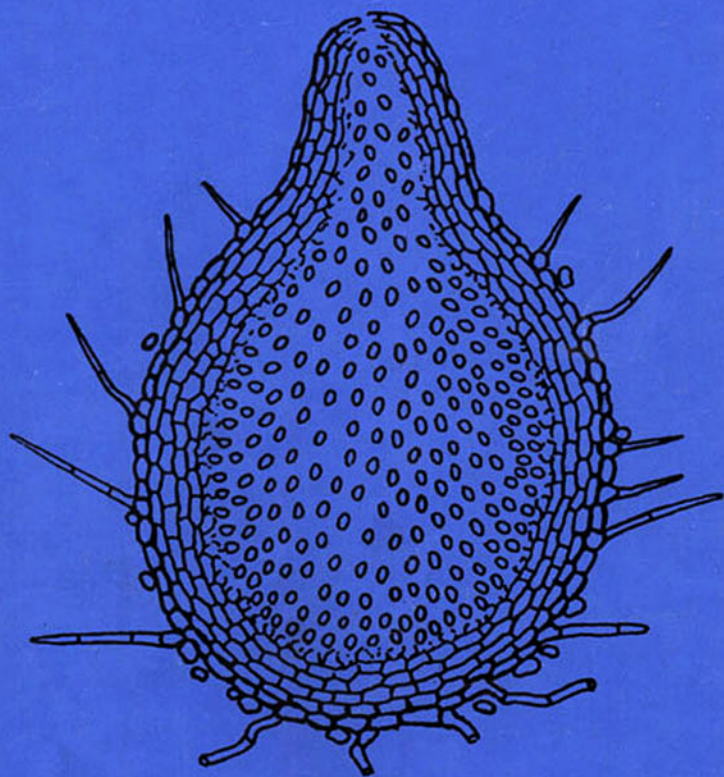


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NOTES ON CREPIDOTUS
FROM MEXICO AND THE SOUTH-EASTERN USA

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Thirteen *Crepidotus* taxa collected in Mexico and the south-eastern USA are described and illustrated. One new infraspecific taxon of *C. croceitinctus* is proposed, var. *aurantiacus*. Type material of *C. ellipsoideus* and *C. occidentalis* has been re-examined.

The Systematics Agenda 2000 (Anonymus, 1994) is a fierce call to discover and describe the global species diversity. Many undescribed species still await detection especially among the fungi from tropical and subtropical regions. However, the situation is not the same in all genera, some as the genus *Crepidotus* (Fr.) Staude in the Americas are fairly well known due to the contributions of Singer (1973) and Hesler & Smith (1965). Recent investigations (Senn-Irlet & de Meijer, 1998; Bandala et al., 1999; Bandala & Montoya, 2000a, b) have contributed to a better understanding of the variability of several species and the identity with European material but have not revealed many new species. This study aims to add additional observations towards a predictive classification system in *Crepidotus*. Further, it is a small contribution to the knowledge of Mexican fungi and to the All-Taxa-Biodiversity-Inventory in the Great Smoky Mountains National Park, Tennessee and North Carolina, which is an extensive biodiversity project in the USA (Norvell, 1999; Kaiser, 1999). Two of us (IK and HV) could profit from an invitation to Mexico and the south-eastern USA in 1996, during which 18 collections of the genus *Crepidotus* were made, belonging to 13 taxa. Besides common species, like *C. applanatus*, some more rare taxa, e.g. *C. cinnabarinus*, were found and one new infraspecific taxon of *C. croceitinctus* is proposed.

MATERIALS AND METHODS

The microscopic structures were observed in dried material. Fragments of the carpophore were mounted in Congo red, heated and examined in 5% KOH. Per collection 20 spores were measured and the population limits estimated (see Senn-Irlet, 1995).

The macroscopic descriptions are from the first and third author, respectively; the first author provided colour slides of all collections, and the second author is responsible for the microscopic examinations and the identification. The material is kept in WU (Vienna University herbarium) and in TENN (University of Tennessee, Knoxville). Within the two subgenera the taxa are arranged alphabetically.

Crepidotus subgenus *Crepidotus*

Crepidotus calolepis (Fr.) P. Karst — Fig. 1

Crepidotus calolepis (Fr.) P. Karst., Bidr. Känn. Finl. Nat. Folk 32 (1879) 414.
Selected literature: Senn-Irlet (1995).

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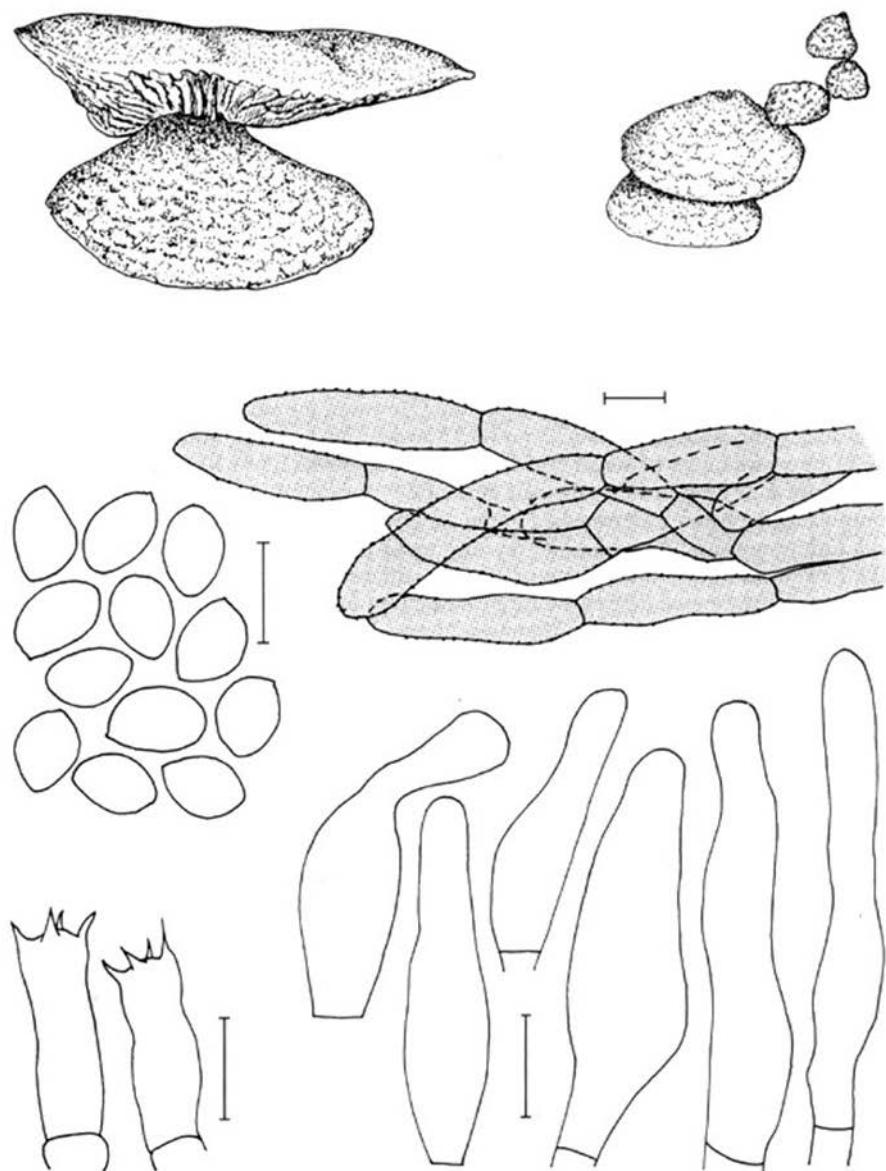


Fig. 1. *Crepidotus calolepis* (no. 6620). Carpophores, scale-forming hyphae, spores, basidia, and cheilocystidia. Bar = 10 μ m.

Pileus 5–60 mm in diam., young nearly circular to semicircular, then applanate semi-circular to flabelliform, strongly hygrophanous, completely red-brown finely appressed scaly, scales lighter ochre when young, sometimes young only fine brownish fibrillous, when old and in wet weather conditions smooth, mat, macroscopically only with thin gelatinous layer, sometimes very water-soaked and then gelatinous layer not obvious, margin sharp, striate, young dirty yellowish, then ochraceous, golden-ochre, moist pale grey to ochraceous or ochre-brown, dry cream to ochre-yellow. Lamellae shortly decurrent to straightly adnate to deeply sinuate, ventricose, in normal distance to slightly crowded, with 1–3 lamellulae, young whitish to dirty cream or pale leathery brown, buff, later watery-cream, old dirty ochraceous to greyish brown, very old dark red-brown due to the mature spores; with lamellar edge concolourous or sometimes slightly brownish, smooth to finely denticulate. Stipe short or absent, not visible from above, cream, finely whitish tomentose to finely fibrillose. Context thin, watery, cream to whitish, smell none, taste mild.

Spores $6.6\text{--}9.1 \times 4.5\text{--}6.2 \mu\text{m}$, $Q = 1.3\text{--}1.6$, av. vol. = $119 \mu\text{m}^3$, broadly ellipsoid to ellipsoid, smooth, thick-walled, strongly coloured ochre-brown. Basidia $18\text{--}23\text{--}(28) \times 6\text{--}7.5 \mu\text{m}$, 4-spored, clamps absent. Cheilocystidia $27\text{--}50 \times 6\text{--}11 \mu\text{m}$, narrowly conical or narrowly lageniform, more rarely cylindrical. Pileipellis scales formed by bundles of $7\text{--}13 \mu\text{m}$ wide hyphae composed of botuliform short cells. Subpellis with a narrow band of filamentous, $2\text{--}3 \mu\text{m}$ wide hyphae embedded in a gelatinous matrix. Lamellar trama towards the subhymenium with filamentous, slightly gelatinised hyphae, in the central part of short-celled, much wider hyphae, tending to form a jig-saw like structure. Pigment brown, encrusting and intracellular in scale-forming hyphae of the pileipellis.

Ecology — Saprotrophic, gregarious on rotten logs of *Quercus* in sclerophilous oak forest or mixed forest (*Quercus*, *Arbutus*, *Pinus*, *Juniperus*).

Collections examined. MEXICO: Federal State Mexico, El Ocotal, Chapa de Mota near Mexico City, 8 July 1996 I. Krisai-Greilhuber & H. Voglmayr no. 6615 & 6620 (WU 20098); *ibid.*, 9 July 1996 I. Krisai-Greilhuber & H. Voglmayr no. 6635.

When covered completely with red-brown scales as in coll. 6620 and with an elastic consistency, this species is easily recognised already in the field. However, the density of red-brown scales may vary greatly in this species, as in coll. 6615 where the coloured scales were hardly visible on the fresh watery carpophores, but distinctly visible in the microscopic examination.

Singer (1973), in his 'stirps mollis', tried to describe this great variation in thickness of the gelatinous layers, in the density of the brownish scales and in the shape pattern of the cheilocystidia with several species and infraspecific names. Coll. 6620 would key out under *C. calolepis* ssp. *tigrensis* var. *januarius*, and coll. 6615 under *C. variisporus*, however without the characteristic variable spore forms.

This group around *Crepidotus mollis* and *C. calolepis* is best recognised with a rather broad species concept. Species of this group show a world-wide distribution and are frequently found fruiting. Morphological studies of accidentally sampled carpophores seem hardly ever to give a sound classification. First results of molecular analyses of the genetic diversity of the large subunit of ribosomal DNA (Senn-Irlet & Hofstetter, 1996, plus unpublished results) show little variation, indicating a very close relationship at least of these analysed collections. Crossing experiments are still lacking.

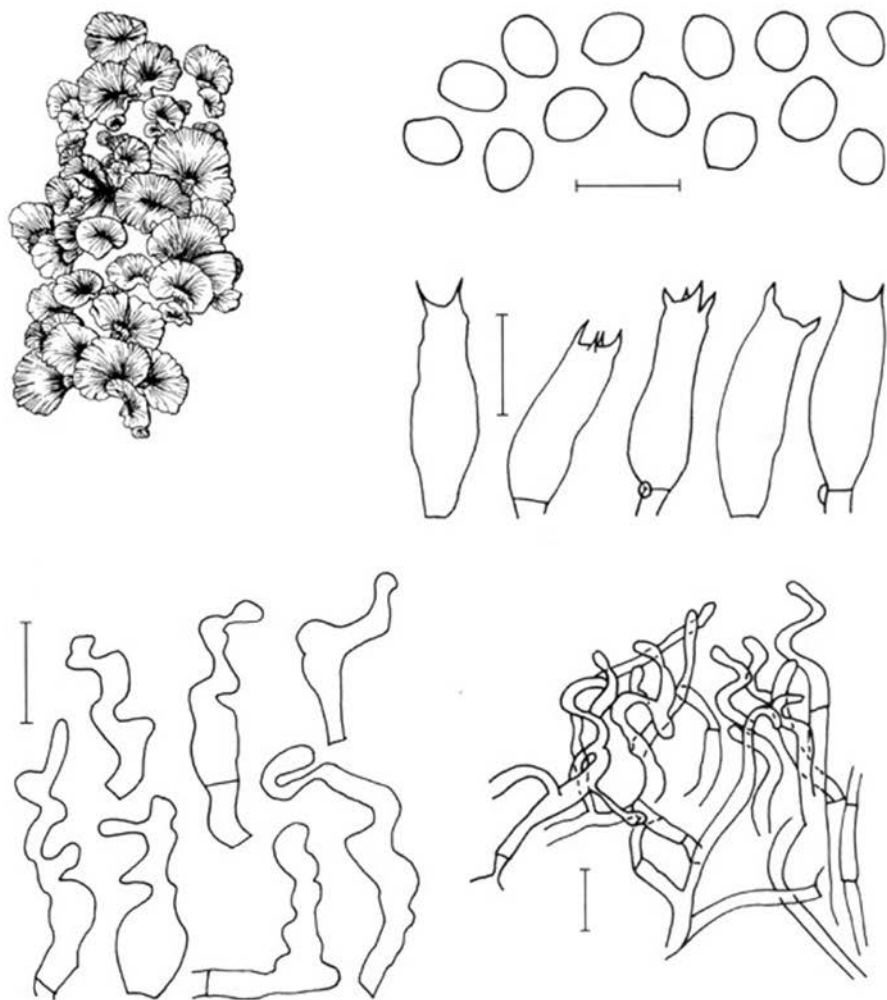


Fig. 2. *Crepidotus albidus* (no. 6522). Carpophores, spores, basidia, cheilocystida, and pileipellis. Bar = 10 μ m.

***Crepidotus* subgenus *Dochmiopus* (Pat.) Pilát**

Crepidotus subgenus *Dochmiopus* (Pat.) Pilát, *Atl. Champ. Eur.* 6 (1948) 12.

***Crepidotus albidus* Ellis & Everh. — Fig. 2**

Crepidotus albidus Ellis & Everh., *Philadelphia Acad. Nat. Sci. Proceed.* (1895'1894') 322.

Selected literature: Bandala & Montoya (2000b).

Pileus 8–12 mm in diam., nearly circular, shell-shaped, kidney-shaped, regularly crenulate at the margin; white, finely tomentose hairy. Lamellae regular, crowded, narrow, with lamellulae, young white, soon pale grey-brown. Stipe absent. Context thin, smell none, taste mild. Spore-print dark chocolate brown (very dark for a *Crepidotus*).

Spores $5.9\text{--}6.9 \times 4.7\text{--}5.6 \mu\text{m}$, $Q = 1.1\text{--}1.3$, av. vol. = $88 \mu\text{m}^3$, broadly ellipsoid, smooth, thick-walled, dark brown. Basidia $17\text{--}21 \times 5.5\text{--}6.5 \mu\text{m}$, 4-spored, clamped. Cheilocystidia $18\text{--}30 \times 3\text{--}5 \mu\text{m}$, cylindrical, strongly flexuous and contorted. Pileipellis a trichoderm with a turf of erect and contorted hyphae. Clamps rare. No pigment.

Ecology — Saprotrophic, gregarious on rotten branch of deciduous tree in subtropical mixed forest (mainly *Quercus*, *Pinus*).

Collection examined. MEXICO: Federal State Mexico, Nevada de Toluca, Temascaltepec, 5 July 1996, I. Krisai-Greilhuber & H. Voglmayr no. 6522.

This species with small carpophores is easily recognised due to its microscopic characters: strongly contorted cheilocystidia and a trichoderm with similar contorted hyphae. This species is only known from the Americas (Hesler & Smith, 1965; Singer, 1973; Senn-Irlet & de Meijer, 1998).

Crepidotus applanatus (Pers.) Kumm. var. *applanatus* — Fig. 3

Crepidotus applanatus (Pers.) Kumm. var. *applanatus*, Führ. Pilzk. (1881) 74.

Selected literature: Senn-Irlet (1995).

Pileus 10–30 mm in diam., flat flabelliform or spatulate, margin first enrolled, distinctly striate, hygrophanous, from faintly marginally to strongly striate when moist, young whitish, old only near the stipe whitish to ivory, elsewhere dirty grey-brown to dirty buff, pale creamy grey, with faint pinkish tinge, translucent, smooth. Lamellae very crowded, sinuate, slightly ventricose, decurrent, dirty grey to concolorous to pileus, only slightly more brownish pink, lamellar edge white, denticulate. Stipe short, eccentric or rudimentary, not visible from above, ivory and hairy. Context thin, watery, concolorous, not gelatinous, whitish, smell none, taste mild.

Spores $4.0\text{--}6.4 \times 3.9\text{--}6.3 \mu\text{m}$, $Q = 0.94\text{--}1.1$, av. vol. = $78 \mu\text{m}^3$, globose, punctate-warty, rather thin-walled, rather pale brown. Basidia $18\text{--}22 \times 5\text{--}6 \mu\text{m}$, 4-spored, clamped. Cheilocystidia $34\text{--}52 \times 6\text{--}10 \mu\text{m}$, cylindrical, slightly lageniform, subcapitate. Pileipellis a cutis of rather broad $8\text{--}13 \mu\text{m}$ wide hyphae, with scattered pileocystidia. Lamellar trama with rather inflated hyphae ($14\text{--}20 \mu\text{m}$ wide) and with oleiferous hyphae. Clamps abundant in all parts of the carpophore. Pigment not observed in trama and pellis.

Ecology — Saprotrophic, gregarious on rotten logs of deciduous trees in mixed forests (*Pinus*, *Quercus*, *Rhododendron*, *Tsuga*, *Liriodendron*, *Toxicodendron*).

Collections examined. USA: North Carolina, Macon County, Nantahala National Forest, Blue Valley, 4 Aug. 1996, I. Krisai-Greilhuber & H. Voglmayr no. 7062 (WU 20097); Tennessee, Blount County, Great Smoky Mountains National Park, Abrams Creek, Little Bottoms Trail, c. 3 miles from Ranger Station, 28 July 1996, I. Krisai-Greilhuber & H. Voglmayr no. 6915.

The two collections differ in spore size, best seen when expressed as the calculated mean spore volume: $49 \mu\text{m}^3$ and $107 \mu\text{m}^3$. As has been shown by Senn-Irlet (1995) these represent extremes of values observed also in European collections.

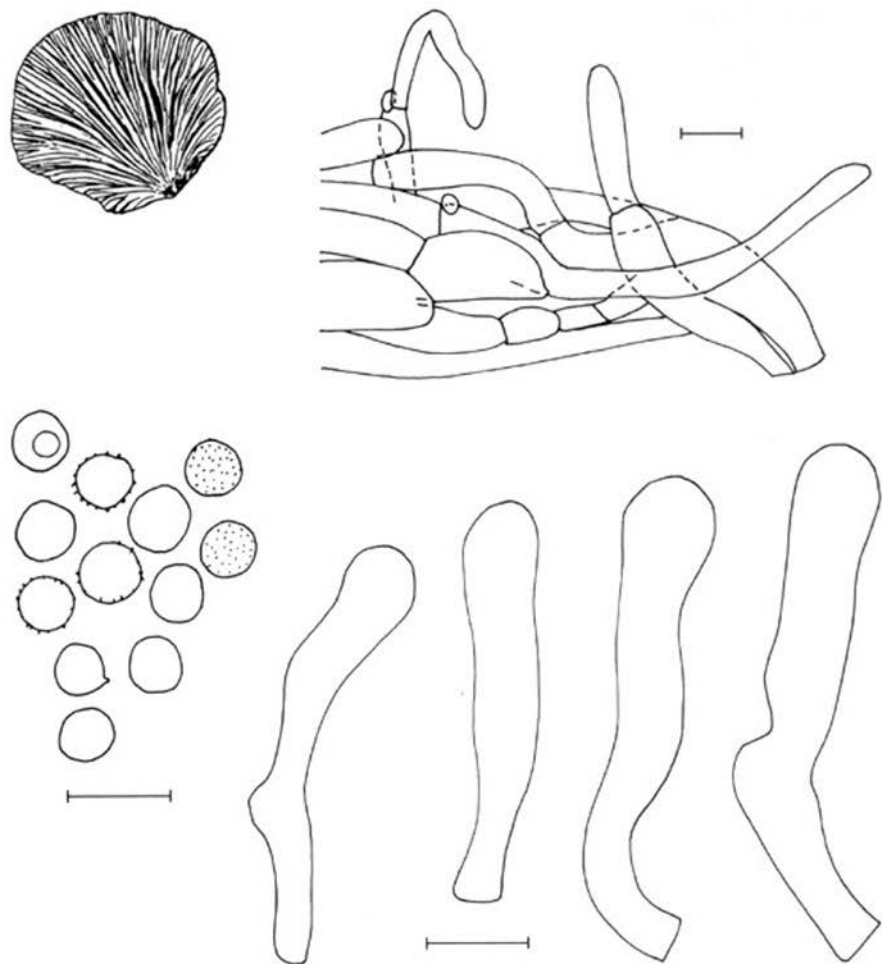


Fig. 3. *Crepidotus applanatus* (no. 6915). Carpophores, pileipellis, spores, and cheilocystidia. Bar = 10 μ m.

***Crepidotus cinnabarinus* Peck — Fig. 4**

Crepidotus cinnabarinus Peck, Bull. Torrey Bot. Club 22 (1895) 489.

Selected literature: Luther & Redhead (1981), Cifuentes et al. (1989), Senn-Irlet (1995), Bandala & Montoya (2000a).

Pileus 8 mm in diam., pleurotoid, circular to flabelliform, cinnabarine, distinctly tomentose. Lamellae crowded, cream, lamellar edge straight, even and cinnabarine. Stipe absent. Context thin, whitish.

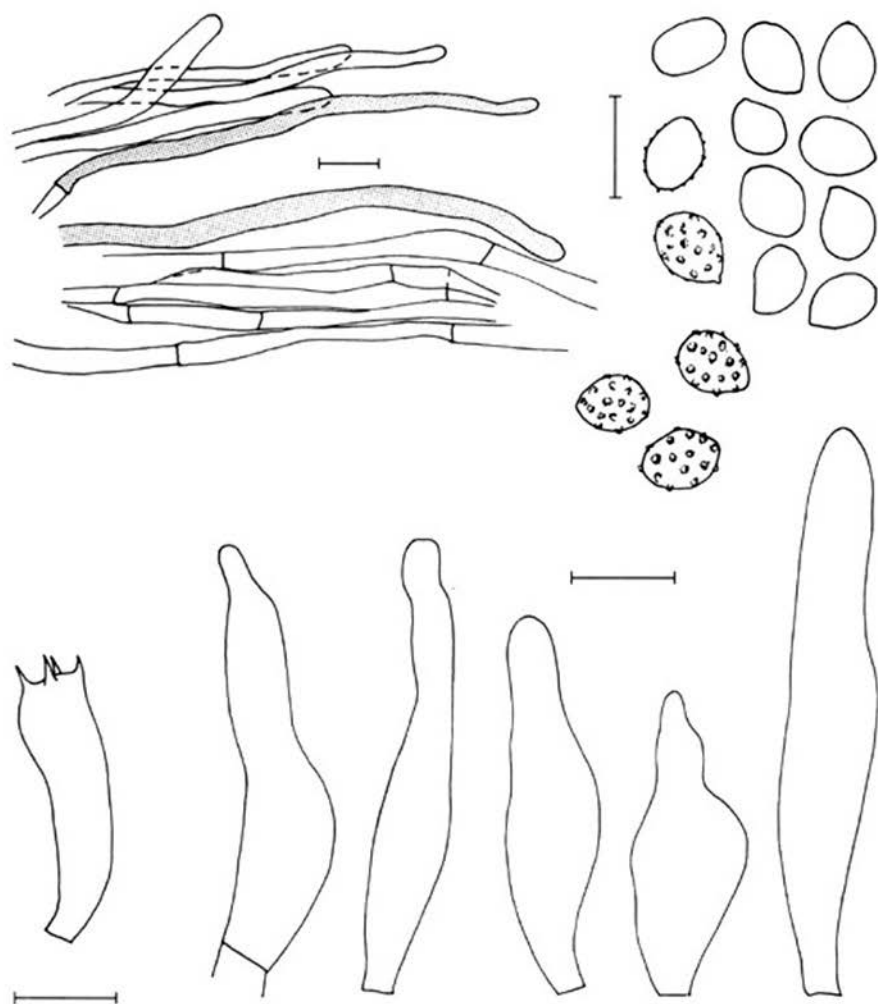


Fig. 4. *Crepidotus cinnabarinus* (no. 6678). Pileipellis, spores, basidium, and cheilocystidia. Bar = 10 μm .

Spores 6.7–8.9 \times 5.2–6.4 μm , $Q = 1.2$ –1.5, av. vol. = 139 μm^3 , broadly ellipsoid, marbled, medium dark brown. Basidia 25–31 \times 6.5–8 μm , 4-spored, clamps absent. Cheilocystidia 31–58 \times 7–11 μm , cylindrical, tapering. Pileipellis a transition between a trichoderm and a cutis. Clamps absent. Pigment red, intracellular, in hyphae of pileipellis and in cheilocystidia, dissolving in ammonia.

Ecology — Saprotrophic, solitary on rotten branch of *Quercus* (?) in a mixed oak forest (*Quercus*, *Pinus*, *Abies*).

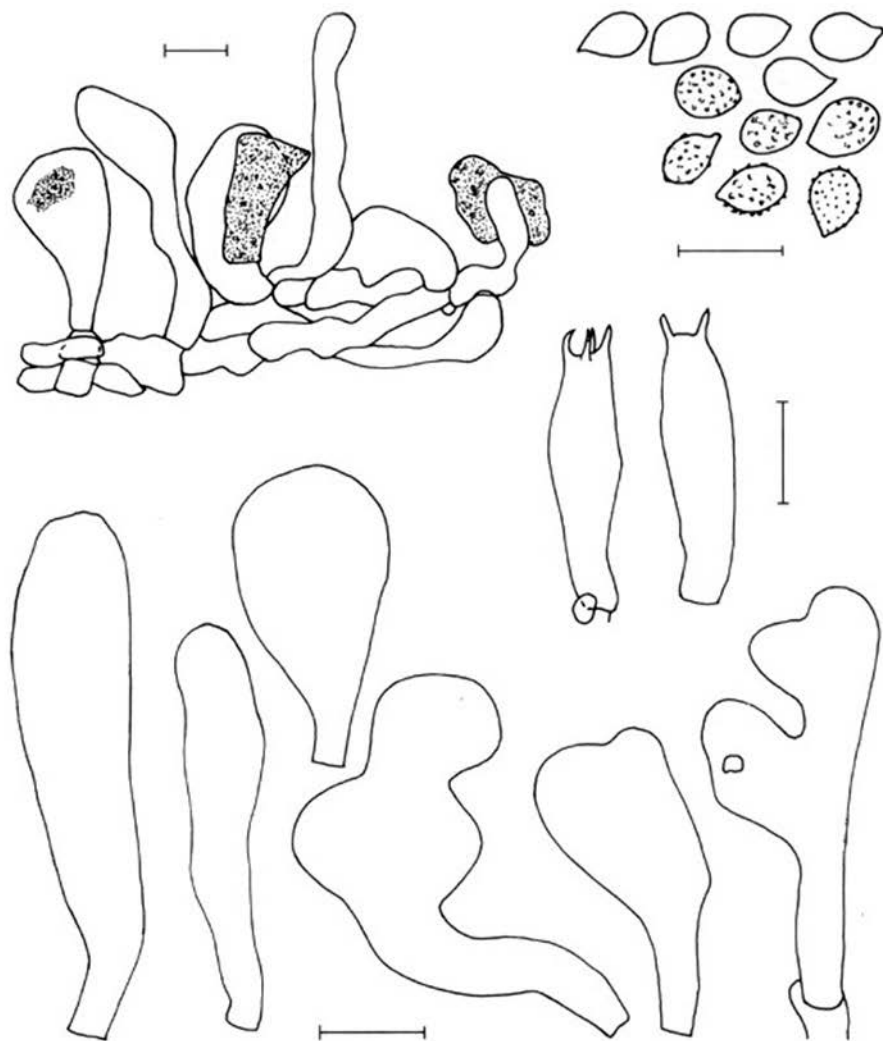


Fig. 5. *Crepidotus croceitinctus* var. *aurantiacus* (no. 6561). Pileipellis, spores, basidia, and cheilocystidia. Bar = 10 μ m.

Collection examined. MEXICO: Federal State Tlaxcala, Volcanic belt, City Tlaxcala E of Mexico City, 5 July 1996, I. Krisai-Greilhuber & H. Voglmayr no. 6678 (WU 20096).

The cystidia are distinctly shorter than found in the scanned European material. Yet, with its striking colour, the absence of clamps and the dissolving pigment it is a very typical and distinctive species already known from Mexico (Bandala-Muñoz et al., 1988; Cifuentes et al., 1989).

Crepidotus croceitinctus Peck

Crepidotus croceitinctus Peck, Ann. Rep. N.Y. Stat. Mus. 39 (1886) 72.

Selected literature: Bandala & Montoya (2000a).

Short species characteristics (detailed descriptions see the varieties below). Pileus semi-circular, flabelliform, somewhat conchate, becoming plane, never white, with yellow, orange or buff colours. Spores not longer than 7.5 μm , globose to broadly ellipsoid, distinctly warty, in SEM verruculose (*C. carpaticus* type sensu Senn-Irlet, 1995); pileipellis a cutis with repent hyphae with often some scattered erect outgrowths or pileocystidia, at times with turfs of erect shorter hyphae. Cheilocystidia with or without excrescences, in the first case often di- to polymorphic, up to 14 μm broad.

KEY TO THE VARIETIES OF *CREPIDOTUS CROCEITINCTUS*

- 1a. Pileus deep orange, pileipellis sharply separated from pileitrama, with amorphous clumps of yellow pigment var. *aurantiacus*
 b. Pileus buff to yellow, pileipellis not sharply separated from pileitrama, amorphous clumps of yellow pigment absent var. *croceitinctus*

Crepidotus croceitinctus seems to be restricted to the Americas where it has been reported from Michigan, New York and Tennessee (Hesler & Smith, 1965), from Central America (Bandala & Montoya, 2000a) and from Argentina and Brazil (Singer, 1973; Senn-Irlet & de Meijer, 1998). The collections made during the present investigations showed some important differences from typical *C. croceitinctus*, which lead us to propose a new variety.

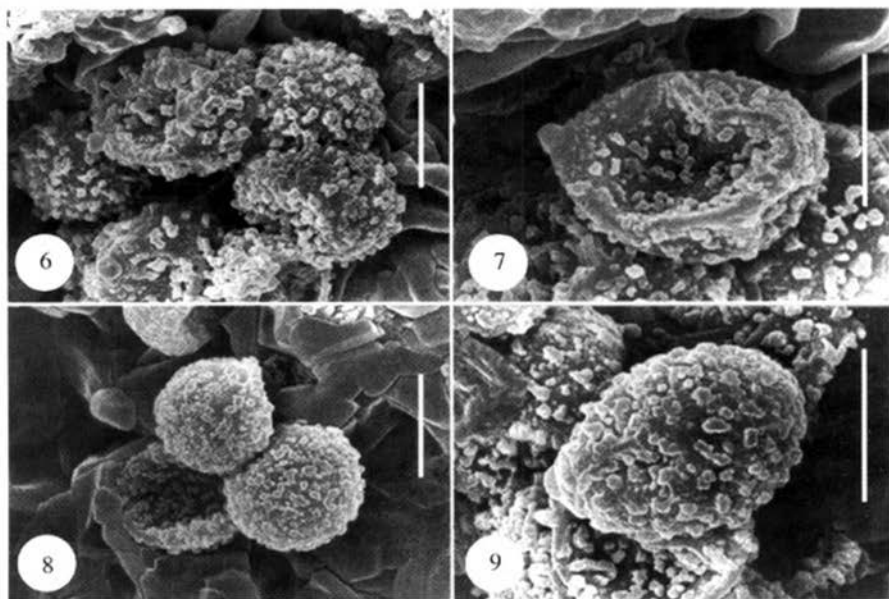
***Crepidotus croceitinctus* var. *aurantiacus*, var. nov.** — Figs. 5–7, Plate 1

A typo differt pileo aurantiaco. In zona subtropicalis.

Holotypus: *I. Krisai-Greilhuber & H. Voglmayr 6561*, 6-VI-1996, Mexico, Mexico Federal State, Temascaltepec (WU 20093) (isotypus in TENN).

Pileus 20–70 mm in diam., pleurotoid, flabelliform, shell-shaped, young margin slightly striate, hygrophanous, fresh vividly yellow-orange, orange to brown-orange, old fading to greyish yellow, tomentose, upper side with white felty mycelium near point of attachment. Lamellae crowded, first cream yellow, then rusty orange. Stipe short, eccentric, later rudimentary or absent, cream yellow, then orange. Context thin, smell mouldy-sour, taste mild.

Spores 5.3–7.5 \times 4.5–5.9 μm , Q = 1.0–1.5, av. vol. = 90 μm^3 , broadly ellipsoid, with faint suprahilar depression, distinctly warty, with warts visible in optical section and with myxosporium, in exsiccates easily collapsed, strongly coloured. Basidia 18–31 \times 6–7.5 μm , 4-spored, clamped. Cheilocystidia 25–60 \times (7–) 9–15.5 μm , clavate to broadly clavate, rarely broadly cylindrical or lageniform, almost vesiculose, rarely broadly cylindrical, sometimes apically depressed or even forked and branched, hyaline. Pileipellis sharply separated from the pileitrama, with a rather complex morphology, a transition between a cutis and a hymeniderm with many erect pileocystidia; hyphae rather short-celled, 5–12 μm wide. Pileocystidia variable, broadly clavate, narrowly lageniform to flexuous mixed with some filiform, cylindrical ones. Clamps abundant everywhere. Pigment yellow, brownish yellow, amorphous in water, as amorphous exudates around some pileocystidia or intracellular, membranous and scattered between the repent hyphae of the pileipellis, in addition locally finely encrusting.



Figs. 6–9. SEM pictures of basidiospores. — Fig. 6, 7. *Crepidotus croceitinctus* var. *aurantiacus* (no. 6483). Fig. 8, 9. *Crepidotus croceitinctus* var. *croceitinctus* (no. 6927). Bars = 3 μ m.

Ecology — Saprotrophic, gregarious on rotten twigs of deciduous trees in moist subtropical mixed forests (mainly *Pinus*, *Quercus*).

Collections examined. MEXICO: Federal State Mexico, Nevada de Toluca, Temascaltepec, c. 130 km W Mexico City, 6 July 1996. I. Krisai-Greilhuber & H. Voglmayr no. 6561 (WU 20093 - holotype, TENN - isotype). *ibid.*, c. 132 km W Mexico City, El Povador, 4 July 1996. I. Krisai-Greilhuber & H. Voglmayr no. 6483 (WU 20094).

The protologue of *Crepidotus croceitinctus* as well as the descriptions by Hesler & Smith (1965), Singer (1973), Bandala & Montoya (2000a) and others never mention such a deep orange to almost fulvous colour of the pileus. The abundance of pigment causing these intense colours is also seen in the microscopic analysis. The presence of such yellow exudates forming amorphous clumps is not mentioned in any of the studies mentioned. In other collections of *C. croceitinctus* previously examined (Senn-Irlt & de Meijer, 1998) no such yellow extracellular pigments have ever been observed.

In the type collection (no. 6561) the pileipellis structure is very well preserved and therefore we can emphasize for the first time the very special type of pileipellis within the genus *Crepidotus*. Even if Hesler & Smith (1965), Singer (1973) and in detail Bandala & Montoya (2000a) describe a more or less loose turf of erect hyphae mixed with some very undifferentiated pileocystidia, they do not mention such a unique type within the genus as found in our collections. The dense turf of variously shaped pileocystidia and the amorphous masses

of yellow pigment, sometimes encrusting the pileocystidia form a very well differentiated pileipellis. In our second collection (no. 6483) no amorphous clumps of yellow pigments were noticed beside still very obvious amounts of intracellular, membranous and finely incrusting yellow pigments. Nevertheless, we suggest that also this collection belongs to the same taxon on account of the other fully matching characters, especially the dense turf of pileocystidia.

As the colour and the structure of the pileipellis are so striking in our collections, we propose the status of a new variety. Further collections have to show if intermediate forms exist or whether this coloured form is better regarded as a species of its own.

***Crepidotus croceitinctus* var. *croceitinctus* — Figs. 8–10**

Crepidotus subcroceitinctus Hesler & A. H. Sm., N. Am. Sp. Crep. (1965) 139.

Pileus 5–40 mm in diam., semicircular, old nearly circular, kidney-shaped, shell-shaped, very flat, margins near point of attachment sometimes overlapping, near point of attachment whitish-cream, other parts vividly golden ochre, completely whitish to cream hairy-tomentose, hygrophanous, shortly and indistinctly striate when moist, very young upper side pale lemon yellow and margin enrolled, margin staying pale very long. Lamellae sinuate, ventricose, lamellar edge white denticulate, young pale lemon yellow, later vividly golden ochre with orange tinge (more vivid than pileus surface). Stipe short, 1–2 mm long, pale lemon yellow, soon sessile, young and old whitish tomentose. Context thin, paler watery cream, smell none, taste mild.

Spores 5.5–6.5 × 4.4–5.7 μm , $Q = 1.0\text{--}1.3$, av. vol. = 81 μm^3 ($n = 22$), globose to broadly ellipsoid, broadly ellipsoid, strongly warty, visible in optical section, easily broken, with strongly coloured walls. Basidia 21–26 × 6–7 μm , 4-spored, clamped, broadly clavate. Cheilocystidia 31–52 × 5–8 μm , cylindrical, flexuous, contorted, branched and antlerlike. Pileipellis a transition between a cutis and a trichoderm of repent 4–7 μm wide hyphae with some erect 10–20 × 3–5 μm filiform outgrowths. Pigment absent in pileitrama and lamellar trama. With scattered small crystals on some loosely repent hyphae of the pileipellis.

Ecology — Saprotrophic, gregarious on rotten trunk of a deciduous tree in a mixed forest (*Tsuga*, *Liriodendron*, *Pinus*, *Quercus*, *Rhododendron*).

Collection examined. USA: Tennessee, Blount County, Great Smoky Mountains National Park, Abrams Creek, Little Bottoms Trail, c. 3 miles off Ranger Station, 28 July 1996, J. Krisai-Greilhuber & H. Voglmayr no. 6927 (WU 20095).

In a very accurate study Bandala & Montoya (2000a) give an emended description of *Crepidotus croceitinctus* based on type studies and collections from Mexico. The pileus colour of this rather variable species is described as ranging from yellowish white to egg-yellow. Another morphological character, the shape of the cheilocystidia, otherwise a very distinctive character in *Crepidotus*, seems to vary considerably from simple clavate to flexuous, contorted, or knobbed, circumscribed as dimorphic by Singer (1973). This variability has already been noted by several authors but only the comparative study of Bandala & Montoya (2000a) allowed for a comprehensive picture of this taxon which should include *Crepidotus subcroceitinctus*. Our collection shows a lamellar edge with a majority of flexuous, contorted cheilocystidia with some outgrowths.

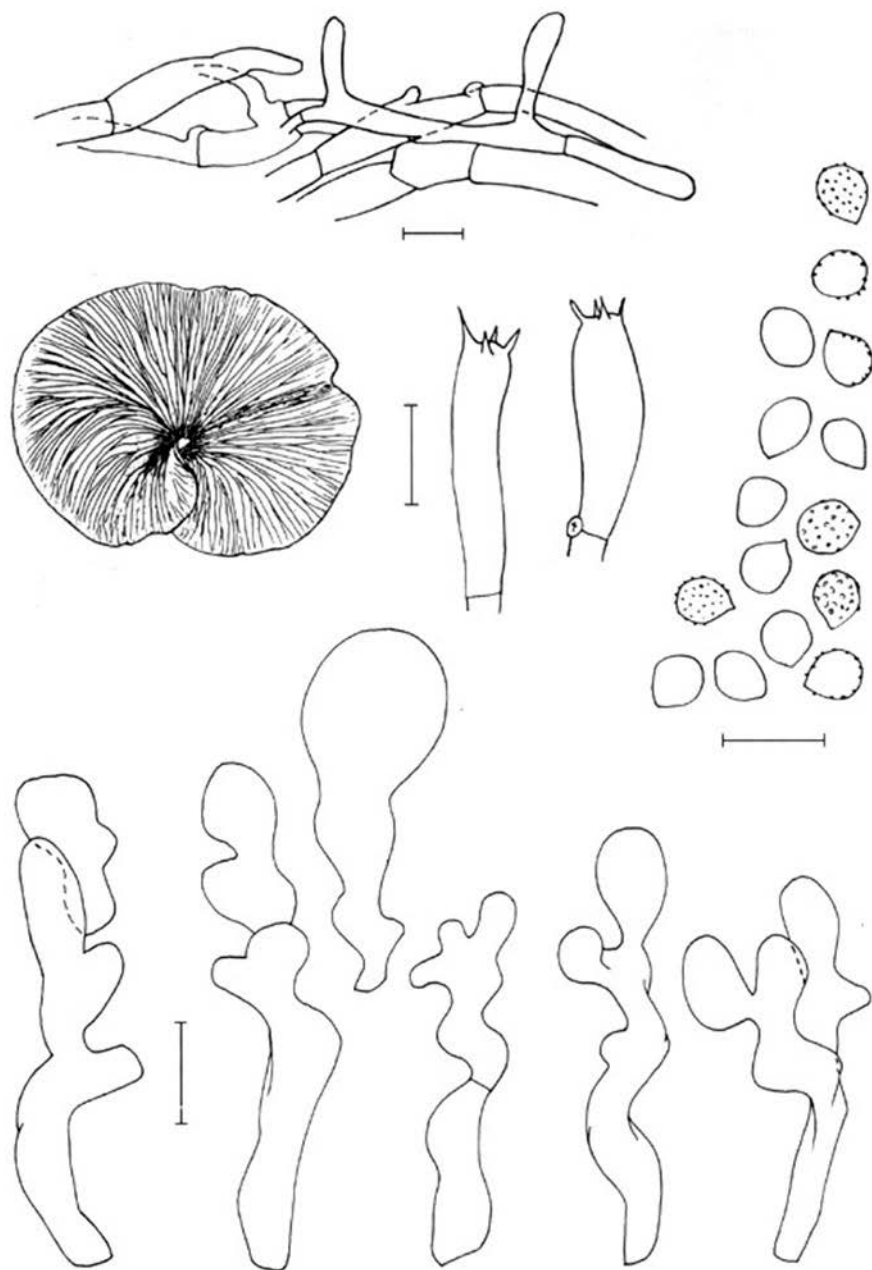


Fig. 10. *Crepidotus croceitinctus* var. *croceitinctus* (no. 6927). Pileipellis, carpophore, basidia, spores, and cheilocystidia. Bar = 10 μ m.

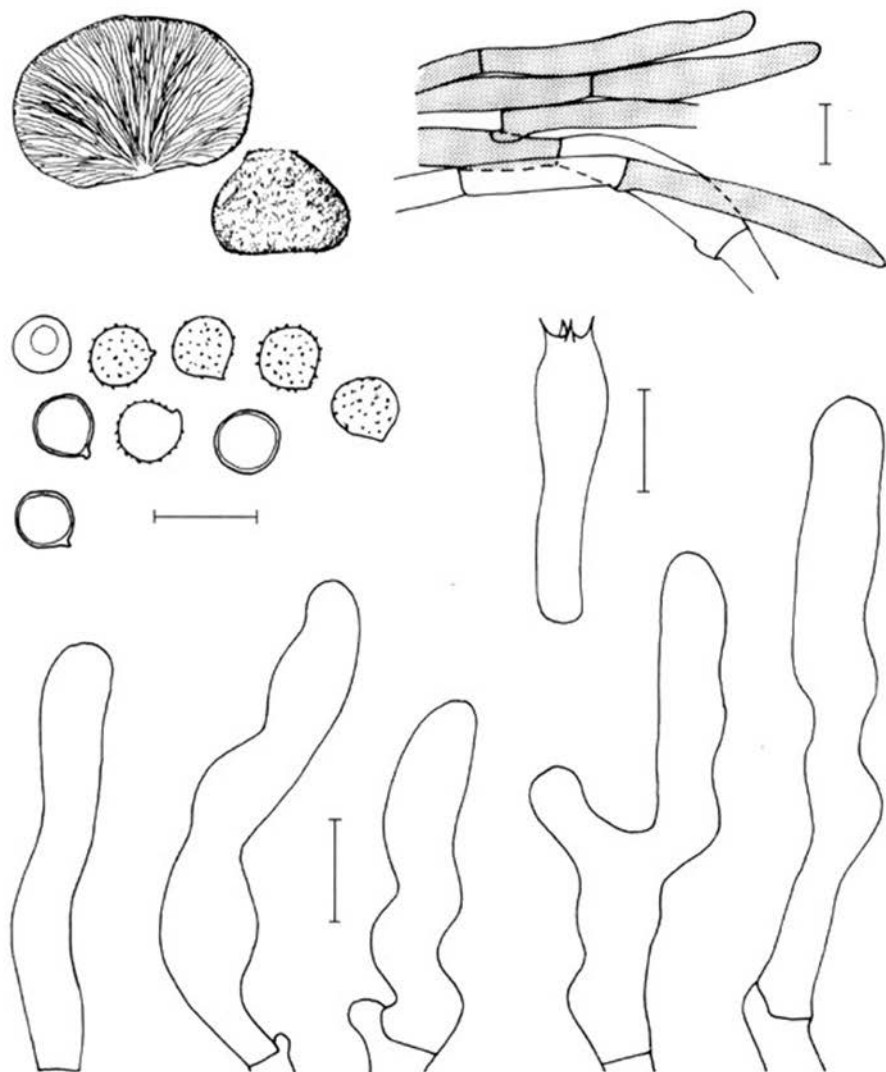


Fig. 11. *Crepidotus crocophyllus* (no. 7006). Carpophores, scale forming hyphae, spores, basidia, and cheilocystidia. Bar = 10 μ m

***Crepidotus crocophyllus* (Berk.) Sacc. — Fig. 11**

Crepidotus crocophyllus (Berk.) Sacc., Syll. Fung. 5 (1887) 886.
Selected literature: Senn-Irlet (1995).

Pileus 10–90 mm in diam., pleurotoid, circular, heart- to kidney-shaped, pale pinkish brown with fine fibrillous orange- to red-brown scales, hygrophanous, margin distinctly

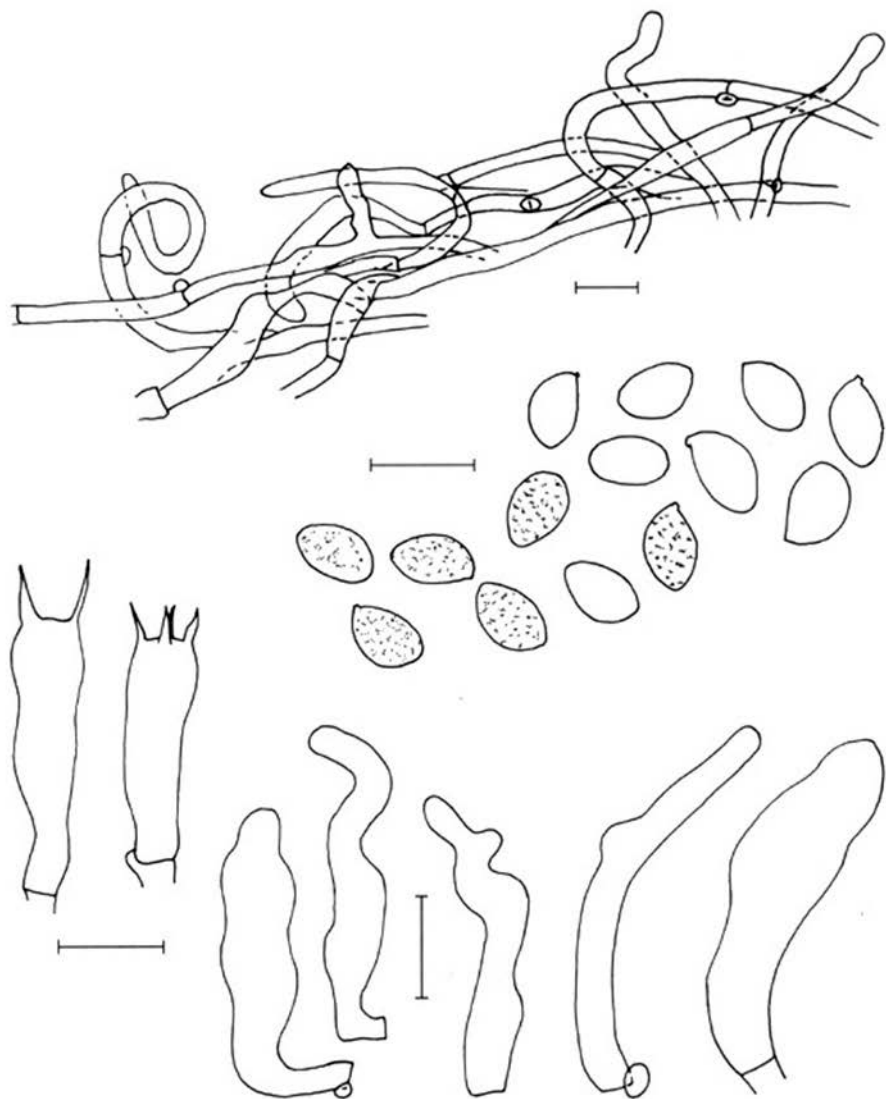


Fig. 12. *Crepidotus furcatus* (no. 6523). Pileipellis, spores, basidia, and cheilocystidia. Bar = 10 μ m.

enrolled and smooth, heavily striate, mat, not gelatinous, above point of attachment finely tomentose. Lamellae slightly crowded, dirty orange to grey-ochre, lamellar edge straight, concolorous. Stipe absent. Context thin, concolorous to pileus, smell none, taste mild.

Spores 5.5–6.8 \times 5.4–6.7 μ m, $Q = 0.95$ –1.1, av. vol = 120 μ m³, globose, punctate-warty, rather thin-walled, rather pale brown. Basidia 26–33 \times 5–6 μ m, 4-spored, clamped.

Cheilocystidia 38–61 × 6–12 µm, cylindrical, rarely lageniform, flexuous, in both collections a few branched. Pileipellis a cutis covered with dense trichodermal clusters of coloured scales with undifferentiated, 40–60 × 6–8 µm terminal cells. Clamps abundant everywhere. Pigment golden brown, parietal and in addition encrusting, present in the scale-forming hyphae and at times in the subhymenium.

Ecology — Saprotrophic, gregarious on rotten logs of deciduous trees in mixed forests (*Liriodendron*, *Quercus*, *Tsuga*, *Pinus*).

Collections examined. USA: Tennessee, Blount County, Great Smoky Mountains National Park, Cades Cove, 2 August 1996, I. Krisai-Greilhuber & H. Voglmayr no. 7006; *ibid.*, Abrams Creek, Little Bottoms Trail, c. 3 miles off Ranger Station, 28 July 1996, I. Krisai-Greilhuber & H. Voglmayr no. 6928 (WU 20099).

The orange colours of the scaly pileus and the orange lamellae make this a rather easy recognisable species. The shape of the cheilocystidia seems to be more variable than in the type collection. While the holotype shows only rather simple cylindrical cheilocystidia of 30–50 × 6–8 µm, the two presented collections exhibit both at least some branched cheilocystidia, as has been observed in some European collections as well, where also subcapitate forms have been encountered.

The species is of scattered occurrence throughout the northern hemisphere.

***Crepidotus furcatus* Hesler & A.H. Sm. — Fig. 12**

Crepidotus furcatus Hesler & A.H. Sm., N. Am. Sp. Crep. (1965) 115 [Figs. 12, 13 (holotype of *C. ellipsoideus*)].

Selected literature: Hesler & Smith (1965).

Pileus 15–30 mm in diam., circular to undulating-wavy, young off-white, old pale cream, surface tomentose to finely villose. Lamellae crowded, pale pinkish brown. Stipe absent. Context thin, smell spicy, taste mild.

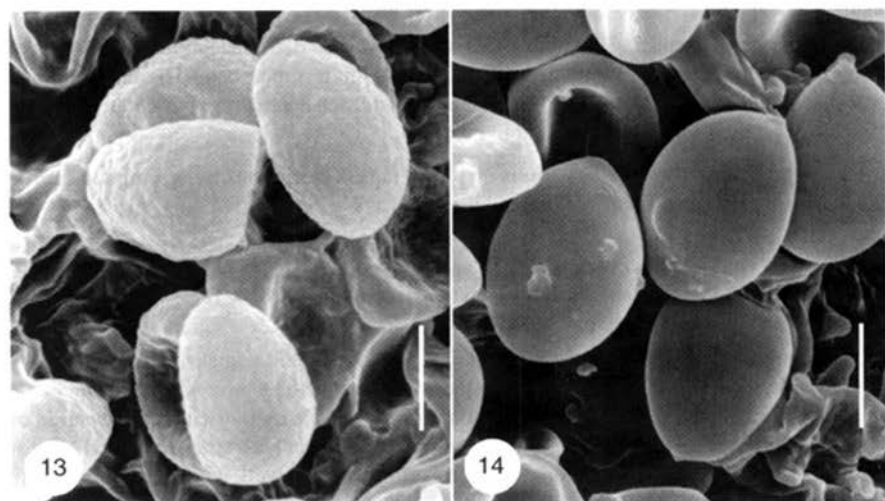
Spores 7.1–9.2 × 4.7–6.3 µm, Q = 1.3–1.7, av. vol. = 130 µm³, ellipsoid, with a faint suprahilar depression, faintly ornamented, marbled, rugulose-verruculose, rather thin-walled with faintly coloured walls. Basidia 23–29 × 7–9 µm, 2- and mixed 4-spored, clamped. Cheilocystidia 23–35 × 6–12 µm, cylindrical, contorted, rarely angled, broadly clavate and flexuous. Pileipellis a transition between a cutis and a trichoderm with crooked and bent hyphae and undifferentiated terminal cells. Pileitrama regular. Clamps abundant. Pigment not present.

Ecology — Saprotrophic, gregarious on rotten trunk of a deciduous tree in a broad-leaved subtropical oak forest.

Collection examined. MEXICO: Federal State Mexico, Nevada de Toluca, Temascaltepec, c. 132 km W. of Mexico City, Avandaro Mexico, 6 July 1996, I. Krisai-Greilhuber & H. Voglmayr no. 6523.

Our specimen fits the description of *Crepidotus furcatus* rather well, a species of temperate to boreal North America with somewhat contorted cheilocystidia, distinctly coiled and crooked hyphae on the pileipellis and a spore size of 7–10 × 4.5–5.5 µm. However, there is a difference in the colour of the lamellae: whereas Hesler & Smith (1965) state it to be clay to ochre-tawny in colour, we have seen distinct pinkish brown tints.

In section *Crepidotellae* Hesler & A.H. Sm. more very similar species have been described. The separation of *C. ellipsoideus* Hesler & A.H. Sm. and *C. furcatus* is solely



Figs. 13, 14. SEM pictures of basidiospores. — Fig. 13. *Crepidotus ellipsoideus* (holotype). Fig. 14. *Crepidotus occidentalis* (holotype). Bars = 3 μ m.

Table I. Distinctive characters of *Crepidotus ellipsoideus* and *C. furcatus* based on the original descriptions (Hesler & Smith, 1965).

Characters	<i>C. ellipsoideus</i>	<i>C. furcatus</i>
Spore size (μ m)	(5.5–)6–8 \times (4–)4.5–5.5	7–10 \times 4.5–5.5(–6)
Spore ornamentation	punctate	minutely punctate
Cheilocystidia size (μ m)	26–45(–66) \times 5–12(–17)	30–50 \times 4–10
Cheilocystidia shape	ventricose, clavate, crooked, contorted, at times forked	cylindrical, clavate, contorted, more rarely forked or knobbed
Spore ornamentation in SEM	faintly rugulose-vermiculose (holotype; see Fig. 13)	

based on small differences in spore size (see Table I). A re-examination of the type of *C. ellipsoideus* Hesler & A.H. Sm. showed a faintly rugulose-vermiculose spore ornamentation when examined under the SEM (see Fig. 13) and most spore sizes within the overlapping range. More observations are needed to decide if these two species are conspecific.

Crepidotus obscurus Hesler & A.H. Sm. — Fig. 15

**Crepidotus obscurus* Hesler & A.H. Sm., N. Am. Sp. Crep. (1965) 128.
Selected literature: Hesler & Smith (1965).

Pileus 5–30 mm, white, white fibrillose, slightly tomentose, finally almost smooth near the margin, older dirty brownish, flat, semicircular to circular, sometimes wavy or lobed.

