To the Graduate Council:

I am submitting a dissertation written by Juan Luis Mata entitled "Taxonomy and systematics of *Lentinula*, *Gymnopus* and *Rhodocollybia* (Agaricales, Fungi), with emphasis on oak forests of southern Costa Rica." I have examined the final paper copy of this thesis for form and content and recommend that it be accepted in partial fulfillment of the requirements for the degree of Doctor of Philosophy, with a major in botany.

Ronald H. Petersen, Major Professor

We have read this dissertation
And recommend its acceptance:

Karen W. Hughes

Edward E. Schilling

Sally P. Horn

Accepted for the Council:

Vice Provost and Dean of
Graduate Studies

TAXONOMY AND SYSTEMATICS OF *LENTINULA, GYMNOPUS*, AND *RHODOCOLLYBIA* (AGARICALES, FUNGI), WITH EMPHASIS ON OAK FORESTS OF SOUTHERN COSTA RICA

A Dissertation
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Juan Luis Mata
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DEDICATION

This dissertation is dedicated to my wife Ingrid and to our children. Also, I want to dedicate this dissertation to my parents Lic. Grace Eugenia Greenwood Quirós and Dr. Leonardo de Jesús Mata Jiménez in recognition to their effort and merit in academy, research, and professional service in the areas of microbiology and public health in Guatemala and in Costa Rica.

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ABSTRACT

This dissertation comprises a taxonomic and systematic study of *Lentinula*, *Gymnopus* and *Rhodocollybia* as found in the Talamanca Mountains of southern Costa Rica, based primarily on morphological descriptions and supported by mating studies and ITS1-5.8S-ITS2 nuclear ribosomal DNA sequence analyses. Each morphological description includes analysis of extant or authentic material, examination of the type specimen when possible, and a nomenclatural revision.

The genus *Lentinula* in the Americas comprises three distinct morphological, biological and phylogenetic species. *Lentinula boryana* and *L. raphanica* form two intersterility groups distinguished by micromorphology and ITS nrDNA. Assignment of correct names to each species was based on examination of all type specimens of synonymous names for the *L. boryana* species complex. Both *L. boryana* and *L. raphanica* have a widespread tropical-subtropical distribution but only *L. raphanica* seems to fruit in the Gulf Coast region of the United States. *Lentinula aciculospora* is the third species known to fruit in the Talamanca Mountains of Costa Rica. Based on ITS nrDNA sequence analysis, all *Lentinula* species in the Americas form a phylogenetic clade separated from those species in the *L. edodes* biological species group from Asia and Australia.

The genus *Rhodocollybia* in the Talamanca Mountains in this study comprises eight morphological species, six of which share dextrinoid spores as a common taxonomic characteristic. *Rhodocollybia dotae*, *R. lignitilis*, and *R. pandipes* are described as new species and *R. popayanica* and *R. prolixa* var. *distorta* represent range extensions from South America and Europe respectively. Mating studies in *R. pandipes* demonstrated that it forms a biological species across the Talamanca Mountains. *Rhodocollybia amica* and *R. tablensis*, both with no observed dextrinoid basidiospores, and five other Costa Rican *Rhodocollybia* species were phylogenetically related to each

other, and together with temperate species form a distinct clade apart from *Gymnopus* and *Lentinula* in analyses using the ITS1-5.8S-ITS2 nrDNA region.

A total of 18 morphological species in *Gymnopus* were found fruiting in the Talamanca Mountains. *Gymnopus* sect. *Levipedes* includes four species, with *G. nubicola* and *G. spongiosus* previously not reported for Costa Rica. New proposed species in *Gymnopus* sect. *Vestipedes* include *G. alnicola*, and *G. cylindricus*, and *G. biformis* var. *lobatus* and *G. biformis* var. *parvulus* are reported as two new varieties. *Gymnopus collybioides*, *G. confluens*, *G. dichrous*, *G. fibrosipes*, *G. impudicus*, *G. polyphyllus*, and *G. pseudo-omphalodes* represent new reports for the Talamanca Mountains. Mating studies in several species including *G. lodgeae, G. collybioides,* and *G. impudicus* revealed a tetrapolar mating system except in the case of *G. omphalodes* and *G. pseudolodgeae* in which a bipolar mating system was observed. Intercollection compatibility. i.e. a biological species, was observed in *G. dryophilus*, *G. lodgeae*, *G. omphalodes* and *G. impudicus*.

Based on sequence analyses of the ITS1-5.8S-ITS2 nrDNA region *Rhodocollybia* appears to be phylogenetically more closely related to *Gymnopus* than to *Lentinula*. All species in *Rhodocollybia* seem to form a monophyletic clade. Species previously placed in *Gymnopus* sect. *Vestipedes* seem not to be monophyletic with members of sect. *Levipedes* nested therein. Sections *Impudicae* and *Peronati* are proposed as new combinations under *Gymnopus* in order to accommodate those species phylogenetically segregated from sect. *Vestipedes*. Sequences from Costa Rican specimens are phylogenetically related to those representing temperate morphological species in *Rhodocollybia* and *Gymnopus*, including *R. maculata* and *G. fusipes*, type species for these genera.

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PART I

General Introduction

Agaricoid mushrooms are placed in the Kingdom Fungi, Phylum Basidiomycota, Class Hymenomycetes, Order Agaricales (Alexopoulos et al, 1996). Around 15 families are currently recognized in the Agaricales with Tricholomataceae making up one of the most diverse and numerous in number of genera and species (Singer, 1986). Basidiomata of tricholomatoid agarics are typically fleshy, consisting of a stipe and a pileus with lamellae covered by a hymenium of basidia producing white spores (Alexopoulos et al, 1996; Hawksworth et al, 1983).

Ever since Linnaeus (1753) produced a comprehensive treatise that systematically classified plants and relatives based on binomial names, biologists have used this practical application to pursue the goal of doing taxonomy with other groups of living organisms. In the area of mycology, Persoon (1801) and Fries (1821) were among the first to apply the Linnaean classification system to write all-inclusive taxonomic treatments of fungi. Many of the generic and species names used today were mentioned in their works. Because the Friesian classification system relies so much on field character identification, the taxonomy of certain groups of mushrooms sometimes has been confusing. Such is the case of *Lentinula* Earle (1909), *Gymnopus* Roussel (1806) and *Rhodocollybia* Singer (1939), the last two associated with the genus name *Collybia* Staude (1857).

Monographic and floristic studies based on morphological traits have been produced in *Lentinula* (Pegler, 1975, 1983b) and *Gymnopus* and *Rhodocollybia* (Antonín and Noordeloos, 1997; Halling, 1983; Lennox, 1979). In present times, however, studies of mushrooms incorporate genetic and molecular techniques to help answer questions in taxonomy and systematics that are otherwise difficult using only traditional morphological approaches. Systematic and biogeographic studies based on DNA sequence analyses have contributed to better understanding of the genus *Lentinula* (Hibbett and Vilgalys, 1993; Hibbett et al, 1995; Hibbett, 2001; Mata et al 2001) and mating studies have been used to elucidate species concepts in the *G. dryophilus* complex (Vilgalys and Miller, 1983; 1987) and in *G. subnudus* (Murphy, 1997). Recently

Lentinula, *Gymnopus* and *Rhodocollybia* have been placed in the same phylogenetic clade based on large subunit nuclear ribosomal DNA sequence analysis (Moncalvo et al, 2000; 2002).

Basidiomata representing species of *Gymnopus* and *Rhodocollybia* have been collected and studied in more detail in temperate regions (Halling, 1983; Lennox, 1979; Antonín and Noordeloos, 1997) than in tropical countries (Dennis, 1970; Guzmán et al, 1992; Pegler, 1983a). The Talamanca Mountains in southern Costa Rica house large tracts of oak forests that have a distinct agaric mycota associated to them (Gómez, 1986; Halling, 2001; Kapelle, 1996; Mueller and Halling, 1995). The need for more taxonomic studies in *Lentinula*, *Gymnopus* and *Rhodocollybia*, whose basidiomata fruit in oak-forest habitat in the Neotropics, in combination with a country providing adequate conditions for mushroom collection allowed this project to become a reality.

This dissertation is composed of four parts. The first three parts are represented by articles on *Lentinula* already published in peer-reviewed mycological journals. The first one describes *L. aciculospora*, a new species in Costa Rica, based on morphological and genetic studies. The second part deals with the study of all type specimens of names in synonymy with *L. boryana* in order to elucidate the application of correct names for two genetically intersterile groups of *Lentinula* fruiting in the Americas. The third part provides a detailed description of these intersterility groups, *L. boryana* and *L. raphanica*, and a phylogenetic analysis of the whole genus, including all morphological forms of *L. edodes*. The fourth part covers the genera *Rhodocollybia* and *Gymnopus* from the Talamanca Mountains of southern Costa Rica. In this final part 26 morphological species are described, some including genetic studies, and at the end a phylogenetic analysis of these species in relation to others collected in temperate regions is provided.

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PART II

A new species of *Lentinula* (Agaricales) from Central America

This chapter is a thesis-formatted version of a paper with the same title published in the journal *Mycoscience* in 2000 by Juan L. Mata and Ronald H. Petersen.

Juan L. Mata and Ronald H. Petersen. 2000. A new species of *Lentinula* (Agaricales) from Central America. Mycoscience 41: 351-355.

Abstract

Lentinula aciculospora sp. nov. is described from a single Province in Costa Rica. Stature and habit of basidiomata of this species are similar to some other *Lentinula* taxa (i.e. *L. edodes*), but the elongate-cylindrical shape of basidiospores is a distinctive character of *L. aciculospora*. The mating system of *L. aciculospora* is tetrapolar as also found in other species of the genus, but this species is genetically isolated from *L. boryana*, and all morphological variants of *L. edodes*.

Key words: *Lentinula*, Agaricales, biological species, mating systems, Costa Rica, *Quercus*.

Introduction

Lentinula Earle has become the object of recent research, especially with the application of molecular studies (Nicholson et al., 1997; Hibbett, 1992; Hibbett et al., 1995, 1998) to sort out Old and New World taxa. Among Old World populations, including the cultivated *L. edodes* (Berk.) Pegler, several distinguishable morphological variants occur, which while sexually intercompatible (Mori et al., 1974; Shimomura et al., 1992) produce identifiable RFLP signatures (Nicholson et al., 1997) and definite phylogenetic ITS-based lineages (Hibbett et al., 1998). These Old World taxa have received much attention because of their economic importance (Chiu et al., 1999; Hibbett and Donoghue, 1996; Royse and May, 1987) along with morphological and molecular revisions to assess their taxonomic circumscription (Molina et al., 1992; Pegler, 1975, 1983).

Lentinula boryana (Berk. & Mont.) Pegler and *L. guarapiensis* (Speg.) Pegler are the two morphospecies reported in the New World (Pegler, 1983), the latter known only from its type collection (Pegler, 1983). Until recently, only *L. boryana* was thought to fruit in subtropical and tropical America (Guzmán et al., 1997; Pegler, 1983), but preliminary genetic data, supported by morphological (Petersen et al., 1998) and phylogenetic analysis (Hibbett et al., 1998; Hibbett, 1999), show a third species also to be present. In recent field trips to Central America, basidiomata from the suspected third *Lentinula* have been collected and will be reported in a subsequent paper. The purpose of this paper is to describe a fourth species of *Lentinula* in the New World, this one known only from the Talamanca Mountains in Costa Rica.

Materials and methods

Morphological observations. Sections of dried specimens were re-hydrated in 95% ethanol and water, then placed in 3% KOH, observed in phase contrast, or with Congo Red and phloxine with bright field microscopy. Melzer's reagent was used to conduct amyloidy tests.

Terminology. Colors in quotation marks are from Ridgway (1912) and alphanumeric codes in parentheses from Kornerup and Wanscher (1978). Basidiospore measurements and statistics are as follows: Spore length x spore width is given for all measured basidiospores in terms of their range of variation; x = range of the minimum and maximum arithmetic means of spore length by spore width for all spores measured; Q = spore length divided by spore width in any spore, indicated as a range of variation in n spores measured; Qx = arithmetic mean of Q-values; n = total number of spores measured; numbers in parentheses = number of collections and specimens studied; **M** = specimens used for morphological examination; and **P** = collections/cultures used for pairings. As of this moment, the name *Armillaria raphanica* Murrill is applied temporarily to those collections made in the Gulf Coast region of North America and Puerto Rico, belonging to a distinct mating complex (i.e. biological species) from *L. boryana*, and also well-defined by micromorphological characters and ITS sequences (Hibbett, 1999; Petersen et al., 1998). Ongoing studies of appropriate type specimens will

reveal the most priorable name for this taxon and an appropriate combination will be adopted.

Cultures. Culture techniques followed those described by Gordon and Petersen (1991). Spore deposits were obtained from fresh basidiomata placed on the inside of a tilted sterile Petri dish containing malt extract agar (MEA, 1.5% Difco malt extract, 2% Difco bacto-agar, 1L distilled water). Germinating spores were harvested and individually transferred to fresh MEA plates. Full grown mycelium was inspected for absence of clamp connections to ascertain monokaryon state. Small agar blocks with mycelium were placed in small vials (4ml cap) containing sterile water for long-term storage at room temperature.

Self-cross. Eleven single-basidiospore isolates (SBIs) from collection 56421 (TENN) (*L. aciculospora*) were paired in all combinations. Subsequently, rows and columns of the resulting grid were adjusted to reveal patterns of compatibility.

Intercollection pairings. In all cases, four SBIs from collection 56421 (TENN) (*L. aciculospora*) were paired with four SBIs of collections from *A. raphanica*, *L. boryana* and all the morphological variants of *L. edodes*. In all cases, pairings totaled four.

Material studied. *Lentinula aciculospora*: COSTA RICA, Prov. San José, Co. Pérez Zeledón, C.A.T.I.E. Experimental Forest of Villa Mills, 9° 33' 03" N, 83° 40' 56" W; 2880 m, 22.VI.95, coll. RHP, no. 7901 (TENN 53813) **M**; same location, 10.VI.97, coll. R.E. Halling, no. REH 7717 **M** ; same location, 30.VI.98, coll. RHP, no. 9447 (TENN 56421, HOLOTYPE) **M**, **P**; same location, 9.VII.98, coll. JL Mata, no. 10026 (TENN 56738) **M**; same location, 20.VI.99, coll. J.L. Mata, no. 10410 (TENN 58105) **M**; Prov. San José, Co. Pérez Zeledón, Cuerricí Biol. Station, 9° 33'17" N, 83° 40' 04" W, 2560 m, 21.VI.99, coll. J.L. Mata, no. 10418 (TENN 58112) **M**. *Lentinula boryana*: COSTA RICA, Prov. San José, vic. Cerro Chonta, 9° 41' 35" N, 83° 56' 19" W, 16.VI.95, coll. RHP, no. 7829 (TENN 53819) **P**; MEXICO, Est. México, vic. Valle de Bravo,

6.VII.96, coll. M. Rodríguez, no. 8436 (TENN 55265) **M, P**. *Lentinula_edodes*: JAPAN, Pref. Tochigi, vic. Lake Chuzenjiko, 27.IX.89, coll. I. Izawa, no. 2306 (TENN 48220) (*edodes* morph) **P**; NEPAL, Ilma, V.95, coll. H.R. Bhandary, no. HRB 7683 (spore print only) (*edodes* morph) **P**; AUSTRALIA, New South Wales, Watagan Mountains, Boarding House Dam, 21.V.91, coll. RHP, no. 3577 (TENN 50062) (*lateritia* morph) **P**; NEW ZEALAND, North Island, vic. Lower Hutt, Wainuiomata, Orongorongo Trailhead, 25.V.94, coll. A.S. Methven, no. 7563 (TENN 53718) (*novaezelandiae* morph) **P**. *Armillaria_raphanica*: UNITED STATES, Florida, Gadsdon Co., Torreya State Park, 30° 34' 35" N, 84° 56' 57" W, 30.V.97, coll. K.W. Hughes, no. 9156 (TENN 54887) **M, P**; Commonw. Puerto Rico, Luquillo Nat. Forest, El Verde Biol. Station, 18° 19' 19" N, 65° 49' 12" W, 9.VI.98, coll. E. Lickey, no. 9564 (TENN 56477) **P**.

Results

Description of the new species

Lentinula aciculospora Mata & R.H.Petersen, sp. nov. Figs. 1-2

Pileus 20-150 mm latitudo, primo convexus, deinde plano-convexus, demum depressus; superficies laevis, primo vinoso-spadicea, dein ochraceo-salmonea, vel ochraceo-aurantiaca, squamulis adpressis, pyramidalis, late dispositis ad marginem pilei, caryophyllaceo-bubalinis; margo semper decurvatus, caryophyllaceo-cinnamomeus; velum partiale plerumque appendiculatum, cortinoideum, laete caryophyllaceo-cinnamomeum. Contextus latus, albus, vel laete caryophyllaceo-bubalinus. Lamellae primo adnatae, adnexescentes, approximatae, exalbidae vel vinoso-bubalinae, aliquando vinaceo-marginatae. Stipes 30-70 x 10-20 mm, cylindricus, basi obclavata vel clavata, squamulis numerosis, subannularibus, laceratis, rubiginosis vel luteo-brunneis vestitus; consistentia solida, tenax. Odor et sapor vix sensibilis.

Basidiosporae 5.6-8.8 x 1.6-2.8 µm, cylindricae vel subreniformes, hyalinae, inamyloideae, laeves. Cheilocystidia vermiformia, anguste clavata vel anguste

sphaeropedunculata, aliquando apice lobata, hyalina, laevia. Pleurocystidia et caulocystidia absentia. Trama lamellae subregularae. Pileipellis repens. Trama pilei 500-100 µm crassa, intertexta, lacunosa. Hyphae fibulatae, tenui- vel crassitunicatae, hyalinae, inamyloideae.

Habitatio: gregarius, in ligno Quercus spp. Holotypus: Costa Rica, Prov. San José, no. 56421 (TENN).

Morphology Pileus 20-150 mm broad, convex at first, later broadly convex to plano-convex, finally depressed; surface dry, smooth or with scattered, appressed, ± pyramidal squamules over disc, becoming more widely spaced and suberect to erect towards pileus margin; disc surface 'claret brown' (9E8) at first, 'ochraceous salmon' (6A6), 'amber brown' (6D8), to 'ochraceous orange' (6B7), outward 'tawny' (6C6, 6C7) to 'orange cinnamon' (7B6); margin always inrolled, 'pinkish cinnamon' (7B5), 'Hay's russet' (8D7), 'walnut brown' (7D6), 'pinkish buff' (6A3). Partial veil mostly appendiculate, heavily cortinoid to pholiotoid, 'light pinkish cinnamon' (7A2). Pileus flesh 10 - 15 mm thick at center of disc, solid, white to 'pale pinkish buff' (6A2), with subtle line between pileus and stipe flesh thick, white to abruptly 'warm sepia' (7F6) at base. Odor and taste negligible. Lamellae 2-6 mm deep, adnate at first, becoming adnexed, not seceding, close to crowded, hardly ventricose, off-white at first, 'pinkish buff' (6A3) to 'vinaceous buff' (9B2), bruising slowly and sometimes appearing marginate to 'pinkish buff' (6A3, 5A3), 'avellaneous' (7B3) or 'vinaceous russet' (8D4). Stipe 30-70 x 10-20 mm, cylindrical, expanded to obclavate at base; surface dry, beset with numerous subannulate, lacerate scales; surface 'pinkish cinnamon' (7A2) to 'tawny' (6C6); scales, 'Hay's russet' (8D7) to 'walnut brown' (7D6); consistency tough (Fig. 1).

Epicutis ± 20-50 µm thick; hyphae 3-6 µm broad, yellow tan in mass, hyaline singly, repent (except in erumpent warty scales), compacted, interwoven, undifferentiated; thin- to thick-walled (wall up to 0.7 µm thick). Hyphae in scales erumpent, tightly packed, ending in weathered or senescent hyphal fragments. Subpellis 100-250 µm thick; hyphae 3-16 µm broad, hyaline, thick-walled (wall up to 1.5 µm

thick), tightly interwoven (except in frequent lacunae). Pileus trama 500-1000 μm thick, loosely interwoven, but generally radially oriented, lacunose; hyphae 6-12 μm broad, thick-walled (wall 1.5-2.5 μm thick), pale straw in mass, hyaline singly, inamyloid. Lamellar trama subregular to tightly interwoven in mediostratum; hyphae 2-4 μm wide, thin- to thick-walled (wall up to 0.5 μm thick), pale straw in mass, hyaline singly; lateral strata irregularly parallel, less tightly packed, abruptly differentiated into subhymenium; hyphae hyaline, more or less thin-walled. Subhymenium rudimentary, pseudoparenchymatous. Hymenium composed solely of basidia and basidioles. Basidia (Fig. 2a) 18-25 x 4-7 μm (n = 25), narrowly clavate to subcylindrical, with clamp connections, four-sterigmate; sterigmata slender, small. Cheilocystidia (Fig. 2b) 16-34 x 4-10 μm (n = 20), collybioid, vermiform to narrowly clavate or narrowly sphaeropedunculate, somewhat inflated at least apically, often gnarled or terminally bluntly lobed; wall thin, smooth; in some cases, lamellar margins sterile. Pleurocystidia absent. Stipitipellis composed of loosely entangled, short or branched hyphae 4-10 μm broad, forming floccons; medullary layer strictly parallel; medullary hyphae 3-12 μm broad, tightly compacted, thin- to thick-walled (wall up to 1.5 μm thick), straw to yellowish brown in mass, hyaline singly, inamyloid. Caulocystidia absent.

Basidiospores (Fig. 2c) white in mass, 5.6-8.8 x 1.6-2.8 μm [x = 6.4-7.7 x 2.2-2.5 μm; Q = 2.29-4.00, Qx = 2.97; n = 104(4/5)], cylindrical to subreniform, hyaline, inamyloid, thin-walled, smooth, easily collapsed; hilar appendix small, obscure; contents usually bivacuolate, sometimes homogeneous.

Habitat gregarious, on fallen oak wood. Vegetation: *Quercus costaricensis* Liebmann and *Quercus* spp.

Etymology: *acicularis* (Lat., adj.), narrow, referring to the shape of the basidiospores.

Holotype: Costa Rica, Prov. San José, no. 56421 (TENN); with cultures under no. 9447.

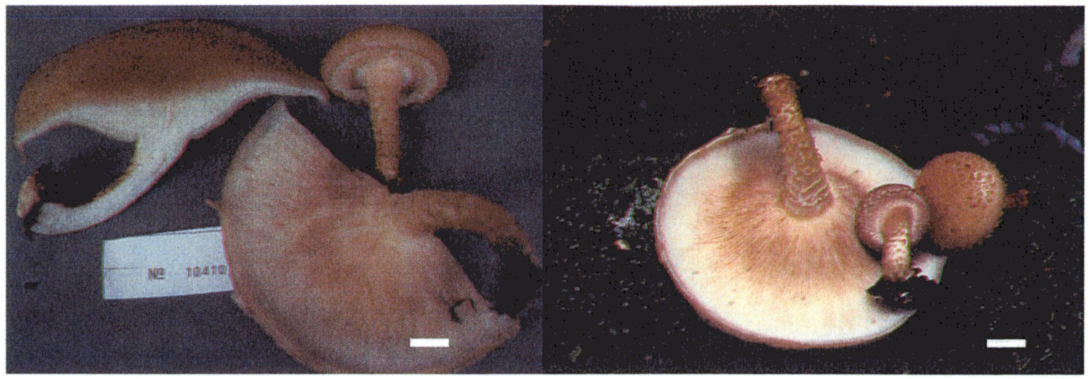


Fig.1. Basidiomata of *Lentinula aciculospora*. 58105 (TENN). Scale bar aprox. 2 cm.

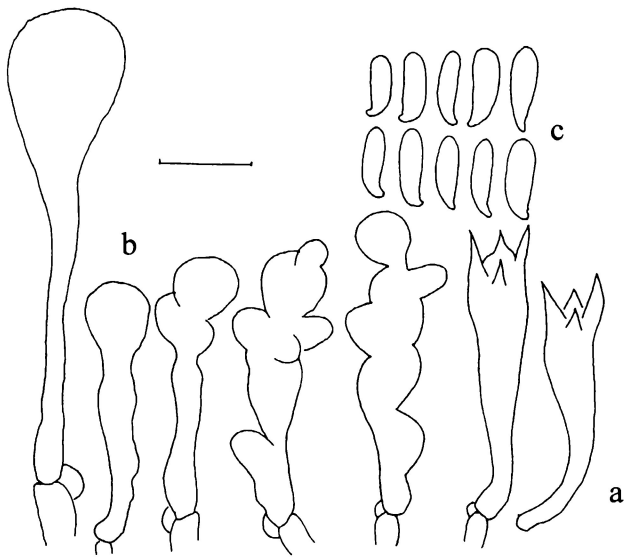


Fig. 2. Microscopic structures of *Lentinula aciculospora*. 56421 (TENN, HT). a. Basidia. b. Cheilocystidia. b. Basidiospores. Scale bar equals 10 μm.

Self-cross. When 11 SBIs were paired in all combinations, a tetrapolar mating system was revealed (Fig. 3). Isolates 4*, 8 = A1B1; 1*, 2, 5, 6, 10 = A2B2; 9* = A2B1; 3*, 7, 11 = A1B2. Subordinate mating types were assigned based on 9 x 1/2/5/6/10 = "flat" = common-A. Within-mating type flat contact zone morphologies (i.e. 1/2/5/6/10 x 1/2/5/6/10) rendered "flat" observations of 9 x 1/2/5/6/10 less credible, however. "Flat" contact zone morphology was indistinct, with a clear but ill-defined crevasse. No barrage morphologies were observed.

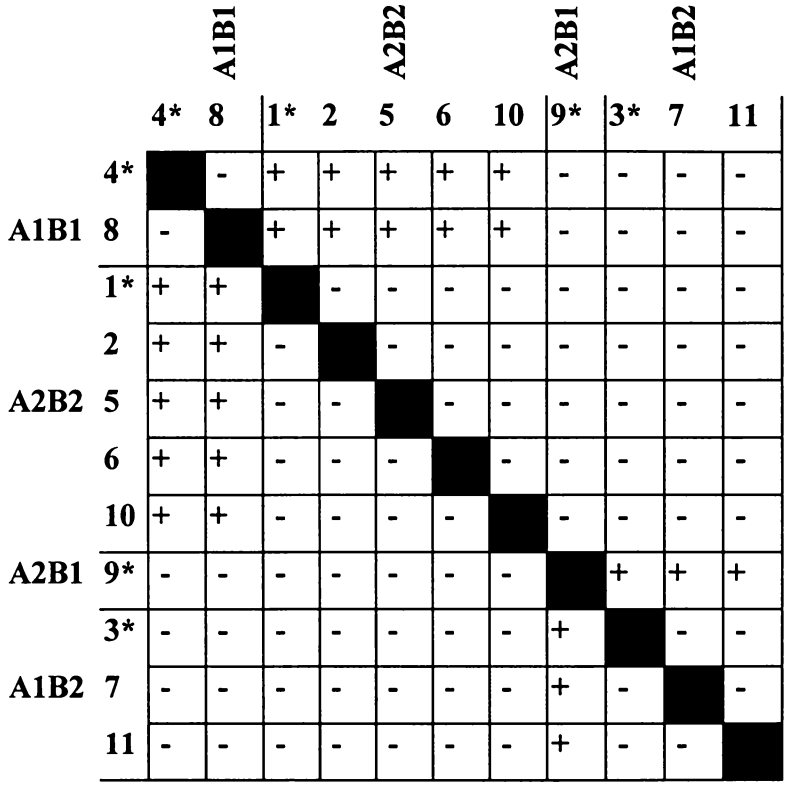
Culture characters. Growth rate averaged one cm radius in six weeks. Colonies on MEA were white, radially combed, sparsely silky, becoming more aerial toward the colony margin. Aside from occasional "picket fence" hyphae and some mild lethal reactions, little hyphal differentiation was noted.

Intercollection pairings. In pairing experiments with all collections listed above, all examined pairings of *L. aciculospora* with *A. raphanica*, *L. boryana* and all morphological variants of *L. edodes* were incompatible. In some instances, "non-self" reactions were noted.

Discussion

Habit and stature of basidiomata of *L. aciculospora* resemble those of other species in the genus, and are similar in size to the morphological variants of *L. edodes* (Pegler, 1983), but notably larger than those of *L. boryana* or *Armillaria raphanica*. Basidiomata of *L. aciculospora* are darker in color than those reported for the morphospecies *L. lateritia* (Berk.) Pegler and nearer to those of the *L. edodes* morphospecies (Pegler, 1983). *Lentinula boryana* and *A. raphanica* present pinkish buff to yellowish white colors in their pilei, separating them from *L. aciculospora* in the New World.

Lentinula aciculospora shares the following microscopic characters with other taxa in the genus: 1) undifferentiated pileus epicutis; 2) loosely interwoven pileus trama consisting of thin- to thick-walled hyphae; 3) clamp connections in all tissues; and 4)



			A1B1			A2B2				A2B1		A1B2	
		4*	8	1*	2	5	6	10	9*	3*	7	11
	4*		-	+	+	+	+	+	-	-	-	-
A1B1	8	-		+	+	+	+	+	-	-	-	-
	1*	+	+		-	-	-	-	-	-	-	-
	2	+	+	-		-	-	-	-	-	-	-
A2B2	5	+	+	-	-		-	-	-	-	-	-
	6	+	+	-	-	-		-	-	-	-	-
	10	+	+	-	-	-	-		-	-	-	-
A2B1	9*	-	-	-	-	-	-	-		+	+	+
	3*	-	-	-	-	-	-	-	+		-	-
A1B2	7	-	-	-	-	-	-	-	+	-		-
	11	-	-	-	-	-	-	-	+	-	-	

Fig. 3. Self-cross of *Lentinula aciculospora* 56421 (TENN, HT) showing a tetrapolar mating system. The sign – means no clamp connections observed, flat contact reaction; + means clamp connections present. SBIs with an * have been selected as tester strains.

absence of pleurocystidia. However, the distinctive cylindrical shape of the spores in *L. aciculospora* separates it from all others. Q spore ratio of 2.97 for *L. aciculospora* is larger than those of examined collections of *A. raphanica* and *L. boryana* 2.15 and 1.91 respectively (Mata et al., unpub. data), and larger than those reported by Pegler (1983) for taxa of the Old World. Cheilocystidia in *L. boryana*, 19-40 x 5-14 µm, are broadly clavate to sphaeropedunculate and larger than those in *L. aciculospora* (Mata et al., unpub. data). Cheilocystidia of *L. aciculospora* are similar in shape and size to those of *A. raphanica*, 17-28 x 3-10 µm, but *L. aciculospora* lacks caulocystidia (Mata et al., unpub. data).

All collections thus far have been gathered within a high altitude forest region in the Talamanca Mountain Range, all on logs of probably Quercus spp., and all at the same season of the year. *Lentinula boryana* has been collected in several localities of the Talamanca Mountains whereas *A. raphanica* has been collected there once (Mata et al., unpub. data). In all cases, depauperate, rain-soaked basidiomata of *L. aciculospora* were also found, and it was concluded that basidiomata easily became waterlogged and rapidly degenerated, a condition not readily observable with other *Lentinula* there.

Basidiospores of other *Lentinula* taxa germinate rapidly (within 24 hrs) on MEA, and colonies grow rapidly. In *L. aciculospora*, germination occurs in 7-10 days, and colonies are slow-growing compared to congeneric taxa. All taxa within *Lentinula* appear to exhibit a tetrapolar mating system, so it is not surprising to find this in *L. aciculospora*. Nevertheless, *L. aciculospora* appears to be genetically isolated from all other *Lentinula* taxa used in this project. The only taxon reported in the genus but not used in this project is *L. guarapiensis*, a species only known from its type specimen (Nicholson et al., 1997; Pegler, 1983).

Acknowledgements

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PART III

Type specimen studies in New World *Lentinula*

This chapter is a thesis-formatted version of a paper with the same title published in the journal *Mycotaxon* in 2001 by Juan L. Mata and Ronald H. Petersen.

Juan L. Mata and Ronald H. Petersen. 2001 Type specimen studies in New World *Lentinula*. 2001. Mycotaxon 79: 217-229.

Abstract

Discovery of two biological species within putative *Lentinula boryana* led to examination of type specimens of names previously listed in synonymy under *L. boryana*. Shape of cystidia and spore dimensions were diagnostic in assigning each type collection to a morphological species. *Armillaria raphanica* is the most priorable basionym representing the "non-*boryana*" taxon, and the epithet is recombined in *Lentinula*.

Key words: nomenclature, specimen studies

Introduction

Lentinula boryana (Berk. & Mont.) Pegler and *L. guarapiensis* (Speg.) Pegler have been considered to be the only New World species within *Lentinula*, the latter species known only from its type collection (Pegler, 1983a). Until now, *L. boryana* has been reported from the tropics of Mexico, Central America, Caribbean landmasses and South America to subtropical North America (Hibbett et al, 1998; Nicholson et al, 1997; Mata et al., 2000; Murrill, 1916; Pegler, 1983a; Petersen et al, 1998).

Mating experiments involving several specimens of putative *L. "boryana"* from Mexico, Costa Rica, Puerto Rico, Florida and the Gulf Coast area of North America revealed the presence of two intersterility groups (Petersen et al, 1998), a conclusion supported by independent genetic and molecular data (Hibbett et al, 1998; Nicholson et al, 1997). Macroscopically, basidiomata of the two intersterility groups were

indistinguishable, but microscopic analyses showed that they could be separated into two morphotaxa distinguished by the shape of cheilocystidia, caulocystidia, and basidiospore dimensions (Mata et al, 2000; Petersen et al, 1998). Consequently, these micromorphological characters gained more taxonomic importance when identifying *L. boryana* and sister taxa. Previous comprehensive studies which included *L. boryana* and putatively synonymous names left the presence of cystidia unnoticed (Singer, 1952, 1955; Pegler, 1975, 1983a,b) although these structures were reported by other authors (Horak, 1968; Guzmán et al, 1997).

The objective of this paper was to determine which of the two biological species is to be represented by the name *L. boryana*, and to apply the most priorable name (if any) to the other biological species. To answer this question, examination of type specimens of epithets listed by Pegler (1983a) as synonymous under *L. boryana* was carried out. This paper reports on the morphological characters of type specimens and develops a nomenclator for the two morphospecies. In those cases where designation of type specimens was not explicit (Pegler, 1975, 1983a,b) or type specimens were missing, proper typification has been proposed according to Art. 9.9, 9.10 and 9.11 of the ICBN (Greuter, 2000).

Material and methods

Morphological observations. All macroscopic observations were performed on dried material. Fungal contaminants detected on basidiomata were removed with a fine-tipped brush. Sections of basidiomata were rehydrated first in 95% ethanol and then in water for as long as 24 hours, and then placed in 3% KOH. Preparations were observed under phase contrast microscopy or bright field with Congo Red and/or phloxine. Melzer's reagent was used to test for amyloidity.

Terminology. Basidiospore measurements and statistics are as follow: range of spore length X spore width is given for all measured basidiospores; n = total number of spores measured, x = arithmetic mean of spore length and spore width for all spores

measured; Q = spore length divided by spore width, indicated as a range of variation in n spores measured; Qx = arithmetic mean of Q-values. Color names in quotations are from Ridgway (1912), names followed by alphanumerical codes in parentheses are from Kornerup and Wanscher (1978).

Results

Agaricus boryanus Berkeley & Montagne. 1849. Ann. Sci. Nat., Bot. Ser. 3, 11: 235.

HOLOTYPE: Brazil, Bahia, Blanchet, no. 24, K [!]

≡ *Collybia boryana* (Berk. & Mont.) Saccardo. 1887. Syll. Fung. 5: 240.

≡ *Gymnopus boryanus* (Berk. & Mont.) Murrill. 1916. North Amer. Fl. 9: 370.

≡ *Armillaria boryana* (Berk. & Mont.) Murrill. 1939. Bull. Torrey Bot. Club 66: 31.

≡ *Lentinus boryanus* (Berk. & Mont.) Singer. 1955. Sydowia 9: 379.

≡ *Lentinula boryana* (Berk. & Mont.) Pegler. 1975. Kavaka 3: 19.

Basidiomata three, pressed and glued on white cardboard; two basidiomata with lamellae upwards, the other showing surface details of the pileus; one basidiome with a complete stipe, one basidiome partially damaged by insects. **Pileus** 3-4 cm diam, circular in shape, probably convex when fresh, surface subvelutinous to subfibrillose (but epicutis apparently missing over some areas), apparently hygrophanous at disc, translucent outwards, light brown (5C6 to 5D6) at disc, darker yellowish brown (5E8) towards margin. **Lamellae** adnate, but seceding (probably during drying), forming a pseudo-collarium, close to crowded, now ± concolorous with pileus; margin entire or eroded; lamellulae in at least three tiers. **Stipe** now 1.5 X 0.2-0.3 cm, central; ± widened at apex and base, striate to sulcate, fibrillose near apex, becoming twisted downwards, in middle section bumpy to warty, dark brown (6F8). No evidence of annulus or velar remains.

Pileipellis a compact, repent layer of interwoven hyphae; hyphae 4-12 μm diam, ± gelatinized, with clamp connections, hyaline singly, light brown in mass; walls thin.

Pileus trama loosely interwoven; hyphae 4-16 μm diam, with clamp connections, hyaline; walls 2-4 μm thick. **Lamellar trama** parallel; hyphae 4-14 μm diam, with clamp connections; walls up to 1.6 μm thick. Hymenium composed of two elements: 1) **basidia** 12-16 X 3-4 μm, clavate; sterigmata 1-4; and 2) **basidioles** equal in size to basidia, clavate to ampullaceous; numerous. **Pleurocystidia** not observed. **Cheilocystidia** (Fig. 1a) 14-21 X 4-10 μm, clavate to broadly clavate, uncommon, often collapsed; apex obtuse. **Stipe trama** parallel; hyphae 2.5-7 μm diam, with clamp connections, hyaline singly, straw color in mass; walls 1-2 μm thick. **Caulocystidia** (Fig. 1b) 12-44 X 3-8 μm, clavate to subventricose; apex obtuse, sub-mucronate or sub-ampullaceous. **Basidiospores** (Fig. 1c) 4.8-6.4 X 2.4- 3.2 μm, (n = 20, x = 5.9 X 2.7 μm, Q = 1.71-2.67, Qx = 2.24), ellipsoid, subovoid to subcylindrical, compressed laterally or partially collapsed, hyaline, inamyloid; wall smooth, thin.

Comment: Singer (1955) and Pegler (1975, 1983a,b) overlooked the presence of cystidia (cheilo-, caulo-) in this type. Because of the preservation process of the type basidiomata, tissues became cemented, giving them a caramelized appearance. As a result, initial distinction of individual cells under light microscopy was very difficult. Only after prolonged re-hydration and squashing of the tissues could cheilocystidia, caulocystidia, and basidiospores be observed. Shape of cheilocystidia and caulocystidia in *A. boryanus* range from clavate to broadly clavate (i.e. subsphaeropedunculate) closely resembling the shape of those structures in modern collections of *L. boryana* (Guzmán et al, 1997; Mata et al, 2000; Petersen et al, 1998). Basidiospore dimensions and other microstructures of this type collection also coincide with those observed by us in fresh material. Singer (1955) reported gelatinization of the hymenophoral and pileus trama, which we have observed in some modern collections, but like Pegler (1983a) we do not consider this phenomenon as taxonomically informative as spores and cystidia, because of its irregular manifestation. Our observations agree with those of previous authors on other characters such as disposition of lamellar trama, hyphal width, and arrangement and composition of the pileus epicutis.

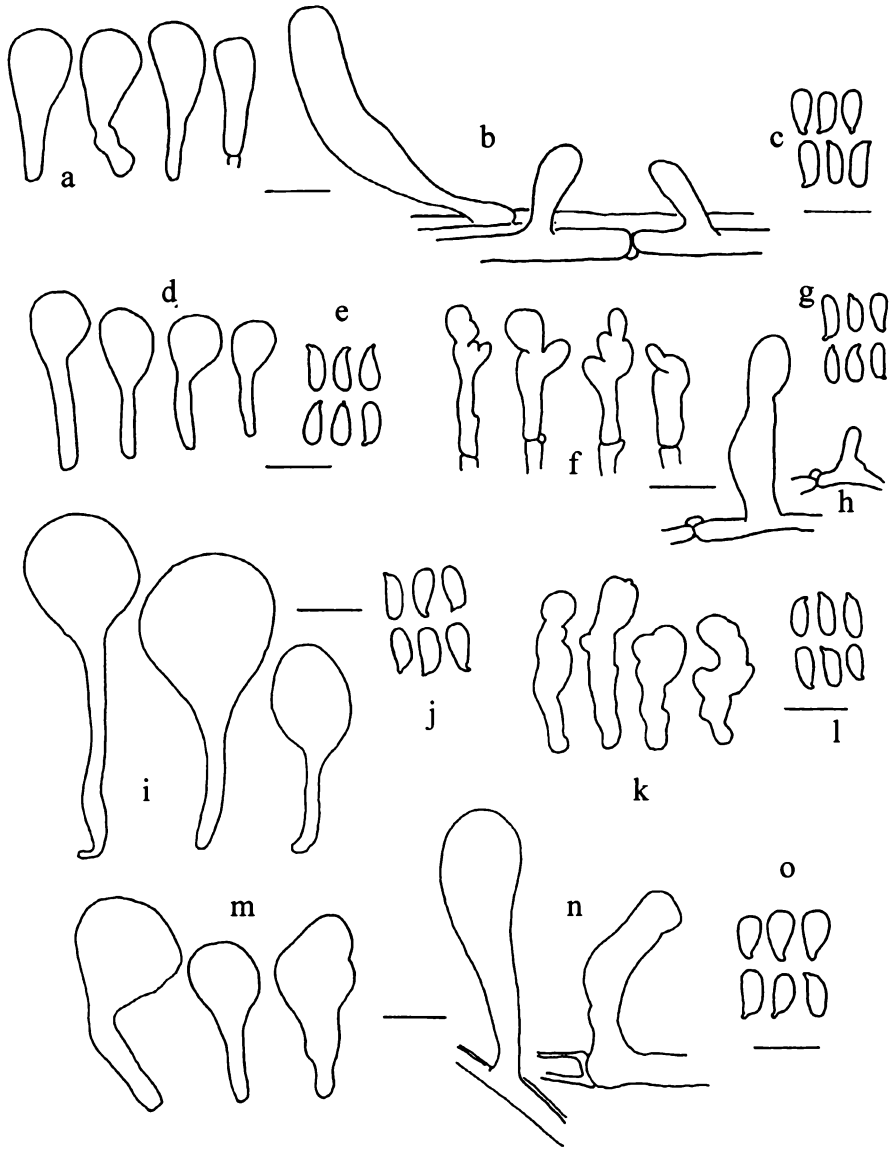

Fig. 1. Micromorphology of type specimens of *A. boryanus, A. ixodes, A. raphanica, A. umbilicata,* and *G. alliaceus.*

A. boryanus. a) cheilocystidia, b) caulocystidia, c) spores. *A. ixodes*. d) cheilocystidia, e) spores. *A. raphanica*. f) cheilocystidia, g) caulocystidia, h) spores. *A. umbilicata*. i) cheilocystidia, j) spores. *G. alliaceus*. k) cheilocystidia, l) spores. *L. cubensis*. m) cheilocystidia, n) caulocystidia, o) spores. Scale bar = 10 μm for all line drawings.

Lentinula boryana is the most priorable name for any morphospecies of *Lentinula* in the New World exhibiting the above combination of micromorphological characters.

Agaricus ixodes Montagne. 1854. Ann. Sci. Nat., Bot. Ser. 4, 1: 95.

HOLOTYPE: Guyana, Leprieur, no. 982 (ex Patouillard # 4055) FH [!]

≡ *Collybia ixodes* (Mont.) Saccardo. 1887. Syll. Fung. 5: 232.

≡ *Lentinus ixodes* (Mont.) Singer. 1952. Lilloa 25: 479.

Basidiomata two, pressed flat, moldy, one complete, the other broken. **Pileus** about 40 mm diam, probably convex when fresh, tan; surface glabrous; margin curved. **Lamellae** seceding, crowded, narrow. **Stipe** about 30 X 3 mm, slightly eccentric, fibrillose, with scattered squamulae, tough. No evidence of veil or annulus.

Pileipellis a compacted, repent layer of interwoven, radially arranged filamentous hyphae; hyphae 2-8 μm diam, pigment-encrusted, light brown in mass, with clamp connections; wall thin. **Pileus trama** loosely interwoven; hyphae 4-10 μm diam, hyaline, with clamp connections; wall up to 3 μm thick. **Lamellar trama** regular to subregular, sometimes interwoven; hyphae 2-20 μm diam, hyaline, with clamp connections; wall up to 1.6 μm thick. **Basidia** 15-18 X 4-7 μm, clavate; sterigmata 4. **Pleurocystidia** not observed. **Cheilocystidia** (Fig. 1d) 15-23 X 4-8 μm, clavate to broadly clavate, occasionally sphaeropedunculate; wall smooth, thin. Lamella margin sometimes fertile. **Stipe trama** parallel; hyphae 3-9 μm diam, straw colored, with clamp connections; wall up to 1.6 μm thick. **Caulocystidia** not observed. **Basidiospores** (Fig. 1e) 5.2-7.2 X 2.4-3.2 μm (n = 40, x = 6.1 X 2.7 μm; Q = 1.75-3.00, Qx = 2.27), subcylindrical to ellipsoid, ± lacrymoid-ellipsoid in side view, hyaline, inamyloid; wall thin, smooth.

Comment: Singer (1955) reported cystidioles but under our criterion these are basidioles. In our study, size and shape of cheilocystidia and basidiospores were similar to those of the type specimen of *A. boryanus*. The lamellar margin was caramelized and

cheilocystidia were not as readily evident as in the type of *A. boryanus*. Absence of caulocystidia can be explained by the preservation condition of the material. We conclude that *A. ixodes* is a taxonomic synonym of *A. boryanus*.

Armillaria raphanica Murrill. 1943. Mycologia 35: 423.

HOLOTYPE [FLAS, F 17949, missing; J. Kimbrough, pers. comm.]. LECTOTYPE [*hic designatus*]: U.S.A., Florida, Sanchez Hammock, near Gainesville, 23.VII.1938, col. West & Murrill, TENN 16075 ["part of type" *teste* Murrill] [!], ISOLECTOTYPE FH [!],

Basidiomata six, in excellent condition, two attached to woody substrate. **Pileus** now 15-35 mm diam, broadly convex; surface smooth, now light brown (5C7 to 6D8), margin incurved to inrolled. **Lamellae** seceding (apparently from drying), crowded, light brown (6C8 to 6C5); margin entire, several lamellulae of different lengths. **Stipe** now 20-30 X 2-4 mm, central to eccentric, ± equal to slightly enlarged at base; surface fibrillose to scaly towards base, longitudinally sulcate and somewhat twisted, similarly colored as pileus; consistency tough.

Pileipellis a simple cutis; hyphae 3-6 μm diam, filamentous, tightly interwoven, hyaline singly, light brown to tan in mass, inamyloid, with clamp connections; wall thin. Oleiferous hyphae occasional. **Pileus trama** 200-400 μm thick, loosely interwoven; hyphae 6-16 μm diam, hyaline, with clamp connections; wall 0.8-1.6 μm thick. **Lamellar trama** regular to subregular; hyphae 6-8 μm diam, hyaline, inamyloid, with clamp connections; wall thin to 0.8 μm thick. Subhymenium 8-15 μm thick, 3-4 cells thick, not clearly distinct from trama. **Basidia** not observed. Basidioles 11-19 X 3-6 μm, clavate to ampullaceous, hyaline, inamyloid. **Pleurocystidia** not observed. **Cheilocystidia** (Fig. 1f) 17-28 x 5-8 μm, mostly clavate, rarely sphaeropedunculate, lobed or knobbed at apex, hyaline, inamyloid; with clamp connections. Not readily evident, often collapsed. **Stipe trama** parallel; hyphae 4-14 μm diam, hyaline singly, yellowish in mass, with clamp connections; walls 2-4 μm thick. **Caulocystidia** (Fig. 1g) 18-24 X 4-5 μm, cylindrical or

clavate, flexed; apex obtuse, or as small knobs or outgrowths, scattered. **Basidiospores** from paratype at TENN (Fig. 1h) 4.8-6.0 X 2.4-2.8 μm (n = 20, x = 5.4 X 2.6 μm; Q = 1.71-2.67, Qx = 2.09), ellipsoid to subcylindrical, hyaline, inamyloid, wall thin wall, smooth.

Comment: Murrill (1943) distinguished this taxon from *A. boryanus* (*Armillaria boryana sensu* Murrill) by the closer and narrower gills, and a radish odor when drying. Spores could not be found in any basidiomata of the specimen at FH but they were measured from the type at TENN. The size of the basidiospores reported by Murrill (1943), 5 X 2 μm, falls within the range of our measurements, and spore dimensions are similar to other type specimens of *Lentinula*. Cheilocystidia and caulocystidia, not reported by Murrill, are relatively smaller than those found in any of the types of synonyms under *L. boryana*, are much more contorted and knobbed, and less apically inflated. Cystidia and spore shape and dimensions fit well with those observed in modern collections of *Lentinula* from the Gulf Coast of the U.S.A., Puerto Rico, Costa Rica and Venezuela which are supported by molecular and genetic data (Hibbett, 1999; Mata et al, 2000; Petersen et al, 1998). The holotype at FLAS (F 17949) could not be located (Kimbrough, pers. com.) and TENN 16075 serves as the best collection to be designated here as the lectotype for the name *A. raphanica*.

Following the dictates of the ICBN, we have chosen *A. raphanica* over *Gymnopus alliaceus* as the correct epithet for the "non-*boryana*" biological species on which we will report in another publication.

Armillariella umbilicata Patouillard. 1899. Bull. Soc. Mycol. France 15: 191.

LECTOTYPE [*hic designatus*]: Guadeloupe, Basse Terre, Camb Jacob, 20.III.1898, coll. Duss, no. 579, FH [!]

≡ *Armillaria umbilicata* (Pat.) Saccardo & Sydow. 1902. Syll. Fung. 16: 19.

Basidiomata 15, moldy, in fair condition. **Pileus** 0.5-40 mm diam, convex when young, aplanate-umbilicate with age; surface glabrous, now wrinkled, dark brown; margin inrolled, persistent. **Lamellae** seceding, close to crowded, rufous to rusty brown; margin entire. **Stipe** 15-20 X 1-3 mm, central, equal; surface striate to sulcate, fibrillose, dark brown.

Pileipellis a repent layer of compacted, filamentous hyphae; hyphae 3-6 μm diam, hyaline singly, light brown in mass, with clamp connections; wall thin. **Pileus trama** loosely interwoven; hyphae 4-9 μm diam, hyaline, with clamp connections; wall up to 2.4 μm thick. **Lamellar trama** regular; hyphae 4-10 μm diam, hyaline, with clamp connections; wall up to 0.8 μm thick. **Basidia** 15-22 X 6-7 μm, clavate; sterigmata 4. **Pleurocystidia** not observed. **Cheilocystidia** (Fig. 1i) 20-42 X 9-18 μm, sphaeropedunculate, occasionally broadly clavate; apex obtuse, occasionally knobbed, hyaline; wall smooth. **Stipe epicutis** parallel; hyphae 4-11 μm diam, hyaline singly, light brown in mass, with clamp connections; wall 1.6-2.4 μm thick. **Caulocystidia** not observed. **Basidiospores** (Fig. 1j) 5.6-7.2 X 2.4-3.2 μm (n = 24, x = 6.1 X 2.9 μm, Q = 1.75-2.50, Qx = 2.07), ellipsoid to subcylindrical, hyaline, inamyloid; wall thin, smooth.

Comment: Basidiospore and cheilocystidia measurements resemble those found in *A. boryanus*. Also, the presence of a fugacious veil included in Patouillard's description recalls that of *L. boryana.* No explicit designation of a holotype was indicated in the protologue for the name *A. umbilicata*, and there is a need for lectotypification because Pegler (1983a) only referred to this collection as the "type". We consider *A. umbilicata* to be a taxonomic synonym of *A. boryanus.*

Gymnopus alliaceus Murrill. 1943. Mycologia 35: 425.

HOLOTYPE: U.S.A., Florida, Magnesia Springs, 15.VIII.1938, coll. W.A. Murrill, F 17940, FLAS [!]

Basidiomata ten, in fair condition. Some basidiomata partially eaten by insects, some with eroded pileus surface. **Pileus** now 30-60 mm diam, broadly convex, ± depressed, glabrous (now wrinkled); margin curved to incurved. **Lamellae** seceding, close to ± crowded, narrow, beige to brown; margin entire. **Stipe** central, equal to ± broad at apex, striate to sulcate.

Pileipellis a repent layer of interwoven filamentous hyphae, worn off in most basidiomata; hyphae 3-8 μm diam, hyaline singly, light brown in mass, with clamp connections; wall thin. **Pileus trama** interwoven; hyphae 6-18 μm diam, with clamp connections; wall up to 2.4 μm thick. Oleiferous hyphae embedded in pileus trama. **Lamellar trama** parallel; hyphae 4-14 μm diam, hyaline, with clamp connections; wall up to 1.6 μm thick. **Basidia** 17-18 X 4-5 μm, clavate; sterigmata 4. **Pleurocystidia** not observed. **Cheilocystidia** (Fig. 1k) 12-24 X 4-5 μm, mostly clavate, some flexed; apex obtuse or knobbed; uncommon. Lamella edge fertile. **Stipe trama** parallel; hyphae 5-12 μm diam, with clamp connections; wall 0.8-3.2 μm thick. **Caulocystidia** not observed. **Basidiospores** (Fig. 1l) 5.6-6.4 X 2.4-3.2 μm, (n = 20, x = 6.0 X 2.8 μm, Q = 1.75-2.67, Qx = 2.19), narrowly ellipsoid, hyaline, inamyloid; wall thin, smooth.

Comment: All the observed microscopic characters in *G. alliaceus* are similar to those of the type specimen of *A. raphanica*. Basidiospore measurements of 5-6 x 4 μm, recorded by Murrill (1943), fall into the range reported here. Cheilocystidia are similar to those found in *A. raphanica*. Murrill (1943) did not observe any cystidia, a hint that these are not readily distinguishable from other hymenial elements. The onion-like odor reported by Murrill (1943), however, resembles that of *A. raphanica*, and has been noted in some of our field collections.

We consider *G. alliaceus* as a taxonomic synonym under *A. raphanica*. Basidiomata of both type specimens exhibit the same combination of macroscopic and microscopic characters. The ICBN directs us to choose between simultaneously published, taxonomically indistinguishable names, and we have done so.

Lentinus cubensis Berkeley & Curtis. 1869. J. Linn. Soc., Bot. 10: 302.

LECTOTYPE [*hic designatus*]: Cuba, coll. C. Wright, no. 115, K [!]

≡ *Lentinula cubensis* (Berk. & Curt.) Earle. 1909. Bull. New York Bot. Gard. 5: 416.

Basidiomata two, moldy; one is pressed flat. **Pileus** 12-24 mm diam, circular, probably convex; surface ± smooth, yellow brown (5C7, 5E8); margin slightly incurved. **Lamellae** adnate, seceding, forming a "pseudo-collarium", close, pale orangeyish (5A4) where not covered by mold; margin entire or eroded; lamellulae in at least two tiers. **Stipe** 12-15 X 1-2 mm, central to slightly eccentric, ± widened at apex and base, appearing sulcate, fibrillose to ± scaly. No veil or annulus observed.

Pileipellis a repent layer of compacted, interwoven hyphae, embedded in a ± gelatinous matrix; hyphae 2-6 μm diam, hyaline singly, yellow-brown in mass, with clamp connections; wall thin. **Pileus trama** loosely interwoven; hyphae 6-16 μm diam, hyaline, with clamp connections; wall 1-3 μm thick. **Lamellar trama** parallel at first, becoming irregular to interwoven at lamellar edge; hyphae 4-10 μm diam, with clamp connections; wall 2-4 μm thick. Subhymenium poorly developed. Hymenium composed of two elements: 1) **basidia** 12-19 x 4-7 μm, clavate; sterigmata 1-4, most with only two visible sterigmata; and 2) **basidioles** numerous, equal in size to basidia, clavate or ampullaceous. **Pleurocystidia** none. **Cheilocystidia** (Fig. 1m) 16-30 x 5-14 μm, clavate to broadly clavate, or sphaeropedunculate, mostly collapsed. **Stipe trama** parallel; hyphae 2-7 μm diam, hyaline singly, straw color in mass, with clamp connections; wall up to 1.5 μm thick. **Caulocystidia** (Fig. 1n) 12-32 x 5-12 μm, cylindrical to broadly clavate, with obtuse or mucronate apex. **Basidiospores** (Fig. 1o) 4.8-6.0 x 2.4-3.2 μm (n = 40, x = 5.5 X 2.9 μm, Q = 1.71-2.33, Qx = 1.95), ellipsoid, subovoid, hyaline; wall smooth, thin.

Comment: Earle (1909) proposed *Lentinula* as a new genus and designated *L. cubensis* as its type species, but *A. boryanus* is an earlier epithet for the species (see Singer, 1955). Unlike Horak (1968), we found basidiospores on a paradermal section of the pileus, the spore ranges and averages being similar to those observed in *A. boryanus*. Cheilocystidia match those of the type of *A. boryanus*, although some cheilocystidia present outgrowths which are not uncommon in more recent collections of *L. boryana*.

There is no explicit designation of a lectotype specimen for *L. cubensis* in previous studies, and collection Wright no. 115 is cited as the "K type" in the two of the three keystone works for *L. boryana* (Pegler, 1975, 1983a, b). It is the intention here to resolve this situation by explicitly designating Wright no. 115 as the lectotype specimen of *L. cubensis*. Microscopic examination of paratype specimens Wright no. 80, 121, 122 (FH) confirmed their identity as *L. cubensis*. Like previous workers (Pegler 1975, 1983a, b; Singer, 1952, 1955) we consider *L. cubensis* to be a taxonomic synonym of *A. boryanus*.

Lentinus detonsus Fries. 1851. Nov. Symb. Mycol., p. 38.

HOLOTYPE: Costa Rica, Oersted, missing. NEOTYPE [*hic designatus*]: Costa Rica, Prov. San José, Jardín de Dota, 3.5 km W. of Inter-American Highway at El Empalme, 9° 42' 52" N, 83° 58' 28" W, elev. 2220 m, 16.VI.1995, coll. R.E. Halling, field no. 7814 (TENN 53824) [!]

≡ *Lentinula detonsa* (Fr.) Murrill. 1911. Mycologia 3:28.

Basidioma one, air-dried, in good condition. **Pileus** convex when young, in age lobate to depressed, disc "avellanous" to "tilleul buff" (7B3 - 7B2). **Lamellae** subventricose, free, close, off-white to "tilleul buff" (7B2); margin entire. **Stipe** somewhat bulbous at base, scrobiculate, concolorous with gills, at base "army brown" to

"natal brown" (8D5 - 8E6); flesh white; consistency tough; veil apparently absent. Odor of cut lamellae very fungus-like, taste not acrid

Pileipellis a layer of repent, tightly interwoven hyphae; hyphae 2-8 μm diam, pigmented light brown, with clamp connections; wall thin. **Pileus trama** loosely interwoven; hyphae 6-16 μm diam, constricted at septa, with clamp connections; wall 2-4 μm thick. **Lamellar trama** regular to subregular; hyphae 3-7 μm diam; walls up to 1 μm thick. **Basidia** 21-26 X 6-7 μm, clavate; sterigmata 4. **Pleurocystidia** not observed. **Cheilocystidia** (Fig. 2a) 25-36 X 10-14 μm, clavate, broadly clavate to sphaeropedunculate, with clamp connections; apex obtuse or irregularly lobed. **Stipe trama** parallel; hyphae 3-6 μm diam, with clamp connections; walls up to 1.5 μm thick. **Caulocystidia** (Fig. 2b) 20-25 X 5-8 μm, clavate, uncommon to rare; apex obtuse or irregularly lobed. **Basidiospores** (Fig. 2c) 6.0-8.0 X 3.2-4.0 μm (n = 20, x = 7.0 X 3.4 μm; Q = 1.78-2.25, Qx = 2.06), ellipsoid, ovoid, hyaline, inamyloid; wall thin, smooth .

Comment: An Oersted specimen under *Lentinus detonsus* is not resident at UPS (herb. Fries) or C (herb. Oersted). With no holotype or lectotype material available, we are obliged to neotypify the name in order to secure its identity. The neotype is from the same country of origin (Costa Rica), exhibits the morphological characters of *L. boryana*, and therefore serves to tie more firmly the name *L. detonsus* and *L. boryana* as listed by Pegler (1983a). Cystidia and basidiospores found in this neotype are typical for *L. boryana*. The name *Lentinula detonsa* has been misapplied to taxa collected along the Gulf Coast of the U.S.A. (Metzler and Metzler, 1992), which actually correspond to *A. raphanica*.

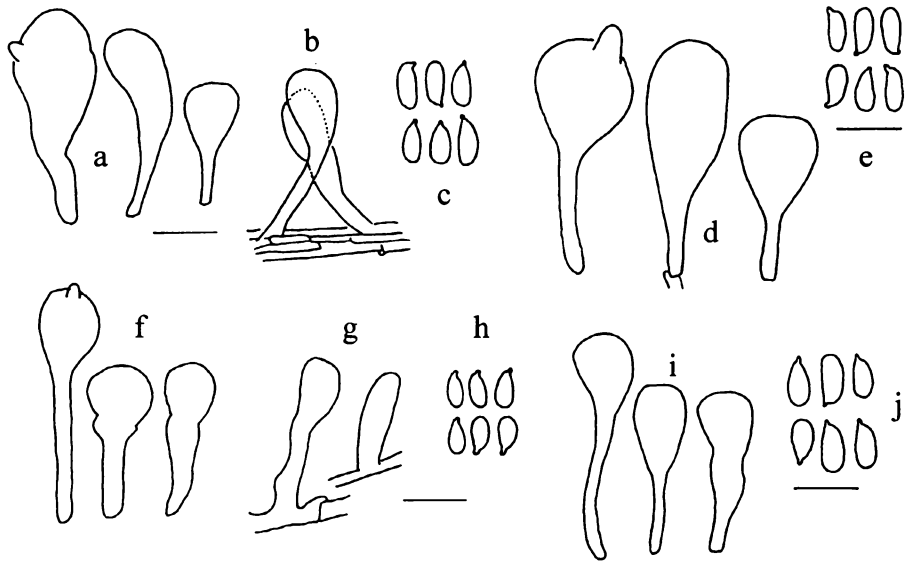

Fig. 2. Micromorphology of type specimens of *L. detonsus*, *L. proximus*, *L. puiggarii,* and *L. leprieurii.*

L. detonsus. a) cheilocystidia, b) caulocystidia, c) spores. *L. proximus*. d) cheilocystidia, e) spores. *L. puiggarii*. f) cheilocystidia, g) caulocystidia, h) spores. *P. leprieurii*. i) cheilocystidia, j) spores. Scale bar = 10 μm for all line drawings.

Lentinus proximus Berkeley & Curtis. 1869. J. Linn. Soc., Bot. 10:302.

LECTOTYPE [*hic designatus*]: Cuba, C. Wright, no. 117, FH [!].

Basidiomata two, moldy, lamellae partially eaten by insects. **Pileus** convex; surface ± smooth to fibrillose; margin inrolled. **Lamellae** adnate-seceding, forming a "pseudo-collarium", close, ± broad; margin entire or eroded; lamellulae in two tiers. **Stipe** central; ± widened at base, appearing sulcate, fibrillose. No evidence of veil or annulus.

Pileipellis a compact, repent layer of interwoven hyphae; hyphae 3-6 µm diam, hyaline singly, light brown in mass, with clamp connections; wall thin. **Pileus trama** loosely interwoven; hyphae 6-14 µm diam, hyaline, with clamp connections; wall 1-3 µm thick. **Lamellar trama** parallel; hyphae 4-12 µm diam, with clamp connections; wall 0.5-2 µm thick. Subhymenium poorly developed. **Basidia** 14-18 X 4-7 µm, clavate, often collapsed; sterigmata 1-4, often only two visible. **Pleurocystidia** not observed. **Cheilocystidia** (Fig. 2d) 12-33 X 5-12 µm, clavate to broadly clavate, often collapsed; apex obtuse, or irregularly lobed to branched. **Stipe trama** parallel; hyphae 2-10 µm diam, hyaline singly, straw colored in mass, with clamp connections; wall 0.8-2.4 µm thick. **Caulocystidia** not observed. **Basidiospores** (Fig. 2e) 5.6–7.2 X 2.4-3.2 µm, (n = 20, x = 6.4 X 2.9 µm, Q = 2.0-2.67, Qx = 2.19), ellipsoid, subovoid to subcylindrical, hyaline, inamyloid; wall thin, smooth.

Comment: All micromorphological characters conform to those described for *A. boryanus* (see above). The stipes of the basidiomata are very contaminated with mold and confirmation of caulocystidia was not possible. Previous workers did not expressly designate a lectotype for *L. proximus* (Pegler, 1975, 1983a), and now we propose collection Wright no. 117 to serve this function. We consider *L. proximus* to be a taxonomic synonym of *A. boryanus*.

Lentinus puiggarii Spegazzini. 1919. Bol. Acad. Nac. Cienc. Córdoba 23: 387.

HOLOTYPE: Brazil, Apiahy, L. Puiggari, no. 86, LPS [!].

Basidiomata five, all in very deteriorated and moldy condition, broken or worn off, some eaten by insects. **Pileus** 5-15 mm diam; surface glabrous, brownish with olive-gray hues. **Lamellae** seceding, close, somewhat narrow, brown; margin eroded in most basidiomata. **Stipe** central, striate-sulcate, somewhat wider at base, light brown; consistency tough. On wood.

Pilepellis a repent layer of tightly interwoven filamentous hyphae; hyphae 3-6 µm diam, hyaline singly, light brown in mass, with clamp connections; wall thin. **Pileus trama** ± loosely interwoven; hyphae 4-10 µm diam, hyaline, with clamp connections; wall 0.8-2.4 µm thick. **Lamellar trama** regular; hyphae 4-12 µm diam, hyaline, with clamp connections. Hymenium 12-20 µm thick; basidia indistinguishable from basidioles; sterigmata collapsed. **Pleurocystidia** not observed. **Cheilocystidia** (Fig. 2f) 19-32 X 4-8 µm, clavate to broadly clavate or sphaeropedunculate, often collapsed. **Stipe trama** parallel; hyphae 4-12 µm diam, hyaline singly, yellowish in mass, with clamp connections; wall 0.8-2.4 µm thick. **Caulocystidia** (Fig. 2g) 21-25 X 4-8 µm, cylindrical to clavate, flexous. **Basidiospores**: (Fig. 2h) 4.4-5.6 X 2.0-2.8 µm (n = 14, 5.1 X 2.4 µm, Qx = 1.71-2.80, Q = 2.16), lacrymoid in side view, ellipsoid to subovoid in profile, collapsed, hyaline, inamyloid; wall thin, smooth.

Comment: On the original voucher folder there are drawings of five basidiospores, 6 X 3 µm, ellipsoid in profile. Singer (1952) provided descriptions of what he considered *L. puiggarii* from fresh material, the basidiospores measuring 5-7.3 X 2.8–4.3 µm, but gave no report of cystidia. However, Singer (1952) thought this species to be related to the types of *A. ixodes* and *L. cubensis*, both synonyms of *A. boryanus*. In our view, cheilocystidia, caulocystidia and basidiospore dimensions match those found on the type specimen of *A. boryanus*.

Panus leprieurii Montagne 1854. Ann. Sci. Nat., Bot. Ser. 4, 1:121 [*non Lentinus leprieurii* Mont. 1854]
LECTOTYPE [*hic designatus*]: Guyana, Cayenne, Leprieur, no. 980, K [!]

Basidioma one, pressed flat, glued to cardboard, severely covered by mold, broken at stipe. **Pileus** about 6 mm diam, probably convex, color not detectable, margin inrolled. **Lamellae** seceding, close, narrow, margin brown to hazel brown, apparently smooth. **Stipe** nearly central, short, striated longitudinally. On wood.

Pileipellis a simple cutis; hyphae 2-4 μm diam, filamentous, radially oriented, somewhat gelatinized, hyaline singly, light brown in mass, with clamp connections; wall thin. **Pileus trama** interwoven; hyphae 3-11 μm diam, hyaline singly, yellowish in mass, with clamp connections; wall thin. **Lamellar trama** subregular; hyphae 2-8 μm diam, hyaline, with clamp connections; wall thin. Hymenium composed of two elements: 1) **basidia** 15-21 X 5-7 μm, clavate, hyaline; wall thin; sterigmata 4; and 2) **basidioles** similarly sized, clavate to mucronate. **Pleurocystidia** not observed. **Cheilocystidia** (Fig. 2i) 18-27 X 5-8 μm, mostly clavate, few sphaeropedunculate, some knobbed; apex obtuse, hyaline; wall thin. **Stipe epicutis** parallel; hyphae 3-8 μm diam, hyaline singly, yellowish in mass, with clamp connections; wall thin to 1.6 μm thick. **Caulocystidia** not observed. **Basidiospores** (Fig. 2j) 5.2-7.2 X 2.8-3.6 μm (n = 27, x = 6.0 X 3.0 μm; Q = 1.75-2.29, Qx = 1.98), pip-shaped in side view, ellipsoid to subcylindrical in profile, hyaline; wall thin, smooth.

Comment: The protologue description by Montagne (1854) of the type specimen coincides with habit and stature of modern day *L. boryana*. Montagne (1854) noticed a veil, scales on the pileus, tough consistency, and seceding lamellae typical of *Lentinula*. Cheilocystidia are not readily distinguished in size from basidioles, but distinct by their shape, but are similar to the cheilocystidia of the type specimen of *A. boryanus*. Heavy

mold infestation made detection of caulocystidia impossible. Pileus epicutis gelatinization is similar to that observed in other type specimens under *A. boryanus*.

Collection Leprieur no. 980 is designated here as the explicit lectotype of *P. leprieurii*. The basidiome represents a younger stage of the paratype collection Leprieur no. 981, which we also have examined.

