

Cryptolepiota, a new sequestrate genus in the Agaricaceae with evidence for adaptive radiation in western North America

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Abstract: Phylogenetic analyses based on nLSU and ITS sequence data indicate that the sequestrate genus *Gigasperma* is polyphyletic. *Gigasperma cryptica*, which is known only from New Zealand, has affinities with the Cortinariaceae whereas *G. americanum* and two additional undescribed taxa from western North America are derived from *Lepiota* within the Agaricaceae. The three North American taxa appear to be recently evolved and are closely related. They occur in similar environments and form a well supported clade indicating that adaptive radiation has occurred within this group of fungi. An independent genus with sequestrate fructifications, *Cryptolepiota* is proposed to accommodate the three species in this clade. *Cryptolepiota microspora* and *C. mengei* are described as new, and *G. americanum* is transferred to *Cryptolepiota*. *Gigasperma cryptica* is illustrated and compared with the species of *Cryptolepiota*.

Key words: Basidiomycota, Cortinariaceae, fungi, *Gigasperma*, phylogenetics

INTRODUCTION

The genus *Gigasperma* was erected by Horak (1971) to accommodate two sequestrate fungi from Tasmania and New Zealand. *Gigasperma* is characterized by its hypogeous habit and distinctive smooth, subglobose to globose, thick-walled basidiospores. As proposed by Horak, the genus comprised two taxa, *G. cryptica* Horak and *G. clelandi* (Rodway) Horak,

although *G. clelandi* subsequently was transferred to the monotypic genus *Horakiella* by Castellano and Trappe (1992). A few years later *Gigasperma americanum* Kropp and Hutchison was added to the genus so that it currently contains two species, *G. cryptica* and *G. americanum* (Kropp and Hutchison 1996).

Gigasperma cryptica is known only from New Zealand where it occurs with species of *Nothofagus* (Castellano and Trappe 1992). On the other hand, *Gigasperma americanum* thus far has been found only in the western United States. It is rarely collected and occurs in stands of *Cercocarpus ledifolius* Nutt., a shrub or small tree in the Rosaceae that grows in semi-arid habitats throughout much of western North America. Because *C. ledifolius* is one of the Rosaceae that form ectomycorrhizas Kropp and Hutchison (1996) suggested that *G. americanum* might be ectomycorrhizal, although this has not been demonstrated experimentally.

Despite the fact that both *Gigasperma* species are hypogeous and have a similar spore morphology, the enormous geographical distance separating their populations raises doubts about their relatedness. Moreover collections of additional, potentially undescribed material that shared morphological affinity with *Gigasperma* indicate that evolutionary radiation might have occurred within this group in western North America. The objectives of this work were: (i) to assess the monophyly of *Gigasperma* sensu lato and (ii) to describe the two new taxa from North America and determine their phylogenetic positions.

MATERIALS AND METHODS

Microscopic features were measured from material mounted in Melzer's reagent with oil immersion at 1000 \times , and photomicrographs were taken with interference contrast. Spore measurements are reported as averages (20 spores) and ranges, whereas measurements for the spore wall and the other cells are given as ranges. Spore dimensions are given as length then width.

DNA was extracted with the method of Kropp et al. (1996). Briefly, dried herbarium material was crushed in liquid nitrogen before being mixed with an extraction buffer (0.7 M NaCl, 0.05 M Tris-HCL [pH 8], 0.01 M EDTA, 1% β -mercaptoethanol, 1% CTAB) and incubated at 60°C. The mixture was emulsified in chloroform:isoamyl alcohol (24:1 by volume) and centrifuged. The DNA was precipitated from the supernatant with cold isopropyl alcohol. Standard PCR protocols were used to amplify the portion of the nuclear large ribosomal subunit (nLSU) between primers LROR and LR5 (Moncalvo et al. 2000, Vilgalys and

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Hester 1990), and the internal transcribed spacer (ITS) also was amplified between primers ITS4 and ITS5 (White et al. 1990). Direct sequencing of the PCR products was done with primers LROR and LR5 for the nLSU and ITS4 and ITS5 for ITS. The sequences used for the phylogenetic analysis were obtained with dye terminator methods with an ABI 3730 DNA Analyzer and deposited in GenBank (TABLE I).

Initial BLAST queries with sequences from the *Gigasperma* species indicated that they belong to the agaricoid clade of Matheny et al. (2006). Thus, taxon sampling to assess the monophyly of *Gigasperma* was focused on this clade with members of the tricholomatoid clade of Matheny et al. (2006) being used to root the tree. To assess the phylogenetic positions of the North American taxa relative to *Lepiota*, taxon sampling was based on Vellinga (2004). The nLSU and ITS sequences for each sampled taxon were concatenated and aligned with Clustal X (Thompson et al. 1997). All taxa used in the analysis were represented by both ITS and nLSU sequences (TABLE I). Alignments are available in TreeBASE, accession number 11566. A total of 1819 characters was used to assess the monophyly of *Gigasperma* (FIG. 1), and 1702 characters were used to assess the position of the North American taxa relative to the genus *Lepiota* (FIG. 2).

The aligned dataset was analyzed with MrBayes 3.1 (Ronquist and Huelsenbeck 2003). Tree searches were performed with a time reversible model of evolution (Maddison 1994, Rodriguez et al. 1990) and a discrete gamma distribution with six substitution types and some invariant sites (GTR+G+I) was assumed. Posterior probabilities were approximated by sampling every 100 trees simulated with the Markov chain Monte Carlo method (MCMC). The initial runs were conducted with eight active MCMC chains, heated at 0.2, and started with a randomly chosen neighbor joining tree. The first MCMC run was iterated for 1 000 000 generations, and three subsequent MCMC simulations were done with 1 000 000 generations, sampling every 100th tree. A majority consensus tree was calculated from the last 7000 sampled trees from a 10 000-tree dataset using all runs to recover the posterior probabilities of the internal nodes with the SUMT command in MrBayes. The results showed the convergence of two independent MCMC chains as described in (Ronquist and Huelsenbeck 2003) with a standard deviation between the two independent chains less than 0.001. The potential scale reduction factors for all convergence statistics approached 1.001 for all parameters. Posterior probability support measures for nodes with less than a 90 percent posterior probability support are not shown.

