TOWARD A REVISION OF THE GENUS *PHYLLOPORUS* (BOLETACEAE):

SYSTEMATICS AND PHYLOGENY OF SPECIES FROM VARIOUS PARTS OF THE WORLD

by

MARIA-ALICE NEVES

A dissertation submitted to the Graduate Faculty in Biology in partial fulfillment of the requirements for the degree of Doctor of Philosophy, The City University of New York

2007

This manuscript has been read and accepted for the Graduate Faculty in Biology in satisfaction of the dissertation requirement for the degree of Doctor of Philosophy.

20th August, 2007

Dr. Roy E. Halling
The New York Botanical Garden
Chair of Examining Committee

20th August, 2007

Dr. Richard L. Chappell
Executive Officer

Dr. Joseph Rachlin
The City University of New York

Dr. Dominick Basile
The City University of New York

Dr. Timothy J. Baroni
State University College at Cortland

Dr. David S. Hibbett
Clark University

Dr. Wm. Wayt Thomas
The New York Botanical Garden

Supervisory Committee

The City University of New York

Abstract

TOWARD A REVISION OF THE GENUS *PHYLLOPORUS* (BOLETACEAE):

SYSTEMATICS AND PHYLOGENY OF SPECIES FROM VARIOUS PARTS OF THE WORLD

by

MARIA-ALICE NEVES

Advisor: Dr. Roy E. Halling

The Boletaceae are composed of approximately 26 genera and 415 species (Singer 1986). The family is monophyletic and its species usually have large fleshy basidiomata with a central stipe. The spores are mostly elongate and are produced in tubular or rarely lamellate hymenophores. Around 90% of the members of this family form ectomycorrhizae with several genera of trees in most forest ecosystems. Like other fungi, they are also a very important component of the biogeochemical cycle of the forests where they grow. Some species are economically important due to their high value as a food, for example, *Boletus edulis*, known as porcini mushroom or cèpe.

Phylloporus is somewhat of a morphological oddity in the Boletaceae because its species produce a lamellate rather than tubular hymenophores although other basidioma characters, spore morphology, and chemical and molecular data support the placement of this genus in the Boletaceae. The results of Binder (1999) suggest that *Phylloporus* is the sister group to the *Xerocomus subtomentosus* group, species of which produce tubular hymenophores. However, despite several broad-scale phylogenetic studies of the Boletaceae, the phylogenetic relationships of *Phylloporus* remain unclear. Previous phylogenies of this group include only two species of *Phylloporus*, one native to Europe and the other to North America. While the majority of *Phylloporus* species have a tropical distribution, no phylogenetic study of this group has included tropical species.

This work treats twenty-six species from various parts of the world, including 19 tropical taxa. Seven of the species in this work are new to science and two taxa are unnamed. The nomenclature regarding the *Phylloporus rhodoxanthus* complex is clarified. The phylogenetic analysis here presented includes the largest selection of *Phylloporus* species in a phylogenetic study to-date. Phylogenetic relationships of selected species of *Phylloporus*, *Xerocomus*, and *Aureoboletus* were estimated using maximum parsimony, maximum likelihood, and Bayesian methods. Those algorithms analyzed sequences of the partial nuclear large subunit ribosomal DNA, the internal transcribed spacer region, and a combined dataset with both genes. *Aureoboletus auriporus* was used as the outgroup for the LSU and the combined dataset. The results suggest that *Phylloporus* is a monophyletic genus that is separate from the genus *Xerocomus*.

To my parents, Vitória and Amilcar,

my sister Lu, my brother Amilcar

and my nephew Caio.

Acknowledgements

I sincerely thank Dr. Roy Halling for agreeing to be my advisor, for his guidance, encouragement, and support throughout these four years. I also express my gratitude to the other members of the committee: I am very grateful to Dr. David Hibbett, who received me in his laboratory at Clark University and guided me and made valuable suggestions to my work. Thanks to Tim Baroni for his suggestions and advice, especially during the field trip to Thailand. Dr. Joseph Rachlin gave great contributions to the project during the second exam. Dr. Dominick Basile was an inspiration during the SEM classes and his enthusiasm and suggestions are greatly appreciated. Thanks to Dr. Wayt Thomas for the valuable discussions during the development of this work and for helping me be reabsorbed as a scientist in Brazil.

Manfred Binder was an exceptional professor, colleague, and friend at the Hibbett laboratory in Worcester, and I would like to express my gratitude to him. Mike Baxter helped with all the SEM preparation at the SEM laboratory at Lehman College.

I am very grateful to Todd Osmundson, for his friendship, the great company in various trips, and all his inputs and thoughts. To Nathan Smith for the love, patience, and all the time spent editing this work. To Aleja Vasco, my Colombian little sister, for her friendship, company, and help in so many aspects that it is impossible to list them. To Fabián Michelangeli for the valuable comments and suggestions and Larry Kelly for the help in different moments of my grad student life in New York.

For the friendship and company in different phases of my New Yorker life: Doug Daly, Amy Keller, Michael Sundue, Ya-Yi Huang, Tanja Schüster, Olga Marder, Don Wheeler, João Condack, Vanessa Rivera, and Sara Branco (in Chicago).

Financial support from the following institutions and foundations has enabled me to conduct my academic work, laboratory work, herbarium visits, and collecting trips: Coordenação de Aperfeiçoamento de Pessoal de Nível Superior (CAPES, Brazil), The New York Botanical Garden (the Institute of Systematic Botany and the Cullman Program for Molecular Systematics Studies), Lehman College of CUNY, and Friends of the Farlow fellowship. A National Geographic grant and a National Science Foundation grant awarded to Dr. Roy Halling that supported my field trips to Costa Rica and Thailand, respectively. A National Science Foundation grant awarded to Dr. David Hibbett to do the molecular work at Clark University. A National Science Foundation grand awarded to Dennis Desjardin that allowed us to go collect in Thailand.

I thank the curators and individuals that made collections available for my studies: Drs. Dennis Desjardin (SFSU), Don Pfister (FH), Egon Horak, Gastón Guzmán (XAL), Gregory Mueller (F), Juan L. Mata, Laura Guzmán, Marc Ducousso, Terry Henkel.

My sincere thanks to all the colleagues and friends at the New York Botanical Garden.

Finally, I thank some colleagues in Brasil that have been important in the course of my mycological life in Brasil: Dr. Clarice Leite (UFSC), Dr. Marina Capelari (IBot), and Dr. Aristóteles Góes Neto (UEFS).

Table of Contents

List of Tables

List of Illustrations

CHAPTER 1: INTRODUCTION AND LITERATURE REVIEW

Introduction

The Boletaceae sensu Singer (1986) (Fungi: Basidiomycetes) are composed of approximately 26 genera and 415 species of conspicuous stipitate-pileate or gastroid mushrooms that typically produce tubular hymenophores (Kirk et al. 2001). However, a recent molecular phylogeny (Binder & Hibbett 2006) suggests a different family concept that includes 38 genera.

The species in the Boletaceae are represented in most forest ecosystems because they form ectomycorrhizae with several host tree species. This obligate fungus-plant symbiosis has an important role in these forests because it is involved in processes such as nutrient cycling, nutrient uptake, and decomposition of organic matter. Roughly 90% of the species of the boletes are potentially ectomycorrhizal and they may represent 18% to 25% of all ectomycorrhizal fungi, according to previous surveys (Agerer 1987-1998, Hawksworth et al. 1995). Some of the plant families that have ectomycorrhizal relationships with boletes include Betulaceae, Ericaceae, Fabaceae (Caesalpinoideae, Mimosoideae), Fagaceae, Myrtaceae, Pinaceae, and Salicaceae.

Several species are also important economically due to their high value as food, like *Boletus edulis* and some related species, popularly known as porcini mushrooms or cèpe.

The species in the Boletaceae (Order Boletales sensu Jülich, 1981; Order Agaricales, Suborder Boletineae sensu Singer, 1986; and Binder & Hibbett [2006]) are conspicuous mushrooms, that normally produce large fleshy basidiomata with a central stipe. The spore print color can be olivaceous, brownish, yellowish, or vinaceous; the spores are mostly elongate or fusiform. In many species, the putrescent flesh turns blue in some parts when injured or exposed. This bluing is the result of the oxidation of pulvinic acid derivatives, like variegatic, xerocomic, and atrotomentinic acid, which are sometimes produced either by hyphae in the basidiomata or mycelia in culture (Hellwig 1999, Singer 1986). The basidiomata produce tubular, or more rarely lamellar, hymenophores with divergent or bilateral trama, and the basidiospores are pigmented. In combination, these morphological characteristics support this family as a monophyletic group. The monophyly of the family has also been tested using the partial nuclear large subunit ribosomal DNA sequences (nuc-lsu rDNA), and these molecular studies support the phylogenetic conclusions based on morphological data (Binder 1999, Bresinsky & Besl 2003). One conspicuous morphological exception is found in *Phylloporus* Quélet, a genus in which the hymenophore has apparently become secondarily lamellate or subporoid (Besl & Bresinsky 1997), and the systematics of that genus is the focus of this dissertation.

Taxonomic History of the Boletaceae and *Phylloporus*

The name Boletaceae was proposed for the first time by Chevallier in 1826 as a family separate from the Agaricaceae. Boletaceae are distinct from Agaricaceae because they have a tubular hymenophore, which in many cases can be separated from the context, and a firmer consistency of the flesh. Chevallier included five genera in the family: *Polyporus, Cladosporus, Physisporus, Boletus,* and *Fistulina*. At this time *Phylloporus* was considered a member of the Agaricaceae under the name *Agaricus pelletieri* Léveillé because it had a lamellate hymenophore. It was later recognized by

Quélet (1888) as part of the Boletaceae because its overall morphology suggested a close relationship to other species in that group.

Based on the same basic characteristics, Singer (1986) kept the boletes and its relatives in the suborder Boletineae of the order Agaricales, dividing the suborder into three families: Paxillaceae, Gomphidiaceae, and Boletaceae. Singer's concept of Paxillaceae includes lamellate taxa with white, yellowish white, or brown spore prints, and Gomphidiaceae includes lamellate taxa with deep gray, deep olive-gray, or blackish spore prints. The Boletaceae are divided into six subfamilies, one of which (Xerocomoideae) includes *Phylloporus*. Singer also includes *Tubosaeta* Horak and *Xerocomus* Quélet in this subfamily with taxa that have brown, olive-brown, olivaceous, or ferrugineous spore prints, and bilateral hymenophoral trama. These three genera are distinguished by (1) the hymenophore, which is lamellate in *Phylloporus* and tubular in *Xerocomus* and *Tubosaeta* (Fig. 1-1), and (2) the cystidia, which are fusiform in *Phylloporus* and *Xerocomus* and setoid in *Tubosaeta* (Singer 1945, 1986).

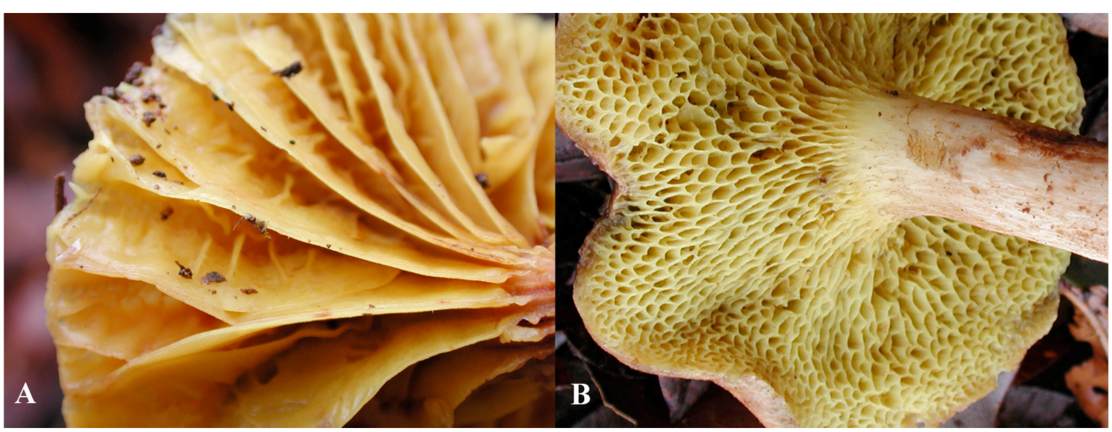

Figure 1-1: Hymenophore morphology in Xerocomoideae (sensu Singer): **A.** *Phylloporus phaeoxanthus*. **B.** *Xerocomus* sp.

The classification proposed by Jülich (1981) maintains a broader concept of the family Boletaceae including sequestrate taxa and the xerocomoid taxa that Singer placed in different subfamilies. Characteristics that unite Jülich's Boletaceae are the presence of yellow-brown, brown, or olivaceous brown spore prints; broadly ellipsoid to elongate, smooth or verrucose basidiospores; and the absence or rarity of clamp connections.

A new family, the Xerocomaceae, including Singer's subfamily Xerocomoideae (Singer 1945) was proposed by Pegler & Young (1981). That family was composed of eight genera that form a closely related group (*Boletellus* Murr., *Gymnopaxillus* E. Horak, *Paxillogaster* E. Horak, *Phylloboletellus* Singer, *Phylloporus* Quél., *Singeromyces* M. M. Moser, *Tubosaeta* E. Horak, and *Xerocomus* Quél.). Within this group the spore ornamentation is variable, from coarsely ornamented *Boletellus* spores to smooth or weakly ornamented *Xerocomus* and *Phylloporus* spores. Pegler & Young (1981) thought *Phylloporus* was closely related to *Xerocomus* and considered smooth spores more advanced than ornamented ones. They also suggested that the hymenophore configuration should be regarded as secondarily lamellate and derived from a tubular condition.

Høiland (1987) suggested a tentative system based on morphological and chemical characters of the Boletales that included seven families: Paxillaceae, Coniophoraceae, Chamonixiaceae, Boletaceae, Rhizopogonaceae, Gomphidiaceae, and Strobilomycetaceae. Høiland's analysis did not support the distinction of Xerocomaceae and Boletaceae as two separate families.

Phylogenetic works based on molecular data (Binder 1999, Binder & Hibbett 2006), including *P. rhodoxanthus* and *P. pelletieri,* showed *Phylloporus* to be nested within

Xerocomus sensu stricto. Binder (1999) maintained them as two independent genera but noted the sister group relationship of *Phylloporus* with the *Xerocomus subtomentosus* complex (*X. illudens, X. lanatus,* and *X. subtomentosus*) based on molecular and morphological characters (Fig. 1-2). Similar results were seen when four collections of *Phylloporus* were included in the phylogeny (Binder & Hibbett 2006, Fig. 1-3). The two genera are distinguished by the lamellate hymenophore produced by *Phylloporus* in contrast to the wide tubulate hymenophore seen in *Xerocomus*.

Based solely on DNA sequences of two *Phylloporus* species, *P. pelletieri* from Europe and *P. rhodoxanthus* from North America, Bresinsky & Besl (2003) reduced the genus to synonymy with *Xerocomus* and suggested that the non-European species, with the exception of the North American *P. rhodoxanthus*, should be placed in a new genus. No new name was suggested and no tropical taxa were included in the analyses. Nevertheless, this synonymy exacerbates the systematic problems of the genus especially since *Xerocomus* is not considered a monophyletic genus (Binder 1999, Binder & Hibbett 2006). Until recently, taxonomic delimitation of *Phylloporus* has been controversial and remained insufficiently resolved (Binder 1999, Bresinsky & Besl 2003).

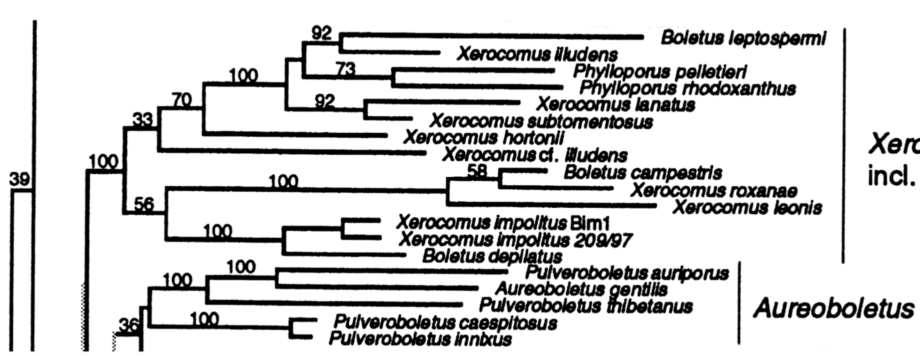

Figure 1-2: Part of the tree showing the relationships among the genera *Aureoboletus*, *Phylloporus*, *Pulveroboletus*, and *Xerocomus* based on LSU (Binder 1999).

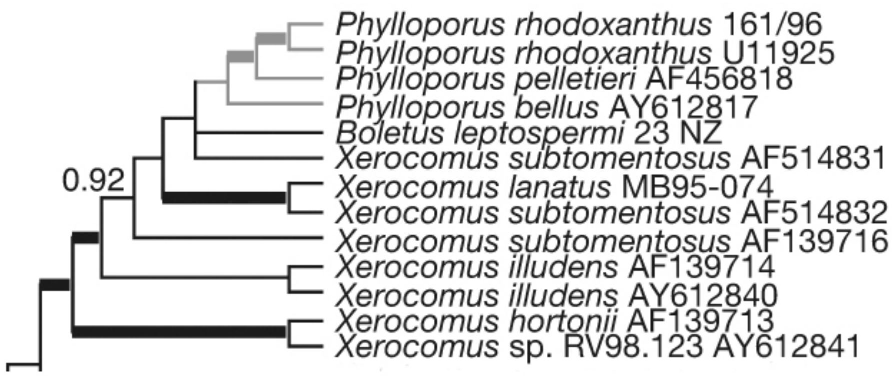

Figure 1-3: Part of the tree showing the relationships among *Phylloporus*, *Boletus*, and *Xerocomus* based on LSU (Binder & Hibbett 2006).

The genus *Phylloporus* was created by Quélet in 1888 to include *Agaricus pelletieri* Léveillé, which is from Europe. There are now 70 species described for the genus and most of these species are native to the tropics (Table 1-1).

The first work that mentioned dividing *Phylloporus* into sections was by Singer (1978), in which he erected section *Manausenses* based on *Phylloporus manausenses*, a species from the Amazonian region of Brazil. In this same paper, Singer (1978) mentioned that the type species of the genus should be included in section *Phylloporus*, with the majority of the species in the genus. Singer, in his last edition of the *Agaricales in Modern Taxonomy* (Singer 1986), recognized two sections in *Phylloporus*: section *Phylloporus* and section *Manausenses*. In a paper on *Phylloporus* from Costa Rica, Singer and Gómez (1984) published a key to the sections *Manausenses* and *Phylloporus*, and cited section *Sulphurei*, noting that the latter does not occur in Costa Rica. There was neither a Latin diagnosis nor authorship for section *Sulphurei*. Another section, *Fibulati*, was later erected by Singer et al. (1990) to accommodate *P. fibulatus*, a species with clamp connections. The authors were not aware of Heinemann & Rammeloo's (1987a) work on *Phylloporus* from Africa, where they had published section *Fibulati* to accommodate *P. pseudopaxillus*. In the same work (Heinemann & Rammeloo 1987a), another four sections were published, among them, section *Phylloporus*. The other three sections were named section *Oxydabiles*, section *Immutabiles*, and section *Tubipedes*.

Table 1-1: *Phylloporus* species, their distribution, and possible hosts. Names in boldface are included in the systematics chapter of this dissertation.

SPECIES	DISTRIBUTION	POSSIBLE HOSTS
P. albocarnosus Heinem.	Zambia, Zaire	Caesalpinioideae
***P. alborufus* Neves & Halling**	Costa Rica	*Quercus*
P. ampliporus Heinem. & Rammeloo	Zambia, Zaire	Caesalpinioideae
***P. arenicola* A.H. Sm. & Trappe**	USA	*Pinus*
P. ater (Beeli) Heinem.	Congo	Caesalpinioideae
***P. aurantiacus* Halling & G.M. Muell.**	Costa Rica	*Quercus*
P. australiensis Watling	Australia	*Casuarina*
***P. bellus* (Massee) Corner**	Costa Rica, Indonesia, Japan, Mexico, Singapore	*Quercus, Castanopsis*
***P. bellus* var. *cyanescens* Corner**	Australia, Malaya	*Quercus, Castanopsis*
***P. bogoriensis* Höhn.**	Singapore	*Dipterocarpus*
***P. boletinoides* A.H. Sm. & Thiers**	Belize, United States	*Pinus, Quercus*
P. borneensis Corner	Borneo	Fagaceae
P. brunneolus Corner	Malaya	Fagaceae
***P. caballeroi* Singer**	Argentina, Bolivia, Costa Rica, Panama	*Alnus jorullensis, A. cuminata*
P. carmineus Heinem.	Zaire	Caesalpinioideae
***P. centroamericanus* Singer & L.D. Gómez**	Costa Rica, Mexico	*Quercus, Magnolia, Chusquea*
P. cingulatus Corner	Singapore	*Fagaceae*
P. clelandii Watling	Australia	*Eucalyptus*
P. coccineus Corner	Singapore	*Fagaceae*
P. colligatus Neves & Henkel	Guyana	*Dicymbe*
P. curvatus Neves & Halling	Thailand	*Castanopsis*

Table 1-1: Cont.

SPECIES	DISTRIBUTION	POSSIBLE HOSTS
P. depressus Heinem.	Zaire	Caesalpinioideae
***P. dimorphus* Neves & Halling**	Thailand	*Castanopsis*
***P. fibulatus* Singer, Ovrebo & Halling**	Colombia	*Quercus humboldtii*
P. flavidulus Corner	Borneo	Fagaceae
P. flavipes Rick	Brazil	?
***P. foliiporus* (Murrill) Singer**	Japan, United States	*Pinus, Quercus*
P. gomphidioides Heinem. & Rammeloo	Burundi	Caesalpinioideae
P. guanacastensis L.D. Gómez	Costa Rica	?
***P. guzmanii* Montoya & Band.-Muñoz**	Mexico	*Pinus, Quercus*
P. gymnocystis Singer	Brazil	Leguminoseae
P. hyperion (Cooke & Massee) Singer	Australia	*Casuarina*
P. incarnatus Corner	Singapore	Fagaceae
***P. infuscatus* Neves & Halling**	Thailand	*Castanopsis*
***P. leucomycelinus* Singer**	Australia (?), United States	*Fagus, Quercus, Eucalyptus* (?)
P. luteobasalis Heinem. & Rammeloo	Congo	Caesalpinioideae
P. luxiensis M. Zang	China	?
P. manausensis Singer	Brazil	*Neea,* Euphorbiaceae
P. nigrescens Heinem. & Rammeloo	Zaire	Caesalpinioideae
***P. novae-zelandiae* McNabb**	New Zealand	*Nothofagus*
P. ochraceobrunneus Corner	Malaya	Fagaceae
P. orientalis Corner	Australia, Borneo, Singapore	Myrtaceae, Fagaceae
***P. orientalis* var. *brevisporus* Corner**	Singapore	*Acacia, Casuarina, Allocasuarina (?), Eucalyptus*
P. parvisporus Corner	Singapore	*Fagaceae*
P. pelletieri (Lév.) Quél.	Europe	*Castanea, Fagus, Larix, Picea, Quercus*
P. phaeosporus Corner	Borneo	*Fagaceae*
P. phaeoxanthus Singer & L.D. Gómez	Colombia, Costa Rica, Mexico	*Quercus*

Table 1-1: Cont.

SPECIES	DISTRIBUTION	POSSIBLE HOSTS
P. phaeoxanthus var. *simplex* Singer & L.D. Gómez	Costa Rica	*Quercus*
P. pratensis Rick	Brazil	?
P. pseudopaxillus Heinem. & Rammeloo	Kenya	Caesalpinioideae
***P. pumilus* Neves & Halling**	Indonesia	*Dipterocarpus*
P. purpurellus Singer	Colombia, Costa Rica	*Quercus*
P. purpureus (Beeli) Heinem.	Zaire	Caesalpinioideae
P. purpureus var. *ambiguus* Heinem.	Burundi, Zaire	Caesalpinioideae
P. rhodophaeus Heinem. & Rammeloo	Zaire	Caesalpinioideae
***P. rhodoxanthus* (Schwein.) Bres.**	United States	*Fagus, Quercus*
***P. rubiginosus* Neves & Halling**	Thailand	*Castanopsis, Dipterocarpus*
P. rubriceps Corner	Borneo	Fagaceae
P. rufescens Corner	Singapore	Fagaceae
***P. scabripes* Ortiz-Santana & Neves**	Belize	*Pinus, Quercus*
P. scabrosus M. Zang	China	?
P. squamosus Corner	Malaysia	*Quercus*
P. stenosporus Corner	Malaysia	*Quercus*
P. sulcatus (Pat.) E.-J. Gilbert	Indochina	?
P. testaceus Heinem. & Gooss.-Font.	Zaire	Caesalpinioideae
P. testaceus var. *bisporus* Heinem.	Zaire	Caesalpinioideae
P. tubipes Heinem.	Congo	Caesalpinioideae
P. tunicatus Corner	Malaya	Fagaceae
P. veluticeps (Cooke & Massee) Pegler & T.W.K. Young	Australia	?
P. viridis (Berk.) Singer	Brazil	?
Phylloporus sp.1	Australia	*Acacia*
Phylloporus sp.2	Costa Rica	*Quercus*

Molecular Phylogeny

The monophyly of the Boletales has been demonstrated in phylogenetic studies based on single or multigene analysis (Binder & Hibbett 2006, Hibbett & Thorn 2001). Nuclear large subunit ribosomal DNA sequences have been traditionally used in fungi to create single gene phylogenies and, even though the support for deeper nodes is generally poor, many of the sister group relationships within the order have been clarified using this technique (Hibbett et al. 1997, Hibbett & Thorn 2001). Combined with morphological studies, these phylogenies include a large number of taxa and have allowed scientists to make evolutionary inferences and to compare the results with earlier systematic classifications.

A large clade has now been resolved within the Agaricomycetidae that contains the Agaricales, Atheliales, and Boletales, (Hibbett & Binder 2002). The sister group relationship between Atheliales and Boletales was inferred by Larsson et al. (2004) and is supported in the multigene phylogeny by Binder & Hibbett (2006). Within Boletales, eight major lineages were resolved based on their multigene approach (Binder & Hibbett 2006). Tapinellineae formed a basal clade, agreeing with Kretzer & Bruns (1999), together with Coniophorineae. Serpulaceae, including *Austropaxillus* (Bresinsky et al. 1999), and Hygrophoropsidaceae formed independent groups; the more derived groups were formed by Suillineae, Sclerodermatineae, Paxillineae, and Boletineae (Binder & Hibbett 2006). The results of Binder & Hibbett (2006) do not support the Paxillineae clade and show it as a sister group to Boletineae. For this reason, the authors propose to merge these two suborders to include Paxillaceae (together with Boletaceae) in the Boletineae.

At the family level, Boletaceae are monophyletic (Binder 1999, Bresinsky & Besl 2003, Hibbett & Thorn 2001, Moncalvo et al. 2000). However, the relationships among genera are poorly resolved in the LSU phylogenies (Binder 1999, Binder & Hibbett 2006) and several of the larger genera in the family, widely accepted in the Boletineae sensu Singer (1986), are not supported as monophyletic. *Xerocomus* species in the *subtomentosus* complex are supported in phylogenetic analyses as closely related to *Phylloporus*, but the genus appears to be a polyphyletic group (Binder 1999, Binder & Hibbett 2006). Recent studies on *Xerocomus* using LSU (Peintner et al. 2003) and ITS (Taylor et al. 2006) sequence data confirm that the genus is not monophyletic. Interestingly, no *Phylloporus* species were included in those studies.

Ecology

The ecological requirements of *Phylloporus* have not been well studied. Data collected during field work and from the literature are presented here to elucidate this aspect of their natural history.

By the same token, little is known about the actual ectomycorrhizal relationships of *Phylloporus* species. Tree associates are usually presumed. Table 1-1 shows a list of the species in the genus, their distribution, and their possible host trees.

Most species of *Phylloporus* that occur in the Western Hemisphere are associated with *Quercus* forests, as has been observed for other genera in the Boletaceae (Mueller & Halling 1995). Among the 20 species known from the Western Hemisphere, 13 are associated (not necessarily exclusively) with *Quercus*, mostly in neotropical montane forests. The presence of both temperate and tropical elements in Central American forests suggests co-evolution and adaptive radiation for the neotropical boletes associated with montane tree species, and suggests that symbiosis has driven the migration of these fungi to the neotropics (Mueller & Halling 1995).

Phylloporus arenicola is interesting in this aspect because it is the only known species that grows in sandy soil and is associated exclusively with *Pinus*. *Phylloporus boletinoides* (United States, Belize), *P. foliiporus* (United States), *P. guzmanii* (Mexico), and *P. scabripes* (Belize) are also associated with *Pinus*, but not necessarily in sandy soils, and they all also grow in association with *Quercus*. *Phylloporus caballeroi* grows under *Alnus* in Argentina, Bolivia, and Costa Rica. *Phylloporus colligatus* is associated with Caesalpinioideae (*Dicymbe* species) in Guyana. *Phylloporus manausensis* occurs in Brazil in campinaranas (white sand openings in rain forests). Information about its tree hosts is poorly known, but possible associations are with *Neea*, Arecaceae, or Euphorbiaceae (Singer et al. 1983).

In the Eastern Hemisphere, the African species are associated with Caesalpinioideae trees. Fourteen species of *Phylloporus* have been described for Central Africa, but unfortunately none of these species were available for study. *Phylloporus pelletieri*, in Europe, is associated with a range of different genera in the Fagaceae and Pinaceae including *Castanea*, *Fagus*, *Larix*, *Picea*, and *Quercus*. In Southeast Asia, *P. bogoriensis*, *P. pumilus*, and *P. rubiginosus* are associated with *Dipterocarpus*, and the last species is also associated with *Castanopsis*. Watling & See (1998) cite the Boletaceae, together with Amanitaceae and Russulaceae, as one of the important components of the ectomycorrhizal community in the Dipterocarpaceae forests of the Malaysian Peninsula. In Thailand, *Phylloporus curvatus*, *P. dimorphus*, and *P. infuscatus* were collected in

Castanopsis forests. The only *Phylloporus* species known from New Zealand is associated with *Nothofagus*, suggesting that other species associated with this tree might be found in similar forests in South America (Chile and Argentina) where *Nothofagus* is common. In Australia the seven known species are associated with *Allocasuarina*, *Casuarina*, *Castanopsis* (Fagaceae, not in Australia), or *Eucalyptus*.

According to Ladurner and Simonini (2003), *P. pelletieri*, in Europe, is a generalist in its mycorrhizal symbiosis, with five known host genera in two different families (Fagaceae: *Castanea*, *Fagus,* and *Quercus*; Pinaceae: *Larix* and *Picea*).

Phylloporus bellus appears to be the most cosmopolitan species in the genus, occurring in both Western (Costa Rica, Mexico) and Eastern (Indonesia, Japan, Singapore) Hemispheres. This species is associated with *Quercus* in the Western Hemisphere and with *Castanopsis* in the Eastern Hemisphere, corroborating the observations made by Mueller and Halling (Mueller & Halling 1995). These authors have observed that species of Boletaceae that occur both in and outside of neotropical *Quercus* forests (e.g., *P. bellus*) have distributions that coincide with the distribution of species of Fagaceae in the Western Hemisphere.

Like African *Phylloporus* species, *P. colligatus* from Guyana has been found associated with Caesalpinioideae trees, suggesting the possibility that *P. colligatus* might be related to the African taxa. However, without the observation of African specimens and a deeper morphological and molecular study of the species from these two continents, it will be impossible to confirm this phylogenetic relationship.

Like the majority of boletes, most *Phylloporus* species produce basidiomata in late summer and fall in both Northern and Southern Hemispheres. This corresponds to the rainy season and, in regions where the seasons themselves are not strongly marked, the amount of rain seems to be the determining factor for basidiomata production. *Phylloporus arenicola* has a different pattern and has been collected during the summer, fall, and winter in California (United States). A longer production of basidiomata throughout the year was also reported for some African species, which have been collected in nine out of the 12 months of the year. Further collections in Central and South America, Southeast Asia, and Africa are needed to better understand the phenology of *Phylloporus* species.

Mycogeography

Of the 70 species known in the genus, only one, *P. pelletieri*, is found in Europe, the others occur elsewhere (Fig. 1-3). Explorations covering North America and Costa Rica have shown that these are diversity hot spots for the genus. Fewer collections have been documented from South America (Argentina, Brazil, Colombia) and Mexico (Montoya & Bandala 1991, Neves & Capelari accepted, Singer et al. 1990), but the presence of montane forests with temperate elements at tropical latitudes strongly suggests that these regions are potentially additional hot spots for *Phylloporus*. *Phylloporus* species have been collected and documented from Malaysia (Corner 1970, 1972), Australia (Watling & Gregory 1991), and Central Africa (Heinemann & Rammeloo 1987b).

Most species of *Phylloporus* are distributed in the Southern Hemisphere, with only three species (*P. bellus*, *P. caballeroi*, and *P. leucomycelinus*) known from both Southern and Northern Hemispheres, and 15 species reported exclusively from the Northern Hemisphere.

Of the 15 species in the Northern Hemisphere, *Phylloporus pelletieri* is the only one known from Europe. Five species have been collected in the United States. One single report was found in the literature that cites the presence of *P. rhodoxanthus* in Canada, however this is not verifiable since there is no voucher mentioned (Roberts et al. 2004). *Phylloporus arenicola* is endemic to the West Coast of the United States. *Phylloporus boletinoides* has been collected in the United States and in Belize and probably represents another example of a southward co-migration of ectomycorrhizal fungi with *Quercus* (Halling 2001). There are three species occurring in Central and northern South America that are also associated with *Quercus*. These species appear to be endemic to neotropical *Quercus* forests: *P. centroamericanus* (Costa Rica and Mexico), *P. phaeoxanthus* (Colombia, Costa Rica and Mexico), and *P. purpurellus* (Colombia and Costa Rica).

Thirteen out of the 15 species in the Northern Hemisphere are found in Central America. Six endemic taxa have been found so far and described for Belize (*P. scabripes*), Costa Rica (*P. alborufus*, *P. aurantiacus*, *P. guanacastensis*, *P. phaeoxanthus* var. *simplex*), and Mexico (*P. guzmanii*).

There are 14 species of *Phylloporus* that have been reported from Central Africa. Another seven are in Australia, with four endemic species (*P. australiensis*, *P. clelandii*, *P. hyperion*, and *P. veluticeps*). The only species known from New Zealand, *P. novae-zealandiae*, is endemic to that country (McNabb 1971). Two endemic species have been reported for China (*P. luxiensis* and *P. scabrosus*), four for Thailand (*P. curvatus*, *P. dimorphus*, *P. infuscatus* and *P. rubiginosus*), and five for Malaysia (*P. brunneolus*, *P. ochraceobrunneus*, *P. squamosus*, *P. stenosporus* and *P. tunicatus*).

Ten species are found in South America and, among those, seven are endemic to different parts of the continent: *P. fibulatus* to Colombia, *P. colligatus* to Guyana, and five species to Brazil (*P. flavipes*, *P. gymnocystis*, *P. manausensis*, *P. pratensis*, and *P. viridis*) (Singer et al. 1983).

Among the species with clamp connections present in all tissues, two are associated with Caesalpinioideae trees, *P. colligatus* in Guyana and *P. pseudopaxillus* in Kenya. *Phylloporus fibulatus*, which is associated with *Quercus*, is found in South America (Colombia).

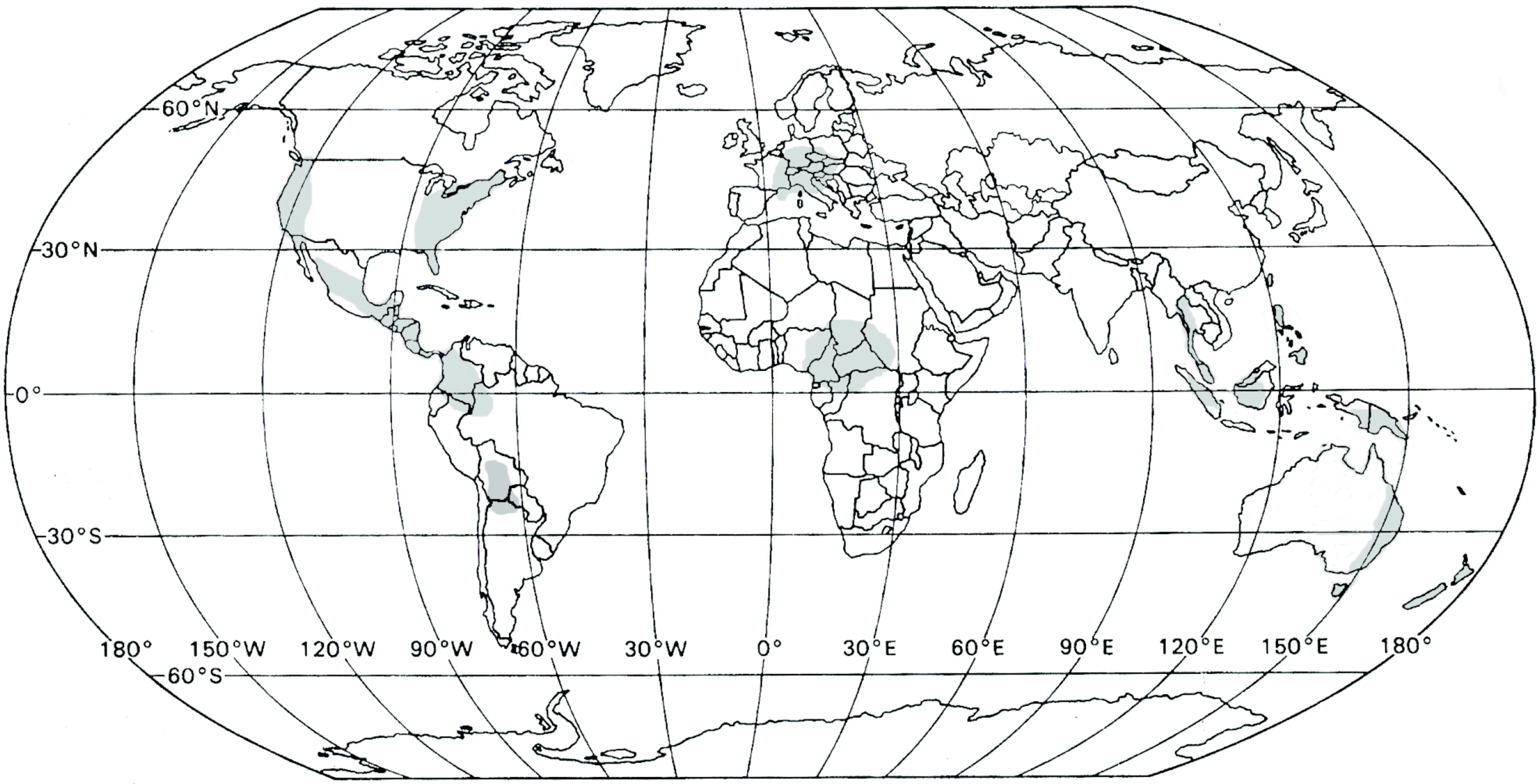

Figure 1-4: World distribution of species of *Phylloporus*.

