* R. Heim & Cailleux emend. Guzmán, and others, cf. Guzmán et al. 2003, 2004, 2005). This is the first record in Mexico of a species belonging to section *Stuntzii*. This species brings to 12 the number of *Psilocybe* species documented from Jalisco, nine of which have the bluing reaction (Guzmán 1998).

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# Taxonomic status and new localities for *Ganoderma ravenelii*

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**Abstract**—The taxonomic status of *Ganoderma ravenelii* is discussed; furthermore, the species is recorded from two new localities in the USA and probably from Mexico. A complete redescription and illustrations of the species are provided.

**Key words**— *Ganoderma curtisii*, *G. meredithiae*, North Carolina, Texas

## Introduction

Based on the absence of resinous bands in the context and basidiospore shape and size, Steyaert (1980) described *Ganoderma ravenelii* from two collections originally from southern USA. The type specimen was previously identified as *G. curtisii* (Berk.) Murrill, a very closely related species. Steyaert himself (1980) suggested that both taxa might eventually represent just varieties of a single species. According to Moncalvo & Ryvarden (1997), the taxonomic status of these two species remain unclear.

*G. ravenelii* has not been reported since its original description, probably because it was erroneously determined as *G. curtisii* or as another species. Gilbertson & Ryvarden (1986) and Adaskaveg & Gilbertson (1988) mentioned neither *G. ravenelii*, nor *G. curtisii* in their monographs on polypores from North America.

In a reevaluation of the *Ganodermataceae* from Mexico during which the type of *G. ravenelii* was studied, Torres-Torres & Guzmán-Dávalos (2005 and unpublished data) suggested the presence of *G. ravenelii* in the area but could not establish the species with confidence because the available specimen was too immature. During a visit to the Chicago Field Museum Herbarium (F),

collections labeled as *G. lucidum* or *Ganoderma* sp. but agreeing with Steyaert's description were found and studied in detail. In addition, specimens labeled as *G. curtisii* at F and IBUG were studied in order to compare the two species. Unfortunately, the type of *G. curtisii* could not be located at K or BR and so was not studied.

Critical analysis of the features that Steyaert (1980) used to delimit the two species support his conclusion of the presence of two different taxa. Here we provide a full description and illustrations of *G. ravenelii* and include additional features not considered by Steyaert. The species is recorded for the second time in the world from two new localities in the USA and probably from Mexico.

## Materials and methods

All materials (except the *G. ravenelii* holotype obtained on loan from K) were studied at F and IBUG. Herbaria abbreviations follow Holmgren et al. (1990). The key colors are from Kornerup & Wanscher (1963). Micromorphologic observations were made from material mounted in 5% KOH. The basidiospore shape was determined from a Q coefficient (length-width; Bas 1969) from at least 20 randomly selected basidiospores from each collection. The drawings of the microscopic structures were made using a 100x oil-immersion objective on a Zeiss K7 or Zeiss Axioscop 40 microscope.

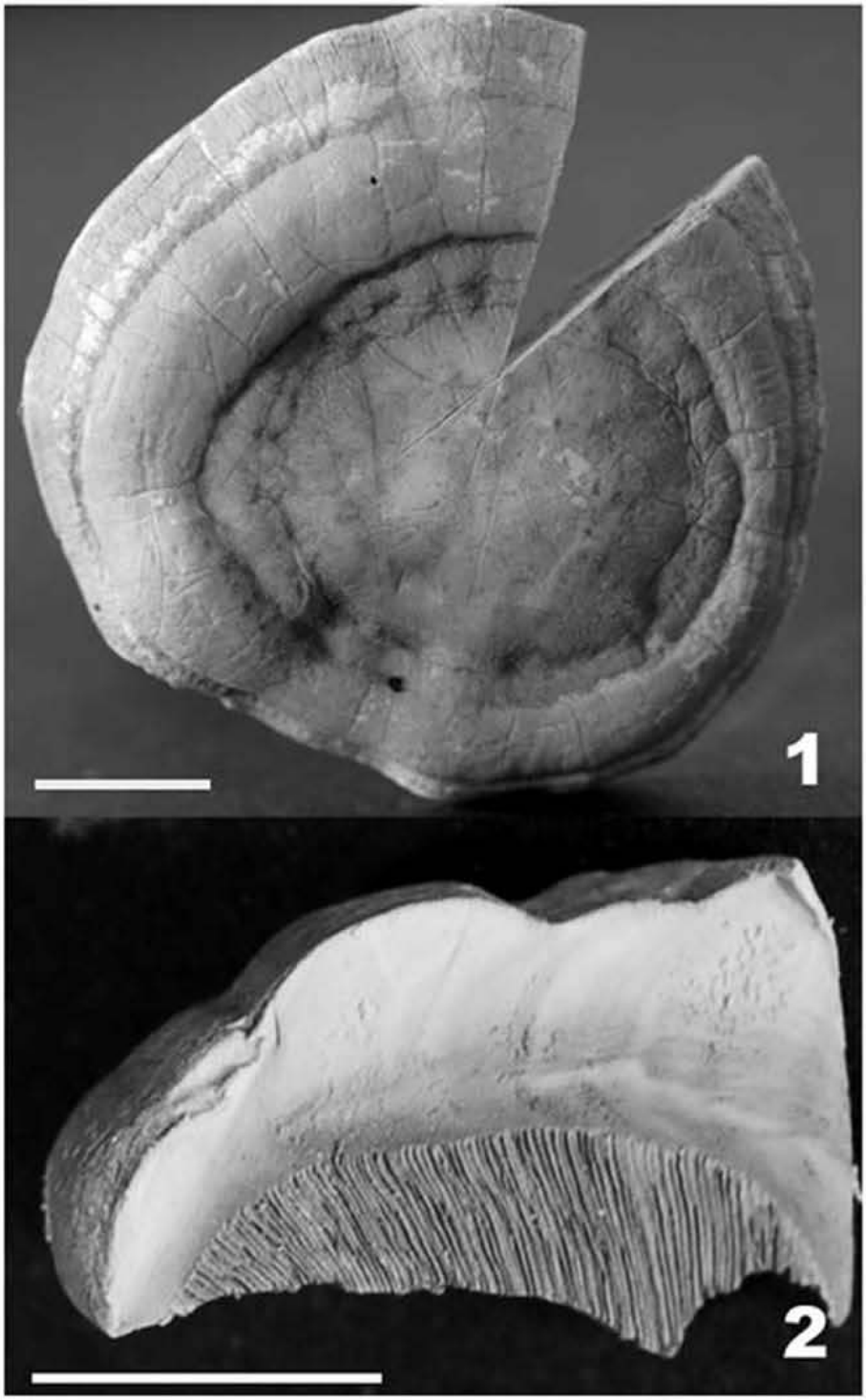
## Taxonomy

***Ganoderma ravenelii*** Steyaert,

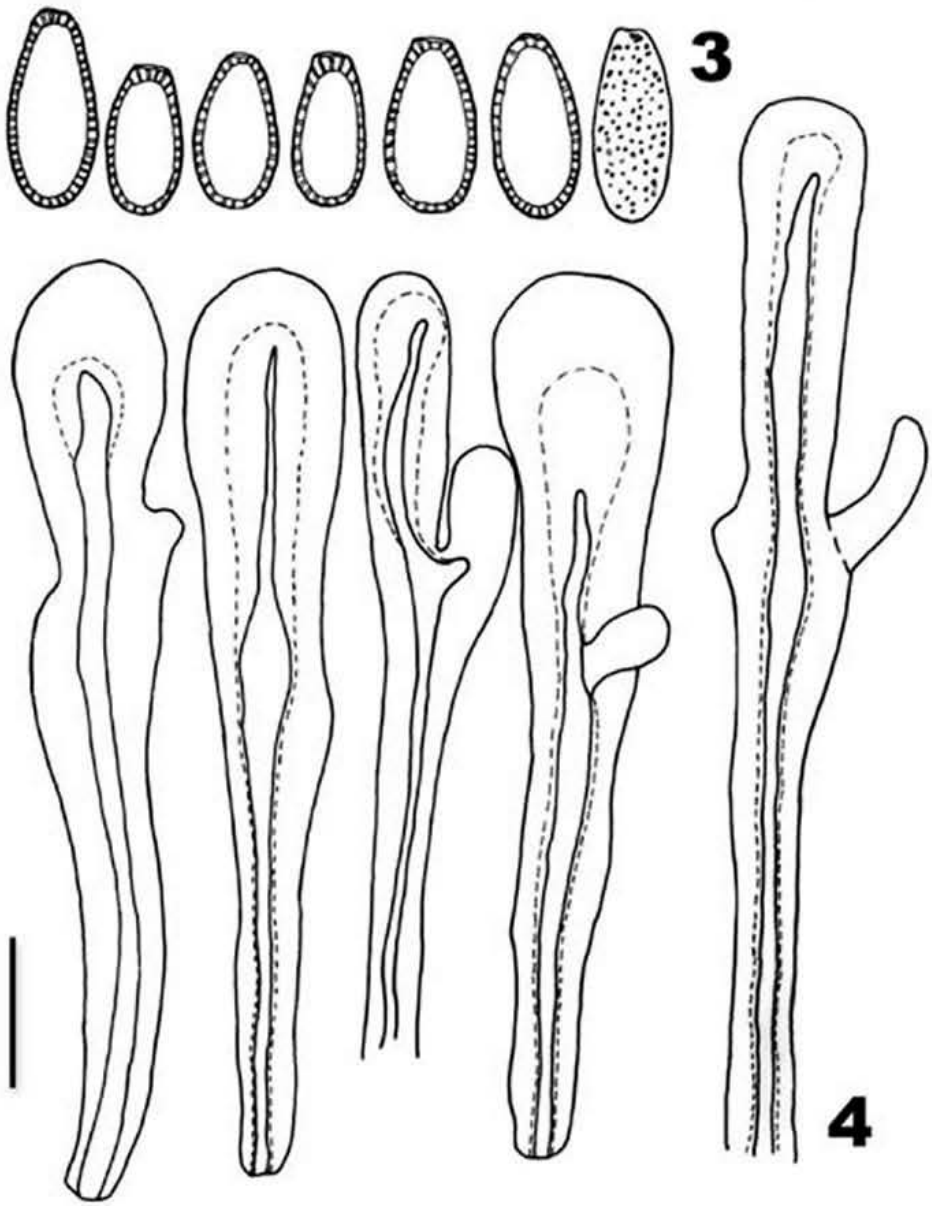
Bulletin du Jardin Botanique National de Belgique 50(1-2): 146, 1980. **Figs. 1-6**

BASIDIOMATA 4.8–6.7 × 3.9–4.5 × 1–1.5 cm, annual, stipitate, single, sometimes imbricate, corky to woody. PILEUS pleuro to mesopodal, rounded flabelliform, reniform to circular, upper surface slightly convex to plane; surface glabrous, smooth to slightly tuberculate, soft, semi-dull, occasionally shiny all over the surface, concentrically sulcate; with a laccate crust, easy to penetrate with fingernail, generally easily removed; maize (4A6), deep yellow (4A8), golden-yellow (5B8) to yellowish-brown (5C8), more or less homogeneous, or with zonations of these tones, occasionally covered by cinnamon (6D6) basidiospores; margin concolorous, entire, thick, rounded to truncate, smooth. STIPE 1-10 × 1.1-1.6 cm, eccentric to central, cylindrical to flattened, solid; surface smooth to tuberculate, dull to shiny, with a laccate crust, easy to remove or not, red-wine almost black, generally darker than pileus.

CONTEXT 0.7–1.3 cm thick, corky to slightly fibrous, duplex, azonate, pale orange to light orange (5A3, 5A4), brown-sienna (6D7) close to the tubes, without resinous bands. PORES 2–3 per mm, angular to rounded; pore surface

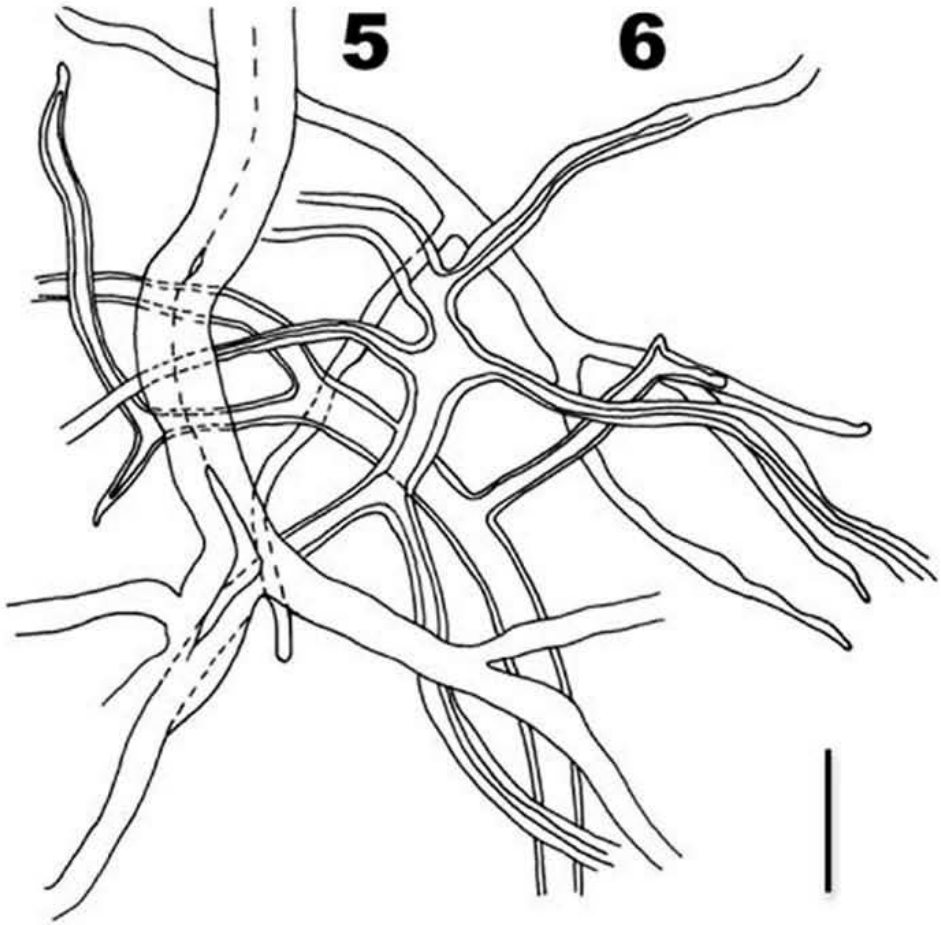



Figs. 1-2. Macroscopic features of *Ganoderma ravenelii* (D.P. Lewis 3428).
1: pileus, 2: context without resinous bands and tubes. Scale bar = 1 cm.




Figs. 3-4. Microscopic features of the holotype of *Ganoderma ravenelii*.
3: basidiospores, 4: cuticle cells. Scale bar = 8 µm.

yellow (2A3) to pale yellow (3A3), darkening to brown (6D8) when bruising or aging; tubes 0.5–1 cm long, unstratified, pale vinaceous-brown to vinaceous-brown (8E4). HYPHAL SYSTEM di-trimitic. CONTEXTUAL TRAMA with generative hyphae no observed; skeletal hyphae 3.2–8.8 µm diam., thick-walled to solid, non-septate, arboriform, yellowish to yellowish-brown, predominant; binding




Figs. 5-6. Microscopic features of the holotype of *Ganoderma ravenelii*.
5: skeletal hyphae, 6: binding hyphae. Scale bar = 8 µm.

hyphae 5.6–8.8 µm diam., thick-walled to solid, non-septate, yellowish. HYMENOPHORAL TRAMA as contextual trama. PILEIPELLIS with cuticle cells 56–80 × 7.2–16 µm, broadly clavate, generally entire or with one lateral protuberance, thick-walled to solid, generally multistratified, yellowish, amyloid in Melzer's reagent; generative hyphae up to 4 µm diam., thin-walled, with conspicuous clamps, hyaline, difficult to observe; skeletal hyphae 4–6.4 µm diam., thick-walled to solid, non-septate, arboriform, yellowish to almost hyaline, scarce; binding hyphae 2–8 µm diam., thick-walled to solid, non-septate, yellowish to yellowish-brown. BASIDIOSPORES (10.2–) 10.8–15.2 × 5.2–7.2 µm, 12.4 × 6.1 µm on average, Q = 1.78–2.72, oblong to cylindrical, apex truncate, with apical germ pore, yellowish-brown; perisporium wrinkled, hyaline to yellowish-red; exosporium with inter-walled pillars up to 0.4 µm thick, free. BASIDIA not observed. CYSTIDIA absent.

Specimens examined. **USA,** South Carolina, Aiken, on the ground, s. data, H.W. Ravenel 2936 (K, HOLOTYPE). Florida, Columbia, Camp O'Lena State Park, s. data, H.S. Dybas s.n. (F). North Carolina, Columbus Co., Reaves Ferry, on tree roots, 18 Oct 1934, W.C. Coker s.n. (F). Texas, Hardin, Larsen Preserve, 21 Dec 1982, D. P. Lewis 3428 (F). **MEXICO,** Jalisco, Municipality of San Cristóbal de la Barranca, km 17 road Guadalajara-San Cristóbal de la Barranca, Milpillas brook, oak forest with tropical influence, altitude 1460 m, 28 Jul 1987, C. Téllez 1025 (IBUG).

Specimens of *Ganoderma curtisii* examined. **USA,** North Carolina, Ardmore Woods, on old oak stump, 10 Aug 1932, P.O. Schallert s.n.; Columbus, Reaves Ferry, on tree roots, 18 Oct 1934, W.C. Coker s.n.; Forsyth, Winston-Salem, in yard, 30 Sep 1934, P.O. Schallert s.n.; Jackson, Whiteside Mt., 12 Jun 1934, P.O. Schallert s.n.; Durham, Durham Duke University Forest, on a stump, Aug 1949, W.L. Culberson s.n. Georgia, Thomas, Thomasville, 19 Jul 1947, H. Field s.n. Louisiana, Calcasieu, Lake Prien, on the ground in pine woods, 30 Oct 1948, F. Drouet s.n. Texas, Hardin, Big Thicket National Preserve, Lance Rosier Unit Cotton Road, on wood, 15 Jun 1983, D.P. Lewis 3515 (all in F). **MEXICO,** Jalisco, Municipality of Zapopan, Bosque La Primavera, 8 km from Prolongación Mariano Otero, oak forest, altitude 1500 m, 20 Jul 2004, M.G. Torres-Torres 526; La Primavera town, Avenida San Antonio, oak forest, altitude 1650 m, 20 Jul 2004, M.G. Torres-Torres 527; Municipality of San Cristóbal de la Barranca, Rancho Pericos, oak forest, altitude 1800 m, 24 Jul 2004, M.G. Torres-Torres 532, Municipality of Colotlán, 14.5 km road Colotlán-Carrizal, oak forest, altitude 1600 m, 2 Aug 2004, M.G. Torres-Torres 541; Municipality of Unión de Tula, Ejutla, near to San Gaspar, oak forest with *Bursera* sp. and *Acacia* sp., 24 Aug 2004, M.G. Torres-Torres 554 (IBUG).

## Discussion

Macromorphologically, *Ganoderma ravenelii* is closely related to *G. curtisii* (Steyaert 1980, and see Table 1 below). Besides the features emphasized by Steyaert (1980) to separate *G. ravenelii* from *G. curtisii* (e.g., absence of resinous bands in the context, shape and size of basidiospores), other important features can be identified: union of the stipe with the pileus, basidiospore pillars, and cuticle cells (Table 1).

*Ganoderma curtisii* produces basidiomata with lateral or occasionally eccentric stipes, resinous bands in the context, cuticle cells that are occasionally laterally or apically branched, ellipsoid to oblong basidiospores [(9.2–)10.4–12(-13.6) × 5.6–8 µm, 11.2 × 6.6 µm on average] with pillars slightly thicker and subfree (free dots mixed with short anastomosed to shortly elongated structures). For *G. ravenelii,* Steyaert (1980) reported cuticle cells of 20 × 8–10 µm, surely a mistake, because his figure 7 shows other proportion. Both *G. curtisii* and *G. ravenelii* can co-exist in the same geographic area, but the morphological features distinguish them as separate species. Based on these morphological differences, we recognize *G. ravenelii* and *G. curtisii* as two distinct species. On the other hand, *G. meredithiae* Adask. & Gilb., another very closely related species (see Table 1), is differentiated by its obligate growth on pines and morphologically distinct cuticle cells (Adaskaveg & Gilbertson 1988).

TABLE 1. Morphologic features of *Ganoderma curtisii* and *G. ravenelii*.

| SPECIES | *G. CURTISII* | *G. MEREDITHIAE** | *G. RAVENELII* |
|---|---|---|---|
| STIPE | generally lateral | sessile, or stipe central to lateral | eccentric to central |
| CONTEXT | duplex | relatively homogeneous or duplex (?) | duplex |
| RESINOUS BANDS | present | present | absent |
| BASIDIOSPORE SIZE | (9.2–)10.4–12(–13.6) × 5.6–8 µm, average 11.2 × 6.6 µm | 9.5–11.5 × 5.5–7 µm | (10.2–) 10.8–15.2 × 5.2–7.2 µm, 12.4 × 6.1 µm |
| BASIDIOSPORE FORM | Q = 1.5–1.88, ellipsoid to occasionally oblong | ellipsoid | Q = 1.78–2.72, oblong to cylindrical |
| BASIDIOSPORE PILLARS | subfree | probably free | free |
| CUTICLE CELLS | clavate, without or with two to three apical or lateral protuberances, occasionally lateral or apical branched | highly variable from nearly spherical or reniform to clavate, often strongly branched or lobed | clavate, generally entire or with one lateral protuberance |

*Information for *G. meredithiae* from Adaskaveg & Gilbertson (1988)

The majority of the specimens labeled *G. lucidum* (Curtis) P. Karst. in F were re-determined as *G. curtisii* or *G. ravenelii*. Steyaert (1980) recorded *G. ravenelii* from South Carolina and Florida. The species is now additionally recorded from North Carolina and Texas. Our examination of one immature specimen with features very similar to *G. ravenelii* but possessing few basidiospores leads us to believe that the species probably occurs in Mexico. This Mexican material of *G. ravenelii* differs from the many Mexican specimens of *G. curtisii* we examined (Torres-Torres & Guzmán-Dávalos 2005, specimens in this paper, and other unpublished data).

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# Three new species of *Parmotrema* containing salazinic acid from the coast of São Paulo State, southeastern Brazil

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**Abstract** — During a survey of *Parmeliaceae* species in natural ecosystems and urbanized coastal areas of São Paulo State, southeastern Brazil, three new salazinic acid-containing *Parmotrema* species were discovered namely, *P. anchietanum*, *P. asperum* and *P. hypermaculatum*.

**Key words** — *Parmotrema cetratum*, *Parmotrema despectum*, *Parmotrema expansum*, *Parmotrema permaculatum*, *Parmotrema ruptum*

## Introduction

The genus *Parmotrema* A.Massal. is characterized by broad rotund lobe apices, absence of pseudocyphellae, frequent occurrence of marginal cilia, naked lower margins, simple rhizinae, and thick-walled, ellipsoid ascospores (Brodo et al. 2001, Nash & Elix 2004). More than three hundred species are known worldwide (Nash & Elix 2004), 93 of which occur in Brazil (Marcelli 2004).

This study describes three new species of *Parmotrema* containing salazinic acid. These were discovered by the authors during an investigation of the large lobed species of *Parmeliaceae* on the coast of São Paulo State in Brazil (Benatti 2005), for the most part situated between the municipalities of Ubatuba (23°02'S, 45°04'W) and Itanhaém (24°11'S, 46°47'W). This region included urbanized areas, rocky shores, and mangrove and restinga forests as the predominant vegetation types.

All three species are saxicolous, collected from rocky coastal areas growing fully exposed to sunlight. Two of the new species produce soredia, while the third lacks vegetative propagules.

Although we have included sufficient specific descriptive information about the three new taxa, more detailed morphological and chemical comparisons with other similar species can be found in Benatti (2005).

## Materials and methods

Specimens were distinguished by morphological characters using standard stereoscopic and light microscopes. Anatomical sections, including those of apothecia and pycnidia, were made with a razor blade by hand. The chemical constituents were checked by spot tests with potassium hydroxide (K), sodium hypochlorite (C) and para-phenylenediamine (P), and also examined under UV light (360 nm). Chemical constituents were identified by thin-layer chromatography (TLC) using solvent C (Bungartz 2001), high performance liquid chromatography (HPLC) (Elix et al. 2003) and comparison with authentic samples.

Since we have encountered problems dealing with the many morphological terms present in the literature, we specify here that in our concept lacinules represent adventitious, ribbon-like secondary outgrowths from the primary lobe margins. Lobules are similar, but short and rounded.

For each new species diagnosis refers exclusively to holotype characters and the English descriptions and comments to all the material studied.

## The species

***Parmotrema anchietanum*** Marcelli, Benatti & Elix, **sp. nov.** **Fig. 1**
MB 511104

DESCRIPTIO: *Thallus sublaciniatus, 12.0 cm latus, cinereus, albidus. Lobi angustati, 1.0–3.0 (–5.5) mm lati, parce maculati, apicibus subtruncatis, lacinulis marginalibus abundantibus, magnitudine 0.2–3.2 × 0.2–1.0 mm, margine ciliato, cilia simplicia, frequentia et regulariter distributa, 0.2–1.6 mm longa; soralia capitata, submarginalia, ad pustulas ephemeras oriunda, in soredia subgranulata caducia erumpentia; subtus niger, ambitum versus castaneus, nudus vel rhizinosus, rhizinae simplices vel furcatae, frequenter agglutinata; medulla albida; apothecia ignota; conidiomata frequentia, conidia brevia filiformia, 6.5–11.0 × 1.0 µm; atranorinam, chloroatranorinam, acidum salazinicum et acidum consalazinicum continens.*

HOLOTYPE–Brazil, São Paulo State, Municipio Ubatuba, Anchieta Island, Saco Grande, rocky shore, 23°02'S, 45°04'W, 2 m alt., saxicolous, leg. A.A. Spielmann, L.S. Canêz & D.F. Peralta 557b, 23-XI-2003 (SP, isotypes in B and NY).

THALLUS up to 16 cm in diameter, coriaceous, saxicolous, light gray darkening in the herbarium, sublaciniate to weakly laciniate; LOBES usually irregularly

branched, occasionally becoming subdichotomous, 1.0–3.0 (–5.5) mm wide, younger lobes imbricate but soon becoming very crowded, adnate to weakly ascending, loosely attached; UPPER SURFACE continuous then irregularly cracked, smooth to partially rugose, APICES almost flat to subconcave, subtruncate to truncate, MARGINS crenate to irregularly dissected, weakly ascending, subundulate, lacinulate, ciliate; MACULAE weak at the center to distinct in the younger parts, punctiform to linear but never reticulate, laminal, sometimes developing into cracks; LACINULES short, regularly distributed at the lobe margins and apices, abundant, usually simple, more rarely furcate or irregular, flat, 0.2–3.2 × 0.2–1.0 mm, truncate or acute, frequently mixed with small, irregular adventitious lobules, underside concolorous with the lower margin; CILIA black, simple, 0.20–1.60 × ca. 0.05 mm, frequent along the margins. MEDULLA white, pigments absent, but sometimes with reddish parts due the hydrolysis and oxidation of the salazinic acid. SORALIA capitate to irregular, originating from pustules (see below), infrequently coalescing, often exposing the lower cortex when eroded; SOREDIA subgranular to granular, ±coarse. PUSTULES submarginal or subapical on the lobes or subapical at the lacinulae, also frequently appearing on thalline ridges, ephemeral and soon bursting into soredia. ISIDIA absent. LOWER SURFACE black, shiny, smooth to subpapillate; MARGIN brown, shiny, smooth to subpapillate, 1.0–3.0 mm wide, usually naked to partially rhizinate; RHIZINES black even at the margin, simple, furcate or rarely irregular, 0.20–1.40 (–3.60) × 0.05–0.10 mm, frequent to dense in some parts, frequently becoming agglutinated, scattered. APOTHECIA absent. PYCNIDIA submarginal and common, sometimes on the lacinulae, with black ostioles; CONIDIA short filiform, 6.5–11.0 × ca. 1.0 µm.

SPOT TESTS: upper cortex K+ yellow, UV–; medulla K+ yellow→red, C–, KC similar to K but weaker due to the C, P+ yellow, UV–.

TLC/HPLC: cortical atranorin (minor), chloroatranorin (minor); medullary salazinic acid (major), consalazinic acid (minor).

PARATYPES — Brazil, São Paulo State, Municipality of Ubatuba, Anchieta Island, Saco Grande rocky shore, 23°02'S, 45°04'W, 2 m alt., saxicolous, leg. A.A. Spielmann, L.S. Canêz & D.F. Peralta 557a, 23-XI-2003 (S); idem, over granite on the rocky shore in the shade, 23°02'S, 45°04'W, 2 m alt., leg. L.S. Canêz & A.A. Spielmann 607b, 23-XI-2003 (SP); idem, over granite on the rocky shore, 23°02'S, 45°04'W, 2 m alt., leg. L.S. Canêz & A.A. Spielmann 620, 23-XI-2003 (SP).

COMMENTS– This species is characterized by the sublaciniate lobes with lacinulate margins, somewhat resembling large specimens of *Parmelinopsis* or *Bulbothrix*, or even small specimens of *Parmotrema reticulatum* (Taylor) M. Choisy.

*Parmotrema anchietanum* is further characterized by the pale gray upper cortex, the ciliate margins and formation of pustular soralia, especially at the

subapical parts of the lobes and lacinules. Although maculae are apparent in parts of the upper cortex, especially in the young, peripheral lobes, they are much less prominent than those observed in species formerly accommodated in *Canomaculina* or *Rimelia*. Indeed, the maculae are barely visible in the central parts of the thallus because of the very pale gray colour of the cortex.

Unlike other sorediate *Parmotrema* species, which commonly have a cream or white coloured margin below the soralia, this species has an entirely brown lower margin.

The lower surface of *P. anchietanum* is only partially nude at the margins due to an irregular pattern of rhizine distribution, some lobes having completely nude lower margins, while others are rhizinate close to the edges, or even entirely rhizinate. The majority of rhizines are simple, but they are mixed with a few furcate or irregularly branched ones, and are frequently agglutinated at the initial stages. The species formely attributed to *Canomaculina* and *Rimelia* have dimorphic or squarrosely branched rhizines, respectively, but these were not observed in *P. anchietanum*. This species has a very smooth and shiny lower cortex, with a few papillate parts and is sometimes randomly cracked.

The development of small, marginal lacinules is quite common in *P. anchietanum*. The soralia are most common in the subapical region of the lacinules, more rarely occurring submarginally or on thalline ridges. They originate from foam-like, ephemeral pustules, which soon burst and form caducous soredia, frequently exposing the upper part of the lower cortex. Sometimes these soredia have a weakly yellowish tinge, possibly due to hydrolysis and oxidation of salazinic acid, since no pigment was found in the medulla.

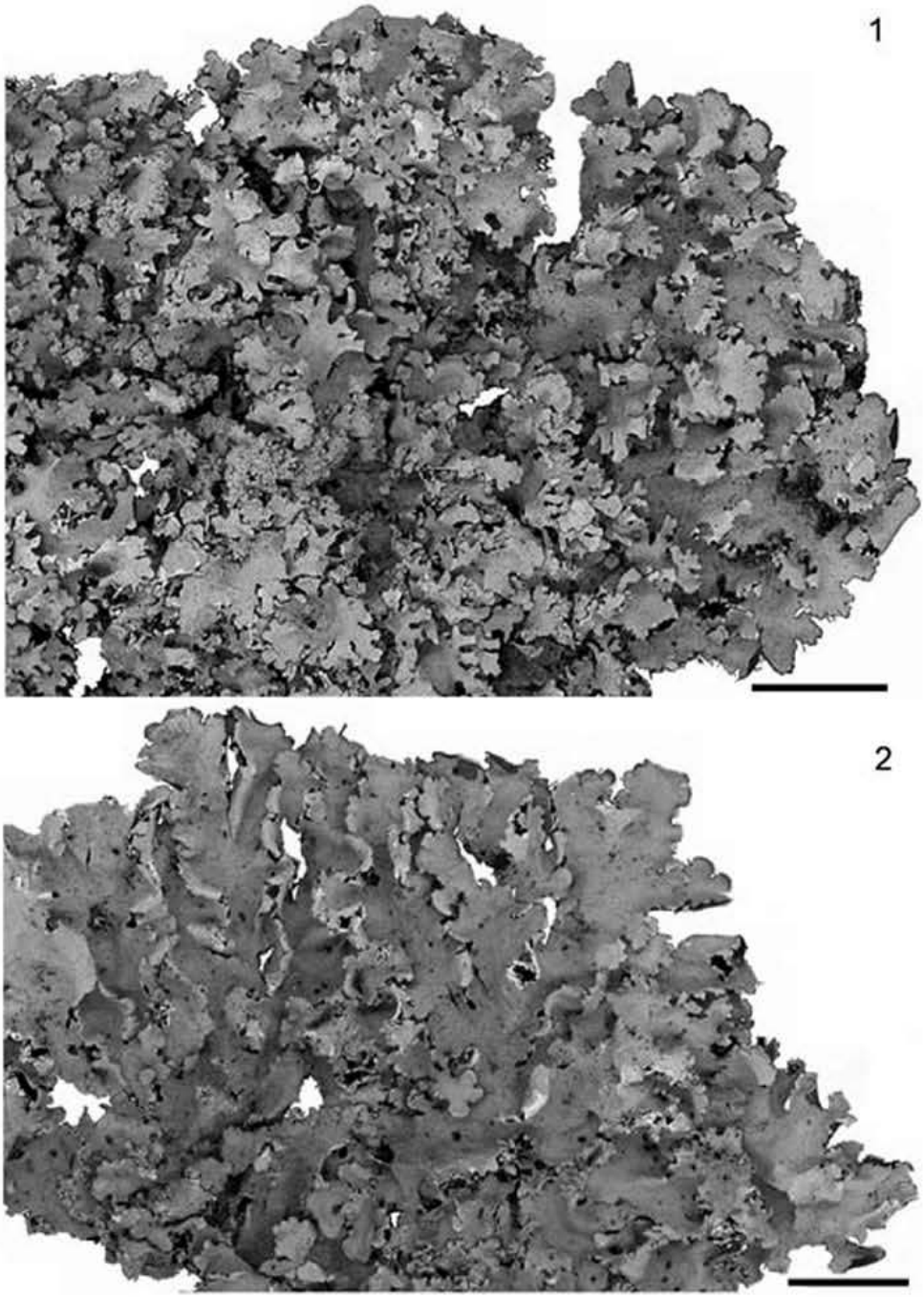
Some specimens exhibited traces of unidentified fatty acids (or other substances) in TLC, substances that were also observed in some specimens of *P. asperum* collected at the same locality (see below).

This species is named after Anchieta Island, the only known collection locality on the northern coast of São Paulo State, Brazil.

***Parmotrema asperum*** Benatti, Marcelli & Elix, **sp. nov.** **Fig. 2**

MB 511105

DESCRIPTIO: *Thallus sublobatus, fragmentatus, 11.0 cm latus, lactineocinereus. Lobi angusti, (1.0–) 2.0–6.5 mm lati, dense maculati, apicibus subrotundatis vel subtruncatis, lacinulis marginalibus satis irregulariter formantes, 0.2–2.2 × 0.2–0.9 mm, margine ciliatis, cilia simplicia et irregulariter distributa, 0.2–0.8 (–1.3) mm longa; soralia capitata, subapicalia, ad pustulas ephemeras oriunda, in soredia granulata et agglutinata incompleta erumpentia; subtus niger, ambito castaneus nudus vel pro parte modice rhizinatus, rhizinae simplices vel furcatae, aliquando agglutinatae; medulla albida; apothecia ignota; conidiomata frequentia, conidia brevia filiformes, 6.5–11.0 × 1.0 µm; atranorinam, chloroatranorinam, acidum salazinicum, acidum consalazinicum et acidum divaricaticum continens.*




**Figures 1-2. 1.** *Parmotrema anchietanum* (part of the holotype).
**2.** *Parmotrema asperum* (part of the holotype).
Bars = 10 mm.

HOLOTYPE–Brazil, São Paulo State, Municipio de Ubatuba, Ilha Anchieta, Saco Grande rocky shore, 23°02'S, 45°04'W, 2 m alt., saxicolous, leg. *A.A. Spielmann*, L.S. Canêz & D.F. Peralta 587a, 23-XI-2003 (SP, isotypes in US and S).

THALLUS fragile, up to 11 cm in diameter, coriaceous, saxicolous, milky gray becoming darker in the herbarium, lobate; LOBES (1.0–) 2.0–6.5 mm wide, irregularly branched, younger lobes overlapping laterally, then becoming crowded, adnate to weakly ascending, loosely attached; UPPER SURFACE continuous, then irregularly cracked, smooth to ±rugose, APICES ±flat to subconcave, subrotund to subtruncate or irregular, MARGIN crenate to irregularly dissected, ±ascending, subundulate, sublacinulate, irregularly ciliate; MACULAE distinct, linear to somewhat reticulate, denser at the center, laminal, sometimes forming cracks; LACINULES short and irregularly spreading at the apices and lobe margins, frequently intermixed with small irregular lobules, simple or irregular, flat, 0.2–2.2 × 0.2–0.9 mm, truncate or acute, underside concolorous with the lower margin; CILIA black, simple, 0.2–0.8 (–1.3) × ca. 0.05 mm, absent on some young lobes, but frequent to irregular on mature lobes. MEDULLA white, pigments absent, but ±reddish spotted due to hydrolysis and oxidation of salazinic acid. SORALIA capitate to irregular, originating from pustules (see below), ±coalescing, frequently agglutinated due to incomplete eruption of the pustules, SOREDIA granular, coarse, agglutinated. PUSTULES subapical on the lobes or lacinules, sometimes also developing on thalline ridges, ephemeral and soon breaking down into soredia, but often incompletely so. ISIDIA absent. LOWER surface black, shiny, smooth to rugose and papillate, MARGIN shiny, brown, smooth to subpapillate, 1.0–3.0 mm wide, naked to partially rhizinate; RHIZINES black even when growing at the margin, simple, furcate or ±irregular, 0.20–1.70 (–2.80) × 0.05–0.10 mm, scattered but frequent to dense in some parts, ±becoming agglutinated. APOTHECIA absent. PYCNIDIA common, submarginal and on the lacinules, with black ostioles; CONIDIA short filiform, 6.5–11.0 × ca. 1.0 µm.

SPOT TESTS: upper cortex K+ yellow, UV–; medulla K+ yellow→red, C–, KC similar to K but paler, P+ yellow, UV–.

TLC/HPLC: cortical atranorin (minor) and chloroatranorin (minor); medullary salazinic acid (major), consalazinic acid (minor), and divaricatic acid (trace).

PARATYPES — Brazil, São Paulo State, Municipality of Ubatuba, Anchieta Island, Saco Grande rocky shore, 23°02'S, 45°04'W, 2 m alt., saxicolous, leg. A.A. Spielmann, L.S. Canêz & D.F. Peralta 561 (B), 609 (SP), 613, 23-XI-2003; idem, 23°02'S, 45°04'W, 2 m alt., over granite in a shaded place on the rocky shore, leg. L.S. Canêz & A.A. Spielmann 607, 23-XI-2003 (SP).

COMMENTS– The habit and maculation of *Parmotrema asperum* resembles that of *P. reticulatum*, and it has narrower and less rounded lobes than species of the *P. cristiferum* (Taylor) Hale group. This species has a characteristic milky

gray colour due to the strongly maculate upper cortex, and the margins are frequently irregularly lacinulate and ciliate.

Both *P. anchietanum* and *P. asperum* differ from the other salazinic acid containing *Parmotrema* species by producing pustular, subapical soralia on the lobes, lacinules, and thalline ridges. Like *P. anchietanum*, the margins of the lower surface of *P. asperum* are brown and in part naked, in part rhizinate with mainly simple rhizines. The surface below the soralia is also brown.

In contrast to *P. asperum*, *P. anchietanum* has a darker gray upper cortex, an alternative disposition and ontogeny of the pustular soralia, and more uniformly ciliate margins. The two species also differ in the nature of the lobes and lacinules. Thus the thallus of *P. asperum* has broader, subrotund lobes with typically short, irregular, unevenly distributed, adventitious marginal lacinules and lobules which are more common on older lobes whereas *P. anchietanum* has narrower, sublaciniate lobes with more regularly distributed marginal lacinules which are distinctly linear. Furthermore, *P. anchietanum* has poorly developed, punctiform maculae which are, for the most part, restricted to the very young lobes, whereas in *P. asperum* and *P. hypermaculatum* (see below), the maculae are often large and dense all over the upper surface where the irregularly dispersed algal cells give rise to small greenish spots scattered over the upper cortex.

The soralia of *P. asperum* are subapical and apical on the lobes and lacinules, but are less common on the thalline ridges. Coarse or granular 78–223 (–305) µm soredia originate from pustules, which only partially burst open, such that the soredia remain agglutinated instead of being shed. The soredia in *P. anchietanum* are finer (52–137 µm) due to the more complete breakdown of the pustules. As a consequence the soredia are often completely shed thus exposing the black upper side of the lower cortex. Also, the soralia of *P. anchietanum* only rarely reach the extremities of the lobes and lacinules, leaving a small but constant esorediate area at the apices.

The specimens of *P. asperum* contained traces of divaricatic acid, which may have originated from sorediate *Dirinaria* species growing nearby. The soredia from such species are often washed on to adjacent lichens. Although some specimens of *P. anchietanum* showed a similar faint spot in TLC, no substances other than the salazinic and consalazinic acids were detected by HPLC.

This species is named after the very granular, agglutinated soredia, which derive from rough soralia following the incomplete breakdown of the pustules, giving the thallus a characteristic, coarse appearance.

***Parmotrema hypermaculatum*** Marcelli, Benatti & Elix, sp. nov. **Fig. 3**

MB 511106

DIAGNOSIS: *Species thallo similis Parmotremati expanso, sed subcoriaceus non fragilis, ciliis plerumque simplicibus, superne cortex dense maculatus, medulla acidum protocetraricum continens, sporis ellipsoideis minoribus (10.0–14.0 × 6.5–9.0 µm) episporio 1.0 µm et conidiis filiformibus majoribus plerumque 9.0–14.0 × 1.0 µm differt.*

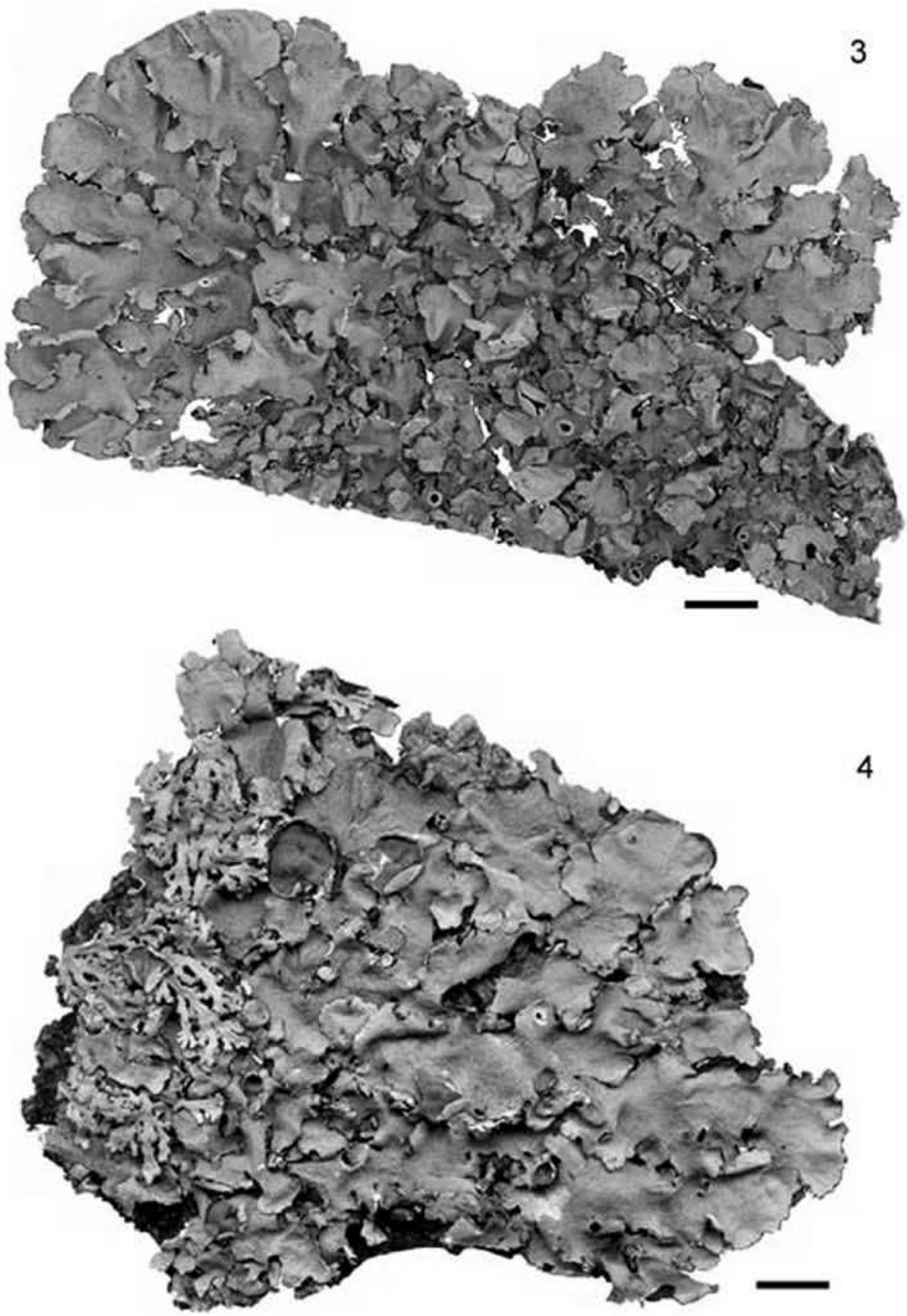
HOLOTYPE–Brazil, São Paulo State, Municipio Itanhaém, Cibratel quarter, rocky shore between Praia dos Sonhos and Praia de Itanhaém, 24°11'S, 46°47'W, 25 m alt., on the leeward side of a small grass-covered granitic rock, leg. M.P. Marcelli 4198, 10-I-1989 (SP, isotypes in CANB and G).

THALLUS up to 19 cm in diameter, subcoriaceous, saxicolous, grayish becoming darker gray on storage, lobate; LOBES (2.0–)4.0–11.0 mm wide, irregularly branched, overlapping laterally and crowded at the thallus center, adnate to weakly ascending, loosely attached; UPPER SURFACE smooth to scrobiculate, continuous, becoming irregularly cracked to almost rimose-reticulate in some parts, APICES slightly revolute, subrounded to irregular, MARGIN smooth to highly incised and crenate, weakly ascending or mostly ±flat, irregularly dissected and sublacinulate in older parts, ciliate; MACULAE laminal and amphithecial, usually distinct, but faint in young apothecia, linear to reticulate, very dense, often developing into cracks; LACINULES short and irregular in size and shape, unevenly dispersed at the apices and along the margins of older lobes, admixed with some small, irregular adventitious lobules, simple or irregularly branched, flat, 0.5–2.3 × 0.4–1.7 mm, apices truncate or rarely acute, underside concolorous with the lower margin; CILIA black, usually simple, rarely furcate or irregularly branched, 0.2–2.6 × 0.05–0.15 mm, sparse to frequent along the margins; MEDULLA white, pigments absent, but sometimes reddish spotted due to hydrolysis and oxidation of salazinic acid. SORALIA, PUSTULES and ISIDIA absent. LOWER SURFACE black, shiny, smooth to subrugose, subvenate or subpapillate; MARGIN shiny, brown, smooth, rugose to papillate, 1.0–5.0 mm wide, naked, cream coloured or variegated under fertile lobes; RHIZINES black, simple, furcate or irregularly branched, 0.2–2.2 (–3.4) × 0.05–0.15 mm, frequent to dense, scattered. APOTHECIA submarginal and common, concave to urceolate, becoming ±flat and sometimes distorted when old, 0.5–7.6 mm wide, substipitate, margins smooth, eciliate; amphithecium and stipe smooth, turning rugose when older; DISC brown, epruinose, imperforate, ±incised when old; ASCOSPORES ellipsoid, 10.0–14.0 × 6.5–9.0 µm, epispore ca. 1.0 µm wide; PYCNIDIA submarginal and common, abundant on lobules or lacinules, with black ostioles; CONIDIA filiform (7.0–) 9.0–14.0 × ca. 1.0 µm.

SPOT TESTS: upper cortex K+ yellow, UV–; medulla K+ yellow→red, C–, KC similar to K but paler, P+ yellow, UV–.

TLC/HPLC: cortical atranorin (minor) and chloroatranorin (minor); medullary salazinic acid (major), consalazinic acid (minor), and protocetraric acid (trace).

PARATYPE — Brazil, São Paulo State, Municipality of Itanhaém, Cibratel quarter, rocky shore between Praia dos Sonhos and Praia de Itanhaém, 24°11'S, 46°47'W, 10 m alt., on steep granitic rock near the sea, leg. M.P. Marcelli, M.M. Marcelli & M.M. Marcelli 6224, 03-IV-1989 (SP).




**Figures 3-4. 3.** *Parmotrema hypermaculatum* (part of the holotype).
**4.** *Parmotrema ruptum* (holotype).
Bars = 10 mm.

Table 1. A comparison of *P. hypermaculatum* with the lectotype of *P. ruptum* and the holotype of *P. expansum*.

| | *P. EXPANSUM* | *P. RUPTUM* | *P. HYPERMACULATUM* |
|---|---|---|---|
| Lobe width (mm) | 6–10 | 4–6 | 4–11 |
| Maculae | effigurate (!, S) | reticulate | markedly reticulate |
| Cilia (mm) | often furcate 0.5–1.2 | simple 0.2–0.5 (!) | usually simple 0.2–2.6 |
| Apothecium | imperforate (S) | imperforate | imperforate |
| Ascospore size (µm) | 17.5–21×10–14 (!) | 10–12.5×6.0–7.5 (S) | 10–14×6.5–9 |
| Conidia size (µm) | 6–9 × 0.5 | 10–12.5 × 1 (S) | 9–14 × 1 |
| Medullary acids | salazinic* | salazinic* | salazinic (major), consalazinic (minor), protocetraric (trace) |

(*) Data from literature. (!) Differs significantly from original description. (S) A.A. Spielmann personal communication.

COMMENTS – *Parmotrema hypermaculatum* is a saxicolous species that lacks vegetative propagules and has a very densely reticulate-maculate upper cortex that sometimes becomes irregularly cracked without becoming reticulate-rimose. The maculae often become so enlarged that they restrict the grayish-green colored areas with algae to small, scattered spots. The margins are moderately ciliate. The apothecia are imperforate, shortly stipitate, and have an eciliate thalline exciple, which is smooth when young but becomes rugose and incised with age.

According to Hale (1977), *Parmotrema expansum* Hale, a very similar species, is distinguished by its subimbricate lobes, frequently furcate cilia and an upper cortex that becomes somewhat rimose-reticulate in the central parts due to the agglomeration of age-dependent cracks, thus more closely resembling those species formerly placed in *Rimelia*. In addition the ascospores of *P. expansum* are considerably larger than those of *P. hypermaculatum* (17.5–21.0 × 10.0–14.0 µm vs. 10.0–14.0 × 6.5–9.0 µm) and the conidia somewhat shorter (6.0–9.0 × 1.0 µm vs. 7.0–14.0 × 1.0 µm).

Lynge´s description of *Parmelia rupta* [*Parmotrema ruptum* (Lynge) Hale] (Lynge 1914) cites only one corticolous specimen (lectotype in S!, Fig. 4), which differs from *P. hypermaculatum* by having much narrower lobes (4–6 mm vs. 4-11 mm wide), shorter (0.5–1.0 mm vs. 0.2–2.6 mm long), simple cilia and

small subglobose spores (9.0–11.0 × 5.5–8.0 µm vs. 10.0-14.0 x 6.5-9.0 µm). Hale (1960) commented that Lynge's specimen was very similar to *Parmotrema cetratum* (Ach.) Hale, apart from the nude lower margins. We confirmed this to be the case with the type resembling a small, weakly reticulate, weakly cracked, elacinulate specimen of *P. cetratum* with nude lower margins. No medullary substance other than salazinic acid is reported for *P. ruptum*.

The original descriptions of *P. ruptum* and *P. expansum* recorded significant differences in lobe width, cilia ramification and ascospore size (Lynge 1914, Hale 1977). Following a comparison of the lectotype of *P. ruptum* with the holotype of *P. expansum*, we consider that these differences are indeed, significant. Thus, the cortex of *P. ruptum* is maculate in a reticulate pattern, whereas *P. expansum* has typical, effigurate maculae (A.A. Spielmann, pers. comm.) which do not resemble *Rimelia*. Consequently, we do not accept these names as synonyms as recommended by Hale & DePriest (1999), but consider them to be similar, possibly related species from this very complicated, salazinic acid containing group (Table 1).

*Parmotrema permaculatum* (Hale) Kurok. is a further species that resembles *P. hypermaculatum*, but it is corticolous, has a more coriaceous thallus, and sessile, perforate apothecia. Moreover, the lobes of *P. permaculatum* are broader (8-15 mm vs. 4-11 mm wide), the ascospores somewhat larger (13.0–16.0 × 8.0–10.0 µm vs. 10–14 × 6.5–9 µm), and lacinules and lobules are absent (Hale 1971, Kurokawa 2001).

According to Kurokawa (2001), *P. despectum* Kurok. has a very coriaceous thallus with a weakly maculate upper cortex, sparse to rare cilia, ±perforate apothecia with a ± rugose amphithecium. Despite some morphological and ascospore similarities with *P. hypermaculatum*, *P. despectum* has a more regular pattern of lobule formation along the margins, with more rounded lobules, while *P. hypermaculatum* has irregular, truncate lobules or lacinules which are scattered and spread irregularly along the margins.

Although Benatti (2005) mentioned caperatic acid detected in TLC, this substance was not confirmed in HPLC analysis.

This species name (*hypermaculatum*) refers to the strong, often dense, reticulate pattern of maculae developed over the entire upper cortex.

Additional comments on other related species containing salazinic acid such as *Parmotrema eurysacum* (Hue) Hale and *P. mantiqueirense* Hale can be found in Benatti (2005).

A more detailed overview of the many problems regarding the salazinic acid-containing group of *Parmotrema* species without vegetative propagules can be found in Spielmann (2005).

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This work could not have been accomplished without the support of the Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPq) — for the masters scholarship to the first author and the research support to the second author.

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MYCOTAXON

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# *Dacampia cladoniicola* sp. nov. (*Ascomycota, Dacampiaceae*) on *Cladonia* sp. from Turkey

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**Abstract** — *Dacampia cladoniicola* is described as new from four collections of squamules of an unidentified *Cladonia* sp. in western Turkey. It differs from the other members of the genus in the shorter and relatively narrow ascospores. The new species is the first *Dacampia* species reported on *Cladonia*.

**Key words** — lichenicolous fungi, lichens, France

## Introduction

In the course of determining lichenicolous fungi from Turkey, one new genus (Hawksworth & Halıcı 2007) and eight new species have been described as new to science (Halıcı et al. 2005, 2007a, b; Halıcı & Hawksworth 2007a, b). Also, many taxa were reported as new records for the country (Halıcı et al. 2006, 2007a, b, c). Here we describe a new species of *Dacampia* growing on *Cladonia*.

The genus *Dacampia* (*Ascomycota, Dothideales, Dacampiaceae*) currently comprises seven species (Halıcı & Hawksworth 2007b). Recently Halıcı & Hawksworth (2007b) provided a key and synopsis to the species, along with drawings of the ascospores. In that study, it was found that *Dacampia* species are generally restricted in their host range, and mostly occur on foliose genera, including *Peltigera*, *Solorina*, and *Leptogium*. However, the type species of the genus; *D. hookeri*, is an independent lichen (Henssen 1995).

## Material and methods

The type material of the new species is deposited in ANES. Specimens were examined with an Olympus BH-2 research microscope fitted with Nomarski

differential interference contrast optics and a drawing tube. Photomicrographs were prepared on a Nikon Eclipse 80i. Sections were prepared by hand and examined in I (Lugol's iodine and Metzler's iodine, with [K/I] and without [I] pre-treatment with 10% KOH), 10% KOH alone, and water. Ascospore measurements were made in water and 10% KOH; the extreme values outside the main range are given in parentheses. The length/breadth (l/b) ratio of the ascospores is given in the same way.

## The species

***Dacampia cladoniicola*** Halici & A.O.Türk, **sp. nov.** FIGURES 1-2

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*Dacampia species insignis ascosporis 3-transseptatiis et 1(-2)-longiseptatis, (9.5–)10.5–12(–12.5) × (4.5–)5.5–6.5 µm (n = 20), l/b = (1.6–)1.9–2.2(–2.6).*

Typus: Turkey, Manisa, Salihli, Western part of Demirköprü Dam, 38°39'N, 28°20'E, alt. 285 m, on squamules of *Cladonia* sp. on mosses, 15 August 2006, leg. M. Candan (ANES 11038 – holotypus).

Etymology: The epithet "*cladoniicola*" refers to the host *Cladonia*.

Description: Lichenicolous, on the squamules of *Cladonia* sp., causing bleaching and apparently suppressing podetia production in infected squamules, pathogenic. Ascomata perithecioid, arising singly, immersed at first with only the ostiole and surrounding zone externally visible, semi-immersed (⅔ to ½) at maturity, 150–200 µm diam, 1–2 per areole, black, subglobose. Ostiole not papilliform, ~ 30 µm diam. Exciple composed of 6–8 layers of angular pseudoparenchymatous cells, textura angularis, 25–50 µm thick, the individual cells somewhat radially compressed, reddish brown to brown, individual cells 8–12 × 6–8 µm in vertical section, smooth, walls ca 1 µm thick. Hamathecium of cellular pseudoparaphyses, abundant, septate, branched and anastomosed, 1.5–2 µm wide; centrum Lugol's and Metzler's solution (after pre-treatment with 10% KOH) I–. Asci elongate-clavate, very shortly stalked, bitunicate in structure, 8-spored in mature asci, (28–)32 × 10–14 µm. Ascospores irregularly biseriately arranged and overlapping in the asci, ellipsoid, pale brown, rounded to somewhat broadly pointed at the apices, muriform, with 3 transsepta and 1(–2) longisepta, smooth-walled, slightly constricted at the septa, cells similarly coloured, lacking a conspicuous gelatinous sheath, (9.5–)10.5–12(–12.5) × (4.5–)5.5–6.5 µm ($n$ = 20), l/b = (1.6–)1.9–2.2(–2.6). Conidiomata not observed.

Ecology and distribution: The species appears to be pathogenic as bleaching is seen in the infected squamules of the host, and podetia production is suppressed. The new species is known from four collections, all from western Turkey. Also, Paul Diederich (pers. comm.) informed us about a further specimen of the new species: France, Alpes-de Haute-Provence, au nord de

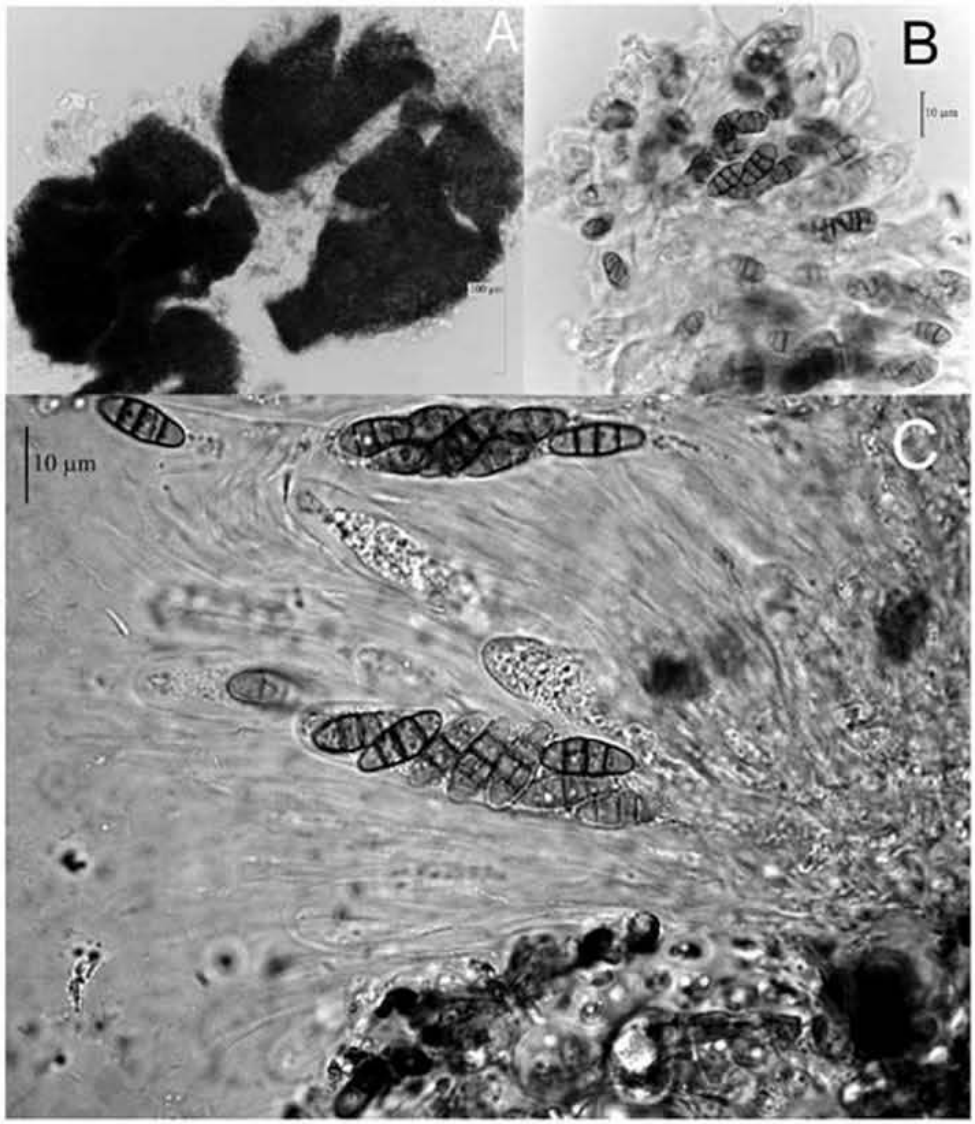


**Fig. 1.** ***Dacampia cladoniicola*** (holotype). A, Surface view of ascomatal wall showing the angular pseudoparenchymatous cells in K. B, Ascospores in K. C, Asci and arrangement of ascospores in asci and pseudoparaphyses in K. Scales A = 100 µm, B = 10 µm, C = 10 µm.

St-Etienne-les-argues, vers Notre Dame de Lure, on terricolous *Cladonia*, 27 May 2006, P. Diederich 12964 (herb. Diederich). Like in the Turkish specimens, the host has only squamules and no podetia. The infected squamules are not bleached (as in the Turkish specimens), but much darker, almost black. As the host genus has a wide distribution in the world, the species should be searched for elsewhere.

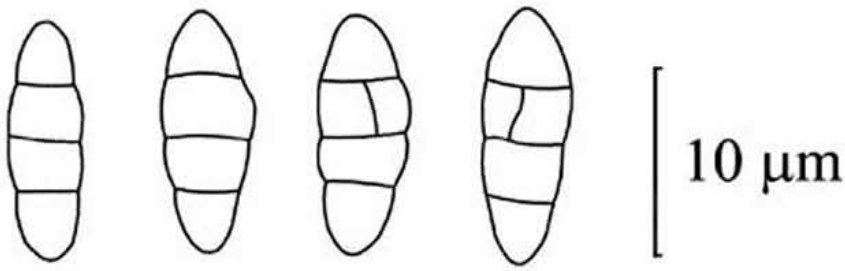


**Fig. 2.** *Dacampia cladoniicola* (holotype). 8 ascospores outline. Scale = 10 µm.

OBSERVATIONS: *Dacampia cladoniicola* is separated from the other members of the genus in having much shorter and relatively narrow ascospores. Also, there is no previous report of any *Dacampia* species on *Cladonia* spp. The narrowest ascospores in the genus are seen in *Dacampia leptogiicola* where they measure 21–25 × 5–6.5 µm, but that species has much longer and also 7-transseptate ascospores and grows on cyanolichens in the genera *Pannaria* and *Leptogium* (Halıcı & Hawksworth 2007b). The other species of the genus with 3-transseptate ascospores, *D. rufescentis* [ascospores (23–)24.5–27 × 11–13 µm] and *D. engeliana* [18–25 × 8–10 µm] have much longer and wider ascospores as well, and grow on different hosts (Halıcı & Hawksworth 2007b).

ADDITIONAL SPECIMENS EXAMINED: Turkey, Manisa, Kula, Southernwest part of Kenger Village, 38°36'N, 28°28'E, alt. 830 m, on squamules of *Cladonia* sp. on mosses, 16 August 2006, leg. M. Candan (ANES 11039); Manisa, Köprübaşı, Western part of Demirköprü Dam, 38°39'N, 28°20'E, alt. 285 m, on squamules of *Cladonia* sp. on mosses, 16 August 2006, leg. M. Candan, (ANES 11042); Eskişehir, Bozdağ, Türkmen Hill, 39°54'N, 30°41'E, alt. 1500 m, on squamules of *Cladonia* sp. on soil, 21 November 2006, leg. M. Candan & T. Tay (ANES 11045).

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# Rare and noteworthy lepiotaceous species (*Basidiomycota, Agaricales, Agaricaceae*) from Israel

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**Abstract**—Four species of lepiotaceous fungi are reported as new to Israel: *Leucoagaricus pilatianus*, *Lepiota apatelia*, *L. subgracilis* and *Macrolepiota konradii*. The second is new for Asia.

**Key words**—*Leucocoprineae*, *Lepioteae*, Near East

## Introduction

Although the family *Agaricaceae* has been the focus of interest for many studies (e.g. Kühner 1936, Babos 1979, Wasser 1980, Bon 1981, Candusso & Lanzoni 1990, Guzmán & Guzmán-Dávalos 1992, Vellinga 2001), it has not been included in inventories of Israeli mycobiota. However, three out of the four agaricaceous tribes as delimited by Singer (1986) and Wasser (1980) are represented in Israel: *Agariceae* Pat., *Leucocoprineae* Singer, and *Lepioteae* Fayod. Wasser (1995, 1996, 1997, 1998, 2000, 2002) has published his reevaluations of earlier data regarding Israeli taxa in the tribe *Agariceae* and reported on 43 taxa representing four genera in *Agariceae* (Wasser 2002). Other tribes, which have not been well studied, require further study. Fragmentary data on some *Leucocoprineae* and *Lepioteae* species is available in more general publications on the higher basidiomycetes of Israel (Avizohar-Hershenzon 1967, Binyamini 1973, 1974, 1975, 1976a,b,c, 1984, 1989; Didukh et al. 2002, 2004; Reichert & Avizohar-Hershenzon 1953, 1955, 1959). Our literature review identified the following 28 lepiotaceous species: *Chlorophyllum molybdites* (G. Mey.) Massee, *Macrolepiota excoriata* (Schaeff.) M.M. Moser, *M. fuligineosquarrosa* Malençon,

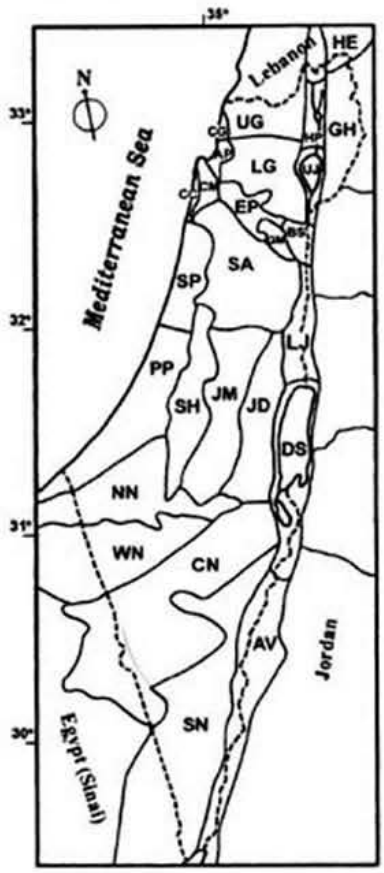


**Figure 1:** Nature regions of Israel: AP – Akko Plain; AV – Arava Valley; BS – Beit Shean Valley; CC – Carmel Coast; CG – Coast Galilee; CM – Carmel Mount; CN – Central Negev; DS – Dead Sea Area; EP – Esdraelon (Yizreel) Plain; GH – Golan Heights; GM – Gilboa Mount; HE – Hermon Mount; HP – Hula Plain; JD – Judean Desert; JM – Judean Mts.; LG – Lower Galilee; LJ – Lower Jordan Valley; NN – Northern Negev; PP – Philistean Plain; SA – Samaria; SH – Shefela; SN – South Negev; SP – Sharon Plain; UG – Upper Galilee; UJ – Upper Jordan Valley; WN – Western Negev.

*M. mastoidea* (Fr.) Singer, *M. procera* (Scop.) Singer, *M. prominens* (Fr.) M.M. Moser, *M. rachodes* (Vittad.) Singer, *Leucoagaricus carneifolius* (Gillet) Wasser, *La. leucothites* (Vittad.) Wasser, *La. littoralis* (Menier) M. Bon & Boiffard, *La. wichanskyi* (Pilát) M. Bon & Boiffard, *Chamaemyces carmelensis* M. Didukh & Wasser, *Ch. fracidus* (Fr.) Donk, *Leucocoprinus birnbaumii* (Corda) Singer, *Lc. cretatus* Locq. ex Lanzoni, *Lepiota alba* (Bres.) Sacc., *L. brunneoincarnata* Chodat & C. Martin, *L. brunneolilacea* M. Bon & Boiffard, *L. castanea* Quél., *L. clypeolaria* (Bull.) P. Kumm., *L. echinella* Quél. & G. E. Bernard, *L. helveola* Bres., *L. lilacina* (Quél.) Boud., *L. micropholis* (Berk. & Broome) Sacc., *L. oreadiformis* Velen., *L. scobinella* (Fr.) Gillet, *L. serena* (Fr.) Sacc., and *L. setulosa* J.E. Lange.

As a result of our investigation, four species of lepiotaceous fungi are reported from Israel for the first time: *Leucoagaricus pilatianus, Lepiota apatelia, L. subgracilis* and *Macrolepiota konradii.*

## Materials and methods

Specimens were collected in Israel during the period 2006-2007 and deposited at the Herbarium of the Institute of Evolution, University of Haifa (HAI).

Distribution of species (Figure 1) is noted according to the natural regions of Israel as defined by Feinbrun-Dothan & Danin (1998). Species concepts and tribe circumscriptions follow Wasser (1985, 1989, 1993). General ecological information is drawn from Wasser (1993), Guzmán & Guzmán-Dávalos (1992), and Vellinga (2001). Micromorphological characteristics of our specimens were observed using Melzer's reagent, Congo red, cotton blue and Cresyl blue.

## Results

Four new records of lepiotaceous fungi are added to the mycobiota of Israel; these are: *Leucoagaricus pilatianus, Lepiota apatelia, L. subgracilis* and *Macrolepiota konradii,* one of these, *Lepiota apatelia* is a new record for Asia. Our investigations also show that species are found primarily in *Quercus, Eucalyptus,* and *Pinus* groves of northern Israel.

## Taxonomy

***Leucoagaricus pilatianus*** (Demoulin) Bon & Boiffard, Doc. Mycol. 6(24): 45. 1976 **Figure 2**

**Bas.:** *Lepiota pilatiana* Demoulin, Lejeunia, n.s. 39: 11. 1966.

**Syn.:** *Lepiota rufovelutina* var. *sanguinescens* Pilát, Sborn. Nar. Muž. v Praže, 11B(2): 16. 1953.

*Lepiota rufovelutina* var. *subrubens* Wichanský, Česká Mykol. 14: 49. 1960.

*Lepiota pilatiana* var. *subrubens* (Wichanský) Demoulin, Lejeunia, n.s. 39: 12. 1966.

*Leucocoprinus pilatianus* (Demoulin) M.M. Moser, Kleine Kryptogamenflora, 3Aufl. IIb/2: 186. 1967 (nom. inval.).

*Leucocoprinus pilatianus* (Demoulin) Wasser, Novit. Syst. Plant. Vasc. et non Vasc. 4: 219. 1978.

*Leucocoprinus pilatianus* var. *subrubens* (Wichanský) Wasser, Nov. Sist. Niz. Rast. 14: 221. 1978 ('1977').

*Leucocoprinus badhamii* var. *pilatianus* (Demoulin) Krieglst., Beitr. Kenntn. Pilze Mitteleur. 7: 57. 1991.

*Leucoagaricus salmoneophyllus* Bon & Guinb., Doc. Mycol. 22(88): 31. 1993.

*Leucoagaricus pilatianus* var. *subrubens* (Wichanský) Migl. & A. Gennari, Riv. Micol. 41(4): 292. 1999 ('1998').

*Leucoagaricus pilatianus* var. *salmoneophyllus* (Bon & Guinb.) Migl. et A. Gennari, Riv. Micol. 41(4): 293. 1999 ('1998').

Misapplied - *Lepiota rufovelutina* Velen. sensu Pilát, Klič k určovaní našich hub: 424. 1951, Sborn. Nar. Muž. v Praže, 11B(2): 12-18. 1953.

Pileus 5–8 cm in diam., convex-applanate, with low umbo, slightly incurved margin, brownish-reddish, toward the center darker, velvety or silky, sometimes almost smooth, with age cracked, flesh between cracks cream-white. Surface on damaging discoloring to reddish, then becoming vinaceous-brownish, brown. Lamellae free, with collarium, thin, crowed, slightly ventricose, whitish or light cream-colored with a concolorous fimbriate edge, on handling staining

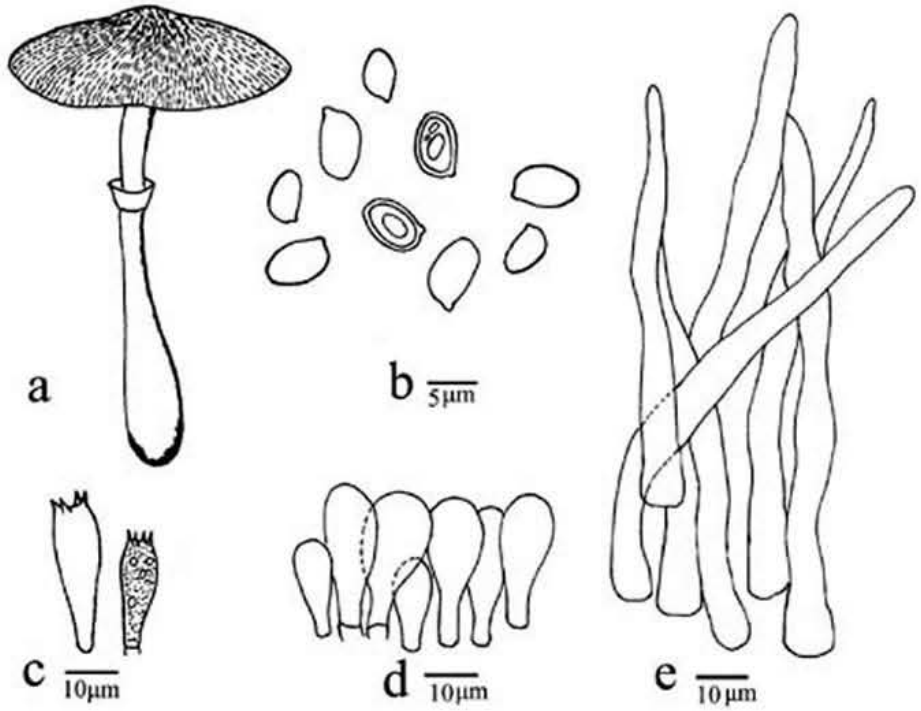


**Figure 2:** *Leucoagaricus pilatianus* (HAI 343).
a – basidiomata, b – basidiospores, c – basidia, d – cheilocystidia, e – elements of the pileipellis

reddish, on drying light pink. STIPE 6–11×0.8–1.0 cm, central, straight, hollow, sometimes twisted, cylindrical, up to 0.5 cm wide at apex and gradually bulbous 1.5 cm wide base, white and smooth above annulus, and brownish-reddish, solid, smooth, silky, shiny, slightly fibrillose under ring. RING situated 6–7 cm above stipe base, sometimes in the middle of the stipe, simple, 0.5–0.7 cm wide, infundibuliform, whitish, with a dark brown margin, stable. CONTEXT fleshy, whitish, on exposure becoming red, without smell, taste none. SPORE PRINT whitish.

BASIDIOSPORES 5.5–9×3.5–4.5 µm, ovate to ellipsoid, sometimes amygdaloid, with a lateral apiculus, with a germ pore, which is often invisible, colorless, smooth, thin walled, weakly dextrinoid, congophilous, cyanophilous, metachromatic. BASIDIA 4-spored, 17–25×6.0–8.0 µm, clavate, hyaline, mostly thin-walled. PLEUROCYSTIDIA absent. CHEILOCYSTIDIA (21)28–43×8.0–12(13–14) µm, variable in shape, mostly clavate, without apical excrescences, smooth, not pigmented. HYMENOPHORAL TRAMA irregular. PILEIPELLIS consisting of densely packed, long erect cystidioid hairs, 80–350×5–15 µm, wide in base, gradually narrow at apex; apex rounded, with pale yellowish, brownish, pinkish

pigment. Almost all hyphae are incrusted with reddish-brownish crystals. STIPITIPELLIS consists of long cylindrical pale colored hyphae 3–10 µm in wide. CLAMP-CONNECTIONS absent.

SPECIMENS EXAMINED: **Israel.** GH: Ein Teo, in *Eucalyptus* groves, leg. Y. Ur, 21.02.07, det. A. Kosakyan (HAI 343) (Figure 6).

HABITAT AND DISTRIBUTION: Solitary to gregarious in small groups, saprotrophic in soil, in deciduous copse on loamy nutrient-rich soil. Known from EUROPE: British Isles, Belgium, Ukraine, France, Italy, Austria, Hungary, Czech Republic, Slovak Republic, Romania, Spain, Russia (North Caucasus); ASIA: Israel (first record), Georgia (Caucasus); AFRICA: Morocco.

DISCUSSION: *Leucoagaricus pilatianus* is a rare species for Asia. First recognized as *Lepiota rufovelutina* Velen. sensu Pilát, the taxon was found to have features in conflict with the concept of *L. rufovelutina* var. *subrubens* as described by Wichanský (1960) by Demoulin (1966), who transferred it to *L. pilatiana* var. *subrubens*. Moser (1967) later transferred *Lepiota pilatianus* to *Leucocoprinus* based on the presence of germ pore in spores and differences in microchemical reactivity and pileipellis. Vellinga (2001) next transferred *Leucocoprinus pilatianus* to *Leucoagaricus* based on differences in habitus of pileus, heteromorphic basidia, presence of pseudoparaphyses; this transfer was also supported by molecular data. *Leucoagaricus salmoneophyllus* was recently described (Bon & Guinberteau 1993) as an independent species based on its small habitus, smell, and different tint; Gennari & Migliozzi (1999), who regarded these differences as varietal, proposed *Leucoagaricus pilatianus* var. *salmoneophyllus* (Gennari & Migliozzi 1999) for the taxon, now regarded as synonymous with *Leucoagaricus pilatianus*.

Relatively larger spores and basidia distinguish our specimens from those of Demoulin (1966). The specimens described by Candusso & Lanzoni (1990) have smaller pilei, shorter stipes, more variable and larger cystidia. In Wasser (1993), specimens are described as thin, without umbos, and with more variable cystidia and pleasant smell and taste. Basidiospores from specimens collected in The Netherlands are more ellipsoid than amygdaloid (Vellinga 2001) while larger fleshy pilei, more amygdaloid basidiospores, and longer pileal elements distinguish specimens described from Canaria (Bañares & Beltran 1981).

*La. pilatianus* is most closely related to *La. badhamii*, from which it differs by the absence of pigmentation in cheilocystidia, cheilocystidial shape, and stipitipellis hyphal structure. The problem of species independence of *La. pilatianus* and *La. badhamii* and differences in the ecology and distribution is discussed in detail by Demoulin (1966), Josserand (1974), Candusso & Lanzoni (1990), Wasser (1993). *Leucoagaricus pilatianus* is also close to *La. aurantiovergens* A. Gennari & Migl. (Gennari & Migliozzi 1999), described

TABLE 1. Important distinguishing characteristics of *Leucoagaricus pilatianus* and related *Leucoagaricus* species

| SPECIES | *LA. PILATIANUS* | *LA. BADHAMII* | *LA. JUBILAEI* | *LA. AURANTIO-VERGENS* | *LA. MELAGRIS* | *LA. GEORGINAE* |
|---|---|---|---|---|---|---|
| REFERENCE | Personal observation | Candusso &Lanzoni 1990 | Reid 1990 | Gennari & Migliozzi 1999 | Vellinga 2001 | Wasser 1993 |
| PILEUS (*P*) & STIPE (*S*) [in cm] | *P* 5.0–8.0 *S* 6.0–11.0 × 0.8–1.5 | *P* 5.0–8.0 *S* 8.0–12.0 × 0.5–1.0 | *P* 2.5–4.5 *S* 3.0–6.0 × 0.4–1.0 | *P* 3.0–6.0 *S* 5–80 × 0.5–1.5 | *P* 1.3–8.0 *S* 2.0–6.5 × 0.3–1.2 | *P* 1.0–2.5 *S* 1.5–4.5 × 0.2–0.3 |
| CONTEXT [color after exposure] | Red | Fleetingly red | First yellow, then red | Orange (stipe only) | Yellow, then red, then vanishing | Red |
| SPORES [in µm] | 5.5–9 × 3.5–4.5 | 6.0–8.5 × 3.5–4.7 | 6.0–8.0 × 3.5–4.5 | 7.0–10.0 × 3.4–5.0 | 7.5–11.5 × 5.5–8.0 | 6.0–8.0 × 3.0–4.5 |
| CYSTIDIA [in µm] | (21)28–43 × 8–12(14) hyaline | 30–60×20 apical rame, brown granules | 20–46(6) × 8–15 | 35–60 × 8–16 clavate-cylindrical | 20–65 × 8–17 | 30–60 × 10–16 lanceolate with apical elongation |
| PILEIPELLIS ELEMENTS [in µm] | Long erect cystidioid hairs (80–350 × 5–15) | Long (100–400 × 10–20) (short cells intermixed) | Clavate-cylindric elements 40–200 × 6–20) | Trichoderm with clavate-cylindric elements | Long (50–170 × 10–20) (short cells intermixed) | Trichoderm with clavate-cylindric elements |

from Italy, from which it differs by basidiospore size, cystidia, and discoloration of stem to orange when scratched. *La. pilatianus* differs from *La. jubilaei* (Joss.) Bon in size, cystidial shape, and context discoloration when cut (Wasser 1993). Table 1 compares *Leucoagaricus pilatianus* and related species.

***Lepiota apatelia*** Vellinga & Huijser,
Belg. J. Bot. 131: 196. 1999 ('1998') **Figure 3**

Misapplied- *Lepiota cristatoides* sensu Bizio et al., Riv. Micol. 36: 226-227. 1993; sensu Huijser & Vellinga, Arnolds et al., Overz. Paddest. Nederland: 290-291. 1995; sensu Kelderman, Parasolzw. Zuid-Linburg: 132-133. 1994; sensu Vellinga & Huijser, Coolia 40: pl. 3. 1997.

PILEUS 1.5–2 cm in diam., plano-convex to applanate, with low umbo, slightly incurved edge, orange brown to yellow-brownish, with little darker center, around center breaking up into concentrically arranged adnate patches, with faintly velar remnants at the margin. LAMELLAE free, thin, crowded, cream-yellowish, with slightly pink tinge, with eroded whitish edge. STIPE 3–3.5×0.2–0.3 cm, with 0.4–0.5 cm wide base, cylindrical, smooth, hollow, cream, from up to base discoloring slightly brown-reddish, which is vanishing later on. White

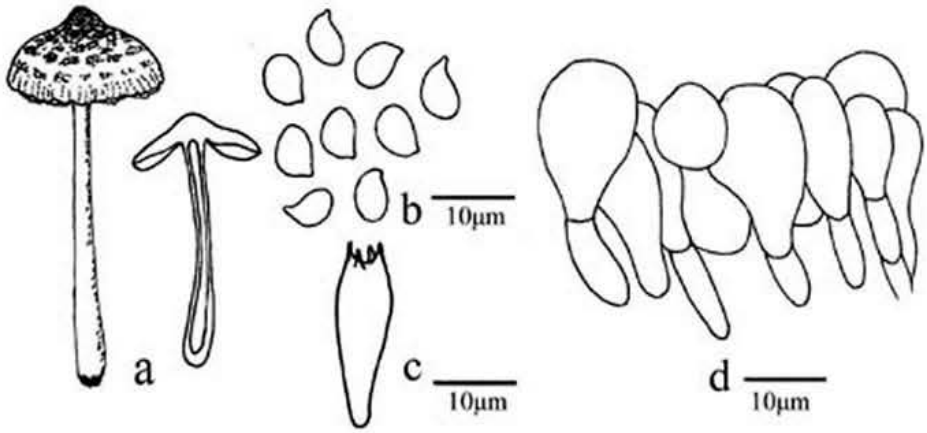


**Figure 3:** *Lepiota apatelia* (HAI 33).
a – basidiomata, b – basidiospores, c – basidium, d – elements of the pileipellis.

mycelial cord on the base. RING white, very membranous, not distinct and removed very easily. CONTEXT whitish-cream, in base brownish, with strong unpleasant smell, taste is unknown. SPORE PRINT whitish.

BASIDIOSPORES 5–7×3.5–4.5 μm, ellipsoid to oblong, with small lateral apiculus, pale brown in Melzer's reagent, weakly metachromatic. BASIDIA 4-spored, 22–30×7–9 μm, clavate, thin-walled. CHEILOCYSTIDIA and PLEUROCYSTIDIA absent. HYMENOPHORAL TRAMA regular. PILEIPELLIS consisting of hymeniderm, made up of clavate terminal thick-walled elements, 12–35×8–10 μm, colored to weakly brown intercellular pigment. STIPITIPELLIS consists of cylindrical long colorless hyphae with 3–4 μm wide. CLAMP-CONNECTIONS present.

SPECIMENS EXAMINED: **Israel.** UG: Safsufa forest, under *Quercus calliprinos*, leg. Y. Ur, 19.11.06, det. A. Kosakyan (HAI 33) (Figure 6).

HABITAT AND DISTRIBUTION: Gregarious, often in big groups, saprotrophic in soil, in deciduous woods, also mine waste heaps. Known from EUROPE: the Netherlands, Austria, Germany, Italy; ASIA: Israel (first record).

DISCUSSION: *Lepiota apatelia* is a very rare species that Vellinga & Huijser (1999) described recently from the Netherlands. *Lepiota apatelia* has often been misidentified (e.g., Bizio et al. 1993, Kelderman 1994, Vellinga & Huijser 1997), and in literature sometimes we can find it as *Lepiota cristatoides*. *Lepiota apatelia* is macroscopically very similar to *L. cristata*, from which it is easily distinguished by shape of spores, absence of distinct annulus, and a different smell. It is also very close to *L. cristatoides*, from which it differs by size, non-smooth, orange-brown pileus, absence of annulus, and basidiospore

TABLE 2. Important distinguishing characteristics of *Lepiota apatelia* and related *Lepiota* species

| SPECIES | *L. APATELIA* | *L. CRISTATA* | *L. CRISTATOIDES* | *L. HYMENODERMA* | *L. THIERSII* |
|---|---|---|---|---|---|
| REFERENCE | Personal observation | Wasser 1980 | Vellinga 2001 | Candusso & Lanzoni 1990 | Sundberg 1989 |
| PILEUS (*P*) & STIPE (*S*) [in cm] | *P* 1.5–2 *S* 3–3.5 × 0.2–0.5 ring not distinct | *P* 1.5–5 *S* 2.5–7 × 0.2–0.5 ring distinct | *P* 0.9–3.0 smoother, pinker *S* 1.5–5 × 1–3 ring distinct | *P* 1.6–2.5 *S* 3–6.5 × 0.18–0.27 without ring | *P* 0.8–4 *S* 3.0–6.0 × 0.25–0.5 fibrillose zone, no ring |
| CONTEXT (*P* = PILEUS; *S* = STIPE) | *P* whitish *S* brown-reddish Odor of strong rubber; | *P* whitish *S* pinkish Odor of strong rubber; taste unpleasant | *P* whitish *S* pinkish Odor of strong rubber | *P* white to cream *S* orangish Odor both fruity & rubberlike | *P*&*S* white & unchanging Odor & taste farinaceous |
| SPORES [in µm] | 5–10 × 2.5–3.5 truncate, narrowly triangular, metachromatic | 6–8.5 × 2.5–3 ellipsoid, pale | 3.5–5.5 × 2.5–3.5 ellipsoid to oblong, metachromatic | 4.5–6. × 2.5–3 ellipsoid to oblong, metachromatic | 4.7–6.3 × 3–3.9 ovoid-ellipsoid, not dextrinoid |
| PLEURO-CYSTDIA | absent | absent | absent | absent | absent |
| CHEILO-CYSTDIA | absent | presen | absent | present | absent |

microchemical reactions. The also close *Lepiota thiersii* Sundb., described from California (Sundberg 1989), produces nondextrinoid basidiospores, a less well-developed velium partiale, clavate to spheropedunculate pileipellis elements, and farinaceous smell. *Lepiota apatelia* and related species are compared in Table 2.

***Lepiota subgracilis*** Kühner ex Wasser,
Ukr. Bot. J. 35(5): 517. 1978 **Figure 4**

Syn.: *Lepiota gracilis* (Quél.) J.E. Lange, Dansk bot. Ark. 2(3): 24. 1915, nom. illegit., *Lepiota gracilis* Peck 1899.
*Lepiota subgracilis* Kühner, Bull. Soc. Mycol. France 52: 231. 1936 (nom. inval.).
*Lepiota kuehneriana* Locq., Friesia 5: 296. 1956.
*Lepiota latispora* (Kühner ex Wasser) Bon, Doc. Mycol. 11(43): 30. 1981.
Misapplied- *Lepiota gracilis* sensu J.E. Lange, Flora Agaricina Danica I: 28. 1935, non sensu Locq. Bulletin Soc. Linn. Lyon 14: 50. 1945.

PILEUS 1.8 cm in diam., thin, fleshy, first campanulate, after expanding to plano-convex, with low umbo, at center dark red-brown, with small dense fibrills,

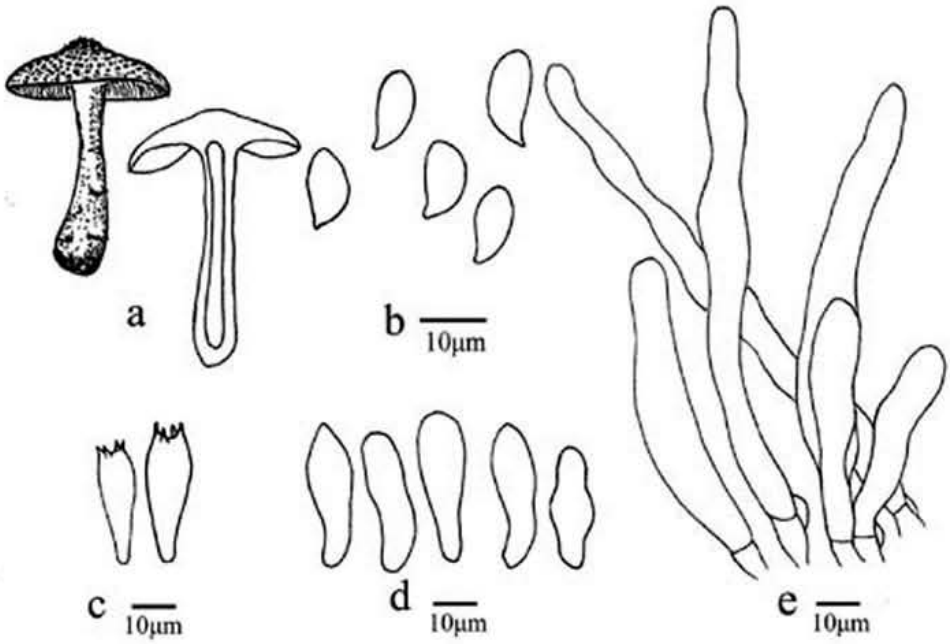


**Figure 4:** *Lepiota subgracilis* (HAI 328).
a – basidiomata, b – basidiospores, c – basidia, d – cheilocystidia, e – elements of the pileipellis

paler near margin, with brownish granular squamules, on whitish-ochraceous background. LAMELLAE free, crowded, wide, creamy white, cream-colored, very pale grey brown with age, with finely serrate to almost even, white age. STIPE 3.0×0.4 mm, cylindrical, slightly enlarging at the base, hollow, cream to pale ochraceous, with lilaceous tinge, fibrillose or squamulose at base. RING usually present as white wooly remnants of the partial veil, but not distinct, and disappearing soon after collection. CONTEXT whitish in pileus, concolorous with surface and shiny in stipe. Without smell and taste. SPORE PRINT whitish.

BASIDIOSPORES 10.5–11.5×4.5–5 µm, amygdaliform to oblong, with lateral apiculus, dextrinoid, congophilous, metachromatic. BASIDIA 4-spored, 20–30×7–12 µm, clavate. PLEUROCYSTIDIA absent. CHEILOCYSTIDIA 20–45×7–12 µm, variable in shape, cylindrical, clavate, lageniform, utriform, colorless. HYMENOPHORAL TRAMA regular. PILEIPELLIS with elongated elements 50–300×6–12 µm, some hyphae with one septa and brownish walls. STIPITIPELLIS made up of long cylindrical hyphae 7–10 µm wide. CLAMP CONNECTIONS present.

SPECIMENS EXAMINED: **Israel.** CM: Ramat Hanadiv, in *Pinus* groves, leg. Y. Ur, 19.02.07, det. A. Kosakyan (HAI 328) (Figure 6).

HABITAT AND DISTRIBUTION: Solitary or in small groups, saprotrophic in soil, on clayey soils, often rich in nutrients, in deciduous woods. Known from

TABLE 3. *Lepiota subgracilis* characters as published by different authors

| REFERENCE | CURRENT PAPER (2008) | WASSER (1980) | CANDUSSO & LANZONI (1990) | VELLINGA (2001) |
|---|---|---|---|---|
| PILEUS (*P*) & STIPE (*S*) [in cm] | *P* 1.8 *S* 3 × 0.4 | *P* 2–4 *S* 2–5 × 0.2–0.5 | *P* 1.5–3.5 *S* 2–4.5 × 0.3–0.6 | *P* 2–4.8 *S* 4–9 × 0.2–0.65 |
| PILEIPELLIS ELEMENTS [in µm] | Elongated (50–300×6–12), some hyphae with one septum; basal clavate elements with brownish walls | — | Elongated (100–400×8–13); clavate on base (30–50×8–14) with brownish walls | Elongated (55–360×8–13) with 0–1 septa & grey-brown walls; with/without basal clavate elements |
| STIPITIPELLIS HYPHAE | Long, cylindrical, 7–10 µm diam | — | — | Long, cylindrical walls pale yellow 4–6 µm diam |
| SPORES [in µm] | 10.5–11.5×4.5–5 amygdaliform to oblong, apiculus lateral | 9–12 × 4.5–6 fusiform-ellipsoid, apiculus lateral | 10–14 × 4.5–5 subfusiform to slightly ellipsoid apiculus lateral | 9–13.5 × 4–6.5 amygdaliform to oblong, broadly fusiform or ovoid |
| CHEILOCYSTIDIA [in µm] | 20–45×7–12 cylindrical, clavate, lageniform, utriform | 23–26×6–8 clavate | 20–27×7–14 lageniform, ventricose | 15–43×6.5–11 cylindrical, subcapitate, narrowly clavate, lageniform to utriform |
| ODOR | Odorless | Pleasant, quickly disappearing | Occasionally slightly fungoid | Not distinct, when old reminiscent of *L. cristata* |

EUROPE: British Isles, the Netherlands, Italy, France, Czech Republic, Hungary, Ukraine. ASIA: Russia (Far East-Primorye Territory), Sri-Lanka, Israel (first record). NORTH AMERICA: USA. SOUTH AMERICA: Argentina.

DISCUSSION: *Lepiota subgracilis* is relatively common throughout the world. Kühner (1936) considered the name used by Lange (1935) *L. gracilis* as *L. subgracilis*, to be invalid based on a missing Latin diagnosis. The validation by Wasser (1978) is considered to be the accepted citation (Candusso & Lanzoni 1990). *Lepiota kuehneriana* and *L. latispora* are considered synonyms of *L. subgracilis*, because their type specimens differ only in larger size and distinct smell (Locquin 1956, Bon 1981).

*Lepiota subgracilis* is quite variable. Our specimens have smaller pilei and larger, more variable cystidia than those described in Wasser (1980). Specimens described by Candusso & Lanzoni (1990) have relatively smaller spores, while those described by Vellinga (2001) have larger pilei and stipes, larger basidiospores, and shorter, narrower pileipellis hyphae. Detailed descriptions of *L. subgracilis* as given by different authors are compared in Table 3.

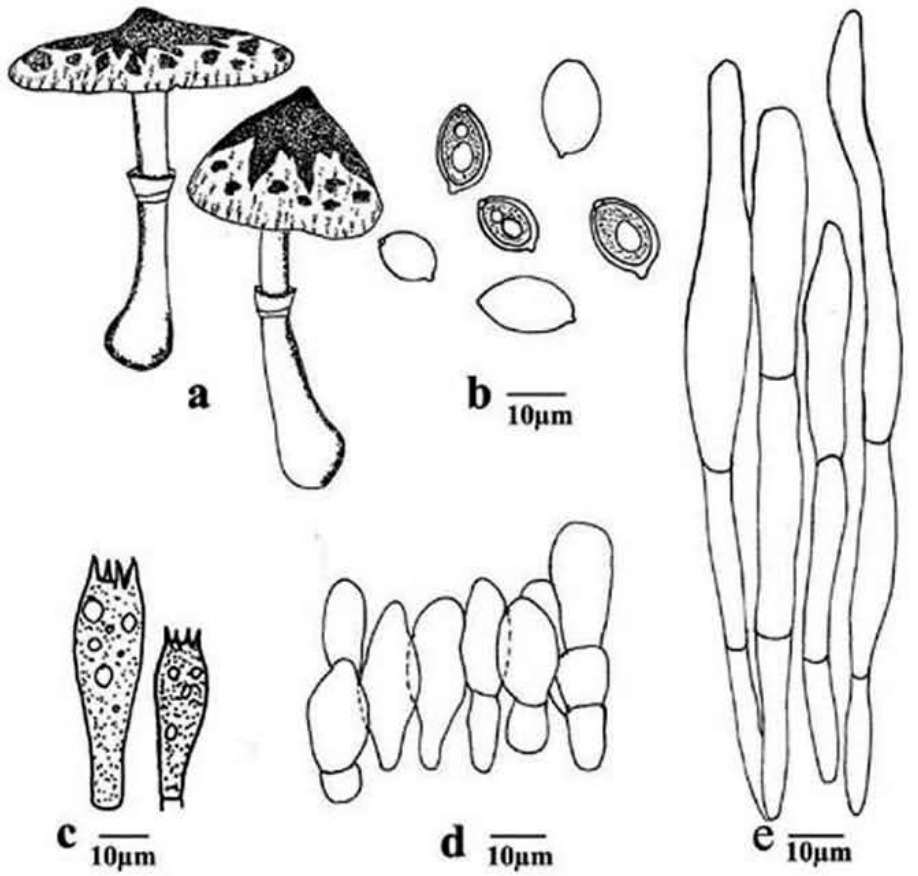


Figure 5: *Macrolepiota konradii* [HAI 200(M). 205. 294].
a – basidiomata, b – basidiospores, c – basidia, d – cheilocystidia, e –elements of the pileipellis

***Macrolepiota konradii*** (Huijsman ex P.D. Orton) M.M. Moser,
Kleine Kryptogamenflora, 3Aufl., IIb/2: 185. 1967 **Figure 5**

**Bas.:** *Lepiota konradii* Huijsman ex P.D. Orton, Trans Brit. Mycol. Soc. 43: 283. 1960.
**Syn.:** *Lepiota excoriata* var. *konradii* Huijsman, Medded. Nedl. Mycol. Ver. 28: 18. 1943. (nom. inval.).
*Leucocoprinus maublancii* Locq., Bull. mens. Soc. Linn. Lyon, 14: 92. 1945.
Misapplied- *Lepiota excoriata* subsp. *mastoidea* sensu Konrad & Maublanc, Icons Selectae Fungorum I: Pl. 10. 1928.
*Lepiota gracilenta* sensu Rea, Brit. Basidiom.: 66. 1922.

PILEUS 5–11 cm in diam., fleshy, when young hemispherical to campanulate, later convex-applanate, with low umbo, pileal surface towards the margin without pellicle, whitish, grey, dingy-grey-brownish, thin-silky-fibrillose, surface layer disrupted on asteroid manner, in the centre dark umbrous, sometimes black-brown, pinkish-brownish. Pileal margin straight, incurved in old specimens,

TABLE 4. Important distinguishing characteristics of *Macrolepiota konradii* and related *Macrolepiota* species

| SPECIES | *M. KONRADII* | *M. MASTOIDEA* | *M. RICKENII* | *M. AFFINIS* | *M. FULIGINEO-SQUARROSA* |
|---|---|---|---|---|---|
| REFERENCE | Personal observation | Candusso & Lanzoni 1990 | Candusso & Lanzoni 1990 | Candusso & Lanzoni 1990 | Didukh et al. 2002 |
| PILEUS [in cm] | 5–11 umbo low, disc umbrous, to black-brownish; margins whitish to grey, surface disrupted in asteroid fashion | 8–12 umbo sharply protruding, dark disc scales appressed fine floccose; margins whitish, cracked | 6–12 disc umbonate & with light brown scales | 5–12 umbo acute, brownish-pink scales on disc; margins creamy cracked, | 6–8 umbo obtuse, cuticle breaks to form central patch surrounded by small concentric squamules on light brown ground |
| PILEIPELLIS — TRICHODERM ELEMENTS [in µm] | Long (50–300 × 9–15), cylindrical, intercellular pigments brownish, incrusted toward base | Long (80–180 × 6–12), cylindrical, walls with/ without pale incrustations | Cylindrical (8–14 diam), walls with/ without pale incrustations | Long, cylindrical (6–12 diam), pigments intercellular, non-incrusting | Disc elements terminal (112.5–200 × 15–22.5) & palisade (117.5–125 × 12.5–17.5); Margin elements cylindrical |
| STIPE [in cm] | 7–15.5 × 0.5–1.5 (2.5–3 at base) | 11–16 × 0.3–1.1 (1.5–2.2 at base) | 10–16 × 1–2 (2–2.5 at base) | 7–13 × 0.6–1 (small bulb at base) | 8–10× 1–1.5 , (≤2.7 diam basal bulb) |
| SPORES [in µm] | 11.4–15.5(20) × 7–9.2(10) ellipsoid, ellipsoid-ovate | 11–16.5(19) × 7.5–10.5 ellipsoid | 12–17 × 7.5–10 ellipsoid | 10–16(20) × 7–9.5(11) ellipsoid | 12.5–17 × 7.5–10 ovoid, subelliptic, almond-shaped |
| CHEILO-CYSTIDIA [in µm] | 17–40 × 10–13 clavate, subfusiform | 20–50 × 12–15 fusiform | 15–33 × 6–15 ventricose-subfusiform | 20–45 × 8–15 ventricose-fusiform | 45–50 × 10–15 slightly clavate, fusiform |

on drying undulate. LAMELLAE free, with small collarium, crowded, broad, with even edge, easily separating from pileus, white, later pale creamy. STIPE 7–15.5×0.5–1.5 cm, central, cylindrical, sometimes slightly twisted, enlarged up to 2.5–3 cm at the base, solid, brownish, grooved, sometimes covered with umbrous adpressed scales, formed by cracking of the surface layer. RING apical, wide, double crown, upper side brown, underside creamy, easily separating, mobile, often with bifurcate edge. CONTEXT dense, white-creamy, unchanging on exposure. Smell and taste pleasant fungal. SPORE PRINT whitish-creamy.