RESULTS

The Bayesian analyses indicated that *Gigasperma* as currently circumscribed is not monophyletic but that all taxa fall within the agaricoid clade of Matheny et al. (2006). *Gigasperma cryptica* has a well supported affinity to the Cortinariaceae, whereas *G. americanum* and two undescribed taxa have an affinity to the Agaricaceae (FIG. 1). The results of our analysis are

consistent overall with the findings of Matheny et al. (2006), although there are differences in tree topology, potentially because we sampled far fewer taxa than they did.

In our analysis the taxa we placed in *Cryptolepiota* were divided by *Lepiota erminea* (Fr.) Gillet (FIG. 1), although posterior probability support for this position of *L. erminea* in this clade was less than 90%. With further analysis the position of *L. erminea* appeared to stabilize and all three *Cryptolepiota* species were monophyletic with good support. The clade comprising *Cryptolepiota* is supported as being derived from *Lepiota* and the analysis also supports the proposition that all three taxa are distinct species (FIG. 2).

Even though the topology of our phylogram differs somewhat from that published for *Lepiota* by Vellinga (2003), the overall patterns observed in both studies are consistent. In the analysis (FIG. 2) we recovered clusters of *Lepiota* species corresponding to clades 1 and 4 as well as to clades 2/3 combined by Vellinga (2003) with good posterior probability support.

DISCUSSION

Based on our phylogenetic analysis, the genus *Gigasperma* as currently understood is polyphyletic. *Gigasperma cryptica*, which is known from New Zealand, has affinities to the Cortinariaceae, whereas *G. americanum* and two morphologically similar undescribed taxa from North America belong in the Agaricaceae (FIG. 1). The three North American species are closely related and are recently derived from *Lepiota* clade 1 of Vellinga (2003) (FIG. 2).

We propose the genus *Cryptolepiota* to accommodate the North American taxa. The three *Cryptolepiota* species share unique ecological and morphological attributes and form a monophyletic lineage that appears to represent radiation within *Lepiota*. *Cryptolepiota* is the result of budding, the divergence of a new lineage from an extant parental taxon (Mayr and Bock 2002). As a taxon *Cryptolepiota* differs sharply from its parental taxon macroscopically, microscopically and in its dispersal ecology, indicating that significant evolutionary change has taken place. The use of an independent genus for these species accordingly is warranted.

The recognition of *Cryptolepiota* makes *Lepiota* paraphyletic, however we agree with Hörandl and Steussy (2010) that paraphyletic groups are natural units of classification that arise automatically as taxa evolve within monophyletic groups. Strict adherence to cladistic classification (in this case classifying the sequestrate taxa as species of *Lepiota*) has the advantage of being clear-cut. However when paraphyly is rigorously avoided during classification

TABLE I. Species and GenBank accession numbers for the sequence data used to generate the phylograms (Figs. 1, 2)

Taxon	ITS	LSU
<i>Agaricus bitorquis</i>	AF482829	AY176342
<i>Agaricus campestris</i>	DQ486682	DQ110871
<i>Agrocybe erebia</i>	DQ484056	DQ457663
<i>Bolbitius vitellinus</i>	DQ200920	AY691807
<i>Calvatia craniiformis</i>	DQ112625	DQ112625
<i>Calvatia gigantea</i>	AJ617492	AF518603
<i>Chlorophyllum agaricoides</i>	DQ200928	AY700187
<i>Chlorophyllum molybdites</i>	AY243618	U85303
<i>Chlorophyllum rachodes</i>	AY243608	AY176345
<i>Clitocybe nebularis</i>	DQ486691	DQ457658
<i>Coprinellus disseminates</i>	AY787669	AF041525
<i>Coprinopsis atramentaria</i>	DQ486694	DQ457661
<i>Coprinus comatus</i>	AY854066	AY635772
<i>Coprinus comatus</i>	U85334	AF041529
<i>Coprinus sterquilinus</i>	AF345821	AF041530
<i>Cortinarius exlugubris</i>	GU233338	GU233409
<i>Cortinarius olivaceofuscus</i>	AY669585	AY669585
<i>Cryptolepiota americana</i> (UTC00143916)	HQ020412	EU130550
<i>Cryptolepiota menzei</i> (Trappe 7311)	HQ020413	HQ020417
<i>Cryptolepiota menzei</i> (Trappe 18025)	HQ020414	HQ020418
<i>Cryptolepiota microspora</i> (Trappe 17584)	HQ020410	HQ020420
<i>Cryptolepiota microspora</i> (UTC 253060)	HQ020411	HQ020421
<i>Cystoderma amianthinum</i>	DQ192177	DQ154108
<i>Descolea maculate</i>	DQ192181	DQ457664
<i>Endoptychum</i> sp.	HQ020416	HQ020419
<i>Galerina semilanceata</i>	DQ486706	AY038309
<i>Gigasperma cryptica</i> (Trappe 12562)	HQ020415	EU130549
<i>Hebeloma velutipes</i>	AY818351	AY745703
<i>Hypholoma sublateritium</i>	AY818349	AY635774
<i>Inocybe asterospora</i>	HM060326	HM060325
<i>Laccaria bicolor</i>	FJ845417	EU522777
<i>Laccaria ochropurpurea</i>	FJ168600	AY700200
<i>Lepiota aspera</i>	AY176354	AY207219
<i>Lepiota clypeolaria</i>	AY176361	AY176362
<i>Lepiota cristata</i>	GQ203806	DQ457685
<i>Lepiota cristatoides</i>	AY176363	AY176364
<i>Lepiota echinella</i>	AY176366	AY176367
<i>Lepiota ermine</i>	AY176357	AY176358
<i>Lepiota forquignonii</i>	AY176370	AY176371
<i>Lepiota fuscovinacea</i>	AY176372	AY176373
<i>Lepiota hystrix</i>	AY176377	AY176378
<i>Lepiota lilacea</i>	AY176379	AY176380
<i>Lepiota magnispora</i>	AF391023	AY176381
<i>Lepiota ochraceofulva</i>	AY176386	AY176387
<i>Lepiota spheniscispora</i>	AF391004	AY176404
<i>Leucoagaricus americanus</i>	AY176407	AF482891
<i>Leucoagaricus barssii</i>	DQ911600	DQ911601
<i>Leucoagaricus cinerascens</i>	AY176408	AY176411
<i>Leucoagaricus leucothites</i>	AF482865	AY207226
<i>Lycoperdon marginatum</i>	EU833661	AF261485
<i>Lycoperdon perlatum</i>	AJ237627	AJ406583
<i>Macrolepiota clelandii</i>	AF482838	AF482882
<i>Macrolepiota procera</i>	U85310	AF482880
<i>Mallocybe dulcamara</i>	DQ221106	AY700196
<i>Mythicomyces corneipes</i>	DQ404393	AY745707
<i>Pholiota squarrosa</i>	DQ494683	DQ470818