Morphology

MACROSCOPIC FEATURES

The macroscopic morphology of *Phylloporus* species is variable due to developmental and environmental factors. Some morphological characters, such as basidioma color and size, have reduced taxonomic importance in certain species because they are extremely variable depending on moisture, amount of light, and age of the basidiomata.

Pileus. The size of the pileus in *Phylloporus*, as in other genera in the Boletaceae (Ladurner & Simonini 2003, Smith & Thiers 1964), can vary considerably and it is usually not a characteristic of taxonomic value. However, in cases when the basidiomata are at the extreme range of the diameter of the pileus, it may be used as an informative character (e.g., *P. pumilus*). The shape of the pileus in *Phylloporus* usually varies from convex in young basidiomata to plane or flat and, in some cases, becoming depressed to infundibuliform when mature. The margin of the pileus is in general even, but can be inrolled at the beginning of development in many taxa. The surface of the pileus is consistently dry and even and mostly smooth or finely tomentose. The color is variable and it will change according to the water content, the amount of light during the development of the basidioma, and age. In general, the color of the pileus ranges from yellow ochraceous to dark red or brown, fading to yellowish tones with age or under high moisture conditions.

Hymenophore. Most taxa in *Phylloporus* have a lamellate hymenophore, a characteristic of the genus that separates it from other genera in the family. The lamellae are invariably intervenose, and in some species this is more pronounced. A few species have a more alveolate or highly anastomosed hymenophore either near the stipe or throughout most of the hymenophore. In any case, the lines of the constituent lamellae can still be followed, distinguishing this form from the truly tubulate hymenophores found in other boletes. *Phylloporus boletinoides*, *P. fibulatus*, *P. rhodophaeus*, *P. pumilus*, *P. purpureus* var. *ambiguous*, and *P. testaceus* are taxa that have some degree of anastomosis in their hymenophores.

The attachment of the hymenophore is usually adnexed or decurrent. Sometimes in older specimens the lamellae detach from the stipe and appear free. The lamellae are somewhat thick and are subdistant to distant in spacing, never thin and never closely spaced. Characters of the lamellar edges are of no taxonomic value because they are always smooth or eroded, with erosion possibly due to aging. The color is always yellow, in some species with orange (*P. aurantiacus*) or olivaceous tones (*P. boletinoides*, *P. scabripes*), especially in mature basidiomata. The reaction of the hymenophore to bruising is a taxonomically important character and can be negative, when there is no change in color, or positive when the bruised part stains blue, blue-green, pink, reddish, or brown. Sometimes the blue reaction may take a few seconds to appear and in other cases the reaction will become brownish after a few seconds (e.g., *P. dimorphus*). This reaction also varies with age and the amount of moisture in the basidioma; in general, collections that are old or wet will exhibit a weaker reaction.

Stipe. The length and diameter of the stipe are of little taxonomic importance in *Phylloporus*, except for species that are at the extremes of the size range (e.g., *P. pumilus*). The shape of the stipe can vary from equal to tapering downwards, and is never bulbous as seen in other boletes. It is usually solid, and in some species (*P. boletinoides*, *P. rhodoxanthus*, *P. tubipes*) it sometimes becomes hollow with age. The surface is dry, smooth or subpruinose, rarely subscabrous or scabrous (*P. scabripes*), or in some cases ridged when the hymenophore is decurrent and the lamellae form lines on the top of the stipe (*P. foliiporus*, *P. rhodoxanthus*). The stipe can have the same color as the pileus, an important character in some species such as *P. aurantiacus* and *P. rubiginosus*. In general, the stipe is slightly lighter and has yellow tones compared to the pileus. When the stipe is pruinose or subscabrous, the pruina or the scabers will sometimes have a darker reddish or brownish tone. Usually the color is lighter at the base of the stipe and tends to be yellowish or creamy white.

The color of the basal mycelium can be white or yellow and has a traditional importance in the taxonomy of the genus because it is often used to distinguish species.

The context of the pileus and the stipe is usually white to yellow in the fresh specimens, with the exception of *P. infuscatus*, a species from Thailand in which the context is brown. As in other boletes, the context may oxidize and change color when exposed; a bluing reaction is common in some species and this character is considered taxonomically important at the species level. Odor and flavor tend to be mild in all taxa and have no taxonomic value in the genus.

Macrochemical reactions. The contact of NH_4 or its vapors to the pileus, flesh, or hymenophore of the basidiomata has been studied in Boletaceae (Baroni 1978) and can contribute to the taxonomy of *Phylloporus* if carefully and systematically applied. The reaction consists of a change of color that in general turns the tissues blue or greenish. Other colors observed are tones of purple, reddish, or pink. The reaction on the pileus surface can be blue to blue-green (as seen in most of the species that have a positive reaction), bright green (*P. scabripes*), or purple to reddish (*P. dimorphus* and *P. infuscatus*). Two species included in this work (*P. curvatus* and *Phylloporus* sp. 2) have a distinct pink reaction on the flesh when NH_4 is applied.

The use of Melzer's reagent in mounting slides, or the fleeting amyloid reaction (see chapter 2), gives a bluing reaction that was observed in all the species studied in this work. It is therefore not a characteristic with taxonomic value and the differences among the species tested are only in subjective terms related to the intensity of the blue color of the reaction.

MICROSCOPIC FEATURES

Basidiospores. *Phylloporus* species in general produce a brown olivaceous spore deposit – a characteristic of the genus that, combined with the lamellate hymenophore, separates it from other genera in the family. Most taxa have boletoid spores that are subfusoid to fusoid, but a few species have ovoid spores (*P. aurantiacus*, *P. guzmanii*, and *P. infuscatus*), and one species shows both morphologies (*P. dimorphus*). The spores in most species are inamyloid under Melzer's reagent, but some species have dextrinoid or amyloid reactions. Although a good tool for taxonomy, some problems might arise when using Melzer's reaction. In some cases only a few spores appear dextrinoid, and sometimes it is clear only when they are seen in mass but not individually. The reaction is also probably related to the viability of the spore or the age of the basidioma. Amyloidity has been observed in *P. alborufus*, *P. foliporus*, *P. infuscatus*, and *P. orientalis* var. *brevisporus*. Unfortunately, other authors like Corner (Corner 1970) and Heinemann & Rammeloo (Heinemann & Rammeloo 1987b) did not use Melzer's reagent in their observations and therefore it is not known if the species described in their work have this reaction.

All *Phylloporus* spores appear smooth under the light microscope and the majority of the spores are smooth when observed under the scanning electron microscope. *Phylloporus rhodoxanthus*, *P. testaceus*, *P. rhodopheus*, and *P. gomphidioides* have spores with a bacilate surface, theserepresent the most ornamented spores in the genus. The spore surface in both *Phylloporus guzmanii* and *P. phaeoxanthus* is finely scrobiculate to rough.

Lamellae. The basidia are not valuable taxonomically in *Phylloporus* species. They are mostly clavate, 4-sterigmate, and range from 17 µm to 23 µm (*P. pumilus*) to 47 µm to 65 µm (*P. guzmanii*) in length. A few 5-sterigmate basidia were observed in *P. colligatus*.

The presence of cystidia in *Phylloporus* is one of the characteristics that distinguishes it from *Paxillus* and *Austropaxillus* (Bresinsky et al. 1999), neither of which have cystidia. The cystidia morphology is quite variable, with the most common being the fusoid type. Some species have versiform cystidia that are variable within the same specimen; this is a diagnostic character, for example, in *P. phaeoxanthus* var. *simplex*. *Phylloporus centroamericanus* and *P. rubiginosus* have thick-walled cystidia, distinguishing these from other species in the genus. The cheilocystidia, in general, are similar to the pleurocystidia, and in many cases can only be distinguished by their position on the lamellae.

Two basic types of hymenophoral trama have been described in Boletales: (1) the bilateral trama (or the *Phylloporus*-type) and (2) the divergent trama (or the *Boletus*-

type). Since the time that these two types of hymenophoral trama were described for boletes by Lohwag & Peringer (1937), there have been arguments about the taxonomic value of this character for the family. Singer (1945) observed diverse species and accepted the terms as good characters. In the same paper, however, he affirmed that the difference between the two types is quantitative, not qualitative. Snell & Dick (1958) have suggested a method to distinguish the two types based on the separation of the tubes of *Boletus* and *Xerocomus* species when the basidoma is broken from the pileus surface towards the hymenophore. This method proved to be inapplicable to *Phylloporus* and results in the assumption (according to the authors that described it) that all species of *Phylloporus* have the *Phylloporus*-type of hymenophoral trama. However, when the trama was observed microscopically this was not always true. Although most species have a bilateral trama, the divergent type was also observed. Some species have a bilateral trama that becomes divergent towards the edge of the lamellae (e.g., *P. curvatus*), or the divergent trama is present in young basidiomata and then develops into a bilateral trama (e.g., *P. alborufus*). Watling & Gregory (1991) described Australian species as having the divergent type of trama (*P. australiensis*, *P. cingulatus*, *Phylloporus* sp. 1) and the same was observed in a few neotropical species (*P. boletinoides*, *P. fibulatus*). Unfortunately, Heinemann and Rammeloo (1987b) and Corner (1970, 1972) did not include this character when they described species from Africa and from Malaysia, respectively.

Ladurner and Simonini (2003) reported that, in species with divergent trama, the hymenophore can always be removed from the pileal context and the round marks of the tube attachments can be seen on the context. They also affirm that this is never the case for species of *Xerocomus*. As seen in *Xerocomus*, the species of *Phylloporus* that I tested for this method did not exhibit the results that Ladurner and Simonini reported for species with divergent trama. Therefore I conclude that the structure of the hymenophoral trama is not a characteristic of taxonomic importance in *Phylloporus*.

Pileus. The pileus surface (pileipellis) is mostly constituted of cylindric hyphae forming a trichodermium. In some species the pileipellis hyphae are in a hymeniform layer formed by the apices of the cylindric hyphae (*P. centroamericanus*, *P. curvatus*, and *P. orientalis*). Sometimes, when the tissue is mounted in KOH, brown or yellow content exudates from the colored hyphae of the pileipellis. The pileal trama in general is formed by cylindric hyphae that are thin-walled, hyaline, and either parallel or interwoven.

Stipe. Caulocystidia are often present on the stipe surface (stipitipellis) along the hyphae that form the trichodermium. The stipitipellis hyphae generally give rise to caulocystidia, which can be in clusters or not. They can also form dermatocystidia where sterile basidia are sometimes present, or they can form chains of short cells (*P. phaeoxanthus*, *Phylloporus* sp. 2). The trama of the stipe is usually parallel and vertically oriented, formed by cylindrical, thin-walled, and hyaline hyphae.

Clamp connections. Clamp connections were believed to be absent in *Phylloporus*, and their absence was used as another character to separate *Phylloporus* from *Paxillus*. Whereas in *Paxillus* the clamps are always present in every tissue, they are rare in some species of *Phylloporus* and seen only in some tissues (e.g., at the base of some basidia in *P. colligatus* and in some hyphae in the hymenophoral trama in *P. foliiporus*). They are only abundant in two *Phylloporus* species where they are seen in most of the septa (*P.*

fibulatus and *P. pseudopaxillus*). Heinemann & Rammeloo (1987b) first reported clamp connections in *P. pseudopaxillus*, from Africa, arguing that it could be a species of *Paxillus* but keeping the species in *Phylloporus* due to the presence of cystidia and the hymenophore characteristics. Singer (1945) noticed rare clamps in *P. foliiporus* from the United States. Later, Singer et al. (1990) published *P. fibulatus* from Colombia. *Phylloporus colligatus* is a species from Guyana in which clamp connections were seen at the base of some basidia.

References

Agerer, R. 1987-1998. Colour atlas of ectomycorrhizae. 1st-11th ed. Einhorn-Verlag, Schwäbisch Gmünd. pp.

Baroni, T.J. 1978. Chemical Spot Test Reactions -- Boletes. Mycologia 70: 1064-1076.

Besl, H. and Bresinsky, A. 1997. Chemosystematics of Suillaceae and Gomphidiaceae (suborder Suillineae). Plant Systematics and Evolution 206: 223-242.

Binder, M. 1999. Zur molekularen systematik der Boletales: Boletineae und Sclerodermatineae subordo nov. Ph.D. University of Regensburg, pp. 147.

Binder, M. and Hibbett, D.S. 2006. Molecular systematics and biological diversification of Boletales. Mycologia 98: 971-981.

Bresinsky, A. and Besl, H. 2003. Beiträge zu einer Mykoflora Deutschlands: Schlüssel zur Gattungsbestimmung der Blätter-Leisten- und Röhrenpilze. Regensburger Mykologische Schriften 11: 5-236.

Bresinsky, A., Jarosch, M., Fischer, M., Schonberger, I. and Wittmann-Bresinsky, B. 1999. Phylogenetic relationships within *Paxillus* s.l. (Basidiomycetes, Boletales): separation of a Southern hemisphere genus. Plant Biology 1: 327-333.

Chevallier, F.F. 1826. Flore générale des environs de Paris. ed. Ferra, Paris. 248 pp.

Corner, E.J.H. 1970. *Phylloporus* Quél. and *Paxillus* Fr. in Malaya and Borneo. Nova Hedwigia 20: 793-822.

Corner, E.J.H. 1972. *Boletus* in Malaysia. ed. Government Printing Office, Singapore. 263 pp.

Halling, R.E. 2001. Ectomycorrhizae: Co-evolution, significance, and biogeography. Annals of the Missouri Botanical Garden 88: 5-13.

Hawksworth, D.L., Kirk, P.M., Sutton, B.C. and Pegler, D.N. 1995. Ainsworth and Bisby's dictionary of fungi. 8th ed. CAB International University Press, Cambridge. 616 pp.

Heinemann, P. and Rammeloo, J. 1987a. Novitates generis Phyllopori (Boletineae). Bulletin du Jardin Botanique National de Belgique 57: 272-274.

Heinemann, P. and Rammeloo, J. 1987b. *Phylloporus* (Boletineae). Flore Illustreé des Champignons d'Afrique Centrale 13: 277-309.

Hellwig, V. 1999. Universität München, pp. 228.

Hibbett, D.S. and Binder, M. 2002. Evolution of complex fruiting-body morphologies in homobasidiomycetes. Proceedings of the Royal Society of London. Series B. Biological Sciences 269: 1963-1969.

Hibbett, D.S., Pine, E.M., Langer, E., Langer, G. and Donoghe, M.J. 1997. Evolution of gilled mushrooms and puffballs inferred from ribosomal DNA sequences. Proceedings of the National Academy of Sciences of the United States of America 94: 12002-12006.

Hibbett, D.S. and Thorn, R.G. 2001. Basidiomycota: Homobasidiomycetes. In The Mycota VII Part B Vol. VII (Ed, Lemke, M.M.) Springer-Verlag, Berlin.

Høiland, K. 1987. A new approach to the phylogeny of the order Boletales (Basidiomycotina). Nordic Journal of Botany 7: 705-718.

Jülich, W. 1981. Higher Taxa of Basidiomycetes. ed. J. Cramer, Vaduz. 485 pp.

Kirk, P.M., Cannon, P.F., David, J.C. and Stalpers, J.A. 2001. Ainsworth and Bisby's dictionary of the fungi. 9th ed. CAB International University Press, Cambridge. 665 pp.

Kretzer, A.M. and Bruns, T.D. 1999. Use of atp6 in fungal phylogenetics: an example from the Boletales. Molecular Phylogenetics and Evolution 13: 483-492.

Ladurner, H. and Simonini, G. 2003. *Xerocomus* s.l. 8. ed. Edizioni Candusso, Alassio. 527 pp.

Larsson, K.H., Larsson, E. and Kõljalg, U. 2004. High phylogenetic diversity among corticioid homobasidiomycetes. Mycological Research 108: 983-1002.

Lohwag, H. and Peringer, M. 1937. Zur anatomie der Boletaceae. Annales Mycologici 35: 295-331.

McNabb, R.F.R. 1971. Some new and revised taxa of New Zealand Basidiomycetes (Fungi). New Zealand Journal of Botany 9: 355-370.

Moncalvo, J.-M., Lutzoni, F.M., Rehner, S.A., Johnson, J. and Vilgalys, R. 2000. Phylogenetic relationships of agaric fungi based on nuclear large subunit ribosomal DNA sequences. Systematic Biology 49: 278-305.

Montoya, L. and Bandala, V.M. 1991. Studies on the genus *Phylloporus* in Mexico, I. Discussion of the known species and description of a new species and a new record. Mycotaxon 41: 471-482.

Mueller, G.M. and Halling, R.E. 1995. Evidence for high diversity of Agaricales (Fungi) in neotropical montane *Quercus* forests. In Biodiversity and conservation of neotropical montane forests (Ed, Churchill, S., Balslev, H, Forero, E, Luteyn, J) New York Botanical Garden, Bronx, pp. 302-312.

Neves, M.A. and Capelari, M. accepted. A preliminary checklist of the Boletales from Brazil and notes on Boletales specimens at the Instituto de Botânica (SP) Herbarium, São Paulo, SP, Brazil. Sitientibus.

Pegler, D.N. and Young, T.W.K. 1981. A natural arrangement of the Boletales, with reference to spore morphology. Transactions of the British Mycological Society 76: 103-146.

Peintner, U., Ladurner, H. and Simonini, G. 2003. *Xerocomus cisalpinus* sp. nov., and the delimitation of species in the *X. chrysenteron* complex based on morphology and rDNA-LSU sequences. Mycological Research 107: 659-679.

Quélet, L. 1888. Flore mycologique de la France et des pays limitrophes. ed. Paris. 492 pp.

Roberts, C., Ceska, O., Kroeger, P. and Kendrick, B. 2004. Macrofungi from six habitats over five years in Clayoquot Sound, Vancouver Island. Canadian Journal of Botany 82: 1518-1538.

Singer, R. 1945. The Boletineae of Florida with notes on extralimital species. II. the Boletaceae (Gyroporoideae). Farlowia 2: 223-303.

Singer, R. 1978. Notes on Bolete taxonomy II. Persoonia 9: 421-438.

Singer, R. 1986. The Agaricales in Modern Taxonomy. 4th ed. Koeltz Scientific Books, Königstein, Germany. 981 pp.

Singer, R., Araujo, I. and Ivory, M.H. 1983. Ectotrophically Mycorrhizal Fungi of the Neotropical Lowlands, Especially Central Amazonia. 77. ed. Lubrecht & Cramer Ltd, 352 pp.

Singer, R. and Gómez, L.D. 1984. The Basidiomycetes of Costa Rica. III. The genus *Phylloporus* (Boletaceae). Brenesia 22: 163-181.

Singer, R., Ovrebo, C.L. and Halling, R.E. 1990. New species of *Phylloporus* and *Tricholomopsis* from Colombia, with notes on *Phylloporus boletinoides*. Mycologia 82: 452-459.

Smith, A.H. and Thiers, H.D. 1964. A contribution toward a monograph of North American species of *Suillus*. Ann Arbor, Michigan. 116 pp.

Snell, W.H. and Dick, E.A. 1958. Notes on boletes. X. A few miscellaneous discussions and a new subspecies. Mycologia 50: 57-65.

Taylor, A.F.S., Hills, A.E., Simonini, G., Both, E.E. and Eberhardt, U. 2006. Detection of species within the *Xerocomus subtomentosus* complex in Europe using rDNA–ITS sequences. Mycological Research 110: 276-287.

Watling, R. and Gregory, N.M. 1991. Observations of the boletes of the Cooloola Sandmass, Queensland and notes on their distribution in Australia. Part 3. Lamellate taxa. Edinburgh Journal of Botany 48: 353-391.

Watling, R. and See, L.S. 1998. Ectomycorrhizal fungi associated with members of the Dipterocarpaceae in Peninsular Malaysia - II. Journal of Tropical Forest Science 10: 421-430.

CHAPTER 2: A CONTRIBUTION TOWARD A MONOGRAPH OF *PHYLLOPORUS*

Abstract

A systematic study of *Phylloporus* is presented. Twenty-six species of *Phylloporus* are described; seven of them are new for science. The new taxa are *P. alborufus* from Costa Rica, *P. colligatus* from Guyana, *P. pumilus* from Indonesia, *P. curvatus*, *P. rubiginosus*, *P. dimorphus*, and *P. infuscatus* from Thailand. The species descriptions are supplemented with keys and distribution maps for collections from Australia, Belize, Colombia, Costa Rica, Germany, Guyana, Indonesia, Italy, Malaysia, Mexico, Panama, Slovakia, Thailand, and USA.

Introduction

Phylloporus is one of the most diverse genera in the Boletaceae in terms of number of species. The type species, *P. pelletieri*, grows in temperate Europe but the genus is mostly pantropical and approximately 70 species have been described from various regions of the world. This number will probably increase as new collections are made from tropical regions that have not been properly inventoried.

Corner (1970) recorded 15 species of *Phylloporus* in Malaysia and Borneo. Of the 15 species, 13 were new for science. Based on the high species diversity of *Phylloporus* in this region, Corner suggested that the origin of the genus was in Malaysia. However, this suggestion is debatable because other potentially diverse regions, such as Australia and the neotropics, have not been properly surveyed. For example, Watling & Gregory (1991) recorded 13 species for Australia. Three species of the genus have been collected in

Colombia (Halling 1989, Singer et al. 1990), and two species are known from Mexico (Montoya & Bandala 1991). Five species are known from North America, mostly distributed in the United States, four on the east coast and one rarely collected species on the west coast (Singer 1945, Smith & Trappe 1972). Costa Rica, one of the most explored neotropical regions in terms of fungi collections, has 5 species (Halling et al. 1999, Singer & Gómez 1984).

Corner (1970) also suggested that species of *Phylloporus* are connected to species of trees in the Fagaceae. The morphology of many recently described neotropical taxa indicates that closely related species co-migrated from North America into Central and South America. The co-migration of mycorrhizal communities has been suggested by different authors and observed in many other fungal species (Halling 2001).

In this paper, 26 species of *Phylloporus* are described, among which seven are new for science and two are distinct but remain incompletely described and unnamed due to lack of adequate material. An identification key and distribution maps of the species are also presented. The descriptions begin with the type species, *Phylloporus pelletieri*, and are followed by the other species in alphabetical order.

Materials and Methods

The collections cited in this work were studied macro- and microscopically following traditional mycological methods (Largent 1986, Largent et al. 1977). Sections were mounted in 5% KOH solution and Melzer's reagent after being rehydrated with ethanol and water. The sections were observed with an Olympus BH-2 light microscope. The Imler's reaction is cited by Watling & Gregory (1991) in their studies of the boletes from the Cooloola Sandmass, Queensland; this reaction is also called the fleeting amyloid reaction (Ladurner & Simonini 2003). To test for Imler's reaction, a small piece of the lamellae (3 mm broad slice from the lamellae) is mounted in Melzer's reagent and carefully squeezed between the slide and cover slip. This reaction is visible to the naked eye when the mounted slide is placed against white paper. The pieces of lamellae will turn blue when the reaction is positive. The reaction can vary from a light, inconspicuous blue to a very dark blue, and it will always vanish within a few minutes. When observed under the microscope, the reaction appears to be located in the trama hyphae, but due to the thickness of the piece used it is hard to determine the exact location.

In the descriptions of the basidiospores, Q is the mean length/width quotient. Color terms and codes (e.g., 5D3) are those of Kornerup & Wanscher (1978) and were noted only for species that were observed when they were fresh. Line drawings were made with the aid of a drawing tube. Herbarium acronyms are from Holmgren et al. (1990).

Scanning electron microscopy (SEM) studies of the spores were conducted at Lehman College, City University of New York. Fragments of the hymenophore were removed from dried basidiomata, mounted directly on aluminum stubs (EMS#75610) using carbon adhesive tabs (EMS#77825-12), and coated with 10 nm of gold using a Hummer II sputter coater. The basidiospores were examined with a Hitachi S-2700 scanning electron microscope operating at 10KeV.

Maps of distribution of species were prepared using ArcGIS 9 (ESRI, Inc.) and include only the specimens that were examined in this treatment.

The following descriptions were generated from a Delta database (Dallwitz 1980, Dallwitz et al. 1993 onwards).

Taxonomy

Phylloporus Quélet, Flore Mycologique de la France (Paris). 409. 1888.

Type species: *Agaricus pelletieri* Lév. in Crouan, Florule du Finistère: 81. 1867.

Pileus fleshy, centrally stipitate, convex to flat and in some cases becoming infundibuliform; surface dry, tomentose to subtomentose, sometimes reacting positively to NH_4OH (becoming blue or in rarer cases purplish). Context usually thick and fleshy, except for very small specimens, typically whitish or pale yellow, rarely tan brown. Odor and taste never very evident.

Hymenophore lamellate or rarely subporoid, in many cases intervenose, often decurrent, sometimes descending the stipe, but also adnate, bright yellow to ochraceous yellow or yellowish, cyanescent or not when bruised.

Stipe central, dry, stright or curved, cylindrical or tapering downwards, usually filled, rarely hollow in older specimens; surface smooth or very finely reticulate to pruinose or squamulose, to finely subscabrous; basal mycelium white or yellow.

Spore deposit brown with olive tinges; spores commonly fusoid or less frequently ovoid, yellowish to ochraceous and smooth under the light microscope, smooth or with a very fine bacillate rugosity under SEM.

Basidia clavate, four-sterigmate. Hymenial cystidia present and conspicuous, in general fusoid. *Hymenophoral trama* phylloporoid (bilateral) or boletoid (divergent). Clamp connections typically absent, but present in three species.

Mostly terrestrial, presumably forming ectomycorrhizae, probably with Caesalpinioideae (*Dicymbe*), Casuarinaceae, Dipterocarpaceae, Fagaceae, Mimosaceae (*Acacia*), Myrtaceae, or Pinaceae.

Nomenclatural clarification

Phylloporus was erected by Quélet (1888) with a single species, *Agaricus pelletieri* Lév. (1867) from Europe, which serves as the nomenclatural type. One can only assume Quélet was unaware of the American taxon *Agaricus rhodoxanthus*, a species published earlier by Schweinitz (1822) and now placed in *Phylloporus*. Konrad & Maublanc (1928) published text and a color plate of a taxon that they called *P. rhodoxanthus* and treated *P. pelletieri* as a later synonym. Later, Singer (1938) relegated these two species to subspecies of *P. rhodoxanthus*. *Phylloporus rhodoxanthus* ssp. *europaeus* was used for the European taxon based on Konrad & Maublanc (1928) plate 392 (erroneously cited as pl. 420 by Singer) of *P. rhodoxanthus*. At the same time, *P. rhodoxanthus* ssp. *americanus* was used for the American taxon of *P. rhodoxanthus* described by Schweinitz from North Carolina. These were valid names at the time because the Botanical Code did not recognize autonyms. *Phylloporus bogoriensis* Höhn. and *P. sulcatus* (Pat.) E.-J. Gilbert were maintained as separate species (Singer 1938).

Phylloporus rhodoxanthus and *P. pelletieri* were the most widely used names until Singer (1945) published a description of *P. rhodoxanthus* ssp. *foliiporus* and detailed discussions of ssp. *bogoriensis*, ssp. *americanus*, and ssp. *europaeus* with a key to the four subspecies. In this publication, Singer indicated that *P. pelletieri* is the same as *P. rhodoxanthus* ssp. *europaeus*.

The name *P. rhodoxanthus* (Schwein.) Bres. ssp. *rhodoxanthus* was first used by Singer (1978). At that time, his concept of this subspecies was based on basidiomata mixed in with the type specimen of *Naucoria mexicana* Murrill housed at NY. The cited basidiomata was examined and it corresponds to *P. centroamericanus*. That specimen has no nomenclatural bearing or status in *Phylloporus*. Still, it is not clear what Singer (1978) considered to be the type of the genus because he did not explicitly indicate *P. pelletieri* as such. Later, Singer & Gómez (1984) stated that *P. rhodoxanthus* is an American species.

Key to the Species of *Phylloporus*

1. Basal mycelium white.

2. Pileus red-brown, 2.5-3.5 cm in diameter, becoming blue or blue-green when exposed to NH_4.

3. Flesh unchanging when exposed, cystidia lanceolate 62.3-84 x 7-11.2 µm, sometimes encrusted.. *P. alborufus*

3.Flesh cyanescent when exposed, cystidia ampullaceous, 50-71 x 11-12 µm, never encrusted .... *P. leucomycelinus*

2.Pileus brown or olive.

4. Pileus olive green, flesh ochraceous brown, unchanging, spores ovoid*P. infuscatus*

4. Pileus brown, flesh white or yellow, unchanging or not, spores fusoid or subfusoid.

5. Hymenophore alveolate or subtubular.

6. Pileus smaller than 1 cm in diameter, blue when exposed to NH_4, hymenophore yellow... *P. pumilus*

6. Pileus 2-4 cm in diameter, with NH_4 violet or orangish, hymenophore olivaceous... *P. boletinoides*

5. Hymneophore lamellate.

7. Flesh unchanging when exposed, associated with *Alnus*.............. *P. caballeroi*

7. Flesh changing or rarely not when exposed.

8. Reaction of NH_4 on pileus negative................................... *Phylloporus sp.1*

8. Reaction of NH_4 on pileus positive (blue or blue lilac).

9. NH_4 on pileus consistently blue, cystidia thick walled (2-4 µm) ....*P. centroamericanus*

9. NH_4 on pileus blue lilac, cystidia thin walled.

10. Spores 10.5-11.9 µm long .... *P. orientalis* var. *brevisporus*

10. Spores less than 10 µm long.

11. Pileus 6 cm or less in diameter, flesh unchanging or bluing when exposed...*P. bellus*

11. Pileus 6–10 cm in diameter, flesh reddening when exposed............*P. bogoriensis*

1. Basal mycelium yellow.

12. Flesh unchanging when exposed, NH_4 reaction on pileus positive, or, if negative then spores ovoid.

13. Pileus orange, spores ovoid... *P. aurantiacus*

13. Pileus colored otherwise, spores fusoid.

14. Lamellae unchanging when bruised.

15. Pileus olive, associated with *Pinus* (growing on sandy soil) ....*P. arenicola*

15. Pileus brown, associated with *Quercus*, *Fagus*, *Larix*, or *Picea*, never in sandy soil.

16. Cystidia non-encrusted...*P. pelletieri*

16. Cystidia encrusted.

17. Stipitipellis composed of cells in chains .... *P. phaeoxanthus*

17'. Stipitipellis composed of cylindric hyphae....................*P. rhodoxanthus*

14. Lamellae bluing when bruised.

18. Stipe finely subscabrous..*P. scabripes*

18. Stipe smooth or finely punctuate.

19. Spores 8.4-9.8 x 3.5-4.2 µm, clamp connections present at the base of the basidia, but not abundant ....*P. colligatus*

19. Spores 9.1-12.6 x 3.5-4.9 µm, clamp connections never present................ ....*P. novae-zelandiae*

12. Flesh staining blue when exposed, NH_4 reaction on pileus negative.

20. Spores ovoid, or both ovoid and fusoid.

21. Spores both ovoid and fusoid, pileus ochraceous brown, associated with *Castanopsis* .... *P. dimorphus*

21. Spores ovoid, pileus dark red brown, associated with *Quercus* and *Pinus*......*P. guzmanii*

20. Spores never ovoid.

22. Clamp connections abundant in all tissues .... *P. fibulatus*

22. Clamp connections absent or rare.

23. NH_4 reaction on pileus surface negative...................................*Phylloporus sp.2*

23. NH_4 reaction on pileus surface positive.

24. Pileus not larger than 3.5 cm diameter when mature, spores 7.7-10.5 x 3.5-4.2 µm, flesh staining pink with NH_4...*P. curvatus*

24. Pileus larger than 4 cm diameter when mature, spores 9.8-12.6 x 3.5-4.9 µm, flesh staining blue with NH_4.

25. Cystidia melleous at the apex, pileus red with yellowish stipe, associated with *Quercus* or *Pinus* in the United States...*P. foliiporus*

25. Cystidia hyaline, basidioma entirely red, associated with *Castanopsis* or *Dipterocarpus* in Thailand..*P. rubiginosus*

(1) **Phylloporus pelletieri** (Lév.) Quél., Fl. Mycol. France (Paris). 409. 1888.

Figure 2-1, 2-28

Agaricus pelletieri Lév. in Crouan, Florule du Finistère 81. 1867.

Agaricus paradoxus Kalchbr., Icon. Sel. Hymenomyc. Hung. 2: 27. 1874.

Paxillus paradoxus (Kalchbr.) Cooke, Grevillea 5:6. 1876.

Flammula paradoxa (Kalchbr.) Sacc., Syll. Fung. 5: 810. 1887.

Phylloporus rhodoxanthus ssp. *europaeus* Singer, Rev. Mycol. (Paris) 3:171. 1938.

Pileus 3–8 cm broad, at first obtusely convex, with age plano-convex or plane or plano-depressed, dry, even, becoming finely cracked; disc smooth, at first dark reddish brown, then olive-ochre (tinges); surface matted tomentose, becoming glabrous, with KOH blue, with NH_4 blue. *Flesh* pale pinkish brown, staining absent; odor not distinctive; taste inconspicuous; with NH_4 no reaction. *Hymenophore* lamellate, subdecurrent or adnate. *Lamellae* close, anastomosing, shallowly intervenose, forked, when young bright yellow, with age yellowish green, staining absent; edges even. *Stipe* 2–7 cm long, 4–20 mm wide, equal or tapering downwards, curved, dry; upper half when young floccose (finely), yellow, with age longitudinally ribbed; lower half when young floccose (finely), yellow; *base* yellow, staining not present; interior solid; flesh at base when young yellow or whitish yellow; staining not present or more frequently dull purple, with NH_4 no reaction. *Basal mycelium* yellow. *Fleeting-amyloid reaction* positive.

Basidiospores olive brown in mass, (7.7–)9.8–11.9 µm long, 3.5–4.2 µm wide, mean Q = 2.54, lacrymoid, smooth, smooth or finely rugulose to smooth under SEM, weakly dextrinoid, in KOH pale yellow. *Basidia* 41.3–78.4 µm long, 8.4–14 µm wide, clavate, hyaline, 4-sterigmate. *Hymenial cystidia* 54.6–63.7 µm long, 17.5–20.3 µm wide, moderately abundant, thin walled, hyaline or gray brown contents (in cheilocystidia), cylindric or fusoid-ventricose, encrusting pigment absent. *Hymenophoral trama* bilateral; hyphae cylindric, hyaline, inamyloid. *Pileipellis hyphae* a trichodermium, in KOH hyaline, inamyloid; elements 6–12 µm wide, cylindric, smooth, thin walled; slight granular content present. *Pileus trama* hyphae hyaline, inamyloid, thin walled. *Stipitipellis hyphae* vertically oriented, interwoven, giving rise to a caulohymenium (on the upper half of the pileus), subcylindric or cylindric, hyaline. *Stipe trama* hyphae parallel, cylindric. *Clamp connections* absent.

MYCORRHIZAL HOSTS: *Fagus*, *Larix*, *Picea*, *Quercus*, *Castanea*.

DISTRIBUTION: According to Ladurner and Simonini (2003), the species is widespread in Europe, except for Ireland, Albania, Greece, Finland and Iceland, where there are no reports of collections.

Three collections were examined, one from Slovakia, one from Italy, and one from Austria.

Singer (1945) mentioned a von Höhnel collection deposited in FH as "good European material from St. Johann in Tyrol." I examined this collection and it agrees with the *P. pelletieri* description by Ladurner & Simonini (2003, as *Xerocomus pelletieri* (Lév.) Bresinsky & Manfr. Binder). Singer (1945) considered these two ssp. as distinct, but closely related, taxa based on geographic distribution and on differences of the hymenophore configuration. Even though the macroscopic differences between this taxon and *P. rhodoxanthus* are not very conspicuous (*P. pelletieri* has more anastomosed lamellae and a more velvety pileus), microscopically there are other characteristics to separate them. *Phylloporus pelletieri* has shorter spores that are smooth or finely rugulose under SEM and has longer basidia, while the spores of *P. rhodoxanthus* have a bacillate ornamentation under SEM. In addition, molecular analyses (see LSU and ITS phylogenies in this work) support these as distinct taxa. Therefore, *P. pelletieri* is here affirmed as a valid species and the type species of the genus.

(2) **Phylloporus alborufus** Neves & Halling sp. nov.