SUMMARIZED NOMENCLATORS (for detailed nomenclators see above)

Lentinula boryana (Berk. & Mont.) Pegler. 1975. Kavaka 3:19.

≡ *Agaricus boryanus* Berkeley & Montagne. 1849. Ann. Sci. Nat., Bot. Ser. 3, 11: 325

= *Lentinus detonsus* Fries. 1851. Nov. Symb. Mycol. [Saccardo 5:597]

= *Panus leprieurii* Montagne. 1854. Ann. Sci. Nat., Bot. Ser. 4, 1: 121 [*non Lentinus leprieurii* Mont. 1854]

= *Agaricus ixodes* Montagne. 1854. Ann. Sci. Nat., Bot. Ser. 4, 1: 95.

= *Lentinus cubensis* Berkeley & Curtis. 1869. J. Linn. Soc., Bot. 10: 302.

= *Lentinus proximus* Berkeley & Curtis. 1869. J. Linn. Soc., Bot. 10:302.

= *Armillariella umbilicata* Patouillard. 1899. Bull. Soc. Mycol. France. 15: 191.

= *Lentinus puiggarii* Spegazzini. 1919. Bol. Acad. Nac. Cienc. Córdoba 23: 387.

Lentinula raphanica (Murrill) Mata & R.H. Petersen *comb. nov.*

≡ *Armillaria raphanica* Murrill. 1943. Mycologia 35: 35:422.

= *Gymnopus alliaceus* Murrill. 1943. Mycologia 35: 425.

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PART IV

The genus *Lentinula* in the Americas

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Abstract

Recent studies have reported only *Lentinula boryana* and *L. guarapiensis* from the American continent, the latter known only from its type specimen. We report at least three extant species. Two widely distributed species, *L. boryana* and *L. raphanica comb. nov.*, are discussed in this paper, and a new species is being described elsewhere. The geographic ranges of both species apparently overlap throughout tropical and subtropical America except for the Gulf Coast region of the United States, from which only *L. raphanica* can be reported. *Lentinula boryana* and *L. raphanica* differ in basidiome micromorphology, especially cheilocystidial form, spore dimensions, sexual compatibility profiles, and ITS sequences.

Key words: Agaricales, consensus tree, intersterility group, nomenclature

Introduction

The genus *Lentinula* is best known for the extensively cultivated Shiitake mushroom *Lentinula edodes* (Berk.) Pegler (Chiu et al, 1999; Hibbett and Donoghue, 1996; Pegler, 1983). *Lentinula edodes* and *L. boryana* (Berk. & Mont.) Pegler were the first species to be placed in this genus (Pegler 1975) followed by *L. lateritia* (Berk.) Pegler, *L. novaezelandiae* (Stev.) Pegler, and *L. guarapiensis* (Speg.) Pegler in a monograph of the genus (Pegler, 1983). *Lentinula aciculospora* Mata & Petersen, is a newly described species from southern Central America (Mata and Petersen, 2000). *Lentinula edodes*, *L. lateritia* and *L. novaezelandiae* have an Asian-Australasian (Old World) distribution (Hibbett et al, 1995, 1998; Pegler, 1983), while the remaining species occur only in the American continent (New World). Mushrooms in the genus *Lentinula*

are saprobic, causing wood decay on logs of broadleaf trees, especially Fagales (*Quercus* and allies) (Hibbett and Donoghue, 1996; Pegler, 1983).

Old World morphospecies differ macro- and microscopically, but all collections tested have been mating-compatible, suggesting that they could form one single biological species, i.e. *L. edodes* (Mori et al, 1974; Shimomura et al, 1992). Old World *Lentinula* populations produce identifiable isoenzyme patterns (Fukuda and Tokimoto, 1991), RFLP signatures (Fukuda et al, 1994) and definite phylogenetic ITS-based lineages (Hibbett et al, 1995, 1998). Hibbett et al (1998) discussed an alternative classification for *Lentinula* based on evolutionary species concepts in order to better reflect phylogenetic relationships in *L. edodes* (Hibbett, 1992; Hibbett et al, 1995, 1998) and to encourage the conservation of genetic diversity of *Lentinula* (Hibbett and Donoghue, 1996). Molecular phylogenies reveal at least five species in Old World *Lentinula* (Hibbett et al, 1995) and molecular clock analysis supports the view that present day distribution is the result of the combination of vicariance events and island hopping (Hibbett, 2001).

The mating systems reported for basidiomata belonging to *L. boryana* and all morphological variants of *L. edodes* are tetrapolar (Fox et al, 1994; Mata and Guzmán, 1989b; Petersen et al, 1998; Takemaru, 1961). *Lentinula boryana*, however, is genetically isolated and morphologically distinct from *L. edodes* (Guzmán et al, 1997; Mata and Guzmán, 1989a; Pegler, 1983). Based on Pegler's (1983) monograph, it has been assumed that only *L. boryana* fruited in the American continent because *L. guarapiensis* is a species only known from its type collection (Guzmán et al, 1997; Nicholson et al, 1997; Pegler, 1983). Inference from results of studies on the type specimen of *L. guarapiensis* made by Singer (1952) and Pegler (1983) suggest this name may be doubtful. For these reasons the latter is not included in this paper.

New World *Lentinula* has been suggested as the earliest lineage to diverge within the genus while evolution of Old World species has taken place more recently (Nicholson

et al, 1997), an assumption supported by the higher levels of sequence divergence found between New World *Lentinula* than those from Asia-Australasia (Hibbett et al, 1998). Thon & Royse (1999) reported two separate clades or lineages within *L. boryana*, one clade with nearly identical ITS sequences for collections from Brazil and Venezuela, concluding that either sexual reproduction over long geographical range or rapid population expansion could explain this similarity. All of these previous conclusions were based on the assumption that all New World collections represented a single species name, *L. boryana*. However, Petersen et al (1998) noted that two intersterility groups existed within *L. boryana* and could be distinguished by ITS patterns and basidiome micromorphology.

Over several years, specimens of putative *L. boryana* were gathered from several geographic localities in the American Continent, and many of them were vouchered by mono- and dikaryon cultures. Discovery of a new species, *L. aciculospora*, (Mata and Petersen, 2000) and the revelation of two distinct intersterility groups within *L. boryana*, also supported by DNA analyses, impose an obligation to characterize these biological species in the American Continent. The objective of this paper is to describe such species and compare them in relation to other *Lentinula* species.

Materials and methods

Definition of terms. Terms used below are defined as follows: 1) single-basidiospore isolate (SBI) = a culture resulting from the germination of a single basidiospore harvested from spore prints from basidiomata in nature; 2) morph or morphological species = collections of basidiomata which differ sufficiently in morphology (macro- and/or micro-) to have been described as separate taxa; 3) biological species (i.e. intersterility group) = collections of one or more morphs which exhibit sexual intercompatibility in pairings among SBIs; 4) sexual compatibility = ability of hyphae of two monokaryon isolates to anastomose, resulting in nuclear migration (uni- or bilateral) and a dikaryon mycelium exhibiting clamp connections (in some other fungal groups, dikaryon hyphae do not exhibit clamp connections, but the definition applies to

Lentinula); 5) partial compatibility = in a given pairing experiment, less than 100% compatibility among SBIs (a numerical value only, not a measure of anastomoses or nuclear migration).

Colors in quotation marks are from Ridgway (1912) and those followed by an alphanumeric code in parentheses are from Kornerup and Wanscher (1978). Basidiospore statistics are as follows: A = mean of length and width for all measured spores ± standard deviation; Q = minimum and maximum quotients, spore length divided by spore width, for all measured spores; Qx = mean of Q for all measured spores ± standard deviation; n = total number of spores measured; numbers after backslash = number of specimens studied.

Specimens used. In the list of specimens utilized below B = basidiomata examined morphologically, SBI = single-basidiospore isolates used in mating studies. Unless otherwise noted, specimens with CLO, ET, FB, HRB and REH numbers are deposited in TENN, those with an F are in FLAS, and TJB numbers are in CORT. GB numbers correspond to sequences deposited in GenBank.

Lentinula boryana. BRAZIL. BAHIA: no date, *Blanchet* 24 (HOLOTYPE of *Lentinula boryana*, K); Apiahy, *L. Puiggari 86* (HOLOTYPE of *Lentinus puiggarii*, LPS). PARANA: Quatro Barras, Borda do Campo, Morro Anhangava, 1100 m, 15-VIII-1999, *A.R. de Meijer FB 10827* (GB AY016440), B, SBI. COSTA RICA. CARTAGO: Palo Verde, 24-VI-2000, *J.L. Mata & R.H. Petersen FB 11008,* B, SBI. SAN JOSE: Jardín de Dota, 3.5 km W. of Inter-American Highway at El Empalme, 9°42'52" N, 83°58'28" W, 2220 m, 16-VI-1995, *R.H. Petersen & R.E. Halling FB 7814* [NEOTYPE of *Lentinus detonsus,* Des. Mata & Petersen (2001)], B, SBI. Same locality, 15-VI-1995, *R.H. Petersen FB 7810,* B, SBI. Same locality, 28-III-1995, *R.E. Halling, REH 7414,* B. KM 66 on Inter-American Highway, Finca El Jaular, 9°30'61"N, 83°52'05" W, 2234 m, 1-VII-1998, *R.H. Petersen FB 9469,* B, SBI. San Gerardo de Dota, 16-VI-1995, *R.H. Petersen FB 7829,* B, SBI. CUBA: no date, *C. Wright 115* [LECTOTYPE of *Lentinus cubensis*, K, Des. Mata & Petersen (2001)]. No date, *C. Wright 117* [LECTOTYPE of

Lentinus proximus, FH, Des. Mata & Petersen (2001)]. GUADELOUPE: Basse Terre, Camb Jacob, 20-III-1898, *Duss 579* [LECTOTYPE of *Armillariella umbilicata*, FH, Des. Mata & Petersen (2001)]. GUYANA: Cayenne, no date, *Leprieur 980* [LECTOTYPE of *Panus leprieurii*, K, Des. Mata & Petersen (2001)]. No date, *Leprieur 982* (HOLOTYPE of *Agaricus ixodes*, FH). MEXICO. MEXICO: vic. Temescaltepec, El Polvorin Mt., 2200 m, 4-VII-1996, *S. Sierra FB 8407,* B, SBI. Rd. to Valle de Bravo, KM 21 (RTE 18), 2700 m, 5-VII-1996, *J. Cifuentes FB 8421,* B, SBI. vic. Valle de Bravo, RTE 861, 4 KM from intersection, 2400 m, 6-VII-1996, *R.H. Petersen FB 8436* B, SBI. 15 KM N of Chapa de Mota, 2600 m, 9-VII-1996, *R.H. Petersen & M. Rodriguez FB 8467,* B, SBI. Dist. Federal, Ajusco, 22-VI-1996, *A. Estrada Torres ET 3784*, spore print only, SBI. Unknown locality, no date, *A. Estrada Torres ET 3349*, spore print only, SBI. PANAMA. PANAMA: Gatun Lake, Barro Colorado Island, 24-V-1997, *C.L. Ovrebo CLO 3521,* B. Same locality, 24-V-1997, *C.L. Ovrebo CLO 3522,* B.

Lentinula raphanica. COSTA RICA. PUNTARENAS: Coto Brus, San Vito, 17-VI-1995, R.H. Petersen *FB 7841*, B. Coto Brus, Hacienda La Amistad, 8°54'22" N, 82°47'40" W, 1328 m, 5-VII-1998, J. Cifuentes FB 9929, B, SBI. Coto Brus, Las Alturas Biological Station, OTS, on the trail to Cerro Chia, 8°56'59" N, 82°50'02" W, 1520 m, 21-VI-2000, J.L. Mata & R.H. Petersen *FB 10497*, B, SBI. TRINIDAD: Port of Spain, 14-V-1921, F.J. Seaver, F 18710, B. UNITED STATES: FLORIDA, Alachua Co., vic. Gainesville, Sanchez Hammock, 23-VII-1938, E. West & W.A. Murrill, *F 17949* [ISOTYPE of Armillaria raphanica (FH), PARATYPE (TENN 16075)]. Gainesville, 1-X-1944, W.A. Murrill (as Lentinus glabratus), TENN 10174, B. Same locality, 14-VII-1938, W.A. Murrill, TENN 16072, B. Same locality, 27-VIII-1947, W.A. Murrill, (TENN 18453), B. Same locality, 23-VII-1941, W.A. Murrill *F 18142*, B. Same locality, 27-VII-1947, W.A. Murrill *F 19741*, B. Same locality, Austin Carey Hatchet Creek Area, 20-VIII-1996, T.J. Baroni & J. Kimbrough *TJB 8078*, B. Magnesia Springs, 15-VIII-1938, W.A. Murrill *F 17940* (HOLOTYPE of Gymnopus alliaceus, FLAS). Columbia Co., Camp O' Leno, 1-X-1939, W.A. Murrill *F 20089*, B. Gadsden Co., Bristol, Torreya State Park, 30°34'58" N, 84°56'95" W, 30-V-1997, K.W. Hughes, *FB 9156* (GB AY016441), B, SBI. Leon Co., Tarquin State Rec. Area, RTE. 276, 30°27'26"

N, 84°38'58" W, 29-V-1997, E.B. Lickey *FB 9217*, SBI. Liberty Co., Bristol, Apalachicola National Forest, White Oak Camp, 30°06'50" N, 85°04'84" W, 30-V-1997, K.W. Hughes *FB 9153*, B. Locality unknown, Newman's Lake, 14-VII-1938, E. West, Arnold & W.A. Murrill *F 18144*, B. Locality unknown, 8-VIII-1996, T.J. Baroni *TJB 8002*, B. LOUISIANA: E. Baton Rouge Par., Baton Rouge, Burden Plantation, Rural Life Museum, 30°25'11" N, 91°07'17" W, 25-V-1997, R.H. Petersen *FB 9119*, B, SBI. E. Feliciana Par., Francisville, St. Francis St., James John Audubon Historic Site, 30°47'84" N, 91°18'43" W, 22-V-1997, M. Blackwell *FB 9078*, B, SBI. Iberia Par., Avery Island, Jungle Gardens, 30°22'74" N, 91°38'59" W, 27-V-1997, K.W. Hughes *FB 9142*, SBI; St. Tammany Par., Fountainbleu State Park, 30°20' N, 90°02' W, 24-V-1997, E.B. Lickey *FB 9207*, SBI. St. Martin Par., Lafayette, Lake Martin, Rookery Rd., Forest to right, 30°47'85" N, 91°18'44" W, 23-V-1997, R.H. Petersen *FB 9097*, B, SBI. Same locality, 23-V-1997, R.H. Petersen *FB 9098*, B, SBI. Same locality, 23-V-1997, R.H. Petersen *FB 9099*, B, SBI. Same locality, 23-V-1997, A.D. Sime *FB 8682*, SBI. PUERTO RICO: Rio Grande, Palmer, Luquillo National Forest, El Verde Biological Station, 18°19'31" N, 65°49'20" W, 320 m, 9-VI-1998, E.B. Lickey *FB 9564* (GB AY016442), B, SBI. TEXAS: Hardin Co., Sour Lake, Aker's residence, 11-VII-1997, D.P. Lewis, TENN 57168, B; Texas Nature Conservancy Roy Larson Sandyland Sanctuary, 10-VI-2000, J.L. Mata *FB 10479*, B, SBI. VENEZUELA. AMAZONAS: Cerro de la Neblina, near Neblina base camp, along Rio Mawarinuma, 0°50' N, 66°10' W, 140 m, 15-II-1985, R.E. Halling *REH 4264*, B.

Lentinula aciculospora. COSTA RICA. SAN JOSE: Pérez Zeledón, Villa Mills, CATIE Experimental Station, 9°33'03" N, 83°40'56" W, 2880 m, 30-VI-1998, *R.H. Petersen FB 9447* (HOLOTYPE, TENN; GB AY016443), B, SBI. Cuerricí Biological Station, 9°33'17" N, 83°40'04" W, 2560 m, 21-VI-1999, *J.L. Mata FB 10418* (GB AY016444).

Lentinula edodes. AUSTRALIA. NEW SOUTH WALES: Watagan Mountains, Boarding House Dam, 21-V-1991, *R.H. Petersen FB 3577* (*lateritia* morph) B, SBI. JAPAN. TOCHIGI: vic. Lake Chuzenjiko, 27-IX-1989, *I. Izawa, FB 2306* (*edodes* morph) B, SBI. NEPAL: Ilma, V-1995, *H.R. Bhandary HRB 7683* (*edodes* morph) SBI,

spore print only. NEW ZEALAND. NORTH ISLAND: vic. Lower Hutt, Wainuiomata, Orongorongo Trailhead, 25-V-1994, *A.S. Methven FB 7563* (*novaezelandiae* morph) B, SBI.

Isolation and storage of cultures. In most cases, single-basidiospore isolates were obtained using techniques described by Gordon and Petersen (1992) and Petersen et al (1999). All cultures were maintained on malt extract (Difco 1.5%) agar (Difco Bacto 2.0%) medium (MEA) in double distilled water (ddH_2O). When spores were received as a spore print, serial dilutions of spores in sterile ddH_2O water were plated onto MEA, and germinating spores were excised after appropriate growth. Most dikaryon cultures were the product of overgrowth of germinating spores in high concentrations. Cultures are stored using the agar-disc-in-water method described by Burdsall and Dorworth (1994).

Sexual compatibility tests among New World collections. In all cases, SBIs of New World collections were examined to eliminate clamp-bearing (i.e. dikaryon) isolates. In these experiments pairings = four. In similar studies on other taxa, smaller numbers of pairings have been employed (Murphy and Miller, 1993), but we considered that a somewhat larger sampling might be required to detect partial compatibility. For certain strains collected late in the study (i.e. collections 9564 from Puerto Rico, and 9469, 9929 from Costa Rica), SBI crosses were made between the new "unknown" genotype and representative strains from identified collections (n = four).

Intercontinental sexual compatibility tests. Previous experiments (Mori et al, 1974; Shimomura et al, 1992) had shown that SBIs from the Old World morphological variants of *L. edodes* (i.e. *L. edodes*, *L. lateritia*, *L. novaezelandiae*) were intercompatible. Exemplars from Old World *Lentinula* morphs were judged eligible for pairings with exemplar collections of New World taxa (i.e. *L. boryana*, *L. raphanica*, *L. aciculospora*). In these experiments pairings = 24. This experimental design allowed each monokaryon isolate to confront two monokaryons from the potential mate and was devised in order to provide an adequate sample for detecting partial compatibility. The

crossing design was described by Petersen and McCleneghan (1996) and Petersen and Hughes (1998).

Morphology. Sections of dried specimens were re-hydrated in 95% ethanol and water, then placed in 3% KOH and observed in phase contrast, or stained with Congo Red and phloxine followed by bright field microscopy. Melzer's reagent was used to conduct amyloidity tests. For basidiomata examined early in the study, notes were taken on all micromorphological features, but as the study progressed, identification of basidiomata was made chiefly on observations of cheilocystidia and spores. In all cases, at least 10 (usually 20) structures were measured.

Molecular analysis. Monokaryon and dikaryon cultures were grown, and DNAs were extracted as described by Hughes et al (1999). The nuclear ribosomal region was PCR-amplified using primers ITS 1F and ITS 4B (Gardes and Bruns, 1993) and sequenced using primers ITS4 and ITS5 as described by Hughes et al (1999). Sequences were aligned and corrected using the Seqlab program in GCG (GCG, 2000). Parsimony analyses were performed using PAUP 4.0 (Swofford, 1996) using a branch and bound search process. Characters were unordered and equally weighted, addition sequence = furthest, maxtree setting = 100, and MulTrees option was in effect. Gaps were treated as missing data or as a fifth base. In one analysis, *Gymnopus dryophilus* (Bull.: Fr.) Murrill and *G. subnudus* (Ellis ex Peck) Halling were used as outgroups. In the second analysis, *Gymnopus* sequences were deleted and *L. edodes* sequences (Hibbett et al, 1998) were used as the outgroup. One hundred bootstraped replicates were used to evaluate node support. Sequences were deposited with GenBank (AY016440-AY016444) and the aligned sequence data were deposited in TreeBASE (Study accession number S591, Matrix accession number M890).

Results

Sexual compatibility among New World collections. When four randomly selected SBIs of each collection were crossed with four SBIs of all other collections listed

below two intersterility groups were defined (Fig. 1). These intersterility groups comprised the following collections: group I = 7810, 7814, 7829, 8407, 8421, 8436, 8467, 9469, ET-3349, ET-3784; group II = 8682, 9078, 9097, 9099, 9119, 9153, 9156, 9207, 9217, 9929, 9564. Collections from intersterility group I were collected from tropical and subtropical Mexico to Costa Rica. Collections from intersterility group II came from the North American Gulf Coast, Costa Rica, and Puerto Rico. Pairing experiments showed that *L. aciculospora* was sexually isolated from both of the above groups (Mata and Petersen 2000), and it was not included in subsequent mating experiments.

Intercontinental pairing experiments. As previously reported (Mori et al 1974; Shimomura et al 1992), there was no incompatibility among SBIs from Old World *Lentinula* morphs (Fig. 1). There was no compatibility between SBIs from *L. edodes* and intersterility group I (*L. boryana*), intersterility group II (*L. raphanica*), or *L. aciculospora* (Mata and Petersen, 2000).

Morphology. Although neither New World intersterility group could be reliably linked to a species epithet, one group was assumed to represent *L. boryana*. As explained in the description and nomenclature for each species below, micromorphological characters can be used to distinguish the two morphotaxa among all New World collections. This distinction led to application of the name *L. boryana* (for intersterility group I) and *L. raphanica* (for intersterility group II).

Lentinula boryana (Berk. & Mont.) Pegler. 1975. Kavaka 3:19.

≡ *Agaricus boryanus* Berkeley & Montagne. 1849. Ann. Sci. Nat., Bot. Ser. 3, 11: 325. [!]

= *Lentinus detonsus* Fries. 1851. Nov. Symb. Mycol. [!]

= *Panus leprieurii* Montagne. 1854. Ann. Sci. Nat., Bot. Ser. 4, 1: 121. [!]

= *Agaricus ixodes* Montagne. 1854. Ann. Sci. Nat., Bot. Ser. IV, 1: 95. [!]

= *Lentinus cubensis* Berkeley & Curtis. 1869. J. Linn. Soc., Bot. 10: 302. [!]

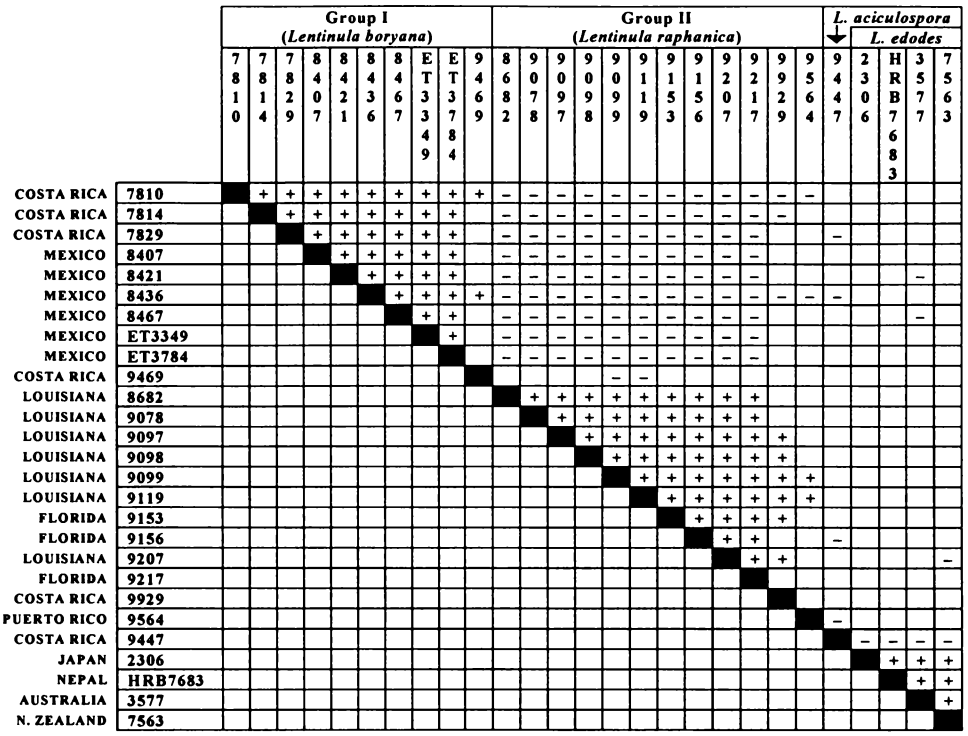

Fig. 1. Sexual compatibility grid. Intersterility group I (*L. boryana*) is composed of collections from Costa Rica and Mexico; intersterility group II (*L. raphanica*) is composed of collection from the Gulf Coast states of USA, Puerto Rico, and Costa Rica. Pairings between group I and group II were n = 4. Pairings of *L. edodes* morphs with exemplars of group-I, group-II and *L. aciculospora* were n = 24. No compatibility was observed between *L. edodes* and any New World *Lentinula*. Compatibility tests for *L. aciculospora* also reported by Mata and Petersen (2000). The symbol "+" means 100% compatibility, an "-" 0% compatibility. Blank spaces means no pairings were done.

= *Lentinus proximus* Berkeley & Curtis. 1869. J. Linn. Soc., Bot. 10:302. [!]

= *Armillariella umbilicata* Patouillard. 1899. Bull. Soc. Mycol. France. 15: 191. [!]

= *Lentinus puiggarii* Spegazzini. 1919. Bol. Acad. Nac. Cienc. Córdoba 23: 387. [!]

Pileus 18-25 mm wide, convex to convex with depressed center, or shallowly conchate when young, in age convex to plano-convex, broadly convex with depressed center; surface glabrous, not striate, hygrophanous, suede-like ± overall at first, eventually very finely appressed toward the margin, often areolate by maturity, disc at first "avellanous", "tilleul buff", "pinkish cinnamon", "cinnamon", "ochraceous tawny", "light ochraceous buff", "cinnamon buff", light brown (6C5 - 6D5), in age "clay color", golden brown (5D7), grayish orange (5B4/3); margin inrolled, smooth, becoming puckered to striate, with fine floccose veil remnants at margin, white to "pale ochraceous-buff" (4A2). Flesh up to 5 mm thick at disc, white, unchanging; odor none, pleasant to mild, rarely of old cabbage; taste none to mild, agaricoid, unpleasant in throat. Lamellae adnate, adnexed to seceding, subventricose to ventricose, close to crowded, white at first, eventually off white, "pale ochraceous-buff", "tilleul buff", "light ochraceous buff", "pale pinkish cinnamon" (6A2), "pinkish buff" (6B4), yellowish white (4A2); margin entire, or uneven when young, becoming even with age; lamellulae numerous. Stipe 10-35 X 5-10 mm, curved, ± equal to slightly broader below, sometimes with a slightly expanded base; surface fibrillose to scrobiculate, flocculose, squamulose, dry, white to yellowish white (4A2), "cinnamon buff", "pinkish buff", or concolorous with gills, scales "pale pinkish cinnamon", stipe base occasionally with white innate zone, the very base "xanthine orange", "verona brown", "army brown" to "natal brown", "vinaceous russet" to "cameo brown"; stipe flesh white, solid, unchanging; consistency tough. Annulus absent. Rhizomorphs extensive, off white, mellowing to ivory color on exposure. Habitat gregarious on *Quercus* spp. wood and other hardwoods; in forest vegetation.

Pileipellis a repent cutis; hyphae 2-8 µm diam., tightly interwoven, some hyphae erect, with diverticulate growths, pigmented, with clamp connections; wall thin. Pileus trama loosely interwoven, forming lacunae; hyphae 3-16 µm diam., hyaline, constricted at septa, with clamp connections; wall 2-4 µm thick. Lamellar trama regular to

subregular, interwoven toward the edge; hyphae 2-14 µm diam., hyaline, with clamp connections; wall thin to 3 µm thick. Subhymenium ramose, 3-4 cells thick. Basidia 19-26 X 4-7 µm, clavate to sphaero-pedunculate; sterigmata four. Pleurocystidia absent. Cheilocystidia (Fig. 2a,b) 19-40 X 5-14 µm, clavate, ventricose to sphaeropedunculate, some with diverticulae, with clamp connections; apex obtuse. Lamella edge fertile. Stipitipellis parallel; hyphae 2-11 µm diam., hyaline, frequently septate, with clamp connections; walls 1.5-6 µm thick. Caulocystidia (Fig. 2c) 20-44 X 4-8 µm, cylindrical, clavate to irregularly lobed or flexed, uncommon to rare; knobs or small projections common. Basidiospores (Fig. 2d) 4.8-8.0 X 2.4-4.0 µm (A = 6.1 ± 0.59 X 3.3 ± 0.35 µm, Q = 1.30-2.67, Qx = 1.91 ± 0.23, n = 225/12), ellipsoid to subovoid in side view, ellipsoid to sub-cylindrical in front view, hyaline, inamyloid; wall smooth, thin.

Commentary. The description of *L. boryana* by Pegler (1983) is adequate for the American collections listed above, including specimens not vouchered by cultures (see specimens used under Material and methods), except for the presence of cheilocystidia and caulocystidia on our collections (not observed by Pegler). Cheilocystidia were sphaeropedunculate to broadly mitten shaped (Fig. 2a,b), and ranged from scattered to abundant on lamellar edges. Spores of *L. boryana* are more oblong than subcylindrical as summarized from the mean ratio, Q = 1.91.

Lentinula raphanica (Murr.) Mata & R.H. Petersen comb. nov.

≡ *Armillaria raphanica* Murrill. 1943. Mycologia 35: 35:422. [!]

= *Gymnopus alliaceus* Murrill. 1943. Mycologia 35: 425. [!]

Pileus convex at first, then broadly convex to shallowly depressed; surface glabrous to suede like, not striate, often wrinkled inward, hyigrophanous overall; disc at first "russet" to "ochraceous tawny", then "vinaceous cinnamon", "pinkish cinnamon", "cinnamon buff", "pale cinnamon-pink", outwards "pinkish cinnamon", "pinkish buff" to off white, bruising "walnut brown"; velar remnants sometimes as appendiculate scales near margin, more or less zonate; margin strongly inrolled, appendiculate at first, with

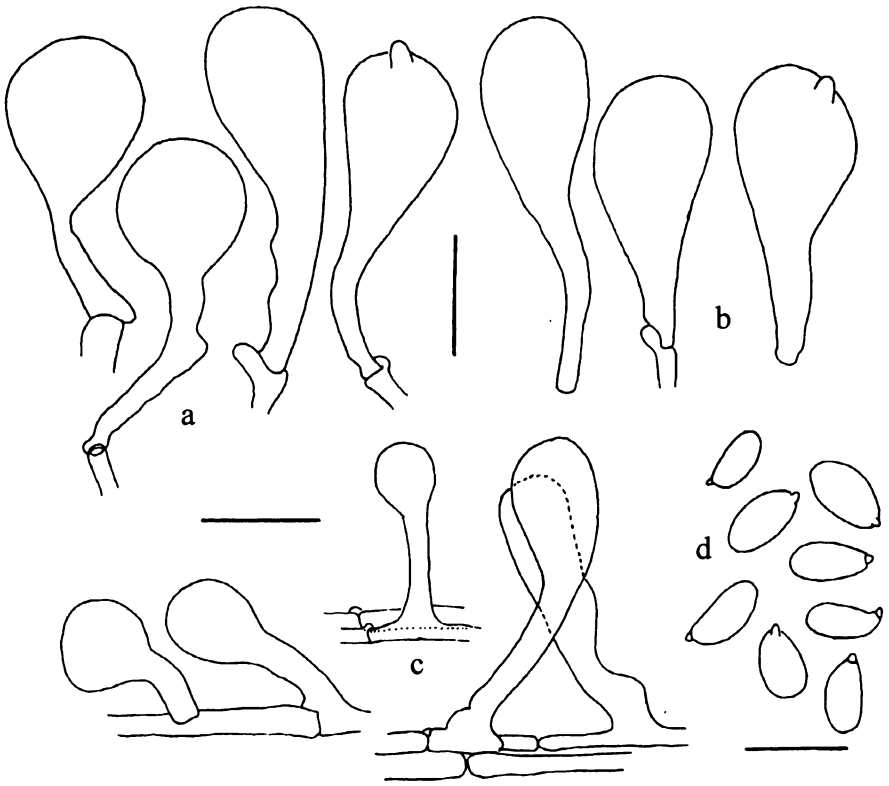

Fig. 2. *Lentinula boryana*. a,b. cheilocystidia. c. caulocystidia. d. basidiospores. Scale bar = 10 μm. FB 7810 = a, c, d; FB 8421 = b.

membranous white veil. Flesh up to 3 mm at disc, off white to "cream-buff"; odor none to mild; taste none at first soon with unpleasant, cabbage-like, aftertaste. Lamellae free to seceding, delicate, close to crowded, narrow to subventricose, thin, "pale pinkish cinnamon", "pale cinnamon-pink", to off-white; edge smooth, eroded in age. Lamellulae numerous. Stipe central to eccentric, equal, sometimes twisted; surface scurfy to shaggy, above with erect floccose fibrillose scales, longitudinally lined, "pinkish buff", "pale cinnamon pink", to off-white towards base, in some bruising brown (7D8) to red-brown (8E6); consistency tough. Annulus absent. Habitat gregarious, on *Quercus* spp wood and other hardwood; in forest vegetation.

Pileipellis repent cutis, rarely embedded in gelatinous matrix; hyphae 1.5-7(10) µm diam., tightly interwoven, occasionally with short, erect or diverticulated outgrowths, pigment-encrusted, sometimes banded, light brown, with clamp connections; wall thin. Pileus trama loosely interwoven; hyphae 3-16 µm diam., hyaline, constricted at septa, with clamp connections; wall up to 1- 5 µm thick. Rarely, oleiferous hyphae present, 3-5 µm diam. Lamellar trama regular to subregular, becoming interwoven towards edge; hyphae 2-12 µm diam., hyaline, with clamp connections; walls thin to 2 µm thick. Subhymenium ramose, 2-4 cells thick. Basidia 16-26 X 4-7 µm, clavate to sphaeropedunculate; sterigmata (2-3)-4, up to 8 µm long. Pleurocystidia absent. Cheilocystidia (Fig. 3a,b) (13)17-28(50) X 3-10 µm, mainly clavate, fusoid, contorted, branched, rarely ampullaceous, with clamp connections; apex obtuse, mucronate, knobbed or with diverticulate outgrowths. Lamella edge sometimes fertile. Stipitipellis parallel; hyphae 3-14 µm diam; walls up to 1-4 µm thick. Caulocystidia (Fig. 3c) 10-35 X 3-7 µm, cylindrical, clavate or flexuous, apex obtuse, knobbed or with outgrowths. Basidiospores (Fig. 3d) 4.8-7.2 X 2.0-3.6 µm (A = 5.7 ± 0.58 X 2.7 ± 0.32 µm, Q = 1.50-3.00, Qx = 2.16 ± 0.28, n = 215/10), narrowly ellipsoid to subovoid in side view, sub-cylindrical in front, hyaline, inamyloid; wall thin, smooth.

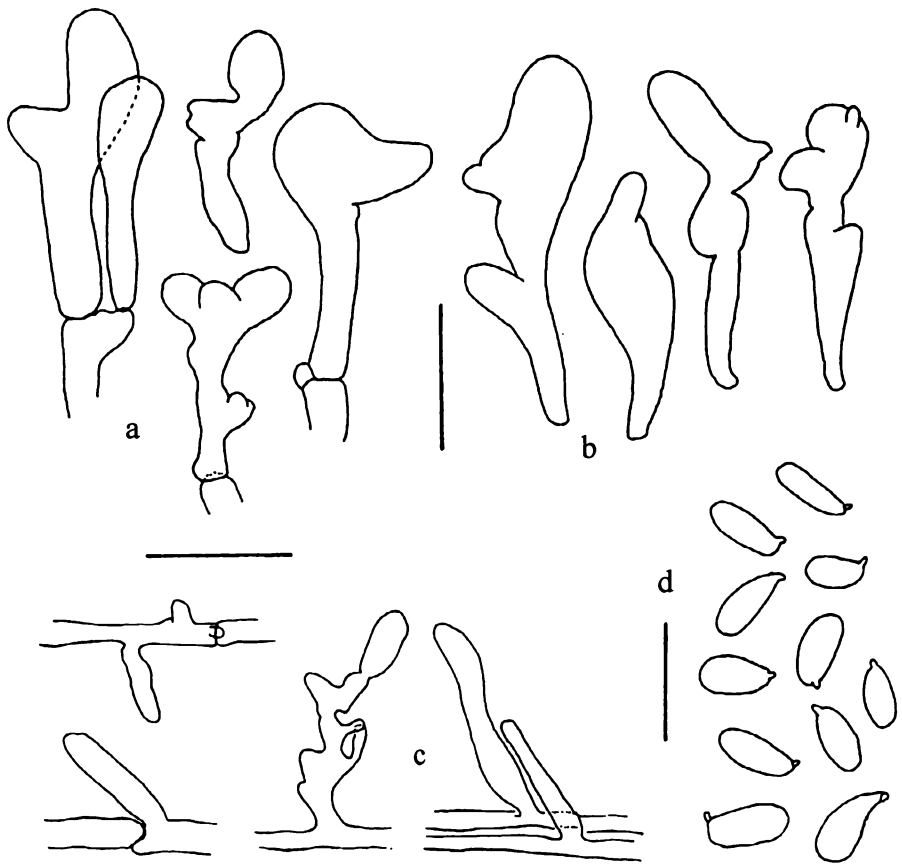

Fig. 3. *Lentinula raphanica*. a,b cheilocystidia. c. caulocystidia. d. basidiospores. Scale bar = 10 μm. FB 9099 = 3a, c, d; FB 9153 = 3b.

Commentary. *Lentinula raphanica* can be distinguished from *L. boryana* by the shape of the cheilocystidia and basidiospore dimensions. Cheilocystidia of *L. raphanica* are clavate and frequently knobbed or diverticulate at the apex and somewhat smaller in size. Spores of *L. raphanica* have a narrower shape, more subcylindrical than oblong, judged by its higher mean ratio of Q = 2.16. Caulocystidia are also different, more clavate and knobbed in *L. raphanica* than in *L. boryana*. Included in this description are basidiomata from USA and Venezuela, not vouchered by cultures but with identical features for this species.

Nomenclature. The assumption that the common American *Lentinula* species can be represented by the same name (Pegler, 1983) has some risk. We have examined the type specimen of *L. boryana* and contrary to findings reported by Pegler (1975, 1983) we found a few cheilocystidia, after examining numerous sections of the basidiomata. We assume that long preservation and exposure of the lamellar edges to friction (paper-on-paper, glue, etc) have extirpated most or all of the cheilocystidia of the type specimen of *L. boryana*. The intersterility group I organisms, however, consistently exhibit inflated, sometimes sphaeropedunculate cheilocystidia and so we have persisted in using *L. boryana* for basidiomata with such features.

In order to secure a name for intersterility group II, it was necessary first to examine type specimens of all synonymous names under *L. boryana* (Pegler, 1983). *Armillaria raphanica* Murrill, gathered near Gainesville, Fla., is the most priorable species epithet for organisms in intersterility group II. The type specimen exhibits clavate, knobbed cheilocystidia and subcylindrical spores, features observed consistently in all basidiomata of extant *Lentinula* from intersterility group II. Thus, it is necessary to transfer the *A. raphanica* species epithet to *Lentinula.* A complete taxonomic study and nomenclature revision on this and other type specimens will be published in another paper (Mata and Petersen, 2001).

Molecular studies. When *G. dryophilus* and *G. subnudus* sequences were used as the outgroup, bootstrapped consensus trees using gaps first as missing data and second as a fifth character identified two major clades, a clade consisting of *L. edodes* isolates and a clade consisting of the New World species, *L. aciculospora, L. boryana,* and *L. raphanica* (results not shown).

While *G. subnudus* could be aligned with *Lentinula* sequences, *G. dryophilus* was more divergent resulting in regions of ambiguous alignment. In order to achieve better alignment, New World sequence data were reanalyzed using *L. edodes* sequences as an outgroup and treating gaps as missing data or as a fifth base. For gaps as missing data, there were 529 constant characters, 58 parsimony-uninformative characters and 244 parsimony-informative characters. There were 225 most parsimonious trees of length = 418 steps, consistency index = 0.88, homoplasy index = 0.12, and retention index = 0.94. For gaps treated as a fifth base, four trees of length = 754 were recovered. There were 392 constant characters, 54 parsimony uninformative characters and 385 parsimony-informative characters. The tree length was 745, consistency index = 0.85, homoplasy index =s 0.15 and the retention index = 0.94. In spite of significant differences in tree length between the two analyses, bootstrapped consensus trees were identical, with *L. aciculospora* basal to both *L. boryana* and *L. raphanica* (Fig. 4). In all analyses, *L. aciculospora*, *L. boryana* and *L. raphanica* formed three discrete well-supported clades, consistent with morphological and mating data.

Discussion

This study has shown that *L. boryana* and *L. raphanica* are widespread taxa occurring in subtropical and tropical regions of North America, the Caribbean, Central America, and South America. The two species are apparently parapatric in Brazil and Venezuela north to Mexico and the Caribbean, but only *L. raphanica* seems to fruit in the Gulf Coast of continental United States. *Lentinula aciculospora* has been collected only in high elevation oak forests in southern Costa Rica (Mata and Petersen, 2000).

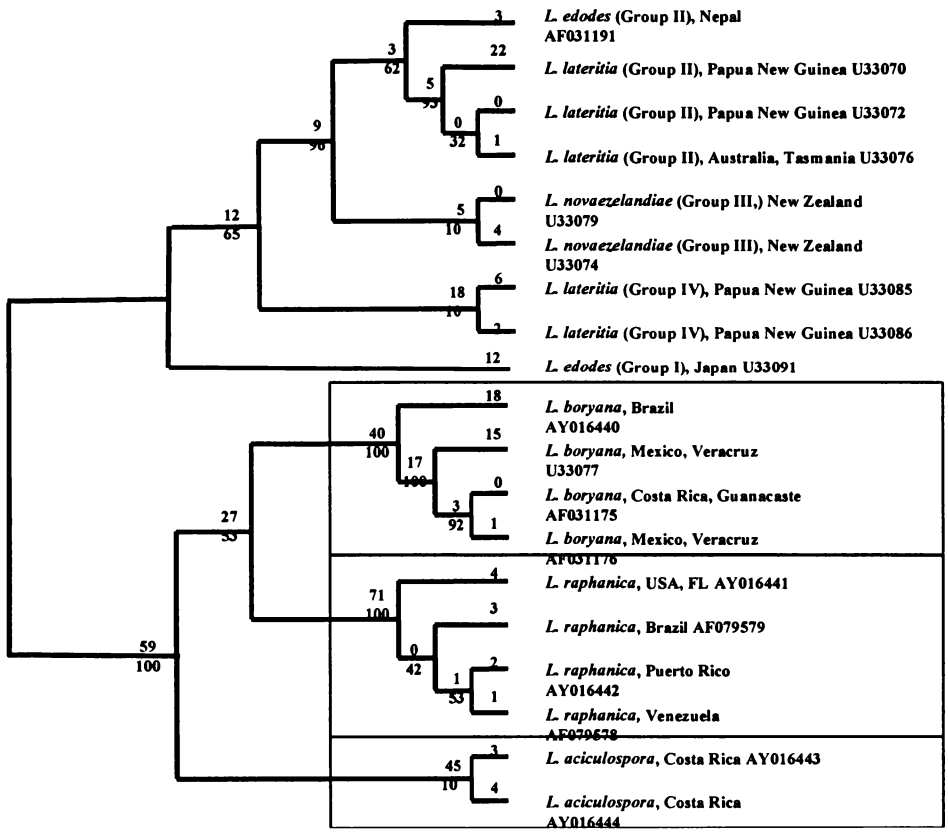

Fig. 4. Bootstrap consensus tree of *Lentinula* collections with gaps treated as missing. A total of 225 most parsimonious trees were obtained with a length of 418. Number of parsimony-informative characters was 244. Consistency index (CI) = 0.8774. Homoplasy index (HI) = 0.1226. The number on top of a line is the number of steps, and the number below a line is the bootstrap value for the node to the right of the number.

The habit and stature of basidiomata of *L. boryana* and *L. raphanica* are similar, but their stature is smaller than that of basidiomata of *L. aciculospora,* which macroscopically more closely resemble morphological variants of *L. edodes* (Mata and Petersen, 2000). Basidiomata of *L. boryana* and *L. raphanica* present pinkish buff to yellowish white colors in their pilei, whereas those of *L. aciculospora* are darker in color and nearer to those morphological variants of *L. edodes* (Mata and Petersen, 2000; Pegler, 1983). All New World *Lentinula* have similar microscopic characters such as an undifferentiated pileus epicutis, a loosely interwoven pileus trama consisting of thin- to thick-walled hyphae, clamp connections in all tissues, and absence of pleurocystidia. The distinctive cylindrical shape of the spores in *L. aciculospora* (Q = 2.97) separates it from *L. raphanica* (Q= 2.16) and *L. boryana* (Q = 1.91). Cheilocystidia in *L. boryana* are broadly clavate to sphaeropedunculate, and are larger than those of *L. aciculospora.* Cheilocystidia of *L. raphanica* are similar in shape and size to those of *L. aciculospora*, but the latter lacks caulocystidia (Mata and Petersen, 2000). Proper analysis of type specimens has elucidated the identity of *Lentinula* taxa on the American continent, and *L. raphanica* is the most priorable name for intersterility group II.

All New World *Lentinula* exhibit a tetrapolar mating system and from a biological species perspective they appear to be reproductively isolated from each other and from the *L. edodes* biological species of the Old World (Mata and Guzmán, 1989b; Petersen et al, 1998). ITS ribosomal DNA sequences of *L. boryana* and *L. raphanica* are mutually exclusive and differ from those of all morphological variants in the *L. edodes* complex (Hibbett, 1992; Hibbett, et al. 1995, 1998; Nicholson et al, 1997; Thon and Royse, 1999).

Previous ITS phylograms show putative *L. boryana* separated into two clades (Hibbett et al, 1998; Thon and Royse, 1999) one represented by voucher collections from Louisiana (USA), Puerto Rico, and Venezuela, now established as *L. raphanica.* *Lentinula boryana* and *L. raphanica* are also separable by nuclear large subunit (LSU) rDNA (Hibbett, 2001), β-tubulin gene sequences (Thon and Royse, 1999) and RFLP profiles of ITS DNA (Petersen et al, 1998), although collections of *L. boryana*, even from

small geographical areas (Nicholson et al., 1997) are heterogeneous for some RFLP patterns.

When ITS sequences of all species of *Lentinula* are compared, the genus can be divided into two major groups, one including *L. boryana*, *L. raphanica*, and *L. aciculospora* and the other including the morphological variants of *L. edodes*. *Lentinula aciculospora* from Costa Rica appears between the two major complexes and is basal to *L. boryana* and *L. raphanica* (Fig. 4). In contrast to conclusions by Nicholson et al (1997), these findings would suggest that *L. boryana* is derived and is not the oldest lineage to diverge within the genus. In a major phylogeny of the Agaricales based on LSU rDNA sequences compiled by Moncalvo et al (2000), one strain of *Lentinula* labeled as *L. edodes* appears nested within several taxa belonging to collybioid Tricholomataceae, supporting the taxonomic placement of the genus by Pegler (1983).

Based on morphology, sexual compatibility, and molecular phylogenetic reconstructions, there appears to be a taxonomic hiatus between the Old World taxon (*L. edodes* and its variants) and the New World entities (Hibbett, 2001). Retention of sexual intercompatibility among widely scattered collections of *L. edodes* in the face of morphological variation and sequence differences reflects the highly conserved nature of the sexual recognition system. In turn, molecular and morphological variations evolve over extended time, leading to the conclusion that Old World *Lentinula* is quite old. Vicariance events and dispersal routes discussed by Hibbett (2001) may help understand why New World species differ in morphology, sexual compatibility and molecular phylogeny.

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PART V

The genera *Gymnopus* and *Rhodocollybia* in the Talamanca Mountains of Costa Rica.

Introduction

Taxonomy.

THE GENUS *COLLYBIA.* The name *Collybia* (*collybus* = coins) was proposed first by Fries (1821) as an infra-generic rank (*tribus*) within the genus *Agaricus*. Fries' (1821) tribe *Collybia* embraced Persoon's (1801) section *Gymnopus* as a synonym. According to Fries (1821) basidiomata in tribe *Collybia* had a fleshy to membranous, convex to plane pileus, soon with a depressed center, glabrous, and dry surface, inrolled margin; lamellae free to attached, never decurrent, unequal, juiceless, entire, white, rarely gray or becoming yellow, and a hollow and smooth stipe, somewhat cartilaginous, slender, equal, often radicating; small stature, on decaying wood or soil, solitary or gregarious.

Later, Fries (1838) prepared a more elaborated concept of tribe *Collybia* (and subtribe *Genuinae*) by dividing it into four sections - *Striaepedes*, *Vestipedes*, *Laevipedes*, and *Tetrophanae* - separated according to stipe surface and color, and spacing of lamellae. Presently, the first three sections constitute the main body of *Gymnopus* (Antonín and Noordeloos, 1993, 1997).

Staude (1857) was the first mycologist to imply the use of *Collybia* at the genus level (Donk, 1949a, 1949b). Because he defined *Collybia* as a genus (= *Gattung* in German) modern mycologists accept Staude (1857) as the authority for the genus *Collybia* (Antonín and Noordeloos, 1997; Antonín et al, 1997; Halling, 1983) as proposed and discussed by Donk (1949a, 1949b, 1962). Gillet (1878) was the first to use *Collybia* explicitly as a genus name and Kummer (1882) made the first explicit combinations in *Collybia*. It was *C. tuberosa* (Bull.: Fr.) Kummer which became designated as the type species for *Collybia* (Donk, 1949a, 1949b, 1962, 1964).

Quélet (1888) presented an improved classification and concept for *Collybia* using substrate and insertion of the stipe as diagnostic characters. Kühner and Romagnesi

(1953) made a significant contribution by further delimiting *Collybia*, segregating section *Tetrophanae* based on dark colors present in the pilei and lamellae.

Mycologists have tried to follow concepts of *Collybia sensu* Fries (1821, 1838) and as a consequence have brought different understandings of this name. Fries' broad concept of *Collybia*, based on few distinctive macroscopic characters, has led mycologists to coin and apply the term "collybioid" to a broad range of mushrooms displaying those macroscopic characters used by Fries (1821, 1838). These rather unspecific macroscopic features are represented in many genera in the family Tricholomataceae, and at the same time basidiomata may present a wide range of phenotypic variation making identification based on relatively few field characters more difficult. Current positive identification of collybioid taxa includes the use of micromorphology, as well as biochemical and stain reactions, and substrate description (Largent, 1986; Lennox, 1979; Petersen, pers. com.). Some collybioid genera similar to or segregated from *Collybia* are mentioned or discussed briefly below.

Basidiomata of the genus *Marasmius* are characterized by having tough consistency and revival properties (i.e. resuming their natural shape and function in moist condition after drying) when compared to those of *Collybia* with more fleshy fruitbodies. Basidiomata representing *Marasmius* are also separated from those of *Collybia s.l.* by the structure of the pileus epicutis and chemical reactions (Antonín and Noordeloos, 1997; Singer, 1986). Basidiomata of the genus *Marasmiellus* are also quite similar to those of *Collybia* due to a very similar pileipellis structure but basidiomata of *Marasmiellus* present an insititious stipe (Antonín et al, 1997). Other noteworthy collybioid genera segregated from *Collybia* are *Crinipellis*, *Flammulina, Baeospora, Strobilurus,* and *Oudemansiella* based on stature and habitat of basidiomata, pileipellis structure, presence of pileocystidia and amyloidity of basidiospores; *Calocybe* and *Lyophyllum* for the presence of basidial siderophilous granulation; *Caulorhiza* and *Xerula* for the presence of a radicating stipe (Antonín and Noordeloos, 1997; Halling, 1983; Kühner and Romagnesi, 1953; Lennox, 1979; Métrod, 1952). The aforementioned genera have been

proposed at different times by different mycologists and with the fundamental comprehensive works done by Singer (1951, 1962, 1986) these have become accepted as independent and valid genera.

Recently, Antonín et al (1997) proposed to restrict the generic circumscription of *Collybia* by defining more stringently its characters. They suggested dividing *Collybia* into three genera, namely *Collybia*, *Gymnopus,* and *Rhodocollybia.* The concept of *Collybia* was reduced to represent those small collybioid mushrooms fruiting on mummified basidiomata of *Russula* and *Lactarius* or with a stipe growing from a sclerotium as represented by its type species, *C. tuberosa* (Antonín and Noordeloos, 1997; Antonín et al, 1997, Hughes et al, 2000). The genus *Gymnopus* became what was typically understood as collybioid mushrooms comprising species mainly from sections *Vestipedes*, *Levipedes*, *Subfumosae,* and *Iocephalae* (*sensu* Singer, 1986). *Rhodocollybia* was proposed for species mainly from section *Striipedes* (*sensu* Singer, 1986) with pinkish colored, pseudoamyloid spores.

THE GENUS *GYMNOPUS.* Persoon (1801) was the first to coin the name *Gymnopus* (*gymno* = naked, *pus* = footed) for which he designated a section within the genus *Agaricus* containing 137 species separated primarily on pileus color. However, Roussel (1806) has been accepted by modern mycologists as the authority of the genus name *Gymnopus* (Antonín and Noordeloos, 1997; Antonín et al, 1997; Donk, 1949b, 1962). In his work, Roussel (1806) listed the species using binomial names preceded by Gymnope [= *Gymnopus*] thus suggesting the application of it as a generic name.

Fries (1815, 1818) initially adopted *Gymnopus* (from Persoon's works) as an infrageneric name under *Agaricus*, but hardly any species placed there by Fries corresponded to those by Persoon (1801). With time, however, Fries (1821, 1838) amended his concept of *Gymnopus*, considering it to be a synonym of tribe *Collybia* in genus *Agaricus*. Gray (1821) was the first mycologist to use *Gymnopus* explicitly at the generic rank, but the recognition of his authority for the use of this name became invalid

based on nomenclature grounds (Donk 1949a, 1949b). It is thought that Gray may have adopted the concept and name of *Gymnopus* from Persoon (Petersen, pers. com.).

The need to bring stability to the use and application of names led Earle (1909) to designate a type species for *Gymnopus* (*G. longipes* ≡ *Ag. longipes*), a choice rejected later because it was not part of Fries' (1821, 1838) original species list under *Collybia* (≡ *Gymnopus*) (Donk, 1949a, 1949b, 1962). Instead, *G. fusipes* (Bull.: Fr.) Gray was proposed as the type species to represent the name *Gymnopus* (Donk, 1949b, 1962).

Ambiguity in the use of binomial names for taxa under *Gymnopus* and *Collybia* prompted agreement over conservation of use of the latter name (Donk, 1962, 1964). In spite of this, a few mycologists like Murrill (1916) believed in the name *Gymnopus* using it extensively for tropical and subtropical taxa, later put by others under synonymy with *Collybia* (or other related taxa). As mentioned in the paragraphs above, the use of *Gymnopus* has gained acceptance with its reintroduction at the genus rank (Antonín and Noordeloos, 1997; Antonín et al, 1997, Desjardin et al, 1997, 1999; Halling, 1996a). The genus *Gymnopus* is divided into sections *Gymnopus*, *Iocephalae, Levipedes,* and *Vestipedes* (Antonín and Noordeloos, 1997; Antonín et al, 1997).

THE GENUS *RHODOCOLLYBIA.* Singer (1939) proposed the genus name *Rhodocollybia* (*rhodo* = rosy) for those species in *Collybia* with a pink spore deposit. The type species designated by Singer was *R.* (= *Collybia*) *maculata* (Alb. & Schw.: Fr.) Kummer. However, soon after, Singer placed *Rhodocollybia* in synonymy under *Collybia* in his subsequent works (Singer 1951, 1962, 1986). Lennox (1979) proposed new combinations of species epithets under *Rhodocollybia* while Halling (1983) used it as a subgeneric name under *Collybia* to accommodate those species with pinkish spore deposit, some with dextrinoid spore reaction in Melzer's reagent and thickened spore walls.

LENTINULA: A CLOSE RELATIVE TO *GYMNOPUS* AND *RHODOCOLLYBIA.* Earle (1909)

proposed the genus name *Lentinula* with *Lentinula cubensis* Berk. & Curt. as the type specimen. However, acceptance of this generic name was not immediate as several authors considered it to be a synonym under *Lentinus* (i.e. Singer, 1986). Pegler's (1975) classification proposal of *Lentinus* considered that some species should be placed under *Lentinula* because its hyphal structure was monomitic (as opposed to dimitic in *Lentinus*).

Pegler (1983a) monographed the genus *Lentinula* and placed it in family Tricholomataceae, tribe *Collybieae* Imai, suggesting a taxonomic relationship with *Collybia s.l.* (i.e. *Gymnopus* and *Rhodocollybia*). Current molecular and genetic studies in *Lentinula* support Pegler's (1983a) taxonomic treatment (Hibbett and Vilgalys, 1991, 1993; Mata et al, 2001; Petersen et al, 1998). Macroscopically, basidiomata of *Lentinula* are more robust than most *Gymnopus* and *Rhodocollybia*, but share similarity in microscopic elements such as spores, cystidia, and hyphal structure and arrangement (pers. obs.). Additionally, molecular data show *Lentinula* to form a monophyletic group, phylogenetically very close to *Gymnopus* and *Rhodocollybia* (Hibbett and Vilgalys, 1993; Nicholson et al, 1997; Mata et al, 2001; Moncalvo et al, 2000, 2002).

Taxonomic characteristics. For the purpose of this dissertation, special importance was given to certain microscopic characters such as hyphal orientation of the pileipellis, spore wall color reaction to Melzer's reagent, and presence or absence of cystidia, since all these are relevant for distinguishing species and delimiting sections in the genera (Halling, 1983; Antonín and Noordeloos, 1993, 1997).

PILEUS EPICUTIS. Below, pileus epicutis is referred to as the tissue that forms the uppermost surface of the pileus and that can be distinguished as such from hyphae of the pileus trama. Two basic types of hyphal structure are recognized for this study. The first one is a pileus epicutis composed of hyphae that are not radially arranged, as seen in a radial section, and that are frequently branched, i.e. not cylindrical. This kind of hyphal structure is commonly referred to as a "dryophila type" cutis, characteristic of *G. dryophilus*.

The second kind of pileus epicutis is a simple cutis, composed of hyphae that are cylindrical, not branched, and radially oriented. Hyphae in this kind of cutis can either present lateral knobs or projections, referred to as diverticula, or not. In the simple cutis construction, terminal cells are referred to those uppermost hyphae that may extend upwards from the usually repent disposition of the rest. In the case of *Gymnopus*, presence or absence of diverticula in hyphae of the pileus epicutis has been used by some mycologists (Halling, 1983; Pegler, 1983b; Singer, 1986) to place taxa in two different sections, namely sect. *Subfumosae* (with diverticula) and sect. *Vestipedes* (without diverticula). In this work, the concepts of Antonín & Noordeloos (1997) are followed in the sense that sect. *Subfumosae* is incorporated into sect. *Vestipedes* to accommodate species represented by basidiomata with wide variation in pileus epicutis structure observed in the genus (Antonín et al, 1997).

Hyphae of the pileus epicutis in *Gymnopus* and *Rhodocollybia* are typically inamyloid in presence of Melzer's reagent, meaning they remain hyaline or become yellow (Largent et al, 1977; Singer, 1986). In the course of microscopic examination, in some cases, however, hyphae that were pigment-incrusted became pale orange to orange-brown colored, differently colored as defined for a typical dextrinoid reaction, i.e brown, reddish brown to wine red (Largent et al, 1977; Singer, 1986).

CYSTIDIA. Cheilocystidia and pleurocystidia are sterile elements, the first ones located at the lamellar margin and the latter along the lamellar sides, and both are morphologically distinct from basidia and basidioles. Most species in *Gymnopus* and *Rhodocollybia* present cheilocystidia of a wide range of shapes and sizes. While pleurocystidia were easier to detect in a cross section, cheilocystidia in several cases were not evident at first hand and light pressure on section mounts was needed in order to distinguish their contours and dimensions. The term caulocysidia has been applied to terminal cells of hyphae in the stipe epicutis, that clearly could be identified as such. In

some cases caulocystidia will be part of an entangled mat of hyphae, forming fascicles or not.

BASIDIOSPORES. The color of basidiospores in mass in species of *Rhodocollybia* is mostly cream, whereas in those of *Gymnopus* is mostly white. Basidiospores of *Rhodocollybia* and *Gymnopus* are hyaline (as observed in water or alkaline mounts) but those of *Rhodocollybia* sometimes will stain pale brown to reddish brown in presence of Melzer's reagent and stain dark blue in presence of cotton blue (Halling, 1983; Largent et al, 1977; Lennox, 1979; pers. obs.). The latter color changes are referred to as dextrinoid and cyanophilous respectively, while the lack of color change is described as inamyloid and acyanophilous. Basidiospore size and shape present a broad range in both genera.

Species concepts. Species, as a nomenclatural rank, is the fundamental unit in classification and systematics (Judd et al, 1999; Petersen, 1976). Ideally, species should be readily recognizable and as such, morphologically distinctive characters have been essential to distinguish them in practice (Judd et al, 1999). Humans detect organisms with their senses, especially sight, so sensory characters have been the primary evidence used in species definitions (Mayr, 1996). Morphology is also the most accessible source of data about evolutionary relationships (a goal in systematics) and is the only basis for the recognition of most species today, of which the Agaricales are an example (Singer, 1986). However, modern data from other sources such as molecular, ecological, cytological, and genetic analyses are becoming more important tools applied for species demarcations (Judd et al, 1999).

Species concepts are the philosophical principles through which researchers communicate their notion of the term species (Mayr, 1996; Petersen and Hughes, 1999). The idea that species are genetically distinct and reproductively isolated from each other is implied in most species concepts (Mayr, 1996). Different species concepts have been elaborated for different kinds of organisms (i.e. sexually vs. asexually reproductive) and as many as 22 species concepts have been postulated up to date (Mayden, 1997). The

main species concepts tested and applied in fungal taxonomy and systematics are the morphological (MSC), biological (BSC), and phylogenetic species (PSC) concepts.

The MSC is the oldest species concept in use and is still the basis for modern taxonomic treatments (i.e. Singer, 1986). In the case of Agaricales, species are defined using sensory characters of basidiomata. The object identified through such characters is referred to as a morphospecies. The MSC is also known as a typological species, because morphological traits of the type specimen form the basis for comparison to other basidiomata (Mayr, 1996). In many cases use of the MSC is insufficient to account for morphological variation in taxa observed in nature, or on the contrary when morphologically similar populations constitute a biological entity (i.e. biological species). The BSC is a concept that emerged as an alternative to the MSC in an attempt to order and relate taxa in a more natural way. The BSC states that a species is a group of actually or potentially interbreeding natural populations that are reproductively isolated from other such groups (Mayr, 1942, 1996). In Agaricales, the BSC is tested principally on the occurrence of heterothallic mating systems and when possible, compared to the MSC (Petersen and Hughes, 1999). A negative aspect of the BSC is that it is not applicable to asexually reproducing organisms (i.e. imperfect fungi) where other criteria may apply (Brasier, 1997). The PSC has been proposed as an alternative to the BSC and MSC to cope with their inherent problems, such as exclusion of asexually reproducing organisms and plasticity of morphological characters. The PSC stipulates that a single lineage of ancestral-descendant populations will remain distinct from other such lineages showing its own evolutionary tendencies and historical fate (Judd, 1999; Mayden, 1997). The PSC can prove to be valuable in resolving relationships for certain sexual and asexual fungi when the BSC or MSC cannot provide strong resolution (Brasier, 1997).

SPECIES CONCEPTS IN *GYMNOPUS, RHODOCOLLYBIA* AND *LENTINULA.* Most of the studies with these genera, including *Collybia s.s.*, have been strictly based on morphological analyses and therefore would fall into the MSC (Antonín and Noordeloos, 1997; Halling, 1983; Lennox, 1979; Noordeloos, 1995; Singer, 1986). Phylogenetic

analysis supported by morphology studies led to the proposal of a new genus related to *Collybia s.s.* (Hughes et al, 2000). Studies testing the MSC and the BSC led to the proposal of new species in the *G. dryophilus* complex (Vilgalys and Miller 1983, 1987a, 1987b). Vilgalys (1986, 1991) reported on phenetic and phylogenetic studies with species in *Gymnopus* sect. *Levipedes* in an attempt to gain insight on species concepts and speciation processes.

Petersen (1995a) compiled a list of the mating systems of several taxa in the genus *Collybia s.l.* and concluded that, with the exception of *G. subnudus* reported as unifactorial by Murphy and Miller (1993), all taxa exhibited bifactorial mating systems. Recently, however, there has been a second report of a unifactorial mating system for the genus, in *C. omphalodes* (Mata and Petersen, 1999). Gordon and Petersen (1991) proposed that different mating systems may be represented within one genus, as in *Marasmius* (Tricholomataceae) where mating systems appeared to be consistent at some infrageneric rank, and could be used as a helpful criterion to establish evolutionary relationships (Petersen, 1995a). Within *Gymnopus*, mating studies with the *G. dryophilus* species complex have led to the proposal of new species, *G. brunneola* in North America, and *G. alpinus* in Europe (Vilgalys and Miller, 1983, 1987b). In a complimentary study, *G. dryophilus* from Europe and North America proved to be fully sexually intercompatible in spite of morphological variation of basidiomata (Vilgalys, 1991).

In a population biology study, Murphy (1997) found two intersterility groups within the morphospecies *G. subnudus* occurring in the Appalachian Mountains, morphologically very similar but associated with different leaf litter types. In a population structure study Marcais et al (1998) used somatic incompatibility and DNA amplification to demonstrate that population structure of *G. fusipes* from two oak forests stands in France was heterogeneous and composed of many genets.

Vilgalys (1986) demonstrated the monophyly of the *G. dryophilus* complex with a cladistic and phenetic analysis of *Gymnopus* sect. *Levipedes* and proposed that the

species in that section were more closely related to those of section *Striipedes* (i.e. *G. fusipes*) than to those of sections *Vestipedes* and *Maculatae* (i.e. *Rhodocollybia*). Ongoing biochemical research (Villaruel-Ordaz and Cifuentes, 1999) is corroborating the partitioning of *Collybia s.l.* into *Collybia s.s.*, *Gymnopus*, and *Rhodocollybia*. Separation of *Collybia s.s.* into two genera has been proposed by Hughes et al (2000) after a phylogenetic study based on sequences from ribosomal ITS and large subunit regions was conducted. Other results from molecular analyses are improving resolution of taxa and their placement in major phylogenies (Moncalvo et al, 2000, 2002).

According to Pegler (1983a) *Lentinula* included five morphospecies, from which *L. boryana* (Berk. & Mont.) Pegler and *L. guarapiensis* (Speg.) Pegler were the only species reported to occur in the New World. Recently, however, Petersen et al (1998) demonstrated the presence of two intersterility groups (i.e. biological species) for putative tropical and subtropical basidiomata of *L. boryana*. The two intersterility groups could also be distinguished by micromorphological characteristics such as cheilocystidia and spores. These findings led to examination of all type specimens of species epithets listed as synonyms under *L. boryana* (Pegler, 1983a) so that correct names could be assigned to each of the biological species. As a result, the names *L. boryana* and *L. raphanica* were assigned to specimens in each of the two intersterility groups (Mata and Petersen, 2001a; Mata et al, 2001). Simultaneously, a third species, *L. aciculospora*, known only from Costa Rica, was discovered based on morphology and mating studies (Mata and Petersen, 2000).

Nicholson et al (1997) provided a phylogenetic analysis based on ribosomal DNA restriction fragment polymorphisms for *Lentinula*, postulating *L. boryana* to be the earliest divergent lineage, and supporting current classification of the genus based on the MSC. Hibbett et al (1995, 1998) proposed at least five phylogenetic species for the *L. edodes* complex based on ITS sequence analysis, and recently explained geographic distribution of these mushrooms in the American Continent using molecular clock dating (Hibbett, 2001).

Ecology and importance. Mushrooms of the genera *Gymnopus* and *Rhodocollybia* are typically saprobes on leaf litter, woody debris and/or soil, or associated with a specific substrate (Antonín and Noordeloos, 1993, 1997; Halling, 1983). Mushrooms in *Lentinula* are lignicolous and reportedly have been associated with plants in the Fagales (Pegler, 1975, 1983a). Like other members of related genera in the Tricholomataceae, these fungi are considered to be important in the recycling of nutrients in forest ecosystems and therefore ubiquitous in many different geographical areas (Murphy, 1997; Singer, 1986).

Except for the commercially grown shiitake, *L. edodes*, most of these mushrooms do not have a major economic value. *Gymnopus confluens* has been reported to contain an alkaloid of pharmaceutical importance of undetermined value and *G. peronatus* has been reported to contain muscarine and epimuscarine (Singer, 1986). *Gymnopus fusipes* is associated with root rots of oak trees (Marcais et al, 1998).

Generalities of the Talamanca Mountains. The Talamanca Mountain Range starts SW of the Irazú Volcano and ends at the Chiriquí Volcano, in Panamá (Fig. 1). This mountain range is the longest orographic system in Costa Rica, extending 320 km in length and distributed among four political provinces. The mountains run along a NW-SE axis, their Pacific flanks descending abruptly in contrast to the gradual sloping towards the Caribbean basin (Gómez, 1986; Kappelle, 1996). There are ten massifs reaching above 2950 m in elevation, and most of the oak-dominated forest habitat is located from 1800 to 3400 m in elevation. An extensive system of protected areas including forest reserves, national parks, indigenous reserves, and other protected zones has been set aside along these mountains.

The climate of the oak forests in the Talamanca mountains is very humid, ranging from temperate to cold, with a moderate water deficit on the Pacific slopes (Coen, 1983; Herrera, 1985). There is a short dry period from December to January, and a rainy period

of almost eight months. Mean annual temperature at about 2000 m is 18 °C and the mean annual precipitation is between 2500-3000 mm; both temperature and rainfall decrease with altitude (Herrera, 1985; Kappelle, 1996).

