

## 1.0 INTRODUCTION

Malaysia is considered as one of the world's twelve mega diverse countries for most of the groups of organisms (Malaysia's National Policy on Biological Diversity, 1998). The forests in Malaysia are the most complicated in the world (Corner, 1972). Yet the diversity of macrofungi in Malaysia is poorly known. There is no accurate figure for the fungal diversity in Malaysia, and many taxonomic groups remains undocumented for the country (Noorlidah *et al.*, 2005a; Noorlidah *et al.*, 2005b). In Malaysia, the earlier records of macrofungi were by Chipp (1921). Corner (1970, 1972) had documented the Malaysian bolete diversity with 140 species from Peninsular Malaysia of which 100 were new to science. Later, Corner (1974) had described another 20 additional species from Borneo. However, Corner (1972) predicted the existence of 300 bolete species in Malaysia. There were new records and clarifications from Malaysia by Watling and Hollands (1990) and Watling (2000). According to Lee (2005), 66% of the species of ectomycorrhizal fungi from Malaysia are undescribed. One of the major factors hindering identification of fungi in Malaysia is a lack of monographic treatments of genera (Lee *et al.*, 2003; Lee, 2005). Therefore, strengthening the systematic studies on boletes in Malaysia is important for sufficient and accurate documentation their diversity.

Corner (1972) had four genera of boletes (*Boletus*, *Gyroporus*, *Heimiella*, *Strobilomyces*), with the remaining distributed in subgenera (*Austroboletus*, *Boletellus*, *Ixocomus*, *Leccinum*, *Pulveroboletus*, *Tylopilus* and *Xerocomus*). Pegler and Young (1981) presented a new classification of boletes based on spore morphology and recognized 35 genera in six families. On the other hand, Singer (1981, 1986) presented an overview of bolete genera in one family where he recognized 26 genera world wide. From the 26 genera of boletes (*sensu* Singer, 1986), they are now distributed in four families as Boletaceae, Boletinellaceae, Gyroporaceae, and Suillaceae. The Boletaceae

family has been variously interpreted by different authorities to include as many as 31 different genera (Halling *et al.*, 2007).

Boletes are freshly pored mushrooms in the order Boletales. This order includes some gilled mushrooms in the families Gomphidiaceae and Paxillaceae, which have the same fleshy texture and similar micromorphological and molecular phylogenetic characteristics (Hawksworth *et al.*, 1995; Seehanan *et al.*, 2008). The Boletaceae is a monophyletic family of macrofungi with largely fleshy basidiomata, with tubular hymenium positioned on the underside of the pileus (Corner, 1972; Halling *et al.*, 2007). The Boletaceae is composed of 26 genera and 415 species (Singer, 1986). The boletes are important component of the biogeochemical cycle of the tropical forests because they form ectomycorrhizae with several genera of trees in tropical forest ecosystems (Lee *et al.*, 1997, 2002; Lee, 2005; Watling *et al.*, 1995). In Malaysia, mycorrhizal trees such as Dipterocarpaceae are predominant in lowland forests (Lee *et al.*, 1995, 1997, 2002; Lee and Chang, 2003; Watling, 1994; Watling and Lee, 1995, 1998, 1999) followed by other families of trees such as Fagaceae, Leguminosae and Myrtaceae (Lee, 2005). About 90% of the species from Boletaceae are ectomycorrhizal, and may represent 18- 25% of all ectomycorrhizal fungi (Halling *et al.*, 2007). Boletes are the second components only to Russulaceae to form ectomycorrhizal symbioses in lowland dipterocarp forests of Peninsular Malaysia (Lee *et al.*, 2002). The humidity and weather condition in tropical forests support a wide diversity of boletes. However, problems are encountered when documenting their diversity in tropical forests due to the seasonality of fruiting, short-lived and solitary fruiting bodies (Corner, 1972; Tan *et al.*, 2007).

In this study, the morphological characteristics including macromorphology and micromorphology are used to differentiate and delimit the boletes species.

## **1.1 OBJECTIVES OF STUDY**

The objectives were:

- a. to document the biodiversity of Boletaceae in diverse habitats in Peninsular Malaysia.
- b. to describe new records and species from Peninsular Malaysia.

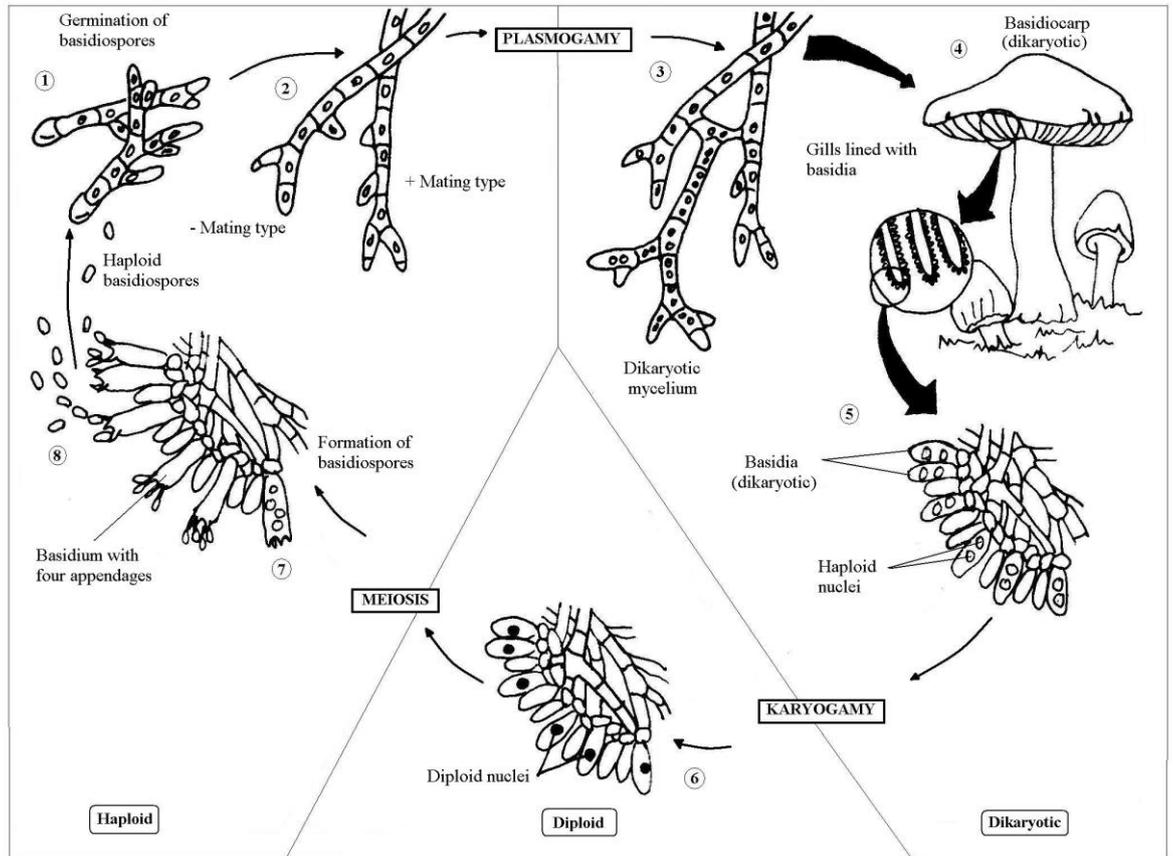
## 2.0 LITERATURE REVIEW

### 2.1 Basidiomycetes and its life cycle

Basidiomycetes are fungi that typically form large fruiting bodies visible without the aid of a microscope. Fungi are the third natural kingdom, just as important as animals and plants. As work on the ecology of fungi proceeds, it has become apparent that the world of plants is incredibly dependent on fungi (Philips *et al.*, 2006). Fungi break down leaf litter and dead wood and thus ensure that the surface of the world has a fertile layer of soil rather than being a heap of detritus. It is through an intimate relationship of fungi with the roots of the trees and plants, the mycorrhizal relationships, that the most important contribution is made (Philips *et al.*, 2006).

The life cycle of basidiomycetes (Figure 2.1) can be divided into three essential phases: basidiospore, vegetative mycelium and the fruiting body (Miles *et al.*, 1997). Haploid basidiospores germinate in a suitable environment and grow into short-lived haploid mycelia. Undifferentiated hyphae from two haploid mycelia of opposite mating type undergo plasmogamy, creating a dikaryotic mycelium that grows faster and ultimately crowds out, the parent haploid mycelium. The mycelium of some mushrooms form mycorrhizae with trees. Environmental cues such as rain, temperature changes, and, for mycorrhizal species, seasonal changes in the plant host, induce the dikaryotic mycelium to form compact masses that develop into mushrooms. Cytoplasm streaming in the mycelium and from the attached mycorrhizae swells the hyphae of mushrooms, causing them to "pop up" overnight. The dikaryons of basidiomycetes are long-lived, generally producing a new crop of basiocarps each year. Karyogamy occurs in the terminal dikaryotic cells that line the surfaces of the gills or pores. Each cell swells to form a diploid basidium, which rapidly undergoes meiosis and yields two or four haploid nuclei. The basidium then grows four appendages, and one haploid nucleus enters each appendage and develops into a basidiospore. When mature, the basidiospores are

propelled slightly (by electrostatic forces) into the spaces between the gills or pores. After the spores drop below the cap, they are dispersed by the wind (Miles *et al.*, 1997).

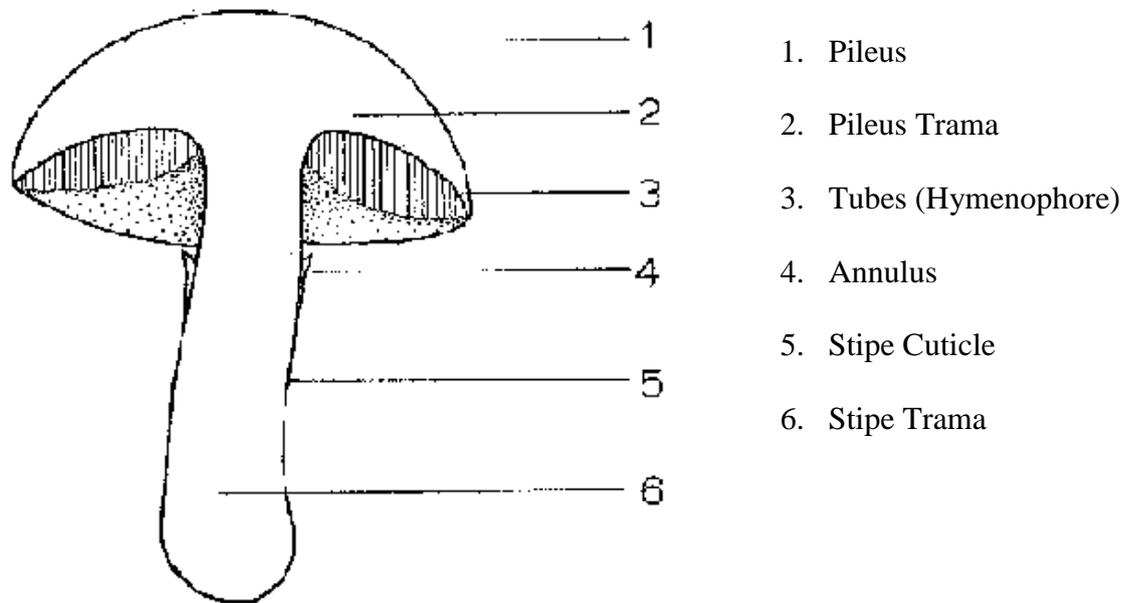


**Figure 2.1:** Life cycle of basidiomycetes. (modified from Campbell *et al.*, 2000)

### 2.1.1 Boletaceae

Boletaceae (known as boletes) belong to the order Boletales (phylum Basidiomycota, kingdom Fungi). The boletes are characterized primarily by the formation of spores, or reproductive bodies, on a highly specialised, microscopic structure known as a basidium (plural, basidia). Boletes belong to the group of Homobasidiomycetes and their basidium is unicellular. The fruit body of the boletes is similar in appearance to the typical mushroom except that, in the boletes, tubes have replaced the lamellae, on the under surface of the cap (Figure 2.2). Because of the

presence of these tubes and their pores, boletes are often called fleshy pore fungi (Thiers, 1975).



**Figure 2.2:** Longitudinal section through bolete basidiocarp. (Thiers, 1975)

## 2.2 Taxonomy history of Boletaceae in Malaysia

In Malaysia, the earlier records of macrofungi were by Chipp (1921). However, the descriptions are simple. Most of the species were described in short descriptions and the type species were unknown. Without question, the monographic works of Corner (1970, 1972, 1974) are the standards from which assessments of Malaysian bolete diversity are measured. One hundred new species out of 140 were described in his compilation of *Boletus* in Malaysia (Corner, 1972). In a week's collecting in Sarawak, Corner (1974) recorded 20 species of boletes among which six were undescribed. Corner (1972) predicted that the existence of 300 bolete species in Malaysia was easily possible. This figure is close to an all too conservative global estimate (Hawksworth *et al.*, 1995). In a Benefactors' Lecture to the British Mycological Society, Corner (1993) rhetorically suggested that the numbers of hymenomycetes in Malesia might be three or four

thousand. According to Lee (2005), 66% of the species of ectomycorrhizal fungi are undescribed for Malaysia. If this figure were applied just to the boletes, then we might expect numbers to approach 500 species. Thus, by necessity, when attempting any determination of a bolete specimen, a first appraisal must involve a look at Corner's (1970, 1972, 1974) treatises. According to Lee *et al.* (2003) and Lee (2005), one of the major factors hindering identification is lack of monographic treatments of genera, although there have been some new records, clarifications, and determinations as newer material has become available (Watling and Hollands, 1990; Watling, 2000). As with many understudied groups of organisms, especially those with high potential of occurring in unexplored habitats, accurate identification is hampered by the apparent high diversity and insufficient documentation of this diversity (Corner, 1972). Therefore, a strengthening of systematic studies on boletes in Malaysia is important for a sufficient and accurate documentation of this diversity.

### **2.3 Ecology and distribution**

In most cases, basidiomata of boletes are found in forest and woodland communities where they have been implicated or proven to be ectomycorrhizal symbionts (Singer, 1986). Such is the case for the lowland forests of Dipterocarpaceae predominant in Malaysia (Lee *et al.*, 1995, 1997, 2002, 2003; Watling, 1994; Watling and Lee, 1995, 1998, 1999) along with other families of trees represented by Fagaceae, Leguminosae, and Myrtaceae (Lee, 2005).

Since the majority of boletes is assumed to be ectomycorrhizal, they would then play a significant role in forest health and maintenance by facilitating water and nutrient uptake and such has been documented for Malaysian dipterocarps (Lee and Alexander, 1994). Corner (1935, 1993) studied the systematics of tropical fungi in Malaysia, Singapore, Borneo and the Solomon Islands during 1930s. He reported that there are two

rainy seasons in Singapore and Malaysia: March to May and August to October or December. During the rainy season, the advent of heavy rain will stimulate the dormant mycelium of boletes to immediate production of basidiomata. Because of the bimodal seasonality first documented by Corner (1935), diligent surveillance and continued monitoring over extended time periods are of high priority in order to assess accurately the true diversity, ecological implications, and distribution of these fungi. Lee *et al.* (2003) continue to assess and document these associations in Malaysian forests.

## **2.4 Mycorrhizal Associations**

The boletes are forest-inhabiting fungi. The restriction to the forests is due to the necessity of the formation of mycorrhizal associations in order to survive. This type of association is an intimate relationship between the vegetative mycelium of the fungus and the feeder roots of the associated tree. The mushroom profits by obtaining nutrients and water from the host tree. The benefits afforded to the tree by the fungus are by accumulating certain minerals from the soil that are subsequently available to the tree and increase the absorptive surface of the roots. Obviously, if such associations are essential for the survival or development of the tree, these fungi assume considerable significance in reforestation practices (Thiers, 1975).

It is difficult to trace the connection between the basidiocarp and the roots of the tree since the fungal hyphae are microscopic. Therefore, most mycorrhizal associations are only presumed and cannot easily be confirmed except by cultural practices. Such assumptions are based upon repeated observations that a specific bolete always occurs in close proximity to the same kind of tree. However, DNA sequence comparisons have confirmed specific associations (Osmundson *et al.*, 2007). Some species appear capable

of forming mycorrhizal associations with a rather wide range of trees; others are seemingly highly restricted (Thiers, 1975).

## **2.5 Morphological characters (Largent *et al.*, 1977; Thiers, 1975; Smith *et al.*, 1071)**

### **2.5.1 Macromorphological characters**

#### **2.5.1.1 Pileus and context**

Bolete pilei are similar to most large agaric genera (Corner, 1972). They are typically large, often reaching 15 cm or more in diameter, and rarely as small as 2 – 4 cm in diameter. Characteristically, they are more or less convex or bulbous in outline when young, becoming plane or plano-convex when mature. Colors range from almost black to many different shades of brown, pink, or bright red. The colors, with a few conspicuous exceptions, are relatively constant for a given species and change only slightly as the basidiocarp matures. When the surface of the pileus is bruised, many boletes show color change (Thiers, 1975).

The nature of the surface of the pileus is of considerable taxonomic significance in the boletes (Thiers, 1975, Corner, 1972). The surface may vary from glabrous, or bare, to fibrillose, to velvety, to tomentose. In some species, the pileus might display a squarrose or squamulose appearance. When they grow older, they become split, or rimose on the margin and become checked, or areolate. As periods of dry weather can be prolonged, the areolations sometimes become deep and strongly pronounced which results in a frustose condition of the pileus.

The margin or edge of the young pileus in most boletes is entire, smooth and even and with no ornamentations. The flesh or context of the pileus is usually soft and spongy, and with a high water content. The context is some shade of yellow or white, and rarely with other colors such as pink or black.

### 2.5.1.2 Hymenophore

The most distinctive feature of boletes is the hymenophore, which consists of soft, moist, putrescent tubes. The hymenium forms the inner lining of these tubes. The pores of the tubes are mostly angular, varying from almost square to rectangular, and range in size from 0.5 – 2 mm in diameter. In a few boletes such as *Phylloporus*, the pores are so large and elongated that they resemble lamellae and are described as lamellate (Corner, 1970; Thiers, 1975). The pores are usually concolorous with the tubes.

### 2.5.1.3 Stipe

The stipe of boletes are usually central and rarely have an eccentric attachment, except in *Gastroboletus*. The overall shape of the stipe is equal to subclavate or clavate. The color of the bolete stipe ranges from white to yellow to pink to darker colors, such as red, brown, or almost black. Frequently the background color is overlain by some other pigment, resulting in a blending of colors. Ornamentations on the surface of the stipe are often a contrasting color, and usually darken with age.

The surface of the stipe is dry, moist or viscid. Typically, it is glabrous, tomentose, fibrillose, or fibrillose-scaly and reticulate. The ridges forming the reticulum are composed of basidia and cystidia or can be an extension of the tubes down the stipe. The base of the stipe is often clothed with distinctively coloured mycelium. Another important taxonomic feature associated with the surface of the stipe is an annulus or a partial veil.

The context of the stipe is typically the same as in the pileus. The sequence of color changes in the stipe apex when exposed is of considerable taxonomic value in boletes. The stipe is usually solid, but in a some genus, such as *Gyroporus*, it is hollow, at least in the basal portion (Thiers, 1975).

#### 2.5.1.4 Chemical reactions

Chemical tests are sometimes used to rapidly distinguish between closely-related or morphologically similar species of mushrooms, or characters to group species into subsections of a genus. Pigments present in the fungal hyphae are dissolved or react differently with various chemicals, and the color reactions may be used as taxonomic characters. The context and tubes of many boletes change color when exposed to the air or damaged. The characteristic blue discoloration, or bluing, seen in a number of species may result from the production of pigments and other metabolites.

The Boletaceae were considered to be absolutely homogenous in the production of pigments and other metabolites, which basically consist of pulvinic acids and derivatives. Members of the genera *Boletus*, *Boletellus*, *Pulveroboletus* and *Xerocomus* normally produce pulvinic acid derivatives like variegatic acid and xerocomic acid (Bresinsky and Besl, 1978; Gill and Steglich, 1987; Binder and Bresinsky, 2002). Exceptions are the genera *Austroboletus*, *Gyroporus*, *Tylopilus* and *Xanthoconium* in which positive proof for pulvinic acid derivatives is still lacking (Bresinsky and Besl, 1978). Recent study, retipolides form the major pigments in presently analysed of new genus *Retiboletus*. This group of secondary metabolites is unique in the Boletales and this new genus (Binder and Bresinsky, 2002).

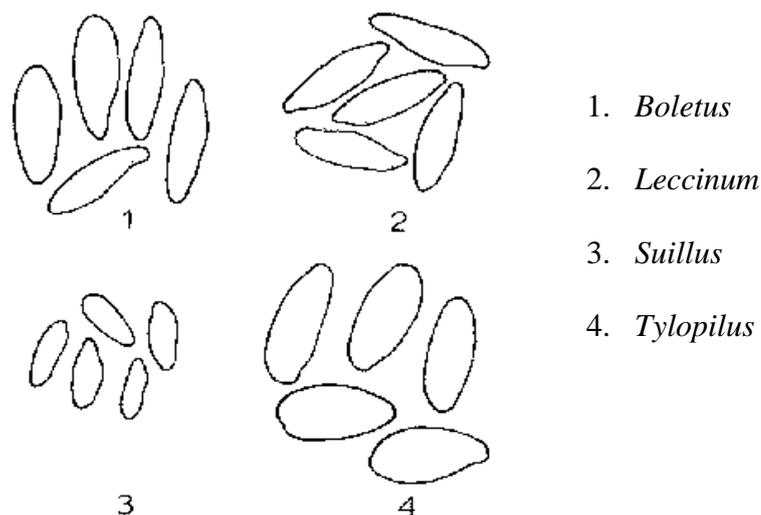
#### 2.5.1.5 Smell and taste

The taste and odor of the context of most boletes are mild and inoffensive. There are species with peppery or acrid taste. *Boletus calopus*, *B. rubripes*, and possibly a few others have a noticeably bitter taste, and some, such as *Suillus pungens* and *S. acerbus*, have somewhat harsh and unpleasant tastes (Thiers, 1975).

## 2.5.2 Micromorphological characters

### 2.5.2.1 Basidiospores

The basidiospores are of diagnostic value in the taxonomy and systematics of the boletes. Prints of the spores in mass are of significance, and spore prints are often necessary. The spore print varies in color from a shade of brown to yellow to flesh or dark pink. Generally, the spores are elongate and cylindric to fusoid or ellipsoid in face view (Figure 2.3). The spore length averages from 9 – 15  $\mu\text{m}$  and the width from 4 – 5  $\mu\text{m}$ . "Giant" spores are sometimes seen in mounts from many different species and are presumably produced by one- or two-spored basidia. The bolete spores show little or no change in color when mounted in water, dilute solutions of potassium hydroxide. However, when mounted in Melzer's reagent, they may either remain unchanged or color changes may occur. The change to rusty brown, bright rust red or a tawny color of the walls is known as dextrinoid reaction. Several species have been found in which the spore wall gives a typical blue-black, or amyloid, reaction when mounted in Melzer's reagent. The chemical reaction responsible for this color change is not fully understood, especially since rarely do all the spores show the same reaction (Thiers, 1975).



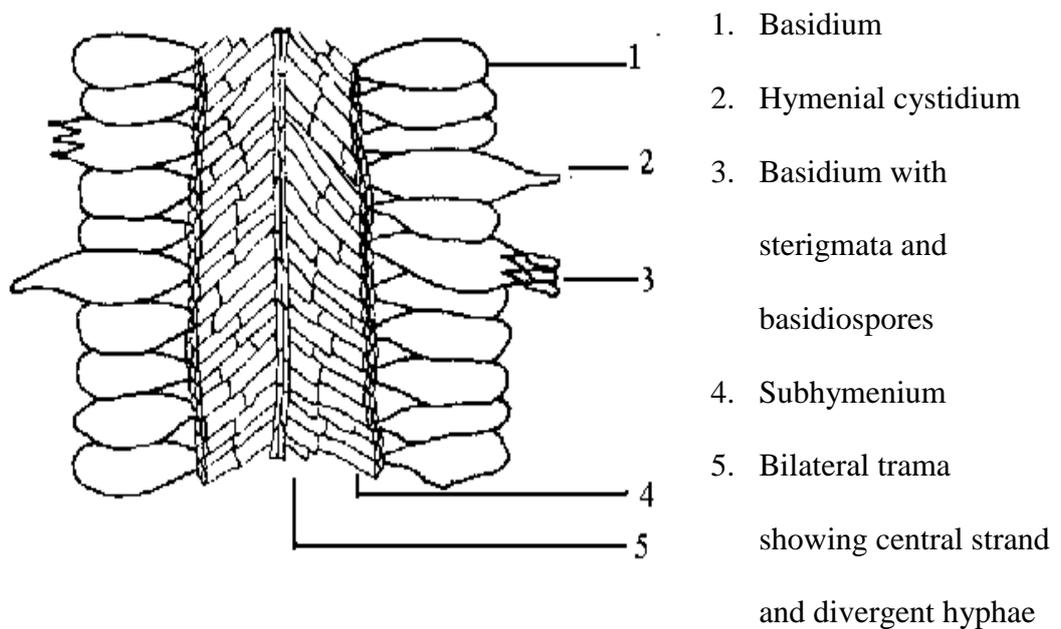
**Figure 2.3:** Bolete basidiospores. (Thiers, 1975)

### 2.5.2.2 Hymenium

The hymenium is the palisade or layer of basidia and associated cells that forms the inner lining of the tubes and may extend down the stipe if the surface is reticulate (Figure 2.4). Basidia are the most important cell, which develop as terminal cells of the hyphae and spore formation occur on the basidia. The basidia are rather large, club-shaped cells ranging from 20 – 30  $\mu\text{m}$  in length and from 7 – 12  $\mu\text{m}$  in width. At the apex four sterigmata will form, and eventually each will bear a single basidiospore.

Basidioles are also commonly seen in the hymenium. These may be undeveloped basidia or basidioid cells that never produce basidiospores. They appear to have the same origin as the fertile basidia, but their true function is not understood.

A third type of cell commonly found in the hymenium of most boletes is the cystidium (Figure 2.4). These cells are sterile and arise as differentiated hyphal tips. They are often highly distinctive in size and shape. Most often they are clavate to fusoid or ventricose in shape with an elongated to obtuse or mucronate apex. They are commonly thin-walled, but may be thick-walled in some species including *Phylloporus*. The cystidia are typically hyaline or only weakly pigmented when mounted in water. There is often a marked color change to reddish brown or black in potassium hydroxide or in Melzer's reagent. Therefore, they will be referred to as hymenial cystidia to distinguish them from the cystidia that sometimes occur on the stipe or pileus cuticle. The role of the cystidia is uncertain, but they may act as organs of excretion, serve as air traps, or preventing evaporation of moisture from the hymenial surface (Thiers, 1975).

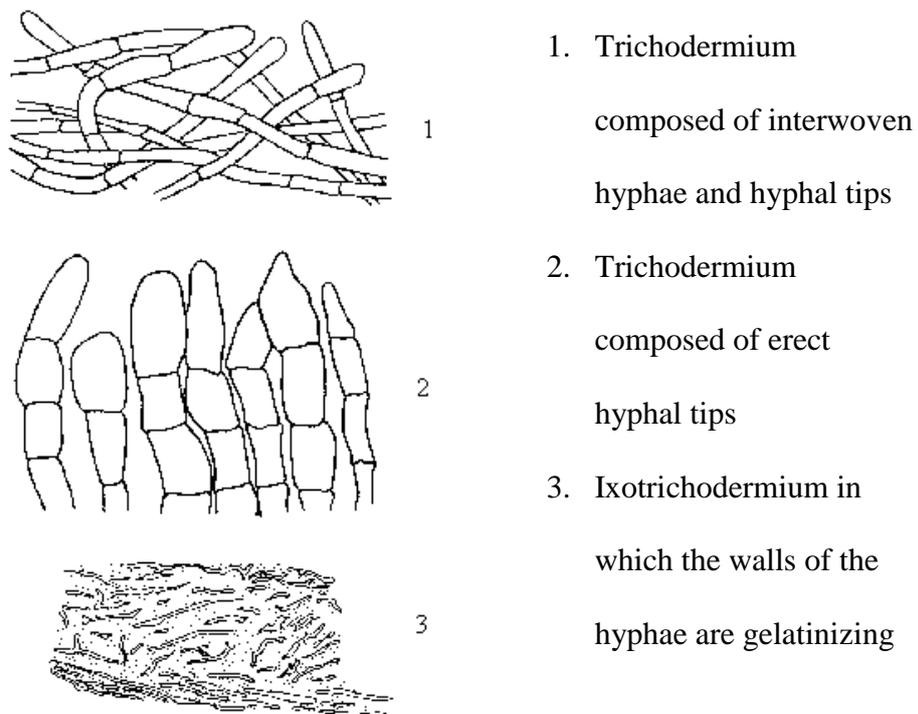


**Figure 2.4:** Longitudinal section through the bolete hymenophore (Thiers, 1975).

### 2.5.2.3 Structure of pileipellis

One of the most important and useful anatomical features from a taxonomic point of view in the boletes is the microscopic structure of the cuticular or external layer of the pileus, and to a lesser extent of the stipe. The cuticle is usually easily distinguished from the pileus trama (Figure 2.5). More characteristic of the boletes is the trichodermial type of cuticle, which is composed of interwoven hyphae. However, there is considerable variation in the trichodermial cuticle, and noticeable changes may occur when the basidiocarp matures. If at all possible, young specimens should be checked, for the hyphae of a tangled or interwoven trichodermium may collapse with age in some boletes and give the appearance of a cutis. In others the hyphal tips may remain more or less erect and highly differentiated, forming a hymeniform cuticle. If the hyphal tips become clustered or aggregated, the surface of the cap appears tomentose or scaly. Another variation in the trichodermium occurs in many species of *Suillus* is ixotrichodermium where the walls of the hyphae gelatinize.

The chemical reaction of some of the boletes in Melzer's reagent is not understood and neither is its significance in establishing species relationships within the genus. However, since the reaction is constant for a given taxon, it is used as a taxonomic character. Pileocystidia or sterile, highly differentiated cells somewhat similar to the cystidia in the hymenium are very rarely present in the cuticle of the boletes (Thiers, 1975).



**Figure 2.5:** Type of pileipellis. (Thiers, 1975)

#### 2.5.2.4 The structure of stipitipellis

The cuticle of the stipe shows less variation than the pileus cuticle (Figure 2.5). Most commonly, the surface is differentiated as a cutis, but trichodermial or ixotrichodermial types also occur. Caulocystidia on stipe is similar to the cystidia in the hymenium and give similar color changes when mounted in potassium hydroxide (Thiers, 1975).

### 2.5.2.5 Clamp connections

Clamp connections are short, inconspicuous branches located at the cross walls or septa of the hyphae and form a by-pass around the septations (Largent *et al.*, 1977, Thiers, 1975). They are present in some of the boletes and are most readily found in the cuticular hyphae, at the base of the basidium or, less frequently, in the basal tomentum of the stipe. The significance of clamp connections is not fully understood since they are often not present at every septation.

## 2.6 Taxonomic discrepancies

### 2.6.1 Family level [Boletaceae Chevall., *Fl. gén. env. Paris (Paris): 248 (1826)*]

The classification of the boletes "officially" begins with the publication of *Systema Mycologicum* by Elias Fries in 1822. In that publication he placed all species of fleshy pore fungi in the single genus *Boletus*. Soon thereafter various systems for the division of this large taxon into smaller units were proposed, beginning with the appearance of S. F. Gray's *Natural Arrangement of British Plants* (1821). He placed all the boletes in *Suillus*, *Leccinum*, or *Pinuzza*. Since the time of these two historic works, other arrangements or systems for the classification of boletes have been presented. Among these was the contribution by Quélet (1888), in which he erected several additional genera. Several years later, Gilbert (1931) in *Les Bolets* recognized eleven different genera (Thiers, 1975; Smith *et al.*, 1971).

The most elaborate classification system of the boletes is by Singer in 1962. He recognized two families, the Boletaceae and Strobilomycetaceae, and a total of eighteen genera. This system was based on the worldwide bolete flora and has been rather widely adopted, especially in European countries. In 1971, Smith and Thiers, in *The Boletes of Michigan*, followed a system which differed from Singer's in that all species were placed

in the family Boletaceae and only eleven genera were recognized. The reduction in the number of genera resulted, in part, from the combining of *Suillus* and *Boletinus*, the elimination of other genera, such as *Xerocomus*, *Xanthoconium*, *Porphyrellus*, and *Paragyrodon*, and the transfer of the genus *Phylloporus*, a lamellate genus often classified with the boletes, to the family Paxillaceae, as has been recommended by Watling. Furthermore, because of the obvious affinities of species of *Gastroboletus* to the boletes, this genus was placed in the Boletaceae (Thiers, 1975).

### 2.6.2 Genus level

Even after recent changes in taxonomy that have moved many members out of the Boletaceae, it remains a large family with many genera. The classification of genera in Boletaceae may differ according to different mycologists. Currently, 35 genera are recognized in Boletaceae, which collectively contain 787 species in 10 edition of Dictionary of Fungi, Kirk *et al.* (2008). Halling *et al.* (2007), suggested 31 genera as show below (Table 2.1).

**Table 2.1** Genera of Boletes s.s Genera reported from Malaysia are indicated in bold.

(Source: Halling *et al.*, 2007)

<i>Afroboletus</i>	<i>Phlebopus</i>
<i>Aureoboletus</i>	<i>Phylloboletellus</i>
<i>Austroboletus</i>	<i>Phylloporus</i>
<i>Boletellus</i>	<i>Porphyrellus</i>
<i>Boletochaete</i>	<i>Pseudoboletus</i>
<i>Boletinellus</i>	<i>Pulveroboletus</i>
<i>Boletus</i> (including <i>Xerocomus</i> )	<i>Retiboletus</i>
<i>Bothia</i>	<i>Rubinoboletus</i>

<i>Chalciporus</i>	<i>Sinoboletus</i>
<i>Fistulinella</i> (including <i>Mucilopilus</i> )	<i>Strobilomyces</i>
<i>Gyrodon</i>	<i>Suillus</i>
<i>Gyroporus</i>	<i>Tuboseta</i> (including <i>Setogyroporus</i> )
<i>Heimioporus</i>	<i>Tylopilus</i>
<i>Leccinellum</i>	<i>Veloporphyrellus</i>
<i>Leccinum</i>	<i>Xanthoconium</i>
<i>Paragyrodon</i>	

- 1) *Afroboletus* Pegler, D.N.; Young, T.W.K., 1981, Transactions of the British Mycological Society 76(1): 130.

Type taxon: *Afroboletus pterosporus* (Singer) Pegler & T.W.K. Young 1981

- 2) *Aureoboletus* Pouzar, Z., 1957, Česká Mykologie 11(1): 48.

Type taxon: *Aureoboletus gentilis* (Quél.) Pouzar, Z., 1957, Česká Mykologie 11(1): 48.

- 3) *Austroboletus* (Corner) Wolfe, C.B. Jr., 1979, Bibliotheca Mycologica 69: 64.

Type taxon: *Austroboletus dictyotus* (Boedijn) Wolfe 1980.

- 4) *Boletinellus* Murrill, W.A., 1909, Mycologia 1(1): 7.

Type taxon: *Boletinellus merulioides* (Schwein.) Murrill 1909.

- 5) *Boletellus* Murrill, Mycologia 1: 9. 1909.

Type taxon: *Boletus ananas* M.A. Curtis 1848.

- 6) *Boletochaete* Singer, R., 1944, Mycologia 36: 358.

Type taxon: *Boletochaete spinifera* (Pat. & C.F. Baker) Singer 1944.

- 7) *Boletus* Gray, S.F., 1821, A natural arrangement of British plants 1: 640.

Type taxon: *Boletus luteus* Linnaeus, C., 1753, Species Plantarum: 1177.

- 8) ***Bothia*** Halling, R.E.; Baroni, T.J.; Binder, M., 2007, *Mycologia* 99(2): 311.  
Type taxon: *Bothia castanella* (Peck) Halling, T.J. Baroni & Manfr. Binder  
2007.
- 9) ***Chalciporus*** Bataille, 1908, *Bull. Soc. Hist. nat. Doubs* 15:39.  
Type taxon: *Chalciporus piperatus* (Bull.) Bataille 1908.
- 10) ***Fistulinella*** Hennings, P., 1901, *Botanische Jahrbücher für Systematik,  
Pflanzengeschichte und Pflanzengeographie* 30: 43.  
Type taxon: *Fistulinella staudtii* Henn. 1901.
- 11) ***Gyrodon*** Opatowski, 1836, *Vergleichende Morphologie und Biologie der Pilze,  
Mycetozoen und Bacterien* 2(1):5.  
Type taxon: *Gyrodon sistotremoides* Opat. 1836.
- 12) ***Gyroporus*** Quélet, L., 1886, *Enchiridion Fungorum in Europa media et praesertim  
in Gallia Vigentium*: 161.  
Type taxon: *Gyroporus cyanescens* (Bull.) Quélet. 1886
- 13) ***Heimioporus*** E. Horak, *Sydowia* 56: 237. 2005. nom. nov. for: *Heimiella* Boedijn  
(1951) non Lohmann (1913).  
Type taxon: unknown
- 14) ***Leccinellum*** Bresinsky, A.; Besl, H., 2003, *Regensburger Mykologische Schriften*  
11: 231.  
Type taxon: *Leccinellum nigrescens* (Richon & Roze) Bresinsky & Manfr.  
Binder 2003.
- 15) ***Leccinum*** Gray, S.F., 1821, *A natural arrangement of British plants* 1: 646.  
Type taxon: *Leccinum aurantiacum* (Bull.) Gray 1821.
- 16) ***Paragyrodon*** (Singer) Singer, R., 1942, *Annales Mycologici* 40: 25.  
Type taxon: *Paragyrodon sphaerosporus* (Peck) Singer 1942.