Figure 2-2, 2-31

ETYMOLOGY: from Latin albo (white) + rufus (red), referring to the white basal mycelium and the red pileus

Pileus 2.5–3.5 cm broad, at first plane, with age plano-depressed, dry, entire, becoming even; disc even, at first dark reddish brown (8E7, 8E8), then brownish red; margin inrolled (slightly when young), when young deep red brown; surface matted tomentose, becoming subtomentose, with NH_4 blue. *Flesh* 3–4 mm thick, yellow, staining absent; odor mild; taste mild. *Hymenophore* lamellate, decurrent. *Lamellae* subdistant, 3–4.5 mm wide, not anastomosing, intervenose, when young bright yellow (2A7), with age yellow, staining blue; edges even. *Stipe* 2.3–5.1 cm long, 6 mm wide, equal, sinuate, dry; upper half when young fibrillose, white with a red brown line next to the hymenophore, with age fibrillose or subfibrillose; lower half when young subfibrillose, brownish red (7C5), with age slightly roughened-fibrillose or subfibrillose, red or buff tan (ground color); fibrils on upper half when young brown or reddish (8E8); base yellow, staining not present; interior solid; flesh above when young whitish yellow, with NH_4 blue. *Basal mycelium* white. *Fleeting-amyloid reaction* positive.

Basidiospores 9.8–11.2 µm long, 3.5–4.2 µm wide, mean Q = 2.72, subfusoid, smooth, with SEM smooth to finely rugulose, slightly amyloid, in KOH ochraceous. *Basidia* 24.5–32.2 µm long, 7–7.7 µm wide, clavate, hyaline, 4 -sterigmate. *Hymenial cystidia* 62.3–84 µm long, 7–11.2 µm wide, numerous on sides and edges of lamellae, thin walled, hyaline, lanceolate, encrusting pigment present (in some). *Hymenophoral trama* bilateral, with age divergent; hyphae cylindric, hyaline, amyloid. *Pileipellis hyphae* a trichodermium, in KOH pale yellow or hyaline, inamyloid; cylindric, smooth, thin walled, granular content present (some). *Pileus trama* hyphae hyaline, smooth, thin walled. *Stipitipellis hyphae* vertically oriented, parallel, giving rise to clusters of caulocystidia, cylindric or clavate, hyaline. *Stipe trama* hyphae parallel, cylindric. *Clamp connections* absent.

MYCORRHIZAL HOST: *Quercus*.

DISTRIBUTION: This species is only known from the type locality: San Gerardo de Dota, San José, Costa Rica. It is possible that it will be found in other places in Costa Rica and possibly Mexico and Colombia, where oak forests are also present.

This taxon is phylogenetically related to the American *P. leucomycelinus*, also a red pileate fungus with white basal mycelium (see LSU and ITS phylogeny). While *P. alborufus* was found in a *Quercus* forest, *P. leucomycelinus* is normally associated with *Fagus*, but may be found with *Quercus*. Compared to *P. leucomycelinus, Phylloporus alborufus* spores are slightly shorter and some cystidia have encrusting pigment.

(3) **Phylloporus arenicola** A.H. Sm. & Trappe, Mycologia 64: 1138–1153. 1972.

Figure 2-3, 2-30

Pileus 1.5–4.5 cm broad, at first plane, with age convex or depressed, dry, even; disc subtomentose, at first olive, then olive-ochre; margin when young olive, then olive-brown or olive-ochre; surface velutinous, becoming subtomentose or tomentose, with NH_4 red or purple. *Flesh* pale yellow, staining absent; odor absent; taste mild; with $FeSO_4$ olivaceous. *Hymenophore* lamellate, adnexed. *Lamellae* subdistant, simple, when young bright yellow, with age mustard yellow, staining absent; edges even. *Stipe* 4–6 cm long, 6–8 mm wide, tapering downwards, curved or sinuate, dry; upper half when young pruinose, with age subpruinose; lower half when young pruinose, dull yellow; pruina on upper half when young brown; base bright yellow, staining not present; interior solid; yellow, flesh at base when young bright yellow, staining not present. *Basal mycelium* yellow. *Fleeting-amyloid reaction* positive (weakly).

Basidiospores 9–12 µm long, 4–5 µm wide, mean Q = 2.3, ovoid or lacrymoid, smooth, weakly dextrinoid, in KOH olive-hyaline. *Basidia* 38–60 µm long, 9–13 µm wide, clavate, hyaline, 4 -sterigmate. *Hymenial cystidia* 52–86 µm long, 9–16 µm wide, numerous on sides and edges of lamellae, thin walled, hyaline, subfusoid or fusoid-ventricose, encrusting pigment absent. *Hymenophoral trama* bilateral; hyphae cylindric, 9–13 µm wide, hyaline. *Pileipellis hyphae* a trichodermium, in KOH pale yellow or hyaline, inamyloid; cylindric or elongated, encrusted with pigment (in Melzer's), thin walled, granular content absent. *Pileus trama* hyphae hyaline, smooth. *Stipitipellis hyphae* vertically oriented, parallel, giving rise to versiform caulocystidia (rare), 44.8–45.5 µm long, 15.4–16.1 µm wide, cylindric, hyaline. *Stipe trama* hyphae parallel, cylindric, hyaline, somewhat dextrinoid. *Clamp connections* absent.

MYCORRHIZAL HOST: *Pinus*.

DISTRIBUTION: Collections of *P. arenicola* are from the west coast of the USA in Washington, Oregon, and Northern California.

This olive colored *Phylloporus* is known from sandy soils in the western USA and is not as common as *P. rhodoxanthus* in the east. *Phylloporus arenicola* does not turn blue when injured; its pileus cuticle turns violet-fuscous or reddish with NH_4, and the dimensions of the basidiome are in general smaller than those observed for *P. rhodoxanthus*. The forests with which each taxon is associated also differ between the two species. *Phylloporus arenicola* is associated with *Pinus* (*Pinus contorta* in the type location, Tillamook County, Oregon, USA), while the hosts for *P. rhodoxanthus* are species of *Quercus*. Phylogenetically, both species are in the same clade, forming a group that also includes *P. phaeoxanthus*, a species from Costa Rica with yellow basal mycelium that is also associated with *Quercus* in Central (and possibly South) America.

(4) **Phylloporus aurantiacus** Halling & G.M. Muell., Mycotaxon 73: 64. 1999.

Figure 2-4, 2-31

Pileus 1–3 cm broad, at first plane, with age depressed, dry; disc even, orange; margin even, when young orange to yellow orange, then yellow orange; surface matted tomentose to tomentose, becoming matted subtomentose, maculose color spots absent or present (sometimes), yellow, with NH_4 no reaction. *Flesh* 2–5 mm thick, yellow, staining absent; odor mild; taste mild. *Hymenophore* lamellate, decurrent. *Lamellae* close, 1–3 mm wide, not anastomosing, rarely somewhat intervenose, orange to orangish brown, staining green; edges even. *Stipe* 3–5 cm long, 4–5 mm wide, equal, strict, dry; floccose to fibrillose, red to orange (with yellow undertones); base yellow, staining not present or green; interior solid; flesh above yellow, staining not present, flesh at base yellow, staining not present. *Basal mycelium* bright yellow. *Fleeting-amyloid reaction* positive.

Basidiospores 5.6–7.7 µm long, 3.5–5 µm wide, mean Q = 1.6, oblong to ovoid, smooth, weakly dextrinoid, in KOH straw yellow. *Basidia* 35–42 µm long, 8–10.5 µm wide, cylindric to clavate, hyaline, 4 -sterigmate. *Hymenial cystidia* 60–90 µm long, 7–10 µm wide, more common on sides of lamellae, thin walled, hyaline, subfusoid to subcylindrical, encrusting pigment absent. *Hymenophoral trama* bilateral; yellow brown in the central stratum. *Pileipellis hyphae* a trichodermium (tangled), in KOH orange to yellow ochraceous, inamyloid; elements 3.5–7 µm wide, cylindric to elongated, smooth, thin walled, granular content absent. *Pileus trama* interwoven, hyphae hyaline, inamyloid, with elements 3.5–10 µm wide, smooth (but dissolving a bright yellow pigment in KOH), thin walled. *Stipitipellis hyphae* vertically oriented, parallel, giving rise to clusters of caulocystidia, 35–90 µm long, 7–10 µm wide, clavate to subfusoid, hyaline to orange. *Stipe trama* hyphae parallel, cylindric, hyaline, inamyloid. *Clamp connections* absent.

MYCORRHIZAL HOST: *Quercus*.

DISTRIBUTION: This species is known only from one site in Cartago Province in the northern Talamancas, Costa Rica.

When Halling et al. (1999) described this species from Costa Rica, they considered it to be phenetically closest to *P. coccineus*, described by Corner (1970) from Singapore. Both species possess yellow basal mycelium and ovoid spores, an uncommon shape in the genus. However, Corner’s fungus has cyanescent, yellow flesh and larger spores. The flesh of *P. aurantiacus* is not cyanescent. The diagnostic characteristics are the color of the basidiome, the yellow flesh, and the presence of maculose areas on some of the basidiomes.

(5) **Phylloporus bellus var. bellus** (Massee) Corner, Nova Hedwiga 20: 798. 1970.

Figure 2-5,2-29, 2-31, 2-33, 2-34

Flammula bella Massee, Kew Bull. 74. 1914.

Pileus 1–3.3(–6) cm broad, at first convex, with age plane or depressed or eventually concave, dry; dark brown to yellow orange; surface subtomentose to subvelutinous, becoming granular-fibrillose or subsquamulose, with NH_4 blue to purple (sometimes fading to latter color). *Flesh* white to pale yellow (with age), staining absent or blue. *Hymenophore* lamellate, decurrent. *Lamellae* subclose to distant, not anastomosing, somewhat intervenose, a few forked, sulphur to primrose yellow, staining absent or blue to slightly green. *Stipe* 1–4.4 cm long, 1.3–7.5(–9.5) mm wide, mostly equal or subequal or clavate (occasionally), dry; upper half when young glabrous to subglabrous, yellow to light cinnamon; lower half when young subglabrous to glabrous, sordid yellow to light cinnamon, with age finely pustulate; base white; interior solid; flesh above white to whitish yellow or pale cinnamon, staining not present or blue; flesh at base whitish yellow or pale cinnamon, staining not present or blue. *Basal mycelium* white. *Fleeting-amyloid reaction* positive.

Basidiospores olive in mass, (8–) 9–10 µm long, (2.7–) 3.5–4.5 (–5.5) µm wide, mean Q = 2–3.3, lacrymoid to subfusoid, smooth, inamyloid, in KOH golden yellow to light brown melleous. *Basidia* 29–38 µm long, 6.5–10 µm wide, clavate, hyaline, 4 -sterigmate. *Hymenial cystidia* 32–81 µm long, 7.5–18 µm wide, numerous on sides and edges of lamellae, thin walled, ampullaceous or clavate-ventricose to ventricose to utriform to obtuse to mucronate, encrusting pigment sometimes present. *Hymenophoral trama* bilateral. *Pileipellis hyphae* a trichodermium or forming a palisade, in KOH pale yellow; elongated or short (eg. 11–16 x 10 µm, but mostly 20–28 x 6–7 µm), smooth, thin walled. *Stipitipellis hyphae* vertically oriented, parallel, giving rise to dermatocystidia, 30–55(–87) µm long, 5–20 µm wide, with incrusting pigment present. *Clamp connections* absent.

MYCORRHIZAL HOSTS: *Quercus* and *Castanopsis*.

DISTRIBUTION: First described from Singapore, Singer & Gómez (1984) cite collections from Japan, Mexico, and Costa Rica.

The diagnostic features are the white basal mycelium, the cyanescent lamellae, and thin-walled non-incrusted hymenial cystidia. Collection REH7733 (Costa Rica) has spores of two sizes and agrees with Singer's description for Costa Rican collections. Collection REH8710 (USA) agrees with REH7733 but has only the smaller spores.

(6) **Phylloporus bogoriensis** Höhn., Sitzber. Kaiserl. Acad. Wiss. Wien Math.-naturw. Kl. 123: 98, n.838. 1914.

Figure 2-6, 2-33, 2-34

Phylloporus rhodoxanthus ssp. *bogoriensis* (Höhn) Singer, Farlowia 2:282. 1945.

Pileus 6–10 cm broad, at first convex, with age plane or concave, dry, even; disc even, at first brownish gray or cinnamon brown, then cinnamon brown; margin even, when young cinnamon brown or chestnut brown, then light brown or ochraceous brown; with NH_4 blue or a pale violet circle around the drop. *Flesh* 4–10 mm thick, pale yellow, staining red or reddish brown or grayish red; odor mild; taste mild; with NH_4 no reaction. *Hymenophore* lamellate, decurrent. *Lamellae* close or subdistant, not anastomosing, shallowly intervenose, when young grayish yellow or ochraceous yellow, with age yellowish brown, staining blue or reddish brown (sometimes); edges even. *Stipe* 3–4(–10) cm long, 4–9 mm wide, equal or subclavate, strict or curved, dry; upper half when young pruinose, yellow, with age subpruinose; lower half when young villous, brown or gray brown; base brown, staining not present; interior solid; flesh above when young buff, staining blue or reddish brown, flesh at base buff. *Basal mycelium* white. *Fleeting-amyloid reaction* positive.

Basidiospores olive brown in mass, 7.7–9.1 µm long, 3.5–4.2 µm wide, mean Q = 2.33, subfusoid, smooth, inamyloid, in KOH pale yellow. *Basidia* 30–32 µm long, 9–10 µm wide, clavate, hyaline, 4 -sterigmate. *Hymenial cystidia* 45.5–51.1 µm long, 9.8–11.2 µm wide, more common on sides of lamellae, thin walled, gray brown contents, fusoid or cylindric, encrusting pigment present. *Hymenophoral trama* divergent; hyphae cylindric, 6–13 µm wide, hyaline. *Pileipellis hyphae* a trichodermium; smooth, thin walled, granular content present. *Pileus trama* hyphae hyaline, smooth, thin walled. *Stipitipellis hyphae* vertically oriented, parallel, giving rise to a caulohymenium, 24.5–25.9 µm long, 5.6–6.3 µm wide, clavate, with incrusting pigment present. *Stipe trama* hyphae parallel, cylindric, hyaline. *Clamp connections* absent.

MYCORRHIZAL HOST: *Dipterocarpus*.

DISTRIBUTION: The type specimen is a Corner collection from northern Borneo. The species has been reported also from Singapore by Corner (1970). Watling & Gregory

(1991) published a description of *P.* aff. *bogoriensis* from Queensland. The description here is based on two collections from Indonesia and Malaysia.

The collections examined here agree with the type description, bearing spores that are more towards the smaller range. Watling and Gregory (1991) suggested that a new taxon might be recognized for an Australian collection, since the spores are a little larger then the type collection. *Phylloporus bogoriensis* is kept as an autonomous species and not as a subspecies of the *rhodoxanthus* complex. This decision is supported by morphological data (the red staining flesh and smaller spores in *P. bogoriensis*), as well as the molecular data (LSU and ITS) and agrees with Corner (1970) and Watling and Gregory (1991).

(7) **Phylloporus boletinoides** A.H. Sm. & Thiers, Contr. N. Amer. Sp. *Suillus*. 105. 1964.

Figure 2-7, 2-29

Pileus 2–4 cm broad, at first convex, with age plano-convex, dry, even; disc subtomentose or velutinous, at first cinnamon brown or dark vinaceous brown, then cocoa brown or cinnamon brown; margin inrolled, brown; surface tomentose, becoming glabrous or matted tomentose, with NH_4 violet purple with an ochre orange circle around the drop. *Flesh* white, staining absent; odor mild; taste mild; with NH_4 purple, with $FeSO_4$ grayish green (slightly). *Hymenophore* lamellate or subporoid (more poroid near the stipe), decurrent. Tubes 2–5 mm long, light yellow (with olive buff tinges), becoming olive, unchanging when injured; pores 2–3 mm wide, pallid yellow or olive buff, then deep olive buff, unchanging when injured. *Stipe* 3–5 cm long, 0.5–1 mm wide, equal or tapering downwards, strict or curved, dry; upper half when young glabrous, yellow, with age longitudinally ribbed (ribs sometimes anastomosing); lower half when young glabrous, light cinnamon, buff tan; base pallid, staining not present; interior solid, or hollow (at the base, with age); flesh yellow, staining not present. *Basal mycelium* white. *Fleeting-amyloid reaction* positive (weak).

Basidiospores 10.5–11.9 µm long, 4.2–4.9 µm wide, mean Q = 2.46, subfusoid or oblong fusoid or cylindric (rarely), smooth, dextrinoid, in KOH ochraceous. *Basidia* 25.9–31.5 µm long, 7–8.4 µm wide, clavate, hyaline, 4 -sterigmate. *Hymenial cystidia* 60.2–81.9 µm long, 7–10.5 µm wide, more common towards the edge of lamellae, thin walled, hyaline (often with brownish content when at the edge of the lamellae), fusoid or fusoid-ventricose, encrusting pigment absent. *Hymenophoral trama* divergent; hyphae cylindric, 1.5–5 µm wide, hyaline or melleous hyaline, subgelatinous in KOH. *Pileipellis hyphae* a trichodermium, in KOH yellow ochraceous, inamyloid; elements 7–11 µm wide, cystidioid or cylindric, thin walled. *Pileus trama* interwoven, hyphae hyaline. *Stipitipellis hyphae* vertically oriented, giving rise to a caulohymenium. *Stipe trama* hyphae hyaline. *Clamp connections* absent.

MYCORRHIZAL HOSTS: *Pinus*, *Quercus*.

DISTRIBUTION: The type was found in Florida and the species also has been collected in Alabama and Texas (Singer et al. 1990). A collection from Belize was examined (TJB9681) and is the first record of *P. boletinoides* outside the USA (Ortiz-Santana et al. 2007).

Phylloporus boletinoides can be distinguished from other *Phylloporus* due to its hymenophore characteristics; the configuration is alveolate to subporoid, more anastomosed than other species, and the color is not bright yellow as in most species, but has a yellowish tone with olive tints. When dried out, the hymenophore loses a lot of its color and becomes a dull brownish ochraceous.

(8) **Phylloporus caballeroi** Singer, Beih. Sydowia 7: 101. 1973.

Figure 2-8, 2-30, 2-31, 2-32

Pileus 1.7–4.5 cm broad, at first pulvinate to plano-convex, with age frequently subumbonate to umbonate, dry; disc uneven or smooth, at first brown to dark brown, then brown to dark reddish brown; margin smooth, cinnamon brown; surface tomentose to velutinous (often fracturing into small brown fibrils on yellowish ground), becoming glabrous (often) or tomentose to velutinous, with NH_4 strongly blue. *Flesh* white or yellowish white, staining absent; odor absent or fruity. *Hymenophore* lamellate, subdecurrent to sinuate to adnate. *Lamellae* subdistant to close, not anastomosing, sometimes intervenose, when young yellow, staining absent or light blue green. *Stipe* 2.4–4.6 cm long, 2.5–7 mm wide, equal or tapering downwards, dry; upper half when young glabrous to subglabrous, pallid, with age yellow pruinose to fibrillose or subreticulate, ground color yellow; lower half when young subglabrous to glabrous, pallid, with age fibrillose to punctate, brown to yellow; interior solid; flesh white or whitish yellow, staining not present or rusty-ochraceous. *Basal mycelium* white. *Fleeting-amyloid reaction* positive.

Basidiospores brownish olive in mass, (9–)10–12(–12.7) µm long, 4–5(–6) µm wide, mean Q = 2.21, mostly subfusoid or subellipsoid to subglobose (few), smooth, dextrinoid. *Basidia* (30.5–)32–37(–40) µm long, (7.5–)8.5–9.5(–11) µm wide, clavate, hyaline, (2–)4 -sterigmate. *Hymenial cystidia* 40–105 µm long, 7.5–14 µm wide, thin walled, hyaline, ampullaceous or fusoid [with obtuse tip but often with constrictions] or utriform, encrusting pigment absent. *Hymenophoral trama* bilateral. *Pileipellis hyphae* a trichodermium and forming a palisade, in KOH yellow or brownish; elements 6–18.5 µm wide, elongated, smooth, thin walled, granular content absent. Intercalary cells subisodiametric, 10–14 µm long, 8.5–10 µm wide. *Pileus trama* interwoven, hyphae hyaline, inamyloid, smooth, thin walled. *Stipitipellis hyphae* vertically oriented, parallel or interwoven, giving rise to dermatocystidia, 25–42 µm long, 5–10 µm wide, mostly clavate, hyaline, with incrusting pigment present (pale melleous and granular). *Clamp connections* absent.

MYCORRHIZAL HOST: *Alnus acuminata*.

DISTRIBUTION: First described from collections gathered in northern Argentina, the species is widespread under the montane neotropical alder from that country north to at least Costa Rica. Collections examined in this work are from Argentina, Bolivia, and Panama.

The ectomycorrhizal association along with the white basal mycelium are good field characters to identify this taxon. Singer & Gómez (1984) compared the Argentinean specimen to Costa Rica collections and suggested that the latter were larger, older collections with tomentose stipes and that the basidiomes collected in Argentina were still young and therefore glabrous due to the age. The collection from Panama agrees with the Costa Rican specimen described in possessing a larger basidiome with a tomentose stipe. The question posed by Singer on whether the collections from Costa Rica and Panama are a different species or a Central American race of *P. caballeroi* cannot be answered until more material is available. The Bolivian material also agrees with the description from the Costa Rica specimen.

This species might be closely related to *P. leucomycelinus* according to the molecular phylogeny, because the sequences obtained from the Panamanian collection place it inside the *P. leucomycelinus* clade. *Phylloporus caballeroi* has a glabrous then fibrillose stipe, short cystidia, and cinnamon brown pilei compared to *P. leucomycelinus,* which has a squamulose stipe, longer cystidia, and red pilei.

The type specimen at F has been annotated with the correct number cited in the protologue (T5150).

(9) **Phylloporus centroamericanus** Singer & L.D. Gómez, Brenesia 22: 169. 1984.

Figure 2-9, 2-29, 2-31, 2-32

Pileus 2–3 cm broad, at first convex, with age plane, dry, even, becoming cracked; disc even, at first brown(7F6), then tan (6D7, 6C6); margin even, when young brown (6E7, 6E8), then tan (6D7); surface subtomentose, becoming finely areolate, with NH_4 blue or blue-green. *Flesh* 5–9 mm thick, white or light yellow (2A4), staining absent or blue green (rare); odor mild; taste mild. *Hymenophore* lamellate, decurrent. *Lamellae* subdistant, 1–5.5 mm wide, not anastomosing, later rather strongly intervenose, when young yellow (3A7) or sulphur yellow (2A7), with age yellowish brown, staining blue; edges even. *Stipe* 3.5–4 cm long, 5–8 mm wide, tapering downwards, sometimes eccentric or curved, dry; upper half when young coarsely subpruinose to finely subscabrous, yellowish (4A2); lower half when young finely subscabrous to subpruinose (coarse), white or yellow (near the base), with age coarsely subpruinose to finely subscabrous; pruina on upper half brown, on lower half brown (6D7); base white, staining not present; interior solid; flesh above white slightly pinkish brown in midportion, staining not present, flesh at base white, staining not present. *Basal mycelium* white. *Fleeting-amyloid reaction* positive.

Basidiospores olive brown in mass, 8–16 µm long, 3.5–5.5 µm wide, mean Q = 2.7, subfusoid, smooth, with SEM finely rugulose, dextrinoid, in KOH light brown melleous. *Basidia* 28–38 µm long, 6.2–8 µm wide, clavate, hyaline, 4 -sterigmate. *Hymenial cystidia* 35–138 µm long, 10–25 µm wide, more common towards the edge of lamellae, thick walled (2-4 µm) hyaline, often clavate to ventricose to fusoid to fusoid mucronate to utriform, encrusting pigment present. *Hymenophoral trama* bilateral; hyphae cylindric, hyaline, in KOH yellow. *Pileipellis hyphae* hymeniform and forming a palisade, in KOH yellow (exuding a yellow pigment on KOH), inamyloid; elements 5–24 µm wide, elongated or subisodiametric or cellular, encrusted with pigment, thin walled, granular content absent. Intercalary cells isodiametric to subisodiametric, 15–32 µm long, 12–24 µm wide. *Pileus trama* interwoven, hyphae hyaline, inamyloid, smooth, thin walled. *Stipitipellis hyphae* vertically oriented, parallel, giving rise to versiform caulocystidia or clusters of caulocystidia, 40–80 µm long, 10–20 µm wide, clavate to ventricose or fusoid, with incrusting pigment present (hyaline to melleous). *Stipe trama* hyphae parallel to subparallel, cylindric, hyaline, inamyloid. *Clamp connections* absent.

MYCORRHIZAL HOST: *Quercus*.

DISTRIBUTION: *Phylloporus centroamericanus* was originally described from Costa Rica and was recently found in Mexico (Montoya & Bandala 1991).

This species is characterized by the red-brown pileus, white mycelium at the stipe base, and conspicuous encrusted hymenial cystidia with thick walls. It is one of the most common species in oak forests of the Cordillera Talamanca of Costa Rica.

(10) **Phylloporus colligatus** Neves & T.W. Henkel sp. nov.

Figure 2-10, 2-32

ETYMOLOGY: colligatus (connections), due to the presence of clamp connections

Pileus 1–1.7 cm broad, at first plano-convex, dry, even, becoming matted subtomentose; reddish brown (with orangish overtones); with NH_4 blue (instantly, but rapidly changing to dull burgundy brown). *Flesh* 1–5 mm thick, off-white, staining absent; odor not distinctive. *Hymenophore* lamellate, decurrent or adnate. *Lamellae* subdistant, forked, when young light yellow, with age yellow, staining brighter yellow; edges even. *Stipe* 3–4.5 cm long, 2–4 mm wide, equal, curved, dry; upper half when young glabrous, dull orangish; lower half orangish yellow; interior solid; flesh above when young off white (sometimes yellowing upon exposure). *Basal mycelium* pale yellow. *Fleeting-amyloid reaction* positive (weakly).

Basidiospores 8.4–9.8 µm long, 3.5–4.2 µm wide, mean Q = 2.36, subfusoid, smooth, inamyloid, in KOH pale yellow. *Basidia* 34.3–37.8 µm long, 6.3–8.4 µm wide, clavate, pale yellow, 4 -sterigmate (rarely 5). *Hymenial cystidia* 42–45.5 µm long, 7–7.7 µm wide, more common on sides of lamellae, thin walled, hyaline, fusoid or lanceolate, encrusting pigment absent. *Hymenophoral trama* bilateral; hyphae cylindric, 4.9–7 µm wide, yellowish. *Pileipellis hyphae* an ixotrichodermium, in KOH pale yellow; hyphae cylindric, thin walled. *Pileus trama* interwoven; hyphae hyaline. *Stipitipellis hyphae* vertically oriented, parallel, subcylindric or cylindric, yellow. *Stipe trama* hyphae parallel, cylindric, pale yellow, 6.3–10.5 µm wide. *Clamp connections* present (rare, but present at the base of some basidia and at septa of the hymenophoral trama hyphae).

MYCORRHIZAL HOST: *Dicymbe*.

DISTRIBUTION: This species is only known from Guyana, where the type collection was found.

This taxon has some similarities to *P. phaeoxanthus* but the cystidia are not incrusted like in the Costa Rican species. It could be confused with *P. phaeoxanthus* var. *simplex* from which it differs from in the fusoid shape of the cystidia and the presence of clamp connections at the base of the basidia. This taxon is possibly phenetically related to African species due to the association with trees on the Caesalpinioideae. The only clamped species described by Heinemann and Rammeloo (1986) from Africa is *P. pseudopaxillus*. However, the African species is larger, has longer spores, and the clamp connections are seen in all parts of the basidiome. See comments under *P. fibulatus* and *P. foliiporus,* the other two clamped species presented here.

(11) **Phylloporus curvatus** Neves & Halling sp. nov.

Figure 2-11, 2-33

ETYMOLOGY: curvatus (curved), due to the curved stipe

Pileus 0.8–3.5 cm broad, at first convex or uplifted, with age plano-depressed to infundibuliform, dry, even; at first brownish orange or pale red brown (8F6), then ochraceous brown (8E5, 8D5); margin smooth; becoming subsquamulose, with NH_4 pale brown (with pinkish tints). *Flesh* pale yellow, staining pale blue; odor absent; taste inconspicuous; with NH_4 light pink. *Hymenophore* lamellate, adnexed. *Lamellae* subdistant, not anastomosing, sometimes inconspicuously intervenose, mostly simple, when young yellow (3A4), staining absent or light blue; edges even. *Stipe* 1–2 cm long, 2–5 mm wide, equal, curved, dry; upper half when young finely squamulose, whitish cream (4B2), with age longitudinally ribbed; on lower half when young orangish ochraceous; base pale yellow; interior solid; flesh when young pale yellow. *Basal mycelium* yellow, or pale yellow. *Fleeting-amyloid reaction* positive.

Basidiospores 7.7–10.5 µm long, 3.5–4.2 µm wide, mean Q = 2.36, subfusoid, smooth, slightly dextrinoid, in KOH light brown melleous. *Basidia* 21.7–28 µm long, 7–8.4 µm wide, clavate, hyaline, 4 -sterigmate. *Hymenial cystidia* 47.6–59.5 µm long, 9.8–12.6 µm wide, numerous on sides and edges of lamellae, thin walled, hyaline, fusoid or subcylindrical, encrusting pigment absent. *Hymenophoral trama* bilateral or divergent (at the edge of the lamellae); hyphae cylindric, (4.9–)5.6–8.4 µm wide, hyaline, inamyloid. *Pileipellis hyphae* hymeniform, in KOH yellow; thin walled, intercalary cells cylindric. *Pileus trama* radial, hyphae hyaline, with elements 5.6–7 µm wide, smooth, thin walled. *Stipitipellis hyphae* vertically oriented, parallel, giving rise to clusters of caulocystidia, 18.9–36.4 µm long, 7–10.5 µm wide, clavate, hyaline. *Stipe trama* hyphae parallel, cylindric, hyaline, 6.3–11.2 µm wide. *Clamp connections* absent.

MYCORRHIZAL HOST: *Castanopsis*.

DISTRIBUTION: From northern Thailand, this species is only known from the type locality.

Of the three collections examined, all basidiomes are of a small diameter and have a distinctly curved stipe. The small size, the curved stipe, and the light pink reaction with NH_4 on the pileus are diagnostic.

(12) **Phylloporus cyanescens** (Corner) Neves & Halling, comb. nov.

Figure 2-5, 2-34

Phylloporus bellus var. *cyanescens* Corner, Niova Hedwigia. 799. 1970.

Pileus 4–6 cm broad, at first convex, with age plane or eventually concave, dry; dark brown to cocoa brown; surface subtomentose to barely subvelutinous, becoming areolate with age, with NH_4 a blue-green flash then light lavender. *Flesh* white, staining pale blue to dark gray. *Hymenophore* lamellate, decurrent. *Lamellae* subdistant, anastomosing, bright yellow, staining blue-green. *Stipe* 4-5 cm long, 8–15 mm wide, mostly equal, dry; upper half subpubescent to matted subtomentose, white to light yellow; lower half subpubescent to matted subtomentose, white; base white; interior solid; flesh white, staining pale blue then grayish fuscous. *Basal mycelium* white. *Fleeting-amyloid reaction* positive.

Basidiospores olive in mass, 9–12.5 µm long, (2.7–) 3.5–5.5 µm wide, mean Q = 2–3.3, lacrymoid to subfusoid, smooth, inamyloid, in KOH golden yellow to light brown melleous. *Basidia* 29–38 µm long, 6.5–10 µm wide, clavate, hyaline, 4 -sterigmate. *Hymenial cystidia* 32–81 µm long, 7.5–18 µm wide, numerous on sides and edges of lamellae, thin walled, ampullaceous or clavate-ventricose to ventricose to utriform to obtuse to mucronate, encrusting pigment sometimes present. *Hymenophoral trama* bilateral. *Pileipellis hyphae* a trichodermium or forming a palisade, in KOH pale yellow; elongated or short (eg. 11–16 x 10 µm, but mostly 20–28 x 6–7 µm), smooth, thin walled. *Stipitipellis hyphae* vertically oriented, parallel, giving rise to dermatocystidia, 30–55(–87) µm long, 5–20 µm wide, with incrusting pigment present. *Clamp connections* absent.

MYCORRHIZAL HOSTS: *Quercus* and *Castanopsis*.

DISTRIBUTION: First described from Malaya (Corner 1970), the species also have been reported from and Australia (Watling & Gregory 1991).

The diagnostic features are the white basal mycelium, the cyanescent lamellae and flesh that turn grayish, and thin-walled non-incrusted hymenial cystidia. *Phylloporus cyanescens* has longer spores and stronger, cyanescent flesh when compared to *P. bellus*. The molecular phylogeny does not support a similarity based on the collections included in the analyses, since *cyanescens* is positioned in a different clade from the two collections of *P. bellus* var. *bellus*. Singer (1978) conjectured that var. *cyanescens* could be *P. foliiporus* and not a variety of *P. bellus*. However, the cystidia of var. *cyanescens* are longer and not melleous at the apex as in *P. foliiporus*. The molecular phylogenies based on LSU and ITS genes also separate these three taxa.

A new combination is suggested to elevate var. *cyanescens* to the species rank based on the morphological differences between the two taxa and on the phylogeny provided by the LSU and ITS analysis.

(13) **Phylloporus dimorphus** Neves & Halling sp. nov.

Figure 2-12

ETYMOLOGY: dimorphus (two forms), due to the presence of two differently shaped spores

Pileus 6.2–7.9 cm broad, at first convex, with age plano-convex, dry, entire or irregularly pitted, becoming irregularly pitted; disc smooth, at first brownish yellow, then ochraceous brown (5C4); margin slightly inrolled, ochraceous; surface finely felted, becoming subfibrillose, with NH_4 vinaceous red. *Flesh* pale yellow, staining pale blue;

odor slightly acrid; with NH_4 no reaction. *Hymenophore* lamellate, decurrent. *Lamellae* subdistant, anastomosing, shallowly intervenose, bright yellow (2A6), staining blue (then brown); edges eroded. *Stipe* 6–7 cm long, 1.1 mm wide, tapering downwards, strict or curved, dry; upper half longitudinally ribbed, pale yellow to light ochraceous (5B4, 4B4); lower half longitudinally ribbed or scurfy, white or buff tan; with age yellow (concolorous with the lamellae); base pale yellow, staining not present; interior solid; with age whitish yellow, with NH_4 no reaction. *Basal mycelium* yellow. *Fleeting-amyloid reaction* positive.

Basidiospores 8.4–9.8 µm long, 3.5–4.2 µm wide, mean Q = 2.36, subfusoid or fusoid; or 7–7.7 µm long, 4.2–4.9, µm wide, mean Q = 1.69, ovoid to subglobose, smooth, inamyloid, in KOH ochraceous. *Basidia* 28–30.8 µm long, 7–7.7 µm wide, clavate, hyaline or pale yellow (some), 4 -sterigmate. *Hymenial cystidia* 53.2–60.9 µm long, 7.7–8.4 µm wide, more common towards the edge of lamellae, thin walled, hyaline, clavate-ventricose or cylindric, encrusting pigment absent. *Hymenophoral trama* bilateral; hyphae cylindric, 7.7–10.5 µm wide, hyaline, inamyloid. *Pileipellis hyphae* a trichodermium, in KOH hyaline, inamyloid; elements (4.9–)6.3–8.4(–10.5) µm wide, cylindric, smooth, thin walled, granular content absent, intercalary cells cylindric. *Pileus trama* interwoven, hyphae light yellow, inamyloid, smooth, thin walled. *Stipitipellis hyphae* vertically oriented, parallel, giving rise to clusters of caulocystidia, 51.5–76.3 µm long (31.5–49), 8.4–25.2 µm wide (12.6–16), clavate or subfusoid (on the lower half) or sphaeropedunculate (on the upper half), hyaline, with incrusting pigment present (sometimes). *Stipe trama* hyphae parallel, cylindric, hyaline, inamyloid. *Clamp connections* absent.

MYCORRHIZAL HOST: *Castanopsis*.

DISTRIBUTION: Only known from northern Thailand.

The two different spore morphologies are evident in the collection and a very diagnostic character for this taxon. The different shape of the caulocystidia at the base versus the apex of the stipe, and the vinaceous red reaction with NH_4 on the pileus are also diagnostic. *Phylloporus bellus* produces spores in two size classes in some collections, but the caulocystidia are consistent in morphology. The vinaceous red reaction to NH_4 is not seen in *P. bellus*.

(14) **Phylloporus fibulatus** Singer, Ovrebo, & Halling, Mycologia 82: 452. 1990.

Figure 2-13, 2-32

Pileus 1.1–3.5 cm broad, at first convex, with age plane or plano-depressed, dry; disc even, at first yellow, then dull yellow; margin even to uneven, when young yellow, then brown; surface woolly, with KOH ochre, with NH_4 brown (deep purple brown at junction of pileus surface and trama). *Flesh* 4–5 mm thick, yellow, staining absent or slightly blue; odor fragrant; taste mild; with $FeSO_4$ olivaceous, with KOH ochre. *Hymenophore* lamellate to subporoid, decurrent. *Lamellae* subdistant, 2–3 mm wide, anastomosing (at half height or when young), shallowly intervenose, boletinoid (not distinctly), when young yellow, with age yellowish green to yellowish gray, staining absent; with $FeSO_4$ olivaceous, with KOH ochre. *Stipe* 1.5–4.5 cm long, 2.5–9 mm wide, equal or tapering downwards, sometimes eccentric or curved, dry; upper half slightly roughened-fibrillose, yellow (with reddish zone near apex); lower half slightly roughened-fibrillose, yellow; base pale yellow to yellow, staining not present; interior solid; flesh yellow, staining not present, with $FeSO_4$ olivaceous, with KOH ochre. *Basal mycelium* yellow to pale yellow. *Fleeting-amyloid reaction* positive.

