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Posgrado en Ciencias Biológicas  
Orientación Sistemática Vegetal



**SISTEMÁTICA DE *Ganoderma***  
**(*Fungi, Basidiomycota, Ganodermatales*):**  
**ASPECTOS MORFOLÓGICOS, MOLECULARES Y**  
**QUÍMICOS**

TESIS

Presentada como requisito parcial para obtener el grado de

**DOCTORA EN CIENCIAS BIOLÓGICAS**  
**ORIENTACIÓN SISTEMÁTICA VEGETAL**

Presenta

**MABEL GISELA TORRES TORRES**

Las Agujas, Zapopan, Jalisco, 3 de agosto de 2007

**SISTEMÁTICA DE *Ganoderma***  
**(*Fungi, Basidiomycota, Ganodermatales*):**  
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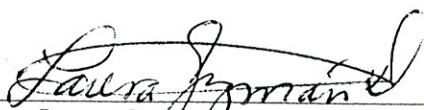
Por

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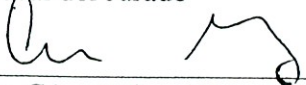
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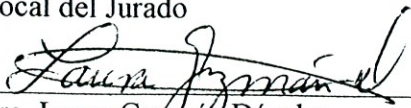
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**ACTA DE REUNIÓN DE LA JUNTA ACADÉMICA**

En las instalaciones del Instituto de Botánica del Departamento de Botánica y Zoología del CUCBA, el día 28 de junio a las 10:00 hrs se reunió la Junta Académica Extraordinaria de Posgrado del CUCBA con la finalidad de analizar la solicitud de designación de sinodales para examen de grado de la alumna del Doctorado en Ciencias Biológicas, **Mabel Gisela Torres Torres**, por lo que después de revisar su propuesta y analizar los candidatos que se proponen se:

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## RESUMEN

En la familia *Ganodermataceae* se han descrito ocho géneros, de los cuales *Amauroderma*, *Ganoderma*, *Haddowia* y *Humphreya* han sido aceptados por autores modernos. Los límites taxonómicos entre los géneros no son claros y muchas especies continúan con una posición taxonómica dudosa. La particularidad de la familia es la presencia de basidiosporas con doble pared y exosporium ornamentado. De acuerdo con el concepto actual, *Ganoderma* tiene la superficie del pileo opaca o brillante, sistema hifal trimítico y basidiosporas con pilares libres a anastomosados y con poro germinal. El género presenta una amplia distribución, principalmente tropical y tiene gran interés fitopatológico y medicinal. En este trabajo se realizaron análisis filogenéticos con caracteres morfológicos y moleculares para conocer sus relaciones inter e infragenéricas. *Ganoderma* presenta un gran número de triterpenos, por esta razón se analizaron caracteres químicos para probar su utilidad en estudios filogenéticos y taxonómicos. Se estudiaron más de 400 especímenes de 43 especies, incluyendo 32 tipos, de *Ganoderma* y géneros relacionados; además de ejemplares del grupo externo. Se construyó una matriz con 45 caracteres morfológicos para 51 especies. Para el análisis filogenético se secuenció la región del ITS del DNAr de 31 especímenes de *Ganodermataceae* y dos del grupo externo. Los análisis filogenéticos se realizaron usando máxima parsimonia, en el programa PAUP 4.0b10. En el estudio de triterpenos se analizaron 55 especímenes de *Ganoderma* y géneros relacionados; los extractos crudos fueron analizados por Espectrometría de Gases acoplado a Masas. En el estudio macro y micromorfológico se encontraron como caracteres novedosos la estructura y color del contexto, estructura de las basidiosporas y células del pileipellis, ya que fueron útiles en la taxonomía y filogenia de *Ganoderma*. Como resultados taxonómicos se describió una nueva especie, se aclaró la posición taxonómica de 16 especies y se citan nuevos registros para Brasil, Cuba, EUA y México. En cuanto a los análisis filogenéticos, con los datos morfológicos no se obtuvo una resolución completa del cladograma, específicamente dentro del clado de *Ganoderma*. Sin embargo, *Amauroderma*, *Elfvigia*, *Ganoderma*, *Haddowia*, *Humphreya* y *Tomophagus* fueron soportados con un alto valor de bootstrap, lo cual sugiere que éstos son géneros válidos. Con datos moleculares no se obtuvo resolución pero cuatro clados fueron soportados con valor de bootstrap: *Elfvigia*, *Humphreya*, *Magoderma* y *Tomophagus*. Las especies que corresponden a *Ganoderma* se separaron en varios clados, algunos de ellos con soporte estadístico. Los grupos formados son congruentes con las características morfológicas. Los resultados sugieren que *Ganoderma* esta conformado sólo por especies con superficie brillante, pileipellis tipo crustotricodermis, basidiosporas con ápice nunca obtuso y con pilares libres a anastomosados. En los análisis de los triterpenos se logró establecer que muchos compuestos permanecen estables aun después de que los especímenes son herborizados y que hay al menos 15 triterpenos que pueden ser usados con fines taxonómicos y filogenéticos.



## INTRODUCCIÓN

### A. Antecedentes

El género *Ganoderma* P. Karst. agrupa organismos que se caracterizan por presentar basidiomas anuales, bianuales o perennes; pileados, dimidiados a flabeliformes, sésiles, estipitados central o lateralmente. La superficie del pileo es lisa, opaca o brillante con una cutícula distintiva, de color café en varios tonos a púrpura oscuro; los poros son de pequeños a medios, con borde entero; los tubos son generalmente estratificados, blanquecinos, ocráceos a color café; el contexto va de blanquecino a color café. El sistema hifal es trimítico; las hifas generativas son de pared delgada, presentan fíbulas, hialinas a amarillentas y muy difíciles de observar; las hifas esqueléticas son de pared gruesa a sólida, amarillas a color café claro y pueden ser arboriformes (se ramifican profusamente en el ápice, con una parte basal no ramificada y con los extremos ramificados adelgazados hacia la punta) o aciculiformes (no ramificadas con el extremo agudo); las hifas conectivas de pared gruesa a sólida, ramificadas, hialinas a amarillentas. Los cistidios son infrecuentes, difíciles de observar en especímenes secos. Las basidiosporas son truncadas, de grandes a muy grandes (7-25  $\mu\text{m}$  de longitud), amarillas a color café, ovoides a elipsoides, con un delgado perisporium, exosporium ornamentado con pilares y un endosporium muy grueso (Furtado 1965, Bazzalo & Wright 1982, Corner 1983, Núñez & Ryvarden 2000, Torres-Torres *et al.* 2007).

*Ganoderma* presenta distribución mundial, desde zonas templadas hasta climas tropicales. Crecen sobre madera o raíces enterradas, en la que ocasionan podredumbre blanca. Algunas especies pueden actuar como parásitas facultativas de cultivos perennes, como: caucho, té, palmas aceiteras, café y árboles de madera dura, o ser saprobias de una gran variedad de hospederos: *Acacia*, *Acer*, *Cocos*, *Ocotea*, *Pinus*, *Quercus*, *Robinia*, *Ulmus*, entre otros (Bazzalo & Wright 1982, Miller *et al.* 1995, Seo & Kirk 2000).

Karsten (1881) describió el género *Ganoderma* con base en *Polyporus lucidus* W. Curt.: Fr., caracterizado por presentar el pileo y estípites laqueados (Telleira 1980). El mismo autor en 1889 erigió el género *Elfvíngia* P. Karst., para incluir las especies no laqueadas, con *Boletus applanatus* Pers. como especie tipo (Ryvarden 2000). El género *Ganoderma* fue aceptado por Patouillard (1889), quien hizo un análisis más amplio, aportando como carácter distintivo la particularidad del ápice truncado de las basidiosporas.

Hasta 1971, *Ganoderma* se consideraba dentro de la familia *Polyporaceae*; Donk lo clasificó en la familia *Ganodermataceae* (Donk, 1971). Las características distintivas que se tomaron en cuenta para segregar esta familia fueron: la presencia de esporas ornamentadas, de doble pared, e hifas esqueléticas ramificadas o aciculiformes (Donk, 1964, 1971). Se han descrito ocho géneros dentro de la familia: *Amauroderma*, *Elfvíngia*, *Ganoderma*, *Haddowia*, *Humphreya*, *Magoderma*, *Tomophagus* y *Trachyderma* (Karsten 1881, Murrill 1905, Imazeki 1939, Steyaert 1972). Algunos de estos géneros no han sido aceptados y las especies se siguen considerando dentro de *Ganoderma*. *Elfvíngia* es aceptado a nivel de subgénero de *Ganoderma*, para las especies con superficie

del píleo opaca. *Trachyderma* se considera un nombre inválido (Moncalvo & Ryvardeen 1997) y *Tomophagus* es considerado como sinónimo de *Ganoderma* por la mayoría de los autores.

Aunque Furtado (1965) consideró que la característica de presencia de laca en la superficie del píleo no era un criterio de clasificación de las especies en *Ganoderma*, debido a que algunos especímenes la pueden perder por acción del clima, sol, o la edad, autores como Corner (1983) y Ryvardeen (1991) la han utilizado. Estos autores han referido a las especies con píleo laqueado como del complejo *Ganoderma lucidum* y a las especies con píleo sin laca y por lo tanto opaco como del complejo *G. applanatum*; denominados como los subgéneros *Ganoderma* y *Elfvingia*, respectivamente. Esta clasificación ha sido sustentada con estudios sistemáticos de isoenzimas (Gottlieb *et al.* 1998, Smith & Sivasithamparam 2000a) y secuencias de DNA (Gottlieb *et al.* 2000, Moncalvo *et al.* 1995a, b, Moncalvo 2000, Hong & Jung 2004). Por otro lado, de acuerdo con características de la estructura de la superficie del píleo, Steyaert (1980) estableció cuatro subgéneros (*Ganoderma*, *Elfvingia*, *Plecoderma* Steyaert, *Anamixoderma* Steyaert) y dividió el subgénero *Ganoderma* en dos secciones (*Ganoderma* y *Characoderma* Steyaert). La clasificación de Steyaert (1980) no ha sido apoyada por estudios modernos.

El género ha sido estudiado por diversos autores. Por ejemplo, Patouillard (1889) listó 48 especies de distribución mundial; Murrill (1902, 1908) publicó una sinopsis de las especies conocidas hasta ese entonces de Norte América y en su publicación de poliporaceos tropicales describió algunas de América tropical (Murrill 1915). Furtado (1967) publicó comentarios sobre especies de Brasil de contexto pálido; Steyaert (1972) hizo una revisión de las especies depositadas en los herbarios de Bogor y Leiden. Telleira (1980) consideró cinco especies para España, las cuales determinó con base en la naturaleza del píleo, presencia del estípote y aspecto del contexto. Bazzalo y Wright (1982) hicieron un estudio de las especies del complejo *Ganoderma lucidum* conocidas de Argentina; Corner (1983) realizó la descripción y comentarios de algunas especies tropicales. Zhao *et al.* (1983, 1984) realizaron un estudio taxonómico de la familia *Ganodermataceae* en China, en donde incluyeron especies tropicales. Gilbertson y Ryvardeen (1986) realizaron un compendio de las especies de Norte América. Moncalvo y Ryvardeen (1997), en un estudio nomenclatural de *Ganodermataceae*, registraron 290 nombres, muchos de los cuales son probablemente sinónimos taxonómicos. Ryvardeen (2000) registró seis especies de América tropical, entre las que describió dos especies nuevas. Recientemente, Moradali *et al.* (2007) realizaron un estudio sobre *Ganoderma* en Irán. Sin embargo, a pesar de que se han realizado numerosos estudios, existen todavía muchos conflictos taxonómicos y nomenclaturales en el género, en parte debido a la poca profundización en el estudio morfológico de manera sistemática.

En la última década se han propuesto varias alternativas para una mejor comprensión y conocimiento del género, las cuales han incluido análisis filogenéticos con isoenzimas o marcadores moleculares, además de pruebas bioquímicas, y más recientemente determinación de especies con metabolitos secundarios (Su *et al.*, 2002), aunque este último aún se encuentra en sus primeras fases. Gottlieb *et al.* (1998) y Smith & Sivasithamparam (2000a) realizaron un

estudio de perfiles isoenzimáticos para discriminar taxa específicos del sur de América del Sur y Australia, respectivamente, en los cuales tuvieron resolución a nivel de subgéneros: *Ganoderma* y *Elfvigia*. También se han utilizado marcadores como los nucleosidos (adenosina, uridina) y bases (uracilo), a través de la aplicación de cromatografía electrocinética micelar, para discriminar especies medicinales a partir de extractos de *G. lucidum* (Curtis) P. Karst., *G. japonicum* (Fr.) Sawada y "*G. capsules*" (Cheung *et al.*, 2001). Kimura *et al.* (2002) aislaron fracciones de triterpenos de cuerpos fructíferos de *G. lucidum* con diferentes actividades antimetastásicas y antitumorales. Un aldehído considerado como un nuevo compuesto fue aislado y elucidado por resonancia magnética nuclear y espectrometría de masas en *G. applanatum* por Ming *et al.* (2002). El análisis de compuestos triterpenoides por HPLC permitió discriminar entre especies de *G. lucidum* y *G. tsugae*; además de redeterminar especies de *G. resinaceum* mal nominadas por caracteres macromorfológicos (Su *et al.*, 2002). Se han aislado polisacáridos y triterpenos oxigenados, entre los que se determinaron estereoisómeros o isómeros posicionales (Lin *et al.* 1991, Lin *et al.* 1995, Choi & Sa 2000, Zhu *et al.* 2000, Kimura *et al.* 2002, Cao & Lin 2003, Shiao (2003).

La monofilia del género ha sido comprobada por análisis filogenéticos con caracteres moleculares, el cual incluye a los subgéneros *Ganoderma* y *Elfvigia*. Moncalvo *et al.* (1995b), Gottlieb *et al.* (1998, 2000), Moncalvo (2000) y Smith & Sivasithamparam (2000b) utilizaron la región interna transcrita (ITS) del DNA ribosomal (DNAr); la cual permitió segregar grupos; sin embargo, se presentaron algunas discrepancias en las relaciones filogenéticas, porque muchas de las especies han sido erróneamente determinadas. Moncalvo (2000) además utilizó la secuencia del gen de la manganeso superóxido-dismutasa, que no resolvió algunas relaciones entre las especies. Gottlieb *et al.* (2000) estudiaron especies sudamericanas definidas morfológicamente, a través de polimorfismos de conformaciones monocatenarias (SSCP) acopladas a PCR y variación en la secuencia del ITS, en donde encontraron relaciones interesantes, pero no pudieron resolver todas las relaciones infragenéricas. Sokol *et al.* (1999) determinaron que *G. australe* (Fr.) Pat. y *G. adpersum* (Schulzer) Donk, por un lado, y *G. pfeifferi* Bres. y *G. resinaceum* Boud., por el otro, son especies independientes con base en el análisis de la región ITS. Moncalvo (2000) y Hong & Jung (2004) incluyeron por primera vez en sus análisis a *G. colossus* (Fr.) C.F. Baker y *G. tsunodae* Yasuda; especies de adscripción dudosa, las cuales quedaron basales o como grupo hermano de *Ganoderma*. Otras especies con posición taxonómica dudosa no han sido incluidas hasta ahora en ningún análisis. Por otro lado, no se ha realizado ningún análisis filogenético con datos morfológicos.

## **B. Justificación**

Con base en los caracteres morfológicos usados hasta ahora, las relaciones filogenéticas y taxonomía del género *Ganoderma* no han podido ser resueltas, lo que ha originado una gran multiplicidad de nombres; inclusive varias especies han sido descritas nuevamente y otras han sido colocadas bajo el mismo nombre. La plasticidad existente en los caracteres macromorfológicos, la similitud de los caracteres micromorfológicos, la deficiencia de descripciones modernas, la no

disponibilidad de buen material, así como el desconocimiento de la literatura, son quizá parte de las explicaciones del por qué la proliferación de nombres. Estas, entre otras razones, no han permitido un conocimiento extenso del género y en especial de las especies tropicales de América, en donde se han realizado pocos estudios taxonómicos. Por otro lado, la interpretación y discusión de las relaciones filogenéticas generada con base en caracteres moleculares no es muy completa, debido al poco conocimiento que se tiene sobre las especies; es así como una misma especie aparece en diversos clados sin muchas explicaciones posibles.

El estudio de las relaciones filogenéticas entre las especies es una alternativa para una mejor comprensión del género. En general se han realizado sólo con especies europeas, incluyendo pocas especies tropicales. Por otro lado, los estudios filogenéticos en su mayoría han sido realizados con caracteres moleculares, sin que otros caracteres hayan sido explorados a profundidad.

En el presente estudio se planteó realizar un análisis sistemático del género *Ganoderma*, considerando en particular especies poco estudiadas del continente Americano, a través de la revisión de especímenes depositados en los herbarios y recién recolectados. Para el estudio se utilizaron datos macro y micromorfológicos, así como caracteres moleculares; además se estudió la estabilidad de triterpenos como posible fuente de caracteres químicos en la taxonomía de *Ganoderma*.

### **C. Objetivos**

#### **a. Objetivo General**

Realizar un estudio sistemático del género *Ganoderma*, con el fin de contribuir al conocimiento de las relaciones infragenéricas, como una estrategia hacia su aprovechamiento y conservación.

#### **b. Objetivos específicos**

1. Analizar caracteres macro y micromorfológicos importantes en el estudio sistemático del género.
2. Buscar nuevos caracteres morfológicos útiles en el análisis filogenético y taxonomía del grupo.
3. Determinar si las secuencias del ITS del DNAr son caracteres moleculares útiles en el análisis filogenético.
4. Realizar análisis filogenéticos con los caracteres propuestos.
5. Determinar estabilidad de caracteres químicos que pudieran tener uso potencial en el análisis filogenético.
6. Contribuir al conocimiento taxonómico y a la comprensión de la distribución del género en América.

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## CAPÍTULO I

### ESTUDIO MORFOLÓGICO Y TAXONOMÍA DE *GANODERMA*

#### RESUMEN

Tradicionalmente, los caracteres macro y micromorfológicos han sido utilizados para la delimitación a diferentes niveles taxonómicos y en diferentes grupos de hongos. Cada grupo taxonómico está definido por un conjunto particular de caracteres; que puede ser diferente al que define a otro, ya que los caracteres pueden variar drásticamente de un grupo a otro, así como su nivel de importancia. En *Ganoderma* los caracteres morfológicos han sido muy conflictivos para la delimitación de las especies, debido a que se han aplicado conceptos muy amplios y erróneos para definir a las especies, usando caracteres diferentes bajo el mismo nombre específico. Esto ha creado una gran confusión aunado a la plasticidad que muchos de ellos presentan. Otro inconveniente en el estudio morfológico es que en muchas especies sólo se conoce el tipo, por lo que no se ha podido profundizar en la variabilidad y congruencia de algunos caracteres.

Con el fin de conocer el conjunto de caracteres que definen a *Ganoderma*, en este trabajo se realizó un estudio morfológico, mediante la revisión de aproximadamente 400 especímenes de todo el mundo, incluyendo 32 tipos en su mayoría de las especies tropicales y subtropicales. Los materiales fueron recolectados por la autora, estaban depositados en el Herbario IBUG o fueron solicitados en préstamo a los herbarios BPI, BR, COL, ENCB, FH, H, INBIO, K, NY, O, PC, SP, UPS, VEN, XAL.

Este capítulo consta de dos artículos previamente publicados, tres artículos en revisión para su publicación y tres manuscritos de artículos en revisión, todos relacionados con la descripción de caracteres morfológicos y su aplicación en la resolución de problemas taxonómicos. Se proponen caracteres morfológicos novedosos, además de los usados tradicionalmente por otros autores y se retoman otros que no han sido usados de manera sistemática. Se describe una nueva especie, se reportan 40 nuevos registros, diez de ellos son registrados por segunda ocasión para el mundo. Además, se elaboraron las descripciones de 20 especies que no tenían descripción moderna y se complementó redescubrieron tres especies. *Ganoderma areolatum* fue excluida de *Ganoderma* y sinonimizado con *Navisporus floccosus*.



Notas sobre la variación morfológica de *Ganoderma curtisii*  
(Ganodermatales, Ganodermataceae) en México

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Morphological variation of *Ganoderma curtisii* (Ganodermatales,  
Ganodermataceae) in Mexico

**Abstract.** A macro and micromorphological study of selected *Ganoderma curtisii* specimens collected in Mexico were done. Traditional taxonomic methods were used, making dissections through a trajectory from the base to the border of the pileus. The macromorphological variation according to the state of maturation, presence or absence of shine and color of the pileus, and form, color and size of the stipe, was described; some relationships were established with the microscopic structures: spores, structure of the pilear surface and hyphal system. From the observed relationships, the most relevant was the yellow to orange-yellow pileus without shine related with the cuticle poor developed, with cells not well-formed, thin-walled or absent, present in young adult, immature or intemperized specimens. In adult specimens, the cuticle cells from pileus margin are hyaline, with a very visible lumen and generally septate, different from the cuticle cells from basidiomata center, which are yellow to yellow-brown and thick-walled.

**Key words:** cuticle cells, pilear surface, spores, micromorphology

**Resumen.** Se realizó un estudio macro y micromorfológico de especímenes seleccionados de *Ganoderma curtisii* recolectados en México. Se usaron métodos de taxonomía tradicional, a través de disecciones trazando una trayectoria desde la base hacia la periferia del píleo. Se describe la variación macromorfológica de acuerdo con el estado de madurez, color, presencia y ausencia de brillo del píleo, y forma, color y tamaño del estípite; se establecen algunas relaciones con las estructuras microscópicas: estructura de la cubierta de la superficie pilear y sistema hifal. De las relaciones observadas, la más relevante fue el píleo amarillo a amarillo-anaranjado y sin brillo con la cutícula mal desarrollada, con células mal formadas, de pared delgada o ausentes, presentes en especímenes adultos jóvenes, inmaduros o intemperizados. En especímenes adultos las células de la cutícula de la periferia del píleo son hialinas, con un lumen muy visible y generalmente septadas, a diferencia de las células de la cutícula del centro del basidioma que son de color amarillo a café-amarillento y de pared gruesa.

**Palabras clave:** células de la cutícula, superficie pilear, esporas, micromorfología.

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Introducción

*Ganoderma curtisii* (Berk.) Murrill es una especie abundante en bosques de *Pinus-Quercus*, en donde crece en el suelo

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como parásito de las raíces de *Quercus*, o sobre madera muerta ocasionando pudrición blanca. Fue citada por Moncalvo y Ryvarden [15] de Norteamérica. Japón, China, India y África. En México ha sido registrada casi de toda la república [2, 3, 8, 9, 11, 15, 16, 17, 18], creciendo desde los

1000 hasta los 2000 msnm. *Ganoderma curtisii* presenta una gran plasticidad morfológica y dentro del género es una de las especies que mayor variación macromorfológica presenta; pero no es claro que factores la ocasionan y si toda esta variación representa una misma especie. Por otro lado, en la literatura se cuenta con pocas descripciones detalladas y no se ha estudiado la variación micromorfológica [10, 16, 18]. Los caracteres microscópicos más estudiados en la delimitación de las especies de *Ganoderma* son las esporas y estructura de la cubierta de la superficie pilear; se han realizado algunos estudios comparativos intra e interespecíficos [1, 5, 7, 10, 19, 20, 21], pero la variación micromorfológica asociado a la morfología macroscópica y al estado de madurez del basidioma dentro de una especie no ha sido evaluada a profundidad. En este trabajo se estudiaron especímenes representantes de la variación morfológica presentada en *G. curtisii*, con el fin de observar los patrones que siguen estas relaciones.

### Materiales y métodos

Se revisaron especímenes depositados en IBUG y ENCB; además se realizaron salidas de campo *ex profeso* para recolecta de material. Para el estudio de la variación morfológica se seleccionaron especímenes con base en los siguientes criterios: estado de madurez del basidioma, color del pileo, color del estípite, permanencia del brillo en el pileo y permanencia de la cutícula. Los cortes se realizaron trazando una trayectoria desde la base del pileo hacia la periferia. Para el estudio macro y micromorfológico de los especímenes se siguió la metodología de Corner [4], Furtado [7], Largent [13] y Largent *et al.* [14]. Los colores de los basidiomas fueron descritos de acuerdo a Korerup y Wanscher [12]. Las descripciones y mediciones de las estructuras microscópicas se realizaron en montajes con hidróxido de potasio (KOH) al 5%, y se observó la reacción

dextrinoide de las esporas e hifas con el reactivo de Melzer. Se midieron al menos 20 esporas de cada espécimen y se calculó el coeficiente Q (longitud/ancho). Los basidiomas fueron fotografiados tratando de representar la mayor variabilidad posible.

## Resultados y discusión

### Descripción de la especie

*Ganoderma curtisii* (Berk.) Murrill, North Amer. Flora 9: 120, 1908.

= *Fomes curtisii* (Berk.) Cooke, Grevillea 13 (68): 118, 1885.

= *Polyporus curtisii* Berk., Hooker's J. Bot. Kew Gard. Misc. 1: 101, 1849.

= *Scindalma curtisii* (Berk.) Kuntze, Revis. gen. pl. (Leipzig) 3: 518, 1898.

Basidioma 40-90 x 55-190 x 10-16 mm, estipitado, corchoso a leñoso. Pileo reniforme, dimidiado o circular, único o cespitoso, generalmente opaco en especímenes inmaduros, con brillo en adultos, que puede perder en su totalidad o quedar remanentes, ocasionalmente conservan el brillo en toda la superficie, amarillo claro (46A), amarillo profundo (4A8), color amarillo-oro (5B8), café-amarillento (5C8), café-violeta (11F8) a café-violeta muy oscuro casi negro, más o menos homogéneo, o con zonaciones de estas tonalidades, en ocasiones cubierto de esporas color canela (6D6); superficie uniforme a ligeramente abollada, ocasionalmente radialmente rugosa, adultos con zonaciones formadas por surcos, glabra; margen entero, a veces lobulado, redondeado a truncado, blanco-amarillento (4A2) a color café claro (7E8). Estípite de 25-270 x 13-70 mm, lateral o excéntrico a ocasionalmente central, aplanado a cilíndrico, sólido; superficie opaca a brillante, lisa, amarilla pálida (3A3), color amarillo-oro (4A4, 5B8) a vino tinto oscuro casi negro, generalmente más oscuro que el pileo, cutícula que se desprende o no. Cutícula brillante, color café-violeta, café-

rojiza a casi negra, que se desprende o no, al desprenderse deja ver superficie amarillo profundo (4A8), ocasionalmente color crema-amarillento (5A2). Contexto de 7-13 mm de grosor, duplex, con una franja amarillo profundo (4A8) inmediatamente después de la cutícula, luego anaranjado pálido a anaranjado claro (5A3-5A4) cerca a la cutícula y color café-sienna (6D7) a excepcionalmente café-amarillento (6F8) cerca de los tubos: corchoso a ligeramente fibroso, con 2 a 4 bandas resinosas, azonado. Poros 2-4 por mm, angulosos a redondeados: superficie de los poros amarilla (2A3), amarilla pálida (3A3) a color amarillo-oro (4A4, 5B8), se mancha de color café (6D8); tubos 3-10 mm de longitud, con o sin estratificación, color café-vináceo (8E4).

Sistema hifal trimitico. Trama del contexto con hifas generativas hasta de 3.2 µm de diámetro, con fibulas, hialinas, difíciles de observar, generalmente colapsadas; hifas esqueléticas de 1.6-5.6 µm de diámetro, arboriformes, pared gruesa a sólida, amarillentas a color café-amarillento, predominantes; hifas conectivas de 3.2-4.8 µm de diámetro, pared gruesa a sólida, amarillentas a color café-amarillento. Cutícula semejante a un himenodermo, células de la cutícula de 43.2-96 x 7.2-16.4 µm, claviformes, con divertículos ocasionales o abundantes, tanto laterales como en el ápice, con ramificaciones laterales, pared sólida a gruesa, a veces multiestratificada, amarillentas, ligeramente amiloides después de 24 horas en reactivo de Melzer; hifas generativas de 2.4-3.2 µm de diámetro, fibulas conspicuas, hialinas a amarillentas, difíciles de observar, generalmente colapsadas; hifas esqueléticas de 1.6-4.8 µm de diámetro, arboriformes, pared gruesa a sólida, amarillas a color café-amarillento, predominantes; hifas conectivas de 1.6-4.8 µm de diámetro, pared gruesa a sólida, amarillentas a color café-amarillento. Basidiósporas de (9.2-)10.4-12.8(-13.6) x 5.6-8 µm, Q = 1.5-1.88, elipsoides a oblongas, ápice truncado; doble pared (0.5-0.8 µm de grosor), la externa rugosa, amarillo-rojiza; la interna rugosa, gruesa, color café-rojizo, con columnelas interparietales, de 0.5-0.64 µm de diámetro, anastomosadas;

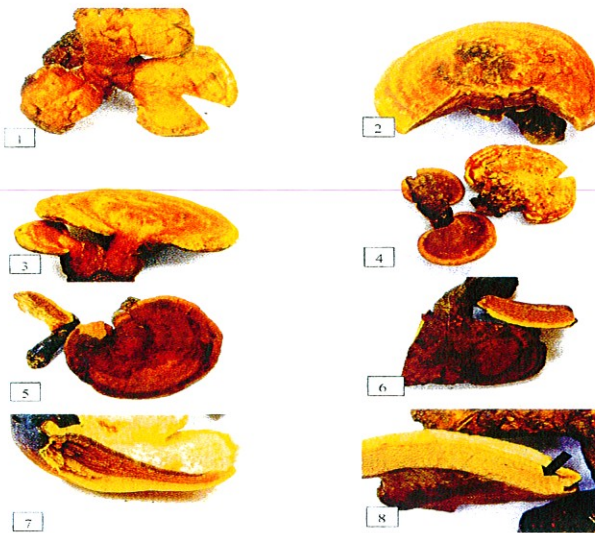
con poro apical y apéndice hilar visible; color café-amarillento, inamiloides. Basidios de 20 x 7.2 µm, hialinos, escasos. Cistidios y otros elementos estériles ausentes.

Hábitat. Solitario o gregario, en bosques de pino, encino, encino-pino y mesófilo de montaña con encinos, altitud de 1600-2170 m.s.n.m.

Material estudiado. JALISCO, Municipio de Colotlán, 16.5 km al O de Colotlán por el camino a Carrizal, agosto 2, 2004, *M.G. Torres-Torres 541* (IBUG); Municipio de Zapopan, Bosque La Primavera, aproximadamente a 8 km de la entrada por prolongación Mariano Otero, julio 20, 2004, *M.G. Torres-Torres 526* (IBUG); Municipio de Mazamitla, 5 km al O de Mazamitla, Los Cazos, febrero 15, 1994, *H. Orozco 5* (IBUG); a 5 km de la Manzanilla de La Paz rumbo a Mazamitla, octubre 6, 1984, *L. Guzmán-Dávalos 1723* (IBUG); Municipio de Mezquitic, La Cebolleta, agosto 15, 1997, *L. Villaseñor-Ibarra 282* (IBUG); Municipio de Cuquio, Las Cruces, octubre 12, 1980, *J. Mejía-Jiménez s.n.* (IBUG); Municipio de Cuautitlán, brecha Las Joyas-Manantlán, septiembre 23, 1983, *A.G. Valenzuela s.n.* (IBUG); Municipio de Mascota, 800 m después de La Campana, km 83.5 carretera Guadalajara-Mascota, agosto 17, 1998, *L. Guzmán-Dávalos 7447* (IBUG); Municipio de Tequila, Volcán de Tequila, km 8 de la brecha a la estación de microondas, julio 30, 1986, *J.A. Pérez de la Rosa s.n.* (IBUG). HIDALGO, Municipio de Tenango de Doria, Temapa, noviembre 20, 1969, *J. Gimete 152-A* (ENCB). MORELOS, 5 km al O de Tepoztlán, cerca de la autopista a Cuautla, septiembre 3, 1967, *M. Frias Neve 18* (ENCB).

#### Análisis de caracteres morfológicos

Las plasticidad morfológica en *Ganoderma curtisii* puede darse a todos los niveles pero más notoriamente a nivel macroscópico (figs. 1-8). Algunas de estas variaciones pueden ser manifestaciones de la variación de las estructuras microscópicas (Tabla 1, figs. 9-22). Para este trabajo se consideraron que los basidiomas eran adultos cuando



Figuras 1-8. Caracteres y variación morfológica de basidiomas de *Ganoderma curtisii*. 1. pilcos espesos, amarillos a color café-amarillento, opacos (M.G. Torres-Torres 541); 2. pilcos color café-amarillento con zonaciones café-amarillento, opaco, que brillan más oscuro que el pilco central, atemperizado (L. Villaseñor-Ibarra 282); 3. pilco color amarillo-oro, opaco, estipe corto, sin brillo, concoloro a más oscuro que el pilco (J. A. Pérez de la Rosa s.n.); 4. pilco de color café-violeta, café-amarillento a amarillo-oro, de opaco a brillante, estipe corto, lateral, brillante, más oscuro que el pilco (J. Mejía-Jiménez s.n.); 5. pilco color café-violeta, brillante, estipe largo, lateral, brillante (L. Guzmán-Dávalos 1723); 6. pilco cubierto de esporas, color café-violeta casi negro; estipe largo con coloro con el pilco (A. G. Valenzuela s.n.); 7. superficie de los pecos amarilla, contexto duplex (L. Guzmán-Dávalos 7447); 8. la flecha indica las hifas de sustancia resinosa en el contexto (A. G. Valenzuela s.n.).

Tabla 1. Caracteres macro y micromorfológicos de *Ganoderma curtisii*.

Especímenes	Madurez del basidioma	Pilco	Cutícula y superficie pilcar	Células cutícula (µm)	Esporas (µm)
<i>L. Villaseñor-Ibarra 282</i>	Adulto	Opaco, color amarillo-oro con tonos café-amarillento, zonado	Se desprende, superficie crema-amarillenta	36-40 x 5.6-8, pared delgada, protuberancias apicales y laterales, poco diferenciadas	(10-)10.4-12 x 6-7.2, Q 1.44-1.88
<i>J. Mejía-Jiménez s.n.</i>	Adulto	Brillante, en algunas partes el brillo se pierde, color café-violeta cerca de la base, luego color café-amarillento a amarillo-oro, sin zonación	Se desprende, superficie amarilla	68-88 x 8.8-16.4, las de la periferia con pared subgruesa, las maduras pared gruesa, multiestratificada, protuberancias apicales y laterales, ramificaciones laterales	10.4-12.8(-13.6) x 6-7.2, Q 1.55-1.88
<i>M.G. Torres-Torres 526</i>	Adulto	Opaco en la periferia a brillante cerca de la base, color café-violeta en la base, el resto amarillo, zonado	Se desprende, superficie amarilla	72-96 x 8.8-16, pared gruesa, multiestratificada, protuberancias apicales y laterales, ramificaciones laterales	(9.2-)9.6-11.2 x 5.6-6.8(-7.2), Q 1.5-1.75
<i>H. Orco 5</i>	Adulto	Brillo que se pierde, color café-violeta, zonado	No se desprende fácilmente, superficie amarilla	64-88 x 12-16, pared gruesa, multiestratificada, protuberancias apicales y laterales, ramificaciones laterales	9.6-11.2 x 5.6-7.2, Q 1.56-1.63
<i>J. Giménez 152-A</i>	Adulto	Muy brillante, color vino tinto muy oscuro, borde más claro, zonado	No se desprende fácilmente, superficie crema-amarillenta	44-64 x 8.8-12.8, pared gruesa, multiestratificada, protuberancias apicales y laterales, ramificaciones laterales	11.2-12.8 x 6.8-8, Q 1.55-1.88
<i>M. Frias Neve 18</i>	Adulto	Brillante, color café-violeta, zonado	No se desprende	56-72 x 9.6-14, pared gruesa, multiestratificada, sin protuberancias, ni ramificaciones laterales	10.4-11.2 x 6.4-7.2, Q 1.56-1.75
<i>L. Guzmán-Dávalos 1723</i>	Adulto	Brillante, color café-violeta, zonado	No se desprende fácilmente, superficie amarilla	36-64 x 10.4-18.6, pared gruesa, multiestratificada, protuberancias apicales y laterales, ramificaciones laterales	10.4-12 x 6.4-7.2(-7.6), Q 1.44-1.88
<i>A.G. Valenzuela s.n.</i>	Adulto	Opaco, color café-violeta muy oscuro casi negro, zonado	No se desprende fácilmente, superficie crema-amarillenta	56-88 x 9.6-20, pared gruesa, multiestratificada, sin protuberancias, ni ramificaciones laterales	(9.6-)10.4-11.2(-12) x (5.6-)6-7.2, Q 1.63-1.83
<i>J.A. Pérez de la Rosa s.n.</i>	Adulto joven	Opaco, color amarillo-oro, zonado	Mal formada	Indiferenciadas, pared delgada, difíciles de observar	9.6-11.2 x 5.6-6.4, Q 1.63-1.81
<i>L. Guzmán-Dávalos 7447</i>	Adulto joven	Opaco, color café-amarillento cerca de la base a amarillo en la periferia, sin zonación	Se desprende, superficie amarilla	48-72 x 7.2-10, pared delgada a gruesa pero difícil de observar, protuberancias apicales y laterales	9.6-11.2 x 5.6-7.2, Q 1.5-1.69(-1.71)
<i>M.G. Torres-Torres 541</i>	Inmaduro	Opaco, color café-amarillento, sin zonación	Mal formada	No diferenciadas	Inmaduras, escasas

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presentaban esporas completamente desarrolladas; adultos jóvenes cuando tenían esporas desarrolladas, pero el mayor porcentaje era de esporas jóvenes, e inmaduros cuando no presentaban esporas o éstas no eran maduras.

**Pileo.** La variación más visible en el pileo está relacionada con el color y el brillo. El color del pileo puede ser amarillo, café-amarillento (figs. 1, 3), café-violeta (fig. 5) o muy oscuro casi negro (fig. 6), o presentar una combinación de ellos (figs. 2, 4); y pueden ser brillantes o opacos (figs. 1-6). Los especímenes muy jóvenes o intemperizados presentan un pileo de amarillo a color café-amarillento y en la mayoría de los casos no alcanzan a desarrollar la estructura cuticular que confiere el brillo (figs. 1-2).

Los especímenes adultos aunque pueden perder el brillo, no pierden el color originado por los compuestos que conforman la cutícula (fig. 6). Por otro lado, los especímenes jóvenes carecen generalmente de zonaciones marcadas por surcos en la superficie, que son muy marcadas en los especímenes adultos. La cantidad de estas zonaciones también puede variar aunque no se observa un patrón que pueda explicar esta variación.

El contexto duplex (figs. 7, 8) y las bandas resinosa (fig. 8) son características distintivas de *G. curtisii*, ya que son invariables independientemente de la edad y el estado de basidioma. Las bandas resinosa en ocasiones no son visible en la periferia del pileo y los colores del contexto pueden llegar a ser un poco más oscuros, pero no se pierde la característica duplex.

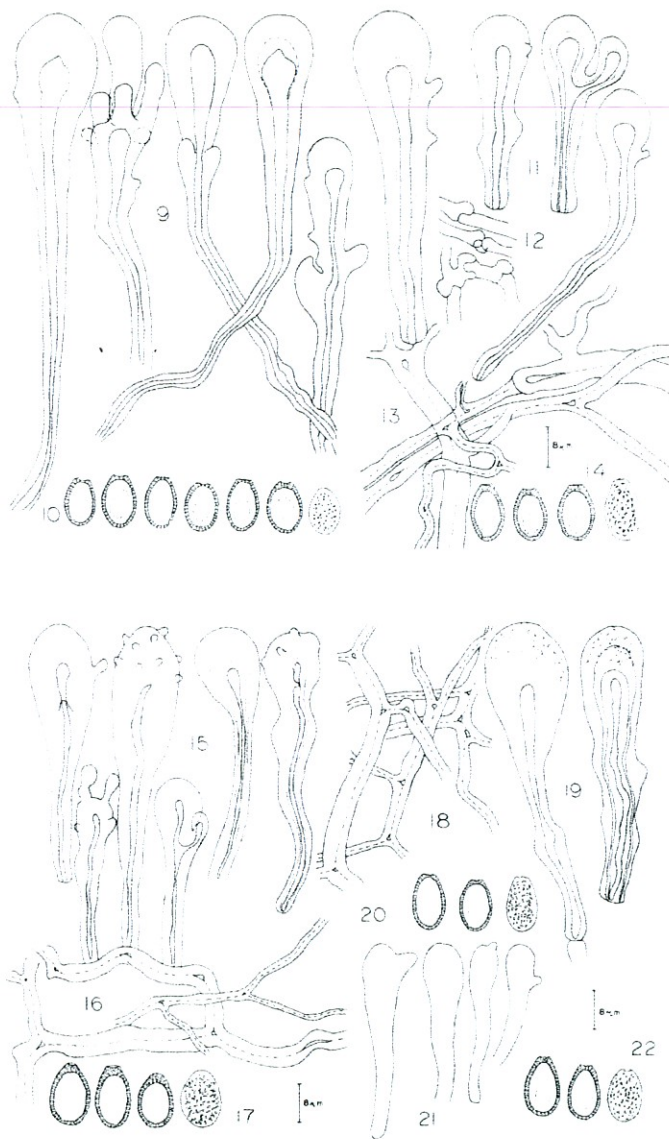
**Cutícula.** En las especies laqueadas del género *Ganoderma*, la capa más externa del pileo ha sido denominada cutícula [4, 6, 10, 18, 19, 20], la cual está compuesta principalmente de células terminales claviforme o cilíndricas, las cuales probablemente son las responsables de secretar una sustancia parecida a la laca, que confiere el brillo al basidioma. En *G. curtisii* la cutícula se puede desprender especialmente en especímenes adultos jóvenes. En éstos, la cutícula es mucho más delgada que en materiales adultos y las

células de la cutícula son generalmente más cortas y de pared no multiestratificada, lo cual posiblemente confiere fragilidad a la estructura de la superficie pilear.

**Estípites.** Su mayor variación en *G. curtisii* se presenta en la longitud y forma, ya que puede ser muy largo o muy corto con relación al diámetro del pileo, además de ser cilíndrico o aplanado y en algunos casos adelgazarse hacia la base. Estos dos caracteres parecen estar asociados con el tipo de sustrato donde crecen; si están enterrados sobre una raíz, el estípites es generalmente largo y cilíndrico, en contraposición con el estípites corto y aplanado cuando crecen sobre madera. Por otro lado, el color puede variar según el estado de madurez e intemperización, siendo notablemente más oscuros que el pileo en especímenes adultos y poco intemperizados (fig. 5).

**Basidiosporas.** Los basidiomas de adultos jóvenes tienden a presentar en promedio esporas maduras más pequeñas. La pared es de 0.5-0.64 µm de grosor (figs. 14, 20, 22), pero el espécimen *J. Gimarc 152-A* presentó esporas ligeramente más grandes, de 11.2-12.8 (-13.6) x 6.8-8 µm, con pared más gruesa, de 0.8 µm, con columnelas más gruesas, con el ápice en ocasiones redondeado y reticulado (fig. 17). El ejemplar *M.G. Torres-Torres 526* presentó esporas con pared más delgada, de menos de 0.5 µm y columnelas más angostas (fig. 10).

**Células de la cutícula.** Su forma básica es de estrecha a ampliamente claviforme o cilíndrica; con o sin protuberancias y ramificaciones (figs. 9, 11, 15, 19, 21). Haddow [10] reportó células de la cutícula de 15-25 µm de longitud, pero en este trabajo fueron significativamente más largas. Ojeda-López *et al* [18] describieron especímenes mexicanos de *G. curtisii* con células enteras, lo mismo que Haddow [10] para materiales de EUA; sin embargo, existe una marcada tendencia a presentar células con protuberancias laterales y apicales, y ramificaciones laterales. Dos especímenes presentaron células sin protuberancias: *A.G. Valenzuela* s.n. (fig. 19) y *M. Frias-Neve 18*; su cutícula



Figuras 9-22. Estructuras microscópicas de *Ganoderma curtisii*. 9: células de la cutícula, 10: esporas. 18: sistema hifal (M.G. Torres-Torres 526). 11: células de la cutícula, 12: hifas generativas, 13: hifas esqueléticas y conectivas, 14: esporas (L. Guzmán-Dávalos 1723). 15: células de la cutícula, 16: hifas esqueléticas y conectivas, 17: esporas (J. Gimete 152-A). 19: células de la cutícula, 20: esporas (A.G. Valenzuela s.n.). 21: células de la cutícula, 22: esporas (L. Villaseñor-Ibarra 282).

quebradiza no permitió realizar buenos cortes, limitando la observación a pocas células, por lo que la evaluación de este carácter no fue completa. Se encontraron diferencias notables de acuerdo con el sitio de corte (centro o periferia) y la madurez del basidioma, con relación al brillo y la presencia y grosor de la cutícula. En especímenes completamente desarrollados las células de la cutícula de la periferia del pileo son generalmente hialinas a amarillentas y la pared es difícil de observar, en ejemplares jóvenes sus células también son difíciles de observar, generalmente amarillentas y con una pared no muy bien desarrollada. Por otro lado, la ausencia de brillo y el pileo completamente amarillo en especímenes inmaduros, o el pileo color amarillo-oro a café-amarillento en materiales adultos pero intemperizados, se relaciona con ausencia de células de la cutícula o muy mal formadas (fig. 21). Las células de la cutícula en ejemplares intemperizados son notablemente más pequeñas (fig. 21). Lo anterior permite conjeturar la relación directa de las células de la cutícula con la producción de la sustancia que confiere el brillo y color a los basidiomas. Además se observó que el tamaño de las células de la cutícula varía de un espécimen a otro y depende del grosor y diferenciación de la cutícula.

Dos especímenes presentaron células de la cutícula con características muy peculiares con relación al resto de los materiales estudiados. El espécimen *J. Gimete 152-A* presentó células de pared más gruesa, con una inusual y excesiva cantidad de protuberancias apicales (fig. 15); además su pileo es muy lustroso y de color vino tinto. El ejemplar *M.G. Torres-Torres 526*, además de la característica de las esporas mencionada anteriormente, tiene células de la cutícula excepcionalmente más largas (fig. 9).

**Sistema hifal.** El diámetro, color y forma de las hifas son más o menos homogéneas en todos los especímenes revisados, sin importar el estado de madurez o las características macroscópicas (figs. 12, 13, 16, 18). Existe una ligera tendencia a que las hifas conectivas sean más visibles en ejemplares jóvenes y en la base del pileo.

## Conclusiones

En México los basidiomas estipitados de *Ganoderma* asociados a los bosques de pino-encino han sido citados como *Ganoderma curtisii*. En este trabajo encontramos que el color y persistencia de la cutícula del pileo, están directamente relacionadas con el tamaño, grosor de la pared y color de las células de la cutícula, lo cual es también probablemente cierto para las otras especies de *Ganoderma* con cutícula laqueada. Tradicionalmente las células de la cutícula para esta *G. curtisii* han sido descritas como enteras; en los materiales revisados se encontró una gran variabilidad con respecto a la presencia de ramificaciones y diverticulaciones. Sin embargo, no existe relación aparente entre éstas y el aspecto macromorfológico. El tamaño de las columnelas de las esporas fue variable entre los ejemplares; este carácter no había sido descrito ni considerado previamente. En conclusión, *Ganoderma curtisii* es una especie muy variable macromorfológicamente, pero es muy probable que parte de la variación micromorfológica presente, corresponda al menos a dos especies. Se requieren de más estudios, en particular con caracteres moleculares, que permitan dilucidar si la variación observada representa una o varias especies.

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## CAPÍTULO I, PARTE B

### Mycologia

#### Study of morphological features of *Ganoderma* subgenus *Ganoderma* in Mexico

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**Abstract:** A discussion on the important morphological features for determination of species in subgenera *Ganoderma* is presented. More of 120 specimens were checked, which included twelve species of *Ganoderma* from Mexico, viz. *G. colossus*, *G. curtisii*, *G. mexicanum*, *G. oerstedii*, *G. oregonense*, *G. perturbatum*, *G. resinaceum*, *G. sessile*, *G. sessiliforme*, *G. subincrustatum*, *G. weberianum* and *G. zonatum*. Color and resinous deposits of the context, structure of basidiospores, and protuberances of the cuticle cells are the most important features for characterization of the species.

**Key words:** cuticle cells, basidiospores structure, resinous deposit

### INTRODUCTION

The genus *Ganoderma* P. Karst. is characterized by its double-walled, mostly colored basidiospores, with truncate or subacute apex and ornamented exosporium and includes species both with a dull and laccate pileus. The pileus cuticle has been differently characterized and according to Furtado (1965a) in the subgenus *Elfvigia* it is a derm of the palisadoderm type, while in the subgenus *Ganoderma* consists of a layer of cells in a hymeniderm. Based in Cléménçon (2004) definitions, the cuticle in *Elfvigia* is a trichoderm and in subgenus *Ganoderma* is a crustohymeniderm. The classification of *Ganoderma* in two subgenera has been supported by phylogenetic analyses (Moncalvo 2000, Hong and Jung 2004); nevertheless there are many problems in the resolution below subgeneric level.

There have been several studies of the known American *Ganoderma* species, such as Bazzalo and Wright (1982), Gilbertson and Ryvarden (1986), Gottlieb and Wright (1999) and Ryvarden (2000, 2004), where the latter recorded 20 species of the two subgenera in the Neotropics.

The micromorphological structures of some species or groups in *Ganoderma* have been studied among others by Haddow (1931), Heim (1962),

Furtado (1965a, b, 1967), Steyaert (1967, 1972, 1975), Pegler and Young (1973), Corner (1983) and Adaskaveg and Gilbertson (1988a). More recently, Gottlieb and Wright (1999) made a detailed study of the basidiospores in an attempt to solve some of the intraspecific problems. However, these studies have been restricted to a rather limited number of species and few of them made use of the cuticle cells as a critical character in their evaluation of the species concepts.

Through the study of more than 120 specimens, mainly Mexican, and 22 holotypes, we studied the morphological variation of the structures of *Ganoderma* subgenus *Ganoderma*, with the following aims: 1) to do a detailed study of current and potential structures useful in the identification of the species and in the phylogenetic analyses, and 2) to give a better circumscription of the species.

## MATERIALS AND METHODS

*Specimens examined.*—The specimens for this study came from the Mexican herbaria ENCB, IBUG and XAL, specimens collected by the first author, as well as specimens borrowed from the following herbaria: BPI, FH, H, K, NY, O, PC, SP and UPS. Herbaria abbreviations follow Holmgren et al (1990).

*Macro and micromorphological observations.*—Descriptions of the macromorphological features of the basidiomata were made mainly according to the following characters: adhesion to the substrate, shape, consistency, weight, width and length; pileus color, consistency, shiny; tubes stratification, length, color, pores per mm, pore surface color; context stratification, width and color, resinous deposits. Besides the micromorphological features traditionally used, the descriptions include the following: basidiospore apex, disposition and size of pillars of the basidiospores; size, shape, and incrustations of the cuticle cells. The color references were made according to Komerup and Wanscher (1963). Microscopical observations were made from sections mounted in 10% KOH and Melzer's reagent; besides Congo red, floxine and cotton blue were used. Basidiospore shape was determined according to Q (length-width, Bas 1969) of 20 randomly selected basidiospores. Only mature cuticles cells were described. In the macrochemical reactions 10% KOH was used. The majority of the morphologic terms are according to Vellinga (1998), Furtado (1965a) and Clémenton (2004). The description of the pileipellis was done according to Clémenton (2004).

*Taxonomy.*—Generally the determination of the Mexican specimens was made through comparison with the type or types of related species. Furthermore, the keys of Bazzalo and Wright (1982) and Ryvarden (2004) were used, besides the descriptions of Steyaert (1972) and Corner (1983).

## RESULTS

*Macromorphological features.*—The shape and attachment to the substrate of the basidiomata are of rather restricted value for taxa delimitation; however, the consistency, weight and basidioma thickness proved to be of value in the taxonomic determinations. *Ganoderma colossus* (Fr.) C.F. Baker and *G. oregonense* Murrill have a spongy and light weighted basidiomata, while *G.*

*zonatum* Murrill has a woody-corky basidioma but nevertheless light weighted. *Ganoderma mexicanum* Pat., *G. sessile* Murrill, *G. sessiliforme* Murrill and *G. subincrustatum* Murrill are woody and have a relatively light-weighted basidiomata. *Ganoderma colossus*, *G. oregonense*, *G. oerstedii*, *G. sessile* and *G. zonatum* are generally robust and *G. mexicanum*, and *G. sessiliforme* have slender basidiomata.

The more important features of the pileus context for the delimitation of the species are the following: structure, consistency and presence of resinous deposits. The structure of the context is classified as: duplex, relatively homogeneous or homogeneous. In the duplex context there is an abrupt change, with two separate, contrasting colors, one upper generally light-colored, and a lower darker one close to the tubes, although the shades will vary to some extent (FIG. 1). Relatively homogeneous is used when there is an evident color difference between the upper and lower parts but without abrupt color changes (FIGS. 2, 3, 4) while the homogeneous context presents only one color (FIG. 5). In some cases, there is a very thin darker line just above of the tubes, absent in some specimens, in this case the context was considered as homogeneous.

Only three types of context consistency are recognized. Two of them: soft-spongy and fibrous are easy to distinguish; but there is an intermediate state between soft and fibrous that is difficult to delimitate. In this study, the third type was considered as fibrous-spongy. In some basidiomata there are resin-like deposits in the context. These deposits are hard and brittle and could be dull or shiny. Often they form continuous bands or discrete bodies, called resinous bands or resinous incrustations, respectively (FIGS. 2, 4, 6).

#### *Micromorphological features*

**Basidiospores.** In accordance with the Q coefficient as defined by Bas (1969), the majority of the species have ellipsoid to oblong basidiospores: while *G. perturbatum* (Lloyd) Torrend and *G. zonatum* have spores distinctively widely ellipsoid to ellipsoid, and oblong to cylindrical, respectively. The *Ganoderma* basidiospores have a cap in the apex (FIG. 7), which is obtuse and hyaline; after drying, the cap generally collapses and breaks, making the basidiospore apex truncate (FIG. 8). However, in some species the cap is permanent or very small and the basidiospores have a subacute apex, which is the case in *G. colossus*, *G. oregonense* and *G. perturbatum* (FIGS. 9, 10, 11), while the remaining species have evidently truncate basidiospores.

The inter-walled pillars on the basidiospores are thin to thick: thin (< 0.4  $\mu\text{m}$ ) as in *G. mexicanum* and *G. zonatum* (FIG. 12), intermediate (0.5-0.6  $\mu\text{m}$ ) as in *G. curtisii* (Berk.) Murrill, *G. resinaceum* Boud., *G. sessile*, *G. sessiliforme*, *G. subincrustatum* Murrill and *G. weberianum* (Bres. et Henn. ex Sacc.) Steyaert, and thick (0.6-1  $\mu\text{m}$ ) as in *G. colossus*, *G. oregonense*, *G. perturbatum* (FIG. 13) and *G. oerstedii*. The pillars can be: free, subfree, partially anastomosed and reticulate (FIGS. 14, 15, 16, 17). The free pillars appear as dots on the basidiospore surface while free dots mixed with short anastomosed to shortly elongated structures are classified as subfree. The term partially anastomosed is used when more than two pillars are grown together and form an irregular surface,

and reticulate is when the ornamentation is present as an almost complete net. According to Cléménçon (2004), all *Ganoderma* basidiospores have a thin, hyaline to reddish-yellow and smooth or rugose perisporium; an ornamented and colored exosporium, and a thick and reddish-brown or yellowish-brown endosporium.

**Structure of the pilear cover.** The pilear surface is formed by a cell layer similar to a hymeniderm, although in the majority of the species the cells do not originate from the same level as in the true hymeniderm. The type of hymeniderm in *Ganoderma* was defined by Cléménçon (2004) as a crustohymeniderm, which is characterized by having thick-walled cells apically covered by a resinous mass. The size of the cells must be interpreted with care as it varies between species, but also in the same specimen according to where the section has been made. Most species have club like apical cells although there are species with branched or knobby cells besides the club shaped cells.

The shape of the crustohymeniderm cells are: cylindrical as in *Ganoderma perzonatum* Murrill, almost cylindrical to narrowly-clavate as in *G. weberianum*, widely-clavate as in *G. perturbatum* and clavate in the majority of the species. The wall may be thin as in *G. colossus* or thick as in the rest of the species. The thick wall may be multistratified, which it is related to the maturity of the basidiomata; some species, as *G. perturbatum*, have an evidently multistratified wall. Some young cuticle cells present secondary septa, more evident in the cuticle cells of the periphery of the basidiomata; furthermore, the cuticle cells of the periphery have a tendency to be lighter and shorter. Also granulations or incrustations can be present in the cuticle cells, in the first case, the granules are easily dissolved in KOH while the incrustations remain because they are difficult to be removed with alkali; an example of this situation is in *G. perzonatum*.

In the cuticle cells there are transitions from smooth clublike cells to antler-like cells. There are serial forms in which the characters change progressively, but a predominant form is always observed. Preliminary three large groups might be found: entire, lobulate-branched and branched crustohymeniderm. 1) The entire crustohymeniderm has entire cuticle cells, cylindrical and/or cylindrical with slightly wide apex, cylindrical to narrowly-clavate, cylindrical with subcapitate apex or narrowly-clavate to widely clavate, very occasionally with maximum two generally lateral protuberances, e.g. *G. resinaceum*, *G. perturbatum*, *G. sessile* (FIG. 19a). 2) The lobulate-branched crustohymeniderm can be subdivide in: a) crustohymeniderm with entire, clavate cuticle cells, with one to three round, short and thick protuberances and/or branches, but conserving the clavate shape, e.g. *G. nitidum*, *G. zonatum* (FIG. 19b); b) cuticle cells with one to two thick and long branches and up to seven short and thick protuberances, the clavate shape is generally lost, e.g. *G. oerstedii*, *G. subincrustedum* (FIG. 19c). 3) The branched crustohymeniderm can be of two types in which the clavate shape is generally lost: a) with two to three thin and long branches, with small protuberances, e.g. *G. orbiforme* (FIG. 19d). and b) the most evident cells have many antler-like branches and small protuberances, e.g. *G. multicornis* (FIG. 19e).

**Hyphal system.** In all species the hyphal system is di-trimitic, and the generative and binding hyphae are generally difficult to observe. The generative hyphae all have clamps at the septa and are hyaline to yellowish and in almost all the species they are branched. Skeletal hyphae are of the arboriform type, thick-walled to solid, yellowish to yellowish-brown, apically branched and thin in the crustohymeniderm, wider in the context. Binding hyphae are generally thinner than skeletal hyphae, hyaline to yellowish-brown.

**Cystidia.** The cystidia were studied following Vellinga (1998), who define them as sterile, differentiated, terminal elements in the hymenium; according with this definition *G. colossus* and *G. oregonense* present cystidia (FIG. 18). These structures are difficult to observe but were present in the majority of specimens of the two species.

## COMMENTARIES OF THE STUDIED SPECIES

*Ganoderma colossus* (Fr.) C.F. Baker, *Brotéria* 425, 1918.

≡ *Thomophagus colossus* (Fr.) Murrill, *Torreya* 5: 197, 1905.

= *Ganoderma obockense* Pat., *Bull. Soc. Mycol. Fr.* 3: 119, 1887.

*Specimens examined.* **Veracruz**, *G. Guzmán* 35708 (XAL), *F. Ventura* 12195 (ENCB); **Quintana Roo**, *G. Guzmán* 20516 (XAL); **Chiapas**, *G. Castillo* 2803 (XAL).

*Other specimens examined.* **COSTA RICA**, HOLOTYPE of *Ganoderma colossus* (UPS). **SOMALIA**, LECTOTYPE of *Ganoderma obockense* (PC).

Furtado (1965b), Adaskaveg and Gilbertson (1988), Ryvar den and Johansen (1980) and Gilbertson and Ryvar den (1986) described cuticle cells as entire, and Nuñez and Ryvar den (2000) and Ryvar den (2000, 2004) considered them slightly diverticulated; the specimens studied had some cells with branched apex, besides the diverticula. Nuñez and Ryvar den (2000) and Ryvar den (2004) mentioned cuticle cells apically encrusted, which was not observed in Mexican materials. Ryvar den and Johansen (1980) described thick-walled cuticle cells; nevertheless the cells of the Mexican specimens are thin-walled, as described by Gilbertson and Ryvar den (1986). On the other hand, cystidia as defined here, were not mentioned by other authors, but classified as cystidiols by Ryvar den and Johansen (1980). Furthermore, sclerified generative hyphae are described for this species.

Steyaert (1972) suggested that *Ganoderma oregonense* might be a temperate variety of *G. colossus*. The two species are similar in several characters, as they both have soft, spongy, very light in weight and pale context, and large basidiospores with subacute apex. However, they differ in other characters, especially in the dark color of the basidiomata and basidiospores with free pillars in *G. oregonense*. Phylogenetic analyses (Moncalvo et al 1995, Moncalvo 2000) indicated little relationship between the two species. A recent molecular study (Hong and Hung 2004) suggested that *G. colossus* might be placed in *Thomophagus* (Fr.) Murrill. However, more molecular studies using other molecular markers from several genes will be necessary to settle its generic position.

***Ganoderma curtisii* (Berk.) Murrill, North Amer. Flora 9: 120, 1908.**

*Selected specimens examined.* Jalisco, M.G. Torres-Torres 526, 541, 554, L. Guzmán-Dávalos 7447 (IBUG); Hidalgo, J. Gimete 152-A (ENCB); Morelos, M. Frias Neve 18 (ENCB).

*Other specimens examined.* USA, HOLOTYPE of *Ganoderma ravenelii* (K).

*Ganoderma curtisii* is perhaps one of the most morphologically variable species of the genus. Torres-Torres and Guzmán-Dávalos (2005) recently discussed this morphological variation in Mexican specimens. The species was described with club-shaped cuticle cells and this concept is the same as used by many authors (Haddow 1931, Steyaert 1980, Ojeda-López et al 1986); nevertheless we found a great variability in respect to the number of protuberances in the cuticle cells. In general, the basidiospores are  $8.5\text{--}12 \times 5.6\text{--}8 \mu\text{m}$  (Overholts 1953, Steyaert 1980, Ojeda-López et al 1986, Torres-Torres and Guzmán-Dávalos 2005), but Overholts (1953) indicated some basidiospores can be up to  $13\mu\text{m}$ , an observation we confirm. Overholts did not study the cuticle cells but in the specimens studied by us those with long basidiospores had diverticulated cuticle cells. A more detailed examination should be performed to see whether one or two species are involved.

Two species: *G. meredithiae* Adask. & Gilb. and *G. ravenelii* Steyaert are morphologically similar to *G. curtisii*. The first one is mainly differentiated by its cuticle cells, which are diverticulated, and being restricted to *Pinus* (Adaskaveg and Gilbertson 1988b). The second has basidiospores  $10\text{--}14.5 \times 5\text{--}6.5 \mu\text{m}$  according to Steyaert (1980), and  $11.2\text{--}15.2 \times 5.2\text{--}7.2 \mu\text{m}$ , oblong to cylindrical in the type, with no resinous bands in the context. It is probable that *G. ravenelii* occurs in Mexico; one specimen (*C. Téllez 1025*) has basidiospores distinctively oblong to cylindrical, but it is a young specimen and very few basidiospores were measured.

***Ganoderma mexicanum* Pat., Bull. Soc. Myc. Fr. 14: 54, 1898.**

*Specimens examined.* Mexico, LECTOTYPE (FH-4823).

Because of the bad condition of the type specimen a detailed study was not possible; nevertheless, its cuticle cells and basidiospores are remarkable. Few basidiospores were checked because most of them were in bad state. We think that probably they have thick pillars ( $0.4 \mu\text{m}$ ). On the other hand, we think that the context is homogenous but it could be relatively homogenous. *Ganoderma mexicanum* is close related with *G. sessile*, but the last one has bigger basidiospores [ $11.2\text{--}14.4\text{--}16.4 \times 7.2\text{--}8.8 \mu\text{m}$ , instead of  $8\text{--}10 \times 5.6\text{--}8 \mu\text{m}$  of *G. mexicanum*] and duplex context.

***Ganoderma oerstedii* (Fr.), Torrend, Bull. Torrey bot. Club 29: 606, 1902.**  
= ***Ganoderma tuberculosum* Murrill, North American Flora 9: 123, 1908.**

*Selected specimens examined.* Guerrero, M.G. Torres-Torres 546 (IBUG); Jalisco, M.G. Torres-Torres 408, 563 (IBUG); Oaxaca, M.G. Torres-Torres 573, 575 (IBUG).

*Other specimens examined.* COSTA RICA, NEOTYPE of *Ganoderma oerstedii* (UPS). HONDURAS, LECTOTYPE of *Ganoderma tuberculosum* (NY).

Ryvarden (2000, 2004) described the cuticle cells as entire; nevertheless, although the neotype of *G. oerstedii* is in a bad state, cuticle cells with protuberances and branches were presented. On the other hand, the basidiospores of the types recorded by Ryvarden (2000, 2004) are larger ( $12-15 \times 8-10 \mu\text{m}$ ) than we observed in the types [ $11-12.8 \times 7.2-9.4 (-10.5) \mu\text{m}$  in *G. oerstedii*, and ( $9.6-$ )  $10.4-12.8 \times (7.2-)$   $8-9.6 \mu\text{m}$  in *G. tuberculosum*] and in the Mexican specimens.

***Ganoderma oregonense* Murrill, North Amer. Flora 9: 118, 1908.**  
= ***Ganoderma sequoiae* Murrill, North Amer. Flora 9: 119, 1908.**

*Selected specimens examined.* Estado de México, S. Acosta 653 (IBUG), G. Guzmán 4675, (ENCB); Hidalgo, M.L. Aguirre-Jones s.n. (ENCB); Veracruz, G. Guzmán 28886 (XAL).

*Other specimens studied.* USA, LECTOTYPE of *Ganoderma oregonense* (NY); LECTOTYPE of *Ganoderma sequoiae* (NY).

This species is closely related to *Ganoderma tsugae* Murrill, an independent species; however it is difficult to define the characters to separate them. Overholts (1953) suggested thickness and length of the tubes in *G. oregonense* as the main features to distinguish them; furthermore he described smaller basidiospores for *G. tsugae*, of  $9-11 \times 6-8 \mu\text{m}$ . Gilbertson and Ryvarden (1986) considered the large size of the basidiomata, large pores and wider basidiospores as the diagnostic features of *G. oregonense*; they described basidiospores  $13-17 \times 8-10 \mu\text{m}$  for *G. oregonense* and  $13-15 \times 7.5-8.5 \mu\text{m}$  for *G. tsugae*. On the other hand, they considered a homogeneous context for *G. oregonense*. Overholts (1953) and Gilbertson and Ryvarden (1986) described a slightly darker layer in the context next to the tubes in *G. tsugae*. Although only two specimens of *G. tsugae* were observed in this study, we suggest size and thickness of the basidiomata, and number of pores per mm set in the differences seen here as the most important features in the separation of the species, until more specimens will be examined.

***Ganoderma perturbatum* (Lloyd) Torrend, Bróteria Bot. 18: 34, 1920.**

*Specimens examined.* Colima, Grupo Ecológico Forestal Tonatiuh s.n. (IBUG).

*Other specimens studied.* BRAZIL, LECTOTYPE of *Ganoderma perturbatum* (BPI, Lloyd herb. num. 55740).

Steyaert (1967) suggested that *Ganoderma perturbatum* could be a form of *G. lucidum*, as some macromorphological features and the shape of the

basidiospores are similar. The neotype of *G. lucidum* (Finland, Fennici Exsiccati 239, s.date, H) selected by Gottlieb et al (1999) has whitish context and cuticle cells narrower than *G. perturbatum*. Ryvar den (2000) considered that *G. perturbatum* is a synonym of *G. resinaceum*, but the last one has thinner, sessile to substipitate basidiomata (as well as *G. lucidum* in some cases) and truncate, ellipsoid basidiospores, with abundant free pillars, and relatively narrow cuticle cells (see below).

*Ganoderma perturbatum* is close to *G. dorsale* (Lloyd) Torrend and they were suggested as synonyms by Steyaert (1967). The revision of the type of *G. dorsale* (Region Grande do Sul, on buried wood, s.date, BPI, Lectotype) suggests that they are independent species, differing mainly in the duplex context and cuticle cells with unistratified wall, generally with granular incrustation apex. It seems related to *G. concinnum* Ryvar den (Ryvar den 2000) (Colombia, Chocó, Municipality of Riosucio, Sautata, Parque Nacional Katios, 28–30 Jun 1978, *L. Ryvar den 16840*, O, Holotype). *Ganoderma concinnum* has thin resinous bands and duplex context, relatively longer stipe, and longer and thinner basidiospores [12–14 × 8–9(–10) µm].

***Ganoderma resinaceum* Boud., Bull. Soc. Mycol. Fr. 5: 72, 1889.**

*Selected specimens examined. Colima, G. Guzmán 11647 (SP).*

Although the type in PC was not checked by us, two collections in PC were examined. One of them is an authentic specimen (s. locality, Sep 1890, *J.L. Boudier s.n.*, on Chêne liège trunk), and the other one is incorrectly labeled as the type specimen (TUNISIE, Aîn Drahem, on Chêne liège trunk, 7 Sep 1895, *N. Patouillard s.n.*). The Mexican material coincides with the type description made by Gottlieb and Wright (1999), except that they reported cuticle cells negative in Melzer's reagent. Ryvar den (2000, 2004) described smaller basidiospores (9–11.5 × 5–7 µm), but on the other hand, a positive reaction of the cuticle cells in Melzer's reagent. The material from PC also present a positive reaction of the cuticle cells in Melzer's reagent.

*Ganoderma resinaceum* is a species with many synonyms; the Index Fungorum ([www.indexfungorum.org](http://www.indexfungorum.org)) indicates twelve taxonomic synonyms. Gottlieb and Wright (1999) did not accept the synonymy with *G. argillaceum* Murrill, *G. chaffangeonii* Pat., *G. praelongum*, *G. sessile*, *G. sessiliforme*, *G. subincrustedum*. Steyaert (1972) considered seven synonyms for the species; Bazzalo and Wright (1982) suggested the synonymy with 10 species including *G. nitidum* Murrill; Ryvar den (2000, 2004) accepted eleven but not *G. nitidum*. It would be desirable to have a common and widely accepted concept of the species and to differentiate it from allied species preferably supported by DNA sequencing. It may be that we are confronted with a variable species slightly differentiated into varieties or forms in some regions of the world. We studied the types of *G. areolatum* Murrill (Mexico, Lectotype, NY), *G. argillaceum* (Cuba, Lectotype, NY), *G. nitidum* (Honduras, Lectotype, NY; fragment in BPI), *G. perturbatum* (see above), *G. praelongum* (Cuba, Lectotype, NY; fragment in BPI), *G. pulverulentum* (Grenada Island, Lectotype, NY), *G. sessile* (see below), *G. sessiliforme* (see below), *G. subfornicatum* Murrill (Honduras, Lectotype, NY)



and *G. subincrustatum* (see below) and concluded with a slight doubt that they are independent species.

***Ganoderma sessile* Murrill, Bull. Torrey bot. Club 29: 604, 1902.**

*Selected specimens examined.* **Hidalgo**, S. & J. Peck s.n. (SP 124133, ex-BCI 3079); **Jalisco**, G. Guzmán 17888 (IBUG).

*Other specimens examined.* **USA**, LECTOTYPE (NY); L.M. Underwood s.n., May 1897, (NY); Bedford Park, on stumps of oak, 1 Jun 1902, s.coll. (NY); s.coll. (NY 12.123, Timber and Forest

Steyaert (1972, 1980) based on the morphology of the basidiospores suggested that the species was a taxonomic synonym of *G. resinaceum*, opinion that was followed by Bazzalo and Wright (1982) and Ryvardeen (2000, 2004). Nevertheless, *G. resinaceum* is different in several aspects: context light reddish-brown without resinous bands and almost cylindrical cuticle cells with a diffuse granulated apex.