- 17) *Phlebopus* (R. Heim) Singer, R., 1936, *Annales Mycologici* 34: 326.  
Type taxon: *Phlebopus colossus* (R. Heim) Singer 1936.
- 18) *Phylloboletellus* Singer, R.; Digilio, A.P.L., 1951, *Lilloa* 25: 438.  
Type taxon: *Phylloboletellus chloephorus* Singer 1952.
- 19) *Phylloporus* Quélet, *Fl. Mycol. France (Paris)* 409. 1888.  
Type taxon: *Phylloporus pelletieri* (Lév.) Quél.
- 20) *Porphyrellus* E.-J. Gilbert, 1931, *Les Livres du Mycologue Tome I-IV, Tom. III: Les Bolets*: 99.  
Type taxon: *Porphyrellus porphyrosporus* (Fr. & Hök) E.-J. Gilbert 1931.
- 21) *Pseudoboletus* Šutara, J., 1991, *Ceská Mykologie* 45(1-2): 2  
Type taxon: *Pseudoboletus parasiticus* (Bull.) Šutara 1991.
- 22) *Pulveroboletus* Murrill, W.A., 1909, *Mycologia* 1(1): 9  
Type taxon: *Pulveroboletus ravenelii* (Berk. & M.A. Curtis) Murrill 1909.
- 23) *Retiboletus* Binder, M.; Bresinsky, A., 2002, *Feddes Repertorium Specierum Novarum Regni Vegetabilis* 113(1-2): 36  
Type taxon: *Retiboletus ornatipes* (Peck) Manfr. Binder & Bresinsky 2002.
- 24) *Rubinoboletus* Pilát, A.; Dermek, A., 1969, *Ceská Mykologie* 23(2): 81  
Type taxon: *Rubinoboletus rubinus* (W.G. Sm.) Pilát & Dermek 1969.
- 25) *Sinoboletus* Zang, M., 1992, *Mycotaxon* 45: 223  
Type taxon: *Sinoboletus duplicatoporus* M. Zang 1992.
- 26) *Strobilomyces* Berkeley, M.J., 1851, *Journal of Botany (Hooker)* 3: 78  
Type taxon: *Strobilomyces strobilaceus* (Scop.) Berk. 1851.
- 27) *Suillus* P. A. Micheli 1729, *Nova Plantarum Genera*: 126; ex S.F. Gray, 1821, *A Natural Arrangement of British Plants* 1: 646  
Type taxon: *Suillus luteus* (L.) Roussel 1821.

28) *Tuboseta* Horak

Type taxon: unknown.

29) *Tylopilus* Karsten, P.A., 1881, Revue mycologique, Toulouse 3(9): 16

Type taxon: *Tylopilus felleus* (Bull.) P. Karst. 1881.

30) *Veloporphyrillus* Singer, R.; Gómez, L.D., 1984, Brenesia 22: 293

Type taxon: *Veloporphyrillus pantoleucus* L.D. Gómez & Singer 1984.

31) *Xanthoconium* Singer, R., 1944, Mycologia 36: 361.

Type taxon: *Xanthoconium stramineum* (Murrill) Singer 1944.

## 2.7 Edibility and economic importance

Many boletes are well known for their quality as edible fungi. *Boletus edulis* is one of the best of all mushrooms and is excellent when used either fresh or dried. In the Finnish cuisine, *Boletus edulis* known as the King Bolete is universally considered to be the tastiest culinary mushroom. Peppery bolete (*Chalciporus piperatus*) has an extremely strong taste, and has been used in place of pepper (Carluccio, 2003). Other boletes considered to be top quality are *Boletus zelleri*, *B. mirabilis*, *Leccinum aurantiacum*, *L. manzanitae*, and *L. insigne*. Unfortunately, these species are common in Europe and America but not in the tropics. Consumption of boletes in Malaysia is not common. This is mostly due to the poor knowledge of boletes in Malaysia and the production of boletes in tropical forest is few.

Most of the boletes are not poisonous but some are mildly toxic. For example *Boletus eastwoodiae*, is known to cause stomach disorders. This species is not a tropical species and might not found in this part of the world. In some parts of the world, they believe eating any bolete in which the context turns blue when exposed to air are poisonous. However, some species like *Xerocomus badius* with this characteristic are not

poisonous. It is strongly advised that anyone interested in eating unfamiliar boletes do so in small amounts at first so that any unfavorable reactions may be detected.

## 2.8 Medicinal value

Not many medicinal uses have been reported in boletes because of the difficulty of cultivation of many species. Most of the medical properties can only be tested from extract of the fruiting body. Therefore a large number of clean and fresh collections are usually needed to extract enough bioactive compounds for testing. This quantity is problematic for collections from tropical forests single or limited fruiting bodies are reported in the wild. Study on *Boletus badius* (known as *Xerocomus badius*) showed that it contained Theanine (N-ethyl- $\gamma$ -glutamine or  $\gamma$ -glutamylethylamide), a non-protein forming amino acid that occurs in the bay boletes (Casimir *et al.*, 1960). A number of studies have recently investigated the physiological and pharmacological potential of theanine, associated most commonly with tea leaves (*Camellia sinensis*). These studies suggest that theanine may be useful for promoting relaxation (Lu *et al.*, 2004), reducing blood pressure (Yokogoshi *et al.*, 1995), inhibiting the negative effects of caffeine (Kakuda *et al.*, 2000), enhancing anti-tumor activity (Sadzuka *et al.*, 2000), providing neuroprotective effects (Egashira *et al.*, 2004) and providing anti-obesity effects (Zheng *et al.*, 2004).

A recent study had demonstrated a promising method for producing theanine by using submerged fermentation of *Boletus badius* mycelium and response surface methodology (Li *et al.*, 2008). Also, there are studies on antioxidant activity by *Boletus badius*. The methanolic extract of dried *B. badius* was analyzed for antioxidant activity in different systems including reducing power, free radical scavenging, superoxide anion radical scavenging, total antioxidant activity, and metal chelating activities (Elmastas *et*

*al.*, 2006). In addition, studies showed that *B. badius* consist of antitumor effects (Ohtsuka *et al.*, 1973) and neuronal inhibition (Moldovan *et al.*, 2001).

### 3.0 MATERIALS AND METHODS

#### 3.1 Basidiomes collection

Specimens of boletes were collected from their natural habitat from thirteen different forests in Peninsular Malaysia (Table 3.1). The forests can be separated into two different types, lowland rain forests and highland forests. Lowland rain forests have elevation from 28m to 640m while highland forests have elevation of more than 700m. (Plate 3.1).

**Table 3.1:** Sampling sites for the bioersivity of Boletaceae.

No	Area	Location
	Tropical Lowland Rainforests	
1	Pasoh Forest Reserve, Negeri Sembilan.	N 02°58'07.6" E 102°17'49.2"
2	Ulu Bendul Forest Reserve, Negeri Sembilan.	N 02°43'39.3" E 102°04'38.2"
3	Sungai Chongkak Forest Reserve, Hulu Langat, Selangor.	N 03°12'00.3" E 101°50'07.4"
4	Batang Kali forest area, Selangor.	N 03°26'54.4" E 101°41'12.65"
5	Ulu Gombak forest area, Selangor.	N 03°15'01.4" E 101°44'59.9"
6	Endau Rompin National Park, Johor (Peta).	N 02° 32' 30.6" E 103° 22' 08.9"
7	Endau Rompin National Park, Johor (Selai).	N 02°25'59.4" E 103°14'49.5"
8	Balik Pulau, Penang.	N 05°21'12.9" E 100°11'59.1"
9	Kuala Lipis, Pahang.	N 04° 15' 31.7" E 101°32' 52.9"
10	Kampung Buku, Gunung Raya, Pulau Langkawi, Kedah.	N 06°22'42.3" E 99°49'21.5"
	<b>Highland Forests</b>	
11	Fraser's Hill, Pahang	N 03°43'36.0" E 101°43'00.6"
12	Sungai Palas, Cameron Highland, Pahang	N 04°31'47.0" E 101°23'46.2"
13	Bukit Tinggi, Pahang	N 03°23'41.4" E 101°50'35.6"

When collecting a specimen, several stages of development such as the young as well as mature individuals were necessary for identification (Halling, 2005). Once collected, the mushrooms were handled carefully. The specimens were placed into a covered box or wrapped with aluminum foil or waxed paper to prevent the specimens from drying up. The wrapped mushrooms were placed in a sturdy basket, box, or bag, and carried to the laboratory or working site.



**Figure 3.1:** Location of the selected areas for sample collecting in Peninsular Malaysia

(Source: <http://malaysia.tourism-asia.net/map-of-malaysia.html>).



**Plate 3.1:** Selected collecting sites. Lowland rain forests: **a.** Endau Rompin National Park, Johor (Peta). **b.** Pasoh Forest Reserve, Negeri Sembilan; Highland forests: **c.** Fraser's Hill, Pahang. **d.** Sungai Palas, Cameron Highland, Pahang.

### **3.2 Spore print**

Spore prints were prepared immediately from the specimens collected from field. A mature fruiting body was selected to do the spore prints because too old or young fruiting bodies do not give a spore print. Before preparing the spore print, the stalks of mushrooms were cut off and the cap placed on a white paper. The cap was covered with a container to prevent moisture loss. After several hours, the colour of the spore prints were observed and recorded.

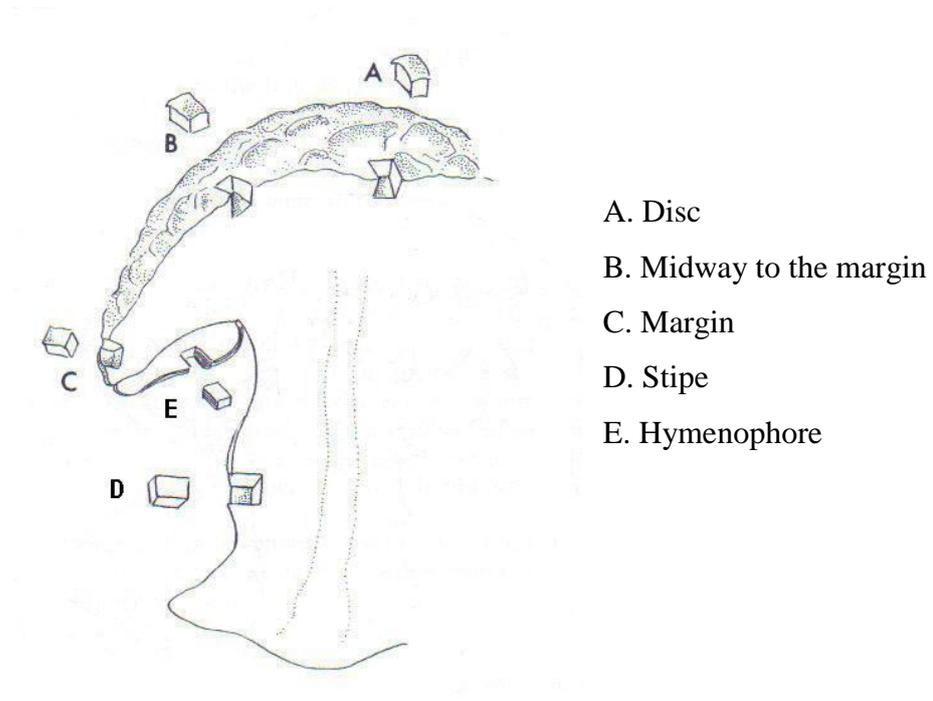
### **3.3 Morphological description**

The macromorphology of specimens were recorded before the preparation of herbarium specimens. Descriptions of the characteristics of fresh specimens are important because the features disappear after the specimens are dried. The descriptions include color, pileus shape, surface features, hymenophore features, size and the odor or taste if present (Largent, 1986). The colour change after the context was exposed to air or staining with ammonium hydroxide ( $\text{NH}_4\text{OH}$ ) was important to note. A 16X hand lens was aid the observation of macromorphological features. Color terms and notations in parentheses were from Kornerup and Wanscher (1978).

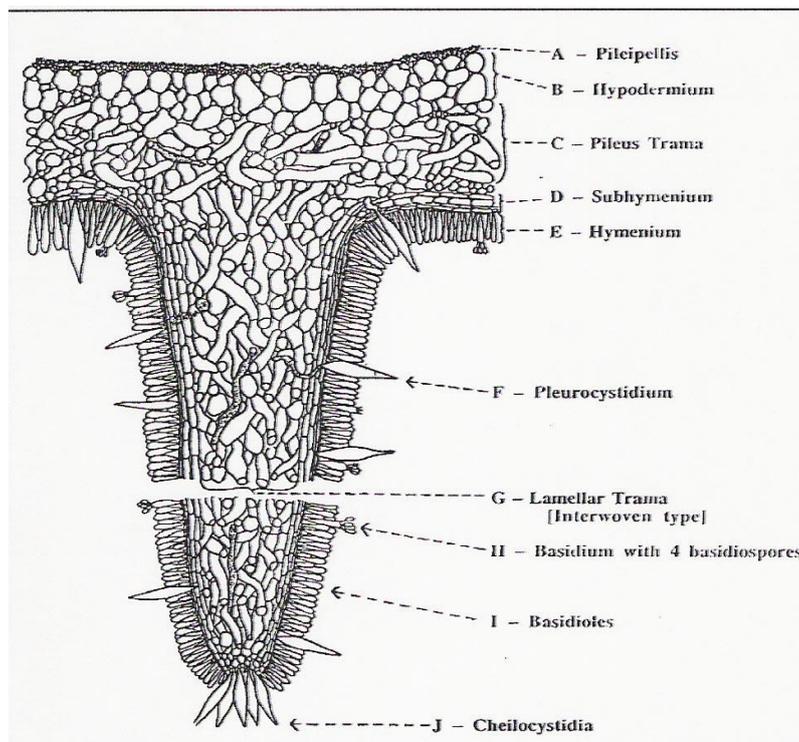
Of the microscopic features of boletes, the basidiospores are of considerable diagnostic value in the taxonomy and systematics of the group. Small segments were cut from the dried specimen (Figure 3.2). Spores, hymenophoral, pileus and stipe elements were mounted in 3% KOH, Melzer's reagent and 10% of  $\text{NH}_4\text{OH}$  to observe the basidiospores, basidia, cystidia, pileipellis and stipe tissue (Largent *et al.*, 1977) (Figure 3.3). Then, the segment was observed using a Nikon Alphaphot-2 microscope with

drawing tube. The microscopic structures and characteristics were recorded and illustrated.

Spore statistics include:  $x_m$ , the arithmetic mean of the spore length by spore width ( $\pm$  standard deviation) for n spores measured in a single specimen;  $x_{mr}$ , the range of spore means, and  $x_{mm}$ , the mean of spore means ( $\pm$  SD) where more than one specimen is available; Q, the quotient of spore length by spore width in any one spore, indicated as a range of variation in n spores measured;  $Q_m$ , the mean of Q-values in a single specimen ( $\pm$  SD);  $Q_{mr}$ , the range of  $Q_m$  values and  $Q_{mm}$ , the mean of  $Q_m$  values where more than one specimen is available ( $\pm$  SD) (Tan, 2007).



**Figure 3.2:** Blocks of tissue taken for micromorphological examination. (modified from Largent *et al.*, 1977).



**Figure 3.3:** Cross section of lamellae for micromorphological examination. (modified from Largent *et al.*, 1977).

### 3.4 Preparation of herbarium specimens

After recording the characteristics and photography, the specimens were placed in an electrical oven and dried slowly at a temperature of 55-65 °C overnight. The specimens were then prepared as herbarium specimens for further examination of micromorphological features and DNA studies. The herbarium specimens were labelled and packed according to the KLU code, location and date. The herbarium specimens were stored in an incubator at room temperature.

Dried collections were accessioned into the University of Malaya Herbarium (KLU) at University of Malaya, Kuala Lumpur.

## CHAPTER 4 Results and Discussion

### 4.1 Diversity of Boletaceae

Collections of Boletineae in Peninsular Malaysia were carried out from 2003 to 2007. A total of 52 collections of boletes were identified to 10 genera and 30 different species. The species included the following genera: *Afroboletus*, *Aureoboletus*, *Austroboletus*, *Boletellus*, *Boletus* (including *Xerocomus*), *Phylloporus*, *Pulveroboletus*, *Strobilomyces*, *Tylopilus* and *Xanthoconium*. Four species are documented as new to science (*Afroboletus malaysianus*, *Phylloporus megaporinus*, *Pulveroboletus claroflavus* and *Tylopilus favo-ballouii*), while three species are documented as new records in Malaysia (*Tylopilus plumbeoviolaceus*, *Tylopilus rubrobrunneus* group and *Xanthoconium violaceofuscus* comb. prov.). Twenty-two species reported by other researchers were re-collected and one specimen (*Phylloporus* sp. 1) has not been identified to species level due to the single specimen and similarity of the species with taxon from Australia which remains as *Phylloporus* sp. 1. (Figure 4.6a, 4.6b).

The result shows that the most suitable collecting of boletes in Peninsular Malaysia is from March to April and August to September. This is due to the raining season and the fruiting season for basidiomycetes. Most of the boletes were collected from lowland dipterocarp forests from West Peninsular Malaysia. Some of the species were found in highland forests such as new species *Phylloporus megaporinus*. Species such as *Tylopilus ballouii*, *Tylopilus albo-ater*, *Tylopilus nigropurpureus* and *Pulveroboletus ridleyi* can be found in both lowland and highland forests. *Afroboletus malaysianus* and *Boletellus corneri* were only collected from Pulau Langkawi which at the northern of Peninsular Malaysia. This isolated island shows a good diverse of boletes. Further collecting and study should be carried out to document the diversity of boletes in this island.

#### 4.1.1 Key to Boletaceae of Peninsular Malaysia

- 1a. Hymenium lamellate to subporoid, decurrent on stipe . . . . . 2
- 1b. Hymenium tubulose, usually not decurrent on stipe . . . . . 8
- 2a. Mycelium at the base of stipe yellowish white; pileus brown . . . . . 3
- 2b. Mycelium at the base of stipe white; pileus dark brown or reddish  
brown . . . . . 1. *Phylloporus bogoriensis* [p. 37]
- 3a. Context and tubes/pores brown or blue when bruised. . . . . 4
- 3b. Context and tubes/pores no colour changed when bruised . . . . . 6
- 4a. Pores alveolate . . . . . 2. *Phylloporus megaporinus* [p. 40]
- 4b. Pores lamellate . . . . . 5
- 5a. Pileus squamulose; NH<sub>4</sub>OH stains red on  
context . . . . . 3. *Phylloporus brunneolus* [p.43]
- 5b. Pileus tomentose; NH<sub>4</sub>OH negative on context . . . . .  
. . . . . 4. *Phylloporus cf. orientalis* [p. 46]
- 6a. Context white . . . . . 5. *Phylloporus cf. sulcatus* [p.49]
- 6b. Context yellowish white . . . . . 7
- 7a. Pileus large, >70 mm diam., light brown; NH<sub>4</sub>OH stains red on  
context . . . . . 6. *Phylloporus sp. 1* [p.51]

- 7b. Pileus small, <20mm diam., grayish brown; NH<sub>4</sub>OH not stains on  
context ..... 7. *Pyloporus cf. bellus* [p. 55]
- 8a. Pileus and stipe with dry universal veil; forming a peronate annulus or annular  
zone on stipe, often with sulfur yellow colors, staining blue when bruised or  
colour did not change; basidiospores smooth; base of stipe white or  
yellow ..... 9
- 8b. Pileus and stipe lacking dry universal veil; if veil is present, then basidiospores  
ornamented; base of stipe white ..... 12
- 9a. Pileus and stipe viscid; hymenium turning brown; basidiospores 11 – 14 × 6 – 7  
µm, ellipsoid or boletoid ..... 8. *Pulveroboletus cf. viridis* [p.59]
- 9b. Pileus and stipe dry, pulverulent or scaly-squamulose. .... 10
- 10a. Pileus olive, scaly or squamulose; context szans blue when exposure;  
basiodiospores small, 7 – 9 × 5 – 6 µm. ....  
..... 9. *Pulveroboletus viridisquamosus* [p.61]
- 10b. Pileus pulverulent from yellowish veil; context stains blue when exposure or  
colour did not change; basidiospores larger ..... 11
- 11a. Pores <1 mm diam.; context changing blue when bruised; stipe solid; mycelium  
at base of stipe white ..... 10. *Pulveroboletus ridleyi* [p.63]
- 11b. Pores >1 mm diam.; context not staining when bruised; stipe hollow; mycelium  
at base of stipe yellow ..... 11. *Pulveroboletus claroflavus* [p.68]

- 12a. Pores/tubes white, yellow or red; spore print bright yellow-brown, olive, olive-brown, brown or dark brown . . . . . 13
- 12b. Pores/tubes pink, vinaceous, grey-brown to very dark brown or black; spore print pinkish to reddish brown, vinaceous-brown or black . . . . . 21
- 13a. Basidiospores short ellipsoid or elongate-fusoid, with conspicuous longitudinal ridges/furrows and transversal striations . . . . . 14
- 13b. Basidiospores ovoid, ellipsoid or elongate-fusoid, smooth, longitudinal ridges/furrows and transversal striations absent . . . . . 16
- 14a. Pileus viscid; stipe subalveolate-reticulate; basidiospores 11 – 14 × 9 – 10 μm, black, dextrinoid, with longitudinal and conspicuous wings/furrows . . . . . 12. *Afroboletus malaysianus* [p.69]
- 14b. Pileus dry; stipe not reticulate; basidiospores olive-brown, inamyloid, with longitudinal ridges/furrows and transversal striations . . . . . 15
- 15a. Pileus brown, smooth or velvety; stipe surface yellow to reddish brown; basidiospores 12 – 15 × 6 – 8 μm, subfusoid to elongate fusoid, with longitudinal ridges, transversal striations absent . . . 13. *Boletellus corneri* [p.72]
- 15b. Pileus with red scales; stipe surface red, fibrillose; basidiospores 19 – 26 × 9 – 11 μm, subfusoid with longitudinal ridges/furrows, transversal striations present . . . . . 14. *Boletellus emodensis* [p.75]

- 16a. Spore print bright yellow-brown; pileus dark violet-brown, pileipellis  
hymeniform; pileus and stipe turning blue in KOH and NH<sub>4</sub>OH; basidiospores  
8 – 11 × 4 – 5 μm, ovoid or ellipsoid . . . . .  
. . . . . 15. *Xanthoconium violaceo-fuscus* [p. 78]
- 16b. Spore print olive-brown; pileus variously coloured, pileipellis usually a  
trichodermium or an ixotrichodermium; pileus and stipe not turning blue with  
KOH and NH<sub>4</sub>OH; basidiospores elongate fusoid . . . . . 17
- 17a. Pileus dry; stipe fibrillose or reticulate; context colour changed or did not  
change when bruised . . . . . 18
- 17b. Pileus viscid, rugulose; stipe glabrous; context colour did not change when  
bruised; basidiospores  
9.5 – 11 × 5 – 5.5 μm . . . . . 16. *Aureoboletus thibetanus* [p.82]
- 18a. Pileus large >30 mm diam., yellow, orange, red, brown, or grayish brown;  
hymenium yellow, when bruised colour change or  
negative . . . . . 19
- 18b. Pileus small <10 mm diam., violet-red to dark-red; hymenium yellow, blue  
when bruised; basidiospores 8 – 10 × 5 – 6 μm . . 17. *Boletus patouillardii* [p.85]
- 19a. Pileus brown to grayish brown; context and hymenium changed brown or no  
colour changed when bruised; stipe smooth, fibrillose or lacunose . . . . . 20
- 19b. Pileus orange-yellow to red; context and hymenium changed blue when bruised;  
finely reticulate at apex of stipe; basidiospores 6 – 7.5 × 4.5 – 5.5  
μm . . . . . 18. *Boletus cf. craspedius* [p.87]

- 20a. Stipe smooth or fibrillose; pileus brown to grayish brown, pileipellis hyphae inamyloid; context and hymenium turning violet or lilac-red with NH<sub>4</sub>OH; basidiospores 12 – 17 × 5 – 7 μm . . . . . 19. *Boletus phaeocephalus* [p.90]
- 20b. Stipe with conspicuous, lacunose net; pileus grayish brown, pileipellis hyphae dextrinoid; context and hymenium turning brown when bruised or with NH<sub>4</sub>OH; basidiospores 11 – 14 × 4 – 6 μm . . . . . 20. *Boletus peltatus* [p.93]
- 21a.(12b) Basidiospores ornamented . . . . . 22
- 21b. Basidiospores smooth . . . . . 23
- 22a. Pileus with black to dark gray scales; hymenium black in age, staining black when bruised, reaction with NH<sub>4</sub>OH negative; basidiospores 10 – 12 × 9 – 10 μm, globose, echinate with slender conical spines, black in KOH . . . . . 21. *Strobilomyces foveatus* [p.95]
- 22b. Pileus lacking dark colored scales; hymenium becoming pink, red or sometimes black in age, reaction with NH<sub>4</sub>OH red; basidiospores 16.5 – 18 × 6 – 6.5 μm, elongate-fusoid, with fine pit-like perforations, translucent purple in KOH . . . . . 22. *Austroboletus longipes* [p.98]
- 23a. Pileus dark fuscous or black . . . . . 24
- 23b. Pileus yellow, orange, olive green, purple or brown . . . . . 25
- 24a. Stipe with conspicuous, reticulate net; basidiospores 10 – 14 × 5 – 6 μm, ellipsoid or ovoid . . . . . 23. *Tylopilus nigropurpureus* [p.102]
- 24b. Stipe smooth or fibrillose; basidiospores 12 – 16 × 4 – 6 μm, elongate-ellipsoid or subfusoid . . . . . 24. *Tylopilus albo-ater* [p.105]

- 25a. Mycelium at base of stipe yellow; pileus olive-yellow or olive-brown,  
rugose. . . . . 26
- 25b. Mycelium at base of stipe white; pileus with other colour, smooth, velvety or  
subtomentose . . . . . 27
- 26a. Stipe yellow, pubescent; context yellow, no colour changed when bruised,  
reaction with NH<sub>4</sub>OH negative; basidiospores 9 – 11 × 4.5 – 5.5 μm, ellipsoid  
or boletoid . . . . . 25. *Tylopilus virens* [p.108]
- 26b. Stipe yellow at apex but pinkish red on lower half; context yellow, no colour  
changed when bruised, reaction with NH<sub>4</sub>OH negative; basidiospores 10 – 14 ×  
5 – 6 μm. . . . . 26. *Tylopilus pernanus* [p.111]
- 27a. Pileus orange; context and hymenium no colour changed when bruised, reaction  
with NH<sub>4</sub>OH negative; taste not distinctive; basidiospores bean shaped or  
ovoid . . . . . 28
- 27b. Pileus not orange; colour of context and hymenium changed or no changed  
when bruised or with NH<sub>4</sub>OH; taste bitter; basidiospores subfusoid. . . . . 29
- 28a. Pores round or ovoid, <1 mm diam. . . . . 27. *Tylopilus ballouii* [p.114]
- 28b. Pores favoloid, >1 mm diam. . . . . 28. *Tylopilus favo-ballouii* [p.117]
- 29a. Basidiospores 11 – 14 × 5 – 6 μm, dextrinoid; context and hymenium changing  
brown when bruised . . . . . 29. *Tylopilus rubrobrunneus* group [p.120]
- 29b. Basidiospores 9 – 11 × 3 – 4 μm, inamyloid; context and hymenium no colour  
changed when bruised. . . . . 30. *Tylopilus plumbeoviolaceus* [p.123]

#### 4.1.2 Enumeration of taxa

1. *Phylloporus bogoriensis* Höhn. Stizber. Kaiserl. Acad. Wiss. Wien Math.-naturw. Kl. 123: 89 (1914). (Fig. 4.1a, 4.2b, Plate 4.1 E)

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

*Pileus* 75 – 94 mm broad, plane with uplifted and undulating margin, dry, smooth to velvety, dark brown (6F5) at disc, light brown (5D6) at margin, turning green at first with NH<sub>4</sub>OH then slowly to red. *Context* 12 – 17mm thick at disc area, 2mm thick at margin, soft and spongy, white, turn brown when exposed, no changed with NH<sub>4</sub>OH. *Hymenophore* 6 – 7 mm thick, lamellate, decurrent with even edges, deep yellow (4A8), close with 3 – 5 lamellulae per 5 mm in 2 series, brown when exposed, no colour changed with NH<sub>4</sub>OH. *Stipe* 47 – 60 mm × 10 mm (apex), 7 mm (base), central, tapering downward and slightly flexuous, dry, fibrillose, grayish yellow to light brown (4B4), with interior solid and firm, brown when exposed, negative reaction with NH<sub>4</sub>OH, yellowish white at the base.

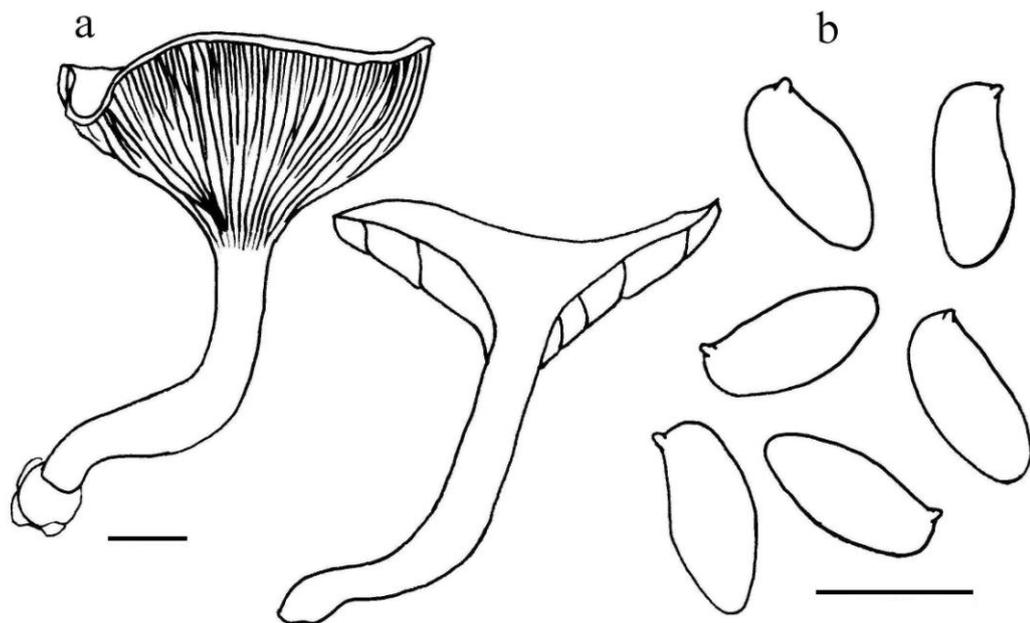
*Basidiospores* 9 – 12 × 4 – 6 μm [  $x_m = 10.57 \pm 0.86 \times 5.83 \pm 1.38 \mu\text{m}$ ,  $x_{mr} = 10.24 - 10.57 \times 4.76 - 5.83 \mu\text{m}$ ,  $x_{mm} = 10.41 \pm 0.23 \times 5.30 \pm 0.76 \mu\text{m}$ ,  $Q = 1.80 - 2.40$ ,  $Q_m = 2.11 \pm 0.16$ ,  $Q_{mr} = 2.11 - 2.25$ ,  $Q_{mm} = 2.18 \pm 0.10$ ,  $n = 55$  spores per two collections], olive brown, ellipsoid to subfusoid, smooth, subdextrinoid, hyaline in KOH. *Basidia* (25-) 32 – 40 × (9-) 10 – 12 (-17) μm, 4-sterigmate, 4 – 5 μm long, long subclavate, hyaline in KOH, inamyloid. *Pleurocystidia* 75 – 135 × 16 – 22 μm, long subventricose to cylindrical in shape. *Cheilocystidia* (48-) 52 – 78 (-92) × 15 – 19 μm, subventricose to cylindrical in shape, thin-walled, brown in KOH, slightly dextrinoid. *Pileipellis* with a long pile of hyphae more than 200 μm high, smooth, thin-walled, hyaline, inamyloid.

*Stipitipellis* forming a parallellocutis with caulocystidia, (31-) 32 – 46 (-54) × 7 – 11 (-13)  $\mu\text{m}$ , long clavate, smooth, thin-walled, slightly dextrinoid. *Clamp connections* absent.

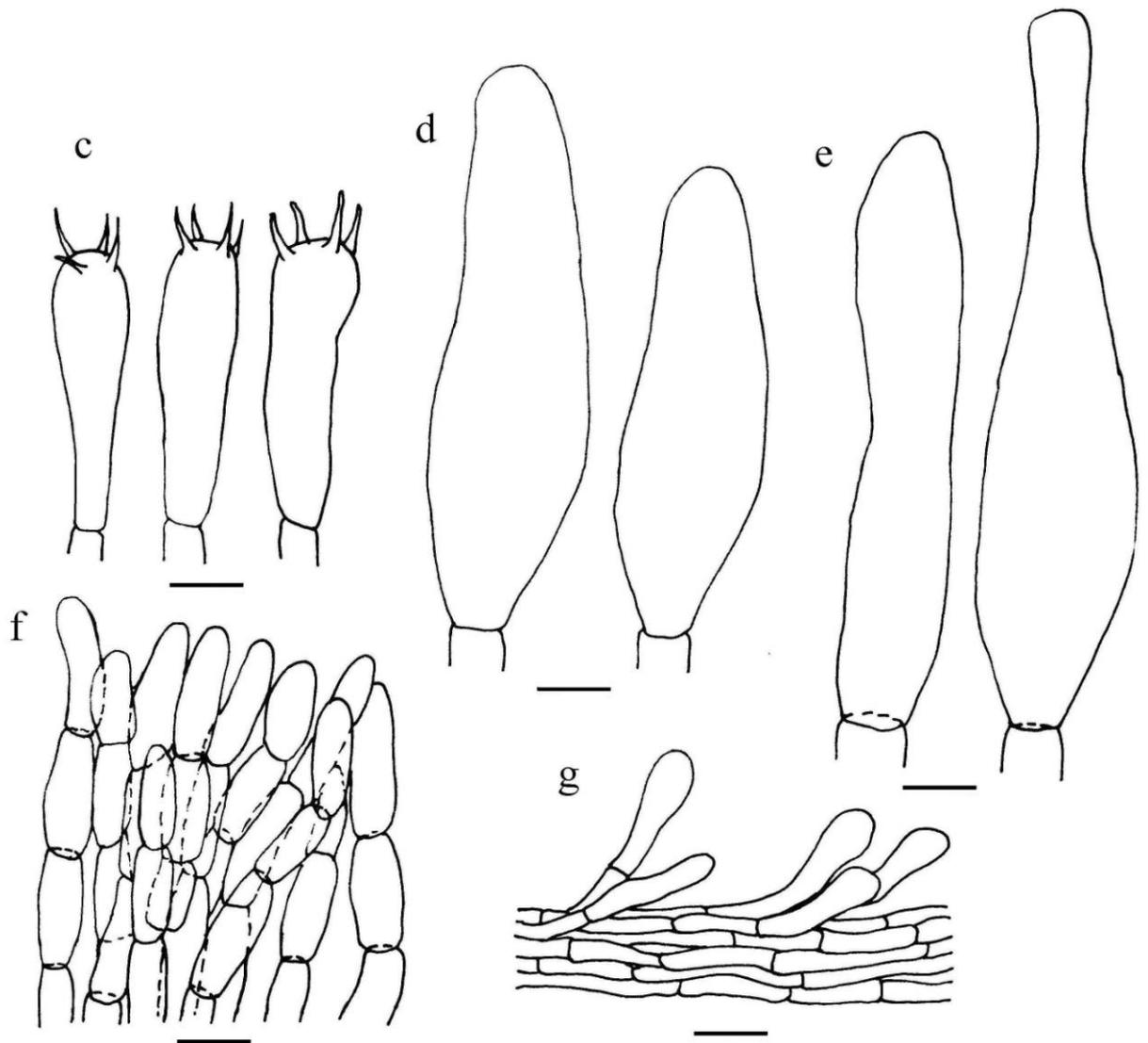
*Habit, habitat, and distribution:* gregarious and scattered on soil in forests of lowland Dipterocarp forest; reported from Peninsular Malaysia and Singapore.

*Material examined:* MALAYSIA. Negeri Sembilan, Pasoh, Hutan Simpan, 2°58'7"N, 102°17'49"E, 111 m, 20 September 2006, *leg. Chan H.T.* (KLU-M1075). Pahang, Kuala Lipis, Kampung Orang Asli, Pos Lenjang, N 04° 15' 31.7", E 101°32' 52.9", 461m, 6 September 2006, *leg. Chan H.T.* (KLU-M1076).

*Commentary:* This resembles *P. bellus* which has velvety surfaces, non-cyanescent and non- red-black colour-change, have gills rather than pores and non reticulate at the stipe. This species rather reddening to browning tissues, white mycelium at stipe base and generally uniform brown pileus appear diagnostic which fits the description from Corner's species and Hoehnal.



**Figure 4.1a :** *Phylloporus bogoriensis* Höhn. (KLU- M1075). **a.** Basidiomes **b.** Basidiospores. Bars: a = 20 mm. b = 10  $\mu\text{m}$ .



**Figure 4.1b:** *Phylloporus bogoriensis* Höhn. (KLU- M1075). **c.** Basidia. **d.** Cheilocystidia. **e.** Pleurocystidia. **f.** Pileipellis. **g.** Stipitipellis. Bars: c – e = 10  $\mu\text{m}$ . f = 25  $\mu\text{m}$ . g = 20  $\mu\text{m}$ .