Basidiospores brownish olive in mass, (8–)8.5–10(–11) µm long, 3–4(–4.5) µm wide, mean Q = 2.7, subfusoid, smooth, inamyloid, in KOH ochraceous. *Basidia* 23–41 µm long, 6–8.5 µm wide, narrowly clavate, hyaline to pale yellow, 4 -sterigmate. *Hymenial cystidia* (29–)50–104 µm long, 6–10.4 µm wide, thin walled, hyaline, cylindric with the mid-portion swollen to narrowly fusiform ventricose, with the apex sometimes subcapitate. *Hymenophoral trama* divergent, with age interwoven; hyphae cylindric, 3.5–14 µm wide, hyaline, in KOH yellow; subhymenial hyphae 2.8–3 µm wide. *Pileipellis hyphae* interwoven or a trichodermium, in KOH yellow; elements 3.5–9.2 µm wide, cylindric and elongated, thin walled, granular content absent or present. *Pileus trama* interwoven, hyphae hyaline to light yellow, inamyloid, with elements 3.5–17 µm wide, thin walled. *Stipitipellis hyphae* vertically oriented, interwoven, 3.5–5.8 µm wide, cylindric. *Stipe trama* hyphae parallel, cylindric, pale yellow, inamyloid, 4.6–17 µm wide. *Clamp connections* numerous.

MYCORRHIZAL HOST: *Quercus*.

DISTRIBUTION: This species was first described from Colombia and more collections have been there found since. It has not been found in other neotropical forests yet, although it is likely due to similar forest types and conditions seen elsewhere in the montane neotropics.

Diagnostic features include the abundance of clamp connections, the absence of a blue reaction to ammonia, and the subporoid hymenophore. *Phylloporus fibulatus* is distinguished from *Phylloporus colligatus*, described above, by the amount of clamp connections and the reaction of NH_4 in the pileus. In addition *P. fibulatus* has cylindric to fusiform cystidia and a more subporoid hymenophore, compared to the ventricose to subcapitate cystidia and lamellate hymenophore of *P. colligatus*. See comments under *P. foliiporus*, also a clamped species, to distinguish the two taxa. Another species with a very obvious subporoid hymenophore is *P. pumilus*, which is much smaller and lacks clamp connections. Singer et al. (1990) created Section Fibulati to accommodate *P. fibulatus*, however, a section with the same name had been erected earlier by Heinemann & Rammeloo (1986) to include *P. pseudopaxillus*, a clamped species from central Africa which differs from *P. fibulatus* by the simple lamellae and larger spores.

(15) **Phylloporus foliiporus** (Murrill) Singer, Persoonia 9: 424. 1978.

Figure 2-14, 2-29

Gomphidius foliiporus Murrill, Mycologia 35:432. 1943.

Phylloporus rhodoxanthus ssp. *foliiporus* (Murrill) Singer, Farlowia 2:280. 1945.

Pileus 1.8–14.5 cm broad, at first convex, with age plane or plano-depressed, dry, even; disc subtomentose,; margin even, when young dark reddish brown or maroon, then tan or ochraceous brown; surface subtomentose, becoming glabrous, with NH_4 blue green. *Flesh* white, staining blue (when fresh and young); odor mild; taste mild; with NH_4 no reaction. *Hymenophore* lamellate, decurrent. *Lamellae* subdistant, not anastomosing, intervenose, forked, when young bright yellow, with age yellowish brown, staining blue; edges even. *Stipe* 1.6–5.5 cm long, 2–40 mm wide, equal or tapering downwards, strict, dry; upper half when young finely subpruinose ridged, yellow, with age finely subpruinose ridged, grayish yellow; lower half when young floccose, olivaceous, grayish green; fibrils on upper half when young red brown, with age light brown, on lower half when young gray brown; base yellow, staining not present; interior solid; flesh when young whitish yellow, staining blue. *Basal mycelium* yellow, or pale yellow. *Fleeting-amyloid reaction* positive.

Basidiospores brownish olive, 10.5–12.6 µm long, 4.2–4.9 µm wide, mean Q = 2.54, subfusoid, smooth, amyloid, in KOH ochraceous. *Basidia* 32.2–35 µm long, 5.6–6.3 µm wide, clavate, hyaline, 4 -sterigmate. *Hymenial cystidia* 38.5–50.4 µm long, 11.9–14 µm wide, more common on sides of lamellae, thin walled, hyaline or honey colored contents (at the apex), fusoid or clavate, encrusting pigment present (at the apex). *Hymenophoral trama* bilateral; hyphae cylindric, 7–8.4 µm wide, hyaline, amyloid. *Pileipellis hyphae* a trichodermium (with appressed hyphae), in KOH pale yellow or hyaline, inamyloid; inflated, encrusted with pigment (brownish cytoplasmic content), thin walled, granular content absent. *Pileus trama* hyphae hyaline, inamyloid, thin walled. *Stipitipellis hyphae* vertically oriented, parallel, giving rise to clusters of caulocystidia, 42.7–44.1 µm long, 12.6–13.3 µm wide, clavate, yellow. *Stipe trama* hyphae parallel, cylindric, hyaline, inamyloid. *Clamp connections* present (rare, in the hymenophoral trama).

MYCORRHIZAL HOSTS: *Pinus*, *Quercus*.

DISTRIBUTION: The species is known from southern USA; it has been collected in Florida and was recently found in Alabama.

One of the most diagnostic characteristics in this taxon is the presence of hymenial cystidia with a melleous colored, sometimes constricted apex. Although Murrill did not mention the presence of cystidia, they are present in the type material. Singer (1945) described the presence of the cystidia, noting the colored apex but without emphasizing it as a diagnostic character. Other Singer collections deposited in FH possess the melleous cystidia.

This is the third of the clamped species included in this work. *Phylloporus fibulatus* has a larger number of clamp connections and lacks the melleous cystidia. *Phylloporus fibulatus* has smaller spores and the surface of the pileus turns brown when exposed to NH_4, compared to *P. foliiporus* that turns blue-green when exposed to NH_4. *Phylloporus colligatus* spores are also smaller than the ones of *P. foliiporus*, the NH4 reaction is blue at first but turns into a burgundy color, and the cystidia lack the melleous apex. The African *P. pseudopaxillus* have a larger number of clamp connections that are seen in all parts of the basidiome.

In 1978, Singer included *P. bellus* var. *cyanescens* as a synonym of *P. foliiporus*. These two taxa look alike but they do not reside in the same clade and show some molecular divergence. Morphologically, *P. bellus* var. *cyanescens* has longer cystidia and the melleous contents are mostly seen in the cheilocystidia but not in the pleurocystidia.

Phylloporus rhodoxanthus ssp. *foliiporus* was considered by Singer (1945) as a geographic race of *P. rhodoxanthus* and later (Singer 1978) was elevated to species rank. Phillips and Kibby in Phillips (1991), without citing Singer, also elevated it to the species level, creating a superfluous combination and status for this taxon.

(16) **Phylloporus guzmanii** Montoya & Band.-Muñoz, Mycotaxon 41: 473. 1991.

Figure 2-15, 2-30

Pileus 1.5–4.5 cm broad, at first convex, with age convex or plano-convex, dry; disc velutinous, at first vinaceous red or dark purple, then dark reddish brown (with yellowish tinges); margin undulated, yellowish brown; surface velutinous, becoming tomentose.

Flesh pale yellow, staining blue; odor mild; taste mild. *Hymenophore* lamellate, decurrent. *Lamellae* close or subdistant, not anastomosing, intervenose, when young bright yellow, with age mustard yellow, staining blue or reddish brown; edges even. *Stipe* 25–50 cm long (65), 4–8 mm wide, equal, strict, dry; upper half subfibrillose, purplish, brown; lower half subfibrillose, brown; base brown, staining not present; interior solid; flesh above when young pale yellow; flesh at base when young mustard yellow. *Basal mycelium* yellow. *Fleeting-amyloid reaction* positive (weakly).

Basidiospores (6.4–)7.2–8.8(–10.4) µm long, (3.2–)4–4.8(–5.6) µm wide, mean Q = 1.8, globose or ovoid, smooth, with SEM finely scrobiculate or bumpy inamyloid, in KOH pale yellow or greenish. *Basidia* 47–65.6 µm long, (5.6–)7.2–8 µm wide, clavate, hyaline, 4 -sterigmate. *Hymenial cystidia* (41.6–)45–105.6(–116.8) µm long, 5.6–11(–12) µm wide, moderately abundant, thin walled, hyaline, subfusoid or sublageniform. *Hymenophoral trama* bilateral; hyphae cylindric, (3.2–)4.8–10 µm wide, hyaline or yellowish. Subhymenial hyphae 2.4–3.2 µm wide. *Pileipellis hyphae* a trichodermium, in KOH yellowish, inamyloid; elements (20–)22–78(–80) µm wide, subclavate, smooth, thin walled. *Pileus trama* hyphae hyaline, smooth, thin walled. *Stipitipellis hyphae* vertically oriented, parallel, cylindric, yellow. *Stipe trama* hyphae parallel, cylindric, hyaline, inamyloid. *Clamp connections* absent.

MYCORRHIZAL HOSTS: *Pinus*, *Quercus*.

DISTRIBUTION: This species is known only from northwest Mexico.

This is another rare *Phylloporus* species with subglobose or ovoid spores. The collections were originally identified as *P. coccineus* Corner by Pérez-Ramirez et al.

(1986); however, the latter has broader pleurocystidia, larger cheilocystidia, and shorter epicutis elements.

(17) **Phylloporus infuscatus** Neves & Halling sp. nov.

Figure 2-16, 2-33

ETYMOLOGY: infuscatus (darkened), because of the somber color of the context

Pileus 2.7–3.2 cm broad, at first convex, with age plano-convex, dry, subrugulose; at first dark olive (4E6, 4F5), then olive-ochre to brown (5F5); with NH_4 vinaceous red. *Flesh* ochraceous brown or brown (6E4), staining absent; odor faint; taste mild; with NH_4 vinaceous red. *Hymenophore* lamellate, decurrent. *Lamellae* subdistant, not anastomosing, rarely intervenose, mostly simple, when young bright yellow (3A6), with age yellow, staining light blue; edges even. *Stipe* 3.4 cm long, 4–5 mm wide, equal, strict, dry; upper half when young pruinose, dark gray greenish (4B4) or yellow (pulverulent on the very top), with age finely pruinose, grayish green; lower half when young pruinose, pale yellow (5B3); pruina on upper half when young yellow; base pale yellow; interior solid; flesh above when young ochraceous brown or brown, staining not present, flesh at base when young yellow, staining not present. *Basal mycelium* white. *Fleeting-amyloid reaction* positive.

Basidiospores 6.3–7.7 µm long, 3.5–4.2 µm wide, mean Q = 1.81, oblong or ovoid, smooth, weakly amyloid, in KOH greenish or hyaline. *Basidia* 25.9–30.8 µm long, 6–7 µm wide clavate, hyaline, 4 -sterigmate. *Hymenial cystidia* 49–63 µm long, 10.5–13.3 µm wide, numerous on sides and edges of lamellae, thin walled, hyaline, fusoid or cylindric or clavate, encrusting pigment absent. *Hymenophoral trama* bilateral; hyphae cylindric. *Pileipellis hyphae* a trichodermium, in KOH tan (some); elements (7–)9.1–15.4 µm wide, in sphaerocyst-like chains, encrusted with pigment (red vinaceous exudate comes out when cuts are mounted in KOH), thin walled. *Stipitipellis hyphae* vertically oriented, parallel, giving rise to clusters of caulocystidia, 5.6–8.4 µm wide, subcylindric or cylindric (with pruina on the surface), yellow (in some hyphae), with incrusting pigment present (fine crystal-like incrustations at the surface). *Stipe trama* hyphae parallel, cylindric, hyaline, inamyloid. *Clamp connections* absent.

MYCORRHIZAL HOST: *Castanopsis*.

DISTRIBUTION: Collected only once in northern Thailand.

This taxon has unusual characteristics for a *Phylloporus*. The hymenophore features and the microscopic characteristics together with molecular data confirm placement in *Phylloporus*. The green color of the pileus is diagnostic and so is the dark color of the flesh, rather than the usual red-brown pilei and the light yellowish or whitish color of the flesh in the rest of the genus. It is important to note that the color of the flesh is not due to oxidation.

(18) **Phylloporus leucomycelinus** Singer, Persoonia 9: 426. 1978.

Figure 2-17, 2-29

Phylloporus leucomycelinus Singer & Gómez, Brenesia 22: 176. 1984. superfluous

Phylloporus rhodoxanthus ssp. *albomycelinus* Snell & Dick, Boleti of Northeastern North America p.47. 1970. nom. nud.

Phylloporus rhodoxanthus ssp. *leucomycelinus* Singer, Röhrlinge p.91. 1965. nom. nud.

Pileus 2.8–3.4 cm broad, at first pulvinate, with age convex, dry, becoming rivulose cracked; when young pale red brown; then deep red brown; surface subvelutinous, with NH_4 blue green. *Flesh* yellowish white, staining blue and bright yellow (in the lower part of the pileus) or cinnamon (immediately underneath the cuticle); odor mild; taste mild. *Hymenophore* lamellate, decurrent. *Lamellae* subclose or close, not anastomosing, intervenose, when young brownish yellow, with age yellowish brown, staining blue; edges even. *Stipe* 2.7–4.5 cm long, 3–5 mm wide, tapering downwards, strict or curved, dry; upper half when young punctate, red brown, with age punctate, pale brown; lower half when young punctate, pale brownish red; pruina on upper half when young red brown; base pallid, staining not present; interior solid; flesh above when young whitish yellow, staining bright yellow, flesh at base when young whitish yellow, staining not present. *Basal mycelium* white. *Fleeting-amyloid reaction* positive.

Basidiospores olive brown in mass, 10–12.5(–13.5) µm long, 3.3–4.5(–4.8) µm wide, mean Q = 2.88, subfusoid, smooth, dextrinoid, in KOH pale yellow. *Basidia* 20–25 µm long, 6–7 µm wide, ventricose, hyaline, 4 -sterigmate. *Hymenial cystidia* 50–71 µm long, (6–)11–12 µm wide, moderately abundant, thin walled, hyaline, ampullaceous, encrusting pigment absent. *Hymenophoral trama* bilateral; hyphae cylindric, melleous hyaline. *Pileipellis hyphae* a trichodermium, in KOH pale yellow or hyaline, inamyloid; elements 18–50 µm wide, cylindric, smooth, thin walled. *Pileus trama* hyphae hyaline, inamyloid, smooth, thin walled. *Stipitipellis hyphae* vertically oriented, parallel, giving rise to dermatocystidia or clusters of caulocystidia, 49.7–53.9 µm long, 8.4–11.2 µm wide, clavate, hyaline. *Stipe trama* hyphae parallel, cylindric, hyaline, inamyloid. *Clamp connections* absent.

MYCORRHIZAL HOSTS: *Fagus*, maybe *Quercus*, possibly *Eucalyptus* (in Australia).

DISTRIBUTION: This taxon has been reported for the eastern United States. Singer (1986) mentioned a collection from Canada that might be *P. leucomycelinus*, but he did not include the collection number and the specimen was not found in FH or in F.

This species has the same distribution range as *P. rhodoxanthus* in the eastern United States, and it has been confused with it, even though the two species can be easily set apart by the white color (versus yellow) of the basal mycelium in *P. leucomycelinus*. Since *P. rhodoxanthus* was the only known species in the USA until Singer validly published *P. leucomycelinus*, many collections had been identified as a "white basal mycelium form" of *P. rhodoxanthus*. Diagnostic characteristics of this taxon include the non-cyanescent flesh and the white basal mycelium.

Phylloporus leucomycelinus is mostly cited as occurring under fagaceous forests. It is possible that it is associated with *Quercus*, since it has been collected around *Quercus* trees. Watling and Gregory (1991) collected in Australia what they considered to be a *P. leucomycelinus* in a *Eucalyptus* forest, however it is likely that the Australian collection belongs to a different taxon.

This taxon was mentioned for the first time by Singer (1945) as a variety of *P. rhodoxanthus* ssp. *americanus* with white basal mycelium. Later, Singer (1965) used the name *P. rhodoxanthus* spp. *leucomycelinus* for the first time in a key, but no Latin diagnosis and no type were included. In 1970, Snell & Dick published *P. rhodoxanthus* ssp. *albomycelinus* based on Singer's (1945) notes, but this also is considered a *nomen nudum* because no type or Latin diagnosis were included. The legitimate publication of the species was made by Singer (1978) when he published the taxon as *P. leucomycelinus*

(Singer) ex Singer even though it was not a new combination. In 1984, the species was again published as new by Singer and Gómez (1984), using the same type. They did not cite Singer's 1978 publication but incorrectly cited *P. rhodoxanthus* ssp. *leucomycelinus* Sing. apud Snell & Dick as a synonym.

(19) **Phylloporus novae-zelandiae** McNabb, New Zealand J. Bot., 9: 358. 1971.

Figure 2-18, 2-34

Macroscopic data from the protologue:

Pileus 2.5–9.5 cm broad, at first plano-convex, with age plano-depressed, dry, even; disc finely felted, at first dark brown, then cocoa brown or yellowish brown; margin inrolled or finely creviced; surface finely felted, with NH_4 red or dark. *Flesh* pale yellow, staining absent; odor mild; taste mild; with NH_4 no reaction. *Hymenophore* lamellate, decurrent. *Lamellae* subdistant, anastomosing, intervenose, when young bright yellow, with age yellow, staining blue green; edges blunt. *Stipe* 2.5–5.5 cm long, 5–15 mm wide, subbulbous, strict, dry; upper half when young finely felted, pale brown or brown, with age finely felted, pale yellow; lower half when young felted, pale yellow; base pale yellow, staining not present; interior solid; flesh above when young pale brownish white, staining not present. *Basal mycelium* yellow. *Fleeting-amyloid reaction* positive.

Basidiospores olive brown in mass, 9.1–12.6 µm long, 3.5–4.9 µm wide, mean Q = 2.58, subfusoid, smooth, on SEM smooth to finely rugulose, inamyloid, in KOH melleous. *Basidia* 33–45 µm long, 8–10 µm wide, clavate, hyaline, 4 -sterigmate. *Hymenial cystidia* 50–75 µm long, 9.5–16.5 µm wide, moderately abundant (scattered), thin walled, hyaline, ventricose rostrate, encrusting pigment absent. *Hymenophoral trama* bilateral; hyphae cylindric, hyaline, inamyloid. *Pileipellis hyphae* a trichodermium, in

KOH brownish, inamyloid; elements 12–18 μm wide, cylindric or subclavate, smooth, thin walled, granular content absent. *Pileus trama* hyphae hyaline, smooth, thin walled. *Stipitipellis hyphae* vertically oriented, parallel, subcylindric or cylindric. *Stipe trama* hyphae parallel, cylindric, hyaline, inamyloid. *Clamp connections* absent.

MYCORRHIZAL HOST: *Nothofagus*.

DISTRIBUTION: This taxon is known from New Zealand, where it has been collected a few times in Karameae, on the west coast.

Diagnostic characteristics for this species include the unchanging context, a slow bluing of the damaged lamellae, and the absence of reaction with NH_4 on the pileus.

(20) **Phylloporus orientalis** var. **brevisporus** Corner, Nova Hedwigia 20: 809. 1970.

Figure 2-19, 2-34

Pileus 3.5–8 cm broad, at first plano-convex or plane, with age uplifted, dry, even, becoming areolate; disc subtomentose, at first cinnamon brown, then brownish red; margin even; surface matted subtomentose, with NH_4 blue green or dull lilac. *Flesh* white, staining blue; odor mild; taste mild; with NH_4 blue. *Hymenophore* lamellate, adnexed or subdecurrent. *Lamellae* subdistant, not anastomosing, intervenose, when young yellow, with age mustard yellow, staining blue; edges even. *Stipe* 4–8 cm long, (6–)10–15 mm wide, equal or tapering downwards, strict, dry; upper half when young subpruinose, white or pale yellow; lower half when young matted subpruinose, brown, with age subpruinose, pale brown; pruina on upper half when young cinnamon; base white, staining not present; interior solid; flesh above when young white, with age pale brown pinkish, staining not present, with NH_4 no reaction. *Basal mycelium* white. *Fleeting-amyloid reaction* positive.

Basidiospores 10.5–11.9 µm long, 3.5–4.9 µm wide, mean Q = 2.67, subfusoid, smooth, amyloid, in KOH pale yellow. *Basidia* 25.9–32.2 µm long, 7.7–9.1 µm wide, clavate, hyaline, 4 -sterigmate. *Hymenial cystidia* (44–)52.2–79.8 µm long, 10.5–13.3(–17.5) µm wide, numerous on sides and edges of lamellae, thin walled, hyaline, fusoid to lanceolate or cylindric, encrusting pigment absent or present (rare). *Hymenophoral trama* divergent; hyphae cylindric, 8.4–9.8 µm wide, yellowish, dextrinoid. *Pileipellis hyphae* hymeniform or an ixotrichodermium, in KOH brownish, inamyloid; elements 6.3–7.7 µm wide, cylindric, smooth, thin walled, granular content absent. *Pileus trama* hyphae ochraceous, inamyloid, smooth, thin walled. *Stipitipellis hyphae* vertically oriented, parallel, giving rise to dermatocystidia or clusters of caulocystidia, 20.3–27.3 µm long, 7–9.8 µm wide, cylindric or clavate, yellow brown contents. *Stipe trama* hyphae parallel or compacted, cylindric, hyaline, inamyloid. *Clamp connections* absent.

MYCORRHIZAL HOSTS: *Eucalyptus*, *Acacia*, *Allocasuarina*.

DISTRIBUTION: The species is known from northern Borneo and Australia.

Australian material was considered by Watling & Gregory (1991) to be closely related to *P. foliiporus* but differs because of the cystidia that lack the melleous color. The large size of the basidiomata and the blue or lilac reaction to NH_4 in the pileus are distinct characters for this taxon. Watling & Gregory (1991) described *P. orientalis* but noted that, due to the spore size, their collections from the Cooloola Sandmass possibly belong to var. *brevisporus*. They mentioned a negative Imler's reaction for the collections they examined. The basidia described here are smaller than the ones described by Corner (1970) from northern Borneo and Watling & Gregory (1991) from Australia. However, the spore size also corresponds to the var. *brevisporus*.

(21) **Phylloporus phaeoxanthus var. phaeoxanthus** Singer & L.D. Gómez, Brenesia 22: 171. 1984.

Figure 2-20, 2-29, 2-31, 2-32

Pileus 2.5–4 cm broad, at first plano-convex, with age plane, dry; disc even, vinaceous brown (8F5); margin even, brown (6D7, 6D6); surface tomentose, with NH_4 blue green or dull lilac (eventually). *Flesh* 4–10 mm thick, white, staining absent; odor mild; taste mild; with NH_4 blue green or dull lilac. *Hymenophore* lamellate, decurrent. *Lamellae* close, 1–8 mm wide, anastomosing to subporoid, yellow (3A7), staining absent; edges even. *Stipe* 2–3 cm long, 6–8 mm wide, equal to tapering downwards, curved, dry; upper half when young finely pruinose, pale brown (6D6) to brown (7E7); lower half when young finely pruinose, pale brown to brown; base pale yellow to yellow, staining not present; interior solid; flesh above white to whitish yellow to yellow, staining not present, flesh at base white to yellow, staining not present. *Basal mycelium* yellow. *Fleeting-amyloid reaction* positive (weakly).

Basidiospores (6.5–)8.5–12.3(–14) µm long, (3.3–)4.3–4.8 µm wide, mean Q = 2.84, subfusoid, smooth, with SEM finely bacillate or bumpy, inamyloid, in KOH light brown melleous. *Basidia* 23–40 µm long, 6–9 µm wide, clavate, hyaline, 4 -sterigmate. *Hymenial cystidia* 35–90 µm long, 9–20 µm wide, moderately abundant, thin walled, hyaline, ventricose to fusoid to utriform, encrusting pigment present. *Hymenophoral trama* bilateral; up to 15 µm wide, hyaline, in KOH finely melleous ochre. *Pileipellis hyphae* a trichodermium and forming a palisade, in KOH yellow, inamyloid; elements 6–12 µm wide, elongated, smooth, thick walled. Intercalary cells subisodiametric to isodiametric, 10–12 µm long, 8–10 µm wide. *Pileus trama* interwoven, hyphae hyaline, inamyloid. *Stipitipellis hyphae* palisadic, with short cells arranged in chains, vertically oriented, parallel to interwoven, giving rise to clusters of caulocystidia, 15–45 µm long, 7–13 µm wide, ventricose to fusoid, with incrusting pigment sometimes present. *Stipe trama* hyphae parallel to subparallel, cylindric, hyaline, inamyloid. *Clamp connections* absent.

MYCORRHIZAL HOST: *Quercus*.

DISTRIBUTION: This species is known from Costa Rica, Colombia and Mexico.

(22) **Phylloporus phaeoxanthus var. simplex** Singer & L.D. Gómez, Brenesia 22: 172. 1984.

Figure 2-21, 2-31

Pileus 2–6.5 cm broad, at first plano-convex, with age plane, dry; vinaceous brown (8F5) to light brown; margin even, brown (6D7, 6D6); surface finely scaly, scales dark brown, with NH_4 blue green. *Flesh* white, staining absent; odor mild; taste mild; with NH_4 blue. *Hymenophore* lamellate, decurrent. *Lamellae* close, with venations, bright yellow (3A8), staining absent; edges even. *Stipe* 3–4.5 cm long, 5–8 mm wide, equal to tapering downwards, dry; upper half when young finely pruinose, pale brown (6D6); lower half when young finely pruinose, pale brown to brown; base pale yellow to yellow, staining not present; interior solid; flesh above white to whitish yellow to yellow, staining not present, flesh at base white to yellow, staining not present. *Basal mycelium* yellow. *Fleeting-amyloid reaction* positive (weakly).

Basidiospores 8.5–12.3(–14) µm long, 4.3–4.8 µm wide, mean Q = 2.84, subfusoid, smooth, with SEM finely bacillate or bumpy, dextrinoid, in KOH light brown melleous. *Basidia* 23–40 µm long, 6–9 µm wide, clavate, hyaline, 4 -sterigmate. *Hymenial cystidia*

30–101.5 µm long, 9–28 µm wide, moderately abundant, thin walled, hyaline, versiform, encrusting pigment absent. *Hymenophoral trama* bilateral; hyaline, in KOH finely melleous ochre. *Pileipellis hyphae* a trichodermium and forming a palisade, in KOH yellow, inamyloid. *Pileus trama* interwoven, hyphae hyaline, inamyloid. *Stipitipellis hyphae* palisadic, with short cells arranged in chains, vertically oriented, parallel to interwoven, giving rise to clusters of caulocystidia, 15–45 µm long, 7–13 µm wide, ventricose to fusoid. *Stipe trama* hyphae parallel to subparallel, cylindric, hyaline, inamyloid. *Clamp connections* absent.

MYCORRHIZAL HOST: *Quercus*.

DISTRIBUTION: Variety *simplex* is more common than var. *phaeoxanthus* in Costa Rica.

The yellow mycelium at the stipe base, brown pileus, unchanging context, palisadic arrangement of hyphal elements in the pileipellis, the cells arranged in chains in the stipitipellis, and blue ammonia reaction are diagnostic. Variety *simplex* is a known variant that differs from var. *phaeoxanthus* because it has dextrinoid basidiospores and hymenial cystidia that are thin walled, unencrusted, larger, and versiform with a really wide range in size. The molecular phylogeny indicates a close relationship among *P. phaeoxanthus* (both varieties), *P. arenicola*, and *P. rhodoxanthus*; the latter two North American species that also have yellow basal mycelium. *Phylloporus arenicola* differs by a reddish to purple reaction with NH_4, and has a ribbed upper part of the stipe and cystidia that are non-encrusted and shorter than the cystidia of *P. phaeoxanthus*.

(23) **Phylloporus pumilus** Neves & Halling sp. nov.

Figure 2-22, 2-33

ETYMOLOGY: pumilus, smaller than other species in the genus

Pileus 0.5–0.9 cm broad, at first convex or plano-convex, with age plano-convex, dry, even; disc subtomentose, at first cocoa brown or dark brown (areolate tufts, with white between); margin smooth; becoming matted tomentose, with NH_4 dark or blue (around the drop). *Flesh* white, staining absent; odor mild. *Hymenophore* alveolate, decurrent. Tubes dull yellow, becoming wax yellow; then concolor with tubes, unchanging when injured. *Stipe* 0.7–1 cm long, less than 1 mm wide, equal, strict or curved, dry; upper half when young subpruinose, pinkish brown. *Basal mycelium* white. *Fleeting-amyloid reaction* positive (weakly).

Basidiospores 10.5–11.9 µm long, 3.5–4.9 µm wide, mean Q = 2.67, subfusoid, smooth, with SEM smooth, in KOH greenish or melleous. *Basidia* 17.5–23.1 µm long, 6.3–7 µm wide, clavate, hyaline, 4 -sterigmate. *Hymenial cystidia* 52.5–63 µm long, 5.6–11.2 µm wide (4.2 on the top when lanceolate), numerous on sides and edges of lamellae (abundant), thin walled, hyaline, fusoid or lanceolate, encrusting pigment absent. *Hymenophoral trama* bilateral; 3.5–4.9(–5.6) µm wide, hyaline. *Pileipellis hyphae* a trichodermium, dextrinoid; cylindric, smooth, thin walled. *Stipitipellis hyphae* vertically oriented, parallel, giving rise to clusters of caulocystidia (some capitate), 3.5–4.9 µm wide, cylindric. *Stipe trama* hyphae parallel. *Clamp connections* absent.

MYCORRHIZAL HOST: *Dipterocarpus*.

DISTRIBUTION: Two collections of this taxon were gathered in Indonesia (Java) and are only known from the type locality.

This diminutive species is very distinct due to the clearly alveolate hymenophore and the small size of the basidiomes, which fruited abundantly on the soil substrate.

(24) **Phylloporus rhodoxanthus** (Schwein.) Bres., Fung. Trident. 2(14): 95. 1900.

Figure 2-23, 2-30

Agaricus rhodoxanthus Schwein., Schr. Naturf. Ges. Leipzig 1: 83. 1822.

Paxillus flavidus Berk, London J. Bot. 6:315. 1847.

Gomphidius rhodoxanthus (Schwein.) Sacc., Syll. Fung. 5: 1887.

Phylloporus rhodoxanthus ssp. *americanus* Singer, Rev. Mycol. (Paris) 3: 171. 1938.

Pileus (2.5–)6–7.5 cm broad, at first obtusely convex or convex or subumbonate, with age subdepressed, dry, even; disc even, at first tawny olivaceous; when young cinnamon brown; surface subtomentose or tomentose, becoming fibrillose tomentose, with NH_4 blue. *Flesh* buff, staining absent; odor mild; taste mild. *Hymenophore* lamellate, decurrent. *Lamellae* subclose or close, not anastomosing, intervenose, when young deep yellow, with age yellowish brown, staining absent; edges even. *Stipe* 3.3–4.1 cm long, 5–8 mm wide, equal, strict, dry; upper half when young pustulate, yellow, with age longitudinally ridged, yellow; base pale yellow or yellow, staining not present; interior solid, or soon hollow; flesh above when young buff, flesh at base when young buff, staining cinnamon. *Basal mycelium* yellow. *Fleeting-amyloid reaction* positive.

Basidiospores 10–12(–14.5) µm long, 3.5–4.5(–5.5) µm wide, mean Q = 2.7, subfusoid, smooth, with SEM bacillate, inamyloid, in KOH melleous. *Basidia* 28–37 µm long, (6–)6.8–9 µm wide, clavate, hyaline, 4 -sterigmate. *Hymenial cystidia* 40–95 µm long, 9–27 µm wide, numerous on sides and edges of tubes, thin walled, hyaline, ventricose or clavate-ventricose, encrusting pigment present (scarcely in a few). *Hymenophoral trama* bilateral; hyphae cylindric, hyaline, inamyloid. *Pileipellis hyphae* a trichodermium, inamyloid; elongated, encrusted with pigment, thin walled, granular content absent. Intercalary cells subisodiametric, 16 µm long, 14 µm wide. *Stipitipellis hyphae* vertically oriented, parallel, giving rise to dermatocystidia (clavate to ventricose), cylindric. *Stipe trama* hyphae parallel, cylindric, hyaline, inamyloid. *Clamp connections* absent.

MYCORRHIZAL HOSTS: *Quercus*, *Fagus*, *Pinus*.

DISTRIBUTION: This species is widely distributed in oak and beech forests in the USA.

The species was originally described from a collection in North Carolina but there is no information on the location of the type specimen. Representative material deposited in FH as *Phylloporus rhodoxanthus* ssp. *americanus* is here designated as the neotype: Singer, July 22 – August 25, 1946, near Mountain Lake, Giles Co., Virginia, under pines (*Pinus rigida*), no collection number [as *P. rhodoxanthus* (Schwein.) Bres. ssp *americanus* Sing. (type ssp.)].

This is the most widely used name in *Phylloporus*. The taxon as it was apparently conceived, and now recognized, occurs only in the eastern United States along with other taxa. The species has a yellow basal mycelium, the flesh and other parts of the basidiome do not stain blue, and the application of NH_4 on the pileus surface gives an immediate blue reaction. The species concept is bound up in a complicated history, including the synonymy with *P. pelletieri* and the creation of a complex that included a number of subspecies: *americanus*, *bogoriensis*, *europeaus*, *foliiporus*, *leucomycelinus*, and *sulcatus*. These have all been elevated and are easily recognized at the species level.

(25) **Phylloporus rubiginosus** Neves & Halling sp. nov.

Figure 2-24, 2-33

ETYMOLOGY: rubiginosus (reddish with metallic tinge), with regard to the color of the pileus and the stipe

Pileus 3.3–6.1 cm broad, at first convex or plano-convex, with age applanate, dry, even, becoming cracked; when young dark reddish brown (6D6, 6E6) or deep red brown, then yellow orange (7F6, 8E7); surface squamulose, becoming subsquamulose (fracturing in some parts where the yellow flesh can be seen), with NH_4 blue. *Flesh* pale yellow, staining blue (sometimes slowly, after 30 seconds); odor mild; taste inconspicuous. *Hymenophore* lamellate, decurrent (separating from the stipe when older). *Lamellae* close or subdistant, not anastomosing, shallowly intervenose, when young yellow (4A6, 4B5), with age orangish yellow, staining blue; edges even. *Stipe* 3.5–5.5 cm long, 2–3 mm wide, equal, curved, dry; upper half when young finely pruinose, red brown, with age longitudinally ribbed; lower half when young finely pruinose (towards the middle, the pruina become smaller), pale brownish red (8E7); interior solid; flesh above when young pale yellow, staining blue, flesh at base when young yellow. *Basal mycelium* yellow. *Fleeting-amyloid reaction* positive.

Basidiospores 9.8–11.2 µm long, 3.5–4.9 µm wide, mean Q = 2.5, subfusoid, smooth, weakly dextrinoid, in KOH straw yellow. *Basidia* 23.8–24.5 µm long, 7–8.4 µm wide, clavate, hyaline, 4 -sterigmate. *Hymenial cystidia* 88.9–100.8 µm long, 9.8–11.2 µm wide, numerous on sides and edges of lamellae, thick walled (1-2 µm), hyaline, fusoid or cylindric, encrusting pigment absent. *Hymenophoral trama* divergent; hyphae cylindric, 5.6–8.4 µm wide, hyaline. *Pileipellis hyphae* a trichodermium, in KOH brownish, inamyloid; elements (4.2–)5.6–7.7 µm wide, cylindric, encrusted with pigment (brown exudate when cuts are put in KOH), thin walled. *Pileus trama* interwoven, hyphae light yellow. *Stipitipellis hyphae* vertically oriented, parallel, giving rise to clusters of caulocystidia, 32.9–41.3 µm long, 9.8–15.4 µm wide, clavate or sinuous clavate, yellow brown contents, with incrusting pigment present (brown exudate is released when cuts are immersed in KOH). *Stipe trama* hyphae parallel, cylindric, pale yellow. *Clamp connections* absent.

MYCORRHIZAL HOSTS: *Castanopsis, Dipterocarpus*.

DISTRIBUTION: This taxon is only known from two collections in northern Thailand.

The dark red color of the pileus and stipe is notable. Other diagnostic characters include the blue reaction with the application of NH_4, the slowly cyanescent flesh, and the thick walled hymenial cystidia. The caulocystidia at the apex of the stipe tend to be more strictly clavate, while the ones at the middle and base are sinuous clavate.

(26) **Phylloporus scabripes** Ortiz & Neves, Fung. Diversity, in press.

Figure 2-25, 2-30

Pileus 2.8–6.5(–7.5) cm broad, convex, dry, even, becoming tomentose; at first pale red brown, then tan; margin inrolled; surface tomentose, with NH_4 bright green (then slowly vinaceous lilac). *Flesh* 10 mm thick, yellowish white or yellow, staining absent; odor not distinctive; taste mild; with NH_4 lilac just under the pileipellis. *Hymenophore* lamellate, decurrent. *Lamellae* close, 7 mm wide, simple, when young yellow, with age olive, staining reddish brown in a few seconds; edges even. *Stipe* 3–5(–6) cm long, 7–15 mm wide, equal, strict, dry; upper half when young squamulose, pale yellow, light cinnamon or red brown; lower half pale yellow, with age squamulose; scabers on upper half when young red brown; base pale yellow, staining not present; interior solid; flesh whitish yellow, staining not present. *Basal mycelium* bright yellow. *Fleeting-amyloid reaction* positive.

Basidiospores 9.8–12.6 µm long, 3.5–4.9 µm wide, mean Q = 2.67, subfusoid, smooth, slightly dextrinoid, in KOH straw yellow or olive-hyaline. *Basidia* 33.6–38.5 µm long, 8.4–9.8 µm wide, cylindric, hyaline, 4 -sterigmate. *Hymenial cystidia* 42.7–79.1 µm long, 12.6–14.7 µm wide, more common towards the edge of lamellae, thin walled, hyaline, cylindric or clavate, encrusting pigment absent. *Hymenophoral trama* bilateral; hyphae cylindric, hyaline, amyloid. *Pileipellis hyphae* a trichodermium, in KOH yellow ochraceous, inamyloid; smooth, thin walled. *Pileus trama* interwoven, hyphae ochraceous (pigment dissolving in KOH), inamyloid, thin walled. *Stipitipellis hyphae* vertically oriented, parallel, giving rise to versiform caulocystidia, 16.1–80.5 µm long, 7–18.9 µm wide, fusoid or clavate or ventricose, hyaline. *Stipe trama* hyphae parallel, cylindric, inamyloid. *Clamp connections* absent.