***Ganoderma sessiliforme* Murrill, Bull. New York. Bot. Gard. 8: 149, 1912.**

*Selected specimens examined.* **Morelos**, LECTOTYPE of *Ganoderma sessiliforme* (NY), G. Guzmán 2078 (ENCB).

Ryvardeen (2000) suggested this species to be a synonym of *Ganoderma resinaceum*; nevertheless, this species has light reddish-brown context, longer cuticle cells, and longer basidiospores with free pillars. Macromorphologically, *G. sessiliforme* may be confused with *G. sessile*, which has resinous bands and duplex context, larger basidiospores and cuticle cells and basidiospores with free pillars.

***Ganoderma subincrustatum* Murrill, North Amer. Flora 9: 122, 1908.**

*Selected specimens examined.* **Jalisco**, O. Vargas 316, G. López-Damián 49 (IBUG); **Nuevo León**, González-Velásquez 556 (ENCB); **Quintana Roo**, R. Valenzuela 6429 (ENCB); **Veracruz**, G. Guzmán 287, F. Ventura 1312 (ENCB).

*Other specimens examined.* **JAMAICA**, LECTOTYPE of *Ganoderma subincrustatum* (NY).

*Ganoderma subincrustatum* was considered a synonym of *G. resinaceum* by Bazzalo and Wright (1982); on the other hand Ryvardeen (1985) suggested that could be *G. lucidum sensu lato*. Gottlieb and Wright (1999) recorded it from Argentina as an independent taxon of *G. resinaceum*. The species was not considered by Steyaert (1972, 1980), Corner (1983) and Ryvardeen (2004). Because of the morphology of the basidiomata and the anastomosed pillars of the basidiospores, we consider that it is an independent species.

***Ganoderma weberianum* (Bres. et Henn. ex Sacc.) Steyaert, *Persoonia* 7(1): 79, 1972.**

*Selected specimens examined.* **Jalisco, M.G. Torres-Torres 690 (IBUG).**

*Ganoderma rivulosum* Pat. & Har. is a synonym according to Steyaert (1972) and Corner (1983). Corner (1983) also included "*G. lauterbachii* Henn." as another synonym, but Steyaert (1972) considered it as a different species, because in the specimens examined by him the chlamydospores were absent. The name "*G. lauterbachii*" is not valid, because the combination has never formally made (Moncalvo and Ryvarden 1997). *Ganoderma weberianum* is also close related to *G. microsporum* R.S. Hseu (Hseu et al 1989) with similar macro and micromorphological features but with smaller basidiospores ( $6-9 \times 4.5-6.5 \mu\text{m}$ ), longer cuticle cells and absence of chlamydospores in the context. Based in the study of the isotype, Wang et al (2005) considered *G. microsporum* as synonym of *G. weberianum*. They found chlamydospores and resinous incrustations in the isotype although they were not indicated in the protologue. It has to be mentioned that the occurrence of chlamydospores is variable in the genus and should be treated with care, it depends also partly where and at which stage the section was made.

***Ganoderma zonatum* Murrill, *Bull. Torrey bot. Club* 29: 606, 1902.**

*Selected specimens examined.* **Jalisco, A. Cervantes 1 (IBUG); Nayarit, E. Fanti 514 (IBUG).**

*Other specimens examined.* **USA, LECTOTYPE (NY).**

*Ganoderma sulcatum* Murrill is a synonym of *G. zonatum* according to Lloyd (1915), Bazzalo and Wright (1982), Gottlieb and Wright (1999) and Ryvarden (2000, 2004). This is the first record for Mexico and on different substratum (hard wood); previously was recorded growing only on palms. Corner (1983) described specimens from Brazil, Peru and Bolivia, suggesting they belong to *G. zonatum*, but his descriptions are deviating. The specimens recorded from Argentina by Bazzalo and Wright (1982) are substipitate and have rugose basidiospores; maybe they correspond to a different species.

## DISCUSSION

It is clear that the species concepts in *Ganoderma* are variable according to the author. Historically *Ganoderma* has been treated as a genus with a great plasticity or homogeneity of its morphological features. Although Bazzalo and Wright (1982) and other authors considered the color of the context of little value, we found that this character, together with the presence of the resin-like deposits, is important in the separation of taxa. The resin-like deposit was named melanoid deposits by Gottlieb and Wright (1999), while Steyaert (1980) used "melanoid substance". Ryvarden (2000) used the term "resinous bands" which is adopted here but with a wider definition to include resinous bands and resinous incrustations.

The basidiospores of *Ganoderma* have been traditionally described with double wall, with a thin exosporium and a thick endosporium, echinulate or with inter-wall pillars, with truncate apex (Haddow 1931, Overholts 1953, Corner 1983, Tham 1998, Ryvar den 2004, among others). According to Cl  men  on (2004) the terms exosporium and endosporium have been wrongly applied. Bazzalo and Wright (1982) suitably used the terms perisporium and endosporium; however, they considered the ornamentation originated from the endosporium, which is erroneous because it is the exosporium that forms the ornamentations (Cl  men  on 2004). According with the definition of Kirk et al (2001), the term echinulate is erroneously used; so we prefer to describe the inner ornamentation as pillars. Bazzalo and Wright (1982) and Gottlieb and Wright (1999) observed the basidiospore pillars under the Scanning Electronic Microscope, and proposed them as useful features in the determination of the species. Some taxa have basidiospores with a truncate apex, which occurs when the cap of the apex is collapsed. The cap is described as "bouchon perisporique" by Heim (1962), as apical papilla by Furtado (1962), as umbo by Corner (1983) and as cap by Tham (1998). Other species have subacute apex, in these cases, the basidiospore has no cap or it is very small. The term subacute apex was used by Corner (1983), and described by Steyaert (1972) as spores with unchanged apex; nevertheless this character has not been used systematically. We consider that it is an important feature in the separation of the taxa; for example, it is one of the reasons for treating *G. perturbatum* as a different species from *G. resinaceum* and related to *G. dorsale*. *Ganoderma conccinum* Ryvar den, *G. dorsale*, *G. longistipitatum* Ryvar den (Venezuela, Holotype O!) and *G. perturbatum* form a distinctive group of species by their stipitate basidiomata and the subacute basidiospore apex. The shape of the basidiospores is also an important factor in separation of taxa. Based on its oblong to cylindrical basidiospores, *G. ravenelii* and *G. zonatum* can be differentiated from species with similar macromorphological characteristics like: *G. curtisii* and *G. orbiforme* (Fr.) Ryvar den (Guinea, Holotype UPS!).

The surface of the pileus is a complex structure in which the crustohyphenoid cells, hyphae and resinous-like substances are immersed. Many discussions on its importance in the taxonomy of the genus have been made; however, its current use is limited, perhaps by the problems to find precise terms for their definition. Much of the complexity is caused by the great variability in the branching pattern of the cells in the same basidioma, creating confusion and difficulty to find a collective term. On the other hand, the interpretation of the development state is complex, restricting the use to the sizes and shapes. Torres-Torres and Guzm  n-D  valos (2005) found that the cuticle cells are very variable in size and shape depending on the site of the cut and on the maturity of the basidiomata. For this reason, for taxonomic purposes only mature cells must be compared.

Imazeki (1939) and Steyaert (1980) proposed classifications based in the type of dermis; the definitions of the last one were partially adopted by Gottlieb and Wright (1999). However, previously Bazzalo and Wright (1982) considered that the types of dermis described by Steyaert (1980) and Imazeki (1939) were not useful for delimitation of species, although were useful for large groups. Although Gottlieb and Wright (1999) made only schematic illustrations of the cuticle cells without description of the inherent variation, they considered the cutis type and

basidiospore ornamentation useful for delimitation of taxa; which were confirmed in the present investigation. There is a complex of species including *G. boninense*, *G. multiplicatum* (Mont.) Pat., *G. orbiforme*, *G. subfornicatum* and *G. oerstedii* (Fr.) Torrend, all with yellowish-brown to reddish-brown context and cuticle cells with protuberances and/or branches; where the differences are in the features of the basidiospores and in the shape and quantity of branches in the cuticle cells. Karsten in 1881 did not mention cystidia in his original description and his non-cystidiate concept was accepted by many mycologists (Haddow 1931, Heim 1962, Furtado 1965b, 1967, Steyaert 1967, 1972, Bazzalo and Wright 1982, Gilbertson and Ryvarden 1986, Ryvarden 1991, 2000). Demelius in 1919 cited by Corner (1983) referred the presence of acute cystidioles in *G. applanatum* and *G. lucidum*; however, we consider true cystidia, following the concept of Vellinga (1998), to be present in *G. colossus* and *G. oregonense*.

Although, Ryvarden (2004) published a mycota of Ganodermataceae in the neotropics, many names remain doubtful because many of the descriptions have been based only in the type material and because the name *Ganoderma lucidum* s.l., has perpetuated in herbaria and publications.

#### ACKNOWLEDGMENTS

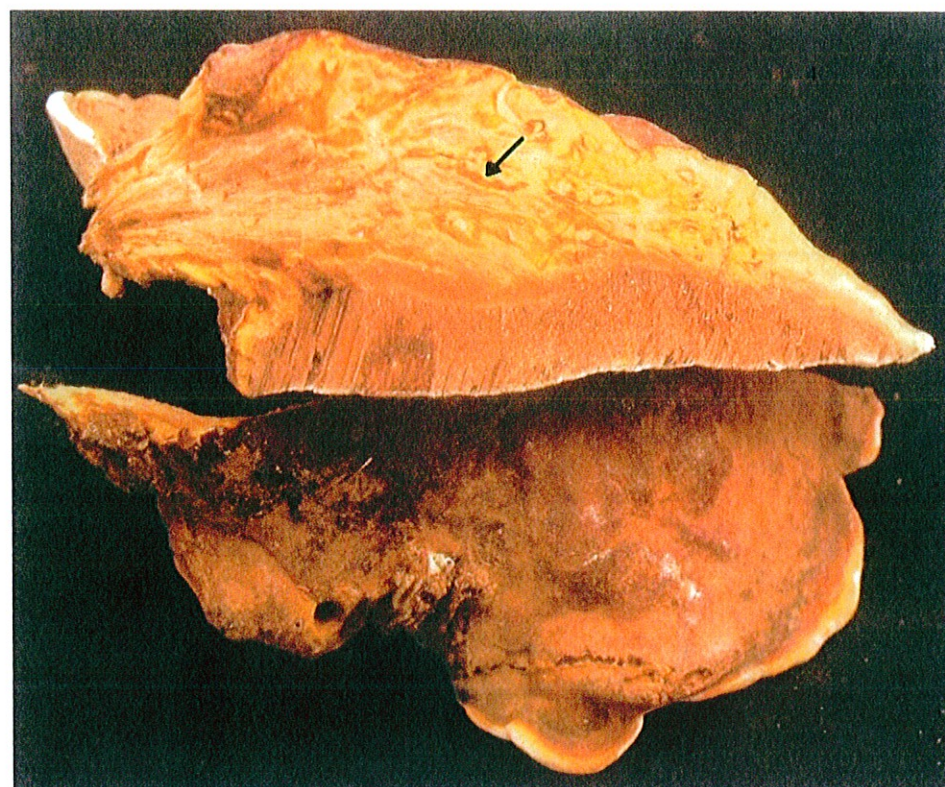
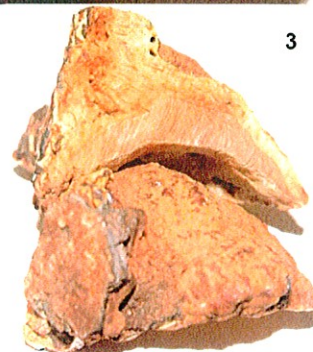
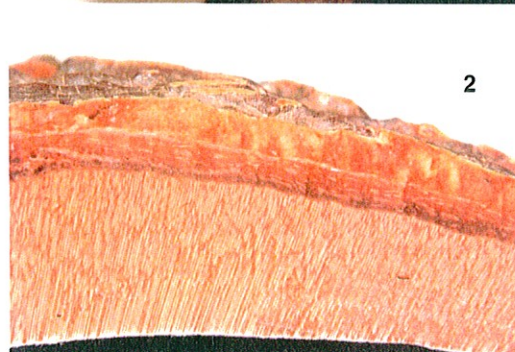
We are grateful to the curators of BPI, ENCB, FH, H, K, NY, O, PC, SP, UPS and XAL. Thanks are due to Universidad de Guadalajara (CA-23 50052, PROCOFIN 7388401), CONACYT (42957) and PROMEP (103.5/03/2580). The first author thanks Oslo University for a grant to visit O herbarium, Red Latinoamericana de Botánica for a grant to visit SP herbarium (RLB-05-P5), COLCIENCIAS and Universidad Tecnológica del Chocó for economic help for her Doctoral studies in the Universidad de Guadalajara, México.

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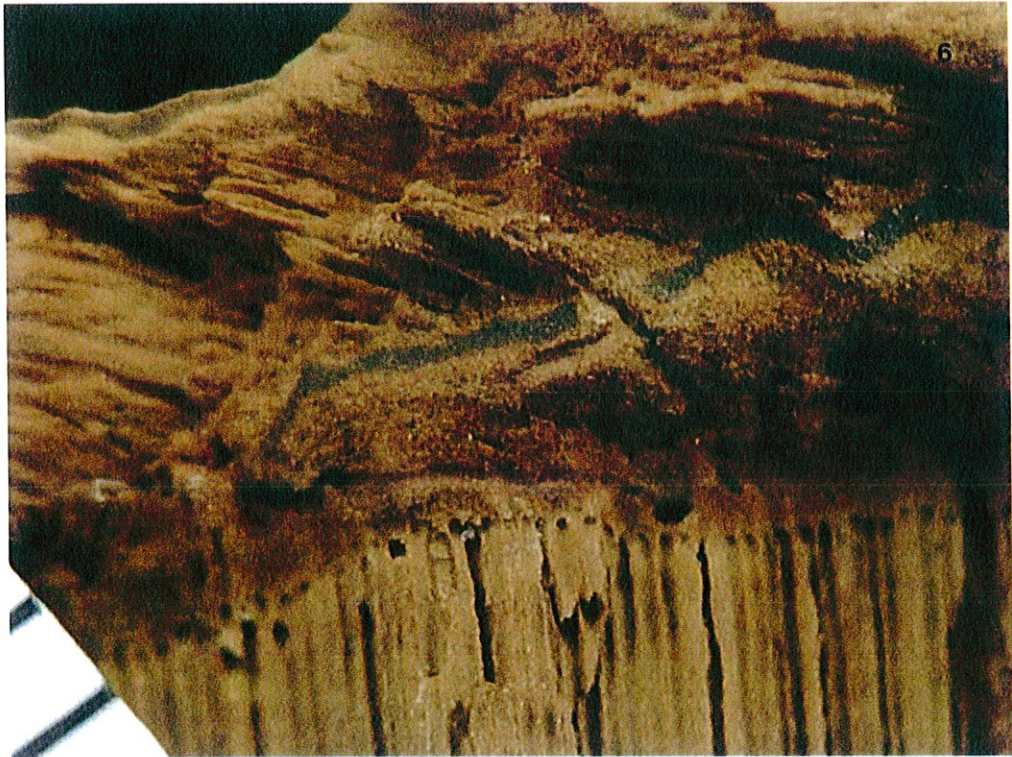
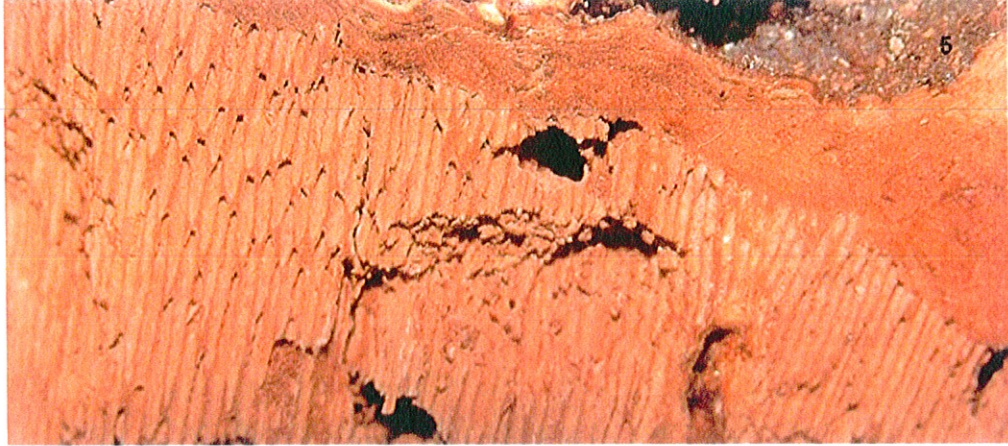
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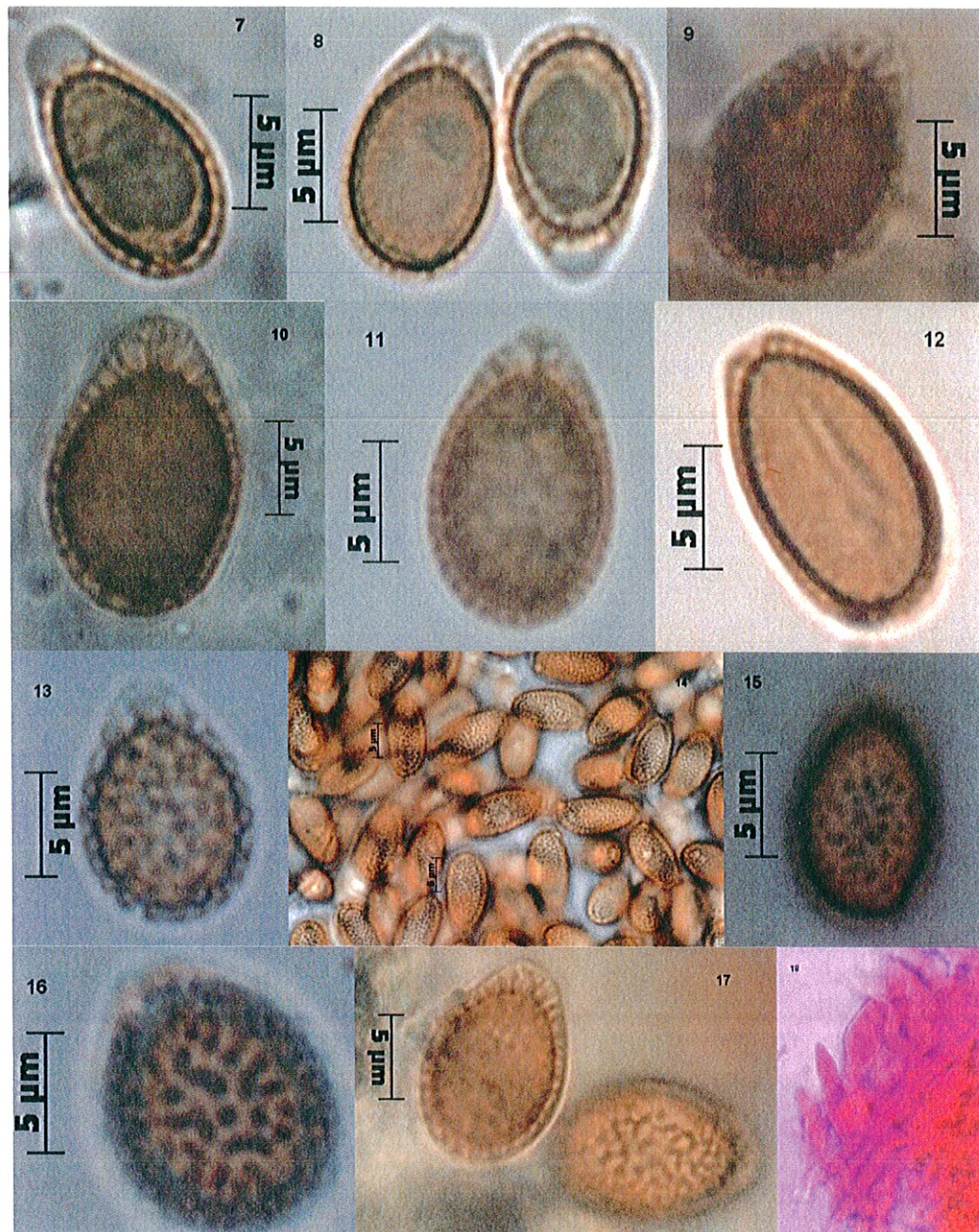


FIGS. 1–4. Macromorphological features of *Ganoderma*. 1. Duplex context of *G. sessile*. 2–4. Relatively homogeneous context. 2. With very evident resinous bands of *G. tuberosum*. 3. Context of *G. sessiliforme*. 4. With resinous incrustations of *G. weberianum*.



FIGS. 5–6. Macromorphological features of *Ganoderma*. 5. Homogeneous context of *G. resinaceum*. 6. Resinous bands in the context of *G. sessile*.





FIGS. 7–18. Micromorphological features of *Ganoderma*. 7–8. Basidiospores of *G. argillaceum*: 7. With cap. 8. Apex with cap or truncate. 9–11. Basidiospores with subacute apex: 9. *G. perturbatum*. 10. *G. colossus*. 11. *G. oregonense*. 12. Basidiospores with thin pillars of *G. zonatum*. 13. Basidiospores with thick pillars of *G. perturbatum*. 14–17. Arrangement of basidiospores pillars: 14. Free of *G. zonatum*. 15. Subfree of *G. oregonense*. 16. Partially anastomosed of *G. perturbatum*. 17. Subreticulate of *G. colossus*. 18. Cystidia of *G. oregonense* in floxina.

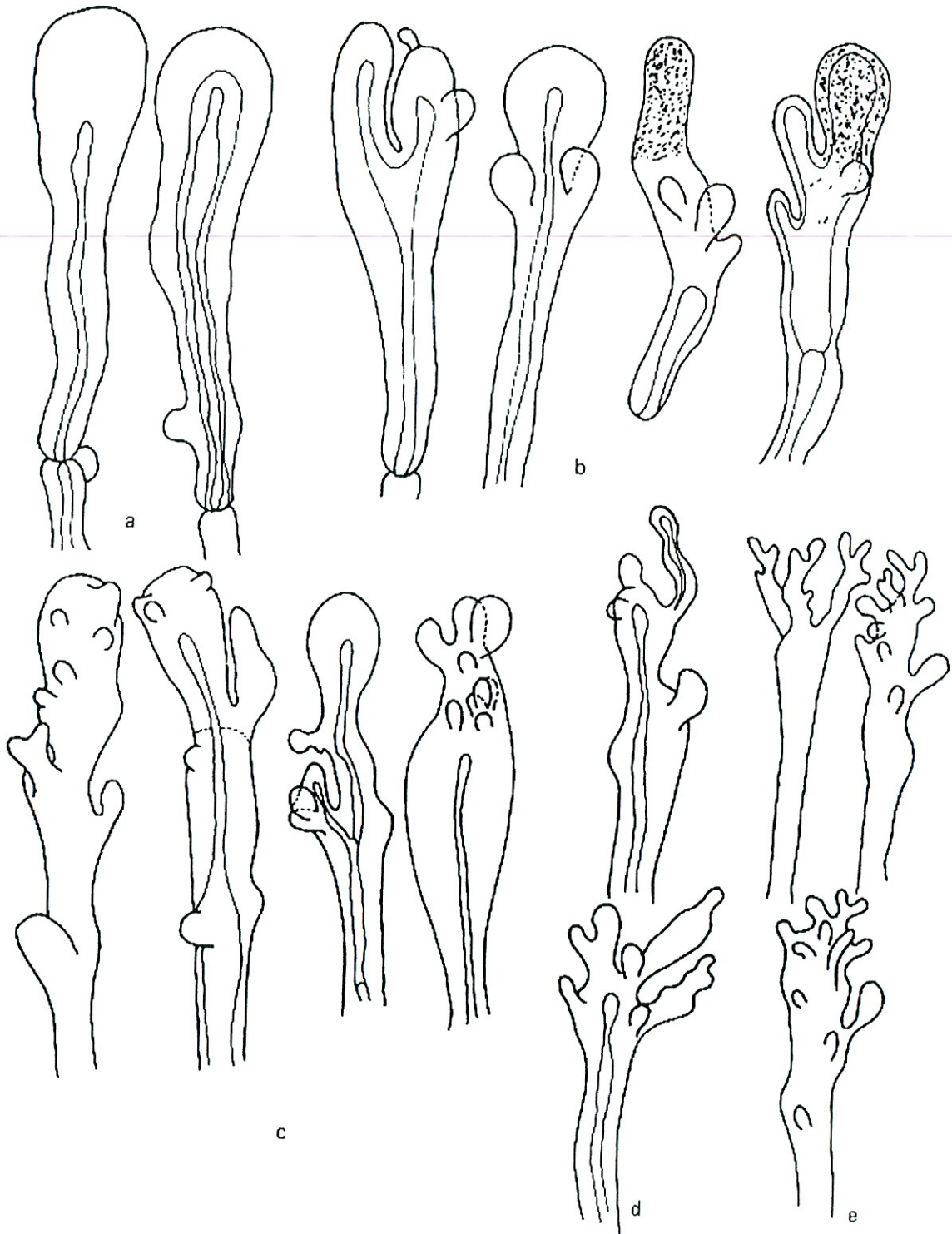


FIG. 19. Cuticle cells of the crustohymeniderm: a. Entire of *G. perturbatum* and *G. sessile*. b. Lobulate-branched of *G. nitidum* and *G. zonatum*. c. With one to two thick and long branches and up to seven short and thick protuberances from *G. oerstedii* and *G. subincrustatum*. d. Branched from *G. orbiforme*. e. Antler-like from *G. multicornis*. Bar = 8  $\mu$ m.

## CAPÍTULO I, PARTE C

### Mycologia

#### *Ganoderma* subgenus *Ganoderma* in Mexico

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**Abstract:** Twelve species of *Ganoderma* subgenus *Ganoderma* are reported from Mexico, viz. *G. colossus*, *G. curtisii*, *G. mexicanum*, *G. oerstedii*, *G. oregonense*, *G. perturbatum*, *G. resinaceum*, *G. sessile*, *G. sessiliforme*, *G. subincrustatum*, *G. weberianum* and *G. zonatum*. From them, *G. perturbatum* and *G. subincrustatum* are recorded by the second time in the world; *G. weberianum* is new to America, while *G. oerstedii* and *G. zonatum* are new to Mexico. Descriptions and illustrations of each species and a key of the Mexican species of the subgenus are provided.

**Key words:** distributions, morphology, new record, taxonomy

### INTRODUCTION

*Ganoderma* P. Karst. included two subgenera: *Ganoderma* and *Elfvigia* (P. Karst.) Imaz. The first is characterized by its glossy pileus surface and hymenodermic pileipellis; in contrast, *Elfvigia* has dull pileus without cuticle cells.

Eight species of the subgenera *Ganoderma* have previously been reported from Mexico: *G. colossus* (Fr.) C.F. Baker, *G. curtisii* (Berk.) Murrill, *G. lucidum* s. l., *G. mexicanum* Pat., *G. oregonense* Murrill, *G. resinaceum* Boud., *G. sessile* Murrill and *G. sessiliforme* Murrill (Patouillard 1898, Murrill 1912, Furtado 1965, Welden and Guzmán 1978, Guzmán-Dávalos and Guzmán 1979, Guzmán 1983, Anel and Guzmán 1987). Many of these species were probably mistakenly determined. The species with dull pileus surface (subgenus *Elfvigia*) as *G. applanatum* (Pers.) Pat., *G. australe* (Fr.) Pat. and *G. lobatum* (Schwein.) G.F. Atk., known from Mexico (Guzmán 1972, 1977), were not considered in this paper.

The main aims of this study were: 1) to describe in detail and to illustrate the species, 2) to give a better circumscription of the species and 3) to provide a reliable record of the Mexican species in subgenus *Ganoderma*.

## MATERIALS AND METHODS

*Specimens examined.*— This paper is based on 40 specimens collected in the field by one of us (Torres-Torres) and examination of more than 80 specimens deposited in Mexican herbaria. Furthermore, 22 types (holotypes and lectotypes) were included. The specimens for this study came from the Mexican herbaria ENCB, IBUG and XAL, as well as specimens borrowed from the following herbaria: BPI, FH, H, K, NY, O, PC, SP and UPS. Herbaria abbreviations follow Holmgren et al (1990).

*Macro and micromorphological observations.*—Morphological features descriptions were made from fresh and herbaria material. The color references were made according to Kornerup and Wanscher (1963). Microscopical observations were made from sections mounted in 10% KOH and Melzer's reagent, besides Congo red, floxine and cotton blue were used. Basidiospore shape was determined according to Q (length-width, Bas 1969) of 20 randomly selected basidiospores. In the macrochemical reactions 10% KOH was used. The drawings of microscopical structures were made with a 100x oil-immersion objective, in a three-dimensional plane to permit a better interpretation of the structures.

*Taxonomy.*—Generally the determination of the Mexican specimens was made through comparison with the type or types of related species not present in Mexico. Furthermore, the keys of Bazzalo and Wright (1982) and Ryvar den (2004) were used, besides the descriptions of Steyaert (1972) and Corner (1983).

## RESULTS

In the present paper descriptions of the species and a key to the Mexican species of *Ganoderma* subgenus *Ganoderma* as well as related species not yet recorded in Mexico are provided. Out of those reported here, five are new records for Mexico, and from them one is new to America, while two are recorded for the second time to the world. A detailed description of each species is provided, including three species apparently without a modern description: *G. mexicanum*, *G. perturbatum* (Lloyd) Torrend and *G. weberianum* (Bres. & Henn. ex Sacc.) Steyaert. Based on the revision of the types, the descriptions of *G. colossus*, *G. oerstedii* (Fr.) Torrend and *G. oregonense* are corrected.

In the key, besides the twelve species of *Ganoderma* subgenus *Ganoderma* from Mexico, other tropical and temperate species not known from Mexico are included, i.e. *Ganoderma capense* (Lloyd) Teng, *G. conccinum* Ryvar den, *G. dorsale* (Lloyd) Torrend, *G. longistipitatum* Ryvar den, *G. meredithiae* Adas. & Gilb., *G. multicornis* Ryvar den, *G. nitidum* Murrill, *G. orbiforme* (Fr.) Ryvar den, *G. ravenelii* Steyaert and *G. subfornicatum* Murrill. These species are taxonomically close related with Mexican species and were included in the key for comparison purposes and to facilitate the determination of the Mexican species. *Ganoderma tsugae* Murrill previously recorded from Mexico was not found in this study.

**KEY OF THE MEXICAN SPECIES** (also related not Mexican species are included)

1. Basidiomata light weighted, spongy, robust and generally large .....2
1. Basidiomata with other features .....4
  2. Pileus yellow, yellowish-orange, brownish-orange but never reddish-black; basidiospores 14–19 × 9–13 μm, reticulated ..... *G. colossus*
  2. Pileus with reddish-black tones .....3
3. Basidiomata rounded-flabelliform to occasionally reniform, 6–20 cm thick, context generally duplex; basidiospores 10–15 × 7–10 μm ..*G. oregonense*
3. Basidiomata reniform to flabelliform, up to 5 cm thick, generally stipitate, context homogeneous; basidiospores 9–13 × 6–8 μm.....*G. tsugae* (not in this study)
  4. Context duplex .. .....5
  4. Context relatively homogeneous to homogeneous .....12
5. Basidiomata stipitate, at times substipitate when growing on wood .....6
5. Basidiomata sessile to substipitate .....11
  6. Basidiospores with subacute apex .....7
  6. Basidiospores distinctively truncate .....9
7. Basidiomata generally less than 6 × 7 cm; basidiospores 12–14 × 8–9 μm.....8
7. Basidiomata generally bigger, basidiospores 15–17 × 10–11 μm .....
  - .....*G. longistipitatum* (not in this study)
  8. Context without resinous deposit, cuticle cells with incrustations in the apex.....*G. dorsale* (not in this study)
  8. Context with resinous deposit, cuticle cells without incrustations in the apex.....*G. concinnum* (not in this study)
9. Basidiospores oblong to cylindrical, 11–14 × 5–8 μm .....
  - .....*G. ravenelii* (not in this study)
9. Basidiospores ellipsoid to oblong; resinous bands in the context very evident; basidiospores 9–13 × 6–8 μm .....10
  10. Cuticle cells entire or occasionally with two to three protuberances.....*G. curtisii*
  10. Cuticle cells with many protuberances and branches; restricted to *Pinus* forest ..... *G. meredithiae* (not in this study)
11. Pileus with narrow zonation, context with resinous bands only in the base; cuticle cells very diverticulated, antler-like; basidiospores 11–13 × 7.5–8 μm .....*G. multicornum* (not in this study)
11. Pileus with wider zonation, context with resinous bands almost to the periphery; cuticle cells entire, basidiospores 12–16 × 8–10 μm ... *G. sessile*
  12. Cuticle cells entire or with occasional protuberances (maximum two).....13
  12. Cuticle cells distinctively diverticulated .....19
13. Basidiomata stipitate, basidiospores with subacute apex, pillars thick and partially anastomosed.....*G. perturbatum*
13. Basidiomata sessile to substipitate, basidiospores truncate, pillars different ..... 14

14. Context pale to very light brown, basidiospores with subfree pillars ..... 15
14. Context yellowish-brown to reddish-brown ..... 16
15. Pileus crust very hard, difficult to penetrate with fingernail, context bruising to yellow; cylindrical to narrowly clavate cuticle cells with granulations in the apex, basidiospores  $8-10 \times 6-7 \mu\text{m}$  ..... *G. weberianum*
15. Pileus crust easy to penetrate with fingernail, context not bruising; clavate cuticle cells, without granulations in the apex, basidiospores  $9-11 \times 6-8 \mu\text{m}$  ..... *G. sessiliforme*
16. Basidiospores with partially anastomosed pillars ..... 17
16. Basidiospores with free pillars ..... 18
17. Basidiomata broadly attached, plane; context without resinous deposit; basidiospores  $9-11 \times 5-7 \mu\text{m}$  ..... *G. capense* (not in this study)
17. Basidiomata generally with a contracted base to substipitate, concave to infundibuliform; context with resinous incrustations close to the base; basidiospores  $9-12 \times 6-8 \mu\text{m}$  ..... *G. subincrustatum*
18. Cuticle cells clavate, almost cylindrical to occasionally widely clavate, basidiospores  $11-14 \times 6-8 \mu\text{m}$  ..... *G. resinaceum*
18. Cuticle cells narrowly clavate to cylindrical with subcapitate apex, basidiospores  $8-10 \times 6-7 \mu\text{m}$  ..... *G. mexicanum*
19. Basidiomata woody-corky, very light weighted, pileus violet-brown with yellowish-orange shades; basidiospores  $11-14 \times 5-8 \mu\text{m}$ , oblong to cylindrical ..... *G. zonatum*
19. Basidiomata and pileus with other characteristics; basidiospores widely ellipsoid to ellipsoid and ellipsoid to oblong ..... 20
20. Basidiospores  $9-13 \times 6-8 \mu\text{m}$ , pillars  $< 0.4-0.6 \mu\text{m}$ , free ..... 21
20. Basidiospores with thick pillars ( $> 0.6 \mu\text{m}$ ) and partially anastomosed ..... 22
21. Cuticle cells with the clavate shape conserved, generally with one to three short and thick protuberances and/or branches ..... *G. nitidum* (not in this study)
21. Cuticle cells commonly with a constriction, one to two thick and long branches, up to seven short and thick protuberances ..... *G. subfornicatum* (not in this study)
22. Context with resinous bands evident throughout the context; cuticle cells occasionally with one to two thick and short branches and with up to seven short and thick protuberances; basidiospores  $9-13 \times 7-10 \mu\text{m}$ , widely ellipsoid to ellipsoid, partially anastomosed pillars, almost reticulated ..... *G. oerstedii*
22. Context with resinous incrustations only in the base of the basidiomata; cuticle cells with long branches, up to seven short and thick protuberances; basidiospores  $11-13 \times 6-8 \mu\text{m}$ , ellipsoid to oblong, with subfree pillars ..... *G. orbiforme* (not in this study)

## TAXONOMY

*Ganoderma colossus* (Fr.) C.F. Baker, *Brotéria* 425, 1918.

FIG. 1

≡ *Thomophagus colossus* (Fr.) Murrill, *Torreyia* 5: 197, 1905.

= *Ganoderma obockense* Pat., *Bull. Soc. Mycol. Fr.* 3: 119, 1887.

*Basidiomata* 13–26 × 16–24 cm, up to 9 cm thick, annual, sessile to substipitate, triquetrous, mainly single, never imbricate, soft, spongy, very light in weight, context thicker than tubes. *Pileus* rounded to flabelliform, convex to plane; surface glabrous, smooth, soft, glossy to dull, first shiny, then dull, with a laccate crust, cracked after drying, easy to remove and to penetrate with fingernail, without zonation; deep-yellow (4A8), lighter towards the periphery, then yellowish-orange (5B8) or eventually darkening with age; margin lighter than the base, entire, thick, rounded, smooth. *Context* 4.7–8 cm thick, soft, spongy, homogeneous, azonate, cream (4A3) to orange-white (5A3), without resinous bands. *Pores* 2–4 per mm, angular to rounded, soft; pore surface white to cream (4A2) when fresh, darkening to ochraceous or brownish-orange (6C5) when aging or drying; tubes 0.3–2 cm long, unstratified, pale to vinaceous-brown (8E4). *Hyphal system* dimitic in the context and hymenium, and trimitic in the pileipellis. *Contextual trama* with generative hyphae 1.6–3.2 µm diam., thin-walled, with large and conspicuous clamps, non-branched to generally branched, hyaline to yellowish, scarce and difficult to observe; sclerified generative hyphae up to 3.2 µm diam., yellowish, scarce; skeletal hyphae 3.2–5.6 µm diam., thick-walled (small lumen), non-septate, non-branched to moderately arboriform, yellowish, predominant; binding hyphae friable. *Hymenophoral trama* with generative hyphae 3.2–4.8 µm diam., thin-walled, with large and conspicuous clamps, with strong branching and very short cells, hyaline to yellowish, abundant and predominant; skeletal hyphae 3.2–4 µm diam., thick-walled, non-septate. *Pileipellis* with cuticle cells 32–75 × 5.6–14.4 µm diam, narrow to broadly clavate, with or without lateral or apical small protuberances, some with short branches in the apex, thin-walled, yellowish, negative in Melzer's reagent; generative hyphae 2.4–4 µm diam., thin-walled, with large and conspicuous clamps, generally branched, partly interwoven in dense clusters, hyaline to yellowish, abundant and predominant; sclerified generative hyphae up to 3.6 µm diam., subthick-walled up to 1 µm, branched, hyaline to yellowish, only observed in Congo Red; skeletal hyphae 2.4–6 µm diam., subthick-walled up to 0.8 µm, non-septate, non-branched to arboriform, yellowish; binding hyphae 1.6–2 µm diam., subthick-walled up to 0.8 µm, non-septate, yellowish. *Basidiospores* 14.4–19.2 × (8.8–) 9.6–11 (–13.2) µm, Q = 1.36–1.75, ellipsoid to oblong, apex acute to subacute, without visible apical germ, only observed in immature basidiospores, yellowish-brown, negative in Melzer's reagent; perisporium wrinkled, hyaline to reddish-yellow; exosporium with inter-walled pillars up to 0.7 µm thick, forming an incomplete reticule; endosporium wrinkled. *Basidia* 29.6 × 8 µm, clavate, hyaline. *Basidiohum* 19.2–24 × 8.8–12 µm, broadly clavate, hyaline. *Cystidia* 18.4–24 × 5.6–6.4 µm, conical to broadly conical, thin-walled, hyaline to yellowish.

*Specimens examined.* **Veracruz**, *G. Guzmán* 35708 (XAL), *F. Ventura* 12195 (ENCB), *J. Pérez-Ortiz* 1016 (ENCB); **Quintana Roo**, *G. Guzmán* 20516 (XAL); **Chiapas**, *G. Castillo* 2803 (XAL), *A.M. Suárez* 81 (ENCB).

*Other specimens examined.* **COSTA RICA**, LECTOTYPE of *Ganoderma colossus* (UPS); **SOMALIA**, LECTOTYPE of *Ganoderma obockense* (PC).

*Habitat.* Solitary, in secondary tropical forest, secondary low tropical rain forest or grassland; on wood, ground or volcanic sand.

*Distribution.* Tropical species recorded from Africa, Asia, Australia, Brazil, Costa Rica, USA and Venezuela. In Mexico its distribution is mainly in the tropical zone south of the country.

*Remarks.*—A spongy basidiomata, light weighted, laccate, easily indented crust, large and subreticulate basidiospores and thin-walled cuticle cells, make this a distinctive species. In general, the macro and micromorphological features, especially shape and size of basidiomata, basidiospores and cystidia, are consistent in specimens of this species. The Mexican specimens corresponded with the type except that the latter has abundant chlamyospores.

We checked type of *Ganoderma obockense*, and confirmed it as synonym of *G. colossus*, as previously established by Furtado (1965), Steyaert (1972) and Ryvarden and Johansen (1980). *Ganoderma colossus* is a rare species with remarkable features. It was reported from Mexico for the first time by Murrill (1905) from Yucatán, and it has been recorded from tropical and subtropical vegetation of Mexico (Guzmán 1977), Oaxaca (Welden and Guzmán 1978), Quintana Roo (Guzmán 1983) and Veracruz (Welden and Guzmán 1978). It is a new record for Chiapas.

***Ganoderma curtisii* (Berk.) Murrill, North Amer. Flora 9: 120, 1908.** FIG. 2

*Basidiomata* 4–9 × 5.5–19 × 1–1.6 cm, annual, stipitate, single, sometimes imbricate, corky to woody, context generally wider than tubes. *Pileus* rounded-flabelliform, reniform to circular, upper surface slightly convex to plane; surface glabrous, smooth to slightly tuberculate, soft, generally dull in immature specimens, shiny when mature, occasionally shiny all over the surface; with laccate crust, easy to penetrate with fingernail, generally easily removed; slightly radially rugose, some materials concentrically sulcate; maize (4A6), deep yellow (4A8), golden-yellow (5B8), yellowish-brown (5C8), violet-brown (11F8) to very dark violet-brown almost black, more or less homogeneous, or with zonations of these tonalities, occasionally covered by cinnamon (6D6) basidiospores; margin generally lighter, yellowish-white (4A2) to light brown (7E8), entire, at times lobulate, thick, rounded to truncate, smooth. *Stipe* 25–270 × 13–70 mm, lateral or eccentric to occasionally central, flattening to cylindrical, solid; surface dull to shiny, smooth, yellow pale (3A3), golden-yellow (4A4, 5B8) to red-wine almost black, generally darker than pileus, laccate crust that removes or not. *Context* 0.7–1.3 cm thick, corky to slightly fibrous, duplex, azonate, pale orange to light orange (5A3, 5A4), brown-sienna (6D7) to exceptionally yellowish-brown (6F8) close to tubes, with resinous bands up to the margin. *Pores* 2–4 per mm, angular to rounded; pore surface yellow (2A3), pale yellow (3A3) to golden-yellow (4A4, 5B8), darkening to brown (6D8) when bruising or aging; tubes 0.3–1 cm long, unstratified to occasionally stratified, pale vinaceous-brown to vinaceous-brown (8E4). *Hyphal system* di-trimitic. *Contextual trama* with generative hyphae up to



3.2  $\mu\text{m}$  diam., thin-walled, with large and conspicuous clamps, non-branched, hyaline to yellowish, scarce and difficult to observe, generally collapsed; skeletal hyphae 1.6–5.6  $\mu\text{m}$  diam., thick-walled to solid, non-septate, arboriform, yellowish to yellowish-brown, predominant; binding hyphae 3.2–4.8  $\mu\text{m}$  diam., thick-walled to solid, non-septate, yellowish to yellowish-brown. *Hymenophoral trama* as contextual trama. *Pileipellis* with cuticle cells 43.2–96  $\times$  7.2–16.4  $\mu\text{m}$ , broadly clavate, entire or with two to three apical or lateral protuberances, occasionally lateral or apical branched, thick-walled to solid, at times multistratified, yellowish, slightly amyloid in Melzer's reagent after 24 h; generative hyphae 2.4–3.2  $\mu\text{m}$  diam., thin-walled, with conspicuous clamps, hyaline to yellowish, abundant but difficult to observe; skeletal hyphae 1.6–4.8  $\mu\text{m}$  diam., thick-walled to solid, non-septate, arboriform, yellowish to yellowish-brown, predominant; binding hyphae 1.6–2  $\mu\text{m}$  diam., thick-walled to solid, non-septate, yellowish to yellowish-brown, not observed in some specimens, more easily observed in young basidiomata or close to the base of the pileus in adult specimens. *Basidiospores* (9.2–) 10.4–12 (-13.6)  $\times$  5.6–8  $\mu\text{m}$ , Q = 1.5–1.88, ellipsoid to oblong, apex truncate, with apical germ pore, yellowish-brown; perisporium wrinkled, hyaline to yellowish-red; exosporium with inter-walled pillars 0.5–0.6  $\mu\text{m}$  thick, subfree; endosporium wrinkled. *Basidia* 29  $\times$  7.2  $\mu\text{m}$ , clavate, hyaline, scarce. *Cystidia* absent.

*Specimens examined.* **Jalisco**, M.G. Torres-Torres 526, 527, 532, 541, 554 (IBUG), H. Orozco 5 (IBUG), L. Villaseñor-Ibarra 282 (IBUG), J. Mejía-Jiménez s.n. (IBUG), A.G. Valenzuela s.n. (IBUG), L. Guzmán-Dávalos 1723, 7447 (IBUG), J.A. Pérez de la Rosa s.n. (IBUG); **Hidalgo**, J. Gimete 152-A (ENCB); **Morelos**, M. Frias Neve 18 (ENCB).

*Other specimens examined.* **USA**, HOLOTYPE of *Ganoderma ravenelii* (K).

*Habitat.* Solitary or gregarious; in oak, oak-pine and mesophytic forests; on wood or commonly on ground.

*Distribution.* Recorded from Africa, China, India, Japan, Mexico and USA.

*Remarks.*—The species is characterized mainly by its stipitate basidioma, pileus color and the lacquer that disappears very easily, combined with its occurrence in temperate or subtropical forests, but always associated to oak. Other diagnostic features are the duplex context with resinous bands. *Ganoderma curtisii* has an apparent wide distribution, nevertheless was not recorded by Corner (1983) neither by Gilbertson and Ryvarden (1986). In Mexico, *G. curtisii* is a common species in oak and pine-oak forests, but also was found in subtropical forests with oak.

*Ganoderma mexicanum* Pat., Bull. Soc. Myc. Fr. 14: 54, 1898.

FIG. 3

*Basidiomata* 9.5  $\times$  15 cm, up to 2 cm thick in the base, annual, sessile to substipitate, single, woody, but light in weight when dried, context wider than tubes. *Pileus* flabelliform, applanate; surface glabrous, rugose, hard, semiglossy to dull, with a laccate crust, thin, not easily cracked or removed, but easy to penetrate with fingernail, slightly concentrically sulcate; reddish-black close to the base to brown-violet (11F5) in the periphery, almost homogeneous; margin henna (7E8) to concolorous, lobulate, thin, smooth. *Context* up to 1.5 cm thick in the base, corky to slightly fibrous, homogeneous, azonate, cinnamon (6D7), without resinous bands. *Pores* 3–4 per mm, angular to rounded; pore surface cinnamon

(6D7); tubes up to 0.5 cm thick, unstratified, cinnamon (6D7). *Hyphal system* dimitic. *Contextual trama* with generative hyphae not observed; skeletal hyphae yellowish, damaged. *Hymenophoral trama* as contextual trama. *Pileipellis* with cuticle cells  $36\text{--}80 \times 5.6\text{--}12 \mu\text{m}$ , narrowly clavate to cylindrical, sometimes with a broad apex, generally without protuberances neither branches, thick-walled, multistratified, yellowish, dextrinoid in Melzer's reagent but apex slightly grayish; generative hyphae  $3.2\text{--}4 \mu\text{m}$  diam., thin-walled, with conspicuous clamps, hyaline; skeletal hyphae  $5.6\text{--}9.6 \mu\text{m}$  diam., solid to thick-walled, commonly septate, generally not arboriform, apex obtuse, yellowish, slightly grayish in Melzer's reagent. *Basidiospores*  $8\text{--}10 \times 5.6\text{--}8 \mu\text{m}$ ,  $Q = 1.41\text{--}1.47$ , ellipsoid, apex shortly truncate, with apical germ pore, dark yellow, negative in Melzer's reagent; perisporium smooth, reddish-yellow; exosporium with inter-walled pillars less than  $0.4 \mu\text{m}$  thick, free; endosporium smooth. *Basidia* not observed. *Cystidia* absent.

*Specimens examined.* **Mexico**, Tepalcingo, LECTOTYPE (FH-4823).

*Habitat.* On hardwoods.

*Distribution.* Known only from the type locality.

*Remarks.*—The type is in a bad state, but some remarkable features were observed: narrowly clavate cuticle cells and unbranched to scarcely branched, skeletal hyphae generally with obtuse apex, in combination with small basidiospores with an apparently smooth perisporium. The species has not been mentioned in subsequent studies neither has been recorded by Mexican authors; nevertheless, we consider it as a valid species by its unique features.

***Ganoderma oerstedii* (Fr.) Torrend, Bull. Torrey bot. Club 29: 606, 1902.** FIG. 4  
= *Ganoderma tuberculosum* Murrill, North American Flora 9: 123, 1908.

*Basidiomata*  $8\text{--}25 \times 7\text{--}26 \times 3\text{--}4.2$  cm, average 2.5–3.5 cm thick, up to 3.5–4.5 cm thick in the base, perennial, sessile, widely attached, single to generally imbricate, woody, generally context narrower than tubes, but in young specimens the context wider than tubes. *Pileus* flabelliform, rounded-flabelliform to semicircular, plane, thickening towards the base; surface glabrous, bumpy, soft when fresh, hard when drying, generally glossy, shiny not easily lost, with a laccate crust, not cracking, not easily removed, easy to penetrate with fingernail, concentrically sulcate; reddish-brown (9F8, 8F8) close to the base, gradually changing to orange-brown, henna (7E8) to deep yellow (5B8) toward the margin in some specimens, or generally fully violet-brown (10F8, 11F6, 11F7), at times terra-cotta (7D7) by basidiospores over the surface; margin whitish, lighter than pileus to concolorous, entire to slightly lobulate, thin to thick, rounded to obtuse, smooth to sulcate. *Context* 1.5–4 cm thick, average 2–3 cm, thinner toward the periphery, fibrous, relatively homogeneous, concentrically zonate, yellowish-brown (6F8) to orange-brown or dark reddish-brown (7F6, 7F8) close to the tubes, with resinous bands from the base to half or more of the basidioma. *Pores* 4–5 per mm, angular to rounded, woody; pore surface whitish, yellowish-white (1A2) to pale yellow (1A3), butter-yellow (4A5) when fresh, darkening to ochraceous when aging, bruising or drying; tubes 0.6–1.8 cm long, individual layer 0.4–0.9 cm, unstratified to stratified, concolorous with lower part of the context. *Hyphal system* trimitic. *Contextual trama* with generative hyphae not observed; skeletal hyphae  $3\text{--}4.8 \mu\text{m}$  diam., solid, non-septate, non-branched to arboriform, golden-

yellow to yellowish-brown. *Hymenophoral trama* as contextual trama. *Pileipellis* with cuticle cells  $56\text{--}88 \times 4.8\text{--}18 \mu\text{m}$  diam., narrowly clavate to clavate, generally with two branches and up to seven short and thick protuberances, thick-walled, golden-yellow, first reddish in groups, negative to occasionally slightly amyloid in Melzer's reagent after 36 h; generative hyphae  $3.2\text{--}4.8 \mu\text{m}$  diam., thin-walled, non-septate, branched, hyaline; skeletal hyphae  $3.2\text{--}7.6 \mu\text{m}$  diam., thick-walled to solid, septate to non-septate, arboriform with many branches, golden-yellow to yellowish-brown; binding hyphae  $3.0\text{--}6.2 \mu\text{m}$  diam., solid, non-septate, yellowish to yellow, difficult to observe. *Basidiospores*  $10.4\text{--}13.6 \times 7.2\text{--}8.8$  ( $-9$ )  $\mu\text{m}$ ,  $Q = 1.3\text{--}1.5$ , ellipsoid, apex truncate, with apical germ pore, yellowish-brown, negative in Melzer's reagent; perisporium wrinkled, hyaline; exosporium with inter-walled pillars up to  $0.6 \mu\text{m}$  thick, partially anastomosed; endosporium wrinkled. *Basidia* not observed. *Cystidia* absent.

*Specimens examined.* Guerrero, M.G. Torres-Torres 546 (IBUG); Jalisco, M.G. Torres-Torres 408, 563 (IBUG), O. Cárdenas-Hernández 12 (IBUG), A. Gaspar 25 (IBUG), J.J. Manzano 480 (IBUG); Oaxaca, M.G. Torres-Torres 573, 575 (IBUG).

*Other specimens examined.* COSTA RICA, NEOTYPE of *Ganoderma oerstedii* (UPS). HONDURAS, LECTOTYPE of *Ganoderma tuberculosum* (NY).

*Habitat.* Solitary or gregarious; evergreen tropical forests or secondary tropical or subtropical vegetation; on living trees and dead wood.

*Distribution.* Recorded from Argentina, Caribbean Island, Costa Rica, and Honduras. Now from Mexico.

*Remarks.*—The diagnostic characters of this species are the color of the basidiomata and context with resinous bands; furthermore, cuticle cells with protuberances and/or branches and its partially anastomosed basidiospore pillars. The examined specimens are in accordance with the types mentioned above, except that these have generally wider ellipsoid basidiospores ( $Q = 1.17\text{--}1.51$  in *G. oerstedii* neotype, and  $Q = 1.17\text{--}1.4$  in *G. tuberculosum* holotype). *Ganoderma oerstedii* is together with *G. subincrustatum* the most common species in urban zones all over the country (especially as a parasite of many kinds of trees). The specimens were incorrectly identified and deposited in herbaria as *G. lucidum*, *G. resinaceum* and *G. sessile*. This is the first record of *G. oerstedii* for Mexico.

***Ganoderma oregonense* Murrill, North Amer. Flora 9: 118, 1908.** FIG. 5

*Basidiomata*  $7\text{--}23 \times 10\text{--}30$  cm, up to 12 cm thick, annual, mainly sessile to substipitate, generally with contracted base, single or rarely imbricate, soft when fresh, very light in weight when dry, generally context wider than tubes. *Pileus* rounded-flabelliform, convex; surface glabrous, smooth to occasionally bumpy after drying, spongy, dull to semiglossy with remainders of shine, with a laccate crust, cracked after drying and easy to penetrate with fingernail but not easy to remove, azonate to slightly zonate; dark reddish-brown to almost black close to the base, then violet-brown (10F) to henna (7E8), lighter in the periphery, some specimens completely reddish-black; margin lighter than the base to concolorous, entire, thick, rounded, smooth. *Substipe* when present  $4\text{--}13 \times 4\text{--}4.5$  cm, cylindrical, reddish-black, darker than pileus. *Context*  $2.4\text{--}11.3$  cm thick, soft-corky, duplex, azonate, orange-white (5A2) or cream-orange-pink above, light brown or sunburn to raw-sienna (6D5, 6D7) near the tubes, with an apricot (5B6)

thin fringe below the laccate crust, without resinous bands. *Pores* 3–4 per mm, angular to rounded; pore surface yellowish-white (3A2) to orange-brown or raw-sienna (6D7); tubes 0.3–0.6 cm thick, stratified to unstratified, fragile, light vinaceous-brown, almost concolorous with the lower part of the context. *Hyphal system* trimitic. *Contextual trama* with generative hyphae up to 5.2  $\mu\text{m}$  diam., thin-walled, with conspicuous clamps, non-branched, hyaline, scarce and difficult to observe; skeletal hyphae 2.4–10  $\mu\text{m}$  diam., thick-walled (narrow lumen), septate near the apex, non-branched to arboriform, hyaline to light amber-brown, predominant; binding hyphae 4–7.2  $\mu\text{m}$  diam., thick-walled, non-septate, hyaline to amber-brown, abundant. *Hymenophoral trama* with generative hyphae 3.2–4.8  $\mu\text{m}$  diam., very short cells, thin-walled, with large and conspicuous clamps, with many branches, hyaline to yellowish, abundant and predominant; skeletal hyphae 3.2–4  $\mu\text{m}$  diam., thick-walled, non-septate, arboriform, negative in Melzer's reagent; binding hyphae up to 4  $\mu\text{m}$  diam., thick-walled, non-septate, hyaline to yellowish. *Pileipellis* with cuticle cells 44–102.4  $\times$  6.4–20  $\mu\text{m}$  diam., narrow to broadly clavate, generally with conspicuous basal clamps, without protuberances or only a single one, thick-walled, yellowish, strongly to slightly amyloid in Melzer's reagent; generative hyphae 2.4–5.6  $\mu\text{m}$  diam., thin-walled, with conspicuous clamps, hyaline; skeletal hyphae 3.2–9.6  $\mu\text{m}$  diam., thick-walled up to 0.8  $\mu\text{m}$  diam., non-septate, generally arboriform, yellowish; binding hyphae 3.2–8  $\mu\text{m}$  diam., thick-walled, non-septate, yellowish. *Basidiospores* 10.8–14.4  $\times$  7.2–8.8 (–9.6)  $\mu\text{m}$ ,  $Q = 1.5$ –1.78, ellipsoid to oblong, apex subacute, without visible apical germ pore, only observed in immature basidiospores, yellowish-brown, negative in Melzer's reagent; perisporium wrinkled, exosporium with inter-walled pillars 0.6–0.8  $\mu\text{m}$  thick, subfree; endosporium wrinkled. *Basidia* 24–40  $\times$  7.2–8.8  $\mu\text{m}$ , clavate, hyaline. *Cystidia* 16–28  $\times$  3.2–5.6  $\mu\text{m}$ , fusiform to narrowly utriform, some with scarce protuberances, thin-walled, hyaline to yellowish.

*Specimens examined.* **Estado de Mexico**, *S. Acosta* 653 (IBUG), *G. Guzmán* 4675, 4939 (ENCB), *E. González* 282 (ENCB); **Hidalgo**, *M.L. Aguirre-Jones s.n.* (ENCB); **Veracruz**, *G. Guzmán* 28886 (XAL).

*Other specimens studied.* **USA**, LECTOTYPE of *Ganoderma oregonense* (NY); HOLOTYPE of *Ganoderma sequoiae* (NY).

*Habitat.* Solitary; in *Pinus hartwegii*, *Pinus-Abies*, *Abies* spp., *Abies religiosa* forests; on wood of *Picea*, *Pinus* or *Abies*.

*Distribution.* Species recorded from Alaska, Canada, Central and South America, Mexico and USA.

*Remarks.*—The more important macromorphological features for its identification are the dark color of the pileus, contrasting with the pale colored context, and the large and spongy basidiocarp very light in weight. Cystidia have previously not been described for this species. The lectotype of *G. oregonense* has relatively smaller basidiospores, 10.8–12.8 (–13.6)  $\times$  7.2–8  $\mu\text{m}$  diam., than the Mexican specimens. A related species is *G. sequoiae* Murrill, which has basidiospores (12.8–) 13.6–16 (–17.6)  $\times$  (7.2–) 8–9.6  $\mu\text{m}$ . The lectotype of *G. oregonense* is in a bad state, including its context, in which a duplex character was not observed. In the nomenclatural study of Moncalvo and Ryvarden (1997), under the type specimen of *G. oregonense* the date is not mentioned and the name of the collector is given as the locality. *Ganoderma nevadense* Murrill was given as a

synonym of *G. oregonense* by Steyaert (1980) and Ryvarden (1985) but the type was not examined in this study. A specimen from Quintana Roo of *G. colossus* was mistakenly identified as *G. oregonense* by Guzmán (1963), and later corrected by him (Guzmán 1983).

***Ganoderma perturbatum* (Lloyd) Torrend, Bróteria Bot. 18: 34, 1920. FIG. 6**

*Basidiomata* 4.5–5 × 5.5–7.5 × 0.8–1.3 cm, perennial, stipitate, single, never imbricate, corky to woody, context slightly thinner or almost as wide than tubes. *Pileus* reniform, convex to plane; surface glabrous, bumpy, hard but possible to penetrate with fingernail, mainly very shiny although dull regions might be observed, with a laccate crust, difficult to remove, generally with abundant radial zonation, totally violet-brown (darker than 11F7 or 11F8) to fully black, homogenous, occasionally covered with cinnamon (6D6) basidiospores; margin generally lighter than pileus, oxide-red (8E8), entire, thick, obtuse to truncate, sulcate. *Stipe* 5.5–13 × 1.1–1.2 cm, lateral, cylindrical, solid, surface shiny, violet-brown (11F8) very dark to black, concolorous to generally darker than pileus. *Context* 0.3–0.5 cm thick, fibrous, relatively homogeneous, azonate, raw-sienna or light brown (6D7), gradually changing to slightly darker next to the tubes, resinous incrustations not very visible. *Pores* 3–4 per mm, angular to rounded, woody; pore surface yellowish-white (3A2) to pale yellow (4A3), darkening to brown (6D8) when bruising or aging; tubes 0.4–0.8 cm long, indistinguishable stratified, concolorous with context. *Hyphal system* dimitic. *Contextual trama* with generative hyphae 2.4–3.6 µm diam., thin-walled, with large and conspicuous clamps, branched, hyaline, scarce and difficult to observe; skeletal hyphae 2.4–7.2 µm diam., generally solid to thick-walled, non-septate, arboriform, yellowish to yellowish-brown. *Hymenophoral trama* as contextual trama. *Pileipellis* with cuticle cells 40–64 × 7.2–16 µm, broadly clavate to occasionally conic, without or up to two diverticules, mainly solid to thick-walled, multistratified wall, generally with a distinctive darker inner stratum, with refringent content, yellow to yellowish-brown in group, content immediately black with Melzer's reagent, walls slightly amyloid after 14 h; generative hyphae 2.8–4 µm diam., thin-walled, conspicuous clamps, hyaline, abundant close to cuticle cells, difficult to observe; skeletal hyphae 2.4–2.8 µm diam., generally thick-walled to solid, apex septate, arboriform, yellowish-brown. *Basidiospores* 11.2–12.8 × (7.6–) 8–9.6 µm, Q = 1.25–1.5, broadly ellipsoid to ellipsoid, apex subacute, slightly visible apical germ pore, yellowish-brown; perisporium wrinkled, exosporium with inter-walled pillars 0.72–0.8 µm thick, partially anastomosed; endosporium wrinkled. *Basidia* not seen. *Cystidia* absent.

*Specimens examined.* **Colima**, Grupo Ecológico Forestal Tonatiuh s.n. (IBUG).

*Other specimens studied.* **BRAZIL**, LECTOTYPE of *Ganoderma perturbatum* (BPI, Lloyd herb. num. 55740).

*Habitat.* Solitary or gregarious; tropical forests; on wood or ground.

*Distribution.* Brazil and Mexico

*Remarks.*—The species may easily be recognized by its dark and shiny pilear surface, the remarkable subacute basidiospores and the characteristic cuticle cells. Few species in *Ganoderma* have broad ellipsoid basidiospores with subacute apex and cuticle cells with a distinctive dark yellow inner stratum. In the type specimen, the cuticle cells are generally wider (48.8–88 × 7.2–20 µm) than in the

specimens examined. *Ganoderma perturbatum* is rare and was only known from the type locality. It is presented now the first record in Mexico.

*Ganoderma resinaceum* Boud., *Bull. Soc. Mycol. Fr.* 5: 72, 1889.

FIG. 7

*Basidiomata* 9–12.5 × 7–10 cm, up to 2 cm thick in the base, annual, single to imbricate, woody-spongy, context narrower than tubes. *Pileus* rounded-flabelliform, convex to plane; surface glabrous, smooth to slightly bumpy, slightly soft, glossy, with a laccate crust, cracked, easy to remove and to penetrate with fingernail, broadly zonate; violet-brown (11F7), darker than 11F7 to dark red in almost all the surface, gradually changing to orange (5A7) toward the margin or fully violet-brown (11F7), darker in adult basidiomata, with terra-cotta (7D7) basidiospores over the surface; margin brownish-orange to concolorous, entire, thin to thick, acute to rounded, smooth. *Context* 0.4–1 cm thick, fibrous-spongy, homogeneous, azonate, light reddish-brown, with an apricot (5B6) thin fringe below the laccate crust, without resinous bands. *Pores* 3–5 per mm, angular to rounded, woody; pore surface yellow (3A2) when fresh, darkening to ochraceous or yellowish-brown (6C5) when aging or drying; tubes 0.5–1 cm long, unstratified, concolorous with the context. *Hyphal system* dimitic. *Contextual trama* with generative hyphae not observed; skeletal hyphae 1.6–8 µm diam., thick-walled, generally solid, non-septate, non-branched to arboriform, moderately branched, golden-yellow. *Hymenophoral trama* as the contextual trama. *Pileipellis* with cuticle cells 46.5–71.3 × 8.6–13.2 µm diam, clavate, almost cylindrical to clavate, generally without protuberances neither branches, some with occasional lateral or apical protuberances and/or branches, thick-walled to solid, apex with granulations, brownish-yellow, amyloid in Melzer's reagent; generative hyphae 2.4–3.1 µm diam., thin-walled, with conspicuous clamps, hyaline to yellowish, abundant; skeletal hyphae 3.1–6.2 µm diam., thick-walled to occasionally solid, non-septate, arboriform, with few branches, yellowish-brown. *Basidiospores* (10.5–) 11.2–13.6 × 6.5–7.4 (–8.1) µm, Q = 1.29–1.5, ellipsoid, apex truncate, with visible apical germ pore, yellowish-brown; perisporium smooth, hyaline; exosporium with inter-walled pillars 0.3–0.4 µm thick, free; endosporium wrinkled. *Basidia* not observed. *Cystidia* absent.

*Specimens examined.* Colima, G. Guzmán 11647 (IBUG).

*Habitat.* Mainly gregarious; in tropical forests; on wood.

*Distribution.* Pantropical extending into warmer parts of the temperate zone.

*Remarks.*—Macromorphologically *Ganoderma resinaceum* may be confused with many species: *G. boninense* Pat., *G. praelongum* Murrill and *G. pulverulentum* Murrill, among others. Nevertheless, the particular combination of characters it makes possible to identify without major problems. The basidiomata color, the corky-spongy context without resinous bands, almost cylindrical to clavate cuticle cells, and the ellipsoid basidiospores, with free and relatively thin pillars are diagnostic features of the species. Many specimens are deposited in Mexican herbaria and published as *G. resinaceum* or *G. sessile* (i.e. Welden and Guzmán 1978, Guzmán-Dávalos and Guzmán 1979, Ojeda-López et al 1986, Anel and Guzmán 1987), but they correspond to *G. subincrustatum* or *G. oerstedii*. The truth is that *Ganoderma resinaceum* is rare in Mexico, and only a single collection has been recorded.

*Basidiomata* 5.5–13 × 5–13 cm, 1–3 cm thick in the base, annual, sessile, single to imbricate, woody-corky, light in weight, context the same wide than the tubes. *Pileus* semicircular, rounded flabelliform to flabelliform, conchate to convex; surface glabrous, bumpy, slightly to radially rugose, hard, glossy, with a laccate crust, not cracking, slightly easy to remove, easy to penetrate with fingernail, concentrically sulcate mainly toward the margin; violet-brown (10F6) or photo-brown (9F8) in the 80 to 90% of the surface, reddish-brown (8F8) to brownish-orange (6C8) in the periphery, or fully violet-brown very dark almost black, occasionally with raw sienna (6D7) basidiospores over the surface; margin whitish, henna (7E8), lighter than pileus or concolorous, entire, thin, rounded to acute, smooth. *Context* up to 1.5 cm thick in the base, 0.7–0.9 cm average, fibrous-corky, duplex, azonate, pale-orange to light-orange (5A3, 5A4) above and reddish-golden to light brown (6C7) close to the tubes; resinous bands generally diffuse and difficult to observe, almost up to the margin. *Pores* 4–5 per mm, angular to rounded, woody; pore surface yellowish-white (3A2), darkening to brown (6D8) when bruising or aging; tubes 0.8–1 cm thick, up to 1.4 cm in the base, unstratified, generally concolorous with the inferior part of the context. *Hyphal system* trimitic. *Contextual trama* with generative hyphae 2.4–4 µm diam., thin-walled, with large and conspicuous clamps, unbranched, hyaline to yellowish, difficult to observe; skeletal hyphae 1.6–12 µm diam., generally solid, non-septate, arboriform, very branched, yellowish to golden-yellow, predominant; binding hyphae 1.6–4 µm diam., solid, non-septate, hyaline to yellowish, scarce. *Hymenophoral trama* as contextual trama. *Pileipellis* with cuticle cells (40.3–) 60–88 × 8–16 µm, clavate, generally without or with one lateral protuberance, thick-walled, generally multistratified, golden-yellow, content immediately black with Melzer's reagent, cells amyloid immediately; generative hyphae 1.6–4 µm diam., thin-walled, with conspicuous clamps, branched, hyaline to yellowish, abundant; skeletal hyphae 2.4–9.6 µm diam., generally solid, non-septate, arboriform, yellow, predominant; binding hyphae 1.6–4 µm diam., generally solid to thick-walled, non-septate, hyaline to yellowish, notably thinner and paler than skeletal hyphae, in some specimens not observed. *Basidiospores* 11.2–14.4 (–16.4) × 7.2–8.8 µm, Q = 1.5–1.86, ellipsoid to oblong, apex truncate, with apical germ pore, yellowish-brown, negative in Melzer's reagent; perisporium wrinkled, hyaline; exosporium with inter-walled pillars 0.56–0.64 µm thick, subfree; endosporium wrinkled. *Basidia* not seen. *Cystidia* absent.

*Specimens examined.* **Hidalgo**, S. & J. Peck s.n. (SP 124133, ex-BCI 3079); **Jalisco**, G. Guzmán 17888 (IBUG), G. Nieves 27-A (IBUG).

*Other specimens examined.* USA, LECTOTYPE (NY); L.M. Underwood s.n., May 1897, (NY); Bedford Park, on stumps of oak, 1 Jun 1902, s.coll. (NY); s.coll. (NY 12.123, Timber and Forest Diseases Survey); L.M. Underwood s.n., May 1894, (NY); W.N. Loug s.n., 28 Nov 1911 (NY).

*Habitat.* Mainly solitary; subtropical vegetation; on living trees (i.e. *Salix* sp. and *Quercus* sp.) or on dead deciduous wood.

*Distribution.* Recorded from Argentina, Mexico and USA.

*Remarks.*—The more distinctive features of the species are the basidiomata color, the spongy-corky consistency, duplex context with resinous bands which at times may be difficult to observe and basidiospores with short, thick and subfree pillars.

The Mexican specimens are in accordance with the lectotype and with the description of Gottlieb and Wright (1999).

Since Murrill described *G. sessile* based on three specimens (Murrill 1902), out of which he did not select any type, there have been many wrong interpretations of the typification. Later, Murrill (1908) cited New York as the type locality and mentioned more specimens. Nowadays five different specimens are marked as types in NY. Bazzalo and Wright (1982) mentioned the following: "New York, Bedford Park, V-1902 (Holotype of *G. sessile* Murr., NY)". On the other hand, Ryvar den (1985) selected the specimen "New York, Bedford Park. 1. Jan. 1902. Collector unknown" as the neotype. However, Moncalvo and Ryvar den (1997) and Ryvar den (2000) considered the collection NY 12.123 as the lectotype, because the neotypification of Ryvar den (1985) was considered superfluous. Moreover, Gottlieb and Wright (1999) selected the specimen Underwood s.n., considered by Murrill (1908), as the lectotype. The basidiospores in the Murrill's original description are  $9\text{--}11 \times 6\text{--}8 \mu\text{m}$ , which are different from the basidiospores in all the "type specimens", which measure  $12\text{--}14.4 \times 7.2\text{--}8.8 \mu\text{m}$ .