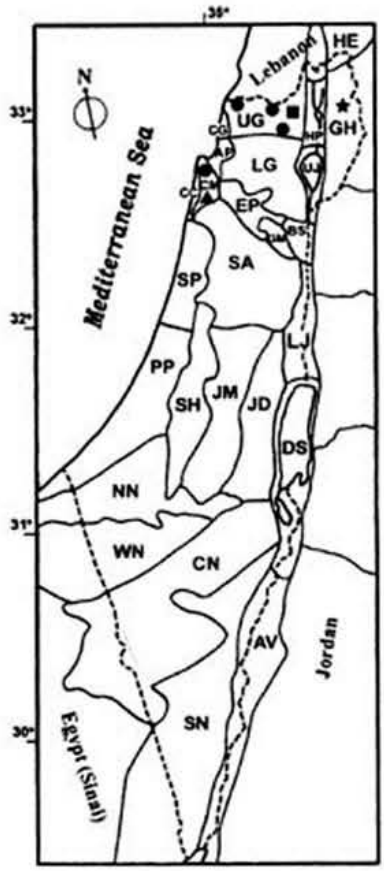


**Figure 6:** Distribution of *Leucoagaricus pilatianus*, *Lepiota apatelia*, *L. subgracilis* and *Macrolepiota konradii* in Israel.

★ – *Leucoagaricus pilatianus*

■ – *Lepiota apatelia*

▲ – *Lepiota subgracilis*

● – *Macrolepiota konradii*

Basidiospores 11.4–15.5(20)×7–9.2(10) µm, ellipsoid, ellipsoid-ovate with a germ pore, with hyaline cap (sometimes slightly visible), with lateral apiculus, with a large refractive droplet, smooth, thick-walled, dextrinoid, congophilous, cyanophilous, metachromatic. Basidia 4-spored, 30–50×10–12(14) µm, clavate. Pleurocystidia absent. Cheilocystidia 20–40×10–13 µm, clavate, subfusiform. Hymenophoral trama irregular. Pileipellis as a trichoderm, with long cylindrical elements, 50–200×9–14 µm, with intercelular brownish pigment, often incrusted at base. Stipitipellis with narrow cylindrical colorless hyphae, 3–10 µm wide, with a brown pigment. Clamp-connections present.

Specimens examined: **Israel.** UG: Mt. Meron, National Park, under *Pinus*, leg. Y. Ur, 14.01.2005, det. S.P. Wasser. UG: Near Mt. Meron, under *Quercus* and *Pinus*, leg. Y. Ur, 03.01.2005, det. S.P. Wasser. CM: Derex Nof Carmel, in *Quercus* groves, leg. Y. Ur, 19.01.2007, det. A. Kosakyan. UG: Goran Park, leg. Y. Ur, 9.02.07, det. A. Kosakyan [HAI 200(M), 205, 294] (Figure 6).

Habitat and distribution: Solitary or in small groups, in broad-leaved and mixed forests, parks, often meadows and fields. Known from Europe: British Isles, Denmark, Belgium, Switzerland, the Netherlands, France, Italy, Hungary,

Austria, Germany, Czech Republic, Slovak Republic, Romania, Poland, Ukraine, Belarus, Moldova. Asia: Russia (Far East-Primorye Territory), Viet Nam, Israel (first record).

Discussion: Konrad & Maublanc (1924-37) treated the species as *Lepiota excoriata* subsp. *mastoidea*. Huijsman (1943) renamed the taxon *Lepiota excoriata* var. *konradii* in honor of the Swiss mycologist but provided no Latin diagnosis. Locquin (1945), not knowing about Huijsman's publication, renamed the same fungus as *Leucocoprinus maublancii*. Orton (1960) proposed *Lepiota konradii*, which Moser (1967) later transferred to *Macrolepiota*. Vellinga (2001) considers *Macrolepiota konradii* identical to *M. mastoidea* and *M. rickenii*, which differ from each other only in pileus color; she also cites molecular evidence supporting the group as one taxon (Vellinga, 2001). However, as there are obvious morphological differences in habitus, size and shape of pileal elements, and pileus color and form, we choose to treat the three as independent species pending further studies. Table 4 compares *M. konradii* and its related species.

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# Studies in lichens and lichenicolous fungi: further notes on North American taxa

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**Abstract** — *Abrothallus hypotrachynae*, *Phaeographis atromaculata*, and *Thailoloma hypoleptum* are reported as new to North America. The generic placement of *Megalaria beechingii* and *M. pulverea* is discussed. Conforming to the new generic concepts in the *Graphidaceae* several new combinations are proposed for North America taxa: *Fissurina illiterata* (for *Graphis illiterata*), *Graphis xylophaga* (for *Graphina xylophaga*), *Leiorreuma explicans* (for *Phaeographina explicans*), *Phaeographis asteroides* (for *Phaeographina asteroides*), and *Platythecium floridanum* (for *Graphis floridana*). *Miriquidica mexicana* is placed in synonymy with *M. scotopholis*, and detailed discussion of the taxon is provided.

1. ***Abrothallus hypotrachynae*** Etayo & Diederich, Bib. Lich., 84: 16. 2002.

*Abrothallus hypotrachynae* was described (Etayo 2002) from specimens of *Hypotrachyna* distributed widely in sub-tropical Central and South America. Considering the phytogeographic affinities of the southeastern Coastal Plain of North America (Beeching 2007, Harris 1995, Lendemer 2006) the discovery of this taxon in North America is not surprising. The North American material was found on thalli of the fertile, lividic acid complex containing taxon *Hypotrachyna livida* (Taylor) Hale, which is widespread and common in eastern North America (Brodo et al. 2001). Presently, no other species of *Abrothallus* has been found on *Hypotrachyna* in North America, and thus the lichenicolous fungus can be recognized by its host as well as the small black apothecia that contain K+ blue-green pigments and have a greenish pruina on the disc, and octosporous asci with 2-celled brown ascospores (11-14 x 5.0-5.3µm).

Specimens Examined – **U.S.A. GEORGIA. DEKALB CO.:** Mt. Arabia Trail, near covered bridge, Arabia Mountain Park, 17.xii.2006, *S.Q. Beeching s.n.* (hb. Lendemer). **FLORIDA. LIBERTY CO.:** Torreya State Park, 9.vii.2007, *E.A. Tripp 297 & K. Deregibus* (hb. Lendemer).

2. ***Fissurina illiterata*** (R.C. Harris) Lendemer, **comb. nov.**
MycoBank MB 511041.

*Graphis illiterata* R.C. Harris, Some Florida Lichens, p. 20. 1990. TYPE: U.S.A. Florida, Duval Co., Big Talbot Island State Park, 17.xii.1987, *R.C. Harris 21236* (NY!, holotype).

Harris (1990) separated *Graphis illiterata* from *G. subnitidula* Nyl. ex Tuck., by its larger ascospores (15-17 x 6-7µm), obscure fissurine lirellae, and ecorticate thallus. Originally reported from Florida (Harris 1990) it has subsequently been reported from North Carolina (Lendemer & Yahr 2004). Staiger (2002) transferred *G. subnitidula* to the genus *Fissurina* and, following her generic concepts *G. illiterata* should also be placed in that genus. The combination is made here.

3. ***Graphis xylophaga*** (R.C. Harris) Lendemer, **comb. nov.**
MycoBank MB 511042.

*Graphina xylophaga* R.C. Harris, Some Florida Lichens, p. 14. 1990. TYPE: U.S.A. Florida, Duval Co., Big Talbot Island State Park, trail to "Scrubby Bluff", 17.xii.1988, *R.C. Harris 23900* (NY!, holotype).

Due to the generic concepts employed in the *Graphidaceae* at the time of its description, Harris (1990) described this taxon in the genus *Graphina*. Following the generic circumscriptions of Staiger (2002) *Graphina xylophaga* should be placed in the genus *Graphis*. It is characterized by its laterally carbonized non-striate exciple, lack of lichen substances, and hyaline muriform, I+ blue-violet ascospores (65-90 x 20-30µm, 1-(2)/ascus). The combination is made here.

4. ***Leiorreuma explicans*** (Fink) Lendemer **comb. nov.**
MycoBank MB 511163.

*Phaeographina explicans* Fink, in Hedrick, Mycologia, 25: 313. 1933. TYPE: U.S.A. Alabama, near Montgomery, 1916, *R.P. Burke s.n.* (MICH!, holotype).

For some time the first author (JCL) has been confused as to the identity of *P. explicans* because it is keyed out as lacking norstictic acid in Harris (1995). Examination of the type specimen (and a paratype in NY) revealed the presence of norstictic acid. *Phaeographina explicans*, described by Fink (in Hedrick 1933) from Alabama and Mississippi, U.S.A. is a widespread species in the southeastern United States recognized by its relatively short unbranched lirellae with poorly developed white margins, fully carbonized exciple, inspersed hymenium, brown submuriform ascospores (8/ascus, 24-31 x 9-12µm, 1-2 x 6 celled), and the presence of norstictic acid (although occasional acid deficient

specimens are known). In this species, the hypothecium and lower portions of the hymenium become carbonized in addition to the entire exciple, although the degree of carbonization is not as great as that illustrated for other species of *Leiorreuma* by Staiger (2002). As a result of the aforementioned character, following the generic concepts presented by Staiger (2002), the species would seem best placed in *Leiorreuma*. The new combination is made here.

5. ***Megalaria beechingii*** Lendemer, Opuscula Philolichenum, 4: 39. 2007.

TYPE: U.S.A. Georgia, Rabun Co., Lake Burton Wildlife Management Area, 17.ix.2006, *J.C. Lendemer et al. 7700* (NY!, holotype; CANL!, MSC!, UCR!, HB. LENDEMER!, isotypes).

As has been repeatedly discussed (Ekman & Tønsberg 1996, Fryday 2004, Lendemer 2007) the genus *Megalaria* Hafellner, as defined by Ekman & Tønsberg (1996), consists of several groups of related species that could potentially be recognized at the generic level. Ekman (2001) however, found that division of *Megalaria* was not supported by molecular data and thus the genus remained as circumscribed by Ekman & Tønsberg (1996) until Kalb (2007) described the genus *Catillochroma* to accommodate a group of taxa previously referred to *Megalaria*. This group of taxa essentially differs from the type of *Megalaria*, *M. grossa* (Pers. ex Nyl.) Hafellner, in having the exciple composed of an outer layer of prosoplectenchymatous hyphae and an inner layer of "textura intricata" filled with minute crystals and numerous open spaces between the hyphae. The "double" layered exciple of *Catillochroma* contrasts with the uniformly prosoplectenchymatous exciple of *Megalaria grossa*. Among the species transferred to *Catillochroma* was *M. pulverea* (Borrer) Hafellner & E. Schreiner, which is comparable in excipular anatomy and chemistry to the recently described southern Appalachian endemic *M. beechingii*. As a result of this relationship, the first author (JCL) was curious if *M. beechingii* should be transferred to *Catillochroma* as well. Thus, in addition to material of *Catillochroma*, specimens of *M. beechingii*, *M. grossa*, and *M. pulverea* were examined.

The type species of *Catillochroma* is *C. endochroma* (Fée) Kalb (basionym *Lecanora endochroma* Fée), a species that had not previously been treated in the genus *Megalaria*. While all of the species examined share the character of a double layered exciple, the outer layer of the exciple of *C. endochroma* and *C. intermiscens* (Nyl.) Kalb is composed of loose, thick walled hyphae with enlarged terminal cells that swell significantly in KOH. *Megalaria beechingii*, and *M. pulverea* do not share these characters, and rather have an outer layer more comparable to the species remaining in *Megalaria* in consisting of comparably compact thin walled hyphae without the terminal cell enlarged.

In addition to the differences in excipular anatomy, Kalb (2007) stressed that all species of *Catillochroma* contain zeorin in addition to atranorin, whereas

*Megalaria* in its newly restricted sense lacks zeorin. Of further interest is the fact that all of the species included in *Catillochroma* contain zeorin, whereas the species included in *Megalaria* lack that substance. Unfortunately, the type of *Megalaria* lacks any lichen substances whereas the majority of other species placed in *Megalaria* produce atranorin, a character shared by *Catillochroma*. The chemical disparity between *M. grossa* and the bulk of *Megalaria* mirrors the considerable morphological differences between the type of the genus and the rest of the species currently placed in *Megalaria*. Could a similar situation have occurred in *Catillochroma*, where the type species differs in excipular characters and apothecial pigmentation from some of the species included in the genus? The argument could be made to remove *M. beechingii* to *Catillochroma* as it is clearly distinct from *M. grossa*, but a similar argument could be made for retaining the species in *Megalaria* as it differs significantly from *C. endochroma*.

As is evidenced by the recent discovery of the affinities of *Lecanora endochroma*, and as has been discussed by Fryday (2004), there are additional species of *Megalaria* s. lat. remaining in other genera, as well as several apparently undescribed taxa. The possibility that these unstudied taxa may shed light on the relationship between *M. grossa* and the rest of *Megalaria* as well as *Catillochroma* should be considered. Until the status of these genera is investigated with molecular data, and a detailed study of *Megalaria* is carried out including the species recently described by Fryday (2004, 2007), Lendemer (2007), and Jagadeesh et al. (2007) the first author advocates a conservative approach, recognizing *Catillochroma* with *C. endochroma* as the type and including any species with similar excipular anatomy such as *C. intermiscens*. Concurrently it seems logical to suggest that *M. beechingii*, *M. pulverea*, and any other species with a similar excipular type be retained in *Megalaria* until their generic placement is confirmed with molecular methods.

6. ***Miriquidica scotopholis*** (Tuck.) B.D. Ryan & Timdal,
in Nash et al., Lichen Flora of the Greater Sonoran Region, 2: 363. 2004.

*Biatora scotopholis* Tuck., Lich. Calif., p. 24. 1866. TYPE: U.S.A, California, sandstone rocks on coast, *H.N. Bolander s.n.* (NY! [Amherst College 73765], lectotype **designated here**).

*Lecidea scotopholis* (Tuck.) Herre, Proc. Wash. Acad. Sci., 12: 80. 1910.

*Psora scotopholis* (Tuck.) Fink, Lich. Flora of the United States, p. 213. 1935.

*Psorula scotopholis* (Tuck.) Gotth. Schneid., Bib. Lich., 13: 141. 1980 "1979".

*Lecanora scotopholis* (Tuck.) Timdal, Nord. J. Bot., 4(4): 539. 1984.

**Syn. nov.** *Miriquidica mexicana* Rambold, Sipman & Hertel, Mycotaxon, 58: 319. 1996. TYPE: MEXICO, Baja California, 23 km SE of El Rosario along Route 1, 78 km on road from San Quintín to Parador Punta Prieta 30°02'N 115°31'W, 200 m., 5.i.1989, *H. Sipman 24923* (B!, holotype)

Though common in central and southern California as well as Baja California, *Miriquidica scotopholis* is rarely collected, partly because many thalli are sterile. It can be mis-determined as a member of the *Lecidea atrobrunnea* group or as *Rhizocarpon bolanderi* (Tuck.) Herre based on the general aspect of its areolate thallus and its lecideine apothecia.

In its nomenclatural history, the taxon has been placed in six genera and has been described twice.

The taxon was first described by Tuckerman (1866) from a Bolander collection from sandstone rocks on the coast of California. Tuckerman (1888) later reported it from the Dallas of the Columbia in Oregon from a Hall collection made in 1871. We examined the Hall specimen (FH!) and found it does not contain miriquidic acid. Although it looks very similar to *M. scotopholis* it is probably a member of the *Lecidea atrobrunnea* group. Timdal (1984) published TLC data for the holotype, reporting miriquidic acid and four accessory unknowns. The holotype was at ASU during the Sonoran lichen flora project. Bruce Ryan prepared a modern description (Nash et al. 2004a). It was apparently lost in the mail when the loan was returned to FH after Ryan's death. Thankfully, an isotype is among the material retained at Amherst College after the transfer of Tuckerman's herbarium to Harvard University. The specimen was subsequently donated to NY in 1989 and is here selected as the lectotype.

Based probably on its lecideine apothecia, Herre (1910) transferred *M. scotopholis* to *Lecidea*. He reported it from Mt. San Bruno on sandstone at 1000 ft, and on the Santa Cruz Peninsula to 1800 ft. Hasse (1913) reported the species from Yosemite Valley and Catalina Island as common in higher ranges of cismontane California. The specimens documenting these reports need to re-examined. Fink (1935), based on its sub-squamulose thallus, transferred the species to *Psora*. Schneider (1980) transferred the species to *Psorula* but it differs from the other member of the genus, *P. rufonigra* (Tuck.) Gotth. Schneid., in "having filiform conidia, a different ascus type, brown epihymenium, loose paraphyses, pale hypothecium, and different chemistry" (Nash et al. 2004a). Timdal (1984) recognized these differences, but because the ascus type of *M. scotopholis* is intermediate between a *Bacidia*-type and a *Lecanora*-type, he placed it in *Lecanora*. Eventually Ryan & Timdal (Nash et al. 2004a) transferred it in to the genus *Miriquidica*. They saw no specimens other than the type during the Sonoran Flora project and reported it as "expected but not confirmed from south of coastal, central California".

Based on collections from Baja California, where the taxon is common, Rambold et al. (1996) described *Miriquidica mexicana*; they were unfamiliar with *M. scotopholis*, which at that time was included in *Lecanora*. Bjorn Owe-Larsson, while traveling with the second author (KK) in 2004 pointed out the species as *M. mexicana* in the San Jacinto and Santa Monica Mountains.

They reported it as new to California (Knudsen & Owe-Larsson 2005). They recognized there were serious problems keying out specimens as either *M. mexicana* or *M. scotopholis* using the key for the genus in Nash et al. (2004a). After numerous collections, and waiting for the holotype to hopefully be found, we re-examined the problem. We found *M. mexicana* does not differ from other specimens in California referable to *M. scotopholis*. The two descriptions in Nash et al. (2004a) erect a false division in the range of variability in epihymenial color, spore width, thallus color, whether or not erumpent apothecia have an evanescent thalline margin, and color of apothecial margin. *Miriquidica mexicana* is proposed here as synonymous with *M. scotopholis*. In the protologue of *M. mexicana* (Rambold et al. 1996) the chemistry is listed as miriquidic acid or lobaric acid. Specimens with lobaric acid have since been described as the new species *Protoparmelia ryaniana* van den Boom, et al. (van den Boom et al. 2007). That species has maritime distribution, is a juvenile parasite, and is known from North Baja California in Mexico, and Point Loma (*Knudsen 8242*, UCR) and Catalina Island in California. Further, chemical analysis of the type of *M. mexicana* revealed that it contains several unknowns in addition to miriquidic acid, and is chemically identical to the lectotype (and all other specimens of this taxon) we have examined.

A new description is supplied due to the confusion in Nash et al. (2004a) and the segregation of *Protoparmelia ryaniana* (van den Boom et al. 2007). The protologue for *M. mexicana* is detailed and can be referred to for more information (Rambold et al. 1996).

Description. Thallus crustose, epilithic, contiguous to dispersed, areolate, up to 1 mm thick, often covering areas up to 5 cm or more, confluent with other thalli. Usually it is surrounded by a distinct black dendroid-branched prothallus. Areoles 0.10-1.5 mm diam, becoming convex, or twisted, angular to round, the rim often upturned, undulate or crenulate, prominent and black, sometimes white pruinose. The attachment can elongate and areoles become thicker and imbricate, and appear squamulose. The surface of areoles is reddish-brown to almost black, rarely a yellowish-brown, usually shiny. Upper cortex is up to 50 µm thick, the upper layer brown-pigmented with an epinecral layer, the lower layer hyaline. The lower surface is corticate, black-brown. The attachment is broad, over one-half of the diameter of areoles. The alga layer is thin, uneven, interrupted. Photobiont is chlorococcoid green algal, 7-12 µm diam. Medulla, white, thick, I-, prosoplectenchymatous. The apothecia are erumpent, 0.3-1 mm in diam., becoming sessile, lecideine, rarely with thalline margin which does not persist, but with scattered algal cells in the exciple. The disc is black, plane to convex, reddish-brown when wet, epruinose, with prominent black to grayish margin, sometimes white pruinose. The exciple is up to 90 µm wide,

outer layers shades of olive and brown becoming hyaline within, inspersed with crystals of miriquidic acid (Rambold et al. 1996) and scattered algal cells. Hymenium is up to 50 µm tall, I- or I+ blue, hyaline below, greenish to brown in upper part including epihymenium; epihymenium 7-12 µm tall; paraphyses 1.5-2.5 µm wide at mid-level, rarely branched or anastomosing, apices expanded to 4-5 µm, sometimes with brown pigment. The hypothecium is up to 100+ µm thick with crystals of lichen substances in lower area (Rambold et al. 1996). Asci 30-45 x 7-14 µm wide. The outer wall of ascus is I+ blue. "Tholus max. 7-8 µm, min.5-6 µm high, without or with small ocular chamber, with narrow axial body" (Rambold et al. 1996) with an almost *Lecanora*-type stain. Ascospores hyaline, simple, ellipsoid, 6-11 x 3-5 µm wide. Condiomata pycnidial, black, immersed, mostly 0.1 mm, usually near edges of areoles. Conidiophores "similar to type V of Vobis [1980]" (Rambold et al. 1996). Conidia long filiform 20-40 x 0.7-1 µm.

Chemistry. miriquidic acid and four or five unknowns.

Distribution. Washington to Baja California (Rambold et al. 1996, Nash et al. 2004a), on acid rock, from 13-1602 m, associated with Coastal Sage Scrub, *Adenostoma fasciculatum* or *A. sparsifolium* chaparral, mesic mixed chaparral, oak woodland, pinyon pine woodland, or conifer or mixed oak-conifer forest. It occurs usually in mixed saxicolous lichen associations often with *Aspicilia* species. In a recent study of San Jacinto Mountains, mostly above 2000 meters, no *M. scotopholis* was found though it is abundant below 1600 meters (Knudsen & Kramer 2007). Thomson (1997) reported it as far north as Saskatchewan in Canada but we saw no vouchers from that study.

Illustrations. Rambold et al. (1996), p. 321; Nash et al. (2004b).

Discussion. The thallus is often sterile and appears blackish and undistinguished in the field and is easily ignored. The prothallus is distinctive but not always evident. In southern California and Baja it is usually locally abundant on hard or decaying outcrops, rocks, and even on pebbles of basalt, volcanic rock, granite, and sandstone. The species is easily separated from the *Lecidea atrobrunnea* group by ascus stain and chemistry because none of the *Lecidea* species contain miriquidic acid. *Protoparmelia ryaniana* has lobaric acid, narrower spores (2.5-3 µm), shorter filiform conidia (15-16 µm long) and a strictly maritime distribution. *Rhizocarpon bolanderi* has large three-septate to murifom spores, two per ascus. In the range of *M. scotopholis* in central and southern California we collected no other *Miriquidica* species, although *M. garovaglii* is similar to *M. scotopholis* it differs in having stictic acid in its medulla and larger ascospores; and has been collected on San Clemente Island in Los Angeles County, California, by Charis Bratt, the specimen determined by Alan Fryday (Shirley Tucker, pers. comm.)

Selected Specimens Studied – **U.S.A. CALIFORNIA. HUMBOLDT CO.:** Bald Mountain, 18 km E of Arcata, on open rocky ridge top, 40°52'N 123°52'W, 920 m., on rock, iii.2001, *B. McCune et al. 25825* (hb. McCune). **ORANGE CO.:** Santa Ana Mountains, Weir Canyon, along Windy Ridge Road 33°50'11"N 117°43'43"W, 280 m., on sandstone, 19.v.2006, *K. Knudsen 6181.3* (UCR); Santa Ana Mountains, Santiago Peak below transmitter tower, 33°42'52"N 117°32'03"W, 1633 m., on boulders and rock outcrops, 22.vi.2004, *J.C. Lendemer 2848 & K. Knudsen* (hb. Lendemer). **RIVERSIDE CO.:** Menifee Hills, Bundy Canyon, 33°37'32"N 117°14'23"W, 487 m., on rock, 23.vi.2004, *J.C. Lendemer 4185 & K. Knudsen* (hb. Lendemer); Santa Ana Mountains, Elsinore Peak, 33°35'32"N 117°21'01"W, 926 m., on volcanic rock in chaparral, 25.xi.2005, *K. Knudsen 4376* (UCR); San Jacinto Mountains, Idyllwild, near Inspiration Point, 33°43'36"N 116°45'03"W, 1602 m., on granite boulders in full sun in *Pinus jefferyii* forest, 4.viii.2005, *K. Knudsen 3439* (UCR). **SAN BENITO CO.:** Pinnacles National Monument, along Chalone Creek, 36°29'05"N 121°10'03"W, 321 m., on volcanic rock in riparian woodland, *K. Knudsen 4146 & B. Hill* (SBBG, UCR). **SAN BERNARDINO CO.:** Joshua Tree National Park, Key's Ranch, 34°02'83"N 116°09'64"W, 1268 m., on granite, *K. Knudsen 3037 & T. la Doux* (UCR); Granite Mountains, Sweeney Granite Mountain UC Reserve, canyon above Yucca Bajada Camp, 34°48'52"N 116°38'37"W, 1237 m., on granite, 12.xii.2006, *K. Knudsen 7991.1 & R. Muertter* (UCR). **SAN DIEGO CO.:** Cuyamaca Mountains, Cuyamaca State Park, above Sweetwater River, 32° 54' 41"N, 116° 34' 30"W, 1222 m, on granite, 10.x.2007, *K. Knudsen 9125* (SD); Palomar Mountain, Palomar State Park, below Nate Harrison Grade, along Adams Trail, 33°20'41"N 116°55'14"W, 1475 m., on hard granite in shade of oak-conifer woodland, 13.v.2005, *K. Knudsen 2934.1 & M. Knudsen* (UCR). **SAN LUIS OBISPO CO.:** San Simeon, San Simeon State Park, coast southeast of state park headquarters on bluff above shore, 33°38'25"N 121°08'28"W, 18 m., on volcanic rock in costal sage scrub, 10.i.2007, *K. Knudsen 8207* (FH, PRM 857299, hb. Lendemer). **WASHINGTON. KITTITAS CO.:** by Columbia River, 8 km S of I-90 bridge, 50 km ESE of Ellensberg, 47°120°N, 185 m., viii.1987, *B. McCune 17059* (hb. McCune).

### 7. *Phaeographis asteroides* (Fink) Lendemer, **comb. nov.**

MycoBank MB 511164.

*Phaeographina asteroides* Fink, Mycologia, 19: 219. 1927. TYPE: Porto Rico, in an open field near Mayaguez, *B.E. Fink 981* (MICH, holotype; NY!, isotype).

*Phaeographis asteroides* was described by Fink (1927) from Puerto Rico and subsequently reported from North America by Harris (1990). The white margins of the lirellae, which are immersed and initially hide the disc, are rather distinctive and make this taxon fairly easy to recognize in the field. Unfortunately the type collection is in poor condition, likely due to the age of the thallus, and the margins of the lirellae have been distinctly abraded or eroded. All of the North American collections of this species are from Florida. Continued field work in the southeastern United States will likely reveal it to be more widespread. In addition to the distinctive white margined lirellae, the species can be recognized by the fully carbonized exciple, hymenium that is not inspersed, brown submuriform ascospores (8/ascus, 28-35 x 10-15µm, 6-8 x (1)-2-4 celled), and lack of lichen substances.

8. ***Phaeographis atromaculata*** (A.W. Archer) A.W. Archer, Telopea, 11: 75. 2005.

Syn. *Phaeographis kalbii* Staiger, Biblio. Lich., 85: 332. 2002.

**Syn. nov.** *Phaeographis illitoraticola* Lendemer, R.C. Harris & Yahr, nom. nud., Evansia, 21(3): 128. 2004.

When the first author (JCL) and his colleagues first encountered this species in coastal North Carolina, USA they assigned it a tentative name since they presumed it was an undescribed species (Lendemer & Yahr 2004) related to the then recently described *Phaeographis kalbii* which differed in having two-spored asci according to Staiger (2002). While those authors were preparing a description of the North American material, Archer (2006) placed *P. kalbii* in synonymy with *P. atromaculata* noting the asci to be monosporous in Australian material. Since the North American material shared this character it was clear further study was needed. After comparison of the North American material with the illustrations provided by Archer (2006) and Staiger (2002), as well as photographs of typical South American material kindly provided by Robert Lücking, it was clear that the North American populations belonged to *P. atromaculata*. Following Archer (2006) *P. atromaculata* has a pan-tropical distribution, and the occurrence of such a widely distributed taxon is not unexpected in the southeastern United States. The species is easily recognized by its circular to somewhat elongate apothecia, often with ragged thalline margins, asci containing a single large brown densely muriform ascospore, and the presence of norstictic acid in the thallus. The present collections indicate the species to be restricted to maritime habitats in North America, but further collecting my reveal it to have a wider habitat distribution in the more tropical parts of the southeastern United States. These are the first reports of *P. atromaculata* from North America.

Specimens Examined. – **U.S.A. NORTH CAROLINA. BRUNSWICK CO.:** Bald Head Island, 5.vii.2003, *J.C. Lendemer 1622 & R. Yahr* (hb. Lendemer), *R. Yahr 4848* (DUKE), *R. Yahr 4855* (DUKE). **CARERET CO.:** Cape Lookout National Seashore Shackleford Banks, 19.iii.2003, *J.C. Lendemer et al. 738* (hb. Lendemer); Theodore Roosevelt Natural Area, 20.iii.2003, *J.C. Lendemer et al. 706* (hb. Lendemer), *R. Yahr 4691* (DUKE), *R. Yahr 4734* (DUKE).

9. ***Platythecium floridanum*** (Tuck.) Lendemer, **comb. nov.**

MycoBank MB 511043.

*Graphis floridana* Tuck., Syn. N. Amer. Lich., 2: 126. 1888. TYPE: U.S.A., East Florida, 1877, *J. Donnell Smith s.n.* (FH-TUCK! [ex herb. C.F. Austin], lectotype selected here; NY!, isolectotype).

*Graphina floridana* (Tuck.) R.C. Harris, Some Florida Lichens, p. 14. 1990.

At the time of its description, Tuckerman (1888) considered *Graphis floridana*, to be closely related to related to *Graphis grammitis* Fée (= *Platythecium grammitis* (Fée) Staiger). Because of its submuriform ascospores Harris (1990) transferred the species to *Graphina* Müll. Arg. Following the current

generic concepts proposed by Staiger (2002) this taxon should be placed in *Platythecium* Staiger, and the new combination is made here. *Platythecium floridanum* is a distinctive species because of its long branching lirellae that lack carbonization, the presence of abundant norstictic acid in the thallus, and hyaline submuriform ascospores (15-20 x 7-9µm, 8/ ascus).

10. ***Thalloloma hypoleptum*** (Nyl.) Staiger, Bib. Lich., 85: 437. 2002.

*Graphis hypolepta* Nyl., Acta Soc. Sci. Bot. Fenn., 7: 472. 1863.

**Syn. nov.** *Graphis anguinoides* R.C. Harris nom. nud., More Florida Lichens, p. 12. 1995.

Harris (1995) published *Graphis anguinoides* as a nomen nudum. When Staiger (2002) revised the generic concepts in the *Graphidaceae* she transferred the related species *G. anguina* (Mont.) Nyl., to *Thalloloma* Trevis. While the first author (JCL) was working on this paper Richard Harris suggested the material he referred to *G. anguinoides* might be conspecific with *T. hypoleptum*, a species that is widely distributed in tropical Central/South America (Staiger 2002). Examination of the material in light of Staiger's treatment confirms this, and *T. hypoleptum* should be added to the North American checklist. The species is common on *Ilex, Magnolia,* and *Taxodium* in subtropical portions of the southeastern United States, and although the specimens reported here are from Florida, the species will likely be found elsewhere in the region as further collections accumulate. *Thalloloma hypoleptum* can be recognized by the creamy white thallus with inconspicuous brownish lirellae lacking any carbonization. The ascospores are eight to the ascus, hyaline, and transversely septate (20-30 x 6-9µm), with (5)-8-10 cells. The thallus in all of the North American material examined to for this paper was UV+ yellow due to the presence of lichexanthone, however a chemotype lacking lichexanthone was reported by Staiger (2002).

Specimens Examined. – **U.S.A. FLORIDA. BAKER CO.:** Along C.R. 127, 26.xi.1996, *R.C. Harris 39298* (NY). **BAY CO.:** N of Co. Rd. 388, 1.xii.1994, *R.C. Harris 35768* (NY). **COLLIER CO.:** Big Cypress National Preserve, 9.xii.1992, *R.C. Harris 30179* (NY). **COLUMBIA CO.:** Osceola National Forest, 2.i.1991, *W.R. Buck 19077* (NY). **ESCAMBIA CO.:** Along Pineville Road ca. 2 mi W of Co. Rd. 97A, 8.xii.1993, *R.C. Harris 31950* (NY). **FLAGLER CO.:** Along Co. Rd. 304 at Sweetwater Creek, 6.i.1996, *R.C. Harris 37407* (NY). **JEFFERSON CO.:** E of St. Rd. 59, 12.xii.1993, *R.C. Harris 32323* (NY). **LAFAYETTE CO.:** Just W of Fla. Hwy. 51, 29.xi.1994, *R.C. Harris 35548* (NY). **LEVY CO.:** Black Point Swamp, 30.xi.1992, *R.C. Harris 29390* (NY). **LIBERTY CO.:** 21.5 mi E of Wilma, 28.xii.1990, *R.C. Harris 26111* (NY). **WAKULLA CO.:** Apalachicola National Forest, 1.i.1991, *R.C. Harris 26209* (NY). **SUMTER CO.:** Withlacoochee State Park, 5.xii.1996, *R.C. Harris 39843-A* (NY).

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# *Ascomycetes* of Sonora, Mexico. 1: The Ajos-Bavispe National Forest Reserve and Wildlife Refuge

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**Abstract** — Eight species of *Ascomycetes* from the Ajos-Bavispe National Forest Reserve and Wildlife Refuge, located in Sonora, Mexico are recorded for the first time in the Mexican mycobiota: *Cenangium yuccae, Diatrype standleyi, Eutypa koschkelovae, E. podanthi, Gloniopsis praelonga, Hypocrea scutelliformis, Hysterium insidens,* and *H. truncatulum*. Photomicrographs, descriptions, and some ecological observations are presented.

**Key words** — *Helotiales, Hypocreales, Xylariales, Hysteriales*

## Introduction

The Ajos-Bavispe National Forest Reserve and Wildlife Refuge (ABNFR) is located in the northeastern part of the state of Sonora in Mexico, and has five mountain ranges. The vegetation is mainly pine forest, pine-oak forest, oak forest, gallery forest, and microphyllous desert scrub. The biological diversity catalogued for the ABNFR includes a little over 1,230 vascular plant species, along with 358 vertebrates and 92 diurnal butterflies (Guerra 1998).

There are some contributions to the taxonomy of *Ascomycetes* from Sonora (Esqueda et al. 1992, Pérez-Silva et al. 1996, San Martín et al. 1999a,b,c), but only three from ABNFR for *Aphyllophorales* (Montaño et al. 2006) and *Myxomycetes* (Moreno et al. 2006, Lizárraga et al. 2007). Therefore, these are the first records of *Ascomycetes* from ABNFR and are new to the Mexican mycobiota.

The collections studied were obtained from seven types of vegetation in the ABNFR that were sampled seasonally from fall 2004 to summer 2005: pine-

Table 1. Sampling sites on the Ajos-Bavispe National Forest Reserve and Wildlife Refuge.

| Site | N | W | Altitude | Vegetation |
|---|---|---|---|---|
| Municipality of Cananea | | | | |
| 1. El Campamento | 30°58′22″ | 109°57′38″ | 1997 m | POF-GF |
| Municipality of Fronteras | | | | |
| 2. El Frijolito | 30°56′35″ | 109°57′21″ | 2286 m | POF |
| 3. La Valdeza | 30°38′06″ | 109°47′22″ | 1546 m | OOF |
| Municipality of Cumpas | | | | |
| 4. Km 8 Moctezuma to La Antena road | 29°58′53″ | 109°39′52″ | 818 m | MDS |
| 5. La Antena | 30°00′02″ | 109°33′29″ | 1653 m | PF |
| 6. La Selva | 29°57′41″ | 109°36′55″ | 881 m | SS |
| 7. El Mezquital | 29°57′26″ | 109°38′23″ | 882 m | M |

Vegetation types: pine-oak forest associated with gallery forest (POF-GF); pine forest (PF); pine-oak forest (POF); oak open forest (OOF); microphyllous desert scrub (MDS); mesquite (M); subtropical scrub (SS).

oak forest associated with gallery forest, pine forest, pine-oak forest, oak open forest, microphyllous desert scrub, mesquite, and subtropical scrub. The seven sites were geo-referenced with a GPS Garmin 12XL, using Datum NAD-27 for digital image processing.

The specimens were collected and conserved following the recommended mycological techniques for *Ascomycetes* (Dennis 1978, Breitenbach & Kranzlin 1981). Species identification was based on Ellis & Everhart (1892), Seaver (1978), Dennis (1978), and Sivanesan (1984); and specific literature as Rappaz (1987), Barr (1990), and Chacón (2004, 2005). The specimens have been deposited in the macromycetes collection of the Centro de Estudios Superiores del Estado de Sonora (CESUES), with some duplicates in the Herbarium of the Instituto de Ecología, A.C. (XAL) in Xalapa, Veracruz.

## Species List

***Cenangium yuccae*** Clem. & E.G. Clem. ex Seaver, N. Amer. Cup-fungi. Inoperc.: 303. 1951

Apothecia 0.8 to 2 mm, erumpent, hysterothecioid at first with involute margins; when mature open and cup-shaped; hymenium yellowish when fresh

to light brown when dry, with a darker brown exterior; sessile appearance but fixed to substrate by a short stalk, robust. Asci 90-115 × 7-11 µm, cylindrical, octosporate, base ending in a short stipe, apical pore amyloid. Ascospores 9-12 × 6-8 µm, ellipsoid, thin-walled, uniseriate in the ascus. Paraphyses 3-5 µm in diam., filiform, hyaline and septate.

SPECIMENS STUDIED: LOCALITY 2, leg. F. Méndez & S. Gómez, 23.VIII.2005, on dry *Yucca* sp. leaf, in ecotone with pine-oak forest, *CESUES 5994, Chacón-5575 (XAL).*

OBSERVATIONS The Mexican collection studied is consistent with *C. yuccae* as described by Seaver (1978) based on specimens from Colorado and California in the U.S.A. This author described the species as having slightly smaller apothecia (1 mm), and paraphyses (2 µm in diam.). Worldwide, this taxon has been recorded on rare occasions, and this is the first record in the Mexican mycobiota.

***Hypocrea scutelliformis*** Berk. & Ravenel, in Ellis & Everhart, N. Amer. Pyrenomyc.: 80. 1892

Stromata 3 mm in diam., discoidal with some dents in center, apparently sessile but short-stalked, robust, dark with brown-reddish powder. Surface dotted with perithecia ostioles; context whitish to light brown. Perithecia 100-150 × 6-120 µm, globose to subglobose, arranged in one layer (monostichous), on rare occasions polystichous; neck very short. Asci 60-80 × 4-6 µm, cylindrical-clavate, inamyloid. Ascospores 2-4 × 2.5-4.5 µm, globose to subellipsoidal, some narrower at one end, wall minutely warted, hyaline with greenish tones, octosporate at first, ending with 16 part-spores, each with an oil droplet at the center.

SPECIMENS STUDIED: LOCALITY 7, *leg.* A. Sánchez, 24.II.2005, solitary, on fallen branches in mesquite vegetation, *CESUES 5994.*

OBSERVATIONS: The Mexican collection agrees well with the description of *H. scutelliformis* given by Seaver (1910) and Ellis & Everhart (1892). Among its distinguishing characteristics are discoidal stromata and asci octosporate at first, but ending in 16 part-spores. Including this record, three species of *Hypocrea* are known for Mexico; the others are *H. citrina* var. *americana* Canham, cited for Morelos and Hidalgo (Chacón & Guzmán 1983) and *H. rufa* (Pers.: Fr.) Fr., recorded for Veracruz (Welden & Guzmán 1979).

***Diatrype standleyi*** Fairm., Mycologia 10: 240. 1918

Stromata (1.5-) 2-2.5 mm in diam., erumpent, partially covered with remnants of host bark, dark, sometimes with slight purplish-reddish tones; surface dotted with perithecia ostioles; context brown to light brown. Perithecia (including neck) 550-620 × 190-290 µm, globose to piriform, mono- or polystichous. Ostioles with 3-4 apical openings radially arranged. Asci 60-100 ×

7-10 µm, cylindrical-clavate, base ending in a thin stipe, apical pore inamyloid. Ascopores 10-13 × 3-4 µm, alantoid, pale yellowish to brownish-red in mass, irregularly biseriate in ascus.

SPECIMENS STUDIED: LOCALITY 1, *leg.* F. Méndez & S. Gómez, 21.I.2005. On fallen *Quercus* sp. leaves in pine-oak forest associated with gallery forest, *CESUES* 5400. *Ibidem*, 24.VIII.2005, *CESUES 5841, Chacón-5613 (XAL).*

OBSERVATIONS: The Mexican material fits with *D. standleyi*, as described by Rappaz (1987) based on specimens from New Mexico, U.S.A. Similar species include *D. praeandina* (Speg.) Rappaz, recorded for Argentina, but the latter has larger stromata (3-20 × 1-2 mm), while its asci (40-50 × 5-6 µm), ascospores (9-12 × 2.2-2.8 µm), and perithecia (200-400 µm) are smaller.

***Eutypa koschkelovae*** Frolov, Nov. Sist. niz. Rast. 7: 194. 1970

Stromata irregular, spread out, 1-10.5 × 1-2.5 mm, occasionally linear, appearance superficial to erumpent, dark, partially covered by bark remnants; context light yellowish-brown; surface dotted with perithecia ostioles. Perithecia 230-440 × 160-340 µm, globose to subglobose. Ostioles 180-295 × 90-160 µm, prominent, with 3-4 radially arranged, linear openings at the apex. Asci (32-) 35-45 × 5-9 µm, cylindrical-clavate, base ending in a long, thin stipe, apical pore inamyloid. Ascospores 6.5-10 × 1.5-2.5 µm, allantoid, hyaline to pale yellowish, ochraceous color in mass.

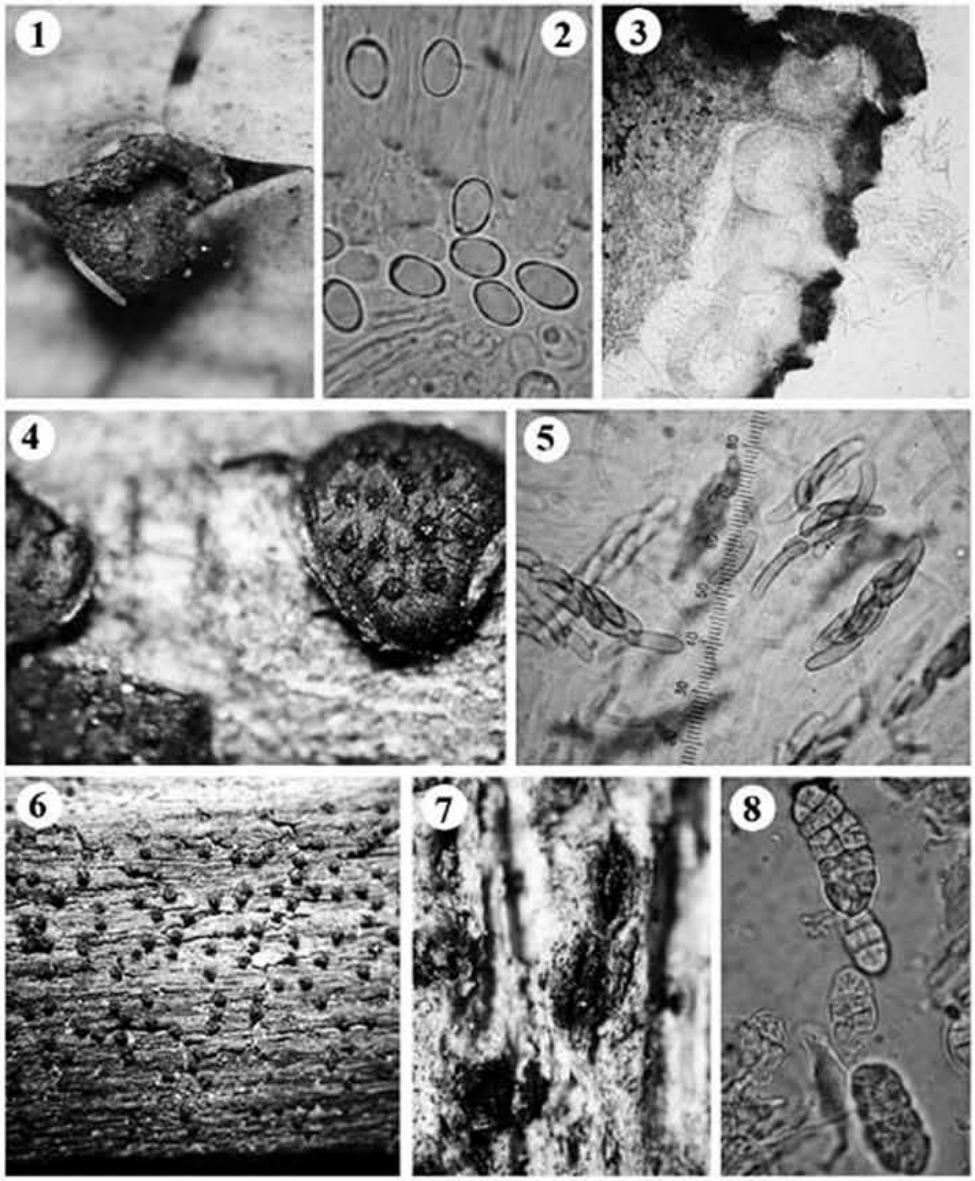
SPECIMENS STUDIED: LOCALITY 4, *leg.* F. Méndez & S. Gómez, 25.VIII.2005, on bark and fallen trees in microphyllous desert scrub, *CESUES 6167, Chacón-6631 (XAL).*

OBSERVATIONS: All macro- and microscopic characteristics fit with E. koschkelovae, as cited by Rappaz (1987) based on a Russian collection from *Calligonum* sp. (*Polygonaceae*). The host was not identified in the Sonoran collection. According to this author, the species was previously only known from its type locality (Russia). This is the second documentation of this species for the rest of the world and the first for the Americas.

***Eutypa podanthi*** Speg., Bol. Acad. Nac. Ci. Cordoba 25: 47. 1921

Stromata one to several cm long, flat, spread out, dark gray with blackish dots from the perithecia ostioles; context minute or absent. Perithecia 440-500 × 430-500 µm, globose, embedded in wood. Ostioles 200-450 × 210-260 µm, having 3-4 apical openings radially arranged. Asci 50-65 × 6-8 µm, cylindrical-clavate, base ending in a thin stalk, apical pore inamyloid. Ascospores 9-12(-14) × (2.8-)3(-3.5) µm, alantoid, yellowish to brown in mass, irregularly biseriate in the ascus.

SPECIMENS STUDIED: LOCALITY 7, *leg.* F. Méndez & S. Gómez, 27.VIII.2005, on dead *Prosopis velutina* wood in mesquite vegetation, *CESUES 6352, Chacón-5673, 5676 (XAL).*




**Figs. 1-8.** Ascomycetes of Sonora, Mexico. 1-2: *Cenangium yuccae*, 1: apothecium, 2: ascospores. 3: *Hypocrea scutellaeformis*, longitudinal section of the stroma showing perithecia. 4-5: *Diatrype standleyi*, 4: stroma, 5: asci and ascospores. 6: *Eutypa podanthi*, stromatic surface showing ostioles. 7-8: *Gloniopsis praelonga*, 7: hysterothecia, 8: ascospores.

OBSERVATIONS: The Mexican collection agrees with *E. podanthi*, as described by Rappaz (1987) based on material collected by Spegazzini in Chile on *Podanthus mitiqui*, but the host was different, and the spore size was smaller (9-13.8 × 2.2-2.5 µm) in the Chilean material. The Spegazzini collection dates from 1918 and,

according to Dr. C. Carmaran (Researcher at the University of Buenos Aires, Argentina, pers. com.) the Chilean material only has two perithecia that are in very poor condition. This opens the possibility for the Mexican material to be proposed here as the epitype. This species, together with *E. koschkelovae*, was only known for the type locality.

***Gloniopsis praelonga*** (Schwein.) Underw. & Earle, Bull. Alabama Agric. Exp. Sta. 80: 196. 1897

Hysterothecia 0.8-1.2 × 0.4-0.7 mm, erumpent to superficial, ellipsoid to fusoid tips, open or partially closed in dehydrated specimens; exterior dark, hymenium same color or lighter. Smooth margin or with fine longitudinal sulcations. Asci 65 × 12 µm, cylindrical-clavate, short-stipitate, apical pore inamyloid. Ascospores 16-29 × 8-13 µm, muriform, hyaline to pale yellow, elliptical to ovoid with obtuse tips.

SPECIMENS STUDIED: LOCALITY 5, *leg.* F. Méndez & S. Gómez, 26.VIII.2005, solitary to gregarious, on bark of fallen trees in pine-oak forest, *CESUES 6239, Chacón-6658 (XAL).*

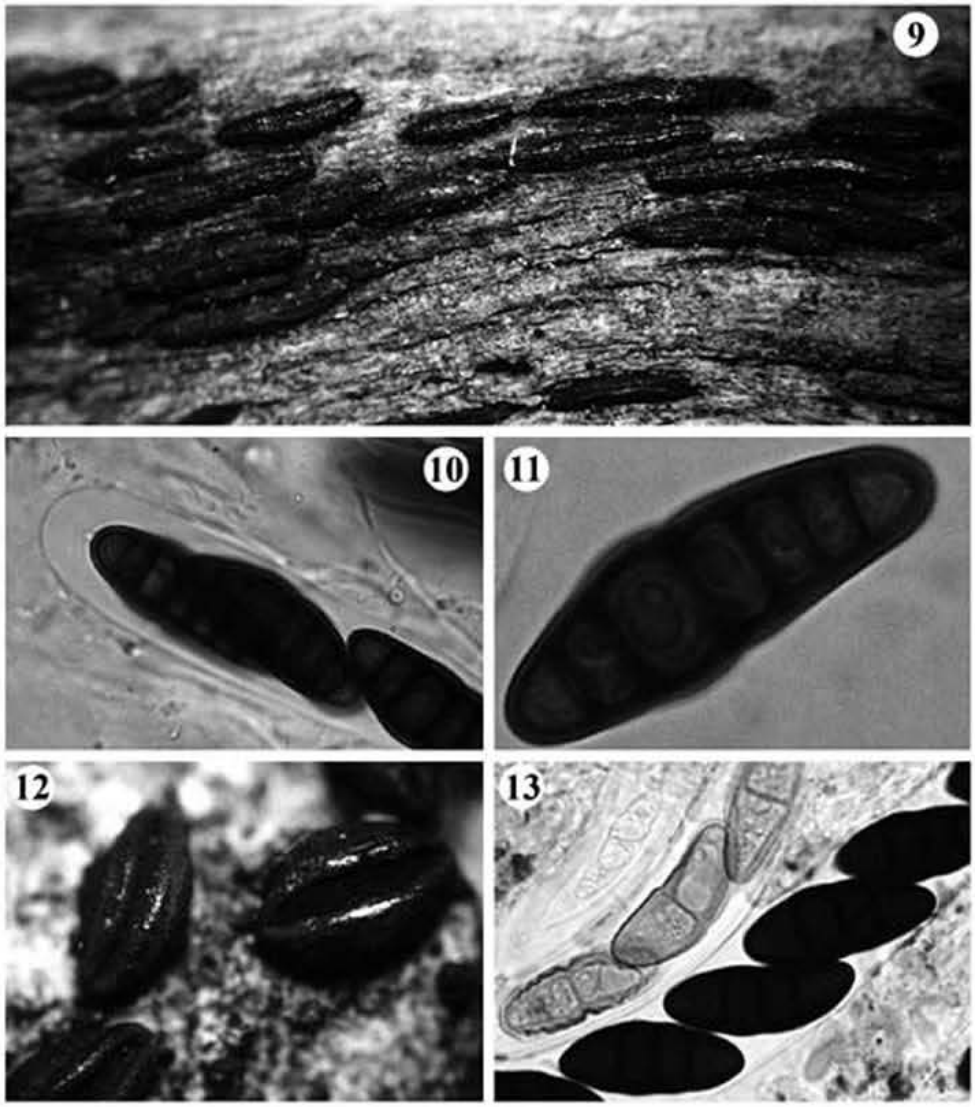
OBSERVATIONS: The Sonoran collection fits with *G. praelonga*, as described by Barr (1990) based on specimens from Italy and the United States of America, but ascus size was larger (60-120 × 15-25 µm) in those countries. Barr (1990) recorded high morphological variability for the macro- and microscopic characters of this species, resulting in the same species having been cited several times as a new species and thus increasing its list of synonyms. Kirk et al. (2001) recognized two species for the genus: *G. praelonga* and *G. smilacis* (Schwein. : Fr.) Underw. & Earle; both taxa were studied by Barr (1990).

***Hysterium insidens*** Schwein., Trans. Amer. Phil. Soc., N.S. 4(2): 244. 1832

Hysterothecia 0.7-4.8 mm long, subellipsoidal with tapered bases, linear and nearly parallel, erumpent to superficial; central part with straight longitudinal opening, dark with opaque to bright tones, external surface smooth, carbon-like appearance. Asci 155-195 × 18-22 µm, cylindrical-clavate, bitunicate, base ending in a short stipe, apical pore inamyloid. Ascospores 32-45 × 11-14 µm, elliptical to subfusoid, brown-olive tone, with 6-8 septi, central part of apical region a little wider than the rest, irregularly biseriate in ascus.

SPECIMENS STUDIED: LOCALITY 3, *leg.* F. Méndez & S. Gómez, 25.VIII.2005, solitary or gregarious, on the bark of branches and fallen trunks in oak forest clearings, *CESUES 5965, Chacón-6627( XAL).*

OBSERVATIONS: The Mexican collection fits with descriptions of Ellis & Everhart (1892), Dennis (1978), and Sivanesan (1984), who analyzed specimens from Africa, North America, and Europe, but ascus size was different. Ellis & Everhart (1892) observed asci measuring 75 × 15 µm, Dennis (1978): 90 × 17 µm, and




**Figs. 9-13.** Ascomycetes of Sonora, Mexico. 9-11: *Hysterium insidens*, 9: hysterothecia, 10: apical part of ascus with ascospores, 11: ascospore. 12-13: *Hysterium truncatulum*, 12: hysterothecia, 13: partial section of asci with ascospores.

Sivanesan (1984): 120 × 10-18 µm. As no further differences were noted, our specimens were identified as *H. insidens*.

***Hysterium truncatulum*** Cooke & Peck, in Cooke, Bull. Buffalo Soc. Nat. Sci. 3: 33. 1875

Hysterothecia 1.5-2 × 0.8-2.5 mm, erumpent, elongate to elliptical-fusoid, with obtuse, carbon-like tips, center dotted with longitudinal opening when closed,

dome-like appearance when open, external surface dark with radially striated margins and speckled with reddish dust. In open specimens, the hymenium is orangy red. Asci 220-345 × 15-20 $\mu$m, cylindrical-clavate, bitunicate, base with a short stipe, inamyloid. Ascospores 31-43 × 10-13 µm, cylindrical-ellipsoid with tips rounded to occasionally subapiculate, dark brown to reddish brown, triseptate, some with central cells darker and more compact at central septum level, uniseriate in the ascus.

SPECIMENS STUDIED: LOCALITY 6, *leg.* F. Méndez & S. Gómez, 27.VIII.2005, on the bark of branches and fallen trunks in subtropical scrub, *CESUES 6334, Chacón-5692 (XAL).*

OBSERVATIONS: The Sonoran collection agrees with the descriptions of Saccardo (1883) and Ellis & Everhart (1892). The distinguishing characteristics for this taxon are its dark brown, tri-septate ascospores, some with dark cells in the center and light ones at the tips. Guzmán (1983) recorded the first *Hysteriales* for Mexico: *Hysterium angustatum* Pers. for the Mexican state of Quintana Roo.

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MYCOTAXON

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# Lepiotaceous fungi in California, U.S.A. 6. *Lepiota castanescens*

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**Abstract**—*Lepiota castanescens* is a red-staining species in the *Leucoagaricus/Leucocoprinus* clade of the *Agaricaceae*. It is very close to the European *Leucoagaricus croceovelutinus* and the Australian *Lepiota haemorrhagica*. Basidiocarps stain red with ammonia, KOH and age, spores have an apical papilla, cheilocystidia are lageniform to clavate with apical excrescence; and the pileus covering outside the central patch is made up of repent hyphae of different length and diameter. The type collection and modern collections are described.

**Key words**—biodiversity, DNA sequence-based phylogeny, morphology, western North America

## Introduction

The mycoflora of California is rich in lepiotaceous fungi, the white-spored agaricoid members of the *Agaricaceae* (Sundberg 1967, Vellinga 2004a, b). In particular the *Leucoagaricus/Leucocoprinus* clade is represented by many species (Vellinga 2004a, b). One of these, a small but striking species whose basidiocarps turn brick to dark red with age and stain orange-red with ammonia, is identified as *L. castanescens*. The European species *La. croceovelutinus* (Bon & Boiffard) Bon & Boiffard, and the Australian species *Lepiota haemorrhagica* Cleland are its sister taxa in sequence analyses (Vellinga 2004a), and together they form a small and distinct clade, which differs from species such as *La. brunnescens* (Peck) Bon, *La. badhamii* (Berk. & Broome) Singer, and *La. americanus* (Peck) Vellinga (Vellinga 2004a, b) in the staining reaction. The basidiocarps of those species also change color when the cells are damaged, but they ultimately stain black(ish) and have a green reaction in ammonia and KOH.

Here we describe some features of the type collection, give a modern description of the species, and discuss the similarities and differences with its close relatives.

## Material and methods

Terminology for descriptive terms follows Vellinga (2001). Munsell (1976) indicated by Mu. in front of the code, and Ridgway (1912) (colour names are in " ") have been used to standardize colours.

The notation [90,6,6] indicates that measurements were made on 90 spores in six samples from six collections; avl stands for average length, avw for average width, Q for quotient of length and width and avQ for average quotient. The abbreviation *L.* is used for *Lepiota* and *La.* for *Leucoagaricus*. Herbarium abbreviations are according to Holmgren & Holmgren (1998).

Standard molecular methods were applied (e.g. Vellinga et al. 2003); the primer pair ITS1F and ITS4 were used both for PCR and sequencing (Gardes & Bruns 1993), and the phylogenetic analyses were performed with PAUP* version 4.0 (Swofford 2002). All nrITS sequences have been deposited in GenBank; accession numbers are listed with the collections.

## Taxonomic part

***Lepiota castanescens*** Murrill, Mycologia 4: 234. 1912. **Figs 1-4, 6, 7**

DESCRIPTION OF TYPE COLLECTION (Murrill 397 (NY)) (figs 1, 6):

Murrill (1912): "Pileus small, thin, convex to subexpanded, prominently umbonate, 2-3 cm. broad; surface dry, densely appressed-fibrillose, white to rose-colored, glabrous and darker-red on the umbo, the entire surface changing to castaneous on drying; lamellae free, crowded, narrow, plane, white, becoming fumosous on drying; spores ellipsoid, smooth, pointed, strictly hyaline, 7-8 × 3-4 μ; stipe tapering upward, slender, slightly fibrillose, hollow, about 6 cm. long and 2-5 mm. thick, white or rose-tinted, changing to castaneous on drying; annulus superior, fixed, ample, persistent, white, changing to castaneous on drying."

BASIDIOSPORES [20,1,1] in side-view 7.1-8.4 × 4.1-4.7 μm, avl × avw = 7.6 × 4.5 μm, Q = 1.55-1.86, avQ = 1.7, oblong-amygdaloid often with small apical papilla, in frontal view ovoid with or without papilla, congophilous, slowly colouring red-brown in Melzer's reagent, metachromatic in Cresyl Blue, uni-guttulate, slightly thick-walled. BASIDIA 4-spored. Lamella edge sterile; CHEILOCYSTIDIA hard to revive, 23-35 × 9-15 μm, clavate, with 7-25 × 5 μm moniliform to cylindrical excrescence, with brownish content in ammonia. PILEUS COVERING structure hard to discern, with chains of isodiametrical cells and elongate hyphae present; pigment brown and intracellular. CLAMP CONNECTIONS not observed.

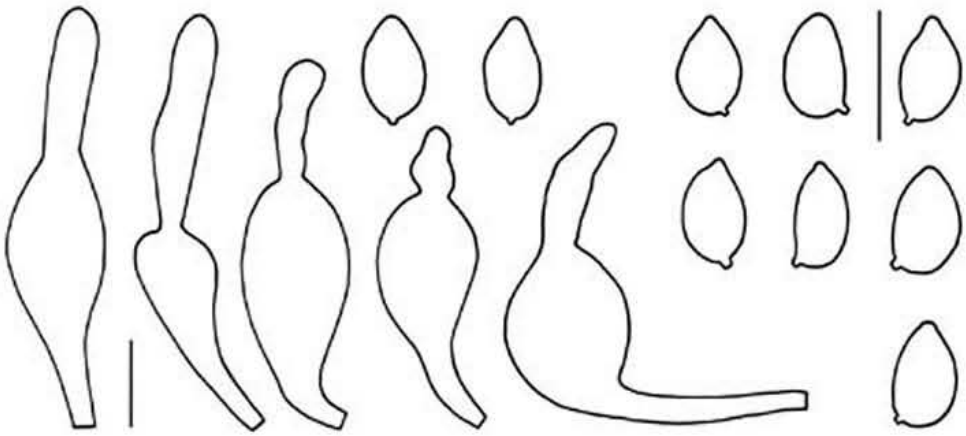

Fig. 1. *Lepiota castanescens* – spores and cheilocystidia from type collection. Scale bars 10 µm.

DESCRIPTION OF MODERN MATERIAL (figs 2-4, 6):

CHARACTERISTICS—Basidiocarps slender, with relatively small pileus, whitish at first, but discolouring orange-red to red in all parts when touched and with age; lamellae first yellow then orange with ammonia.

PILEUS 8-45(-50) mm, when young truncate-conical or hemispherical, expanding to plano-convex, applanate and finally plano-concave, most often with low, broad umbo, sometimes set in shallow depression, rarely without umbo, smooth or velvety-felted to plush-like tomentose at centre, around centre with radially arranged more or less coarse fibrillose or cobwebby scales, when young whitish, but soon discolouring to dark orange-brown, red-brown to almost black at centre (Mu. 2.5 YR 3/4, 2.5 YR 3-4/4; "Vandyke brown", "liver brown" to "burnt umber") and at fibrils (Mu. 2.5 YR 6/6-8, 2.5 YR 4/6; "Verona brown", "walnut brown", "hay's russet" to "liver brown"); background white at first, changing to orange with rain, age, or bruising (Mu. 2.5 YR 4/8; "carrot red"); margin fibrillose when young, exceeding lamellae. LAMELLAE, L = around 50, l = 1(-3), moderately crowded, free, rounded near stipe and slightly ventricose, whitish at first, but soon pale pink ("light ochraceous-buff", "apricot buff") and flushed orange-red in places to completely red ("orange cinnamon" to "kaiser brown"); edge conspicuously set with cystidia, white at first, changing to red when touched, and with age. STIPE 25-80(-220) × 1-7.5 mm, cylindrical but widening downwards, especially in lower 1 cm, up to 1.5 times width at half length, when young whitish, to slightly pinkish, innately lengthwise fibrillose, becoming red instantaneously when touched (Mu. 10 R 5-4/8), and orange-red to red with age and other damage (rain e.g.), concolorous with pileus, hollow. ANNULUS funnel-shaped, or with a small ascending, rarely descending, cuff with

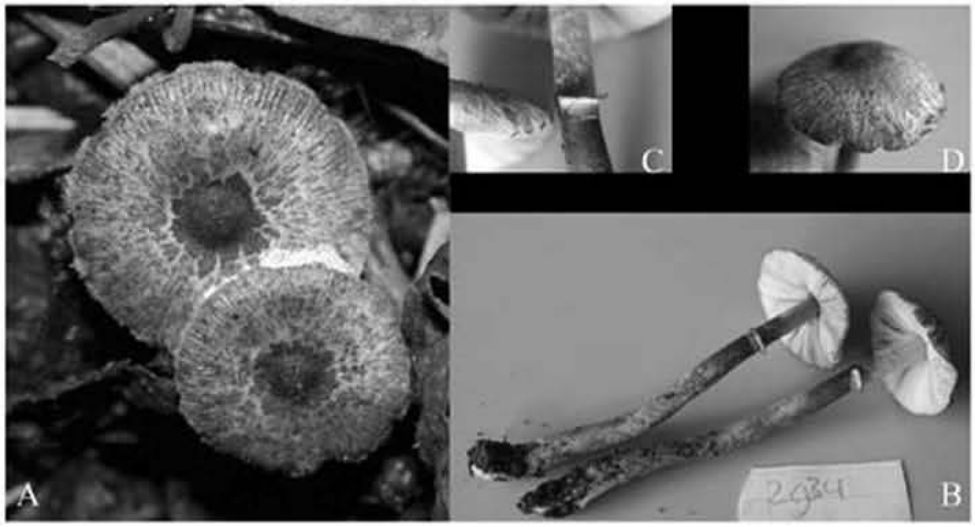


Fig. 2. *Lepiota castanescens* – A. collection ecv3077; B, C and D. collection ecv2934. All photos by John Lennie.

a (small) flaring part, white at first, changing to red to red-brown, especially at outside, and on edge and there almost black; inner part staying white for a longer time. CONTEXT very thin in pileus, whitish, not or changing to carrot orange to light scarlet ("light ochraceous-salmon" to "vinaceous-rufous") when cut; in stipe whitish, reddish when cut. SMELL none. TASTE not recorded. SPORE PRINT color white. CHEMICAL REACTIONS: Ammonia on lamellae first yellow, then orangey red.

BASIDIOSPORES [105,7,7] in side view 5.9-8.3 × 3.5-4.7 µm, avl × avw = 6.8-7.3 × 4.0-4.2 µm, Q = 1.5-2.1, avQ = 1.66-1.78, amygdaliform with small apical papilla and conspicuous hilar appendage, in frontal view oblong with apical papilla, slightly thick-walled, often with one, more rarely with two, guttules, dextrinoid, congophilous, and metachromatic in Cresyl blue, without germ pore. BASIDIA 14-26 × 6-9 µm, the majority 4-spored, a few 2-spored, close to lamella edge with rusty brownish contents. Lamella edge sterile; CHEILOCYSTIDIA 17-62 (in total) × 7-19 µm, clavate, narrowly lageniform, lageniform to utriform, often narrowly clavate gradually or abruptly changing into 5-30 × 3-8 µm cylindrical, moniliform to capitate, apical excrescence, with reddish rusty brown contents in ammonia. PLEUROCYSTIDIA absent. PILEUS COVERING at umbo a dense layer of iso-diametrical cells, 8-30 × 7-30 µm giving rise to occasional elongate cylindrical or slightly fusiform cells, 25-45 × 7-13 µm; around umbo a cutis of strands of repent hyphae with uplifted terminal elements, more sparsely covering pileus surface and with thinner hyphae towards margin; terminal elements cylindrical to slightly inflated, with rounded apex, 27-90 × 8-12.5 µm; all elements of pileus covering with dark red-brown walls, rarely incrusted-

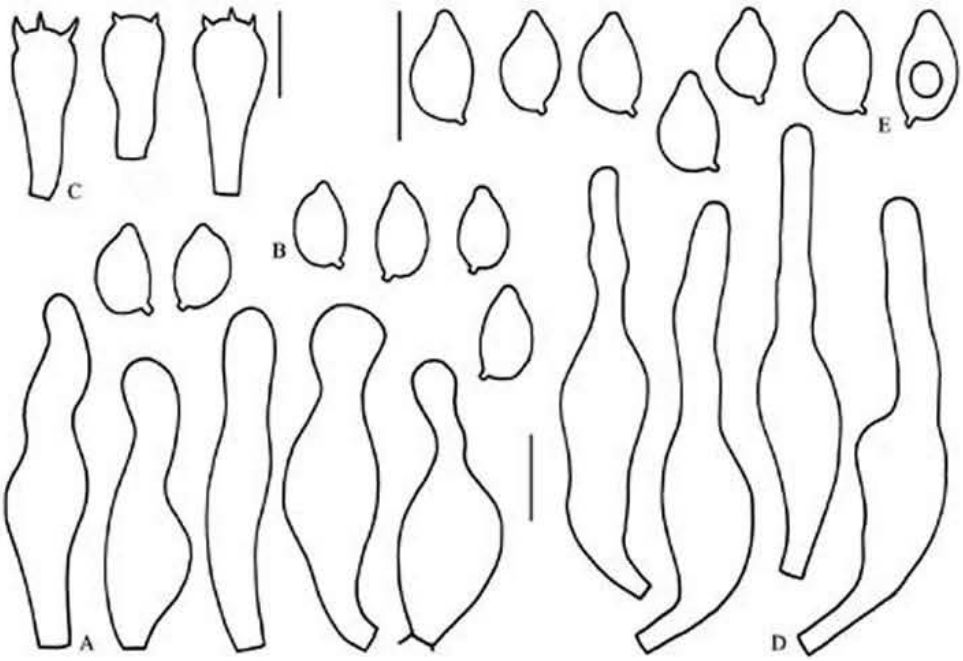


Fig. 3. *Lepiota castanescens*, microscopical characters – spores (B and E), basidia (C) and cheilocystidia (A and D).
A-C from collection ecv3424; D and E from collection R. Pastorino.
Scale bars 10 µm.

pigmented, and some cells with intracellular pigment. STIPE COVERING a cutis of cylindrical cells, 3-5 µm wide, with brown to orange-brown pigments, some with excrescences. CLAMP CONNECTIONS absent.

HABITAT & DISTRIBUTION – Gregarious in small groups, rarely solitary, terrestrial and saprotrophic, in various forest types, e.g. Monterey Cypress (*Cupressus macrocarpa*) groves, in mixed *Alnus-Picea sitchensis* forest, or mixed conifer coastal forest. Known from Washington, and coastal California, reported from Humboldt Co. in the north to San Mateo Co., south of San Francisco.

COLLECTIONS EXAMINED – U.S.A., Washington, around Seattle, 20 Oct.–1 Nov. 1911, W.A. Murrill 397 (Holotype, NY); King Co., Seattle, Fauntleroy Park, 27 Oct. 2003, J.M. Birkebaek 80 (WTU; Genbank nrITS EU166350). California, Humboldt Co., Patrick's Point S.P., 23 Sept. 1966, W.J. Sundberg 777 (SFSU; as *L. rubrofolia*); Patrick's Point S.P. near Mussel Rock, 23 Oct. 2003, E.C. Vellinga 3076 and 3077 (UC); Prairie Creek S.P., 23 Sept. 1966, W.J. Sundberg 790 (SFSU; as *L. rubrofolia*); ibidem, 8 Oct. 1966, W.J. Sundberg 881 (SFSU; as *L. rubrofolia*); Prairie Creek Redwood NP, near Wolf Creek, 26 Oct. 2003, E.C. Vellinga 3115; Mendocino Co., Jackson State Demonstration Forest, 22 Nov. 2003, E.C. Vellinga 3131 (UC; Genbank nrITS EU166351); Marin Co., Audubon Canyon Ranch, Picher Canyon, 6 Dec. 1977, Calhoun 77-394 (SFSU; as *L. rubrofolia*); ibidem, Galloway Canyon, 13 Nov. 1980, Calhoun 80-168 (SFSU; as *L. rubrofolia*); Muir Woods, 20 Nov. 1966, Madden 539 (SFSU; as *L. rubrofolia*); Point Reyes, near Bolinas,

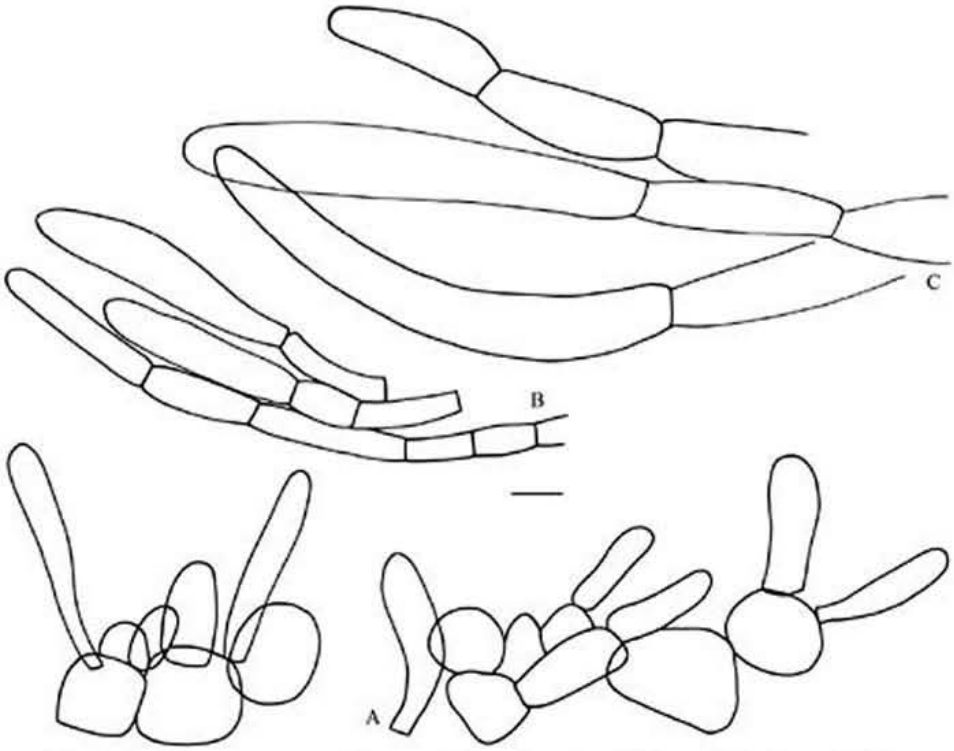


Fig. 4. *Lepiota castanescens*, pileus coverings – A. centre of pileus (collection ecv3142); B and C. radiating fibrils around umbo (B from collection ecv3424; C from collection R. Pastorino). Scale bar 10 µm.

25 Nov. 2003, R.L. Pastorino (UC; Genbank nrITS EU166352); Contra Costa Co., Tilden regional park, 23 Nov. 2001, E.C. Vellinga 2739 (UC); San Mateo Co., San Francisco Watershed, 10 Dec. 1999, E.C. Vellinga 2398 (UC); ibidem, 8 Dec. 2000, E.C. Vellinga 2596 (UC; Genbank nrITS AF482860, as '*Leucoagaricus* sp.'); ibidem, 13 Dec. 2002, E.C. Vellinga 2934 (UC); ibidem, 5 Dec. 2003, E.C. Vellinga 3142; ibidem, 2 Dec. 2005, E.C. Vellinga 3424 (UC).

ADDITIONAL COLLECTIONS EXAMINED

*Leucoagaricus croceovelutinus* – The Netherlands, prov. Noord-Holland, Aerdenhout, Naaldenveld, 27 Oct. 1977, E. Kits van Waveren (L); Amsterdamse Waterleidingduinen, Vogelenzang, 13 Oct. 1979, C. Bas 7561A (L); ibidem, 18 Sept. 1982, E.C. Vellinga 473 (L); prov. Zuid-Holland, isl. of Voorne, Westvoorne, Quackjeswater, 7 Oct. 1980, J. Schreurs 531 (L); prov. Limburg, Bemelen, 9 Oct. 1991, E.C. Vellinga 1779 (L); Cadier en Keer, 6 Oct. 2004, H.A. Huijser (herb. Huijser; Genbank nrITS EU166349); Elsloo-Geulle, Bunderbos, 19 Sept. 1998, E.C. Vellinga 2229 and 2243 (L) (Genbank nrITS AF482862, LSU AF482889).

*Lepiota haemorrhagica* – Australia, Victoria, 52 km No of Orbost on the Bonang Road, Martins Creek, alt. 320 m, 17 May 2000, K.R. Thiele 2652 (MEL) (Genbank nrITS AY176374, LSU AY176375).

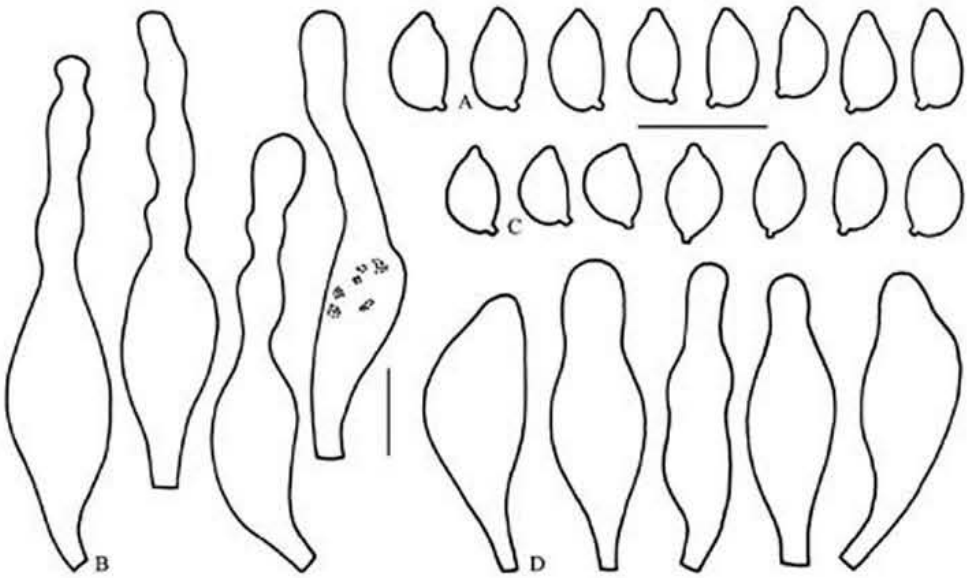


Fig. 5A, B. *Leucoagaricus croceovelutinus* – A. spores; B. cheilocystidia (collection H.A. Huijser, 6 Oct. 2004). Scale bars 10 µm.

Fig. 5C, D. *Lepiota haemorrhagica* – C. spores; D. cheilocystidia (collection K.R. Thiele 2652). Scale bars 10 µm.

COMMENTS – Murrill's description and the type collection of *L. castanescens* fit very well what we have been calling either "*L. rubrofolia*" (Sundberg 1967), "*L. carmineobasidia*" (Vellinga 2004a) (both names are manuscript names by Sundberg (1967)) or more recently "American representatives of *La. croceovelutinus*". The type collection was studied previously by Smith (1966), whose spore sizes are a little smaller than what we found.

*Lepiota castanescens* is easy to recognize because of its reddish colours and the distinct central patch on the pileus surrounded by fibrillose scales. Microscopically the combination of spore shape, cystidia and structure of the pileus covering is characteristic.

Only in very early stages are the basidiocarps predominantly white, but the slightest touch changes them into reddish mushrooms. The specimens display a huge range in size, from minute with a slender 1 mm wide stipe and an 8 mm wide pileus to robust with a long and 7.5 mm wide stipe and an up to 50 mm wide pileus. Microscopically there is also huge variation, especially in size and shape of the cheilocystidia (fig. 3).

The structure of the pileus covering at the umbo differs strikingly from that of the squames surrounding the umbo. The same is true for the other two species in the group. The descriptions of the pileus covering of *La. croceovelutinus* vary from author to author (compare for instance the original description by Bon &

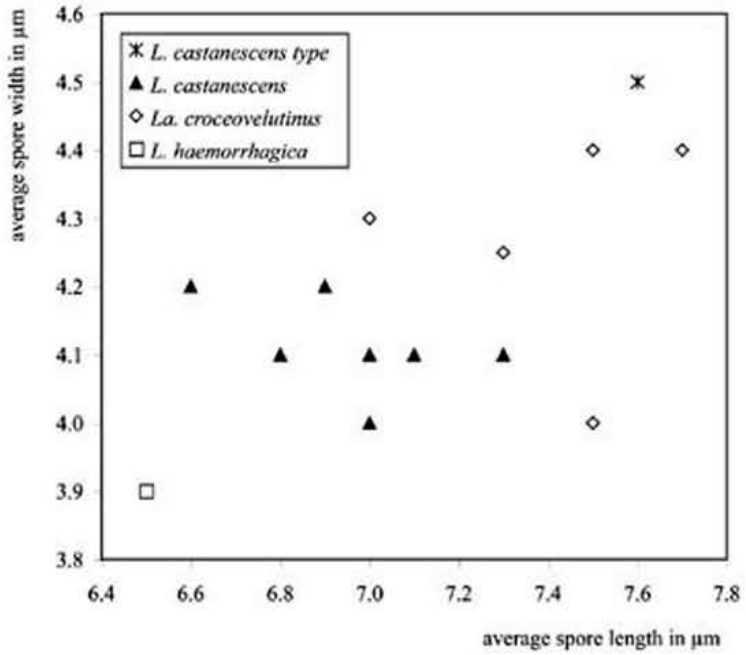


Fig. 6. Scatter diagram of average spore sizes for collections of *L. castanescens* including the type, *La. croceovelutinus*, and *L. haemorrhagica*.

Boiffard (1972) and those by Candusso & Lanzoni (1990), Kelderman (1994), and Reid (1990)). This discrepancy is caused by the fact that the structure of the umbo and that of the fibrillose covering surrounding it are very different. In *L. castanescens* the central calotte is made up of globose to iso-diametrical cells with emerging cylindrical elements (fig. 4A), whereas the scales are composed of repent radially arranged hyphae (fig. 4B, C).

The amygdaloid spore shape with the apical papilla is not unique in the *Agaricaceae*: *Leucoagaricus pepinus* Heinem. with spores (8.5-)10.2-14.5(-17) × (5.5-)6.5-8.2(-8.5) µm (Heinemann 1973) and *Leucocoprinus pepinosporus* Heinem. with spores 11.2-13.4 × 7.4-8.1 µm (Heinemann 1977) are other examples, but both lack the striking colour changes of *La. croceovelutinus*, and have much bigger spores with a bigger papilla.

Several species in *Leucoagaricus* section *Piloselli* (Kühner ex) Singer possess similarly shaped cheilocystidia (e.g. *L. fuliginescens* Murrill and *La. badhamii*), but these species differ in their reaction with ammonia.

*Lepiota castanescens* forms together with *La. croceovelutinus* and *L. haemorrhagica* a distinct group in the *Leucoagaricus/Leucocoprinus* clade, apparently not closely related to species in *Leucoagaricus* sect. *Piloselli* such as *La. badhamii* with a green reaction in ammonia and spore without a germ pore

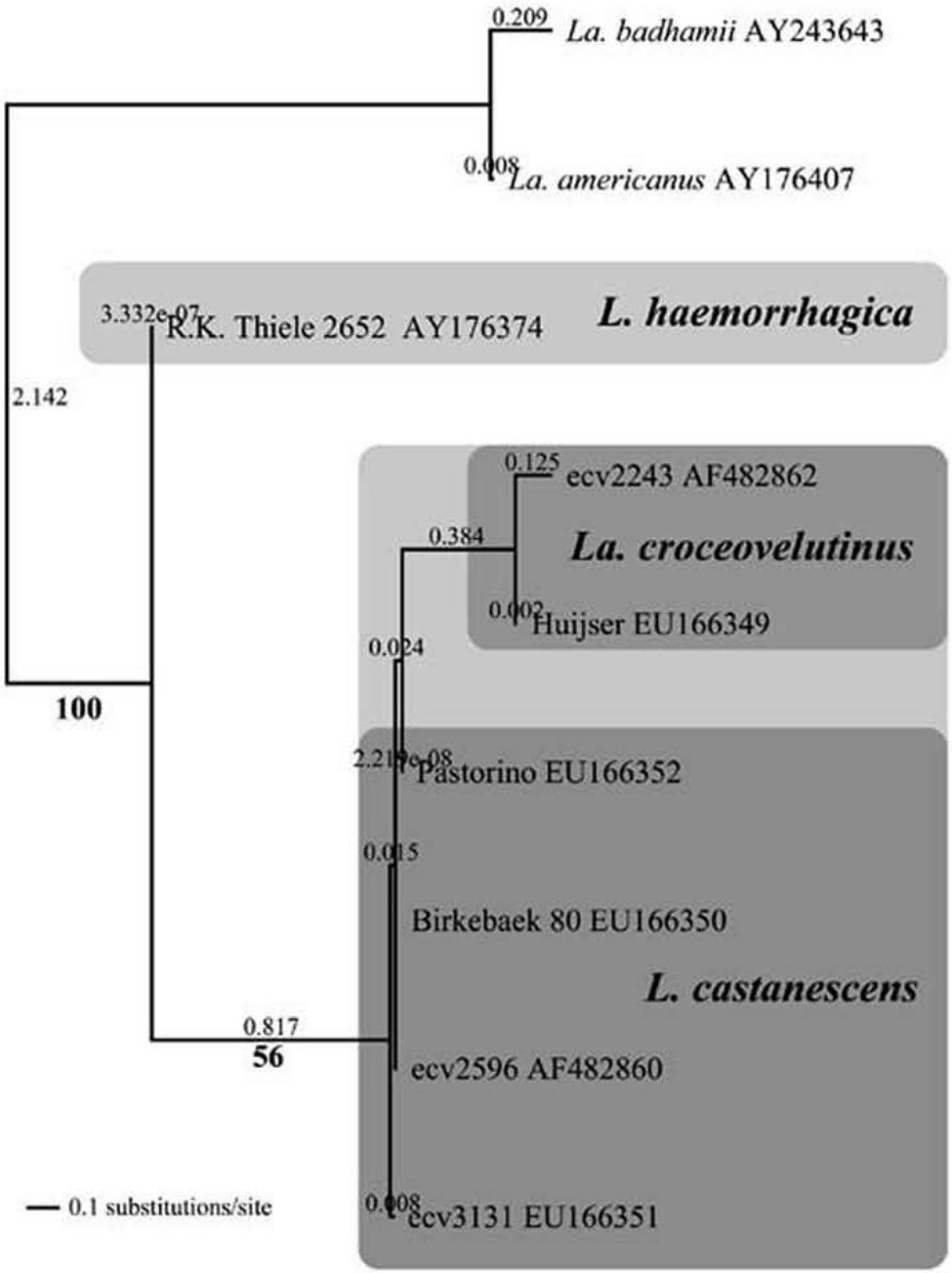


Fig. 7. Phylogenetic relationships among reddening species of *Leucoagaricus* inferred from Maximum Likelihood analyses of a dataset of nrITS sequences; 60 characters were informative; the HKY85 variant was used as model. Bootstrap values (in bold) are based on 100 replicates. *Leucoagaricus badhamii* and *La. americanus* were used as outgroup. All sequences have been deposited in Genbank.

and *La. americanus* and allies, also with a green reaction with ammonia, but with spores with a germ pore.

*Leucoagaricus croceovelutinus* differs in nrITS sequences, and in general has more robust and less slender basidiocarps than *L. castanescens*. The long slender stipes of the latter might be caused by environmental factors like a thick litter layer. Besides the differences in nrITS, also gene sequences of RPB2 show differences in base pair composition between the two North American and European collections sequenced (data not shown).

*Lepiota haemorrhagica* from Australia has smaller cheilocystidia (fig. 5C, D) than *La. croceovelutinus* (fig. 5A, B) and *L. castanescens* (fig. 3D), but is in general appearance very similar to both (Cleland 1931, 1934; Grgurinovic 1997). The average spore size for the one collection we studied is given in fig. 6, but Grgurinovic (1997) listed bigger and wider spores for the type collection: 6.6-9.6 × 4.0-5.8 µm, on average 7.4 × 4.9 µm. The differences in nrITS sequences are quite small. The three taxa differ mostly at the molecular level and in geography; the morphological differences are small and subtle. Despite an extensive literature review we have not been able to find other species with this set of characters. It is remarkable that the closest relative of the Northern Hemisphere species is found in Australia.

We refrain from making the combination in *Leucoagaricus* for *L. castanescens* and *L. haemorrhagica* as the taxonomy of the *Leucoagaricus/Leucocoprinus* clade is still in flux (see Vellinga 2004b).

The actual distribution areas of all three taxa are not known. *Lepiota castanescens* is known from Washington and coastal, northern California. It has not been found in the eastern states of the U.S.A. The European taxon is known from western Europe (from Denmark south to Italy and Spain (e.g. Heilmann-Clausen 1992, Lange 1995, Bon 1993, Chiusa 1999, Candusso & Lanzoni 1990, Consiglio et al. 2004, Bolets Catalunya 1998), and from the United Kingdom eastwards to Hungary (e.g. Reid 1990, Vellinga 2001, Babos 1995). *Lepiota haemorrhagica* is known from South Australia and Victoria (Cleland 1931, 1934; Grgurinovic 1997, Shepherd & Totterdell 1988).

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# Taxonomy of the fungus commonly known as *Stropharia aurantiaca*, with new combinations in *Leratiomyces*

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**Abstract** — The taxonomy and nomenclature of the fungus commonly referred to as *Stropharia aurantiaca* was investigated. Molecular analysis of the ribosomal RNA large subunit gene confirmed the exclusion of the species from *Stropharia*. The results from further molecular and morphological comparison with type species indicated that '*S. aurantiaca*' is congeneric with a number of other taxa currently placed in *Leratiomyces*, *Stropharia* and *Weraroa*. These taxa are transferred to *Leratiomyces* and an emended diagnosis and brief discussion of this genus are provided.

**Key words** — LSU, strophariod, woodchip, sequestrate

## Introduction

The suitability of woodchip mulch as a substratum for the colonisation of fungi has become increasingly evident in recent years, with an ever-growing number of species being recorded from it from many parts of the world. During the past twenty years or so the mulching of garden beds with woodchips has become a routine practice in Britain (Shaw et al. 2004), providing a specialised habitat that is considered by Shaw & Kibby (2001) to be unique and not otherwise found in nature. Either as mulch or in composting piles, woodchip proves surprisingly ideal for a wide range of fungi from various groups, most conspicuously amongst the larger fungi, including common species found more usually in other habitats, and otherwise scarce species which can fruit in abundance. In Britain, these are exemplified by the bird's-nest fungi *Cyathus olla*, *C. striatus* and

*Crucibulum laeve*, and by agarics such as *Volvariella gloiocephala*, *V. hypopithys*, and *Macrocystidia cucumis*. Some woodchip fungi have even proved to be first recorded from this substratum, notably *Melanoleuca verrucipes* (Shaw & Kibby 2001) and *Agrocybe putaminum* (Henrici 2001) in Britain, while others have proved to be undescribed and of unknown origin. Perhaps the most recent example of this is *Agrocybe rivulosa*, recently described from woodchips in the Netherlands (Nauta 2003). This species was first recorded in Britain from Staffordshire in 2004 and is now spreading rapidly in England with at least 8 collections to date (Henrici 2006a,b). *Psilocybe cyanescens* was perhaps the first of the introduced woodchip fungi to be recorded in Britain, being first described from the Royal Botanic Gardens, Kew, in 1911 (Dennis & Wakefield 1946). An unpublished compilation of woodchip fungi on file at Kew now totals at least 250 species. Their origin, ecology and mode of dispersal are all little understood aspects in need of further study.

One of the commonest and most distinctive of the woodchip fungi has been known as *Stropharia aurantiaca*, a species readily recognised by its orange cap and dark spores, often fruiting in large numbers over extensive areas of woodchip mulch (e.g. Pegler & Legon 1998, Shaw & Kibby 2001). Ironically, it has become clear that this distinctive and widespread species has been consistently misidentified and the name misapplied, and the search for an appropriate name has proved difficult. Guzmán (1983) noted this confusion, and the problem was recently highlighted by Fortey (2004), who has carefully and clearly summarised the situation. *Stropharia aurantiaca*, which unfortunately lacks type material and is represented only by a painting (reproduced in Fortey 2004), has a similar orange cap but is a slender agaric as illustrated and described by Cooke (1887, as *Agaricus squamosus* f. *aurantiacus*). This may also occur on woodchips but can be identified with *S. thrausta* (= *S. squamosa* var. *thrausta*), a scarce taxon which, based on morphology and DNA analysis, has been shown by Jahnke (1984) to be no more than a colour form of *S. squamosa*. This taxon, morphologically identical with *S. squamosa*, lacks chrysocystidia whereas the species in question has abundant chrysocystidia and is quite distinct (Fortey 2004). It continues to be reported worldwide and is now known throughout much of Europe (Noordeloos 1999), North America (Arora 1986), Australia (Daams 1991) and New Zealand (Taylor 1981). In Britain, it was first recognised in 1957, being reported from sawdust in Somerset by Orton (1960) who noted the abundant chrysocystidia. Reid (1966) described and illustrated a subsequent collection from Surrey in November 1957. It is now a regular and often abundant coloniser of woodchip mulch, occasionally also found away from woodchip in natural habitats. However, its mode of dispersal and colonisation are, as for other wood-chip fungi, little understood. At the Royal Horticultural Society's garden at Wisley in Surrey, DNA analysis of mulch and

Table 1. Specimen details and GenBank numbers for sequences generated in this study.

| Species | Locality | Collector | Voucher | GenBank |
|---|---|---|---|---|
| *Stropharia aurantiaca* | UK, Surrey, Wisley, Royal Horticultural Society Garden | Bridge & Prior, 2004 | KC1700 (culture) | AM747624 |
| *Stropharia percevalii* | UK, Surrey, Leatherhead | Shaw, 2000 | K(M)77890 | AM747625 |
| *Weraroa cucullata* | California, Sierra County, Chapman Creek | Saylor, Cazares & Castellano, 1987 | PDD 58490 (Trappe 9507) | EU019232 |
| *Weraroa erythrocephala* | New Zealand, Waipori Valley | Beever, Pennycook & Johnston, 2000 | PDD 71777 (Beever 1879) | EU019233 |
| *Weraroa novae-zelandiae* | New Zealand, Ketetahi | Lebel & Camacho, 1995 | PDD 65096 (Trappe 15563) | EU019234 |
| *Weraroa novae-zelandiae* | New Zealand, Waitakere Range | Beever , 1997 | PDD 67194 (Beever 1547) | EU019235 |
| *Leratiomyces atrovirens* | New Caledonia, Monts des Koghis | Johnston & Beever, 2003 | PDD 88863 (Beever 2202) | EU183546 |

soil showed somewhat inconsistent results but in general found that the fungus is present in soil that may then provide a reservoir for colonisation of the mulch (Bridge & Prior 2007).

The origin of '*Stropharia aurantiaca*' (the woodchip fungus) has been unclear but was suggested by Watling & Gregory (1987) to be Australia, where they consider the native *Psilocybe ceres* to be identical with it. This conclusion was also followed by Daams (1991), and by Grgurinovic (1997). *Psilocybe ceres* is described as slender, glabrous, with elongate stipe, and to occur on soil and rarely on dung (Cleland 1934; Grgurinovic 1997, as *S. aurantiaca*). Pegler (1965), who examined the holotype of *P. ceres*, lodged in K, illustrated the spores and reported the presence of a pseudorhiza (e.g., rhizomorphs) at the stipe base. He also reported the presence of pleurocystidia with refractive inclusions, hence showing that, although the species is described in the protologue as slender, it cannot be identified with *S. thrausta*. The holotype collection comprises sectioned, rather fragmentary fruitbodies not suitable for DNA study. Although given in the protologue as 'testacea', a descriptive note with the specimen reads: 'pileus rufus red, gills watery dirty grey with black tinge turning darker, numerous; stem fibrillose, striate, concolorous towards the base'. Re-examination of this collection by BMS reveals whitish basal rhizoids as

well as chrysocystidia and spores identical to those of '*S. aurantiaca*'. Although cheilocystidia could not be recovered, the characters noted, as well as the lower stipe being concolorous with the cap, fully agree with '*S. aurantiaca*' and there seems every reason to accept it as conspecific with that fungus and therefore to provide the earliest name for it. Although '*S. aurantiaca*' has a clear preference for woodchips, a few collections in more natural habitat away from woodchip have been made in England.

A search of the literature has revealed that a later synonym for the species in question is provided by *Nematoloma rubrococcineum* (Balletto 1967). This was described from France 'inter detrito permixta', and agrees well in all respects including spore characters and the possession of chrysocystidia. A coloured plate of the species was published by Nonis (1994) who recombined it as *Hypholoma rubrococcineum*, and another by Mazza (1994) who noted the name as a possible synonym of '*Stropharia aurantiaca*'.

The recent molecular study of the nuclear large ribosomal subunit gene (LSU) of a wide range of agarics by Moncalvo et al. (2002) recognised 117 clades and recovered '*S. aurantiaca*' (as *H. aurantiacum*) in a small clade with sequences from *S. magnivelaris, Leratiomyces similis* and *Weraroa erythrocephala*. This clade was placed in the strophariod clade group, but was well separated from both *Stropharia* (typified by *S. aeruginosa* (Curtis) Quél.) and *Hypholoma* (typified by *H. fasciculare* (Huds.) P. Kumm.). This indicates that *S. aurantiaca* cannot be placed in the genus *Hypholoma* nor in *Stropharia*, but may belong either in *Leratiomyces* as typified by *L. similis*, or in *Weraroa*. An appropriate genus for *Psilocybe ceres* therefore requires careful consideration based not only on basidiocarp morphology but, especially, on analysis of DNA sequences of this and related taxa. *Weraroa* Singer is a genus of sequestrate agarics popularly known as 'tobacco-pouch fungi'. They have dark spores and coloured pilei and are recognised as close relatives of *Stropharia* (Singer 1960). In Moncalvo et al. (2002), *W. erythrocephala* was recovered with '*H. aurantiacum*' and *L. similis*, and thus *Weraroa* may provide a generic name for '*S. aurantiaca*'. However, *W. virescens* was placed in a different section of their grouping, indicating that *Weraroa* is polyphyletic, and the type species, *W. novae-zelandiae*, was not included in their analysis. In the present study, we report LSU sequence data for some further species, including *W. novae-zelandiae*, and clarify their taxonomy.

## Methods

**DNA extraction and PCR**— The partial LSU sequence of '*S. aurantiaca*' was obtained from material collected at the Royal Horticultural Society Garden, Wisley, UK in 2005 and DNA was extracted as described in Bridge & Prior (2007).

A small section of dried stipe (approx. $2mm^3$) of *S. percevalii* (K(M)77890) was disrupted with a micro-homogeniser in a microcentrifuge tube in 300 µl of SDS containing buffer, and DNA was extracted according to Cenis (1992). PCR was undertaken with primers LR6 (Vilgalys & Hester 1990) and LR0R (Moncalvo et al. 1995) with Qiagen "Ready to go beads". PCR conditions were an initial heating to 94°C for 2 min, followed by 30 cycles of 94°C for 2min, 55°C for 1 min, 72°C for 1 min, followed by a final extension of 72°C for 10 min. PCR products for '*S. aurantiaca*' and *S. percevalii* were sequenced by a commercial sequencing service. *Weraroa* and *L. atrovirens* DNA was obtained from portions of recently dried herbarium material (Table 1). DNA was isolated and purified using DNeasy Plant Mini kit (Qiagen, USA). PCR was performed with primers LR5 (Vilgalys & Hester 1990) and LR0R with FastStart Taq (Roche, USA) supplemented with 0.4µg/µl of BSA. PCR conditions were an initial heating to 95°C for 4 min, followed by 19 cycles of 94° C for 1min, 55°C for 30 sec, 72°C for 1 min, followed by 14 cycles of 94°C for 30sec, 60°C for 30sec, 72°C for 1 min, followed by a final extension of 72°C for 15 min. Sequencing of the PCR products was performed with ABI PRISM BigDye Terminator Ready Reaction Kit V3.1 (Applied Biosystems, USA) and run on an Applied Biosystems ABI PRISM 3100 Avant Genetic Analyzer.

**Phylogenetic analysis**— Our LSU sequences for '*S. aurantiaca*' and *S. percevalii* were compared with representative sequences of the /psychedelia, /hypholoma, /stropharia, /pholiota, /magnivelaris and /psilocybe clades, (clades 106 and 109-113) as recovered by Moncalvo et al. (2002), a sequence for *S. squamosa* (AY207302), as well as sequences for *W. novae-zelandiae, W. erythrocephala* and *W. cucullata* obtained in this study. Sequences were aligned over 882bp (inc. gaps) in CLUSTALW and maximum likelihood trees were constructed in Phylip. Tree construction assumed a transition/transversion ratio of 2 and constant rate variation. A majority rule consensus tree was obtained and this was rooted with *Phlebia uda* (= *Mycoacia uda*).

## Results

Our analysis of the LSU sequences (Fig. 1) grouped the reference sequence into the 'genus level' clades described by Moncalvo et al. (2002). The LSU sequences did not contain sufficient variability to be reliably used for species comparisons as is demonstrated by the two sequences from *Weraroa novae-zelandiae* that appear separated within the /psychedelia clade although their sequences only differ by one transition and one transversion. At the generic level the LSU analysis confirms that the red *W. erythrocephala*, together with *W. cucullata*, are closely related to *Leratiomyces similis* and to '*S. aurantiaca*'. However, the type species, *W. novae-zelandiae* (G. Cunn.) Singer, belongs to

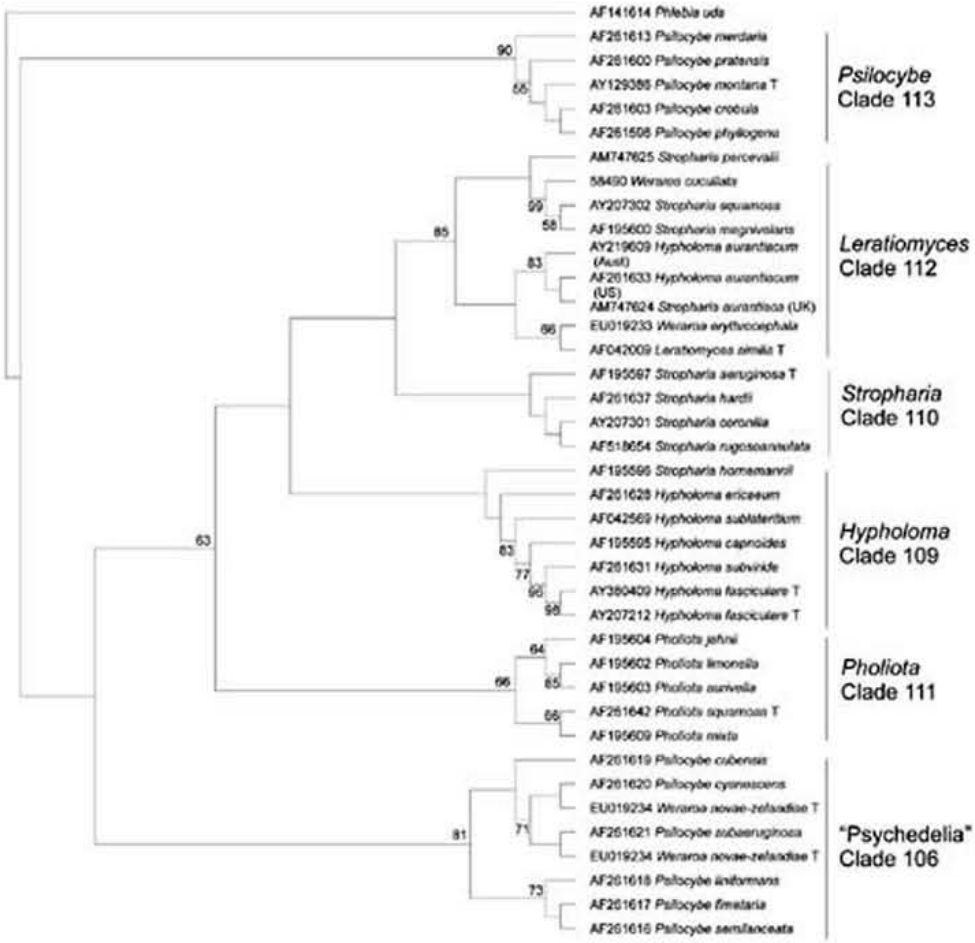


Figure 1. Consensus ML tree of LSU sequences from strophariod and related taxa. Clade numbers are as given in Moncalvo et al. (2002), numbers at branch points are bootstrap values where greater than 50. T after species indicates type species for genus.

a different clade nesting together with hallucinogenic psilocybin-containing species currently referred to *Psilocybe*, in the /psychedelia clade of Moncalvo et al. *Weraroa* therefore does not offer an appropriate genus for '*S. aurantiaca*'. It could provide an appropriate genus for the /psychedelia clade, but Redhead et al. (2007) propose that *Psilocybe* should be conserved with the circumscription of the /psychedelia clade with *P. semilanceata* as its conserved type. Our study also confirms the close relationship between '*S. aurantiaca*' and *Leratiomyces similis*, a sequestrate species described from New Caledonia. This is the type of *Leratiomyces* that, therefore, is an appropriate genus for '*S. aurantiaca*' and its allies. The sequestrate character of the type and other species so far referred to *Leratiomyces* in itself has little taxonomic value. The inclusion of sequestrate

and non-sequestrate species in the same genus is supported by the molecular evidence and has been done recently for other agaricoid fungi, for example in *Cortinarius* (Peintner et al. 2002), and in *Lactarius* and other *Russulaceae* as discussed by Eberhardt & Verbeken (2004).