2. *Phylloporus megaporinus* Chan H.T. & Halling R.E., *sp. nov. prov.*

(Fig. 4.2, Plate 4.1 C)

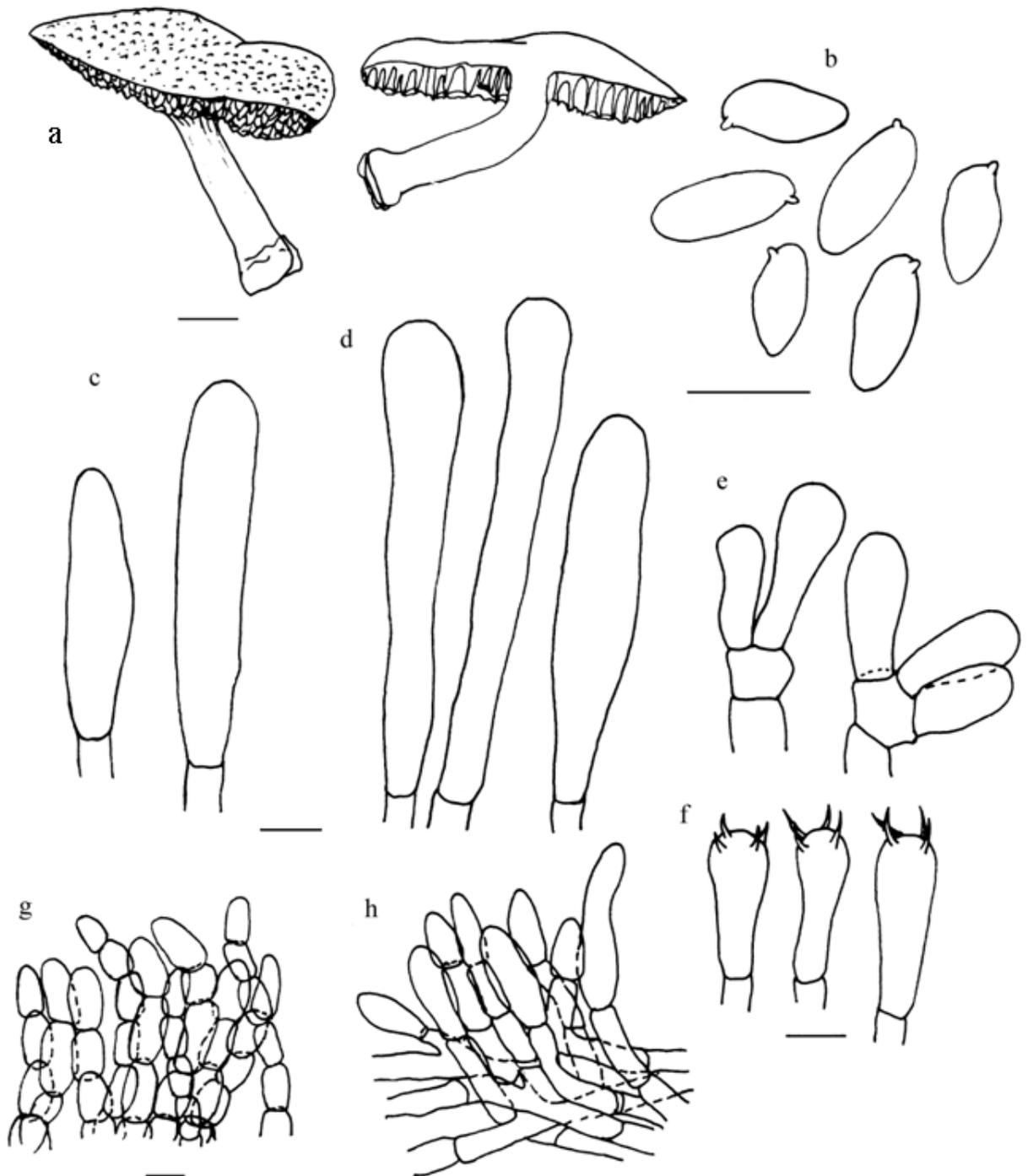
*Pileus* 26 - 57 mm broad, convex to plano-convex, dry, with margin decurved margin and striate, squamose, brownish orange (5C4), turning purplish red to black with NH<sub>4</sub>OH. *Context* 5 mm thick at disc, 2 mm thick at margin, soft and spongy, white, turning red when exposed, orange red with NH<sub>4</sub>OH. *Hymenophore* 4 - 9 mm thick; lamellae with deep anastomoses to almost tubulose, 3 pores per 5 mm, adnexed, yellow (3A7), turning blue when bruised, turning red with NH<sub>4</sub>OH. *Stipe* 35 - 45 mm long, 4 - 7 mm broad, central, equal to slightly clavate, dry, fibrillose, brown (5D4); context solid, white, red when exposed, red with NH<sub>4</sub>OH, with white basal mycelium.

*Basidiospores* (-6) 8 - 12.5 × 4 - 6 μm [ $x_m = 10.60 \pm 1.47 \times 4.98 \pm 0.44$  μm, Q = (1.5-) 1.8 - 2.5,  $Q_m = 2.161 \pm 0.25$ , n = 30 spores per one collections], ellipsoid to ovate to subfusoid, smooth, olive brown, inamyloid, hyaline in KOH. *Basidia* 25 - 32 (-37) × 7 - 10 (-11) μm, 4-sterigmate, clavate, hyaline in KOH, inamyloid. *Cheilocystidia* 75 - 89 (-91) × 12 - 13 μm, cylindrical to subclavate, hyaline in KOH, inamyloid, thin-walled. *Pleurocystidia* (46-) 60 - 70 × 11 - 12 μm, subclavate to cylindrical, hyaline in KOH, inamyloid, thin-walled. *Hymenophoral trama* parallel to subdivergent, with hyaline elements, inamyloid. *Pileipellis* a structure of moniliform hyphae, forming a pile of end cells 15 - 22 μm in broad, thin-walled, hyaline, slightly dextrinoid. *Stipitipellis* a trichodermium of scattered cylindrical to subclavate to subventricose cystidia; *caulocystidia* 15 - 50 × (5-) 13 - 17 (-19) μm, cylindrical to subclavate to subventricose, thin-walled, hyaline, inamyloid. *Clamp connections* absent.

*Habit, habitat, and distribution:* solitary to gregarious on soil in highland forests.

*Material examined:* MALAYSIA. Pahang, Fraser Hill, Jalan Air Terjun Jeriau. N 03°43'36.0", E 101°43'00.6", 680m; 1 January 2007, *leg. Chan H.T.* (KLU-M1077).

*Commentary* This is a large example of a *Phylloporus* with radially and conspicuously elongated pores. It readily stains blue on the hymenophore and the context turns red when exposed. Such combinations of features are unknown for Malaysian phyllopori. The nearest might be *P. cingulatus* described by Corner (1970).



**Figure 4.2:** *Phylloporus megaporinus* Chan H.T. & Halling R.E. *sp. nov. prov.* (KLU-M1077). **a.** Basidiomes **b.** Basidiospores. **c.** Pileocystidia. **d.** Cheilocystidia. **e.** Basidiales. **f.** Basidia. **g.** Pileipellis. **h.** Claulocystidia. Bars: a = 100 mm. b = 20  $\mu\text{m}$ . c – f & h = 10  $\mu\text{m}$ . e = 20  $\mu\text{m}$ .

3. *Phylloporus brunneolus* Corner, Nova Hedwigia 20: 802. 1970.

(Fig. 4.3, Plate 4.1 F)

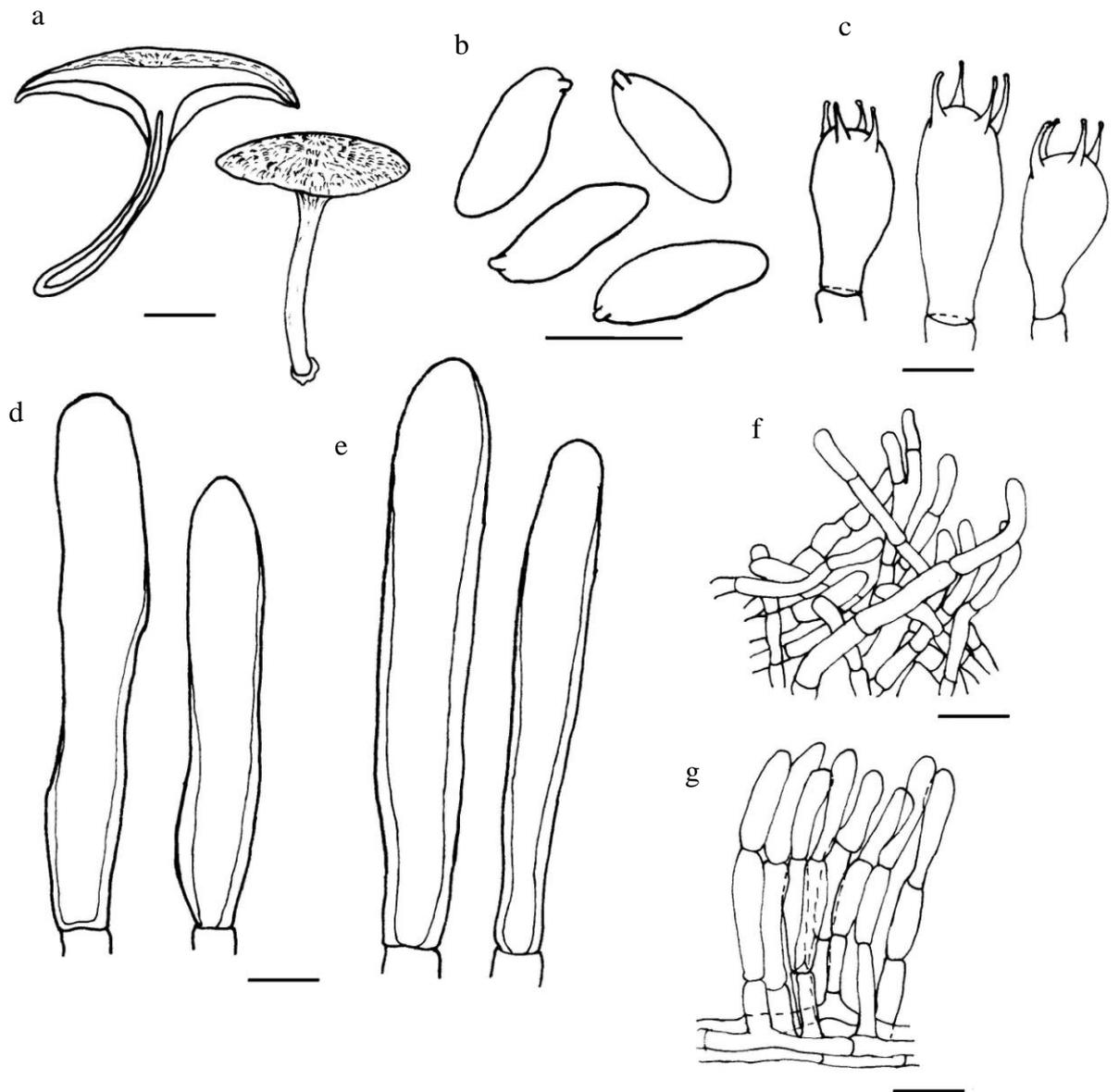
*Pileus* 10 – 50 mm broad, convex to slightly depressed on the center of disc, dry, straight to slightly decurved at the margin, becoming even and slightly wavy with age, squamulose, light brown (5D6) to brown (6E8), not bruising. *Context* 2 - 4 mm thick at disc, 1- 2mm thick at margin, soft and spongy, yellowish white (2A2), turning blue when exposed, red with NH<sub>4</sub>OH. *Hymenophore* 2 - 6 mm thick, lamellate, decurrent with eroded edges, yellow (2A7), subdistant with 5 lamellulae per 5 mm in 2 series, turning blue when bruised, negative reaction with NH<sub>4</sub>OH. *Stipe* 17 - 30 × 2 – 5 mm, central to slightly eccentric, equal to slightly flexuous, dry, fibrillose, dark brown (6F6), white at the base, with fistulose context, no colour changed when exposed.

*Basidiospores* (10-) 11-14 (-15) × 5 -6 μm [  $x_m = 13.08 \pm 1.26 \times 6.46 \pm 1.02$  μm,  $x_{mr} = 10.55 - 13.08 \times 5.08 - 6.46$  μm,  $x_{mm} = 12.18 \pm 1.79 \times 5.77 \pm 0.97$  μm,  $Q = (1.67-)$  1.83 – 2.36 (- 2.5),  $Q_m = 2.22 \pm 0.21$ ,  $Q_{mr} = 2.08 - 2.22$ ,  $Q_{mm} = 2.15 \pm 0.41$ ,  $n = 50$  spores per 2 collections], ellipsoid to subfusoid, smooth, inamyloid, hyaline in KOH. *Basidia* (25-) 32 - 35(-46) × 9-11 μm, 4-sterigmate, clavate, hyaline in KOH, inamyloid. *Pleurocystidia* (51-) 60 – 69 (-87) × (10-) 11 – 14 μm, metuloid in shape, hyaline in KOH, inamyloid, thick-walled. *Cheilocystidia* (61-) 80 – 90 × (10-) 12 - 13 μm, metuloid, hyaline in KOH, inamyloid, thick-walled. *Hymenophoral trama* divergent (boletoid), with hyaline elements, inamyloid. *Pileipellis* a palisade trichodermium or and enterocutis clavate leptocystidia, smooth, thin-walled, hyaline, inamyloid. *Caulocystidia* a trichodermium of caulocystidia, (27-) 32 - 45 × 7 - 9 (-13) μm, cylindrical to subclavate, smooth, thin-walled, inamyloid. *Clamp connections* absent.

*Habit, habitat, and distribution:* gregarious on soil in forests of highland oak trees; reported from Peninsular Malaysia, Terengganu, Kemaman and Bukit Kajang.

*Material examined:* MALAYSIA. Pahang, Fraser Hill, Pine Trail, N 03°44'27.0", E 101°47'30.6", 980m, 8 January 2006, *leg. Chan H.T.* (KLU-M1078); Jalan Air Terjun Jeriau, N 03°43'36.0", E 101°43'00.6", 970m; 8 January 2006. *leg. Sumaiyah A.* (KLU-M1079).

*Commentary:* Collection KLU-M1078 matches Corner's but with the addition of cyanescent context and slightly larger spores. The finely squamulose pileus with brown squamules and pale context showing through, the cyanescent lamellae, and stipe base with white mycelium appear diagnostic. KLU-M1079 is an old, water soaked specimen, but appears to agree in all essentials.



**Figure 4.3:** *Phylloporus brunneolus* Corner (KLU-1078). **a.** Basidiomes **b.** Basidiospores. **c.** Basidia. **d.** Pleurocystidia. **e.** Cheilocystidia. **f.** Pileipellis. **g.** Stipitipellis. Bars: a = 10 mm. b – e = 10  $\mu$ m. f – g = 20  $\mu$ m.

4. *Phylloporus cf. orientalis* Corner, Nova Hedwigia 20: 809. 1970.

(Fig. 4.4a, 4.4b, Plate 4.1 D)

*Pileus* 54 mm broad, plano-concave, dry, with incurved and eroded margin, tomentose, pale orange (5A3), not bruising, negative reaction with NH<sub>4</sub>OH. *Context* 4 mm thick, soft and spongy, white, blue when exposed, negative reaction with NH<sub>4</sub>OH. *Hymenophore* lamellate 7 mm thick, with anastomosis, decurrent, yellow (1A7), subdistant with 4-5 lamellulae per 5 mm with 2 series, bruising blue, negative reaction with NH<sub>4</sub>OH. *Stipe* 22 mm long, 4 mm broad, central to eccentric, equal to slightly bulbous, dry, fibrillose, golden brown (5D7), white basal mycelium; context solid blue when exposed, negative reaction with NH<sub>4</sub>OH.

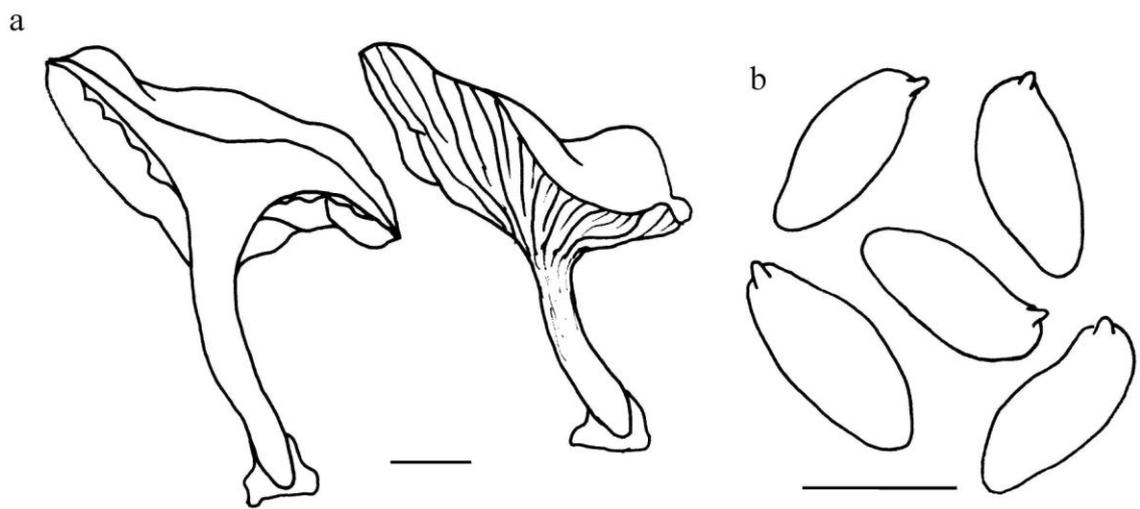
*Basidiospores* 10 - 14 × (4-) 5 - 6 μm [  $x_m = 12.14 \pm 1.42 \times 5.60 \pm 0.56$  μm, Q = 1.67 - 2.80, Q<sub>m</sub> = 2.19 ± 0.29, n = 31 spores per 1 collections], narrow ellipsoid to long subfusoid, smooth, olive brown in mass, inamyloid, hyaline in KOH. *Basidia* (25-) 32 - 35 (-46) × 9 - 11 μm, 4-sterigmate, clavate, hyaline in KOH, inamyloid. *Cheilocystidia* 68 - 89 (-97) × (16-) 19 - 23 (-38) μm, clavate in shape, hyaline in KOH, inamyloid, thick-walled. *Pleurocystidia* 65 - 81 (-87) × 15 - 21 (-26) μm, clavate to cylindrical in shape, hyaline in KOH, inamyloid, thick-walled. *Hymenophoral trama* 6 - 14 μm broad, divergent (boletoid), with hyaline elements, inamyloid. *Pileipellis* a hymeniform layer of long pileocystidia, 100 - 130 μm high; *pileocystidia* 50 - 67 × 11 - 17 (-19) μm, long subclavate to subventricose, smooth, thin-walled, hyaline, inamyloid. *Stipitipellis* a trichodermium of caulocystidia, 25 - 34 (-42) × (8-) 10 - 13 μm, clavate to subventricose, smooth, thin-walled, hyaline, inamyloid. *Clamp connections* absent.

*Habit, habitat, and distribution:* solitary on soil in forests of lowland Dipterocarpaceae.

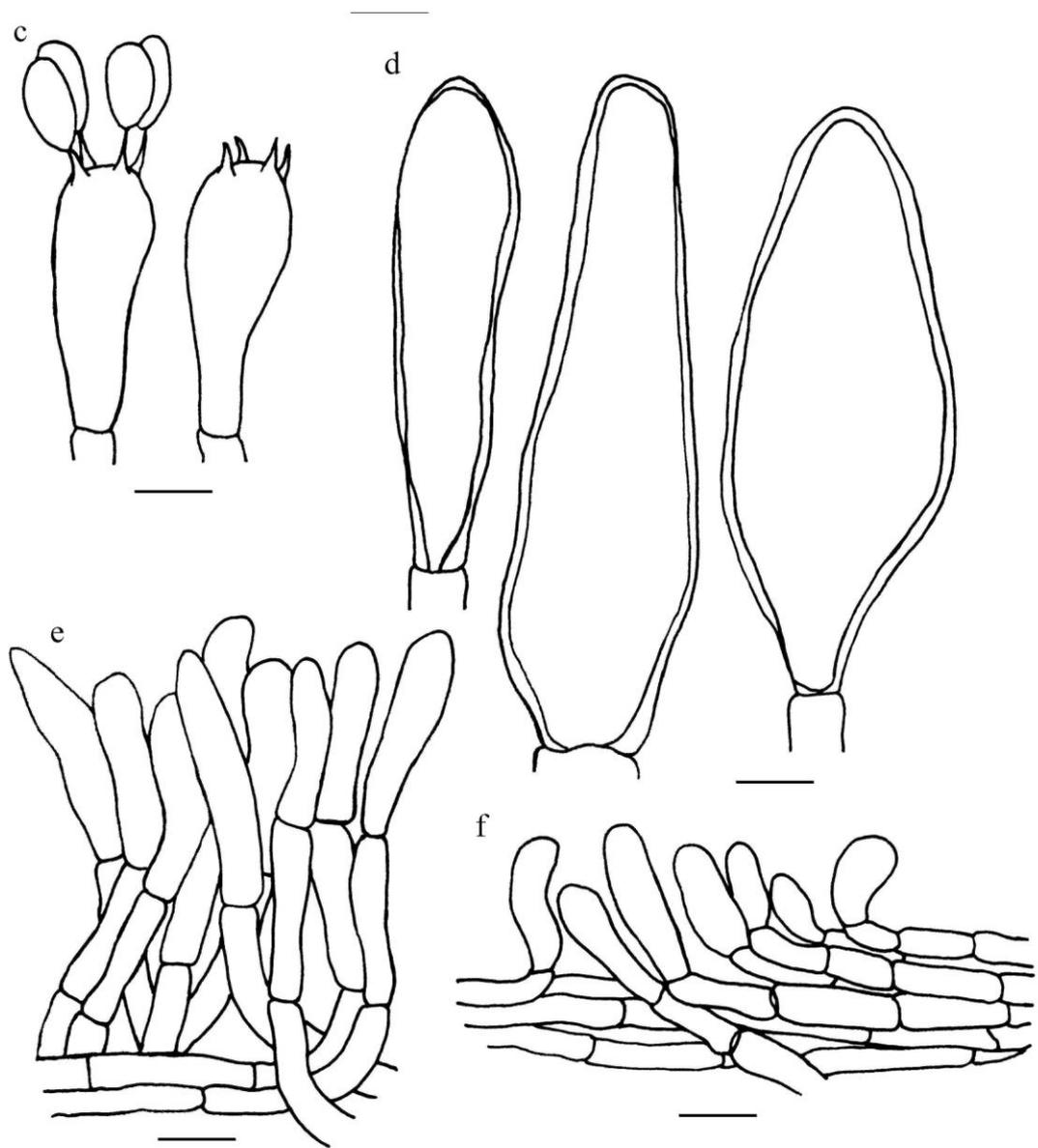
*Material examined:* MALAYSIA. Johor. Endau Rompin National Park (Peta).

N 02° 32' 30.6", E 103° 22' 08.9", 28 m, 13 July 2005, leg. Chan H.T. (KLU-M1080).

*Commentary:* This solitary, mature basidiome has brown pileus, white context and white at the stipe base, the cyanescent tissues (not reddening or blacking) fits to Corner's species. The spore size (10 - 14 × (4-) 5 - 6 μm) pointing toward *P. orientalis*. However, in KLU-M1080, the hymenial cystidia are broader and with slightly thickened walls which is different from thin-walled Corner's description.



**Figure 4.4a:** *Phylloporus cf. orientalis* Corner (KLU-M1080). **a.** Basidiomes. **b.** Basidiospores. Bars: a = 10 mm. b = 10 μm.



**Figure 4.4b:** *Phylloporus cf. orientalis* Corner (KLU-M1080). **c.** Basidia. **d.** Cheilocystidia. **e.** Pileipellis. **f.** Stipitipellis. Bars: a = 10 mm. b – d = 10  $\mu\text{m}$ . e – f = 20  $\mu\text{m}$ .

5. *Phylloporus* cf. *sulcatus* (Pat.) Gilbert, Les bolets, p. 88. 1931, ss Corner.

(Fig. 4.5, Plate 4.1 B)

*Pileus* 32 mm broad, convex, dry, with straight to slightly decurved margin, velvety to subtomentose, brown (7E4). *Context* 3 mm thick at disc, 1 mm thick at margin, soft and spongy, white, no colour changed when exposed, negative reaction with NH<sub>4</sub>OH. *Hymenophore* 4 mm thick, lamellae, decurrent with smooth edges, yellow (2A7), subdistant with 1 – 2 lamellulae per mm with 2 series, slightly veined at base, not bruising, negative reaction with NH<sub>4</sub>OH. *Stipe* 32 mm × 4 mm, central, equal and cylindrical, dry, fibrillose, light brown at apex, brown at base, with white basal mycelium, no colour changed when exposed and negative reaction with NH<sub>4</sub>OH.

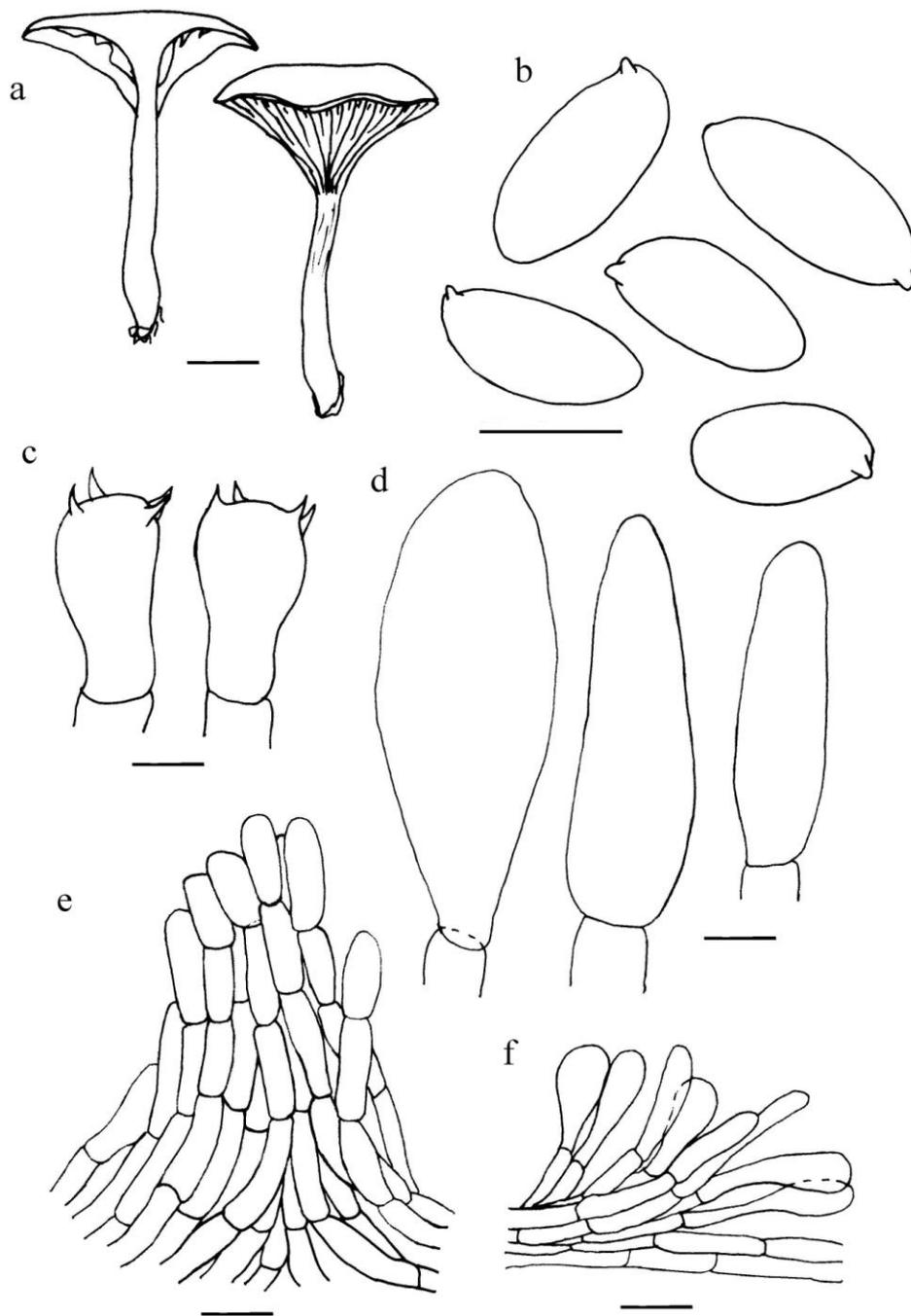
*Basidiospores* (9.5-) 10 – 14 (-15) × (4.5-) 5 – 6 μm [  $x_m = 12.33 \pm 1.70 \times 5.40 \pm 0.48$  μm,  $Q = (1.81-) 1.90 - 2.73 (- 2.8)$ ,  $Q_m = 2.29 \pm 0.29$ ,  $n = 20$  spores per collection], ellipsoid to subfusoid, smooth, inamyloid, hyaline in KOH. *Basidia* (19-) 23 – 25 (-31) × (6-) 8 – 10 (-11) μm, 4-sterigmate, clavate, hyaline in KOH, inamyloid. *Hymenial cystidia* (35-) 40 – 60 (-70) × 10 – 15 μm, clavate to cylindrical, hyaline in KOH, inamyloid, thin-walled. *Hymenophoral trama* divergent (boletoid), with hyaline elements, inamyloid. *Pileipellis* a palisade trichodermium, 120 – 140 μm high, smooth, thin-walled, hyaline, inamyloid. *Caulocystidia* forming a trichodermium 20 – 40 μm high, cylindric to subclavate to clavate, smooth, thin-walled, inamyloid. *Clamp connections* absent.

*Habit, habitat, and distribution:* solitary on soil under a Dipterocarp tree; reported from Peninsular Malaysia.

*Material examined:* MALAYSIA. Johor. Endau Rompin National Park, Kuala Jasin, N 02° 32' 30.6", E 103° 22' 08.9", 28m, 17 July 2005, leg. Chan H.T. (KLU-M1081).

*Commentary:* Again, the single basidiome is brown, has a white basal mycelium on the stipe, and lacks any color change when bruised. The cystidia are not fusoid, rather

they are broadly cylindrical, thin-walled but not abundant as in most species. Supposedly, the species was noted to have a sulcate stipe apex. Such a feature was not noted for KLU-M1081. Corner (1970) had not seen fresh material.



**Figure 4.5:** *Phylloporus cf. sulcatus* Pat. (KLU-M1081). **a.** Basidiomes **b.** Basidiospores. **c.** Basidia. **d.** Hymenial cystidia **e.** Pileipellis. **f.** Stipitipellis. Bars: a = 10 mm. b-f = 10  $\mu$ m.

6. *Phylloporus* sp. 1.

(Fig. 4.6a, 4.6b, Plate 4.1 A)

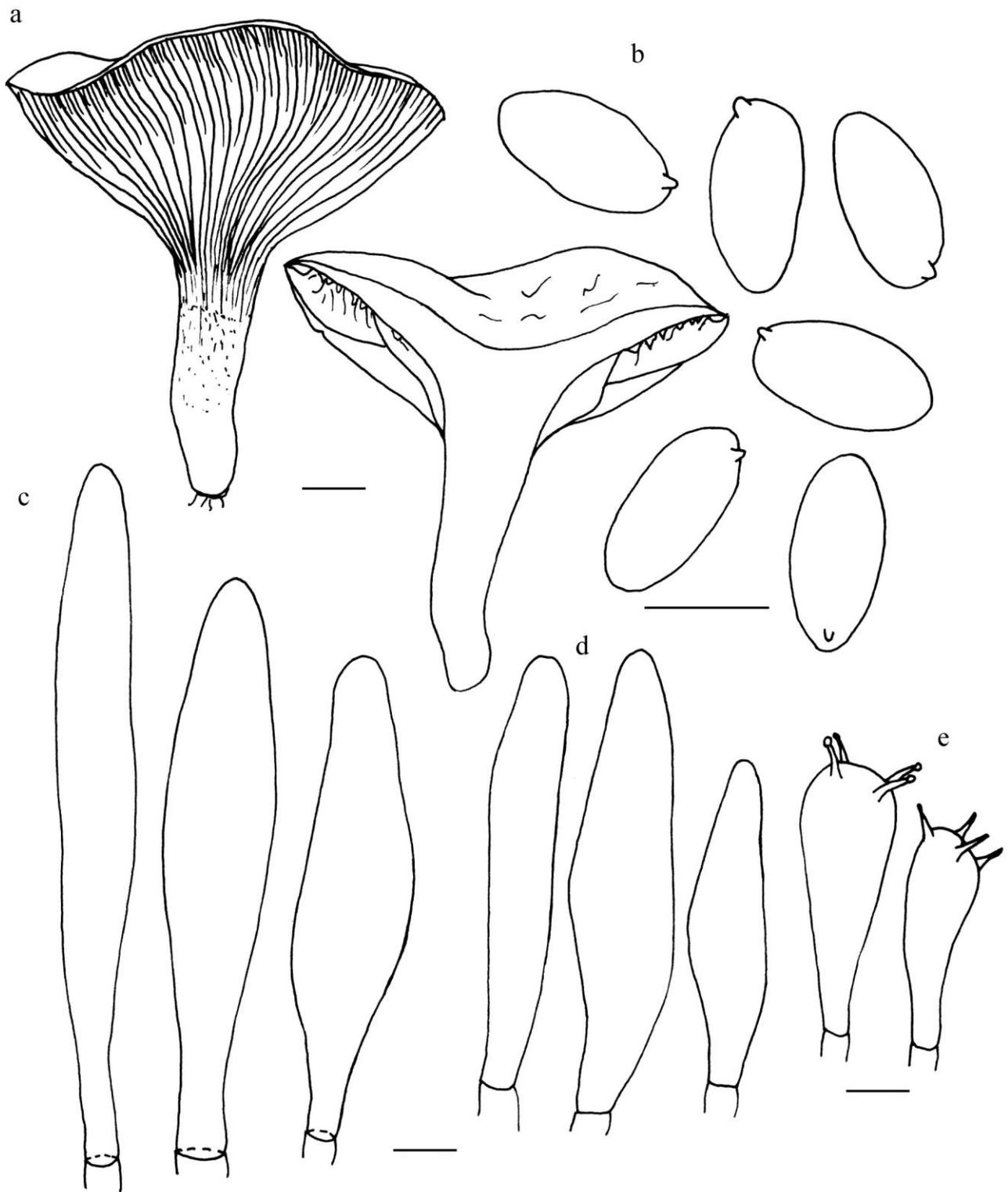
*Pileus* 72 mm broad, depressed at disc, with arched margin, dry, velvety, light brown (5D5), reddish orange with NH<sub>4</sub>OH. *Context* 10 mm thick at disc, 1 mm thick at margin, soft and spongy, yellowish white (2A2), no changed when exposed, negative reaction with NH<sub>4</sub>OH. *Hymenophore* 6 mm thick, lamellate, deeply decurrent with slightly eroded edges, yellow (2A7), close with 30 lamellulae in 2 series, intervenose, no changed when exposed, negative reaction with NH<sub>4</sub>OH. *Stipe* 44 × 14 mm at the apex, 9 mm broad at the base, central, tapering down, dry, punctate surface with extension of lamellae at apex, light brown (6F6) to brownish white (4A2), white at the base, with solid context, no colour changed when exposed, negative reaction with NH<sub>4</sub>OH.

*Basidiospores* (9-) 11 – 13 (-14) × 5 – 6 μm [ $x_m = 11.73 \pm 1.14 \times 5.73 \pm 0.55$  μm, Q = 1.83 – 2.60, Q<sub>m</sub> = 2.06 ± 0.20, n = 30 spores per one collections], ellipsoid to subfusoid, smooth, light olive green, inamyloid, hyaline in KOH. *Basidia* 30.0 – 34.5 × 8.9 – 10.6 μm, 4-sterigmata, clavate, hyaline in KOH, inamyloid. *Pleurocystidia* 52.0 – 64.0 × (9.6-) 13.6 – 16 μm, cystidia shorter than cheilocystidia, subventricose to subcylindrical, hyaline in KOH, inamyloid, thin-walled. *Cheilocystidia* 51.2 – 88.3 × 11.4 – 12.6 (-13.6) μm, long cylindrical to subventricose, hyaline in KOH, inamyloid, thin-walled. *Hymenophoral trama* divergent, with hyaline elements, inamyloid. *Pileipellis* a trichodermium of long cylindrical pile hyphae, 2 – 4 separate hyphae with - 120 μm, smooth, thin-walled; *pileocystidia* 38 – 48 × 6 – 10 μm, slightly brownish, inamyloid. *Stipitipellis* a palisade trichodermium of caulocystidia 50 – 60 μm high; *caulocystidia* 25 – 40 × 11 – 18 μm, short subclavate to clavate, smooth, thin-walled, inamyloid. *Clamp connections* absent.

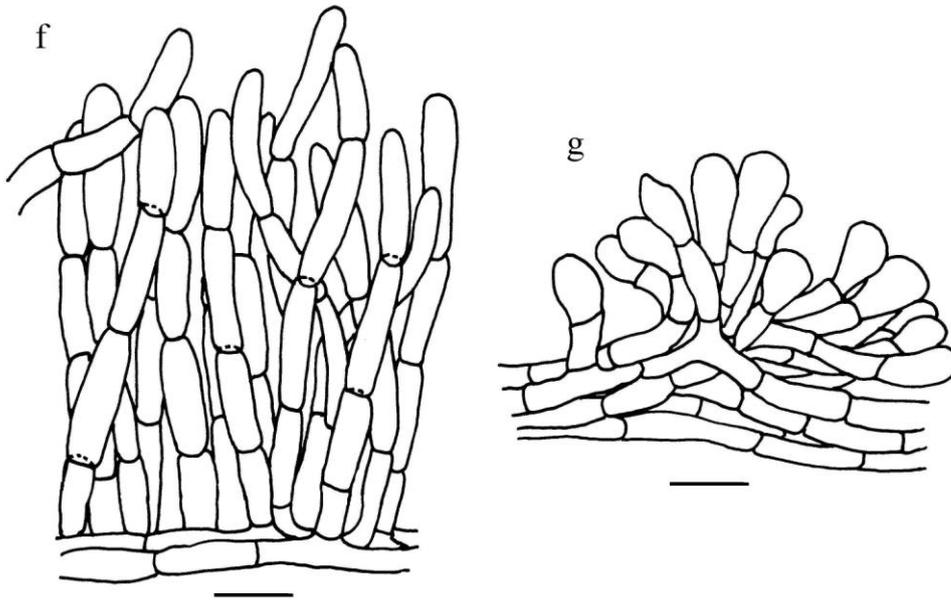
*Habit, habitat, and distribution:* solitary on soil under *Syzygium* sp. in forests of lowland Dipterocarpaceae.