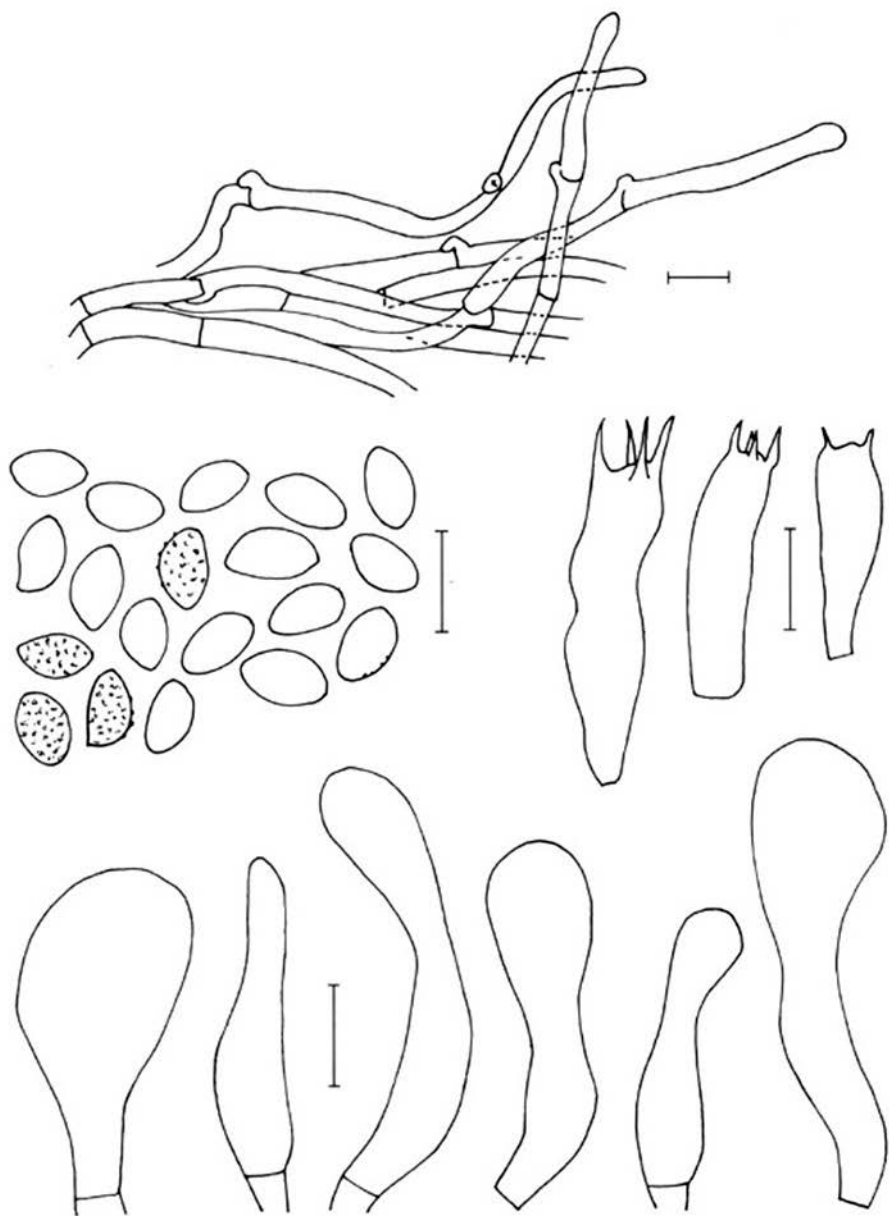


Fig. 15. *Crepidotus obscurus* (no. 7058). Pileipellis, spores, basidia, and cheilocystidia. Bar = 10 μ m.

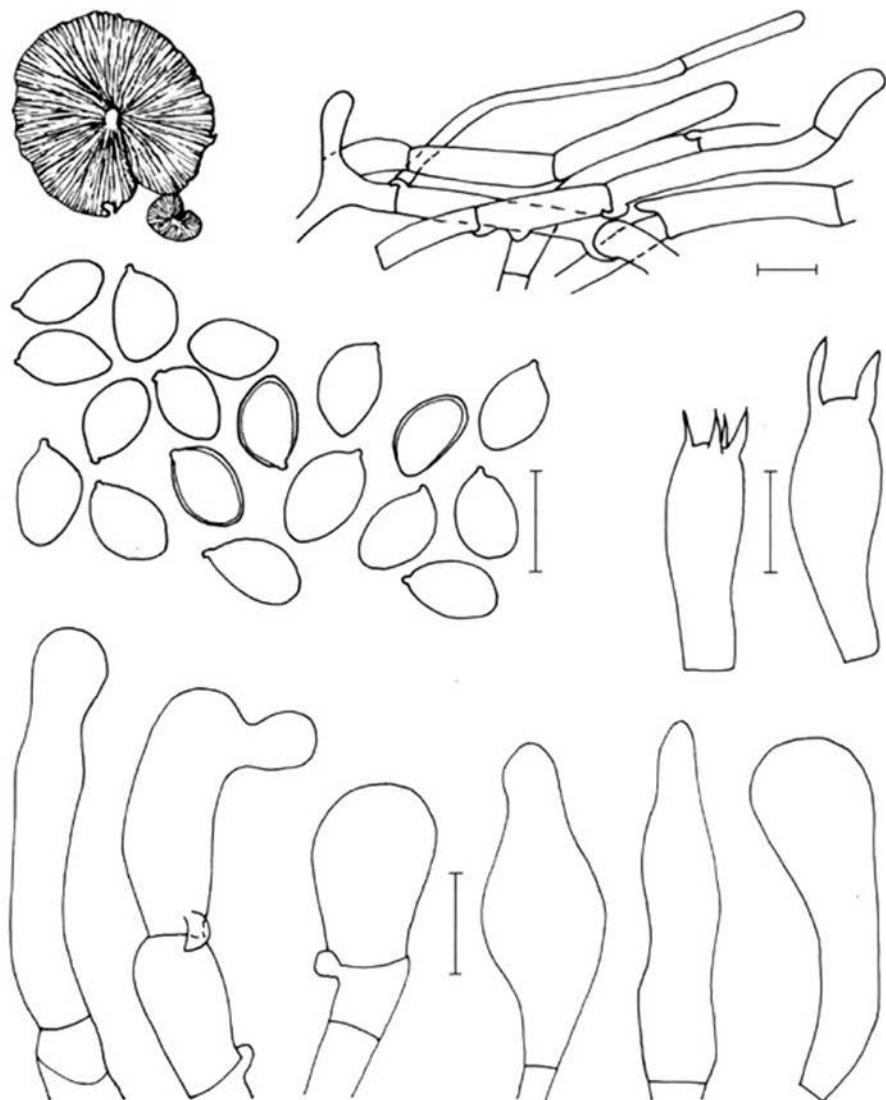


Fig. 16. *Crepidotus occidentalis* (no. 6566). Carpophores, pileipellis, spores, basidia, and cheilocystidia. Bar = 10 μ m.

Lamellae crowded, white then orange to dirty brown. Stipe 2–3 mm long, eccentric, white. Context thin, whitish, smell and taste mild.

Spores $6.5\text{--}8.9 \times 4.3\text{--}5.4 \mu\text{m}$, $Q = 1.37\text{--}1.8$, av. vol. = $94 \mu\text{m}^3$, ellipsoid, slightly amygdaliform in side view, faintly marbled, rugulose-verruculose, moderately thin-walled, rather pale brown. Basidia $22\text{--}32 \times 6\text{--}7 \mu\text{m}$, 4-spored, clamped. Cheilocystidia $30\text{--}48 \times 7\text{--}10 \mu\text{m}$ of two types, cylindrical, narrowly lageniform i.e. bottle-shaped, or broadly clavate up to $18 \mu\text{m}$ broad to almost sphaeropedunculate. Pileipellis a cutis of more or less repent $3\text{--}4 \mu\text{m}$ wide hyaline hyphae, with some ascending hyphae. Clamps abundant. No pigment found in trama and pellis.

Ecology — Saprotrophic, gregarious on rotten twig of a deciduous tree in a mixed forest.

Collection examined. USA: North Carolina, Macon County, Nantahala National Forest, Blue valley, 4 Aug. 1996, I. Krisai-Greilhuber & H. Voglmayr no. 7058 (WU 20100).

Crepidotus obscurus is characterised by the ellipsoid, rugulose-verruculose spores and the shape of the cheilocystidia which are of two distinct types. It is a member of the species-rich section *Crepidotellae* Hesler & A.H. Sm.

Crepidotus occidentalis Hesler & A.H. Sm. — Fig. 16

Crepidotus occidentalis Hesler & A.H. Sm., N. Am. Sp. Crep. (1965) 103 (Figs. 14, 16).

Selected literature: Hesler & Smith (1965).

Pileus 1–3.5 cm in diam., pleurotoid, tomentose-hairy, greyish white to silver-grey, old brown-grey. Lamellae pink-brown grey, lamellar edge concolorous. Stipe present, short. Context thin, white, smell indistinct, taste mild.

Spores $8.1\text{--}10.1 \times 5.4\text{--}6.7 \mu\text{m}$, $Q = 1.3\text{--}1.7$, av. vol. = $176 \mu\text{m}^3$, ellipsoid, smooth, rather thick-walled, and distinctly brown-coloured. Basidia $23\text{--}29 \times 7\text{--}9 \mu\text{m}$, 4-spored, clamped. Cheilocystidia $23\text{--}35 \times 6\text{--}12 \mu\text{m}$, lageniform, apex acute and tapering or rounded and subcapitate, some angled. Pileipellis a cutis with narrow, $2 \mu\text{m}$ wide, long, undifferentiated terminal cells (as 'hairs') on a pileitrama with distinctly broader hyphae. Trama not jigsaw like, without any gelatinous layer. Clamps abundant. No pigment present in trama and pellis.

Ecology — Saprotrophic, gregarious on rotten branch of a deciduous tree in a subtropical evergreen oak forest.

Collection examined. MEXICO: Federal State Mexico, Nevada de Toluca, Temascaltepec, c. 132 km W. of Mexico City, 6 July 1996, I. Krisai-Greilhuber & H. Voglmayr no. 6566.

On account of the thick-walled spores which are smooth and deeply coloured and the pileipellis type with distinctly narrow hyphae this species resembles *Crepidotus autochthonus* J. Lange and has to be placed in section *Autochthoni* Senn-Irlet. A re-examination of the holotype of *C. occidentalis* showed the same spore type (Fig. 14), although the shape of the cheilocystidia differs from our collection: whereas the holotype exhibits throughout cylindrical, slightly flexuous cheilocystidia of $34\text{--}55 \times 5\text{--}7 \mu\text{m}$ (as described and depicted by the authors!) our collection has shorter, thicker and in shape more variable ones. More collections are needed to decide if our collection still falls within the range of *C. occidentalis* or if it has to be recognised as a different species.

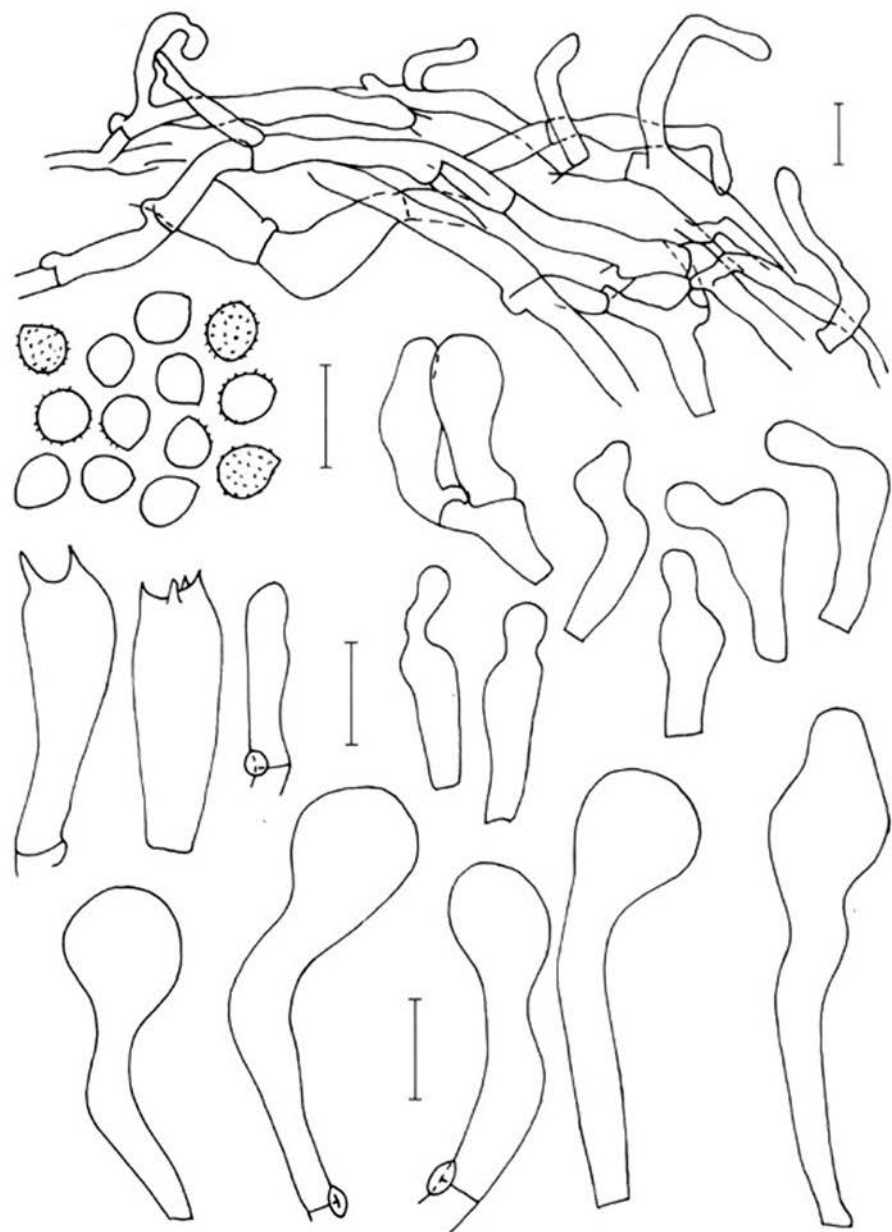


Fig. 17. *Crepidotus palmarum* (no. 6730). Pileipellis, spores, pleurocystidia, basidia, and cheilocystidia. Bar = 10 μ m.

***Crepidotus palmarum* Sing. — Fig. 17**

Crepidotus palmarum Sing. in Sing. & Digilio, Lilloa 25 (1952, '1951') 406.

Crepidotus luridus Sing., Beih. Nova Hedwigia 44(1973) 479.

Crepidotus luridus var. *oaxacae* Sing., Beih. Nova Hedwigia 44 (1973) 479.

Selected literature: Senn-Irlet & de Meijer (1998), Bandala & Montoya (2000a).

Pileus 5–15 mm, flabelliform, smooth, striate when moist, hygrophanous, dirty whitish, ivory. Lamellae slightly distant, lamellar edge smooth, concolorous, young dirty whitish, then with pinkish brown tinge. Stipe short, present only in very young fruit-bodies, soon sessile, and then only a bit thicker at point of attachment. Context thin, smell none, taste mild.

Spores $5.8-6.3 \times 4.6-5.6 \mu\text{m}$, $Q = 1.1-1.3$, av. vol. = $82 \mu\text{m}^3$, subglobose to broadly ellipsoid warty-punctate, with warts clearly visible in optical section, moderately coloured. Basidia $23-29 \times 7-10 \mu\text{m}$, 4-spored, clamped. Cheilocystidia $32-51 \times 4-5 \times 10-12 \mu\text{m}$, clavate-subcapitate to distinctly capitate (i.e. vesiculose), sometimes slightly flexuous. Hymenium full of basidioles, which can be interpreted as cystidioid elements ('pleurocystidia'), $16-21 \times 4-7 \mu\text{m}$, cylindrical to lageniform, in the upper part flexuous. Pileipellis a cutis of moderately broad repent hyphae, with erect, angled, bent terminal cells.

Ecology — Saprotrophic, gregarious on rotten trunk of *Quercus* in a mixed, humid, subtropical deciduous oak forest.

Collection examined. MEXICO: Federal State Nayarit, City Tepic, near the W coast, Villarruez-Orbaz, 18 July 1996, I. Krisai-Greilhuber & H. Voglmayr no. 6730.

With Singer (1973) this collection keys out as *Crepidotus luridus* var. *oaxacae* Singer on account of the combination of subglobose warty spores and vesiculose cheilocystidia. A figure even illustrates the shape of the pleurocystidioid terminal cells. With the exception of the substrate – not palms – the characters are the same as for *Crepidotus palmarum*. A careful comparison of the different types led Bandala et al. (1999) to conclude that these species are conspecific. We agree with this conclusion as also our observations on a collection from Brazil growing on decayed leaf petioles of a palm tree (Senn-Irlet & de Meijer, 1998) show a striking similarity with the Mexican specimen.

This collection shows a feature encountered in several, especially tropical specimens of *Crepidotus*: the hymenium is full of cystidioid elements, which can be termed as pleurocystidia (Hesler & Smith, 1965). As we firmly believe that such elements do not deserve any taxonomic status because they may simply be a modification due to climatic conditions or unknown biotic triggers (bacteria?) we abandon even the idea of describing a variety of this species as this has been done by several authors.

***Crepidotus subverrucisporus* Pilát — Fig. 18**

Crepidotus subverrucisporus Pilát, Studia bot. cech. 10 (1949) 151.

Selected literature: Senn-Irlet (1995), Bandala et al. (1999).

Pileus 8–40 mm in diam., typically flabelliform, margin strongly undulating, young pure white, old cream-white, watery and slightly translucent when moist, dry cottony, mat, tomentose, older near point of attachment light yellowish (similar to *Crepidotus luteolus* (Lamb.) Sacc.) and strigose-hairy to scaly. Lamellae crowded, first pure white, soon dirty pinkish, not directly brownish or only when very old. Stipe absent. Context white, watery, smell fruity, taste mild to slightly bitter.

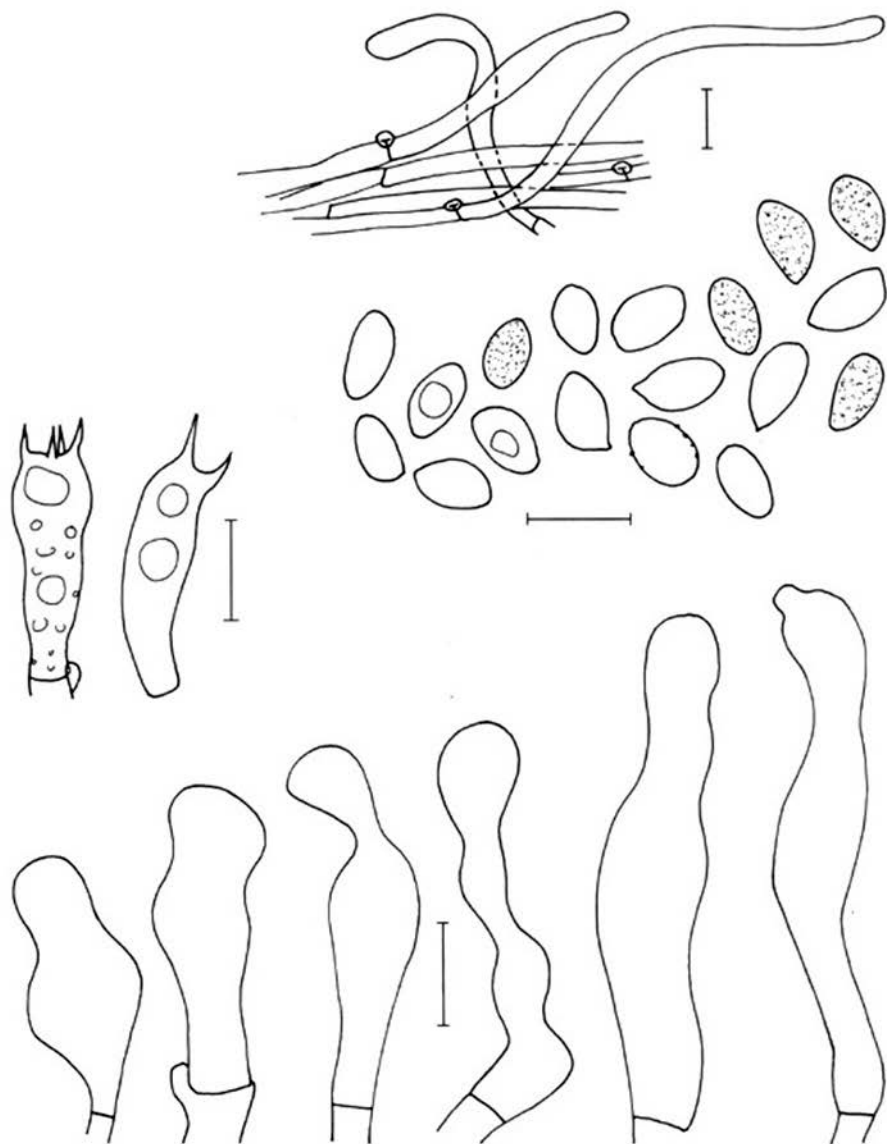


Fig. 18. *Crepidotus subverrucisporus* (no. 6474). Pileipellis, spores, basidia, and cheilocystidia. Bar = 10 μ m.

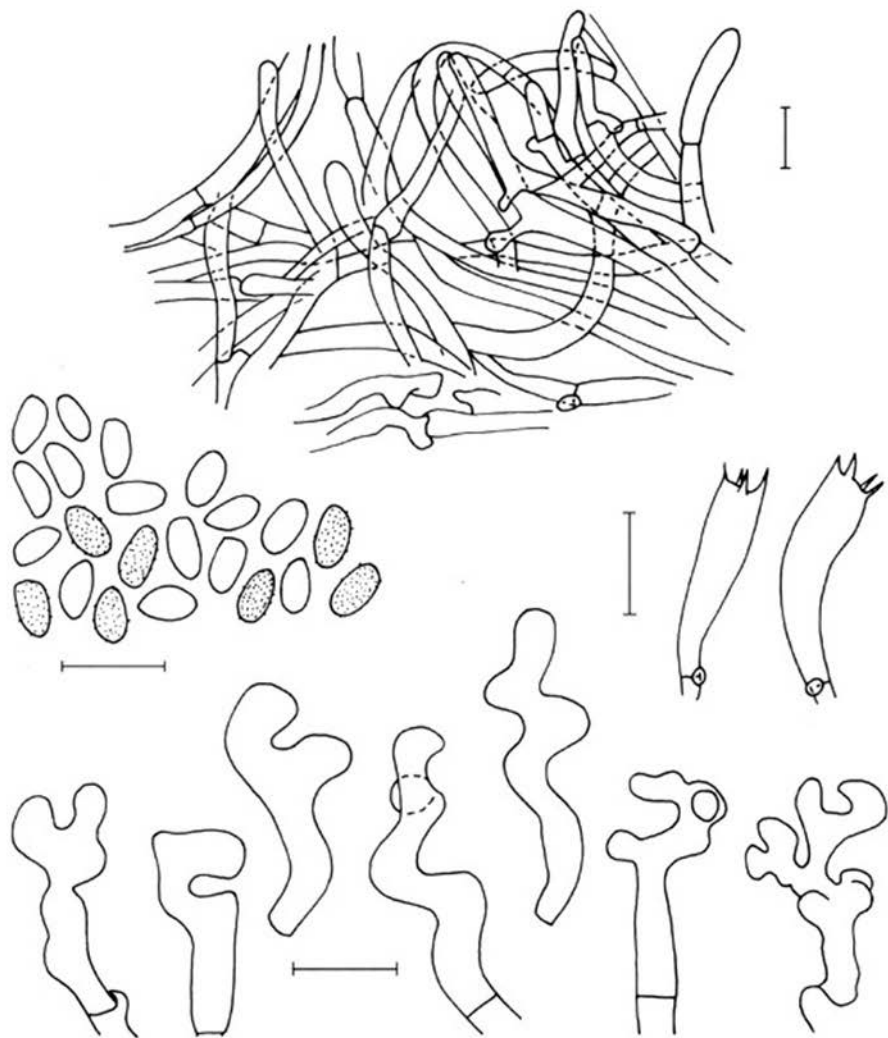


Fig. 19. *Crepidotus variabilis* (no. 6612). Pileipellis, spores, basidia, and cheilocystidia. Bar = 10 μ m.

Spores 7.2–9.1 \times 4.3–5.3 μ m, $Q = 1.5$ –1.9, av. vol. = 97 μ m³, ellipsoid to oblong, faintly to coarsely marbled, rugulose, rather thin-walled, not too dark brown. Basidia 22–24 \times 6–8 μ m, 4-spored, clamped. Cheilocystidia 30–53 \times 7–10 μ m cylindrical, more rarely narrowly lageniform, bottle-shaped, slightly flexuous, often slightly capitate. Pileipellis a cutis, with a rather dense hypocutis. Clamps abundant. No pigment.

Ecology — Saprotrophic, gregarious on rotten *Quercus*(?) twig in subtropical mixed forest (mainly *Quercus*, *Pinus*).

Collection examined. MEXICO: Federal State Mexico, Nevada de Toluca, Temascaltepec, c. 132 km W. of Mexico City, El Polvador, 4 July 1996, I. Krisai-Greilhuber & H. Voglmayr no. 6474 (WU 20092).

The character combination indicates a close relationship with a species described from Europe, i.e. *Crepidotus subverrucisporus*. Recent studies by Bandala et al. (1999) with type studies and SEM observations on the spore ornamentation could demonstrate that indeed Mexican collections can be attributed to this species.

Crepidotus variabilis (Pers.: Fr.) Kumm. — Fig. 19

Crepidotus variabilis (Pers.: Fr.) Kumm., Führ. Pilzk. (1871) 74.
Selected literature: Senn-Irlet (1995).

Pileus 5–10 mm in diam., circular to shell-shaped, carpophores partly fused, white, white tomentose-hairy, not gelatinous. Lamellae straight adnate to sinuate, crowded, lamellar edge white denticulate, young white, then dirty brownish, no pink tinge. Stipe up to 3 mm long, white, tomentose. Context thin, white, smell none, taste mild.

Spores 5.6–6.4 × 2.9–4.0 µm, Q = 1.5–2.0, av. vol. = 37 µm³, elongate to subcylindrical, distinctly finely punctate-warty, with warts hardly visible in optical section, moderately coloured. Basidia 19–25 × 4–6 µm, 4-spored, clamped. Cheilocystidia 20–30 × 4–5 µm, cylindrical to narrowly clavate, flexuous, often branched and contorted, with finger-like protuberances. Pileipellis a thick trichoderm of undifferentiated, often bent hyphae. Clamp-connections in all parts of the carpophore. No pigment observed in trama and pellis.

Ecology — Saprotrophic, gregarious on rotten twig of deciduous tree in a sclerophilous *Quercus* forest.

Collection examined. MEXICO: Federal State Mexico, El Ocotil, Chapa de Mota near Mexico City, 8 July 1996, I. Krisai-Greilhuber & H. Voglmayr no. 6612.

In all respects identical with European collections of this species known to us. Reported from several places in North America (Hesler & Smith, 1965), however apparently not yet known from the Neotropics.

ACKNOWLEDGEMENTS

We thank Margrit Kummer (Bern) for inking the figures and providing us with line drawings of the carpophores based on colour slides, and H. Halbritter and F. Zweili for taking the SEM pictures. Ron H. Petersen and Joaquin Cifuentes Blanco are gratefully acknowledged for providing excellent support to IKG and HV during their trip to the south-eastern USA and Mexico. IKG wishes to thank the Hesler Fund and HV the Lindsay S. Olive Fund for financial support, and the Highlands Biological station for providing working and living facilities. We also thank the herbarium curators for the loans of type material.

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CONTRIBUTIONS TOWARDS A MONOGRAPH OF PHOMA
(COELOMYCETES) VIII

Section *Paraphoma*: Taxa with setose pycnidia

J. DE GRUYTER¹ & G.H. BOEREMA²

In this paper eleven species of *Phoma* with obvious setose pycnidia, grouped in the section *Paraphoma*, are documented and described. Most of these species were formerly classified in *Pyrenochaeta*. The following new taxa have been proposed: *Phoma briardii* nom. nov., *Phoma carteri* nom. nov., *Phoma glycinicola* nom. nov., *Phoma indica* (T.S. Viswan.) comb. nov., *Phoma setariae* (H.C. Greene) comb. nov. and *Phoma leveillei* var. *microspora* var. nov. Indices on host/substratum-fungus and fungus-host relations are included and short comments on the ecology and distribution of the taxa are given.

The following sections have already been treated in this series of 'Contributions towards a Monograph of *Phoma*': sect. *Phoma* sensu stricto (De Gruyter & Noordeloos, 1992; De Gruyter et al., 1993, 1998), sect. *Peyronellaea* (Boerema, 1993), sect. *Plenodomus* (Boerema et al., 1994, 1996; Boerema & de Gruyter, 1999), sect. *Heterospora* (Boerema et al., 1997, 1999), sect. *Sclerophomella* (Boerema & de Gruyter, 1998) and sect. *Phyllostictoides* (Van der Aa et al., 2001). For the collective and differentiating characters of these sections see Van der Aa et al. (1990) and Boerema (1997).

The present paper deals with the section *Paraphoma*, originally described as a separate genus.

Phoma sect. **Paraphoma** (Morgan-Jones & J.F. White) Boerema

Phoma sect. *Paraphoma* (Morgan-Jones & J.F. White) Boerema in Van der Aa et al., Stud. Mycol. 32 (1990) 7.

Paraphoma Morgan-Jones & J.F. White, Mycotaxon (1983) 59–60.

Type: *Paraphoma radicina* (McAlpine) Morgan-Jones & J.F. White. — *Phoma radicina* (McAlpine) Boerema (this paper no. 3).

The species of this section are characterized by a copious production of mainly septate setae on the surface of the relatively thick-walled, pseudoparenchymatous and distinctly ostiolate pycnidia. The conidia are always one-celled both in vivo and in vitro. The setae may be stiff or rather hyphal-like and either short or relatively long. They may be scattered over the entire surface of the pycnidium as shown in the type species of the section, *Phoma radicina* (Fig. 1A), but often they are most abundant around the ostiole (Fig. 1B and Table I). Pycnidia with mainly setae around the ostiole superficially closely resemble those of the genus *Pyrenochaeta* De Not. emend. Schneider (1979). That genus, however, is charac-

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terized by elongated, branched conidiophores instead of simple doliiform or ampulliform conidiogenous cells³.

Some species of this section produce single chlamydo-spores, solitary or in series and complexes. So far none of the species has been associated with a teleomorph.

It is curious that most species of *Phoma* sect. *Paraphoma* are typical soil fungi, often associated with monocotyledonous plants (Gramineae, Amaryllidaceae, Iridaceae, Liliaceae, Orchidaceae and Zingiberaceae).

MATERIAL AND METHODS

The isolates used in this study were obtained by the Plant Protection Service (PD) and deposited at the CBS (Utrecht, formerly Baarn). The methodology used conforms with that described in Contributions I–1 & 2 of this series (De Gruyter & Noordeloos, 1992 and De Gruyter et al., 1993). See also Contribution VII (Boerema & de Gruyter, 1998). Growth-rate of colonies on oatmeal agar (OA), malt agar (MA) and cherry-decoction agar (CA) are diameters after 7 days, unless otherwise stated.

KEY TO THE SPECIES AND VARIETIES OF PHOMA SECT. PARAPHOMA

Based on characteristics *in vitro*; see also the Appendix

- 1a. Chlamydo-spores absent 2
- b. Chlamydo-spores present 9
- 2a. Characteristic fragmentation of hyphae occurs (Fig. 13), colony greenish to rosy vinaceous/orange on OA, conidia 3.5–6.0 × 1.5–3.0 µm, soil- and air-borne saprophyte, probably cosmopolitan 1. *P. septicialis*
- b. Fragmentation of hyphae absent 3
- 3a. Conidia very small, subglobose, not exceeding 3.5 µm, colony greenish, often with coral pigmentation, on bark and wood *Quercus* spp. in North America and Europe 2. *P. carteri*
- b. Conidia exceeding 3.5 µm 4
- 4a. Colony distinct pale luteous on OA, due to a diffusible pigment production, conidia 3.0–6.0 × 1.0–3.0 µm, cosmopolitan soil-borne fungus, saprophytic, particularly on root surfaces; also isolated from cysts of nematodes 3. *P. radicina*
- b. Colony on OA greenish, greyish, brownish or vinaceous 5

3) It should be noted that at present also included in *Pyrenochaeta* are species producing both undifferentiated discrete conidiogenous cells and conidiogenous cells integrated on branched conidiophores in a single pycnidium, e.g. *Pyrenochaeta corni* (Bat. & A.F. Vital) Boerema et al. (1996) and *Pyrenochaeta dolichi* Mohanty (1958). With reference to the discussion of the taxonomic position of the latter by Grondona et al. (1997) we now think that these species can be better treated as 'setose species' of *Pleurophoma* Höhn. Our recent isolates of the type species of that genus, *Pleurophoma pleurospora* (Sacc.) Höhn., and the related *Pleurophoma cava* (Schulzer) Boerema et al. (1996) also showed the presence of both undifferentiated and integrated conidiogenous cells. All these species have probably more affinity with *Phoma* than with true species of *Pyrenochaeta* (teleomorph, where known, belonging to *Herpotrichia* Fuckel, see Schneider, 1979). Concerning *Pyrenochaeta dolichi*, Grondona et al. (1997) also produce evidence which supports this idea.

- 5a. Colony vinaceous on OA, due to production of pigmented grains of exudate, conidia $4.0-6.0 \times 2.0-2.5 \mu\text{m}$, cosmopolitan soil-borne root pathogen of *Allium* spp., esp. *A. cepa*, also in rhizosphere of other crops 4. *P. terrestris*
- b. Colony on OA greenish, greyish or brownish 6
- 6a. Conidia not exceeding $5.5 \mu\text{m}$ 7
- b. Conidia exceeding $5.5 \mu\text{m}$ 8
- 7a. Colony greenish to greyish on OA, conidia $4.0-5.5 \times 2.0-2.5 \mu\text{m}$, cosmopolitan soil fungus, saprophytic, or opportunistic root pathogen 5a. *P. leveillei* var. *leveillei*
- b. Colony greenish on OA, conidia $3.5-4.5 \times 1.5-2.0 \mu\text{m}$, probably also a cosmopolitan soil fungus 5b. *P. leveillei* var. *microspora*
- 8a. Average length-width ratio (Q) conidia < 3 , colony white to greyish/greenish/brownish on OA, conidia $5.0-7.5 \times 2.0-2.5 \mu\text{m}$, sclerotial bodies covered by short setae present, a pathogen of *Glycine* spp. in Africa 6. *P. glycinicola*
- b. Average length-width ratio (Q) conidia > 3 , colony colourless to greenish/brownish on OA, conidia cylindrical to allantoid, $4.5-7.0 \times 1.0-2.0 \mu\text{m}$, sclerotial bodies absent, soil-borne fungus in Europe, also recorded from roots 7. *P. briardii*
- 9a. Growth-rate fast, 50-70 mm on OA, conidia highly variable and relatively large, $3.5-10.5 \times 1.5-4.5 \mu\text{m}$, colony greenish on OA, common soil-borne fungus in India, opportunistic pathogen 8. *P. gardeniae*
- b. Growth-rate slow to moderate, 10-45 mm on OA 10
- 10a. Growth-rate slow, 10-15 mm on OA, conidia $4.0-5.5 \times 1.5-2.5 \mu\text{m}$, colony brownish on OA and MA, NaOH reaction greenish (not an E⁺ reaction), on leaf spots of *Saccharum officinarum* in India 9. *P. indica*
- b. Growth-rate slow to moderate, ≥ 25 mm on OA 11
- 11a. Growth-rate slow to moderate, 25-30 mm on OA, conidia $3.0-5.0 \times 1.5-2.0 \mu\text{m}$, colony greyish to greenish on OA, NaOH reaction negative, soil-borne fungus in western Europe, especially in agricultural fields, isolated from cysts of nematodes
10. *P. terricola*
- 11b. Growth-rate moderate, 35-45 mm on OA, conidia $4.0-6.0 \times 2.0-2.5 \mu\text{m}$, colony vinaceous on OA, NaOH reaction vinaceous/violet on OA (not an E⁺ reaction), common soil fungus in North and South America; world-widely associated with root rot of *Allium* spp., esp. *A. cepa*; roots of other plants also may be affected 4. *P. terrestris*

HOST/SUBSTRATUM-FUNGUS INDEX

(Including the Appendix)

Plurivorous (but often with preference for monocotyledonous plants, see below): *P. briardii* (no. 7) (common in Europe), *P. gardeniae* (8) (common in India), *P. leveillei* var. *leveillei* and var. *microspora* (5a, 5b) (worldwide), *P. radicina* (3) (recorded from Australia, Eurasia and North America), *P. septicialis* (1) (recorded from Europe and Africa), *P. terrestris* (4) (common in America, but also elsewhere), *P. terricola* (10) (common in Europe).

Isolated from soil: *P. briardii* (7), *P. leveillei* var. *leveillei* (5a), *P. radicina* (3), *P. septicialis* (1), *P. terrestris* (4), *P. terricola* (10).

Isolated from cysts of phytonematodes: *P. radicina* (3), *P. terricola* (10).

Isolated from air: *P. gardeniae* (8), *P. septicialis* (1).

Isolated from water: *P. leveillei* var. *microspora* (5b).

HOSTS NOTED IN THIS PAPER

Fagaceae

- Quercus* spp. no. 2: *P. carteri*
 (Disease: 'Pyrenochaeta-Dieback', [Europe and North America]
 but the *Phoma* is probably not the
 primary cause)

Leguminosae

- Glycine* spp. no. 6: *P. glycinicola*
 (Disease: Leaf Spot) [widespread in Africa]
Glycine max (roots) no. 1: *P. septicidialis*
 [Africa]

Monocotyledonae:

Amaryllidaceae

- Narcissus* sp. (roots) no. 5a: *P. leveillei* var. *leveillei*
 [Europe]

Gramineae

- Oryza sativa* (roots) no. 5a: *P. leveillei* var. *leveillei*
 no. 4: *P. terrestris*
 [records from Asia and North America]

- Pennisetum typhoides* no. 11: *P. setariae* (Appendix)
 (Disease: Leaf Spot) [record from Africa]

- Saccharum officinarum* no. 9: *P. indica*
 (Disease: Leaf Spot) [recorded in India]

- no. 11: *P. setariae* (Appendix)
 [record from South America]

- Secale cereale* (roots) no. 3: *P. radicina*

- no. 5a: *P. leveillei* var. *leveillei*
 [both worldwide distributed]

- no. 7: *P. briardii*
 [so far only known from Europe]

- Setaria lutescens* (roots) no. 4: *P. terrestris*
 (Disease: Leaf Spot) no. 11: *P. setariae* (Appendix)

- [both records from North America]

- Zea mays* (roots) no. 4: *P. terrestris*
 [record from North America]

Iridaceae

- Iris* spp. (roots) no. 3: *P. radicina*
 [record from Europe]

Liliaceae

- Allium* spp., esp. *A. cepa* no. 4: *P. terrestris*
 (Disease: Pink Root) [worldwide]

Orchidaceae

- Phalaenopsis* sp. no. 7: *P. briardii*
 [record from Europe]

Zingiberaceae

- Elettaria cardamomum* no. 5b: *P. leveillei* var. *microspora*
 [record from Central America]

FUNGUS-HOST RECORDS IN THIS PAPER

- P. briardii* (7) e.g. *Secale cereale*, *Milium effusum*
(Gramineae)
Phalaenopsis sp.
(Orchidaceae)
- P. carteri* (2) *Quercus* spp.
(Fagaceae)
- P. gardeniae* (8) e.g. *Gardenia jasminoides*
(Rubiaceae)
Arachis hypogaea
(Leguminosae)
- P. glycinicola* (6) *Glycine* spp.
(Leguminosae)
- P. indica* (9) *Saccharum officinarum*
(Gramineae)
- P. leveillei* var. *leveillei* (5a) e.g. *Oryza sativa*, *Secale cereale*
(Gramineae)
Narcissus sp.
(Amaryllidaceae)
Fragaria × *ananassa*
(Rosaceae)
- P. leveillei* var. *microspora* (5b) e.g. *Elettaria cardamomum*
(Zingiberaceae)
- P. radicina* (3) e.g. *Secale cereale*
(Gramineae)
Iris spp.
(Iridaceae)
Lycopersicon esculentum
(Solanaceae)
Malus sylvestris
(Rosaceae)
- P. septicalis* (1) e.g. *Glycine max*
(Leguminosae)
- P. setariae* (11) (Appendix) e.g. *Pennisetum typhoides*, *Saccharum officinarum*, *Setaria lutescens*
(Gramineae)
- P. terrestris* (4) e.g. *Allium* spp., esp. *A. cepa*
(Liliaceae)
Oryza sativa, *Setaria lutescens*, *Zea mays*
(Gramineae)
Calathea crocata
(Maranthaceae)

Table I. Characteristics of setae in species of *Phoma* sect. *Paraphoma*.

<i>Phoma</i> sect. <i>Paraphoma</i>	setae short, up to 100 µm	setae of moderate length	setae long, exceeding 200 µm	setae mainly around ostiole	setae scattered over pycnidium
<i>P. indica</i> (9)	+			+	
<i>P. gardeniae</i> (8)	+			+	
<i>P. terricola</i> (10)	+			+	
<i>P. leveillei</i> var. <i>microspora</i> (5b)	+			+	+
<i>P. setariae</i> (11)	+			+	
<i>P. carteri</i> (2)		+			+
<i>P. glycinicola</i> (6)		+		+	+
<i>P. briardii</i> (7)			+	+	+
<i>P. leveillei</i> var. <i>leveillei</i> (5a)			+		+
<i>P. radicina</i> (3)			+		+
<i>P. septicidialis</i> (1)			+		+
<i>P. terrestris</i> (4)			+	+	

DESCRIPTIVE PART

Section *Paraphoma*

1. *Phoma septicidialis* Boerema — Figs. 2, 13

Phoma septicidialis Boerema in Boerema & Dorenbosch, Versl. Meded. plziektenk. Dienst Wageningen 153 (Jaarb. 1978) (1979) 20. — *Pyrenochaeta telephii* Allesch., Ber. bayer. bot. Ges. 4 (1896) 33; not *Phoma telephii* (Vestergr.) Kesteren, Neth. J. Pl. Path. 78 (1972) 117.

Selected literature. Boerema & Dorenbosch (1979).

Description in vitro

OA: growth-rate 22–40 mm, (14 days: 52–77 mm), regular to slightly irregular, with felty, (pale) olivaceous grey/grey olivaceous aerial mycelium; colony citrine/greenish olivaceous to dull green, rosy vinaceous to orange towards margin; reverse similar.

MA: growth-rate 21–39 mm, (14 days: 41–72 mm), regular to slightly irregular, with compact finely floccose to felty aerial mycelium; colony grey olivaceous/dull green, or honey/amber, with buff to rosy buff near margin; reverse similar.

CA: growth-rate 21–31 mm (14 days: 40–63 mm), regular to slightly irregular, with felty, white to grey olivaceous aerial mycelium; colony honey or dull green; reverse greenish olivaceous/honey or dull green, with olivaceous/olivaceous black or leaden grey/leaden black near centre.

Pycnidia setose, 70–170 µm diam., globose to subglobose, solitary or confluent, with 1 (or 2) non-papillate ostioles, honey to olivaceous, later olivaceous black; setae relatively long, exceeding 200 µm, spread over the upper surface; walls made up of 2–8 layers of cells, sometimes partly thicker due to protruding of cells into the pycnidial cavity, outer

layers pigmented; with white coloured conidial exudate; abundant, mainly on the agar; micropycnidia present, 25–50 µm diam. Conidiogenous cells 3–6 × 3–6 µm, bottle-shaped. Conidia aseptate, 3.5–5(–6) × 1.5–3 µm, av. 4.0–4.5 × 1.8–2.3 µm, Q = 1.5–3.0, av. Q = 2.0–2.3, subglobose to ellipsoidal, with several small or large guttules.

Chlamydospores absent.

NaOH spot test: rosy vinaceous margin may discolour to livid violet/purple on OA.

Crystals absent.

Note: a characteristic fragmentation of the hyphae occurs (Fig. 13).

Ecology and distribution. In Europe a widespread soil- and air-borne saprophyte ('pioneer flora'). The fungus is also found in Africa and may be equally common in others parts of the world. The epithet '*septicidalis*' refers to the easy fragmentation of the hyphae in vitro.

Representative cultures. CBS 112.79 (PD 74/1018) ex air, Finland; CBS 101636 (PD 86/1186) ex root *Glycine max* (Leguminosae), Zimbabwe.

2. *Phoma carteri* De Gruyter & Boerema, *nom. nov.* — Fig. 3

Pyrenochaeta minuta J.C. Carter, Bull. Ill. nat. Hist. Surv. 21 (1941) 214 [replaced synonym; type specimen pure culture isolated from bark of *Quercus palustris*, coll. J.C. Carter, Xenia, Clay County, Illinois, USA, Oct. 8, 1937]; not *Phoma minuta* Wehm., Mycologia 38 (1946) 318, nor *Phoma minuta* Alcalde, An. Jard. bot. Madr. 10 (1952) 233.

Selected literature. Carter (1941).

Description in vitro

OA: growth-rate 23–25 mm (14 days: 47–57 mm), regular to somewhat irregular, with finely floccose/finely woolly, (pale) olivaceous grey aerial mycelium; colony olivaceous buff/greenish olivaceous to grey olivaceous, often with a coral pigmentation; reverse similar.

MA: growth-rate 19–20 mm (14 days: 49–51 mm), regular to somewhat irregular, with compact woolly to floccose, pale olivaceous grey aerial mycelium; colony buff to citrine/greenish olivaceous, with olivaceous grey at centre, also with salmon to flesh coloured patches, with amber margin; reverse similar.

CA: growth-rate 23–24 mm, (14 days: 48–61 mm), regular, with finely floccose to finely woolly, (pale) olivaceous grey aerial mycelium; colony greenish olivaceous to pale luteous, often with a coral pigmentation; reverse similar, with olivaceous/olivaceous black at centre.

Pycnidia setose, 80–230 µm diam., globose, solitary or confluent, with 1 (or 2) non-papillate ostioles, greenish olivaceous/olivaceous, later olivaceous black; setae of moderate length, up to 200 µm, spread over the upper surface; walls made up of 2–6 layers of cells, sometimes partly thicker due to protruding of cells into the pycnidia cavity, outer layers pigmented; with buff/rosy buff coloured conidial exudate; on the agar and in aerial mycelium. Conidiogenous cells 3–5 × 3–6 µm, globose to bottle-shaped. Conidia aseptate, 2.5–3.5 × 2–2.5 µm, av. 3.1 × 2.3 µm, Q = 1.0–1.6, av. Q = 1.3, subglobose, with 1(–2) minor guttules.

Chlamydospores absent.

NaOH spot test: coral pigmentation discolours to violet on OA, amber pigmentation discolours to orange on MA.

Crystals absent.

Ecology and distribution. Isolated from discoloured bark and wood of different species of oaks (*Quercus alba*, *Q. palustris* and *Q. suber*) in North America (USA, Illinois) and Europe (the Netherlands, Spain). Although die-back has been attributed to this fungus (USA), it is probably only an opportunistic pathogen.

Representative culture. CBS 101633 (PD 84/74) ex *Quercus* sp. (Fagaceae), the Netherlands.

3. *Phoma radicina* (McAlpine) Boerema — Figs. 1A, 4

Phoma radicina (McAlpine) Boerema in Boerema & Dorenbosch, Versl. Meded. plziektenk. Dienst Wageningen 153 (Jaarb. 1978) (1979) 20. — *Pyrenochaeta radicina* McAlpine, Fung. Dis. Stone-Fruit Austr. (1902) 127. — *Paraphoma radicina* (McAlpine) Morgan-Jones & J.F. White, Mycotaxon 18 (1983) 60.

Selected literature. Morgan-Jones & White (1983), Boerema (1985).

Description in vitro

OA: growth-rate 29–30 mm (14 days: 56–57 mm), regular, with woolly, pale olivaceous grey aerial mycelium; colony pale luteous, due to production of a diffusible pigment, with coral concentric zones; reverse pale luteous to amber.

MA: growth-rate 22–24 mm (14 days 29–44 mm), regular, with compact, finely floccose to woolly, greenish grey aerial mycelium; colony olivaceous grey to greenish grey, with amber due to production of a diffusible pigment; reverse citrine to amber, partly olivaceous to olivaceous black.

CA: growth-rate 25–27 mm (14 days: 52–54 mm), regular, with finely floccose to woolly, (pale) olivaceous grey aerial mycelium; colony buff to pale olivaceous grey/greenish grey; reverse pale luteous to sienna/dark brick.

Pycnidia setose, 180–450 µm diam., globose to subglobose, mostly solitary, with 1 (or 2) non-papillate or papillate ostioles, honey/olivaceous, later olivaceous black; setae relatively long, exceeding 200 µm, spread over the upper surface; walls made up of 3–7 layers of cells, outer layers pigmented; with off-white to buff coloured conidial exudate; abundant, mainly on the agar. Conidiogenous cells 4–7 × 3–7 µm, bottle-shaped. Conidia aseptate, (3–)4–6 × (1–)2–3 µm, av. 5.4 × 2.6 µm, Q = 1.7–2.4, av. Q = 2.1, ellipsoidal to subglobose, usually with several guttules.

Chlamydospores absent.

NaOH spot test: a greenish discolouring may occur on OA.

Note: red pigmented grains of exudate, resembling small crystals, are produced in culture media.

Ecology and distribution. Recorded from a wide variety of woody and herbaceous plants in Australia, Eurasia and North America. Very often isolated from root surfaces (e.g. *Iris* spp. and *Secale cereale*). Also from bulbs, cysts of nematodes and various soil samples. The fungus may be regarded as harmless or saprophytic. It represents the type of the section *Paraphoma*.

Representative culture. CBS 111.79 (PD 76/437) ex *Malus sylvestris* (Rosaceae), the Netherlands; CBS 102875 (PD 78/1097) ex *Lycopersicon esculentum* (Solanaceae), Germany.

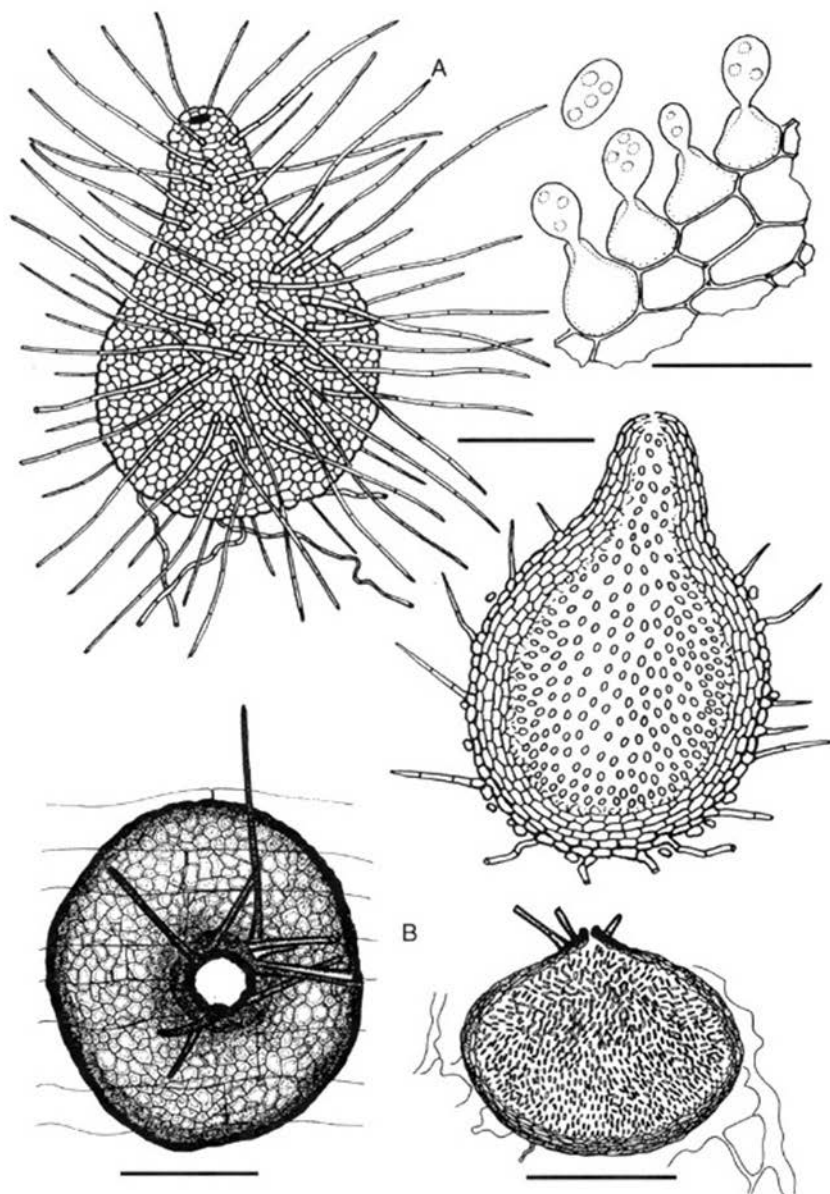
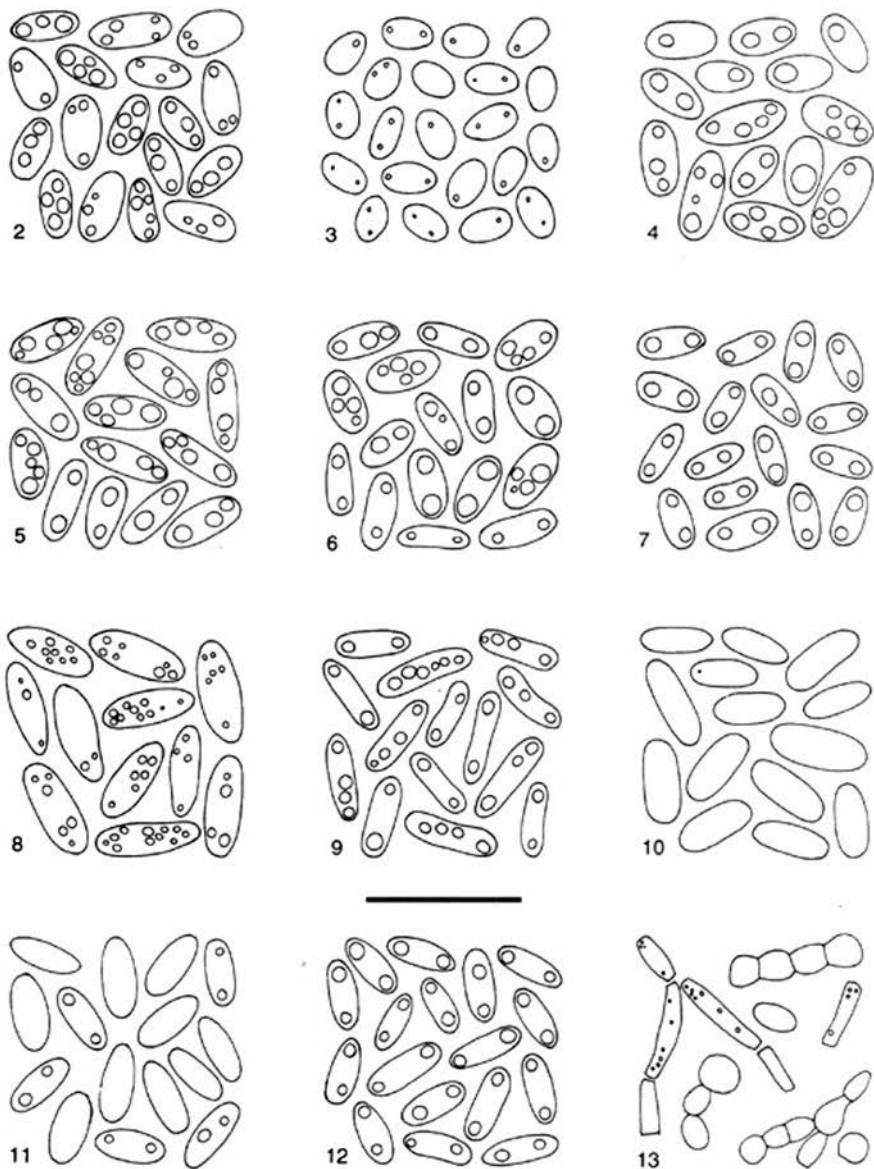


Fig. 1. A. *Phoma radicina*, type species of *Phoma* sect. *Paraphoma*. Surface view and vertical section of pycnidium with setae scattered over the entire pycnidial wall. Inner part of wall with conidiogenous cell; B. *Phoma terrestris*, surface view and vertical section of pycnidium with setae around the ostiole. Drawing A after Morgan-Jones & White (1983; with permission), B after Punithalingam & Holliday (1973; with permission). — Bar = 100 μ m for pycnidia and 10 μ m for conidiogenous cells and conidia.



Figs. 2–12. Conidia. 2. *Phoma septicaldis*; 3. *Phoma carteri*; 4. *Phoma radicina*; 5. *Phoma terrestris*; 6. *Phoma leveillei* var. *leveillei*; 7. *Phoma leveillei* var. *microspora*; 8. *Phoma glycinicola*; 9. *Phoma briardii*; 10. *Phoma gardeniae*; 11. *Phoma indica*; 12. *Phoma terricola*; Fig. 13. *Phoma septicaldis*, fragmentation of the hyphae. — Bar = 10 μ m.

4. *Phoma terrestris* H. N. Hansen — Figs. 1B, 5, 14

Phoma terrestris H. N. Hansen, *Phytopathology* 19 (1929) 699. — *Pyrenochaeta terrestris* (H. N. Hansen) Gorenz, J. Walker & Larson, *Phytopathology* 38 (1948) 838; not *Phoma terrestris* Saksena, Nand & A. K. Sarbhoy, *Mycopath. Mycol. appl.* 29 (1966) 86 [= *Phoma multirostrata* var. *macrospora* Boerema; de Gruyter et al., 1998].

Selected literature. Hansen (1929), Punithalingam & Holliday (1973), Boerema (1985).

Description in vitro

OA: growth-rate 35–45 mm after 7 days, regular, with felty to finely woolly, pale olivaceous grey aerial mycelium; colony brick to vinaceous, or partly primrose, often with dull green patches; reverse similar, often with fulvous to rust patches.

MA: growth-rate 15–40 mm after 7 days, regular or slightly irregular, with felty to finely woolly, pale olivaceous grey to smoke grey aerial mycelium; colony rosy vinaceous to vinaceous/brick, with buff near margin and hazel at centre; reverse similar, brown vinaceous at centre.

CA: growth-rate 28–35 mm after 7 days, regular, with felty to finely woolly, pale olivaceous grey aerial mycelium; colony rosy vinaceous to vinaceous or vinaceous buff to hazel; reverse similar, partly grey olivaceous/olivaceous grey, and hazel to olivaceous/olivaceous black at centre.

Pycnidia setose, 120–370 µm diam., globose to subglobose, solitary or confluent, with 1(–3) usually papillate ostioles, honey, later olivaceous to olivaceous black; setae relatively long, exceeding 200 µm, mainly concentrated around the ostiole; walls made up of 4–11 layers of cells, outer layer(s) pigmented; with white coloured conidial exudate; scattered or in concentric rings, mostly on the agar. Conidiogenous cells 4–8 × 4–7.5 µm, globose to bottle shaped. Conidia aseptate, 4–6 × 2–2.5 µm, av. 5.0 × 2.3 µm, Q = 1.8–2.9, av. Q = 2.2, ellipsoidal, with several distinct guttules.