*Hypholoma rubrococcineum* has also been referred to the later genus *Stropholoma* (Balletto 1989), typified by *Stropholoma squamosum*, an invalid name. *Stropholoma* was employed by Noordeloos (1999) as a subgenus of *Psilocybe*, to include non-fasciculate species with dry stem, spores longer than 10 µm, and with chrysocystidia either present or absent.

Combinations in *Leratiomyces* for *Psilocybe ceres* as well as other species shown by DNA sequence analysis to be closely related are proposed here. The rather complex nomenclature applied to these fungi is also given. Ironically, the misinterpretation of *Stropharia aurantiaca* has resulted in the most extensive synonymy for the scarcest of the species.

*Stropharia squamosa* was not included in the Moncalvo et al. (2002) study, but was considered by Walther et al. (2005). Their sequence (AY207302) was recovered within the *Leratiomyces* clade supporting the previously suggested close relationship between *S. squamosa* and '*S. aurantiaca*'. A second sequence available in GenBank also labeled as *S. squamosa* (AF261640) was substantially different from all of the sequences used in this study. It showed 97-99% homology with species of *Coprinus* and *Coprinopsis* and so was not considered further. There are no available LSU sequences for *S. thrausta* and repeated attempts failed to obtain a sequence from our material. We did obtain an ITS sequence from one specimen (K(M)83498) and this gave 90% or higher homology with ITS sequences from '*S. aurantiaca*' and related specimens (results not shown).

## Taxonomy

***Leratiomyces*** Bresinsky & Manfr. Binder, Z. Mykol. 64(1): 80 (1998)
- = *Le Ratia* Pat., Bull. Trimestriel Soc. Mycol. France 23: 52 (1907) (nom. inval., Art. 20.3)
- = *Le-Ratia* Pat. ex Sacc. & Trotter, Syll. fung. 21: 468 (1912) (nom. illegit., non *Le-Ratia* Broth. & Paris (1909), *Musci*)

= *Nematoloma* sect. *Stropholoma* Singer, Sydowia 2: 36 (1948)
- = *Stropholoma* (Singer) Balletto, Micol. Ital. 18: 36 (1989) (nom. inval., Art. 33.4)
- = *Psilocybe* subgen. *Stropholoma* (Singer) Noordel., Persoonia 16: 127 (1995)

**Type:** *L. similis* (Pat.) Bresinsky & Manfr. Binder, Z. Mykol. 64(1): 80 (1998)
= *Le Ratia similis* Pat., Bull. Trimestriel Soc. Mycol. France 23: 52 (1907)

EMENDED DIAGNOSIS — Basidiomata hymenogastroid to agaricoid and velate, pileate, stipitate or rarely almost sessile, usually with basal pseudorhiza, terrestrial or lignicolous; pileus typically subglobose to pyriform, but usually convex to umbonate or acute, moist or viscid, yellow to tawny or red, smooth

or with small scales; spores ovate to ellipsoid, with germ pore, pale to dark coloured; chrysocystidia present or absent; clamp connections present.

Four species have been referred to *Le Ratia*, all described from New Caledonia and transferred to *Leratiomyces* by Bresinsky & Binder (1998). They were discussed by Heim (1968). Of these, *L. smaragdinus* (Pat.) Bresinsky & Manfr. Binder and *L. atrovirens* (R. Heim) Bresinsky & Manfr. Binder do not belong in this genus (see 'Excluded species'). *Leratiomyces coccinea* (Massee & Wakef.) Bresinsky & Manfr. Binder has not received further study. As currently understood the genus therefore contains 8 species and is widespread in both tropical and temperate regions. Although typically secotioid, most species now referred there actively discharge their spores. They are variable as noted in the emended diagnosis and it seems that the genus cannot be concisely defined morphologically.

***Leratiomyces ceres*** (Cooke & Massee) Spooner & Bridge, **comb. nov.**
MycoBank MB 511252.

**Basionym:** *Agaricus ceres* Cooke & Massee, Grevillea 16: 72 (1888)
= *Psilocybe ceres* (Cooke & Massee) Sacc., Syll. fung. 9: 140 (1891)

= *Nematoloma rubrococcineum* Balletto, Bull. Trimestriel Soc. Mycol. France 83: 217 (1967)
= *Stropholoma rubrococcineum* (Balletto) Balletto, Micol. Ital. 18: 36 (1989) (nom. inval., Arts 33.4, 43.1)
= *Hypholoma rubrococcineum* (Balletto) Nonis, Rivista Micol. 37: 109 (1994)

= *Stropharia aurantiaca* [ss auct., misapplied]

Specimen examined: Australia, Melbourne (near), 7 June 1887, J. Reader 35 (holotype)

Selected descriptions and illustrations: Balletto (1967, as *Nematoloma rubrococcineum*), Fuhrer (2005, as *S. aurantiaca*), Orton (1960, as *S. aurantiaca*), Pegler & Legon (1998, as *S. aurantiaca*), Reid (1966, as *S. aurantiaca*), Taylor (1981, as *S. aurantiaca*), Watling & Gregory (1987, as *S. aurantiaca*).

***Leratiomyces cucullatus*** (Shope & Seaver) Beever & D.-C. Park, **comb. nov.**
MycoBank MB 511253.

**Basionym:** *Bolbitius cucullatus* Shope & Seaver, Mycologia 27: 649 (1935)
= *Weraroa cucullata* (Shope & Seaver) Thiers & Watling, Madroño 21(1): 2 (1971)

Selected descriptions and illustrations: Arora (1979, as *Weraroa*), Castellano et al. (1989, as *Weraroa*), Thiers & Watling (1971, as *Weraroa*).

***Leratiomyces erythrocephalus*** (Tul. & C. Tul.) Beever & D.-C. Park, **comb. nov.**
MycoBank MB 511254.

**Basionym:** *Secotium erythrocephalum* Tul. & C. Tul. [as Tul.], in Raoul, Ann. Sci. Nat., Bot. Sér. 3, 2: 115 (1844)

= *Weraroa erythrocephala* (Tul. & C. Tul.) Singer & A.H. Sm., Bull. Torrey Bot. Club 85: 329 (1958)
= *Clavogaster erythrocephalus* (Tul. & C. Tul.) Lintott, Cass History & Science: 339 (1977) (nom. inval., Art. 33.4)

Selected descriptions and illustrations: Cunningham (1944, as *Secotium*), Singer & Smith (1958, as *Weraroa*), Soop (2005, as *Weraroa*), Taylor (1981, as *Weraroa*).

***Leratiomyces magnivelaris*** (Peck) Bridge & Spooner, **comb. nov.**
MycoBank MB 511255.
**Basionym:** *Stropharia magnivelaris* Peck, Harriman Alaska Expedition: 44 (1905)
= *Nematoloma magnivelare* (Peck) Singer, Agaricales in Modern Taxonomy, ed. 4: 564 (1986)
= *Psilocybe magnivelaris* (Peck) Høil., in Knudsen & Hansen, Nordic J. Bot. 11(4): 481(1991)

Selected descriptions and illustrations: Kytövuori (1990, as *Stropharia*), Noordeloos (1998, 1999, as *Psilocybe*), Watling & Gregory (1987, as *Psilocybe percevalii*)

***Leratiomyces percevalii*** (Berk. & Broome) Bridge & Spooner, **comb. nov.**
MycoBank MB 511257.
Basionym: *Agaricus percevalii* Berk. & Broome, Ann. Mag. Nat. Hist., Ser. 5, 3: 206 (1879)
= *Stropharia percevalii* (Berk. & Broome) Sacc., Syll. fung. 5: 1016 (1887)
= *Psilocybe percevalii* (Berk. & Broome) P.D. Orton, Notes Roy. Bot. Gard. Edinburgh 29: 80 (1969)

Selected descriptions and illustrations: Kytövuori (1990, as *Stropharia*), Noordeloos (1998, 1999, as *Psilocybe*), Rald (1989, as *Stropharia*)

***Leratiomyces squamosus*** (Pers.) Bridge & Spooner, **comb. nov.**
MycoBank MB 511256.
**Basionym:** *Agaricus squamosus* Pers., Syn. meth. fung.: 409 (1801)
= *Stropharia squamosa* (Pers.) Quél., Mém. Soc. Émul. Montbéliard, 2e Sér., 5: 348 (1873)
= *Geophila squamosa* (Pers.) Quél., Enchir. fung.: 111 (1886)
= *Nematoloma squamosum* (Pers.) Singer, Sydowia 2: 36 (1948)
= *Psilocybe squamosa* (Pers.) P.D. Orton, Notes Roy. Bot. Gard. Edinburgh. 29: 80 (1969)
= *Hypholoma squamosum* (Pers.) Urbonas, Lietuvos T.S.R. Mokslu Akad. Darb. C, 4(72): 12 (1975)
= *Stropholoma squamosum* (Pers.) Balletto, Micol. Ital. 18: 36 (1989) (nom. inval., Arts 33.4, 43.1)

Selected descriptions and illustrations: Breitenbach & Kränzlin (1995), Guzmán (1983, as *Psilocybe*), Kytövuori (1990, as *Stropharia*), Noordeloos (1999, as *Psilocybe*), Watling & Gregory (1987, as *Psilocybe*).

***Leratiomyces squamosus*** var. ***thraustus*** (Schulzer ex Kalchbr.) Bridge & Spooner, **comb. nov.**

MycoBank MB 511258.

**Basionym:** *Agaricus thraustus* Schulzer ex Kalchbr., Icon. select. Hymenomyc. Hung.: 30 (1873) & pl. 15 f. 2

= *Stropharia thrausta* (Schulzer ex Kalchbr.) Sacc., Syll. fung. 5: 1016 (1887)

= *Agaricus squamosus* var. *thraustus* (Schulzer ex Kalchbr.) Cooke, Handb. Brit. fung., ed. 2: 199 (1887)

= *Stropharia squamosa* var. *thrausta* (Schulzer ex Kalchbr.) Massee, Brit. fung.-fl. 1: 402 (1892)

= *Psilocybe thrausta* (Schulzer ex Kalchbr.) Bon, Bull. Trimestriel Soc. Mycol. France 85(4): Suppl., Atlas Pl. 182 (1970) [1969]

= *Hypholoma thraustum* (Schulzer ex Kalchbr.) Urbonas [as '*thrausta*'], Lietuvos T.S.R. Mokslu Akad. Darb. C, 4 (72): 14 (1975)

= *Psilocybe squamosa* var. *thrausta* (Schulzer ex Kalchbr.) Guzmán, Nova Hedwigia Beih. 74: 321 (1983)

= *Agaricus thraustus* var. *aurantiacus* Cooke, Ill. Brit. fung. 562 (555) (1885)

= *Stropharia percevalii* var. *aurantiaca* (Cooke) Sacc., Syll. fung. 5: 1016 (1887)

= *Agaricus squamosus* f. *aurantiacus* (Cooke) Cooke, Handb. Brit. fung., ed. 2: 199 (1887) [some may prefer to interpret this combination as a form of *A. squamosus* var. *thraustus*]

= *Stropharia squamosa* var. *aurantiaca* (Cooke) Massee, Brit. fung.-fl. 1: 402 (1892)

= *Stropharia aurantiaca* (Cooke) S. Imai, J. Fac. Agric. Hokkaido Imp. Univ. 43: 267 (1938)

= *Nematoloma aurantiacum* (Cooke) Guzmán, Nova Hedwigia Beih. 51: 114 (1975) (nom. inval., Art. 33.4)

= *Hypholoma aurantiacum* (Cooke) Faus, in Moreno & Faus, Bol. Soc. Micol. Castellana 7: 70 (1982)

= *Nematoloma aurantiacum* (Cooke) Guzmán ex Singer, Agaricales in Modern Taxonomy, ed 4 (Koenigstein): 564 (1986)

= *Stropholoma aurantiacum* (Cooke) Balletto, Micol. Ital. 18(1): 36 (1989) (nom. inval., Arts 33.4, 43.1)

= *Psilocybe aurantiaca* (Cooke) Noordel., Persoonia 16(1): 128 (1995)

Selected descriptions and illustrations: Fortey (2004, as *Psilocybe*), Guzmán (1983, as *Psilocybe*), Imazeki & Hongo (1987, as *Naematoloma squamosum* var. *thraustum*), Noordeloos (1999, as *Psilocybe squamosa* var. *thrausta*).

## Excluded species

The LSU sequence available for *Leratiomyces smaragdinus* places it in the /agrocybe clade (clade 80; Moncalvo et al. 2002) and our analysis confirmed this placement. Our analysis of the LSU sequence of *Leratiomyces atrovirens* showed 95-97% homology to a number of species of *Coprinopsis* and *Psathyrella* suggesting that this species should be placed within Moncalvo et al. (2002) / psathyrella clade (clade 89). Walther et al. (2005) recovered an LSU sequence for *Agrocybe pediades* as a sister group to *Stropharia squamosa*. The sequence used

in that study (AY207142) differs by some 4% from two other LSU sequences reported for this species (AJ871493 & DQ110872) that are 99% similar to each other. The correct placement of *A. pediades* is unclear.

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# Three new records for the lichen biota of Turkey

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**Abstract** - Three lichen species, *Cladonia borealis*, *C. dimorpha*, and *Sphinctrina tubiformis*, are new to the lichen flora of Turkey. For each a short description is presented.

**Key words** - Trabzon, Giresun

## Introduction

Lichenology in Turkey started seriously only in the last two decades. So far a total of 360 papers refer to lichens from Turkey (John 2004). For Trabzon province in the eastern Black Sea region 518 species have been reported (John 1995 and references therein, John 1999, 2000, 2002; John & Breuss 2004, John & Nimis 1998, John et al. 2000, Kınalıoğlu 2007b, Yazıcı 1996, 1999, 2006; Yazıcı & Aslan 2002, 2005) and for Giresun province only 320 species (Aslan & Yazıcı 2006, John & Breuss 2004, Kınalıoğlu 2004, 2005, 2006, 2007a; Kınalıoğlu & Engin 2004, Küçük 1990, Yazıcı & Aslan 2005, 2006). The present paper is a further contribution to the lichen flora of these provinces.

## Materials and methods

Samples were collected at three different sites in Trabzon and Giresun provinces between 17 August 2005 and 14 January 2007. They were identified using various floras and identification keys (e.g. Brodo et al. 2001, Goward 1999, Osyczka 2006, Purvis et al. 1992, Wasser & Nevo 2005, Wirth 1995). Specimens are kept in the herbarium of Faculty of Science and Arts, Giresun University, Giresun.

## Species recorded

***Cladonia borealis*** S. Stenroos

Primary thallus squamulose, persistent, lobed, 3-5.5 mm across, with a dark yellow or orange tint at the base below. Podetia 2-2.5 cm tall, 4-10 mm across,

with rounded areoles to continuously corticate at base, without soredia or granules. Cups goblet-shaped, margins smooth or proliferating. Apothecia bright red, common, on the cup margins. Podetia PD-, K-, C-, KC+ yellow (Brodo et al. 2001, Goward 1999, Osyczka 2006, Wirth 1995).

Grows on soil, humus, mossy rocks or rarely decaying wood in full sun from sea level to the mountain tops. Known from North America, Finland, Gemany, Greenland, Svaldbard (Brodo et al. 2001, Goward 1999, Osyczka 2006, Wirth 1995).

Turkey, Trabzon, Araklı, S of Kızılkaya Yaylası, on soil, 40° 40′ 21″ N, 40° 1′ 24″ E, 2350 m, 17 August 2005, Kınalıoğlu 1445.

*C. dimorpha* S. Hammer

Primary thallus squamulose, persistent. Podetia green to greyish, 2.7-3.5 cm tall, 2-2.6 mm across. Cup interior containing strongly convex granules or areoles, or both, occasionally also microsquamules, usually inclined, bearing numerous proliferations, these to more than 1 cm tall, in part longitudinally fissured. Apothecia dark brown, on the cup margins. Medulla PD+ orange (Goward 1999).

Grows on soil and mossy rocks in open, coastal localities at lower elevations. Distribution poorly known, reported from western North America and western Eurasia (Goward 1999).

Turkey , Trabzon, Araklı, SE of Paska lar Yaylası, on soil, 40° 40′ 3″ N, 40° 1′ 41″ E, 2400 m, 17 August 2005, Kınalıoğlu 1442.

*Sphinctrina tubiformis* A. Massal.

Not developing an autonomous thallus. Apothecia 0.16-0.4 mm tall; stalk short to absent, 0-1.2 times as long as wide, dark brown or black; head 0.14-0.36 mm in diam., globose, usually partly broadened vertically and slightly irregular, bright black or dark brown; true exciple dark brown in section, K-. Ascospores 10-15.5 x 5.5-9 µm, broadly ellipsoid, the end pointed, with distinctive surface ornamentation of longitudinally arranged, coarse ridges (Purvis et al. 1992).

Commensalistic to pathogenic on *Pertusaria leioplaca*, occasionally on other *Pertusaria* species. Known from Europe, North, Central & South America, Africa, Asia, Fiji (Purvis et al. 1992).

Turkey , SW of Giresun city centre, on *Pertusaria* sp., 40° 54′ 20″ N, 38° 19′ 24″ E, 5 m, 14 January 2007, Kınalıoğlu 1240.

## Acknowledgements

I would like to thank Prof. Teuvo Ahti (Fnland) for the identification of *Cladonia borealis* and *Cladonia dimorpha* and Dr. Javier Etayo (Spain) for that of *Sphinctrina tubiformis*. I also thank peer-reviewers Owe-Larsson & Sipman for their contributions on revising article.

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# New species of operculate discomycetes (*Ascomycota, Pezizomycetes*) for Israeli mycobiota

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**Abstract**—Three species from the genera *Helvella* and *Morchella* were recorded for the first time for Israel. Morphological and habitat descriptions, general distribution, illustrations, and taxonomical discussions on *Morchella elata*, *Helvella chinensis* and *H. spadicea* are presented in this paper. Descriptions of all species are provided for Israeli samples.

**Key words**—Mediterranean, morels, taxonomy, *Pezizales*

## Introduction

The study of operculate discomycetes has been neglected in Israel since the publications of Avizohar-Hershenzon & Nemlich (1974, 1978), Binyamini (1972a, b, 1973a, b, 1984, 1986, 1989, 1993, 1994), Nemlich & Avizohar-Hershenzon (1972, 1975, 1976a, b), and Rayss (1940, 1947, 1953). The *Morchella* and *Helvella* species diversity of Israel is poorly known. Only a few surveys of the two genera have been published. The most notable ones include Rayss (1940), Nemlich & Avizohar-Hershenzon (1972), and Binyamini (1984, 1986, 1989). After reviewing regional mycological reports, we found that the following species from the genera *Morchella* and *Helvella* exist in Israel: 2 species of *Morchella* [*M. conica* Pers., *M. esculenta* (L.) Pers.] and 10 species of *Helvella* [*H. acetabulum* (L.) Quél., *H. atra* J. Koenig, *H. crispa* (Scop.) Fr., *H. elastica* Bull., *H. ephippium* Lév., *H. lacunosa* Afzel., *H. leucomelaena* (Pers.) Nannf., *H. pezizoides* Afzel., *H. phlebophora* Sacc., *H. queletii* Bres.].

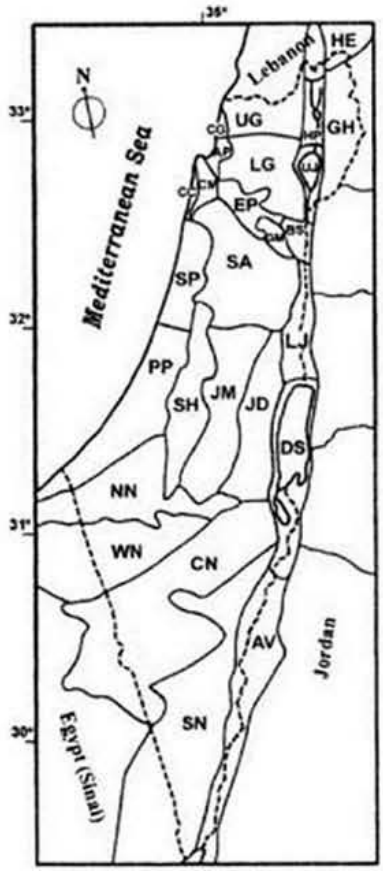



**Figure 1:** Accepted abbreviations of nature regions of Israel: AP – Akko Plain; AV – Arava Valley; BS – Beit Shean Valley; CC – Carmel Coast; CG – Coast Galilee; CM – Carmel Mount; CN – Central Negev; DS – Dead Sea Area; EP – Esdraelon (Yizre'el) Plain; GH – Golan Heights; GM – Gilboa Mount; HE – Hermon Mount; HP – Hula Plain; JD – Judean Desert; JM – Judean Mts.; LG – Lower Galilee; LJ – Lower Jordan Valley; NN – Northern Negev; PP – Philistean Plain; SA – Samaria; SH – Shefela; SN – South Negev; SP – Sharon Plain; UG – Upper Galilee; UJ – Upper Jordan Valley; WN – Western Negev.

## Materials, methods & results

Collected specimens are preserved in the herbarium of the Institute of Evolution, University of Haifa (HAI, Haifa, Israel). The microscopic characteristics were observed with the Carl Zeiss-amplival microscope. Microscopic photos were taken with a "Sony Camera". The chemical reagent used in the microscopic examination was Melzer's reagent. Fungal material was mounted on a microscope slide and examined in water using a light/dark field microscope with or without phase contrast at ×20, ×40, and ×100 (oil immersion). For statistical calculations, 30–40 ascospores, asci, and paraphyses were measured for every preparation.

Distribution of species is shown (Figure 1) using the natural regions of Israel as described by Feinbrun-Dothan & Danin (1998).

As a result of our investigation, three new species of operculate discomycetes (*Morchella elata, Helvella chinensis,* and *H. spadicea*) have been added to data on Israeli mycobiota. Detailed descriptions, iconography, locations and dates of collections in Israel, habitat and general distribution are given. Taxonomic discussions of mentioned species are presented below.

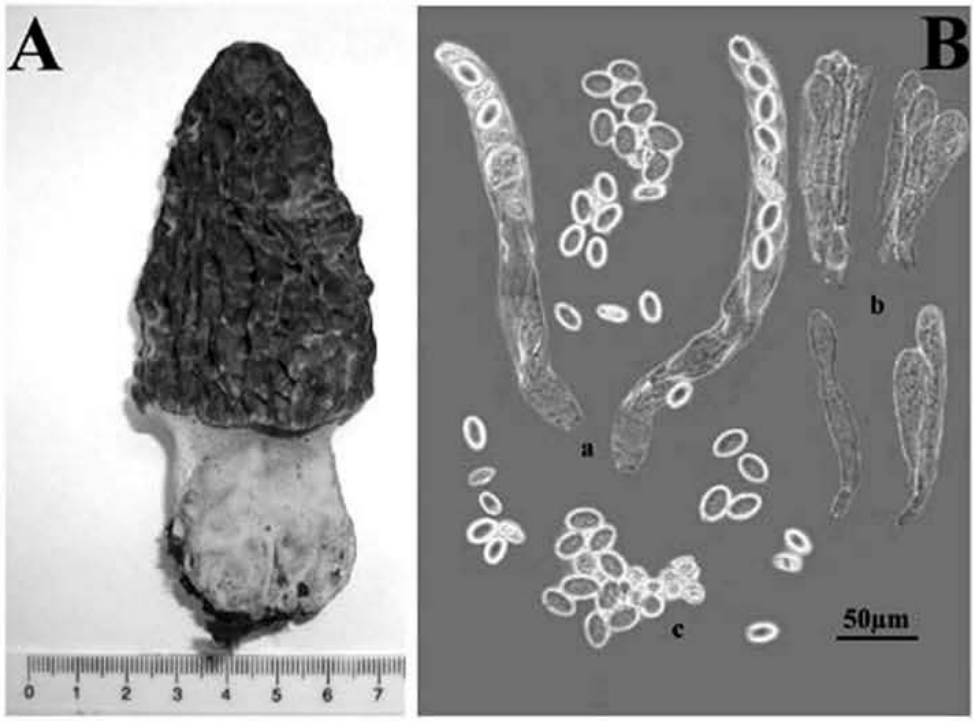



**Figure 2.** *Morchella elata*.
A – ascocarp; B – microscopic structure: a – asci; b – paraphyses; c – ascospores.

## Description and discussion of species

***Morchella elata*** Fr. Syst. Mycol. 2: 8. 1822 Figure 2
Icon.: Dennis [1978] 1981: pl. 1B), Breitenbach & Kränzlin (1984: pl. 2).

### Macromorphology

Ascocarps 40–150 mm high, 20–50 mm broad, narrowly to broadly conical, occasionally more rounded, obtuse to ovoid-conical; surface of parallel to meandering ridges and cross-ribs, pubescent when young in some forms; color at first greyish to ochre-brown, occasionally pinkish to blackish overall; with age the ridges become dark grey to blackish brown, the pits lighter, ochre to grey-brown; margin when young, overlapping the stipe at attachment, less so in age. Flesh elastic, whitish, thin, firm, brittle, interior, hollow; odor earthy to fungal; taste not investigated. Stipe 20–70 mm high, 10–60 mm thick, hollow, cylindrical, equal to enlarged above and below, the base with longitudinal folds; surface typically whitish to light grey-ochre, pubescent.

### Micromorphology

Asci up to 241–286 × 16.1–20.7 µm, 8-spored. Ascospores 18.4–23 × 9.2–16.1 µm, Q = 1.4–2, non amyloid, ellipsoid, smooth, sometimes with small

TABLE 1. Important distinguishing characteristics of *Morchella elata* as cited by different authors

| AUTHOR | ASCOCARP HABITUS | ASCI | SPORES | PARAPHYSES |
|---|---|---|---|---|
| PERSONAL DESCRIPTION | 40–150 mm, narrowly to broadly conical, greyish to ochre-brown. Stipe 20–70× 10–60 mm, hollow, whitish to light grey-ochre, base with longitudinal folds | ≤241–286 × 16.1–20.7 µm | Ellipsoid, 18.4–23 × 9.2–16.1 µm | Cylindrical or slightly enlarged upwards, 8–17 µm diam |
| SMICKAYA (1980) | 40–100 mm, conical to obtuse, brown or olive-brown. Stipe 40–60× 15–20 mm, hollow, whitish, base bulbous | 280–300 × 18–20 µm (cylindrical) | Ellipsoid, 17.5/20–22/25 × 11–15 µm | Cylindrical, enlarged upwards, 300×3 µm |
| DENNIS [1978] 1981 | ≤ 100 mm, cylindrical to slightly conical, yellowish brown, dark grey or black. Stipe hollow, white or yellowish, cylindrical | ≤ 300 × 20 µm | Ellipsoid, 18–25 × 11–15 µm, (cream-colored) | Cylindrical or slightly enlarged upwards, 8–17 µm diam (multiseptate) |
| BREITENBACH & KRÄNZLIN (1984) | 50–150 mm, conical, honey-brown, red to black-brown. Stipe hollow,ochre, furfuraceous, wrinkled | 250–300 × 20 µm | Ellipsoid, 18–25 × 12–16 µm (hyaline) | Septate,branched towards slightly clavate base 17 µm diam |
| ELLIS & ELLIS [1988] 1998 | 100 mm, cylindrical to sharply conical. Stipe hollow, white or ochraceous, furfuraceous | ——— | 18–25 × 11–14 µm | ——— |

droplets on both ends (outside the spore wall), spores creamy to pale-tawny in deposit. PARAPHYSES cylindrical or slightly enlarged upwards, hyaline, filiform, multiseptate, and branched toward the base, with slight clavate inflations, 8–17 µm broad.

SPECIMENS EXAMINED. **Israel:** UG, Hanita forest (*Quercus calliprinos*), on the ground, 08. III. 2007; 20. III. 2007, leg. Y. Ur, det. G. Barseghyan (HAI-D-017; D-023; Figure 5).

HABITAT AND DISTRIBUTION: This species is primarily a humus saprotroph, but also can be mycorrhizal (Buscot 1994). The fruiting bodies can appear after fires and are found on soil. EUROPE: UK, Germany, Italy, Norway, Finland, Switzerland, Poland, Czech Republic, Ukraine, Estonia. ASIA: Tajikistan, Kazakhstan, Pakistan, Israel. NORTH AMERICA: USA (California). AUSTRALIA.

DISCUSSION: *Morchella elata* was found in Israel for the first time. It is often confused with old specimens of *M. conica* (Breitenbach & Kränzlin 1984).

Microscopically, all morels are very similar to each other but mostly differ morphologically. *M. elata* is considered by some taxonomists to be synonymous with, or part of a complex of black morel species or subspecies including *M. angusticeps* Peck and *M. conica* (Bunyard et al. 1995, Wipf et al. 1999). The ascomata of *M. elata* are distinguished from other black morels by smooth, ivory-white stalks in younger fruiting bodies, by steel-grey tones in the ridges and pits of the pileus, and by the production of spores larger than those of *M. angusticeps* (Weber 1988). Generally, we can find different descriptions by different authors, which are presented in Table 1.

***Helvella chinensis*** (Velen.) Nannf. & L. Holm,
in Lundell et al., Publ. Herb. Univ. Uppsala 18: 5. 1985 **Figure 3**

- *Macropodia chinensis* Velen., Novit. Mycol.: 200. 1940.

=*Octospora villosa* Hedw., Descr. micr.-anal. musc. frond. 2: 54. 1789.

- *Helvella villosa* (Hedw.) Dissing & Nannf., Svensk bot. Tidskr. 60: 330. 1966, nom. illegit., non *H. villosa* Schaeff. 1774.
- *Helvella dissingii* Korf, Mycotaxon 31(2): 381. 1988.

**Icon.:** Hedwig (1789: pl. 19 B, as *Octospora villosa*), Dissing & Nannfeldt (1966: pl. 1.3, 2.1, as *Helvella villosa*), Ellis & Ellis [1988]1998: pl. 23. 220, as *H. villosa*).

### Macromorphology

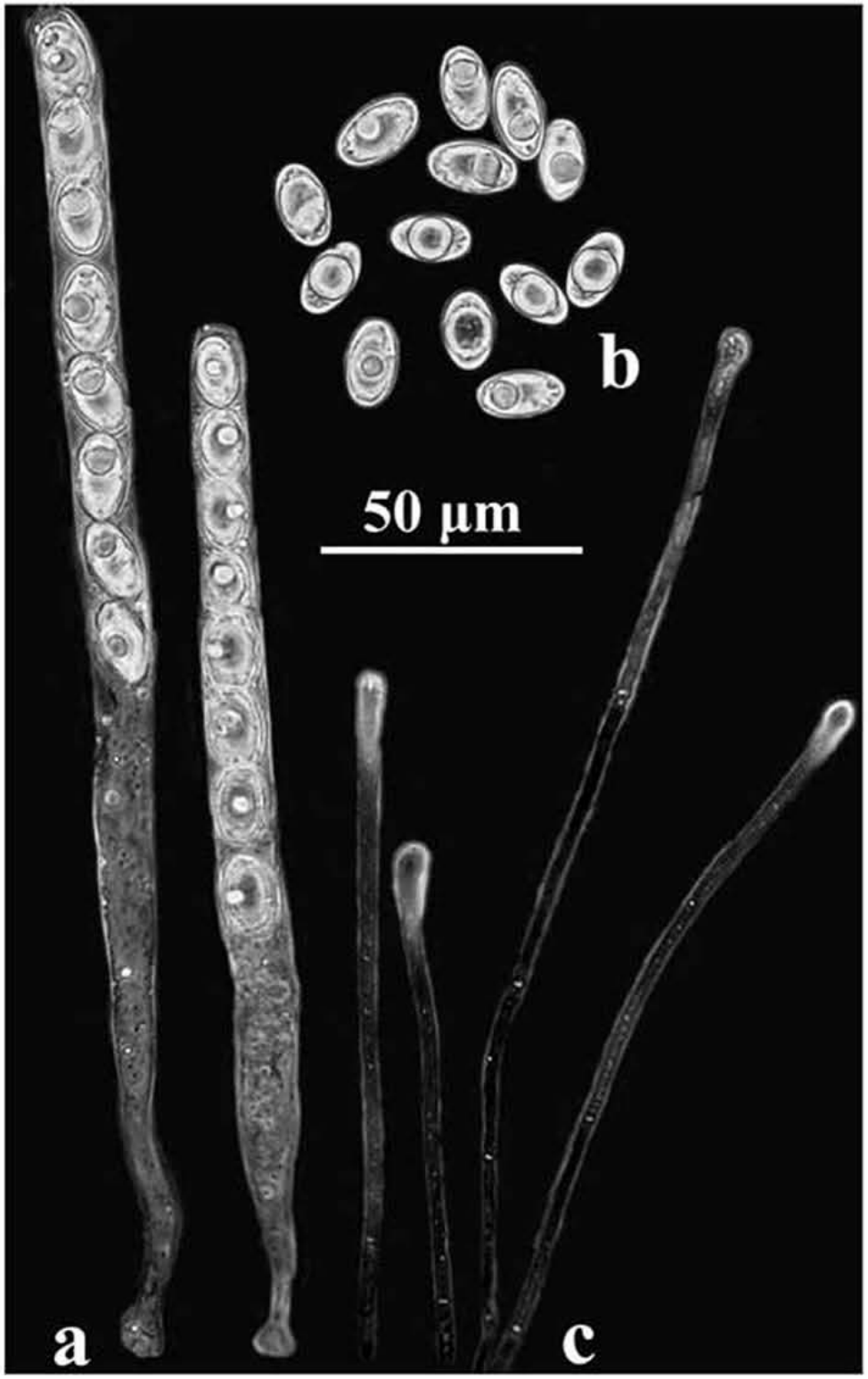
Ascocarps 10–25 mm wide, cup-shaped, compressed when young, becoming saucer-shaped at maturity, splitting into several irregular lobes, exterior pubescent (slightly hairy) to villose; margin sinuous or wavy, bent inward; grey, grayish-brown to steel grey, under side greyish to dark brown to steel grey. Hymenium smooth and dull, dark grey to grey-brown. Flesh whitish, odor and taste indistinct. Stipe 10–15 mm long, cylindrical, solid, downy, thick, same color as underside of cup, becoming whitish or paler at the base.

### Micromorphology

Asci cylindrical, attenuated in the base, measure 230–250 × 11.5–13.8 µm, 8-spored. Ascospores 16.1–18.4 × 9.2–11.5 µm, Q = 1.75–1.6, oblong to ellipsoid, with one large and 0 to 5 small oil-droplets, usually smooth. Paraphyses cylindrical, tips with slight clavate in the apex, 4.6–6.9 µm broad. Hyphal outgrowths on the excipulum with oblong cells.

Specimens Examined. Israel: CM, near University of Haifa, on the burnt ground in woods after forest fires in the mountains, *Quercus* and *Pinus* mixed forest. 12.IV.2001, leg. S. Reshetnikov, det. G. Barseghyan (HAI-D-30; Figure 5).

Habitat and distribution: Humus saprotroph; very rare. Edibility not known. On sandy soil in damp woods and along streams under hardwoods, early summer to fall, often after fires. Europe: UK, Switzerland, Germany, Belgium, Italy, Poland, Norway, Czech Republic. Asia: Israel, China, West Pakistan. North America: USA. Australia.


b
50 µm
a
c

Discussion: *Helvella chinensis* is found in Israel for the first time.

*Helvella chinensis* has been confused with *H. macropus* (Pers.) P. Karst. *H. chinensis* tends to be darker in color and less villose than *H. macropus*. Moreover *H. chinensis* has a greater tendency for the cup to split into irregular lobes at maturity than *H. macropus*. These two similar species not only have macromorphological differences, but we noticed some differences in micromorphology: *H. chinensis* has smaller spore sizes and smoother surfaces, than *H. macropus*, which has more expressed warts exosporial warts. It could also be confused with *H. ephippium* and *H. queletii* (Breitenbach & Kränzlin 1984).

***Helvella spadicea*** Schaeff., Fung. Bavar. Palat. Nasc. 4: 112. 1774 Figure 4

=*Helvella leucopus* Pers., Mycol. Eur. 1: 213. 1822.

=*Helvella albipes* Fuckel, Jb. nassau. Ver. Naturk. 23-14: 344. 1870.

=*Helvella heganii* Copel., Ann. Mycol. 2: 510. 1904.

**Misapplied:** *Helvella monachella* Scop., sensu Quél., Enchir. Fung.: 273. 1886.

### Macromorphology

Ascocarps 15–30 mm high, 10–15 broad, saddle-shaped with two lobes, upper surface dark brown to black, under surface whitish to creamy buff, glabrous (smooth), margin curving toward stipe. Flesh thin, elastic, whitish, with insignificant smell and taste. Stipe white to pale cream, terete (round), hollow, equal or tapering toward top, 15–25 mm long, 5–10 mm thick, smooth, flattered or pitted near base.

### Micromorphology

Asci are cylindrical, non amyloid, attenuated in the base, measure 300–350 × 18.4–23 µm, 8-spored. Ascospores of 16.1–18.4 × 9.2–11.5 µm, ellipsoid, hyaline, smooth, with large central oil-drop, and with many small oil drops around large drop. Paraphyses of 230–250 × 4.6–6 µm cylindrical, slender, septate, forked, and clavate in the apex.

Specimens Examined. Israel: UG, Park Goran, on the ground in *Quercus calliprinos* frorest, 9.II.2007, leg. A. Rotenberg, det. G. Barseghyan (HAI-D-32; Figure 5).

Habitat and distribution: Humus saprotroph; very rare. Inedible. Europe: UK, Germany, Austria, France, Italy, Belgium, the Netherlands, Czech Republic, Romania, Hungary, Portugal. Asia: Israel, Turkey, Kyrgyzstan. North America: USA. Africa: Algeria.

Discussion: *Helvella spadicea* was found in Israel for the first time. Classical species very easy to identify by its dark coloration of two-lobed, saddle-shaped

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Figure 3. Microscopic structure of *Helvella chinensis*: a – asci with eight smooth ascospores; b – ascospores with oil-drops; c – paraphyses with slightly inflated in the apex.

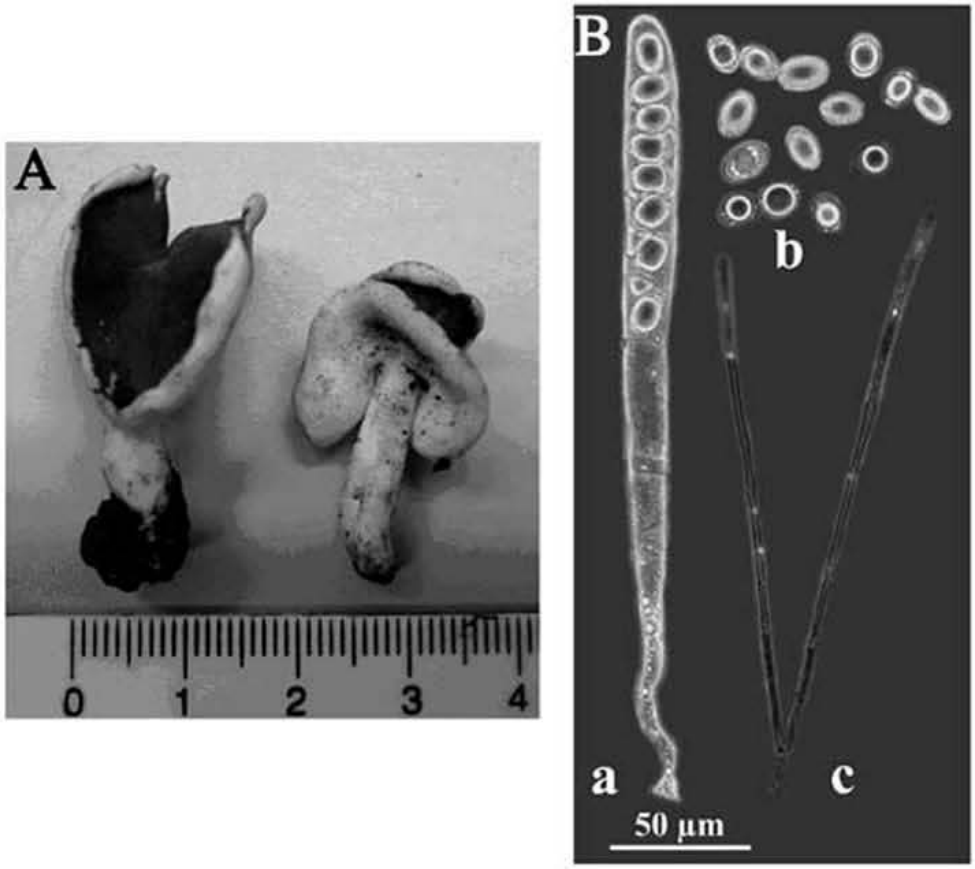


**Figure 4.** *Helvella spadicea.*
A – ascocarps; B – microscopic structure: a – asci with eight smooth ascospores; b – ascospores with oil-drops; c – paraphyses forked and inflated in the apex.

ascocarp with smooth, white, cylindrical stipe. It is different from other species of genus *Helvella,* but it can be confused with *H. lacunosa* and *H. fusca* Gillet. The last ones have deep furrows. Ascomata of *H. lacunosa* have the same coloration as *H. spadicea,* but stipe greyish and furrowed. Ascomata of *H. fusca* have dark brown or black coloration, with a little reddish tint, stipe white, deeply furrowed.

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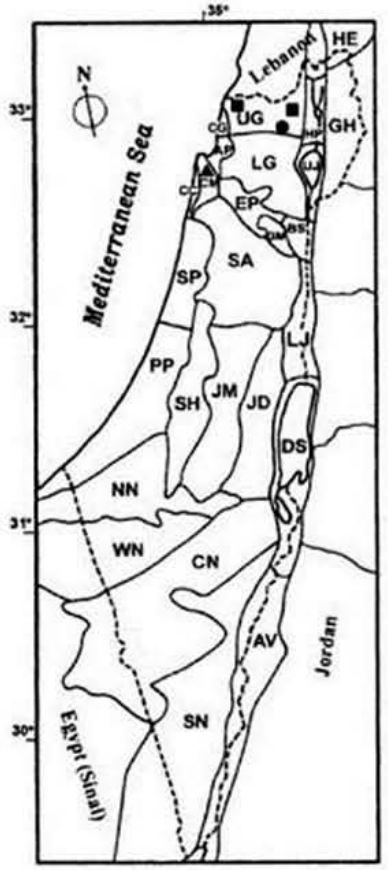


Figure 5. Distribution of *Morchella elata*, *Helvella chinensis* and *H. spadicea* in Israel.

■ — *Morchella elata*

▲ — *Helvella chinensis*

● — *Helvella spadicea*

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MYCOTAXON

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# Type studies on South American *Strophariaceae*: 1. *Pholiota varzeae* from the Brazilian Amazon

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**Abstract** — The study of the holotype of *Pholiota varzeae* revealed the presence of numerous acanthocytes in its rhizomorphs, generally accepted as an exclusive generic character of the genus *Stropharia*. The proposed new combination, *S. varzeae*, is based in morphological and anatomical characters. The taxonomic discussion is accompanied by detailed macroscopic and microscopic descriptions and illustrations.

**Key words** — *Agaricales*, *Pholiota* section *Albivelatae*, Amazon mycobiota, várzea vegetation, nomenclature

As part of ongoing studies on the genus *Stropharia* (Fr.) Quél. (*Strophariaceae*, *Agaricales*) in south Brazil (Cortez & Silveira 2008), the type specimens of some South American taxa were studied. In this work, the holotype collection of *Pholiota varzeae*, collected from the Brazilian Amazon and described by Singer (1989), is detailed and discussed and its transference to the genus *Stropharia* is proposed.

## Materials and methods

The holotype collection of *Pholiota varzeae*, on loan from Chicago's Field Museum (F), was examined. Thin sections of the dried basidiomes were mounted in 5% KOH and 1% Congo Red solutions and examined microscopically. A minimum of 25 measurements were taken for all microstructures and microdrawings were made assisted by a light tube. For basidiospores, $Q$ represents the ratio of length width, $Q_m$ the mean value of $Q$, and $n$ the number of measured basidiospores. The macroscopic description (including Maerz & Paul (1930) color codes, shown here in quotation marks) is adapted from the original Latin diagnosis (Singer 1989).

## Taxonomy

***Stropharia varzeae*** (Singer) Cortez, **comb. nov.** FIGS. 1-7

MYCOBANK MB 511191

BASONYM: *Pholiota varzeae* Singer, Fieldiana, Bot. 21: 113, 1989.

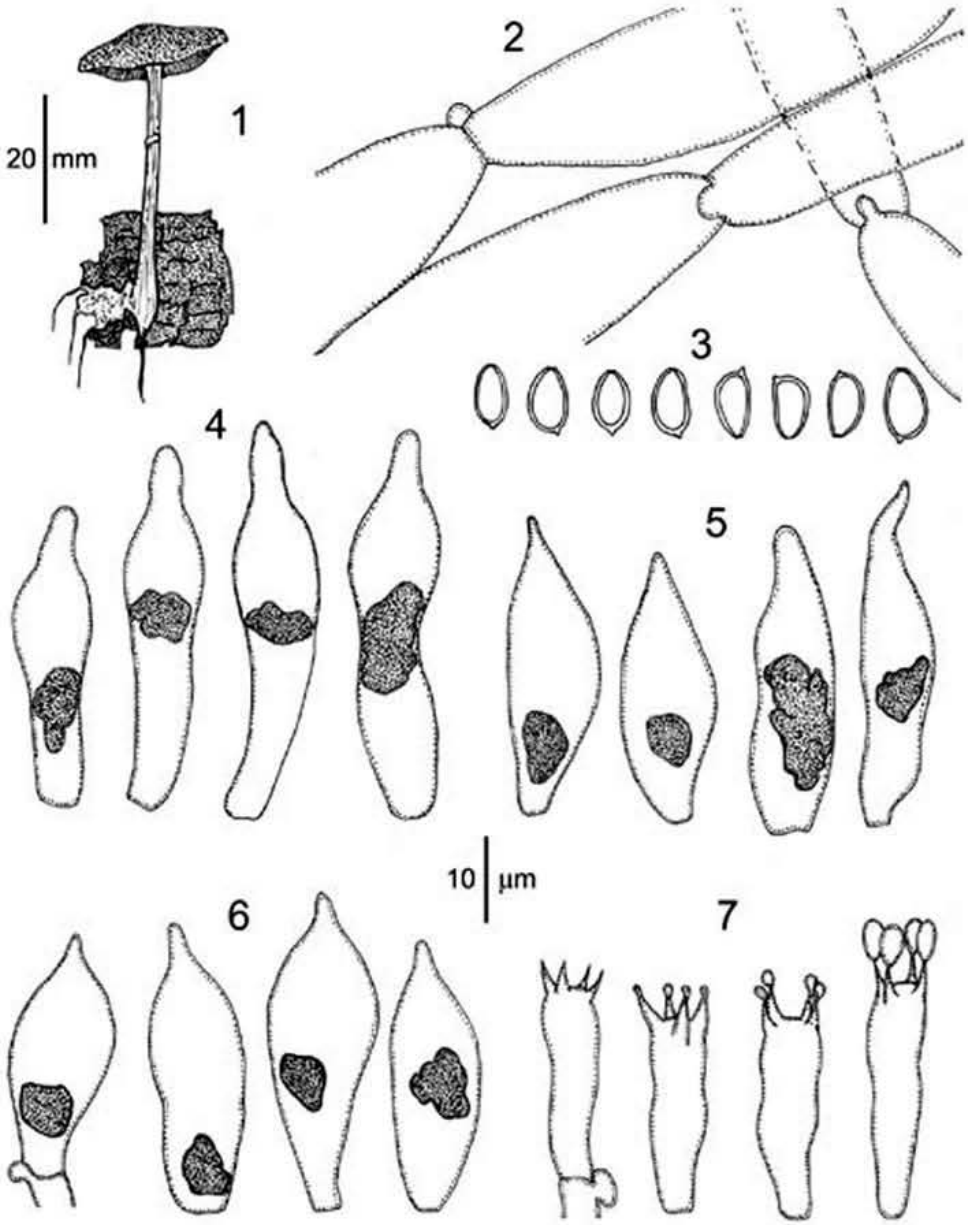
PILEUS 21–30 mm diam, convex and umbonate to subumbonate to finally depressed; rusty ("henna") with the disc reddish-brown ("mascara" to "Maracaibo"), surface dry; context whitish. LAMELLAE adnate, close, brownish ("Cuban sand", "atmosphere" to "kis kilim"), with a conspicuous whitish margin. STIPE 22–46 × 2.5–7.5 mm, central, cylindrical or with a slightly attenuate apex and expanded base; cream to grayish colored, with longitudinal dark brown fibrils; abundant white rhizomorphs and basal mycelium abundant at the base. VEIL forming a membranous, slightly grooved and whitish annulus. SPORE PRINT "falcon".

BASIDIOSPORES 6–7.5(–8) × 4–4.5(–5) µm, $Q$= 1.50–1.75(–1.80), $Q_m$= 1.65, $n$= 30, subellipsoid to ovoid in both face- and side-view, some slightly reniform in profile, smooth and thick-walled, with a reduced germ-pore; yellowish brown in KOH. BASIDIA 20–27 × 6–7.5 µm, clavate, 4-spored. PLEUROCYSTIDIA 35–46 × 9.5–13 µm, as chrysocystidia, fusoid or subclavate, with obtuse to rounded apex, thin-walled, with yellowish to greenish contents. CHEILOCYSTIDIA (24–) 29–38 × (8–)10–13 µm, as chrysocystidia, fusoid to clavate, with a mucronate apex, similar to pleurocystidia, but slightly smaller; thin-walled, with yellowish to greenish contents. PILEIPELLIS a trichodermium, hyphae inflated to subcylindrical, clamped, smooth and thin-walled, hyaline, 11–17(–21) µm diam. CONTEXT formed of interwoven, hyaline, thin-walled, and clamped hyphae, 8–16 µm diam. GILL TRAMA regular, composed of hyaline, thin-walled, cylindrical to inflated hyphae, 6–9(–11) µm diam. STIPITIPELLIS composed of hyaline, non-gelatinized, filamentous, smooth and thin-walled hyphae, 4–6.5 µm diam. CAULOCYSTIDIA 29–47 × 9–12 µm, as chrysocystidia, mainly clavate or some fusoid, similar to the pleurocystidia, abundant on the stipe apex. ACANTHOCYTES numerous, observed only on rhizomorphs' surface. CLAMP CONNECTIONS present.

HABITAT: gregarious on decayed palm and dicotyledonous wood in tropical flooded forest ("várzea" vegetation).

EXAMINED SPECIMEN: BRAZIL. Amazonas: Paraná do Januacá, near to Santa Luzia, Lago do Castanho, in "várzea" vegetation, 23.V.1980, *Singer B 12177* (F 1030768 – holotype).

REMARKS: The membranous annulus, the presence of chrysocystidia as pleuro-, cheilo- and caulocystidia, and especially the presence of acanthocytes in the type specimens of *Pholiota varzeae* support its transfer to *Stropharia*. Singer (1989) included *P. varzeae* in *Pholiota* section *Albivelatae* A.H. Sm. & Hesler; most species of this section have been assigned to *Stropharia* due the presence of acanthocytes (cf. Norvell & Redhead 2000), an exclusive feature of this genus




Figs. 1-7. *Stropharia varzeae* (Holotype, F 1030768)
1. Basidiome. 2. Hyphae of the pileipellis. 3. Basidiospores.
4. Pleurocystidia. 5. Caulocystidia. 6. Cheilocystidia. 7. Basidia.

(Farr 1980). An additional distinguishing feature of the species is its habitat in "várzea" vegetation, growing on palm and wood debris (Singer 1989).

Except for only minor differences between the basidiospore size ranges described by Singer (1989) and those reported here, the measurements of the

basidia, cystidia, and pileipellis hyphae in the type were slightly larger than those presented by Singer (1989). Singer also did not report cheilocystidia in this species, considering them "*veris haud differentiatis*". Cheilocystidia as well as pleuro- and caulocystidia were checked in the type specimens and we consider it as chrysocystidia.

Singer (1989) related his *Pholiota varzeae* to *P. cubensis* Earle, another species occurring in Brazil (Pegler 1997, Cortez & Silveira 2008). The latter species differs mainly by its more robust habit and absence of chrysocystidia on gill edge and stipitipellis. Norvell & Redhead (2000) recently transferred *P. cubensis* to *Stropharia*, renaming it *S. earlei* Norvell & Redhead. Another similar taxon in *Pholiota* section *Albivelatae* is *P. apiahyna* Speg., which differs from *S. varzeae* by the fusoid to clavate and mucronate pleurocystidia and the non-chrysocystidioid cheilocystidia.

Based on all above-cited features and following the criterion of the presence of acanthocytes as a synapomorphic feature of *Stropharia*, *P. varzeae* is considered here as an authentic member of this genus and thus the above new combination is proposed. *Stropharia* has proved to be a more diverse than previously documented in Brazil. To determine if *S. varzeae* is in fact restricted to the "várzea" vegetation, as Singer (1984, 1989) stated, will deserve future mycological investigations in Amazon region.

## Acknowledgements

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# Lichens from the Batman, Mardin, Osmaniye, and Sivas regions of Turkey

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**Abstract** — A total of 205 lichen taxa (198 species, 2 subspecies, 4 varieties, 1 forma) and one lichenicolous fungus were identified from 25 different localities in the Batman, Mardin, Osmaniye and Sivas regions of Turkey. *Cladonia metacorallifera*, *Caloplaca flavocitrina*, *Caloplaca variabilis* f. *ocellulata*, *Phaeophyscia chloantha* and *Usnea wasmuthii* are new to Turkey. 130 species and one lichenicolous fungus (*Lichenodiplis lecanorae*) are new records from Osmaniye. Moreover, 50 taxa are reported as new from Mardin, 35 from Sivas, and 23 from Batman. Distribution and substrata are summarized for the complete annotated species list posted at http://www.mycotaxon.com/resources/weblists.html

**Keywords** — *Ascomycetes*, flora, biota

## Introduction

Compared to other countries, not many lichen studies have been conducted in Turkey previous to this decade, so that the lichen biota of Turkey remains poorly known. Recently, however, there has been a substantial increase in the number of lichenological papers for Turkey (Halıcı & Aksoy 2006, Yazıcı 2006, Yazıcı & Aslan 2007, Yazıcı & Aptroot 2007). Although previous lichenological

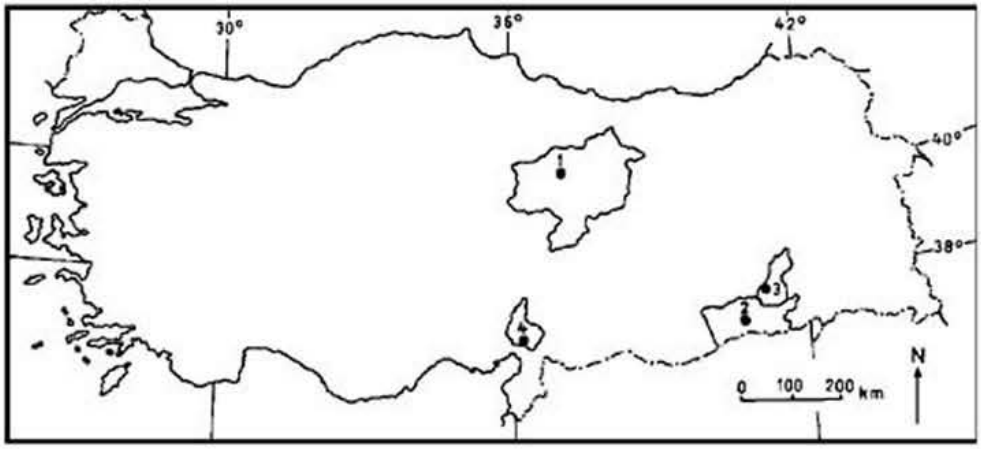


Figure 1. Map of the study area showing the four provinces dealed with in this paper.
1: Sivas, 2: Mardin, 3: Batman, 4: Osmaniye

research (John 2002, John & Türk 2006, John et al. 2000, Hafellner & John 2006, Nimis & John 1998, Steiner (1921) has been reported for Batman, Osmaniye and Sivas, no lichen records have thus far been reported for Mardin (Midyat). Additionally, several regions in Batman, Osmaniye, and Sivas remain still unexplored. This present paper provides information on the lichen biota in the above regions of Turkey.

Sivas and Batman have continental climates. Steppe vegetation is dominant in plateaux and hills. Temperatures range from a winter low of –34.6 °C to a summer high of 38.3 °C. The mean annual humidity is 80.0%. Mean annual rainfall is 420 mm (Akman 1990).

Osmaniye has a typical mediterranean climate. Forest and shrub fields are quite large (42%). Lowlands are seen culture plants, macchia between 500–600 m and coniferous trees in upper fields (Baytop et al. 1963).

Mardin (Midyat) is influenced by both mediterranean and continental climates. The fields are mostly uneven in Mardin and mountains are not very hgh. *Quercus* communities grow in high fields of the mountains (Baytop et al. 1963). The highest precipitation is 115.8 mm in March. The lowest temperature is –2.6 °C in February while the highest 42.5 °C in July (Akman 1990).

Batman forests are composed of *Quercus*, *Juglans*, *Populus*, *Platanus*, *Pistacia*, *Pyrus* and *Prunus* (Baytop et al. 1963). The summer mean temperature is 28 °C, and the winter mean is 3.9 °C in winters. Mean annual precipitation is 713.4 mm (Akman 1990).

## Material and methods

Specimens were collected from 25 different areas between 15 August 2006 and 09 February 2007. The study areas are randomly distributed in four provinces

of Turkey: Batman, Mardin, Osmaniye and Sivas. Lichen samples were dried at room temperature and studied using stereo and compound light microscopes. Secondary metabolites were identified by spot test (Giralt 2001, Halonen et al. 1988, Mayrhofer 1988, Poelt 1974, Purvis et al. 1992, Wirth 1995). All specimens studied were deposited in the herbarium of the Biology Department, Faculty of Sciences and Arts, Karadeniz Technical University.

## Results

The taxonomic survey of Batman, Mardin, Osmaniye and Sivas yielded 205 lichen taxa (198 species, 2 subspecies, 4 varieties, 1 forma) representing 70 genera in 31 families in the *Ascomycota*. Additionally, one lichenicolous fungus was identified. Of these, 146 taxa were collected in Osmaniye, 50 in Mardin, 39 in Sivas, and 24 in Batman. New records per province include 130 lichens and the lichenicolous fungus *Lichenodiplis lecanorae* for Osmaniye, 50 lichens for Mardin, 35 lichens for Sivas, and 23 lichens for Batman.

## Discussion

*Caloplaca flavocitrina*, *Caloplaca variabilis* f. *ocellulata*, *Cladonia metacorallifera*, *Phaeophyscia chloantha* and *Usnea wasmuthii* are new records for Turkey.

Of the 205 taxa, 111 are crustose, 56 foliose, 26 fruticose, and 2 leprose. Further, 86 taxa were epiphytic, 88 saxicolous, eight terricolous, 4 epiphytic or saxicolous, 5 epiphytic or terricolous, and 3 terricolous or saxicolous. Substrates were varied, with 5 taxa growing on decayed barks, 9 on mosses, 10 on limestone, 21 on conifereous trees, and 29 on deciduous trees.

*Caloplaca*, *Cladonia*, *Lecanora*, *Rinodina*, *Verrucaria* and *Physcia* are the common genera in the area researched. Among the common genera, *Caloplaca* is the most diverse (represented by 17 species), with 17 taxa occurring in Mardin and 9 in Sivas. The members of the genus *Cladonia* were only found in Osmaniye. Foliose genera such as *Xanthoria*, *Phaeophyscia*, *Physcia*, *Melanelia*, *Parmelia*, *Melanelixia*, *Melanohalea*, were mostly found in Osmaniye and Sivas regions on mostly deciduous trees i.e. *Carpinus*, *Prunus*, *Pyrus*, *Quercus*, and *Salix*. Saxicolous crustose lichens were found in all the stations and are very common, especially in Osmaniye (Büyük Kuyucak district), Osmaniye (Center); Zorkun high plateau (Sarıüce District), Sivas (İşhanı village), Sivas (Center); Mardin (Midyat); Acırlı village and Batman (Arıca village).

*Aspicilia calcarea*, *Verrucaria dufourii*, *V. calciseda* and *V. muralis*, which are determined as indicator species of lime and calcareous substratum, was found only in Osmaniye: Sarıüce, Yoncalı, Mardin: Midyat, Acırlı village and Batman: Arıca, which are situated near a lime pit.

Species considered sensitive by Purvis et al. (1992) and Wirth (1995), including *Usnea*, *Ramalina farinacea*, *R. calicaris*, *R. fraxinea*, *Physcia aipolia*,

*Lobaria pulmonaria, Pertusaria pertusa, P. amara* and *P. coccodes,* were found in the study area, especially in Osmaniye.

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MYCOTAXON

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# Santa Catarina Island mangroves 1 – First report of *Myxomycetes* on *Avicennia schaueriana*

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**Abstract** — Mangroves are transitional coastal ecosystems linking marine and terrestrial environments, typically found in tropical areas. The total mangrove area in Brazil is one of the largest in the world, where austral limits/borders are in the State of Santa Catarina (Southern Brazil). During a survey about mangrove mycobiota on Santa Catarina Island, there were also collected and identified species of myxomycetes: *Stemonitis fusca*, *S. splendens* and *Physarum pezizoideum*. All specimens were gathered on *Avicennia schaueriana*. These species were collected for the first time in Brazilian mangroves and these are the first records of myxomycetes on *A. schaueriana*.

**Key words** — taxonomy, neotropical

## Introduction

Mangroves are typically tropical vegetal formations located on the transition between terrestrial and aquatic environments, growing along low and protected shores in environments of sedimentation, like estuaries, salt lagoons and deltas (Ayala 2004). Along the American continent's Atlantic coast, the austral limit of mangroves occurrence is at the city of Laguna, located at latitude of 28° 55' S (Cintrón & Schaeffer-Novelli 1980). In Brazil, these ecosystems are well represented, corresponding to one of the six largest mangrove areas in the world (Lacerda 1984).

The occurrence of myxomycetes in mangroves appears to be rare (Nieves-Rivera & Stephenson 2004). Even though the mangrove ecosystems provide a range of potential habitats for myxomycetes, it is possible that regular inundation by the sea inhibit their regular development (Ing 1994).The first mangrove myxomycete species reported was *Arcyria cinerea* (Bull.) Pers., which was collected in 1968 (Oahu, Hawaii) on a branch of *Rhizophora mangle* L. (Kohlmeyer 1969). Afterward, Lee & Baker (1973) reported *Arcyria virescens* G. Lister, *Ceratiomyxa* sp. and *Physarum* sp. from living roots of *R. mangle* at

the same locality. Ing & Hnatiuk (1981) gathered *Physarum globuliferum* (Bull.) Pers. on mangrove poles, in Aldabra Atoll, western Indian Ocean. In Puerto Rico, Nieves-Rivera & Stephenson (2004) reported the occurrence of *Stemonitis splendens* on *R. mangle* in Cabo Rojo.

In spite of myxomycetes' ecological significance for these ecosystems, as organic matter decomposers, only one study on Brazilian myxobiota in mangroves was carried out in northern and northeastern Brazil, in Pernambuco, Sergipe and Pará states (Cavalcanti et al. 2000). In this survey, thirteen taxa, gathered on *R. mangle* and *Laguncularia racemosa* (L.) C.F.Gaertn. mangrove trees were identified.

The myxomycete studies in Santa Catarina state had started by the end of the XIX century, with E. Ule's and A. Möller's collections. A total of 47 taxa of myxomycetes are recorded for Blumenau, Joinville, Itajaí, Tubarão and Florianópolis (Cavalcanti & Fortes 1994). So far, no myxomycete species have been gathered in mangrove areas in the State of Santa Catarina.

The main objective of the present work is to expand the myxobiota knowledge of Brazilian mangroves.

## Materials and methods

Santa Catarina Island is located in the central-east of the State of Santa Catarina (27°35' S and 48°32' W) and belongs to Florianópolis municipality. Mangroves are set up only in the western shores of the island, where there are low-energy environments. The four largest mangroves on the island are located in the following river mouths: Ratones (29° 30' 00" S and 48° 27' 00" W), Saco Grande (28° 37' 30" S and 48° 27' 30" W), Itacorubi (27° 34' 14" S and 48° 30' 07" W) and Tavares (27° 38' 40" S and 48° 30' 17" W).

The typical mangrove trees from these areas are *Avicennia schaueriana* Stapf & Leechm. ex Moldenke, *L. racemosa* and *R. mangle*. The species *A. schaueriana* (also known as black-mangrove or "siriuba") forms phytophysiognomic dominance of these ecosystems (Bigarella 1946, Souza-Sobrinho et al. 1969).

During a survey of xylophilous fungi in Santa Catarina Island mangroves, from May/2005 to August/2006, myxomycete specimens were also collected, always being found on the typical mangrove trees. The specimens were analyzed macro- and micromorphologically, identified using specialized literature and compared with herbarium collections.

Colours are coded according to Munsell (1975). Herbarium names are abbreviated according to the Index Herbariorum (Holmgren et al. 1990). The examined materials are preserved in Herbarium FLOR.

Descriptions, comments, illustrations and a key of the species collected are given, as well as a key for the determination of myxomycete genera collected in mangroves around the world.

## Results and discussion

A total of six myxomycete specimens were collected and identified as: *Stemonitis fusca*, *S. splendens* and *Physarum pezizoideum*. All the species had *A. schaueriana* bark as their substrate.

The three species were gathered from the Itacorubi mangrove and only one *S. splendens* specimen was gathered from the Saco Grande mangrove. There were no myxomycetes collected on the Ratones or the Tavares River mangroves.

### Key for the determination of myxomycete species gathered on Santa Catarina Island mangroves, SC, Brazil

1 Discoid or saucer-shaped sporothecae, deposits of calcium carbonate present, spores 10-13 µm ..... *Physarum pezizoideum*

1' Cylindrical sporothecae, deposits of calcium carbonate absent, spores 8-10 µm ..... 2

2 Sporocarps 9-15 mm high, spore surface with warty-reticulate ornamentation ..... *Stemonitis fusca*

2' Sporocarps 13-19 mm high, spores surface with warty ornamentation ..... *S. splendens*

## Descriptions

***Physarum pezizoideum*** (Jungh.) Pavill. & Lagarde, Bull. Soc. mycol. Fr. 19:87, 1903. FIGURES 1-2

Sporocarps single or caespitose, gregarious, stalked. Sporothecae flat discoid to saucer-shaped, 2.5-4.0 mm high. Peridium squamulose, calcium carbonate present, light grey (5YR 7/1) to reddish grey (10YR 6/1), dehiscence irregular. Capillitium threads hyaline in transmitted light, nodes covered with calcarium. Stalk slender, opaque, striate, 1.5-4.0 mm high, reddish brown (2.5 YR 4/4) to dark reddish brown (2.5YR 3/4). Spores globose, thin-walled, with warty ornamentation on the surface, 10-13 µm, pale violet in 1% KOH. Spore-mass dark reddish brown (5YR 3/2).

SUBSTRATE — *A. schaueriana* dead tree.

DISTRIBUTION — cosmopolitan.

SPECIMENS EXAMINED — BRAZIL. SANTA CATARINA: **Florianópolis**. Ilha de Santa Catarina. Manguezal do Itacorubi. col. Trierveiler-Pereira & Maccarini *161*, 29.I.2006 (*FLOR 31878*). BRAZIL. SANTA CATARINA: **Florianópolis**. Ilha de Santa Catarina. Manguezal do Itacorubi. col. Trierveiler-Pereira & Marcon-Baltazar *191*, 24.II.2006 (*FLOR 31879*).

OTHER SPECIMENS EXAMINED — *P. compressum*: BRAZIL. RIO GRANDE DO SUL: **Porto Alegre**. Morro Santana. col. Rodrigues *081*. 15.III.1984 (*ICN 56693*). BRAZIL. RIO GRANDE DO SUL: **Porto Alegre**. Morro Santana. col. Rodrigues *204*. 29.VI.1984 (*ICN 56816*). BRAZIL. RIO GRANDE DO SUL: **Porto Alegre**. Morro Santana. col. Rodrigues

*272.* 12.IX.1984 (*ICN 56884*). BRAZIL. Rio Grande do Sul: Porto Alegre. Morro Santana. col. Rodrigues *173.* 31.V.1984 (*ICN 56785*). ***P. pezizoideum***: ARGENTINA. col. Castro. 3/V/1974 (*BAFC 23293*).

Comments — The spores of the examined specimens were larger than those described by Farr (1964) and Ukkola & Härkönen (1996). *P. pezizoideum* resembles *Physarum polycephalum* Schwein., but the former has more pronounced spore ornamentation and different stalk morphology (Camino et al. 2005). In the field, this species also resembles *Badhamia gigantospora* Ukkola & Härk. (Ukkola & Härkönen 1996), but that species has larger spores (19-22 µm). Only one record of *P. pezizoideum* is known in the State of Santa Catarina, from the city of Blumenau (Jahn 1902).

***Stemonitis fusca*** Roth, Bot. Mag. (Römer & Usteri) 1(2): 26, 1787. FIGURES 3-4

Sporocarps single, gregarious, stalked. Sporothecae cylindrical, 7-14 mm high, dark brown (7.5YR 3/2). Stalk black (7.5YR N 2/), translucent, hollow, 7-13.5 mm high. Columella central, running to the apex of the sporotheca, then subdividing into several branches. Capillitium dark reddish brown in transmitted light, without calcic deposits. Spores globose, thin-walled, with warty-reticulate ornamentation on the surface, 8-10 µm, purplish brown in 1% KOH. Spore-mass dusky red (2.5YR 3/2).

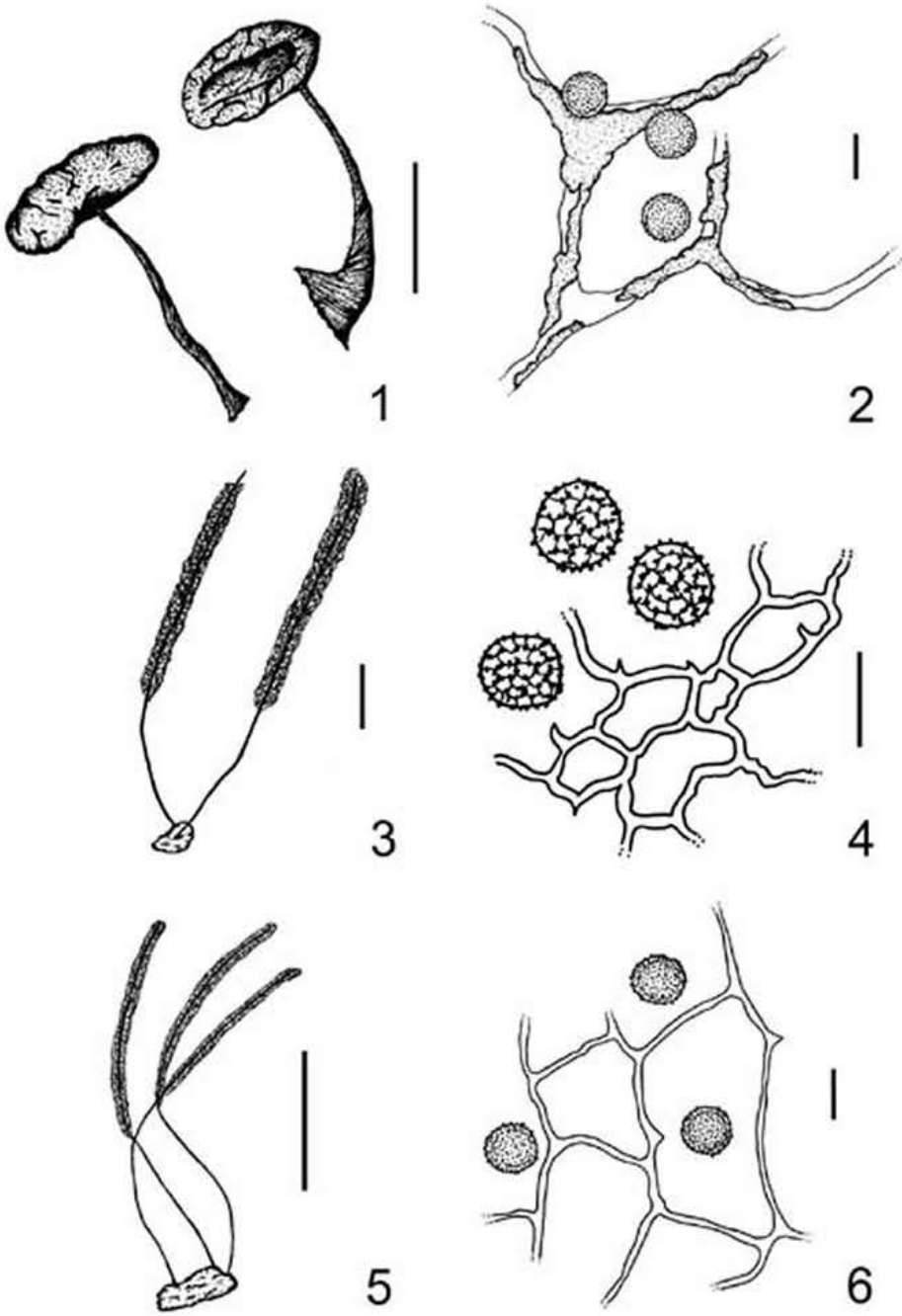
Substrate — *A. schaueriana* dead tree.

Distribution — cosmopolitan.

*Specimens examined* — BRAZIL. Santa Catarina: Florianópolis. *Ilha de Santa Catarina. Manguezal do Itacorubi. col. Trierveiler-Pereira & Name 010. 01.IV.2005 (FLOR 31878).* BRAZIL. Santa Catarina: Florianópolis. *Ilha de Santa Catarina. Manguezal do Itacorubi. col. Trierveiler-Pereira & Marcon-Baltazar 192. 24.II.2006 (FLOR 31879).*

*Other specimens examined* — BRAZIL. Rio Grande do Sul: Porto Alegre. *Morro Santana. col. Rodrigues 081. 15.III.1984 (ICN 56693).* BRAZIL. Rio Grande do Sul: Porto Alegre. *Morro Santana. col. Rodrigues 204. 29.VI.1984 (ICN 56816).* BRAZIL. Rio Grande do Sul: Porto Alegre. *Morro Santana. col. Rodrigues 272. 12.IX.1984 (ICN 56884).* BRAZIL. Rio Grande do Sul: Porto Alegre. *Morro Santana. col. Rodrigues 173. 31.V.1984 (ICN 56785).*

Comments — *S. fusca* is a very ordinary species with very variable morphology (Teixeira 1971). Castillo et al. (1997) concluded that this is a species variable in the size of sporocarp, stalk, capillitium and in colour. The material is similar to other specimens examined, being different only in the darker coloured sporocarps (ICN 56785) and with a slightly smaller spores (ICN 56884). Essentially, the spore ornamentation pattern (warty-reticulate) characterize the species. In the State of Santa Catarina, *S. fusca* has been previously reported from the cities of Blumenau (Jahn 1902), Itajaí (Hennings 1896) and Florianópolis (Cavalcanti & Fortes 1994).




**Figures 1-6. 1-2. *Physarum pezizoideum*.** 1: sporangia (scale bar = 1 mm), 2: capillitium detail and spores (scale bar = 10 µm). **3-4. *Stemonitis fusca*.** 3: sporangia (scale bar = 1 mm), 4: capillitium detail and spores (scale bar = 10 µm). **5-6. *Stemonitis splendens*.** 5: sporangia (scale bar = 5 mm), 6: capillitium detail and spores (scale bar = 10 µm).

***Stemonitis splendens*** Rostaf.,
Śluzowce monogr. (Paryz): 195, 1874 ('1875') FIGURES 5-6

Sporocarps single, gregarious, stalked. Sporothecae cylindrical, 13-19 mm high, dark reddish brown (5YR 3/2) to dark brown (7.5YR 3/2). Stalk black (7.5YR N 2/), translucent, hollow, 13-18 mm high. Columella central, reaching nearly to the sporotheca apex. Capillitium dark reddish brown in transmitted light, without calcaric deposits. Spores globose, thin-walled, with warty ornamentation on the surface, 8-10 µm, purplish brown in 1% KOH. Spore-mass very dark greyish brown (10YR 3/2).

SUBSTRATE — *A. schaueriana* dead tree.

DISTRIBUTION — cosmopolitan.

SPECIMENS EXAMINED — BRAZIL. SANTA CATARINA: **Florianópolis.** Ilha de Santa Catarina. Manguezal do Itacorubi. col. Trierveiler-Pereira & Maccarini *145*. 29.I.2006 (*FLOR 31880*). BRAZIL. SANTA CATARINA: **Florianópolis.** Ilha de Santa Catarina. Manguezal do Saco Grande. Marcon-Baltazar & Regolin *200*. 27.IV.2006 (*FLOR 31881*).

OTHER SPECIMENS EXAMINED — BRAZIL. RIO GRANDE DO SUL: **Porto Alegre.** Morro Santana. col. Rodrigues *294*. 30.XI.1984 (*ICN 56908*). BRAZIL. RIO GRANDE DO SUL: **Porto Alegre.** col. Rodrigues *320*. 12.IV.1986 (*ICN 56934*).

COMMENTS — The spores of the compared material are slightly smaller (7-8 µm), however, Rodríguez-Palma & Estrada-Torres (1996) noticed that *S. splendens* is a highly variable species, showing differences in the size and colour of the sporocarps, mesh size, expansions in the capillitium and columella apex, and the colour, size and ornamentation of the spores. In the State of Santa Catarina, the species has been previously reported from Blumenau (Jahn 1902) and Florianópolis (Cavalcanti & Fortes 1994). In Puerto Rico, it was found growing on *Rhizophora mangle* decayed fallen trunks (Nieves-Rivera & Stephenson 2004).

To facilitate the identification of myxomycete genera gathered in mangrove areas, the following key was made, based on bibliographic references (Teixeira 1971, Rodrigues & Guerrero 1990, Moreno et al. 2001).

**Key for the determination of myxomycete genera on mangrove trees**

1. Pseudocapillitium present and columella absent ..... *Licea*
1' Capillitium and columella present ..... 2
2. Sporocarps stalked and tiny (up to 0.5 mm tall) ..... *Echinostelium*
2' Sporocarps stalked or sessile, taller than 0.5 mm ..... 3
3. Spores produced externally to sporangium. ..... *Ceratiomyxa*
3' Spores produced internally to sporangium ..... 4

4. Sporocarp with some calcarium material ..... 5
4' Sporocarp without calcarium material ..... 7
5. Capillitium with calcium carbonate present ..... *Physarum*
5' Capillitium with calcium carbonate absent. ..... 6
6. Peridium without calcium carbonate ..... *Diachea*
6' Peridium with calcium carbonate ..... *Didymium*
7. Columella present and capillitium without ornamentations. ..... 8
7' Columella absent and capillitium ornamented ..... 10
8. Sporotheca globose, peridium persistent and iridescent ..... *Lamproderma*
8' Sporotheca cylindrical, peridium evanescent, with or without a superficial net arising from the capillitium ..... 9
9. Superficial net present, stalk base fibrous ..... *Stemonitis*
9' Superficial net absent, stalk base not fibrous ..... *Comatricha*
10. Capillitial threads ornamented with well- defined spirals ..... *Hemitrichia*
10' Capillitial threads ornamented with spines, warts, rings or half-rings, reticulations or poorly-defined spirals. ..... 11
11. Capillitial threads short, simple or slightly ramified, rarely forming a complete net. ..... *Perichaena*
11' Capillitial threads long, well-ramified and anastomosed, forming a complete isodiametric net ..... *Arcyria*

## Conclusions

Myxobiota in mangroves is still barely known throughout the world. The present work contributes to this knowledge. These are the first records of myxomycetes in Brazilian mangroves and represents the first occurrences of myxomycetes on *Avicennia schaueriana*.

At this time, sixteen myxomycete taxa are known for Brazilian mangroves, demonstrating an important research field for these organisms.

Inventorying the diversity of organisms, especially communities of lower organisms such as fungi and myxomycetes, supplies information that can be used as tools for conservation and management of mangrove forests.

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# *Myxomycetes* of Sonora, Mexico. 4: Sierra de Alamos-Rio Cuchujaqui Biosphere Reserve

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**Abstract** – Thirty-one species of myxomycetes from the protected area Sierra de Alamos - Rio Cuchujaqui were studied. Fifteen taxa are new records for the Mexican state of Sonora: *Cribraria fragilis, Diachea bulbillosa, D. leucopodia, Diderma effusum, D. hemisphaericum, Fuligo megaspora, Licea castanea, Oligonema schweinitzii, Physarella oblonga, Physarum auriscalpium, P. bogoriense, P. cinereum, P. viride, Stemonaria longa* and *Stemonitis mussooriensis*. Including these new records, eighty-one taxa have now been described for the Sonoran myxobiota. The species studied are discussed and photomicrographs of their macro- and microscopic characters are provided for some of them.

**Key words** – myxomycota, chorology, taxonomy, SEM

## Introduction

The Sierra de Alamos – Rio Cuchujaqui region was decreed as a wildlife protection area in 1996 and a Biosphere Reserve by UNESCO on September 18, 2007. This protected region, which is located at 27°12'–26°53' N and 109°03'–108°29' W, comprises an area of 92,889 ha in southeastern Sonora in the municipality of Alamos. The climate is subhumid, with rain in summer, and the vegetation is mainly microphyllous desert scrub, tropical deciduous forest, oak forest, and pine-oak forest. Tropical dry forest covers 64.5% of the total area of Alamos.

This forest is characterized by high-elevation vegetation with mostly mesophilic and hydromorphic species, along with some spinose shrubs and succulent plants. An outstanding adaptive characteristic of the plants that comprise this type of vegetation is the nutrient reservoir contained in their root system. This makes it possible for the plant to quickly respond to rainfall in summer and also allows plants to survive and grow during drought. The latter phenomenon is documented for trees such as *Bursera laxiflora* S. Watson, *B. stenophylla* Sprague & L. Riley, *Ceiba acuminata* (S. Watson) Rose, *Ficus trigonata* L., *Haematoxylum brasiletto* H. Karst., *Lysiloma watsonii* Rose and *Taxodium mucronatum* Ten., which are found along the Cuchujaqui River. Other plants that are representative of this tropical forest include *Acacia cochliacantha* Willd., *Ambrosia ambrosioides* (Cav.) W.W. Payne, *Croton flavescens* Greenm., *Ipomoea arborescens* (Willd.) G. Don, *Pachycereus pecten-aboriginum* (Engelm.) Britton & Rose, *Pithecellobium sonorae* S. Watson, *Prosopis glandulosa* Torr. and *Vitex mollis* Humb. et al. Some macromycetes have been reported in association with these tropical deciduous forest plants in the Alamos Mountains (Esqueda et al. 1999).

There are 81 taxa of myxomycetes known for Sonora, including the fifteen new records for the Sierra de Alamos – Rio Cuchujaqui region. The distribution of the taxa in Mexico follows Moreno et al. (2007).

## Materials and methods

The collections studied were gathered mainly in the field, but some were cultivated in moist chambers. Samples for light microscopy were mounted in Hoyer's medium and PVA following Schnittler & Novozhilov (1996) and Koske & Tessier (1983). Spores were measured, to include surface structures such as spines or warts, with an oil immersion lens. For ultramicroscopic studies, the material to be examined was rehydrated in concentrated ammonium hydroxide (28–30%) for 30 minutes, dehydrated in aqueous ethanol (70%) for 30 minutes, fixed for 2 hours in pure ethylene glycol dimethyl ether (= 1,2-dimethoxymethane) and finally immersed in pure acetone for at least 2 hours. This was followed by critical point drying and sputtering with gold-palladium.

The micrographs were taken at the University of Alcalá using a Zeiss DSM-950. This technique uses very little material (one sporocarp, a part of it or only a small portion of spores). Terminology used to describe spore ornamentation follows that of Rammeloo (1975a,b). For each species, the locality is included (see details in Table 1); collectors are abbreviated as follows: S. Chacón (SC), J. Cifuentes (JC), M. Esqueda (ME), S. Gómez (SG), M. Lizárraga (ML), E. Pérez-Silva (EPS), M. Rivera (MR), A. Sánchez (AS) and R. Valenzuela (RV); date and herbarium are also given. First records for the Sonoran myxobiota are indicated by an asterisk.

Table 1. Sampling localities in the Sierra de Alamos - Rio Cuchujaqui protected area.

| Localities | N | W | Elevation | Vegetation |
|---|---|---|---|---|
| 1. La Cañita | 26° 59' 31" | 108° 38' 59" | 643 m | TDF, OF |
| 2. El Aguaje | 26° 56' 49" | 108° 41' 41" | 491 m | TDSF, GF |
| 3. El Cuzalito | 26° 58' 33" | 108° 39' 33" | 713 m | OF |
| 4. El Platanar | 26° 59' 27" | 108° 40' 40" | 635 m | TDSF |
| 5. Huerta Vieja | 27° 02' 05" | 109° 02' 54" | 563 m | TDF |
| 6. Mesa del Trigo | 26° 58' 13" | 108° 41' 19" | 583 m | TDSF/SVSH |
| 7. Palo Injerto | 27° 03' 04" | 108° 43' 52" | 399 m | TDF/SVS |
| 8. Promontorios | 27° 00' 53" | 109° 02' 11" | 536 m | TDF |
| 9. San Pedro | 27° 03' 53" | 108° 43' 44" | 444 m | TDF |

TDF: Tropical deciduous forest; OF: Oak forest; TDSF: Tropical deciduous and subdeciduous forest; GF: Gallery forest; SVS: Secondary vegetation of shrubs; SVSH: Secondary vegetation of shrubs and herbs.

## Species List

***Arcyria denudata*** (L.) Wettst., Verh. Zool.-Bot. Ges. Wien 35: Abh. 535 (1886)

Specimens studied: Locality 5, leg. EPS, ML, ME, AS & MR, 21.XI.2005, on the decayed wood of *Ipomoea arborescens*, *CESUES* 7263.

Observations: This species has been cited for the Mexican states of Chiapas, Chihuahua, Jalisco, Morelos, Nuevo Leon, Puebla, Quintana Roo, Sonora, Tabasco, Tamaulipas, Veracruz and Yucatan and is commonly distributed in the tropical and subtropical regions of Mexico.

***Comatricha elegans*** (Racib.) G. Lister, Guide Brit. Mycetozoa, ed. 3: 31 (1909)

Specimens studied: Locality 1, leg. ML, ME, AS & MR, 23.XI.2005, on *Pseudotsuga menziesii* wood, *CESUES* 7276. It was cultivated in a moist chamber (01.XII.2005), fruiting on 20.XII.2005. Ibidem, 10.I.2006, *CESUES* 7284, *CESUES* 7279. Locality 9, on *Pseudotsuga menziesii* wood, leg. ML, ME, AS & MR, 23.XI.2005, *AH* 31917. It was cultivated in a moist chamber (01.XII.2005), fruiting on 15.XII.2005.

Observations: *Comatricha elegans* was recently described for Chihuahua by Lizárraga et al. (2005b). The authors published photomicrographs of sporocarps by transmitted light and spore ornamentation under SEM. In Mexico, its known distribution is Chihuahua, Jalisco, Sonora, Tabasco and Veracruz.

***Comatricha tenerrima*** (M.A. Curtis) G. Lister, Guide Brit. Mycetozoa, ed. 4: 39 (1919)

Specimens studied: Locality 5, leg. EPS, ML, ME, AS & MR, 21.XI.2005, on the decayed wood of *Quercus* sp., *CESUES* 7262.

OBSERVATIONS: *Comatricha tenerrima* is characterized by sporocarps attenuating at the apex and spores 7-8 µm diam., ornamented as baculate under SEM with a stelliform to coralloid apex (Lizárraga et al. 1999a, 2005b, 2007). This taxon has been reported for Baja California, Chihuahua, Guerrero, Jalisco, Quintana Roo, Sonora, Tlaxcala, Veracruz and Yucatan.

****Cribraria fragilis*** Lado & Estrada, in Estrada et al., Mycologia 93(4): 744 (2001) FIGS. 1–5

SPECIMENS STUDIED: LOCALITY 8, leg. ML, ME, SC, RV & JC, 12.IX.2006, on decayed cactus, *AH* 31918.

OBSERVATIONS: The characteristic features of this species are the small sporocarps with violet hues, ca. 0.2-0.3 mm in total height, conspicuous calyculus, absence of peridial net at maturity; spores 8-9 µm diam., subglobose, with conspicuous longitudinal bands which form depression-like valleys where prominent and scattered warts are located visible under SEM; and its succulenticolous habitat. It can be distinguished from *Cribraria violacea* Rex by the globose and warted spores of the latter species; and from *Cribraria zonatispora* Lado et al., by the ellipsoidal shape and inconspicuous verrucose spores of the latter species. *C. zonatispora* also has succulenticolous habitat. It was described as a new species for the Mexican state of Morelos by Estrada et al. (2001).

****Diachea bulbillosa*** (Berk. & Broome) Lister, in Penzig, Myxomyc. Fl. Buitenzorg: 45 (1898) FIGS. 6–7

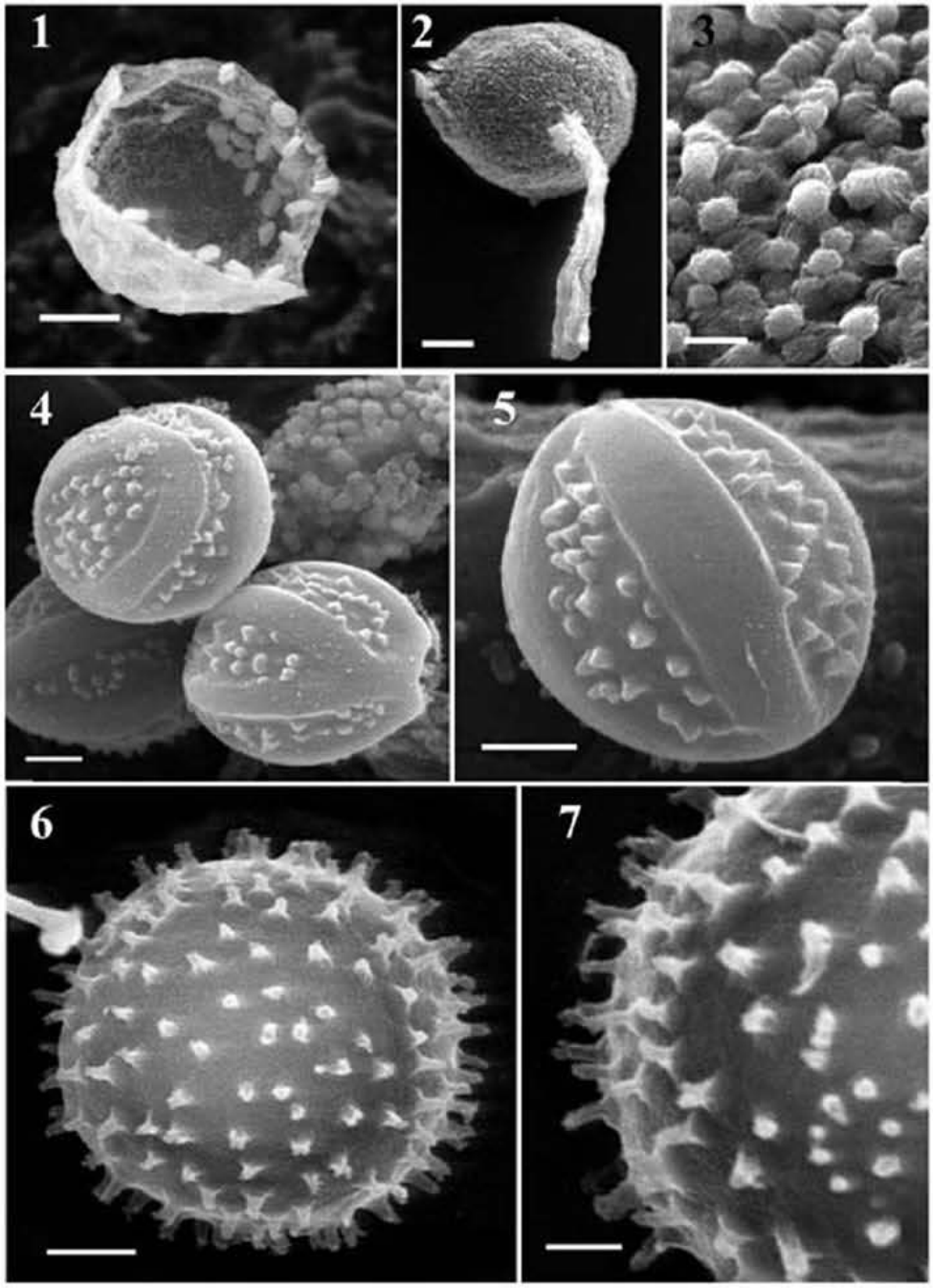
SPECIMENS STUDIED: LOCALITY 4, leg. ML, ME, SC, RV & JC, 14.IX.2006, on dried *Quercus* sp. leaves, *AH* 31919. Ibidem, LOCALITY 2, *CESUES* 7300.

OBSERVATIONS: The distinctive characters of this species are its stalked sporocarps; globose sporotheca; calcareous stalk, whitish to ochraceous-white, rigid and fragile; spores 9-11 µm diam., globose, conspicuously spinose; capillitium with dark purple filaments, (2-)3-4 µm diam., tapering with discoloration at the apex. Mexican collections showed spore ornamentation strongly spinose under SEM as was recently described for Cuban collections by Camino et al. (2005). It was previously recorded for the Mexican states of Jalisco, Quintana Roo and Veracruz.

****Diachea leucopodia*** (Bull.) Rostaf., Sluzowce Monogr.: 190 (1874)

SPECIMENS STUDIED: LOCALITY 2, leg. ML, ME, SC, RV & JC, 14.IX.2006, on dried *Quercus* sp. leaves, *CESUES* 7299.

OBSERVATIONS: This cosmopolitan species was reported for Baja California, Chihuahua, Estado de Mexico, Jalisco, Morelos, Nuevo Leon, Sinaloa, Tlaxcala and Veracruz.




FIGS. 1–5 *Cribraria fragilis* AH 31918: 1. Detail of inner side of calyculus. 2. Detail of outer side of calyculus and stalk. 3. Detail of outer side of calyculus ornamentation. 4. Spores. 5 Spore with longitudinal bands and scattered warts. FIGS. 6–7 *Diachea bulbillosa* CESUES 7300: 6. Spore. 7. Detail of spore ornamentation. Scale bars. 1 = 500 µm. 2 = 20 µm. 3-6 = 2 µm. 7 = 1 µm.

**Diderma effusum* (Schwein.) Morgan, J. Cincinnati Soc. Nat.Hist. 16: 155 (1894)

SPECIMENS STUDIED: LOCALITY 3, leg. ML, ME, SC, RV & JC, 15.IX.2006, on *Quercus* sp. leaves, *CESUES* 7307.

OBSERVATIONS: The distinctive characters of this taxon are the sessile and flattened sporocarps; globose to subglobose spores, 8–10 µm diam., verruculose with faint clusters of larger warts. *Diderma cubense* Berk. & M.A. Curtis is a synonym that was recently confirmed by Camino et al. (2005). These authors studied the type specimens. Although this is a cosmopolitan species (Martin & Alexopoulos 1969), there are few records for Mexico: Chiapas, Chihuahua, Jalisco, Quintana Roo, Veracruz and Yucatan.

**Diderma hemisphaericum* (Bull.) Hornem., Fl. Dan. 33: 13 (1829)

SPECIMENS STUDIED: LOCALITY 8, leg. ML, ME, SC, RV & JC, 12.IX.2006, on *Quercus* sp. leaves, *CESUES* 7289.

OBSERVATIONS: This cosmopolitan species is commonly reported for Mexico: Baja California, Chiapas, Chihuahua, Nuevo Leon, Quintana Roo, Tabasco, Tlaxcala, Veracruz and Yucatan.

*Diderma spumarioides* (Fr.) Fr., Syst. Mycol. 3: 104 (1829)

SPECIMENS STUDIED: LOCALITY 4, leg. ML, ME, SC, RV & JC, on dried *Quercus* sp. leaves, *AH* 31920.

**Observations:** The studied material is characterized by its densely clustered fructifications but with individual sporocarps amongst them, subglobose, whitish, embedded in a hypothallus; peridium thin, calcareous. Spore-mass black, globose, violet hues by transmitted light, 11–12 µm diam., ornamented with verrucae that are regularly distributed on the surface. The features observed in the collections agree with descriptions of Lizárraga et al. (2007). It has been cited for the Estado de Mexico, Jalisco, Quintana Roo, Sonora, and Yucatan.

*Echinostelium apitectum* K.D. Whitney, Mycologia 72(5): 954 (1980) FIG. 8

SPECIMENS STUDIED: LOCALITY 1, leg. ML, ME, AS & MR, 23.XI.2005, on *Prosopis* sp. wood, *AH* 31916. It was cultivated in a moist chamber (01.XII.2005), fruiting on 15.XII.2005.

OBSERVATIONS: Previously reported in Mexico for Baja California Sur, Chihuahua, Sonora, and Tlaxcala.

**Fuligo megaspora* Sturgis, Colorado Coll. Stud. Sci. Ser. 12: 443 (1913) FIG. 9

SPECIMENS STUDIED: LOCALITY 4, leg. ML, ME, SC, RV & JC, 14.IX.2006, on dried *Quercus* sp. leaves, *AH* 31904, *CESUES* 7302.

OBSERVATIONS: This taxon is easily recognized by its whitish aethalia and typically large, dark spores, 15-20 µm diam., ornamented by strong crests, which

are sometimes subreticulate. Under SEM, the spore ornamentation comprises irregular crests, which are occasionally anastomosed with the apex denticulate-verrucose. It has previously been recorded for some Mexican states: Colima, Jalisco, Nuevo Leon, Quintana Roo, Tlaxcala, and Yucatan.

***Fuligo septica*** (L.) F.H. Wigg., Prim. Fl. Holsat.: 112 (1780)

SPECIMENS STUDIED: LOCALITY 1, leg. ML, AS, ME & MR, growing on *Pseudotsuga menziesi* wood at 1.2 m high, *CESUES* 7280.

OBSERVATIONS: A cosmopolitan taxon (Martin & Alexopoulos 1969) that has been widely cited for Mexico (Moreno et al. 2007).

***Hemitrichia calyculata*** (Speg.) M.L. Farr, Mycologia 66(5): 887 (1974)

SPECIMENS STUDIED: LOCALITY 5, leg. EPS, ML, ME, AS & MR, 21.XI.2005, on the decayed wood of *Quercus* sp., *CESUES* 7265. LOCALITY 2, leg. ML, ME, AS & MR, 22.XI.2005, on the decayed wood of *Quercus* sp., *CESUES* 7273.

OBSERVATIONS: This species is regularly reported from Mexico: Baja California, Baja California Sur, Chiapas, Chihuahua, Guerrero, Hidalgo, Jalisco, Morelos, Nuevo Leon, Oaxaca, Quintana Roo, Sinaloa, Sonora, Tabasco, Tamaulipas, Veracruz, and Yucatan. This taxon is common in tropical zones.

***Hemitrichia parviverrucospora*** (Lizárraga, G. Moreno & Illana) G. Moreno & Illana, in Pérez-Silva et al., Mycotaxon 77: 187 (2001)

SPECIMENS STUDIED: LOCALITY 5, leg. EPS, ML, ME, AS & MR, 21.XI.2005, on decayed *Quercus* sp. wood, *CESUES* 7264. LOCALITY 2, on the decayed wood of a palm tree, leg. ML, ME, AS & MR, 22.XI.2005, *CESUES* 7271. Ibidem, on *Quercus* sp. wood, *CESUES* 7272.

OBSERVATIONS: This species is recognized by its capillitium with very spiny ornamentation with spines frequently 4–7 µm long, spores reticulate with small warts within the meshes, visible only under SEM (Lizárraga et al. 1999b, Pérez-Silva et al. 2001). This species has been reported only for Central Africa and Mexico (Baja California Sur, Guerrero, Sinaloa and Sonora).

****Licea castanea*** G. Lister, J. Bot. 49: 61 (1911)

SPECIMENS STUDIED: LOCALITY 9, leg. ML, ME, AS & MR, 24.XI.2005, on *Pseudotsuga menziesii* wood, *AH* 31915. It was cultivated in a moist chamber (24.II.2006), fruiting on 02.III.2006.

OBSERVATIONS: This species is characterized by its minute fructifications, ca. 0.1–0.3 mm in total height, yellowish brown to chestnut brown, dehiscence by polygonal plates; peridium overlaid with abundant granular material, inferior inner surface exhibiting numerous papillae. Spores globose, pale yellow in transmitted light, smooth, thick-walled, and with a conspicuous pale area. It is cited for Chihuahua, Tlaxcala, and Yucatan.

***Lycogala flavofuscum*** (Ehrenb.) Rostaf., in Fuckel, Jahrb. Nassauischen Vereins Naturk. 27–28: 68 (1873) FIGS. 10–12

SPECIMENS STUDIED: LOCALITY 6, leg. ML, ME, SC, RV & JC, 14.IX.2006, on living *Quercus* sp. wood, *AH* 31910. Ibidem, LOCALITY 4, on decayed wood, 21.XI.2005, *CESUES* 7270.

OBSERVATIONS: *Lycogala flavofuscum* is characterized by its pale to colourless pseudocapillitium comprising irregular tubes, 10–35 µm diam., ornamented by small warts or spines and its globose and transparent spores of 4–6 µm in diam, with an incomplete reticulum that is barely perceptible. With the SEM, one can observe that the reticulum is formed of several meshes of different widths, with a mesh-free zone and smooth reticulate walls. Reported for Baja California, Chiapas, Distrito Federal, Guanajuato, Jalisco, Michoacan, Sonora, Tamaulipas, and Tlaxcala.

***Macbrideola decapillata*** H.C. Gilbert, Stud. Nat. Hist. Iowa Univ. 16: 158 (1934)

SPECIMENS STUDIED: LOCALITY 1, leg. ML, ME, AS & MR, 23.XI.2005, on *Quercus* sp. wood, *CESUES* 7277. It was cultivated in a moist chamber (01.XII.2005), fruiting on 9.XII.2005. Ibidem, 10.I.2006, *CESUES* 7283.

OBSERVATIONS: *Macbrideola decapillata* has been reported previously from the Mexican states of Chihuahua, Puebla, and Sonora. This rare species was described and photographed during recent research in Chihuahua (Lizárraga et al. 2003), Puebla (Keller & Braun 1977), and Sonora (Moreno et al. 2006).

***Metatrichia horrida*** Ing, Trans. Brit. Mycol. Soc. 47(1): 51 (1964)

SPECIMENS STUDIED: LOCALITY 5, leg. EPS, ML, ME, AS & MR, 21.XI.2005, on the rotten wood of *Quercus* sp., *CESUES* 7261. LOCALITY 6, on the decayed wood of *Acacia cochliacantha*, leg. ML, ME, AS & MR, 21.XI.2005, *CESUES* 7269.

OBSERVATIONS: A detailed description of this species and its comparison to *Metatrichia vesparium* (Batsch) Nann.-Bremek. ex G.W. Martin & Alexop., was given by Moreno et al. (1997), based on collections from Baja California Sur. Reported for Baja California Sur, Guerrero, Quintana Roo, Sinaloa, Sonora, Veracruz and Yucatan.

