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A new genus to accommodate *Gymnopus acervatus* (Agaricales)

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Abstract: Phylogenies based on ITS and LSU nrDNA sequences show *Agaricus* (*Gymnopus*) *acervatus* as unique within the *Gymnopus/Rhodocollybia* complex. These phylogenies imply that a separate genus is necessary, and *Connopus* is proposed. Intraspecific morphological and DNA-based variation within *C. acervatus* suggests that a western North American clade might be reproductively isolated from the eastern North American/Scandinavian clade and that in this species complex the European and eastern North American clade might be conspecific. A Scandinavian exemplar is selected for bar-coding. Two GenBank sequences with name-phylogenetic placement inconsistencies are identified.

Key words: bar-coding, biogeography, intraspecific variation, *Rhodocollybia*, taxonomy, Tricholomataceae

INTRODUCTION

Recent contributions to systematics of Agaricales have provided multigene phylogenetic reconstructions (Hibbett 2006; Hibbett et al. 2007; Matheny et al. 2006, 2007; Moncalvo et al. 2002) varying widely in scope. Additional phylogenies at the branch tips of these phylogenies have elucidated smaller taxonomic groups, Grand 2004 (*Lentinus/Panus*); Hedh et al. 2008 (*Paxillus involutus*); Hughes et al. 2001 (*Collybia*); Hughes et al. 2007 (*Megacollybia*); Hughes and Petersen 2004 (*Lentinellus*); Jin et al. 2001 (*Panellus*); Krueger 2002 (*Polyporus*); Lickey et al. 2003 (*Arto-Artomyces/Clavicornia*); Mata et al. 2004 (*Rhodocollybia*), 2007 (Omphalotaceae); and Redhead et al. 2001 (coprinoid mushrooms). An ancillary effect of fine-grain studies at tree tips has been to identify anomalies for further research. One such anomaly is *Gymnopus acervatus*.

Gymnopus acervatus forms densely caespitose or connate basidiomata (FIG. 1) on reddish brown stipes 4–16 cm high, often found in moss on rotting conifer logs or stumps in moist northwestern North America,

eastern North America and western Europe. In traditional morphology-based systematic treatments of Agaricales (more recently known as euagarics) *Agaricus acervatus* Fries has been among species considered “collybioid”. Once Fries (1836:92) recognized segregate genera from *Agaricus*, *A. acervatus* was accepted as belonging in subg. *Levipedes* of *Collybia*. Kühner and Romagnesi (1953) included *M. acervatus* in *Marasmius* sect. *Peronati*, but their concept of *Marasmius* was greatly expanded over that accepted now. Halling (1983) continued placement in *Collybia* sect. *Levipedes*. The species was not mentioned by Singer (1986).

Antonin and Noordeloos (1997) included *G. acervatus* in *Gymnopus* section *Vestipedes* together with *G. confluens* (Pers.:Fr.) Antonin et al., *G. peronatus* (Bolt.:Fr.) Antonin et al., *G. luxurians* (Peck) Murrill, *G. putillus* (Fr.:Fr.) Antonin et al. and others. Diagnostic characters for sect. *Vestipedes* were (p 22): “Stipe surface hairy or tomentose; pileipellis a simple cutis without rameales- or dryophila-structure.” Further on (p 32) additional characters for subsection *Vestipedes* were: “pileipellis usually a simple cutis with weakly to distinctly coralloid or diverticulate terminal elements; lamellae edge usually sterile with well differentiated cheilocystidia.” Of these characters *G. acervatus* does not exhibit a stipe vesture (the tomentum of the lower stipe cannot be considered as vesture but adventitious and superficial), does not form a pileipellis with coralloid or diverticulate terminal cells and lacks well differentiated cheilocystidia (see Antonin and Noordeloos 1997, FIG. 16 and acknowledged on p 59–60). In short *G. acervatus* is a discordant element in subsection *Vestipedes*.

To separate *G. acervatus* from *Rhodocollybia* it is necessary to circumscribe *Rhodocollybia* or at least to list diagnostic characters for comparison. This task is made more difficult because the *R. maculata* (typus generis) complex differs significantly morphologically and molecularly from the *R. butyracea* complex. Of these two, the molecular sequence *G. acervatus* is more similar to that of the *R. butyracea* complex.

Three relatively recent significant papers have summarized taxonomic characters of *Rhodocollybia*. Although numerous characters could be applied to multiple genera, these limiting characters appear in all these summaries: (i) “spore print pinkish yellow to pale pinkish brown, never white” (Antonin and Noordeloos 1997, p 115; Knudsen and Vesterholt 2008, p 307); (ii) occasional spores dextrinoid

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FIG. 1. *Connopus acervatus* TENN061292 from Newfoundland, Canada. Numerals on the label are 4 mm high.

(Antonin and Noordeloos 1997, p 115; Mata et al. 2004, p 337; Knudsen and Vesterholt 2008, p 115); (iii) spores cyanophilous, often thick-walled (Knudsen and Vesterholt 2008, p 115; Mata et al. 2004, p 337; Antonin and Noordeloos 1997, p 115); and (iv) “rather large and fleshy basidiomata” (Mata et al. 2004, p 337) or “usually fleshy, putrescent” (Antonin and Noordeloos 1997, p 115).

Spore print of *G. acervatus* is white or off-white, never yellowish or pinkish. Spores of *G. acervatus* are thin-walled, never dextrinoid and never cyanophilous. Basidiomata of *G. acervatus* are hardly fleshy, more mycenoid in stature than rhodocollybioid, and almost always in connate clusters, not gregarious. Comparison of color plates by Antonin and Noordeloos (1997), Knudsen and Vesterholt (2008), Ryman and Holmåsen (1984), Phillips (1991), McNeil (2006) and Trudell and Ammirati (2009) clearly distinguish *G. acervatus* from *R. butyracea* and allied species.

Mata et al. (2006, FIG. 2) included a single ITS sequence for *G. acervatus* in a large phylogeny emphasizing *Gymnopus*. In that analysis unalignable highly variable portions of the ITS sequence were excluded. *Gymnopus acervatus* occurred on a long branch between *Gymnopus* and *Rhodocollybia*, suggesting that it was a unique taxon, but the dataset was so limited that no conclusion could be reached and

discussion concerning placement of *G. acervatus* was almost lacking. Because of uncertainties about relationships based on ITS sequences *G. acervatus* was not included in the remaining phylogenetic analyses based on complete ITS sequence data. In this paper we used nuclear ribosomal ITS and LSU sequences to show that *G. acervatus* is not part of *Gymnopus* s.l. as defined by Mata et al. (2006) but that it apparently falls within Omphalotaceae near *Rhodocollybia*. A new genus, *Connopus*, is proposed and infraspecific clades within *Connopus acervatus* are discussed.

MATERIALS AND METHODS

Abbreviations: PhC = phase contrast microscopy, GSMNP = Great Smoky Mountains National Park, TENN = herbarium of the University of Tennessee, TFB = Tennessee field book, in which notes on fresh specimens are recorded.

Methods for collection and documentation of specimens for DNA extractions and for molecular analyses are given in Mata et al. (2004). PCR primers were ITSIF and ITS4 for the ITS region (Gardes and Bruns 1993, White et al. 1990) and LR0R and LR7 for the nrLSU region (Moncalvo et al. 2002). Sanger dideoxy sequencing primers were ITS5, ITS4, LR5 and LR0R (White et al. 1990, Moncalvo et al. 2002). Cloning was accomplished with the Promega pGEM-T cloning vector and JM109 competent cells using manufacturer directions