*Ganoderma sessile* has been cited from many localities in USA and Mexico, but because it has previously been treated as a synonymy of *G. resinaceum* the distribution is uncertain. Gottlieb and Wright (1999) cited it from Argentina. *Ganoderma sessile* is one of the most commonly cited species from Mexico (e.g. Guzmán 1977). Nevertheless the majority of the specimens in Mexican herbaria correspond to *G. oerstedii* and *G. subincrustatum*. *Ganoderma sessile* s.str. is rare in Mexico and the collections cited above are the first confirmed records.

***Ganoderma sessiliforme* Murrill, Bull. New York. Bot. Gard. 8: 149, 1912.**

FIG. 9

*Basidiomata*  $3.5 \times 5$  cm, up to 1.5 cm thick in the base, very thin towards the margin, annual, sessile to substipitate, single or occasionally imbricate, woody, but light in weight. *Pileus* flabelliform, somewhat concave to convex; surface glabrous, radially rugose, hard, glossy, with a laccate crust, not cracking, not easily removed, penetrable with fingernail, slightly concentric-sulcate; violet-brown (11F8), then caput-mortuum (8F7) in the 80% of the surface to reddish-brown (8E8), deep orange (6A8) in the periphery, occasionally with cinnamon (6D6) basidiospores over the surface; margin whitish, generally lighter than pileus, entire, thin, smooth. *Substipe*  $2 \times 1\text{--}1.5$  mm, lateral, flattened, solid, surface shiny, red-wine to almost black, darker than pileus. *Context* 0.3 cm average, up to 0.7 cm thick in the base, fibrous, homogeneous to relatively homogeneous, azonate, orange-white (5A2), with a deep yellow (4A8) fringe below the laccate crust, without resinous bands. *Pores* 3–5 per mm, angular to rounded, woody; pore surface yellowish-white (3A2), darkening to brown (6D8) when bruising or aging; tubes 0.1–0.3 cm thick, up to 1 cm in the base, unstratified, sunburn (6D7), darker than context. *Hyphal system* trimitic. *Contextual trama* with generative hyphae  $1.6\text{--}3.2 \mu\text{m}$  diam., thin-walled, with large and conspicuous clamps, non-branched, hyaline to yellowish; skeletal hyphae  $2.4\text{--}9.6 \mu\text{m}$  diam., mainly solid to some thick-walled, non-septate, arboriform, scarcely branched, yellowish, predominant; binding hyphae  $1.6\text{--}3.2 \mu\text{m}$  diam., solid, non-septate, yellowish, scarce. *Hymenophoral trama* as the contextual trama. *Pileipellis* with cuticle cells  $32\text{--}52$  ( $\sim 60.8$ )  $\times 8\text{--}12 \mu\text{m}$ , clavate,



generally without or with scarce lateral diverticules, thick-walled, at times multistratified, golden-yellow, content immediately black with Melzer's reagent, cells amyloid after 48 h, predominant the shorter cells; generative hyphae 2.4–4  $\mu\text{m}$  diam., thin-walled, with conspicuous clamps, hyaline to yellowish, abundant, difficult to observe; skeletal hyphae 1.6–7.2  $\mu\text{m}$  diam., mainly solid to some thick-walled, non-septate, arboriform but with few branches, yellowish-brown, predominant; binding hyphae 1.2–2.4  $\mu\text{m}$  diam., solid to thick-walled, non-septate, hyaline to yellowish, notably thinner and paler than skeletal hyphae. *Basidiospores* 8.8–11.2 (–12)  $\times$  6.4–8 (–8.4)  $\mu\text{m}$ ,  $Q = 1.28\text{--}1.65$ , ellipsoid to oblong, apex truncate, with apical germ pore, yellowish-brown, negative in Melzer's reagent; perisporium wrinkled, hyaline; exosporium with inter-walled pillars 0.56–0.64  $\mu\text{m}$  thick, subfree and in some basidiospores free; endosporium wrinkled. *Basidia* not observed. *Cystidia* absent.

*Specimens examined.* **Morelos**, LECTOTYPE of *Ganoderma sessiliforme* (NY), G. Guzmán 2078 (ENCB).

*Habitat.* Mainly gregarious; tropical forest with *Quercus*; on dead wood.

*Distribution.* Recorded from Argentina and Mexico.

*Remarks.*—The more important distinctive features of this species are the thin, flabelliform, conchate pileus, light context without resinous bands, basidiospores with short, thick and subfree pillars and generally short cuticle cells. The studied specimens, including the type, are in accordance with the description of Gottlieb and Wright (1999), who recorded it from Argentina. *Ganoderma sessiliforme* has not been mentioned from Mexico since it was described in 1912, thus, the one cited above is the second record from the country and it comes from the same region as the type.

***Ganoderma subincrustatum* Murrill, North Amer. Flora 9: 122, 1908.** FIG. 10

*Basidiomata* 7.5–10  $\times$  12–12.5 cm, average 1.5 cm thick, up to 2.3 cm thick in the base, perennial, substipitate to stipitate, with a contracted base, single to imbricate, woody, context wider than tubes. *Pileus* flabelliform, rounded-flabelliform to circular, concave to infundibuliform; surface glabrous, slightly bumpy, soft when fresh, hard when drying, generally glossy, in some changing to dull, with a laccate crust, not cracking, not easily removed, easy to penetrate with fingernail, concentrically sulcate; reddish-brown (9F8, 8F8) to burn-sienna (7D8) close to the base, gradually changing to henna (7E8), deep yellow (5B8) toward the margin in some specimens, with age fully dark reddish-brown, at times terracotta (7D7) basidiospores over the surface; margin whitish or lighter than pileus, entire to slightly lobulate, thick, rounded, smooth. *Stipe* 1.5–4.3  $\times$  1.5–2 cm, short to large and thick, cylindrical, lateral to central, solid, reddish-black, darker than pileus. *Context* 1.1–1.7 cm thick, average 0.5–0.7 cm, fibrous-corky, relatively homogeneous, concentrically zonate, a narrow apricot (5B6) zone close to the pileus, and otherwise dark-brown (7F7), with discontinuous resinous bands from the base to half or more of the basidiomata, with whitish mycelium close to the base. *Pores* 4–6 per mm, angular to rounded, woody; pore surface whitish, yellowish-white (3A2) to pale yellow (4A2, 4A3) when fresh, darkening to ochraceous or yellowish-brown (6C5) when bruising; tubes 0.4 cm long, lighter to concolorous with lower part of the context. *Hyphal system* trimitic. *Contextual trama* with generative hyphae not observed; skeletal hyphae 3–4.8  $\mu\text{m}$  diam.,

solid, non-septate, non-branched to arboriform, golden-yellow to yellowish-brown. *Hymenophoral trama* as contextual trama, generative hyphae 3.2–5.6 µm diam., thin-walled, non-septate, branched, hyaline; apex of the hyphae visible in the tube lumen, simple or branched, rounded to tapering, or submoniliform, hyaline. *Pileipellis* with cuticle cells 32–80 (–96) × 5.6–14.4 µm diam., narrow clavate to clavate, generally with one or two protuberances or branches, thick-walled, golden-yellow, first reddish in group, negative to occasionally slightly amyloid in Melzer's reagent after 36 h; generative hyphae not observed; skeletal hyphae 3.2–7.6 µm diam., solid to thick-walled, septate to non-septate, arboriform with many branches, golden-yellow to yellowish-brown; binding hyphae 3.2–5.6 µm diam., solid, non-septate, yellowish to yellow, scarce. *Basidiospores* 9.6–12.4 × 7.2–8.4 µm, Q = 1.3–1.58, ellipsoid, few broadly ellipsoid, apex truncate, with apical germ pore, yellowish-brown; perisporium wrinkled, hyaline; exosporium with inter-walled pillars up to 0.6 µm thick, partially anastomosed, endosporium wrinkled. *Basidia* not observed. *Cystidia* absent.

*Specimens examined.* **Jalisco**, *O. Vargas* 316 (IBUG), *G. López-Damián* 49 (IBUG), *G. Nieves* 56 (IBUG); **Nuevo León**, *González-Velásquez* 556 (ENCB); **Quintana Roo**, *R. Valenzuela* 6429 (ENCB); **Veracruz**, *C. Rojas* s.n. (ENCB), *G. Guzmán* 2873 (ENCB), *F. Ventura* 1312 (ENCB).

*Other specimens examined.* **JAMAICA**, LECTOTYPE of *Ganoderma subincrustatum* (NY).

*Habitat.* Solitary or gregarious; in evergreen tropical forest, deciduous tropical forest, *Pinus-Quercus* forests, *Pinus-Eucalyptus* artificial forest, xerophytic bush or subtropical secondary vegetation; on living trees, on dead wood or on the ground as parasite of roots.

*Distribution.* Recorded from Argentina and Jamaica, and now from Mexico.

*Remarks.*—*Ganoderma subincrustatum* is macromorphologically similar to *G. boninense*, *G. oerstedii* and *G. pulverulentum* by the colors of pileus surface and context. Macromorphologically, this species is recognized by the gradual change of the pileus colors, the concave to infundibuliform, and generally rounded-flabelliform to circular pileus, the relatively homogeneous context without resinous bands in the periphery and the basidiospores with anastomosed pillars. Moreover, the generative hyphae in the hymenium have a distinctive tapering apex, very characteristic in the examined specimens and also in *G. applanatum*.

*Ganoderma subincrustatum* together with *G. oerstedii* and one species not yet identified were among the most common species in the urban tropical and subtropical zone in Mexico, especially as a parasite of many kinds of trees. *Ganoderma subincrustatum* also was found as parasite in tropical, subtropical and *Pinus-Quercus* forests. The specimens were labeled and deposited in herbaria as *G. lucidum*, *G. resinaceum*, *G. sessile* or as *Ganoderma* sp. This is the first record of *G. subincrustatum* for Mexico.

***Ganoderma weberianum* (Bres. et Henn. ex Sacc.) Steyaert, *Persoonia* 7(1): 79, 1972.**

FIG. 11

*Basidiomata* 4.5–8 × 7–14.5 cm, up to 2 cm thick, annual, sessile to substipitate, with a contracted base, single to imbricate, woody, context slightly wider than tubes. *Pileus* rounded-flabelliform to flabelliform, sometimes

conchate, convex to plane; surface glabrous, rivulose to slightly radially rugose, hard, glossy to dull, with a laccate crust, not cracking, difficult to penetrate with fingernail, easily lost leaving the surface relatively homogeneously dull, zonate; violet-brown (11F7) or darker to dark red in almost all the surface, except to the margin where gradually changes to orange (5A7), or fully violet-brown (11F7), with terra-cotta (7D7) basidiospores over the surface; margin pure white or yellowish, entire to slightly lobulate, thin, smooth. *Substipe* when present 3–5 × 1–3.5 cm, short and thick, thinner toward the base, horizontal, slightly darker than pileus, solid. *Context* 1.1–1.7 cm thick, fibrous, relatively homogeneous, zonate, lighter than orange-white (5A2), darkening to light brown (7D6) close to the tubes, changing to yellow when cut, with resinous incrustations throughout the context. *Pores* 2–3 per mm, angular to round, woody; pore surface yellowish-white (3A2) when fresh, darkening to ochraceous or yellowish-brown (6E8) when aging and drying; tubes 0.5–1 cm long, unstratified, concolorous with lower part of the context. *Hyphal system* dimitic. *Contextual trama* with generative hyphae not observed; skeletal hyphae 1.6–8 µm diam., thick-walled, generally solid, non-septate, non-branched to arboriform, moderately branched, golden-yellow. *Hymenophoral trama* as contextual trama. *Pileipellis* with cuticle cells 48–57.6 × 5–8.8 µm, cylindrical to narrow clavate, generally without protuberances nor branches, some with lateral short protuberances, thick-walled to almost solid, apex with granulations, golden-yellow, slightly amyloid in Melzer's reagent after 24 h; generative hyphae not observed; skeletal hyphae 4–6.4 µm diam., thick-walled to solid, non-septate, non-branched to arboriform with few branches, apex rounded and slightly wider, golden-yellow. *Basidiospores* 8.8–10.4 × 6.4–7.2 µm, Q = 1.29–1.5, ellipsoid, few broadly ellipsoid, apex truncate, with apical germ pore, yellowish-brown; perisporium wrinkled, hyaline; exosporium with inter-walled pillars up to 0.5 µm thick, subfree; endosporium wrinkled. *Chlamydospores* 10–11.2 µm, globose, thick-walled, with inter-walled pillars, yellowish-brown, scarce, negative in Melzer's reagent. *Basidia* not observed. *Cystidia* absent.

*Specimens examined.* **Jalisco**, *T. Cuevas s.n.* (IBUG), *L. Guzmán-Dávalos 9556, 9569* (IBUG), *M.G. Torres-Torres 690* (IBUG). R. Nava 309, 27 agosto 2004 (ENCB).

*Habitat.* Mainly gregarious; *Pinus-Quercus* forest; on wood.

*Distribution.* Recorded from Africa, Australia, Indonesia, Malaysia, New Guinea, Samoa Island, Singapore and Taiwan, and now from Mexico.

*Remarks.*—The remarkable features of *Ganoderma weberianum* are a pale context that changes to yellow when cut in fresh condition and with shiny and thick resinous incrustations, frequently with chlamydospores, and relatively narrow and very thick-walled to solid, almost cylindrical cuticle cells. Furthermore, the pileus is very hard as in subgenus *Elfvingia* and the laccate crust is difficult to indent with the fingernail. The examined specimens are in accordance with the descriptions of Steyaert (1972) and Corner (1983), except they mentioned the inter-pillars in the basidiospores barely visible. Steyaert (1972) described two forms of *Ganoderma weberianum*: one with thin and long cuticle cells (30 × 7–8 µm) and without or with few chlamydospores, and the other with thick and short cuticle cells (20 × 10–12 µm) and abundance of chlamydospores. Nevertheless, according to his pictures, the relationship wide/long indicates the cuticle cells are longer than he described. On the other hand, Corner (1983) described narrow and

long cuticle cells and abundant chlamydospores. The Mexican materials have narrow and long cuticle cells as in Corner specimens but scarce and smaller chlamydospores; Corner (1983) described them as  $12\text{--}19 \times 12\text{--}16 \mu\text{m}$ . Although *G. weberianum* had been reported from Asia (Steyaert 1972), it was not considered by Nuñez and Ryvarden (2000). Corner (1983) suggested the possibility of its occurrence in America. This is the first record of *G. weberianum* in America.

*Ganoderma zonatum* Murrill, *Bull. Torrey bot. Club* 29: 606, 1902. FIG. 12

*Basidiomata* 4–7 × 5–8 cm, up to 3 thick in the base, perenne, sessile to substipitate, sometimes effused-reflexed, single, woody but very light in weight, context narrower than tubes. *Pileus* dimidiate to semicircular, broadly attached, appanate; surface glabrous, slightly bumpy, shiny, with a laccate crust, cracked and removed when cut or aged, easy to penetrate with fingernail, slightly concentrically sulcate; reddish-brown (8F8, 9F8), becoming lighter than deep orange (7E7) toward periphery, generally homogeneous except in the margin, covered with cinnamon (6D6) basidiospores; margin yellowish-white, or as the pileus but lighter, entire, thick, obtuse, sulcate, with margins of previous season one over the other. *Context* up to 1.5 cm thick in the base, 0.9 cm average, corky-fibrous, almost homogeneous, zonated, henna-brown (7E8) to dark-brown (7F8), with a golden-yellow (5B7) to deep yellow (4A8) thin fringe below the laccate crust, without resinous bands. *Pores* 3–5 per mm, angular to rounded; pore surface yellowish-white (3A2), darkening to brown (6D8) when bruising; tubes up to 1.5 cm thick, indistinctly stratified, concolorous with the context. *Hyphal system* dimitic. *Contextual trama* with generative hyphae not observed; skeletal hyphae 2.4–8  $\mu\text{m}$  diam., solid to thick-walled, non-septate, arboriform, with few branches, yellowish to yellowish-brown. *Hymenophoral trama* with generative hyphae 2.4–3.2  $\mu\text{m}$  diam., thin-walled, with conspicuous clamps, hyaline, abundant; skeletal hyphae as in contextual trama. *Pileipellis* with cuticle cells 40–80 × 5.6–18.4  $\mu\text{m}$ , clavate, with two to three lateral protuberances and branches, generally without protuberances in the apex, thick-walled, at times multistratified, golden-yellow, with incrustations on all cell, content immediately black with Melzer's reagent, cells strongly amyloid after 12 h; generative hyphae 1.6–4.8  $\mu\text{m}$  diam., thin-walled, with conspicuous clamps, hyaline to yellowish, abundant, difficult to observe; skeletal hyphae 2.8–7.2  $\mu\text{m}$  diam., solid to thick-walled, arboriform, yellowish-brown. *Basidiospores* (11.2–) 12–14.4 × 5.6–7.6 (–8.4)  $\mu\text{m}$ , Q = (1.57–) 1.67–2.15, oblong, apex truncate, with apical germ pore, yellowish-brown, negative in Melzer's reagent; perisporium smooth, reddish-yellow; exosporium with inter-walled pillars less than 0.4  $\mu\text{m}$  thick, free pillars, endosporium semi-wrinkled. *Basidia* not observed. *Cystidia* absent.

*Specimens examined.* Jalisco, A. Cervantes 1 (IBUG); Nayarit, O. Vargas 13 (IBUG), E. Fanti 514 (IBUG).

*Other specimens examined.* USA, LECTOTYPE (NY).

*Habitat.* Single; planted forest of *Pinus* and deciduous tropical forests; on dead wood.

*Distribution.* Recorded from Africa, Argentina, Java, USA and now Mexico. This species has a wide tropical and subtropical distribution.

*Remarks.*—The species is easily recognized because in the edge of the pileus there are many imbricate margins and the basidioma although is not spongy, is very light in weight. Also, its oblong basidiospores are very characteristic. Murrill (1902) described basidiospores  $8-10 \times 4-6 \mu\text{m}$ , smaller than the ones observed in Mexican specimens, but subsequent authors studied the type and described them as  $10-15 \times 5-8 \mu\text{m}$  (Overholts 1953, Bazzalo and Wright 1982, Gottlieb and Wright 1999, Ryvar den 2000, 2004), which are in accordance with the studied specimen. Furthermore, we also examined the lectotype and observed basidiospores  $(11.2-12-13 (-14) \times 5.6-7.2 (-8) \mu\text{m}$ . In the Mexican materials the color of the pileus surface is more or less homogeneous and the surface remains shiny, while in the type the surface is dull and presents color zonations [reddish-brown (8F8, 9F8), deep orange (7E7) and golden-yellow (5B8)].

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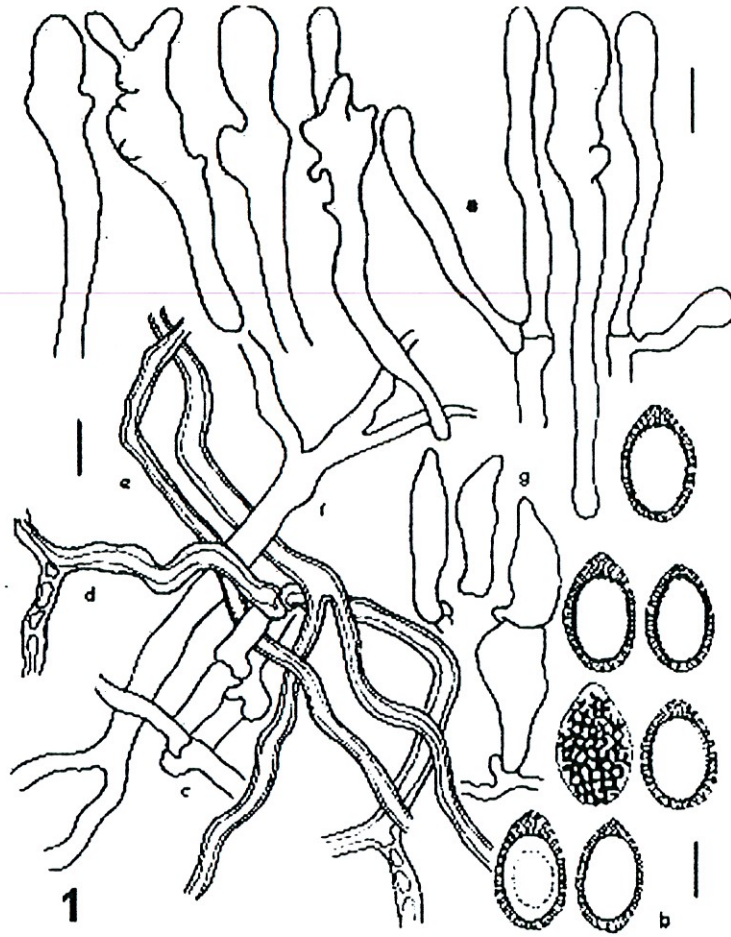


FIG. 1. Micromorphological features of *Ganoderma colossus*: a. Cuticle cells. b. Basidiospores. c–f. Hyphal system of crustohymenodermis: c. Generative hyphae. d. Sclerified generative hyphae. e. Skeletal hyphae. f. Binding hyphae. g. Cystidia. Bar = 8  $\mu$ m.

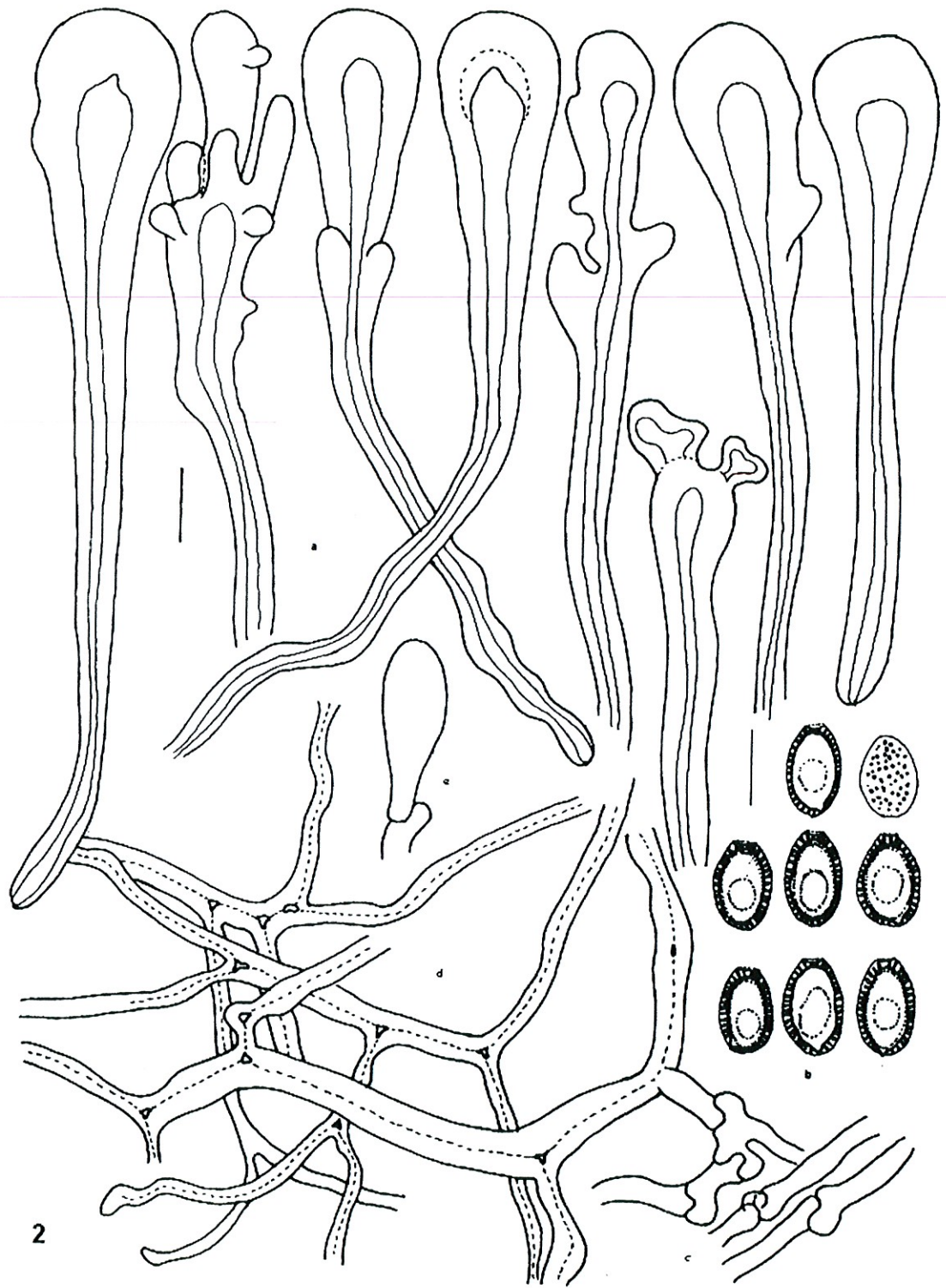


FIG. 2. Micromorphological features of *Ganoderma curtisii*: a. Cuticle cells. b. Basidiospores. c-d. Hyphal system of crustohymenodermis: c. Generative hyphae. d. Skeletal hyphae. e. Basidiolium. Bar = 8  $\mu$ m.



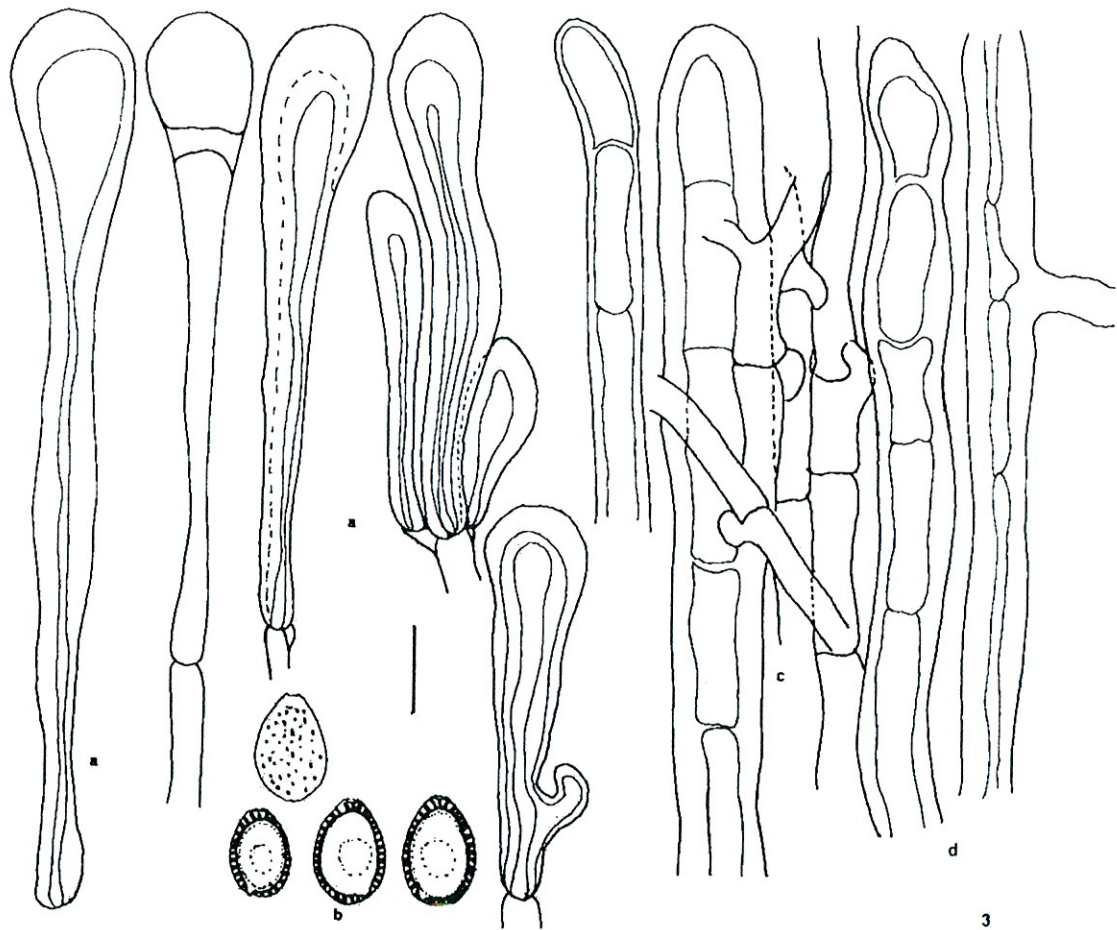


FIG. 3. Micromorphological features of *Ganoderma mexicanum*: a. Cuticle cells. b. Basidiospores. c–d. Hyphal system of crustohymenodermis: c. Generative hyphae. d. Skeletal hyphae. Bar = 8  $\mu$ m.

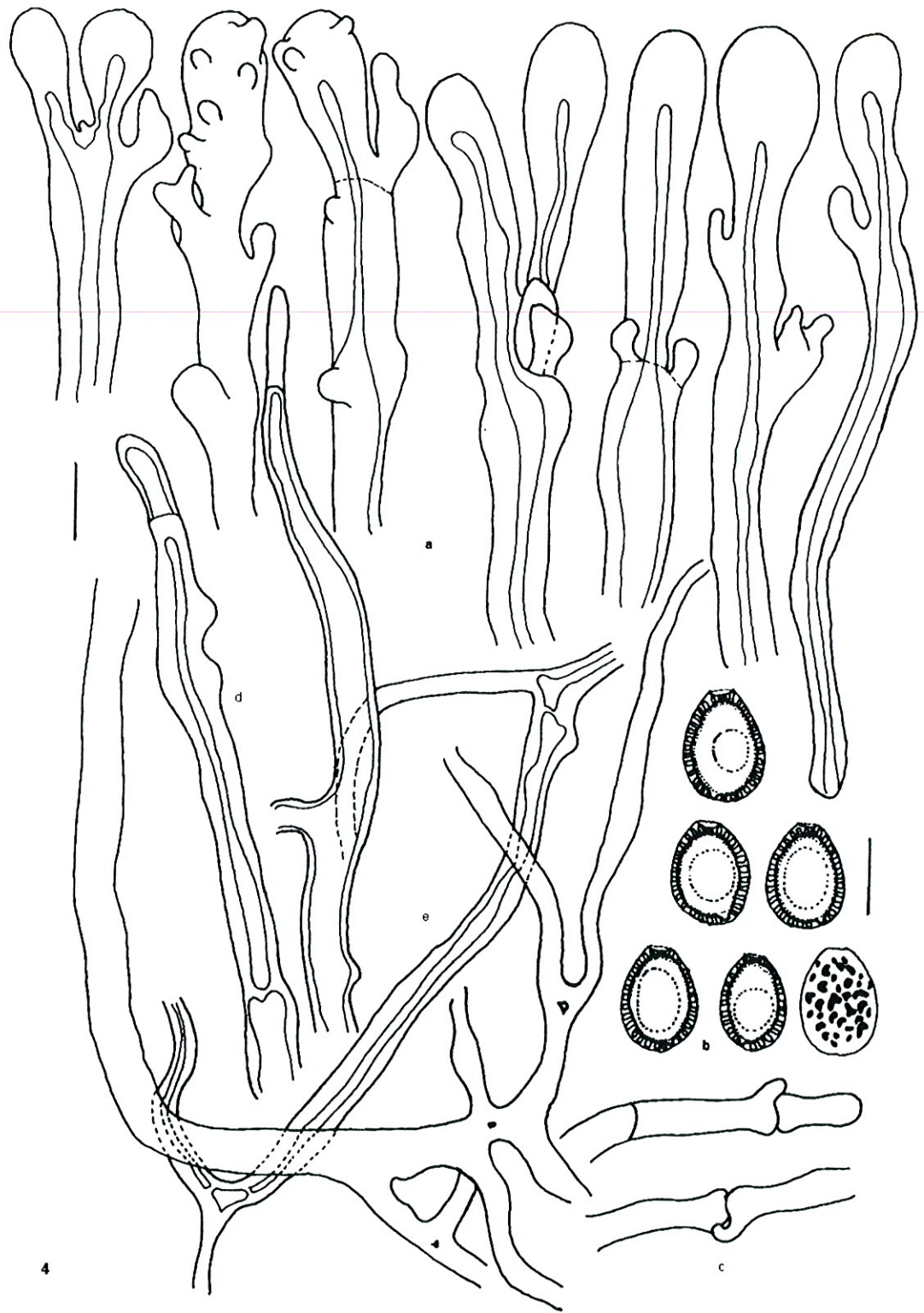


FIG. 4. Micromorphological features of *Ganoderma oerstedii*: a. Cuticle cells. b. Basidiospores. c–d. Hyphal system of crustohymenodermis: c. Generative hyphae. d. Skeletal hyphae. e. Binding hyphae. Bar = 8  $\mu$ m.

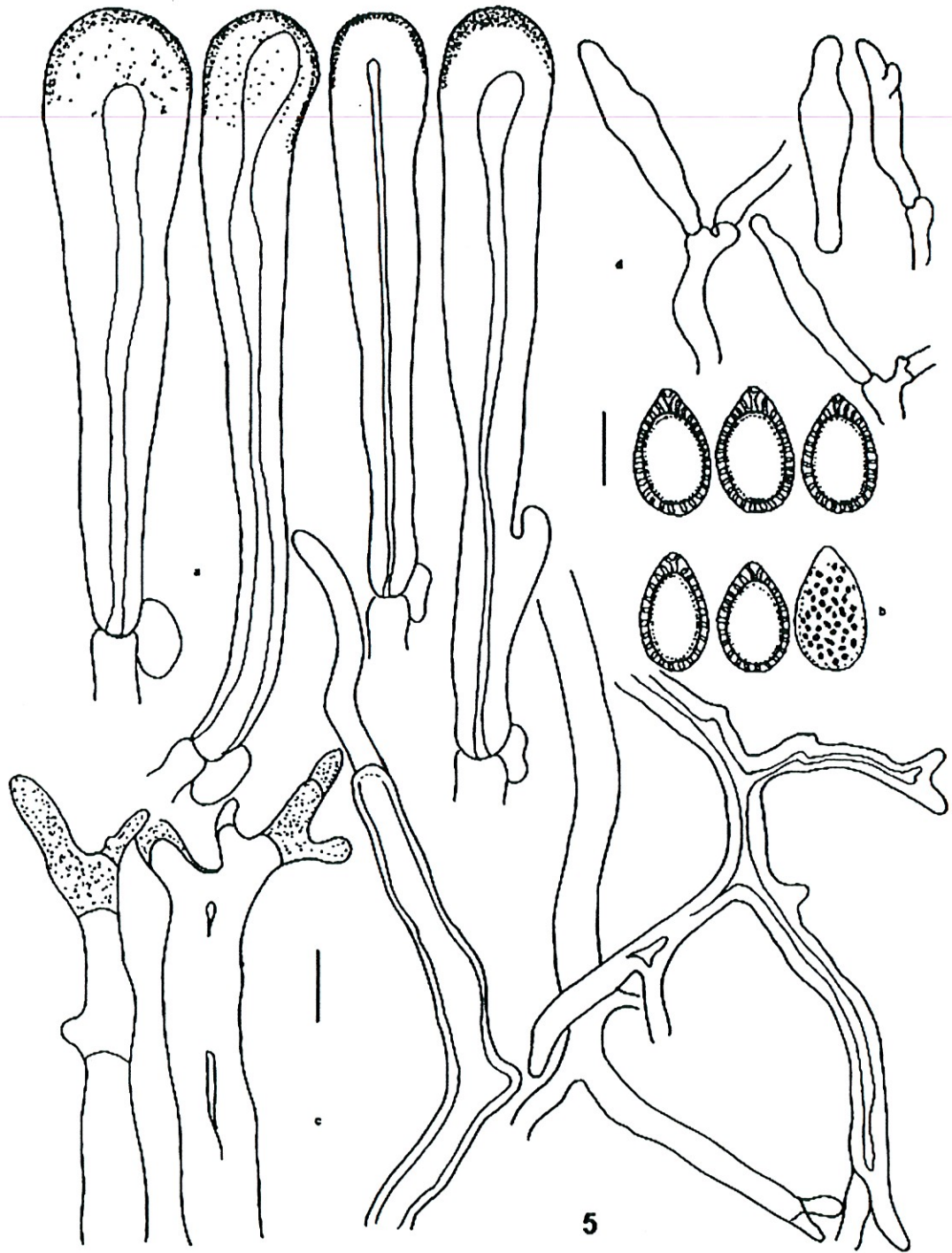


FIG. 5. Micromorphological features of *Ganoderma oregonense*: a. Cuticle cells. b. Basidiospores. c. Skeletal hyphae of crustohymenodermis. d. Cystidia. Bar = 8  $\mu$ m.

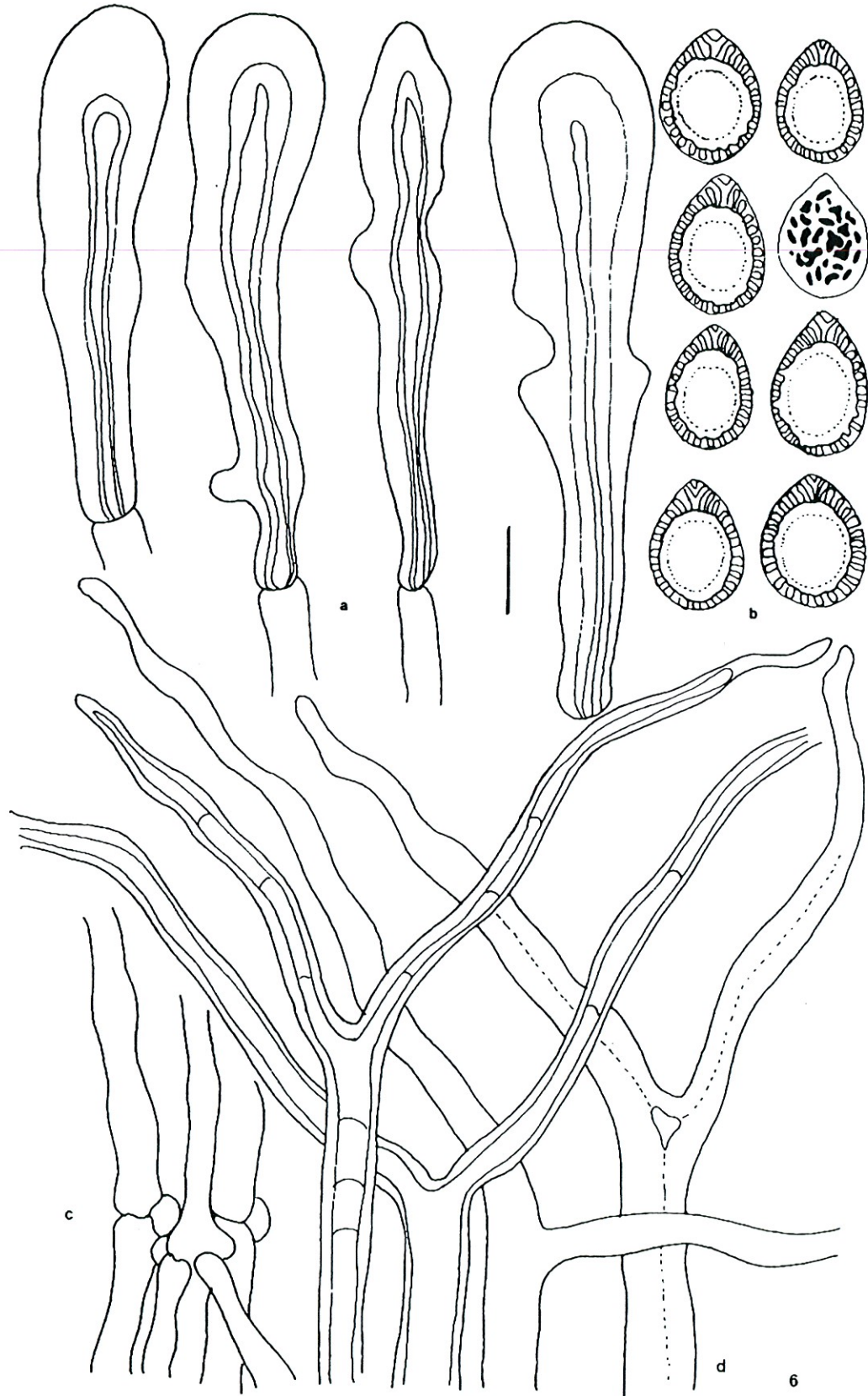


FIG. 6. Micromorphological features of *Ganoderma perturbatum*: a. Cuticle cells. b. Basidiospores. c–d. Hyphal system of crustohymenodermis: c. Generative hyphae. d. Skeletal hyphae. Bar = 8  $\mu$ m.

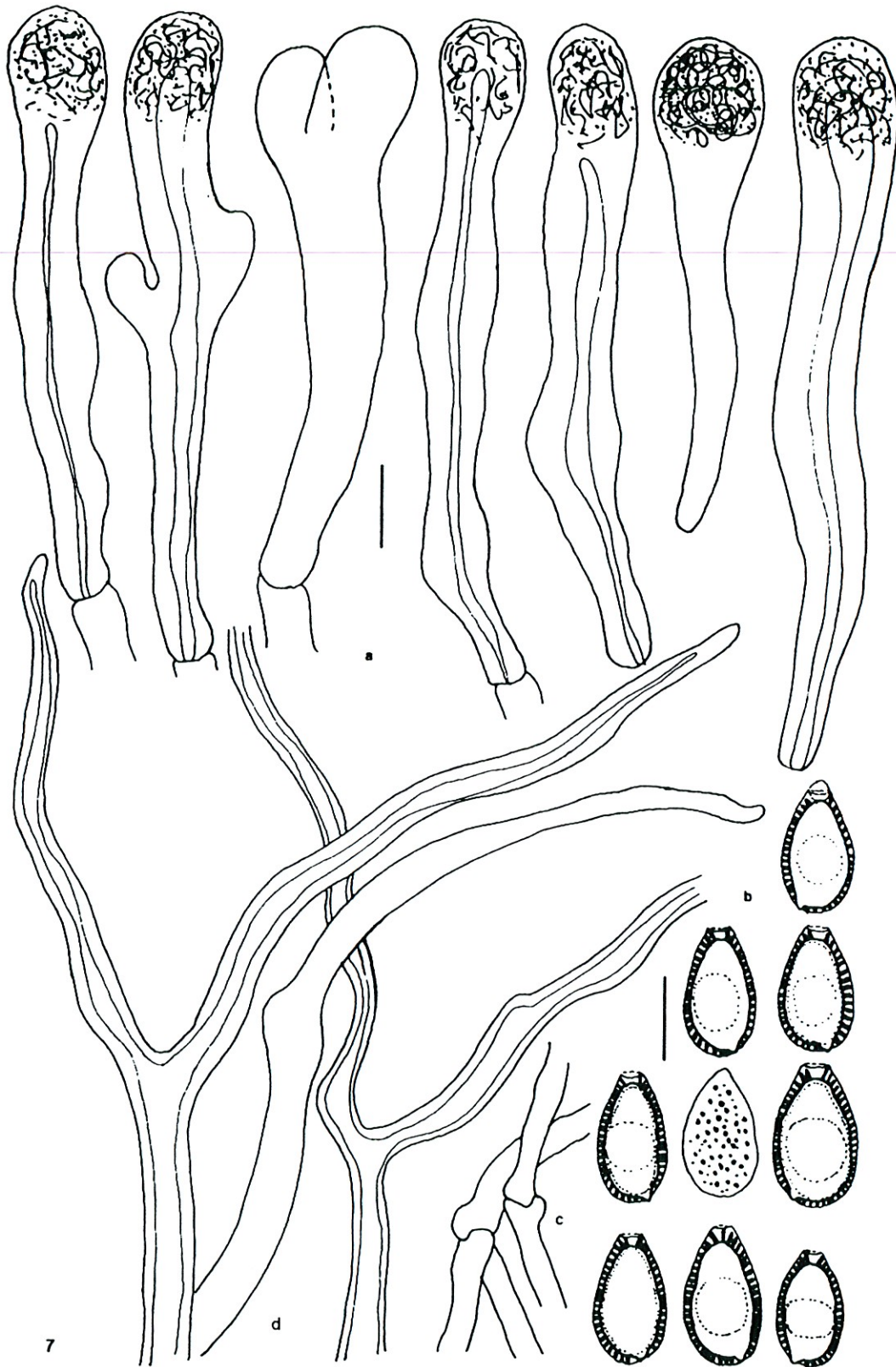


FIG. 7. Micromorphological features of *Ganoderma resinaceum*: a. Cuticle cells. b. Basidiospores. c–d. Hyphal system of crustohymenodermis: c. Generative hyphae. d. Skeletal hyphae. Bar = 8  $\mu$ m.

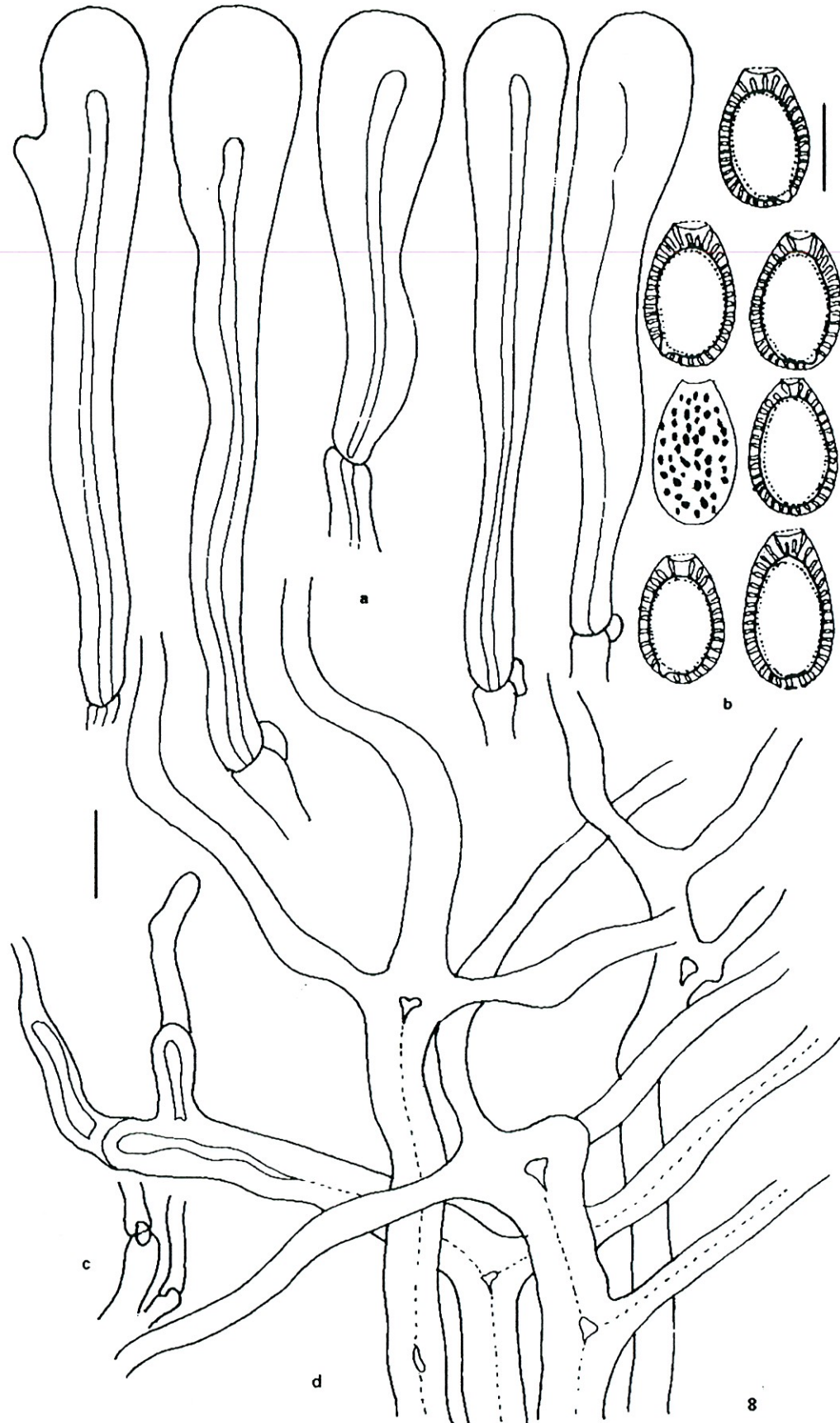


FIG. 8. Micromorphological features of *Ganoderma sessile*: a. Cuticle cells. b. Basidiospores. c–d. Hyphal system of crustohymenodermis: c. Generative hyphae d. Skeletal hyphae. Bar = 8  $\mu$ m.

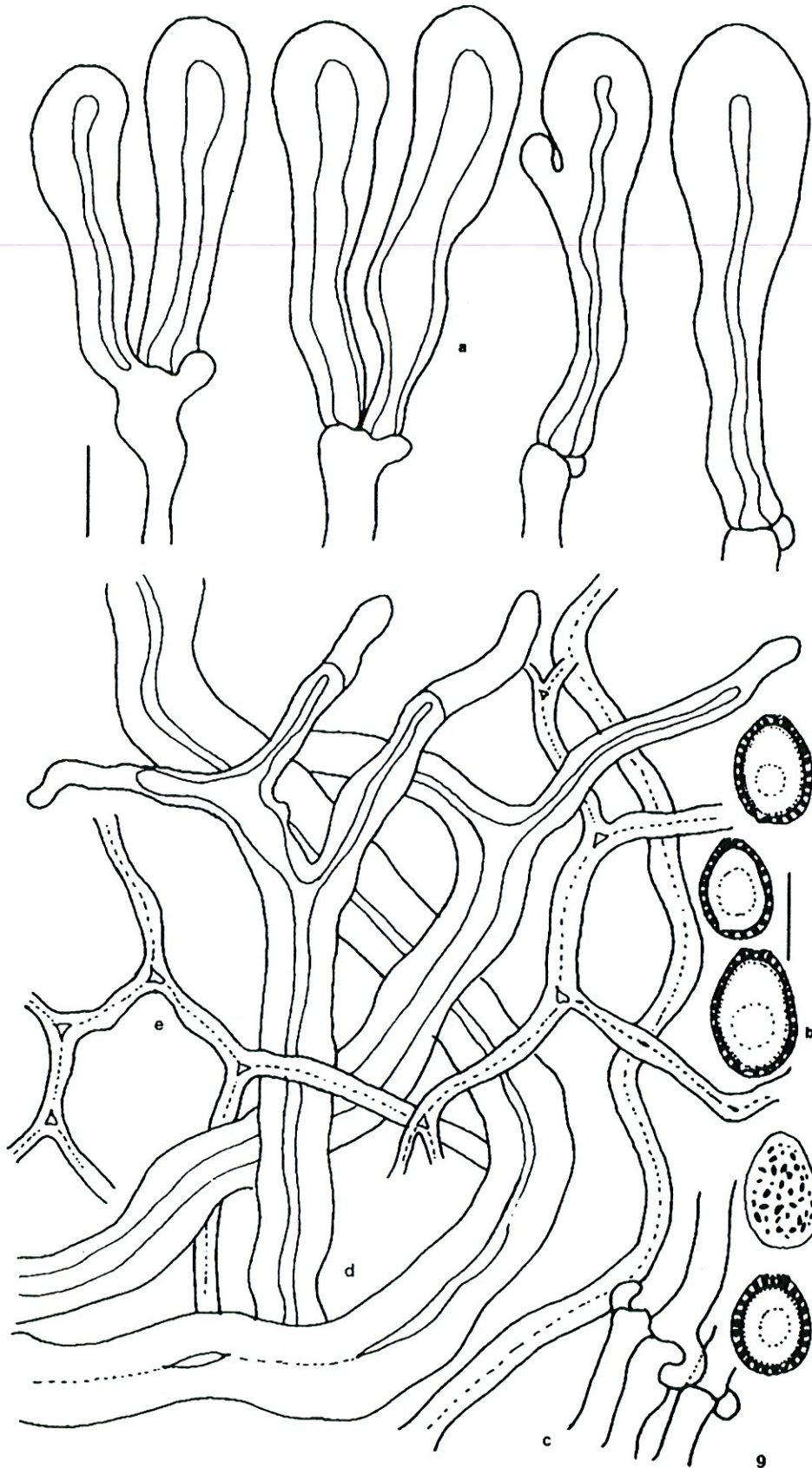
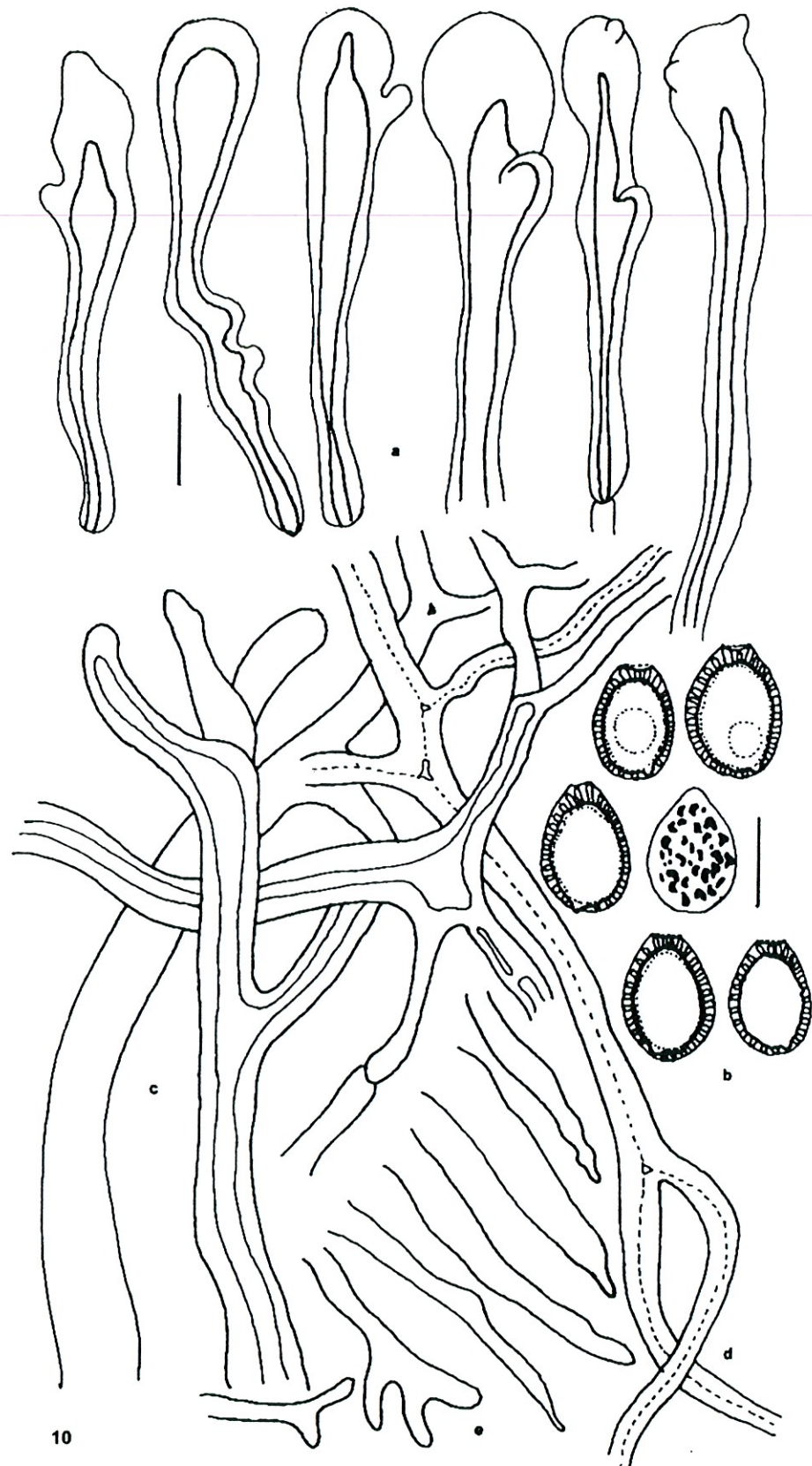


FIG. 9. Micromorphological features of *Ganoderma sessiliforme*. a. Cuticle cells. b. Basidiospores. c–d. Hyphal system of crustohymenodermis: c. Generative hyphae. d. Skeletal hyphae. e. Binding hyphae. Bar = 8  $\mu$ m.



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FIG. 10. Micromorphological features of *Ganoderma subincrustatum*: a. Cuticle cells. b. Basidiospores. c–d. Hyphal system of crustohymenodermis: c. Skeletal hyphae. d. Binding hyphae. e. Terminal hyphae of the hymenium. Bar = 8  $\mu$ m.



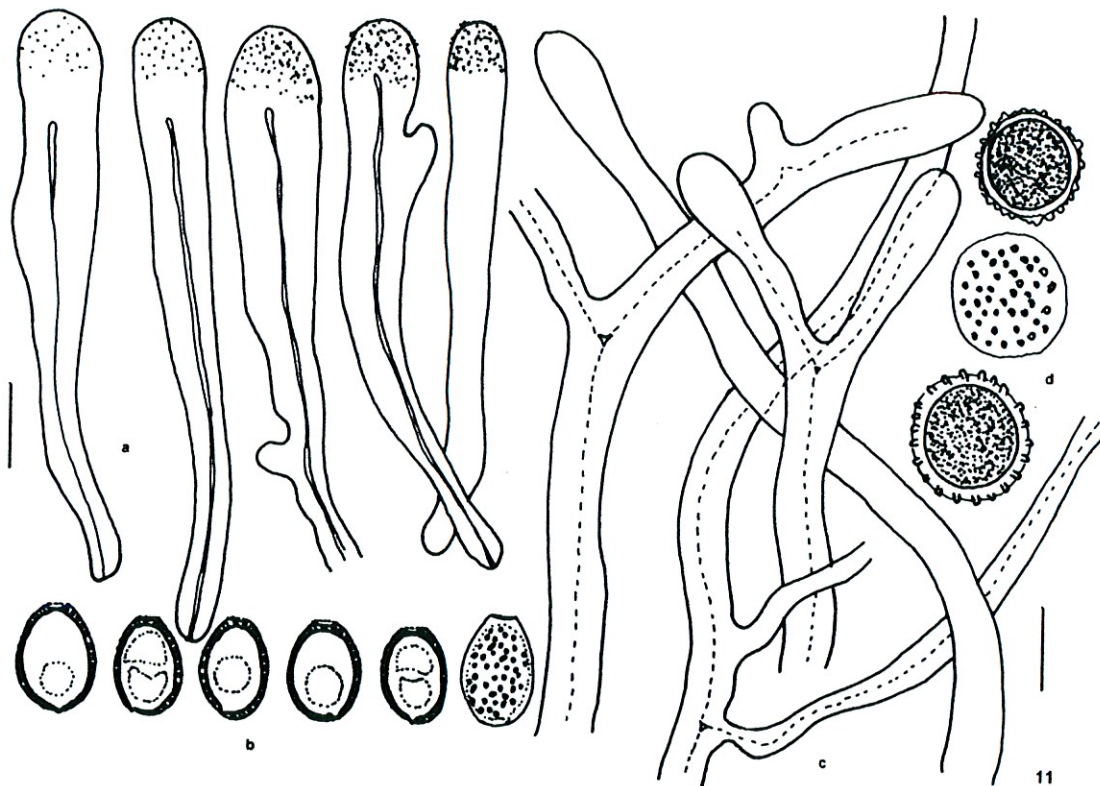


FIG. 11. Micromorphological features of *Ganoderma weberianum*. a. Cuticle cells. b. Basidiospores. c. Skeletal hyphae of crustohymenodermis. d. Chlamydospores. Bar = 8  $\mu$ m.

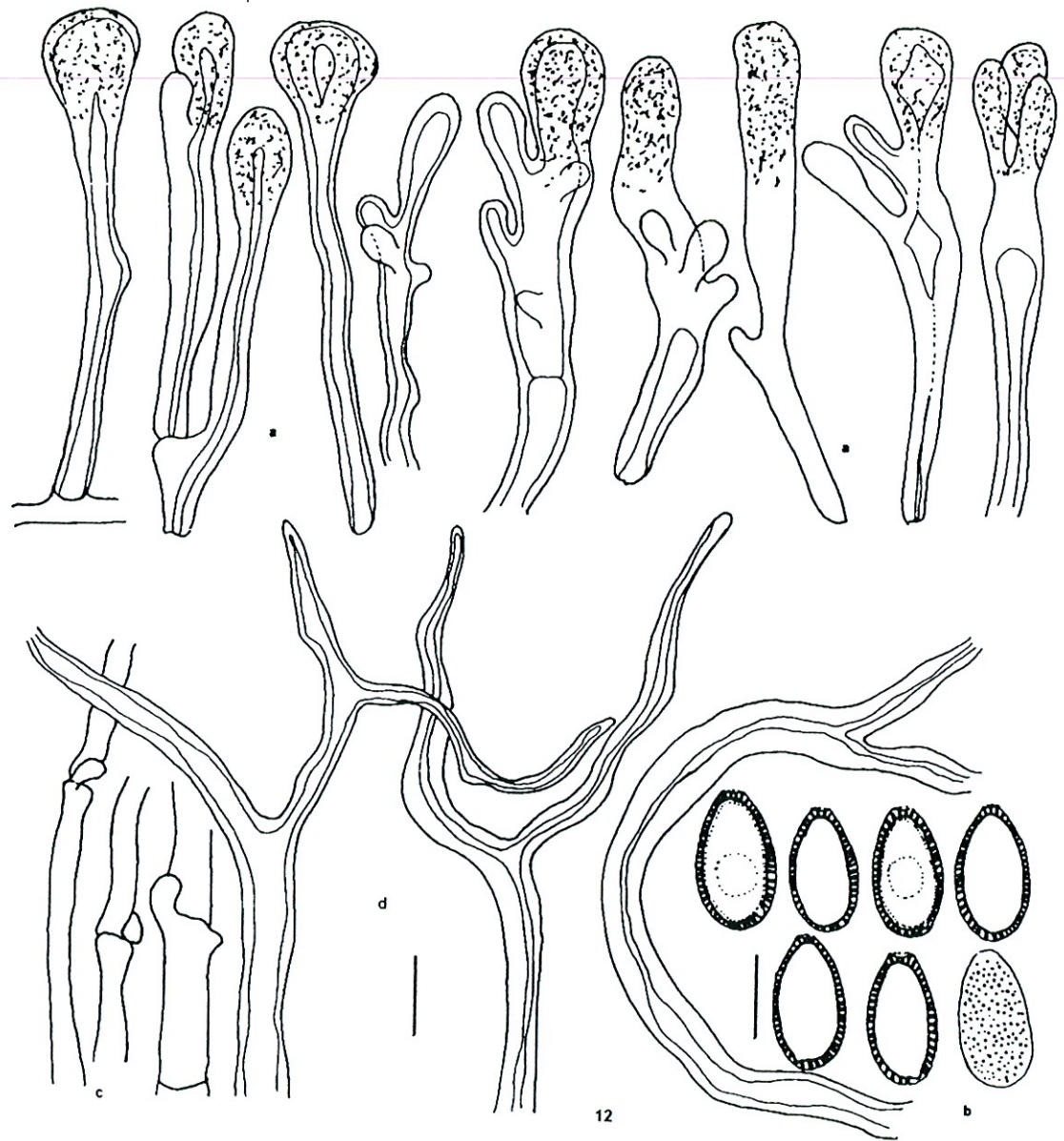


FIG. 12. Micromorphological features of *Ganoderma zonatum*: a. Cuticle cells. b. Basidiospores. c–d. Hyphal system of crustohymenodermis: c. Generative hyphae. d. Skeletal hyphae. Bar = 8  $\mu$ m.

## CAPÍTULO I, PARTE D

### Fungal Diversity

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#### Occurrence of *Ganoderma* on Brazil and new records of poorly known species

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Seventeen species of *Ganoderma* are described to Brazil; six of them are recorded for second or third time for the world and twelve are new records for Brazil.

**Key words:** Basidiospores, cuticle cells, *Ganodermataceae*.

#### Introduction

*Ganoderma* is a genus with mainly a tropical distribution and few species restricted to temperate regions. According with Ryvardeen (2004), *Ganoderma* is represented in the Neotropic by 20 species, many of them only known from the type locality. It is arguable the validity of some taxa because few specimens or only the type have been studied (Torres-Torres *et al.*, unpublished data). Brazil is a tropical country with a great mycological diversity; however, ten *Ganoderma* species have been recorded: *G. applanatum* (Pers.) Pat., *G. australe* (Fr.) Pat., *G. colossus* (Fr.) C.F. Baker, *G. curtisii* (Berk.) Murrill, *G. dorsale* (Lloyd) Torrend, *G. lucidum* (Curtis) P. Karst, *G. opacum* (Berk. & Mont.) Pat., *G. orbiforme* (Fr.) Ryvardeen, *G. parvulum* Murrill, *G. resinaceum* Boud. and *G. tornatum* (Pers.) Bres. (Rajchenber & de Meijer 1990, Loguercio Leite & Wright 1991, Lehmkuhl Gerber 1996, de Meijer 2001, Sotão *et al.* 2002, Gibertoni & Cavalcanti 2003, Góes-Neto *et al.*, 2003). Some of these species were mistakenly determined and the occurrence of others has not been recently confirmed. In many cases *G. applanatum* and *G. lucidum sensu lato* are the species commonly reported (Bononi *et al.* 1981, Sotão *et al.* 1991, Jesus 1993). On the other hand, some

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Brazilian thesis on polyporaceous fungi from specific areas did not record species of *Ganoderma*, confirming the poor knowledge of the genus. Furthermore, there are not specific works on *Ganoderma* from Brazil; the taxa recorded until now are only in lists, so they have not been described either illustrated.

Continuing with the work of Torres-Torres *et al.* (2005), the goal of the present study was to contribute to the knowledge about the geographic range of the species of *Ganoderma*, as well as consolidate the morphological features that permit the delimitation of the species. A review of the specimens of almost all the geographic areas in Brazil (Figure 1) deposited in SP and EMBRAPA was made; furthermore specimens and types deposited in other herbaria were checked. Here we describe 17 species from Brazil; six of them are recorded for second or third time for the world and twelve are new records for Brazil.

## Materials and methods

**Specimens studied.** The studied material was checked in EMBRAPA (personal collection of A.A.R. de Meijer) and SP herbaria. The type materials were requested to the herbaria BPI, FH, NY, O and UPS. Herbaria abbreviations follow Holmgren *et al.* (1990).

**Macro and micromorphological observations.** The key colours are from the colour reference of Kõrnerup & Wanscher (1963). Micromorphologic observations were made from material mounted in 10% KOH and Melzer's reagent. In the case of basidiospores the characters considered were: size, shape, apex, thickness and disposition of the pillars. The basidiospores shape was determined according to Q coefficient (length-width, Bas 1969) of 20 randomly selected but mature basidiospores. The shape, size, protuberances, branches and granulations of the cuticle cells were considered. The structures were measured through Axio Vision 4 software in a Zeiss Axioscop 40 microscope. The microscopic structure draws were made with a 100x oil-immersion objective, in a Zeiss K7 and a Zeiss Axioscop 40 microscopes.

## Results

### Dichotomy key

1. Pileus dull, generally grayish-brown to brown, cuticle hard ..... 2
1. Pileus glossy, degrading yellow, violet-brown to reddish-brown almost black, cuticle easy to penetrate with fingernail ..... 4
2. Cuticle very hard, generally more than 3 mm thick, basidiospores 8-13 x 6-7 µm..... 3
2. Cuticle hard, generally less than 3 mm thick, without resinous bands in the context, basidiospores 7-9 x 5-6 µm..... *G. applanatum*
3. Basidiospores with inter-walled pillars 0.3-0.4 µm thick, free..... *G. australe*
3. Basidiospores with inter-walled pillars 0.5-0.8 µm thick, anastomosed ..... *G. brownii*
4. Basidiomata stipitate, basidiospores with subacute apex..... 5
4. Basidiomata sessil to stipitate, basidiospores with truncate apex..... 7

5. Context relatively homogeneous, brown, cuticle cells without granulations..... *G. perturbatum*
5. Context duplex, light brown to brown, ..... 6
6. Cuticle cells with granulations..... *G. dorsale*
6. Cuticle cells without granulations..... *G. conccinum*
7. Context pale ..... 8
7. Context light brown to brown..... 11
8. Context duplex, basidiospores 11-14 x 7-9  $\mu\text{m}$ , with free pillars..... *G. sessile*
8. Context relatively homogeneous, basidiospores 8-10 x 6-7  $\mu\text{m}$ ..... 9
9. Context without resinous deposits, cuticle cells claviform, without granulations..... *G. sessiliforme*
9. Context with resinous deposits, cuticle cells cylindrical, with granulations..... 10
10. Pileus surface difficult to penetrate with fingernail, context changing to yellow when cut, basidiospores with subfree pillars..... *G. weberianum*
10. Pileus easy to penetrate with fingernail, context unchanging, cuticle cells with concentric elongate granulations in the apex, basidiospores with free pillars..... *G. perzonatum*
11. Context homogeneous, cuticle cells entire, almost cylindrical to cylindrical. basidiospores 11-13 x 6-7  $\mu\text{m}$ , with free pillars..... *G. resinaceum*
11. Context relatively homogeneous..... 12
12. Cuticle cells entire..... 13
12. Cuticle cells with protuberances or branches..... 14
13. Basidiospores 9-11 x 6-7  $\mu\text{m}$ , with free pillars..... *G. mexicanum*
13. Basidiospores 9-13 x 6-8  $\mu\text{m}$ , with anastomosed pillars..... *G. pulverulentum*
14. Cuticle cells with up to 14 lateral or apical protuberances, basidiospores 8-10 x 6-7  $\mu\text{m}$ , with free pillars..... *G. multiplicatum*
14. Cuticle cells with less protuberances, basidiospores 9-13 x 5-7  $\mu\text{m}$ ..... 15
15. Basidiomata stipitate, cuticle cells with up to four protuberances, without branches, basidiospores 10-13 x 5-7  $\mu\text{m}$ ..... *G. elegantum*
15. Basidiomata sessile to substipitate, cuticle cells different from above. basidiospores 9-12 x 7-8  $\mu\text{m}$ ..... 16
16. Cuticle cells commonly with a constriction, generally with up to five protuberances and one to two branches, apex with ferruginous granulations, basidiospores with free pillars..... *G. subfornicatum*
16. Cuticle cells irregular, with up to 10 lateral or apical protuberances or branches, without granulations in the apex, basidiospores with subfree pillars..... *G. orbiforme*

### Description of Brazilian species

A complete description is presented for the first, second or third records and only some micromorphologic data are provided for previously known species from Brazil.

*Ganoderma applanatum* (Pers.) Pat., Hyménomic. Eur. (Paris): 143 (1887).

**Basidiospores** 7-9.6 x 5.6-6.4  $\mu\text{m}$ , Q = 1.4-1.57, ellipsoid, apex truncate, yellowish-brown, negative in Melzer's reagent; perisporium wrinkled, hyaline; exosporium with inter-walled pillars 0.3-0.4  $\mu\text{m}$  thick, free.

*Specimen studied.* **BRAZIL**, Paraná, Curitiba, São José dos Pinhais, ADEA Reserva Biológica Cambuí, on dead palm trunk, 4 March 1980, A.A.R. de Meijer 386 (EMBRAPA); Antonina, Marumbí, Parque Marumbí, Rio do Nune, on dead dicotyledon trunk, 12 December 1987, A.A.R. de Meijer 962A (EMBRAPA).

*Remarks.* The species is characterized by its cuticle less than 3 mm thick, context without resinous deposits and small basidiospores. This species is very common with a wide distribution.

*Ganoderma australe* (Fr.) Pat., Bull. Soc. Myc. Fr. 5: 65 (1890).

≡ *Polyporus australis* Fr. Elench. Fung. 1: 108, 1828.