Chlamydospores may be present, globose to subglobose, solitary or aggregated, ochraceous to olivaceous, with greenish guttules, intercalary or terminal, 6–12 µm diam.

NaOH spot test: brick to vinaceous pigments becoming vinaceous/violet on OA.

Note: vinaceous or amber pigmented grains of exudate, resembling small crystals, may be produced in culture media.

Ecology and distribution. This well-known causal organism of Pink Root of onion (*Allium cepa*) is apparently a widely distributed soil fungus in North America (USA and Canada) and probably also a common soil inhabitant in some regions of South America (Argentina, Brazil, Venezuela). Records from Europe, Africa and Australia are generally associated with the cultivation of onions or other crops of *Allium* (leek, shallot, garlic and chive). The fungus is frequently isolated from the roots of grasses (e.g. *Setaria lutescens*) and other herbaceous plants, but usually without any disease symptoms. However, the roots of maize plants (*Zea mays*) and rice (*Oryza sativa*) may also be affected. The fungus is characterized by a red pigment in the mycelium and this easily distinguishes it from the morphologically very similar *Phoma leveillei* Boerema & Bollen var. *leveillei* (no. 5a) and from *Phoma terricola* Boerema (no. 10).

Representative cultures. CBS 377.52 ex *Allium cepa* (Liliaceae), CBS 732.97 (PD 94/379) ex *Calathea crocata* (Maranthaceae), the Netherlands; CBS 335.87 (PD 2000/8963) ex *Allium cepa* (Liliaceae), Senegal.

5a. *Phoma leveillei* Boerema & G.J. Bollen var. *leveillei* — Fig. 6

Phoma leveillei Boerema & G.J. Bollen, Persoonia 8 (2) (1975) 115, var. *leveillei* [autonym created by the separation of the variety *microspora*, see below]. — *Vermicularia acicola* Moug. & Lév. apud Lévillé, Anns Sci. nat. (Bot.) III, 9 (1848) 259–260 [as 'Moug. Lév.']; not *Phoma acicola* (Moug. & Lév.) Sacc., Sylloge Fung. 3 (1884) 100 [as '(Lév.) Sacc.']; = *Sclerophoma pythiophila* (Corda) Höhn.]. — *Pyrenochaeta acicola* (Moug. & Lév.) Sacc., Sylloge Fung. 3 (1884) 220 [as '(Lév.) Sacc.']. — *Pyrenochaeta phlogis* Masee, Bull. misc. Inf. R. bot. Gdns Kew (1907) 241; not *Phoma phlogis* Room., Revue mycol. 6 (1884) 160 [= *Phoma acuta* (Hoffm.: Fr.) Fuckel subsp. *acuta* f. sp. *phlogis*, see Boerema et al., 1994: 465].

Pyrenochaeta oryzae Shirai ex I. Miyake, J. Coll. Agric. imp. Univ. Tokyo 2 (4) (1910) 255–256; not *Phoma oryzae* Catt., Arch. Bot. crittog. Pavia 2–3 (1877) 118; not *Phoma oryzae* Cooke & Masee, Grevillea 16 (1887) 15 [= *Phoma minutispora* P.N. Mathur, see de Gruyter & Noordeloos, 1992: 75]; not *Phoma oryzae* Hori, 'Nosakubutsu-Biyogatu' (1903) 111–113 [in Japanese].

Pyrenochaeta lupini Sibilia, Annali Bot. 18 (1930) 284; not *Phoma lupini* Ellis & Everh., Bull. Washburn [Coll.] Lab. nat. Hist. 1 (1884) 6 [see de Gruyter et al., 1992: 375]; not *Phoma lupini* Buchw. in Möller, Fungi Faeröes 2 (1958) 153.

Pyrenochaeta calligoni Kratzev apud Schwarzman & Kratzev, Trudy Inst. Bot., Alma Ata 9 (1961) 45; not *Phoma calligoni* Murashk., Trans. Agric. Forest. Omsk 9 (1928) 6.

Pyrenochaeta spinaciae Verona & Negru in Negru & Verona, Mycopath. Mycol. appl. 30 (1966) 309–310; not *Phoma spinaciae* Bubák & K. Krieg. in Bubák, Anns mycol. 10 (1912) 47 [= *Phoma betae* Frank, see Boerema et al., 1987].

Pyrenochaeta anthyllidis Manoliu & Mítítíuc, Reprim nov. Spec. Regni veg. [Feddes Reprim] 87 (1976) 142.

Selected literature. Dorenbosch (1970), Boerema & Hamers (1989).

Description in vitro

OA: growth-rate 21–24 mm (14 days: 40–48 mm), regular to somewhat irregular, with woolly, (pale) olivaceous grey aerial mycelium; colony grey olivaceous/olivaceous grey to dull green; reverse similar.

MA: growth-rate 16–20 mm (14 days: 30–40 mm), regular to somewhat irregular, with compact, finely floccose to woolly, (pale) olivaceous grey aerial mycelium; colony olivaceous grey, becoming grey olivaceous at margin; reverse olivaceous black, with olivaceous buff/grey olivaceous to leaden grey at margin.

CA: growth-rate 19–24 mm (14 days: 40–45 mm), regular to somewhat irregular, with compact, felty to woolly, (pale) olivaceous grey aerial mycelium; colony buff to grey olivaceous/olivaceous grey; reverse olivaceous grey to purplish grey, buff/saffron at margin.

Pycnidia setose, 180–270 µm diam., globose to subglobose, with usually 1, non-papillate or slightly papillate ostiole, olivaceous to olivaceous black; setae relatively long, exceeding 200 µm, spread over the upper surface; walls made up of 2–7 layers of cells, outer layers pigmented; with white to buff coloured conidial exudate; abundant, scattered or in concentric rings, on the agar as well as in aerial mycelium. Conidiogenous cells 3–7 × 3–7 µm, subglobose to bottle-shaped. Conidia aseptate, 4–5.5 × 2–2.5 µm, av. 4.4 × 2.4 µm, Q = 1.6–2.3, av. Q = 1.8, subglobose to ellipsoidal, with 2 or more, distinct guttules.

Chlamydospores absent, but hyphal swollen cells may occur.

NaOH spot test: negative.

Note: luteous to ochraceous pigmented grains of exudate, resembling small crystals, may be produced in culture media.

Ecology and distribution. A worldwide soil fungus (Eurasia, North America, Africa, Australia), regarded as a collective species with much variability in morphological and physiological characters. Generally it behaves like a saprophyte; all listed synonyms being associated with necrotic plant tissue. However, the basal and underground parts of monocotyledonous plants may be affected by it (reported from e.g. *Oryza sativa*, *Secale cereale* and *Narcissus* spp.). The fungus has been confused with morphologically very similar soil fungi: *Phoma terrestris* Hansen (no. 4; characterized by the production of a red pigment) and *Phoma terricola* Boerema (no. 10; distinguished by abundant production of chlamydospores).

Representative cultures. CBS 260.65 ex wheat field soil, Germany; CBS 101634 (PD 76/416) ex *Fragaria* (\times) *ananassa* (Rosaceae), the Netherlands.

5b. *Phoma leveillei* var. *microspora* De Gruyter & Boerema, var. nov. — Fig. 7

A varietate *leveillei* conidiis minoribus ($3.5-4.5 \times 1.5-2 \mu\text{m}$) et setis vulgo brevioribus quam 100 μm differens.

Holotypus: HLB 999-242399, cultura exsiccata, viva CBS 102876, isolatus ex aqua in Yugoslavia.

Description in vitro

OA: growth-rate 26–28 mm (14 days: 52 mm), regular, with finely woolly, pale olivaceous grey aerial mycelium; colony grey olivaceous to dull green/dark herbage green; reverse similar to olivaceous.

MA: growth-rate 24–26 mm (14 days 47–49 mm), regular, with compact woolly/finely floccose, pale olivaceous grey aerial mycelium; colony buff, (pale) olivaceous grey at centre; reverse pale luteous to olivaceous grey, with leaden grey/leaden black at centre.

CA: growth-rate 15–17 mm (14 days: 27–29 mm), irregular, with compact, finely woolly to finely floccose, grey olivaceous aerial mycelium; colony grey olivaceous to dull green; reverse similar with fulvous patches, and an olivaceous black centre.

Pycnidia setose, (20–)80–270 μm diam., globose to subglobose, solitary or confluent, with 1 (or 2) non-papillate ostioles, greenish olivaceous/olivaceous, later olivaceous black; setae relatively short, up to 100 μm , spread over the upper surface, more densely around the ostiole; walls made up of 2–7 layers of cells, outer layers pigmented; conidial exudate off-white; scattered or in concentric rings, mainly on the agar. Conidiogenous cells 3–6 \times 3–6 μm , bottle-shaped. Conidia aseptate, $3.5-4.5 \times 1.5-2 \mu\text{m}$, av. $4.0 \times 1.7 \mu\text{m}$, $Q = 1.7-2.8$, av. $Q = 2.3$, ellipsoidal to oblong, with 2 distinct guttules.

Chlamydospores absent.

NaOH spot test: a pale greenish discolouring may occur, but this is not specific.

Crystals absent.

Ecology and distribution. In saprophytic behaviour and apparently worldwide distribution (SE Europe and Central America (fruits of *Elettaria cordamamum*, Zingiberaceae)) this newly recognized variety resembles the very variable and ubiquitous *Phoma leveillei*. Morphologically, however, it is distinguished by significantly smaller conidia and shorter setae.

Representative culture. CBS 102876 (PD 75/911) ex water, Lake of Skadar, Yugoslavia (Montenegro).

6. *Phoma glycinicola* De Gruyter & Boerema, *nom. nov.* — Fig. 8

Pyrenochaeta glycinis R. B. Stewart, Mycologia 49 (1957) 115 [replaced synonym; holotype on leaf spot of *Glycine max*, coll. R. B. Stewart, Jimma, Ethiopia, Sept. 15, 1955, BPI, compare Schneider, 1979]; not *Phoma glycinis* Sawada, Spec. Publ., Coll. Agric., Nat. Taiwan Univ. 8 (1959) 129.

Selected literature. Stewart (1957).

Description in vitro

OA: growth-rate 18–25 mm (14 days: 35–48 mm), regular, with scarce, finely felty, white aerial mycelium; colony white to pale olivaceous grey/pale dull green to brick; reverse similar.

MA: growth-rate 15–25 mm (14 days: 30–49 mm), regular to irregular due to outgrowths of sectors, with floccose to coarsely floccose, white to salmon aerial mycelium; colony white to salmon due to aerial mycelium, with pale olivaceous grey to grey olivaceous/greenish olivaceous patches; reverse salmon to saffron, with pale luteous or greenish olivaceous patches, olivaceous at centre.

CA: growth-rate 15–25 mm (14 days: 30–37 mm), regular to irregular due to outgrowths of sectors, with felty to floccose, white to pale greenish grey aerial mycelium; colony colourless, to white due to aerial mycelium, with pale greenish grey/glaucous grey to greenish olivaceous patches; reverse similar or with salmon/saffron, and a dull green to olivaceous/olivaceous black centre.

Pycnidia setose, 70–240 µm diam., globose to irregular, solitary or confluent with 1(–3) non-papillate or papillate ostioles, honey/olivaceous, later olivaceous black; setae of moderate length, up to 200 µm, spread over the upper surface, more densely around the ostiole; walls made up of 4–11 layers of cells, outer layers pigmented; with rosy buff to salmon/saffron coloured conidial exudate; abundant, mainly in concentric rings, both on and in the agar, and in aerial mycelium as well. Conidiogenous cells 4.5–9.5 × 4–6.5 µm, bottle-shaped. Conidia aseptate, 5–7.5 × 2–2.5 µm, av. 6.0 × 2.3 µm, Q = 2.2–3.3, av. Q = 2.6, ellipsoidal to subglobose, with several small guttules.

Sclerotia present, up to 600 µm, covered with very short, setae (up to 10 µm), globose to subglobose, solitary or confluent. The cell-structure of these sclerotial bodies resembles those of the pseudoparenchymatous 'pyncnosclerotia' found in some species of *Phoma* sect. *Sclerophomella* (Höhn.) Boerema et al. (Boerema & de Gruyter, 1998); after addition of iodine only the contents of the cells become red, not the walls of the cells.

Chlamydospores absent, but dark red mycelial fragments occur due to crystallization of the pigments.

NaOH spot test: brick pigments which may turn to greenish blue.

Note: reddish pigmented grains of exudate, resembling small crystals, may be produced in culture media.

Ecology and distribution. Recorded as serious pathogen of *Glycine* spp.: Leaf Spot in Africa (Ethiopia, Zambia, Zimbabwe). The primary indigenous host is probably *Glycine javanica*. Varieties of soybean, *Glycine max* (originally native of eastern Asia), appear to be very susceptible. The leaf spots, at first small, reddish brown, soon become necrotic and may fall out, giving the plants a very ragged appearance. In susceptible varieties of soybean leaf abscission is the most damaging aspect of the disease.

Representative culture. IMI 294986 ex *Glycine max* (Leguminosae), Zambia.

7. *Phoma briardii* De Gruyter & Boerema, *nom. nov.* — Fig. 9

Pyrenochaeta leptospora Sacc. & Briard, *Revue mycol.* 11 (1889) 16 [replaced synonym; holotype on stem pieces of *Milium effusum*, coll. P.A. Briard 'no. 6', near Troyes, France, PAD]. — *Pyrenochaeta spegazziniana* Trotter, *Sylloge Fung.* 25 (1931) 190 [illegitimate: nomenclaturally superfluous name⁴]; not *Phoma leptospora* Speg., *Fungi Chilens.* (1910) 145, nor *Phoma leptospora* Sacc., *Annls mycol.* 11 (1913) 553; not *Pyrenochaeta leptospora* Speg., *Ann. Mus. Buenos Aires* 20 (1910) 354.

Description in vitro

OA: growth-rate 20–22 mm, (14 days: 44–46 mm), regular, with finely velvety, pale olivaceous grey aerial mycelium; colony colourless to grey olivaceous/olivaceous; reverse similar.

MA: growth-rate 16–21 mm, (14 days: 32–41 mm), regular to somewhat irregular, with velvety to compact felty, (pale) olivaceous grey aerial mycelium; colony buff/honey to hazel/olivaceous; reverse similar to pale luteous, (pale) mouse grey at centre.

CA: growth-rate 23–26 mm (14 days: 44–48 mm), regular, with (finely) felty, pale olivaceous grey to greenish glaucous aerial mycelium; colony vinaceous buff/fawn to pale grey olivaceous, colourless near margin; reverse vinaceous buff to fawn/hazel.

Pycnidia 70–265 µm diam., setose, globose to subglobose, solitary to confluent, with usually 1 papillate or non-papillate ostiole, honey/citrine, later olivaceous to olivaceous black; setae relatively long, exceeding 200 µm, spread over the upper surface, more densely around the ostiole; walls made up of 2–6(–10) layers of cells, outer layers pigmented, with white coloured conidial exudate; scattered, both on and in the agar; micropycnidia present, mainly 20–50 µm diam. Conidiogenous cells 3–7 × 3–6 µm, sometimes with a slightly elongated neck at a later state, bottle-shaped. Conidia aseptate, 4.5–7 × (1–)1.5–2 µm, av. 5.6 × 1.6 µm, Q = 2.8–5.2, av. Q = 3.6, cylindrical to allantoid, with two or more distinct guttules.

Chlamydospores absent.

NaOH spot test: negative.

Crystals absent.

Ecology and distribution. This fungus has been repeatedly isolated from soil in agricultural fields in Europe (especially Germany and the Netherlands). The French type collection on stem debris of millet had already indicated a soil-inhabiting fungus. Other records refer specifically to roots of Monocotyledonae, Gramineae (e.g. *Secale cereale*) and Orchidaceae (*Phalaenopsis* sp.). So far there are no data on pathogenicity.

Representative culture. CBS 101635 (PD 71/1027) ex *Secale cereale* (Gramineae), the Netherlands.

8. *Phoma gardeniae* (S. Chandra & Tandon) Boerema — Figs. 10, 15

Phoma gardeniae (S. Chandra & Tandon) Boerema in Boerema & Dorenbo., *Versl. Meded. plziektenk. Dienst Wageningen* 156 (Jaarb. 1979) (1980) 27.

Pyrenochaeta gardeniae S. Chandra & Tandon, *Mycopath. Mycol. appl.* 29 (1966) 274–275.

Selected literature. Chandra & Tandon (1966), Boerema & Dorenbosch (1980).

4) Trotter probably intended to replace the later homonym *Pyrenochaeta leptospora* Speg. with *Pyrenochaeta spegazziniana*, but listed the latter as a substitute for *P. leptospora* Sacc. & Briard.

Description in vitro

OA: growth-rate 50–68 mm, regular, with finely floccose, grey olivaceous to olivaceous grey aerial mycelium; colony grey olivaceous to greenish olivaceous towards margin, or colourless with grey olivaceous to dull green sectors; reverse olivaceous grey to leaden grey/leaden black, or with grey olivaceous to olivaceous grey sectors.

MA: growth-rate 55–73 mm, regular to somewhat irregular, with abundant, floccose to woolly, grey olivaceous aerial mycelium; colony grey olivaceous to olivaceous grey towards margin, or colourless with greenish olivaceous/olivaceous grey to dull green sectors; reverse grey olivaceous to leaden grey/leaden black, or with greenish olivaceous to olivaceous grey sectors.

CA: growth-rate 59–78 mm, regular, with floccose to woolly, (pale) olivaceous grey aerial mycelium; colony greenish olivaceous to olivaceous, or colourless with greenish olivaceous to olivaceous sectors; reverse olivaceous to leaden grey at centre, or buff to rosy buff with olivaceous sectors.

Pycnidia setose, 50–180 µm diam., globose to irregular, solitary or confluent, with usually 1 slightly papillate or non-papillate ostiole, olivaceous to olivaceous black; setae relatively short, up to 100 µm, concentrated around ostiole; walls made up of 3–8 layers of cells, or filling nearly the entire cavity, outer layers pigmented; with white to flesh-coloured conidial exudate; scattered, both on and in the agar as well as in aerial mycelium. Conidiogenous cells 4–8 × 4–7 µm, bottle-shaped. Conidia aseptate, (3.5–)5–8.5(–10.5) × (1.5–)2–3.5(–4.5) µm, av. 5.6–7.0 × 2.7–3.0 µm, Q = 1.4–3.4, av. Q = 2.1–2.4, ellipsoidal to ovoid, with or without several small guttules.

Chlamydospores present, 6–15 µm, globose to subglobose, solitary or aggregated, ochraceous to olivaceous, with greenish guttules, usually intercalary.

NaOH spot test: a weak reddish/brownish discoloring may occur, but this is not specific. Crystals absent.

Ecology and distribution. The original Indian isolate of this species was made from leaf spots of the cape jasmine, *Gardenia jasminoides*. In India it seems to be a common soil-borne fungus, which may act as an opportunistic pathogen of woody as well as herbaceous plants (once isolated from *Arachis hypogaea*). The fungus is also reported from Curaçao. It has been confused with *Phoma leveillei* Boerema & G.J. Bollen var. *leveillei* (no. 5a), but can be easily distinguished from the latter by its highly variable, relatively large conidia and the production of chains of chlamydospores.

Representative cultures. CBS 626.68 (IMI 108771) ex *Gardenia jasminoides* (Rubiaceae), India; CBS 302.79 (PD 79/1156) ex air, Netherlands Antilles (Curaçao).

9. **Phoma indica** (T.S. Viswan.) De Gruyter & Boerema, *comb. nov.* — Fig. 11

Pyrenochaeta indica T.S. Viswan., Curr. Sci. 26 (1957) 118 [basionym; holotype on leaf spot of *Saccharum officinarum*, Poona, India, AMH-11; dried culture of type in Herb. IMI-062569(b)].

Description in vitro

OA: growth-rate 12 mm after 7 days (14 days: 28–30 mm), regular, with sparse, felty, pale olivaceous grey aerial mycelium; colony olivaceous, reverse similar.

MA: growth-rate 16–17 mm after 7 days (14 days: 40–41 mm), regular, with felty, white to smoke grey/pale olivaceous grey aerial mycelium; colony greyish due to aerial mycelium, grey olivaceous to olivaceous near margin; reverse similar.

CA: growth-rate 14–15 mm after 7 days (14 days: 23–25 mm), regular, with felty, white to smoke grey/pale olivaceous grey aerial mycelium; colony greyish due to aerial mycelium, hazel near margin; reverse hazel, with mouse grey at centre.

Pycnidia (on type herbarium material) setose, 55–240 µm diam., globose to subglobose, solitary or confluent, with 1 or 2 papillate ostioles, citrine/honey, later sienna to olivaceous/olivaceous black; setae relatively short, up to 100 µm, mainly concentrated around the ostiole; walls made up of 3–12 layers of cells, outer layer(s) pigmented; with (pale) luteous coloured conidial exudate; scattered, both on and in the agar. Conidiogenous cells 4–6 × 3–6 µm, globose to bottle shaped. Conidia aseptate, 4–5.5 × 1.5–2.5 µm, av. 4.8 × 2.0 µm, Q = 2.0–3.4, av. Q = 2.4, ellipsoidal, usually with 2 guttules.

Chlamydospores present, 5–11 µm diam., globose to subglobose, solitary or in short chains, occasionally clustered, olivaceous with greenish guttules, mostly terminal.

NaOH spot test: positive on OA and MA, herbage green (not E reaction).

Crystals absent.

Ecology and distribution. Found on the whitish centre of fusiform dirty brownish leaf spots of sugar-cane, *Saccharum officinarum* in India. There are no data on pathogenicity. On the spots a species of *Melanospora* was also found. *Phoma indica* produces significantly smaller conidia than *P. setariae* (no. 11), recorded in Brazil from leaf spots of sugar-cane.

Representative culture. IMI 062569 ex *Saccharum officinarum* (Gramineae), India.

10. *Phoma terricola* Boerema — Figs. 12, 16

Phoma terricola Boerema, Versl. Meded. plziektenk. Dienst Wageningen 163 (Jaarb. 1984) (1985) 38–39; not *Phoma terricola* 'Agnihotr.'. Soil Sci. 91 (1961) 135 [a nomen nudum erroneously adopted in Mathur, Coelom. India (1979) 185].

Pyrenochaeta decipiens Marchal, Champ. copr. 6 (1891) 8; not *Phoma decipiens* Mont., Fl. chil. cell. 7 (1852) 488.

Selected literature. Boerema (1985).

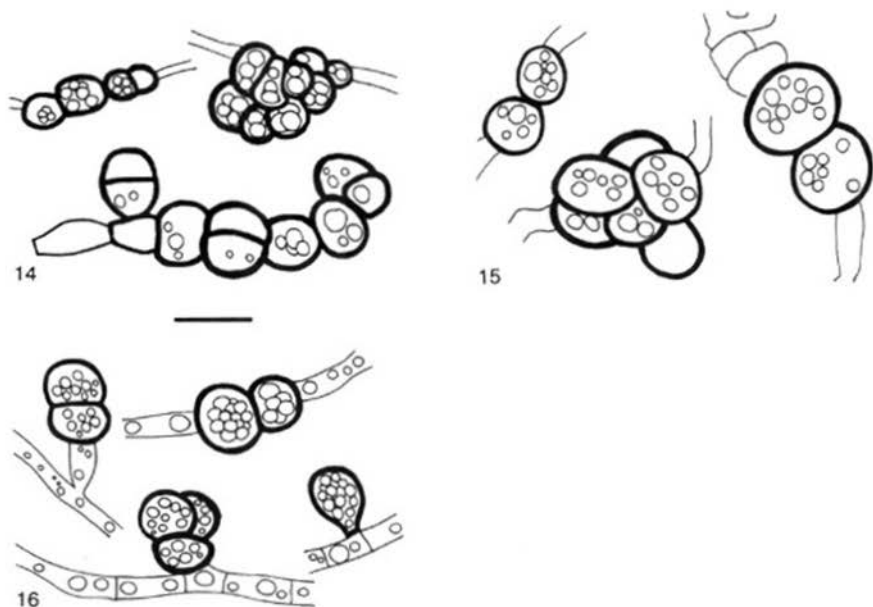
Description in vitro

OA: growth-rate 25–29 mm (14 days: 46–51 mm), regular to somewhat irregular, with finely floccose, pale olivaceous grey to pale greenish grey aerial mycelium; colony pale olivaceous grey/grey olivaceous to dark herbage green/dull green; reverse dull green/olivaceous to leaden grey/leaden black.

MA: growth-rate 20–22 mm (14 days: 39–40 mm), regular to somewhat irregular, with finely felty, pale olivaceous grey aerial mycelium; colony olivaceous grey/grey olivaceous to dull green; reverse similar, olivaceous to leaden grey/leaden black.

CA: growth-rate 22–24 mm (14 days: 42–46 mm), regular to somewhat irregular, with floccose, white to pale olivaceous grey aerial mycelium; colony grey olivaceous to olivaceous grey; similar, with fawn to hazel and leaden grey patches.

Pycnidia setose, 130–250 µm diam., globose, usually solitary, with usually 1, non-papillate or slightly papillate ostiole, honey to citrine, later olivaceous black; setae relatively short, up to 100 µm, mainly concentrated around the ostiole; walls made up of 2–5 layers of cells, outer layer(s) pigmented; with white coloured conidial exudate; scattered or in concentric rings, both on and in the agar. Conidiogenous cells 3–6 × 3–7 µm, globose to bottle-shaped. Conidia aseptate, 3–5(–5.5) × 1.5–2 µm, av. 3.6–4.4 × 1.5–1.6 µm, Q = 1.8–3.3, av. Q = 2.5–2.8, ellipsoidal to subcylindrical, with usually 2 guttules.



Figs 14–16. Chlamydospores. 14. *Phoma terrestris*; 15. *Phoma gardeniae*; 16. *Phoma terricola*. — Bar = 10 μ m.

Chlamydospores present, globose to subglobose, solitary or aggregated, ochraceous to olivaceous, with greenish guttules, intercalate or terminal, 6–10 μ m diam.

NaOH spot test: negative.

Crystals absent.

Ecology and distribution. A soil fungus recorded in western Europe (Belgium, the Netherlands). Probably widespread occurring in agricultural fields. For example, it has been found in the rhizosphere of wheat (*Triticum aestivum*) and beet (*Beta vulgaris*), and isolated from cysts of the golden nematode of potatoes (*Globodera pallida*). The pycnidia of this fungus are very similar to those of *Phoma leveillei* Boerema & G.J. Bollen var. *leveillei* (no. 5a), but can be easily distinguished by an abundant production of chlamydospores. It also morphologically resembles *Phoma terrestris* H.N. Hansen (no. 4), but does not produce red pigment.

Representative culture. CBS 343.85 (PD 85/1044) ex cyst of *Globodera pallida*, the Netherlands.

APPENDIX

Insufficiently known species

Apart from the seventeen *Pyrenochaeta* synonyms of the *Phoma* species treated in this paper in section *Paraphoma*, Schneider (1979) listed 23 other *Pyrenochaeta* spp. which, on account of their original descriptions or type material, should also belong to *Phoma*.

Most of these taxa are only known from a single collection. However, Schneider (1979) noted that three differently named African/American collections from leaf spots on *Pennisetum*, *Saccharum* and *Setaria* spp. refer to one and the same species. An old isolate of this pathogen appears to be available (CBS 333.39 from *Saccharum officinarum* in Brazil), but has remained sterile. Without doubt a *Phoma* species of sect. *Paraphoma* is involved. Below it is transferred from *Pyrenochaeta* to *Phoma* with a resumé of its characteristics on the hosts.

11. *Phoma setariae* (H. C. Greene) De Gruyter & Boerema, *comb. nov.*

Pyrenochaeta setariae H. C. Greene, Trans. Wis. Acad. Sci. Arts Lett. 53 (1964) 211[–212] [basionym; holotype on leaf of *Setaria lutescens*, coll. T. F. Hubb, near Pine Bluff, Dane County, Wisconsin, USA, Sept. 1964, WIS].

Pyrenochaeta sacchari Bitanc., Arquivos Inst. Biol., São Paulo 9 (1938) 301; not *Phoma sacchari* (Cooke) Sacc., Sylloge Fung. 3 (1884) 166; not *Phoma sacchari* Gutner apud Bond.-Mont., Gutner & Novos., Acta Inst. Bot. Acad. Sci. URSS, Ser. II, Fasc. 3 (1936) 789 [= *Phoma gutneri* Pons, Fitopat. Venezolana 3 (2) (1990) 40[–42].

Pyrenochaeta penniseti Kranz, Sydowia 22 (1968) 360–361.

Selected literature. Schneider (1979).

Description in vivo

Pycnidia (initially epiphyllous, later also amphigenous, in oval-fusiform, often confluent, first pale buff, later brownish or vinaceous spots with narrow darker border) in majority setose, 100–150 µm diam., subglobose with usually 1 papillate ostiole, brownish, lighter at the base and darker toward the ostiole; setae short, continuous, 15–75 µm, uniform, mostly around the ostiole; wall made up of 2–5 layers of cells, outer layer(s) pigmented; conidial exudate whitish. Conidia aseptate, 6–10(–12) × 2.5–4 µm, broadly ellipsoidal, subcylindrical to subfusoid or irregular, with usually 2 distinct guttules.

Ecology and distribution. Possibly a widely distributed weak pathogen of Gramineae, which only becomes noticeable in conditions favourable for spread. The records refer to *Pennisetum typhoides* in Africa (Guinea, Nigeria), *Saccharum officinarum* in Brazil and *Setaria lutescens* in North America: Leaf Spot. As the infection progresses the first infected leaves may die back completely.

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With the treatment of this section *Paraphoma* we made grateful use of the type studies of *Pyrenochaeta* spp. by Dr. Roswitha Schneider, former mycologist at the 'Institut für Mikrobiologie', BBA, Berlin-Dahlem. Living and/or dried cultures were obtained from CBS, Baarn/Utrecht, NL and IMI, Egham, UK. Dr. R. T. A. Cook kindly improved the English of this paper. Thanks are also due to Dr. W. Gams for the Latin description.

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THE GENUS *AMANITA* IN THE PAKARAIMA MOUNTAINS OF GUYANA

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Six species of *Amanita* (Amanitaceae, Basidiomycota, Fungi), collected in the Pakaraima Mountains of Guyana, are described; four as new and two as new records for Guyana: *A. aurantiobrunnea* spec. nov., *A. calochroa* spec. nov., *A. cyanopus* spec. nov., *A. perphaea* spec. nov., *A. lanivolva* Bas. and *A. xerocybe* Bas. All six species have been collected in forests dominated by *Dicymbe* (Caesalpinaceae) and are assumed to be ectomycorrhizal. *Amanita perphaea* is commonly eaten by Patamona Indians and is called 'Pulutukwe'.

Ectomycorrhizal fungi, and specifically the genus *Amanita*, were long thought to be well developed in the northern Temperate Zone, but poorly represented in the tropics (Gilbert, 1941b). This viewpoint began to shift with the advent of mycofloristic studies in the paleotropics, where numerous studies have demonstrated an increasingly wide diversity of ectomycorrhizal fungi associated primarily with caesalpinoid legumes in Africa and with Dipterocarpaceae and Fagaceae in Asia (e.g. Heinemann, 1954; Corner & Bas, 1962; Corner, 1972; Watling & Lee, 1995; Buyck et al., 1996). In the lowland forests of the neotropics, however, documentation of ectomycorrhizal associations is limited to the pioneering work of Singer and colleagues in the Central Brazilian Azon and Central America, and Moyersoen in southern Venezuela (Singer & Araujo, 1979; Singer et al., 1983, 1991; Moyersoen, 1993).

Knowledge of *Amanita* in South America is limited. *Amanita* species have been recorded with the vegetation types of the pampas (but a small number of species recorded from the pampas are probably non-ectomycorrhizal), open dry forests and, to a limited extent, lowland rain forests (Bas, 1969). Dennis (1970) described only three species for Venezuela, among which there was only one species (*A. antilliana* Dennis from Trinidad) from tropical lowland forests. Subsequent description of new species from Amazonian Brazil by Bas (1978) and Andean Colombia by Tulloss et al. (1992), among others, did much to reverse this trend of thought, but the number of known *Amanita* species from the continent still remained relatively low.

Recent investigations of the macromycota of Guyana, in northeastern South America, have revealed an extensive assemblage of putatively ectomycorrhizal fungi, including *Amanita*, associated with trees of the legume genus *Dicymbe* Spruce ex Benth. in Benth. Hookf. (Caesalpinaceae, tribe Amherstieae) (Henkel, 1999; Henkel et al., 2000; Miller & Henkel, 2000). These include, along with *Amanita*, members of the basidiomycete families Boletaceae, Russulaceae, Cortinariaceae, Cantharellaceae, Clavariaceae, and Hygrophoraceae, as well as the hypogeous ascomycete genus *Elaphomyces*. These fungi are tightly

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spatially associated with forest stands dominated by ectomycorrhizal *Dicymbe* species, in particular *D. corymbosa* Spruce ex Benth. and *D. altsonii* Sandwith, which occur in a landscape-scale mosaic interposed with largely anectotrophic mixed forest in the Pakaraima Mountains of west-central Guyana (Fanshawe 1952, 1955; Steege et al., 1993; Henkel, 2000). Discovery of these *Dicymbe*-dominated ectotrophic forests in Guyana has greatly extended the known range of ectomycorrhizal associations in South American rain forests.

Terry Henkel, during the period 1993 to 1998, made 5 collecting expeditions to the Upper Ireng River basin along the western border of Guyana with Brazil. He kindly allowed his Guyanese *Amanita* collections be examined and analysed by the first author, under guidance of the third author. As a preliminary result, four new species and two species previously described from the Brazilian Amazon are recorded and described in the present paper.

The genus *Amanita* consists largely of terrestrial, forest-inhabiting fungi with a rich biogeographic history (Bas, 1969). There are over 600 species known world-wide, with the genus divided into two subgenera based on the amyloidity of the spores and the structure of the margin of the pileus, viz. the subgenera *Lepidella* and *Amanita*, and the macroscopic and microscopic features of the volva being a primary character for dividing the subgenus *Lepidella* into four sections and the subgenus *Amanita* into two sections (Bas, 1969).

COLLECTING SITE DESCRIPTION

The research site was located along the Upper Ireng River which forms the border between Guyana¹ and Brazil in the South Central Pakaraima Mountains (general area: 5° 5' N; 59° 58' W). This region of western Guyana constitutes the eastern extension of the Guayana¹ Highlands, a distinct phytogeographic province characterized by sedimentary strata overlying the igneous Guiana¹ Shield basement, oligotrophic soils, and a highly autochthonous, endemicized flora (Gansser, 1954; Maguire, 1970; Gibbs & Barron, 1993). Elevations range from 700 m at riverside to 1800 m along the highest ridgelines. The resulting terrain is of high relief and characterized by deeply cut creek and river valleys amidst highly eroded plateaus and pinnacled ridgelines.

Annual rainfall for the remote area can only be inferred from records spanning the years 1935–1947 at a Potaro River site located approximately 100 km to the northeast, at the eastern edge of the Pakaraima escarpment (Fanshawe, 1952). The annual mean for the 12 year period was 3866 mm with a pronounced peak in May and June, a lesser peak in December and January, with relatively drier periods between. Since 1991, the timing of annual wet and drier periods at the Ireng site have, in general, coincided with those indicated by the Potaro data (Henkel, pers. obs.). At the Ireng site, during the onset of the rainy season in May–June 1998, a total precipitation of 585 mm was recorded for a 27 day period, and during May–June 1999 490 mm for a 30 day period, which were in accordance with the mean values of 500–600 mm/month for May and June at the Potaro site. For macromycetes, annual peak production of sporocarps appears to occur during the May–June period (Henkel, unpubl.). Regional temperature records were unavailable, but during the equivalent periods of 1998 and 1999 temperatures were remarkably constant at the Ireng site (daily max.: 25–29°C; min.: 19–21°C). In general, these data correspond to a classification of Submeso-

1) 'Guyana': the country formerly called British Guiana; 'Guayana': the concerning phytogeographical province; 'Guiana': the geological region underlain by the Guiana Shield.

thermic Ombrophilous Climate as indicated by Berry et al. (1995), defined as annual precipitation exceeding 2000 mm with minimal or no dry season (i.e. where monthly precipitation falls below 100 mm), with average annual air temperatures ranging between 18°C and 24°C.

Vegetation of the region corresponds to the Dry Evergreen Forest Formation of the Pakaraima Montane Region (Fanshawe, 1952). Fanshawe considered this formation to predominate throughout the entire sandstone belt composing the Pakaraima Mountains, being subdivided into various associations and faciations with attendant dominant tree species according to degree of soil drainage, the series running from sclerophyllous savanna on the most poorly drained sites to well-drained, high canopy mixed forest, and 'clump wallaba' forest dominated by species of *Dicymbe*. *Dicymbe*, in particular *D. corymbosa*, forms extensive mono-specifically dominant stands of five ha and larger on the lower toe slopes of the mountains (Myers, 1936; Fanshawe, 1955; Richards, 1996). These *Dicymbe* stands have, by tropical standards, extremely low woody plant diversity, with roughly 40 species >10 cm dbh per ha, with 80–95% of the basal area of the stand composed of *D. corymbosa* (Henkel, 2000). Mixed forests of the area, generally lacking in *Dicymbe*, have 70–90 woody species per ha, well represented by species of *Eschweilera* Mart. ex DC. (Lecythidaceae), *Ocotea* Aubl. (Lauraceae), *Inga* Mill. (Mimosaceae), and *Eperua* Aubl. (Caesalpinaceae) (Henkel, unpubl.). Low-lying forests with poor drainage are generally dominated by *Micrandra glabra* (R.E. Schult.) R.E. Schult. (Euphorbiaceae) and *Mora excelsa* Benth. (Caesalpinaceae).

Soils are poor due to the sandstone nature of the parent materials. Soil impoverishment is further evidenced by the universal presence of tannin-rich 'blackwater' streams throughout the region (Janzen, 1974).

MATERIALS AND METHODS

Fungi were collected from an area of approximately 10 km² surrounding a series of previously established base camps (Henkel, 1999). Basidiomata were examined in the field for fresh characteristics. Colour characteristics were coded according to Kornerup & Wanscher (1981) and described subjectively. Spore deposits, when obtained, were examined for fresh colour characteristics. Basidiomata were dried slowly over charcoal and subsequently placed in containers with silica gel to prevent spoilage in the excessively humid conditions.

All anatomical studies were performed on dried specimens. Microscopic examination of tissues were done in 5% and 10% NH₄OH or in 5% KOH. Stains used were Congo red and the spores were observed in Melzer's reagent and measured in 10% NH₄OH and 5% KOH. Bas found that in cases where amyloidity was dubious, soaking overnight in Melzer's reagent and observing in fresh reagent the next morning made amyloidity show more clearly. For spores at least 10 individuals per sample were measured and for basidia at least 6 individuals. Anatomical details were determined with an Olympus CHA microscope containing bright field and phase contrast optics and line drawings were made with the help of a drawing tube.

Because of rather primitive circumstances in the field, the dried specimens were often not in optimal condition. It turned out that even in good looking specimens it sometimes was very difficult or impossible to get the tissues of the trama of the lamellae, the stipe, and the pileus to reinflate.

Herbarium designations: BRG – National Herbarium of Guyana, University of Guyana, Georgetown, Guyana; DUKE – Department of Botany, Duke University, Durham, North Carolina, USA; L – Nationaal Herbarium Nederland, Universiteit Leiden Branch, Leiden

Other abbreviations used were: 'Q' = length/breadth ratio; 'Spores [20/2]' = twenty spores from two specimens measured; 'R' = radius of pileus.

TAXONOMIC PART

Hitherto, not all sections of *Amanita* are represented in the set of collections from the Pakaraima Mountains that has been analysed. The species recognised are treated in taxonomic order. A survey of the subgenera and sections they belong to is given here:

Amanita subgenus *Lepidella*

Section *Phalloideae*: *A. aurantiobrunnea*

Section *Validae*: *A. cyanopus*, *A. perphaea*

Section *Amidella*: not represented

Section *Lepidella*: not represented

Amanita subgenus *Amanita*

Section *Vaginatae*: several specimens collected, but none yet analysed.

Section *Amanita*: *A. calochroa*, *A. lanivolva*¹, *A. xerocybe*

Subgenus **LEPIDELLA**

Section **Phalloideae**

Amanita aurantiobrunnea C. Simmons, T. Henkel & Bas, *spec. nov.* — Fig. 1.

Plate 2

Pileus 30–70 mm latus, convexus vel applanatus, postea plano-concavus, interdum obtuse umbonatus, margine laevis, aurantio-brunneus vel aurantiacus, viscidus, margine fragmentis volvae minutissimis, albidis vel concoloribus saepe instructus. Lamellae liberae, (sub)confertae, albae; lamellulae attenuatae. Stipes 64–88 × 7–16 mm, basi bulbosus, 22–35 mm latus, annulatus. Volva membranaceo-saccata, alba vel pallide brunneo-salmonea. Annulus apicalis, floccoso-membranaceus, albus, facile diffractus.

Sporae 6.8–9.4 × 5.5–7.5 µm, subgloboosae vel late ellipsoideae vel ellipsoideae, amyloideae. Basidia 4-sporigera. Fragmenta volvae supra marginem pilei cellulis turgidis, (sub)globoosis vel pyriformibus, 25–95 × 15–50 µm, terminalibus vel catenulatis hyphisque albis vel brunneis composita. Limbus volvae externus hyphis praecipue constructus. Fibulae absentes.

Typus: *T. Henkel 6431*. Guyana, Pakaraima Mountains, Upper Ireng River, 22.V.1998 (BRG holotypus, L isotypus).

Fruit-body terrestrial, solitary, medium-sized and fleshy. Pileus 30–70 mm in diam., convex to applanate, plano-concave with age, sometimes with low broad umbo, viscid when wet, tacky when dry, rich orangish tan (close to K&W 5B8) to light orange at margin (K&W 5A5), with white context and often with inconspicuous, tiny, whitish to concolorous volval fragments on outermost margin. Lamellae free, thin, close to rather close, white, with entire

1) The taxonomic position of *A. lanivolva* is not quite clear. It is placed here in section *Amanita* because it has non-amyloid spores combined with a sulcate-striate pileus margin and a distinctly bulbous base, but it has a saccate volva, which is unique in section *Amanita*.

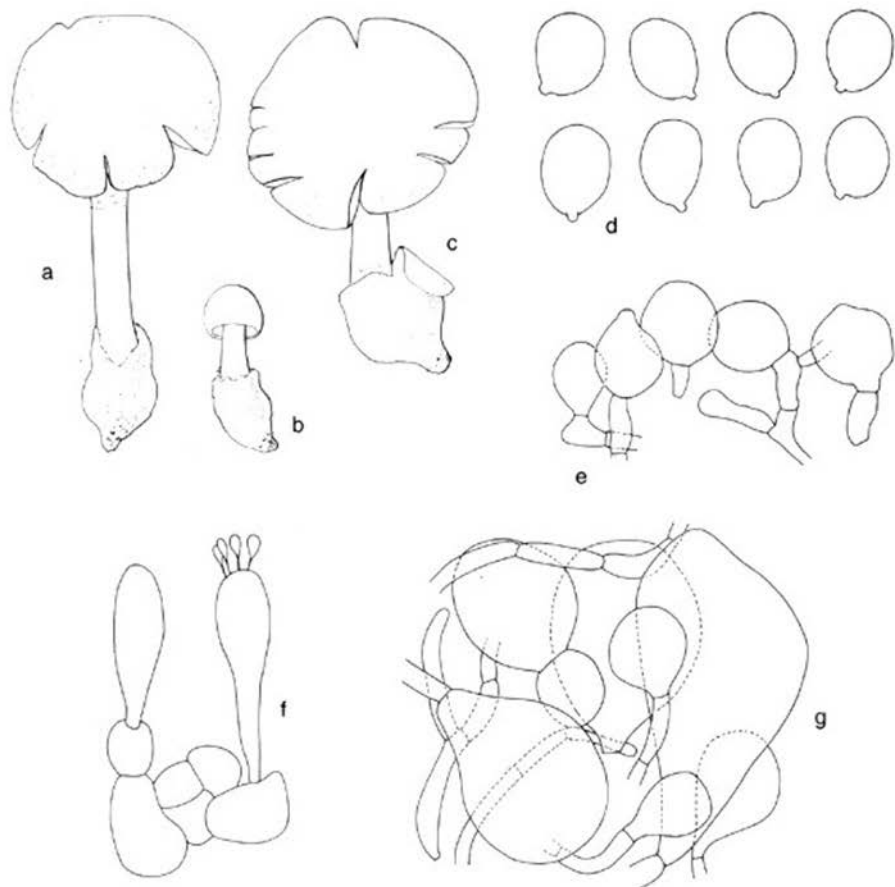


Fig. 1. *Amanita aurantiobrunnea*. a–c. Basidiocarps, $\times 0.5$; d. spores, $\times 1500$; e. elements of marginal tissue of lamella, $\times 500$; f. basidia and subhymenium, $\times 1000$; g. tissue of minute fragment of volva on margin of pileus, $\times 500$ (a, d–f: holotype; b: TH 6655; c & g: TH 6898).

edge. Lamellulae attenuate. Stipe 64–88 \times 7–16 mm, slightly attenuated upwards, white with white floccose squamules when young but glabrescent with age; bulb 22–35 mm in diam., enclosed in a firm, adnate, white to pale brownish salmon, membranous, saccate to limbate volva attached to upper part of bulb with limb free and spreading or more or less appressed against stipe. Annulus apical, white, floccose-membranous, rarely skirt-like, often reduced to small white scales on apex of stipe, rarely forming white appendages at margin of pileus. Spore print not available.

Spores [65/5] (6.2–)6.8–9.4 \times (5.0–)5.5–7.5(–7.9) μm , $Q = (1.0\text{--})1.1\text{--}1.35\text{--}(1.45)$, aver. $Q = (1.1\text{--})1.15\text{--}1.35$, subglobose to broadly ellipsoid to ellipsoid, with thin to very slightly thickened walls, amyloid, with relatively broad and short, rounded hilar appendix. Basidia 27–45 \times 7–11 μm , clavate, 4-spored, with up to 5 μm long sterigmata, clampless. Sub-

hymenium pseudoparenchymatic, composed of subglobose to (larger) angular, up to 17 μm wide elements. Marginal tissue an uneven strip of mainly globose to broadly ellipsoid, thin-walled to slightly but distinctly thick-walled, colourless elements $11\text{--}28 \times 13\text{--}25 \mu\text{m}$, sometimes in short chains and occasionally with apical papilla, attached to frequently branching, $3\text{--}6.5 \mu\text{m}$ wide hyphae. Hymenophoral trama bilateral. Pileipellis an ixocutis of densely interwoven, $3.8\text{--}5.2\text{--}(6.6) \mu\text{m}$ wide, agglutinated, thin-walled, slightly inflated hyphae without clamps; suprapellis colourless in NH_4OH , subpellis pale yellow; superficial hyphae sometimes carrying ovoid, ellipsoid and elongate elements of up to $98 \times 25 \mu\text{m}$. Tiny volval remnants on margin of pileus consisting of colourless to brownish hyphae carrying terminal, single or more rarely catenulate, globose to pyriform, thin- to slightly thick-walled elements measuring $27\text{--}97 \times 17\text{--}49 \mu\text{m}$. Volval limb: outer surface a dense tissue of mainly longitudinal to interwoven, clampless, $1.7\text{--}5.5 \mu\text{m}$ wide hyphae and a few scattered, isolated, subglobose to broadly ovoid and ellipsoid, inflated, thin-walled, colourless elements $25\text{--}60 \times 21\text{--}34 \mu\text{m}$; the inner tissue composed of irregularly disposed, branching $2.2\text{--}6.2 \mu\text{m}$ wide hyphae and many subglobose, ellipsoid, ovoid, and elongate, inflated, terminal cells $28\text{--}60 \times 25\text{--}35 \mu\text{m}$; inner surface, particularly at outermost limb, with globose to pyriform inflated cells, $28\text{--}76 \times 25\text{--}49 \mu\text{m}$, embedded in a tissue of $2.5\text{--}5.5 \mu\text{m}$ wide hyphae, pale yellow in NH_4OH . Trama of stipe composed of longitudinal hyphae carrying acrophysalides up to $308 \times 56 \mu\text{m}$. No clamp-connections observed.

Habitat & distribution — Slope forest, dominated by *Dicymbe corymbosa* (Paluwayek), on grey sands. Thus far only known from western Guyana.

Collections examined. GUYANA: Pakaraima Mountains, Upper Ireng River: 1 km west of Kurutuik Falls, on adjacent ridges, 6 April 1998, *T. Henkel et al.* (TH 6852; BRG); Forest adjacent to Sukabi River, 1–2 km upstream from confluence with Ireng River, 22 May 1998, *T. Henkel et al.* (TH 6431; holotype BRG, isotype L); Sakaliu River, 1–2 km upstream from confluence of Ireng River, 25 May 1998, *T. Henkel* (TH 6445; BRG); north to south running ridge about 1 km west of confluence of Ireng and Sukabi Rivers, 27 May 1998, *T. Henkel et al.* (TH 6655; BRG, L); east bank of Iring River, 1 km downstream from Kurutuik Falls, 6 June 1998, *T. Henkel* (TH 6898; BRG).

Amanita aurantiobrunnea clearly belongs to section *Phalloideae* because of the amyloid spores, the saccate to limbate volva attached to the upper part of the bulb and the deeply coloured pileipellis. Within this section very few species have colours that are reminiscent of those in the present one.

Amanita aureomonile Tulloss & Franco-Molano (Tulloss et al., 1992) from Colombia has a bright yellow pileus with a darker disc, a pale yellow stipe and a bright yellow annulus.

Amanita subjunquillea S. Imai from Japan, as illustrated by Imai in Gilbert (1941a), seems close to *A. aurantiobrunnea*, but has a more ochraceous yellow pileus, a more coherent annulus, and more globose spores ($7.0\text{--}8.0 \times 7.0\text{--}7.5 \mu\text{m}$ according to Gilbert (1941a) and $(6.0\text{--})6.5\text{--}8.5\text{--}(9.0) \times 6.0\text{--}8.5\text{--}(9.0) \mu\text{m}$, $Q = 1.0\text{--}1.2$, according to Yang, 1997).

It should be mentioned here that the original *A. gayana* (Mont.) Sacc., described by Montagne (1853) from Chile, also has a glabrous red to orange pileus with a non-sulcate margin (specifically mentioned), a white stipe and annulus, and a white saccate volva and thus could represent a species of section *Phalloideae* too, although it seems to have no bulbous base. We prefer to consider the Chilean taxon a dubious one as long as no material has been collected again in Chile and the Melzer's reaction of the spores remains unknown.

Montagne's name has been misapplied by Singer (1969) and Garrido & Bresinsky (1985) to the species now known as *A. aurantiovelata* Schalkw. & G.M. Jansen (1982) which has

non-amyloid spores, a sulcate pileus margin, and orange-yellow volval warts on the pileus and the base of the stipe.

Amanita aurantiobrunnea has a peculiar character, that, as far as we know, has not yet been described in other species of section *Phalloideae*, viz. a thin, white to coloured, friable inner layer at least on the outer limb of the volva that is responsible for the frequent presence of rather inconspicuous, small, thin volval patches on the outermost margin of the pileus.

Because of slight irregularities on the surface of the spores seen in the light microscope, the spores have been scanned with a scanning electron microscope. When enlarged 10.000× they appeared, however, to be perfectly smooth.

Section *Validae*

Amanita cyanopus C. Simmons, Henkel & Bas, *spec. nov.* — Fig. 2, Plate 4

Pileus 65 mm latus, plano-concavus, margine laevis, sordide pallide vel obscure griseo-caeruleus, fragmentis volvae applanatis, coactis, concoloris decoratus praesertim prope margine, minute fibrillosus. Lamellae liberae, confertae, sordide cremeo-alutaceae, margine pallide caeruleae. Stipes 95 × 12 mm, basi bulbosus 18 mm latus, leviter radicans, pallide caeruleus, squamulis flocculosis minutis griseis obsitus, bulbo fragmentis volvae verruciformibus vivide caeruleis obtectis, annulatus. Odor ingratus.

Spores 7.4–8.7 × 5.6–7.4 μm, late ellipsoideae vel ellipsoideo-elongatae, amyloideae. Basidia 4-sporigera. Fragmenta volvae cellulis turgidis, 9–33 × 7–19.5 μm, terminalibus vel breve catenulatis, olivaceo-brunneis hyphisque rarioris composita. Fibulae absentes.

Holotypus: *T. Henkel 7083*, Guyana, Pakaraima Mts, Upper Ireng River, 30.V.1999 (BRG).

Fruit-body medium-sized, rather slender, solitary. Pileus 65 mm in diam., plano-concave, with smooth margin, rather dull greyish to bluish turquoise (= greyish lavender blue to dark, slightly greyed blue, K&W 23D3–24D8), with concolorous, flat, felted volval patches particularly near margin, minutely, rather innately fibrillose. Lamellae free, crowded, thickish, occasionally forking, dull dark greyish tan (according to field notes; in colour slide looking rather pale cream-buff), with finely uneven, light blue edge. Stipe 95 × 12 mm, equal throughout, but with large, slenderly napiform, somewhat rooting bulb, 55 × 18 mm, annulate, light bluish under a coating of minute, grey, floccose scales, but bulb covered with small bright blue, conical volval warts and ridges on a paler blue to whitish background. Annulus present, apical (broken off close to apex of stipe in single specimen available). Smell unpleasant (according to collector distinctly of 'chlorine'). Spore print not available.

Spores [25/1] 7.4–8.7(–9.0) × (5.0–)5.6–7.4 μm, Q = (1.15–)1.25–1.45(–1.6), aver. Q = 1.36, broadly ellipsoid to oblong, with rather small hilar appendix, thin-walled, smooth, colourless, amyloid. Basidia 29–38 × 9.4–10.2 μm, 4-spored, clampless. Subhymenium pseudoparenchymatic, consisting of globose to ellipsoid, ovoid and broadly clavate cells, about 11–18 × 11–18 μm. Marginal tissue a rather broad, (in NH₄OH) colourless strip of thin-walled globose to ellipsoid, ovoid and broadly clavate cells 14–29(–42) × 12.5–20.5 μm, at least partly in chains. Hymenophoral trama: bilateral (very difficult to study in present specimen). Pileipellis (in scalp): suprapellis an ixocetus of 1.4–4.2 μm, interwoven (at centre as well as near margin) hyphae with unevenly distributed, (in NH₄OH) olivaceous brown, intracellular pigment, distant by gelatinification, over a subpellis of (almost) colourless, more radial, 2–7 μm wide hyphae. Volval remnants made up of globose, ellipsoid, ovoid, clavate, and irregularly shaped inflated elements 9–33 × 7–19.5 μm, with (in NH₄OH) dark, slightly olivaceous brown, unevenly distributed intracellular pigment, terminal or in

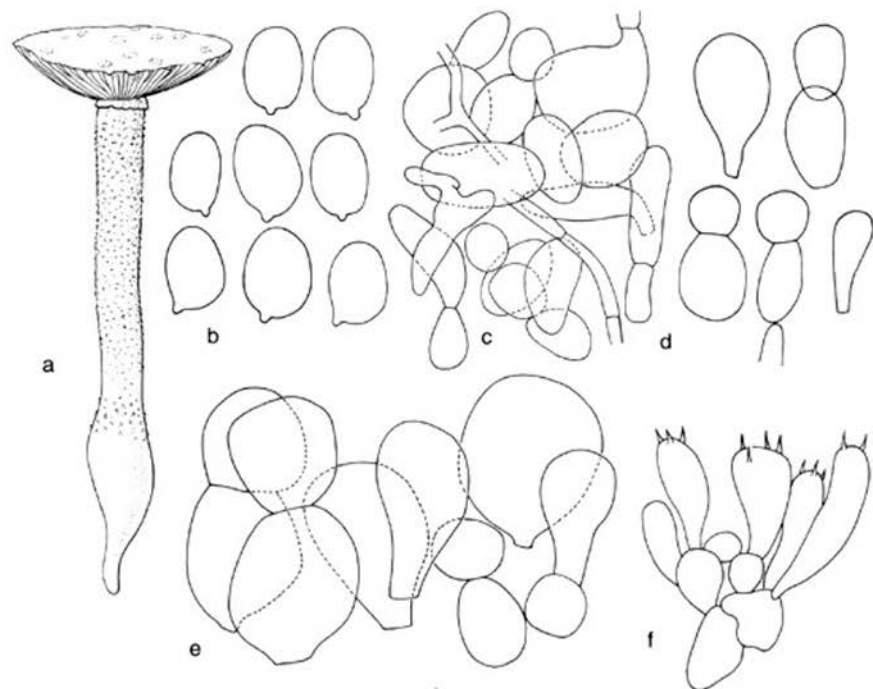


Fig. 2. *Amanita cyanopus*. a. Basidiocarp, $\times 0.5$; b. spores, $\times 1500$; c. slightly crushed scalp of volval patch on pileus, $\times 500$; d. terminal elements from volval patch on pileus, $\times 500$; e. elements of marginal tissue of lamella, $\times 1000$; f. basidia and subhymenium, $\times 1000$ (all from holotype).

terminal chains on relatively scarce 2.8–7.6 μm wide, brown, branching hyphae. Volval remnants on base of stipe not studied. Trama of stipe acrophysalidic, with 3.7–4.3 μm wide, brown, septate hyphae and abundant acrophysalides 83–154 \times 10–23 μm . Clamp-connections absent.

Habitat & distribution — On root mat, but rooting into mineral soil, in forest of predominantly *Dicymbe corymbosa* (Paluwayek). Known only from the type locality in western Guyana.

Collection examined. GUYANA: Potaro-Siparuni, Pakaraima Mountains, Upper Ireng River, west bank of Yuarka River, 1 km upstream from juncture with Suruwabaru Creek, 30 May 1999, *T. Henkel et al.* (TH 7083; holotypus BRG).¹

Although only one dried specimen (fortunately with colour slide) was available, we do not hesitate to describe the taxon concerned as a new species in section *Validae*, because of its unique colours, viz. the greyish blue to dark blue pileus with concolorous volval patches, the pale blue stipe with greyish floccose dots and the bright blue volval warts on the pale blue bulb.

1) Recently a second fruit-body has been collected (*T. Henkel s.n.*), but that has not yet been analysed.

The only species bearing a slight resemblance to *A. cyanopus* is *A. odorata* Beeli from the Congo (1931), also illustrated and described by Gilbert (1941a) with a greyish green pileus with brown volval warts. However, that species has been placed in section *Lepidella* by Bas (1969), because of its appendiculate pileus margin and elongate to subcylindrical spores ($Q = 1.9-3.0$). Moreover it has a smell like bitter almonds.