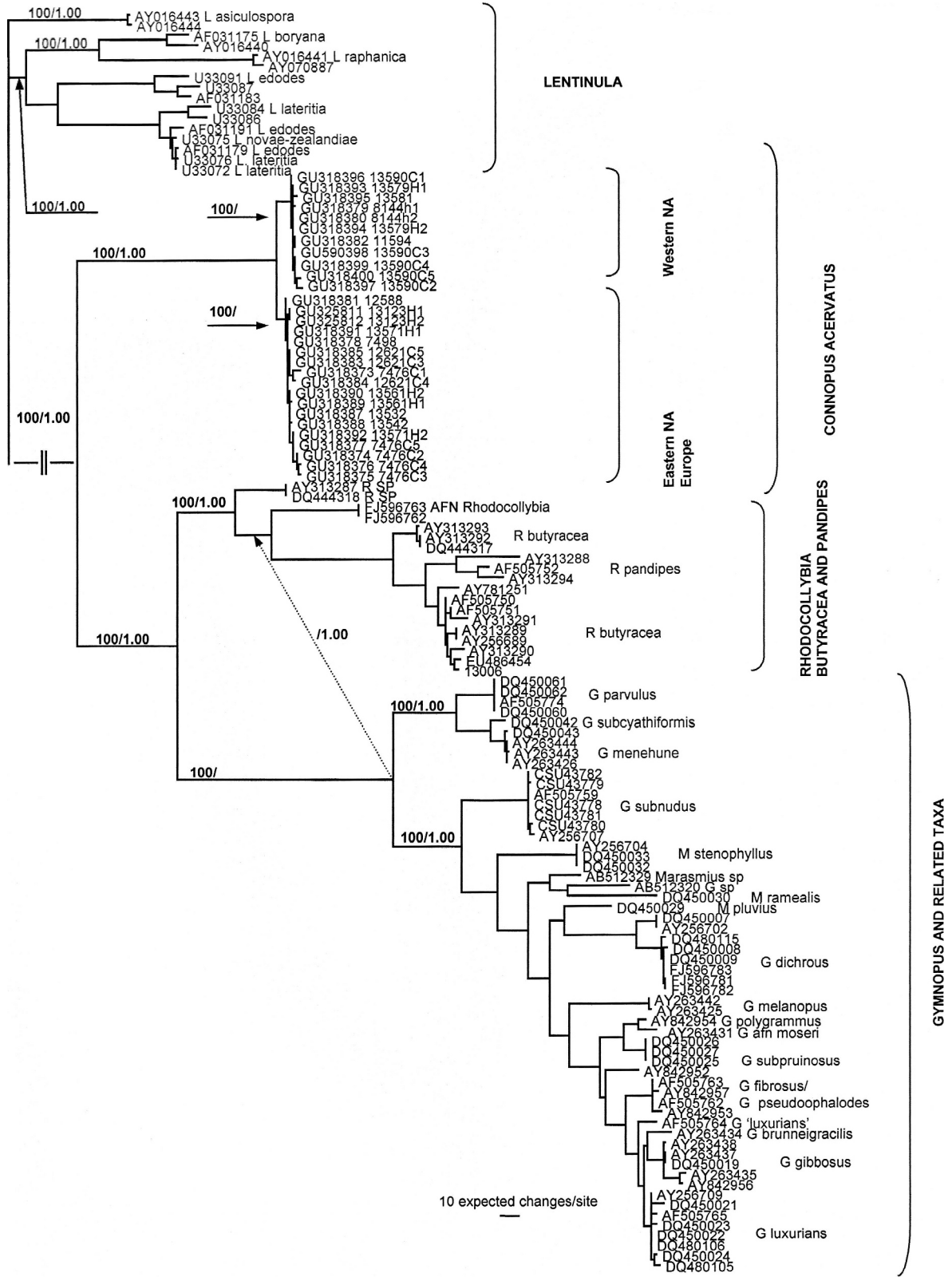


FIG. 2. One of 1000 equally parsimonious trees 1950 bp long based on the ribosomal ITS region. Regions of uncertain alignment were excluded from the analysis (121 bp in ITS1 between bases 214 and 335; 107 bp in ITS2 between bases 753 and 860). Of 792 total characters the number of parsimony informative characters was 511. Gaps were considered informative and were treated as a fifth base. Bootstrap and Bayesian posterior probabilities are to the left of each supported node.

(Promega Corp., Madison, Wisconsin). Cloning was required when a DNA sequence was heterozygous for more than one simple (1–2 bp) insertion or deletion event (indel).

Three datasets were examined: (i) “GCR ITS”, an ITS dataset consisting of the 100 closest ITS sequences to *G. acervatus* in GenBank by BLAST match plus all *G. acervatus* sequences used in this study. (All *Gymnopus* and *Rhodocollybia* sequences used in Mata et al. [2004, 2006] are available through GenBank.) In this dataset 228 characters in unalignable regions of the ITS sequences were excluded from the analyses. (ii) “C ITS” dataset consisting of full length ITS *Gymnopus* (*Connopus*) *acervatus* sequences. (iii) “GCR LSU” dataset consisting of LSU sequences from GenBank within Omphalotaceae. Data from ITS and LSU regions were analyzed separately because of marked differences in variability and non-overlapping datasets.

For both ITS and LSU analyses parsimony analysis was carried out with PAUP* 4b (Swofford 2002). Bootstrap support was computed with 1000 bootstrap replicates. The starting trees were obtained via stepwise addition. One tree was held at each step; MAXTREES was 1000. The branch-swapping algorithm was tree-bisection-reconnection. All characters had equal weight and were unordered. The model selected by Modeltest (Posada and Crandall 1998) for the “C” ITS ribosomal dataset consisting of *Gymnopus* (*Connopus*) *acervatus* sequences was HKY + Γ . The HKY model of evolution provides for equal transition and transversion rates with equal rates among sites (Hasegawa et al. 1985).

The model selected for the “GCR ITS” dataset consisting of *Gymnopus*, *Rhodocollybia*, *G. acervatus* was TVM + I + Γ , which recognizes four transition rates. The TVM model of evolution is not available in MrBayes therefore the GTR + I + Γ model was used for Bayesian analysis. The model selected for the “GCR LSU” dataset was GTR + I + Γ . The general time reversible model assumes a symmetric substitution matrix with each pair of substitutions occurring at a different rate and unequal base frequencies. Bayesian analysis was performed with MrBayes (Huelsenbeck et al. 2001) using two chains and 500 000 generations at settings appropriate to each selected model. Chains converged after approximately 50 000 generations. Bayesian analyses yielded posterior partition probability estimates that largely mirrored bootstrap results from parsimony. Preliminary percent sequence differences were estimated for the *Gymnopus* (*Connopus*) ITS dataset with the uncorrected distances program in GCG (GCG 2000). Sequence pairs showing maximum and minimum sequence divergence were realigned manually, and percent sequence difference was calculated manually from these sequence pairs.

To test whether alternate topologies of the nLSU dataset, in which *G. acervatus* was imbedded within *Rhodocollybia* could be rejected, a constrained tree, in which *G. acervatus* and *Rhodocollybia* were monophyletic sister clades, was constructed in Mesquite (Maddison et al. 2009). This tree was compared with the Bayesian consensus tree (FIG. 4) using the Shimodaira-Hasegawa test (Shimodaira and Hasegawa 1999, Shimodaira et al. 1999). (GenBank accession numbers for sequences used in this study are in TABLE I.)

RESULTS

We include results of phylogenetic analyses based on the 100 closest ITS BLAST matches to *G. acervatus* (FIG. 2). In both Bayesian and parsimony analysis *G. acervatus* forms a well supported clade that is distinct from the *Rhodocollybia butyracea/pandipes* clade and from *Gymnopus*. *Rhodocollybia* is known to be composed of at least two divergent elements, a *Rhodocollybia maculata*-associated clade and a *Rhodocollybia butyracea*-associated clade (Mata 2004, 2006, FIG. 2). The top 100 BLAST matches to *G. acervatus* did not include members of the *R. maculata*-associated clade or several *Gymnopus* species treated in Mata et al. (2006). The placement of *Gymnopus* with respect to the *Rhodocollybia butyracea*-associated clade differed among analyses. In parsimony analysis *Gymnopus* and *Rhodocollybia* were sister clades; in Bayesian analysis *Gymnopus* appeared basal to the *R. butyracea*-associated clade.

Within *G. acervatus* are two distinct, well supported clades (FIGS. 2, 3). Clade 1 is composed of collections from Idaho, Washington and Alaska. Clade 2 is composed of collections from Scandinavia, Newfoundland and Great Smoky Mountains National Park where it is found at high altitudes in relic spruce-fir islands. Percent ITS sequence difference between *G. acervatus* clades 1 and 2 was 2.99–3.80. Within Clade 1 percent sequence difference was 0–1.22%. Within Clade 2 percent sequence difference was 0–0.82%. The two clades are separated by 17 consistent synapomorphies (average ITS region = 710 bp), and there is no current evidence of hybridization between the two clades. BLAST queries in GenBank indicated that neither *Gymnopus* nor *Rhodocollybia* were a good match for *G. acervatus* (closest BLAST match was 84%). ITS sequences for conspecific individuals rarely differ by more than 3% (see Hughes et al. 2009 and references therein).

We recorded results of phylogenetic analyses based on the ribosomal LSU region that included a wider taxon sampling within Omphalotaceae (FIG. 4). In Bayesian analysis *G. acervatus* appears as a long branch between *Rhodocollybia butyracea* and *R. maculata* clades, making *Rhodocollybia* polyphyletic. In parsimony analysis *G. acervatus* is also within the *Rhodocollybia* clade but both *Rhodocollybia butyracea* and *R. maculata* clades are basal to the *G. acervatus* clade and support for this relationship was poor. Within *G. acervatus* Scandinavian and eastern NA collections were basal to collections from Idaho. We constrained *G. acervatus* as a sister clade of *Rhodocollybia* and obtained 100 constrained MP trees. None of the 100 constrained trees were significantly worse than the Bayesian consensus tree with the Shimodaira-Hasegawa test ($P = 0.392\text{--}0.894$).

TAXONOMY

Connopus R.H. Petersen, gen. nov.

Mycobank MB516031

Type species: *Agaricus acervatus* Fries. 1821. Syst. Mycol. 1:122.

≡ *Collybia acervata* (Fr.) Kummer. 1871. Führ. Pilzk.: 114

≡ *Gymnopus acervatus* (Fr.) Murrill. 1916. North Amer. Fl. 9:362.

≡ *Marasmius acervatus* (Fr.) Pears. and Dennis. 1948. Trans. Brit. Mycol. Soc. 31:158.

Basidiomata connata, collybioidea vel mycenoidea. Pileus immaturus convexus, maturus plano-convexus, hygrophanus, levis, pallide-brunneus ad rubro-brunneus. Lamellae librae, secedens, confertis. Stipite rubro-brunneo ad purpureo-brunneo, glabro, fistuloso, cum tomentum album ad basim. Hab: in musci in silvae coniferae in hemispheris septentrionalis.