The actual formation of the Costa Rican mainland started with the upheaval of the chain of Mesoamerican islands (Occidental Archipelago) during the Mesozoic (upper Jurassic). During the Superior Miocene there was lifting of the Mesoamerican isthmus that gave birth to the Talamanca Mountain Range. The rocks present in this Cordillera are composed mainly of Tertiary marine and volcanic sediments. The closure of the Costa Rica-Panamá isthmus was completed at about 3 MYA (Castillo-Muñoz, 1983; Rich and Rich, 1983).

Oak forests. The geological process of forming a 'land bridge' between the northern and southern continental masses provided an opportunity for the migration, establishment and exchange of biota (Hooghiemstra and Cleef, 1995; Mueller and Halling, 1995; Rich and Rich, 1983). It has been documented that high biodiversity in Costa Rica is supported, in part, by a wide range of environments (Kappelle, 1996). These environments are a result of marked precipitation patterns, complex topography, diverse soil structure and composition, and recent ecological and geological events from the Quaternary Period (Kappelle, 1996). Among the most important floristic elements associated to the Holarctic region is the genus *Quercus* (Fagaceae). Oaks have migrated southward through the Central American Isthmus, reaching northern Colombia about 330,000 years ago (Hooghiemstra and Cleef, 1995). *Quercus* trees are predominant components of the forest of the Talamanca Mountains, where at least seven species have been reported occurring above 2000 m. (Kappelle, 1996).

Agaric fungi are important elements in the balance and function of oak forests, because *Quercus* trees are obligate symbionts with them (Trappe, 1962). These fungal associations, called ectomycorrhizae, together with saprobic agarics are vital in the recycling of nutrients and decomposition of organic matter (Alexopoulous et al, 1996;

Trappe, 1962). Many described agarics from Costa Rican forests match species circumscriptions of temperate taxa, indicating that migration of fungal taxa probably took place along with their hosts (Halling, 1996b; Halling and Ovrebo, 1987; Mueller, 1996; Tulloss et al, 1992). However, recent analysis suggests that there may be high levels of endemism for certain ectomycorrhizal taxa in neotropical montane oak forests (Mueller and Halling, 1995), providing an alternative view to the idea of long-distance viable basidiospore dispersal and favoring concepts of speciation through geographic and reproductive isolation.

Objectives. The main objective of this study has been to determine the presence of species of *Lentinula, Gymnopus,* and *Rhodocollybia* in the Talamanca Mountains of Costa Rica.

The following were specific objectives during the research:

1. To determine the presence of taxa of *Lentinula*, *Gymnopus,* and *Rhodocollybia,* known from other parts of the world, in the Talamanca Mountains. For this, species will be described principally by morphological characteristics, and when possible, supported by genetic and molecular analyses.

2. To correctly provide nomenclatural combinations in *Gymnopus* and in *Rhodocollybia* for appropriate taxa currently placed in *Collybia*. Nomenclatural status for previously described species names is reviewed, and when possible, type specimen material has been consulted. New species are described following taxonomic and nomenclatural guidelines.

3. To test if phylogenetic relationships obtained from ITS DNA molecular data are consistent with the current taxonomic placement, at a sectional level.

4. To determine if *Lentinula* is phylogenetically more closely related to *Gymnopus* than to *Rhodocollybia.*

5. To produce an identification key for species of *Rhodocollybia* and *Gymnopus* in the Talamanca Mountain range.

Materials and methods

Field collections. Field trips to the Talamanca Mountains took place in the years 1998, 1999 and 2000. Areas where material was collected for this research, including those visited by other mycologists in other years, are shown in Fig. 1. All collecting sites are accessible by automobile in a one-day roundtrip from nearby lodging facilities that provide adequate installations for proper processing of specimens. Therefore, remote locations, such as those on the Caribbean slopes of the Talamanca Mountains, lacking these basic conditions, or where access is more limited either because of poor roads or complicated travel logistics, or heavy deforestation impeding good collecting, were not visited. Most of the collections from the Talamanca Mountains are from the NW and SW portions on the Pacific watershed.

Morphology analysis. All specimens were processed according to customary field collection and laboratory techniques as described by Largent (1986) and Largent et al (1977). Colors of basidiomata were matched with those of Kornerup and Wanscher (1978) and different color codes, used by other collectors, were converted to Kornerup and Wanscher colors when possible. All color pictures are from field collections made by the author or by Roy E. Halling (REH numbers). All collected material was air-dried, boxed, data-based, in TENN for subsequent microscopic study. Duplicates were deposited with USJO (University of Costa Rica).

Light microscopy, with a Leitz Laborlux microscope, was used for the study of micromorphology of basidiomata. Sections of different parts of the basidiomata were first placed in water mixed with 190 proof ethanol and then mounted in 3-5% aqueous

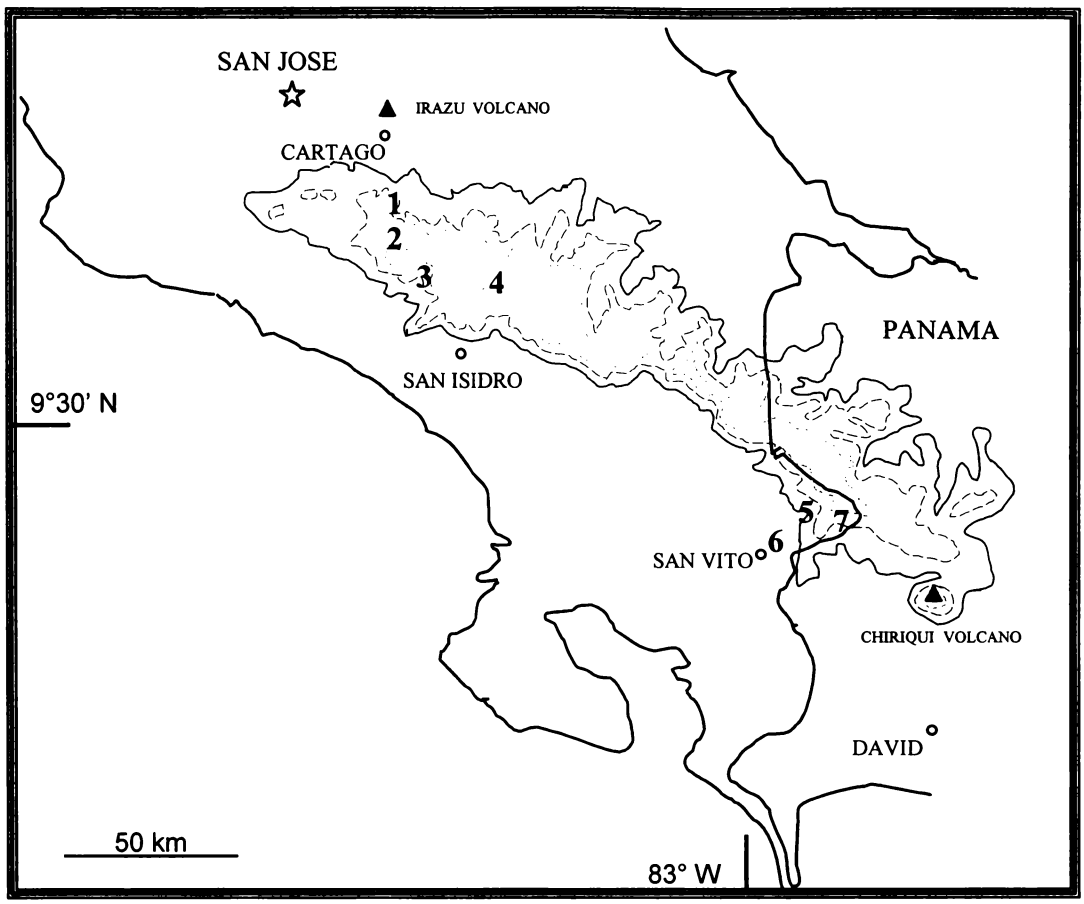


Fig. 1. Orography of the Talamanca Mountain Range and most visited collecting sites.

1. Estrella & Palo Verde; 2. Jardín de Dota, La Chonta & Finca El Jaular; 3. San Gerardo de Dota; 4. Villa Mills & Cuericí Biological Station; 5. Wilson Botanical Gardens; 6. Las Alturas/OTS; 7. Hacienda La Amistad.

Contour lines: — 1000 m ------1500 m ------ 2000 m ····· 2500 m 3000 m

solution of KOH for measurement and drawing of microstructures. All measurements and drawings were done using an oil immersion objective at 1250 X magnification. Drawings were done with the aid of a drawing tube. Melzer's reagent was used to test for amyloid reactions in tissues and spores, and Congo Red and phloxine were used to stain structures for better contrast when needed.

All basidiospore measurements for each species description include a first set of numbers corresponding to the minimum to maximum range of length X minimum to maximum range of width in µm (micrometers), followed by the number of spores per collection measured, the mean of spore length X spore width (x), range of spore quotients (Q), this being spore length divided by spore width, and average quotient (Qx).

Morphological descriptions of species are ordered according to species epithets, and in the case of *Gymnopus* also into sections. A number assigned sequentially within genera precedes each species.

HERBARIUM AND TYPE MATERIAL. Herbarium specimens were required for morphological comparison, and in some cases for DNA extraction. Loans from herbaria included The New York Botanical Garden (NY), Field Museum of Natural History, Chicago (F), Instituto Spegazzini, La Plata, Argentina (LPS), Museum National D'Histoire Naturelle, Paris, France (PC), Farlow Herbarium, Harvard University (FH), Royal Botanic Gardens Kew, England (K), Instituto de Biodiversidad, Costa Rica (INB), New York State Museum, Albany (NYS), and National Museum in Prague, Czechoslovakia (PRM). All herbarium material was annotated.

ABBREVIATIONS. Notes and cultures vouchered by specimens deposited in TENN are represented by TFB (Tennessee Field Book) numbers. Specimens from other herbaria are represented with different acronyms; the most common ones are REH (Roy E. Halling) and EFM (Ana E. Franco-Molano). Holotype is abbreviated as HT, isotype as IT, and paratype as PT. Melzer's reagent is abbreviated as IKI (iodine/potassium iodide).

An S follows specimens from which DNA was extracted, sequenced, and used in molecular analysis. Single basidiospore isolate is abbreviated as SBI.

Genetic studies.

CULTURES. Spore deposits were obtained from basidiomata collected in the field, either directly on 2% malt extract agar (MEA) or on aluminum foil as described by Gordon and Petersen (1991) and Petersen and Krisai-Greilhuber (1996). Single basidiospores germinating on MEA plates were isolated and inoculated to fresh MEA plates. In the laboratory, spore prints on aluminum foil were diluted at different concentrations in sterile water, and inoculated onto fresh MEA plates. When basidiospores germinated, the isolation procedure followed that mentioned above. In addition to obtaining monokaryon isolates, a dikaryon culture for each collection was established. All cultures were stored in sterile water cryovials (Burdsall and Dorworth, 1994) and accessioned in the CULTENN database.

MATING STUDIES. Whenever possible the mating system for each taxon was determined. This was done when at least 12 single basidiospore isolates (SBIs) were paired in all possible combinations (Petersen and Krisai-Greilhuber, 1996); in some cases this procedure was attempted with a smaller number of SBIs. From such self-crosses, tester strains were selected for future recognition crosses. When there was suspicion of cryptic species, or to confirm a biological species (intersterility group), intercollection pairings were performed. This was done by pairing at least four SBIs of each collection once. When readings were not clear this procedure was repeated using up to eight or 12 SBIs. Compatibility of SBIs was determined by the production of clamp connections, and incompatibility by their absence. When there was doubt about compatibility results, mycelium stained with DAPI was observed under epifluorescence microscopy to ascertain nuclear condition (Petersen, pers. com.).

Molecular studies. A total of 26 sequences from Costa Rican specimens and 16 from specimens collected in North America or Europe representing species placed in *Rhodocollybia* and *Gymnopus* were used for DNA extraction and sequencing. Non Costa Rican sequences were generated based on availability of monokaryon cultures (see below) and were intended to give better support and credibility to the phylogenetic analysis.

DNA EXTRACTION AND SEQUENCING. DNA for direct sequencing was extracted from sections of basidiomata preserved in sodium chloride-hexadecyltrimethylammonium bromide (NaCl-CTAB) or from monokaryon cultures grown in potato dextrose broth following standard protocols (Hughes, 1998, Hughes et al, 1999; White et al, 1990). When the extraction product included excess amounts of polysaccharides or other impurities it was cleaned using the GENECLEAN III Kit (Bio 101, 1999/2000).

The nuclear ribosomal internal transcribed spacer ITS 1-5.8 S-ITS 2 (ITS) region was amplified using forward primers ITS 5 or ITS 1F and reverse primers ITS 4 or ITS 4B (White et al, 1990; Gardes and Bruns, 1993). The polymerase chain reaction (PCR) protocols were: initial melting cycle of 94 ° C for 4 minutes, 35 repetitions of a three step amplification cycle of 94 ° C for 1 minute, and 52 ° C for 1 minute, ending with an extension cycle of 72 ° for 1 minute. PCR products were directly purified using Amicon® Microcon®-PCR Centrifugal Filter Devices (Millipore, 2000). Sequencing reaction protocols with primers ITS 5 and ITS 4 for the purified product were: 25 repetitions of a three step amplification cycle of 96 °C for 0.1 min, 50 °C for 0.05 min and 60 °C for 4 min, ending with an extension cycle of 4 °C. This product was cleaned with isopropanol or ethanol/sodium acetate (K. Hughes, pers. comm.) or with the Centri-Sep™ Protocol (Sambrook et al, 1989). The cleaned product was sequenced using an automated ABI 373 DNA sequencer (ABI Prism Dye Terminator cycle sequencing, Perkin-Elmer, Inc.).

Sequences were manually corrected and aligned using the SeqLab program in the Genetics Computer Group package (GCG, 2001). Sequences within and across genera were very different and many gaps were needed to generate the best possible alignment, resulting in a dataset of 1069 characters. Automatic alignment with the program CLUSTALX (Thompson et al, 1997) produced a similar matrix. All sequences were deposited in GenBank (AF505747-AF505788). The aligned sequences used for this dissertation appear are shown in Appendix A.

Five sequences representing four morphospecies in *Lentinula* and one sequence representing *G. dryophilus* were downloaded from GenBank and added to the data matrix.

MOLECULAR DATA ANALYSIS. The aligned ITS nrDNA sequences were analyzed using the software package for phylogenetic analysis using parsimony, PAUP 4b, for the construction of phylogenetic trees according to procedures and criteria by Swofford et al (1996). The function BestFit in the program SeqLab (GCG, 2001) was used to compare percentage of similarity between two sequences representing the same morphological species. The percentage of similarity is represented by an optimal alignment of the best segment of similarity between two sequences and is found by increasing gaps to maximize number of matches (GCG, 2001).

The settings for the neighbor-joining analysis included distance as the criterion. Distance measure used was the Kimura 2-parameter. Ties were broken systematically, if encountered. Bootstrap replicates used were 1000.

The settings for the heuristic search included maximum parsimony as the optimal criterion. All 1232 characters had equal weight, from which 512 were constant, 210 were parsimony-uninformative, and 510 were parsimony-informative. Gaps were treated as missing and multistate characters were interpreted as uncertainty. Starting trees were obtained via stepwise addition and one tree was held at each step, addition sequence was

random, with 100 replicates. Branch-swapping algorithm was tree-bisection-reconnection (TBR) with steepest descent option not in effect. Initial 'MaxTrees' setting was 100. Polytomies were created if maximum branch length was zero, 'MulTrees' option was in effect, and topological constraints were not enforced. Bootstrap replicates used were 1000. The sequence representing *L. edodes* was used to root the phylogenetic trees.

Results

Molecular analyses. The result of the neighbor-joining analysis is shown in Fig. 2 and those of parsimony in Fig. 3 and Fig. 4. The topology of the neighbor-joining tree (distance as optimality criterion) is very similar to those generated from the parsimony analysis. Parsimony and neighbor-joining analyses were performed with matrices containing only sequences representing *Gymnopus* or *Rhodocollybia*, or both combined without *Lentinula*, but relationships among species within the phylogenetic tree and robustness of data were the same as those shown in Figs. 2-4. A different matrix of sequences was created by omitting regions with ambiguous alignment to perform parsimony and distance based analyses. The outcome was very similar to those of the original dataset, including low values for consistency, homoplasy, and retention indices and similar relationships between species.

All trees show species in *Rhodocollybia* to be more closely related to species in *Gymnopus* than to those representing *Lentinula*. Phylogenetic relationships among species in *Lentinula* are discussed in detail in Chapter 4 of this dissertation. Morphological species placed in *Rhodocollybia* appear as a weakly supported clade (Figs. 2, 4). The best-supported node within this genus is that including *R. pandipes* and *R. butyracea*. Two other clades are formed witin *Rhodocollybia*, one including *R. maculata*, the type species of the genus, and the other with newly proposed species in the genus. Of particular interest is the placement of *R. amica* and *R. tablensis* within *Rhodocollybia*, despite the absence of dextrinoid spores, a micromorphological characteristic used to distinguish *Rhodocollybia* from *Gymnopus*. Both *R. tablensis* and *R. amica* appear,

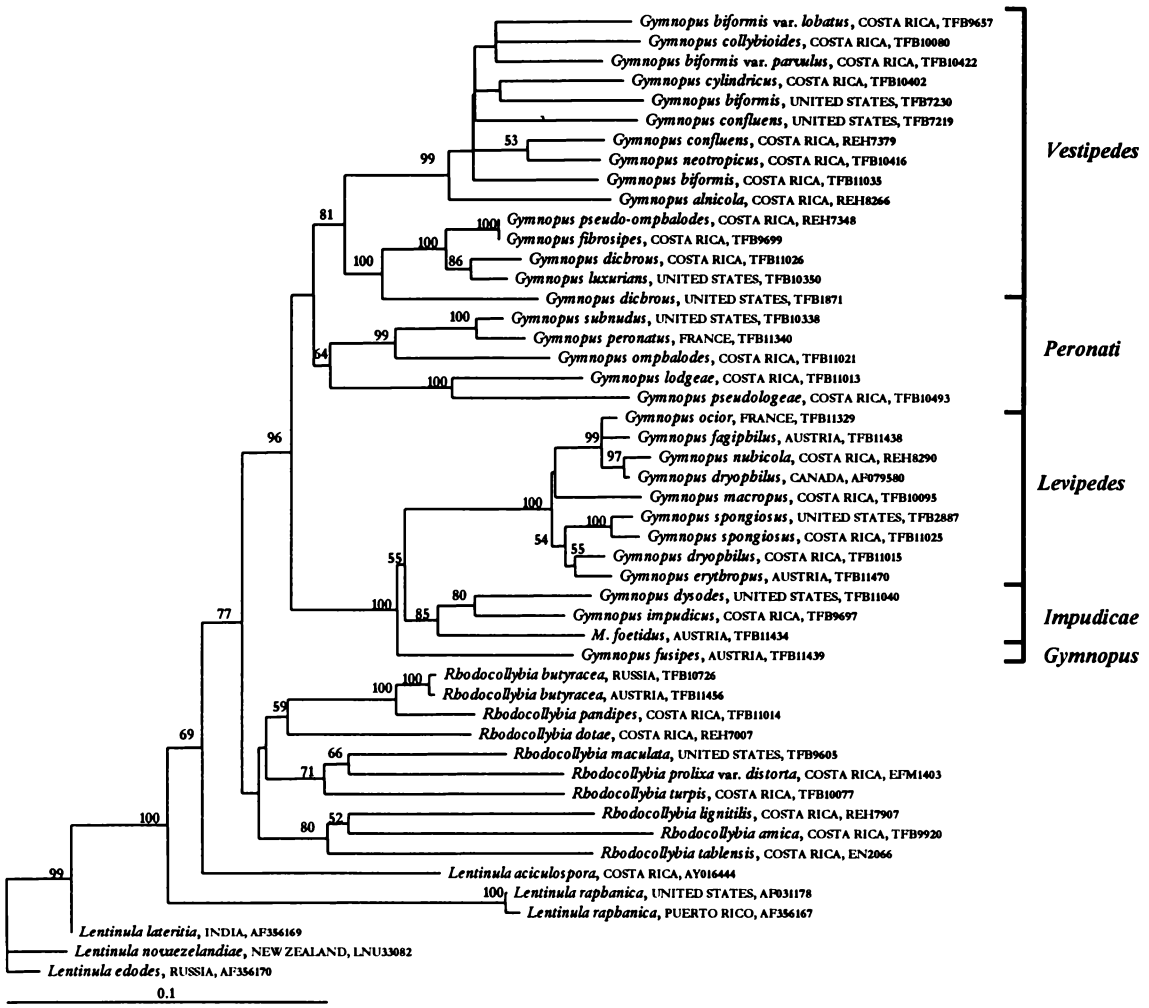


Fig. 2. Neighbor-joining tree based on nrDNA ITS1-5.8S-ITS2 sequences of selected species of *Lentinula*, *Rhodocollybia*, and *Gymnopus*. Analysis uses the Kimura 2-parameter distance measure. Numbers at nodes represent bootstrap values over 50% from 1000 replicates. Scale bar represents substitutions/site. Names of clades represent sections in *Gymnopus*.

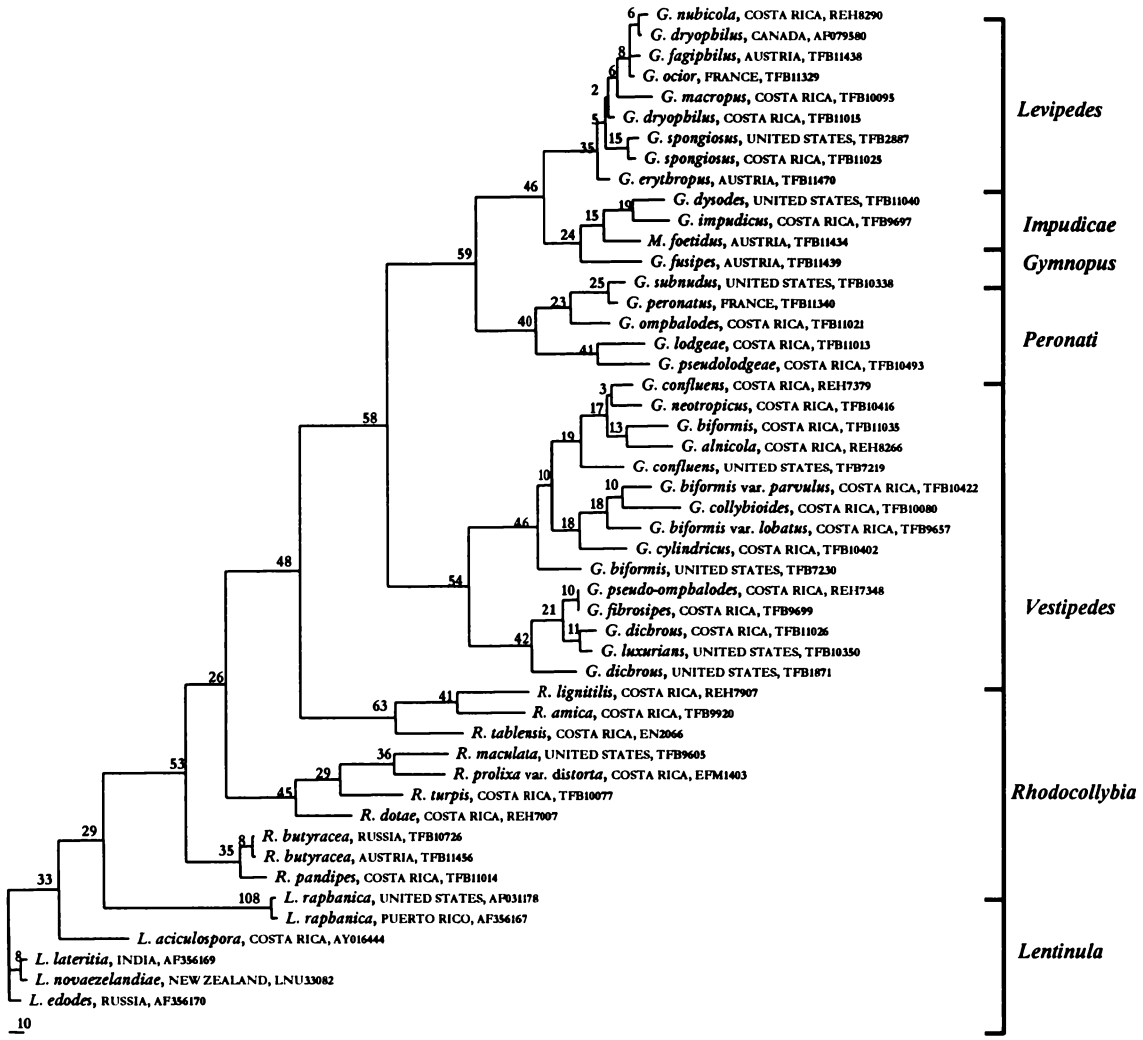


Fig. 3. One of twelve most parsimonious trees generated from a heuristic search of nrDNA ITS1-5.8S-ITS2 sequences of selected species of *Lentinula*, *Rhodocollybia*, and *Gymnopus*. Numbers represent branch lengths. Tree length = 2245; CI = 0.5390; HI = 0.4610; RI = 0.6966. Names of clades represent sections in *Gymnopus*.

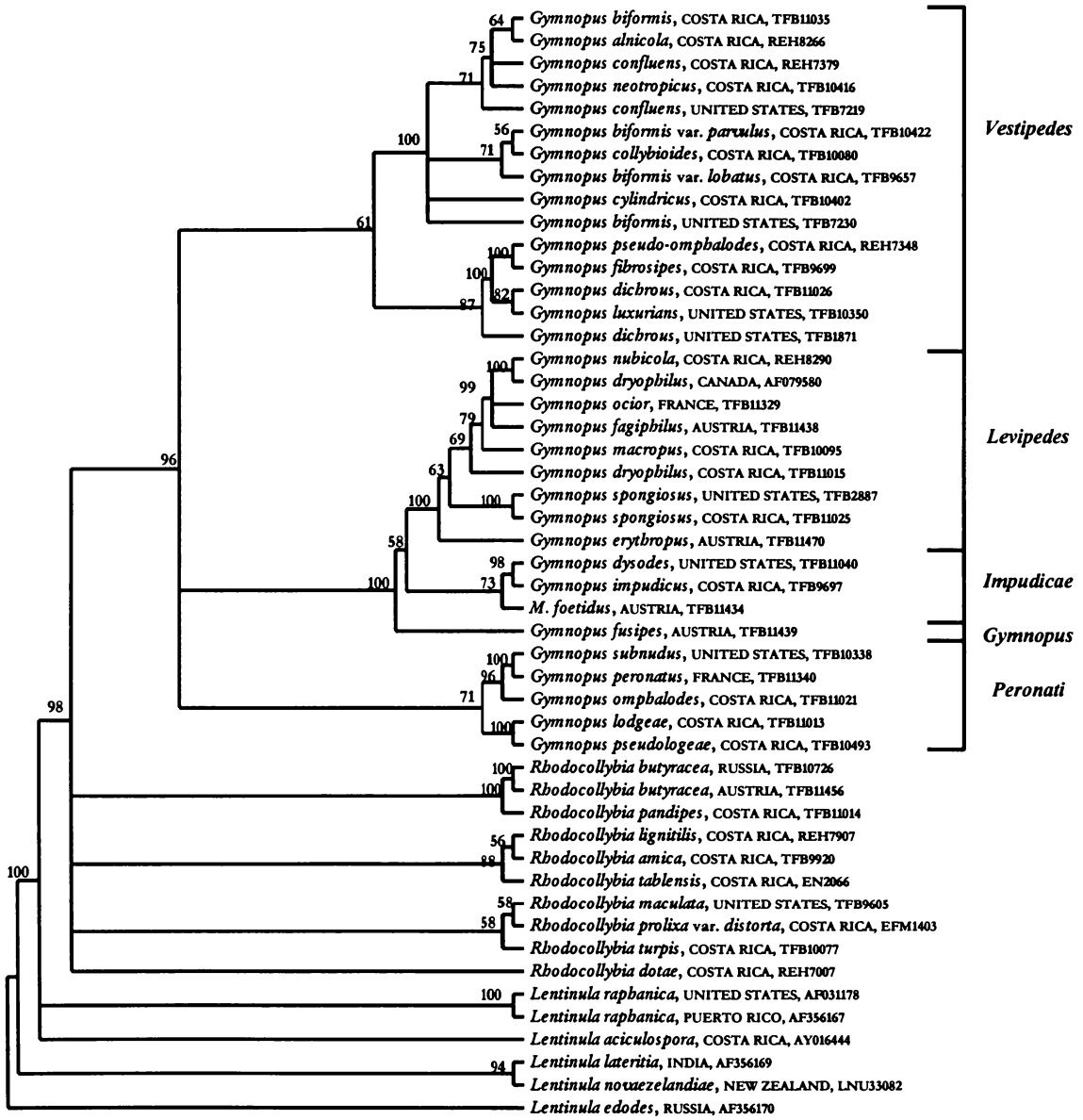


Fig. 4. Strict consensus of twelve most parsimonious trees generated from a heuristic search of nrDNA ITS1-5.8S-ITS2 sequences of selected species of *Lentinula*, *Rhodocollybia*, and *Gymnopus*. Numbers represent bootstrap values over 50% from 1000 replicates. . Tree length = 2245; CI = 0.5390; HI = 0.4610; RI = 0.6966. Names of clades represent sections in *Gymnopus*.

however, to be well-integrated with other species placed *Rhodocollybia* and not with those of *Gymnopus*. Phylogenetic analyses excluding sequences from other *Rhodocollybia* showed these two species basal to those placed in *Gymnopus* (data not shown). Branch lengths in *Rhodocollybia* are long (Figs. 2, 4).

All morphological species placed in *Gymnopus* form a well-supported clade in the neighbor-joining (Fig. 2) and parsimony (Fig. 4) based analyses. Sequences representing Costa Rican, United States and European specimens placed in sect. *Levipedes* appear to form a well-supported phylogenetic clade within *Gymnopus*. While the sequence of *G. nubicola* is closest to a GenBank sequence of *G. dryophilus* (99% similar), sequences of *G. spongiosus* are strongly related to each other.

Species placed in sect. *Vestipedes* (*sensu* Antonín and Noordeloos, 1997) appear not to form a monophyletic clade, but Costa Rican species are phylogenetically related to species described from temperate regions.

Gymnopus fusipes, the type species of the genus, appears to be more closely related to *G. dysodes* and *G. impudicus* placed in sect. *Vestipedes* by Antonín and Noordeloos (1997) but placed here in sect. *Impudicae* (Figs. 2-4). Species in sect. *Levipedes*. are related to this section and to *G. fusipes* (sect. *Gymnopus*). *Marasmiellus foetidus* appears to be phylogenetically related to species in sect. *Impudicae* all of which have fetid-smelling basidiomata as a common morphological characteristic (see discussion for more analysis).

Gymnopus omphalodes apppears more closely related to *G. subnudus* and *G. peronatus*, which form a clade together with *G. lodgeae* and *G. pseudolodgeae* (Figs. 2-4). Presence of pleurocystidia has been reported in all of these species (Antonín and Noordeloos, 1997; Murphy, 1997; Pegler, 1983b). In this dissertation, these species are placed in sect. *Peronati*.

The clade representing sect. *Vestipedes* is that where *G. confluens*, the type species of the section (Antonín and Noordeloos, 1997; Halling, 1993) is located (Figs. 2-4). Within sect. *Vestipedes* is a larger and weakly-supported clade formed by members of the *G. biformis* complex, *G. neotropicus, G. alnicola* and *G. confluens* from Costa Rica and the United States. Costa Rican *G. dichrous* appear in a clade including *G. fibrosipes*, *G. pseudo-omphalodes* and *G. luxurians*, all with a lignicolous habitat. North American *G. dichrous* seems to be basal to this clade.

Taxonomic study.

THE GENUS *RHODOCOLLYBIA*

Key to species of *Rhodocollybia* of the Talamanca Mountains, Costa Rica.

1. Basidiospores inamyloid and acyanophilous.....2
1. Basidiospores dextrinoid and cyanophilous.....3

2. Basidiomata developing brown spots; lamellae white to ivory; pleurocystidia present; spores 4.8-6.0 X 2.8-4.0 µm (Q = 1.30-2.00).....(7) *R. tablensis*
2. Basidiomata not developing any spots; lamellae yellowish; pleurocystidia absent; spores 4.0-6.0 X 2.4-3.6 µm (Q = 1.33-2.33).....(1) *R. amica*

3. Basidiomata developing brown to black spots or changing color upon bruising or when cut.....4
3. Basidiomata with no spots or color changes.....6

4. Pileus becoming strongly sulcate to plicate; lamellae yellowish; wound spots turning black; odor strong; spores 4.2-6.8 X 2.8-4.0 µm (Q = 1.56-1.69).....(8) *R. turpis*
4. Pileus slightly or not sulcate; wound spots not turning black.....5

5. Basidiomata fruiting directly from wood, clustered; lamellae pale orange; wound spots frequent; cheilocystidia flexuous with forked and/or knobbed apex; spores 5.2-7.2 X 3.2-4.8 µm (Q = 1.53-1.60).....(3) *R. lignitilis*

5. Basidiomata on wood or soil, gregarious; lamellae yellowish to pale orange; wound spots infrequent; cheilocystidia long, cylindrical; spores 6.4-10.4 X 3.6-5.2 µm (Q = 1.75-1.86).....(2) *R. dotae*

6. Spores subglobose, 4.0-5.6 X 3.2-4.4 µm (Q = 1.10-1.50).....(6) *R. prolixa* var. *distorta*

6. Spores mostly ellipsoid.....7

7. Pileus with reddish brown, brown to tan colors; lamellae white to pinkish; stipe tapering and with bent base; spores 4.4-9.6 X 2.8-4.8 µm (Q = 1.70-2.13).....(4) *R. pandipes*

7. Pileus brown to pink, or white in age; lamellae white, fimbriate; stipe tapering without bent base; spores 6.4-7.6 X 3.2-4.0 µm (Q = 1.80-2.25).....(5) *R. popayanica*

***Rhodocollybia* (Singer) Mata, emend.**

≡ *Rhodocollybia* Singer. 1939. Schweiz. Z. Pilzk. 17: 71.

Type species *R. maculata* [NEOTYPE: SWEDEN. Småland, Femsjö, 14.IX.1973, Huijsman 73.97 (L).] *n.v.*

Basidiomata usually fleshy, with bruise spots or not; pileus epicutis a cutis or ixocutis; cheilocystidia and pleurocystidia present; spore print cream to pinkish; basidiospores inamyloid or dextrinoid, acyanophilous or cyanophilous; ITS phylogenetic placement related to *R. maculata.*

Section *Rhodocollybia*

Type species *Rhodocollybia maculata.*

(1) *Rhodocollybia amica* Mata, *sp. nov.* Fig. 5

Basidiomata armeniaca; odore fragans et condimentatum; lamellae albo luteae; stipite equalis, constancia carnosa. Basidiosporae 4.0-6.0 X 2.4-3.6 µm.

HOLOTYPE [*hic designatus*]: COSTA RICA. Puntarenas Prov., Coto Brus Co., La Amistad Lodge, 5.VII.1998, col. RH Petersen, *TFB 9920* (TENN).

Etymology: *amica* (Latin) = friendly; geographical epithet for the National Park La Amistad.

Pileus 20-115 mm diam, at first convex, with age applanate to shallowly depressed; surface glabrous, shiny, at disc apricot gold (5B6), fading with age; margin at first incurved to curved, with age decurved and undulating; context very thin, concolorous with surface. **Lamellae** adnate, up to 9 mm broad, close, yellowish (3A2); margin even; lamellulae in three tiers of different lengths. **Stipe** 50-90 X 4-8 mm, cylindrical; surface glabrous, hygrophanous, light brown (4B3); interior hollow; consistency fleshy, brittle. Odor fragrant or spicy; taste fungous. **Habitat** on soil; scattered (Fig. 5a).

Pileus epicutis (Fig. 5b) a simple cutis; hyphae 2-8 µm diam, tightly repent, radially oriented, yellowish in mass, hyaline singly, inamyloid, with clamp connections; wall thin; terminal cells occasionally erect. Gloeopleurous hyphae uncommon, 6-10 µm diam. **Pileus trama** loosely interwoven to ± radially oriented; hyphae 4-16 µm diam, hyaline, inamyloid, with clamp connections; wall thin to 1 µm thick. **Lamellar trama** regular; hyphae 3-12 µm diam, hyaline, inamyloid, with clamp connections; wall thin; mediostratum not distinct from lateral trama; subhymenium narrow, pseudoparenchymatous. **Basidia** (Fig. 5c) 18-21 X 4-5 µm, clavate; sterigmata four; basidioles 14-17 X 3-5 µm, clavate to subfusoid-mucronate. **Pleurocystidia** absent. Lamellar margin fertile. **Cheilocystidia** (Fig. 5d) inconspicuous, 24-40 X 3-6 µm, mostly

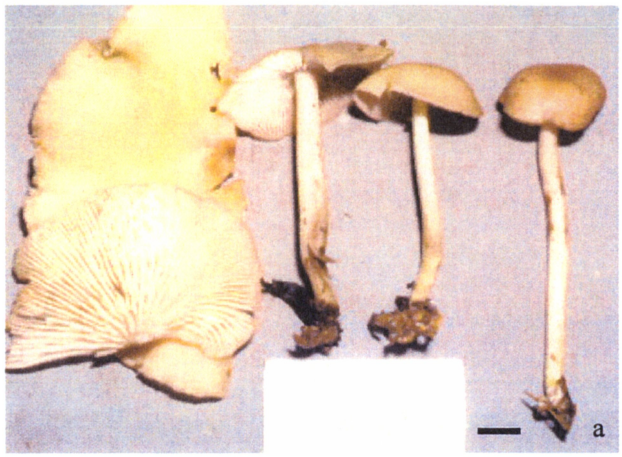
cylindrical in profile; apex obtuse, knobbed, or with a long appendages. **Stipe epicutis** parallel; hyphae 2-14 µm diam, cylindrical, yellowish in mass, hyaline singly, inamyloid, with clamp connections; wall up to 2 µm thick. Gloeopleurous hyphae rare, up to 8 µm diam. **Caulocystidia** (Fig. 5e) forming a dense mat of entangled hyphae, typically 2-3 µm diam, cylindrical, branched or furcate, often knobbed or diverticulate.

Basidiospores (Fig. 5f) yellowish (2A2-4A2, 4A4) in mass, 4.0-6.0 X 2.4-3.6 µm (n = 40, x = 5.2 X 3.0 µm, Q = 1.33-2.33, Qx = 1.78), ellipsoid to subovoid, hyaline, inamyloid; wall smooth, thin; hilar appendix lateral.

Commentary: This specimen has a strong resemblance to mushrooms placed in *Rhodocollybia*, but dextrinoid spores, an important taxonomic character distinguishing this genus, were not detected. More collections are needed to verify absence/presence of dextrinoid spores. Stature of basidiomata and spore print color in *R. amica* resemble those reported for other basidiomata representing species in *Rhodocollybia*, especially those in the *R. butyracea* group. Basidiomata of *R. amica* could be confused with those of *G. dryophilus*, which have a different pileus epicutis configuration.

Rhodocollybia amica appears phylogenetically related to species placed in *Rhodocollybia*, and is closely related to *R. lignitilis,* and to *R. tablensis*, another rhodocollybioid species in which dextrinoid spores were not observed (Figs. 2-4). Phylogenetic placement in combination with morphology are the reasons to propose this species in *Rhodocollybia.*

Material examined: COSTA RICA. Puntarenas Prov., Coto Brus Co., Hacienda La Amistad, 8° 54'22" N, 82° 47'40" W; 1330 m, 5.VII.1998, col. RH Petersen, *TFB 9920* (TENN 56662, HT) **S.**



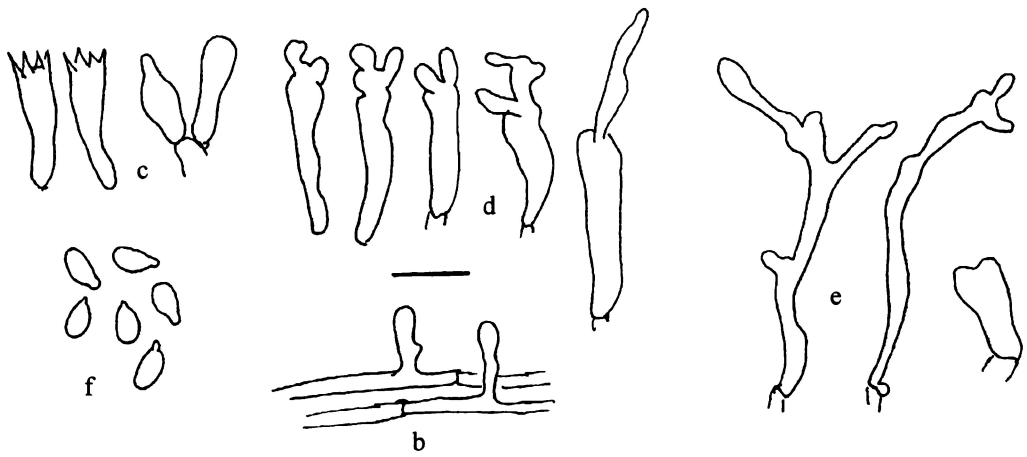


Fig. 5. *Rhodocollybia amica*. a. basidiomata (TFB 9920); b. elements of pileus epicutis; c. basidia and basidioles; d. cheilocystidia; e. caulocystidia; f. basidiospores. Scale bar equals 10 µm for line drawings, 1 cm for picture.

(2) *Rhodocollybia dotae* Mata, *sp. nov.* Fig. 6

Pileo rubellus-aureus ad pallidus-aurantiacus. Lamellae approximatae, subflavidae. Stipe concolorus ad pileo aut pallens. Odore et sapore nullis aut mitis. Basidiosporae 6.4-10.4 X 3.6-5.2 µm.

HOLOTYPE [*hic designatus*]: COSTA RICA. San José Prov., Dota Co., Jardín de Dota, 3.5 km W of Interamerican Highway, 9° 42' 52" N 83° 58' 28" W, 2220 m, 13.VII.1993, col. RE Halling, Mueller, Pine, *REH-7007* (NY).

Etymology: *dota* (Latin) = to endow; geographical epithet for Dota County

Pileus 20-80 mm diam, convex to broadly umbonate; surface radially fibrillose, watery, hygrophanous, orange brown (7C/D8), fading to pale orange brown (6C/D7/6, 6A3),with a few brown spots; margin translucent-striate, entire to irregularly undulate; context thin, watery brown. **Lamellae** adnexed, close, cream to yellowish (4/5A2/3); margin uneven to serrulate. **Stipe** 70-120 X 5-10 mm, equal, sometimes curved; surface white fibrillose above, brownish tan or concolorous to pileus below. Odor and taste mild. **Habitat** on soil or wood, under *Quercus* spp. and *Q. copeyensis* dominant vegetation; gregarious (Fig. 6a).

Pileus epicutis a cutis; hyphae 2-8(-10) µm diam, cylindrical, not branching, ± gelatinized, radially oriented, pigment-incrusted, straw colored in mass, hyaline singly, inamyloid, with clamp connections; wall thin. **Pileus trama** interwoven; hyphae 2-16 µm diam, hyaline, inamyloid, with clamp connections; wall thin. Gloeopleurous hyphae, uncommon, up to 10 µm diam; contents refractive. **Lamellar trama** regular to subregular; hyphae 2-16 µm diam, hyaline, inamyloid, with clamp connections; wall thin. Subhymenium pseudoparenchymatous, 2-4 cells thick, little differentiated. **Basidia** (Fig. 6b) 26-33 X 5-7 µm, clavate; sterigmata four; basidioles (Fig. 6c) 24-32 X 5-7 µm, clavate, sometimes with an obtuse or knobbed apex. **Pleurocystidia** absent.

Cheilocystidia (Fig. 6d) 14-42 X 4-7 μm, clavate to cylindrical, often flexed and with a diverticulate outgrowth at apex, with clamp connections. **Stipe epicutis** parallel; hyphae 4-16 μm diam, light brown in mass, hyaline singly, inamyloid, with clamp connections; wall up to 0.8 μm thick. **Caulocystidia** cylindrical, arising from a ± loose mat of hyphae, 2-4 μm diam.

Basidiospores (Fig. 6e) 6.4-10.4 X 3.6-5.2 μm (n = 40/2, x = 7.8 X 4.3 μm, Q = 1.54-2.20, Qx = 1.81), pip-shaped in side view, ellipsoid in face view, hyaline, inamyloid or dextrinoid (light brown to tan or honey color), and cyanophilous; wall thin or thick, smooth; apiculus ± central to lateral, bent, hyaline.

Commentary: The presence of dextrinoid spores in combination with a simple cutis place basidiomata of this taxon in *Rhodocollybia*. The most similar basidiomata to those of *R. dotae* are those of *R. turpis* but the latter has much darker colored pilei, with gills more distantly spaced, and with a stronger aroma. Also, spore and cheilocystidia dimensions for *R. turpis* are much smaller than those observed in both collections of *R. dotae*. Another taxonomic relative that presents spots on its basidiomata is *R. lignitilis*, distinguished by a smaller stature and strict lignicolous habit. All of the observed morphological characteristics in *R. dotae* are very similar to those characterizing *Rhodocollybia* and provide ground for placing it under this genus name.

Rhodocollybia dotae is phylogenetically related to other species placed in *Rhodocollybia* (Figs. 2,3). In the strict consensus tree *R. dotae* appears to be the most distantly related to other species in the genus (Fig. 4).

Material examined: COSTA RICA. San José Prov., Dota Co., Jardín de Dota, 3.5 km W of Interamerican Highway, 9° 42'52" N, 83° 58'28" W, 2220 m, 13.VII.1993, col. RE Halling, Mueller, Pine, *REH-7007* (NY, HT) S; same location, 17.VI.1994, col. E. Franco-Molano, RE Halling, L Umaña, M Mata, *EFM 1224* (NY).



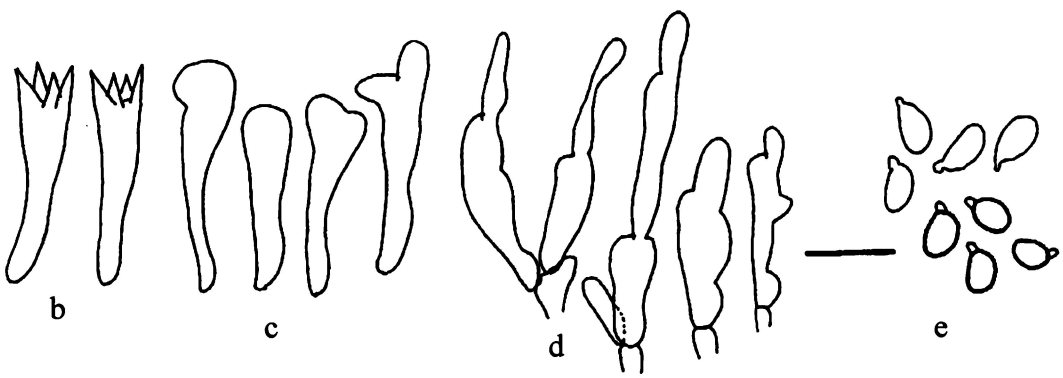


Fig. 6. *Rhodocollybia dotae*. a. basiodiomata (REH-7007); b. basidia; c. basidioles; d. cheilocystidia; e. basidiospores. Scale bar equals 10 μm for line drawings, 1 cm for picture.

(3) *Rhodocollybia lignitilis* Mata, *sp. nov.* Fig. 7

Pileo et lamellae cum maculae porphyraceae quando contunderis. Lamellae aggregatae, aurantiacae albidae. Stipite subclavatus, in materia ligna. Cheilocystidia flexuosae cum apice furcatus aut nodosus. Basidiosporae 4.8-7.2 X 3.2-4.8 µm.

HOLOTYPE [*hic designatus*]: COSTA RICA: San José Prov., Dota Co., San Gerardo, Hotel de Montaña Savegre, 29.VI.1998. col. JL Mata, *TFB 9667* (TENN). ISOTYPE: PANAMA: Chiriquí Prov., Bugabá, Cerro Punta, Parque Internacional La Amistad, Sendero Retoño, 8° 51' N, 82° 34' W, 2280 m, 2.X.1999, col. RE Halling, Duguay, González, *REH 7907* (NY).

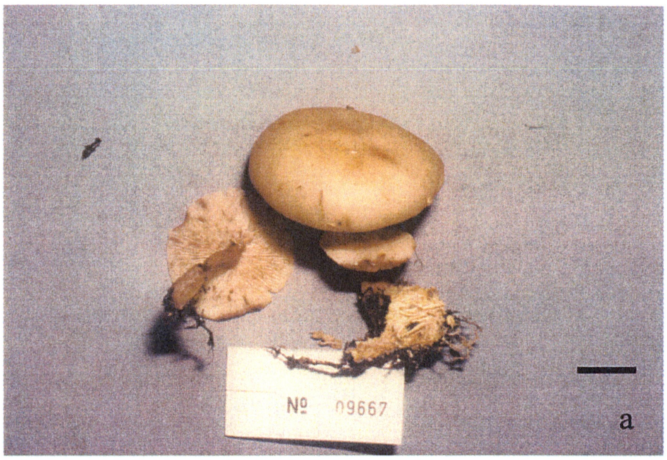
Etymology: *lignitilis* (Latin) = growing on wood.

Pileus 20-70 mm diam, convex, broadly convex to applanate, sometimes with a low umbo; surface glabrous or finely fibrillose, ± lubricous to the touch, shiny, at first light brown to butter-yellow (4A5), pale orange (5A3), with age cinnamon brown (6D6-7), developing brown to reddish brown irregular spots when bruised; margin curved to decurved; context 2-6 mm thick at the center, concolorous to surface, unchanging. **Lamellae** adnexed to adnate, very narrow, crowded to extremely crowded, light orange (4A4, 5A3), unchanging or staining brown when bruised; margin ± even to slightly uneven; lamellulae in several tiers of different lengths, some anastomosing. **Stipe** 30-80 X 3-8 mm, cylindrical, equal to broadening towards base; surface glabrous to fibrillose, moist or wet, superficially ribbed, sometimes twisted, dark orange brown (6E7) or concolorous to lamellae above, more pinkish brown towards base; interior hollow; consistency cartilaginous, tough. Basal mycelium not evident. Odor pleasant, sweet, or mild; taste farinaceous to slightly bitter. **Habitat** on wood; clustered (Fig. 7a).

Pileus epicutis a cutis; hyphae 2-10 µm diam, repent, cylindrical, unbranched, radially oriented, with clamp connections, some with incrusted pigment; terminal cells suberect or repent, flexuous; wall thin. **Pileus trama** ± loosely interwoven; hyphae 2-12 µm diam, hyaline, with clamp connections; wall thin. **Lamellar trama** regular; hyphae 2-12 µm diam, hyaline, sometimes turning light orange in IKI, with clamp connections; wall thin. Subhymenium 2-3 cells thick, pseudoparenchymatous. **Basidia** (Fig. 7b) 24-43 X 5-8 mm, clavate, yellowish brown in mass, hyaline singly; sterigmata four; wall thin; basidioles (Fig. 7c) similar in size to basidia, clavate, ampullaceous to mucronate. **Pleurocystidia** absent. **Cheilocystidia** (Fig. 7d) obscure, 25-48 X 4-8 µm, long clavate, typically strangulate, hyaline, inamyloid; apex furcate, knobbed to lobed; wall thin. **Stipe epicutis** parallel; hyphae 4-14 µm diam, yellowish in mass, hyaline singly, inamyloid, with clamp connections; wall thin to 2.0 µm thick. **Caulocystidia**, if present, short, intertwining in a dense mat of cylindrical to coralloid hyphae, 2-5 µm diam; wall up to 1.6 µm diam.

Basidiospores (Fig. 7e) 5.2-7.2 X 3.2-4.8 µm (n = 40/2, x = 6.0 X 38 µm, Q = 1.30-1.75, Qx = 1.57), broadly ellipsoid to subovoid, hyaline, mostly inamyloid, sometimes dextrinoid (light brown to rosy brown), and cyanophilous; wall smooth, thin to thick; hilar appendix lateral.

Commentary: Because of the stains on the pileus and lamellae, basidiomata of *R. lignitilis* resemble those of the *R. maculata* group. In Costa Rica, basidiomata of *R. lignitilis* are different from those of *R. turpis*, which exhibit a conspicuous sulcate-plicate pileus. Spores of *R. dotae* are much larger (6.4-10.4 X 3.6-5.2 µm). Because of the lubricous pileus surface, basidiomata of *R. lignitilis* resemble distantly those of *R. pandipes*, which differ by a bent stipe. Spores of *R. prolixa* var. *distorta* are subglobose (4.0-5.6 X 3.2-4.4 µm). Other species in *Rhodocollybia* may fruit on woody substrates, but *R. lignitilis* has been collected at two distant locations in the Talamanca Mountains, both strictly on wood.



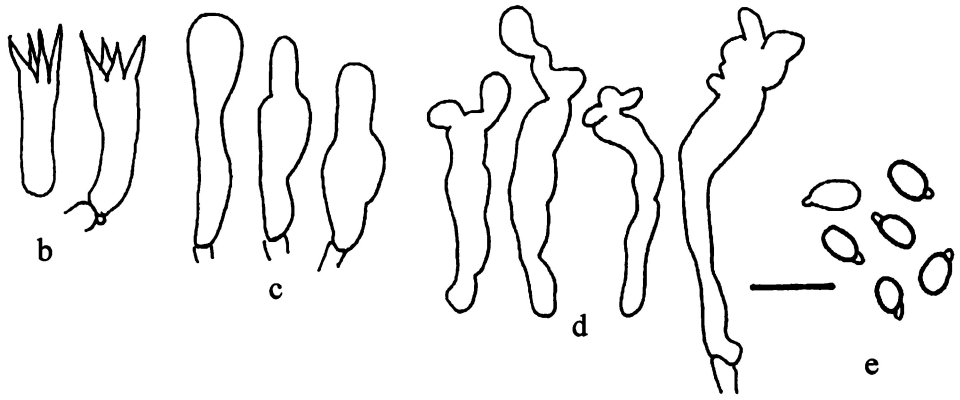


Fig. 7. *Rhodocollybia lignitilis*. a. basidiomata (TFB 9667); b. basidia; c. basidioles; d. cheilocystidia; e. basidiospores. Scale bar equals 10 μm for line drawings, 1 cm for picture.

Rhodocollybia lignitilis appears phylogenetically related to *R. amica* and *R. tablensis* (Figs. 2-4). Morphological characteristics and ITS phylogenetic signal justify proposal of this species in *Rhodocollybia.*

Material examined: COSTA RICA: San José Prov., Dota Co., San Gerardo, Hotel de Montaña Savegre, 29.VI.1998, col. JL Mata, *TFB 9667* (TENN 56628, HT). PANAMA: Chiriquí Prov., Bugabá, Cerro Punta, Parque Internacional La Amistad, Sendero Retoño, 8° 51' N, 82° 34' W, 2280 m, 2.X.1999, col. RE Halling, Duguay, González, *REH 7907* (NY) **S.**

(4) *Rhodocollybia pandipes* Halling & Mata, *sp. nov.* Figs. 8-10

Pileo convexo, aliquando umbonato; superficies glaber, griseo-brunneus ad luteo-brunneus. Odore et sapore mites. Lamellae aggregatae, albidae ad cremeae. Stipite cum basis geniculata et contracta. Basidiosporae 6.4-7.2 X 3.2-4.0 μm.

HOLOTYPE [*hic designatus*]: COSTA RICA. San José Prov., La Chonta, South of Interamerican Highway through oak woods toward Laguna Chonta, 9° 41' 58" N 83° 56' 31" W, 2400 m, 6.VI.1996, col. RE Halling, J Ammirati *REH 7630* (NY).

Etymology: *pandus* (Latin) = bent; *pes* (Latin) = foot.

Pileus 20-70 cm diam, when young parabolic, with age broadly conic, convex to plano-convex with a low broad umbo; surface at first moist and greasy, glabrous, with age hygrophanous and dry, radially appressed fibrillose, disc brown to reddish brown (5E7, 6E6/8), cinnamon brown to caramel brown (6D7/6), brownish orange (5C5/8), towards margin yellow brown to gray brown (6C/D5, 6C6-5C6), pale orange (5A3), grayish yellow (4B4/3) to cream (4A2-3A2); margin even, curved or uplifted, at first non-striate,

with age crenulate-striatulate, translucent; context 2-5 mm thick, white, unchanging. **Lamellae** adnexed, soon seceding to free, crowded to very crowded, 1-6 mm broad, at first white, cream (2/4A2), with age developing a pale flesh pink tint (5A3); margin at first even, with age eroded to subserrate, or fimbriate; lamellulae in two to three tiers. **Stipe** 50-120(-180) X 4-10 mm, subclavate to clavate, terete or flattened, base up to 20 mm diam in some, frequently pinched, tapered, contorted to bent at the base often subradicating; surface glabrous to subfibrillose-striate, sometimes twisted fibrillose, with scattered fine white pruina, above concolorous with pileus, buff pinkish, (5A4/3), grayish orange (5C4), near base pinkish brown (5C4/5), orange brown (6C8), reddish brown (6/7E7/8, 7/8F8) to dark red brown (9E8), but soon discoloring light orange brown (5A5) to cream (3/4A3), and sometimes with yellowish tints, at tip usually white; interior white, at first pithy, with age hollow; mycelium at base white; rhizomorphs white, pinkish buff, occasional. Reaction to $FeSO_4$ (-). Odor fragrant, mild, pungent, or similar to spearmint; taste mild, mealy, or subraphanoid. **Habitat** in leaf litter, humus and soil, in forests of *Quercus* spp; solitary to gregarious (Fig. 8a,b).

Pileus epicutis a cutis; hyphae 2-6(-8) μm diam, repent, cylindrical, unbranched, radially oriented, somewhat loosely interwoven, pigment-incrusted, banded, pale brown in mass, hyaline singly, with clamp connections; wall thin; outermost hyphae, ± gelatinized or not; terminal cells occasionally knobbed and with diverticula. **Pileus trama** interwoven to loosely interwoven, to ± lacunose; hyphae 4-16(-24) μm diam, hyaline in mass and singly, inamyloid; wall thin. Gloeopleurous hyphae, common to uncommon, 4-8 μm diam. **Lamellar trama** regular to subregular; hyphae 3-12(-20) μm diam, hyaline in mass and singly, inamyloid, with clamp connections; wall thin. Gloeopleurous hyphae, up to 8 μm, uncommon. **Basidia** (Fig. 8c) 17-32 X 5-8 mm, clavate, hyaline, occasionally sclerified and dextrinoid; wall thin; sterigmata four; basidioles 16-22 X 4-5 μm, mostly narrowly clavate. **Pleurocystidia** absent. Lamellar margin fertile. **Cheilocystidia** (Fig. 8d) 24-43 X 3-10 μm, mostly cylindrical and ± strangulate, occasionally short clavate, broadly clavate, or long clavate, occasionally concatenate, many collapsed and prostrate, hyaline; wall thin; apex obtuse, knobbed, or

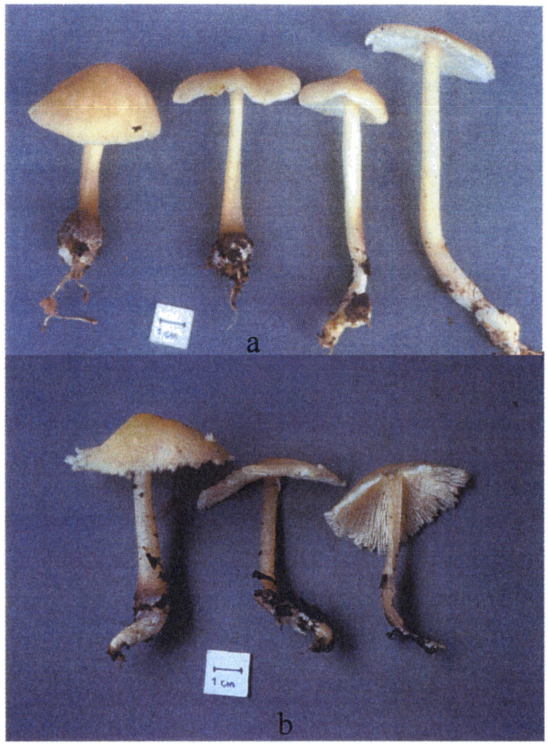
diverticulated; not readily evident. **Stipe epicutis** parallel; hyphae 2-18(-24) μm, yellowish in mass, hyaline singly, inamyloid, with clamp connections; wall thin to 1.6 μm thick. Gloeopleurous hyphae, occasional, 3-5 μm diam. **Caulocystidia** if present, forming an entangled mat of cylindrical elements, 2-9 μm diam.

Basidiospores (Fig. 8e) pinkish cream in deposit; 4.4-9.6 X 2.8-4.8 μm (n = 470/16, x = 6.4 X 3.6 μm, Q = 1.20-2.50, Qx = 1.77), broadly ellipsoid to subovoid, hyaline, typically inamyloid, often dextrinoid (orange brown); wall smooth, thin to thick; hilar appendix lateral.

Commentary: Basidiomata of *R. butyracea* closely resemble those of *R. pandipes* but are different by the predominance of gray-brown to yellow-brown colors of the pilei, and by their more fragile consistency. The tips of stipes of the basidiomata of *R. pandipes* are distinctive in that they tend to bend, and then abruptly taper at the base. From all the species of the genus collected in the oak forests of Talamanca, *R. pandipes* seems to be the most widespread and common. A specimen from Colombia (EFM 835 [NY]) closely resembles those of *R. pandipes*, but the collector reported bruising on the pileus, a feature absent from all collections from Costa Rica.

Rhodocollybia pandipes is well-supported in a clade together with *R. butyracea* from Europe (Figs. 2-4). Sequences for *R. butyracea* are 99% similar between them and 95% similar with TFB 11014 representing *R. pandipes*.

Mating studies. A self-cross of 12 SBIs of collection TFB 11014 yielded data consistent with a tetrapolar mating system (Fig. 9). All four mating types were identified based on clamp connection observation. Mating types A_1B_2 and A_2B_1 were arbitrarily designated based on clamp formation between 1/6/15/16 X 3/4/5/8. Mating types A_1B_1 and A_2B_2 were designated as subordinates. Two mating types are suspected for SBI no. 2. Vilgalys & Miller (1982) reported a tetrapolar mating type for *R. butyracea* from North America which are the same results previously reported for European collections



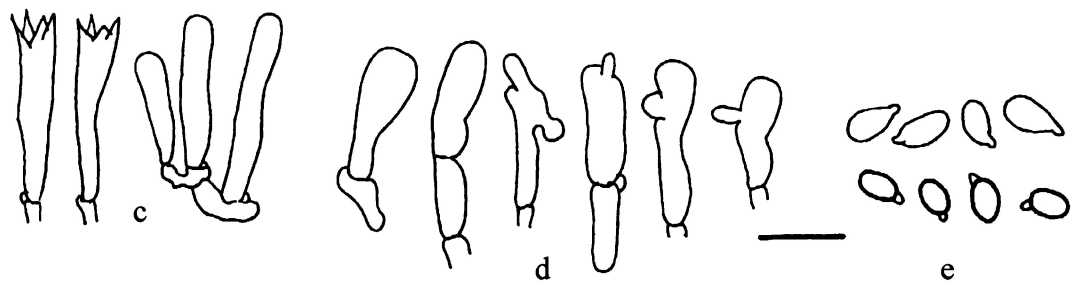


Fig. 8. *Rhodocollybia pandipes*. a,b. basidiomata (a = TFB 11023, b = TFB 10420); c. basidia and basidioles; d. cheilocystidia; e. basidiospores. Scale bar equals 10 μm for line drawings.

(Lamoure, 1989). Intercollection crosses with the available SBIs for some collections of *R. pandipes* showed that compatibility between different geographic locations of the Talamanca Mountains was rather low, and SBIs from TFB 11023 (Villa Mills-CATIE) did not form any clamp connections with any other collections (Fig. 10). Biologically, there may be some reproductive barriers between populations of these separated locations within the Talamanca Mountains, but morphological examination did not reveal any differences between the basidiomata of these collections, which may suggest presence of a cryptic species. More studies, however, are needed to understand the genetics of this newly described species.

Material examined: COSTA RICA. Alajuela Prov., Cerro Palmira, 1800-2000 m, in mixed *Quercus* forest, VI/1986, col. LD Gómez & RM Alfaro *LDG 24779* (F); Cartago Prov., Palo Verde, ± 4.5 km east of Interamerican Highway, near town of Palo Verde, 9° 46'34" N, 83° 56'42" W, 1600 m, 29.V.1996, col. RE Halling, *REH 7581* (NY); San José Prov., La Chonta, south of Interamerican Highway through oak woods toward Laguna Chonta, 9° 41'58" N, 83° 56'31" W, 2400 m, 7.VI.1994, col. RE Halling & TJ Baroni, *REH 7242* (NY); same location 19.VIII.1995, col. E Franco-Molano, J Torres, M Mata, *EFM-1381* (NY); same location, 6.VI.1996, col. RE Halling, J Ammirati REH 7630 (NY, HT); Dota Co., vic. Copey de Dota, 12 km south from Copey on road to Providencia, 9° 35'19" N, 83° 53'00" W, 2795 m, 28.VI.2000, col. col. RE Halling, L. Norvell, RH Petersen & JL Mata, *TFB 11027* (TENN 58635); Jardín de Dota, 3.5 km W of Interamerican Hwy at El Empalme, 9° 42'52" N, 83° 58'28" W, 2220 m, 13.VII.1993, col. RE Halling, G Mueller, E Pine, J Carranza, *REH 7016* (NY); same location, 01.VII.1998, col. RE Halling, RH Petersen & JL Mata, *TFB 9680* (TENN 56641); same location, 10.VIII.1998, col. JL Mata, *TFB 10033* (TENN 56745); same location, 25.VI.2000, col. RE Halling, L. Norvell, RH Petersen & JL Mata, *TFB 11014* (TENN 58622) **S**; San Gerardo de Dota, 1.5 Km W on road toward San Gerardo, 9° 35'47" N, 83° 47'55" W, 2860 m, 9.VI.1997, col. RE Halling & Felsch, *REH 7714* (NY); San Gerardo de Dota, Savegre, 5 Km SW of Cerro de la Muerte, 9° 33" N, 83° 48" W, 2500 m, 6.X.1994, col. GM Mueller, *no 4714* (F); Albergue de Montaña Savegre, 26.VI.2000, col.

					A_1B_2				A_2B_1	A_1B_1		A_2B_2	A1B2 + A2B2
		3	4	5	8	15	16	6	1	10	7	9	
	3*		-	-	-	+	+	+	+	-	-	-	-
	4	-		-	-	+	+	+	+	-	-	-	-
	5	-	-		-	+	+	+	+	-	-	-	-
A_1B_2	8	-	-	-		+	+	+	+	-	-	-	-
	15	+	+	+	+		-	-	-	-	-	-	+
	16	+	+	+	+	-		-	-	-	-	-	+
	6*	+	+	+	+	-	-		-	-	-	-	+
A_2B_1	1	+	+	+	+	-	-	-		-	-	-	-
A_1B_1	10*	-	-	-	-	-	-	-	-		+	+	+
	7*	-	-	-	-	-	-	-	-	+		-	+
A_2B_2	9	-	-	-	-	-	-	-	-	+	-		-
$A_1B_2 + A_2B_2$	2	-	-	-	-	+	+	+	-	+	+	-	

Fig. 9. Self-cross pairings of SBIs from *Rhodocollybia pandipes* TFB 11014. + = presence of clamp connections; - = absence of clamp connections; * = tester strains. Isolate numbers and assigned mating types top and side. SBI 2 may be amphithallic or the product of two germinating spores.

		9680		
Jardín de Dota	9680		11014	
Jardín de Dota	11014	4/4		11023
CATIE - Villa Mills	11023	0/4	0/4	11027
Copey de Dota	11027	1/4	0/4	-

Fig. 10. Intercollection pairings from *Rhodocollybia pandipes*. All pairings are n = 4. Pairing between TFB 11014 and TFB 9680 is with tester strains of TFB 11014.

RE Halling, L Norvell, RH Petersen & JL Mata, *TFB 11019* (TENN 58627); Pérez Zeledón Co., Villa Mills, CATIE Experimental Forest of Villa Mills, 9° 33'03" N, 83° 40'55" W, 2880 m, 22.VI.1995, col. RE Halling & RH Petersen, *TFB 7899* (TENN 53838); same location, 9.VI.1996, col. RE Halling, J Ammirati, M Mata, *REH 7653* (NY); same location 30.VI.1998, col. JL Mata, *TFB 9669* (TENN 56630); same location, 20.VI.1999, col. JL Mata, *TFB 10412* (TENN 58107); same location 23.VI.2000, col. RH Petersen & JL Mata, *TFB 11003* (TENN 58611), *TFB 11004* (TENN 58612); same location 27.VI.2000, col. RH Petersen, RE Halling & JL Mata, *TFB 11023* (TENN 58631); vic. Cerro de la Muerte, near La Georgina, 3200 m, 1.VIII.1986, col. R Singer, *B 14565* (F); Estación Biológica Cuericí, 21.VI.1999, col. JL Mata, *TFB 10420* (TENN 58114).

Rhodocollybia butyracea (Bull.: Fr.) Lennox. [1979. Mycotaxon 9: 218.] AUSTRIA. Lower Austria, Weinviertel, Kuhenring, west of Eqqenburg, 48° 38' N, 15° 45' E, 24.IX.2001, col. H Vogelmayr, *TFB 11456* (TENN 59317) **S**. RUSSIA. NW of Leningrad, Research Station Otradne, 14.IX.2000, *TFB 10726* (spore print only) **S**. UNITED STATES, North Carolina, Jackson Co., vic Cashiers, 12.VII.1999, col. JL Mata, *TFB 10450* (TENN 58144); same location and date, *TFB 10452* (TENN 58146). South Carolina, Oconee Co., vic. Walhalla, 13.VII.1999, col. JL Mata,, *TFB 10460* (TENN 58154).