****Oligonema schweinitzii*** (Berk.) G.W. Martin, Mycologia 39(4): 460 (1947) FIGS. 13–20

SPECIMENS STUDIED: LOCALITY 2, leg. ML, ME, AS & MR, 23.XI.2005, on the decayed wood of *Quercus* sp, *CESUES* 7274. LOCALITY 7, leg. ML, ME, SC, RV & JC, 13.IX.2006, on rotten wood of *Quercus* sp., *AH* 31908.

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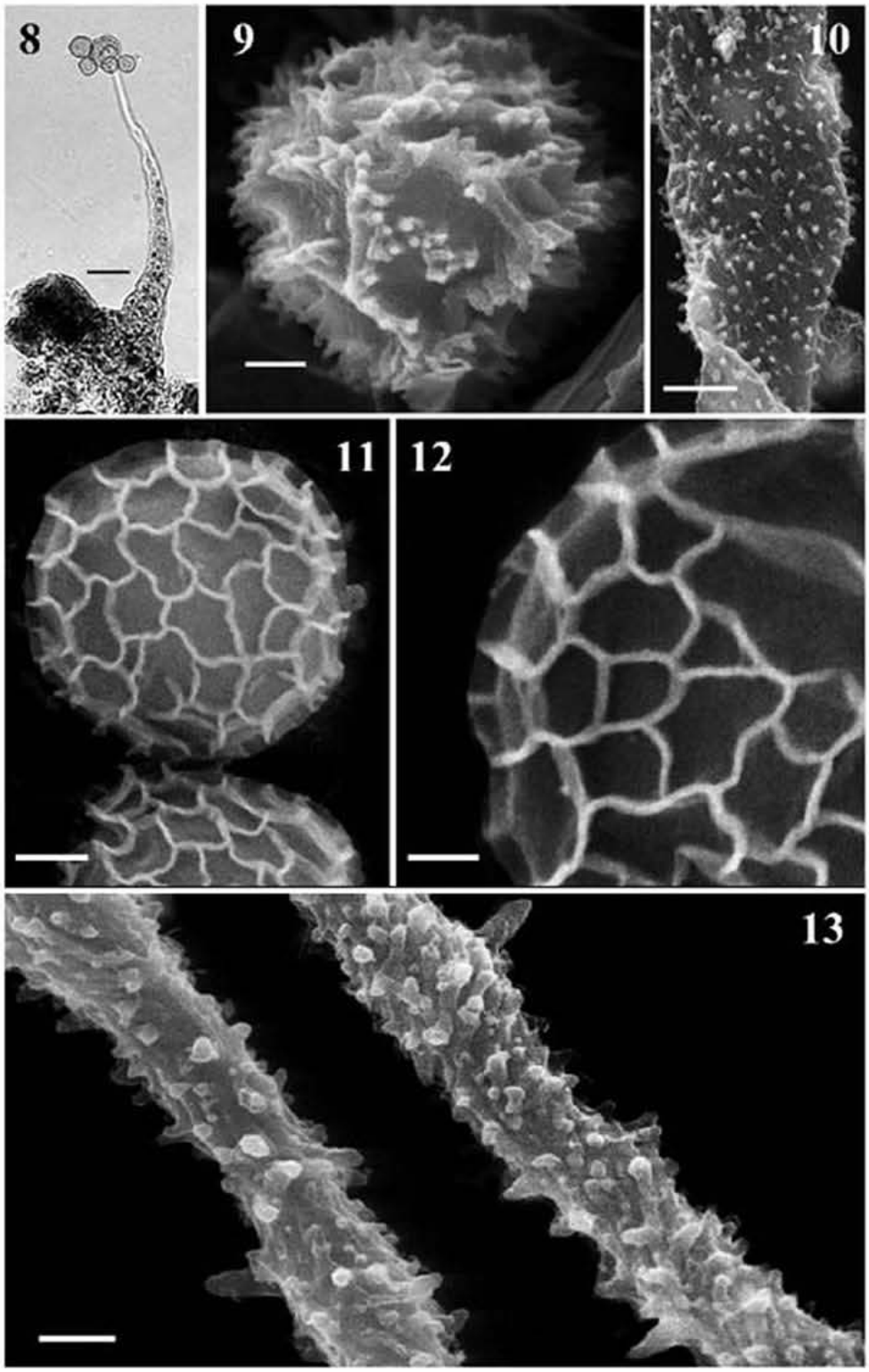
FIG. 8 *Echinostelium apitectum* AH 31916: Fructification and spores. FIG. 9 *Fuligo megaspora* AH 31904: Spore. FIGS. 10–12 *Lycogala flavofuscum* AH 31910: 10. Detail of capillitium. 11. Spore. 12. Detail of spore ornamentation. FIG. 13 *Oligonema schweinitzii* AH 31908: Capillitium.
Scale bars. 8 = 10 µm. 9 and 13 = 2 µm. 10 = 5 µm. 11 = 1 µm. 12 = 0.5 µm.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OBSERVATIONS: *Oligonema schweinitzii* is characterized by its densely clustered, sessile, and subglobose sporocarps sometimes cylindrical to obconical in shape, over 1 mm in length and 0.5-0.9 mm in diam.; peridium is membranous, thin, and shiny, with the inner surface ornamented by dense warts visible with SEM but barely perceptible in transmitted light. Spore-mass yellow, light yellow by OM, globose to subglobose, 10-12 µm in diam, reticulate, with the reticule formed by several wide meshes somewhat irregular in shape. Under SEM, reticule formed of smooth walls, the upper portions of which are composed of smaller meshes of irregular widths. Capillitium is formed by yellow elaters, 3-4 µm in diam., and bearing warts that are conspicuous under the SEM.

The Mexican collection is characterized by its subglobose densely clustered sporocarps; verrucose inner peridium; narrow and wart-bearing capillitium and reticulate spores with meshes conspicuously wide and sometimes fragmented. It differs from the descriptions indicated for this species by authors such as Martin & Alexopoulos (1969), Rammeloo (1984), Neubert et al. (1993), and de Haan (2003) owing to the verrucose capillitium and the lack of smooth elaters that bear fine and smooth dextrose spirals.

A closely related species is *Oligonema flavidum* (Peck) Peck, which also has reticulated spores but with meshes that are closer; thus, the spore morphology is very different compared with Mexican collection, as we can see in the plates by Rammeloo (1984). For this reason we decided to identify it as *O. schweinitzii*. There was only one previous report of the genus *Oligonema* in Mexico: *O. schweinitzii,* which was reported by MacBride (1899) without specifying the locality. Thus, it has been considered as a dubious record in the catalogue of the myxomycetes of Mexico.

***Perichaena depressa*** Lib., Pl. Crypt. Arduenna: 378 (1837)

SPECIMENS STUDIED: LOCALITY 2, leg. ML, ME, AS & MR, 22.XI.2005, on the decaying wood of *Quercus* sp., *CESUES* 7275.

OBSERVATIONS: Cited for Baja California, Baja California Sur, Chihuahua, Morelos, Puebla, Quintana Roo, Sinaloa, Sonora, Tlaxcala, Veracruz, and Yucatan. A cosmopolitan species that is easily recognized by its sporocarps, which are flat and bear a lateral dehiscence line that runs around them.

****Physarella oblonga*** (Berk. & M.A. Curtis) Morgan,
J. Cincinnati Soc. Nat. Hist. 19: 7 (1896)

SPECIMENS STUDIED: LOCALITY 8, leg. ML, ME, SC, RV & JC, 12.IX.2006, on the decayed bark of *Quercus* sp., *CESUES* 7288, *CESUES* 7287.

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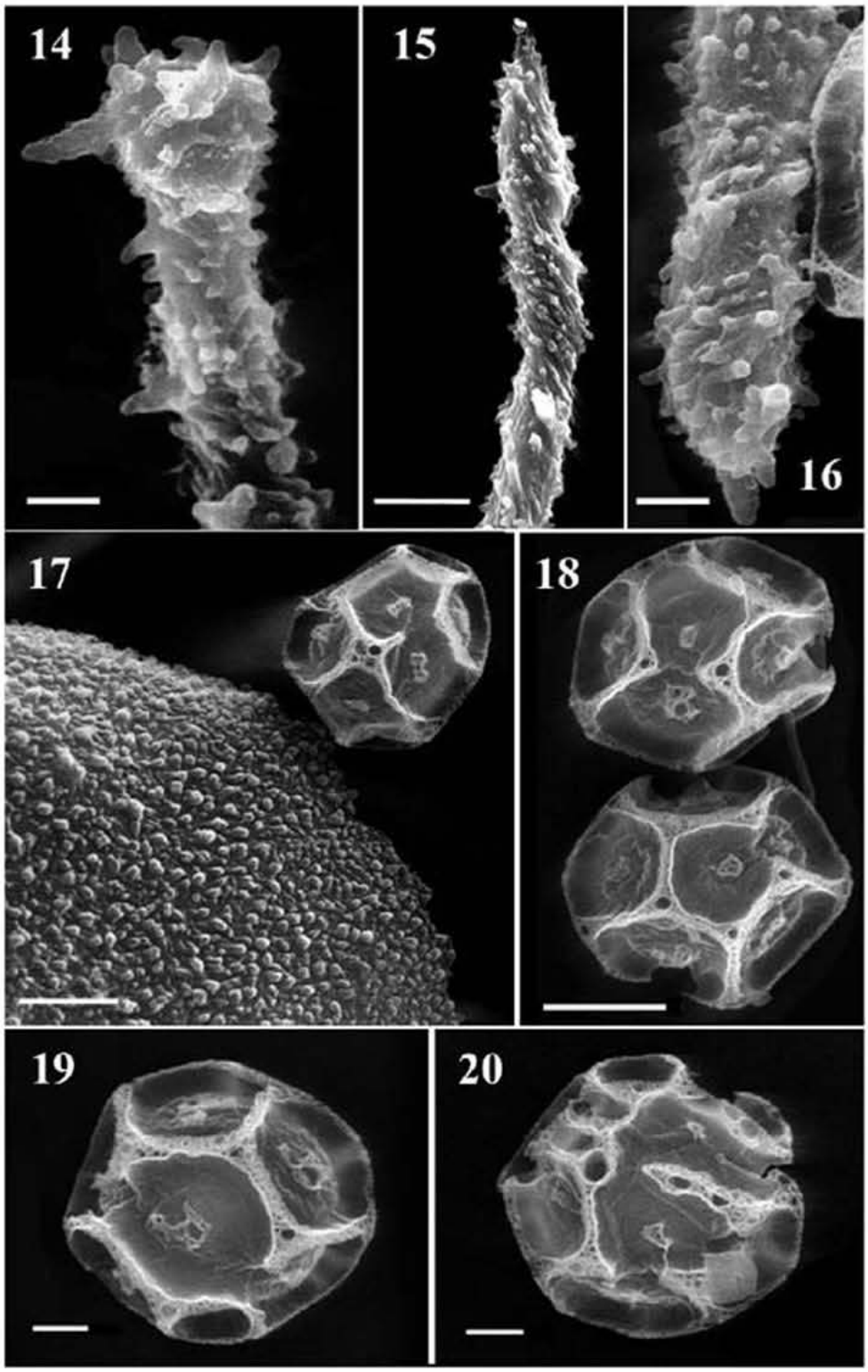
FIGS. 14–20 *Oligonema schweinitzii* AH 31908: 14–16. Detail of the thread ends of the capillitium. 17. Detail of inner side of peridium ornamentation. 18–20. Variation in spore ornamentation. Scale bars. 14, 16, 19-20 = 2 µm. 15, 17-18 = 5 µm.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OBSERVATION: *Physarella oblonga* was cited previously for Chiapas, Quintana Roo, Sinaloa, Veracruz, and Yucatan. This species has a rare tropical distribution in Europe.

* ***Physarum auriscalpium*** Cooke, Ann. Lyceum Nat. Hist. New York 11: 384 (1877)

SPECIMENS STUDIED: LOCALITY 2, leg. ML, ME, SC, RV & JC, 14.IX.2006, on the fallen leaves of *Quercus* sp., *AH* 31921.

OBSERVATIONS: This taxon is recognized by its sessile sporocarps, varying from subglobose to short plasmodiocarps; peridium orange; capillitium physaroid with whitish nodules. Spores 8-9 µm diam., globose, ornamented with spines and small groups of spines. Under SEM, spore ornamentation consists of tight rods in a semi-regular arrangement. It has been cited only for Baja California (Moreno et al. 2007).

* ***Physarum bogoriense*** Racib., Hedwigia 37: 52 (1898) FIGS. 21–22

SPECIMENS STUDIED: LOCALITY 8, leg. ML, ME, SC, RV & JC, 12.IX.2006, on a fallen branch of *Quercus* sp., *AH* 31922.

OBSERVATIONS: *Physarum bogoriense* is characterized by its sporocarps with a double peridium: the external peridium is calcareous, yellow, and dehiscent by patches, while the internal peridium is membranous with an iridescent greyish colour. Spores 7-8 µm in diam., with conspicuous spiky ornamentation and compact groups of thicker spines. Under SEM, the ornamentation is shown as tightly packed rods. Cited from Guerrero, Nuevo Leon, Quintana Roo, Tlaxcala, Veracruz, and Yucatan.

* ***Physarum cinereum*** (Batsch) Pers., Neues Mag. Bot. 1: 89 (1794)

SPECIMENS STUDIED: LOCALITY 7, leg. ML, ME, SC, RV & JC, 13.IX.2006, on bark of *Quercus* sp., *AH* 31914.

OBSERVATIONS: This species has fructifications that vary from globose sporocarps to short plasmodiocarps, whitish, with abundant pale nodules at the capillitium that are more or less subglobose in shape. Spores 9-10 µm in diam., violet in mass and light violet by transmitted light with groups of larger and darker warts. Cited from Baja California, Chihuahua, Guerrero, Jalisco, Nuevo Leon, Quintana Roo, Tlaxcala, Veracruz, and Yucatan. It is a cosmopolitan species (Martin & Alexopoulos 1969).

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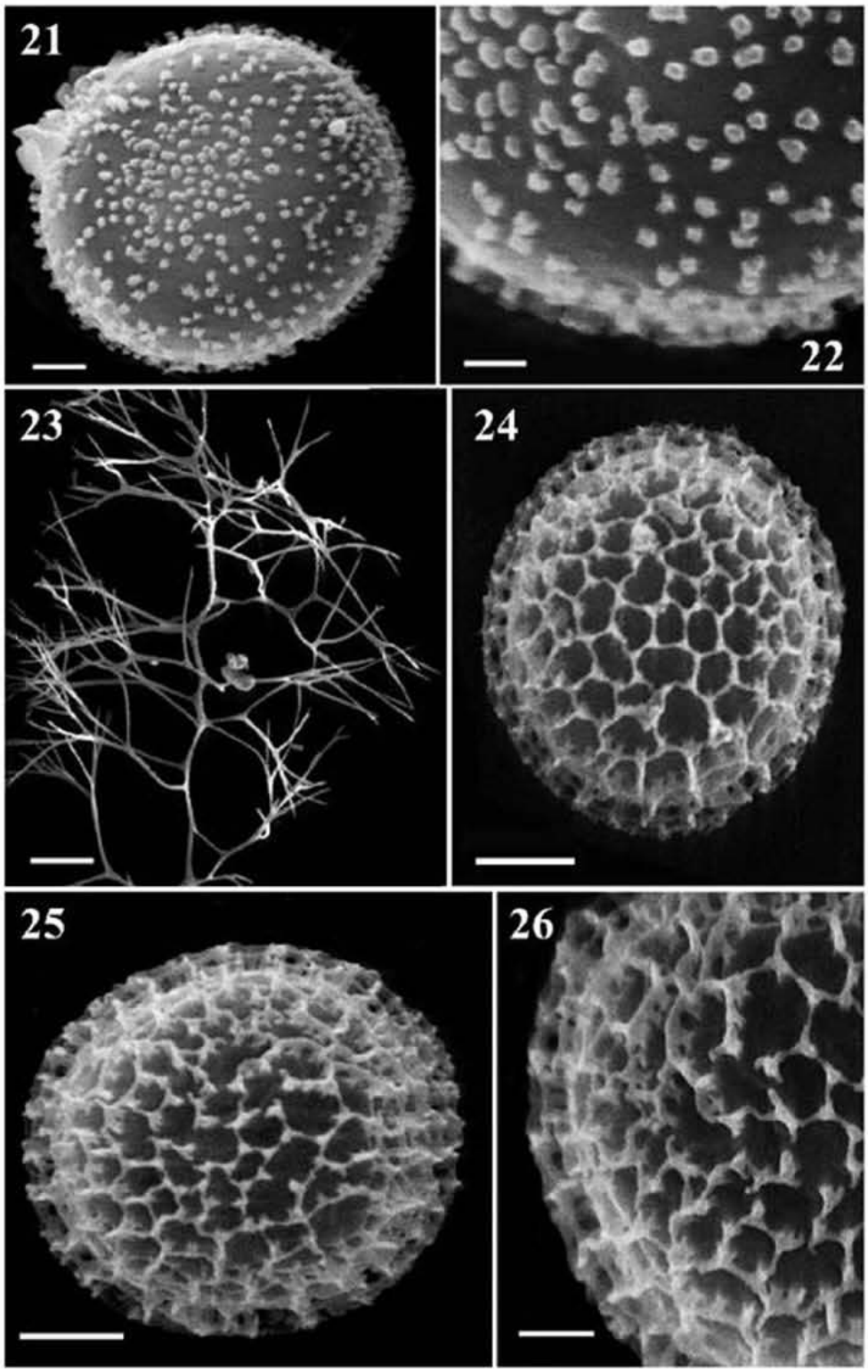
FIGS. 21–22 *Physarum bogoriense* AH 31922: 21. Spore. 22. Detail of spore ornamentation. FIGS. 23–26 *Stemonaria longa* AH 31905: 23. Detail of the thread ends of the capillitium. 24–25. Variation in spore morphology. 26. Detail of spore ornamentation.

Scale bars. 21 and 26 = 1 µm. 22 = 0.5 µm. 23 = 20 µm. 24-25 = 2 µm.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***Physarum compressum*** Alb. & Schwein., Consp. Fung. Lusat.: 97 (1805)

SPECIMENS STUDIED: LOCALITY 7, leg. ML, ME, SC, RV & JC, 13.IX.2006, on wood remains of *Quercus* sp., *CESUES* 7293.

OBSERVATIONS: This cosmopolitan species has been reported for Baja California Sur, Chiapas, Jalisco, Morelos, Nuevo Leon, Quintana Roo, Sinaloa, Sonora, and Veracruz.

***Physarum globuliferum*** (Bull.) Pers., Syn. Meth. Fung.: 175 (1801)

SPECIMENS STUDIED: LOCALITY 6, leg. ML, ME, AS & MR, 21.XI.2005, on the decomposing wood of *Acacia cochliacantha*, *CESUES* 7268.

OBSERVATIONS: *Physarum globuliferum* is characterized by its stalked sporocarps; globose, whitish, sporotheca, with columella; calcareous whitish stalk; spores 7–8 µm diam., verrucose, with groups of more conspicuous and thicker warts on its surface. *P. tenerum* is a closely related species that differs from the former by its columella in a globose sporotheca, a calcareous stalk that is brownish-yellow and larger spores, which are 8–11 µm in diam. Previously reported from Chiapas, Quintana Roo, Sonora, Veracruz, and Yucatan.

****Physarum viride*** (Bull.) Pers., Ann. Bot. (Usteri) 15: 6 (1795)

SPECIMENS STUDIED: LOCALITY 6, leg. ML, ME, SC, RV & JC, 14.IX.2006, on the rotten wood of *Quercus* sp., *CESUES* 7306.

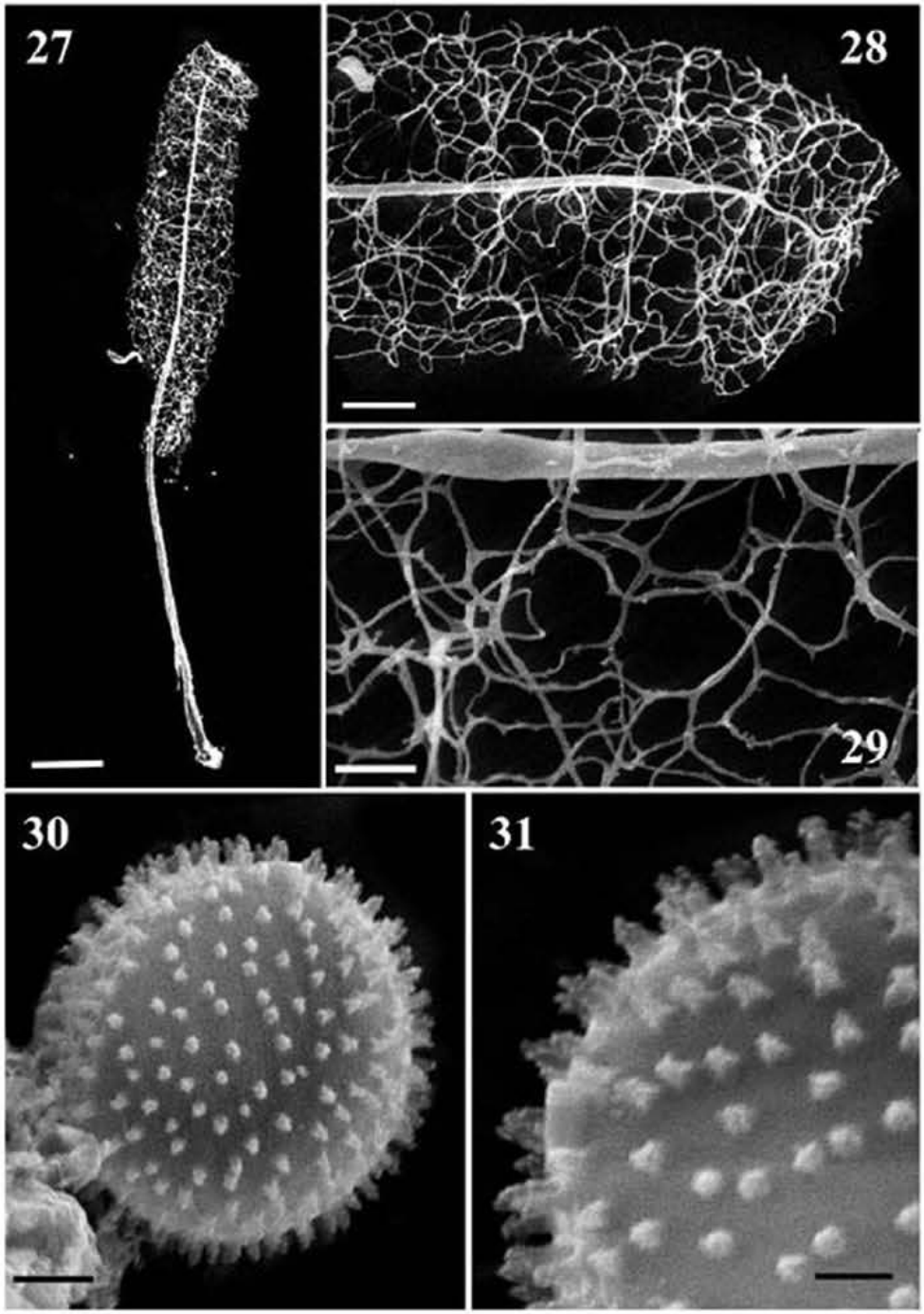
OBSERVATIONS: This cosmopolitan species is cited for Chihuahua, Estado de Mexico, Guerrero, Nuevo Leon, Quintana Roo, Tlaxcala, Veracruz, and Yucatan.

****Stemonaria longa*** (Peck) Nann.-Bremek., R. Sharma & Y. Yamam., in Nannenga-Bremekamp et al., Proc. Kon. Ned. Akad. Wetensch., C. 87(4): 453 (1984) FIGS. 23–26

≡ *Comatricha longa* Peck, Annual Rep. New York State Mus. 43:70 (1890)

SPECIMENS STUDIED: LOCALITY 9, leg. ML, ME, SC, RV & JC, 13.IX.2006, on the decomposing remains of *Polyporaceae*, *CESUES* 7291. Ibidem, LOCALITY 2, 14-IX-2006, on the decomposing wood of *Quercus* sp., *AH* 31905, *CESUES* 7298.

OBSERVATIONS: This species is characterized by its quite long, pendulous sporocarps (more than 10 mm long), and by its open capillitium, with free branches and dichotomous terminations; spores 8-10 µm in diam., globose, which appear verrucose and reticulated by transmitted light. Under SEM, the reticulum is clearly observed, plus abundant meshes and pierced walls. This species has been cited for Chiapas, Jalisco, Quintana Roo, Veracruz, and Yucatan.




FIGS. 27–31 *Stemonitis mussooriensis* AH 31923: 27. Sporotheca. 28. Detail of sporocarp apex. 29. Detail of capillitium. 30. Spore. 31. Detail of spore ornamentation.

Scale bars. 27 = 200 µm. 28 = 50 µm. 29 = 20 µm. 30 = 2 µm. 31 = 1 µm.

***Stemonitis fusca*** Roth, Bot. Mag. (Römer & Usteri) 1(2): 26 (1787)

SPECIMENS STUDIED: LOCALITY 5, on wood remains of *Quercus* sp., leg. ML, ME, EPS, AS & MR, 21.XI.2005, *CESUES* 7260.

OBSERVATIONS: Previously reported for Baja California, Baja California Sur, Chiapas, Chihuahua, Jalisco, Nuevo Leon, Puebla, Quintana Roo, Sonora, Tabasco, Tamaulipas, Tlaxcala, Veracruz, and Yucatan. This cosmopolitan species is frequently found in Mexico.

****Stemonitis mussooriensis*** G.W. Martin, K.S. Thind & Sohi, Mycologia 49(1): 128 (1957) FIGS. 27–31

SPECIMENS STUDIED: LOCALITY 5, leg. ML, ME, SC, RV & JC, 12.IX.2006, on cortex of a liana, *AH* 31923.

OBSERVATIONS: This apparently rather rare species is characterized by its stalked fructifications 1.5–2.2 mm in total height, fruiting in small groups. Sporotheca 1.1–1.6 x 0.3–0.4 mm, cylindrical, dark brown. Peridium evanescent. Stalk short, about one third the total height of the sporocarp. Membranous hypothallus. Capillitium violet-brown which forms an inner and loose net with few free terminations; it may break towards the apex, and bears a more or less abundant number of spines. Central columella tapering towards the apex. Spore mass brownish-black, dark violet by transmitted light, 10–12 µm in diam., globose, conspicuously spiny ornamented, occasionally bearing a pale coloured area. Under SEM, spore ornamentation appears to be composed of long rods with irregular apexes and slightly denticulate.

This collection has been compared to the type material of *S. mussooriensis*, with which it coincides in all characters. There is only one report of *S. mussooriensis* cf., belonging to Veracruz (Lado et al. 2003), which reported smaller spores that did not exceed 10 µm in diam., with spines about 0.5 µm long.

***Trichia affinis*** de Bary, in Fuckel, Jahrb. Nassauischen Vereins Naturk. 23–24: 336 (1870)

SPECIMENS STUDIED: LOCALITY 2, leg. ML, ME, SC, RV & JC, 14.IX.2006, on the woody remains of *Quercus* sp., *CESUES* 7301.

OBSERVATIONS: A SEM study in which the elaters and spore ornamentation can be appreciated for the Sonoran collections was done by Pérez-Silva et al. (2001). This species has been cited for Quintana Roo, Sonora, and Yucatan.

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MYCOTAXON

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# *Acaulosporaceae* from El Palmar National Park, Entre Ríos, Argentina

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**Abstract** — The occurrence of arbuscular mycorrhizal fungi belonging to the genus *Acaulospora* in El Palmar National Park (Entre Ríos Province, Argentina) is reported. In this work *A. entreriana* sp. nov. is described and *A. denticulata*, *A. dilatata*, *A. elegans*, *A. foveata* and *A. rehmii* are reported and illustrated for the first time in Argentina. The distribution area of *A. bireticulata*, *A. delicata*, *A. excavata*, *A. laevis*, *A. mellea*, *A. scrobiculata* and *A. spinosa* was enlarged.

**Key words** — *Glomeromycota*, taxonomy, palm forest

## Introduction

El Palmar National Park is one of the most floristically diverse national parks in Argentina. With over 700 vascular plant species (Biganzoli et al. 2001), the park shelters an important concentration of palm trees, *Butia yatay* (Mart.) Becc., that represents a characteristic edaphic community and one of the last *B. yatay* remnants in the region. Such protected areas have proved to serve as propitious places to conserve floristic biodiversity and associated microorganisms (Hawksworth 1991).

Amongst those symbiotic with land flora, arbuscular mycorrhizal fungi (AMF), which associate with 82% of plant species, are the most common soil fungi (Wang & Qiu 2006). Their ubiquity alone makes them an important component in the soil microbial biomass, and they are directly involved in crucial processes at the plant-soil interface (Harley & Smith 1983, McGee et al. 1989).

The abundance and diversity of AMF in El Palmar National Park is still unknown, although the park's protected environment makes it ideal for

studying these microorganisms. Also, the park offers a wide range of vegetation including scrubland, marsh, palm forest, grassland and gallery forest. In brief, it is clear that vegetation and associated microorganisms within such areas should be protected (APN 1994).

Arbuscular mycorrhizal fungi represent four orders (Schüssler 2007) within the class *Glomeromycetes* (Cavalier-Smith 1988) and phylum *Glomeromycota* (Schüssler et al. 2001). Currently the *Glomeromycota* comprises 10 families and 13 genera (http://AMF-phylogeny.com 2007).

Of the arbuscular mycorrhizal genera in El Palmar National Park, *Acaulospora* species are the most dominant. Gerdemann & Trappe (1974) segregated the genus *Acaulospora* from *Endogone* to accommodate the type species, *Acaulospora laevis*, and another species included in their first key. Berch (1985) later added a third species and emended the generic description.

*Acaulospora* species possess a sporiferous saccule that is developmentally and structurally indistinguishable from the saccule produced by *Archaeospora* species (Morton & Redecker 2001). However, phylogenetic analyses suggest that this structure, which first appeared in *Archaeosporaceae*, was conserved within the *Acaulosporaceae* clade but lost during evolution of *Glomeraceae* (Redecker et al. 2000). Currently accepted *Acaulospora* species include 18 with smooth spore surfaces and 15 with ornamented outer spore walls. Oehl et al. (2006) provide the most recent key to the ornamented *Acaulospora* species.

The aim of this work is to describe and illustrate *Acaulospora* species recorded for the first time for Argentina and expand the known distribution for species previously cited for the country.

## Materials and methods

### Study area

Soil samples were taken from El Palmar National Park (58° 17' W, 31° 50' S), Entre Rios Province, Argentina.

We selected 5 sites with different characteristics: 1-SCRUBLAND, dominated by *Baccharis dracunculifolia* and *Eupatorium buniifolium*; 2-MARSH displayed a different proportion of *Cyperaceae* and *Gramineae* were the following genera *Scirpus*, *Andropogon*, *Bromus* predominate; 3-PALM FOREST *Butia yatay* palm savannah; 4-GRASSLAND principal type physiognomical with grass and herbs of variable height and density; 5-GALLERY FOREST with exotic species as *Melia*, *Ligustrum*. Rhizospheric soil samples were collected using a composite random sampling method. Each sample contained three replicates.

### Trap culture

In each of the 5 sites, three trap cultures were set up following Oehl et al. (2003). An autoclaved substrate consisting of a mixture of soil:perlite:vermiculite (3:1:1, w/w/w)

was used. The AMF inocula containing 20 g per trap plant of unsterilized soil from field samples were placed in 27x17x20 cm pots.

*Lolium perenne, Medicago sativa* and *Plantago lanceolata* seeds were surface-sterilized with sodium hypochlorite (10% v/v) for 10 min and thoroughly rinsed with sterilized water. After germination, seedlings were selected for uniform size and then transplanted into pots, three of each species per trap culture. These pots were placed in a greenhouse at 24 ± 1°C day/ 20 ± 1°C night, and a 16-h photoperiod was provided by incandescent and cool-white lamps; the plants were fertilized with a nutritive solution (Cabello 1997).

### Sampling of trap cultures

Trap cultures were grown for 90 days, then 2 soil core samples (15 $cm^3$; sampling depth, 10 cm) were taken from each pot for the extraction of AMF spores.

### AMF spore isolation and identification

AMF were studied by spore extraction from soil samples or trap culture. Spores were extracted by wet-sieving and decanting (Gerdemann & Nicolson 1963) and the modified sucrose density gradient centrifugation method of Walker et al. (1982).

Fungal spores were examined using an optic microscope and mounted on polyvinyl-lactic acid-glycerine (PVLG) (Koske & Tessier 1983) or PVLG mixed 1:1 (v/v) using Melzer's reagent (Brundrett et al. 1994). Scanning electron microscope (SEM) was used to observe the different ornamentation in spore walls. Specimens were compared with original species described at the Germoplam Bank Institute Spegazzini, La Plata, Argentina; reference isolates described by the Internacional Culture Collection of Arbuscular and Vesicular-Arbuscular Micorrhizal Fungi (INVAM, USA, http://invam.caſwvu.edu), and Błaszkowski, http://www.agro.ar.szczecin.pl/~jbłaszkowski/. Terminology of the spore structure follows Stürmer & Morton (1999), as adapted by INVAM and modified by Oehl et al. (2006).

Figures 9–10 were photographed using a digital Olympus camera (model SP-350) on a stereoscopic microscope (Olympus SZ61); Figures 3–5, 7–8, and 11–14 were photographed on a Leitz Dialux 20EB compound microscope. Photographs (fig. 1, 2 and 6) were taken on a scanning microscope. Spores were prepared for scanning electron microscopy by rinsing them in distilled water, piercing them with a fine needle, and allowing them to air-dry on a metal stub. Dried spores were sputter-coated with gold/palladium and observed with a Jeol JSM-6360 LV microscope. Specimens mounted in PVLG and a mixture of PVLG and Melzer's reagent were deposited at LPS (La Plata, Spegazzini) herbarium.

## Results

Nineteen species belonging to *Acaulosporaceae* were identified. Twelve spore morphotypes could be unequivocally assigned to species of *Acaulospora*, of which one was found to be a new species and six spore types could not be identified to species level.

## Taxonomy

*1. Acaulospora bireticulata* E.M. Rothwell & Trappe,
Mycotaxon 8: 472. 1979 FIGS. 1-2

MATERIAL EXAMINED: ARGENTINA. Entre Rios: *El Palmar National Park.* VIII-2004, *Velázquez.* Isolated from soil sample coming from scrubland, marsh, palm forest, grassland and pot cultures associated with *L. perenne, M. sativa* and *P. lanceolata.* LPS, slide N° 47994.

DISTRIBUTION AND HABITAT: *A. bireticulata* has a worldwide distribution. Rothwell & Trappe (1979) originally recovered spores of this species from soil sample collected under *Sassafras albidum* growing in Kentucky. Miller et al. (1985) isolated this fungus from the root zone of *Malus domestica* in Michigan. Błaszkowski (1989, 1997) reported it in Poland. Lugo & Cabello (1999) in autochthonous mountain grassland in Central Argentina and Schalamuk et al. (2006) also reported it for agroecosystems associated with wheat crops.

GENERAL NOTES: This material agreed with the original descriptions of *A. bireticulata.*

*2. Acaulospora delicata* C. Walker, C. M. Pfeiffer & Bloss,
Mycotaxon 25: 622. 1986

MATERIAL EXAMINED: ARGENTINA. Entre Rios: *El Palmar National Park.* V-2004, *Velázquez.* Isolated from soil sample from scrubland, marsh, palm forest, grassland and gallery forest and pot cultures associated with *L. perenne, M. sativa* and *P. lanceolata.* LPS, slide N° 47995.

DISTRIBUTION AND HABITAT: *A. delicata* has been cited from *Celtis tala* and *Scutia buxifolia* forests in Argentina (Irrazabal et al. 2004) and from a wheat monoculture (Schalamuk et al. 2006).

GENERAL NOTES: This material agreed with the original descriptions of *A. delicata.*

*3. Acaulospora denticulata* E. Sieverd. & Toro,
Angewandte Botanik 61: 217. 1987 FIG. 3

SPORES single in the soil, and laterally on the neck of a sporiferous saccule, pale orange-brown to dark orange-brown, globose to subglobose; 120-180 µm diam.

SUBCELLULAR STRUCTURE OF SPORES consists of 3 spore walls and 2 inner germinal walls.

SPORE WALL composed of 3 layers ***(swl1-3).*** *LAYER 1* ***(swl1)*** hyaline, 0.6-1.5 µm thick, and sloughing detached from sporiferous saccule. *LAYER 2* ***(swl2)*** 0.6-0.8 µm thick, with stubby yellow-brown "knobs" attached. Circular to oblong projections arise from the "knobs" and have a smooth outer edge or form a polygon of 6 sides, each projection has a central cavity. *LAYER 3* ***(swl3)*** a single

hyaline layer, <0.8 µm thick, which is detectable only when it separates from the spore wall.

GERMINAL WALL 1 ***(gw1)*** formed by 2 layers ***gw1l1*** 0.4-0.6 µm thick and ***gw1l2*** 1.2-1.4 µm thick. This wall is clearly visible because it separates readily from the spore wall.

GERMINAL WALL 2 ***(gw2)*** consists of 2 adherent layers. *LAYER 1* ***(gw2l1)*** 0.6-1.0 µm thick, with granular excresences. *LAYER 2* ***(gw2l2)*** "amorphous" 3-10 µm thick in PVLG, staining red-purple to dark red-purple in Melzer's reagent.

SPORIFEROUS SACCULE hyaline, mostly globose, 130-160 µm diam.

CICATRIX not observed.

MATERIAL EXAMINED: ARGENTINA. Entre Rios: *El Palmar National Park*. IX-2004, *Velázquez*. LPS, slide N° 47996.

DISTRIBUTION AND HABITAT: *A. denticulata* was isolated from soil collected in Cauca, Colombia (Sieverding & Toro 1987). *A. denticulata* was found associated with woody and herbaceous vegetation in soils of El Palmar National Park. *A. denticulata* was found in soil samples emerging from 4 treatments, scrubland, marsh, palm forest and gallery forest. The spore density of *A. denticulata* in soil samples was 2-4 in 100 g dry soil. The fungi accompanying *A. denticulata* were *Glomus etunicatum* W. N. Becker & Gerd. and *Glomus sp.*

MYCORRHIZAL ASSOCIATIONS: *A. denticulata* was associated with woody and herbaceous vegetation.

GENERAL NOTES: *A. denticulata* was not recovered in trap culture. *A. denticulata* was reported by Menendez et al. (2001) in natural and cultivated grasslands in Argentina. However, this species seemed to be *Entrophospora infrequens* (I. R. Hall) R. N. Ames & R. W. Schneid. rather than *A. denticulata* as shown in the figure.

***4. Acaulospora dilatata*** J. B. Morton, Mycologia 78: 641. 1986 FIGS. 4-5

SPORES single in the soil, and laterally on the neck of a sporiferous saccule, pale yellow to brown, mostly globose to subglobose; 100-130 µm diam.

SUBCELLULAR STRUCTURE OF SPORES consists of 1 spore wall and 2 inner germinal walls.

SPORE WALL composed of 3 layers ***(swl1-3)***. *LAYER 1* ***(swl1)*** hyaline, <0.5 µm thick, and sloughing, showing a clear detachment from the sporiferous saccule. *LAYER 2* ***(swl2)*** laminate, smooth, pale yellow-brown, 2.8-5.5 µm thick. At maturity, the pore between spore and saccule neck is closed by continuous sublayers of this layer, sealing in spore contents. *LAYER 3* ***(swl3)*** pale yellow-brown, 1-4 µm thick, and somewhat flexible when it is separated from layer 2 of the spore wall. This layer also appears to have sublayers (laminae) which can

separate more readily from each other than those of layer 2, forming folds that resemble numerous separate "inner walls".

GERMINAL WALL 1 ***(gw1)*** 2-layered hyaline wall that separates easily from the spore wall and thus it can be seen readily. Layers separate slightly in many spores but they can also be adherent and appear as a single layer. *LAYER 1* ***(gw1l1)*** <0.5 µm thick, folding slightly when separated from layer 2. *LAYER 2* ***(gw1l2)*** 0.5-1.0 µm thick. This wall has some inherent rigidity because it breaks with the spore wall.

GERMINAL WALL 2 ***(gw2)*** consists of 2 adherent hyaline layers. *LAYER 1* ***(gw2l1)*** 0.5-1.2 µm thick, presents granular excrescences. *LAYER 2* ***(gw2l2)*** plastic, amorphous, 1.0 µm thick, becoming red purple to dark red purple in Melzer's reagent.

SPORIFEROUS SACCULE hyaline, mostly globose to subglobose, occasionally irregular, 100-135 µm diam. Saccule wall consist of 1 layer with smooth surface, 1.0-1.4 µm thick.

CICATRIX very narrow lip circumscribing the scar, circular to oval-shaped.

MATERIAL EXAMINED: ARGENTINA. Entre Rios: *El Palmar National Park*. V-2004, *Velázquez*. LPS, slide N° 47997.

DISTRIBUTION AND HABITAT: *A. dilatata* was found in 2 coal minesoils in Preston; also on a roadside embankment with acid mine drainage near Sabraton (Morton 1986). *A. dilatata* was found associated with woody and herbaceous vegetation in soils of El Palmar National Park. *A. dilatata* was found in soil samples and trap cultures from 5 treatments, scrubland, marsh, palm forest, grassland, and gallery forest. The spore density of *A. dilatata* in soil samples was 44-156 in 100 g dry soil, the gallery forest evidenced the highest density, whereas in trap culture was 3-21 in 100 g dry soil. The fungi accompanying *A. dilatata* were *G. etunicatum* and *Glomus sp.*

MYCORRHIZAL ASSOCIATIONS: *A. dilatata* was associated with woody and herbaceous vegetation and trap cultures with *L. perenne, M. sativa,* and *P. lanceolata.*

GENERAL NOTES: *A. dilatata* spores resembled *Acaulospora lacunosa* J.B. Morton in colour, size and shape, but *A. lacunosa* presented a distinctive ornamentation in the external wall.

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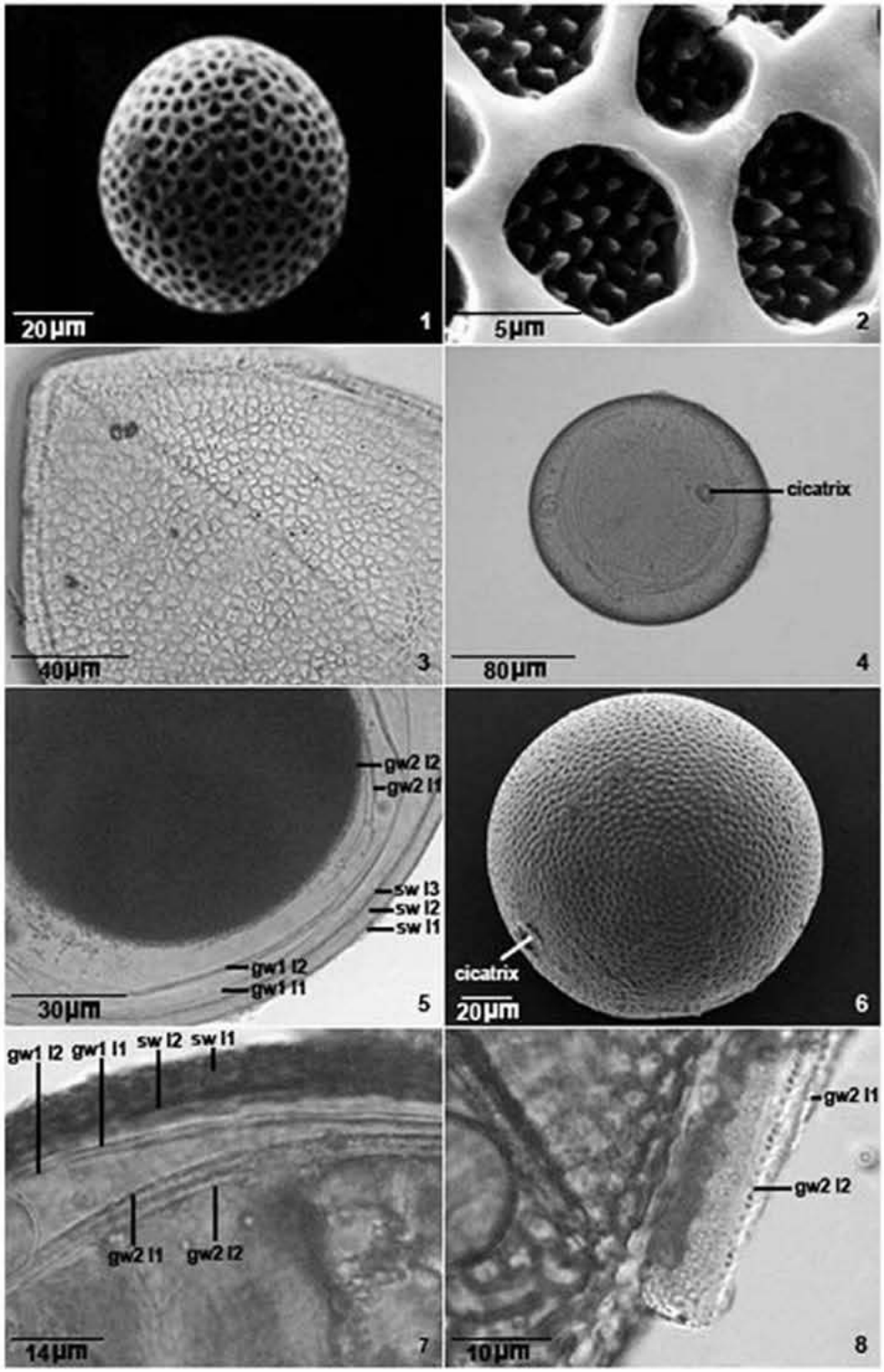
**Figures 1-2.** *Acaulospora bireticulata.* **Fig. 1.** Mature spore. Scanning electron microscopy. **Fig. 2.** Ornamented wall. Scanning electron microscopy. **Figure 3.** *A. denticulata,* ornamented wall. **Figures 4-5.** *A. dilatata.* **Fig. 4.** Mature spore with cicatrix. **Fig. 5.** Broken spores with 3-layered spore wall (*sw*l1-3), and 2 inner germinal walls (*gw*1*l*1-2 and *gw*2*l*1-2). *Gw*2*l*2 becoming red purple to dark red purple in Melzer's reagent. **Figures 6-8.** *A. elegans.* **Fig. 6.** Mature spore with cicatrix. Scanning electron microscopy. **Fig. 7.** Broken spores with 2-layered spore wall (*sw*l1-2), and 2 inner germinal walls (*gw*1*l*1-2 and *gw*2*l*1-2). **Fig. 8.** Bilayered germinal wall 2 (*gw*2*l*1-2) with granular excrescences.


20 µm
1
5µm
2
40µm
3
cicatrix
80µm
4
gw2 l2
gw2 l1
sw l3
sw l2
sw l1
gw1 l2
gw1 l1
30µm
5
cicatrix
20µm
6
gw1 l2
gw1 l1
sw l2
sw l1
gw2 l1
gw2 l2
14µm
7
gw2 l1
gw2 l2
10µm
8

**5. *Acaulospora elegans*** J.W. Trappe & Gerd., Mycologia Memoir 5: 34. 1974 FIGS. 6-7-8

SPORES single in the soil, and laterally from the neck of a sporiferous saccule. Dark brown, globose to subglobose; 140-280 x 145-330 µm diam.

SUBCELLULAR STRUCTURE OF SPORES consists of 1 spore wall and 2 inner germinal walls.

SPORE WALL composed of 2 layers ***(swl1-2)***. *LAYER 1* ***(swl1)*** hyaline, continuous with the wall of the hyphal neck subtending the saccule. *LAYER 2* ***(swl2)*** consisting of laminae within which spines are crowded. Light brown spines 2.0 µm wide x 0.5 µm high, with total layer as much as 12 µm. An alveolate reticulum of hyaline ridges is superimposed on the spines. Alveoli are 4-8 µm long.

GERMINAL WALL 1 ***(gw1)*** consists of 2 thin adherent flexible layers ***(gw1l1-2)***, which almost appear to be of equal thickness, presenting granular excresenses.

GERMINAL WALL 2 ***(gw2)*** consists of 2 hyaline layers ***(gw2l1-2)***, granular excresenses, the outher layer thinner than the inner 1, which produces a red-brown reaction in Melzer's reagent.

SPORIFEROUS SACCULE pale brown, mostly globose to ellipsoid, 150-240 µm diam.

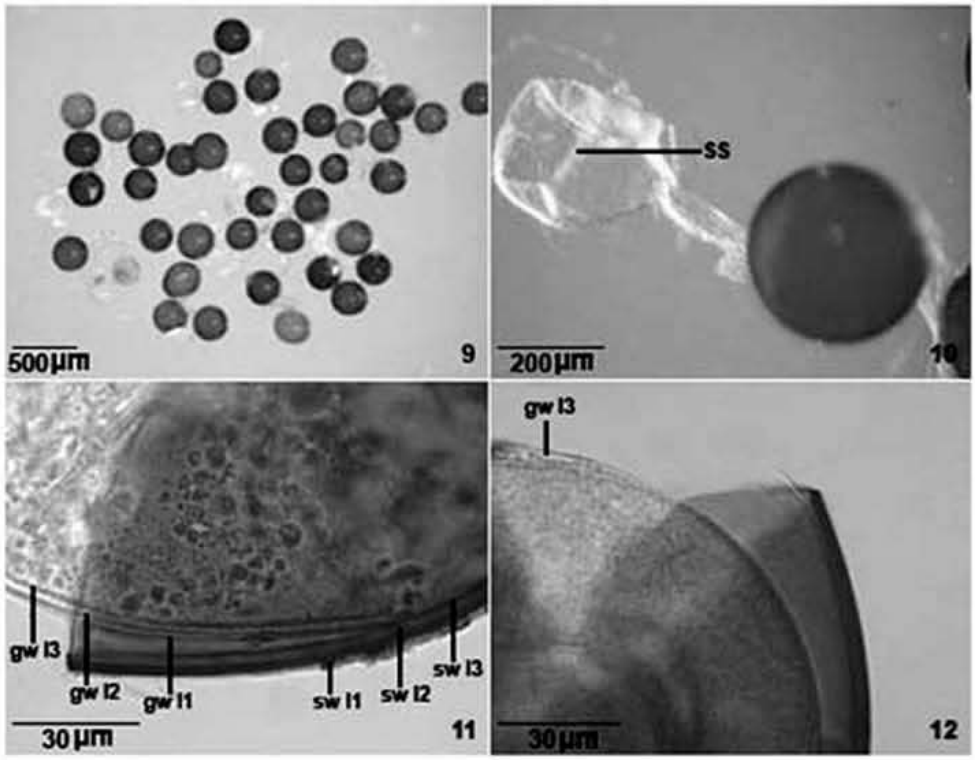
CICATRIX not observed.

MATERIAL EXAMINED: ARGENTINA. Entre Ríos: *El Palmar National Park*. XI-2006, *Velázquez*. LPS, slide N° 47998.

DISTRIBUTION AND HABITAT: *A. elegans* has a widely distributed in coastal sands of northern California to southwestern Washington (Gerdemann & Trappe 1974). *A. elegans* was found in trap cultures containing soil from marsh of El Palmar National Park. The spore density of *A. elegans* in trap culture was 53 in 100 g dry soil. The fungus accompanying *A. elegans* was *G. etunicatum*.

MYCORRHIZAL ASSOCIATIONS: *A. elegans* was associated in trap cultures with *L. perenne, M. sativa* and *P. lanceolata*.

GENERAL NOTES: spores of *A. elegans* resembled *A. bireticulata* except for pattern of ornamentation in layer 2 (swl2) of the spore wall. Gerdemann & Trappe (1974) described the spore wall as a 3-layered reticulum over "angular processes" 1 µm high, whereas the spore wall of *A. elegans* was described as a single-layered reticulum over crowded spines only 0.5 µm high. Therefore, concerning these differences these species would be considered as separate. The germinal wall ornamentation in El Palmar National Park *A. elegans* spores is quite different from that characterized by INVAM. Spores found in El Palmar exhibited granular excresences in 2 germinal walls (*gw*1-2) whereas INVAM spores are only in *gw2l2*.




**Figures 9-12.** *Acaulospora entreriana.* **Figure 9.** Spores. **Figure 10.** Spore laterally on the neck of a sporiferous saccule (ss). **Figure 11.** Broken spores with 3-layered spore wall (*swl*1-3) and 3-layered germinal wall (*gwl*1-3). **Figure 12.** Broken spores with *gwl*3 ornamented with teeth.

**6. *Acaulospora entreriana*** M.S. Velázquez & Cabello, **sp. nov.** Figs. 9–12

MycoBank MB 511190

*Sporae singulare in solo efformtae, lateraliter gestae ad collum sacculi sporangiferi; brunneae, globosae, subglobosae vel ovoideae 260-300 x 280-330 µm diam. Sacculus sporifer hyalinus, globosus vel subglobosus, aliquando irregularis 240-280 µm diam. Sporae tunicis duobus, tunica exterior et interior. Tunica exterior in totum 4,5 µm crassa, stratis tribus: stratum exterius tenue et evanescens, hyalinum; stratum medium laminatum, brunneum; stratus interius hyalinum. Tunica interior stratis tribus 3,6 µm in totum, hyalina, stratis uno hyalino minutis granulatis; stratis duobus hyalino; stratis tribus conspicue dentata, dentibus 0,5µm longis. Formans vesicular-arbuscular mycorrhizae.*

ETYMOLOGY: *entreriana,* referring to Entre Ríos Province where the species was first found.

TYPUS: isolated from trap culture at the Institute of Botany Spegazzini, Universidad Nacional de La Plata, Argentina. XI-2006, *Velázquez.* Holotypus LPS, slide N° 47993.

SPORES single in the soil, and laterally on the neck of a sporiferous saccule, brown, globose to subglobose; 280-300 µm diam, sometimes ovoid; 260–300 x 280-330 µm.

SUBCELLULAR STRUCTURE OF SPORES consists of 1 spore wall and inner germinal wall. Neither spore nor germinal walls react in Melzer's reagent.

SPORE WALL composed of 3 layers ***(swl1-3)***, 4.5 µm thick. *LAYER 1* ***(swl1)*** evanescent, hyaline, 0.9 µm thick, rarely present in mature spores, continuous with the wall of a soporiferous saccule. *LAYER 2* ***(swl2)*** smooth, light brown to dark brown, (1.2-)2.7(-4.6) µm thick. *LAYER 3* ***(swl3)*** smooth, composed of 3–4 tightly attached laminae, hyaline, (0.7)1.0(-2.7) µm thick, commonly adhered to layer 2.

GERMINAL WALL 3-layered, hyaline, ***(gwl1-3)*** 3.6 µm thick. *LAYER 1* ***(gwl1)*** smooth, hyaline, covered with small beads, (0.7-)2.7(-1.2) µm thick. *LAYER 2* ***(gwl2)*** smooth hyaline. *LAYER 3* ***(gwl3)*** hyaline 0.9-1.5 µm thick in PVLG, ornamented with teeth 0.5 µm high.

SPORIFEROUS SACCULE hyaline, mostly globose to subglobose, occasionally irregular, 240-280 µm diam. Distance from saccule to spore 100-150 µm.

CICATRIX not observed.

DISTRIBUTION AND HABITAT: *A. entreriana* was found in trap cultures built up with soil from scrubland, palm forest, grassland, and gallery forest of El Palmar National Park. The spore density of *A. entreriana* in trap culture was 4-200 in 100 g dry soil, the scrubland evidenced the highest density. The fungi accompanying *A. entreriana* were *A. delicata, E. infrequens, Glomus claroideum* N. C. Schenck & G. S. Sm. and *G. etunicatum*.

MYCORRHIZAL ASSOCIATIONS: Forms vesicular arbuscular mycorrhizae. *A. entreriana* was associated in trap cultures with *L. perenne, M. sativa* and *P. lanceolata*.

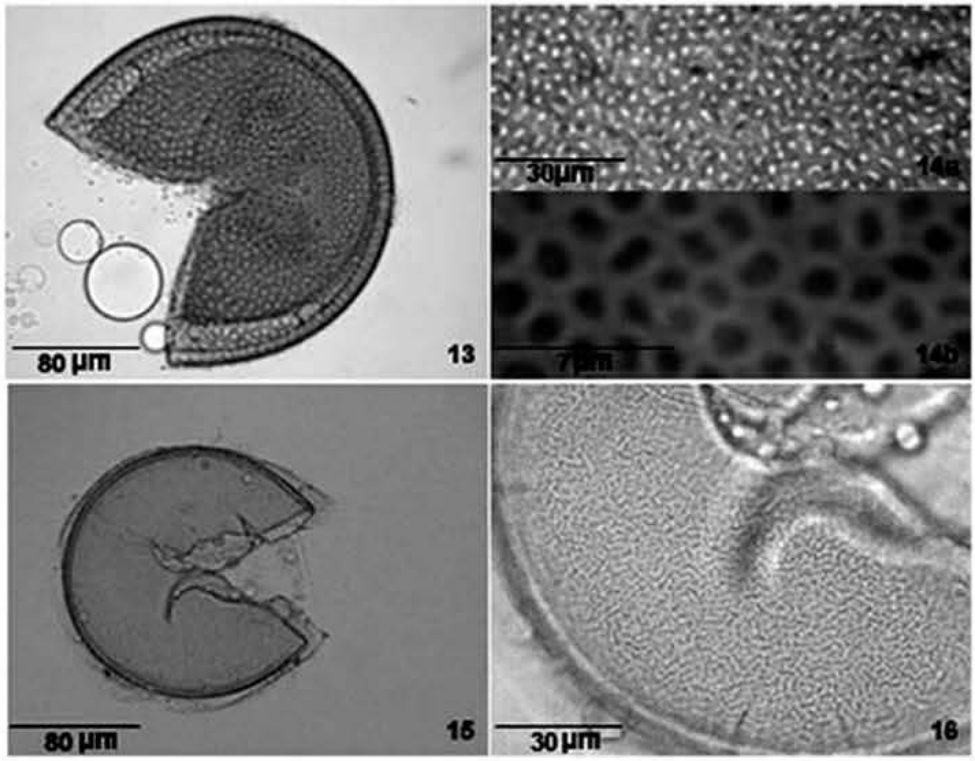
GENERAL NOTES: In colour and shape, *A. entreriana* spores resemble those of *Acaulospora koskei* J. Błaszk. (Błaszkowski 1995), except larger and with an ornamented inner wall. The wall in *A. entreriana* did not react in Melzer's, whereas the smooth wall of *A. koskei* turned red-purple in the same reagent. Both *A. entreriana* and *A. splendida* E. Sieverd. et al. are the only species known in *Acaulospora* that have a single germinal wall, a character that is very important considering that germinal walls are involved in germination. *A. entreriana* walls do not react in Melzer reagent, a character shared with *A. polonica* J. Błaszk.

*A. entreriana* spores were not recovered in soil samples.

***7. Acaulospora excavata*** Ingleby & C. Walker, Mycotaxon 50: 100. 1994

MATERIAL EXAMINED: ARGENTINA. Entre Ríos: *El Palmar National Park*. VIII-2004, *Velázquez*. Isolated from soil sample from scrubland, marsh and palm forest. LPS, slide N° 47999.

DISTRIBUTION AND HABITAT: this species was found for the first time at Ivory Coast, Africa. Later it was found in Central Argentina mountain grasslands (Lugo & Cabello 1999) and agroecosystems (Schalamuk et al. 2006).




**Figures 13-14.** *Acaulospora foveata.* **Figure 13.** Mature spore in Melzer's reagent. **Figure 14 a-b.** Ornamented wall. **Figures 15-16.** *A. rehmii.* **Figure 15.** Broken spores. **Figure 16.** Broken spores with labyrinthiform ornamentation.

GENERAL NOTES: This species was not recovered on trap plants. This material agrees with the original descriptions of *A. excavata.*

*8. Acaulospora foveata* J. M. Trappe & Janos,
Mycotaxon 15: 516. 1982 FIGS. 13-14 a, b

SPORES single in the soil, and laterally on the neck of a sporiferous saccule, red-orange to dark red-orange, globose to subglobose; 240-320 µm diam.

SUBCELLULAR STRUCTURE OF SPORES consists of 1 spore wall and 2 inner germinal walls.

SPORE WALL composed of 3 layers ***(swl1-3).*** *LAYER 1* ***(swl1)*** hyaline, 2.0 µm thick, rarely present in mature spores. *LAYER 2* ***(swl2)*** 9-18 µm thick at maturity. Initially a single layer, forms in undulations that establish the shape and depth of surface concave circular to ovoid depressions (8-12 µm across and 0.5-3 µm deep in the mature layer). *LAYER 3* ***(swl3)*** 3-5 µm thick, produces a dark red-brown reaction in Melzer's reagent.

GERMINAL WALL 1 ***(gw1)*** consists of 2 adherent layers ***(gw1l1-2),*** both are of near-equal thickness, ranging from 0-6-1.0 µm thick.

GERMINAL WALL 2 *(gw2)* consists of 2 adherent layers *(gw2l1-2)*, produces pinkish red to a reddish-purple reaction in Melzer's reagent.

SPORIFEROUS SACCULE not observed.

CICATRIX not observed.

MATERIAL EXAMINED: ARGENTINA. Entre Ríos: *El Palmar National Park.* VIII-2007, *Velázquez.* LPS, slide N° 48000.

DISTRIBUTION AND HABITAT: *A. foveata* was found in wet tropical forests in Mexico, Costa Rica and Panama (Janos & Trappe 1982). *A. foveata* was found in trap cultures built up with soil from palm forest of El Palmar National Park. The spore density of *A. foveata* in trap culture was 6 in 100 g dry soil. The fungi accompanying *A. foveata* were *A. delicata* and *G. etunicatum.*

MYCORRHIZAL ASSOCIATIONS: *A. foveata* was associated in trap cultures with *L. perenne, M. sativa* and *P. lanceolata.*

GENERAL NOTES: *A. foveata* spores were not recovered in soil samples. This material agreed with the original descriptions of *A. foveata.*

***9. Acaulospora laevis*** J. W. Gerd. & Trappe, Mycologia Memoir 5: 33. 1974

MATERIAL EXAMINED: ARGENTINA. Entre Ríos: *El Palmar National Park.* V-2004, *Velázquez.* Isolated from soil sample from scrubland, marsh, palm forest, grassland and gallery forest. LPS, slide N° 48001.

DISTRIBUTION AND HABITAT: *A. laevis* has a worldwide distribution. Abundant from the coast of northern California to Washington. Also reported from Florida, Australia, New Zeland, Pakistan and Scotland. *A. laevis* was found associated with mountain grasses in Central Argentina (Lugo et al. 1999) and with *C. tala* and *S. buxifolia* forests (Irrazabal et al. 2004).

GENERAL NOTES: *A. laevis* spores were not recovered on trap plants.

***10. Acaulospora mellea*** Spain & N. C. Schenck, Mycologia 76: 689. 1984

MATERIAL EXAMINED: ARGENTINA. Entre Ríos: *El Palmar National Park.* V-2004, *Velázquez* Isolated from soil sample from scrubland, marsh, palm forest, grassland and gallery forest and pot cultures associated with *L. perenne, M. sativa* and *P. lanceolata.* LPS, slide N° 48002.

DISTRIBUTION AND HABITAT: *A. mellea* was found in Poland in uncultivated and cultivated soils (Błaszkowski 1993) and associated with dune plants (Błaszkowski et al. 2002). In Central Argentina, it was found associated with mountain grassland (Lugo & Cabello 1999) and agroecosystems (Schalamuk et al. 2006).

GENERAL NOTES: Our material agreed with the original descriptions of *A. mellea.*

***11. Acaulospora rehmii*** Sieverd. & S. Toro,
Angewandte Botanik 61: 219. 1987 Figs. 15-16

SPORES single in the soil, and laterally on the neck of a sporiferous saccule, pale yellow to light brown, globose to subglobose 90-160 µm diam.

SUBCELLULAR STRUCTURE OF SPORES consists of 1 spore wall and 2 inner germinal walls.

SPORE WALL composed of 3 layers ***(swl1-3)***. *LAYER 1* ***(swl1)*** evanescent, hyaline, 1.0 µm thick, continuous with the wall of a sporiferous saccule, rarely present in mature spores. *LAYER 2* ***(swl2)*** laminate, pale yellow, 4-10 µm thick, ornamented with labyrinthiform folds. *LAYER 3* ***(swl3)*** semiflexible, hyaline, 0.5 µm thick, rarely separated from layer 2.

GERMINAL WALL 1 **(gw1)** consists of 2 tightly adherent hyaline, semiflexible layers. *LAYER 1* ***(gw1l1)*** and ***layer 2 (gw1l2)*** reach 0.5-1.0 µm thick. These layers tightly adhere to each other and hence are quite difficult to differentiate.

GERMINAL WALL 2 **(gw2)** composed of 2 adherent layers. *LAYER 1* ***(gw2l1)*** flexible, hyaline, 0.5-1.0 µm thick, covered with granules. *LAYER 2* ***(gw2l2)*** flexible, hyaline, 0.6-1.4 µm thick in PVLG.

SPORIFEROUS SACCULE hyaline, globose to subglobose, 90-150 µm diam, usually collapsed and detached in mature spores.

CICATRIX not observed.

MATERIAL EXAMINED: ARGENTINA. Entre Ríos: *El Palmar National Park*. IX-2006, *Velázquez*. LPS, slide N° 48003.

DISTRIBUTION AND HABITAT: *A. rehmii* was originally isolated from a crop field at Caicedonia, Valle de Cauca, Colombia (Sieverding & Toro 1987). This fungus has been associated with cassava, beans, sorghum and *Crotalaria* species. Additionally, Sieverding & Toro (1987) found *A. rehmii* spores in a culture containing a soil sample from Brasilia, Brazil. Wu et al. (1995) isolated *A. rehmii* spores from among roots of *Phyllostachys pubescens* growing in a garden at Chitou Experimental Station, National Taiwan University.

*A. rehmii* was found in trap cultures containing soil from the El Palmar National Park palm forest. Its trap culture spore density was 6 per 100 g dry soil. The fungi accompanying *A. rehmii* were *G. etunicatum* and *Glomus sp.*

MYCORRHIZAL ASSOCIATION: *A. rehmii* was associated in trap cultures with *L. perenne, M. sativa* and *P. lanceolata.*

GENERAL NOTES: *A. rehmii* is a species easily recognized by the unique labyrinthiform ornamentation of its layer 2 spore wall.

***12. Acaulospora scrobiculata*** J. M. Trappe, Mycotaxon 6: 363. 1977

MATERIAL EXAMINED: ARGENTINA. Entre Ríos: *El Palmar National Park.* V-2004, *Velázquez.* Isolated from soil sample from scrubland, marsh, palm forest, grassland and gallery forest and trap cultures associated with *L. perenne, M. sativa* and *P. lanceolata.* LPS, slide N° 48004.

DISTRIBUTION AND HABITAT: *A. scrobiculata* was originally described from spores collected in Mexico (Trappe 1977). This fungus has a global distribution and is found in many localities in the U.S.A. (Friese & Koske 1991, Gemma & Koske 1989, Koske 1987, 1988), Canada (Dalpé 1989), Australia (Koske 1975), China (Zhang et al. 1992), Taiwan (Wu & Chen 1986), and Argentina (Lugo & Cabello 1999).

GENERAL NOTES: The spores of our material correspond in colour, form and size with those originally described.

***13. Acaulospora spinosa*** C. Walker & Trappe, Mycotaxon 12: 515. 1981

MATERIAL EXAMINED: ARGENTINA. Entre Ríos: *El Palmar National Park.* V-2004, *Velázquez.* Isolated the soil sample from scrubland, marsh, palm forest, grassland and gallery forest. LPS, slide N° 48005.

DISTRIBUTION AND HABITAT: This fungus has a worldwide distribution and is found in many localities in the U.S.A. and Mexico. *A. spinosa* was found in a mountain grassland in Central Argentina (Lugo & Cabello 1999). Irrazabal et al. (2004) also record it from *C. tala* and *S. buxifolia* forests.

GENERAL NOTES: *A. spinosa* spores were not recovered in trap plants.

## Discussion

These species of *Acaulosporaceae* were identified in field samples and in trap culture; a total of 19 species were detected in El Palmar National Park. *Acaulospora* diversity in El Palmar National Park was found to be higher than that reported for other ecosystems (Lugo & Cabello 1999, 2002, Lugo et al. 1999, Irrazabal et al. 2004) or agroecosystems (Albornoz & Catania 1996, Menéndez et al. 2001, Schalamuk et al. 2006) in Argentina. AM fungal communities dominated by *Acaulospora* spp. have been reported in other forested ecosystems (Helgason et al. 1998, Merryweather & Fitter 1998a, b), and in alluvial soils (Lovelock et al. 2003).

The decrease of anthropoid disturbance in protected areas such as El Palmar National Park is well known to markedly favour diversity conservation (Jasper et al. 1991). On the other hand, heterogeneity of the environment involving scrubland, marsh, palm forest grassland and gallery forest play an essential role in keeping the abundance and distribution of these fungi.

## Acknowledgements

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# *Hymenoscyphus ginkgonis* sp. nov. growing on leaves of *Ginkgo biloba*

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**Abstract** – An interesting *Hymenoscyphus* species was collected on the leaves of *Ginkgo biloba* in Korea. Morphological characteristics and sequence analysis of the ITS rDNA indicate that the fungus represents a distinct new species, named here as *Hymenoscyphus ginkgonis*.

**Key words** – *Helotiaceae*, ITS rDNA, sequence analysis, taxonomy

## Introduction

*Ginkgo biloba* L. ("ginkgo tree"), the only extant species within the genus *Ginkgo*, is common in the temperate zone, especially China, Korea and Japan. Since ginkgo tree originated about 200 million years ago, it is often called a living fossil. Some fungal species have been reported as pathogens, saprobes or symbionts of *G. biloba* (Aoki 1997, Fontana 1985, Vasilyeva & Mel'nik 2006), but there have been no records of discomycetous fungi on gingko tree substrate.

During our mycofloristic research in Korea, we collected an apothecial ascomycete growing on fallen leaves of *G. biloba*. The fungus, referred to *Hymenoscyphus* by careful macro- and microscopic observation, did not match any known species in the genus. In this paper we describe this fungus as a new species of *Hymenoscyphus* based on morphological characteristics and sequence analysis of the ITS region (ITS1-5.8S-ITS2) of rDNA.

## Materials and methods

Free-hand sections of fresh materials were mounted in distilled water, lactic acid, lacto-cotton blue and Melzer's reagent in case of needs. Dried materials were revived in 3% aqueous KOH. Photographic works were carried out with the aid of a differential interference contrast microscope (Zeiss AX10) equipped

* *Author for correspondence*

Table 1. Sequence data used for phylogenetic analysis.

| Species | Source | Habitat | Accession No. |
|---|---|---|---|
| *Hymenoscyphus caudatus* (P. Karst.) Dennis | HMAS 82057 | Herbaceous stem | AY348576 |
| | HMAS 82060 | Unknown | AY348577 |
| | HMAS 82063 | Herbaceous stem | AY348578 |
| | HMAS 82073 | Herbaceous stem | AY348579 |
| *H. crataegi* Baral & R. Galan | F156966 | Leaves of *Crataegus monogyna* | DQ431177 |
| *H. epiphyllus* (Pers.) Rehm ex Kauffman | HMAS 82075 | Fallen leaves | AY348580 |
| | HMAS 82076 | Unknown | AY348581 |
| | H.B.7054 | Bark of *Quercus* sp. | DQ431180 |
| *H. fructigenus* (Bull.) Fr. | ARON3264.H | Decaying nut of *Quercus robur* | AJ430396 |
| | F109077 | Unknown | DQ431169 |
| | F115882 | Fruits of *Quercus robur* | DQ431171 |
| | F156965 | Twigs of *Abies* sp. | DQ431176 |
| *H. fucatus* (W. Phillips) Baral | HMAS 75902 | Unknown | AY348583 |
| *Hymenoscyphus* sp. | KUS-F51352 | Fallen leaves of *Ginkgo biloba* | EU096525* |
| | KUS-F51854 | Fallen leaves of *Ginkgo biloba* | EU219982* |
| *H. globus* W.Y. Zhuang & Yan H. Zhang | HMAS 82107 | Wet hardwood | AY348593 |
| *H. immutabilis* (Fuckel) Dennis | HMAS 71809 | Rotten leaves of *Populus* sp. | AY348584 |
| | F137632 | Leaves of *Ulmus* sp. | DQ431174 |
| | F155012 | Decayed leaves of *Salix caprea* | DQ431175 |
| *H. lasiopodius* (Pat.) Dennis | HMAS 71820 | Dead root of *Carex* sp. | AY348585 |
| | HMAS 71821 | Dead root of *Carex* sp. | AY348586 |
| | HMAS 75878 | Dead root of *Carex* sp. | AY348587 |
| *H. macroguttatus* Baral, B. Declereq & Hengstm | H.B. 7034 | Leaves of *Acer pseudoplatanus* | DQ431179 |
| *H. scutula* (Pers.) W. Phillips | HMAS 82093 | Fern | AY348590 |
| | HMAS 82098 | Herbaceous stem | AY348591 |
| *H. serotinus* (Pers.) W. Phillips | F093261 | Rotten wood of *Fagus sylvatica* | DQ431168 |
| | F115891 | Branches of *Fagus sylvatica* | DQ431173 |
| | F159526 | *Fagus sylvatica* | DQ431178 |
| | BR020 | *Tamarix gallica* debris | DQ431167 |
| *Lachnum virgineum* (Batsch) P. Karst. | AFTOL-ID 49 | Unknown | DQ491485 |

* Sequences obtained from the present study.

with a digital camera (AxioCam MRc5). The specimens were deposited in the herbarium of Korea University, KUS.

Extraction of genomic DNA was undertaken according to the method outlined by Lee & Taylor (1990). The entire ITS region was amplified by PCR using primers ITS1 and ITS4 (White et al. 1990) and purified using a QIAquick gel extraction kit (Qiagene, Hilden, Germany). And then the products were

directly sequenced on an automatic sequencer (ABI Prism TM 377 DNA Sequencer) with primers identical for PCR. Sequence data were introduced and edited with DNAstar (DNAstar, Inc., Madison, Wis.). To construct a tree twenty-eight sequences were obtained from GenBank and compared with the present fungus (listed in Table 1). Phylogenetic analysis was performed using the neighbor-joining method (Saitou & Nei 1987) in PAUP* ver. 4b10 (Swofford 2002). Relative robustness of the branches was estimated by bootstrapping of 1000 replication. *Lachnum virgineum* (Batsch) P. Karst. (DQ491485), member of the *Hyaloscyphaceae* was selected as outgroup for analysis.

## Results and discussion

The *Hymenoscyphus* species we collected is characterized by its yellowish apothecia with a long stipe, elliptic-clavate ascospores and paraphyses with a deep violet apex. In a mixed pile of fallen leaves, the fruiting bodies of this fungus were found only on the leaves of ginkgo tree, and not on other substrate

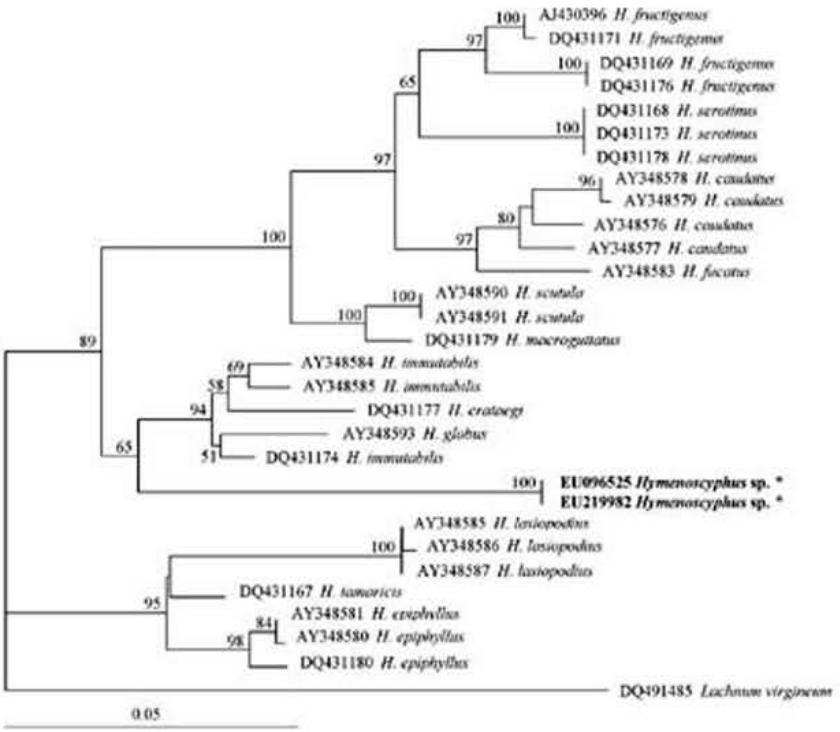


**Fig. 1.** Neighbor-joining tree of *Hymenoscyphus* species based on the ITS rDNA. Numbers above the branches are the bootstrap values (1000 replication, values smaller than 50% not shown). The numbers of nucleotide changed among taxa are represented by branch length and scale bar equals the number of nucleotide substitution per site. Taxa sequenced from this study marked with asterisk (*). *Lachnum virgineum* was chosen as outgroup.

such as leaves of grasses, oaks and other broad-leaved trees. This indicates that this species has a substrate-specific property on ginkgo leaves.

At first glance, the macroscopic features such as its long-stipitate apothecia and the discoloration of dried disc are reminiscent of *H. serotinus* (Pers.) W. Phillips. The measurements also agree with the descriptions of *H. serotinus* for most structures. However, pigmentation of the paraphyses in dried materials separates them: the pigment in *H. serotinus* is yellow to light brown (Dumont & Carpenter 1982), while that in *H.* sp. is deep violet. *Hymenoscyphus serotinus* was reported as associated with woody substrates, mainly of *Fagus sylvatica* in Europe (Lizoň 1992) and on herbaceous stems and leaves in South America (Dumont & Carpenter 1982). Zhuang (1996) reported this fungus (as *Lanzia serotinus*) on leaf blades of *Ulmus* sp. in China, but there have been no additional records of *H. serotinus* growing on leaves of woody plants, including ginkgo tree. *Hymenoscyphus caudatus* (P. Karst.) Dennis, reported on various leaves, is also similar to the present fungus but is distinguished by its shorter stipe (≤ 2 mm long).

The morphological discrepancy is supported by high genetic distances from other species of *Hymenoscyphus*. In the neighbor-joining tree, sequence data of the *Hymenoscyphus* sp. (EU096525 and EU219982) were independently nested within the genus *Hymenoscyphus* (Fig. 1). The next closest clade, consisting of *H. immutabilis* (Fuckel) Dennis, *H. globus* W.Y. Zhuang & Yan H. Zhang, and *H. crataegi* Baral & R. Galan, grouped with *Hymenoscyphus* sp. with moderate bootstrap value of 65% but shared significant sequence dissimilarities with the latter fungus (9.4–9.9%). They also differed morphologically spore shape, ectal excipular structure, and ecological habitat. In contrast *H. serotinus* and *H. caudatus* were distinctly separated from *Hymenoscyphus* sp. despite their morphological similarity. Baral et al. (2006) already noted such discordance between morphological and molecular data of some *Hymenoscyphus* species. Based on morphological features and molecular evidence, we describe and illustrate the fungus as a new species, *H. ginkgonis*.

***Hymenoscyphus ginkgonis*** J.G. Han & H.D. Shin, **sp. nov.** **Fig. 2**

MycoBank MB511133

*Apothecia superficialia, longe stipitata. Disco 0.5–2.0 mm diametro, plana vel anguste concava, albido ab pallide sufflavo, sicco fusco-brunnea. Receptaculo glabro, constans albido ab sufflavo. Stipes tenuis, usque 7 mm longus. Excipulum ectale ex textura prismatica compositur, cellulis hyalinis tenuiter tunicatis, 15–29 x 6–11 µm. Excipulum medullosum textura porrecta vel intricata compositum, hyphis 2–3 µm latis. Asci cylindraceo-clavati, octospori, poro iodo caerulescentia, 72–98 x 8–11 µm. Ascosporae irregulariter biseriatae, elliptico-clavatae, hyalinae, rectae vel minute curvatae, unicellulares vel 1-septatae, 17–22 x 3–4 µm. Paraphyses cylindraceae, sicco fusco-violaceae, septatae, apicibus anguste clavatis, usque ad 2 µm latis.*

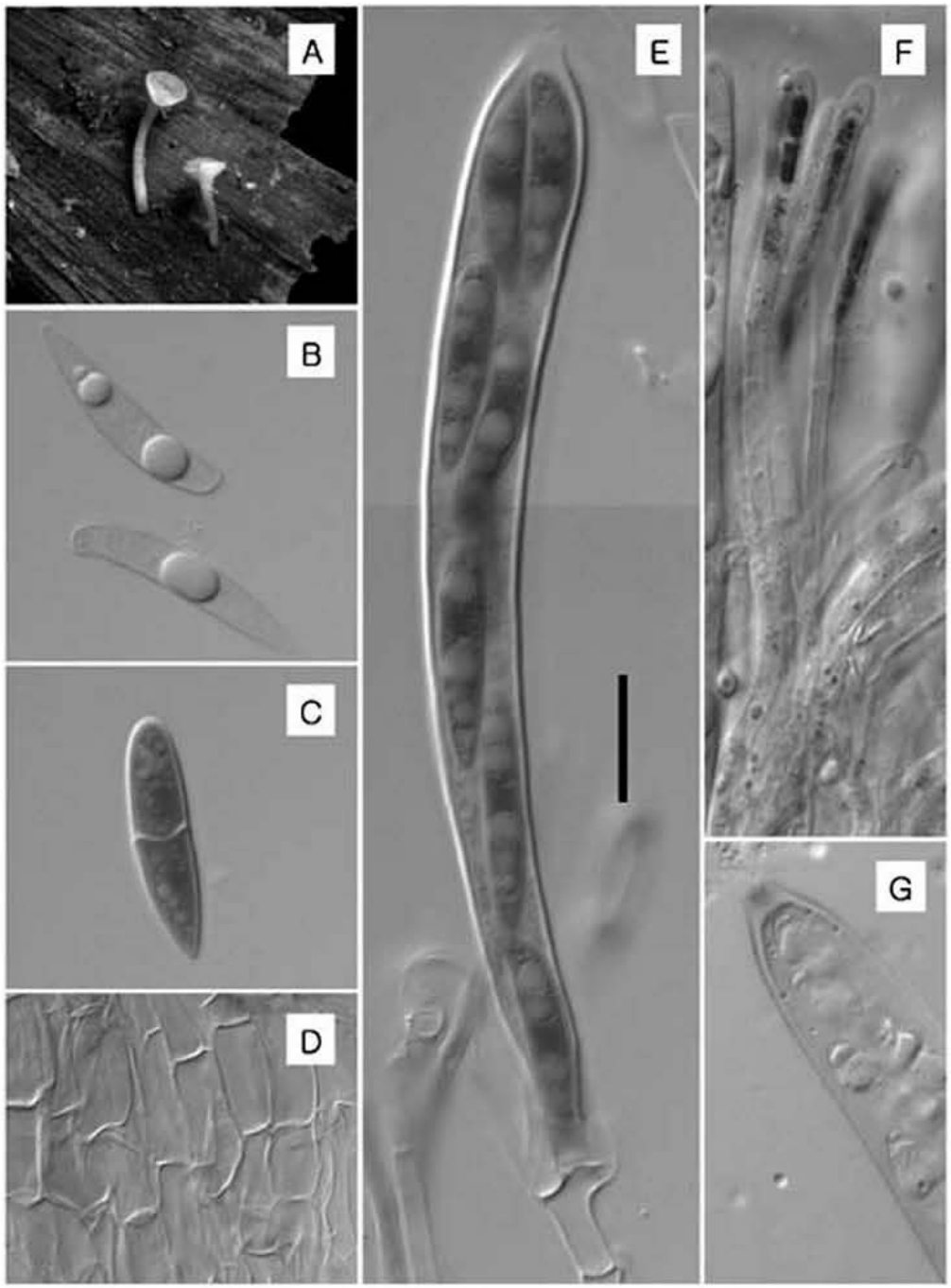


**Fig. 2.** *Hymenoscyphus ginkgonis* (KUS-F51352). A: apothecia occurring on a fallen leaf of *Ginkgo biloba*, B: ascospores containing guttules, C: 1-septate ascospore, D: ectal excipulum composed of prismatic cells, E: ascus arising from simple septum, F: apex of paraphyses turning deep violet in dried materials, G: apical pore blued in Melzer's reagent.

Scale bar: 4 mm for A, 10 μm for B, C, E, F and G, 20 μm for D.

HOLOTYPE — Korea, Wonju, Mons Chiak, alt. 430m, 37°23'56" N, 128°2'57" E, ad folia *Ginkgo biloba*, 15 September 2006, leg. J.G. Han & H.D. Shin (KUS-F51352).

ETYMOLOGY — "*ginkgonis*" refers to the host plant on which the fungus was collected.

APOTHECIA superficial, disc-shaped and long-stipitate. DISC 0.5–2.0 mm in diameter, flat to slightly concave, white to pale yellowish when fresh, turning dark brown when dry. RECEPTACLE smooth, concolorous with the hymenium when fresh, but, no discoloration occurred when dry. STIPE up to 7 mm long, slender, white. ECTAL EXCIPULUM composed of thin-walled, hyaline, prismatic cells 15–29 x 6–11 µm in the flanks of the cup, becoming textura porrecta at the stipe, hyphae 2–3 µm wide. MEDULLARY EXCIPULUM composed of textura porrecta to textura intricata. ASCI arising from simple septa, cylindric-clavate, hyaline, 8-spored, apex conical, apical pore blued in Melzer's reagent, 72–98 x 8–11µm. ASCOSPORES irregularly biseriate, elliptic-clavate, pointed at one end, straight to slightly curved, hyaline, smooth, containing several guttules, 0–1-spetate, 17–22 x 3–4 µm. PARAPHYSES cylindric, slightly swelling upward or not, up to 2 µm wide at the apex, apex turning deep violet in dried material, septate, equal to or only slightly exceeding the asci.

SPECIMENS EXAMINED: Korea, Wonju, Mt. Chiak National Park, alt. 430m, 37°23'56" N, 128°2'57" E, on fallen leaves of *Ginkgo biloba*, 15 September 2006, leg. J.G. Han & H.D. Shin (Holotype: KUS-F51352); same locality, 30 August 2007 (KUS-F51817); Korea, Yangpyeong, Experimental Forest of Korea University, alt. 240m, N37°24'49.25" E127°43'5.32", on fallen leaves of *G. biloba*, 10 September 2007, leg. H.D. Shin (KUS-F51854).

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MYCOTAXON

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# Additions to the knowledge of lignocellulolytic basidiomycetes in forests from Santa Catarina, Southern Brazil

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**Abstract** — An updated checklist of the lignocellulolytic basidiomycetes in Santa Catarina State with 110 species distributed in 19 families and six orders (*Agaricales, Hymenochaetales, Polyporales, Russulales, Auriculariales, Tremellales*) is presented. The complete checklist is available on http://www.mycotaxon.com/resources/weblists.html and as Electronic Guide BASC (Basidiomycetes of Santa Catarina) as well on http://www.cienciasbiologicas.ufsc.br/bot/micologia/index.htm.

## Introduction

*Basidiomycota* comprises almost 30,000 species with *Basidiomycetes* representing approximately 70% (more of the 20,000 spp.) of the phylum (Kirk et al. 2001, David 2002). The basidiomycetes include wood decomposing organisms in most ecosystems causing white and brown rots in tropical areas where they are very abundant (Nobles 1971, Dix & Webster 1995, Deacon 1997, Anagnost 1998, Highley & Dashek 1998, Bononi & Grandi 1999). In Southern Brazil, the Santa Catarina State (26°–30° S lat, 48°30'–54° W long) presents two major vegetation types: the Atlantic Rain Forest and the Atlantic Semi-deciduous Forest (Morellato & Haddad 2000). In these ecosystems, Polypore fungi were first studied by Europeans Mycologists already in the 19th century (Loguercio-Leite 1990). The contributions by Brazilian researches from the Mycology Laboratory (BOT/CCB/UFSC) in Santa Catarina State has been taking place since 1990, with several works based on collections from the Atlantic Forest (Loguercio-Leite & Wright 1991ab, Loguercio-Leite 1992, 1993, 1994; Loguercio-Leite & Wright 1995, Gerber 1996, Gerber & Loguercio-Leite 1997, Loguercio-Leite & Gerber 1997, Loguercio-Leite & Wright 1998, Loguercio-Leite et al. 1998, Gerber et al. 1999, Neves & Loguercio-Leite 1999, Gerber & Loguercio-Leite 2000, Gonçalves 2001, Loguercio-Leite et al. 2001, 2002; Groposo & Loguercio-

Leite 2005). The purpose of this study was to estimate the species diversity of basidiomycetes in Santa Catarina State.

## Material and methods

This study is based on the collections deposited in the herbarium FLOR since 1983 (Holmgren et al. 1990). Basidiomes were studied using macroscopic (shape, size, colors, types of hymenophore) and microscopic (presence/absence of structures, dimensions, basidiospores) characters (Ryvarden 1991). Measurements were made from slide preparations stained with 1% aqueous phloxine and 5% KOH. Melzer's reagent was used to define wall chemical characteristics. Specimens were identified to species using specialized references and comparing collections with BAFC, ICN and PACA Herbaria collections. Nomenclature, taxonomy and author citation followed Kirk et al. 2001 and databases: Centraalbureau voor Schimmelcultures – CBS (http://www.cbs.knaw.nl/databases/) and Index Fungorum – IFS (http://www.indexfungorum.org/Names/Names.asp). For geographic references consult the map of Santa Catarina State in Groposo & Loguercio-Leite (2005).

## Results and discussion

As a contribution to the general checklist of the lignocellulolytic fungi (*Basidiomycetes*) in Santa Catarina State a total of 110 species is presented. The taxa are distributed in 19 families and six orders. *Polyporales* is the most representative order with 74 species in 12 families, followed by *Hymenochaetales* with 25 species in two families. The high diversity of *Polyporales* found in the present study agrees with the results of other inventories of the basidiomycetes on subtropical Southern Brazil (Groposo & Loguercio-Leite 2005) and also in tropical Northeastern Brazil (Gibertoni et al. 2004). Consequently, *Polyporaceae* is the most representative family with 45 species, followed by *Hymenochaetaceae* with 23 species. *Phellinus* s. l. is the genus with the highest number of species with 16 taxa. Most of the reported species showed a tropical distribution: 47 neotropical (almost 42.7%) and 42 pantropical species (38.2%). Only 8 and 13 species are considered widely distributed (7.3%) and cosmopolitan (11.8%), respectively. Up to now, these 110 species added here to the original list of lignocellulolytic fungi (Groposo & Loguercio-Leite 2005) increases to 157 species the total diversity of these organisms in Santa Catarina State, Brazil. The results revealed a high mycodiversity in Santa Catarina State, which corresponds to the knowledge of the basidiomycetes in the Atlantic Forest of Southern Brazil. From the total 157 species, 75.8% (119 species) are tropical (pantropical and neotropical). Besides the 4 species known only from Brazil [*Amauroderma corneri* Gulaid & Ryvarden, *Antrodiella multipileata* C.L. Leite & J.E. Wright, *Hennings brasiliensis* (Speg.) Speg., *Skeletocutis roseolus* (Rick

ex Theiss.) Rajchenb.] cited earlier by Groposo & Loguercio-Leite (2005), we report ten additional species endemic to this country: *Phellinus bambusarum* (Rick) M.J. Larsen, *Ceriporiopsis cystidiata* C.L. Leite et al., *Rigidoporus amazonicus* Ryvarden, *Diplomitoporus dilutabilis* C.L. Leite & J.E. Wright, *Rubroporus carneoporis* C.L. Leite et al., *Tyromyces crassisporus* C.L. Leite & J.E. Wright, *Tyromyces hypocitrinus* (Berk.) Ryvarden, *Wrightoporia porilacerata* C.L. Leite et al., *Pyrofomes fulvoumbrinus* (Bres.) A. David & Rajchenb., and *Protomerulius substuppeus* (Berk. & Cooke) Ryvarden. Of these, six are known only to Santa Catarina State. Only eight of the 157 taxa cause brown rot [*Fistulina hepatica* (Schaeff.) With., *Fomitopsis feei* (Fr.) Kreisel, *Gloeophyllum striatum* (Sw.) Murrill, *Antrodia albida* (Fr.) Donk, *Laetiporus sulphureus* (Bull.) Murrill, *Amylosporus bracei* (Murrill) A. David & Rajchenb., *Wrightoporia avellanea* (Bres.) Pouzar, and *Wrightoporia porilacerata*]. Consequently, 93-95% of the lignocellulolytic fungi from Santa Catarina caused white rots, confirming the trends given for tropical and subtropical areas by several authors (Nobles 1971, Gilbertson 1980, Nakasone 1996).

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# *Bulbothrix viatica*, a new species of *Parmeliaceae* from Brazil

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**Abstract–** *Bulbothrix viatica* is here proposed as new. It is a very common species that has been confused with *Bulbothrix subcoronata* but differs in its brown undersurface, larger ascospores, and wider lobes.

**Key words–** lichens, bulbate cilia, Rio Grande do Sul

The lichen genus *Bulbothrix* Hale (*Parmeliaceae*) is diagnosed by the bulbate marginal cilia, pale grey upper cortex, and production of atranorin (Hale 1974, 1976). *Relicina* (Hale & Kurok.) Hale also has bulbate cilia, but the cortex is yellowish colored, with usnic acid (Hale 1975, Elix 1996).

At present 48 species of *Bulbothrix* are known (Farr et al. 1999), 20 of which have been recorded from Brazil (Marcelli 2005) and six from the state Rio Grande do Sul (Spielmann 2006). Furthermore, six new species recently discovered in south and southeastern Brazil are in the process of publication (Canêz & Marcelli 2007, Jungbluth et al. 2008). This supports Hale's (1976) earlier assertion that Brazil is the center of diversity for the genus.

During our *Parmeliaceae* research along the roadsides and slopes in the central region of the Rio Grande do Sul State (Spielmann 2005), we found an interesting species of *Bulbothrix* that is described for the first time below.

***Bulbothrix viatica*** A.A. Spielm. & Marcelli, **sp. nov.** FIG. 1

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*Thallus adnatus, cinereo-glaucescens, membranaceus, margine bulbato-ciliatus, isidiis sorediisque destitutus, subtus brunneus, rhizinosus, rhizinis nigris vel atrobrunnescentibus, simplicibus. Apothecia coronata, sporae 12.0–16.5 × 7.5–10.0 µm, episporio 1–2 µm crasso. Thallus K+ flavescens, medulla K+ primo lutescens, dein valde aurantiaca; atranorinam et acidum norsticticum continens.*

HOLOTYPUS– Brazil, Rio Grande do Sul State, Municipio Santa Cruz do Sul, margin of the highway RST-287, km 102, 29°41'03.3"S, 52°25'33.6"W, 150 m alt., roadside, on *Eucalyptus* branch, 28 July 2003, A.A. Spielmann 389 (SP)

THALLUS greenish gray, lobate, adnate, membranaceous, saxicolous or corticolous, 1.5–7.5 cm in diameter; LOBES (SUBLACINIAE) irregularly branched, laterally overlapping, 1–4 mm wide, with rounded tips, margins crenate to crenate-incised (sublacinulate); distal surface smooth to slightly rugose, becoming rugose and cracked toward the center; LACINULAE absent; MACULAE absent; CILIA black, bulbate, simple, frequent, located at the lobe and crenae axils, with one or rarely two apices per bulb, 0.05–0.70 × 0.02–0.15 mm (including bulb). PUSTULAE absent. SOREDIA absent. ISIDIA absent. MEDULLA white or sometimes with some K– light orange points. LOWER SURFACE brown to dark-brown, dull, smooth to rugose; MARGIN dark-brown, 1.0–1.5 mm wide, bare, with attenuate transition to the central parts, smooth to rugose or sometimes veined at the edge; RHIZINES black to dark brown, simple, base not or just slightly bulbate, 0.10–0.50 × 0.01–0.04 mm, abundant, evenly dispersed. APOTHECIA urceolate to concave, 1.5–8.0 mm in diameter, sessile to substipitate, laminal, margin crenate, coronate, coronal bulbs sometimes with ciliate apices, amphithecium smooth, sometimes with scarce bulbs, disc brown, matt, epruinose, imperforate; EPITHECIUM 2.5–12.0 µm thick; HYMENIUM 55–75 µm tall; SUBHYMENIUM 12.5–25.0 µm thick; ASCOSPORES ellipsoid to narrowly ellipsoid, 12.0–17.5 × 7.5–10.0 µm, episporium 1.0–2.0 µm thick. PYCNIDIA submarginal to laminal, conspicuous, with or without a prominent margin, ostiole black; CONIDIA bifusiform (sometimes hardly distinct), 5.0–7.5 × ca. 1.0 µm.

CHEMISTRY: cortex K+ yellow, UV–; medulla K+ yellow → strongly orange or orange-red, C–, KC–, P+ orange-yellow, UV–; containing atranorin (cortex), norstictic acid and connorstictic acid (medulla).

PARATYPES: Brazil, Rio Grande do Sul State. Herveiras Municipality, 29°27'12.5"S, 52°37'57.7"W, 540 m alt., on a roadside *Eucalyptus* trunk, open place, *A.A. Spielmann, L.S. Canêz & C. Trentin* 714, 24 January 2004. Idem, Santa Cruz do Sul Municipality, margin of highway RST-287, Km 102, 29°41'03.3"S, 52°25'33.6"W, 150 m alt., roadside, on an *Eucalyptus* branch, *A.A. Spielmann* 389, 28 July 2003. Idem, Sinimbu Municipality, Cava Funda, 29°27'41.8"S, 52°31'11.7"W, 500 m alt., corticolous, on roadside, open place, *A.A. Spielmann* 1318, 12 February 2003; idem, 29°27'33.4"S, 52°31'05.1"W, 520 m alt., saxicolous, on roadside, open place, *A.A. Spielmann & L.S. Canêz* 713, 05 May 2004; idem, ca. 1 Km from Linha Almeida, 29°23'20.2"S, 52°30'21.9"W, on fallen branch at roadside, open place, *A.A. Spielmann & L.S. Canêz* 1336, 05 January 2004. Idem, Sobradinho Municipality, margin of highway RST-481, next to the crossroads, 29°24'20.2"S, 53°01'25.9"W, 375 m alt., corticolous, on bush at roadside, open place, *A.A. Spielmann* 341, 17 July 2003.

COMMENTS–The absence of vegetative propagules, coronate apothecia, a brown to dark-brown undersurface, a medulla with norstictic acid (K+ yellow →

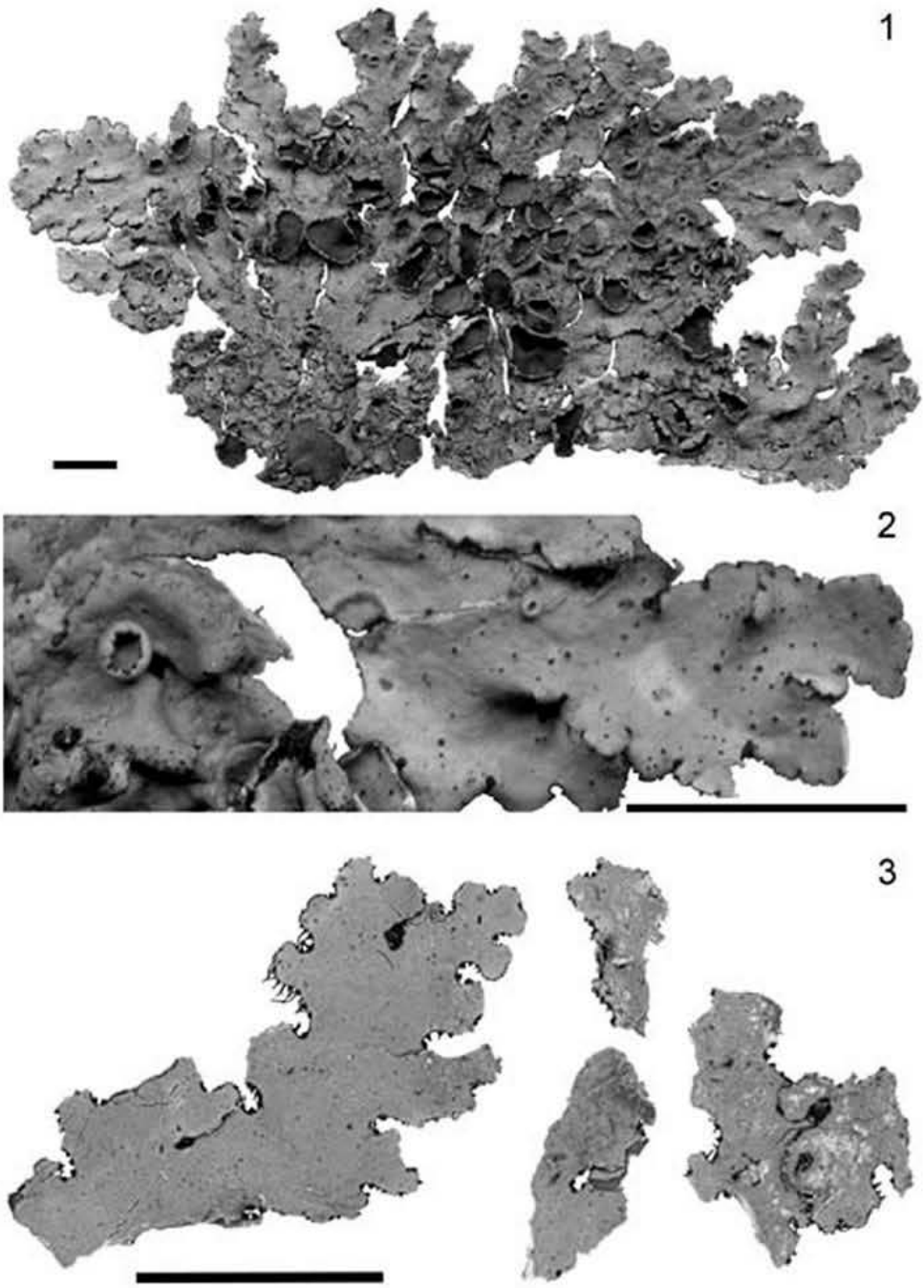


**Fig. 1.** *Bulbothrix viatica* (holotype).
**Fig. 2.** *Bulbothrix viatica* (detail of holotype).
**Fig. 3.** *Bulbothrix subcoronata* (the complete holotype).
Bars = 5 mm.

intense orange or orange-red), large (12.0–17.5 × 7.5–10.0 µm) ascospores and wide (1–4 mm) lobes differentiate *Bulbothrix viatica* from related species.

The studied specimens were at first identified as *Bulbothrix subcoronata* (Müll. Arg.) Hale, since all available keys and descriptions attribute this name to any *Bulbothrix* material having a brown undersurface, large (7–20 × 5–10 µm) ascospores, and lobes 0.5–3.0 mm wide (Hale 1976, Fleig 1985, Marcelli 1993, Ribeiro 1998, Eliasaro 2001).

However, *B. subcoronata* was originally described [*Parmelia subcoronata*] as having small (ca. 5 µm long), broadly ellipsoid to subglobose ascospores (Müller 1887). Analysis of the holotype (G!, FIG. 2) shows that it has a shiny black underside and lobes just 0.5–1.0 mm wide. Furthermore, the ascospores were found to measure actually 5.0–7.5 × 4.0–5.5 µm, with an episporium ca. 0.5 µm (Benatti, pers. comm.).

Therefore, it is highly possible that the specimens cited in the literature listed above as *Bulbothrix subcoronata* belong, in fact, to *B. viatica*.

This species is relatively common along roadsides in the studied area; therefore the specific epithet *viatica* (= growing along roads, fide Stearn 1992) was chosen.

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MYCOTAXON

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# Taxonomic revision of the genus *Cladosporium* s. lat. 8. Reintroduction of *Graphiopsis* (= *Dichocladosporium*) with further reassessments of cladosporioid hyphomycetes

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**Abstract** — *Graphiopsis* is shown to be an older, valid name for the recently introduced genus *Dichocladosporium*. The new name *Cladosporium vincicola* is introduced and the new combinations *Fusicladium britannicum* (= *Cladosporium britannicum*) and *F. psammicola* (= *Exosporium psammicola*, *Cladosporium psammicola*) are proposed. The fungus represented by the invalid name *Cladosporium indigoferae* is described and illustrated. Due to insufficient material, however, its generic affinity could not be resolved.