Basidiomata connate in small to large numbers, collybioid or mycenoid, often with long stipes (FIG. 4). Pileus strongly convex to hemispherical when young, plano-convex by maturity, strongly hygrophanous, smooth, slippery when wet, tan-brown to ruddy brown. Lamellae free to adnexed, seceding early leaving a purplish ring on inner pileus around stipe apex, off-white, close. Stipe ruddy brown to purplish brown, glabrous above, fistulose, developing a white pruina or thin tomentum where protected downward. Flavor and odor negligible. Habitat usually in deep polytrichaceous moss juxtaposed to conifer logs or stumps in temperate to cool forests of the northern hemisphere. Pileipellis a lax trichoderm with occasional, weakly banded, erect terminal cells. Hyphae conspicuously clamped throughout. Basidia four-spored (FIG. 5A). Pleurocystidia absent. Cheilocystidia occasional to absent, slender-lecythiform. Basidiospores (FIG. 5B) small, ellipsoid to cylindrical, hyaline, thin-walled, inamyloid.

Specimens examined.—CANADA: Newfoundland and Labrador, La Nauche Provincial Park, trail to waterfall, 47°10.056'N, 52°53.600'W, 187 m, 15.IX.2006, coll. RHP, TFB 12621 (TENN 61292). FINLAND: Etelä-Häme Prov., Padasjoki, Vesijako Strict Nature Reserve, 15, IX, 1994, coll. RHP, TFB 7476 (TENN53516); Padasjoki, Vesijako Strict Nature Reserve, 15, IX, 1994, coll. RHP, TFB 7491 (TENN53643); RUSSIA: Leningrad Reg., vic. Lodynoe Pole, just outside Nizhnesvirsky Preserve, 30.VIII.1999, coll. RHP, TFB 10646 (TENN 58235); Lodeynopolsky, Kut-Lakhta, 30.VIII.1999, coll. RHP, TFB 10647 (TENN 58236); vic. Lodynoe Pole, just outside Nizhnesvirsky Preserve, 30.VIII.1999, coll. RHP, TFB 10647 (TENN 58236). SWEDEN: Närke, vic. Örebro, Uggelhöjdens Nature Preserve, 59°13.719'N, 14°38.725'E, 11.IX.2008, coll. RHP, A. Methven TFB 13532 (TENN 62879); vic. Örebro, Ullavi klint, 59°22.947'N, 5°00.947'E, 12.IX.2008, coll. RHP, A. Methven, TFB 13542 (TENN 62889); Västergötland, Upphärad, strax SO Kroken, 21.IX.1991, coll. L. & A. Stridvall, TFB 4174 (TENN 50311); vic. Landvetter,

Klippans Nature Reserve, 57°40.0123'N, 12°28.790'E 16.IX.2008, coll. RHP, A. Methven TFB 13571 (TENN 62917); vic. Brobacken, Risveden Nature Reserve, trail to Sälsson Lake, 57°58.317'N, 12°19.016'E, 17.IX.2008, coll. RHP, A. Methven, TFB 13575 (TENN 62921). UNITED KINGDOM: Scotland, Pitlochry, Kindrogan Field Centre, 3.IX.1997, coll. R. Watling, TFB 7000 (TENN 55919); vic. Pitlochry, Kindrogan Field Centre, 56°44.878'N, 003°32.896'W, 3.1997, coll. RHP, TFB 3840 (TENN 55817). UNITED STATES: Alaska, vic. Juneau, Douglas Island, Otter Point Trail, 58°17.980'N, 134°40.119'W, 20.IX.1995, coll. RHP, TFB 8225 (TENN 53990); Idaho, Bonner County, vic. Priest Lake, Spokane Mushroom Club foray, 28.IX/2002, foray collection, TFB 11594 (TENN 59487); McCall County, vic. McCall, vic. Brundage Ski Resort, Bear Basin Road, 5.IX.2008, leg. & det. A.D. Wolfenbarger, ADW 055 (TENN 62824); vic. McCall, Bear Basin Road, vic. Goose Lake Road, 5.IX.2008, coll. S. Trudell, ADW 0054 (TENN 62825); Massachusetts, Hampshire County, Amherst, IX.1964, coll. P.D. Olexia, det. L.R. Hesler, PDO 61 (TENN 28257); Tennessee, Sevier County, GSMNP, Indian Gap, 7.VII.1946, coll. L.R. Hesler, det. A.H. Smith, TENN 17637; Sevier County, GSMNP, Indian Gap, 6.VI.1949, coll. L.R. Hesler, TENN 18992; Sevier County, GSMNP, Indian Gap, Appalachian Trail, 11.VIII.2005, coll. D.J. Lodge, det. E.G. Lickey, TFB 12588 (TENN 61148); Sevier County, GSMNP, Indian Gap, Appalachian Trail north, 30.VII.2006, coll. KW Hughes, TFB 13123 (TENN 61213); Washington, King County, Baring, vic. Barkley's Lake, 30.IX.2002, coll. RHP, TFB 11595 (TENN 59488); Mount Rainier Natl. Park, vic. Longmire, east-facing slope, 26.X.1967, coll. RHP, det. D.E. Stuntz, TENN 30112; Pend Oreille County, Metalline Falls, Forest Service road 310 N of Metalline Falls, 19.XI.2008, coll. D. Parker, TFB 13590 (TENN 62990); Whatcom County, vic. Acme, between Mosquito Road and Timling Falls, 2.X.2002, coll. RHP, TFB 9869 (TENN 58494); Whatcom County, Lake Louise Road, government land, 9.X.1992, coll. RHP and KWH, TFB 5810 (TENN 52213).

Because the ITS phylogeny segregates two general populations of *C. acervatus* (Euro-Scandinavia-eastern North America and moist western North America), careful examination of representative specimens from these regions was undertaken. Three characters proved somewhat distinctive.

First, using mostly northeastern North American material Halling (1983; <http://www.nybg.org/bsci/col/vestiped.html#acervatus>) described cheilocystidia as uncommon, often buried among basidia, fusoid and variously lobed. His illustration depicted one digitate and one somewhat lobed individual. In material from Massachusetts (TENN 28257) and high-altitude southern Appalachian Mountains (TENN 18992, TFB 13123; TFB 12588) such cheilocystidia are common and relatively conspicuous. Antonin and Noordeloos (1997) described cheilocystidia as “absent or present, 18–60 × 2–9 μm, subcylindrical, clavate, fusiform or slightly coralloid”,