***Amanita perphaea* C. Simmons, T. Henkel, & Bas, *spec. nov.* — Fig. 3, Plate 5**

Pileus usque ad 150 mm latus, initio convexus, postea plano-convexus vel depressus, obscure griseo-brunneus, nitidus, obscure fibrillosus, verrucis conicis, obscure griseo-brunneis ornatus, margine initio laevus, postea sulcato-striatus. Lamellae liberae, subconfertae, albae vel cremeae. Stipes 60–150 × 10–20 mm, basi bulbosus, obscure griseo-brunneus, fibrillosus, deorsum fragmentis volvae griseo-brunneis inconspicuis instructus. Annulus apicalis (sub)membranaceus, evanescens, pallidus.

Sporae 6.1–7.4 × 4.9–6.9 μm . (sub)globoseae, amyloideae. Basidia 4-sporigera. Fragmenta volvae cellulis turgidis, 25–126 × 20–44 μm , subglobosis, ellipsoideis vel clavatis, brunneis, hyphisque copiosis composita. Fibulae absentes.

Holotypus: *T. Henkel* 6229, Guyana Pakaraima Mts, Upper Ireng River, 15.II.1997, (BRG).

Fruit-bodies terrestrial, solitary, large and fleshy. Pileus up to 150 mm in diam., plano-convex, with depressed centre, grey and shiny, darkening towards centre, with sulcate-striate margin, 4–6 grooves per 10 mm, covered with dark grey conical volval warts over entire surface but more concentrated at central region. Lamellae free, slightly crowded and slightly thickened, ventricose, white to cream, with entire edge; lamellulae truncate to subtruncate. Stipe 60–150 × 10–20 mm, central, tapering upwards, with bulbous base, fibrillose, grey and darkening with age; volval remnants forming grey ridge-like fragments at base on upper part of bulb (not visible in dried specimen). Annulus (sub)membranous, in young specimen still attached to lamellae, in matured specimen apical, descending, skirt-like, pallid, easily torn. Spore print not available.

Spores (present only in oldest specimens) [40/5] (5.6–)6.1–7.4 × 4.9–6.9 μm , $Q = 1.0-1.1-1.3$, aver. $Q = 1.05-1.08$, globose to subglobose, rarely broadly ellipsoid, with slightly thickened wall, amyloid, with prominent hilar appendix. Basidia 27–34 × 5–11 μm , clavate, 4-spored, clampless. Subhymenium composed of subglobose to ovoid elements, 12–13 × 8–9 μm . Marginal tissue a rather broad strip of 1.7–4.1 μm wide hyphae \pm parallel to lamella edge and many, mainly subglobose to ellipsoid and pyriform elements, 15–29 × 14–22 μm . Hymenophoral trama divergent with narrow central stratum of parallel hyphae and diverging zones with subglobose, ovoid and ellipsoid elements single and terminal or in short terminal chains. Pileipellis (at $\pm 0.25R$ from centre) consisting of interwoven to subradial, fairly dark, broad, up to 7.5 μm wide hyphae with intracellular and encrusting pigments and scattered long inflated elements of up to 24 μm wide (also with intracellular and encrusting pigments); incrustations very distinct; refractive hyphae also present, on top of a rather thick almost colourless gelatinised layer. Volval remnants on pileus (in wart taken from centre) consisting of very abundantly branched, 4–7 μm wide, irregularly disposed, colourless to pale brown hyphae carrying subglobose, ellipsoid and elongate elements, 25–126 × 20–44 μm , usually in erect position, except just above pileipellis and there often in periclinal position, with brown vacuolar and encrusting pigments, particularly dark just above pileipellis, but paler towards apex (strongly pigmented layer with periclinal inflated elements with hand lens visible as dark line between base of wart and gelatinised pileipellis). Volval remnants on stipe consisting of mass of irregular hyphae with very few longi-

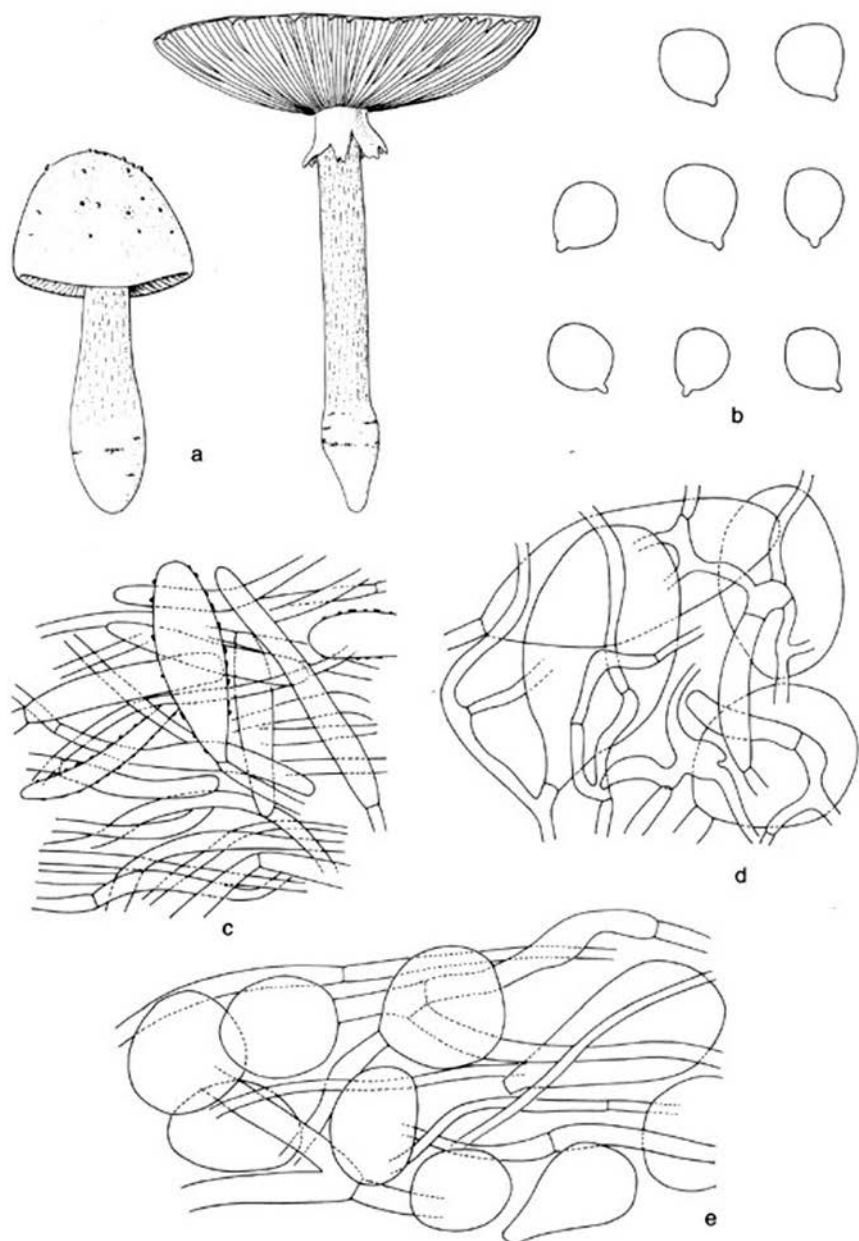


Fig. 3. *Amanita perphaea*. a. Basidiocarps, $\times 0.5$; b. spores, $\times 1500$; c. scalp of pileipellis from above, $\times 500$; d. elements of volval remnants on pileus, $\times 500$; e. marginal tissue of lamella, $\times 1000$ (all from holotype).

tudinal inflated cells of up to $43 \times 14 \mu\text{m}$. Trama of stipe acrophysalidic with elongate cells, $115\text{--}355 \times 24\text{--}41 \mu\text{m}$. No clamp-connections observed.

Habitat & distribution — Riverine and adjacent slope forest dominated by *Dicymbe corymbosa* (Paluwayek) with scattered hardwoods including *Caryocar* sp. in wet bottomlands, on sandy soils with thick root mat and organic accumulations. So far known only from western Guyana.

Collections examined. GUYANA: Potaro-Siparuni, Pakaraima Mountains, Upper Ireng River: Slopes adjacent to east bank of Sukabi River near mouth of Kukuinang Creek, 1.5 km along south bank of Kukuinang Creek, 16 Feb. 1997, T. Henkel et al. (TH 6255; BRG, L); 0.2–1 km downstream from Kurutuk Falls, slopes adjacent to west side of river, 15 Feb. 1997, T. Henkel et al. (TH 6229; BRG holotype, L isotype).

At first it was thought that the collections described above represented *Amanita phaea* Bas nom. prov. (1978) from Brazilian Amazonia, described provisionally because the material available does not have spores. But the pileipellis of *A. phaea* and *A. perphaea* differs considerably. In *A. phaea* the superficial rather dark hyphae only have intracellular pigment, whereas in *A. perphaea* there is an additional encrusting brown pigment. Moreover there are elongate up to $24 \mu\text{m}$ wide inflated elements, probably all terminal, in the pileipellis of *A. perphaea*, which are lacking in *A. phaea*.

Macroscopically the two taxa differ as well. In *A. perphaea* the volval warts and patches on the pileus are as dark as the pileipellis and have a pallid apex or centre, whereas in *A. phaea* they are uniformly pallid. The aspect of the pileipellis differs also, viz. in *A. perphaea* dark and fibrillose with fibers strongly appressed to a paler background and in *A. phaea* more uniformly greyish brown.

In section *Validae*, where *A. perphaea* clearly belongs, several dark grey-brown species with small spores have been described from different parts of the world.

Amanita fritillaria f. *malayensis* Corner & Bas (1962) resembles *A. perphaea* rather strongly, but has more and smaller volval warts on the pileus, a grey annulus at about the half-way point of the stipe and no elongated inflated elements and apparently no encrusting pigment in the pileipellis. According to Corner & Bas (1962) *Amanita fritillaria* (Berk.) Sacc. f. *fritillaria* has more ellipsoid spores (aver. $Q = 1.3\text{--}1.35$).

Another dark species from south-eastern Asia is *Amanita pilosella* Corner & Bas (1962), which has several characters in common with *A. perphaea*, but has considerably smaller fruit-bodies, more powdery to subvillose volval remnants on the pileus and a grey annulus with blackish edge. It should be mentioned here that *A. pilosella* has the same type of elongate inflated elements in the pileipellis as does *A. perphaea*.

Amanita tristis Corner & Bas (1962) from Singapore has smaller and more ellipsoid spores ($4.9\text{--}6.1\text{--}7.0 \times 4.3\text{--}4.6 \mu\text{m}$, aver. $Q = 1.3\text{--}1.35$) and the volval warts on the pileus and the base of the stipe are entirely pale greyish to whitish.

Amanita fuscobrunnea A.E. Wood (1997) from Australia has smaller fruit-bodies, a grey pileus with dark pyramidal warts and a stipe entirely decorated with brown powdery granules.

Amanita echinulata Beeli (1927, 1935) from the Congo was placed by Gilbert (1940) in section *Lepidella* (then called *Aspidella*). However, Bas (1969) demonstrated that it belongs to section *Validae* and suggested a position near *A. pilosella*. It has a dark brown pileus with very dark conical warts and a dark brown stipe with a dark grey-brown annulus. Its type has small globose to subglobose spores ($5.5\text{--}6.5 \times 4.5\text{--}5.5 \mu\text{m}$; pers. obs. C. Bas.).

Amanita morrisii Peck (1910) from North America also has a very dark pileus, at least when young. However, it has no conical volval warts but pallid volval patches on the initially dark grey-brown to blackish brown pileus and, according to Tulloss (1991), larger, more ellipsoid spores ($7-9.5 \times 5.5-7 \mu\text{m}$, $Q = 1.28-1.42$).

Amanita spissacea S. Imai (1933) from Japan also has crust-like volval patches, in this case very dark grey-brown on a paler background and has brown powdery scales on the stipe. This species has globose to subglobose spores. Yang (1997) suggested that *A. spissacea* is a synonym for *A. fritillaria*.

Since so far pigments in the pileipellis of *Amanita* have always been recorded as a vacuolar, it is remarkable that the very dark superficial hyphae on the pileus of *A. perphaea* have an additional encrusting pigment. This could mean that these hyphae represent a fibrillose, strongly fragmented inner layer of the volva. Encrusting pigments in the volval tissues of *Amanita* have been recorded before.

Amanita perphaea is commonly eaten by Patamona Indians and is called 'Pulutukwe'.

Subgenus AMANITA

Section Amanita

Amanita calochroa C. Simmons, T. Henkel & Bas, *spec. nov.* — Fig. 4, Plate 6

Pileus 18–30 mm latus, primo hemisphaericus vel convexus margine laevis, postea applanatus margine valde sulcatus, centro applanatus vel depressus, fragmentis volvae pulveraceis rubris vel aurantio-rubris dense obsitus. Lamellae liberae, subdistantes, albae vel cremeae; lamellulae truncatae. Stipes 40–60 × 2.5–5 mm, basi bulbosus, cremeus; pars supera bulbi aurantio-pulveracea, exannulatus.

Sporae 6.3–7.8 × 5.5–7.8 μm , globosae vel late ellipsoideae, inamyloideae. Fragmenta volvae cellulis turgidis, 19–43 × 13–21 μm , subglobois, ovoideis, ellipsoideis vel elongatis composita. Fibulae absentes.

Typus: Guyana, Pakaraima Mts, Ireng River, 22.V.1998, T. Henkel et al. (TH 6426; BRG holotypus, L isotypus).

Fruit-bodies terrestrial, gregarious, small, and fragile. Pileus 18–30 mm in diam., at first hemispherical to convex with smooth margin and flattened centre, later applanate with strongly sulcate-striate (about 0.3 to 0.5R), somewhat crenulate margin and depressed centre, densely covered with a bright red to orange-red (K&W 10A8 to 8A8), pulverulent to minutely subtomentose volval layer with age at margin breaking up into orange-red granules on a yellowish to cream background (in dried state pale yellow and covered with melon-yellow (5A6) powdery substance). Lamellae free, subdistant, thickish, narrow, with even to eroded edges, white to pale cream; lamellulae truncate. Stipe 40–60 × 2.5–5 mm, slightly tapering upwards, with 3–7.5 mm wide, sometimes slightly rooting, bulbous base, white to pale yellow, and lightly covered with orange powdery substance forming a denser pulverulent orange zone on upper part of bulb. Annulus absent. Spore print not available.

Spores [25/2] 6.3–7.8(–8.4) × 5.5–7.8 μm , $Q = 1.0-1.2(-1.4)$, aver. $Q = 1.06-1.19$, globose to broadly ellipsoid, rarely ellipsoid, with smooth, thin walls, usually with one oil droplet, with rather narrow, but relatively prominent, hilar appendix, colourless, non-amyloid. Basidia 24–38 × 8–11.5 μm , clavate, shorter and broader near edge, 19–21 × 12.5–13.5 μm , 4-spored, clampless. Subhymenium pseudoparenchymatic (reinflating very badly); elements short and broad. Marginal tissue present as small \pm conical teeth, pale yellowish in NH_4OH or just a strip of rather amorphous tissue; teeth consisting of subglobose, broadly ellipsoid and pyriform elements, 9–26(–36) × 8.5–18 μm , at least partly attached terminally

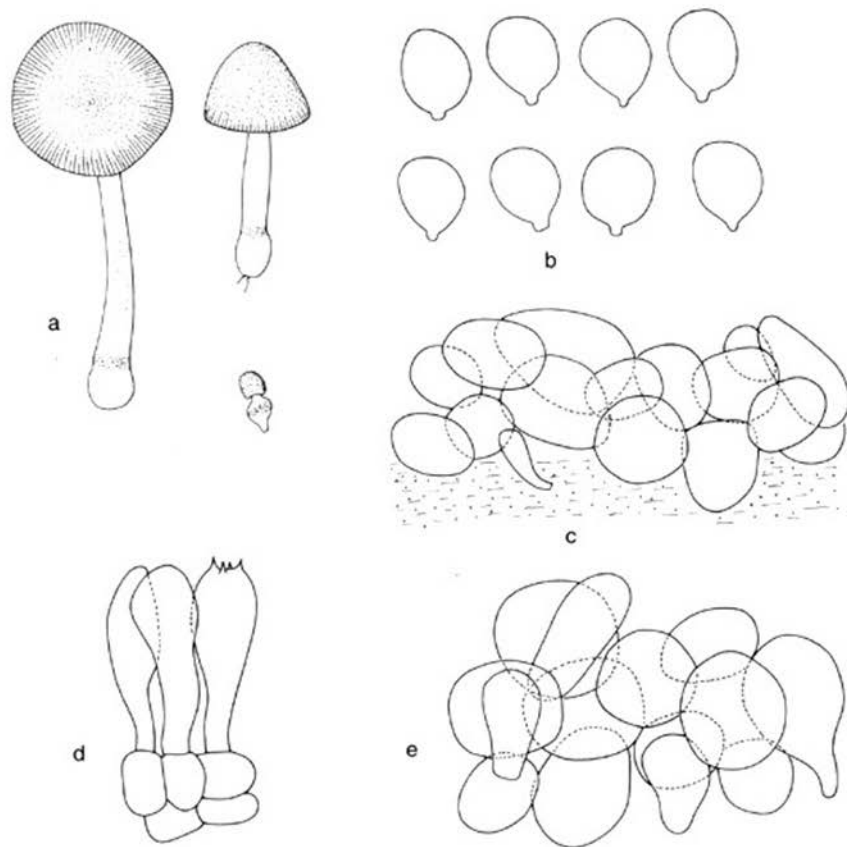


Fig. 4. *Amanita calochroa*. a. Basidiocarps, $\times 1$; b. spores, $\times 1500$; c. velar remnants on gelatinized pileipellis, $\times 500$; d. basidia and subhymenium, $\times 1000$; e. marginal tissue of lamella, $\times 500$ (all from holotype).

to hyphae. Hymenophoral trama bilateral, with a thick subhymenium and rather wide to narrow central stratum. Pileipellis consisting of a strongly gelatinised (in 5% KOH) layer about $70\ \mu\text{m}$ wide, consisting of \pm radial, $2\text{--}4.5\ \mu\text{m}$ wide hyphae, in upper part disintegrated. Volval remnants on pileus consisting of a mass of broadly ellipsoid to ovoid, rarely subglobose or elongate inflated elements, $(14\text{--})19\text{--}43 \times (14\text{--})13\text{--}21\text{--}(30)\ \mu\text{m}$, with thin walls, pale yellow in NH_4OH , probably at least partly in short chains, but connection of elements not clearly seen (nearly all elements remaining slightly to strongly collapsed); inflated elements much more abundant than hyphae. Volval remnants on stipe similar to those on pileus, but with an even higher ratio of inflated elements to hyphae. Context of stipe made up of firm, pale, brownish orange tinged 'cortex' and a wide, fluffy, whitish pith, with few, relatively small, up to $175 \times 27\ \mu\text{m}$ large acrophysalides; at centre consisting of loose tissue of $3\text{--}6\ \mu\text{m}$ wide interwoven to longitudinal hyphae. No clamp-connections observed.

Habitat & distribution — Terrestrial, gregarious in litter mats accumulated in crooks of the trunks of *Dicymbe corymbosa* (Paluyawek) in riverine and adjacent slope forest dominated by *D. corymbosa*, and *D. altsonii* (Edubayek) with *Micrandra glabra* (R.E. Schult.) R.E. Schult. (Euphorbiaceae; Suruwayek), *Moronobea* Aubl. spec. (Guttiferae; Morombayek), and other mixed hardwoods on sand soils with thick organic matter accumulations on grey sands and exposed sandstone boulders and cliff faces. So far known only from the Pakaraima Mountains in western Guyana.

Collections examined. GUYANA: Potaro-Siparuni, Pakaraima Mountains, Upper Ireng River: 0.75–1.5 km downstream from Kurutuik Falls, forest near ridge trail and slopes adjacent river, 14 Feb. 1997, T. Henkel et al. (field notes only; BRG); Forest adjacent Sukabi River, 1–2 km upstream from confluence with Ireng River, 22 May 1998, T. Henkel & L. Williams (TH 6426, holotype BRG, isotype L); Mt Kukuinang, fringing forest around southern edge of savannah, about 3 km south-west from peaks of mountain, 25 May 1998, T. Henkel et al. (TH 6589; BRG, L).

Amanita calochroa is a beautiful species with gregariously growing small fruit-bodies of which the pileus is covered by bright red to orange-red powdery volval remnants. As in addition it has non-amyloid spores, a sulcate-striate pileus margin and a stipe with a bulbous base, it finds its place in section *Amanita*. Within this section several small to medium-sized species with a red or orange pileus and a friable volva are known, but those differ from *A. calochroa* in the following ways:

Amanita aurantiovelata Schalkw. & G.M. Jansen (1982) described from Chile. Originally the collector thought the present taxon to represent *A. aurantiovelata*, but that species has considerably larger spores (8–11.5 × 6.5–8 µm), abundant clamp-connections, and larger fruit-bodies with at least some conical volval warts.

Amanita parcovolvata (Peck, 1900) E.J. Gilbert (1941a) from North America has a glabrous, viscid, yellow-orange to red pileipellis with dirty yellow to cream warts to patches. Moreover its spores are much larger and ellipsoid to nearly cylindrical.

Amanita rubrovolvata S. Imai (1939) described from Japan and occurring widely in Eastern Asia, differs by the presence of an annulus, a volval layer on the pileus that tends to break up into patches and warts, and has larger, more (sub)globose spores.

Amanita armeniaca A.E. Wood (1997) from Australia (Sydney) is a species with a bright orange pileus with membranous, dull cream volval patches near the centre and has an annulus. In addition its spores are larger ((7.4–)8.1–10.2 × 6.9–9.6 µm).

Amanita subfrostiana Z. Yang (1997) described from south-west China and Tibet, has a red to orange pileus, with yellowish to orange, powdery to floccose volval remnants. But it differs by having an annulus, larger spores and clamps.

Amanita bingensis Beeli (1931) described from the Congo region has an orange-yellow powdery volva and lacks an annulus; its spores are much smaller (5–6 × 3–4.5 µm).

Amanita chrysoleuca Pegler (1983), discovered on Martinique and also occurring on the Virgin Islands (Miller et al., 2000), and in Panama and Florida (Tulloss, pers. comm.), resembles the present species in several aspects, such as small size, strongly friable volva, lacking annulus and bright colours, but has a more orange-yellow to yellow pileus and somewhat larger (7–9.5 × 4.5–6 µm), considerably more ellipsoid to oblong spores (aver. Q = 1.56), according to Miller et al. (2000) even still larger (7–10(–10.5) × 5–7 µm, aver. Q = 1.31).

***Amanita lanivolva* Bas — Fig. 5, Plate 3**

Amanita lanivolva Bas, Persoonia 10 (1978) 12.

Fruit-body solitary, medium-sized, slender, terrestrial. Pileus 30–70 mm in diam., first plano-convex, later with wide central depression, with sulcate-striate margin, (original colour not recorded, but in colour-slide from pale brown at margin, K&W about 6C4, to dark brown at centre, about 7E6), glabrous or with a small cluster of grey, conical volval fragments or of small grey volval patches at centre. Lamellae free, crowded, narrow, cream; lamellulae truncate. Stipe 65–88 × 6–8 mm, tapering upwards, exannulate, white, glabrous, at base slightly, but distinctly bulbous with membranous, rather narrowly saccate, grey-brown tomentose to tomentose-squamulose volva enclosing lower 1/4 to 1/3 of stipe like a sock. Spore print not available.

Spores [40/3] 7.4–9.8 × 5.0–6.6(–7.2) µm, Q = 1.3–1.65, aver. Q = 1.4–1.55, broadly ellipsoid to ellipsoid with very slightly thickened wall, non-amyloid, usually with oil droplet, with rather broad hilar appendix. Basidia 24–33 × 8–11 µm, 4-spored, with inconspicuous clamp-connection. Subhymenium made up of subglobose to broadly ellipsoid, 8–25 µm wide elements. Marginal tissue consisting of a broad, sterile, ± amorphous strip of tissue, with outer cells difficult to reinflate, but between amorphous layer and edge of hymenium with quite a few (sub)globose to broadly ellipsoid and broadly clavate elements of up to 20 µm wide and with slightly thickened wall. Hymenophoral trama bilateral. Pileipellis consisting of a gelatinised suprapellis of colourless, thin-walled, 2–3 µm wide, interwoven hyphae, over a subpellis of 4–8 µm wide, interwoven brown hyphae. Volval remnants on pileus consisting of very loosely interwoven brown hyphae carrying (sub)globose, ovoid and broadly ellipsoid brown elements 56–78 × 19–29 µm, with slightly thickened walls. Trama of stipe acrophysalidic with up to 175 × 40 µm large terminal elements. Clamp-connections abundant.

Habitat & distribution — Riverine swamp forest on flat ground dominated by *Dicymbe altsonii* (Edubayek) and *D. corymbosa* (Paluwayek) with scattered associated hardwoods. Known from Brazilian Amazonia and western Guyana.

Collections examined. GUYANA: Pakaraima Mountains, Upper Ireng watershed: Upper Ireng River, forest adjacent Sukabi River, 1–2 km upstream from confluence with Ireng River, 22 May 1998, *T. Henkel & L. Williams* (TH 6432; BRG, L); Upper Ireng River, forested slopes adjacent last bank of river, 27 May 1998, *T. Henkel* (TH 6640; BRG, L); Mt. Kubinoang, fringing forest around southern edge of savannah, about 3 km south-west from peak of mountain, 25 May 1998, *T. Henkel et al.* (TH 6593; BRG, L).

This is the second time that *A. lanivolva* is recorded and the first time from outside Brazilian Amazonia. The present description agrees rather well with the original one, except that in some specimens from Guyana a few small, conical volval warts are present at the centre of the pileus.

There is no longer any doubt about the presence of a small, but distinct basal bulb in this species. Because of this bulb *A. lanivolva* does clearly not belong to section *Vaginatae*, but to section *Amanita*, where, however, it stands well apart from all the other species on account of its saccate volva.

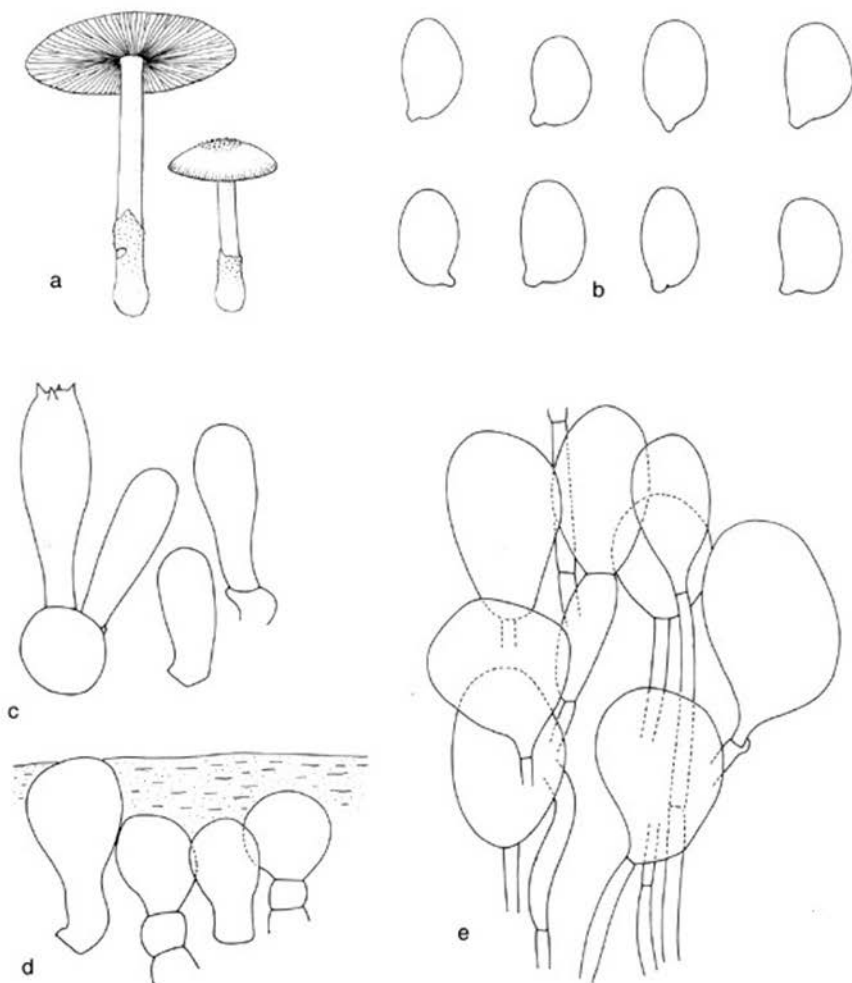


Fig. 5. *Amanita lanivolva*. a. Basidiocarps, $\times 0.5$; b. spores, $\times 1500$; c. young basidia and subhymenium, $\times 1000$; d. elements of marginal tissue of lamella, $\times 1000$; e. slightly crushed longitudinal section of volval wart on centre of pileus, $\times 500$ (a & e: TH 6432; b–d: TH 6640).

***Amanita xerocybe* Bas — Fig. 6, Plate 7**

Amanita xerocybe Bas, Persoonia 10 (1978) 7.

Fruit-body medium-sized, slender, fragile, solitary to gregarious. Pileus 20–46 mm, young hemispherical to paraboloid, later plano-convex with broadly rounded umbo or with slightly depressed centre, with broad widely sulcate-striate margin (0.25–0.65R), cream (K&W 4A2 – 4A3) with dark red-brown to fairly dark orange-reddish brown (in colour-slides 7E8 to 7D8, later between 7D7 and 7C8) powdery-granular substance over entire

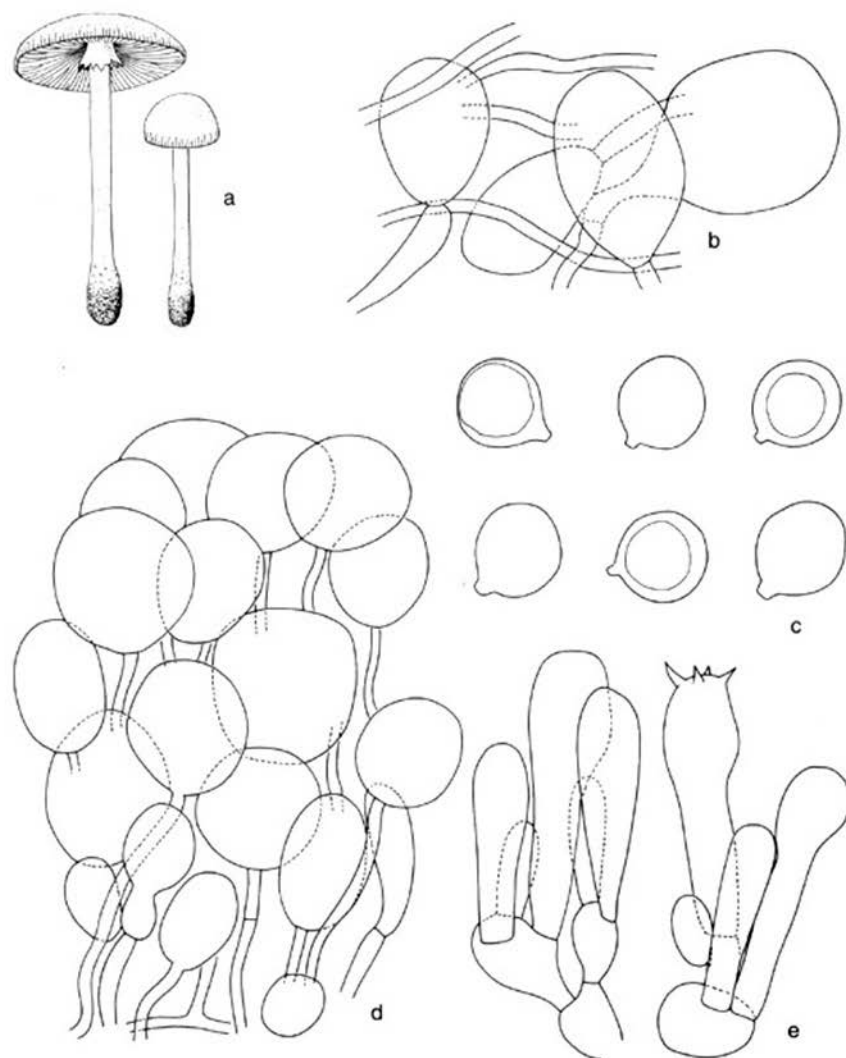


Fig. 6. *Amanita xerocybe*. a. Basidiocarps, $\times 0.5$; b. marginal tissue of lamella, $\times 1000$; c. spores, $\times 1500$; d. velar remnants on pileus, 0.25 R from centre, $\times 500$; e. basidia and subhymenium, $\times 1000$ (a: reconstructed; b & c: TH 6261; d & e: TH 6434).

surface but more concentrated at central region. Lamella free, distant, narrow, but becoming ventricose, white to cream, with minutely subflocose-subgranular, concolorous edge; lamellulae not found. Stipe up to 82×6 mm, slightly tapering upwards, with up to 9 mm wide, clavate, bulbous base, pallid with minute brown granules all over, at base with scattered, brown, small, irregular, wart-like volval fragments. Annulus submembranous, skirt-

like, but very fragile (lacking in all dried specimen studied, but clearly visible on colour-slide), white and sulcate-striate above and orange-brown with darker brown granules below. No spore print available.

Spores [49/3] $6.0-9.0 \times 6.0-8.6(-9.4) \mu\text{m}$, $Q = 1.0-1.1$, aver. $Q = 1.03-1.04$, globose to subglobose, with normal hilar appendix, with smooth, slightly thickened wall, non-amyloid. Basidia $37-51(-62) \times (8.0-9.0-12.0(-14.0) \mu\text{m}$, 4-spored, clampless. Subhymenium consisting of irregularly subglobose and subellipsoid elements, $15-19 \times 10-14 \mu\text{m}$. Marginal tissue a rather broad strip of amorphous, partly gelatinised tissue with irregularly disposed, globose to broadly ellipsoid, terminal elements, $20-42(-51) \times 13.5-25(-32) \mu\text{m}$, on $2.5-4 \mu\text{m}$ wide hyphae. Trama of gills bilateral. Pileipellis consisting of a non-gelatinised layer of $2.5-6 \mu\text{m}$ wide, colourless, thin-walled hyphae between adnate volval tissue and trama of pileus. Volval remnants on pileus (near centre) pale yellowish brownish in NH_4OH , in scalp seen from above: an irregular arrangement of globose, ellipsoid and ovoid, thin-walled, terminal elements, $22-49(-70) \times (12.5-18-41(-58) \mu\text{m}$; in radial section: made up of single, erect, terminal, inflated elements on $3-5.2 \mu\text{m}$ wide, septate hyphae. Volval remnants on stipe similar to those on pileus, but hyphae more abundant. Trama of stem acrophysalidic: inflated elements up to $325 \times 24 \mu\text{m}$. Clamp-connections absent.

Habitat & distribution — Riverine swamp forest and adjacent slope forest dominated by *Dicymbe corymbosa* (Paluwayek) and other mixed hardwoods, on sand soils with thick organic accumulations and on exposed sandstone and cliff boulders. Known now from Brazilian Amazonia, and from western Guyana

Collections examined. GUYANA: Potaro-Siparuni, Pakaraima Mountains: Upper Ireng watershed, Suruwaburu Creek, 1–2 km upstream from juncture with Jurka River, immediate riverine environs, 2 March 1997 (TH 6217; BRG); Upper Ireng River, forest adjacent Sukabi River, 1–2 km upstream from confluence with Ireng River, 22 May 1998 (TH 6434; BRG); Upper Ireng River, Mt. Kukuinang, fringing forest on western side of savannah, ± 3 km WSW from mountain peak, 25 May 1998 (TH 6614; BRG, L); Upper Ireng River, 0.2–1 km downstream from Kurutuik Falls, slopes adjacent west side of river, 15 Feb. 1997 (TH 6261; BRG, L).

Amanita xerocybe Bas, described in 1978 from Brazilian Amazonia, is recorded for the second time and for the first time from outside Brazil.

The specimens from Guyana usually have a somewhat smaller pileus and a relatively longer marginal striation, whereas the granular-subfloccose volval remnants have a somewhat more reddish brown colour. However, all the other characters being in agreement with the protologue of *A. xerocybe*, we do not hesitate to consider the material from Guyana conspecific with that from Brazilian Amazonia.

Amanita xerocybe is a very characteristic species that belongs to section *Amanita* because of its non-amyloid spores, friable volva, and bulbous base of the stipe.

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CHEMOTAXONOMICAL AND MORPHOLOGICAL OBSERVATIONS
IN THE GENUS OMPHALOTUS (OMPHALOTACEAE)

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Comparative thin-layer chromatography – for the first time applied to *Omphalotus olivascens* var. *olivascens*, *O. olivascens* var. *indigo*, *O. nidiformis*, and to *O. mexicanus* – revealed strikingly similar pigment patterns for all *Omphalotus* species except *O. mexicanus*. Atromentin, thelephoric acid and pulvinic acid derivatives were found in dried material and/or culture extracts of all species. Illudin S and illudin M were detected in *O. mexicanus* by high-performance liquid chromatography-atmospheric pressure chemical ionisation mass spectrometry. Data on morphological features of all described *Omphalotus* and *Lampteromyces* species are listed, illustrated, and summarized in a key. New combinations in the genus *Omphalotus* are proposed for *Lampteromyces japonicus* and *L. mangensis*.

In classical fungal taxonomy morphological-anatomical characters are often used exclusively to classify specimens. Unfortunately, many fungi show only few 'good' morphological characters. Therefore, if possible, additional methods should be applied to elucidate their taxonomic relationships. Besides eco-physiological (e.g. host specificity) or molecular (e.g. DNA sequences) characters the investigation of pigment patterns still helps to clarify discrete taxonomic limits. Following Singer (1986) the order Boletales is mainly characterized by the occurrence of "pigments of the variegatic acid type or derivatives (or otherwise related to pigments commonly found in boletes)". The detection of certain Boletales pigments in members of the genera *Omphalotus* Fayod and *Lampteromyces* Sing. was one of the main motives of Bresinsky & Besl (1979b) to include them into the Paxillaceae. Further investigations (Kämmerer et al., 1985) focused on physiological characters of some species of the afore mentioned genera revealed relevant differences to other members of the Paxillaceae: based on both the occurrence of sesquiterpenes of the illudane type and the formation of so called 'white soft-rot', features not found in other members of the Paxillaceae, the new family Omphalotaceae Bresinsky was established. Until recently most chemotaxonomical studies in the genus *Omphalotus* were focused on two species only: *O. olearius* and *O. illudens*. Therefore, the objective of the present study was to gather additional chemotaxonomical and morphological data aiming at an improvement of the taxonomic structure of the genus which is still unclear and incomplete.

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METHODS

Microscopic descriptions were made from sections or pieces of tissue taken from dried basidiomata, mounted in 2.5% KOH (pigment topology was studied in aqua dest. because most of the pigments are soluble in KOH). A Nikon Labophot microscope with an automatic photographic system (Sony Multiscan Video Printer UP-930) was used for light microscopy (LM). Drawings of anatomical features are based on video prints. Video prints of spores taken from a spore print or from the stipe were used for measurements also (sample size for each collection > 30). Measurements are given in the form (minimum) mean \pm standard deviation (maximum). Colour notations are mainly based on original descriptions.

For thin layer chromatography (TLC) the different strains were cultivated on MEA plates (malt extract agar, Merck 5398) for 20 days at 20°C and two hours light per day. The cultures (each with 20 ml medium) were liquefied at about 90°C and extracted with hot ethanol acidified with a few drops of 1N HCl. This raw extract was shaken out three times in a separating funnel with an equal amount of pure ethyl acetate. The ethyl acetate phase was dried with anhydrous sodium sulfate, filtrated and evaporated to dryness at 50°C. The residue was resolved in 1.5 ml ethyl acetate. Twenty μ l of these extracts were spotted on silica gel TLC-plates (Merck 1.05721) with toluene : ethyl formate : formic acid (10 : 5 : 3) as a solvent according to Bresinsky (1974). If bands were too weak for characterization, the amount of spotted extracts was increased to 30 or 40 μ l. For separating thelephoric acid, methyl ethyl ketone : H₂O : formic acid (250 : 25 : 1) was used as solvent. Pigment patterns were examined in daylight and under UV (366 nm). For determining pulvinic acid derivatives a K₃[Fe(CN)₆]/NaHCO₃ solution was used as spray reagent: variegatic acid and xerocomic acid change their colour from yellow to blue. Incubating the chromatogram in ammonium vapour for half an hour changes the colour of these two pigments to red. Atromentic acid shows no colour change with K₃[Fe(CN)₆]/NaHCO₃ but turns violet with ammonium (Bresinsky, 1974). Thelephoric acid was identified following Bresinsky & Rennschmied (1971). For reference the following compounds were used: xerocomic acid and variegatic acid isolated from *Boletinus cavipes*, thelephoric acid from *Thelephora terrestris*, atromentin from *Paxillus atrotomentosus*, and atromentic acid provided by Dr. H. Besl (Regensburg, Germany). Isolation procedures were carried out following Kögl & Postowsky (1924), Bresinsky & Orendi (1970), and Bresinsky & Rennschmied (1971).

For detection and identification of illudin S and illudin M by high-performance liquid chromatography – atmospheric pressure chemical ionisation – mass spectrometry (HPLC-APCI-MS) liquid cultures of *O. mexicanus* were used according to Kirchmair et al. (1999). Illudin S and illudin M standards were kindly provided by Trevor C. McMorris, San Diego, USA. The HPLC system consisted of a low-pressure gradient micro pump (model Rheos 2000, Flux Instruments, Karlskoga, Sweden) controlled by a personal computer, a vacuum degasser (Knauer, Berlin, Germany), and an injector (model Cheminert C3-2006, Valco Instruments Co. INC., Houston, TX, USA) with a 5 μ l sample loop. LC separations were performed with a gradient of 10–42% acetonitrile in 0.1% aqueous acetic acid in 20 min at a flow-rate of 250 μ l/min. The Nucleosil ODS (3 μ m, 100 Å) column packing material was obtained from Macherey & Nagel (Düren, Germany) and packed into a 150 \times 2 mm i.d. stainless steel column (Grom, Herrenberg, Germany) with the help of an air-driven high-pressure packing pump (Knauer, Berlin, Germany).

APCI-MS3 was performed on a Finnigan LCQ quadrupole ion trap mass spectrometer (Finnigan MAT, San Jose, CA, USA) equipped with the atmospheric pressure chemical

ionisation ion source. For LC-MS analysis with heat assisted pneumatic nebulisation, a corona current of 5 μ A at 5 kV and a sheath gas flow of 60 arbitrary units were employed. The temperatures of the heated nebuliser and the heated pileusillary were set at 450 and 200°C, respectively. Highly selective MS³ mass spectra for illudin S and illudin M were obtained using the transitions 247.1→217.1→full scan from 60–250 u and 231.1→213.2→full scan from 60–250 u, respectively, with a relative collision energy of 15%. Total ion chromatograms and mass spectra were recorded on a personal computer with the Navigator software version 1.2 (Finnigan).

Material studied

Cultures. For a list of examined cultures see Table I.

Omphalotus illudens (Schwein.) Bresinsky & Besl.

Collections examined. BELGIUM: Liège, Nivezé, 20 Sept. 1997, *J. Prados*, BR 70376.51; Namur, Briquemont-Rochefort, 13 Sept. 1981, *P. Heinemann*, BR 21676.45; Namur, Couvin, 22 Sept. 1977, *C. Cnobs*, BR 12717.10, BR 12714.09; Namur, Ciergnon-Briquemont, 8 Sept. 1987, *P. Heinemann*, BR 2704.85. — USA: Bethesda MD, 7422 Hampton Lane, around stumps of hardwood trees, 7 Sept. 1967, *Ann Tenks*, OKM 6086; Michigan, Ann Arbor, 20 Oct. 1973, *A.H. Smith*, OKM 9597.

Omphalotus mexicanus Guzmán & Mora.

Collections examined. MEXICO: Sierra del Tigre, Región de Mazamitla, carretera de Tamazulajiquilpan, Jalisco, 23 June 1983, *G. Guzmán*, XAL 23474; Cerca de San Cristobal de Las Casas, Chiapas, 23 June 1983, *G. Guzmán*, XAL 29252.

Omphalotus nidiformis (Berk.) O.K. Mill.

Collection examined. AUSTRALIA: NSW, Blue Mts, Mt Wilson, on bark of living trees in the rain forest, 7 Apr. 1983, *E. Horak*, ZT 2144.

Omphalotus olearius (D.C.: Fr.) Sing.

Collections examined. AUSTRIA: near Vienna, on hardwood trees, 1 Oct. 1952, *M. Moser*, IB 1952/115. — CROATIA: Veli Losinj, St. Juan, on soil (roots?) between *Pinus halepensis*, 6 Oct. 1997, *M. Kirchmair*, IB 1997/776; Mali Losinj, Èikat Bay, on *Pinus halepensis*, 9 Oct. 1997, *M. Kirchmair*, IB 1997/777; Mali Losinj, Èikat Bay, on *Quercus ilex*, 9 Oct. 1997, *M. Kirchmair*, IB 1997/778; Aéroport Losinj, Èunski Bay, on *Cistus sp.*, 7 Oct. 1997, *M. Kirchmair*, IB 1997/779. — FRANCE: Port Man, Ile de Cros, Var, on *Quercus ilex*, 30 Oct. 1977, *M. Moser*, IB 1977/222; Port Man, Ile de Cros, Var, on *Quercus ilex*, 31 Oct. 1978, *M. Moser*, IB 1978/492. — ITALY: Prov. Parma, Monte Penna, on stump of *Castanea sativa*, 12 Oct. 1994, *M. Kirchmair*, R. Pöder, IB 1994/904; Prov. Parma, Marzocco, on *Quercus sp.*, 6 Oct. 1996, *M. Kirchmair*, IB 1996/674; Sicilia, Fascio tre, under *Quercus cerris*, 13 Nov. 1998, *R. Piérart*, BR 96550.35; Aressano, 24 Sept. 1997, *M. Moser*, IB 1997/780. — KENYA: Mt Elgon, under olive trees, 8 May 1964, *P.H. Erwin*, K 50515. — TANZANIA: Magamba Forest Reserve, Lushoto, W. Usambora Mts, Tanga Prov., on decaying wood, 23 Apr. 1968, *D.N. Pegler*, K 50514.

Omphalotus olivascens var. *indigo* Moreno, Esteve-Rav., Pöder & Ayala.

Collections examined. MEXICO: Baja California Norte, Santa Rosa, on stump of *Quercus agrifolia*, 10 Nov. 1993, *G. Moreno*, AH 16295; Baja California Norte, San Antonio de las Minas, on stump of *Quercus agrifolia*, 14 March 1990, *G. Moreno*, AH 13187; Baja California Norte, Las Lomas, San Antonio de las Minas, on stump of *Quercus agrifolia*, 2 Feb. 1996, *G. Moreno*, IB 1996/731.

Omphalotus olivascens var. *olivascens* Bigelow, O.K. Mill. & Thiers.

Collections examined. USA: California, San Mateo Co., Junipero Serra Park, San Bruno, on oak stumps, 4 Dec. 1960, *H.D. Thiers*, HDT 8547; California, Cleveland National Forest, Decker Canyon, on a stump of *Quercus agrifolia*, 12 March 1984, *O.K. Miller Jr.*, OKM 20911.

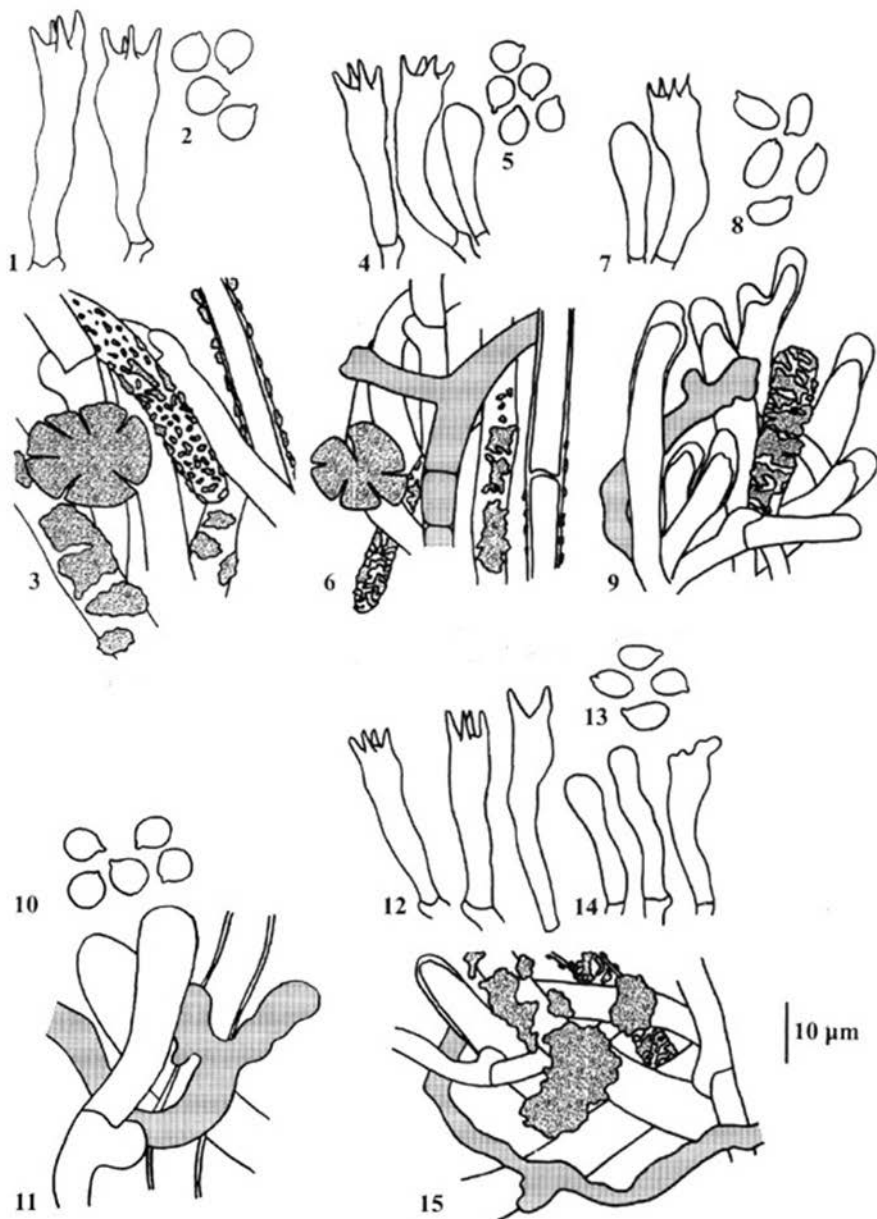
Omphalotus subilludens (Murrill) Bigelow.

Collection examined. USA: Austin, Texas, on hardwood, 3 Nov. 1983, *O.K. Miller Jr.*, OKM 20850.

RESULTS

Morphological observations

Relevant morphological characters and observations are discussed here and presented in Tables II & III and in Figs. 1–34.



Figs. 1–15. Microanatomical features of *Omphalotus* taxa: basidia and basidioles, basidiospores, cheilocystidia (14), and elements of the pileipellis. *Omphalotus olearius* (1–3), *O. illudens* (4–6), *O. aff. olearius*, 'Kenya collection' (7–9), *O. aff. olearius*, 'Tanzania collection' (10–11), and *O. subilludens* (12–15). Refractive hyphae (light grey) and pigment incrustations (dark grey) are highlighted by shadings.

Table 1. List of cultured strains studied. Acronyms: CBS = Centraalbureau voor Schimmelcultures, Baarn, The Netherlands; VT = Virginia Polytechnic Institute and State University, Blacksburg, Virginia, USA; UT = University of Tennessee, Texas, USA; OLE = Institute of Microbiology, Innsbruck, Austria.

Strain	Species	Location	Substrate	Date
CBS 101.448	<i>Omphalotus olivascens</i> var. <i>indigo</i>	Baja California, Mexico	<i>Quercus agrifolia</i>	14 Jan. 1995
CBS 101.447	<i>Omphalotus olivascens</i> var. <i>indigo</i>	Baja California, Mexico	<i>Quercus agrifolia</i>	1 Feb. 1995
VT 455	<i>Omphalotus olivascens</i> var. <i>olivascens</i>	California, USA	unknown	25 Nov. 1970
VT 1178	<i>Omphalotus olivascens</i> var. <i>olivascens</i>	California, USA	<i>Quercus agrifolia</i>	10 Feb. 1982
OLE 1	<i>Omphalotus olearius</i>	Prov. Parma, Italy	<i>Castanea sativa</i>	12 Oct. 1994
OLE 2	<i>Omphalotus olearius</i>	Prov. Parma, Italy	<i>Quercus</i> sp.	6 Oct. 1996
CBS 141.34	<i>Omphalotus illudens</i>	USA	unknown	1934
VT 0452	<i>Omphalotus illudens</i>	California, USA	unknown	10 Dec. 1971
VT 1946	<i>Omphalotus nidiformis</i>	Western Australia	on buried wood	10 June 1989
VT 1948	<i>Omphalotus nidiformis</i>	Western Australia	on dead <i>Banksia</i> <i>menziesii</i>	11 June 1989
VT 1949.01	<i>Omphalotus nidiformis</i>	Western Australia	<i>Acacia</i>	8 June 1989
VT 1490	<i>Omphalotus nidiformis</i>	Western Australia	unknown	13 July 1980
CBS 660.85	<i>Omphalotus subilludens</i>	Texas, USA	decaying wood stump	1967
CBS 101.446 (= UT 4866)	<i>Omphalotus mexicanus</i>	Guatemala	decaying wood stump	—
CBS 446.69	<i>Lampteromyces japonicus</i>	Japan	decaying <i>Fagus</i> sp.	March 1969
CBS 374.51	<i>Lampteromyces japonicus</i>	Japan	unknown	Feb. 1951

Omphalotus olearius (D.C.: Fr.) Sing. — Figs. 1–3

Omphalotus olearius (D.C.: Fr.) Sing., Lilloa 22 ('1949' 1951) 181.

Omphalotus illudens (Schwein.) Bresinsky & Besl — Figs. 4–6

Omphalotus illudens (Schwein.) Bresinsky & Besl, Beih. Sydowia 8 (1979) 106.

By many mycologists *O. illudens* is seen as conspecific with *O. olearius* (e.g. Watling & Gregory, 1989). A greenish staining of the pileipellis when treated with 25% ammonia solution or 30% KOH could be observed in all examined species of *Omphalotus*, a reaction easily recognisable when small fragments are squeezed between two glass slides. In contrast to other reports (e.g. Kuyper, 1995) refracting hyphae were found in the pileipellis of all *Omphalotus* species but they are rather scarce in most *O. olearius* collections. Thus, the only distinct morphological differences between *O. olearius* and *O. illudens* are the form and colour of the basidiomata: umbonate and uniformly yellowish orange coloured basidiomata are characteristic for *O. illudens*. A papilla like umbo was never observed in *O. olearius*; its pileus was always darker (usually reddish brown) than lamellae and stipe.

Pegler (1977), who considered *O. olearius* and *O. illudens* as one species, reported the occurrence of *O. olearius* in East Africa. His collection from Tanzania (K 50514; Figs. 10–11) consists of very small specimens: pilei about 2 cm in diameter, stipe 2 × 0.5 cm;

Table II. Morphological characters in *Omphalotus* and geographic distribution of taxa.

Characters	<i>O. olearius</i>	<i>O. illudens</i>	<i>O. subilludens</i>	<i>O. olivascens</i> var. <i>olivascens</i>	<i>O. olivascens</i> var. <i>indigo</i>	<i>O. nidiformis</i>	<i>O. mexicanus</i>
Distribution	southern Europe	USA, east coast, northern Europe?	southern USA, Mexico	USA, Texas, California	Mexico, Baja California	Australia, Tasmania	Mexico, Guatemala
Colours							
Pileus	orange to dark reddish brown	yellow orange	orange to dark reddish brown	dull orange to dark olive orange	olive orange,	whitish to blackish brown	bluish black to blackish
Lamellae	deep yellow, orange yellow	orange	orange, orange brown, reddish brown	olive to olive yellow	olive to olive yellow	whitish with a pink touch to yellowish orange	bluish black to blackish
Stipe	deep yellow, orange yellow	orange	deep yellow, orange yellow	olive to olive yellow, brick-red	olive to olive yellow, brick-red	whitish at the tip, brownish below	bluish black
Context (pileus, stipe)	deep yellow, orange yellow	deep yellow	deep yellow, orange yellow	dull orange olivaceous	bluish grey to blue violet	pure white	blue violet
Spores							
Length (L) (μm)	(5.0)6.4 \pm 0.7 (8.0)	(4.6)5.0 \pm 0.3 (5.7)	(6.3)7.0 \pm 0.5 (8.1)	(6.2)7.4 \pm 0.5 (8.8)	(6.7)7.9 \pm 0.6 (9.7)	(5.0) 6.0 \pm 0.6 (7.0)	(5.4) 6.0 \pm 0.3 (6.7)
Width (W) (μm)	(4.6)5.7 \pm 0.6 (7.6)	(4.0)4.7 \pm 0.3 (5.2)	(4.2)4.8 \pm 0.3 (5.8)	(5.8)6.9 \pm 0.6 (8.2)	(6.0)7.4 \pm 0.6 (9.0)	(4.6) 5.5 \pm 0.6 (7.0)	(4.6)5.2 \pm 0.3 (5.8)
L/W	1.1 \pm 0.1	1.1 \pm 0.1	1.5 \pm 0.2	1.1 \pm 0.1	1.1 \pm 0.1	1.1 \pm 0.1	1.2 \pm 0.1
Volume (μm^3)	(56)110 \pm 32 (239)	(40)58 \pm 8 (77)	(57) 86 \pm 18 (145)	(112)184 \pm 42 (295)	(128)232 \pm 52 (412)	(55)96 \pm 28 (187)	(69)85 \pm 10 (104)
Pileipellis							
Refractive hyphae	rare	numerous	numerous	numerous	numerous	numerous	numerous
Incrusting pigment	orange to brownish	orange to brownish	orange to brownish	orange to brownish, violet	orange to brownish, violet	orange to brownish, violet	violet

Table III. Morphological characters in *Lampteromyces* and geographic distribution of taxa. Data are based on the original descriptions (Kawamura, 1915; Li & Hu, 1993; Zang, 1979).