**Key words** — anamorphs, cladosporium-like

## Introduction

Based on a new phylogenetic and morphological circumscription of the heterogeneous genus *Cladosporium* Link (David 1997, Braun et al. 2003, Schubert et al. 2007b), attempts to redescribe and reassess the numerous species previously assigned to this genus (Dugan et al. 2004) have recently been made (Braun & Schubert 2007, Heuchert et al. 2005, Schubert 2005a,b; Schubert & Braun 2004, 2005a,b, 2007; Schubert et al. 2006, 2007a,b). In a recently published issue of 'Studies in Mycology' dedicated to cladosporioid fungi (Crous et al. 2007b), all aspects and kinds of hyphomycetes previously assigned to *Cladosporium* s. lat. (Crous et al. 2007a) have been addressed using standardized cultures and molecular approaches. This volume provides an important basis for the preparation of a projected monograph of *Cladosporium*.

In the present paper, the nomenclature of a recently introduced cladosporioid genus is discussed and some species, which are only known from type material and other herbarium specimens, are redescribed and reassessed.

## Materials and methods

The collections examined were described, mounted in distilled water, using oil immersion (bright field and phase contrast), but without any staining, by means of standard light microscopy (Olympus BX 50, Hamburg, Germany). The collections examined are deposited at the herbaria BPI, CUP, HAL, IMI, LE, PAD, PPMH and W (abbreviations according to Holmgren et al. 1990).

## Nomenclature and taxonomy

### 1. *Graphiopsis* – an older valid name for *Dichocladosporium*

*Cladosporium chlorocephalum* (= *C. paeoniae*) is a common, widespread hyphomycete of peony, characterized by dimorphic fruiting, and causing distinct necrotic leaf-blotch symptoms on living leaves, and with a second type of conidiophore and conidia on rotten, overwintered stems. Based on detailed re-examinations of this fungus in vivo and in vitro, supplemented by sequence data of the ITS and LSU gene regions, Schubert et al. (2007a) clearly demonstrated that this fungus has to be excluded from *Cladosporium* s. str. It is retained in the *Davidiellaceae* (*Capnodiales*), but forms a clade separate from the *Davidiella* Crous & U. Braun cluster. The placement of this fungus in a separate genus, '*Dichocladosporium*' was supported by its phylogenetic position and morphological peculiarities of the conidiophores, conidiogenous loci and hila. Unfortunately it was overlooked that *D. chlorocephala* (≡ *Periconia chlorocephala*), the type species of the former genus, is also the type species of an older, forgotten genus, *Graphiopsis*. Trail (1889) probably misinterpreted the name *P. chlorocephala* since he assigned Scottish collections on *Carex* spp., *Juncus effusus* and *Phragmites australis* to this species, which is confined to *Paeonia* spp. But peony was not listed under the hosts cited. This is, however, nomenclaturally irrelevant since Trail (1889: 76) clearly cited *Periconia chlorocephala* as type species of *Graphiopsis*. Hence, the following nomenclatural reassessment and correction is necessary:

***Graphiopsis*** Trail, Scott. Naturalist (Perth) 10: 75. 1889

[non *Graphiopsis* Bainier 1907].

TYPE SPECIES: *G. chlorocephala* (Fresen.) Trail.

≡ *Dichocladosporium* K. Schub., U. Braun & Crous, Stud. Mycol. 58: 96. 2007.

***Graphiopsis chlorocephala*** (Fresen.) Trail, Scott. Naturalist (Perth) 10: 76. 1889.

NEOTYPE: On dead stems of *Paeonia officinalis*, Germany, Sachsen-Anhalt, Halle (Saale), Botanical garden, 16 Mar. 2005, K. Schubert (HAL 1924 F). Isoneotype: CBS-H 19869. Culture ex neotype: CBS 121523 = CPC 11969.

≡ *Periconia chlorocephala* Fresen., Beitr. Mykol. 1: 21. 1850.
≡ *Haplographium chlorocephalum* (Fresen.) Grove, Hardwicke's Sci.-Gossip 21: 198. 1885.
≡ *Cladosporium chlorocephalum* (Fresen.) E.W. Mason & M.B. Ellis, Mycol. Pap. 56: 123. 1953.
≡ *Dichocladosporium chlorocephalum* (Fresen.) K. Schub., U. Braun & Crous, Stud. Mycol. 58: 96. 2007.
= *Cladosporium paeoniae* Pass., in Thümen, Herb. Mycol. Oecon., Fasc. IX, No. 416. 1876, and in Bot. Jahresber. (Just) 4: 235. 1876.
= *Periconia ellipsospora* Penz. & Sacc., Atti Reale Ist. Veneto Sci. Lett. Arti, Ser. 6, 2: 596. 1884.
= *Cladosporium paeoniae* var. *paeoniae-anomalae* Sacc., Syll. Fung. 4: 362. 1886.
= *Haplographium chlorocephalum* var. *ovalisporum* Ferraris, Fl. Ital. Cryptog., Hyphales: 875. 1914.

2. ***Cladosporium vincicola*** U. Braun & K. Schub., **nom. nov.** FIG. 1

MYCOBANK, MB 511302

≡ *Cladosporium vincae* Moesz, Bot. Közlem. 23: 123. 1926, nom. illeg., non *C. vincae* Fairm., 1911.

MATERIAL EXAMINED: HUNGARY. Near Budapest, on living leaves of *Vinca herbacea* (*Apocynaceae*) infected by *Puccinia vincae* (DC.) Plowr., 25 Apr. 1926, W. Moesz (W 10216: lectotype, selected here). RUSSIA. St. Petersburg, Botanical Garden of the Komarov Botanical Institute, on living leaves of *Vinca minor*, 7 May 2007, V.A. Mel'nik (HAL 2069 F).

LEAF SPOTS usually initiated terminally or laterally, later spreading, covering large leaf segments, often more than 50 % of the leaf blade, finally entire leaves turning necrotic, shape and size of the lesions variable, dark brown to pale grayish brown, later dingy gray to grayish white, margin indefinite or with a narrow to moderately wide dark brown to blackish border or halo, sometimes with rather diffuse discolorations. CAESPITULI amphigenous, mainly hypophyllous, punctiform, scattered, dark brown. MYCELIUM internal, forming immersed stromatic aggregations, 10–80 µm diam., composed of swollen hyphal cells, pigmented, thick-walled, up to 12 µm diam. CONIDIOPHORES in small to moderately large fascicles, loose to moderately dense, occasionally solitary, arising from substomatal stromatic hyphal aggregations, emerging through stomata, erect, straight, subcylindrical to moderately geniculate-sinuous, rarely subnodulose, unbranched, 15–100 × 3–7.5 µm, at the very base sometimes up to 10 µm wide, 0–4(–6)-septate, pale to medium olivaceous or olivaceous-brown, wall 0.5–1 µm thick, one-layered, smooth to faintly rough-walled; conidiogenous cells integrated, terminal or intercalary, 10–35 µm long; conidiogenous loci distinctly coronate, somewhat protuberant, 1.5–2.5 µm wide and ca. 1 µm high. CONIDIA in simple or branched chains, ramoconidia lacking, secondary ramoconidia (sensu Schubert et al. 2007b) and conidia ellipsoid-ovoid, obovoid, fusiform, rarely subcylindrical, (4.5–)6–25(–33) × (3–)4–8(–9) µm, 0–3(–4)-septate, pale olivaceous to olivaceous-brown, wall thin, ca. 0.5 µm, sometimes distinctly two-layered and up to 1 µm thick, distinctly verruculose,

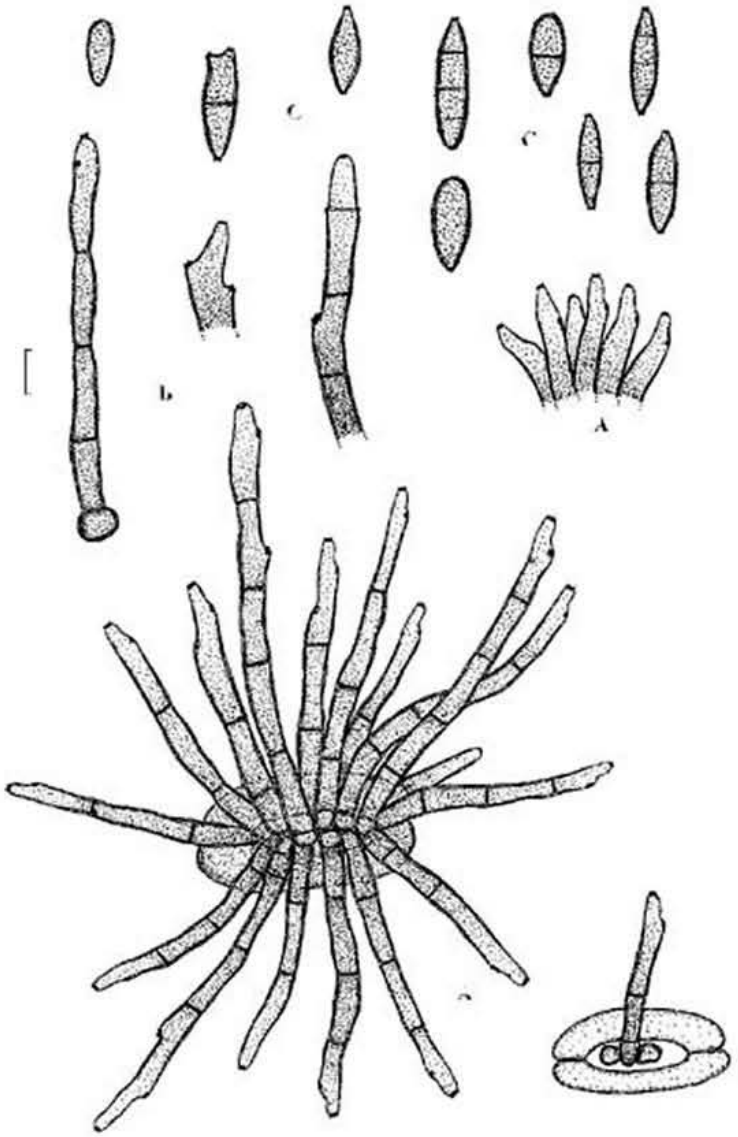

Fig. 1. *Cladosporium vincicola*.
A. Conidiophore fascicle. B. Conidiophores. C. Conidia.
Scale bar = 10 µm. U. Braun *del.*

apex rounded in conidia formed singly, attenuated in catenate conidia, base rounded, with an abruptly protuberant hilum or attenuated, 1–2 µm diam., occasionally with microcyclic conidiogenesis.

COMMENTS: Morphologically this species resembles *Cladosporium aecidiicola* Thüm. However, *C. vincicola* is undoubtedly biotrophic, forming distinct leaf lesions, with fasciculate conidiophores emerging through stomata, and is not associated with rust aecia. The fascicles of conidiophores arise from

well-developed, large stromata. The conidia of *C. vincicola* agree well with those of *Cladosporium herbarum* (Pers.) Link s. str., but the conidiophores and the arrangement of the conidiogenous loci is different. The conidiophores in *C. vincicola* are non-nodulose, i.e., the conidiogenous loci are not confined to nodulose swellings as in *C. herbarum*. *Cladosporium vincae* Fairm. (material examined: on dead leaves of *Vinca minor*, USA, New York, Lyndonville, 6 May 1910, C.E. Fairman, CUP-F2873(24-68), holotype) is identical with *Cladosporium macrocarpum* Preuss, i.e., the conidiophores are distinctly nodulose with conidiogenous loci confined to swellings, and the conidia are rather broad, 5.5–11 µm, mostly 8–10 µm.

3. ***Fusicladium britannicum*** (M.B. Ellis) U. Braun & K. Schub., **comb. nov.** FIG. 2
MycoBank, MB 511303
≡ *Cladosporium britannicum* M.B. Ellis, More Dematiaceous Hyphomycetes: 328. 1976.

ILLUSTRATIONS: Ellis (1976: 327, Fig. 245 C), Ellis & Ellis (1985: Pl. 19, Fig. 190).

MATERIAL EXAMINED: UK. WALES, Pwee-y-Faeda Estate, on rotten wood of *Quercus* sp. (*Fagaceae*), 13 May 1973, collector unknown (IMI 175936: holotype).

COLONIES on rotten wood, effuse, dark to blackish brown, villose. MYCELIUM usually immersed; hyphae sparingly branched, 1–3 µm wide, septate, brown, with solitary or aggregated swollen hyphal cells, 3–10 µm diam., subglobose, brown, wall up to 1 µm thick, but genuine stromata lacking. CONIDIOPHORES solitary to loosely aggregated, arising from immersed hyphae or swollen hyphal cells, erect, straight, subcylindrical to flexuous, sinuous, rarely slightly geniculate, unbranched, 80–350 × 3–6(–8) µm, pluriseptate throughout, medium to dark brown, tip somewhat paler, wall slightly thickened, up to 1 µm wide, smooth; conidiogenous cells integrated, terminal, 10–30 µm long; conidiogenous loci inconspicuous to subconspicuous by being subdenticulate, apex truncate to somewhat convex, 1–2.5 µm diam., unthickened or almost so, but somewhat darkened-refractive. CONIDIA in simple or branched chains, (9–)12–20 × (4–)5–8 µm, primary conidia obovoid, apex rounded, base short obconically truncate, secondary conidia ellipsoid-ovoid, fusiform, both ends attenuated, tips (hila) truncate, pale brown to medium brown, wall thin, 0.5–1 µm wide, smooth, hila (1–)1.5–2.5(–3) µm diam., unthickened or almost so, but often somewhat darkened-refractive.

COMMENTS: Due to the non-coronate structure of the conidiogenous loci and conidial hila, this species has to be excluded from *Cladosporium* s. str. The subdenticulate, unthickened, but somewhat darkened-refractive loci and hila agree well with those of *Fusicladium* Bonord. (Schubert et al. 2003). Without cultures and molecular sequence analyses, it is rather difficult to distinguish *Fusicladium* anamorphs [*Venturiaceae*] from species of the genus *Cladophialophora* Borelli [*Herpotrichellaceae*] (Crous et al. 2007c). However,

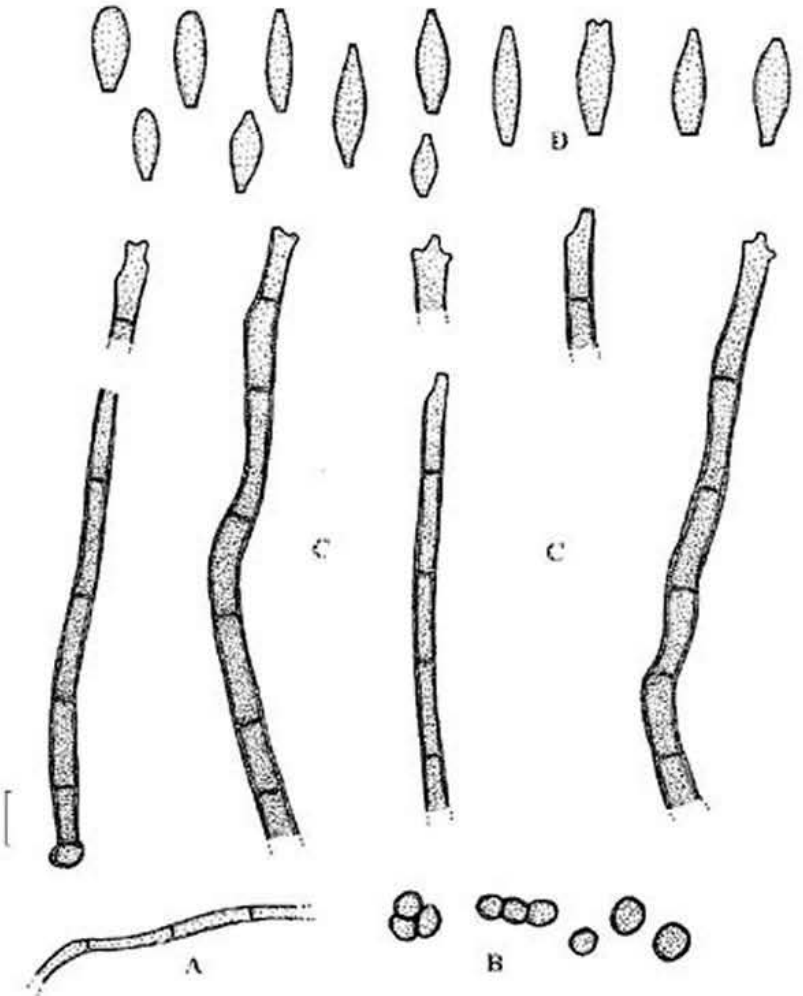


Fig. 2. *Fusicladium britannicum*.
A. Hyphae. B. Swollen hyphal cells. C. Conidiophores. D. Conidia.
Scale bar = 10 µm. U. Braun *del.*

based on the structure of the rather coarse, long and wide conidiophores, this species has been placed in *Fusicladium*.

4. ***Fusicladium psammicola*** (Sacc.) U. Braun & K. Schub., **comb. nov.** FIG. 3

MycoBank, MB 511304

= *Exosporium psammicola* Sacc., in Saccardo & Trotter, Ann. Mycol. 11: 420. 1913.

= *Cladosporium psammicola* (Sacc.) Morgan-Jones & W.B. Kendr., Canad. J. Bot. 50(9): 1817. 1972.

ILLUSTRATION: Morgan-Jones & Kendrick (1972: 1818, Fig. 1).

MATERIAL EXAMINED: LIBYA. Ras Carrac, Magna Syrte, on dead leaves of *Psamma arenaria* [= *Ammophila arenaria*] (*Poaceae*), 18 May 1913, A. Trotter (PAD: holotype).

COLONIES on dead leaves, punctiform to subeffuse, dark brown to blackish, scattered to dense. MYCELIUM immersed, occasionally partly superficial, branched, septate, subhyaline to brown, 2–5 µm wide, forming irregularly radiating strands and aggregations of swollen hyphal cells, 3–9 µm wide, brown, wall somewhat thickened, septate, usually constricted at the septa, often confluent, forming irregular plates or stromata, up to 50 µm diam., immersed to erumpent. CONIDIOPHORES solitary, in loose groups or loose to dense fascicles, arising from hyphal aggregations or stromata, erect, subcylindrical-conical, unbranched, 5–35 × 3–6 µm (according to the original description up to 70 µm long), aseptate or septate, pale brown, wall thin to slightly thickened, smooth to faintly rough-walled; conidiogenous cells integrated, terminal or conidiophores reduced to conidiogenous cells, often with a single conidiogenous locus, sometimes with sympodial proliferation and several loci, 1.5–2 µm diam., truncate to slightly convex, unthickened, but often distinctly darkened (pigmented). CONIDIA solitary, occasionally in short chains, ellipsoid-ovoid, fusiform, short subcylindrical, 10–25 × 4–7 µm, 0–3-septate [rarely with up to

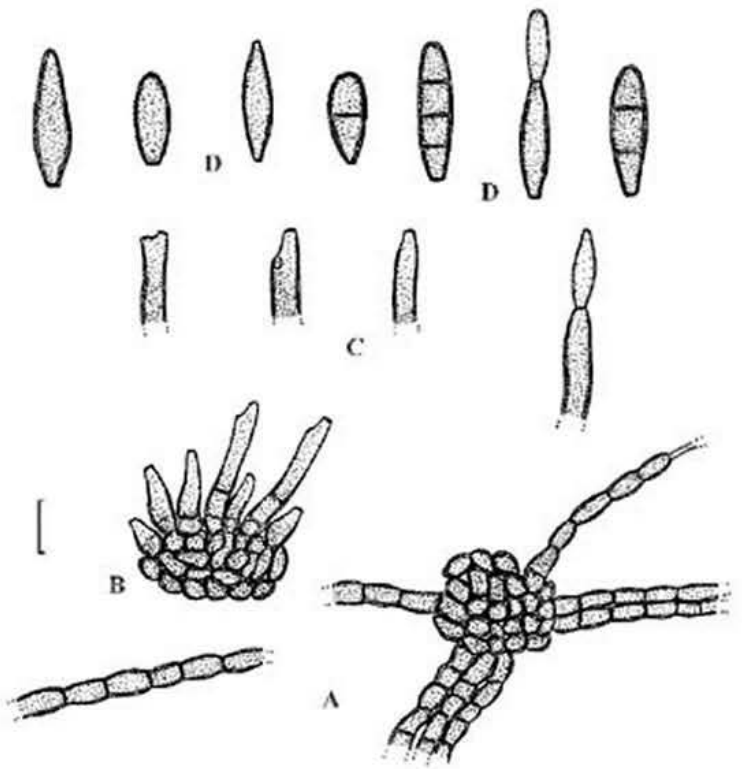


Fig. 3. *Fusicladium psammicola*.
A. Hyphae, hyphal aggregations and strands.
B. Fasciculate conidiophores. C. Conidiogenous cells. D. Conidia.
Scale bar = 10 µm. U. Braun *del.*

5 septa, according to Morgan-Jones & Kendrick (1972)], pale to medium brown or olivaceous-brown, wall thin to slightly thickened, smooth or almost so to faintly rough-walled, apex obtuse or somewhat attenuated, rarely subtruncate, base attenuated, with a truncate hilum, 1.5–2 µm diam., unthickened, but often distinctly darkened.

COMMENTS: On account of clear differences between this species and the basic features of the genus *Exosporium* Link, Morgan-Jones & Kendrick (1972) reallocated *E. psammicola* to *Cladosporium* s. lat. However, due to the non-coronate conidiogenous loci and conidial hila, this species has to be excluded from *Cladosporium* s. str. The presence of subdenticulate, truncate, unthickened, but darkened-refractive scars suggests this species should be assigned to *Fusicladium* (Schubert et al. 2003), an assignment supported by the structure of the radiating hyphal strands and stromatic plates as well as the broad conidia, mostly formed singly. *Cladophialophora* is morphologically similar, but without cultures and molecular sequence analyses it is difficult to distinguish the two genera. However, characteristically radiating hyphal strands and stromatic plates are usually not formed in the latter genus, and the conidia are usually more slender and formed in long acropetal chains.

5. ***Cladosporium indigoferae*** Sawada,

Special Publ. Coll. Agric. Natl. Taiwan Univ. 8: 196. 1959, nom. inval. FIG. 4

= **fusicladium/cladophialophora-like**

ILLUSTRATION: Sawada (1959: Pl. 3, Figs 8–9).

MATERIAL EXAMINED: TAIWAN. PREF. TAIPEI, Taipei, on dead stems of *Indigofera tinctoria* (*Fabaceae*), 18 Aug. 1909, K. Sawada (BPI 427230: type); Taiwan, 18 Aug. 1942, K. Sawada (PPMH: authentic material).

COLONIES on dead stems, without any lesions (probably saprobic), punctiform, scattered or effuse, brown to blackish, somewhat velvety. MYCELIUM internal, forming one- to rarely multi-layered hyphal strands, composed of more or less globular, often somewhat inflated, 4–11 µm diam., smooth, thick-walled, olivaceous-brown cells, giving raise to conidiophores, but typical stromata lacking, additional hyphae smooth, 2–3 µm wide, paler. CONIDIOPHORES solitary or in small loose groups, arising from swollen hyphal cells or hyphal strands, erect, straight to somewhat curved, occasionally somewhat geniculate-sinuous, unbranched or rarely branched, 15–100(–146) × 3–4.5(–5) µm, septate, yellowish olivaceous-brown, wall smooth, slightly thickened; conidiogenous cells integrated, terminal or intercalary, 7–20 µm long, sympodial, with a single or few conidiogenous loci, truncate to slightly convex, 1–2 µm wide, barely thickened, but somewhat darkened-refractive. CONIDIA in branched chains, subglobose, ellipsoid-ovoid, cylindrical, straight, 4–26 × 3–5(–7) µm, 0–3-septate, not to somewhat constricted at the septa, pale olivaceous, wall

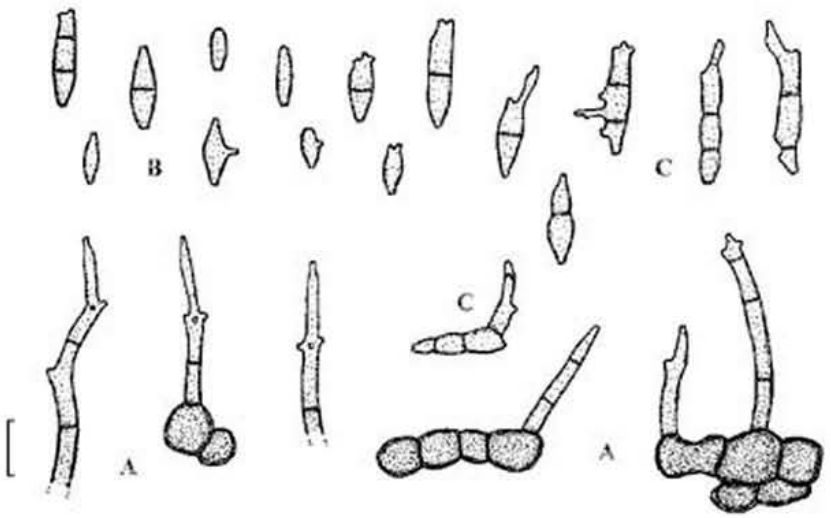


Fig. 4. *Cladosporium indigoferae*.
A. Conidiophores arising from swollen hyphal cells.
B. Conidia. C. Conidia (germination, with microcyclic conidiogenesis).
Scale bar = 10 µm. U. Braun *del.*

thin to slightly thickened, smooth, apex rounded to attenuated, base usually short obconically truncate, hila truncate, 1–2 µm diam., barely thickened, but somewhat darkened-refractive. Microcyclic conidiogenesis observed.

COMMENT: *Cladosporium indigoferae* is an invalid name, published without a Latin description. The type material examined is rather sparse, but some conidiophores and conidia, morphologically agreeing with the features of *Fusicladium* and *Cladophialophora*, has been found. However, the material examined is in too poor a condition for a final treatment and validation, which should be based on cultural characteristics and supported by molecular DNA sequence analyses.

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# Polypores new to Israel — 1: Genera Ceriporiopsis, Postia and Skeletocutis

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**Abstract** — *Ceriporiopsis balaenae*, *Postia rancida*, *P. subcaesia*, and *Skeletocutis percandida* are described and their microstructural features are illustrated according to new material collected in northern Israeli forests. Their distribution, habitat, and taxonomic relationships with other species are discussed.

**Key words** — Mediterranean, annual tinder fungi, diversity, taxonomy

## Introduction

This paper is the first in a series devoted to the study of rare polypores in or new to Israel and contains information on four species from the genera *Ceriporiopsis* Domański, *Postia* Fr., and *Skeletocutis* Kotl. et Pouzar. These genera comprise a lot of annual non-stipitate polypores that have long been attributed to the large genus *Tyromyces* P. Karst. The taxonomy of this unclear genus is rather complicated, and "...its analysis requires much more detailed knowledge of the tropical species" (Corner 1989). In currently accepted taxonomy, the generic delimitation is based mainly on the presence of fiber hyphae in *Skeletocutis*, while fiberless *Postia* and *Ceriporiopsis* are distinguishable on the basis of basidiospore, basidium and hyphal morphology as well as the type of rot

produced, with brown rot characteristic only to *Postia* within the group. The rest of *Tyromyces* remains an assemblage of subdimitic, white rot-producing, sessile polypores, related to *Panus* Fr. on the one hand, and to *Phlebia* Fr. on the other (Corner 1989, Spirin & Zmitrovich 2003).

Seven *Postia* species have been previously reported for Israel by Binyamini (1981) within the boundaries of the traditionally accepted genus *Tyromyces*, namely, *T. fragilis* (Fr.) Donk, *T. floriformis* (Quél.) Bondartsev & Singer, *T. stypticus* (Pers.) Kotl. & Pouzar, *T. mappa* (Overh. & J. Lowe) Ryvarden, *T. tephroleucus* (Fr.) Donk, *T. hibernicus* (Berk. & Broome) Ryvarden, and *T. lacteus* (Fr.) Murrill. Most of these descriptions were repeated in the book "Larger Fungi of Israel (*Ascomycotina* and *Basidiomycotina*)" (Binyamini 1984). At a later date, two other species – *Tyromyces balsameus* (Peck) Murill (Binyamini 1987) and *Postia inocybe* (A. David & Malençon) Jülich (Czederpiltz et al. 2004) – were added to the list of Israeli *Postia* (in the modern sense of this generic name). Here, *Postia rancida* and *P. subcaesia* are treated as new for Israel.

The genus *Ceriporiopsis* was previously not known in Israel; the presence of *Ceriporiopsis balaenae* is indicated in the humid oak forests of Golan Heights. *Skeletocutis percandida* is widely distributed in the West Mediterranean (Ryvarden & Gilbertson 1994); its easternmost locality is reported below.

## Material and methods

Morphological characteristics of our specimens were examined by using a light/dark field microscope (Carl Zeiss Axiostar 1122-100). Measurements were taken using an immersion objective; a total of 30 spores from each specimen were measured. In order to prevent a variation of spore size, 5% of measurements were excluded from the end of each size variation range and are given in parentheses. Potassium hydroxide solution 10% (KOH) and medicinal iodine were used for microscopical analyses. A map (Fig. 1) showing the natural regions of Israel was used in order to demonstrate each new found species distribution. The material used in our analyses can be found in the herbarium of the Institute of Evolution, University of Haifa (HAI, Haifa, Israel).

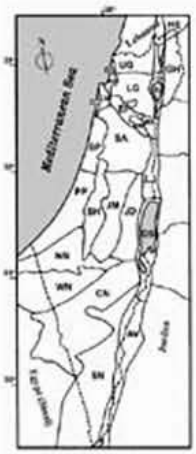
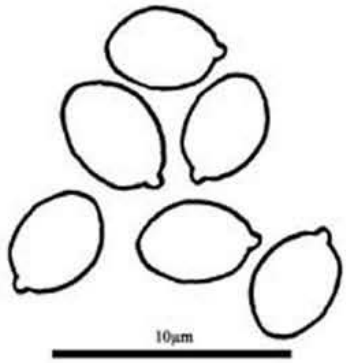



**Fig. 1.** Accepted abbreviations of nature regions of Israel: AP – Akko Plain; AV – Arava Valley; BS – Beit Shean Valley; CC – Carmel Coast; CG – Coast Galilee; CM – Carmel Mount; CN – Central Negev; DS – Dead Sea Area; EP – Esdraelon (Yizreèl) Plain; GH – Golan Heights; GM – Gilboa Mount; HE – Hermon Mount; HP – Hula Plain; JD – Judean Desert; JM – Judean Mts.; LG – Lower Galilee; LJ – Lower Jordan Valley; NN – Northern Negev; PP – Philistean Plain; SA – Samaria; SH – Shefela; SN – South Negev; SP – Sharon Plain; UG – Upper Galilee; UJ – Upper Jordan Valley; WN – Western Negev (Feinbrun-Dothan et al. 1998).

## Taxonomic descriptions

***Ceriporiopsis balaenae*** Niemelä, Nat. Canad. 112: 449 (1985). FIG. 2
≡ *Porpomyces balaenae* (Niemelä) Spirin & Zmitr., Karstenia 43: 80 (2003).

BASIDIOCARPS resupinate, annual, soft when fresh, hard when dry (dry specimen), adnate. HYMENOPHORE poroid, as a single tube layer up to 3 mm thick, pores angular, irregular, 1–2 per mm, honey-yellow, with slightly dentate dissepiments. MARGIN narrow, mucedinous and somewhat lighter than remaining fruitbody. CONTEXT whitish and very thin (barely observable), up to 0.3 mm thick. HYPHAL SYSTEM monomitic. HYPHAE hyaline, with clamps, thin- to slightly thick-walled, branched, 2–4 µm broad (Fig. 2c). CYSTIDIA or other sterile elements lacking. BASIDIA clavate with a basal clamp, 7–25 × 4–5 µm (Fig. 2b). BASIDIOSPORES ellipsoid to subglobose, smooth, hyaline and thin-walled, (3.4–)3.5–4.9 × (1.9–)2–3.5(–3.6) µm, non-amyloid (Fig. 2a).



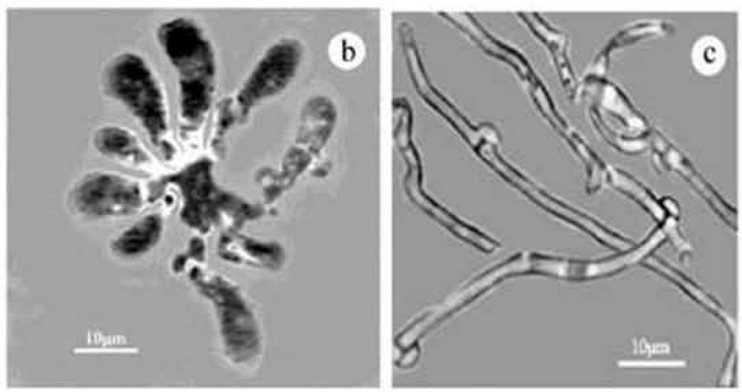




**Fig. 2.** *Ceriporiopsis balaenae* (HAI 0109).
*a* – basidiospores, *b* – basidia, *c* – contextual hyphae.

GENERAL DISTRIBUTION AND HABITAT: *Ceriporiopsis balaenae* is a rare species, originally described from Canada (Niemelä 1985). Nowadays, its scarce distribution includes: EUROPE (Finland, France, Sweden, Russia, Czech

Republic, Slovakia); Asia (Russian Far East, Israel – Fig. 3); also known from the Caribbean (Puerto Rico).

Note: Being associated with a white rot, *C. balaenae* can be easily distinguished in the field due to white or cream-colored resupinate basidiocarps, soft consistence (when fresh) and turning honey-colored on drying. Its closest relative is, evidently, the East-Asian *C. cremea* (Parmasto) Ryvarden (Vampola & Pouzar 1996, Spirin & Zmitrovich 2003), which differs in having strongly cordonic basidiocarps and somewhat longer spores (up to 5.4 µm long according to Spirin & Zmitrovich 2003, up to 6 µm – according to Kinnunen & Niemelä 2005). The other closely related taxon is *C. consobrina* (Bres.) Ryvarden. According to Vampola (Vampola, pers. com. 2008), *C. balaenae* is very closely related to *C. consobrina*, distributed in the Mediterranean. We refer our material to *C. balaenae*, primarily based on spore shape and size (the spores of *C. consobrina* are somewhat wider and subglobose). More comprehensive studies are needed for this species complex.

SPECIMENS EXAMINED: *Ceriporiopsis balaenae*. **ISRAEL. Golan Heights:** Massada, mixed forest of *Quercus calliprinos* and *Q. boissieri*, on deciduous wood, 10.11.2006, leg. Y. Ur, det. I.V. Zmitrovich, D. Tura and V.F. Malysheva (HAI 0109); Bar'am, mainly *Quercus calliprinos* forest, on oak bark, 09.03.2007, leg. & det. I.V. Zmitrovich, D. Tura and V. Malysheva (HAI 0110). **CANADA. Québec:** Poste-de-la-Baleine, *Salix planifolia*, 12.08.1982, leg. & det. T. Niemelä (H, holotype). **RUSSIA. Leningrad Reg.:** Lisino, *Salix caprea*, 03.09.1960, leg. M.A. Bondartseva, det. W.A. Spirin & I.V. Zmitrovich (LE 19420). **FINLAND. Inarin Lappi:** Utsjoki, *Salix*, 16.08.1987, leg. & det. H. Kotiranta (H, dupl. in LE).

*Ceriporiopsis cremea*. **RUSSIA. Kamchatka:** Klyuchi, *Populus suaveolens*, 18.08.1960, leg. & det. E. Parmasto (TAA 13599, isotype in LE).

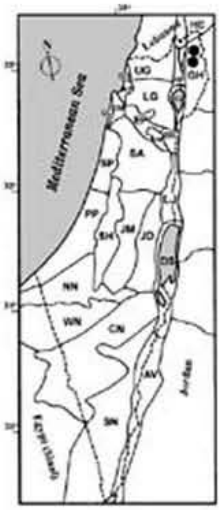



Fig. 3. Distribution of *Ceriporiopsis balaenae* in Israel.

***Postia rancida*** (Bres.) M.J. Larsen & Lombard, Mycotaxon 26: 272 (1986). FIG. 4

≡ *Poria rancida* Bres., Fung. Trident. 2: 96 (1900).

Basidiocarps resupinate, soft, with rancid taste (fresh material). Hymenophore poroid, as a single tube layer up to 5 mm thick. Pore surface white to pale cream, pores angular, 3–4 per mm, with slightly dentate dissepiments. Context white, present as a thin layer up to 1 mm thick. Margin white, cottony and thinning outwards. Hyphal system monomitic. Tramal hyphae hyaline, clamped, richly branched and thin-walled, 2–4 µm wide; contextual hyphae sparingly branched, thin-walled and broader (up to 5 µm across) than ones in tube trama (Fig. 4c). Cystidia not seen. Basidia clavate with a basal clamp, 15–21 × 3–7

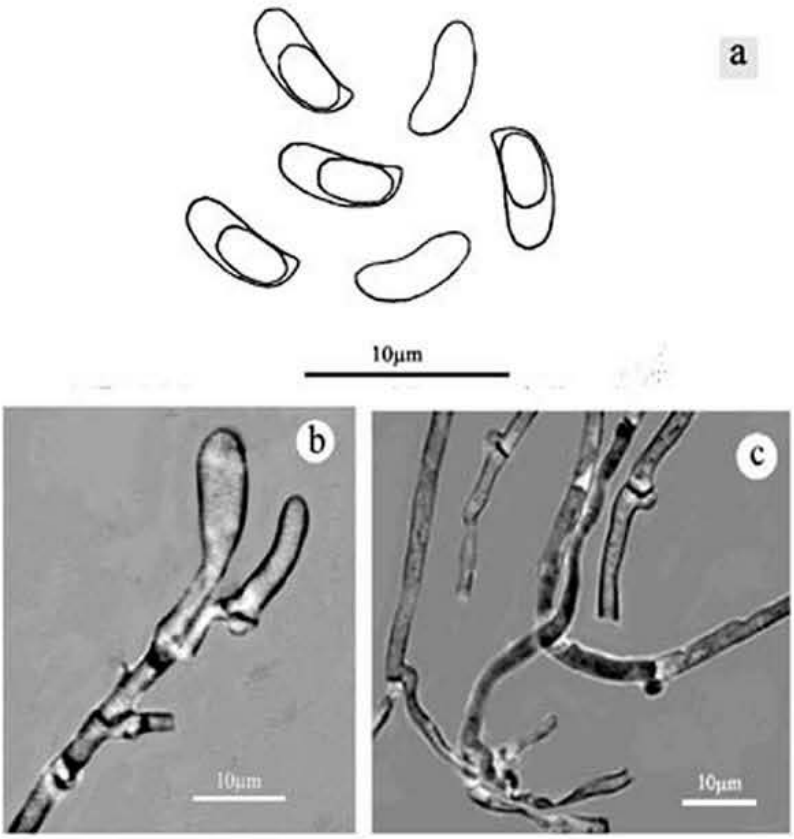



**Fig. 4.** *Postia rancida* (HAI 0108).
*a* – basidiospores, *b* – basidia, *c* – contextual hyphae.

µm (Fig. 4b). BASIDIOSPORES cylindrical, smooth, thin-walled, with oil-drops inside (usually one large oil-drop in the middle part of the spore), 5.9–7.9 × (1.9–)2–3(–3.1) µm, non-amyloid, acyanophilous (Fig. 4a).

GENERAL DISTRIBUTION AND HABITAT: *Postia rancida* occurs mostly in coniferous forests, growing on dead wood of *Pinus* and *Picea*, and more rarely in hardwood forests. Associated with a brown cubical rot. *P. rancida* is a rare species, its general distribution includes: EUROPE (England, Norway, Italy, Poland); NORTH AMERICA (USA); ASIA (Israel – Fig. 5, China).

NOTE: *Postia rancida* can be easily confused with other species of this genus. The main reliable feature of this polypore is the spore size. However, *P. rancida* might be confused with a closely related species *Rhodonia placenta* (Fr.) Niemelä et al. (Niemelä et al. 2005). The spores of the latter are quite similar to those of *P. rancida*, and its common bright rose color is absent in some collections, replaced by cream, pale-lilac or even ochraceous tints. Renvall (Ryvarden & Gilbertson 1994) was of the opinion that *P. rancida* could be a pale

form of *Rhodonia placenta*. Further studies are needed to clarify the identity of these species. In comparison to other *Postia* species, *P. rancida* is the closest relative of *Postia mappa* (Overh. & J. Lowe) M.J. Larsen & Lombard, which also occurs in Israel. Both species are very similar, but *P. mappa* has slightly larger spores (6.5–8.5 µm long) and 1–3 pores per mm. *P. rancida* and *P. mappa* were discussed by Spirin et al. (2006) as possible members of *Rhodonia*.

SPECIMENS EXAMINED: *Postia rancida*. **ISRAEL. Upper Galilee:** Biriya, *Pinus halepensis* forest, on coniferous cone, 07.03.2007, leg. & det. I.V. Zmitrovich, D. Tura and V.F. Malysheva (HAI 0108).

*Postia mappa*. **ISRAEL. Judean mountains:** Shaar Hagai, *Pinus* sp. forest, on dry branches of pine, 26.01.1981, leg. & det. N. Binyamini (TELA).

*Rhodonia placenta*. **RUSSIA. Nizhny Novgorod Reg.:** Kilemarsky Nat. Res., *Picea abies*, 17.08.2004, leg. & det. W.A. Spirin (H) (cream-coloured specimen). **FINLAND. Etelä-Häme:** Mustiala, *Pinus sylvestris*, 27.07.1892, leg. & det. P.A. Karsten (H, holotype of *Physisporus albolilacinus* P. Karst.).

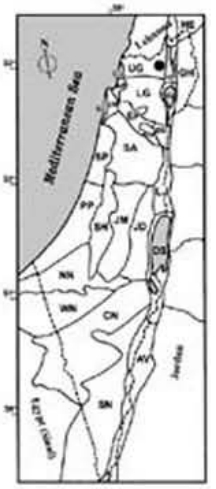



Fig. 5. Distribution of *Postia rancida* in Israel.

***Postia subcaesia*** (A. David) Jülich, Persoonia 11: 424 (1982). FIG. 6
≡ *Tyromyces subcaesius A. David*, Bull. Soc. Linn. Lyon 43: 120 (1974).

Basidiocarp pileate to effused-reflexed. Pileus semicircular, ca 2 × 1 × 0.5 cm, soft in fresh condition, fragile when dry (Fig. 6a). Upper surface weakly zonate, minutely uneven, tomentose with small tufts of hyphae (barely seen with naked eye), middle part bluish (dark blue to blackish on drying). Margin white, thinning outwards. Hymenophore poroid, as a single layer up to 5 mm thick, concolorous with pore surface. Pores 4–5 per mm, white-grayish, angular. Context white, present as a thin layer up to 2 mm thick. Hyphal system monomitic. Generative hyphae hyaline, with clamps, branched, slightly sinuous, thin-walled to thick-walled, wider in the context (up to 5 µm across; Fig. 6d). Cystidia not seen. Basidia cylindrical to lageniform, with a basal clamp and 4 sterigmata, 11–15 × 3–4.5 µm (Fig. 6b). Basidiospores cylindrical to allantoid, smooth, hyaline, thin-walled with oil-drops inside (usually two droplets), 4.9–5.9 × (0.9–)1–1.5(–1.6) µm, non- or weakly amyloid; bluish-gray in mass (Fig. 6c).

General distribution and habitat: According to our current knowledge on the species limits within the *Postia caesia*-complex, *P. subcaesia* is known as growing on various hardwood substrates from: Europe (England, Ireland,

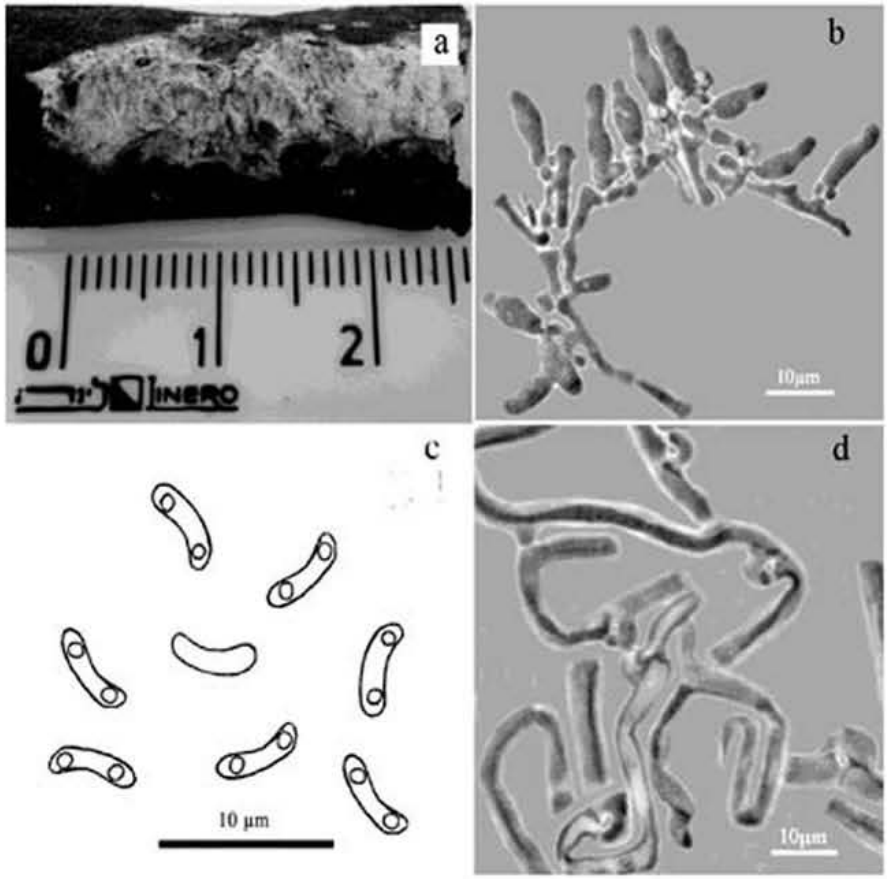



Fig. 6. *Postia subcaesia* (HAI 0107).
*a* – upper view of basidiocarps, *b* – basidia, *c* – basidiospores, *d* – contextual hyphae.

Norway, Czech Republic, Sweden, Germany, Switzerland, France, Denmark, Italy, Belgium, Poland, Ukraine, the Netherlands, Slovakia, Hungary, Greece, Russia); Asia (Israel – Fig. 7, India, China); North America (USA, Canada).

Note: Members of the *Postia caesia*-complex have an isolated position within the genus *Postia*. All are bluish, medium- to small-sized polypores causing a brown rot; the species do not produce chlamydospores in culture (David 1980), and their basidiospores are faintly amyloid. The name *Cyanosporus* McGinty might be applied to this group (Pieri & Rivoire 1998) if raised to generic rank.

There are many difficulties with the species limits in this complex. David (1974, 1980) was the first to split *P. caesia* sensu lato into separate species; other species were added later by Niemelä et al. (2001) and Pieri & Rivoire (2005). *Postia subcaesia* and *P. alni* Niemelä & Vampola belong to the narrow-spored species within the *Postia caesia* complex (spore width does not exceed 1.5 µm), while *P. caesia* (Schrad.) P. Karst. s. str., *P. luteocaesia* (A. David) Jülich, and *P. mediterraneocaesia* M. Pieri & B. Rivoire constitute the wide-spored group

(spore width mostly more than 1.5 µm). *Postia alni*, the closest relative of *P. subcaesia*, differs by its smaller fruitbodies and matted upper surface; its tramal hyphae swell inwards and become gelatinized in KOH. However, there are some confusing East-Asian collections that do not fit well with both *P. alni* and *P. subcaesia*. Recent molecular data (Yao et al. 2005) suggest that the taxonomy of this group should be more complicated than currently found.

SPECIMENS EXAMINED: ISRAEL. SAMARIA: Reihan, *Pinus halepensis* and *Quercus calliprinos* forest, on unidentified substrate, 19.02.2007, leg. Y. Ur, det. I.V. Zmitrovich, D. Ţura and V.F. Malysheva (HAI 0106). UPPER GALILEE: Meron, *Pinus halepensis* and *Quercus calliprinos* forest, on deciduous wood, 09.03.2007, leg. & det. I.V. Zmitrovich, D. Ţura and V.F. Malysheva (HAI 0107). FRANCE. ISÈRE: region de Crémieu, *Malus*, 10.1968, leg. & det. A. David (H ex LY-AD 652, isotype of *Tyromyces subcaesius*). MÈRE BOITIER: Saone et Loire, *Quercus*, 29.07.1965, leg. & det. A. David (H ex LY-AD 150).

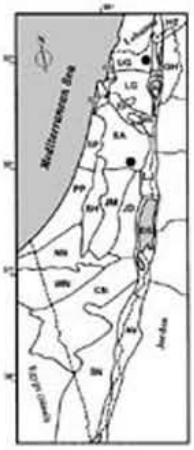


Fig. 7. Distribution of *Postia subcaesia* in Israel.

***Skeletocutis percandida*** (Malençon & Bertault) Jean Keller, Persoonia 10: 353 (1979). FIG. 8

= *Poria percandida* Malençon & Bertault, Acta Phytotax. Barcinon. 8 : 35 (1971).

BASIDIOCARP annual, resupinate, sprawled on wood as a thin layer, up to 1–1.5 mm thick, soft when fresh, hard when dry (dry specimen), peelable (Fig. 8). HYMENOPHORE poroid, as a single whitish tube layer up to 0.5 mm thick. Pores round, 3–4 per mm, with slightly thick dissepiments, pore surface white to cream with fine pinkish tint. MARGIN white, thin, byssoid to cordonic, broadly sterile. CONTEXT very thin, cottony, white-colored. HYPHAL SYSTEM dimitic. GENERATIVE HYPHAE hyaline with clamps, branched, easily encrusted, thin- to thick-walled, 2–3 µm wide. SKELETAL HYPHAE hyaline, sinuous, sparingly branched and thick-walled, 2–5 µm wide. CYSTIDIA not seen, but fusoid cystidioles are present in the hymenium. BASIDIA ovoid to clavate, 15–20 × 5–7 µm, with a basal clamp and 4 sterigmata. BASIDIOSPORES cylindrical (slightly curved), hyaline, smooth, thin-walled with oil drops inside, (6.4–)6.5–8 × 2–3(–3.1) µm, non-amyloid.

GENERAL DISTRIBUTION AND HABITAT: *Skeletocutis percandida* is widely distributed in the Mediterranean (David 1982, Vampola & Forstinger 1993, Bernicchia 2005, Pieri & Rivoire 2007). To the best of our knowledge, its general distribution includes: EUROPE (Italy, France, Spain, Portugal, Greece); ASIA (China, Israel – Fig. 9); AFRICA (Zimbabwe).


**Fig. 8.** *Skeletocutis percandida* (HAI 0111)effused basidiocarp with byssoid cordonic margin.

NOTE: This species is thoroughly described by David (1982) and Niemelä (1998). It is characterized by strongly cordonic basidiocarps and relatively long cylindrical spores 4.9–7.2 × 2.2–2.8 µm. *S. alutacea* (J. Lowe) Jean Keller is another cordonic *Skeletocutis* species known in the Northern Hemisphere; but differs by smaller pores and spores (Niemelä 1998, Spirin 2005). An apical incrustation on skeletal hyphae occurs at the dissepiments edges in both *S. alutacea* and *S. percandida*; this feature is not reported for other *Skeletocutis* species (only generative hyphae are encrusted), except *S. novae-zelandiae* (G. Cunn.) P.K. Buchanan & Ryvarden (Buchanan & Ryvarden 1988), which could be closely related to them. Taxonomy of these species is poorly worked-out.

SPECIMENS EXAMINED: *SKELETOCUTIS PERCANDIDA*. **ISRAEL. UPPER GALILEE:** Meron, *Pinus halepensis* and *Quercus calliprinos* forest, on coniferous wood, 09.03.2007, leg. I.V. Zmitrovich, D. Tura and V.F. Malysheva, det. W.A. Spirin (HAI 0111). – *SKELETOCUTIS ALUTACEA*. **RUSSIA. NIZHNY NOVGOROD REG.:** Razino, *Pinus sylvestris*, 05.08.1998, leg. & det. W.A. Spirin (LE 208266).

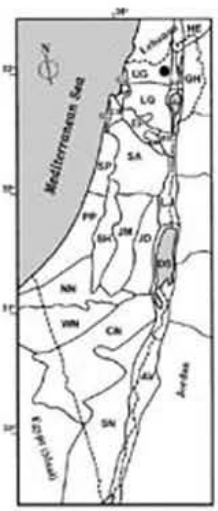



**Fig. 9.** Distribution of *Skeletocutis percandida* in Israel.

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MYCOTAXON

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# South Florida microfungi: a new species of *Stanjehughesia* (hyphomycetes) from *Sabal* palm

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**Abstract**—*Stanjehughesia floridensis* anam. sp. nov. is described and illustrated from rachides and petioles of dead leaves of *Sabal* sp. in southeastern Florida, USA. The fungus is characterized by conidiophores reduced to monoblastic, brown, cylindrical, ampulliform or lageniform, solitary or clustered conidiogenous cells and obclavate to cylindric-obclavate, smooth, brown, 5-9 euseptate conidia with dark bands at the septa. It is compared with morphologically similar taxa, and a synoptical table of accepted species of *Stanjehughesia* is provided.

**Key words**—anamorphic fungi, palm fungi, *Sporidesmium*

## Introduction

During a short-term survey of saprophytic microfungi occurring on plant debris from southeastern Florida, an interesting hyphomycetous anamorph was abundantly collected on *Sabal* palm dead leaves. The fungus possesses conidiogenesis and conidial features that clearly suggest a placement within the genus *Stanjehughesia* Subram. (Subramanian 1992, Wu & Zhuang 2005), but morphologically differs from all previously known species. Therefore it is described here as new. The type specimen and other specimen examined are deposited in the Herbarium of the U. S. National Fungus Collections (BPI).

## Taxonomic description

***Stanjehughesia floridensis*** G. Delgado, anam. sp. nov. Figs. 1-6

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*Ad fungus anamorphicus, hyphomycetes, pertinens. COLONIAE in substrato naturali effusae, nigrae. MYCELIUM superficiale, ex hyphis ramosis, septatis, pallide brunneae vel brunneae, 1.5–2 µm latis compositum. CONIDIOPHORA absentia. CELLULAE CONIDIOGENAE monoblasticae, determinatae, plerumque intercalares, cylindricae, ampulliformes vel lageniformes, erectae, rectae vel flexuosae, crassitunicatae, laevia, discretae vel gregares,*

*brunneae vel atro-brunneae, 5-9 × 2-3.5 µm, ad basim 5-6 µm crassa, at apicem 1.5-2 µm crassa et truncatae.* CONIDIORUM SECESSIO *schizolytica.* CONIDIA *holoblastica, solitaria, sicca, acrogena, recta vel leviter curvata, obclavata vel cylindrico-obclavata, brunneae, laevia, 5-9 euseptata, leviter constricta ad septa, 32-48 × 6-7 µm, cellula apicalis rotundata vel leviter spatulata, pallide brunnea, cellula basalis obconica, saepe pallide brunneae, 5-7 × 4-4.5 µm, in hilum truncatum breviter attenuata.* TELEOMORPHOSIS *ignota.*

HOLOTYPE — **UNITED STATES. FLORIDA: Broward Co.,** POMPANO BEACH, on rachides and petioles of dead leaves of *Sabal* sp., V.27.2007, coll. G. Delgado (BPI 878270).

ETYMOLOGY — Latin, *floridensis*, referring to the state of Florida, where the fungus was collected.

Anamorphic fungi, hyphomycetes. COLONIES on natural substrate effuse, black. MYCELIUM superficial, composed of branched, septate, pale brown to brown hyphae, 1.5-2 µm wide. CONIDIOPHORES absent. CONIDIOGENOUS CELLS monoblastic, determinate, mostly intercalary, cylindrical, ampulliform or lageniform, erect, straight or flexuous, thick-walled, smooth, solitary or disposed in caespitose clusters to large extensive groups, brown to dark brown, 5-9 × 2-3.5 µm, 5-6 µm wide at the bulbous base, 1.5-2 µm wide at the truncate apex. CONIDIAL SECESSION schizolytic. CONIDIA holoblastic, solitary, dry, acrogenous, straight or slightly curved, obclavate to cylindric-obclavate, brown, smooth-walled, 5-9 euseptate, with dark bands and slightly constricted at the septa, 32-48 × 6-7 µm; apical cell rounded or slightly spathulate, light brown; basal cell obconical, often paler, 5-7 × 4-4.5 µm, tapered to a narrow, truncate hilum. TELEOMORPH unknown.

OTHER SPECIMENS EXAMINED—UNITED STATES. FLORIDA: Broward Co., POMPANO BEACH, on rachides and petioles of dead leaves of *Sabal* sp., XI.2004, coll. G. Delgado (BPI 877806A).

## Discussion

Subramanian (1992) reassessed the anamorphic genus *Sporidesmium* Link and introduced *Stanjehughesia* to accommodate a group of five species characterized by solitary, euseptate conidia and very reduced or absent conidiophores consisting of simple conidiogenous cells. Reblová (1999) did not accept Subramanian's arrangement, considering it to be schematic, diagnostically valuable but phylogenetically unacceptable. She preferred to retain the type species, *Stanjehughesia hormiscioides* (Corda) Subram. in *Sporidesmium*, as *Sp. hormiscioides* Corda. However, additional species have been described or transferred to *Stanjehughesia* following Subramanian's concept (McKenzie 1995, Mena et al. 2001, Wu & Zhuang 2005). Wu & Zhuang provided an account of the genus and described four species collected on bamboo culms and dead branches from China. They retained the specific name *St. vermiculata* (Cooke) Subram. (=*Sp. vermiculatum* (Cooke) M.B. Ellis) for a specimen collected on rotten wood and bark of *Fagus sylvatica* L. and *Quercus* sp. Hughes

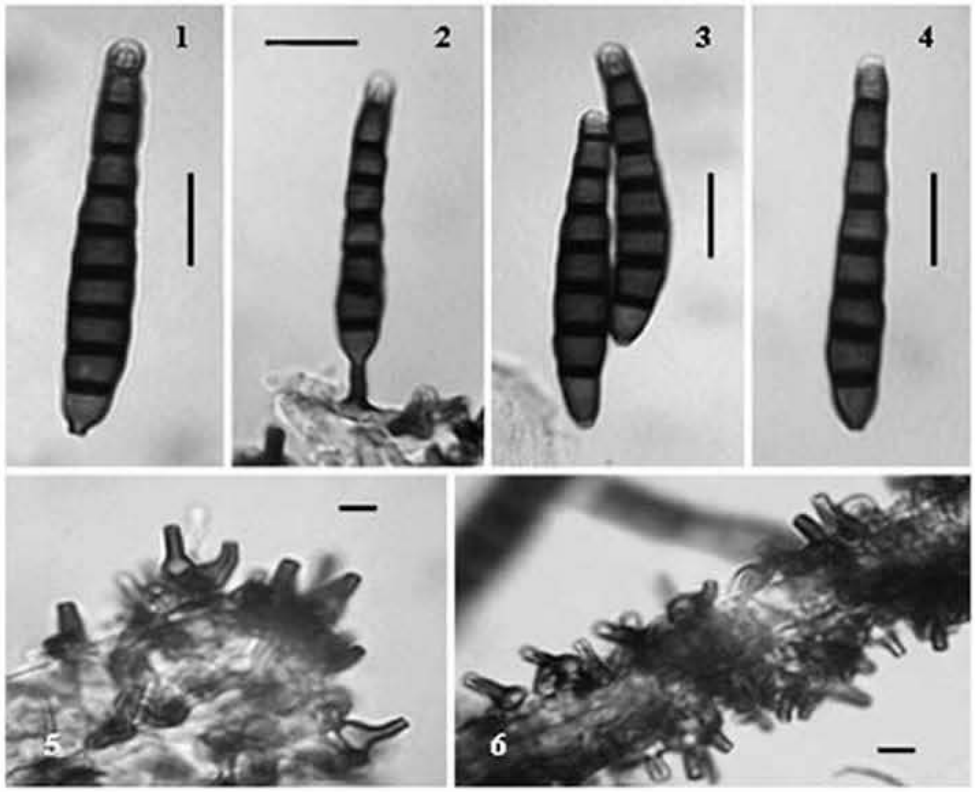


**Fig. 1-6.** *Stanjehughesia floridensis*, from holotype (BPI 878270). 1, 3-4. Conidia. 2. Conidium attached to a conidiogenous cell. 5-6. Conidiogenous cells on natural substrate.
Scale bars: 1-4 = 10 µm; 5-6 = 5 µm.

(1958) previously considered *Sp. vermiculatum* and *Sp. caespitulosum* (Ellis & Everh.) M.B. Ellis (=*St. caespitulosa* (Ellis & Everh.) Subram.) to be synonyms of *Sp. hormiscioides* (=*St. hormiscioides*). The descriptions and illustrations provided by Ellis (1958, 1976) show that *Sp. vermiculatum* and *Sp. hormiscioides* are remarkably similar and should be considered conspecific under their current placement in *Stanjehughesia*, whereas *St. caespitulosa* differs enough to be retained as a separate species. Reblová (1999) also discussed the variations in conidiophore morphology of another species, *St. larvata* (Cooke & Ellis) Subram. (=*Sp. larvatum* Cooke & Ellis) under natural and culture conditions, retaining the latter name for the anamorph of *Miyoshiella larvata* Reblová. Several collections having distinct, macronematous, multicelled conidiophores and 0-1 proliferating conidiogenous cells support Reblová's opinion to maintain this anamorph in *Sporidesmium* (Hughes & Illman 1974, Matsushima 1975, Reblová 1999, Wu & Zhuang 2005).

Recent phylogenetic studies suggest that *Sporidesmium* and morphologically similar genera including *Stanjehughesia* are a polyphyletic, artificial assemblage

Table 1. Morphological features of accepted species of *Stanjehughesia*.

| Species | Conidiophores | Conidiogenous cells | | Conidia | | | |
|---|---|---|---|---|---|---|---|
| | | Shape | Size (mm) | Shape | Septation | Wall texture | Size (mm) |
| *St. caespitulosa* | Absent | Bulbous | 12-15 × 8-11 | Cylindrical to subfusiform | 8-20 euseptа | Smooth | 65-150 × 13-17 |
| *St. decorosa* | Present, reduced | Cylindrical | 20-30 × 10-15 | Obclavate | 10-14 euseptate | Verrucose, striated | 170-200 × 12-17 |
| *St. fasciculata* | Present, reduced | Obclavo-cylindrical to obclavate | Up to 30 | Obclavate or fusiform | 9-15 euseptate, 1-2 distoseptate at the apex | Verruculose | 60-118 × 7-11 |
| *St. floridensis* | Absent | Cylindrical, ampulliform or lageniform | 5-9 × 2-3.5 | Obclavate to cylindric-obclavate | 5-9 euseptate | Smooth | 32-48 × 6-7 |
| *St. fusiformis* | Absent | Cylindrical to ampulliform | 11-15 × 5-7 | Fusiform, obclavate to obclavate-rostrate | 6-8 euseptate | Smooth | 65-85 × 7-8 |
| *St. hamatiella* | Absent | Cylindrical to ampulliform | 7-14 × 5-7 | Obclavate to obclavate-rostrate and hamate | 11-14 euseptate, 6-9 distoseptate | Smooth | 70-140 × 7-8.5 |
| *St. hormiscioides* | Absent | Obclavate to flask shape | 10-25 × 5-11 | Subcylindrical to cylindric-fusoid | 12-26 euseptate | Smooth | 65-270 × 10-17 |
| *St. minima* | Absent | Cylindrical to ampulliform | 8-11 × 2-3.5 | Fusiform or ellipsoidal | 6-8 euseptate | Smooth | 34-48 × 8.5-10 |
| *St. nigroacus* | Absent | Lageniform to doliiform | 8-14 × 7-12 | Acicular | 23-36 euseptate | Smooth | 130-214 × 10-12 |
| *St. polypora* | Absent | Cylindrical to ampulliform | 13-18 × 3.5-5 | Fusiform, obclavate to obclavate-rostrate | 15-20 euseptate | Smooth | 110-165 × 10-12 |

of unrelated anamorphs with affiliations to *Dothideomycetes* and *Sordariomycetes*, and the morphological characters currently used to delimit the genera are not phylogenetically significant (Shenoy et al. 2006). In the present case however, Subramanian's concept of *Stanjehughesia* is followed and considered valid for diagnostic purposes, until further investigations continue refining the taxonomic status and phylogenetic relationships among *Sporidesmium* and related anamorphs (Shenoy et al. 2007). Accordingly, *Stanjehughesia* would currently comprise ten species, whose morphological features are compared in Table 1.

Some *Stanjehughesia* species fit well with the generic concept of *Janetia* sensu Goh & Hyde (1996). These authors formally expanded the original concept of Ellis (1976), based on *Janetia euphorbiae* M.B. Ellis, to include species producing obclavate or cylindrical, euseptate or distoseptate conidia borne on monoblastic and/or polyblastic denticulate conidiogenous cells on usually integrated and single conidiophores, but also aggregated in synnematal conidiomata. Despite the morphological similarities, *Stanjehughesia* species can be separated from *Janetia* on the basis of their monoblastic, non-denticulate conidiogenous cells and lignicolous habitat (Mena et al. 2001, Wu & Zhuang 2005).

*Stanjehughesia floridensis* is morphologically close to *St. caespitulosa* and *St. hormiscioides* (Ellis 1958, 1976), which also have smooth-walled, brown conidia with dark bands associated with the septa. They also share similarly shaped conidiogenous cells that are solitary or disposed in caespitose clusters to large groups, terminally and laterally arranged on the hyphae. However, *St. caespitulosa* has cylindrical to subfusiform, 8-20 euseptate, larger conidia (65-150 × 13-17 µm) and larger conidiogenous cells (12-15 × 8-11 µm). *St. hormiscioides* is distinguished from the new species by its clavate when young, fusiform when mature, also larger conidia (65-270 × 10-17 µm), with 12-26 septa, and larger conidiogenous cells (10-25 × 5-11 µm). Another species, *St. minima* W.P. Wu (Wu & Zhuang 2005) resemble *St. floridensis* in conidial length, texture, color, number of septa and the morphology of conidiogenous cells, but differs in having fusiform or ellipsoidal, wider conidia (8.5-10 µm) without dark bands at the septa.

Among species of *Janetia* with similar conidial morphology, *J. euphorbiae* (Ellis 1976) could be compared with *St. floridensis*, but the presence of polyblastic conidiogenous cells usually bearing 2-4 thick denticles and 3-6 septate, shorter conidia (18-36 µm) clearly separate both taxa. *Janetia capnophila* S. Hughes (Hughes 1983) is also morphologically close to *St. floridensis* in having inflated, constantly monoblastic conidiogenous cells and narrowly obclavate conidia that are rounded at the apex and conico-truncate at the base. However, *J. capnophila* has (7-)9-13(-16) septate, larger conidia (80-125(-145) × 10.8-13.5(-16.2) µm), with conspicuously darker basal cells and up to 7 conidial cells that may

become conidiogenous. It also occurs intimately associated with sooty moulds, developing a thin, sparse, irregular network of hyphae on leaf surfaces.

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# Type studies in the genus *Crepidotus*

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**Abstract** — Type collections of six *Crepidotus* species described from Chile, Sri Lanka, and the USA were studied. Microscopic characters are described for each taxon. Macroscopic features provided in the original diagnoses have been supplemented by the micromorphologic variations noted in each specimen; these comparisons support our conclusion that each of the type specimens shares the same set of features exhibited by a previously described species and thus represents part of its variation. We therefore propose that *Crepidotus brunswickianus* should be synonymised with *C. croceitinctus*; *C. cuneiformis*, *C. cystidiosus* and *C. truncatus* with *C. applanatus*; *C. grummosopilosus* with *C. calolepis*; and *C. maximus* with *C. albescens*. Descriptions, illustrations and discussions are provided.

**Key words** — *Crepidotaceae*, new synonyms, taxonomy, wood-inhabiting fungi

## Introduction

Many recent publications have addressed old *Crepidotus* species descriptions in an effort to uncover taxonomically reliable microscopic characters. Ranges of variability for micromorphological features such as cheilocystidia, pileipellis, trama tissues, and basidiospores have become better known for many taxa. SEM examinations of basidiospores have also revealed important spore ornamentation patterns useful for recognizing lineages in the genus (Bandala & Montoya 2000ab, Bandala et al. 2006, Gonou-Zagou & Delivorias 2005, Horak & Desjardin 2004, Pegler & Young 1972, Senn-Irlet 1995, Senn-Irlet & De Meijer 1998). A number of described *Crepidotus* taxa still need additional documentation of anatomical patterns (especially microscopical) to place them within the current taxonomic system of the genus. The morphological patterns of variation reported for some taxa seem, however, to be associated more with morphologic plasticity among individual specimens than with taxonomically

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significant support for species isolation. In this paper we compare *C. albescens*, *C. applanatus*, *C. calolepis*, and *C. croceitinctus* with six type specimens and eight collections from Chile, the USA and Sri Lanka (Ceylon) . Reexamination revealed that the extremes of some macro- and microscopic features overlap within the range of the morphological variation reported by one or another of these species. From this we conclude that the six types examined do not represent discrete taxonomic entities but rather should be assigned to a previously described species. Descriptions, illustrations and discussions of each are presented.

## Materials and methods

Hand sections of dried basidiomes were mounted and examined microscopically in 3% KOH or Congo red with all measurements taken in KOH. Basidiospore measurement notations and SEM analytical methodology follow Bandala et al. (1999, 2006) and Bandala & Montoya (2000a, 2004). $\overline{X}$ refers to the length × width of *n* basidiospore mean (for one specimen) or range of means (for more than one specimen); similarly, *Q* corresponds to the mean or range of means of the basidiospore length/width ratio. Herbarium acronyms follow Holmgren et al. (1990).

## Taxonomy

***Crepidotus brunswickianus*** (Speg.) Sacc.,
Syll. Fung. 9: 116, 1891. PLATES 1 & 7A-C
Bas.: *Agaricus brunswickianus* Speg., Bol. Acad. Nac. Cien. Córdoba 11: 13, 1887.

HOLOTYPE: CHILE. TIERRA DE FUEGO: Voces Bay, May 1882, *C. Spegazzini 17022* (LPS, Spegazzini Fungi Patagonici No. 19).

OTHER MATERIAL STUDIED — ARGENTINA. NEHUEQUÉN: Quetrihué, 15.V.1952, *R. Singer M 641*. Puerto Manzano, 18.V.1952, *R. Singer M 736* (both at LIL); 9.IV. 1963, *R. Singer 3365* (MICH). VENEZUELA. MERIDA: S of Santo Domingo, Laguna Negra and Laguna de Los Palos, 31.VII.1958, *R.W.G. Dennis 1760* (K) (all as *C. brunswickianus*).

Macroscopic description provided by Spegazzini (1887):

*"... PILEUS dimidiatus... -3 × 1.5 in. ... antice rotundatus, postice subreniformis v. cuneatus, in STIPITE brevissimo, albo, dense irregulariterque papilato-tomentosulo, crassiusculo productus, margine integro, incurvo donatus, glaber, laevissimus, pallide melleus v. fulvo-lutescens, sub iove pluvio hygrophanus; LAMELLAE pileo concolores confertiusculae, polimacriae, antice rotundato-acutatae, postice attenuato-decurrentes, non confluentes, acie integerrimae..."*

Microscopic features recovered from the holotype:

BASIDIOSPORES 7–8.5 (–9.5) × (4.5–) 5–6.5 (–7) µm, $\overline{X}$= 7.6 × 5.7 µm, *Q* = 1.30, broadly ellipsoid, at times rather subglobose, suprahilar depression not or weakly developed, verrucose to moderately coarsely verrucose, ochraceous to yellowish-brown, ornamentation darker, then orangish-brown in mass, thick-

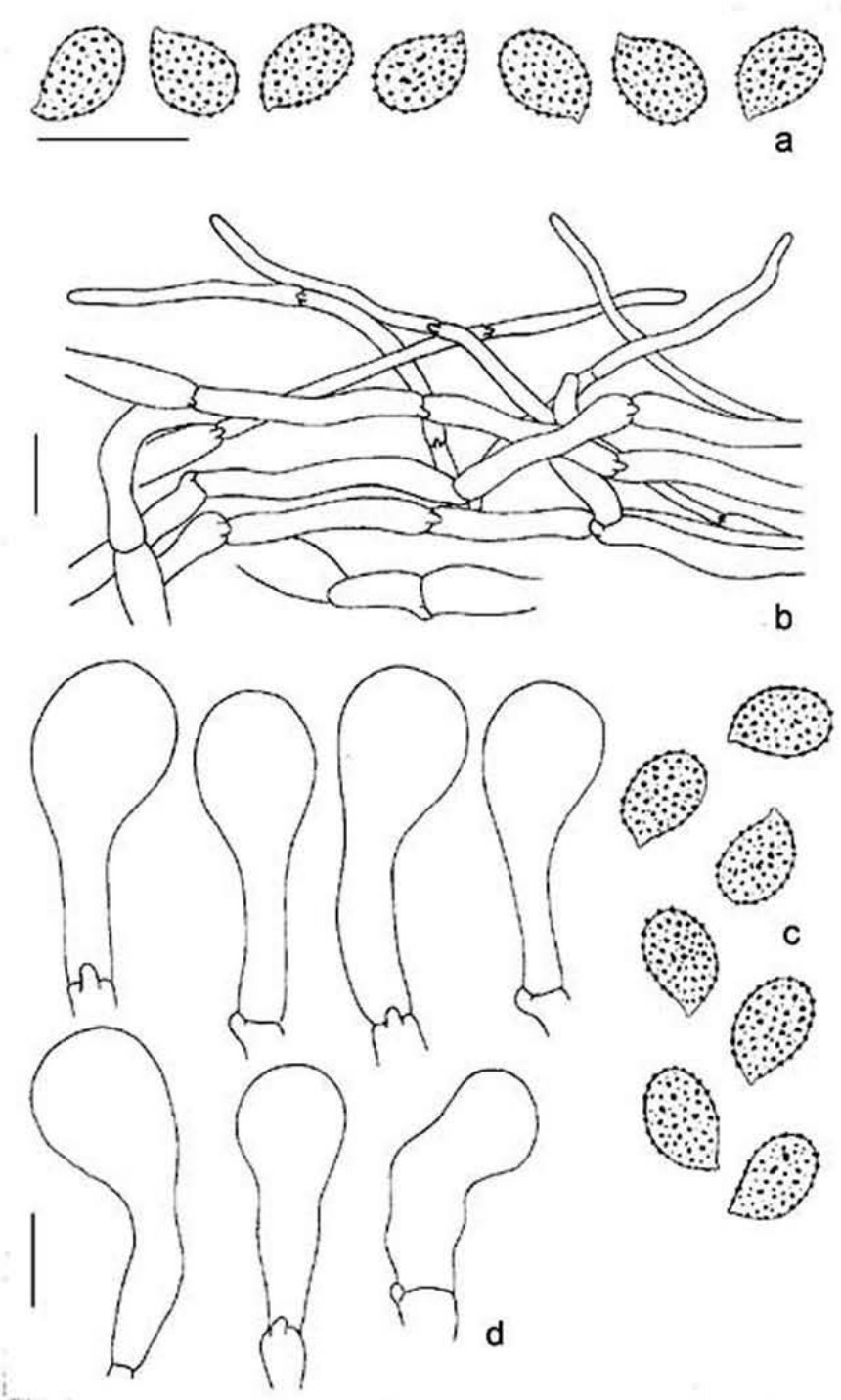


Plate 1. *Crepidotus croceitinctus*. a. Basidiospores (Spegazzini 17022, holotype of *Agaricus brunswickianus*). b. Pileipellis. c. Basidiospores. d. Cheilocystidia (Singer 3365). Scale bar = 10 µm, except b = 15 µm.

walled (≤ 0.5µm wide); under SEM the ornamentation consists of distinctive rounded or more or less truncate, hemispheric to conical (occasionally somewhat rod-like), isolate verrucae, at times two or more verrucae are joined appearing irregular in shape or forming short bulges. Tissues hardly revived with KOH but TRAMA of both PILEUS and HYMENOPHORE consists of non-gelatinized, hyaline, clamped hyphae. BASIDIA, CHEILOCYSTIDIA and PILEIPELLIS not recovered.

COMMENTS — The type consists of incomplete basidiome fragments in such poor condition that some characters appear to be missing completely. As Singer (1947, 1953) also noted, only basidiospore and some tissue characters could be recovered from Spegazzini's specimen. The holotype basidiospores recall the group of taxa close to *C. palmarum* Singer and *C. croceitinctus* in *Crepidotus* section *Sphaerula* Hesler & A.H. Sm. (sensu Senn-Irlet 1995, Senn-Irlet & De Meijer 1998). The basidiospore data, combined with the macroscopic features described by Spegazzini (1887), suggest that *C. brunswickianus* is conspecific with *C. croceitinctus*, a species characterized by basidiomes that are distinctly yellow (or yellowish to orange), possess broadly ellipsoid, verrucose basidiospores, clavate or irregularly (due to outgrowths) clavate cheilocystidia, and a pileipellis that is basically a cutis (Bandala & Montoya 2000b, Pereira 1990, Senn-Irlet & De Meijer 1998, Singer 1973). Specimens from Argentina and Venezuela possessing similar microcharacters as *C. croceitinctus* and originally referred by Dennis (1961) and Singer (1973) to *C. brunswickianus* have basidiospores and cheilocystidia measuring 5.5–9(–9.5) × 4.5–7.5(–8) µm ($\overline{X}$ = 6.5–8.4 × 5.2–6.9 µm, $Q$ = 1.20–1.40) and (25–)28–55 × 4–10(–13) × (apex) 8–17(–19) µm respectively (Plate 1b-d). SEM micrographs (Plate 7a-c) reveal that the basidiospores of all samples show the shape and ornamentation that are distinctive of *C. croceitinctus*. The similarity in microcharacters and the fact that both Dennis and Singer described their specimens as having yellow pilei support our hypothesis that the South American collections represent a variation of *C. croceitinctus*. Illustrations in Horak (1964) and data in Lazo (1971) "... chapeau... jaune orange... Lames oranges... spores 7–8.3 × 5 µm, spinuleuses... cheilocystidia 25–30 × 6–9.5 µm, claviformes, versiformes..." also support this conclusion. Therefore, we assign the holotype of *C. brunswickianus* to *C. croceitinctus* and propose the following synonymy:

***Crepidotus croceitinctus*** Peck, Ann. Rep. N.Y. Stat. Mus. 39: 72, 1886.
Syn.: *Agaricus brunswickianus* Speg., Bol. Acad. Nac. Cien. Córdoba 11: 113, 1887.
≡ *Crepidotus brunswickianus* (Speg.) Sacc., Syll. Fung. 9: 116, 1891.

Singer & Digilio (1951) and Horak (1979) appear to have referred collections characterized by ellipsoid basidiospores and cylindric to +/- narrowly lageniform cheilocystidia to *C. brunswickianus*. These could be better referred to *C. luteolus* (Lambotte) Sacc. or *C. icterinus* Singer (cf. descriptions in Senn-Irlet 1995, Singer 1973).

***Crepidotus cuneiformis*** Pat.,

Bull. Soc. Mycol. France 18: 173, 1902. PLATES 2A, 3A & 7D

HOLOTYPE: **GUADALOUPE.** Bois des Basses Terres, no date, *P. Duss 31* (FH, Patouillard Herbarium No. 405).

Macroscopic description provided by Patouillard (1902):

*"... CHAPEAU convexe-plan, incurve en avant, striolé sur les bords, atténué en coin en arrière, CHARNU, mou, glabre, brun pâle, large de 8-12 millim. LAMES larges, inégales, brunnâtres, peu serrées, molles, se prolongeant jusqu'au point d'insertion..."*

Microscopic features recovered from the holotype:

BASIDIOSPORES 5.5–7 × 5.5–6.5 (–7) µm, $\overline{X}$ = 6.1 × 5.9 µm, $Q$ = 1.03, globose to subglobose, some somewhat cuneate towards the apiculus, finely and densely spinulose to spinulose-verruculose seen under light microscope, baculate when observed under SEM, i.e. with more or less homogeneous, short or moderately long, rod-like or conical protuberances which are apically truncate, small verrucae can be also present among the bacules; thick-walled (≤ 0.5 µm thick), yellowish to yellowish-brown. BASIDIA and elements of hymenium collapsed and not recovered. CHEILOCYSTIDIA not observed, lamellae edges collapsed and hardly revived with KOH. PILEIPELLIS very collapsed, but apparently a cutis composed of hyaline (occasionally pale yellowish hyphae are intermixed), thin-walled, cylindric hyphae, 5–10 µm diam., with undifferentiated terminal elements; in some segments the hyphae at times show a kind of incrusted, colorless, residual material. PILEUS TRAMA with hyaline, cylindric to subventricose, thin-walled hyphae 5–20 (–30) µm diam., some oleiferous-like hyphae (yellowish) present. HYMENOPHORAL TRAMA subregular to subirregular, composed of cylindric to subventricose, thin-walled, hyaline hyphae, with some barely punctate segments. CLAMP CONNECTIONS present in all tissues.

COMMENTS— Macroscopically, elements of the type collection recall small forms of *C. applanatus*. The shape, texture, and color of pileus recorded by Patouillard (1902) fit well within the range of variability of this species. Although Hesler & Smith (1965) also noted the collapsed lamellae edges in the holotype, they observed few clavate cheilocystidia. Pegler (1983) reported clavate to ventricose or ampullaceous cheilocystidia in Patouillard's specimen. The cheilocystidia in combination with the globose, baculate (seen under SEM) basidiospores and pileipellis cutis bearing colorless hyphae, support considering the Duss 31 specimen as conspecific with *C. applanatus*. The "...brun pâle..." pileus of *C. cuneiformis*, apparently the primary character used by Hesler & Smith (1965) to separate it from the *C. applanatus* group is here regarded more as an intermediate form of *C. applanatus* than representing an independent species. Previously Singer (1947) referred the Duss 31 specimen to *C. aquosus* Murrill, which, however, is distinguished from *C. applanatus* by long, narrowly sublageniform cheilocystidia (Singer 1973, Senn-Irlet & De Meijer 1998). The

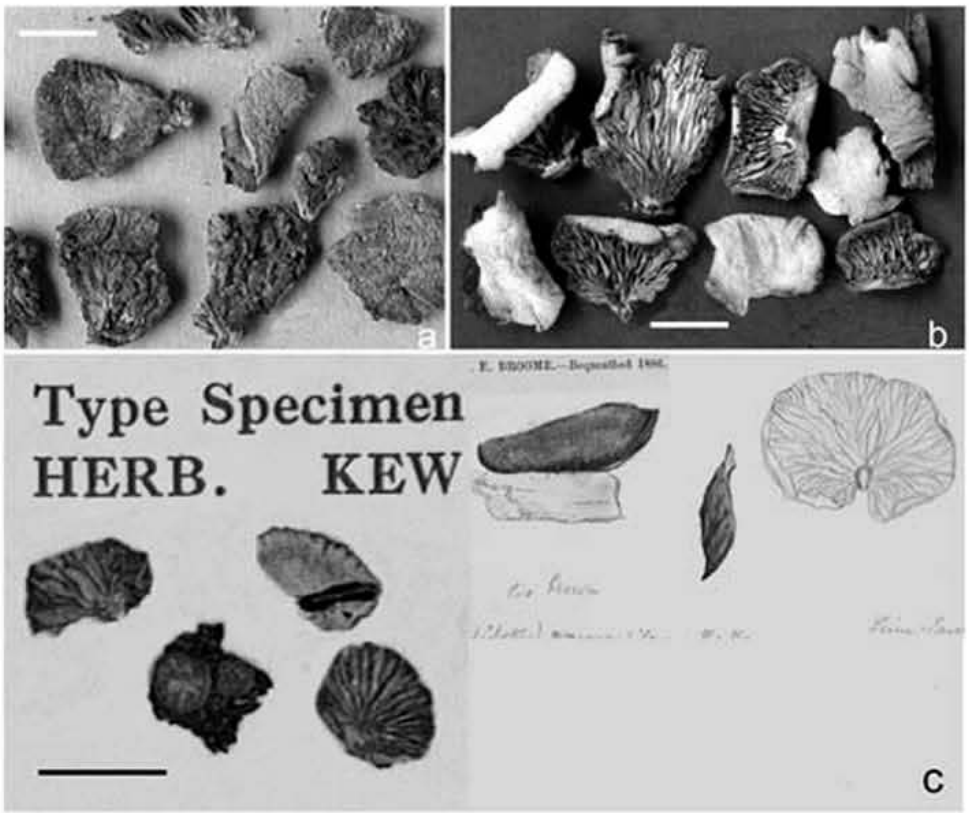


Plate 2. a-b: *Crepidotus applanatus* (a. Duss 31, holotype of *C. cuneiformis*. b. Smith 66854, holotype of *C. cystidiosus*). c. *Crepidotus calolepis* (Thwaites 954a, lectotype of *C. grumosopilosus*).
Scale bar = 10 mm, except a = 5 mm.

specimen from Mexico that Singer (1973) referred to *C. cuneiformis* (Singer M 8038, F) differs from the holotype and represents instead *C. palmarum* (Bandala & Montoya 2004). Since the Duss 31 specimen macro- and microscopically falls within the range of variation reported for *C. applanatus*, we propose the following synonymy:

***Crepidotus applanatus*** (Pers.) P. Kumm., Führ. Pilzk. p. 74, 1871.
Bas.: *Agaricus applanatus* Pers., Obs. Mycol. 1: 8, 1796.
Syn.: *Crepidotus cuneiformis* Pat., Bull. Soc. Mycol. France 18: 173, 1902.

***Crepidotus cystidiosus*** Hesler & A.H. Sm.,
N. Am. Sp. *Crepidotus*, p. 43, 1965. PLATES 2B, 3B-E & 7E-F

HOLOTYPE: USA. MICHIGAN: Colonial Point Hardwoods, 22.VII.1963, *A.H. Smith 66854* (MICH).

OTHER MATERIAL STUDIED— MEXICO. OAXACA: San Agustin, 10.VII.1969, *R. Singer M 8413* (F). USA. MICHIGAN: Cheboygan Co., Maple River, Univ. Michigan Biol. Stat., 24.VI.1956, *H.D. Thiers 2668* (MICH) (both as *C. cystidiosus*).

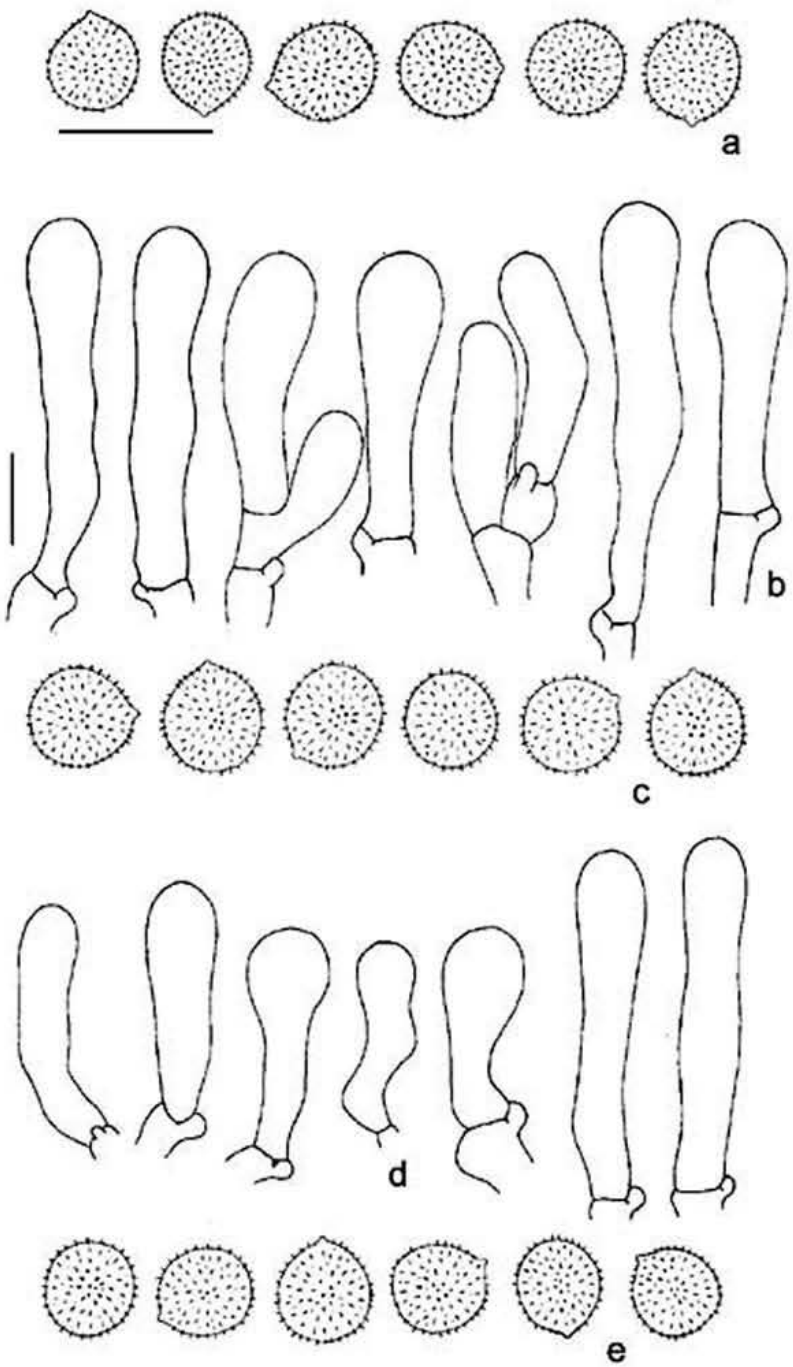


Plate 3. *Crepidotus applanatus*. a. Basidiospores (Duss 31, holotype of *C. cuneiformis*). b. Cheilocystidia. c. Basidiospores (Smith 66854, holotype of *C. cystidiosus*). d. Cheilocystidia e. Basidiospores (Singer M 8413). Scale bar = 10 µm.

Macroscopic description provided by Hesler & Smith (1965):

> "... Pileus 1.5–6 cm broad, sessile, white or pallid, hygrophanous, becoming alutaceous to buff in age, spathulate, dimidiate, or reniform, glabrous or slightly tomentose, margin striate when moist. Context thin, white, soft; odor mild, taste fungoid or mild, soon becoming somewhat bitter-astringent. Lamellae close, medium broad, white, becoming dull clay color, edges fimbriate..."

Microscopic features recovered from the holotype:

Basidiospores 5–6.5 (–7) × 5–6.5 (–7) µm, $\overline{X}$ = 6.0 × 5.8 µm, $Q$ = 1.02, globose to subglobose, spinulose to spinulose-verruculose seen under light microscope, baculate when observed under SEM, i.e. with more or less homogeneous, conical or rod-like, apically truncate protuberances, also small verrucae can be present among the bacules; thick-walled (≤ 0.5 µm thick), yellowish to yellowish-brown. Basidia (20–) 22–28 (–30) × 6–8 (–10) µm, clavate, tetrasporic, at times mono- or bisporic, hyaline, clamped. Pleurocystidia absent; sterile bodies resembling amorphous basidia or anomalous basidioles occasionally present, inconspicuous. Cheilocystidia 23–40 × 5–7 µm, numerous, clavate, narrowly clavate or subclavate, often more or less narrowly utriform, somewhat flexuous, apex 6–9 µm broad, rounded, at times subcapitate, hyaline, thin-walled, clamped. Pileipellis a cutis composed of more or less compactely arranged, hyaline, thin-walled, cylindric or somewhat ventricose hyphae 5–15 µm diam., occasionally some segments pale yellowish; terminal elements scarce, undifferentiated or at times more or less narrowly lageniform. Pileus trama with hyaline, cylindric to subventricose, thin-walled hyphae, 4–15 (–20) µm diam., some oleiferous-like hyphae (yellowish) present. Hymenophoral trama subregular, with cylindric to subventricose, thin-walled, hyaline hyphae, 4–15 µm diam., some oleiferous-like hyphae (yellowish) present. Clamp connections present in all tissues.

Comments— Except for the so-called pleurocystidia described by Hesler & Smith (1965), the type specimen of *Crepidotus cystidiosus* is identical to *C. applanatus*. Macro- and microscopically, A.H. Smith 66854 shows all the characters of *C. applanatus*, i.e. size, shape and color of pileus, globose, spinulose basidiospores that bear a baculate ornamentation pattern when observed under SEM, clamped hyphae, and a pileipellis cutis lacking brown incrusted pigments (cf. descriptions in Josserand 1937, Senn-Irlet 1995, Gonou-Zagou & Delivorias 2005). Analysis of tangential sections reveals that the hymenial elements on the lamellar face are more or less homogeneous with regard to size and shape (clavate, narrowly clavate or subcylindric). Occasional sterile bodies, apparently the so-called pleurocystidia, were detected (if somewhat ambiguously recognized) among the hymenial elements. Such structures, which possibly represent abnormal basidia or basidioles, can hardly be interpreted as true cystidia and do not support recognition of A.H. Smith 66854 as an

independent taxon. Other materials from Oaxaca and Michigan identified as *C. cystidiosus* by Singer (1973) and Hesler & Smith (1965) also do not show clear evidence of sterile bodies that can be reliably interpreted as pleurocystidia. The unpredictable occurrence of such structures, similarly observed by other authors in different samples of *Crepidotus*, reduces their value for taxonomic purposes (Bandala & Montoya 2000a, Gonou-Zagou & Delivorias 2005, Senn-Irlet 1995, Senn-Irlet & De Meijer 1998, Singer 1973). The macro- and microscopic features exhibited by the holotype support placing this taxon in synonymy with *C. applanatus* as follows:

***Crepidotus applanatus*** (Pers.) P. Kumm., Führ. Pilzk. p. 74, 1871.
Bas.: *Agaricus applanatus* Pers., Obs. Mycol. 1: 8, 1796.
Syn.: *Crepidotus cystidiosus* Hesler & A.H. Sm., N. Am. Sp. *Crepidotus*, p. 43, 1965.

***Crepidotus grumosopilosus*** (Berk. & Broome) Sacc.,
Syll. Fung. 5: 884, 1887. PLATES 2C, 4A-C & 7G-H
Bas.: *Agaricus grumosopilosus* Berk. & Broome, J. Linn. Soc. Bot. 11: 546, 1871.

LECTOTYPE: SRI LANKA (CEYLON). Peradeniya, January 1896, *Thwaites 954a* (K) (selected by Pilát 1950).

Macroscopic description by Berkeley & Broome (1871) for the specimen no. 954a, cum icone, on dead wood. Jan. 1869:

"... PILEUS suborbicular or reniform, rarely flabelliform, at first resupinate, then reflexed, white, striate, at first marked with radiating matted hairs which gradually acquire a more or less ferruginous tint and form scattered spots; STEM very short or quite obsolete; GILLS 2 lines or more broad, ventricose, pale ferruginous... "

Microscopic features recovered from the lectotype:

BASIDIOSPORES (7–) 8–10 × (5–) 5.5–7 µm, $\overline{X}$ = 9.0 × 6.4 µm, $Q$ = 1.40, ellipsoid, some more or less amygdaliform in side view, smooth (even under SEM), thick-walled (≤ 0.5 µm thick), yellowish-brown. BASIDIA 17–25 (–30) × (8–) 9–10 µm, clavate to subcylindric, tetrasporic, hyaline, thin-walled. PLEUROCYSTIDIA absent. CHEILOCYSTIDIA 23–60 × 4–9 (–11) µm, numerous, narrowly utriform to narrowly sublageniform, occasionally subcylindric to narrowly clavate or subclavate, apex rounded 3–9 µm wide, occasionally subfusiform, somewhat flexuous, embedded in gelatinous material, hyaline, clampless. PILEIPELLIS a compactly arranged, interrupted cutis, composed of cylindric to subventricose, yellowish to yellowish-brown hyphae 5–15 (–20) µm wide, thin- or thick-walled (< 0.5 µm thick), with incrusting pigment, at times the incrustations producing discontinuous lines, occasionally some hyaline hyphae are intermixed; terminal hyphae undifferentiated or somewhat attenuated, prostrated or semierect. PILEUS TRAMA consisting of hyaline, cylindric to subventricose or short-bifurcate, thin-walled hyphae 3–20 µm wide, some recalling puzzle-like elements, forming a compactly arranged, gelatinized tissue, without any distinctively refringent layer in tangential

section. HYMENOPHORAL TRAMA with a mediostratum, subregular, composed of cylindric to subventricose, compactly arranged, hyaline, non-gelatinized hyphae, 3–20 µm wide; laterostrata poorly differentiated, gelatinized, composed of hyaline, thin-walled, filamentous hyphae 3–7 µm wide. CLAMP CONNECTIONS absent.

COMMENTS— In his revision of species from Ceylon (Sri Lanka) described by Berkeley & Broome, Petch (1924) pointed out that the type collection of *Agaricus grumosopilosus*, Thwaites 954a cum icone, consisted of a mixture of specimens representing different species. Years later, Pilát (1950) recognized *C. grumosopilosus* as a smooth-spored species, which he lectotypified by selecting three specimens from Thwaites 954a. Pilát identified a fourth specimen bearing ornamented spores within this same collection as *C. truncatus*. Pegler (1986), who later examined Thwaites 954a and also distinguished two separate entities, referred the smooth-spored taxon (separated as Thwaites 954a p.p.) to *C. melleus* (Berk. & Broome) Petch and the taxon with ornamented spores (Thwaites 954a p.p. cum ic.) to *C. grumosopilosus*. There are, however, now in K two collections numbered 954a, **both** labeled as type specimens of *C. grumosopilosus*. [NOTE: another species also recorded by Berkeley & Broome (1871) as 954 (and 954b) is *C. epicrocinus* (Berk & Broome) Sacc., not discussed here]. One of the K 954a collections is coded "K(M): 49551" and the other is coded "K(M): 42400". Our reexamination of both collections reveals that they are indistinguishable from previously described species, suggesting that *Agaricus grumosopilosus* should be reduced to a synonym. This conclusion, which partly agrees with that of Pilát (1950) but opposes that of Pegler (1986), led us to reevaluate the nomenclatural and taxonomic status of the material kept at K.

Collection 954a [49551], unlike collection 954a [42400], is accompanied by a color drawing of some of its basidiomes (Plate 2c). The herbarium sheet, as noted by both Pilát (1950) and Pegler (1986), accompanies drawings by Berkeley showing smooth, ellipsoid basidiospores. Collection 954a [49551] is thus interpreted as representing the specimens included in the protologue and studied by the aforementioned authors. As collection 954a [49551] contains mixed specimens representing different species, then the macroscopic data in the diagnosis (see above) presumably embraced features of all specimens in this collection. In fact, our study revealed that the pileipellis of all basidiomes consists of pigmented, often incrusted, thick-walled hyphae, which macroscopically constitute brown fibrils or squamules actually observed on the pileus surface (when analyzed under magnification) and mentioned in the original description. Five of seven basidiomes in this collection (Plate 2c) possess smooth spores and two have ornamented spores. As observed by Pilát (1950), these two latter basidiomes represent a taxon with a more or less flabelliform, plane-convex pileus, a non-striate margin and laterally attached to

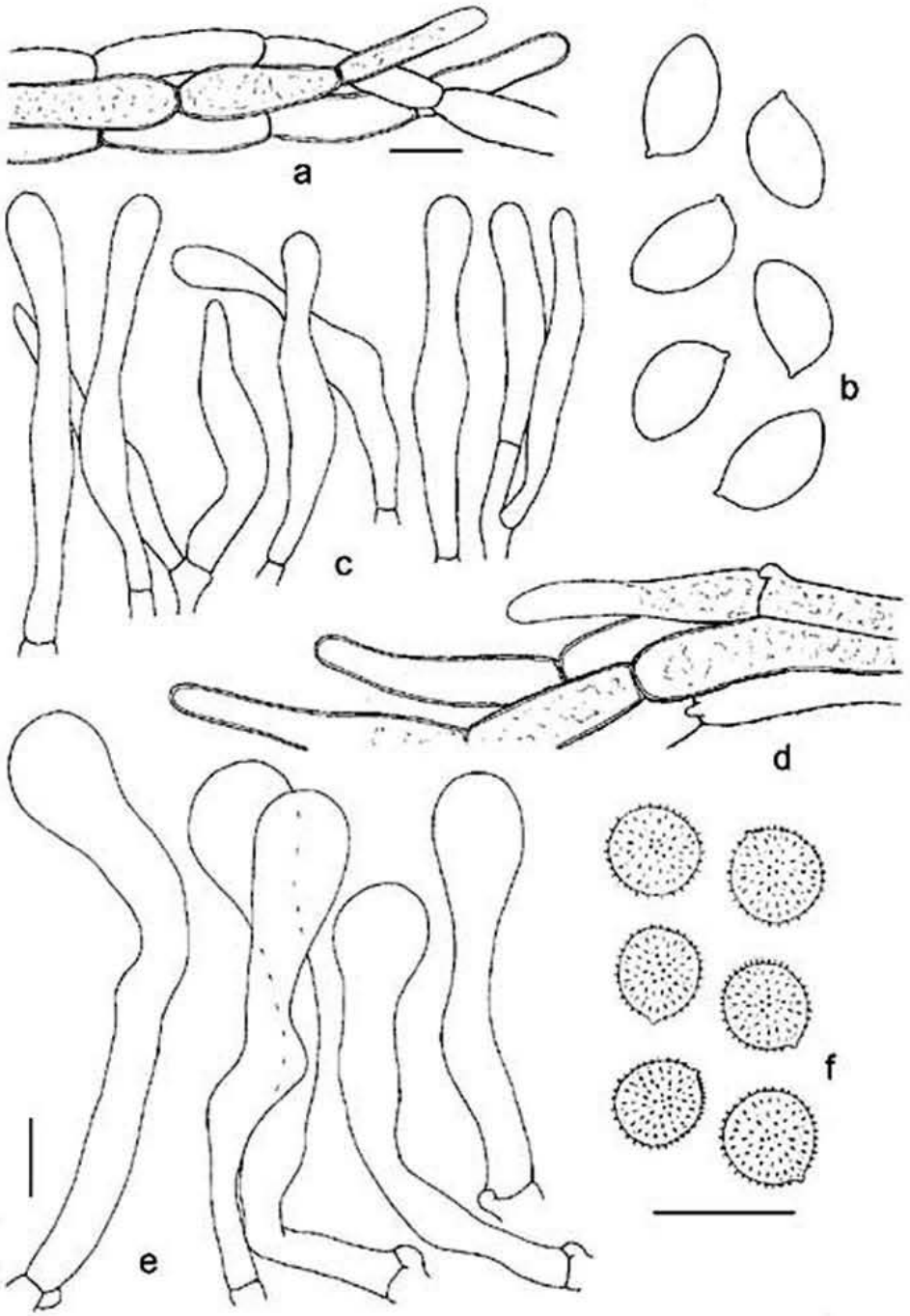


Plate 4. a-c: *Crepidotus calolepis*. a. Pileipellis. b. Basidiospores.
c. Cheilocystidia (Thwaites 954a, lectotype of *C. grumosopilosus*).
d-f: *Crepidotus crocophyllus*. d. Pileipellis. e. Cheilocystidia. f. Basidiospores (Thwaites 954a.1).
Scale bar = 10 µm, except a & d = 15 µm.

the substratum. However, the remaining five smooth-spored basidiomes are distinct even in the dried state due to the subcircular to rounded flabelliform (somewhat reniform when immature, as seen from the hymenophore), convex, striate pileus, with some more or less dorsally attached to substratum reminiscent of an effuse-reflexed pileus. We found among these five basidiomes some that appear with those depicted in Berkeley's color drawing accompanying the collection (Plate 2c). From this, we concluded that these five basidiomes agree better with the protologue (ICBN, Art. 9) than the material selected by Pegler (1986). Pilát's earlier lectotypification (1950) is, therefore, supported.