TABLE I. GenBank accession numbers for sequences used in this study

| GenBank accession number | Herbarium number | Collection number or strain | Species in tree | Geographic origin |
|--------------------------|------------------|-----------------------------|-----------------------------------|--------------------------|
| AY745709 (LSU) | CUW-PBM2201 | PBM2201 | <i>Anthracophyllum archeri</i> | Not given |
| AF261324 (LSU) | TENN56925 | TFB4419 | <i>Anthracophyllum lateritium</i> | USA, Louisiana |
| AF261327 (LSU) | JMCR.143 | JMCR.143 | <i>Caripia montagnei</i> | Not given |
| DQ444310 (ITS) | TENN59487 | TFB11594 | <i>Connopus acervatus</i> | USA, Idaho |
| GU318382 (ITS) | TENN59487 | TFB11594 | <i>Connopus acervatus</i> | USA, Idaho |
| GU318381 (ITS) | TENN61148 | TFB12588 | <i>Connopus acervatus</i> | USA, GSMNP |
| GU318383-85 (ITS) | TENN61292 | TFB12621c3-c5 | <i>Connopus acervatus</i> | Canada, Newfoundland |
| GU325811-12 (ITS) | TENN61213 | TFB13123 | <i>Connopus acervatus</i> | USA, Tennessee, GSMNP |
| GU325811-12 (ITS) | | TFB13123h1, h2 | <i>Connopus acervatus</i> | USA, GSMNP |
| GU318387 (ITS) | TENN62879 | TFB13532 | <i>Connopus acervatus</i> | Sweden |
| FJ750255 (LSU) | | | | |
| GU318388 (ITS) | TENN62889 | TFB13542 | <i>Connopus acervatus</i> | Sweden |
| FJ750253 (LSU) | | | | |
| GU318389-90 (ITS) | Specimen missing | TFB13561h1, h2 | <i>Connopus acervatus</i> | Sweden |
| GU318391-92 (ITS) | TENN62917 | TFB13571h1,h2 | <i>Connopus acervatus</i> | Sweden |
| GU318393-94 (ITS) | TENN62824 | TFB13579h1,h2 | <i>Connopus acervatus</i> | USA, Idaho |
| FJ750261 (LSU) | | | | |
| GU318395 (ITS) | TENN62825 | TFB13581 | <i>Connopus acervatus</i> | USA, Idaho |
| FJ750260 (LSU) | | | | |
| GU318396-400 (ITS) | TENN62990 | TFB13590c1-c5 | <i>Connopus acervatus</i> | USA, Washington |
| GU318373-377 (ITS) | TENN53516 | TFB7476c1-c5 | <i>Connopus acervatus</i> | Finland |
| FJ750259 (LSU) | | | | |
| GU318378 (ITS) | TENN53596 | TFB7498 | <i>Connopus acervatus</i> | Finland |
| FJ750256 (LSU) | | | | |
| GU318379-80 (ITS) | TENN53962 | TFB8144h1,h2 | <i>Connopus acervatus</i> | USA, Alaska |
| GU318318 (ITS) | TENN61148 | TFB12588 | <i>Gymnopus acervatus</i> | GSMNP Tennessee, USA |
| FJ750259 (LSU) | | | | |
| GU318383-85 (ITS) | TENN61292 | TFB12621 | <i>Gymnopus acervatus</i> | Canada, Newfoundland |
| FJ750254 (LSU) | | | | |
| AF223172 (LSU) | | CBS 174.48 | <i>Gymnopus acervatus</i> (?) | Not given |
| AY639408 (LSU) | SFSU-AWW113 | AWW113 | <i>Gymnopus afn menehune</i> | Not given |
| AY639409 (LSU) | SFSU-AWW10 | AWW10 | <i>Gymnopus afn moseri</i> | Not given |
| AY263431 (ITS) | SFSU AWW10 | AWW10 | <i>Gymnopus afn. Moseri</i> | Not given |
| AY639410 (LSU) | SFSU-AWW118 | AWW118 | <i>Gymnopus aurantipes</i> | Not given |
| AY639411 (LSU) | SFSU-AWW116 | SFSU-AWW116 | <i>Gymnopus bicolor</i> | Not available |
| AF261336 (LSU) | DUKE-RV98/32 | RV98/32 | <i>Gymnopus bififormis</i> | Not available |
| DQ450056 (LSU) | TENN58624 | TFB11016 | <i>Gymnopus bififormis</i> | Costa Rica |
| FJ750264 (LSU) | TENN60951 | TFB12836 | <i>Gymnopus bififormis</i> | New Zealand |
| AY639412 (LSU) | SFSU-AWW01 | AWW01 | <i>Gymnopus brunneigracilis</i> | Not given |
| AY263434 (ITS) | SFSU AWW01 | AWW01 | <i>Gymnopus brunneigracilis</i> | Not given |
| AY207164 (LSU) | GLM 45930 | | <i>Gymnopus confluens</i> | Germany |
| DQ457670 (LSU) | TENN062527 | PBM 2711 | <i>Gymnopus contrarius</i> | USA, Colorado |
| | | AFTOL-ID 1758 | | |
| FJ596783 (ITS) | TENN61128 | TFB12567 | <i>Gymnopus dichrous</i> | USA, NC, GSMNP |
| FJ596782 (ITS) | TENN60673 | TFB12506h1,h2 | <i>Gymnopus dichrous</i> | USA, TN, GSMNP |
| FJ596781 (ITS) | TENN60673 | TFB12506h1 | <i>Gymnopus dichrous</i> | USA, TN GSMNP |
| AY256702 (ITS) | TENN56726 | TFB10014 | <i>Gymnopus dichrous</i> | USA, North Carolina |
| DQ480115 (ITS) | Missing | TFB10829 | <i>Gymnopus dichrous</i> | USA, North Carolina |
| DQ450009 (ITS) | TENN60161 | TFB11554 | <i>Gymnopus dichrous</i> | USA, North Carolina |
| FJ596783 (LSU) | TENN61128 | TFB12567 | <i>Gymnopus dichrous</i> | North Carolina, USA |
| DQ450008 (ITS) | TENN48637 | TFB2028 | <i>Gymnopus dichrous</i> | USA |
| DQ450007 (ITS) | TENN53792 | TFB7920 | <i>Gymnopus dichrous</i> | USA |
| AY639413 (LSU) | SFSU-AR099 | AR099 | <i>Gymnopus diminutus</i> | Not given |
| AF291305 (LSU) | FO 21603 | FO 21603 | <i>Gymnopus dryophilus</i> | Germany |

TABLE I. Continued

| GenBank accession number | Herbarium number | Collection number or strain | Species in tree | Geographic origin |
|--------------------------|------------------|-----------------------------|---|---------------------|
| AF042595 (LSU) | DUKE-RV83/180 | RV83/180 | <i>Gymnopus dryophilus</i> | not available |
| FJ596766 (LSU) | TENN60015 | TFB11786 | <i>Gymnopus dryophilus</i> | Tennessee, USA |
| FJ596767 (LSU) | | | | |
| AY640619 (LSU) | TENN57012 | TFB9952 | <i>Gymnopus dryophilus</i> | North Carolina, USA |
| AJ406564 (LSU) | Not given | GEL4613 | <i>Gymnopus dryophilus</i> incorrect id? | not available |
| FJ750265 (LSU) | TENN61125 | TFB12563 | <i>Gymnopus dysodes</i> | Tennessee, USA |
| DQ449994 (LSU) | TENN59140 | TFB11039 | <i>Gymnopus earleae</i> | Tennessee, USA |
| AY207167 (LSU) | GLM 45932 | GLM 45932 | <i>Gymnopus erythropus</i> | not available |
| AY842953 (ITS) | Not given | PR23TN | <i>Gymnopus fibrosipes</i> | Not given |
| AF505763 (ITS) | TENN56660 | TFB9699 | <i>Gymnopus fibrosipes</i> | Costa Rica |
| AM946450 (LSU) | C-42389 | C42389 | <i>Gymnopus fusipes</i> | Denmark |
| AY639414 (LSU) | NYBG-REH6509 | REH6509 | <i>Gymnopus fusipes</i> | Not given |
| AY256710 (LSU) | TENN59217 | TFB11333 | <i>Gymnopus fusipes</i> | France |
| AY256711 (LSU) | TENN59300 | TFB11439 | <i>Gymnopus fusipes</i> | Austria |
| AF135795 (LSU) | TENN55904 | TFB6985 | <i>Gymnopus fusipes</i> | United Kingdom |
| AY639415 (LSU) | SFSU-AWW12a | AWW12a | <i>Gymnopus gibbosus</i> | Not given |
| AY639417 (LSU) | SFSU-AWW112 | AWW112 | <i>Gymnopus gibbosus</i> | Not given |
| AY263435 (ITS) | SFSU AWW112 | AWW112 | <i>Gymnopus gibbosus</i> | Not given |
| AY263437 (ITS) | SFSU AWW66 | AWW66 | <i>Gymnopus gibbosus</i> | Not given |
| AY263438 (ITS) | SFSU AWW95 | AWW95 | <i>Gymnopus gibbosus</i> | Not given |
| AY842956 (ITS) | Not given | PRH | <i>Gymnopus gibbosus</i> | Not given |
| AY207166 (LSU) | F-GLM 45933 | | <i>Gymnopus hariolorum</i> | Germany |
| AY639418 (LSU) | SFSU-AWW03 | AWW03 | <i>Gymnopus indoctus</i> | Not given |
| DQ449984 (LSU) | TENN52970 | TFB6520 | <i>Gymnopus ioccephalus</i> | North Carolina, USA |
| DQ480106 (ITS) | DUKE VLUX | | <i>Gymnopus luxurians</i> | Not given |
| DQ480105 (ITS) | DUKE 54 | | <i>Gymnopus luxurians</i> | Not given |
| AY639421 (LSU) | SFSU-DEH1304 | DEH1304 | <i>Gymnopus luxurians</i> | Not given |
| AY256709 (LSU) | TENN57910 | TFB10350 | <i>Gymnopus luxurians</i> | USA, North Carolina |
| AY256709 (ITS) | TENN57910 | TFB10350 | <i>Gymnopus luxurians</i> | USA, North Carolina |
| DQ450023 (ITS) | TENN57914 | TFB10355 | <i>Gymnopus luxurians</i> | USA, South Carolina |
| AF505764 (ITS) | TENN58634 | TFB11026 | <i>Gymnopus luxurians</i> | Costa Rica |
| DQ450019 (ITS) | DEH2318 | TFB11585 | <i>Gymnopus luxurians</i> | USA, Hawaii |
| DQ450024 (ITS) | TENN59547 | TFB11711 | <i>Gymnopus luxurians</i> | Dominican Republic |
| DQ450022 (ITS) | TENN50619 | TFB4283 | <i>Gymnopus luxurians</i> | Switzerland |
| DQ450021 (ITS) | TENN50937 | TFB4439 | <i>Gymnopus luxurians</i> | USA, Tennessee |
| AY263442 (ITS) | SFSU AWW50 | AWW50 | <i>Gymnopus melanopus</i> | Not given |
| AY639422 (LSU) | SFSU-AWW54 | AWW54 | <i>Gymnopus melanopus</i> | Not given |
| AY263425 (ITS) | SFSU AWW 54 | AWW54 | <i>Gymnopus melanopus</i> | Not given |
| AY639423 (LSU) | SFSU-AWW02 | AWW02 | <i>Gymnopus menehune</i> | Not given |
| AY639425 (LSU) | SFSU-AWW87 | AWW87 | <i>Gymnopus menehune</i> | Not given |
| AY639424 (LSU) | SFSU-AWW02 | AWW02 | <i>Gymnopus menehune</i> | Not given |
| AY263443 (ITS) | SFSU AWW15 | AWW15 | <i>Gymnopus menehune</i> | Not given |
| AY263444 (ITS) | SFSU AWW87 | AWW87 | <i>Gymnopus menehune</i> | Not given |
| AY263426 (ITS) | SFSU DED5866 | DED5866 | <i>Gymnopus menehune</i> | Not given |
| DQ450043 (ITS) | DEH2320 | TFB11587 | <i>Gymnopus menehune</i> | USA, Hawaii |
| DQ450035 (LSU) | TENN54460 | TFB11005 | <i>Gymnopus mesoamericanus</i> | Costa Rica |
| AY639426 (LSU) | SFSU-AWW05 | AWW05 | <i>Gymnopus nonnulus</i> | Not given |
| DQ450060 (ITS) | TENN58113 | TFB10419 | <i>Gymnopus parvulus</i> | Costa Rica |
| DQ450061 (ITS) | TENN58115 | TFB10421 | <i>Gymnopus parvulus</i> | Costa Rica |
| AF505774 (ITS) | TENN58116 | TFB10422 | <i>Gymnopus parvulus</i> | Costa Rica |
| DQ450062 (ITS) | TENN58119 | TFB10425 | <i>Gymnopus parvulus</i> | Costa Rica |
| AY207168 (LSU) | F-GLM 45934 | | <i>Gymnopus peronatus</i> | Germany |
| AF223173 (LSU) | CBS 426.79 | culture only | <i>Gymnopus peronatus</i> | Not given |
| DQ450017 (LSU) | TENN50540 | TFB4204 | <i>Gymnopus peronatus</i> | Sweden |