(5) *Rhodocollybia popayanica* (Halling) Halling. 1997. Mycotaxon 63: 365. Fig. 11
≡ *Collybia popayanica* Halling. 1989. Mycologia 81:872.

HOLOTYPE: COLOMBIA. Dept. Cauca, Municipio de Tunía, km 93 on Pan-American Hwy from Cali to Popayán, Reserva Forestal "EL Guayabo", 1800 m, 19.V.1987, Halling 5272, (FUP) (Isotype, NY) [!].

Pileus 25-35 mm diam, convex to plane, with a low umbo; surface glabrous to finely appressed-fibrillose, moist and hygrophanous, white; margin finely striate; context thin, white, unchanging. **Lamellae** adnexed, crowded, narrow, white; margin fimbriate.

Stipe 50-80 X 3-5 mm, gradually broader below, then tapering to a point; surface glabrous to obscurely fibrillose-striate, white; interior pithy. Odor and taste pungent, non-idiosyncratic. **Habitat** on soil, humus, *Q. corrugata* vegetation; gregarious (Fig. 11a).

Pileus epicutis a cutis; hyphae 2-12 µm diam, cylindrical, occasionally somewhat inflated, radially arranged, hyaline, inamyloid, with clamp connections; wall thin; outermost hyphae ± gelatinous; terminal hyphae repent, sometimes ± coralloid. **Pileus trama** interwoven to ± radially oriented; hyphae 8-20 µm diam, hyaline, inamyloid, with clamp connections; wall thin. **Lamellar trama** regular; hyphae 4-16 µm diam, hyaline, inamyloid, with clamp connections, wall thin. Gloeopleurous hyphae rare, up to 5 µm diam. **Basidia** (Fig. 11b) 22-28 X 4-7 µm, clavate, sterigmata four; basidioles 16-21 X 3-5 µm, clavate. **Pleurocystidia** absent. **Cheilocystidia** (Fig. 11c) 20-52 X 7-16 µm, clavate, sphaeropedunculate, or pedicellate; apex obtuse, lobed, or with diverticula; wall thin. **Stipe epicutis** parallel; outermost hyphae 2-10 µm diam, cylindrical; inner hyphae up to 25 µm diam, inflated, hyaline, inamyloid; with clamp connections; wall thin. **Caulocystidia** absent.

Basidiospores (Fig. 11d) 6.4-7.6 X 3.2-4.0 µm (n = 20, x = 6.8 X 3.4 µm, Q = 1.80-2.25, Qx = 2.01), ellipsoid in side view and face view, mostly hyaline and inamyloid, some dextrinoid (pale reddish brown) and cyanophilous; wall smooth, thin or thick; apiculus slightly bent.

Commentary: Basidiomata of the sole Costa Rican specimen seem to be aged, judging from the white overall color reported by the collector, and fit Halling's (1989) description of Colombian material. Examination of the isotype led me to conclude that all elements observed in the Costa Rican specimen were very similar to those of the isotype, with basidiospores and cheilocystidia having an identical match. Fimbriate lamellar margins and a pungent smell and taste are particularly distinguishing for this taxon. Halling (1989) considered *R. popayanica* to be taxonomically related to the *R. maculata* group, although his specimens did not show changes in colors upon bruising or cutting.

Until now, this species had only been reported from Colombian oak forests (Franco-Molano et al, 2000; Halling, 1989).

Material examined: COLOMBIA. Dept. Cauca, Municipio de Tunía, km 93 on Pan-American Hwy from Cali to Popayán, Reserva Forestal "EL Guayabo", 1800 m, 19.V.1987, col. RE Halling, *REH 5272* (NY, IT). COSTA RICA. Puntarenas Prov., Coto Brus Co., Las Mellizas, Zona Protectora Las Tablas, Sitio Tinieblas 8° 54' N, 82° 6' W, 1450 m, 6.VI.2001, col. RE Halling, B Buyck, R Aldana-Gómez, E Navarro, *REH 8127* (NY).

(6) *Rhodocollybia prolixa var distorta* (Fr.) Antonín, Halling & Noordeloos. 1997. Mycotaxon 63: 365. Fig. 12

≡ *Agaricus distortus* Fries. 1838. Epicr. Syst. Mycol.: 84.

≡ *Collybia distorta* (Fr.) Quélet. 1872. Mém. Soc. Emul. Montbéliard 2: 93.

≡ *Marasmius distortus* (Fr.) P. Karst. 1889. Krit. Öfvers. Finl. Basidsv: 101.

≡ *Rhodocollybia distorta* (Fr.) Singer. 1942. Ann. Mycol. 41: 88.

NEOTYPE: SWEDEN, Västergötland, Trollhättan, Gärdhen, 5 km. SE Störtekleven, 19.VIII.1977, *L & A Stridvall 77/095*, (L) *teste* Antonín & Noordeloos 1997, *n.v.*

Pileus 80-100 mm diam, convex to applanate; surface wet, hygrophanous, yellowish (4A2), becoming whitish; margin undulating; context up to 8 mm thick, whitish. **Lamellae** adnexed, close to crowded, narrow, 3 mm broad, white. **Stipe** up to 120 X 10 mm, central, equal; surface fibrillose, near gray orange (6B4). Odor mild; taste mild, pungent. **Habitat** on soil, in mixed forest with *Quercus spp*; gregarious.

Pileus epicutis a cutis; hyphae 4-6 μm diam, cylindrical, not branching, not gelatinized, radially oriented, ± interwoven, light brown in mass, hyaline singly, inamyloid, with clamp connections; wall thin. **Pileus trama** interwoven; hyphae 4-12 μm diam, hyaline, inamyloid, with clamp connections; wall up to 1 μm thick. Gloeopleurous

hyphae, occasional, up to 4 µm diam. **Lamellar trama** regular; hyphae 4-12 µm diam, hyaline, inamyloid, with clamp connections; wall thin. **Basidia** (Fig. 12a) 19-22 X 5-7 µm, clavate; sterigmata four; basidioles (Fig. 12b) 14-19 X 3-6 µm, clavate to cylindrical. **Pleurocystidia** not observed. Lamellar edge fertile, heteromorphic. **Cheilocystidia** (Fig. 12c) obscure, 12-33 X 4-7 µm, short clavate, with clamp connections; apex occasionally lobed or with outgrowths. **Stipe epicutis** parallel; hyphae 4-18 µm diam, frequently with diverticula, pale yellowish in mass, hyaline singly, inamyloid, with clamp connections; wall up to 1 µm thick. **Caulocystidia** arising and forming a dense, interwoven mat of hyphae, 2-4 µm diam, cylindrical, often contorted, occasionally knobbed, with clamp connections.

Basidiospores (Fig. 12d) 4.0-5.6 X 3.2-4.4 µm (n = 25, x = 5.0 X 3.9 µm, Q = 1.10-1.50, Qx = 1.28), subglobose in side view and in face view, hyaline, mostly inamyloid, some weakly dextrinoid (light orange brown), acyanophilous, not metachromatic; wall smooth, thin or slightly thickened in dextrinoid spores.

Commentary: Because some of the subglobose spores observed were weakly dextrinoid, this specimen must be placed under *Rhodocollybia.* All characteristics observed and measured fit into *R. prolixa* var. *distorta*, a taxon described originally from Europe (Antonín & Noordeloos, 1997). In North America, basidiomata of *R. prolixa* var. *distorta* appear to be similar to those of *R. badiialba* which reportedly has much darker colored basidiomata (Halling, 1983). Concomitantly, this specimen is very similar to the type of *C. subdryophila* in the shape and dimensions of most micromorphological characteristics (spores, cystidia, hyphae), but no conclusive dextrinoid reaction (yellow brown) of the spores from the type was detected. Annotations on the type made by R.E. Halling and R. Vilgalys indicate absence of dextrinoid spores and tissues. The dried Costa Rican specimen looks similar to the type of *C. subdryophila. Rhodocollybia prolixa* var. *distorta* constitutes a new report for Costa Rica. The name *C. distorta* (= *R. prolixa* var. *distorta*) has been reported from the United States (Halling, 1983), México (Guzmán et al, 1992) and Jamaica (Dennis, 1968) but voucher material from these

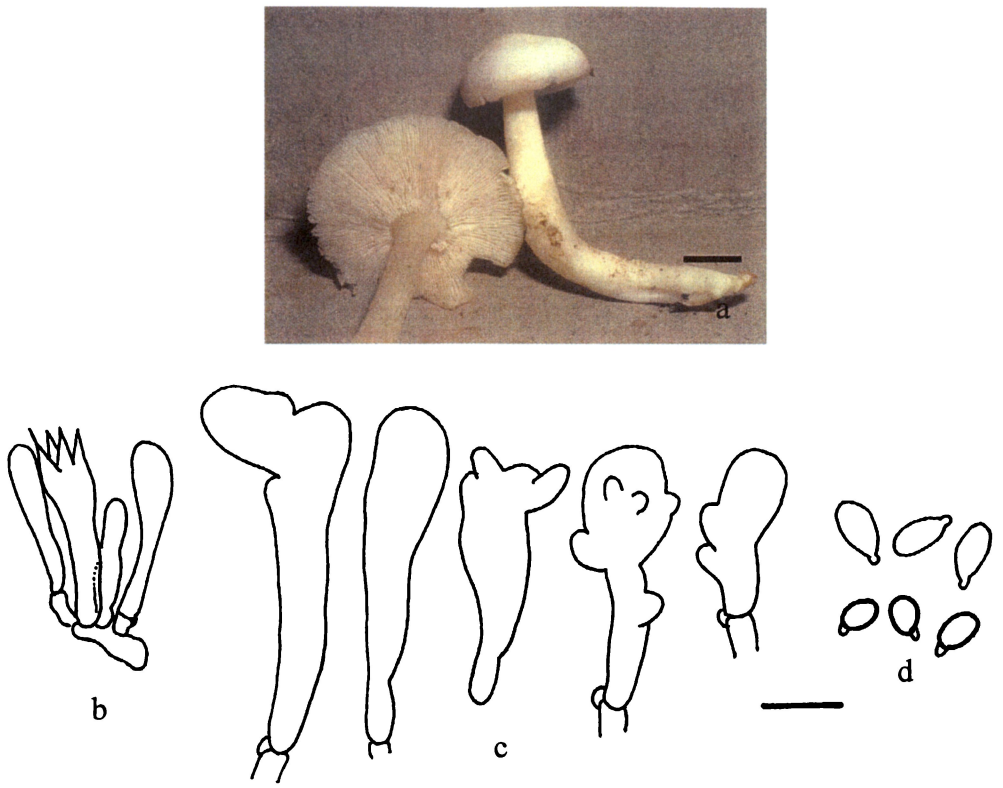


Fig. 11. *Rhodocollybia popayanica*. a. basidiomata (REH 8127); b. basidium and basidioles; c. cheilocystidia; d. basidiospores. Scale bar equals 10 μm for line drawings, 1 cm for picture.



Fig. 12. *Rhodocollybia prolixa var. distorta* (EFM 1403). a. basidia; b. basidioles; c. cheilocystidia; d. basidiospores. Scale bar equals 10 μm.

regions was not studied. The type specimen of *R. prolixa* var. *distorta* was not available for study before the conclusion of this dissertation.

Rhodocollybia prolixa var. *distorta* is placed within the *Rhodocollybia* clade in both neighbor-joining and parsimony analyses (Figs. 2-4). It appears phylogenetically closely related to *R. maculata*, the type species of the genus.

Material examined: COSTA RICA. San José Prov., La Guaria, 9° 36' N, 83° 59' W, 1900 m, 21.VIII.1995, col. E Franco-Molano, J Torres, M Mata, *EFM-1403* (NY) **S.** UNITED STATES. North Carolina, Chapel Hill, Fern Walk near Sparrow's Pond, col. WC Coker, 2.X.1908, *no. 22634*, HT of *Collybia subdryophila* Atkinson; (CUP-A) [!].

Rhodocollybia maculata (A. & S.: Fr.) Singer. [1939. Schweiz. Z. Pilz. 17:71.] UNITED STATES. North Carolina, Macon Co, Standing Indian Campground, 35° 04'47" N, 83° 31'83" W, 26.V.1998, col. E Lickey, TFB 9605 (TENN 56566) **S.**

(7) *Rhodocollybia tablensis* Mata, *sp. nov.* Fig. 13

Pileo convexo, subumbonatus. Sapore amarus neglegere. Lamellae aggregatae, albidae. Stipite radicans, albus, subinde cum maculae fuscae. Cheilocystidia et pleurocystidia praescentis. Basidiosporae 4.8-6.0 X 2.8-4.0 µm.

HOLOTYPE [*hic designatus*]: COSTA RICA, Puntarenas Prov., Zona Protectora Las Tablas, Quijada del Diablo, 11.VI.2000, col: E.Navarro, *EN 2067* (INB)

Etymology: *tablensis* (Latin) = geographical epithet for the protected zone of Las Tablas.

Pileus 16-32 mm diam, campanulate to slightly mammilate; surface fibrillose, disc vinaceous pink (7D5/6), near rust brown (6D8), outward off-white; margin incurved

to decurved, even; context 2-5 mm broad, white. **Lamellae** free, up to 2 mm broad, crowded, white to ivory (4A3-4B4); margin even; lamellulae in several tiers of different lengths. **Stipe** 100-200 X 4-7 mm, central, equal to obbclavate, sometimes with a radicating tip; surface fibrillose, white, sometimes with brown spots, near walnut borwn (7E7); interior solid, white. Rhizomorphs white. Odor not distinctive; taste unpleasant or slightly bitter. **Habit** on soil; gregarious (Fig. 13a).

Pileus epicutis (Fig. 13b) a cutis; hyphae hyaline singly, inamyloid, with clamp connections; wall thin; suprapellis ± 40 μm broad; hyphae of suprapellis 2-4 μm diam, radially oriented, slightly interwoven, slightly gelatinized, pigment-encrusted, with numerous erect terminal cells, flexuous; subpellis ± 200 μm broad; hyphae of subpellis 4-12 μm diam, radially oriented. **Pileus trama** loosely interwoven; hyphae 8-16 μm diam, hyaline, inamyloid, with clamp connections; wall up to 0.8 μm thick. **Lamellar trama** strictly regular; hyphae 4-10 μm diam, hyaline, inamyloid, with clamp connections; wall thin. **Basidia** (Fig. 13c) 22-32 X 7-12 μm, broadly clavate to fusoid ventricose; sterigmata four; basidioles 12-18 X 3-4 μm, narrowly clavate to clavate. **Pleurocystidia** (Fig. 13d) abundant, 28-84 X 8-16 μm, broadly clavate, ventricose-fusoid, inflated, voluminous; wall thin. **Cheilocystida** (Fig. 13e) 26-36 X 5-7 μm, slender, clavate to cylindrical, mixed with pleurocystidia. **Stipe epicutis** parallel; hyphae 4-14 μm, hyaline singly, inamyloid; wall up to 0.8 μm thick. **Caulocystidia** (Fig. 13f) common, up to 20 X 6 μm, short clavate, occasionally furcate.

Basidiospores (Fig. 13g) reported as white in mass, 4.8-6.0 X 2.8-4.0 μm (n = 20, x = 5.4 X 3.4 μm, Q = 1.30-2.00, Qx = 1.61), broadly ellipsoid to obovoid in side view and in face view, hyaline, inamyloid, acyanophilous, not metachromatic; wall thin, smooth; apiculus slightly lateral.

Commentary: Like *R.. amica*, this specimen has the stature of a *Rhodocollybia*, but no thick-walled, dextrinoid spores were found. Also, the presence of stains on the stipe, similar to those found in *R. maculata*, make this specimen a good candidate for

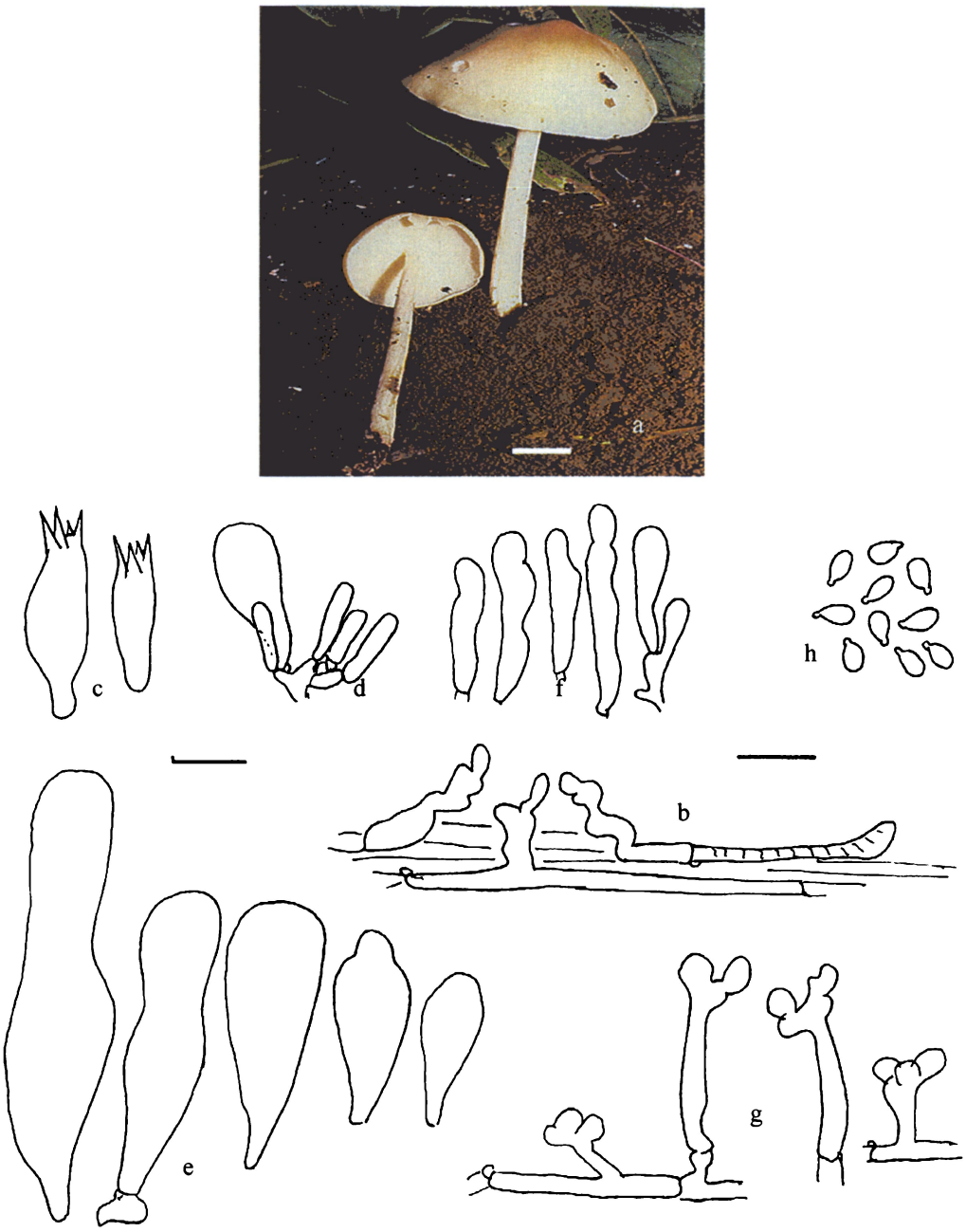


Fig. 13. *Rhodocollybia tablensis*. a. basidiomata (EN 2066); b. elements of pileus epicitis; c. basidia; d. basidioles and pleurocystidium; e. pleurocystidia; f. cheilocystidia; g. caulocystidia; h. basidiospores. Scale bar equals 10 μm for line drawings, 1 cm for picture.

Rhodocollybia. Presence of pleurocystidia in this species is shared with *R. monticola*, an undescribed species from South America (R.E. Halling, pers. com.). Certain members in *Gymnopus* sect. *Peronati* like *G. lodgeae* and *G. pseudolodgeae* possess conspicuous hymenial cystidia but are smaller in stature, and their basidiomata can be distinguised by the sulcate and much darker pilei and more distantly spaced lamellae.

Rhodocollybia tablensis is phylogenetically related to *R. amica* and it is nested within the *Rhodocollybia* clade (Figs. 2-4). Morphological characteristics in combination with apparent phylogenetic placement seem to justify the proposal of *R. tablensis* in *Rhodocollybia*.

Material examined: COSTA RICA, Puntarenas Prov., Zona Protectora Las Tablas, Quijada del Diablo, 11.VI.2000, col: E.Navarro, *EN 2066* (INB 3108789) **S**, *EN 2067* (INB 3108790, HT).

(8) *Rhodocollybia turpis* (Halling) Halling. 1997. Mycotaxon 63: 366. Fig. 14

≡ *Collybia turpis* Halling. 1989. Mycologia 81: 879.

HOLOTYPE: COLOMBIA. Dept. Antioquia, Mun. Guarne, near Centro Experimental Piedras Blancas, ± 14 Km E of Medellín, 2350 m, under *Q. humboldtii*, 10.XI.1988, col. DE Desjardin & RE Halling, *REH 6077*, (NY) [!].

Pileus 10-70 mm broad, at first conico-convex, with age broadly convex to applanate, somewhat umbonate; surface glabrous, ± lubricous, sulcate, at first brown yellowish (4A4), with age or when handled orange brown (6D8, 8E8) at center, yellowish (4A5/6) towards margin, staining brown to black when bruised; margin sulcate-striate, ± translucent, curved to incurved; context thin, concolorous to pileus. **Lamellae** adnate, up to 4 mm, subdistant, yellowish orange (3A6), buff 4B3), staining brown when bruised; margin even or emarginate, sometimes brown; lamellulae in two tiers. **Stipe** 40-90 X 3-8 mm, equal or broader at center or base, base occasionally tapering, fistulose; surface

fibrillose, hygrophanous, sometimes twisted, becoming a little velvety at apex, light yellow (2A4), changing to orange brown when bruised; interior hollow; consistency fibrous. Odor farinaceous, strong; taste pleasant, mild. **Habitat** on leaf litter or woody debris; gregarious (Fig. 14a).

Pileus epicutis a cutis; hyphae 2-6 μm diam, cylindrical, repent, unbranched, radially oriented, yellowish in mass, hyaline single, with clamp connections; wall thin; terminal elements clavate to subglobose. Gloeopleurous hyphae scattered, brownish in color. **Pileus trama** interwoven to ± radially oriented; hyphae 4-14 μm diam, yellowish in mass, hyaline singly, with clamp connections; wall thin. **Lamellar trama** subregular to interwoven; hyphae 3-9 μm diam, hyaline, with clamp connections; wall thin. **Basidia** (Fig. 14b) 32-41 X 6-9 μm, clavate to cylindrical, projecting from hymenium layer, evident; sterigmata four; basidioles of similar shape and size. Lamellar margin sterile. **Pleurocystidia** absent. **Cheilocystidia** (Fig. 14c) 12-28 X 5-7 μm, clavate, sub-ampullaceous, flexuous, with clamp connections; not readily evident; apex obtuse or mucronate. **Stipe epicutis** parallel; hyphae 4-12 μm diam, with clamp connections; wall thin. **Caulocystidia** (Fig. 14d) 28-44 X 5-7 μm, similar to cheilocystidia, arising from a loose mat of short-celled hyphae.

Basidiospores (Fig. 14e) 4.2-6.8 X 2.8-4.0 μm (n = 19, x = 5.8 X 3.4 μm, Q = 1.40-2.14, Qx = 1.69), ellipsoid to broadly ellipsoid, or lacrymoid in side view, hyaline, inamyloid, occasionally dextrinoid (reddish brown); wall smooth, thin or thick.

Commentary: *Rhodocollybia turpis* has been collected in oak forests from Colombia and Costa Rica (Halling, 1989). Its conspicuously sulcate pileus, yellow-orange lamellae that develop spots upon bruising and soon turn black, and strong odor are unique field characters. All macroscopic and microscopic characters match those seen in the type specimen. In the Talamanca Mountains of Costa Rica, *R. turpis* has been collected mostly in Estrella de Cartago. Along with *R. popayanica*, *R. turpis* is found also



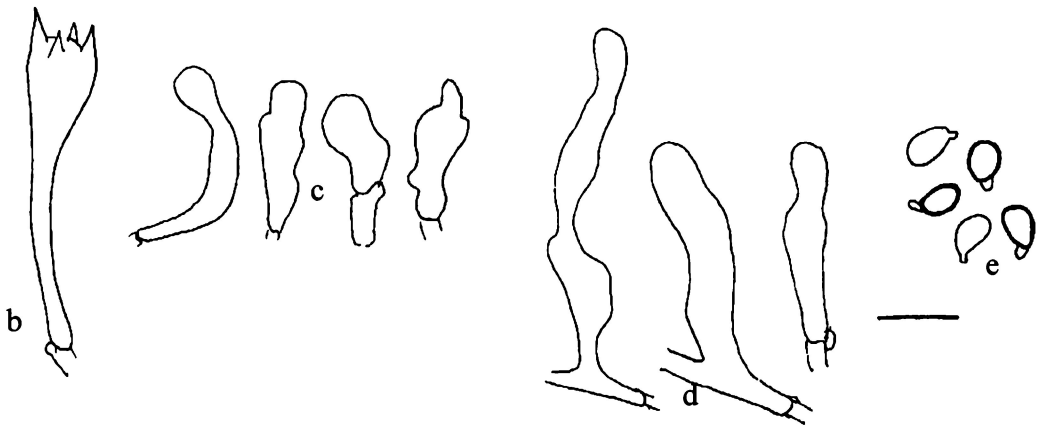


Fig. 14. *Rhodocollybia turpis*. a. basidiomata (TFB 9655); b. basidium; c. cheilocystidia; d. caulocystidia; e. basidiospores. Scale bar equals 10 μm for line drawings, 1 cm for picture.

in Colombian oak forests (Franco-Molano et al, 2000; Halling, 1989) and, until now, their northernmost distribution limit seemed to be in southern Costa Rica.

Rhodocollybia turpis is phylogenetically related to *R.. maculata,* and *R. prolixa* var. *distorta* (Figs. 2-4). Basidiomata of all these species are reported to develop spots or stains (Antonín and Noordeloos, 1997; Halling, 1983).

Material examined: COSTA RICA: Cartago Prov., vic. Estrella, Palo Verde; 9° 46'59" N, 83° 56'71" W, ± 1700 m, 13.VI.1995, col. RE Halling & RH Petersen, *TFB 7688* (TENN 53789); same location, 28.VI.1998, col. JL Mata, & RH Petersen, *TFB 9655* (TENN 56616); same location, 16.VI.1999, col. JL Mata, *TFB 10077* (TENN 58017) **S**; Puntarenas Prov., Coto Brus Co., vic. San Vito, Las Cruces Biological Station, Ridge trail, 8° 47'9" N, 82° 57'31" W, 1100 m., 12.X.1992, col. RE Halling, G Mueller, B Strack *REH 7347* (NY) *teste* RE Halling, *n.v.*

THE GENUS *GYMNOPUS*

Key to species in *Gymnopus* of the Talamanca Mountains, Costa Rica.

1. Pileus epicutis composed of repent, short, branched hyphae, not radially oriented.....2
1. Pileus epicutis composed of repent, cylindrical, unbranched hyphae, radially oriented.....5

2. Basidiomata with stipe tissues turning green in KOH; pileus with reddish brown tints; rhizomorphs cream colored; spores 6.4-8.8 X 3.6-4.4 µm (Q = 1.73-2.20).....(13) *G. spongiosus*
2. Basidiomata with no tissues turning green in KOH.....3

3. Basidiomata fruiting in Páramo vegetation; rhizomorphs with pinkish tints; spores 4.8-5.6 X 2.4-3.6 µm (Q = 1.50-2.33).....(12) *G. nubicola*
3. Basidiomata associated with oak forest vegetation; rhizomorphs white, cream to yellowish.....4

4. Stipe long and slender in relation to pileus diameter, with persisting orange colors; cheilocystidia mostly short, clavate; spores 6.0-8.4 X 3.2-4.0 µm (Q = 1.60-2.57).....(11) *G. macropus*
4. Stipe obclavate, base frequently cleft, bulbous; cheilocystidia clavate, often contorted and/or with diverticula; spores 4.8-6.8 X 2.4-3.6 µm (Q = 1.56-2.29).....(10) *G. dryophilus*

5. Pleurocystidia present.....6
5. Pleurocystidia absent.....8

6. Pileus at first brown, with age fading to off-white, crenate; lamellae close; cheilocystidia narrowly clavate; spores 6.4-8.0 X 3.2-3.6 µm (Q = 1.70-2.75).....(15) *G. omphalodes*
6. Pileus brown to tan throughout aging, sulcate; lamellae subdistant, cheilocystidia clavate to lanceolate.....7

7. Pleurocystidia up to 190 µm long; spores 6.4-8.0 X 2.8-3.6 µm (Q = 1.75-2.71).....(14) *G. lodgeae*
7. Pleurocystidia up to 80 µm long; spores 4.8-6.4 X 2.4-3.6 µm (Q = 1.56-2.29).....(16) *G. pseudolodgeae*

8. Basidiomata with foetid or fenugreek odor.....9
8. Basidiomata with mild or no distinct odor.....10

9. Basidiomata with strong foetid to pungent odor and taste; pileus convex to

convex-umbilicate, reddish-brown to brown, discoloring around margin, lamellae close, white to off-white; stipe with pale brown to pinkish tints; cheilocystidia clavate to sphaeropedunculate; spores 6.0-8.0 X 2.8-4.0 µm (Q = 1.78-2.29).....(9) *G. impudicus*

9. Basidiomata with fenugreek or sweetish odor, taste bitter; pileus conico-convex, light brown, not discoloring at margin; lamellae close to crowded; off-white to cream; stipe brown orange to purple brown; cheilocystidia clavate to cylindrical; spores 4.8-6.4 X 2.4-3.6 µm (Q = 1.50-2.83).....(27) *G. polyphyllus*

10. Spores 7.2-12.8 µm long.....11

10. Spores 4.0-9.2 µm long.....16

11. Basidiomata fruiting only under *Alnus acuminata*; pileus dark brown; lamellae with lilac tints; spores 7.6-9.6 X 3.2-4.0 µm (Q = 2.00-2.78).....(17) *G. alnicola*

11. Basidiomata fruiting in oak forest vegetation.....12

12. Habitat lignicolous.....13

12. Habitat on leaf litter, humus and/soil.....14

13. Pileus broadly convex, crenate-sulcate, pale brown; lamellae adnexed at first, seceding into pseudocollarium, subdistant; cheilocystidia voluminous; spores 8.8-12.8 X 4.0-5.6 µm (Q = 1.71-2.45).....(24) *G. dichrous*

13. Pileus obtusely convex to subumbonate, striate-sulcate, brown to dark brown; lamellae adnexed, not seceding into a pseudocollarium, crowded; cheilocystidia not volumnious; spores 8.0-9.6 X 3.6-4.8 µm (Q = 1.82-2.22).....(28) *G. pseudo-omphalodes*

14. Pileus sulcate; lamellae close to subdistant; broad, spores 7.2-9.6 X 3.6-5.2 µm (Q = 1.64-2.44).....(26) *G. neotropicus*

14. Pileus not deeply sulcate; lamellae crowded, narrow.....15

15. Basidiomata with orange-gray tints; lamellar margin fimbriate; cheilocystidia frequently lobed; spores 7.2-8.4 X 3.2-4.8 µm (Q = 1.80-2.63).....(22) *G. confluens*
15. Basidiomata with orange-brown tints; lamellar margin even; cheilocystidia typically cylindrical; spores 9.6-11.2 X 3.2-4.0 µm (Q = 2.10-3.63).....(23) *G. cylindricus*

16. Pileus more than 40 mm diam, conico-convex; stipe compressed-sulcate; habitat on humus or soil; spores 6.4-8.8 X 3.2-4.0 µm (Q = 1.78-2.44).....(25) *G. fibrosipes*
16. Pileus less than 40 mm diam, with age depressed-umbilicate; stipe terete, not sulcate; habitat on leaf litter.....17

17. Lamellae never forming a pseudocollarium, crowded, white; spores 6.0-8.8 X 3.2-4.0 µm (Q = 1.75-2.88).....(21) *G. collybioides*
17. Lamellae often forming a pseudocollarium, close, off-white.....18

18. Stipe densely pubescent to strigose, reddish brown; spores 6.4-9.2 X 2.4-4.8 µm (Q = 1.80-2.88).....(18) *G. biformis*
18. Stipe not densely pubescent to pruinose.....19

19. Stipe base blackening; cheilocystidia typically lobate; spores 6.4-8.4 X 3.2-4.0 µm (Q = 1.63-2.50).....(19) *G. biformis var. lobatus*
19. Stipe base not blackening; cheilocystidia clavate, subfusoid or cylindrical; spores 5.6-8.0 X 2.8-3.6 µm (Q = 1.60-2.75).....(20) *G. biformis var. parvulus*

***Gymnopus* (Pers.) Roussel. 1806. Fl. Calavados, 2nd ed.: 62.**
Type species *Gymnopus fusipes* [EPITYPE: FRANCE. Dept. L. & Ch., Montrichard, 19.IX.1955, OF Uffelie, (L)] *n.v.*

Section *Impudicae* (Antonín and Noordeloos) Mata, *comb. nov*

≡ *Gymnopus* sect. *Vestipedes* subsect. *Impudicae* Antonín and Noordeloos. 1997. Libri Botanici 17: 66.

Type species *G. impudicus* [NEOTYPE: SWEDEN. Västergötland, Göteborg, Slottsskogen. 4.IX.1942. col. T Nathorst-Windahl, *no. 3243* (PRM)] [!]

Basidiomata with fetid, unpleasant, or strong odor; pileus epicutis composed of cylindrical, diveticulate, radially oriented hyphae.

(9) *Gymnopus impudicus* (Fr.) Antonín, Halling & Noordeloos. 1997. Mycotaxon 63: 364. Figs. 15-17

≡ *Marasmius impudicus* Fr. 1838. Epicr. Syst. Mycol. 377.

≡ *Collybia impudica* (Fr.) Singer. 1943. Ann. Mycol. 41: 11.

≡ *Micromphale impudicus* (Fr.) PD Orton. 1960. Trans. Br. Mycol. Soc. 43: 178.

NEOTYPE: SWEDEN. Västergötland, Göteborg, Slottsskogen. 4.IX.1942. col. T Nathorst-Windahl, *no. 3243* (PRM)] [!].

Pileus 10-30(-50) mm diam, at first convex, with age broadly convex, convex umbilicate, irregularly convex to applanate; surface fibrillose, shiny, hygrophanous or dry, with age translucent-crenate toward margin, disc reddish brown (7D6), dark brown (7-9F8), agate brown (7E8) fading to brown (7E6), cinnamon brown (6D6/7), center sometimes pale; margin at first even and curved, soon crenate and undulating, uplifted, translucent, paler than disc, off-white to tan; context thin, white to watery brown. **Lamellae** adnate, adnexed to seceding, very narrow, crowded to close, rarely subdistant, white to white yellowish (2-4A2); margin even to finely fimbriate; lamellulae in three to five tiers of different lengths, some anastomosing or with intervenose projections. **Stipe** 20-70(-80) X 2-6 mm, equal, sometimes ± broader at base or apex, terete to compressed and cleft; surface subpruinose above to pruinose below, when young near apex cream, gray orange (6B5, 5A3) to orange cream (5/6A2), downwards dark brown (6E8), agate brown (7E6), grayish brown (5C4, 6E5) to light brown (6-7D-E5), occasionally blackening at base; vesture whitish; interior hollow; consistency tough. Mycelium and

rhizomorphs white, creamy or with yellowish tints. Odor alliaceous, pungent, spicy, strongly fragrant or none; taste acrid, mealy, mild or not distinctive. **Habitat** on leaf litter and humus, under *Cupressus spp* or mixed *Quercus spp* vegetation; gregarious to cespitose (Fig. 15a,b).

Pileus epicutis (Fig. 15c) a cutis; hyphae 2-12 µm diam, cylindrical, repent, not branched, radially oriented, occasionally to frequently diverticulate, pigment-encrusted, light brown to olive brown in mass, hyaline singly, light orange in IKI, with clamp connections; wall thin; terminal cells, cylindrical or short to broadly clavate, prostrate, semierect to erect, usually diverticulate, frequently arising at hyphal septa. **Pileus trama** interwoven to radially oriented; hyphae 2-10 µm diam, hyaline, inamyloid, with clamp connections; wall thin. **Lamellar trama** subregular to interwoven; hyphae 2-12 µm diam, hyaline, inamyloid, with clamp connections; wall thin; subhymenium pseudoparenchymatous. **Basidia** (Fig. 15d) 18-28(-34) X 5-6 µm, clavate; sterigmata four; basidioles 16-24(-28) X 4-5 µm, mostly clavate or cylindrical, occasionally submucronate. **Pleurocystidia** absent. Lamellar margin sterile or fertile. **Cheilocystidia** (Fig. 15e) 14-32(-90) X 4-8 µm, narrowly clavate to clavate, sphaeropedunculate, often furcate or flexed, occasionally developing long appendages, concatenate, with clamp connections; arising from horizontal hyphae. **Stipe epicutis** strictly parallel; hyphae 2-12 µm diam, yellowish in mass, hyaline singly, inamyloid, with clamp connections; wall thin. **Caulocystidia** abundant, up to 100 X 6 µm, cylindrical, ± strangulate, septate, in fascicles or entangled and forming a mat.

Basidiospores (Fig. 15f) (5.6-)6.0-8.0(-8.8) X 2.8-4.0 µm (n = 207/10 x = 7.0 X 3.6 µm, Q = (1.56-)1.78-2.29(-2.44), Qx = 1.98), ellipsoid or lacrymoid in side view, ellipsoid in face view, hyaline, inamyloid; wall thin, smooth.

Commentary: *Gymnopus impudicus* is a species originally described from Europe, characterized by its garlic-like odor, similar to the odor noted in most specimens collected under this name during this study. There is no other Costa Rican *Gymnopus*

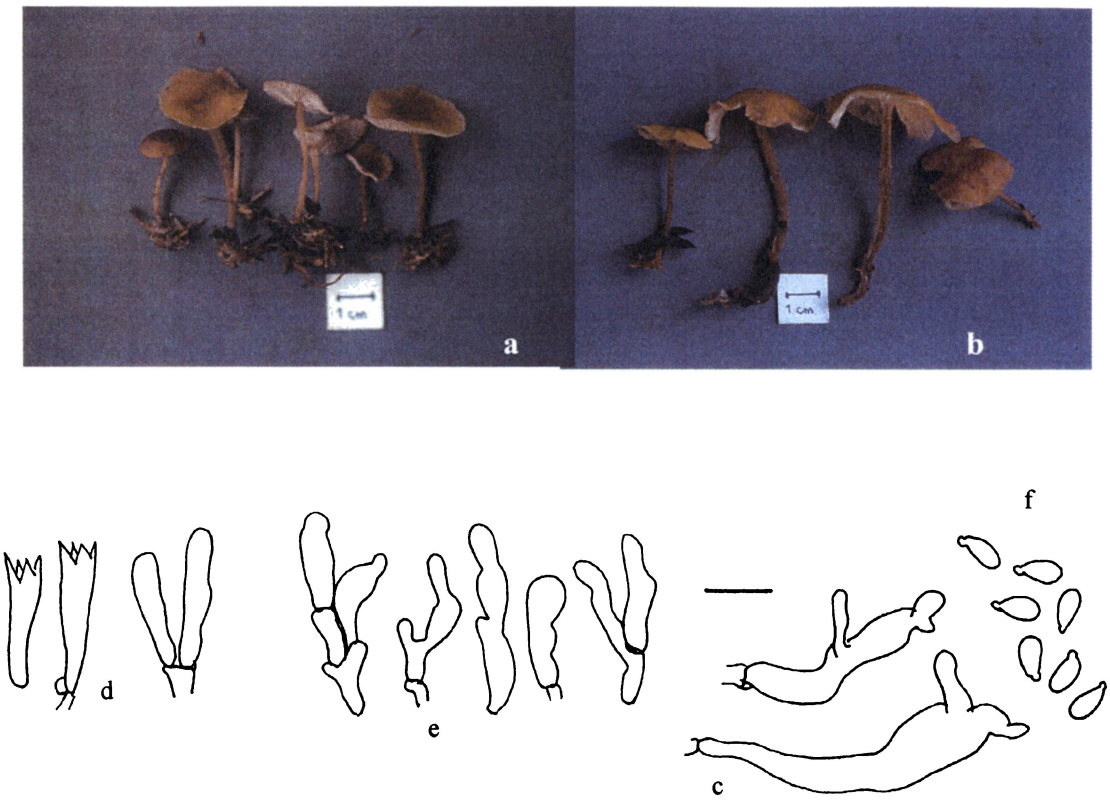


Fig. 15. *Gymnopus impudicus*. a,b. basidiomata (a = TFB 10088, b = TFB 10433); c. elements of pileus epicutis; d. basidia and basidioles; e. cheilocystidia; f. basidiospores. Scale bar equals 10 µm for line drawings.

with such a strong foetid odor. Costa Rican *G. polyphyllus* has more a fenugreek odor and it can be distinguished micromophologically from *G. impudicus* by smaller spores (4.8-6.4 X 2.4-3.6 μm) and long, clavate cheilocystidia (20-42 X 4-8 μm). Other important characteristics in *G. impudicus*, noted in the collections made for this study, were the reddish colors of the pileus that fade to pale brown or white at the margin, lamellae usually white and crowded, and stipes orange and pink to cream colored, sometimes with a black base. Microscopic characteristics observed in Costa Rican specimens, especially spore dimensions and shape of cheilocystidia, were very similar to those present in the neotype specimen. Basidiomata of the neotype specimen have light brown colors and broader lamellae than those of the Costa Rican specimen (dried material compared).

As noted by Antonín & Noordeloos (1997) basidiomata of *G. impudicus* are variable in many morphological traits but easily distinguished from similar species with foetid odor like *G. dysodes*, characterized by a sulcate pileus and distant lamellae. *Gymnopus brassicolens* is recognized by a tapering and blackening stipe and *G. hariolorum* by the pale colored basidiomata (Antonín and Noordeloos, 1997).

Habitat of Costa Rican collections was also very similar to that reported by Antonín & Noordeloos (1997), especially those fruiting at higher elevations in the Talamanca Mountains under *Cupressus* spp, a taxonomic relative of the European *Juniperus*. Basidiomata occur, however, in other kinds of forest associations.

Costa Rican *G. impudicus* appears phylogenetically related to *G. dysodes* and *M. foetidus* (Figs. 2-4). Basidiomata of all three species are reported to have unpleasant to fetid odor (Antonín and Noordeloos, 1997; Halling, 1983). These three species are putatively phylogenetically closely related to *G. fusipes*, the type species of *Gymnopus,* but no distinct morphological characteristic is shared by all of these species.

Mating studies. The self-cross of nine SBIs from collection TFB 10436 representing Costa Rican *G. impudicus* resulted in a tetrapolar mating system (Fig. 16) which confirmed Lamoure's (1989) previous report for this species. All four mating types were identified. Mating types A_1B_2 and A_2B_1 were assigned arbitrarily based on presence of clamp connections in 7/9 X 1/3/4/10/11 and A_1B_1 and A_2B_2 were assigned as subordinates. Intercollection pairings demonstrated that specimens from La Amistad were compatible with each other (Fig. 17).

Material examined: COSTA RICA. Cartago Prov., vic. Estrella, Palo Verde, 9° 46'59" N, 83° 56'71" W, 1700 m, 13.VI.1995, col. RE Halling & RH Petersen, *TFB 7690* (TENN 53782); same location, 28.VI.1998, col. JL Mata, *TFB 9662* (TENN 56623); same location, 10.VIII.1998, col. JL Mata, *TFB 10031* (TENN 56743); same location, 16.VI.1999, col. JL Mata & R Muñoz, *TFB 10079* (TENN 58019). Puntarenas Prov, Coto Brus Co. Sabalito, La Amistad Lodge, Sienaga and Surtuo Trails, 8° 54'22" N, 82° 47'40" W; 1560 m, 3.VII.1998, col. RH Petersen & JL Mata, *TFB 9683* (TENN 56644); same location, 4.VII.1998, col. RH Petersen & JL Mata, *TFB 9687* (TENN 56648), *TFB 9691* (TENN 56652), *TFB 9694* (TENN 56655); same location, 5.VII.1998, col. RH Petersen & JL Mata, *TFB 9697* (TENN 56658) **S**, *TFB 9700* (TENN 56661); same location, 23.VI.1999, col. JL Mata & I González, *TFB 10426* (TENN 58120), *TFB 10428* (TENN 58122); same location, 24.VI.1999, col. JL Mata & I González, *TFB 10433* (TENN 58127), *TFB 10434* (TENN 58128), *TFB 10435* (TENN 58129), *TFB 10436* (TENN 58130), *TFB 10437* (TENN 58131), *TFB 10438* (TENN 58132), *TFB 10439* (TENN 58133). San José Prov., Dota Co., Jardín de Dota, 3.5 km W off Inter American Hwy, 9° 42'42" N, 83° 53'28" W, 2220 m, 13.VII.1993, col. RE Halling, GM Mueller, E Pine, J Carranza, *REH 7008, REH 7010* (NY); same location, 1.VII.1998, col. RE Halling, RH Petersen & JL Mata, *TFB 9676* (TENN 56637); same location, 18.VI.1999, col. JL Mata, *TFB 10088* (TENN 58083), *TFB 10097* (TENN 58092). SWEDEN. Västergötland, Göteborg, Slottsskogen. 4.IX.1942. col. T Nathorst-Windahl, *no 3243* (HT, PRM). UNITED STATES. Michigan, Washtenaw Co., Ann Arbor, 1.X.1936, col. AH Smith, *AHS 4978* (NY). Wisconsin, Sand Co., Aldo Leopold Preserve, near Madison,

			A_1B_2					A_2B_1	A_2B_2	A_1B_1
		7	9	1	3	4	10	11	2	8
	7*		-	+	+	+	+	+	-	-
A_1B_2	9	-		+	+	+	+	+	-	-
	1	+	+		-	-	-	-	-	-
	3*	+	+	-		-	-	-	-	-
	4	+	+	-	-		-	-	-	-
	10	+	+	-	-	-		-	-	-
A_2B_1	11	+	+	-	-	-	-		-	-
A_2B_2	2*	-	-	-	-	-	-	-		+
A_1B_1	8*	-	-	-	-	-	-	-	+	

Fig. 16. Self-cross pairings of SBIs from *Gymnopus impudicus* TFB 10436; + = presence of clamp connections; - = absence of clamp connections; * = tester strains. Isolate numbers and assigned mating types top and side.

		9687								
La Amistad	9687		9694							
La Amistad	9694	**4/4**		9697						
La Amistad	9697	0/4	**4/4**		9700					
La Amistad	9700	**4/4**	**4/4**	0/4		10031				
Palo Verde	10031	**4/4**	0/4	0/4	**1/4**		10433			
La Amistad	10433	x	x	x	x	x		10435		
La Amistad	10435	x	x	x	x	x	**4/4**		10436	
La Amistad	10436	x	x	x	x	x	**2/4**	**3/4**		10437
La Amistad	10437	x	x	x	x	x	**2/4**	**4/4**	**3/4**	

Fig. 17. Intercollection pairings from *Gymnopus impudicus*. Numbers indicate ratio of pairings where clamp connections were observed; x = no pairing was performed. All locations are from the Talamanca Mountains.

22.VI.1991, col. RE Halling, *REH 6570* (NY).

Gymnopus dysodes (Halling) Halling. [1997. Mycotaxon 63: 364.] UNITED STATES. New York, Bronx Co., NY Botanical Garden, 23.VIII.1984, col. RE Halling, *REH 3806* (NY); Tennessee, Knox Co., UTK campus, 4.IX.2001, col. JL Mata, *TFB 11040* (TENN 59141) **S**. Virginia, Hanover Co., near Montpelier, Rte. 671, 0.5 miles S of Rte 54, IX.1999, col. E Lickey, *TFB 10474* (TENN 58367).

Gymnopus fusipes (Bull.: Fr.) S.F. Gray. [1821. Nat. Arr. Brit. Plants 1: 604.] AUSTRIA. Vic. Vienna, Lower Austria, Dist. Hainburg, south Wolfstahl Wald, 48° 07' N, 7° 17' E, 23.IX.2001, col. RH Petersen, *TFB 11439* (TENN 59300) **S**. FRANCE. Rhone-Alpes, Dept. Savoire, 10.IX.2001, col. E Grand, *TFB 11333* (TENN 59217).

Marasmiellus foetidus (Sow.: Fr.) Antonín, Halling & Noordeloos. [1997. Mycotaxon 63: 366.] AUSTRIA. vic. Vienna, Lower Austria, Dist. Hainburg, south Wolfstahl, Wangheimer Wald, 48° 07' N, 17° 00' E, 23.IX.2001, col. H Vogelmayr, *TFB 11434* (TENN 59294) **S**.

Section *Levipedes*

Type species: *Gymnopus dryophilus*.

Pileus epicutis composed of short, branched, not radially arranged hyphae.

(10) *Gymnopus dryophilus* (Bull.: Fr.) Murrill. 1916. N. Amer. Flora 9: 362. Figs. 18-20

≡ *Agaricus dryophilus* Bull.: Fries. 1821. Syst. Mycol. 1: 124.

≡ *Omphalia dryophilus* (Bull.: Fr.) S.F. Gray. 1821. Nat. Arr. Br. Pl.: 612.

≡ *Collybia dryophila* (Bull.: Fr.) Kummer. 1871. Führ. Pilzk. 115.

≡ *Marasmius dryophilus* (Bull.: Fr.) Karsten. 1889. Krit. Ofvers. Finlands Basidsvamp. 103.

≡ *Collybidium dryophilum* (Bull.: Fr.) Murrill. 1911. Mycologia 3: 101.

NEOTYPE: SWEDEN. Uppsala, Uppsala city park (Stadskogen), 17.VI.1984, col. R Vilgalys & S Ryman *Vilgalys 84/181*, (VPI) *teste* Halling 1996 *n.v.*

Pileus 10-45 mm diam, convex, plano-convex to irregularly applanate or uplifted; surface glabrous to finely appressed-fibrillose, shiny, ± lubricous to the touch, at center orange (5/6C8), orange brown (6E8-7E7, 7E8, 6D7/6), dark brown (7E7) or butter yellow (4A5), outwards pale brown (6C/D6/5), pale orange (5C7 - 5D7, 5B4, 4A4) or cream (2/3A2); margin even, uplifted or undulating, superficially striate, translucent; context white or concolorous to surface, unchanging. **Lamellae** adnexed to seceding, ± 1-3 mm broad, close to crowded, at first white, with age pale yellow (2A2), cream yellowish (3/4A3), yellowish (2A4-3A4) to cream (4/5A2); margin even; lamellulae in two to three tiers. **Stipe** 20-75 X 3-6 mm, terete to clavate, base sometimes bulbous and bent, compressed to somewhat cleft, twisted; surface glabrous, base sometimes strigose, at apex almost white, pale yellow (3A4) to yellow brown, towards base orange (4B8-5D6), brown orange (5C7-5B6, 6B/C8), sometimes with yellowish tints; interior hollow; consistency fleshy. Rhizomorphs at base and on substratum, white, orange cream, pale ochraceous buff, or yellowish. Odor not distinctive, mild, pleasant to floral; taste mild, fungous to ± acrid. **Habitat** on leaf litter, woody debris, and grassy patches near forest; grouped to cespitose (Fig. 18a,b).

Pileus epicutis a cutis; hyphae 2-16 µm diam, not cylindrical, frequently branched, not radially oriented, occasionally with diverticula, lightly pigmented or not, inamyloid, with clamp connections; wall thin. **Pileus trama** interwoven; hyphae 6-14 µm diam, hyaline, inamyloid to brick orange in IKI, with clamp connections; wall thin. **Lamellar trama** regular to subregular; hyphae 6-16 µm diam, hyaline, inamyloid to brick orange in IKI, with clamp connections; wall thin; subhymenium parenchymatous. **Basidia** (Fig. 18c) 24-28 X 5-7 µm, clavate, sterigmata four. **Pleurocystidia** absent. **Cheilocystidia** (Fig. 18d) 24-32 X 5-8 µm, clavate to cylindrical, furcate or not, with clamp connections; apex obtuse, knobbed or with diverticula. **Stipe epicutis** parallel; hyphae 3-10 µm diam, inamyloid; wall thin. **Caulocystidia** 24-60 X 3-9 µm, cylindrical to clavate, flexuous or contorted, single or entangled in fascicles; common to uncommon.

Basidiospores (Fig. 18e) 4.8-6.8 X 2.4-3.6 μm (n = 140/7, x = 5.6 X 3.0 μm, Q = 1.56-2.29, Qx = 1.87) lacrymoid in side view, ellipsoid to obovoid in face view, hyaline, inamyloid; wall smooth, thin.

Commentary: *Gymnopus dryophilus* is a commonly collected agaric and therefore is well known and well documented. There seems to be some variation in the color of basidiomata as well as in the shape of the base of the stipe. In the Talamanca Mountains of Costa Rica, the only other taxon that is morphologically similar is *G. macropus*, which can be distinguished by a much more slender stipe and brighter orange tints.

Similarly colored basidiomata of North American *G. subsulphureus,* and European *G. aquosus* can be distinguished from those of *G. dryophilus* by the pinkish colored rhizomorphs and clavate to sphaeropedunculate cheilocystidia respectively (Antonín and Noordeloos, 1997; Vilgalys and Miller, 1983, 1987a). *Gymnopus dryophilus* seems to be a cosmopolitan species (Antonín & Noordeloos, 1997; Dennis, 1970; Halling, 1983; Pegler, 1983b). Description of Colombian specimens (Franco-Molano et al, 2000) is very similar to that reported here. In oak forests of the Talamanca Mountains this species is conspicuous in most sites, and seems to occur in a wide range of altitudes, in comparison to other taxa within *Gymnopus*.

Costa Rican *G. dryophilus* seems to be distantly related to that of North America in all phylogenetic analyses (Figs. 2-4). The sequence for *G. dryophilus* deposited in GenBank (by others) is more similar to that of *G. nubicola* than to Costa Rican *G. dryophilus*. Pairwise sequence comparison of Costa Rican *G. dryophilus* with that deposited in GenBank resulted in 96% similarity.

Mating studies. A self-cross with 12 SBIs from collection TFB 11015 resulted in a tetrapolar mating system (Fig. 19), as previously reported for this species (Lamoure, 1989). All four mating types were identified. Mating types A_1B_2 and A_2B_1 were assigned



Fig. 18. *Gymnopus dryophilus*. a,b. basidiomata (a= TFB 10081, b = REH 7901); c. basidium and basidioles; d. cheilocystidia; e. basidiospores. Scale bar equals 10 µm.

arbitrarily based on clamp formation between 1/3/4/6/14 X 5/8/15 and mating types A_1B_1 and A_2B_2 were assigned as subordinates. SBIs of collections from the Talamanca Mountains were intercompatible in different percentages, which can be interpreted as a well-spread biological species, demonstrated also for North American and European populations (Vilgalys and Miller 1983, 1987b). Collections TFB 9647 (Bosque del Niño, Central Volcanic Mountains) and TFB 10081 (El Jaular) were incompatible with all SBIs from other collections (Fig. 20) but were not crossed to test if they formed an intersterility group. Morphological examination of the corresponding voucher specimens did not reveal any differences from other specimens within *G. dryophilus*. No intercollection pairings with collections of *G. dryophilus* from other regions of the world were attempted.

Material examined: COSTA RICA. Puntarenas Prov., Coto Brus Co., Sabalito District, Hacienda La Amistad, 8° 54'22" N, 82°47'40" W; 1330 m, 3.VII.1998, col. RH Petersen & JL Mata, *TFB 9684* (TENN 56645); same location, 3.VII.1998, col. RH Petersen, *TFB 9686* (TENN 56647). San José Prov., Dota Co., Jardín, 3.5 Km E of Interamerican Highway at El Empalme, 9° 42'52" N, 83° 58'28" W, 2220 m, 1.VII.1998, col. RH Petersen, *TFB 9674* (TENN 56635); same location, 18.VI.1999, col. JL Mata, *TFB 10092* (TENN 58087); same location, 25.VI.2000, col. RH Petersen & JL Mata, *TFB 11015* (TENN 58623) **S**, *TFB 11017* (TENN 58625); same location, 2.VI.2001, col. RE Halling, B Buyck, R Aldana-Gómez, *REH 8091* (NY); La Chonta, S of Interamerican Highway toward Laguna/Cerro Chonta, 9° 41'56" N, 83° 56'31" W, 2400 m, 11.VI.2001, col. RE Halling, B Buyck, R Aldana-Gómez *REH 8176* (NY); Finca El Jaular, forest patch along creek from main house, 9° 39'39" N, 83° 52'01" W, 2300 m, 17.VI.1999, col. JL Mata, *TFB 10081* (TENN 58021). PANAMA. Chiriquí Prov., Bugaba, Cerro Punta, Parque Internacional La Amistad, Sendero Retoño, 8° 51' N, 82° 34' W, 2280 m, 20.X.1999, col. RE Halling, Duguay & Gonzalez, *REH 7901* (NY). UNITED STATES. North Carolina, Jackson Co., vic. Cashiers, 12.VII.1999, col. JL Mata, *TFB 10456* (TENN 58150). South Carolina, Oconee Co., vic Walhalla, 13.VII.1999, col. JL Mata, *TFB 10461* (TENN 58155). Tennessee, Sevier Co., GSMNP, Sugarlands Visitor Center,

						A1B2			A_2B_1		A_1B_1	A_2B_2
		1	3	4	6	14	5	8	15	2	12	7
	1*		-	-	-	-	+	+	+	-	-	-
	3	-		-	-	-	+	+	+	-	-	-
	4	-	-		-	-	+	+	+	-	-	-
	6	-	-	-		-	+	+	+	-	-	-
A1B2	14	-	-	-	-		+	+	+	-	-	-
	5*	+	+	+	+	+		-	-	-	-	-
	8	+	+	+	+	+	-		-	-	-	-
A_2B_1	15	+	+	+	+	+	-	-		-	-	-
	2*	-	-	-	-	-	-	-	-		-	+
A_1B_1	12	-	-	-	-	-	-	-	-	-		+
A_2B_2	7*	-	-	-	-	-	-	-	-	+	+	

Fig. 19. Self-cross pairings of SBIs from *Gymnopus dryophilus* TFB 11015. + = presence of clamp connections; - = absence of clamp connections; * = tester strains. Isolate numbers and assigned mating types top and side.

	9647		9674					
Bosque del Niño	9647		9674					
Jardín de Dota	9674	0/4		9684				
La Amistad	9684	0/4	0/4		9686			
La Amistad	9686	0/4	2/4	2/4		10081		
El jaular	10081	-	0/4	0/4	0/4		10092	
Jardín de Dota	10092	-	-	2/4	2/4	-		11015
Jardín de Dota	11015	-	-	2/4	-	-	4/4	

Fig. 20. Intercollection pairings from *Gymnopus dryophilus*. - = no pairing was performed. Numbers indicate ratio of pairings where clamp connections were observed. All locations are from the Talamanca Mountains except for Bosque del Niño.

24.V.1998, col. JL Mata, *TFB 9604* (TENN 56323).

Gymnopus fagiphilus (Velen.) Antonín, Halling & Noordeloos. [1997. Mycotaxon 63: 364.] AUSTRIA. Vic. Vienna, Lower Austria, Dist. Hainburg, south Wolfstahl Wald, 48° 07' N, 7° 17' E, 23.IX.2001, col. H Vogelmayr, *TFB 11438* (TENN 59299) **S.**

Gymnopus erythropus (Pers.: Fr.) Antonín, Halling & Noordeloos. [1997. Mycotaxon 63: 364.] AUSTRIA. Vienna, Dist 2, Maurewald, 48° 09' N, 16° 15' E, col. H. Vogelmayr, *TFB 11470* (TENN 59329) **S.**

Gymnopus ocior (Pers.) Antonín & Noordeloos. [1997. Mycotaxon 63: 365.] FRANCE. Rhone-Alpes, Dept. Savoire (73), St. Jean de Chevelu, 45° 42' N, 5° 48' E, 10.IX.2001, col. RH Petersen, *TFB 11329* (TENN 59213) **S.**

(11) *Gymnopus macropus* Halling. 1996. Brittonia 48: 490. Fig. 21
HOLOTYPE: COLOMBIA. Antioquia, Guarne, 14 km E of Medellín, Centro Experimental Piedras Blancas, 15.V.1987, col. RE Halling, *REH 5263* (NY) [!].

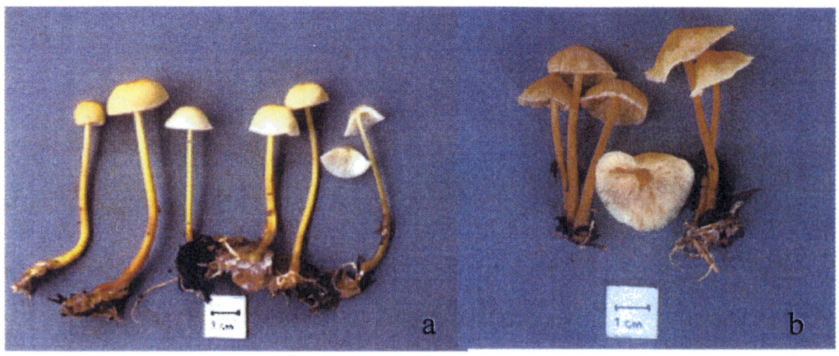
Pileus 14-45 mm diam, at first convex to subumbonate, with age broadly convex to applanate, or shallowly depressed; surface glabrous to fibrillose, hygrophanous, outwards becoming striate-translucent, at center orange (6D8, 6D7, 5C8), gray orange (6B5/6), yellow orange (4B8), outwards pale orange (5A5/4), with age cream to off white; margin curved, striate or rugulose, translucent, cream buff (4A2/4); context thin, concolorous to surface. **Lamellae** adnate to adnexed, sometimes seceding, up to 3 mm broad, close to crowded, off white, cream (2A2), yellowish (3/4A2), pale pinkish cinnamon (6A2) to pinkish buff (6A3); margin even; lamellulae in several tiers of different lengths, sometimes anastomosing. **Stipe** 40-130 X 2-4 mm, terete, equal to compressed-sulcate at center, base sometimes widened into a semi-bulb; surface glabrous to fibrillose, at base sometimes hirsute, near apex yellowish (3B6), pale orange yellow (4A4) to brown orange (6B7-6C7), near base orange (5A8), mikado orange (6A6), orange brown (6D8), at base dark orange (5B8) to orange rufous (7B7); interior hollow; consistency fleshy to cartilaginous. Mycelium at base and substratum white to cream

(4A2), yellowish, pale ochraceous salmon (3A3) to warm buff (5A4). Odor pleasant, sweetish, spicy or fungous; taste not distinctive to ± raphanoid. **Habitat** on leaf litter and soil; gregarious to cespitose (Fig. 21a,b).

Pileus epicutis a cutis; hyphae 3-8 (-12) μm diam, not cylindrical, frequently branched, occasionally with diverticula, not radially oriented, pale yellow in mass, hyaline singly, inamyloid, with clamp connections; wall thin. **Pileus trama** loosely interwoven; hyphae 2-11(-14) μm diam, hyaline, inamyloid, with clamp connections; wall thin to 0.8 μm thick. **Lamellar trama** subregular; hyphae 2-8 μm diam, hyaline, inamyloid, with clamp connections; wall thin. **Basidia** (Fig. 21c) 20-26 X 5-7 μm, clavate; sterigmata four; basidioles 16-26 X 5-6 μm, clavate. **Pleurocystidia** absent. **Cheilocystidia** (Fig. 21d) 12-32 X 4-12 μm, broadly clavate, some furcate or sphaeropedunculate, with clamp connections; apex obtuse or occasionally with diverticula. **Stipe epicutis** parallel; hyphae 2-12 μm diam, frequently septate, pale yellow in mass, hyaline singly, inamyloid, with clamp connections; wall thin to 1 μm thick. Gloeopleurous hyphae rare, up to 8 μm diam. **Caulocystidia** 12-48 X 4-9 μm, clavate to subglobose, occasionally lobed or with diverticula, semierect to erect, single or in fascicles; wall up to 0.8 μm thick.

Basidiospores (Fig. 21e) 6.0-8.4 X 3.2-4.0 μm (n = 80/4, x = 7.2 X 3.5 μm, Q = 1.60-2.57, Qx = 2.05), lacrymoid to pip shape in side view, ellipsoid in face view, hyaline, inamyloid; wall smooth, thin.

Commentary: Basidiomata of *G. macropus* are very similar to those of *G. dryophilus*. One important difference is that stipes in *G. macropus* tend to be more slender, and longer in proportion to the pileus diameter, in contrast to those of *G. dryophilus* that sometimes present a clavate to bulbous, fistulose base. Microscopically, spores measurements are consistently 1-2 μm longer than those of *G. dryophilus*. Cheilocystidia in *G. macropus* are variable, tending to wear off at the apex in aging basidiomata, but are more broadly clavate than those found in *G. dryophilus*.



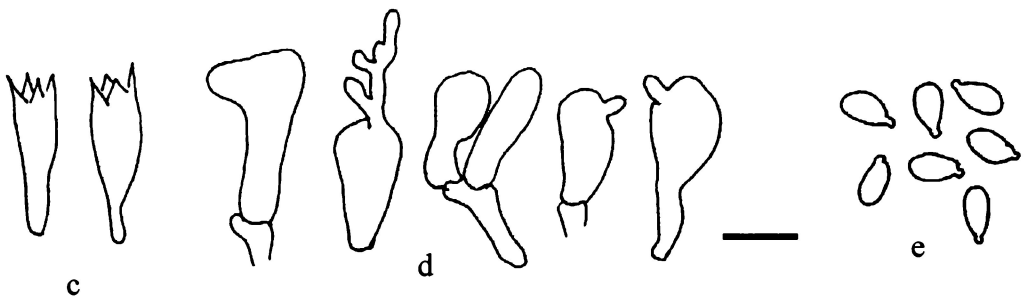


Fig. 21. *Gymnopus macropus*. a,b. basidiomata (a = TFB 10095, b = TFB 11024); c. basidia; d. cheilocystidia; e. basidiospores. Scale bar equals 10 µm for line drawings.

Gymnopus macropus appears to be more related to *G. dryophilus* and allied species than to *G. spongiosus* (Figs. 2-4). The sequence of *G. macropus* is 95% similar to that of Costa Rican *G. dryophilus*.

Material examined: COLOMBIA. Antioquia: Guaren, 14 Km E of Medellín, Centro Experimental Piedras Blancas, 15.V.1987, col. RE Halling, *REH 5263* (NY, HT). COSTA RICA. Puntarenas Prov., Coto Brus Co., vic. San Vito de Coto Brus, Finca Las Alturas, NW boundary w/buffer zone of Parque La Amistad, 8° 47' N, 82° 54' W, 1550 m, 8.XI.1989, col. RE Halling *REH 6339* (NY); same location, on the trail to Cerro Chia; 1520 m, 21.VI.2000, col. RH Petersen & JL Mata, *TFB 10490* (TENN 58598); Sabalito Co., Estación Biológica Las Alturas, 8° 56'59" N, 82° 50'02" W, 1420 m, 25.V.1994, col. E Franco-Molano, M Mata, J Johnson, L Umaña, *EFM 1188* (NY); Hacienda La Amistad, Higueron trail, 8° 54'22" N, 82° 47'40" W; 1330 m, 4.VII.1998, col. RH Petersen & JL Mata, *TFB 9693* (TENN 56654); San José Prov., Dota Co., Jardín de Dota, 3.5 Km W of Interamerican Highway at Empalme. 9° 42'52" N, 83° 53'28" W, 2220 m, 5.VI.1996, col. RE Halling, J Ammirati & J Torres, *REH 7616* (NY); same location, 1.VII.1998, col. RE Halling, RH Petersen & JL Mata, *TFB 9675* (TENN 56636); same location, 18.VI.1999, col. JL Mata, *TFB 10095* (TENN 58090) **S**; same location, 25.VI.2000, col. RH Petersen & JL Mata, *TFB 11011* (TENN 58619); Perez Zeledón Co., Villa Mills, Estación Experimental CATIE, 9° 33'03" N, 83° 40'56" W, 2880 m, 22.VI.1995, col. RH Petersen, *TFB 7911* (TENN 53806); Estación Biológica Cuericí, 9° 33'30" N, 83° 40'00" W, 2650 m, 27.VI.2000, col. RE Halling, RH Petersen, L Norvell & JL Mata, *TFB 11024* (TENN 58632).

(12) *Gymnopus nubicola* Halling. 1996. Brittonia 48: 492. Fig. 22

HOLOTYPE: ECUADOR. Pichincha/Napo: Nr. Papallacta, along rd. from Quito to Papallacta, páramo, 30.IV.1987, ± 4000 m, 00° 19' N, 78° 11' W, col. RE Halling, *REH 5219* (NY) [!].

Pileus 25-50 mm diam, convex to plano-convex, sometimes uplifted; surface glabrous, subviscid, reddish brown (8F8), brown (7E8) to brownish yellow (5C-D7); context white, unchanging. **Lamellae** adnexed, close, white to yellowish white, 4A2; margin even to uneven. **Stipe** 30-60 X 2-8 mm, terete to compressed, equal to slightly broader below; surface glabrous, moist to dry, pale yellow above, orange below; interior becoming hollow; basal rhizomorphs pale pinkish buff. Odor and taste mild. **Habitat** on soil, humus, in páramo mixed with bamboo; gregarious (Fig. 22a).

Pileus epicutis a cutis; hyphae 4-12 μm diam, not cylindrical, frequently branching, not radially arranged, sometimes with knobs, pale tan in mass, inamyloid, with clamp connections; wall thin. **Pileus trama** ± radially oriented and somewhat interwoven; hyphae 3-12 μm diam, hyaline, inamyloid, with clamp connections; wall thin. **Lamellar trama** regular; hyphae 2-14 μm, hyaline, inamyloid, with clamp connections; wall thin. **Basidia** (Fig. 22b) 18-23 X 5-6 μm clavate; sterigmata four; basidioles 16-23 X 4-6 μm, clavate to cylindrical. **Pleurocystidia** absent. **Cheilocystidia** (Fig. 22c) 28-36 X 4-19 μm, cylindrical to broadly clavate; apex typically branched and diverticulate; wall thin. **Stipe epicutis** parallel; hyphae 3-20 μm diam, innermost hyphae broader, pale tan in mass, hyaline singly, inamyloid, with clamp connections; wall thin. **Caulocystidia** 24-50 X 4-9 μm, cylindrical to clavate, occasionally strangulate, erect to semi-erect; uncommon.