Characters	<i>L. japonicus</i>	<i>L. mangensis</i>	<i>L. luminescens</i>
Distribution	Japan	Hunan, China	Xizang (Tibet), China
<i>Colours</i>			
Pileus	light brown, with yellowish or rosy tinges	white, sometimes with bluish tinges	white to pale purplish
Lamellae	white, sometimes with yellowish tinges	white to bluish violet	white
Stipe	light brown	white to pale bluish violet	white
Context (pileus, stipe)	white; dark purplish at the transition pileus-stipe	white, bluish violet the transition pileus-stipe	no data
<i>Size</i>			
Pileus		10–15 cm	5–9 cm
Flesh	1.3–2 cm		no data
Lamellae	9–18 mm	4–7(–11) mm	no data
Stipe	1.4–2.5 × 1.5–3 cm	2–3 × 2.5–3 cm	0.3–0.5 × 0.5–0.6 cm
Spores	smooth, 13–17 µm	smooth, 9.5–15 µm	rough, 12–14 µm
Cheilocystidia	none	none	fusiform
Gill edge	entire	entire	serrate
Annulus	present	present	absent

spore size (4.5) 5.2 ± 0.3 (5.8) × (4.2) 4.8 ± 0.3 (5.4) µm; quotient = 1.1 ± 0.1 ; volume = (41.8) 62.2 ± 12.0 (83.4) µm³; pileipellis with numerous refractive hyphae; other elements incrustated with pale yellow pigment. These characters resemble those of *O. illudens*. The collection from Kenya (K 50515; Figs. 7–9) consists of one specimen only: it differs from *O. olearius* by its distinctly elliptic spores: (6.3) 6.9 ± 0.5 (7.9) × (3.3) 3.8 ± 0.3 (4.5) µm; quotient = 1.8 ± 0.1 ; volume = (36.4) 53.9 ± 11.1 (82.7) µm³. The pileipellis is characterized by many hyphal tips with strongly thickened walls, some refractive hyphae, and few hyphal elements that are weakly incrustated with a pale yellow pigment. This set of features could not be found in any other investigated *Omphalotus* collection.

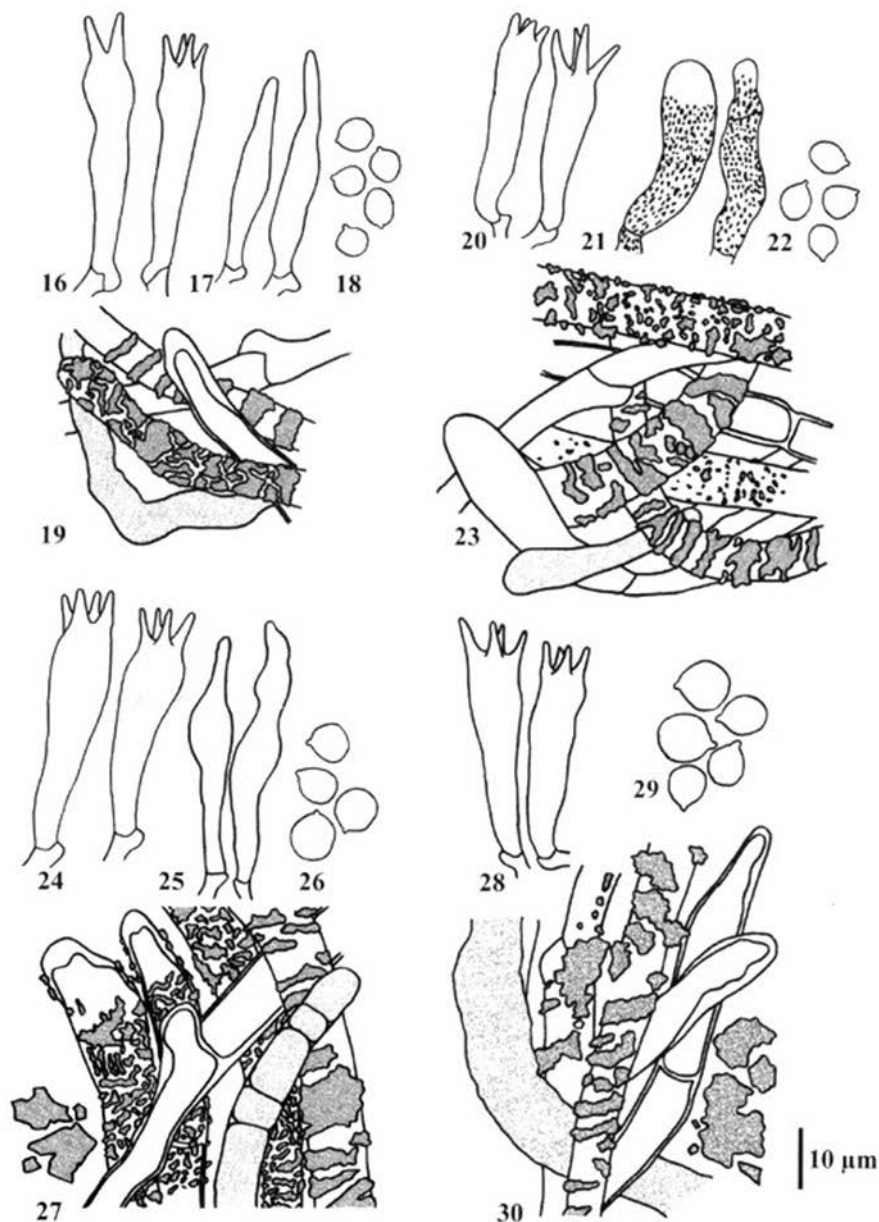
Omphalotus subilludens (Murrill) Bigelow — Figs. 12–15

Omphalotus subilludens (Murrill) Bigelow, Sydowia 35 (1982) 67.

This species is mainly characterized by its distinctly elliptic spores (quotient = 1.5 ± 0.2). The macroscopic characters are essentially the same as in *O. olearius* (Table II).

Omphalotus nidiformis (Berk.) O. K. Mill. — Figs. 16–19

Omphalotus nidiformis (Berk.) O. K. Mill., Mycol. Helv. 2 (1994) 93.



Figs. 16–30. Microanatomical features of *Omphalotus* taxa: basidia, basidiospores, cheilocystidia, terminal cells of the hymenophoral trama (21), and elements of the pileipellis. *Omphalotus nidiformis* (16–19), *O. mexicanus* (20–23), *O. olivascens* var. *olivascens* (24–27), and *O. olivascens* var. *indigo* (28–30). Refractive hyphae (light grey) and pigment incrustations (dark grey) are highlighted by shadings.

Populations of this taxon show at least two distinct colour forms: the colour of pileus ranges from white to brownish black. The context is always white (Miller, 1994). Size and shape of spores are of the *O. olearius* type (Table II).

Omphalotus mexicanus Guzmán & Mora — Figs. 20–23

Omphalotus mexicanus Guzmán & Mora, Bol. Soc. Mex. Mic. 18 (1983) 117.

Mora & Guzmán (1983) described amyloid elements in the gill trama (they called them 'pseudocystidia') and in the pileipellis. In our study amyloid elements could be found neither in the gill trama nor in the pileipellis. However, claviform to irregularly cylindrical terminal cells (Fig. 21) which are densely covered with very fine, dark violet crystal needles are rather frequent in the gill trama when observed in water. Also some hyphae in the pileipellis are incrustated with such crystals. This striking pigment dissolves in 2.5% KOH (the medium stains greenish) but remains unchanged in Melzer's reagent.

Omphalotus olivascens Bigelow, O.K. Mill. & Thiers var. **olivascens** & **Omphalotus olivascens** var. **indigo** Moreno, Esteve-Rav., Pöder & Ayala — Figs. 24–30

Omphalotus olivascens Bigelow, O.K. Mill. & Thiers var. *olivascens*, Mycotaxon 3 (1976) 363.

Omphalotus olivascens var. *indigo* Moreno, Esteve-Rav., Pöder & Ayala, Mycotaxon 48 (1993) 218.

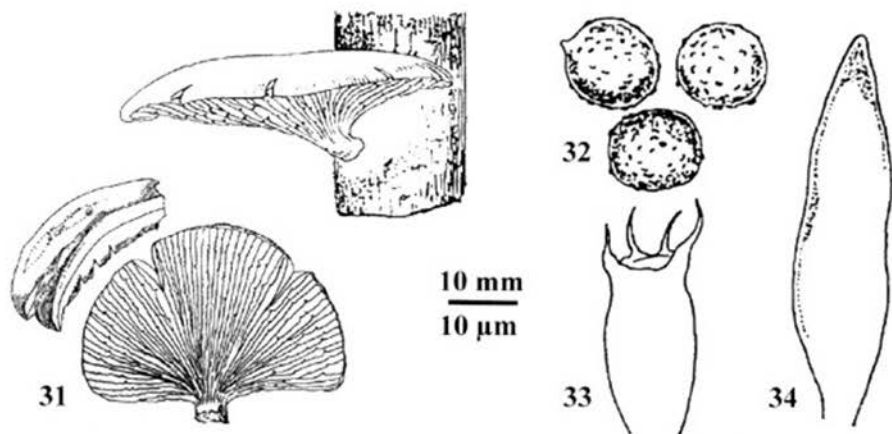
The two varieties of *O. olivascens* are distinguished from other species of *Omphalotus* by distinctive olive overtones in all parts of their basidiomata. The spores are usually larger as in other species of the genus (Table II), although Bigelow et al. (1976) mentioned that "some specimens have some smaller spores (5–7 µm) which lead to ranges which overlap with those of the other species". *Omphalotus olivascens* var. *indigo* shows in general the same features as the type variety but differs in its distinctly bluish grey to blue violet context (Moreno et al., 1993).

Lampteromyces Sing.

Lampteromyces Sing., Mycologia 39 (1947) 79–80.

The genus was originally based on one species: *L. japonicus* (Kawam.) Sing. (Singer, 1947). Singer (1986) delimited this genus from *Omphalotus* by its voluminous spores (up to 17 µm in diameter) and the presence of a veil. Chemical characters such as the occurrence of *Boletales* pigments and of sesquiterpenes of the illudane type are very similar in *L. japonicus* and in *Omphalotus* species (Kämmerer et al. 1985). Until now two further species in the genus *Lampteromyces* have been described: *L. luminescens* M. Zang and *L. mangensis* J. Li & X. Hu.

Lampteromyces luminescens is described to have no veil, punctate spores, fusiform cheilocystidia, serrate gill edges and, compared to the other two species, a relative slender stipe (Zang, 1979). For more characters see Table III and Figs. 31–34. Microchemical reactions (e.g. amyloidity of spores) are not mentioned in the original description. Although no material was available for our studies it seems likely that *Lampteromyces luminescens* belongs to *Lentinellus*.



Figs. 31–34. *Lampteromyces luminescens*: basidiomata (31), basidiospores (32), basidium (33), cheilocystidium (34); after Zang (1979, 1984).

Lampteromyces mangensis differs from *L. japonicus* mainly in its more or less whitish colours. The description of this species sounds very similar to Corner's diagnoses of the Malaysian *Pleurotus olivascens* Corner (Corner, 1981), a species already discussed to be included in *Lampteromyces* by Singer (1986). No authentic material of *L. mangensis* and *P. olivascens* could be studied by us.

THIN LAYER CHROMATOGRAPHY OF PIGMENTS

Pigments from dried material

With the exception of *O. mexicanus* a very similar pigment pattern could be observed for all species (Fig. 35, Table IV). All species were characterized by the presence of atromentin and thelephoric acid although these pigments were produced in quite different quantities: In the two varieties of *O. olivascens* very large amounts of atromentin could be detected whereas in *O. olearius* and *O. nidiformis* only traces of this pigment were found. Medium quantities of atromentin were produced by *O. illudens*, *O. mexicanus*, and *O. subilludens*. Thelephoric acid was abundant in all species except in *O. olearius* and *O. subilludens* where only small amounts were detectable.

Pigments from cultures

A survey of the known Boletales pigments in *Omphalotus* and *Lampteromyces* is given in Table IV. Atromentin and thelephoric acid were found in all culture extracts. Variegatic acid, although not detectable in dried material, was present in *Lampteromyces japonicus* and all *Omphalotus* strains. The red pigment variegatorubin, found in all culture extracts, might be an artefact caused by oxidation during extraction (Gill & Steglich, 1987). The presence of xeroconic acid could be shown in cultures of *O. illudens*, *O. mexicanus* and the two varieties of *O. olivascens*. The cyclopentanoid 'gyroporin' and the pulvinic acid derivate 'atromentinic acid', which had been found by Bresinsky & Besl (1979b) in *O. illudens*, *O. olearius* and *L. japonicus* were not clearly detectable in this study. Moreover, a

number of unidentified pigments was found in all strains but the individual patterns did not indicate substantial differences between species because the amounts of produced pigments varied strongly within different strains or collections of one species and, consequently, weak bands of pigments might have been masked (Fig. 36, Table V).

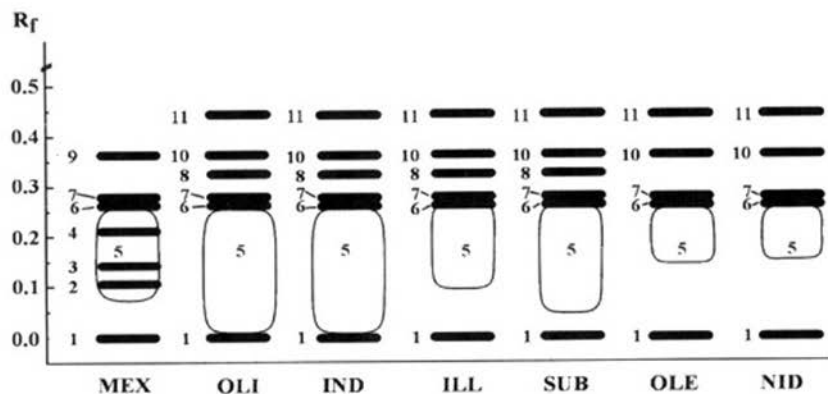


Fig. 35. Schematic representation compiling TLC pigment patterns from dried material. Acronyms: MEX = *O. mexicanus*; OLI = *O. olivascens* var. *olivascens*; IND = *O. olivascens* var. *indigo*; ILL = *O. illudens*; SUB = *O. subilludens*; OLE = *O. olearius*; NID = *O. nidiformis*. The different pigment bands (indicated by numbers) are characterised in Table V.

Table IV. Pigments in *Omphalotus* and *Lampteromyces* (C = from cultures; D = from dried material; F = from fresh basidiomata). Numerical indices refer to the initial detection of pigments: (1) Kämmerer et al., 1985; (2) Sullivan et al., 1971; (3) Sullivan & Guess, 1969; (4) Bresinsky & Besl, 1979b; (5) Singh & Anchel, 1971; (6) Bresinsky & Besl, 1979a.

	<i>O. illudens</i>	<i>O. olearius</i>	<i>O. olivascens</i> var. <i>olivascens</i>	<i>O. olivascens</i> var. <i>indigo</i>	<i>O. mexicanus</i>	<i>O. nidiformis</i>	<i>O. subilludens</i>	<i>L. japonicus</i>
Atromentin	C, D	C ⁽¹⁾ , D	C, D	C, D	C, D	C, D	C ⁽²⁾ , F ⁽³⁾	C ⁽¹⁾ , D
Atromentinic acid	C ⁽⁴⁾ , F ⁽⁵⁾	C ⁽⁴⁾	-	-	-	-	-	C ⁽¹⁾
Gyoporin	C ⁽⁶⁾	C ⁽⁴⁾	-	-	-	-	-	C ⁽¹⁾
Thelephoric acid	C, D, F ⁽⁶⁾	C, D	C, D	C, D	C, D	C, D	C ⁽²⁾ , D	C ⁽¹⁾
Variagatic acid	C	C ⁽⁴⁾	C	C	C	C	C	C
Variogatorubin	C	C	C	C	C	C	C	C
Xerocomic acid	C	C ⁽⁴⁾	C	C	C	-	-	-

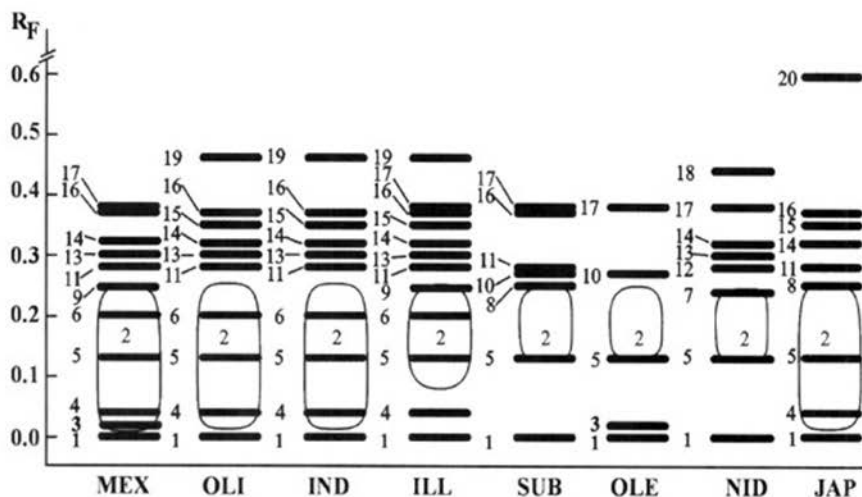


Fig. 36. Schematic representation compiling TLC pigment patterns from culture extracts. Acronyms: MEX = *O. mexicanus*; OLI = *O. olivascens* var. *olivascens*; IND = *O. olivascens* var. *indigo*; ILL = *O. illudens*; SUB = *O. subilludens*; OLE = *O. olearius*; NID = *O. nidiformis*; JAP = *Lampteromyces japonicus*. The different pigment bands (indicated by numbers) are characterised in Table VI.

Table V. Characterisation of the bands shown in Fig. 35 (pigment patterns from extracts of dried material).

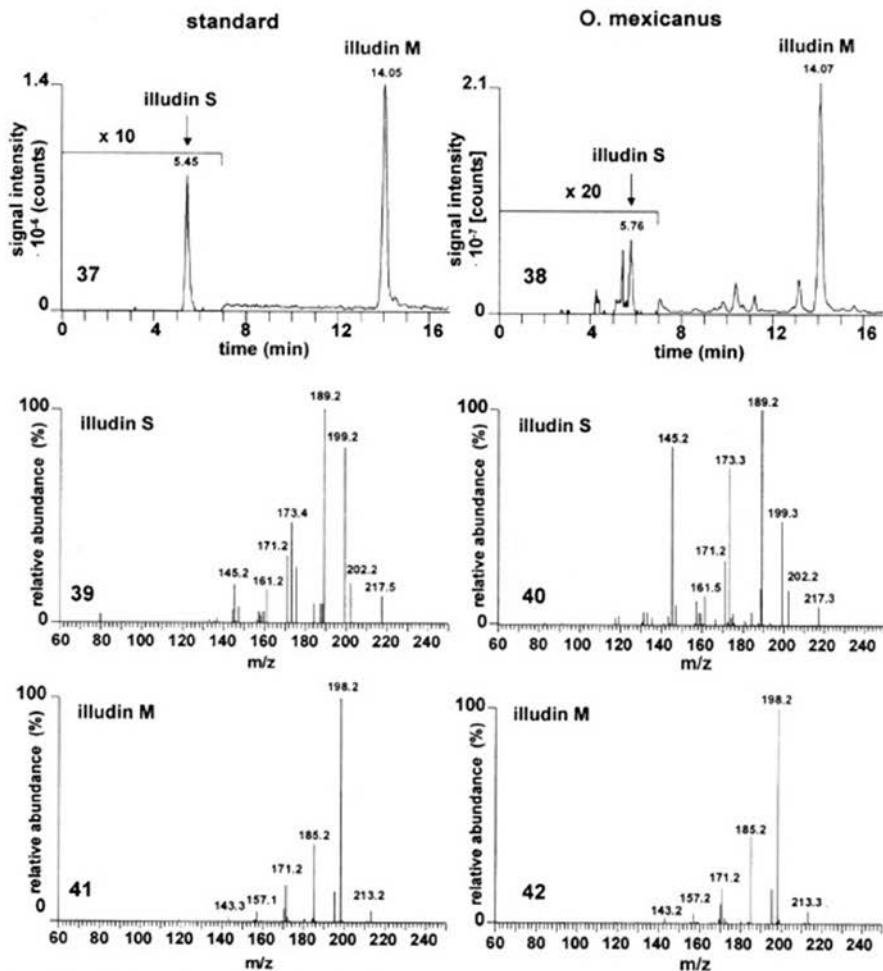
Nr.	Rf-value	daylight	UV 366 nm	Species
1	0	—	—	starting point
2	0.10	yellow	dark	<i>O. mexicanus</i>
3	0.15	turquoise	dark	<i>O. mexicanus</i>
4	0.22	red	yellow	<i>O. mexicanus</i>
5	0–0.25 (atromentin)	greenish	dark	<i>O. mexicanus</i> , <i>O. olivascens</i> var. <i>olivascens</i> , var. <i>indigo</i> , <i>O. olearius</i> , <i>O. nidiformis</i> , <i>O. illudens</i> , <i>O. subilludens</i>
6	0.27	yellow	yellow	<i>O. mexicanus</i> , <i>O. olivascens</i> var. <i>olivascens</i> , var. <i>indigo</i> , <i>O. olearius</i> , <i>O. nidiformis</i> , <i>O. illudens</i> , <i>O. subilludens</i>
7	0.28	orange	yellow	<i>O. mexicanus</i> , <i>O. olivascens</i> var. <i>olivascens</i> , var. <i>indigo</i> , <i>O. olearius</i> , <i>O. nidiformis</i> , <i>O. subilludens</i>
8	0.33	yellow	dark	<i>O. olivascens</i> var. <i>olivascens</i> , var. <i>indigo</i> , <i>O. illudens</i> , <i>O. subilludens</i>
9	0.37	turquoise	blue	<i>O. mexicanus</i>
10	0.37	yellow	blue	<i>O. olivascens</i> var. <i>olivascens</i> , var. <i>indigo</i> , <i>O. olearius</i> , <i>O. nidiformis</i> , <i>O. illudens</i> , <i>O. subilludens</i>
11	0.46	yellow	orange	<i>O. olivascens</i> var. <i>olivascens</i> , var. <i>indigo</i> , <i>O. olearius</i> , <i>O. nidiformis</i> , <i>O. illudens</i> , <i>O. subilludens</i>

Table VI. Characterisation of the bands shown in Fig. 36 (pigment patterns from culture extracts).

Nr.	R _f	daylight	UV 366 nm	Pigment	Species
1	0	—	—	—	Starting point
2	0	greenish	dark	atromentin	<i>O. olivascens</i> var. <i>olivascens</i> , var. <i>indigo</i> , <i>O. illudens</i> , <i>O. subilludens</i> , <i>O. olearius</i> , <i>O. nidiformis</i> , <i>O. mexicanus</i> , <i>L. japonicus</i>
	0.25	—	—	—	—
3	0.02	red	dark	unknown	<i>O. olearius</i> , <i>O. mexicanus</i>
4	0.04	yellow	dark	unknown	<i>O. olivascens</i> var. <i>olivascens</i> , var. <i>indigo</i> , <i>O. illudens</i> , <i>O. mexicanus</i> , <i>L. japonicus</i>
5	0.13	yellow	dull yellow	variegatic acid	<i>O. olivascens</i> var. <i>olivascens</i> , var. <i>indigo</i> , <i>O. illudens</i> , <i>O. subilludens</i> , <i>O. olearius</i> , <i>O. nidiformis</i> , <i>O. mexicanus</i> , <i>L. japonicus</i>
6	0.20	yellow	yellow	xeroconic acid	<i>O. olivascens</i> var. <i>olivascens</i> , var. <i>indigo</i> , <i>O. illudens</i> , <i>O. mexicanus</i>
7	0.24	yellow	yellow	unknown	<i>O. nidiformis</i>
8	0.25	red	dark	unknown	<i>O. subilludens</i> , <i>L. japonicus</i>
9	0.26	yellow	dark	gyroporin?	<i>O. illudens</i> , <i>O. mexicanus</i>
10	0.27	blue	dark	unknown	<i>O. subilludens</i> , <i>O. olearius</i>
11	0.28	yellow	dark	unknown	<i>O. olivascens</i> var. <i>olivascens</i> , var. <i>indigo</i> , <i>O. illudens</i> , <i>O. subilludens</i> , <i>O. mexicanus</i> , <i>L. japonicus</i>
12	0.28	red	orange	unknown	<i>O. nidiformis</i>
13	0.30	yellow	yellow	unknown	<i>O. olivascens</i> var. <i>olivascens</i> , var. <i>indigo</i> , <i>O. illudens</i> , <i>O. nidiformis</i> , <i>O. mexicanus</i>
14	0.32	red	dark rubin	variegato-	<i>O. olivascens</i> var. <i>olivascens</i> , var. <i>indigo</i> , <i>O. illudens</i> , <i>O. nidiformis</i> , <i>O. mexicanus</i> , <i>L. japonicus</i>
15	0.35	yellow	dark	unknown	<i>O. olivascens</i> var. <i>olivascens</i> , var. <i>indigo</i> , <i>O. illudens</i> , <i>L. japonicus</i>
16	0.37	red	dark	unknown	<i>O. olivascens</i> var. <i>olivascens</i> , var. <i>indigo</i> , <i>O. illudens</i> , <i>O. subilludens</i> , <i>O. mexicanus</i> , <i>L. japonicus</i>
17	0.38	yellow	dark	unknown	<i>O. illudens</i> , <i>O. subilludens</i> , <i>O. olearius</i> , <i>O. nidi-</i> <i>formis</i> , <i>O. mexicanus</i>
18	0.44	yellow	dark	unknown	<i>O. nidiformis</i>
19	0.46	yellow	dark	unknown	<i>O. olivascens</i> var. <i>olivascens</i> , var. <i>indigo</i> , <i>O. illudens</i>
20	0.59	yellow	dark	unknown	<i>L. japonicus</i>

DETECTION OF ILLUDIN S AND ILLUDIN M

Earlier studies reported the occurrence of the sesquiterpenes illudin S and illudin M in *Lampteromyces japonicus* (Nakanishi et al., 1965) and in many *Omphalotus* species (Anchel et al., 1952; Nair et al., 1983; Kirchmair et al., 1999). In this study *O. mexicanus* was shown to produce both illudin S and illudin M. The chromatographic retention times of illudin S and illudin M were obtained by injection of reference solutions in acetonitrile. Using a 150



Figs. 37–42. Detection and identification of illudins in an ethyl acetate extract of *O. mexicana* by high-performance liquid chromatography-mass spectrometry. For chromatographic and mass spectrometric conditions refer to method section; 37, 38. reconstructed ion chromatograms detecting the transitions 247.1→217.1→full scan from 60–250 u and 231.1→213.2→full scan from 60–250 u, specific for illudin S and M; 39 & 40. MS³ spectra of the illudin S reference standard and illudin S in *O. mexicana*, respectively; 41, 42. MS³ spectra of the illudin M reference standard and illudin M in *O. mexicana*, respectively.

× 2 mm I.D. column packed with a Nucleosil ODS stationary phase and applying a gradient of 10–42% acetonitrile in 0.1% aqueous acetic acid in 20 min, illudin S eluted first at 5.4 min and illudin M at 14.1 min (Figs. 37, 38). Analysis of the MS³ spectra under the peaks at 5.4 min and 14.1 min of the *O. mexicana* extract revealed fragment ions characteristic for illudin S and illudin M, respectively (Figs. 39–42). The high specificity of chromatographic separation in combination with MS³ and the congruence of the obtained mass spectra

with those of the reference standards prove the presence of both illudin S and illudin M in *O. mexicanus*.

DISCUSSION

Dried material is useful for morphological research but many characteristic pigments are rapidly degraded. Therefore, herbarium specimens may be useful for chemotaxonomical fingerprints but not for the identification of group characteristic metabolites. For extensive comparative chemotaxonomical studies fresh basidiomata are usually not available. Thus, culture extracts provide a suitable alternative. But it should be mentioned that the pigment pattern in native basidiomata need not be identical to that in mycelium cultures (Bresinsky, 1974).

In all *Omphalotus* and *Lampteromyces* species typical Boletales pigments of the pulvinic acid type as well as sesquiterpenes of the illudane type were found. As already discussed by Kämmerer et al. (1985) the combination of these two features is highly characteristic for *Omphalotus* and *Lampteromyces* and cannot be found in any other mushroom group. Because of the uniqueness of the metabolites illudin S and illudin M Nair et al. (1983) proposed the placement of *Lampteromyces* in *Omphalotus*. Kämmerer et al. (1985) stressed that in contrast to the more irregular hymenophoral trama in *Lampteromyces*, *Omphalotus* has a distinctly divergent gill anatomy. In the present study a distinctly divergent tramal structure could not be observed in *Omphalotus* (see also Singer, 1986). Thus, the large spores and the occurrence of a veil in *Lampteromyces* remain as reliable distinguishing characters. In other genera, e.g. in *Armillaria* or in *Pleurotus*, species with and without veil have been described. Moreover, sequence analyses of the ribosomal ITS1-5.8S-ITS2 region demonstrated that *L. japonicus* clustered within the clade of *Omphalotus* and was not assigned to a separate clade (Stolz, 1999). So it seems logical to regard *Lampteromyces* as a synonym of *Omphalotus*. The following new combinations are necessary:

Omphalotus japonicus* (Kawam.) Kirchm. & O.K. Mill., *comb. nov.

Basionym: *Pleurotus japonicus* S. Kawamura, Journ. Coll. Sci. Imp. Univ. Tokyo 35 (3) (1915) 2.

Omphalotus mangensis* (J. Li & X. Hu) Kirchm. & O.K. Mill., *comb. nov.

Basionym: *Lampteromyces mangensis* J. Li & X. Hu, Acta Sci. Nat. Univ. Norm. Hunan 16 (1993) 188.

Lampteromyces luminescens M. Zang differs in macro- and microscopical characters from *Omphalotus* species. Luminosity of the basidiomata is the only character it shares with *Omphalotus* species. Its rough spores, fusiform cheilocystidia and the serrate gill edges make an affiliation to *Lentinellus* probable.

In literature, the delimitation of the different *Omphalotus* species is controversial. *Omphalotus mexicanus* and *O. nidiformis*, for example, can be accepted as good morpho-species on account of their unique characters (Table II): the entire basidioma of the former is bluish black, and terminal cells covered with violet crystal needles can be found in its hymenophoral trama. White lamellae and a white context are typical for *O. nidiformis* (Miller, 1994), features it shares with *O. japonicus* and *O. mangensis*. However, the latter two species are characterized by an annular zone at the stipe apex and much bigger spores. The opinion that *O. mexicanus* and *O. nidiformis* are separate species is supported by the

findings of Petersen & Hughes (1998) who reported only a low sexual compatibility of *O. nidiformis* with other *Omphalotus* species. The results of their RFLP analyses of the ribosomal ITS1-5.8S-ITS2 region confirmed the relatively isolated position of these two taxa (Hughes & Petersen, 1998).

There has long been disagreement whether or not the American *O. illudens* and the European *O. olearius* are two different species. Mating experiments affirmed that *O. illudens* ranks as species from North America (Petersen & Hughes, 1998). But it remains still unclear if beside *O. olearius* also *O. illudens* exists in Europe since there are only few morphological separating characters: the colour of *O. illudens* is paler (uniformly yellowish orange), the pileus is umbonate and refractive hyphae are abundant in the pileipellis. Using these criteria it seems that the collections from Belgium belong to *O. illudens* while all collections from the mediterranean region can be recognised as *O. olearius*.

Omphalotus subilludens is morphological very similar to *O. olearius* but differs in its elliptic spores and numerous refractive hyphae in the pileipellis. *Omphalotus olivascens* is distinguished from *O. olearius* mainly by its more olive colours (the context is blue in its variety *indigo*), the abundant refractive hyphae, and significantly bigger spores. The olive and blue colours are due to the very high amount of the pigment atromentin (Kirchmair & Griesser, 1999). Petersen & Hughes (1998) noticed a very high level of sexual intercompatibility between collections of the three taxa mentioned before. Moreover, Hughes & Petersen (1998) found no differences in the RFLP patterns between *O. olearius* and *O. subilludens*. *Omphalotus olivascens* differed in one restriction site from the two former taxa. The findings of these authors indicate that these three taxa belong to one biological species. Nevertheless, since they are geographically well isolated and differ at least in two morphological characters, they can be considered as different morphospecies in agreement with a morphological species concept (see Kuyper, 1988).

KEY TO THE SPECIES OF OMPHALOTUS

- | | |
|---|---|
| 1a. Basidioma annulate | 2 |
| b. Basidioma not annulate | 3 |
| 2a. Entire basidioma more or less white | <i>O. mangensis</i> |
| b. Basidioma brownish | <i>O. japonicus</i> |
| 3a. Context in pileus and stipe ± white | <i>O. nidiformis</i> |
| b. Context coloured | 4 |
| 4a. Spores elliptical (quotient = 1.5 ± 0.2) | <i>O. subilludens</i> |
| b. Spores spherical (quotient = 1.1 ± 0.1) | 5 |
| 5a. Basidioma somewhere with olive tinges | 6 |
| b. Basidioma without olive tinges | 7 |
| 6a. Context olivaceous, not blue | <i>O. olivascens</i> var. <i>olivascens</i> |
| b. Context distinctly blue, bluish grey | <i>O. olivascens</i> var. <i>indigo</i> |
| 7a. Entire basidioma dark blue to bluish black | <i>O. mexicanus</i> |
| b. Basidioma not so | 8 |
| 8a. Basidioma uniformly yellow orange, pileus umbonate, pileipellis with numerous refractive hyphae | <i>O. illudens</i> |
| b. Pileus usually ± orange brown, darker than the lamellae, infundibuliform, refractive hyphae usually rare | <i>O. olearius</i> |

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CERATOBASIDIUM ALBASITENSIS

A new *Rhizoctonia*-like fungus isolated in Spain

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Ceratobasidium albasitensis is described as a new species based on morphological data and on phylogenetic reconstruction from rDNA sequences, including representatives of other related species. Isolates of this species were found in several localities within Albacete province (SE of Spain). This taxon is part of the *Rhizoctonia* s.l. species complex and is placed in the genus *Ceratobasidium* (Stat. Anam. = *Ceratorhiza*) on account of its macro- and micromorphological features. In addition, two new methods to induce sexual sporulation on plates are described.

The form-genus *Rhizoctonia* is currently considered to be a heterogeneous assemblage of fungal taxa, which do not form asexual spores, but have certain significant morphological characteristics in common. The *Rhizoctonia* complex is now split into at least seven or eight genera, according to different authors (Moore, 1987; Andersen, 1996). *Rhizoctonia* s.l. has considerable ecological and economic importance because it occurs worldwide and different isolates within the complex may live as saprotrophs or as symbionts (such as those associated with terrestrial orchids). Many isolates are also effective in protecting plants from a number of fungal diseases (Sneh, 1998), promoting plant growth. Conversely, many other isolates cause significant losses in agriculture and forestry; currently *Rhizoctonia* diseases have been described in more than 200 plant species. At least 120 epithets referring to *Rhizoctonia* can be found in the literature, where until recently very few attempts to clarify genera and species concepts had been made (Sneh et al., 1991; Andersen & Stalpers, 1994; Roberts, 1999).

Teleomorphs of fungi that show the morphological characteristics of *Rhizoctonia* species with binucleate hyphal compartments have usually been reported as belonging to *Ceratobasidium* D.P. Rogers, with the exception of *Epulorhiza repens* (N. Bernard) Moore (= *Rhizoctonia repens* Bernard), which has been found by some authors to have a perfect state in *Tulasnella* Schroet. (*T. deliquescens* (Juel) Juel = *T. calospora* (Boud.) Juel s. auct.) (Sneh et al., 1991). *Ceratobasidium* is regarded as close to *Thanatephorus* Donk, being considered by some authors (e.g. Stalpers & Andersen, 1996; Roberts, 1999) as part of a generic complex, where delimitation presents some difficulties. Anamorphic *Rhizoctonia*-like fungi from these two genera have traditionally been classified on the basis of the number of nuclei per hyphal compartment, considering *Thanatephorus* as multinucleate and *Ceratobasidium* as binucleate, although the nuclear condition remains unknown for many taxa. Another important handicap to understanding the taxonomy of these fungi is the fact that most studies on *Rhizoctonia* s.l. are performed with cultures where sexual reproductive structures are

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usually not or hardly observed, so that delimitation of species is based on the morphology and physiological features of the asexual morphs. Because of this, the development of new methods for inducing teleomorph formation constitutes a valuable tool to clarifying and correlating *Rhizoctonia* taxonomy.

Some non-pathogenic or hypovirulent isolates within the form-genus *Rhizoctonia* have been shown to be highly effective biocontrol agents (Sneh, 1998). In the majority of studies to date, most of the known isolates capable of protecting plants and preventing diseases have been found to belong to *Ceratobasidium*. In preliminary studies (data not shown), isolates of the new species described here have proved to possess protective properties in several plant species against pathogenic isolates of *Rhizoctonia solani* Kühner (= *Thanatephorus cucumeris* (A.B. Frank) Donk), and other fungal pathogens such as *Fusarium* spp., *Alternaria* spp., *Penicillium* spp., etc.

As stated by some authors (e.g. Mordue et al., 1989; Andersen, 1996), the employment of integrated approaches to solve taxonomic problems in *Rhizoctonia* (including morphometrical, cultural, biochemical, ecological and molecular data) could lead to establishing more accurate and natural classifications within this group of organisms. In the present work, molecular phylogeny of binucleate *Rhizoctonia* isolates was undertaken based on sequence data from the ribosomal ITS region. The isolates included those from Albacete, several testers used for defining anastomosis groups within binucleate *Rhizoctonia*, and miscellaneous sequences of *Ceratobasidium* retrieved from GenBank. The results provided additional evidence (at sequence level) for validating the new taxon presently described.

METHODS

Ceratobasidium albasitensis isolates

Root samples of saffron corms and pine seedlings were collected from six locations in Albacete. *Ceratobasidium* isolates were collected by washing roots with tap water to remove adhering soil particles and placing both root segments and rhizobial soil particles on 1.5% water agar amended with chloramphenicol (250 mg/ml). Some of the resulting colonies were identified as belonging to *Ceratohiza* s. Moore (1987) (= binucleate *Rhizoctonia*) by observing typical morphological features such as hyphal branching, number of nuclei, colour of the cultures, sclerotia formation or dolipore septa. Cultures were then transferred and maintained on potato-dextrose-agar (PDA) at room temperature. The number of nuclei per cell was determined using a staining method as described by Julián et al. (1997).

Formation of perfect state

Two undescribed methods were used to induce formation of basidiocarps by the isolates. The first method consisted of a modification of the one described by Hietala et al. (1994). In this first procedure, sterile radish seedlings (ethanol 70% 2'; sodium hypochloride 5% with Tween 0.005% 15' and at least five washes in distilled water), were pregerminated by incubating them on 15% water-agar plates at 24°C in the dark. After two days, the seedlings were transferred to Petri-dishes containing 20 ml sterile water with two 5 × 5 mm blocks of a 7 days old colony of the fungus, and incubated at room temperature with natural lighting. Teleomorph production started between 3 and 10 days after fungal inoculation.

A second new method was developed in our laboratory, consisting of a modification of some of the previously described methods (i.e. Flentje, 1956; Murray, 1982, 1984) based

on transferring isolates from high nutrient agar to low nutrient agar. Briefly, basidiomes were obtained by growing cultures of the isolates on PDA (potato-dextrose-agar) for 5–6 days and then transferred to plates of 15% water-agar containing small pieces (two blocks of 10 cm² approx. per plate) of leaves or twigs from several plant species such as *Nerium oleander* L., *Prunus laurocerasus* L., *Ligustrum vulgare* L., *Pinus halepensis* Miller, etc., previously surface sterilized as described above. Hymenial production started (depending upon the strains) between 7 and 15 days after the transfer of the colonies.

Phylogenetic reconstruction

For phylogenetic analysis, the entire sequence of the ITS regions of 37 binucleate *Rhizoctonia* isolates, representing several *Ceratobasidium* teleomorphs, was determined. The sequences were aligned and processed for phylogenetic reconstruction. All the isolates used in this work are listed in Table I, including the isolates of *C. albasitensis*, all available AG testers of binucleate *Rhizoctonia* described up to date and some *C. cornigerum* and *C. cerealis* isolates from the CBS collection (Baarn, The Netherlands). In addition, several ITS sequences including *C. oryzae-sativae*, *C. cerealis*, *Waitea circinata* and *Serendipita vermifera* s. Roberts (1999) (= *Sebacina vermifera*) were retrieved from GenBank. DNA isolation, PCR and DNA sequencing procedures used have been previously described (Boysen et al., 1996). Alignments of the ITS regions were performed using the multiple alignment program CLUSTALW (Thompson et al., 1994). Phylogenetic analysis of the aligned sequences was performed using maximum-parsimony with the heuristic-search algorithm of the Phylogenetic Analysis Using Parsimony (PAUP) program 3.1.1 (Swofford, 1993) with gaps treated as missing data. The trees were rooted with *Agaricus bisporus* as an outgroup. The data were resampled with 1000 bootstrap replicates (Felsenstein, 1985) by using the heuristic search option of PAUP. The percentage of bootstrap replicates that yielded each grouping was used as a measure of statistical confidence. A grouping found in 90% of bootstrap replicates was considered statistically significant. Similar trees were also obtained from the distance matrix of Jukes and Cantor using the neighbour-joining method of the program PHYLIP 3.5 (Felsenstein, 1993).

Ceratobasidium albasitensis V. González & V. Rubio, spec. nov. — Fig. 1

Basidiocarpus albidus, resupinatus, sparsus, inconspicuus, tenuis, pelliculosus vel pulveraceus quando recens et in aridis. Hyphae subcylindricae hyalinae, laxae, partim incrassatae tunicatae, ramis angulis rectis, fibulatae destitutae, 3.5–5.5(–6.5) µm latae. Basidia subglobosa vel sphaericopedunculata, producta aut singula aut quasi racemis aggregata ex hyphis subcylindricis, (16.2–)18.2–20.8(–24) × 7–11 µm, 4(–5) sterigmata, longissima, cornuta, subcurvata, saepe cum septis adventitiis prope apices (19–)22–35.7 × 1.9–3 µm. Sporae subglobosae vel latae ellipsoideae (Q = 1.2–1.4), laeves, hyalinae, inamilioideae, raro iterativae prope laterales, 5–7(–8) × (3.5–)4–5.5(–6) µm. Hyphae moliniformiae amplae, tumoribus, 13–15 µm latae. Status anamorphosis *Ceratohiza*.

In terra ad *Crocus sativus* L. et *Pinus halepensis* Miller.

Holotypus: Hispania, Albacete, Tobarra, 7 Nov. 1996, O. Salazar & M.C. Julián, in herb. Alcalá (AH 26603) conservatur.

Etymology: referring to the geographical origin of the isolates.

Among the nine isolates identified as a new species, three of them fructified repeatedly with both of the two methods used for teleomorph induction. Thus descriptions of both macro- and microscopic characters from the sexual stage are based on fructifications from these three isolates.

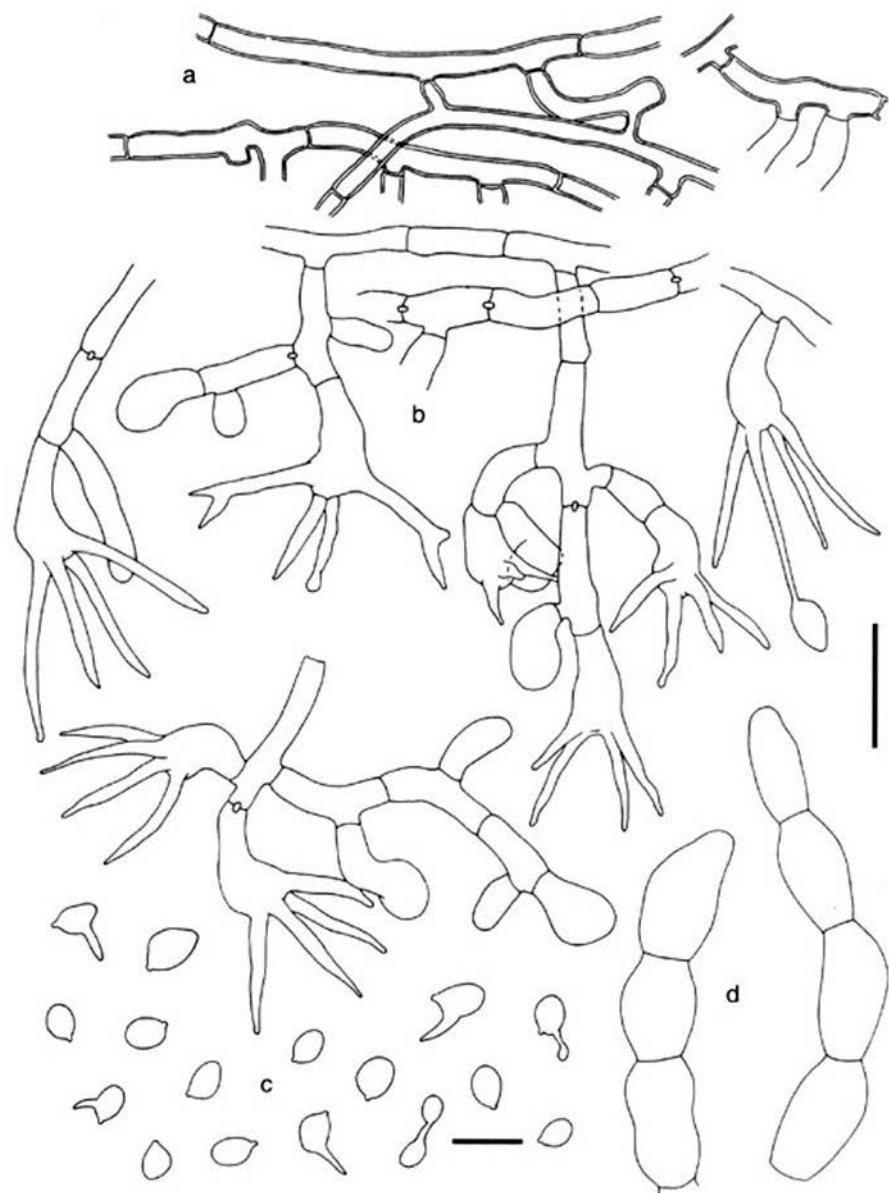


Fig. 1. *Ceratobasidium albasitensis*. a. Subicular hyphae; b. hymenial layer (basidia and basidioles); c. spores; d. molinioid cells. Bars: 10 & 20 μm respectively.

Table I. Isolates used in this study; host, size of ITS1, ITS2, total rDNA-ITS region, and EMBL or GenBank accession numbers. a) Isolates of *C. albasitensis*; b) tester isolates of binucleate *Rhizoctonia* (*Ceratobasidium* spp.); c) other isolates.

a) Isolate	Isolated from	ITS1	ITS2	Total	Accession No.	
Eab-F1	<i>Crocus sativus</i>	184	231	570	AJ242875	
Eab-F2	<i>Crocus sativus</i>	185	230	570	AJ242876	
Eab-F3	<i>Crocus sativus</i>	184	230	569	AJ242877	
Eab-F5	<i>Crocus sativus</i>	184	230	569	AJ242878	
Eab-F6	<i>Crocus sativus</i>	184	230	569	AJ242879	
Eab-F7	<i>Crocus sativus</i>	184	232	571	AJ242880	
Eab-S1	<i>Crocus sativus</i>	185	231	571	AJ242881	
Eab-S5	<i>Crocus sativus</i>	184	230	569	AJ242885	
Eab-S6	<i>Pinus halepensis</i>	184	230	569	AJ242886	
b) Isolate	AG	Isolated from	ITS1	ITS2	Total	Accession No.
C-662	AG-A	Soil	188	223	566	AJ242890
BN4	AG-A (CAG2)	Soil	188	223	566	AJ242891
SIR-2	AG-Bo	<i>Ipomoea batatas</i>	185	230	570	AJ242892
C-455	AG-Bb	<i>Oryza sativa</i>	185	230	570	AJ242893
70B	AG-C	Soil	189	223	567	AJ242894
C-610	AG-D	unknown	189	223	567	AJ242895
Rh155	AG-E	unknown	190	223	568	AJ242896
C-653	AG-G	unknown	183	232	570	AJ242897
AV-2	AG-I	<i>Artemisia</i> sp.	183	229	567	AJ242898
184	AG-J	unknown	183	229	567	AJ242899
FA59209	AG-K	unknown	193	237	585	AJ242900
76146	AG-L	unknown	190	222	567	AJ242901
TC1	AG-N	unknown	186	230	571	AJ242902
76150	AG-P	unknown	187	230	572	AJ242903
c) Isolate	Isolated from	ITS1	ITS2	Total	Accession No.	
<i>R. cerealis</i>	<i>Poa annua</i>	212	231	598	AF063019	
C. o-s 2	<i>Oryza sativa</i>	226	249	630	AJ000192	
C o-s 1	<i>Oryza sativa</i>	227	249	631	AJ000191	
C o-s 3	<i>Oryza sativa</i>	226	249	630	AJ000193	
C o-s 4	<i>Oryza sativa</i>	226	248	631	AJ000194	
Rh2815	<i>Vicia faba</i>	188	228	571	U19962	
521	soil	190	230	575	U19950	
C1 (CAG-1)	<i>Festuca</i>	198	241	594	AJ301903	
C2 (CAG-2)	<i>Pittosporum</i>	198	238	591	AJ301899	
C4 (CAG-3)	<i>Juniperus</i>	191	243	589	AJ301900	
C5 (CAG-5)	<i>Taxus</i>	190	240	585	AJ301901	
C6 (CAG-6)	<i>Erigeron canadensis</i>	185	236	576	AJ301902	
C8 (CAG-7)	<i>Pittosporum</i>	190	245	590	AJ302006	
C11	<i>Triticum aestivum</i>	210	231	596	AJ302007	
C13	<i>Triticum aestivum</i>	210	231	596	AJ302009	
C12	<i>Triticum aestivum</i>	210	231	596	AJ302008	
Eab-aB	<i>Medicago sativa</i>	228	242	625	AJ302010	
Eab-S3	<i>Crocus sativus</i>	185	233	573	AJ242883	
<i>W. circinata</i> I	<i>Oryza sativa</i>	213	198	566	AJ000196	
<i>W. circinata</i> II	<i>Oryza sativa</i>	212	198	565	AJ000195	
<i>A. bisporus</i>	unknown	294	207	656	AJ301619	
<i>S. vermifera</i>	<i>Orchidaceae</i>	171	199	525	AF202728	

Basidiomata whitish to almost hyaline, resupinate, thin, hymenium consisting of a hypochnoid, pruinose, pellicular layer over the water surface (when fruiting in sterile water with radish seedlings) or on the outer surface of the agar, near the margins of the Petri-dish, more rarely covering the whole plate surface and even occasionally colonizing vegetable debris added to the medium (i. e. pine twigs, *Nerium* leaves, etc).

Basidia globose when young, then pyriform to sphaeropedunculate (16.2–)18.2–20.8(–24) × 7–11 µm, produced directly from basal hyphae or in raceme-like groups. Sterigmata 4 or 5, very long (more than twice the length of the basidium), (19–)22–35.7 × 1.9–3 µm, cornute, curved or straight, easily collapsing and often with adventitious septa occurring near the tips. Basidiospores broadly ellipsoid to ovoid (Q = 1.2–1.4, average 1.26) in front view, amygdaliform to slightly citriform in side view, 5–7(–8) × (3.5–)4–5.5(–6) µm (average 6.42 × 4.19 µm) (n = 21), apiculate, hyaline, smooth, not amyloid, germinating by a lateral germ-tube and more rarely by repetition. Subicular hyphae hyaline, thick to thin-walled, 3.5–5.5(–6.5) µm in diam. Moniloid hyphae occurring among the hymenial tissues, consisting of inflated, barrel-shaped elements, up to 15 µm in diam. Clamp-connections absent in all tissues.

Habitat — Saprotrophic on rhizospherical soil and healthy saffron corms (*Crocus sativus* L.) and pine seedlings (*Pinus halepensis* Miller) in agricultural ground.

Material studied. SPAIN: Albacete, Tobarra, 28 Nov. 1996, in agricultural ground with *Crocus sativus* L., leg. O. Salazar & M.C. Julián (holotype, AH 26603); ibidem, 7 July 1996, AH 26605; Albacete, Aguas Nuevas, 7 July 1996, leg. O. Salazar & M.C. Julián, AH 26604.

A system of anastomosis grouping based on hyphal fusion is widely accepted as the basis for recognizing groups among the several taxa that constitute the form-genus *Rhizoctonia* (Ogoshi, 1975; Sneh et al. 1991). These methods have been extensively employed for both multinucleate and binucleate *Rhizoctonia* isolates, instead of taxonomical approaches dealing with teleomorph-based systems. Several authors have pointed out the difficulty in differentiating species within *Ceratobasidium* (Ogoshi et al., 1983). Although there are some reports in the literature of well-defined taxa based on the morphology of the teleomorphic phase [i. e. *C. oryzae-sativae* Gunnell & R.K. Webster (1987); *C. ramicola* Tu, Roberts & Kimbrough (1969)], many of the teleomorphs from the different anastomosis groups recognized within 'binucleate *Rhizoctonia*' (AG-E, AG-L, AG-I, AG-K, etc) obtained in the laboratory by indirect methods, although assigned to *Ceratobasidium*, are generally not defined at species level (Sneh et al., 1991).

Thanatephorus, the teleomorphic stage of multinucleate *Rhizoctonia* (e.g. *R. solani*) is the closest genus to *Ceratobasidium*. The systematic position of both genera within the Basidiomycetes has been previously studied (Talbot, 1965; Eriksson & Ryvardeen, 1973; Stalpers & Andersen, 1996; etc.) and the relationships of the *Ceratobasidiaceae* with the Heterobasidiomycetes (mainly *Tulasnellaceae* and *Tremellaceae*) profusely discussed. The systematic arrangement still remains unclear, although they are presently placed among the *Ceratobasidiaceae* (Ceratobasidiales, Basidiomycetes) within the Homobasidiomycetes (Hawksworth et al., 1995). Recently, Roberts (1999) suggested a rearrangement of the classification proposed in Hawksworth et al. (1995). He proposed simplifying the current nomenclatural status of genera within the *Ceratobasidiaceae* by recognizing just three genera within the family: *Ceratobasidium*, *Thanatephorus* and *Waitea*. As stated by several authors (e.g. Stalpers & Andersen, 1996; Roberts, 1999), the differences used to distinguish these genera are gradual, although it is commonly accepted that *Thanatephorus* is applied to those

mostly parasitic fungi with hypochnoid, sometimes gelatinized basidiomes with ellipsoid to barrel-shaped basidia formed from vertically branching, cymose hyphae (mostly containing more than two nuclei per compartment), while the name *Ceratobasidium* is applied to taxa with ceraceous fruitbodies with ovoid to sphaeropedunculate basidia arising in raceme-like groups from the subicular hyphae (generally with two nuclei per compartment). The convenience of accepting genus *Waitea* as a member of the *Ceratobasidiaceae* will be discussed below.

The sizes of the ITS region (ITS1, 5.8S and ITS2) of the different isolates are listed in Table I. The ITS1 region varied in length from 171 to 228 bp, the ITS2 region varied from 198 to 249 bp. It was regularly observed that the ITS1 region was slightly shorter than the ITS2 for all the isolates used in the study. The 5.8S region was highly conserved: all the isolates analyzed were 155 bp in length, only minor variations in nucleotide sequence were detected among isolates and this variation was identical in isolates belonging to the same species. The percentage of similarity in the total ITS region among the main clusters recognized in the phylogenetic tree (labelled from G1 to G9) is summarized in Table II.

Phylogenetic trees based on ITS sequences were obtained with PAUP. PHYLIP analysis did not differ in the topology of the strongly supported branches. The parsimony analysis of these characters generated 100 equally parsimonious trees with the minimum length tree of 1008 steps, with consistency (CI) and retention (RI) indexes of 0.6577 and 0.8214 respectively. One of these most parsimonious trees is shown in Fig. 2. Parsimony analysis of the ITS region supports in general the morpho- and cytological criteria used for defining and delimitating species within *Ceratobasidium*.

With the exception of *Ceratobasidium cornigerum* (AG-Bo and AG-P), *C. cerealis* (AG-D) and *C. oryzae-sativae* (AG-Bb), when more than one isolate of the same species was sequenced, they always clustered together in the same branch, or at least in adjacent branches.

Phylogenetic analysis grouped all the binucleate taxa with a *Ceratobasidium* teleomorph in seven groups, most of them well supported by bootstrap indexes. These binucleate groups clustered apart from the isolates of *Waitea circinata* and *Serendipita vermifera*. Several authors have discussed the position of these last two taxa, usually considered within the concept of *Rhizoctonia* s.l. according to the morphological features exhibited by their anamorphic states. Moore (1978, 1987), employing electron microscopy to characterize the ultrastructure of the septal pore apparatus, designated those binucleate *Rhizoctonia* anamorphs with a perfect state in the genus *Tulasnella* as belonging to *Epulorhiza*, while Andersen and Moore (Moore, 1996) erected the epithet *Opadorhiza* to accommodate *Rhizoctonia globularis* Saksena & Vaartaja, a taxon with a teleomorph in *Sebacina* s.l., very similar to *Epulorhiza* but with a different septal pore apparatus.