The microscopic information provided above is based on the aforementioned five basidiomes, which we recognize as representing the lectotype. This material was separated as Thwaites 954a (i.e. Thwaites 954a cum icone) inside the collection K (M): 49551. The set of features exhibited by these specimens, such as smooth spores, gelatinized tissues, clampless hyphae and pileipellis with brown pigmented hyphae (Figs. 4a-c & 7g-h) fall, however, within the range of variation recorded for *C. calolepis*. We interpret, therefore, the specimens as representing this species and for this reason consider *C. grumosopilosus* as a later synonym:

***Crepidotus calolepis*** (Fr.) P. Karst., Bidr. Känn. Finl. Nat. Folk 32: 414, 1879.
Bas.: *Agaricus calolepis* Fr., Öfvers. K. Vetensk.-Akad. Förh. 30: 5, 1873.
Syn.: *Agaricus grumosopilosus* Berk. & Broome, J. Linn. Soc. Bot. 11: 546, 1871.
≡ *Crepidotus grumosopilosus* (Berk. & Broome) Sacc., Syll. Fung. 5: 884, 1887.

The two specimens with ornamented spores, i.e. *C. grumosopilosus* sensu Pegler or *C. truncatus* as identified by Pilát (cf. the type study of this latter below), are indistinguishable from *C. crocophyllus*. These two materials exhibit the following remarkable set of characters (Figs. 4d-f & 7i): basidiospores (5.5–)6–7.5 × (5.5–)6–7(–7.5) µm, $\overline{X}$ = 6.7 × 6.5 µm, $Q$ = 1.03, globose, spinulose (baculate under SEM), clamped hyphae, variably clavate or subutriform cheilocystidia (45–) 50–90 (–95) × 5–9 (–10) × (apex) 10–18 (–20) µm, and pileipellis with brown pigmented, often incrusted, thick-walled hyphae, (6–) 9–15 µm wide. We separated these two specimens as Thwaites 954a.1 inside collection K(M): 49551. On the other hand, collection Thwaites 954a K(M): 42400 consists of a single basidiome that has the same features cited above for *C. calolepis*. The herbarium sheet is also accompanied by Berkeley drawings showing smooth, ellipsoid basidiospores; macroscopically, the presence of brown fibrils on the pileus surface (seen under magnification) is remarkable. Relevant microscopic features of this specimen are:

Basidiospores (7.5–)8–10(–10.5) × 5.5–7(–7.5) µm, $\overline{X}$ = 8.9 × 6.4 µm, $Q$ = 1.40, ellipsoid, smooth. Cheilocystidia hyaline, clampless, more or less narrowly sublageniform, gelatinized (the lamellar edge is collapsed and could barely be revived in KOH). Pileipellis a cutis composed of cylindric

to subventricose, yellowish-brown hyphae, 5–10 µm wide, with or without incrusting pigment. PILEUS TRAMA and HYMENOPHORAL TRAMA with gelatinized hyphae. CLAMP CONNECTIONS absent. It is not clear whether this latter material together with the lectotype were originally a single sample gathered by Thwaites.

***Crepidotus maximus*** Hesler & A.H. Sm.,
N. Am. Sp. *Crepidotus*, p. 100, 1965. **PLATES 5 & 71-K**

HOLOTYPE: USA. TENNESSEE: Ball Camp Pike, 7.XI.1936, *R.L. Hesler and S.A. Cain 9718* (TENN).

Macroscopic description provided by Hesler & Smith (1965):

"...PILEUS 4–13 cm broad, sessile, somewhat imbricate, dimidiate, cuneate or broadly flabelliform, convex, then more or less plane, or the margin wavy and the midportion more or less depressed, white or whitish, becoming tinted "light ochraceous-buff", coarsely rivulose-reticulated, innately fibrillose, distinctly viscid or almost glutinous when wet, soon dry, not hygrophanous, margin incurved and even. CONTEXT thick, tough, rather turgid, pallid. LAMELLAE radiating, white at first, becoming "snuff brown", "argus brown" or "Brussel's brown", with a purple-brown tint where bruised, close, linear, relatively narrow (up to 5 mm), narrowed at the ends, edges white-fimbriate, at times gelatinous..."

Microscopic features recovered from the holotype:

BASIDIOSPORES (6–)6.5–8 × 3.5–4.5(–5) µm, $\overline{X}$ = 7.2 × 4.1 µm, $Q$ = 1.76, ellipsoid to more or less narrowly amygdaliform, somewhat depressed adaxially and then more or less narrowly reniform, weakly attenuated towards the apex but rounded, smooth (even under SEM), thick-walled (<0.5 µm wide), yellowish to yellowish-brown. BASIDIA 20–28 × 4–7 µm, tetrasporic, occasionally bi- or monosporic, clavate, thin-walled, hyaline, clamped. PLEUROCYSTIDIA absent. CHEILOCYSTIDIA (62–)73–121(–126) × (3–)4–5 µm, numerous, subcylindric, more or less elongate-subclavate or slightly broadened downwards and then elongate and more or less narrowly lageniform, apex rounded, (4–)5–8(–9) µm wide, neck occasionally bifurcate, often somewhat sinuous towards the base, prominently projected above the hymenium level, hyaline, thin- or slightly thick-walled (<0.5 µm thick), clamped, slightly gelatinized producing a more or less dense, refringent layer on lamellae edge. PILEIPELLIS a broad (± 300–450 µm thick) not gelatinized layer of somewhat loosely interwoven or entangled, more or less filamentous hyphae, 3–5 µm wide, forming a cutis or a transition between a loose cutis and a loose trichoderm, with a variable number of undifferentiated, repent or often erect terminal elements; most hyphae hyaline (some of their segments occasionally pale yellowish), moderately sinuous, often curved, at times bifurcate or with short, lateral outgrowths, clamped, thin- or slightly thick-walled (≤ 0.5 µm thick), smooth or at times some segments weakly punctuate (colorless). PILEUS TRAMA (in tangential section) differentiated in

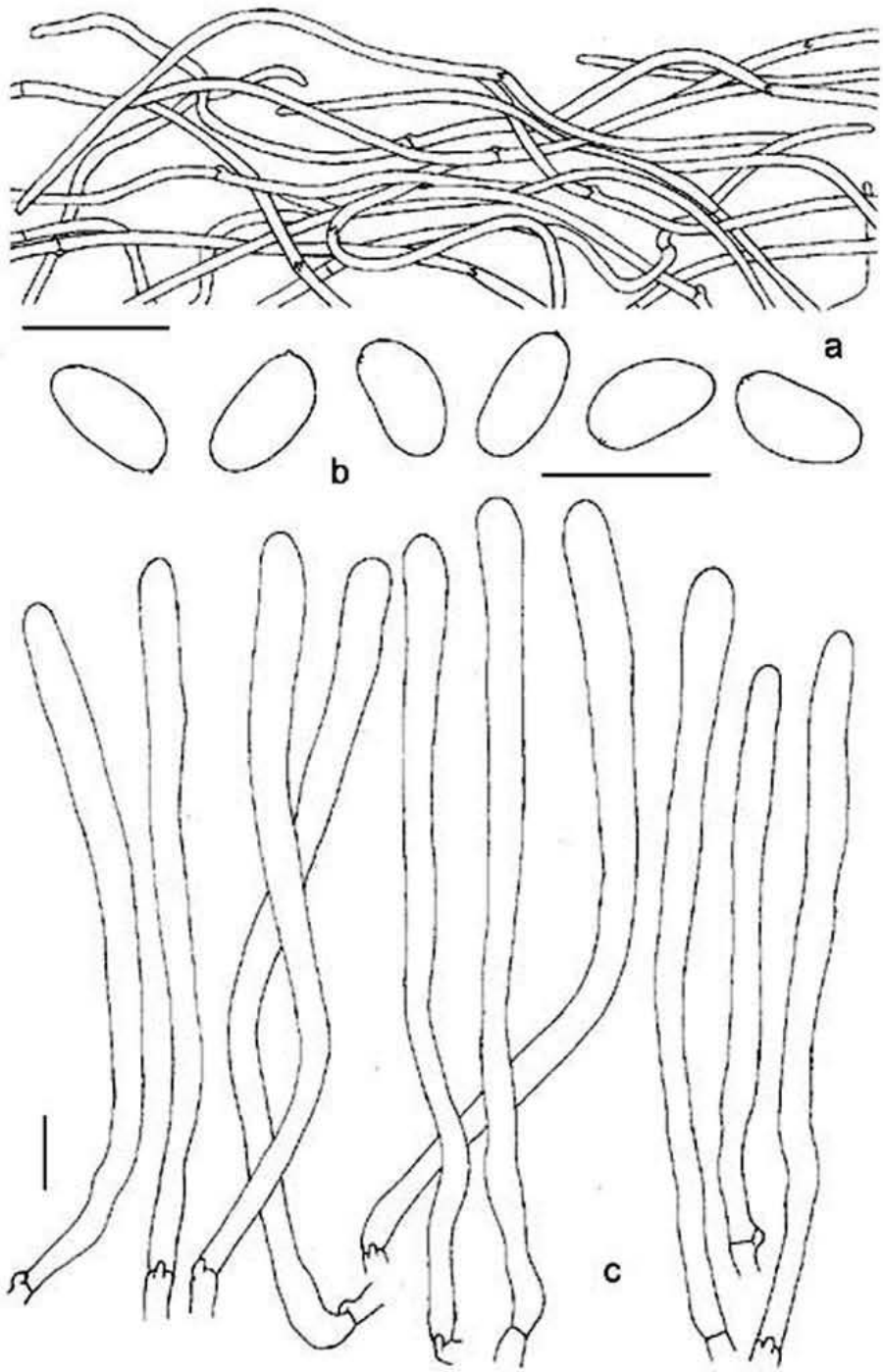


Plate 5. *Crepidotus albescens*. a. Pileipellis. b. Basidiospores.
c. Cheilocystidia (Hesler & Cain 9718, holotype of *C. maximus*).
Scale bar = 10 µm, except a = 20 µm.

two layers, one beneath pileipellis, gelatinized, refringent, variable in thickness (± 200–400 µm thick), composed of more or less filamentous hyphae (1–) 2–4 µm wide, hyaline or at times with some segments with a dense, yellowish content, thin-walled, smooth or occasionally obscurely punctuate (colorless), flexuous, more or less radially oriented and loosely interwoven; below that layer is a distinctive, moderately compact, gelatinized but not refringent stratum comprising most of the pileus context and composed of colorless, thin-walled, subcylindric to subventricose, simple or bifurcate hyphae, 5–15 (–20) µm wide. HYMENOPHORAL TRAMA with a mediostratum, subregular, composed of cylindric to subventricose, compactly arranged, hyaline, not gelatinized hyphae, 4–15 µm wide; laterostrata refringent, gelatinized, composed of filamentous, colorless to pale yellowish, thin-walled hyphae, 2–4 (–5) µm wide, loosely and more or less divergently arranged. CLAMP CONNECTIONS present in all tissues.

COMMENTS— Microscopically Hesler & Cain 9718 exhibits all the distinctive characters of *C. albescens* that include the moderately reniform, smooth basidiospores, elongate cheilocystidia, clamped hyphae, entangled cutis and gelatinized tissues (Hesler & Smith (1965) under *C. phaseoliformis* Hesler & A.H. Sm., Redhead 1984, pers. obs.). The pileus size and the color change of gills when bruised (i.e. with a purple-brown tint) were the main features Hesler & Smith (1965) used to separate *C. maximus* from *C. betulae* Murrill, the latter equated with *C. albescens* after type study (we shall discuss *C. albescens* and related species in a separate paper). Certainly the type specimen supporting *C. maximus* possesses a somewhat large pileus, but it perhaps corresponds to the upper limit of pileus size in *C. albescens*. Microscopically, the fine fibrillose pileus covering forms a dense hyphal layer that is slightly thicker than that observed in other collections. The pileus size, the apparent color change of lamellae, and even the thickness of the pileipellis layer showed by Hesler & Cain 9718 seem to fall within the range of variability to be expected for a single taxon rather than indicate a taxonomic separation. The color change of the lamellae needs more observation before it can be considered discriminative. Since macro- and microscopically the holotype falls within the range of variation of *C. albescens*, we consider that *C. maximus* is not an independent taxon:

***Crepidotus albescens*** (Murrill) Redhead, Sydowia 37: 255, 1984.
Bas.: *Geopetalum albescens* Murrill, N. Am. Fl. 9: 299, 1916.
Syn.: *Crepidotus maximus* Hesler & A.H. Sm., N. Am. Sp. *Crepidotus*, p. 100, 1965.

***Crepidotus truncatus*** Petch, Ann. Roy. Bot. Gard. Perad. 9: 226, 1924. **PLATES 6-7I.**

HOLOTYPE: SRI LANKA (CEYLON). CENTRAL PROVINCE: Kandy District, Peradeniya, 19.XII.1914, *T. Petch 4384* (K).

OTHER MATERIAL STUDIED— MEXICO. CHIAPAS: along road from Finca Suspiro to El Pozo, 4.VIII.1969, *R. Singer M 8965* (F, as *C. truncatus*).

Macroscopic description provided by Petch (1924):

> "... PILEUS elliptical, lobed behind, broadly convex, 4 × 2.5 cm., pallid, covered with a thin layer of matted fibrils, strongly tomentose and white at the point of attachment, striate from the margin over half the pileus when moist; GILLS not crowded, very broad (up to 1 cm.), ventricose, pallid then brownish, truncate behind, terminating at an excentric, white, tomentose area under the point of attachment...".

Microscopic features recovered from the holotype:

BASIDIOSPORES 5.5–6.5 × 5.5–6.5 µm, $\overline{X}$ = 6.2 × 6.1 µm, $Q$ = 1.02, globose, spinulose to spinulose-verruculose (under light microscope), baculate under SEM, i.e. with conical or rod-like, apically truncate protuberances, small verrucae can be present among the bacules; thick-walled (≤ 0.5 µm thick), yellowish-brown. BASIDIA clavate, tetrasporic, hyaline, clamped. PLEUROCYSTIDIA absent. CHEILOCYSTIDIA hardly revived with KOH, the lamellae edge is somewhat collapsed but some clavate forms were observed (or part of their upper extremes with rounded apex), hyaline, thin-walled, clamped. PILEIPELLIS a cutis composed of hyaline, thin-walled hyphae 2–5(–7) µm wide, occasionally some segments pale yellowish; terminal elements undifferentiated or somewhat narrowly lageniform, at times somewhat erect then recalling a more or less trichodermoid arrangement. PILEUS TRAMA with hyaline to yellowish, cylindric to subventricose, thin-walled hyphae. HYMENOPHORAL TRAMA subregular, with cylindric to subventricose, thin-walled, hyaline hyphae. *Clamp connections* present in all tissues.

COMMENTS — The combination of features exhibited by the holotype, such as the globose, spinulose (baculate under SEM) basidiospores, clavate cheilocystidia, pileipellis cutis with colorless hyphae and clamp connections, indicates that *C. truncatus* is the same as *C. applanatus*. The macroscopic variation described by Petch (1924) also falls within the range of variation reported for *C. applanatus* (Gonou-Zagou & Delivorias 2005, Hesler & Smith 1965, Josserand 1937, Nordstein 1990, Pilát 1948, Senn-Irlet 1995). Singer (1955) found *C. truncatus* similar to either *C. applanatus* or *C. nephrodes* sensu Singer but later (Singer 1973: 369) treated *C. truncatus* as an autonomous species, segregating it from *C. applanatus* based on pileus color: *C. applanatus* with a whitish pileus and a marginal zone tinged argillaceous-gray when fresh, not brownish unless in age when dried and *C. truncatus* with a brown or pale brown pileus (although this detail does not agree with the diagnosis, see above). We have reexamined a collection from Mexico that Singer (1973) identified as *C. truncatus*; Singer M 8965 is likewise morphologically indistinguishable from *C. applanatus*, representing a small form with:

BASIDIOSPORES 5.5–7 × (5–)5.5–6.5 µm, $\overline{X}$ = 6.1 × 5.9 µm, $Q$ = 1.04, globose, baculate (under SEM), and CHEILOCYSTIDIA (15–)20–55 × (3–)4–9

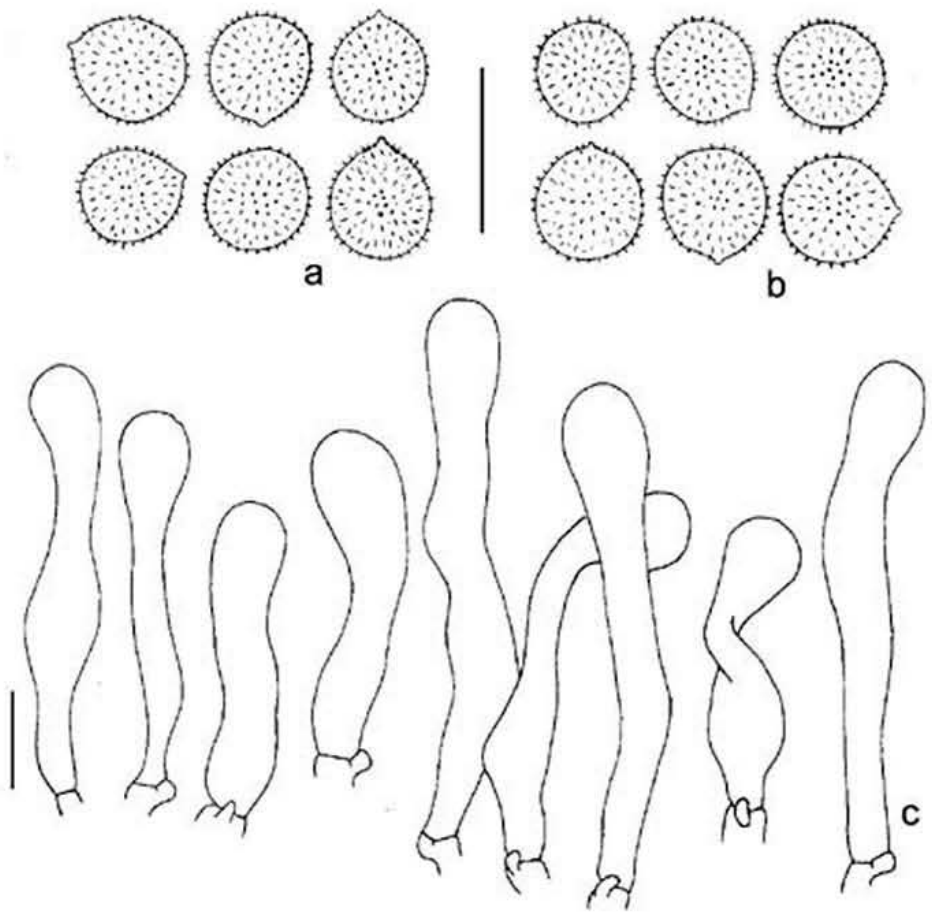


Plate 6. *Crepidotus applanatus*. a. Basidiospores (Petch 4384, holotype of *C. truncatus*). b. Basidiospores. c. Cheilocystidia (Singer 8965). Scale bar = 10 µm.

µm, apex 3–9(–12) µm wide, numerous, clavate, narrowly clavate or more or less narrowly utriform, hyaline thin-walled, clamped (Plate 6b-c).

From the above, we concluded that the Sri Lankan type collection of *C. truncatus* represents *C. applanatus* and is a later synonym:

***Crepidotus applanatus*** (Pers.) P. Kumm., Führ. Pilzk. p. 74, 1871.
Bas.: *Agaricus applanatus* Pers., Obs. Mycol. 1: 8, 1796.
Syn.: *Crepidotus truncatus* Petch, Ann. Roy. Bot. Gard. Perad. 9: 226, 1924.

As noted in the earlier discussion on *C. grumosopilosus*, Pegler (1986) considered *C. truncatus* and *C. grumosopilosus* to be conspecific. We shall discuss the taxonomy of *Agaricus nephrodes* Berk. & M.A. Curtis, which Pegler (1977) considered synonymous to *Crepidotus truncatus*, in a separate paper devoted to the species related to *C. crocophyllus* (cf. also Ripková et al. 2005).

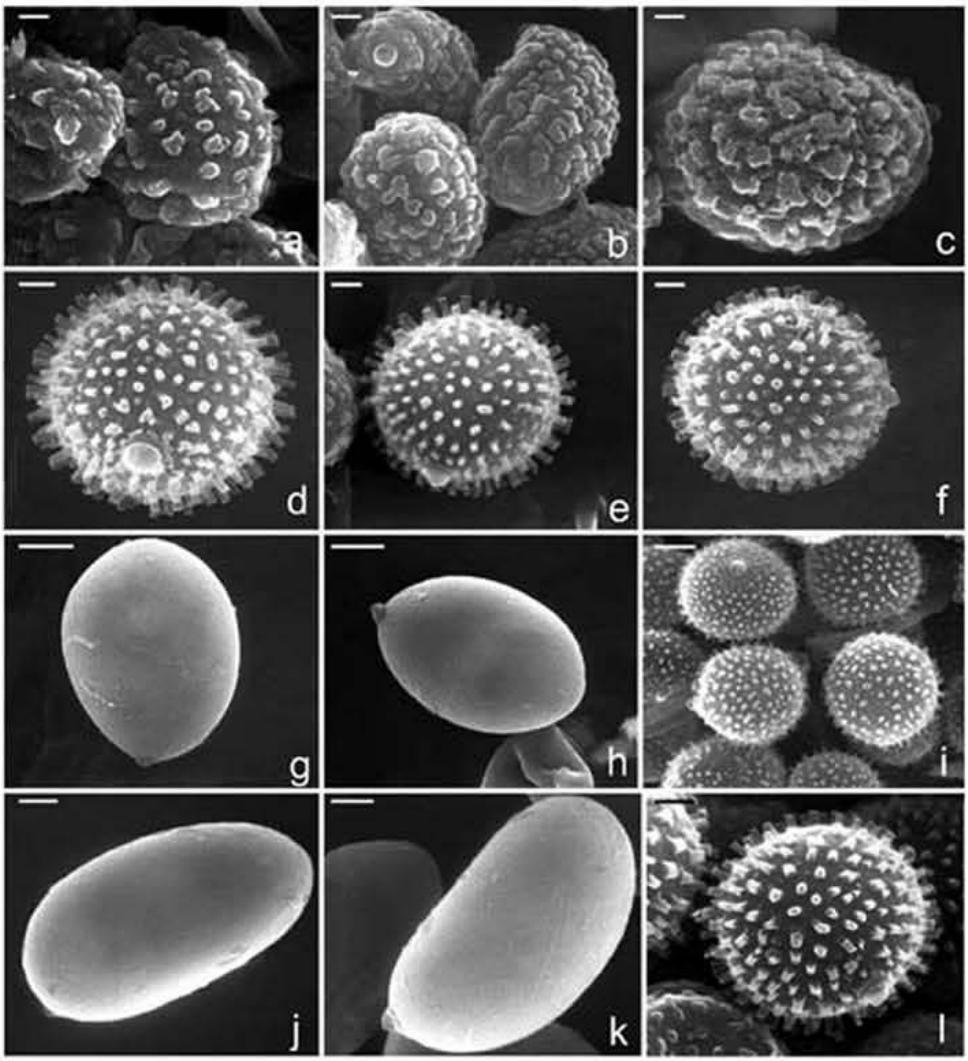


Plate 7. a-c: *Crepidotus croceitinctus* (a. Spegazzini 17022, holotype of *C. brunswickianus*. b. Singer 3365. c. Dennis 1760). d-f: *Crepidotus applanatus* (d. Duss 31, holotype of *C. cuneiformis*. e. Smith 66854, holotype of *C. cystidiosus*. f. Singer M 8413). g-h: *Crepidotus calolepis* (Thwaites 954a, lectotype of *C. grumosopilosus*). i: *Crepidotus crocophyllus* (Thwaites 954a.1). j-k: *Crepidotus albescens* (Hesler & Cain 9718, holotype of *C. maximus*). l. *Crepidotus applanatus* (Petch 4384, holotype of *C. truncatus*). Scale bar = 1 µm, except g-i = 2 µm.

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# *Sporopodium isidiatum* (*Pilocarpaceae*), new from Papua New Guinea and Sri Lanka, with a key to the world's *Sporopodium* lichen species

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**Abstract** — The new lichen species *Sporopodium isidiatum* is described from Papua New Guinea and Sri Lanka. A key to all known species of *Sporopodium* is provided, as well as a synoptic table of their secondary chemical compounds.

**Key Words** — *Ectolechiaceae, Micareaceae*

## Introduction

The lichen family *Pilocarpaceae* was reinstated by Vězda (1986) to accommodate mainly foliicolous species in the genera *Byssoloma*, *Fellhanera* and *Byssolecania*. However, Lücking & al. (1994) and Lücking (1999) suggested that the family should be emended to include *Ectolechiaceae*, i.e. taxa with characteristic campylidioid anamorphs centered in the genus *Sporopodium*. A recent molecular phylogenetic analysis, based on Bayesian tree sampling and maximum likelihood analysis of mtSSU sequences (Andersen & Ekman 2005), not only confirmed this view but showed that also *Micareaceae* have to be submerged within *Pilocarpaceae*.

Whereas the phylogenetic relationships of genera within this large and diverse clade remain unclear, inventories of tropical regions continuously yield new and undescribed species, especially in the large genera *Byssoloma* and *Fellhanera*, but also in the smaller genera, such as *Sporopodium*. The latter is distinguished by the following combination of characters (Lücking 1999, Santesson 1952, Sérusiaux 1986, Vězda 1986): finely farinose thallus, sometimes with verrucae; branched and anastomosed paraphyses; usually a single muriform spore per

ascus (8 in one species); campylidia hood-like, made of a conidia-producing cavity connected to a tiny platform which seems to be covered under a hood-like lobe; and conidia non-septate, ellipsoid to guttuliform (1-septate, with the distal cell rounded and the proximal one ellipsoid-bacillar in a single species). Photobiont cells are present in the epithecium and the conidiogenous layer in some species.

In his world-wide monograph on foliicolous lichens, Santesson (1952) recognized six taxa of *Sporopodium* (four species and two varieties); several further species have eventually been recognized (Aptroot & Sipman 1993, Aptroot & al. 1997, Elix & al. 1995, Lücking 1999 & 2008, Lücking & Kalb 2002, Lücking & Lumbsch 2001, Lücking & Santesson 2002, Øvstedal & Elix 2007, Santesson & Lücking 1999). Most species are foliicolous but many can also occur on twigs, and two species are so far only known from bark.

A very conspicuous species, with verrucose-isidiate thalli and large campylidia with a black lobe, is here described as new to science; it is known from Papua New Guinea and Sri Lanka. A key to all described species in the genus is presented, as well as a synopsis of the diverse secondary chemical compounds they produce.

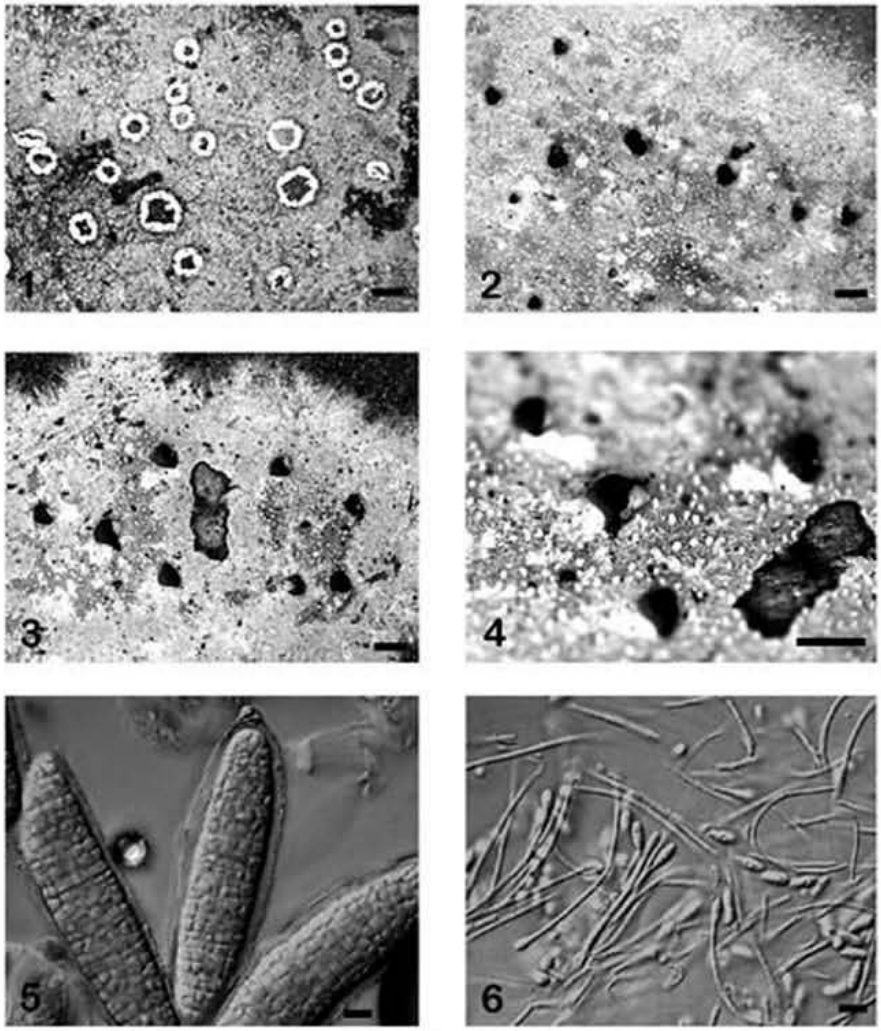
## The species

***Sporopodium isidiatum*** Sérus. & Lücking **sp. nov** Figs. 1-6

MycoBank MB 511310

*Ab aliis speciebus Sporopodium differt thallo isidiato et campylidiis atris.*

Thallus corticolous or foliicolous, usually circular, c. 1.0-1.5 cm in diam. (largest seen: c. 2.5 cm), hardly delimited at the margins where free-living radiate hyphae may occur and thus form a whitish prothallus, continuous but formed of scattered to aggregated tiny patches at the margins; surface finely farinose, not corticated and rather matt, pale greenish to pale grey, usually with a bluish hue; verrucae always present, most probably representing genuine isidia, cylindrical and erect, c. 0.1-0.2(-0.3) mm in height and 0.05-0.1 mm in diam., regularly distributed over the thallus surface, albeit more numerous towards the center of the thallus, never branched, nor aggregated, of the same color as the thallus, filled with large, irregular oxalate crystals. Photobiont a species of the *Chlorococcaceae*, most probably *Trebouxia*. Cyanobacteria always present on the thallus surface, forming loose to compact, vermicular to granular, bluish grey to almost turquoise blue cephalodia, extensively covering large parts of the thallus; cyanobacterium a species of *Scytonema* with rare false branching, almost all trichomes with heterocysts, densely embedded by hyphae. Campylidia present on thalli not producing apothecia, all pointing in the same direction and looking like the ventilating pipes of a steamer; made of a




Figs. 1-6. *Sporopodium isidiatum*. 1. Thallus with apothecia. 2-4. Thallus with campylidia. 5. Ascospores. 6. Conidia and conidiogenous cells. 1 & 5: holotype; 2-4 & 6: Papua New Guinea, Gahavisuka, E. Sérusiaux 13762-81. Scale bars: 1-4 = 1 mm; 5-6 = 10 µm.

robust „socle" containing the conidia-producing cavity vertically connected to a tiny, laterally laid down cupula which is almost completely covered by a hood-like lobe; socle 0.4-0.5(-0.6) mm in diam., campylidium (when mature) total height 0.7-1 mm, and (seen from above) 0.5-0.8 mm for the largest dimension (width of the cupula); outer surface pale orange to brownish at the socle level, turning into dark brown to almost pure black on the covering lobe and the cupula; inner parts of the tissues forming the covering lobe and the cupula, as well as the bottom of the conidiogenous layer, with numerous, tiny, orange brown crystals that do not dissolve in K; conidiogenous cells cylindrical, 20-25

× 1-1.5 µm; conidia abundantly produced, apically arising one at a time, ellipsoid to slightly clavate, usually lacriform (the distal end rounded and the proximal one pointed, usually with an apiculus), hyaline, non-septate, 8-10 × 3(-3.5) µm; conidia accumulating in a globose, pale orange mass at the „mouth" of the campylidium; no photobiont cells seen in the conidiogenous layer, nor in the conidial mass. APOTHECIA present on thalli not producing campylidia, biatorine, rounded or slightly irregular, strongly constricted at their base, 0.9-1.2 mm when fully mature, 0.3-0.4 mm in height; margin persistently strongly prominent, 0.1-0.15(-0.2) mm thick, whitish, or with a very pale yellowish-orange hue, smooth to downy, crenulate and somewhat swollen, with a few radial fissures; disc greyish to almost black, not or slightly pruinose, plane. Excipulum well-developed, typically paraplectenchymatous in its inner parts, containing numerous, tiny, orange brown crystals that do not dissolve in K, 90-100 µm thick under the hypothecium, up to 140 µm in lateral parts; hypothecium orange to brownish, not significantly changing in K; hymenium dark under the dissecting microscope for unknown reason, but hyaline when examined with the photonic microscope, c. 110-130 µm thick; paraphyses numerous, branched and anastomosed, c. 1 µm thick; asci clavate, of the *Sporopodium*-type, 100-125 × c. 25 µm; ascospores single in the asci, strongly muriform, narrowly ellipsoid, with a distinct halo (easily seen in water), 91-118 × 19-25 µm.

CHEMISTRY: 2,7-dichlorolichexanthone (major), zeorin (major), pannarin (minor), 7-chloro-6-O-methylnorlichexanthone (minor), 2,7-dichloro-6-O-methylnorlichexanthone (minor) and 2-chlorolichexanthone (determined by HPLC and HPTLC).

NOTES — *Sporopodium isidiatum* is readily recognized by its verrucose-isidiate thallus, and large and conspicuous campylidia and (when present) apothecia. Moreover the almost black color of the campylidia lobes forms a strong contrast with the white to slightly bluish thallus, and this feature is characteristic of the species.

As summarized in Tab. 1, the secondary chemical compounds produced in the genus are numerous and diverse. *Sporopodium isidiatum* is characterized by the production as major compounds of 2,7-dichloro-lichexanthone and zeorin, a combination not found in any other species.

DISTRIBUTION AND ECOLOGY — *Sporopodium isidiatum* is known from two localities in Papua New Guinea, in the montane forest zone (1300-2450 m), where it grows on living leaves in well-preserved stands. It must be quite rare as — although many rainforest localities have been studied in several parts of the country — *S. isidiatum* has been detected in only two. Interestingly, it was also found corticolous at a much lower elevation in Sri Lanka. The species is thus likely to be widespread in tropical S-E Asia.

TABLE 1. Secondary chemical compounds produced by *Sporopodium* species

| COMPOUNDS/SPECIES | ae | an | au | ci | fl | is | li | lm | lu | ma | ph | pi | xa |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| **XANTHONES** | | | | | | | | | | | | | |
| Arthothelin | xx | x | xxx | | x | | x | | | | | | x |
| Isoarthothelin | | x | xx | | x | | x | x | | xx | | | x |
| Asemone | | x | x | | x | | x | | | | | | x |
| 3-O-methylasemone | | | | | | | | x | | | | | |
| 2-Chlorolichexanthone | | | | | | x | | | | | | | |
| 4-Chlorolichexanthone | | | | x | | | | | | | | | |
| 4-Chloro-3-O-methylnorlichexanthone | | | | x | | | | | | | | | |
| 4-Chloro-6-O-methylnorlichexanthone | | | | x | | | | | | | | | |
| 5-Chloronorlichexanthone | | | | x | | | | | | | | | |
| 2,5-Dichlorolichexanthone | | | | | | | | x | | | | | |
| 2,7-Dichlorolichexanthone | | | | | x | xxx | | | | | | | x |
| 4,5-Dichlorolichexanthone | | | | x | | | | | | | | | |
| 5,7-Dichlorolichexanthone | | | | | x | | | x | | | | | x |
| 2,7-Dichloro-6-O-methylnorlichexanthone | | | | | | x | | | | | | | |
| 4,5-Dichloro-6-O-methylnorlichexanthone | | | | x | | | | | | | | | |
| 5,7-Dichloro-3-O-methylnorlichexanthone | | | | | x | | | x | xx | xxx | x | | x |
| 4,5-Dichloronorlichexanthone | xx | | | | | | | | | | | | |
| 5,7-Dichloronorlichexanthone | | | | | x | | | | | | | | x |
| 2,4,5-Trichlorolichexanthone | | | | | | | | x | | | | | |
| 2,5,7-Trichlorolichexanthone | | | | | x | | | | | | | | x |
| 2,5,7-Trichloro-3-O-methylnorlichexanthone | | | | | x | | | | xx | xxx | x | | x |
| 4,5,7-Trichlorolichexanthone | | | | | | | | x | | | | | |
| 7-Chloro-6-O-methylnorlichexanthone | | | | | | x | | | | | | | |
| Chodatin | | | | | | | | x | | | | | |
| Thiophanic acid | xxx | x | xxx | | x | | x | | | | | | x |
| Vinetorin | | | | x | | | | | | | | | |
| **DEPSIDES AND DEPSIDONES** | | | | | | | | | | | | | |
| Atranorin | | | | | | | | | | | | x | |
| Methylbarbate | | | | | | | | xxx | | | | | |
| Argopsin | | x | | | | | x | | | | | | |
| Pannarin | | | | | x | x | | | xx | | x | x | x |
| **TRITERPENOIDS** | | | | | | | | | | | | | |
| Zeorin | | x | xx | x | x | xxx | x | | | | x | x | x |
| **VULPINIC ACID DERIVATIVES AND DIBENZOFURANES** | | | | | | | | | | | | | |
| Vulpinic acid | | | | | | | | | xxx | | | | |
| Isousnic acid | xx | | | | | | | | | | | | |
| Usnic acid | xxx | | | | | | | | | | | | |

ae= *aeruginascens*, an = *antonianum*, au = *aurantiacum*, ci = *citrinum*, fl = *flavescens*, is = *isidiatum*, li = *leprieurii*, lm = *leprosum*, lu = *lucidum*, ma = *marginatum*, ph = *phyllocharis*, pi = *pilocarpoides*, xa = *xantholeucum*.

Specimens examined — Papua New Guinea: Madang prov., S side of Ramu river, Bundi village, along the road to Bundi Gap, 5°44.9'S 145°14.1'E, 1300-1500 m, forested slope, foliicolous, Nov. 1995, E. Sérusiaux 16609 (LG—holotypus, F—isotypus). Eastern Highlands prov., Gahavisuka Provincial Park, 11 km N of Goroka, 6°01'S 145°25'E, 2300-2450 m, little disturbed mossy mountain forest, Aug. 1992, E. Sérusiaux 13762-81 (LG, F). Sri Lanka: Southern prov., Galle distr., Kanneliya Forest, 1 km N of forestry bungalows, 6°15'N 80°22'E, 90 m, corticolous, March 1978, G. Thor 583 (S).

## Key to all known species of *Sporopodium*

1a Thallus or reproduction organs distinctly (citrine) yellow or orange ..... 2

1b Thallus or reproduction organs not distinctly yellow or orange [campylidia and apothecial margins sometimes pale yellow to pinkish orange] ..... 7

2a Thallus (at least parts of it, mainly near apothecia and campylidia), campylidia and apothecia yellow or orange ..... 3

2b Thallus bluish, apothecial margin grey to whitish, campylidia bright yellow ..... 4

3a Thallus, apothecia (mainly margins) and campylidiapale to bright orange; apothecial margin smooth ..... [pantropical] ***Sp. aurantiacum*** (Zahlbr.) Lücking

3b Thallus lemon green, apothecia (mainly margins) and campylidia bright lemon yellow; apothecial margin slightly crenulate..... .....[tropical Asia, New Guinea] ***Sp. lucidum*** Aptroot & Sipman

4a Thallus distinctly verrucose; apothecial margin irregular or slightly crenulate ... 5

4b Thallus smooth or with scattered, indistinct, low verrucae; apothecial margin smooth or almost so ..... 6

5a Verrucae irregularly papillose, with a thinly pruinose to pilose surface; thallus with usnic and isousnic acid, and dichloro- and trichlorolichexanthones ..... ..... [neotropical] ***Sp. aeruginascens*** Lücking & Lumbsch

5b Verrucae regular and smooth; thallus with zeorin, together with chloro- and dichlorolichexanthones ..... [pantropical] ***Sp. citrinum*** (Zahlbr.) Elix & al.

6a Thallus smooth or with scattered, indistinct, low verrucae; base of campylidia well-developed, usually broader and larger than the lobe, and thus the most conspicuous part of the campylidium ..... [eastern paleotropics] ***Sp. flavescens*** (R. Sant.) Vezda

6b Thallus smooth; base of campylidia reduced and thus the lobe being the most conspicuous part of the campylidium ... [amphipacific] ***Sp. subflavescens*** Lücking

7a Verrucae always distinct; thallus usually greyish to pale greenish grey *or* with a bluish tinge ..... 8

7b Verrucae absent, or scattered, indistinct or low; thallus usually bluish grey ..... 13

8a Thallus coarsely verrucose to isidiate; campylidia with a blackish brown to pure black lobe; thallus with a bluish tinge [tropical Asia and New Guinea] ***Sp. isidiatum***

8b Thallus finely verrucose, not isidiate; campylidia with a light to dark brown lobe; thallus usually without a bluish tinge ..... 9

9a Prothallus typically present, woolly, of loosely interwoven hyphae, very similar to that produced by *Lasioloma* species; apothecial margin with tiny hairs. . . . . [pantropical] ***Sp. antonianum*** Elix & al.

9b Prothallus not woolly; apothecial margin without hairs . . . . . 10

10a Apothecial margin smooth or almost so, hardly raised . . . . . 11

10b Apothecial margin irregular or thick, crenulate, and raised . . . . . 12

11a Apothecial disc non-pruinose; campylidia with a distinct socle and a rather small, usually dark brown lobe. . . . . [pantropical] ***Sp. leprieurii*** Mont.

11b Apothecial disc pruinose; campylidia without a distinct socle and with a large, brownish lobe . . . . . [neotropical and Africa] ***Sp. pilocarpoides*** (Zahlbr.) Lücking & Kalb

12a Apothecia 0.8-1.4 mm in diam., disc with spreading masses of torulose hyphae . . . . . [New Guinea] ***Sp. muscicola*** Lambley & Sérus.

12b Apothecia 0.5-0.8 mm in diam., disc smooth. . . . . [tropical Asia] ***Sp. argillaceum*** (Müll. Arg.) Zahlbr.

13a Thallus C and KC + orange, (containing methyl barbatate);apothecia unknown . . . . . [neotropical] ***Sp. leprosum*** Øvstedal & Elix

13b Thallus C and KC -; apothecia present or absent . . . . . 14

14a Ascospores 8/ascus; campylidia unknown . . . . . ***Sp. octosporum*** Lücking

14b Ascospores single in the ascus; campylidia usually present . . . . . 5

15a Conidia 1-septate, distal cell rounded and proximal cell ellipsoid-bacillar; campylidia with reduced socle and a large, greyish lobe with a whitish gray pruina . . . . . [tropical Africa] ***Sp. podosphaera*** Lücking & R. Sant.

15b Conidia non-septate, ellipsoid to guttuliform; campylidia without pruina . . . . . 16

16a Apothecia irregular, with thickly pruinose margin; campylidial wall paraplectenchymatous throughout; campylidia often pale yellowish . . . . . [pantropical] ***Sp. xantholeucum*** (Müll. Arg.) Zahlbr.

16b Apothecia regularly rounded; campylidial wall in most parts prosoplectenchymatous; campylidia usually pale grey . . . . . 17

17a Apothecial margin prominent, pale orange to pinkish; pannarin and zeorin absent . . . . . [amphipacific] ***Sp. marginatum*** Lücking

17b Apothecial margin not prominent, pale grey to whitish, pannarin and zeorin present . . . . . [pantropical] ***Sp. phyllocharis*** (Mont.) A. Massal.

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We wish to thank very much our colleague and friend Goran Thor for having made his collection from Sri Lanka available to us, through the courtesy of the herbarium in Stockholm (S). Drs. P. Diederich, D. Ertz and H. J. M. Sipman are also warmly thanked for their precious help in the preparation of the manuscript.

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# Taxonomic studies of *Alternaria* from China 11. Three large-spored new species

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**Abstract** — Three large-spored new species in the genus *Alternaria*: *A. amphicarpaeae* and *A. sojae* on plants of *Fabaceae* and *A. tribuli* on *Zygophyllaceae* are reported. *A. amphicarpaeae* and *A. sojae* are morphologically distinguishable from each other, and are also different from the other reported species of *Alternaria* on *Fabaceae*. *Alternaria tribuli* is the first species of *Alternaria* reported on *Zygophyllaceae*. Type specimens are deposited in the Herbarium of Shandong Agricultural University: Plant Pathology (HSAUP) and the Herbarium of Henan Agricultural University: Fungi (HHAUF).

**Key words** — taxonomy, hyphomycetes, leaf spots

In the course of a survey of mitosporic fungal pathogens of important weeds, three new taxa of *Alternaria* were found on *Amphicarpaea edgeworthii, Glycine soja* and *Tribulus terrestris* in Shandong Province and Henan Province of China. They are described as follows.

***Alternaria amphicarpaeae*** Meng Zhang & T.Y. Zhang, **sp. nov.** **FIGURE 1**

MycoBank MB 511321

*Conidia solitaria, obclavata, obpyriformia vel ovoidea, recta, subhyalina vel pallide brunnea, septa transversa 5–8, septa longitudinalia vel obliqua 2–6, constricta, 50–70 × 15.5–22µm, rostra filiformia, septata, hyalina, 54–140 × 2–3µm.*

Holotype: on leaf spots of *Amphicarpaea edgeworthii* Benth.: Taian, Shandong Province, China, leg. M. Zhang, Jun. 2003, *HSAUP*$_{23}$*0635* (=*ZM*$_{23}$*0135*, dried cultures on PCA + filter paper).

Leaf spots circular or irregular, pale brown in the central area, with dark brown margins, 3–6 mm in diameter. Colonies on PCA greyish brown, velvety, effuse,

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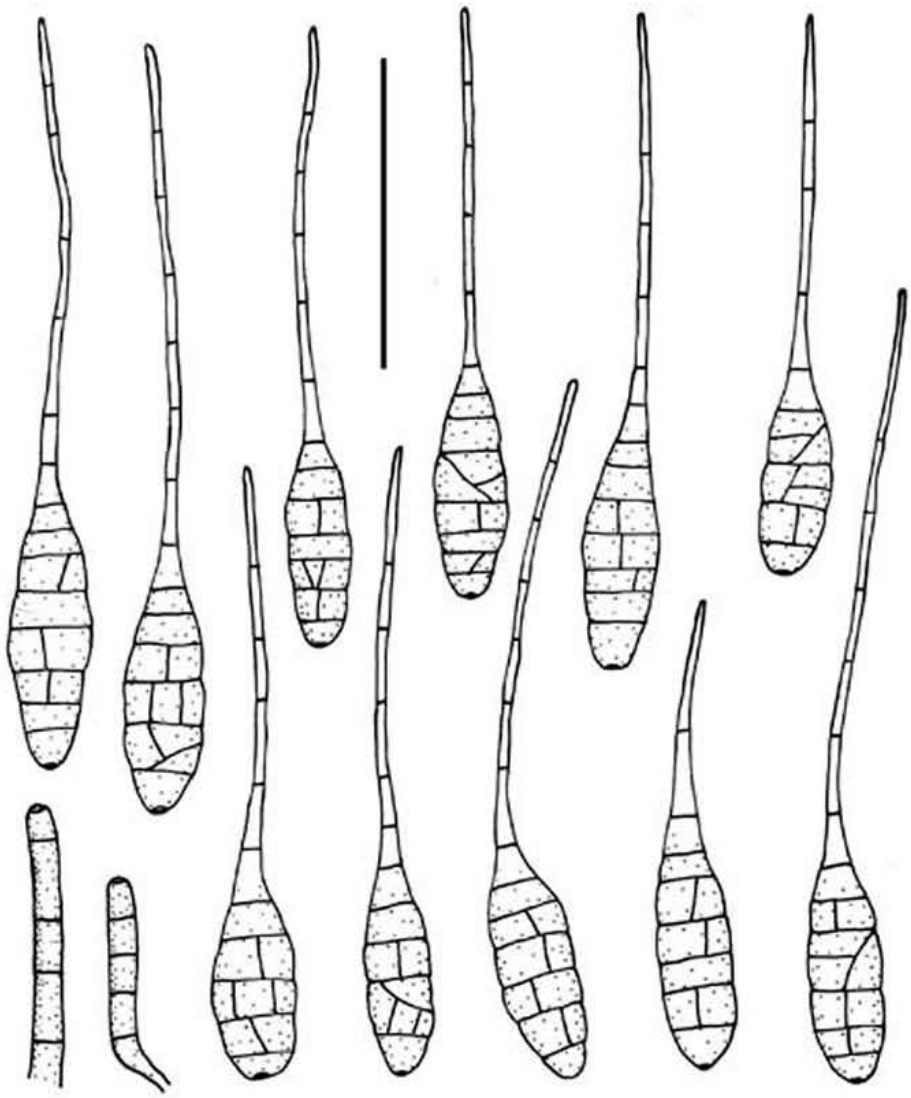

**FIGURE 1** *Alternaria amphicarpaeae*
Conidia and conidiophores on PCA + filter paper. (ex holotype, bar=50μm)

sporulating sparsely. On PCA + filter paper sporulating freely. Conidiophores arising singly, terminally or laterally from hyphae, simple, straight or curved, smooth-walled, septate, pale brown, 39–78 × 6–9μm. Conidia obclavate or obpyriform, base obtuse, straight, subhyaline to pale brown, smooth-walled, with 5–8 transverse septa and 2–6 longitudinal or oblique septa, constricted at some main septa, 50–70 × 15–22μm, av. 59.5 × 19.5μm. Rostra long, filiform, straight, septate, hyaline, 54–140 × 2–3μm.

*Alternaria amphicarpaeae* is similar to *A. cassiae* Jurair & A. Khan and *A. longirostrata* T.Y. Zhang & Meng Zhang in conidium morphology. The main distinctions between them are the conidium size range and shape. The conidia of *A. cassiae* (the description of original authors: 92–332 × 28–40μm; the description of E.G. Simmons: the body of conidia 65–90 × 20μm; Simmons 1982) are obviously larger than those of this new fungus. The conidia of *A. longirostrata* are narrower (av. 17.4μm) and its conidium rostra are distinctly longer (av. 180.5μm; Zhang et al. 2003) than those of the present fungus. Additionally, the light coloured conidia of *A. amphicarpaeae* are very helpful in distinguishing it from the other *Alternaria* species on plants of *Fabaceae*.

***Alternaria sojae*** Meng Zhang & T.Y. Zhang, **sp. nov.** FIGURE 2

MycoBankMB 511324

*Conidia solitaria, raro in catenis bisporis, fusiformia vel ellipsoidea, attenuata versus basim angustam, laevia, pallide brunnea, 4–12 transversa septa, 2–9 longitudinalia vel obliqua septa, constricta, 80–103 × 20–31.5μm, rostra vel pseudorostra filiformia, hyalina, 75–350 × 2–3μm.*

*Holotype: on leaf spots of Glycine soja Siebold & Zucc.: Luoyang, Henan Province, China, leg. M. Zhang, Oct. 2005, HHAUF $_{cs}$ 0360 (=ZM $_{cs}$ 0360, dried cultures on PCA).*

Leaf spots circular or subcircular, brown to black, 3–6mm in diameter. Colonies on PDA grey-brown, velvety, effuse, no sporulation. Colonies on PCA grey, velvety, effuse, sporulating freely. Conidiophores arising singly, terminally or laterally from hyphae, simple, straight or curved, smooth-walled, septate, medium brown, paler toward the apex, 70–156 × 4.5–6.5μm. Conidia fusiform or ellipsoidal, gradually subulate from the lower-middle toward the base, medium brown, smooth-walled, with 4–12 transverse septa and 2–9 longitudinal or oblique septa, 80–103 × 20–31.5μm. Rostra or pseudorostra very long, filiform, straight, sometimes curved, hyaline, 75–350 × 2–3μm, the apex of pseudorostra swollen to 4.5 μm wide.

The new species has the largest conidia of any *Alternaria* thus far described on *Fabaceae*. Its characteristic large, fusiform conidia and long filamentous rostra or pseudorostra are very useful in separating it from all other *Alternaria* species described from this plant family.

***Alternaria tribuli*** Meng Zhang & T.Y. Zhang, **sp. nov.** FIGURE 3

MycoBank MB 511325

*Conidia solitaria, obclavata vel obpyriformia, recta, pallide brunnea vel moderate brunnea, 3–8 transversa septa, 0–1–4 longitudinalia vel obliqua septa, partim constricta, crassa, 31–62 × 11–17.5μm. Rostra filiformia, septata, subhyalina, 50–190 × 2–3μm.*

Holotype: on leaf spots of *Tribulus terrestris* L.: Taian, Shandong Province, 2003, Coll. M. Zhang., *HSAUP $_{cs}$ 0700* (=*ZM $_{cs}$ 0200*, dried cultures on PCA).

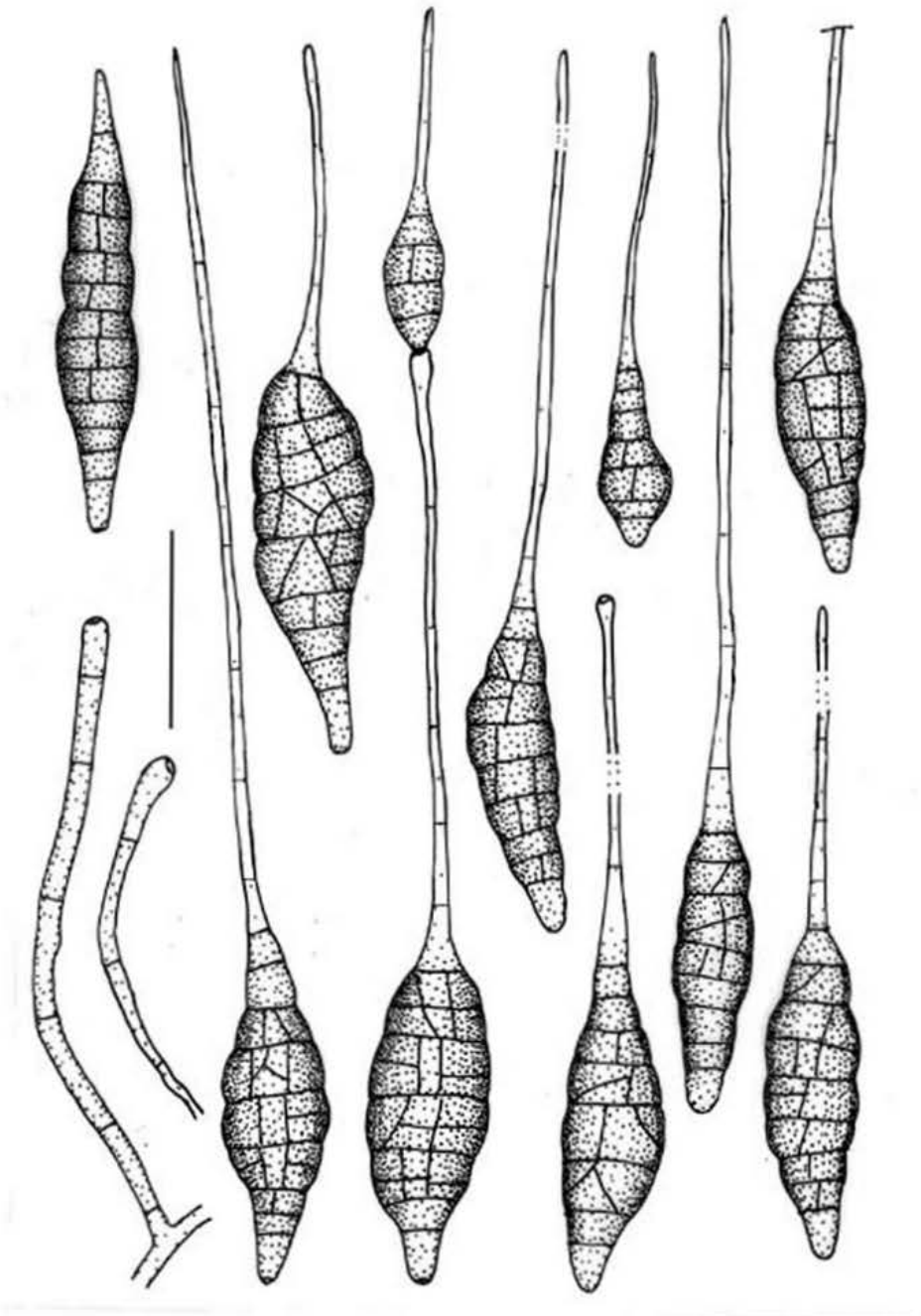

FIGURE 2 *Alternaria sojae*
Conidia and conidiospores on PCA. (ex holotype, bar=50μm)

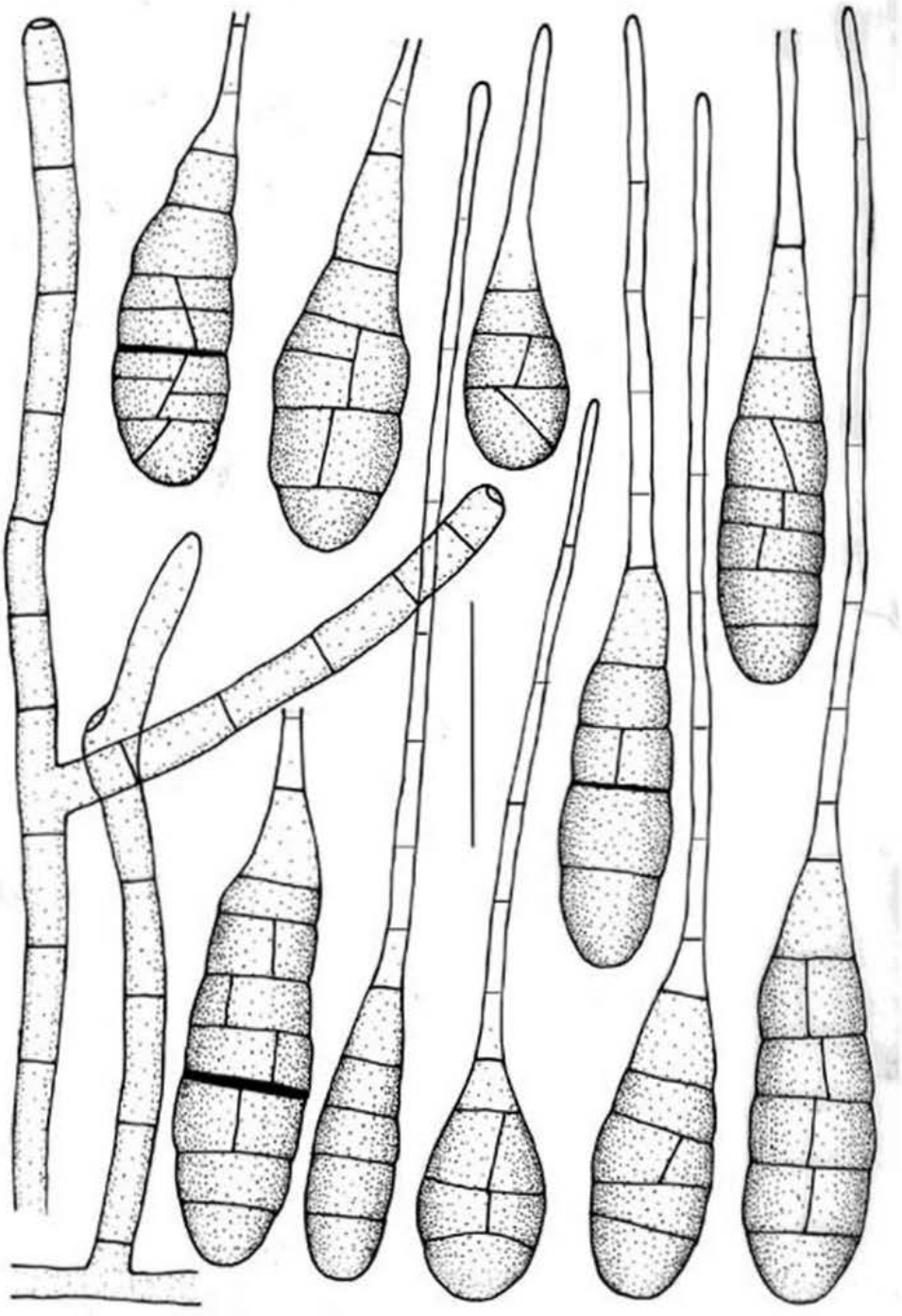

FIGURE 3 *Alternaria tribuli*
Conidia and conidiophores on PCA + filter paper. (ex holotype, bar=30μm)

Leaf spots subcircular or irregular, pale brown, 2–4mm in diameter. Colonies on PCA grey, velvety, effuse, sporulating sparsely. On PCA + filter paper sporulating freely. Conidiophores arising singly, terminally or laterally from hyphae, simple or branched, straight or curved, smooth-walled, septate, pale brown, paler toward the apex, 60–175 × 4.5–6.5µm. Conidia obclavate or obpyriform, pale brown to medium brown, smooth-walled, with 3–8 transverse septa and 0–1–4 longitudinal or oblique septa, constricted and thickened at some septa, 31–62 × 11–17.5µm, av. 47.5 × 15µm. Rostra filiform, straight, sometimes curved, subhyaline, 50–190 × 2–3µm.

This is the first species of *Alternaria* reported on *Zygophyllaceae*.

## Acknowledgments

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# Taxonomic studies of *Alternaria* from China 12. Three taxa on *Paeonia suffruticosa*

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**Abstract** — Three species of *Alternaria* on *Paeonia suffruticosa* are reported: *A. suffruticosae*, *A. suffruticosicola*, and *A. tenuissima*. The first two taxa are described as new species. *A. suffruticosae* is the first large-spored *Alternaria* species reported on plants of the family *Ranunculaceae*. *A. suffruticosicola* differs from other species reported on *Ranunculaceae* by its characteristic *Alternaria infectoria*-like sporulation pattern. Type specimens are deposited in the Herbarium of Henan Agricultural University: Fungi (HHAUF).

**Key words** — taxonomy, hyphomycetes, leaf spots

In the course of a survey of pathogenic fungi of *Paeonia suffruticosa* Andrews in Henan Province, three taxa of *Alternaria* were found. Two of them are new species. We describe them as follows.

***Alternaria suffruticosae*** Meng Zhang, Z.S. Chen & T.Y. Zhang, **sp. nov.** FIGURE 1
MycoBank MB 511326

*Conidia solitaria, obclavata, obpyriformia vel ovoidea, recta, moderate brunnea vel brunnea, transverse 4–10-septata, longitudinaliter vel obique 2–8-septata, constricta, 50–100 × 19–28µm, rostra filiformia, hyalina, 120.5–225 × 2.5–3.5µm.*

HOLOTYPE: on leaf spots of *Paeonia suffruticosa*: Zhengzhou, Henan Province, 2006, Coll. M. Zhang and Z. S. Chen, *HHAUF* $_{DS}$ *0572–1=ZM* $_{DC}$ *0572–1*, dried cultures on PCA+ filter paper.

Leaf spots circular or irregular, brown to black, 3–10mm in diameter. Colonies on PDA brown, velvety, effuse, no sporulation. Colonies on PCA grayish-brown, velvety, effuse, sporulating sparsely. On PCA + filter paper sporulating freely. Conidiophores arising singly or in groups terminally or laterally from hyphae,


FIGURE 1 *Alternaria suffruticosae*
Conidia and conidiophores on PCA + filter paper. (ex holotype)

simple, straight or curved, smooth-walled, septate, pale brown, paler toward the apex, 60–93 × 4–6μm. Conidia ovoid, obpyriform or obclavate, medium brown to brown, smooth-walled, with 4–10 transverse septa and 2–8 longitudinal or oblique septa, some conidial cells bulge to one side, 50–100 × 19–28μm. Rostra filiform, long, straight or slightly curved, subhyaline, 120–225 × 2.5–3.5μm.

FIGURE 2 *Alternaria suffruticosicola*
Conidia, conidiophores and sporulation patterns on PCA + filter paper.
(ex holotype, all bars=30μm)

***Alternaria suffruticosicola*** Meng Zhang, Z.S. Chen & T.Y. Zhang, **sp. nov.** FIGURE 2
MYCOBANK MB 511327

*Conidiophora solitaria, pallide brunnea, erecta, septata, 20–50 × 4–5.5μm. Conidia catenulata, obclavata vel ellipsoidea, recta, pallide brunnea vel medio brunnea, septa transversa 2–6, septa longitudinalia vel obliqua 0–1–3, partim constricta, 13–40 × 7.5–11.5μm. Rostra curta vel nulla, pseudorostra cylindrica, simplicia vel ramosa, septata, subhyalina, apice inflata, 3–57 × 2–4.5μm.*

HOLOTYPE: on leaf spots of *Paeonia suffruticosa*: Zhengzhou, Henan Province, 2006, Coll. M. Zhang and Z. S. Chen, *HHAUF* $_{DS}$ *0572–2=ZM* $_{DS}$ *0572–2*, dried cultures on PCA + filter paper.

Leaf spots circular or irregular, brown to black, 3–10mm in diameter. Young colonies (5–7 d) on PCA greyish brown, velvety, effuse, sporulating freely. Conidiophores arising singly, terminally or laterally from hyphae, simple, straight or curved, smooth-walled, septate, pale brown, paler toward the apex, 20–50 × 4–5.5μm. Conidia catenulate, obclavate or ellipsoidal, pale brown

to medium brown, smooth-walled, with 2–6 transverse septa and 0–1–3 longitudinal or oblique septa, constricted at some septa, 13–40 × 7.5–11.5μm. Rostra short or none; pseudorostra commonly produced from any cells of conidia, simple or branched, straight or curved, sometimes apex geniculate and inflated, subhyaline to pale brown, septate, 3–57 × 2–4.5μm.

*A. suffruticosae* and *A. suffruticosicola* occur on the same leaf spots of *Paeonia suffruticosa*. But they are morphologically distinguishable from each other. There are three previously reported species of *Alternaria* on *Ranunculaceae*. One is *Alternaria tenuissima* on *Ranunculus asiaticus* L. Zhang et al. 2003, The second is *Alternaria thalictrina* Ondřej on *Thalictrum aquilegiifolia* L. (Ondřej 1974). The third is *Alternaria thalictricolor* Y.L. Guo on *Thalictrum macrorhynchum* Franch. (Guo 1999). The conidia of *A. suffruticosae* is obviously bigger than those of the three reported species. It is also the first large-spored species of *Alternaria* on *Ranunculaceae* with a long filamentous beak. The sporulation patterns of *A. suffruticosicola* belong to those of the *Alternaria infectoria* species-group; individual sporulation clumps are relatively open; the pseudorostra of main central conidia in the chains are usually longer than conidia that produce them. Therefore it is different from other reported species of *Alternaria* on *Ranunculaceae*.

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≡ *Helminthosporium tenuissimum* Kunze (1818), in Nees & Nees, Nova Acta Phys.-Med. Acad. Caes. Leop.-Carol. Nat. Cur. 9: 242.
≡ *Macrosporium tenuissimum* (Kunze) Fr. (1832), Syst. Mycol. 3: 374.
= *Alternaria humicola* Oudem. (1902), Arch. Néerl. Sci. Exact. Nat. 7: 292.

Specimen: on leaf spots of *Paeonia suffruticosa*: Luoyang, Henan Province, 2006, Coll. M. Zhang and Z.S. Chen, *HHAUF* $_{cf}$ *0468 (=ZM* $_{cf}$ *0468)*.

*A. tenuissima* is a very familiar species of *Alternaria* reported on various substrates. It also occurs on leaf spots of *Paeonia suffruticosa*, the spots circular or irregular, brown to black, 3–7mm in diameter.

## Acknowledgments

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Zhang TY, Zhang JZ, Chen WQ, Zhang M, Sun GY, Gao MX. 2003. Flora Fungorum Sinicorum, 16: *Alternaria*. Beijing: Science Press, 1–283 + Plates (in Chinese).

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# *Pluteus neotropicalis* (*Pluteaceae, Agaricales*), a new species from tropical-subtropical Mexico

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**Abstract** — The new species *Pluteus neotropicalis* occurs in habitats situated in tropical and subtropical regions in the states of Colima, Quintana Roo, and Veracruz, Mexico. Morphologically, *P. neotropicalis* is most closely related to the northern temperate *P. thomsonii*, from which it is differentiated by its obtuse and short rostrate cystidia.

**Key words** — agaric, *Basidiomycota*, *Celluloderma*, *Pluteus thomsonii*

## Introduction

The majority of *Pluteus* Fr. species have been described from temperate regions, with comparatively few taxa reported from the tropics. In Mexico, *Pluteus* species have been studied by Singer (1973), Cifuentes & Guzmán (1981), Candusso et al. (1994), Rodríguez & Guzmán-Dávalos (1997, 1999, 2000, 2001), Ramírez-Guillén & Guzmán (2003), Rodríguez et al. (1997, 2004), and Rodríguez (2005). So far, thirty three species of *Pluteus* have been reported from Mexico (Rodríguez 2005). Several of those occur in tropical-subtropical habitats, e.g. *Pluteus aethalus* (Berk. & M.A. Curtis) Sacc., *P. horridus* Singer, *P. leucocyanescens* Singer, *P. oligocystis* Singer, and *P. triplocystis* Singer from tropical rain forests; *P. martinicensis* Singer & Fiard from deciduous tropical forests (Rodríguez 2005), and *P. xylophilus* (Speg.) Singer from subtropical and pine-oak forests (Ramírez-Guillén & Guzmán 2003, Rodríguez 2005).

Below we describe a new species of *Pluteus* collected from tropical-subtropical forests that includes one collection (Guzmán 30963) previously indicated as Ramírez-Guillén (1998) representing a probable new taxon.

## Material and methods

Microscopic observations were made from sections of basidiomata mounted in 3 % KOH solution. Basidiospore shape was determined according to Bas (1969) using the Q coefficient (length-width ratio) determined from 20 randomly selected basidiospores. Descriptive terms follow Vellinga (1998).

## Taxonomy

***Pluteus neotropicalis*** O. Rodr.-Alcántar, **sp. nov.** FIGS. 1-6

MycoBank MB 511284

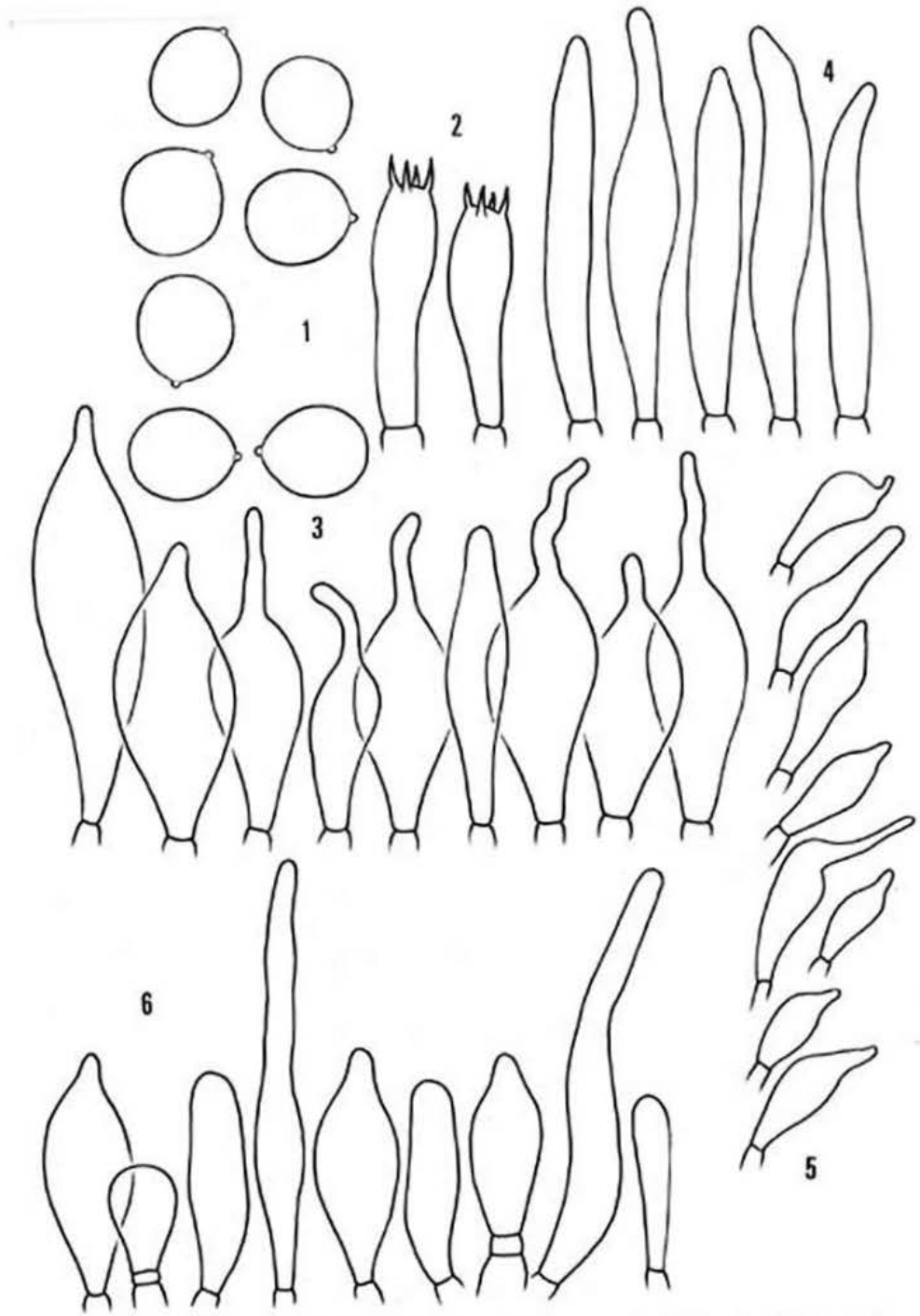
*Pileus 10-18 mm latus, marginem pallidiorem versus striato-sulcatus, velutinus, ad discum rugosovenosus, griseo-brunneus vel rubellus-brunneus. Lamellae confertae, ventricosae, albidae vel roseae, ad aciem subfimbriatae, haud coloratae. Stipes 12-22 × 1-2 mm, centralis, basim versus glaber vel floccosus, albido-griseus dein brunneus. Sporae 5.5-7(-8) × (4-) 5-6 µm, subglobosis vel late ellipsoideis, laevis, subcrassus tunicatus (0.8 µm), hyalinis. Cheilocystidia (et pleurocystidia valde similia) 32-62.5(-68.5) × 10-19(-23)µm, fusiformia vel claviformia, apicaliter rostro 5-28 µm longlongo et obtuso frequenter instructa, abundantes. Pileipellis typi Mixtini, cellulae subglobosae, ellipsoideaes vel claviformia compositus, 17-45 × 12-21(-28) µm, pileocystidia 42.5-67.5 × (9.5-)10-19(-20) µm, anguste utriformia, lageniformia vel fusiformia, saepe mucronata, pigmento brunneolo instructa. Caulocystidia (15-)25-61(-67.5) × (6-) 10-15 (-23.5) µm, cheilo- et pleurocystidiis similia, hyalina. Fibulae nullae. Ad lignum putridum. Mexico. Holotypus G. Guzmán 20644 (XAL).*

Holotypus: Mexico, Quintana Roo: Municipality of Felipe Carrillo Puerto, 4.XI.1981, G. Guzmán 20644 (XAL).

Etymology: *neotropicalis*, named based on its neotropical distribution.

Pileus 10-18 mm broad, conic-campanulate when young, convex to plane in mature specimens, margin striate-sulcate, surface velutinous, wrinkled or venose in the disk, at first grayish-brown to reddish-brown in center, light brown, pinkish-brown to yellowish-brown upon drying or in age, dry. Lamellae free, crowed, ventricose, at first whitish, gradually becoming pink; entire or fimbriate, concolorous edges. Stipe 12-22 × 1-2 mm, central, equal or slightly enlarged towards base, fragile, minutely pruinose, floccose near base, brown with whitish-grayish or pink tinges, with whitish mycelium at base. Context concolorous, unchanging upon exposure. Basidiospores 5-6 (-7) × (4-) 4.5-5.5 (-6) µm, Q = 1.05-1.15 (-1.25), suglobose or globose (rarely broadly ellipsoid), smooth, thin-walled, with refringent content, hyaline.

Basidia 24-28 × (6.5-) 7-8 (-11) µm, 4-spored, subclavate, hyaline, clampless. Cheilocystidia 25-45 (-62.5) × 7-15 (-19) µm, slender fusoid or broadly fusoid, at apex with long (5-28 µm), cylindrical projection, thin-walled, hyaline, plasmatic pigment absent. Pleurocystidia 60-100 × 10-16 µm, subcylindrical or slender fusoid, thin-walled, hyaline, intermixed (near lamellar edges) with cheilocystidia-like cells, scattered. Pileipellis (sect. *Celluloderma*,




FIGS.1-6: *Pluteus neotropicalis* (Holotype). 1: basidiospores (x 2000), 2: basidia (x 1000), 3: cheilocystidia (x 1000), 4: pleurocystidia (x 1000), 5: caulocystidia (x 500), 6: pileipellis (x 500).

subsect. *Mixtini*) a celluloderm, composed of vesiculose, clavate, cylindrical or broadly fusoid cells, 25-60 × 6-20 µm, intermixed with slender fusoid, thin-walled, hyaline pileocystidia (60-100 × 10-15 µm). Oleiferous hyphae absent. CAULOCYSTIDIA (15-) 20-61 (-67.5) × (6-) 8-15 µm, shape like cheilocystidia, rarely subfusoid, thin-walled, hyaline, numerous. Clamp connections absent.

HABITAT – Lignicolous, gregarious, in tropical evergreen forests and subtropical montane cloud forests.

MATERIAL EXAMINED – MEXICO: **Colima:** Municipality of Ixtlahuacán, 5 km near to Agua de la Virgen towards the town 26 de Julio, alt. 590 m, 26.IX.1996, *O. Rodríguez 1107* (paratype IBUG). - **Quintana Roo:** Municipality of Felipe Carrillo Puerto, Zona de Chunyaxché, near to road Puerto Morelos to Carrillo Puerto, 4.XI.1981, *G. Guzmán 20634* (paratype ENCB); *G. Guzmán 20644* (holotype XAL, isotypes ENCB, IBUG, ZT). - **Veracruz:** old road Xalapa to Coatepec, near to km 6, Municipality of Coatepec, Congregación Zoncuantla, alt. 1270 m, Dr. Gastón Guzmán's house, 14.IX.1994, *G. Guzmán 30963* (paratype XAL); 1.XII.1998, *G. Guzmán 32758* (paratype XAL).

ADDITIONAL MATERIALS EXAMINED REFERRING TO *PLUTEUS THOMSONII* - ENGLAND: West Farleigh, near Maidstone (ex herb. M. J. Berkeley, K-93764, holotype). - SPAIN: Prov. Madrid, El Plantío La Florida, 2.X.1975, *M. Piera s.n.* (AH-945). - SWITZERLAND: Kerns OW (Kernwald), quadrant 1966, alt. 600 m, 17.VIII.1992, *FK-1708-92 KI* (NMLU).

OBSERVATIONS - *Pluteus neotropicalis* is distinguished by the size and shape of the cheilocystidia, the pileipellis structure, and habitat. Due to the lacunose-venose, grayish-brown pileus, the most closely related species based on morphology is *P. thomsonii* (Berk. & Broome) Dennis, which is reported from Europe, North America and North Africa (Citérin & Eyssartier 1998) and cited as restricted to temperate habitats (Orton 1986, Vellinga 1990).

Erroneously, Rodríguez & Guzmán-Dávalos (1999) previously referred collection Rodríguez1107 from Mexico to *P. thomsonii*. However, our subsequent comparison of additional Mexican material and European *P. thomsonii* collections reveal that in *P. thomsonii* the cheilocystidia have longer (≤ 50 µm) acute apices. The type of *Pluteus thomsonii* is in fragmentary condition; only basidiospore size and shape could be confirmed, as all other micromorphological structures had collapsed. Based on their description of the tropical habitat and cystidia with short rostrate apices, the report by Wartchow et al. (2004) of *P. thomsonii* from a subtropical rain forest in Brazil should probably be referred to *P. neotropicalis*,.

*Pluteus insidiosus* Vellinga & Schreurs, described from Europe, is also similar morphologically, but the pileipellis hymeniderm is composed exclusively of claviform or obovoid cells and lacks distinctive pileocystidia (Vellinga & Schreurs 1985, Vellinga 1990, Breitenbach & Kränzlin 1995). A further morphologically related species is *P. diverticulatus* Corriol, also reported from Europe, which can be separated by its diverticulate and polymorphic cheilocystidia, pileipellis structure, and caulocystidial shape (Corriol 2003).

## Acknowledgement

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# *Basidiopycnides albertensis* gen. et sp. nov., a new anamorphic fungus with phylogenetic affinities in the *Atractiellales (Basidiomycota)*

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**Abstract** — A new anamorphic genus, *Basidiopycnides*, and its type species, *Basidiopycnides albertensis*, are described. Strains of *Basidiopycnides albertensis* produce what superficially appeared to be a *Graphium*-like conidial state with percurrently proliferating annellophores. Detailed morphological and molecular data analysis showed these isolates represent a new taxon that belongs to the *Atractiellales (Basidiomycota)*.

**Key words** — bark beetles, DAPI staining, molecular phylogeny

## Introduction

In 1987 collections of bark beetle infested material from various coniferous hosts yielded isolates resembling synnematous species of either *Graphium* Corda or *Pesotum* J.L. Crane & Schokn. But since then, species of these and other similar genera have been studied extensively. Seifert & Okada (1993) proposed that *Graphilbum* H.P. Upadhyay & W.B. Kendr., *Pesotum*, *Hyalopesotum* H.P. Upadhyay & W.B. Kendr., *Phialographium* H.P. Upadhyay & W.B. Kendr., *Pachnodium* H.P. Upadhyay & W.B. Kendr., and *Graphiocladiella* H.P. Upadhyay were all synonymous with *Graphium*. However, later detailed morphological and molecular taxonomic studies showed that *Graphium* and *Pesotum* are distinct (Okada et al. 1998b, 2000), and that while *Graphium* species are linked phylogenetically to the *Microascales*, *Pesotum* species are linked to the *Ophiostomales* (Okada et al. 1998b, 2000; Jacobs et al. 2003). Therefore, *Ophiostoma* anamorphs formerly placed in *Graphilbum*, *Hyalopesotum*,

*Phialographium*, *Pachnodium*, and *Graphiocladiella* now should be treated under *Pesotum* J.L. Crane & Schokn. emend. G. Okada & Seifert (Okada et al. 1998b, p. 1503; 2000).

Although two variants of percurrently proliferated conidiogenous cells were recognized in *Graphium* species (Seifert & Okada 1993, Okada et al. 1998b, 2000), both are said to be enteroblastic annellidic: one form produces nodular annellations; the second a mixture of nodular and dense annellations. In contrast conidiogenesis in *Pesotum* is either holoblastic or enteroblastic phialidic (Okada et al. 1998b).

Five cultures isolated from material collected in Banff National Park, Alberta, Canada, exhibited serial percurrent formation and secession of holoblastic conidia that resulted in the formation of proliferated annellophores sensu Kiffer & Morelet (2000), with elongate intervals between the successive annellations that mark spore-production sites. However, the nodular annellations seen in some *Graphium* species were absent. The conidia ultimately aggregated into slimy masses, but the conidiophores were neither verticillate nor penicillate. Their macronematous conidiomata also differed markedly from synnemata produced by *Pesotum* and *Graphium* species, in that they developed from simple erect hyphal initials that produced basally constricted branches from just below the septa. These branches curved sharply upward immediately after initiation, and grew parallel to the parent element; each branch produced several new branches in a similar manner. At the apices of the ultimate branches, the annellophores were formed. There was a wide range of sizes noted amongst the conidiophores observed, and when conidiomata coalesced laterally, top-shaped structures were formed with globules of conidia at their apices. This developmental pattern is different from those of synnemata as described by Seifert (1985) and Seifert & Okada (1990).

As the conidiogenesis and conidiophore branching of our strains were similar to certain basidiomycete species, e.g., *Stilbotulasnella conidiophora* Bandoni & Oberw. (1982), *Gloeosynnema ochroleucum* (Penz. & Sacc.) Seifert & G. Okada (1988), *Gloeosynnema roseum* Matsush. (1995), *Filosporella annelidica* (Shearer & J.L. Crane) J.L. Crane & Shearer (1977), and species of the ascomycete genus *Pyxidiophora* Bref. & Tavel (Blackwell & Malloch 1989), we used sequence analyses of the 18S ribosomal RNA gene (18S rDNA) and rDNA internal transcribed sequences (ITS), and incident-light fluorescence microscopy to determine their phylogenetic relationship and nuclear status.

## Materials and methods

**Strains studied and morphology.** Five isolates, whose collection details are given under the new species description, were obtained from host material from which beetles had been removed at the collection site according to

Hutchison & Reid (1988) and Eyjólfsdóttir (1990). These have been maintained at 4 °C in darkness on slants of malt extract agar (Difco, MI; Fisher Scientific Fair Lawn, NJ) plus yeast extract (MEYE, Hausner et al. 2003) and culture descriptions on agar were also prepared according to Hausner et al. (2003). Isolates were also grown on autoclaved *Pinus mugo* Turra and *Pinus sylvestris* L. twigs embedded aseptically in both 2.0 % water agar or MEYE in Petri dishes at 20 °C to see if different forms of fruiting occurred. The twig culture technique was repeated with our strains using three separate sets of twig-plates inoculated in combination with one of three strains of *Clonostachys rosea* (Link) Schroers et al. (1999); the latter had successfully induced perithecial formation in strains of *Ceratocystiopsis falcata* (E.F.Wright & Cain) H.P. Upadhyay [ = *Cornuvesica falcata* (E.F. Wright & Cain) Viljoen et al.] (Hutchison & Reid 1988, Kawchuk et al. 1993). Inoculated twig plates lacking the *Clonostachys* strains served as controls. Portions of both living and dried specimens have been deposited in the University of Alberta Microfungus Collection and Herbarium (UAMH), Devonian Botanic Garden, and are also held in the Biological Sciences Department [WIN(M)] of the University of Manitoba.

Morphological structures used to identify isolates were mounted on slides in either 85 % lactic acid or 85 % lactic acid/water solutions (1:15 by volume), and cover slips were ringed with nail varnish prior to observation. But the pure 85 % lactic acid mounts were placed on a slide warmer at 40 °C for 24 hours before ringing, while the lactic acid/water mounts were ringed and studied immediately; the latter yielded clearer preparations (Eyjólfsdóttir 1990). At least 50 measurements of spores were recorded for statistical analysis, and either a Leitz Ortholux II or a Zeiss Photomicroscope II was used for observations, drawings and photography. We used Munsell Soil Color charts (Kolmorgen Corp., Baltimore, MD) to categorize for colours, and Gams (1971) for descriptions of colony texture and conidiophore organization.

**Nuclear staining and incident-light fluorescence microscopy.** Aseptically, in a laminar flow chamber, sterile slides were dipped in sterile molten malt extract (2 g malt extract, 20 g agar, 1 l tap water). One side was wiped clean, and the slide placed agar side up on a sterile bent glass rod in a sterile Petri dish moist-chamber lined with several layers of moistened sterile paper toweling. Slides were inoculated aseptically at the centre with a spore mass from a fresh culture of the selected test strain, and the Petri dishes were incubated at 20 °C for 7 days. If at that time growth had occurred, slides were fixed in 100 % methanol and stained with DAPI (4', 6-diamadino-2-phenylindole) according to Hamada & Fujita (1983) as described by Okada et al. (1998a). But we replaced the 2-mercapto-ethylamine in the staining solution with 2-mercaptoethanol (Aldrich Chemicals, Milwaukee, WI); the latter had been shown to be highly effective in fungi (M. Young, personal communication).

When maintained on ice in a refrigerator at 4 °C, these slides yielded good results for up to 5 days after preparation. The nuclei were visualised using a Zeiss Axio Imager and the images collected digitally with an AxioCam Mrm camera.

**DNA extraction and amplification protocols.** DNA extraction, purification, and agarose electrophoresis protocols were those of Hausner et al. (1992). Whole cell DNA served as the template for amplifying DNA fragments of interest using the Invitrogen-BRL PCR System (Buffer and Taq polymerase, Invitrogen, Fredrick, MD). Primers SSUZ and LSU4 (Hausner et al. 2005) were used to amplify the ITS regions, and primers SSJ and SST were employed to recover the 18S rDNA. The PCR primer sequences, amplification conditions, sizes of the expected PCR products, and preparation of sequencing templates for ITS, and 18S rDNA fragments have been described previously (Hausner & Reid 2004, Hausner & Wang 2005, Hausner et al. 2005). DNA sequencing templates were prepared with the aid of the Promega Wizard SV Gel and PCR clean-up system (Promega, Madison, WI). Purified double-stranded PCR products were sequenced in both directions using the cycle-sequencing protocols performed according to the manufacturers' recommendations (Perkin Elmer Applied Biosystems, Foster City, CA), and automated Fluorescent DNA sequence analysis was performed using an ABI Prism 310 Genetic Analyzer system (PEAB at the University of Calgary, DNA sequencing facility, Calgary, AB).

**Analyses of DNA sequence data.** Forty-seven 18S rDNA sequences were aligned with CLUSTAL-X (Thomson et al. 1997) and, when appropriate, modified with the alignment editor program GeneDoc v2.5.010 (Nicholas et al. 1997, http://www.psc.edu/biomed/genedoc). The alignment (EMBL-Align database accession: ALIGN_001145) covered 1742 positions and the 18S rDNA sequence of *Saccharomyces cerevisiae* Meyen ex E.C. Hansen var. *cerevisiae* served as the outgroup in phylogenetic analysis. First, programs contained within PHYLIP (Felsenstein 2006, Version 3.66; http://evolution.genetics.washington.edu/phylip/getme.html) were used to resolve phylogenetic relationships among the tested sequences, and the data set was then analyzed with DNAPARS (maximum parsimony) and DNADIST (F84 setting). From the latter, the distance matrix generated was utilized in the NEIGHBOR program (NJ setting) for inferring a phylogenetic tree. The phylogenetic estimates were evaluated using the bootstrap procedure (SEQBOOT 1000 replicates; and CONSENSE) in PHYLIP. The 18S rDNA data was also analyzed with the Tree-Puzzle (TP) program [maximum likelihood (ML) phylogenetic analysis using quartets and parallel computing (Schmidt et al. 2002)]. The settings for the quartet puzzling algorithm were as follows: 10000 puzzling steps, transition/transversion parameter estimated from data set, HKY evolutionary model

(Hasegawa et al. 1985). However tree topologies were essentially identical to those of parsimony and NJ analysis. Finally the MrBayes (version 3.1) program (Ronquist & Huelsenbeck 2003; Ronquist 2004) was used for Bayesian analysis. The NEXUS file format necessary for the alignment (input) file was generated with the file converting option available within the DAMBE (Xia 2001). The DNA substitution model setting for Bayesian analysis was: GRT, gamma distribution with 4 gamma rate parameters. The model of DNA substitution was chosen based on evaluating the 18S rDNA alignment with the Modeltest 3.0 program (Posada & Crandall 1998). The Bayesian inference of phylogenies was initiated from a random starting tree, and four chains were run simultaneously for 250000 generations; trees were sampled every 100 generations. The first 25 % of trees generated were discarded ("burn-in") and the remaining trees were used to construct both a 50 % majority rule consensus tree and compute the posterior probability values.

## Results

Our fungus morphologically resembled anamorphs of both some ascomycetous and basidiomycetous taxa, but the nuclear staining and fluorescent microscopy clearly showed that individual hyphal cells, conidiogenous cells, and conidia were very rarely other than binucleate (see under Taxonomy). This dikaryotic condition suggests these isolates are the anamorph of a basidiomycete.

This staining technique produced surprisingly good nuclear staining and, unexpectedly, it also showed the proliferated annellophores extremely well.

To investigate the molecular phylogeny of our fungus, the 18S rDNA sequences were determined for two of our isolates [UAMH 10782 and 10785], these were identical (GenBank EF406118), and were used as queries in a Blastn (Altschul et al. 1997) NCBI database search. The results showed they were closely related to the holotypes of *Basidiopycnis hyalina* Oberw. et al. and *Proceropycnis pinicola* M. (Oberwinkler et al. 2006), both monotypic genera. At least partially by comparing 18S rDNA sequences of their two new species with a variety of basidiomycete sequences, six of which were from species of the *Atractiellales*, they placed their new species within the order *Atractiellales*, class *Atractiellomycetes*, subphylum *Pucciniomycotina*, division *Basidiomycota* (Bauer et al. 2006).

Because our strains closely resembled *Basidiopycnis hyalina* in some features, 18S rDNA sequences of the following species were included in the phylogenetic analysis: *Basidiopycnis hyalina*, *Proceropycnis pinicola*, *Phleogena faginea* (Fr.) Link, *Helicogloea lagerheimii* Pat., and *Atractiella solani* (Cohn & J. Schröt.) Oberw. & Bandoni (Fig. 1A). Various ascomycete taxa that morphologically resembled our isolates were also included, e.g. *Ophiostoma ulmi* (Buisman) Nannf.(has a *Pesotum* anamorph) and several *Graphium* species. Phylogenetic analysis of the 18S rDNA data using four different (NJ, Parsimony, Bayesian,

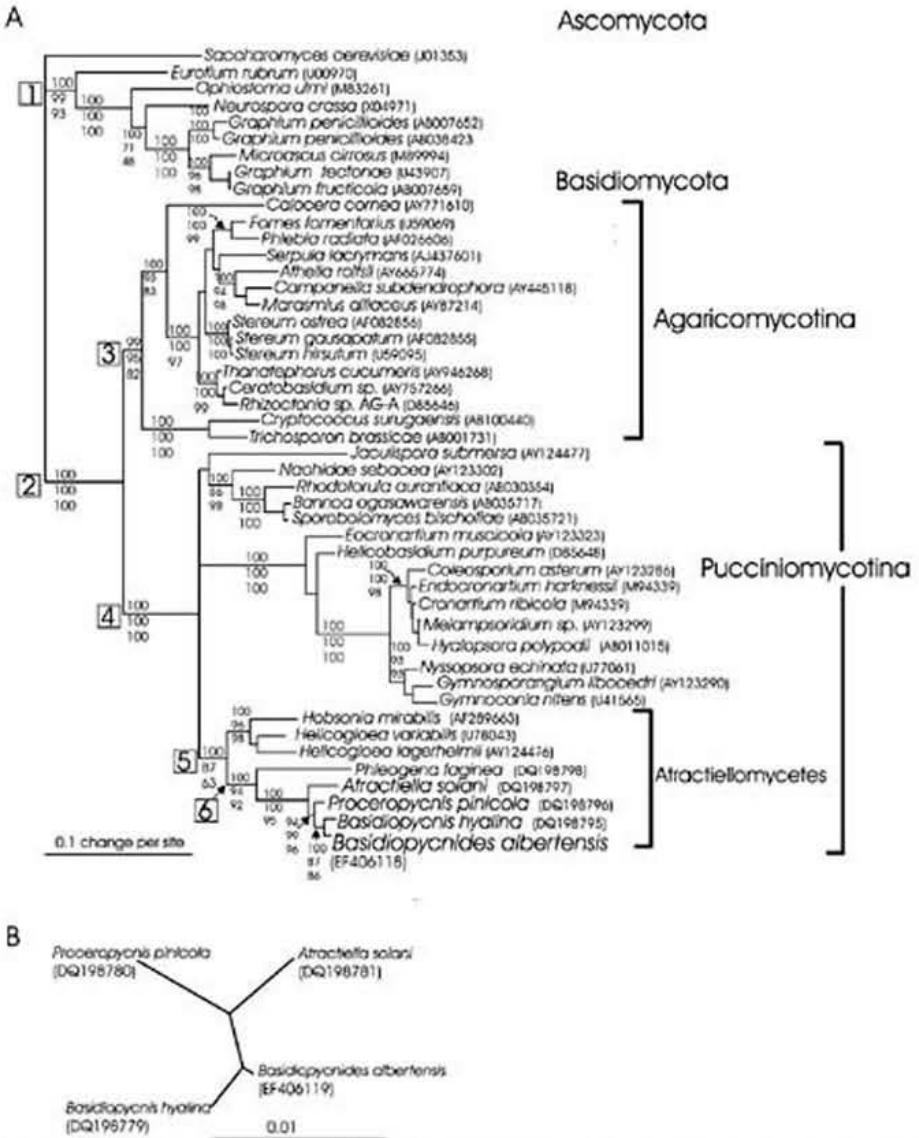


Fig. 1: Phylogenetic analysis of the 18S rDNA sequence data. A. Phylogenetic tree for *Basidiopycnides albertensis* within the *Atractiellales*. Tree is based on a 50 % majority rule consensus tree obtained from Bayesian inference. The first number at the nodes above the line represents the posterior probability values obtained from Bayesian analysis. The second and third number (below the lines) at the nodes indicate the level of support based on bootstrap analysis (Felsenstein 1985; % based on 1000 bootstrap replicates) in combination with NJ and PARS analysis respectively. The 18S rDNA alignment can be retrieved from the EMBL-Align database under the following accession: ALIGN_001145. B. The unrooted Neighbour-Joining (PHYLIP, NJ, DNADIST: setting K84) phylogenetic network showing the relatedness among partial ITS region sequences for four species of the *Atractiellales* including *Basidiopycnides albertensis*. For those sequences that were obtained from NCBI the GenBank accession numbers are provided for in brackets following the species names. The phylogenetic tree (A) and dendrogram (B) were drawn with the TreeView program (Page 1996) using the PHYLIP tree outfiles, and annotations were added to the figures with the aid of Corel Draw (Corel Corporation and Corel Corporation Limited, Ottawa, Canada).

Maximum-likelihood as implemented in TreePuzzle) methods for inferring evolutionary relationships yielded trees with essentially similar topologies (Fig. 1A).

This 18S rDNA data set also separated members of the *Ascomycota* from those of the *Basidiomycota* and, as expected from previously proposed phylogenies (Sikaroodi et al. 2001, Swann et al. 2001, Bauer et al. 2006, Blackwell et al. 2006), placed members of the *Agaricomycotina* and *Pucciniomycotina* of the *Basidiomycota* into specific clades. The 18S rDNA sequences of UAMH 10782 and 10785 grouped within a clade that included species of the class *Atractiellomycetes* (see Fig 1A, node 5). More precisely, our strains, under the name of *Basidiopycnides albertensis* (see Taxonomy) , were placed within the *Atractiellales* in a monophyletic clade that contains *Basidiopycnis hyalina, Proceropycnis pinicola, A. solani,* and *Phleogena faginea* (Fig. 1A, node 6). Most notably, the18S rDNA sequences of these strains differed by only 3 substitutions from the 18S rDNA sequence of *B. hyalina,* but there were 5 and 6 nucleotide differences between the UAMH strains and the sequences of *A. solani* and *Proceropycnis pinicola,* respectively.

The rDNA ITS regions (ITS1, 5.8S rDNA, ITS2) for all of our five strains (UAMH 10782 to UAMH 10786) were sequenced (GenBank EF406119), and these ITS regions of 551 bp, were identical. Partial ITS region sequences (405 bps) from *Basidiopycnis hyalina* (DQ198779), *P. pinicola* (DQ198780) and *A. solani* (DQ198781) were compared with those of our strains (Fig. 1B) and, excluding gaps, there were 16 nucleotide differences between the ITS sequences of the UAMH strains and that of *B. hyalina*, compared to 46 and 52 nucleotide differences between our strains and the ITS sequences of *P. pinicola* and *A. solani* respectively.

These results show that our isolates are distinct from related species, and we propose a new species in a new anamorphic basidiomycete genus.

## Taxonomy

***Basidiopycnides*** J. Reid, Eyjólfsd. & G. Hausner, **gen. nov.**

MycoBank MB510902

*Hyphomycetus. Hyphae hyalinae, leaves, septatae sine fibulis, cellulae binucleatae. Conidiophora ab initio ex hypha singulare erecta constantia; postea regulatim multo ramificantia, sic habitu mononemata vel macronemata. Conidiophora singula suis lateribus adhaerescentia, habitu saepe turbinata. Cellula conidiogena terminalis, annellophorum percurrens, primo ad apicem conidium singulare formans posteaque, primum conidium secessum, successive conidia additicia holoblastica formans. Conidia in guttas mucosas aggregata, unicellularia, hyalina, laevigata; oblonga vel breviter clavata apicibus obtusis, sed basibus truncatis cum fimbriis curtis basilaribus. Conidia plerumque tempore germinationis nova annellophora emittunt.*

Etymology: From the generic name *Basidiopycnis*, and -ides (Latin, = like, connected with)

**Type species:**

***Basidiopycnides albertensis*** J. Reid, Eyjólfsd. & G. Hausner, **sp. nov.**

MycoBank MB 510903 Figs. 2 - 6.

*Hyphomycetus. Coloniae in agaro cum extracto malti fermentique ad 20 °C aut 16-32 mm diametro post 12 dies aut 26-80 mm post 21 dies in obscuritate, aut 13-17 mm post 12 dies in luce obscuritateque alternanti; albae vel dilutissime brunneae; demum crassis muscosis massis conidiorum obtectae. Hyphae 1.5-3.0 µm diametro, hyalinae, laeves sine fibulis. Cellulae binucleatae. Conidiophora 220-400 µm longa, mononemata vel macronemata. Rami proxime infra septa orientes, saepe in origine attenuati, autem cito sursum curvati dilatatique. Cellulae conidiogenae terminales, holoblasticae, cylindricae vel basi leniter contractae, annellophorae percurrens 25-90 (100) x 2.5-3.4 (4.0) µm. Situs conidiogeni successivi 0.5-7.0 µm distantes. Conidia 6.5-16(-18.5)(sd=11.45 ± 2.78) X 2.5-4.0 (-4.5)(sd=3.45 ± 0.65) µm in guttas mucosas aggregata, unicellularia, hyalina, laevigata, oblonga vel breviter clavata; basibus truncatis cum fimbriis; per hyphas vel annellophora secundaria germinantia.*

Holotype: Canada, Alberta; Taylor Lake Hiking Trail, Banff National Park. Dried colonies on sterile pine twigs embedded in malt extract/yeast extract agar (MEYE) isolated from bark beetle galleries in *Pinus contorta* var. *latifolia* Engelm. bark, collected 23 September 1987, J. and B. Reid, derived from UAMH 10782.

Isotype: WIN(M)1397

Etymology: From the provincial name, Alberta.

Colonies on MEYE white to very pale brown (10YR8/2 to 8/3) with even margins in alternating light and dark at 20 °C, margins white (10YR8/2) to translucent. Dark grown colony margins often irregular due to faster growing hyphae developing short, lateral dendroid branches around the margins, but these are less pronounced in alternating light. White (10YR8/2) beneath areas of denser mycelium, but colourless elsewhere. Odour lacking or indistinct. Attaining 16-32 mm diam in 12 days or 26-80 mm in 21days in darkness, and 13-17 mm in 12 days in alternating light and dark, all at 20 °C. Hyphae hyaline, smooth-walled, 1.5-3.6 µm wide, cells binucleate, aerial hyphae collapsing in older cultures. Clamps and chlamydospores lacking. Conidiophores erect, initially arising from single hyphal elements, phalacrogenous, mononematous, semi-macronematous or macronemetous, the latter often coalescent laterally and then resembling top-shaped conidiomata with glistening apices (Figs. 2,3).