Basidiospores (Fig. 22d) 4.8-5.6 X 2.4-3.6 μm (n = 20, x = 5.4 X 3.1 μm, Q = 1.50-2.33, Qx = 1.76), ellipsoid in side view and face view, hyaline, inamyloid; wall smooth, thin. Spore print white to cream (2A2).

Commentary: *Gymnopus nubicola* is a species described from the páramo vegetation of the Ecuadorian Andes. Basidiomata are similar to those of *G. dryophilus* and especially *G. subsulphureus*, because of the pinkish color of the rhizomorphs (Vilgalys and Miller, 1983), and different from other members in the section by having a



Fig. 22. *Gymnopus nubicola*. a. basidiomata (REH 8290); b. basidia and basidioles; c. cheilocystidia; d. basidiospores. Scale bar equals 10 μm for line drawings, 1 cm for picture.

negative reaction to alkali. Basidiomata of *G. nubicola* exhibit more reddish tints in their pilei, whereas those of *G. dryophilus* are yellower. Spore and cheilocystidia measurements are somewhat smaller than those of South American specimens reported by Halling (1996a). All macro- and microscopical characteristics as well as habitat observed in the sole Costa Rican collection match with those of the type.

The sequence of *G. nubicola* is 99% similar to one representing *G. dryophilus* deposited in GenBank (AF079580) and 96% similar to that of Costa Rican *G. dryophilus*. *Gymnopus nubicola* appears phylogenetically related to European *G. fagiphilus* and *G. ocior* (Figs. 2-4).

Material examined: COSTA RICA. San José Prov., Cerro de la Muerte, 9° 40" N, 83° 45" W, 3491 m, 17. XI.2001, col. RE Halling & Kirchgessner, *REH 8290* (NY) **S.** ECUADOR. Pichincha/Napo: Nr. Papallacta, along rd. from Quito to Papallacta, páramo, 30.IV.1987, ± 4000 m, 00° 19' N, 78° 11' W, col. RE Halling, *REH 5219* (NY, HT).

(13) *Gymnopus spongiosus* (Berk. & Curt.) Halling. 1996. Brittonia 48: 489. Figs. 23,24

≡ *Marasmius spongiosus* Berkeley & Curtis. 1849. Hooker's J. Bot. Kew Gard. Misc. 1: 100.

≡ *Collybia spongiosa* (Berk. & Curt.) Singer. 1949. Lilloa 22: 201.

HOLOTYPE: UNITED STATES. South Carolina, VI.1847, *Curtis 1257*, (K), *teste* Halling 1996; *n.v.*

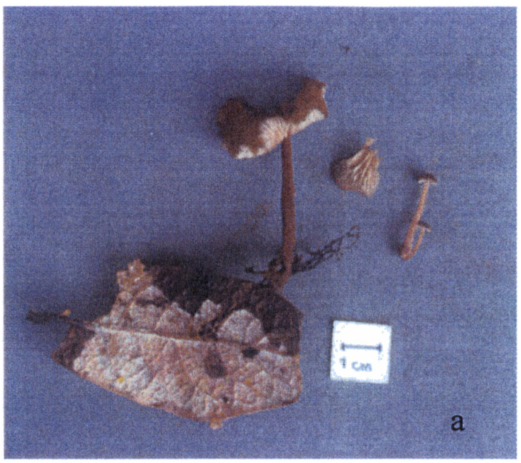
Pileus 5-30 mm across, planate to shallowly depressed, or ± umbonate; surface glabrous, shiny, disc at first reddish brown (7F8-7E8), outwards with age brown (7D7-6E7); margin convex and smooth at first, whitish, uplifted and undulating with age; context thin. **Lamellae** adnexed at first, free with age, ± narrow to 2 mm, subdistant, at first off-white, with age cream (4A3); margin even; lamellulae in one to two tiers. **Stipe** 15-50 X 1-3 mm, ± equal; surface covered with a vesture of hairs, more densely so

towards base; hairs yellowish; surface at first brown (6D7-6E7), with age reddish brown (8E8); interior hollow; consistency brittle. Mycelium at base and substrate cream colored. Odor and taste not recorded. **Habit** on decayed leaves, gregarious (Fig. 23a).

Pileus epicutis (Fig. 23b) a cutis; hyphae not cylindrical, frequently branched, not radially oriented, some with incrusted pigment, yellow-brown in mass, hyaline singly, inamyloid, with clamp connections; wall thin. **Pileus trama** interwoven; hyphae 4-12 μm diam, hyaline singly, inamyloid; walls thin. **Lamellar trama** regular to subregular; hyphae 2-10 μm diam, hyaline, with clamp connections; wall thin; subhymenium 10-12 μm wide. **Basidia** (Fig. 23c) 24-40 X 5-9 μm, clavate; sterigmata four. **Pleurocystidia** absent. **Cheilocystidia** (Fig. 23d) obscure, 14-36 X 5-10 μm, clavate to ampullaceous, some with outgrowths or knobbed; wall thin. **Stipe epicutis** parallel; hyphae 2-20 μm, cylindrical, yellowish in mass, hyaline singly, with clamp connections; wall up to 4 μm thick. **Caulocystidia** abundant, 60-110 X 6-8 μm, cylindrical to occasionally clavate, hyaline, with clamp connections. All tissues turning light green in KOH.

Basidiospores (Fig. 23e) 6.4-8.8 X 3.6-4.4 μm (n = 25; x = 8.0 X 4.1 μm, Q = 1.73-2.20, Qx = 1.96), pip-shaped to lacrymoid in side view, ellipsoid to ovoid in face view, hyaline, inamyloid; wall smooth, thin; hilar appendix visible.

Commentary: this species is recognized by its green reaction to alkali in the tissues, especially those of the stipe, in combination with the "dryophila" type epicutis. It is different from *G. fuscopurpureus*, reported in México and western United States (Halling, 1990), in that it lacks heavy granular pigmentation in the hyphae of the pileus epicutis. *Gymnopus spongiosus*, a species originally described from North America, has been also recently reported from oak forests in Colombia (Franco-Molano et al, 2000; Halling, 1989).



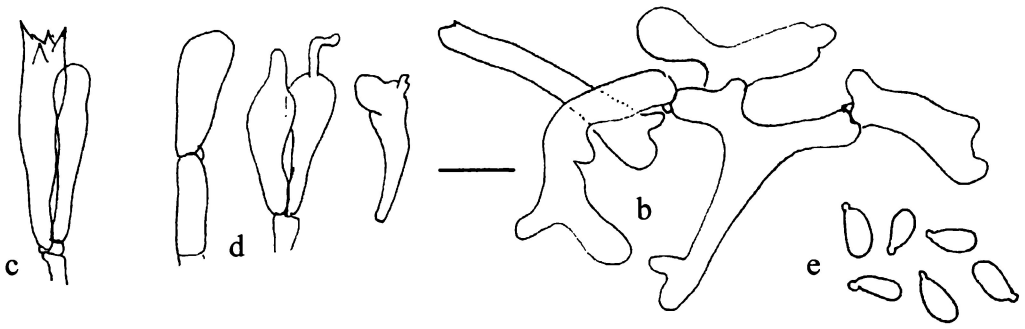


Fig. 23. *Gymnopus spongiosus*. a. basidiomata (TFB 11025); b. elements of pileus epicutis; c. basidia and basidioles; d. cheilocystidia; e. basidiospores. Scale bar equals 10 μm for line drawings.

The sequence of Costa Rican *G. spongiosus* is almost identical (98% similarity) to the one from the United States and is related phylogenetically with a sequence of *G. erythropus* from France (Figs. 2-4).

Mating studies. A self-cross with 12 SBIs from the sole Costa Rican collection of *G. spongiosus* TFB 11025 resulted in a tetrapolar mating system (Fig. 24). This is the first report of the mating system for this particular species, communicated by Mata & Petersen (2001b). Three mating types were identified. Mating types A_1B_2 and A_2B_1 were assigned arbitrarily based on clamp connection formation between 7/11/17/22/35 X 4/32 and the third mating type was assigned as a subordinate. Other members in section *Levipedes* exhibit a tetrapolar mating system (see Petersen, 1995a).

Material examined: COSTA RICA, San José Prov., Pérez Zeledón Co., Villa Mills, Estación Experimental CATIE, 9° 33'03" N, 83° 40'56" W, 2880 m, 27.VI.2000, col. JL Mata, RH Petersen, RE Halling & L Norvell, *TFB 11025* (TENN 58633) **S.** UNITED STATES. Wisconsin, Sand Co., Leopold Memorial Reserve, 23.VI.1990, col. RH Petersen, *TFB 2887* (TENN 49077) **S.**

Section *Peronati* (Kühner) Mata, *comb. nov.*

≡ *Marasmius* sect. *Peronati* Kühner. 1933. Botaniste. 25: 85.

Type species: *Gymnopus peronatus* [NEOTYPE: UNITED KINGDOM. Yorkshire, Halifax, Elland Park Wood, 7.IX.1996, A Leonard, E.] *n.v*

Pileus epicutis composed of cylindrical, radially oriented hyphae; pleurocystidia present, conspicuous or inconspicuous.

(14) *Gymnopus lodgeae* (Singer) Mata, *comb. nov.* Figs. 25-27

≡ *Collybia lodgeae* Singer. 1989. Fieldiana, Bot. 21: 23.

HOLOTYPE: COSTA RICA. Heredia Prov., Sarapiquí, Puerto Viejo de Sarapiquí, OTS-La Selva Biological Station, Research Trail, 23.XII.1979, col. J Lodge *CR 294*, (F) [!].

						A_1B_2		A_2B_1		A_1B_1	$A_2B_1 + A_2B_2$
		7	11	17	22	35	4	32	28	36	24
	7*		-	-	-	-	+	+	-	-	-
	11	-		-	-	-	+	+	-	-	-
	17	-	-		-	-	-	+	-	-	-
	22	-	-	-		-	+	+	-	-	-
A_1B_2	35	-	-	-	-		-	+	-	-	+
	4*	+	+	-	+	-		-	-	-	-
A_2B_1	32	+	+	+	+	+	-		-	-	-
	28*	-	-	-	-	-	-	-		-	+
A_1B_1	36	-	-	-	-	-	-	-	-		+
$A_2B_1 + A_2B_2$	24	-	-	-	-	+	-	-	+	+	

Fig. 24. Self-cross pairings of SBIs from *Gymnopus spongiosus* TFB 11025. + = presence of clamp connections; - = absence of clamp connections; * = tester strains. Isolate numbers and assigned mating types top and side. SBI no 24 may be amphithallic or the product of two germinating spores.

Pileus 20-80 mm diam, at first convex-campanulate and umbonate, with age broadly convex-campanulate to applanate; surface glabrous to velutinous, hygrophanous, sulcate halfway toward margin or entirely, brown (7E8, 6E7), when hygrophanous light grayish brown (5B4, 5D6) to beige, at umbo brown (6/F8) and always remaining darker than disc; margin at first incurved, then curved, crenate to sulcate, translucent; context thin, cream or concolorous to pileus. **Lamellae** adnexed to adnate, up to 6 mm broad, close to subdistant, yellowish (3A2-4A2) to creamy gray (4A2-5B3); margin even or finely serrulate; lamellulae in two to three tiers of different lengths. **Stipe** 50-110 X 4-7 mm, cylindrical, equal, ribbed longitudinally, sometimes with widened, twisted base; surface fibrillose, hygrophanous, upwards brown (7E8), orange brown (5C5), tan (4A3) or concolorous to pileus surface, downwards dark brown (6F8), with age brown black; interior hollow; consistency fibrous-fleshy, but brittle. Basal mycelium white. Odor not distinctive; taste mealy. **Habitat** on wood or humus; solitary to gregarious (Fig. 25a).

Pileus epicutis (Fig. 25b) a cutis; hyphae 2-9(-12) μm diam, cylindrical, repent, rarely with diverticula, radially oriented, pigmented light brown in mass, hyaline singly, inamyloid, with clamp connections; wall thin; terminal cells abundant, cylindrical, contorted or ventricose, erect. **Pileus trama** loosely interwoven, ± radially oriented; hyphae 3-16(-24) μm diam, pale yellow to hyaline in mass, hyaline singly, inamyloid, with clamp connections; wall thin to 1.6 μm thick. Gloeopleurous hyphae occasional, up to 6 μm diam. **Lamellar trama** regular to irregular near pileus trama, interwoven towards margin; hyphae 3-14(-24) μm diam, hyaline, inamyloid, with clamp connections; wall thin. **Basidia** (Fig. 25c) 23-38 X 4-7 μm, clavate, hyaline; sterigmata four; basidioles 24-33 X 3-4 μm, clavate. **Pleurocystidia** (Fig. 25d) numerous, 60-190 X 8-20 μm, fusoid, ventricose, lanceolate, originating from lamellar trama, hyaline; wall smooth, thin. Lamellar margin sterile. **Cheilocystidia** (Fig. 25e) 44-130 X 4-13 μm, fusoid to clavate, or similar in form to pleurocystidia; apex sometimes knobbed or forked; wall thin; numerous. **Stipe epicutis** parallel; hyphae 2-10 μm diam, yellowish in mass, hyaline

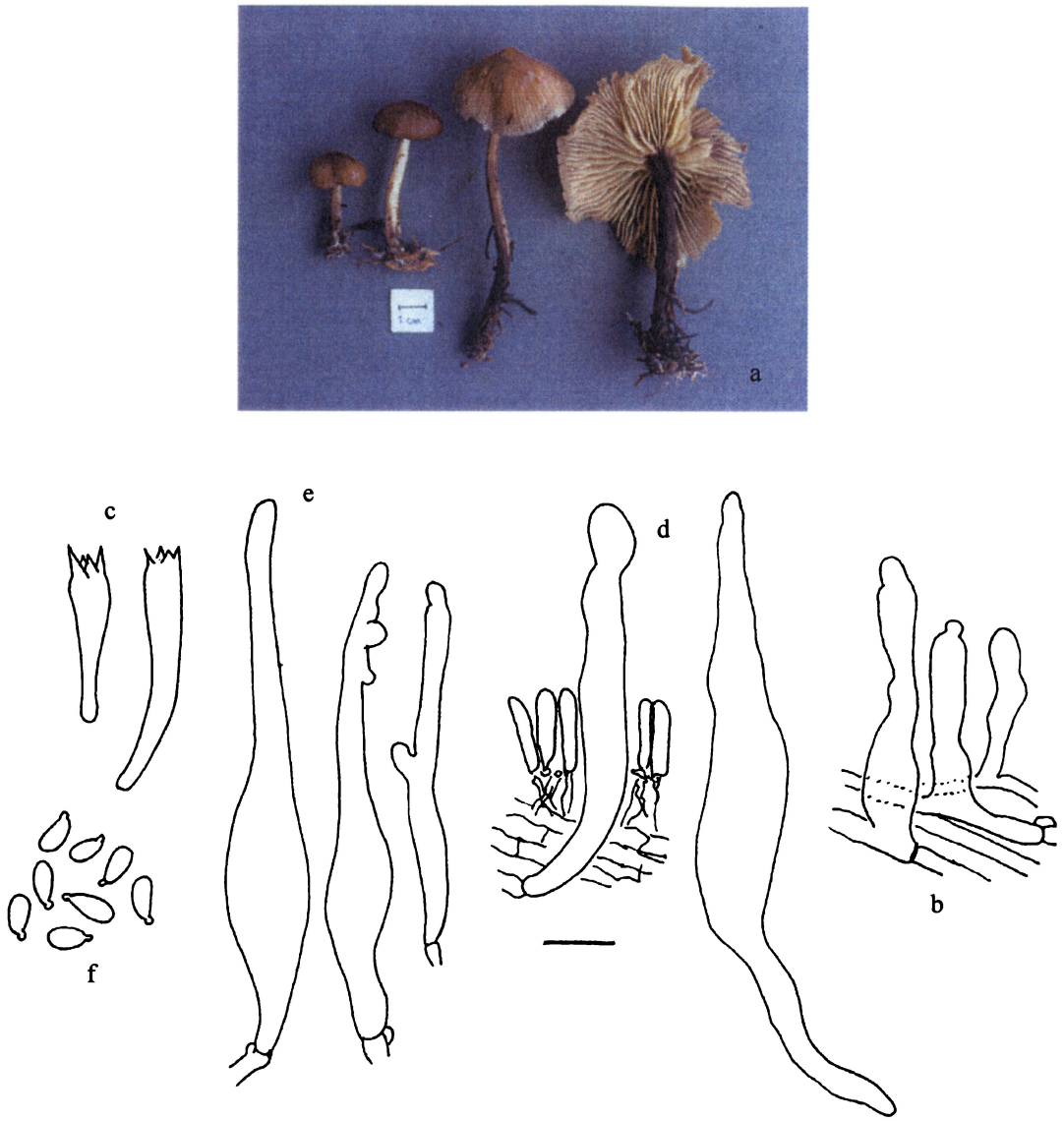


Fig. 25. *Gymnopus lodgeae*. a. basidiomata (TFB 11013); b. elements of pileus epicutis; c. basidia; d. pleurocystidia; e. cheilocystidia; f. basidiospores. Scale bar equals 10 μm for line drawings.

singly, with clamp connections; wall thin to 0.8 μm thick. **Caulocystidia** numerous, 44-92 X 5-8 μm, cylindrical, flexuous, single or in entangled clusters, hyaline.

Basidiospores (Fig. 25f) (5.6-)6.4-8.0(-8.8) X (2.4-)2.8-3.6(-4.0) μm (n = 50/3, x = 6.6 x 3.2 μm; Q = 1.75-2.71, Qx = 2.10), lacrymoid in side view, ellipsoid to subovoid in face view, hyaline, inamyloid; wall smooth, thin.

Commentary: Examination of the type specimen of *G. lodgeae* confirmed morphological identity of basidiomata collected in the Talamanca Mountains during this study. Basidiomata of *G. lodgeae* are recognized by their conical umbo and inrolled pileus margins in early stages, usually together with a twisted, fibrous stipe. These features are found in larger basidiomata of other members in *Gymnopus* and only those of *G. neotropicus* are similar in stature. Pleurocystidia are also present in *G. pseudolodgeae*, *G. omphalodes* and *R. tablensis* that can be distinguished from those of *G. lodgeae* by smaller size (50-68 X 10-17 μm in *G. omphalodes*, 40-80 X 8-12 μm in *G. pseudolodgeae* and 28-84 X 8-16 μm in *R. tablensis*).

Gymnopus lodgeae forms a phylogenetic couplet with *G. pseudolodgeae* (Figs. 2-4). Sequences of the two species are only 89% similar. Also, *G. lodgeae* is phylogenetically closely related to other memebrs in sect. *Peronati*.

Mating studies. A tetrapolar mating system for *G. lodgeae* was reported by Mata & Petersen (2001b) (Fig. 26). Three mating types were identified, based on clamp connection production. Designation of mating types A_1B_2 and A_2B_1 was based on the observation of clamp connections of 5/6/7/9/10/11/13 X 1/4/12/14. Only one SBI could be recovered from the collection from El Jaular and it was crossed with five SBIs from collection TFB 11013 from Jardín de Dota, resulting in partial compatibility (Fig. 27).

Material examined: COSTA RICA. Heredia Prov., Sarapiquí, near Puerto Viejo de Sarapiquí, OTS-La Selva Biological Station, Research Trail, 23.XII.1979, col. J

								A_1B_2	A_1B_1				A_2B_1
		11	13	5	6	10	9	7	2	1	4	12	14
	11		-	-	-	-	-	-	-	+	+	+	+
	13	-		-	-	-	-	-	-	+	+	+	+
	5*	-	-		-	-	-	-	-	+	X	+	+
	6	-	-	-		-	-	-	-	+	+	+	+
	10	-	-	-	-		-	-	-	+	+	+	+
	9	-	-	-	-	-		-	-	+	+	+	+
A_1B_2	7	-	-	-	-	-	-		-	+	+	+	+
A_1B_1	2*	-	-	-	-	-	-	-		-	-	-	-
	1*	+	+	+	+	+	+	+	-		-	-	-
	4	+	+	X	+	+	+	+	-	-		X	-
	12	+	+	+	+	+	+	+	-	-	X		-
A_2B_1	14	+	+	+	+	+	+	+	-	-	-	-	

Fig. 26. Self-cross pairings of SBIs from *Gymnopus lodgeae* TFB 9678. + = presence of clamp connections; - = absence of clamp connections; * = tester strains. Isolate numbers and assigned mating types top and side. X = mycelia did not meet.

		FB 11013				
		2	3	4	6	9
FB 11030	10	+	-	-	X	-

Fig. 27. Intercollection pairings from *Gymnopus lodgeae*. Only one SBI recovered from TFB 11030 (El Jaular) was crossed with five from TFB 11013 (Jardín de Dota); + = presence of clamp connections; - = absence of clamp connections; X = mycelia did not meet.

Lodge, *CR 294* (F). San José Prov., Dota Co., Jardín de Dota, 3.5 km W off Interamerican Hwy at El Empalme, 9° 42'52" N, 83° 53'28" W, 2220 m, 1.VII.1998, col. JL Mata, *TFB 9678* (TENN 56639); same location, 18.VI.1999, col. JL Mata, *TFB 10096* (TENN 58091); same location, col. JL Mata & RH Petersen, *TFB 11013* (TENN 58621) **S**; Finca El Jaular, 29.VI.2000, col. RH Petersen & JL Mata, *TFB 11030* (TENN 58638).

Gymnopus peronatus (Bolt.: Fr.) Antonín, Halling & Noordeloos. [1997. Mycotaxon 63: 365.] AUSTRIA. Vic. Vienna, Lower Austria, Dist. Hainburg, south Wolfstahl, Wangheimer Wald, 48° 07' N, 17° 00' E, 23.IX.2001, col. H Vogelmayr, *TFB 11436* (TENN 59297). FRANCE. Rhone-Alpes, Dept. Savoire (73), Commune St. Germain La Chambotte, 45° 46' N, 5° 53' E, 10.IX.2001, col. PA Moreau, *TFB 11340* (TENN 59219) **S**.

(15) *Gymnopus omphalodes* Mata, *comb. nov.* Figs. 28-30

≡ *Marasmius omphalodes* Berkeley. 1856. Hooker, Journ. Bot. & Kew Misc. 8: 138.

≡ *Collybia omphalodes* (Berk.) Dennis. 1951. Trans. Br. Mycol. Soc. 34: 443.

≡ *Marasmiellus omphalodes* (Berk.) Singer ex Singer. 1973. Beih. Nova Hedw. 44: 118.

HOLOTYPE: BRAZIL. Amazonas, Sao Jeronimo, II. 1853, Spruce 131, (K), *teste* Pegler (1983), *n.v.*

Pileus 15-38 mm diam, at first convex, with age broadly convex, applanate and umbonate, rarely slightly depressed; surface glabrescent to finely fibrillose, entirely crenate, translucent, at umbo brown (6E8-7E8), light brown (6D/E8), outwards pale brown (6C6), cream with faint brownish hues, with age off-white; margin curved, crenate, translucent; context very thin, concolorous to surface. **Lamellae** adnate to free, narrow, crowded to close, white to off-white; margin even; lamellulae in two tiers of different lengths, some anastomosing. **Stipe** 20-60 X 2-4 mm, equal, terete; surface with a fine, white hairy vesture, at apex reddish orange (7E8), brown to buff (6B3), soon reddish brown (7E8-7F8), downwards with age becoming almost entirely black; interior

hollow; consistency ± cartilaginous or ± brittle. Basal mycelium white. Odor pleasant or farinaceous; taste ± acrid. **Habitat** on wood or leaf litter, under *Cupressus spp* or mixed *Quercus spp* vegetation; gregarious (Fig. 28a).

Pileus epicutis a simple cutis; hyphae 4-8(10) µm diam, cylindrical, ± radially oriented, pigment-incrusted, brown in mass, hyaline singly, inamyloid, with clamp connections; wall thin; terminal cells common, cylindrical, prostrate or erect, single or in fascicles, occasionally diverticulate. **Pileus trama** ± loosely interwoven; hyphae 4-16 µm diam, pale yellowish in mass, hyaline singly, inamyloid, with clamp connections; wall thin to 0.8 µm thick. Gloeopleurous hyphae occasional, 22-5 µm diam. **Lamellar trama** subregular to interwoven near margin; hyphae 4-8 µm, hyaline, with clamp connections; wall thin. **Basidia** (Fig. 28b) 18-24 X 3-6 µm, clavate, hyaline, inamyloid; sterigmata four; basidioles numerous, similarly sized and shaped as basidia or submucronate. **Pleurocystidia** (Fig. 28c) 50-68 X 10-17 µm, ventricose, fusoid, or broadly clavate, apex obtuse, mucronated or pointed, hyaline, inamyloid; common. Lamellar margin sometimes fertile. **Cheilocystidia** (Fig. 28d) 24-36 X 4-7 µm, narrowly clavate to clavate, sometimes subventricose; apex frequently with appendages. **Stipe epicutis** parallel; hyphae 3-12 µm diam, inner hyphae broader, yellowish in mass, hyaline singly, with clamp connections; wall up to 1.6 µm thick. **Caulocystidia** entwined, forming a dense mat, up to 300 x 6 µm diam, cylindrical, slightly moniliform, frequently clamped.

Basidiospores (Fig. 28e) (6.0-)6.4-8.0(-8.8) X (2.8-)3.2-3.6(-4.0) µm (n = 60/3 x = 7.2 X 3.3 µm, Q = 1.70-2.75, Qx = 2.20), pip shaped to sublacrymoid in side view, ellipsoid to subcilindrical in face view, hyaline, inamyloid; wall smooth, thin; hilar appendix small.

Commentary: this is a common leaf litter agaric, whose basidiomata can be recognized by their brown grayish to almost white colors of the pileus with a stipe that becomes black with age. Microscopically, similar species to *G. omphalodes* are *G. lodgeae,* which can be distinguished by a much larger size and sulcate pileus, and from *R.*



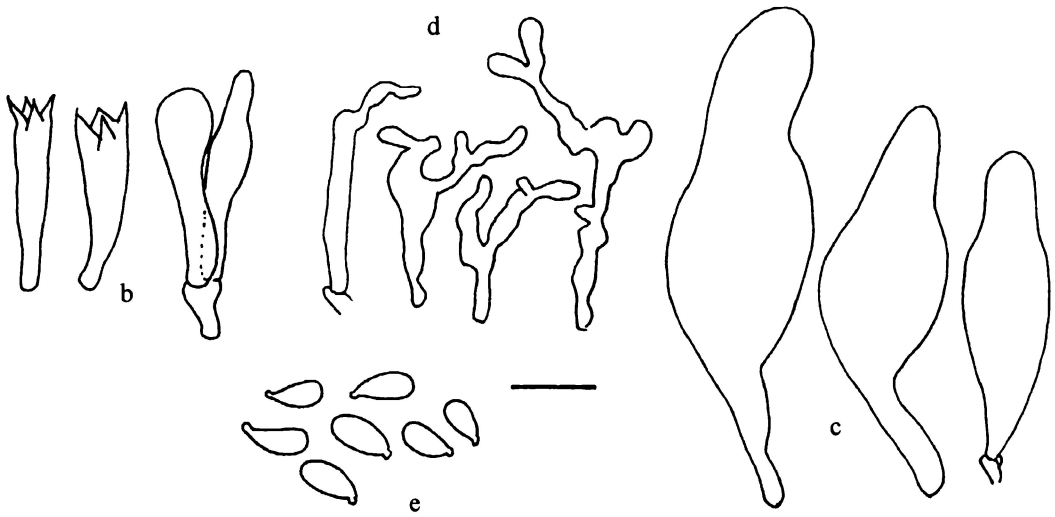


Fig. 28. *Gymnopus omphalodes*. a. basidiomata (TFB 10427); b. basidia and basidioles; c. pleurocystidia; d. cheilocystidia; e. basidiospores. Scale bar equals 10 μm for line drawings.

tablensis, which has smaller spores (4.8-6.0 X 2.8-4.0 μm) and crowded lamellae. Because of the prominent and common sub-erect to erect terminal cells of the pileus epicutis this taxon was considered to belong in *Marasmiellus* (Singer, 1973). In my work this kind of observed structure of the pileus epicutis, including terminal cells, is similar to that in *G. lodgeae*, *G. neotropica* and in a lesser degree in *G. impudicus*. Examination of the type specimen of *G. omphalodes* is pending.

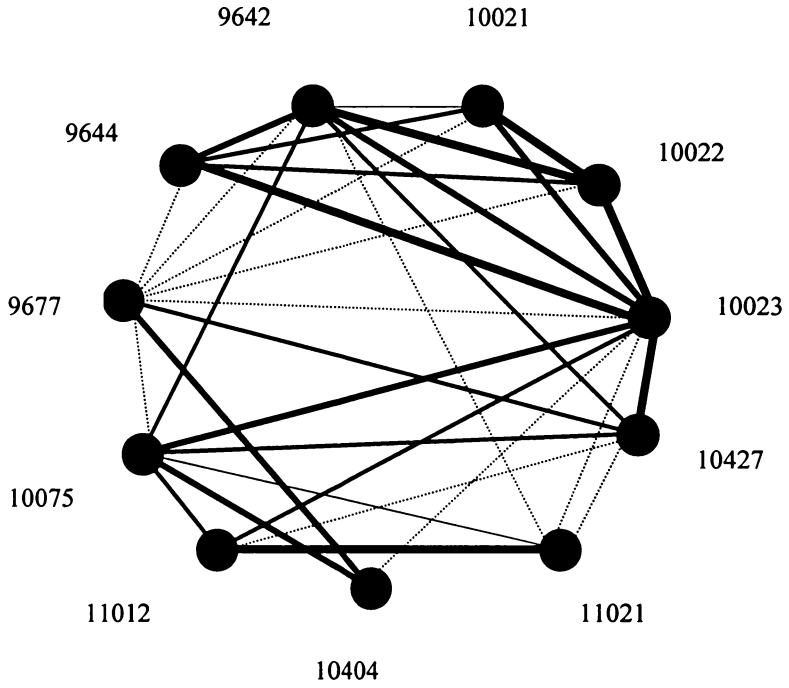
Gymnopus omphalodes appears phylogenetically related to *G. subnudus* and *G. peronatus*. The three species form a well-defined clade related to the *G. lodgeae* and *G. pseudolodgeae* clade (Figs. 2-4).

Mating studies. A bipolar mating system was reported for *G. omphalodes* by Mata & Petersen (1999) and is shown in Fig 29. In that paper it was also suggested that a cryptic species could be present in the Talamanca Mountains as judged by the mating behavior of one collection from Estrella (TFB 9677) with those from the Central Volcanic Mountains. However, later pairings with new material from other regions in Talamanca confirmed that *G. omphalodes* is a coherent biological species across these two mountainous zones of Costa Rica (Fig. 30).

Material examined: COSTA RICA: Alajuela Prov., Grecia Co., Reserva Forestal de Grecia, Bosque del Niño, 10° 8'38" N, 84° 14'62" W, 1750 m, 27.VI.1998, col. JL Mata, *TFB 9642* (TENN 56603), *TFB 9643* (TENN 56604), *TFB 9644* (TENN 56604), *TFB 9653* (TENN 56614); same location 8.VIII.1998, col. JL Mata, *TFB 10021* (TENN 56733), *TFB 10022* (TENN 56734), *TFB 10023* (TENN 56735). Cartago Prov., El Guarco Co., vic. Estrella, 9° 47'6" N, 83° 57'57" W, 1700 m, 13.VI.1995, col. RE Halling & RH Petersen, *TFB 7687* (TENN 53790); same location, 16.VI.1999, col. JL Mata, *TFB 10075* (TENN 58015). Puntarenas Prov., Coto Brus Co., Sabalito, vic La Amistad Lodge, Pizote Trail, 8° 54'22" N, 82° 47'40" W, ± 1330 m, 23.VI.1999, col. JL Mata & I

		A_1											A_2
		5	2	3	4	13	1	8	9	10	11	12	7
	5		-	+	+	+	+	+	+	+	+	+	+
A_1	2*	-		+	+	+	+	+	+	+	+	+	+
	3	+	+		-	-	-	-	-	-	-	-	-
	4	+	+	-		-	-	-	-	-	-	-	-
	13	+	+	-	-		-	-	-	-	-	-	-
	1*	+	+	-	-	-		-	-	-	-	-	-
	8	+	+	-	-	-	-		-	-	-	-	-
	9	+	+	-	-	-	-	-		-	-	-	-
	10	+	+	-	-	-	-	-	-		-	-	-
	11	+	+	-	-	-	-	-	-	-		-	-
	12	+	+	-	-	-	-	-	-	-	-		-
A_2	7	+	+	-	-	-	-	-	-	-	-	-	

Fig. 29. Self-cross pairings of SBIs from *Gymnopus omphalodes* TFB 10023. + = presence of clamp connections; - = absence of clamp connections; * = tester strains. Isolate numbers and assigned mating types top and side.



Location	Collection Number
Bosque del Niño, Central Volcanic Range	TFB 9642, TFB 9644, TFB 10021, TFB 10022, TFB 10023
Estrella	TFB 9677
Jardín de Dota	TFB 10075, TFB 11012
San Gerardo de Dota	TFB 10404, TFB 11021
La Amistad	TFB 10427

Fig. 30. Recognition grid for intercollection pairings from *Gymnopus omphalodes.* All matings are n = 4 SBIs. Compatibility based on clamp connection formation; ━━ 100% compatibility, ━━ 75% compatibility, —— 50% compatibility, —— 25% compatibility, no compatibility

González, *TFB 10427* (TENN 58121). San José Prov., Dota Co., Jardín de Dota, 3.5 km W off Interamerican Hwy at El Empalme, 9° 42'52" N, 83° 53'28" W, 2220 m, col. RH Petersen & JL Mata, *TFB 9677* (TENN 56638); same location, 25.VI.2000, col. JL Mata, RH Petersen, RE Halling & L Norvell, *TFB 11012* (TENN 58620); Pérez Zeledón Co., Hotel de Montaña Savegre, 9° 32'42" N, 83° 48'39" W, 2300 m, 19.VI.1999, col. JL Mata, *TFB 10404* (TENN 58099); same location, 26.VI.2000, col. JL Mata, RH Petersen, RE Halling & L Norvell, *TFB 11021* (TENN 58629) **S.**

(16) *Gymnopus pseudolodgeae* Mata, *sp. nov.* Figs. 31,32

Pileo convexo-umbonato ad plano-depreso; superficies velutinus, sulcatus, porphyreus. Lamellae distantes, roseo-fuscae. Stipite albidus ad nigricantis vetustatis. Habito lignicola. Pleurocystidia et cheilocystidia similares ad G. lodgeae cum basidiosporae 4.8-6.4 X 2.4-3.6 µm.

HOLOTYPE [*hic designatus*]: COSTA RICA. Puntarenas Prov., Coto Brus Co., vic. San Vito, Estación Biológica Las Alturas, OTS, on the trail to Cerro Chia, 1520 m, 21.VI.2000, col. RH Petersen & JL Mata, *TFB 10493* (TENN).

Etymology: *pseudo* (Greek) = false, resembling *G. lodgeae*.

Pileus 10-30(-40) mm diam, at first convex-umbonate, with age gradually expanding to applanate-depressed; surface finely velvety, strongly hygrophanous, deeply sulcate almost entirely, ± translucent, at first reddish brown (8E6-7E7), with age tan to light brown; margin at first incurved, with age curved to expanded; context very thin. **Lamellae** adnate, ± broad, distant, pinkish brown (7B4-2); margin even; lamellulae in two tiers of different lengths. **Stipe** 30-65 X 2-5 mm, ± equal, twisted; surface fibrillose, at apex at first off-white to cream or slightly reddish brown, with age turning black; interior hollow; consistency fibrous. Odor not distinctive; taste mealy to raphanoid.

Habitat on wood chips of fallen, decomposed log; gregarious (Fig. 31a).

Pileus epicutis a simple cutis; hyphae 2-8 μm diam, radially oriented, not branched, diverticula or knobbed, frequently septate, with clamp connections, pigment-incrusted light olivaceous brown in mass, hyaline singly, inamyloid; wall thin; terminal cells sub-erect to erect, up to 12 μm long. **Pileus trama** interwoven to ± radially oriented; hyphae 2-16 μm diam, hyaline, inamyloid, with clamp connections; wall thin to 1.6 μm thick. **Lamellar trama** subregular; hyphae 2-14 μm diam, hyaline, inamyloid, with clamp connections; wall thin. **Basidia** (Fig. 31b) 24-33 X 4-6 μm, narrowly clavate; sterigmata four. **Pleurocystidia** (Fig. 31c) 40-80 X 8-12 μm, mostly fusoid ventricose, hyaline, with clamp connections; wall thin. **Cheilocystidia** (Fig. 31d) (16-)32-48 X 6-12 μm, cylindrical, clavate to fusoid, hyaline, inamyloid, with clamp connections; apex obtuse diverticulate. **Stipe epicutis** parallel; hyphae 2-20 μm diam, hyaline, inamyloid, with clamp connections; wall thin. Gloeopleurous hyphae rare, up to 7 μm diam. **Caulocystidia** 24-32 X 5-8 μm, clavate to ventricose, or as small knobs, with clamp connections.

Basidiospores (Fig. 31e) 4.8-6.4 X 2.4-3.6 μm (n = 28, x = 5.6 X 3.1 μm, Q = 1.56-2.29, Qx = 1.82) ellipsoid in side view, obovoid in face view, hyaline, inamyloid; wall smooth, thin.

Commentary: Basidiomata of this taxon are very similar to those of *G. lodgeae*, but spores and cystidia of *G. pseudolodgeae* are smaller. *Gymnopus subpruinosus* seems to match macroscopically but no previous author has reported presence of pleurocystidia (Dennis, 1951; Desjardin et al, 1999; Pegler, 1983b). Basidiomata of *G. omphalodes* have much paler pilei and bases of stipes become black with age and pleurocystidia are much broader and basidiospores longer.

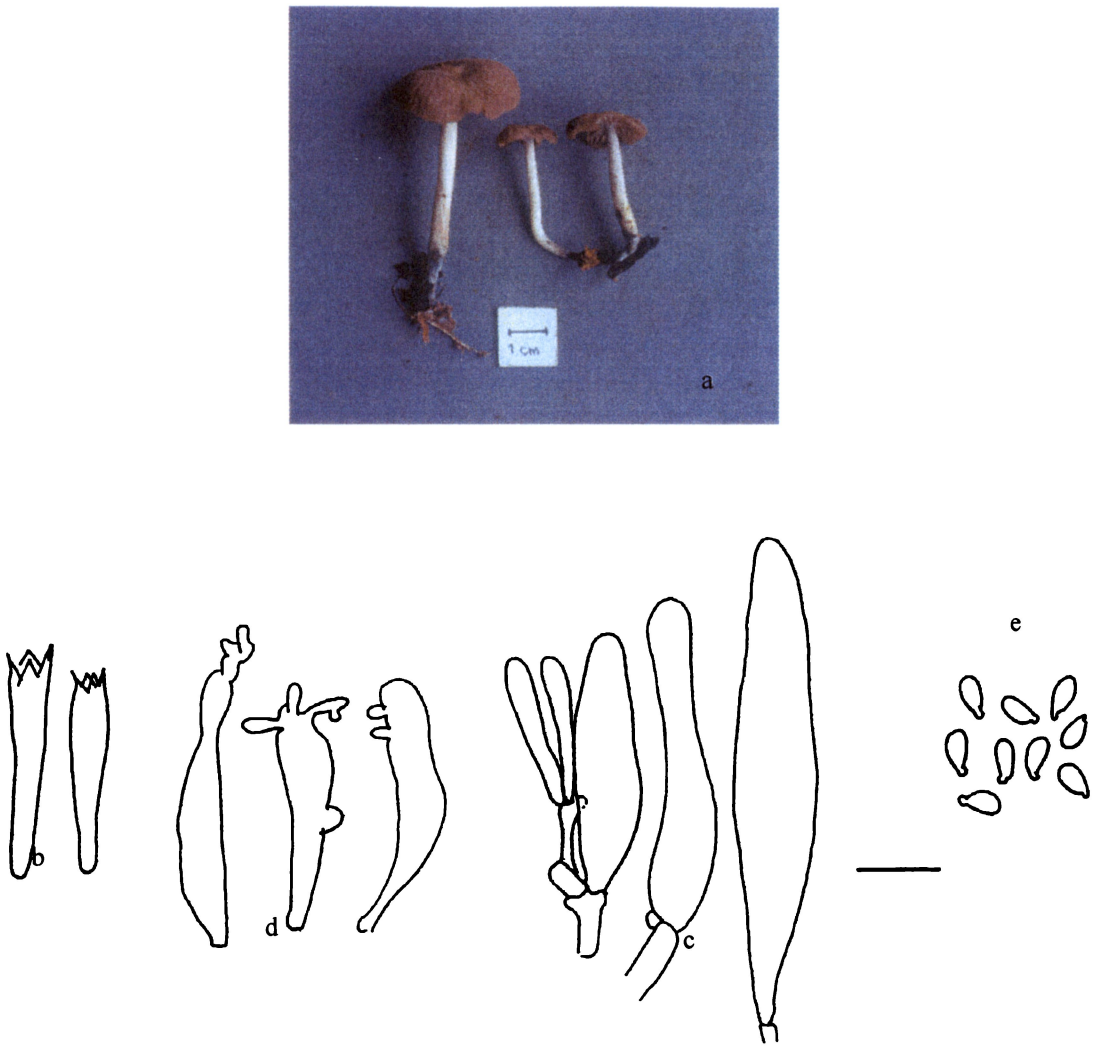


Fig. 31. *Gymnopus pseudolodgeae*. a. basidiomata (TFB 10493); b. basidia; c. pleurocystidia; d. cheilocystidia;. e. basidiospores. Scale bar equals 10 µm for line drawings.

Gymnopus pseudolodgeae appears phylogenetically closely related to *G. lodgeae* and to a sister clade formed by *G. subnudus*, *G. peronatus*, and *G. omphalodes* (Figs. 2-4).

Mating studies. The self-cross of 12 SBIs from the sole collection (TFB 10493) of *G. pseudolodgeae* resulted in a bipolar mating system (Fig. 32). This is the second report of such mating system, together with *G. omphalodes*, for *Gymnopus* in Costa Rica. The two mating types A_1 and A_2 were assigned arbitrarily. An intercollection pairing of SBIs from TFB 10493 with those from TFB 11013 representing *G. lodgeae*, n = 8, resulted in 100% incompatibility (data not shown).

Material examined: COSTA RICA. Puntarenas Prov., Coto Brus Co., vic. San Vito, Estación Biológica Las Alturas, OTS, on the trail to Cerro Chia, 1520 m, 21.VI.2000, col. RH Petersen & JL Mata, *TFB 10493* (TENN 58601) **S.**

Section *Vestipedes*

Type species *Gymnopus confluens.*

Pileus epicutis composed of cylindrical, radially oriented hyphae; pleurocystidia absent.

(17) *Gymnopus alnicola* Halling, sp. nov. Fig. 33

Pileo plano-convexo, fusco, albido in centro; carne concoloram pileo. Odore et sapore mitis. Lamellae albida ad palidae lialacea. Stipite equalis, pubescente, roseo-bruneo ad fusco. In Alnus vegetatio. Basidiosporae 8.8-13.6 X 3.2-4.8 μm.

HOLOTYPE [*hic designatus*]: ECUADOR. Napo Prov., Papallacta to Baeza Road, 3.8 Km E. of Papallacta, 30.IV.1987, col. RE Halling, *REH 5224 A*, NY]

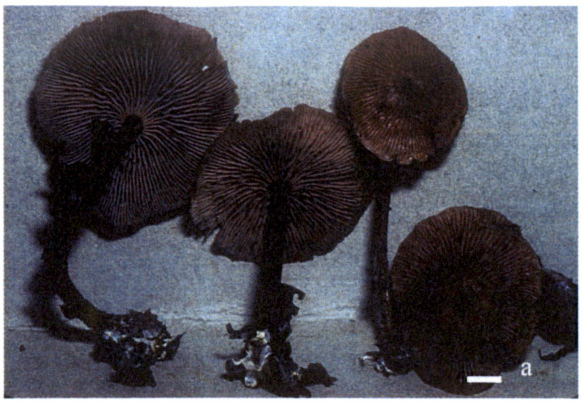
										A_1		A_2
		5	6	8	9	10	13	2	4	3	11	7	1
	5		-	-	-	-	-	-	-	-	+	+	+
	6	-		-	-	-	-	-	-	-	+	+	+
	8	-	-		-	-	-	-	-	-	+	+	+
	9	-	-	-		-	-	-	-	-	+	+	+
	10	-	-	-	-		-	-	-	-	+	+	+
	13	-	-	-	-	-		-	-	-	-	+	+
	2	-	-	-	-	-	-		-	-	-	+	+
	4	-	-	-	-	-	-	-		-	+	+	-
A_1	3*	-	-	-	-	-	-	-	-		+	+	-
	11	+	+	+	+	+	-	-	+	+		-	-
	7	+	+	+	+	+	+	+	+	+	-		-
A_2	1*	+	+	+	+	+	+	+	-	-	-	-	

Fig. 32. Self-cross pairings of SBIs from *Gymnopus pseudolodgeae* TFB 10493. + = presence of clamp connections; - = absence of clamp connections; * = tester strains. Isolate numbers and assigned mating types top

Etymology: from *Alnus* spp.; *cola* (Latin) = dweller

Pileus (10-)35-60 mm diam, obtusely convex, campanulate, convex to plano-convex, depressed to sharply depressed on disc; surface moist, hygrophanous, finely striate to sulcate, at first dark brown (7/8F8, 9F5), fading pale tan to white in depression, with age gray orange (6B5) to orange brown (6C-D4); margin at first smooth and even, with age striate-translucent, ± lacerate; context thin, watery brownish. **Lamellae** adnate to adnexed, crowded or close to subdistant, less than 1 mm broad, at first white to yellowish white, with age brown orange (6/7C3), sometimes with a pale lilac tint; margin even, minutely pruinose to fimbriate; lamellulae in three tiers of different lengths. **Stipe** 50-100 X (1-)3-8 mm, equal to slightly broadened towards base, flattened and compressed to cleft; surface dry, with white pubescence, more so at apex, at apex concolorous with lamellae, pale pinkish brown, brown (7F5) at first, at base and eventually overall with age gray brown (7D3) or concolorous to pileus; interior hollow. Rhizomorphs and binding mycelium white. Odor and taste none to mild. Chemical reactions Syring (-), p-Cresol: orange brown at stipe. **Habitat** on leaf litter, under *Alnus acuminata* vegetation; gregarious (Fig. 33a).

Pileus epicutis (Fig. 33b) a cutis; hyphae 2-8 µm diam, cylindrical, some with diverticulate outgrowths, radially oriented, heavily pigment-encrusted, olive-brown in mass, hyaline singly, orange-brown in IKI, with clamp connections; wall thin. Terminal cells 16-64 X 3-6 µm, mostly cylindrical, some with diverticula, prostrate to erect, singly or in fascicles, common. **Pileus trama** interwoven to ± radially oriented; hyphae 4-12 µm diam, hyaline, inamyloid, with clamp connections; wall thin. **Lamellar trama** irregular to interwoven; hyphae 3-10 µm diam, hyaline, inamyloid, with clamp connections; wall thin. **Basidia** (Fig. 33c) 19-31 X 5-9 µm, clavate; sterigmata four; basidioles 19-34 X 5-8 µm, cylindrical to clavate, occasionally mucronate or subfusoid. **Pleurocystidia** absent. **Cheilocystidia** (Fig. 33d) (19-)28-45 X 512 µm, cylindrical, narrowly clavate, sphaeropedunculate or submucronate, some flexed; prostrate, collapsing and not easy to



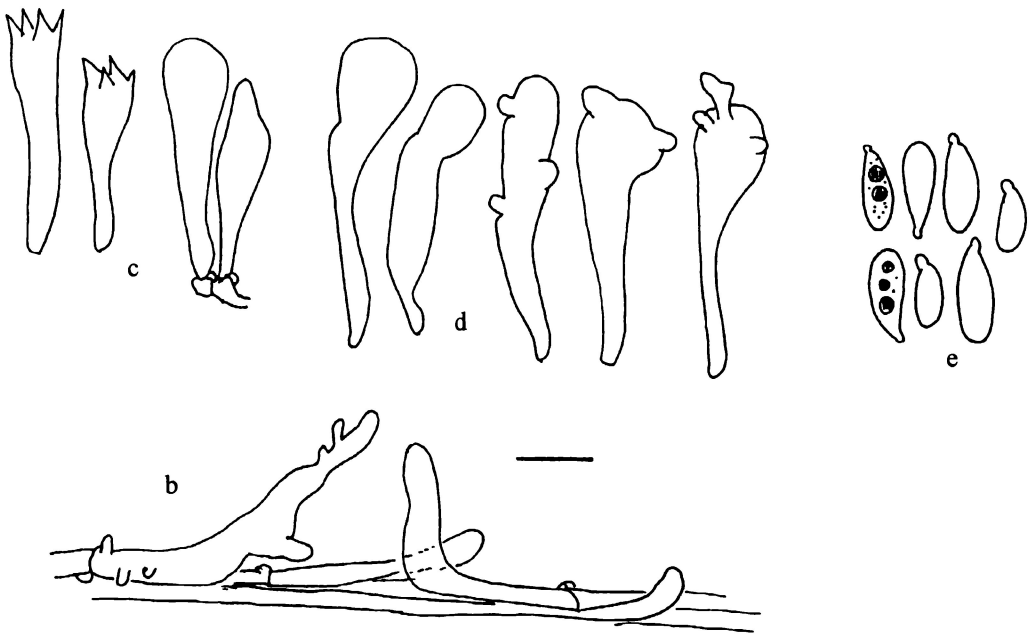


Fig. 33. *Gymnopus alnicola*. a. basidiomata (REH 8266); b. elements of pileus epicutis; c. basidia and basidioles; d. cheilocystidia; e. basidiospores. Scale bar equals 10 μm for line drawings, 1 cm for picture.

see; apex sometimes knobbed or with diverticulate outgrowths. **Stipe epicutis** parallel; hyphae 4-14 µm diam, cylindrical, outermost hyphae pigmented like pileus epicutis, with clamp connections; wall up to 0.8 µm thick. **Caulocystidia** abundant, cylindrical, 2-4 µm diam, some with diverticulate outgrowths, entangled, some septate, with clamp connections.

Basidiospores (Fig. 33e) (6.8-)7.6-9.6(-13.6) X 3.2-4.0 µm (n = 100/4, x = 8.6 X 3.7 µm, Q = (1.88-)2.00-2.78(-3.11), Qx = 2.33), narrowly ellipsoid or lacrymoid in side view and in face view, hyaline, inamyloid; wall smooth, thin; contents often multiguttulate.

Commentary: This is a new species, fruiting under *Alnus acuminata*, found by RE Halling. The collybioid characters of the basidiomata in combination with a cutis structure of the pileus epicutis and hyaline, inamyloid spores place this taxon in *Gymnopus*. Basidiomata of *G. confluens* and *G. cylindricus* are similar to those of *G. alnicola* but microscopically can be separated by spore size and shape of cheilocystidia, respectively. While the four specimens from Ecuador are very similar in all morphological characters, a third collection from Costa Rica is a little different. When dried specimens were compared to each other, the Costa Rican material presented much larger basidiomata, pilei more sulcate, lamellae appearing more distant and broader, and a stipe vesture denser and beige colored, than the Ecuadorian material. Most notable is that spore length is much bigger in the Costa Rican basidiomata while cheilocystidia are similar. Because this species was first noted from Ecuador, the type collection designated here is from that locality.

Gymnopus alnicola is phylogenetically related to others representing Costa Rican specimens of *Gymnopus* sect. *Vestipedes* including *G. neotropicus*, *G. biformis* and *G. confluens* (Figs. 2-4).

Material examined: COSTA RICA. Cartago Prov., El Guarco, Tapantí, Parque Nacional Tapantí, Macizo de La Muerte, Area de Conservación La Amistad Pacífico, 9° 41'06" N, 83° 52'30" W, 2600 m, 6.VII.2001, col. RE Halling, *REH 8266* (NY) **S.** ECUADOR. Napo Prov., Papallacta to Baeza Road, 3.8 Km E. of Papallacta, 30.IV.1987, col. RE Halling, *REH 5224 A* (NY, HT); same location, ± 8-9 Km E. of Papallacta, at Puente Guango, in *Alnus* forest, 30.IV.1987, col. RE Halling, *REH 5225, REH 5226, REH 5227* (NY).

(18) *Gymnopus biformis* (Pk.) Halling. 1997. Mycotaxon 63: 363. Figs. 34, 35

≡ *Marasmius biformis* Peck. 1903. Bull. N.Y. State Mus. 67: 25.

≡ *Collybia biformis* (Pk.) Singer. 1962. Sydowia 15: 55.

HOLOTYPE: UNITED STATES. New York, Rennselaer Co., Sandlake, VIII.1902, col. CH Peck, (NY)[!].

Pileus (5-)12-40 mm diam, broadly convex to depressed; surface fibrillose, opaque, entirely crenate to sulcate, brown, 6E8-6D7, at depression beige or tan; margin crenate, translucent; context very thin. **Lamellae** adnate, adnexed to seceding, in some forming a pseudocollarium, ± broad, close to subdistant, white to cream (3A3-4A3); margin even; lamellulae in two tiers of different lengths, with intervenose projections. **Stipe** 25-60 X 1-2 mm, equal, terete; surface covered overall densely pruinose, at times strigose, background color brown, rusty brown to dark brown; vesture white to creamy; interior becoming hollow; consistency ± cartilaginous. Mycelium rhizomorphic, off-white. Odor fungous; taste not recorded. **Habitat** on ± fresh leaves, on top of leaf litter; gregarious (Fig. 34a).

Pileus epicutis (Fig. 34b) a cutis; hyphae 2-8 μm diam, cylindrical, radially oriented, not branching, with diverticula, heavily pigment-encrusted, brown, inamyloid, with clamp connections; wall thin; terminal cells common, cylindrical to clavate, occasionally with diverticula, semierect to erect, dark brown; wall thickened, in some

becoming sclerified and black. **Pileus trama** interwoven to ± radially oriented; hyphae 2-10(-16) μm diam, hyaline, inamyloid, with clamp connections; wall thin. Gloeopleurous hyphae rare, up to 6 μm diam. **Lamellar trama** subregular to interwoven; hyphae 2-8 μm diam, hyaline, inamyloid, with clamp connections; wall thin. **Basidia** (Fig. 34c) 20-28 X 6-7 μm, clavate; sterigmata four, occasionally two; basidioles 23-26 X 5-7 μm, clavate to subcapitate. **Pleurocystidia** absent. Lamellae edge sterile. **Cheilocystidia** (Fig. 34d) 16-36 (-50) X 6-12 μm, clavate to sphaeropedunculate, occasionally lobed, knobbed or furcate, or septate, with clamp connections. **Stipe epicutis** parallel; hyphae 3-12 μm diam, innermost hyphae up to 14 μm diam, frequently septate, yellowish to light brown in mass, hyaline singly, inamyloid, with clamp connections; wall up to 1 μm thick. **Caulocystidia** up to 200 X 6 μm, cylindrical, strangulate, septate, with clamp connections, often in fascicles or densely packed; arising from a dense mat of hyphae, 2-6 μm diam.

Basidiospores (Fig. 34e) 6.4-9.2(-11.2) X 2.4-4.8(-6.0) μm (n = 105/5, x = 8.1 X 3.7 μm, Q = 1.80-2.88, Qx = 2.21) lacrymoid to subfusoid in side view, subcilindrical to narrowly ellipsoid in face view, hyaline, inamyloid; wall smooth, thin.

Commentary: Examination of the type specimen of *G. biformis* confirmed the identity of the basidiomata collected in Costa Rica. Basidiomata are very similar to and easily confused with those of *G. subnudus* (Halling, 1983). Besides the previously reported distinctions between the two species (Halling, 1983), it can be added that cheilocystidia in basidiomata of *G. biformis* tend to have a considerable variation in shape (i.e. typically contorted, frequently lobed, knobbed or diverticulate), and the range of basidiospore length is lower, by 1-2 μm, than that reported for the type specimen and natural populations of *G. subnudus*, i.e. 8.6-10.4(-12) μm (Desjardin, 1989; Halling, 1983).

Tropical to subtropical species similar to *G. biformis* are *G. menehune* described from Hawaii, *G. subcyathiformis* from México, *Collybia johnstonii* from the Caribbean,

and *G. collybioides* from Brazil. Lamellae of basidiomata of *G. menehune* are closer together and do not form a pseudocollarium as in *G. biformis*; cheilocystidia are abundant, clavate to lobed. Basidiomata of *G. collybioides* are much smaller and lamellae are much more crowded than those of *G. biformis*, but microscopic elements are similar. The type specimen of *G. subcyathiformis* is very similar in lamellar distance and stipe vesture to basidiomata of *G. biformis* collected for this project, as are spore dimensions, but presence of cheilocystidia in *G. subcyathiformis* could not be ascertained positively. All of the above-mentioned species have diverticulate projections on the hyphae of the pileus epicutis. *Collybia johnstonii* is a collybioid agaric similar in stature and habit to *G. biformis*, with pileus more grayish colored, stipe less pubescent, and cheilocystidia more nodulose (Dennis, 1951; Pegler, 1983b; see material examined below).

Costa Rican specimens of *G. biformis* were morphologically identical (when compared in dried condition) to those specimens from the United States which also are sexually intercompatible (data not shown). All of these specimens are characterized by having a densely pubescent to strigose stipe with white-cream vesture. Lamellae are close to distant, cream yellowish and pileus often uplifted when dried, rugulose-sulcate, sometimes with the center of the disc cream colored. *Gymnopus biformis* seems to be present in oak-forest habitats in southern Costa Rica and in Colombia, and has been recently reported in Japan (Miyamoto et al, 1998).

Costa Rican *G. biformis* is phylogenetically related to G. *alnicola*, *G. confluens*, and *G. neotropicus* (Figs. 2-4). Pairwise sequence comparison of Costa Rican and United States *G. biformis* resulted in 92% similarity.

Mating studies. Only collection TFB 11035 yielded some SBIs. The self-cross (n = 5) resulted in an apparent tetrapolar mating system (Fig. 35). With so few SBIs used, this report must be taken with caution. It must be said that micromorphology of this collection revealed that the hymenium contained almost exclusively two-sterigmate basidia which may have a direct influence in mating data as noted by Petersen (1995b).

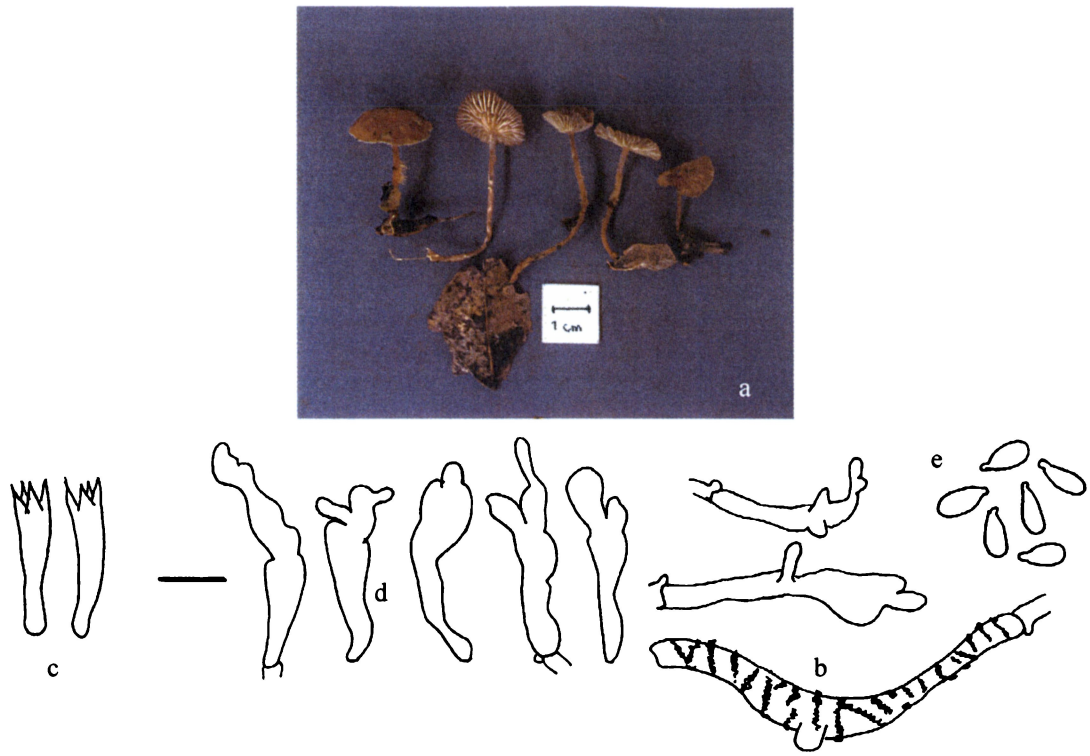


Fig. 34. *Gymnopus biformis* a. basidiomata (TFB 10093); b. elements of pileus epicutis; c. basidia; d. cheilocystidia; e. basidiospores. Scale bar equals 10 µm for line drawings.

		A_1B_1	A_1B_2			A_2B_1
		1	2	4	9	7
A_1B_1	1*		-	-	-	-
A_1B_2	2*	-		-	-	+
	4	-	-		-	+
	9	-	-	-		+
A_2B_1	7*	-	+	+	+	

Fig. 35. Self-cross pairings of SBIs from *Gymnopus biformis* TFB 11035; + = presence of clamp connections; - = absence of clamp connections; * = tester strains. Isolate numbers and assigned mating types top and side.

To my knowledge, no report on the mating system for *G. biformis* has been published. Intercollection pairings (n = 4) of TFB 11035 (Costa Rica) with TFB 7230 (*G. biformis*, United States), TFB 9657 (*G. biformis* var. *lobatus*), and TFB 10425 (*G. biformis* var. *parvulus*) resulted in 100 % intersterility (data not shown).

Material examined: COLOMBIA. Dept. Nariño, Mun. Pasto, 11 km E of Chachagüí, Bosque El Común, *Q. humboldtii* vegetation, 22.XI.1998, col. RE Halling, *REH 6133* (NY). COSTA RICA. Cartago Prov., EL Guarco Co., near Estrella, col. JL Mata, *TFB 11035* (TENN 58642) S. Puntarenas Prov., Coto Brus Co., Sabalito, La Amistad Lodge, Fila Palmital, Sendero Higuerón, 8° 54'52" N, 82° 46'50" W, 1300 m, 18.VI.1995, col. RE Halling, R Tulloss, C Shanks, RH Petersen, *REH 7431* (NY); same location, 4.VII.1998, col. JL Mata & RH Petersen, *TFB 9696* (TENN 56657). San José Prov., Dota Co., Jardín de Dota, 3.5 km W of Interamerican Hwy at El Empalme, 9° 42'52" N, 83° 58'28" W, 2220 m, 15.VI.1995, col. RE Halling, R Tulloss, C Shanks, RH Petersen, *REH 7422* (NY); same location, 18.VI.1999, col. JL Mata, & RH Petersen, *TFB 10093* (TENN 58088). UNITED STATES. Georgia, Rabun Co., Double Bridges, 21.VII.1994, col. DBG Nichol, *TFB 7664* (TENN 56628). New York, Rennselaer Co., Sandlake, VIII.1902, col. CH Peck, HT of *M. biformis*, (NYS). Louisiana, Baton Rouge Co., Baton Rouge, Burden Research Park, 25.V.1995, col. SC McCleneghan, *TFB 9516* (TENN 56273); Tammany Parish, Fountainbleu State Park, 24.V.1995, col. RH Petersen, *TFB 9111* (TENN 55740), *TFB 9127* (TENN 55753). South Carolina, Oconee Co., Walhalla Fish Hatchery Rd., 20.VII.1994, col. RH Petersen, *TFB 7230* (TENN 53558).

Gymnopus menehune Desjardin, Halling & Hemmes. [1999. Mycologia 91(1): 173.] UNITED STATES. Hawaii, Hawaii, Puna District, MacKenzie State Park on the SE coast, 3.VII.1993, col. DE Desjardin, D Hemmes & M Goldsmith, *DED 5866* (NY, IT), *DED 6515* (NY, PT).

Gymnopus subcyathiformis (Murr.) Desjardin, Halling & Hemmes. [1999. Mycologia 91(1): 175.] MEXICO. Colima, 3/4.I.1910, col. Edna & WA Murrill, *no. 615*, (NY, HT).

Gymnopus subnudus (Ellis ex Peck) Halling. [1997. Mycotaxon 63: 365.]

UNITED STATES. Louisiana. Baton Rouge Co., E. Baton Rouge, 25.V.1995, col. SC McCleneghan, *TFB 9120* (TENN); North Carolina, Macon Co., Blue Valley, Forest Service Rd 89, 14.VI.1989, col. RH Petersen, *TFB 1804* (TENN 48444); Coweeta Hydrological Laboratory, 22.VI.1989, col. RH Petersen, *TFB 1818* (TENN 48353); Whitesides Cove Rd, trail from "Summer Chapel", 8.VII.1999, col. RH Petersen, *TFB 10338* (TENN 57899) **S.**

Collybia johnstonii (Murr.) Dennis. [1951. Trans. Brit. Mycol. Soc. 34: 437.] COLOMBIA. Dept. Antioquia, Mun. Cocorna, Refugio Cañón del Río Claro, 5° 55' N, 74° 50' W, 200-500 m, 22.XI.1986, col. RE Halling, *REH 5047* (NY); Reserva Guayabo, 19.V.1987, col. RE Halling, *REH 5268* (NY).

(19) *Gymnopus biformis* var. *lobatus* Mata, *var. nov.* Figs. 36, 37

Basidiomata similaris ad Gymnopus biformis sed distinctas per cheilocystidae lobatis.

Etymology: *lobatus* (Latin) = partly divided into a number of rounded segments.

Pileus 10-25 mm diam, at first convex, soon broadly convex, with a shallow umbo or slightly umbilicate; surface glabrescent to radially fibrillose, dry, opaque, at first finely striate but soon almost entirely crenate-sulcate, or rugulose, translucent towards margin, at first reddish brown (7E7-6/7E6), orange brown (7/6E8), soon light brown (6D6), in some buff or beige near margin, with age pale brown or cream buff at center; margin curved to straight, even at first, soon uneven and crenate to sulcate, translucent, with age lobed or cracked; context very thin. **Lamellae** adnate to adnexed, with age seceding and forming a pseudocollarium, narrow, in some arched sinuate, close to subdistant, at first white to off-white, soon cream (3/4A2); margin even; lamellulae in two to three tiers of different lengths, in some anastomosing frequently. **Stipe** (5-)30-70 X 1-3 mm, at first equal and terete, with age sulcate and compressed, a little broader at apex; surface entirely pruinose, more so with age and apically, at first entirely reddish

brown (6/7F8, 6/7E5), purplish (8/9E8) or light brown (6E5/7), sometimes near apex creamy to grayish and blackening at base; interior hollow; consistency cartilaginous, tough. Basal mycelium a small disc, frequently fanning out opposite from side of base insertion, cream (4A2). Odor not distinctive; taste fungous. **Habitat** on ± freshly fallen leaves, on upper layer of leaf litter; scattered (Fig. 36a).

Pileus epicutis (Fig. 36b) a cutis; hyphae 2-8 μm diam, cylindrical, radially oriented, repent, not branched, rarely with small diverticula, heavily pigment-incrusted, dark olive brown in mass, hyaline singly, orange in IKI, with clamp connections; wall thin; terminal cells erect or semi-erect, occasionally in fascicles. **Pileus trama** loosely interwoven to interwoven, rarely ± radially oriented; hyphae 2-12(-14) μm diam, hyaline, inamyloid, with clamp connections; wall thin to 1 μm thick. **Lamellar trama** subregular to irregular, sometimes interwoven; hyphae 2-8 μm diam, hyaline, inamyloid, with clamp connections; wall thin; subhymenium pseudoparenchymatous, 2-3 cells broad. **Basidia** (Fig. 36c) 22-27 X 6-8 μm, clavate; sterigmata four; basidioles 20-24 X 4-6 μm, clavate to submucronate. **Pleurocystidia** absent. Lamellar margin sterile. **Cheilocystidia** (Fig. 36d) 28-45 X 5-10 μm, clavate or bi-, tri-lobate, sometimes furcate, rarely with diverticula, with clamp connections. **Stipe epicutis** parallel; hyphae 2-12 μm diam, pale brown in mass, hyaline singly, inamyloid, with clamp connections; wall up to 0.8 μm thick. **Caulocystidia** up to 120 X 6 μm diam, cylindrical, strangulate, sometimes septate and in fascicles; apex obtuse or knobbed.

Basidiospores (Fig. 36e) (5.6-)6.4-8.4(-9.6) X (2.8-)3.2-4.0 μm (n = 110/6, x = 7.3 X 3.4 μm, Q = 1.63-2.50, Qx = 2.10) pip shaped or lacrymoid in side view, narrowly ellipsoid in face view, hyaline, inamyloid; wall smooth, thin.