On the basis of these studies, both genera must be considered as peripheral to *Rhizoctonia* in the modern concept of the complex. Nevertheless, Roberts (1999) included *Waitea* as a member of the *Ceratobasidiaceae*, considering the genus as very close to (if not synonymous with) *Thanatephorus* based on morphological criteria. Phylogenetic reconstruction carried out in the present study is not in concordance with this hypothesis, and suggests moving *Waitea* out of the *Ceratobasidiaceae*. Furthermore Johanson et al. (1998), in a study employing a PCR-based method to discriminate between the different taxa involved in the rice sheath-blight disease complex, showed that isolates from *Rhizoctonia oryzae* (teleomorph = *Waitea circinata*) clustered apart from those of *R. solani* and *R. oryzae-sativae*

Tree length = 1008
 CI = 0.6577
 RI = 0.8214
 RC = 0.5403

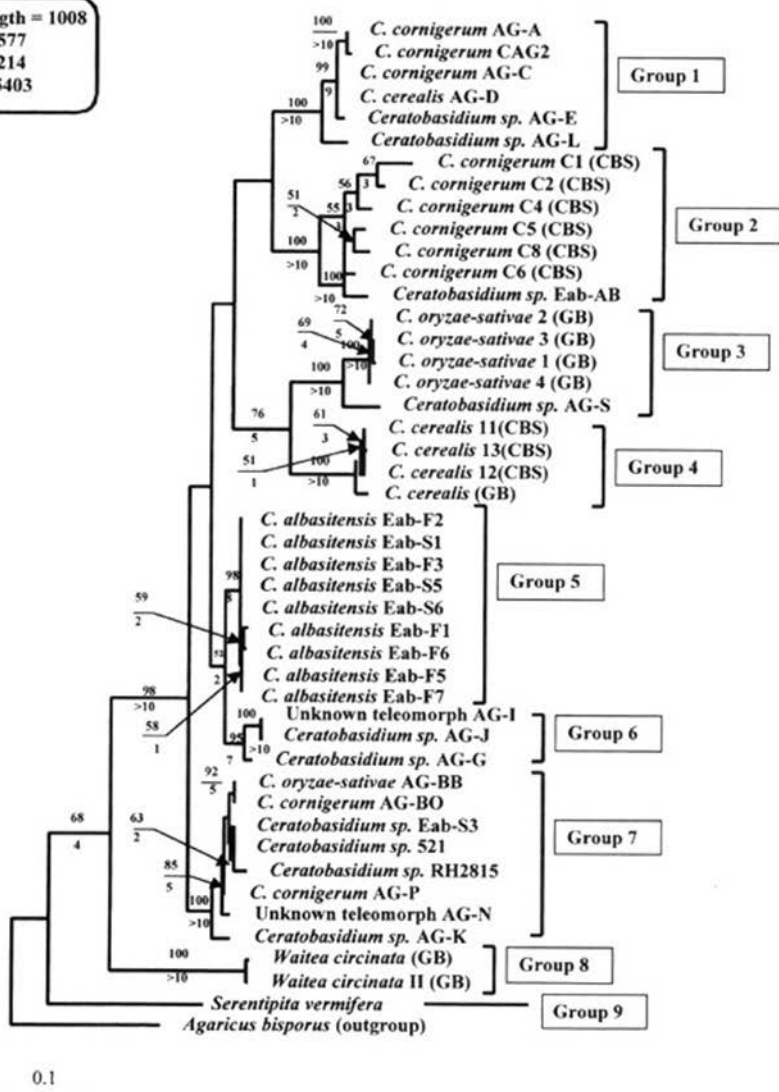


Fig. 2. One of the most parsimonious trees generated from a branch and bound algorithm in PAUP 3.1.1, using CLUSTALW for alignment. The trees were rooted with the sequence of one isolate of *Agaricus bisporus* (Agaricaceae). The numbers above branches indicate the bootstrap percentage from 1000 bootstrap replicates (for values greater than 50%). Decay indices up to 10 steps are indicated below branches. Horizontal lengths represent genetic distances.

Table II. Percentage of sequence divergence among the nine clusters recognized in the phylogenetic tree.

	G1	G2	G3	G4	G5	G6	G7	G8	G9
G1	0.00– 6.27	14.09– 17.84	13.03– 18.24	14.89– 17.38	10.84– 12.80	10.44– 13.02	10.48– 14.61	31.67– 33.48	40.64– 43.48
G2		3.15– 13.88	15.16– 22.80	15.75– 25.22	10.57– 14.38	12.07– 13.99	11.63– 15.94	34.07– 40.23	42.90– 44.56
G3			0.16– 8.46	16.46– 17.75	13.23– 15.85	11.80– 15.06	12.89– 17.82	36.19– 39.46	42.03– 43.61
G4				0.34– 1.53	13.84– 15.09	13.67– 15.43	14.78– 16.65	38.25– 39.19	43.01– 43.72
G5					0.00– 1.06	4.56– 6.30	6.27– 8.35	30.56– 31.52	40.32– 41.71
G6						0.18– 2.88	7.34– 10.02	29.83– 31.15	40.08– 40.32
G7							0.35– 4.37	29.61– 30.85	39.05– 41.83
G8								0.53	45.24– 45.37
G9									—

(teleomorph = *Ceratobasidium oryzae-sativae*). The systematic position of *Serendipita vermifera*, as revealed by molecular phylogeny, agrees with the conclusions previously reported by some authors (e.g. Roberts, 1999; Andersen, 1996; etc). The first author segregated some taxa from the genus *Sebacina* Tul. (*Tremellales* s. Hawksworth et al., 1995) into at least four genera: *Ceratosebacina* P. Roberts, *Endoperplexa* P. Roberts, *Serendipita* P. Roberts and *Hauerslevia* P. Roberts, all of them included by the author in the Exidiales. In addition, Andersen (1996) employed both ultrastructural and molecular methods (including RFLP analysis) to distinguish *Waitea* and *Sebacina* s.l. from other forms within *Rhizoctonia*.

Group 7 includes several AG testers of binucleate *Rhizoctonia* such as AG-Bo, AG-Bb, AG-P, AG-N and AG-K, one binucleate isolate from Spain previously identified as *Ceratobasidium* sp., together with the isolates 521 and 2815, and two *Rhizoctonia* isolates previously reported in the literature as *R. solani* AG-4 (Boysen et al. 1996). This cluster separates from the rest of the binucleate isolates (100% bootstrap value), forming a heterogeneous assemblage of taxa, including two strains of *C. cornigerum*, one of *C. oryzae-sativae*, and several AG tester isolates with insufficient or completely unknown sexual morphs. In spite of the nomenclatural heterogeneity, nucleotide divergence within this branch ranges from 0.35 to 4.37%, suggesting that the taxonomic status of some of these AG testers must be revised.

Group 6 includes three AG tester isolates (AG-I, AG-G and AG-J), all of them without any well-defined teleomorph. Recently, Masuhara et al. (1994) reported the teleomorphic features of some isolates of AG-I, and characterized them as probably belonging to the species complex *Ceratobasidium cornigerum*, although these authors reported two sexual morphs, *C. cornigerum* and *C. pseudocornigerum*, for several isolates. The other two strains, AG-G and AG-J are reported in the literature (Sneh et al., 1991) as belonging to *Ceratobasidium*, without defining any specific epithet.

Group 5 brings together all the strains determined as belonging to the new species proposed here (with a 98% bootstrap value). Isolates from this branch ranged between 0.00 and 1.06% in nucleotide divergence (the lowest values found in any of the 9 clades), suggesting that strains from clade 5, although isolated from different localities (and even from different hosts) could represent a well-defined and homogeneous taxon within *Ceratobasidium*.

Groups 4 and 3 branch together (supported by a 76% bootstrap value) and contain several *Ceratobasidium cerealis* isolates (Group 4) and a set of *C. oryzae-sativae* strains (Group 3) with one binucleate AG tester (AG-S) referred to in the literature as *Ceratobasidium* sp. Group 4 included sequences obtained from CBS isolates, together with one sequence obtained from the GenBank, and all of them showed a nucleotide divergence ranging from 0.34 to 1.53%, which suggests a very low intraspecific variation for this taxon. In addition, the other isolate (AG-D) with a sexual morph defined as *C. cerealis*, clustered in Group 1, together with several *C. cornigerum* AG testers. *Ceratobasidium gramineum* (= *C. cerealis*; *Corticium gramineum*) was proposed (Oniki et al., 1986) as the teleomorphic state for AG-D (CAG-1) strains. These authors obtained some sexual fructifications from AG-D strains and compared morphological measures of their hymenial components with those of *Corticium gramineum* Ikata & T. Matsuura and *Ceratobasidium cereale* Murray & Burpee, concluding that these three taxa must be considered conspecific. After critical revision of the dimensions reported for *Corticium gramineum* and *C. cerealis* in Oniki et al. (1986), these last two taxa could not be considered as synonyms. Thus, teleomorphs exhibited by AG-D strains can be assigned to the taxa described by Murray & Burpee (1984), but not to *Corticium gramineum*. On the other hand, dimensions reported for *C. cereale* by these last authors fit into the range of measures given by Rogers (1935) for *C. cornigerum*. Furthermore, a recent study (Toda et al., 1999) on several AG-D isolates from turfgrass, differentiate two subgroups (I and II) within the AG by means of rDNA and RAPD analyses. In summary, molecular and morphological data suggest the existence of two teleomorphic forms for this AG; one of them probably closely related to other AG groups (i.e. AG-C, AG-A, etc) with a *C. cornigerum* teleomorph, and other groups defined by possessing a *C. cerealis* teleomorph.

Group 3 includes four sequences of *C. oryzae-sativae* retrieved from GenBank and one AG-S tester isolate. The original host of this last strain is not mentioned in the literature and, although no teleomorphic state has been defined up to date for this tester, it could also represent a member of this taxon. Phylogenetic reconstruction suggests that this well-known rice pathogen could constitute a natural taxon, in spite of the nucleotide divergence observed for the group (ranging from 0.16 to 8.46%) and the heterogeneous geographical origin of the samples analyzed.

Group 2 includes some strains from the CBS culture collection (with a 100% bootstrap value), all of them deposited under the name of *Ceratobasidium cornigerum*, plus one isolate from Albacete (Spain), previously identified (using teleomorph induction methods) as

C. cornigerum s.l. Within this clade, nucleotide divergence ranges from 3.15 to 13.88%, indicating high rates of heterogeneity at sequence level. Furthermore, the CBS strains were deposited as representative isolates from several of the anastomosis groups defined for binucleate *Rhizoctonia* in America (Burpee et al., 1980a; 1980b). These molecular data suggest that American AGs represent a closely related pool of taxa where the different groups could constitute populations of the same collective taxon with different rates of somatic isolation.

Group 1 and Group 2 formed a common branch. Group 1 isolates are 5 Japanese AG testers (AG-A, AG-C, AG-D, AG-E and AG-L) and one isolate from CAG2. With the exception of AG-D (discussed above), teleomorphs indicated in the literature from members of this group are mostly *Ceratobasidium cornigerum* or unnamed *Ceratobasidium* spp. Several authors (e.g. Ogoshi et al., 1983) have proposed correlating most of the American binucleate AGs with the Japanese groups. The phylogenetic reconstruction carried out in this work suggests considering the different AGs described for binucleate *Rhizoctonia* (both American and Japanese isolates) as members of the same collective taxon, where relationships between the different groups of isolates (in terms of somatic compatibility) need to be revised, due to the fact that correlations based on hyphal anastomosis are not well reflected at sequence level. Cubeta et al. (1991), employing molecular methodologies (RFLP analysis) to characterize most of the AGs of binucleate *Rhizoctonia* species, were able to separate 13 of the 21 AGs defined for binucleate *Rhizoctonia*, although the relatedness and correlations between the American and Japanese groups were not exactly consistent with those proposed by Ogoshi et al. (1983).

With respect to the nutritional behaviour of the taxa included in the study, molecular data supports the differentiation of the several life styles within *Ceratobasidium*. Phylogenetic reconstruction suggests the existence of at least two monophyletic trends in the genus. One of them is linked to pathogenicity, and includes Group 1 and Group 2, where most of the isolates are reported in the literature as pathogenic to several plant species (Burpee et al., 1980b; Ogoshi et al., 1983; Toda et al., 1999; etc.), plus Groups 3 and 4, which include rice and grass pathogens. The other branches in the phylogenetic tree involve mostly saprotrophic taxa (Group 5), as well as other isolates not previously reported as pathogens (e.g. AG-I, AG-P, AG-N, etc.), or even described as plant protective isolates (e.g. AG-G) (Leclerc-Potvin et al., 1999).

In summary, phylogenetic analysis of the ITS region supported the definition of *Ceratobasidium albasitensis*, an undescribed binucleate taxon well differentiated from other taxa described in the genus by morphometrical methods (see discussion below). Furthermore, phylogenetic reconstruction allowed the outlining of some hypotheses on the relationships among the rest of the taxa within *Ceratobasidium*. Thus, *C. cornigerum*, the most common and widespread taxon of the genus, has been revealed in our studies as a large complex of taxa (including different species, varieties, ecotypes and populations), where a critical revision of the species concept, as well as a redefinition of the criteria for defining anastomosis groups is still required.

On account of the presence of two nuclei per hyphal compartment, saprotrophic behaviour, presence of a *Ceratorhiza* anamorph and the micromorphological features exhibited (including hymenial structure and basidial shape), we place the new species described in the genus *Ceratobasidium*. *Ceratobasidium albasitensis* is characterized by its soil habitat, large and sphaeropedunculate basidia, ovoid to subellipsoid spores and extremely large sterigmata (up to 35.7 µm), and clearly differs from other described *Ceratobasidium* species.

Table III. A microscopic comparison between *Ceratobasidium albasitensis*, *C. stridii*, *Thanatephorus obscurum* and *C. cornigerum*.

species	spores	Q	sterigmata	habitat
<i>C. albasitensis</i>	5-7(-8) × 4-5.5(-6)	1.2-4.0	-35.7 µm long	saprotrophic ¹
<i>C. stridii</i>	5.5-7.5 × 3-5	1.5-1.9	-8 µm	saprotrophic ²
<i>T. obscurum</i>	8-10 × 6-7	1.2-1.4	-1.2-1.4 µm	saprotrophic ³
<i>C. cornigerum</i>	(6.6-)7-11.5 × 3.5-6	1.9-2.0	-12 µm long	saprotrophic, parasitic and symbiotic ^{3,4}

1) This work; 2) Eriksson & Ryvardeen, 1973; 3) Roberts, 1999; 4) Rogers, 1935.

Ceratobasidium stridii J. Erikss. & Ryvardeen, a rare north temperate taxon characterized by having small basidiospores of 5.5-7 × 3-4 µm, resembles *C. albasitensis*, but, based on the literature available (Eriksson & Ryvardeen, 1973; Roberts, 1999), spores of *C. stridii* are oblong to fusiform (Q = 1.5-1.9). In addition, spores from *C. stridii* were reported to produce secondary spores mostly apically, a feature not constantly present in the genus and absent in the taxon presently described. The most common and ubiquitous taxon of the genus, *C. cornigerum* (Bourd.) D.P. Rogers, seems to be a complex of several different taxa, which some authors (e.g. Roberts, 1999) consider within a broad species concept. Nevertheless, the last author has pointed out a possible splitting of the complex among several of the existing synonyms on the basis of genetic distinctions. In this sense, DNA sequencing in the present study suggests separating taxa such as *C. cereale* from *C. cornigerum* s. str. Concerning the new species proposed here, *C. cornigerum* differs from *C. albasitensis* in having subceraceous basidiomes with smaller, laterally stalked cuboid to papillate basidia and larger, ellipsoid to fusiform, sometimes subcylindrical (Q = 1.4-2.0) spores up to 12.5 µm long and shorter sterigmata up to 12 µm long (Rogers, 1935; Eriksson & Ryvardeen, 1973; Jülich, 1989; Roberts, 1999). *Ceratobasidium obscurum* D.P. Rogers, a taxon misreported as an orchid associate (Warcup & Talbot, 1967) and presently combined as *Thanatephorus obscurum* (D.P. Rogers) P. Roberts (Roberts, 1998), has spores resembling those of *C. albasitensis*, being subglobose to broadly ellipsoid with Q = 1.2-1.4 (but not overlapping in size range). This taxon must be definitively considered as a *Thanatephorus* species as revealed by Roberts (1998) in a study of the type collection, where this author reports oblong basidia forming hymenial palisades and a saprotrophic habitat on rotten wood of *Ulmus* sp.

Morphological measures of *C. albasitensis* and some other *Ceratobasidium* species are summarized in Table III.

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**BOLBITIUS ELEGANS, A STRIKING NEW SPECIES FROM
SOUTHERN SPAIN**

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Bolbitius elegans spec. nov., a new species with gasteroid habit, is described and illustrated. Its taxonomic relationships with other taxa of *Bolbitius* and members of presumably related secotioid-gasteroid genera are discussed.

While studying the Basidiomycota of several habitats in southern Spain (Andalucía, provinces of Málaga, Huelva and Cádiz), the elegant basidiocarps of a bolbitiaceous agaric attracted our attention by their *Gastrocybe*- or *Galeropsis*-like habit. The weakly deliquescent basidiocarps with brilliant golden-yellow pileus and pale yellow stipe have exclusively been found on horse dung in open and grazed meadows on limestone.

The present taxon has previously been recorded as *Gastrocybe* sp. in the checklist referring to the Basidiomycota in or near the endemic *Abies pinsapo* forests in Andalucía (Ortega et al., 1996). On several later occasions, material in perfect condition was recollected which subsequently allowed a thorough analysis of the taxonomically relevant macro- and microscopic features of the ephemeral basidiocarps. Despite the fact that the basidiocarps are mimicking taxa of gasteroid *Bolbitiaceae*, it turned out that all morphological characters observed in the new taxon clearly indicate that this species belongs to the genus *Bolbitius* Fr. and not, as earlier suspected, to any of the associated secotioid-gasteroid genera (Heim, 1950, 1968; Watling & Gregory, 1981).

MATERIAL AND METHODS

SEM-photographs of the basidiospores, prepared by the critical point method, were taken with a Zeiss-DSM 950. For microscopic examination the material was mounted in KOH (4%), NH₄OH (2%) and Congo Red-solution.

The description of *Bolbitius elegans* is based upon data taken from collections found within close vicinity at three different localities. The holotype is deposited in the Herbarium AH (Alcalá, Madrid); isotypes and paratypes are lodged both in GDAC (Herbarium Granada, Spain) and ZT (Zurich, Switzerland).

Bolbitius elegans E. Horak, G. Moreno, A. Ortega & Esteve-Rav., *spec. nov.* —
Figs. 1–4

Pileus 10–35 × 4–12 mm, obtuse cylindrico, dein conicocampanulatus, secotioideus, luteus vel sulphureus, viscidus vel glutinosus, substriatus, fragilis, deliquescens. Lamellae liberae vel subattenuatae, densissimae, ochraceae, deliquescentes. Stipes 40–110 × 1–4 mm, cylindricus, stramineus, pruinosis.

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fistulosus, fragilis. Velum nullum. Caro fragilissima. Odor saporque nulli. Basidiosporae $12-15(-18) \times 6.5-9(-12) \mu\text{m}$, leves, ellipsoideae, poro conspicuo instructae, crassetunicatae. Basidia $23-30 \times 11-13 \mu\text{m}$, sphaeropedunculata, 4-sporigera. Cheilocystidia usque ad $45 \mu\text{m}$ diam., globosa vel ovoidea, conspicua, hyalina. Pleurocystidia nulla. Caulocystidia $70 \times 9-13 \mu\text{m}$, polymorphica. Pileipellis ex cellulis clavatis, $40-60 \times 12-25 \mu\text{m}$, ixohymenidermium formantibus. Fibulae nullae. Ad fimum equinum.

Holotypus: Hispania, Málaga, prov. Ronda, Nava de San Luis, 2 Nov. 1994, Alcoba, Esteve-Raventós, Horak & Ortega, AH 19372 (holotypus). Isotypus: GDAC 39965, paratypus: ZT 5273, AH 19577.

Pileus $10-35 \times 4-12 \text{ mm}$, at first narrowly cylindrical-conical with obtuse apex, then conical-campanulate, not or barely expanding, colour varying between golden-yellow, egg-yellow to lemon-yellow when young, gradually changing to grey-ferruginous or ochraceous-brown in ageing and overmature basidiocarps, surface smooth, viscid to strongly glutinous when moist, margin straight weakly transparent-striate, attached to stipe in young and collapsing pilei, but separated in mature specimens, veil remnants absent. Lamellae very crowded, straight and radially arranged (rarely anastomosing but never lacunose), free to adnexed to stipe, slightly ventricose, ochraceous to ochre-orange at maturity, rapidly deliquescent, autolysis beginning at the whitish edges. Stipe $40-110 \times 1-4 \text{ mm}$, cylindrical, flexuose, fistulose, brittle, lemon-yellow when fresh, then straw-yellow to isabel, at first conspicuously pruinose to furfureous, becoming glabrous with age, dry, hollow, solitary or in groups, rarely cespitose, veil remnants absent. Context in pileus and stipe very fragile. Smell fungoid. Taste not recorded. Spore print brilliant rust orange.

Basidiospores $12-15(-18) \times 6.5-9(-12) \mu\text{m}$, smooth (also in SEM), ellipsoid, sometimes slightly phaseoliform and constricted in profile, with up to $0.6 \mu\text{m}$ thick walls, orange-brown in KOH (2%), with broad germ pore ($1.2-1.8 \mu\text{m}$ diam.), mostly with central apiculus, occasionally with attached remnants of sterigmata. Basidia $23-30 \times 11-13 \mu\text{m}$, sphaeropedunculata, 4-spored, sterigmata up to $3.5 \mu\text{m}$. Cheilocystidia up to $45 \mu\text{m}$ in diam., globose to ovoid, forming a nearly homogeneous sterile band on the lamellar edges, sometimes forming two or even three aggregated rows, easily collapsing, thin-walled, hyaline. Hymenial trama soon collapsing. Pleurocystidia absent. Caulocystidia up to $70 \times 9-13 \mu\text{m}$, conspicuous, polymorphic, usually cylindrical to clavate or slenderly vesiculose, without irregular, finger- or knob-like excrescences (cf. *Bolbitius vitellinus*), thin-walled, hyaline. Pileipellis an ixohymeniderm, composed of long clavate elements $40-60 \times 12-25 \mu\text{m}$, embedded in gel, with yellowish plasmatic pigment, hypoderm of cylindrical, parallel hyphae (up to $3 \mu\text{m}$ in diam.). Clamp-connections absent at all septa of the hyphae and basidia.

Collections studied. SPAIN: Málaga, Nava de San Luis, 2 Nov. 1994, on horse dung in pastures, Alcoba, Esteve-Raventós, Horak & Ortega, AH 19372 (holotype); same locality, GDAC 39965 (isotype); Ronda, Sierra de las Nieves, Pilones, Casa forestal Felix Rodriguez de la Fuente, 2 Nov. 1994, same habitat, Horak 5273 (ZT, paratype); Huelva, Aracena, 27 Nov. 1999, on donkey dung in pastures, R. Galán, G. Moreno & L. Romero de la Osa, AH 19577 (paratype).

DISCUSSION

The most distinctive characters of *Bolbitius elegans* are the yellow colours of the gasteroid, subdeliquescent basidiocarps, the well-developed ixohymeniderm of the pileipellis and the conspicuous, vesiculose to subglobose cheilocystidia on the lamella edges.

In the field the basidiocarps of this taxon with conical and non-expanding pilei can easily be mistaken either for an aberrant form belonging to the *B. vitellinus*-complex or for a species related to the heterogeneous group of gasteroid Bolbitiaceae (Watling & Gregory, 1981).

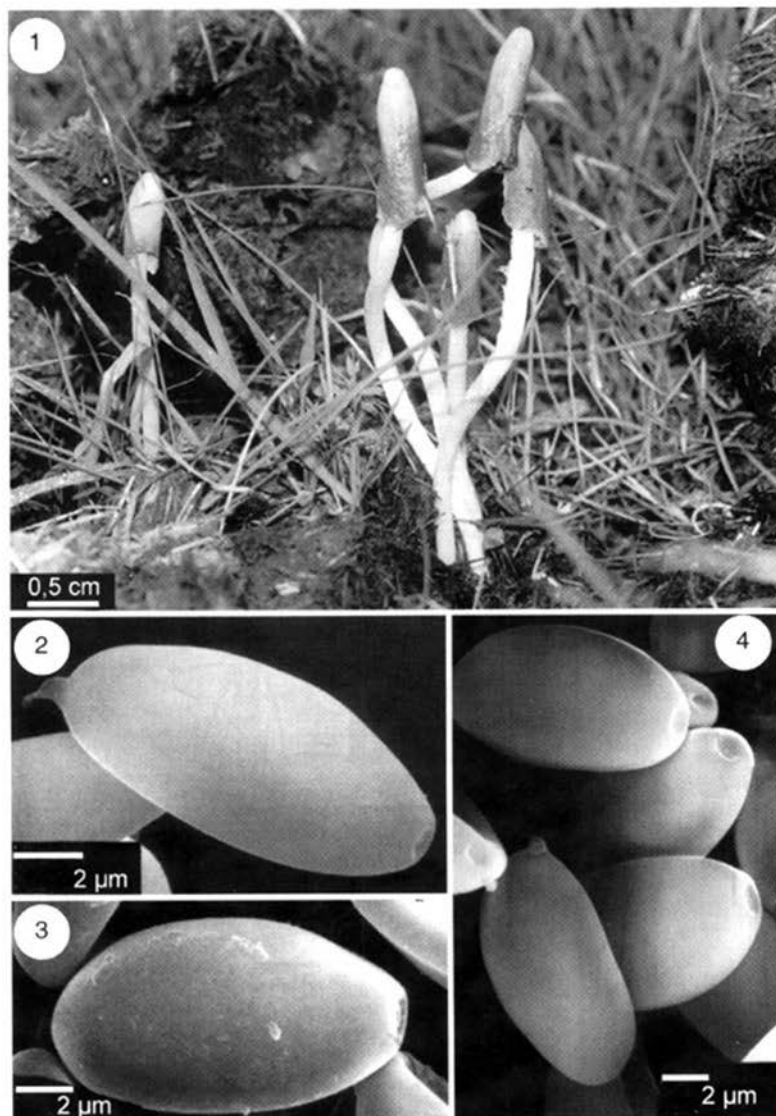


Fig. 1. *Bolbitius elegans* (AH 19372, holotype). — 1. Basidiocarps; 2–4. basidiospores showing germ pore.

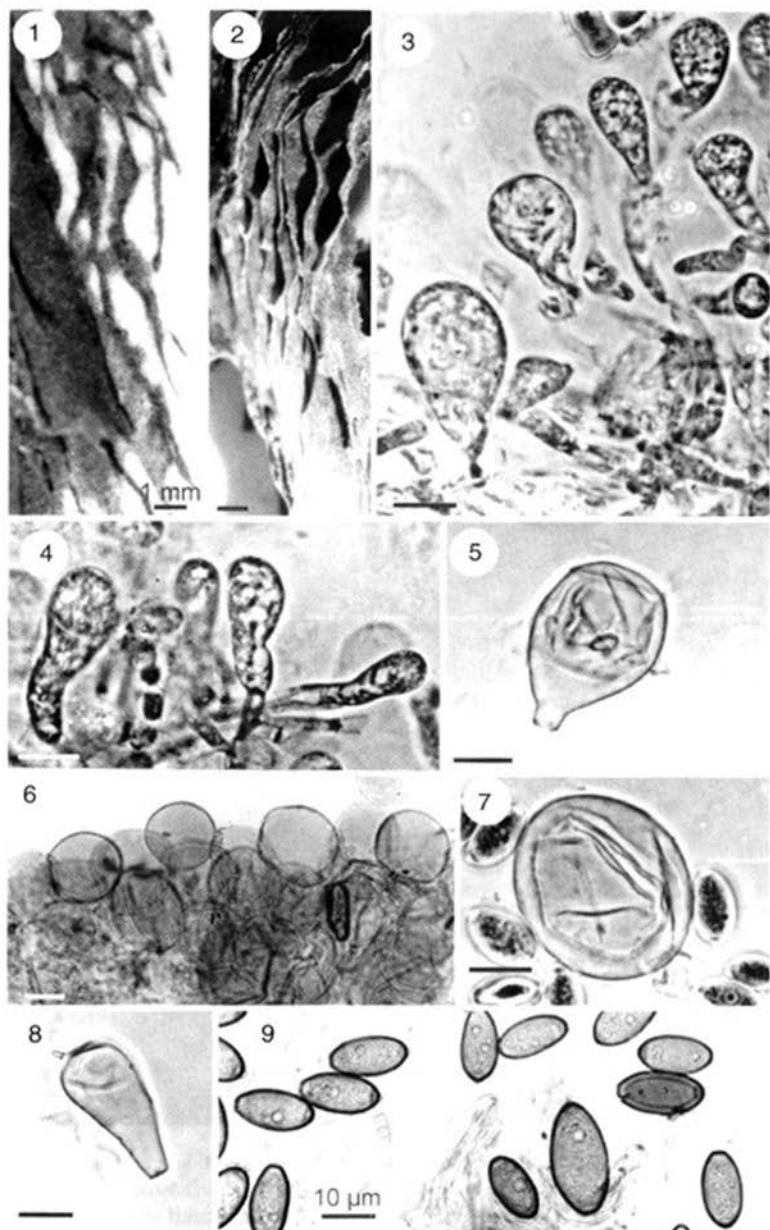


Fig. 2. *Bolbitius elegans* (AH 19372, holotype). — 1 & 2. Collapsed and agglutinated lamellae near pileal insertion (bar = 1 mm); 3 & 4. cells of pileipellis forming an ixohymeniderm (bar = 10 μ m); 5. edge of lamella with cheilocystidia (bar = 10 μ m); 6 & 7. cheilocystidia (bar = 10 μ m); 8. basidium; 9. spores.

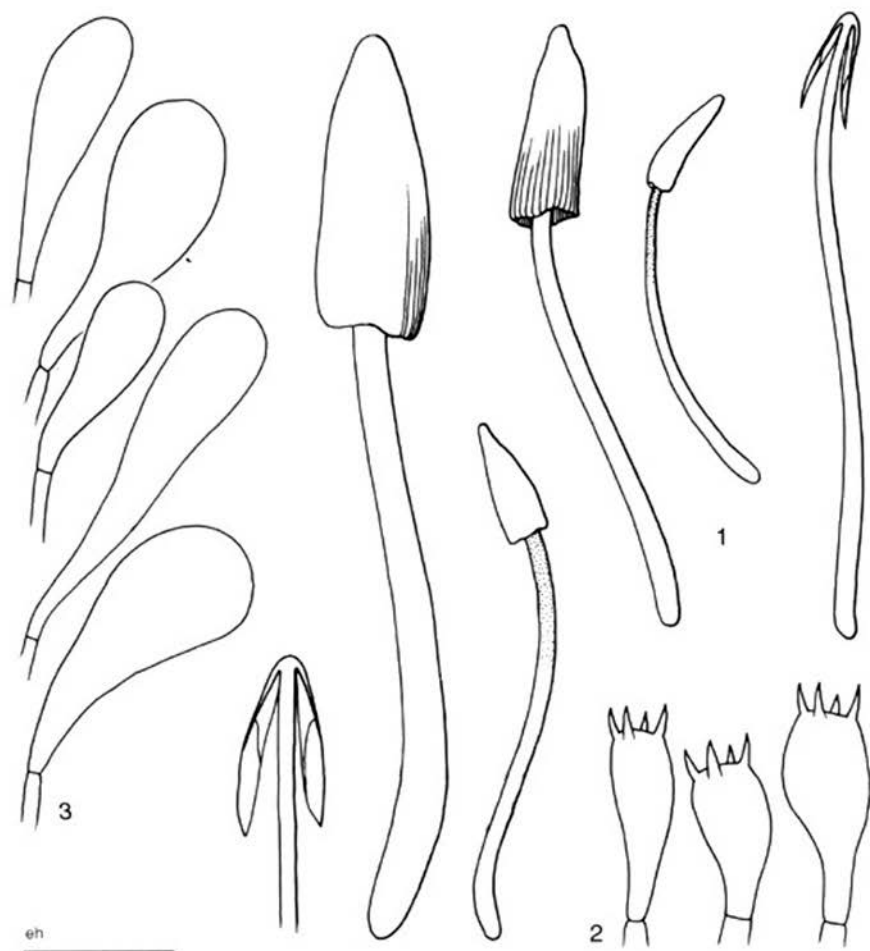


Fig. 3. *Bolbitius elegans* (ZT 5273, paratype). — 1. Basidiocarps (bar = 20 mm); 2. basidia ($\times 1000$, bar = 20 μm); 3. caulocystidia ($\times 1000$, bar = 20 μm).

The pileal ixohymeniderm composed of elongate, clavate cells packed into a gelatinous matrix, clearly suggests a taxon belonging to the genus *Bolbitius*. Some species of *Conocybe* Fayod, which, however, are non-deliquestent, may also have a slightly gelatinized pileipellis, e.g. *C. coprophila* (Kühner) Kühner and *C. rickenii* (Schaeff.) Kühner. In comparison to these taxa the thickness of the ixohymeniderm in *B. elegans* exceeds that of the hymenoderma.

Following the taxonomic concepts proposed by Bon (1992), only a few species of *Bolbitius* (considered as *Conocybe* by some authorities) actually have typical lecythiform cheilocystidia, e.g. *B. tener* Berk. & Broome (= *Conocybe lactea* ss. auct.), *B. lacteus* J.E. Lange and *B. crispus* (Longyear) Bon. In *B. elegans*, however, the edges of the lamellae

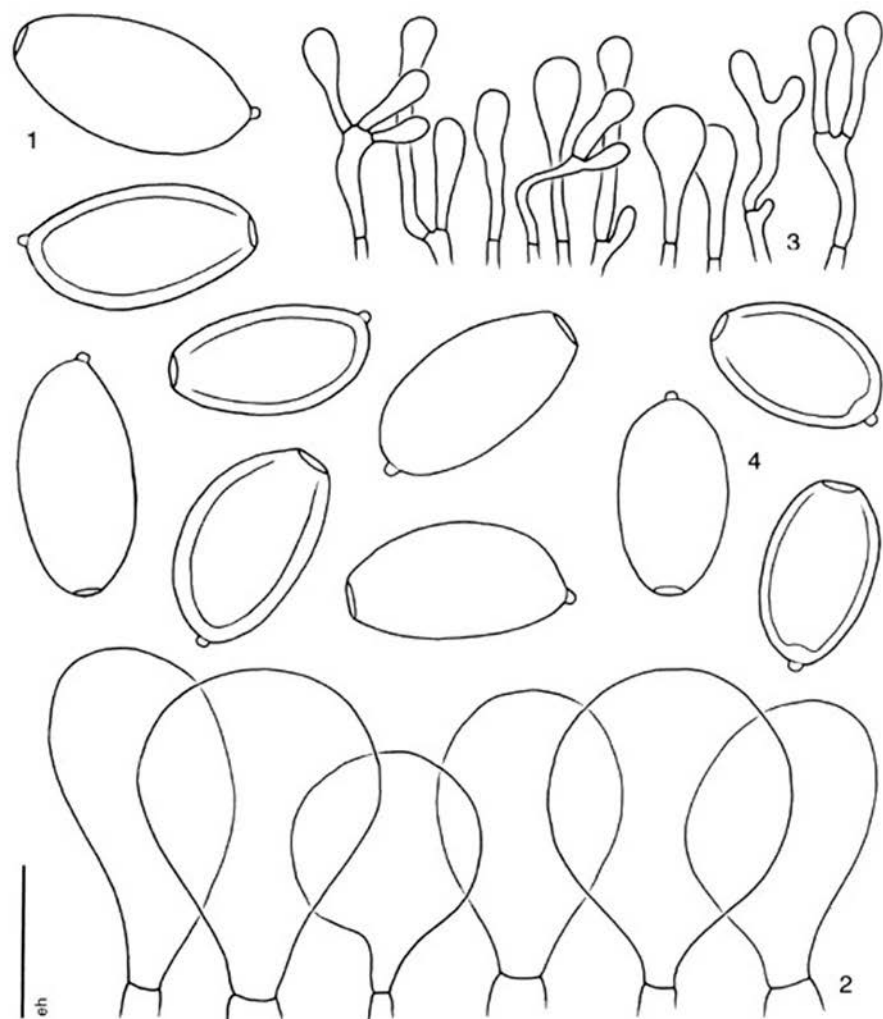


Fig. 4. *Bolbitius elegans* (ZT 5273, paratype). — 1. Basidiospores ($\times 2000$, bar = 10 μm); 2. cheilocystidia ($\times 1000$, bar = 20 μm); 3. pileipellis (ixohymeniderm, $\times 500$, bar = 40 μm). — *Gastrocybe lateritia* Watling (E 18098). 4. Basidiospores ($\times 2000$, bar = 10 μm).

are exclusively composed of large, broadly clavate to globose-vesiculose cheilocystidia. This distinctive character separates *B. elegans* from related taxa belonging to the *B. vitellinus*-complex, in which the shape of the cheilocystidia and/or caulocystidia usually ranges from fusiform or lageniform to irregular (Horak, 1968; Enderle et al., 1985; Cacialli et al., 1995).

Depending on edaphic and microclimatic conditions, the basidiocarps of many taxa of *Bolbitius* more or less exhibit a tendency to deliquesce. This phenomenon is also observed in the new species from Spain. In contrast to other taxa, the rather fast decomposition process observed in *B. elegans* does not allow the narrowly conical pileus to expand and thus the basidiocarps retain the secotioid aspect.

In the opinion of Watling (1968), *Gastrocybe* is probably a secotioid ally of *Bolbitius*. The description of the type species, *G. lateritia* Watling (collected in Michigan, USA), emphasizes the hymeniform pileipellis, formed by pyriform, hyaline cells (Babos, 1987). Exclusively based on the assumption of a passive discharge of the basidiospores, Watling (1968) concludes that *G. lateritia* should be considered a secotioid genus. However, it must be stressed that many taxa of *Coprinus* Pers., developing typical agaricoid basidiocarps, are also characterized by both deliquescence and passive discharge of the basidiospores.

The same year as Watling proposed the new genus *Gastrocybe*, Heim (1968) described *Bolbitius rogersii* (ad. int., nom. nud.) from Africa. This species is defined as a "*Galeropsis* charnu ou *Bolbitius* subangiocarpe?". The macromorphological features of this taxon recall *B. elegans* but the two species are readily separated by their microscopic characters.

Subsequently, Singer (1975) and Singer & Ponce de Leon (1982) have considered Heim's invalidly published taxon as a synonym of *Gastrocybe lateritia* Watling, whose macrocharacters clearly point to its intermediate taxonomic position between bolbitious taxa with agaricoid and secotioid basidiocarps. In the case of *G. lateritia*, microscopic features (e.g. lecythiform cheilocystidia) support a close affinity to *Conocybe* sect. *Candidae* (Kühner ex Singer) Bon. Despite the conspicuous, clavate-vesiculose cheilocystidia observed in the new Spanish agaric, there is no doubt that *Bolbitius elegans* also belongs to the complex of rather polymorphic taxa centred around *B. vitellinus*.

In the course of evaluating the identity of *Bolbitius elegans*, an extensive literature search [supported by the data published in Murrill (1917), Watling & Gregory (1981) and Watling (1982)], referring to taxa described as belonging to either *Bolbitius* or *Pluteolus* has been carried out. The protologues of about 50 species have been screened in detail but no description was discovered which matches the specific characters observed in the new Spanish representative of *Bolbitius*.

The procedure also included taxa of secotioid genera (Moreno et al., 1989) characterized by basidiocarps whose general habit recall those of *Bolbitius elegans*. Accordingly, the following secotioid species have been critically compared and subsequently found not to be conspecific:

Cyttarophyllopsis cordispora R. Heim (1968).

Galeropsis angusticeps (Peck) Singer (1963) (Bas.: *Galera angusticeps* Peck, Bull. Torrey Bot. Club 24 (1897) 143).

Galeropsis aporos Courtecuisse (1993).

Gastrocybe iberica G. Moreno, Illana & Heykoop (1987).

Gastrocybe lateritia Watling (1968).

Panaeolopsis nirimbi Watling & Young (1983).

Additional collections examined for comparison

Bolbitius coprophilus (Peck) Hongo, Mem. Fac. Liberal Arts Educ. Shiga Univ., Nat. Sci. 9 (1959) 82. CZECH REPUBLIC: Moravia, Paskov, in caldario SEMBRA, 21 Feb. 1989, det. Kuthan, CS 89-40 (ZT 6252).

Bolbitius variicolor Atkinson. Stud. Am. Fungi (1900) 154. CZECH REPUBLIC: Moravia, Paskov, in caldario SEMPRA, 6 March 1989, det. Kuthan, CS 89-51 (ZT 6253).

Bolbitius vitellinus (Pers.: Fr.) Fr. Epicrisis (1838) 254. SWITZERLAND: GR, Davos, Teufi, 1680 m, on rotting hay, 27 July 1964, det. Horak, 64-212 (ZT); GR, Susch, 1440 m, in manured meadow, 17 Sept. 1975, det. Horak, 6351 (ZT); SO, Olten, Kappel, among rotting grass, Oct. 1963, det. Horak, 63-263 (ZT). TI; Verzasca, Alpe Tencia near Brione, in manured grassland, 1500 m, 14 Oct. 1966, det. Horak, 66-690 (ZT). — USA: TN, Johnson Co., Shady Valley, among rotting leaves on soil, 17 June 1987, det. Horak, 3963 (ZT).

Gastrocybe lateritia Watling. Michigan Bot. 7 (1968) 20. SPAIN: Barcelona, on lawn in newly arranged garden, autumn 1984, Tabarés, det. Watling, 18100 (E). — CANADA: Ontario, Senator O'Connor school, in lawn, 14 Aug. 1983, Miller, det. Watling, 18098 (E). — ITALY: Rome, in lawn with *Poa pratensis*, July 1984, det. Watling, 18099 (E).

ACKNOWLEDGEMENTS

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THREE NEW SPECIES OF ENTOLOMA FROM KERALA STATE, INDIA

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Three new species of *Entoloma* s.l. from Kerala State, India are described, illustrated and discussed. *Entoloma haematinum*, a very small, bright red, omphalinoid species, reminiscent of *Hygrocybe cantharellus*, is unique because of its quadrate-cuboid spores; it fits well in subgenus *Omphaliopsis*. *Entoloma nubilum* and *E. carneum* are both characterised by their small, pleurotoid basidiocarps. The first is related to the species of subgenus *Leptonia*; the second fits better in subgenus *Claudopus*. Comments are given on the taxonomic position of the new species.

Following the publication of a preliminary account of the genus *Entoloma* s.l. from Kerala State, India (Manimohan et al., 1995), several interesting collections belonging to the genus have subsequently been made as part of a continuing study of the agaric mycota of this region. Based on some of these collections, three new species of *Entoloma* are described, illustrated and discussed below. Materials and methods adopted are the same as those given in the paper cited above. Colour names given in quotation marks and colour codes refer to Komerup & Wanscher (1978). All collections cited are deposited in the Nationaal Herbarium Nederland, Leiden (L).

Entoloma haematinum Manim., Leelav. & Noordel., *spec. nov.* — Figs. 1–3

Basidiocarpus parvus, omphalinoidus. Pileus 5–15 mm latus, convexus, haematinus, translucidus, primo pellucido-striatus, postea sulcato-striatus, glaber. Lamellae subdecurrentes vel decurrentes, aurantiae, subconfertae, lamellis intermixtae, marginibus concoloratae et integrae. Stipes 10–20 × 0.5–1.5 mm, centralis, cylindricus, cavus, haematinus. Odor nullus. Sporae 6.5–8.5 × 6–8 µm, quadratae. Basidia 25–31 × 9–10 µm, clavata, 2- vel 4-sporigera. Acies lamellarum fertilis. Cheilocystidia et pleurocystidia nulla. Cuticula pilei ex hyphis repentibus, 7.5–20 µm latis composita. Fibulae nullae.

Holotypus: India, Kerala State, Malappuram District, Nilambur Teak Forest, 14 Nov. 1997, Manimohan M740a (L).

Etymology: haematinum (Latin), blood red.

Basidiocarps very small, omphalinoid, delicate, translucent, reminiscent of *Hygrocybe cantharellus*. Pileus 5–15 mm diam., hemispheric to convex, with or without a slight central depression; with straight margin, not hygrophanous, initially finely translucently striate, bright red or blood red (exact shade of red not available in Komerup & Wanscher, 1978; nearest shade 9C8), smooth, glabrous, becoming finely radially sulcate with age. Lamellae well-developed, moderately crowded, without or with lamellulae of 1 or 2 lengths, sub-decurrent to deeply decurrent, up to 2 mm broad, 'light orange' (8A4, 5A5, 6A4, 6A5) with entire, concolorous edge. Stipe 10–20 × 0.5–1.5 mm, central, terete, equal with slightly enlarged base, hollow, concolorous with pileus or slightly paler, smooth, glabrous, with scanty basal mycelium. Context 'light orange' (6A4, 6A5), less than 0.5 mm thick. Odour and taste not distinctive.

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2) Nationaal Herbarium Nederland, Universiteit Leiden branch, P.O. Box 9514, 2300 RA Leiden, The Netherlands.

Spores $6.5\text{--}8.5 \times 6\text{--}8$ ($7.6 \pm 0.65 \times 7.4 \pm 0.6$) μm , $Q = 1.00\text{--}1.08$, $Q_m = 1.03$, $n = 20$, cuboid, mostly quadrate in profile, rarely 5-sided, with slightly concave facets. Basidia $25\text{--}31 \times 9\text{--}10$ μm , clavate, 2- or 4-spored; sterigmata less than 5 μm long. Lamella-edge fertile. Hymenial cystidia absent. Hymenophoral trama regular; hyphae made up of inflated, up to 16 μm wide elements with hyaline, thin walls. Pileipellis a cutis of cylindrical hyphae, made up of short, inflated elements, $7.5\text{--}20$ μm wide, with clavate terminal elements, $20\text{--}60 \times 7.5\text{--}23$ μm , thin-walled, with a yellowish plasmatic pigment, without encrustations. Pileitrama interwoven; hyphae made up of inflated, $6\text{--}25$ μm wide, thin-walled elements, with yellowish plasmatic pigment. Stipitipellis a cutis of $3\text{--}15$ μm wide, thin-walled hyphae, with yellowish plasmatic pigment, without encrustations; frequently showing bundles of caulocystidia, $25\text{--}35 \times 11\text{--}14$ μm , inflated-clavate, with yellowish plasmatic pigment. Stipititrama made up of parallelly arranged hyphae; elements $60\text{--}90 \times 3\text{--}15$ μm , thin-walled, with a yellowish plasmatic pigment. Clamp-connections absent. Oleiferous hyphae rare.

Habitat — Saprotrophic, on the ground, amongst decaying leaf litter, scattered. November – December.

Specimens examined. INDIA: Kerala State, Malappuram District, Nilambur Teak Forest, 14 Nov. 1997, Manimohan M740a (holotype, L.); 18 Nov. 1997, Manimohan M740b; 31 Dec. 1997, Manimohan M740c.

Entoloma haematinum is a rather unusual species with its very small, bright red, omphalinoid basidiocarps. In addition, some microscopical features, such as cuboid spores, absence of both clamp-connections and pigment encrustations, plasmatic pigment and presence of inflated-clavate caulocystidia are characteristics of this species. Red basidiocarps are extremely rare in *Entoloma* and according to Horak (1980), the Indian species *Entoloma nanum* (Masse) E. Horak is the only one known to show this colour amongst the entire range of Indomalayan and Australasian species of *Entoloma* studied by him. *Entoloma nanum* is easily distinguished by its wine-red to ochre-red, conico-companulate pileus, adnato-adnexed lamellae, smaller, isodiametric-polygonal spores and cheilocystidia. Extensive searches in the literature revealed no comparable species from other geographical regions. The taxonomic position of this species is also interesting. We place it in the subgenus *Omphaliopsis* Noordel. since it does not fit in subgenus *Claudopus* (A. Gillet) Noordel. because of the colours and pigments. Cuboid spores are rare in subgenus *Omphaliopsis*, but do occur in some African species (Romagnesi & Gilles, 1979). As most species of subgenus *Paraleptonia* Romagn. ex Noordel. have clamp-connections and faint encrustations in the pileipellis, it is difficult to place the new species in that subgenus.

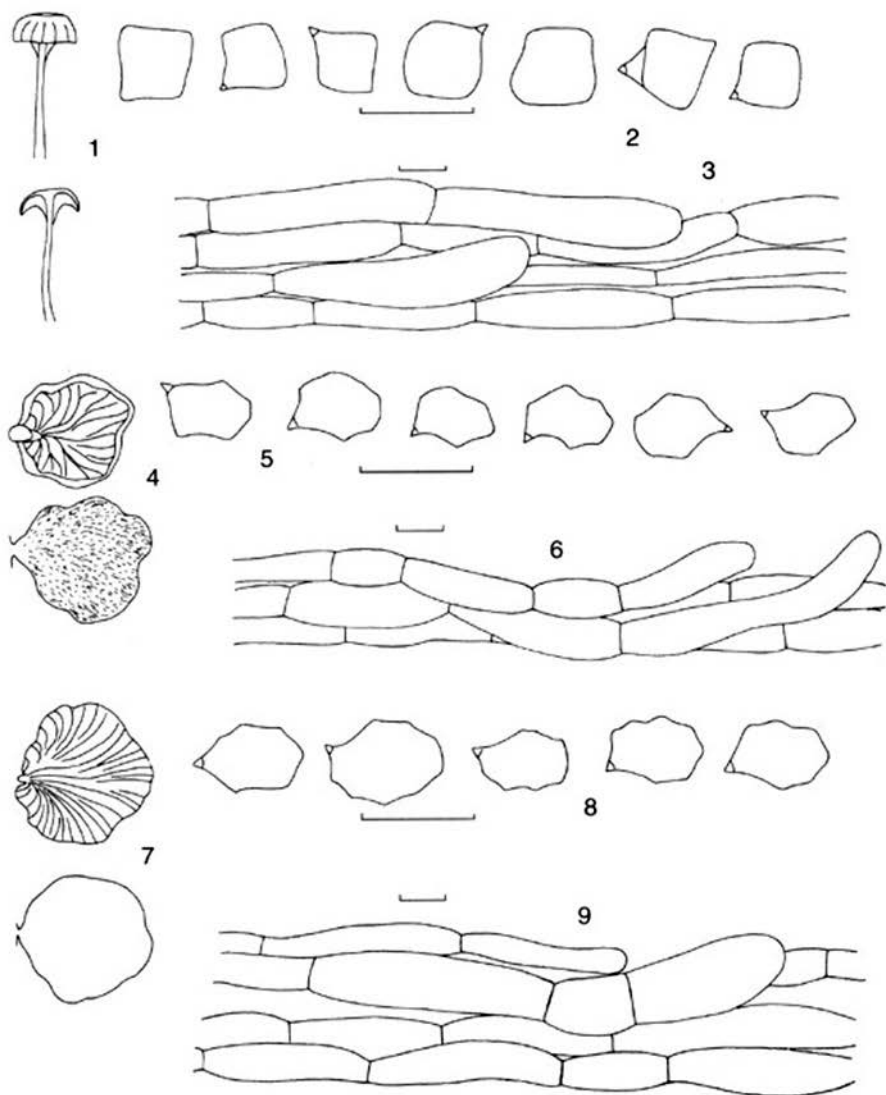
***Entoloma nubilum* Manim., Leclav. & Noordel., spec. nov. — Figs. 4–6**

Basidiocarpus parvus, pleurotoidus. Pileus 4–8 mm latus, cupulato reniformis vel subflabelliformis, lividus, estriatus, glaber. Lamellae adnexae, primo caesiae, postea aurantio-griseae, subremotae, lamellulis intermixtae, marginibus concoloratae et integrae. Stipes brevissimus, lateralis. Odor nullus. Sporae $7.5\text{--}9 \times 5\text{--}6$ μm , heterodiametrico-ovatae, 6–7 angulatae. Basidia $30\text{--}40 \times 8.5\text{--}10$ μm , clavata, 4-sporigera. Acies lamellarum fertilis. Cheilocystidia et pleurocystidia nulla. Cuticula pilei ex hyphis repentibus, $5\text{--}12.5$ μm latis composita. Fibulae nullae.

Holotypus: India, Kerala State, Malappuram District, Nilambur Teak Forest, 14 Nov. 1997, Manimohan M739a (L).

Etymology: nubilum (Latin), greyish blue.

Basidiocarps small, pleurotoid. Pileus 4–8 mm, cupulate-reniform to subflabelliform to almost unguulate with incurved entire margin which becomes lobate and fissurate with



Figs. 1-3. *Entoloma haematinum*. 1. Basidiocarp; 2. spores; 3. elements of pileipellis. — Figs. 4-6. *Entoloma nubilum*. 4. Basidiocarp; 5. spores; 6. elements of pileipellis. — Figs. 7-9. *Entoloma carneum*. 7. Basidiocarp; 8. spores; 9. elements of pileipellis. — Scale bars = 10 μ m.

age; not hygrophanous, not translucently striate, 'ink blue' (20F4), finely appressed hairy to almost smooth. Lamellae with lamellulae of 2–5 lengths, subdistant, adnexed, up to 2 mm wide, initially 'bluish grey' (20C2), becoming 'orange grey' (6B2) with age, with entire, concolorous slightly bluish edge. Stipe strongly reduced to almost absent, lateral, knob-like, with dimensions less than 1 × 1 mm; surface concolorous with the pileus, densely pruinose to tomentose. Context thin, white, less than 1 mm thick. Odour and taste not distinctive. Dried specimens retaining the blue colour.

Spores $7.5-9 \times 5-6$ ($8 \pm 0.4 \times 5.2 \pm 0.34$) μm , $Q = 1.33-1.8$, $Q_m = 1.54$, $n = 20$, heterodiametric-ovate with 6 or 7 concave sides in profile. Basidia $30-40 \times 8.5-10$ μm clavate, 4-spored; sterigmata up to 5 μm long. Lamella-edge fertile. Cheilocystidia and pleurocystidia absent. Hymenophoral trama regular; hyphae 3.5–15 μm wide, thin-walled, hyaline. Pileipellis a cutis of repent hyphae, 5–12.5 μm wide, often constricted at septa, thin-walled, with dark blue plasmatic pigment, without encrustations. Pileitrama parallel-interwoven; hyphae 3.5–18.5 μm wide, thin-walled, hyaline. Clamp-connections and oleiferous hyphae absent.

Habitat — Saprotrophic, on decaying leaves partially buried in soil or directly on humus-rich soil, scattered, November.

Specimens examined. INDIA: Kerala State, Malappuram District, Nilambur Teak Forest, 14 Nov. 1997, Manimohan M739a (holotype, L); 18 Nov. 1997, Manimohan M739b.

Entoloma nubilum is characterised by the combination of small, pleurotoid basidiocarps, dark blue, non-striate pileus, lateral, rudimentary stipe, heterodiametric-ovate spores with 6 or 7 concave facets in profile, cutis-type pileipellis with plasmatic pigment and absence of both pigment encrustations and clamp-connections. *Claudopus cyanomelaenus* Boedijn is the only blue *Claudopus* reported so far from Indomalaya and Australasia (Horak, 1980). Not much is known about its variability, however, since the species has only been recorded once from the type locality in Sumatra. Horak (1980) could not trace the type material. It is nonetheless clear that *Entoloma nubilum*, though similar to *C. cyanomelaenus* in overall morphology, differs in certain features. According to Horak (1980), the pileus of *C. cyanomelaenus* turns red in KOH, whereas the pileus of the present collections does not. The spores of *E. nubilum* are also distinctly smaller than those of *C. cyanomelaenus*. These features, along with the geographical difference, have led us to describe the collections from Kerala State as a new species. *Entoloma cyaneum* (Murrill) Hesler, another blue pleurotoid species described from Florida, is somewhat similar to the present collections. However, it is a lignicolous species with adnate lamellae, a well-developed, white stipe, slightly larger, 5 or 6 sided spores and colourless pileipellis hyphae.

The taxonomic position of *Entoloma nubilum* and *Claudopus cyanomelaenus* is intriguing. They certainly do not fit in the current concept of *Claudopus* (treated as either a 'genus' or a 'subgenus') because of the blue plasmatic pigment and the total lack of pigment encrustations. It is probably better to consider them as reduced forms in subgen. *Leptonia*, sect. *Cyanula*. As in other agaric genera (e.g. *Marasmius*, *Marasmiellus*, *Psilocybe*) pleurotoid basidiocarps are found in different parts of the infrageneric classification system and are by no means unique. For this reason we strongly believe that the pleurotoid habit in *Entoloma* is likewise polyphyletic, and may have occurred several times in the course of evolution of that genus. *Claudopus* in the sense of Horak (1980) is therefore an unnatural, heterogeneous assemblage of non-related species. For that reason, the following new combination is made here.

Entoloma cyaneomelaenus (Boedijn) Manim., Leelav. & Noordel., *comb. nov.*Basionym: *Claudopus cyaneomelaenus* Boedijn, Rec. Trav. bot. Neerl. 26 (1929) 419.**Entoloma carneum** Manim., Leelav. & Noordel., *spec. nov.* — Figs. 7–9

Basidiocarpus parvus, pleurotoidus. Pileus 5–35 mm latus, sublabelliformis, primo albidus, postea incarnatus, estriatus, glaber. Lamellae liberae, incarnatae, subconfertae, lamellulis intermixtae, marginibus concoloratae et integrae. Stepes brevissimus, lateralis vel excentricus, solidus. Odour nullus. Sporae 7.5–10 × 5–7 µm, heterodiametrico-ovatae, noduloso-angulatae. Basidia 30–39 × 9–11.25 µm, clavata, 4-sporigera. Acies lamellarum fertillis; cheilocystidia et pleurocystidia nulla. Cuticula pilei ex hyphis repentibus 3–21 µm latis composita. Hyphae fibulatae.

Holotypus: India, Kerala State, Kannur District, Pulikurumba, 23 Sept. 1997, Manimohan M729a (L).
Etymology: carneum (Latin), flesh-coloured.

Basidiocarps small to medium-sized, pleurotoid. Pileus 5–35 mm diam., convex, sublabelliform with lateral or excentric attachment; with initially straight, entire, with age upturned, irregularly lobate, fissile margin, not hygrophanous, not translucently striate, not zonate, whitish, becoming flesh-coloured (6B3, 6B2) with age, glabrous, with a silky sheen. Lamellae with lamellulae of several lengths, moderately crowded to crowded, free to remote, up to 3 mm wide, initially flesh-coloured (6B3, 6B2), becoming 'red-haired' (6C4), with entire, concolorous edge. Stipe up to 2 × 1 mm, rudimentary, excentric to lateral, terete, solid; concolorous with pileus, finely pruinose under a lens; with well-developed basal mycelium, and with mycelial cords. Context thin, flesh-coloured (6B2). Odour and taste not distinctive.

Spores 7.5–10 × 5–7 (8.6 ± 0.86 × 6.1 ± 0.55) µm, Q = 1.25–1.55, Qm = 1.4, n = 20, heterodiametric-ovate to almost heterodiametric-elliptic, irregularly nodulose-angular in side-view. Basidia 30–39 × 9–11.25 µm, broadly clavate, 4-spored; sterigmata up to 4.5 µm long. Lamella-edge fertile. Cheilocystidia and pleurocystidia absent. Hymenophoral trama regular, made up of 3.5–25 µm wide, short-celled, thin-walled, hyaline hyphae. Pileipellis an undifferentiated cutis of repent hyphae, 3–21 µm wide, short-celled, thin-walled, hyaline, without encrustations. Pileitrama parallel-interwoven; hyphae 4.5–20 µm wide, made up of short, hyaline, thin-walled elements. Stipititrama composed of frequently rather inflated, up to 40 µm wide, thin-walled, hyaline hyphae. Stipitipellis an undifferentiated cutis with recurved cylindrical hyphal tips frequently projecting out. Clamp-connections and oleiferous hyphae rarely observed in the hymenophoral trama.