TABLE I. Continued

| GenBank accession number | Herbarium number | Collection number or strain | Species in tree | Geographic origin |
|--------------------------|------------------|-----------------------------|-----------------------------------|---------------------|
| AY842954 (ITS) | Not given | PR2542TN | <i>Gymnopus polygrammus</i> | Puerto Rico |
| AF042596 (LSU) | DUKE-RV182.01 | | <i>Gymnopus polyphyllus</i> | Not given |
| AY842957 (ITS) | Not given | PR24TN | <i>Gymnopus pseudomphalodes</i> | Not given |
| AF505762 (ITS) | NYBG REH 7348 | REH 7348 | <i>Gymnopus pseudomphalodes</i> | Not given |
| AY639427 (LSU) | SFSU-AWW126 | AWW126 | <i>Gymnopus sepiiconicus</i> | Not given |
| AF261326 (LSU) | DUKE-JEJ.PR.213 | JEJ.PR.213 | <i>Gymnopus</i> sp. | Puerto Rico |
| AF261334 (LSU) | DUKE-RV.PR.98.08 | PR.98.08 | <i>Gymnopus</i> sp. | Puerto Rico |
| AF261335 (LSU) | DUKE-RVPR98.13 | RVPR98.13 | <i>Gymnopus</i> sp. | Puerto Rico |
| AF261333 (LSU) | DUKE-RVPR98.46 | RVPR98.46 | <i>Gymnopus</i> sp. | Puerto Rico |
| FJ750263 (LSU) | TENN58602 | TFB10494 | <i>Gymnopus</i> sp. nov. | Costa Rica |
| AB512320 (ITS) | Not given | Strain 7090106 | <i>Gymnopus</i> sp. | Japan, Okinawa, |
| DQ450042 (ITS) | TENN59550 | TFB11714 | <i>Gymnopus subcyathiformis</i> | Dominican Republic |
| CSU43779 (ITS) | Not given | JFM 1302 | <i>Gymnopus subnudus</i> | USA |
| CSU43780 (ITS) | Not given | JFM 1480 | <i>Gymnopus subnudus</i> | USA |
| CSU43781 (ITS) | Not given | JFM 1482 | <i>Gymnopus subnudus</i> | USA |
| CSU43778 (ITS) | Not given | JFM 898 | <i>Gymnopus subnudus</i> | USA |
| CSU43782 (ITS) | TENN48353 | TFB1818 | <i>Gymnopus subnudus</i> | USA |
| AF505759 (ITS) | TENN57899 | TFB10338 | <i>Gymnopus subnudus</i> | USA, North Carolina |
| FJ750262 (LSU) | TENN61138 | TFB12577 | <i>Gymnopus subnudus</i> | Tennessee, USA |
| AY256707 (ITS) | Missing | TFB6928 | <i>Gymnopus subnudus</i> | USA. MN |
| AY639429 (LSU) | SFSU-DED6674 | DED6675 | <i>Gymnopus subpruinosis</i> | Not given |
| AY842952 (ITS) | Not given | Isolate PRA | <i>Gymnopus subpruinosis</i> | Puerto Rico |
| DQ450025 (ITS) | TENN59474 | TFB11063 | <i>Gymnopus subpruinosis</i> | USA, Hawaii |
| DQ450027 (ITS) | TENN59477 | TFB11066 | <i>Gymnopus subpruinosis</i> | USA, Hawaii |
| DQ450026 (ITS) | TENN56242 | TFB9529 | <i>Gymnopus subpruinosis</i> | USA, California |
| AY639430 (LSU) | SFSU-AWW106 | AWW106 | <i>Gymnopus termiticola</i> | Not given |
| AY639432 (LSU) | SFSU-AWW127 | AWW127 | <i>Gymnopus vitellinipes</i> | Not given |
| AY016444 (ITS) | TENN58112 | TFB10418 | <i>Lentinula asiculosp.ora</i> | Costa Rica |
| AY016443 (ITS) | TENN56421 | TFB9447 | <i>Lentinula asiculosp.ora</i> | Costa Rica |
| AY016440 (ITS) | | AAR de Meijer #3700 | <i>Lentinula boryana</i> | Brazil |
| AF031175 (ITS) | Not given | R.G. Thorn 960624/09 | <i>Lentinula boryana</i> | Costa Rica |
| AF042579 (LSU) | ATCC42962 | | <i>Lentinula edodes</i> | Not given |
| U33087 (ITS) | TMI1633 | | <i>Lentinula edodes</i> | Thailand |
| AF031191 (ITS) | TMI1546 | | <i>Lentinula edodes</i> | Nepal |
| AF031179 (ITS) | Duke RV95-376 | RV95-376 | <i>Lentinula edodes</i> | Australia |
| AF031183 (ITS) | Not given | Strain STCL125 | <i>Lentinula edodes</i> | China |
| U33084 (ITS) | TMI1485 | | <i>Lentinula lateritia</i> | PNG |
| U33086 (ITS) | TMI1502 | | <i>Lentinula lateritia</i> | PNG |
| U33072 (ITS) | CLARK UNIV | DSH 92-147 | <i>Lentinula lateritia</i> | PNG |
| U33076 (ITS) | TENN50062 | TFB3577 | <i>Lentinula lateritia</i> | Tasmania |
| U33091 (ITS) | TMI818 | | <i>Lentinula nov.ae-zelandiae</i> | Japan |
| U33075 (ITS) | Not given | NZFS 210 | <i>Lentinula nov.ae-zelandiae</i> | New Zealand |
| AY016441 (ITS) | TENN54887 | TFB9156 | <i>Lentinula raphanica</i> | USA, Florida |
| AY016442 | TENN56477 | TFB9564 | <i>Lentinula raphanica</i> | Puerto Rico |
| DQ450029 (ITS) | TENN55766 | TFB9168 | <i>Marasmiellus aff. pluvius</i> | USA, TN GSMNP |
| AY256708 (LSU) | TENN59540 | TFB9889 | <i>Marasmiellus juniperinus</i> | USA, Louisiana |
| AF261330 (LSU) | Not given | HN2270 | <i>Marasmiellus opacus</i> | Not given |
| AF261329 (LSU) | Not given | JEJ.574 | <i>Marasmiellus opacus</i> | Not given |
| DQ450030 (ITS) | TENN50324 | TFB4727 | <i>Marasmiellus rameales</i> | Sweden |
| DQ450030 (LSU) | | | | |
| AY207236 (LSU) | F-GLM 45958 | | <i>Marasmiellus ramealis</i> | Germany |
| AY639435 (LSU) | SFSU-DED5258 | DED5258 | <i>Marasmiellus synodicus</i> | Not given |
| AY639436 (LSU) | BRNM568- | | <i>Marasmius alliaceus</i> | Not given |