Macronematous conidiophores 220-400 µm long, with each branch narrowed at its point of emergence from immediately below a septum in the originating hypha, curving upward sharply and widening to normal size; the vast majority have a basal septum above the curve, and all are terminated by percurrently proliferating annellophores (Figs. 3,4,5); producing slimy conidial masses; medium-embedded hyphae may produce crescent-shaped conidial masses. Conidiogenous cells holoblastic, terminal, integrated, percurrent, forming conspicuous annellophores 25-150 X 2.5-3.5 (-4.0) µm; these are at

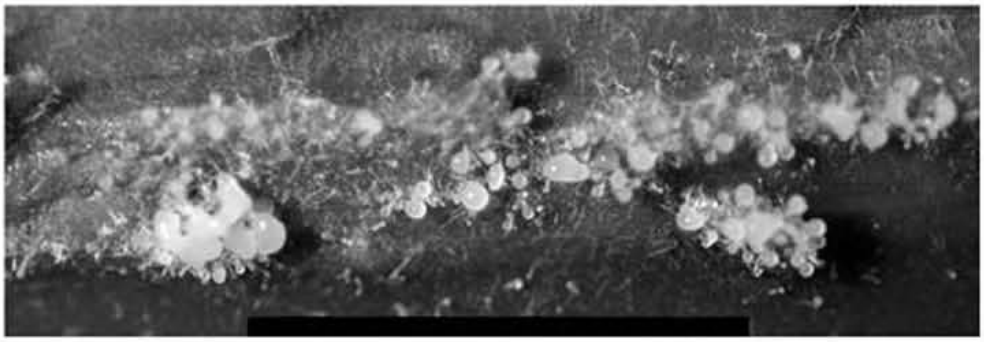

Fig. 2: Conidiomata of *Basidiopycnides albertensis* (strain: UAMH 10784 = WIN(M) 720) in various stages of development, growing on an autoclaved pine twig in agar. Bar = 10 mm.

first parallel-sided but taper gradually towards the apex, and are occasionally either slightly constricted or form slightly swollen nodular annellations at their base. ANNELLATIONS occur at intervals of 0.5-7.0 µm, with the longer intervals formed first. CONIDIA (Figs. 3,6) unicellular, hyaline, binucleate, smooth-walled, oblong to occasionally short clavate, with an obtuse apex and truncate base that may be slightly narrowed, but with a short but definite frill, 6.5-16(-18.5) (sd=11.45 ± 2.78) X 2.5-4.0(-4.5) (sd=3.45 ±0.65) µm, narrowed bases 2.5 ± 0.8 µm, produced singly, but aggregating in slimy masses in culture, germinating by germ tubes or directly produced annellophores.

TELEOMORPH: Not observed.

ADDITIONAL ISOLATES EXAMINED (PARATYPES). Same site and same date as the holotype: UAMH 10783 = WIN(M) 719 from the same tree as holotype; UAMH 10784 = WIN(M) 720, UAMH 10785 = WIN(M) 1399, and UAMH 10786 = WIN(M) 1425 were from three different trees.

Individual dark grown isolates varied slightly in growth rates and colony morphologies, but in alternating light and dark growth they were very similar. And in all cases secondary conidium production greatly increased the size of the slimy conidial masses on the surface of a colony. Importantly, the nature of the conidiomata formed in twig cultures was the same as on agar plates, e.g., size variation, branching pattern, conidium size, etc.

Occasionally conidia remain attached at the conidiogenous locus; this caused bending in the annellation zone (Fig. 3). However, this is different from the situation where seceded conidia sometimes remain adherent after secession; presumably because of the presence of mucilaginous material on either walls of the conidia or conidiogenous cell. When viewed with phase contrast optics, both conidium initials and most conidia have a halo surrounding them (Fig. 6); that may be a mucilaginous sheath.

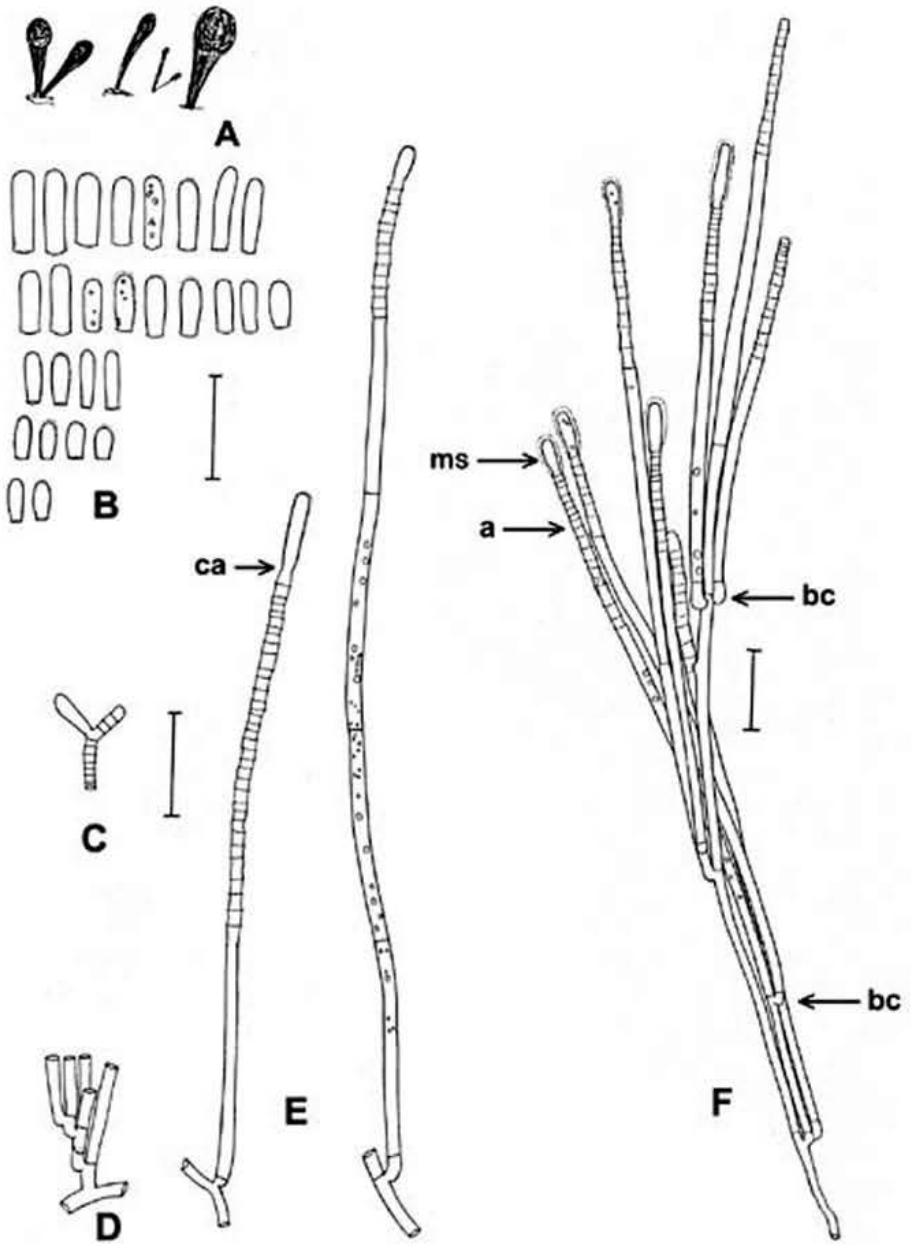


Fig. 3: Line drawings of *Basidiopycnides albertensis* (strain: UAMH 10786 = WIN(M) 1397). A. Habit sketch of individual conidiophores aggregations; not to scale. B. Conidial size variation. Some conidia have guttules and / or sheaths. Bar = 20 µm. C. An annellation zone from which a conidium was displaced in a sympodular manner. D. Basal branching of a complex conidiophore. E. Two mononematous conidiophores (annellophores), one highly guttulate. When a conidium is formed initially, it is often subtended by a slight constriction in the originating conidiogenous cells (ca). Bar for C, D, and E = 20 µm. F. Young conidioma with branched conidiophores arising from a single element; conidium with mucilaginous sheath (ms); annellation (a); branch constriction (bc). Bar = 20 µm.

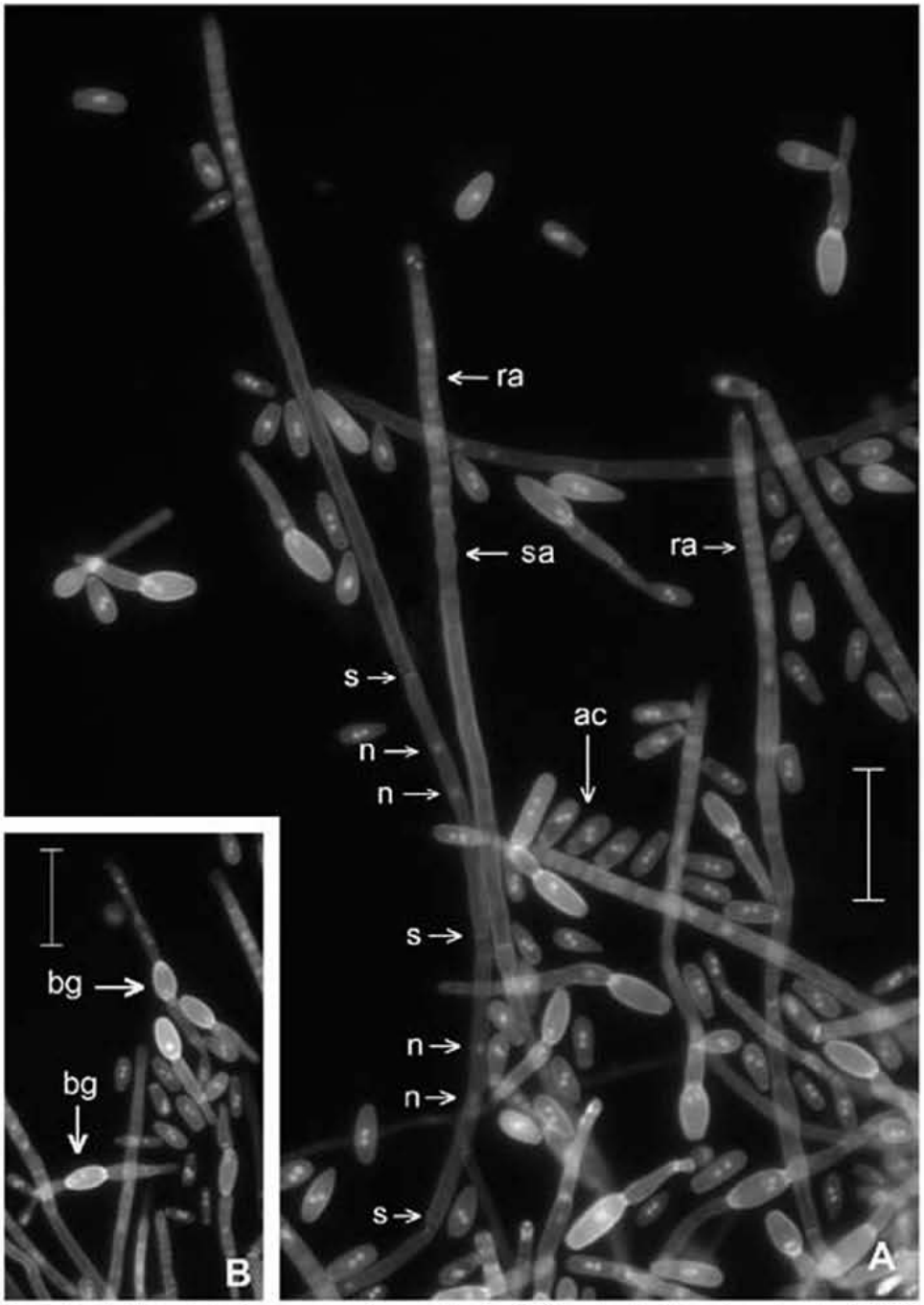


Fig. 4: An incident-light fluorescent micrograph of a DAPI-stained slide culture (strain: UAMH 10784 = WIN(M) 720]. A. Binucleate hyphal cells; septa (s) and nuclei (n). Swellings on the annellophore base (sa). Laterally displaced seceded conidia (ac) still adhering to a proliferated annellophore. Repetitively produced annellations (ra). Bar = 20 µm. B. Two bipolar germinating conidia (bg). Bar = 20 µm. In both A and B, the conidia are regularly binucleate and, on germinating, successive conidiogenous sites are often narrower.

## Discussion

None of the strains of *Basidiopycnides albertensis* ever produced basidiomata in culture, and their conidiomata superficially resemble synnemata as characterized by Seifert (1985) and Seifert & Okada (1990). However, our DAPI nuclear staining results that showed the hyphal cells and the conidia are consistently binucleate, and the DNA sequence phylogeny data combined, provide strong evidence that this species is a member of the Basidiomycota closely allied to, but not identical with, *Basidiopycnis hyalina* of the *Atractiellales*. And while morphological similarities were evident in these two species, e.g., the manner of conidiophore branching, size and shape of conidia and the latter's aggregation into slimy masses. Neither significant annellations nor pronounced annellation intervals were described or figured for *Basidiopycnis hyalina* – herein we use annellation to denote the sites where single conidia are produced, and annellation interval(s) the distance(s) between two such successive sites – but it was said to produce annellides.

In some features *Basidiopycnides albertensis* also resembles the basidiomycete *Stilbotulasnella conidiophora*. However, Bandoni & Oberwinkler (1982) state the annellations in *S. conidiophora* are inconspicuous and only clearly visible when stained, but its annellation intervals as figured are significant. In *Basidiopycnides albertensis* the annellations are seen easily in both water or KOH mounts (Fig. 5). Thus we assume that significant annellation intervals are lacking in *Basidiopycnis hyalina*, since Bandoni & Oberwinkler (1982) illustrated such for *S. conidiophora* where visualization was markedly improved in stained mounts, while Oberwinkler et al. (2006) who also used stained preparations did not record these features in *Basidiopycnis hyalina*. Nor did they record conidial germination by direct production of annellophores in *Basidiopycnis hyalina* as we found in *Basidiopycnides albertensis*; they only reported germination by production of hyphae. Indeed, the conidia of *Basidiopycnides albertensis* typically germinate by the production of either uni- or bipolar secondary annellophores. These can, and do, produce multiple secondary conidia (Figs. 4A, B). Finally, *Basidiopycnides albertensis* is only known from a single location from several *Pinus contorta* var. *latifolia* trees at a single location, while *Basidiopycnis hyalina* occurs in galleries of multiple species of bark-beetles infesting both *Picea abies* (L.) H. Karst. and *Pinus sylvestris* in Germany, Italy and Switzerland (Oberwinkler et al. 2006).

The unusually percurrently proliferating annellophores produced by *Basidiopycnides albertensis* are not unique. Similar annellophores that develop as extensions of conidiophores and conidiophore branches are reported in *Filosporella* [= *Rogersia annelidica* Shearer & J.L. Crane (1976)], but this has very different conidia. Blackwell & Malloch (1989) described the hyaline imperfect state of a *Pyxidiophora* sp. as having 200-300 µm long conidiophores

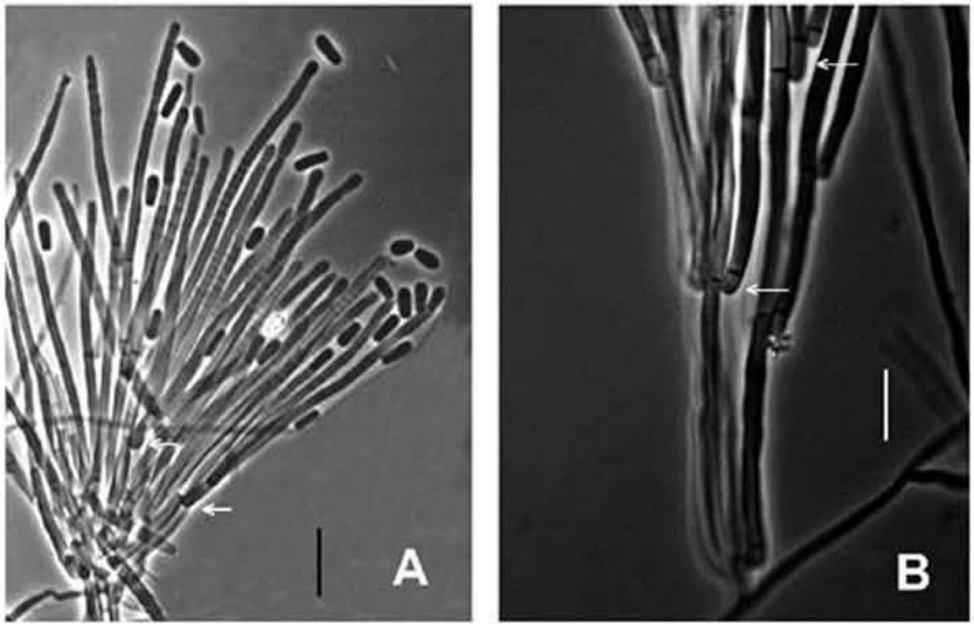


Fig. 5: Phase contrast micrographs of conidiomata (strain UAMH 10786 = WIN(M) 1397). A. An apparent aggregation of at least three conidiomata arising from at least three separate hyphal elements (see base). Note the constricted branch bases (arrows), and the very regular intervals between successive annellations. Bar = 20 µm. B. The base of a conidioma arising from a parent hyphal element shown in two different focal planes. The constricted bases of branching elements can be clearly seen, even in successive branches (arrows). Note on the right the "three level branching pattern of the conidiophore" sensu Jacobs et al. (2003, Fig 12); on the left the conidioma is derived from a single branch arising from a vegetative hyphal element. Bar = 10 µm.

and percurrently proliferating conidiogenous cells; this too was ruled out after examination of a prepared slide loaned by Dr. Malloch. Indeed, such annellophores have been reported in a wide variety of fungi (Glawe 1989), sometimes being produced concurrently with sympodially proliferated conidiogenous cells e.g. in *Diatrype albopruinosa* (Schwein.) Cooke (Glawe & Rogers 1982).

*Basidiopycnides albertensis* strains resemble superficially species of *Graphium* and *Pesotum* (Seifert & Okada 1993, Okada et al. 1998b) but differ in the branching nature in the conidiomata of *Basidiopycnides albertensis*; lengths of both annellophores and conidiation site intervals; the presence of only slight swelling of some of the basal annellation intervals in the annellophores (Fig. 5), rather than the significant nodulation found in *Graphium* spp.; and the conidiogenous cells that are markedly different to those found in *Pesotum* spp. But the basal branching in *Basidiopycnides albertensis* is strikingly similar to the "Three-level branching pattern of the conidiophore" that illustrates how conidiophores are produced in the capitulum of *Graphium laricis* K. Jacobs et

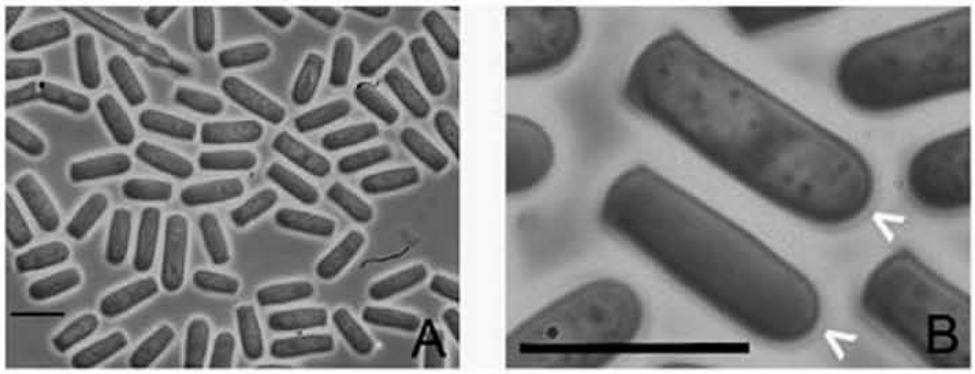


Fig. 6: Phase contrast micrographs of conidia (strain UAMH 10786 = WIN(M)1397). A. Oblong conidia with truncated basal frill [compare with Jacobs et al. 2003, Fig 10]. Bar = 10 µm. B. Conidia. The presumed sheath is indicated by arrows. Bar = 10 µm.

al. (2003, Fig. 12), except in the latter the branches do not have constricted bases at their points of origin.

When first reported in the literature (Bauer et al. 2006, pp. 43, 57-58, Fig. 1), *Basidiopycnis hyalina* was treated as one of two new *Atractiellomycetes* with "pycnidial basidiocarps", and only designated by a herbarium number in the discussion in that paper. Later, only *Basidiopycnis hyalina* was confirmed to produce a "pycnidial basidiocarp", i.e., a structure resembling a pycnidium in which basidia and basidiospores are produced; the second species, *Proceropycnis pinicola*, was treated as a new anamorphic fungus (Oberwinkler et al. 2006). However, the latter authors reported that the basidiomata of *Basidiopycnis hyalina* were only produced when strains were paired with other fungi, but not in collections from host material.

We never found basidiomata of *Basidiopycnides albertensis* either on host material or under any culture conditions.

Oberwinkler et al. (2006, p. 644) did not propose an anamorph genus for their asexual phase of *Basidiopycnis hyalina*, stating "We were not able to find morphological characteristics sufficient for separating these basidiomycetes anamorphs on a generic level and, therefore, do not propose a new genus for the anamorph of *B. hyalina*." In support of this they refer specifically to *Stilbotulasnella* Oberw. & Bandoni, a monotypic genus based on *S. conidiophora*, and two species of *Pistillaria* Fr. (Koski & Perrin 1971); the latter two are now considered species of *Typhula*: *T. micans* (Pers.) Berthier and *T. setipes* (Grev.) Berthier (Berthier, 1976). But an anamorph genus is required because criteria other than conidiomatal structure separate organisms such as *S. conidiophora* and *T. micans* from *B. hyalina*. For example, *Basidiopycnis hyalina* has simple septate pores surrounded by atractosomes (Oberwinkler et al. 2006), while *S. conidiophora* has dolipore septa, lacking parenthosomes. We did not examine

pore structure in *Basidiopycnides albertensis*. And both *T. setipes* and *T. micans* have clamp connections (Koski & Perrin 1971), but *Basidiopycnis hyalina* (Oberwinkler et al. 2006), *S. conidiophora* and *Basidiopycnides albertensis* do not.

These factors, plus the fact that *Basidiopycnides albertensis* did not produce a teleomorph state, made it difficult to determine the various relationships. But it suggests strongly that relationships should not be assumed based simply on the production of presumptive "enteroblastic annelloconidia". It is doubtful *Graphium* spp. sensu Okada et al. (2000) are closely related to the organisms discussed above, but the method of conidial production in species such as *Graphium laricis* (Jacobs et al. 2003), *Graphium penicillioides* Corda (Okada et al. 2000) or *Remersonia thermophila* (Fergus) Seifert & Samson (Seifert et al. 1997) are superficially strikingly similar to that of *Basidiopycnis hyalina*, *Basidiopycnides albertensis* and *S. conidiophora*. It is also highly unlikely that *Graphium* spp. consistently produce binucleate conidia as observed in *Basidiopycnides albertensis*. Probably, the superficial similarity in conidiogenesis observed in the genera/species listed above simply reflects convergent evolution of a similar, but not identical, characteristic that has been selected for in different groups of unrelated fungi. The production of large masses of sticky spores on upward pointing conidiophores would facilitate their being brushed onto the bodies of insects, and thus favor their dissemination by insects. With the inclusion of sequence data along with morphological criteria, *Basidiopycnides albertensis* can be distinguished from "*Graphium*-like" anamorphs that evolved within both ascomycetous and basidiomycetous lineages. In order to facilitate a better understanding of the biology of these organisms we need to designate a phylogenetic classification. It is likely that more "*Graphium*-like" fungi will be recovered from bark beetle galleries and these organisms should be placed into appropriate genera.

Throughout we have used the term annellophore in reference to conidiogenesis in *Basidiopycnides albertensis*, but this is contrary to the recommendation by Kendrick (1971, p. 261) that the term annellophore should be replaced with annellide to mean annellated conidiogenous cell. However, annellated was defined in that same Proceedings chapter (Kendrick 1971, p. 254) as "condition of a conidiogenous cell which has undergone a number of **very short** percurrent vegetative proliferations, each of which terminated in the production of a single holoblastic conidium. Each --- a **narrow band** of wall material encircling the conidiogenous cell, is called an annellation." (The bold usage is ours). Although subsequently many mycologists have followed this recommendation, we do not feel the term annellide, as defined above, is appropriate for the conidiogenous structure seen in *Basidiopycnides albertensis*. And problems with this general usage of the term annellide have been noted before.

Hammill (1972), in one of a series of papers on conidiogenesis in fungi based on transmission electron microscopy studies, drew attention to an unusual form of such in *Monotosporella sphaerocephala* (Berk. & Broome) S. Hughes [= *Acrogenospora sphaerocephala* (Berk. & Broome) M.B. Ellis (1971)]. Here, the subsequent unusual pattern of conidium maturation aside, initial development involved percurrent growth of intraconidiophore hyphae through a previously formed conidium-delimiting septum.

Cole & Samson (1979) using scanning electron microscopy confirmed, and expanded on, Hammill's work and concluded, page 78, "--- it is probable that several genera of annellated Deuteromycetes develop in this manner". But what is significant, from our point of view, is that the annellations on the conidiophore are produced at significant intervals, not very short percurrent vegetative proliferations (see above).

The term annellide was defined by Sutton (1980, p. 642) as "a holoblastic percurrently proliferating conidiogenous cell", a more succinct but nonetheless accurate rendering of Kendrick's (1971, p. 258) definition. And an example of the confusion that existed at that time is to be seen in Bandoni & Oberwinkler (1982, p. 1879) who were unsure as to the true nature of the conidiogenous cells in *S. conidiophora* and wrote "Although we have used the term annellide in reference to the conidiogenous cells, it remains to be demonstrated they are homologs of structures so designated in ascomycetes and hyphomycetes". Then, in a progressive step, Sutton and his colleagues (Minter et al. 1983, pp. 117-118) argued that at least two sets of developmental processes were being defined by the term annellide. This view was supported by both Wang (1990) and Kiffer & Morelet (2000), who both resurrected and defined their concept of an annellophore and redefined an annellide. We concur an annellophore is really an annellated conidiophore; see Wang (1990, pp. 53-56) for a precise description of the postulated process, and that annellophore is the appropriate term to describe conidiogenesis in *Basidiopycnides albertensis*, and quite possibly fungi such as *S. conidiophora* as well. It is probable that transmission electron microscopic pictures will show that the two latter species develop annellophores in a manner consistent with Wang's (1990) theory and, as a consequence, the term annellophore should be reinstated.

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# A new species of *Candelabrochaete* (*Polyporales, Basidiomycota*)

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**Abstract** — A new species of *Candelabrochaete* (*Corticiaceae, Polyporales, Basidiomycota*) is described and illustrated from specimens collected in two islands of the Macaronesian region, Faial (Azores) and Madeira. Affinities with other close species as *C. africana, C. langloisii* and *C. septocystidia* are discussed

**Key words** — *Aphyllophorales*, Macaronesia

## Introduction

The genus *Candelabrochaete* was described by Boidin (1970) to include species characterized by a resupinate, adnate, smooth or tuberculate basidioma, brittle when dry; with a monomitic hyphal structure, consisting of wide, short, clampless hyphae, branched at right angles; with projecting septocystidia, subcylindrical to suburniform basidia, sometimes with linear repetition, and spores oblong to ellipsoid, thin-walled, smooth, inamyloid, acyanophilous.

Actually the genus comprises ten species with a worldwide distribution that are differentiated primarily based on basidiospore morphology (Table 1): subglobose in *C. dispar* Hjortstam & Ryvarden (Hjortstam & Ryvarden 1986); ellipsoid in *C. adnata* Hjortstam (Hjortstam 1995), *C. cirrata* Hjortstam & Ryvarden (Hjortstam & Ryvarden 1986), *C. eruciformis* (G. Cunn.) Stalpers & P. K. Buchanan (Stalpers & Buchanan 1991), *C. langloisii* (Pat.) Boidin (Boidin 1970), *C. magnahypha* (Burt) Burds. (Burdsall 1984), *C. simulans* Hjortstam

(Hjortstam 1995) and *C. verruculosa* Hjortstam (Hjortstam 1983); cylindrical to subcylincrical in *C. africana* Boidin (Boidin 1970); and allantoid in *C. septocystidia* (Burt) Burds. (Eriksson & al. 1978). The Macaronesian material has spores that are reniform to ellipsoid.

Roberts (2000) transferred *Peniophora mexicana* Burt to *Candelabrochaete,* making the new combination *C. mexicana* (Burt) P. Roberts without studying the type material. According to Burdsall (1984), *P. mexicana* has thick-walled spores and pseudocystidia, characteristics suggesting a relationship with the genera *Phanerochaete* P. Karst., *Candelabrochaete,* and *Hypochnicium* J. Erikss. We think that further studies are necessary to ascertain the correct position of this taxon and whether it should or should not be included in *Candelabrochaete.*

## Materials and methods

A total of 12 specimens were collected during mycological forays to two islands of the Macaronesian region, Faial (belonging to the central group of the Azores archipelago) at the end of the winter of 2005 and Madeira during the summer of 2000 and autumn of 2006.

The material was studied following classical methods for the corticiaceous fungi: thin, freehand sections from each specimen were mounted in KOH (5%) and/or Melzer's reagent. These sections were examined under an Olympus BH50 microscope. M. Dueñas prepared line drawings with the aid of a Leica camera lucida. Color names are from the ISCC-NBS Color Names (Kelly & Judd 1976). Abbreviations include MD (M. Dueñas), Tell. (M.T. Telleria), and TFC (University of La Laguna Herbarium, where the E. Beltrán & J.L. Rodríguez-Armas collections are deposited).

## Taxonomy

***Candelabrochaete macaronesica*** M. Dueñas, Telleria & Melo, **sp. nov.** FIGS. 1 & 2
MYCOBANK MB 511289

*Fructificatio resupinata, laeviter ad substratum unita, lutea-aurantiaca, cum margine fibrata, clariore. Systema monomiticum hypharum, fibulae rarissimae; septocystidia usque ad 150 × 8-11 µm; basidia quattuor sporarum, subcylindricorum ad subclavata (12-) 16-18.5 × 5-5.5 µm; sporae subcilyndricis ad anguste ellipticae, 5-6.5 × 2-3 µm. Holotypus MA-Fungi 72924.*

HOLOTYPE: **PORTUGAL, AZORES, Faial,** Horta, Ponta do Varadouro, 26SLH4570, 42 m, on *Picconia azorica,* 21 Feb 2005, M. Dueñas, 10467MD, MA Fungi 72924.

ETYMOLOGY: from the Macaronesian region

KEY CHARACTERS — Fructification resupinate, adnate, yellow to orange yellowish, margin differentiated, fibrillose; hyphal system monomitic, basal hyphae slightly thick-walled, with scattered clamps, forming a thick subiculum

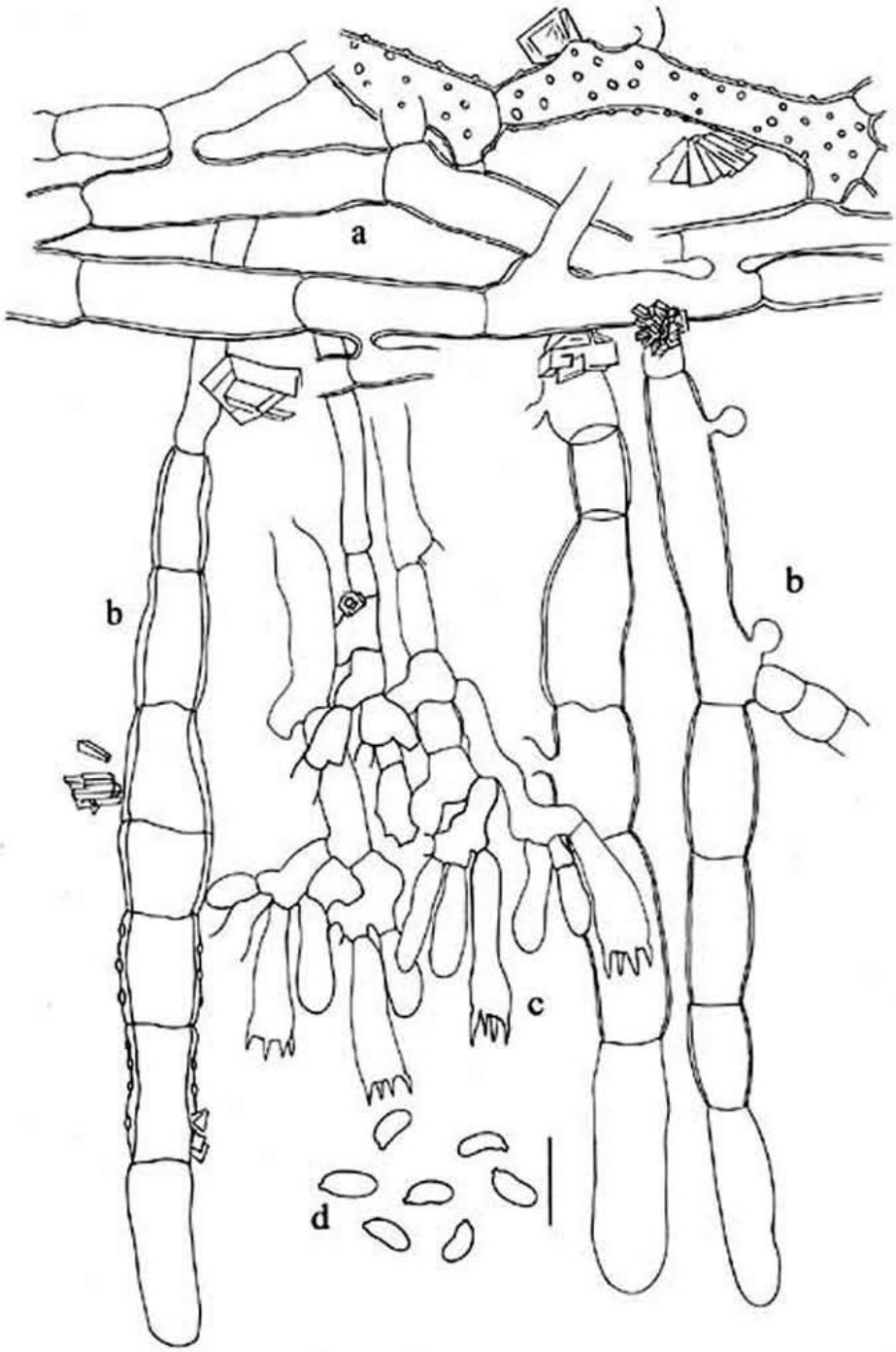


FIG. 1. *Candelabrochaete macaronesica*, MA-Fungi 72924 (holotype).
a: basal hyphae; b: septocystidia; c: basidia; d: basidiospores.
(Bar = 10 µm).

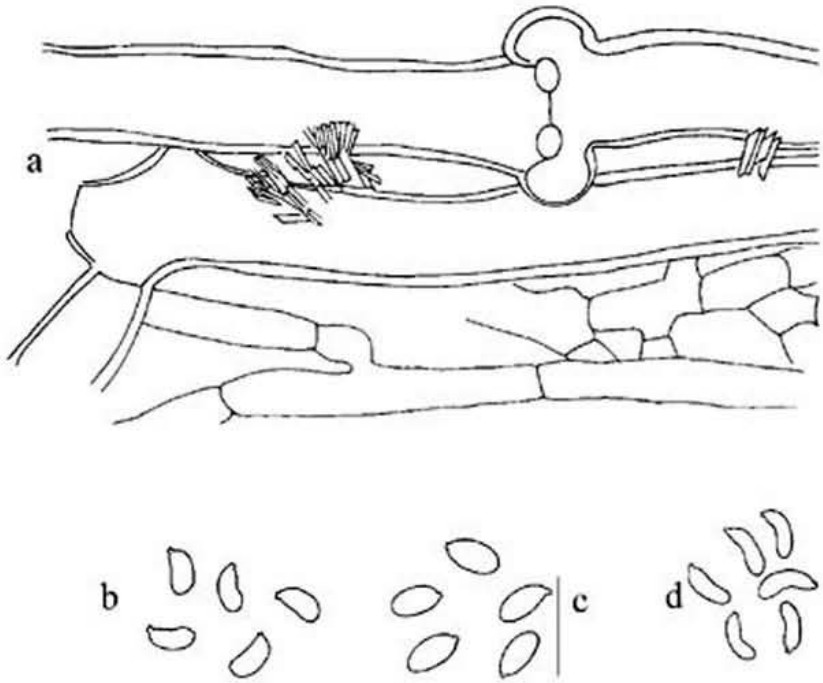


FIG. 2. *Candelabrochaete macaronesica*, MA-Fungi 72925, a: hyphae; b: basidiospores. *Candelabrochaete africana*, LY 5495 (holotype), c: basidiospores. *Candelabrochaete septocystidia*, MA-Fungi 7168, d: spores. (Bar = 10 µm).

visible under the lens; septocystidia up to 150 × 8-11 µm; basidia subcylindrical to subclavate (12-)16-18.5 × 5-5.5 µm; basidiospores cylindrical to narrowly ellipsoid, adaxially concave, 5-6.5 × 2-3 µm.

BASIDIOME resupinate, adnate, up to 2 mm thick, pellicular to membranaceous, yellow/orange-yellowish [70.I.OY/71.m.OY], pink in contact with KOH (5%); SUBICULUM lighter; HYMENOPHORE mainly smooth, under the lens pilose from the projecting cystidia; MARGIN differentiated, fibrillose, white.

HYPHAL SYSTEM monomitic; basal hyphae slightly thick-walled, 4-10 µm wide, short celled, sometimes with attached organic material giving them an irregular aspect, as a rule with simple septa — but specimen 10881MD with scattered clamps (Fig. 2) — , loosely interwoven, forming a thick subiculum visible under the lens; subhymenial hyphae thin-walled, 4-6 µm wide, more densely interwoven. SEPTOCYSTIDIA cylindrical, arising from the basal hyphae and projecting through the hymenium, up to 150 × 8-11 µm, with barrel shaped cells, thin to slightly thick-walled, sometimes ramified; especially the basal hyphae and cystidia often provided with a cover of yellowish crystalline matter.

BASIDIA without linear repetition, subcylindrical to subclavate, (12-)16-18.5 × 5-5.5 µm, with 4 sterigmata. SPORES cylindrical to narrowly ellipsoid, adaxially concave, thin-walled, smooth, hyaline, 5-6.5 × 2-3 µm, non-amyloid, non-cyanophilous.

ECOLOGY AND DISTRIBUTION — This new species has been collected from two islands, Faial (Azores) and Madeira, fruiting on decayed wood of *Picconia azorica* and *Occotea foetens*.

ADDITIONAL MATERIAL STUDIED (Paratypes) — **PORTUGAL. AZORES: Faial,** HORTA, PONTA DO VARADOURO, 26SLH4570, 42 m, on decayed wood, 21 Feb 2005, 16059Tell., MA Fungi 72929; 16061Tell., MA Fungi 72932; 16063Tell., MA Fungi 72926; on *Picconia azorica*, 10466MD, MA Fungi 72927; 10470 MD, MA Fungi 72933; 10473MD, MA Fungi 72928; 15182TFCMic; 15187TFCMic; 15193TFCMic. **MADEIRA: Santana,** FAJÃ DA NOGUEIRA, 28SCB2325, 850 m, on *Ocotea foetens*, 26 Jun 2000, 14108 Tell., MA Fungi 72934; **Santana,** FAJÃ DA NOGUEIRA, MONTADO DO LEACKOK, 28SCB2024, 850 m, on trunk of *Ocotea foetens*, 19 Oct 2006, 10880MD, MA Fungi 72930; 10881MD, MA Fungi 72925; 10884bisMD, MA Fungi 72931 [together with *Hyphoderma argillaceum*].

COMMENTS *C. macaronesica* resembles *C. langloisii* in the colour changing to pink in the presence of KOH. However, in the last species, the basidiospores in *C. langloisii* are also laterally depressed but are larger, 7-9.5 × 3-4 µm (Boidin 1970).

The presence of clamps on basal hyphae is not a frequent character in *Candelabrochaete*; however, *C. magnahypha*, described from Florida, has basal hyphae with scattered clamps (Burdsall 1984) but with ellipsoid and larger (6-7(-8) × 3-4 µm) spores.

The spores of this new species are similar in size to those of *C. verruculosa* and *C. africana*, but the former has grandinioid to odontoid hymenophores while the latter differs in its subcylindrical, laterally convex spores. Furthermore, *C. africana* has thick-walled hyphae and cystidia and basidia present linear repetition, as also noted for *C. verruculosa*. *C. septocystidia*, which was also collected from the same locality in the Azores, differs from *C. macaronesica* in the allantoid shaped and smaller sized (4-5 × 1.5-2 µm) spores (FIG. 2).

## Acknowledgments

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TABLE 1. Diagnostic characters for described species of *Candelabrochaete*

| SPECIES | HYMENOPHORE | COLOUR CHANGE IN KOH | CYSTIDIA SIZE (µm) | BASIDIA SIZE (µm) | SPORES | |
|---|---|---|---|---|---|---|
| | | | | | SHAPE | SIZE (µm) |
| *C. adnata* | smooth | no | 40-60 × 6-8 | 17-20 × 4-5 | ellipsoid | 6.5-7 (-8) × 2.8-3.5 |
| *C. africana* | smooth | no | up to 90 × 10 | 13-17 × 5-6 | subcylindrical | 4.5-6 × 2-3 |
| *C. cirrata* | tuberculate | no | up to 40 × 10 | 17 × 6-7 | ellipsoid | 6-6.5 × 4-5 |
| *C. dispar* | grandinioid to odontioid | no | 80-120 (-200) × 6-9 (-10) | (12-)15-17 (-25) × 4-5 (-6) | subglobose | 5-5.5 × 4-4.5 |
| *C. eruciformis* | smooth to tuberculate | no | 40-90 × 6-10 | 24-30 × 5-6 | ellipsoid | 7-9 × 3-5 |
| *C. langloisii* | smooth | yes | 75-200 × 9-15(-21) | 15-18 × 6.5-8 | ellipsoid | 7-9.5 × 3-4 |
| *C. magnahypha* | smooth | no | 100-150 × 9-12 | 12-15 × 5.5-6 | ellipsoid | 6-7 (-8) × 3-4 |
| *C. mexicana* | continuous to pubescent | no | 90-150 × 9-20 | 9-11.5 × 6.5-7 | ellipsoid to broadly ellipsoid | 7-10 (-2) × (4-) 5-6 |
| *C. septocystidia* | smooth | no | 60-150 × 5-9(-12) | 12-18 × 4.5-6.5 | allantoid | 4.5-6.5 × 1.5-2 |
| *C. simulans* | smooth | no | 100-250 | 20-25 × 4-5.5 | ellipsoid | 4.5-5.5 × 3-3.2 |
| *C. verruculosa* | grandinioid to odontioid | no | 70-140 × 8-12 | 15-20 × 4.5-5 | ellipsoid | 5.5-6 × 3-3.25 |
| *C. macaronesica* | smooth | yes | 70-150 × 8-11 | (12-)16-18.5 × 5-5.5 | subcylindrical to narrowly ellipsoid | 5-6.5 × 2-3 |

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# Nomenclatural notes. 12. Untangling Hedwig's *Octospora villosa*: *Helvella fibrosa* comb. nov.

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**Abstract** — *Octospora villosa* was described and illustrated by Hedwig in 1789, and has been reassigned first as a variety in *Peziza*, and later at species rank in *Fuckelina*, *Cyathipodia*, and *Helvella*. Hedwig's species was sanctioned (at varietal level) by Fries, who treated it as *Peziza macropus* [var.] β *hirta*, an unintentional error in attributing to this taxon the epithet of a different Hedwig species. Error upon error has plagued Hedwig's species name, and an attempt here is made to untangle the most flagrant of these. Names in current use are *Helvella villosa*, *H. chinensis*, and *H. dissingii*. None turns out to be correct under the International Code of Botanical Nomenclature. Overlooked by all recent authors is the species name *Peziza fibrosa* proposed by Wallroth in 1833 as a new name to be substituted for *O. villosa* since that epithet could not be transferred to the genus *Peziza*, it being preoccupied there by Persoon's *P. villosa* of 1801. The new combination *Helvella fibrosa* is formally proposed, and an epitype specimen for *O. villosa* is designated.

**Keywords** — homonyms, sanctioned names, errors, corrected synonymy

## *Octospora villosa* Hedwig

One of the most distinctive and beautiful plates of a *Helvella* is that provided by Hedwig (1789) as Tab. 19, B. It is an unmistakable species, closely resembling *Helvella macropus* (Pers.) P. Karst. but differing markedly in its dark hymenial color. Probably the best description of Hedwig's species is that by Dissing & Nannfeldt (1966) in which they proposed the new combination *Helvella villosa* (Hedw. ex Kuntze) Dissing & Nannf., which of course they could have cited as *H. villosa* (Kuntze) Dissing & Nannf. At the time they proposed this name, the 1961 edition of the International Code of Botanical Nomenclature (ICBN) still had as a starting point for *"fungi caeteri"* Fries's Systema Mycologicum (1821-32). This explains how "Kuntze" was part of their citation of authors. *O. villosa* was a pre-starting-point name, and apparently the first author to transfer *O.*

*villosa* to another genus was Kuntze (1891), who mentioned it as a species of his new genus *Fuckelina* Kuntze [itself an invalid later generic homonym of *Fuckelina* Sacc. (Saccardo, 1875)]. There are at least two earlier uses of the name *Helvella villosa* (some with the alternative generic name orthography of "*Elvela*") for other fungi, but these, too, were pre-starting-point names and did not constitute valid competing names under the 1961 ICBN. With the change of starting points for all fungi in the 1981 ICBN to a starting point of Linnaeus's *Species Plantarum* (1753), the Dissing & Nannfeldt combination became a later homonym, and thus illegitimate.

A major error was mine (Korf, 1988a), when in an attempt to provide a correct name in *Helvella* for Hedwig's species I proposed that it be called *Helvella dissingii* Korf. On publication of this name honoring my dear colleague, Henry Dissing, I was advised by him in an e-mail of a paper I had overlooked, Lundell et al. (1985), wherein Nannfeldt & Holm had earlier noted that *H. villosa* (Kuntze) Dissing & Nannf. was now a later homonym. They decided that the earliest available name for Hedwig's species was that of a species described by Velenovský (1939) as *Macropodia chinensis* Velen. They therefore proposed a new combination, *Helvella chinensis* (Velen.) Nannf. & L. Holm. I immediately pointed out my error (Korf, 1988b) in the errata page of a revision of my chapter in volume 5 of the reprint edition of Boudier's Icones Mycologicae. At the time of writing this paper (December 2007), the Index Fungorum website <www.indexfungorum.org/names/names.asp> erroneously still lists *H. dissingii* as the correct name for this taxon. As pointed out below, the choice of *H. chinensis* as the correct name was also an error.

## *Peziza fibrosa*, a long forgotten obligate synonym

No recent author working with *O. villosa* seems to have realized that the name *Peziza fibrosa* Wallr. (Wallroth, 1833) was not proposed as a new species, but rather as a new name for *Octopsora villosa* Hedw., which was carefully cited as its synonym, including a reference to Hedwig's plate 19. Wallroth could not transfer Hedwig's species to *Peziza* because it would have become a later homonym of *Peziza villosa* Pers. published in 1801. Wallroth's name is typified by Hedwig's plate, the same illustration that serves as the lectotype of *O. villosa* itself and all the names based thereon. The name *P. fibrosa* was accepted by Kickx (1867), who provided additional information on specimens from Flanders. It was later transferred to *Macropodia* by Saccardo (1889), who also included the reference to Hedwig's name in the synonymy and to Kickx's treatment. Strangely, Nannfeldt, as well as Dissing, I, and all other recent authors, seem never to have taken *P. fibrosa* or *M. fibrosa* (Wallr.) Sacc. into account, despite the latter being a frequently cited synonym of Hedwig's *O. villosa*. Wallroth's species apparently

provides the earliest epithet available for the species when treated in *Helvella*. The synonymy was clearly accepted by Kickx (1867), Saccardo (1889), Kuntze (1891), Mussat (1901), and Boudier (1907).

## A chronicle of the major errors in date order

### Fries, Systema Mycologicum (1821-32)

When Fries established his variety *Peziza macropus* var. *hirta* (Syst. mycol. 2: 57. 1822), he cited it as based upon "Octosp. hirta *Hedw. Musc. fr. 2. p. 54, t. 19. f. B.*" His major error was that he confused two Hedwig species names, *Octospora villosa* Hedw., which appears on that page and plate, and *O. hirta* Hedw., which appears elsewhere and was treated by Fries as a synonym of *Peziza scutellata*. Fries later recognized his error, and corrected the citations in his index (Syst. Mycol. 3 (index): 124. 1832) to indicate that *O. villosa* was the correct basionym for the variety. He did not, however, change the varietal epithet to "villosa," which surely would have been his decision ten years earlier had he not made the name error.

### Streintz, Nomenclator Fungorum (1862)

This normally useful and quite exhaustive compendium of early names failed to record *Peziza fibrosa* Wallr., although other species from that work are cited.

### Saccardo, Sylloge Fungorum 8 (1889)

Since Hedwig's *O. villosa* is not preoccupied in *Macropodia*, there was no reason to accept Wallroth's name in making the combination *Macropodia fibrosa*. Saccardo should have proposed a new combination based on Hedwig's name, not Wallroth's name. Both Saccardo and Mussat (1901) assumed Fries's (sanctioned) *P. macropus* [var.] β *hirta* (Hedw.) Fr. was a synonym of Persoon's earlier treatments of a homonymous *P. macropus* [var.] β *hirta* Pers., which Fries did not mention or sanction.

### Kuntze, Revisio Genera Plantarum 2: 852. (1891)

This author proposed a new genus, *Fuckelina*, including Hedwig's species as *F. villosa* (Hedw.) Kuntze, one of several new combinations, unaware that there was an earlier generic name *Fuckelina* (Saccardo 1875).

### Arnould, Bull. Soc. Mycol. France 14:111. (1893)

This paper includes the new combination *Leptopodia villosa* (Schaeff.) Boud., an error in which *Helvella villosa* Schaeff. was incorrectly assumed to be an earlier synonym of *Octospora villosa* Hedw. Schaeffer's name is unquestionably that of another species. This combination is thus not a synonym of *O. villosa*, but

a misapplication. Arnould was in contact with Boudier, and knew of Boudier's (1885) treatment of the *Discomycetes,* and of the generic name *Leptopodia* Boud. which had been proposed there. But whether Boudier approved of the transfer to *Leptopodia* or of the change from Hedwig to Schaeffer as the author of the epithet is unkown.

**Boudier, Liste préliminaire (1904)**

The generic name *Cyathipodia* Boud. is generally treated as having been published in his Histoire et Classification des Discomycètes d'Europe (Boudier 1907), but three species were combined under this generic name by Boudier in this 1904 document, *C. villosa* (Hedw.) Boud., *C. dupainii* (Boud.) Boud., and *C. corium* (O. Webereb.) Boud. No generic description appears there. The generic name *Cyathipodia* appears not to have been formally proposed prior to Boudier's Histoire (Boudier 1907), so that none of these three combinations can be considered as validly published in 1904.

**Lundell, Nannfeldt & Holm, Publ. Herb. Univ. Upsal. 18 (1985)**

The choice of *Helvella chinensis* (Velen.) Nannf. & L. Holm as the correct name was not an error under the 1981 ICBN, but the authors' ignorance of the nomenclatural status and availability of *Peziza fibrosa* Wallr. was an error. Their assumption that the earliest available synonym was *Macropodia chinensis* Velen., not published until 1939, seems in retrospect to have been naïve. Any earlier taxonomic synonym found would automatically have reduced their combination to synonymy. *Peziza fibrosa* was published over a century earlier.

**Korf, in Boudier's Icones Mycologicae (rev. edition) 5: 215. (1986)**

In this work I cited the correct name as "*Helvella villosa* (Hedw. : Fr.) Diss. & Nannf." and a synonym as "*Cyathipodia villosa* (Hedw. : Fr.) Boud." These formulations were used to indicate that Fries was the sanctioning author. My reasoning was that since Fries had listed *Octospora villosa* Hedw. in the index to volume 3 of the sanctioning work, he had clearly accepted and thus sanctioned the name. I should not, however, have used the formulation ": Fr." since Hedwig's name was only sanctioned at varietal rank, not at species rank. It is incontrovertible that Fries did accept *Octospora villosa* Hedw. as the basionym for his treatment of *Peziza macropus* var. *hirta* when he corrected his original mistake in the Index. The epithet "*hirta*" was and is clearly a *lapsus calami* that was never changed to "*villosa.*"

**Korf. Mycotaxon 31: 381. (1988a)**

Here my errors were twofold — first, in not being aware of the paper by Lundell et al. (1985) and thus in proposing the superfluous new name *Helvella*

*dissingii* as mentioned above and secondly, of course, in being equally unaware of the nomenclatural status of *Peziza fibrosa*.

**Korf, Chapter reprint. Errata p. 48 (1988b)**

In this document I accepted the name proposed by Lundell et al. (1985), which I should not have done had I been aware of Wallroth's valid epithet.

## The corrected synonymy

***Helvella fibrosa*** (Wallr.) Korf, **comb. nov.**

MycoBank: MB 511344

= *Peziza fibrosa* Wallr., Fl. Crypt. Germ. 2: 498. 1833 (basionym), [Nomen novum, typus: *Octospora villosa* Hedw. *non Peziza villosa* Pers., Syn. meth. fung. 2: 655. 1801.]

= *Octospora villosa* Hedw., Descr. micro-anal. musc. p. 54, t. 19. fig. B. 1789. Epitype (selected here): Lundell, Nannfeldt & Holm, Fungi exsiccati Suecici praesertim Upsaliensis # 3262. 1985. (UPS).

= *Peziza macropus* [var.] β *hirta* (Hedw. : Fr.) Fr., Syst. Mycol. 2: 57. 1822. [Basionym *O. villosa* Hedw. Epithet *"hirta"* was an unintentional 1822 error (acknowledged 1832, Index p. 124) for *"villosa."*]

= *Macropodia fibrosa* (Wallr.) Sacc., Syll. Fung. 8: 160. 1889.

= *Fuckelina villosa* (Hedw.) Kuntze, Rev. Gen. Pl. 2: 852. 1891.

= *Cyathipodia villosa* (Hedw.) Boud., Hist. classific. discomyc. Europe p. 39, 1907.

= *Helvella villosa* (Hedw.) Dissing & Nannf., Svensk Bot. Tidskr. 60: 330. 1966. [ut *H. villosa* (Hedw. ex Kuntze) Dissing & Nannf.], later homonym, non *H. villosa* Schaeff. 1774.

= *Helvella dissingii* Korf, Mycotaxon 31: 381. 1988, superfluous name.

= *Macropodia chinensis* Velen., Novitat. Mycol. p. 200. 1939.

= *Helvella chinensis* (Velen.) Nannf. & L. Holm, in Lundell, Nannfeldt & Holm, Publ. Herb. Univ. Upsal. 18: 5. 1985.

**Misapplication:** *Leptopodia villosa* (Schaeff.) Boud., in Arnould, Bull. Soc. Mycol. France 9: 111. 1893.

= *Helvella villosa* Schaeff., Fung. Bavar. Palat. nasc. 4: 114. 1774.

### Acknowledgments

I wish to thank Drs. Shaun Pennycook (Mycotaxon's Nomenclature Editor) and Wen-ying Zhuang for acting as presubmission reviewers, and Dr. Pennycook especially for initially calling this problem to my attention and for his stimulating correspondence in regard to various provisions of the ICBN and for his many corrections of my text.

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MYCOTAXON

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# *Camarotella brasiliensis* sp. nov. (*Phyllachoraceae*) on *Syagrus schizophylla* (*Arecaceae*) from Brazil

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**Abstract** — A new *Camarotella* species found in the States of Bahía and Sergipe, Brazil, on an arecaceous host (*Syagrus schizophylla*) is here described. The new species, *C. brasiliensis*, is distinguished from the other congeneric species by ascospore size, a persistent mucous sheath surrounding the ascospores, and ascus shape and width.

**Key words** — neotropical fungi, *Phyllachorales*, palm, ascomycetes

## Introduction

*Arecaceae* are plants of pantropical distribution with major diversity in Brazil (Alves & Demattê 1997). Hyde et al. (1997) listed 1,580 fungal species on palms, including 650 ascomycetes, 270 basidiomycetes, 400 hyphomycetes and 260 coelomycetes. In Brazil a total of 92 fungal species were recorded on arecaceous species by Mendes et al. (1999). Among them the ascomycetes predominated with 46 species. Sanchez et al. (2003) issued a preliminary report of a new species of *Camarotella* Theiss. & Syd. on *Syagrus schizophylla* (Mart.) Glassman (*Arecaceae*) from the state of Sergipe, collected in 1997. In the course of preparing this paper, however, the authors discovered that the same fungus had been found earlier in 1986, in Valença, Bahía. This new species is now described and named.

## Materials and methods

Specimens of *Syagrus schizophylla* collected in Bahía were incorporated into the CEPEC Herbarium in Itabuna, Bahía, and those from the coastal area

of Sergipe, Brazil deposited in the Mycological Collection of the Herbarium of the University of Brasília (UB- Col. Micol.). Observations under the stereomicroscope were followed by study of squash preparations and stromatic sections made with a freezing microtome. Morphological features were described, measured, and photographed using a Zeiss-Axiophot E microscope. In most cases, the samples were stained with lacto-glycerol cotton blue and the slides sealed with nail polish. A minimum of fifty replicates were made of each spore dimension.

## Taxonomy

***Camarotella brasiliensis*** C.A.P. Souza, Vitória, J.L. Bezerra, Inácio & Dianese, **sp. nov.** FIGS. 1-2.

MycoBank MB 511338

*Stromata nigra 0.2–0.6 × 1.4–2.6 mm, elongata, unilocularia, raro multilocularia, subepidermalia, erumpentia. Ascomata 171–427 × 209–684 µm, subglobosa, erumpentia, ostiolata, externa cum pariete texturae angularis composita. Periphyses hyaline, 1.0–2.5 µm diam. Paraphyses 1.6–2.7 µm lata, filiformes, hyalina, septata, ramosae, copiosae. Asci 72–168 × 14–48 µm, clavati, unitunicati, octospori. Ascosporae 36–60 × 5–7 µm, hyalinae, pallido-brunneae, elongato-fusiformes, aseptatae, multiguttulatae, vagina mucosa irregularis involutae. Conidiomata ad 600 µm diam. Cellulae conidiogenae 1.0–1.5 µm diam. Conidia 3.0–14.5 x ca 0.5 µm, filiformia, recta vel sigmoidea, aseptatae, hialina.*

LESIONS black linear pustules along the foliar veins, 0.8–1.4 x 0.5–0.8 mm. STROMATA 0.2–0.6 (mean 0.5) × 1.4–2.6 (mean 1.9) mm, strongly erumpent, mostly unilocular, black discrete or confluent, mostly epiphyllous, occasionally hypophyllous, flat at the base, brown to black, sometimes coated by a temporary yellow pigment, carbonaceous, subglobose to ellipsoid, rough; walls pseudoparenchymatous, black, opaque, formed by host cells colonization. ASCOMATA 171–427 (mean 307) × 209–684 (mean 402) µm, subglobose, erumpent through the stromata, ostiolate, with outer wall composed of TEXTURA ANGULARIS, ostiole central, aplanate, rounded, slightly ribbed, lined internally by hyaline periphyses. PERIPHYSES hyaline, 1.0–2.5 µm diam. PARAPHYSES 1.6–2.7 µm diam, hyaline, filiform, septate, branched, abundant. ASCI 72–168 (mean 125) × 14–48 (mean 25) µm, clavate, unitunicate, short-stalked, thin-walled, 8-spored. ASCOSPORES 36–60 (mean 46) × 5–7 (mean 6.5) µm, hyaline to pale brown, elongate-fusiform, aseptate, multiguttulate, surrounded by an irregular wide mucilaginous sheath. CONIDIOMATAL STROMATA erumpent from the epidermal and hypodermal host cells, sometimes subcuticular, pulvinate, subglobose, but sometimes a little elongate, carbonaceous, black, shiny, disposed as the ascal stromata, 0.2–0.6 mm wide. CONIDIOMATA immersed in the stroma, dimidiate, black, 320 µm wide and 176 µm high, of thin walls, thicker at the base. CONIDIOGENOUS CELLS phialidic, lining the whole pycnidial locule, arranged in palisade, hyaline, branched, 1.0–1.5 µm wide,

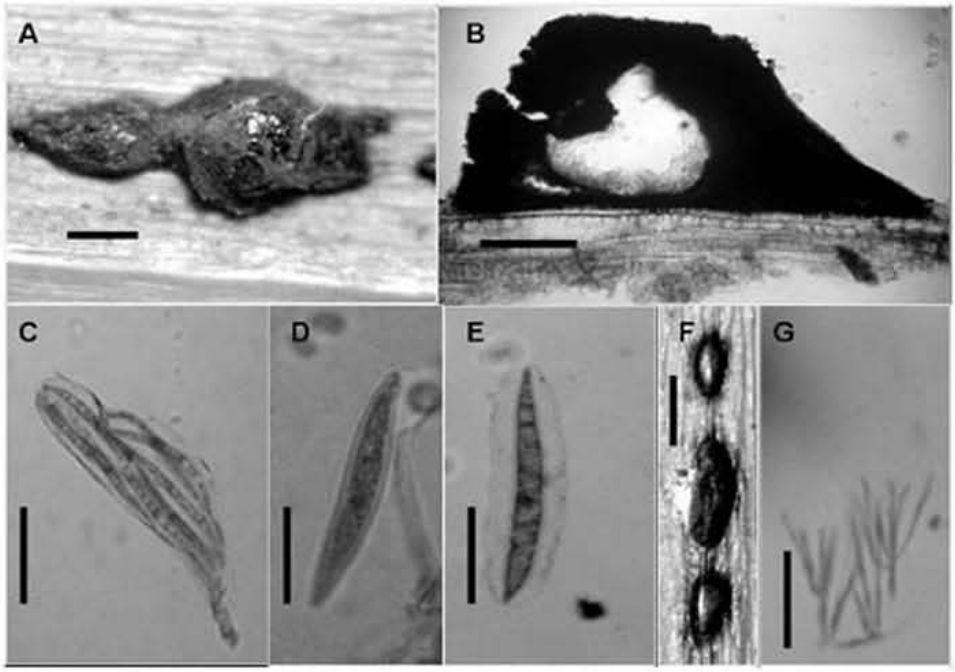


FIG. 1. *Camarotella brasiliensis* on *Syagrus schizophylla*: A. Perithecial stroma (bar = 0,5 mm); B. Lengthwise section of perithecia (bar = 50 µm); C. Asci (bar = 30 µm); D-E. Ascospores (note mucilaginous sheath surrounding spores) (bar = 30 µm); F. Pycnidial stromata (bar = 0.5 mm); G. Conidiophores and conidia. (bar = 30 µm).

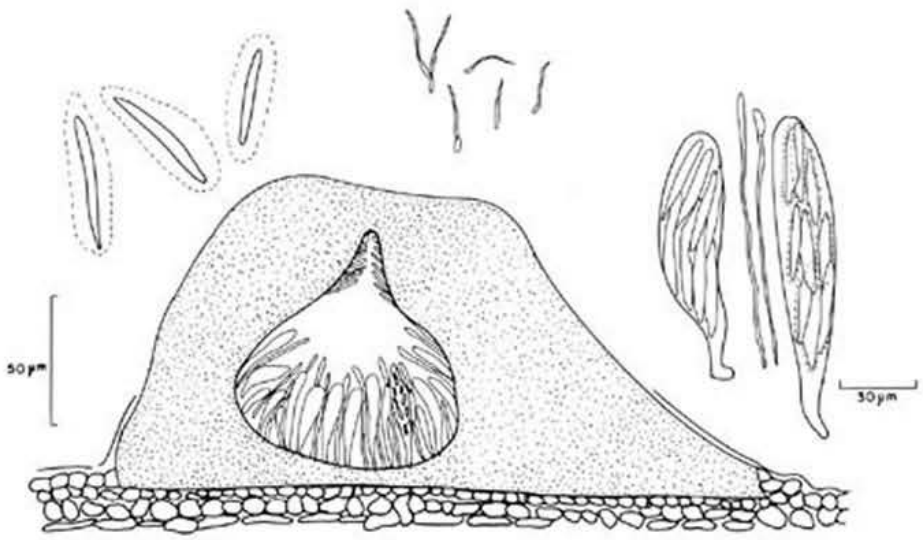


FIG. 2. *Camarotella brasiliensis* on *Syagrus schizophylla*: A. Perithecium with asci, paraphysis, ascospores and periphysis (bar = 50 µm). B. Asci and paraphysis (bar = 30 µm). C. Ascospores with mucous sheath (bar = 30 µm). D. Conidiophore and conidia (bar = 30 µm).

apex acuminate. CONIDIA filiform, hyaline, curved, sigmoid, acropleurogenous, 3.0–14.5 x 1.0 µm, aseptate.

SPECIMENS EXAMINED. BRAZIL. BAHÍA: Municipality of Valença, on leaflets of *Syagrus schizophylla*, 25 Feb 1986, *leg.* L. Hage 1946, (CEPEC Herbarium Mycological collection, 686, **holotype**). SERGIPE: north coastal region, on living leaves of *Syagrus schizophylla*, 10 April 1997, *leg. Dulce Warwick* 03, (UB- Col. Micol. 13.720).

## Discussion

Theissen & Sydow (1915) described the genus *Camarotella* as having thick walled asci and hyaline, four-celled ascospores. Petrak (1940) later revised this genus to correct Theissen & Sydow's description. According to Petrak, the asci are unitunicate and the ascospores unicellular. Clements & Shear (1931) and Wehmeyer (1975) treated the genus *Camarotella* but ignored the changes introduced by Petrak (1940).

Hyde & Cannon (1999), who examined parasitic fungi causing tar spots and similar diseases on palms, reevaluated the genus *Camarotella* and defined the following diagnostic characters: stromata that are black, pulvinate, strongly erumpent, and disposed in linear rows; anamorphs formed in thin-walled stromatic locules with pointed conidiogenous cells of percurrent proliferation and filiform, acuminate, curved conidia; one to several ascomata per stroma that are elongated towards the foliar veins; 8-spored asci that are unitunicate, cylindrical or cylindrical-clavate in shape and with or without an apical ring; ascospores that are short-fusiform, hyaline to light brown, unicellular, and surrounded by a mucilaginous sheath. *Camarotella acrocomiae* (Mont.) K.D. Hyde & P.F. Cannon was recognized as the type for this genus.

Hyde & Cannon (1999) further suggested a close relationship among species of *Camarotella*, *Oxodeora* K.D. Hyde & P.F. Cannon, and *Coccostromopsis* Plunkett. Species of *Oxodeora* produce stromata emerging through the leaf epidermis in a linear pattern, similar to those of *C. brasiliensis*; however, *Oxodeora* ascospores are ornamented with wide irregular ridges. *Coccostromopsis* stromata are superficial, pulvinate, and contain several locules, but the stromata are gelatinous with a yellowish outer layer when young, which distinguishes them from the *C. brasiliensis* specimens studied.

*Coccodiella* Hara is another black phyllachoraceous genus. *Camarotella* species form pulvinate leathery stromata as opposed to *Coccodiella* species with stromata that are flat, somewhat gelatinous, and with constricted bases (Cannon 1996, Hyde & Cannon 1999). Another major distinction lies in the conspicuous gelatinous sheath surrounding *Camarotella* ascospores that is lacking in *Coccodiella* (Hyde & Cannon 1999).

*Camarotella* species also differ from those of *Sphaerodothis* (Sacc. & P. Syd.) Shear. *Sphaerodothis* ascomata are never erumpent and produce short cylindrical asci and globose brown ascospores.

Two other *Camarotella* species are known to occur on *Arecaceae* — *C. acrocomiae* (ascospores 20–29 × 7.5–10 µm) and *C. costaricensis* (F. Stevens) K.D. Hyde & P.F. Cannon (ascospores 15–20 × 9–10 µm). The Bahía and Sergipe specimens have much longer (36–60 µm) and narrower (5–7 µm) ascospores that are distinctly fusiform and multiguttulate, with up to 8 large guttules per ascospore. These differences clearly show that the fungus belongs to a new *Camarotella* species, here designated as *C. brasiliensis*.

### Key to *Camarotella* species

1. Ascospores short-fusiform, not exceeding 30 µm long ..... 2
1'. Ascospores elongate-fusiform, multiguttulate, exceeding 30 µm long ... *C. brasiliensis*
2. Ascospores oblong to ellipsoid, blunt at both ends, usually brown ..... *C. acrocomiae*
2'. Ascospores fusoid, acute at both ends, usually hyaline to yellowish ... *C. costaricensis*

### Acknowledgements

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MYCOTAXON

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# *Postia ptychogaster*, an unusual two-stage polypore new to Italian mycobiota

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**Abstract** — *Postia ptychogaster* is reported from Italy for the first time. It was collected on a dead rotten trunk of *Abies alba* in a natural park, and was associated with a brown cubical rot. This species is well characterized by basidiomes closely associated with a typical *Ptychogaster* anamorphic stage forming large conidiomata. A full description, including photographs of both basidiomes and conidiomata and line drawings of microscopic characters, is provided.

**Key words** — biodiversity, wood-inhabiting fungi, chlamydospores, conidial stage, *Oligoporus*

## Introduction

In the *Oligoporus/Postia* assemblage (*Agaricomycetes*, *Polyporales*, *Fomitopsidaceae*) taxonomy is still somewhat unsettled and in flux (Niemelä 2005, Niemelä et al. 2005, Spirin et al. 2006). Apart from Yao et al. (1999), thorough molecular analyses on these taxa have not yet been performed, and are urgently needed. In the meanwhile, according with Vesterholt & Knudsen (1997), Pieri & Rivoire (2006), and Wei & Dai (2006), the genus *Postia* Fr. s.l. encompasses saprotrophic polypores characterized by a soft, effused-reflexed or flabelliform-pileate basidiome, annual or short-lived, white to pale coloured (some species have a rusty-brown upper surface), a monomitic hyphal system with thin- to distinctly thick-walled hyphae with clamp connections, small, allantoid, narrowly ellipsoid to sub-cylindrical spores, a brown rot and occurrence mainly on gymnosperms. Some species have well-differentiated cystidia and are able to form chlamydoconidia both in nature and in pure culture (Gilbertson & Ryvarden 1987, Ryvarden & Gilbertson 1994, Stalpers 2000, Niemelä 2005, Pieri & Rivoire 2006).

Field mycological investigations in an artificial plantation of *Picea abies* (L.) H. Karst. and *Abies alba* Mill. located in the "Parco naturale della Collina di Superga", a park hill area near Turin, Piedmont (northern Italy), yielded several specimens of *Postia ptychogaster*. This polypore, until now unknown from Italy (Ryvarden & Gilbertson 1994, Bernicchia 2005, Onofri et al. 2005), was found on a decayed fallen trunk of *Abies alba*, both in its anamorphic and its teleomorphic state. The aim of this paper is to provide a complete description of this rare species and to extend its geographic and hosts range.

## Materials and methods

Basidiomes and conidiomata were examined both macro- and microscopically with a stereo microscope (Leica MZ12) and a compound microscope (Leica DM4500B), respectively. Photographs were taken using a Leica DFC320 digital camera mounted on the compound microscope. The description of the microscopic features is based on fresh specimens. Sections were mounted in distilled water, in lactic acid plus acid fuchsine, in 5% potassium hydroxide, in Melzer's reagent, and in Cotton Blue. For conidia, basidia, and other structures at least 30 individuals were measured. The following abbreviations are used: Q = length to width ratio of the spores in side view; Qm = average quotient; IKI stands for Melzer's reagent, and CB is the abbreviation of Cotton Blue. $CB^+$ and $CB^-$ means cyanophilous and acyanophilous, respectively; $IKI^-$ means inamyloid and indextrinoid.

The abbreviation *P.* stands for '*Postia*' and *Pt.* for '*Ptychogaster*'. The colour notations in brackets are taken from Kornerup & Wanscher (1978), indicated as M. before a colour code. All examined material is deposited and kept at GDOR (Herbarium of the Museo Civico di Storia Naturale Giacomo Doria, Mycologia section, Genova, Italy). Herbarium abbreviations follow Holmgren & Holmgren (1998).

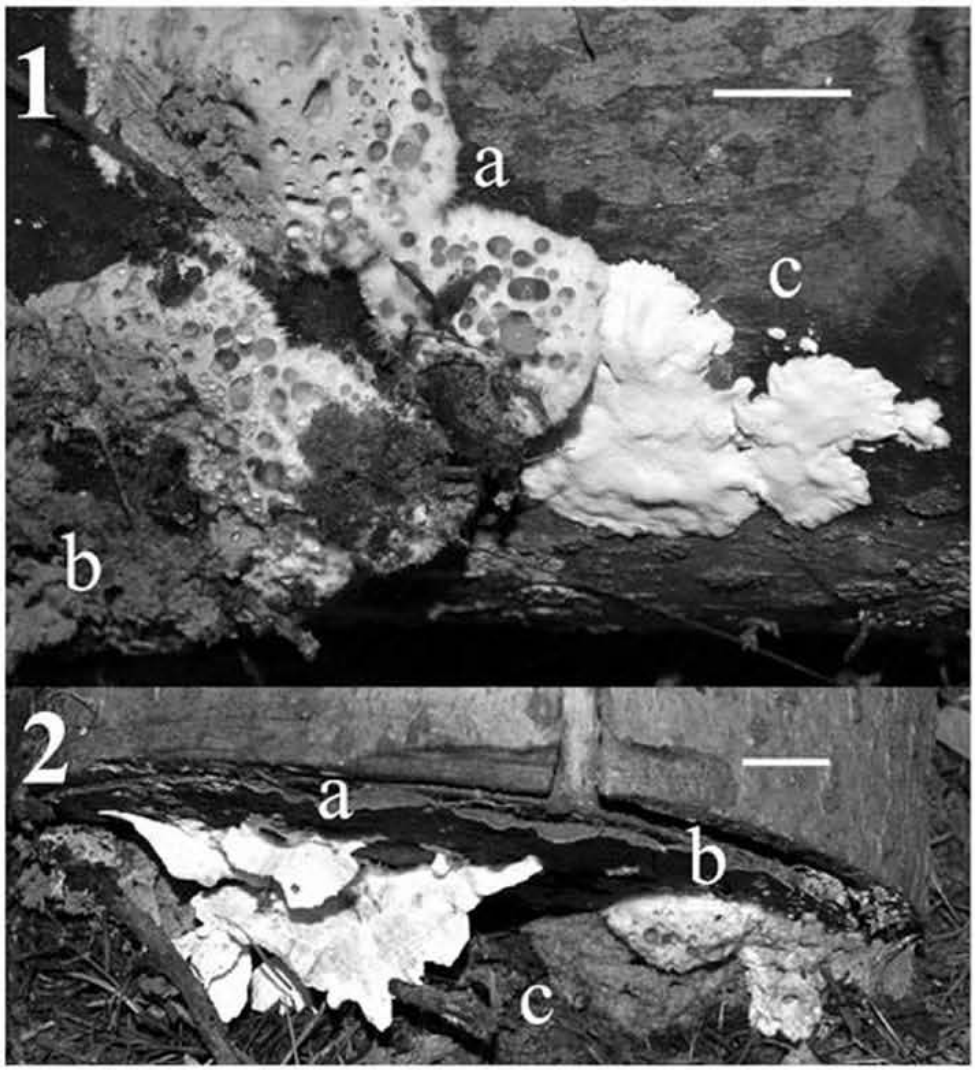
## Taxonomy

***Postia ptychogaster*** (F. Ludw.) Vesterh., in Knudsen & Hansen, Nordic J. Bot. 16: 213 (1996) FIGS. 1-9

= *Oligoporus ustilaginoides* Bref.; *Tyromyces ptychogaster* (F. Ludw.) Donk; *Oligoporus ptychogaster* (F. Ludw.) Falck & O. Falck

Anamorph: *Ptychogaster fuliginoides* (Pers.) Donk, Proc. K. Ned. Akad. Wet. 74: 124 (1972) [= *Ptychogaster pulverulentus* (Sowerby) Stalpers; *Ptychogaster albus* Corda; *Ptychogaster flavescens* Falck & O. Falck].

**Anamorphic, conidial stage** (FIGS. 1A,C; 2B,C; 3-8). Conidioma [= chlamydosporocarp sensu Clémençon (2004), ptychogasteroid or chlamydosporic fruitbody sensu Domański (1972)] always preceding basidioma development, as a hemispherical pulvinate, cushion-shaped structure, 2-5 cm in diam.,

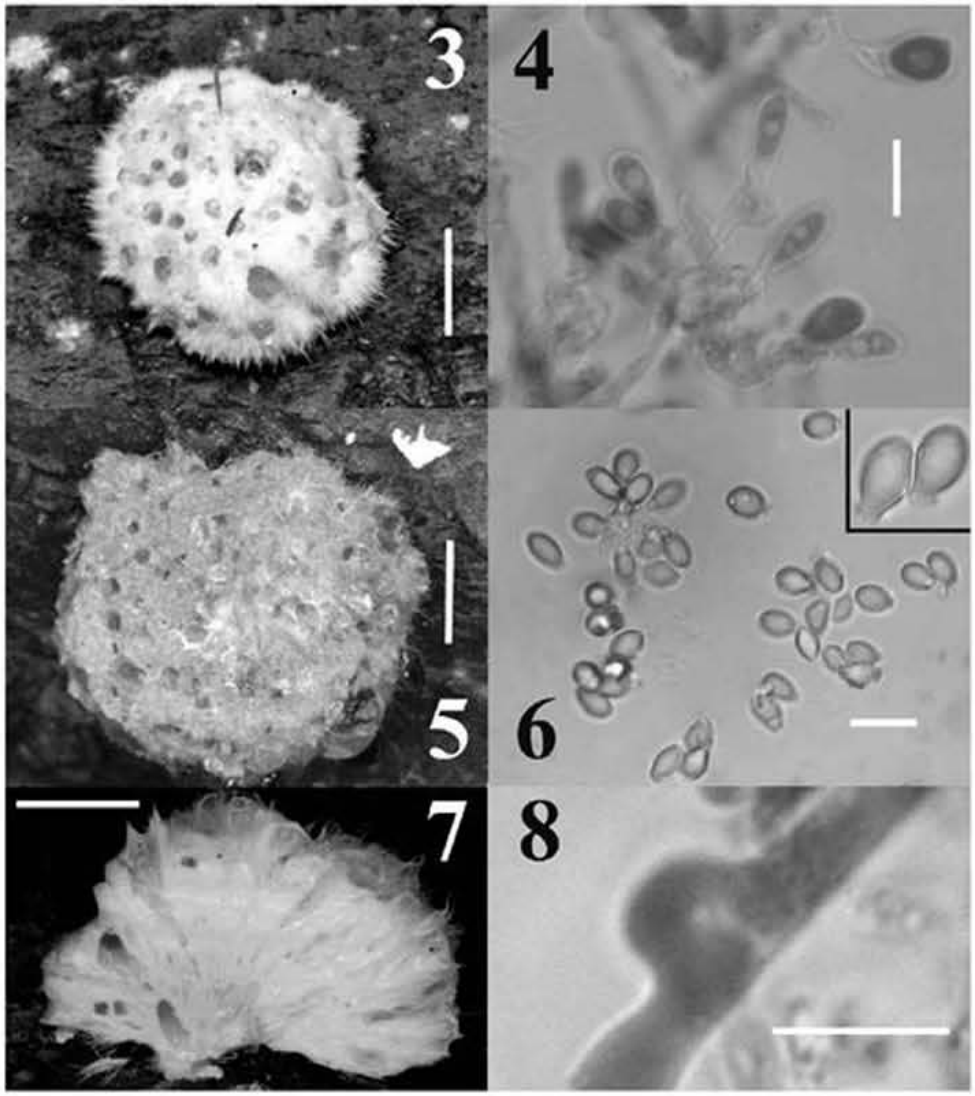



**Figs. 1–2.** *Postia ptychogaster*. **Fig. 1: a.** Young conidiomata. **b.** Effuse-resupinate basidiome. **c.** Powdery mass of chlamydoconidia. **Fig. 2: a.** Pileate basidiomes. **b.** Young conidiomata. **c.** Powdery mass of chlamydoconidia. Scale bars = 10 mm.

1-2.5(3) cm thick, usually shortly stipitate; when young white, sometimes with pink hues, loose and cheesy, spongy, watery, easily collapsing, slightly concentrically zoned in vertical cross section, exuding small amber-brown to cherry-red droplets [M. (6-9)A8; reminiscent of primordia of *Abortiporus biennis* (Bull.) Singer and *Hydnellum peckii* Banker], with an aculeate-villose surface consisting of soft, tufted hairs; when mature disintegrating into an ochraceous, powdery mass of conidia [at this stage it can be mistaken for an old myxomycete (Ryvarden & Gilbertson 1994, Clémençon 2002)]. By confluence

the conidiomata can form irregular masses reaching 6-8 cm or more in extent. Inner part of the conidioma prosenchymatic, at first consisting mainly of 70-110 µm wide synnema-like bundles of hyphae, running radially from a single basal core; at maturity composed of hyphae and brown powdery masses of conidia. Hyphae hyaline, thin- to slightly thick-walled, (3-)4.8(-7.5) µm wide, with abundant clamp connections. Clamps usually sprouting, perforate, often with a considerable open space between the hypha and the anastomosing body [medallion type (Stalpers 1978, Clémençon 2004)]. The conidiogenesis is probably holoblastic. Conidiophores (conidiogenous hyphae) hardly differentiated from vegetative hyphae, indiscrete, often arising from clamps, branched, each branch typically slightly swollen at the apex, with a clamp at some distance from the swelling, and developing a terminal blastoconidium. Blastoconidia light brown in mass [M. 6D(7-8)], arising from swollen parts of the conidiogenous hyphae, attached with a broad base, at first hyaline and thin-walled, brown-pigmented and thick-walled when mature (chlamydoconidia), often with a slightly flattened or truncated end, ovoid-ellipsoid, 5.0-8.0 × 3.5-5.0 µm in size, on average 7.05 × 4.52 µm, Q = (1.2-)1.4-1.7(-1.8), Qm = 1.56, smooth, IKI⁻, CB⁻, seceding rhexolytically and often with vestigial remnants of the subtending cell (clamped mother cell).

**Teleomorphic stage** (Figs. 1B, 2A, 9). Basidioma short-lived, annual, below or close to the imperfect stage, polymorphic, effused-resupinate to reflexed -pileate, and then spathulate-flabelliform or reniform, 1-4(-5) cm wide, 3-6 mm thick, projecting 1.5-4 cm, always with a narrowed or resupinate base, single or sometimes occurring in loose imbricate clusters. Upper surface, when present, glabrous to slightly pubescent, white to light cream-grey [M. 5B(2-3)], zonate, soft, fragile, when fresh with small, round and hygrophanous flecks. Lower surface white, ochraceous-cream when dry, pores irregular, lacerate, angular-elongated to sinuose, 2-3(-4) per mm, with slightly denticulate edges. Context 1-2 mm thick, white, fibrillose; tubes whitish to cream, (1-)2-3(-4) mm long. Taste at first mild, slightly bitter after long mastication. Odour indistinct. Hyphal system monomitic, hyphae hyaline, 3.75-5.15 µm wide, colourless, thin- to slightly thick-walled, strongly interwoven, with frequent branching, especially from clamps, easily disarticulating, with abundant close to perforate (medallion) clamp connections and sometimes with clamp complexes of 2-3 close-set clamps. Pileus covering, a cutis made up of parallel arranged hyphae, up to 6 µm wide. Cystidia or other sterile hymenial elements absent. Basidia 4-spored, clavate, 15-17 × 5-6 µm, sterigmata up to 4 µm long, with a basal clamp. Basidiospores ellipsoid, 4-5 × 2-2.5(-3) µm, on average 4.15 × 2.15 µm, Q = 1.7-2.1, Qm = 1.9, flattened on one side, hyaline, thin-walled, smooth, usually uniguttulate, IKI⁻, CB⁻.




**Figs. 3-8.** *Postia ptychogaster*. Macro- and micromorphological characters of the conidiomata. **3.** Young conidioma. **4.** Chlamydoconidia formation. **5.** Old conidioma. **6.** Mature detached chlamydoconidia. In square, two chlamydoconidia with vestigial remnants of the subtending cell. **7.** Conidioma in section. **8.** Large clamp connection (medallion type). (All from collection GDOR 07101401). Scale bars 3,5,7 = 10 mm; 4,6,8 = 10 µm.

Habitat: saprotrophic, causing a brown cubical rot, on a dead, decayed trunk of *Abies alba*, fruiting together with *Hymenochaete cruenta* (Pers.) Donk.

Material examined: 14/10/2007, numerous specimens, Parco naturale della Collina di Superga, 500 m a.s.l. (Baldissero Torinese, Turin, Italy), leg. L. Latino, GDOR 07101401.

## Discussion

*Postia ptychogaster* is a well-circumscribed species based on the effused-reflexed to pileate basidiomes, the absence of hymenial cystidia and the presence of a distinct and independent *Ptychogaster* anamorphic stage forming well-organized and large, pulvinate conidiomata. This imperfect stage always precedes the basidiome formation; conidiomata are far more frequent than basidiomes and are often found without the poroid stage (e.g. Domański 1972, Ryvarden & Gilbertson 1994, Clémençon 2002, Pieri & Rivoire 2006). In the Italian specimens, conidiomata were found strictly connected to the basidiomes. *P. ptychogaster* is reported as widespread in North and Central-Eastern Europe (Jülich 1984, Pegler & Saunders 1994, Ryvarden & Gilbertson 1994, Bondartseva 1998), although a rarely described and illustrated species, and was suspected to be endemic to Europe (Ryvarden & Gilbertson 1994). However, it has recently been found in Canada (Lamoureux 2007) and probably in the United States (UBC Botanical Garden Forums 2006). Our specimens represent the first collection of *P. ptychogaster* from Italy, and one of the rare reports from Mediterranean countries (South Europe). Well-known as a saprotrophic, brown-rot decay polypore, almost exclusively found on *Picea*, rarely on *Larix* and *Pinus*, and once noted on *Betula* (Domański 1972, Jülich 1984, Ryvarden & Gilbertson 1994, Niemelä 2005), it was collected in Italy on *Abies alba*.

As regard to the anamorphic stage, the genus *Ptychogaster* Corda, which encompasses the conidial stages of some species of *Postia*, is characterized by hyphae organized or not in a conidioma, provided with round medallion clamp connections at all septa and by one-celled conidia with a thick pigmented wall, born terminally, laterally or intercalary, mostly in branched chains and separated from the conidiogenous cell by a cross-wall, obovoid or broadly clavate, with a truncate, 2-3 µm wide base or truncate at both ends. The conidia are liberated by lysis of the mother (stalk) cell, remnants of which often remain attached to the base of the conidia (rhexolytic secession) (Ulbrich 1941, von Arx 1973, Sigler & Carmichael 1976, Stalpers 1984, 2000). The conidia form powdery masses. The genus *Ptychogaster* is morphologically closely related to *Sporotrichum*, which differs especially by the absence of clamp connections, and includes the anamorphs of *Laetiporus*, *Pycnoporellus* and *Phanerochaete*.

Our collection agrees fairly well with the descriptions and illustrations given by a number of authors (e.g. Domański 1972, Jahn 1970, 1979; Erkkilä & Niemelä 1986, Ryvarden & Gilbertson 1994, Bondartseva 1998, Stalpers 2000, Clémençon 2002, 2004; Bernicchia 2005, Niemelä 2005), except for one aspect of the conidiogenetic process: von Arx (1973), Sigler & Carmichael (1976) and Stalpers (1984, 2000), based on observations in pure culture, described conidia as formed in terminal clusters and/or in chains, whereas in our microscopic

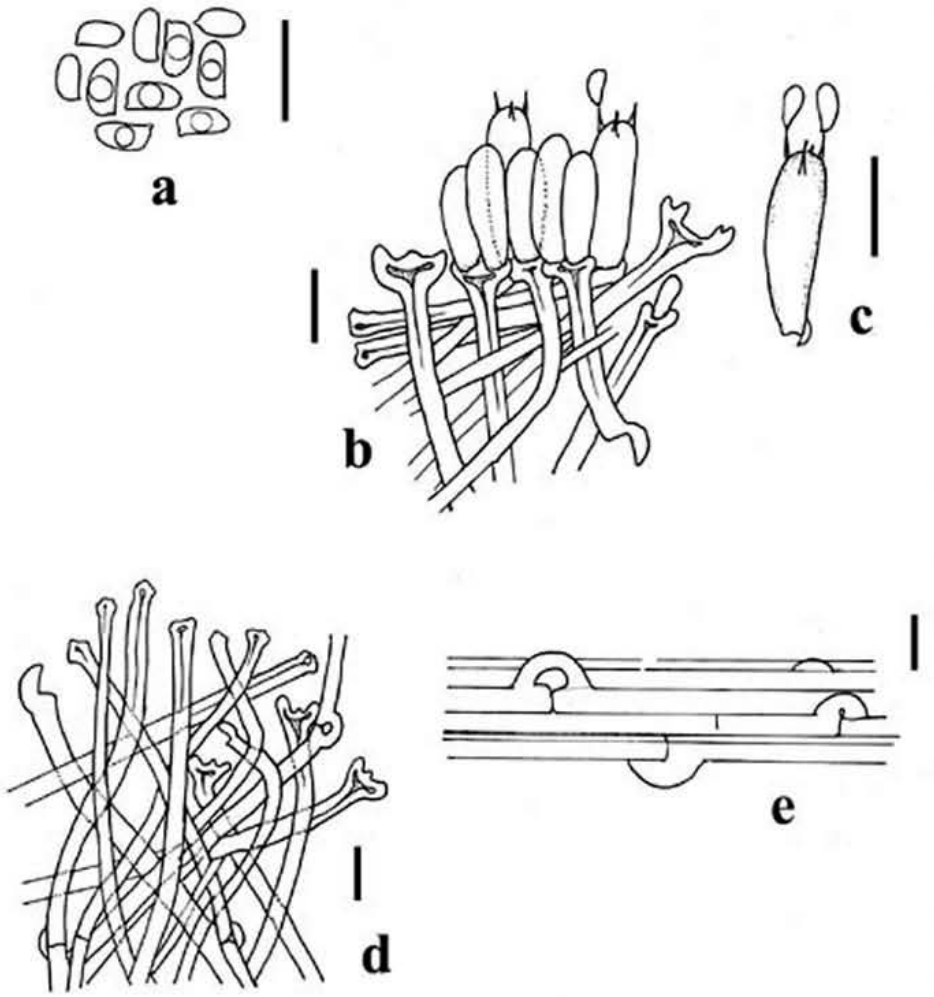



**Fig. 9.** *Postia ptychogaster*. Micromorphological characters of the basidiomes. a. Basidiospores. b. Subhymenial and tramal hyphae. c. Basidium. d. Contextual hyphae. e. Pileus covering hyphae. (All from collection GDOR 07101401). Scale bars = 10 µm. (Courtesy of Mido Traverso).

analysis of in natura conidiomata, only conidia borne singly and terminally are produced. It may be that the conidiogenesis pattern in cultures tends to be slightly different from that of hyphae growing on natural substrate. Regarding conidial measurements, reports range between 4.5-7 × 3.5-4.5 µm (Jahn 1970, 1979) to 5-10 × 3.5-7 µm (Ryvarden & Gilbertson 1994). Our measurements fits well with those of Fallahyan (1964; 5.5-6.5 × 5 µm), Bondartseva (1998; 5-8 × 3.5-5.5 µm), Stalpers [1978, 2000; (5.5-)6.5-8.5(-10) × 3.5-5(-5.5) µm]

and Pieri & Rivoire [2006; 5-6.5(-7) × 4-5(-5.5) µm]. Some authors named the disseminula of *Ptychogaster* spp. as chlamydospores: in our opinion, these structures more correctly should be called conidia or chlamydoconidia, as asexual propagules formed after mitotic events (in agreement with David 1980 and Stalpers 2000).

*P. rennyi* (Berk. & Broome) Rajchenb. (anamorph: *Pt. citrinus* Boud.) is regarded as the closest taxon among the *Postia* species developing conidia in pure culture and/or in nature (Ryvarden & Gilbertson 1994, Gilbertson & Ristich 1997, Stalpers 2000, Pieri & Rivoire 2006): it differs in having strictly resupinate-effused, very brittle basidiomes and slightly smaller chlamydoconidia at the margin of the basidiomes or below the collapsed tubes, forming a yellow powdery mass.

*Pt. rubescens* Boud. was first described from France (Boudier 1887); it produces conidia 4-6.5 × 3-4.3 µm in size (Domański 1972, Ryvarden Gilbertson 1994, Pieri & Rivoire 2006) in pink, fragile, hemispherical conidiomata especially on conifer timber (Stalpers 2000, Pieri & Rivoire 2006). It was suspected to represent the anamorph of either *P. guttulata* (Peck) Jülich (Davidson et al. 1946, Fidalgo 1958, Domański 1972), *P. floriformis* (Quél.) Jülich (Erkkilä & Niemelä 1986, Pieri & Rivoire 1998, 2006; Stalpers 2000) or *Punctularia atropurpurascens* (Berk. & Broome) Petch (Stalpers 1978). In our opinion it is very close (if not identical), and hardly distinguishable from *Pt. fuliginoides*. In addition, these anamorph/teleomorph connections do not seem to have definitively been established so far.

The doubtful taxon *Multiporus chlamydoformans* Falck & O. Falck (1937) described as producing a whitish, poroid, effused basidiome, with hyphae up to 7 µm wide and ellipsoid spores 5 × 3-3.5 µm, is characterized by the presence of conidiomata of a brown *Ptychogaster* anamorph, with conidia ellipsoid to subcylindrical, 10-15 × 9-12 µm in size.

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# *Chalciporus africanus*, a new bolete species from Africa

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**Abstract** — A new bolete species, *Chalciporus africanus*, has been discovered in the rain forest of Cameroon. It is the first representative of its genus to be described from the African continent.

**Key words** — *Boletales*, taxonomy

## Introduction

The present knowledge of African boletes refers to about 150 species collected from only a few localities. Comprehensive contributions towards the knowledge of the Central African taxa were published by Heinemann (1951, 1953, 1954a, b, 1955, 1960, 1964a, b, 1966), Heinemann & Rammeloo (1980, 1982, 1983a, b, 1986a, b, 1987, 1989), Rammeloo & Heinemann (1982, 1987, 1988, 1990, 1995) and Rammeloo et al. (1988). East African taxa were reported by Watling & Turnbull (1992, 1994).

Several genera of the *Boletaceae* sensu Singer (1986) have not been recorded in Africa, notably, *Chalciporus* Bataille. Despite this genus having representatives in temperate northern hemisphere, tropical and sub-tropical regions, it was surprising that no records occurred in Africa. Macro-morphologically *Chalciporus* is well characterised by pink or reddish hymenophore, the presence of yellow mycelium at the base of its stipe (which lacks a reticulum or asperulation) and the absence of velar remnants on both cap and stipe. The cystidia are usually conspicuous, the smooth spores are elongated or short ellipsoid. The spore prints typically lack olive tinges. Clamp connections are absent and in fresh specimens the mediostratum of the hymenophore is often tinged red.

During a collecting trip to the Dja Forest Reserve in the Eastern Province of Cameroon, a new species of *Chalciporus* was gathered on a river edge under *Uapaca guineensis*.

## Materials and methods

The macroscopic description is based on field notes and photographs. Codes (between square brackets) and names for colours correspond with the Methuen Handbook of Colour (Kornerup & Wanscher 1983).

The microscopic structures were observed in a 1% Congo-red solution and Melzer's reagent. Measurements were performed using an Olympus BX51 light microscope, with digital camera and AnalySIS® Five imaging software (Soft Imaging System GmbH). Mean values (in italics) ± 1.96 × standard deviations, and minimum-maximum values (between brackets) are given for all microstructures and derived parameters (length/width ratios). For the statistical data 75 basidiospores and 10 basidia and cystidia were measured. The holotype material and the additional collections are deposited at the National Botanic Garden of Belgium's herbarium 'BR' (abbreviation following Holmgren et al. 1990).

## Taxonomy

***Chalciporus africanus*** Degreef & De Kesel, **sp. nov.** FIGURES 1 & 2

MYCOBANK MB 511404

*Pileus 10–30 mm, cinnamomeus dein suffusco-rufus, aliquantulus lurido-maculatus, siccus, subtomentosus. Tubi adnati dein subdecurrentes vel decurrentes, pallidi-flavi dein rubri. Pori concolores ut ad instar tuborum, paulatim rufescentes ab centro. Stipes 30–55 x 3–5 mm, cylindricus vel subclavatus, griseo-croceus sed rubello-fuscus infra tubos, ad apicem minute furfuraceus, mycelium basale flavum. Caro crassa, albida in centro pilei et in basim stipitis, alibi rubella, immutabilis. Basidiosporae (7.1–10.3 × 3.3–4.2 µm) ellipsoideae vel fusoideae, leves, pallido-melleae, inamyloideae. Basidia (30–38× 7–9 µm) cylindrica vel subclavata, tetraspora. Pleurocystidia (35–57× 7–10 µm) cylindracea, anguste lageniformia vel anguste clavata. Cheilocystidia (35–62 × 11–16 µm) pedunculata vel lageniformia. Pileipellis ex hyphis haud gelatinosis trichodermium formantibus. Fibulae desunt.*

HOLOTYPUS: AFRICA, CAMEROON, Eastern Province, Dja Forest Reserve, Somalomo, 7.IV.2007, *Degreef* 517 (BR).

PILEUS 10–30 mm in diameter, hemispherical to sub-umbonate, becoming plano-convex; cinnamon [6D6] then brownish red [9C6] to dull red [9B4] with sparse pale yellow [3A3] stains; surface dry, flocculose; margin obtuse, straight; context thick especially at the centre of the pileus (up to 10 mm), white to cream-colour [4A3] with pale red to pink [10A3] shades towards the stipe. TUBES sub-decurrent then decurrent, up to 5 mm long, light yellow [4A4] then reddish brown [9E6] to oxblood red [9E7]. PORES irregular, up to 1 mm in diameter; concolorous with the tubes, progressively becoming red from the centre to the margin when aging, pores at the margin often paler. STIPE 30–55 × 3–5 mm, central, cylindrical to sub-clavate with base up to 10 mm

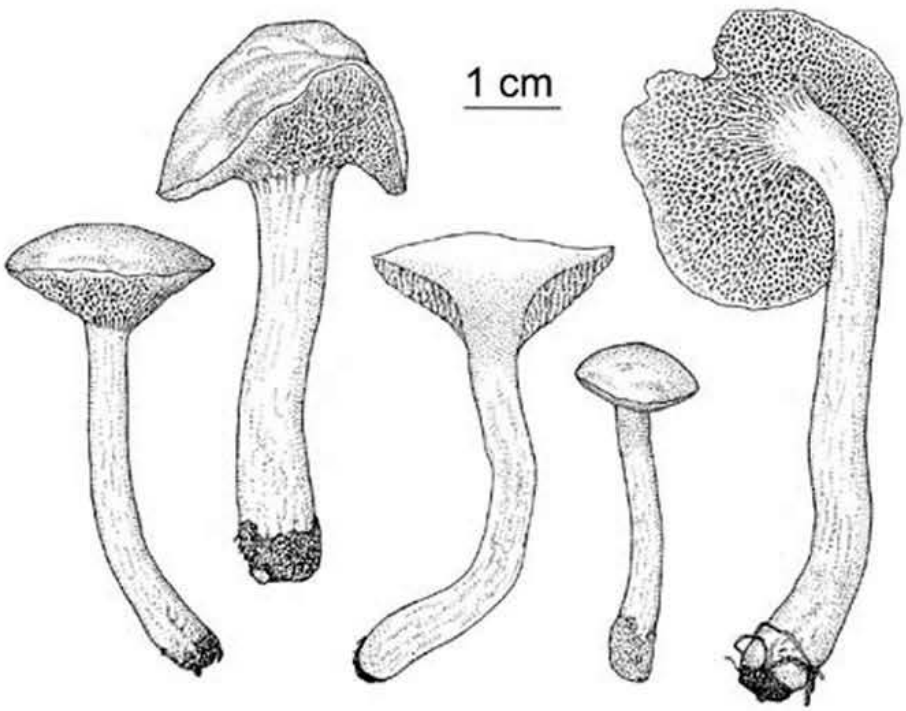


Fig. 1. *Chalciporus africanus* (*Degreef* 517, holotype). Basidiomes.

wide; solid, fibrous; reddish brown [9E6] immediately below the tubes, greyish orange [5B4] elsewhere, with cinnamon to cognac brown [6D6–6E7] floccules in its upper third. Odour agreeable, weak. TASTE mild. SPORE PRINT dark blond [5D4]. MYCELIUM yellow.

BASIDIOSPORES (7.1–)7.5–*8.7*–9.9(–10.3) × (3.3–)3.4–*3.8*–4.2(–4.2) µm, (l/w = (1.9–)1.97–*2.31*–2.65(–2.7)), broadly elliptical to broadly fusiform, with a very moderate suprahilar depression, weakly pigmented (under the microscope), uni- or biguttulate, smooth, inamyloid. BASIDIA (30–)29.1–*33.5*–37.9(–38) × (7–)7.1–*8.3*–9.5(–9) µm, cylindrical to narrowly clavate, hyaline or containing small granules while immature, mostly 4-spored. PLEUROCYSTIDIA (35–)37.3–*49.5*–61.7(–57) × (7–)6.7–*8.5*–10.3(–10) µm, cylindrical to narrowly lageniform, sometimes narrowly clavate, emergent, thin-walled, concolorous with the hymenium, hyaline, without crystals or incrustations. CHEILOCYSTIDIA (35–)32.2–*43.2*–54.2(–62) × (11–)10.5–*13.1*–15.7(–16) µm,

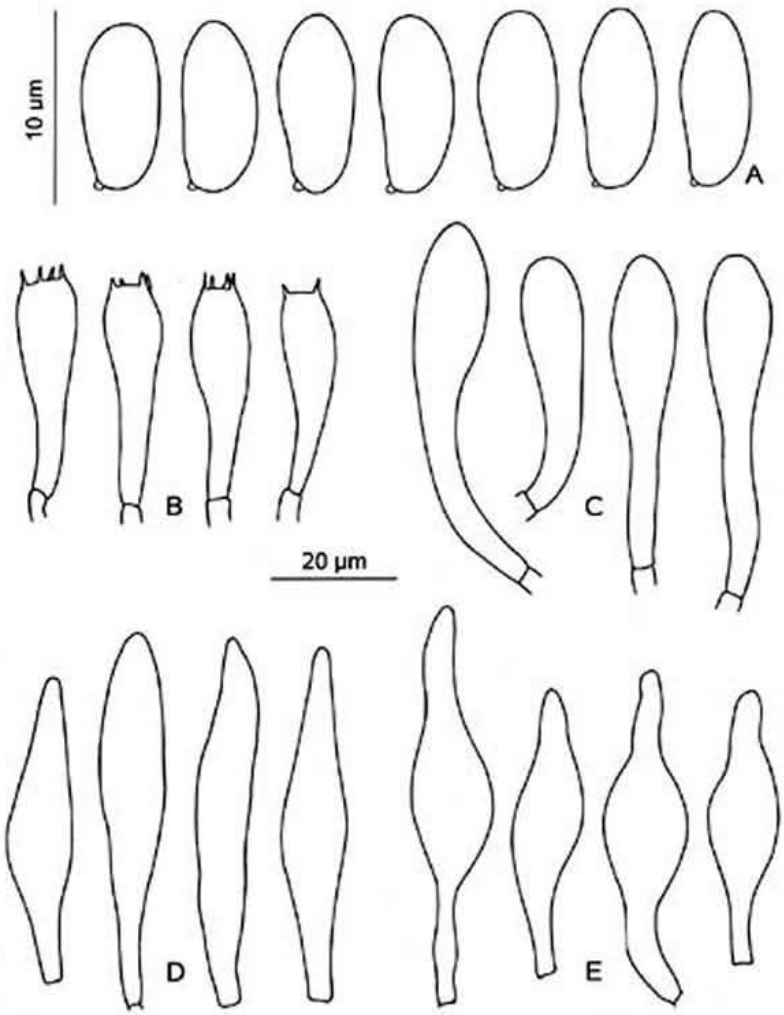


Fig. 2. *Chalciporus africanus* (*Degreef* 517, holotype).
A. basidiospores; B. basidia; C. caulocystidia; D. pleurocystidia; E. cheilocystidia.
(A: vertical scale bar; B-E: horizontal scale bar).

narrowly lageniform to lageniform, sometimes pedunculate, emergent, thin-walled, hyaline, without crystals or incrustations. PILEIPELLIS a trichoderm, collapsing into a cutis, 50–70 µm thick, consisting of parallel, pigmented, non-amyloid, thin-walled hyphae, 40–80 × 5–7 µm; terminal cells narrowly clavate to clavate, 20–30 × 10–12 µm, with scattered incrustations; no mucilagineous layer. STIPITIPELLIS composed of hyaline, thin-walled hyphae, 20–90 × 5–10

µm; caulocystidia from floccules narrowly clavate, (38–)33.6–*46.4*–59.2(–58) × (7–)5.6–8.8–12(–11) µm. TRAMA composed of hyaline or reddish, thin-walled hyphae, up to 14 µm wide, mostly parallel or very slightly divergent from the mediostratum, oleiferous hyphae absent. CLAMP CONNECTIONS absent.