*Material examined:* MALAYSIA. Johor. Endau Rompin National Park (Peta). Kompleks Pejabat T.N.E.R., Aboretum Trial, N 02° 31'47.9", E 103° 24' 49.9", 20 m, 22 May 2007, *leg. Chan H.T.* (KLU-M1082).

*Commentary:* This collection consists of a single large basidiome with a pale yellow brown pileus surface. Also, it is not cyanescent or rufescent in any part, has pale yellow context and a white basal mycelium at the base of the stipe. With these features, it is not close to any taxon discussed by Corner (1970). It is close to a taxon from Australia included as "*Phylloporus* sp.1" included in an unpublished key written by M.A. Neves (2010).



**Figure 4.6a: *Phylloporus* sp. 1 (KLU-M1082).** **a.** Basidiomes. **b.** Basidiospores.  
**c.** Cheilocystidia. **d.** Pleurocystidia. **e.** Basidia. Bars: a = 10 mm. b – e = 10  $\mu$ m.



**Figure 4.6b** : *Phylloporus* sp. 1 (KLU-M1082). **f.** Pileipellis. **g.** Stipitipellis. Bars: f – g  
= 20  $\mu\text{m}$ .

7. *Phylloporus* cf. *bellus* (Masse) Corner, Nova Hedwigia 20: 798. 1970.  
(Fig. 4.7, Plate 4.1 G)

*Pileus* 18 mm broad, plane with revolute margin, dry, slightly areolate at disc, velvety at margin, greyish brown (6F3), not bruising. *Context* 1.5 mm thick at disc, soft and spongy, yellowish white (3A3), not bruising. *Hymenophore* 2 mm thick, lamellae, decurrent with even edges, yellow (3A6), subdistant with 16 - 21 lamellulae in 2 series, not bruising. *Stipe* 20 × 3 mm, central, equal to slightly tapering upward to slightly flexuous, dry, pruinose at apex, fibrillose at base, light brown (5D5), white at the base, with context solid, slowly brown when exposed. .

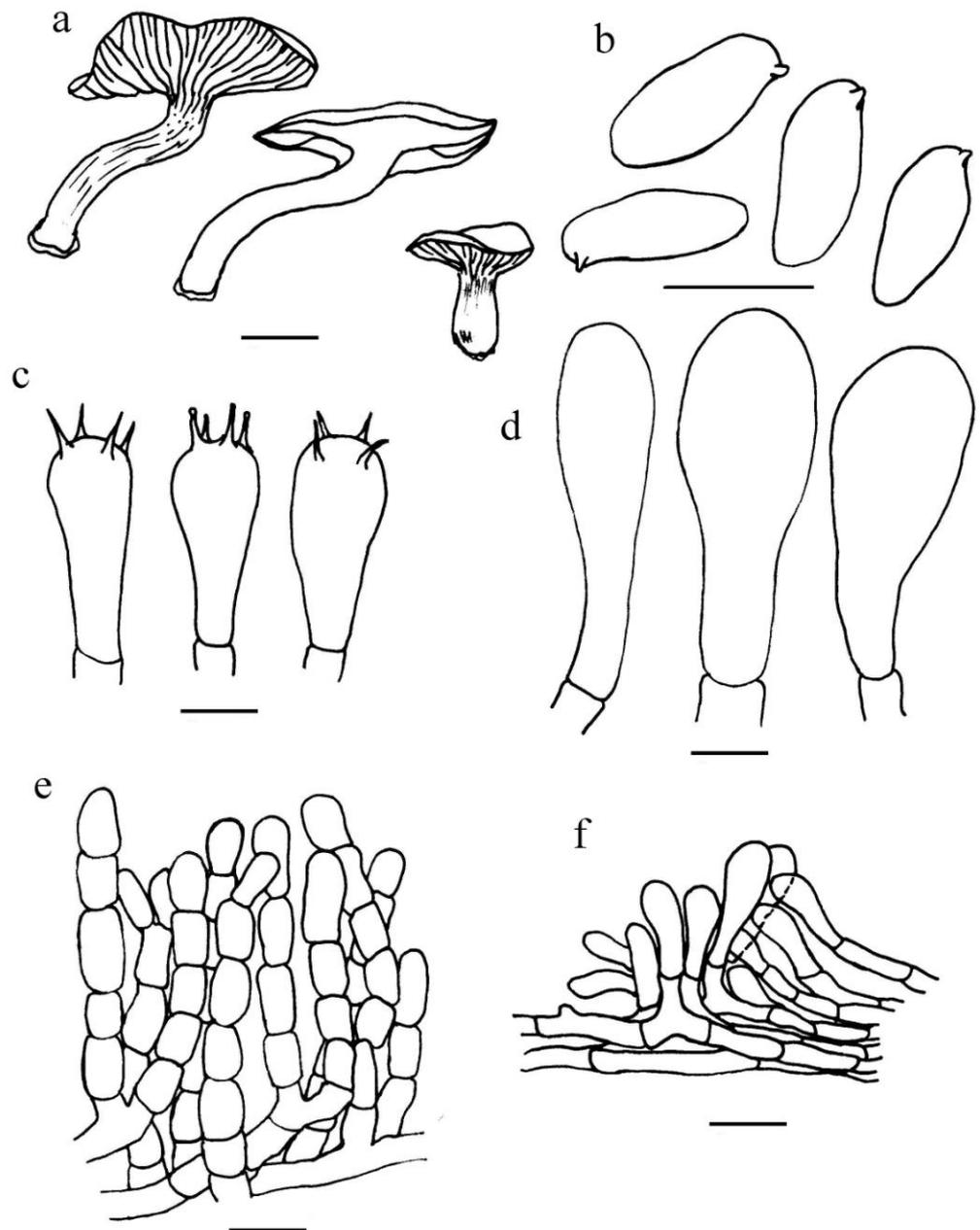
*Basidiospores* (10-) 11-14 (-15) × 5 -6 (-7) μm [  $x_m = 12.35 \pm 1.14 \times 5.30 \pm 0.57$  μm,  $Q = (1.43-) 2.0 - 2.8$ ,  $Q_m = 2.40 \pm 0.36$ ,  $n = 20$  spores per one collection], ellipsoid to subfusoid, smooth, inamyloid, hyaline in KOH. *Basidia* 28 – 30 (-31) × 12 – 14 μm, 4-sterigmate, length of sterigmata 4 – 6 μm, clavate, hyaline, inamyloid. *Pleurocystidia* (41-) 44 – 53 × (13-) 14 – 16 (-20) μm, same with cheilocystidia. *Cheilocystidia* (49-) 58 – 67 (-77) × (15-) 16 - 21 μm, long clavate, hyaline, inamyloid. *Hymenophoral trama* divergent (boletoid), with hyaline elements, inamyloid. *Pileipellis* a structure of moniliform hyphae, forming a pile of end cells with 30 – 40 × 15 – 20 μm, thin-walled, hyaline, inamyloid. *Stipitipellis* a trichodermial palisade of caulocystidia 50 μm high, oleiferous hyphae present; *caulocystidia* 20 – 30 × 8 – 12 μm, subclavate to clavate, smooth, thin-walled, inamyloid. *Clamp connections* absent.

*Habit, habitat, and distribution:* solitary to gregarious on soil in Dipterocarp forests. Reported from Peninsular Malaysia and Sarawak.

*Material examined:* MALAYSIA. Selangor, Hulu Langat, Sungai Chongkak Forest Reserve, N 03°12'00.3", E 101°50'07.4", 160m. 16 August 2006. *leg. Chan H.T.* (KLU-M1083).

*Commentary:* Two basidiomata with brown pileus and stipe, and a subtle cyanescence in the white context come close to this species. However, it also has some

resemblance to *P. bogoriensis*. In reading Corner's commentaries of these two taxa, there appears to be some overlap in diagnostic features.



**Figure 4.7:** *Phylloporus* cf. *bellus* (Masseé). (KLU-M1083). **a.** Basidiomes **b.** Basidiospores. **c.** Basidia. **d.** Hymenial cystidia. **e.** Pileipellis. **f.** Stipitipellis. Bars: a = 5 mm. b – d = 10  $\mu$ m. e – f = 20  $\mu$ m.



**Plate 4.1:** A: *Phylloporus* sp. 1 ( $\times 4.8$ ); B: *Phylloporus* cf. *sulcatus* ( $\times 2.1$ ); C: *Phylloporus megaporinus* ( $\times 3.8$ ); D: *Phylloporus* cf. *orientalis* ( $\times 3.6$ ); E: *Phylloporus bogoriensis* ( $\times 6.5$ ); F: *Phylloporus brunneolus* ( $\times 3.3$ ); G: *Phylloporus* cf. *bellus* ( $\times 1.2$ ); H: *Tylopilus pernanus* ( $\times 1.3$ ). Bar = 10mm.

8. *Pulveroboletus cf. viridis* Heinem. & Gooss., Bull. Jard. Bot. Etat Brux. 21: 306. 1951,  
ss Corner. (Fig. 4.8, Plate 4.2 C)

≡ *Boletus viridis* (Heinem. & Gooss.) Corner, *Boletus* in Malaysia, p. 197. 1972.

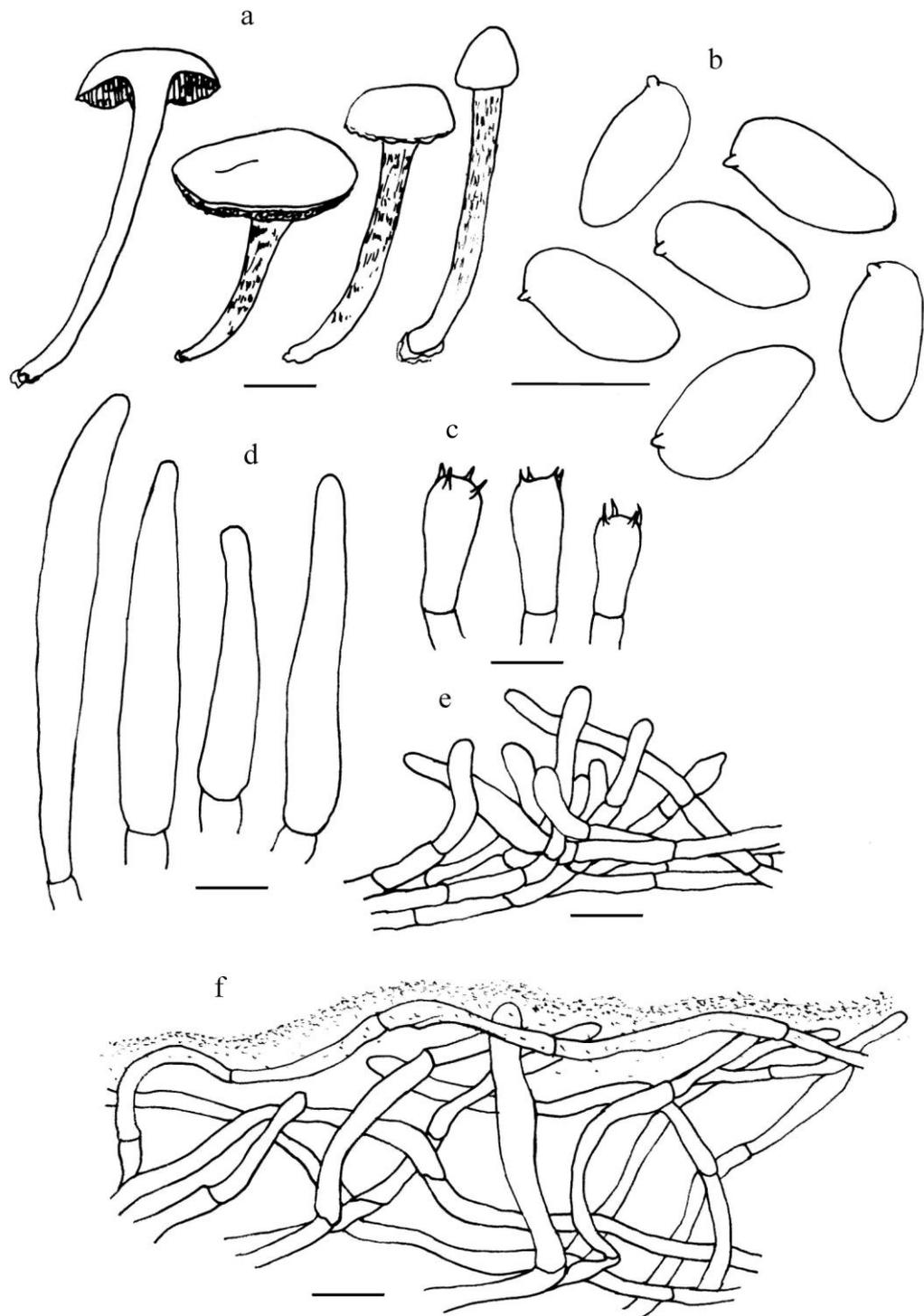
*Pileus* 20 – 28 mm broad, conical when young, convex when mature, viscid, with decurved margin when young, becoming straight with appendiculate sterile extension white (1A1), negative reaction with NH<sub>4</sub>OH. *Context* 4 mm thick, 1 mm thick at margin, white, no colour changed when exposed, negative reaction with NH<sub>4</sub>OH. *Hymenophore* 4 – 5 mm thick, tubulose, adnate, white when young (1A1), light brown (5D8) to greyish green (1D4) when mature or old, with concolorous pores, 2 – 3 pores per mm, no colour changed when exposed, turning slightly pink with NH<sub>4</sub>OH. *Stipe* 30 – 46 × 6 mm at apex, 3 – 4 mm at base, central, tapering downward, white (1A1), slightly scabrous with viscid surface, no colour changed when bruised or negative reaction with NH<sub>4</sub>OH, with white basal mycelium; interior white and solid.

*Basidiospores* (10-) 11 – 14 × 6 – 7 μm [  $x_m = 12.07 \pm 1.07 \times 6.18 \pm 0.36$  μm,  $Q = 1.67 - 2.17$ ,  $Q_m = 1.95 \pm 0.12$ ,  $n = 30$  spores per one collection], ellipsoid to boletoid, olive green, smooth, inamyloid. *Basidia* 12 – 16.8 × 5.2 – 7.2 μm, 4-sterigmate, short-clavate, hyaline, inamyloid. *Hymenial cystidia* (32-) 42.4 – 60 × (5.6-) 7.2 – 8.0 μm, cylindrical with narrow tip, thin-walled, hyaline, inamyloid. *Pileipellis* an ixotrichodermium of hyphae and cystidia, 100 -120 μm high, thin-walled, hyaline, inamyloid. *Stipitipellis* a trichodermium of caulocystidia, with end cells cylindrical to subventricose, hyaline, inamyloid. *Clamp connections* absent.

*Habit, habitat, and distribution:* scattered on soil in forests of Dipterocarpaceae (*Shorea* sp.).

*Material examined:* MALAYSIA. Negeri Sembilan, Pasoh, Hutan Simpan, N 2°58'7", E 102°17'49", 111 m, 20 September 2006, leg. Chan H.T. (KLU-M1084).

*Commentary:* This collection is deceptively similar to the illustration of this taxon as portrayed by Corner (pl. 11, 3; 1972). Also, Corner describes the spores as “ferruginous ochraceous in mass” which would point to *Tylopilus* s.l. He also said the spores were bright golden yellow under the microscope. The illustration shows a basidiome with a hollow stipe. My collection was nearly pure white and viscid with only a hint of pale greenish, the spores were nearly hyaline and shorter (cf. 13-17 x 4-5  $\mu\text{m}$ ), and the stipe was solid. Also, there are squamules on the stipe and an appendiculate sterile extension of the pileus margin. However, there was only the slightest indication of a connection between the margin and stipe in very young material.



**Figure 4.8:** *Pulveroboletus cf. viridis* (KLU-M1084). **a.** Basidiomes **b.** Basidiospores. **c.** Basidia. **d.** Pleurocystidia. **e.** Cheilocystidia. **f.** Pileocystidia. Bars: a = 10 mm. b – f = 10  $\mu$ m.

9. *Pulveroboletus viridisquamosus* Watling, Turnbull, & Lee, Bull. Soc. Mycol. France  
122: 340. (Fig. 4.9, Plate 4.2 D)

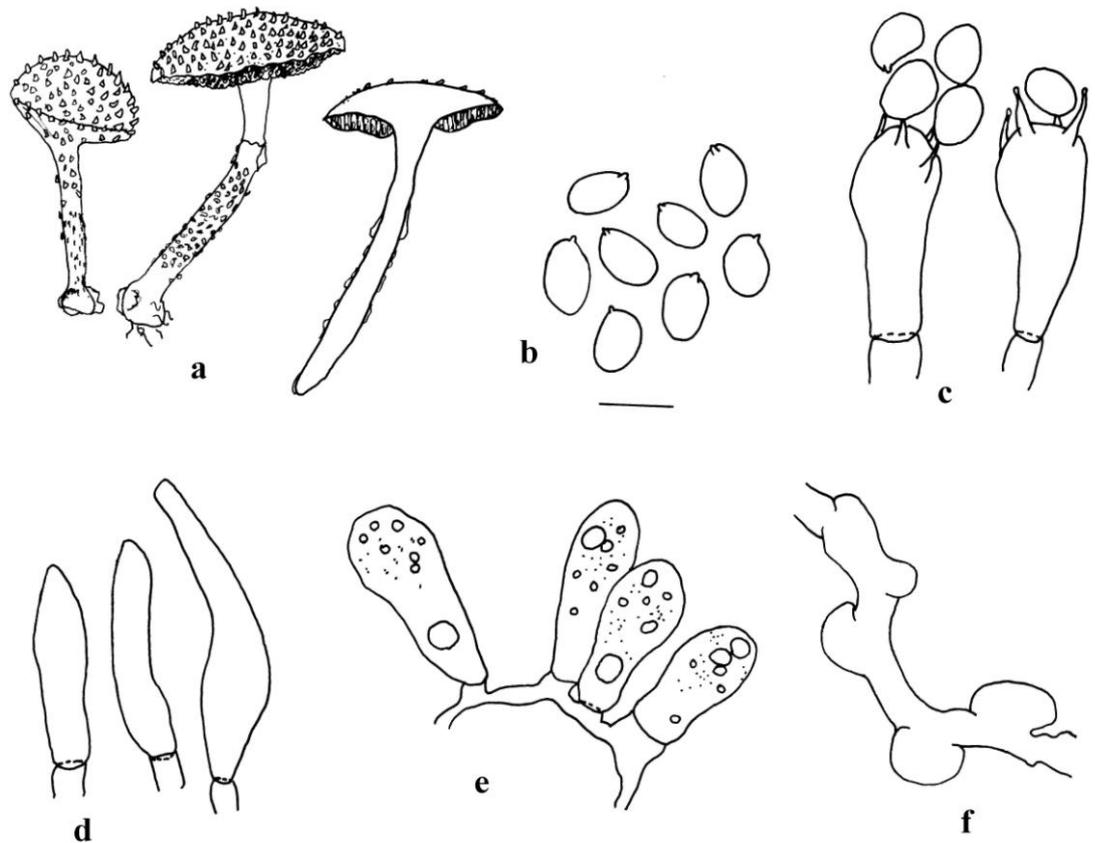
*Pileus* 55 – 73 mm broad, convex, dry, scaly surface; disc pastel green (30A4) with olive scales (2E4); margin straight with appendiculate edge, orange with NH<sub>4</sub>OH. *Context* 15 mm thick at disc, 0.1 cm thick at margin, white, soft and spongy, bruising slightly blue, negative reaction with NH<sub>4</sub>OH. *Hymenophore* tubulose, 6 mm thick, deeply depressed to free; *tubes* olive yellow (2C7), blue when injured, blue with NH<sub>4</sub>OH; *pores* 2 pores per mm, concolorous with tubes. *Stipe* 6-9 cm long, 0.9-1 cm broad, equal to slightly tapering downward, central, dry, deeply scaly to powdery, pedant annulus present; yellowish green (30B7), orange with NH<sub>4</sub>OH; interior solid, context yellowish green (30B8); *basal mycelium* yellowish white.

*Basidiospores* (6-) 7 – 9 (-10) × 5 - 6 μm, [ $x_m = 7.73 \pm 0.86 \times 5.43 \pm 0.49$  μm,  $Q = 1.0 - 2.0$ ,  $Q_m = 1.42 \pm 0.23$ ,  $n = 30$  spores per collection], smooth, ovoid to subglobose to subellipsoid, olive brown in mass, hyaline in KOH, inamyloid. *Hymenial cystidia* (29-) 30 – 35 (-45) × 4 – 6 (-9) μm broad, cylindrical to slightly ventricose, thin-walled, hyaline in KOH, inamyloid. *Pileipellis* trichodermium palisade with bright yellow enterocutis structure, thin-walled, branching hyphae occur, inamyloid. *Stipitipellis* a trichodermium, with hyphae (3-) 5 – 6.5 μm broad, branching, with oleiferous hyphae present, inamyloid. *Clamp connections* absent.

*Habit, habitat, and distribution:* gregarious on soil in forests of Dipterocarpaceae (*Shorea* sp.), reported from Negeri Sembilan, Peninsular Malaysia.

*Material examined:* MALAYSIA. Negeri Sembilan, Pasoh, Hutan Simpan, N 2°58'7", E 102°17'49", 111 m, 20 September 2006, leg. Chan H.T. (KLU-1085).

*Commentary:* This species was firstly reported by Watling *et al.*, (2006). The species has distinct olive green with pileus and stipe ornamented with tufts of powdery component. The smooth and almost globose spores differentiate this species with *Afroboletus luteolus* (Heinem.) Pegler & Young from Africa which has a similar appearance of colour (Watling *et al.*, 2006). It was collected at the Pasoh Forest which is the same location where Watling collected. This species is common in Pasoh Forest.



**Figure 4.9:** *Pulveroboletus viridisquamosus* Watling, Turnbull, & Lee, Bull (KLU-M1085). **a.** Basidiomes. **b.** Basidiospores. **c.** Basidia. **d.** Cheilocystidia. **e.** Basidiales. **f.** Oleiferous hyphae. Bars: a = 20 mm. b – f = 10  $\mu$ m.

10. *Pulveroboletus ridleyi* (Masse) Watling, Bull. Soc. Mycol. France 122: 340. 2006.

(Fig. 4.10, Plate 4.2 F)

≡ *Boletus ridleyi* Masse, Kew Bull. Misc. Inform. XX: 20. 1901, *non* Masse 1909.

≡ *Pulveroboletus icterinus* (Pat. & C.F. Baker) Watling, Not. Roy. Bot. Gard. Edinburgh 46: 413. 1990.

≡ *Boletopsis icterinus* Pat. & C.F. Baker, J. Straits Branch Royal Asiatic Soc. 78: 68. 1918.

*Pileus* 12 – 29 mm broad, convex, dry, pulverulent with yellow powder, yellow (1A7, 3A6), with decurved margin, bruising slightly blue. *Context* 2 - 4 mm thick, soft and spongy, white, bruised blue when exposed, negative reaction with NH<sub>4</sub>OH. *Hymenophore* (2-) 3 – 4 mm thick, tubulose, sinuate, greyish yellow (1B4) when young, greyish green (1D4) when mature, with concolorous pores less than 1 mm broad, bruising blue, slightly pink with NH<sub>4</sub>OH. *Stipe* (25-) 30 – 43 × 5 – 6 mm at apex, 3 – 4 mm broad at base, central, tapering downward, dry, scabrous with yellow powder above the ring, opaque yellow (3A6), no colour changed when bruised or with NH<sub>4</sub>OH. White basal mycelium; interior white and solid, blue when exposed, red with NH<sub>4</sub>OH.

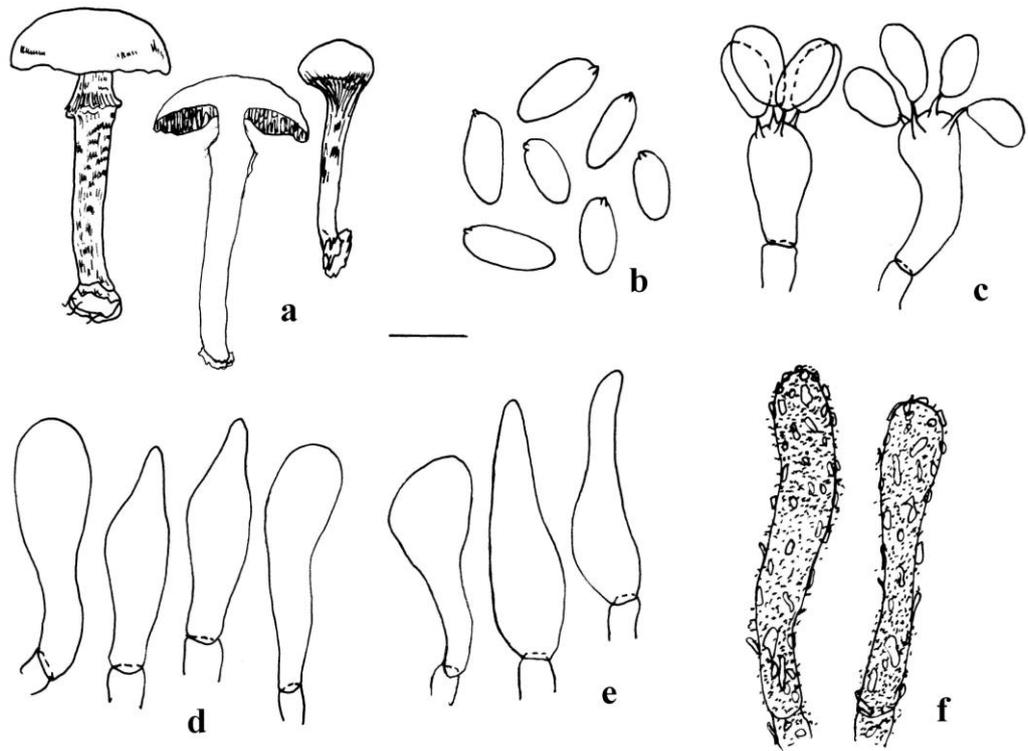
*Basidiospores* (8-) 9 – 11 × 4 – 5 (-6) μm [  $x_{mr} = 8.38 - 10.25 \times 4.25 - 5.58$  μm,  $x_{mm} = 9.45 \pm 0.96 \times 4.87 \pm 0.67$  μm, Q = 1.5 – 2.5,  $Q_{mr} = 1.84 - 2.07$ ,  $Q_{mm} = 1.96 \pm 0.12$ , n = 60 spores per 3 collections], ellipsoid to subfusoid, smooth, inamyloid. *Basidia* (13-) 14 – 20 × 7 – 10 μm, 4-sterigmate, short-clavate, hyaline, inamyloid. *Pleurocystidia* 25 – 30 (-35) × (7-) 8 – 10 μm, ventricose to clavate, thin-walled, hyaline, inamyloid. *Cheilocystidia* 26 – 30 × (6-) 7 – 10 (-11) μm, ventricose to clavate, hyaline, inamyloid, thin-walled. *Hymenophoral trama* divergent (boletoid), with hyaline elements, inamyloid. *Pileipellis* an ixotrichodermium, 45 – 57 × 5 – 9 μm broad, thin-walled,

powdery elements at pileocystidia, hyaline, inamyloid. *Stipitipellis* a trichodermium of caulocystidia, hyaline, inamyloid, with powdery elements absent. *Clamp connections* absent.

*Habit, habitat, and distribution:* gregarious on soil in forests of Dipterocarpaceae (*Shorea* sp.); reported from Peninsular Malaysia and Sarawak as well as Singapore.

*Material examined:* MALAYSIA. Pahang, Taman Negara Endau Rompin, Selai, 02°25'59.4" N, 103°14'49.5" E, 536 m, 18 February 2006, *leg. Chan H.-T.* (KLU-M1086); Pahang, Fraser's Hill, Jeriau Waterfall road, N 03°43'36.0", E 101°43'00.6", 680 m, 8 January 2006, *leg. Chan H.-T.* (KLU-M1087); Kedah, Langkawi Island, Datai, N 06°22'42.3", E 99°49'21.5", 59 m, 12 April 2003, *leg. Tan Y.S.* (KLU-M1088).

*Commentary:* This common species easily recognizable from the yellow colour which frequent species that develops at odd times during the year as well as in agaric fruiting season. This species look similar to *Pulveroboletus ravenelii* where both have yellow colour and changes to blue on injury. However, this species lack the dull red pileus surface under the sulphur yellow and with slender stipe, 3 – 6 mm thick.



**Figure 4.10:** *Pulveroboletus ridleyi* Masee (KLU-M1086). **a.** Basidiomes **b.** Basidiospores. **c.** Basidia. **d.** Pleurocystidia. **e.** Cheilocystidia. **f.** Pileocystidia. Bars: a = 10 mm. b-f = 10  $\mu$ m.

11. *Pulveroboletus claroflavus* Chan H.T. & Halling R.E., *sp. nov. prov.*  
(Fig. 4.11, Plate 4.2 G)

*Pileus* 20 – 30 mm broad, convex to slightly plane when mature, dry, pulverulent, straight to slightly decurved at margin, margin with pulverulent veil, yellow (1A7), negative reaction with NH<sub>4</sub>OH. *Context* 4 mm thick at disc, 1 – 2 mm thick at margin, solid and spongy, white (2A2), neither changing when exposed nor with NH<sub>4</sub>OH. *Hymenophore* 5 mm thick, tubulose, uncinata, pale yellow when young, light brown when mature, blue when exposed and stains blue with NH<sub>4</sub>OH; *pores* 4 – 5 per 5 mm, concolorous with tubes. *Stipe* 20 – 45 × 4 – 5 mm, central, hollow, slightly tapering downward, dry, slightly scabrous at veil, yellow (3A6), negative reaction with NH<sub>4</sub>OH, no colour changed when exposed. *Basal mycelium* with pale yellow rhizomorphs.

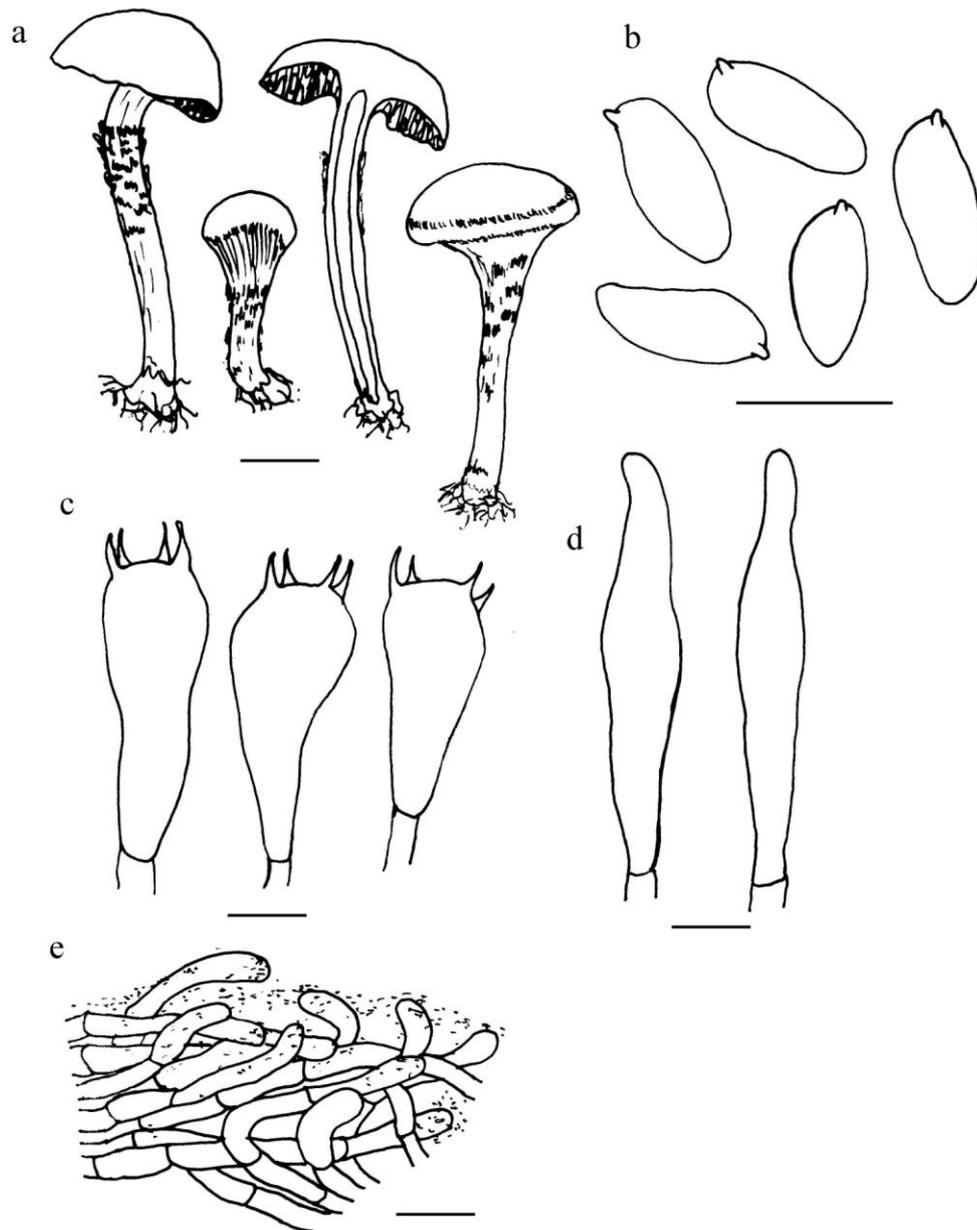
*Basidiospores* (9-) 10 – 13 (-14) × 4 – 7 μm [  $x_m = 11.23 \pm 1.32 \times 5.23 \pm 0.82$  μm,  $x_{mr} = 11.10 - 11.23 \times 4.90 - 5.23$  μm,  $x_{mm} = 11.17 \pm 0.09 \times 5.07 \pm 0.23$  μm,  $Q = 1.67 - 2.55$ ,  $Q_m = 2.18 \pm 0.31$ ,  $Q_{mr} = 2.18 - 2.27$ ,  $Q_{mm} = 2.23 \pm 0.06$ ,  $n = 40$  spores per 2 collections], ellipsoid to boletoid, smooth, olivaceous brown in light microscope, inamyloid, hyaline in KOH. *Basidia* (20-) 30 – 44 × 10 – 12 μm, 4-sterigmata, long clavate, hyaline in KOH, inamyloid. *Hymenial cystidia* (35-) 43 – 55 (-60) × (5-) 6 – 10 (-11) μm, long ventricose with thin-walled, hyaline in KOH, inamyloid. *Hymenophoral trama* divergent (boletoid), with hyaline elements, inamyloid. *Pileipellis* with interwoven cystidia, pileocystidia encrusted with yellow granular pigment, 3 – 5 μm in broad, inamyloid. *Stipitipellis* an interwoven layer of caulocystidia, encrusted with yellow granular pigment, thin-walled, hyaline and inamyloid. *Clamp connections* absent.

*Habit, habitat, and distribution:* gregarious on soil in forests of highland oak trees, grow under *Lithocarpus* spp.

*Material examined:* MALAYSIA. Pahang, Fraser Hill, Jalan Air Terjun Jeriau, N 03°43'36.0", E 101°43'00.6", 970m; 9 January 2006. *leg.* Chan H.T. (KLU-M1089);

Jalan Valley, N 03° 42' 41.5", E 101° 44' 18.6", 1253 m; 25 September 2003. *leg. Tan Y.S.* (KLU-M1090).

*Commentary:* This taxon is provisionally distinguished from the sympatric *P. icterinus* (see below) by possessing longer basidiospores. Otherwise, the lemon yellow colors are quite similar. Both taxa do not possess the red or brownish red pileus surface (under the yellow pulverulent veil) of *P. ravenelii* (Berk. & M.A. Curt.) Murrill.



**Figure 4.11:** *Pulveroboletus claroflavus* Chan H.T. & Halling R. *sp. nov. prov.* (KLU-M1089). **a.** Basidiomes. **b.** Basidiospores. **c.** Basidia. **d.** Hymenial cystidia. **e.** Pileipellis. Bars: a = 10 mm. b – d = 10  $\mu$ m. e = 20  $\mu$ m.

12. *Afroboletus malaysianus* Chan H.T. & Halling R.E. *sp. nov.*

(Fig. 4.12a, 4.12b, Plate 4.2 A,B)

*Pileus* 66 mm, convex with slimy and dull surface, glabrous, smooth, with a combination of dark brown and brown colors. *Context* 14 mm thick, stuffed, brown in colour. *Hymenophore* tubulose, concolourous with pores, 20 mm in thick; *pores* irregular 1-2 mm broad, reddish brown. *Stipe* 80 x 15 mm, cylindrical, central, dry and dull, shallowly and broadly subalveolate-reticulate throughout length, yellowish brown, white at base; *stipe context* broadly hollow, yellowish brown, turn brown when exposed.