TABLE I. Continued

| GenBank accession number | Herbarium number | Collection number or strain | Species in tree | Geographic origin |
|--------------------------|------------------------|-----------------------------|--------------------------------|--------------------------|
| AY207234 (LSU) | F-GLM45959 | GLM 45959 | <i>Marasmius alliaceus</i> | Germany |
| AY635776 (LSU) | TENN55620 | TFB8960 | <i>Marasmius alliaceus</i> | Russia |
| AF261585 (LSU) | Not given | HN4730 | <i>Marasmius androsaceus</i> | Not given |
| AY639437 (LSU) | SFSU-DED6628 | DED6628 | <i>Marasmius applanatipes</i> | Not given |
| AY639438 (LSU) | SFSU-DED6628 | | <i>Marasmius copelandii</i> | Not given |
| AF261332 (LSU) | DAOM175382 | DED6628 | <i>Marasmius scorodonius</i> | Not given |
| AF261331 (LSU) | Not given | JEJ.586 | <i>Marasmius scorodonius</i> | Not given |
| DQ450006 (LSU) | TENN50346 | TFB4749 | <i>Marasmius scorodonius</i> | Switzerland |
| EU522806 (LSU) | Not given | TM03_419 | <i>Marasmius scorodonius</i> | Canada |
| AB512329 (ITS) | Not given | Strain 0801Y51 | <i>Marasmius</i> sp. | Japan, Okinawa, |
| DQ450032 (ITS) | TENN59444 | TFB11558 | <i>Marasmius stenophyllus</i> | USA, North Carolina |
| DQ450033 (ITS) | TENN59449 | TFB11559 | <i>Marasmius stenophyllus</i> | USA, North Carolina |
| AY207240 (LSU) | GLM 45964 | | <i>Micromphale foetidum</i> | Germany |
| AF261328 (LSU) | DUKE-JEJ.VA.567 | | <i>Micromphale foetidum</i> | USA, Virginia |
| AF042628 (LSU) | DUKE-RV83/67 | RV83/67 | <i>Micromphale perforans</i> | Not available |
| AF042577 (LSU) | DUKE-RVPR1308 | RVPR1308 | <i>Neonothopanus nambi</i> | Puerto Rico |
| AF135175 (LSU) | DUKE- RVPR27 | RVPR27 | <i>Neonothopanus nambi</i> | Puerto Rico |
| AF135172 (LSU) | Not given | JM | <i>Omphalotus japonicus</i> | Not given |
| AF042621 (LSU) | Not given | T1946.8 | <i>Omphalotus nidiformis</i> | Not available |
| DQ470816 (LSU) | CBS 102282 | Culture only | <i>Omphalotus olearius</i> | Slovenia? |
| FJ596762 (ITS) | TENN59896 | TFB11778h1 | <i>Rhodocollybia</i> afn. | USA, TN, GSMNP |
| FJ596763 (ITS) | TENN59896 | TFB11778h2 | <i>Rhodocollybia</i> afn. | USA, TN, GSMNP |
| AY639439 (LSU) | SFSU- DLLargent9199 | DLLarge9199 | <i>Rhodocollybia badiialba</i> | Not given |
| AY207163 (LSU) | GLM 46024 | | <i>Rhodocollybia butyracea</i> | Germany |
| AY639440 (LSU) | NYBG-REH6705 | REH6705 | <i>Rhodocollybia butyracea</i> | Not available |
| DQ444317 (ITS) | VT OKM27562 | OKM27562 | <i>Rhodocollybia butyracea</i> | USA |
| EU486454 (ITS) | UBC F16294 | | <i>Rhodocollybia butyracea</i> | Canada |
| AF505750 (ITS) | Missing | TFB10726 | <i>Rhodocollybia butyracea</i> | Russia |
| AY781251 (ITS) | Not given | olrim421 | <i>Rhodocollybia butyracea</i> | Sweden |
| AF505751 (ITS) | TENN59317 | TFB11456 | <i>Rhodocollybia butyracea</i> | Austria |
| AY313291 (ITS) | TENN59317 | TFB11456 | <i>Rhodocollybia butyracea</i> | Austria |
| GU318386 (ITS) | TENN60927 | TFB13006 | <i>Rhodocollybia butyracea</i> | USA, Tennessee, GSMNP |
| FJ740252 (LSU) | | | | |
| GU318386[ITS] | TENN60927 | TFB13006 | <i>Rhodocollybia butyracea</i> | USA, GSMNP |
| FJ750252 [LSU] | | | | |
| FJ750251 (LSU) | TENN61089 | TFB13085 | <i>Rhodocollybia butyracea</i> | New Zealand |
| AY313293 (ITS) | TENN53580 | TFB7452 | <i>Rhodocollybia butyracea</i> | Sweden |
| AY313292 (ITS) | Missing | TFB8250 | <i>Rhodocollybia butyracea</i> | USA |
| AY313290 (ITS) | TENN56303 | TFB8801 | <i>Rhodocollybia butyracea</i> | Mexico |
| AY313289 (ITS) | TENN55660 | TFB9000 | <i>Rhodocollybia butyracea</i> | Turkey |
| AY639441 (LSU) | SFSU-DED5873 | DED5873 | <i>Rhodocollybia laulaha</i> | Hawaii |
| AM946462 (LSU) | TAA147123 | | <i>Rhodocollybia maculata</i> | Estonia |
| AY639880 (LSU) | TENN062341 | PBM2481 | <i>Rhodocollybia maculata</i> | Isolate=AFTOL-ID 540 |
| AF042597 (LSU) | DUKE-RV94/175 | RV94/175 | <i>Rhodocollybia maculata</i> | Not available |
| AF505752 (ITS) | TENN58622 | TFB11014 | <i>Rhodocollybia pandipes</i> | Costa Rica |
| AY313288 (ITS) | TENN59546 | TFB11707 | <i>Rhodocollybia pandipes</i> | Dominican Republic |
| AY313294 (ITS) | TENN53838 | TFB7899 | <i>Rhodocollybia pandipes</i> | USA, North Carolina |
| AY313287 (ITS) | TENN58798 | TFB10712 | <i>Rhodocollybia</i> sp. | Greenland |
| DQ444318 (ITS) | TENN58798 | TFB10712 | <i>Rhodocollybia</i> sp. | Greenland |
| AY207235 (LSU) | GLM 45960 | | <i>Setulipes androsaceus</i> | Germany |

BRNM = Moravian Museum; C = Copenhagen; F = Field Museum of Chicago; NZFS = New Zealand Forest Service; SFSU = San Francisco State University, TENN = University of Tennessee; TMI = Tottori Mycological Institute; h = within collection haplotype; c = clone number.

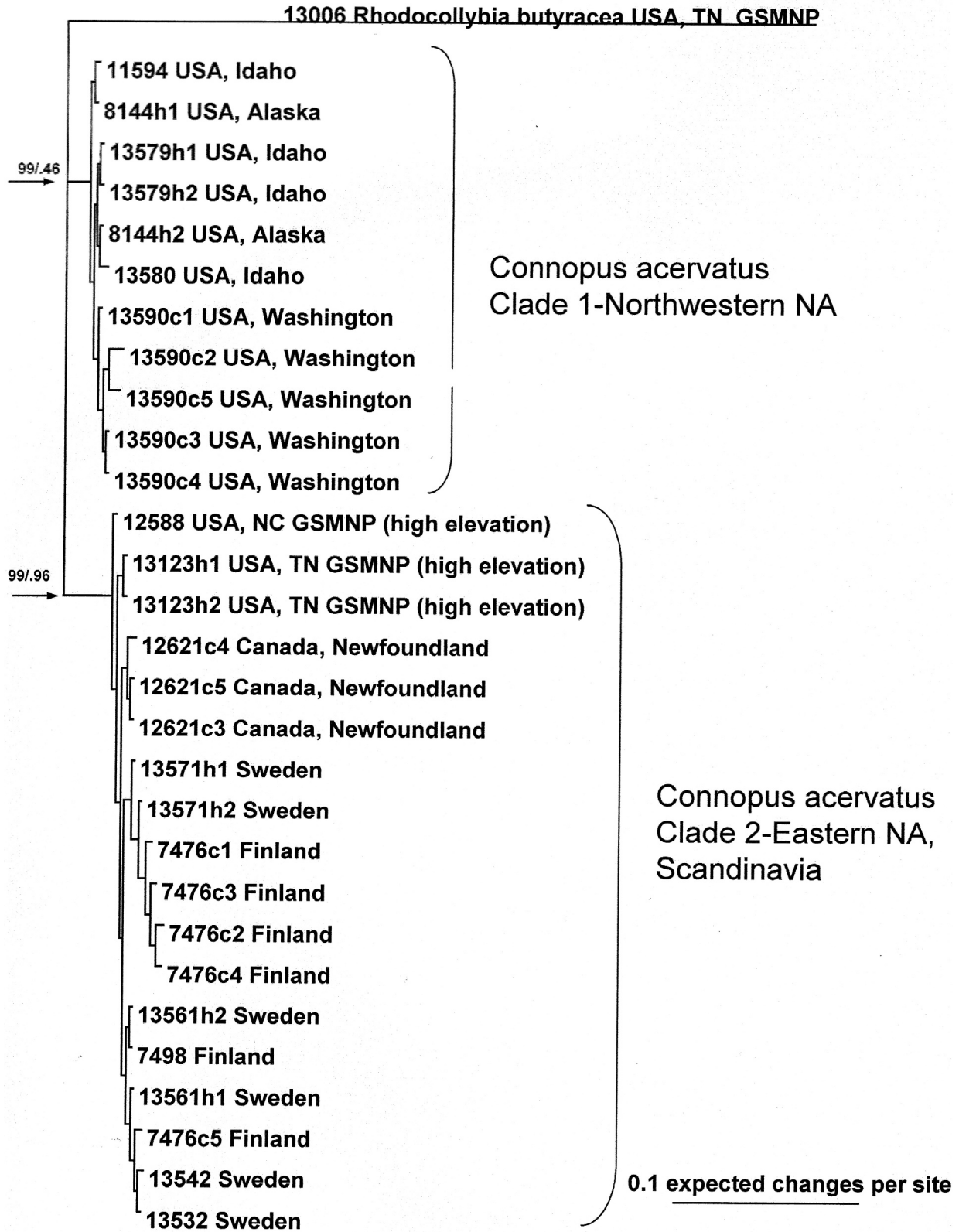


FIG. 3. Bayesian majority rule consensus tree based on complete ribosomal ITS sequences. Bootstrap and Bayesian posterior probabilities are to the left of each supported node.