ECOLOGY AND DISTRIBUTION: Rain forest, scattered along river's edge, growing under the tree *Uapaca guineensis* (*Euphorbiaceae*).

SPECIMENS EXAMINED– CAMEROON. EASTERN PROVINCE, DJA FOREST RESERVE, near Somalomo, N03°23'39" E12°43'25". *Degreef* 506, 6.IV.2007. Ibid. *Degreef* 516, 7.IV.2007. Ibid. *Degreef* 517, 7.IV.2007 (HOLOTYPE-BR).

COMMENTS — *Chalciporus africanus* is similar to the temperate *C. rubinus* (W.G. Sm.) Singer but differs due to its larger and more elongated spores, unchanging context and prominent reddish colour of its cap. As a consequence, *C. africanus* bridges the generic difference between *Chalciporus* and *Rubinoboletus* Pilát & Dermek. The close relationship between *C. africanus* and *C. rubinus* confirms Singer's statement that *Boletus rubinus* W.G. Sm. is a good species of *Chalciporus*, making *Rubinoboletus* a synonym of *Chalciporus* (Singer 1986). The genus *Rubinoboletus* was created to accommodate *C. rubinus* based solely on its globose spores (Pilát & Dermek 1969). We think that all globose-spored taxa subsequently combined in, or described under, *Rubinoboletus* should be placed elsewhere. In this context, sub-globose spores are to be expected in any alliance of elongated spores.

Macroscopically *Chalciporus africanus* is similar to the temperate and widely distributed *C. piperatus* (Bull.: Fr.) Bataille. The latter, however, is a much taller species with a peppery taste, longer (average 9.8 µm) spores, and incrusted cystidia (Muñoz 2005, see fig. 33, p. 275).

*Chalciporus africanus* is possibly also close to the tropical *Boletus* (*Xerocomus*) *rubriporus* Corner from Malaysia (Corner 1972), a species that now resides in *Chalciporus* (Singer 1986). *C. rubriporus* and *C. africanus* share a similar general habitus and pileipellis arrangement of loosely interwoven hyphae that are not arranged in a pile. *C. rubriporus*, however, has larger spores, more elongate cystidia, and discolouring pores and tubes.

Singer (1986) stated that *Chalciporus* has 'erratic' mycorrhizal associations, i.e. a wide range of hosts. *Chalciporus* is known to associate with *Pinus*, *Fagus*, *Quercus*, *Betula*, *Nothofagus* and *Cistus*. *Chalciporus africanus* however, seems to be specific as it was exclusively found under the endemic *Uapaca guineensis*, a large tree typically growing along rivers and rivulets. In Africa, this host is frequently reported to be associated with many other ectomycorrhizal boletes (Thoen 1993), but taxa of *Chalciporus* had not previously been recorded.

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# Diversity of nivicolous myxomycetes in the Gorce mountains – a low-elevation massif of the Western Carpathians

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**Abstract** — Nivicolous myxomycetes constitute an ecologically well-defined group of organisms requiring the particular set of environmental conditions that occur in mountain habitats with long-lasting winter snow cover. In Europe, relatively complex data sets relating to this group are available only from the Alps and mountains of the Iberian peninsula, whereas there is almost no data from other mountainous areas. In order to contribute to the knowledge of the diversity and distribution of this group in the central European mountains, we carried out the first comprehensive study in the Carpathians, one of the major parts of the European mountain system. A survey of the diversity of nivicolous myxomycetes was conducted in a low-elevation area of the Gorce Mountains (Western Carpathians, Poland), including fieldwork during six seasons. We recorded 18 species, ten of which are reported for the first time from Poland. *Diderma niveum*, *Lepidoderma chailletii* and *Lamproderma ovoideum* repeatedly occurred very abundantly and represented an important element in the spring phenological aspect of glades and shrub communities of the study area.

**Key words** — biogeography, taxonomy

## Introduction

Nivicolous myxomycetes constitute an ecologically well-defined group of organisms requiring a particular set of environmental conditions that occur in mountain habitats with long-lasting winter snow cover, at the edge of melting snow in spring. There is an increasing body of data relating to the world distribution of this group, but it is still so scarce and unequally distributed that a major obstacle remains with respect to approaching general problems. In Europe, relatively complex data sets are available, mainly from the Alps (e.g. Meylan 1931, 1932; Schinner 1982, Cochet & Bozonnet 1984, Bozonnet et al. 1991, Meyer 1986, Meyer et al. 1996, Neubert et al. 1993, 1995, 2000; Singer

et al. 2001ab, Poulain et al. 2002a) and from the mountains of the Iberian peninsula (e.g. Sánchez et al. 2002, Moreno et al. 2003a, Lado 2004, Lado et al. 2005). Other mountain regions have not been investigated thus far, or only incidentally collected data are available (e.g. Ronikier & Ronikier 2007). Systematic regional investigations are needed to develop a better understanding of the biogeography and ecology of this group.

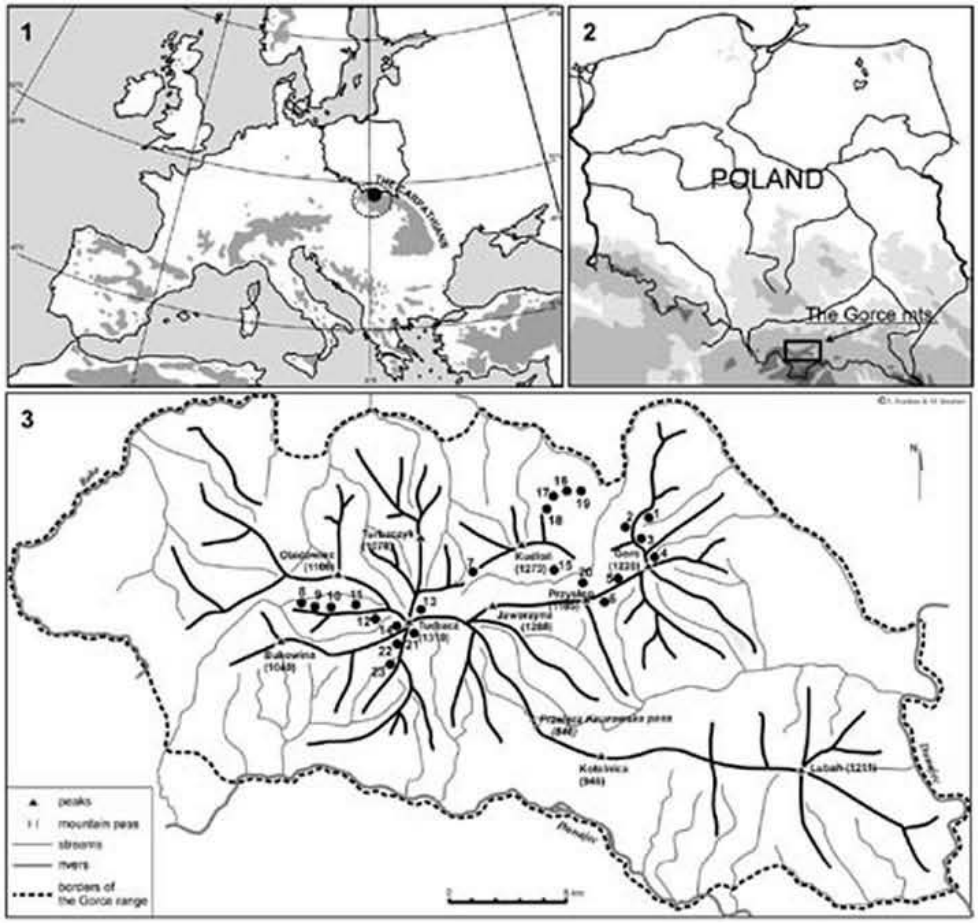
The Carpathians form one of the major elements of the European alpine system (Ozenda 1985), but data concerning nivicolous myxomycetes from this range are very scarce. Only seven strictly nivicolous species have been reported thus far from incidental observations in the Western Carpathians. They are *Diderma alpinum*, *D. niveum*, *Lamproderma ovoideum*, *L. sauteri*, *Lepidoderma chailletii*, *Physarum albescens* and *Trichia alpina* (Wichanský 1962, 1963; Drozdowicz 1985, 1988, 1995; Komorowska & Drozdowicz 1996). The first observations have recently been reported from the Eastern Carpathians (Drozdowicz 1997, 2000; Krivomaz et al. 2005).

Herein, we present the first more complete report from a mountain massif in the Carpathians. Based on several field prospects, we investigated the diversity of nivicolous myxomycetes in the Gorce Mountains, a model lower-mountain massif representative of the greater part of the Western Carpathians. Specific aims of this study were (1) to outline the diversity of nivicolous slime moulds in the Western Carpathian massif and (2) to make a contribution to the knowledge of the distribution of this group in the central European mountains, an area very poorly investigated in this respect.

## Materials and methods

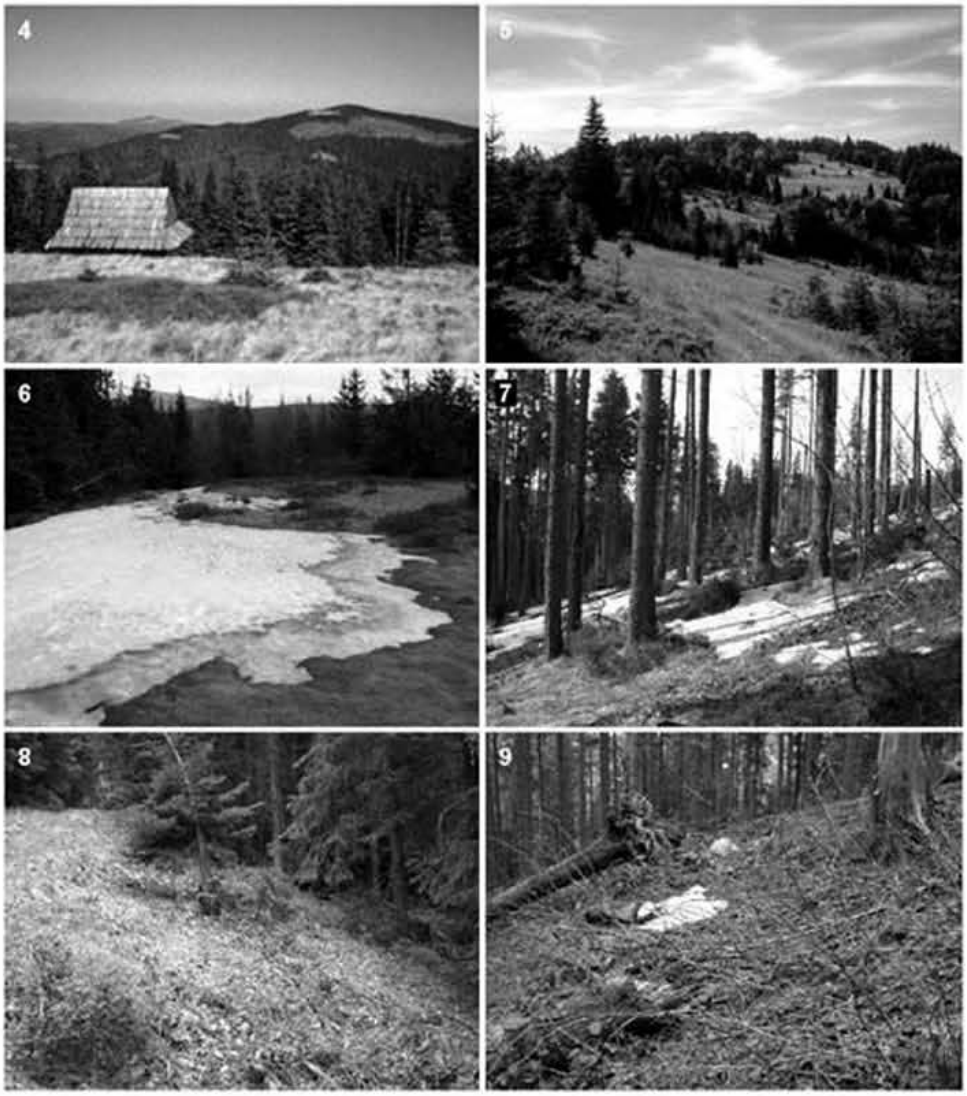
### The study area

The Carpathian arch is divided into three main biogeographical units; these are the Western, Eastern and Southern Carpathians. The Gorce Range forms a part of the Western Carpathians (Figs. 1-3). These mountains cover an area of approximately 550 km$^2$ and represent a typical landscape of this part of the Carpathians, dominated by lower-mountain areas built of flysch (Figs. 4-5). As one of the most representative parts of the Western Carpathians, a large part of the Gorce massif is protected as a national park. The Gorce Mountains reach an elevation of 1310 m a.s.l., but only about 10% of the massif exceeds an elevation of 1000 m. The mean temperature of the year in the highest areas is estimated at 2–3°C, while the mean temperature of January is -6.2°C. The snow cover is usually present from November to April, up to 170 days on northern exposures, where patches of snow still persist in May (Kornaś 1955, Miczyński 2006). Two elevational vegetation zones can be distinguished in the mountains: the lower montane belt with beech, fir and spruce forests (up to 1150–1200 m




Figs. 1-3. Location of the area of research. 1. Location of the Carpathians in Europe; Western Carpathians embedded with dashed line. 2. Location of the Gorce massif. 3. The Gorce Range; dots – localities of sampling, numbers refer to those in Table 1.

a.s.l.) and the upper montane belt dominated by spruce forests (Kornaś 1955). Natural treeless (subalpine and alpine) vegetation is absent from the massif due to moderate elevations, but the presence of numerous anthropogenic semi-natural glades, due to multicentennial pastoral husbandry, is a characteristic feature of this area, increasing the landscape diversity (Figs. 4-6). Changes in land use during the last decades, especially the abandonment of grazing and hay-making, launched transformation of open glades into shrub communities with *Vaccinium myrtillus*, followed by expansion of young trees (Figs. 5-6). All of these conditions form a potentially diverse mountain environment for nivicolous myxomycetes, but not characterized by subalpine or alpine habitat features (Figs. 6-9).




Figs. 4-9. Area of research. 4, 5. Typical landscape of the Gorce massif with spruce montane forest and abandoned pastures with young trees in succession. 6-9. Localities of sampling: 6. mountain meadow, 7. forest interior, 8. small clearing by path, inside forest, 9. shrub vegetation inside forest.

Table 1: Localities of collections in the Gorce mountains (Western Carpathians).

| N° | Locality, habitat, date of collection, collector | Geographic coordinates | Elevation |
|---|---|---|---|
| [1] | Upper part of the Głębieniec valley, N ridge of the Gorc Mtn., E slope, by a path, shrubs with *Rubus* sp.; 18 Apr 2004; *leg. AR, MR* | 20°14'47" E 49°34'50" N | 1030 m |
| [2] | Upper part of the Głębieniec valley, N ridge of the Gorc Mtn., E slope, about 200 m N from the glade Świnkówka, steep slope in beech forest, shrubs with *Rubus* sp.; 18 Apr 2004; *leg. AR, MR* | 20°14'24" E 49°34'32" N | 1060 m |

| | | | |
|---|---|---|---|
| [3] | NW part of the glade Gorc Kamienicki (N ridge of the Gorc Mtn.), meadow; 18 Apr 2004; *leg. AR, MR* | 20°14'56" E 49°34'17" N | 1130–1150 m |
| [4] | S part of the glade Gorc Kamienicki (N ridge of the Gorc Mtn.), meadow; 18 Apr 2004; *leg. AR, MR* | 20°15'07" E 49°34'03" N | 1150 m |
| [5] | Ridge between the Gorc Mtn. and the Przysłop Mtn., S slopes, the glades Chyzniocka and Przysłop Dolny, meadow; 18 Apr 2004; *leg. AR, MR* | 20°13'19" E 49°33'13" N | 1080–1090 m |
| [6] | Ridge between the Gorc Mtn. and the Przysłop Mtn., S slopes, the glade Polana Przysłop Dolny, meadow; 18 Apr 2004; *leg. AR, MR* | 20°12'45" E 49°33'01" N | 1150 m |
| [7] | Ridge from the Borek pass to the Kudłoń Mtn., the glade Przysłopek (S slopes), meadow; 18 Apr 2004; *leg. AR, MR* | 20°09'21" E 49°33'55" N | 1100 m |
| [8] | Ridge of the Średni Wierch Mtn., above the village Obidowa, below the glade Stusy, degraded beech forest with spruce; 13 May 2006; *leg. AR, MR* | 20°02'32" E 49°32'52" N | 880 m |
| [9] | Ridge of the Średni Wierch Mtn., above the village Obidowa, surroundings of the glade Stusy, meadow; 13 May 2006; *leg. AR, MR* | 20°02'40" E 49°32'48" N | 950 m |
| [10] | Ridge of the Średni Wierch Mtn., above the village Obidowa, the glade Tynowe, meadow; 13 May 2006; *leg. AR, MR* | 20°03'19" E 49°32'49" N | 1020 m |
| [11] | Ridge of the Średni Wierch Mtn., above the village Obidowa, small glades close to the Średni Wierch Mtn., meadow; 13 May 2006; *leg. AR, MR* | 20°03'19" E 49°32'49" N | 1090 m |
| [12] | Ridge of Turbacz Mtn., the Rozdziele Mtn., at the red tourist trail, spruce forest; 13 May 2006; *leg. AR, MR* | 20°05'52" E 49°32'57" N | 1190 m |
| [13] | Ridge of Turbacz Mtn., surroundings of the glade Turbacz, N slopes, meadow; 13 May 2006; *leg. AR, MR* | 20°06'36" E 49°32'50" N | 1230 m |
| [14] | Turbacz Mtn., summital area, spruce forest (upper montane belt); 13 May 2006; *leg. AR, MR* | 20°06'41" E 49°32'34" N | 1280–1310 m |
| [15] | Range of Kudłoń Mtn., below the glade Stawieniec, edge of forest; 21 Oct 2006; *leg. AC* | 20°11'34" E 49°33'45" N | 1000 m |
| [16] | NE ridge of the Kudłoń Mtn., the glade Jaworzynka, meadow; 20 Apr 1983; *leg. AD* | 20°12'25" E 49°35'15" N | 1020 m |
| [17] | NE ridge of the Kudłoń Mtn., the glade Podskały, meadow; 20 Apr 1983; *leg. AD* | 20°11'53" E 49°35'12" N | 1000 m |
| [18] | NE ridge of the Kudłoń Mtn., the glade Adamówka, meadow; 20 Apr 1983; *leg. AD* | 20°11'29" E 49°34'43" N | 1050 m |
| [19] | NE ridge of the Kudłoń Mtn., E edge of the glade Jaworzynka, meadow; 4 Apr 1981; *leg. AD* | 20°12'43" E 49°35'17" N | 990 m |
| [20] | Kamienica valley, the Przysłop Mtn.; 5 May 2006; *leg. AC* | - | - |
| [21] | Turbacz Mtn., vicinity of the refuge; 25 May 2001; *leg. AD* | 20°07'10" E 49°32'34" N | 1270 m |
| [22] | S ridge of the Turbacz Mtn., the glade Długie Młaki, meadow; 19 May 2000; *leg. AD* | 20°06'28" E 49°32'03" N | 1200 m |
| [23] | S ridge of the Turbacz Mtn., the glade Rusnakowa, meadow; 19 May 2000; *leg. AD* | 20°06'19" E 49°32'02" N | 1150 m |

Collector abbreviations: AC – Andrzej Chlebicki; AD – Anna Drozdowicz; AR – Anna Ronikier; MR – Michał Ronikier.

### Field work and methods of analysis

Field collecting was made in spring in 1981, 1983, 2000, 2001, 2004 and 2006, in various sites of the Gorce Mountains, mainly in the area of the Gorce National Park. Single collections made in 2006 and kindly provided by A. Chlebicki were also taken into account. In total, 23 sites were investigated (Table 1, Fig. 3). Collections were made in open places such as glades and meadows, close to the melting snow, as well as inside forest, at snow patches or in places where the snow disappeared. All collections were deposited in KRAM or KRA, with some duplicates in MA-Fungi and in the private herbarium of Marianne Meyer.

Standard methods used in the taxonomy of myxomycetes were applied to study the collected material. Analysis of microscopic characters and all microscopic measurements were performed on permanent preparations fixed in Hoyer's medium; measurements were made under the oil immersion ×100 objective. For each collection 20 spores were measured; the reported spore size does not include spore ornamentation. The scanning electron microscopy (SEM) observations were carried out with a Hitachi S-4700 microscope, using 10 kV voltage and working distance of about 12 mm. The material was prepared in a sequence of acetone dilutions (50% – 100%) followed by the critical point drying procedure and coating with gold.

Nomenclature used for myxomycetes follows Hernández-Crespo & Lado (2005; http://www.nomen.eumycetozoa.com).

## Results

In total, 148 collections were gathered in the Gorce Mountains within the framework of this study. Eighteen species were identified from this material, all but *Didymium dubium* considered to be strictly nivicolous (Bozonnet et al. 1991). Ten taxa are new to Poland; they are marked with an asterisk "*" in the list below. Numbers of collection sites given in bold in brackets refer to TABLE 1.

***Diderma alpinum*** (Meyl.) Meyl.

SPECIMENS EXAMINED. [**10**], on stems of *Rubus* sp., KRAM M-1157, duplicate in herb. Marianne Meyer; [**18**], on grass culms, mosses, beech leaves and other plant remnants, KRA MYXO-520; KRA MYXO-523; [**19**], on grass culms or plant remnants, KRA MYXO-525; KRA MYXO-526.

****Diderma globosum*** var. ***europaeum*** Buyck

SPECIMENS EXAMINED. [**2**], on fallen twigs and leaves of *Fagus sylvatica*, fallen twigs and needles of *Picea abies*, on stems of *Rubus* sp., KRAM M-1151, duplicate in MA-Fungi; [**4**], on stems of *Rubus* sp., KRAM M-1149.

***Diderma niveum*** (Rostaf.) T. Macbr.

SPECIMENS EXAMINED. [4], on stem of *Rubus* sp. or *Vaccinium myrtillus*, KRAM M-1152; KRAM M-1154; [5], on stems of *Rubus* sp. or *Vaccinium myrtillus*, KRAM M-1150; KRAM M-1207; [11], on stems of *Vaccinium myrtillus* or grass culms, KRAM M-1158, duplicate in MA-Fungi; KRAM M-1156; [13], on stems of *Vaccinium myrtillus*, KRAM M-1206; KRAM M-1208; [14], on stems of *Vaccinium myrtillus*, KRAM M-1153, duplicate in MA-Fungi; [18], on fallen twigs of *Picea abies* and *Abies alba*, beechnut husks and mosses, KRA MYXO-522; [19], on beech leaves, grass culms and leaves of *Veronica officinalis*, KRA MYXO-518; KRA MYXO-524; [22], on stems of *Vaccinium myrtillus*, grass culms and needles of *Picea abies*, KRA MYXO-534; KRA MYXO-535; KRA MYXO-537.

COMMENTARY. According to the literature, *D. niveum* forms sporangia larger than (0.8)1 mm in diameter (e.g. Neubert et al. 1995, Moreno et al. 2003b). Some of our collections are characterized by sporangia smaller than 1 mm, which is characteristic of *D. microcarpum* Meyl. (Meylan 1924, Neubert et al. 1995, Moreno et al. 2003b). According to Meylan (1924), the capillitium of *D. microcarpum* should have pale colour similar to that of *D. globosum*. We measured sporangia of all our collections (10-20 measurements per collection). In only one collection (KRAM M-1207) were all sporangia smaller than 1 mm (0.66–0.94 mm) and the capillitium was pale macroscopically (but rigid and brown in transmitted light). All other collections have sporangia both smaller than 1 mm and bigger than 1 mm (on average 0.61–2.04) and dark, rigid capillitium. We identified all our specimens as *D. niveum*, although the collection KRAM M-1207 might be placed in *D. microcarpum*. Collections of *D. niveum* from the herbarium KRA mentioned above were previously published under the name *D. alpinum* (Drozdowicz 1985).

****Didymium dubium*** Rostaf.

SPECIMENS EXAMINED. [2], on stems of *Rubus* sp., KRAM M-1128; KRAM M-1205; [4], on stems of *Rubus* sp., KRAM M-1126; [5], on stems of *Rubus* sp., KRAM M-1127; [14], on plant stems, KRAM M-1135.

****Lamproderma aeneum*** Mar. Mey. & Poulain

SPECIMENS EXAMINED. [5], on stems of *Rubus* sp., KRAM M-1211, duplicate in herb. Marianne Meyer; [21], on a fern petiole, KRA MYXO-527. [9], on stems of *Rubus* sp. KRAM M-1261, duplicate in herb. Marianne Meyer.

COMMENTARY. Our specimens are characterized by slightly larger spores (10–12 µm) than stated in the original description, however, the variation of spore size for all the collections examined by the authors of the species (Poulain et al. 2002a) is also larger than in the type specimen.

***Lamproderma carestiae*** (Ces. & De Not.) Meyl.

= *Lamproderma atrosporum* Meyl.

SPECIMENS EXAMINED. [2], on stems of *Rubus* sp., KRAM M-1230; [3], on stems of *Rubus* sp., KRAM M-1269; [4], on stems of *Rubus* sp., KRAM M-1218; KRAM M-

1248; together with *L. ovoideum* – KRAM M-1216 and KRAM M-1247; [5], on stems of *Rubus* sp., KRAM M-1226; KRAM M-1227; KRAM M-1228; [8], on stems of *Rubus* sp., KRAM M-1242, duplicate in MA-Fungi; [9], on stems of *Vaccinium myrtillus* or plant remnants, KRAM M-1237; KRAM M-1238; [10], on stems of *Rubus* sp. or *Vaccinium myrtillus*, KRAM M-1234; KRAM M-1235; [11], on stems of *Rubus* sp., KRAM M-1241; [12], on stems of *Vaccinium myrtillus*, KRAM M-1232; [14], on stems of *Rubus* sp. and *Vaccinium myrtillus*, KRAM M-1267 (together with *L. ovoideum*); [22], on stems of *Vaccinium myrtillus*, KRA MYXO-538.

COMMENTARY. The sporangia in all of our specimens are stipitate or, rarely, substipitate. Three forms are present in our material, differing from each other by the size of spores and pattern of ornamentation. One group is characterized by large spores, (12)13–17(19) µm, covered with isolated, regularly arranged spines about 1 µm long, the second form has smaller spores, 10–12(13) µm in diameter, covered with 0.5–1 µm long spines arranged in a (sub)reticulate pattern and the third form has smaller spores (10.5–12 µm) covered with irregularly arranged long spines (about 1 µm long), which tend to merge. One specimen (KRAM M-1237) has ornamentation, consisting of spines tending to fuse in groups. The great variability in spore size and ornamentation in *L. carestiae* is known and many different patterns (warts isolated, regularly arranged, irregularly arranged, forming subreticulate pattern) can be found (e.g. Neubert et al. 2000) in small-spored as well as large-spored and sessile as well as stipitate collections. In such a wide range of variability there are two possible interpretations: either distinguishing many species (or lower taxa) with a very narrow variation, or recognition of only one very variable taxon. Because in our opinion myxomycetes, especially nivicolous taxa, are very variable due to unstable environmental conditions in their habitats, we prefer to treat species in a wide sense and therefore we consider the variability in *L. carestiae* as rather continuous and prefer to treat it as one very variable species. Poulain et al. (2002b), who proposed the new generic name *Meriderma* for the outstanding complex species of *L. carestiae* (as "complexe «*atrosporum*»") recognize several taxa on a species level. Apart from *L. carestiae*, we consider *L. cribrarioides* a distinct taxon at species rank due to its distinct genetic position (Martin et al. 2003).

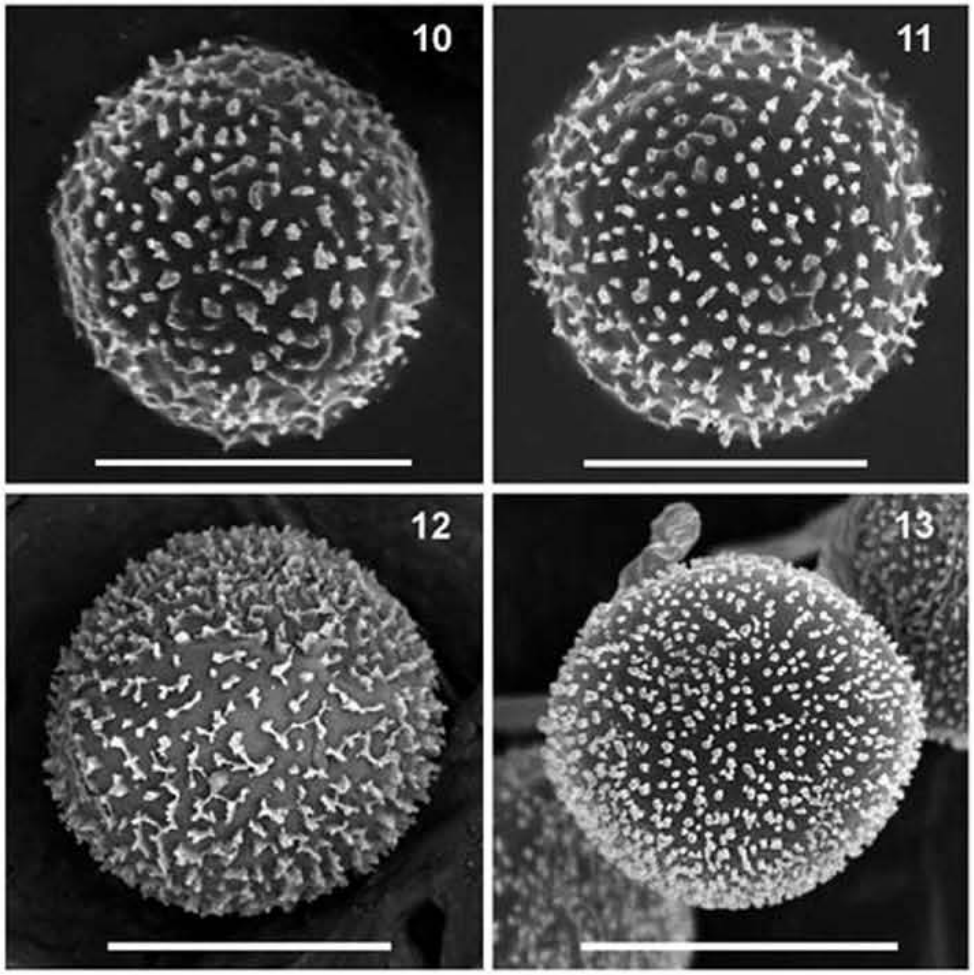
* ***Lamproderma cribrarioides*** (Fr.) R.E. Fr.

= *Lamproderma atrosporum* var. *pseudocribrarioides* Mar. Mey. et al.

SPECIMENS EXAMINED. [9], on stems of *Vaccinium myrtillus*, KRAM M-1214.

* ***Lamproderma ovoideoechinulatum*** Mar. Mey. & Poulain FIG. 10–12

SPECIMENS EXAMINED. [4], on stems of *Rubus* sp., KRAM M-1217; KRAM M-1288; [5], on stems of *Rubus* sp., KRAM M-1254; KRAM M-1222, duplicate in herb. Marianne Meyer; [7] on stems of *Rubus* sp., KRAM M-1287, duplicate in herb. Marianne Meyer; [9], on plant remnants, KRAM M-1236, duplicate in MA-Fungi and herb. Marianne




Figs. 10-12. spores of *Lamproderma ovoideoechinulatum*. 10. coll. KRAM M-1287. 11. coll. KRAM M-1233. 12. coll. KRAM M-1243. 13. a single spore of *Lamproderma ovoideum*, coll. KRAM M-1219. Bars = 10 µm.

Meyer; [10], on grass and *Carex*, on stems of *Vaccinium myrtillus* and other plant remnants, on a stem and needles of a young spruce, KRAM M-1262, duplicate in herb. Marianne Meyer; KRAM M-1263, duplicate in herb. Marianne Meyer; KRAM M-1264, duplicate in herb. Marianne Meyer; KRAM M-1233, duplicate in MA-Fungi and herb. Marianne Meyer; [14], on stems of *Vaccinium myrtillus*, KRAM M-1243, duplicate in MA-Fungi and herb. Marianne Meyer.

COMMENTARY. We identified this species on the basis of its spore ornamentation, which is in form of spines more than 0.5 µm long, longer and more clearly visible in light microscope than are those of *L. ovoideum*. Under SEM the spore ornamentation of *L. ovoideoechinulatum* was slightly different from that of *L. ovoideum*. The lectotype of *L. ovoideum* has spores covered with regularly

arranged, isolated spines (Moreno et al. 2002). This feature was also observed in our collections of *L. ovoideum* (Fig. 13), while the spores of our collections of *L. ovoideoechinulatum* are covered with irregularly arranged spines tending to fuse into ridges or even a subreticulate pattern (Fig. 10-12). According to the original description (Poulain & Meyer 2005), *L. ovoideoechinulatum* is also characterized by a pyriform shape of the sporangia, but this feature was not well visible in our collections.

***Lamproderma ovoideum*** Meyl. FIG. 13

SPECIMENS EXAMINED. [1], on stems of *Rubus* sp., KRAM M-1249; KRAM M-1250; [3], on stems of *Rubus* sp. and other plant remnants, KRAM M-1219, duplicate in herb. Marianne Meyer; KRAM M-1229, duplicate in herb. Marianne Meyer; KRAM M-1251; KRAM M-1252; [4], on stems of *Rubus* sp., KRAM M-1216, duplicate in MA-Fungi; KRAM M-1245; KRAM M-1246; KRAM M-1247; [5], on stems of *Rubus* sp. or on plant remnants, KRAM M-1221; KRAM M-1253; [6], on plant remnants, KRAM M-1255; KRAM M-1256; [9], on plant remnants, KRAM M-1257, duplicate in MA-Fungi; KRAM M-1258; [10], on stems of *Rubus* sp., KRAM M-1265, duplicate in herb. Marianne Meyer; [12], on stems of *Rubus* sp. or plant remnants, KRAM M-1212, duplicate in herb. Marianne Meyer; KRAM M-1231, duplicate in herb. Marianne Meyer; [14], on stems of *Rubus* sp. or *Vaccinium myrtillus*, KRAM M-1267; KRAM M-1268; [20], on plant remnants, KRAM M-1244, duplicate in herb. Marianne Meyer; [21], on a fern petiole, KRA MYXO-536; [22], on fern petioles or stems of *Vaccinium myrtillus*, KRA MYXO-528; KRA MYXO-529; KRA MYXO-530; KRA MYXO-532.

COMMENTARY. We distinguished *L. ovoideum* from *L. ovoideoechinulatum* on account of spore ornamentation, which is in the form of usually low, isolated spines in *L. ovoideum* (Fig. 13) (see remarks under *L. ovoideoechinulatum* above).

****Lamproderma pulveratum*** Mar. Mey. & Poulain

SPECIMENS EXAMINED. [5], on stems of *Rubus* sp., grass leaves and other plant remnants, KRAM M-1224, duplicate in herb. Marianne Meyer; KRAM M-1223, duplicate in MA-Fungi and herb. Marianne Meyer; KRAM M-1225, duplicate in herb. Marianne Meyer; [9], on stems of *Rubus* sp. or *Vaccinium myrtillus*, KRAM M-1260, duplicate in herb. Marianne Meyer; [11], on stems of *Rubus* sp. or *Vaccinium myrtillus*, KRAM M-1239, duplicate in herb. Marianne Meyer; KRAM M-1240, duplicate in herb. Marianne Meyer; KRAM M-1266.

COMMENTARY. Some of our collections possess all characteristic features of *L. pulveratum* (presence of lime crystals in peridium, "rough" peridium surface, dark, well visible warts covering spores), while in other collections, some of those features are lacking. The collections lacking crystals in peridium and possessing more delicately warted spores were identified by us as *L.* cf. *pulveratum*, as they seem to have an unclear position in the group of small-spored nivicolous species of *Lamproderma*.

***Lamproderma sauteri*** Rostaf.

SPECIMENS EXAMINED. [11], on remnants of *Centaurea* sp., KRAM M-1213, duplicate in MA-Fungi; [22], on grass culms, KRA MYXO-531.

****Lamproderma spinulosporum*** Mar. Mey. et al.

SPECIMENS EXAMINED. [22], on grass culms, KRA MYXO-533.

***Lepidoderma aggregatum*** Kowalski

SPECIMENS EXAMINED. [13], on stems of *Vaccinium myrtillus*, KRAM M-1148; [17], on stems of *Vaccinium myrtillus* and *Veronica officinalis*, on grass culms, KRA MYXO-502; KRA MYXO-514.

COMMENTARY. Based on the study of the type collection of *Lepidoderma aggregatum*, Moreno et al. (2004) concluded that this species hardly differs from *L. chailletii* and they synonymized both species. We could find in our material two distinct morphotypes. The majority of collections were characterized by a single peridium covered with whitish-greyish or whitish-ochraceous lime scales, giving the rough appearance of the surface. These specimens represent in our opinion *L. chailletii* (see below). The second morphotype was characterized by a double peridium. The outer layer, very distinctly separated from the inner one, was densely covered by very small crystals forming a continuous, shining crust. These specimens represent in our opinion *L. aggregatum*.

****Lepidoderma chailletii*** Rostaf.

SPECIMENS EXAMINED. [2], on stems and leaves of *Rubus* sp., leaves of *Fagus sylvatica* KRAM M-1147, duplicate in MA-Fungi; KRAM M-1199; [4], on stems of *Rubus* sp., KRAM M-1142, KRAM M-1143; [5], on stems of *Rubus* sp. or *Hypericum maculatum*, culms of grasses, beech leaves, other plant remnants, KRAM M-1144; KRAM M-1145, duplicate in MA-Fungi and herb. Marianne Meyer; KRAM M-1146; [6], on a twig, on stems of *Vaccinium myrtillus* and grass culms, KRAM M-1141; KRAM M-1204; [7], on stems of *Rubus* sp., KRAM M-1197; [8], on stems of *Rubus* sp., KRAM M-1196; [9], on stems of *Vaccinium myrtillus*, KRAM M-1198; KRAM M-1201; [10], on stems of *Vaccinium myrtillus*, KRAM M-1202; [14], on stems of *Vaccinium myrtillus* and other plants, KRAM M-1195; KRAM M-1200; [17], on stems of *Vaccinium myrtillus* and *Veronica officinalis*, on grass culms and beech leaves, KRA MYXO-503; KRA MYXO-504; KRA MYXO-505; KRA MYXO-506; KRA MYXO-507; KRA MYXO-508; [19], on a stone, on grass culms, stems and leaves of *Veronica officinalis*, KRA MYXO-513; on grass culms, KRA MYXO-512; KRA MYXO-511; on grass culms, leaves of beech, stems and leaves of *Veronica officinalis*, KRA MYXO-510; KRA MYXO-509; KRA MYXO-539.

COMMENTARY. Some collections from the locality [19] were previously published under the name *L. carestianum* (Rabenh.) Rostaf. (Drozdowicz 1985).

***Physarum albescens*** Ellis ex T. Macbr.

SPECIMENS EXAMINED. [9], on stems of *Vaccinium myrtillus*, KRAM M-1131, duplicate in herb. Marianne Meyer; [13], on stems of *Vaccinium myrtillus*, KRAM M-1132; [14], on stems of *Vaccinium myrtillus*, KRAM M-1136, duplicate in MA-Fungi; KRAM M-1140.

**Physarum alpestre* Mitchel et al.

Specimens examined. [2], stems and leaves of *Rubus* sp., beech leaves, KRAM M-1130, duplicate in MA-Fungi; [4], stems of *Rubus* sp., KRAM M-1134; [5], on plant remnants, KRAM M-1129; [10], on grass culms, KRAM M-1133; KRAM M-1138; [14], on plant stems, KRAM M-1137; [16], on grass culms and remnants of *Carlina acaulis*, KRA MYXO-516; [17], on grass culms, KRA MYXO-515.

**Physarum vernum* Sommerf.

Specimens examined. [18], on leaves of beech and stems of *Vaccinium myrtillus*, KRA MYXO-500.

*Trichia alpina* (R.E. Fr.) Meyl.

Specimens examined. [2], on fallen twigs, stems of *Rubus* sp., a leaf of beech, KRAM M-1125, duplicate in MA-Fungi and herb. Marianne Meyer; KRAM M-1139; [8] on stem of *Rubus* sp., KRAM M-1194; [13], on stems of *Vaccinium myrtillus*, KRAM M-1155; [15] on fallen twig of *Acer pseudoplatanus*, KRAM M-1193.

Commentary. One collection of *Trichia alpina* (site [15]) is particularly noteworthy from a phenological point of view, as it was made not in spring but in October, before the first snow fall, on a twig of *Acer pseudoplatanus*. The collection is typical, well developed and seems to be fresh, as the short plasmodiocarps are still closed, therefore they were probably formed just before collection, and not persisting from the spring period. Interestingly, also Meylan (1929, 1932) mentions an autumnal collection of this species from the Swiss Jura Mts., also occurring on twigs of *Acer* and *Sorbus*. He did not notice any morphological differences, but remarked that these phenologically distinct collections could represent biologically distinct forms.

## Discussion

A considerable diversity of nivicolous myxomycetes was found in the Gorce Mountains, and almost all collected species were strictly nivicolous. The presence of these organisms was repeatedly connected with a mass occurrence of some species, such as *Diderma niveum*, *Lepidoderma chailletii* or *Lamproderma ovoideum*, which played an important role in the springtime aspect of glades and shrub communities. Thanks to the long-term observations carried out during spring time of six years we could not only obtain a general picture of the diversity of nivicolous myxomycetes of the lower parts of the Carpathians, but also find more rare species noticed during regular, repeated observations. Our results show that nivicolous slime moulds can be found not only in alpine and subalpine zones of high mountains but they can also occur abundantly in low-mountain massifs. The lowest elevation in the present collections was 880 m a.s.l., but snowbank myxomycetes were collected in the Polish

Carpathians even lower. So far, the lowest collection of *Diderma globosum* var. *europaeum* was found at 583 m a.s.l. (K. Nowak, unpublished data). According to our observations, *Diderma alpinum, D. niveum, Lamproderma sauteri, Lepidoderma chailletii, Physarum alpestre* and *P. vernum*, which were mentioned as dominating in the highest elevations of Scottish or Spanish mountains (Ing 1998, Lado 2004), are also present not only in the lowest locations of montane meadows, but also of montane forest in the Gorce mts., and some of them are very common here (*Lepidoderma chailletii* and *D. niveum* were among the most frequently encountered and the most abundantly occurring species).

Most collections from the Gorce Mountains were found by us in open areas such as mountain meadows, but it is noteworthy that we also found several specimens in a loose but regular canopy beech or spruce forest, where no open areas such as glades or meadows were present. It should be noted, however, that during the period of snow melting in spring broadleaved trees are still devoid of leaves, therefore, even in a regular canopy beech forest there is an abundance of light in the forest bottom. The occurrence of nivicolous species at patches of melting snow remaining between trees was also observed by Lado (2004) in the Spanish mountains.

Nivicolous myxomycetes are usually found in close vicinity (up to a few meters) of melting snowbanks (e.g. Stephenson & Johnston 2003), and this was also true in the case of our study. But, interestingly, several of our collections were found also in places devoid of snow cover for a long time. Such places were usually located at the lowest elevations, in regions where even small snow patches were already absent. Very delicate sporangia of slime moulds are usually very easily damaged by wind or rain; therefore, they are most likely to be found just after formation. During rainy springs it is difficult to find any specimens even close to the snowbanks, because they are damaged during transformation from plasmodium to sporangia. Collections made during a dry and sunny spring of 2006 demonstrated that in long periods of favourable weather conditions the sporangia of nivicolous species can persist for a considerable length of time after snow melting.

There are still too few data sets for general conclusions about distribution and ecological preferences of nivicolous species in Europe. Our study shows that the Carpathians, like the Alps and Iberian mountains, harbour diverse communities of nivicolous myxomycetes, confirming the wide distribution of these organisms. A comparison of the low-mountain and high-mountain areas of the Carpathians is being planned to estimate how the ecological conditions at various climatic belts influence the diversity of snowbank slime moulds.

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# A new species of *Pleurocollybia* (*Tricholomataceae; Agaricales; Basidiomycetes*) from Belize

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**Abstract**—A new species, *Pleurocollybia imbricata*, is described from the Maya Mountains of Belize and a new combination in *Pleurocollybia* is proposed. A key to the known species of *Pleurocollybia* is also provided.

**Keywords**—agarics, Doyle's Delight, siderophilous inclusions, taxonomy

## Introduction

*Pleurocollybia* Singer was proposed as a new monotypic genus to accommodate *Gymnopus praemultifolius* Murrill (Murrill 1945) based on several features: eccentric stipe, clampless hyphae, minute basidiospores (very small for an agaric, e.g. "2.7-3.5 × 2.5-3.2 µm"), and lack of necropigments (Singer 1947). Singer (1947) compared *Pleurocollybia* to two morphologically similar genera, *Callistosporium* Singer and *Podabrella* Singer (now considered a synonym of *Termitomyces*, Frøslev et al. 2003), but separated *Pleurocollybia* from them by the eccentric stipe and very small basidiospores. *Podabrella* (= *Termitomyces*)

produces a reddish/pinkish colored spore deposit while those of *Pleurocollybia* and *Callistosporium* are white. *Podabrella* (= *Termitomyces*) also produces siderophilous bodies in the basidia, while siderophilous bodies are not present in *Pleurocollybia*. *Callistosporium* has abundant brightly colored necropigments in the basidiospores, basidia and tramal hyphae, while these pigments are not present in *Pleurocollybia*. A BLAST search based on ITS sequences from *Pleurocollybia imbricata* match closely to several ITS sequences of exemplars of *Callistosporium*. Although not definitive, this similarity in ITS sequences may indicate a close relationship with *Callistosporium* as Singer originally suspected (Singer 1947) and seems to be indicated by a study of the nlsu rDNA region as well since *Callistosporium* and *Pleurocollybia* are found in the same clade, the /callistosporioid clade (Moncalvo et al. 2002).

Since 1947, eight more species have been added to *Pleurocollybia*, including four new species described from the neotropics by Singer (Singer 1955, 1963, 1969, 1978), and 2 new combinations into *Pleurocollybia* from existing neotropical taxa (Singer 1970, 1986). Two additional species were placed in the genus by recombinations of existing taxa, one from Sri Lanka/Africa (Pegler 1977) and one from New Zealand (Horak 1971). With the addition of this newest taxon, and the new combination proposed in this paper, we now know of 11 species that belong in the genus, with the greatest number being New World neotropical taxa (9 species).

*Pleurocollybia* is characterized by a combination of distinctive features: the stipe is typically eccentric or lateral or lacking, only a truly central stipe is known for two species, thus the habit is pleurotoid or collybioid-pleurotoid, the lamellae are crowded and very narrow, adnate or adnexed or emarginate-adnexed, the lamellulae are truncate, the basidiospores are small and white in deposits, smooth, inamyloid, the basidia lack siderophilous/cyanophilous bodies, the basidiospores, basidia and hyphae of the trama lack of necropigments, and the habitat for the basidiomata is typically lignicolous on rotting wood. Clamps may be present or absent.

A key to all known species is provided.

## Materials and methods

Color notations in the macroscopic descriptions are from Kornerup & Wanscher (1978). Methods used in preparation of microscopic structures were those of Baroni (1981). Testing for cyanophilic reactions of spore walls and for cyanophilic bodies in basidia was carried out as follows: un-revived dry lamella fragments were gently heated over a flame in a drop of cotton blue/lactic acid (Singer 1986) on a clean glass slide; when the mountant began to release vapor (not boiled), the fragment was removed and placed in a clear drop of lactic acid at room temperature and washed to remove excess dye. This fragment was finally

transferred to a fresh drop of clear lactic acid at room temperature on a clean slide to make a squash mount. It has previously been shown that siderophilous inclusions in basidia can be determined by using the cotton blue/lactic acid test described above (Baroni 1981), since siderophilous inclusions stain in a similar fashion in cotton blue. All measurements of anatomical features were made in mounts of 3% KOH under an oil immersion lens. The designations used for basidiospore measurements are those of Baroni & Horak (1994) where n = number of spores measured, Q = range of length/width of individual spores and $Q_{m}$ = mean of those Q values. All measurements were made with an Olympus BHS light microscope under Hoffman interference optics using an ocular micrometer or by using a semi-automated image analysis system (a GTCO digitizer pad and Metrics5 software written by Dr. David Malloch). Descriptive statistical analysis of the measurements was obtained using EXCEL 5.0 and SigmaStat 1.0. All illustrations of microscopic features were made with the aid of a drawing tube and the final plates were prepared using a WACOM pen drawing tablet and Adobe Illustrator 10. All longitude/latitude readings listed were made by hand held GPS (GARMIN Etrex Vista) set on the WGS84 Datum standard or the UTM standard.

## Taxonomy

### New species

***Pleurocollybia imbricata*** T.J. Baroni, Lodge & D. L. Lindner, **sp. nov.**

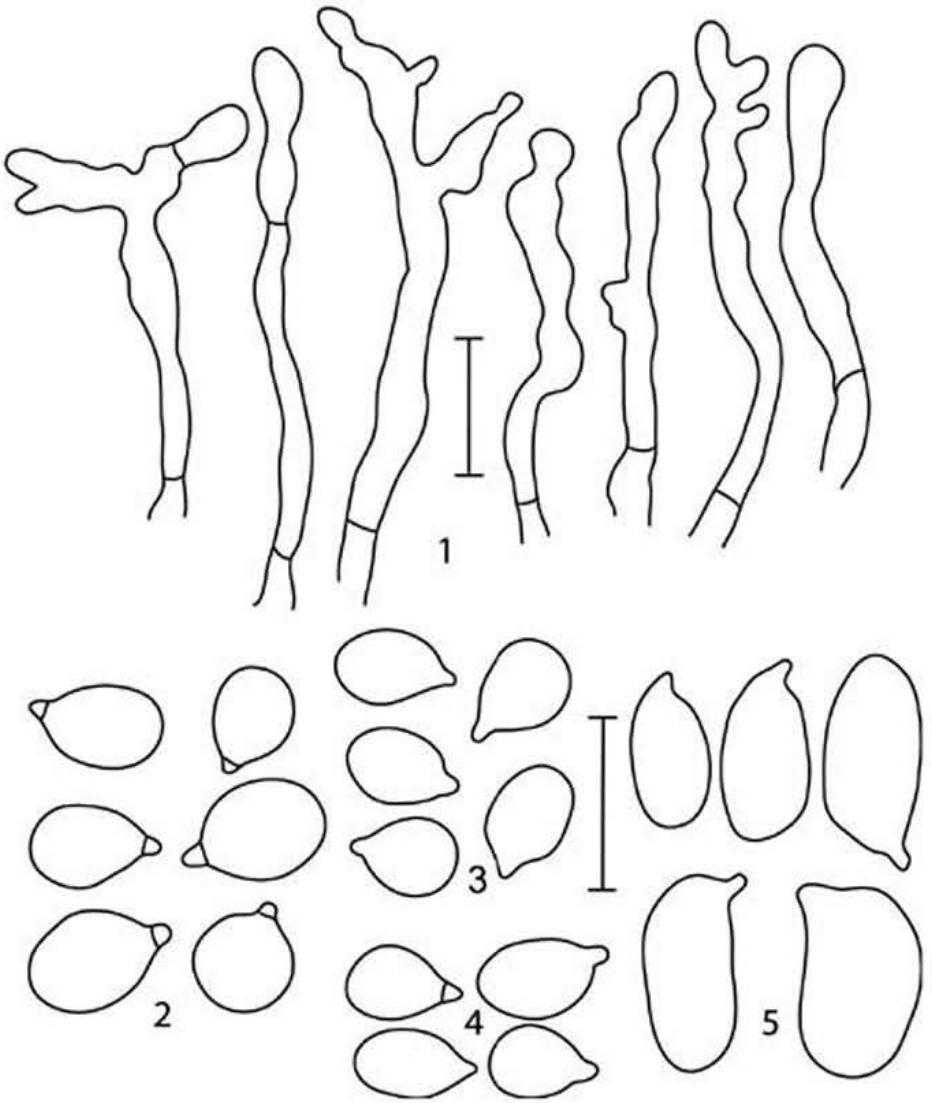
Mycobank MB510698 FIGS. 1-2, 6

*Basidiomata pleurotoidea, imbricata, in subiculo crasso albo implicito, in ligno putrido. Ab congeneribus differt basidiomatibus dense imbricato compaginatis ut videtur stipite carentibus, marginibus pilei profunde incisis lobatisque, pileo incano pallide canido-brunneo, odore farinaceo, sapore amaro farinaceo, basidiosporis parvis subglobosis, 3-4 × 2-3.2 um, cheilocystidiis cylindricis contortis.*

HOLOTYPE T. J. Baroni 9847, Maya Mountains, Belize (BRH; ISOTYPES, CORT, NY)

ETYMOLOGY — From the Latin *imbricatus*. The species is named for its overlapping pilei, like roof tiles.

BASIDIOMATA pleurotoid, densely imbricate and produced from a thick white matted subiculum on decaying woody substrate. PILEUS pale grayish buff or grayish orange or grayish brown (5–6B2–3, Alabaster, Birch Bark, Flesh or 5C2–3, Birch Grey, Brownish Orange), 15–60 mm broad, plano-convex becoming plane (or when young and on top of the woody substrate infundibuliform! – only one basidioma out of 30 or more), irregularly dimidiate, subspathulate, deeply incised, lobed, lobes overlapping, producing small conchate "caps" from margins of lobes, densely imbricate-shelving, hoary canescent over most of surface with darker brown colors below canescence, canescence often grown over or around small bits of woody debris trapping particles on the surface, margin inrolled.




Figs. 1-2: *Pleurocollybia imbricata* (9847 TJ Baroni, Isotype):
Fig. 1, cheilocystidia, scale bar = 10 µm. Fig. 2, basidiospores.
Fig. 3 *Pleurocollybia densifolia* (F.S. Earle 578, Holotype) — basidiospores.
Fig. 4 *Pleurocollybia praemultifolia* (F17468, Holotype) — basidiospores.
Fig. 5. *Pleurocollybia amara* (F17336, Holotype) — basidiospores.
Scale bar for all basidiospores = 5 µm.

Context watery grayish brown, approx. 1 mm thick. LAMELLAE grayish brown (± concolorous with pileus) but also with flesh pinkish hues, adnexed or adnate, extremely crowded with numerous irregular tiers of truncate lamellulae, some so tightly crowded they have grown together, frequently producing ball-like clumps of white pubescent outgrowths on the fused lamella edges, moderately

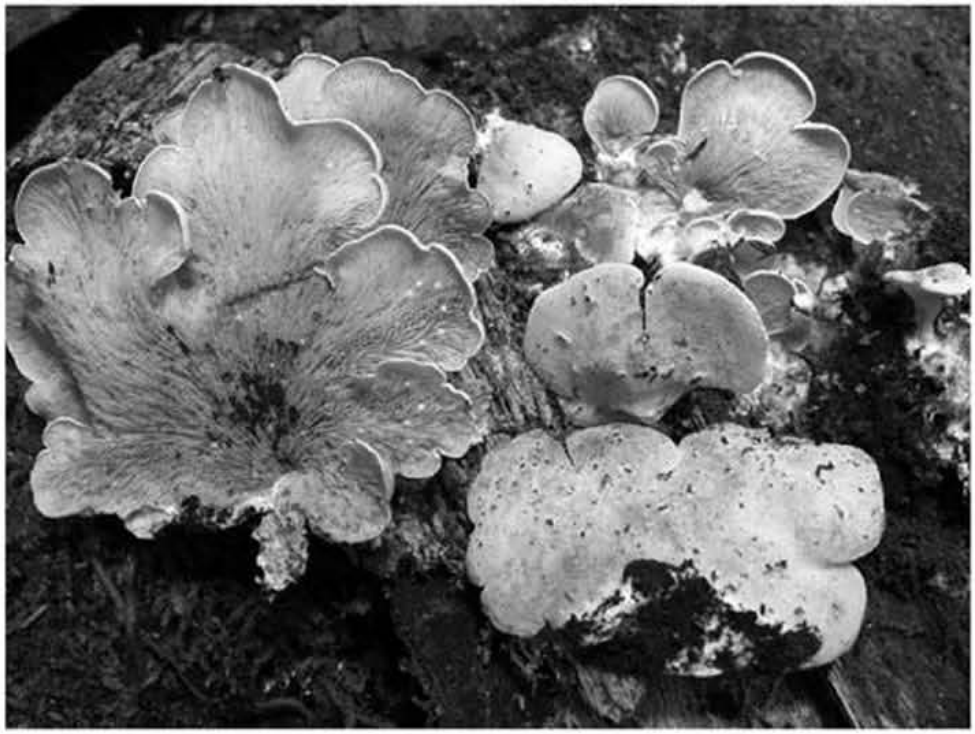


Fig. 6. *Pleurocollybia imbricata* basidiomata in situ. TYPE (9847 T. J. Baroni). 1×.

broad (± 1 mm), edges ± even, concolorous with faces. STIPE central at first but quickly strongly eccentric or lateral and highly reduced, approx. 1 mm × 1 mm, densely white fibrillose appressed; all stipes arising from a dense, thick whitish subiculum covering the woody substrate. ODOR fruity at first or when cut, farinaceous when flesh is crushed. TASTE bitter!! and farinaceous.

BASIDIOSPORES 3–4 × 2–3.2 um, (n=41, $L_m$ = 3.3 ± 0.19, $W_m$ = 2.6 ± 0.32, Q= 1.07–1.3 (–1.6), $Q_m$ = 1.26 ± 0.14), subglobose or broadly ellipsoid, round in polar view, smooth, with a very small apiculus (< 0.5 µm), hyaline, inamyloid, weakly cyanophilic or acyanophilic. BASIDIA 8–12.1 × 4–5.6 µm, 4-sterigmate, broadly clavate, hyaline, lacking cyanophilic bodies. CHEILOCYSTIDIA hyaline, scattered or abundant, cylindrical, frequently contorted, often branched, some subclavate, some septate, (14–) 22–36 × 2.4–3.2 µm. PLEUROCYSTIDIA absent. LAMELLAR TRAMA composed of parallel hyphae, cylindric or slightly inflated, 4–16 um in diam, cells mostly short. PILEUS CONTEXT hyaline, a compact layer of short cylindrical hyphae, 4.8–16.2 um in diam. PILEIPELLIS hyaline, 60–120 um deep, of loosely entangled, erect, cylindrical hyphae, 1.6–4.8 um in diam., mostly trichodermial in aspect, hyphal ends frequently contorted, and often branched. CLAMP CONNECTIONS absent.

HABIT: Lignicolous on downed decaying large (40-60 cm diam) dicotyledenous log, over mosses on the side and underside of the log. August.

MATERIAL EXAMINED: Belize; Cayo District, Maya Mountains, Doyle's Delight UTM 81843W 24593W, 1035 m alt., on north trail to creek from summit, in creek bed area, 13 August 2004, T. J. Baroni 9847 (with Dan Lindner), (HOLOTYPE: BRH; ISOTYPES: NY, CORT).

ADDITIONAL MATERIAL EXAMINED: *Gymnopus praemultifolius*, USA, Florida, Alachua County, Gainesville area, on decaying hardwood log, shade, 7 July 1938, collected and determined W. A. Murrill, F17468 (HOLOTYPE: FLAS ). *Gymnopus densifolius* Murrill, Jamaica, Port Antonio, on a much decayed stump, 23 November 1902, F. S. Earle 578 (HOLOTYPE: NY)

COMMENTS: *Pleurocollybia imbricata* is distinguished from other species in this genus by the lateral obscure stipes arising from a thick whitish subiculum, the deeply incised and frequently lobed margins on the imbricate pilei, the hoary-canescent grayish pileus surface and the conspicuous cheilocystidia (FIGS. 1 & 6). Macroscopically, this taxon may be confused with *P. apoda*, *P. paradoxa*, or *P. praemultifolia*. *P. imbricata* is similar to *P. apoda* and *P. paradoxa* due to the absence or obscurity of stipes at maturity. *P. imbricata* is different from both of these taxa, because it produces at maturity imbricate shelving basidiomata. *P. apoda* and *P. paradoxa* do not produce imbricate shelving basidiomata. In addition, *P. imbricata* has cheilocystidia. *P. apoda* and *P. paradoxa* lack cheilocystidia. *P. imbricata* has a hoary-canescent pileaus surface, while *P. apoda* and *P. paradoxa* have glabrous pileus surfaces.

*Pleurocollybia imbricata* is similar to *P. praemultifolia*, in that, they both have imbricate basidiomata and they produce conspicuous cheilocystidia. However, *P. imbricata* has a pale grayish or grayish brown colored hoary-canescent pileus and the individual basidiomata are produced from a dense whitish subiculum on highly reduced, lateral obscure stipes, while *P. praemultifolia* has a chestnut colored glabrous pileus and the conspicuous stipes are merely eccentric and not arising from a subiculum. The basidiospores of *P. imbricata* are subglobose or broadly ellipsoid (FIG. 2) and are larger than the spores of *P. praemultifolia* (FIG. 4).

## New Combinations

***Pleurocollybia amara*** (Murrill) Singer ex T. J. Baroni & Bocsusis, **comb. nov.**
MYCOBANK MB510957 FIG. 5
BASIONYM: *Gymnopus amarus* Murrill, Proc. Fla. Acad. Sci. 7:109. 1945
≡ *Collybia amara* Murrill, Proc. Fla. Acad. Sci. 7:127. 1945, nom. alt.

Singer (1975, 1986) listed *Pleurocollybia amara* as one of the seven, and then eight, species he accepted in the genus. However, after searching the literature (Mueller & Wu 1997; http://www.speciesfungorum.org/Names/Names.asp), it became obvious to us that Singer never validly published the new combination

he had originally discussed (Singer 1970). We borrowed the type and the information we obtained is described below. *Gymnopus amarus* belongs in *Pleurocollybia* and we validate that new combination here.

The following description of macroscopic features is from Murrill (1945).

> "Pileus conic-convex, not expanding, cespitose, 2 cm broad; surface dry, smooth, glabrous, avellaneous, the disk tinged with isabelline, margin even, entire, inflexed; context thin, gray, opaque, bitter at once, odorless; lamellae adnate, inserted, narrow, close, white, unchanging, entire to eroded; spores ellipsoid, smooth, hyaline, uniguttulate, 5–6 × 3–4 μ; stipe tapering upward, smooth, glabrous, white, 3 × 0.3–0.6 cm. Type collected by West and Murrill on a much-decayed pine log in Sugarfoot Hammock, near Gainesville, Fla., August 4, 1938 (F17336). Gray above and white below, with bitter flesh."

Microscopic features were obtained from our examination of the holotype:

BASIDIOSPORES 5.6–7.2 × 3–4 μm (n = 10, $L_m$ = 6.27 ± 0.57, $W_m$ = 3.37 ± 0.35, Q = 1.75–2, $Q_m$ 1.86 ± 0.10), ellipsoid in face and profile, round in polar view, smooth, hyaline, inamyloid, acyanophilous. BASIDIA 19.4–24.3 × 5.6–6.4 μm, 4-sterigmate, clavate, hyaline, lacking cyanophilous bodies. HYMENIAL CYSTIDIA absent. CLAMPS present on hyphae in hymenium.

HABITAT: Lignicolous on well decayed pine log. August.

> Material examined: USA, Florida, Alachua County, Gainesville area, Sugarfoot Hammock, 4 August 1938, West and Murrill F17336 (HOLOTYPE: FLAS)

COMMENTS: The small, inamyloid, acyanophilous, smooth basidiospores, crowded, narrow lamellae, truncate lamellulae (?), lack of necropigments, lack of cyanophilous bodies in the basidia, bitter taste and habitat on decaying wood indicate this species belongs in *Pleurocollybia* even though the stipe is not conspicuously and consistently eccentric (Singer 1970). Singer (1986) clearly accepted this species in *Pleurocollybia* but never formally made the combination he considered some time earlier (Singer 1970). He did not make the combination earlier because this species was one of just a few taxa of *Pleurocollybia* that possessed clamp connections on its hyphae and did not show clearly eccentric stipe attachments. Since this taxon is not commonly collected (we only know of the type collection), it has not been the subject of a molecular phylogenetic analysis as yet.

Unfortunately the holotype of *G. amarus* consists of only broken fragments and these are not plentiful nor in very good condition. One can determine that the lamellae are crowded and narrow, but that is all. Most of the remaining lamellae and lamellulae on the pileus fragments have been shattered at the pileus connection so that few whole structures are left. It is not possible to determine if the lamellulae where truncate on the few pileus fragments remaining. However, Singer (1970) indicates that *Gymnopus amarus* has the typical truncate lamellulae.

Table 1 – Abbreviated bibliographic and distribution information for species of *Pleurocollybia*.

| Taxon | published as *Pleurocollybia* | Distribution |
|---|---|---|
| *P. amara* | (2008) This publication | Florida |
| *P. apoda* Singer | 1955 Mycologia 47: 769 | Panama |
| *P. brunescens* | 1973 Beih. Sydowia 7: 17 | Cuba |
| *P. cibaria* Singer | 1963 Bol. Soc. Argent. 10: 207 | Peru |
| *P. cremea* (G. Stev.) E. Horak | 1971 N.Z. J. Bot. 9: 415 | New Zealand |
| *P. densifolia* (Murrill) Singer | 1986 Agar. In Mod. Tax., Ed.4, p. 280 | Jamaica |
| *P. imbricata* | (2008) This publication | Florida |
| *P. paradoxa* Singer | 1969 Beih. Nova Hedwigia 29: 55 | Chile |
| *P. praemultifolia* (Murrill) Singer | 1947. Mycologia 39(1): 80 | Florida |
| *P. pulcherrima* Singer | 1978 Nova Hedwigia 29: 12 | Colombia |
| *P. versiformis* (Berk.) Pegler | 1977 Kew Bull. Add. Ser. VI: 96 | Sri Lanka (Type) & Tanzania |

## Clarifications

***Pleurocollybia brunescens*** (Earle) Singer,
[as *brunnescens*] *Beih. Sydowia* 7: 17 (1973)
■ *Geopetalum brunescens* Earle, 1906, In. An. Est. Cent. Agr. de Cuba 1:235, non
[= '*Micromphale brunnescens* Earle', sensu Dennis 1953, lapsus calami].

In the process of obtaining information on all known taxa of *Pleurocollybia*, we discovered misinformation in the published literature that had been followed by several authors and was cited in Index Fungorum incorrectly. [The information in Index Fungorum has since been corrected after we pointed out the problems.] The basionym for *Geopetalum brunescens* Earle was cited incorrectly in the literature as a *Micromphale brunnescens* (sic) Earle by Dennis (1953), Pegler (1977, 1983) and Singer (1973) apparently based on an error in citation by Dennis (1953). There is no *Micromphale brunnescens* described in the literature, by Earle or any other author. Pegler (1987) obviously recognized the error by correctly listing the synonyms of *Pleurocollybia brunnescens* (sic) as *Geopetalum brunnescens* (sic) but incorrectly maintaining the spelling of the specific epithet as *brunnescens* instead of the original and perfectly correct *brunescens*.

## Key to the known species of *Pleurocollybia*

1. Stipe present and typically eccentric or lateral at maturity, rarely central ..... 2
1. Stipe absent or obscure at maturity ..... 9
2. Pileus ± 4 mm broad when dried; stipe 11 × 0.4 mm; pileus, stipe and basal mycelium violet colored; spores 3–4.5 × 2.5–3 µm (Singer 1978) ..... *P. pulcherrima*
2. Pileus larger; stipe broader; violet colors absent ..... 3
3. Fruit bodies white or cream color ..... 4
3. Fruit bodies tan or cinnamon-brown or chestnut brown or grayish ..... 5
4. Pileus 20-100 mm broad, white, spathulate, translucent, non-striate; spores 5.5–8 × 3.5–4.5 µm (Pegler 1977) ..... *P. versiformis*
4. Pileus 15–35 mm broad, cream-white or deep cream color, orbicular or reniform; spores 2.5–3.5 × 2.5 µm, weakly amyloid (Segedin 1996!) ..... *P. cremea*
5. Pileus 10–15 mm broad, cinnamon-brown; stipe pallid, 10 mm × 2 mm, eccentric with maturity, pruinose especially above; spores globose or subglobose, (4–5 µm Murrill 1916; 2.3–2.8 × 1.7–2.2 µm short ellipsoid, Singer 1970; 3.2–4 × 2.4–3.2 µm, ! from Holotype, Fig. 3) ..... *P. densifolia*
5. Pileus larger than 15 mm ..... 6
6. Pileus spathulate, pale grayish brown; stipe lateral, white, silky; spores globose 3-4 µm (Earle 1906) ..... *P. brunescens*
6. Pileus orbicular; stipe eccentric or central ..... 7
7. Gregarious on soil among moss; stipe central; taste mild ..... *P. cibaria*
7. Lignicolous; taste bitter ..... 8
8. Basidiomata densely imbricate; pileus livid chestnut, 35-45 mm broad, odor of anise, taste bitter; stipe eccentric, pruinose, pale yellowish-white; cheilocystidia present, cylindrical-contorted, narrowly clavate, some branched; spores 1.5–2.0 µm (Murrill 1945), but 2.8–3.2 × 2.4–2.8 µm (! from Holotype) ..... *P. praemultifolia*
8. Basidiomata not imbricate; pileus avellaneous or disc isabelline, 20 mm broad, odorless, taste bitter; stipe central, glabrous, white; cheilocystidia absent; spores ellipsoid, 5-6 × 3–4 µm (Murrill 1945; 5.6–7.2 × 3–4 µm TYPE!) ..... *P. amara*
9. Basidiomata imbricate shelving; pileus 15–60 mm broad, margins deeply incised and frequently lobed, pale-grayish buff or grayish-brown, surface hoary-canescent; cheilocystidia present; spores 3–4 × 2–3.2 µm, subglobose or ellipsoid ..... *P. imbricata*
9. Basidiomata not imbricate; pileus margin not incised or lobed, surface glabrous; cheilocystidia absent ..... 10
10. Pileus up to 42 mm broad, pale ochraceous; spores 2.7–3.0 × 1.8–2.0 µm, ellipsoid ..... *P. apoda*
10. Pileus up to 11 mm broad, pale cinnamon brown; spores 3–5 × 2–2.5 µm, ellipsoid ..... *P. paradoxa*

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# A distinct species of *Cordyceps* on coleopterous larvae hidden in twigs

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**Abstract** — A new distinct caterpillar fungus *Cordyceps neosuperficialis* is formally described. Its ascomata grow on coleopterous larvae hidden in small woody twigs and possess obviously superficial perithecia. It was discovered in the Dinghushan Biosphere Reserve, Guangdong Province, China. The holotype (GDGM 24809) is deposited in the Herbarium of Guangdong Institute of Microbiology (GDGM), Guangzhou.

**Key words** — *Ascomycetes, Clavicipitaceae, Coleoptera,* new species

## Introduction

More than 400 species of *Cordyceps* have been described (Kobayasi & Shimizu 1983, Ito & Hirano 1996). At least 130 species, varieties and formae have been reported in China, and 32 taxa recorded in Guangdong province (Bi et al. 1993, Song et al. 2006). Recently, the authors re-examined some of the materials from Guangdong and discovered that the collections previously labeled as '*Cordyceps superficialis*' from Dinghushan Biosphere Reserve of Guangdong are not completely identical to that American species and are different from any other known taxa. Some new specimens were collected to make further observation and a new species is proposed as follows.

## Materials and methods

The description of the new species was based on the examination of the specimens. Herbarium abbreviation follows the Index Herbariorum: GDGM

* corresponding author.

represents the Herbarium of Guangdong Institute of Microbiology. The holotype was deposited in GDGM. The photographs were taken from the holotype. The colour description was according to Kornerup & Wanscher (1978). Tissues were mounted in 5% KOH for microscopic examination. Authors' abbreviations were according to Kirk & Ansell (1992).

## Taxonomy

***Cordyceps neosuperficialis*** T.H. Li, Chun.Y. Deng & B. Song, **sp. nov.** **Figs. 1-4**

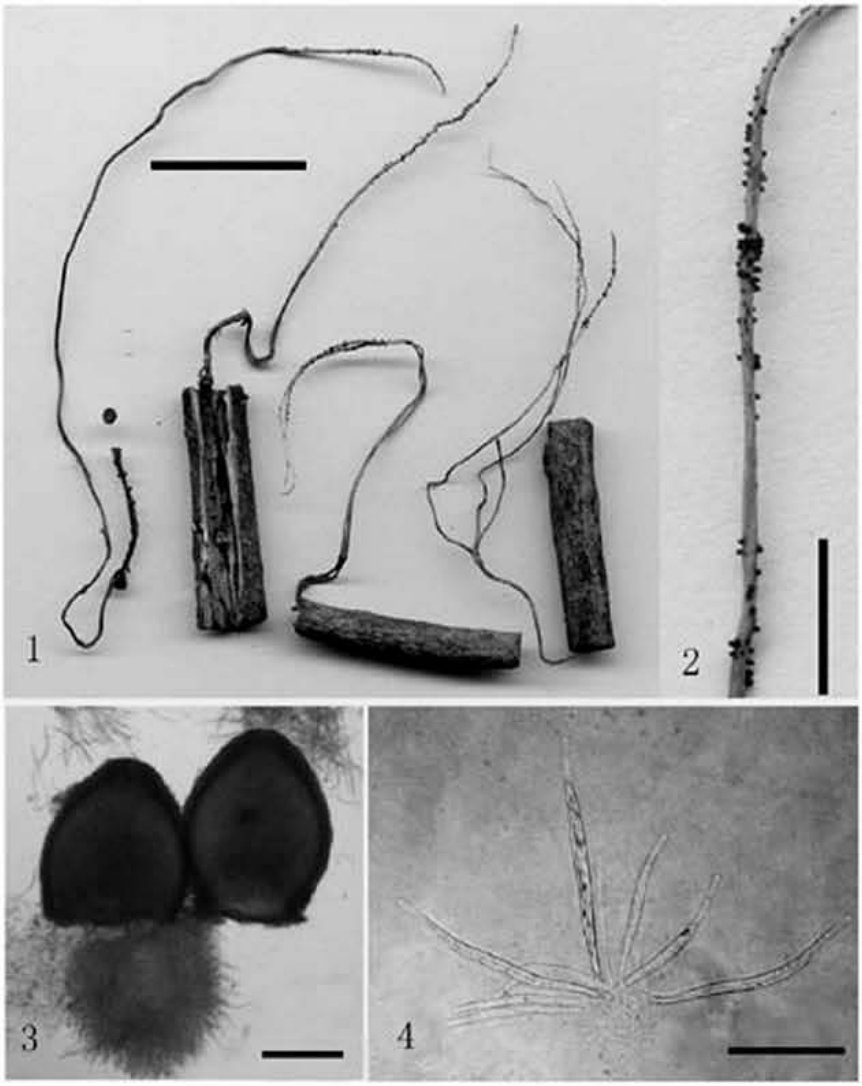
MycoBank MB 510989

=*Cordyceps superficialis* sensu Bi et al (1983: 118;1993:12);
non *C. superficialis* (Peck) Sacc., Syll. Fung. 2: 574, 1883.

*Stroma ex larvo Coleoptero 1.7-2.5 cm longo et 2-2.5 mm crasso, saepe flexuosum, simplicium vel ramosum, exilis, 6-12 cm longum, 0.5-1.5 mm crassum, dilute aurantiacum, griseo-aurantiacum vel brunneolo-aurantiacum, juvenili apice sterili canum vel subalbidum, perithecis numerosis.* Perithecia *fusca, dense superficialia, ovoidea vel subconica, 220-550 × 220-450 µm.* Asci *helminthoidei, peranguste cylindrici vel filiformes, 145-210 × 4-6 µm, hyalini, terminatione capitulo perlucidulo tumidulo, 8-sporis.* Ascosporae *filiformes, 140-180 × 0.8-1.1 µm, hyalinae, multiseptatae, secedentes, in partisporas 4.5-7.5 µm longas.*

Stroma arising from the head or two ends of host-insects, unbranched to branched, slender, long cylindrical, up to 6-12 cm long, 0.5-1.5 mm thick, usually flexuous, upper portion ashy or pale grey (1B1) when young (perithecia not yet borne), lower and mature portion pale orange, greyish orange to orange brown (5A3, 5B4 to 6B4), colour practically unchanged when dried; fertile portion not swollen, not clearly delimited from sterile portion, up to 6 cm long, gradually tapering upwards, apiculate at terminal, with or without a sterile apex, often with smaller perithecia near apex. Perithecia superficial on superior periphery of stroma, gregarious to caespitose, ovoid to subconical, 220-550× 220-450 µm, usually 400-550 × 400-450 µm when matured, brown, dark orange brown to dark brown (6E5, 6F6 to 6F5), perithecial ostiola slightly exserted, from which spouting white ascospores when matured and humid. Asci helminthoid to narrowly cylindrical or filiform, 145-210 × 4-6 µm, with a transparent crown at apex, acuminate at posterior base, walls gradually broken at maturity, with eight ascospores denuded. Ascospores filiform, slightly shorter than asci, 140-180 × 0.8-1.1 µm, multiseptate, breaking up into 4.5-7.5 µm long part-spores.

Habitat, distribution — On the larvae of *Coleoptera*. The larvae are up to 1.7-2.5 cm long, 2-2.5 mm across, hidden in piths of fallen twigs which were cut off by the insects into small segments measured 2.5-3.5 × 0.4-0.8 cm, usually covered with fallen small branches and leaves in broad-leaf forest. China (Guangdong).




**Figs. 1-4:** *Cordyceps neosuperficialis* (holotype).
1. Ascomata. 2-3. Perithecia. 4. Asci and ascospores.
Bars: 1=20mm; 2=10mm; 3=200μm; 4=100μm

Collections Examined — CHINA, Guangdong, Dinghushan Biosphere Reserve. 10 May 2007, Li, T. H. & Deng, C. Y. (GDGM 24809; holotype designated here); 27 April 1981, Bi, Z.S. & Li, T. H. (GDGM 4568); 14 May 1981, Bi, Z.S. (GDGM 4811); 23 April 1982, Bi, Z.S. & Li, T. H. (GDGM 6621).

Comments—According to the taxonomic system of Kobayasi (1982), the new species should be placed in Section *Racemella* under Subgenus *Eucordyceps*. Among the known taxa, four species are quite similar to the new species in the characters of slender stroma and superficial perithecia, and their differences are

pointed out below. As compared with the new species, the American species *C. superficialis* possesses much shorter and darker stroma and obviously larger perithecia and part-spores (Saccardo 1883, Ellis 1892, Shimizu 1997); another American taxon, *C. michiganensis* Mains, has paler perithecia and much longer part-spores (Mains 1934, Shimizu 1997); the Asian caterpillar fungus, *C. agriota* A. Kawam. from Japan, has a acicular stroma and darker blackish perithecia (Kobayasi & Shimizu 1983, Shimizu 1997); the second Asian species, *C. mrciensis* Aung et al. from Thailand arising from spider, produces smaller perithecia, thicker asci, and ascospores that do not break into part-spores (Aung et al. 2006).

The most distinct macroscopic character of the new species is that its host larvae were hidden in piths of fallen twigs, which were cut off by the insects into small segments. Specimens cited above for the new species were originally misidentified as *C. superficialis* (Bi et al. 1983, 1993), but re-examination showed that no specimens completely identical to that American species have been found in China.

## Acknowledgments

The authors thank Dr. Roy Watling of Royal Botanic Garden Edinburgh and Dr. Wen-Ying Zhuang of Institute of Microbiology, Chinese Academy of Sciences for reviewing the manuscript. Thanks are also due to Dr. Zhu L. Yang of Kunming Institute of Botany, Chinese Academy of Sciences for the discussion on the paper. This study was supported by the National Natural Science Foundation of China (No. 30470012; 30470013), the Natural Science Foundation of Guangdong (No. A06020222, E05202480) and Guangdong Key Laboratory of Microbial Culture Collection and Application.

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# *Cordyceps guangdongensis* sp. nov. from China

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**Abstract**—A new species of *Cordyceps* discovered in Guangdong of China, *Cordyceps guangdongensis*, is formally introduced. The fungus grows on *Elaphomyces* sp. in a broadleaf forest. The closest species is *C. japonica*, but these are different in sizes of perithecia and part spores, and with only 93.9% sequence similarity of ITS1-5.8S-ITS2 region. The holotype is deposited in the Herbarium of Microbiology Institute of Guangdong Province.

**Key words**—*Ascomycetes*, *Clavicipitaceae*, mycogenous fungi

## Introduction

*Cordyceps* is a megagenus that contains about 450 species, including 32 species parasitic on hypogeous fungi, mainly *Elaphomyces* (Stensrud et al. 2005, Kobayasi & Shimizu 1983). However, some names are treated as taxonomic synonyms. There are about 130 taxa of *Cordyceps* reported in China, including 7 species and 1 forma associated with *Elaphomyces* (Liang 2007, Song et al. 2006). Guangdong Province, located in the south of China, is rich in *Cordyceps* species. More than 30 species of the genus occurring there have been recorded (Song et al. 2006, Bi et al. 1993). In an investigation of the Erhuangzhang Nature Reserve, Yangchun County, Guangdong Province, collections of a *Cordyceps* species parasitic on *Elaphomyces* sp. was discovered. It turns out to be a new species and is formally described.

## Materials and methods

### Specimens and isolates

Morphological and molecular data were obtained from fresh and dried specimens collected from Erhuangzhang Nature Reserve. Specimens are

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deposited in the Herbarium of Microbiology Institute of Guangdong Province (GDGM). The isolate of *C. japonica* Lloyd (IFO9647) was from the culture collection at the Institute for Fermentation, Osaka, Japan, maintained on PDA medium (potato 200g/L, glucose 20g/L, agar 20g/L). Liquid culture with YMPD medium (glucose 10g/L, polypeptone 5 g/L, yeast extract 3 g/L, malt extract 3g/L ) was performed at 20-25 °C, with a shake speed of 150r/m. Mycelia were harvested, washed with sterile water for 2-3 times, and squeezed to dry for DNA extraction.

### Morphological observations

Ten stromata of the specimens were measured. Color descriptions followed Kornerup & Wanscher (1978). Micro-anatomical characters were studied with light microscopy. 40 perithecia, 40 part spores, and 20 asci were measured. Perithecia, asci and part-spores were respectively measured and pictured at magnifications of ×100, ×400, ×400 or ×1000. Authors' abbreviations were according to Kirk & Ansell (1992).

### DNA extraction, amplification and sequencing

Genomic DNA was extracted from dried specimen and fresh mycelia followed the benzyl chloride method (Zhu et al. 1994). To eliminate the interference of RNA, RNAse (20ng/μl) was added to the DNA solution and bathed at 37°C for 30 min. DNA examples were checked on 1.0% agarose.

The PCR amplification was performed on a T3000 Thermocycler (Biometra, Germany), with a total of 50μl reaction mixture, containing 5μl 10× buffer with 25mM $MgCl_2$, 2μl 10mM dNTPs, 2μl 10mM ITS4, 2μl 10mM ITS5, 0.4μl 5U/μl Taq polymerase, 2μl 50× diluted DNA template and 36.6μl double distilled water. The cyclic thermal program was as follows: an initial 4 min denaturation at 95 °C; 30 cycles of 1 min at 94 °C, 40s at 55 °C, 1.5 min at 72 °C, and termination with a 6 min elongation at 72 °C. A negative control ($ddH_2O$) was included. 5μl product was determined by electrophoresis with 1.0% agarose. Sequencing was finished by Guangzhou Top Genomics, Ltd., with TA clone. Sequences were submitted to GenBank.

### Data analysis

DNA sequences of the new species and *C. japonica* were tested with BLAST in GenBank. According to the sequence similarities, 10 sequences of the closest species were retrieved from GenBank for construction of the consensus tree with *C. militaris* as outgroup. Their Genbank Accession numbers were listed in Table 1. The 11 ITS1-5.8S-ITS2 sequences were selected with BioEdit referring to the sequence of the new species. Preliminary multiple alignments were generated with ClustalX 1.83. The phylogenetic tree was constructed with MEGA3.1 (Kumar et al. 2004). Neighbor-joining method was performed

Table 1. Sequences used in phylogenetic tree construction

| Species | GenBank Accession No |
|---|---|
| *C. capitata* | AJ786557 |
| *C. guangdongensis* | EU039881 |
| *C. inegoensis* | AB027368 |
| *C. japonica* | EU039882 |
| *C. jezoensis* S. Imai | AB027365 |
| *C. longisegmentis* Ginns | AJ786568 |
| *C. militaris* (L.) Link | AJ786569 |
| *C. ophioglossoides* | AJ786586 |
| *C. paradoxa* | AB027369 |
| *C. subsessilis* | AJ786599 |
| *Tolypocladium cylindrosporum* W. Gams | AB208110 |
| *T. inflatum* W. Gams | AJ786592 |

in MEGA3.1, selecting the 'complete deletion' mode to consider deletions. Confidence values for individual branches were determined by bootstrap analysis (1000 replications). Four sequences of ITS1-5.8S-ITS2 were selected based on phylogenetic study and morphological similarity. Their similarity was compared using BioEdit.

## Results

### Taxonomy

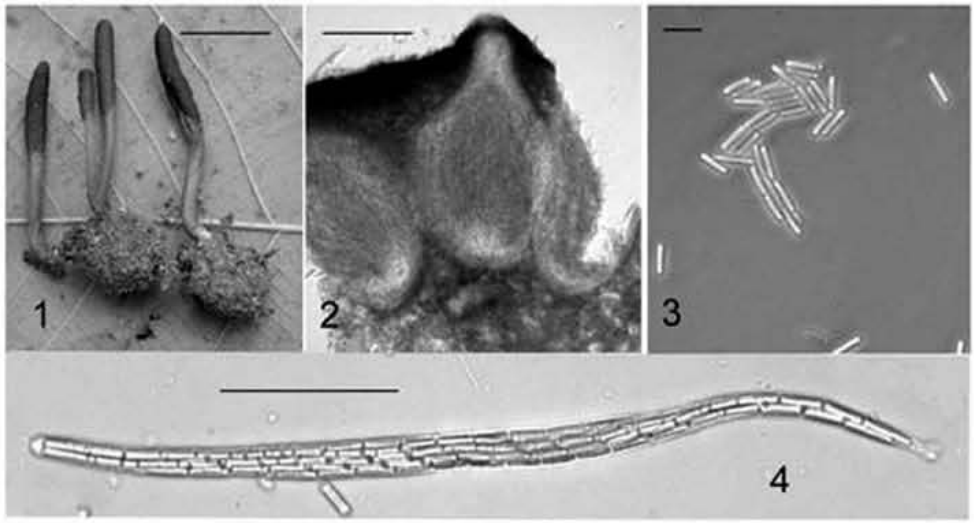
***Cordyceps guangdongensis*** T.H. Li, Q.Y. Lin & B. Song, **sp. nov.** **Figs. 1-4**

MycoBank MB 511126

■ *Cordyceps japonica* f. *guangdongensis* T.H. Li, Q.Y. Lin & B. Song (nom. nudum), in Song et al., Journal of Microbiology 27(1): 60, 2007.

*Stromata solitaria vel plures ex Elaphomycete, simplicia, cylindrica vel clavata, 3-7 cm longa, carnosa. Area fertilis terminalis, cylindrica, 1-3 cm longa, 5-8 mm crassa, olivacea, olivaceo-brunnea vel sepiacea. Stipes cylindricus, 2-4 cm longus, 4-6 mm crassus, olivaceo-luteus, olivaceo-griseus vel griseus.* Perithecia *dense immersia, ellipsoidea vel ovoidea, 245-495 × 160-320 µm.* Asci *helminthoidei, 195-270 × 7-10 µm, 8-ascosporis.* Ascosporae *levis, filiformes, 180-260 × 2.0-3.7 µm, multiseptatae, in partisporas 10-17 µm longas.*

Stromata 1 to 3 in a group arising from fruitbodies of Elaphomyces sp., cylindrical to clavate, 3-7 cm long, tender fleshy; upper fertile portion cylindrical, rounded at top, 1-3 × 0.5-0.8 cm, olive, dark olive, olive brown to golden gray or brownish gray (3E3, 3F3, 4F3 to 4F4), sometimes furrowed at lower part, without a sterile apex; perithecial ostiola punctiform, not protruding from surface of stroma; lower sterile portion 2-4 × 0.4-0.6 cm, color varied from yellowish gray, olive, grayish olive, olive-gray to gray (3C2, 3C3, 3C4 to 3D4), usually paler at base and near fertile portion which is light yellow to grayish




**Figs. 1-4.** *Cordyceps guangdongensis* 1. Stromata (bar = 3cm). 2. Perithecia (bar = 200μm). 3. Part spores (bar= 15μm). 4. Ascus (bar= 50μm). All from holotype.

yellow or dull yellow (3A5, 3B4-3B7) and then becoming yellowish gray, olive gray to gray (3D1, 3D2 to 3D3), with white mycelium around the base and part of substrate. PERITHECIA deeply embedded in stroma, ellipsoid to ovoid, 245-495 × 160-320 μm. ASCI helminthoid, 195-270 × 7-10 μm, with a transparent and slightly swollen crown 3.5-5.0 × 5.0-7.0 μm at apex, acuminate at posterior end, 8-spored. ASCOSPORES 180-260 × 2.0-3.7 μm, filiform, multiseptate, breaking up into part spores; part spores 10-17 μm long, with truncated ends.

HABITAT —scattered to gregarious on *Elaphomyces* sp. buried in soil in broadleaf forest.

SPECIMENS EXAMINED —China, Guangdong Province, Yangchun County, Erhuangzhang Nature Reserve, alt. 50 m, 23 IV 2005, Li T.H., Wu L.M., Chen M.B. & Huang H., (GDGM 24020; holytype); 2 V 2006, alt. 50 m, Li T.H., Lin Q.Y. & Huang H. (GDGM 24851).

## Sequence analysis

The ITS1-5.8S-ITS2 sequences of *C. guangdongensis* and *C. japonica* (GenBank Acc. No. EU039882) had 611 bp and 626 bp respectively. Sequence of *C. japonica* obtained in this study is almost identical with that by Stensrud et al. (2005) (GenBank Acc. No.: AB027366), only with two different base pairs, while *C. guangdongensis* and *C. japonica* are with 36 different base pairs. In analysis of the ITS sequence, the similarity between *C. guangdongensis* and *C. japonica* is 93.9% (Table 2). Although they are closely related as shown in the consensus tree, their genetic distance is the same as that between *C. inegoensis* Kobayasi and *C. paradoxa* Kobayasi, as well as that between *C. subsessilis* Petch and *C. ophioglossoides* (Ehrh.) Link (Fig. 5).

Table 2. Similarity matrix of ITS sequences of four *Cordyceps* species

| Taxa | *C. guangdongensis* | *C. japonica* | *C. jezoensis* | *C. ophioglossoides* |
|---|---|---|---|---|
| *C. guangdongensis* | — | | | |
| *C. japonica* | 93.9% | — | | |
| *C. jezoensis* | 89.3% | 87.4% | — | |
| *C. ophioglossoides* | 73.4% | 71.4% | 77.3% | — |

## Discussion

Among the *Cordyceps* species parasitic on *Elaphomyces* spp., including the well known *C. ophioglossoides* and *C. capitata* (Holmsk.) Link, the most similar species, morphologically, to the new species is *C. japonica*. However, the two species are distinctively different in the size of perithecia and partial spores. Both the perithecia (500-550 × 200-250μm) and the part spores (10-25 × 2.5-3.0μm) of *C. japonica* (Kobayasi &Shimizu 1983) are longer than those of the new species. According to the study based on analysis of ITS sequences from 32 specimens of seven *Cordyceps* species, sequences of the same species from different localities are almost invariable (Stensrud et al. 2005). In this study, the ITS sequences of *C. guangdongensis* and its closest species *C. japonica* have 36 different base pairs. The 93.9% sequence similarity and obvious genetic distance indicate that they are different species.

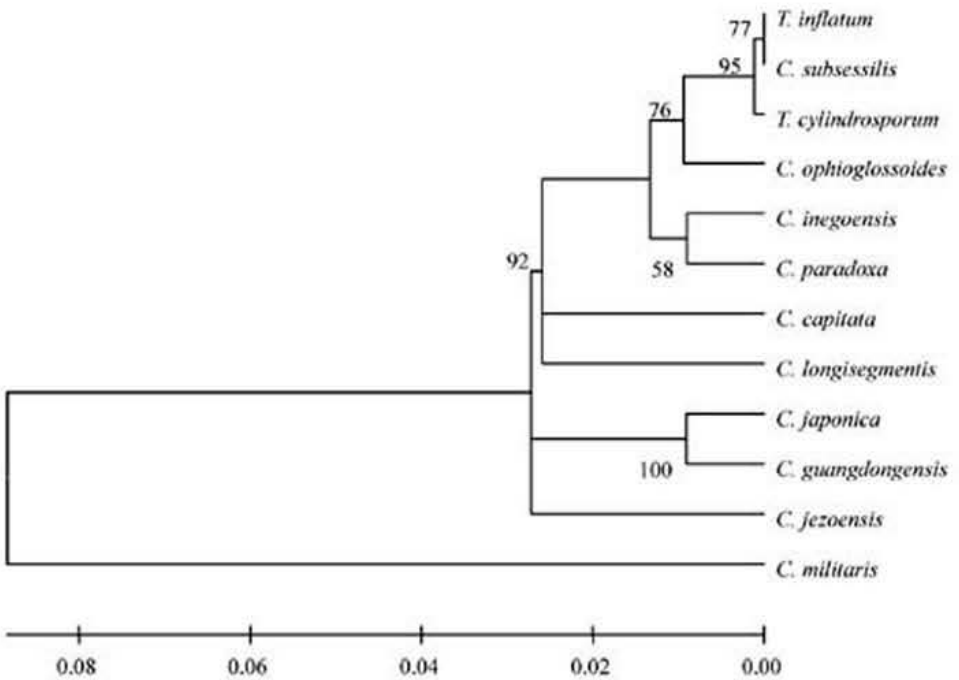



Fig. 5. Consensus tree, based on ITS sequences of 12 taxa of *Cordyceps*.

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# A study of the types and additional materials of *Clitocybe pseudophyllophila* and *Clitocybe subcandicans*

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**Abstract** — The holotypes and additional materials of *Clitocybe pseudophyllophila* and *C. subcandicans*, originally described from China in 1985, were critically restudied. New evidence proved that they should be placed under *Clitopilus* for their pinkish basidiospores with longitudinal ridges. All re-examined specimens labeled as *C. pseudophyllophila* and *C. subcandicans* were identical to *Clitopilus crispus*.

**Key words** — *Agaricales*, taxonomy, revision

## Introduction

Some clitocybeoid species of *Clitopilus* (Fr. ex Rabenh.) P. Kumm. are macroscopically similar to those of *Clitocybe* (Fr.) Staude; however, their basidiospores obviously differ. Basidiospores of *Clitocybe* are smooth, while those of *Clitopilus* are longitudinally ridged (Singer 1986). The two genera can be possibly confused if the characters of basidiospores have not been observed carefully. Recently the authors critically re-examined the holotypes and additional materials of *Clitocybe pseudophyllophila* and *Clitocybe subcandicans* originally described from Guangdong Province of China. The results show that both represent a species of *Clitopilus* with pinkish longitudinally ridged basidiospores. Therefore, detailed description based on the re-examination is presented and the taxonomic status is discussed as follows.

## Materials and methods

The specimens examined are preserved in GDGM (Herbarium of Guangdong Institute of Microbiology, Guangdong Province, China). Herbarium abbreviation follows Holmgren & Holmgren (1998).

*corresponding author

Tissues were mounted in 5% aqueous KOH for microscopic examination. The abbreviation [n/m/p] designates *n* basidiospores measured from *m* basidiomata *in* p collections. Dimensions of basidiospores including the apiculus are given with notation of the form (a)b–c(d). The range b–c contains a minimum of 90% of the measured values with the outliers a and d given in parentheses. Q refers to the basidiospore length/width ratio; **Q** refers to the average Q of all basidiospores ± sample standard deviation. Scanning electron micrographs were produced with a FEI-XL30 scanning electron microscope (SEM) generally running at 10 kev.

## Taxonomy

**1. *Clitocybe pseudophyllophila*** Z.S. Bi & G.Y. Zheng, Guihaia 5: 364, 1985

FIGS. 1–2

BASIDIOMATA (FIG. 1) small to medium in size. PILEUS 2–6 cm broad, convex to applanate, often slightly depressed at centre, white to chalk white; margin incurved, finely subfimbriate, with radial fine ridges extending nearly halfway to the disc. CONTEXT white. LAMELLAE 2–3 mm wide, decurrent, white or cream-coloured to pinkish. STIPE 2–6 × 0.3–0.8 cm, subcylindrical, central to excentric, white, smooth, sometimes finely fibrillose, especially at base.

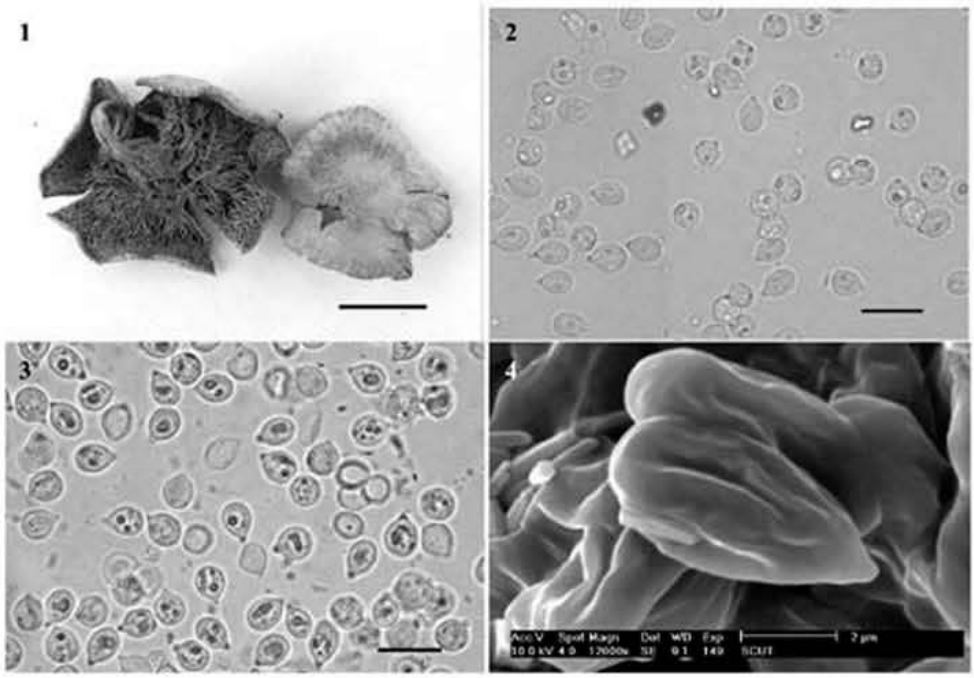
BASIDIOSPORES (FIG. 2) [175/10/5] (5.9–)6.8–8.5(–9.6) × (4.0–)4.5–5.5(–6.0) µm [Q = (1.20–)1.30–1.60(–1.70), **Q** = 1.52 ± 0.13], ovoid, broadly ellipsoid to ellipsoid in profile and face view, angled in polar view with 9–11 facets, longitudinally ridged. BASIDIA 15–24 × 6.4–8 µm, clavate, 4-spored, rarely 1-, 2- or 3-spored; sterigmata about 3 µm long. SUBHYMENIUM consisting of short segments, 2–5 µm wide. HYMENOPHORAL TRAMA more or less regular, with thin-walled hyphae 3–7 µm wide. PLEUROCYSTIDIA and CHEILOCYSTIDIA absent. CLAMP CONNECTION absent.

HABITAT: on soil in broad-leaved forest.

SPECIMENS EXAMINED—CHINA, GUANGDONG PROVINCE, GUANGZHOU, Huanghuagang Park, 5.VIII. 1983, W.L. Zheng [HOLOTYPE, GDGM-6027]; ibid, 2. IV. 1984, T.H. Li [GDGM-6148]; ibid, 2. IV. 1984, Z.S. Bi [GDGM-6309]; ibid, 16. VII. 1984, T.H. Li [GDGM-6340]; Guangzhou Zoological Garden, 26. IV. 1984, T.H. Li [GDGM-6201].

**2. *Clitocybe subcandicans*** Z.S. Bi, Guihaia 5: 365, 1985 FIGS. 3–4

BASIDIOMATA small in size. PILEUS 0.8–2.1(–4) cm broad, convex to applanate, often slightly depressed at centre, white to chalk white; margin incurved, finely subfimbriate, with radial fine ridges extending nearly halfway to the disc. CONTEXT white. LAMELLAE decurrent, white. STIPE 1.2–4.0 × 0.1–0.7 cm, subcylindrical, central to excentric, white, smooth, sometimes finely fibrillose, especially at base.




**Figs. 1-2:** *Clitocybe pseudophyllophila* (holotype). 1. Basidiomata (dry material). 2. Basidiospores in LM; **Figs. 3-4:** *Clitocybe subcandicans* (holotype). 3. Basidiospores in LM. 4. Basidiospores in SEM. Bars: 1 = 1cm; 2 = 10 µm; 3 = 10 µm; 4 = 2 µm

The microscopic characters (Fig. 3–4) of the specimens are practically the same as those of *C. pseudophyllophila*.

HABITAT: on soil in broad-leaved forest.

SPECIMENS EXAMINED—CHINA, GUANGDONG PROVINCE, ZHAOQING COUNTY, Dinghushan Biosphere Reserve, 16.IV. 1981, T.H. Li [HOLOTYPE, GDGM-5478]; ibid, 3. IV. 1981, J.Q. Liang [GDGM-4428]; ibid, 17.VII. 1980, Z.S. Bi [GDGM-4387]; ibid, 8. VIII. 1981, T.H. Li [GDGM-5320].

REMARKS — The longitudinal ridges (Fig. 4) on the basidiospores of *C. pseudophyllophila* and *C. subcandicans* indicate that they represent a species of *Clitopilus*. Their main characters, including the white pileus with radial fine ridges at margin as well as the ovoid, broadly ellipsoid to ellipsoid basidiospores with 9–11 facets and (5.9–)6.8–8.5(–9.6) × (4.0–)4.5–5.5 (–6.0) µm in size, are identical to those of *Clitopilus crispus* Pat. (Patouillard 1913, Baroni & Watling 1999, Yang 2000). Therefore, *C. pseudophyllophila* and *C. subcandicans* should be placed as synonyms with *C. crispus*.

The specimens cited above were misidentified as species of *Clitocybe* because the longitudinal ridges on their basidiospores were overlooked. Bi & Zheng

(1985) considered that *C. pseudophyllophila* was different from *C. subcandicans* in spore print colour, i.e., pinkish in *C. pseudophyllophila* and white in *C. subcandicans*. However, recent observation showed that the basidiospores from type specimen of *C. subcandicans* were pinkish as well. The reason for the mistake might be that the specimens were young and, thus, the pinkish colour of lamellae and spore print was not so obvious with less mature spores.

*C. crispus* was first described from Vietnam (Patouillard 1913). It was reported for the first time from China by Yang (2000). Yang & Zang (2003) considered that the species is a tropical Asian element. While re-examining the specimens of *Clitopilus* in China, the present authors found that it was quite common in tropical Yunnan and Guangdong. There are also some similar species with white to whitish basidiomata occurring in tropical Asian areas, including *C. apalus* (Berk. & Broome) Petch var. *apalus*, *C. amygdaliformis* Zhu L. Yang, and *C. orientalis* T.J. Baroni & Watling. A detailed comparison among them has been made by Yang (2007).

## Acknowledgments

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# Errata

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| | |
|---|---|
| p.150, lines 8–9 | for: *P. dysthales* forma *keralense* Manim. & Noordel.<br>read: *Entoloma dysthales* var. *keralense* Manim. & Noordel. |
| p.331, line 23 | *for:* Castellano MA, Smith JE, O'Dell T, Cázares E, Nugent S. 2003.<br>*read:* Castellano MA, Cázares E, Fondrick B, Dreisbach T. 2003. |

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