**Basidiospores** 8.4-12 x 6-7.2  $\mu\text{m}$ , Q = 1.4-1.57, ellipsoid, apex truncate, yellowish-brown, negative in Melzer's reagent; perisporium wrinkled, hyaline; exosporium with inter-walled pillars 0.3-0.4  $\mu\text{m}$  thick, free.

*Specimen studied.* **BRAZIL**, Paraná, Curitiba, São José dos Pinhais, ADEA Reserva Biológica Cambuí, 3 February 1979, A.A.R. de Meijer 23 (EMBRAPA); on stump, 15 July 1979, A.A.R. de Meijer 98 (EMBRAPA).

*Remarks.* The species is characterized by its thick and very hard cuticle, more than 3 mm thick; although young specimens can have a thinner but hard cuticle. Other diagnostic feature is the context with resinous deposits. *Ganoderma australe* is differentiated from *G. applanatum*, because the last one has smaller basidiospores and cuticle less than 3 mm thick. It is a cosmopolitan species; nevertheless, frequently underestimated because bad determinations.

*Ganoderma brownii* (Murrill) Gilb., Mycologia 53: 505 (1961).

≡ *Elfyngia brownii* Murrill, Western Polypores 5: 29, 1915.

**Basidiomata** 6.5-16 x 6.5-15 x 1.4-2 cm, perenne, sessile, single to imbricate, woody. **Pileus** round-flabelliform to circular, generally plane; surface glabrous, bumpy, dull, concentrically sulcate; with a crust 0.6-0.8 mm, not cracking, difficult to penetrate with fingernail; brown (7F7), with basidiospores over the surface; margin concolorous, entire, thin to thick, obtuse, sulcate. **Context** 0.3-0.9 cm thick, fibrous, azonate, homogeneous, reddish-brown (9F8); generally with resinous deposits close to the base of the pileus. **Pores** 4-5 per mm, angular to round, woody; pore surface yellow (3A2) to chrome-yellow (2A8); tubes 0.4-1.5 cm thick, unstratified to stratified, concolorous with the inferior part of the context. **Hyphal system** trimitic. **Pileipellis** an crustotricoderm; terminal elements 6.8-9.3  $\mu\text{m}$  wide, solid-walled, with apex 7-10  $\mu\text{m}$  wide, golden-yellow; generative hyphae not observed; skeletal hyphae 3.7-9.3  $\mu\text{m}$  diam., generally solid to thick-walled, non-septate, arboriform, yellowish-brown; binding hyphae not observed. **Basidiospores** 9.3-11.2 x 6.6-7.4  $\mu\text{m}$ , Q = 1.25-1.5, widely ellipsoid to ellipsoid, apex truncate, yellowish-brown, negative in Melzer's reagent;

perisporium wrinkled, hyaline; exosporium with inter-walled pillars 0.6-0.8  $\mu\text{m}$  thick, anastomosed. *Basidia* not seen. *Cystidia* absent.

*Specimens examined.* USA, California, Strawberry Canyon, on dead *Umbellularia*, November 1910, V.S. Brown 307 (NY, Lectotype). BRAZIL, São Paulo, Reserva Biológica de Paranapiacaba, 25 August 1987, M. Capelari 1723 (SP).

*Remarks.* The species is characterized by its pileus cuticle very hard and thick, more than 5  $\mu\text{m}$  thick, context reddish-brown, pore surface yellow, and big and widely ellipsoid to ellipsoid basidiospores. The Brazilian specimen agrees with the type, except that this has litter bigger basidiospores (9-12 x 6.5-8  $\mu\text{m}$ ). Gilbertson & Ryvarden (1986) suggested that the species was restricted to California. This is the first record of the species for Brazil and outside USA.

*Ganoderma conccinum* Ryvarden, Mycologia 92: 183 (2000).

Fig. 2

*Basidiomata* 1.4-5.5 x 2-10 x 0.5-1.5 cm, annual, stipitate, single, woody-corky, light in weight. *Pileus* round-flabelliform to reniform, convex to generally plane; surface glabrous, bumpy, semiglossy to glossy, concentrically sulcate; with a laccate crust, not cracking, difficult to remove, easy to penetrate with fingernail; garnet-red (11E8) to violet-brown (11F8) almost homogeneous in the adult, violet-brown (11F8) degrading to garnet-red (11E8) towards the periphery in the younger, with basidiospores over the surface; margin whitish to yellow in the young to concolorous in the adult, entire, acute in the young to obtuse and sulcate in the adult. *Stipe* 3.5-19 x 0.3-1.5 cm, lateral, cylindrical, solid, context duplex as basidiomata context; surface smooth to tuberculate, generally very shiny, red-wine almost black, generally darker than pileus. *Context* 0.2-0.6 cm thick, up to 1.2 cm in the base of the pileus, fibrous, azonate, duplex, pale-orange to light-orange (5A3, 5A4) above, light brown (6D7) toward the tubes; generally with resinous bands very thin and inconspicuous, only one specimen with thick lines. *Pores* 6-8 per mm, angular to round, woody; pore surface yellow (3A2); tubes 0.5-0.7 cm thick, unstratified to stratified, concolorous with the inferior part of the context. *Hyphal system* dimitic. *Contextual trama* with generative hyphae not observed; skeletal hyphae 6.2-6.8  $\mu\text{m}$  diam., generally solid to thick-walled, septate in the apex, arboriform, yellowish. *Hymenophoral trama* as contextual trama. *Pileipellis* with cuticle cells 35.2-46.5 x 6.5-10  $\mu\text{m}$ , claviform, entire or rarely with one lateral protuberance; thick-walled, golden-brown, without granulations in the apex; dextrinoid with Melzer's reagent; generative hyphae not observed; skeletal hyphae 6.2-6.8  $\mu\text{m}$  diam., generally solid to thick-walled, non-septate, arboriform, yellowish. *Basidiospores* (11-) 11.8-14 x 8-9 (-10)  $\mu\text{m}$ , Q = 1.4-1.65, ellipsoid, apex subacute, with apical germ pore difficult to observe, yellowish-brown, negative in Melzer's reagent; perisporium wrinkled, hyaline; exosporium with inter-walled pillars 0.6-0.8  $\mu\text{m}$  thick, anastomosed. *Basidia* not seen. *Cystidia* absent.

*Specimens examined.* COLOMBIA, Chocó, Municipality of Riosucio, Sautatá, Parque Nacional Katios, 28-30 June 1978, Ryvarden 18640 (O, Holotype). BRAZIL, Paraná State, Curitiba, Centro Politécnico, Jardim das Americas, on buried roots of living dicotyledon, 18 January 1989, A.A.R. de Meijer 1194 (EMBRAPA); Parque Boriquí, on soil, 5 March 1993, A.A.R. de

Meijer 2533 (EMBRAPA); RBC, on forest soil, 24 November 1994, A.A.R. de Meijer 2961 (EMBRAPA); Reserva Biológica Cambuí, on buried dicotyledon, 20 May 1995, A.A.R. de Meijer 2278 (EMBRAPA); Jundiá do Sul, Fazenda Monte Verde, on decayed dicotyledon, on buried wood, 4 February 1996, A.A.A.R. de Meijer 3253 (O, EMBRAPA); Rio Grande do Sul, Pelotas, Horto Forestal, buried, 9 June 1959, E.C. Santos 29 (SP).

*Remarks.* The species can be distinguished by its stipitate basidiomata, duplex context, cuticle cells without granulations and basidiospores with subacute apex. Ryvarden (2000) suggested that the long stipe was an important feature; nevertheless, we found specimens with very short stipe. Ryvarden (2000) described basidiospores 12-14 x 7-8  $\mu\text{m}$ , but we found basidiospores wider (8-10  $\mu\text{m}$ ) in the type deposited in O. The species is close related to *Ganoderma dorsale*, mainly distinguish for the presence of granulations in the apex of cuticle cells in the last one. *Ganoderma conccinum* was recorded by Ryvarden & de Meijer (2002) from Brazil, so this is the second record for the species for the country.

*Ganoderma dorsale* (Lloyd) Torrend, Brotéria Bot. 18: 32 (1920). Fig. 3  
≡ *Polyporus dorsalis* Lloyd, Mycol. Writ. 5: 658, 1915.

**Basidiomata** 2-4 x 3-3.5 x 0.7-1.1 cm, annual, stipitate, single, woody-corky, light in weight. **Pileus** round-flabelliform to reniform, convex to generally plane; surface glabrous, bumpy, semiglossy to glossy, concentrically sulcate; with a laccate crust, not cracking, difficult to remove, easy to penetrate with fingernail; garnet-red (11E8) degrading to violet-brown (11F8), almost homogeneous in the adult, without basidiospores over the surface; margin yellowish to concolorous, entire, acute to obtuse, smooth to sulcate. **Stipe** 8.7-10 x 0.7-1 cm, lateral, cylindrical, solid, context duplex as basidiomata context; surface smooth to tuberculate, generally very shiny, red-wine almost black, generally darker than pileus. **Context** 0.2-0.5 cm thick, up to 0.7 cm in the base of the pileus, fibrous, azonate, duplex, caramel (6C6) above, lighter than dark brown (6F8) toward the tubes; without resinous bands. **Pores** 4-5 per mm, angular to round, woody; pore surface yellow (3A2); tubes 0.1-0.7 cm thick, unstratified, concolorous with the inferior part of the context. **Hyphal system** trimitic. **Contextual trama** with generative hyphae not observed; skeletal hyphae 3.1-9.3  $\mu\text{m}$  diam., solid to generally thick-walled, septate in the apex, arboriform, yellow to yellowish-brown; binding hyphae 0.8-4.2  $\mu\text{m}$  diam., solid, non-septate, yellowish, scarce. **Hymenophoral trama** as contextual trama. **Pileipellis** with cuticle cells 46-72 x 7.2-13.6  $\mu\text{m}$ , claviform, entire or with one lateral protuberance; thick-walled, golden-brown, with granulations in the apex; dextrinoid with Melzer's reagent; generative hyphae not observed; skeletal hyphae 4.4-7.4  $\mu\text{m}$  diam., generally solid to thick-walled, non-septate, arboriform, yellowish. **Basidiospores** 11.8-13 x 8-9.3  $\mu\text{m}$ , Q = 1.33-1.57, ellipsoid, apex subacute, with apical germ pore difficult to observe, yellowish-brown, negative in Melzer's reagent; perisporium wrinkled, hyaline; exosporium with inter-walled pillars 0.6-0.8  $\mu\text{m}$  thick, anastomosed. **Basidia** not observed. **Cystidia** absent.

*Specimens examined.* BRAZIL, Rio Grande do Sul, on buried wood, s. date, R.J. Rick s.n. (BPI, Lectotype); Pelotas, Horto Botânico, Instituto Agronômico do Sul, 16 June 1959, E.C. Santos 76 (SP); São Paulo, Iepe, about 5



km of Porto Alvarado, along of the Rio Paranapanema, Fazenda CAPI, 9 February 1965, G. Eiten, L.T. Eiten & H. Kimura 6009 (SP).

*Remarks.* The species is characterized by its stipitate basidioma, cuticle cells with granulations in the apex and basidiospores with subacute apex. The species did not have a modern description and was not included in the last treatment on Neotropical Polypores (Ryvarden 2004). Nevertheless, Moncalvo & Ryvarden (1997) wrote down that this species is common in Brazil, and also they recorded it from Florida, Honduras, Java, Nicaragua, Malaysia, Philippines and Singapore. Besides this, there are not records of the species in the last 80 years. *Ganoderma dorsale* was considered a synonym of *G. perturbatum* by Steyaert (1967) and of *G. lucidum s.l.* by Ryvarden (1990). The species is close related with *G. conccinum* and *G. perturbatum*; the tree species present stipe, basidiospores with subacute apex and anastomosed pillars. This is the second record for the species from Brazil.

*Ganoderma elegantum* Ryvarden, Synopsis Fungorum 19: 81-82 (2004). Fig. 4

**Basidiomata** 1.7 x 2 x 1 cm, annual, stipitate, single, woody-corky. **Pileus** round-flabelliform, plane; surface glabrous, glossy, concentrically sulcate; with a laccate crust, not cracking, easy to penetrate with fingernail; reddish-brown (11F8) homogeneous, without basidiospores over the surface; margin whitish, entire, acute to obtuse, smooth. **Stipe** 8.7 x 1.3 cm, lateral, cylindrical, solid; surface smooth, very shiny, red-wine almost black, generally darker than pileus. **Context** 0.9 cm thick, spongy-fibrous, azonate, relatively homogeneous, yellowish-brown (5E8) degrading to eyes-brown (7F7); with resinous bands that do not extend up to the periphery. **Pores** 6-7 per mm, angular to round, woody; pore surface yellowish (2A3); tubes 0.1 cm thick, unstratified, concolorous with the inferior part of the context. **Hyphal system** dimitic. **Contextual trama** with generative hyphae not observed; skeletal hyphae 2.5-8.5 µm diam., solid to generally thick-walled, non-septate, arboriform, yellow to yellowish-brown. **Hymenophoral trama** with generative hyphae not observed; skeletal hyphae 2.5-6.2 µm diam., solid to generally thick-walled, non-septate, arboriform, yellow to yellowish-brown. **Pileipellis** with cuticle cells 29.8-52.7 x 6.2-10.5 µm, claviform, entire or with up to four lateral or apical protuberances; subthick-walled, golden-yellow, without granulations in the apex; negative to apex slightly amyloid with Melzer's reagent; generative hyphae not observed; skeletal hyphae 5.6-8.7 µm diam., subthick-walled to solid, non-septate, arboriform, yellowish-brown. **Basidiospores** 10-12 x 5-7 µm, Q = 1.4-1.65, oblong to ellipsoid, apex subacute, with apical germ pore difficult to observe, yellowish-brown, negative in Melzer's reagent; perisporium wrinkled, hyaline; exosporium with inter-walled pillars 0.6-0.8 µm thick, free; endosporium wrinkled. **Basidia** not observed. **Cystidia** absent.

*Specimen examined.* **BRAZIL**, Rondônia, Jarú, near to the airport, on wood, 10 October 1986, M. Capelari & R. Maziera 962 (SP).

*Remarks.* *Ganoderma elegantum* was recently described from Ecuador (Ryvarden 2004). The specimen examined had many basidiomata immature and few with basidiospores, but in general the features agree with Ryvarden (2004); although he described cuticle cells without protuberances and apically widened. This is the second record of the species for the world.

**Basidiomata** 6.2-11.2 x 9-19 x 1.6-2.5 cm, annual, sessile, single, woody, but light in weight when dry. **Pileus** round-flabelliform to semicircular, plane to convex; surface glabrous, rugose, semiglossy to dull, with a laccate crust, thin, not easily cracked or removed, but easy to penetrate with fingernail, concentrically sulcate; reddish-black in a 90% to brown-violet (11F5) in the periphery, almost homogeneous; margin henna (7E8) to concolorous, slightly lobulate, thin to thick, obtuse, smooth. **Context** 0.4-0.8 cm thick, fibrous, azonate, relatively homogeneous, pale-orange to apricot (5A3, 5B6) above, cognac (6E7) toward the tubes; with resinous bands until half of basidiomata. **Pores** 3-5 per mm, round, woody; pore surface pale yellow (2A3); tubes 0.8-1.1 cm thick, unstratified, concolorous with the inferior part of the context. **Hyphal system** dimitic. **Contextual trama** with generative hyphae 1.8-3.8  $\mu\text{m}$  diam., thin-walled, hyaline, non-branched; skeletal hyphae 1.9-4.4  $\mu\text{m}$  diam., generally solid, non-septate, arboriform, yellowish. **Hymenophoral trama** as contextual trama. **Pileipellis** with cuticle cells 35.2-72.4 x 6.8-10.5  $\mu\text{m}$ , claviform but apically widened, entire or rarely with one lateral protuberance; very thick-walled, golden-brown to yellowish-brown, without granulations in the apex; dextrinoid with Melzer's reagent; generative hyphae 3.8-5  $\mu\text{m}$  diam., thin-walled, hyaline, abundant; skeletal hyphae 5-9.3  $\mu\text{m}$  diam., thick-walled to generally solid, non-septate, arboriform, yellowish-brown. **Basidiospores** 9.3-10.6 x 6.2-7.4  $\mu\text{m}$ , Q = 1.42-1.54, ellipsoid, apex truncate, yellowish-brown, negative in Melzer's reagent; perisporium wrinkled, hyaline; exosporium with inter-walled pillars 0.4  $\mu\text{m}$  thick, subfree. **Basidia** not seen. **Cystidia** absent.

*Specimens examined.* **MEXICO**, Estado de México, D. de Jonacatepec, Tepalcingo, 22 October 1890, s. coll (FH, Lectotype). **BRAZIL**, São Paulo, Tremembé, Horto Forestal de Cantareira, on trunk of *Araucaria angustifolia*, 3 March 1953, C.D.F. Pacheco s.n. (SP).

*Remarks.* The species is characterized by its relatively homogenous context, cuticle cells apically widened and small basidiospores with a subfree pillars. Also, it is notably that the skeletal hyphae are little branched. The studied specimen agrees with the type of *Ganoderma mexicanum*, but the type apparently has a homogenous context without resinous bands; a possible explanation is that the type is in very bad state and a big part of the context was destroyed. The species was only known from the type locality. This is the second record for the world.

*Ganoderma multiplicatum* (Mont.) Pat., Bull. Soc. Myc. Fr. 5: 74 (1889). Fig. 6  
= *Polyporus multiplicatus* Mont., *Annls Sci. Nat., Bot., sér. 4, 1: 128. 1854.*

**Basidiomata** 5-7 x 4-12 x 1-3 cm, perenne, sessile to substipitate, single, woody. **Pileus** round-flabelliform with contracted base, plane to slightly convex; surface glabrous, smooth to bumpy, glossy, with a laccate crust, thin, not easily cracked or removed, but easy to penetrate with fingernail, concentrically sulcate; very dark reddish-black to dark violet-brown (11F5) in a 80-90%, degrading to rust-brown (6E8) to dark orange (5B8); margin whitish, entire to slightly lobulate, obtuse, smooth. **Context** 0.6-0.9 cm thick, fibrous, zonate, relatively homogeneous, light yellowish-brown changing to light brown (6D7) toward the

tubes; with resinous bands. *Pores* 4-6 per mm, angular to round, woody; pore surface pale yellow (2A3); tubes 0.4-0.8 cm thick, unstratified, concolorous with the inferior part of the context. *Hyphal system* dimitic. *Contextual trama* with generative hyphae not observed; skeletal hyphae 3.8-7.5  $\mu\text{m}$  diam., subthick-walled to generally solid, non-septate, arboriform, yellow. *Hymenophoral trama* with generative hyphae not observed; skeletal hyphae 2.2-3.8  $\mu\text{m}$  diam., subthick-walled to generally solid, non-septate, arboriform, yellow. *Pileipellis* with cuticle cells 38.5-62 x 5.6-10  $\mu\text{m}$ , irregular, up to 14 lateral or apical protuberances; very thick-walled, golden-yellow, without granulations in the apex; amyloid with Melzer's reagent; generative hyphae 2-3.8  $\mu\text{m}$  diam., thin-walled, hyaline, abundant; skeletal hyphae 1.9-3.2  $\mu\text{m}$  diam., solid-walled, non-septate, arboriform, yellowish-brown. *Basidiospores* 8-9.9 x 6.2-6.8  $\mu\text{m}$ , Q = 1.3-1.47, ellipsoid, apex truncate, yellowish-brown, negative in Melzer's reagent; perisporium wrinkled, hyaline; exosporium with inter-walled pillars 0.4-0.5  $\mu\text{m}$  thick, free. *Basidia* not seen. *Cystidia* absent.

*Specimens examined.* BRAZIL, Alagoas, Maceió, Reserva da Ibama, 28 August 1992, R. Maziero s.n. (SP); Rio de Janeiro, Mata de Tijuca, 4 May 1966, J.S. Furtado s.n. (SP); Rondônia, Jarú, right margin of Rio Jarú, 3 October 1986, M. Capellari & R. Maziero 693 (SP); São Paulo, Parque do Estado, Instituto de Botânica, on wood, 23 January 1970, B. Skvortzov s.n. (SP); Sergipes, Estação Ecológica Santa Isabel, 9 December 2003, R.H. Marino s.n. (SP).

*Remarks.* The species is characterized by its basidiomata reddish-black, context relatively homogenous, small basidiospores and cuticle cells with many protuberances. The specimens agree with Gottlieb & Wright (1999) and Ryvar den (2000) descriptions, except that the last author described cuticle cells entire or with few small apical protuberances. The species was described from Guyana, and it has been recorded from Argentina (Gottlieb & Wright 1999) and Brazil (Ryvar den 2000). According with Ryvar den (2000), the species has a Pantropical distribution; nevertheless, there are not records of the species from Brazil. On the other hand, some mistakenly identified specimens as *Ganoderma lucidum* and other species were found in SP and EMBRAPA. Here we record the species for the first time for some localities.

*Ganoderma orbiforme* (Fr.) Ryvar den, Mycologia 92: 187 (2000). Fig. 7  
 = *Polyporus orbiformis* Fr., Epicr. Syst. Mycol. (Upsaliae): 463, 1838.

*Basidiomata* 4-6.5 x 5-8 x 1.5-2 cm, perenne, sessile, single to imbricate, woody. *Pileus* round-flabelliform, plane to convex; surface glabrous, bumpy, glossy, with a laccate crust, thin, not easily cracked or removed, but easy to penetrate with fingernail, concentrically sulcate; very dark reddish-black in a 80-90% degrading to light-brown (6D8) toward the periphery; margin light-brown (6D8), entire, obtuse, generally sulcate. *Context* 0.8-1 cm thick, fibrous, zonate, relatively homogeneous, light golden-brown (5D) degrading to rust-brown (6E8) toward the tubes; with discontinuous resinous bands. *Pores* 4-8 per mm, round, woody; pore surface yellow (3A6); tubes 0.7-0.9 cm thick, unstratified to stratified, concolorous with the inferior part of the context. *Hyphal system* trimitic. *Contextual trama* with generative hyphae not observed; skeletal hyphae 2.5-8.8  $\mu\text{m}$  diam., generally solid, non-septate, arboriform, yellow to yellowish-brown. *Hymenophoral trama* with generative hyphae 2.5-3.1  $\mu\text{m}$  diam., thin-

walled, hyaline, branched, scarce; skeletal hyphae 2.5-4.4  $\mu\text{m}$  diam., generally solid, non-septate, arboriform, yellow; binding hyphae 1.9-2.5  $\mu\text{m}$ , thick-walled, hyaline to yellowish, scarce. *Pileipellis* with cuticle cells 33.5-62 x 4.4-9.3  $\mu\text{m}$ , irregular, up to 10 lateral or apical protuberances or branches; thick-walled, golden-yellow to golden-brown, without granulations in the apex; strongly amyloid with Melzer's reagent; generative 2.5-5  $\mu\text{m}$  diam., thin-walled, hyaline, branched, abundant; skeletal hyphae 3.8-8  $\mu\text{m}$  diam., solid-walled, non-septate, arboriform, golden-yellow to yellowish-brown. *Basidiospores* 9-11.2 (-13) x 6.9-8 (-8.6)  $\mu\text{m}$ , Q = 1.38-1.5, ellipsoid, apex truncate, yellowish-brown, negative in Melzer's reagent; perisporium wrinkled, hyaline; exosporium with inter-walled pillars 0.4-0.5  $\mu\text{m}$  thick, subfree. *Basidia* not seen. *Cystidia* absent.

*Specimens examined.* **GUINEA**, s. data, s. coll., s. date, A. Afzelius s.n. (UPS, Holotype). **BRAZIL**, Rio de Janeiro, Angra de Reis, Ilha da Gilpoia, 4 March 1956, O. Fidalgo F-365 (SP); Paraná, Paranaguá, Ilha do Mel, on dead standing trunk of *Ocotea pulchella*, 6 May 1989, A.A.R. de Meijer 1234 (EMBRAPA); São Paulo, Agua Funda, Secretaria de Agricultura, on trunk of *Pinus* sp., M.A. de Jesús s.n. (SP); Teilha do Jardim Botânico de São Paulo, 12 February 2004, A.M. Guggliota & G.R. Leal 1205 (SP).

*Remarks.* The species is characterized by its very dark almost black pileus, relatively homogenous context, cuticle cells with many protuberances and branches, and basidiospores with subfree pillars. The specimen agrees with type, although the type is one basidioma in bad state, with only remains of the context, where it can not be seen if it is relatively homogenous, nor the resinous bands, but it was observed its colour: rust-brown (6E8). Ryvarden (2000, 2004) described basidiospores broadly ellipsoid, which do not agree with its measures (10-11 x 6-7  $\mu\text{m}$ ) or with the type. *Ganoderma orbiforme* is close related to *G. multiplicatum* but the last has smaller basidiospores with free pillars and their cuticle cells only have short protuberances. The species probably has a wider tropical distribution; it was recorded for the first time from Paraná (Ryvarden & de Meijer, 2002). Here, we recorded for the first time from other states: Rio de Janeiro and São Paulo.

*Ganoderma perturbatum* (Lloyd) Torrend, Bróteria Bot. 18: 34 (1920). Fig. 8  
= *Polyporus perturbatus* Lloyd, Mycol. Writ. 5, Let. 68: 11, 1918.

*Basidiomata* 4 x 5 x 1.2 cm, perenne, stipitate, single, woody-corky, light in weight. *Pileus* reniform, convex; surface glabrous, bumpy, semiglossy to glossy, concentrically sulcate; with a laccate crust, not cracking, difficult to remove, easy to penetrate with fingernail; violet-brown (darker than 11F7 or 11F8) almost homogeneous, without basidiospores over the surface; margin whitish to yellow, entire, obtuse, sulcate. *Stipe* 4.7 x 1 cm, lateral, cylindrical, solid, very dark violet-brown (11F8) to black, concolorous to generally darker than pileus; surface smooth to tuberculate, shiny, red-wine almost black, darker than pileus. *Context* 0.4 cm thick, fibrous, azonate, relatively homogenous, raw-sienna or light brown (6D7), gradually changing to slightly darker next to the tubes, resinous incrustations not very visible. *Pores* 3-4 per mm, angular to round, woody; pore surface yellow (3A2); tubes up to 0.8 cm thick, stratified, concolorous with the inferior part of the context. *Hyphal system* trimitic. *Contextual trama* with generative hyphae not observed; skeletal hyphae 2.5-7.5  $\mu\text{m}$  diam., generally solid to thick-walled, non-septate in the apex, arboriform,

golden-yellow; binding hyphae 1.9-2.2  $\mu\text{m}$  diam., thick-walled, yellowish, scarce. *Hymenophoral trama* as contextual trama. *Pileipellis* with cuticle cells 43.5 x 83.8  $\mu\text{m}$ , claviform, entire or rarely with one lateral protuberance; thick-walled, multistratified, golden-brown, without granulations in the apex; slightly amyloid with Melzer's reagent; generative hyphae 3.2  $\mu\text{m}$  diam., thin-walled, hyaline, scarce; skeletal hyphae 4.4-8  $\mu\text{m}$  diam., generally thick-walled, septate in the apex, arboriform, yellow to yellowish-brown. *Basidiospores* (9.9-) 10.8-12.4 (-13) x 8-9.3 (-9.9)  $\mu\text{m}$ ,  $Q = 1.14-1.43$ , broadly ellipsoid to ellipsoid, few subglobose, apex subacute, with apical germ pore difficult to observe, yellowish-brown, negative in Melzer's reagent; perisporium wrinkled, hyaline; exosporium with inter-walled pillars 0.6-0.8  $\mu\text{m}$  thick, anastomosed. *Basidia* not seen. *Cystidia* absent.

*Specimens examined.* BRAZIL, Lageado, s. date, R. Rick s.n. (BPI, Lectotype); Regiao Grande do Sul, on wood dead buried, 22 April 1961, J.P. da Costa Neto s.n. (SP).

*Remarks.* The distinctive features are the basidiomata stipitate, relatively homogeneous context, cuticle cells entire with multistratified wall and basidiospores with a subacute apex and anastomosed pillars. More likely this species displays a wider tropical distribution, but due to its little knowledge it has not been registered from other places. Steyaert (1967) synonymized it with *Ganoderma dorsale* and Ryvarden (1990) considered in the *Ganoderma lucidum* complex; nevertheless, the species have unique features. This is second record of the species for the world and the second for Brazil.

*Ganoderma perzonatum* Murrill, North Amer. Flora 9: 121 (1908). Fig. 9

*Basidiomata* 3.5-10 x 3-12 x 0.6-1.8 cm, annual, substipitate, single to imbricate, woody-corky, light in weight. *Pileus* flabelliform to round-flabelliform; surface glabrous, bumpy, glossy to semiglossy, concentrically sulcate; with a laccate crust, not cracking, difficult to remove, easy to penetrate with fingernail; violet-brown (11F8) almost homogeneous, degrading to garnet-red (11E8) towards the periphery, without basidiospores over the surface; margin whitish to generally concolorous, entire, thin, smooth. *Context* 0.3-0.9 cm thick, 0.7 cm average, fibrous, zonate, relatively homogeneous, pale-orange to light-orange (5A3, 5A4) above, degrading to light brown (6D7) toward the tubes; with resinous bands. *Pores* 4-6 per mm, angular to round, woody; pore surface pale-yellow (2A3) to sun-yellow (2A5); tubes 0.5-0.7 cm thick, unstratified to stratified, concolorous with the inferior part of the context. *Hyphal system* dimitic. *Contextual trama* with generative hyphae not observed; skeletal hyphae 6-13  $\mu\text{m}$  diam., generally solid to thick-walled, non-septate, arboriform. *Hymenophoral trama* as contextual trama. *Pileipellis* with cuticle cells 50-86 x 6.2-13.6  $\mu\text{m}$ , cylindrical, apex obtuse to rarely subcapitate, entire or with one lateral protuberance; thick-walled, generally multistratified in the apex, golden-yellow, with concentric elongate granulations in the apex; dextrinoid with Melzer's reagent; generative hyphae not observed; skeletal hyphae 5-9.3  $\mu\text{m}$  diam., generally solid-walled, non-septate, arboriform, yellowish to golden-yellow. *Basidiospores* 8.3-10.2 x 5.9-7.4  $\mu\text{m}$ ,  $Q = 1.25-1.33 -1.53$ , widely ellipsoid to ellipsoid, apex truncate, with apical germ pore, light yellowish-brown, negative in Melzer's reagent; perisporium wrinkled, hyaline; exosporium with inter-walled

pillars up to 0.3  $\mu\text{m}$  thick, free; endosporium wrinkled. *Basidia* 15.5-18.6 x 8-11  $\mu\text{m}$ , utriform, hyaline. *Cystidia* absent.

*Specimens examined.* CUBA, Santiago de Las Vegas on *Mango* log, F. S. Earle 309 (UPS, Lectotype). BRAZIL, Apaná, Grande do Curua, Bailipe, Igarape, on dead wood, 22 September 1988, H. Sotão & coll. 88.18.13 (SP); Mangué, Ilha de Maraca, on dead wood, January 1989, H. Sotão & coll. 88.21.06 (SP); Pará, Santarem, s. data, B. Lowy 22813 (SP); Rio Grande do Sul, Porto Alegre, Parque Farroupilha, on *Salix babylonica* trunk, 14 Jun 1960, J.C. Paim Costa s.n. (SP).

*Remarks.* The species can be identified by its pale context, remarkable cuticle cells cylindrical, apex obtuse to subcapitate, entire with concentric elongate granulations in the apex and small basidiospores. The studied specimens agree with the type, except that in the type the resinous bands are inconspicuous. Ryvarden (2000) described oblong ellipsoid basidiospores but of the same size described here. This is the second record of *Ganoderma perzonatum* for the world.

*Ganoderma pulverulentum* Murrill, North Amer. Flora 92: 121 (1908). Fig. 10

*Basidiomata* 10 x 15 x 1.5 cm, annual, sessile, imbricate, woody-corky, light in weight. *Pileus* round-flabelliform, plane; surface glabrous, bumpy, soft-corky, semiglossy, concentrically sulcate; with a laccate crust, not cracked, generally difficult to remove, easy to penetrate with fingernail; very dark reddish-black, almost black close to the base of the pileus, degrading to photo-brown (9F8), henna (7E8) to oxid-red (8E8) in the periphery, with basidiospores over the surface; margin whitish, lobulate, thin, smooth. *Context* 0.7-0.9 cm, fibrous-corky, relatively homogenous, zonate, ochre-yellow (5B6) degrading to dark brown (7F7) close to the tubes; with discontinuous resinous bands. *Pores* 4-5 per mm, angular to round, woody; pore surface cream to pale-yellowish (3A3), darkening when bruising or aging; tubes 0.7-1 cm thick, stratified, concolorous with the inferior part of the context. *Hyphal system* trimitic. *Contextual trama* with generative hyphae not observed; skeletal hyphae 2.8-12.8  $\mu\text{m}$  diam., generally solid, non-septate, arboriform or not, very branched, yellowish to golden-yellow, predominant; binding hyphae 1.6-4  $\mu\text{m}$  diam., solid, non-septate, hyaline to yellowish, scarce. *Hymenophoral trama* as contextual trama. *Pileipellis* with cuticle cells 37.2-59 x 8-16  $\mu\text{m}$ , narrowly clavate to clavate, generally entire; thick-walled, generally multistratified, apex with scarce granulations, golden-yellow to yellowish-brown, amyloid in Melzer's reagent; generative hyphae 4-4.8  $\mu\text{m}$ , thin-walled, with conspicuous clamps, hyaline; skeletal hyphae 2.8-7.4  $\mu\text{m}$  diam., generally thick-walled, at times septate, arboriform, very branched, yellowish-brown, predominant; binding hyphae 1.6-2.8  $\mu\text{m}$  diam., subthick-walled, non-septate, hyaline to yellowish, scarce. *Basidiospores* 9.6-12.8 x 6.2-8  $\mu\text{m}$ , Q = 1.41-1.64, ellipsoid, apex truncate, with apical germ pore, yellowish-brown, negative in Melzer's reagent; perisporium semi-wrinkled, reddish-yellow; exosporium with inter-walled pillars 0.4-0.5  $\mu\text{m}$  thick, partially anastomosed; endosporium smooth. *Basidia* not seen. *Cystidia* absent.

*Specimens examined.* **INDIA**, Grenada Island, on dry manchinell, W.E. Broadway s.n. (NY, Lectotype). **BRAZIL**, Bahia, Correntina, on trunk, 28 January 1967, D.M. Vital s.n. (SP).

*Remarks.* The distinctive features are the basidioma light in weight, cuticle cells entire with granulations and basidiospores with anastomosed pillars. The studied specimen agrees with the type, except that the type has thinner cuticle cells (8-11.2  $\mu\text{m}$  wide) and conspicuous granulations. The species is close related with *Ganoderma resinaceum* and *G. praelongum*. This is the second record of the species for the world.

*Ganoderma resinaceum* Boud., Bull. Soc. Mycol. Fr. 5: 72 (1889). Fig. 11  
= *Ganoderma chaffangeonii* Pat., Bull. Soc. Mycol. Fr. 5: 74, (1889).

*Basidiomata* 2.4-5 x 3-6.5 x 0.9-1.5 cm, annual, single, woody-spongy. *Pileus* round-flabelliform, convex to plane; surface glabrous, smooth to slightly bumpy, slightly soft, glossy, broadly concentrically sulcate; with a laccate crust, cracked, easy to remove and to penetrate with fingernail; violet-brown (darker than 11F7) to dark red in almost all the surface, gradually changing to orange (5A7) toward the margin or fully violet-brown (11F7), darker in adult basidiomata, with terra-cotta (7D7) basidiospores over the surface; margin whitish, brownish-orange to concolorous, entire, thin to obtuse, smooth. *Context* 0.4-1.3 cm thick, fibrous-spongy, azonate, homogeneous, light reddish-brown (7E7), with an apricot (5B6) thin fringe below the laccate crust, without resinous bands. *Pores* 4-5 per mm, angular to circular, woody; pore surface paste yellow (2A4) to yellow (3A2) when fresh, darkening to ochraceous or yellowish-brown (6C5) when aging or drying; tubes 0.1-0.5 cm long, stratified, concolorous with the context. *Hyphal system* trimitic. *Contextual trama* with generative hyphae not observed; skeletal hyphae 1.9-6.8  $\mu\text{m}$  diam., thick-walled, generally thick-walled to solid, non-septate, arboriform or not, moderately branched, golden-brown; binding hyphae not observed. *Hymenophoral trama* generative hyphae 1.9-3.1  $\mu\text{m}$  diam., thin-walled, hyaline, scarce; skeletal hyphae 2.8-7.5  $\mu\text{m}$  diam., generally thick-walled to solid, non-septate, arboriform or not, moderately branched, golden-brown; binding hyphae 2.8-4.4  $\mu\text{m}$  diam., generally thick-walled to solid, non-septate, golden-yellow. *Pileipellis* with cuticle cells 34-59 x 6.2-9.3  $\mu\text{m}$ , narrowly clavate, almost cylindrical, generally without protuberances neither branches, some with one lateral branch, thick-walled to solid, apex with granulations, brownish-yellow, amyloid in Melzer's reagent; generative hyphae 1.9-3.1  $\mu\text{m}$  diam., thin-walled, with conspicuous clamps, hyaline to yellowish, abundant; skeletal hyphae 1.9-4.3  $\mu\text{m}$  diam., subthick-walled to thick-walled, non-septate, sometimes arboriform, yellowish-brown; binding hyphae 1.9-3  $\mu\text{m}$  diam., thick-walled, non-septate, yellowish-brown. *Basidiospores* 11.2-12.5 x 6.5-7.4  $\mu\text{m}$ , Q = 1.5-1.72, ellipsoid to oblong, apex truncate, with apical germ pore, yellowish-brown; perisporium smooth, hyaline; exosporium with inter-walled pillars up to 0.4  $\mu\text{m}$  thick, free; endosporium wrinkled. *Basidia* not observed. *Cystidia* absent.

*Specimens examined.* **BRAZIL**, Mato Grosso, Rio Alto Juruena, August 1962, M. Mee s.n. (SP).

*Remarks.* This species is recognized by its basidiomata generally very dark and with homogenous brown context, furthermore by its cuticle cells entire and

cylindrical, and its basidiospores relatively big and with free pillars. *Ganoderma resinaceum* is a rare species, but organisms with many different features have been included under this name. For example, the materials de Meijer 1418, 2776 and 3238 (EMBRAPA) cited by Ryvar den & de Meijer (2002) under *G. resinaceum* correspond to another species. For this reason, and based in the literature its distribution is uncertain. The species had not been recorded from this area of Brazil.

*Ganoderma sessile* Murrill, Bull. Torrey. Bot. Club. 29: 64 (1902). Fig. 12

**Basidiomata** 5.5–13 × 5–13 cm, 1–3 cm thick in the base, annual, sessile, single to imbricate, woody-corky, light in weight, context the same wide than the tubes. **Pileus** semicircular, rounded flabelliform to flabelliform, conchate to convex; surface glabrous, bumpy, slightly to radially rugose, hard, glossy, with a laccate crust, not cracking, slightly easy to remove, easy to penetrate with fingernail, concentrically sulcate mainly toward the margin; violet-brown (10F6) or photo-brown (9F8) in the 80 to 90% of the surface, reddish-brown (8F8) to brownish-orange (6C8) in the periphery, or fully violet-brown very dark almost black, occasionally with raw sienna (6D7) basidiospores over the surface; margin whitish, henna (7E8), lighter than pileus or concolorous, entire, thin, rounded to acute, smooth. **Context** up to 1.5 cm thick in the base, 0.7–0.9 cm average, fibrous-corky, duplex, azonate, pale-orange to light-orange (5A3, 5A4) above and reddish-golden to light brown (6C7) close to the tubes; resinous bands generally diffuse and difficult to observe, almost up to the margin. **Pores** 4–5 per mm, angular to rounded, woody; pore surface yellowish-white (3A2), darkening to brown (6D8) when bruising or aging; tubes 0.8–1 cm thick, up to 1.4 cm in the base, unstratified, generally concolorous with the inferior part of the context. **Hyphal system** trimitic. **Contextual trama** with generative hyphae 2.4–4 µm diam., thin-walled, with large and conspicuous clamps, unbranched, hyaline to yellowish, difficult to observe; skeletal hyphae 1.6–12 µm diam., generally solid, non-septate, arboriform, very branched, yellowish to golden-yellow, predominant; binding hyphae 1.6–4 µm diam., solid, non-septate, hyaline to yellowish, scarce. **Hymenophoral trama** as contextual trama. **Pileipellis** with cuticle cells (40.3–) 60–88 × 8–16 µm, clavate, generally without or with one lateral protuberance, thick-walled, generally multistratified, golden-yellow, content immediately black with Melzer's reagent, cells amyloid immediately; generative hyphae 1.9–2.8 µm diam., thin-walled, with conspicuous clamps, branched, hyaline to yellowish, abundant; skeletal hyphae 2.4–9.6 µm diam., generally solid, non-septate, arboriform, yellow, predominant; binding hyphae 1.6–4 µm diam., generally solid to thick-walled, non-septate, hyaline to yellowish, notably thinner and paler than skeletal hyphae, in some specimens not observed. **Basidiospores** 11.2–13.6 × 7.4–9.3 µm, Q = 1.33–1.63, ellipsoid to oblong, apex truncate, with apical germ pore, yellowish-brown; perisporium smooth, hyaline; exosporium with inter-walled pillars 0.5–0.6 µm thick, subfree; endosporium wrinkled. **Basidia** not observed. **Cystidia** absent.

*Specimens examined.* USA, New York, Bedford Park, on *Quercus* trunk, s. date, s. coll., (NY, Lectotype). BRAZIL, s. date, s. coll. (SP).

*Remarks.* The species is easily recognized by its pale context, cuticle cells entire and big basidiospores. The Brazilian specimen agrees with the type, but this



has slightly bigger basidiospores, 12-14.4 x 7.2-8.8 (-9.6)  $\mu\text{m}$ . This is the first record of the species for Brazil.

*Ganoderma sessiliforme* Murrill, Bull. New York. Bot. Gard. 8: 149 (1912).

Fig. 13

$\equiv$  *Fomes sessiliformis* (Murrill) Murrill, Bull. New York. Bot. Gard. 8: 153, 1912.

**Basidiomata** 4-7 x 5.7-10 x 1.1-1.2 cm, sessile to substipitate, annual, single or occasionally imbricate, woody, but light in weight. **Pileus** flabelliform to semicircular, somewhat conchate to convex; surface glabrous, rugose to radially rugose, hard, semiglossy, slightly concentrically sulcate; with a laccate crust, not cracking, easy to remove and to penetrate with fingernail; darker than violet-brown (9F8) in the 80% of the surface degrading to wine-red (11D8) toward the periphery, without basidiospores over the surface; margin whitish to mandarine-orange (6B8), entire to slightly lobulate, acute to obtuse, smooth. **Substipe** 3 x 1-1.5 cm, horizontal, flattened, solid, surface shiny, red-wine to almost black, darker than pileus. **Context** 0.5-0.7 cm, fibrous, azonate, homogeneous to relatively homogeneous, orange-white (5A2) to cocoa (6E6), with a deep yellow (4A8) fringe below the laccate crust, without resinous bands. **Pores** 3-4 per mm, angular, woody; pore surface pale-yellow to pastel-yellow (3A3, 3A4), darkening to brown (6D8) when bruising or aging; tubes 0.4-0.6 cm thick, darker than context. **Hyphal system** dimitic. **Contextual trama** with generative hyphae not observed; skeletal hyphae 2.5-11.2  $\mu\text{m}$  diam., mainly solid to some thick-walled, non-septate, arboriform, branched, yellowish to yellow, predominant. **Hymenophoral trama** as the contextual trama. **Pileipellis** with cuticle cells 52.7-68.2 x 6.8-11.8  $\mu\text{m}$ , clavate, generally entire, thick-walled, golden-yellow, without granulations, slightly amyloid with Melzer's reagent; generative hyphae not observed; skeletal hyphae 3.7-7.4  $\mu\text{m}$  diam., subthick to thick-walled, non-septate, arboriform but with few branches, yellowish to golden-yellow, predominant. **Basidiospores** 8.7-9.9 (-10.3) x 6.3-7.1  $\mu\text{m}$ , Q = 1.31-1.57, ellipsoid, apex truncate, with apical germ pore, yellowish-brown, negative in Melzer's reagent; perisporium wrinkled, hyaline; exosporium with inter-walled pillars 0.4-0.5  $\mu\text{m}$  thick, partially anastomosed, but only in some zones and in some basidiospores free; endosporium wrinkled. **Basidia** not observed. **Cystidia** absent. **Chlamydospores** 11-15.5 x 7.4-10.5  $\mu\text{m}$ , Q = 1.35-1.6 (-2), ellipsoid, with one or both apexes mucronate, thick-walled, yellowish.

**Specimens examined.** MEXICO, Morelos, Municipality of Cuernavaca, on dead wood, 24-27 December 1909, E. & L. Murrill 392 (NY, Lectotype). BRAZIL, Rio Grande do Sul, Porto Alegre, Parque Saint Hillaire, on wood, 30 Jun 1960, M.H. Homrich s.n. (SP).

**Remarks.** This species has a basidiomata generally conchate, context pale without resinous deposits and basidiospores relatively small. *Ganoderma sessiliforme* was not included in the last treatment on Neotropical Polypores (Ryvarden 2004). This is the first record of the species for Brazil. It is a rare species only known from Argentina, Brazil and Mexico.

*Ganoderma subfornicatum* Murrill, North Amer. Flora 9: 121 (1908). Fig. 14

**Basidiomata** 3.4-10.5 x 4-8 x 1.3-1.8 cm, perenne, substipitate to rarely with a long stipe, with a contracted base, single, woody. **Pileus** round-flabelliform, generally plane; surface glabrous, bumpy, slightly to radially rugose, very shiny, remarkable concentrically sulcate; with a laccate crust, difficult to remove, hard but easily to penetrate with fingernail; totally reddish-brown (darker than 9F8) almost black, homogenous, generally covered with cinnamon (6D6) basidiospores; margin concolorous with the pileus, entire, thick, obtuse to truncate, sulcate. **Substipe** 1.4-2 (-5.3) x 1.2-2.3 cm, generally short and thick, cylindrical, horizontal or lateral, concolorous with the pileus, solid. **Context** 0.4-0.8 cm thick, fibrous, relatively homogeneous, yellowish-brown (4B7) above, gradually changing to rust-brown (6E8) next to the tubes; with resinous bands. **Pores** 4-6 per mm, round, woody, very small; pore surface brown (6D8); tubes 0.6-1 cm long, stratified, concolorous with base of the context. **Hyphal system** trimitic. **Contextual trama** with generative hyphae 3.2 µm diam., thin-walled, with large and conspicuous clamps, hyaline, scarce and difficult to observe; skeletal hyphae 5.6-11.2 µm diam., generally solid, not-branched or with few branches, golden-yellow to yellowish-brown, predominant; binding hyphae 1.2-3.2 µm diam., solid, non-septate, hyaline to yellowish, scarce, notably thinner and paler than skeletal hyphae. **Hymenophoral trama** as contextual trama. **Pileipellis** with cuticle cells 37.2-62 x 5-9.3 µm, not claviform, commonly with a constriction, generally with up to five protuberances and one to two branches, mainly solid to thick-walled, not multistratified wall, apex with ferruginous granulations, yellowish, slightly amyloid with Melzer's reagent; generative hyphae 2.4-4.3 µm diam., thin-walled, with conspicuous clamps, branched, hyaline to yellowish, abundant; skeletal hyphae 4.3-8 µm diam., generally solid, occasionally septate, arboriform, golden-yellow; binding hyphae 1.9-3.7 µm diam., solid, non-septate, yellowish to golden-yellow, abundant, notably thinner and paler than skeletal hyphae. **Basidiospores** 9.6-11.8 x 6.4-8 µm, Q = 1.41-1.69, ellipsoid to oblong, apex subacute, slightly visible apical germ pore, yellowish; perisporium semi-wrinkled, exosporium with inter-walled pillars up to 0.4 µm thick, subfree; endosporium semi-wrinkled. **Basidia** not seen. **Cystidia** absent.

*Specimens examined.* **HONDURAS**, s. loc., on dead wood, 1906, *M.E. Peck s.n.* (NY, Lectotype). **BRAZIL**, Paraná, Paranaguá, Ponta do Sul, on wood, 10 October 1968, G. Hatschbach 20106 (SP); Pernambuco, Recife, Dois Irmãos, 1 May 1956, s.coll.; (SP); Antonina, Parque Morumbi, Rio do Nunes, on dead dicotyledon trunk, 12 December 1987, A.A.R. de Meijer 962 (EMBRAPA); Rondônia, Rio Ji-Paraná, Acampamento JP-15, on wood, 8 December 1987, M. Capelari 1770 (SP); São Paulo, Cananéia, Ilha Comprida, 2 km from the beach, on wood, 25 February 1983, O. Yano & J.R. Pirani 5977 (SP); Sergipe, SE Estação Ecológica Santa Isabel, 11 November 2003, U. da Silva-Aragão s.n.(SP).

*Remarks.* This is a conspicuous species by its shiny and dark basidioma that contrasts with a pale context with resinous bands; micromorphologically it is distinguished by its cuticle cells with many protuberances. The species was not included in the last treatment on Neotropical Polypores (Ryvarden 2004). According with Moncalvo & Ryvarden (1997) the species had been reported throughout the tropics. This species was recorded from Brazil by Rajchenberg & de Meijer (1990); later Ryvarden & de Meijer (2002) mentioned that this report was incorrect, because the materials corresponded to *G. orbiforme*. However, we studied those specimens and one (de Meijer 1234) is *G. orbiforme* and the other

(de Meijer 962) comprises two basidiomata, one is *G. subfornicatum* and the other one (marked as de Meijer 962A) is *G. applanatum* (see above). *Ganoderma subfornicatum* is recorded for the second time from Brazil.

*Ganoderma weberianum* (Bres. et Henn. ex Sacc.) Steyaert, Persoonia 7(1): 79 (1972). Fig. 15

= *Ganoderma rivulosum* Pat. & Har., Bull. trimest. Soc. myc. Fr. 22: 119, 1906.

**Basidiomata** 4-5.5 x 4-5 x 0.9-1.3 cm, annual, substipitate, with a contracted base, single to imbricate, woody. **Pileus** semicircular, round-flabelliform to flabelliform, convex to plane; surface glabrous, rivulose to slightly radially rugose, hard, glossy to dull, with a laccate crust, not cracking, difficult to penetrate with fingernail, easily lost leaving the surface relatively homogeneously dull, zonate; reddish-black to violet-brown (11F7) or darker in almost all the surface, except to the margin where gradually changes to orange (5A7), or fully violet-brown (11F7), with terra-cotta (7D7) basidiospores over the surface; margin pure white or yellowish, entire to slightly lobulate, thin, smooth. **Substipe** when present 0.9-1.5 x 1-1.5 cm, short and thick, thinner toward the base, horizontal, slightly darker than pileus, solid. **Context** 0.6-1 cm thick, fibrous, relatively homogeneous, zonate, light-yellow (4A4), darkening to reddish-brown (8F7) close to the tubes, changing to yellow when cut, with resinous incrustations throughout the context. **Pores** 4-5 per mm, angular to round, woody; pore surface yellowish-white (3A2) to sun-yellow (2A5) when fresh, darkening to ochraceous or yellowish-brown (6E8) when aging and drying; tubes 0.2-0.4 cm long, unstratified, concolorous with lower part of the context. **Hyphal system** dimitic. **Contextual trama** with generative hyphae not observed; skeletal hyphae 1.6-8 µm diam., thick-walled, generally solid, non-septate, non-branched to arboriform, moderately branched, golden-yellow. **Hymenophoral trama** as contextual trama. **Pileipellis** with cuticle cells 56.4-74.4 x 6.2-9.9 µm, cylindrical to narrowly clavate, entire, thick-walled to solid, apex with granulations, golden-yellow, slightly amyloid in Melzer's reagent; generative hyphae 1.9-3.7 µm diam., thin-walled, with conspicuous clamps, non-branched, hyaline to yellowish; skeletal hyphae 4.3-7.4 µm diam., thick-walled to solid, non-septate, non-branched to arboriform with few branches, apex rounded and slightly wider, golden-yellow. **Basidiospores** 8.8-9.3 x 6.8-7.2 µm, Q = 1.29-1.5, ellipsoid, few broadly ellipsoid, apex truncate, with apical germ pore, yellowish-brown; perisporium wrinkled, hyaline; exosporium with inter-walled pillars up to 0.5 µm thick, subfree; endosporium wrinkled. **Basidia** 13.6-17.3 x 8-9.3 µm, widely claviform, hyaline. **Cystidia** absent. **Chlamydospores** 9.3-10.5 µm, globose, thick-walled, with inter-walled very thick pillars, yellow.

*Specimens examined.* BRAZIL, São Paulo, S. Paulo Brotas, km 216-217 of Rodovia, Região de Cerrado, 11 January 1962, A. Milanez & D. Altimari s.n. (SP).

*Remarks.* *Ganoderma weberianum* may be recognized due to its hard cuticle, pale context that changes to yellow when cut, whit resinous incrustations, cuticle cells cylindrical with granulations and small basidiospores. The specimen agrees with Wang *et al.* (2005) description; who recorded the species from China, except that they described clavate cuticle cells although in their pictures are shown cylindrical. This is the second record for America and the first for Brazil.

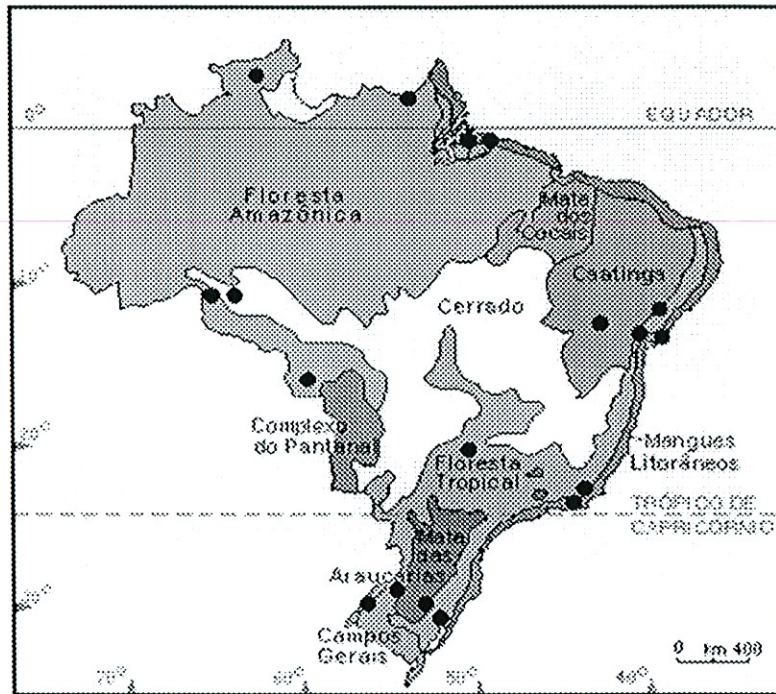
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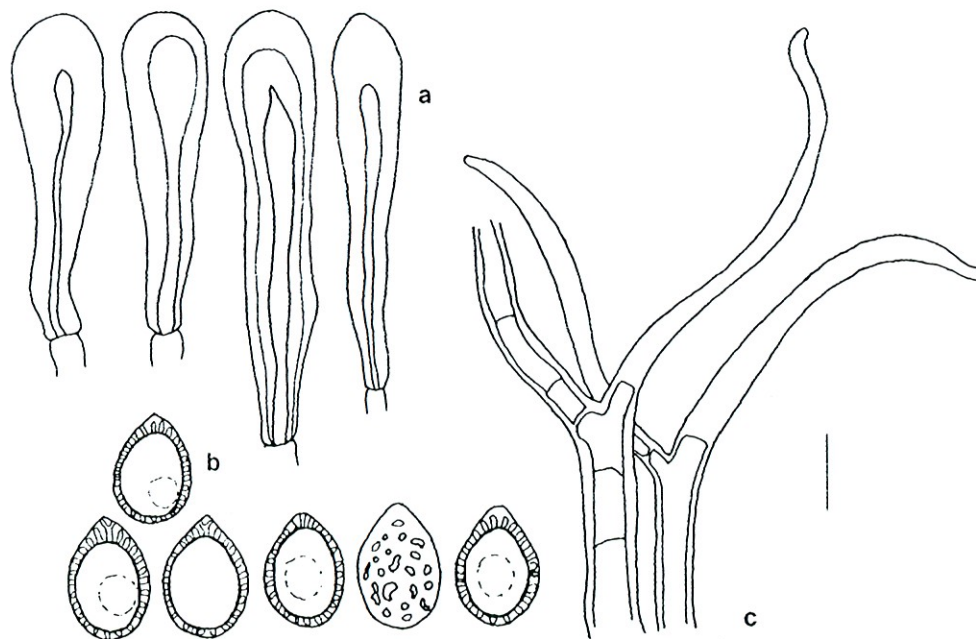
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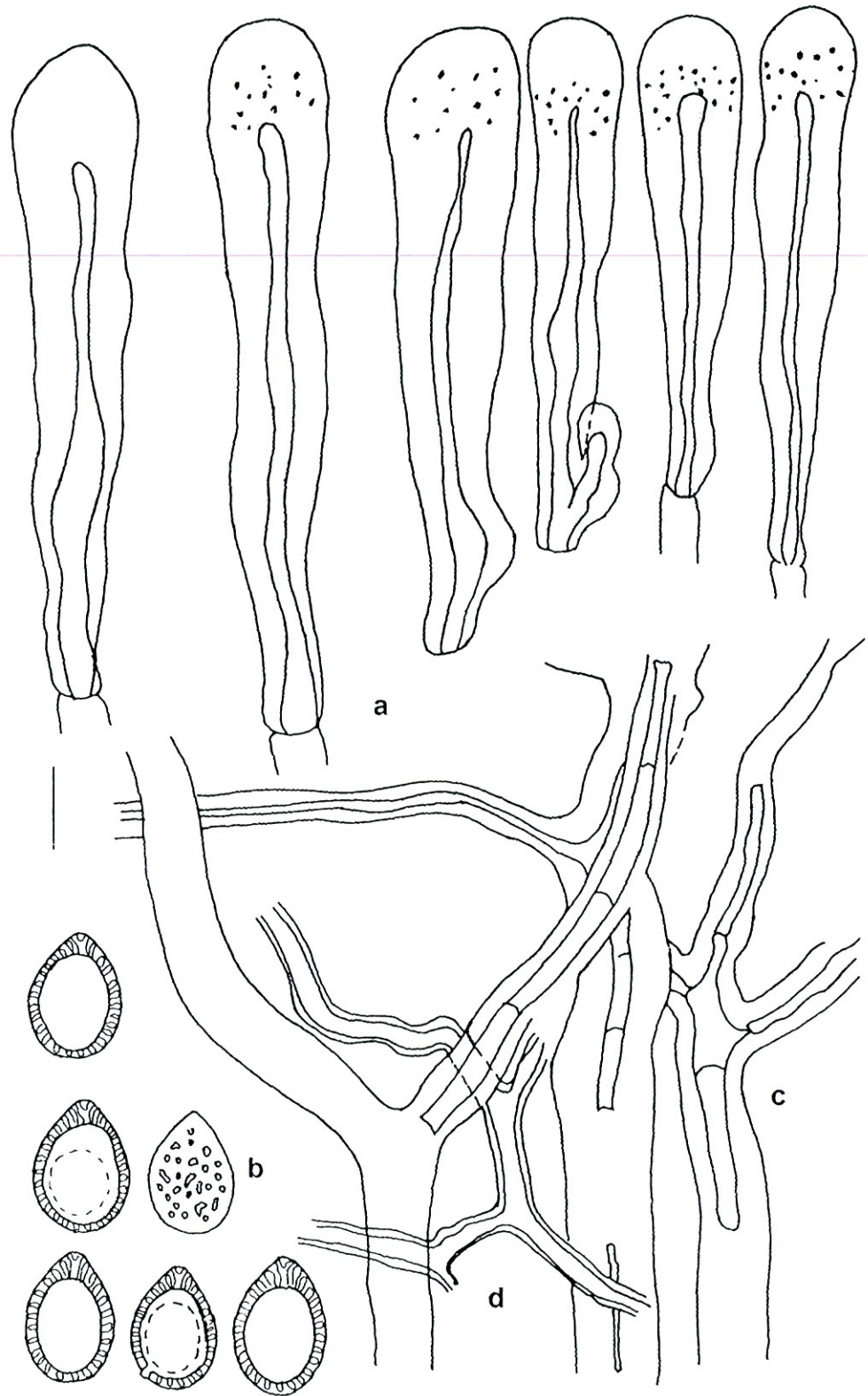
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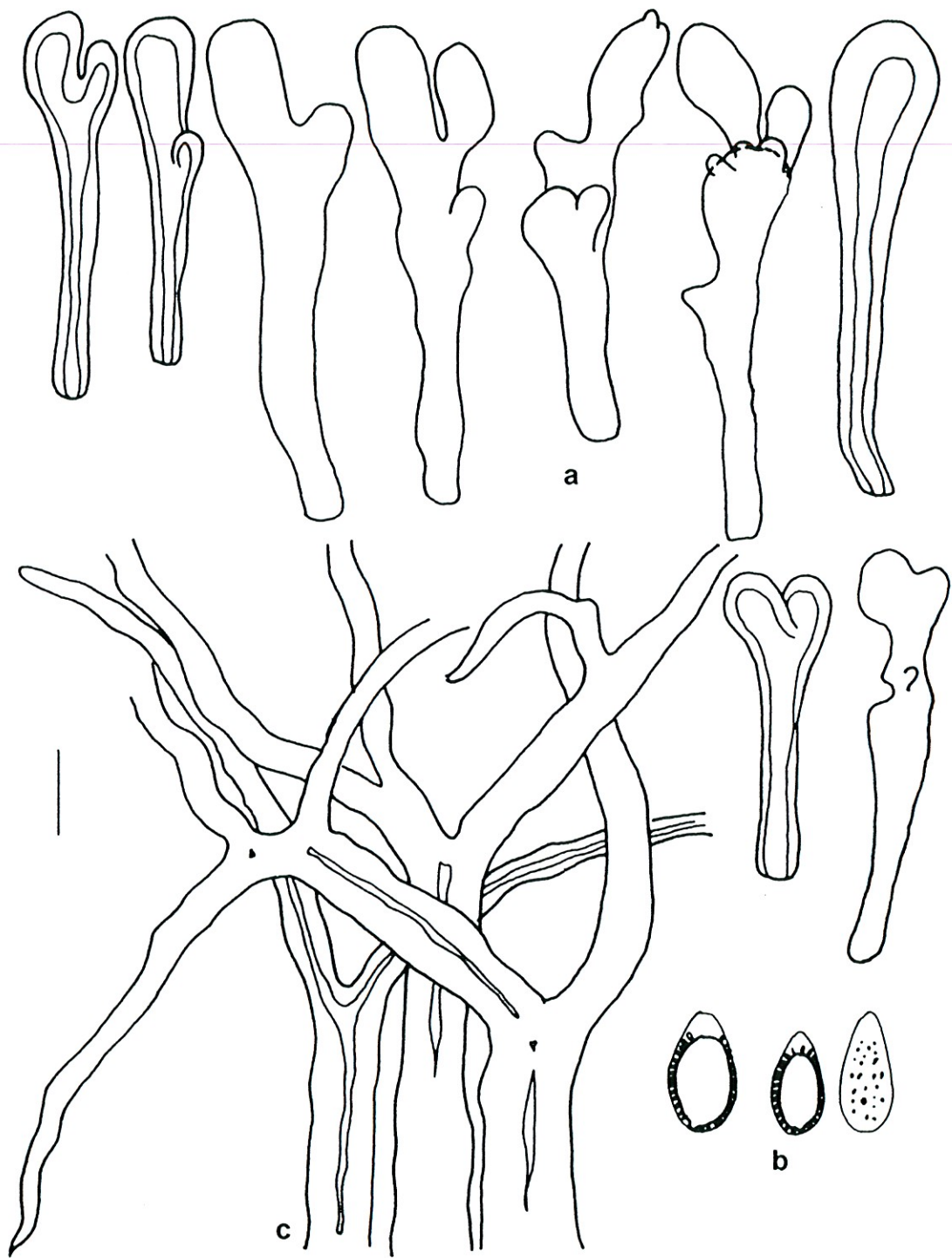
**Fig. 1.** Map of Brazil vegetation. The points show the studied localities (Map from [www.ibge.gov.br](http://www.ibge.gov.br))



**Fig. 2.** Micromorphological features of *Ganoderma conccinum*: a. Cuticle cells. b. Basidiospores. c. Skeletal hyphae. Bar = 8  $\mu$ m.

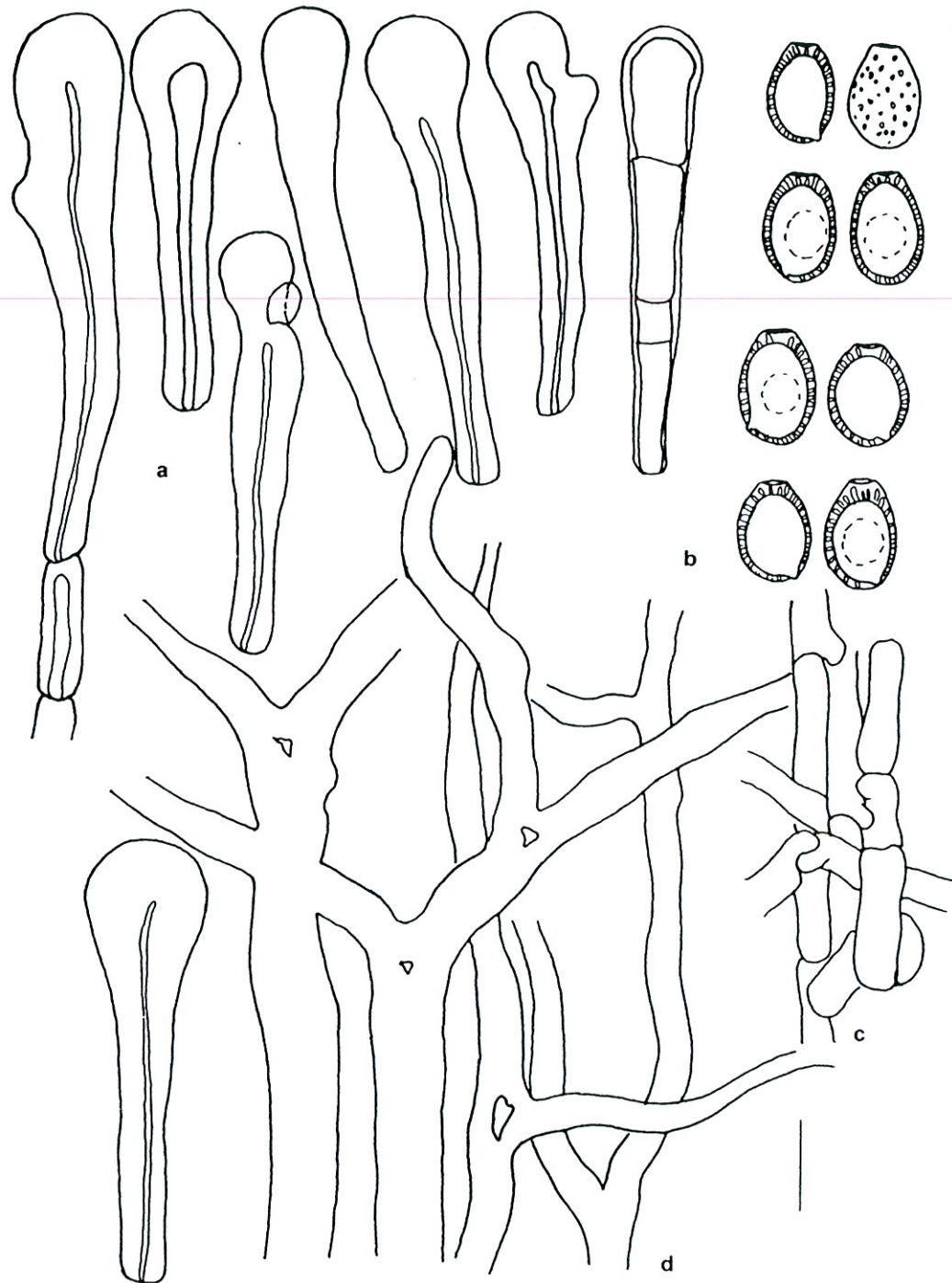


**Fig. 3.** Micromorphological features of *Ganoderma dorsale*: a. Cuticle cells. b. Basidiospores. c–d. Hyphal system of crustohymenodermis: c. Skeletal hyphae. d. Binding hyphae. Bar = 8  $\mu$ m.

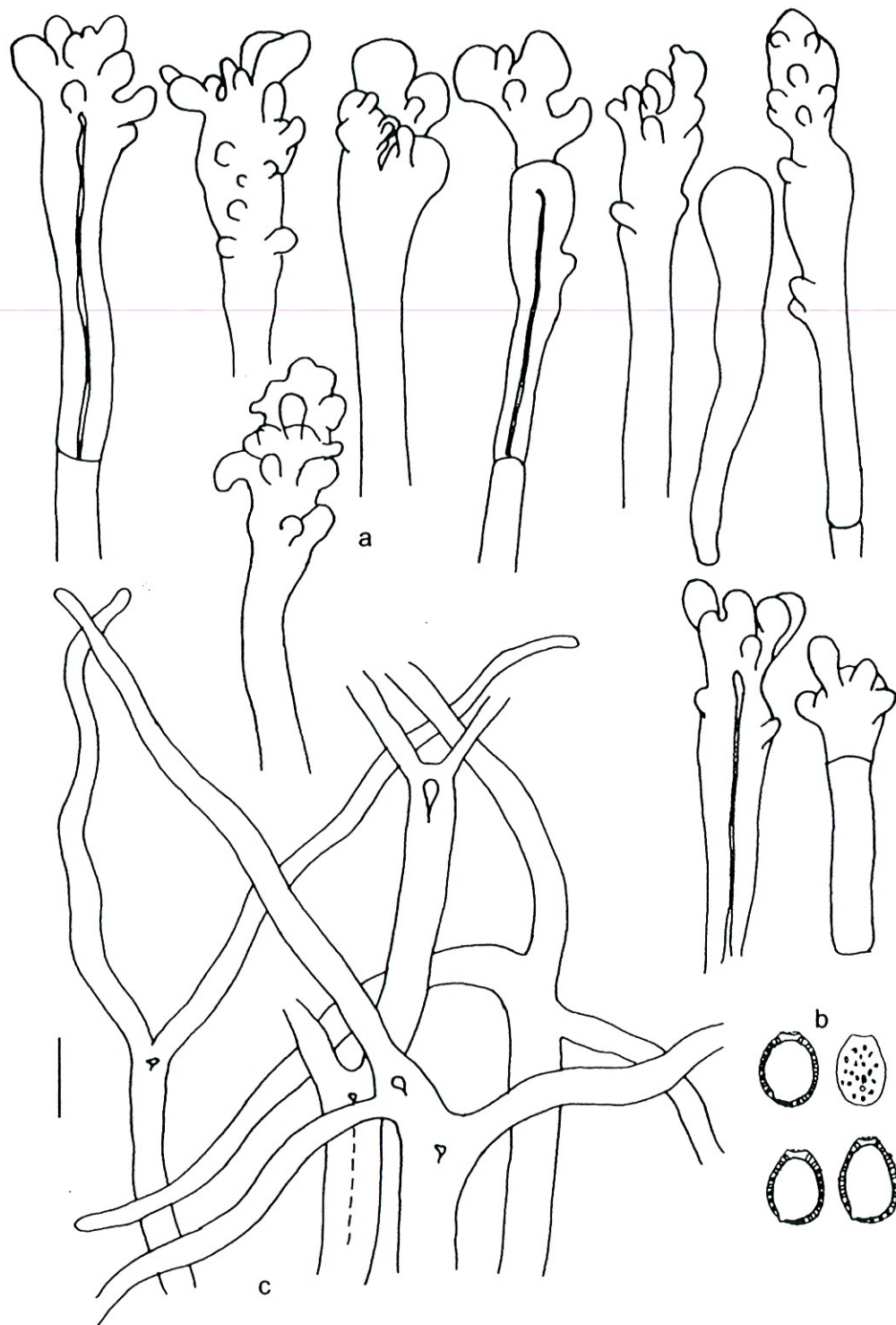


**Fig. 4.** Micromorphological features of *Ganoderma elegantum*: a. Cuticle cells. b. Basidiospores. c. Hyphal system of crustohymenodermis: Skeletal hyphae. Bar = 8  $\mu$ m.