TABLE I. Continued

Taxon	ITS	LSU
<i>Psathyrella rhodospora</i>	DQ267129	AY645058
<i>Stropharia ambigua</i>	AY818350	AY646102
<i>Thaxterogaster campbellae</i>	DQ328102	AF388747
<i>Tricholoma aestuans</i>	DQ494699	AY700197
<i>Tubaria confragosa</i>	DQ267126	AY700190
<i>Tulostoma brumale</i>	EU784434	AF336272

important ecological information or evolutionary processes such as budding are potentially neglected. Phylograms are extremely informative, but they are ultimately an abstraction of the evolutionary processes that would have happened. To be most useful classifications should be capable of integrating morphological or ecological information with the information contained in phylograms.

The idea that sequestrate Basidiomycota have been independently derived multiple times from their epigeous relatives is well established in the mycological literature. The most familiar examples are fungi in genera such as *Gastrospora* or *Rhizopogon* that are derived from the boletes (Kretzer and Bruns 1997, Grubisha et al. 2001). However numerous sequestrate taxa also have been derived from other mushroom groups, such as the Cortinariaceae and the Russulaceae (Miller et al. 2001, Peinter et al. 2001), and some are even derived from less frequently studied groups such as the Albatrellaceae (Albee-Scott 2007a). Evolution of sequestrate or puffball taxa appears to have occurred frequently within the Agaricaceae. The nLSU analysis of Vellinga (2004) shows at least nine sequestrate or puffball groups of fungi derived within this family, although more also are reported (<http://www.indexfungorum.org/names/names.asp>). These lineages are widely scattered among the clades comprising the family. For instance *Longula texensis* (Berk. & M.A. Curtis) Zeller, *Endoptychum depressum* Singer & A.H. Smith and *Gyrophragmium dunali* (Fr.) Zeller are allied to *Agaricus* whereas the stalked puffballs *Montagnea* sp. and *Podaxis pistillaris* (L.) Fr. have been linked to *Coprinus* (Geml 2004, Hopple and Vilgalys 1999). However the most recent results of Vellinga et al. (2011) indicated that *P. pistillaris* and *Coprinus* may not be as closely allied as previously thought. On the other hand the puffball genera *Lycoperdon*, *Calvatia* and sometimes *Bovista* tend to form an independent clade together within the Agaricaceae (Hibbett et al. 1997, Vellinga 2004).

The development of sequestrate fruiting bodies like those of *Cryptolepiota* in conjunction with dispersal by small mammals has been suggested to be adaptive for fungi living in xeric habitats (Thiers 1984, Trappe et al. 2009). The unusually thick basidiospore walls of

the *Cryptolepiota* species also might be an adaptation to survive under dry conditions or to withstand passage through the digestive tracts of small mammals. In this case the emergence of a sequestrate form within an extant parental taxon corresponds to an early stage in the secotioid drive of Albee-Scott where an adaptive response to environmental stress is to become hypogeous (2007b). All three species of *Cryptolepiota* occur at mid-elevation, in relatively arid habitats in the western United States, and speciation within this group potentially resulted from adaptive radiation under xeric conditions after the appearance of the sequestrate form.

Given the phylogenetic affinity of the *Cryptolepiota* species to *Lepiota*, the presence of metachromatic spores in all three species is unusual. Metachromatic spores are characteristic of *Chlorophyllum*, *Macrolepiota*, *Leucoagaricus* and *Leucocoprinus* but not of *Lepiota*. Nonetheless some species of *Lepiota* have metachromatic basidiospores and therefore their presence in *Cryptolepiota* is not totally unexpected (Vellinga 2003, Lange 2008a). Based on the phylogenetic studies of Vellinga (2003, 2004), metachromatic spores have evolved several times in the Agaricaceae because the genera with metachromatic spores appear in different clades. Vellinga (2003) indicated that even within *Lepiota* species such as *L. cristatoides* Einhell. *L. ochraceofulva* P.D. Orton, or *L. lilacea* Bres. with metachromatic spores occur in different clusters. Taxa such as *L. erminea* or *L. clypeolaria* (Bull.) P. Kumm. in Clade 1 of Vellinga (2003) that are closely related to *Cryptolepiota* do not have metachromatic spores (Lange 2008a). Of note, recent work by Vellinga et al. (2011) indicated that *Lepiota* might be polyphyletic so that the classification of species currently placed in this genus probably will be re-evaluated as future work reveals more about the relationships among these fungi.