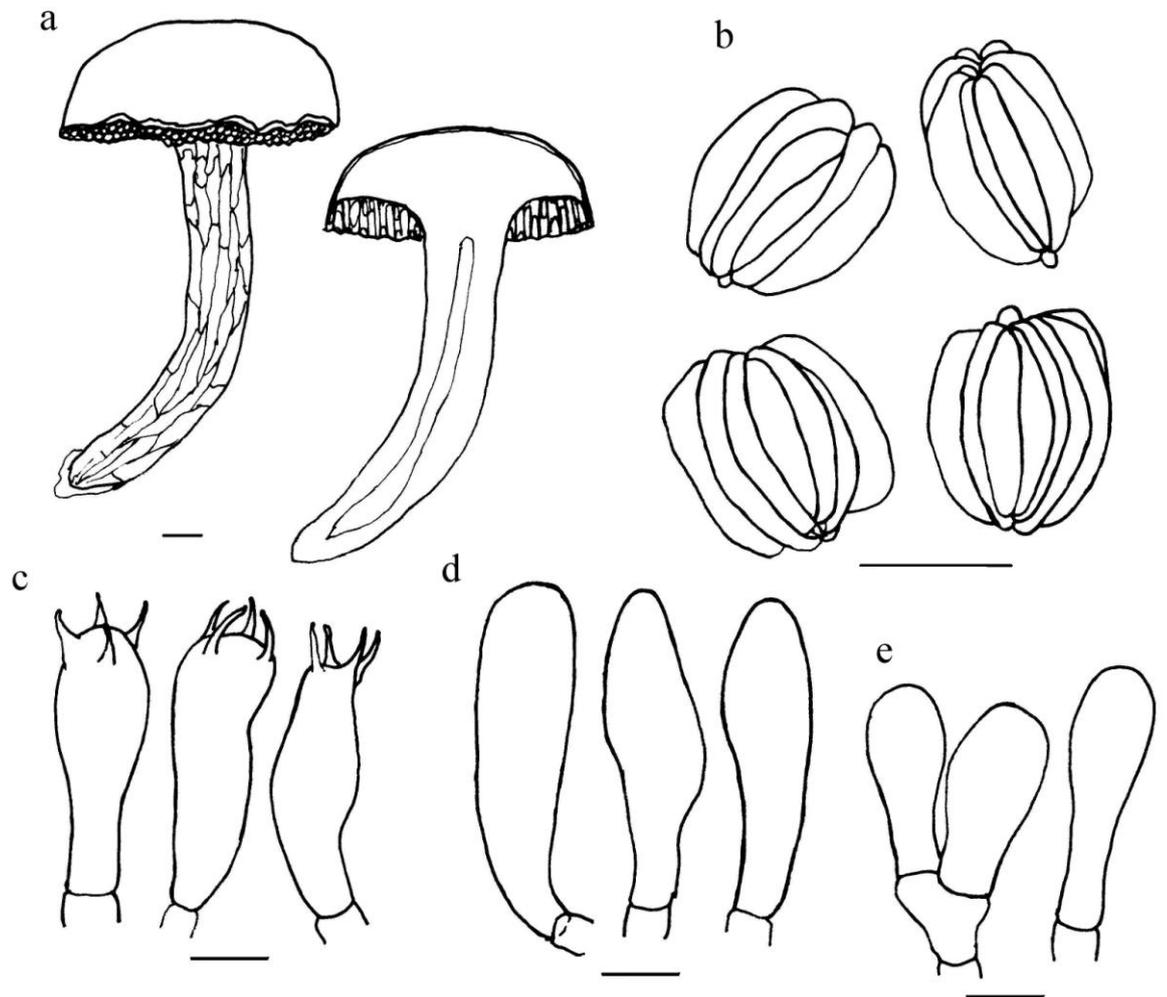
*Basidiospores* black (10-) 11-14 (-15) × (-8) 9 - 10 μm, [ $x_m = 12.45 \pm 1.12 \times 9.4 \pm 0.69$  μm,  $Q = 1.1 - 1.67$ ,  $Q_m = 1.33 \pm 0.16$ ,  $n = 30$  spores per collection], dextrinoid in Melzer's Reagent, short ellipsoid, longitudinally and conspicuously winged with the wings nearly as broad as the spore body, with basal thickened rim around the sterigmatal appendage. *Hymenophoral trama* parallel, with hyaline to slightly pale black elements, inamyloid. *Basidia* (20-) 25 - 34 × 8 - 10 μm, 4-sterigmate, clavate, hyaline, inamyloid. *Hymenophoral cystidia* 26.2 - 40.8 (-48) × (5.8-) 6.4 - 8.8 (-10.4) μm, clavate, thin-walled, hyaline, inamyloid. *Pileipellis* an ixotrichodermium, 60 - 80 μm thick, with branched hyphae, dextrinoid; *pileocystidia* clavate, 32 - 45 × 6 - 10 μm, dextrinoid. *Stipitipellis* a hymeniform with 60 - 80 μm thick; *caulocystidia* long ventricose, 54 - 70 (-80) × (8.8-) 12 - 16 μm, thin-walled, dextrinoid. *Clamps connection* absent.

*Habit, habitat, and distribution:* Solitary in soil. Known only from a single collection.

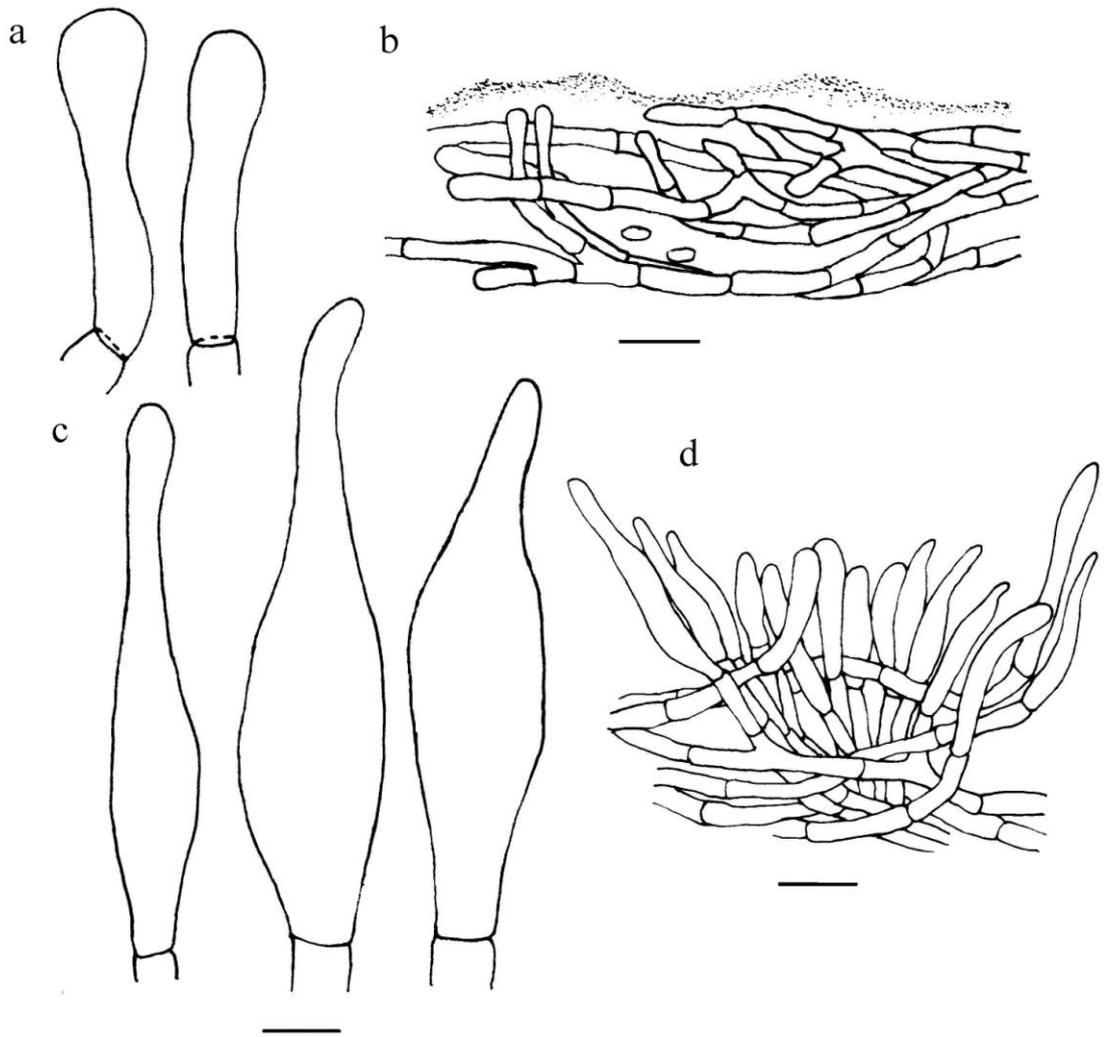
*Material examined:* MALAYSIA. Kedah, Langkawi, Kampung Buku, Gunung Raya, N 6°21'51.9", E 99°47'25.8", 6 April 2004, *leg. Tan Y.S.* (KLU-M1091).

*Commentary:* the genus *Afroboletus* has so far only been found in equatorial Africa and in many ways the African material is similar to *Strobilomyces* (Pegler and

Young, 1981). This single specimen of *Afroboletus malaysianus* first found in northern Peninsular Malaysia extends the distribution of the genus outside the African continent. This new distribution needs serious attention with regard to biogeography of the Boletineae. This specimen with black, ornamented spores with widely spaced longitudinal costae and basal thickened rim is not found in *Strobilomyces* and *Boletellus*.



**Figure 4.12a:** *Afroboletus malaysianus* (KLU-M1091). **a.** Basidiomes. **b.** Basidiospores. **c.** Basidia. **d.** Hymenial cystidia. **e.** Basidiales. Bars: a = 10 mm. b – e = 10  $\mu$ m.



**Figure 4.12b:** *Afroboletus malaysianus* (KLU-M1091). **a.** Pileocystidia. **b.** Pileipellis. **c.** Caulocystidia. **d.** Stipitipellis. Bars: a & c = 10  $\mu\text{m}$ . b & d = 20  $\mu\text{m}$ .

13. *Boletellus corneri* Klofac & Krisai, Ö st. f. Pilzk. 1: 32. 1992. (Fig. 4.13, Plate 4.3 I)

≡ *Boletus fallax* Corner, *Boletus* in Malaysia, p. 99. 1972.

≡ *Boletellus fallax* (Corner) Watling, Not. Roy. Bot. Gard. Edinburgh 46: 407. 1990,  
*non* Singer (1983).

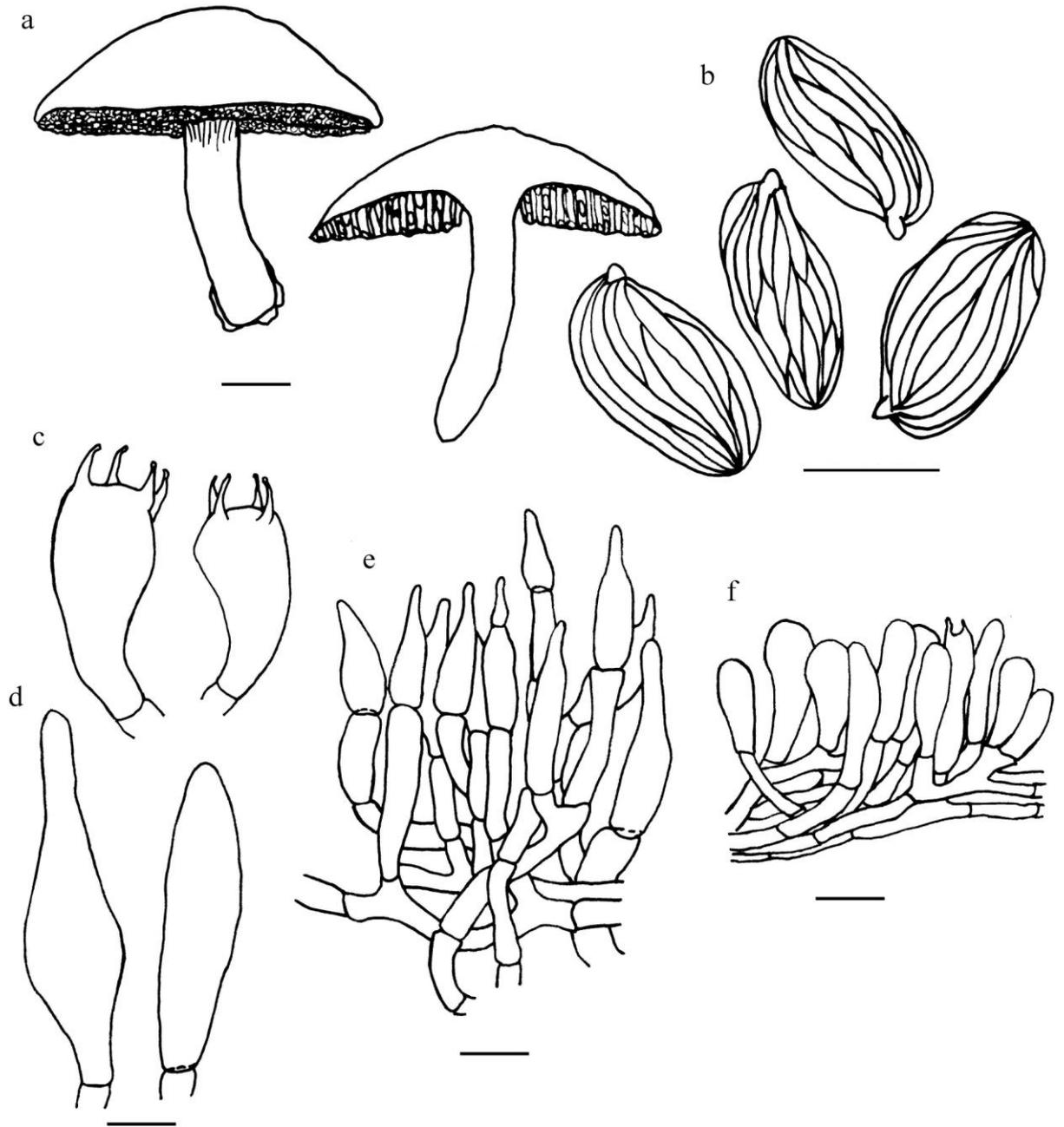
*Pileus* 70 – 107 mm broad, convex, dry, smooth to velvety with straight and smooth margin, brown; context bruising blue when exposed. *Hymenophore* tubulose, adnexed to free, yellow (3A7), with pores more than 1 mm broad, bruising blue when injured. *Stipe* 60 × 15 mm, central equal, cylindrical, dry, granular, yellow (3A7) at apex, reddish brown (8D7) at base, no colour changed when bruised, with white basal mycelium; interior yellowish white with solid stipe.

*Basidiospores* 12.1 – 15.1 (-16.4) × 6.1 – 8.1 μm [  $x_m = 14.04 \pm 0.85 \times 7.2 \pm 0.49$  μm,  $Q = 1.70 - 2.78$ ,  $Q_m = 1.94 \pm 0.11$ ,  $n = 30$  spores per one collection], finely longitudinally winged, subfusoid to elongated boletoid, inamyloid. *Basidia* 27.1 – 35 × 11 – 12 μm, 4-sterigmate, clavate, hyaline, inamyloid. *Hymenial cystidia* 35.0 – 40.0 × 6.8 – 8.2 (-10.4) μm, subclavate to subventricose, thin-walled, hyaline, inamyloid. *Pileipellis* a hymeniform structure, forming a long pile of hyphae; the end cells 26 – 45.6 × (7.7-) 10.0 – 17.3 μm, ventricose, thin-walled, hyaline, inamyloid. *Stipitipellis* with scattered clavate cystidia and sterile basidia; *caulocystidia* (23.8-) 25.8 – 35.6 × (8.5-) 10.2 – 13.9 (-16.3) μm, clavate to subclavate, hyaline, inamyloid. *Clamp connections* absent.

*Habit, habitat, and distribution:* solitary on soil in lowland Dipterocarp forests. Reported from Peninsular Malaysia and Singapore.

*Material examined:* MALAYSIA. Kedah. Langkawi Island, N 06°25'36.0", E 99°45'00.6", 200 m, 15 March 2003, leg. Tan Y.S. (KLU-M1092).

*Commentary* : This single specimen of *Boletellus* was first reported by Corner (1972) as *Boletus fallax* found in Western Peninsular Malaysia. This single specimen was collected in Northern of Peninsular Malaysia. The new combination by Watling (1990) is superseded by a previous one (Singer 1983) for a Murrill taxon from Florida, USA. Klofac and Krisai (1992) provided the new name for Corner's distinctive fungus from Malaysia.



**Figure 4.13:** *Boletellus corneri* Klofac & Krisai (KLU-M1092). **a.** Basidiomes **b.** Basidiospores. **c.** Basidia. **d.** Hymenial cystidia. **e.** Pileipellis. **f.** Stipitipellis. Bars: a = 20 mm. b – d = 10  $\mu$ m. e – f = 20  $\mu$ m.

14. *Boletellus emodensis* (Berk.) Singer, Ann. Mycol. 40: 19. 1942.

(Fig. 4.14, Plate 4.2 H)

≡ *Boletus emodensis* Berk. Fung. Hook. tab. TCCCLXX, Hook Journ. 1851, pg. 48.

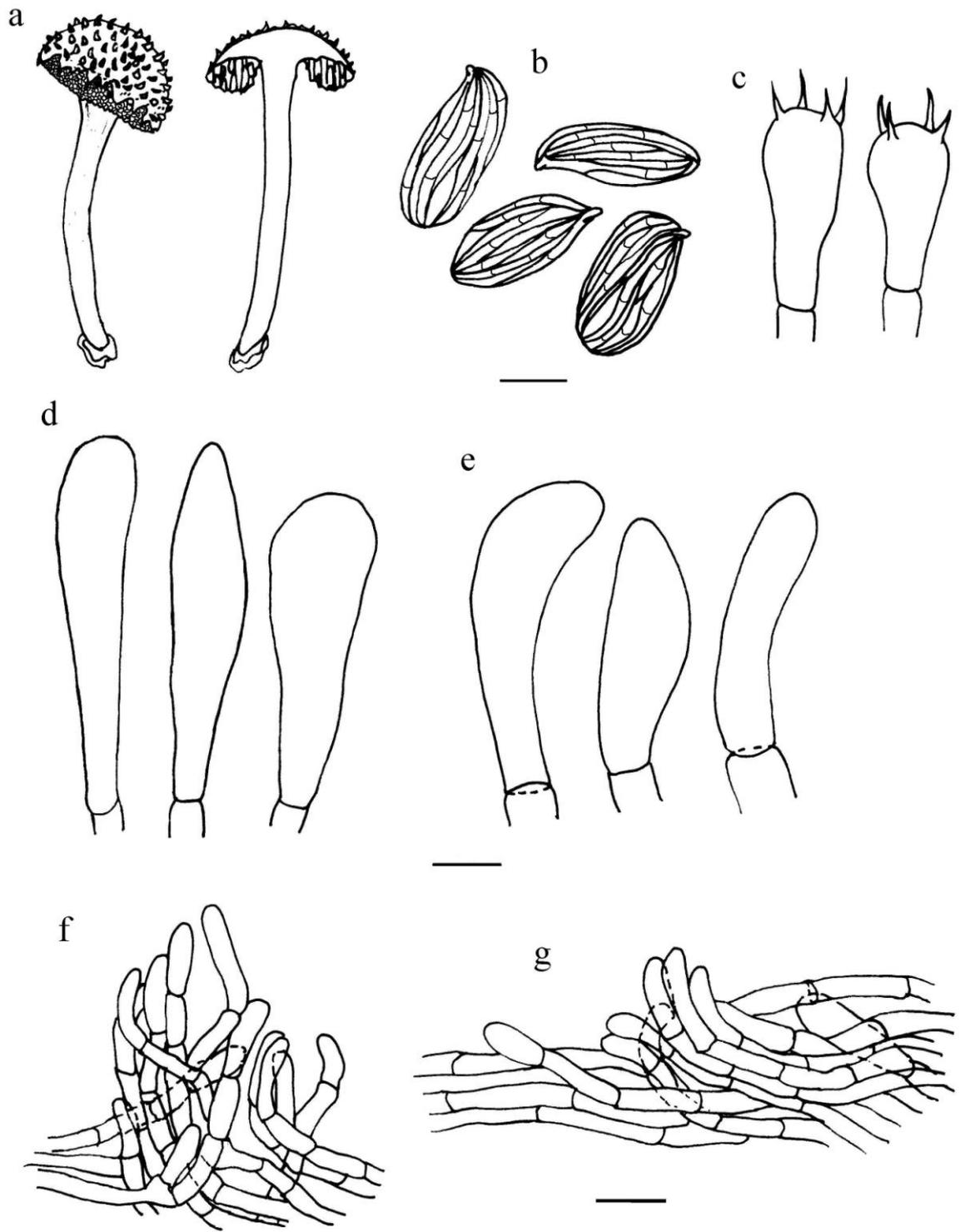
*Pileus* 25- 55 mm broad, convex to plano-convex, dry, scaly red to violet red (10C8, 10F6), decurved margin with appendiculate edge, yellow with NH<sub>4</sub>OH. *Context* 5 - 10 mm thick, yellowish white (2A2), discoloring blue, yellow with NH<sub>4</sub>OH. *Hymenophore* tubulose, 4 - 10 mm thick, adnexed to deeply depressed to free; *tubes* yellow (4A8) to olive yellow (2C7), blue when injured; *pores* 3 - 4 per 0.5 cm, concolorous with tubes, bruising blue to dark blue. *Stipe* 37- 78 mm long, 4 - 10 mm broad, equal to slightly tapering downward, central, dry, fibrillose, red (11C7) to deep violet brown (11F4), with interior solid and rigid, yellowish white (2A2), discoloring blue; *basal mycelium* white.

*Basidiospores* (18 -) 19 - 26 (-30) × (8-) 9 - 11 (-12) μm [  $x_m = 22.33 \pm 4.10 \times 10.13 \pm 0.86$  μm,  $x_{mr} = 22.33 - 23.94 \times 9.08 - 11.52$  μm,  $x_{mm} = 23.18 \pm 0.67 \times 10.13 \pm 1.06$  μm,  $Q = 1.90 - 2.78$ ,  $Q_{mr} = 1.79 - 2.65$ ,  $Q_{mm} = 2.28 \pm 0.24$ ,  $n = 130$  spores per 5 collections], longitudinally winged with cross striate, subfusoid, inamyloid. *Basidia* (21-) 25 - 32 × (9-) 10 - 12 (-13) μm, 4-sterigmate, clavate, hyaline, inamyloid. *Hymenophoral trama* parallel, inamyloid. *Hymenial cystidia* (32-) 35 - 44 (-50) × 8 - 12 (-14) μm, cylindrical to subclavate to subventricose, thin-walled, inamyloid. *Pileipellis* a palisade trichodermium, about 100 μm in height, with branched element, inamyloid. *Stipitipellis* a trichodermium of caulocystidia (34-) 38 - 43 (-46) × 9 - 12 (-14) μm, cylindric to subclavate, smooth, thin-walled, inamyloid. *Clamp connections* absent.

*Habit, habitat, and distribution:* solitary on soil in lowland Dipterocarp forests and highland oak tree forests (*Lithocarpus* sp.), common species in tropical forest worldwide, reported from Peninsular Malaysia, Borneo and Singapore.

*Material examined:* MALAYSIA. Negeri Sembilan, Pasoh, Hutan Simpan, N 2°58'7", E 102°17'49", 111 m, 20 September 2006, *leg. Chan H.T.* (KLU-M1093); Hutan Lipur Ulu Bendul, N 02°43'39.3", E 102°04'38.2", 170m, 26 May 2007, *leg. Chan H.T.* (KLU-M1094); Pahang, Fraser's Hill, Jeriau waterfall road, N 03°43'36.0", E 101°43'00.6", 970m, 30 May 2007, *leg. Chan H.T.* (KLU-M1095); Johor, Selai, Endau Rompin National Park, Takah Tinggi trail, N 02°25'59.4", E 103°14'49.5", 120m, 16 February 2006, *leg. Chan H.T.* (KLU-M1096); Selangor, Ulu Gombak forest area, N 03°15'01.4", E 101°44'59.9", 7 April 2005, *leg. Chan H.T. & Sumaiyah A.* (KLU-M1097).

*Commentary :* This very shaggy fungus appears either solitary or in twos on tree root or shadow places. It is very common at certain times of the year. It grows on either lowland or highlands forests. Pores are yellow, and stain instantly blue-green on bruising. The marginal veil eventually forms shaggy fragments with maturity. There are several species close to *Boletus emodensis*, but none has the minute cross striations on the spores. *Boletus ananiceps* is similar to *Boletellus emodensis*, but tends to be scaly and is wholly wine pink tinted. According to Corner (1975), *Boletellus ananas* may represent old and discoloured specimens of *Boletellus emodensis*. Certainly, this species may discolour with age, but *Boletellus ananas* has pale fruit-body from the outset.



**Figure 4.14:** *Boletellus emodensis* (Berk.) Singer (KLU-M1093). **a.** Basidiomes **b.** Basidiospores. **c.** Basidia. **d.** Hymenial cystidia. **e.** Caulocystidia **f.** Pileipellis. **g.** Stipitipellis. Bars: a = 20 mm. b – e = 10  $\mu$ m. f – g = 20  $\mu$ m.

15. *Xanthoconium violaceofuscus* (Chiu) Chan H.T. & Halling, *comb. prov.*

(Fig. 4.15, Plate 4.2 E)

= *Boletus violaceofuscus* Chiu, *Mycologia* 40: 210. 1948.

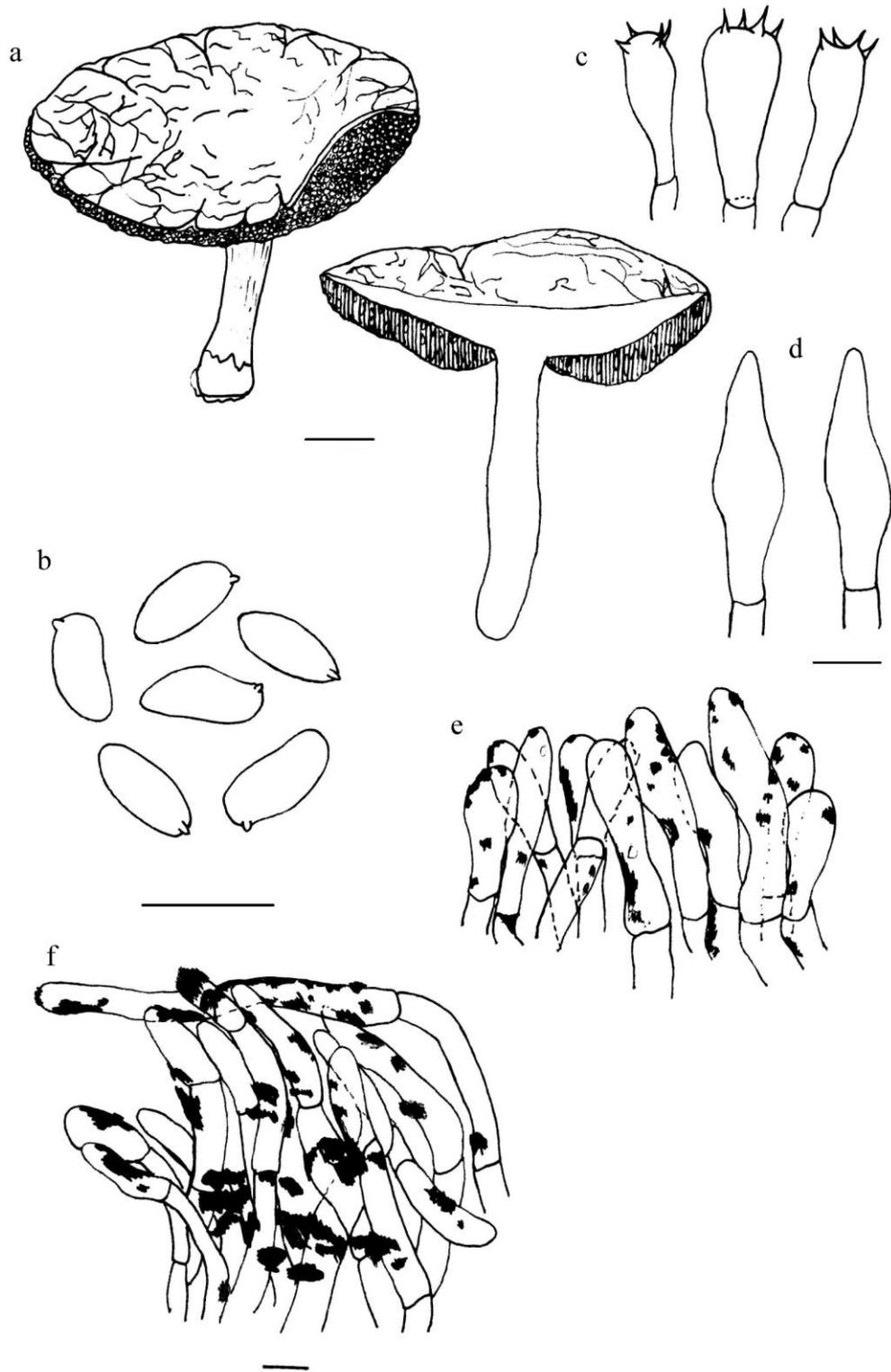
*Pileus* 120 mm broad, broadly convex, dry and scaly surface; dark violet brown (11F6, 11F7) scale with pale yellow background, matured fruiting body, straight margin with even edge, eroded when matured, no colour changed when exposed. *Context* thick with soft and spongy flesh, white in colour, no colour changed when exposed, blue green in NH<sub>4</sub>OH. *Hymenophore* tubulose, 4 - 6 mm thick, yellow (3A7, 3B7), free, slowly brown when exposed; *pores* concolorous with tubes; fine pores with 2 - 3 pores per mm, brown when bruised. *Stipe* 11 × 9 cm broad, equal and cylindrical to slightly bulbous, central, dry, broadly reticulate at apex, fibrillose at base, violet brown (11F7) at apex, dark violet brown (12F8) at base, interior solid, context white, blue green with NH<sub>4</sub>OH; *basal mycelium* white.

*Basidiospores* (7-) 8 - 11 × 4 - 5 μm, [ $x_m = 9.53 \pm 1.12 \times 4.65 \pm 0.40 \mu\text{m}$ ,  $Q = (-1.4) 1.78 - 2.25$ ,  $Q_m = 2.06 \pm 0.30$ ,  $n = 20$  spores per collection] smooth, ovoid to ellipsoid, olive brown, inamyloid. *Basidia* 23 - 29 × 7 - 11 μm, 4 sterigmata with 2 - 3 μm length, clavate, hyaline, inamyloid. *Hymenophoral trama* with cells 7 - 16 μm in broad, divergent with hyaline elements, inamyloid. *Hymenial cystidia* (31-) 39 - 43 μm × 9 - 12 μm, subventricose to ventricose, thin-walled, inamyloid. *Pileipellis* a hymeniform structure, with black pigment randomly spread around the hyphae, the end cells 7 - 10 μm broad, thin-walled, inamyloid. *Stipitipellis* with scattered clavate cystidia in hymeniform arrangement; black pigment present as in pileipellis. *Caulocystidia* 20 - 41 × 6 - 4 μm, clavate to subventricose, dextrinoid. *Clamp connections* absent.

*Habit, habitat, and distribution:* solitary on soil in lowland dipterocarp forest. (Dipterocarpaceae); reported from Southern China.

*Material examined:* MALAYSIA. Negeri Sembilan, Ulu Bendul Forest Reserve. N 02°43'39.3", E 102°04'38.2", 170m, 17 February 2004, *leg. Tan Y.S., Chan H.T.* (KLU-M1098).

*Commentary:* This is a first report of the genus in Malaysia. The species was reported as *Boletus violaceofuscus* by Chiu in 1948 from China. The hymeniform elements in the pileipellis and stipitipellis along with a color change to blue-green with ammonia distinguishes it from *Boletus*. The genus is also primarily distinguished by the bright yellow brown color of the spore deposit.



**Figure 4.15:** *Xanthoconium violaceofuscus* (Chiu) Chan H.T. & Halling, *comb. prov.* (KLU-M1098). **a.** Basidiomes **b.** Basidiospores. **c.** Basidia. **d.** Hymenial cystidia. **e.** Claulocystidia. **f.** Pileipellis. Bars: a = 20 mm. b = 20  $\mu$ m. c – f = 10  $\mu$ m.



**Plate 4.2:** A: *Afroboletus malaysianus* ( $\times 4.4$ ); B: Scanning electron microscopy of basidiospore ( $10\ \mu\text{m}$ ); C: *Pulveroboletus cf. viridis* ( $\times 1.7$ ); D: *Pulveroboletus viridisquamosus* ( $\times 4.9$ ); E: *Xanthoconium violaceofuscus* ( $\times 8.0$ ); F: *Pulveroboletus ridleyi* ( $\times 1.9$ ); G: *Pulveroboletus claroflavus* ( $\times 2.0$ ); H: *Boletellus emodensis* ( $\times 3.6$ ).

**Bar = 10mm**

16. *Aureoboletus thibetanus* (Pat.) Hongo & Nagasawa Rep. Tottori Mycol. Inst. 18: 133.  
1980. (Fig. 4.16, Plate 4.3 D)

≡ *Boletus thibetanus* Pat., *Bull. Soc. mycol. Fr.* 11: 196 (1895).

≡ *Pulveroboletus thibetanus* (Pat.) Singer, *Agaric. mod. Tax.*, Edn 4 Koenigstein):  
774 (1986).

≡ *Suillus thibetanus* (Pat.) F.L. Tai, *Syll. fung. sinicorum*: 736 (1979).

*Pileus* 33 mm broad, convex, viscid, rugose, brown (5D7), with smooth margin.  
*Context* 5mm thick at disc, 3mm thick at margin, soft and spongy, slightly brownish white, no colour change when exposed, slightly yellowish with NH<sub>4</sub>OH. *Hymenophore* tubulose, 4 mm thick, free. *Tubes* yellow (2A8), no staining when injured, becoming blue with NH<sub>4</sub>OH, eight pores per mm, pores concolorous with tubes. *Stipe* 45 cm long, 5 mm broad at apex, 7 mm broad at base, subclavate, central, dry, fibrillose, brown (5D7), interior solid, white, no colour change when exposed or with NH<sub>4</sub>OH; *basal mycelium* white.

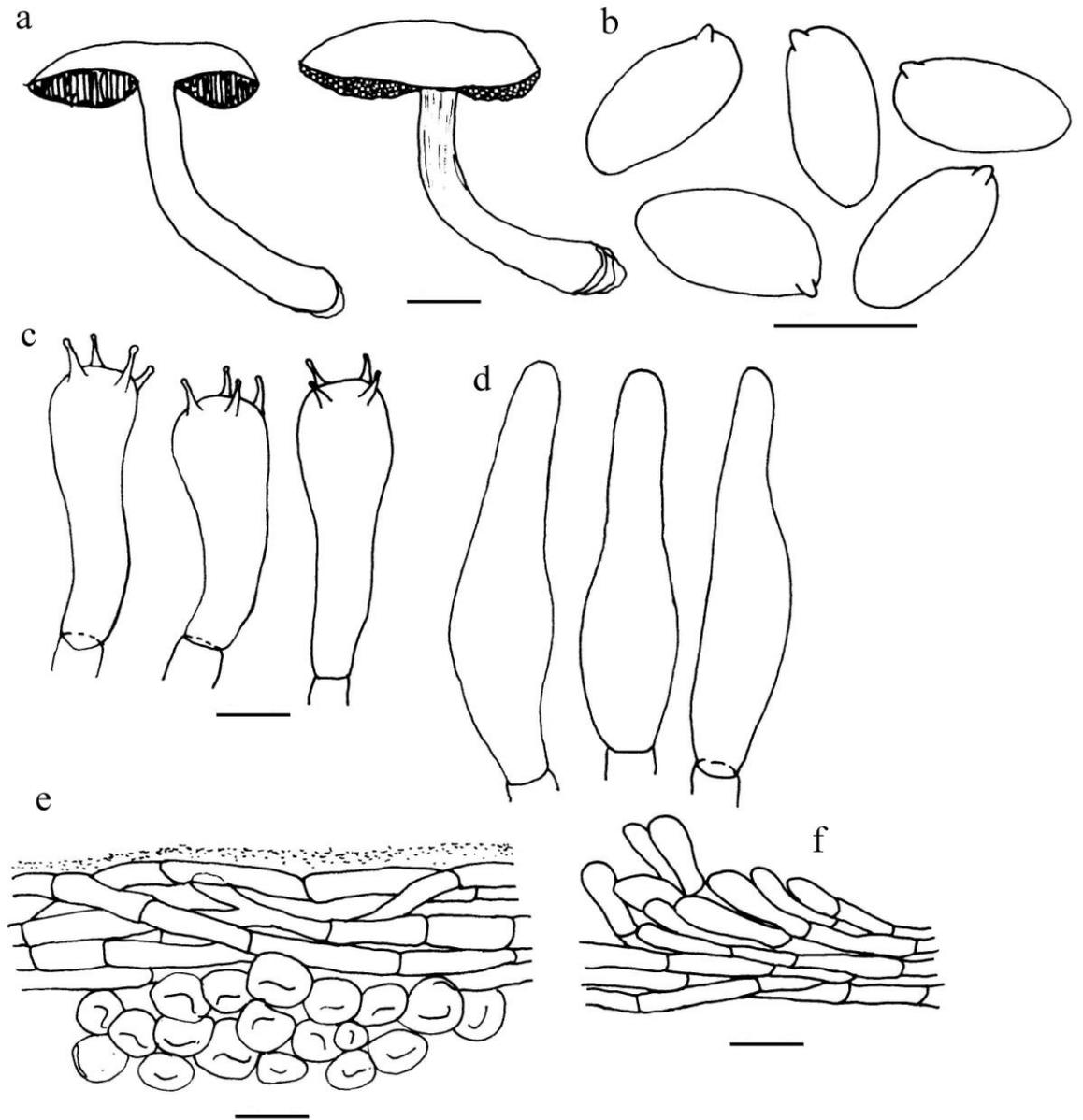
*Basidiospores* olive yellow,  $9.6 - 11.2 \times (4.8-) 5.2 - 5.6 (-6) \mu\text{m}$ , [ $x_m = 10.27 \pm 0.49 \times 5.47 \pm 0.28 \mu\text{m}$ ,  $Q = 1.71 - 2.0$ ,  $Q_m = 1.88 \pm 0.11$ ,  $n = 25$  spores per one collection] smooth, boletoid to ellipsoid to subfusiform, inamyloid. *Basidia*  $25.6 - 37.6 \times (8.0-) 9.6 - 12.0 \mu\text{m}$ , 4-sterigmate, long clavate, hyaline in KOH, inamyloid. *Basidiales*  $22.4 - 28.0 \times 8.0 - 9.6 \mu\text{m}$ , clavate, branching and septate hyphae, hyaline, inamyloid. *Hymenophoral trama* divergent, inamyloid. *Hymenial cystidia*  $45 - 64 \times 12 - 14 \mu\text{m}$ , subventricose, thin-walled, hyaline, inamyloid. *Pileipellis* an ixotrichodermium with inflated hyphae at the bottom,  $120 - 160 \mu\text{m}$  high, hyaline, dextrinoid. *Stipitipellis* with scattered clavate cystidia; *caulocystidia*  $(19.4-) 22 - 33 (-42) \times (7.5-) 10 - 12.5 \mu\text{m}$ , clavate, hyaline, inamyloid. *Clamp connections* absent.