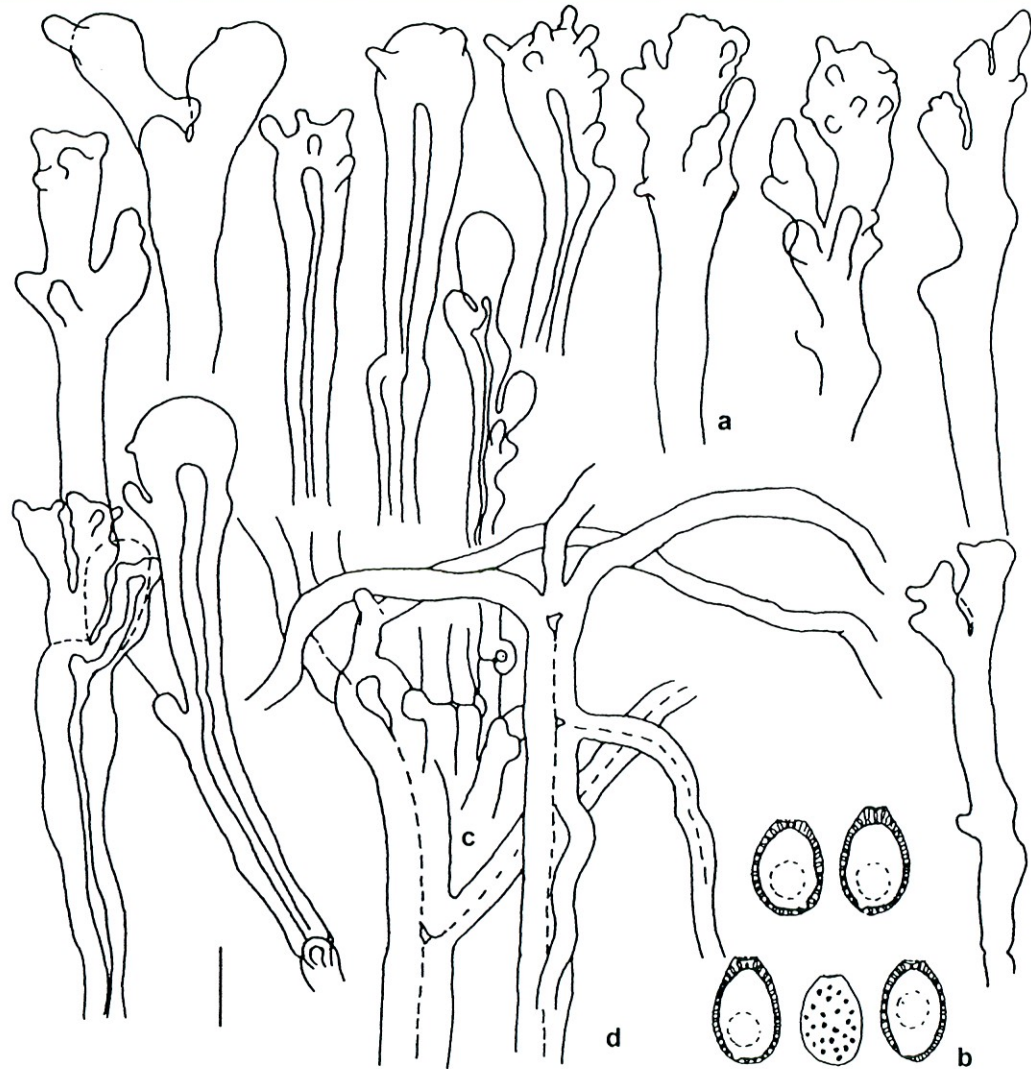




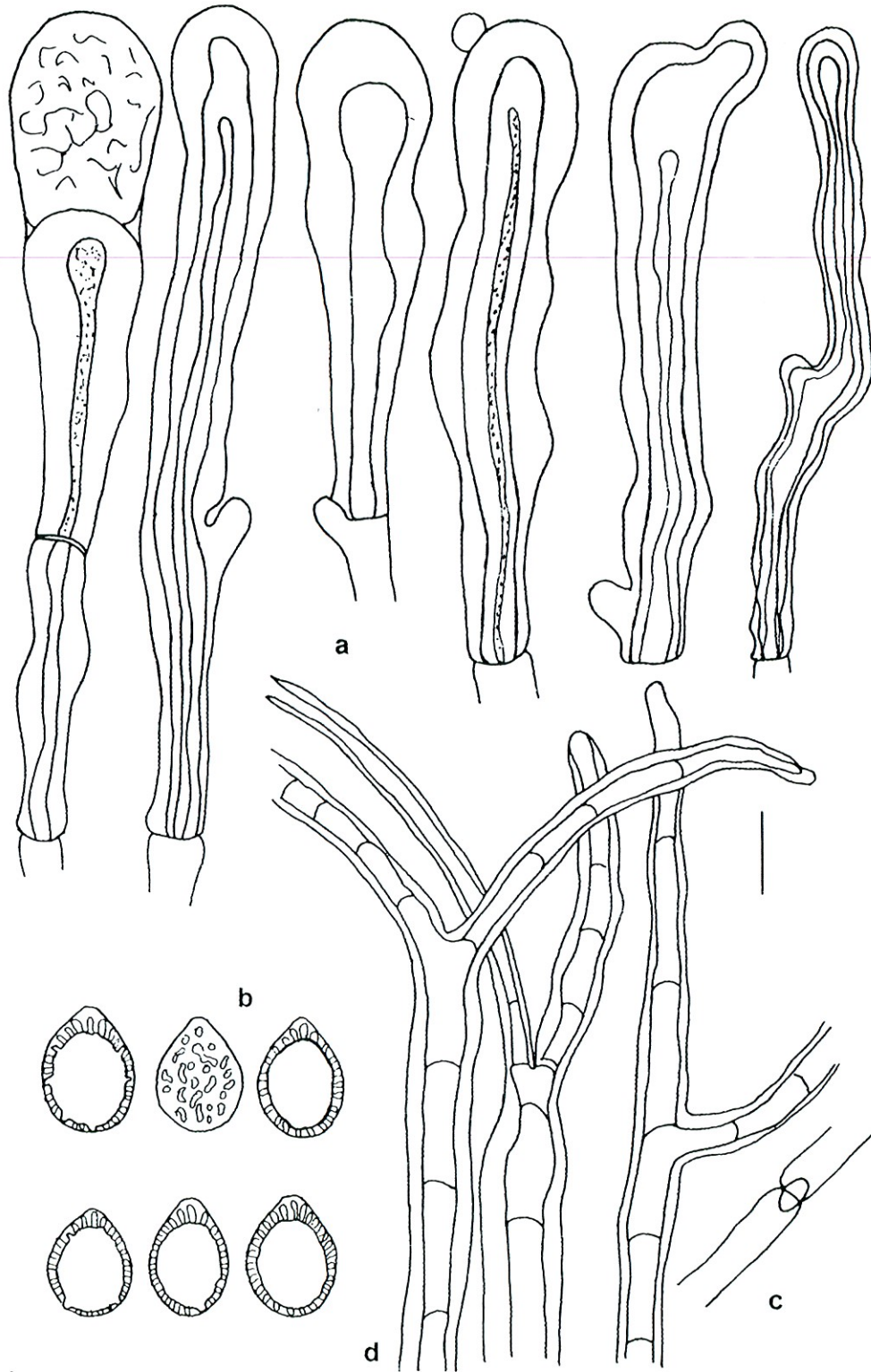
**Fig. 5.** Micromorphological features of *Ganoderma mexicanum*: a. Cuticle cells. b. Basidiospores. c–d. Hyphal system of crustohymenodermis: c. Generative hyphae. d. Skeletal hyphae. Bar = 8  $\mu$ m.



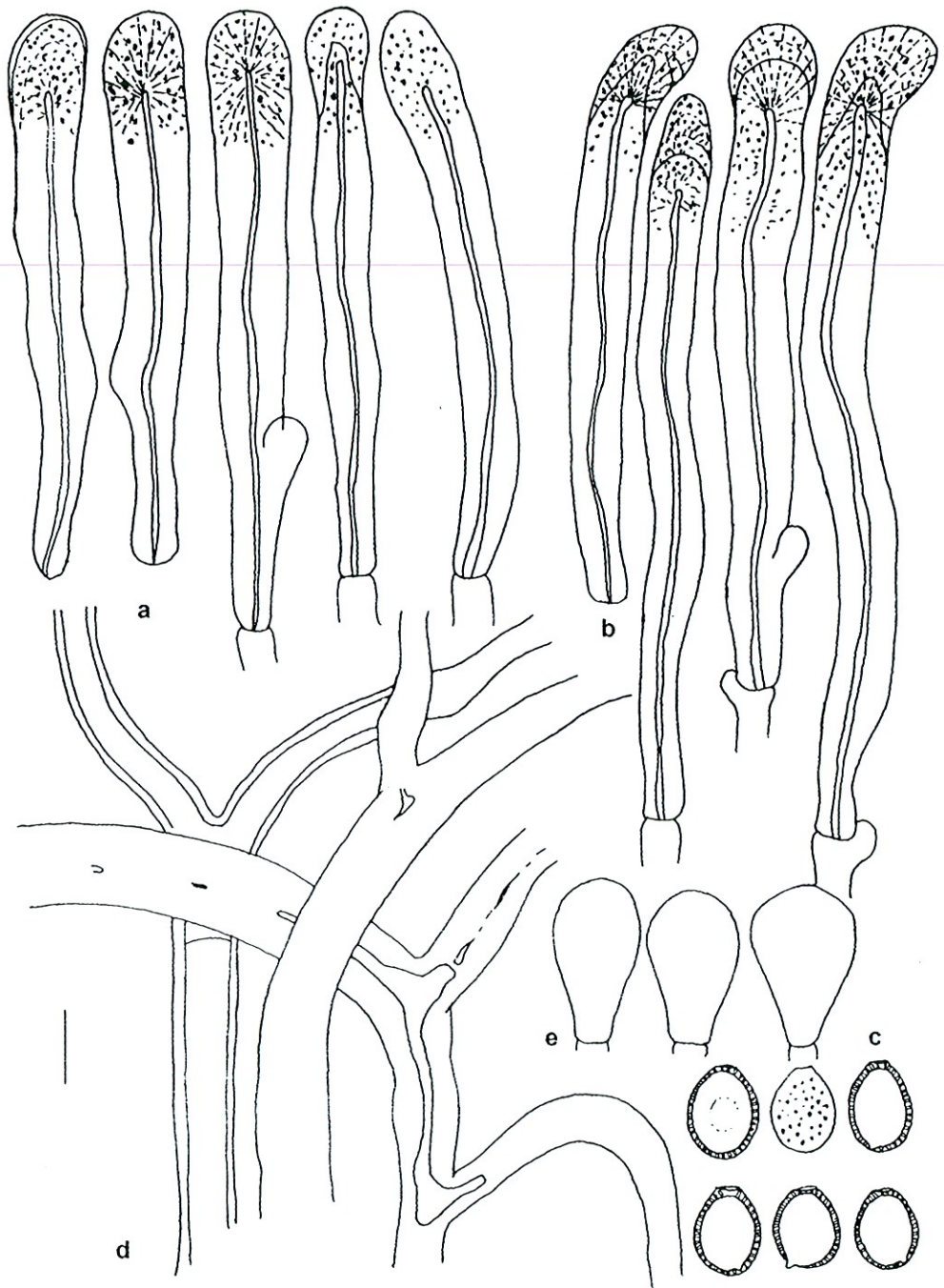
**Fig. 6.** Micromorphological features of *Ganoderma multiplicatum*: a. Cuticle cells. b. Basidiospores. c. Hyphal system of crustohymenodermis: Skeletal hyphae. Bar = 8  $\mu$ m.



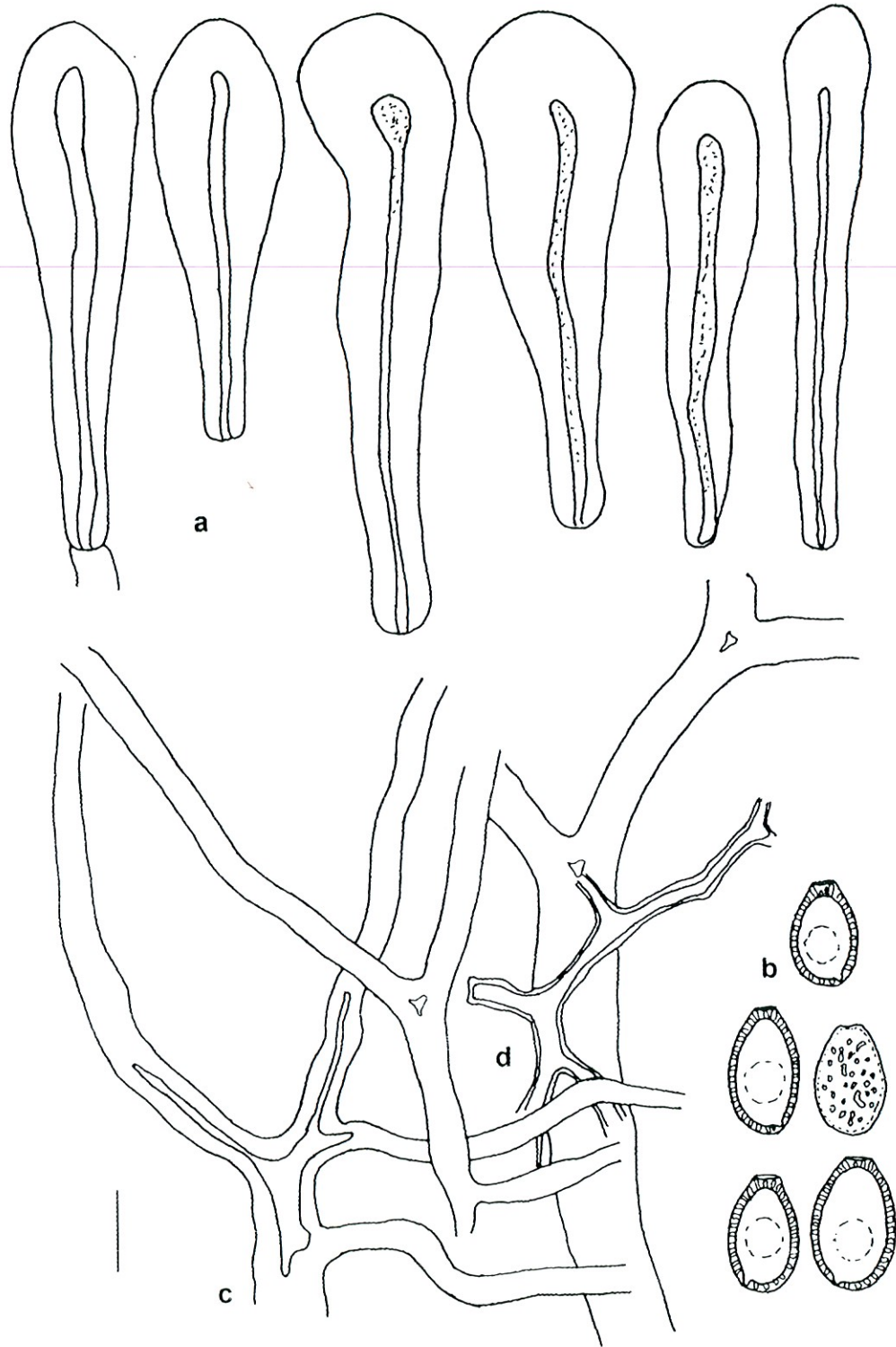
**Fig. 7.** Micromorphological features of *Ganoderma orbiformum*: a. Cuticle cells. b. Basidiospores. c–d. Hyphal system of crustohymenodermis: c. Generative hyphae. d. Skeletal hyphae. Bar = 8  $\mu$ m.



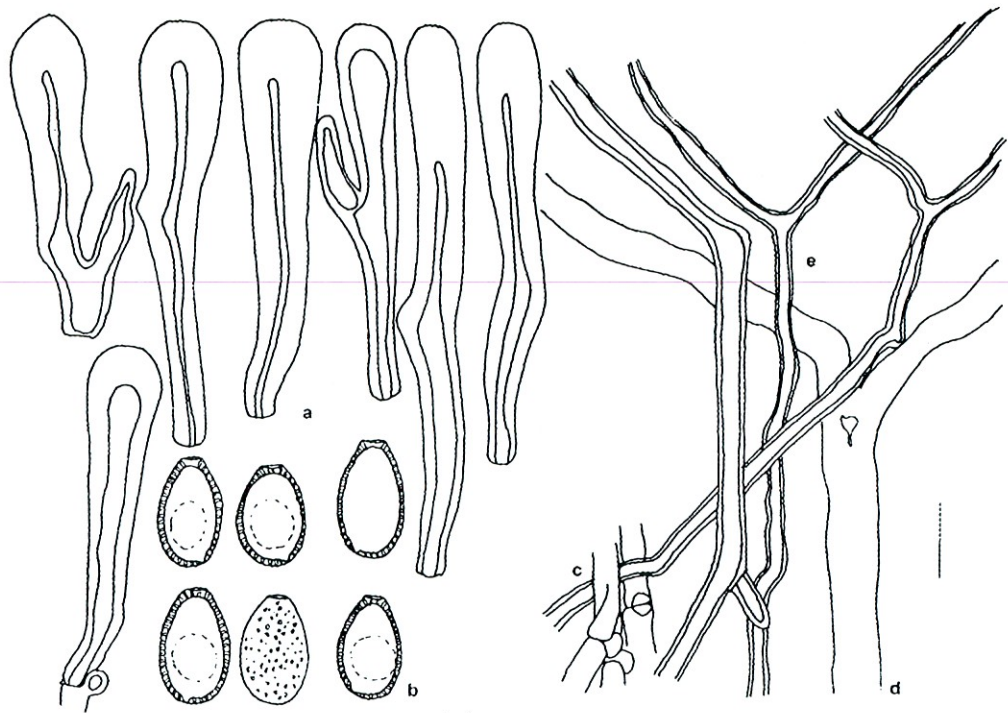
**Fig. 8.** Micromorphological features of *Ganoderma perturbatum*: a. Cuticle cells. b. Basidiospores. c–d. Hyphal system of crustohymenodermis: c. Generative hyphae. d. Skeletal hyphae. Bar = 8  $\mu$ m.



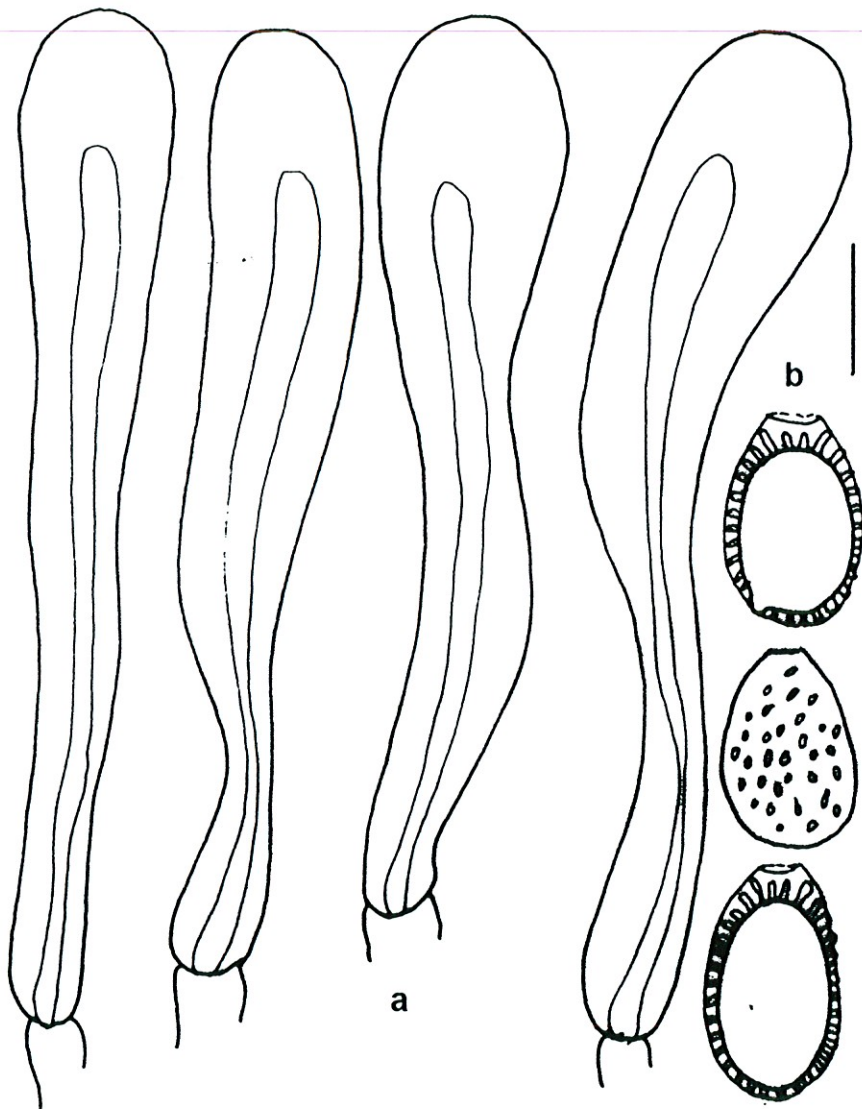
**Fig. 9.** Micromorphological features of *Ganoderma perzonatum*: a-b. Cuticle cells. c. Basidiospores. d. Hyphal system of crustohymenodermis: Skeletal hyphae. E. Basidiola. Bar = 8  $\mu$ m.



**Fig. 10.** Micromorphological features of *Ganoderma pulverulentum*: a. Cuticle cells. b. Basidiospores. c–d. Hyphal system of crustohymenodermis: c. Skeletal hyphae. d. Binding hyphae. Bar = 8  $\mu\text{m}$ .

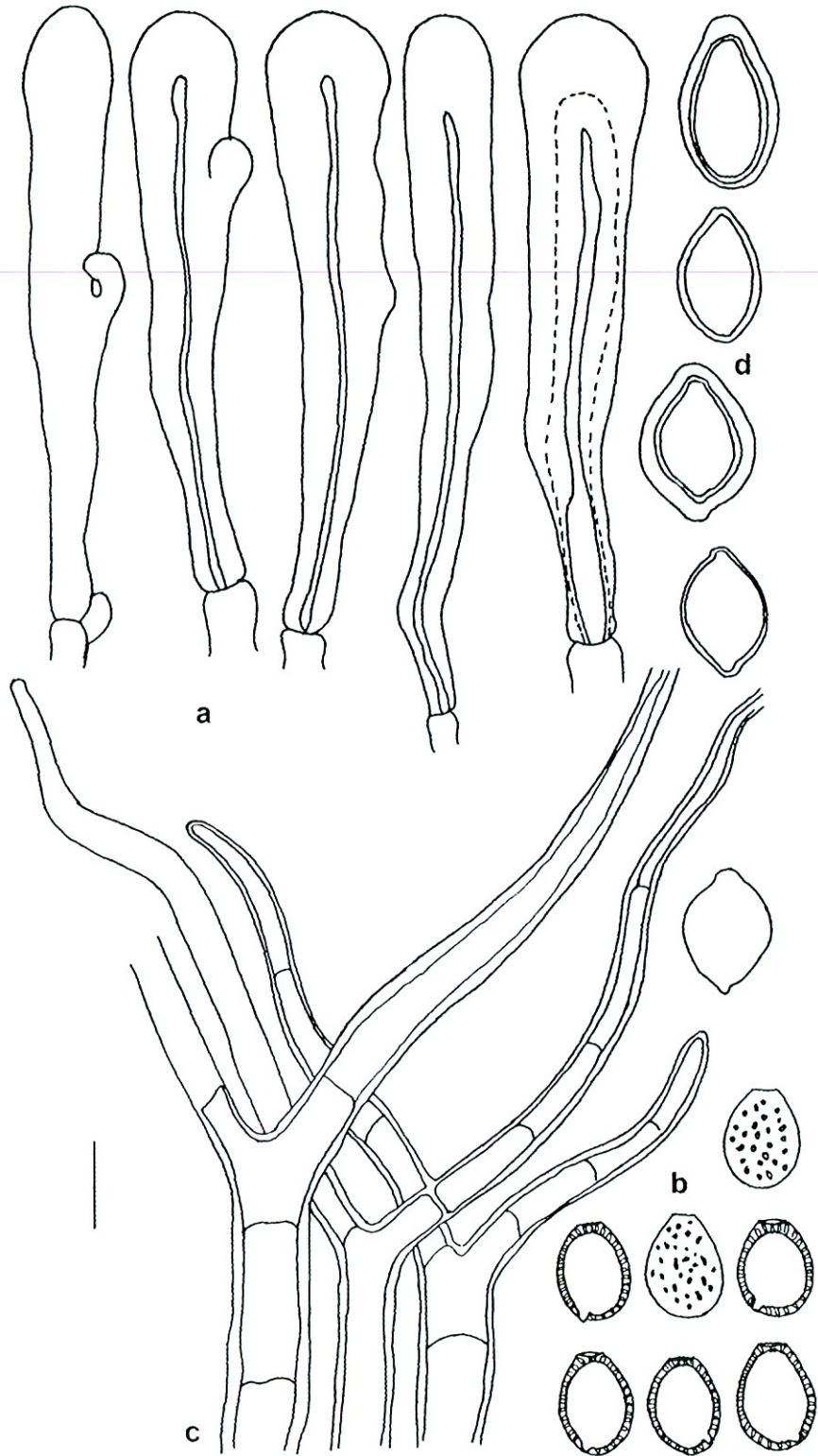


**Fig. 11.** Micromorphological features of *Ganoderma resinaceum*: a. Cuticle cells. b. Basidiospores. c–e. Hyphal system of crustohymenodermis: c. Generative hyphae. d. Skeletal hyphae. e. Binding hyphae. Bar = 8  $\mu$ m.

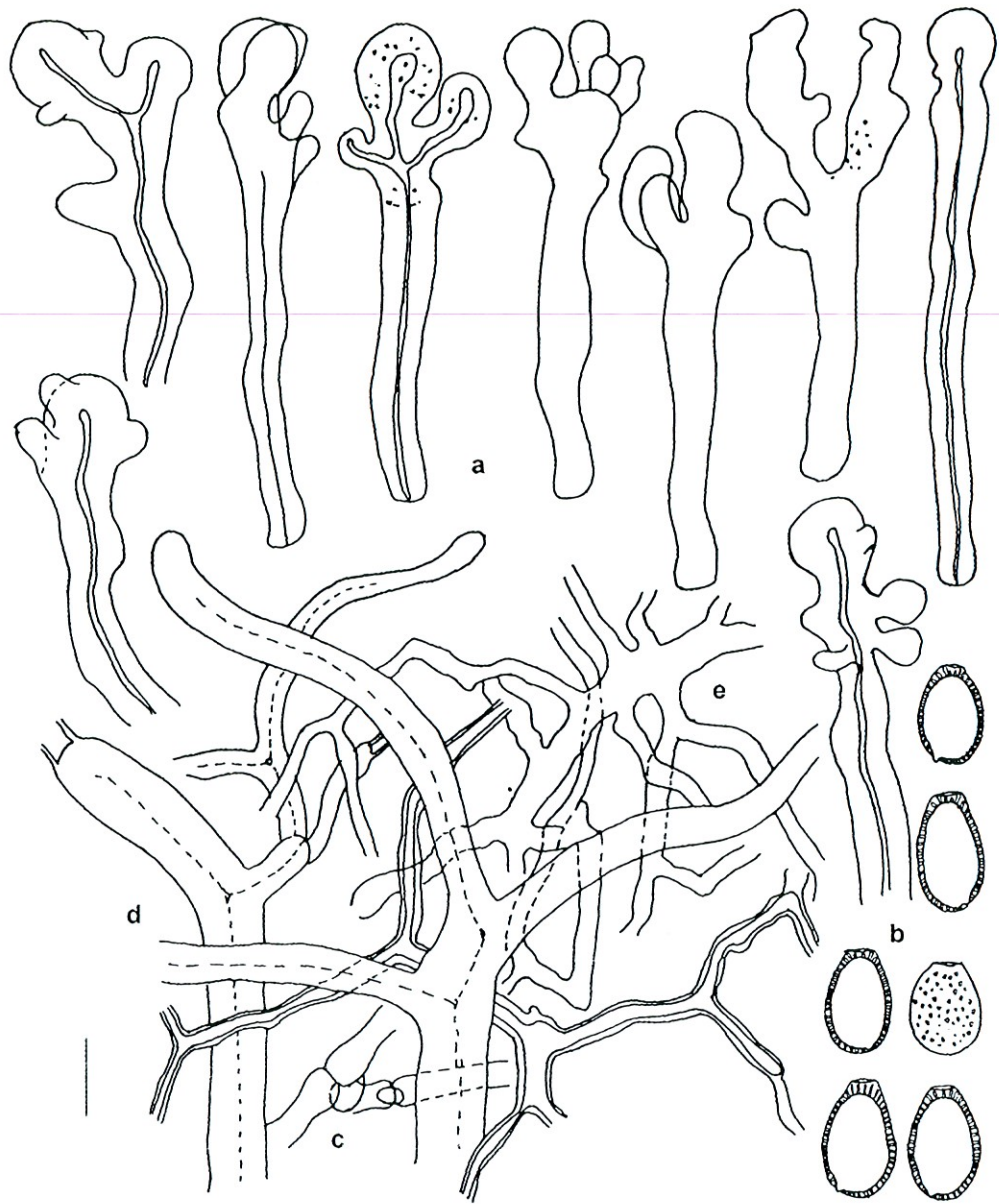


**Fig. 12.** Micromorphological features of *Ganoderma sessile*: a. Cuticle cells. b. Basidiospores. Bar = 8  $\mu$ m.

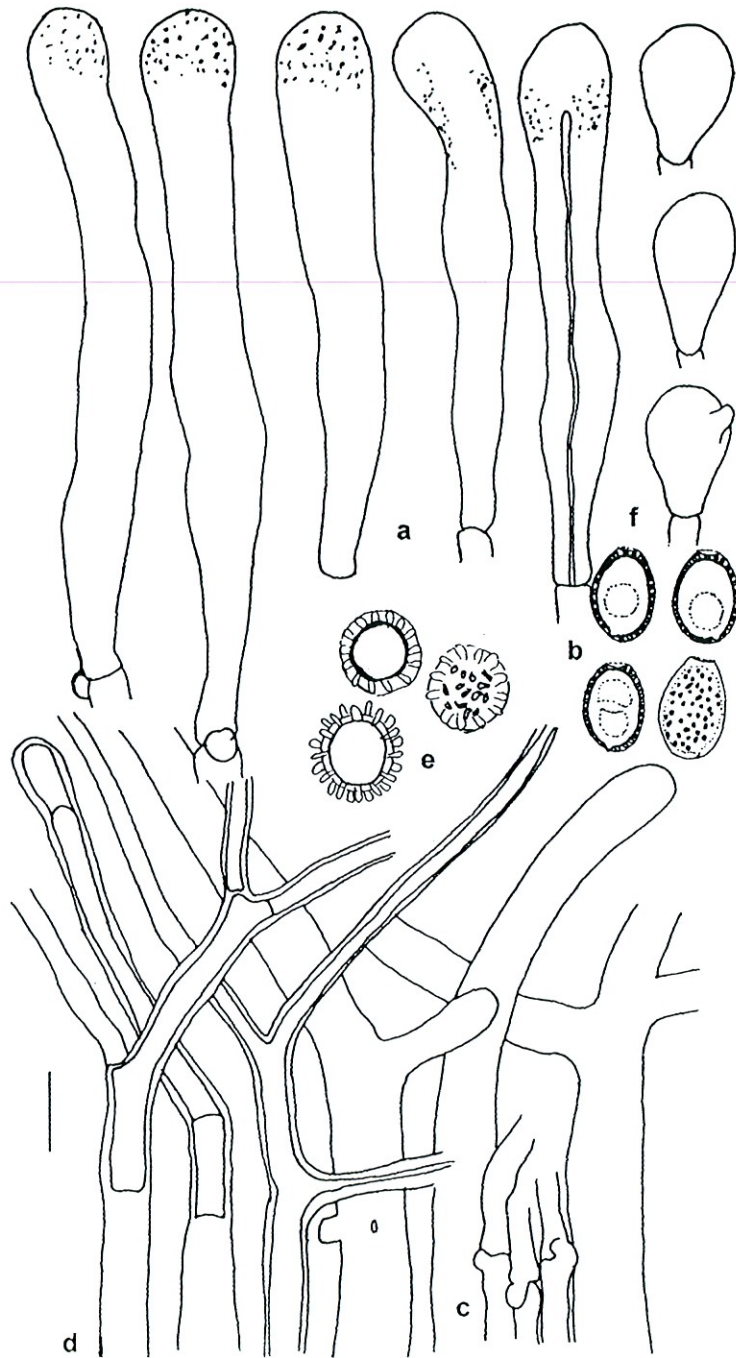




**Fig. 13.** Micromorphological features of *Ganoderma sessiliforme*: a. Cuticle cells. b. Basidiospores. c. Hyphal system of crustohymenodermis: Skeletal hyphae. d. Chlamydospores. Bar = 8  $\mu$ m.



**Fig. 14.** Micromorphological features of *Ganoderma subformicatum*: a. Cuticle cells. b. Basidiospores. c–e. Hyphal system of crustohymenodermis: c. Generative hyphae. d. Skeletal hyphae. e. Binding hyphae. Bar = 8  $\mu$ m.



**Fig. 15.** Micromorphological features of *Ganoderma weberianum*: a. Cuticle cells. b. Basidiospores. c-d. Hyphal system of crustohymenodermis: c. Generative hyphae. d. Skeletal hyphae. e. Chlamydospores. f. Basidium and basidiola. Bar = 8  $\mu$ m.

## CAPÍTULO I, PARTE E

### MYCOTAXON

#### Taxonomic status and new localities for *Ganoderma ravenelii*

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**Abstract**—The taxonomic status of *Ganoderma ravenelii* is discussed; furthermore the species is recorded from two new localities in the USA. A full description and illustrations of the species are made.

**Key words**—*Ganoderma curtisii*, Texas, North Carolina, USA, basidiospores, resinous bands

#### Introduction

Based in the absence of resinous bands in the context and the shape and size of basidiospores, Steyaert (1980) described *Ganoderma ravenelii* from a collection proposed previously by him as the neotype of *G. curtisii* (Berk.) Murrill. According with Steyaert (1980) this material did not fully correspond to Berkeley's description and he selected another neotype for *G. curtisii*. In the original description of *G. ravenelii*, two collections were included: one from South Carolina and the other from Florida. Steyaert (1980) was doubtful about the rank, as species or as variety, of the name he was proposing, until more specimens would be studied. Since its description, this species have not been newly recorded, probably because it was erroneously determined as *G. curtisii* or as another species. It is interesting that Gilbertson and Ryvarden (1986) and Adaskaveg & Gilbertson (1988) did not mention *G. ravenelii*, neither *G. curtisii*, for the North America polypores. According with Moncalvo & Ryvarden (1997) the taxonomic status of these two species remained unclear. In a study on *G. curtisii* in Mexico, where the type of *G. ravenelii* was studied, Torres-Torres et al. (2005) suggested the presence of *G. ravenelii* in Mexico; but it was not established with confidence because the available specimen was too young. In occasion to a visit to the Field Museum herbarium, materials deposited as *G. lucidum* or *Ganoderma* sp. agreeing with the Steyaert's description were found and studied in detail. In addition, specimens of *G. curtisii* were studied in order to compare the two species. The features used by Steyaert (1980) for delimitation of the two species were verified and we concluded that this is a good species. A full description and illustrations of the species are made, including features not considered by Steyaert. On the other hand, the species is recorded by the second time in the world from two new localities also in the USA.

#### Materials and Methods

All materials studied were checked in the Field Museum Herbarium (F), except the holotype, which was requested to K. Herbaria abbreviations follow Holmgren et al. (1990). The key colors are from Komerup & Wanscher (1963). Micromorphologic observations were made from material mounted in 5% KOH. The basidiospores shape was determined according to Q coefficient (length-width, Bas 1969) from at least 20 randomly selected basidiospores from each collection. The draws of the microscopic structures were made with a 100x oil-immersion objective, in a Zeiss K7 and a Zeiss Axioscop 40 microscopes.

## Taxonomy

*Ganoderma ravenelii* (Berk.) Murrill

Figs. 1-6

Bulletin du Jardin Botanique National de Belgique 50(1-2): 146, 1980.

**Basidiomata** 4.8–6.7 × 3.9–4.5 × 1–1.5 cm, annual, stipitate, single, sometimes imbricate, corky to woody. Pileus rounded-flabelliform, reniform to circular, upper surface slightly convex to plane; surface glabrous, smooth to slightly tuberculate, soft, semi-dull, occasionally shiny all over the surface, concentrically sulcate; with a laccate crust, easy to penetrate with fingernail, generally easily removed; maize (4A6), deep yellow (4A8), golden-yellow (5B8) to yellowish-brown (5C8), more or less homogeneous, or with zonations of these tonalities, occasionally covered by cinnamon (6D6) basidiospores; margin concolorous, entire, thick, rounded to truncate, smooth. **Stipe** 1–10 × 1.1–1.6 cm, eccentric to central, cylindrical to flattened, solid; surface smooth to tuberculate, dull to shiny, with a laccate crust, easy to remove or not, red-wine almost black, generally darker than pileus.

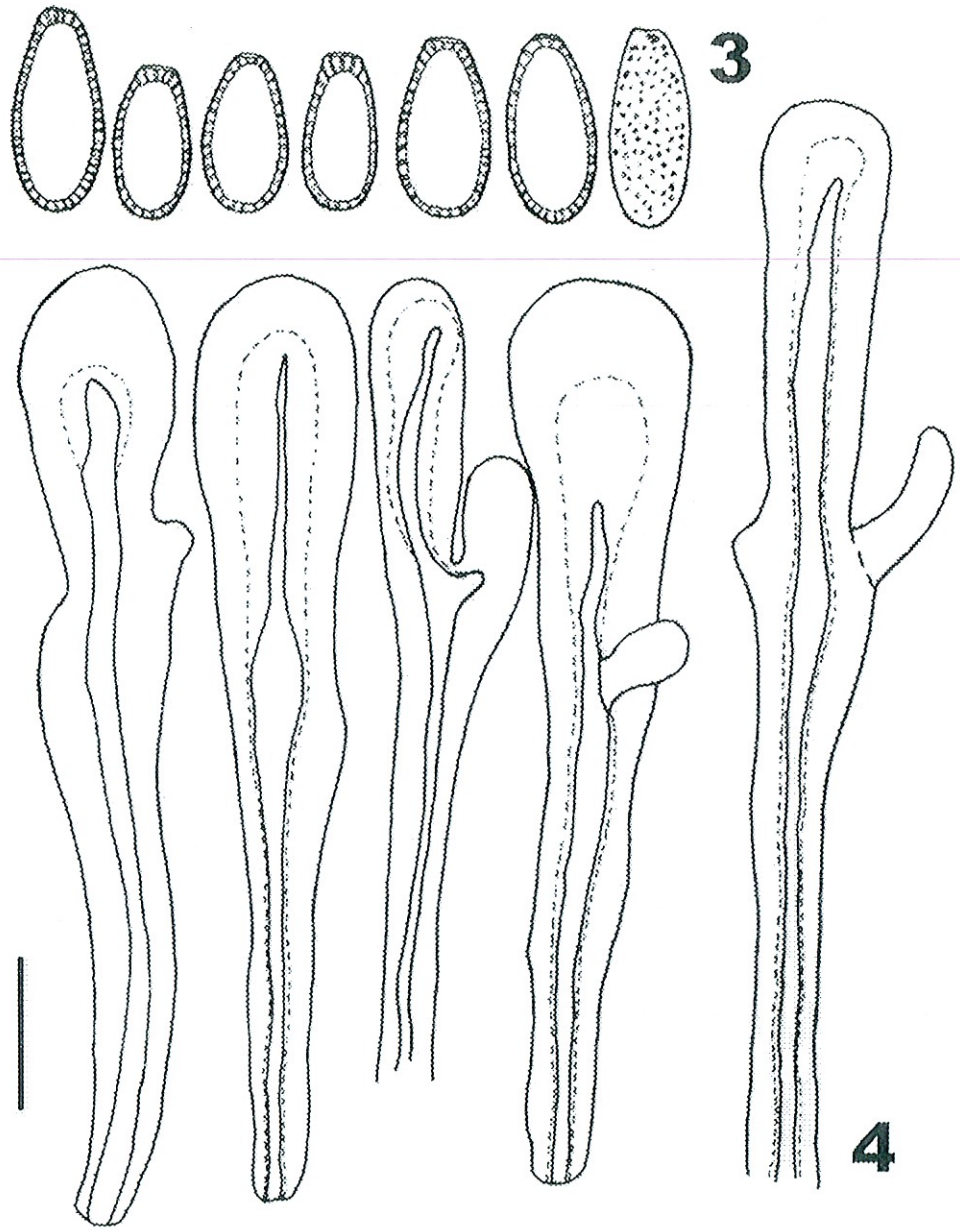
**Context** 0.7–1.3 cm thick, corky to slightly fibrous, duplex, azonate, pale orange to light orange (5A3, 5A4), brown-sienna (6D7) close to the tubes, without resinous bands. *Pores* 2–3 per mm, angular to rounded; pore surface yellow (2A3) to pale yellow (3A3), darkening to brown (6D8) when bruising or aging; tubes 0.5–1 cm long, unstratified, pale vinaceous-brown to vinaceous-brown (8E4). **Hyphal system** di-trimitic. **Contextual trama** with generative hyphae no observed; skeletal hyphae 3.2–8.8 µm diam., thick-walled to solid, non-septate, arboriform, yellowish to yellowish-brown, predominant; binding hyphae 5.6–8.8 µm diam., thick-walled to solid, non-septate, yellowish. **Hymenophoral trama** as contextual trama. **Pileipellis** with cuticle cells 56–80 × 7.2–16 µm, broadly clavate, generally entire or with one lateral protuberance, thick-walled to solid, generally multistratified, yellowish, amyloid in Melzer's reagent; generative hyphae up to 4 µm diam., thin-walled, with conspicuous clamps, hyaline, difficult to observe; skeletal hyphae 4–6.4 µm diam., thick-walled to solid, non-septate, arboriform, yellowish to almost hyaline, scarce; binding hyphae 2–8 µm diam., thick-walled to solid, non-septate, yellowish to yellowish-brown. **Basidiospores** (10.2–) 10.8–15.2 × 5.2–7.2 µm, Q = 1.78–2.72, oblong to cylindrical, apex truncate, with apical germ pore, yellowish-brown; perisporium wrinkled, hyaline to yellowish-red; exosporium with inter-walled pillars up to 0.4 µm thick, free. **Basidia** no observed. **Cystidia** absent.

**Specimens examined.** USA, South Carolina, Aiken, on the ground, s. data, H.W. Ravenel 2936 (K. HOLOTYPE). Florida, Columbia, Camp O'Lena State Park, s. data, H.S. Dybas s.n. (F). North Carolina, Columbus Co., Reaves Ferry, on tree roots, 18 Oct 1934, W.C. Coker s.n. (F). Texas, Hardin, Larsen Preserve, 21 Dec 1982, D. P. Lewis 3428 (F).

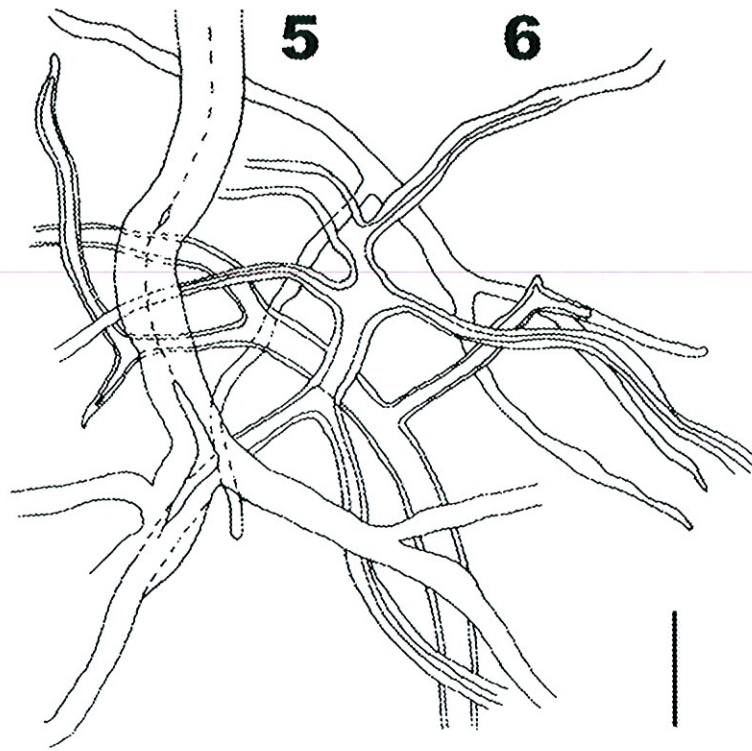
**Specimens of *Ganoderma curtisii* examined.** USA, North Carolina, Ardmore Woods, on old oak stump, 10 Aug 1932, P.O. Schallert s.n.; Columbus, Reaves Ferry, on tree roots, 18 Oct 1934, W.C. Coker s.n.; Forsyth, Winston-Salem, in yard, 30 Sep 1934, P.O. Schallert s.n.; Jackson, Whiteside Mt., 12 Jun 1934, P.O. Schallert s.n.; Durham, Durham Duke University Forest, on a stump, Aug 1949, W.L. Culberson s.n. Georgia, Thomas, Thomasville, 19 Jul 1947, H. Field s.n. Louisiana, Calcasieu, Lake Prien, on the ground in pine woods, 30 Oct 1948, F. Drouet s.n. Texas, Hardin, Big Thicket National Preserve, Lance Rosier Unit Cotton Road, on wood, 15 Jun 1983, D.P. Lewis 3515 (all in F).



Figs. 1-2. Macroscopic features of *Ganoderma ravenelii* (D.P. Lewis 3428). 1: pileus, 2: context without resinous bands and tubes. Scale bar = 1 cm.



Figs. 3-4. Microscopic features of the holotype of *Ganoderma ravenelii*. 3: basidiospores, 4: cuticle cells. Scale bar = 8  $\mu$ m.



Figs. 5-6. Microscopic features of the holotype of *Ganoderma ravenelii*. 5: skeletal hyphae, 6: binding hyphae. Scale bar = 8  $\mu$ m.

### Discussion

Macromorphologically the species is closely related with *G. curtisii* (see table 1). Besides the features (absence of resinous bands in the context and the shape and size of basidiospores) included by Steyaert (1980) to separate *G. ravenelii* from *G. curtisii*, we analyzed other important features: union of the stipe, basidiospore pillars and cuticle cells. So we consider that there is enough evidence to treat *G. ravenelii* as an independent species from *G. curtisii*, and not as a variety of it. *Ganoderma curtisii* presents resinous bands in the context, basidiospores of (9.2–) 10.4–12 (–13.6)  $\times$  5.6–8  $\mu$ m, ellipsoid to oblong, basidiomata with lateral or occasionally eccentric stipe, basidiospore pillars slightly thicker and subfree, and cuticle cells occasionally lateral or apically branched. Steyaert (1980) described for *G. ravenelii* cuticle cells of 20  $\times$  8–10  $\mu$ m, surely a mistake, because his figure 7 shows other proportion. Both species can occur in the same geographic area, but the morphological features permit to distinguish them as separate species. The majority of the specimens of *G. ravenelii* and of *G. curtisii* deposited in F were determined as *G. lucidum* (Curtis) P. Karst. Steyaert (1980) recorded *G. ravenelii* from South Carolina and Florida; this is the second record of the species from USA, from North Carolina and Texas.



Table 1. Morphologic features of *Ganoderma curtisii* and *G. ravenelii*.

Species	<i>G. curtisii</i>	<i>G. ravenelii</i>
Stipe	generally lateral	generally eccentric to central
Context	duplex	duplex
Resinous bands	present	absent
Basidiospores size	(9.2-) 10.4-12 (-13.6) × 5.6-8 µm	(10.2-) 10.8-15.2 × 5.2-7.2 µm
Basidiospores form	Q = 1.5-1.88, ellipsoid to occasionally oblong	Q = 1.78-2.72, oblong to cylindrical
Basidiospores pillars	subfree	free
Cuticle cells	clavate, without or with two to three apical or lateral protuberances, occasionally lateral or apical branched	clavate, generally entire or with one lateral protuberance

#### Acknowledgments

The authors thanks to Adriana de Mello Gugliotta and Cony Decock for their critical review and their valuable comments on this paper. Thanks are due to the curator of K herbarium for the loans of the type for the study. To Miguel de Santiago for inking the draws. M.G Torres-Torres especially thanks to Gregory Mueller and Betty Strack by their assistance and kind hospitality during her stay in the Botanical section of the Field Museum. The first author also gratefully acknowledges the facilities and financial support received from project NUFFIC-Altterra of Wageningen University, and Universidad Tecnológica del Chocó. Funds also were obtained from CONACYT (project CONACYT-SEP-2003-C02-42957) and Universidad de Guadalajara (project 62935).

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## CAPÍTULO I, PARTE F

### A TAXONOMIC REVIEW OF THE SPECIES CONSIDERED AS SYNONYMS OF *GANODERMA RESINACEUM*

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**Abstract.** Morphologic studies of eleven types of the species considered as synonyms of *Ganoderma resinaceum* were made. Modern features were considered to make a re-description of the species. The color of the context, basidiospore pillars and cuticle cells resulted to be the most important characters in the determination of the species.

**Key words.** Complex, morphology, basidiospores, cuticle cells

#### Introduction

The circumscription of the taxa in *Ganoderma* has resulted very often too complicated. In the past many different names were erected for the same species, or, on the other hand, many different species were grouped under the same name. One example for the last case is *Ganoderma resinaceum* Boud. Steyaert (1980) treated it as a complex and considered the size of the basidiospores as the most important feature to group the species. So, he synonymized *G. argillaceum* Murrill, *G. chaffangeonii* Pat., *G. sessile* Murrill, *G. polychromum* (Copel.) Murrill, *G. praelongum* Murrill and *G. subperforatum* G.F. Atk. under *G. resinaceum*. Bazzalo and Wright (1982) accepted these synonyms and besides suggested *G. nitidum* Murrill also as a synonym. Subsequently more synonyms were included by Ryvar den (1985, 2000, 2004) under *G. resinaceum*: *G. areolatum* Murrill, *G. perturbatum* (Lloyd) Torrend, *G. pulverulentum* Murrill, *G. sessiliforme* Murrill, *G. subincrustatum* Murrill, *G. subfornicatum* Murrill, *G. subtuberculosum* Murrill and *G. triviale* Bres. This wide species concept includes organisms with very variable features: structure and colour of the context, size and disposition of the basidiospore pillars, and shape of cuticle cells. Gottlieb & Bazzalo (1999) studied some species of the complex with modern methods and did not accept all the synonymies; nevertheless they did not make full descriptions either illustration of the species. There are no taxonomic reviews that consider modern features for the descriptions of the species of the *G. resinaceum sensu* Steyaert (*op. cit.*), and in many cases there are not descriptions of the species.

With the objective to have a better comprehension of the complex *G. resinaceum sensu* Steyaert, a taxonomic review of the species and modern descriptions including illustrations of each species were made; which will permit us a critical analysis about the taxonomic status of the species. On the other hand, it will also contribute to the knowledge of the geographical distribution of species

poorly known. The type materials of eleven species of the *G. resinaceum* complex were macro and micromorphological checked. The materials included in this study were: *G. argillaceum*, *G. chaffangeonii*, *G. nitidum*, *G. perturbatum*, *G. praelongum*, *G. pulverulentum*, *G. sessile*, *G. sessiliforme*, *G. subincrustatum* and *G. subformicatum*. The type of *G. resinaceum* was not studied because the specimen was not found in PC, nevertheless an authentic specimen was studied and comparisons with description of Gottlieb & Bazzalo (1999) were done. *Ganoderma areolatum* was reviewed but was not included in this paper, because it resulted to be *Navisporus floccosus* (Bres.) Ryvarden (Torres-Torres *et al.*, 2007). From the taxa studied, *G. chaffangeonii* is the unique species that resulted to be a synonym of *G. resinaceum*. The consistency of the basidiomata, presence of resinous deposit in the context, structure, colour and consistency of the context, basidiospores apex, thickness and disposition of the pillars in the basidiospores, and shape of the cuticle cells were considered. We concluded that morphological characters are important aspect to consider in the delimitation of the taxa.

## Materials and Methods

**Specimens studied.** The materials studied were requested to the herbaria BPI, NY and PC. Herbaria abbreviations follow Holmgren *et al.* (1990).

**Macro and micromorphological observations.** Adhesion to the substrate, shape, consistency, width and length; pileus colour; tubes stratification, length, color, pores per mm, pore surface color; context stratification, width and color were the macromorphological features observed for description of the basidiomata. The fruit bodies were compared with the color reference of Kornerup & Wanscher (1963). Micromorphologic observations were made from material mounted in 10% KOH and Melzer's reagent; furthermore other colorants as Congo red, floxine and cotton blue were used. The basidiospore shape was determined according to Q (length-width, Bas. 1969) of 20 randomly selected but mature spores. The description of the pileipellis was done following Cléménçon (2004). The microscopically structures draws were made with a 100x oil-immersion objective, in a Zeiss K7 and Zeiss Axioscop 40 microscop.

## KEY TO THE SPECIES

1. Context duplex; cuticle cells entire .....2
  1. Context relatively homogeneous to homogeneous; cuticle cells entire or diverticulate.....3
  2. Pileus sessile, resinous bands almost to the periphery; basidiospores 12-16 x 8-10  $\mu$ m ..... *G. sessile*
  2. Pileus stipitate, without resinous bands; basidiospores 9-12 x 6-8  $\mu$ m..... *G. praelongum*
3. Brown context, relatively homogenous to homogenous; cuticle cells entire to diverticulate.....4
  3. Pale context, relatively homogenous; cuticle cells entire, without apex granulations.....8
4. Basidiomata stipitate, at times substipitate when growing on wood, basidiospores with subacute apex, pillars thick and partially anastomosed..... *G. perturbatum*

4. Basidiomata sessile, substipitate or with a short stipe, basidiospores distinctively truncate, pillars thick to thin, subfree to partially anastomosed.....5
5. Cuticle cells entire or with few protuberances.....6
  5. Cuticle cells with protuberances.....7
6. Basidiomata generally with a contracted base, concave to infundibuliform, woody; context with resinous incrustations close to the base, relatively homogeneous; basidiospores 9-12 x 6-8  $\mu\text{m}$ , pillars partially anastomosed.....*G. subincrustatum*
  6. Basidiomata generally sessile, plane to convex, spongy-corky; context without resinous incrustations, light reddish-brown homogeneous, basidiospores 11-14 x 6-8  $\mu\text{m}$ , pillars subfree..... *G. resinaceum*
7. Cuticle cells with the clavate shape conserved, generally with one to three short and thick protuberances and/or branches ..... *G. nitidum*
  7. Cuticle cells not claviform, commonly with a constriction, one to two thick and long branches, up to seven short and thick protuberances.....*G. subfornicatum*
8. Basidiospores 9-11 x 6-8  $\mu\text{m}$ , pillars partially anastomosed..... *G. sessiliforme*
  8. Basidiospores 10-13 x 7-10  $\mu\text{m}$ , pillars subfree..... *G. argillaceum*

## Taxonomy

Descriptions of the type specimens are presented, indicating in bold the valid name.

*Ganoderma argillaceum* Murrill, North Amer. Flora 9: 122, 1908. Fig. 1

**Basidiomata** 5-7.5 x 8-11 x 1.5-4 cm, annual, substipitate, single, woody-corky, light in weight. **Pileus** rounded flabelliform to circular; surface glabrous, bumpy, smooth to radially rugose, soft, glossy, without concentric sulcate zonation; with a laccate crust, not cracking, difficult to remove, easy to penetrate with fingernail; reddish-brown or photo-brown (9F8) in the 80 to 90% of the surface to henna (7E8) in the periphery, without basidiospores over the surface; margin whitish, entire, thin to rounded in some specimens, smooth. **Substipe** 1-2 x 1.5 mm, lateral, cylindrical, solid, surface shiny, red-wine to almost black, darker than pileus. **Context** up to 3 cm thick in the base, 2 cm average, fibrous-corky, zonate, relatively homogeneous, pale-orange to light-orange (5A3, 5A4) above and reddish-golden to light brown (6C7) close to the tubes; resinous bands only close to the base. **Pores** 3-4 per mm, angular to circular, woody; pore surface yellowish-white (3A2), darkening to brown (6D8) when bruising or aging; tubes 0.5-1 cm thick, up to 1 cm in the base, unstratified, generally concolorous with the inferior part of the context. **Hyphal system** trimitic. **Contextual trama** with generative hyphae not observed; skeletal hyphae 4-8  $\mu\text{m}$  diam., generally solid to thick-walled, non-septate, arboriform, very branched, yellowish to golden-yellow, predominant; binding hyphae 3.2-5.6  $\mu\text{m}$  diam., solid, non-septate, yellowish to light yellowish-brown. **Hymenophoral trama** as contextual trama. **Pileipellis** with cuticle cells 45.6-68 x 7.2-12  $\mu\text{m}$ , clavate, entire or with one lateral protuberance; thick-walled, generally multistratified, golden-yellow, content immediately black with Melzer's reagent, cells amyloid immediately or after 48 h; generative hyphae not observed; skeletal hyphae 4-8  $\mu\text{m}$  diam., generally solid to thick-walled, non-

septate, arboriform, very branched, yellowish to golden-yellow, predominant; binding hyphae 3.2-5.6  $\mu\text{m}$  diam., solid, non-septate, yellowish to light yellowish-brown, difficult to differentiate from the skeletal hyphae. **Basidiospores** 10.4-13.6 x 7.2-9.6  $\mu\text{m}$ , Q = 1.33-1.6, ellipsoid, apex truncate, with apical germ pore, light yellowish-brown, negative in Melzer's reagent; perisporium wrinkled, hyaline; exosporium with inter-walled pillars up to 0.4  $\mu\text{m}$  thick, subfree; endosporium wrinkled, frequently with smooth basidiospores smaller. **Basidia** not seen. **Cystidia** absent.

*Specimens examined.* CUBA, Province of La Habana, Santiago de las Vegas, on dead mango log, 5 July 1904, F.S. Earle 658 (NY, Lectotype).

*Ganoderma chaffangeonii* Pat., Bull. Soc. Mycol. Fr. 5: 74, 1889. Fig. 2  
= *Ganoderma resinaceum* Boud.

**Basidiomata** only fragments remain, up to 2 cm thick in the base, annual, single, woody-spongy. **Pileus** convex to plane; surface glabrous, slightly bumpy, slightly soft, glossy, broadly zonated; with a laccate crust, cracked, easy to remove and to penetrate with fingernail; violet-brown (11F4), in all the surface; margin concolorous, slightly lobulate, rounded, wrinkle. **Context** 0.6-0.8 cm thick, fibrous-spongy, slightly zonate, homogeneous, light reddish-brown, with an apricot (5B6) thin fringe below the laccate crust, without resinous bands. **Pores** 4-5 per mm, angular to rounded, woody; pore surface yellow (3A2) when fresh, darkening to ochraceous or yellowish-brown (6C5) when aging or drying; tubes 1-1.5 cm long, unstratified, concolorous with the context. **Hyphal system** dimitic. **Contextual trama** with generative hyphae not observed; skeletal hyphae 1.6-8  $\mu\text{m}$  diam., thick-walled, generally solid, non-septate, non-branched to arboriform, moderately branched, golden-yellow. **Hymenophoral trama** as the contextual trama. **Pileipellis** with cuticle cells 46.5-71.3 x 8.6-13.2  $\mu\text{m}$  diam, clavate, almost cylindrical to occasionally widely clavate, generally entire, some with occasional lateral or apical protuberances and branches, thick-walled to solid, apex with granulations, brownish-yellow, amyloid in Melzer's reagent; generative hyphae 2.4-3.1  $\mu\text{m}$  diam., thin-walled, with conspicuous clamps, hyaline to yellowish, abundant; skeletal hyphae 3.1-6.2  $\mu\text{m}$  diam., thick-walled to occasionally solid, non-septate, sometimes arboriform, yellowish-brown. **Basidiospores** 11.2-13.2 (-13.6) x 7.2-8.8 (-9.2)  $\mu\text{m}$ , Q = 1.43-1.6, ellipsoid, apex truncate, with visible apical germ pore, yellowish-brown; perisporium smooth, hyaline; exosporium with inter-walled pillars up to 0.4  $\mu\text{m}$  thick, free; endosporium wrinkled. **Basidia** not observed. **Cystidia** absent.

VENEZUELA, Orinoco, on dead trunk, s.date, *Chaffangeon* 1885 (PC, Lectotype of *Ganoderma chaffangeonii*).

*Ganoderma nitidum* Murrill, North American Flora 9: 123; 1908. Fig. 3  
= *Polyporus nitidum* (Murrill) Overh., Botany of Porto Rico and the Virgin Islands: Mycology: 164 (1926).

**Basidiomata** 2.5-9.5 x 3.2-18 x 1.5-3 cm, perenne, sessile to substipitate, with a contracted base, fully imbricate, woody but light in weight. **Pileus** rounded-flabelliform to reniform, convex to generally plane; surface glabrous, bumpy, hard but easy to penetrate with fingernail, very shiny, remarkable concentrically sulcate; with a laccate crust, difficult to remove; totally reddish-brown almost black (darker than 9F8), homogenous, generally covered with cinnamon (6D6) basidiospores; margin oxide-red (8E8), entire to slightly lobulate, thick, obtuse to truncate, sulcate. **Substipe** 1-1.4 x 1.5-3.3 cm, short and thick, cylindrical, horizontal, concolorous to the pileus, solid. **Context** 0.2-0.5 cm thick, up to 1.8 cm thick in the base, fibrous, relatively homogeneous, azonate, raw-sienna or rust-brown (6E7), gradually changing to dark-brown (7F8) next to the tubes; without resinous bands, a specimen with resinous incrustations only close to base. **Pores** 6-7 per mm, circular, woody; pore surface orange-white (5A2), slightly darkening to brown (6D8) when bruising; tubes 1.5-2 cm long, indistinguishable stratified, brown, lighter than context. **Hyphal system** trimitic. **Contextual trama** with generative hyphae 1.5-4.8  $\mu\text{m}$  diam., thin-walled, with large and conspicuous clamps, branched, hyaline, scarce and difficult to observe; skeletal hyphae 4.8-9.4  $\mu\text{m}$  diam., generally thick-walled to solid, occasionally septate, arboriform or not, golden-yellow to yellowish-brown, predominant; binding hyphae 1.6-4.8  $\mu\text{m}$  diam., solid, non-septate, hyaline to yellowish, scarce, difficult to observe. **Hymenophoral trama** as contextual trama. **Pileipellis** with cuticle cells 48-96 x 5.6-12.8  $\mu\text{m}$ , clavate, without or up to two protuberances, and/or short and thick branches, mainly solid to thick-walled, multistratified wall, golden yellow to yellowish-brown in group, slightly greyish apex with Melzer's reagent; generative hyphae 1.6-4.8  $\mu\text{m}$  diam., thin-walled, conspicuous clamps, hyaline, abundant, difficult to observe; skeletal hyphae 4-9.6  $\mu\text{m}$  diam., generally thick-walled to solid, apex septate, arboriform, dark golden-yellow. **Basidiospores** 9.6-12.8 (-13.6) x 6.4-7.6 (-8)  $\mu\text{m}$ , Q = 1.5-1.88, ellipsoid to oblong, apex slightly truncate, apical germ pore, yellowish; perisporium semi-wrinkled, exosporium with inter-walled pillars up to 0.5  $\mu\text{m}$  thick, partially anastomosed; endosporium semi-wrinkled. **Basidia** not seen. **Cystidia** absent.

*Specimens examined.* HONDURAS, Puerto Sierra, Rio Esperanza, 28 February 1903, P. Wilson 607 (NY, Lectotype; fragment in BPI).

***Ganoderma perturbatum*** (Lloyd) Torrend, Broteria Bot. 18: 34, 1920. Fig. 4  
≡ *Polyporus perturbatus* Lloyd, Mycol. Writ. 5, Let. 68: 11, 1918.

**Basidiomata** 5.5 x 4.5 x 1.1 cm, perenne, stipitate, single, never imbricate, corky to woody. **Pileus** reniform, convex to plane; surface glabrous, bumpy, hard but possible to penetrate with fingernail, mainly very shiny although dull regions might be observed, generally with abundant radial zonation; with a laccate crust, difficult to remove; totally violet-brown almost black (darker than 11F7 or 11F8), homogenous, occasionally covered with cinnamon (6D6) basidiospores; margin generally lighter than pileus, oxide-red (8E8), entire, thick, obtuse to truncate, sulcate. **Stipe** 13 x 1.1 cm, lateral, cylindrical, solid, surface shiny, violet-brown (11F8) very dark to black, concolorous to generally darker than pileus. **Context** 0.3-0.5 cm thick, fibrous, relatively homogeneous, azonate, raw-sienna or light brown (6D7), gradually changing to slightly darker next to the tubes; resinous incrustations not very visible. **Pores** 3-4 per mm, angular to rounded, woody; pore

surface yellowish-white (3A2) to pale yellow (4A3), darkening to brown (6D8) when bruising or aging; tubes 0.4-0.6 cm long, indistinguishable stratified, concolorous with context. *Hyphal system* dimitic. *Contextual trama* with generative hyphae not observe; skeletal hyphae 2.4-7.2  $\mu\text{m}$  diam., generally solid to thick-walled, non-septate, arboriform, yellowish to yellowish-brown. *Hymenophoral trama* as contextual trama. *Pileipellis* with cuticle cells 48.8-88 x 7.2-20  $\mu\text{m}$ , broadly clavate to occasionally conic, without or up to two diverticules, mainly solid to thick-walled, multistratified wall, generally with a distinctive darker inner stratum, with refringent content, yellow to yellowish-brown in group, content immediately black with Melzer's reagent, walls slightly amyloid after 14 h; generative hyphae not observe; skeletal hyphae 2.4-7.2  $\mu\text{m}$  diam., generally thick-walled to solid, apex septate, arboriform, yellowish-brown. *Basidiospores* 11.2-12.8 x 8-9.6  $\mu\text{m}$ , Q = 1.25-1.45, broadly ellipsoid to ellipsoid, apex subacute, slightly visible apical germ pore, yellowish-brown; perisporium wrinkled, exosporium with inter-walled pillars 0.7-0.8  $\mu\text{m}$  thick, partially anastomosed; endosporium wrinkled. *Basidia* not seen. *Cystidia* absent.

*Specimens examined.* BRAZIL, Region Grande do Sul, Lageado, s.date, J. Rev Rick, s.n., Lloyd herb. num. 55740 (BPI. Lectotype of *Ganoderma perturbatum*).

*Ganoderma praelongum* Murrill, North Amer. Flora 9: 121, 1908. Fig. 5

*Basidiomata* 5-6 x 4.5 x 1.5 cm, 2.5 cm thick in the base, annual, stipitate, single, woody-corky, light in weight. *Pileus* rounded flabelliform, plane; surface glabrous, bumpy, slightly radially rugose, soft-corky, glossy, concentrically sulcate mainly toward the margin; with a laccate crust, not cracking, generally difficult to remove, easy to penetrate with fingernail; violet-brown (11F6, 11F6) in the 80 to 90% of the surface, in some part violet-brown very dark almost black, henna (7E8) in the periphery, without basidiospores over the surface; margin slightly lighter than pileus, lobulate, thin, smooth. *Stipe* 7-8 x 1-1.7 cm, lateral, cylindrical, solid, surface shiny, violet-brown (11F8) very dark, generally darker than pileus. *Context* up to 2 cm thick in the base, 0.7-1 cm average, fibrous-corky, azonate, duplex, ochre-yellow (6D7) above and rust brown (6E8) close to the tubes; without resinous bands. *Pores* 2-3 per mm, angular to rounded, woody; pore surface pale-yellowish (3A3), darkening to brown (6D8) when bruising or aging; tubes 0.8-1 cm thick, up to 1.4 cm in the base, unstratified, generally concolorous with the inferior part of the context. *Hyphal system* dimitic. *Contextual trama* with generative not observe; skeletal hyphae 2.8-12.8  $\mu\text{m}$  diam., generally solid, non-septate, arboriform or not, very branched, yellowish to golden-yellow, predominant; binding hyphae 1.6-4  $\mu\text{m}$  diam., solid, non-septate, hyaline to yellowish, scarce. *Hymenophoral trama* as contextual trama. *Pileipellis* with cuticle cells 41.6-93.6 x 6.4-17.6  $\mu\text{m}$ , narrowly clavate and clavate, generally entire or with one lateral protuberance and/or branches; thick-walled, generally multistratified, apex with granulations, golden-yellow to yellowish-brown, with cells slightly amyloid in Melzer's reagent; generative hyphae thin-walled, with conspicuous clamps, hyaline, difficult to measure; skeletal hyphae 4-9.6  $\mu\text{m}$  diam., generally solid, at time septate, arboriform, very branched, yellowish-brown, predominant. *Basidiospores* 9.6-12 x 6.4-8 (-8.4)  $\mu\text{m}$ , Q = 1.38-1.62, ellipsoid, slightly apex truncate, with apical germ pore, yellowish-brown, negative in Melzer's reagent; perisporium semi-wrinkled, reddish-yellow;

exosporium with inter-walled pillars  $<0.4 \mu\text{m}$  thick, partially anastomosed; endosporium smooth. **Basidia** not seen. **Cystidia** absent.

*Specimens examined.* CUBA, Provincia de Santiago de Cuba, Alto Cedro; under big stump in open field or clearing, 19-20 March 1905, *F.S. Earle & L. Murrill* 536 (NY, Lectotype, fragment in BPI).

*Ganoderma pulverulentum* Murrill, North Amer. Flora 92: 121, 1908. Fig. 6

**Basidiomata** 12-25 x 4.5 x 1.5 x 2.5-6 cm, annual, sessile to substipitate, generally imbricate, woody-corky, light in weight. **Pileus** rounded flabelliform to reniform, plane; surface glabrous, bumpy, soft-corky, semiglossy, concentrically sulcate mainly; with a laccate crust, not cracking, generally difficult to remove, easy to penetrate with fingernail; reddish-brown (8F8) to photo-brown (9F8) in the 90% of the surface, in some part violet-brown very dark almost black, henna (7E8) to oxid red (8E8) in the periphery, with basidiospores over the surface; margin whitish, lobulate, thin, smooth. **Stipe** 2.5-3 x 2.5-3 cm, lateral, cylindrical, solid, surface shiny, reddish-black, very dark, darker than pileus. **Context** 0.9-2.5 cm, fibrous-corky, zonate, relatively homogenous, ochre-yellow (5B6) above and dark brown (7F7) close to the tubes; with discontinuous resinous bands. **Pores** 3-4 per mm, angular to rounded, woody; pore surface cream to pale-yellowish (3A3), apparently do not darkening when bruising or aging; tubes 1.2-2.7 cm thick, up to 4 cm in the base, stratified, generally concolorous with the inferior part of the context. **Hyphal system** dimitic. **Contextual trama** with generative not observe; skeletal hyphae 2.8-12.8  $\mu\text{m}$  diam., generally solid, non-septate, arboriform or not, very branched, yellowish to golden-yellow, predominant; binding hyphae 1.6-4  $\mu\text{m}$  diam., solid, non-septate, hyaline to yellowish, scarce. **Hymenophoral trama** as contextual trama. **Pileipellis** with cuticle cells 40-64 x 8-11.2  $\mu\text{m}$ , narrowly clavate and clavate, generally without or with one lateral protuberance and/or branches; thick-walled, generally multistratified, apex with granulations, golden-yellow to yellowish-brown, with cells slightly amyloid in Melzer's reagent; generative hyphae 4-4.8  $\mu\text{m}$ , thin-walled, with conspicuous clamps, hyaline; skeletal hyphae 3.2-5.6  $\mu\text{m}$  diam., generally thick-walled, at time septate, arboriform, very branched, yellowish-brown, predominant. **Basidiospores** 10.4-12.4 x 6.4-8  $\mu\text{m}$ ,  $Q = 1.44-1.62$ , ellipsoid, apex truncate, with apical germ pore, yellowish-brown, negative in Melzer's reagent; perisporium semi-wrinkled, reddish-yellow; exosporium with inter-walled pillars up to 0.4  $\mu\text{m}$  thick, partially anastomosed; endosporium smooth. **Basidia** not seen. **Cystidia** 20-20.8 x 4-5.6  $\mu\text{m}$ , utriform, hyaline, thin-walled.