The taxa in *Cryptolepiota* are nearly indistinguishable macroscopically. All have fragile, hypogeous basidiomata with a thin, white peridium and a loosely labyrinthiform gleba that is grayish when fresh. However their microscopic characters differ and *Cryptolepiota americana* can be easily separated from the other two by its relatively large, globose spores (FIG. 3) and mucronate cystidia (FIG. 4). The spores

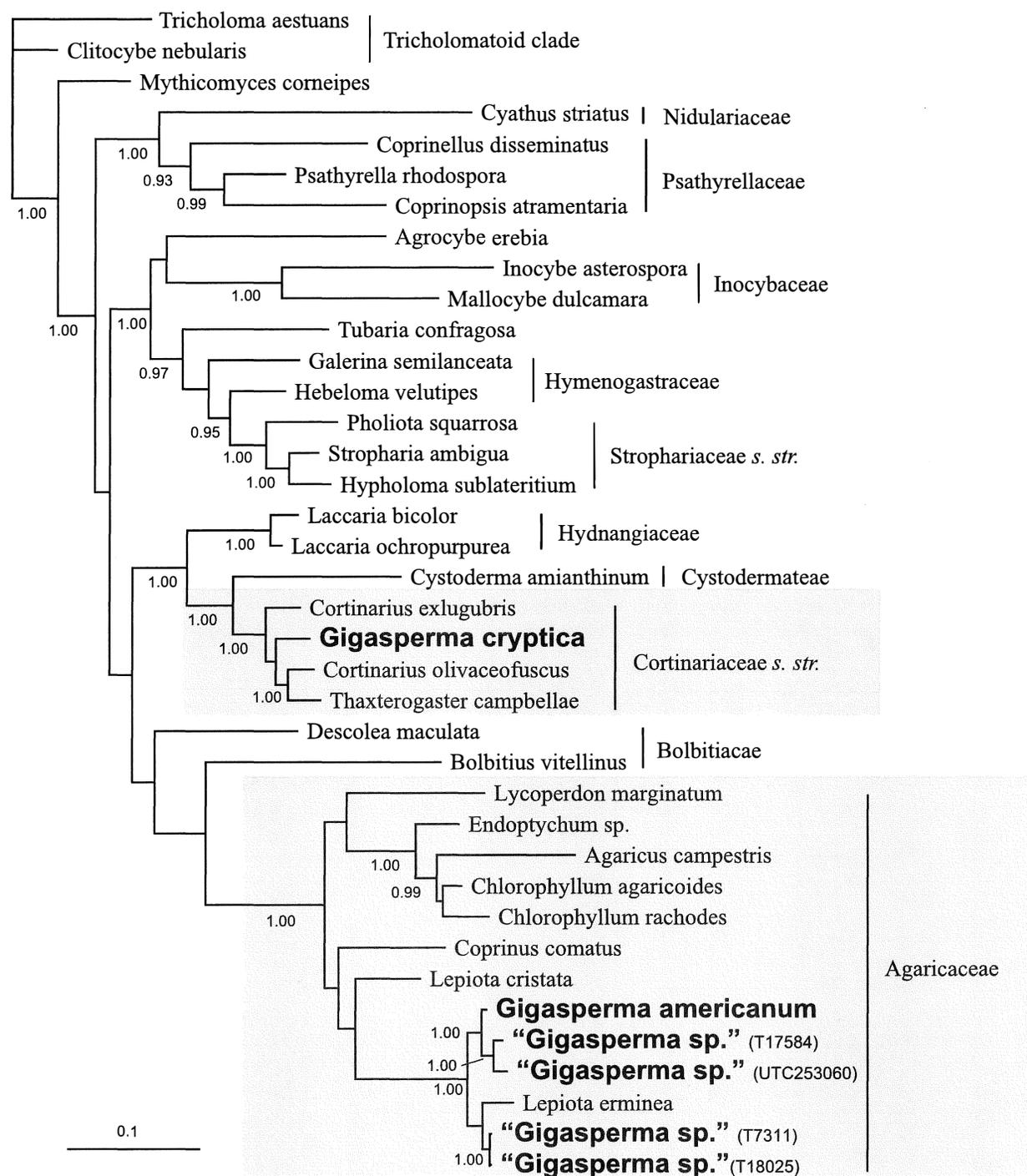


FIG. 1. Bayesian phylogram done to assess the monophyly of *Gigasperma*. Combined nLSU and ITS data were used for the analysis, and members of the tricholomatoid clade were used to root the phylogram. Clades are labeled following Matheny et al. (2006). Support measures for nodes having less than 90% posterior probability support are not shown.

of *C. americana* and *C. microspora* are similar in shape but differ in size and spore wall thickness. Separating *C. microspora* from *C. menzei* is more difficult because their spore characters are much more similar and

both lack cystidia. DNA sequences readily separate them, although the two taxa can be differentiated by differences in spore shape and by sterigmata. The spores of *C. menzei* are typically broadly elliptical (Q