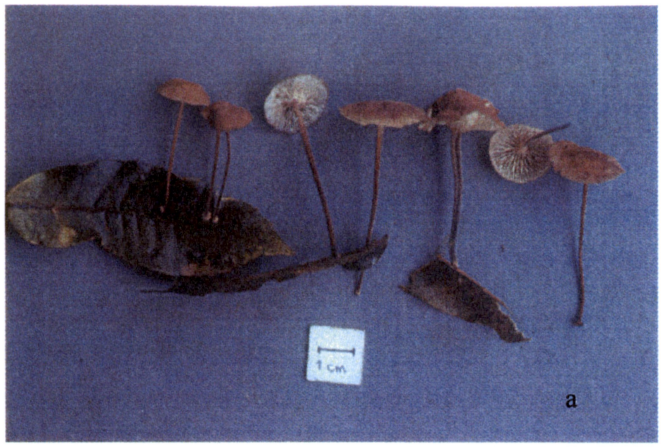
Commentary: Macro- and microscopic characteristics observed in basidiomata representing *G. biformis* var. *lobatus* are very similar to those representing *G. biformis* and its other variety. Basidiomata of *G. biformis* vars *lobatus* appear to lack the densely strigose-pubescent vesture of the stipe seen in those representing *G. biformis*. Also, pilei

seem to be more depressed-umbilicate, with more orange-brown to tan colors, and lamellae less distantly spaced, and stipes that turn black with age. Microscopically, spores seem to be similar, but cheilocystidia appear to be consistently more lobate and furcate than those observed in specimens of var. *biformis* and var. *parvulus*. Despite the various apparent morphological differences it is my intention to keep this name as a variety under *G. biformis* and wait for more collections to be described in order to have a better understanding of the morphological boundaries of this taxon and species complex.

Gymnopus biformis var. *lobatus* and *G. biformis* var. *parvulus* are phylogenetically related to *G. collybioides* and *G. cylindricus* (Figs. 2-4). Pairwise sequence comparisons of *G. biformis* var. *parvulus* with *G. biformis* var. *lobatus* resulted in 93% similarity.

Mating studies. Collections TFB 9657, TFB 9673 and TFB 10034 were paired against each other (n = 4). No clamp connections were observed in any of these pairings (Fig. 37). In pairings of TFB 9657 and TFB 11016 with TFB 7230 and TFB 9111 (n = 4) no clamp connections were observed (data not shown).

Material examined: COSTA RICA. Cartago Prov., vic. Estrella, Palo Verde, 9° 46'59" N, 83° 56'71"W, 1700 m, 28.VI.1998, col. JL Mata & RH Petersen, *TFB 9656* (TENN 56617), *TFB 9657* (TENN 56618) S; same location, 10.VIII.1998, col. JL Mata, *TFB 10034* (TENN 56746); same location, 24.VI.2000, col. JL Mata & RH Petersen, *TFB 11006* (TENN 58614), *TFB 11007* (TENN 58615), *TFB 11009* (TENN 58617). San José Prov., Dota Co., Jardín de Dota, 3.5 km W of Interamerican Hwy at El Empalme, 9° 42'52" N, 83° 58'28" W, 2220 m, 30.VI.1998, col. RE Halling, RH Petersen & JL Mata, *TFB 9673* (TENN 56634); same location, 25.VI.2000, col. JL Mata & RH Petersen, *TFB 11016* (TENN 58624).



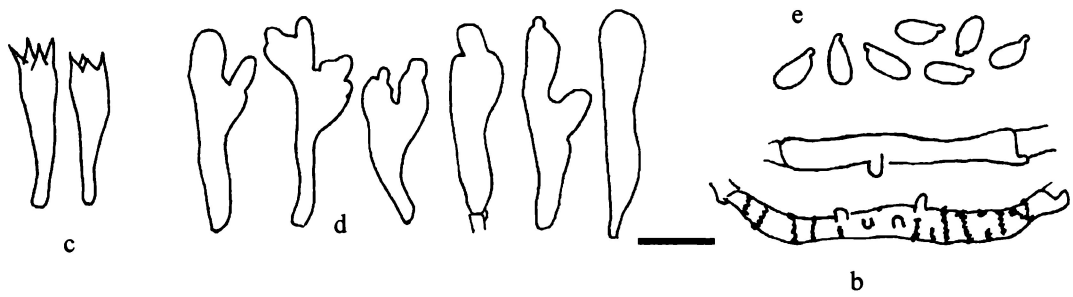


Fig. 36. *Gymnopus biformis var. lobatus* a. basidiomata (TFB 11006); b. elements of pileus epicutis ; c. basidia; d. cheilocystidia; e. basidiospores. Scale bar equals 10 μm for line drawings.

		9657		
Palo Verde	9657		9673	
Palo Verde	9673	0/4		10034
Jardín de Dota	10034	0/4	0/4	

Fig. 37. Intercollection pairings from *Gymnopus biformis* var. *lobatus*. Numbers indicate ratio of pairings where clamp connections were observed.

(20) *Gymnopus biformis* var. *parvulus* Mata, *var. nov.* Figs. 38, 39

Basidiomata similaris ad Gymnopus biformis sed distinctas per staturas parvulus.

Etymology: *parvulus* (Latin) = very small.

Pileus 10-22(-35) mm diam, at first convex to applanate, then somewhat centrally depressed and more notorious with age; surface radially fibrillose, opaque, entirely sulcate-crenate, translucent, when young uniform orange brownish (6D7-6C5), brown to light (5/6D8-5C6/8), at center light orange brown (6C5/6) with age, near margin beige, pinkish buff (6B4/5), tawny buff (5B4/5) to buff (5C4-5B3); margin curved to plane, crenate-sulcate; context very thin. **Lamellae** adnate to free, forming a pseudocollarium, narrow, close, white to off-white (2A2); margin even, fimbriate to eroded; lamellulae in three tiers of different lengths, some anastomosing or with intervenose projections. **Stipe** 30-50(-70) X 1-2(-4) mm, equal, terete to ± compressed, occasionally flaring at apex with age; surface covered entirely with a fine white vesture, towards base brown to reddish brown (7D/E8), dark brown (7F8) to light brown (7E6/7), fading in color apically; interior hollow; consistency tough, cartilaginous. Basal mycelium a disc, white to creamy. Odor not distinctive or pleasant; taste mild or mealy. **Habitat** directly on leaves in leaf litter; scattered (Fig. 38a).

Pileus epicutis (Fig. 38b) a cutis; hyphae 2-8 µm diam, cylindrical, not branched, occasionally diverticulate, radially oriented, ± interwoven, pale brown to pale olive brown in mass, hyaline singly, pale orange in IKI, with clamp connections; wall thin; terminal cells, cylindrical to clavate, occasionally strangulate, erect, frequently with diverticula. **Pileus trama** interwoven to loosely interwoven, rarely ± radially oriented; hyphae 2-10(-16) µm diam, hyaline, inamyloid, with clamp connections; wall thin to 0.8 µm thick. **Lamellar trama** subregular to irregular, rarely interwoven; hyphae 2-8(-12) µm diam, hyaline, inamyloid, with clamp connections; wall thin. **Basidia** (Fig. 38c) 20-38 X 4-10 µm, clavate; sterigmata four; basidioles 17-28 X 4-7 µm, clavate to

cylindrical. **Pleurocystidia** absent. Lamellae edge sterile or fertile. **Cheilocystidia** (Fig. 38d) 13-36 X 4-12 µm, typically broadly clavate, sometimes subfusoid, cylindrical or flexuous; apex frequently lobed, knobbed or with irregular appendages, rarely furcate, with clamp connections. **Stipe epicutis** parallel; hyphae 2-10 µm diam, pale brownish in mass, hyaline singly, inamyloid, with clamp connections; wall up to 1.6 µm thick. **Caulocystidia** up to 160(-260) X 10(-15) µm, cylindrical, strangulate, occasionally septate, with clamp connections; single or forming fascicles; apex obtuse.

Basidiospores (Fig. 38e) 5.6-8.0(-8.8) X 2.8-3.6(-4.8) µm (n = 180/8, x = 7.2 X 3.5 µm, Q = 1.60-2.75, Qx = 2.08) lacrymoid to narrowly ellipsoid in side view, narrowly ellipsoid to subcilindrical in face view, hyaline, inamyloid; wall smooth, thin.

Commentary: Basidiomata of *G. biformis* var. *parvulus* appear to be much smaller in stature than those of var. *biformis* but similar to those of var. *lobatus*. Additionally, like var. *lobatus*, pilei have more orange-brown to tan colors than those in var. *biformis*. Basidiomata of var. *parvulus* are different from those of var. *lobatus* in that the latter have been described as having blackening stipes and pileus colors that do not fade significantly outward to the margin. Lamellar insertion seems to be similar among all varieties in that in young stages they are attached and gradually, as the pileus expands, secede into a pseudocollarium which can be observed well in dried condition. Stipe vesture is similar to that of var. *lobatus* but not as stout or pubescent as that of var. *biformis*. Cheilocystidia in var. *parvulus* are similar to those observed in Costa Rican specimens of var. *biformis*, but contrast with the lobate-furcate shape of those in var. *lobatus*. Spore dimensions are similar in all varieties but the mean and ratio for var. *biformis* (x = 8.1 X 3.7 µm, Qx = 2.21) appear to be higher than those for var. *lobatus* (x = 7.3 X 3.4 µm, Qx = 2.10) and var. *parvulus* (x = 7.2 X 3.5 µm, Qx = 2.08). As with *G. biformis* var. *lobatus* more collections are needed in order to define better morphological boundaries in this species complex.

Mating studies. It was not possible to recover enough SBIs to perform a self-cross. Three SBIs from collection TFB 10422 were paired with six of TFB 10425 (Fig. 39) resulting in 33% compatibility. When SBIs (n = 4) of TFB 10422 and TFB 10425 were paired with TFB 9657 and TFB 11016 (var. *lobatus*) no clamp connections were observed (data not shown). The same result was observed in pairings with SBIs from TFB 7230 and TFB 9111 representing *G. biformis* from the United States (data not shown).

Material examined: COSTA RICA. Cartago, vic. Estrella, Palo Verde, 9° 46'59" N, 83° 56'71" W, 1700 m, 28.VI.1998, col. JL Mata & RH Petersen, *TFB 9663* (TENN 56624). San José Prov., Dota Co., Finca El Jaular, 9° 39'39" N, 83° 52'01" W, 2300 m, 17.VI.1999, col. JL Mata, *TFB 10082* (TENN 58022); same location, 29.VI.2000, col. JL Mata & RH Petersen, *TFB 11028* (TENN 58636); Pérez Zeledón Co., vic. Villa Mills, Km 95 on Inter American Highway past CATIE Experimental Station, Estación Biológica Cuericí, 9° 33'17" N, 83° 40'04" W, 2560 m, 24.VIII.1995, col. E Franco & M Mata, *EFM 1446* (NY); same location, 21.VI.1999, col. JL Mata, *TFB 10419* (TENN 58113), *TFB 10421* (TENN 58115), *TFB 10422* (TENN 58116) **S**, *TFB 10424* (TENN 58118), *TFB 10425* (TENN 58119).

(21) *Gymnopus collybioides* (Speg.) Desjardin, Halling & Hemmes. 1999. Mycologia 91: 175. Figs. 40,41

≡ *Clitocybe collybioides* Spegazzini. 1889. Bol. Acad. Nac. Ci. 11(4): 387.

≡ *Collybia collybioides* (Speg.) Singer. 1950. Lilloa 23: 162.

HOLOTYPE: BRAZIL. Apiahy, IV.1888, col. J. Puiggari, *no 2893*, (LPS) [!].

Pileus 7-30(-40) mm diam, at first campanulate or convex with a sharply depressed disc, with age becoming plane with a shallowly depressed disc; surface moist or dry, radially fibrillose, appressed, disc dark brown (8F8), orange brown (7E7, 6/7C6) with age becoming pale cinnamon brown (6E6-6C6), at center cream or yellowish;

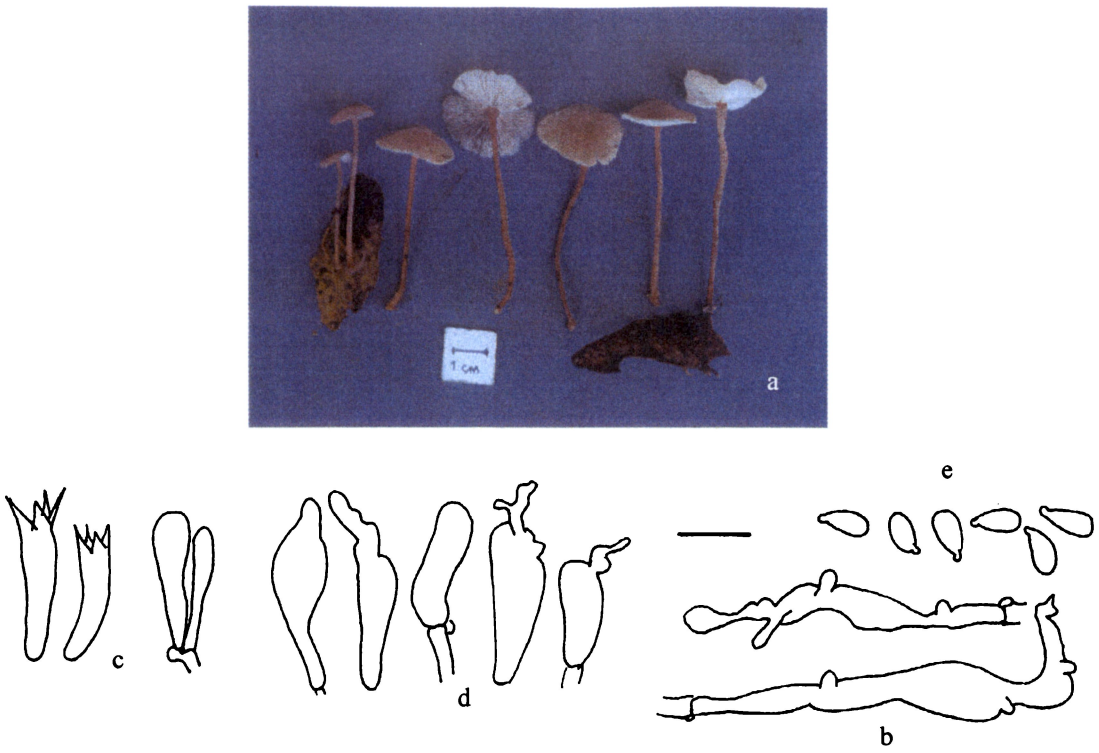


Fig. 38. *Gymnopus biformis* var. *parvulus* a. basidiomata (TFB 11028); b. elements of pileus epicutis ; c. basidia and basidioles; d. cheilocystidia; e. basidiospores. Scale bar equals 10 µm for line drawings.

FB 10422 \ FB 10425	1	8	2	3	5	7
1	-					
1		-				
2			+			
2				+		
3					-	
3						-

Fig. 39. Intercollection pairings from *Gymnopus biformis* var. *parvulus*. Three SBIs of TFB 10422 were crossed against six from TFB 10425. + = presence of clamp connections, - = absence of clamp connections.

margin finely striate to irregular; context thin, watery white to creamy. **Lamellae** adnate to adnexed, extremely crowded, occasionally anastomosed, white; margin even to barely subfimbriate in some; lamellulae in up to five tiers of different lengths. **Stipe** 40-120 X 1-3 mm, equal, strict, terete to compressed; surface overall subpruinose, more strigose-pubescent below, concolorous with lamellae above, darker below, grayish brown, orange brown to brown (6/7D7) or dark brown (6E8-7F8); interior becoming hollow; consistency ± cartilaginous but fragile. Rhizomorphs white. Odor none or farinaceous; taste none or mealy. **Habitat** on humus and leaf litter; solitary to gregarious (Fig. 40a).

Pileus epicutis a cutis; hyphae 2-6(10) µm diam, cylindrical, not branched, radially oriented, occasionally diverticulate, heavily pigment-encrusted, light brown to olive brown in mass, hyaline singly, orangish in IKI, with clamp connections; wall thin; terminal cells semierect to erect, frequent. **Pileus trama** interwoven, ± loosely interwoven to ±- radially oriented; hyphae 2-12 µm diam, hyaline, inamyloid, with clamp connections; wall thin. **Lamellar trama** regular to irregular; hyphae 2-10(-16) µm diam, hyaline, inamyloid, with clamp connections; wall thin. **Basidia** (Fig. 40b) 18-26 X 5-7 µm, clavate; sterigmata four; basidioles 18-22 X 4-8 µm, clavate, subfusoid or slightly mucronate. **Pleurocystidia** absent. Lamellar margin sterile or fertile. **Cheilocystidia** (Fig. 40c) 18-48 X 4-11µm, clavate to broadly clavate, occasionally subfusoid, strangulate or flexed; apex obtuse, knobbed or with diverticulate projections, occasionally furcate. **Stipe epicutis** parallel; hyphae 2-10(-16) µm, frequently septate, yellowish in mass, hyaline singly, inamyloid, with clamp connections; wall up to 1 µm thick. Gloeopleurous hyphae occasional, up to 4 µm diam. **Caulocystidia** (Fig. 40d) up to 120 X 6 µm, cylindrical, ± strangulate, septate, with clamp connections; forming fascicles.

Basidiospores (Fig. 40e) 6.0-8.8(-10.4) X (2.8-)3.2-4.0 µm (n = 155/8, x = 7.5 X 3.6 µm, Q = 1.75-2.88, Qx = 2.08), narrowly ellipsoid or subcilindrical in side view, ellipsoid in face view, hyaline, inamyloid; wall smooth, thin.



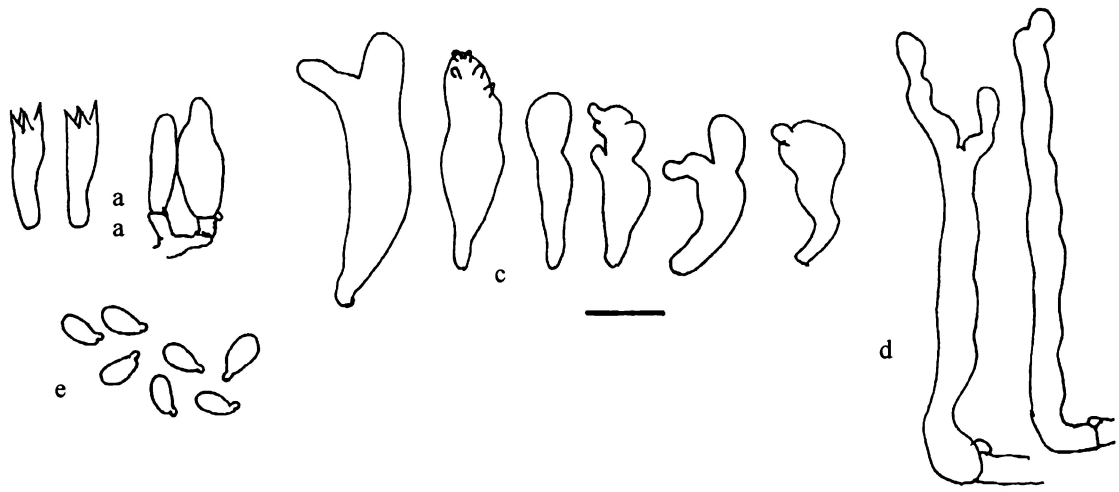


Fig. 40. *Gymnopus collybioides*. a. basidiomata (TFB 9690); b. basidia and basidioles; c. cheilocystidia; d. caulocystidia; e. basidiospores. Scale bar equals 10 μm for line drawings, 1 cm for picture.

Commentary: *Gymnopus collybioides* can be distinguished in the field by the small stature of basidiomata, with a very slender stipe that is proportionally long in relation to the pileus diameter, and white lamellae that are very crowded and do not form a pseudocollarium. Members of the *G. biformis* morphological complex are different because lamellae are not crowded and lamellae are off-white to cream. Specimens collected during this project agreed very well with Spegazzini's type specimen and description of the species, which differ somewhat from Singer's (1952) species circumscription.

In the examination of the holotype specimen, which is in poor condition (i.e. pileus tissues highly contaminated with molds) some putative spores were found (i.e. ellipsoid, hyaline and thin-walled) but cheilocystidia were hard to distinguish. Spores and cheilocystidia fell into the ranges and shapes observed in the specimens collected for this project. The pileus epicutis of the type was definitively a cutis and no pleurocystidia were found. Desjardin et al (1999) transferred this name to *Gymnopus* and considered it taxonomically related to *G. menehune* from Hawaii, and to *G. subcyathiformis* from México, while Singer (1952) considered it to be related to *G. confluens*. In my work, *G. collybioides* seems morphologically very similar to the *G. biformis* complex because of the structure of the pileus epicutis, cheilocystidia and spore shape and dimensions.

The specimens TFB 10491 and EFM 1369 were unusually large for the norm of this species and pilei of the specimens TFB 9692, TFB 10485, and TFB 10587 were more conico-umbonate.

Gymnopus collybioides appears phylogenetically closest to *G. biformis* var. *parvulus* and var. *lobatus* (Figs. 2-4). Together with *G. cylindricus* all of these species share leaf-litter habitat as a common characteristic.

Mating studies. A self-cross with SBIs from collection TFB 9690 resulted in a tetrapolar mating system (Fig. 41). Three mating types were identified. Mating types

		A_1B_1					A_2B_1		A_1B_2				
		1	2	3	7	9	8	16	13	17	4	6	12
	1*		-	-	-	-	-	-	-	-	-	-	-
A_1B_1	2			-	-	-	-	-	-	+	-	-	-
	3*	-	-		-	-	-	+	+	+	+	-	+
	7	-	-	-		-	-	-	+	+	-	-	+
	9	-	-	-	-		-	+	+	-	-	+	+
A_2B_1	8	-	-	-	-	-		+	-	-	-	+	+
	16*	-	-	+	-	+	+		-	+	+	+	+
A_1B_2	13	-	-	+	+	+	-	-		-	-	+	+
	17	-	+	+	+	-	-	+	-		+	-	+
	4	-	-	+	-	-	-	+	-	+		-	+
	6	-	-	-	-	+	+	+	+	-	-		-
	12	-	-	+	+	+	+	+	+	+	+	-	

Fig. 41. Self-cross pairings of SBIs from *Gymnopus collybioides* FB 9690. + = presence of clamp connections; - = absence of clamp connections; * = tester strains. Isolate numbers and assigned mating types top and side. SBIs 4, 6, 12, 17 may be amphithallic or product of two germinating spores.

A_1B_2 and A_2B_1 were assigned arbitrarily based on clamp connection formation between 3/7/8/9 X 13/16 and the third mating type was assigned as a subordinate. Several SBIs appeared to be amphithallic or a product of two germinating spores. The inability to produce a clear result in *G. collybioides* seems to be a pattern in other related taxa like *G. biformis* and its varieties and in *G. subnudus* (see comments in Petersen, 1995a). A self-cross with SBIs from specimen TFB 9687 was completely uninterpretable because no clamp connections were observed (data not shown).

Material examined: BRAZIL. Apiahy, IV.1888, col. J Puiggari, *no 2893*, (LPS, HT). COSTA RICA. Alajuela Prov., Grecia Co., Reserva Forestal de Grecia, Bosque del Niño, 10° 08'38" N, 84° 14'62" W, 1750 m, 27.VI.1998, col. JL Mata, *TFB 9645* (TENN 56606); Cartago Prov., Estrella, 5 km E. of Km 31 of Interamerican Hwy, vic. Estrella, 9° 46'04" N, 83° 57'19" W, 1685 m, 31.V.1994, col. RE Halling, E Franco, M Mata, TJ Baroni, B Strack, L Umaña, *REH 7213* (NY); same location, 16.VI.1999, col. JL Mata & R Muñoz, *TFB 10074* (TENN 58104); Palo Verde, 9° 46'59" N, 83° 56'42" W, 1700 m, 16.VI.1999, col. JL Mata & R Muñoz, *TFB 10080* (TENN) **S**; Puntarenas Prov., Coto Brus Co., Sabalito District, Hacienda La Amistad, 8° 54'22" N, 82° 47'40" W, 1330 m, 3.VII1998, col. RH Petersen & JL Mata, *TFB 9685* (TENN 56646); same location, 4.VII.1998, col. RH Petersen & JL Mata, *TFB 9690* (TENN 56651), *TFB 9692* (TENN 56653); vic. Palmira, 8° 56'59" N, 82° 50'02" W, 21.VI.2000, col. JL Mata & RH Petersen, *TFB 10491* (TENN 58599); Montes de Oro Co., vic. Monteverde, Cañitas, 10° 18' N, 84° 48' W, 1500 m, 17.VIII.1995, col. E Franco, M Mata, J Torres, *EFM 1369* (NY); vic. San Vito, OTS Biological Station, 8° 46'43" N, 82° 57'37" W, 19.VI.2000, col. JL Mata & RH Petersen, *TFB 10484* (TENN 58592), *TFB 10485* (TENN 58593); same location, 20.VI.2000, col. JL Mata & RH Petersen, *TFB 10487* (TENN 58595).

(22) *Gymnopus confluens* (Pers.: Fr.) Antonín, Halling & Noordeloos. 1997. Mycotaxon 63: 364. Fig. 42

≡ *Agaricus confluens* Pers.:Fries. 1821. Syst. Mycol. 1: 123.

≡ *Marasmius confluens* (Pers.) Ricken. 1911. Blatterp. Deutschl. 1-72.

≡ *Collybia confluens* (Pers.: Fr.) Kummer. 1871. Führ. Pilzk.: 117.

NEOTYPE: BELGIUM. prov. Namur. Grande Tinémont near Han-sur-Lesse, 26.IX.1974, col. ME Noordeloos, *no 7479*, (L) *teste* Antonín & Noordeloos, 1997, *n.v.*

Pileus 15-60 mm diam, at first obtusely conic to broadly campanulate, with age applanate or shallowly umbonate; surface moist, hygrophanous, even when moist, finely rugulose-subsulcate when dry, translucent, at disc brown (6E6-8), cinnamon brown (6D-E6-5), when moist, fading to light brown (6D5, 6C4-6) or grayish orange (5B3), near margin sometimes almost white; margin curved to uplifted, crenate, sometimes irregular, translucent; context extremely thin, white and unchanging, or pale tan, cream (2/3A2) with age. **Lamellae** adnexed, extremely crowded, narrow and thin, at first white, with age pale tan or grayish orange (5B3); margin fimbriate; lamellulae in several tiers of different lengths. **Stipe** 50-140 X 1-3(-6) mm, strict, ± equal to slightly broader at apex and base, or compressed and cleft; surface dry, pubescent overall with white hairs, ground color reddish brown (8E8-8E6), purple brown (8F8-9F8), dark brown or gray brown, near apex orange brown (6C8); interior hollow; consistency fibrous and touch. Basal mycelium binding substrate white to pale ochraceous. Odor slightly farinaceous or mild; taste mild. **Habitat** in vegetation of *Quercus spp;* gregarious, on humus (Fig. 42a).

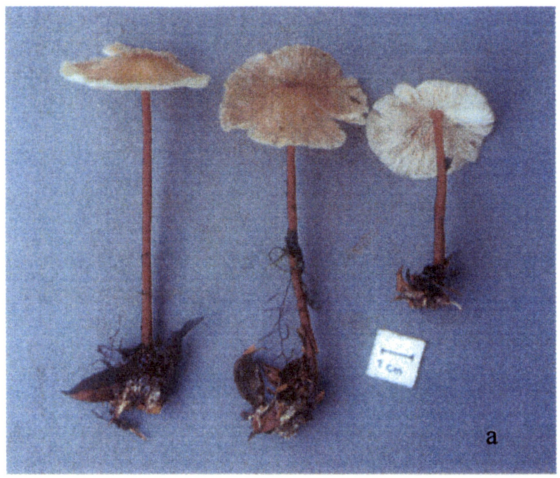
Pileus epicutis (Fig. 42b) a cutis; hyphae 2-6 μm diam, cylindrical, rarely diverticulate, not branched, not gelatinized, radially oriented, ± interwoven, pale yellow in mass, hyaline singly, inamyloid, with clamp connections; wall thin; terminal cells common, prostrate to erect, in fascicles. **Pileus trama** ± loosely interwoven; hyphae 3-14 μm diam, hyaline, inamyloid, with clamp connections; wall up to 0.8 μm thick. Gloeopleurous hyphae rare, up to 4 μm diam. **Lamellar trama** regular to subregular; hyphae 2-10 μm diam, hyaline, inamyloid, with clamp connections; wall thin.

Pleurocystidia absent. Lamellar margin sterile. **Basidia** (Fig. 42c) 21-28 X 5-6 μm, clavate; sterigmata four; basidioles 14-23 X 3-4 μm, clavate or cylindrical. **Cheilocystidia** (Fig. 42d) conspicuous, 14-53 X 5-11 μm, short or long clavate, submucronate, ± flexed, with clamp connections; apex frequently knobbed or lobed, occasionally furcate or ramose. **Stipe epicutis** parallel; hyphae 4-14(-20) μm, frequently septate, pale yellowish in mass, hyaline singly, inamyloid, with clamp connections; wall up to 1.6 μm thick. Gloeopleurous hyphae rare, up to 8 μm diam. **Caulocystidia** up to 200 X 6 μm, cylindrical, flexed, forming an entangled and interwoven mat of hyphae, with clamp connections; apex often furcate.

Basidiospores (Fig. 42e) white yellowish (2A2) in spore deposit, (6.4-)7.2-8.4(-9.6) X 3.2-4.8 μm (n = 85/4, x = 8.3 X 3.8 μm, Q = 1.80-2.63, Qx = 2.19), narrowly ellipsoid or lacrymoid in side view, subfusoid or subcilindrical in face view, hyaline, inamyloid; wall smooth, thin.

Commentary: Macro- and microcharacteristics match those reported for *G. confluens* (Halling, 1983; Antonín & Noordeloos, 1997). *Gymnopus confluens* may be confused with *G. polyphyllus* (Halling, 1983) based on my examination of the type specimen of the latter. Costa Rican specimens appear to exhibit more reddish brown to purple-brown colors in the stipe instead of orange-brown to gray-brown. Field notes for the Costa Rican specimens show a gregarious habit, a condition also reported for specimens from Europe (Antonín and Noordeloos, 1997). When examined in dried condition specimens from United States appeared very similar to those from Costa Rica. No SBIs were obtained from any collection made in Costa Rica.

Costa Rican *G. confluens* appears not phylogenetically closely related *G. confluens* from the United States (Figs. 2-4). Pairwise sequence comparisons, however, of *G. confluens* from Costa Rica and United States resulted in 91% similarity. Costa Rican *G. confluens* seems to be more related to that of *G. neotropicus*.



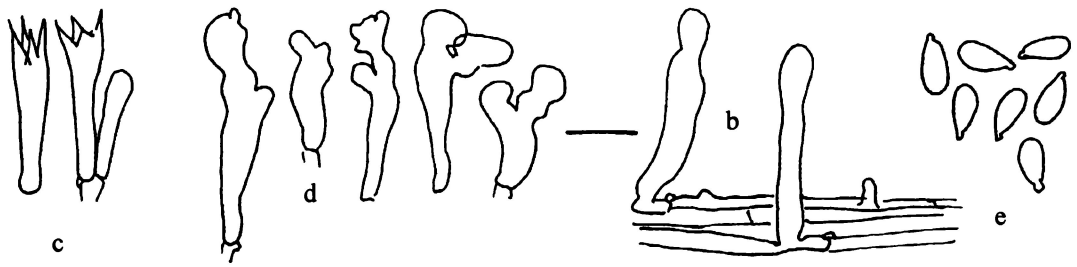


Fig. 42. *Gymnopus confluens*. a. basidiomata (TFB 11005); b. elements of pileus epicutis; c. basidia and basidiole; d. cheilocystidia; e. basidiospores. Scale bar equals 10 μm for line drawings.

Material examined: COSTA RICA. San José Prov., Dota Co., San Gerardo de Dota. ± 500 m along road from Interamerican Highway toward San Gerardo, 9° 42'52" N, 83° 58'28" W, 3000 m, 17.X.1994, col. RE Halling, *REH 7379* (NY) **S**. Pérez Zeledón Co., Villa Mills, Finca Alejandrina, along Interamerican Hwy at km 95 near Hotel La Georgina, 9° 33'43" N, 83° 44'22" W, 3000 m, 23.VI.1994, col. RE Halling, E. Franco, M Mata, L Umaña, *REH 7341* (NY); Estación Experimental CATIE, 9° 33'03" N, 83° 40'56" W, 2880 m, 20.VI.1999, col. JL Mata, *TFB 10411* (TENN 58106); same location, 23.VI.2000, col. JL Mata & RH Petersen, *TFB 11005* (TENN 58613). Puntarenas Prov., Sabalito, Estación Biológica Las Alturas, 8° 56'59" N, 82° 50'02" W, 1420 m, 26.V.1994, col. E Franco-Molano, M Mata, J Johnson, L Umaña, *EFM 1191* (NY). FRANCE. Rhone-Alpes, Dept. Savoire (73) Commune St. Germain La Chambotte, 45° 46' N, 5° 53' E, 10.IX.2001, col. RH Petersen & PA Moreau, *TFB 11335* (TENN 59219). UNITED STATES. New York, Tompkins Co., near Dryden, Ringwood Preserve, 13.IX.1994, col. RE Halling, *REH 3851* (NY); North Carolina, Macon Co., Standing Indian Campground, 35° 04' N, 83° 25' W, col. RE Halling, *REH 6929* (NY); same location, JL Mata & RH Petersen, *TFB 10010* (TENN 56722); short of Mt. Area, 18.VII.1994, col. J Johnson, TFB 7219 (TENN 53522) **S**; Franklin, 35° 4'30" N, 83° 31'48" W, 16.VII.1999, col. JL Mata, *TFB 10472* (TENN 58166).

(23) *Gymnopus cylindricus* Mata, *sp. nov.* Fig. 43

Pileo plano-convexo, glaber, auarntiaco-brunneus. Lamellae adnatae ad adnexas, angustas, confertes, cremicolores. Stipite equalis, concolore pileo ad roseo-brunneo. Sporis acicularis, 8.8-13.6 X 3.2-4.8 µm, cheilocystidia cylindricales, septates.

HOLOTYPE [*hic designatus]*: COSTA RICA. San José Prov. Dota Co., San Gerardo de Dota, Hotel de Montaña Savegre, 9° 32'71" N, 83° 48'39" W, 2300 m, 19.VI.1999, col. JL Mata, *TFB 10402* (TENN).

Etymology: *cylindricus* (Latin) = cylindrical shape of cheilocystidia.

Pileus 8-30 mm diam, convex, plano-convex, with a broad and flattened umbo, or centrally depressed; surface glabrous, radially wrinkled, becoming ± translucent outwards, when young brown (6C6) to dark brown (7F8), with age brown (7E8), light brown (7C5), orange brown (5C7, 6D7) to tan (6C6-6D7), near or at margin always paler than disc, light brown to creamy (4A3); margin curved to plane, striate-crenate, translucent; context thin, concolorous to surface. **Lamellae** adnate to adnexed, very narrow, crowded, at first white, with age off-white (3A2) to cream (3A4, 4A3); margin even; lamellulae in several tiers of different lengths. **Stipe** 40-100 X 1-2(-4) mm, equal, terete or compressed; surface overall white-pruinose, reddish brown (8E6, 7E8), orange brown (6E6) to brown (6D8-6E8), darker than pileus, at apex fading to light brown (7D8); interior hollow; consistency fleshy, brittle. Odor not distinctive or mild; taste not distinctive. **Habitat** on leaf litter; solitary to gregarious (Fig. 43a).

Pileus epicutis (Fig. 43b) a cutis; hyphae 2-8 µm diam, cylindrical, frequently with diverticula, pigment-incrusted, straw colored in mass, orange brown in IKI, hyaline singly, inamyloid, with clamp connections; wall thin; terminal cells occasional, semi-erect to erect, originating at hyphal septa. Gloeopleurous hyphae rare, up to 10 µm diam. **Pileus trama** interwoven; hyphae 2-12 µm diam, hyaline, inamyloid, with clamp connections; wall thin. **Lamellar trama** subregular to irregular; hyphae 2-10 µm diam, hyaline, inamyloid, with clamp connections; wall thin; subhymenium pseudparenchymatous. **Basidia** (Fig. 43c) 20-29 X 5-8 µm, clavate; sterigmata four; basidioles similarly sized as basidia, clavate to mucronate. **Pleurocystidia** absent. Lamellar margin sterile. **Cheilocystidia** (Fig. 43d) 24-65 X 6-9 µm, cylindrical, ± flexed, septate, with clamp connections; apex obtuse, submucronate or lobed. **Stipe epicutis** parallel; hyphae 4-12(-16) µm diam, cylindrical, brown in mass, pale yellow singly, inamyloid, with clamp connections; wall thin to 1 µm thick. **Caulocystidia** (Fig. 43e) up to 170(-300) X 6(-8) µm, cylindrical, septate, with clamp connections, forming an entangled mat; apex obtuse, knobbed or furcate.

Basidiospores (Fig. 43f) (8.8-)9.6-11.2(-13.6) X 3.2-4.0(-4.8) μm (n = 92/5, x = 10.4 X 3.7 μm, Q = 2.10-3.63, Qx = 2.85) subfusoid to aciculate in side view, narrowly ellipsoid to cylindrical in face view, hyaline, inamyloid; wall smooth, thin.

Commentary: *Gymnopus cylindricus* is characterized by the cylindrical shape of the cheilocystidia in combination with aciculate spores. Basidiomata of *G. alnicola* also produce narrow elliptical spores but cystidia are clavate to sphaeropedunculate. Macroscopically *G. cylindricus* is very similar to *G. confluens* and *G. polyphyllus*, but orange colors of the stipe and lack of a sweet-like odor (i.e. fenugreek) distinguish it from these two, respectively. Microscopically, *G. cylindricus* is similar to *G. polyphyllus* in that the hyphae of the pileus epicutis are frequently diverticulate. Pilei of *G. subpruinosus,* a species not yet reported from Costa Rica, are smaller and conspicuously sulcate, and lamellae more greyish orange (Desjardin et al, 1999).

Gymnopus cylindricus appears phylogenetically related to species in the Costa Rican *G. biformis* complex (Figs. 2-4).

Material examined: COSTA RICA. San José Prov., Dota Co., Jardín de Dota, 3.5 km W off Inter American Hwy, 9° 42'42" N, 83° 53'28" W, 2300 m, 18.VI.1999, col. JL Mata, *TFB 10091* (TENN 58086); Finca El Jaular, 9° 39' 39" N, 83° 52' 01" W, 2300 m, 17.VI.1999, col. JL Mata, *TFB 10084* (TENN 58024); San Gerardo de Dota, Hotel de Montaña Savegre, 9° 32'71" N, 83° 48'39" W, 2300 m, 29.VI.1998, col. RH Petersen & JL Mata, *TFB 9664* (TENN 56625); same location 19.VI.1999, col. JL Mata, *TFB 10402* (TENN 58097) **S**; Pérez Zeledón Co., Villa Mills, Estación Experimental CATIE, 9° 33'03" N, 83° 40'56" W; 2880 m, 30.VI.1998, col. JL Mata, *TFB 9671* (TENN 56632).



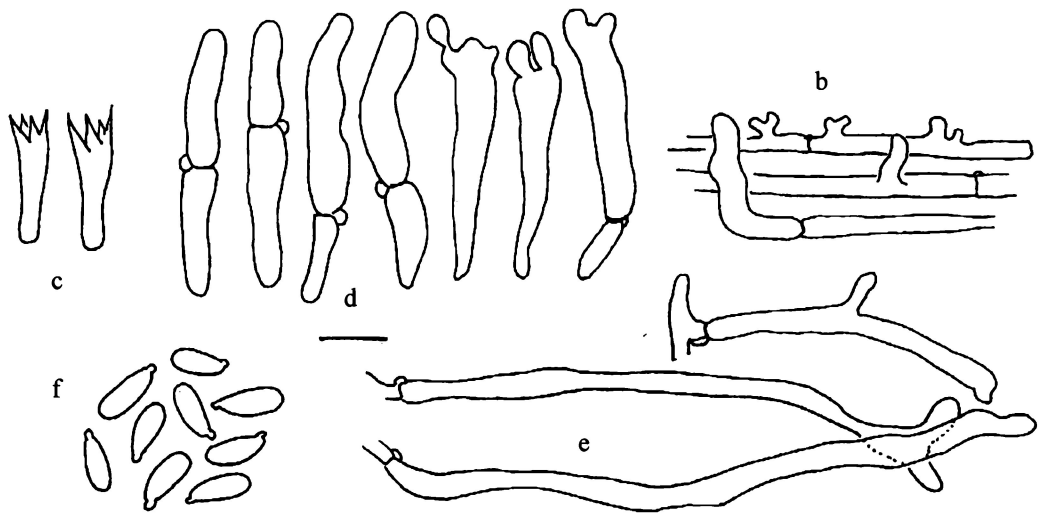


Fig. 43. *Gymnopus cylindricus*. a. basidiomata (TFB 9664); b. elements of pileus epicutis; c. basidia; d. cheilocystidia; e. caulocystidia; f. basidiospores. Scale bar equals 10 µm for line drawings, 1 cm for picture.

(24) *Gymnopus dichrous* (Berk. & Curt.) Halling. 1997. Mycotaxon 63: 364. Figs. 44-46

≡ *Marasmius dichrous* Berkeley & Curtis. 1853. Ann. Mag. Nat. Hist. II, 12: 426.

≡ *Collybia dichrous* (Berk. & Curt.) Gilliam. 1976. Mycotaxon 4(1): 130.

HOLOTYPE: UNITED STATES. South Carolina, Society Hill, VII.1840, *Curtis 2834*, (K), *teste* Halling (1983), *n.v.*

Pileus up to 30 mm diam, broadly convex to applanate, shallowly umbonate; surface radially fibrillose, appearing silky, dry, opaque, sulcate halfway toward margin, at center buff brown (6D6), outward pale brown (5/6C4); margin curved, sulcate; context thin, off white. **Lamellae** adnexed at first, seceding into a pseudocollarium, 2 mm broad (± narrow), subdistant, cream grayish (3A/B2-4A/B2); margin serrulate; lamellulae in three tiers of different lengths. **Stipe** up to 50 X 3 mm, equal, terete, curved; surface finely velvety, hygrophanous, near apex cream colored, downwards brown (6D7); interior hollow; consistency ± cartilaginous. Odor and taste mild. **Habitat** on wood; solitary (Fig. 44a).

Pileus epicutis (Fig. 44b) a cutis; hyphae 2-16 µm diam, cylindrical, repent, radially oriented, ± interwoven, occasionally diverticulate, pigment-incrusted, banded, light brown in mass, orange in IKI, hyaline singly, inamyloid, with clamp connections; wall thin. **Pileus trama** interwoven; hyphae 4-18 µm diam, hyaline, inamyloid, with clamp connections; wall thin. **Lamellar trama** regular; hyphae 2-8 µm diam, hyaline, inamyloid, with clamp connections; wall thin. **Basidia** (Fig. 44c) 29-40 X 6-8 µm, clavate; sterigmata four; basidioles 29-38 X 6-8 µm, clavate. **Pleurocystidia** absent. **Cheilocystidia** (Fig. 44d) 32-60 X 10-18 µm, inflated, clavate to broadly clavate, sphaeropedunculate; apex obtuse or with diverticula. **Stipe epicutis** parallel; hyphae 4-12 µm diam, straw colored in mass, hyaline singly, inamyloid, with clamp connections; wall up to 1 µm thick. **Caulocystidia** arising from a dense mat of hyphae, up to 60 X 17 µm diam, inflated, broadly clavate; apex often with long diverticulate projections.

Basidiospores (Fig. 44e) 8.8-12.8 X 4.0-5.6 μm (n = 20, x = 9.9 X 4.7 μm, Q = 1.71-2.45, Qx = 2.13) pip shaped to lacrymoid, or obovoid in side view, ellipsoid in face view, hyaline, inamyloid; wall thin, smooth; contents often guttulate.

Commentary: Although a tuberculate knob was not noticed in this collection, all other field characteristics of the basidiome were similar to those described by Halling (1986) and other collections from the United States. Most microscopic elements observed in the Costa Rican specimen were similar to those described by Halling (1983), especially cheilocystidia and caulocystidia, which were identical in shape and form, whereas spore and hyphal measurements of the sole Costa Rican specimen were somewhat broader, and basidia longer. Description of the type specimen of *G. dichrous* (Desjardin, 1989) also matched characteristics observed in the Costa Rican specimen. Lignicolous habitat is as reported for *G. dichrous* in the United States and *G. dichrous* is distinguished from other species by its voluminous cheilocystidia. *Gymnopus dichrous* is a new report for the Costa Rican mycoflora.

Costa Rican *G. dichrous* appears in the same clade as *G. luxurians*, *G fibrosipes*, and *G. pseudo-omphalodes*. Pairwise sequence comparison between Costa Rican and United States *G. dichrous* resulted in 86% similarity. *Gymnopus dichrous* from the United States appears to be basal to this clade (Figs. 2-4).

Mating studies. The self-cross of the SBIs of this single specimen resulted in a tetrapolar mating system (Fig. 45), already reported by Petersen (1995a). Four mating types were identified. Mating types A_1B_2 and A_2B_1 were assigned based on clamp connection formation in 3/6/8/12/14 X 1/7/9/11/15. Mating types A_1B_1 and A_2B_2 were assigned as subordinates. Pairings of 10 X 3/8/12/14/16/1/7/9/11/15 rendered a crevasse. Costa Rican *G. dichrous* is incompatible with North American specimens but also intercollection pairings of United States specimens yielded a low percentage of intercollection compatibility (Fig. 46).

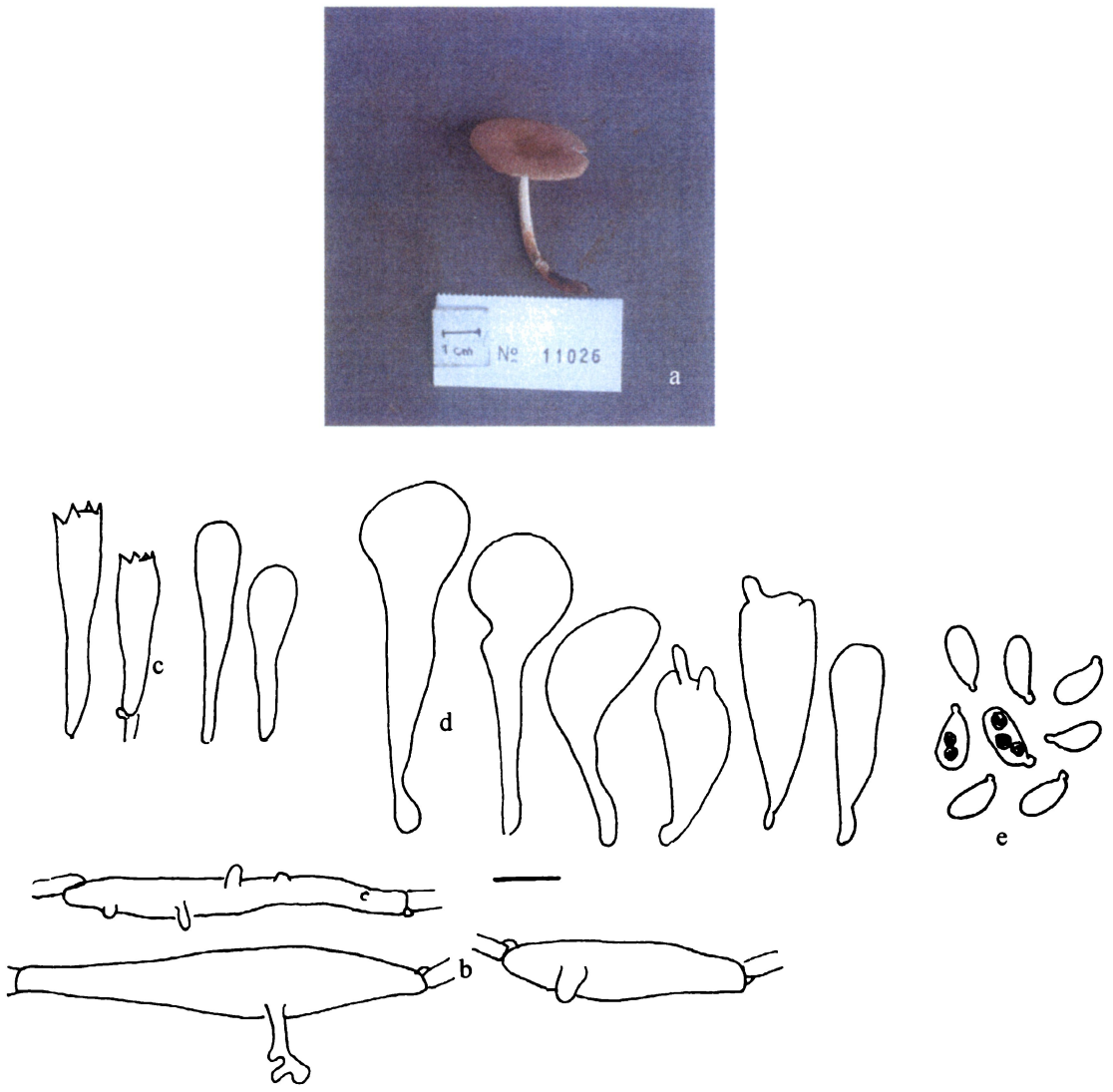


Fig. 44. *Gymnopus dichrous*. a. basidioma (TFB 11026); b. elements of pileus epicutis ; c. basidia and basidioles; d. cheilocystidia; e. basidiospores. Scale bar equals 10 µm for line drawings.

						A_1B_2					A_2B_1	A_1B_1	A_2B_2
		3	8	12	14	6	1	7	9	11	15	4	10
	3*		-	-	-	-	+	+	+	+	+	-	-
	8	-		-	-	-	+	+	+	+	+	-	-
	12	-	-		-	-	+	+	+	+	+	-	-
	14	-	-	-		-	+	+	+	+	+	-	-
A_1B_2	6	-	-	-	-		-	+	+	+	+	-	-
	1	+	+	+	+	-		-	-	-	-	-	-
	7*	+	+	+	+	+	-		-	-	-	-	-
	9	+	+	+	+	+	-	-		-	-	-	-
	11	+	+	+	+	+	-	-	-		-	-	-
A_2B_1	15	+	+	+	+	+	-	-	-	-		-	-
A_1B_1	4*	-	-	-	-	-	-	-	-	-	-		+
A_2B_2	10*	-	-	-	-	-	-	-	-	-	-	+	

Fig. 45. Self-cross pairings of SBIs from *Gymnopus dichrous* TFB 11026. + = presence of clamp connections; - = absence of clamp connections; * = tester strains. Isolate numbers and assigned mating types top and side.

		1871			
United States	1871		10009		
United States	10009	0/4		10014	
United States	10014	0/4	3/4		11026
Costa Rica	11026	0/4	0/4	0/4	

Fig. 46. Intercollection pairings from *Gymnopus dichrous* from Costa Rica and United States. Numbers indicate ratio of pairings where clamp connections were observed.

Material examined: COSTA RICA. San José Prov., Dota Co., 12 km S from Copey on the road to Providencia, 9° 35'19" N, 83° 53'00" W, 2795 m, 28.VI.2000, col. JL Mata & RH Petersen, RE Halling, L Norvell, *TFB 11026* (TENN 58634) **S**. UNITED STATES. North Carolina, Avery Co., Grandfather Mt, along US 221, 1 mile S of main entrance, 24.VII.1998, col. EB Lickey, *TFB 10002* (TENN 56714); Macon Co., Highlands, Rte 28, 6.VII.1989, col. RH Petersen, *TFB 1871* (TENN 48443) **S**; Horse Cave Dr., Bullpen Rd, 35° 1' N, 83° 8' W, 1000 m, 28.VII.1998, col. Q Wu, *TFB 10009* (TENN 56721); Standing Indian Campground, 35° 04'48" N, 83° 25'94" W, 29.VII.1998, col. RH Petersen & JL Mata, *TFB 10013* (TENN 56725), *TFB 10014* (TENN 56726), *TFB 10016* (TENN 56728), *TFB 10017* (TENN 56729).

Gymnopus luxurians (Peck) Murrill. [1916. N. Amer. Fl. 9: 362.] UNITED STATES. North Carolina, Jackson Co., Whitesides Cove Rd., Summer Chapel Trail, 12.VII.1999, col. E Lickey, *TFB 10350* (TENN 57910) **S**. Macon Co., Highlands, 35° 01'10" N, 83° 14'44" W, 14.VII.1999, col. JL Mata, *TFB 10465* (TENN 58159). Tennessee, Knox Co., Knoxville, 2.VII.1999, col. JL Mata, *TFB 10448* (TENN 58142); same location, 25.V.2000, col. KW Hughes & RH Petersen, *TFB 10476* (TENN 58539); Sevier Co., Sevierville, 1.VII.1999, col. RH Petersen, *TFB 10447* (TENN 58141). Sevier Co., 4.VII.1999, col. E Lickey, *TFB 10449* (TENN 58143).

(25) *Gymnopus fibrosipes* (Berk. & Curt.) Mata, *comb. nov.* Figs. 47-48

≡ *Marasmius fibrosipes* Berk. & Curt. 1868. J. Linn. Soc.(Bot.), 10: 293.

≡ *Collybia fibrosipes* (Berk. & Curt.) Dennis. 1951. Trans. Br. Mycol. Soc. 34: 448.

HOLOTYPE: CUBA, Wright 3, on dead wood, (K) [!].

= *Gymnopus oculatus* Murrill. 1916. N. Amer. Flora, 9: 371.

≡ *Collybia oculata* (Murrill) Murrill. 1916. Mycologia 8: 219.

HOLOTYPE: JAMAICA. Troy and Tyre, Cockpit Co., 610 m (2000 ft), 12-14.I.1909, col. WA Murrill & W Harris, *no. 1022*, (NY) [!].

Pileus 20-60 mm diam, at first obtusely campanulate, conico-convex, then broadly convex to plane; surface moist, radially appressed-fibrillose, hygrophanous, at center and with age orangish (5C6-5B7), outwards whitish (4A4-3A3); margin undulating to flared, becoming striated; context thin, whitish, turning a little pinkish when cut. **Lamellae** adnexed to seceding, very narrow, crowded, white to white yellowish (2A2); margin even to finely fimbriate; lamellulae in several tiers of different lengths. **Stipe** 55-100 X 4-10 mm, equal, downwards tapering, compressed to cleft, twisted; surface fibrillose, striate to sulcate, at apex whitish (3A3-2A2), downwards pinkish brown (6B3); interior hollow, similarly colored as surface; consistency fibrous, tough. Odor mild or spicy; taste to radish or bitter. **Habitat** on decaying litter, in *Quercus spp.* vegetation; cespitose (Fig. 47a).

Pileus epicutis a simple cutis; hyphae 2-8 μm diam, cylindrical, repent, radially oriented, not interwoven, pale yellowish to tan in mass, hyaline singly, inamyloid, frequently septate, with clamp connections; wall thin; surface hyphae a little gelatinized. **Pileus trama** loosely interwoven, forming a lacunar tissue (in cross section); hyphae 4-22 μm diam, hyaline, inamyloid, with clamp connections; wall thin. Gloeopleurous hyphae, 4-8 μm diam. **Lamellar trama** regular to subregular, becoming interwoven towards margin; hyphae of mediostratum 8-16 μm diam, hyaline, with clamp connections, narrowing at septum; wall thin; subhymenium ± 5-10 μm thick, densely interwoven, composed of small cells, hyaline. **Basidia** (Fig. 47b) 24-32 X 6-8 μm, clavate; sterigmata four, rarely two; basidioles numerous, 24-28 X 5-6 μm, clavate. **Pleurocystidia** absent. Lamellar margin sometimes fertile, split dichotomously. **Cheilocystidia** (Fig. 47c) obscure, 16-36 X 5-8 μm, clavate, fusoid, lobed, hyaline; wall thin, smooth; apex obtuse, forked or knobbed. **Stipe epicutis** parallel; hyphae 4-16 μm, inner hyphae broader, hyaline to slightly tan colored, with clamp connections; wall thin. Gloeopleurous hyphae 6-10 μm diam. **Caulocystidia** present as a vesture of entangled hyphae, some with short erect terminal cells.

Basidiospores (Fig. 47d) pale yellow (2A2) in mass, 6.4-8.8 X 3.2-4.0 μm (n = 36, x = 7.6 X 3.9 μm; Q = (1.54-)1.78-2.44, Qx = 1.97), pip shaped to amygdaliform in side view, ellipsoid in face view, hyaline, inamyloid; wall smooth, thin; hilar appendix visible.

Commentary: Basidiomata of this specimen are macroscopically similar to those described for *C. fibrosipes* (Dennis, 1951; Guzmán, 1979; Pegler, 1983b) in terms of shape and color of basidiomata, and the cespitose habit, an important feature. However, spores, cystidia and pileipellis deviate somewhat from previous descriptions by other authors. Basidiomata of the holotype specimen are blackened and pressed flat, but lamellae appear to be seceding, a feature observed in the Costa Rican material. The mean spore measurement from the holotype is 5.4 X 3.0 μm (Qx = 1.80) which is about two microns less in length than for the Costa Rican material. Observation of cheilocystidia was obscured by the growth of molds and erosion due to preservation conditions and their presence was not confirmed. Basidiomata of the type specimen of *G. oculatus* were much smaller but colors described in the original description for pileus, and especially lamellae (Murrill, 1916), were very similar when comparing them to the Costa Rican specimen. In spite of spore dimensions being somewhat smaller (x = 6.1 X 3.2 μm Qx = 1.90) than those produced by basidiomata of *G. oculatus*, their shape was identical (i.e. lacrymoid) to those of the sole Costa Rican specimen. Cheilocystidia were also very similar in shape and were inconspicuous in both the type of *G. oculatus,* and in the Costa Rican specimen. Moreover, Dennis (1951) reported "cystidia wanting." The kind of hyphal arrangement in the pileus epicutis places this specimen under *Gymnopus*, so a new combination for the species epithet is proposed.

Mating studies. *Gymnopus fibrosipes* exhibits a tetrapolar mating system according to the self-cross performed with 12 SBIs (Fig. 48). Only three mating types were identified. Mating types A_1B_2 and A_2B_1 were assigned arbitrarily based on presence of clamp connections in 9/11/25 X 5/17/23 and A_1B_1 as a subordinate. SBI 1 may be amphithallic or the product of two germinating spores.





Fig. 47. *Gymnopus fibrosipes*. a. basidiomata (TFB 9699); b. basidia; c. cheilocystidia; d. basidiospores. Scale bar equals 10 μm for line drawings, 1 cm for picture.

Material examined: COSTA RICA. Puntarenas Prov, Coto Brus Co., Sabalito, Hacienda La Amistad, on the road to Contoncito, 8° 54'22" N, 82° 47'40"W; ± 1330 m, col. RH Petersen, RE Halling & JL Mata, *TFB 9699* (TENN 56660) **S**, *REH 7819* (NY). CUBA, Wright 3 (K, HT of *Marasmius fibrosipes*). CUBA. Wright 3, on dead wood, (K, HT of *M. fibrosipes*). JAMAICA. Troy and Tyre, Cockpit Co., 610 m, 12-14.I.1909, col. WA Murrill & W Harris *no. 1022*, (NY, HT of *Gymnopus oculatus*).

(26) *Gymnopus neotropicus* (Singer) Mata, *comb. nov.* Fig. 49

≡ *Collybia neotropica* Singer. 1962. Sydowia 15: 54.

HOLOTYPE: BOLIVIA. La Paz, Nor-Yungas, Coroico, 15.II.1956, col. R Singer, *B 1173* (LIL) [!].

Pileus 50-90 mm diam, broadly convex, umbonate; surface glabrous to finely fibrillose, sulcate entirely, at umbo at first light orange-brown (6D4-7, 6C4), with age reddish brown (7E8-8E8), outwards pale brown, cream (5A2) to almost off white, troughs darker in color than crests, translucent; margin curved, sulcate, translucent; context up to 3(-8) mm diam, white or concolorous to surface. **Lamellae** adnate, sometimes with a decurrent tooth, up to 7(-12) mm broad, subdistant to distant, white, cream (4A2) or grayish yellow (4B4); margin even to uneven; lamellulae in two tiers of different lengths. **Stipe** 60-170 X 4-8 mm, ± equal, cylindrical to slightly flattened, sometimes geniculate, base ± bulbous; surface fibrillose, entirely white-pruinose, more dense at apex, near apex concolorous to lamellae, downwards orange-brown (6C5), reddish brown (7E8-8-7E6) to pale gray brown or tan, sometimes with age blackening; interior pithy to hollow; consistency fibrous or ± cartilaginous, tough or brittle. Basal and binding mycelium white to yellowish. Odor fungous or none; taste mild or none. **Habitat** on decomposed leaf litter/soil, within a *Chusquea* spp patch, along trail; scattered (Fig. 49a).

				A_1B_2			A_2B_1					A_1B_1	$A_1B_2 + A_2B_2$
		11	9	25	17	23	5	4	7	14	24	8	1
	11*		-	-	+	+	-	-	-	-	-	-	-
	9	-		-	-	-	+	-	-	-	-	-	-
A_1B_2	25	-	-		+	-	+	-	-	-	-	-	-
	17*	+	-	+		-	-	-	-	-	-	-	+
	23	+	-	-	-		-	-	-	-	-	-	+
A_2B_1	5	-	+	+	-	-		-	-	-	-	-	-
	4	-	-	-	-	-	-		-	-	-	-	+
	7	-	-	-	-	-	-	-		-	-	-	+
	14*	-	-	-	-	-	-	-	-		-	-	+
	24	-	-	-	-	-	-	-	-	-		-	+
A_1B_1	8	-	-	-	-	-	-	-	-	-	-		-
$A_1B_2 + A_2B_2$	1	-	-	-	+	+	-	+	+	+	+	-	

Fig. 48. Self-cross pairings of SBIs from *Gymnopus fibrosipes* FB 9699. + = presence of clamp connections; - = absence of clamp connections; * = tester strains. Isolate numbers and assigned mating types top and side. SBI no 1 is interpreted as amphithallic or the product of two germinating spores.

Pileus epicutis (Fig. 49b) a cutis; hyphae 2-14(-20) µm diam, cylindrical, radially oriented, loosely arranged, not branched, pale brown to straw colored in mass, hyaline singly, orangish in IKI, with clamp connections; wall thin; terminal cells, erect to semi-erect, frequently diverticulate, forming fascicles; wall up to 0.8 µm. **Pileus trama** interwoven to ± radially oriented; hyphae 2-14(-20) µm diam, hyaline, inamyloid, with clamp connections; wall thin. **Lamellar trama** regular to subregular; hyphae 2-12(-16) µm diam, hyaline, pale orangish in IKI, with clamp connections; wall thin; subhymenium parenchymatous. **Basidia** (Fig. 49c) 26-40 X 6-8(-10) µm, long clavate; sterigmata four; basidioles 19-28 X 4-6 µm, mostly clavate. **Pleurocystidia** absent. Lamellae edge sterile or fertile. **Cheilocystidia** (Fig. 49d) 38-56(-68) X 6-12 µm, clavate, saccate to sphaeropedunculate, with clamp connections; apex obtuse, lobed or furcate, sometimes with appendages. **Stipe epicutis** parallel; hyphae 2-24 µm diam, pale yellowish in mass, hyaline singly, inamyloid, with clamp connections; wall up to 1 µm thick. **Caulocystidia** up to 120 X 6 (-12) µm, clavate, cylindrical, clavate to ampullaceous, with clamp connections, densely entangled; apex obtuse.

Basidiospores (Fig. 49e) 7.2-9.6(-10.4) X 3.6-5.2 µm (n = 80/4, x = 8.4 X 4.3 µm, Q = 1.64-2.44, Qx = 1.97) lacrymoid to pip shaped in side view, ellipsoid to obovoid in face view, hyaline, inamyloid; wall smooth, thin.

Commentary: Basidiomata of this species can be recognized by their large stature, sulcate pileus with relatively distant lamellae and a somewhat fragile consistency. A morphologically similar species is *G. lodgeae*, basidiomata of which can be distinguished by a much darker and fibrous stipe and microscopically by the presence of pleurocystidia. The pileus epicutis in *G. neotropicus* has terminal hyphae with distinct erect diverticula, like those of *G. omphalodes,* a much smaller statured mushroom.

Most morphological characteristics observed in Costa Rican specimens of *G. neotropicus* are similar to those in previous descriptions for other countries (Franco-

Molano et al, 2000; Pegler, 1983b). In Costa Rica, *G. neotropicus* has been collected at different elevations in all mountain ranges.

The type specimen of *G. neotropicus* is in bad condition and lacks the stipe. Dimensions and shape of basidia, basidioles, and spores match those described for Costa Rican specimens. Cheilocystidia observed in the type specimen are narrower and shorter than those of Costa Rican specimens. Diverticula in the hyphae of the pileus epicutis of the type could not be observed. The study of the type specimen agrees with current morphological concepts in *Gymnopus* and a new combination is proposed.

Gymnopus neotropicus from Costa Rica appears phylogenetically related to North American and Costa Rican *G. confluens* and other leaf litter-dwelling species in sect. *Vestipedes* (Figs. 2-4).

Material examined: BOLIVIA. La Paz, Nor-Yungas, Coroico, 15.II.1956, col. R Singer, *B 1173* (LIL, HT). COSTA RICA: Alajuela Prov., San Ramón, Caserío Colonia Palmareña, Reserva Forestal de San Ramón, 1000 m, 23.VII.1993, col. RE Halling, GM Mueller & E Pine, *REH 7053* (NY). Cartago Prov., Estrella, 5 km East of Interamerican Hwy at Km 31, 9° 46'5" N, 83° 57'20" W, 1717 m, 21.VII.1993, col. RE Halling, GM Mueller & E Pine, *REH 7046* (NY); same location 15.XI.1993, col. RE Halling, E Franco, L Umaña & JL Mata, *REH 7168* (NY). Guanacaste Prov., Parque Nacional Santa Rosa, Santa Rosa, bosque húmedo primario, 10° 51'04" N, 85° 36'26" W, 330 m, 6.III.1994, col. RE Halling, M Mata, TJ Baroni, GM Mueller, B Strack & L Umaña, *REH 7226* (NY). San José Prov., Pérez Zeledón Co., vic. Villa Mills, Estación Biológica Cuericí, 9° 33'17" N, 83° 40'04" W, 2560 m, 21.VI.1999, col. JL Mata, *TFB 10416* (TENN 58110) **S**; El Jaular, 9° 33'17" N, 85° 40'15" W, 2900 m, 24.VIII.1995, col. E Franco, J Torres & M Mata, *EFM-1431* (NY).



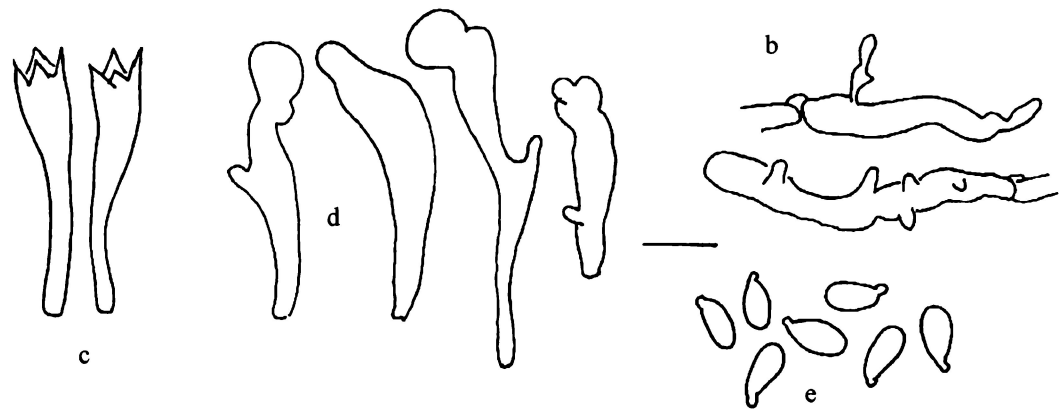


Fig. 49. *Gymnopus neotropicus*. a. basidiomata (TFB 10416); b. elements of pileus epicutis ; c. basidia; d. cheilocystidia; e. basidiospores. Scale bar equals 10 µm for line drawings.

(27) *Gymnopus polyphyllus* (Pk.) Halling. 1997. Mycotaxon 63: 365. Fig. 50

≡ *Marasmius polyphyllus* Pk. 1898. Ann. Rept. N.Y. State Mus. 51: 286.

≡ *Collybia polyphylla* (Pk.) Singer ex Halling. 1983. Mycologia Mem. 8: 85.

HOLOTYPE: UNITED STATES, New York, Essex Co., Minerva, VII.1902, col. CH Peck, (NYS)[!].

Pileus 20-50 mm diam, conico-convex, convex-plane to uplifted; surface glabrous, wet, hygrophanous, translucent, grayish orange (6B5) to caramel brown (6D7) overall or darker at the center, outwards fading to buff or yellowish, often with dark brown spots; margin curved, striate; context 2-3 mm thick, concolorous to surface unchanging. **Lamellae** adnate to adnexed, less than 1 mm broad, crowded, off-white to concolorous with pileus surface; margin even; lamellulae in several tiers of different lengths. **Stipe** 60-150 X 2-4 mm, central, equal to slightly tapered toward base, twisted; surface glabrous to fibrillose, brownish orange (5C3-5C4) to purple brown, upwards light brown, at base sometimes blackening; interior hollow, concolorous. Basal mycelium white. Odor faintly sweetish or fenugreek; taste bitterish. **Habitat** on soil, in mixed forest with *Quercus* spp; cespitose to gregarious (Fig. 50a).

Pileus epicutis (Fig. 50b) a cutis; hyphae 2-9 μm diam, cylindrical, not branched, occasionally diverticulate, not gelatinized, radially oriented, ± interwoven, pale yellow in mass, hyaline singly, inamyloid, with clamp connections; wall thin; terminal cells semi-erect to erect, common. **Pileus trama** ± loosely interwoven to ± radially oriented; hyphae 4-12 μm diam, hyaline, inamyloid, with clamp connections; wall thin to 1 μm thick. **Lamellar trama** regular; hyphae 2-10 μm diam, cylindrical or barrel shaped, hyaline, inamyloid, with clamp connections; wall thin. **Basidia** (Fig. 50c) 16-24 X 4-7 μm, clavate; sterigmata four; basidioles numerous, clavate or mucronate. **Pleurocystidia** absent. Lamellar margin sterile. **Cheilocystidia** (Fig. 50d) conspicuous, 20-42 X 4-8 μm, long clavate, cylindrical, ± flexed. with clamp connections; apex frequently knobbed. **Stipe epicutis** parallel; hyphae 4-22 μm, occasionally diverticula, light brown to pale yellowish in mass, hyaline singly, inamyloid, with clamp connections; wall up to 1.6 μm

thick. **Caulocystidia** up to 50 X 13 μm, cylindrical to strangulate, interwoven in a mat, with clamp connections.