*Habit, habitat, and distribution:* solitary on soil in highland forest (*Lithocarpus* sp.). Reported from Yunnan, China, Singapore and Japan.

*Material examined:* MALAYSIA. Pahang, Fraser Hill, Jeriau Waterfall Road, N 03°43'36.0", E 101°43'00.6", 960 m, 8 January 2006, *leg. Chan H.T.* (KLU-M1099).

*Commentary:* This single collection was collected under a *Lithocarpus* sp. and could possibly be associated with it. Originally described from Tibet by Patouillard (1895). This species were re-collected from Yunnan (Chiu, 1948), Singapore (Corner, 1972) and Sarawak (Corner, 1974) and known as *Boletus thibetanus*. Whereas Singer (1986) places this species in *Pulveroboletus*. According to Corner (1972), he believed that *Boletus flexipes* Masee collected from Singapore were the same fungus. However, this is only based on the illustration of Ridley's but no type material exists in Kew. Watling and Hollands (1990) re-collected this species from Sarawak. Recently, Yang *et al.* (2003) studied the type and additional materials of *Boletus thibetanus* from Yunnan.

This collection agrees with the description by Chiu from Yunnan and the size of the pileus is wide ranging from 20 – 35 mm. Patouillard's specimens from Tibet were larger with pileus 4 – 5 mm wide. The viscid rugulose pileus, glabrous stipe with white base, and no colour change at tissues when bruised point towards this taxon. Unfortunately, this collection consisted only of an old solitary basidiocarp. The pileipellis were observed with corrupted pleurocystidia whereas Yang *et al.* (2003) observed an ixotrichodermium of vertically arranged cystidia.



**Figure 4.16:** *Aureoboletus thibetanus* (Pat.) Hongo & Nagasawa (KLU-M1099). **a.** Basidiomes **b.** Basidiospores. **c.** Basidia. **d.** Hymenial cystidia. **e.** Pileipellis. **f.** Stipitipellis. Scale bars: a = 10 mm. b – d = 10  $\mu$ m. e – f = 20  $\mu$ m.

17. *Boletus patouillardii* Singer, Amer. Midl. Nat. 37: 55. 1947. (Fig. 4.17, Plate 4.4 F)

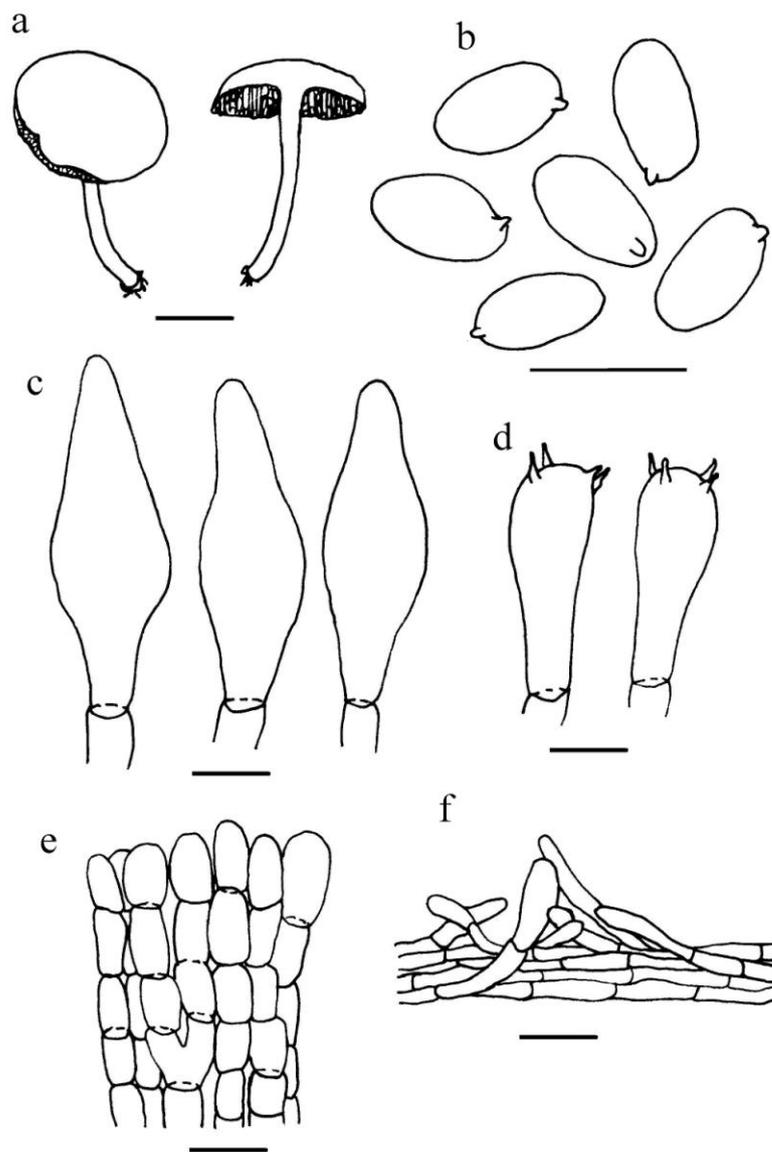
*Pileus* 6 – 10 mm broad, convex, dry, smooth, dark red to violet red (10E8), with straight and even margin, yellow with NH<sub>4</sub>OH. *Context* 1.0 mm thick; *flesh* soft, white (2A1), blue when exposed, yellow with NH<sub>4</sub>OH. *Hymenophore* tubulose, 1.0 – 1.5 mm thick, adnate, yellow (1A8), blue when bruised, orange with NH<sub>4</sub>OH; *pores* 7 – 10 pores per 1 mm, concolorous with tubes, bruising blue. *Stipe* 5 – 10 × 1 mm, flexuous and tapering downwards, central to slightly eccentric, dry, smooth, yellow at apex (3A5), red (10E8) at base, with interior solid, blue when exposed; *basal mycelium* white.

*Basidiospores* 8 – 10 × 5 – 6 μm [  $x_m = 9.02 \pm 0.73 \times 5.43 \pm 0.47$  μm,  $x_{mr} = 8.60 - 9.02 \times 5.43 - 5.50$  μm,  $x_{mm} = 8.81 \pm 0.30 \times 5.47 \pm 0.05$  μm,  $Q = 1.33 - 2.00$ ,  $Q_m = 1.67 \pm 0.20$ ,  $Q_{mr} = 1.57 - 1.67$ ,  $Q_{mm} = 1.62 \pm 0.07$ ,  $n = 40$  spores per two collections], smooth, ellipsoid, inamyloid. *Basidia* 26 – 30 × 9 – 11 μm, 4-sterigmate, long subclavate, hyaline in KOH, inamyloid. *Hymenial cystidia* 75 – 80 × 9 – 12 μm, long cylindrical to long subventricose, smooth, thin-walled, hyaline, inamyloid. *Hymenophoral trama* subparallel to subdivergent, with hyaline elements, inamyloid. *Pileipellis* a structure of moniliform hyphae, > 80 μm, forming a pile of end cells 7 – 12 × 5 – 6 μm, thin-walled, hyaline in KOH, inamyloid. *Stipitipellis* almost smooth with a slight trichodermium of scattered cylindrical to subclavate cystidia, thin-walled, hyaline, inamyloid. *Clamp connections* absent.

*Habit, habitat, and distribution:* gregarious on soil of cut road banks in forests of Dipterocarpaceae; reported from Peninsular Malaysia.

*Material examined:* MALAYSIA. Selangor, Hulu Langat, Forest Reserve Sungai Chongkak, N 03°12'00.3", E 101°50'07.4", 149 m, 6 January 2006, *leg. Chan H.T.* (KLU-M1100); Ulu Gombak, Biodiversity Research Center Ulu Gombak, N 03°19'28.7", E 101°50'35.3", 261 m; 7 April 2005. *leg. Chan H.T.* (KLU-M1101).

*Commentary:* This very small bolete is usually found growing on soil of cut road banks. The dry red pileus and yellow hymenophore that stains blue makes it easy to be recognized. Singer suggested that this species was allied with *B. pernanus* Pat. et. Bak. and resembled *Boletus coccineinanus* Corner. Unfortunately, *B. coccineinanus* has pink pores and larger spores (9.5 – 11.5 x 4.5 – 5.2  $\mu\text{m}$ ) but this species has yellow pores and smaller spores which fit Singer's description.



**Figure 4.17:** *Boletus patouillardii* Singer (KLU-M1100). **a.** Basidiomes. **b.** Basidiospores. **c.** Hymenial cystidia. **d.** Basidia. **e.** Pileipellis. **f.** Stipitipellis. Bars: a = 5 mm. b – d = 10  $\mu\text{m}$ . e – f = 20  $\mu\text{m}$ .

18. *Boletus cf. craspedius* Masee, Kew Bull. 11:76. 1914. (Fig. 4.18, Plate 4.3 G)

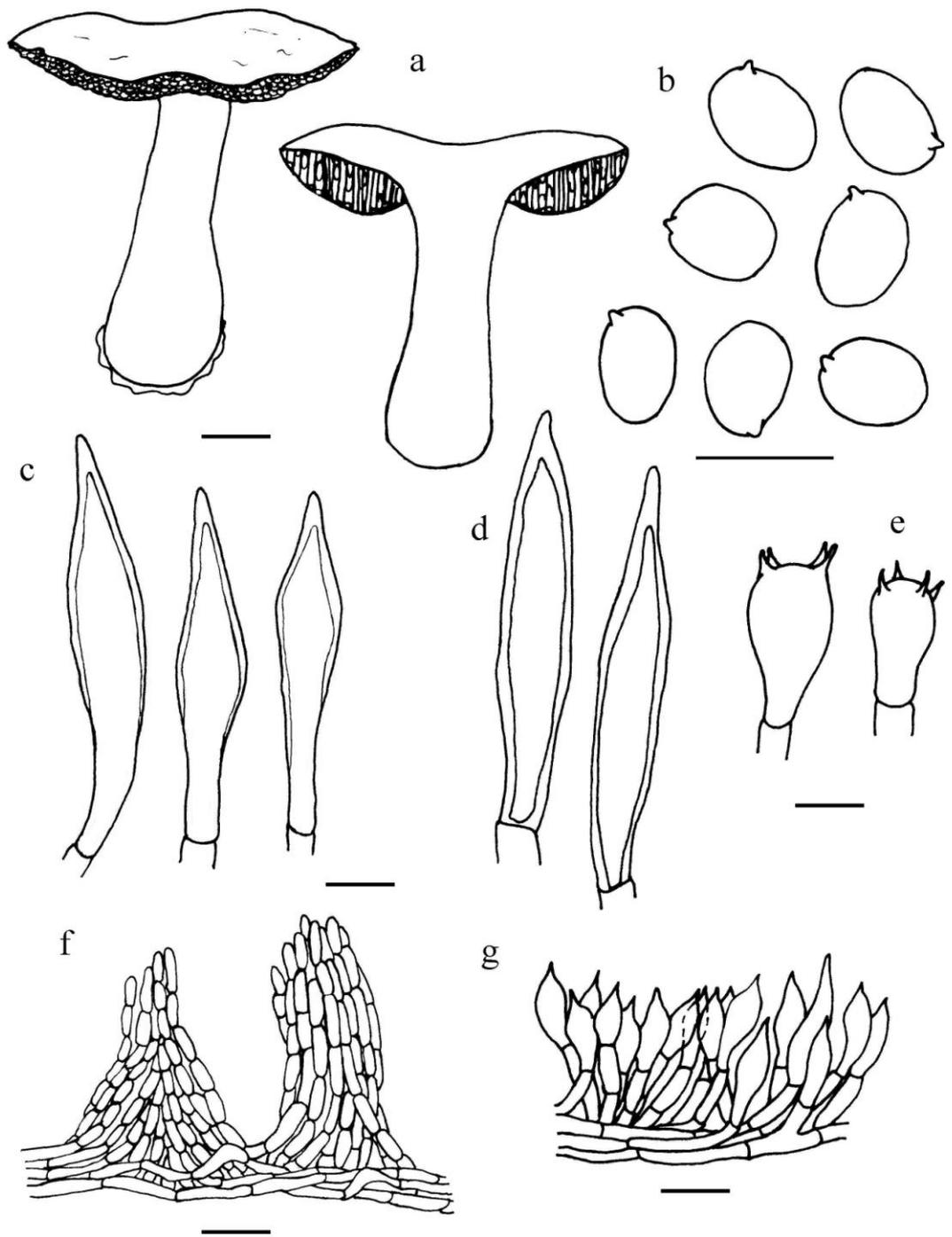
*Pileus* 50 mm broad, convex to broadly depressed, dry and velvety, with even to slightly wavy margin, red when young (10B7), orange yellow (5A7) when mature. *Context* 5 – 10 mm thick, solid, yellowish white, immediately blue when exposed. *Hymenophore* 2 mm thick, tubulose, adnexed to almost free, reddish yellow (4A7), deeply cyanescent; *pores* dark red (9C5) with 3 – 4 pores per mm, deeply cyanescent. *Stipe* 45 × 15 µm, central, subclavate to clavate, dry, fine ventricose from apex to middle, base smooth, orange yellow (5A7) at apex, reddish orange at base (7A8) with interior solid, yellowish white, blue when exposed, with white basal mycelium. *Taste* not distinctive.

*Basidiospores* 6 – 7.6 × 4.6 – 5.5 µm [ $x_m = 6.83 \pm 0.43 \times 5.05 \pm 0.23$  µm,  $Q = 1.20 - 1.45$  (-1.55),  $Q_m = 1.35 \pm 0.09$ ,  $n = 35$  spores per one collection], globose to subglobose, smooth, olivaceous, inamyloid. *Basidia* 14.4 – 22.4 × 4.8 – 9.6 µm, 4-sterigmate, short clavate, hyaline, inamyloid. *Cheilocystidia* (29.5-) 39.2 – 52.3 × 6.0 – 8.8 (-9.6) µm, subventricose to ventricose to digitate, thick-walled at apex and gradually thin-walled towards base, olivaceous in KOH, deeply dextrinoid at apex in Melzer's Reagent. *Pleurocystidia* 48.3 – 52.9 × 7.7 – 9.7 µm, same as cheilocystidia but olivaceous and thick-walled the whole length. *Hymenophoral trama* divergent, with yellow brown elements, inamyloid. *Pileipellis* a trichoderminal palisade with short compact hyphae forming a pile, 80 – 100 µm high; *pileocystidia* 8 – 10 × 2 – 4 µm, short cylindrical, hyaline, inamyloid. *Stipitipellis* a hymenium of caulocystidia 40 – 50 µm high; *caulocystidia* 21.3 – 33.2 (-36.1) × 6.3 – 8.4 (-10.0) µm, smooth, short ventricose-fusiform, thin-walled, hyaline, inamyloid. *Clamp connections* absent.

*Habit, habitat, and distribution:* solitary to gregarious on soil in lowland Dipterocarp forest of Northern Peninsular Malaysia; reported from Singapore.

*Material examined:* MALAYSIA. Kedah. Langkawi Island, Datai, N 06°25'22.0", E 99°40'30.1", 108 m, 15 March 2003, *leg. Tan Y.S.* (KLU-M1102).

*Commentary:* The color illustration of this taxon indicates placement in subsection *Luridi* of *Boletus*. However, the inadequate notes on the fresh condition preclude a more precise determination. In Corner (1972), very few red-pored boletes are discussed preventing further comparison with species known to him.



**Figure 4.18:** *Boletus cf. craspedius* Masee (KLU-M1102). **a.** Basidiomes. **b.** Basidiospores. **c.** Cheilocystidia. **d.** Pleurocystidia. **e.** Basidia. **f.** Pileipellis. **g.** Stiptipellis. Bars: a = 10 mm. b – e = 10  $\mu$ m. f – g = 20  $\mu$ m.

19. *Boletus phaeocephalus* Pat. & C.F. Baker, J. Straits Branch Royal Asiatic Soc. 78:

70. 1918.

(Fig. 4.19, Plate 4.3 F)

*Pileus* 50 mm broad, convex, dry, smooth to velvety, brown to grayish brown (6D5), decurved margin with even edge when young, violet with NH<sub>4</sub>OH. *Context* 8 mm thick at disc, 2mm thick at margin; *flesh* white, discoloring absent, purple red with NH<sub>4</sub>OH. *Hymenophore* tubulose, 6.5 mm thick, adnate, yellowish white to pale white (1A2), discoloring absent; *pores* fine, 3 pores per mm, brown, purple red with NH<sub>4</sub>OH. *Stipe* 49 mm × 12 mm (apex), 9 mm (base), tapering downwards, central, dry, fibrillose, orange to orange brown (6D5); interior solid, discoloring absent. *Basal mycelium* white.

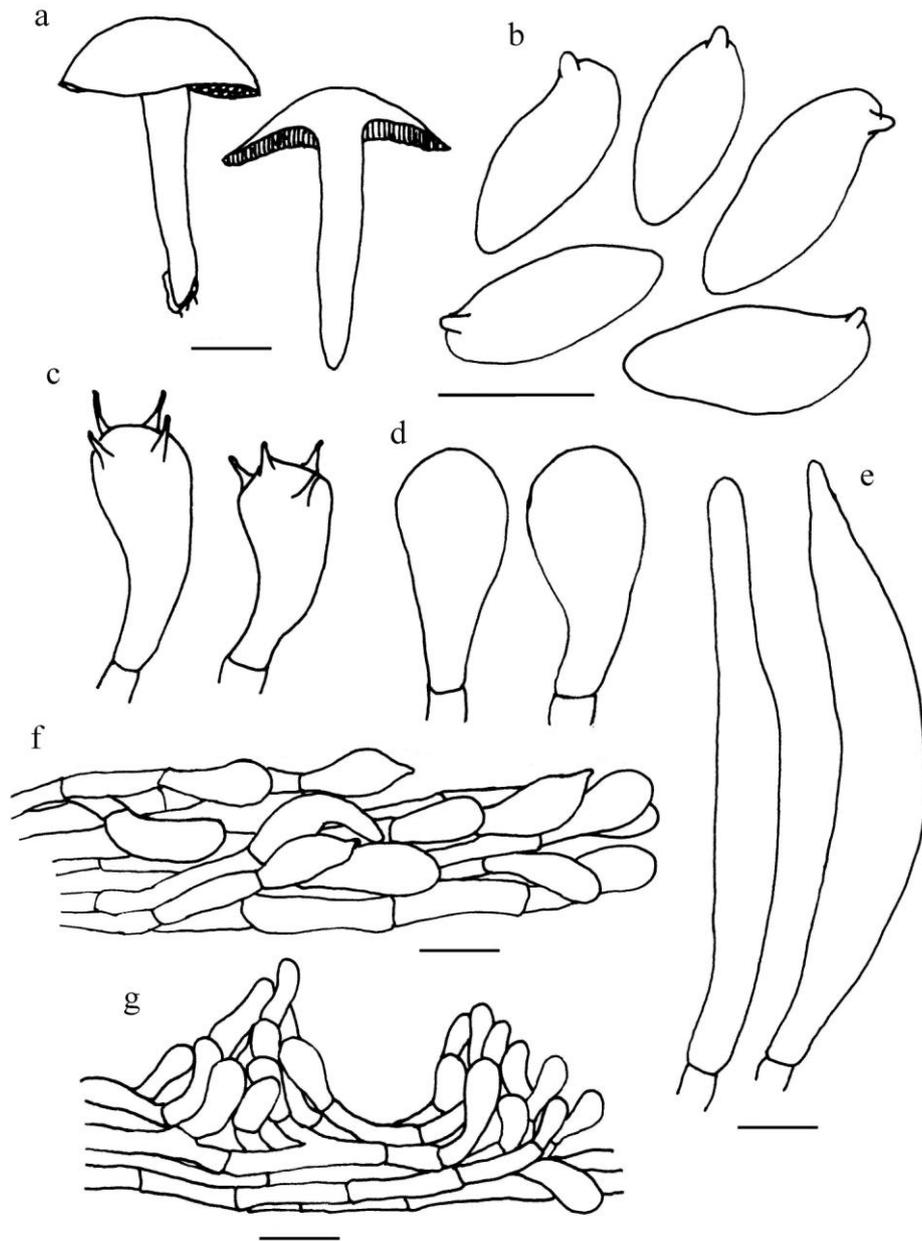
*Basidiospores* (11-) 12 – 17 × 5 – 7 (-9) μm [  $x_m = 14.94 \pm 1.49 \times 6.26 \pm 0.84$  μm,  $x_{mr} = 13.85 - 14.94 \times 5.34 - 6.26$  μm,  $x_{mm} = 14.40 \pm 0.77 \times 5.80 \pm 0.65$  μm,  $Q = 1.78 - 2.73$ ,  $Q_m = 2.40 \pm 0.21$ ,  $Q_{mr} = 1.78 - 3.2$ ,  $Q_{mm} = 2.51 \pm 0.16$ ,  $n = 50$  spores per two collections], subfusoid to ellipsoid, smooth, olive green, inamyloid. *Basidia* (20-) 26 – 36 × 10 – 15 μm, 4-sterigmate, clavate, hyaline in KOH, inamyloid. *Hymenial cystidia* 75 – 80 × 9 – 12 μm, cylindrical to subventricose, smooth, thin-walled, hyaline, inamyloid. *Hymenophoral trama* subparallel to subdivergent, with hyaline elements, inamyloid. *Pileipellis* a trichodermium of pileocystidia and corrupted hyphae, thin-walled, inamyloid. *Stipitipellis* covered by a trichodermium of cystidia, 40 – 50 μm in high, clavate to subventricose, smooth, thin-walled, inamyloid. *Clamp connections* absent.

*Habit, habitat, and distribution:* solitary on soil in lowland Dipterocarp forests; reported from Peninsular Malaysia.

*Material examined:* MALAYSIA. Negeri Sembilan, Hutan Simpan Pasoh, 2°58'7"N, 102°17'49"E, 111 m, 20 September 2006, leg. Chan H.T. (KLU-M1103). Kedah, Langkawi Island, Gunung Raya, Lubuk Semilang, N 06°21'58.4", E 99°47'29.6", 59 m, 8 April 2004, leg. Tan Y.S. (KLU-M1104).

*Commentary:* My material matches that described by Corner (1972) in micromorphology and most macromorphological features. His description of a white reticulum may be a transient feature however. Notable fresh characters include the dry, subtomentose pileus and concolorous stipe with non-cyanescent flesh and pores developing a pale brownish tint. This taxon may very well belong to the eduloid group as surmised by Corner (1972).

Based on the spore size and the macroscopic image, it is possible, but questionable, that KLU-M1103 may be this species, but the exposed flesh is pale orangish yellow and the overall macroscopic notes are not adequate.



**Figure 4.19:** *Boletus phaeocephalus* Pat. & C.F. Baker (KLU-M1103). **a.** Basidiomes. **b.** Basidiospores. **c.** Basidia. **d.** Basidiales. **e.** Hymenial cystidia. **f.** Pileipellis. **g.** Stipitipellis. Bars: a = 10 mm. b – e = 10  $\mu$ m. f – g = 20  $\mu$ m.

20. *Boletus peltatus* Corner & Watling in Watling, Edinburgh J. Bot. 50: 237. 1993.

(Fig. 4.20, Plate 4.3 E)

≡ *Boletus peltatus* Corner, Boletus in Malaysia, p. 136. 1972, *nom. nud.*

*Pileus* 66 – 134 mm broad, convex, dry, smooth to velvety; disc greyish brown (5F3), straight margin with even edge, turning orange with NH<sub>4</sub>OH. *Context* 10 – 17 mm thick, *flesh* soft and spongy, white when young, light yellow (2A4) when mature, discoloring absent, turning red with NH<sub>4</sub>OH. *Hymenophore* tubulose, 6 – 7 mm thick, free; *tubes* yellow (3B7), staining brown when injured, staining dark brown with NH<sub>4</sub>OH, two pores per one mm, concolorous with tubes. *Stipe* 80 – 140 × 8 – 23 mm broad at apex, 11 – 30 mm broad at base, clavate, central, dry, lacerate to lacunose, yellowish brown (5D5) to light brown, interior solid, context white, discoloring brown, bruising blue and slowly turning red with ammonia; *basal mycelium* white.

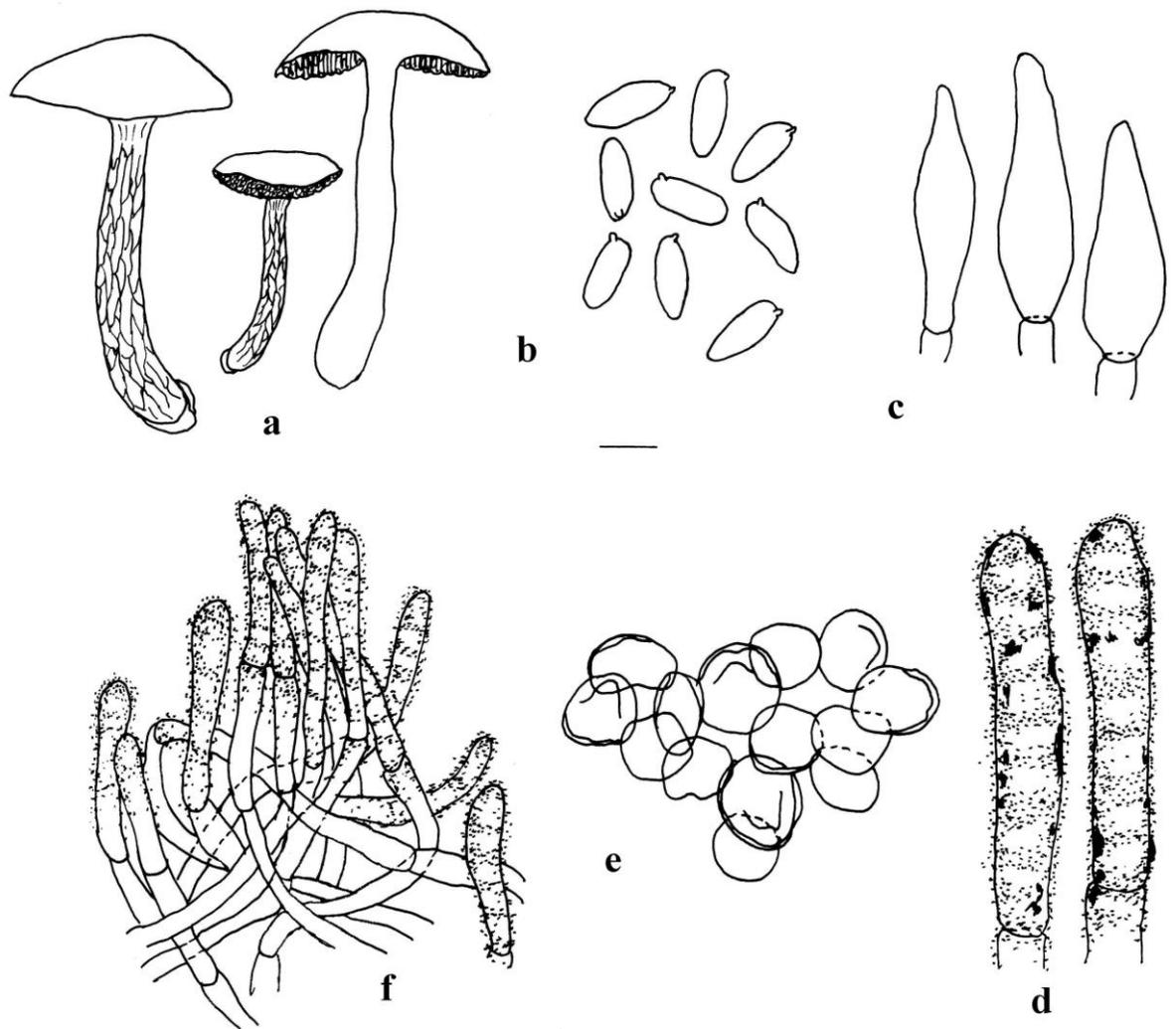
*Basidiospores* olive brown, (10-) 11 – 14 (-15) × 4 – 6 μm, [  $x_m = 12.37 \pm 1.1 \times 5.03 \pm 0.43 \mu\text{m}$ ,  $Q = 2.2 - 2.8$ ,  $Q_m = 2.461 \pm 0.16$ ,  $n = 30$  spores per collection] smooth, ellipsoid to subfusiform, inamyloid. *Hymenophoral trama* inamyloid. *Hymenophoral cystidia* 30 – 43 (-48) μm × (9-) 10 – 14 (-17) μm, ventricose, thin-walled, inamyloid. *Pileipellis* a trichodermium, with incrustated hyphae, (7-) 8 – 10 μm broad, disarticulating, oleiferous hyphae present, dextrinoid. *Stipitipellis* 24 – 40 (-45) × (20-) 25 – 30 (-34) μm; hyphae resembling sphaerocysts, oleiferous hyphae present, inamyloid. *Clamp connections* absent.

*Habit, habitat, and distribution:* gregarious on soil in forests of Dipterocarpaceae (*Shorea parvifolia*, *Calophyllum dioscurii*.); reported from peninsular Malaysia.

*Material examined:* MALAYSIA. Negeri Sembilan, Hutan Simpan Pasoh, 2°58'7"N, 102°17'49"E, 111 m, 20 September 2006, *leg. Chan H.T.* (KLU-M1105).

*Commentary:* First recognized and invalidly described by Corner (1972), this taxon was re-characterized and validly published by Watling (1993) and discussed again

by Watling *et al.* (2006). It is easily recognizable by the uniformly brown to light brown pileus and pale brown, lacerate to lacunose-reticulate stipe with white base. Such stipe ornamentation is reminiscent of *Boletellus russellii* Frost from North East USA, but the spores of the Malaysian taxon are smooth. Watling (1993) compares in detail other unrelated boletes (species of *Austroboletus* and *Boletus*) that have conspicuously ornamented stipes.



**Figure 4.20:** *Boletus peltatus* Corner & Watling in Watling (KLU-M1105). **a.** Basidiomes **b.** Basidiospores. **c.** Cheilocystidia. **d.** Pileocystidia. **e.** Claulocystidia. **f.** Pileipellis. Bars: a = 100 mm. b – d = 10  $\mu$ m. e – f = 25  $\mu$ m.

21. *Strobilomyces foveatus* Corner, *Boletus* in Malaysia, p. 60. 1972.

(Fig. 4.21, Plate 4.3 C)

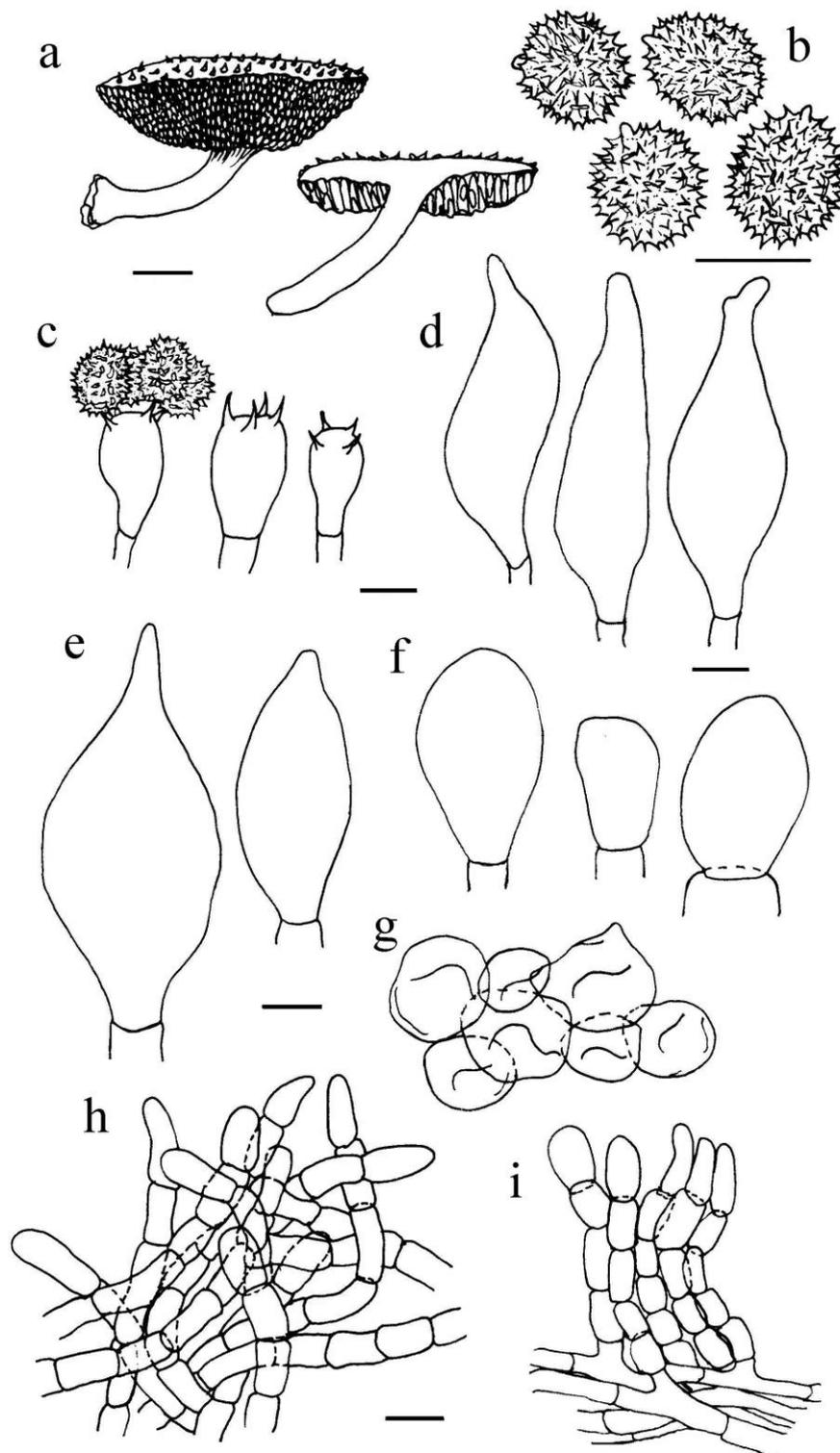
*Pileus* 50 mm broad, convex to plano-convex, dry, scaly surface; disc greyish black (5H1), with a straight margin and appendiculate veil remnants, negative reaction with  $\text{NH}_4\text{OH}$ . *Context* 3 - 4 mm thick, grey to blackish grey (20G1), discoloring black, negative reaction with  $\text{NH}_4\text{OH}$ . *Hymenophore* tubulose, 9 mm thick, adnate. *Pores* black (5H2); 3 pores per 5 mm at disc, 6 - 7 pores per 5 mm at margin, concolorous with tubes. *Stipe* 30 mm long, 5 mm broad, cylindrical and equal, central, dry, fibrillose, brownish black (6H8), with solid interior; *Basal mycelium* brownish black.

*Basidiospores* in fresh deposit dark brown to black, (7-)10 - 12 (-13)  $\times$  (6-) 9 - 10 (-11)  $\mu\text{m}$  [  $x_m = 10.75 \pm 1.58 \times 9.23 \pm 1.26 \mu\text{m}$ ,  $Q = 1.1 - 1.3$ ,  $Q_m = 1.17 \pm 0.07$ ,  $n = 20$  spores per collection], echinulate with slender conical spines, globose to subglobose, inamyloid. *Basidia* (15-) 20 - 27 (-35)  $\mu\text{m} \times$  (10-) 11 - 15  $\mu\text{m}$ , short clavate, 4 sterigmata, inamyloid. *Hymenophoral trama* parallel, inamyloid. *Pleurocystidia* 54 - 67  $\mu\text{m} \times$  (15-) 17 - 22 (-24)  $\mu\text{m}$ , ventricose with narrow obtuse appendage, thin-walled, dextrinoid. *Cheilocystidia* (50-) 58 - 68 (-75)  $\times$  (23-) 24 - 27 (-29)  $\mu\text{m}$ , same as pleurocystidia. *Pileipellis* 18 - 32  $\times$  (8-) 10- 20  $\mu\text{m}$ , a tangled trichodermium with short-celled hyphae forming a pile, black to dark brown in KOH, dextrinoid. *Stipitipellis* with short and compact hyphae forming a pile with inflated sphaerocysts hyphae present; *Caulocystidia* 22.4 - 32.8 (-36.8)  $\times$  13.6 - 20  $\mu\text{m}$ , napiform, thin-walled, black to dark brown, dextrinoid. *Hyphae* without clamp connection.

*Habit, habitat, and distribution:* solitary on soil of *Callerya atropurpurea*; reported from Sarawak by Corner (1972).

*Material examined:* MALAYSIA. Negeri Sembilan, Hutan Simpan Pasoh, 2°58'7"N, 102°17'49"E, 111 m, 20 September 2006, *leg. Chan H.T.* (KLU-M1106).

*Commentary:* From other species in the region described by Corner (1972), *S. foveatus* seems distinguished by the short and broad, moniliform elements forming the pileipellis. Further, the spores are echinulate (neither reticulate nor cristate). Macroscopically, the lack of velar remains on the stipe, the reticulate apex and the adnexed hymenophore appear distinctive.



**Figure 4.21:** *Strobilomyces foveatus* Corner (KLU-M1106). **a.** Basidiomes **b.** Basidiospores. **c.** Basidia. **d.** Pleurocystidia. Pileocystidia. **e.** Cheilocystidia. **f.** Caulocystidia. **g.** Sphaerocysts hyphae. **h.** Pileipellis. **i.** Stipitipellis. Bars: a = 100 mm. b-g = 10  $\mu$ m. h-i = 20  $\mu$ m.

22. *Austroboletus longipes* (Massee) Wolfe, Biblioth. Mycol. 69: 105. 1979 (1980).