MYCORRHIZAL HOSTS: *Pinus*, *Quercus*.

DISTRIBUTION: This is a new species described from Belize and so far only known from that country.

This species is distinctive because of the orangish pileus surface that becomes greenish blue with NH_4, and the reddish scabers on the upper surface of the yellow stipe.

(27) **Phylloporus** sp. 1 sensu Watling & N.M. Greg., Edinburgh J. Bot. 48: 379. 1991.

Figure 2-26, 2-34

Pileus 1.7–2 cm broad, at first convex or plano-convex, dry, even; margin even, when young brown; surface areolate, with NH_4 no reaction. *Flesh* white, staining pale pink; odor mild; taste mild; with NH_4 no reaction. *Hymenophore* lamellate, subdecurrent. *Lamellae* close or subdistant, not anastomosing, sparsely intervenose, mostly simple, when young yellow or light yellow, with age yellow, staining absent; edges even. *Stipe* 0.8–1 cm long, 2–3 mm wide, equal, strict or curved, dry; upper half when young pruinose, white; lower half when young pruinose, pallid; pruina on upper half when young pale brown; base white, staining not present; interior solid; flesh above when young white, staining not present, flesh at base when young white, staining not present, with NH_4 no reaction. *Basal mycelium* white. *Fleeting-amyloid reaction* positive.

Basidiospores 11.2–13.3 µm long, 4.2–4.9 µm wide, mean Q = 2.69, subfusoid, smooth, slightly amyloid, in KOH pale yellow. *Basidia* 24.5–32.9 µm long, 8.4–9.8 µm wide, clavate, pale yellow, 4 -sterigmate. *Hymenial cystidia* 51.8–71.4 µm long, 12.6–15.4 µm wide, numerous on sides and edges of lamellae, thin walled, hyaline, fusoid or swollen or clavate, encrusting pigment present (granular cytoplasmic content). *Hymenophoral trama* divergent, with age bilateral (towards the margin of the lamellae); hyphae cylindric or laticiferous-like, 8.4–10.5 µm wide, hyaline or yellowish, inamyloid. *Pileipellis hyphae* a trichodermium, in KOH yellow ochraceous, inamyloid; cylindric, smooth, thin walled, granular content present. *Pileus trama* hyphae hyaline, inamyloid, smooth. *Stipitipellis hyphae* vertically oriented, parallel, giving rise to clusters of caulocystidia, 39.2–40.6 µm long, 15.4–16.1 µm wide, clavate, light yellow. *Stipe trama* hyphae parallel, cylindric. *Clamp connections* absent.

MYCORRHIZAL HOST: *Acacia*.

DISTRIBUTION: Two collections are known from Australia.

Watling & Gregory (1991) published a description of an unidentified taxon the description of which agrees with another recent collection from Queensland, Australia. Referred to as *Phylloporus* sp. 1 in one of the papers on Cooloola boletes from Queensland, the collection has not been located after a dedicated search in BRI. This is a fairly small fungus, and the diagnostic characteristics include the brownish cap that does not turn blue with NH_4, the white flesh that stains pale pink when exposed, and the large number of pigmented, encrusted cystidia.

(28) **Phylloporus** sp. 2

Figure 2-27, 2-31

Pileus 2–2.25 cm broad, at first convex, with age plano-convex or plane, dry, entire; disc finely scaly, at first dark vinaceous brown, then violet brown (10F4); margin inrolled, when young violet brown; surface scaly, becoming crenate, with NH_4 no reaction. *Flesh* yellowish white, staining blue; odor mild; taste mild; with NH_4 light pink (under pileipellis). *Hymenophore* lamellate, decurrent. *Lamellae* subdistant, simple, when young bright yellow (3A6), with age yellowish green, staining light blue; edges even. *Stipe* 1.5–2.5 cm long, 4 mm wide, equal, curved, dry; brown (7E4), with age finely squamulose, pale brown; base yellow, staining not present; interior solid; flesh when young whitish yellow (2A2), with NH_4 blue. *Basal mycelium* yellow. *Fleeting-amyloid reaction* positive.

Basidiospores 9.1–10.5 µm long, 2.8–3.5 µm wide, mean Q = 3.1, subfusoid or cylindric, smooth, slightly amyloid, in KOH light brown melleous. *Basidia* 23.1–25.2 µm long, 5.6–7 µm wide, clavate, hyaline, 2–4 -sterigmate. *Hymenial cystidia* 63.7–70 µm long, (8.4–)10.5–18.2 µm wide, more common towards the edge of lamellae, thin walled, hyaline, subfusoid or fusoid or ventricose, encrusting pigment present. *Hymenophoral trama* bilateral; hyphae cylindric, 6.3–7.7 µm wide, hyaline, inamyloid. *Pileipellis hyphae* a trichodermium, in KOH yellow ochraceous or brownish, inamyloid; elements 6.3–14 µm wide, inflated or cylindric, smooth, thick walled, granular content absent. *Pileus trama* interwoven, hyphae hyaline, inamyloid, smooth, thin walled. *Stipitipellis hyphae* vertically oriented, parallel, giving rise to chains of short cells (thick walled), 9.8–20.3 µm long, 8.4–9.1 µm wide, subcylindric, yellow brown contents. *Stipe trama* hyphae parallel, cylindric, hyaline, inamyloid. *Clamp connections* absent.

MYCORRHIZAL HOST: *Quercus*.

DISTRIBUTION: This collection was found in Costa Rica in Puntarenas province and so far is the only one known.

The collection is composed of only one basidiome. The stipitipellis with chains of cells is similar to the ones observed in *P. phaeoxanthus*, but with thicker walls. It is included as a distinct species and keyed out separately because of this distinctive feature and the absence of a NH4 reaction on the pileus.

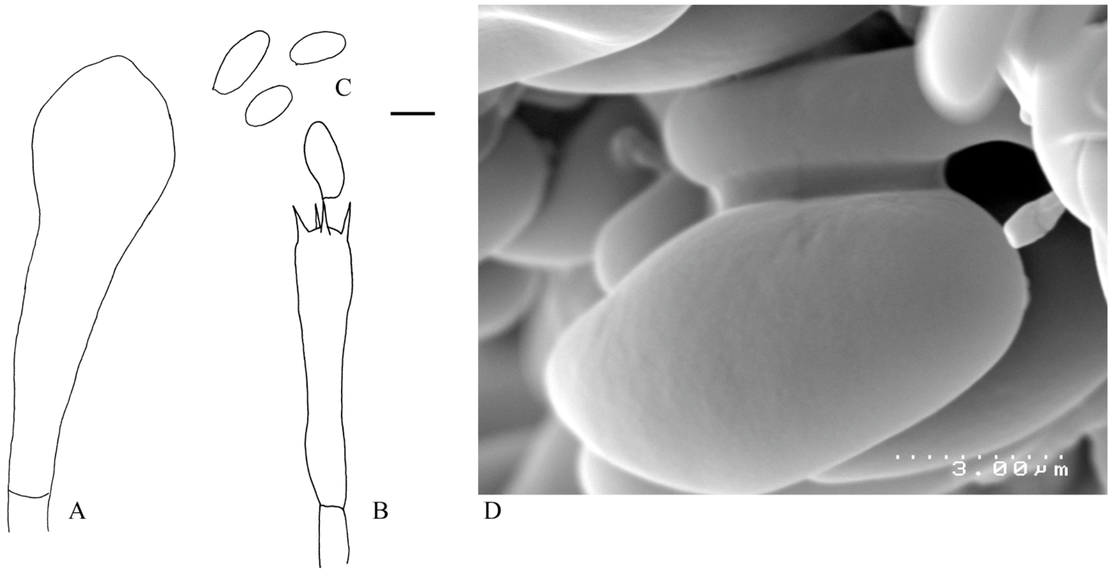

Figure 2-1: *Phylloporus pelletieri*. **A.** Pleurocystidium; **B.** Basidium; **C.** Spores; **D.** SEM of spores. (A–C: Scale bar = 10µm.)

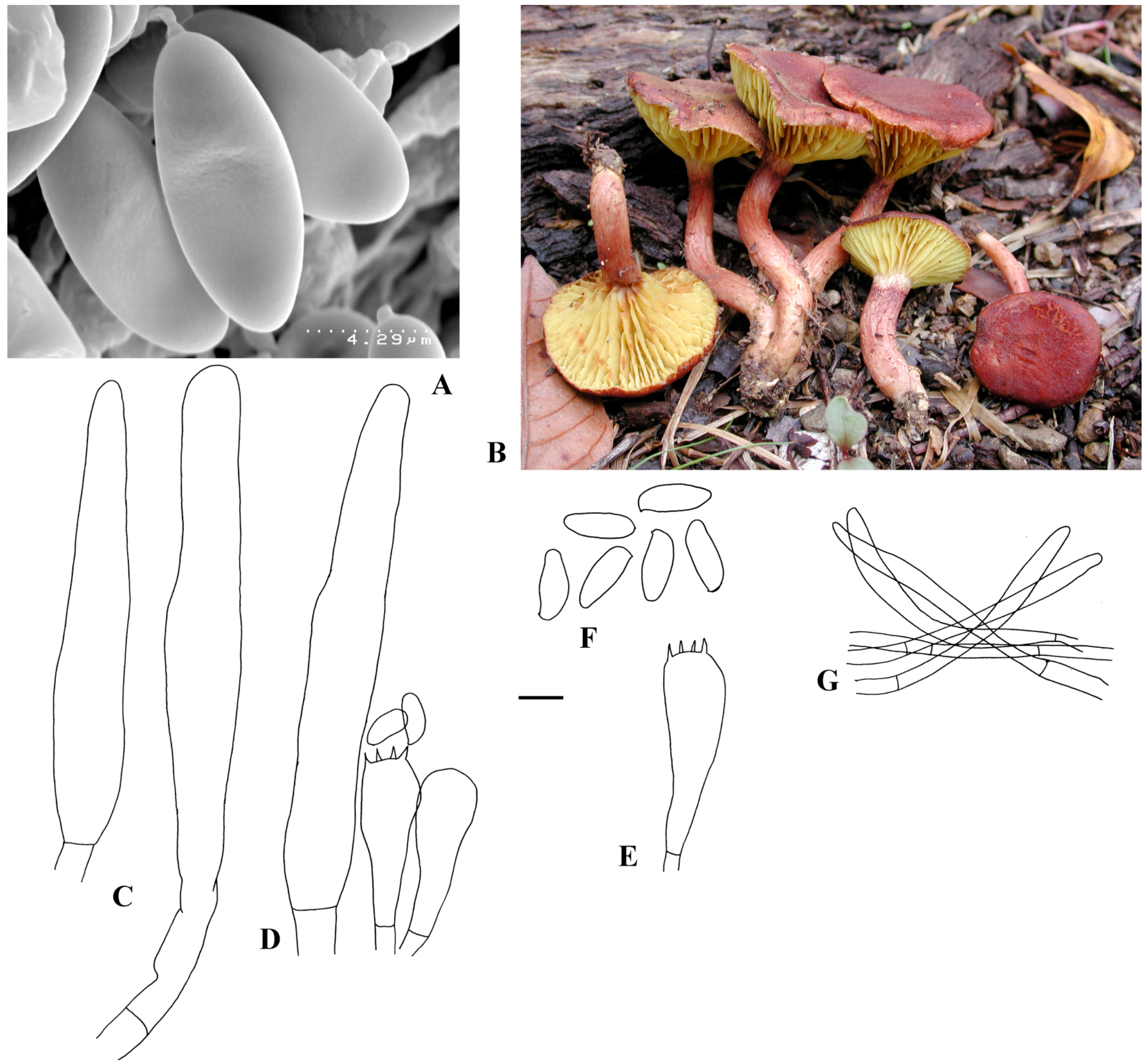

Figure 2-2: *Phylloporus alborufus*. **A.** Scanning electron microscopy of spores. **B.** Basidiomata (MAN22). **C.** Pleurocystidia. **D.** Pleurocystidium, basidium, and basidiole. **E.** Basidium. **F.** Spores. **G.** Stipitipellis hyphae. (C–G: Scale bar = 10µm.)

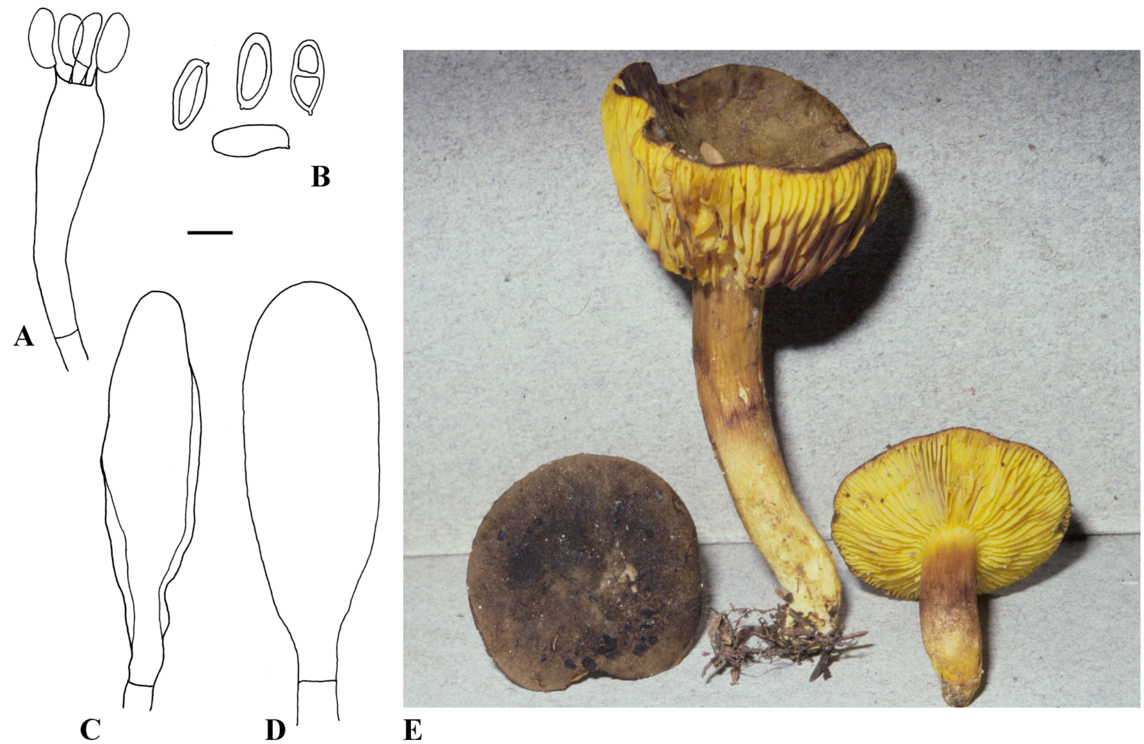

Figure 2-3: *Phylloporus arenicola*. **A.** Basidium. **B.** Spores. **C.** Pleurocystidium. **D.** Caulocystidium. **E.** Basidiomata (REH6951), photograph by R.E. Halling. (A–D: Scale bar = 10μm.)

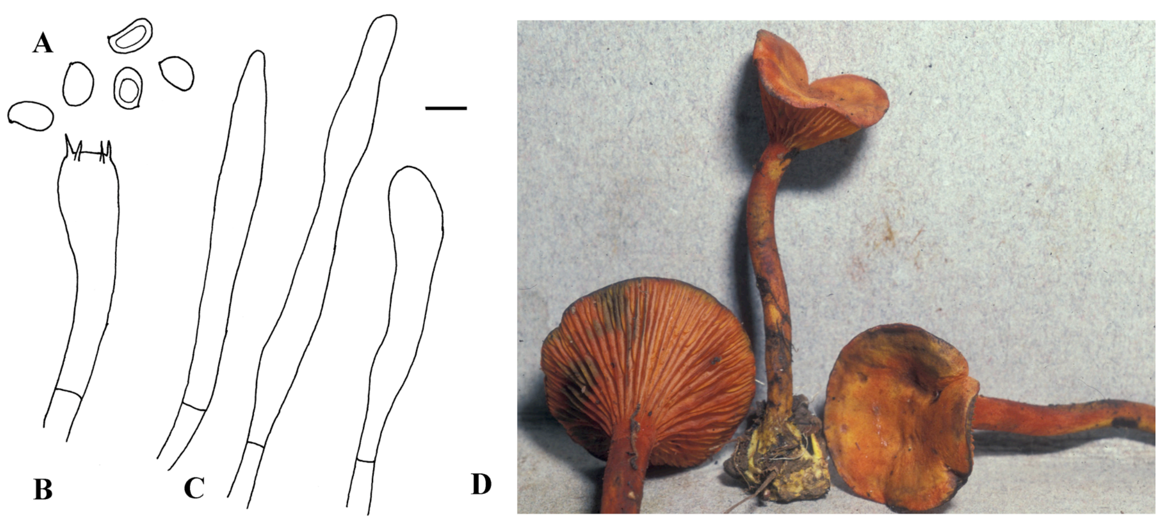

Figure 2-4: *Phylloporus aurantiacus*. **A.** Spores. **B.** Basidium. **C.** Pleurocystidia. **D.** Basidiomata (REH7271), photograph by R.E. Halling. (A–C: Scale bar = 10μm.)

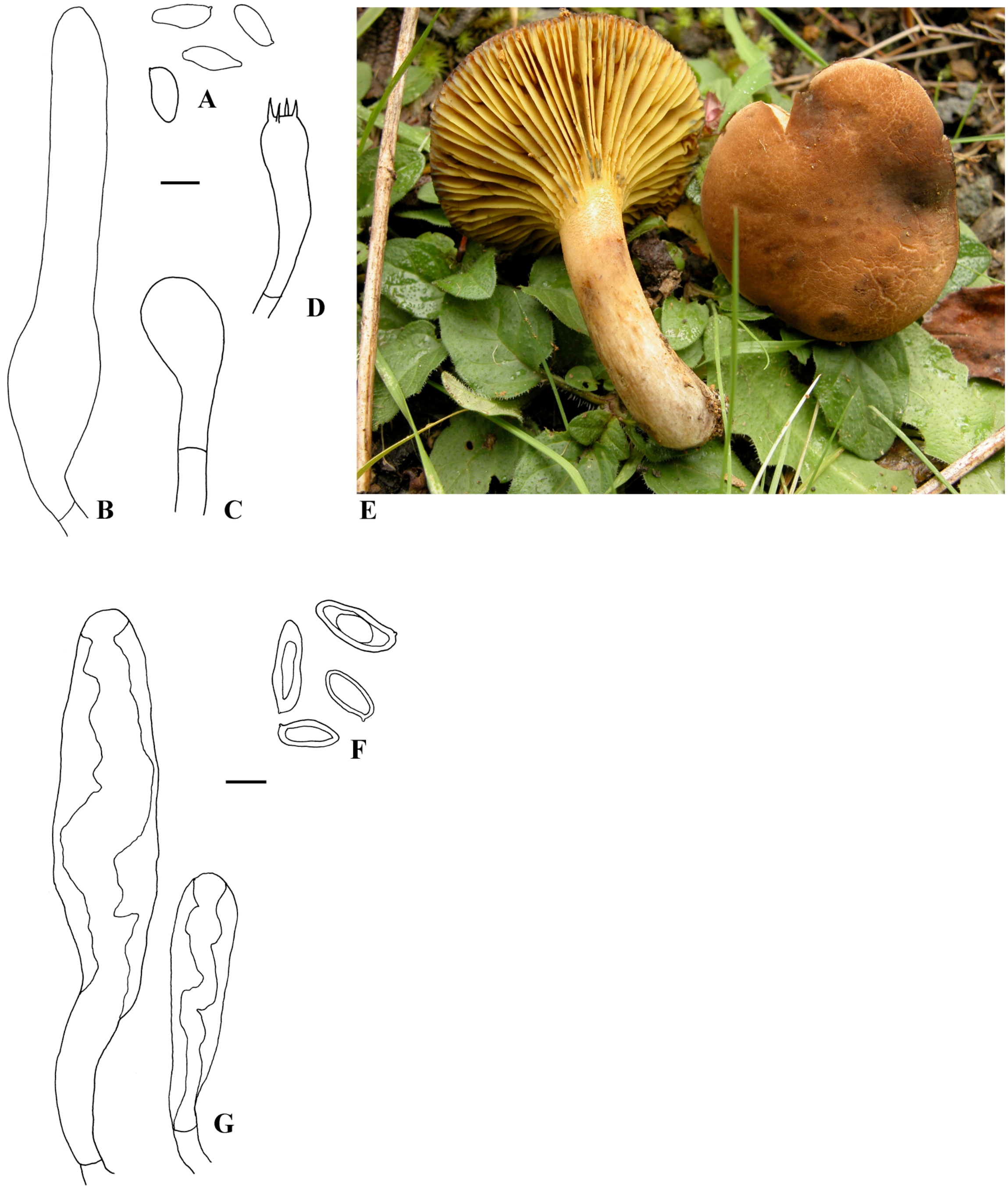

Figure 2-5: *Phylloporus bellus*. **A–E:** *P. bellu*s var. *bellus*. **A.** Spores. **B.** Pleurocystidium. **C.** Caulocystidium. **D.** Basidium. **E.** Basidiomata (REH8681), photograph by R.E. Halling. **F–G:** *P. cyanescens*. **F.** Spores. **G.** Pleurocystidia. (A–D; F–G: Scale bar = 10µm.)

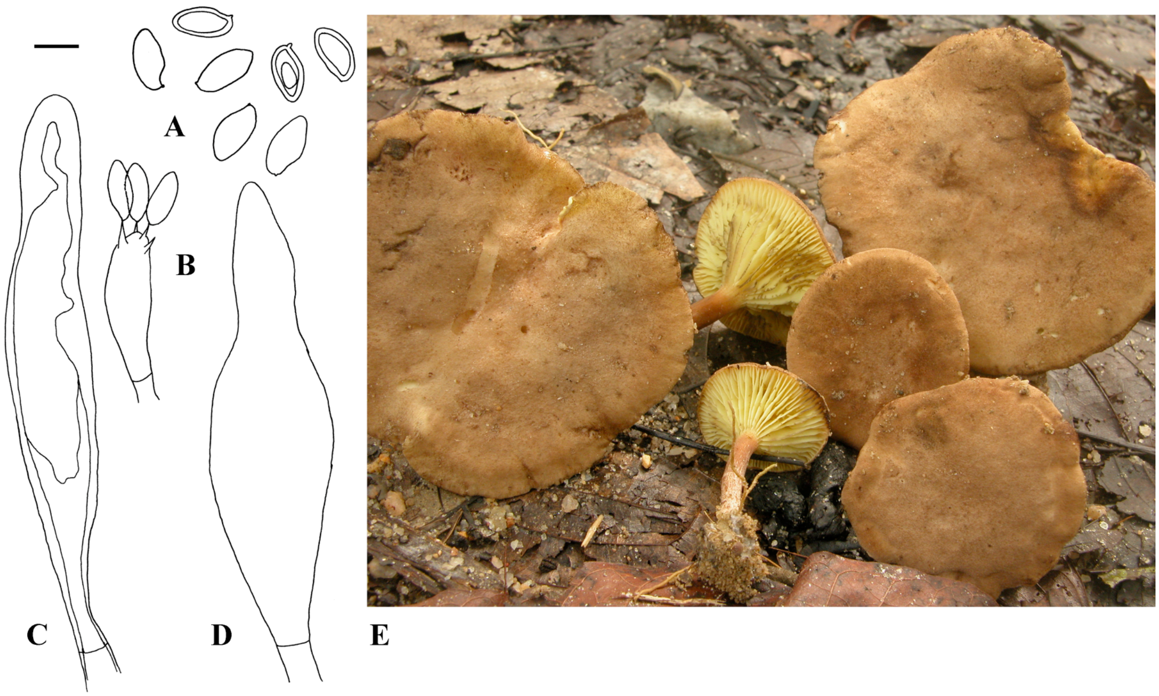

Figure 2-6: *Phylloporus bogoriensis*. **A.** Spores. **B.** Basidium. **C.** Pleurocystidium. **D.** Caulocystidium. **E.** Basidiomata (REH8691), photograph by R.E. Halling. (A–D: Scale bar = 10µm.)

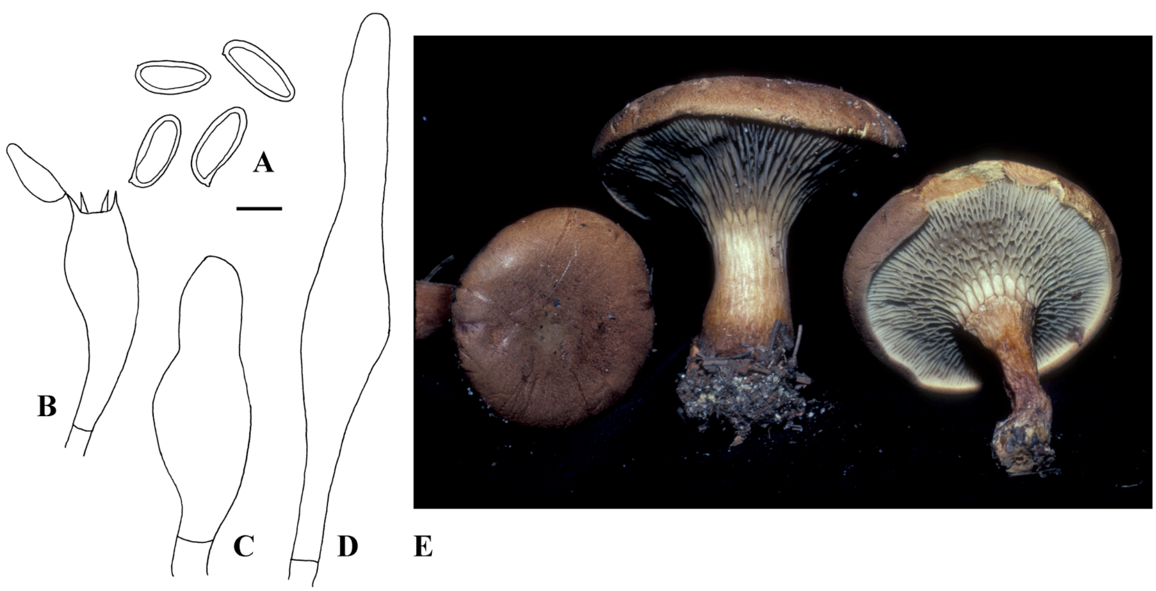

Figure 2-7: *Phylloporus boletinoides*. **A.** Spores. **B.** Basidium. **C.** Caulocystidium. **D.** Pleurocystidium. **E.** Basidiomata (REH3811), photograph by R.E. Halling. (A–D: Scale bar = 10µm.)

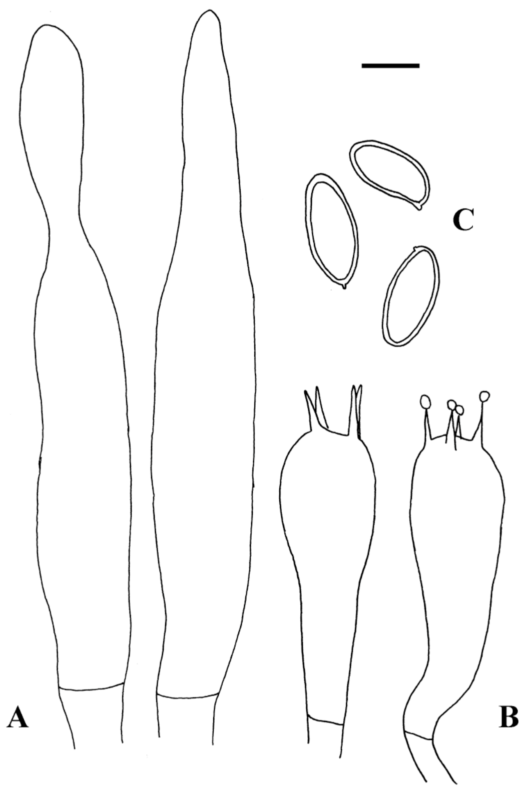

Figure 2-8: *Phylloporus caballeroi*. **A.** Pleurocystidia. **B.** Basidia. **C.** Spores. (A–C: Scale bar = 10μm.)

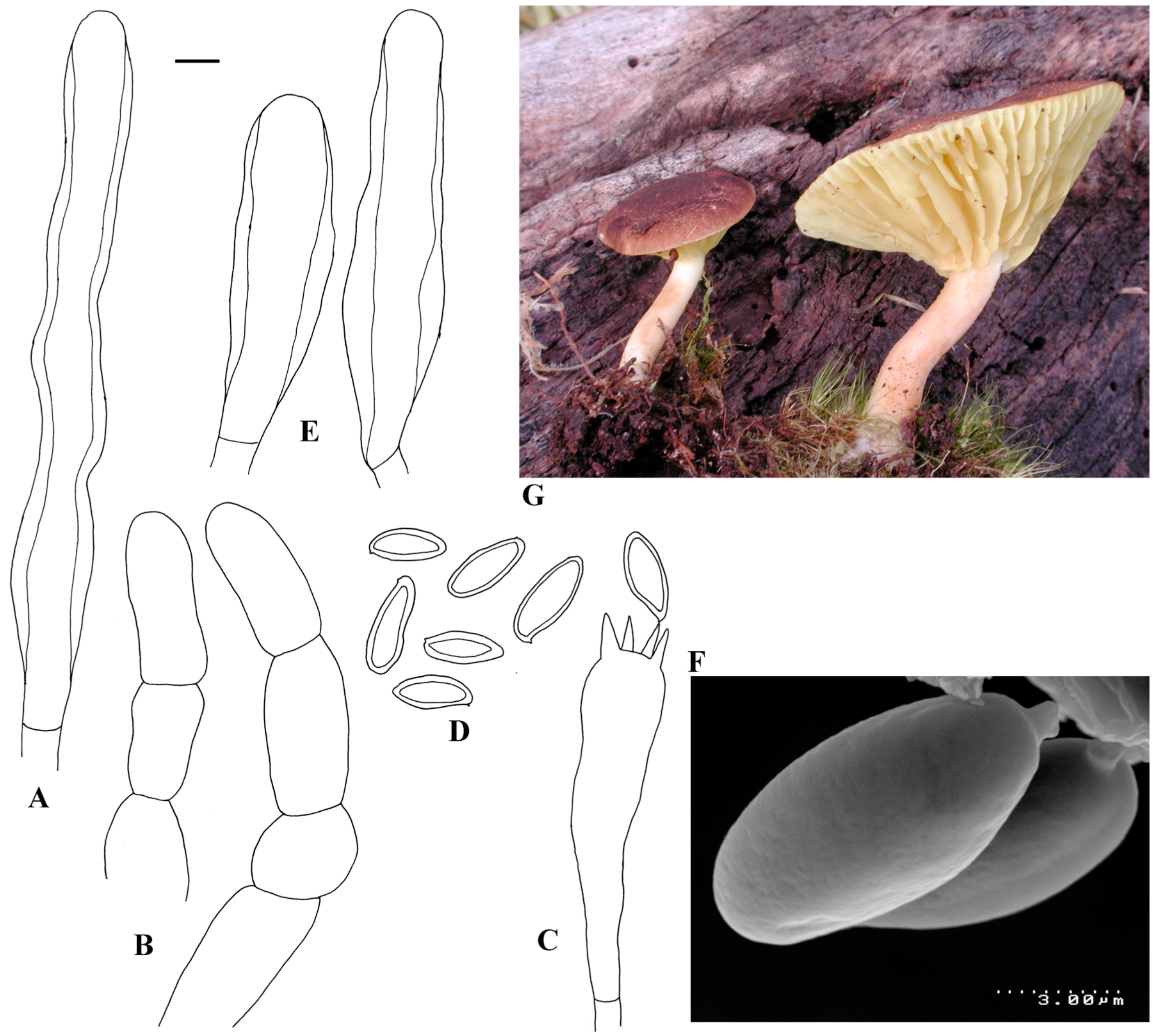

Figure 2-9: *Phylloporus centroamericanus*. **A.** Pleurocystidium. **B.** Pilleipellis hyphae. **C.** Basidium. **D.** Spores. **E.** Pleurocystidia. **F.** SEM of spores. **G.** Basidiomata (MAN57). (A–E: Scale bar = 10μm.)

Figure 2-10: *Phylloporus colligatus*. **A.** Spores. **B.** Pleurocystidia. **C.** Basidia. **D.** Basidiomata (TH8026), photograph by T. Henkel. (A–C: Scale bar = 10μm.)

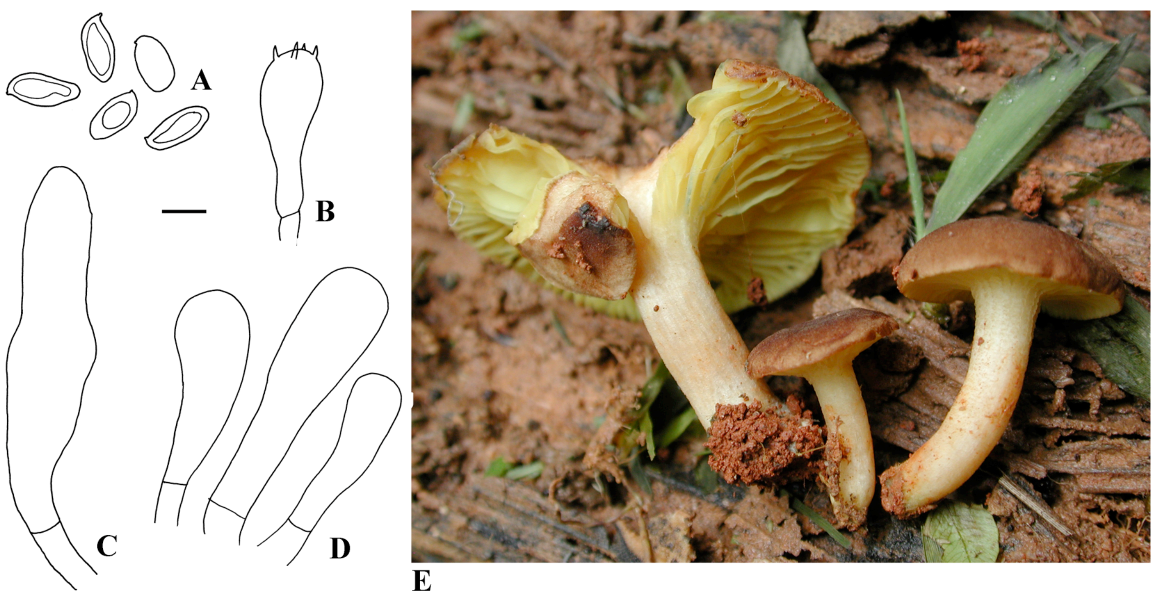

Figure 2-11: *Phylloporus curvatus*. **A.** Spores. **B.** Basidium. **C.** Pleurocystidium. **D.** Caulocystidia. **E.** Basidiomata (MAN104). (A–D: Scale bar = 10μm.)

Figure 2-12: *Phylloporus dimorphus*. **A.** Spores. **B.** Caulocystidium at the top of the stipe. **C.** Basidium. **D.** Pleurocystidium. **E.** Caulocystidium at the base of the stipe. **F.** Basidioma (MAN128). (A–E: Scale bar = 10µm.)

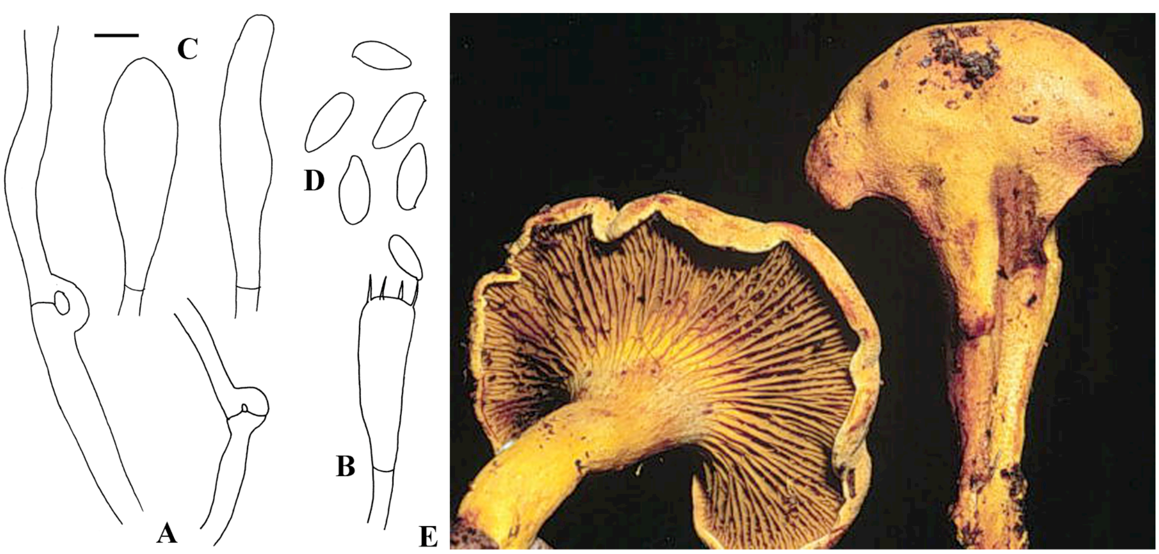

Figure 2-13: *Phylloporus fibulatus*. **A.** Clamp connections. **B.** Basidium. **C.** Pleurocystidia. **D.** Spores. **E.** Basidiomata (REH4983), photograph by R.E. Halling. (A–D: Scale bar = 10µm.)