Basidiospores (Fig. 50e) yellowish (2A2) in mass, 4.8-6.4(-8.0) X 2.4-3.6(-4.) μm (n = 48/2, x = 5.7 X 3.1 μm, Q = 1.50-2.83, Qx = 2.11), lacrymoid to ellipsoid in side view and in face view, hyaline, inamyloid; wall smooth, thin.

Commentary: Basidiomata of *G. polyphyllus* can be mistaken for those of *G. confluens* (Halling, 1983) but can be separated from each other by the distinct odor of the former. Basidiomata of *G. impudicus* also have a strong odor and taste that is more foetid and unpleasant, but pilei are more reddish-brown and lamellae are not as crowded. Hyphae of the pileus epicutis in *G. polyphyllus* produce frequent diverticula, characteristically absent in *G. confluens*. Based on the examination of the type specimen of *G. polyphyllus*, the shape and dimensions of the spores and cheilocystidia, and structure of pileus epicutis, conform well to the Costa Rican specimens. This represents the first report of this species from Costa Rica, and a range extension for the species in the Americas.

Material examined: COSTA RICA. Cartago, vic. Estrella, Palo Verde, 9° 46'59" N, 83° 56'71" W, 1700 m, 28.VI.1998, col. RH Petersen & JL Mata, *TFB 9658* (TENN 56619); Puntarenas Prov., Sabalito, Estación Biológica Las Alturas, 8° 56'59" N, 82° 50'02" W, 1420 m, 26.V.1994, col. E Franco, M Mata, J Johnson, L Umaña, *EFM 1191* (NY). UNITED STATES. New York, Essex Co., Minerva, VII.1992, col. CH Peck, (NYS, HT).

(28) *Gymnopus pseudo-omphalodes* (Dennis) Mata, *comb. nov.* Fig. 51

≡ *Collybia pseudo-omphalodes* Dennis. 1961. Kew Bull. 15: 74.

HOLOTYPE: VENEZUELA. Estado Miranda, Guatopo, 25.VI.1958, *Dennis 1108*, (K) *teste* Pegler (1983), *n.v.*

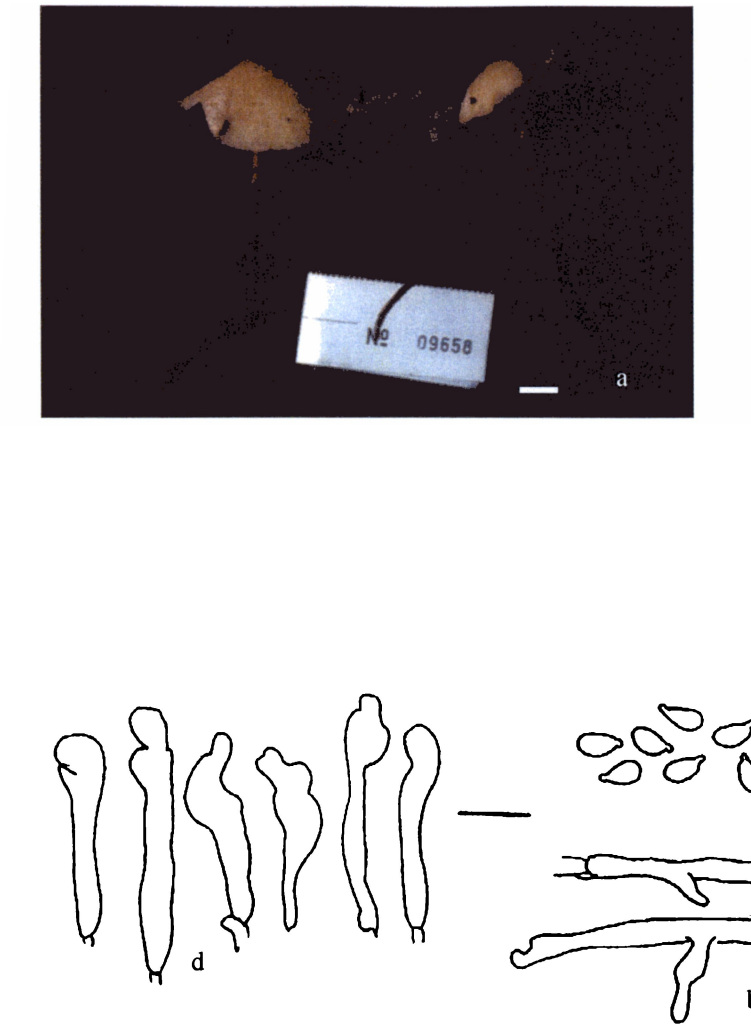


Fig. 50. *Gymnopus polyphyllus*. a. basidiomata (TFB 9658); b. elements of pileus epicutis; c. basidia and basidiole; d. cheilocystidia; e. basidiospores. Scale bar equals 10 μm for line drawings, 1 cm for picture.

Pileus 10-55 mm diam, at first obtusely convex to parabolic, then plano convex to subumbonate; surface fibrillose-streaked, hygrophanous, sometimes lubricous, brown (7E7) or cinnamon brown (6C/D7) overall except disc center when young, with age dark brown (7F8-8F8) at center of disc, outwards eventually orange brown (5C4/5) to white; margin incurved at first, then plane, striate-sulcate; context up to 2 mm broad, white to watery brown (5C4). **Lamellae** adnexed when young, soon pulling away from stipe, up to 3 mm broad, crowded, white to ivory (4B3); margin even to barely subfimbriate, occasionally anastomosed or forked. **Stipe** 30-80 X 3-9 mm, ± equal, but expanded at base, terete but sometimes flattened, sometimes curved at the base, twisted; surface white subpruinose or fibrillose-striate, near apex at first white to ± concolorous with lamellae overall, at base soon darkening cinnamon brown (6/7E7/8); interior becoming hollow . Odor and taste mild. **Habitat** on rotten wood; gregarious (Fig. 51a).

Pileus epicutis a cutis; hyphae 2-8(-30) μm diam, repent, radially oriented, with diverticula, pigment lightly encrusted, yellowish in mass, hyaline singly, inamyloid, with clamp connections; wall thin; terminal cells, cylindrical to inflated, prostrate to semierect, sometimes diverticulate. **Pileus trama** interwoven; hyphae 4-14 μm diam, hyaline, inamyloid, with clamp connections; wall thin. Gloeopleurous hyphae common, up to 4 μm diam. **Lamellar trama** regular; hyphae 4-20(30) μm diam, hyaline, inamyloid, with clamp connections; wall thin. Gloeopleurous hyphae common, up to 4 μm diam. **Basidia** (Fig. 51b) 26-29 X 6-7 μm, clavate; sterigmata four; basidioles 24-29 X 4-7 μm, cylindrical to clavate. **Pleurocystidia** absent. **Cheilocystidia** (Fig. 51c) 32-68 X 6-10 μm, long clavate to broadly clavate, sometimes mucronate, lobed, with apical projections or furcate, with clamp connections. **Stipe epicutis** parallel; hyphae 4-12 μm diam, hyaline singly, inamyloid, with clamp connections; wall up to 0.8 μm thick. **Caulocystidia** abundant, up to 120 X 6 μm, long, cylindrical, septate, forming fascicles or entangled and repent on the surface.

Basidiospores (Fig. 51d) cream (3A4-4A4) in mass, (7.2-)8.0-9.6 X 3.6-4.8 μm (n = 60/3, x = 8.2 X 4.1 μm, Q = 1.82-2.22, Qx = 2.00), broadly ellipsoid or lacrymoid in side view, ellipsoid or obovoid in face view, hyaline, inamyloid; wall smooth, thin; contents multiguttulate.

Commentary: Examined material from Puerto Rico was identical to that from Costa Rica, which macroscopically matches well the original description by Dennis (1961) and basidiomata reported by Pegler. Spore dimensions, however, differ from those reported from the type (not seen) and species description for the Lesser Antilles (Pegler, 1983b), although cheilocystidia are vaguely similar. The pileus epicutis is a strict cutis, with occasional diverticulate hyphae and a new combination in *Gymnopus* for this species epithet is proposed.

Gymnopus pseudo-omphalodes appears phylogenetically related to other lignicolous species such as *G. fibrosipes*, *G. luxurians* and *G. dichrous* (Figs. 2-4).

Material examined: COSTA RICA. Puntarenas Prov., Coto Brus Co., San Vito, OTS Las Cruces Biological Station, Ridge trail, 8° 47'09" N 82° 57'31" W, 1100 m, 12.VIII.1994, col. RE Halling, G Mueller, B Strack, *REH 7348* (NY) **S**; Río Marzo, Finca de Efraín Jiménez, 8° 56'59" N, 82° 50'02" W, 1100 m, 11.IX.1994, col. E Franco-Molano, Ruiz, *EFM 1285* (NY). PUERTO RICO. El Verde, 13.VIII.1988, col. RE Halling & J Lodge, *REH 6013* (NY).

Discussion

A total of eight morphological species placed in *Rhodocollybia* and 18 in *Gymnopus* have been described from specimens collected in the Talamanca Mountains of southern Costa Rica. Five new species are described in *Rhodocollybia*, namely *R. amica, R. dotae*, *R. lignitilis, R. pandipes*, and *R. tablensis* while *R. popayanica* represents a report of range extension north of Colombia (Franco-Molano et al, 2000; Halling, 1989).



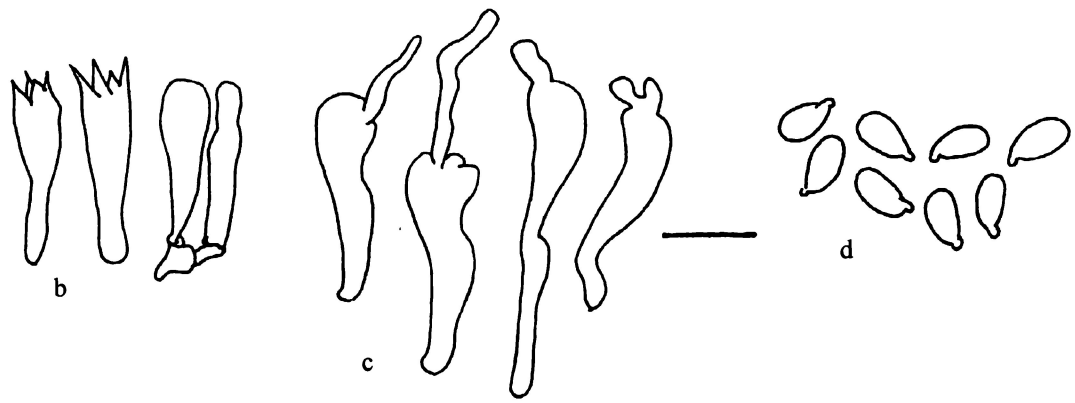


Fig. 51. *Gymnopus pseudo-omphalodes*. a. basidiomata (REH 6013); b. basidia and basidioles; c. cheilocystidia; d. basidiospores. Scale bar equals 10 µm for line drawings, 1 cm for picture.

Within *Gymnopus*, *G. alnicola*, *G. cylindricus*, and *G. pseudolodgeae* are proposed as new to science. New reports to the Talamanca Mountains are *G. biformis*, including two proposed morphological variants, *G. collybioides*, *G. confluens*, *G. dichrous*, *G. fibrosipes*, *G. impudicus*, *G. nubicola*, *G. polyphyllus*, *G. pseudo-omphalodes*, and *G. spongiosus*. At the same time new combinations for *G. lodgeae*, *G. neotropicus* and *G. omphalodes* are proposed based on current morphological circumscription of the genus (Antonín and Noordeloos, 1997; Desjardin et al, 1999; Halling, 1983) and on my examination of type specimens or studies of type specimens by others (Antonín and Noordeloos, 1997; Desjardin, 1989; Halling, 1983). *Gymnopus* seems to be represented by a larger number of species than *Rhodocollybia* in the Talamanca Mountains, a comparison that seems to be consistent in relation to other floristic studies in North America (Halling, 1983; Lennox, 1979), Europe (Antonín and Noordeloos, 1997), Colombia (Franco-Molano et al, 2000), Hawaii (Desjardin et al, 1999), Lesser Antilles (Pegler, 1983b), Mexico (Guzmán et al, 1992; Villaruel-Ordaz et al, 1993) and Venezuela (Dennis, 1970).

Mating studies are an important element in taxonomic studies (Petersen, 1995a, 1995b; Petersen and Hughes, 1999). Prevalence of a tetrapolar mating system in both *Rhodocollybia* and *Gymnopus* has been reported (Petersen, 1995a) and this seems to be the case also for species in the Talamanca Mountains. In this study, *G. omphalodes* and *G. pseudolodgeae* seem to exhibit a bipolar mating system, a phenomenon only previously reported in *G. subnudus* (Murphy, 1997; Murphy and Miller, 1993). In this study, *G. omphalodes* is placed in the same phylogenetic clade with *G. subnudus* and *G. peronatus*, a species with a reportedly tetrapolar mating system (Kniep, 1928). No conclusion can be made for a mating system of Costa Rican *G. biformis* because the number of SBIs used in the self-cross was insufficient but a tetrapolar mating system seems to be present. Readings of self-crosses with other taxa were difficult, either because of suspected amphithallism or the presence of two mating factors in clampless SBI cultures. In this dissertation, new reports of tetrapolar mating systems are provided for *R. pandipes*, *G. collybioides*, *G. fibrosipes*, *G. lodgeae*, and *G. spongiosus.* Previous

reports of tetrapolar mating systems from temperate specimens of *G. dichrous*, *G. dryophilus*, and *G. impudicus* (Petersen, 1995a) are corroborated from those collected in the Talamanca Mountains.

Intercollection mating studies in Costa Rican specimens representing *G. impudicus* and *G. omphalodes*, showed that these two form biological species. Results were not as consistent in *R. pandipes*, *G. dryophilus*, *G. lodgeae* or *G. biformis* var. *parvulus* where a low ratio of clamp connections per mating was observed. Failure to observe clamp connections in matings with SBIs from Costa Rican and United States specimens representing morphospecies of *G. dichrous* and *G. biformis* may indicate presence of sibling species, but more voucher specimens and matings are needed to verify the nature of this phenomenon. Intercollection matings with SBIs from the sole specimen of *G. pseudolodgeae* with those from *G. lodgeae* resulted in 100% incompatibility, and differences in many micromorphological characteristics favored proposal of a new species.

The morphological study, in combination with mating data and phylogenetic analysis, support for the most part the placement of the studied Costa Rican specimens in *Gymnopus* and *Rhodocollybia*. The presence of dextrinoid spores seems to be a taxonomic hiatus at the micromorphological level whether basidiomata should be placed in *Rhodocollybia* or *Gymnopus* since other morphological characteristics such as composition of pileus epicutis and presence of cheilocystidia and caulocystidia are very similar in both genera. While presence of dextrinoid spores in *R. amica* and *R. tablensis* could not be confirmed, the phylogenetic signal places them within *Rhodocollybia* and for this reason the genus circumscription (*sensu* Antonín and Noordeloos, 1997) has been emended in this dissertation. Previous mycologists (Antonín and Noordeloos, 1997; Lennox, 1979) have noted scarcity of dextrinoid spores in *Rhodocollybia*, and *R. amica* and *R. tablensis* could be examples of this situation. Both species are represented by only one or two specimens and more collections are needed to determine with certainty if dextrinoid spores are really absent or not.

While the clade including morphospecies in *Rhodocollybia* is not supported by high bootstrap values, *R. maculata*, the type species of *Rhodocollybia*, appears nested among other *Rhodocollybia* species in all of the phylogenetic analyses, supporting the morphological taxonomic treatment. Sequences representing Costa Rican species of *Rhodocollybia* seem to be very different from each other as shown in the phylograms (Figs. 2,3) and could have been a factor in the outcome of the phylogenetic analyses.

Selected species in *Gymnopus* have been used as outgroups in molecular and phylogenetic studies (Hibbett, 2001; Hibbett and Vilgalys, 1993, Nicholson et al, 1997; Thon and Royse, 1999) or appear to be related to those in *Lentinula* in large-scale phylogenies (Moncalvo et al, 2000, 2002). In this study, ITS members of *Rhodocollybia* appear phylogenetically more related to those of *Gymnopus* than to those of *Lentinula.*

Morphological species placed in *Gymnopus* sect. *Levipedes* are separated from those in sect. *Vestipedes* (sensu Antonín and Noordeloos, 1997) by the presence of a "dryophila type" cutis and are supported as a cohesive clade in the phylogenetic tree. Cladistic and phenetic analysis showed North American species placed in sect. *Levipedes* to be monophyletic when *G. fusipes* was used as an outgroup (Vilgalys, 1986). In the same analysis of that study, *G. spongiosus* appeared phenetically associated with other species (i.e. *G. alkalivirens* and *G. semihirtipes*) whose basidiomatal tissues turned green in alkali (i.e. KOH) but separated from species in the *G. dryophilus* complex. In this study, Costa Rican and North American *G. spongiosus* were phylogenetically closely related with *G. erythropus,* a species whose basidiomata have been reported to stain green in alkali (Antonín and Noordeloos, 1997) but separated from those species represented by basidiomata with no green reaction in their tissues, such as *G. ocior, G. dryophilus* and *G. nubicola. Gymnopus macropus* appears distantly related to *G. dryophilus* in spite of reported morphological similarities (Halling, 1996a).

Gymnopus sect. *Vestipedes* (*sensu* Antonín and Noordeloos, 1997; Antonín et al, 1997), on the contrary, appears divided into several phylogenetic clades. Apparent polyphyly in sect. *Vestipedes* has been proposed by Moncalvo et al (2000, 2002) based on sequence analysis of the nuclear large subunit rDNA region. In Moncalvo et al (2002) placement of *M. foetidus* is near to *G. dryophilus* (placed in sect. *Levipedes*) which is a similar result to the phylogenetic relationships observed with the ITS region in this study (Figs. 2-4). *Gymnopus impudicus*, *G. dysodes,* and *M. foetidus* share the peculiarity of a pungent or disagreeable odor present in their basidiomata and form a small clade separate from other members in sect. *Vestipedes*. While no previous publication is known on a DNA sequence-based phylogenetic placement of *G. fusipes*, the type species of *Gymnopus*, in this study it appears related to the clade representing species placed in sect. *Impudicae*. It appears that there is no distinct morphological characteristic that justifies the phylogenetic relationship between *G. fusipes* and Costa Rican *G. impudicus* (or other species in that section) and more studies are needed to explain this phenomenon. Therefore, *G.fusipes* is placed in its own section as treated by Antonín and Noordeloos (1997). Also, *G. fusipes* appears to be phylogenetically closer to the clade formed by species in sect. *Levipedes* than that composed of species in sect. *Vestipedes*. The results in this dissertation support those found in parsimony analysis using phenetic data which placed *G. fusipes* closer to species in sect. *Levipedes* than to those in sect. *Vestipedes* or in *Rhodocollybia* (Vilgalys, 1986).

The clade representing *Gymnopus* sect. *Vestipedes* includes *G. confluens*, the type species for the section. Within sect. *Vestipedes* several phylogenetic associations merit mention. Lignicolous species such as *G. fibrosipes*, *G. pseudo-omphalodes*, *G. luxurians* and *G. dichrous* appear related, while leaf litter mushrooms of the *G. biformis* morphological complex, *G. collybioides*, *G. cylindricus*, *G. confluens* and *G. neotropicus* appear in a sister clade. More data are needed to justify treatment of each clade as independent taxonomic sections in *Gymnopus* based on these characteristics.

Presence of pleurocystidia in combination with habitat and bitter to acrid taste in basidiomata seem to be shared characteristics of *G. omphalodes*, *G. subnudus* and *G. peronatus*. Therefore, *Gymnopus* sect. *Peronati* has been proposed as a new combination within *Gymnopus* in order to accommodate these species phylogenetically segregated from sect. *Vestipedes*. While the above may be plausible explanations to the observed phylogenetic inferences, other factors and data are needed to fully understand and justify the underlying causes of these relationships.

Basidiomata representing morphological species described originally from North America, South America and Europe are now confirmed to fruit in the Talamanca Mountains of southern Costa Rica. This is the case for *G. biformis*, *G. dichrous*, *G. polyphyllus*, and *G. spongiosus* from the United States (Halling, 1983), *R. popayanica* and *G. nubicola* from Colombia (Halling, 1989), and *G. confluens, G. impudicus* and *R. prolixa* var. *distorta* from Europe and United States whose basidiomata are reported to fruit in mixed deciduous forests including *Quercus* spp (Antonín and Noordeloos, 1997; Halling, 1983). The closure of the gap in the Central American Isthmus during the Pliocene Epoch (Gómez, 1986; Graham, 1995) made possible the southward migration of oak-dominated forests and their fungal associates from North America into the Talamanca Mountains of Costa Rica and northern Colombia (Hooghiemstra and Cleef, 1995; Halling, 2001; Kapelle, 1996; Mueller and Halling, 1995). Simultaneously, Neotropical floristic elements dispersing from South America and the Caribbean found a niche in this newly formed geological unit (Gómez, 1986; Kapelle, 1996). Weber (1959) was the first to document the strong resemblance of high mountain vegetation communities in the Talamanca Mountains to the northern Andes Mountains. Evidence of affinity between eastern North American and Costa Rican-Colombian mycobiota associated with oak-forests has been revised for ectomycorrhizal agarics (Halling, 2001; Mueller and Halling, 1995) and documented for *Gymnopus* and *Rhodocollybia* (Halling 1989, 1996a). Discovery of new species and new reports of range extensions in *Gymnopus* and *Rhodocollybia* proposed in this dissertation reinforce the notion of high

diversity and endemism of agaric fungi in neotropical montane *Quercus* forests (Lodge et al, 1995; Mueller and Halling, 1995).

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PART VI
General conclusions

Modern taxonomy and systematic studies incorporate different resources in order to improve data quality. Morphology is the classic and fundamental tool in mushroom taxonomy and in this study all species have been characterized by this method. Taxonomic placement of examined specimens in any of the three genera using macro- and micromorphological characteristics was consistent with current concepts. Correct application of species epithets to specimens, however, depends on the study of the type specimen or authentic material to which a particular name is tied.

Mating studies are particularly helpful when cryptic or sibling species are suspected or when wide distribution patterns are detected. In this disertation, tetrapolar mating systems seem to be a common denominator in *Lentinula*, *Rhodocollybia* and *Gymnopus*. Bipolar matings systems in *Gymnopus* require more attention to determine the nature and significance in biological terms. Mating studies were the cornerstone in detecting an intersterility group in *L. boryana*, leading to the proposal of a new combination. Intercollection pairings in *R. pandipes*, *G. dryophilus*, *G. impudicus* and *G. omphalodes* along the Talamanca Mountains were useful in characterizing biological species and matching those with morphological species concepts. Conversely, intersterility among intercollection pairings raised the question whether morphologically identical specimens belonged to the same biological species as in the case of *G. dichrous* and *G. biformis*.

Sequence analysis of ITS nrDNA is a powerful approach in systematics. Phylogenetic analysis proved valuable in this dissertation to support morphological placement of taxa in *Lentinula*, *Rhodocollybia* and *Gymnopus* within discrete phylogenetic clades. Costa Rican specimens seem to be phylogenetically closely related to those from Europe and North America and to the type species of each genus.

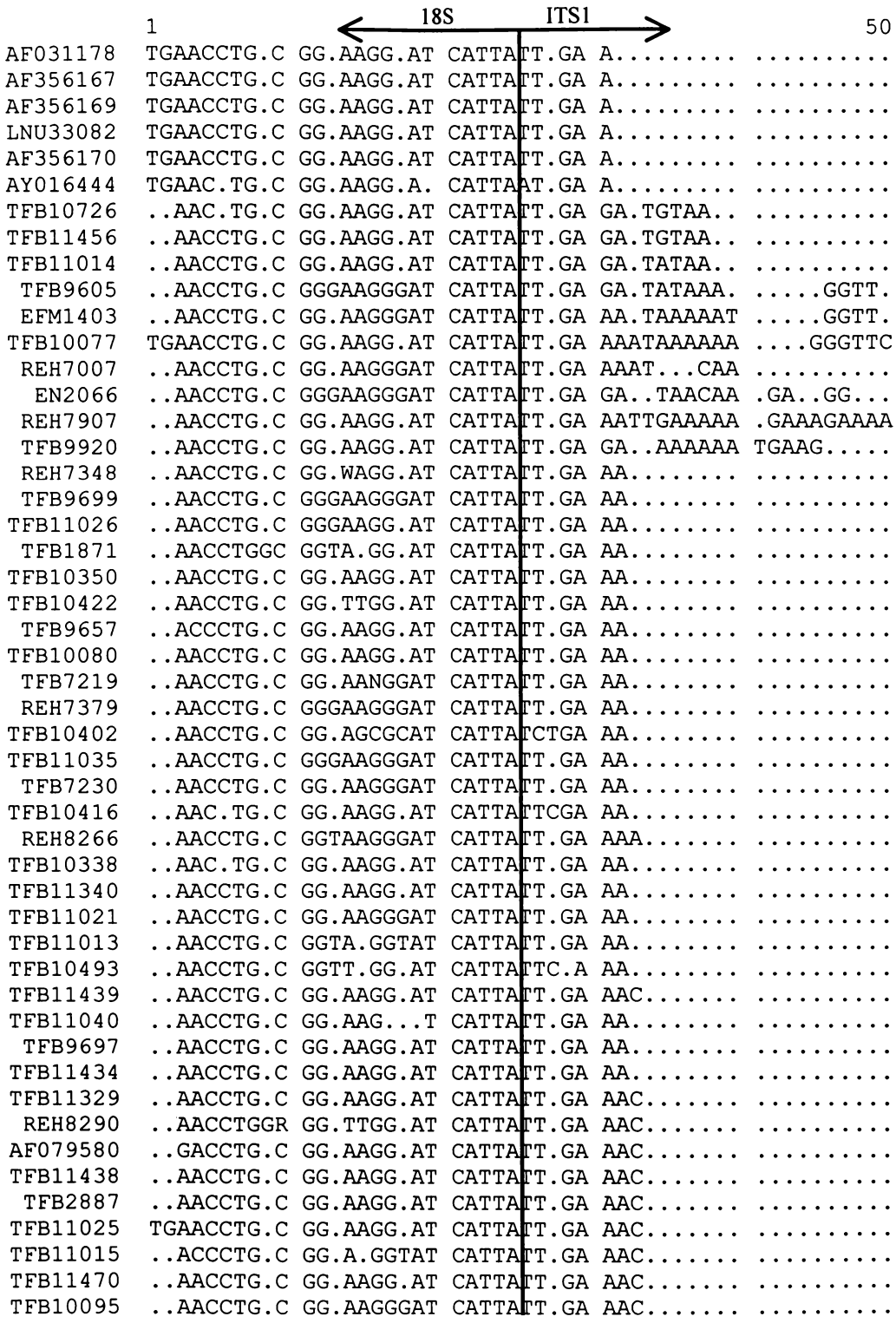
With the input from this dissertation the known number of morphological species in *Lentinula* (3), *Rhodocollybia* (8) and *Gymnopus* (18) in the Talamanca Mountains of southern Costa Rica has increased significantly. The Talamanca Mountains constitute a

distinct physiogeographic unit within Costa Rica whose *Quercus*-dominated forests share floristic elements with northern and southern regions and new reports of species in *Gymnopus* and *Rhodocollybia* described from temperate and Andean regions confirm its unique biogeographic situation.

Appendix

Appendix A

Sequence alignment of the nrDNA ITS region for *Lentinula*, *Rhodocollybia*, and *Gymnopus*. Small subunit (18S), ITS1, 5.8S, ITS2 and large subunit (LSU) are identified by lines. The small subunit (18S), ITS1, 5.8S, ITS2, and large subunit (LSU) regions are identified by lines.