(Fig. 4.22, Plate 4.3 A, B)

≡*Boletus longipes* Massee, Kew Bull. Misc. Inform. 5: 207. 1909, ss Corner (1972).

≡*Porphyrellus longipes* (Massee) Wolfe & R.H. Petersen, Mycotaxon 7: 160. 1978.

≡*Boletus tristis* Pat. & C.F. Baker, J. Straits Branch Royal Asiatic Soc. 78: 70. 1918.

≡*Porphyrellus tristis* (Pat. & C.F. Baker) Singer, Farlowia 2: 118. 1945.

≡*Austroboletus tristis* (Pat. & C.F. Baker) Wolfe, Biblioth. Mycol. 69: 127.

1979(1980).

*Pileus* (30-) 44 – 60 (-73) cm broad, convex, dry, velvety, even to uneven, brown (5E4, 5F7), with decurved to slightly uplifted margin, not bruising, turning red with NH<sub>4</sub>OH. *Context* 5 - 9 mm thick, soft and spongy, white to yellowish white, no colour changed when exposed, red with NH<sub>4</sub>OH. *Hymenophore* tubulose, adnexed to adnate, light yellow (3A5, 4A3), with concolorous pores less than 1 mm broad, bruising red, sometimes with fine black discoloration, red with NH<sub>4</sub>OH. *Stipe* 5.5 - 8 (-11.6) cm long, 4 – 7 (-9) mm broad, central, equal to slightly subclavate, dry, finely costate to finely sublacunose or finely elongate reticulate, otherwise subpruinose, brown to dark brown (5E4, 6F3), white at the base, no colour changed when bruised, red with NH<sub>4</sub>OH with white basal mycelium; interior white with chalky consistency, red with NH<sub>4</sub>OH.

*Basidiospores* (13-) 14-19 (-22) × 6 -7 μm [  $x_m = 16.5 \pm 1.78 \times 6.22 \pm 0.43$  μm,  $x_{mr} = 16.5 - 17.9 \times 6.22 - 6.53$  μm,  $x_{mm} = 17.2 \pm 0.99 \times 6.38 \pm 0.22$  μm, Q = 2.14 – 3.27,  $Q_{mr} = 2.66 - 2.75$ ,  $Q_{mm} = 2.7 \pm 0.06$ , n = 115 spores per 4 collections], ellipsoid to subfusoid, smooth under light microscope, finely pitted under Nomarski differential interference contrast optics, dextrinoid, translucent purple in KOH. *Basidia* (20-) 22-27 × 10-12 (-15) μm, 4-sterigmate, short-clavate, hyaline, inamyloid. *Cheilocystidia* (40-) 49 – 65 (-95) × (13-) 16 - 20 (-22) μm, ventricose to subventricose to subfusoid, hyaline,

inamyloid, thin-walled. *Hymenophoral trama* divergent (boletoid), with hyaline elements, inamyloid. *Pileipellis* a trichodermium, composed of erect to suberect cylindrical elements, (60-) 65 - 108 (-118) × (9-) 10 - 15 (-24) μm, smooth, thin-walled, hyaline or pale brown content, inamyloid. *Stipitipellis* a trichodermium of caulocystidia 29 - 43 × 9 - 15 μm, subcylindric to clavate, smooth, thin-walled, with brown content in KOH, dextrinoid. *Clamp connections* absent.

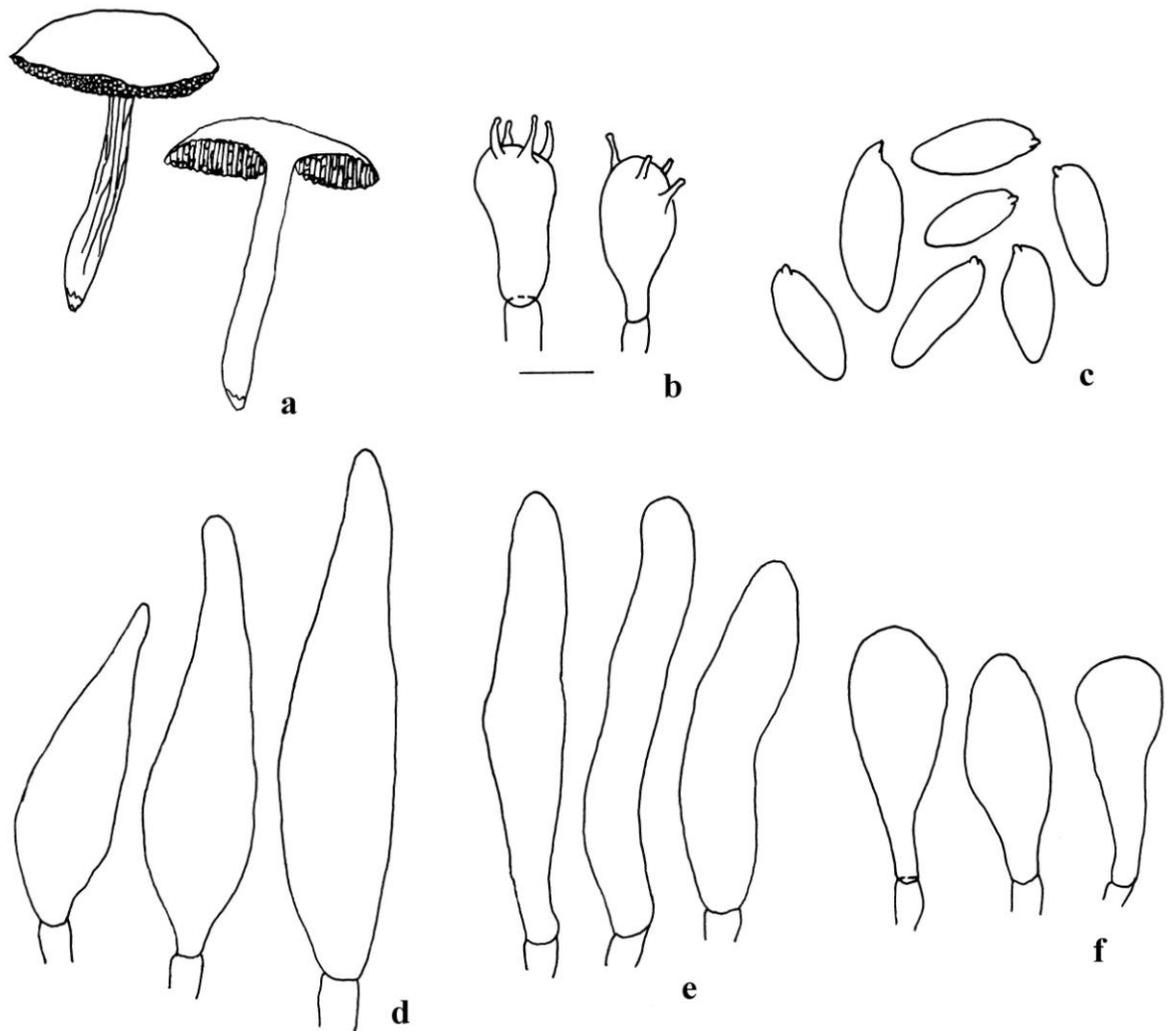
*Habit, habitat, and distribution:* gregarious on soil in forests of Dipterocarpaceae (*Shorea* sp.); reported from peninsular Malaysia and Sarawak as well as Singapore.

*Material examined:* MALAYSIA. Negeri Sembilan, Pasoh, Hutan Simpan, N 2°58'7", E 102°17'49", 111 m, 20 September 2006, leg. Chan H.T. (KLU-M1107, KLU-M1108, KLU-M1109, KLU-M1110).

*Commentary:* The nearly uniform brown colors of the pileus and stipe along with the red staining; yellowish hymenophore and red reaction with alkali are distinguishing macroscopic features. The purple reaction of the spores in alkali is immediately distinctive under the microscope. It should be noted here that the spores appear smooth when examined with the light microscope using bright field optics. However, with the aid of Nomarski differential interference contrast optics, the finely pitted ornamentation is just barely visible. *Boletus longipes* is reported to have dark red brown spores in deposit (Corner 1972).

The description of my material matches the concept provided by Corner (1972) and he placed the species in subgenus *Tylopilus* noting *B. tristis* as synonymous. Singer (1945) placed this latter species in *Porphyrellus*. Later, Wolfe and Petersen (1978) looked at the type specimens of *B. tristis* and *B. longipes* and noted subtle differences in spore ornamentation only seen with the SEM and recognized placement of both species in *Porphyrellus* with synonymy as noted by Corner (1972). In his subsequent monographic work, Wolfe (1979) placed both *B. tristis* and *B. longipes* in *Austroboletus*

because of the spore ornamentation that distinguishes it from the smooth-spored genera *Tylopilus* and *Porphyrellus* and maintained them as distinct species. Recently, Watling (2000) provided an assessment in which he summarized the findings of Wolfe and Petersen (1978) and Wolfe (1979).



**Figure 4.22:** *Austroboletus longipes* (Masse) Wolfe (KLU-M1110). **a.** Basidiomes. **b.** Basidia. **c.** Basidiospores. **d.** Cheilocystidia. **e.** Pleurocystidia. **f.** Caulocystidia. Bars: a = 10 mm. b – f = 10  $\mu$ m.



**Plate 4.3:** A: *Austroboletus longipes* (×4.0); B: Spore of *Austroboletus longipes* (Nomarski differential interference contrast optics, 5 μm); C: *Strobilomyces foveatus* (×3.3); D: *Aureoboletus thibetanus* (×2.2); E: *Boletus peltatus* (×9.0); F: *Boletus phaeocephalus* (×3.3); G: *Boletus* cf. *craspedius* (×3.3); H: *Boletellus corneri* (×7.1).

Bar = 10 mm

23. *Tylopilus nigropurpureus* Hongo, Mem. Shiga Univ. 23: 40. 1973.

(Fig. 4.23a, 4.23b, Plate 4.4 C)

= *Poria nigropurpurea* (Schwein.) Sacc., *Syll. fung.* (Abellini) 6: 304 (1888)

= *Boletus nigropurpureus* Corner, *Boletus in Malaysia*, p. 178. 1972, *non* Schweintiz, *Schr. Naturf. Ges. Leipzig* 1: 99. 1822.

= *Tylopilus nigropurpureus* (Corner) Watling in Watling & Turnbull, *Edinburgh J. Bot.* 51: 336. 1994, *nom. superfl.*

*Pileus* 49 – 80 mm broad, convex, dry, velvety when young, slightly areolate when matured, with even margin edge, deep brownish grey to brownish black (5F2, 5F7, 5F6), negative reaction with NH<sub>4</sub>OH. *Context* 13 – 16 mm at disc, 2 – 4 mm at margin, thick, soft and spongy, white, slowly black when exposed, negative reaction with NH<sub>4</sub>OH. *Hymenophore* 20 – 30 mm, tubulose, adnate, orange grey (5C2) to brownish grey (5D2), with fine pores, 2 – 3 per mm, black when bruised, negative reaction with NH<sub>4</sub>OH. *Stipe* 47 – 60 mm long, 10 - 20 mm broad at apex, 16 – 23 mm at base, central, subclavate to clavate, dry, reticulate surface, orange grey (5C2) at apex, brownish grey (5F2) to black at base; *context* spongy, black when exposed, negative reaction with NH<sub>4</sub>OH, with solid interior and black basal mycelium; *Taste* slightly bitter.

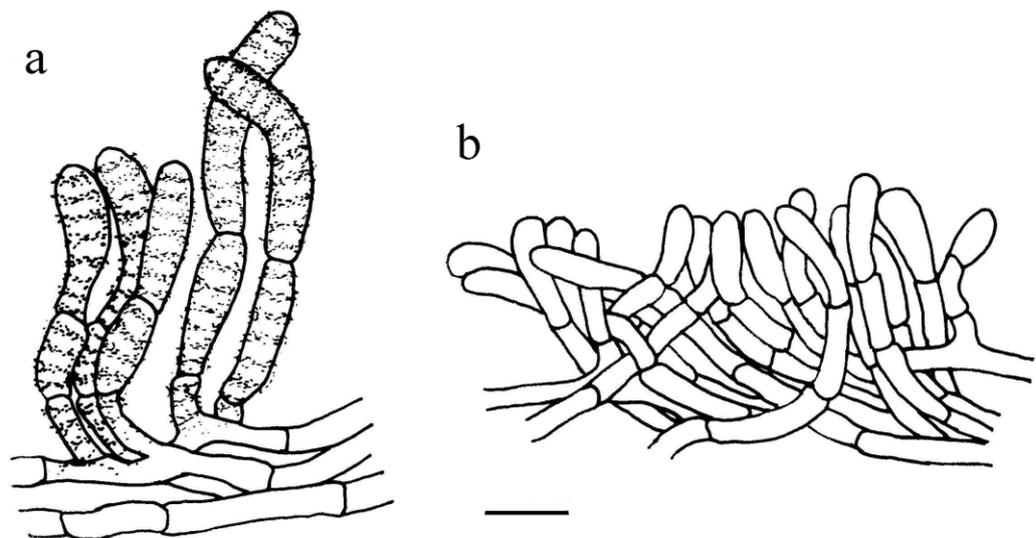
*Basidiospores* olive to olive brown 10 - 14 (-18) × 5 - 6 μm [  $x_m = 11.6 \pm 1.54 \times 5.82 \pm 0.38 \mu\text{m}$ ,  $x_{mr} = 9.6 - 11.6 \times 4.62 - 5.82 \mu\text{m}$ ,  $x_{mm} = 10.68 \pm 1.01 \times 5.22 \pm 0.60 \mu\text{m}$ ,  $Q = 1.67 - 2.17 (-3)$ ,  $Q_{mr} = 1.60 - 2.5 (-3)$ ,  $Q_{mm} = 2.06 \pm 0.05$ ,  $n = 75$  spores per 3 collections], ellipsoid to ovoid, smooth, inamyloid. *Basidia* (22-) 26 – 29 (- 40) × (-11) 12 -14 μm, 4-sterigmate, clavate, hyaline, inamyloid. *Hymenophoral cystidia* 42 - 52 × 13 – 15 (-18) μm, ventricose, thin-walled, inamyloid, hyaline in KOH. *Hymenophoral trama* parallel, with hyaline elements, inamyloid. *Pileipellis* a hymenium of incrustated hyphae, with pileocystidia (8-) 9 – 15 (-16) × 14 – 17 μm, thin-walled, with pale brown

content, dextrinoid. *Stipitipellis* hymeniform with caulocystidia (29-) 30 – 40 × 10 – 14 (-15) μm, subcylindric to clavate, smooth, thin-walled, with brown content in KOH, dextrinoid. *Clamp connections* absent.

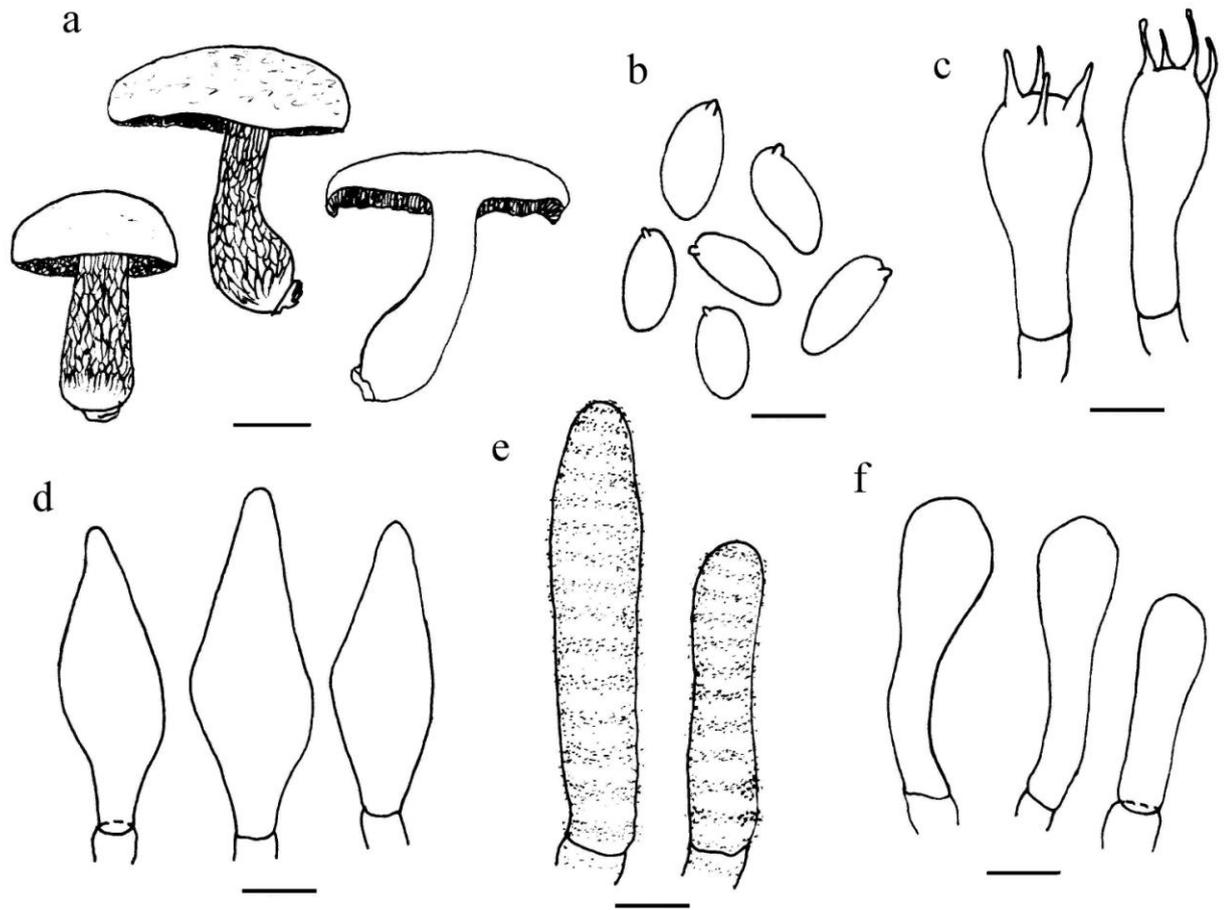
*Habit, habitat, and distribution:* solitary to gregarious on soil in highland forests; reported from Peninsular Malaysia as well as Singapore.

*Material examined:* MALAYSIA. Pahang, Fraser Hill, Jalan Air Terjun Jeriau. N 03°43'36.0", E 101°43'00.6", 970m; 29 November 2003, *leg. Tan Y.S.* (KLU-M1111), 1 January 2007, *leg. Chan H.T.* (KLU-M1112), 30 May 2007, *leg. Chan H.T.* (KLU-M1113).

*Commentary:* This dark brown to black species was first reported by Corner (1972) and placed in subgenus *Tylopilus*. This species is common in Fraser Hill and can be found throughout the year. The species resembles a *Strobilomyces* without veil and ornamented spores. The reticulate stipe differentiates this species from *Tylopilus alboater* whose stipe is smooth.



**Figure 4.23b:** *Tylopilus nigropurpureus* Hongo (KLU-M1113). **a.** Pileipellis. **b.** Stipitipellis. Bars: a – b = 20 μm.



**Figure 4.23b:** *Tylopilus nigropurpureus* Hongo (KLU-M1113). **a.** Basidiomes. **b.** Basidiospores. **c.** Basidia. **d.** Hymenial cystidia. **e.** Pileocystidia. **f.** Claulocystidia. Bars: a = 10 mm. b – f = 10  $\mu$ m.

24. *Tylopilus albo-ater* (Schwein.) Murrill, Mycologia 1: 16. 1909.

(Fig. 4.24, Plate 4.4 B)

≡ *Boletus alboater* Schwein., Schr. Naturf. Ges. Leipzig 1: 95. 1822.

*Pileus* 62 mm broad, convex to plane, dry and areolate, with even to slightly wavy margin edge, deep brownish black (7F8, 5F5), turning slowly black when injured, yellow and slowly black with NH<sub>4</sub>OH. *Context* 8 mm at disc, 1 – 3 mm at margin, solid, grayish white (4B6), slowly reddish pink and then black when exposed, turn yellow and slowly changed to black with NH<sub>4</sub>OH. *Hymenophore*, tubulose, adnexed 6 mm, grayish white to pinkish white (7A2), pores concolourous with tubes, 2 – 3 pores per mm, reddish pink and then black when exposed, yellow slowly then black with NH<sub>4</sub>OH. *Stipe* 45 × 12 μm, central, subclavate to cylindrical, dry, fibrillose, brownish black (5F5) with solid content, grayish white, slowly reddish pink then black when exposed, negative reaction with NH<sub>4</sub>OH, with black basal mycelium; *Taste* a little bitter.

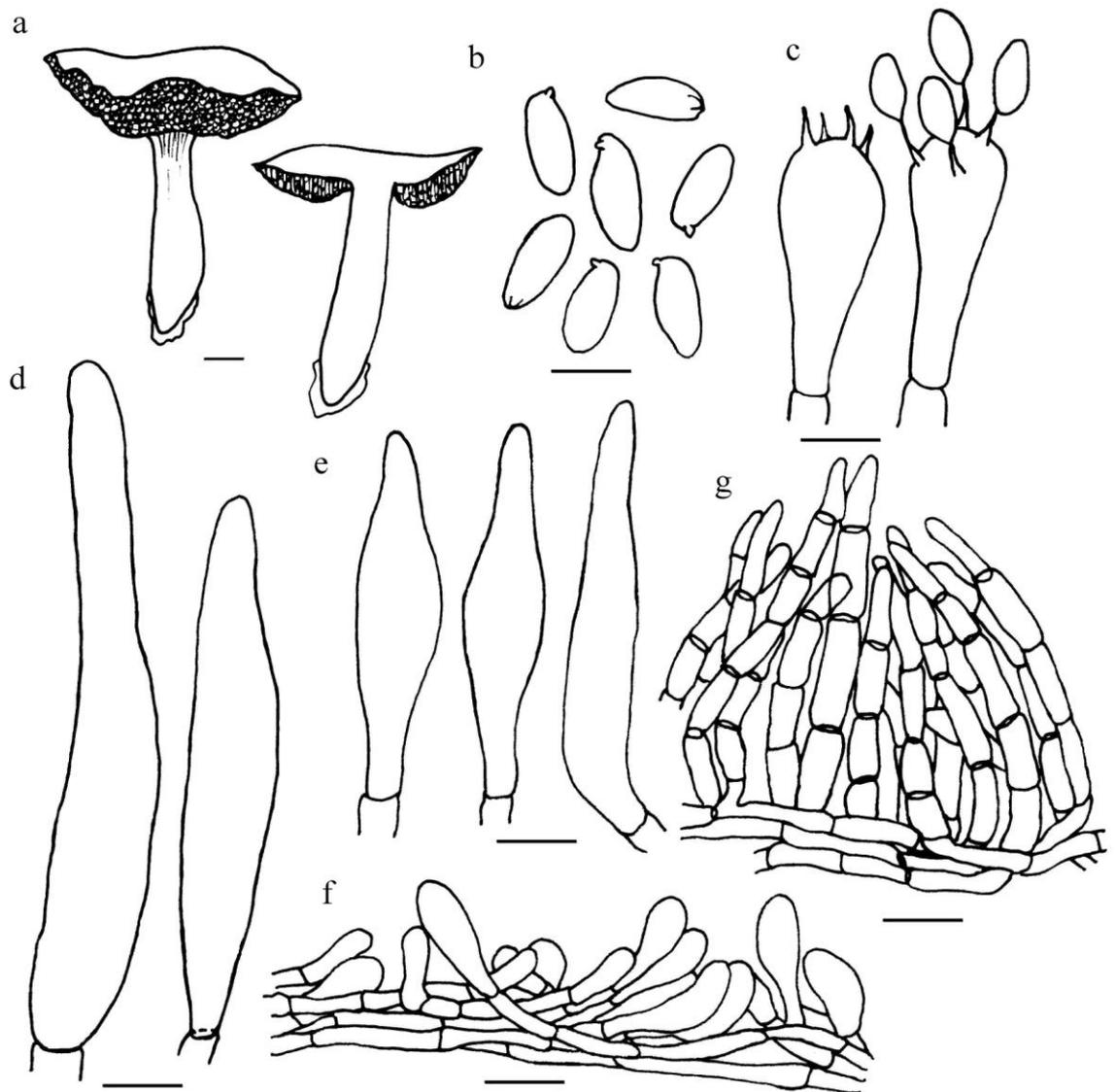
*Basidiospores* 12 – 16 × 4 – 6 μm [  $x_m = 13.7 \pm 1.45 \times 5.25 \pm 0.64$  μm,  $x_{mr} = 9.25 - 15.18 \times 4.15 - 5.87$  μm,  $x_{mm} = 12.71 \pm 3.09 \times 5.09 \pm 0.87$  μm,  $Q = 2.20 - 3.75$ ,  $Q_{mr} = 2.27 - 2.63$ ,  $Q_{mm} = 2.50 \pm 0.20$ ,  $n = 80$  spores per 4 collections], long ellipsoid to subfusiform, smooth, light pinkish brown, inamyloid. *Basidia* 30 – 40 (-45) × (10-) 11 – 13 (-14) μm, 4-sterigmate, clavate, hyaline, inamyloid. *Pleurocystidia* (46-) 49 – 56 (-58) × (9-) 10 – 12 μm, subventricose to ventricose, thin-walled, brown in KOH, dextrinoid. *Cheilocystidia* (43-) 50 – 65 (-102) μm, same characteristics pleurocystidia. *Hymenophoral trama* parallel to subdivergent, with hyaline elements, inamyloid. *Pileipellis* a palisade trichodermi with compact pile hyphae at 110 – 180 μm high; pileocystidia (21-) 28 – 50 (-62) × 11 – 14 (-17) μm, subventricose, with dark brown content, dextrinoid. *Stipitipellis* covered by a trichoderm of short clavate caulocystidia

20 – 40 µm high, caulocystidia (12-) 15 – 23 (-27) × (5-) 7 – 10 µm, smooth, thin-walled, with dark brown content, dextrinoid. *Clamp connections* absent.

*Habit, habitat, and distribution:* solitary to gregarious on soil in highland forests and lowland Dipterocarpaceae forest; reported from Peninsular Malaysia, Singapore and North America.

*Material examined:* MALAYSIA. Selangor, Hulu Langat, Forest Reserve Sungai Chongkak, N 03°12'00.3", E 101°50'07.4", 149 m, 22 July 2005, leg. Chan H.T. and Chan H.H (KLU-M1114); 7 January 2005, leg. Desjardin D. (KLU-M1115); 7 March 2005, leg. Chan H.T. (KLU-M1116). Johor, Selai, Endau Rompin National Park, Takah Tinggi trail, N 02°25'59.4", E 103°14'49.5", 120 m, 17 February 2006, leg. Chan H.T. (KLU-M1117).

*Commentary:* This easily recognized species has a distinctive blackish, velvety cap and a dark, smooth stem. Its pore surface and context is initially white, becomes pinkish and bruises promptly red, then black with injured. The blackish colour can easily confuse with *Tylopilus nigropurpureus*. *T. nigropurpureus* can easily be distinguished from *T. albo-ater* by the absence of reticulate on the stipe.



**Figure 4.24:** *Tylopilus albo-ater* (Schwein.) Murrill (KLU-M1114). **a.** Basidiomes. **b.** Basidiospores. **c.** Basidia. **d.** Cheilocystidia. **e.** Pleurocystidia. **f.** Stipitipellis. **g.** Pileipellis. Bars: a = 10 mm. b – f = 10  $\mu$ m. g = 20  $\mu$ m.

25. *Tylophilus virens* (W.F. Chiu) Hongo, *Mem. Fac.lib. Arts Educ. Shiga Univ.*, Nat. Sci. 14: 46 (1964). (Fig. 4.25, Plate 4.4 G)

≡ *Boletus virens* W.F. Chiu, *Mycologia* 40: 206 (1948)

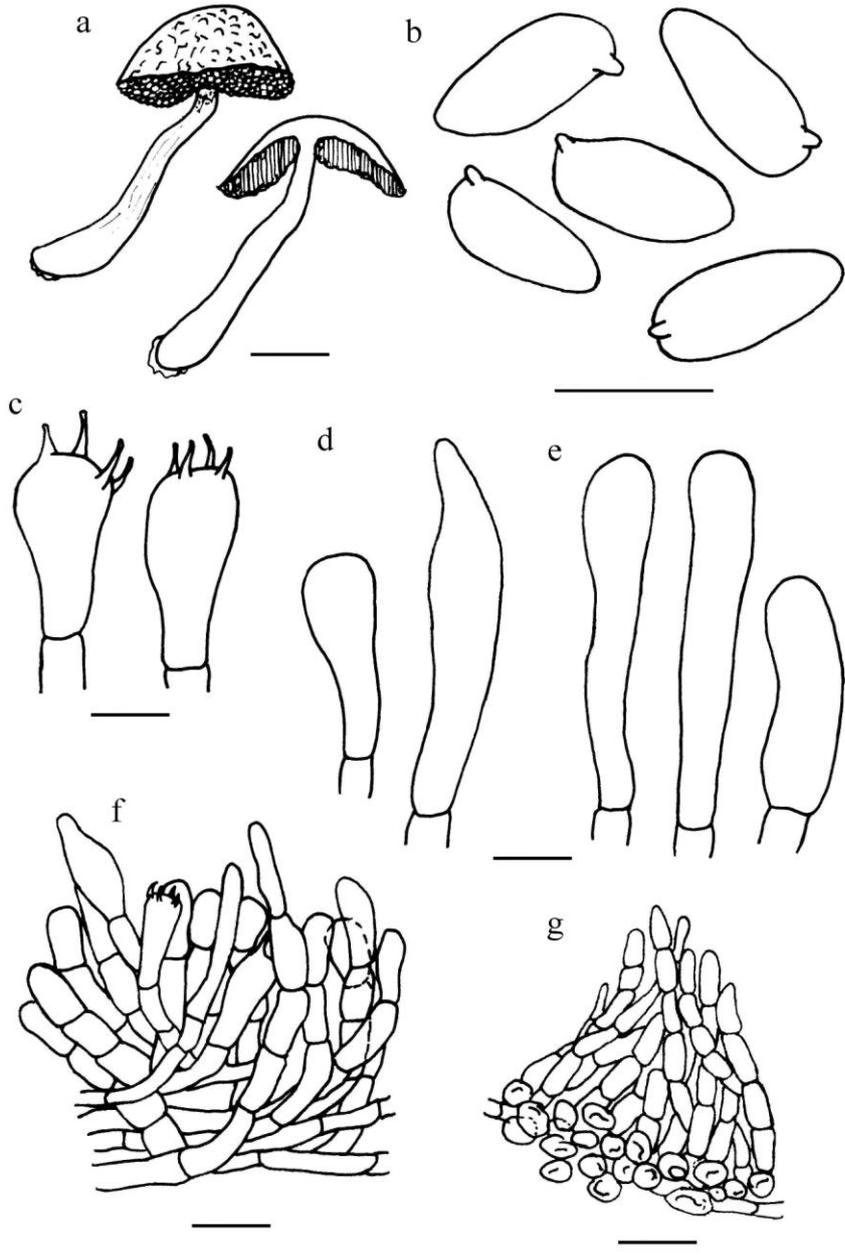
*Pileus* 10 – 20 mm broad, convex, dry, uneven to rugose, yellow (3A7) at margin, olivaceous yellow (4B7) at disc, with straight, even margin, negative reaction with  $\text{NH}_4\text{OH}$ . *Context* 2 – 2.5 mm thick, soft and spongy, yellow (3A5), not bruising, negative reaction with  $\text{NH}_4\text{OH}$ . *Hymenophore* tubulose, free 5 mm thick near apex, 1 – 2 mm thick at margin, pinkish white (5A2). *Pores* concolorous with tubes, 2 – 3 pores per mm, no reaction when exposed or with  $\text{NH}_4\text{OH}$ . *Stipe* 30 mm long, 2 mm broad at apex, 6 mm broad at base, tapering upward to subclavate, central, dry, pubescent, yellow (3A7); interior solid, yellow (3A7), no reaction when exposed or with  $\text{NH}_4\text{OH}$ ; *basal mycelium* yellow. *Taste* mild.

*Basidiospores*  $9.2 - 11.1 \times 4.5 - 5.4 \mu\text{m}$ , [ $x_m = 10.53 \pm 0.49 \times 4.90 \pm 0.29 \mu\text{m}$ ,  $Q = 1.83 - 2.47$ ,  $Q_m = 2.16 \pm 0.14$ ,  $n = 30$  spores per one collection] smooth, ellipsoid to boletoid, pinkish in mass, inamyloid. *Basidia*  $21.6 - 24.0 (-26.0) \times (7.2-) 8.0 - 9.6 \mu\text{m}$ , 4 sterigmate, clavate, hyaline in KOH, inamyloid. *Hymenophoral trama* divergent with hyaline elements, inamyloid. *Pleurocystidia*  $25 - 50 \times 8 - 10 \mu\text{m}$ , subventricose to short clavate, thin-walled, hyaline, inamyloid. *Cheilocystidia*  $30 - 38 \times 6 - 8 (-10) \mu\text{m}$ , long clavate, thin-walled, hyaline, inamyloid. *Pileipellis* a trichodermial palisade with compact pile hyphae at 200 – 320  $\mu\text{m}$  high; inflated hyphae absent; *pileocystidia*  $20 - 25 \times 10 - 16 \mu\text{m}$ , subventricose, hyaline, inamyloid. *Stipitipellis* covered with sterile hymenium of basidia, ventricose to clavate cystidia, 100 – 110  $\mu\text{m}$  high; *caulocystidia*  $21 - 40 (-48) \times 10 - 17 \mu\text{m}$ , thin-walled, hyaline, inamyloid. *Clamp connections* absent.

*Habit, habitat, and distribution*: solitary on soil in lowland Dipterocarp forest, (*Mangifera subsessilifolia*); originally reported from Yunnan China.

*Material examined:* MALAYSIA. Johor, Endau Rompin National Park (Peta). N 02°31'47.8", E 103°22'01.5", 111 m, 21 May 2007, *leg. Chan H.T.* (KLU-M1118).

*Commentary:* This single collection with distinctive olivaceous pileus, yellowish stipe with yellow, solid context, yellow basal mycelium, and no colour changed at the tissues when bruised point toward this taxon. This species was reported by Chiu (1948) in Yunnan. Unfortunately, this collection consisted of only one solitary basidiocarp.



**Figure 4.25:** *Tylopilus virens* (W.F. Chiu) Hongo (KLU-M1118). **a.** Basidiomes. **b.** Basidiospores. **c.** Basidia. **d.** Pleurocystidia. **e.** Cheilocystidia. **f.** Stipitipellis. **g.** Pileipellis. Bars: a = 10 mm. b – e = 10  $\mu$ m. f = 20  $\mu$ m. g = 40  $\mu$ m.

26. *Tylopilus pernanus* (Pat. & C.F. Baker) Watling, Mycologia 2000: 583. 2000.

(Fig. 4.26, Plate 4.1 H)

≡ *Boletus pernanus* Pat. & C.F. Baker, J. Straits Branch Royal Asiatic Soc. 78: 72.

1918.

≡ *Boletus nanus* Masee, Kew Bull Misc. Inform. xx: 208. 1909. *sensu* Corner *non auct.*

≡ *Tylopilus nanus* (Masee) Singer, Agaricales Mod. Tax., p. 736. 1975.

*Pileus* 10 – 20 mm broad, convex, dry velvety surface and becoming rugose toward margin olive brown (4D6) when young fruiting bodies, turning greyish brown (5F3) with maturity, straight margin with even edge, negative reaction with NH<sub>4</sub>OH. *Context* 2 – 3 mm thick, soft and spongy, light yellow (2A5), discoloring absent, no reaction with NH<sub>4</sub>OH. *Hymenophore* tubulose, 2 - 3 mm thick, pinkish white (10A2), free. *Pores* concolorous with tubes, 2 – 3 pores per mm, no reaction when exposed and with NH<sub>4</sub>OH. *Stipe* 13 - 25 mm long, 2 - 4 mm broad at apex, 3 – 4 mm broad at base, cylindrical to slightly tapering upward, central, dry, tomentose, slightly blue at apex near hymenophore, yellow at apex (2A6), pinkish red (8A7) at center and base of stipe; interior solid, yellow, negative reaction with NH<sub>4</sub>OH; *basal mycelium* yellow with orange red rhizomorph.