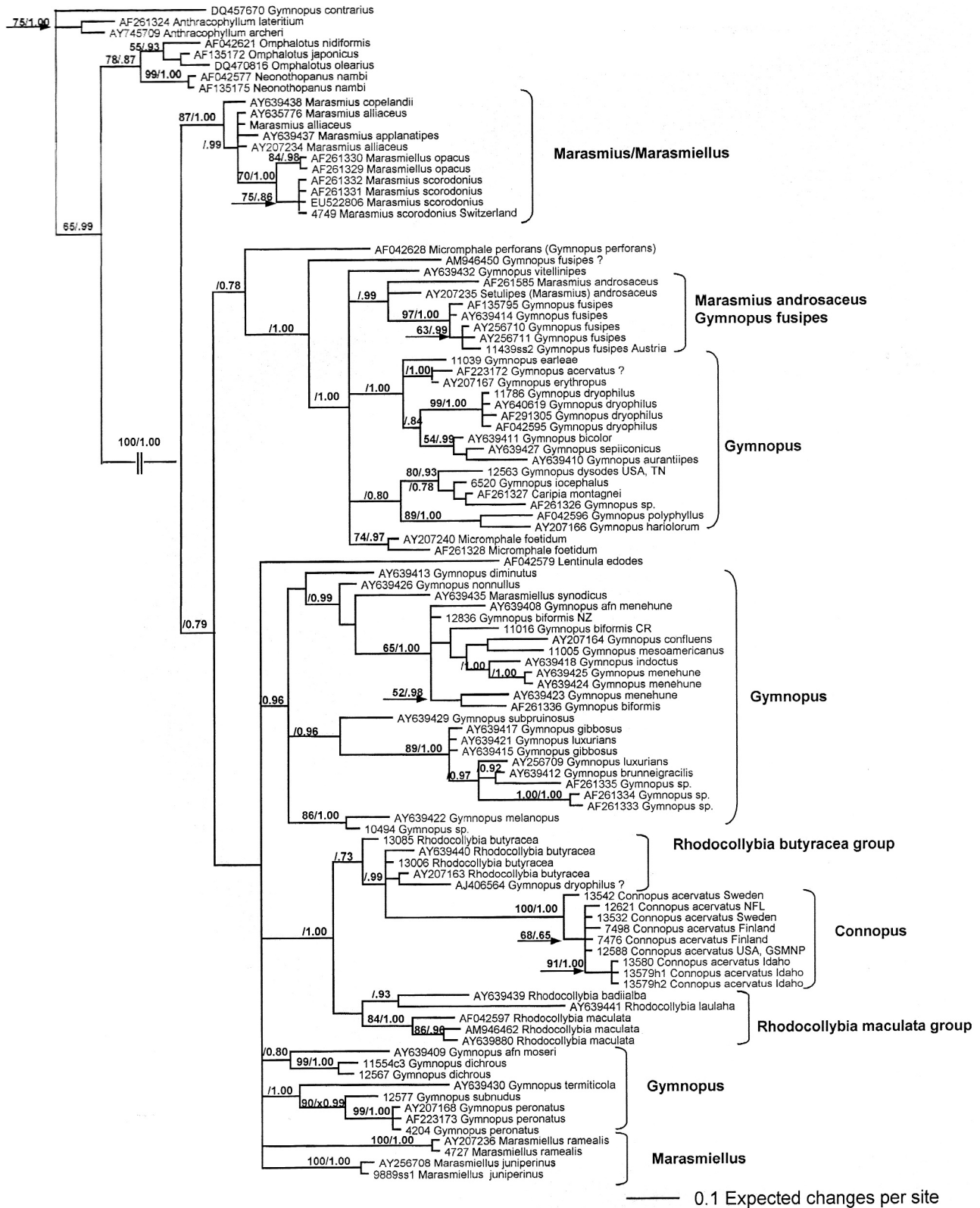


FIG. 4. Bayesian majority rule consensus tree based on ribosomal LSU sequences. Bootstrap and Bayesian posterior probabilities are to the left of each supported node. ? = GenBank name-phylogenetic placement disagreement; Ss = single spore (monokaryon) isolate.

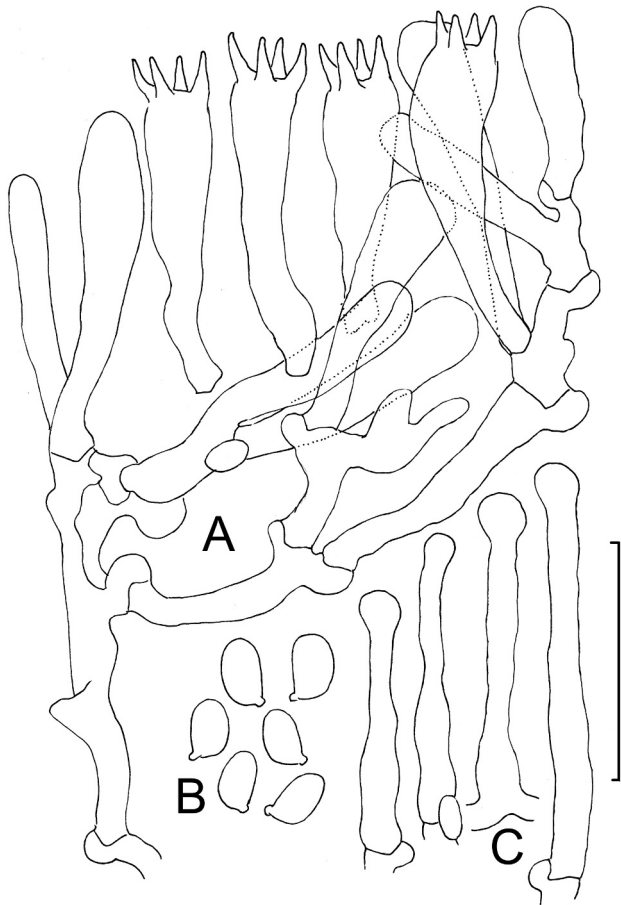


FIG. 5. *Connopus acervatus* TENN58236 from western Russia. A. Basidia. B. Basidiospores. C. Cheilocystidia. Bar = 20 μ m.

but cheilocystidia were not illustrated. A similar taxon, *C. terginoides*, while reportedly differing by "well-differentiated cheilocystidia", was not accepted as a discrete species (Antonin and Noordeloos 1997:60). In material from Russo-Scandinavia (TFB 10646, TFB10647; TFB 13542; TFB 13571; TFB 13575), maritime Canada (TFB 12621) and high-altitude southern Appalachian Mountains (TENN 17637) we see only slender-lecythiform or thermometric structures (FIG. 5C, 1.5–2 μ m diam), emergent up to 15(–20) μ m from the lamellar edge, and even these are rare and capricious (locally common in TFB 13542), but in other Euro-Scandinavian specimens cheilocystidia seem absent. No previous literature describes cheilocystidia of western North American *C. acervatus*. Our observations (TFB 5810, TFB 8225, TFB 11595, TFB 13590) show cheilocystidia locally common, otherwise rare, 3–3.5 \times 30–60 μ m, not capitulate, simply rounded at apex, arising from a conspicuous clamp, hyaline, thin-walled, emergent from hymenium up to 15 μ m.

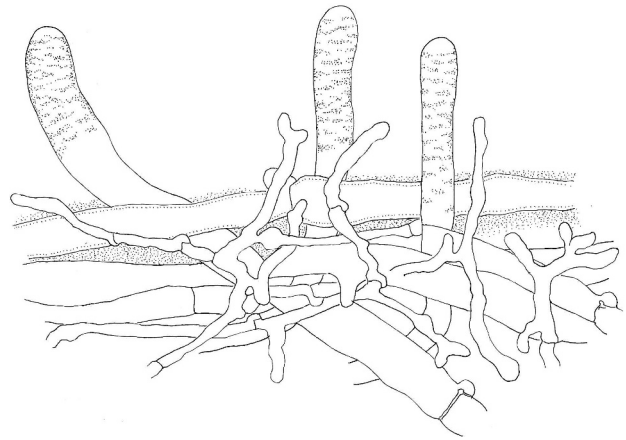


FIG. 6. *Connopus acervatus* TENN 62921. Pileipellis structure of Euro-Scandinavian specimens. Standard bar = 20 μ m. Interhyphal stippling represents shallow glutinous exudate.

Second, pileipellis of Euro-Scandinavian specimens (FIG. 6) was observed as constructed of two hyphal types: (i) relatively seldom branched, wide (6–8 μ m diam), thin-walled, occasionally clamped, hyaline hyphae with common curved, erect, subtly banded terminal cells; and (ii) intermixed, slender (1.5–3 μ m diam), frequently branched, frequently and conspicuously clamped hyphae. The former were illustrated by Antonin and Noordeloos (1997) but not the latter.

Halling (1983) recognized that *Gymnopus acervatus* exhibited a unique pileipellis organization in the genus, although his data emphasized North American collections. He described the pileipellis as "... a tangled trichodermium when young, developing into a layer of repent, cylindrical, branched hyphae, not diverticulate or coralloid, not radially arranged." A young pileipellis was illustrated. Our observation of pileipellis structure of eastern North American specimens showed the same hyphal types as found in Euro-Scandinavian basidiomata, but the wider hyphae were frequently lobed or gnarled, and if banded the banding was indistinct. Instead these hyphae often appeared spotted (PhC) with some round inclusions (FIG. 7).