18S
ITS1
1
50
AF031178 TGAACCTG.C GG.AAGG.AT CATTATT.GA A..... .....
AF356167 TGAACCTG.C GG.AAGG.AT CATTATT.GA A..... .....
AF356169 TGAACCTG.C GG.AAGG.AT CATTATT.GA A..... .....
LNU33082 TGAACCTG.C GG.AAGG.AT CATTATT.GA A..... .....
AF356170 TGAACCTG.C GG.AAGG.AT CATTATT.GA A..... .....
AY016444 TGAAC.TG.C GG.AAGG.A. CATTAAT.GA A..... .....
TFB10726 ..AAC.TG.C GG.AAGG.AT CATTATT.GA GA.TGTAA.. .....
TFB11456 ..AACCTG.C GG.AAGG.AT CATTATT.GA GA.TGTAA.. .....
TFB11014 ..AACCTG.C GG.AAGG.AT CATTATT.GA GA.TATAA.. .....
TFB9605 ..AACCTG.C GGGAAGGGAT CATTATT.GA GA.TATAAA. .....GGTT.
EFM1403 ..AACCTG.C GG.AAGGGAT CATTATT.GA AA.TAAAAAT .....GGTT.
TFB10077 TGAACCTG.C GG.AAGG.AT CATTATT.GA AAATAAAAAA ....GGGTTC
REH7007 ..AACCTG.C GG.AAGGGAT CATTATT.GA AAAT...CAA .....
EN2066 ..AACCTG.C GGGAAGGGAT CATTATT.GA GA..TAACAA .GA..GG...
REH7907 ..AACCTG.C GG.AAGG.AT CATTATT.GA AATTGAAAAA .GAAAGAAAA
TFB9920 ..AACCTG.C GG.AAGG.AT CATTATT.GA GA..AAAAAA TGAAG.....
REH7348 ..AACCTG.C GG.WAGG.AT CATTATT.GA AA..... .....
TFB9699 ..AACCTG.C GGGAAGGGAT CATTATT.GA AA..... .....
TFB11026 ..AACCTG.C GGGAAGG.AT CATTATT.GA AA..... .....
TFB1871 ..AACCTGGC GGTA.GG.AT CATTATT.GA AA..... .....
TFB10350 ..AACCTG.C GG.AAGG.AT CATTATT.GA AA..... .....
TFB10422 ..AACCTG.C GG.TTGG.AT CATTATT.GA AA..... .....
TFB9657 ..ACCCTG.C GG.AAGG.AT CATTATT.GA AA..... .....
TFB10080 ..AACCTG.C GG.AAGG.AT CATTATT.GA AA..... .....
TFB7219 ..AACCTG.C GG.AANGGAT CATTATT.GA AA..... .....
REH7379 ..AACCTG.C GGGAAGGGAT CATTATT.GA AA..... .....
TFB10402 ..AACCTG.C GG.AGCGCAT CATTATCTGA AA..... .....
TFB11035 ..AACCTG.C GGGAAGGGAT CATTATT.GA AA..... .....
TFB7230 ..AACCTG.C GG.AAGGGAT CATTATT.GA AA..... .....
TFB10416 ..AAC.TG.C GG.AAGG.AT CATTATTCGA AA..... .....
REH8266 ..AACCTG.C GGTAAGGGAT CATTATT.GA AAA..... .....
TFB10338 ..AAC.TG.C GG.AAGG.AT CATTATT.GA AA..... .....
TFB11340 ..AACCTG.C GG.AAGG.AT CATTATT.GA AA..... .....
TFB11021 ..AACCTG.C GG.AAGGGAT CATTATT.GA AA..... .....
TFB11013 ..AACCTG.C GGTA.GGTAT CATTATT.GA AA..... .....
TFB10493 ..AACCTG.C GGTT.GG.AT CATTATTC.A AA..... .....
TFB11439 ..AACCTG.C GG.AAGG.AT CATTATT.GA AAC..... .....
TFB11040 ..AACCTG.C GG.AAG...T CATTATT.GA AA..... .....
TFB9697 ..AACCTG.C GG.AAGG.AT CATTATT.GA AA..... .....
TFB11434 ..AACCTG.C GG.AAGG.AT CATTATT.GA AA..... .....
TFB11329 ..AACCTG.C GG.AAGG.AT CATTATT.GA AAC..... .....
REH8290 ..AACCTGGR GG.TTGG.AT CATTATT.GA AAC..... .....
AF079580 ..GACCTG.C GG.AAGG.AT CATTATT.GA AAC..... .....
TFB11438 ..AACCTG.C GG.AAGG.AT CATTATT.GA AAC..... .....
TFB2887 ..AACCTG.C GG.AAGG.AT CATTATT.GA AAC..... .....
TFB11025 TGAACCTG.C GG.AAGG.AT CATTATT.GA AAC..... .....
TFB11015 ..ACCCTG.C GG.A.GGTAT CATTATT.GA AAC..... .....
TFB11470 ..AACCTG.C GG.AAGG.AT CATTATT.GA AAC..... .....
TFB10095 ..AACCTG.C GG.AAGGGAT CATTATT.GA AAC..... .....

```
          51                                                        100
AF031178  .......... ...CTC.TTT GG.TGGGT.G GAC....... ........TG
AF356167  .......... ...CTC.TTT GG.TGGGT.G GAC....... ........TG
AF356169  .......... ....TTTTTT GG.TGG.T.G GAT....... ........TG
LNU33082  .......... ....TTTTTT GG.TGG.T.G GAT....... ........TG
AF356170  .......... ....TTTTTT GG.TGG.T.G GAT....... ........TG
AY016444  .......... .TGC....TT G..TAG.A.G G.T....... ........TG
TFB10726  ...CGGCGCC CTG....TTA G..TTGTT.G .......... ...CAA..TG
TFB11456  ...CGGCGCC CTG....TTA G..TTGTT.G .......... ...CAA..TG
TFB11014  ...CGGCGCC .TGCTGGTTA G..TTCTT.. .......... ..ACAA..TG
 TFB9605  ATT....... .......GGA G...TC.... .......... AAACAAA.TG
 EFM1403  A......... ......TTGA GG..TGT... .......... TTA.....TG
TFB10077  ATA....... .......TGA G..TAGT... .......... ACACAT..TG
 REH7007  .......... ........AA GG........ .......... ...C....TG
  EN2066  .......... ......TTTA ...TTG.... .AGGTGT.CG AATCATA.TG
 REH7907  GA........ ......GGGA GG.TTG.ATG AAA..GT..G TTTTA...TG
 TFB9920  .......... ...TCATT.. .....GTAA. ....TGCTTG .TTCA..TTG
 REH7348  .......... ..GCTTT.GG GG........ .......... ATAC....TG
 TFB9699  .......... ..GCTTT.GG GG........ .......... ATAC....TG
TFB11026  .......... ..GTTTT.GG GGG....... .......... .TAC....TG
 TFB1871  .......... ..TCTCTT.. GG........ .......... ATAT....TG
TFB10350  .......... ..GTTT..GG GG........ .......... .TAC....TG
TFB10422  .......... ..GCTTT..G GG..AGG... .......... .TAC....TG
 TFB9657  .......... ..GCTTT..G GG..GGA... .......... .TAC....TG
TFB10080  .......... ..GCTTT..G GG..AGA... .......... .TAC....TG
 TFB7219  .......... ..GCTTTT.G GG..AGG... .......... .TAC....TG
 REH7379  .......... ..GCTTT..G GG..AGA... .......... .TAC....TG
TFB10402  .......... ..GCTTTT.G GG..GAA... .......... .TAC....TG
TFB11035  .......... ..GCTTTT.G GG..AGA... .......... .TAC....TG
 TFB7230  .......... ..GCTTTT.G GG..GAA... .......... .TAC....TG
TFB10416  .......... ..GCTTTT.G GG..AGA... .......... .TAT....TG
 REH8266  .......... ..GCTTTT.G GG..AAA... .......... .TAC....TG
TFB10338  .......... ..GGTTTTGG GG...AAT.. .......... .TAC....TG
TFB11340  .......... ..GGTTTTGG GG...AAT.. .......... .TAC....TG
TFB11021  .......... .AGTTTT.GG GG...AAT.. .......... .TAC....TG
TFB11013  .......... ..GCTTTTAG GGG..AAT.. .......... .TGC....TG
TFB10493  .......... ..GTTTTTGG GGG..AAT.. .......... .TAC....TG
TFB11439  .......... .TTCTTT... GG..AGATAA G......... .TAC....TG
TFB11040  .......... ...CTTT... GG..AAATGG .......... .TGC....TG
 TFB9697  .......... ...CTTT... GG..AAATAG .......... .TAC....TG
TFB11434  .......... CTTCTTT... GG..AAGTAG .......... .TAC....TG
TFB11329  .......... .TTCTTT... GG..AAATAG .......... .TAC....TG
 REH8290  .......... .TTCTTT... GG..AAATAN .......... .TAC....TG
AF079580  .......... .TTCTTT... GG..AAATAG .......... .TAC....TG
TFB11438  .......... .TTCTTT... GG..AAATAG .......... .TAC....TG
 TFB2887  .......... .TTCTTT... GG..AGATAG .......... .TAC....TG
TFB11025  .......... .TTCTTT... GG..AGATAG .......... .TAC....TG
TFB11015  .......... .TTCTTT... GG..AAATAG .......... .TAC....TG
TFB11470  .......... .TTCTTT... GG..AGATAG .......... .TAC....TG
TFB10095  .......... .TTCTTTT.. GG..AAATAG .......... .TAC....TG
```

```
          101                                                        150
AF031178  TT..... ...GCTGGCT TG...TCTTT CT.AAAAAAG GTTTGCA.TG
AF356167  TT..... ...GCTGGCT TG...TCTTT CT.AAAAAAG GTTTGCA.TG
AF356169  TT..... ...GCTGGCC .......TTT ........GG G....TA.TG
LNU33082  TT..... ...GCTGGCC .......TAT ........GG G....TA.TG
AF356170  TT..... ...GCTGGCC .......TTT ........GG G....TA.TG
AY016444  AT..... ...GCTGGCC C....TCTTA AC...AAAAG G...GCA.TG
TFB10726  T...AGAGTC AGTGCTGGC. ........TT CT.....GAA G....TAT.G
TFB11456  T...AGAGTC AGTGCTGGC. ........TT CT.....GAA G....TAT.G
TFB11014  C...AGAGTC AGTGCTGAC. TGT....... .....AAAAA G....TAT.G
 TFB9605  TTTTT..... ..TGCTGGTC TTT....... CT..ATGGAA G...ACAT.G
 EFM1403  T..... ..TGCTGGCC C........T C.AAAT.AGG G....CAT.G
TFB10077  T..... ..TGCTGGC. .......... CTT.....AG G....CAT.G
 REH7007  TT..... ...GC..AC. .ATAA.TTTA C..... .....TAT.G
  EN2066  T..... ..TGCTGGT. .......... CTT....AGG G....CATTG
 REH7907  TC..... ..TGC..AT. .GTTG..AAA CAAT...AAG G....CAT.G
 TFB9920  T..... ..TGCTGGCC T.......TT CAAATGAAGG G....CAT.G
 REH7348  T..... ..TGCTGGTC T.......TT C.TC...AAG G....CAT.G
 TFB9699  T..... ..TGCTGGTC T.......TT C.TC...AAG G....CAT.G
TFB11026  T..... ..TGCTGGTC TTTT..... ..AC...AAG G....CAT.G
 TFB1871  T..... .CTGCTGGTC TTTT...CTT TTAGTT.AAG G....CATTG
TFB10350  T..... ..TGCTGGTC T......CTT ..AC...AAG G....CAT.G
TFB10422  T..... ..TGCTGG.C TT......CT A.AC...GAG G....CAT.G
 TFB9657  T..... ..TGCTGGTC TT......TT A.ACA..AAG G....CAT.G
TFB10080  T..... ..TGCTGG.C TT......CT A.AA...GAG G....CAT.G
 TFB7219  T..... ..TGCTGG.C TT......TT A.AC...AAG G....CAT.G
 REH7379  T..... ..TGCTGG.C TTT....TTT ..AC...AAG G....CAT.G
TFB10402  T..... ..KGCTGGCC TT......TT A.ATG..AAG G....CAT.G
TFB11035  T..... ..TGCTGG.C TTT.....TA A.AC...AAG G....CAT.G
 TFB7230  T..... ..TGCTGG.C TT.....CTT A.ACG..GAG G....CAT.G
TFB10416  T..... ..TGCTGG.C TTT.....TT A.AC...AAG G....CAT.G
 REH8266  T..... ..TGCTGG.C TTT.....TA A.AC...AAG G....CAT.G
TFB10338  T..... ..TGCTGGCC TTT....TCT AAT.G.GAAG GG...TAT.G
TFB11340  T..... ..TGCTGGCC TTT....TCT AATAG.GAAG GG...TAT.G
TFB11021  T..... ..TGCTGGCC TTT.....GT A.ACA.GAAG G....CAT.G
TFB11013  T..... ..TGCTGGTC TTTC..... ......TAAG G....CAT.G
TFB10493  T..... ..TGCTGGTC TCT....TTT ......TGA. .....CAT.G
TFB11439  T..... ..TGCTGGCC TT......GT A.ATT..GAG G....CAT.G
TFB11040  T..... .TTGCTGACC TT......GT T.AT...GAG G....TAT.G
 TFB9697  T..... ..TGCTGGCC TT......GT A.TA...AAG G....TAT.G
TFB11434  T..... ..TGCTGGCC TT......GT A.AT...GAG G....TAT.G
TFB11329  T..... ..TGCTGGCC TT......GT A.AT...GAG G....TAT.G
 REH8290  T..... ..TGCTGGCC TT......GT A.AT...GAG G....TAT.G
AF079580  T..... ..TGCTGGCC TT......GT A.AT...GAG G....TAT.G
TFB11438  T..... ..TGCTGGCC TT......GT A.AT...GAG G....TAT.G
 TFB2887  T..... ..TGCTGGCC TT......GT A.AT...GAG G....TAT.G
TFB11025  T..... ..TGCTGGCC TT......GT A.AT...GAG G....TAT.G
TFB11015  T..... ..TGCTGGCC TT......GT A.AT...GAG G....TAT.G
TFB11470  T..... ..TGCTGGCC TT......GT A.AT...GAG G....TAT.G
TFB10095  T..... ..TGCTGGCC TT......GT A.AT...GAG G....TAT.G
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          151                                                       200
AF031178  T.GCAC.GT. .......... TCTCCACT.. .AATTT.... ..........
AF356167  T.GCAC.GT. .......... TCTCCACT.. .AATTT.... ..........
AF356169  T.GCAC.AT. .......... CCTCCTCT.. .......... GATT......
LNU33082  T.GCAC.AT. .......... CCTCCTCT.. .......... GATT......
AF356170  T.GCAC.AT. .......... CCTCCTCC.. .......... GATTT.....
AY016444  T.GCTC.GC. .......... .CTCT..... .......... TATA......
TFB10726  T.GCAC.GCT TTACTTG.TA GCTCT..... .AACTTGG.T GATAA.....
TFB11456  T.GCAC.GCT TTACTTG.TA GCTCT..... .AACTTGG.T GATAA.....
TFB11014  T.GCAC.ACT TTGCTTGTTA GCTCT..... .GACTTGGGT GATAA.....
 TFB9605  T.GCAC...T TT..TT.TT. GC........ .CACT....T GA........
 EFM1403  T.GCACTG.T T......... .......... .CACT..... GATTTAG...
TFB10077  T.GCAC.GGT TTAC...... .......... .TACCT...T GAAT......
 REH7007  TCGCAC.GCT T......... .......... .TAT.....T GTTT......
  EN2066  T.GCACTAGT T......... .CTT...... .CACTAAAA. GAAA......
 REH7907  T.GCACTGTT ..GAT..... GCTC....AT CGACT..... GAACTTTTTT
 TFB9920  T.GCAC.GAA TTTT...... GCT......A CTACTTT... GA.CTAATTT
 REH7348  T.GCAC.GTA T......... .CTC.....T AATC...... ..........
 TFB9699  T.GCAC.GTA T......... .CTC.....T AATC...... ..........
TFB11026  T.GCAC.GTA T......... .CTC.....T AATC...... ..........
 TFB1871  T.GCAC.GTA T......... .CTG....AC TGTC...... ..........
TFB10350  T.GCAC.GTA .......... .CTT....CT AATC...... ..........
TFB10422  T.GCAC.GTA T......... .CTTTT.... AATC...... ..........
 TFB9657  T.GCAC.GTA T......... .CTCTT.... AATC...... ..........
TFB10080  T.GCAC.GTA T......... .CTCTT.... AATC...... ..........
 TFB7219  T.GCAC.GTA TTT....... .CTT...... AATTT..... ..........
 REH7379  T.GCAC.GTA TTT....... .CTT.....A AATT...... ..........
TFB10402  T.GCAC.GTA TT........ .CTTT...AA AAT....... ..........
TFB11035  T.GCAC.GTA TTT....... .CTT.....A AATC...... ..........
 TFB7230  T.GCAC.GTA TTTT...... .CTT.....A AATC...... ..........
TFB10416  T.GCAC.GTA TTT....... .CTT.....A GAT....... ..........
 REH8266  T.GCAC.GTA TTT....... ..TTT....A AATC...... ..........
TFB10338  T.GCAC.GTA TT........ .CTC.....T AATC...... ..........
TFB11340  T.GCAC.GTA TT........ .CTC.....T AATC...... ..........
TFB11021  T.GCAC.GTA TT........ .CTC.....T AATC...... ..........
TFB11013  T.GCAC.GTA GTT....... .CTC....TT GATTT..... ..........
TFB10493  T.GCAC.GTA GTT....... .CTC....TT AATCT..... ..........
TFB11439  T.GCAC.GTC TTATTTC... .........T AATC...... ..........
TFB11040  T.GCAC.GT. .......... ACCATTTCT. GATC...... ..........
 TFB9697  T.GCAC.GT. .......... ACTATTTCT. AATC...... ..........
TFB11434  T.GCAC.GT. .......... ACTATTTCT. AATC...... ..........
TFB11329  T.GCAC.GTC TATT...... ACC....... AATT...... ..........
 REH8290  T.GCAC.GTC TATT...... ACC....... AATT...... ..........
AF079580  T.GCAC.GTC TATT...... ACC....... AATT...... ..........
TFB11438  T.GCAC.GTC TATT...... ACC....... AATT...... ..........
 TFB2887  T.GCAC.GTC TAG....... .CTCT..... AATC...... ..........
TFB11025  T.GCAC.GTC TAA....... .CTCT..... AATC...... ..........
TFB11015  T.GCAC.GTC TATT...... ACC....... AATC...... ..........
TFB11470  T.GCAC.GTC TAT....... .CTCT..... AATC...... ..........
TFB10095  T.GCAC.GTC TATT...... ACC....... AATT...... ..........
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          201                                                       250
AF031178  .......... ....C.A.TT TAT...CCAC CTGTGCACCT TTT..GTAGG
AF356167  .......... ....C.A.TT TAT...CCAC CTGTGCACCT TTT..GTAGG
AF356169  .......... ....CTA.TT CAT...CCAC CTGTGCACTT TTT..GTAGG
LNU33082  .......... ....CTA.TA CAT...CCAC CTGTGCACTT TTT..GTAGG
AF356170  .......... ....CTA.TT CAT...CCAC CTGTGCACTT TTT..GTAGG
AY016444  .......... ....TT..TT CAT...CCAC CTGTGCACTT TTT..GTAGG
TFB10726  CCG....... TTTCTTA.TC CAT...CCAC CTGTGCACTT T.TT.GTAGG
TFB11456  CCG....... TTTCTTA.TC CAT...CCAC CTGTGCACTT T.TT.GTAGG
TFB11014  CCG....... TTTCTTA.TC TAT...CCAC CTGTGCACTT T.TT.GTAGG
 TFB9605  CCG....... ......A.TT TATTATCCAC CTGTGCACTT ..TTTGGAGA
 EFM1403  CCAA...... ..TTTTG..T TAT...CCAC CTGTGCACGT .ATT.GTAGG
TFB10077  CCGG...... ..TTTTA.TA TAT...CCAC YTGTGCACTT T.TT.GTAGA
 REH7007  .......... .........T TAT...CCAC CTGTGCACTT TATT.GTAGA
  EN2066  CCGA.TT... .TTATTA.TC CAT...CCAC CTGTGCACTT TGTT.GTAGA
 REH7907  TCTTTCTTTC TTCGCTT.TT CAT...CCAC CTGTGCACTT TTTTTGT...
 TFB9920  .......... ......A.TC CAT...TCAC CTGTGCACTT TTTT.GTAGA
 REH7348  .......... .....TA.TT CAT...CCAC CTGTGCATCT T.TT.GTAGG
 TFB9699  .......... .....TA.TT CAT...CCAC CTGTGCATCT T.TT.GTAGG
TFB11026  .......... .....TA.TT CAT...CCAC CTGTGCATTT T.TT.GTAGA
 TFB1871  .......... .....T..TT CAT...CCAC CTGTGCA.CT TTTT.GTAGA
TFB10350  .......... .....TA.TT CAT...CCAC CTGTGCATCT T.TT.GTAGA
TFB10422  .......... .....CA.TT CAT...CCAC CTGTGCACCT T.TT.GTAGA
 TFB9657  .......... .....CA.TT CAT...CCAC CTGTGCACCT T.TT.GTAGA
TFB10080  .......... .....CA.TT CAT...CCAC CTGTGCACCT T.TT.GTAGA
 TFB7219  .......... .....CA.TT CAT...CCAC CTGTGCACCT T.TT.GTAGA
 REH7379  .......... .....CA.TT CAT...CCAC CTGTGCATCT T.TT.GTAGA
TFB10402  .......... .....CA.TT CAT...CCAC CTGTGCACCT T.TT.GTAGA
TFB11035  .......... .....TA.TT CAT...CCAC CTGTGCATCT T.TT.GTAGA
 TFB7230  .......... .....TA.TT CAT...CCAC CTGTGCACCT T.TT.GTAGA
TFB10416  .......... .....CA.TT CAT...CCAC CTGTGCATCT T.TT.GTAGA
 REH8266  .......... .....TA.TT CAT...CCAC CTGTGCATCT T.TT.GTAGA
TFB10338  .......... .....TA.TT CAT...CCAC CTGTGCACTT T.TT.GTAGA
TFB11340  .......... .....TA.TT CAT...CCAC CTGTGCACTT T.TT.GTAGG
TFB11021  .......... .....TAGTT CAT...CCAC CTGTGCACTT T.TT.GTAGG
TFB11013  .......... .....TATTT CAT...CCAC CTGTGCACTT T.TT.GTAGG
TFB10493  .......... .....TATTT CAT...CCAC CTGTGCACTT .CTT.GTAGG
TFB11439  .......... .....CA.TT CAT...CCAC CTGTGCACTT T.TT.GTAGG
TFB11040  .......... .....TA.TT CAT...CCAC CTGTGCATTT .ATT.GTAGG
 TFB9697  .......... .....TG.TT CAT...CCAC CTGTGCATTT .ATT.GTAGG
TFB11434  .......... .....TA.TT CAT...CCAC CTGTGCACTT .ATT.GTAGG
TFB11329  .......... .....TA.TT CAT...CCAC CTGTGCACTT T.TT.GTAGG
 REH8290  .......... .....TA.TT CAT...CCAC CTGTGCACTT T.TT.GTAGG
AF079580  .......... .....TA.TT CAT...CCAC CTGTGCACTT T.TT.GTAGG
TFB11438  .......... .....TA.TT CAT...CCAC CTGTGCACTT T.TT.GTAGG
 TFB2887  .......... .....TA.TT CAT...CCAC CTGTGCACTT .ATT.GTAGG
TFB11025  .......... .....TA.TT CAT...CCAC CTGTGCACTT .ATT.GTAGG
TFB11015  .......... .....TA.TT CAT...CCAC CTGTGCACTT T.TT.GTAGG
TFB11470  .......... .....TA.TT CAT...CCAC CTGTGCACTT .ATT.GTAGG
TFB10095  .......... .....TA.TC CAT...CCAC CTGTGCATTT T.TT.GTAGG
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          251                                                        300
AF031178  ..AGG.GACC TTTACT..CA ...TTCTATT G.TGATAA.. .....CATTA
AF356167  ..AGG.GACC TTTACT..CA ...TTCTATT G.TGATAA.. .....CATTA
AF356169  ..AGTT...C TTTCAT..CA GG.TTTTGAA C.AGGT.... ..GCTCATTA
LNU33082  ..AGTT...C TTTCAT..CG GG.TATTTAA C.AGGT.... ..GCTCATTA
AF356170  ..AGTT...C TTTCAT..CG GG.TTTTTGA ..AGGT.... ..GCTCATTA
AY016444  ..AGTT...C TTTCTTTTCA .......TAT C.AGTT.... ..GA.CATTC
TFB10726  ..AGTT.TTT ATTA...... GG.TT..... ...GGCTTT. .....CAAG.
TFB11456  ..AGTT.TTT ATTA...... GG.TT..... ...GGCTTT. .....CAAG.
TFB11014  ..AGTT.TTT GTTA.....C GG.TT..... ...GGCTCT. .....CAGG.
 TFB9605  ..TG....CC TT.CTT.... GG.T...... ..GGGC.... ...AAC.TTG
 EFM1403  ..AG.....C TTTT...... .G.TCATTTT TT........ .....C.TTG
TFB10077  ..AGTT...C TT......GC TG.TTTTT.. ....GC.... .....CATG.
 REH7007  ..AGCGTT.T GTTA...... .GATTTTAA. .......... ....CCAA..
  EN2066  ..GGTT.... TTTAA....T AG.TTGACTT TT.GGCTAT. ...TGCAAG.
 REH7907  ..TGTT.... .......... .......... .......... .....GAGA.
 TFB9920  ..AGTT..TC TT........ .GTTTAATTT T......... .....CAAG.
 REH7348  ..AGTT...C TTT.....CA AG.TTGGCTT .......... ....GCA.GG
 TFB9699  ..AGTT...C TTT.....CA AG.TTGGCTT .......... ....GCA.GG
TFB11026  ..AGTT...C TTTT....CA AG.TTGGCTT T...GTTT.. ....GCAAGG
 TFB1871  ..AGTT...C TTTT...... AGATTGGCTT .......... ......ATGG
TFB10350  ..AGTT...C TTT.....CA GG.TTGGCTT .......... ....GCAAG.
TFB10422  A.TGT..CTC GATTA...CA GA.TCGG... ..TTTT.... ....GCAAGT
 TFB9657  A.TGT....C TCTAGTA.CA GA.TTGG... .CTTT..... ....ACGACT
TFB10080  A.TGT....C TCGACTAT.A GA.TCAAA.. ....GTTT.. ..ATG.AA.T
 TFB7219  A.TGT..CTC GTMTT...CA AA.TCGGCT. ..TTT..... ...TGCAAGT
 REH7379  G.TGTT..TC GTTT....CA AA.TCGGC.. TCTT.....T TTCTGCGAGT
TFB10402  A.TGT....C TTGA.TT.CA GA.TTGG... TCC....... ....GCAAGT
TFB11035  A.TGT..CTC CTTT....CA AA.TCGGATT TCTTT..... ..CTGCAGGT
 TFB7230  A.TGT..CTC GTTTT...CA GA.TTGG... ..TTTT.... ...TGCAAGT
TFB10416  A.TGT..CTC TTTT....CA AG.TCGGGTT TCTTTT.... ....GCGAGT
 REH8266  A.TGT....C TTTGTTTCAG AG.TCGGATT TCTTTT.... ....GTAGGG
TFB10338  ..AGTA.CTC TT.......G AG.TTGATT. .......... .GGTAGAAGG
TFB11340  ..AGTA...C TT.GTT...G AG.TTGACT. .......... .GGTAAAAGG
TFB11021  ...GATT..C TTTT.....A GG.TTGACT. .......... .GGTAGA...
TFB11013  .GGG.CTATC TTTT....GA GA.TTGACAT T......... ..........
TFB10493  ..AGTCTGTT GCTTTT..AG GA.TTGACAT T......... ..........
TFB11439  ..AGT...TC TTT.....GA AG........ .........T TGGCTTTAA.
TFB11040  A.GGT....C TTT.....GA AG........ .......... .GGTT.....
 TFB9697  A.GGT....C TTT.....GA AG........ .......... .GGTT.....
TFB11434  G..GTTTTTT TTT.....GA GG.TTGACTT .......... ..GTTGTAGG
TFB11329  ..AGT...TC TTT.....GA AG.TTATTTT .......... ..........
 REH8290  ..AGT...TC TTT.....GA AG.TTATTTT .......... ..........
AF079580  ..AGT...TC TTT.....GA AG.TTATTTT .......... ..........
TFB11438  ..AGT...TC TTT.....GA AG.TTATTTT .......... ..........
 TFB2887  ..AGTTTT.. TTTT...AAA GG.CAACTTT .......... ..........
TFB11025  ..AGT...TT TTTTTAAAAA GG.CAACTTT .......... ..........
TFB11015  ..AGT...TC ATTT....GA GG.CTATTTT .......... ..........
TFB11470  ..AGT...TC TTT.....GA GG.CTATTTT .......... ..........
TFB10095  ..AGT...TC TTT.....GA GG.CTATTTT .......... ..........
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          301                                                        350
AF031178  TGTTGT.... ..TTTCAATA TT.....GTG AGGC...... ..AAAT...G
AF356167  TGTTGT.... ..TTTCAATA TT.....GTG AGGC...... ..AAAT....
AF356169  CGA.GTT... .....AACTT .GG....GAA GGACTAGTT. ........GA
LNU33082  CGA.GTT... .....AACTT .GG....GAA AGACTAGTT. ........GA
AF356170  TGA.GTT... .....A.CTT .GA.....AA AGACTAGTT. ........GA
AY016444  ATT.GTG... ....GAG... TGT......T CAACTGGGTT T......GAA
TFB10726  .TC......G AAA.GACTTT .GTT..AG.T TGGCAT.... ........AA
TFB11456  .TC......G AAA.GACTTT .GTT..AG.T TGGCAT.... ........AA
TFB11014  .TC......G AAA.GACTT. .GTT..AG.T TAGCAT.... ........AA
 TFB9605  GAC.....TT AT.GGTGTGA .GTT...GAT CTGAC..... ..........
 EFM1403  G.CT...TTT A..GGAG.TT .GT...AGAA AAGA...... .........T
TFB10077  TTG.....TA A..TGACTCG .GTA...... AAGTGTT... .........G
 REH7007  .TC.....TG A..TAA.... ..TA...... .......... ..........
  EN2066  TTGT.....G AAAGG.CTT. .GT...GGTA GGGCAA.GTT ...AATTTTT
 REH7907  .......... .......... .......... .......... ..........
 TFB9920  .TCC.....G TTAAGA..TT TGG....AAA AG..ACAAC. ..........
 REH7348  .TCCTTTT.. ..GGG.CATT .GTA.....C AGTTAATTTT .....GA...
 TFB9699  .TCCTTTT.. ..GGG.CATT .GTA.....C AGTTAATTTT .....GA...
TFB11026  .TCCTTT... ...GGACATT .GTGTA.TAT TGGCTAGCAT .....GA..T
 TFB1871  G...TTT... ...CGACCTT .GTA...... .GTTGATTT. .....GA...
TFB10350  .TCCTTT... ...GGACTTT .GTATT.... GGCTAATA.. ....TGT...
TFB10422  GTCCCT.... .AGGGACTTT .GTA.....A ATAATGGTTT ....GGAA..
 TFB9657  GTCTTC.... ACG.GACTTT TGT.....AG AAAT.GGTTT ....GGAA..
TFB10080  GTCTGT.... ACAGG.CTTT .GTA.....G ATTGCGGTCT A....GAA..
 TFB7219  CTCTCT...A GGGAGTTCTT .GTGG....A ATAATGTTTT .....GGA.T
 REH7379  GCCTCT.... AGGGGACCTT .GTA...GGA ATGACGATTT T....GGGA.
TFB10402  ATCTCTCTAG TAGGGATCTT .GTA.....G ATAATGATTT .....GAGG.
TFB11035  GTCTTT.... .AGGGACCCT .GTGAG.GGA ATAATGATTT T....GGA..
 TFB7230  GTCTCT.... .AGGGACCTT .GTG.....A ATATTGATTT T....GAA..
TFB10416  ATCTCT.... .AGGGATCTT .GTAAG.GGA ATAATGATTT T....GGA..
 REH8266  G.CTCTTT.. TAGAGACTCT .GTAAG.GGA TTTATGATTC T....GGG..
TFB10338  ...TCTTT.. ....GACTCT C.TAC..... .TGTTGATTT .....GAA..
TFB11340  ...TCTTT.. ....GACTCT C.TAC..... .TGTCAATTT .....GAA..
TFB11021  .....TTTT. ...CTAC... .......... .AGTCGATTT .....GAA..
TFB11013  .......... ....CACTT. .......... ..GTTGATTT TTT..GAA..
TFB10493  .......... ....CACTT. .......... ..GTTGATTT T....GAA..
TFB11439  .......... TGTTGAC... .......... ..TTCGA... .....AAAAA
TFB11040  .......... ....AGCTTC T........A AGTTAACTT. .....GGTA.
 TFB9697  .......... ....AACTTT C.....TG.A AGTTAACTT. .....GTTA.
TFB11434  .......... CTTTGGCTTA T.....TGCG GGTTGACTTT .....GGA..
TFB11329  .......... ...GTACTTG ......TGCA ATTTGGCTTT .....GAA..
 REH8290  .......... ...GTACTTG ......TGCA ATTTGACTTT .....GAA..
AF079580  .......... ...GTACTTG ......TGCA ATTTGACTTT .....GAA..
TFB11438  .......... ...GTACTTG ......TGCA ATTTGGCTTT .....GAA..
 TFB2887  .......... ...GTACCTG G.....TGCA ATTTGGCTTT T.....AAA.
TFB11025  .......... ...GTACTTG G.....TGCA ATTTCGCTTT T.....AAA.
TFB11015  .......... ...GTACTTG ......TGCA ATTTGGCTTT .....GAA..
TFB11470  .......... ...GTACTTG ......TGCA ATTTGGCTTT ......ACA.
TFB10095  .......... ...GCACTTG ......TGCA ATTTGGTTTT ......AAA.
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          351                                                      400
AF031178  GGAGG..TCC CTT.CTATGT ...CTT...C ACAAATGAT. TCAAAGTATG
AF356167  GGAGG..TCC CTT.CTATGT ...CTT...C ACAAATGAT. TCAAAGTATG
AF356169  AAAGA..... CTT.CTATGT T..CT....T ATAAACTA.. TTGAAGTATG
LNU33082  AAAGA..... CTT.CTATGT T..CT....T ATAAACTA.. TTGAAGTATG
AF356170  CAAGG..... CTT.CTATGT T..CT....T ATAAACCA.. TTGAAGTATG
AY016444  AGGGG..... CTT.CTATGT ...ATTA..T ATAAACTAT. TTGAAGTATG
TFB10726  TAGGA..... CTT.CTATGT ...CTTT... ACAAACCA.. TTGAAGTATG
TFB11456  TAGGA..... CTT.CTATGT ...CTTT... ACAAACCA.. TTGAAGTATG
TFB11014  TGAGA..... CTT.CTATGT ...CTTT... ATAAACCA.. TTGAAGTATG
 TFB9605  AGAGG..... CTT.CTATGT ...CTTT... ACAAAACTAT TTCAAGTATG
 EFM1403  CGAGG..... CTT.CTATGT T..CTTTTTT ACAAACACA. TTGAAGTATG
TFB10077  TGAGG..... CTT.CTATGT ...CTTC.TA ACAAACTA.. TT...GTATG
 REH7007  .......... CTT.CTATGT ...CTT.... ATAAACTA.. TTGAAGTATG
  EN2066  AAAGA..... CAT.CTATGT T..CTTT.AT A.AAACTC.. .....GTATG
 REH7907  TGAGA..... CTT.CTATGT T..CTTT... ACAAACTA.. .AAATG..TG
 TFB9920  AAAAA..... CTTT.TATGT ATACATTTTT ..AAGCTATG .AAAAGTATG
 REH7348  .GAGA..... CTT.CTATGT ...CTTT... ACAAACC.AG TTAA.GCATG
 TFB9699  .GAGA..... CTT.CTATGT ...CTTT... ACAAACC.AG TTAA.GCATG
TFB11026  TGAGG..... CTT.CTATAT ...CTTT... ACAAACC.AG TTAA.GCATG
 TFB1871  .AAGG..... CTT.CTATGT ...CTT.... ACAAACT.AG TTAA.GCATG
TFB10350  .GAGG..... CTT.CTATGT ...CTTT... ACAAACC.AG TTAA.GCATG
TFB10422  .AGGG..... CTT.CTATGT TT..TT.... ACAAACA.AG TTAA.GCATG
 TFB9657  .AGAG..... CTT.CTATGT TT..TT.... ACACACA.AG TTAA.GCATG
TFB10080  .AGAG..... CTT.CTATGT TTT....... ATAAACA.AG TTAA.GCATG
 TFB7219  TAGAG..... CTT.CTATGT T..CTT.... ACAAACTCAG TTAA.GCATG
 REH7379  TAGAG....C CT..CTATGT T...TTT... ACCAACT.AG TTAA.GCATG
TFB10402  .AGAG..... CTT.CTATGT TTT....... ACAAACT.AG TTAA.GCATG
TFB11035  CAGAG..... CTT.CTATGT TTT....... ACAAACA.AG TTAA.GCATG
 TFB7230  TAGAG..... CTT.CTATGT TTTT...... ACAAACA.AG TTAA.GCATG
TFB10416  TAGAG..... CTT.CTATGT T...TTT... ACAAACC.AT TTAA.GCATG
 REH8266  TAAAG..... CTT.CTATGT T..CTT.... ACAAACA.AG TTAA.GCATG
TFB10338  ..GGG..... CTT.CTATGT T..CTT.... ACAAACA.AG TTAA.GCATG
TFB11340  ..GGG..... CTT.CTATGT T..CTT.... ACAAACA.AG TTAA.GCATG
TFB11021  ..GGGA.... TTTCCTATGT TT.CT..... ACAAACT.AT ATAA.GCATG
TFB11013  ..GG.ACTGC CTT.CTATGT ...CTTT... ATACACTTTG TTAATGTATG
TFB10493  .GGGGACTA. CTT.CTATGT ...ATTT... ATACACTC.G TAAATGTATG
TFB11439  AAGGG..... CTT.CTATGT T..CTT.... ACAAACCCA. TTGAAGTATG
TFB11040  AAGGG....C CTT.CTATGT T..CTC.... ACAAACCTA. TTGAAGCATG
 TFB9697  AAGGG..... CTTTCTATGT T..CTC.... ATAAACCTA. TTGAAGTATG
TFB11434  .AAGA..... CTT.CTATGT T...TTT... ACAAACCTAT TTGAAGTATG
TFB11329  ..AGG..... CTT.CTATGT ...CTT.... ATAAACCCA. TTA..GTATG
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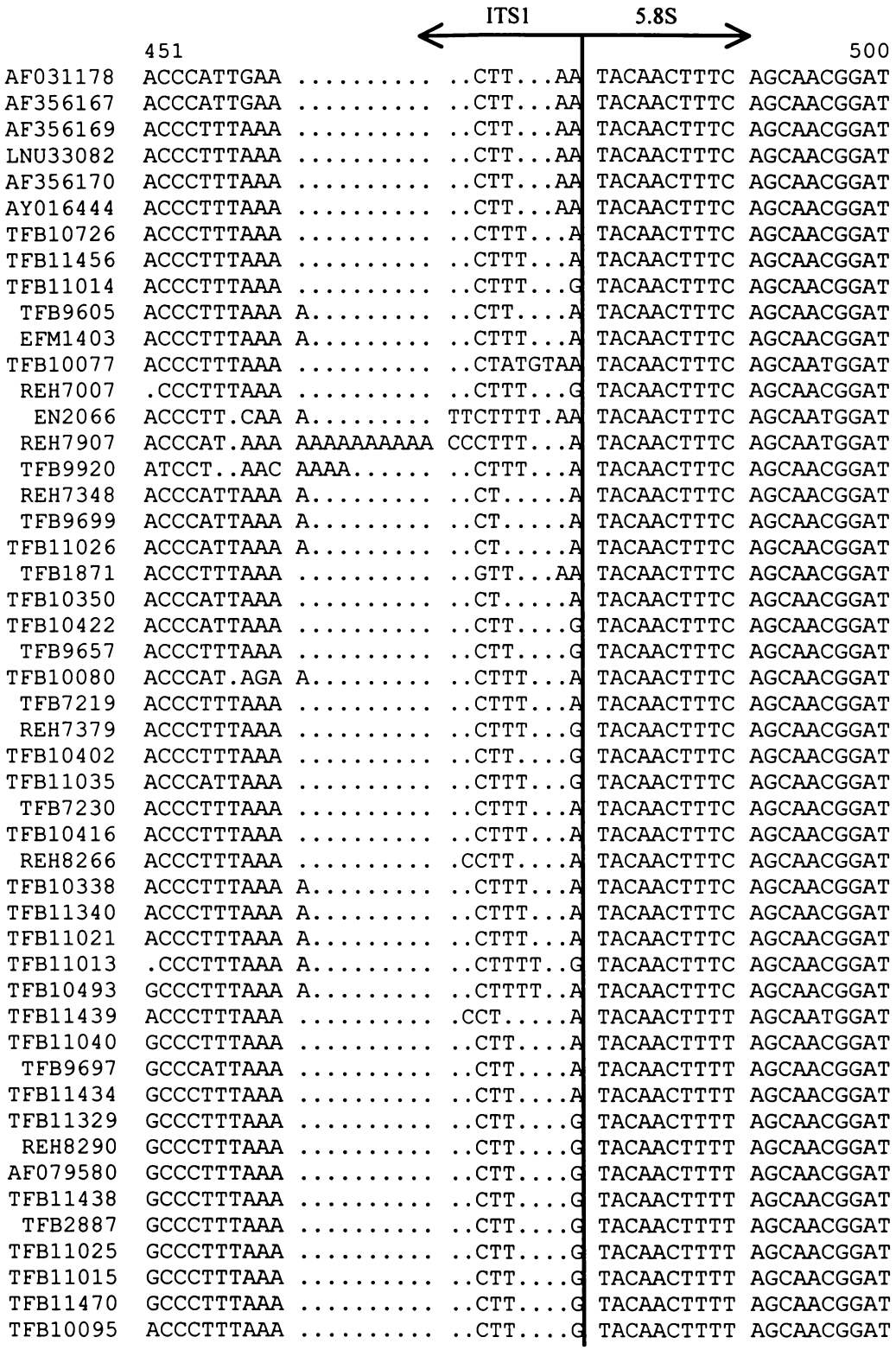
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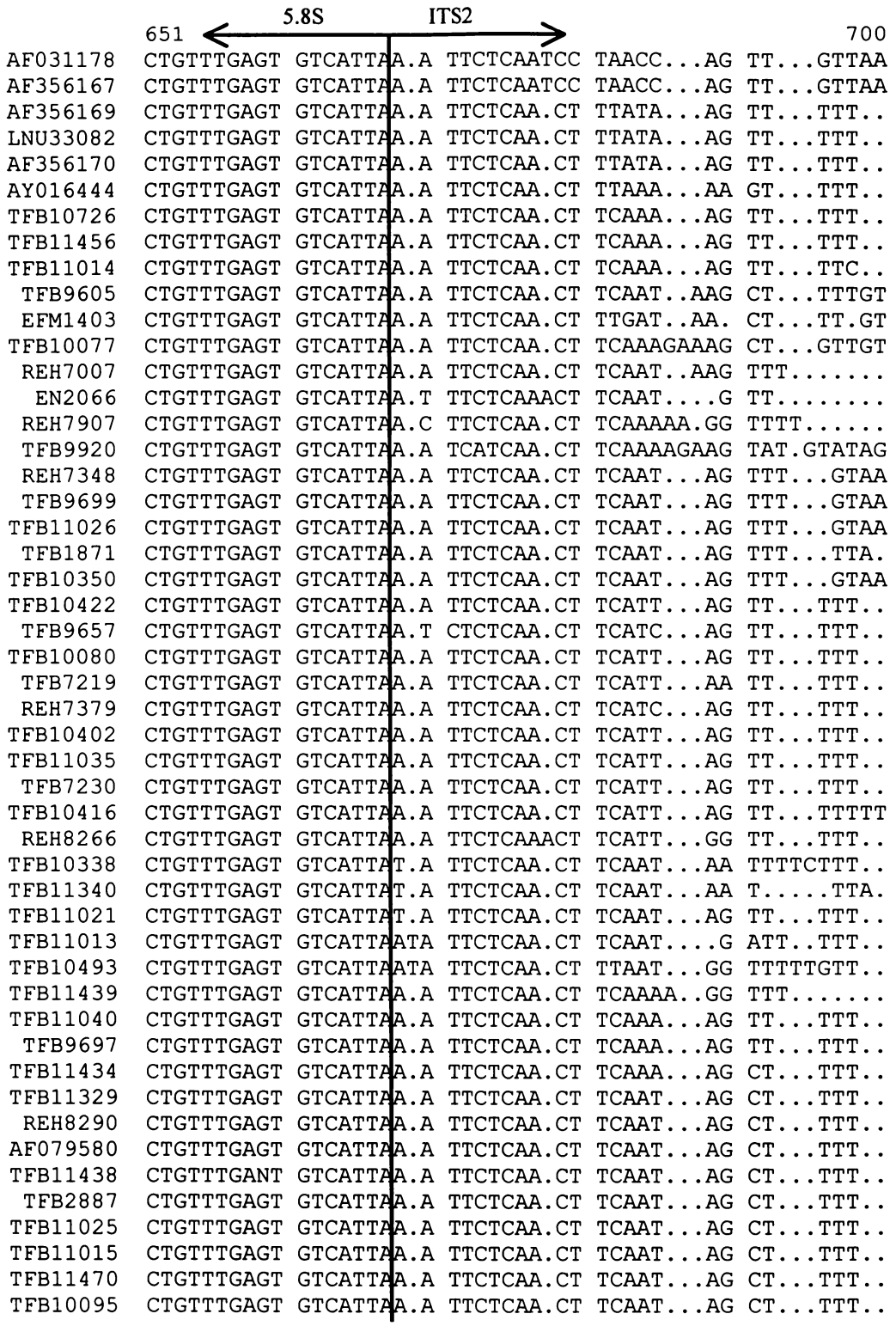
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AF356167 CTGTTTGAGT GTCATTAA.A TTCTCAATCC TAACC...AG TT...GTTAA
AF356169 CTGTTTGAGT GTCATTAA.A TTCTCAA.CT TTATA...AG TT...TTT..
LNU33082 CTGTTTGAGT GTCATTAA.A TTCTCAA.CT TTATA...AG TT...TTT..
AF356170 CTGTTTGAGT GTCATTAA.A TTCTCAA.CT TTATA...AG TT...TTT..
AY016444 CTGTTTGAGT GTCATTAA.A TTCTCAA.CT TTAAA...AA GT...TTT..
TFB10726 CTGTTTGAGT GTCATTAA.A TTCTCAA.CT TCAAA...AG TT...TTT..
TFB11456 CTGTTTGAGT GTCATTAA.A TTCTCAA.CT TCAAA...AG TT...TTT..
TFB11014 CTGTTTGAGT GTCATTAA.A TTCTCAA.CT TCAAA...AG TT...TTC..
TFB9605 CTGTTTGAGT GTCATTAA.A TTCTCAA.CT TCAAT..AAG CT...TTTGT
EFM1403 CTGTTTGAGT GTCATTAA.A TTCTCAA.CT TTGAT..AA. CT...TT.GT
TFB10077 CTGTTTGAGT GTCATTAA.A TTCTCAA.CT TCAAAGAAAG CT...GTTGT
REH7007 CTGTTTGAGT GTCATTAA.A TTCTCAA.CT TCAAT..AAG TTT.......
EN2066 CTGTTTGAGT GTCATTAA.T TTCTCAAACT TCAAT....G TT........
REH7907 CTGTTTGAGT GTCATTAA.C TTCTCAA.CT TCAAAAA.GG TTTT......
TFB9920 CTGTTTGAGT GTCATTAA.A TCATCAA.CT TCAAAAGAAG TAT.GTATAG
REH7348 CTGTTTGAGT GTCATTAA.A TTCTCAA.CT TCAAT...AG TTT...GTAA
TFB9699 CTGTTTGAGT GTCATTAA.A TTCTCAA.CT TCAAT...AG TTT...GTAA
TFB11026 CTGTTTGAGT GTCATTAA.A TTCTCAA.CT TCAAT...AG TTT...GTAA
TFB1871 CTGTTTGAGT GTCATTAA.A TTCTCAA.CT TCAAT...AG TTT...TTA.
TFB10350 CTGTTTGAGT GTCATTAA.A TTCTCAA.CT TCAAT...AG TTT...GTAA
TFB10422 CTGTTTGAGT GTCATTAA.A TTCTCAA.CT TCATT...AG TT...TTT..
TFB9657 CTGTTTGAGT GTCATTAA.T CTCTCAA.CT TCATC...AG TT...TTT..
TFB10080 CTGTTTGAGT GTCATTAA.A TTCTCAA.CT TCATT...AG TT...TTT..
TFB7219 CTGTTTGAGT GTCATTAA.A TTCTCAA.CT TCATT...AA TT...TTT..
REH7379 CTGTTTGAGT GTCATTAA.A TTCTCAA.CT TCATC...AG TT...TTT..
TFB10402 CTGTTTGAGT GTCATTAA.A TTCTCAA.CT TCATT...AG TT...TTT..
TFB11035 CTGTTTGAGT GTCATTAA.A TTCTCAA.CT TCATT...AG TT...TTT..
TFB7230 CTGTTTGAGT GTCATTAA.A TTCTCAA.CT TCATT...AG TT...TTT..
TFB10416 CTGTTTGAGT GTCATTAA.A TTCTCAA.CT TCATT...AG TT...TTTTT
REH8266 CTGTTTGAGT GTCATTAA.A TTCTCAAACT TCATT...GG TT...TTT..
TFB10338 CTGTTTGAGT GTCATTAT.A TTCTCAA.CT TCAAT...AA TTTTCTTT..
TFB11340 CTGTTTGAGT GTCATTAT.A TTCTCAA.CT TCAAT...AA T.....TTA.
TFB11021 CTGTTTGAGT GTCATTAT.A TTCTCAA.CT TCAAT...AG TT...TTT..
TFB11013 CTGTTTGAGT GTCATTAATA TTCTCAA.CT TCAAT....G ATT..TTT..
TFB10493 CTGTTTGAGT GTCATTAATA TTCTCAA.CT TTAAT...GG TTTTTGTT..
TFB11439 CTGTTTGAGT GTCATTAA.A TTCTCAA.CT TCAAAA..GG TTT.......
TFB11040 CTGTTTGAGT GTCATTAA.A TTCTCAA.CT TCAAA...AG TT...TTT..
TFB9697 CTGTTTGAGT GTCATTAA.A TTCTCAA.CT TCAAA...AG TT...TTT..
TFB11434 CTGTTTGAGT GTCATTAA.A TTCTCAA.CT TCAAA...AG CT...TTT..
TFB11329 CTGTTTGAGT GTCATTAA.A TTCTCAA.CT TCAAT...AG CT...TTT..
REH8290 CTGTTTGAGT GTCATTAA.A TTCTCAA.CT TCAAT...AG CT...TTT..
AF079580 CTGTTTGAGT GTCATTAA.A TTCTCAA.CT TCAAT...AG CT...TTT..
TFB11438 CTGTTTGANT GTCATTAA.A TTCTCAA.CT TCAAT...AG CT...TTT..
TFB2887 CTGTTTGAGT GTCATTAA.A TTCTCAA.CT TCAAT...AG CT...TTT..
TFB11025 CTGTTTGAGT GTCATTAA.A TTCTCAA.CT TCAAT...AG CT...TTT..
TFB11015 CTGTTTGAGT GTCATTAA.A TTCTCAA.CT TCAAT...AG CT...TTT..
TFB11470 CTGTTTGAGT GTCATTAA.A TTCTCAA.CT TCAAT...AG CT...TTT..
TFB10095 CTGTTTGAGT GTCATTAA.A TTCTCAA.CT TCAAT...AG CT...TTT..

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          701                                                       750
AF031178  TT.....GAC TGCGTT...A GAATTGGAAC T..GGAGG.T TT..GTTGGC
AF356167  TT.....GAC TGCGTT...A GCATTGGAAC T..GGAGG.T TT..GTTGGC
AF356169  ........AC TTATT..AAA GCTT.GGATA TT.GGAGG.T TT..GCAGGC
LNU33082  ........AC TTCTT..AAA GCTT.GGATA TT.GGAGG.T TT..GCAGGC
AF356170  ........AC TTATT..AAA GCTT.GGATG TT.GGAGG.C TT..GCAGGC
AY016444  .......... .......AAA GCTT.GGATG TTTGGAGG.T TT..GCTGAC
TFB10726  ....ATTAAC TTATT..GAA GCTT.GGATG T..GGGGG.T TT..GTTGGC
TFB11456  ....ATTAAC TTATT..GAA GCTT.GGATG T..GGGGG.T TT..GTTGGC
TFB11014  ....ATTAAC TTATT..GAA GCTT.GGATG T..GGAGGCT T...GTTGGC
 TFB9605  T...AGT... TTATT..GGA GCTT.GGATG T..GGAGG.T T...GCTGGC
 EFM1403  TT........ ..ATT..GAA GCTT.GGATG T..GAAGG.T TT..GCTGGC
TFB10077  TTT......C TT.TT..GGA GCTT.GGTTG T..GGAGG.T TTT.GTTGGC
 REH7007  .......CAC TTATTT.GAG GCTT.GGATG T..GGAGG.T TTTTGTTGGC
  EN2066  .......... .......GAA GCTTTGGATG T..GGGAGCT T...GTTGGC
 REH7907  .........C TTTTTTTGTT GTTT.GGATG T..GGAGG.T TTT.GCTGGT
 TFB9920  .........C T.ATTTTGAA GTTT.GGTTG T..GGGGA.T TT..GCTGGC
 REH7348  .....TGAAC TTTTT..GAA GCTT.GGATG T..GGAGG.T TT..GCTGGC
 TFB9699  .....TGAAC TTTTT..GAA GCTT.GGATG T..GGAGG.T TT..GCTGGC
TFB11026  .....CGAAC TGTT...GAA GCTT.GGATG T..GGAGG.T TTT.GCTGGC
 TFB1871  .....TTAAC TCGTT..GAA GCTT.GGATG T..GGAGG.T TT..GCTGGC
TFB10350  .....TGAAC TGCT...GAA GCTT.GGATG T..GGAGG.T TTT.GCTGGC
TFB10422  ....AT.AAC TGATT..GAA GCTT.GGATG T..GGGGG.T TTT.GCTGGC
 TFB9657  ....ATTAAC TGATTT.GAA GCTT.GGATG T..GGGGGCT T...GCTGGC
TFB10080  ....AT.AAC TGATT..GAA GCTT.GGATG T..GGGGG.T TT..GCTGGC
 TFB7219  ....AT.AAT TGATT..GAA GCTT.GGATG T..GGGGG.T TT..GCTGGC
 REH7379  ....AT.AAC CGATT..GAA GCTT.GGATG T..GGGGG.T TT..GCTGGC
TFB10402  ....AT.AAC TGATT..GAA GCTT.GGATG T..GGGGG.T TT..GCTGGC
TFB11035  ....AT.AAC TAATT..GAA GCTT.GGATG T..GGGGG.T TT..GCTGGC
 TFB7230  ....AT.AAC TGATT..GAA GCTT.GGATG T..GGGGG.T TT..GCTGGC
TFB10416  ....AT.AAC TAGTT..GAA GCTT.GGATG T..GGGGG.T TT..GCTGGC
 REH8266  ...AAT.AAC TGAT...GAA GCTT.GGATG T..GGAGG.T TT..GCTGGC
TFB10338  ...AAT.... .TGTT..GAA GCTT.GGATG T..GGAGGCT TTT.GCTGGC
TFB11340  .......... TTGTT..GAA GCTT.GGATG T..GGAGGCT TTT.GCTGGC
TFB11021  ....CTTTA. TTATT..GAA GTCT.GGATG T..GGAGG.T TTTTGCTGGC
TFB11013  ....GTAGTC ..ATT..GAA GCTT.GGATG T..GGAGGCT TTT.GTTGGT
TFB10493  .......ATC ..ATT..GAA GCTT.GGAAG T..GGAGGGT TTTTGCTGGT
TFB11439  ..TCATTAAC TTTCT..GAA GCTT.GGATG T..GGGGGCT T...GCTGGC
TFB11040  ....ATTAAC TTATT..GGA GCTT.GGATA T..GGGAGCT T...GCTGGC
 TFB9697  ....ATTAAC TTTTT..GAA GCTT.GGATG T..GGGAGCT T...GCTGGC
TFB11434  ....ATTAGC TTATT..GAA GCTT.GGATG T..GGGGGCT T...GCTGGC
TFB11329  ....ACTAGC TTATT..GAT GCTT.GGATG T..GGGAGCT T...GCTGGC
 REH8290  ....ACTAGC TTATT..GAT GCTT.GGATG T..GGGAGCT T...GCTGGC
AF079580  ....ACTAGC TTATT..GAT GCTT.GGATG T..GGGAGCT T...GCTGGC
TFB11438  ....ACTAGC TTATT..GAT GCTT.GGATG T..GGGAGCT T...GCTGGC
 TFB2887  ....ATTAGC TTATT..GAT GCTT.GGATG T..GGGAGCT T...GCTGGC
TFB11025  ....ACTAGC TTATT..GAT GCTT.GGATG T..GGGAGCT T...GCTGGC
TFB11015  ....ACTAGC TTATT..GAT GCTT.GGATG T..GGGAGCT T...GCTGGC
TFB11470  ....ATTAGC TTATT..GAT GCTT.GGATG T..GGGAGCT T...GCTGGC
TFB10095  ....ACTAGC TTATT..GAT GCTT.GGATG T..GGGAGCT T...GCTGGC
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          751                                                        800
AF031178  TTC.......  .....TGTT.  ..CAACTCCT CTA.AAATAT ATCAGTG..G
AF356167  ATC.......  .....TGT..  .CCAACTCCT CTA.AAATAT ATCAGTG..G
AF356169  GTT.......  .....TGT..  ..CAGCTCCT CTT.AAATTT ATTAGTG..G
LNU33082  GTT.......  .....TGT..  ..CAGCTCCT CTT.AAATTT ATTAGTG..G
AF356170  GTT.......  .....TGT..  ..CAGCTCCT CTT.AAATTT ATTAGTG.GG
AY016444  TTTC......  .....TGTTT  ...AGCTCCT CTT.AAATGC ATTAGTG..G
TFB10726  GTCT.....T  TAGA.TGTT.  ..CAGCTCCC CT.TAAATGC ATTAGTG..G
TFB11456  GTCT.....T  TAGA.TGTT.  ..CAGCTCCC CT.TAAATGC ATTAGTG..G
TFB11014  GTCT......  TAGA.TGTT.  ..CAGCTCCT CT.TAAATAC ATTAGTG..G
 TFB9605  GTCC.....T  TGGA.TGTTT  T.CAGCTCRT CT.TAAATGC ATTAGTG..G
 EFM1403  GTCT......  TAGA.TGCTT  T.CAGCTCCT TT.TAAATAT ATTAGTG..G
TFB10077  AGC...GCTT  T.GTTTGT..  ..CAGCTCCT CT.CAAATAT ATCAGTG..G
 REH7007  ATCTC.....  .AGA.TGTT.  ..CAACTCCT CTT.AAATGT ATTAGTG..G
  EN2066  TTT...GTTT  AATAAAC...  ..CAACTCTC CT.TAAACAT ATTAGTG..G
 REH7907  TATCTCGTTT  ..GA..GTTT  ..CAGCTCCT CT.TAAATGC ATTAGTG..G
 TFB9920  AAC...GTCT  CA....GTT.  ..CAGCTTCT CT.TAAATAC ATTAGTG...
 REH7348  GTCT......  TAGTATGTT.  ..CGGCTCCT CTT.AAATGC ATTAGTG..G
 TFB9699  GTCT......  TAGTATGTT.  ..CGGCTCCT CTT.AAATGC ATTAGTG..G
TFB11026  GTCT......  TAGTATGTT.  ..CGGCTCCT CTT.AAATGC ATTAGTG..G
 TFB1871  GTCT......  TAG.ATGTT.  ..CGGCTCCT CTT.AAATGC ATTAGTG.GG
TFB10350  GTCT......  TAGTATGTT.  ..CGGCTCCT CTT.AAATGC ATTAGTG..G
TFB10422  GTCTT.....  .AGA.TGTTT  ..CGGCTCTC CTT.AAATGC ATTAGTA.GG
 TFB9657  GTCTT.....  .AGA.TGTT.  ..CGGCTCTC CTT.AAATGC ATTAGTG.GG
TFB10080  GTCTT.....  .AGA.TGT..  .CCGGCTCCT CTT.AAATGT ATTAGTA.GG
 TFB7219  GTCTTT....  CAGA.TGTTT  T.CGGCTCCC CTT.AAATGC ATTAGTA.GG
 REH7379  GTC....ATT  .AGA.YGTTT  T.CGGCTCCC CTT.AAATGC ATTAGTA.GG
TFB10402  ATCTT.....  .AGATGCTTT  ...GGCTCTC CTT.AAATGC ATTAGTG.GG
TFB11035  GTCC...AAT  GAGA.TGTTT  T.CGGCTCCC CTT.AAAGGC ATTAGTA.GG
 TFB7230  GTCTTT....  .AGA.TGTTT  T.CGGCTCCC CTT.AAATGC ATTAGTA.AG
TFB10416  GTC....ATT  .AGAC.GTTT  TT.GGCTCCC CTT.AAATGC ATTAGTA.GG
 REH8266  ATC....ATT  AAGA.TGTTT  T..GGCTCCT CTT.AAATGC ATTAGTG.GG
TFB10338  ATCTC.....  .AGA.TGTC.  ..CGGCTCCT CTT.AAATGC ATTAGTG..G
TFB11340  ATCTC.....  .AGA.TGTC.  ..CGGCTCCT CTT.AAATGC ATTAGTG..G
TFB11021  ATTGC.....  AAAA.TGCT.  ..CAGCTCCT CTT.AAATAC ATTAGTG..G
TFB11013  GTCTTCTT..  .ACA.CATT.  ...GGCTCCT CT.GAAATGC ATTAGTGGGG
TFB10493  GTCTC.....  TAGA.TATT.  ...AGCTCCT CT.GAAATGC ATTAGTG.GG
TFB11439  GTCT.....T  TGGA.TGTT.  ..CGGCTCTC CT.TAAAGAT ATTAGTGGGG
TFB11040  GTCTT.....  .AGA.TGTT.  ..CGGCTCTC CTT.AAATTT ATTAGTG..G
 TFB9697  GTCTT.....  CAGA.TGTT.  ..CGGCTCTC CTT.AAATTT ATTAGTG..G
TFB11434  GTCT......  CAGA.TGTT.  ..CGGCTCTC CTT.AAAGTT ATTAGTG..G
TFB11329  GTCT......  TAGA.TGTT.  ..CGGCTCTC CTT.AAAGTT ATTAGTG..G
 REH8290  GTCT......  TAGA.TGTT.  ..CGGCTCTC CTT.AAAGTT ATTAGTG..G
AF079580  GTCT......  TAGA.TGTT.  ..CGGCTCTC CTT.AAAGTT ATTAGTG..G
TFB11438  GTCT......  TAAA.TGTT.  ..CNGCTNTN CTT.AAAGTT ATTANTG..G
 TFB2887  GTCT......  TAGA.TGTT.  ..CGGCTCTC CTT.AAAGTG ATTAGTG..G
TFB11025  GTCT......  TAGA.TGTT.  ..CGGCTCTC CTT.AAAGTT ATTAGTG..G
TFB11015  GTCT......  TAGA.TGTT.  ..CGGCTCTC CTT.AAAGTT ATTAGTG..G
TFB11470  GTCT......  TAGA.TGTT.  ..CGGCTCTC CTT.AAAGTT ATTAGTG..G
TFB10095  GTCT......  TAGA.TGTT.  ..CGGCTCTC CTT.AAAGTT ATTAGTG..G
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          801                                                        850
AF031178  ATTTT.AACA ....CCCTGC TTTGTT.GAT CC.....CAA CCTGTTTAAG
AF356167  ATTTT.AACA ....CCCTGC TTTGTTTGAT CC.....CAA CCTGTTTAAG
AF356169  AA........ ....CCCTGT TTTGTTGGTT TT.....CAA CCTT.....G
LNU33082  AA........ ....CCCTGT TTTGTTGGTT TT.....CAA CCTT.....G
AF356170  AA........ ....CCCTGT TTTGTTAGTT CT......AA CCTT.....G
AY016444  AAT....... ......CTTA TTTGTTGAAT TCA.....TT CCAT.....A
TFB10726  AATT...... ....CCC.GT TTGGTTGGT. CCA....... TCTT.....G
TFB11456  AATT...... ....CCC.GT TTGGTTGGT. CCA....... TCTT.....G
TFB11014  AATT...... ....CCC.GT TTGGTTGGT. CCA....... TCTT.....G
 TFB9605  AAC....... ....CCC.GT AGTGTTGGT. CCA....... TCTT.....G
 EFM1403  AGTTTTACAT GTTCCTC.GT TGTGTTGCT. CCA....... TCTT.....G
TFB10077  AATCCTAAAC ..TCCTCCGT TGTGTTGGT. CCA....... CCTT.....G
 REH7007  AA.CTT.... ....CTC.GT TTTGTTGGT. CCAA.....T CCTT.....G
  EN2066  AAA....... ...CCCCAGT TGTGTTGGTT CCA....... TCTT.....G
 REH7907  AGACTTATAA ....CCC.GT TGTGTTGATC CCA....... TTTT.....G
 TFB9920  AA.CTG.... ....TTTAGT TACT..GGT. TCA....... TCGTT....A
 REH7348  AA........ ....CCC.GT TT.GTTGGA. TCAT...... CCAT.....A
 TFB9699  AA........ ....CCC.GT TT.GTTGGA. TCAT...... CCAT.....A
TFB11026  AA........ ....CCC.GT TT.GTTGGA. CTAT...... CCAT.....A
 TFB1871  A......... ...CCCC.GT TT.ATTGGA. CAAT...... CCTT.....G
TFB10350  AA........ ....CCC.GT TT.GTTGGAA CTAT...... CCAT.....A
TFB10422  AA........ ....CCC.GT TT.GTTGG.. CGT....... CCAT.....A
 TFB9657  AA........ ....CCC.GT TT.GTTGG.. CGT....... CCAT.....A
TFB10080  AA........ ....CCC.GT TT.GTTGG.. CGT....... CCAT.....A
 TFB7219  AA........ ....CCC.GT TT.GTTGG.. CGT....... CCAA.....A
 REH7379  AA........ ....CCC.GT TT.GTTGG.. CGT....... CCAT.....A
TFB10402  AA........ ....CCC.GT TT.GTTGG.. CGT....... CCAT.....A
TFB11035  AA........ ....CCC.GT TT.GTTGG.. CGT....... CCTT.....G
 TFB7230  AA........ ....CCC.GT TT.GTTGG.. CGT....... TCAT.....T
TFB10416  AA........ ....CCC.GT TT.GTCGG.. CGT....... CCAT.....A
 REH8266  AA........ ....CCC.GT TT.GTTGG.. CGT....... CCTT.....G
TFB10338  AA........ ....CCC.GT TCAGTTGGG. CTAT...... CCTT.....G
TFB11340  AA........ ....CCC.GT TCAGTTGGG. CTAT...... CCTT.....G
TFB11021  AAT....... ....CCC.AT TTCGTTGGG. CTAT...... TCTT.....G
TFB11013  AATAA..... ....TCT.AC TTCGTTGGGG CTATTGTT.T CCTT.....A
TFB10493  AAA....... ...TGTTTAC TTCGTTGG.A CTATTATT.T CCTT.....A
TFB11439  AA........ ....TGT.CT TTTGTTGGG. CGG......T CCTT.....G
TFB11040  AAA....... ....TCT.CT TT.GTTGGAG TTAGT....C CCT......G
 TFB9697  AAA....... ....TCT.CT TT.GTTGGAG TCTAAT...C CCT......A
TFB11434  AAA....... ....TCT.CT TT.GTTGGGG TGAT...... CCTT.....G
TFB11329  AAA....... ....CCT.CT TT.GTTGGTT .TATT..... CCTT.....A
 REH8290  AAA....... ....CCT.CT TT.GTTGGTT .TATT..... CCTT.....A
AF079580  AAA....... ....CCT.CT TT.GTTGGTT .TATT..... CCTT.....A
TFB11438  AAA....... ....CCT.CT TT.GTTGGTT .TATT..... CCTT.....A
 TFB2887  AAA....... ....CCT.CT TT.GTTGGGT .CATT..... CCTT.....G
TFB11025  AAA....... ....CCT.CT TT.GTTGGGT .TATT..... CCTT.....G
TFB11015  AAA....... ....CCT.TT TT.GTTGGGT .TATT..... CCT......G
TFB11470  ATAAA..... ....CCT.CT TT.GTTAGGT .TATT..... .CTT.....G
TFB10095  AAAA...... ....CCT.CT TT.GTTAGGT .CATT..... .CTA.....A
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          851                                                        900
AF031178  GTG.TGATAA .T..TATCTA CGCTTT.... ......TT.A ACAG......
AF356167  GTG.TGATAA .T..TATCTA CGCTTT.... ......TT.A ACAG......
AF356169  GTG.TGATAA .C..TATCTA CA.TTTTGGT TTGT...... ..........
LNU33082  GTG.TGATAA .T..TATCTA CA.TTTTGGT TTGT.....A ACCT...TA.
AF356170  GTG.TGATAA .T..TATCTA CA.TTTTGG. TGG......A ACCT...TA.
AY016444  GTG.TGATAA .T..TATCTA CACTTT.GGT .......... ..........
TFB10726  GTG.TGATAA .T..TATCTA CGCTTT.GCT TGG...TTCA ACTGT.....
TFB11456  GTG.TGATAA .T..TATCTA CGCTTT.GCT TGG...TTCA ACTGT.....
TFB11014  GTG.TGATAA .T..TATCTA CGCTTT.GTT TGG...TTCA ACTGTT....
 TFB9605  ATG.TGATAG .T..TATCTA CATTTT.GG. TGG...TTCA GCTGT....T
 EFM1403  GTG.TGATAA .T..TATCTA CATCTT.GAT GTG...TTCA GCTGTT.TTT
TFB10077  GTG.TGATAG .T..TATCTA CATCTT.GG. TGG...TTCA ATTGT..TTT
 REH7007  GTG.TGATAA .T..TATCTA CACTTT.GGT TGG...TTCA ACT...AG..
  EN2066  ATGGTGATAA .T..TATCTA CATTTT.GA. TGG...TTCA ACTGT....T
 REH7907  ATG.TGATAA .T..TATCTA CATTTT.AG. TGG..TTTCA GCTTT..GCT
 TFB9920  ATG.TGATAA GTGATATCTA CATT...GC. T.GAT...GA GCC.......
 REH7348  GTG.TGATAA .T..TATCTA CGCTTT.GGT TAG...TTCA ATGAACTCA.
 TFB9699  GTG.TGATAA .T..TATCTA CGCTTT.GGT TAG...TTCA ATGAACTCA.
TFB11026  GTG.TGATAA .T..TATCTA CGCTTT.GG. TTGA..TTCA ATGAACTCT.
 TFB1871  GTG.TGATAA .T..TATCTA CGCTTT.GGT TAG...TTCA ATGAACTCT.
TFB10350  GTG.TGATAA .T..TATCTA CGCTTT.GG. TTGA..TTCA ATGAACTCTT
TFB10422  GTG.TGATAA .T..TATCTA CGCTTT.GGT TAG...TTCA ACAGGCTCTA
 TFB9657  GTG.TGATAA .T..TATCTA CGCTTT.GGT TAG...TTCA ACAAGCTCTA
TFB10080  GTG.TGATAA .T..TATCTA CGCTTT.GGT TAG...TCCA TCAGACTCTA
 TFB7219  GTG.TGATAA .T..TATCTA CGCTTTTGGT TAG...TCTA ACAGGCTCGA
 REH7379  GTG.TGATAA .T..TATCTA CACTTT.GGT TAG...TCTA ACAGGCTCTA
TFB10402  GTG.TGATAA .T..TATCTA CGCTTT.GGT TAG...TTCA ACAGACTCTA
TFB11035  GTG.TGATAA .T..TATCTA CACTTT.GGT TAG...TCTA ACACACTCTG
 TFB7230  GTG.TGATAA .T..TATCTA CACATT.GGT CAG...TTCA ACAGGCTCTA
TFB10416  GTG.TGATAA .T..TATCTA CGCTTT.GGT TAG...TCCG ACAGGCTCTA
 REH8266  GTG.TGATAA .T..TATCTA CACTTT.GGC TAG...TCTG ACAGGCTCTA
TFB10338  GTTGTGATAA .T..TATCTA CGCTTTGGAT TAGTC.TTCA ACAAACTCTT
TFB11340  GTTGTGATAA .T..TATCTA CGCTTTGGAT TAGTC.TCTA ACAAACTCTT
TFB11021  GTG.TGATAA .T..TATCTA CGCTTTGAAT CAG..TTCTA ACAAGCTCTC
TFB11013  GTG.TGATAG GT..TATCTA CGCTTCGGAT TGG...CCCA AT.GGT...T
TFB10493  GTG.TGATAG .T..TATCTA CGCTTTGGAT TGG...TCTA AT........
TFB11439  GTG.TGATAG .T..TATCTA CGCTTT.GGA AAG...TCTA ATGGTTCC..
TFB11040  GTG.TGATAA .T..TATCTA CTCCTT.GGA TGG...TCCA ATGTTTCC..
 TFB9697  GTG.TGATAA .T..TATCTA CATTTT.GGA TAG...TCCA ATGTTTCC..
TFB11434  GTG.TGATAA .T..TATCTA CGCTTT.GGA TAG...TCCA ATGTTTCC..
TFB11329  GTG.TGATAA .T..TATCTA CGCTAT.GAA TAA...TCTA ATGTT.CC..
 REH8290  GTG.TGATAA .T..TATCTA CGCTAT.GAA TAA...TCTA ATGTT.CC..
AF079580  GTG.TGATAA .T..TATCTA CGCTAT.GAA TAA...TCTA ATGTT.CC..
TFB11438  GNG.TGANAA .N..TATCTA CCCTAT.GAA TAA...TCTA ATGTT.CC..
 TFB2887  GTG.TGATAA .T..TATCTA CGCTTC.GGA TAA...CCTA ATGTT.CC..
TFB11025  GTG.TGATAA .T..TATCTA CGCTTT.GGA TAA...CCTA ATGTT.CC..
TFB11015  GTG.TGATAA .T..TATCTA CGCTTT.GAA TAA...TCTA ATGTT.CC..
TFB11470  GTG.TGATAA .T..TATCTA CGCTTT.GAA TAA...TCTA ATGTT.CC..
TFB10095  GTG.TGATAA .T..TATCTA CGCTTTTGAA CAGG..TCTA ATGTT.CC..
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          901                                                        950
AF031178  .......... .......... .......... .......... ..........
AF356167  .......... .......... .......... .......... ..........
AF356169  .......... .......... .......... .......... ..........
LNU33082  CAATAATAAA GCTCTA.... .......... .......... ..........
AF356170  CAATAATAAA GCTCTA.... .......... .......... ..........
AY016444  ..TTAGTTCA CGA....... .......... .......... ..........
TFB10726  ...CAGGTGT GCTT...... ........AG G......... ....CTAGAT
TFB11456  ...CAGGTGT GCTT...... ........AA G......... ....CTAGAT
TFB11014  ...CAGGTGT GCTT...... ........AG G......... ....CTAGAT
 TFB9605  C......... .......... ........AG A......... ...TCTT.TT
 EFM1403  C......... .......... ........AG AGCAG..... TCTTC.TG.T
TFB10077  C.TCAGT.TC .......... .......... .......... .TTTCTTG.A
 REH7007  .TTCAAGTTT T......... ......ATTA .......... .........T
  EN2066  C..AAAACAA AAC....... ......TAAG G......... ......TTTA
 REH7907  C.TC...... .......... ........AG AAA....... ...TCTGATT
 TFB9920  ..TAG..TTC .......... .........A AATT...... ..........
 REH7348  GTGAG.CTTC ......TTT. ........CAG AATTGC.... .........C
 TFB9699  GTGAG.CTTC ......TTT. ........CAG AATTGC.... .........C
TFB11026  ATGAA.CTTC ......TTT. ........CAG AATTGC.... .........T
 TFB1871  ATGAG.TGTT ......TTT. ........CAG ACTTGTTCTG .......GGT
TFB10350  TGGA..CTTC ......TTT. ........CAG AATTGC.... .........T
TFB10422  ATGAAGCT.G TACCGTTTTT ....CTTCAG TACGCTCANG CG.TCTCTGG
 TFB9657  ATGAA.TGTG TACCA.TCTT TTTT.CTCAG AGCGTTTACG CGTTT.CTGG
TFB10080  GTGATGT..A TACTATCT.. .....CTCAG AACGCTTAGG CG.TCTCTGG
 TFB7219  ATGGAAGTTA TACCATTT.. ....TTTCAG GACACTT.CG GTGTTTCTGG
 REH7379  GTGAAGTTTA TACTA..... .......... .......... ..........
TFB10402  ATAAAGTT.A TACTGTTTTT ....CTTCAG AACACTTTAT GTGTTTCTGG
TFB11035  ATGAAA.... .......... ......TCTA TA........ ......CTG.
 TFB7230  ATGAAGTT.A TACCATATAT ....CTTCAG AACACTAA.C GTGTTTCTGG
TFB10416  ATGAAGG... .....TTT.. ......ATAC TG........ ..........
 REH8266  ATGAAG.... .....CTT.. ......ATGC TG........ ..........
TFB10338  .......... AGTGA..... ......TCAA TGCA...... ..........
TFB11340  .......... AGTGG..... ......TCAA TGTA...... ..........
TFB11021  .......... AGTAG..... ......TCAG TGTA...... ..........
TFB11013  ATACT.CT.. A.TGT..... ......TCAT T........T G.........
TFB10493  ATACT.CTTT AGTGTGTAGT T.....GGAG AGA...TACT GT........
TFB11439  ATTGGGATTA GT........ ........AG T.ATT..... ..........
TFB11040  ATTGAGA.TT GTT....... .......CAG TGAAT..... ..........
 TFB9697  ATTGAGA.TT GTT....... .......CAG T.AAT..... ..........
TFB11434  ATTGAAGGTT ATT....... .......CAG T.GAT..... ..........
TFB11329  ATTGAGACTT GAGATTT... .....CTCAG TAGA...... ..AATACTGG
 REH8290  ATTGAGACTT GGGATTT... .....CTCAG TAGA...... ..AATACTGG
AF079580  ATTGAGACTT GGGATTT... .....CTCAG TAGA...... ..AATACTGG
TFB11438  ATTGNGACTT GANATTT... .....CTCAA TANA...... ..AATACTGG
 TFB2887  ATTGGGGCTT GAGATTT... .....CTCAG TAGA...... ..AATACTGG
TFB11025  ATTGGGGCCT GAGATTT... .....CTCAG TAGA...... ..AATACTGG
TFB11015  ATTGGGACTT GAGATTT... .....CTCAG TAGA...... ..AATACTGG
TFB11470  ATTGGGACTT GAGATTT... .....CTCAG TAGA...... ..AATACTGG
TFB10095  ATTGGGACTT GAGATTATTT .....CTCAG TAGA...... ..AATACTGG
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          951                                                        1000
AF031178  ..TTGGTTT. ...CACGAGT TACTCTTT.. .......... ......TGAT
AF356167  ..TTGGTTT. ...CACGAGT TACTCTTT.. .......... ......TGAT
AF356169  .......... .......... .......... .......... ..........
LNU33082  .......... .......... .......... .......... .....TTGGT
AF356170  .......... .......... .......... .......... .....TTGGT
AY016444  .......... .......... .......... .......... .........GT
TFB10726  CC........ .......... .......... .......... ..........
TFB11456  CC........ .......... .......... .......... ..........
TFB11014  CC........ .......... .......... .......... ..........
 TFB9605  TC........ .......... .......... .......... ..........
 EFM1403  TC........ .......... .......... .......... ..........
TFB10077  AC........ .......... .......... .......... ..........
 REH7007  TATTA..... ..TTACT... .......... .......... ..........
  EN2066  TG........ .......... .......... .......... .........TT
 REH7907  TC........ .......... .......... .......... ..........
 TFB9920  .......... .......... .......... .......... ..........
 REH7348  TAG....... ..GCATTC.. .......... .......... ..........
 TFB9699  TAG....... ..GCATTC.. .......... .......... ..........
TFB11026  TAG....... ..GCATTC.. .......... .......... ..........
 TFB1871  T.GGG.TTTT ..GCATTAGG TTGCTTT.AC CTGTTTTCTT CAGATGCCTT
TFB10350  TA........ .GGCATTC.. .......... .......... ..........
TFB10422  TAGGGGTTTT ..GCACTT.. .......... .......... ..........
 TFB9657  TTGGGGTTTT ..GCACTT.. .......... .......... ..........
TFB10080  TAGGGGTTTT ..GCACATT. .......... .......... ..........
 TFB7219  TATGGGTTTT GTGCACTTC. .......... .......... ..........
 REH7379  TG.AGGTTTT ..GCACATTG .......... .......... ..........
TFB10402  CGTGGGTTTT ..GCACTTG. .......... .......... ..........
TFB11035  TG.AGGTTTT ..GCACATTA A......... .......... ..........
 TFB7230  CGTGGGTTTT ..GCACTTTC GTGCTTTTCC TGTTTATTGG GTTTTGCACT
TFB10416  TG.AGGTTTT ..GCACATTG .......... .......... ..........
 REH8266  TG.AGGTTTT ..GCACATTG .......... .......... ..........
TFB10338  .CTTTT.... ..GTACAT.. .......... .......... ..........
TFB11340  .CTTTT.... ..GTACGT.. .......... .......... ..........
TFB11021  .CTTTTG... .TGTACAT.. .......... .......... ..........
TFB11013  ....TTTA.. ...TACA... .......... .......... .........A
TFB10493  ....GTTA.. GTTCAC.... ......TTTA TCTGTTCATT CATTGCATGA
TFB11439  .......... .......... .......... .......... ..........
TFB11040  .......... .......... .......... .......... ..........
 TFB9697  .......... .......... .......... .......... ..........
TFB11434  .......... .......... .......... .......... ..........
TFB11329  TCTGGGTAC. .TGCAATTAA ATT....... .......... ..........
 REH8290  TTTGGGTAC. .TGCAATTAA ATT....... .......... ..........
AF079580  TTTGGGTAC. .TGCAATTAA ATT....... .......... ..........
TFB11438  NCTGGGNAC. .TGCAANTAA ATT....... .......... ..........
 TFB2887  TCTGGGTAT. .TGCAATTAA TTT....... .......... ..........
TFB11025  TCTGGGTAT. .TGCAAGTAA TTT....... .......... ..........
TFB11015  TCTGGGTAT. .TGCAATTAA TTT....... .......... ..........
TFB11470  TCTGGGTAT. .TGCAATTAA TTTAT..... .......... ..........
TFB10095  TTTGGGTAT. .TGCATTT.. .......... .......... ..........
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          1001                                                    1050
AF031178  TTGGGT.... ....TGTGTT G......... ......... .........
AF356167  TTGGGT.... ....TGTGTT G......... ......... .........
AF356169  .......... .......... .......... ......... .........
LNU33082  TTGGGT.... ....TGTTG. .......... CAA....... .........
AF356170  TTGGGT.... ....TGTTG. .......... CA........ .........
AY016444  TTGAAT.... ....TGTTG. .......... CG........ .........
TFB10726  .TGGTT.... .....GTGAT A........T CGCATTAA.. .........
TFB11456  .TGGTT.... .....GTGAT A........T CGCATTAA.. .........
TFB11014  .TGGTT.... .....GTGAT A........T TGCATTA... .........
 TFB9605  .TGGTC.... ...AGGATAT TGCCCTT... ..CATT.... .........
 EFM1403  .TGGTC.... ...AGGATAA GT.....GTT CACATTA... .........
TFB10077  .TGGTT.... ...AAGATA. .......TTG TG.ATAA... .........
 REH7007  .TGGTT.... ...GTGATA. .........T TGCATTA... .........
  EN2066  TTGGT..... ..CGGGATA. .........T TGCAA..... .........
 REH7907  .TGGTT.... ..GAGAATA. .........T TGCAA...GG ATCTATC...
 TFB9920  ..GGT..... ..CT.AAT.T T........G TACAAA.... ..CTATT...
 REH7348  .TGGT..... ..GTGGGTAT T........G TACTT..... .........
 TFB9699  .TGGT..... ..GTGGGTAT T........G TACTT..... .........
TFB11026  .TGGTTT... ..G.GGTTAT T........G TACTT..... .........
 TFB1871  TTGGTGT... .TCTGG..AT T..GGGTGGT TGCATTA... .........
TFB10350  .TGGT..... ..GTGGGTAT T........G TACTT..... .........
TFB10422  ...GTGCTTT ACCTGGGTAT ....GGGTTT TGCGTTTTAA T.........
 TFB9657  ...GTGCTTT ACCTGGGTAT ....GGGTTT TGCGTTTTC. .........
TFB10080  ...GTGCTTT ATCTGGGTAT ....AGGTTT TGCATTTAAT .........
 TFB7219  GGTGTGCTTT GCCTGGGTAT ...GGGGTTT TGCGTACTT. .........
 REH7379  ...GTGCTTT TCCTGAGTAT ...GGGGTTT TGTATTCTTG T.........
TFB10402  ...TTGCTTT ACCTGGGTAT ....GGGTTT TGCATCTT.. .........
TFB11035  ...GTGCTTT TCCTGAGTAT ...GGGGTTT TGTATTTTTT TTTT......
 TFB7230  TTCGTGCTTT TCCTGTTTAT T...GGGTTT TGCACTTTCG TGCTTTTCCT
TFB10416  ...GTGCTTT TCCTGAGTAT ...GGGGTTC TGCGTTCTTA AT........
 REH8266  ...GTGCTTT TCCTGAGTAT ...GGGGTTT TGTAT..TCG .........
TFB10338  .TGGTT.... ...GGGGTA. .........T TGCATTT... .........
TFB11340  .TGGTTT... ...GGGGTA. .........T TGCATTC... .........
TFB11021  .TGGTTT... ...TGGATAA .........C TGCAATCT.. .........
TFB11013  GTGGTT.... ...GGGATA. .........T TGCATTAGCT C.........
TFB10493  GTGGTT.... ...TGGATA. .........T TGCATTA... .........
TFB11439  .....GCG.. ...AT..... .......... ....TAA... .........
TFB11040  .....GCCTT CTGGTGTTTT ...CTGGTTT T......... .........G
 TFB9697  .....GCCTT TGTAGCATTT ...CTGGTTT TTT....... .........G
TFB11434  .....GCCCT T..GTGGTAT TT.CTGGTTT .......... .........G
TFB11329  .TGCTGTAT. ...CTGTAAT .CTC.TCAGT AGC....... .....AATAC
 REH8290  .TGCTGTAT. ...CTGTAAT .CTC.TCAGT AGC....... .....AATAC
AF079580  .TGCTGTAT. ...CTGTAAT .CTC.TCAGT AGC....... .....AATAC
TFB11438  .TGCTGNAT. ...CTGTAAT .CTC.TCAAT AAC....... .....AATAC
 TFB2887  .TGCTATAT. ...CTGTAAT .CTC.TCAGT AGC....... .....AATAC
TFB11025  .TGCTATAT. ...CTGTAAT .CTC.TCAGT AGC....... .....AATAC
TFB11015  .TGCTGTAT. ...CTGTAAT .CTC.TCAGT AGC....... .....AATAC
TFB11470  .TGCTGTAT. ...CTGTAAT .CTC.TCAGT AGC....... .....AATAC
TFB10095  .TGCTATAT. ...CTGTAAT .CTC.TCAGT AGC....... .....AATAC
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          1051                                                      1100
AF031178  .......... .......... .C..TGTGCT CAATCTGTTT CTATTCATT.
AF356167  .......... .......... .CATTGTGCT CAATCTGTTT CTATTCATT.
AF356169  .......... .......... .......... .......... ..........
LNU33082  .......... .........T TTAGTTTGCT CAATCTGTT. CTACTCATT.
AF356170  .......... .........T TTAGTTTGCT CAATCTGTT. CTATTCATT.
AY016444  .......... .........T TGTTCATGCT CAATCTGTA. C.........
TFB10726  .......... .......... ...GTT.GCT TTATCTG... ..........
TFB11456  .......... .......... ...GTT.GCT TTATCTG... ..........
TFB11014  .......... .......... ...GTTTGCT TTATCTG... ..........
 TFB9605  .......... .......... ...GT..GCT TTATCTGTAA TGGCGG....
 EFM1403  .......... .......... ...GTGAGCT CTATCTG... ..........
TFB10077  .......... .......... ...TAAAGCT CTATCTG... ..........
 REH7007  .......... .......... ...ATTTGCT TTATCTG.AA C.........
  EN2066  .......... .......... ...GTTTGCT TTATCTATCT C.........
 REH7907  .......... .......... ...TTTGGCT TTATTCT... ..........
 TFB9920  .......... .......... ...GTTTGCT TTTTT..... ..........
 REH7348  .......... .......... ...GTT.GCT TTACCTGG.. ..........
 TFB9699  .......... .......... ...GTT.GCT TTACCTGG.. ..........
TFB11026  .......... .......... ...GTT.GCT TTACCTGG.. ..........
 TFB1871  .......... .......... ...GTTTGCT CTACCTGG.. ..........
TFB10350  .......... .......... ...GTT.GCT TTACCTGG.. ..........
TFB10422  .......... .......... .....ATGCT TTACCTGA.. .......TTC
 TFB9657  .......... .........A TTA..ATGCT TTACCTGA.. .......TTC
TFB10080  .......... .......... .....ATGCT TTACCTGT.. .........TT
 TFB7219  .......... .......... .TGGTGTGCT TTACCTGTGT .......CTT
 REH7379  .......... .......... ...GAATGCT TTACCTGGTC .......TTT
TFB10402  .......... .......... ...ATATGCT TTATCTGA.. .......TTC
TFB11035  .......... .......... .TTTAATGCT TTACCTGATA .........TT
 TFB7230  GGGTATGTGG GGCTTT.GCA TA.TTATGCT TTGCCTGT.. .........TT
TFB10416  .......... .......... ..GGAATGCT TTACCTGGTC ......TTTT
 REH8266  .......... .........A TAG.AGTACT TTATCTGTTC .........TT
TFB10338  .......... .......... ...GTTTGCT CTGCCTGG.. .........C
TFB11340  .......... .......... ...ATTTGCT TCGCCTGGT. .........TC
TFB11021  .......... .......... ...GTTTGCT TTATCTG... .......TTC
TFB11013  .......... .......... ...TTTTGCT TTATCTG... .........TC
TFB10493  .......... .......... ...GTTTGCT TTGTCTGTCT .......GGC
TFB11439  .......... .......... ...GTTTGCT TTACTTGGG. .........TC
TFB11040  TAGTATTG.. .....CAACT ...AGTTGCT CTACTTGAA. ..........
 TFB9697  TAGTATTG.. .....CGACT T.AGTTTGCT CTACTTGAG. ..........
TFB11434  TAGTACT... .....GCAAC T..AGTTGCT TTGCTTGGG. ..........
TFB11329  TGGTTT..GG GCATTGCAAT TAAATTTGCT TTACCTGTAA ..........
 REH8290  TGGTCT..GG GTATTGCAAT TAAATTTGCT TTGCCTGTA. ..........
AF079580  TGGTCT..GG GTATTGCAAT TAAACTTGCT TTGCCTGTA. ..........
TFB11438  TGGTTT..GG GCATTGCAAT AAAAATTGCT TTACCTGXXX XX.....GAA
 TFB2887  TGGTTT..TG GCATTGCAAT TAAGTTTGCT TGGCCTGT.. .........A
TFB11025  TGGTTT..TG GCATTGCAAT TAAGTTTGCT TGGCCTGT.. .........A
TFB11015  TGGTTT..TG GCATTGCAAT TAAGTTTGCT TTACCTGTA. ..........
TFB11470  TGGTTT..TG GCATTGCAAT TAATTTTGCT TTACCTGTTA .........A
TFB10095  TGGTTT..TG GCATTGCAAT T..TTTTGCT TTACCTGTA. ..........
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          1101                                                    1150
AF031178  ..AGAGTTTG ATACAAAGGG GA........ ....GT.CAT CT..GCTTTC
AF356167  ..AGAGTTTA ATACAAAGGG GA........ ....GT.CAT CC..GCTTTC
AF356169  .......... .......... .......... .......... ..........
LNU33082  ..GGAG.... ..AAAAAGGG AA........ ....GG...T CT..GCTTTC
AF356170  ..GGAG.... ..AAAAAGGG AA........ ....GT...T CC..GCTTTC
AY016444  .......... .....AAGGG AA........ ....G....T CT..GCTTTC
TFB10726  .......... ATTGT..GGG AAATTTAT.. .......... CT..GCTTTC
TFB11456  .......... ATTGT..GGG AAATTTAT.. .......... CT..GCTTTC
TFB11014  .......... AATGT..GGG AAATTTAT.. .......... CT..GCTTTC
 TFB9605  .......... .TT....... .......... ....GT.... CT..GCTATC
 EFM1403  .......TTT ATTG.AGGAA AAGTATAGTT TT........ ....GCTTCC
TFB10077  .......TTT ATTT.GGAGA AA.TTTTGTG AA..GT.... CT..GCTTTC
 REH7007  .......... ATT..GAGAA AA...TATT. ....GT..AT CT..GCTTTC
  EN2066  .......... ATT....GGG TT........ ....GT..AT CT..GCTTTC
 REH7907  .........G ATAGT..GGG TTG..TATGT AT..GC..GT CT..GCTTTC
 TFB9920  .........G A......... .......... ..CTG..TCT CTTTGC....
 REH7348  TTC..ATTAG AGTTT..GGG A......... ........CAT CT..GCTCTC
 TFB9699  TTC..ATTAG AGTTT..GGG A......... ........CAT CT..GCTCTC
TFB11026  TTC..ATTAG AGTTT..GGG A......... ........CAT CT..GCTCTC
 TFB1871  CTC..ATTAG AGTA...GGG AA........ ........TAT CT..GCTCTC
TFB10350  TTC..ATTAG AGTT...GGG A......... ........CAT CT..GCTCTC
TFB10422  TTC..ATTAG AGT....AGG AA........ .........GT CT..GCTCTC
 TFB9657  TTC..ATTAG AGT....AGG AA........ .........CT CT..GCTTTC
TFB10080  TTC..ACTAG AGT....AGG AA........ .........CT CT..GCTTTC
 TFB7219  TTC..ATTAG AGTTT..AGG AA........ .........CT CT..GCTTTC
 REH7379  TTC..ATTAG AGT....AGG AA........ .........CT CT..GCTTTC
TFB10402  TTT..ATTAG AGT....AGG AA........ .........CT CT..GCTTTC
TFB11035  TTC..ATTAG AGT....AGG AA........ .........CT CT..GCTCTC
 TFB7230  TTC..ATTGG AGT....AGG AA........ .........AT CT..GCTTTC
TFB10416  TTC..ATTAG AGT....AGG AA........ .........CT CT..GCTTTC
 REH8266  TTC..ATTAG AGT....AGG AA........ .........CT CT..GCTTTC
TFB10338  T.C..ACTAG AGTGAAAGGG AAA....... ..TTG....A CC..GCTTTC
TFB11340  .AC.TA.TAG AGTGAAAGGG AAA....... ....GT..GA CC..GCTTTC
TFB11021  T....ACTAG AGTGAAAGGG AAA....... .TTTG...AA CC..GCTCTC
TFB11013  .........G AGTTTAATAG AAGT.....T TTTATT..AA CT..GCTTTT
TFB10493  TGTACAC.TG AGTGTTTAAG AAGTT..... ..TTGTTTGC CT..GCTTTC
TFB11439  TCAGAAGAAA AGAT.....A AA........ ....GTT.CT ....GCTTTC
TFB11040  .TCTCAGAGG AAGAA..GGG AA........ ....GCT.CT ....GCTCTC
 TFB9697  .TCTCAGAGG AAGA...GGG AA........ ....GTT... ..TTGCTTTC
TFB11434  .CCTCAGAGG AAAA...GGG AA........ ....GTT... ..TTGCTTTG
TFB11329  GTCTCAAAAG AAAA...GAA AT........ ....GTT... CT..GCTTTC
 REH8290  GTCTCAGAGG AAAA...GAA AA........ ....GTT... CT..GCTTTC
AF079580  GTCTCAGAGG AAAA...GAA AA........ ....GTT... CT..GCTTTC
TFB11438  GTCTCAGAGG AAAA...GAA AA........ ....GTT... CT..GCTTTC
 TFB2887  GCCTCAGAGG AAAA...GGG AA........ ....GTT... CT..GCTTTC
TFB11025  GCCTCAGAGG AAAA...GGG AA........ ....GTT... CT..GCTTTC
TFB11015  GTCTCAGAGG AAAAA..GGG AA........ ....GTT... CT..GCTTTC
TFB11470  NTCTCAGAGG AAAA...GGG AA........ ....G...AA CT..GCTTTC
TFB10095  GTCTCAGAGG AAAA...GAA AA........ .....TT.CT CT..GCTCTC
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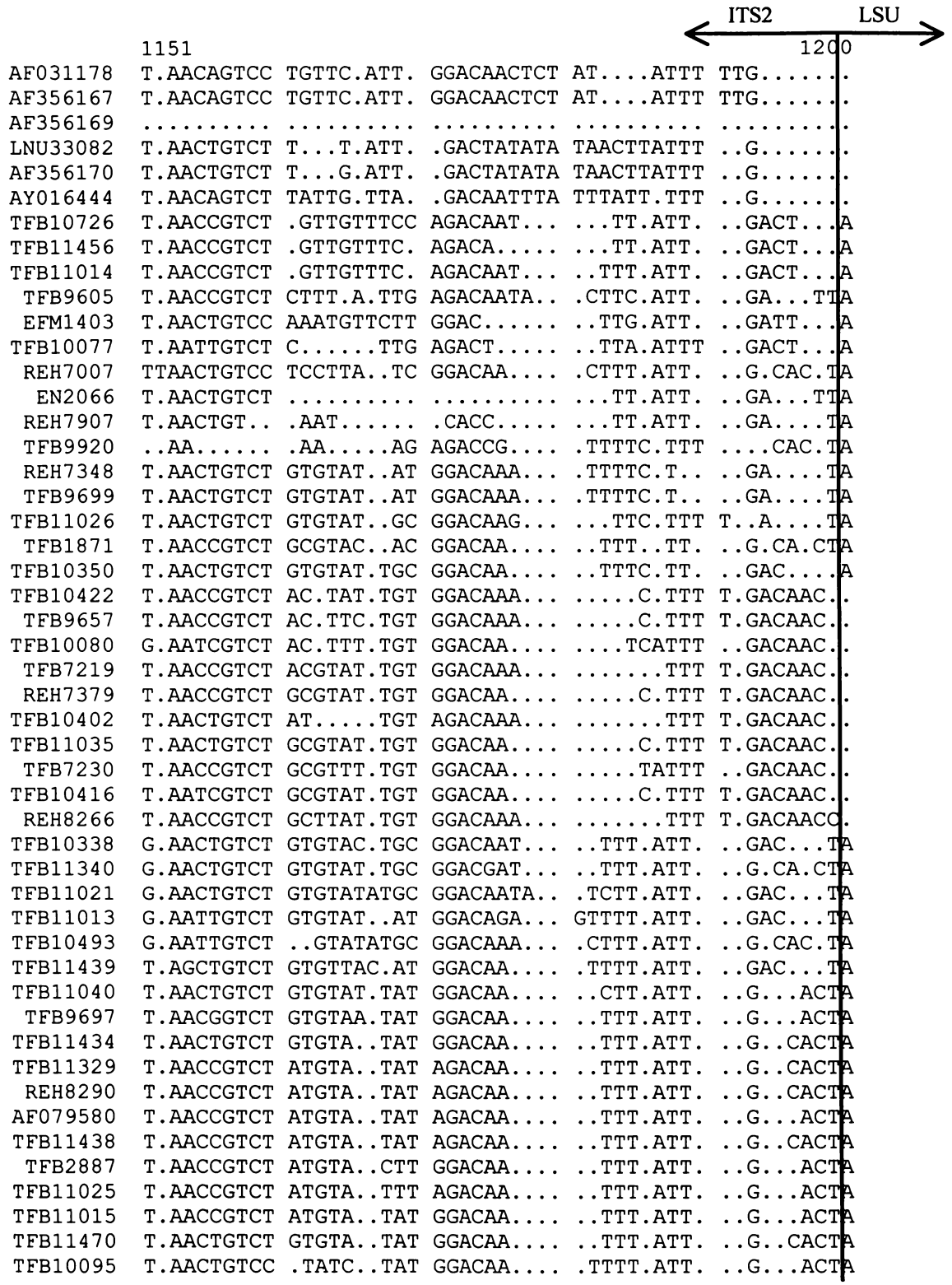
ITS2
LSU
1151
1200
AF031178 T.AACAGTCC TGTTC.ATT. GGACAACTCT AT....ATTT TTG.....
AF356167 T.AACAGTCC TGTTC.ATT. GGACAACTCT AT....ATTT TTG.....
AF356169 ....
LNU33082 T.AACTGTCT T...T.ATT. .GACTATATA TAACTTATTT ..G.....
AF356170 T.AACTGTCT T...G.ATT. .GACTATATA TAACTTATTT ..G.....
AY016444 T.AACAGTCT TATTG.TTA. .GACAATTTA TTTATT.TTT ..G.....
TFB10726 T.AACCGTCT .GTTGTTTCC AGACAAT... ...TT.ATT. ..GACT...A
TFB11456 T.AACCGTCT .GTTGTTTC. AGACA..... ...TT.ATT. ..GACT...A
TFB11014 T.AACCGTCT .GTTGTTTC. AGACAAT... ..TTT.ATT. ..GACT...A
TFB9605 T.AACCGTCT CTTT.A.TTG AGACAATA.. .CTTC.ATT. ..GA...TTA
EFM1403 T.AACTGTCC AAATGTTCTT GGAC..... ..TTG.ATT. ..GATT...A
TFB10077 T.AATTGTCT C.....TTG AGACT..... ..TTA.ATTT ..GACT...A
REH7007 TTAACTGTCC TCCTTA..TC GGACAA.... .CTTT.ATT. ..G.CAC.TA
EN2066 T.AACTGTCT ....TT.ATT. ..GA...TTA
REH7907 T.AACTGT.. .AAT..... .CACC..... ...TT.ATT. ..GA....TA
TFB9920 ..AA..... .AA.....AG AGACCG.... .TTTTC.TTT ....CAC.TA
REH7348 T.AACTGTCT GTGTAT..AT GGACAAA... .TTTTC.T.. ..GA....TA
TFB9699 T.AACTGTCT GTGTAT..AT GGACAAA... .TTTTC.T.. ..GA....TA
TFB11026 T.AACTGTCT GTGTAT..GC GGACAAG... ...TTC.TTT T..A....TA
TFB1871 T.AACCGTCT GCGTAC..AC GGACAA.... ..TTT..TT. ..G.CA.CTA
TFB10350 T.AACTGTCT GTGTAT.TGC GGACAA.... ..TTTC.TT. ..GAC....A
TFB10422 T.AACCGTCT AC.TAT.TGT GGACAAA... .....C.TTT T.GACAAC..
TFB9657 T.AACCGTCT AC.TTC.TGT GGACAAA... .....C.TTT T.GACAAC..
TFB10080 G.AATCGTCT AC.TTT.TGT GGACAA.... ....TCATTT ..GACAAC..
TFB7219 T.AACCGTCT ACGTAT.TGT GGACAAA... ....TTT T.GACAAC..
REH7379 T.AACCGTCT GCGTAT.TGT GGACAA.... .....C.TTT T.GACAAC..
TFB10402 T.AACTGTCT AT.....TGT AGACAAA... ....TTT T.GACAAC..
TFB11035 T.AACTGTCT GCGTAT.TGT GGACAA.... .....C.TTT T.GACAAC..
TFB7230 T.AACCGTCT GCGTTT.TGT GGACAA.... .....TATTT ..GACAAC..
TFB10416 T.AATCGTCT GCGTAT.TGT GGACAA.... .....C.TTT T.GACAAC..
REH8266 T.AACCGTCT GCTTAT.TGT GGACAAA... ....TTT T.GACAACC.
TFB10338 G.AACTGTCT GTGTAC.TGC GGACAAT... ..TTT.ATT. ..GAC...TA
TFB11340 G.AACTGTCT GTGTAT.TGC GGACGAT... ..TTT.ATT. ..G.CA.CTA
TFB11021 G.AACTGTCT GTGTATATGC GGACAATA.. .TCTT.ATT. ..GAC...TA
TFB11013 G.AATTGTCT GTGTAT..AT GGACAGA... GTTTT.ATT. ..GAC...TA
TFB10493 G.AATTGTCT ..GTATATGC GGACAAA... .CTTT.ATT. ..G.CAC.TA
TFB11439 T.AGCTGTCT GTGTTAC.AT GGACAA.... .TTTT.ATT. ..GAC...TA
TFB11040 T.AACTGTCT GTGTAT.TAT GGACAA.... ..CTT.ATT. ..G...ACTA
TFB9697 T.AACGGTCT GTGTAA.TAT GGACAA.... ..TTT.ATT. ..G...ACTA
TFB11434 T.AACTGTCT GTGTA..TAT GGACAA.... ..TTT.ATT. ..G..CACTA
TFB11329 T.AACCGTCT ATGTA..TAT AGACAA.... ..TTT.ATT. ..G..CACTA
REH8290 T.AACCGTCT ATGTA..TAT AGACAA.... ..TTT.ATT. ..G..CACTA
AF079580 T.AACCGTCT ATGTA..TAT AGACAA.... ..TTT.ATT. ..G...ACTA
TFB11438 T.AACCGTCT ATGTA..TAT AGACAA.... ..TTT.ATT. ..G..CACTA
TFB2887 T.AACCGTCT ATGTA..CTT GGACAA.... ..TTT.ATT. ..G...ACTA
TFB11025 T.AACCGTCT ATGTA..TTT AGACAA.... ..TTT.ATT. ..G...ACTA
TFB11015 T.AACCGTCT ATGTA..TAT GGACAA.... ..TTT.ATT. ..G...ACTA
TFB11470 T.AACTGTCT GTGTA..TAT GGACAA.... ..TTT.ATT. ..G..CACTA
TFB10095 T.AACTGTCC .TATC..TAT GGACAA.... .TTTT.ATT. ..G...ACTA

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          1201                                1232
AF031178  CTT.GA.CCT CAA.AT...C A......... ..
AF356167  CTT.GA.CCT CAA.AT...C A......... ..
AF356169  .......... .......... .......... ..
LNU33082  CTT.GA.CCT CAA.AT...C A......... ..
AF356170  CTT.GA.CCT CAA.AT...C A......... ..
AY016444  GTT.GA.CCT CAA.AT...C A......... ..
TFB10726  TTT.GA.CCT CCA.AT...C AGGTAGGA.C TA
TFB11456  TTT.GA.CCT CAA.AT...G AGGTAGGA.C TA
TFB11014  TTT.GA.CCT CAA.AT...C AGGTAGGA.C TA
 TFB9605  TTT.GA.CCT CAA.AT...C AGGTAGGA.C TA
 EFM1403  TT..GA.CCT CAA.AT...C AGGTAGGA.T TA
TFB10077  TTT.GA.CCT CAA.AT...C AGGTAGGA.C TA
 REH7007  TTT.GA.CCT CAA.CAT..C AGGRAGGAAA TA
  EN2066  TT..GA.CCT CAAAN....C AGGTAGGA.C TA
 REH7907  ATTTGA.CCT CAA.AT...C AGGTAGGAAC TA
 TFB9920  TTTTGA.CCT CAA.CT..GC AGGTAGGAAS TA
 REH7348  TTT.GA.CCT CAA.AN...C AGGTAGGA.C TA
 TFB9699  TTT.GA.CCT CAA.AT...C AGGTAGGA.C TA
TFB11026  TTT.GA.CCT CAA.AT...C AGGTAGGA.C TA
 TFB1871  TTATGNACCT CGN.ACNTGC AGGTAGGAAG TA
TFB10350  TT.TGA.CCT CAA.AT...C AGGTAGGA.C TA
TFB10422  .TT.GA.CCT CAA.CT..GC AGGTAGGAAC TA
 TFB9657  .TT.GA.CCT CAA.AT...C AGGTAGGA.C TA
TFB10080  .TT.GA.CCT CAA.CNT..C AGGTAGGA.. ..
 TFB7219  .TT.GA.CCT CAA.A....C AGGTAGGA.C TA
 REH7379  .TT.GA.CCT CAA.CWT..C AGGWAGGA.C T.
TFB10402  .TT.GA.CCT CAA.AT...C AGGTAGGAAC TA
TFB11035  .TT.GA.CCT CAA.AN...C AGGTAGGA.C TA
 TFB7230  .TT.GA.CCT CAA.AT...C AGGTAGGA.C TA
TFB10416  .TT.GRACCT CAA.AT..GC AGGTAGGAAS TA
 REH8266  TTT.GA.CCT CAA.AT...C AGGTAGGA.C TA
TFB10338  TTT.GA.CCT CAA.AT...C AGGTAGGA.C TA
TFB11340  TTT.GA.CCT CAA.AT...C AGGTAGGAAC TA
TFB11021  TTT.GA.CCT CAA.AT...C AGGTAGGA.C TA
TFB11013  TT..GA.CCT CAA.AT...C AGGTAGGAAC TA
TFB10493  TTT.GNACCT CAACA...GC AGGTAGGAAN TA
TFB11439  TTT.GA.CCT CAA.AT...C AGGTAGGA.C TA
TFB11040  TTT.GA.CCT CAA.AT...C AGGTAGGA.C TA
 TFB9697  TTT.GA.CCT CAA.AT...C AGGTAGGAAC TA
TFB11434  TTT.GA.CCT CAA.AT..GC AGGTAGGAAG TA
TFB11329  TTT.GA.CCT CAA.AT..GC AGGTAGGAAC TA
 REH8290  TTATGYACCT CAACA...GC AGGTAGGAAG TA
AF079580  TTT.GA.CCT CAA.AT...C AGGTAGGA.C TA
TFB11438  TT..GNACCT CAACN....C AGGTAGGAAC TA
 TFB2887  TTT.GA.CCT CAA.AT...C AGGTAGGAAC TA
TFB11025  TTT.GA.CCT CAA.AT...C AGGTAGGA.C TA
TFB11015  TTT.GA.CCT CAA.AT...C AGGTAGGA.C TA
TFB11470  TTT.GCACCT CAA.AT..GC AGGTAGGAAG TA
TFB10095  TTT.GA.CCT CAA.AT...C AGGTAGGA.C TA
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VITA

Juan Luis Mata completed his elementary education in 1972 at the Colegio Alemán de Guatemala. In 1977 he obtained his high school diploma from the Humboldt Schule of Costa Rica. During his high school years he also attended Thompson Junior High in Seattle, WA, and Bad Bramstedt Realschule in Germany. In 1984, he obtained a Bachelor's Degree in Biology with a major in botany from the Universidad de Costa Rica. For five years, until 1987, he was a science teacher at a bilingual high school in Costa Rica and after that he worked as a free-lance naturalist tourist guide. In 1994 he was accepted in the Biology Master's program at the Universidad de Costa Rica. The title of his thesis was "Estudio taxonómico y ecológico de los hongos Agaricales en el Bosque del Niño, Reserva Forestal de Grecia". After graduating in 1997, he went to the University of Tennessee in Knoxville, TN, to expand his academic career in mycology.

Juan Luis was born in Boston, Mass. and is the second child to Lic. Grace Eugenia Greenwood and Dr. Leonardo de Jesús Mata. He is married to Ingrid Arce and has two sons, Luis Diego and Juan Carlos.

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