Figure 2-14: *Phylloporus foliiporus*. **A.** Stipitipellis with caulocystidia. **B.** Spores. **C.** Pleurocystidium. **D.** Basidium. **E.** Pleurocystidia. **F.** Basidiomata (JLM1677), photograph by J.L. Mata. (A–E: Scale bar = 10µm.)

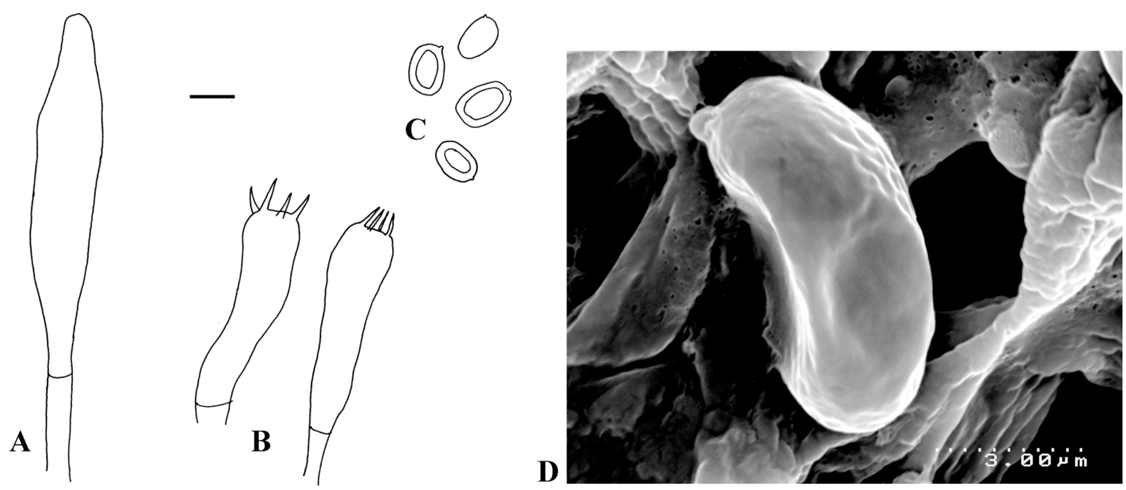

Figure 2-15: *Phylloporus guzmanii*. **A.** Pleurocystidium. **B.** Basidia. **C.** Spores. **D.** SEM of spores. (A–C: Scale bar = 10µm.)

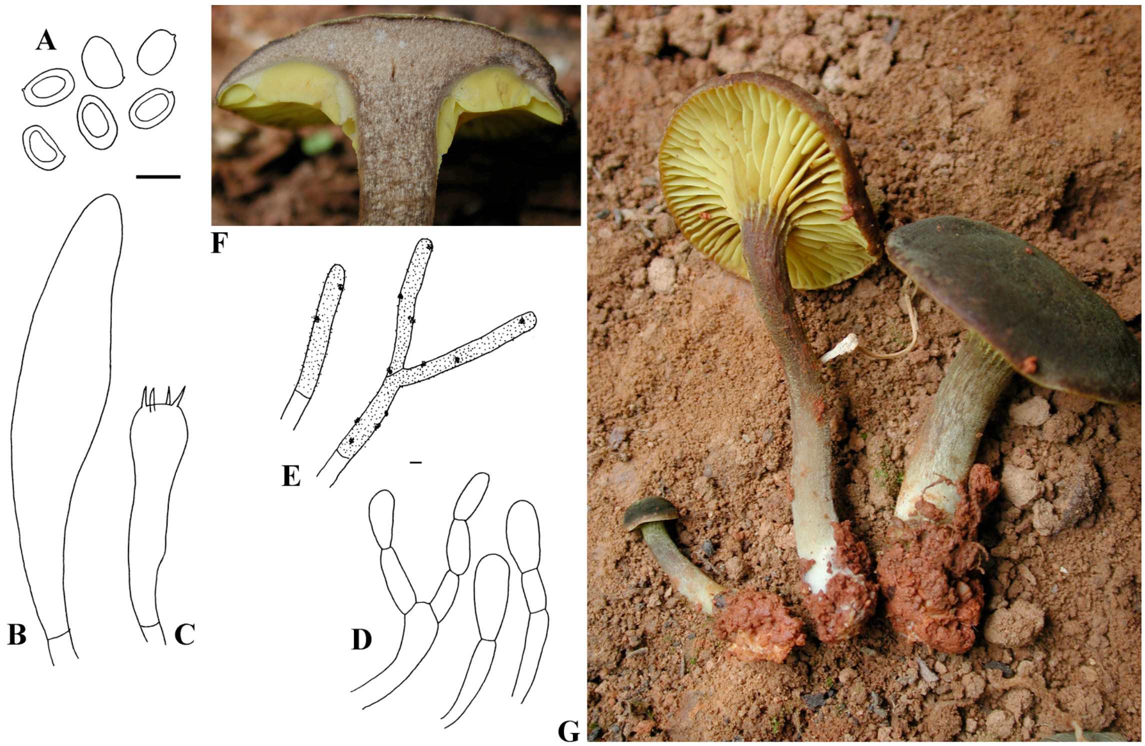

Figure 2-16: *Phylloporus infuscatus*. **A.** Spores. **B.** Pleurocystidium. **C.** Basidium. **D.** Pileipellis hyphae. **E.** Stipitipellis hyphae. **F, G.** Basidiomata (MAN123). (A–C: Scale bar = 10µm; D, E: Scale bar = 10µm.)

Figure 2-17: *Phylloporus leucomycelinus*. **A.** Pleurocystidia. **B.** Stipitipellis with caulocystidia. **C.** Basidium. **D.** Spores. **E.** Basidiomata (REH4582), photograph by R.E. Halling. (A, C–D: Scale bar = 10µm; B: Scale bar = 10µm.)

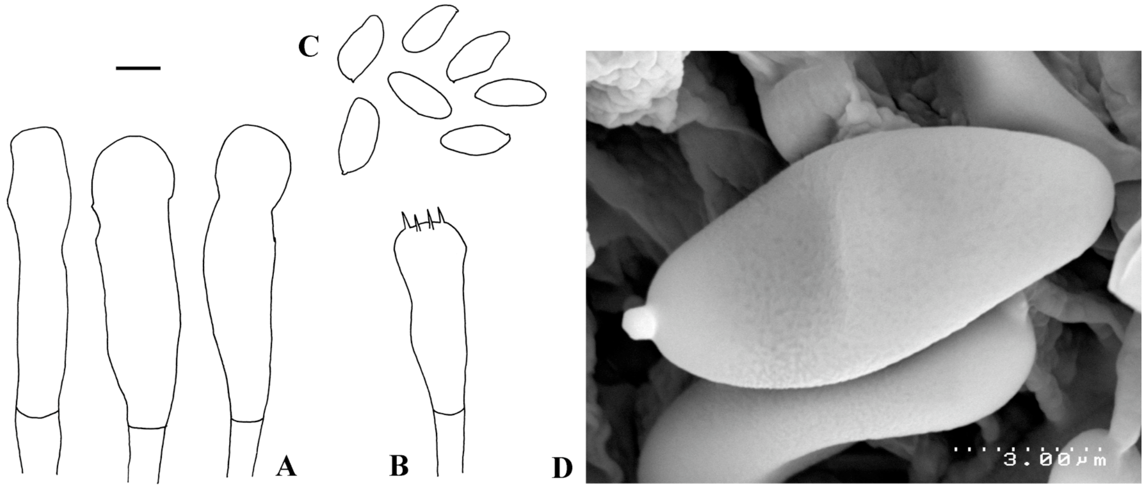

Figure 2-18: *Phylloporus novae-zelandiae*. **A.** Pleurocystidia. **B.** Basidium. **C.** Spores. **D.** SEM of spores. (A–C: Scale bar = 10µm.)

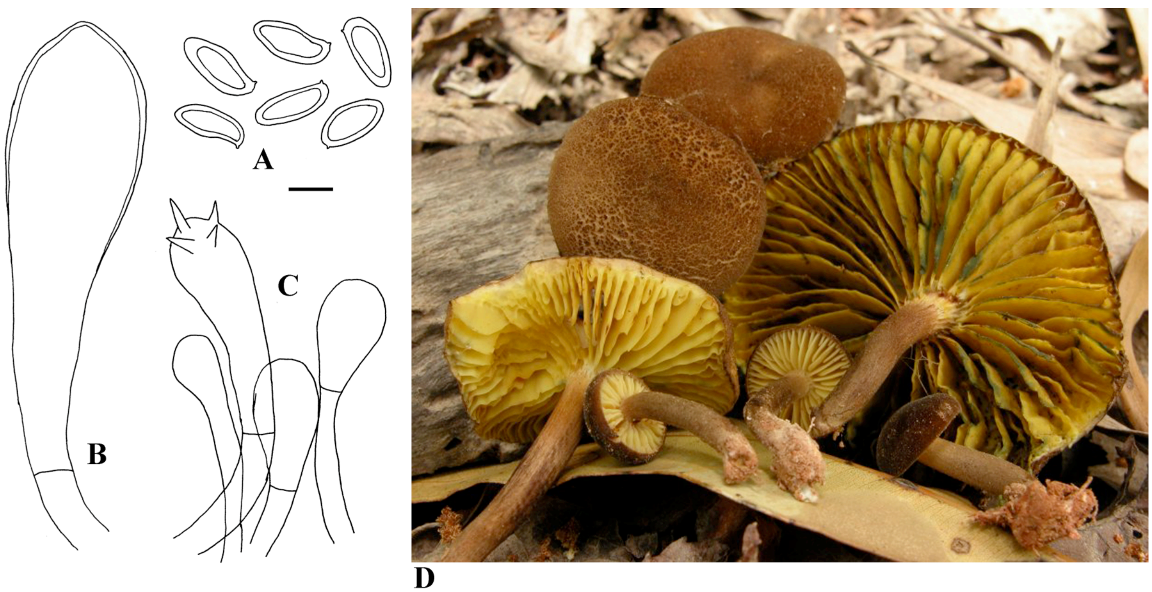

Figure 2-19: *Phylloporus orientalis* var. *brevisporus*. **A.** Spores. **B.** Pleurocystidium. **C.** Stipitipellis hyphae with caulocystidia and basidium. **D.** Basidiomata (REH8755), photograph by R.E. Halling. (A–C: Scale bar = 10µm.)

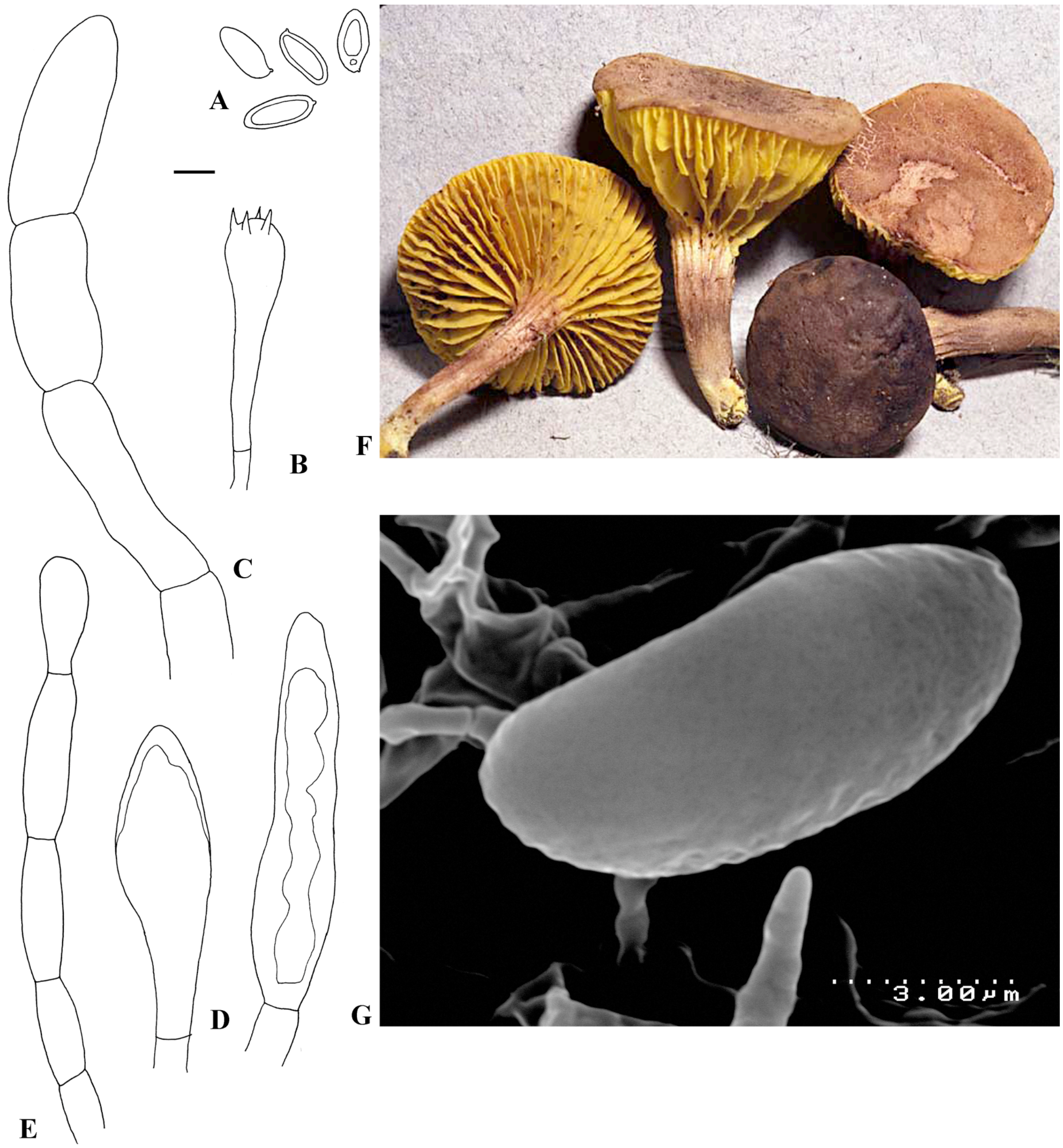

Figure 2-20: *Phylloporus phaeoxanthus* var. *phaeoxanthus*. **A.** Spores. **B.** Basidium. **C.** Pileipellis hyphae. **D.** Cystidia. **E.** Stipitipellis hyphae. **F.** Basidiomata (REH8032), photograph by R.E. Halling. **G.** SEM of spores. (A–E: Scale bar = 10µm.)

Figure 2-21: *Phylloporus phaeoxanthus* var. *simplex*. **A.** Spores. **B.** Pleurocystidium. **C.** Basidiomata (MAN38). (A, B: Scale bar = 10µm.)

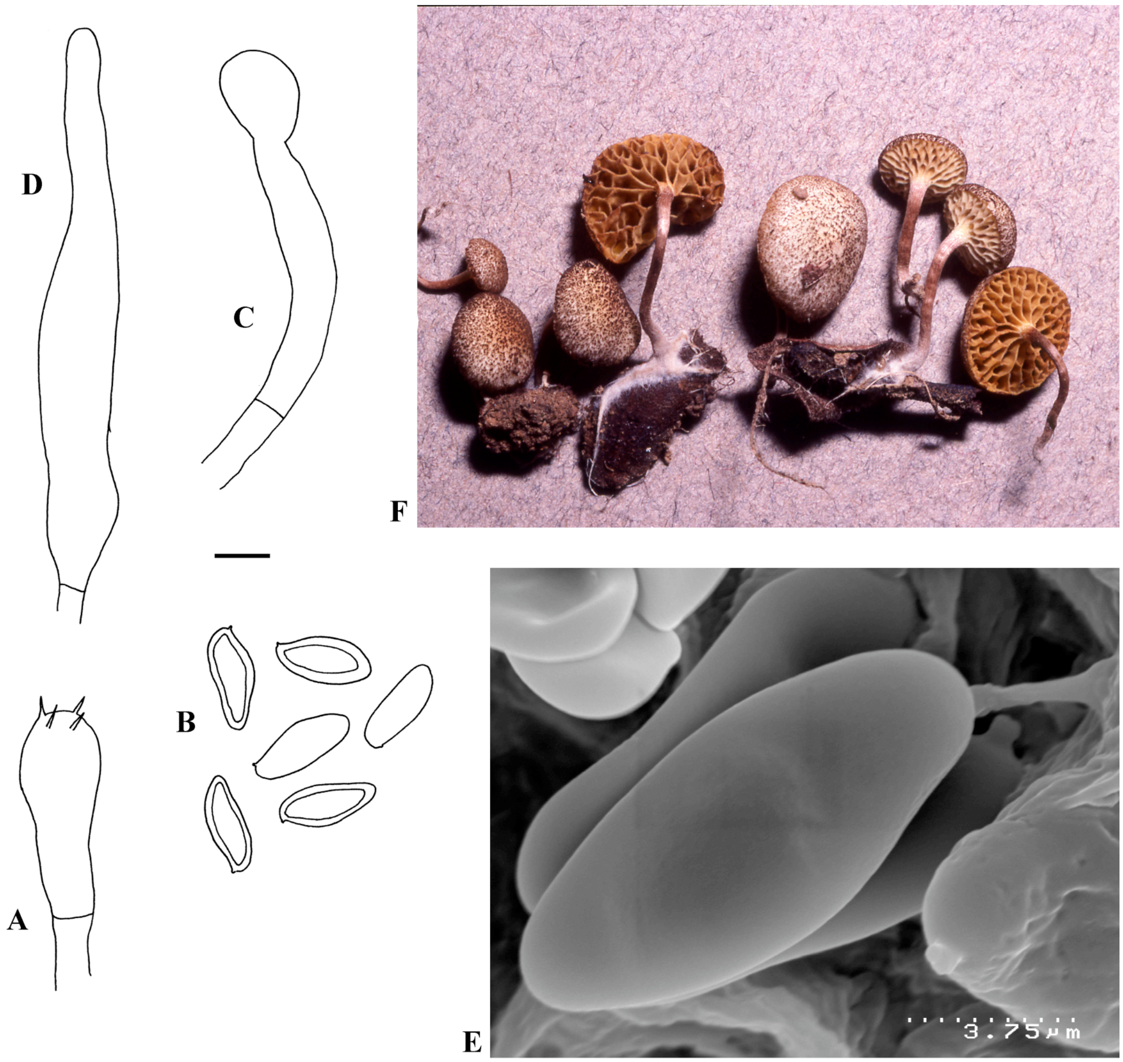

Figure 2-22: *Phylloporus pumilus*. **A.** Basidium. **B.** Spores. **C.** Caulocystidium. **D.** Pleurocystidium. **E.** SEM of spores. **F.** Basidiomata (REH8062), photograph by R.E. Halling. (A–D: Scale bar = 10µm.)

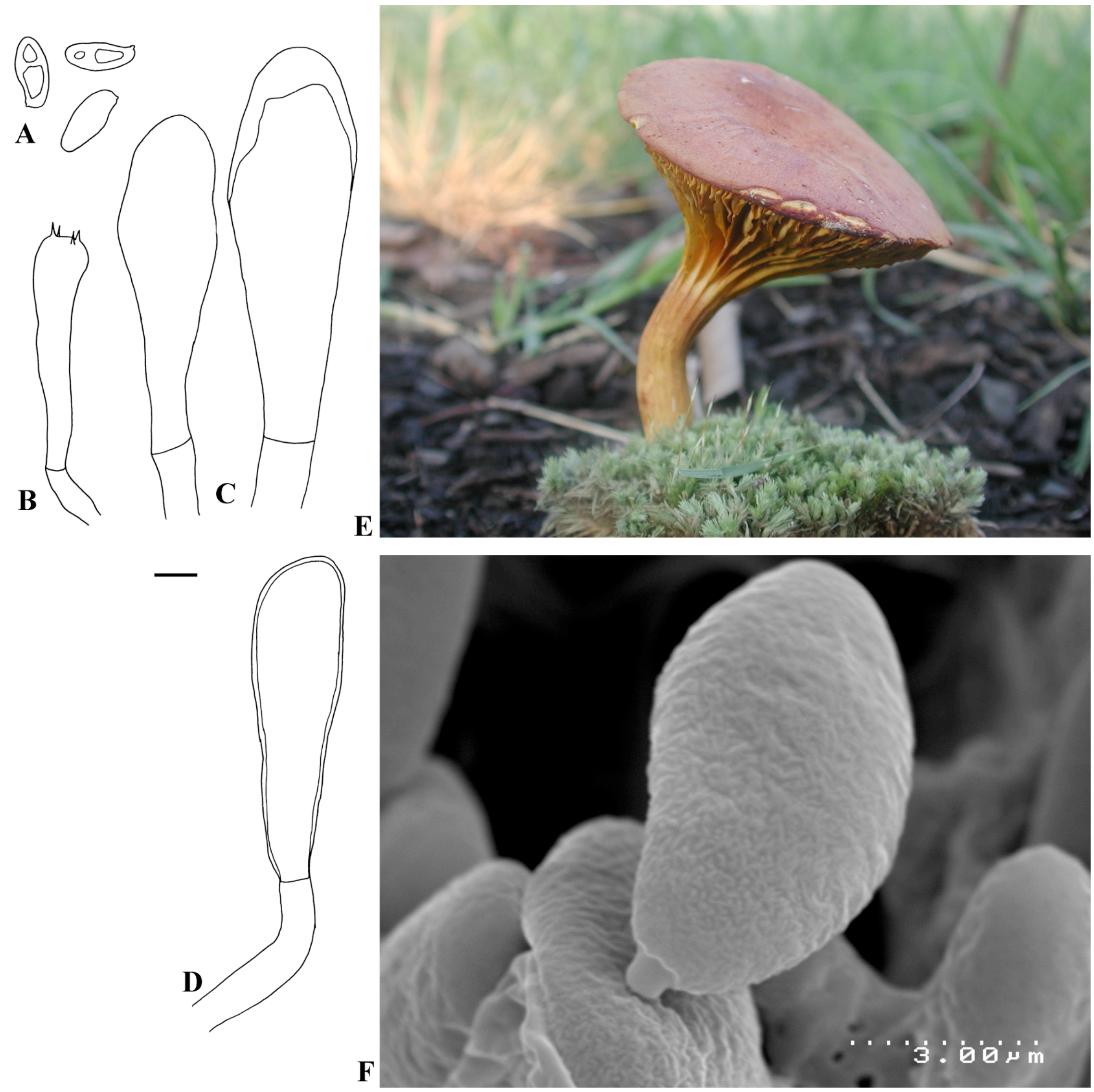

Figure 2-23: *Phylloporus rhodoxanthus*. **A.** Spores. **B.** Basidium. **C.** Pleurocystidia. **D.** Caulocystidia. **E.** Basidioma (MAN75). **F.** SEM of spores. (A–D: Sclae bar = 10µm.)

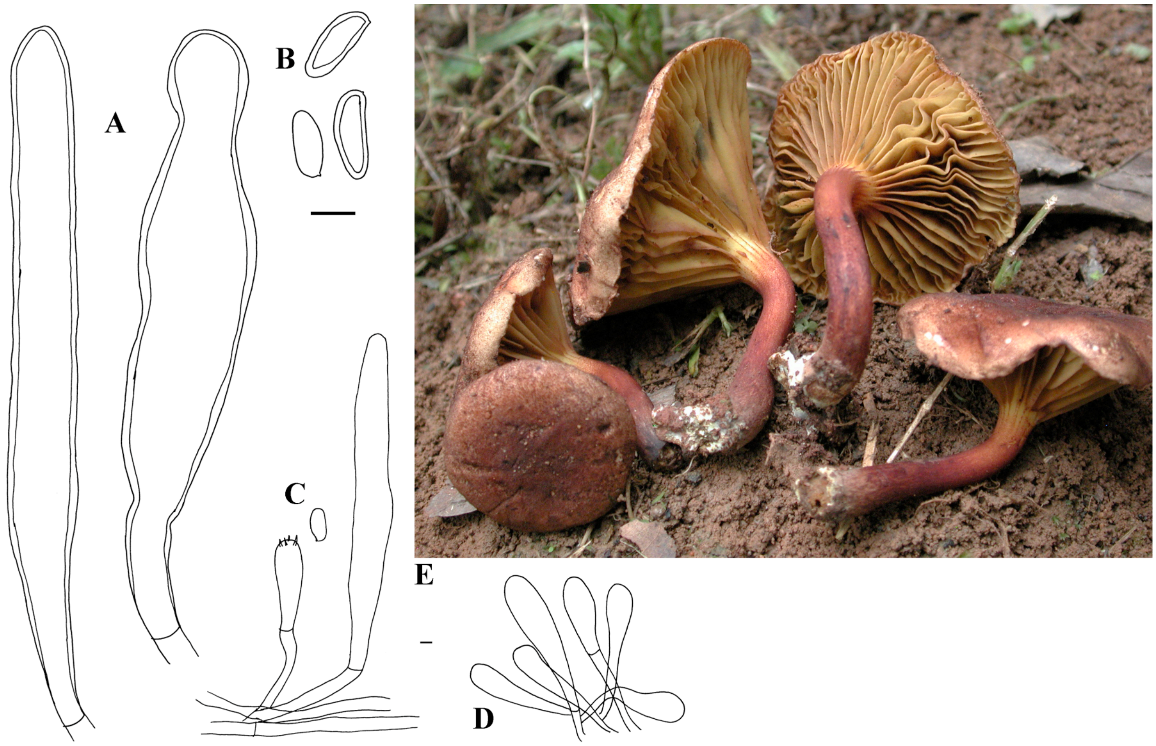

Figure 2-24: *Phylloporus rubiginosus*. **A.** Pleurocystidia. **B.** Spores. **C.** Basidium with Pleurocystidium and spore. **D.** Stipitipellis hyphae with caulocystidia. **E.** Basidiomata (MAN117). (A, B: Scale bar = 10µm; C, D: Scale bar = 10µm.)

Figure 2-25: *Phylloporus scabripes*. **A.** Spores. **B.** Basidium. **C.** Pleurocystidium. **D.** Caulocystidia. **E.** Pileipellis hyphae. **F.** Basidiomata (REH8558), photograph by R.E. Halling. (A–C: Scale bar = 10μm; D, E: Scale bar = 10μm.)

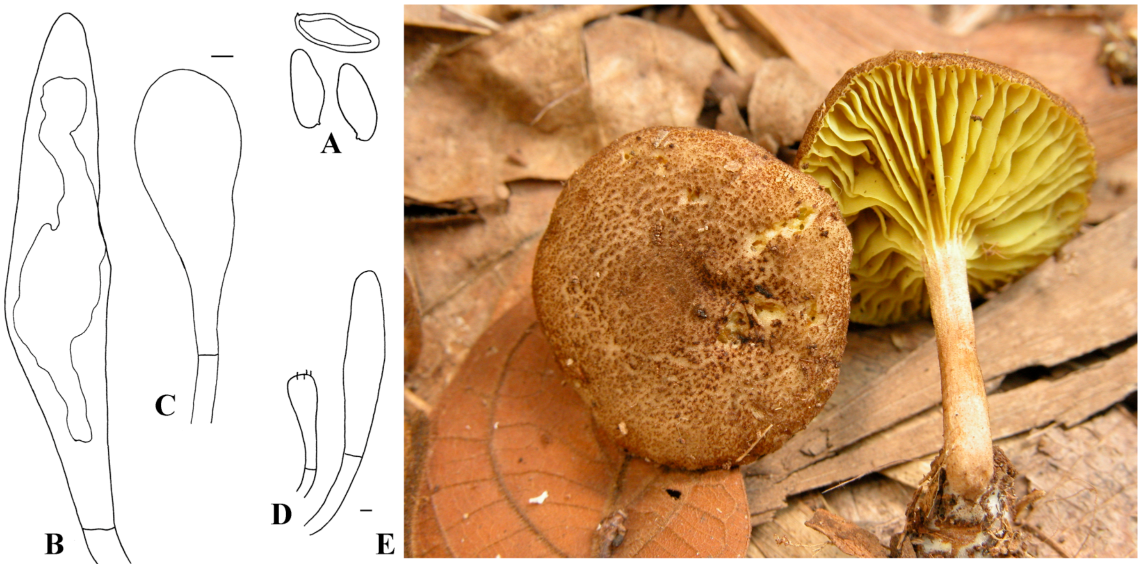

Figure 2-26: *Phylloporus* sp. 1. **A.** Spores. **B.** Pleurocystidium. **C.** Caulocystidium. **D.** Basidium and Pleurocystidium. **E.** Basidiomata (REH8729), photograph by R.E. Halling. (A–C: Scale bar = 10µm; D: Scale bar = 10µm.)

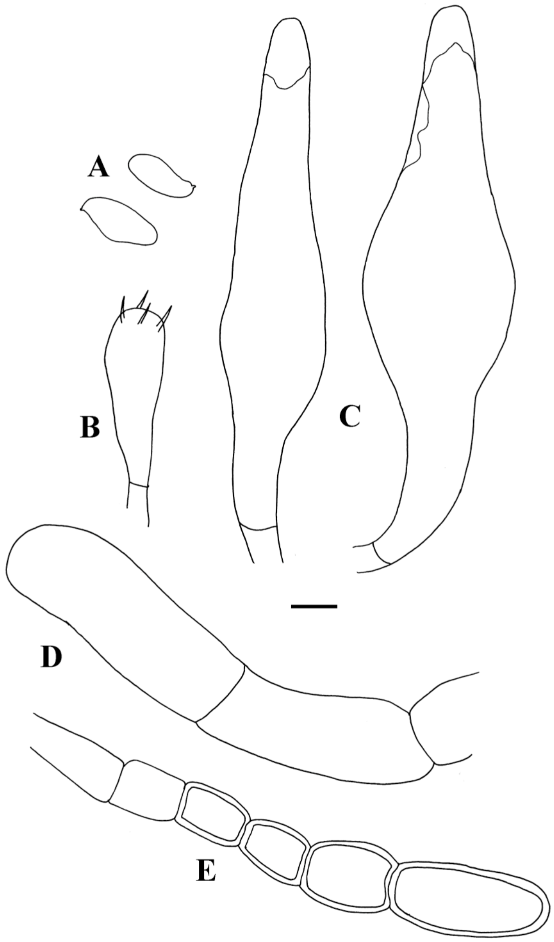

Figure 2-27: *Phylloporus* sp. 2. **A.** Spores. **B.** Basidium. **C.** Pleurocystidia. **D.** Pileipellis hyphae. **E.** Stipitipellis hyphae. (A–E: Scale bar = 10µm.)

List of Material Examined

Phylloporus pelletieri (Lév.) Quél., Flore Mycologique de France: 409. 1888.

MATERIAL EXAMINED: SLOVAKIA. Remetské Hámre village: Vihorlat mountain, Natural Reserve Machnatý 48°51'N, 22°10'60"E, 330 m, 19 September 2001, *Jančovičová & Terray Q 7199c* (SAV). ITALY. Liguria: 20 June 2005, *Rigoni* (NY). AUSTRIA. Tyrol: 1908, *von Höhnel* (FH).

Phylloporus alborufus Neves & Halling sp. nov.

MATERIAL EXAMINED: COSTA RICA. San José: San Gerardo de Dota, ±5 Km southwest of Cerro de la Muerte, Albergue de la Montaña, Savegre, 9º33'N, 83º48'28"W, 2200 m, 11 June 1994, *Neves 22* (HOLOTYPE: USJ, ISOTYPE: NY).

Phylloporus arenicola Smith & Trappe, Mycologia 64: 1138–1153. 1972.

MATERIAL EXAMINED: UNITED STATES. California: Marin Co., Bolinas Ridge Road, 37°52'7"N, 122°30'29"W, 3 January 1992, *Desjardin 5447* (SFSU). Mendocino Co., Jackson State Forest, Highway 409, 38° 20' 56N 120° 46' 23W, 21 November 1992, *Halling 6951* (NY). Sierra Co., Chapman Creek Campground, 37° 55' 8N, 122° 30' 57W, 5 June 1997, *Desjardin 6622* (SFSU).Washington: Clallam Co., Olympic National Forest, Boulder Creek Drainage, 48°15'17"N, 124°15'30"W, 19 October 1991, *Desjardin 5373* (SFSU).

Phylloporus aurantiacus Halling & Mueller, Mycotaxon 73: 64. 1999.

MATERIAL EXAMINED: COSTA RICA. Cartago: near town of Palo Verde, ±4.5 km E of km 31 of Interamericana Highway, 9º46'34"N, 83º56'42"W, 1600 m, 11 June 1994, *Halling 7271* (ISOTYPE: NY).

Phylloporus bellus (Mass.) Corner, Nova Hedwiga 20: 798. 1970.

MATERIAL EXAMINED: COSTA RICA. Guanacaste: Santa Rosa, Guanacaste Conservation Area, 10º51'4"N, 85º36'26"W, 330 m, 22 June 1997, *Halling 7733* (NY). UNITED STATES. Bronx: New York Botanical Garden, 40°67N, 73°94W, 13 July 2005, *Halling 8710* (NY). JAPAN. Tottori prefecture: Tottori, near Tottori Mycological Institute, 7 September 1983, *A4021* (F).

Phylloporus bogoriensis Höhnel, Sitzber. Kaiserl. Acad. Wiss. Wien Math.-naturw. Kl. 123. 98, n.838. 1914.

MATERIAL EXAMINED: INDONESIA. Java: Bogor, Bogor Botanical Garden, 6°35.955'S, 106°47.720'E, 266 m, 12 January 2005, *Desjardin 7785* (SFSU). MALAY. Selangor: Sungai Chongkak Forest Reserve, 3°12'42"S, 101°50'28"E, 18 m, 16 May 2005, *Halling 8691* (NY).

Phylloporus boletinoides A.H. Sm. & Thiers, A contribution toward a monograph of North American species of *Suillus*: 105. 1964.

MATERIAL EXAMINED: UNITED STATES. Florida: Alachua Co., east of Gainesville, 31 July 1958, *Thiers 4960* (HOLOTYPE: MICH); 16 August 1996, *Baroni 8047* (CORT); Sarasota, Myakka Valley Ranches, 12 January 1991, *F3912* (F). Texas: Tyler Co., Big Thicket National Preserve, Turkey Creek, 6 September 1996, *Baroni 8172* (CORT). New Jersey: Burlington Couinty: Penn State Forest, Oswego Lake, 17 August 1984, *Halling 3811* (NY). Alabama: Baldwin County, Orange Beach, 1982, *Lewis 3196* (F). BELIZE. Cayo District: Douglas D'Silva Forest Station, 6 October 2003, *Baroni 9681* (CORT).

Phylloporus caballeroi Singer, Beih. Sydowia, 7:101. 1973.

MATERIAL EXAMINED: ARGENTINA. Jujuy: Lagunas de Yala, 2400 m alt., 14 Feb 1966, *Singer T5051* (HOLOTYPE: F), *T4062* (PARATYPE: F). BOLIVIA. Departamento de La Paz: Province of Sud Yungas, 18-25 km from Unduavi on road to Chulumani, 16º19'S, 67º54"W, 22 March 1990, *Halling 6360* (NY). PANAMA. Chiriqui: Bugaba, Cerro Punta, Parque Internacional La Amistad, Sendero Retoño, 8°51'N, 82°34'W, 2280 m, 20 October 1999, *Halling 7906* (NY).

Phylloporus centroamericanus Singer & Gómez, Brenesia 22: 169. 1984.

MATERIAL EXAMINED: COSTA RICA. Cartago: La Chonta, 2800 m, September 1982, Gómez 18413 (TYPE: F). Puntarenas: Coto Brus, Zona Protectora Las Tablas, Finca La Cafrosa, Camino El Portones por El Tajo, 8°55'34"N, 82°46'W, 1500 m, 12 June 2004, *Neves 51* (NY). San Jose de Dota: La Chonta, South of Interamericana Highway, Cerro Chonta, 9°41'56"N, 83°56'31"W, 2340 m, 14 June 2004, *Neves 57, Neves 58, Neves 59* (NY). San Jose: San Gerardo de Dota, ±500 Km southwest of Cerro de La Muerte, Albergue de la Montaña, Savegre, 9°33'N, 83°48'28"W, 2200 m, 3 June 2004, *Neves 16* plus six collections from same locality (NY).

Phylloporus colligatus Neves, Henkel sp. nov.

MATERIAL EXAMINED: GUYANA. Pakaraima Mountains: Upper Potaro River Basin, 15 Km East of Mountain Ayanganna, 3 Km Soutwest of Potaro base, 5 May 2001, *Henkel 8026* (HOLOTYPE: BRG).

Phylloporus curvatus Neves & Halling sp. nov.

MATERIAL EXAMINED: THAILAND. Chiang Mai Province: Mae Sae, Highway 1095 at Km 55, 19°14'33.6"N, 98°38'29.4"E, 982 m, 3 June 2006, *Neves 104* (HOLOTYPE: MRC, ISOTYPE: NY); 10 June 2006, *Neves 124* (MRC, NY). Sangasabhasri lane to Huai Kok Ma village, Doi Suthep National Park, 18°48'24.3"N, 98°54'38"E, 1150 m, 7 June 2006, *Neves 118* (NY).

Phylloporus cyanescens (Mass.) Neves & Halling, comb. nov.

MATERIAL EXAMINED: AUSTRALIA. Victoria: Otway Range, Colac Otway Shire, Otway State Forest, 38º41'56"S, 143º28'42"E, 72 m, 08 May 2005, *Halling 8681*(NY).

Phylloporus dimorphus Neves & Halling sp. nov.

MATERIAL EXAMINED: THAILAND. Chiang Mai Province: Ban Pha Deng village, Pathummikaram temple, 19°06'28.8"N, 98°44'47.3"E, 1050 m, 12 June 2006, *Neves 128* (HOLOTYPE: NY). Doi Inthanon National Park, Highway 1009 at 25 Km marker, 18°32'19.5"N, 98°33'42.5"E, 1050 m, 5 June 2006, *Neves 111* (NY).