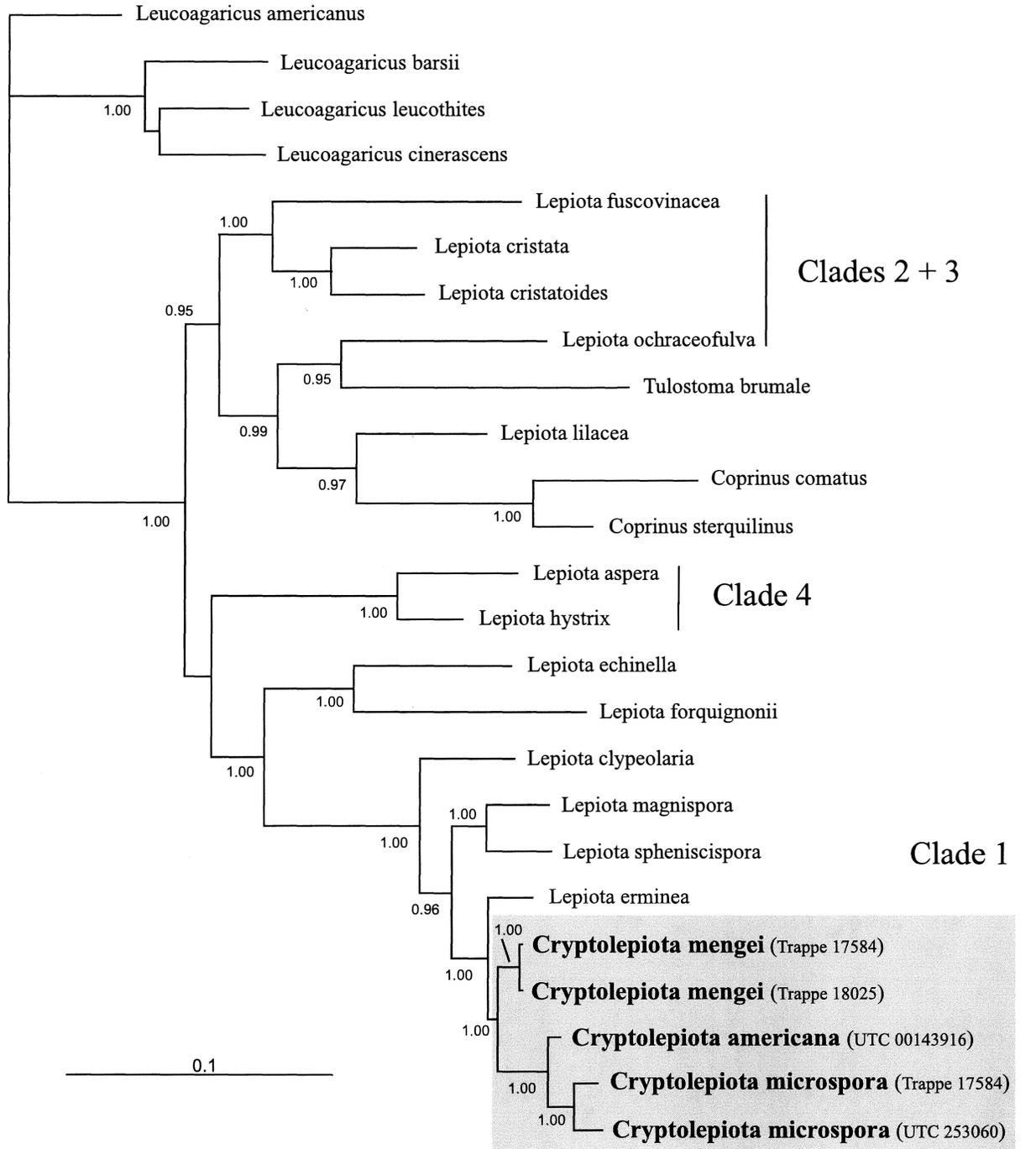


FIG. 2. Phylogram showing the relationship of three *Cryptolepiota* species to *Lepiota*. Bayesian analysis was done to generate the phylogram using combined nLSU and ITS sequences, and members of *Leucoagaricus* were used to root the phylogram. Support measures for nodes having less than 90% posterior probability support are not shown. The labeled clades on the phylogram correspond to those recovered by Vellinga (2003).

= 1.23–1.31), with a *Q* value that differs significantly ($P = 0.01$) from that of *C. microspora*, which has spores that are usually globose to subglobose ($Q = 1.05–1.15$) using a *t*-test (FIG. 3). In addition the

basidia of *C. mengei* typically have four sterigmata that are thick but become granular in appearance, not thick and glassy like those of *C. microspora* and *C. americana* (FIG. 4b, c).

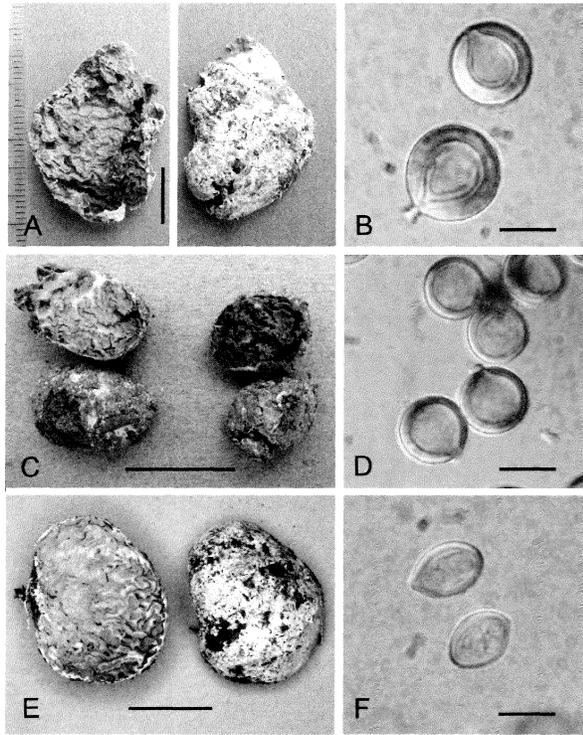


FIG. 3. Comparison of basidiomata and basidiospores for *Cryptolepiota* species. A. Basidiome of *C. americana* (UTCF-00579). Bar = 1 cm. B. Basidiospores of *C. americana*. Bar = 10 µm. C. Basidiomata of *C. microspora* (UTC 253060). Bar = 1 cm. D. Basidiospores of *C. microspora*. Bar = 10 µm. E. Basidiome of *C. menzei* (Trappe 18025). Bar = 1 cm. F. Basidiospores of *C. menzei*. Bar = 10 µm.

The distinctive single-sterigmate basidia of *C. americana* are consistent for all collections we have seen and occur only in this species, whereas basidia with one and two sterigmata occur in *C. microspora*. Variation in basidia structure often accompanies the development of an enclosed hymenium with passive spore discharge (Miller and Miller 1988). For example the number of sterigmata varies within species of *Lycoperdon* and *Calvatia*. Thus the variable numbers of sterigmata produced on the basidia of members of *Cryptolepiota* may relate to development of an enclosed hymenium.