Habitat — Saprotrophic, on decaying stem of palm (*Corypha umbraculifera* L.) in dense imbricate clusters, September.

Specimens examined. INDIA: Kerala State, Kannur District, Pulikurumba, 23 Sept. 1997, Manimohan M729a (holotype, L.); 29 Sept. 1997, Manimohan M729b.

Entoloma carneum is characterised by its palmycolous, pleurotoid basidiocarps which appear in dense imbricate clusters and have a whitish to flesh-coloured, glabrous pileus, free to remote lamellae, a rudimentary, lateral to excentric stipe, spores that are irregularly-nodulose in profile, no cystidia of any kind, a cutis-type pileipellis without encrusting or membranal pigment, and clamp-connections. In the size and shape of spores, in the absence of cystidia, and in the presence of clamp-connections, it is very similar to *Entoloma byssi-sedum* (Pers.: Fr.) Donk, a species widely distributed in temperate and boreal regions. The latter species, however, differs in having a darker, more grey-brown pileus, adnate to decur-

rent lamellae, farinaceous odour and membranal and encrusting pigments in the pileipellis and pileitrama. The palmicolous habitat and growth-form in dense, imbricate clusters are also distinctive for *E. carneum*. *Entoloma depluens* (Batsch: Fr.) Hesler also seems to be very similar, but differs in having silvery-white hairs on pileus and abundant cheilocystidia. For the time being, it seems best to include *Entoloma carneum* in subgenus *Claudopus*, despite the lack of encrusting pigments, although the considerations in the discussion under *E. nubilum* should be taken into account with regard to the taxonomic status of species with a pleurotoid habit.

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**HYDROPS PARADOXUS VAR. XEROPHYTICUS AND
A KEY TO THE TAXA KNOWN FROM EUROPE**

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Hydopus paradoxus var. *xerophyticus*, characterized by its long cystidia, broad spores and habitat in xerophytic basic pastures with communities of *Thymus-Lavandula* spp. is described as a new taxon from Spain. In addition, a key to 22 taxa known from Europe is given.

The genus *Hydopus* (Kühner) Singer ex Singer has been the subject of very few monographical studies, both at European and at world-wide level. The only monograph of this genus is Singer's (1982) which deals only with tropical species. In Europe, one of the first authors who studied this genus was Kühner (1938), who recognised four species though he included them in *Mycena* (Pers.) Roussel. Later, Moser (1983) recognised the genus *Hydopus* as a taxon on its own, and provided the first European key comprising a total of seven species. We also stress the importance of contributions published by Robich (1986, 1990, 1992), Contu & Robich (1998) in Italy, Hausknecht et al. (1997) in Austria and Bas (1999) in the Netherlands.

The present paper describes a new variety of *Hydopus paradoxus* from Spain. The colour codes given in this paper are according to Munsell (1988).

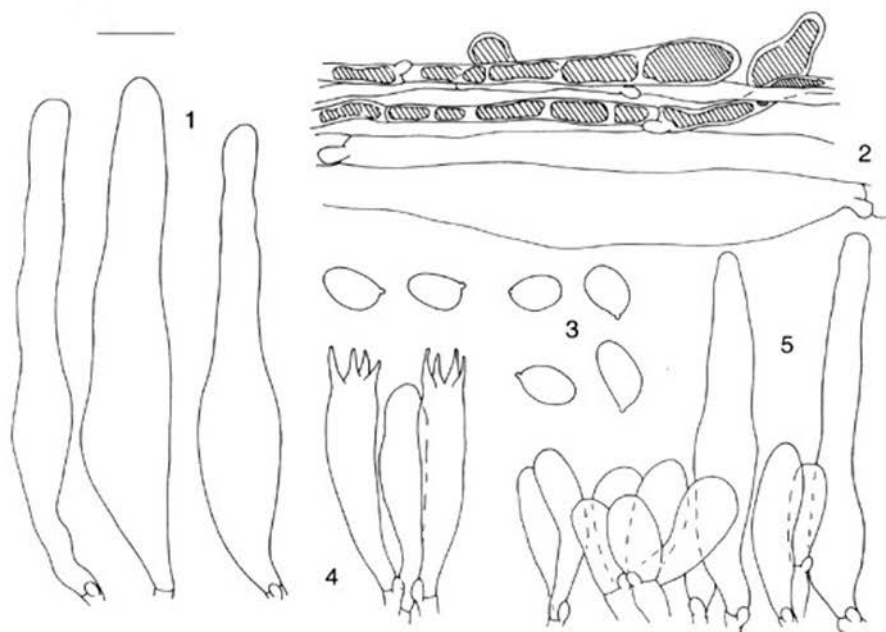
***Hydopus paradoxus* var. *xerophyticus* Esteve-Rav., Villarreal & Heykoop, var. nov. —
Figs. 1–5**

A typo differt sporis latioribus, cystidiis longioribus et habitatione aridiore.

Holotypus: SPAIN, Segovia, Parque Natural de las hoces del Río Duratón, 10 Nov. 1993, leg. F. Esteve-Raventós, M. Heykoop, S.G. Busutil & P.G. Escolar (AH 18987).

Basidiomata gregarious. Pileus 10–30 mm in diam., hemispherical, convex to planoconvex, sometimes with abrupt central papilla, hygrophanous, translucent-striate only when wet, apparently smooth, fairly pruinose under the lens, very dark brown (Mu. 10 YR 2/2) to dark brown (10 YR 3/3), fading to greyish brown (10 YR 4/2), with yellowish brown (10 YR 5/6), crenulate margin. Lamellae ascending, broadly ventricose, deeply emarginate with decurrent tooth, white to dirty greyish when dry, with entire, concolorous edge. Stipe 15–50 × 2–4 µm, cylindrical to tapering upwards, very pale brown (10 YR 7/4), gradually darkening towards the base to greyish brown or dark brown (10 YR 4/2–3), always somewhat paler than the pileus, entirely pruinose-floccose, at the base densely covered with brownish fibrils. Context somewhat cartilaginous, whitish to pale brown under the cortex at the base of the stipe. Smell none.

Spores 7.7–9.8–11.9 × 5.4–6.35–7.3 µm; Q = (1.32–)1.45–1.55–1.65(–1.95); (n = 21), ellipsoid, broadly ellipsoid, or subglobose, smooth, thin-walled, hyaline, with vacuolar inclusions, inamyloid, acyanophilic. Basidia 32–42 × 8.5–10 µm, 4-spored (rarely 2-spored),



Figs. 1–5. *Hydropus paradoxus* var. *xerophyticus* (holotype). 1. Hymenial cystidia; 2. pileipellis; 3. spores; 4. basidia; 5. Caulocystidia. Bar = 15 μ m.

sterigmata 4.5–9 μ m long, hyaline or with vacuolar contents, clamped. Lamella edge heterogeneous. Cheilocystidia and pleurocystidia abundant, (50–)70–110(–137) \times 12–17(–23) μ m, normally sublageniform with long neck, but also subutriform to clavate, mostly thick-walled (–1.5 μ m). Hymenophoral trama regular to subregular, not embedded in gelatinous matter, not dextrinoid, consisting of long and cylindrical hyphae (–27 μ m wide), constricted at the septa. Pileipellis consisting of hyphae 2–5 μ m wide, with numerous prostrate cylindrical, cylindrico-flexuose to subclavate dermatocystidioid elements up to 80 \times 7–12 μ m, cylindrical to clavate, filled with brownish vacuolar contents, not forming a well-developed palisade, and locally forming denses clusters. Subpellis made of wider and shorter elements (15–32 μ m wide), forming a pseudoparenchymatic layer with parietal yellowish pigment. Stipitipellis a cutis of cylindrical, parallel 2–5 μ m wide hyphae with parietal yellowish pigment, with caulocystidioid terminal elements at the stipe apex, very variable, cylindrical, clavate to sublageniform, 30–85 \times 5–10 μ m, thin-walled, forming clusters. Context not dextrinoid, not cyanophilic nor oleiferous elements observed. Sarcodimitic tissues present at the cortical layer of the stipe. Clamps present, but sometimes inconstant.

Habitat — In xerophytic grassland, under *Lavandula stoechas* L. and *Thymus* sp.

Material studied. SPAIN: Segovia, Parque Natural de las hoces del Río Duratón, 5 Nov. 1993, leg. F. Esteve-Raventós, M. Heykoop, S.G. Busutil & P.G. Escolar, AH 18986; *ibidem*, 10 Nov. 1993, AH 18987 (Holotype).

Hydropus paradoxus is an extremely rare taxon; according to our knowledge, it is known only from the type locality in Switzerland (Moser, 1969). The Spanish collections, which grow in very xerophytic shrubland areas with poor, basic soils, seem to be restricted to this vegetation where the presence of *Thymus* and *Lavandula* species seems to be constant. The basidiomata grow directly on soil or more commonly on and around debris under the canopy of these two characteristic Mediterranean plants. Apart from this typical habitat, the broader spores and longer cystidia and basidia, seem to be different from those described in the type collection. Macroscopically both taxa are very similar, and the cartilaginous context and the ventricose, annexed greyish white gills, which characterise this species, are present in both taxa.

KEY FOR THE EUROPEAN SPECIES OF HYDRUPUS

- | | |
|--|---|
| 1. Spores amyloid | 10 |
| 1. Spores inamyloid | 2 |
| 2. Spores globose | 3 |
| 3. Smell spermatic, pileus dark brown, on alpine musci | <i>Hydropus montis-rosae</i> |
| 3. Without these characters | 4 |
| 4. Stipe, context and lamellae with yellowish tinges | |
| | <i>Hydropus floccipes</i> var. <i>luteipes</i> |
| 4. Without yellowish tinges | <i>Hydropus floccipes</i> var. <i>floccipes</i> |
| 2. Spores not globose | 5 |
| 5. Hymenial cystidia absent; alpine distribution | <i>Hydropus dryadicola</i> |
| 5. Hymenial cystidia present | 6 |
| 6. Pileocystidia absent | <i>Hydropus conicus</i> |
| 6. Pileocystidia present | 7 |
| 7. Gloeocystidia present | <i>Hydropus liciosae</i> |
| 7. Gloeocystidia absent | 8 |
| 8. Stipe whitish, spores reniform | <i>Hydropus subalpinus</i> |
| 8. Stipe brownish, spores ellipsoid to subglobose | 9 |
| 9. Spore width $\leq 5.5 \mu\text{m}$; cystidia up to $60 \mu\text{m}$ long | |
| | <i>Hydropus paradoxus</i> |
| 9. Spore width $> 5.5 \mu\text{m}$; cystidia $>$ than $60 \mu\text{m}$ long | |
| | <i>Hydropus paradoxus</i> var. <i>xerophyticus</i> |
| 10. Pileipellis with encrusting pigment | 11 |
| 11. Spores up to $5 \mu\text{m}$ wide, pileipellis without intracellular pigment, sarcodimitic tissues absent, lignicolous | |
| | <i>Dennisiomyces lanzonii</i> |
| 11. Without these characters. | 12 |
| 12. Basidia 4-spored, clamp-connections present | |
| | <i>Hydropus trichoderma</i> var. <i>trichoderma</i> |
| 12. Basidia 2-spored, clamp-connections absent | |
| | <i>Hydropus trichoderma</i> var. <i>lobauensis</i> |
| 10. Pileipellis without encrusting pigment | 13 |
| 13. Context darkening when cut, spores globose | |
| | <i>Hydropus atramentosus</i> |
| 13. Context not darkening, spores not globose | 14 |

14. Pileocystidia absent 15
15. Caulocystidia fusoid, with thick and frequently mucronate apex *Hydropus nitens*
15. Without these characters 16
16. Spores < 4.5 µm wide, base of stipe saffron-yellowish, pileipellis with some diverticulae
Hydropus pseudotenax
16. Spores > 4.5 µm wide, base of stipe without yellowish tinges 17
17. Basidia 2-spored
Hydropus scabripes var. *scabripes*
17. Basidia 4-spored
Hydropus scabripes var. *quadrisporus*
14. Pileocystidia present 18
18. Pleurocystidia present 19
19. Clamp-connections absent, crowded to rather crowded lamellae (L = 20–28, l = 1–5)
*Hydropus fraterniger*¹
19. Clamp-connections present, lamellae distant to subdistant 20
20. Basidia 2-spored, lamellae moderately close to subdistant (with e.g. 18 through-lamellae according to Singer, 1982)
*Hydropus fraterniger*¹
20. Basidia 4-spored, lamellae distant to very distant (L = 12–16, l = 0–1) *Hydropus moserianus*
18. Pleurocystidia absent 21
21. Pileipellis consisting of a dense layer formed by pileocystidia *Hydropus marginellus*
21. Pileipellis not consisting of a dense layer formed by pileocystidia (tropical species growing in European greenhouses) *Hydropus semimarginellus*

1) *Hydropus fraterniger* Singer, *H. fraterniger* s. Hausknecht et al. (1997) and *H. moserianus* Bas are three closely related, though different taxa. *H. fraterniger* s. Hausknecht et al. is characterized by its first dark grey-brown but later brown pileus with paler ochraceous margin, its lamellae with brownish grey edge because of the presence of cheilocystidia with vacuolar pigment, and the absence of clamps. The fruit-bodies on the coloured plate published by Hausknecht et al. (1997) are completely different from *H. moserianus*, especially because of the distant to very distant lamellae in *H. moserianus*. Moreover, the taxon of Hausknecht et al. grows on wood (lying stems of *Abies*). The original *H. fraterniger* Sing. differs from *H. moserianus* by 2-spored basidia (in the latter 4-spored), a (sub)umbonate pileus, white to grey lamellae becoming dark in the region along the edge (in *H. moserianus* grey to dark grey but paler towards the edge), probably less distant lamellae (according to Singer (1982): moderately close to distant, with e.g. 18 through-lamellae), and cystidia without a long narrow neck.

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**TERFEZIA BOUDIERI, FIRST RECORDS FROM EUROPE
OF A RARE VERNAL HYPOGEOUS MYCORRHIZAL FUNGUS**

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Terfezia boudieri is reported for the first time from the Iberian Peninsula and Europe. It is illustrated and compared with material and descriptions of similar *Terfezia* species *T. arenaria*, *T. claveryi* and *T. leptoderma*. Specimens of *T. boudieri* from Spain were also compared with those from Morocco.

Plants belonging to the Cistaceae, particularly species of *Helianthemum*, *Xolantha*, *Cistus*, *Halimium* and *Fumana*, occur in many semi-arid regions in the Mediterranean basin. Most of these plants establish mycorrhizas, and have a specific mycota associated with them, notably hypogeous ascomycetes (species of *Tuber*, *Terfezia*, *Tirmania* and *Picoa*) (Alsheikh & Trappe, 1983a; Moreno et al., 1986; 1991; Pacioni & El-Kholy, 1994; Moreno et al., 2000).

Three species of *Terfezia* have been reported so far from Spain: *T. arenaria* (Moris) Trappe, the most common of those occurring on acid sandy soils; *T. leptoderma* Tul. & C. Tul., which also occurs on acid soils but less frequently; and *T. claveryi* Chatin, typical on calcareous soils. These desert truffles, traditionally called in Spain 'criadillas de tierra' or 'turmas' are collected in spring under annual and perennial plants, often *Helianthemum* and *Xolantha* spp. (Moreno et al., 1986). Their abundance in favourable years, their high market value, and their exportation to Arabia, make them an important natural resource.

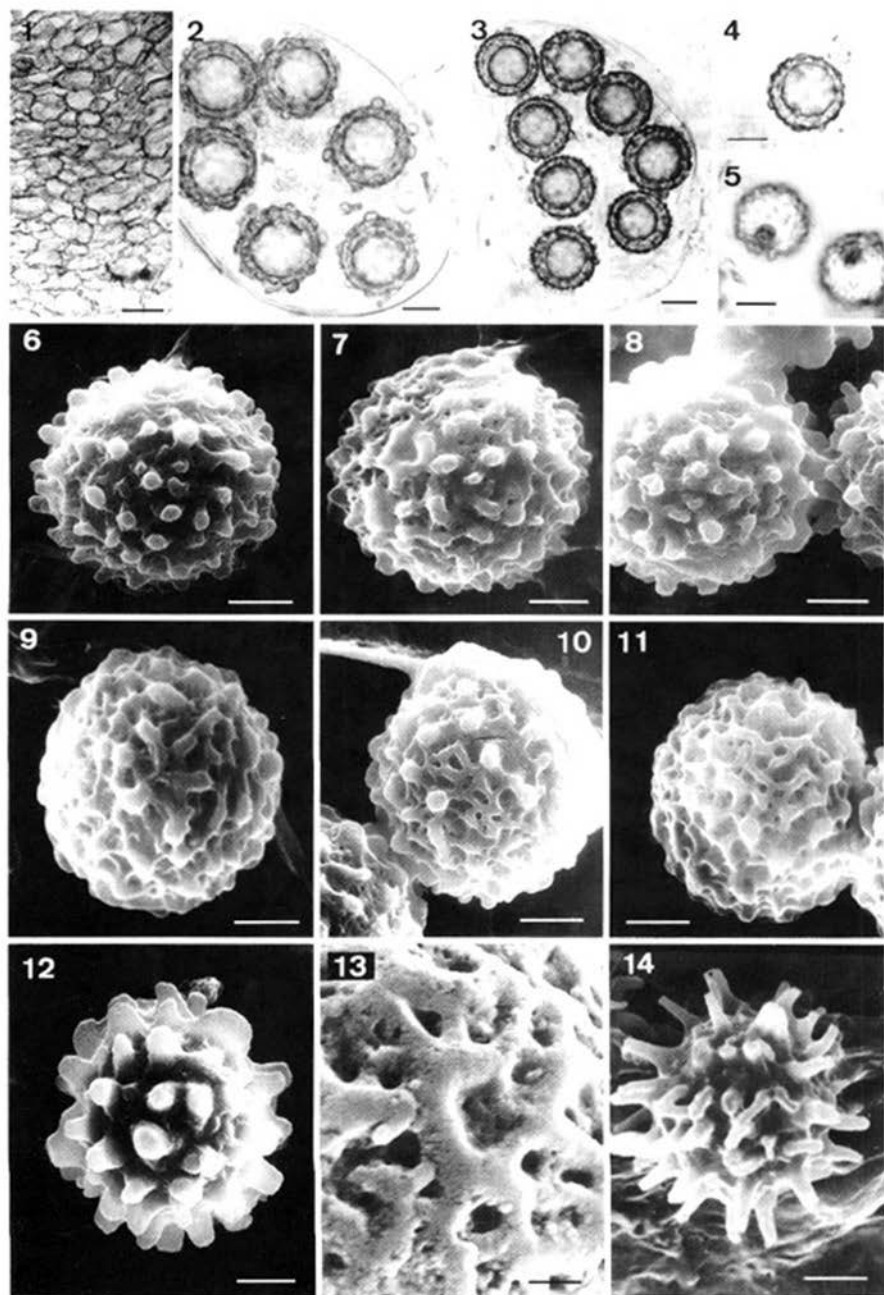
As Moreno et al. (2000) point out, these mycorrhizal fungi play an important role in the maintenance of Mediterranean shrub-lands by preventing erosion and desertification. Hence, it is important to study these fungi and understand their biology and taxonomy more fully. In the present study, we describe a rare and little known species, *Terfezia boudieri* Chatin, not previously reported from Europe. This hypogeous fungus is collected and consumed, without knowledge of its specific identity, in the provinces of Madrid and Toledo on calcareous and gypsiferous soils.

The material from Spain, herein reported, was compared with specimens of *T. boudieri* from Morocco, where this desert truffle is sold in local markets or 'zocos'.

MATERIAL AND METHODS

Microscopic examination and measurement of ascoma elements were carried out in tap water, 10% aqueous NH₄OH, Melzer's reagent and Congo-Red. The photomicrographs were made with a Nikon microscope (Optiphot model), with phase-contrast and automatic photography. The SEM photographs were taken with a Zeiss DSM-950 microscope.

Voucher specimens were deposited in AH (herbarium of Alcalá University, Spain).



Terfezia boudieri Chatin — Figs. 1–11

Terfezia boudieri Chatin, Compt. Rend. Acad. Sci. Paris 113 (1891) 530.

Ascomata hypogeous, isolated, 4–9 cm diam., subglobose, obconical, irregular, depending on whether soils are more or less sandy, with a basal cord or attachment formed by sand and mycelium and connected to roots of vascular plants. Peridium smooth, with adherent sand, brown-red to brown, reminding of potato peels. Gleba yellow-brown with red tones, at maturity with red-brown fertile pockets, surrounded by paler off-white infertile veins. Smell not distinct.

Peridium composed of parenchymatous cells with thick walls. Asci 56–90 µm in diam., globose, (3–4)–6–8-spored, when immature with a short wide pedicel, non amyloid. Spores 18–22 µm diam. (ornamentation included), globose, pale yellow-brown, with a large guttule. Ornamentation 1–3 µm thick and variable depending on maturity. At first reticulate, then more or less warty with large obtuse warts often with a truncate tip, similar to *T. arenaria* (Fig. 12) but with abundant small warts. At maturity with short crests and wide rounded warts over a reticulum, non amyloid.

Collections studied. SPAIN: Fuentidueña del Tajo (Madrid), gypsum shrub-land, associated with *Helianthemum squamatum* (L.) Dum. Cours., March 1997, B. Anta & A. Fernández, AH 22364; Valdeguerra, Colmenar de Oreja (Madrid), on calcareous gypsiferous soil together with *Picoa lefebvrei* (Pat.) Maire, and associated with *Helianthemum salicifolium* (L.) Mill., 18 Apr. 1999, J. Cámara, G. Moreno, K. Kreisel & H. Kreisel, AH 19743; Cabezamesada, Toledo, 2 May 1999, E. Rubio, AH 19581. — MOROCCO: Market of Tanger, 19 May 1997, Laghzaoui Rabie, AH 22363, duplicate in Montecchi Herbarium.

Additional collections studied. (1) *Terfezia arenaria* (Moris) Trappe. SPAIN: Navalmoral de la Mata (Cáceres) associated with *Helianthemum guttatum* (L.) Miller, March 1995, J.L. Manjón & J. Díez, AH 22331 (Fig. 12). (2) *Terfezia claveryi* Chatin. SPAIN: Guadix (Granada) with *Helianthemum salicifolium* (L.) Mill., 30 Apr. 1977, A. Ortega, AH 3813 (Fig. 13). (3) *Terfezia leptoderma* Tul. & C. Tul. SPAIN: Trujillo (Cáceres) with *H. guttatum* (L.) Mill., 26 Feb. 1977, leg. J.M. Hernández, AH 1809 (Fig. 14).

Terfezia boudieri occurs on basic, either calcareous soils or gypsiferous marl. It is characterized by large ascomata, and spores bearing warts over a reticulum.

Terfezia claveryi also occurs in such soils, and is macroscopically very similar. Microscopical examination is required to differentiate one from another based on spore ornamentation. *Terfezia claveryi* has reticulate spores without warts (Fig. 13), whereas the spores of *T. boudieri* have warts over a reticulum (Figs. 2–11).

On acid soils, two other species fruit, namely *T. arenaria* and *T. leptoderma*. Although both acidophilous species occur on sandy sites associated with the same host, they are easy to distinguish from each other. The former has a pale brown gleba with pink tones at maturity while *T. leptoderma* has green-olive tones to its gleba at maturity. These species are also very different in microscopy, the spores of *T. arenaria* bearing warts with obtuse apices (Fig. 12), whereas spores of *T. leptoderma* have long spines (Fig. 14) (Moreno et al., 1986).

←

Figs. 1–11. *Terfezia boudieri* (1–5: AH 19743; 6–11: AH 19581). 1. Parenchymatous cells of ascoma peridium (MO); 2. 6-spored ascus; 3. 8-spored ascus; 4 & 5. detail, spore ornamentation (MO); 6–11. spores showing variation in ornamentation (SEM), from warty over a weak reticulum to reticulate with scattered warts. — 12. *Terfezia arenaria* (AH 22331). Spore with large truncate warts (SEM). — 13. *Terfezia claveryi* (AH 3813). Spore with complete reticulum without warts (SEM). — 14. *Terfezia leptoderma* (AH 1809). Spore with long spines (SEM). — Scale bars: Fig. 1 = 20 µm; Fig. 2–5 = 10 µm; Figs. 6–12 & 14 = 5 µm; Fig. 13 = 1 µm.

There is an excellent iconography of *T. boudieri* by Ceruti (1960), who reported this fungus as a common truffle in northern Africa. However, Malençon (1973) curiously did not mention this species in his broad treatment of North Africa. Pacioni & El-Kholy (1994) described *T. boudieri* from Egypt, and provided the first SEM microphotographs of the spore ornamentation.

In certain areas of the Iberian Peninsula (i.e. the provinces of Madrid and Toledo) *T. boudieri* is consumed by the locals. In certain favourable years, large amounts of this desert truffle are collected. In these regions, it is called 'criadilla or turma', the same name used to refer to the most abundant and commonly collected species in other regions of Spain (i.e. Extremadura, western Spain), namely *T. arenaria*.

Until now, *Terfezia boudieri* has been known to occur only in North Africa and West Asia. It had never been reported from southern Europe, though it was likely misidentified as other *Terfezia* spp. because of its variable spore ornamentation. We observed that the features of the ornamentation depend on the spore maturity (Figs. 6–11). At first, the spores are reticulate with scattered small warts, hence it is easily confused with *T. claveryi*. Later, spores become warty as in *T. arenaria*, but with numerous small warts. Furthermore, we believe the figure published by Montecchi & Lazzari (1993: 214), showing spores with rounded warts over a reticulum, likely represents *T. boudieri* instead of *T. claveryi*.

In the Iberian Peninsula, *T. boudieri* occurs on basic soils associated with *Helianthemum* spp., either with annual species such as *H. ledifolium* (L.) Miller or *H. salicifolium* (L.) Miller or with perennial species such as *H. squamatum* (L.) Dum. Cours., the last one a gypsophilous plant endemic to the Iberian Peninsula.

As Moreno et al. (2000) point out in a previous work, the Iberian Peninsula is located between the African and the European continents. Moreover, these regions share a heliophilous flora and the associated mycota, which suggests this region may play an important role in the migration of mycorrhizal fungi between both continents, particularly of those species associated with *Cistaceae*.

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**DIDERMA CRISTATOSPORUM, A NIVICOLOUS MYXOMYCETE
FROM SPAIN**

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A new species of nivicolous myxomycetes, *Diderma cristatosporum* is described from Spain and compared with the type of *D. subdictyospermum*. LM and SEM photographs of the microscopical characters are provided.

Myxomycetes growing near melting snow have not been well studied in Spain. The first reports were published by Gràcia (1986, 1987), and subsequently by Lado (1992) and Illana et al. (1993) who reported new records from the Sierra de Guadarrama in Central Spain (province of Segovia). Gorris et al. (1999) published some species which fructify in meadows and clearings in the East Pyrenean alpine and higher subalpine belts of the Catalan Pyrenees.

In the present paper we describe a new nivicolous myxomycete species which was also found in the Sierra de Guadarrama (Segovia).

The material studied is deposited in the herbarium of the University of Alcalá (AH).

Images using scanning electron microscopy (SEM) were made following Castillo et al. (1998). The descriptions of spore ornamentation under SEM follow the terminology proposed by Rammeloo (1974, 1975).

***Diderma cristatosporum* A. Sánchez, G. Moreno & Illana, spec. nov. — Figs. 1–10**

Sporocarpia in gregibus, 0.7–1.6 mm diametro, globosae vel subglobosae, sessilia. Peridium duplex, stratum externum albidum, crustaceum, stratum internum membranaceum, hyalinum cinereum. Columella magna, convexa vel hemisphaerica, ferruginosa.

Capillitium 2–5 µm diametro, abundans, fuscum, ramosum, apicibus distinctis. Sporae 12–15 µm, globosae, translucidae, irregulariter coloratae, griseae vel hyalino-griseae, cristis irregularibus sinuosis incrustatae.

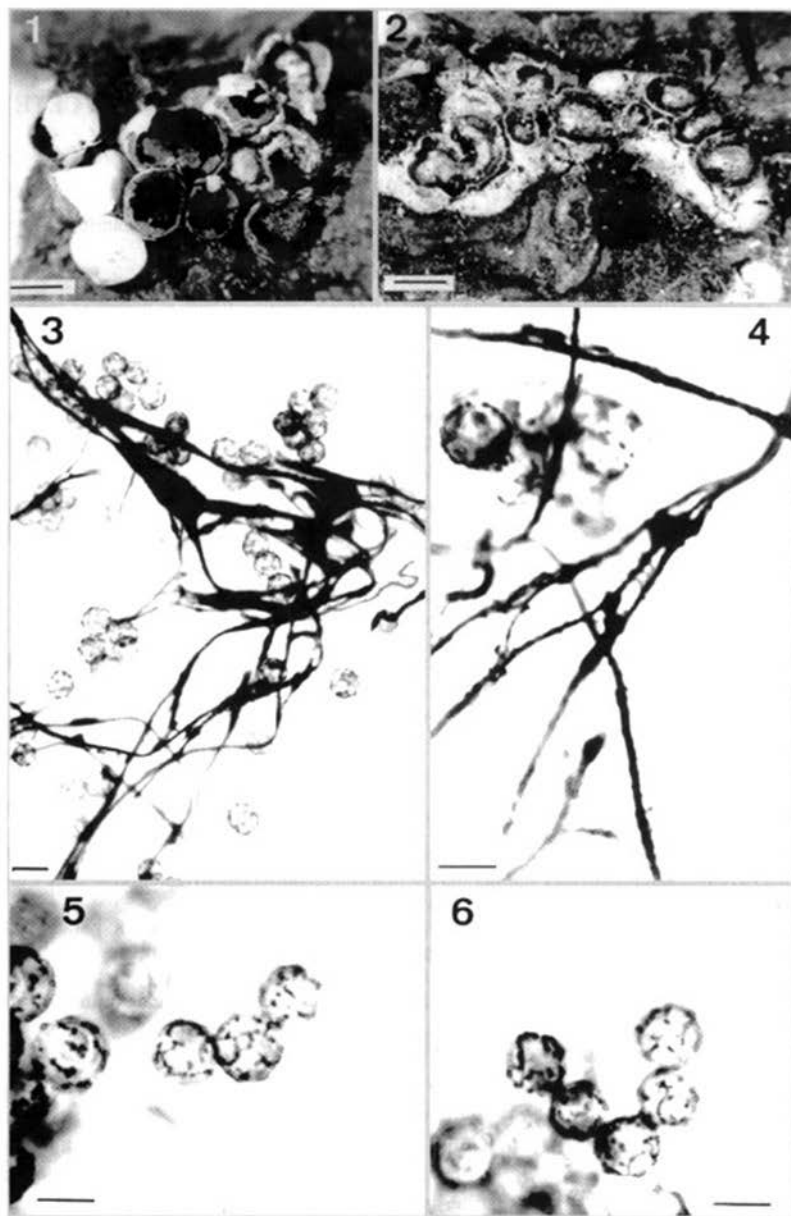
Holotypus: Hispania, Puerto de Navafría 1800 m, Segovia, ad corticem rami emortui *Pini sylvestris* L., 15-V-1997, leg. A. Sánchez, in Herbario AH sub no. 18413 conservatur.

Etymology: Referring to the ridged ornamentation of the spores.

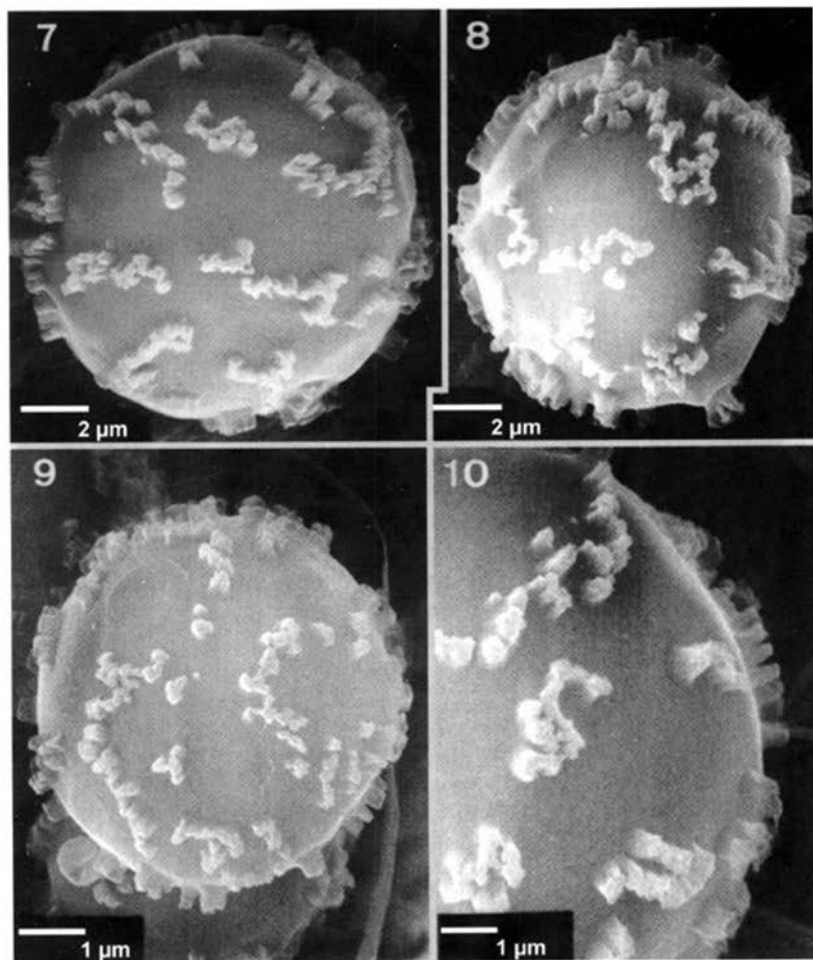
Fructification formed of 15–30 sessile sporocarps. Sporocarps globose to subglobose, some slightly plasmodiocarpous, 0.7–1.6 mm diam. Hypothallus membranous, continuous, whitish with lime incrustation. Peridium clearly double; outer layer very fragile, thick, smooth, irregularly dehiscent, white, inner layer membranous, cinereous and closely applied to the spore mass. Columella hemispherical to elongated, rough, reddish brown.

Capillitium abundant, branched and anastomosed, threads 2–5 µm diam., rigid, flexuous, often with many irregular swellings and membranous expansions, dark brown and some-

1) Corresponding author.



Figs. 1–6. *Diderma cristatosporum*, holotype (AH 18413). 1 & 2. Sporocarps; 3 & 4. capillitium and spores under LM; 5 & 6. spores under LM. — Scale bars: Figs. 1 & 2 = 1 mm; Figs. 3–6 = 10 μ m.



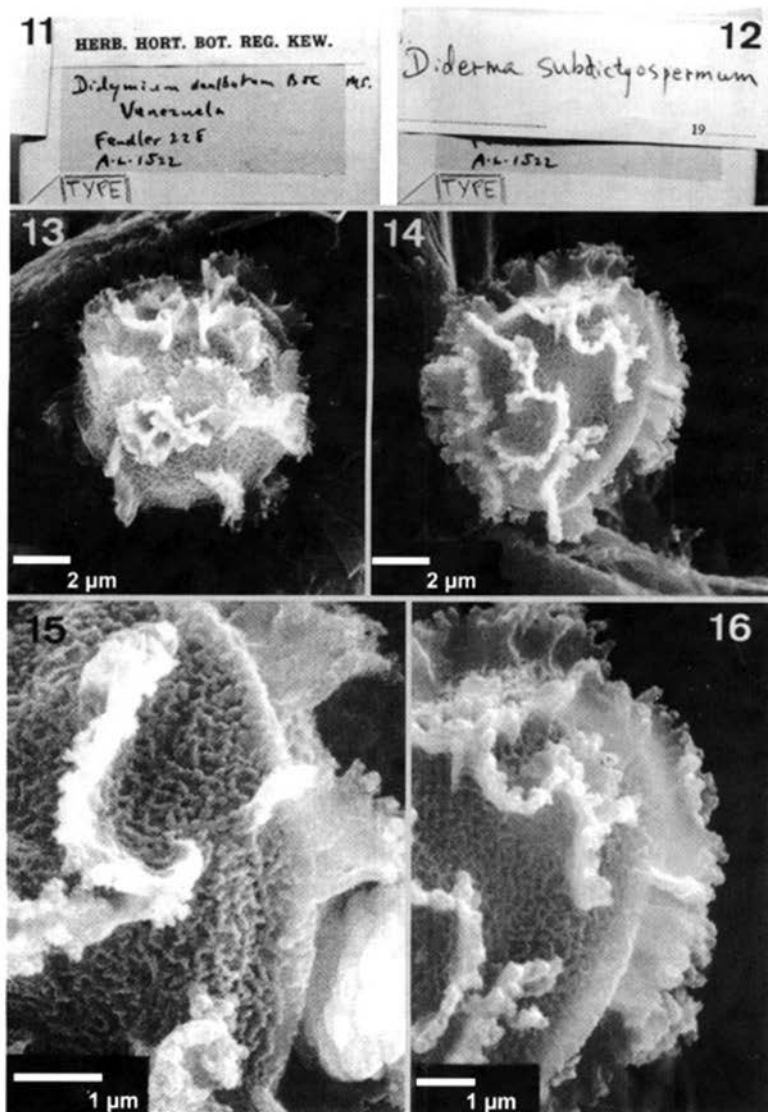
Figs. 7–10. *Diderma cristatosporum*, holotype (AH 18413). 7 & 8. Spores under SEM; 9 & 10. detail of spore ornamentation under SEM.

times colourless at the extremities. Spores free, dark brown in mass, pale purple-grey in the light microscope, globose, 12–15 μm diam., with small, scattered ridges; when observed by SEM, the spore has a smooth surface and the ridges are formed by long bacula united to form sinuous lines.

Habitat — On fallen branches of *Pinus sylvestris* near melting snow.

Distribution — Known only from Spain (province of Segovia).

Collections studied. SPAIN: Segovia, Puerto de Navafría, 1800 m, on bark of dead branches of *Pinus sylvestris* L., 15-V-1997, A. Sánchez, AH 18413 (holotype) and AH 19557.



Figs. 11–16. *Diderma subdictyospermum*, holotype (Fendler 228 in K). 11 & 12. Box and labels of the type; 13 & 14. spores under SEM; 15 & 16. detail of spore ornamentation under SEM.

Diderma cristatosporum is characterized by its sessile sporocarps, double peridium, reddish brown, hemispherical to elongated columella, thick dark brown capillitium and spores with small ridges.

Other *Diderma* species described with reticulate or subreticulate spores are: *Diderma subdictyospermum* (Rostaf.) G. Lister, *D. reticulosporum* Nann.-Bremek., Mukerji & Pasricha and *D. diadematum* Schokn. & J.L. Crane.

Diderma subdictyospermum was originally described from Venezuela, growing on dead leaves and moss. We have studied the type material deposited in Kew (Figs. 11–16): the ornamentation of the spores is different from that of *D. cristatosporum*, viz. very marked ridges and a reticulum on the spore surface that is visible only with SEM.

Diderma reticulosporum described from India by Nannenga-Bremekamp et al. (1984) has stipitate sporocarps, a short, cylindrical columella and subovoid to subglobose spores with an irregular reticulum.

Diderma diadematum described by Schoknecht & Crane (1978) from Illinois (USA) was obtained from moist-chamber cultures of submerged leaf litter of *Acer* sp. and *Taxodium distichum* (L.) Rich. This species possesses sessile sporocarps with a double peridium (the outer peridium is white and often incompletely covers the inner layer), no columella and globose spores, (11–)12–13(–15) μm diam., with large spines arranged in an apparently subreticulate pattern.

ACKNOWLEDGEMENTS

We wish to express our thanks to the Keeper of the Kew Herbarium (K) for the loan of the type of *Diderma subdictyospermum*. We also thank Mr. J.A. Pérez and Mr. A. Priego of the SEM Service of the University of Alcalá for their assistance, and Mr. D.W. Mitchell for revising the English text.

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**COPROTUS ARDUENNENSIS, A NEW SPECIES OF
COPROPHILOUS DISCOMYCETES (PEZIZALES, ASCOMYCOTA)**

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A new coprophilous species of *Coprotus* (Pezizales, Pyronemataceae) is described and reported from five localities of the Ardennes (Belgium). Within the genus, it belongs to a group of species mainly identified by the presence of carotenoids within the paraphyses. It could not be identified as any of the 25 species known, but is closest to *C. ochraceus*.

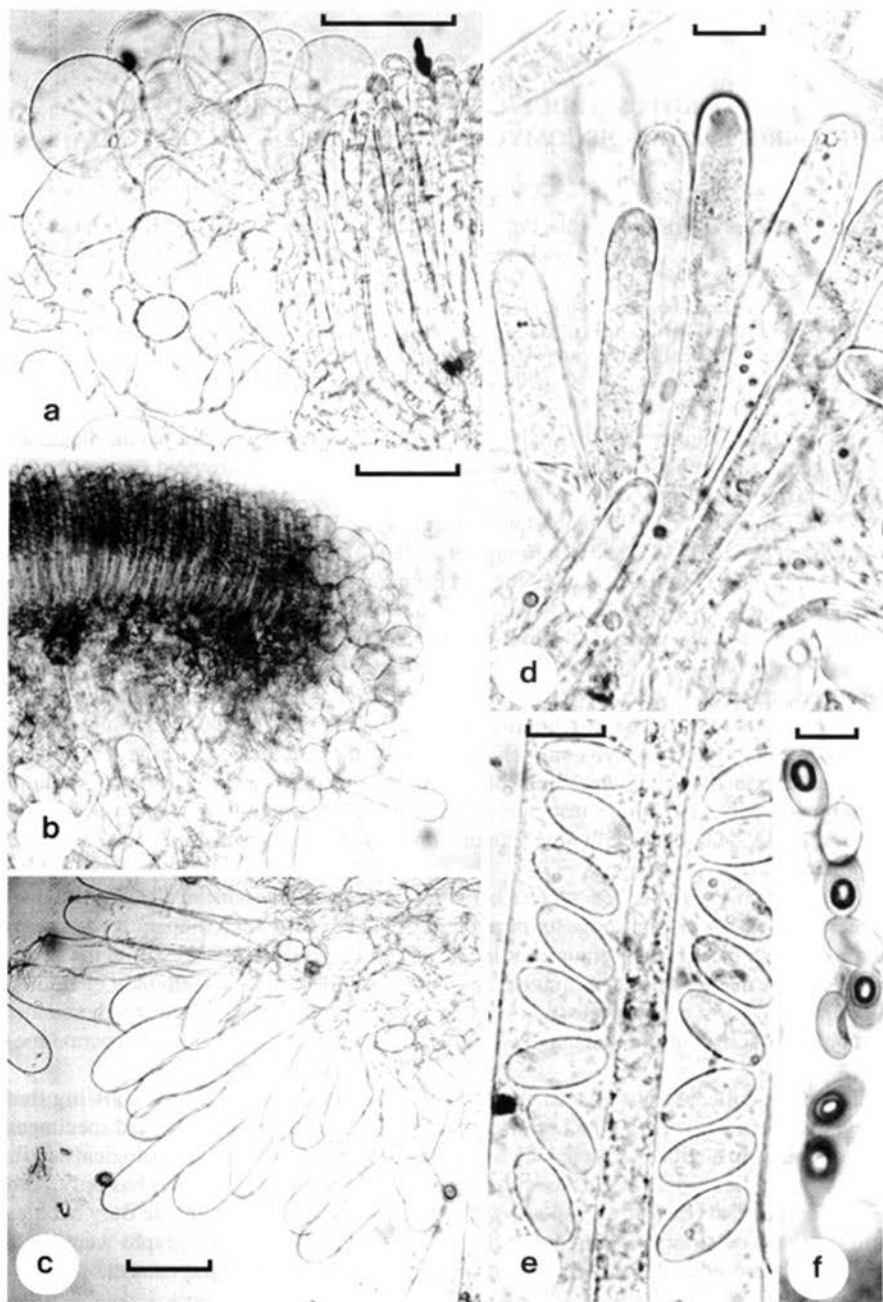
Coprotus Korf & Kimbr. was first suggested by Korf (1954) who later drafted the diagnosis of the genus to comprise mainly coprophilous species with non-amyloid 8-spored asci, smooth, hyaline ascospores and uncinat paraphyses i.e. species traditionally placed within the old heterogeneous *Ascophanus* Boud. (Korf, 1958). Subsequently Kimbrough (1966), Kimbrough & Korf (1967) and Kimbrough et al. (1972) extended the limits of the genus to species with multispored asci, some extracted from *Rhyarobius* Boud., admitting that their paraphyses are not strictly hooked and adding new discovered species. For an up-to-date circumscription of the genus, see Van Brummelen (1998: 427).

In the basic paper of Kimbrough et al. (1972) eighteen species were combined, shortly described and keyed. Moravec (1971), Bell & Kimbrough (1973), Jeng & Krug (1977), Thind et al. (1978), Gibson & Kimbrough (1980), Gené et al. (1993) and Wang (1994) added seven more species. No comprehensive study has been made to assess the validity of the 25 described species, which more often than not are quite obviously difficult to delimit (Kimbrough et al., 1972). For instance, a recent study on coprophilous Pezizales in Italy (Doveri et al., 2000) deals with nine taxa of *Coprotus* of which two could not be assigned to previously described species.

Before dealing with the new species hereafter described we produced a key (not shown here) to the best of our knowledge for taxa referred to *Coprotus*¹. This comparative approach confirmed that the informal groups outlined by Kimbrough et al. (1972) within the genus still hold but that specific delimitations are hard to pin down on the grounds of limited descriptions of most of the species involved. Nevertheless, the material described hereunder combines sufficient differential diagnostic characters to be distinguished as a separate species.

The dung collected was placed in moist chambers for several weeks knowing that *Coprotus* species are among the last discomycetes to appear. Freshly collected specimens were mounted in distilled water in which all measurements were made. Histological details have been studied from freehand sections. Cotton blue in lactic acid (CB) has only been tested for its effects on ripe ascospores with non-elastic walls developing de Bary bubbles artifact. Freehand drawings were made for the illustrations. Photomicrographs were made with light microscopy with a Leitz microscope using a 35 mm Olympus camera.

1) This key and comments on the species involved will be published in the near future.



Coprotus arduennensis J.R. De Sloover, spec. nov. — Figs. 1, 2

Apothecia discoidea, sessilia, dispersa; discus aurantius 0.5–1.5 mm diametro, scabriusculus ob protrudentes ascos. Superficies externa glabra, alba vel pallide lutea. Margo alba, parum prominens. Excipulum ectale textura globulosa, cellulis valde inflatis, 10–45 µm diam. praecipue ad marginem; cellulae marginales valde elongatae praesertim ad basim, usque 200 µm longae. Asci octospori 150–185 × 10–16 µm, late cylindracei vel cylindrico-clavati, inferne leviter attenuati. Ascospores 12.5–15.5 × 6.5–7.5 µm, ellipsoideae, uniseriales, hyalinae, levigatae. Paraphyses cum multis parvis aurantiisque guttulis, cylindraceae omniouque erectae, comparate crassiusculae, inferne septatae et raro ramosae, 6–9 µm diam., raro ad apicem leviter incrassatae (10 µm) obtusaeque.

In fimo fero porcino (*Sus scrofa*) crescens, in calluneto cum sphagnis (*Vaccinietum*).

Typus: *J. De Sloover 00C9*, Vielsalm, A Sacrawé, Belgium, 23.III.2000, (holotypus: herb. *J. De Sloover*; isotypus L 998.171-667).

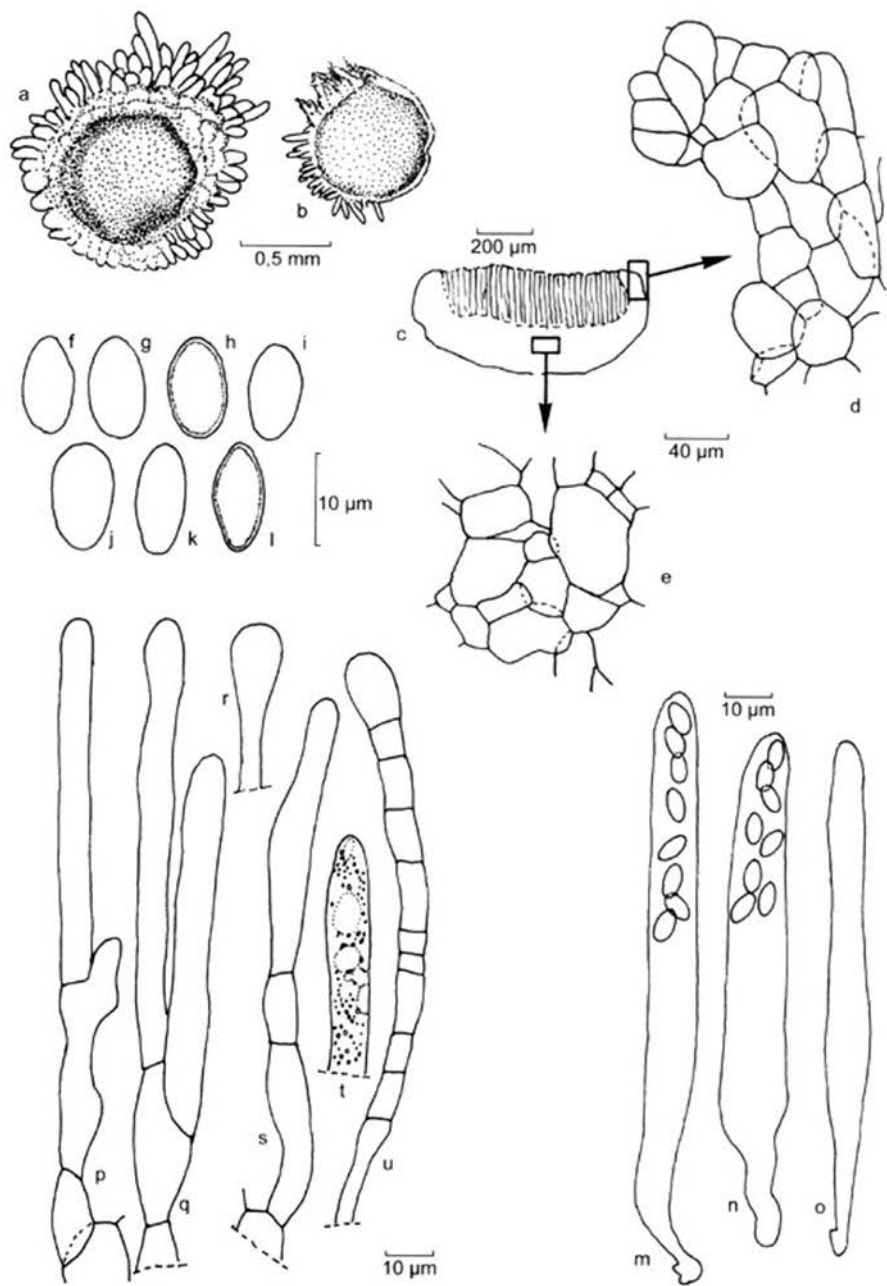
Etymology: from Latin, from the Ardenne country.

Apothecia discoid, superficial, scattered, sessile on an obconical base, 0.5–1.5 mm across, 0.5–0.7 mm high. Receptacle at first subglobular, then expanding and finally high saucer-shaped, light orange turning whitish when dry, smooth with a narrow and rough margin slightly raised above the disk. Disk flat, bright orange, roughened by the slightly protruding asci or paraphyses varying greatly in length, remaining deep orange upon drying. Hymenium 250–400 µm thick. Cortical excipulum 250–300 µm thick near the base, of closely compacted isodiametric globose cells 10–45 µm across, spreading into the lower and then the upper flank made of isodiametric, subglobular-subangular cells 30–40 µm across (textura globulosa to angularis), or oblong cells up to 200 µm long with thin hyaline walls near the margin. Margin made of inflated globular cells causing its rough bumpy appearance. Asci cylindrical or cylindrical clavate gently tapering upwards and downwards from a maximum width below the middle, (150–)160(–185) × (10–)12.8(–16) µm, rounded above, 8-spored (but often with only some of the spores fully developed or rarely 16-spored), the wall not blue in Melzer's reagent. Ascospores uniseriate or rarely biseriate, ellipsoid, hyaline, smooth, (12.5–)14.5(–15.5) × (6.5–)6.8(–7.5) µm. Paraphyses simple or branched below the long upper cell, strictly cylindrical and straight, rather thick, 6–9 µm wide from base to tip (10 µm), 3 or 4-septate (exceptionally more septate), the upper cell (60–)80(–100) µm long, with numerous tiny (up to 1 µm) orange plasmatic oil guttules and large vacuoles.

Habitat — On dung of wild boar in wet heathland.

Specimens examined. BELGIUM: Luxemburg Province, Vielsalm, A Sacrawé (alt. 585 m), on wild boar dung in wet *Vaccinietum* heathland, 23.III.2000, *J. De Sloover 00C9* (holotype of *Coprotus arduennensis*, herb. *J. De Sloover*; isotype L 998.171-667); Petit-Thiers, Grand Fond (alt. 440 m), on deer dung in wet *Molinietum*, 15.VII.2000, *J. De Sloover 00C106*. Liège Province, Büllingen, Holzwarche (alt. 640 m), on wild boar dung in peaty *Molinia* grassland with *Coprotus leucopocillum* Kimbrough et al., *Ascobolus michaudii* Boud., 04.VII.1999, *J. De Sloover 99C111*; Biron, on deer dung in a spruce afforested site, 15.X.2000, *J. De Sloover 00C139*. Namur Province, Oignies-en-Thiérache, Trieu des Cavaliers (alt. 360 m), on deer dung in birch-oak wood with *Ascozonus woolhopensis* (B. & Br.) Boud., *Ascobolus furfuraceus* Pers., *A. albidus* Crouan and *Thelebolus stercoreus* Tode, 20.VIII.2000, *J. De Sloover 00C114*.

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Fig. 1. *Coprotus arduennensis*. a. Longitudinal median section through ripe apothecium: raised margin of bullate cells (left) and hymenium; b. median section through ripe apothecium: hymenium and ectal excipulum with bullate cells and elongated cells on the lower flank; c. lower flank excipulum with external elongated cells; d. paraphyses with granular content and refringent guttules; e. ascospores in two asci flanking one paraphysis; f. ascus with ascospores showing de Bary bubbles in CB. a–c, e: *De Sloover 00C9*, holotype; d & f: *De Sloover 99C111*. — Scale markers: a & c = 50 µm; b = 100 µm; d–f = 10 µm.



Criteria commonly used to delimit species of *Coprotus* are both quantitative and qualitative: apothecia colour and size, excipulum extent, ascus and ascospore form and size, paraphyses form and content. Depending on the relative importance given to these criteria the resulting classification may be quite different.

De Bary bubbles are consistently induced in *Coprotus* spores by the use of special mounting media and they have been regarded as one of their taxonomic features (Kimbrough et al. 1972). To our mind those artifacts created by mounting the spores, e.g. in cotton blue in lactic acid, must only be viewed as a clue to the thickness or rigidity of the spore walls (Baral, 1992): they are produced in the same conditions in quite different taxa such as Pyrenomataceae like *Pulvinula* (pers. obs.) and *Pseudombrophila* (van Brummelen, 1995) or Thelebolaceae like *Coprotiella* (Jeng & Krug, 1976).

Quantitative criteria, like spore and ascus size, are highly variable in species of *Coprotus* from one sample or from one author to another. For *C. ochraceus* (H. & P. Crouan) J. Moravec for instance, ascospore length is reported as 13.5–17.5 μm (Aas, 1983) and as 15–18 (Moravec, 1971; Ellis & Ellis, 1998). Such data are difficult to handle where means are not mentioned. For *C. arduennensis* one of the collections (*J. De Sloover 99C111*) had smaller ascospores (9–)10.5(–13) μm , but agreed with the type in all other aspects: that apparently aberrant sample could perhaps indicate a slightly deviating taxon of which the delimitation is not yet clear.

Sizes of asci and apothecial diameter also vary greatly. Quantitative data are so variable, and simple statistical analysis should be considered essential when describing new taxa but it is far from being a common practice.

Presence vs. absence of carotenoid pigments was proposed by Kimbrough et al. (1972) as a first order qualitative criterion to found groups within the genus. Yellow to bright orange colours are frequently noticed in the hymenium of some species of *Coprotus*, particularly in the paraphyses in granules or oil droplets. Similar but duller shades have been detected both in the ascospores and in the excipulum cells and cell walls. It should be noted that the orange colour of the hymenium, as described for instance in *C. aurora*, is always the sign of carotenoid bound lipids in the paraphyses. On the other hand, yellow or yellowish shades are mostly linked to excipulum wall pigments or to refringent droplets, e.g. in paraphyses. These quite different origins of colour are not to be confused. In Table 1 the 26 known species are set out so that species sharing the same characteristics are brought together. The group with lipid bound orange-yellow pigments in paraphyses is limited to seven among the 26 species listed; most of them have ascospores with light yellowish contents, as well as a small amount of carotenoids in the excipulum mainly on cell walls. Obviously the greatest amounts of carotenoids are located in the paraphyses. It should be noted that this small group of seven species has eight-spored asci. Furthermore, the last nine species listed are completely devoid of any yellow pigments. They usually possess translucent to white apothecia except sometimes when drying. Here also are most of the multisporied species.

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 Fig. 2. *Coprotus arduennensis*. a, b. Habit of fruit-bodies from above; c. median section of an apothecium showing the location of d and e; d. hypothecium cells (textura angularis); e. ectal excipular cells; f–l. ascospores with two (h, l) in optical section; m–o. asci with uniseriate to irregularly biseriolate spores; p–u; paraphyses straight and thick, rarely branched towards basis (p, q) or clavate at apex (r), exceptionally septate (u) with small refractive golden yellow droplets (t); a, b: *De Sloover 00C114*; c–u: *De Sloover 99C111*.

Concerning pigmentation *Coprotus arduennensis* seems related to the group of the other six species with golden yellow droplets in the paraphyses, i.e. *C. breviascus*, *C. vicinus*, *C. aurora*, *C. luteus*, *C. baeosporus*, and particularly *C. ochraceus*.