Phylloporus fibulatus Singer, Ovrebo, & Halling, Mycologia 82: 452. 1990.

MATERIAL EXAMINED: COLOMBIA. Departamento de Antioquia: along road from Santa Rosa de Osos to San José de la Montaña, 25 September 1986, *Ovrebo 2546* (ISOTYPE: NY); Municipio Santa Rosa de Osos, ±11 Km North of Santa Rosa de Osos, road to Aragon, near Llanos de Cuivá, 6°45'N, 75°3'W, 2500 m, 5 September 1986, *Halling 4983* (NY); on road to Aragon, El Chaquiro, 8 November 1988, *Halling 6068* (NY). Departamento de Nariño: Municipio Pasto, 11 Km East of Chachagüi, bosque "El Comun", 22 September 1988, *Halling 6132* (NY); Km 17, road from Pasto to Chachagüi, vereda "La Josefina", 23 September 1988, *Halling 6153* (PARATYPE: NY). Departamento del Tolima: Municipio de Murillo, Cabecera Municipal, Sector El Infierno, 2900 m, 19 April 2005, *Palacio 04* (HUA).

Phylloporus foliiporus Singer, Ovrebo, & Halling, Mycologia 82: 452. 1990.

MATERIAL EXAMINED: UNITED STATES. Florida: Gainsville, Sugarfoot Hammock, 11 July 1938, *F 17747* (HOLOTYPE: FH); *F 17774* plus five collections from same locality (FH). Alabama: Baldwin Co., Meaher State Park, 22 July 2005, *Mata 1677* (NY). JAPAN. Nara: Nara Park, 5 July 1975, *Trappe 4289* (SFSC).

Phylloporus guzmanii Montoya & Band.-Munoz, Mycotaxon 41(2): 473. 1991.

MATERIAL EXAMINED: MEXICO. Guerrero: Municipio de Chilpancingo, Omiltemi, 21 August 1985, *Pérez-Ramirez 565* (ISOTYPE: XAL).

Phylloporus infuscatus Neves & Halling sp. nov.

MATERIAL EXAMINED: THAILAND. Chiang Mai Province: Mae Sae, Highway 1095 at Km 55, 19°14'33.6"N, 98°38'29.4"E, 982 m, 10 June 2006, *Neves 123* (HOLOTYPE: MRC, ISOTYPE: NY).

Phylloporus leucomycelinus Singer, Persoonia 9(4): 426. 1978.

MATERIAL EXAMINED: UNITED STATES. Michigan: Warren Woods, 23 July 1973, Ponce de León & Nash; *Singer N4674* (HOLOTYPE: F). Bronx: New York Botanical Garden, 40°67'N, 73°94'W, 14 September 2003, *Halling 8452*, *Halling 8705* (NY). Massachusetts: Upton, 12 August 2003, *Binder 03-038* (NY); Worcester, Rutland State Park, 27 August 2000, *Binder 00-043* (NY); Lancaster, Devens Reserve Forces Training Area, Slate Quarry, 70 m, 2 September 1998, *D-112* (FH).

Phylloporus novae-zelandiae McNabb, New Zealand Journal of Botany, 9: 358. 1971.

MATERIAL EXAMINED: NEW ZEALAND. Nelson: Karamea, Umere, 8 January 1968, *McNabb 26516* (HOLOTYPE: PDD); plus five collections from the same locality.

Phylloporus orientalis var. **brevisporus** Corner

MATERIAL EXAMINED: AUSTRALIA. Queensland: Mareeba Shire, Mareeba, Davies Creek National Park, Davies Creek Road, after second waterfall, 17°1'53"S, 145°36'23"E, 700 m, 4 February 2006, *Halling 8731* (NY); Daintree National Park, 18 Km North of Cape Tribulation on road to Cooktown, 15°57'46"S, 145°23'23"E, 116 m, 12 February 2006, *Halling 8755* (NY); Atherton Shire, Atherton, Mount Baldy State Forest, Mount Baldy Road, approximately 5 Km west of Atherton-Herberton Highway, 17°20'30"S, 145°24'54"E, 970 m, 13 February 2006, *Halling 8756* (NY).

Phylloporus phaeoxanthus var. **phaeoxanthus** Singer & Gómez, Brenesia 22: 171. 1984.

MATERIAL EXAMINED: COSTA RICA. San José: Jardín de Dota, Route 2, 6 Km west of Empalme, 2000 m, 2 July 1983, *Singer 20583* (HOLOTYPE: F). COLOMBIA. Departamento de Boyacá: near border with Deparatamento de Cudinamarca, Municipio San Miguel de Sema, road from Simijacta to San Miguel de Sema, 9 May 1987, *Halling 5248* (NY). Departamento de Cauca: Municipio de Popayan, vereda Rio Blanco, Hacienda Cantaclaro, 1800 m, 9 May 1990, *Franco-Molano 423* (NY).

Phylloporus phaeoxanthus var. **simplex** Singer & Gómez, Brenesia 22: 171. 1984.

MATERIAL EXAMINED: COSTA RICA. San José: Empalme, 1.1 Km east of La Perla Restaurant, 2000 m, July 1983, *Singer 20623* (HOLOTYPE: F); San Gerardo de Dota, Albergue de La Montaña, Savegre, 5 Km southwest of Cerro de La Muerte, 9°33'2"N, 83°48'27"W, 2350 m, 19 October 1994, *Halling 7388* plus eight collections from the same locality (NY).

Phylloporus pumilus Neves and Halling sp. nov.

MATERIAL EXAMINED: INDONESIA. Java: Haurbentes Park, 6°32.65'S, 106°26.26'E, 300 m, 14 January 2001, *Halling 8062*; (HOLOTYPE: BO, ISOTYPE: NY), *Halling 8063* (NY).

Phylloporus rhodoxanthus (Schwein.) Bres., Fung. Trident. 2(14): 95. 1900.

MATERIAL EXAMINED: UNITED STATES. Virginia: Giles Co., Near Mountain Lake, 22 July – 25 August 1946, *Singer* (no collection number, type ssp. *americanus*) (NEOTYPE: FH). North Carolina: Asheville, 17 July 2004, *Neves 75* (NY); Pennsylvania: Mont Alto, 12 August 2005, *Neves 97* plus two collections (NY). Alabama: Mobile County, 22 May 2005, *Mata 1808* (NY). New York: Cornwall, Black Rock Forest, *Halling 8714* (NY); 19 July 1933, *Singer 667* (FH). Illinois: Cook County, Palos Park Cook County Forest Preserve, Sag Valley Division, Swallow Cliff Woods, 41°40'34"N, 87°51'52"W, 215 m, 19 August 1997, *Campbell 159*; 23 August 2003, *Leacock 5805* (F). Texas: Jasper County, Black Brunch Hunting Club, Road 2&6, 8 June 1990, *Lewis 4378* (F). North Carolina: Macon County, Horse Cove Drive, Bull Pen Road, 1000 m, 20 July 1997, *Quist 42* (F).

Phylloporus rubiginosus Neves & Halling sp. nov.

MATERIAL EXAMINED: THAILAND. Chiang Mai Province: Sangasabhasri lane to Huai Kok Ma village, Doi Suthep National Park, 18°48'24.3"N, 98°54'38"E, 1150 m, 7 June 2006, *Neves 117* (HOLOTYPE: MRC, ISOTYPE: NY), *Neves 119* (MRC, NY).

Phylloporus scabripes Ortiz & Neves, Fungal Diversity, In Press

MATERIAL EXAMINED: BELIZE. Belize District: Western Highway, Foster Property, near Belize Zoo, 17°22'25"N, 88°33'42"W, 50 m, 15 October 2003, *Halling 8558* (NY); Cayo District: Mountain Pine Ridge Reserve, 17°2'5.1"N, 88°56'53.3"W, 450 m, 7 October 2003, *Halling 8531* (NY).

Phylloporus sp.1

MATERIAL EXAMINED: AUSTRALIA. Queensland: Mareeba Shire, Mareeba, Barron Gorge National Park, Barron Gorge Road, Agathis Track, 16°50'8"S, 145°38'16"E, 380 m, 4 February 2006, *Halling 8729* (NY).

Phylloporus sp.2

MATERIAL EXAMINED: COSTA RICA. Puntarenas: Coto Brus, Zona Protectora Las Tablas, Finca La Cafrosa, Camino El Portones por El Tajo, 8°55'34"N, 82°46'W, 1500 m, 12 June 2004, *Neves 50* (NY).

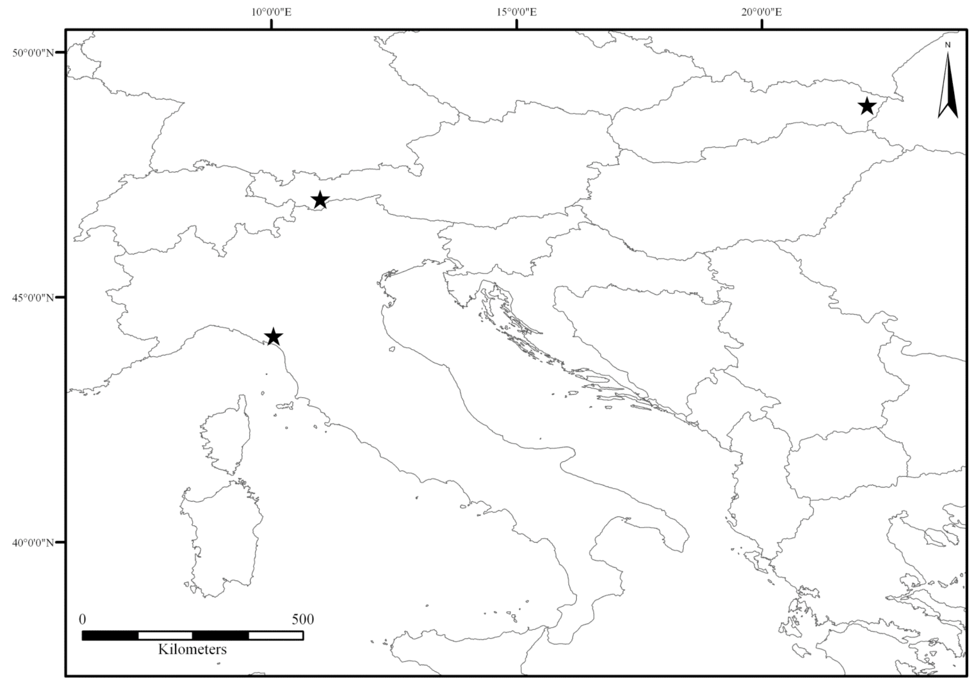

Figure 2-28. Distribution of the collections of *Phylloporus pelletieri* (★) in Europe.

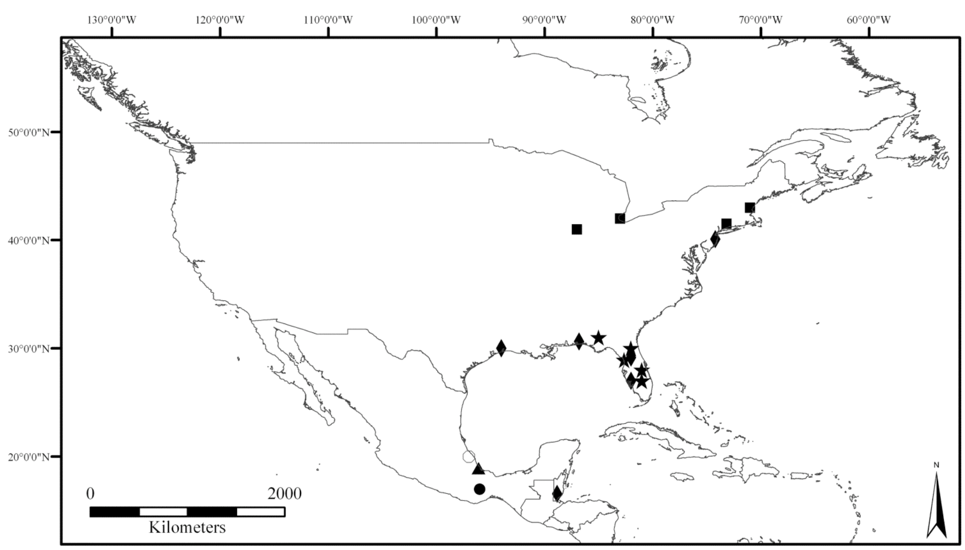

Figure 2-29. Distribution of the collections of *Phylloporus bellus* (●), *P. boletinoides* (♦), *P. centroamericanus* (▲), *P. foliiporus* (★), *P. leucomycelinus* (■), and *P. phaeoxanthus* (○) in the United States and Mexico.

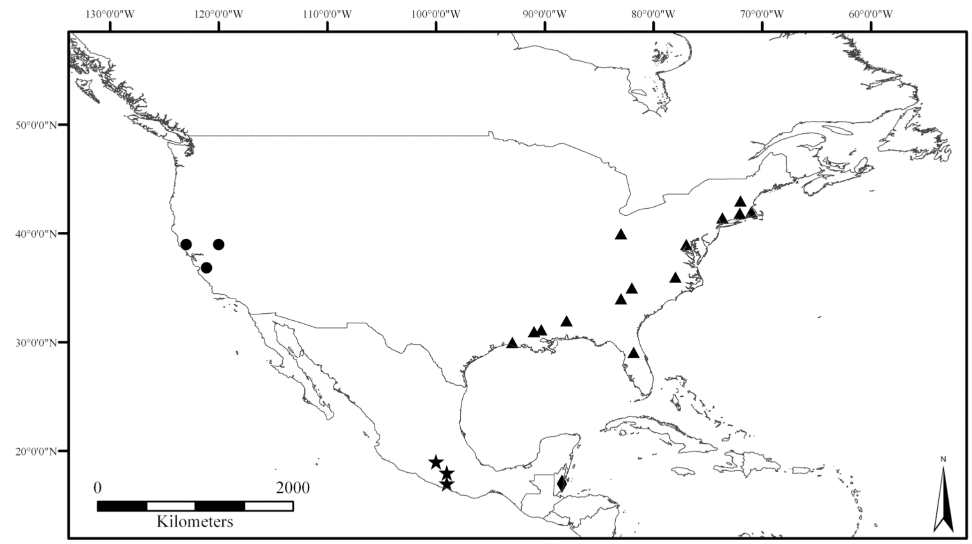

Figure 2-30. Distribution of the collections of *Phylloporus arenicola* (●), *P. rhodoxanthus* (▲), *P. guzmanii* (★), and *P. scabripes* (♦) in the United States and Mexico.

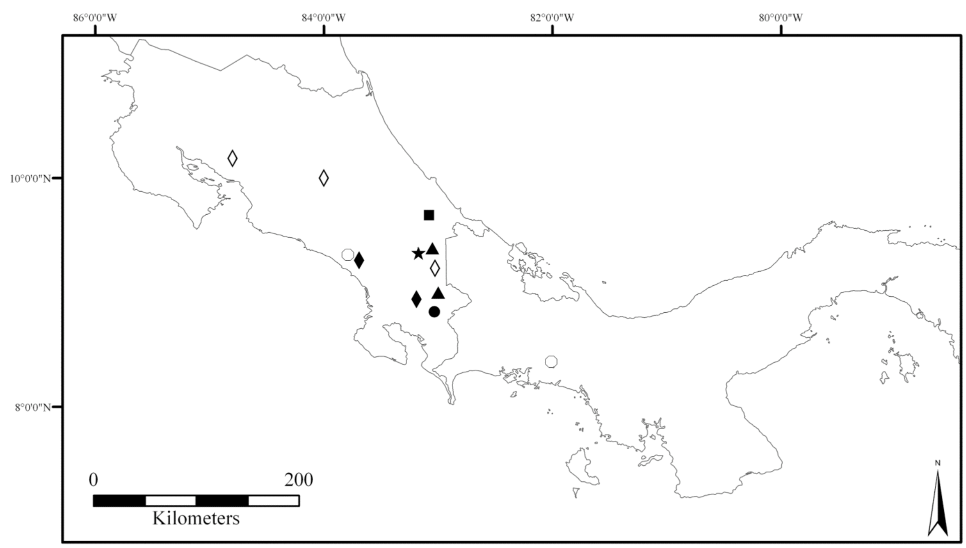

Figure 2-31. Distribution of the collections of *Phylloporus caballeroi* (○), *P. alborufus* (●), *P. aurantiacus* (■), *P. centroamericanus* (▲), *P. bellus* (◊), *P. phaeoxanthus* (♦), and *Phylloporus* sp. 2 (★) in Costa Rica and Panama.

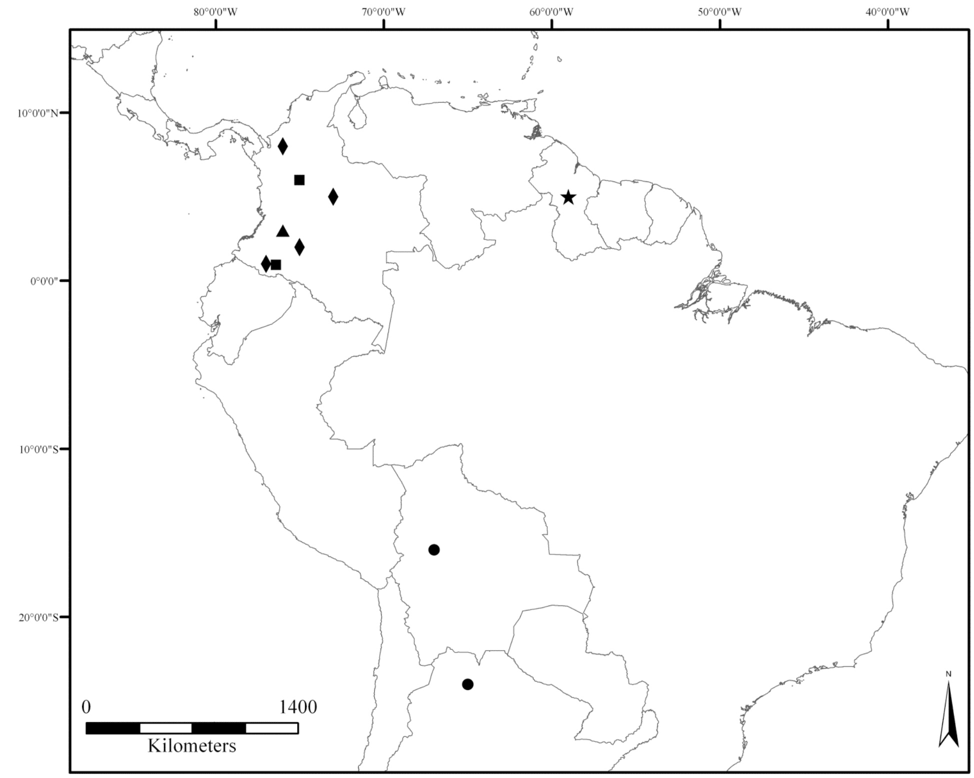

Figure 2-32. Distribution of the collections of *Phylloporus caballeroi* (●), *P. centroamericanus* (▲), *P. colligatus* (★), *P. fibulatus* (■), and *P. phaeoxanthus* (♦) in South America.

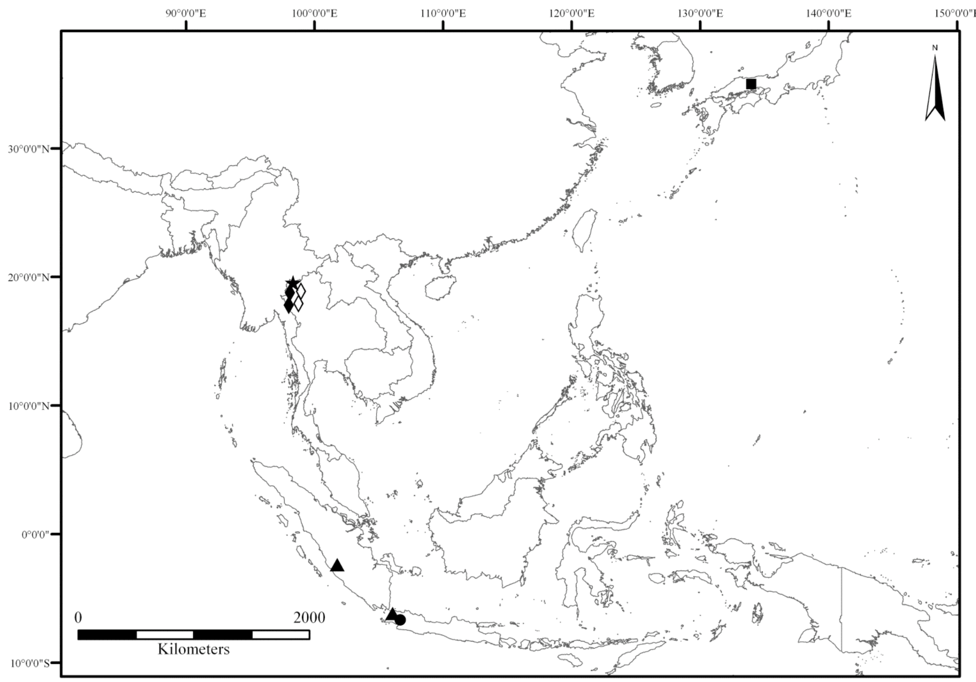

Figure 2-33. Distribution of the collections of *Phylloporus bogoriensis* (▲), *P. bellus* (■), *P. curvatus* (◊), *P. infuscatus* (★), *P. pumilus* (•), and *P. rubiginosus* (♦) in Southeast Asia.

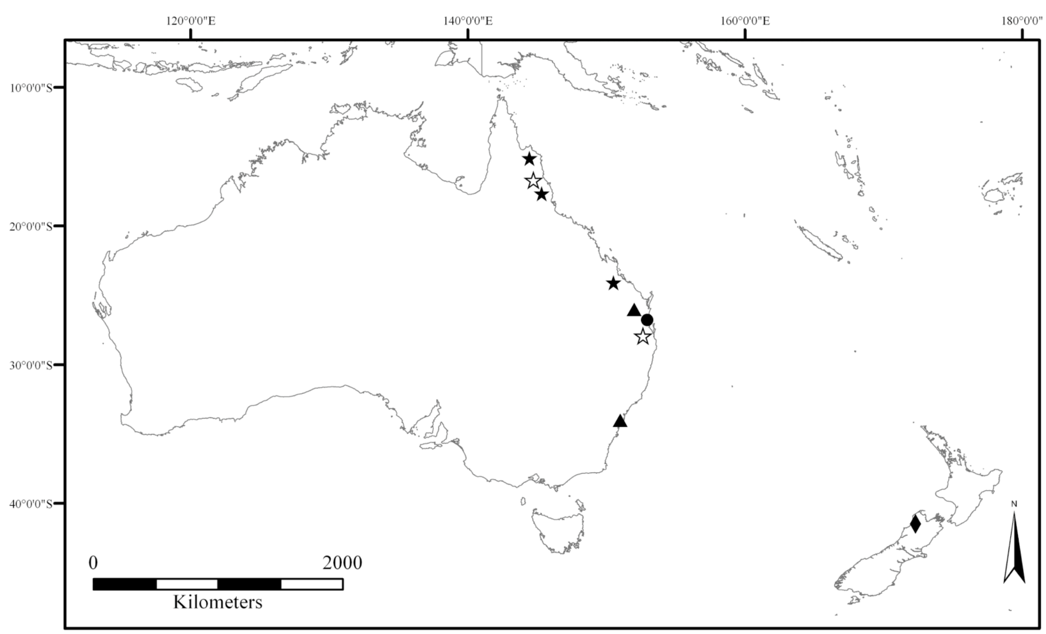

Figure 2-34. Distribution of the collections of *P. cyanescens* (•), *P. bogoriensis* (▲), *P. orientalis* (★), *P. novae-zelandiae* (♦), and *Phylloporus* sp. 1 (☆) in Oceania.

References

Corner, E.J.H. 1970. *Phylloporus* Quél. and *Paxillus* Fr. in Malaya and Borneo. Nova Hedwigia 20: 793-822.

Dallwitz, M.J. 1980. A general system for coding taxonomic descriptions. Taxon 29: 41-46.

Dallwitz, M.J., Paine, T.A. and Zurcher, E.J. 1993 onwards. User's guide to the DELTA system: a general system for processing taxonomic descriptions.

Halling, R.E. 1989. A synopsis of Colombian boletes. Mycotaxon 34: 93-113.

Halling, R.E. 2001. Ectomycorrhizae: Co-evolution, significance, and biogeography. Annals of the Missouri Botanical Garden 88: 5-13.

Halling, R.E., Mueller, G.M. and Dallwitz, M.J. 1999. New *Phylloporus* (Basidiomycetes, Boletaceae) with a key to species in Colombia and Costa Rica. Mycotaxon 73: 63-68.

Heinemann, P. and Rammeloo, J. 1986. *Phylloporus* (Boletineae). Flore Illustreé des Champignons d'Afrique Centrale 13: 277-309.

Holmgren, P.K., Holmgren, N.H. and Barnett, L.C. 1990. Index Herbariorum. 8. New York Botanical Garden Press, 693 pp.

Konrad, P. and Maublanc, A. 1928. *Phylloporus rhodoxanthus* (Scweinitz) Bresadola. Icones Selectœ Fungorum Tome VI: pl. 392.

Kornerup, A. and Wanscher, J.H. 1978. Methuen Handbook of colour. 3rd ed. Methuen Co., London. 243 pp.

Ladurner, H. and Simonini, G. 2003. *Xerocomus* s.l. 8. Edizioni Candusso, Alassio. 527 pp.

Largent, D.L. 1986. How to Identify Mushrooms to Genus I: Macroscopic features. I. 2nd. Mad River Press Inc., Eureka. 166 pp.

Largent, D.L., Johnson, D. and Watling, R. 1977. How to Identify Mushrooms to Genus III: Microscopis features. III. Mad River Press Inc., Eureka. 148 pp.

Montoya, L. and Bandala, V.M. 1991. Studies on the genus *Phylloporus* in Mexico, I. Discussion of the known species and description of a new species and a new record. Mycotaxon 41: 471-482.

Ortiz-Santana, B., Lodge, D.J., Baroni, T.J. and Both, E.E. 2007. Boletes from Belize and the Dominican Republic. Fungal Diversity In Press.

Pérez-Ramírez, L., Villegas, M. and Cifuentes, J. 1986. Descripción de macromicetos poco estudiados en Mexico, II. Revista Mexicana de Micologia 2: 251-257.

Quélet, L. 1888. Flore mycologique de la France et des pays limitrophes. Paris. 492 pp.

Singer, R. 1938. Sur les genres *Ixocomus*, *Boletinus*, *Phylloporus*, *Gyrodon* et *Gomphidius*. Revue de Mycologie 3: 157-177.

Singer, R. 1945. The Boletineae of Florida with notes on extralimital species. II. the Boletaceae (Gyroporoideae). Farlowia 2: 223-303.

Singer, R. 1965. Die Röhrlinge. Teil I. Verlag Julius Klinkhardt, Bad Heilbrunn. 131 pp.

Singer, R. 1978. Notes on Bolete taxonomy II. Persoonia 9: 421-438.

Singer, R. and Gómez, L.D. 1984. The Basidiomycetes of Costa Rica. III. The genus *Phylloporus* (Boletaceae). Brenesia 22: 163-181.

Singer, R., Ovrebo, C.L. and Halling, R.E. 1990. New species of *Phylloporus* and *Tricholomopsis* from Colombia, with notes on *Phylloporus boletinoides*. Mycologia 82: 452-459.

Smith, A.H. and Trappe, J.M. 1972. The higher fungi of Oregon's Cascade Head Experimental Forest and vicinity - I. The genus *Phaeocollybia* (Agaricales) and notes and descriptions of other species in the Agaricales. Mycologia 64: 1138-1153.

Watling, R. and Gregory, N.M. 1991. Observations of the boletes of the Cooloola Sandmass, Queensland and notes on their distribution in Australia. Part 3. Lamellate taxa. Edinburgh Journal of Botany 48: 353-391.

CHAPTER 3: PHYLOGENY OF SELECTED *PHYLLOPORUS* SPECIES BASED ON NUC-LSU DNA AND ITS SEQUENCES

Abstract

The phylogeny of *Phylloporus* (Boletaceae) has not previously been well studied, and the taxonomic relationships of this genus have varied considerably among authors. The following study presents phylogenetic relationships of *Phylloporus* based on two nuclear ribosomal DNA regions, ITS and LSU. The ITS dataset includes 39 collections of *Phylloporus* and five collections of *Xerocomus*, which were included to resolve sister group relationships. The LSU dataset is larger and contains 50 *Phylloporus* and 14 terminals of other genera in the family, including nine *Xerocomus*. A combined analysis of both genes was also performed. The deeper nodes in the phylogeny were not well resolved in any of the analyses, but the results suggest that *Phylloporus* is monophyletic and a sister group of the *Xerocomus subtomentosus* group.

The lamellate hymenophore configuration is a synapomorphy that distinguishes *Phylloporus* from the other genera in the family. The placement of a lamellate genus within Boletaceae suggests that hymenophore evolution is not well understood in the family. This is the first phylogeny of *Phylloporus* and includes 20 species from different geographic regions. The results, although not totally conclusive, indicate that *Phylloporus* is a monophyletic genus that is separate from the genus *Xerocomus* within the Boletaceae.

Introduction

Phylloporus is one of the largest genera in the Boletaceae and species in this genus are represented in tropical forests worldwide (Corner 1970, Heinemann & Rammeloo 1986, Montoya & Bandala 1991, Singer & Gómez 1984, Singer et al. 1990). A few species are also found in North America (Singer 1945, Smith & Thiers 1971) and the type species, *Phylloporus pelletieri*, is from Europe (Ladurner & Simonini 2003). The genus contains 70 species; however many parts of the neotropics and the paleotropics have not been extensively studied, and tropical collections to-date suggest that some of these regions are hot spots for *Phylloporus* diversity, for example Malaysia (Corner 1970), Africa (Heinemann & Rammeloo 1986), Costa Rica (Singer & Gómez 1984), and Colombia (Singer et al. 1990).

Phylloporus species are known to form mycorrhiza with species of Fabaceae (Caesalpinioideae, Mimosoideae), Casuarinaceae, Dipterocarpaceae, Fagaceae, Myrtaceae, and Pinaceae (Halling & Mueller 2002, Heinemann & Rammeloo 1986). Previous studies have shown that some boletes are suspected to have migrated from North America to Central and South America following their host tree (Halling 2001, Mueller & Halling 1995), for example, *Leccinum* and *Quercus* (Halling 1999).

Corner (1972) considered the tubular *Boletus* with elongate fusoid spores to be derived from a lamellate agaric of *Paxillus*-*Phylloporus* affinity and that the characteristic shape of the spore was developed in this ancestor. He postulated that the development of a tubular hymenophore was related to the elongated form of the spores due to the compression of what he called spore-circles inside the tubes of the hymenophore. More ‘primitive’ species would then have spores that are closer to isodiametric form, and more ‘advanced’ species with narrow tubes forming the hymenophore would produce spores that are extended along the longitudinal axis.

Pegler and Young (1981) believed that spore elongation should be seen as a phylogenetic trend, affirmed that the form could be variable in genera regarded as advanced, and cite *Xerocomus* as a genus which has both subglobose and fusoid spored species. Likewise, genera such as *Heimiella*, *Phylloboletellus*, and *Strobilomyces,* considered to be primitive for other reasons, would present uniformity in the subisodiametric outline and lack lateral compression. The same authors (Pegler & Young 1981) postulated that the variation in the spore ornamentation could be interpreted as a phylogenetic series, where the coarsely ornamented spores are more primitive than the smooth ones. They also suggested that the hymenophore configuration should be regarded as secondarily lamellate and derived from a tubular condition.

Phylloporus was considered by Corner (1972) to be primitive due to the lamellate hymenophore. Pegler & Young (1981) defined *Phylloporus* as a derived genus for the same reason, combining that characteristic with another one, the smooth spores, also considered by those authors as derived. It has been reported that *Phylloporus*, as in *Xerocomus*, has both subglobose and fusoid spored species, supporting Pegler & Young’s theory (1981). Diverse molecular phylogenies of the Boletales have shown *Phylloporus* as a derived species that it is placed next to the *Xerocomus subtomentosus* group, towards the top of the lineage (Binder 1999, Binder & Hibbett 2006).

The phylogenetic relationships in the genus *Phylloporus* remain unclear despite several broad-scale studies of the Boletales (Binder & Bresinsky 2002, Binder & Hibbett 2006, Grubisha et al. 2001). A phylogenetic study by Binder (1999) based on molecular data that included *Phylloporus rhodoxanthus* and *P. pelletieri*, placed *Phylloporus* within *Xerocomus* Quél. sensu stricto; however, Binder (1999) maintained *Phylloporus* and *Xerocomus* as independent genera because of morphological differences in the hymenophore configuration. However, he also noted the sister group relationship of *Phylloporus* with the *Xerocomus subtomentosus* complex (*X. illudens*, *X. lanatus*, and *X. subtomentosus*) and suggested that, based on his analyses, these taxa could be treated as a single genus.

Based solely on DNA sequences of two *Phylloporus* species, Bresinsky & Besl (2003) reduced the genus to synonymy with *Xerocomus* and suggested that the non-European species, with the exception of the North American *P. rhodoxanthus*, should be placed in a new genus. No new name was suggested and no tropical taxa were included in the analyses. Nevertheless, this synonymy exacerbates the systematic problems of *Phylloporus* especially since *Xerocomus* is not considered a monophyletic genus (Binder 1999, Binder & Hibbett 2006). The two genera are distinguished by the lamellate hymenophore produced by *Phylloporus* in contrast to the wide tubular hymenophore seen in *Xerocomus*. Even though the hymenophore of some species in *Phylloporus* show a high degree of anastomosis, it is never tubular as in Xerocomus, and this lamellate hymenophore is a characteristic that distinguishes *Phylloporus* from other genera in the family.

The current study provides the most inclusive phylogeny of *Phylloporus* to date, and provides data necessary for studies of character evolution in the genus. One of the questions that can be addressed is: Have lamellate hymenophores in the Boletaceae evolved from tubular hymenophores, or vice versa? If the former hypothesis is correct, then the lamellate hymenophore of *Phylloporus* species has evolved secondarily via morphological reduction and is convergent with lamellate hymenophores in the Agaricales.

Materials and Methods

SPECIMENS

The collections used in this study are listed in Table 3-1. In total, 20 species of *Phylloporus* and seven species of *Xerocomus* are included in the analysis. Sequences of the nuclear ribosomal DNA large subunit (nrDNA-LSU) were generated for 47 *Phylloporus* collections. An additional three LSU sequences were retrieved from GenBank. The LSU dataset also includes 14 sequences from *Xerocomus*, *Tylopilus*, *Boletus*, and *Aureoboletus*. Ribosomal DNA internal transcribed spacer (nrDNA-ITS) sequences were generated for 39 *Phylloporus* collections. Four ITS sequences were generated for *Xerocomus* and one was retrieved from GenBank.

Voucher material was studied and identified to morphological species. Both macromorphological and micromorphological characters were observed. The analyses include *Phylloporus* specimens from Australia, Belize, Costa Rica, Germany, Indonesia, Malaysia, Mexico, Panama, Slovakia, Thailand, and the United States.

MOLECULAR METHODS

Ninety-eight sequences were newly generated for this work, including 55 LSU sequences (47 from *Phylloporus*) and 43 ITS sequences (39 from *Phylloporus*).

The DNA was isolated from recent collections and from herbarium specimens. In the field, a small portion of the material to be used for DNA extraction was preserved in a tube with silica dessicant to be. For the herbarium specimens, a piece of the pileus was used in the extraction. In both cases DNA was extracted from samples using the E.Z.N.A.™ Fungal Miniprep Kit (Qiagen) after the sample of the specimen had been subjected to treatment in a Mini Beadbeater-8 cell disrupter (Biospec Products) to pulverize the material. The nrDNA-LSU sequences were amplified using primers LR5 and LR0R (Moncalvo et al. 2000) and cycle sequenced with these same primers plus the internal primers LR3R and LR3 (Vilgalys & Hester 1990). The primers ITS1-F (Gardes & Bruns 1993) and ITS4 (White et al. 1990) plus 5.8S and 5.8SR (Vilgalys & Hester 1990) were used for amplification and sequencing of the ITS regions (ITS1-5.8S-ITS2). PCR reactions were performed in 25 µl using *Taq* DNA polymerase under the following thermocycling conditions: initial denaturation at 94˚C for 2 min followed by 30 cycles of 94˚C for 30 sec, 55˚C for 1 min, and 72˚C for 1 min, followed by a final extension step of 72˚C for 5 min. Amplification products were purified using Pellet Paint (Novagen) or the QIAquick PCR Purification Kit (QIAGEN Inc.) following the manufacturers' instructions. Sequencing was performed using Big Dye 3.1 chemistry on an Applied Biosystems (3730xl) automated DNA sequencer.

The LSU sequences have proven to be most useful for molecular systematics of boletes at the level of order, genus, and species. This is an advantage because LSU sequences for most taxa in the boletes can be easily aligned (Grubisha et al. 2001, Binder & Hibbett 2002). The ITS region has been used in other genera of fungi with some reliability (Hughes et al. 2001). Although this region is quite variable and sometimes difficult to align across bolete genera, a more inclusive dataset might result in better resolution, especially at the species level.

The combined dataset included 58 *Phylloporus* terminals and 15 non-*Phylloporus* collections. LSU was not amplified successfully in 9 out of the 73 collections included in the analyses. The ITS sequences were not successfully amplified for 27 collections included in this dataset and were then treated as missing data.

The failure to extract amplifiable DNA was probably because of the following: (1) the specimens were not properly dried and (2) the humidity was too high where the specimens were stored. First, if the temperature while drying a specimen is too high, the quality of the DNA will be affected. Second, fungal collections must be stored in a dry location, which discourages insect and mold attacks. In tropical regions, preservation of fungi collections has been difficult for many herbaria because they lack the proper facilities. In many cases, to avoid damage by insects and mold, the herbaria have placed mothballs with the specimens. It is possible that the mothballs also influence DNA extraction.