The distinctive smooth, globose spores of *C. cryptica* are unusual for the Cortinariaceae and do not immediately suggest an affinity to that family. Its spores resemble those of the *Cryptolepiota* spp. (FIGS. 3, 5) but are almost twice as large as spores of *C. americana*. *Gigasperma* and *Cryptolepiota* also are differentiated from one another by their gleba structure; that of *Gigasperma* consists of enclosed chambers whereas the gleba of *Cryptolepiota* spp. is loosely labyrinthiform (FIGS. 3, 5).

The phylogenetic affinity between *Gigasperma cryptica* and *Cortinarius* species, which are almost all ectomycorrhizal, indicates that *C. cryptica* may form ectomycorrhizae with *Nothofagus* spp. In contrast, members of the Agaricaceae are not known to form ectomycorrhizal symbioses. Because *Cryptolepiota* species belong to this family they are probably not ectomycorrhizal.

TAXONOMY

Cryptolepiota Kropp & Trappe, gen. nov.

Mycobank MB511131

Fructificaciones hypogaeae, globosae vel ovoideae; gleba fragilis, loculis labyrinthiformibus; basidia 1–4 spora; spora, globosae vel late ellipticae, 10–20 × 7.5–19.5 µm, membrana crassa. Typus genericus *Cryptolepiota americana* (Kropp & Hutchison) Kropp & Trappe.

Etymology: *Cryptolepiota* refers to the hypogeous (hidden) nature of its three species and its phylogenetic relationship to the agaricoid genus *Lepiota*.

Cryptolepiota americana (Kropp & Hutchison) Kropp & Trappe, comb. nov.

FIGS. 3a, b, 4

Mycobank MB511132

= *Gigasperma americanum* Kropp & Hutchison, Mycologia 88:662–665. 1996. Basionym.

Basidiomata 6–30 × 4–21 mm, subglobose to ovoid, hypogeous, fragile; both gleba and peridium negative with KOH and FeSO₄. Peridium white, thin, finely felt-like with soil particles adhering and embedded, 195–255 µm thick, consisting of interwoven hyphae 2.5–10 µm wide, small clamps common, some hyphae of the outer layer slightly encrusted, gradually grading into trama tissue. Gleba loosely labyrinthiform with plate-like layers of fertile tissue, gray when fresh becoming pale brown when dried; trama plates 66–129 µm wide, of irregularly shaped pseudoparenchymatous cells 8–21 µm wide. Basidiospores 14–(18.2)–20 × 14–(17.1)–19.5 µm, Q = 1.06, globose, spore wall 2.5–4.5 µm thick, metachromatic, many spores with a distinct dextrinoid reaction. Basidia 29–32 × 10–11.5 µm, broadly clavate with single, distinctive, thick, refractive sterigmata 4–7 × 2 µm. Cystidia 27–29 × 10 µm, hyaline, thin-walled, mucronate, not abundant.

Specimens examined: USA. UTAH: Cache County. Near Beaver Mountain Ski Area, under *Cercocarpus ledifolius*, 20 May 1995, B.R. Kropp & L.J. Hutchison (UTCF-00579, HOLOTYPE); Cache County near Beaver Mountain Ski Area, under *Cercocarpus ledifolius*, 21 May 1995, B.R. Kropp & L.J. Hutchison (MICH 00009544, PARATYPE); Summit County. Winter sports area near Park City, under *Cercocarpus ledifolius* and *Quercus gambellii*, 3 Jun 1998, B.R. Kropp (UTC00247364).

Cryptolepiota microspora Kropp & Albee-Scott, sp. nov.

FIG. 3c, d

Mycobank MB563084

Differt a *Cryptolepiota americana* sporis 10–14.5 × 8.5–14.5 µm et globosis vel subglobosis, basidiis monosporis