*Specimens examined.* GRENADA ISLAND, West Indies; on dry manchineel, 4 Sept 1905, *W.E. Brodway s.n.* (NY, Lectotype)

*Ganoderma resinaceum* Boud., Bull. Soc. Mycol. Fr. 5: 72, 1889. Fig. 7  
= *Ganoderma chaffangeonii* Pat., Bull. Soc. Mycol. Fr. 5: 74, 1889.

**Basidiomata** 9-12.5 x 7-10 cm, up to 2 cm thick in the base, annual, single to imbricate, woody-spongy. **Pileus** rounded-flabelliform, convex to plane; surface glabrous, smooth to slightly bumpy, slightly soft, glossy, broadly zonated;



with a laccate crust, cracked, easy to remove and to penetrate with fingernail; violet-brown (11F7), darker than 11F7 to dark red in almost all the surface, gradually changing to orange (5A7) toward the margin or fully violet-brown (11F7), darker in adult basidiomata, with terra-cotta (7D7) basidiospores over the surface; margin brownish-orange to concolorous, entire, thin to rounded, smooth. **Context** 0.4-1 cm thick, fibrous-spongy, azonate, homogeneous, light reddish-brown, with an apricot (5B6) thin fringe below the laccate crust, without resinous bands. **Pores** 3-5 per mm, angular to rounded, woody; pore surface yellow (3A2) when fresh, darkening to ochraceous or yellowish-brown (6C5) when aging or drying; tubes 0.5-1 cm long, unstratified, concolorous with the context. **Hyphal system** dimitic. **Contextual trama** with generative hyphae not observed; skeletal hyphae 1.6-8  $\mu\text{m}$  diam., thick-walled, generally solid, non-septate, non-branched to arboriform, moderately branched, golden-yellow. **Hymenophoral trama** as the contextual trama. **Pileipellis** with cuticle cells 46.5-71.3 x 8.6-13.2  $\mu\text{m}$  diam, clavate, almost cylindrical to occasionally widely clavate, generally without protuberances neither branches, some with occasional lateral or apical protuberances and branches, thick-walled to solid, apex with granulations, brownish-yellow, amyloid in Melzer's reagent; generative hyphae 2.4-3.1  $\mu\text{m}$  diam., thin-walled, with conspicuous clamps, hyaline to yellowish, abundant; skeletal hyphae 3.1-6.2  $\mu\text{m}$  diam., thick-walled to occasionally solid, non-septate, sometimes arboriform, yellowish-brown. **Basidiospores** (10.5-) 11.2-13.6 x 6.5-7.4 (-8.1)  $\mu\text{m}$ , Q = 1.29-1.5, ellipsoid, apex truncate, with visible apical germ pore, yellowish-brown; perisporium smooth, hyaline; exosporium with inter-walled pillars 0.3-0.4  $\mu\text{m}$  thick, free; endosporium wrinkled. **Basidia** not observed. **Cystidia** absent.

*Specimens examined.* s. locality, on Chêne liège trunk, Sep 1890. *J.L. Boudier s.n.*, (PC, Lectotype)

***Ganoderma sessile* Murrill**, Bull. Torrey bot. Club 29: 604, 1902. Fig. 8  
 ≡ *Fomes sessilis* (Murrill) Sacc. & D. Sacc., Syll. fung. (Abellini) 17: 122, 1905.  
 ≡ *Polyporus sessilis* (Murrill) Lloyd, Mycol. Writ. 4 (Syn. Apus): 371, 1915.

**Basidiomata** 5-8 x 7-13 cm, 1-2.5 cm thick in the base, annual, sessile, single to imbricate, woody-corky, light in weight. **Pileus** semicircular, rounded flabelliform to flabelliform, conchate to convex; surface glabrous, bumpy, slightly to radially rugose, hard, glossy, concentrically sulcate mainly toward the margin; with a laccate crust, not cracking, slightly easy to remove, easy to penetrate with fingernail; fully violet-brown (10F6) or photo-brown (9F8) or fully violet-brown very dark almost black, occasionally with raw sienna (6D7) basidiospores over the surface; margin whitish, henna (7E8), lighter than pileus or concolorous, entire, thin to rounded, smooth. **Context** up to 0.3 cm thick in the base, fibrous-corky, azonate, duplex, pale-orange to light-orange (5A3, 5A4) above and reddish-golden to light brown (6C7) close to the tubes; resinous bands generally diffuse and difficult to observe, almost until the margin. **Pores** 4-5 per mm, angular to rounded, woody; pore surface yellowish-white (3A2), darkening to brown (6D8) when bruising or aging; tubes 0.8-1 cm thick, up to 1.4 cm in the base, unstratified, generally concolorous with the inferior part of the context. **Hyphal system** trimitic. **Contextual trama** with generative hyphae 2.4-4  $\mu\text{m}$  diam., thin-

walled, with large and conspicuous clamps, unbranched, hyaline to yellowish, difficult to observe; skeletal hyphae 3.2-12  $\mu\text{m}$  diam., generally solid, non-septate, arboriform, very branched, yellowish to golden-yellow, predominant; binding hyphae 4.8-12  $\mu\text{m}$  diam., solid, non-septate, hyaline to yellowish, scarce. **Hymenophoral trama** as contextual trama. **Pileipellis** with cuticle cells 56-88 x 8-13  $\mu\text{m}$ , clavate, generally without or with one lateral protuberance; thick-walled, generally multistratified, golden-yellow, content immediately black with Melzer's reagent, cells amyloid immediately or after 48 h, predominant the longer cells; generative hyphae 1.6-4.8  $\mu\text{m}$  diam., thin-walled, with conspicuous clamps, branched, hyaline to yellowish, abundant; skeletal hyphae 5.6-6.4  $\mu\text{m}$  diam., generally solid, non-septate, arboriform, very branched, yellow, predominant; binding hyphae 4-11.2  $\mu\text{m}$  diam., generally solid to thick-walled, non-septate, hyaline to yellowish, notably thinner and paler than skeletal hyphae, in some specimens not observed. **Basidiospores** 12-14.4 x 7.2-8.8 (-9.6)  $\mu\text{m}$ , Q = 1.46-2, ellipsoid to oblong, apex truncate, with apical germ pore, yellowish-brown, negative in Melzer's reagent; perisporium wrinkled, hyaline; exosporium with inter-walled pillars 0.56-0.64  $\mu\text{m}$  thick, subfree; endosporium wrinkled. **Basidia** not seen. **Cystidia** absent.

*Specimens examined.* USA, New York, Bedford Park, on *Quercus* trunk, s.date, s.coll., (NY, Lectotype).

*Others specimens examined.* USA, New York, White Plains, on deciduous wood. May 1897, L.M. Underwood s.n. (NY); Bedford Park, on stumps of oak, 1 June 1902, s.coll. (NY); s.loc., on dead fallen trees of *Liquidambar styraciflora*, s.d., s.coll. (NY 12.123, Timber and Forest Diseases Survey); Indianapolis, Greencastle, s.loc., on decaying oak, May 1894, L.M. Underwood s.n. (NY); s.loc., on dead fallen trees of *Liquidambar styraciflora*, 28 November 1911, W.N. Loug s.n. (NY).

*Habitat.* Single or imbricate; subtropical vegetation; on a living trees (i.e. *Salix* sp. and *Quercus* sp.) or on deciduous wood.

*Distribution.* Recorded from Argentina, Mexico and USA.

**Ganoderma sessiliforme** Murrill, Bull. New York. Bot. Gard. 8: 149, 1912.

Fig. 9

$\equiv$  *Fomes sessiliformis* (Murrill) Murrill, Bull. New York. Bot. Gard. 8: 153, 1912.

**Basidiomata** 3.5-4 x 6-8 x 0.8-1.2 cm, up to 2.5 cm thick in the base, sessile to substipitate, annual, single or occasionally imbricate, woody, but light in weight. **Pileus** flabelliform, somewhat concave to convex; surface glabrous, rugose to radially rugose, hard, glossy, slightly concentric sulcate; with a laccate crust, not cracking, not easily removed, penetrable with fingernail; darker than violet-brown (9F8) in the 90% of the surface, then henna (7F7) toward the periphery, with abundant cinnamon (6D6) basidiospores over the surface; margin whitish to concolorous, entire, thin, smooth. **Substipe** 0.6-0.8 x 1-2 cm, lateral, flattened, solid, surface shiny, red-wine to almost black, darker than pileus. **Context** 0.4-0.5 cm average, up to 2 cm thick in the base, fibrous, azonate, homogeneous to relatively homogeneous, orange-white (5A2) to cocoa (6E6),

with a deep yellow (4A8) fringe below the laccate crust, without resinous bands. **Pores** 3-5 per mm, angular to rounded, woody; pore surface pale-yellow to pastel-yellow (3A3-3A4), darkening to brown (6D8) when bruising or aging only in some part; tubes 0.4-0.6 cm thick, up to 1 cm in the base, darker than context. **Hyphal system** trimitic. **Contextual trama** with generative hyphae 1.6-3.2  $\mu\text{m}$  diam., thin-walled, with large and conspicuous clamps, non-branched, hyaline to yellowish; skeletal hyphae 2.4-9.6  $\mu\text{m}$  diam., mainly solid to some thick-walled, non-septate, arboriform, scarcely branched, yellowish, predominant; binding hyphae 1.6-3.2  $\mu\text{m}$  diam., solid, non-septate, yellowish, scarce. **Hymenophoral trama** as the contextual trama. **Pileipellis** with cuticle cells 40-52 x 5.2-8  $\mu\text{m}$ , clavate, generally entire, thick-walled, at times multistratified, golden-yellow, content immediately black with Melzer's reagent, cells amyloid; generative hyphae 2.8-3.2  $\mu\text{m}$  diam., thin-walled, with conspicuous clamps, hyaline to yellowish, abundant, difficult to observe; skeletal hyphae 3.2-7.2  $\mu\text{m}$  diam., mainly solid to some thick-walled, non-septate, arboriform but with few branches, yellowish-brown, predominant; binding hyphae not observed. **Basidiospores** 8.8-11.4 x 6.2-7.8  $\mu\text{m}$ , Q = 1.28-1.65, ellipsoid to oblong, apex truncate, with apical germ pore, yellowish-brown, negative in Melzer's reagent; perisporium wrinkled, hyaline; exosporium with inter-walled pillars 0.5-0.6  $\mu\text{m}$  thick, partially anastomosed, but only in some zones and in some basidiospores free; endosporium wrinkled. **Basidia** not observed. **Cystidia** absent.

*Specimens examined.* MEXICO, Morelos, Municipality of Cuernavaca, on dead wood, 24-27 December 1909, E. & L. Murrill 392 (NY, Lectotype).

*Ganoderma subfornicatum* Murrill, North Amer. Flora 9: 121, 1908. Fig. 10

**Basidiomata** 8 x 9-11 x 2.8 cm, average 2 cm?, perenne, substipitate, with a contracted base, single, woody. **Pileus** redounded flabelliform, generally plane; surface glabrous, bumpy, slightly to radially rugose, hard but easily penetrate with fingernail, very shiny, remarkable concentrically sulcate; with a laccate crust, difficult to remove; totally reddish-brown (darker than 9F8) almost black, homogenous, generally covered with cinnamon (6D6) basidiospores; margin concolorous with the pileus, entire, thick, obtuse to truncate, sulcate. **Substipe** 5.5 x 2 cm, short and thick, cylindrical, horizontal, of the color of the pileus, solid. **Context** 0.4-0.6 cm thick, up to 1 cm thick in the base, fibrous, relatively homogeneous, yellowish-brown above, gradually changing to dark-brown (7F8) next to the tubes; with resinous bands. **Pores** 3-4 per mm, rounded, woody, very small; pore surface brown (6D8); tubes 1.4 cm long, indistinguishable stratified, concolorous with base of the context. **Hyphal system** trimitic. **Contextual trama** with generative hyphae 3.2  $\mu\text{m}$  diam., thin-walled, with large and conspicuous clamps, hyaline, scarce and difficult to observe; skeletal hyphae 5.6-11.2  $\mu\text{m}$  diam., generally solid, generally not branches to few branches, golden-yellow to yellowish-brown, predominant; binding hyphae 1.2-3.2  $\mu\text{m}$  diam., solid, non-septate, hyaline to yellowish, scarce, notably thinner and paler than skeletal hyphae. **Hymenophoral trama** as contextual trama. **Pileipellis** with cuticle cells 56-64 (or more) x 6.4-11.2  $\mu\text{m}$ , not claviform, commonly with a constriction, generally with up to seven protuberances and one to two to branches, mainly solid to thick-walled, not multistratified wall, apex with ferruginous granulations, yellowish, negative with Melzer's reagent; generative hyphae not observe;

skeletal hyphae 5.6-12  $\mu\text{m}$  diam., generally solid, some time septate, arboriform, golden-yellow to yellowish-brown; binding hyphae difficult to measure. **Basidiospores** 9.6-11.8 x 6.4-8)  $\mu\text{m}$ , Q = 1.41-1.69, ellipsoid to oblong, apex subacute, slightly visible apical germ pore, yellowish; perisporium semi-wrinkled, exosporium with inter-walled pillars <0.4  $\mu\text{m}$  thick, subfree; endosporium semi-wrinkled. **Basidia** not seen. **Cystidia** absent.

*Specimens examined.* HONDURAS, s.loc., on dead wood, 1906, M.E. Peck s.n (NY, Lectotype)

***Ganoderma subincrustatum*** Murrill, North Amer. Flora 9: 122, 1908. Fig. 11

**Basidiomata** 7.5 cm, average 1.5 cm thick, up to 2.3 cm thick in the base, perenne, substipitate, with a contracted base, single, woody but light in weight. **Pileus** circular, concave to infundibuliform; surface glabrous, slightly bumpy, soft when fresh, hard when drying, generally glossy, in some changing to dull, concentrically sulcate; with a laccate crust, not cracking, not easily removed, easy to penetrate with fingernail; reddish-brown (9F8, 8F8) to burn-sienna (7D8) close to the base, gradually changing to henna (7E8), deep yellow (5B8) toward the margin in some specimens, with age fully dark reddish-brown, at times terra-cotta (7D7) basidiospores over the surface; margin whitish or lighter than pileus, entire to slightly lobulate, thick, rounded, smooth. **Stipe** 5 x 3 cm, short to large and thick, cylindrical, central, solid, reddish-black, darker than pileus. **Context** 1.1 cm thick, average 0.5-0.7 cm, thinner toward the periphery, fibrous-corky, concentrically zonate, relatively homogeneous, a narrow zone apricot (5B6) close to the pileus, rust-brown to cognac (6E8, 6E7), with discontinuous resinous bands from the base to half or more of the basidioma, with whitish mycelium close to the base. **Pores** 4-6 per mm, angular to rounded, woody; pore surface whitish, yellowish-white (3A2) to pale yellow (4A2, 4A3) when fresh, darkening to ochraceous or yellowish-brown (6C5) when bruising; tubes 0.4 cm long, lighter to concolorous with lower part of the context. **Hyphal system** dimitic. **Contextual trama** with generative hyphae not observed; skeletal hyphae 3-4.8  $\mu\text{m}$  diam., solid, non-septate, non-branched to arboriform, golden-yellow to yellowish-brown. **Hymenophoral trama** as contextual trama, generative hyphae 3.2-5.6  $\mu\text{m}$  diam., thin-walled, non-septate, branched, with a tapering apex, hyaline. **Pileipellis** with cuticle cells 40-64 x 5.6-16  $\mu\text{m}$  diam., narrow clavate to clavate, generally with one or two protuberances or branches, thick-walled, golden-yellow, first reddish in group, negative to occasionally slightly amyloid in Melzer's reagent after 36 h; generative hyphae not observed; skeletal hyphae 3.2-7.6  $\mu\text{m}$  diam., solid to thick-walled, septate to non-septate, arboriform with many branches, golden-yellow to yellowish-brown. **Basidiospores** 9.6-11.2 (-12) x 6.4-7.2 (-8)  $\mu\text{m}$ , Q = 1.4-1.67, ellipsoid, apex truncate, with apical germ pore, yellowish-brown; perisporium wrinkled, hyaline; exosporium with inter-walled pillars up to 0.6  $\mu\text{m}$  thick, partially anastomosed, endosporium wrinkled. **Basidia** 21.6-24 x 10.4-11.2  $\mu\text{m}$ , hyaline. **Cystidia** absent.

*Specimens examined.* JAMAICA, Hope Garden, on log wood stump, 26 October 1902, F.S. Earle 176 (NY, Lectotype; fragment in BPI).

TABLE 1. Comparison of morphologic features between different species of *Ganoderma resinaceum* complex

Species	Color context	Resinous deposit	Size basidiospores ( $\mu\text{m}$ )	Pillars	Cuticle cells
<i>Ganoderma argillaceum</i>	relatively homogeneous, pale to light brown	resinous bands only close to the base	10.4-13.6 x 7.2-9.6, ellipsoid	0.4 $\mu\text{m}$ thick, subfree	clavate, generally without protuberance
<i>Ganoderma chaffangeonii</i>	homogeneous, light reddish-brown	without resinous bands	11.2- 13.2 (-13.6) x 7.2-8.8 (-9.2), ellipsoid	0.3-0.4 thick, free	almost cylindrical to widely clavate, generally without protuberances neither branches
<i>Ganoderma nitidum</i>	relatively homogeneous, light brown to dark-brown	without resinous incrustations	9.6-12.8 (-13.6) x 6.4-7.6 (-8), ellipsoid to oblong	up to 0.5 $\mu\text{m}$ thick, partially anastomosed	clavate, without or up to two protuberances, and/or short and thick branches
<i>Ganoderma perturbatum</i>	relatively homogeneous, light brown to slightly darker	resinous incrustations not very visible	11.2-12.8 x 8-9.6, broadly ellipsoid to ellipsoid	0.7-0.8 $\mu\text{m}$ thick, partially anastomosed	broadly clavate to occasionally conic, without or up to two diverticules
<i>Ganoderma praelongum</i>	duplex, ochre-yellow above and rust brown close to the tubes	without resinous bands	9.6-12 x 6.4-8 (-8.4), ellipsoid	<0.4 $\mu\text{m}$ thick, partially anastomosed	narrowly clavate and clavate, generally without or with one lateral protuberance and/or branches
<i>Ganoderma pulverulentum</i>	ochre-yellow above and dark brown	with discontinuous resinous bands	10.4-12.4 x 6.4-8, ellipsoid	up to 0.4 $\mu\text{m}$ thick, partially anastomosed	narrowly clavate and clavate, generally without or with one lateral protuberance and/or branches
<i>Ganoderma resinaceum</i>	homogeneous, light reddish-brown	without resinous bands	(10.5-) 11.2-13.6 x 6.5-7.4 (-8.1), ellipsoid	0.3-0.4 $\mu\text{m}$ thick, free	almost cylindrical to widely clavate, generally without protuberances neither branches
<i>Ganoderma sessile</i>	duplex, pale-above and light brown	resinous bands generally diffuse	12-14.4 x 7.2-8.8 (-9.6), ellipsoid to oblong	0.56-0.64 $\mu\text{m}$ thick, subfree	clavate, generally without or with one lateral protuberance
<i>Ganoderma sessiliforme</i>	homogeneous to relatively homogeneous, orange-white, with a deep yellow	without resinous bands	8.8-11.2 (-12) x 6.4-8 (-8.4) $\mu\text{m}$ , ellipsoid to oblong	0.5-0.6 $\mu\text{m}$ thick, partially anastomosed	clavate, generally without or with scarce lateral diverticules
<i>Ganoderma subforficatum</i>	relatively homogeneous, yellowish-brown to dark-brown next to the tubes	with resinous bands	9.6-11.8 x 6.4-8), ellipsoid to oblong	0.4 $\mu\text{m}$ thick, subfree	not claviform, commonly with a constriction, generally with up to seven protuberances and one to two to branches
<i>Ganoderma subincrustatum</i>	relatively homogeneous, rust-brown to cognac	discontinue resinous bands	9.6-11.2 (-9.6-11.2 (-12) x 6.4-7.2 (-8), ellipsoid	up to 0.6 $\mu\text{m}$ thick, partially anastomosed	clavate to clavate, generally with one or two protuberances or branches

## Discussion

The type material of *G. resinaceum* was not found in PC, for this reason an authentic material and comparisons with descriptions of Gottlieb & Wright (1999) were made, to make the discussion. The colour context is the more visible feature splitting the complex in two groups: The first group includes species with a brown context as *G. chaffangeonii*, *G. nitidum*, *G. perturbatum*, *G. pulverulentum*, *G. resinaceum*, *G. subincrustatum*, *G. subforficatum*. In contrast, the second group has species with a pale context, it is: *G. argillaceum*, *G. praelongum*, *G. sessile*,

*G. sessiliforme*. Inside of this last group *G. praelongum* and *G. sessile* has duplex context, while that *G. argillaceum* and *G. sessiliforme* has relatively homogenous context. Although all the species with pale context has entire cuticle cells as in *G. resinaceum*, the basidiospore pillars are distinctively different, observation in agreement with Gottlieb & Wright (1999). Other important point is that inside of the group with brown context, *G. nitidum* and *G. subfornicatum* have diverticulate cuticle cells. *Ganoderma perturbatum*, *G. pulverulentum* and *G. subincrustatum* have entire cuticle cells. Nevertheless, the first species has remarkable features as: basidiomata stipitate, basidiospores with subacute apex, thick, short and partially anastomosed basidiospore pillars, and cuticle cells wide. *Ganoderma pulverulentum* and *G. subincrustatum* have context relatively homogenous and basidiospore pillars partially anastomosed, but the first species has basidiomata generally sessile and bigger, woody-corky, and apex of cuticle cells with granulations.

Steyaert (1972, 1977, 1980) gave little importance to the context color arguing on the great variability that may be present in it. But Torres-Torres *et al.* (data unpublished, see chapter IB in this thesis) found that although the shade may vary, the color is constant inside different specimens of the same species. On the other hand, Steyaert (1977), based on Pegler & Young's (1973) work, considered the basidiospore exosporium ornamentation was not variable for specimens with different color context. Besides of the basidiospore size, the main features to synonymy the species on *Ganoderma resinaceum* complex were the thickness and abundance of the pillars, but not their connectivity as was referred by Heim (1962), Steyaert (1967) and Pegler & Young (1973). We found that such pillar connections might be observed under light microscopy. In agreement with Bazzalo & Wright (1999), we clarified on the importance of the disposition of the pillars in the identity of the species.

*Ganoderma polychromum*, *G. subperforatum*, *G. subtuberculosum* and *G. triviale* were not considered in this paper because the types of these species were not available. Some comments about these species may be found in Moncalvo & Ryvarden (1997). Because of its basidiospores, *G. areolatum* does not belong to the genus *Ganoderma*, and for this reason it is described in other paper (Torres-Torres *et al.*, 2007).

*Ganoderma resinaceum* is a species which many mycologists have given a wide concept and thus many synonyms (Steyaert 1972, 1980, Bazzalo & Wright 1982, Ryvarden 2000, 2004). In *G. resinaceum* the weight of the basidiomata, context color, absence of resinous deposits, and thin and subfree pillars of the basidiospores were enough and particular features for the definition of this species. Haddow (1931), Heim (1962), Furtado (1962, 1965a), Steyaert (1975, 1980), Adaskaveg & Gilbertson (1988) made micromorphologic studies in *Ganoderma*; however, in the majority of these works the discussion and the interpretation were limited because few species were systematically compared. We found that there are enough features for the identification of the studied species and for this reason we consider them as independent species from *G. resinaceum*. The conclusion is that *G. nitidum*, *G. perturbatum*, *G. praelongum*, *G. sessile*, *G. sessiliforme*, *G. subfornicatum* are independent species of *G. resinaceum*.

### Acknowledgments

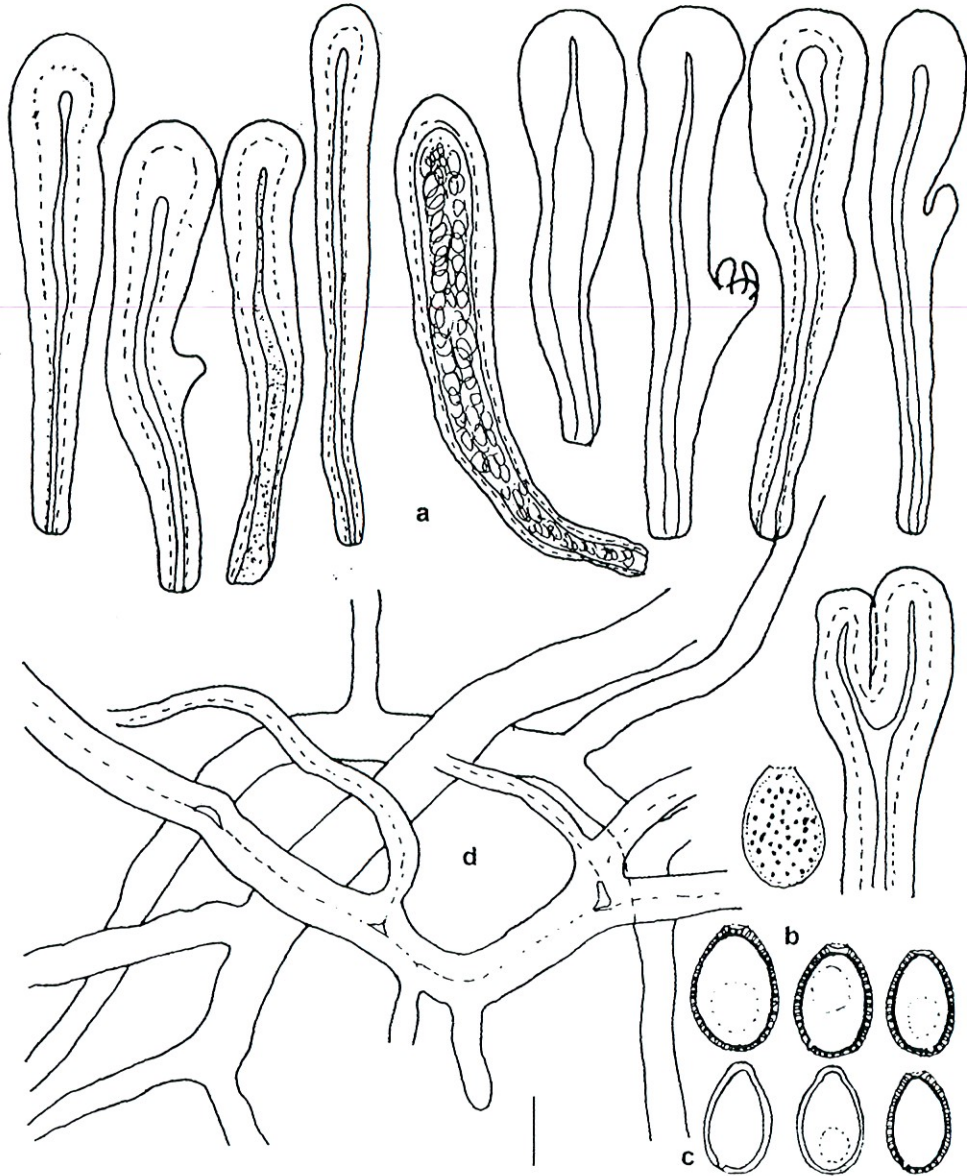
Thanks are due to the curators of BPI, NY and PC herbaria, who kindly proportioned the materials for the study. CONACYT (project CONACYT-SEP-2003-C02-42957) and Universidad de Guadalajara (project 62935) gave financial support. Thanks to Leif Ryvar den by his teaching, time for discussion on *Ganoderma*, and his critical review of this paper. Also, the first author acknowledges Project NUFFIC-Alterra of Wageningen University, COLCIENCIAS and Universidad Tecnológica del Chocó for grants for her Doctoral studies.

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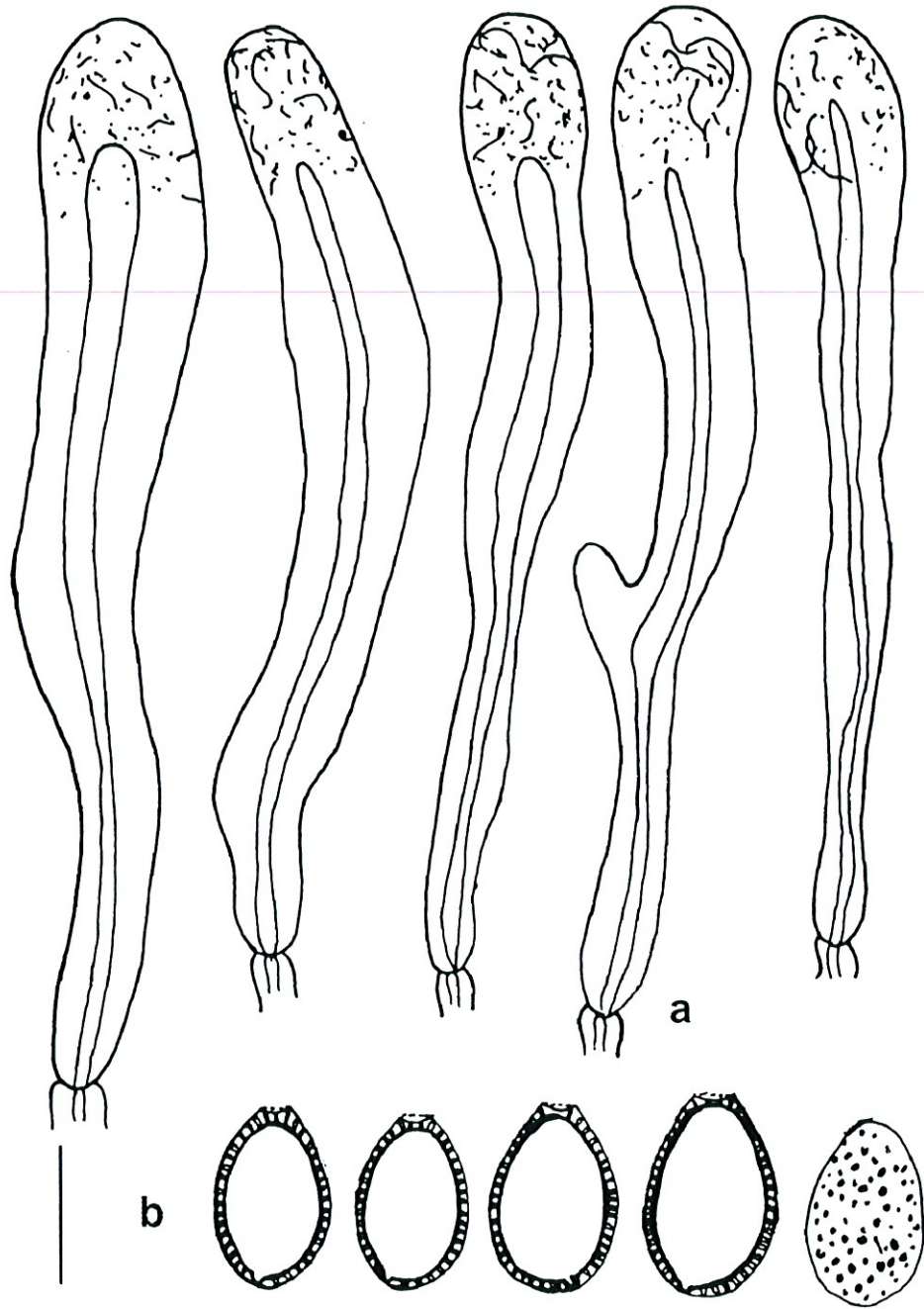
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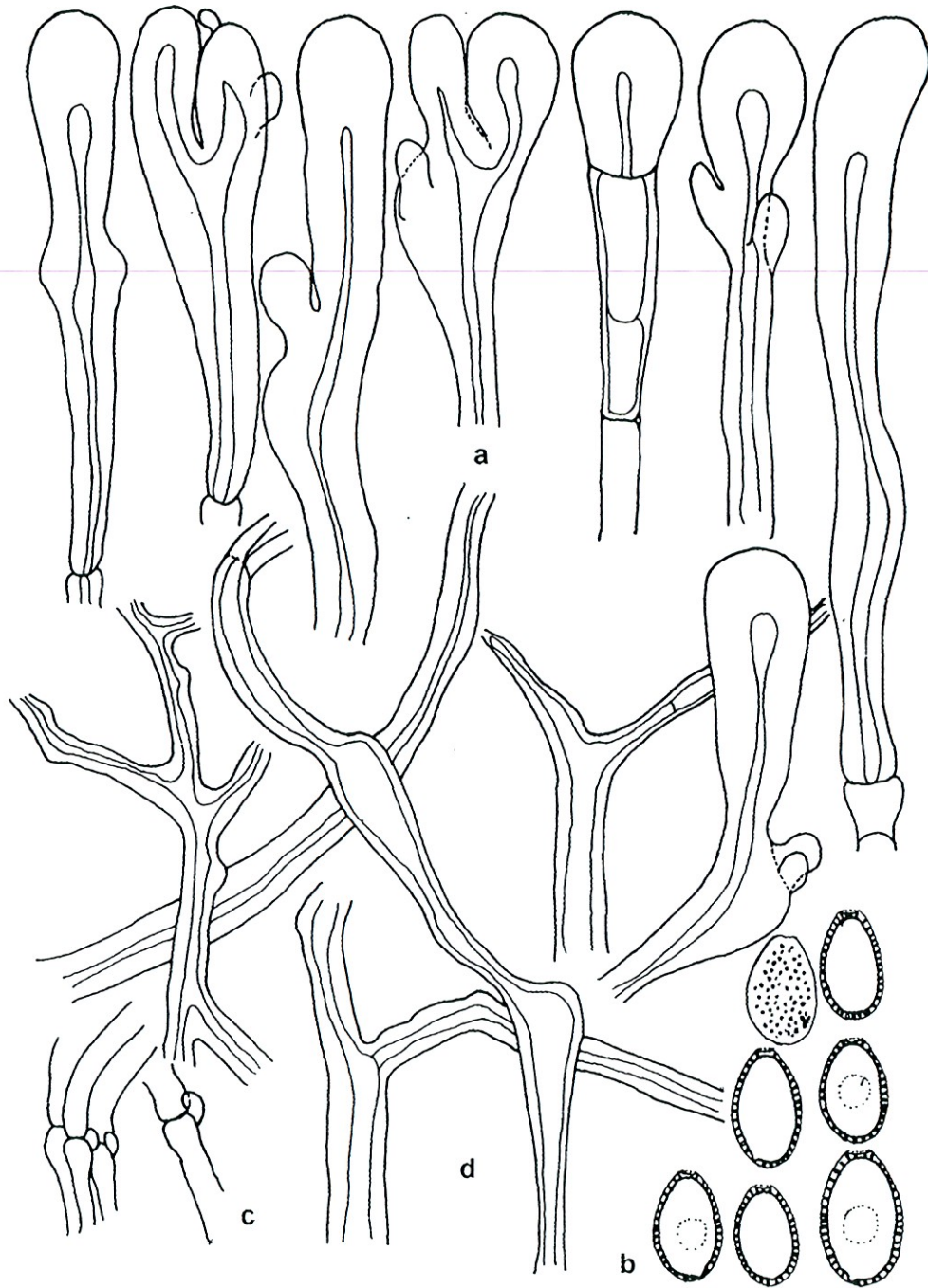




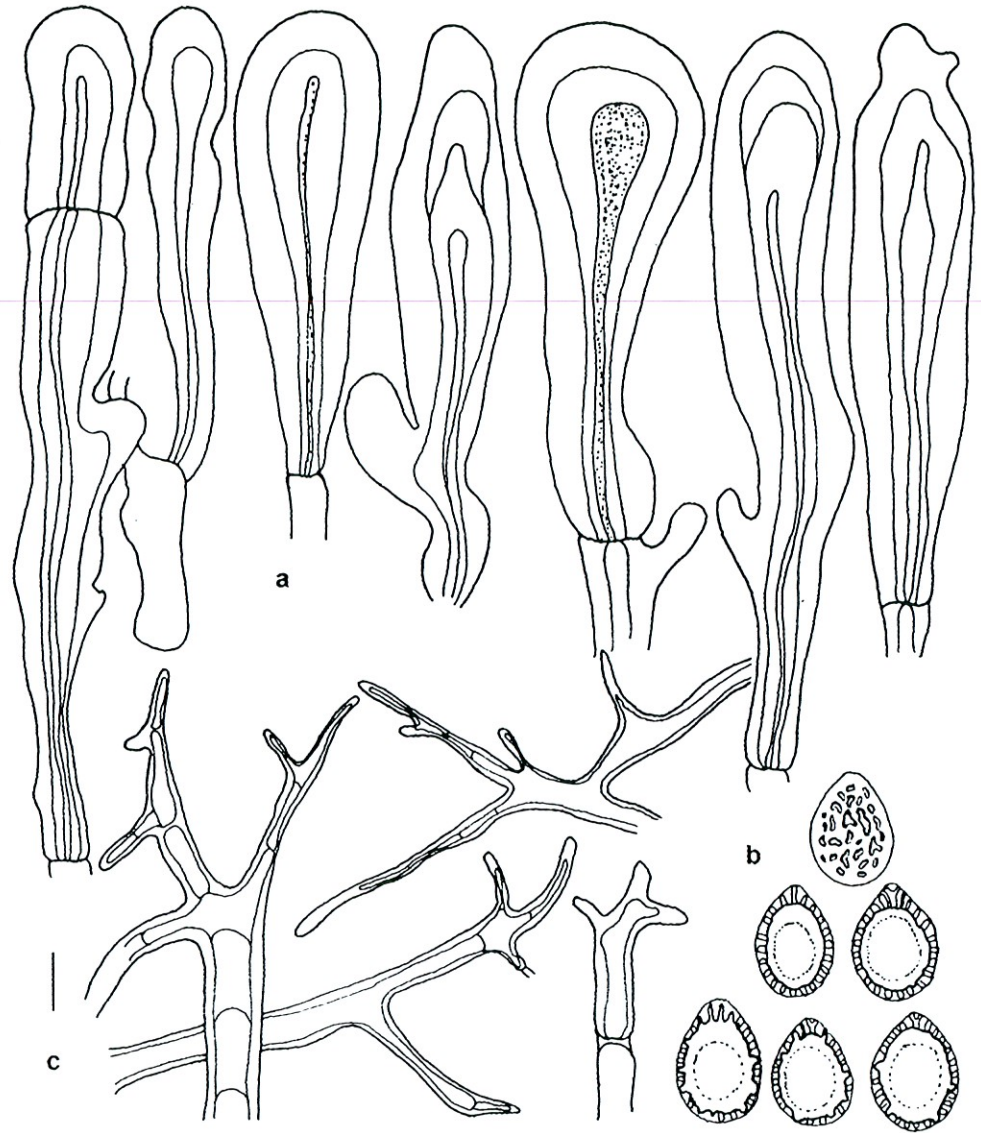
**Fig. 1.** Micromorphological features of *Ganoderma argillaceum*: a. Cuticle cells. b. Basidiospores. c. Immaturity basidiospores. d. Hyphal system of crustohymenodermis: Skeletal hyphae. Bar = 8  $\mu\text{m}$ .



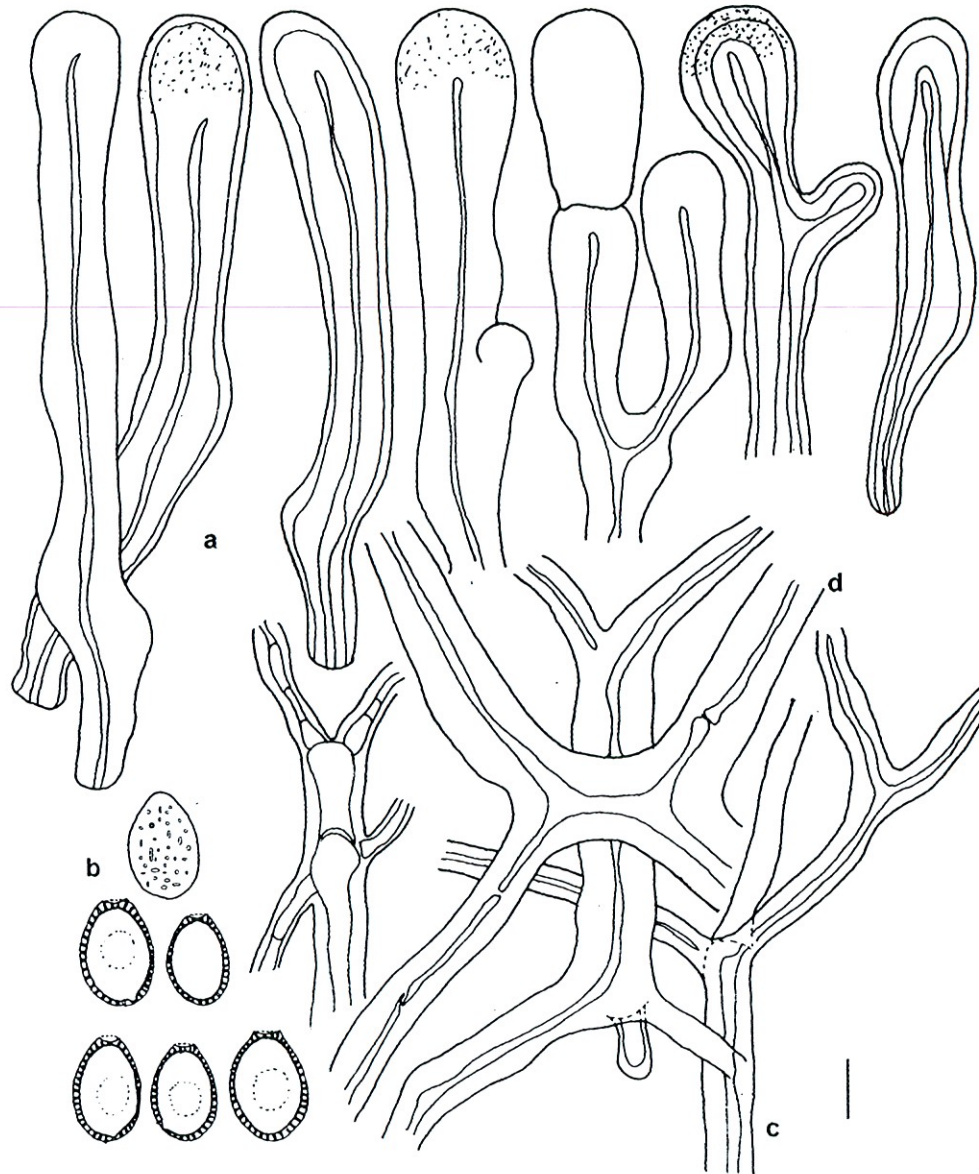
**Fig. 2.** Micromorphological features of *Ganoderma chaffangeonii*: a. Cuticle cells. b. Basidiospores. Bar = 8  $\mu$ m.



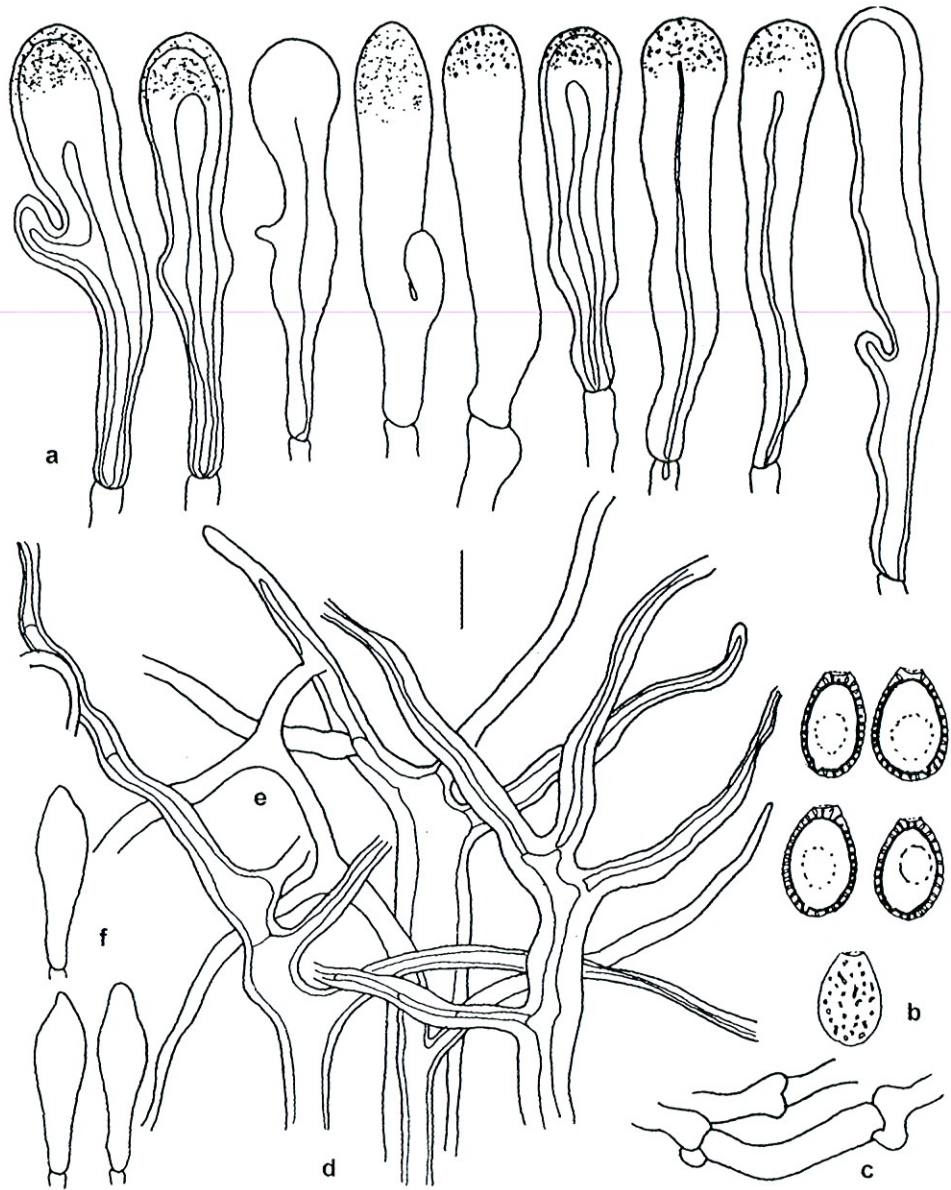
**Fig. 3.** Micromorphological features of *Ganoderma nitidum*: a. Cuticle cells. b. Basidiospores. c-d. Hyphal system of crustohymenodermis: a. Generative hyphae. d. Skeletal hyphae. Bar = 8  $\mu$ m.



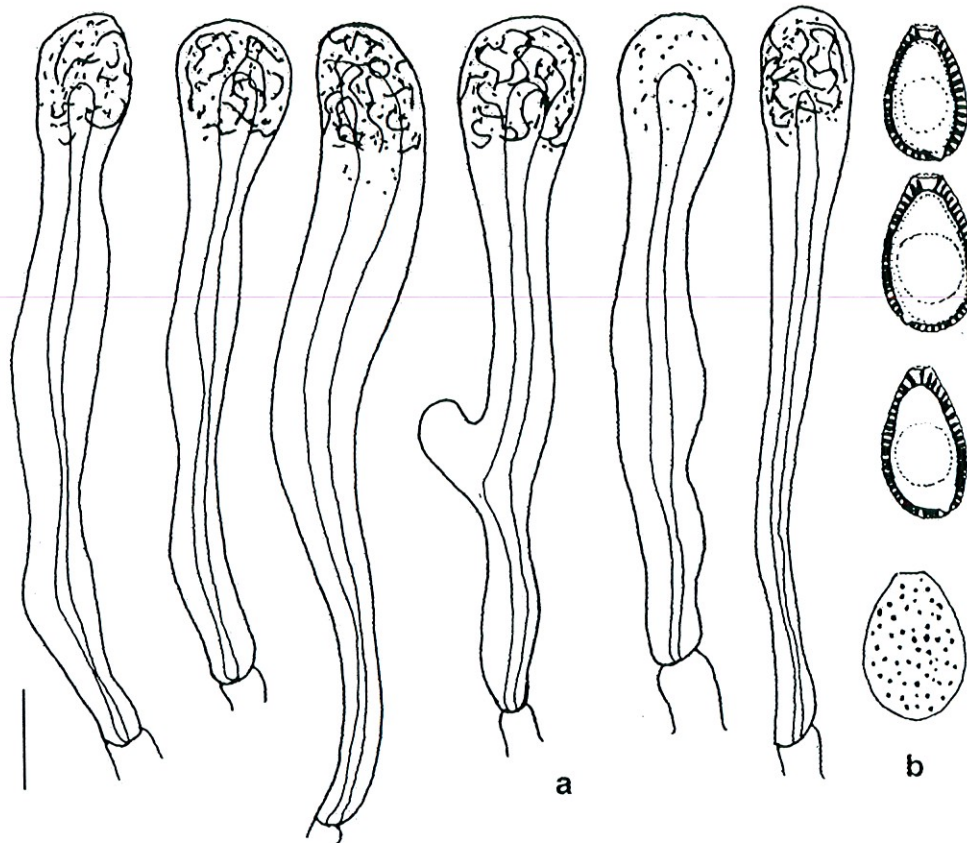
**Fig. 4.** Micromorphological features of *Ganoderma perturbatum*: a. Cuticle cells. b. Basidiospores. c. Hyphal system of crustohymenodermis: Skeletal hyphae. Bar = 8  $\mu$ m.



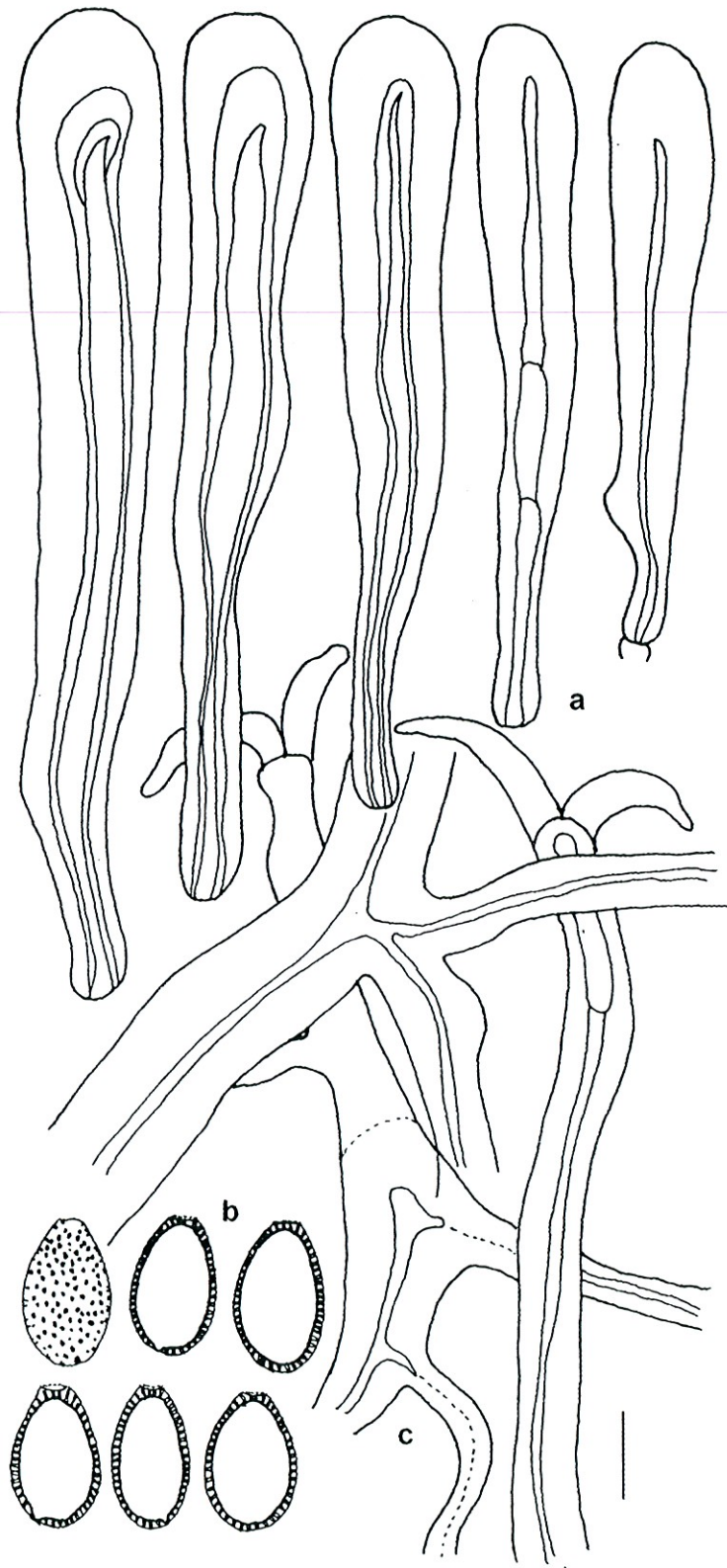
**Fig. 5.** Micromorphological features of *Ganoderma praelongum*: a. Cuticle cells. b. Basidiospores. c-d. Hyphal system of crustohymenodermis: c. Skeletal hyphae. d. Binding hyphae. Bar = 8  $\mu$ m.



**Fig. 6.** Micromorphological features of *Ganoderma pulverulenum*: a. Cuticle cells. b. Basidiospores. c-e. Hyphal system of crustohymenodermis: c. Generative hyphae. d. Skeletal hyphae. e. Binding hyphae. f. Cystidioles. Bar = 8  $\mu$ m.

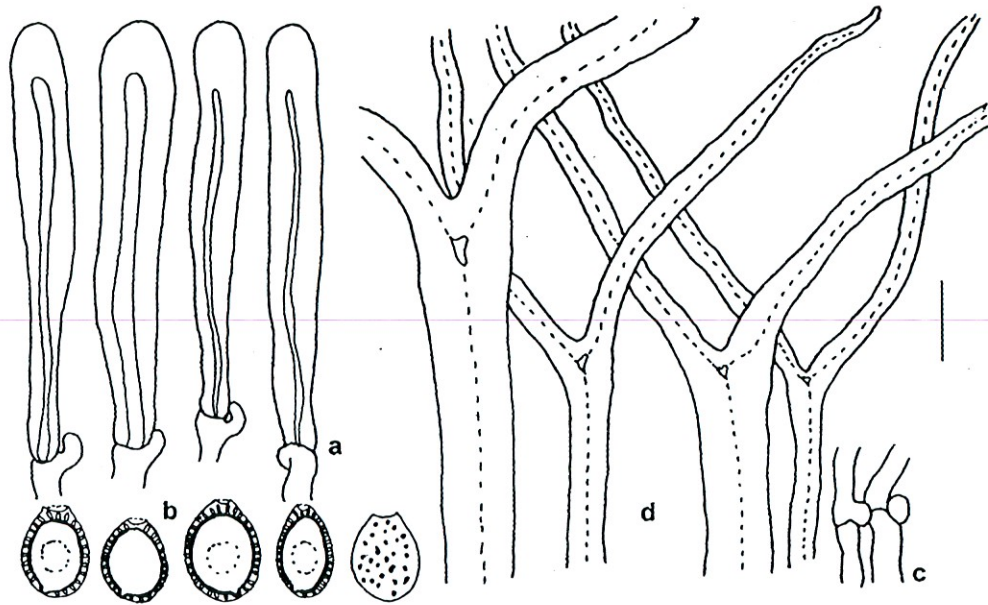


**Fig. 7.** Micromorphological features of *Ganoderma resinaceum*: a. Cuticle cells. b. Basidiospores. Bar = 8  $\mu$ m.

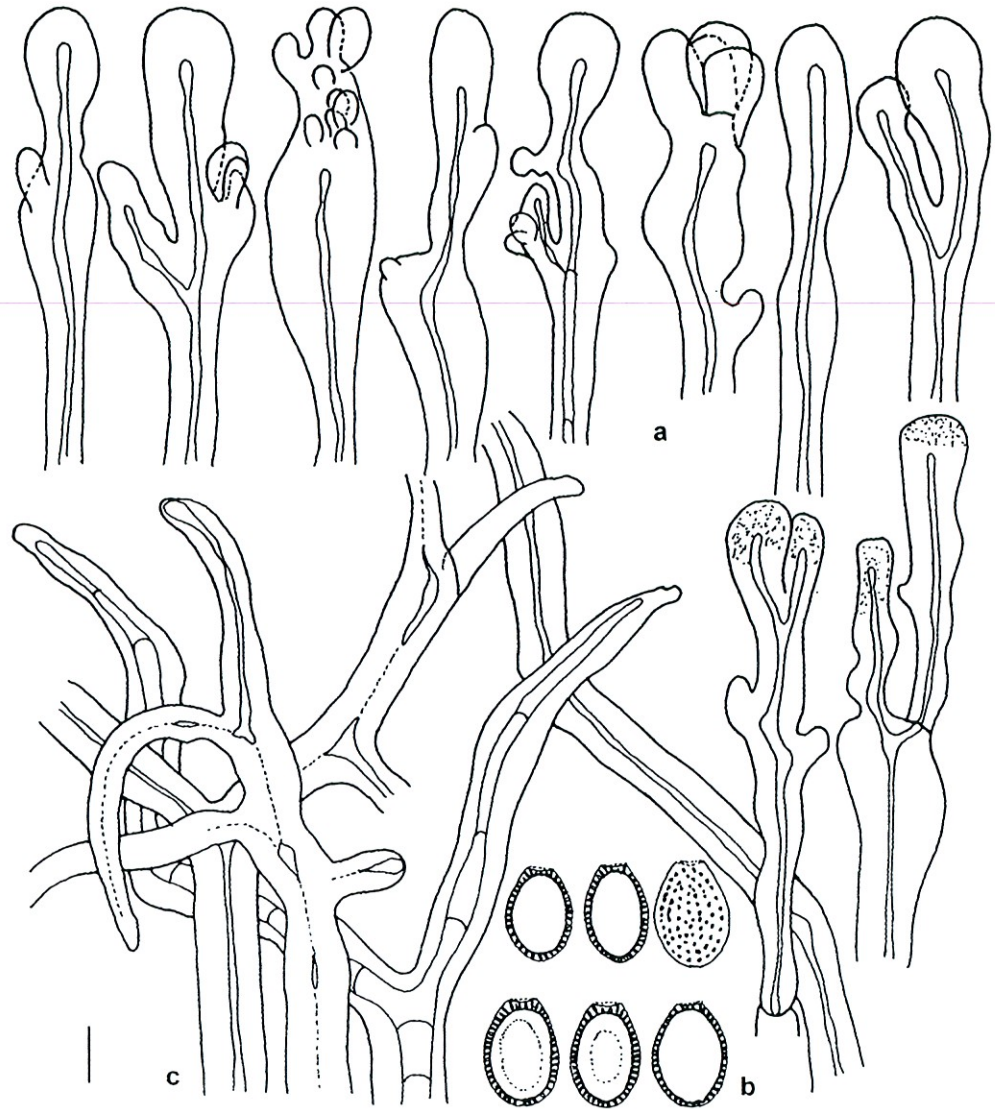


**Fig. 8.** Micromorphological features of *Ganoderma sessile*: a. Cuticle cells. b. Basidiospores. c. Hyphal system of crustohymenodermis: Skeletal hyphae. Bar = 8  $\mu\text{m}$ .

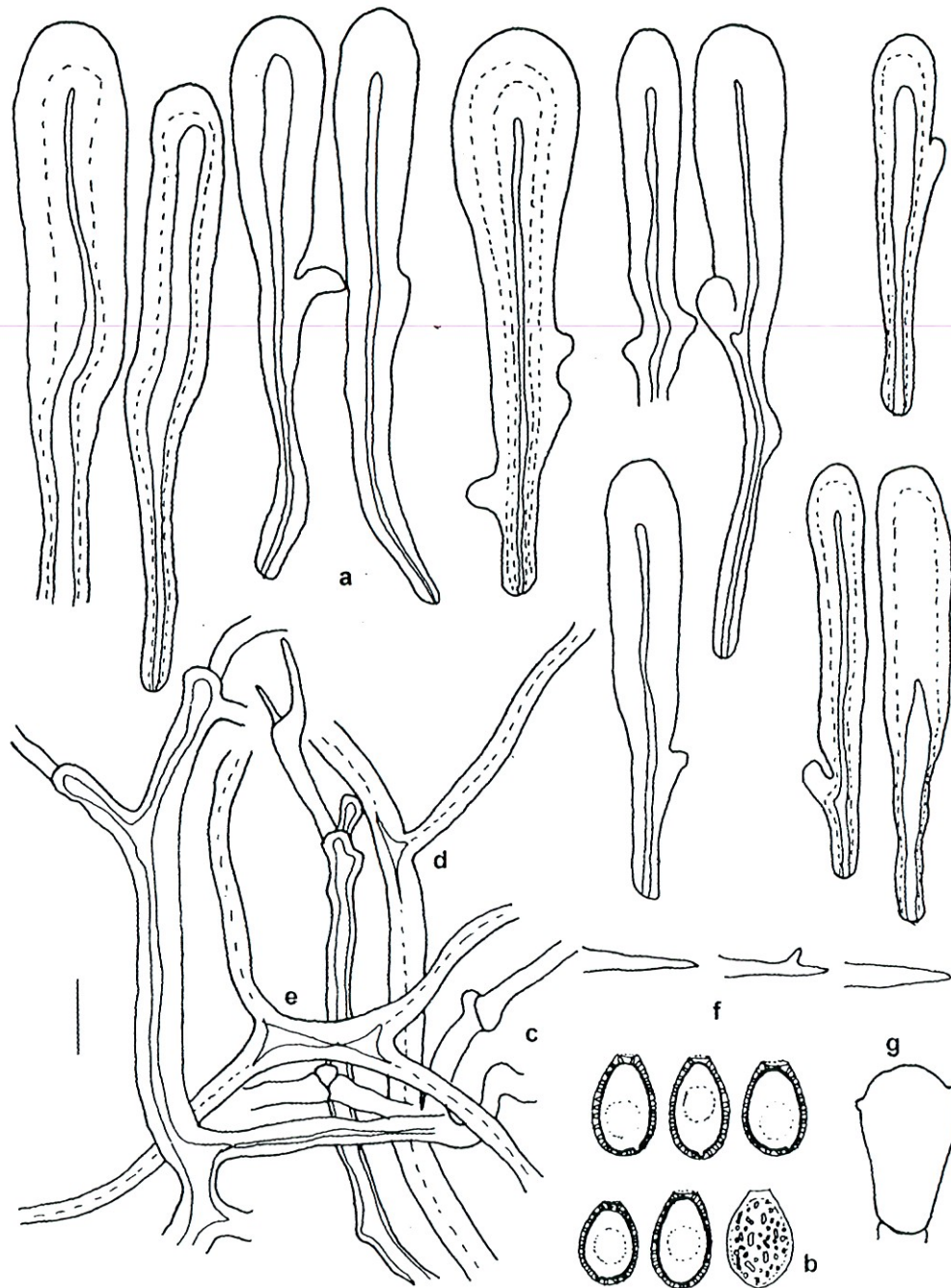




**Fig. 9.** Micromorphological features of *Ganoderma sessiliforme*: a. Cuticle cells. b. Basidiospores. c-d. Hyphal system of crustohymenodermis: c. Generative hyphae. d. Skeletal hyphae. Bar = 8  $\mu$ m.



**Fig. 10.** Micromorphological features of *Ganoderma subfornicatum*: a. Cuticle cells. b. Basidiospores. c. Hyphal system of crustohymenodermis: Skeletal hyphae. Bar = 8  $\mu$ m.



**Fig. 11.** Micromorphological features of *Ganoderma subincrustatum* a. Cuticle cells. b. Basidiospores. c-e. Hyphal system of crustohymenodermis: c. Generative hyphae. d. Skeletal hyphae. e. Binding hyphae. f. Hyphae of hymenophoral trama. Bar = 8  $\mu$ m.

## CAPÍTULO I, PARTE G

### MYCOTAXON

#### *Ganoderma vivianmercedum* sp. nov. and related species

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**Abstract**—A new species of *Ganoderma*, *G. vivianmercedum* is described from Cuba, Brazil and Mexico. The selected type of this species is a specimen that was formerly part of *G. argillaceum* type from Cuba. It is distinguished by its flabelliform basidioma, with a contracted base, conchate, radially rugose pileus, relatively homogenous context and claviform with granulations cuticle cells.

**Key words**—*Ganoderma argillaceum*, *G. perzonatum*, *G. resinaceum*

#### Introduction

In the course of the study of *Ganoderma* developed by the first author, two specimens deposited in NY, marked as the type of *G. argillaceum* Murrill were found; nevertheless, these materials have different features among them. One of the specimens does not coincide with *G. argillaceum* but it is closely related to *G. perzonatum* Murrill and *G. resinaceum* Boud., the first described from Cuba and known only from the type locality. The types or authentic material of *G. argillaceum*, *G. perzonatum* and *G. resinaceum* were studied for comparison with the material in question. In this paper a description of a new species is made, based on one of Murrill's materials of *G. argillaceum*. In addition a full description of *G. perzonatum* is provided.

#### Materials and Methods

Type materials were requested from BPI and NY herbaria, furthermore specimens from IBUG and SP were checked. The colors are based in Kornerup & Wanscher (1963). Micromorphological observations were made through slides of the basidioma mounted in 5% KOH and Melzer's reagent. Basidiospores shape was determined according to Q coefficient (length-width, Bas 1969) of 20 randomly selected spores.

#### Taxonomy

*Ganoderma vivianmercedum* Torres-Torres, sp. nov.

Figs. 1-10

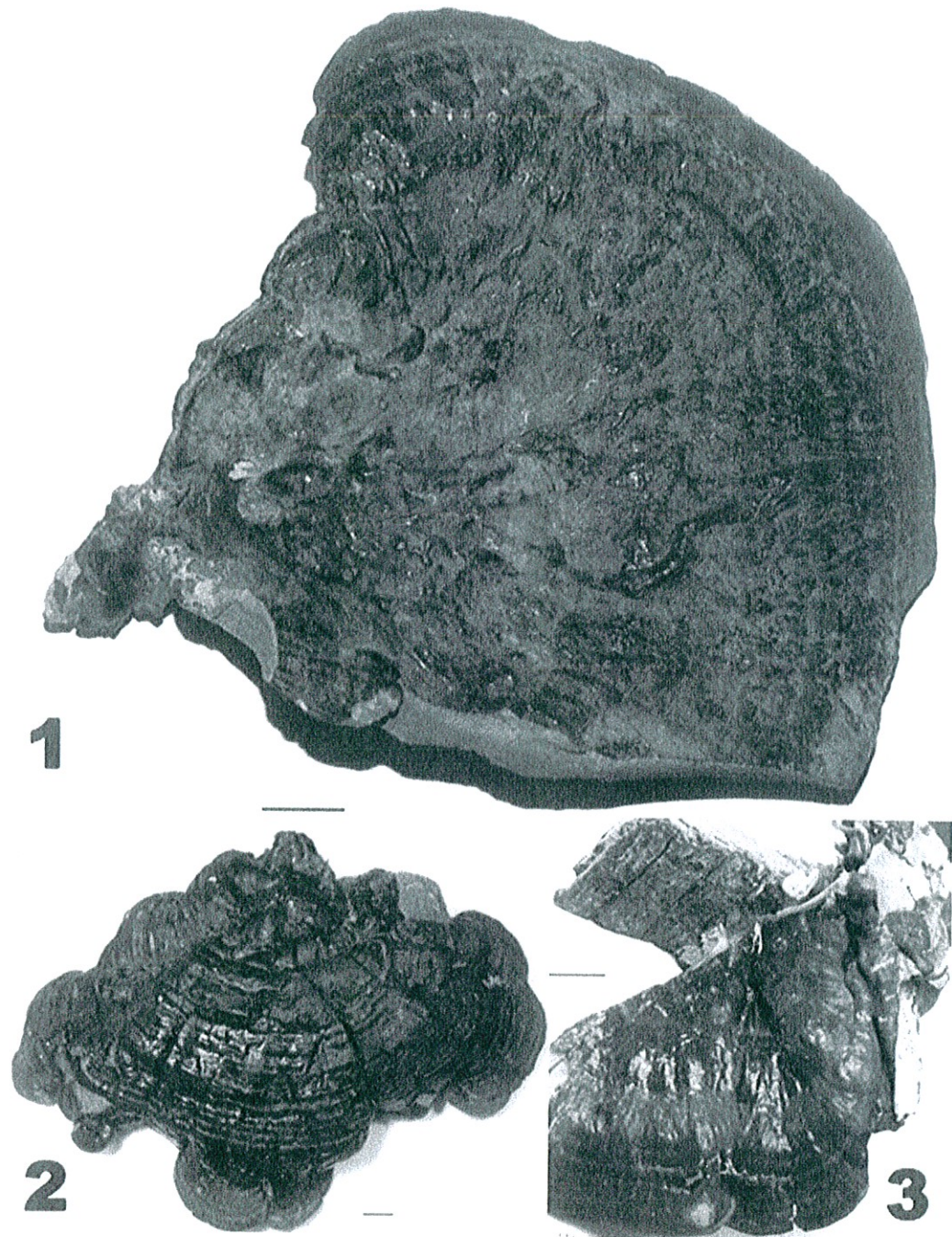
*Fructificatio lignicola, perenne, sessilia vel substipitata; superficies demum rubro-brunnea, crustosa vel laccata; pori 3-5 per mm; contextus pallido-bubalinus vel purpureo-brunneus, non zonae resinosa; systema hypharum trimiticum; hyphae generativae fibulatae; hyphae skeletales nonseptatae, arboriformes; cutis hymenodermiformis, pallide brunnea, amiloidea, elementis entire vel lobata, 36-72 x 7.2-13.6 µm; basidiosporae ellipsoideae, truncate, luteo-brunnea, paries duplexis, cum columnae interparietis, 9.6-11.2 x 6.4-8 µm.*

CUBA, sine data, F.S. Earle s.n. (Holotype: NY).

Etymology: To the memory of Angie Viviana and Rosa Mercedes.

**Basidiomata** 6.5-10 x 8.5-13 x 1.3-1.5 cm, annual, sessile to substipitate, with a contracted base, single, woody-corky, light in weight. **Pileus** flabelliform, conchate; surface glabrous, bumpy, slightly to radially rugose, glossy, concentrically sulcate; with a laccate crust, not cracking,

slightly to radially rugose, glossy, concentrically sulcate; with a laccate crust, not cracking, difficult to remove, easy to penetrate with fingernail; violet-brown (11F7-11F8) in the 80 to 90% of the surface, to henna (7E8-8E8) in the periphery, with basidiospores over the surface; margin whitish to yellowish, generally entire to slightly lobulate, thin, smooth. **Substipe** 2 x 1.3 cm, lateral, cylindrical, solid, surface shiny, red-wine to almost black, darker than pileus. **Context** 0.9-1.3 cm thick, 0.6 cm average, fibrous-corky, zonate, relatively homogeneous, caramel (6C6) above and grading to dark brown (7F7) toward the tubes; with resinous incrustations (not observed in the type). **Pores** 3-5 per mm, angular to circular, woody; pore surface pastel-yellow (2A4), darkening to brown (6D8) when bruising or aging; tubes 0.5-0.9 cm thick, up to 1.2 cm in the base, unstratified, concolorous with the inferior part of the context.