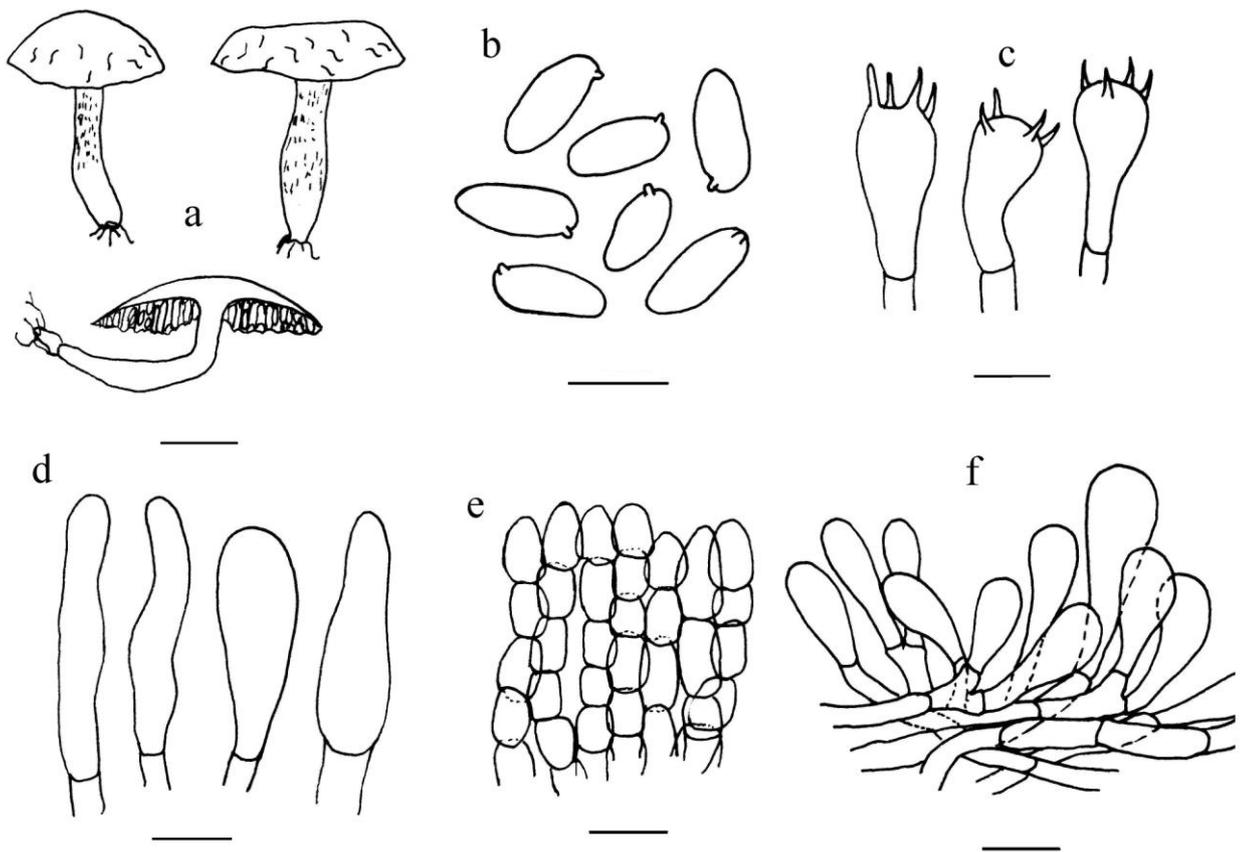
*Basidiospores* (9-) 10 - 14 × 5 - 6 μm, [ $x_m = 12.1 \pm 1.27 \times 5.08 \pm 0.27$  μm, Q = (1.8-) 2.0 – 2.8, Q<sub>m</sub> = 2.398 ± 0.26, n = 30 spores per collection] smooth, ellipsoid, pinkish in mass, dextrinoid. *Basidia* (-12) 16 – 17.6 (18.4) × 6.4 – 7.2 (-8) μm, 4 sterigmata with 2 - 4 μm length, clavate, hyaline in KOH, inamyloid. *Hymenophoral trama* parallel to subdivergent with hyaline elements, inamyloid. *Hymenial cystidia* 24 – 27.5 (-32) μm × (4.8-) 6.4 -8.8 (-9.6) μm, clavate to cylindrical to subventricose, thin-walled, hyaline, inamyloid. *Pileipellis* a structure of moniliform, elements, forming a pile

of hyphae, with end cells  $20 - 25.6 (-34.4) \times 10.4 - 12.8 (-14.4) \mu\text{m}$ , thin-walled, hyaline, inamyloid. *Stipitipellis* with scattered clavate cystidia; *caulocystidia*  $24 - 40 (-45) \times (20-) 25 - 30 (-34) \mu\text{m}$ , hyaline, inamyloid. *Clamp connections* absent.

*Habit, habitat, and distribution:* gregarious on soil in highland forest (*Lithocarpus* sp.); reported from Peninsular Malaysia.

*Material examined:* MALAYSIA. Pahang, Fraser Hill, Jeriau Waterfall Road, N  $03^{\circ}43'36.0''$ , E  $101^{\circ}43'00.6''$ , 970m, 8 January 2006, *leg. Chan H.T.* (KLU-M1119).

*Commentary:* The interpretation and confusion of concepts in this taxon was reviewed by Watling (2000). The entity illustrated and described by Corner (1972) circumscribes a very small bolete with pale olive pileus, chrome yellow stipe base and extremely fine pinkish to pale reddish ornamentation in the mid stipe. This bolete is often found growing on soil of road banks, much like *B. patouillardii*.



**Figure 4.26:** *Tylopilus pernanus* (Pat. & C.F. Baker) Watling (KLU-M1119). **a.** Basidiomes. **b.** Basidiospores. **c.** Basidia. **d.** Hymenial cystidia. **e.** Pileipellis. **f.** Stipitipellis. Scale bars: a = 10 mm. b – d = 10  $\mu$ m. e – f = 20  $\mu$ m.

27. *Tylopilus ballouii* (Peck) Singer, Amer. Midl. Nat. 37: 104. 1947.

(Fig. 4.27, Plate 4.4 H)

≡ *Boletus ballouii* Peck, Bull. New York St. Mus. 157: 22. 1912.

≡ *Rubinoboletus ballouii* (Peck) Heinem. & Rameloo, Bull. Jard. Bot. Nat. Belg. 53: 295. 1983.

*Pileus* 41 - 61 mm broad, subconvex to plane to slightly depressed, dry, smooth, with plane to slightly uplifted margin, orange (6B8), not bruising, orange with NH<sub>4</sub>OH. *Context* 7 mm thick in disc, 1 mm thick at margin, soft and spongy, white, no colour changed when exposed or with NH<sub>4</sub>OH. *Hymenophore* 2 - 5 (-6) mm thick, tubulose, decurrent, yellowish white (1A2), with concolorous pores less than 1 mm broad, bruising brown, negative reaction with NH<sub>4</sub>OH. *Stipe* 39 - 75 mm long, 6 - 14 mm broad at apex, 5 - 9 mm broad at base, central to slightly eccentric, tapering downward, dry, fibrillose, orange (6B8), with context solid and white, no colour changed when bruised or with NH<sub>4</sub>OH, white basal mycelium.

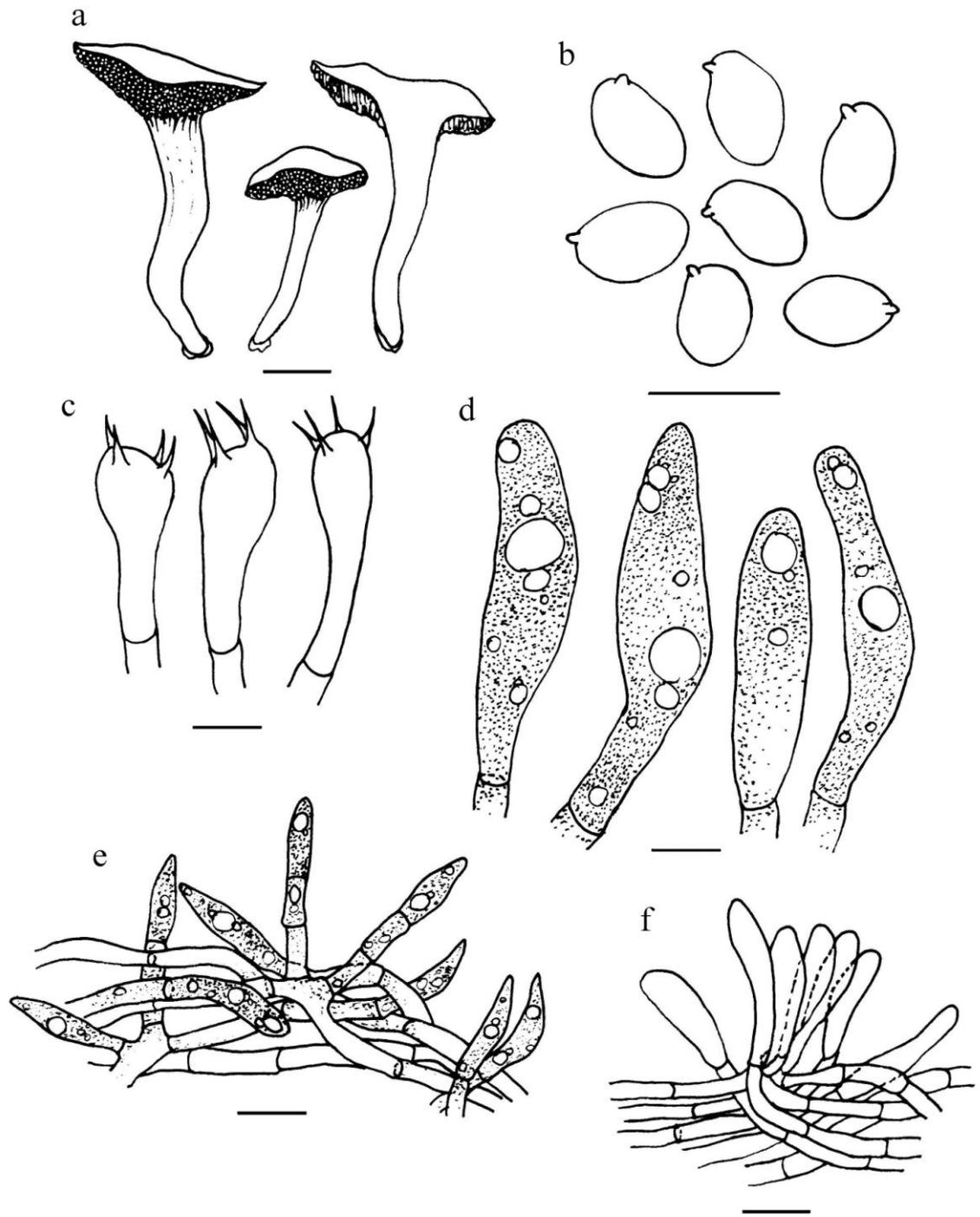
*Basidiospores* 8 - 10 (-13) × 4 - 6 μm [  $x_m = 8.90 \pm 1.20 \times 5.10 \pm 0.46$  μm,  $x_{mr} = 7.70 - 8.90 \times 4.75 - 5.23$  μm,  $x_{mm} = 8.26 \pm 0.48 \times 5.02 \pm 0.20$  μm, Q = 1.60 - 2.00 (-2.25),  $Q_m = 1.75 \pm 0.20$ ,  $Q_{mr} = 1.20 - 2.25$ ,  $Q_{mm} = 1.65 \pm 0.08$ , n = 70 spores per 4 collections], ellipsoid to ovoid, smooth, olive in colour, inamyloid, hyaline in KOH. *Basidia* 32 - 39 × 9 - 12 μm, 4-sterigmate, clavate, hyaline in KOH, inamyloid. *Hymenial cystidia* (42-) 46 - 55 (-61) × (10-) 11 - 12 μm, cylindrical to subclavate to subventricose, oil drop present, brown in KOH, dextrinoid. *Hymenophoral trama* divergent (boletoid), with hyaline elements, inamyloid. *Pileipellis* a trichodermium of pileocystidia and hyphae; *pileocystidia* 20 - 30.4 × 4.8 - 7.2 μm, ventricose, containing yellowish oil drops, thin-walled, inamyloid. *Stipitipellis* covered by a hymenium of

cystidia; *caulocystidia* 39.2 – 52.0 × 8 – 9 (-12.8) μm, clavate, smooth, thin-walled, inamyloid. *Clamp connections* absent.

*Habit, habitat, and distribution:* gregarious on soil in forests of Dipterocarpaceae and Fagaceae; a common species reported from Peninsular Malaysia, Sarawak and Singapore.

*Material examined:* MALAYSIA. Negeri Sembilan, Pasoh, Hutan Simpan, 2°58'7"N, 102°17'49"E, 111 m, 20 September 2006, *leg. Chan H.T.* (KLU-M1120). Pahang, Fraser's Hill, Kindersley trail, N 03°43'36.0", E 101°43'00.6", 970m, 29 September 2003, *leg. Tan Y.S.* (KLU-M1121). Pahang, Fraser's Hill, Jeriau waterfall road, N 03°43'36.0", E 101°43'00.6", 970m, , 25 September 2003, *leg. Tan Y.S.* (KLU-M1122, KLU-M1123).

*Commentary:* Corner (1972) included this taxon in his treatment of Malaysian boletes. However, his illustration and description circumscribe an entity with favoloid hymenophore: “. . . pores 0.8-2 mm, angular with short internal dissepiments, . . .” This morpho-type has been collected in Fraser's Hill and in N Queensland by Halling (*pers. com.*), but is certainly not identical to the form originally described by Peck. However, the Malaysian material described above conforms to material and the type concept from East North America. Still, the collections with favoloid hymenophore are most certainly closely related and are provisionally named *Tylopilus favo-ballouii* *nom. prov.* (KLU-M1124).



**Figure 4.27:** *Tylopilus ballouii* (Peck) Singer (KLU-M1120). **a.** Basidiomes **b.** Basidiospores. **c.** Basidia. **d.** Hymenial cystidia. **e.** Pileipellis **f.** Stipitipellis. Bars: a = 20 mm. b – d = 10  $\mu\text{m}$ . e – f = 20  $\mu\text{m}$ .

28. *Tylopilus favo-ballouii* Chan H. T. & Halling R.E., nom. prov.

(Fig. 4.28, Plate 4.4 E)

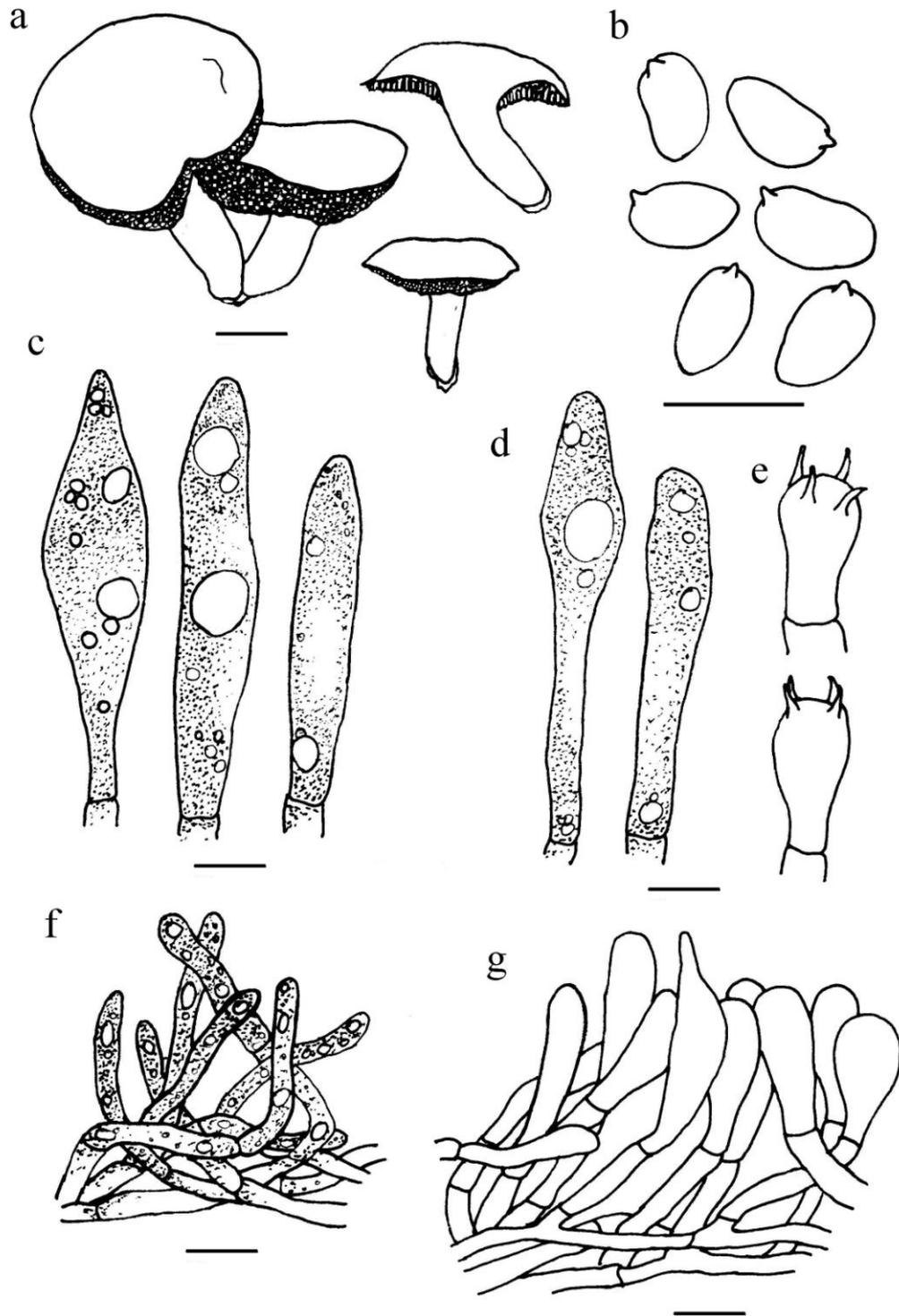
*Pileus* 31 – 82 mm broad, convex, dry, with decurved margin when young, and uplifted when mature, smooth to slightly velvety, orange (6B8). *Context* 6 - 10 mm thick, soft and spongy, white, no colour changed when exposed, negative reaction with NH<sub>4</sub>OH. *Hymenophore* 1.5 – 6 mm thick, tubulose, adnate to slightly decurrent, yellowish white to pale yellow (4A2), with concolorous medium favoloid pores, about 1 per mm, bruising brown, negative reaction with NH<sub>4</sub>OH. *Stipe* 35 - 45 × 11 – 14 mm, central, tapering downward, dry, fibrilose, orange (6B8), negative reaction with NH<sub>4</sub>OH, with white basal mycelium; interior white with solid content, negative reaction with NH<sub>4</sub>OH; *Taste* bitter.

*Basidiospores* (5-) 6 – 9 × 4 – 6 (-7) μm [ $x_m = 7.68 \pm 1.17 \times 5.32 \pm 0.66$  μm, Q = 1.00 – 1.80, Q<sub>m</sub> = 1.48 ± 0.23, n = 30 spores per one collection], ovoid to short ellipsoid, smooth, inamyloid. *Basidia* (14-) 20 – 30 (-32) × 8 – 10 μm, 4-sterigmate, short-clavate, hyaline, inamyloid. *Cheilocystidia* 52 – 67 (-70) × 8 – 12 (-15) μm, ventricose to subclavate to cylindrical, brownish yellow in KOH with oil drop present, dextrinoid, thin-walled. *Hymenial cystidia* (48) 52 – 70 (-72) × 6 – 11 (-15) μm. *Hymenophoral trama* divergent, with hyaline elements, inamyloid. *Pileipellis* a trichodermium, hyaline, with oil drop present, thin-walled, inamyloid. *Stipitipellis* hymeniform with caulocystidia 44 – 60 (-70) × 12 – 16 μm, subclavate to clavate to ventricose, smooth, thin-walled, hyaline, inamyloid. *Clamp connections* absent.

*Habit, habitat, and distribution:* gregarious on soil in highland forest (*Lithocarpus* sp.).

*Material examined:* MALAYSIA. Pahang, Fraser Hill, Jeriau Waterfall Road, N 03°43'36.0", E 101°43'00.6", 960 m, 8 January 2006, leg. Chan H.T. (KLU-M1124).

*Commentary:* The bright orange pigments, the short ellipsoid spores and turn brown as a result of oxidation reaction with injury on the hymenophore shows it is close to *Tylopilus ballouii* (Perk) Singer. However this collection has a favoloid hymenophore and bigger pores which is not identical to the morphological concept originally described by Peck. As mentioned in Corner's (1972) commentary of *Tylopilus ballouii* (Perk) Singer, Corner's collections are most certainly closely related and were included in his treatment in Malaysian boletes as *Tylopilus ballouii*. This confusing morpho-taxon needs further study. Still, the collections with favoloid hymenophore and big pores are provisionally named *Tylopilus favo-ballouii* *nom. prov.*



**Figure 4.28:** *Tylopilus favo-ballouii* Chan H. T., *nom. prov.*(KLU-M1124). **a.** Basidiomes. **b.** Basidiospores. **c.** Pleurocystidia. **d.** Cheilocystidia. **e.** Basidia. **f.** Pileipellis. **g.** Stiptipellis. Bars: a = 20 mm. b – e = 10  $\mu$ m. f – g = 20  $\mu$ m.

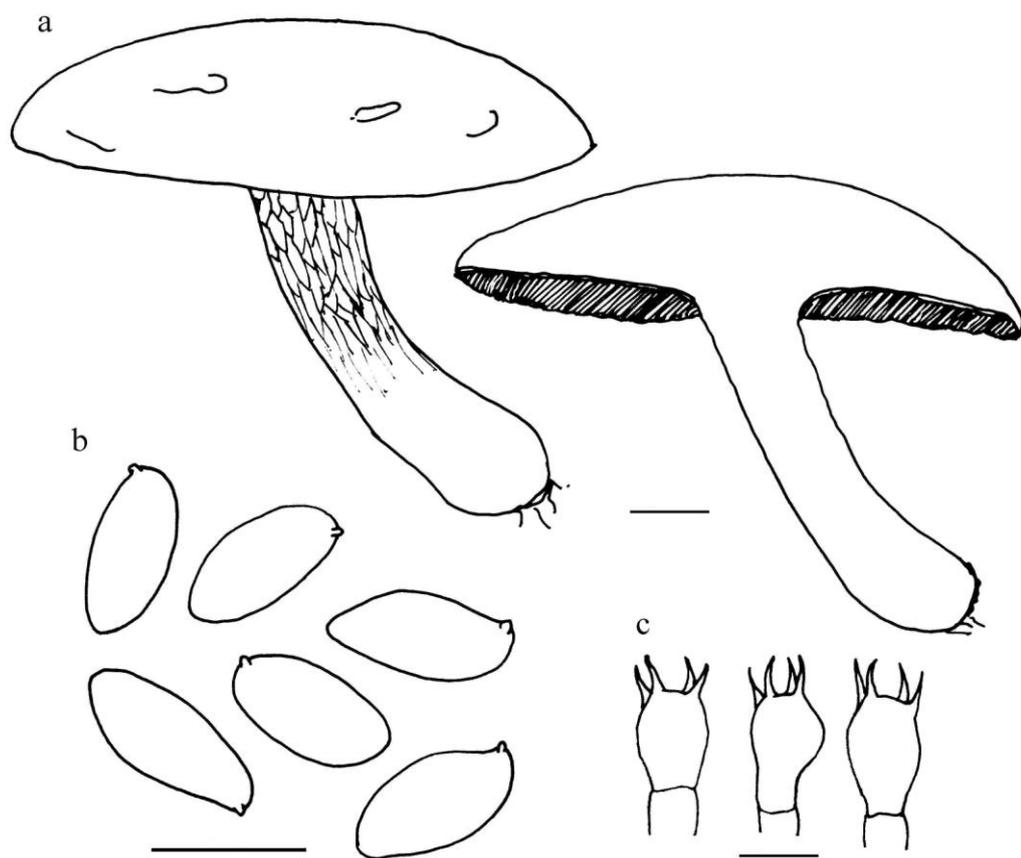
*Pileus* 150 mm broad, plano-convex, dry, subtomentose, with straight and even margin, brown (5F8), turn reddish brown with NH<sub>4</sub>OH. *Context* 30 mm thick in disc, 2 mm thick at margin, soft and spongy, white, turn brown when exposed, negative reaction with NH<sub>4</sub>OH. *Hymenophore* 8 mm thick, tubulose, adnate to slightly depressed around stipe, pinkish white to pale brownish white, with concolorous pores, fine pores 5 – 6 pores per 5 mm, bruising brown, negative reaction with NH<sub>4</sub>OH. *Stipe* 63 × 21 mm, central, subclavate stipe to almost cylindrical, dry, fine reticulate at apex and smooth at base, reddish brown to dark brown (8E8, 8F8), with context solid and white, chalky and easy to break, flesh turn brown when exposed, negative reaction with NH<sub>4</sub>OH, white basal mycelium. *Odor and taste* odor not distinctive and bitter taste.

*Basidiospores* 11 – 14 (-15) × 5 – 6 μm [ $x_m = 12.75 \pm 1.47 \times 5.18 \pm 0.38$  μm,  $Q = 2 – 2.8$  (- 3.0)  $Q_m = 2.47 \pm 0.31$ ,  $n = 30$  spores per one collections], subfusoid, smooth, pinkish colour, inamyloid, hyaline in KOH. *Basidia* 15 – 19 × 8 – 11 μm, 4-sterigmate, short clavate, hyaline in KOH, inamyloid. *Hymenial cystidia* (50-) 64 – 78 (-102) × 12 – 17 μm, fusoid ventricose, brownish in KOH, dextrinoid, thin-walled, with granular absent. *Hymenophoral trama* divergent, inamyloid. *Pileipellis* a trichodermium of matted hyphae, about 120 μm; *pileocystidia* 47 – 52 × 6 – 9 μm, long cylindrical with narrow tip, thin-walled, yellowish in KOH, slightly brownish in Melzer. *Stipitipellis* covered by a trichodermium of cystidia, 30 – 40 μm high, clavate, smooth, thin-walled, yellowish in KOH, slightly brownish in Melzer. *Clamp connections* absent.

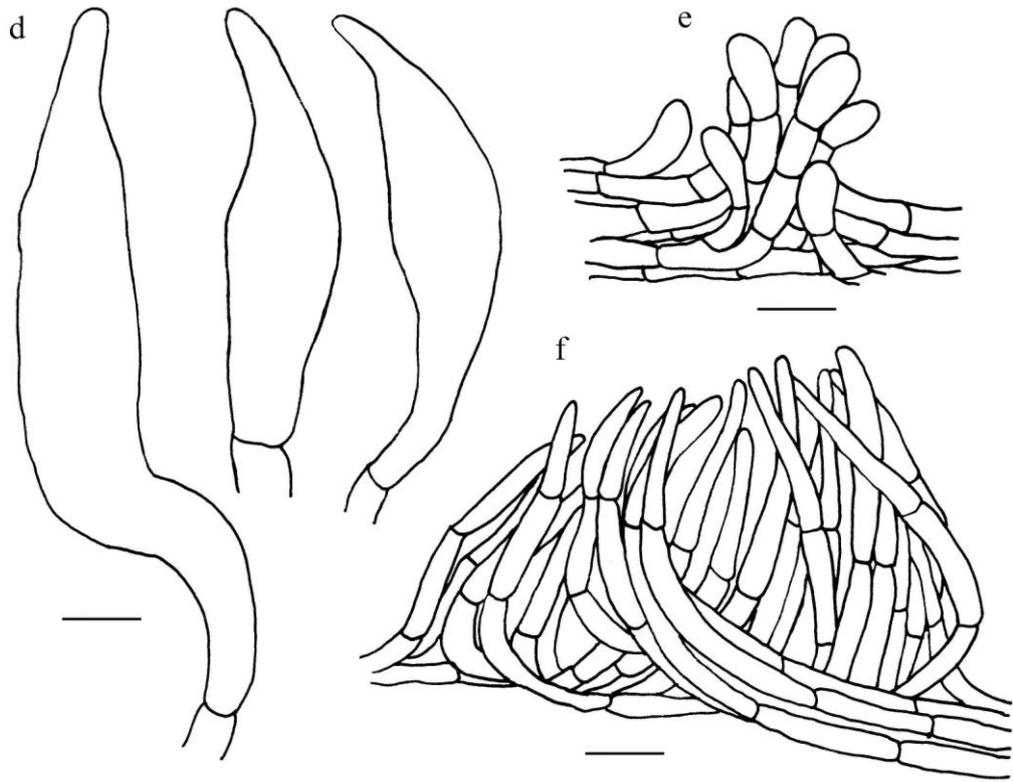
*Habit, habitat, and distribution:* solitary on soil in highland forests of Dipterocarpaceae and Fagaceae.

*Material examined:* MALAYSIA. Pahang, Fraser's Hill, Jeriau waterfall road, N 03°43'36.0", E 101°43'00.6", 1038 m, 8 January 2006, leg. Sumaiyah A. (KLU-M1125).

*Commentary:* The large solitary basidiome recalls this complex of species. The bitter taste and changing context are also consistent and so are the presences of some pale violet to lilac pigments. However, KLU-M1125 has a conspicuous reticulum confined to the apex. Of these large tylopili, the well-known taxa with reticulate stipes are mild tasting, except for *T. felleus* which appears confined to conifer forests in the northern hemisphere.



**Figure 4.29a:** *Tylopilus rubrobrunneus* group (KLU-M1125). **a.** Basidiomes. **b.** Basidiospores. **c.** Basidia. Bars: a = 10 mm. b – c = 10 μm.



**Figure 4.29b:** *Tylopilus rubrobrunneus* group (KLU-M1125). **d.** Hymenial cystidia. **e.** Stipitipellis. **f.** Pileipellis. Bars: d = 10  $\mu\text{m}$ . e – f = 20  $\mu\text{m}$ .

30. *Tylopilus plumbeoviolaceus* (Snell & E.A. Dick) Snell & E.A. Dick. Mycol. 33:33.

1941.

(Fig. 4.30, Plate 4.4 D)

≡ *Boletus plumbeoviolaceus* Snell & E.A. Dick. Mycologia 33:32.1941.

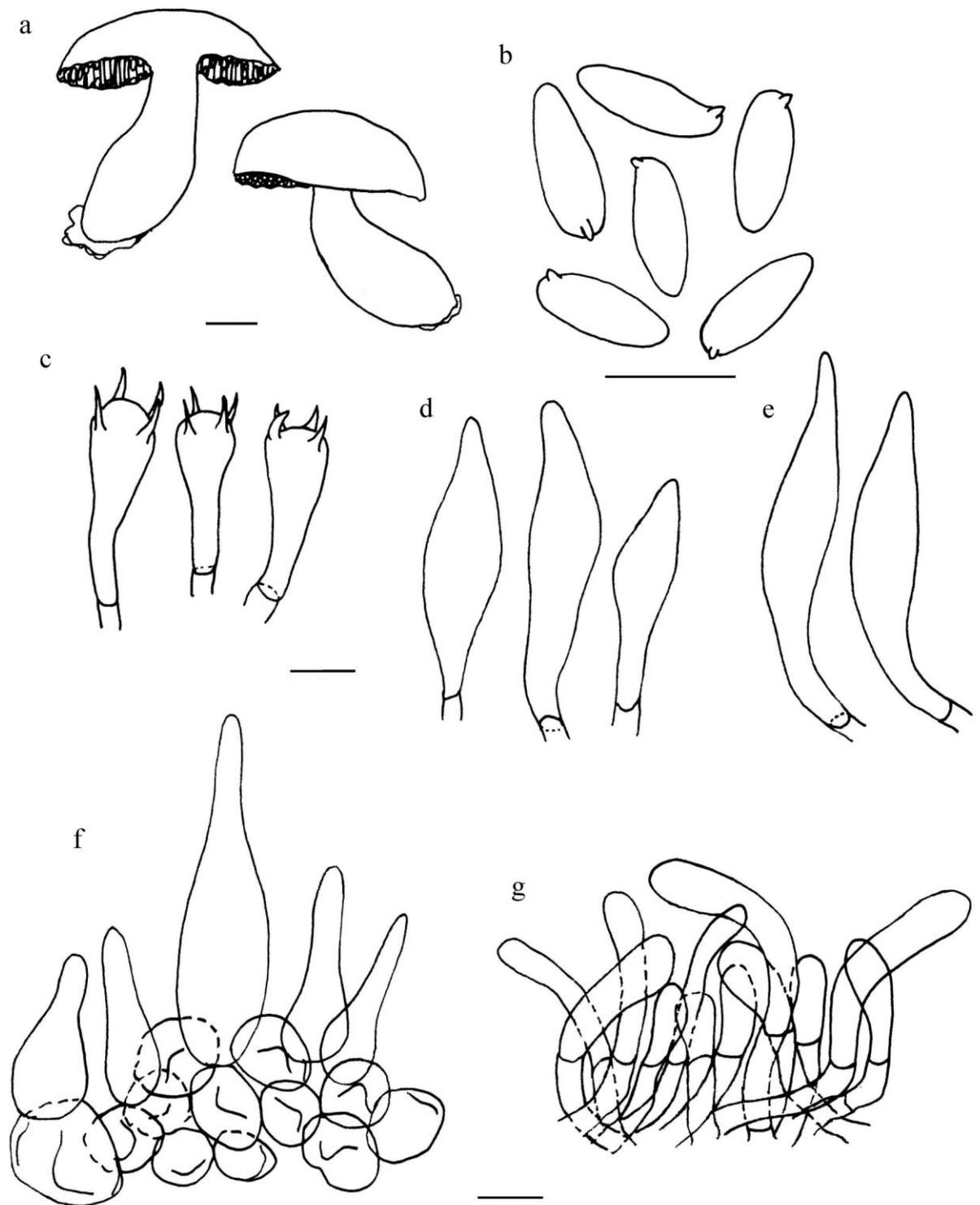
*Pileus* 41 mm broad, convex, dry and smooth, with even margin when mature, purple brown to purple (10E5, 10E4), not bruising. *Context* 6 mm thick, soft and spongy, white, no colour changed when exposed. *Hymenophore* 6 mm thick, tubulose, adnate, white, with concolorous fine pores, 2 – 3 pores per mm, not bruising. *Stipe* 34 mm long, 9 mm broad at apex, 14 mm broad at base, central, clavate, dry velvety, concolourous with pileus, no colour changed when bruised, with white basal mycelium; interior white, solid fresh. *Taste* bitter.

*Basidiospores*  $8.8 - 11.2 \times 3.2 - 4 \mu\text{m}$  [ $x_m = 9.69 \pm 0.78 \times 3.55 \pm 0.43 \mu\text{m}$ ,  $Q = 2.14 - 3.27$ ,  $Q_m 2.76 \pm 0.28$ ,  $n = 30$  spores per one collections], ellipsoid, smooth, dextrinoid, olive in KOH. *Basidia*  $21.6 - 30.4 \times 6.4 - 8.8 \mu\text{m}$ , clavate, 4-sterigmate, length of sterigmata 1.5-2  $\mu\text{m}$ , hyaline, inamyloid. *Cheilocystidia*  $31.2 - 44 (-47.2) \times 6.4 - 8 \mu\text{m}$ , ventricose, hyaline, inamyloid, thin-walled. *Pleurocystidia*  $30.4 - 42.4 (-47.2) \times 6.4 - 8 \mu\text{m}$ , ventricose, thin-walled, hyaline, inamyloid. *Hymenophoral trama* divergent (boletoid), with hyaline elements, inamyloid. *Pileipellis* a trichodermium, composed of erect cylindrical elements,  $40 \times 5 \mu\text{m}$ , smooth, thin-walled, hyaline, inamyloid. *Stipitipellis* with ventricose caulocystidia and sphaerocyst hyphae,  $15.2 - 17.6 \mu\text{m}$  in broad, thin-walled, inamyloid and hyaline in KOH. *Clamp connections* absent.

*Habit, habitat, and distribution:* solitary on soil in forests of Dipterocarpaceae and Fabaceae (*Acacia mangium*); reported from the Western Pacific.

*Material examined:* MALAYSIA. Pulau Pinang, Balik Pulau, N 05°21'12.9", E 100°11'59.1", 450 m, 15 November 2005, leg. Noramly M. (KLU-M1126).

*Commentary:* The purplish brown to dark brown cap and the purplish stem, combined with bitter taste, are distinctive. *Tylopilus plumbeoviolaceus* is mycorrhizal with hardwoods, and widely distributed in the east of Rocky Mountains from Canada to Mexico (Kou, 2004). This species is a first report from Malaysia.



**Figure 4.30:** *Tylopilus plumbeoviolaceus* (Snell & E.A. Dick) (KLU-M1126). **a.** Basidiomes. **b.** Basidia. **c.** Basidiospores. **d.** Pleurocystidia. **e.** Cheilocystidia. **f.** Stipitipellis. **g.** Pileipellis. Bars: a = 10 mm. b – g = 10  $\mu$ m.



**Plate 4.4:** A: *Tylopilus rubrobrunneus* group (top view) ( $\times 10.0$ ); B: *Tylopilus alboater* ( $\times 4.1$ ); C: *Tylopilus nigropurpureus* ( $\times 5.3$ ); D: *Tylopilus plumbeoviolaceus* ( $\times 2.7$ ); E: *Tylopilus favo-ballouii* ( $\times 5.5$ ); F: *Boletus patouillardii* ( $\times 1.0$ ); G: *Tylopilus virens* ( $\times 1.3$ ); H: *Tylopilus ballouii* ( $\times 4.0$ ). Bar = 10mm.

## 5.0 RECOMMENDATIONS AND CONCLUSIONS

In Malaysia, the documentation of Boletaceae based on previous shows that the diversity boletes is poorly known. Further, over the period 1970 to 1992, natural forest in the whole Malaysia was reduced by 19.3 percent, mainly conversion to the agricultural crops, oil palm and rubber (Malaysia's National Policy on Biological Diversity, 1998). This means loss of natural habitats of boletes which are mycorrhizal in large trees. Therefore, there is an urgent need to undertake intensive biological resource inventories and systematic studies to document the boletes, to assess its direct and indirect values and identify the potential threats to biological diversity loss.

In future studies the molecular characterization and phylogenetic relationships should be assessed to support the species concepts. The inclusion of additional taxa and more collections especially from East Peninsular Malaysia are necessary to document the Boletaceae of Malaysia.

In conclusion, total of 52 collections were identified to 30 different species of boletes in West Peninsular Malaysia. The collections included *Afroboletus*, *Aureoboletus*, *Austroboletus*, *Boletellus*, *Boletus* (including *Xerocomus*), *Phylloporus*, *Pulveroboletus*, *Strobilomyces*, *Tylopilus* and *Xanthoconium*. Out of the 30 species that have been identified, four species namely *Afroboletus malaysiancus*, *Phylloporus megaporinus*, *Pulveroboletus claroflavus* and *Tylopilus favo-ballouii* were documented as new to science while another three species - *Tylopilus plumbeoviolaceus*, *Tylopilus rubrobrunneus* group and *Xanthoconium violaceofuscus* were documented as new records in Malaysia. Further, two species, *Afroboletus malaysiancus* and *Xanthoconium violaceofuscum* were first records for Malaysia. Twenty-two species reported by other researchers were re-collected and one specimen of *Phylloporus* sp. has not been identified to species level due to the single specimen collected. It is similar to the taxon from Australia which remains as *Phylloporus* sp. 1.

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## APPENDICES

### Appendix - Buffers and reagents

#### Potassium hydroxide (KOH)

Potassium hydroxide	3 g
Distilled water	97 ml

#### Melzer's reagent

Iodine	1.5 g
Potassium-Iodide	5 g
Chloral Hydrate	100 g
Distilled water	100 ml

#### Ammonium hydroxide solution (NH<sub>4</sub>)OH

Ammonium hydroxide	10 ml
Distilled water	90 ml