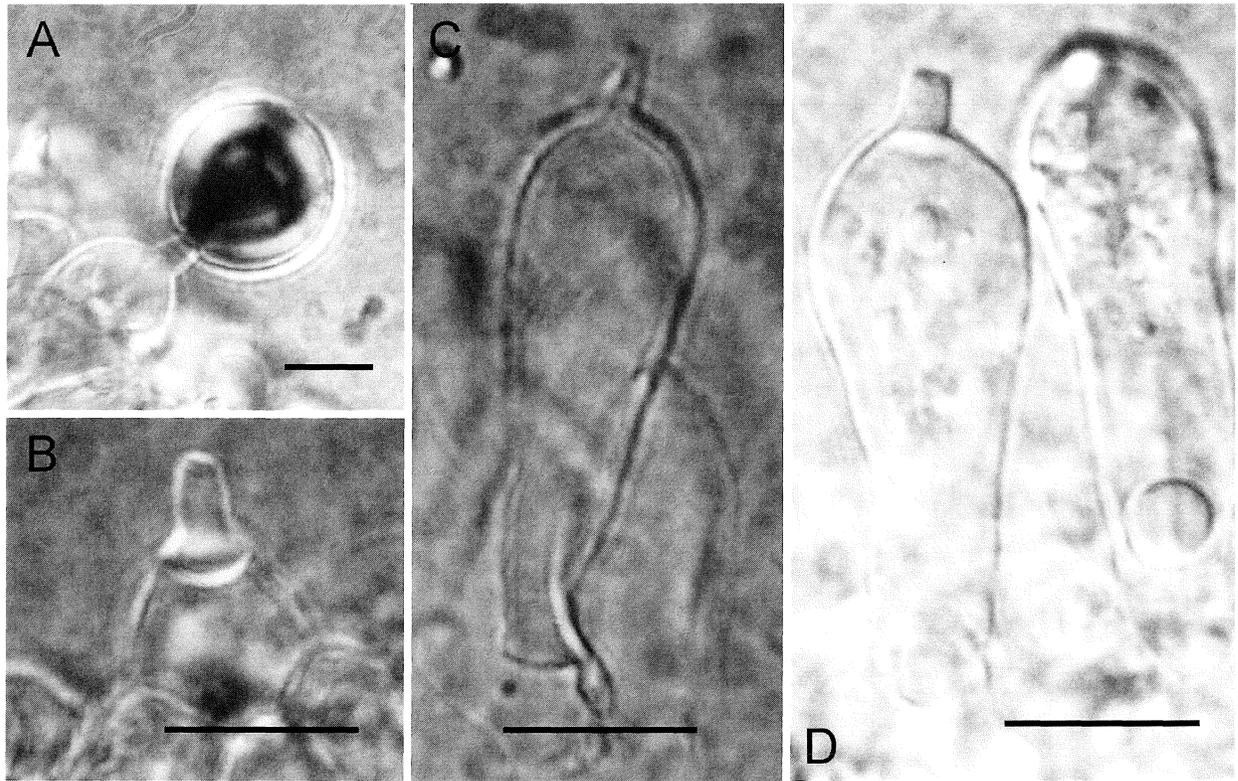


FIG. 4. Microscopic characters for *C. americana*. A. Single-spored basidium with attached basidiospore. Bar = 10 μ m. B. Apex of a basidium showing the thick, refractive sterigma typical for *C. americana*. Bar = 10 μ m. C. Mucronate cystidium, note basal clamp. Bar = 10 μ m. D. Basidium showing the single, thick, refractive sterigma. Bar = 10 μ m.

et bisporis. Holotypus hic designatus 17 Jul 2009, S. Albee-Scott (UTC253060).

Etymology: *microspora* refers to the small spores of this species relative to those of *G. americana*.

Basidiomata 5–14 \times 4–9 mm, subglobose to ovoid, hypogeous, fragile and easily broken when dry. Peridium whitish, thin and more or less felt-like with adhering soil and some soil particles embedded, 150–195 μ m wide, consisting of hyphae 3–5 μ m wide, clamps present but not abundant, gradually grading into trama tissue. Gleba gray when fresh, remaining gray or becoming brown when dried, loosely labyrinthiform with plate-like layers of fertile tissue; tramal plates 72–81 μ m wide, of irregularly shaped pseudo-parenchymatous cells 7–15 μ m wide. Basidiospores 10–(13)–14.5 \times 8.5–(12.5)–14.5 μ m, $Q = 1.05$ –1.15, globose to ovoid, spore wall 1–2.5 μ m thick, metachromatic, only a small proportion of spores dextrinoid. Basidia 25–32 \times 9–10 μ m, broadly clavate, with 1–2 thick refractive, hyaline sterigmata 3–5 \times 2 μ m, identical to those observed for *C. americanum* (FIG. 4b, c). Cystidia not seen, but sterile, clavate, basidium-sized cells that could be either immature basidia or cystidia present in the hymenium.

Specimens examined: USA. UTAH: Rich County. Under *Pinus contorta*, 17 Jul 2009, S. Albee-Scott (UTC 253060, HOLOTYPE); IDAHO: Owyhee County. Slacks Mountain, gregarious in duff under *Cercocarpus*, 22 Jun 1979, E. & J. Trueblood (Trappe 17584, OSC, PARATYPE); CALIFORNIA: Plumas County: Swain Mountain Experimental Forest, 25 Jun 1991, E.A. Geiger (Trappe 11994 OSC, PARATYPE).

Cryptolepiota mengei Kropp & Castellano, sp. nov.

FIG. 3e, f

Mycobank MB563085

Differt a *Cryptolepiota americana* et *Cryptolepiota microspora* sporis 10–16 \times 7.5–12.5 μ m et late ellipticis, basidiis 1–4 sporis et peridiis 275–315 μ m latis. Holotypus hic designatus 23 Apr 1996, E. Cázares (Trappe 18025).

Etymology: *mengei* honors Dr John Menge, accomplished mycologist and mycorrhiza researcher.

Basidiomata 14–40 \times 10–24 mm, ovoid, hypogeous, fragile and easily broken when dry. Peridium whitish, sometimes with salmon hues, thin and somewhat felt-like with adhering soil and embedded soil particles, 275–315 μ m wide, of hyaline hyphae 4–6 μ m wide that have scattered clamps and are sometimes slightly encrusted, gradually grading into trama tissue. Gleba