In *Coprotus* the excipulum is as a rule poorly developed: this is particularly true in species like *C. disculus* (Thind et al., 1978), yet in a few others like *C. baeosporus* (Jeng & Krug, 1977) or *C. sarangpurensis* (Thind et al., 1978) it is rather well differentiated with medullary and ectal layers. Moreover, the hypothecium is absent or at least indistinct as reported, e.g. in *C. ochraceus* (Thind et al., 1978). Even though well differentiated, the excipulum thickness and the number of its layers are only reported for seven species out of 25, from two layers in *C. baeosporus*, up to a maximum of four in *C. vicinus* and up to five or six layers in *C. sexdecimsporus* (Kimbrough et al., 1972). Ectal excipulum thickness goes from 50–95 µm, while the medullary excipulum could reach 425 µm as reported in *C. ochraceus*. While basal and medullary cells are usually isodiametric or slightly elongated [8–30 µm, except in *C. ochraceus* where they may reach 55 µm (Thind et al., 1978)], cells of the ectal excipulum along the margins are clearly elongated in most species reaching up to 100 µm in *C. marginatus*. In this respect *C. dhofarensis* is the only one of its kind, where most top-

Table I. Location of carotenoids in selected cells or walls of species of *Coprotus*: (+) = in small amount, + = present, ++ = in large amount.

	Paraphyses with yellow oil gut-tules	Ascospores slightly yellowish	Excipulum walls and/or cells yellow
<i>C. lacteus</i> (Cooke & W. Phillips) Kimbr., Luck-Allen & Cain ⁵			
<i>C. glaucellus</i> (Rehm) Kimbr. ⁵			(+)
<i>C. marginatus</i> Kimbr., Luck-Allen & Cain ⁵			(+)
<i>C. granuliformis</i> (P. & H. Crouan) Kimbr. ⁵			(+)
<i>C. dextrinoideus</i> Kimbr., Luck-Allen & Cain ⁵			+
<i>C. dhofarensis</i> Gené, ElShafie & Guarro ⁸			+
<i>C. leucopocillum</i> Kimbr., Luck-Allen & Cain ⁵		(+)	
<i>C. duplus</i> Kimbr., Luck-Allen & Cain ⁵		(+)	(+)
<i>C. sexdecimsporus</i> (P. & H. Crouan) Kimbr. ⁵		(+)	(+)
<i>C. disculus</i> Kimbr., Luck-Allen & Cain ⁵		(+)	(+)
<i>C. breviascus</i> (Velen.) Kimbr., Luck-Allen & Cain ⁵	+	(+)	(+)
<i>C. vicinus</i> (Boud.) Kimbr., Luck-Allen & Cain ⁵	+	(+)	(+)
<i>C. aurora</i> (H. & P. Crouan) Kimbr., Luck-Allen & Cain ⁵		++	(+)+
<i>C. ochraceus</i> (H. & P. Crouan) Moravec ^{1,5}	++	(+)	(+)
<i>C. arduennensis</i> J.R. De Sloover	++		
<i>C. luteus</i> Kimbr., Luck-Allen & Cain ⁵	++		
<i>C. baeosporus</i> Jeng & Krug ⁴	++		
<i>C. sphaerosporus</i> Gibson & Kimbr. ³			
<i>C. niveus</i> (Fuckel) Kimbr., Luck-Allen & Cain ⁵			
<i>C. rhyarobioides</i> (Heimerl) Kimbr. ⁵			
<i>C. winteri</i> (E. Marchal) Kimbr. ⁵			
<i>C. albidus</i> (Boud.) Kimbr. ⁵			
<i>C. sarangpurensis</i> K.S. Thind S.C. Kaushal ⁷			
<i>C. trichosurus</i> A.E. Bell & Kimbr. ²			
<i>C. uncinatus</i> Y.Z. Wang ⁹			
<i>C. subcylindrosporus</i> J.M. Moravec ⁶			

1) Aas, 1983; 2) Bell & Kimbrough, 1973; 3) Gibson & Kimbrough, 1980; 4) Jeng & Krug, 1976, 1977; 5) Kimbrough et al., 1972; 6) Moravec, 1971; 7) Thind et al., 1978; 8) Gené et al., 1993; 9) Wang, 1994.

cells on the upper flank of the ectal excipulum elongate upwards forming a fringe of long cells exceeding the surface of the hymenium. Here the marginal structure is far more developed than the one described for *C. marginatus*, but its cells look like the nearby paraphyses and the ones described here for *C. arduennensis*, while *C. dhofarensis* paraphyses have a size typical of those in *Coprotus*. *Coprotus arduennensis* shows a well-differentiated excipulum with globose isodiametric basal cells up to 45 µm, which are of about the same order of size as the largest ones that are known in the genus, in *C. ochraceus*. The sizes of cells at the excipular margin in *C. arduennensis* (exceeding 200 µm) are even larger than the largest reported thus far, in *C. marginatus*. The cells of the raised margin are also considerably enlarged in *C. arduennensis*. It appears that *C. marginatus*, *C. dhofarensis* and *C. arduennensis* form a group of three species where the marginal cells are particularly well developed in different ways.

Thus *C. arduennensis* shares the same characteristics with the six species indicated in Table I as having paraphyses with yellow guttules and especially the most richly pigmented *C. ochraceus*, *C. aurora*, *C. luteus* and *C. baeosporus*. Moreover, the pronounced yellow to orange colour of the apothecium is another feature that *C. arduennensis* has in common with *C. ochraceus* and *C. aurora*. The wide straight cylindrical paraphyses, the larger asci, the smaller ascospores and particularly the large swollen excipular cells are consistent, and sufficient to distinguish *C. arduennensis* from these other species.

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**CORTINARIUS ERYTHROFUSCUS (SUBGENUS TELAMONIA,
SECTION FIRMIORES), A NEW SPECIES FROM SPAIN**

R. MAHIQUES¹ & A. ORTEGA²

The new species *Cortinarius erythrofuscus* Mahiques & A. Ortega (subgen. *Telamonia*, sect. *Firmiores*) is proposed. Its morphological characters are compared with similar taxa (e.g. *Cortinarius petroselineus* Chevassut & Rob. Henry and *Cortinarius casimiri* (Velen.) Huijsman). Its taxonomic inclusion in section *Firmiores* (Fr.) Henn. (subgen. *Telamonia* (Fr.) Trog) is discussed.

In the last few years, the region of Valencia (Eastern Spain) has been extensively studied from a mycological point of view, and particularly regarding the genus *Cortinarius*. Some of the results of this study have been published recently (Mahiques & Ortega, 1997; Ortega & Mahiques, 1995b, 1998). In this contribution we propose *Cortinarius erythrofuscus* Mahiques & A. Ortega as a new species, based on material collected by the first author (R.M.) which shows a combination of characters unknown in *Cortinarius*.

Cortinarius erythrofuscus was collected in abundance (about 100 basidiomata), fructifying in a wide area (around 1500 m²) in two different years, 1998 and 1999. The material studied is deposited in the Herbarium of the University of Granada, Spain (GDAC), the Nationaal Herbarium Nederland, Universiteit Leiden branch, the Netherlands (L) as well as in the private herbaria of R. Mahiques (MES) and F. Martinez (FM).

***Cortinarius erythrofuscus* Mahiques & A. Ortega, spec. nov. — Fig. 1**

Pileo 30–70 mm lato, primum forma conica conico-campanulata, postea hemisphaericus aut convexus, umbonatus. Hygrophanus, colore castaneo rubeo aut castaneo nigricante (simile *Cortinarius vernus* H. Lindstr. et Melot), in medio castaneo quasi nigro, marces colore castaneo ochraceo, sed cum variis maculis radialibus quibusdam castaneis nigricantibus. Cuticula levis, non rugulosa, cum paucis albidulis fragmentis veli, plus manifestis ad marginem. Lamellae emarginatae aut uncinatae, ventricosae, raras (4–5/cm), usque ad 1.1 cm latitudinem, colore castaneo ochraceo satis persistente, arista pallida, flocculosa et serrulata. Stipes 35–100 × 4–15 mm, cylindricus cum base attenuata, colore albidulo, sed prompte adipiscens colorem castaneum ochraceum in dimidia superiore et atrorubentem nigricantem in dimidia inferiore, cum fibrillarum albidularum bandis transversis, base tomentosa alba. Caro colore castaneo atrorubente in pileo, atque in parte inferiore stipis, alba, et prompte, fit cinerea in reliquo stipe. Odor levis, selinus aut herbae corrupta et, quondam, parum certus. Sapor inconspicuus.

Sporae 9–13(–14) × 6–7.5 µm, ellipsoideae, sublacriformoides, aut subcylindricae, cum ornamento medio, crestulis anastomosadis praeditae. Pileipellis conformata ex hyphis cum pigmento castaneo ochraceo parietale et intracellulare.

Circiter 50 basidiomata, gregaria aut connata sub *Quercus pyrenaica*, prope Ciruelos del Pinar (Guadalajara). Leg. R. Mahiques. 19.XI.1998. In herbarium GDAC no. 44213 (Holotypus). In Herbarium L (Isotypus). In herbarium MES no. 3351 (Isotypus). In herbarium FM no. 2303 (Isotypus).

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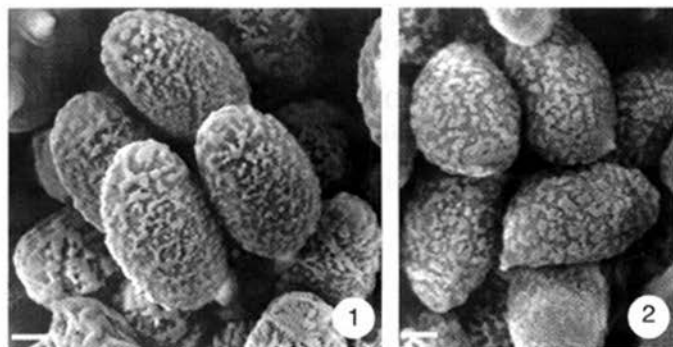


Fig. 1. *Cortinarius erytrophuscus* (holotypus). Spores SEM (bar = 2 μ m). — Fig. 2. *Cortinarius casimiri* (GDAC 42848). Spores SEM (bar = 2 μ m).

Fruit-bodies gregarious to fasciculate or connate. Pileus 30–70 mm diam., at first campanulate-conical, then hemispherical-convex to convex, frequently with a broad umbo, hygrophanous (in most cases, the pileus shows centripetal drying, with some radial narrow bands which remain dark; in other the drying process is simultaneous in margin and disc, retaining a dark concentric zone in between), cigar-brown or dark reddish brown, blackish brown at centre, turning brown-saffron on drying; cuticle smooth, non rugulose, silky, with whitish veil remnants especially at the margin; margin translucently striate. Gills ventricose, sinuate, distant (4–5/cm), moderately wide (0.8–1.1 cm), reddish brown to saffron, with floccose and serrulate whitish edge. Stipe 35–100 \times 4–15 mm, cylindrical, tapering slightly towards the base, whitish when young, quickly turning ochraceous-brown in the upper half, dark reddish brown in the lower half (on handling). Veil whitish, forming some transverse bands on the stipe. Flesh dark reddish brown in the pileus, cortex and lower part of the stipe (similar to *Cortinarius vernus* H. Lindstr. & Melot), brownish in the rest of stipe. Smell mild, reminiscent of parsley or fermented grass (some basidiomata with indistinct smell). Taste mild, sometimes of fermented grass.

Chemical characters — KOH blackish brown on pileus surface, and pileus and stipe context; Phenol-Aniline negative.

Spores (Fig. 1) ($n = 180$) 9–13(–14) \times 5–7.5 μ m; (mean value between 10.0–11.6 \times 5.8–6.7 μ m; average 10.9 \times 6.4 μ m), ellipsoid to amygdaliform, sub-lacrymoid or sub-cylindrical (Q: L/w = 1.4–2; mean value between 1.60–1.72; average 1.66), with a moderately high ornamentation (spore outline serrulate) made up of anastomosing crests. Basidia 4-spored, hyaline or with brownish content. Gill edge heterogeneous, with clavate sterile cells (similar to basidia). Pileipellis formed by a cutis of 3.5–6 μ m wide hyphae, slightly to moderately encrusted with a parietal brownish granular pigment, also with intracellular pigment. Hypodermium with brownish hyphae, 10–20 μ m wide. Hymenophoral trama made up of intracellular and slightly encrusted brownish hyphae. Clamp-connections present in all tissues.

Habitat — On sandy soil under *Quercus pyrenaica*, in mixed forest of *Quercus pyrenaica* and *Pinus pinaster*.

Material studied. SPAIN: Guadalajara, Ciruelos del Pinar, 19.XI.1998, R. Mahiques (GDAC 44213, holotype; L. isotype; MES 3351, isotype; FM 2303, isotype). Idem, 30.X.1999, R. Mahiques (GDAC 44498; MES 3493).

Additional collections studied for comparison

Cortinarius casimiri (Velen.) Huijsman. FRANCE: Arbois, 28.IX.1998, R. Mahiques (MES 3370) [XVI Journées Européennes du *Cortinarius*; under *Fagus sylvatica* and *Quercus robur*]. — SPAIN: Córdoba, near Priego de Córdoba, 18.XI.1996, J. Gómez & B. Moreno-Arroyo (GDAC 42848) [on acid soil, under *Cistus ladanifer*].

Cortinarius petroselinus Chevassut & Rob. Henry. FRANCE: Pezennes, 21.XI.1973 (Herb. Chevassut no. 2333) [Avignon exhibition: on acid soil, under evergreen oaks]; 18.XI.1984, G. Chevassut (Herb. G. Chevassut no. 3362). — SPAIN: Valencia, Pinet, Els Surars, 1.XI.1991, R. Mahiques (MES 1552) [mixed forests of *Quercus suber* and *Pinus halepensis*].

Cortinarius vernus H. Lindstr. & Melot. SPAIN: Granada, Huéneja, Sierra Nevada, 19.V.1996, A. Capilla (GDAC 41046) [1500 m alt., in riparian woods of *Populus nigra* and *Salix atrocinerea*]; Aldeire, Sierra Nevada, 4.IV.1999, A. Capilla (GDAC 44672) [1400 m alt., under *Populus nigra*].

Cortinarius bififormis Fr. SPAIN: Granada, Barranco de S. Sebastián (Natural Park of Sierra de Baza), 7.XII.1990, A. Ortega (GDAC 36716) [under *Pinus sylvestris*].

Cortinarius erythrofuscus is referred to the section *Firmiores* (Fr.) Henn. based on the size of its basidiomata (pileus 3–7 cm diam., stipe 3.5–10 × 0.4–1.5 cm), whitish veil forming silky fibres on the pileus surface, the markedly hygrophanous pileus and spores with moderately high ornamentation. However, *Cortinarius erythrofuscus* can easily be distinguished from other species in this section (e.g. *Cortinarius bififormis* Fr.; *Cortinarius illuminus* Fr.) because of the reddish dark brown basidiomata (essentially the flesh of the pileus and the stipe base), parsley smell and larger spores (Brandrud et al., 1992, 1994).

The parsley smell might suggest a relationship with *Cortinarius rheubarbarinus* Rob. Henry (sect. *Brunnei* Kühner & Romagn. ex Melot). However, the two species are easily distinguished, since *Cortinarius rheubarbarinus* has a different colour and habit, its pileus is hardly hygrophanous and the spores are smaller (Henry, 1956; Brandrud et al., 1992, 1994). Another Mediterranean autumnal taxon with parsley smell is *Cortinarius petroselinus* Chevassut & Rob. Henry (= ? *C. vernus* H. Lindstr. & Melot) (sect. *Erythrini* Melot), which is smaller (pileus 1–4 cm diam., stipe 2–6.5 × 0.2–0.5(–0.6) cm), and has quite different spores (Ortega & Mahiques, 1995a; Ortega & Chevassut, 1998).

Cortinarius casimiri (Velen.) Huijsman (= *C. subsertipes* Kühner, *C. rubellopes* Rob. Henry) has similar spores (Fig. 2) to *C. erythrofuscus*. *Cortinarius casimiri* is – like other taxa of section *Hygrocybe* (Fr.) Nezdobjm. (e.g. *Cortinarius decipiens* (Pers.: Fr.) Fr., *Cortinarius sertipes* f. *contrarius* (J. Geesink) A. Ortega & Mahiques (= *C. sertipes* ss. Kuyper in Arnolds et al., 1995) – smaller, with a pileus of 1–5 cm diameter and a stipe of 2–7(–9) × 0.2–0.5(–0.7) cm, the dark reddish flesh is paler and it lacks the parsley smell (Henry, 1937; Brandrud et al., 1994, 1998).

It should be noted that, because of its macroscopical characters and the large spores, *Cortinarius erythrofuscus* could also be related to some species of section *Uracei* Kühner & Romagn. ex Melot (e.g. *Cortinarius uraceus* Fr., *Cortinarius crassifolius* (Velen.) Kühner & Romagn.). However, the species in this section have darker basidiomata becoming blackish on drying, a yellowish to greenish veil and spores with a very marked ornamentation (Moser, 1983; Brandrud et al., 1990, 1994). Another important character of section *Uracei* is the presence of a distinct greenish pigment in the basidia (Brandrud et al., 1990; Melot, 1990), which is absent in *Cortinarius erythrofuscus*.

Resumen

Se describe una nueva especie *Cortinarius erythrofuscus* Mahiques & A. Ortega (subgen. *Telamonia*, sect. *Firmiores*), recolectado bajo *Quercus pyrenaica* en la provincia de Guadalajara (España). Se compara con otros taxones de caracteres similares como *Cortinarius petroselineus* Chevassut & Rob. Henry y *Cortinarius casimiri* (Velen.) Huijsman. Se discute su inclusión dentro de la sección *Firmiores* (Fr.) Henn. del subgénero *Telamonia* (Fr.) Trog.

ACKNOWLEDGEMENTS

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**GYMNOPUS CASTANEUS, A NEW MEDITERRANEAN SPECIES
FROM SPAIN**

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Gymnopus castaneus, characterized by its reddish-brown colour and lack of hymenial cystidia, is described as new from Spain. In addition, the new combination *Gymnopus brunnescens* (Murrill) Villarreal, Heykoop & Esteve-Rav. is proposed.

Gymnopus castaneus, a new species from central Spain, is described and discussed. On account of its peculiar macro- and microscopical characters combined with the strong farinaceous smell, this new taxon is easily distinguished from other taxa of *Collybia* sensu lato. From a nomenclatural point of view this new species is included in the genus *Gymnopus*, following the treatment of Antonín et al. (1997), though there is still a considerable controversy in adopting this name for many species traditionally recognized within the genus *Collybia* (Bon, 1999).

Colours are given according to the colour code of Munsell (1988).

***Gymnopus castaneus* Villarreal, Heykoop & Esteve-Rav., spec. nov. — Figs. 1–4**

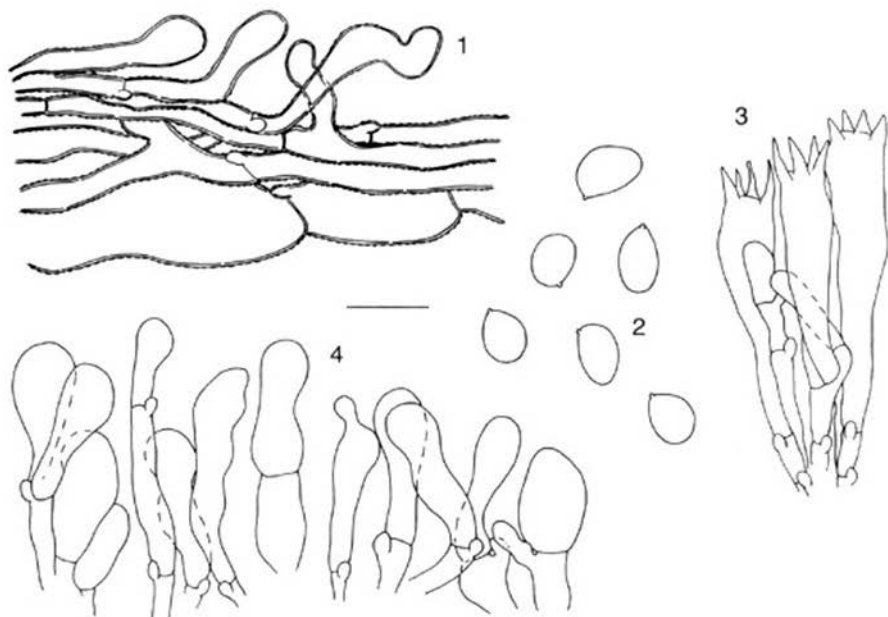
Basidiomata sparsa vel subcongregata. Pileus 3–18 mm latus, ab initio campanulatus vel convexus, dein subinfundibuliformis, haud umboatus, non striatus, toto obscure castaneus, minute velutinus. Lamellae c. 1 mm latae, distantes, emarginatae vel dente-decurrentes, albae, intervenatae. Caro tenuis, alba. Odore saporeque farinaceis. Stipes 8–35 × 1.5–3.5 mm, cylindricus, concolor pileo, toto albo-pruinoso.

Basidia (30–)35–45 × (8.5–)9.5–12 µm, clavata, 4-sporigera, fibulata. Sporae 7.20–8.60–10.30 (–11) × 5.50–6.42–7.65 (–7.70) µm, ellipsoideae vel raro subglobosae, leves, inamyloideae. Cystidia nulla. Trama hymenophori non dextrinoidea, haud in materiam gelatinosam immersae. Pileipellis ex elementis dermatocystidiformibus fasciculatis pseudohymeniformibus vel haud hymeniformiter efformata; dermatocystidia numerosa, cylindracea, cylindraceo-flexuosa vel subclavata, usque ad 55 × 6–9 µm, ascendentes vel suberectae. Hyphae et dermatocystidia pigmento luteolo incrustata. Caulocystidia – 90 × 5–10 µm, descendentes ad basim, dermatocystidiis similia. Hyphae stipitipellis 3–6 µm latae, non dextrinoidea, leves vel raro diverticulatae. Hyphae vasculares praesentia sed dispersae.

Holotypus: Spain, Toledo, Pinar de Almorox, 19 Nov. 1996, F. Esteve-Raventós, C. Sánchez & M. Villarreal (AH no. 21520).

Etymology: referring to the colour of the pileus and stipe.

Basidiomata collybioid to subomphalioid. Pileus 3–18 mm in diam., campanulate to convex when young, then plano-convex with depressed centre, finally infundibuliform, without apical papilla, margin somewhat exceeding the lamellae, at first involute, then inflexed to straight, dry, slightly hygrophanous, not striate, weak red to dusky red (Mu. 10 R 3/4, 4/4), with margin pale red (10 R 5/6), glabrous to fairly sericeous. Context whitish to pale red (10 R 6/4) under the cuticle, not darkening. Smell and taste slightly farinaceous. Lamellae 19–25, c. 1 mm wide, distant, deeply emarginate, with decurrent tooth, ventricose, locally intervenose, especially at their base, whitish to cream in dried material, with the edge entire and concolorous, lamellulae present. Stipe 8–35 × 1.5–3.5 mm, central, cylindrical, equal



Figs. 1–4. *Gymnopus castaneus* (holotype). 1. Pileipellis; 2. spores; 3. basidia; 4. caulocystidia. Bar = 15 μ m.

or tapering upwards (–3.5 mm), rarely compressed and tapering downwards, uniformly pale red (10 R 5/6) except for the pinkish base, apparently smooth, completely pruinose under the lens, more pronounced at the apex.

Spores 7.2–8.6–10.3(–11) \times 5.5–6.4–7.6(–7.7) μ m; Q = 1.22–1.34–1.49; (n = 23), broadly ellipsoid, rarely subglobose, sometimes with a very faint suprahilar depression, smooth, thin-walled, hyaline or with vacuolar inclusions, inamyloid, acyanophilic. Basidia (30–)35–45 \times (8.5–)9.5–12 μ m, clavate, 4-spored, some 2-spored (rarely 1- or 3-spored), sterigmata up to 7 μ m long, hyaline or with coarse vacuolar inclusions, clamped. Cheilocystidia absent, only some cylindrical to clavate elements intermingled with the basidia. Hymenophoral trama regular to subregular, not embedded in gelatinous matter, not dextrinoid, consisting of cylindrical hyphae 40 \times 2.5–6 μ m, fairly thick-walled (0.5–1 μ m). Pileipellis a 'pseudohymeniderm' consisting of hyphae 2–5(–8) μ m wide, fairly thick-walled (–0.5 μ m), with numerous cylindrical, cylindrico-flexuose to subclavate dermatocystidioid elements up to 55 \times 6–9 μ m, rostrate or suberect, not forming a well-developed palisade, and locally forming dense clusters. All hyphae and dermatocystidioid elements with yellowish intraparietal and encrusting pigment. Caulocystidia up to 90 \times 5–10 μ m, descending to the base, similar to the dermatocystidioid elements. Hyphae of the stiptipellis 3–6 μ m wide, not dextrinoid, smooth or with very few isolated projections. Oleiferous hyphae present, but very scarce. No sarcodimitic tissues present. Clamps present at all septa.

Habitat — On soil, mossy banks, among fallen, decaying leaves of *Quercus ilex* ssp. *ballota* (Desf.) Samp. and *Pinus pinea* L.

Material studied. SPAIN: Toledo, La Iglesuela, 13 Dec. 1995, S.G. Busutil, C. Sánchez & M. Villarreal, AH 20475; Toledo, Pinar de Almorox, 19 Nov. 1996, F. Esteve-Raventós, C. Sánchez & M. Villarreal, AH 21520 (holotype); Toledo, Pinar de Almorox, 11 Dec. 1999, R. Izquierdo & M. Villarreal, AH 25421; Toledo, Pinar de Almorox, 30 May 2000, R. Izquierdo & M. Villarreal, AH 27053.

Gymnopus castaneus belongs to sect. *Vestipedes* (Fr.) Antonín, Halling & Noordel., and is characterized by its reddish basidiomata, inamyloid spores, the absence of hymenial cystidia, presence of encrusting and intraparietal pigment and absence of sarcodimitic tissues. Because of the presence of repent to suberect dermatocystidia this new taxon was at first thought to belong to *Hydropus*. However, *G. castaneus* does not have vacuolar pigment, the trama of the stipe is not sarcodimitic, vascular hyphae are very rare, there are no cheiloid and no pleurocystidia, and the spores are non-amyloid. In quite a few species placed in *Hydropus* by Singer (1982) one or more of these characters are lacking, but in none of them are they all lacking. Singer (1982: 9) mentioned 12 species with encrusting pigment, but in 11 of these it occurs in addition to evident vacuolar pigment. Only in *H. brunnescens* did he describe the sole presence of encrusting pigment. We believe that the latter is not a true *Hydropus* but should be included in *Gymnopus* as well. We therefore propose the following new combination:

***Gymnopus brunnescens* (Murrill) Villarreal, Heykoop & Esteve-Rav., comb. nov.**

Basionym: *Omphalina brunnescens* Murrill, Proc. Florida Acad. Sci. 7 ('1944' 1945) 112.

Hydropus brunnescens (Murrill) Singer, Flora Neotropica 32 (1982) 48. Not to be confused with *Collybia brunnescens* Peck, Bull. Torrey Bot. Club. 33 (1906) 214.

Singer (1982: 48) indicated that *G. brunnescens* has amyloid spores and cheilocystidia (which according to him were also observed in other Floridan material). However, we could not observe these characters in the type.

Whilst studying the taxonomic status of this taxon we also considered its inclusion in *Dennisiomyces* Singer on account of the absence of vacuolar pigment and the presence of abundant parietal and encrusting pigment. According to Singer (1982, 1986: 391) *Dennisiomyces* differs mainly from *Hydropus* in the absence of intracellular pigments and the predominantly collyboid to tricholomatoid habit. However, the diagnosis of *Dennisiomyces* (Singer, 1955) only includes cystidiate species with amyloid spores.

The very simple anatomy of this taxon points to its inclusion in the *Collybia-Marasmiellus* complex, where, according to the monograph of that group by Antonín & Noordeloos (1997), it keys out in *Gymnopus* sect. *Vestipedes*. However, the presence of a strong farinaceous smell and the absence of cheilocystidia separate *Gymnopus castaneus* clearly from both subsections *Impudicae* and *Vestipedes*. The most closely related species within section *Vestipedes* is *G. terginus* (Fr.) Antonín & Noordel. The latter is, however, different from *G. castaneus* because of its yellow to reddish brown pileus, its collyboid (never omphalioid) habit, the presence of cheilocystidia, and much narrower spores (up to 3–4.5 µm).

Gymnopus brunnescens, described from Florida is very similar to *G. castaneus* sharing the following characters: 1) similar structure of the pileipellis and stiptipellis; 2) absence of hymenial cystidia; 3) similar pigmentation. The differences between *G. castaneus*, *G. brunnescens* and *G. terginus* are tabulated in Table I.

Table I. A comparison between *G. castaneus*, *G. brunnescens* and *G. terginus*.

	habit	pileus colour	lamellae colour	sporal shape	cheilocystidia	Q (L/l)
<i>Gymnopus castaneus</i>	collybioid to subomphalioid	red-brown	whitish when fresh and dry	broadly ellipsoid to subglobose	absent	1.22– 1.34 –1.49
<i>Gymnopus brunnescens</i>	omphalioid	isabelline	fresh: white or citrinous? dry: orange-brown	narrowly ellipsoid	absent	1.50– 1.80 –2.08(–2.09)
<i>Gymnopus terginus</i>	collybioid	yellow to reddish brown	whitish to reddish brown	oblong to cylindrical	present	2.00

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**DERMOLOMA MAGICUM SPEC. NOV.,
A GRASSLAND FUNGUS MIMICKING PORPOLOMA METAPODIUM**

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Dermoloma magicum is proposed as a new species, characterised by reddening, then blackening basidiocarps, a unique feature within the genus *Dermoloma*. The species has been collected in grasslands in the Netherlands and Scotland. The collections were initially identified as *Porpoloma metapodium*. The differences with that fungus and with related *Dermoloma* species are discussed.

During mycological research in 1994 in limestone grasslands in southern Limburg, the Netherlands, I came across a tricholomatoid agaric with medium-sized, grey-brown basidiocarps which attracted attention because they stained reddish, then black, when damaged. It was identified on the basis of macroscopic characters as *Porpoloma metapodium* (Fr.) Singer, although the basidiocarps were relatively small for that species. The amyloid spores seemed to confirm this identification. The record of *P. metapodium*, listed in the checklist of the Netherlands by Arnolds et al. (1995) was based on this collection. The following year the same fungus was collected in another locality in the same region.

In the framework of a revision of the genus *Porpoloma* for Flora agaricina neerlandica (Arnolds & Noordeloos, 1999) the collections were studied in more detail. It then appeared that the structure of the pileipellis was a hymeniderm of spheropedunculate cells, substantially different from the cylindrical hyphae in the pileipellis of *P. metapodium*. This combination of characteristics places our fungus in the genus *Dermoloma*, where no discolouring species have been described so far. In view of its striking colour change I describe it as *D. magicum*.

Interestingly, I found among the other collections identified as *Porpoloma metapodium* in the Nationaal Herbarium Nederland (L), further material with a hymeniform pileipellis, collected by M.E. Noordeloos in Scotland. It agrees in all relevant characters with *Dermoloma magicum*. It would not be surprising if some other records of *P. metapodium* have also been misidentified in the past.

***Dermoloma magicum* Arnolds, spec. nov. — Fig. 1**

Pileus 25–50 mm latus, conico-convexus vel plano-convexus, interdum umbonatus, brunneo-griseus vel obscure griseo-brunneus, siccus, glaber vel subsquamulosus, interdum rugulosus. Lamellae adnatae vel emarginatae, subdistantes, subcrassae, intervenosae, ventricosae, pallide griseae vel griseo-brunneae. Stipes 30–65 × 4–12 mm, aequalis, fistulosus, pallide griseus, fibrilloso-striatus, flocculosus. Caro compacta, albida, fracta rubescens, dein nigrescens, odore farinaceo vel herbaceo. Basidiomata vulnerata rubescentia, dein nigrescentia.

Basidia 26–38 × 6.5–7.5 µm, clavata, 4-sporigera vel 4-, 2- et 1-sporigera intermixta. Spores 6–9 × (3.5–)4–5(–5.5) µm, ellipsoideae vel ellipsoideae-oblongae, amyloideae. Cystidia nulla. Trama lamellarum subregularis. Pileipellis hymenidermium unistratum, cellulae ovatae, clavatae vel spheropeduncu-

latae, 17–52 × 7–22(–35) µm, saepe tunicis brunneis. Stipitipellis cutis, hyphae 3–8 µm latae. Caulocystidia gregaria, clavata vel subcylindracea, 23–47 × 4–14 µm. Fibulae frequentes. Ad terram in pratis.

Holotypus: The Netherlands, Limburg, Epen, Cotessen, 21.X.1995, *E. Arnolds 6701* (WAG).

Pileus 25–50 mm wide, conico-convex, convex or plano-convex, with or without obtuse umbo, finally sometimes with the centre depressed, not hygrophanous, brownish grey to rather dark grey-brown (K. & W. 6C3, 6C4, 6E5, 6F6); margin even or lacerate, not striate; surface dull, smooth to slightly squamulose near centre, even or radially wrinkled to rugulose, sometimes cracked, exposing white context beneath. Lamellae (L = 28–40, l = 1–5) adnate to emarginate, moderately crowded to subdistant, rather thick, interveined, ventricose, up to 8 mm broad, pale grey to grey-brown (K. & W. 5C4, 6D3). Stipe 30–65 × 4–12 mm, cylindrical or slightly tapering downwards, narrowly fistulose, pale grey to beige, base sometimes ochre-yellow, fibrillose striate lengthwise, in addition minutely floccose, in some specimens (*Arnolds 6701*) with grey flocks. Context firm, compact, whitish, when cut soon discolouring orange-red, then black in places, in particular in base of stipe and above lamellae. Smell farinaceous to herbaceous. All parts of basidiocarp quickly turning orange to red when bruised, after a while (15–30 minutes) blackening. Colour of spore-print unknown.

Spores 6.0–9.0 × (3.5–)4.0–5.0(–5.5) µm, on average 6.7–7.8 × 4.4–4.5 µm, Q = 1.35–2.0(–2.3), av. Q = 1.50–1.75, ellipsoid to ellipsoid-oblong with prominent hilar appendix, often with one or two droplets, amyloid. Basidia 26–38 × 6.5–7.5 µm, clavate, 4-spored or mixed 4-, 2- and 1-spored, in exsiccata with brown content. Lamella-edge fertile. Hymenophoral trama subregular, made up of cylindrical to inflated elements, 35–85 × 5–16 µm. Pileipellis a unistratous hymeniderm, made up of erect, branched hyphae with swollen, ovate, clavate to spheropedunculate cells, 17–52 × 7–22(–35) µm, often slightly thick-walled, with parietal to encrusted brown pigment; sometimes with scattered, subcylindrical pileocystidia, 52–58 × 6–6.5 µm. Stipitipellis a cutis of repent hyphae, 3–8 µm wide, with pale yellowish parietal pigment, in addition sometimes minutely encrusted, producing dense clusters of caulocystidia, in particular near apex of stipe. Caulocystidia subcylindrical to clavate, 23–47 × 4–14 µm, thin-walled, hyaline.

Habitat — Terrestrial, saprotrophic, solitary or in small groups in old, not or weakly fertilised pastures on dry, loamy, often calcareous soil. Aug.–Oct.

Collections examined. THE NETHERLANDS: Limburg, Epen, Cotessen, 21 Oct. 1995, *E. Arnolds 6701* (WAG, holotype); Limburg, Wittem, Nijswiller, 12 Oct. 1994, *E. Arnolds 6549* (WAG).— GREAT BRITAIN: Scotland, Moffat, 24 Aug. 1996, *M.E. Noordeloos 9665* (L).

Dermoloma magicum is rather variable in some characters. In the collection from Scotland the spores are ellipsoid (av. Q = 1.5) and small (6–7.5 × 4–4.5(–5.0) µm), in the collection from Cotessen predominantly ellipsoid-oblong (av. Q = 1.75) and larger: 7.0–9.0 × (3.5–)4.0–5.0 µm. The collection from Nijswiller is intermediate in this respect, and differs from the other collections in the presence of 2- and 1-spored basidia, a feature often observed in *Dermoloma*-species and of no taxonomic significance (Arnolds, 1993). In addition the Nijswiller collection bears some pileocystidia in between the hymenidermal cells (Fig. 1E), a phenomenon not reported before in this genus. They could not be found in the other collections. It is unclear whether this character is taxonomically important or only an accidental anomaly.

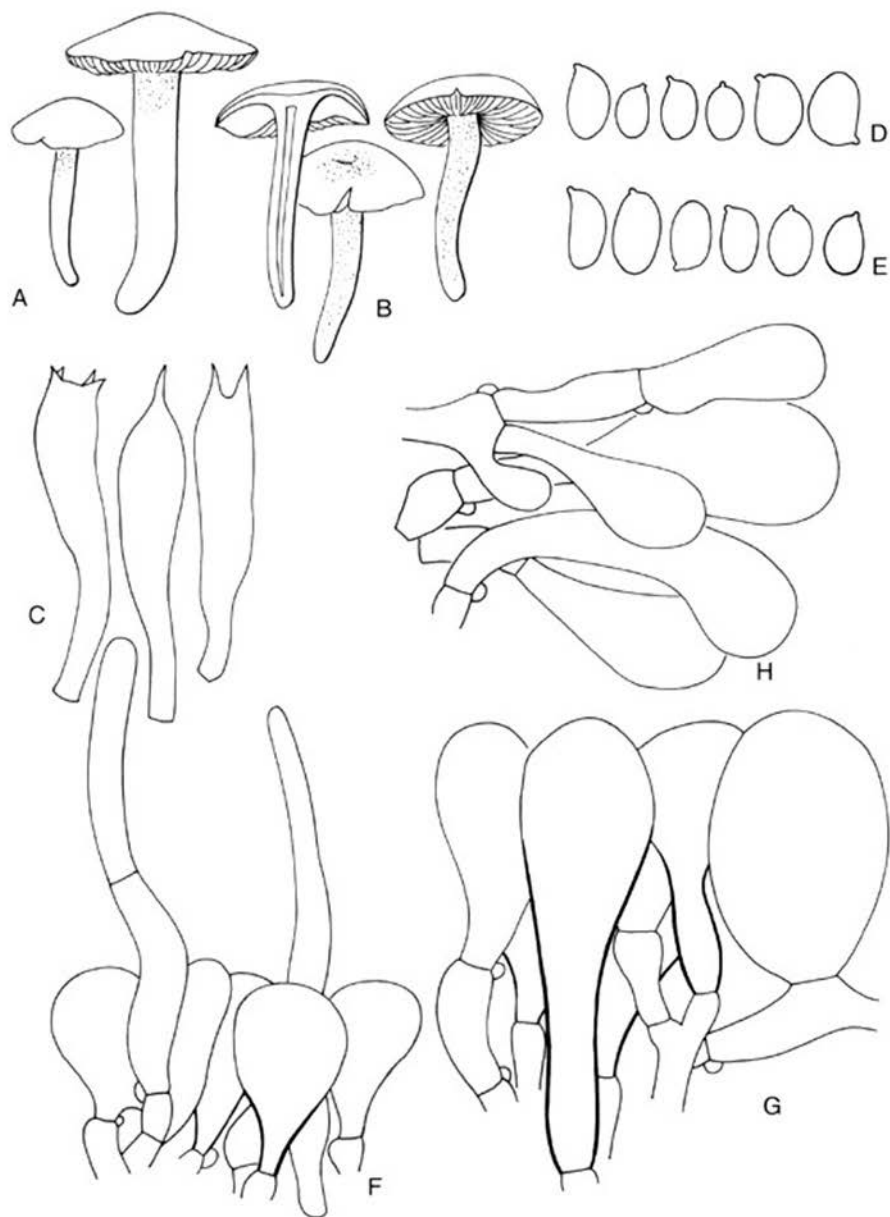


Fig. 1. *Dermoloma magicum*. A, B. Basidiocarps ($\times 0.7$); C. basidia; D, E. spores; F, G. pileipellis; H. caulocystidia (Figs. B–H, $\times 1400$; Figs. B, E, G, H from Arnolds 6701, type; A, C, D, F from Arnolds 6549).

Within the genus *Dermoloma*, *D. magicum* is unique in its reddening, then blackening basidiocarps. In microscopic characteristics this species shows most resemblance to *D. pseudocuneifolium*, for instance in the relatively large spore-size (Arnolds, 1993). As well as lacking the remarkable discoloration, the latter species also differs in its smaller basidiocarps (pileus 7–25 mm, stipe 12–50 × 1–3 mm, lamellae 11–21) with darker colours.

Dermoloma magicum differs from *Porpoloma metapodium* not only in the hymeniform pileipellis, but also in general habit. The latter species has more robust basidiocarps with the pileus 50–100 mm in diameter, the stipe 10–20(–30) mm thick and more numerous lamellae (40–50). In addition the stipe in *Porpoloma metapodium* is concolorous with the pileus, whereas in *D. magicum* it is considerably paler than the pileus. The spores in *P. metapodium* are slightly narrower, measuring 6–8 × 3–4 µm (Arnolds & Noordeloos, 1999).

ACKNOWLEDGEMENT

For critical comments and linguistic improvements I thank Dr. Th.W. Kuyper.

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**BOOKS RECEIVED BY THE NATIONAAL HERBARIUM NEDERLAND
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R. Agerer et al. (eds.). *Descriptions of ectomycorrhiza 5*. (Einhorn Verlag, P.O. Box 1280, D-73502 Schwäbisch Gmünd. 2001). ISSN 1431-4819. Pp. 225, numerous line-drawings. Price: DM 280.-.

In the fifth issue of this series, 33 species of ectomycorrhiza are described, including representatives of the genera *Alpova*, *Arcangeliella*, *Boletus*, *Cortinari*, *Descomyces*, *Lactarius*, *Russula*, *Tomentella* and *Xerocomus*, and in addition some unidentified mycorrhiza. For each species a description is given of the morphological and anatomical characters, colour reactions, and chemical properties, as well as extensive discussions and a list of references. The anatomical characters are illustrated in a very elaborate and accurate way with very clear line-drawings. The series is an invaluable tool for all interested in ectomycorrhiza.

V. Antonín & P. Škubla. *Interesting macromycetes found in the Czech and Slovak Republics*. (Fungi non delineati, pars XI. Libreria Mykoflora, Via Ottone Primo 90, I-17021 Alassio. 2000). Pp. 46, 16 coloured photographs, 15 line-drawings. Price: unknown.

Descriptions of 12 species of agarics and boleti from the Czech and Slovak republics are described and illustrated with line-drawings of the microscopical characters and coloured photographs. Three new taxa are described: *Pluteus favrei*, *Gymnopilus josserandii*, and *Hygrophoropsis aurantiaca* var. *robusta*. In addition one new combination, viz. *Hemimycena delectabilis* var. *bispora* is proposed. All species are extensively described with their macro- and micromorphology and discussed, and references to useful literature are given.

G. Baiano, D. Garofoli & M. Fillippa. *Ascomycetes interessanti del Nord Italia*. (Fungi non delineati, pars XII. Libreria Mykoflora, Via Ottone Primo 90, I-17021 Alassio. 2000.) Pp. 74, 16 coloured photographs, numerous line-drawings. In Italian. Price: unknown.

Volume twelve of *Fungi non delineati* deals with rare ascomycetes from Italy, which are fully described and illustrated with photographs and line-drawings of the microscopical features. Eleven species of *Peziza* (*P. merdae*, *P. acroornata*, *P. coquandi*, *P. saliciphila*, *P. luteoloflavida*, *P. maximovicii*, *P. azureoides*, *P. phlebospora*, *P. pseudovesiculosa*, *P. brunneoatra* and *P. irina*), two species of *Helvella* (*H. aestivalis*, *H. branzeiana* and *H. confusa*) and *Spooneromyces helveticus* and *Microstoma protracta* are included in the work. *Helvella aestivalis* is neotypified with a collection from France.

A. Bidaud, P. Moëgne-Loccoz & P. Reumaux. *Atlas des Cortinaires. Pars X*. (Éditions Fédération Mycologique Dauphiné-Savoie, 70 Rue Edison, F-69330 Meyzieu. 2000.) Pp. 411–526, 62 sheets with descriptions and line-drawings, coloured plates 262–310. In French. Price: FF 580; € 89.-.

Volume ten of this prestigious French series entirely devoted to the large genus *Cortinarius* contains two parts. Pars X(1) is devoted to the subgenus *Myxacium* with 96 taxa described and illustrated, including an impressive number of new species, varieties and forms. The second part (Pars X(2)) is devoted to subgenus *Hydrocybe*, sect. *Castanei* with 24 taxa. Besides the loose-leafed plates and descriptions, each part contains a booklet with taxonomic and nomenclatural comments, and as the authors call 'attempts to dichotomous keys'. Again this publication illustrates the rather big variety encountered in the notoriously difficult genus *Cortinarius*, and an attempt to solve the problems involved with this variability.

A. Bidaud, P. Moëgne-Loccoz & P. Reumaux. *Atlas des Cortinaires. Pars XI*. (Éditions Fédération Mycologique Dauphiné-Savoie, 70 Rue Edison, F-69330 Meyzieu. 2001.) Pp. 527–626, 60 sheets with descriptions and line-drawings, coloured plates 311–357. In French. Price: FF 580; € 89,-.

The eleventh part treats subgenus *Hydrocybe*, sect. *Hydrocybe* (Pars XI(1)) and of subgenus *Phlegmacium* sect. *Calochroi* (Pars X(2)). This part follows the concept of the previous parts of this series, and is equally well got-up. Again many new taxa are proposed.

F.S. Dobson. *Lichens: An illustrated Guide to the British and Irish Species*. 4th ed. (Richmond Publishing Co., P.O. Box 963, Slough, SL2 3RS, England. 2001.) ISBN 0-85546-094-6 (Paperback); 0-85546-093-8 (Hardcover). Pp. 431, with numerous coloured photographs, line-drawings and distribution maps. In English. Price: £ 45 (Hardback), £ 30 (Paperback).

This is the fourth, fully revised edition of the well-known Lichen guide by Dobson. The text has been updated with respect to nomenclature and data on occurrence, distribution and ecology. The book covers the common species in Great-Britain and Ireland and a selection of rarer species. Clear keys and tables to the genera and species are given, as well as elaborate line drawings of microscopic and other diagnostic structures. The c. 450 species are treated alphabetically according to genus.

New in this edition are the full-colour photographs that have been included, which greatly improves the attraction of this book, and facilitates identification. This book certainly will find its way to lichenologists and naturalists in general.

R.T.V. Fox (ed.). *Armillaria Root Rot: Biology and control of Honey Fungus*. (Intercept Ltd., P.O. Box 716, Andover, Hampshire SP10 1YG, United Kingdom. 2000.) Pp. 222, 10 coloured photographs, numerous illustrations. Price: £ 47.50.

This book is entirely devoted to *Armillaria*, the Honey Fungus, and deals with various aspects. The book is written by a collective of 10 authors from the United Kingdom, the Netherlands, and Pakistan. It is divided into five sections. Section one deals with the biology of *Armillaria* with three contributions on biology and life-cycle (R.T. Fox), ecology and epidemiology (A. Termorshuizen), and quantitative aspects of the epidemiology (A. Lamour & M. Jeger). Section two deals with diversity, with chapters on taxonomy and nomenclature (D.N. Pegler), and molecular methods to detect and identify *Armillaria* (A. Pérez-Sierra, D. Whitehead and M. Whitehead). Section three deals with pathogenicity (R.T. Fox). Part four handles with control, with contributions on the extent of losses and aims for managing

Armillaria (R.T. Fox), culturing methods (R.T. Fox), chemical control (J.S. West), and biological and integrated control (F. Raziq). The final section tries to answer all the questions about *Armillaria* (R.T. Fox). And index facilitates the use of the book. This well-edited and printed book on this important pathogenic fungus will give an up-to-date overview of the state of affairs in *Armillaria*, valuable to all pathologists and ecologists confronted with this fungus, as well as teachers and other people intrigued by this organism.

J.C. Frisvad, P.D. Bridge & D.K. Arora (eds.). *Chemical fungal taxonomy*. (Marcel Dekker AG, Hutgasse 4, Postfach 812, CH-4001 Basel. 1998.) ISBN 0-8247-0069-4. Pp. 398, several text-figs. Price: US \$ 195.-.

Recent developments in molecular biology have enabled the use of new techniques, with many implications for fungal taxonomy. However, chemical and molecular approaches have often been applied independently. In this book these approaches have been brought together for the first time. The main instruction for the authors was, according to the editors, to give information on used molecular techniques and their application in systematic mycology. This has resulted in 14 chapters, written by 24 authors. Various subjects are covered in this book: PCR, RFLP, proteins, isozymes, polysaccharides, unsaponifiable lipids, fatty acids, carbohydrates, volatiles, secondary metabolites, all in their relation to fungal taxonomy. Each contribution provides an extensive list of useful references. A cumulative index concludes the book.

A. Gennari. *401 Funghi* (Associazione Micologica Bresadola, Via A. Volta 46, I-38100 Trento. 2001.) Pp. 544, 439 coloured plates, several line-drawings. In Italian. Price: Lit. 57.000 ; € 29.44.

This book presents coloured photographs of excellent quality and descriptions of about 400 fungi to be found in the Italian forests. Emphasis is laid on gilled mushrooms and boletes, especially the edible and poisonous ones. Introductory chapters are quite extensive and deal with general anatomical and ecological characters of fungi. In an appendix the reader is informed how to deal with dangerous vipers which can be encountered during a mushroom collecting trip. The book is recommended for the good quality of its illustrations.

L.J.L.D. van Griensven (ed.). *Science and cultivation of edible Fungi. Proceedings of the 15th International Congress, Maastricht, Netherlands, 15-19 May 2000*. (A. A. Balkema, P.O. Box 1675, NL-3000 BR Rotterdam. 2000.) ISBN 90-5809-143-0. Pp. 1020, many text-figs., 2 vols. Price: US \$ 160; € 160.

These two volumes of congress proceedings cover a wide range of subjects by numerous authors. The contributions are arranged according to various themes. Three keynote lectures on use of agricultural waste materials in the cultivation of fungi, genetics and breeding of *Agaricus bisporus* and medicinally important fungi, are followed by various contributions in the following fields of research:

Physiology of edible fungi
Development and morphogenesis
Genetics and breeding
Substrates

Crop management
 Pests and diseases
 Quality Control
 Medically and industrially important edible fungi
 Environmental aspects of mushroom cultivation
 Economics of the industry and market research
 Mushroom cultivation: teaching and extension

The last part of the book contains the proceedings of technical sessions on
 Cultivation technique of *Agaricus*
 Equipment-Agricultural waste as substrate for the cultivation of edible fungi
 Spawn, substrate, casing and cultivation
 Cultivation technique

The two volumes contain the most up-to-date record on the subject, and should be required reading for all interested in the subject.

G.J. Krieglsteiner (ed.). *Die Großpilze Baden-Württembergs. Band 1: Allgemeiner Teil, Ständerpilze: Gallert-, Rinden-, Stachel- und Porenpilze.* (Eugen Ulmer Verlag, Wollgrasweg 41, D-70599 Stuttgart. 2000.) ISBN 3-8001-3528-0. Pp. 629, numerous coloured photographs, line-drawings and distributions maps. In German. Price: DM 98,-; € 50.11.

Ditto. *Band II: Ständerpilze: Leisten-, Keulen-, Korallen-, und Stoppelpilze, Bauchpilze, Röhrlings- und Täublingsartige.* (2000.) ISBN 3-8001-3531-0. Pp. 620, numerous coloured photographs, line-drawings and distributions maps. In German. Price: DM 98,-; € 50.11.

Two volumes have appeared of this work, which eventually will count four volumes covering all macromycetes found in Baden-Württemberg State, Germany. The first two volumes are devoted to Basidiomycetes. In volume 1 all Heterobasidiomycetidae are treated and of the Homobasidiomycetidae: Aphyllophorales in part (Corticaceae, Coniophoraceae, Lachnocladiaceae, Thelephoraceae, Schizophyllaceae, Ganodermataceae, Hymenochaetaceae and Polyporaceae). Volume 2 includes all other Aphyllophorales, Gasteromycetes, Boletales and Russulales. For each family keys to the genera are given. With each genus, a genus diagnosis is given, followed by a key to the species, and descriptions of the species, often accompanied by a coloured photograph and/or line-drawings of diagnostic characters, and/or a distribution map in Baden-Württemberg. The photographs are often of good quality. Species concepts, however, are sometimes rather wide. The authors brought together a wealth of knowledge on these groups of fungi occurring in their home state, which clearly has a wider interest than for inhabitants of this large German state. We look forward to the forthcoming volumes of this series.

M. Mata. *Macrohongos de Costa Rica Mushrooms.* (Instituto Nacional de Biodiversidad, Costa Rica. 1999.) ISBN 9968-702-30-7. Pp. 253, numerous coloured photographs. In Spanish and English. Price: unknown.

This bilingual fieldguide offers a very nice introduction into the world of macrofungi in Costa Rica, an area which is renowned for its rich biodiversity. After about 50 pages introductory chapters, the author presents descriptions and notes on distribution and ecology of a

selection of species, all provided with a coloured photograph, which generally is of good quality. It is an interesting mixture of European and North American taxa. Recommended for everyone interested in the mushrooms of this unique part of the world.

J. A. Muñoz. *El género Leccinum S.F. Gray en el Norte de España* (Fungi non delineati, pars XIII. Libreria Mykoflora, Via Ottone Primo 90, I-17021 Alassio. 2000.) Pp. 47, 19 coloured photographs, 6 coloured plates, numerous line-drawings. In Spanish. Price: Lit. 16.000; € 8,26.

Volume thirteen of this series is entirely devoted to the genus *Leccinum* (Boletales) in Spain. A dichotomic key to the species is given, followed by extensive descriptions and notes on chorology and taxonomic position of the taxa. Reference is also given to authoritative icons in other publications. Diagnostic microscopical characters, such as spores, cystidia and pileipellis structures are illustrated in the form of line drawings. Coloured plates of good quality with photographs and water-colour paintings illustrate this treatment of *Leccinum*.

C. Papetti et al. (eds.). *Micologia 2000*. (Associazione Micologica Bresadola, Via. A. Volta 46. I-38100 Trento. 2000). Pp. 712, numerous illustrations and coloured pls. In English, Italian, French and Spanish. Price: Lit. 125.000; € 64,56.

On the occasion of the new millennium, the Foundation Centro Studi Micologici of the Associazione Micologica Bresadola took the initiative to invite mycologists from all over the world to contribute a paper to a special publication devoted to recent developments in (taxonomic) mycology. As a result 712 pages are published, containing 56 papers by 90 mycologists, in various languages. Most contributions are on the taxonomy of macromycetes and fimicolous pyrenomycetes, but also articles dealing with ecology and computer-aided tools are included. The book is illustrated with numerous coloured photographs and line-drawings. The AMB can be congratulated with this prestigious publication, which enables the reader to get a good impression of present-day taxonomic mycology.

M. Raillere & M. Gannaz. *Les Ramaria Europeennes* (Fédération Mycologique Dauphiné-Savoie, 22 le Praz du Nant, F-73000 Bassens, France, 1999). Pp. 176, without illustrations. Price: FF 120.00; € 18,29.

This book gives an overview of the European species of genus *Ramaria*, with a short introduction on the genus, followed by a dichotomic key to the European species. The main part of the book consists of so-called fishes, of one page each, for all species of *Ramaria* known from Europe. The fishes contain information on macroscopical and microscopical characters, chemical reactions, comments on related species, and a selection of descriptions and illustrations in literature. A brief glossary explains the terms used in the text, and the book ends with a bibliography and an index to all names used in the text.

M. Ulloa & R.T. Hanlin. *Illustrated Dictionary of Mycology*. (APS Press, Europe Branch Office, Broekstraat 47, B-3001 Heverlee. 2000.) Pp. 448, 1322 black-and-white illustrations. Price: US \$ 99.

This dictionary contains about 4000 mycological terms, for which definitions are given and also the etymology of the term. Many terms are illustrated with either black-and-

white photographs or line-drawings. The result is an impressive hard-bound volume in A4-size which is well printed and easy to use. It will be welcomed by all students in mycology.

G.J.M. Verkley. *A monograph of the genus Pezicula and its anamorphs*. (Studies in Mycology 44. Centraalbureau voor Schimmelcultures, Utrecht. 1999.) ISBN 90-70351-40-4. Pp. 180, 50 black-and-white plates, numerous line-drawings. Price: unknown.

This long awaited worldmonograph of the difficult genus *Pezicula* appeared in the end of 1999. After the works of Groves (1938–1941) no thorough study has been published for a long time, although various authors studied the genus in varying depth. This new monograph is the result of four years of study. In this study also collections were made in the areas where Groves worked in the past, teleomorphs and anamorphs on natural substrate as well as in culture were studied, type-specimens were investigated, and some molecular studies were carried out. Also several species of the closely related genera *Ocellaria* and *Dermea* have been studied.

The result is a thorough work, with 28 pages of introduction about the history of the genus, material and methods, discussion on the relation of *Pezicula* and other genera in the Dermateaceae, and a discussion on the molecular characterization of the cultured species with RFLP. The molecular data seem to support the taxa distinguished morphologically. Keys are given to some related genera, to the species of *Pezicula* and *Neofabraea* based on apothecial characters, and to the anamorphs of *Pezicula* and *Neofabraea* based on characters *in vivo* and *in culture*. As a result of the studies *Ocellaria* is regarded as a synonym of *Pezicula*, and the genus *Neofabraea* is accepted with four species. Three new taxa are described in *Pezicula*. Seventy-six taxa mentioned in relation to *Pezicula* are discussed, leading to six new combinations in other genera.

Of the 26 accepted species in *Pezicula* an extensive description *in vivo* and *in vitro* is given, notes on substrate preference and distribution, and a list of specimens examined, together with a discussion.

Of course, this book is not the final one on *Pezicula* since not all taxa could be evaluated through lack of material. Also, one does not have to agree with all synonyms. But this book is certainly recommended for those interested in the group.



Plate 1. *Crepidotus croceitinctus* var. *aurantiacus* (no. 6483). Fruit-bodies. Photograph by I. Krisai-Greilhuber.



Plate 2. *Amanita aurantiobrunnea*.
Coll. T. Henkel 6898.



Plate 3. *Amanita cyanopus*. Coll. T. Henkel et al.
7083 (holotype).



Plate 4. *Amanita perphaea*. Coll. T. Henkel 6229 (holotype).



Plate 5. *Amanita calochroa*. Coll. T. Henkel 6426 (holotype).



Plate 6. *Amanita lanivolvata*.
Coll. T. Henkel 6432.



Plate 7. *Amanita xerocybe*. Coll. T. Henkel 6228.

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