Figs. 1-3. Basidiomata of *Ganoderma vivianmercedum*. 1: Holotype, 2: Pacheco s.n., 3: Sotao et al. 88.21.26. Scale bar = 1 cm.

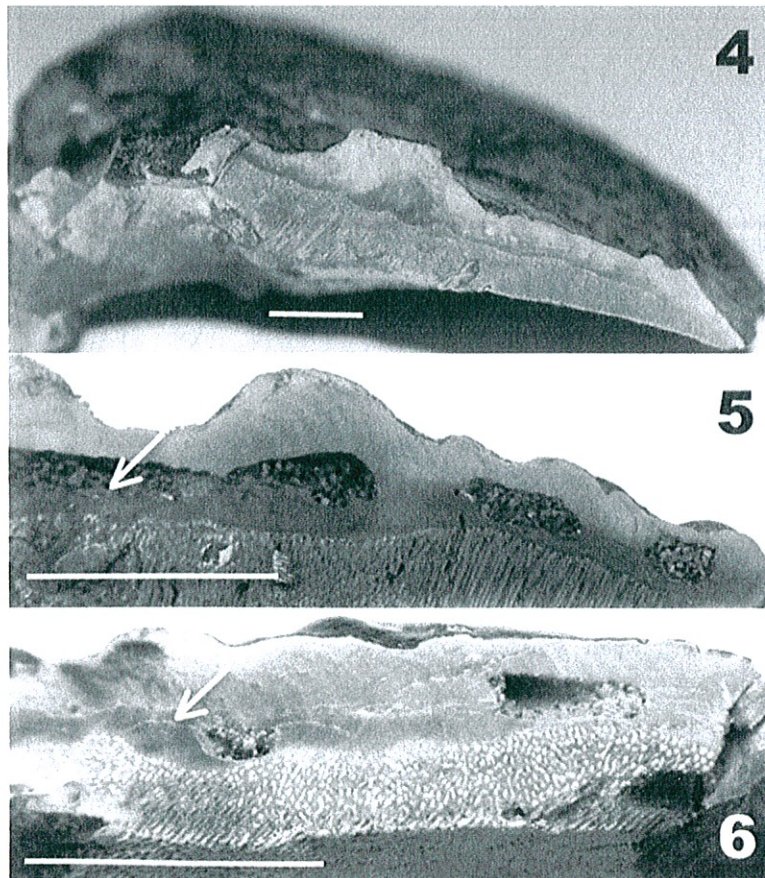
**Hyphal system** trimitic. **Contextual trama** with generative hyphae up to 4  $\mu\text{m}$  diam, hyaline difficult to observed; skeletal hyphae 3.2-12.4  $\mu\text{m}$  diam., generally solid to thick-walled, non-septate, arboriform, very branched, yellowish to golden-yellow, predominant; binding hyphae 3.2-8.8  $\mu\text{m}$  diam., solid, non-septate, yellowish. **Hymenophoral trama** as contextual trama. **Pileipellis** with cuticle cells 36-65 x 7.2-12  $\mu\text{m}$ , narrowly claviform to clavate, entire or with up to two lateral protuberances, not branched or few with two apical branches; thick-walled, unstratified, golden-yellow, with granulations in the apex; cells amyloid in Melzer's reagent; generative hyphae 1.8-4.8  $\mu\text{m}$ , hyaline, branched; skeletal hyphae 3.2-9.4  $\mu\text{m}$  diam., generally solid to thick-walled, non-septate, arboriform, very branched, yellowish to yellowish-brown, predominant; binding hyphae 1.9-5.6  $\mu\text{m}$  diam., solid, non-septate, yellowish to light yellowish-brown, difficult to differentiate from the skeletal hyphae. **Basidiospores** 8.8-11.2 (-12) x 6.4-8  $\mu\text{m}$ , Q = 1.25-1.5, ellipsoid, apex truncate, with apical germ pore, light yellowish-brown, negative in Melzer's reagent; perisporium wrinkled, hyaline; exosporium with inter-walled pillars up to 0.4  $\mu\text{m}$  thick, subfree; endosporium wrinkled. **Basidia** not seen. **Cystidia** absent.

The holotype comprises an almost complete basidioma in good state.

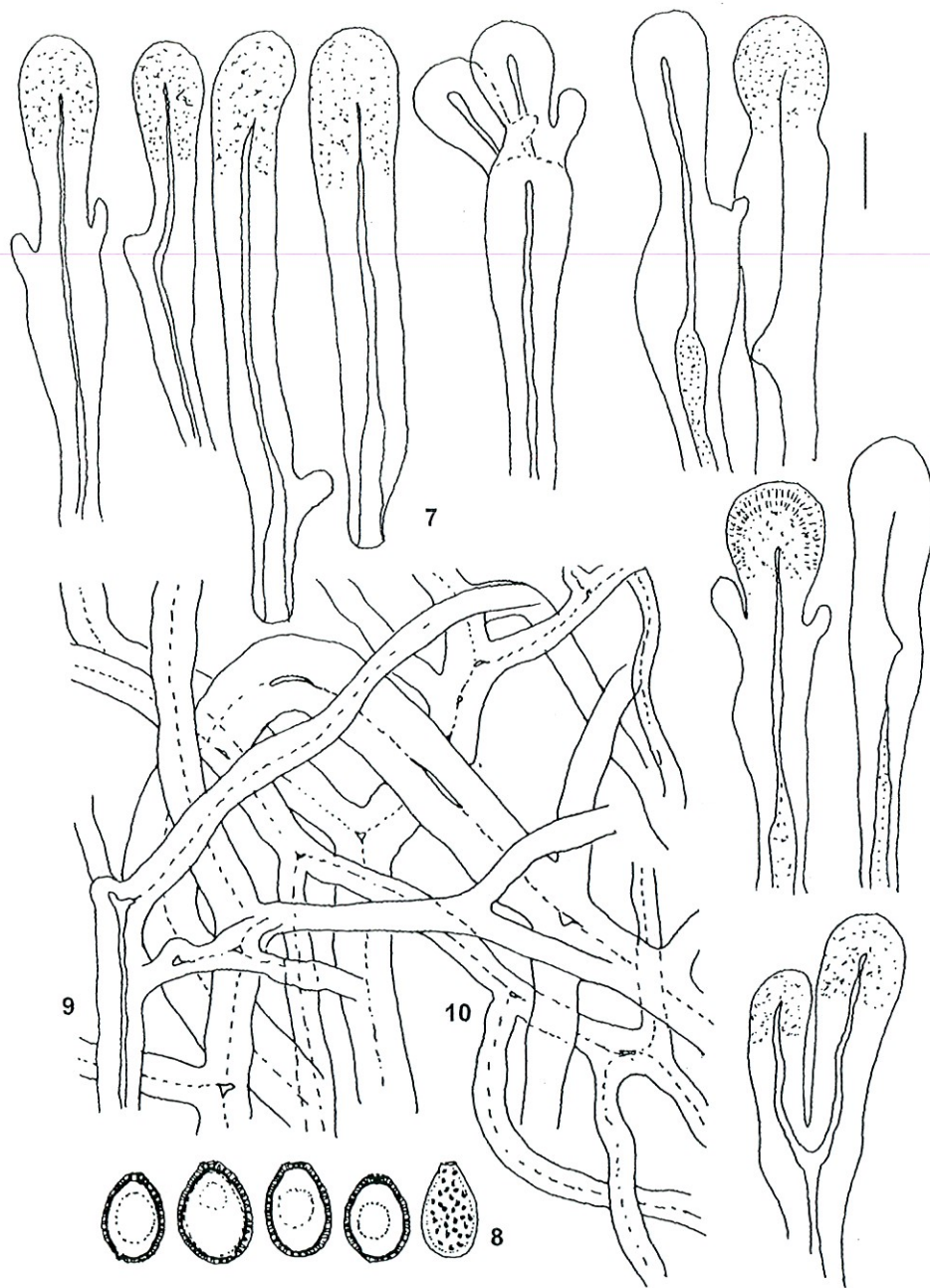
**Substrata** – On a dead wood.

**Habitat** – Mangrove, subtropical and tropical forest.

**Specimens studied** - CUBA, sine data (holotype, NY). BRAZIL, Guaiba, Rio Grande do Sul, Fazenda da Faculdade de Agronomia e Veterinária, em lenho morto, 29 March 1963, J.P. de Costa-Neto s.n. (SP); São Paulo, Tremembé da Cantareira, Villa Amalia, Horto Forestal SPSF, podridão da madeira, 20 May 1952, C.D.F. Pacheco s.n. (SP); Amapá, Ilha de Maracá, tronco de madeira em decomposição, 24 October 1988, H. Sotao et al. 88.21.26 (SP). MEXICO, Jalisco, Zapopan, Centro Universitario de Ciencias Biológicas y Agropecuarias, Departamento de Madera Celulosa y Papel, on dead wood, 21 October 2006, M.G. Torres-Torres 699 (IBUG); Veracruz, Jaltipan, on dead Wood, 19 March 1978, J. Pérez-Ortiz 1278 (ENCB).



Figs. 4-6. Basidiomata context of *Ganoderma vivianmercedum*. 4: Holotype, 5: Pacheco s.n., 6: Sotao et al. 88.21.26. Scale bar = 1 cm.



Figs. 7-10. Microscopy of *Ganoderma vivianmercedum*, holotype. 7: Cuticle cells, 8: basidiospores, 9: skeletal hyphae, 10: binding hyphae. Scale bar = 8  $\mu$ m.

*Ganoderma perzonatum* Murrill, North Amer. Flora 9: 121, 1908.

Figs. 11-13

**Basidiomata** 2.5-8.5 x 4.8 x 0.7-1.5 cm, annual, substipitate, single to imbricate, woody-corky, light in weight. **Pileus** flabelliform; surface glabrous, bumpy, glossy, concentrically sulcate; with a laccate crust, not cracking, difficult to remove, easy to penetrate with fingernail; violet-brown (11F8) almost homogeneous grading to garnet-red (11E8) towards the periphery, with basidiospores over the surface; margin concolorous, entire, thin, smooth. **Context** 0.3-0.9 cm thick, 0.7 cm average, fibrous, zonate, relatively homogeneous, pale-orange to light-orange (5A3,

5A4) above, grading to light brown (6D7) toward the tubes; with resinous bands. **Pores** 4-5 per mm, angular to circular, woody; pore surface pale-yellow (2A3); tubes 0.5-0.9 cm thick, up to 1.2 cm in the base, unstratified to stratified, concolorous with the inferior part of the context. **Hyphal system** dimitic. **Contextual trama** generative hyphae not observed; skeletal hyphae 6.4-13.6  $\mu$ m diam., generally solid to thick-walled, non-septate, arboriform. **Hymenophoral trama** as contextual trama. **Pileipellis** with cuticle cells 70.4-97.6 x 5.6-20.8  $\mu$ m, cylindrical, apex obtuse to rarely subcapitate, entire or with one lateral protuberance; thick-walled, generally multistratified in the apex, golden-yellow, with concentric elongate granulations in the apex; dextrinoid with Melzer's reagent; generative hyphae not observed; skeletal hyphae 6.4-8.8  $\mu$ m diam., generally solid to thick-walled, non-septate, arboriform, yellowish. **Basidiospores** 8.4-10.4 x (6-) 6.4-7.2 (-7.6)  $\mu$ m, Q = (1.25-) 1.36 -1.53, ellipsoid, apex truncate, with apical germ pore, light yellowish-brown, negative in Melzer's reagent; perispodium wrinkled, hyaline; exosporium with inter-walled pillars up to 0.4  $\mu$ m thick, free; endosporium wrinkled. **Basidia** not seen. **Cystidia** absent.

The lectotype is a half basidioma in good condition. In BPI there are fragments of the type, consisting in: a half of a small imbricate basidiomata, two fragments of about 6 x 2.5 cm, and two slices, one including pileus, context and tubes, and the other with the laccate crust in both surfaces and context.

**Substrata** – On mango log.

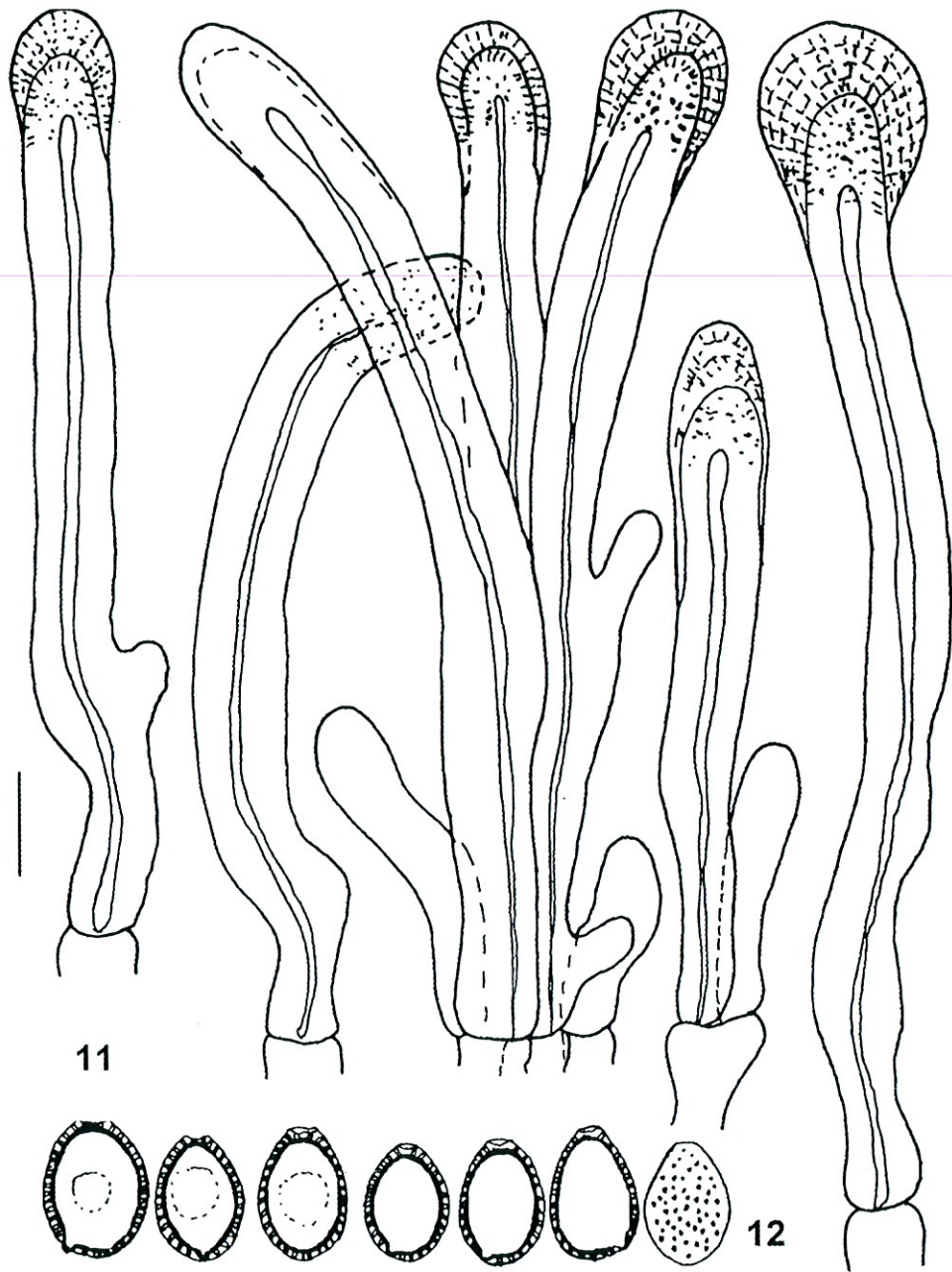
**Habitat** – Not specified.

**Specimens studied** – CUBA, Santiago de Las Vegas, 8 November 1904, F.S. Earle 309 (Lectotype: NY; Isolectotype: BPI).

## Discussion

The type of *Ganoderma argillaceum* deposited in NY is a specimen marked as: CUBA, Province of La Habana, Santiago de Las Vegas, on dead mango log, 5 July 1904, F.S. Earle 658. There is another specimen in NY as *G. argillaceum* from the same collector and probably the same locality with no data, only with a hand written label that says: "part of type, Earle, Cuba". Moncalvo & Ryvardeen (1997) did not make reference to the last material. The last material was checked by J.E. Wright in May of 1967, who wrote the note: "Very much like *G. perzonatum* Murr.!". It was also checked by M.E. Bazzalo in December of 1980, who considered it as a synonym under *G. resinaceum*. We checked this specimen and agreed with Wright's observation that the material did not completely agree with *G. argillaceum* and was close to *G. perzonatum*. However, it does not correspond with *G. perzonatum*, so we are describing the new species. This species is morphologically close to *G. argillaceum*, *G. perzonatum* and *G. resinaceum*; but *G. argillaceum* has greater basidiospores (10.4-13.6 x 7.2-9.6  $\mu$ m) and cuticle cells without granulations. *Ganoderma perzonatum* has slightly smaller basidiospores (8.4-10.4 x 6.4-7.2  $\mu$ m), remarkable cylindrical and large cuticle cells, concentric elongate granulations only in the apex, as discussed above. *Ganoderma resinaceum* has greater basidiospores [11.2-13.6 x 6.5-7.4 (-8.1)  $\mu$ m], cuticle cells narrowly clavate, almost cylindric, and the context is fibrous-spongy, homogeneous, light reddish-brown, without resinous bands. Ryvardeen (2004) suggested a relation of *G. perzonatum* with *G. resinaceum*. We consider these species independent, each one with unique features. Moradali et al. (2007) recorded *G. resinaceum* from Iran, but according with the picture of the basidioma and the macro and micromorphological descriptions, their specimen may be *G. vivianmercedum*. However, they did not describe the granulations on the apex of the pileipellis cells. *Ganoderma vivianmercedum* can be recognized by the flabelliform basidiomata with a contracted based, the relatively homogenous, light brown context and the mainly narrowly claviform to clavate cuticle cells with scattered granulation in the apex. The Brazilian and Mexican specimens have resinous incrustations in the context near to the pileus base, not observed in the type because the basidioma is not cut in the base.





Figs. 11-13. Microscopy of *Ganoderma perzonatum*, lectotype. 11: cuticle cells, 12: basidiospores, 13: skeletal hyphae. Scale bar = 8  $\mu$ m.

#### Acknowledgments

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# MYCOTAXON

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## New data and localities for *Navisporus* in America

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**Abstract** – The genus *Navisporus* is recorded for first time from Mexico, and the species *N. floccosus* is recorded for the first time from Mexico and Cuba. Based on study of the type specimen of *Ganoderma areolatum*, this species is synonymized under *N. floccosus*. Furthermore, *Navisporus sulcatus* is recorded from northern Brazil.

**Key words** – *Polyporaceae*, *Basidiomycota*, taxonomy

## Introduction

*Navisporus* Ryvar den (*Polyporaceae*) (Ryvar den & Johansen 1980) is characterized by its dextrinoid skeletal hyphae and navicular basidiospores. It is a relatively small genus, with six species described, and mainly with a pantropical distribution (Ryvar den 1991). The majority of the species are poorly known, with a very restricted distribution. Four species have been described from the Neotropics: *Navisporus floccosus*, *N. sulcatus*, *N. perennis* Ryvar den & Iturr. and *N. terrestris* Gibertoni & Ryvar den, and two from the Paletropics: *N. africanus* Ryvar den and *N. ortizii* S. Herrera & Bondartseva (Ryvar den & Iturriaga 2003, Gibertoni et al. 2004). Of these species, two are recently described and known only from their type localities: *N. perennis* from Venezuela (Ryvar den & Iturriaga 2003) and *N. terrestris* from Brazil (Gibertoni et al. 2004). *Navisporus floccosus* has been recorded from North America, Africa and Asia (Ryvar den & Johansen 1980) and *N. sulcatus* was cited from Florida (Gilbertson & Ryvar den

1986) and southern Brazil (Rajchenberg & Meijer 1990). The genus has not previously been recorded from Mexico.

In the course of *Ganodermataceae* studies from tropical America, the type specimen of *Ganoderma areolatum*, deposited at NY, was checked. This material has morphological features different from *Ganoderma*. Currently, *Ganoderma* is divided in two subgenera: *Ganoderma* and *Elfvigia*. Subgenus *Ganoderma* is characterized by having a glossy cuticle formed by cuticle cells; in contrast, in subgenus *Elfvigia* the crust is opaque, and does not present differentiated cells. *Ganoderma areolatum* was placed in the subgenus *Ganoderma* (Moncalvo & Ryvarden 1997) and was treated as a synonym of *G. resinaceum* Boud. (Ryvarden 1985), also in subgenus *Ganoderma*.

### Materials and methods

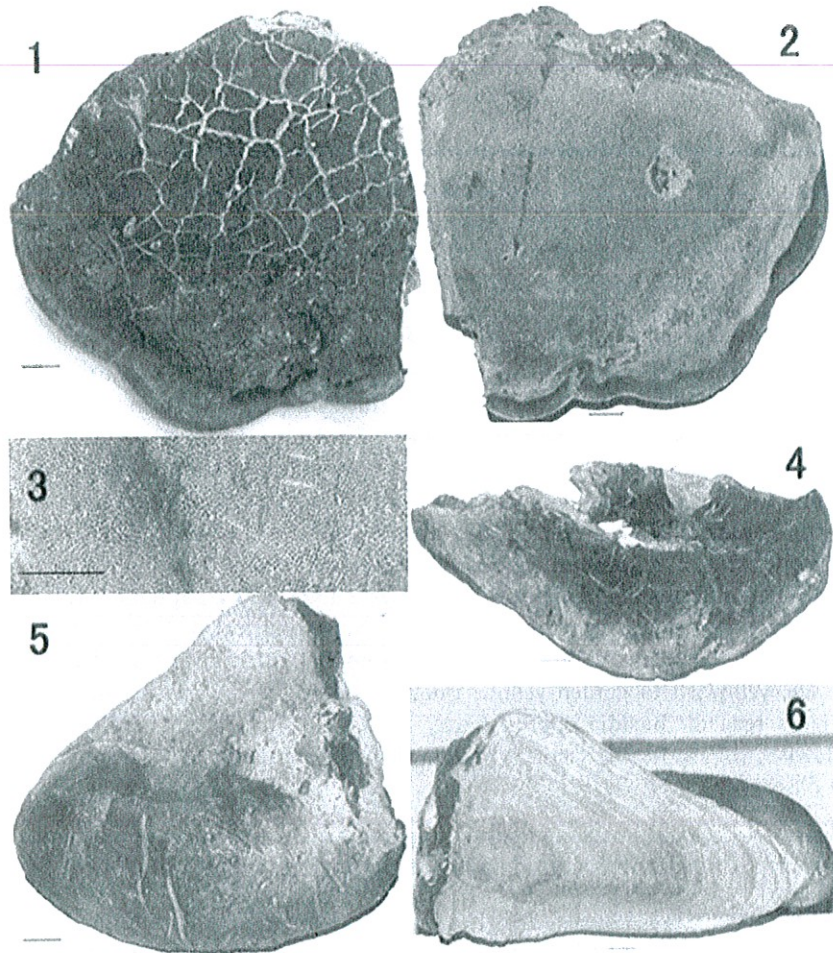
The material studied was requested to ENCB, NY and O herbaria; the new collection made in Mexico was deposited in IBUG. Descriptions of the basidiomata were made according to the following macro features: basidioma size, substrate adhesion, pileus color, consistency; tubes stratification, length, color, pores per mm, pore surface color; context stratification, width, color. The color was described using the key colors of Kornerup & Wanscher (1978). Micromorphological observations were made from material mounted in 5% KOH and Melzer's reagent; measures were made in 5% KOH. Basidiospore shape was determined according to Q coefficient (length-width, Bas 1969) of 20 randomly selected basidiospores. Herbarium abbreviations follow Holmgren et al. (1990).

### Taxonomy

The genus *Navisporus* is reported for the first time from Mexico. *Navisporus floccosus* is recorded from Mexico and Cuba. *Navisporus sulcatus*, an apparently rare species, is recorded from northern Brazil. The type specimen of *Ganoderma areolatum* has morphological features different from *Ganoderma*; here it is synonymized under *Navisporus floccosus*.

From the two species considered, only *Navisporus floccosus* is described in detailed, both macro and micromorphologically, while *N. sulcatus* is described only micromorphologically.

*Navisporus floccosus* (Bres.) Ryvarden Figs. 1-10, 13-16  
in Ryvarden & Johansen, Prelim. Polyp. Fl. E. Afr. (Oslo): 443, 1980.  
= *Trametes floccosa* Bres., Ann. Roy. Inst. Bot. Roma 6: 179, 1896.  
= *Ganoderma areolatum* Murrill, Bull. New York Bot. Gard. 8: 149, 1912.



Figs. 1-6. *Navisporus floccosus*. 1-2: Basidiomata of *Ganoderma areolatum* (holotype), 1: pileus, 2: pore surface, 3-4: basidiomata (L. Guzmán-Dávalos 9904), 3: pore surface, 4: pileus, 5-6: basidiomata (J. Pérez-Ortiz 1016), 5: pileus, 6: context. Scale bar = 1 cm, except 3 = 3 cm.

**Basidiomata** 20-35 x 13-20 x 5.5-7 cm, annual, sessile, broadly attached, single, soft-corky, light weighted, context thinner than tubes. **Pileus** rounded flabelliform, convex; surface glabrous, smooth, dull, without zonation; with a distinctive crust in old specimens, wrinkled, up to 0.5 mm thick; first cream (4A3) darkening to mandarin-orange (6B8) when bruised, grayish-brown (13F) to almost black in adult or old specimens; margin thin to thick, at times rounded, even, cream to grayish-brown. **Context** averaging 2-4 cm thick, up to

7 cm thick at the base, soft, zonate, orange-white (5A2) to pale orange (5A3), darker near the tubes, dark chestnut (6F7), without resinous substance. Pores 2-3 per mm, generally irregular; pore surface pale yellow (4A3), darker when bruising or aging; tubes 2.5-4 cm long, concolorous with pore surface. KOH: pileus black; context, tubes and pore surface blackish-brown.

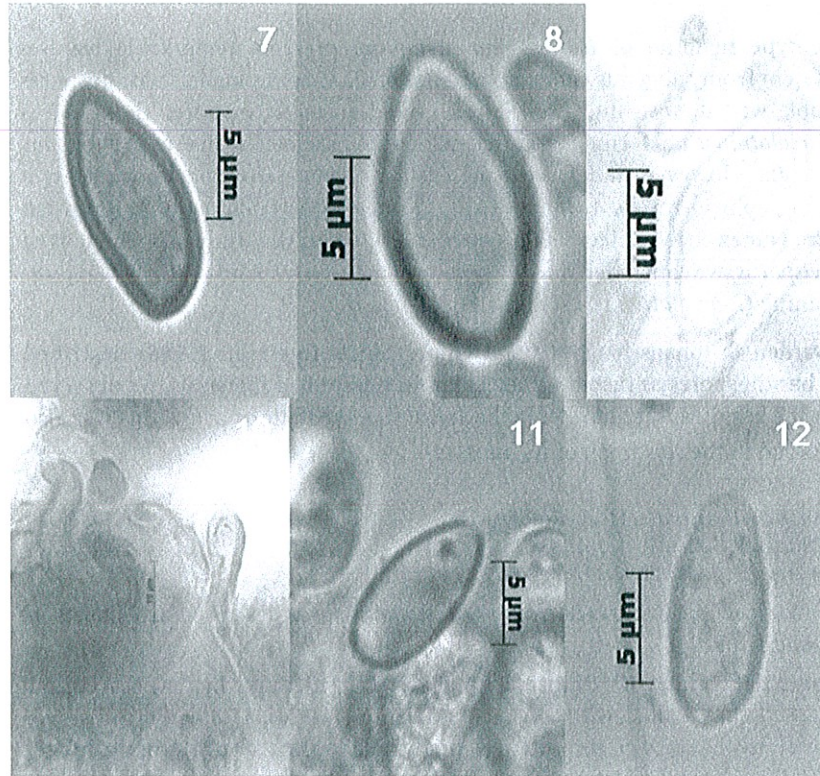
**Contextual trama with dimitic hyphal system;** generative hyphae scarce and collapsed, thin-walled, with conspicuous clamps, yellowish, difficult to observe; skeletal hyphae 1.6-14.4  $\mu\text{m}$  diam., thick-walled (0.4-2.4  $\mu\text{m}$ ), non-septate, generally unbranched to moderately branched, yellowish to yellow, dextrinoid. **Hymenophoral trama with dimitic hyphal system;** generative hyphae 2.4-2.8  $\mu\text{m}$  diam., thin-walled, with conspicuous clamps, yellowish, scarce, difficult to observe; skeletal hyphae up to 6.4  $\mu\text{m}$  diam., thick-walled, non-septate, sometimes branched, yellowish to yellow, slightly dextrinoid to dextrinoid. **Pileus crust** when present formed by erect and rounded apex hyphae as a palisoderm; generative hyphae 3.2-6.4  $\mu\text{m}$  diam., thin-walled to sclerified, with conspicuous clamps, single or multiple, generally branched, hyaline to yellowish, abundant, forming a compact network; skeletal hyphae 4.5-9.6  $\mu\text{m}$  diam., thick-walled (0.8-4  $\mu\text{m}$ ), unbranched to branched, yellow to brown-amber, with granulose content, some septate toward apex and constricted; sclerids present. **Basidiospores** 11.2-13.6 x 5.6-7.2  $\mu\text{m}$ , Q = 1.76-2.29, oblong to cylindrical, amygdaliform to subcylindrical, with the apical side acute to subacute, without germ pore, smooth, with conspicuous lateral apicule, yellowish to golden-yellow; thick-walled (up to 1  $\mu\text{m}$ ), negative in Melzer's reagent. **Basidia** approximately 18.4 x 5.6  $\mu\text{m}$ , clavate, hyaline to yellowish, tetrasporated, scarce. **Cystidia** or other sterile hymenial elements not observed.

**Substrata** - On a dead trunk of a silk-cotton tree, on a living trunk of *Ficus*, and on an unidentified stump, according to the specimens examined.

**Habitat** - Secondary tropical forests.

**Specimens studied** - CUBA, Province of La Habana, Escaleras de Jaruca, 16 June 2001, C. Decock s.n. (O). MEXICO, Colima, near Colima, January 1910, W.A. Merrill & E.L. Merrill 588 (NY, Holotype of *Ganoderma areolatum*); Jalisco, Municipality of Puerto Vallarta, Hotel NH-Kristal, alt. 5 m, 28 October 2005, L. Guzmán-Dávalos 9904 (IBUG); Veracruz, Municipality of Minatitlán, El Remolino, Río Coachapa, 7 August 1977, J. Pérez-Ortiz 1016 (ENCB).

**Remarks** - The studied specimens coincide with Ryvarden (2004), except that this author described basidiospores slightly different (12-15 x 4.5-6.5  $\mu\text{m}$ ) and hyaline; in our case only the immature basidiospores were hyaline. The material L. Guzmán-Dávalos 9904 developed a very thick and black crust over some parts of the pileus; while in the type specimen of *Ganoderma areolatum*



the cuticle is almost uniform over all the pileus. The specimen J. Pérez-Ortiz 1016 does not present this cuticle; moreover it has a very thick margin. Our guess is that the old materials might develop a thick and black cuticle, similar to *Ganoderma* and *Fomes*. In all cases, the Mexican materials studied here were confused with *Ganoderma*. Nevertheless, the macromorphological features that make them different from *Ganoderma* are the irregular, thinner walled and bigger sized pores in *N. floccosus*. Although, *N. floccosus* has black cuticle, pale context and spongy basidiomata as do some of the temperate species of *Ganoderma* (i.e. *G. carnosum* Pat., *G. oregonense* Murrill, *G. tsugae* Murrill and *G. valesiacum* Boud.), these have a peculiar shiny basidiomata and a more fragile cuticle.

The type material of *Ganoderma areolatum* presents remarkable features different from subgenus *Ganoderma*. First of all, *Ganoderma* has basidiospores double-walled, with inter-walled pillars and apical germ pore. The crust in *G. areolatum* is not formed by cuticle cells, but by skeletal hyphae as in subgenus *Elfvingia*. However, in *Elfvingia* the skeletal hyphae are arboriform. After a bibliographical review (Murrill 1912, Corner 1983, Gilbertson & Ryvardeen 1986, Núñez & Ryvardeen 2000, Ryvardeen 2000) and the study of specimens of *Navisporus*, we concluded that the type specimen corresponds with *N. floccosus*, meaning *G. areolatum* is a synonym of *N. floccosus*.

Ryvardeen & Johansen (1980) and Gilbertson & Ryvardeen (1986) described the basidiospores as fusoid or navicular. In the studied materials, we observed that the basidiospores have only the apical side subacute or acute; this was also observed in the specimen of *N. sulcatus*.

*Navisporus sulcatus* (Lloyd) Ryvardeen,  
Nordic J Bot. 3(3): 412, 1983.

Figs. 11-12, 17-19

= *Trametes sulcata* Lloyd, Mycol. Writ. 7: 1146, 1922.

The macroscopic features of *Navisporus sulcatus* are as described by Gilbertson & Ryvardeen (1986).

**Contextual trama with dimitic hyphal system;** generative hyphae scarce and collapsed, thin-walled, with conspicuous clamps, yellowish, difficult to observe; skeletal hyphae 2.4-5 µm diam., thick-walled (0.4-2.4 µm), non-septate, generally unbranched to moderately branched, yellowish to yellow, dextrinoid. **Hymenophoral trama with dimitic hyphal system;** generative hyphae 2.4-2.8 µm diam., thin-walled, with conspicuous clamps, yellowish, scarce, difficult to observe; skeletal hyphae up to 4 µm diam., thick-walled, non-septate, sometimes branched, yellowish to yellow, dextrinoid. **Basidiospores** 9-11.6 x 4.4-6.2 µm, Q = 1.82-2.82, oblong to cylindrical, amygdaliform to subcylindrical, with the apical side acute to subacute, without germ pore, smooth, with conspicuous apicule, yellowish to golden-yellow; thick-walled (up to 1 µm), negative in Melzer's reagent. **Basidia** 20-20.6 x 8.8-9 µm, clavate, hyaline to yellowish, tetrasporated, abundant. **Cystidia** or other sterile hymenial elements not observed.

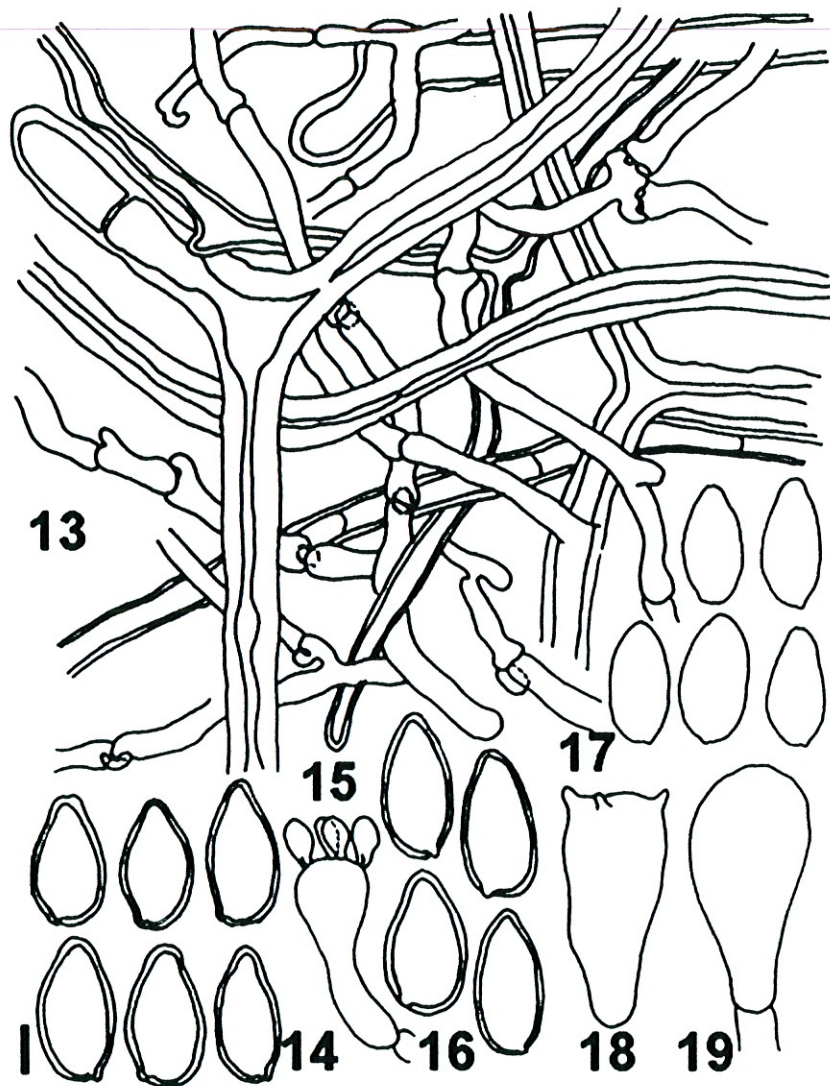
**Substrata** - On *Manihot*.

**Habitat** - Tropical forest.

Specimen studied. Brazil, State of Roraima, Municipality of Boa Vista, Território do Roraima, vicinity of Auaris, 800 m, 31 July 1974, G.T. Prance, O. Fidalgo, B.W. Nelson & J.F. Ramos s.n. (O).

**Remarks** - The studied specimen coincides with Gilbertson & Ryvardeen (1986) and with Gibertoni et al. (2004), except that in this material the duplex context





Figs. 13-19. 13-16: Microscopic features of *Navisporus floccosus*. 13-15: holotype of *Ganoderma areolatum*; 13: elements from the crust, 14: basidiospores, 15: basidium. 16: basidiospores (J. Pérez-Ortiz 1016). 17-19. Microscopic features of *Navisporus sulcatus* (G.T. Prance, O. Fidalgo, B.W. Nelson & J.F. Ramos s.n.); 17: basidiospores, 18: basidium, 19: basidiolum. Scale bar = 4  $\mu$ m.

is not very evident. The species was recorded from south Brazil, State of Paraná (Rajchenberg & Meijer 1990), but no details about its morphological features and ecology were provided. Because of its morphological features the species may be confused with *Corioloopsis*, *Fomes* or *Trametes*, among many others; maybe this confusion is one of the reasons of its rarity.

#### Acknowledgments

Adriana de Mello Gugliotta from Instituto de Botânica da Secretaria de Estado do Meio Ambiente, São Paulo and Gastón Guzmán from Instituto de Ecología, Xalapa, kindly reviewed the manuscript. We are grateful to the curators of NY, O and ENCB, who kindly proportioned the materials from the study. Thanks are due to Universidad de Guadalajara (projects 34961 and 62935 from CA-23, and Fondos Concurrentes), CONACYT (project CONACYT-SEP-2003-C02-42957) and PROMEP (project 103.5/03/2580). The first author thanks Oslo University for a grant to visit O herbarium. She also thanks COLCIENCIAS and Universidad Tecnológica del Chocó for economic help for her Doctoral studies and to Dr. G. Guzmán for his support in the first steps in her studies in fungi.

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## CAPÍTULO II

### ANÁLISIS FILOGENÉTICOS CON DATOS MORFOLÓGICOS Y SECUENCIAS DE DNA

#### RESUMEN

Muchos caracteres morfológicos, usados tradicionalmente en la determinación de especies, no han podido ser usados en el análisis filogenético ante la imposibilidad de codificarlos de manera correcta. En la actualidad, los análisis filogenéticos en *Ganoderma* de secuencias de DNA y otros marcadores, como isoenzimas, han permitido concluir que el género es monofilético y de origen reciente y que se encuentra en diversificación (Moncalvo *et al.* 1995, Moncalvo & Ryvarden 1997, Gottlieb *et al.* 2000). Esta diversificación reciente de *Ganoderma* es evidente en la gran plasticidad del basidioma y de las células del pileipellis, en contraste con la gran uniformidad de las basidiosporas.

En este capítulo se definen caracteres morfológicos útiles para el análisis filogenético. Algunos fueron usados por Moncalvo *et al.* (1995), Gottlieb & Wright (1999a, 1999b) y Gottlieb *et al.* (2000). Los estudios moleculares en *Ganoderma* se han realizado con secuencias de la región interna transcrita (ITS), subunidad grande (LSU) y subunidad pequeña (SSU) del DNA ribosomal (DNAr), así como el gen de la manganeso superóxido-dismutasa (Moncalvo *et al.* 1995a, 1995b, Moncalvo 2000, Smith & Sivasithamparam 2000, Hong & Jung 2004).

En este trabajo el análisis filogenético con datos morfológicos incluyó 38 especies de *Ganoderma*, dos de *Amauroderma*, tres de *Humphreya*, dos de *Magoderma* y seis especies del grupo externo. Se encontró que las basidiosporas son una fuente de caracteres que no había sido completamente aprovechada. Entre ellos se proponen la amplitud de los pilares, truncamiento del ápice y forma como se proyectan los pilares. Para el análisis filogenético con datos moleculares, sólo 33 secuencias del ITS del DNAr pudieron ser incluidas, 31 de *Ganodermataceae* y dos del grupo externo. Este capítulo comprende dos manuscritos de artículos en revisión con los resultados de los análisis filogenéticos con datos morfológicos y moleculares, respectivamente.

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## CAPÍTULO II, PARTE A

### ***Ganodermataceae (Fungi, Basidiomycota): how many genera are there? A morphologic analysis***

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#### **Abstract**

In taxonomic treatments of *Ganodermataceae*, the family has been segregated into eight genera, from which only four have been accepted. Modern phylogenetic analyses with molecular data support two clades in *Ganoderma*, which correspond to *Elfvigia* and *Ganoderma* subgenera, but taxa of the other genera of *Ganodermataceae* have not been included in these molecular studies. Analyses with morphological data have not been conducted. In this study, a phylogenetic analysis of 51 taxa and 45 morphological characters was performed using maximum parsimony in PAUP 4.0b10. As a result, 2304 most parsimonious trees were found of 216 steps. The consensus tree was not fully resolved, but seven clades were supported with a bootstrap value above 50%. The analysis suggests the segregation of *Amauroderma*, *Elfvigia*, *Ganoderma*, *Haddowia*, *Humphreya* and *Tomophagus*. Being *Elfvigia* an independent genus, *Ganoderma*, as was previously considered, is not a natural group.

Keys words. *Amauroderma*, *Elfvigia*, *Ganoderma*, *Haddowia*, *Humphreya*, *Tomophagus*, phylogeny

#### **INTRODUCTION**

*Ganodermataceae* Donk has been defined based on its particular basidiospores with ornamented exosporium and the arboriform or aciculiform hyphal system (Donk 1964, Pegler & Young 1973). In taxonomic treatments of *Ganodermataceae* eight genera have been proposed: *Amauroderma* Murrill, *Elfvigia* P. Karst., *Ganoderma* P. Karst., *Haddowia* Steyaert, *Humphreya* Steyaert, *Magoderma* Steyaert, *Tomophagus* Murrill and *Trachyderma* (Imaz.) Imaz. (Imazeki 1939, Murrill 1905, Steyaert 1972); from which only four have been modernly accepted: *Amauroderma*, *Ganoderma*, *Haddowia* and *Humphreya* (Kirk *et al.* 2001). Some authors considered species of *Haddowia* and *Humphreya* inside *Ganoderma* (Ryvarden 2004, Decock & Herrera-Figueroa 2007). *Ganoderma neurosporum* J.S. Furtado was transferred to *Haddowia* by Teixeira (1992); however, this species was not accepted in *Haddowia* (Ryvarden 2004). *Elfvigia* has been considered as subgenus of *Ganoderma*, supported by phylogenetic analyses with molecular data (Moncalvo *et al.* 1995b, 2000, Smith & Sivasithamparam 2000). *Tomophagus* is a synonym of *Ganoderma* according to

Moncalvo & Ryvarden (1997). On the other hand, some species of *Ganoderma*, as *G. guianensis* Decock & Ryvarden, *G. hildebrandii* Henn., *G. lignosum* Pat. and *G. sculpturatum* (Lloyd) Ryvarden, which have peculiar features, did not have a clear taxonomic position.

Some phylogenetic analyses with molecular data have been made. Nevertheless, these studies have been mainly designed to test the phylogenetic relationships in *Ganoderma*. So, only five of the eight genera proposed in *Ganodermataceae* have been considered (Moncalvo *et al.* 1995a, b, Moncalvo 2000, Smith & Sivasithamparam 2000, Hong & Jung 2004). The problematic species (e.g. *G. guianensis*, *G. hildebrandii*, *G. lignosum*, *G. sculpturatum*) either *Haddowia*, *Humphreya* or *Magoderna*, have not been included. These studies considered the monophily of *Ganoderma* including species with dull and glossy pileus surface, at the moment accepted as *Elfvigia* and *Ganoderma* subgenera, respectively. Recently Moncalvo (2000) and Hong & Jung (2004) found *Ganoderma colossum* (Fr.) C.F. Baker and *G. tsunodae* Yasuda, the type species of *Tomophagus* and *Trachyderma*, respectively, out of the *Ganoderma* core clade and *Amauroderma* as the sister group. *Tomophagus* and *Trachyderma* have been segregated mainly based in the features of the basidiospores and the structure of the pileus surface. Until now, there are doubts if the groups segregated based in morphological features are natural groups.

The molecular analyses are a very important tool in the studies of the phylogenetic relationships, but not always there is material available for its analysis. Ryvarden (1995) and Moncalvo (2000) have discussed about the little utility of the morphological features in phylogenetic analyses; however, none phylogenetic analysis with morphological data has been made. The main objective of this study was to determinate the phylogenetic relationships of the genera in *Ganodermataceae* with morphological data and if the previously proposed genera form natural groups.

## MATERIALS AND METHODS

*Organisms studied.* The materials were collected in the field by the first author, asked in loan, or checked directly in the herbaria. The materials are from BPI, BR, COL, EMBRAPA, ENCB, F, FH, H, IBUG, INBIO, K, NY, O, PC, SP, UPS, VEN and XAL herbaria. More than 400 specimens and 32 types were analyzed (Table 1). An important number of tropical species, which had not been previously studied, was included. The specimens were studied in detail, considering macro- and micro-morphological characters, except *Amauroderma brasiliensis* (Singer) Ryvarden, *A. macrosporum* J.S. Furtado, *Humphreya endertii* Steyaert and *Magoderna infundibuliforme* (Wakef.) Steyaert, which were taken from the literature (Steyaert 1972, Ryvarden 2004). The macroscopic study was made following the methodology established by Teixeira (1946) with some modifications. The microscopic study was made using traditional techniques, with 3 or 10% KOH and Melzer's reagent. The microscopic structures were observed in optic microscopes K-7 Zeiss. When possible, the structures were measured through Axio Vision 4 software in a Zeiss Axioscop 40 microscope. We gave emphasis in characters that had a great potential in the phylogenetic analysis but

were little explored, e.g. granulations, shape and protuberances of pileipellis cells; apex of basidiospores; thickness and disposition of basidiospores pillars.

Table 1. Material studied. Only representative specimens are indicated. IG = Internal group, EG = External group. The herbaria are indicated with capitals before the specimens.

Species		Specimens, Herbaria, Collector
<i>Amauroderma brasiliensis</i> (Singer) Ryvarden	IG	From literature
<i>A. macrosporum</i> J.S. Furtado	IG	From literature
<i>Cryptoporus volvatus</i> (Peck) Shear	EG	IBUG: M.A. Oliva 253, M.G. Torres-Torres 528
<i>Fomitopsis pinicola</i> (Sw.) P. Karst.	EG	IBUG: M.G. Torres-Torres 599, 600, 693
<i>Fomitopsis</i> sp	EG	IBUG: N. Guerrero-Cordero 35
<i>Ganoderma applanatum</i> (Pers.) Pat.	IG	EMBRAPA: A.A.R. de Meijer 386, 962A IBUG: R. Beas 10
<i>G. argillaceum</i> Murrill	IG	NY: F.S. Earle 658 (Lectotype)
<i>G. applanatum</i> (Fr.) Pat.	IG	EMBRAPA: A.A.R. de Meijer 23, 98 IBUG: G. Guzmán 29084
<i>G. brownii</i> (Murrill) Gilb.	IG	NY: V.S. Brown 307 (Lectotype) SP: M. Capelari 1723
<i>G. capense</i> (Lloyd) Teng	IG	NY: P. van der Bijl s.n. (Lectotype)
<i>G. colossium</i> (Fr.) C.F. Baker	IG	UPS: A.S. Oersted s.n. (Lectotype) ENCB: A.M. Suárez 81, F. Ventura 12195 O: N.W. Legon s.n., C. Morsebo s.n., B.M. Spooner 574 XAL: G. Castillo 2803, G. Guzmán 20516, 35708
<i>G. conecinum</i> Ryvarden	IG	O: Ryvarden 16840 (Holotype) EMBRAPA: A.A.R. de Meijer 1194, 2278, 2961, 3253 O: Kyomuhendo s.n., Ryvarden 16840 SP: E.C. Santos 29
<i>G. curtisii</i> (Berk.) Murrill	IG	ENCB: M. Frias-Neve 18, J. Gimete 152-A F: W.C. Coker s.n., W.L. Culberson s.n., F. Drouet, H. Field s.n., D.P. Lewis 3515, P.O. Schallert s.n. (F1119720, F1119722, F1119723) IBUG: L. Guzmán-Dávalos 1723, 7447, J. Mejía-Jiménez s.n., H. Orozco 5, J.A. Pérez de la Rosa s.n., M.G. Torres-Torres 526, 527, 532, 541, 554, A.G. Valenzuela s.n., L. Villaseñor-Ibarra 282
<i>G. dorsale</i> (Lloyd) Torrend	IG	BPI: R.J. Rick s.n. (Lectotype) SP: G. Eiten, L.T. Eiten & H. Kimura 6009, E.C. Santos 76
<i>G. elegantum</i> Ryvarden	IG	SP: M. Capelari & R. Maziera 962
<i>G. guianensis</i> Decock & Ryvarden	IG	O: C. Decock & G. Castillo FG02/02 (Holotype)
<i>G. lignosum</i> Pat.	IG	O: J. Selander 729/1
<i>G. longistipitatum</i> Ryvarden	IG	O: L. Ryvarden 40588 (Holotype)
<i>G. lucidum</i> (Curtis) P. Karst.	IG	H: P.A. Karsten s.n. (Neotype)
<i>G. mexicanum</i> Pat.	IG	FH: S. coll. (Lectotype) SP: C.D.F. Pacheco s.n.
<i>G. multicornis</i> Ryvarden	IG	O: G.J. Samuels s.n. (Holotype)
<i>G. multiplicatum</i> (Mont.) Pat.	IG	SP: M. Capellari & R. Maziero 693, J.S. Furtado s.n., R.H. Marino s.n., R. Maziero s.n., B. Skvortzov s.n.
<i>G. neurosporum</i> J.S. Furtado	IG	BPI: J.S. Stevenson s.n. (Lectotype)
<i>G. nitidum</i> Murrill	IG	NY: P. Wilson 607 (Lectotype, NY; Isolectotype BPI)

<i>G. oerstedii</i> (Fr.) Torrend	<b>IG</b>	O: P. Hind 7 UPS: A.S. Oersted s.n. (Neotype), NY: M.E. Peck s.n. (Lectotype of <i>G. tuberculosum</i> ) IBUG: O. Cárdenas-Hernández 12, A. Gaspar 25, J.J.D. Manzano 480, M.G. Torres-Torres 408, 46, 563, 573, 575
<i>G. orbiforme</i> (Fr.) Ryvarden	<b>IG</b>	UPS: W. Afzelius s.n. (Lectotype) EMBRAPA: A.A.R. de Meijer 1234 SP: O. Fidalgo F-365, A.M. Gugliotta & G.R. Leal 1205, M.A. de Jesús s.n.
<i>G. oregonense</i> Murrill	<b>IG</b>	NY: J.E. Kirkwood s.n. (Lectotype) ENCB: G. Guzmán 4675, 4939 IBUG: S. Acosta 653, M.L. Aguirre-Jones s.n., E. González 282 XAL: G. Guzmán 28886
<i>G. perturbatum</i> (Lloyd) Torrend	<b>IG</b>	BPI: R. Rick s.n. (Lectotype) IBUG: Grupo Ecológico Forestal Tonatiuh s.n. SP: J.P. da Costa Neto s.n. O: Kyomuhendo 05
<i>G. perzonatum</i> Murrill	<b>IG</b>	UPS: F.S. Earle 309 (Lectotype) SP: B. Lowy 22813, J.C. Paim Costa s.n., H. Sotão & coll. 88.18.13, 88.21.06 O: O. Paino-Perdomo 646
<i>G. praelongum</i> Murrill	<b>IG</b>	NY: F.S. Earle & W.A. Murrill 536 (Lectotype, NY, Isolectotype BPI)
<i>G. pulverulentum</i> Murrill	<b>IG</b>	NY: W.E. Broadway s.n. (Lectotype) F: H. Field s.n. (F1119668). O: T. Ferry s.n. SP: D.M. Vital s.n.
<i>G. ravenelii</i> Steyaert	<b>IG</b>	K: H.W. Ravenel 2936 (Holotype) F: W.C. Coker s.n., D.P. Lewis 3428
<i>G. resinaceum</i> Boud.	<b>IG</b>	PC: J.S. Boudier s.n. (Lectotype), Chaffangeon s.n. (Lectotype of <i>G. chaffangeonii</i> ) IBUG: G. Guzmán 11647 SP: M. Mee s.n.
<i>G. sculpturatum</i> (Lloyd) Ryvarden	<b>IG</b>	O: L. Ryvarden 11422
<i>G. sessile</i> Murrill	<b>IG</b>	NY: L.M. Underwood s.n. (Lectotype) IBUG: G. Guzmán 17888 (IBUG). G. Nieves 27-A SP: S & J. Peck s.n., s. coll.
<i>G. sessiliforme</i> Murrill	<b>IG</b>	NY: W.A. Murrill & E.L. Murrill s.n. (Lectotype) ENCB: G. Guzmán 2078 F: E.T. & S.A. Harper s.n. SP: M.H. Homrich s.n.
<i>G. simulans</i> Wakef.	<b>IG</b>	O: T.D. Maitland 556 (Isotype)
<i>G. subfornicatum</i> Murrill	<b>IG</b>	NY: M.E. Peck s.n. (Lectotype) BPI: A.M. Brenes 21375, R. Little 10426 EMBRAPA: A.A.R. de Meijer 962 SP: M. Capelari 1770, G. Hatschbach 20106, U. da Silva-Aragão s.n., O. Yano & J.R. Pirani 5977
<i>G. subincrustatum</i> Murrill	<b>IG</b>	NY: F.S. Earle 176 (Lectotype, NY; Isolectotype, BPI). ENCB: S. Contreras 103, González- Velázquez 556, G. Guzmán 2873, C. Rojas s.n., R. Valenzuela 6429 IBUG: G. López-Damián 49, O. Vargas 316
<i>G. tsugae</i> Murrill	<b>IG</b>	O: K.J. Martin 222



<i>G. tsunodae</i> Yasuda	IG	O: Imazeki 773, T.L. Lui 347
<i>G. weberianum</i> (Bres. & T.Henn. ex Sacc.) Steyaert	IG	ENCB: R. Nava 309 INBIO: E. Fletes 266, 7619 IBUG: T. Cuevas s.n., L. Guzmán-Dávalos 9569, M.G. Torres-Torres 690 SP: A. Milanez & D. Altimari s.n.
<i>G. zonatum</i> Murrill	IG	NY: L.M. Underwood s.n. (Lectotype). F: H.S. Dybas s.n. (F1119728, F1119735, F1119736, F1119737) IBUG: A. Cervantes 1, E. Fanti 514, O. Vargas 13
<i>Humphreya coffeatum</i> (Berk.) Steyaert	IG	SP: G.T. Prance, O. Fidalgo, B.W. Nelson & J.F. Ramos s.n.
<i>H. endertii</i> Steyaert	IG	From literature
<i>H. lloydii</i> (Pat. & Har.) Steyaert	IG	PC: S. coll. (Lectotype)
<i>Magoderma infundibuliforme</i> (Wakef.) Steyaert	IG	From literature
<i>Magoderma</i> sp.	IG	O: R.F. Cain, H.D. Griffin & J.C. Kim
<i>Navisporus floccosus</i> (Bres.) Ryvarden	EG	NY: W.A. & E.L. Murrill 588 (Lectotype of <i>G. areolatum</i> ). ENCB: J. Pérez-Ortiz 1016. IBUG: L. Guzmán-Dávalos 9904 O: C. Decock s.n.
<i>Perenniporia ochroleuca</i> (Berk.) Ryvarden	EG	IBUG: M.A. Oliva 143, L. Guzmán-Dávalos 8274
<i>Polyporus tricholoma</i> Mont.	EG	IBUG: L. Guzmán-Dávalos 7040

*Phylogenetic analysis.* In this study, a phylogenetic analysis of 52 taxa and 45 morphological characters (Tables 1 & 2) was performed using maximum parsimony in PAUP 4.0b10 (Swofford 2000). The analysis includes the eight genera proposed: *Amauroderma*, *Ganoderma*, *Elfvigia*, *Haddowia*, *Humphreya*, *Magoderma*, *Tomophagus*, *Trachyderma* and some species of unclear taxonomic position. In table 1 the names appear as the currently accepted. For example *Haddowia* is represented by *G. neurosporum*. For rooting and character polarization five outgroups were included: *Cryptoporus volvatus*, *Fomitopsis pinicola*, *Navisporus floccosus*, *Perenniporia ochroleuca* and *Polyporus tricholoma*. Heuristic searches were conducted with 1000 replications using TBR, random addition, and MAXTREE set to auto-increase. The support of the branches was calculated using 1000 replications of bootstrap analysis.

## RESULTS AND DISCUSSION

### Character and character states

#### Basidiomata

The basidioma is defined as the organ where basidia and basidiospores are produced. It has received several names, e.g. fruit body, carpophore, basidiocarp, sporocarp, sporophore and receptaculum (Clémenton 2004). All species considered here have a pileate basidioma. In this study, few characters of the basidiomata were codified due to the variation observed. Among *Ganodermataceae*, *Ganoderma* presents the greatest variation of the basidioma,

because weather conditions, locality or microhabitat considerably affect their growing. In the other genera there was less variation.

1. Consistency of the basidioma: 0 = spongy, 1 = woody, 2 = corky-woody, 3 = leathery

The basidioma spongy is mainly formed by generative hyphae. Here *Ganoderma colossium* has a spongy basidioma with a dimitic hyphal system. When the skeletal hyphae are predominant and the hyphal system is trimitic, the basidioma generally have a woody consistency. Corky-woody consistency was defined here for basidioma with di-trimitic hyphal system but an intermediate state between spongy and woody.

2. Light weight of the basidioma: 0 = absent, 1 = present

In some of the specimens studied, the weight of the basidioma does not correspond with the volume. The weight of the basidioma can vary from light to heavy, or something in between. It was difficult to codify all the observed variants, so we consider only the distinctive light weighted basidiomata, present for example in *Ganoderma colossium*, *G. oregonense* and *G. zonatum*. The weight generally corresponds with the hyphal construction, so species with a monomitic system have a consistency generally soft and light weight; nevertheless, the species considered here are those species that have a dimitic or trimitic system but are light weighted, so with no relation with the hyphal system.

### **Pileus**

Generally the pileus is the more conspicuous structure in the fungi and it may be relatively uniform between species. However, in this case because of the little stability inside the species, few characters could be codified.

3. Infundibuliform pileus: 0 = absent, 1 = present

The pileus presents the shape of a funnel. In *Ganodermataceae* only few species have an infundibuliform pileus; this character was registered as present when at least one basidioma has an infundibuliform pileus.

### **Color of the pileus**

The coloration of the pileus in *Ganoderma* seems to be given by a polyphenolic compounds mixture (Núñez & Ryvarde 2000) or by the presence of triterpenes (Rösecke & König 2000). In some species, the surface of the pileus can presents a degradation of colors from light to dark and in different tinges. The color variations can be in agreement with the state of maturity, the weather conditions or the part of the pileus considered, for example becoming darker near the base of the pileus and lighter towards the periphery. Generally, although the basidioma becomes darker when aging, the color degradation is conserved in adults. In other species, it seems that the concentration of triterpenes is higher, given as a result a darker color. That is the case of species with very dark red almost black pileus, as *G. oregonense*, *G. multiplicatum* and *G. nitidum*, that never present color degradation except sometimes a light margin.

4. Degrading from yellow-orange to reddish-brown: 0 = absent, 1 = present

5. Predominantly black-vinaceous: 0 = absent, 1 = present

6. Predominantly dark brown: 0 = absent, 1 = present

7. Beige to light brown: 0 = absent, 1 = present

This character was codified for those species where the triterpenes seem to be absent or the triterpen concentration is low.

8. Glossy surface: 0 = absent, 1 = present

Furtado (1965) defined that the varnished appearance (here as glossy surface) of the basidiomata was due to a substance similar to a lacquer of unknown chemical nature, formed by amorphous exudates secreted by the hyphae of the pilear surface. In subgenera *Elfvigia* the pileus is dull, without brightness. Subgenera *Ganoderma* is characterized by the presence of a glossy surface; however, some species of the *G. lucidum* complex can lose it. It is also very frequent to find young specimens where the pilear surface is very thin and semidull. Some species of the external group present a glossy surface, e.g. *Cryptoporus volvatus* and *Fomitopsis pinicola*.

9. Pileus with resinous substance: 0 = absent, 1 = present

10. Resinous substance soluble in KOH: 0 = absent, 1 = present

The triterpenes of the crust (Rösecke & König, 2000) may be accumulated on the pilear surface to form a resinous mass, which has a dark color (reddish-black to black). This resinous mass can be thin as in the *G. lucidum* complex or thick as in *Elfvigia* subgenus, *Fomitopsis pinicola*, *Humphreya* and *Navisporus floccosus*. When a cut of the pileus is mounted in KOH, the substance may be soluble or insoluble. The first may be easily identified because when the substance is soluble the elements of the pileipellis are easily observed.

### **Pileipellis**

According with Cléménçon (2004) and Kirk *et al.* (2001), the pileipellis is the cellular cortical layer covering the sterile parts of the mature basidiomata, excluding all veil remnants. For *Ganodermataceae*, Furtado (1965) considered the structure of the superficial layer as a derm or as a cortex; the first one for a layer formed by anticlinal hyphae to the pilear surface, and cortex for the structure lacking differentiation. Other studies on the cuticle structure have been made by Corner (1983), Furtado (1965), Haddow (1931), Gottlieb & Wright (1999a, b). We decide to use the general term pileipellis and the classification proposed by Cléménçon (2004) with some exceptions. The genera studied present a diversity of pileipellis types, which were grouped as tomentum, tomentocutis, crustotricoderm, crustohymenidermiform layer and crustohymeniderm. The last two present modified hyphae similar to a hymeniderm. According to Cléménçon (2004) a tomentum is a loose layer with hyphae irregularly arranged, not strongly inflated, sometimes ramified; tomentocutis is a compacted, thin layer of repent, irregularly, sometimes branched, not strongly inflated hyphae; a trichoderm is an intricate layer with erect, irregular to subregular, not or moderately inflated hyphae; hymeniderm is a single layer with pyriform or clavate cells.

In the case of *Ganoderma*, Clémenton (2004) defined the term crustohymeniderm, for the superficial layer with thick-walled cells topped by a resinous substance. We applied the term crustotrichoderm for a trichoderm topped also by a resinous mass. Steyaert (1972) used the term hymenidermiform for a layer similar to a hymeniderm, but with septae at different levels, presented in *Magoderna*. Using the Steyaert's term and following the definition of Clémenton (2004) for a crustohymeniderm, we think that the most appropriate term for the pileipellis of *Magoderna* is crustohymenidermiform layer.

The modified hyphae from the pileipellis of subgenus *Ganoderma* and genus *Magoderna* have been referred by various authors (Haddow 1931, Furtado 1965, Steyaert 1972, Adaskaveg & Gilbertson 1988) as cuticle hyphae, hyphal elements or pileocystidia; in this work they will be treated as pileipellis cells arranged in a crustohymeniderm or a crustohymenidermiform layer.

11. Pileipellis: 0 = tomentum, 1 = tomentocutis, 2 = crustotrichoderm, 3 = crustohymenidermiform layer, 4 = crustohymeniderm

12. Shape of the pileipellis cells: 0 = only cylindrical, 1 = majority claviform, 2 = irregular

13. Protuberances of the cuticle cells: 0 = maximum two, 1 = two to five, 2 = more than seven

The protuberances are projections or prolongations of the hyphal wall generally up to 8  $\mu\text{m}$  long, located apically or laterally. The protuberances are distinguished from short branches because they do not have lumen. Some species present entire cuticle cells (without protuberances) or sometimes with one protuberance. On the other hand, other species have cells with many protuberances; nevertheless, the cuticle cells entire are always observed, intermixed with the ones with many protuberances. For the codification of this character, we considered the maximum number of protuberances although entire cells were present. These protuberances can be or not frequent, depending on the species and location in the pileus. Cells of the cuticle entire or with very few protuberances can be found towards the pileus periphery or/and in immature basidiomata.

14. Wall of the cuticle cells: 0 = thin, 1 = thick

The wall can be thin (< 1  $\mu\text{m}$ ) or thick (> 1  $\mu\text{m}$ ). They can also present many layers projected like several walls; which can be observed through all the structure or only in the apex, that only were codified here as thick wall.

15. Granulations in the cuticle cells: 0 = absent, 1 = present

In some species, the granulations, as small particles especially in the apex, are presented in all cuticle cells. The granulations were considered as present when at least one cell shows them.

16. Hyphae of pileipellis: 0 = inflated, 1 = non-inflated

Besides the modified hyphae mentioned above (see 11 and 12), the pileipellis is composed by generative hyphae, skeletal hyphae or modified skeletal hyphae; which can have an inflated apex. They are recognizable by having a considerably greater diameter than the rest of the hyphae.

## Stipe

The stipe is the stalk that supports the pileus. In *Ganoderma*, many authors have discussed about the utility of this character in the determination of the species, because it can vary according to the position in the substrate, exposure of light, carbon dioxide concentration, among others (Ryvarden 1995, Moncalvo 2000). We think that this is a useful character, considering as a true stipe that who is longer than 5 cm and its tissues are differentiated from the context. Some species can develop a pseudostipe as a continuation of the pileus, which is generally short (less than 5 cm); Steyaert (1980) defined it as pedicel, and we consider it as substipe. The position of the pileus has not been of great importance, because it is very variable inside of the same species. For example, the stipe can be central, lateral or eccentric among individuals of the same specimen.

17. True stipe: 0 = absent, 1 = present

## Context

The context is the sterile part of a basidioma between the tubes and the pilear surface (Núñez & Ryvarden 2000). The traditional taxonomy of *Ganoderma* has used features of the context as dimension, coloration and consistency (Teixeira, 1946). The context comprises good characters because they could be separated as discrete character states.

18. Context structure: 0 = homogeneous, 1 = relatively homogenous, 2 = duplex

19. Beige context: 0 = absent, 1 = present

20. Brown light context: 0 = absent, 1 = present

21. Orangish-brown context: 0 = absent, 1 = present

The context structure is classified as duplex, relatively homogeneous or homogeneous. In the duplex context there is an abrupt change of color, with two separate, contrasting colors, one upper generally light-coloured, and a lower darker one close to the tubes, although the shades will vary to some extent. Relatively homogeneous is used when there is an evident color difference between the upper and lower parts but without abrupt color changes; while the homogeneous context is unicoloured. In some cases, there is a very thin darker line just above of the tubes, absent in some specimens, in this case the context was considered as homogeneous. The color reference was followed from Kornerup & Wanscher (1963).

22. Resinous deposits: 0 = absent, 1 = present

In some species of *Ganodermataceae* there are resin-like deposits in the context, not observed in the external group. These deposits are hard and brittle and could be dull or shiny. It seems that their composition is the same as in the pileus. They can be present as continuous bands or discrete bodies, called resinous bands or resinous incrustations, respectively. Steyaert (1967) named it as "melanoid substance" and Ryvarden (2000) described them as "resinous bands". When the resinous deposits were only present near to the base of the pileus like little bands less than 1 cm long, they were considered as absent.

## Basidiospores

Traditionally the basidiospores of *Ganodermataceae* have been defined as double-walled or thick-walled (Heim 1962, Steyaert 1972, Pegler & Young 1973, Corner 1983, Ryvarden 2004). The basidiospores have a perisporium, exosporium and endosporium. The terms used for the basidiospore wall were according to Cl  men  on (2004). The double-wall corresponds to the perisporium and the exosporium. We only codified the perisporium and exosporium, which are homologous characters in the basidiospores of the Basidiomycetes. The perisporium is defined as a gelatinous to slimy, colorless, often continuous layer that may become disrupted. The exosporium forms the ornamentations. All *Ganoderma* basidiospores have a thin, hyaline to yellowish and smooth or rugose perisporium.

The exosporium is ornamented with projections (pillars) or ornamentations like crests or ridges (Haddow 1931, Furtado 1962, Pegler 1973, Steyaert 1977). Some authors have described these projections as spines or echinules (Patouillard 1889, Steyaert 1977). This unique character is only present in the *Ganodermatacea* family. The basidiospores have been widely used as an important taxonomic character, which include size, shape, thickness wall, color, ornamentation and chemical reaction, among others.

23. Basidiospores dextrinoid: 0 = absent, 1 = present

Some species present basidiospores with a positive reaction with Melzer's reagent; which can be dextrinoid or amyloid, changing to reddish-brown, or grey to blue, respectively. In *Ganoderma* the basidiospores have a negative reaction, but this character is considered for the external group.

24. Basidiospores globose: 0 = absent, 1 = present

25. Basidiospores subglobose: 0 = absent, 1 = present

26. Basidiospores widely ellipsoid: 0 = absent, 1 = present

27. Basidiospores ellipsoid: 0 = absent, 1 = present

28. Basidiospores oblong: 0 = absent, 1 = present

29. Basidiospores cylindrical: 0 = absent, 1 = present

There is a great diversity of shape in the basidiospores of *Ganoderma*. The shape is described according to the Q coefficient (length/width) following Bas (1969). According to the conjunction test, the character was divided qualitatively as independent characters; a specimen can present more than one basidiospore shape but the same basidiospores cannot have several shapes.

30. Basidiospores color: 0 = hyaline, 1 = yellowish, 2 = brown

The coloration is given by the accumulation of pigments in the basidiospores with thick-wall (Ryvarden 1990). In *Ganodermataceae* the basidiospores are golden-yellow to yellowish-brown, so codified as 1 or 2, respectively. The variation in the tonality of yellow and brown were not taken in account because it was impossibility to codify. Some species of the external group have thin-walled and hyaline basidiospores.

31. Perisporium: 0 = absent, 1 = present

32. Exosporium: 0 = absent, 1 = present

33. Ornamented exosporium: 0 = absent, 1 = present

The ornamentation of the basidiospores of *Ganodermataceae* is unique inside the polyporoid fungi, because the exosporium forms pillars or crests. The ornamentation is different for each genus.