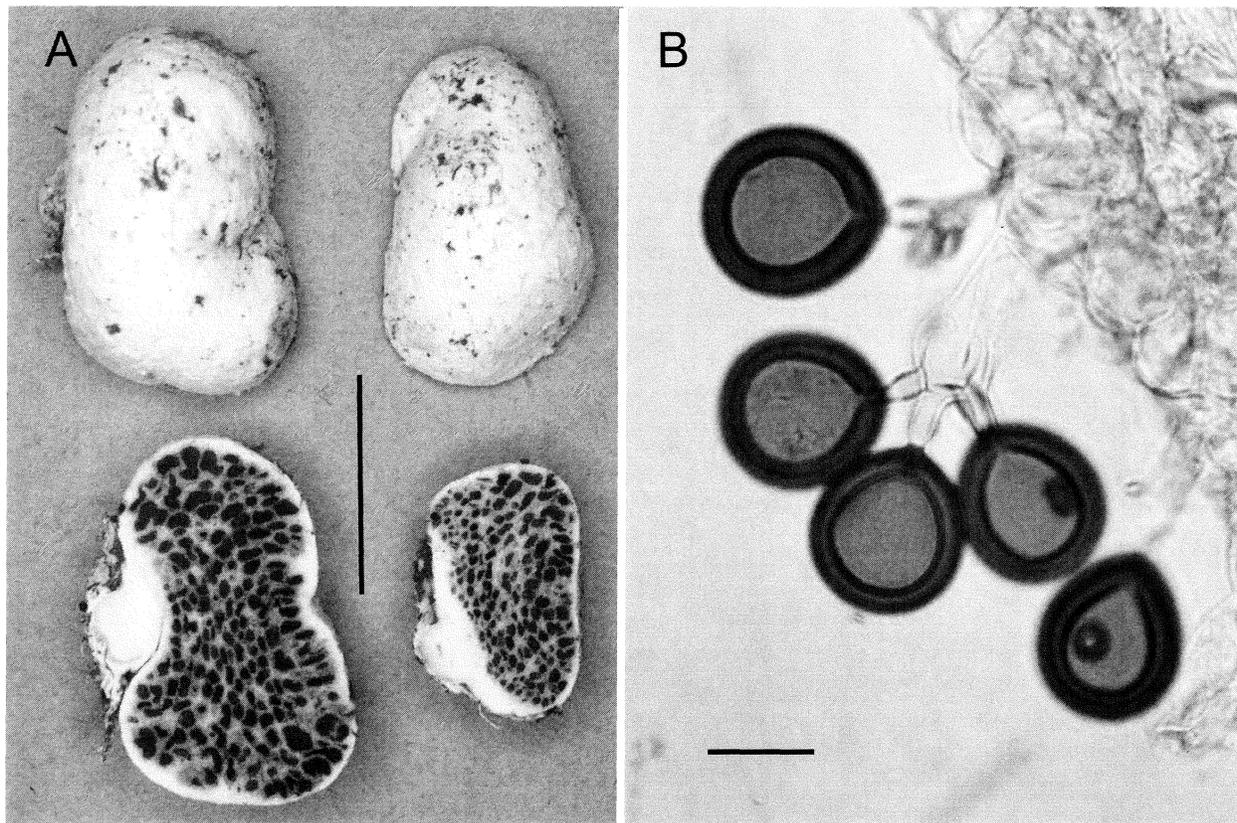


FIG. 5. *Gigasperma cryptica* (Trappe 12562, UTC00247365). A. Basidiome showing the enclosed chambers within the gleba. Bar = 5 mm. B. Basidium with the globose spores typical of the species. Bar = 20 μ m.

pale olive-gray or with greenish yellow hues when fresh, pale brown when dry, loosely labyrinthiform with plate-like layers of fertile tissue; tramal plates 150–195 μ m wide of irregularly shaped, often enlarged pseudoparenchymatous cells 6–28 μ m wide. Basidiospores 10–(13.8)–16 \times 7.5–(10.5)–12.5 μ m, $Q = 1.23$ –1.31, subglobose to ovoid, spore wall 0.5–2 μ m thick, metachromatic, dextrinoid reaction variable, but most collections have at least a few dextrinoid spores. Basidia 18–28 \times 9–12 μ m, broadly clavate, mostly with four sterigmata, sometimes with 1–3 sterigmata, these somewhat thick and 4–5 \times 1 μ m but appearing granular when mature. Cystidia not seen.

Specimens examined: USA. CALIFORNIA: Ventura County. Los Padres National Forest, California 33 milepost 40, 23 April 1996, *E. Cázares* (Trappe 18025, OSC, HOLOTYPE); San Bernadino County. San Bernadino Mountains, Lake Gregory area, 31 May 1981, *T. Dinoff* (Trappe 17662, OSC, PARATYPE); San Bernadino County. San Bernadino Mountains, Strawberry Peak near Lake Arrowhead, 2 Jun 1982, *T. Dinoff* (Trappe 7311, OSC, PARATYPE); Los Angeles County. Angeles National Forest, 17 Apr 1993, *M. Castellano* (Trappe 12984, OSC, PARATYPE); UTAH: Rich County.

Above Bear Lake along US 89 under *Cercocarpus ledifolius*, 22 Jun 1997, *B.R. Kropp* (UTC 253524, PARATYPE).

KEY TO SPECIES OF CRYPTOLEPIOTA

1. Basidiospores 14–20 μ m wide, globose; spore walls 2.5–4.5 μ m thick, mucronate cystidia present in the hymenium *C. americana*
1. Basidiospores typically less than 14 μ m wide, globose to ovoid; spore walls \leq 2.5 μ m thick, mucronate cystidia absent from the hymenium 2
 2. Basidiospores globose to ovoid, 10–14.5 \times 8.5–14.5 μ m ($Q = 1.05$ –1.15); basidia with 1–2 thick, refractive sterigmata resembling those of *C. americana* (see FIG. 4b, d) *C. microspora*
 2. Basidiospores typically broadly ellipsoid, 10–16 \times 7.5–12.5 μ m ($Q = 1.23$ –1.31); most basidia with four sterigmata but sometimes fewer, these sometimes thick but typically not refractive as in FIG. 4b, d *C. mengi*

Gigasperma cryptica Horak, N. Zeal. J. Bot. 9:463–493. 1971. See also Castellano and Trappe 1992. FIG. 5

Basidiome 6 × 25 mm, ovoid to pyriform and somewhat irregular, hypogeous. Peridium white turning cream when dried, dry, smooth to slightly felt-like, 40–70 μm thick when dry, consisting of repent, often compacted hyphae 2–7 μm wide; clamps common. Gleba consisting of dull to reddish brown, closed chambers that are variable in size and shape, up to 0.7 mm across with grayish, sterile tissue between, consisting of irregularly sized cells 13–37 × 12–5 μm; sterile pad at base with a few, scant rhizomorphs attached; columella absent. Basidiospores 30–(36)–41 × 28–(33)–40 μm, Q = 1.08, mostly globose, spore wall 4–8 μm thick, sterigmal attachment conspicuous, some spores dextrinoid, not metachromatic. Basidia 40–60 × 10–30 μm, elongated, mostly four-spored, sometimes three-spored, collapsing in dried material. Cystidia absent.

Specimen examined: NEW ZEALAND. South Island: Canterbury, Craigieburn Forest under *Nothofagus cliffortoides*, 21 Sep 1992, J.M. Trappe (Trappe 12562, UTC00247365).

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