ANALYTICAL METHODS

Sequence chromatograms were edited and contigs assembled using Sequencher 4.5 (GeneCodes, Ann Arbor, Michigan). Nucleotide sequences were aligned using Clustal X 1.83.1 (Thompson et al. 1997) and adjusted by eye using MacClade 4.08 (Maddison & Maddison 2005) where necessary.

The datasets were analyzed using parsimony (MP), maximum likelihood (ML), and Bayesian methods (MB) as described below.

Parsimony analyses were performed in PAUP* 4.0b4 (Swofford 2002). All characters were treated as unordered and equally weighted, with gaps treated as missing data. A heuristic search was performed using 1000 random-addition sequences with one tree held at each step, tree bisection-reconnection (TBR) branch swapping, Multrees option enabled, and branches collapsed when maximum branch length equaled zero. Maxtrees settings were 1000 for the LSU and the ITS analyses and 2000 for the combined analyses. Branch support was assessed using 1000 bootstrap replicates with full heuristic searches, one random addition sequence per bootstrap replicate, and saving one tree per random addition sequence. *Aureoboletus auriporus* was defined as the outgroup for the LSU and the combined dataset because it is a clade sister to the *Xerocomus subtomentosus* complex clade, where *Phylloporus* is included in previous phylogenetic analysis (Binder 1999, Binder & Hibbett 2006). For the ITS dataset, where *Aureoboletus* was not included due to difficulties in the alignment, *Xerocomus illudens*, a taxon basal to *X. subtomentosus* according to the same phylogenetic works (Binder 1999, Binder & Hibbett 2006), was used as the outgroup.

Prior to ML and MB searches, an optimal model of sequence evolution was selected with the program Model Test 3.7 (Posada & Crandall 1998), using one of the most parsimonious trees to evaluate the models. Maximum likelihood analyses were implemented in PAUP* 4.0b10 (Swofford 2002) and Bayesian analyses were implemented in MrBayes 3.1.2 (Huelsenbeck & Ronquist 2001). Posterior probabilities (PP) for the combined dataset were determined by running one cold and three heated chains for 2 million generations in parallel mode, saving trees every 100th generation, and 2000 suboptimal trees at the beginning of the runs were discarded (burn-in phase). For the LSU and ITS datasets, PP were determined by running one cold and three heated chains for 1 million generations in parallel mode, saving trees every 100th generation, with a burn-in phase of 2500 trees for the LSU dataset and 100 trees for the ITS dataset.

Results

PHYLOGENETIC ANALYSIS OF LSU SEQUENCES

The aligned LSU sequences included 64 sequences and 1505 characters with 1117 constant characters, 97 variable but parsimony-uninformative characters, and 294 parsimony-informative positions. Analyses yielded 1000 most parsimonious trees (L=1559 steps, CI=03374, RI=0.5836, RC=0.1969). The phylogenetic analysis using ML resulted in three highest-likelihood trees (ln likelihood=7471.53237), one of which is presented in Figure 3-1.

PHYLOGENETIC ANALYSIS OF ITS SEQUENCES

The ITS sequence alignment included 44 sequences of 797 characters of which 273 were constant, 162 variable but parsimony-uninformative, and 362 parsimony-informative (an additional 491 characters were excluded due to ambiguous alignment). The parsimony analysis was carried out for a restricted subset of 44 taxa (taxa which sequences were not alignable were excluded from the dataset), resulting in 1000 most parsimonious trees (L=1307 steps, CI=0.6320, RI=0.6913, RC=0.4369). The phylogenetic analysis using ML resulted in a single tree (ln likelihood=7273.17334), which is presented in Figure 3-2.

PHYLOGENETIC ANALYSIS OF COMBINED LSU AND ITS SEQUENCES

The combined data set (LSU + ITS) for 73 sequences included 2795 characters with 1588 constant characters, 357 parsimony-uninformative characters, and 850 parsimony-informative characters. The parsimony analysis resulted in 2000 most parsimonious trees (L=3354 steps, CI=0.6035, RI=0.6903, RC=0.4166). The phylogenetic analysis using ML resulted in three trees (xln likelihood=19894.71357), one of which is presented in Figure 3-3.

Discussion

The three methodologies used in this work showed similar results concerning the relationships among the species here included. The differences were observed in the clades where the support was not strong. One maximum likelihood tree is presented for each analysis and the discussion is mostly based on the results of the analysis of the two genes combined.

This study suggests that *Phylloporus* might be a monophyletic genus, closely related to *Xerocomus*. The sister group to *Phylloporus* contains *X. perplexus* and *X. gracilis*, while *X. subtomentosus* comes out in a basal grade to *Phylloporus*. The paraphyly of *Xerocomus* has been seen in other phylogenetic studies (Binder 1999, Binder & Hibbett 2006) and that genus is under examination by different groups of researchers (Peintner et al. 2003, Taylor et al. 2006).

The monophyly of the two isolates of *Phylloporus pelletieri*, the type species of the genus, in the phylogeny is supported by the LSU dataset and by the combined dataset (BS = 97% and 98% respectively), however the position of the species is not resolved with confidence. *Phylloporus pelletieri* came out in the tree in a clade distinct from *P. rhodoxanthus* and morphological data also support these as two distinct species. The American species, *P. rhodoxanthus*, has a lamellate hymenophore and encrusted cystidia, compared to the more alveolate hymenophore and smooth cystidia of *P. pelletieri*, a species from Europe.

Xerocomus subtomentosus, the type species of *Xerocomus*, was also included in the studies and it was resolved along with other *Xerocomus* species at the base of the tree (Figs. 3-1, 3-3).

Within *Phylloporus*, five species form distinct groups with good node support that can be recognized in each phylogeny (LSU, ITS, combined). Species that form these well supported groups (BS > 85%, PP > 0.98) are *P. leucomycelinus*, *P. centroamericanus*, *P. curvatus*, *P. dimorphus*, and *P. pumilus* (labeled from A to E on the trees).

Phylloporus dimorphus (Group B) and *P. curvatus* (Group A) are two new species that formed well supported groups and are closely related based on molecular data. They are both associated with *Castanopsis* forests in northern Thailand but can be morphologically differentiated by the two differently shaped spores of *P. dimorphus* and the light pink reaction of the flesh of *P. curvatus* when in contact with NH_4.

Phylloporus centroamericanus was the most frequently sampled species in this study, with sequences from seven collections included. This species is easily recognizable by its thick-walled cystidia, and it formed a consistent phylogenetic group (Group D) in the analyses. This is one of the most common *Phylloporus* species collected in oak forests in Costa Rica, followed by species in the *Phylloporus phaeoxanthus* complex.

The *P. phaeoxanthus* group (BS=99, PP=1) contains subspecies *phaeoxanthus* and subspecies *simplex*. *Phylloporus phaeoxanthus* ssp. *simplex* is a taxon included in this group that did not exhibit a significant difference at the molecular level; however, ssp. *simplex* can be morphologically distinguished by having incrusted cystidia, compared to the smooth cystidia seen on *P. phaeoxanthus*.

Phylloporus rhodoxanthus presented an interesting pattern, with the taxon placed in two different parts on the tree (Fig. 3-3). Three collections (MAN 98, MAN 99, JLM1808) were within the Thailand species group, with *P. curvatus* and *P. dimorphus*, while two collections (MAN75, MAN8714) were near *P. arenicola* and *P. phaeoxanthus*. However, it is interesting to note that when the gene sequences were analyzed in separate datasets, the position of these collections changed and they formed well-supported groups (BS >85%). In the LSU dataset, the collection JLM1808 from Alabama still came out related to the Thailand group, while the other four collections formed a group with a 90% BS support. When the ITS database was analyzed three *P. rhodoxanthus* collections formed a strongly supported group (BS=100, PP=1).

Phylloporus rhodoxanthus is the most common species in the United States and is found mainly on the east coast, where its distribution overlaps with the distribution of *P. leucomycelinus*. These two species have been confused in the field because *P. leucomycelinus* is not as well known, but they can be separated because *P. leucomycelinus* has a white basal mycelium.

Phylloporus pumilus is a species that forms a distinct group in the analyses (Group C). This is a species from Indonesia with an alveolate hymenophore, and is distinct from the other alveolate species by its diminutive size.

The *P. leucomycelinus* group (Group E) includes the Panamanian collection of *P. caballeroi*; unfortunately no other collections of *P. caballeroi* were successfully amplified for either LSU or ITS. The collection was observed and is correctly identified and distinct from *P. leucomycelinus*. One single collection of *P. leucomycelinus* (4582) fell outside the main group, suggesting a possibly contamination of the isolated DNA, since the specimen has been observed and it is correctly identified. Closely related to the *P. leucomycelinus* group is a new species from Costa Rica, *P. alborufus*, which also has white basal mycelium and a red colored pileus. This species was found in oak forests in Costa Rica and possibly migrated from the Northern Hemisphere to the montane neotropic oak forests as observed in other species (Halling 2001).

The sections proposed for *Phylloporus* by Heinemann & Rammeloo (1987) and by Singer (1945) and Singer et al. (1978) do not seem to follow a natural arrangement. The cited authors used morphological characters such as the oxidation of the flesh when exposed, spore ornamentation under scanning electron microscope, and the presence or absence of clamp connections to create these sections. Sections *Phylloporus*, *Sulphurei*, and *Manausensis* were erected by Singer for species from the Americas, while sections *Phylloporus* sensu Heinemann and Rammeloo, *Oxydabiles*, *Immutabiles*, *Tubipedes*, and *Fibulati* were erected based on African species. When these characters are mapped in the molecular phylogenies it is observed that they are distributed in different clades and do not form the basis of natural arrangements.

A larger sample that would include more species that have been classified in these sections would give a better idea of the relationships within *Phylloporus* and the formation of natural sections. Unfortunately, none of the clamp connection-bearing species (section *Fibulati*) were successfully amplified and therefore these could not be included in the molecular analysis. A geographic pattern was not observed, although the sequences of *Phylloporus* included in this work are from various regions of the world. Unfortunately the African species were not available to study, which constitutes a gap in the phylogenetic analyses.

Yellow pigments have been shown to be present in members of the Boletaceae in different parts of the basidiome (Besl & Bresinsky 1997). *Phylloporus* species present these pigments in the basal mycelium, which constitute a valuable taxonomic character in the genus. In *Phylloporus,* the color of the basal mycelium has been traditionally used to separate species, and it was interesting to note that the species with yellow basal mycelium formed two groups within the genus, being also present in *P. pelletieri* (Fig. 3-3). The white basal mycelium is present in the outgroup species used in this work, and appears to be plesiomorphic in *Phylloporus*.

The results suggest that yellow pigmentation in the basal mycelium is a character that has evolved three times (Fig. 3-3), or, alternatively, was gained once and lost four times, though the former explanation is more parsimonious.

This study might suggest that the lamellate hymenophore evolved once in *Phylloporus*, and it represents a reduction of the tubular hymenophore in the Boletaceae.

Large subunit sequence data have been shown to be useful for the delimitation of groups in Boletales (Binder 1999, Bresinsky et al. 1999) and its utility in delimiting

Phylloporus species within the genus and from *Xerocomus* is demonstrated in the present study. The previous species concept based on the lamellate hymenophore is confirmed by the results of the molecular phylogenetic analyses based on LSU, ITS, and the combined analyses of these two genes. Basal relationships are not resolved, as might be expected based on other studies using LSU only (Moncalvo et al. 2000, Moncalvo et al. 2002, Peintner et al. 2003). The lack of success in amplifying ITS sequences from many of the specimens is probably one of the reasons why better resolution was not acquired for the basal relationships of the combined LSU and ITS analysis presented herein. It is expected that the inclusion of other genes in the dataset will improve the results.

The phylogenetic analyses suggest that *Phylloporus* is a monophyletic group distinct from *Xerocomus*; however, further analyses are needed to determine if the genus is monophyletic because the resolution of the backbone of the tree is not completely resolved. Also, broader analyses including diverse genera in the family and more *Xerocomus* species from different groups are needed to answer this question.

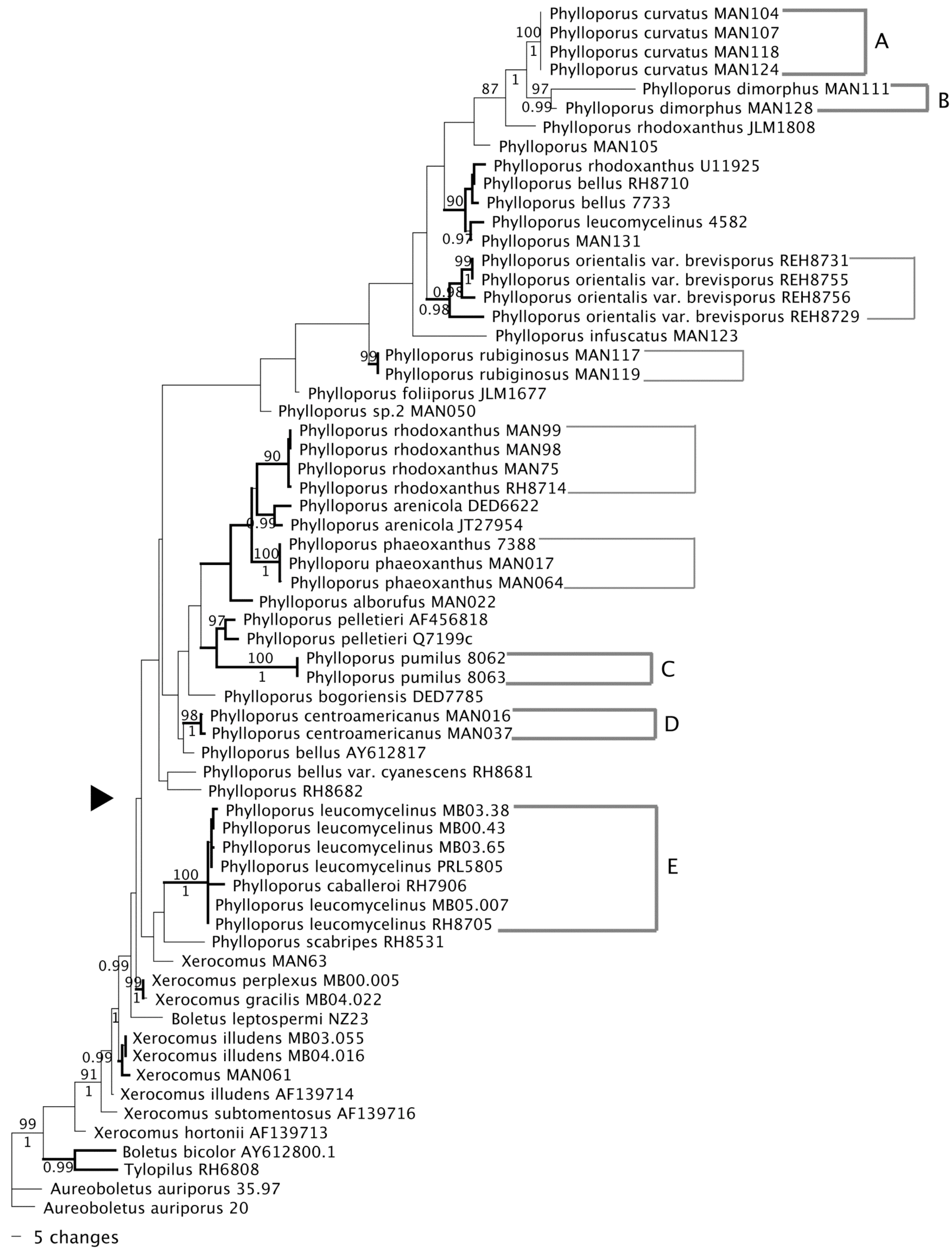

Figure 3-1: Phylogenetic relationships of *Phylloporus* inferred from rDNA-LSU sequences using maximum likelihood. Bootstrap values (>85%) are shown above the branches. Bayesian posterior probability values (probabilities >0.98) are shown below the branches. Branches present in both ML and MP trees are in bold. The letters on the right correspond to the groups mentioned in the text. *Aureoboletus auriporus* was used as outgroup. The arrow marks the node that separates *Phylloporus* from *Xerocomus*.

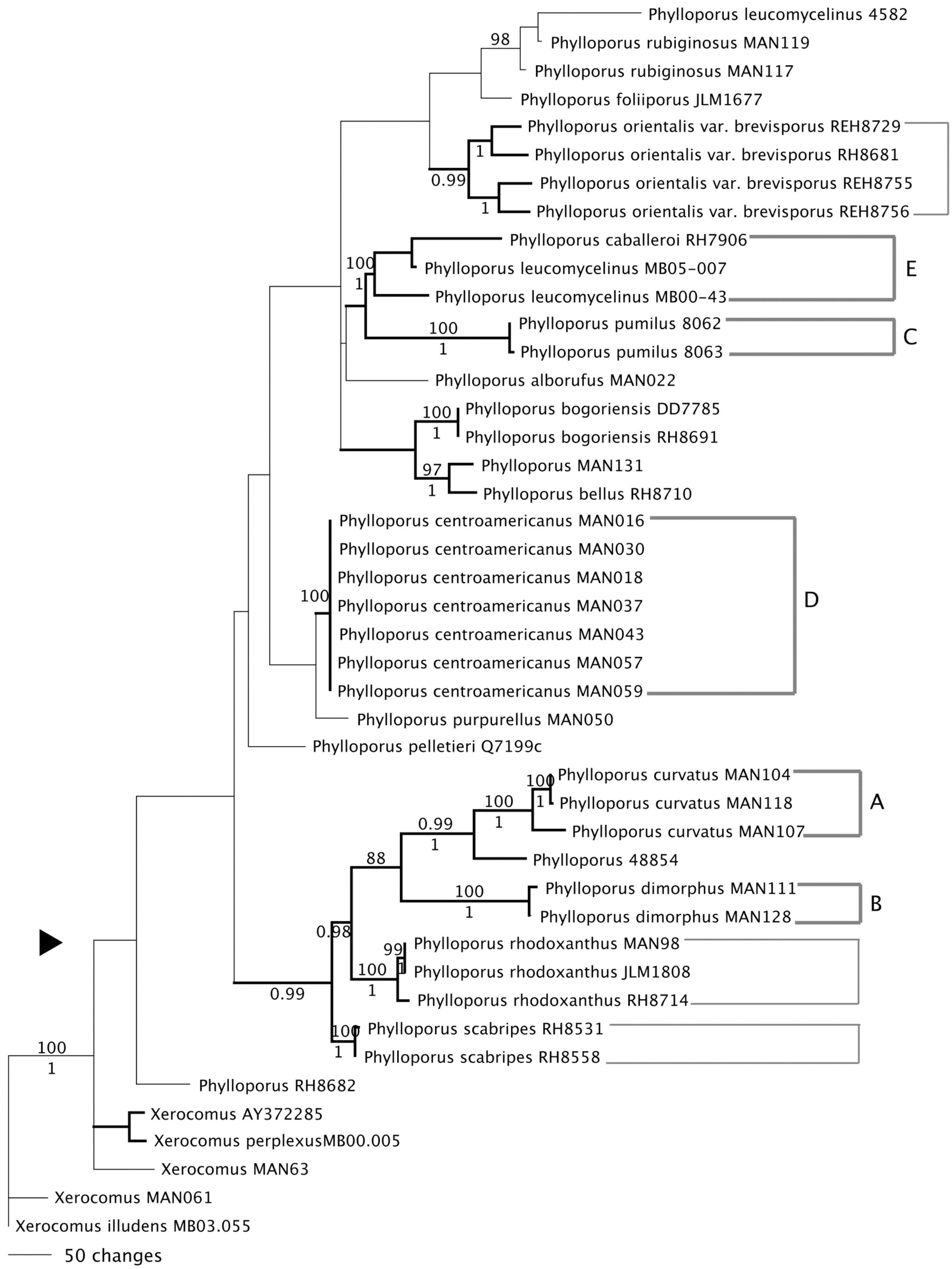

Figure 3-2: Phylogenetic relationships of *Phylloporus* inferred from rDNA-ITS sequences using maximum likelihood. Bootstrap values (>85%) are shown above the branches. Bayesian posterior probability values (probabilities >0.98) are shown below the branches. Branches present in both ML and MP trees are in bold. Groups labeled A – E are the same as in Figure 3-1. *Xerocomus illudens* was used as outgroup. The arrow marks the node that separates *Phylloporus* from *Xerocomus*.

Figure 3-3: Phylogenetic relationships of *Phylloporus* inferred from the combined rDNA-LSU and rDNA-ITS dataset using maximum likelihood. Bootstrap values (>85%) are shown above the branches. Bayesian posterior probability values (probabilities >0.98) are shown below the branches. Branches present in both ML and MP trees are in bold. The groups within the dashed lines represent taxa with yellow basal mycelium. Groups labeled A – E are the same as in Figure 3-1. *Aureoboletus auriporus* was used as outgroup. The arrow marks the node that separates *Phylloporus* from *Xerocomus*.

Table 3-1: Material included in the phylogenetic analysis of *Phylloporus* with the country of provenance, the respective GenBank accession numbers for LSU and ITS sequences, and the herbarium and collection numbers. Some sequences were retrieved from GenBank.

Species	Origin	GenBank accession # LSU	ITS	Collection number
Aureoboletus auriporus (Peck) Pouzar	USAeast	DQ534636		**35.97**
Aureoboletus auriporus (Peck) Pouzar	Costa Rica	x		**MAN20**
Boletus bicolor Peck	USAeast	AY612800		**TH6933**
Boletus leptospermi McNabb	New Zealand	DQ534632		**NZ23**
Phylloporus alborufus Neves & Halling	Costa Rica	x	x	**MAN22**
Phylloporus arenicola A.H. Sm. & Trappe		x		**JT27954**
Phylloporus arenicola A.H. Sm. & Trappe	USAwest	x		**DED6622**
Phylloporus bellus (Massee) Corner	USAeast	x	x	**REH8710**
Phylloporus bellus (Massee) Corner	Japan	**AY612817**		**MCA559**
Phylloporus bellus (Massee) Corner	Costa Rica	x		**REH7733**
Phylloporus bellus var. *cyanescens* Corner	Australia	x	x	**REH8681**
Phylloporus bogoriensis Höhn.	Indonesia	x	x	**DED7785**
Phylloporus bogoriensis Höhn.	Malaysia		x	**REH8691**
Phylloporus caballeroi Singer	Panama	x	x	**REH7906**
Phylloporus centroamericanus Singer & L.D. Gómez	Costa Rica	x	x	**MAN16**
Phylloporus centroamericanus Singer & L.D. Gómez	Costa Rica		x	**MAN18**
Phylloporus centroamericanus Singer & L.D. Gómez	Costa Rica		x	**MAN30**
Phylloporus centroamericanus Singer & L.D. Gómez	Costa Rica	x	x	**MAN37**
Phylloporus centroamericanus Singer & L.D. Gómez	Costa Rica		x	**MAN43**
Phylloporus centroamericanus Singer & L.D. Gómez	Costa Rica		x	**MAN57**
Phylloporus centroamericanus Singer & L.D. Gómez	Costa Rica		x	**MAN59**
Phylloporus curvatus Neves & Halling	Thailand	x	x	**MAN104**
Phylloporus curvatus Neves & Halling	Thailand	x	x	**MAN107**
Phylloporus curvatus Neves & Halling	Thailand	x	x	**MAN118**
Phylloporus curvatus Neves & Halling	Thailand	x		**MAN124**
Phylloporus dimorphus Neves & Halling	Thailand	x	x	**MAN111**
Phylloporus dimorphus Neves & Halling	Thailand	x	x	**MAN128**
Phylloporus foliiporus (Murrill) Singer	USAeast	x	x	**JLM1677**
Phylloporus infuscatus Neves & Halling	Thailand	x		**MAN123**
Phylloporus leucomycelinus (Singer & M.H. Ivory) Singer	USAeast	x	x	**MB00-043**
Phylloporus leucomycelinus (Singer & M.H. Ivory) Singer	USAeast	x		**REH8705**
Phylloporus leucomycelinus (Singer & M.H. Ivory) Singer	USAeast	x	x	**MB05-007**
Phylloporus leucomycelinus (Singer & M.H. Ivory) Singer	USAeast	x		**MB03-65**
Phylloporus leucomycelinus (Singer & M.H. Ivory) Singer	USAeast	x	x	**REH4582**
Phylloporus leucomycelinus (Singer & M.H. Ivory) Singer	USAeast	x		**PRL5805**
Phylloporus leucomycelinus (Singer & M.H. Ivory) Singer	USAeast	x		**MB03-038**
Phylloporus orientalis Corner	Australia	x		**REH8731**
Phylloporus orientalis Corner	Australia	x	x	**REH8755**
Phylloporus orientalis Corner	Australia	x	x	**REH8756**
Phylloporus pelletieri (Lév.) Quél.	Germany	**AF456818**		**Pp1**
Phylloporus pelletieri (Lév.) Quél.	Slovakia	x	x	**Q7199c**
Phylloporus phaeoxanthus Singer & L.D. Gómez	Costa Rica	x		**MAN17**
Phylloporus phaeoxanthus Singer & L.D. Gómez	Costa Rica	x		**MAN64**
Phylloporus phaeoxanthus ssp simplex Singer & L.D. Gómez	Costa Rica	x		**REH7388**
Phylloporus pumilus Neves & Halling	Indonesia	x	x	**REH8063**

Table 3-1: Cont

Species	Origin	GenBank accession # LSU	ITS	Collection number
Phylloporus pumilus Neves & Halling	Indonesia	x	x	**REH8062**
Phylloporus purpurellus Singer	Costa Rica	x	x	**MAN50**
Phylloporus Quél.	Australia	x	x	**REH8682**
Phylloporus Quél.	Thailand	x		**MAN105**
Phylloporus Quél.	Thailand	x	x	**MAN131**
Phylloporus Quél.	China		x	**48854**
Phylloporus rhodoxanthus (Schwein.) Bres.	USAeast	x	x	**JLM1808**
Phylloporus rhodoxanthus (Schwein.) Bres.	USAeast	**U11925**		**SAR 89.457**
Phylloporus rhodoxanthus (Schwein.) Bres.	USAeast	x		**MAN75**
Phylloporus rhodoxanthus (Schwein.) Bres.	USAeast	x		**MAN99**
Phylloporus rhodoxanthus (Schwein.) Bres.	USAeast	x	x	**REH8714**
Phylloporus rhodoxanthus (Schwein.) Bres.	USAeast	x	x	**MAN98**
Phylloporus rubiginosus Neves & Halling	Thailand	x	x	**MAN117**
Phylloporus rubiginosus Neves & Halling	Thailand	x	x	**MAN119**
Phylloporus scabripes Ortiz, T.J. Baroni & Neves	Belize	x	x	**REH8531**
Phylloporus scabripes Ortiz, T.J. Baroni & Neves	Belize		x	**REH8558**
Phylloporus sp.1 sensu Watling	Australia	x	x	**REH8729**
Tylopilus P. Karst.	Australia	x		**REH6808**
Xerocomus gracilis (A.H. Sm. & Thiers) L.D. Gómez	USAeast	x		**MB04-022**
Xerocomus hortonii (Sm. & Thiers) Binder & Besl	USAeast	AF139713		**84.94**
Xerocomus illudens (Peck) Singer	USAeast	**AF139714**		**64.98**
Xerocomus illudens (Peck) Singer	USAeast	x	x	**MB03-055**
Xerocomus illudens (Peck) Singer	USAeast	x		**MB04-016**
Xerocomus perplexus A.H. Sm. & Thiers	USAeast	x	x	**MB00-005**
Xerocomus Quél.			**AY372285**	**not known**
Xerocomus Quél.	Costa Rica	x	x	**MAN61**
Xerocomus Quél.	Costa Rica	x	x	**MAN63**
Xerocomus subtomentosus (L.) Fr.	Germany	**AF139716**		**Xs1**

References

Besl, H. and Bresinsky, A. 1997. Chemosystematics of Suillaceae and Gomphidiaceae (suborder Suillineae). Plant Systematics and Evolution 206: 223-242.

Binder, M. 1999. Zur molekularen systematik der Boletales: Boletineae und Sclerodermatineae subordo nov. Ph.D. University of Regensburg, pp. 147.

Binder, M. and Bresinsky, A. 2002. Derivation of a polymorphic lineage of Gasteromycetes from boletoid ancestors. Mycologia 94: 85-98.

Binder, M. and Hibbett, D.S. 2006. Molecular systematics and biological diversification of Boletales. Mycologia 98: 971-981.

Bresinsky, A. and Besl, H. 2003. Beiträge zu einer Mykoflora Deutschlands: Schlüssel zur Gattungsbestimmung der Blätter-Leisten- und Röhrenpilze. Regensburger Mykologische Schriften 11: 5-236.

Bresinsky, A., Jarosch, M., Fischer, M., Schonberger, I. and Wittmann-Bresinsky, B. 1999. Phylogenetic relationships within *Paxillus* s.l. (Basidiomycetes, Boletales): separation of a Southern hemisphere genus. Plant Biology 1: 327-333.

Corner, E.J.H. 1970. *Phylloporus* Quél. and *Paxillus* Fr. in Malaya and Borneo. Nova Hedwigia 20: 793-822.

Corner, E.J.H. 1972. *Boletus* in Malaysia. ed. Government Printing Office, Singapore. 263 pp.

Gardes, M. and Bruns, T.D. 1993. ITS primers with enhanced specificity for basidiomycetes--application to the identification of mycorrhizae and rusts. Molecular Ecology 2: 113.

Grubisha, L.C., Trappe, J.M., Molina, R. and Spatafora, J.W. 2001. Biology of the ectomycorrhizal genus *Rhizopogon*. V. Phylogenetic relationships in the Boletales inferred from LSU rDNA sequences. Mycologia 93: 82-89.

Halling, R.E. 1999. New *Leccinum* species from Costa Rica. Kew Bulletin 54: 747-753.

Halling, R.E. 2001. Ectomycorrhizae: Co-evolution, significance, and biogeography. Annals of the Missouri Botanical Garden 88: 5-13.

Halling, R.E. and Mueller, G.M. 2002. Agarics and boletes of neotropical oakwoods. In Tropical Mycology Vol. 1 (Eds, Watling, R., Frankland, J.C., Ainsworth, A.M., Isaac, S. and Robinson, C.H.) CAB International, pp. 1-10.

Heinemann, P. and Rammeloo, J. 1986. *Phylloporus* (Boletineae). Flore Illustreé des Champignons d'Afrique Centrale 13: 277-309.

Heinemann, P. and Rammeloo, J. 1987. *Phylloporus* (Boletineae). Flore Illustreé des Champignons d'Afrique Centrale 13: 277-309.

Huelsenbeck, J.P. and Ronquist, F.R. 2001. MrBayes: Bayesian inference of phylogeny. Bioinformatics 17: 754-755.

Ladurner, H. and Simonini, G. 2003. *Xerocomus* s.l. 8. ed. Edizioni Candusso, Alassio. 527 pp.

Maddison, W.P. and Maddison, D.R. 2005. MacClade: Analysis of phylogeny and character evolution. Sinauer Associates, Sunderland.

Moncalvo, J.-M., Lutzoni, F.M., Rehner, S.A., Johnson, J. and Vilgalys, R. 2000. Phylogenetic relationships of agaric fungi based on nuclear large subunit ribosomal DNA sequences. Systematic Biology 49: 278-305.

Moncalvo, J.-M., Vilgalys, R., Redhead, S.A., Johnson, J.E., James, T.Y., Aime, M.C., Hofstetter, V., Verduin, S.J.W., Larsson, E., Baroni, T.J., Thorn, R.G., Jacobsson, S., Clémençon, H. and Miller, O.K.J. 2002. One Hundred and Seventeen Clades of Euagarics. Molecular Phylogenetics and Evolution 23: 357-400.

Montoya, L. and Bandala, V.M. 1991. Studies on the genus *Phylloporus* in Mexico, I. Discussion of the known species and description of a new species and a new record. Mycotaxon 41: 471-482.

Mueller, G.M. and Halling, R.E. 1995. Evidence for high diversity of Agaricales (Fungi) in neotropical montane *Quercus* forests. In Biodiversity and conservation of neotropical montane forests (Ed, Churchill, S., Balslev, H, Forero, E, Luteyn, J) New York Botanical Garden, Bronx, pp. 302-312.

Pegler, D.N. and Young, T.W.K. 1981. A natural arrangement of the Boletales, with reference to spore morphology. Transactions of the British Mycological Society 76: 103-146.

Peintner, U., Ladurner, H. and Simonini, G. 2003. *Xerocomus cisalpinus* sp. nov., and the delimitation of species in the *X. chrysenteron* complex based on morphology and rDNA-LSU sequences. Mycological Research 107: 659-679.

Posada, D. and Crandall, K.A. 1998. Modeltest: testing the model of DNA substitution. Bioinformatics applications 14: 817-818.

Singer, R. 1945. The Boletineae of Florida with notes on extralimital species. II. the Boletaceae (Gyroporoideae). Farlowia 2: 223-303.

Singer, R. 1978. Notes on Bolete taxonomy II. Persoonia 9: 421-438.

Singer, R. and Gómez, L.D. 1984. The Basidiomycetes of Costa Rica. III. The genus *Phylloporus* (Boletaceae). Brenesia 22: 163-181.

Singer, R., Ovrebo, C.L. and Halling, R.E. 1990. New species of *Phylloporus* and *Tricholomopsis* from Colombia, with notes on *Phylloporus boletinoides*. Mycologia 82: 452-459.

Smith, A.H. and Thiers, H.D. 1971. The Boletes of Michigan. ed. University of Michigan Press, Ann Arbor. 428 pp.

Swofford, D.L. 2002. PAUP*. Phylogenetic analysis using parsimony (*and other methods), version 4.0b10 (Altivec). Sinauer Associates, Sunderland.

Taylor, A.F.S., Hills, A.E., Simonini, G., Both, E.E. and Eberhardt, U. 2006. Detection of species within the *Xerocomus subtomentosus* complex in Europe using rDNA–ITS sequences. Mycological Research 110: 276-287.

Thompson, J.D., Gibson, T.J., Plewniak, F., Jenmougin, F. and Higgins, D.G. 1997. The CLUSTAL X windows interface: flexible strategies for multiple sequence alignment aided by quality analysis tools. Nucleic acids research 24: 4876-4882.

Vilgalys, R. and Hester, M. 1990. Rapid genetic identification and mapping of enzimatically amplifiedribosomal DNA from several *Cryptococcus* species. Journal of Bacteriology 172: 4238-4246.

White, T.J., Bruns, T.D., Lee, S. and Taylor, J.W. 1990. Amplification and direct sequencing of fungal ribosomal RNA genes for phylogenetics. In PCR protocols: a guide to methods and applications (Eds, Innis, M.A., Gelfand, D.H., Sninsky, J.J. and White, T.J.) Academic Press, San Diego, pp. 315-322.

EPILOGUE

Even though it is one of the most diverse genera in the Boletaceae, *Phylloporus* has received little previous study beyond descriptions of taxa within restricted geographical areas. *Phylloporus* was originally created to accommodate the European species *Agaricus pelletieri*. Since then, 70 species have been described and the genus is now known to occur in every Hemisphere. The majority of these species are native to the tropics though some, including the type species, are from temperate regions. Over the past four years while working on this dissertation, I studied the systematics, taxonomy, and molecular phylogeny of the *Phylloporus* taxa that I gathered from field trips, other mycologists, and herbaria within and outside of the United States. This work includes 28 species and represents the most comprehensive synthesis of *Phylloporus* to date.

The results of this study suggest that *Phylloporus* is a monophyletic genus closely related to *Xerocomus*. A reduction to synonymy with *Xerocomus* has been recently suggested based on a molecular phylogeny that included *P. pelletieri* (the European type) and *P. rhodoxanthus* (from the United States). However, molecular studies have shown that *Xerocomus* is polyphyletic, suggesting that placing the two genera in synonomy would make their taxonomy more confusing. These two points – the apparent monophyly of *Phylloporus* and the taxonomic confusion resulting from synonomy with *Xerocomus* – present compelling arguments for maintaining *Phylloporus* as a distinct genus.

The morphological variation seen in the lamellae and spores of *Phylloporus,* combined with the phylogenetic position of the genus within the Boletaceae, hint at several interesting questions regarding trends in character evolution within the genus and family.

However, these questions cannot be adequately addressed until a wider sampling of *Phylloporus* species are studied. While the present study compares the broadest geographical sample of *Phylloporus* to date, it nonetheless lacks collections from several regions that are key to understanding evolutionary patterns in the genus; most critical is the lack of African taxa, which were not available for study during the course of this dissertation. It is important that future works on *Phylloporus* include the African species. The distribution maps generated from this study (e.g., Figure 2-32) clearly show gaps where no collections of *Phylloporus* have been made. It is highly likely that some of these gaps result simply from lack of collecting effort, since many of these regions have forests similar to those in areas where *Phylloporus* has been collected. It is likely that the neotropics contain additional, undiscovered diversity, as work in just a few regions (Costa Rica, Colombia, and Mexico) have generated a number of new *Phylloporus* collections and species. More collections are needed from Central and South America to help fully resolve the taxonomy of this genus. Based on the limited field work of a few other workers and myself, the same situation appears to be true for Southeast Asia; of the six newly described species presented in this work, four were collected in Thailand during a single 10-day field trip in 2006. The study of morphological characters combined with molecular phylogenetic analysis will only help clarify aspects of the taxonomy and evolution of the genus and its relationships if a broader sample (both geographically and taxonomically) is studied.

Despite its limitations, this is the first comparative study to sample *Phylloporus* from various regions of the world and the first to include tropical *Phylloporus* species in a phylogenetic analysis. The number of species included is slightly less then 30% of the total number of described species in the genus, so it is quite possible that the phylogenetic relationships presented herein are an accurate, if rough, representation of evolutionary trends. It is my hope that the results presented in this study will stimulate further research on this group of macromycetes.