Pileipellis structure of western North American pilei seems to differ somewhat (FIG. 8). The outermost layer comprises wide (6–8 μ m diam) hyphae tightly interwoven into an almost parenchymatous layer with common erect termini without inclusionary spots or superficial banding, firm-walled, conspicuously clamped. When squashed this tissue does not flatten out as other specimens do but squashes intact or shatters into small aggregations. The subpellis layer is loosely interwoven; hyphae 3.5–5 μ m diam, frequently branched, conspicuously clamped, firm-

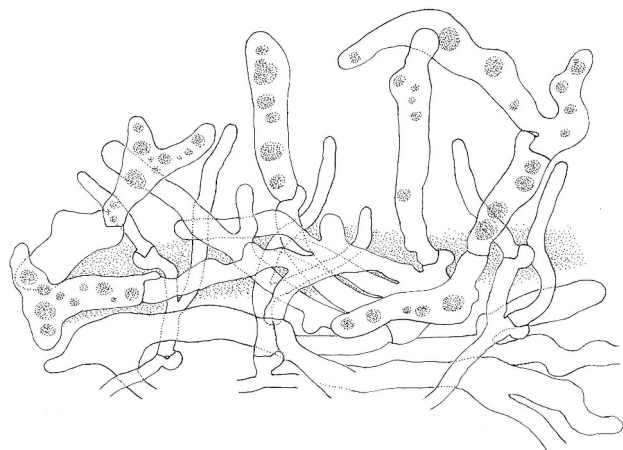


FIG. 7. *Connopus acervatus* TENN61213. Pileipellis structure of eastern North American specimens. Bar = 20 μ m. Interhyphal stippling represents shallow glutinous exudate.

walled, free (not agglutinated or adherent). In short the two hyphal types as found elsewhere are present in this material but differ somewhat in location and congestion.

Pileipellis (as “cortical layer”) structure has been detailed by Cl  men  on (2004). Using that terminology, in radial section the cortical layer of Euro-Scandinavian basidiomata generally conforms to a plagiotrichoderm but constructed of two hyphal types. Eastern North American basidiomata exhibit a cortical layer closer to a loose clavicutis. Western North American material shows a cortical layer more or less like a plagiotrichoderm which, when viewed in tangential section, loosely resembles a clavicutis (see

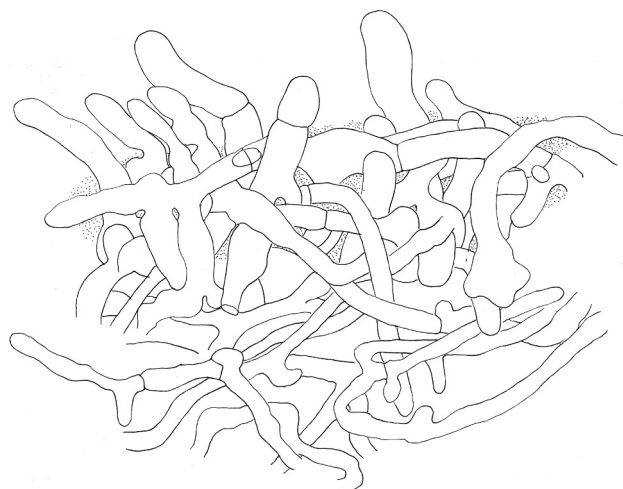


FIG. 8. *Connopus acervatus* TENN62990. Pileipellis structure of western North American specimens. Bar = 20 μ m. Interhyphal stippling represents shallow glutinous exudate.

Cl  men  on, FIG. 9.63), also known as “dryophila-structure”.

Third, basidiospores of Euro-Scandinavian specimens seem slightly larger ($5\text{--}7 \times 3.5\text{--}4.5 \mu\text{m}$), western North American specimens intermediate ($5\text{--}6.5 \times 2.5\text{--}3 \mu\text{m}$) and eastern North America smaller ($4.0\text{--}5.5 \times 2.5\text{--}3 \mu\text{m}$).

A few microscopic characters seem to have escaped description. (i) Especially in Swedish specimens (TFB 13575) caulocystidia are common over the stipe apex, appearing as a delicate, loose hyaline pruina ($30\times$) against the purple-brown stipe surface. Presence of such caulocystidia technically would dictate placement in *Gymnopus* sect. *Vestipedes* but can hardly qualify as vesture. (ii) In squash mounts of both pileipellis (peridermal scalp) and stipe surface small amounts of glutinous material are expressed from the tissues (PhC). This material often takes the form of individual hyphal lengths and must be interpreted as minimal viscid material. When notes accompanying specimens address pileus texture, “viscid” never appears but “wet”, “moist” and “slippery” are common, perhaps referencing this embedding material. Pileipellis hyphal walls are not gelatinized, so the leaching substance appears as a hyphal exudate not a matrix. (iii) All examined basidiomata, regardless of geographic origin, exhibited adventitious white pruina on the lower stipe. This tomentum varies greatly in extent, from several millimeters at stipe base to covering the stipe nearly to the pileus. Upward on the stipe the pruina consists of some superficial, interwoven hyphae with individual slender ($1.5\text{--}2 \times 50\text{--}750 \mu\text{m}$) hyphae extending outward. Downward the hyphae juxtaposed to stipe surface form a loose thatch with more densely scattered extending hyphae. Toward the stipe base the thatch often forms a soft, white tomentum up to 500 μ m thick, and surface hyphae congregate into strigose, apparently coherent synnemata up to 1000 μ m long. The thatch itself often engulfs surrounding moss leaves and other detritus. Individual stipes disappear into the engulfing tomentum at base but remain discrete. Whether a single primordium produces multiple basidiomata or whether a cluster of individual primordia is involved remains unresolved. Thatch hyphae are loosely interwoven, firm-walled, conspicuously and frequently clamped, hyaline and consistent in diameter (3.5–4 μ m).

Notes accompanying specimens often identify conifer logs, stumps or forest, but less commonly specify particular trees. Among them are *Larix* (United Kingdom), *Picea* (Sweden, GSMNP), *Pseudotsuga* and *Thuja* (Washington state), *Picea abies*, *Betula*, occasional *Populus*, with *Pinus sylvestris* overstory (Russia).

DISCUSSION

In a paper on the Omphalotaceae with ribosomal ITS sequences *Gymnopus acervatus* appeared on a long branch between *Rhodocollybia* and *Gymnopus* (Mata et al. 2006, FIG. 2). That phylogeny excluded several unalignable regions from the Omphalotaceae dataset to achieve a phylogeny for Omphalotaceae overall. *Gymnopus acervatus* was excluded from analyses because it could not be reasonably aligned with full length *Gymnopus* or *Rhodocollybia* ITS sequences. Placement of *G. acervatus* within *Gymnopus* is not supported by the current study. At the ITS level *G. acervatus* sequences consistently form a clade well separated from other collybioid genera and are not easily aligned with other collybioid genera in variable regions within ITS1 and ITS2 (FIG. 1). In LSU-based phylogenetic reconstructions the closest taxonomic group to *G. acervatus* was affiliated with *Rhodocollybia*, either between the two major *Rhodocollybia* clades rendering *Rhodocollybia* paraphyletic (Bayesian analysis) or derived from *Rhodocollybia*, but LSU trees constrained so that *G. acervatus* is monophyletic (as in the ITS-based tree) are not significantly different from the unconstrained tree.

Morphologically *G. acervatus* is not a *Rhodocollybia*. *Rhodocollybia* is defined by a pinkish cream spore print. *Gymnopus acervatus* has white spores more consistent with other genera of Omphalotaceae (i.e. *Mycena*, *Marasmius*, *Clitocybe*, *Tricholoma*, etc.) and spores are neither dextrinoid, cyanophilous nor thick-walled. Because "*Gymnopus acervatus*" is neither *Rhodocollybia* nor *Gymnopus* based on ITS and LSU sequences a new genus to accommodate this species is proposed here.

Percent ITS sequence difference between collections in *Connopus* Clade 1 (western North American collections) and Clade 2 (eastern North American and European collections) is at the margin of percent sequence divergence suggestive of different molecular species (2.99–3.80%; see Hughes et al. 2009). While sequence divergence within species vary with species concepts and taxonomic groups, the observed sequence difference, the well supported western North American clade and lack of any observed hybridization suggests that the western North American collections might be a distinct species. While Clade 2 ITS Scandinavian and eastern North America sequences are similar, morphological differences in cheilocystidia, pileipellis structure and basidiospore dimensions were observed. Further study is needed to determine whether collections in clades 1 and 2 are reproductively isolated and/or whether European and eastern North American collections are conspecific.

With several ITS sequences representing *Connopus acervatus* a question could be raised about a "representative sequence" or a candidate for barcode sequence. We consider that the notion of bar-coding should not bypass the International Code of Botanical Nomenclature and its conventions. Thus it follows that because the epithet originated from Fries (1821:121), an exemplar sequence must be based on a southern Swedish collection vouchered by an accepted herbarium specimen. Even within the sequences included in our phylogeny, small but distinct sequence differences can be seen. We suggest that sequence GU318387 (ITS) and FJ750255 (LSU) (TFB 13532 = TENN 62879) serve as exemplar sequences for the purposes of bar-coding. This collection is heterozygous and two sequences were deposited. Within Scandinavia several different haplotypes have been recovered.

GenBank is noted for inaccurate phylogenetic designations and the error rate might be as high as 20% (Bridge et al. 2003, Nilsson et al. 2006). Correction of inaccuracies ultimately must be based on specific published phylogenies that cover the groups in question. As part of this study we identified sequences in GenBank with assigned names that are not congruent with their placement in the phylogeny as follows: large subunit sequence (FIG. 3) AF223172 (Moncalvo et al. 2002) = CBS 174.48 *Gymnopus acervatus*. This sequence is not congruent with true *G. acervatus* but is congruent with *Gymnopus erythropus*, AJ406564 *Gymnopus dryophilus* (E. Langer, no paper cited). This sequence falls within *Rhodocollybia*.

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