34. Pillars: 0 = free, 1 = subfree, 2 = anastomosed, 3 = reticulate, 4 = crested ornamentation

The ornamentation of the exosporium appears like individual elements which are observed like dots, or united themselves to form small lines or a perfect reticulum. Several types of this ornamentation were defined for *Ganoderma*: free pillars, subfree pillars, anastomosed pillars and reticulated ornamentation. The free pillars appear as dots on the basidiospore surface, while free dots mixed with short anastomosed to shortly elongated structures are classified as subfree. The term anastomosed is used when more than two pillars are grown together and form an irregular surface, and reticulate is when the ornamentation is present as an almost complete net. This is a constant character inside of the species. Sometimes, species with basidiospores with subfree pillars present also basidiospores with free pillars, in these cases the character was codified as subfree. Crested ornamentation is an irregular reticulum, more strongly developed along of the longitudinal axis.

35. Basidiospores wall: 0 = non-uniform, 1 = uniform

According with Corner (1983), in *Amauroderma* the wall is equally thick all around the basidiospores; while in *Ganoderma* the wall is apically thickened. In the first case, the pillars are of the same size when they are observed in the light microscope. Genera of the external group that have thick-walled basidiospores have an equally thick wall. An equally or uniform thick-walled basidiospores was codified for *Amauroderma* and *Magoderma*, and wall with the apex apically thickened (non-uniform wall) was codified for *Ganoderma* and *Humphreya*.

36. Pillars wide: 0 = up to 0.4  $\mu\text{m}$ , 1 = 0.5-8  $\mu\text{m}$ , 2 = 0.8  $\mu\text{m}$  or more

When the surface-view of the basidiospores is observed in the light microscope, the ornamentation wide can be measured. The size of the ornamentation was measured through Axio Vision 4 software in a Zeiss Axioscop 40 microscope. There was enough and discrete variation among species in the wide of the ornamentation that could be codified. This variation was observed inside of the species, but there are intervals of wide for each species. This variation goes from very thin (difficult to precise, up to 0.4  $\mu\text{m}$ ) to very thick (more than 8  $\mu\text{m}$ ).

37. Germ pore: 0 = absent, 1 = present

According with Kirk *et al.* (2001), the germ pore is a differentiated, frequently apical area, or hollow, in a spore wall through which a germ tube may come out. For the species included in this study the germ pore is central. Although, the germ pore in *Ganodermataceae* is particular, we only codified the presence or absence of it. There is some variation in the size of the apex, which was not codified because it is the mirror of the character number 40. In some species, as *G. colossum*, *G. conccinum*, *G. dorsale*, *G. oregonense*, *G. perturbatum*, *G. tsugae* and *G. tsunodae*, the germ pore in mature basidiospores is very difficult to

observe, but could be observed in immature basidiospores, so it was codified as present.

38. Basidiospores apex: 0 = papilla, 1 = obtusely conical, 2 = obtuse, 3 = truncate  
The apex of the basidiospore can always have the same form or in some genera of *Ganodermataceae* it can collapse and change. Here we codified the apex of young and freshly discharged basidiospores, before they collapsed, so in *Ganoderma* the basidiospores are obtusely conical giving an even, ovoid outline. In *Amauroderma* and *Magoderna* the apexes of the basidiospores do not collapse, and are always obtuse (Pegler & Young 1973, Corner 1983).

39. Apex collapsed: 0 = absent, 1 = present

40. Shape of the collapsed apex: 0 = truncate, 1 = subacute

In *Ganoderma* and *Humphreya* the basidiospore apexes collapse in dried specimens giving a subtruncate or truncate apex; but in some species of *Ganoderma*, as *G. colossum*, *G. conccinum*, *G. dorsale*, *G. oregonense*, *G. perturbatum*, *G. tsugae* and *G. tsunodae*, the apex after collapsing is not truncate, but subacute. In these species is very rare to find basidiospores with the truncate apex, nevertheless, some basidiospores may be observed.

### Hyphal system

The hyphal system in *Ganodermataceae* is dimitic or trimitic (Furtado 1965, Corner 1983). It has two particular types of skeletal hyphae that are not common between polyporaceae fungi: arboriform and aciculiform hyphae (Furtado 1965). The arboriform skeletal hyphae have long unbranched hyphae which end in a branched tapering end. According to Furtado (1965) “the branching of the arboriform hyphae varies with the species, but it follows a more or less uniform dichotomous pattern. In some species it is rather difficult to distinguish the broken tapering end of the arboriform skeletal hyphae from the binding hyphae”. The aciculiform hyphae are the typical unbranched skeletal hyphae, but with a sharp tip. The generative and binding hyphae collapse very easily, and for this reason are difficult to observe in dried specimens. From the external group, only *Perenniporia* have arboriform skeletal hyphae. The amount of the branches is variable among species but it was not codified because we did not find a form to codify it in discrete states.

41. Arboriform hyphae: 0 = absent, 1 = present

42. Aciculiform hyphae: 0 = absent, 1 = present

43. Hyphal system: 0 = dimitic, 1 = trimitic

44. Skeletal hyphae color: 0 = hyaline, 1 = colored

### Habitat

45. Growing in temperate zone: 0 = absent, 1 = present

Some species of *Ganodermataceae* are restricted to the temperate zones, many times associated with coniferous forest. This character was codified as present even if the species was also present in subtropical forest.



## Phylogenetic analysis

A data matrix with 45 character and 51 taxa was constructed (Table 3). All characters were parsimony-informative, had equal weight and were unordered. As a result 2304 most parsimonious trees were found of 216 steps, with the following indexes: consistency index (CI) = 0.2778, homoplasy index (HI) = 0.7222, retention index (RI) = 0.6579, rescaled consistency index (RC) = 0.1827. The strict consensus tree was not resolved within subgenus *Ganoderma* (Fig. 1). The majority of the clades had bootstraps values above 50%. The *Ganodermataceae* is a natural group, with a bootstrap value 100%. The sister group of *Ganodermataceae* is *Fomitopsis*. In the strict consensus three natural groups were resolved. A first natural group (A) is composed by five clades: *Elfvigia* with a bootstrap of 100% (clade 1), *Tomophagus* and *Trachyderma* (clade 2) with a low support of 50%, *Haddowia* (clade 3), *Humphreya* with a bootstrap of 100% (clade 4), *Ganoderma sculpturatum* (clade 5) and two species unresolved with unclear taxonomic position: *G. guianensis* and *G. lignosum*. A second natural clade (B) represents *Ganoderma* and the third group (C) is *Amauroderma* and *Magoderma* with a bootstrap of 100%. So, this analysis supports at least five genera of eight previously proposed and one not yet classified (clade 5).

*Elfvigia* (clade 1). This clade is formed by species with dull pileus surface, crustotrichoderm pileipellis, brown context, basidiospores small, with truncate apex, free to subfree and thin pillars. *Elfvigia* was proposed as genus by Karsten (Moncalvo & Ryvarde 1997) to include the species of *Ganoderma* with dull surface; nevertheless it was not accepted as genus but as subgenus (Gottlieb 1999a, b, Moncalvo & Ryvarde 1997). This analysis does not support the monophyly of *Ganoderma* if *Ganoderma* and *Elfvigia* are considered inside of the same genus. So, our findings indicate that *Elfvigia* can be considered as a different genus.

*Tomophagus* and *Trachyderma* (clade 2). *Tomophagus* was proposed as a genus by Murrill (1905), with *T. colossus* (Fr.) Murrill, because of the distinctive features of the basidiomata (spongy and pale context). This is a monotypic genus, which had been accepted as synonym of *Ganoderma* by many authors. In this study, *Ganoderma colossus* was in the same clade with *G. tsunodae*. *Trachyderma* was a genus proposed by Imazeki (Moncalvo & Ryvarde 1997) for *G. tsunodae*, but this had not been accepted as genus. The results obtained here agree with the studies of Moncalvo (2000) and Hong & Jung (2004). The first included *G. colossus* and *G. tsunodae*, which were also in the same clade and outside of the core clade of *Ganoderma*. Hong & Jung (2004) only included *G. colossus*, which was basal to the clade of *Ganoderma*. Here, *G. colossus* and *G. tsunodae* form a clade and are the sister group of *Humphreya* clade. *Ganoderma colossus* and *G. tsunodae* share a pale context and reticulate basidiospores with relatively thick pillars.

*Haddowia* (clade 3). This clade is formed by only one species, *Ganoderma neurosporum*, which is the sister group of *Humphreya* (clade 4). The taxonomic position of *G. neurosporum* is unclear, Teixeira (1962) transferred it to *Haddowia* [*H. neurospora* (J.S. Furtado) Teixeira]. Ryvarde (2004) accepted *Haddowia* but not *G. neurosporum* as member of this genus. Studies including more species of

*Haddowia* will be necessary to know if this is a monophyletic group. The main features of *H. neurospora* are dull pileus surface, pale context with resinous incrustations, and basidiospores with thick and crested ornamentation, and germ pore.

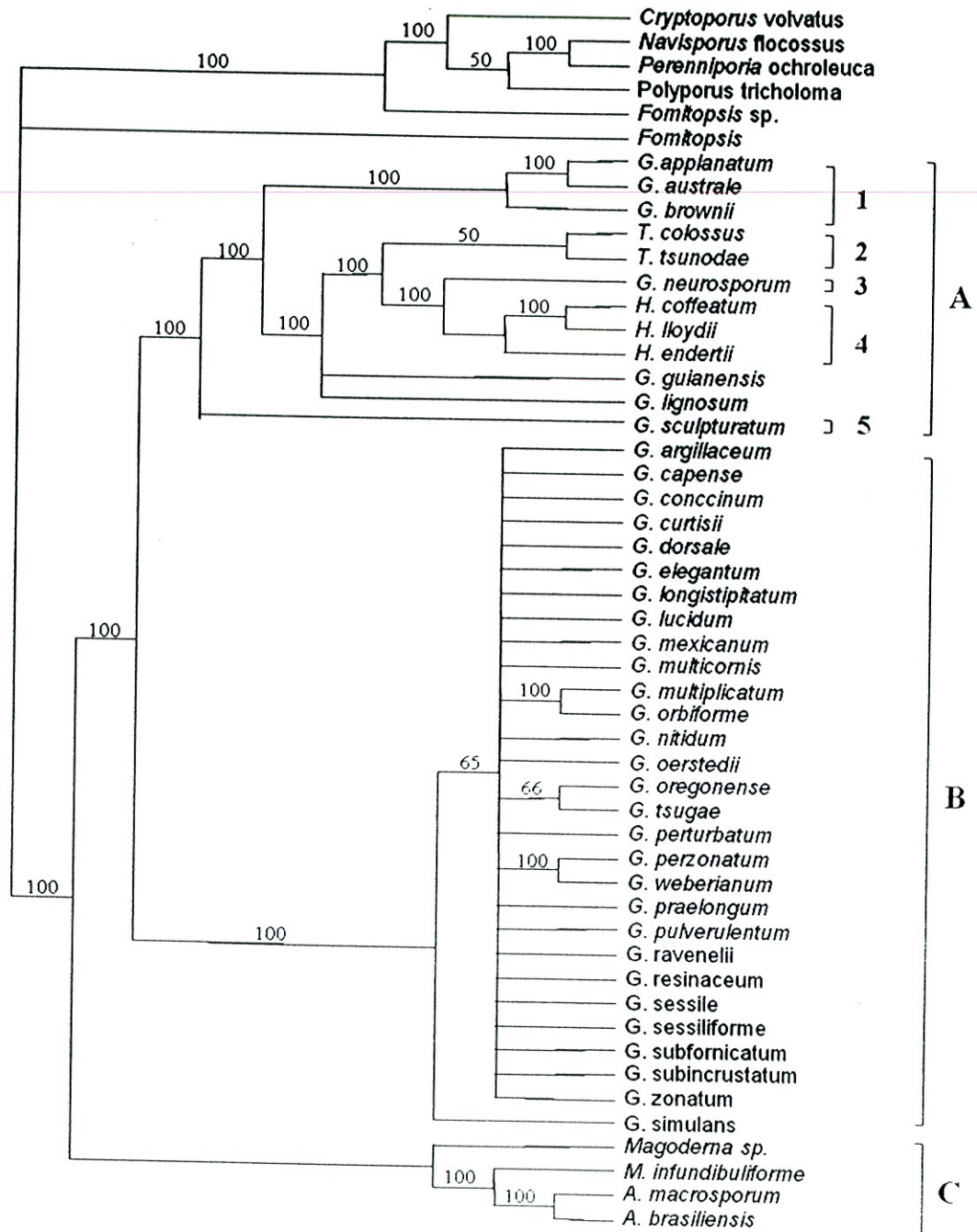


Figure 1. Consensus strict of the 2304 most parsimonious trees with morphological data of *Ganodermataceae* (L = 216, CI = 0.2778, RC = 0.1827). The values of bootstrap are indicated above branches.

*Humphreya* (clade 4). According with our analysis the classification of Steyaert (1972) as genus is supported. This is a natural group characterized by dull pileus

thick and strongly anastomosed to reticulate ornamentation. It is necessary to clarify that *Humphreya* and unclassified species of *Ganoderma* had not been included in previous studies: in this analysis they are in the same clade A along with *Elfvigia*. Ryvar den (2004) did not accepted *Humphreya coffeata* (Berk.) Steyaert, but *Ganoderma coffeatum*.

*Ganoderma sculpturatum* (clade 5). This species were in the base of the Group A; it has particular features: basidioma dull, pale context with resinous incrustations, pileipellis a crustohymenidermiform layer, basidiospores without germ pore, uniform thick-walled, pillars relatively thick and subfree. Morphologically this species is related to *Magoderna* (Steyaert 1972).

*Ganoderma* (clade B). This clade is formed by species with a glossy surface and crustohymeniderm pileipellis, although the infrageneric relationships are not resolved inside this clade. Interestingly, *Ganoderma* and *Elfvigia* subgenera which with molecular data form a monophyletic group (Moncalvo *et al.* 1995b, Moncalvo 2000, Smith & Sivasithamparam 2000, Hong & Jung 2004), did not form a natural group with morphological data.

*Amauroderma* and *Magoderna* (clade C). It is in the base of *Ganodermataceae* which agreed with other studies made with molecular data (Moncalvo *et al.* 1995a, b, Moncalvo 2000, Smith & Sivasithamparam 2000). *Magoderna* was proposed by Steyaert (1972) to include species with dull to semidull pileus surface, pileipellis a crustohymenidermiform layer, and basidiospores without germ pore and uniform thick-walled. The species of *Amauroderma* are characterized by stipitate basidiomata, mainly terricolous habit, dull pileus surface and globose to subglobose basidiospores, without germ pore.

According with our results, the morphological data resolved the phylogenetic relationships of many of the *Ganodermataceae* genera. We include features that in the pass were considered of non-taxonomic value or of limited value to identify *Ganoderma* species (Steyaert 1980, Ryvar den 1995, Moncalvo 2000), e.g. weight of the basidiomata, presence of stipe, color of the context, resinous deposit, shape, protuberances and granulations of the cuticle cells and thickness of basidiospores ornamentation.

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Table 2. Data matrix for 51 species and 45 characters of *Ganodermataceae*.

	10	20	30	40
<i>Cryptoporus volvatus</i>	021000110?	0000100000000010010?	??000?	00101
<i>Fomitopsis</i> sp.	02100001100	??000100000001002010	???	130?00100
<i>Navisporus floccosus</i>	02000110100	??000100010000111010	???	000?00010
<i>Polyporus tricholoma</i>	131000100?	1?001010000000100000	???	020?00000
<i>Perenniporia ochroleuca</i>	021000100?	1?000100010000111010	???	130?10000
<i>Fomitopsis pinicola</i>	01001001100	??00010000000011000?	???	020?00101
<i>Ganoderma applanatum</i>	01000100102	??000001000001002111	1000111010111	
<i>G. australe</i>	01000100102	??0100001100001002111	1000111010111	
<i>G. argillaceum</i>	02110001114	1010001110100001002111	100111010110	
<i>G. brownii</i>	01000100102	??000001000001002111	100111010110	
<i>G. capense</i>	02110001114	1010000010000001102111	1200111110110	
<i>G. colossum</i>	00100001114	1101000100000001101111	1302111111000	
<i>G. conccinum</i>	01010001114	1010012011100001102111	1201111110110	
<i>G. curtisii</i>	02010001114	1010012110100001102111	110111010110	
<i>G. dorsale</i>	02010001114	101101201100001102111	1201111110110	
<i>G. elegantum</i>	02010001114	1010010001100001102111	1100111010110	
<i>G. guianensis</i>	01000100100	??0001000000010011111	101111010110	
<i>G. lignosum</i>	01000100100	??0001000000011011111	100111010110	
<i>G. longistipitatum</i>	02001001114	1010012011000001002111	1201111110110	
<i>G. lucidum</i>	02110001114	1010010100000001002111	1201111010111	
<i>G. mexicanum</i>	02010001114	1010001010100001002111	1000111010110	
<i>G. multicornis</i>	01001001114	2210002110100001102111	1100111010110	
<i>G. multiplicatum</i>	02010001114	2110000001100001002111	1101111010110	
<i>G. neurosporum</i>	02000100100	??001011000010011114	02111011100	
<i>G. nitidum</i>	02101001114	1110000001100001102111	1000111010110	
<i>G. oerstedii</i>	02010001114	1110000001100011002111	1201111010110	
<i>G. orbiforme</i>	02001001114	2210000001000001002111	1101111010110	
<i>G. oregonense</i>	02101001114	1010002110000001102111	110111110110	
<i>G. perturbatum</i>	01001001114	1010010001100011002111	120111110110	
<i>G. perzonatum</i>	01010001114	0011001110000001002111	1000111010110	
<i>G. praelongum</i>	02010001114	1011012110000001002111	1200111010110	
<i>G. pulverulentum</i>	02110001114	1011001011100001102111	1200111010110	
<i>G. ravenelii</i>	02110001114	1010012110000000112111	1000111010110	
<i>G. resinaceum</i>	02110001114	0011000001000001102111	1000111010110	
<i>G. sculpturatum</i>	01001000100	??0001000000010021111	101110?10110	
<i>G. sessile</i>	02110001114	1010002110100001102111	1100111010110	
<i>G. sessiliforme</i>	01010001114	1010001010100001102111	1100111010110	
<i>G. simulans</i>	01001001114	1010000100000001102111	1002010?10110	
<i>G. subfornicatum</i>	02011001114	1110000001100001102111	1000111010110	
<i>G. subincrustatum</i>	02010001114	1010001010100001102111	1201111010110	
<i>G. tsugae</i>	02101001114	0010010100000001102111	1100111110111	
<i>G. tsunoda</i>	01000100100	??00100000001001111	1302111111010	
<i>G. weberianum</i>	01011001114	0011001010100001002111	1100111010110	
<i>G. zonatum</i>	02110001114	1110000001000000112111	1000111010110	
<i>Humphreya coffeata</i>	11000110100	??010100000001001111	1202111011100	
<i>H. endertii</i>	01000100100	??010010000001001111	1202111011100	
<i>H. lloydii</i>	11000110100	??0110100000001001111	1302111011100	
<i>Magoderma</i> sp.nov.	01001001103	00000000100100000111111	110020?11100	
<i>M. infundibuliforme</i>	11001101103	00000000101011001111111	110020?11100	
<i>Amauroderma macrosporum</i>	02001100100	??01001000110000111111	10020?11100	
<i>A. brasiliensis</i>	01000100100	??01010000110000111111	10020?11100	

## CAPÍTULO II, PARTE B

### Relaciones en *Ganoderma* (*Ganodermataceae*, *Basidiomycota*) y géneros afines utilizando secuencias del ITS del DNAr

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#### RESUMEN

Los límites de los géneros en *Ganodermataceae* no están muy bien definidos. Muchos de los géneros propuestos no han sido aceptados y las relaciones filogenéticas inter e infragenéricas no son claras, principalmente debido a que existen confusiones taxonómicas generadas por el uso inadecuado de los nombres. En general, los análisis previos con datos moleculares no han podido ser discutidos a la luz de los caracteres morfológicos. En este trabajo se estudiaron especies de varios géneros de *Ganodermataceae*, principalmente de América tropical, usando el ITS del DNAr con el fin de determinar las relaciones inter e infragenéricas. Aunque no se obtuvo una resolución del cladograma, los clados formados son congruentes con los datos morfológicos. Los clados apoyados con valor de bootstrap sugieren que *Elfvigia*, *Humphreya*, *Magoderma* y *Tomophagus* son géneros válidos.

Palabras claves. *Amauroderma*, filogenia, *Humphreya*, *Tomophagus*

#### INTRODUCCIÓN

Dentro de *Ganodermataceae* Donk se han descrito ocho géneros, de los cuales sólo cuatro de ellos se aceptan actualmente: *Amauroderma* Murrill, *Ganoderma* P. Karst., *Haddowia* Steyaert y *Humphreya* Steyaert (Kirk *et al.* 2001). Los géneros fueron establecidos principalmente con base en las características distintivas de las basidiosporas: grosor uniforme o desigual de la pared y tipo de ornamentación. La determinación de las especies en *Ganodermataceae* ha sido bastante complicada, de allí que muchas especies han sido sinonimizadas. Aunque existen trabajos que han contribuido en gran medida con la definición morfológica de las especies, como los de Gottlieb & Wright (1999a, b), no han sido suficientemente tomados en cuenta. A algunos caracteres morfológicos se les ha dado poco valor taxonómico, porque en muchos casos no se han usado adecuadamente. Esta es una de las razones por las que se ha sugerido que *Ganoderma* es uno de los géneros más complicados dentro de los polyporaceos (Ryvarden 1991). Esta dificultad para trabajar con él se ve reflejada en trabajos que incluyen análisis filogenéticos con datos moleculares, en donde la

misma especie puede estar presente en varios clados, por ejemplo *Ganoderma lucidum* (Curtis) P. Karst. y *G. resinaceum* Boud. (Moncalvo *et al.* 1995a, Smith & Sivasithamparam 2000). Estos dos nombres se han popularizado, considerando un concepto de especie muy amplio y suponiendo que las especies tienen una distribución también muy amplia, lo cual puede no ser completamente cierto. Todavía persisten en *Ganodermataceae* muchos problemas taxonómicos, ya que no se conocen las relaciones filogenéticas entre géneros y entre muchas especies, por lo que permanecen sin una posición taxonómica clara; y algunas especies descritas en otros géneros son a menudo incluidas dentro de *Ganoderma*, dándole a este género un concepto muy amplio, algunos ejemplos son: *Ganoderma amazonense* Weir, *G. hildebrandii* Henn., *G. lignosum* Pat., *G. sculpturatum* (Lloyd) Ryvarden, en los que las relaciones filogenéticas son desconocidas.

*Ganoderma* representa el género con mayor número de especies, mayor plasticidad en los caracteres morfológicos y distribución más amplia de *Ganodermataceae*. En la clasificación actual, *Ganoderma* está integrado por especies con superficie del píleo brillante y opaca; para las que se propusieron los subgéneros *Ganoderma* y *Elfvigia* (Karst.) Imaz. (Moncalvo & Ryvarden 1997), respectivamente. De acuerdo con los análisis con datos moleculares los dos subgéneros forman un grupo monofilético (Moncalvo 2000, Smith & Sivasithamparam 2000, Hong & Jung 2004). A la luz de los caracteres morfológicos, la interpretación de las filogenias moleculares ha sido limitada, porque muchos de los materiales no han sido correctamente determinados; de esta manera, poco se ha discutido acerca de las sinapomorfias que definen los clados en *Ganoderma*. Se han realizado algunos avances en la definición de los géneros de *Ganodermataceae*, por ejemplo Moncalvo (2000) y Hong & Jung (2004) encontraron a *G. colossum* Murrill y *G. tsunodae* Yasuda por fuera del clado de *Ganoderma*, por lo que indicaron que podrían ser segregados como géneros independientes: *Tomophagus* Murrill y *Trachyderma* (Imaz.) Imaz., respectivamente, tal y como lo habían propuesto Murrill e Imazeki (Moncalvo & Ryvarden 1997). Ambas son macroscópicamente muy diferentes de otras especies de *Ganoderma*, por lo que sus hallazgos con datos moleculares estarían confirmados por la morfología. Por otro lado, *Humphreya* Steyaert fue descrito por Steyaert (1972), pero algunos autores lo consideraron como sinónimo de *Ganoderma* (Ryvarden 2004, Decock & Herrera-Figueroa 2007). *Humphreya* no ha sido incluida en los análisis filogenéticos y no se conoce su relación con otros géneros de *Ganodermataceae*.

El objetivo del presente estudio fue encontrar los grupos naturales en *Ganodermataceae* e identificar las sinapomorfias morfológicas que los definen, a través de un análisis filogenético, usando secuencias de la región interna transcrita (ITS) del DNA ribosomal (DNAr). Como en general las especies de América Tropical han sido poco consideradas en los análisis filogenéticos, en este trabajo se trató de incluir un mayor número de ellas.

## **MATERIALES Y MÉTODOS**

*Especímenes estudiados.* Los especímenes incluidos en el estudio fueron proporcionados por los herbarios ENCB, IBUG, INBIO, O y VEN, o fueron recolectados por las dos primeras autoras. Todos los especímenes fueron



estudiados macro y micromorfológicamente, y confirmada su determinación o determinados por la primera autora, excepto el espécimen marcado como *Ganoderma* sp., que erróneamente estaba determinado como *G. stipitatum* (Murrill) Murrill, que no fue estudiado micromorfológicamente. Se extrajo DNA de más de 200 materiales; sin embargo, sólo se obtuvieron secuencias a partir de 32 materiales, los cuales tienen una edad de recolecta entre 1 y 33 años, además se bajaron nueve secuencias de GenBank (Cuadro 1). En total se incluyeron 39 secuencias que corresponden a 25 especies, dos de ellas del grupo externo: *Polyporus tricholoma* Mont. y *Polyporus* sp. Por referencias de la literatura, *Fomitopsis rosea* (Alb. & Schwein) P. Karst y *Cryptoporus volvatus* (Peck) Shear fueron pensados como grupos externos; sin embargo, fueron excluidos de la matriz ante la imposibilidad de alinear sus secuencias.

**Cuadro 1. Especies y especímenes de *Ganoderma*, géneros relacionados y grupos externos usados en este estudio.**

Especies	Colector y número, fecha, herbario	Localidad	GenBank No.
<i>Amauroderma rude</i> var. <i>intermedium</i> J.S. Furtado		Taiwan	X78753 (ITS1), X78774 (ITS2) AJ627583
<i>A. subresinosum</i> (Murrill) Corner			
<i>G. amazonense</i> Weir	F. Fletes 1296, 24 febrero 2000 (INBIO)	Costa Rica	
<i>G. applanatum</i> (Pers.) Pat.		Gran Bretaña	AY884179
	M.G. Torres-Torres 472, 1 julio 2004 (IBUG)	México	
<i>G. carnosum</i> Pat.		Gran Bretaña	AY884175
<i>G. colossum</i> (Fr.) C.F. Baker	A.M. Suárez 81, sin fecha (ENCB)	México	
	C. Morsebo s.n., sin fecha (O)		
	N.W. Legon s.n., sin fecha (O)		
<i>G. concinnum</i> Ryvarden	A.A.A. de Meijer 3252, sin fecha (O)	Brasil	
<i>G. curtisii</i> (Berk.) Murrill	H. Orozco 5, 15 febrero 1994 (IBUG)	México	
	H. Morales-Alarcón 537, 9 octubre 2005 (IBUG)	México	
	M. Mata 1482, 8 agosto 2004 (INBIO)	Costa Rica	
<i>G. lipsiense</i> (Batsch) G.F. Atk.		Finlandia	EF059995
		Finlandia	EF060006
<i>G. lobatum</i> (Schwein.) G.F. Atk.	E. Navarro 5003, 19 julio 2002 (INBIO)	Costa Rica	
<i>G. oerstedii</i> (Fr.) Torrend	M.G. Torres-Torres 407, 27 octubre 2003 (IBUG)	México	
	M.G. Torres-Torres 408, 27 octubre 2003 (IBUG)	México	
	M.G. Torres-Torres y P. Carrillo-Reyes 560, 24 agosto 2004 (IBUG)	México	
	M.G. Torres-Torres 646, 12 agosto 2005 (IBUG)	México	
<i>G. oregonense</i> Murrill	S. Acosta 653, 26 julio 1981 (IBUG)	México	
<i>G. perturbatum</i> (Lloyd) Torrend	Kyomuhendo 5, sin fecha (O)	África	
<i>G. aff. perturbatum</i>	F. Ipulet 1451, 21 abril 2004 (O)	África	
<i>G. perzonatum</i> Murrill	O. Paino-Perdomo 646, 27 diciembre 2000 (O)	República Dominicana	
<i>G. aff. perzonatum</i>	E. Fletes 7275, 8 marzo 2005 (INBIO)	Costa Rica	
<i>G. sculpturatum</i> (Lloyd) Ryvarden	L. Ryvarden 11422, 11 marzo 1973 (O)	África	
<i>G. aff. sessile</i>	L. Guzmán-Dávalos 9918, 21 abril 2006 (IBUG)	China	
<i>Ganoderma</i> sp.	L. Ryvarden 44597, 9-12 marzo 2002 (O)	Ecuador	
<i>G. subformicatum</i> Murrill	R. Liesner y V. Medina s.n., 15 abril 1982, (VEN)	Venezuela	
<i>G. subincrustatum</i> Murrill	González-Velásquez 556, 13 julio 1986 (ENCB)	México	

<i>G. tsugae</i> Murill				DQ206985
<i>G. weberianum</i> (Bres. & Henn. ex. Sacc.) Steyaert	L. Guzmán-Dávalos 9556, 18 septiembre 2004 (IBUG)	México		
	R. Nava 309, 27 agosto 2004 (ENCB)	México		
		Australia		AY569451
<i>Humphreya coffeata</i> (Berk.) Steyaert	L. Ryvarden 45285, 28 octubre 2002 (O)	Belice		
	L. Guzmán-Dávalos 8649, 31 octubre 2001 (IBUG)	Ecuador		
	M.G. Torres-Torres 413, 20 noviembre 2003 (IBUG)	México		
<i>Polyporus tricholoma</i> Mont.	L. Guzmán-Dávalos 7040, 6 diciembre 2005 (IBUG)	México		
<i>Polyporus</i> sp.	E. Fanti 657, 6 diciembre 2005 (IBUG)	México		

*Análisis filogenéticos.* Las secuencias fueron prealineadas en Clustal X (Thompson *et al.* 1997), y luego realineadas a mano en MacClade versión 4.0 (Maddison & Maddison 2000). El análisis filogenético se realizó usando máxima parsimonia, en el programa PAUP 4.0b10 (Swofford 2000). Los espacios fueron tratados como “missing”, todos los caracteres fueron no ordenados y tuvieron igual peso. Se realizó una búsqueda heurística con intercambio de ramas TBR. Se excluyeron del análisis las regiones ambiguas. El soporte para los nodos fue realizado con 1000 réplicas de bootstrap.

## RESULTADOS

Se alinearon un total de 766 nucleótidos después de la adición de espacios, de los que se excluyeron 274 caracteres por estar en regiones ambiguas; 361 caracteres fueron constantes, 39 variables y 82 fueron filogenéticamente informativos. Al menos tres regiones altamente variables fueron identificadas, las cuales fueron excluidas porque su alineación fue muy ambigua. Se obtuvo un total de 3283 árboles más parsimoniosos, de 237 pasos. El índice de consistencia (CI) fue de 0.6245 y excluyendo caracteres no informativos de 0.5412, el de homoplasia (H) de 0.3840 y excluyendo caracteres no informativos de 0.4588, el índice de retención (RI) de 0.3840 y el de consistencia rescalado (RC) fue de 0.4726. En la figura 1 se muestra el consenso estricto de los árboles obtenidos. Con el análisis de las secuencias del ITS no se obtuvo resolución, el cladograma resultó en una politomía pero se recuperaron siete clados con un alto soporte de bootstrap, los cuales se describen a continuación: 1) los tres especímenes de *Humphreya*, uno de Ecuador, uno de México y uno de Belice (como *Ganoderma flaviporum* (Murrill) Sacc. & Trotter), formaron un grupo monofilético, junto con los especímenes de *G. colossum*, un material de *G. sculpturatum*, una secuencia de GenBank determinada como *Amauroderma subresinosum* y *Ganoderma* sp.; 2) las especies del subgénero *Elfvigia*: dos secuencias de Finlandia obtenidas de GenBank, un material de Gran Bretaña y dos de México, con un bajo soporte de bootstrap del 55%, formaron un clado monofilético; 3) *G. tsugae* y *G. carnosum*, dos especies principalmente de zonas templadas, quedaron en el mismo clado con un soporte de 86; 4) otro clado fue el conformado por *G. conccinum*, *G. perturbatum* y *G. aff. perturbatum*; 5) en este clado con un soporte del 88% se agruparon los especímenes de *G. curtisii* con un material determinado como *G. aff. subfornicatum* de Venezuela y con un espécimen de China identificado tentativamente como *G. aff. sessile*; 6) los especímenes de *G. oerstedii* se agruparon con *G. oregonense*; 7) *G. weberianum* formó un grupo con *G.*

*perzonatum* con un bootstrap del 81%. Las especies en las que no se resolvieron sus relaciones fueron: *Amauroderma rude*, *G. amazonense* y *G. subincrustatum*.

## DISCUSIÓN

El ITS es una región del DNA ribosomal que ha sido usada para el estudio de las relaciones infragenéricas en muchos géneros de hongos y específicamente en *Ganoderma* (Moncalvo *et al.* 1995a, b, Moncalvo 2000, Smith & Sivasithamparam 2000). Los primeros estudios filogenéticos en *Ganoderma* fueron realizados por Moncalvo *et al.* (1995a, b), en donde la mayoría de los especímenes analizados fueron de Asia. La relación entre los grupos no fue completamente resuelta; sin embargo, obtuvieron algunos clados. Se destaca el grupo de las especies con superficie no laqueada en uno de los análisis (Moncalvo *et al.* 1995b, fig 9); el cual fue también recuperado en este estudio (grupo 2: *Ganoderma applanatum*, *G. lipsiense* y *G. lobatum*). Tanto en Moncalvo *et al.* (1995b) como en este trabajo las especies con superficie laqueada quedaron agrupadas en varios clados y no se resolvió la relación entre ellos. Smith & Sivasithamparam (2000) realizaron un estudio con ITS para ubicar las especies australianas, en el que incluyeron secuencias de Asia, Europa y América, en el que obtuvieron un consenso estricto resuelto, aunque no muestran el soporte de bootstrap. En su árbol las especies con laca formaron un grupo monofilético con siete clados y las especies sin laca formaron un grupo monofilético, excepto *G. philippii* (Bres. & Henn. ex Sacc.) Bres., una especie con superficie del pileo opaca, que quedó agrupada con especies laqueadas. Un estudio realizado por Torres-Torres & Guzmán-Dávalos (datos no publicados, ver capítulo IIA) con datos morfológicos mostró que *Ganoderma* no es un grupo monofilético cuando se incluyen especies de *Humphreya*, *Trachyderma*, *Tomophagus* y otras especies de afinidad incierta (como *G. neurosporum*). Esto es también evidente en uno de los análisis realizado por Moncalvo *et al.* (1995b, fig 7), en donde *G. formosanum* T.T. Chang & T. Chen, una especie con laca, queda dentro del clado de las especies sin laca.

Smith & Sivasithamparam (2000) y Hong & Jung (2004) tuvieron resolución con el ITS y SSU, respectivamente: en los cuales se recuperó el clado del subgénero *Ganoderma*. Aquí los miembros de ese clado estuvieron repartidos en seis clados (grupos 3 a 7), cada uno con un alto soporte de bootstrap. De acuerdo con los resultados aquí obtenidos se pueden definir siete grupos:

Grupo 1. *Humphreya*, *Tomophagus*, *Magoderma* y *Ganoderma* sp.

El clado está conformado por especies con superficie del pileo opaco o semibrillante, basidiosporas generalmente grandes y con ornamentación compleja y distribución principalmente tropical. En este clado identificamos tres subclados:

Subgrupo 1A. *Humphreya*

De acuerdo con los resultados y con la visión actual de *Ganodermataceae*, éste es un género monofilético, independiente de *Ganoderma*. El clado esta soportado por caracteres morfológicos: todo el basidioma incluyendo el pileo opaco, generalmente de color café, contexto generalmente claro y basidiosporas con una

ornamentación compleja (pilares anastomosados a reticulados). Dentro de este

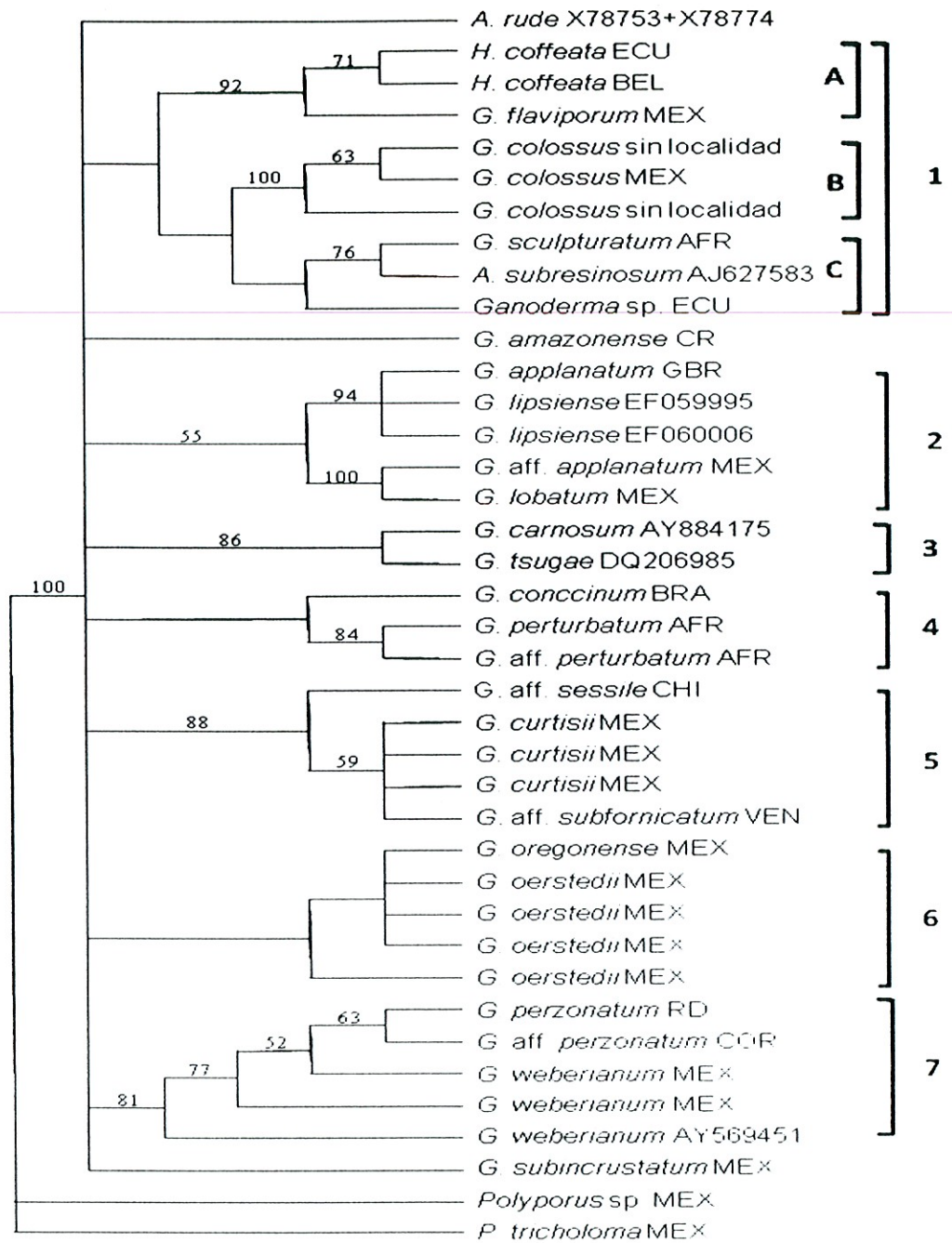


Figura 1. Consenso estricto de 3283 árboles más parsimoniosos usando secuencias del ITS del DNAr. L = 237, CI = 0.6245, RC = 0.4726. El número arriba de la ramas indica el soporte de bootstrap. *Polyporus tricholoma* y *Polyporus* sp. fueron usados como grupos externos. Las letras al lado de cada espécimen indican la procedencia: AFR = África, AUS = Australia, BEL = Belice, BRA = Brasil, CHI = China, CR = Costa Rica, ECU = Ecuador, GBR = Gran Bretaña, FIN = Finlandia, MEX = México, RD = República Dominicana. Las secuencias de GenBank se indican con la clave respectiva. Para explicación de los clados 1-7 y subclados A- C ver discusión.

subclado se pudieron identificar dos grupos: a) los especímenes de Belice y Ecuador que coinciden con las características morfológicas descritas para *Humphreya coffeata* (Berk.) Steyaert, y b) el espécimen de México, que tiene características que coinciden con las descritas para *Ganoderma flaviporum* (Decock & Iturriaga 2007), indicando que los especímenes citados para México por Vázquez & Guzmán-Dávalos (1991) como “*H. coffeatum*” corresponden a *Ganoderma flaviporum*. Decock & Iturriaga (2007) usaron el nombre genérico de *Ganoderma* para las dos especies (*G. coffeatum* y *G. flaviporum*); sin embargo, con base en este análisis y en el análisis con datos morfológicos de Torres-Torres & Guzmán-Dávalos (datos no publicado, ver capítulo IIA), consideramos que *Humphreya* puede conservarse como género y que *Ganoderma flaviporum* es una especie distinta de *Humphreya coffeata*; así se debe hacer una nueva combinación para *Humphreya flaviporum*.

#### Subgrupo 1B. *Tomophagus*

Los especímenes de *Ganoderma colossum* formaron un grupo monofilético; Moncalvo (2000) y Hong & Jung (2004) encontraron éste por fuera del clado de *Ganoderma*, lo cual fue también encontrado por con datos morfológico (Torres-Torres y Guzmán-Dávalos, datos no publicados, ver capítulo IIA). *Tomophagus* fue propuesto por Murrill (Moncalvo & Ryvarden (1997) para incluir a *G. colossum*; sin embargo, autores modernos lo siguen considerando como sinónimo de *Ganoderma*. Existen suficientes sinapomorfias que soportan a *Tomophagus* como un género independiente de *Ganoderma*. *Ganoderma colossum* es un grupo natural definido por un basidioma esponjoso, superficie del pileo semibrillante, contexto claro, basidiosporas grandes y superficie de las basidiosporas reticulada, soportando así el género *Tomophagus*.

Subgrupo 1C. *Magoderma* y *Ganoderma* sp. El clado que incluye a *Ganoderma sculpturatum*, *Amauroderma subresinosum* y *Ganoderma* sp. se caracteriza porque las especies presentan basidioma de color café muy oscuro a negro-rojizo, semibrillante, contexto claro con incrustaciones resinosas, tubos contrastantes con el contexto de color café más oscuro, basidiosporas grandes con pilares relativamente libres, ausencia de poro germinal y ápice obtuso. *Amauroderma subresinosum* presenta características diferentes a las del género *Amauroderma*; asimismo *Ganoderma sculpturatum* no coincide completamente con el género *Ganoderma*. Las dos especies presentan las características descritas por Steyaert (1972) para el género *Magoderma* Steyaert, que no ha sido aceptado por autores modernos. Este análisis soporta esa clasificación y reconoce que este clado corresponde a *Magoderma*. *Ganoderma* sp. estaba marcada en O como *Ganoderma stipitatum*, pero sus características macromorfológicas no coinciden con esta especie; se requieren de mayores estudios, tanto morfológicos como moleculares, para definir la posición de esta especie.

#### Grupo 2. *Elfvingia*

El grupo incluye a *Ganoderma applanatum*, *G. lipsiense* y *G. lobatum* y corresponde al género *Elfvingia* (Moncalvo & Ryvarden 1997); se caracteriza por tener especies con basidioma de color café, superficie del pileo opaca, contexto de color café, basidiosporas generalmente pequeñas, con poro germinal y pilares relativamente libres. Se detectaron dos subclados: tres especímenes de Europa (*G. applanatum* y *G. lipsiense*) quedaron en un mismo clado y las dos especies

americanas quedaron en otro clado, que corresponden *G. lobatum* y *G. aff. applanatum*. *Ganoderma applanatum* tiene una distribución cosmopolita; *G. lipsiense*, que se conoce sólo de Europa, ha sido considerado como sinónimo de *G. applanatum*. *Ganoderma lobatum* parece tener una distribución más tropical.

Grupo 3. *Ganoderma carnosum* y *G. tsugae*

El clado incluye especies descritas de climas templados y restringidas a bosques de coníferas. *Ganoderma carnosum* es conocida sólo de Europa y *G. tsugae* ha sido registrada de Norte América, y está muy relacionada morfológicamente a *G. oregonense*. El clado está definido por el basidioma corchoso, negro-rojizo, contexto pálido, basidiosporas con ápice subagudo y pilares relativamente libres.

Grupo 4. *Ganoderma conccinum*, *G. perturbatum* y *G. aff. perturbatum*

Las especies de este clado no habían sido incluido en análisis filogenéticos anteriores. *Ganoderma conccinum* es una especie recientemente descrita de Colombia (Ryvarden 2000) y *G. perturbatum* fue considerada sinónimo de *G. resinaceum* (Ryvarden 2000). El clado está muy bien definido por caracteres como presencia de estípites, contexto de color café, basidiosporas con el ápice subagudo y pilares anastomosados. Este es un grupo con características muy peculiares dentro de *Ganoderma*.

Grupo 5. *Ganoderma curtisii*, *G. aff. sessile* y *G. aff. subforficatum*

La relaciones filogenéticas de estas especies no eran conocidas, ninguna de ellas había sido incluida en análisis previos, aunque las relaciones con otros grupos no fueron definidas el clado se define por el contexto color café claro con sustancias resinosas, relativamente homogéneo a duplex y basidiosporas con pilares relativamente libres. Las especies presentan distribución tropical. Ryvarden (1985) sinonimizó *G. sessile* bajo *G. resinaceum*; sin embargo, de acuerdo con el estudio morfológico (Torres-Torres y Guzmán-Dávalos, datos no publicados, ver capítulo IF), estas son especies independientes con características propias.

Grupo 6. *Ganoderma oerstedii* y *G. oregonense*

Los cuatro especímenes secuenciados de *G. oerstedii* de México fueron agrupados con *G. oregonense* sin soporte de bootstrap. No es posible definir las sinapomorfias en este grupo; sin embargo, tenemos dudas de que la secuencia de *G. oregonense* sea correcta. Si éste fuera el caso el grupo de las especies templadas (grupo 5) no sería monofilético. *G. oerstedii* no está relacionada con *G. oregonense*, ni desde el punto de vista geográfico ni morfológico.

Grupo 7. *Ganoderma weberianum* y *G. perzonatum*

Tanto en el estudio de Moncalvo *et al.* (1995b) como en el de Smith & Sivasithamparam (2000), las secuencias de los cultivos de *G. weberianum* y *G. microsporum* R.S. Hseu de Asia y Australia quedaron en el mismo clado. De acuerdo con Wang *et al.* (2005) estas especies podrían ser sinónimos. En este estudio los especímenes de *G. weberianum* de México presentaron una secuencia muy similar a la secuencia de Australia del GenBank (AY569451), y formaron un grupo monofilético con *G. perzonatum*. Esta última fue también agrupada con *G. weberianum* en el análisis filogenético con datos morfológicos (Torres-Torres y Guzmán-Dávalos, datos no publicados, ver capítulo IIA). Ryvarden (2000) sugirió que *G. perzonatum* podría ser una variante de *G. resinaceum*, aquí consideramos

que éstas no están filogenéticamente emparentadas y que *G. perzonatum* es una especie diferente. Las características que definen el clado son: contexto pálido, relativamente homogéneo a duplex, células de la cutícula cilíndricas generalmente largas, con incrustaciones y basidiosporas con pilares libres a relativamente libres.

Tres especies no se agruparon: *Amauroderma rude*, *G. amazonense* y *G. subincrustatum*. *Amauroderma rude* tiene hábito terrícola, basidioma estipitado, generalmente de color café y basidiosporas sin poro germinal y ápice obtuso. En estudios previos (e.g. Moncalvo 2000, Smith & Sivasithamparam 2000), *Amauroderma* fue completamente resuelto como el grupo hermano de *Ganoderma*, lo cual no se encontró en este análisis. Estudios posteriores que incluyan más especies de *Amauroderma* podrán confirmar la existencia de este grupo monofilético.

*Ganoderma amazonense* es una especie con superficie del pileo opaca, pero con un contexto claro. Fue clasificada por Moncalvo & Ryvardeen (1997) dentro del subgénero *Elfvingia*; sin embargo, en este estudio quedó por fuera del grupo 2 que corresponde a *Elfvingia*. De acuerdo con Steyaert (1980) y Ryvardeen (2004), *G. amazonense* tiene una distribución Neotropical, pero Corner (1983) además la registró de Asia. Moncalvo & Ryvardeen (1997) indicaron que pueden existir diferencias en las reacciones químicas entre los especímenes de acuerdo a su lugar de origen, lo cual sugiere que probablemente estas variantes geográficas obedecen a especies diferentes.

*Ganoderma subincrustatum* presenta un conjunto de características que la distinguen de los grupos 1 a 7. Morfológicamente es similar a *G. oerstedii*.

Este análisis falló en recuperar la filogenia intergenérica: sin embargo, contrario a las conclusiones de otros estudios (Moncalvo 1995a, b), los resultados obtenidos aquí son altamente congruentes con las características morfológicas. Siete clados pudieron ser definidos, de los cuales tres fueron soportados por un valor alto de bootstrap. Las características de las basidiosporas son sinapomorfias que pueden ser mapeadas en algunos clados de *Ganodermataceae*. Los grupos definidos por estas sinapomorfias son: grupo 1A (basidiosporas con poro germinal grande y pilares gruesos, anastomosados en bandas transversales), grupo 1B (basidiosporas con poro germinal pequeño y pilares relativamente gruesos, reticulados), grupo 1C (basidiosporas sin poro germinal y pilares relativamente gruesos, anastomosados) y grupos 2-7 (basidiosporas con poro germinal y pilares delgados). En los grupos 2-7 la unión de los pilares de las basidiosporas fue muy variable, de libres a anastomosados. Dada la baja resolución encontrada en este análisis, las conclusiones son preliminares.

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## CAPÍTULO III

### ESTUDIO QUÍMICO

#### RESUMEN

El análisis de metabolitos secundarios ha sido utilizado con diversos fines en el estudio de especies, poblaciones o comunidades. En el caso de *Ganoderma* existe un sinnúmero de publicaciones de especies de interés farmacéutico, en donde se han determinado moléculas muy estables con diversas actividades farmacológicas. Estas moléculas bien podrían servir como caracteres taxonómicos para determinar relaciones filogenéticas; sin embargo, los estudios se han restringido a dos o tres especies del género, principalmente a *G. lucidum* y afines. De las muchas sustancias activas que se han aislado de *G. lucidum*, las más sobresalientes son: adenosina, polisacáridos BN3A, BN3B y BN3C, germanio (Ge) y triterpenoides, estos últimos identificados como ácidos ganodéricos A y B (Lin *et al.*, 1991, 1995; Chen *et al.*, 1995; Choi & Sa, 2000; Zhu *et al.*, 2000; Cao & Lin, 2003).

Se realizó una evaluación de triterpenos con 55 especímenes de *Ganoderma* y géneros relacionados y un material de *Cryptoporus*. Se determinó la estabilidad y abundancia de los triterpenos con la finalidad de usarlos potencialmente en el análisis filogenético, además se compara la presencia de estos metabolitos secundarios con los de otras especies reportadas en la literatura con la finalidad de obtener información para su uso posterior. El capítulo consta de manuscritos de artículos en revisión.

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## CAPÍTULO III, PARTE A

### Occurrence of triterpenes in Neotropical species of *Ganoderma* (Fungi, Basidiomycota)

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#### ABSTRACT

It was found that species of *Ganoderma* have a distinctive complex mixture of secondary metabolites, especially triterpenes. To determinate the type of compounds present in the samples, the fruiting bodies of 55 materials from 17 different species of *Ganoderma*, two specimens from one related genera and one species of *Cryptoporus* were examined. The compounds were monitored by TLC, and analyses of the crude extracts were made with GC-MS. More than 70 triterpenes were identified. Some of them have previously been reported for other species of *Ganoderma*, and some were unique for the specimens here studied.

Keywords. America, GC-MS, secondary metabolites, Ergosterol

#### INTRODUCTION

In Asia species of *Ganoderma* (Fungi, Basidiomycota), in special of subgenus *Ganoderma*, have been used in folk medicine since old times for the treatment of various human sufferings, among them: cirrhosis, cancer, fatigue, hypertension, hepatitis and hypercholesterolemia. In the past two decades, there has been a noticeable interest in the study of the secondary metabolites of *Ganoderma*, because of the biologic activities *in vitro* of the extracts of some species (Choi & Sa 2000, Mothana *et al.* 2000, Bao *et al.* 2002, González *et al.* 2002, Luo *et al.* 2002, Wang *et al.* 2002, Iwatsuki *et al.* 2003, Li-Zhen & Zhi-Bin 2003, Lu *et al.* 2004, Lindequist *et al.* 2005, Yang *et al.* 2005, de Silva *et al.* 2006). The extracts of *Ganoderma* are complex mixtures of triterpenes, polysaccharides, proteins, amino acids, nucleotides, alkaloids, steroids, lactones, fatty acids and enzymes (Choi & Sa 2000, Mothana *et al.* 2000, Bao *et al.* 2002). More than 130 triterpenes and related compounds from the fruiting bodies, basidiospores and mycelium have been reported for *Ganoderma*, many of them new and unique for the genus. These have been named for example as ganoderic acids, lucidenic acids, ganodermic acids, ganoderals and ganoderols (Mothana *et*

*al.* 2000, Bao *et al.* 2002, González *et al.* 2002, Luo *et al.* 2002, Wang *et al.* 2002, Iwatsuki *et al.* 2003, Gao *et al.* 2004, Lu *et al.* 2004, Lindequist *et al.* 2005, Yang *et al.* 2005, de Silva *et al.* 2006). The previous studies have focused in the purification and identification of specific compounds present in fractions of extracts. Until now, four to twelve triterpenoid acids have been identified in species of *Ganoderma*. There are no reports to date on the occurrence of these compounds in the crude extracts of *Ganoderma*.

In Asia most of the studied species used have been identified as *Ganoderma lucidum*; nevertheless, this is a commercial name where many species are implicated. There are few studies on the chemical components from American species (González *et al.* 2002). On the other hand, the Gas Chromatography-Mass Spectrometry (GC-MS) is a powerful tool for the analysis of the extracts when many compounds are presented and profiles of the metabolites are required. In fungi, GC-MS methods have been barely used; in the majority of the studies the metabolites had been analyzed by High-Performance Liquid Chromatography (HPLC) of fractions of the extract from the fruiting bodies or mycelium (Hallen *et al.* 2003). In the present study, we analyzed the triterpenes of several species of *Ganoderma* from crude extracts of fresh or stored fruiting bodies using GC-MS. The objective was to make a descriptive study of the number and type of compounds present in samples of different species of *Ganoderma*.

## EXPERIMENTAL SECTIONS

**General Experimental Procedures.** The mass spectrums were carried in a gas chromatograph connected to spectrometry of masses, Agilent Technologies model 6890N. Method: Model of acquisition 60,0 to 800,0 uma. Inserted: Splitless way, gas Helium, heating of 270 °C, pressure 8,77 psi, total flow 24,3 ml/min. 2µl of sample. Column: pressure 8,77 psi, flow 1ml/min, speed 37 cm/seg; time of bullfight 15,58 min; model Agilent 190915-433, HP-5MS, 0,25 30,0 mm x TM x 0,25 mm. Furnace: initial temperature 70 °C (2 min), incline of 33 °C/min, final temperature of 320 °C (6 min). In order to assure that the samples were not contaminated when they were filtrated with the glass syringe, controls were taken and analyzed in the chromatograph. The CH<sub>2</sub>Cl<sub>2</sub> crude extracts were monitored by Thin Layer Chromatography (TLC) in Silice gel 60 PF<sub>254</sub> plate (10 x 20 cm). Spots were detected under a UV light 254 and 365 nm and by spraying 5% phosphomolybdic acid (in ethanol) followed by heating. It was used a movable phase composed by hexane and acetone with a relation of 3:1 and 65:35.

**Fungal Material.** The samples were materials from scientific collections (mycological herbaria) with different ages, degree of maturity and from different regions. The fruiting bodies of 55 materials from 17 different species of *Ganoderma*, two specimens of a related genus and one material of *Cryptoporus volvatus* (Table 1) were included in this study. The materials were recollected and deposited in IBUG herbarium, or provided by the ENCB, IBUG, INBIO, O or XAL herbaria (Holmgren *et al.* 1990).

**Extraction.** The dried fruiting bodies were pulverized in a mill Wiley and sieved through a mesh 40. One gram of the powdered was extracted three times with 30 volumes of dichloromethane (CH<sub>2</sub>Cl<sub>2</sub>) in an ultrasonic bath for 3 hours. The

extracts were filtrated and evaporated to dryness In a Rotavapor Yamato Scientific RE-47 to 100 rpm and 50°C. The concentrated extracts were dissolved in 5 ml of dichloromethane and kept in refrigeration until their use. Previous to injection in the GC-MS, the samples were dissolved in 1 ml dichloromethane. A presence of settled solids was observed after 24 hours storage, to reduce them and avoid problems in the GC-MS, the solution was filtered through a 0.2-µm Millipore filtering unit.

**Table 1. Species and specimens of *Ganoderma*, and related genera.**

Species	Code	Colector & number, date, herbarium	Locality
<i>Cryptoporus volvatus</i> (Peck) Shear	53	G. Martínez s.n., 17 June 1990 (IBUG)	Mexico
<i>G. amazonense</i> Weir	73	E. Fletes 1296, 24 February 2000 (ENCB)	Mexico
<i>G. applanatum</i> (Pers.) Pat.	93	G. Guzmán 7270, 18 February 1971	Mexico
	90	M. Romero 23, 1 julio 2004 (IBUG)	Mexico
<i>G. argillaceum</i> Murrill	83	M.G. Torres-Torres 699, 14 September 2006 (IBUG)	Mexico
<i>G. australe</i> (Fr.) Pat.	84	M.G. Torres-Torres 687, 1 October 2005 (IBUG)	Mexico
	91	M.A. Oliva 253, 15 September 1995 (IBUG)	Mexico
	95	S. coll., s. date (HAJB)	Cuba
<i>G. curtisii</i> (Berk.) Murrill	44	J.A. Pérez de la Rosa, 30 July 1986 (IBUG)	Mexico
	48	O. Rodríguez 1710, 12 August 1981 (IBUG)	Mexico
	49	L. Villaseñor-Ibarra 282, 1 September 1998 (IBUG)	Mexico
	50	O. Vargas 119, 18 September 1988 (IBUG)	Mexico
	54	M.G. Torres-Torres 648, 22 August 2005 (IBUG)	Mexico
	61	H. Morales-Alarcón 537, 9 octubre 2005 (IBUG)	Mexico
	77	M. Mata 1482, 8 agosto 2004 (INBIO)	Costa Rica
	86	M.O. Ocegueda-Reyes 16, 11 June 2001 (IBUG)	Mexico
	87	L. Villaseñor-Ibarra 282, 1 September 1998 (IBUG)	Mexico
	88	O. Vargas 119, 18 September 1988 (IBUG)	Mexico
	89	Moreno 75, s. date (ENCB)	Mexico
	101	L. Guzmán-Dávalos 1723, 6 June 1984 (IBUG)	Mexico
	102	O. Rodríguez 1710, 12 August 1981 (IBUG)	Mexico
<i>G. lobatum</i> (Schwein) G.F. Atk.	56	G. Guzmán 8961, 18 February 1971. (XAL)	Mexico
	68	S. coll., s. date (IBUG)	Mexico
	75	I. López 930, 6 December 1999 (INBIO)	Costa Rica
	76	E. Navarro 5003, 19 julio 2002 (INBIO)	Costa Rica
	85	S. coll., s. date (IBUG)	Mexico
	92	F. Ventura 17311, 9 June 1980 (ENCB)	Mexico
	94	G. Guzmán 8961, 18 February 1971 (XAL)	Mexico
	118	S. coll., s. date (IBUG)	Mexico
<i>G. aff. lucidum</i> (Curtis) P. Karst.	72	L. Guzmán-Dávalos 9921, 21 abril 2006 (IBUG)	China
<i>G. multiplicatum</i> (Mont.) Pat.	74	E. Fletes 1799, 25 July 2000 (INBIO)	Costa Rica
<i>G. oerstedii</i> (Fr.) Torrend	67	M.G. Torres-Torres 696, 28 August 2006 (IBUG)	Mexico
	82	M.G. Torres-Torres 696, 28 August 2006 (IBUG)	Mexico
	96	M.G. Torres-Torres & P. Carrillo-Reyes 560, 24 agosto 2004 (IBUG)	Mexico
	97	G. Guzmán 8749, 22 February 1971 (ENCB)	Mexico
	98	S. coll., s. date (HAJB)	Cuba
	99	S. coll., s. date (IBUG)	Mexico
	116	Cárdenas-Hernández 12 (IBUG)	Mexico
	117	A. Gaspar 25, 20 June 2006 (IBUG)	Mexico
<i>G. oregonense</i> Murrill	45	G. Guzmán 4675, October 1965 (ENCB)	Mexico
<i>G. aff. perturbatum</i> (Lloyd) Torrend	69	L. Guzmán-Dávalos 9919, 21 April 2006 (IBUG)	China

<i>G. aff. perturbatum</i>	70	L. Guzmán-Dávalos 9920, 21 April 2006 (IBUG)	China
<i>G. aff. sessile</i> Murrill		L. Guzmán-Dávalos 9918, 21 abril 2006 (IBUG)	China
<i>G. subincrustatum</i> Murrill	100	C. Rojas s.n. (ENCB)	Mexico
<i>G. weberianum</i> (Bres. & Henn. ex. Sacc.) Steyaert	52	R. Nava 309, 27 agosto 2004 (ENCB)	Mexico
	60	M.G. Torres-Torres 690, 2005 (IBUG)	Mexico
	62	L. Guzmán-Dávalos 9556, 18 septiembre 2004 (IBUG)	Mexico
	80	E. Fletes 7619, 5 June 2005 (INBIO)	Costa Rica
<i>Ganoderma</i> sp. 1	78	E. Fletes 4481, 10 October 2002 (INBIO)	Costa Rica
<i>Ganoderma</i> sp. 2	47	F. Ventura 7314, October 30 1972 (ENCB)	Mexico
<i>Ganoderma</i> sp. 3	81	E. Navarro 4072, 11 November 2001 (INBIO)	Costa Rica
<i>Humphreya coffeata</i> (Berk.) Steyaert	58	A.A.R. de Meijer 3292, 3 February 1990 (O)	Brazil
	79	I. López 3932, 9 October 2002 (INBIO)	Costa Rica

## RESULTS AND DISCUSSION

### Monitoring of compounds in Thin Layer Chromatography (TLC)

The extracts visualized at 356 nm showed at least 16 different compounds, two of which were present at least in 95% of the samples (Fig. 1). The compounds were visualized as fluorescent spots of different colors.

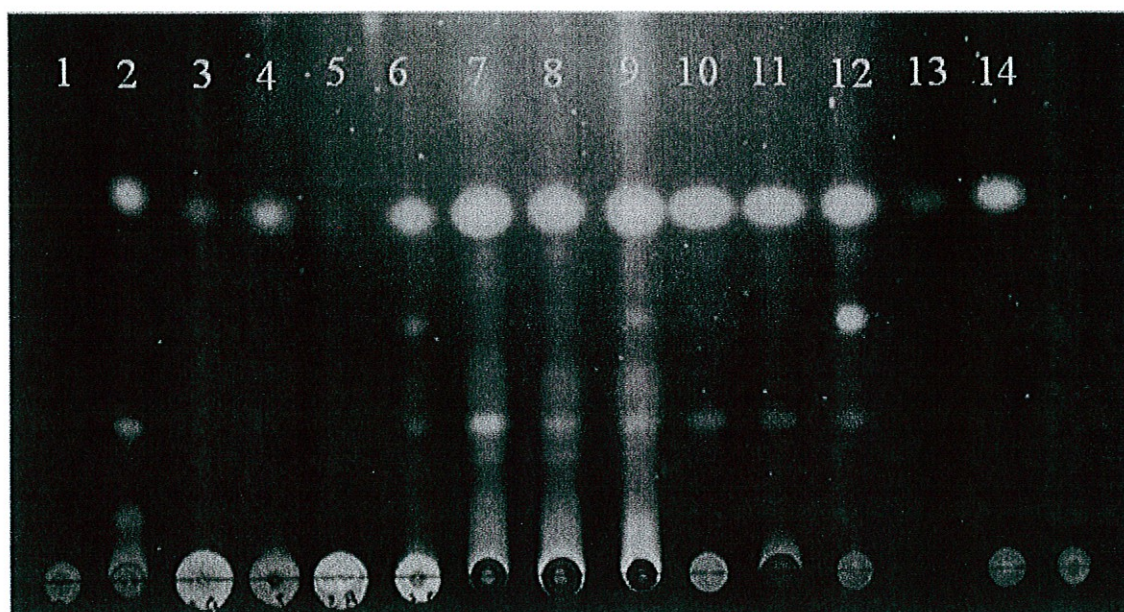


Figure 1. TLC of *Ganoderma*. Two compounds were present in almost all samples. The tracks 1 to 14 shows the chromatography profile of different species.

### Analysis in GC-MS

Approximately 70 compounds of triterpenoid type were tentatively identified through the comparison with the spectrum of the NIST library (Table 2). The compounds were identified based in the probability reported by the equipment software and presence of the majoritary ion in the spectrum. The triterpenes were between 10.907 and 14.3 retention time (rt). In some cases, the retention times were displaced depending of the sample; nevertheless, the Dehydroergosterol 3,5-dinitrobenzoato, 9 (11)-dehydroergosterol, Ergosterol and Ergosta-7,22-dien-3-ol, (3B, 22E) were always in the same retention value, with few exceptions. Different retention times (11.172, 11.278, 11.279, 11.345, 12.032, 12.512) were found for

Ergosterylacetate; it can indicate the presence of similar compounds no yet identified.

Table 2. Identified compounds and their retention time (Rt)

Rt	Compounds	Code of the sample
10.907	Ergosta-5,22-dien-3-ol,	51
10.948-11.039	Stigmasta-4,6,22-trien-3-be-ol	44
11.048	Pregnane-3,11,17,20	63
11.048	Pregnan-20-one...	73,75
10.089	Cholesta-5,7-dien-3-one,4,4-dimethyl	49,63
11.089-11.115	Ergosta-5,22-dien-3-ol,acetate,(3be,22E)	44,46,47,48,63,86,91,94,97,98,117, 118
11.172	Dehydroergosterol 3,5-dinitrobenzoato	49,50,61,65,67,68,76,77,82,83,84,85,86,87,88,89,90,91,92,93,94,95,96,97,98,99,101,102,116,117,118
11.172	9(11)-Dehydroergosterol	44,46,48,54,56,58,63,64,66,69,70,71,72,73,74,84,85,90,91,92,93,94,99,102,116
11.172	5,7,9(11)-Androstatriene, 3-hydroxy-17-ox-	75
11.246	Anthraergosta-5,7,9,22-tetren-ol hexahydrobenzoate	90,93,95,101,102
11.321	Ergosta-5,7,22-trien-3-ol,acetate,(3B,22E)	86
11.246-11.345	Anthiaergostan-5,7,9,16,22-penten	84,86,88,89,90,91,93,95
11.345	oleanoi acid	88,91,96,97
11.403	Anthiergostan-5,7,9,14,-tetraene	86
11.428	B(9a)-Homo-19-norpregna-9(11),9a,16-trien-20-one,3-(dimethylamina)-4,4,14-trimethyl-,(5Be5Al)-	57,68,75,85,91,92,94,93,95,118
11.428	Estrone, benzoate	101,102
11.428	5,16,20-Pregnatriene-3be,20,dioldiacetate	55
11.428	Anthraergosta-5,7,9,(10),14,22-pentene	78
11.428	Carda-4,20(22)-dienoide	96
11.619	Olean-12-ene-3,15,16,21,22,28-hexol,(3be,15al,16al,21be,22al)-	99
11.635	9,10-Secholesta-5,7,10(19)-triene-3,24,25-triol, (3B,5Z,7E)-	117
11.635	4-Chlorocholest-4-en-3-one	98
11.643	Ethyliso-allocholate	92,93,95
11.759	15,17,19,21-Hexatricosatetrayne	93
11.776	Pregn-4-ene-3,11,20-trione,21-hydroxy	79
11.784	5al-Estran-2-one	92
11.792	Cholesta-8,24-dien-3-ol,4-methyl-,(3be,4al)-	82,85,96
11.849	Anthraergostatetraenol	60,82,96,98,99
11.983	4-Norlanasta-17(20),24-diene-11,16-diol-21- oic acid, 3-oxo-16,21-lactone	68
12.024	Anthiergostan-5,7,9,22-tetraen-3-ol	71,83,85,86,88,93,95,97
12.032	12-(9-Anthryl)	44
12.032	Cholest-5-en-19-al	65
12.032	Ergosta -7,22-diene-3,5,6-triol	66
12.078	Cholestanol!7,8-a!cyclobutane,3-methoxy-6-oxo-2'-methylene-	88
12.123	Ergosta 14,22-dien-3-ol(3be,5al,22E)	85,99
12.132	Bufa-20,22-dienoide, 3,14-dihydroxy-, (3á,5á)-	56,68,85,90,92,93,94
12.223-12.248	Ergosterol	44,45,50,51,52,53,57,62,54,64,65,66,69,70,61,67,68,71,72,73,74,75,77,78,79,80,81,82,83,85,84,87,88,89,90,91,92,93,94,95,96,97,98,99,101,102,116,117,118
12.223	7,22-Ergostadienol	59
12.223	Ergosta-7,22-diene-3,5,6-triol	60
12.314-12.348	Ergosta-7,22-dien-3-ol, (3B,22E)	44,45,46,47,48,49,50,54,56,60,68,69,70,75,76,77,84,85,87,88,90,91,92,93,94,95,96,97,98,99,101,102,116,117,118

12.413-12.479	7,22-Ergostadienol	51,67,78,79,82,84
12.479-12.488	4,22-Cholestadien-3-one	47,59,61,71,76,81,85,87,88,89,90,91,92,93,94,99,102
12.488	7,22-Ergostadienone	56,74,78,81,83,84,86,90,91,95,96,97,98,117
12.479	Ergosta-5,8,22-trien-3-ol	53,80
12,479	Ergosta-4,22-dien-3-one	118
12.504	Ergosta-6,22-dien-3-ol	51
12.521	Bufa-20,22-dienolide, 3,14-dihydroxy-, (3á,5á)-	56,68,92,93
12.545	Ergost-7-en-3-ol	44,67,81,82,83,88,89,91,92,117
12.545	Cholest-5-en-19-al,13-acetyloxy	56
12.653	Pregnane'3,11,20,21'tetрил..	46,50,55,70,71,90,91,118
12.661	Ergost-8(14)-en-ol,(3be)	89,91,95,97,98,101,102,118
12.76	ç-Ergostenol	76,77,84,85,86,92,91,93,94,95,96,99,101,118
12.752	Cholest-5-en-19-al,13-acetyloxy	55
12.801	Ergosta-5,24(28)-dien-3-ol	95
12.827	5al-Stigmastane-3be,5be-triol	44
12,843	Ursa-9(11),12-dien-3-ol	89
12.86	Ergosta-5,24(28)-dien-3-ol,(3b)-	85,88
12.86	9,10-Secholesta	92
12,876	Cholesta-8,24-dien-3-ol,4-methyl-,(3be,4al)-	118
12.918	D'Friedoalean'14'en'3'one	92
13.108	Anthraergostatetraenol	83,84,85,88,91
13.299	Ergosta-4,6,8(14),22-tetraen-3-one	61,90,91,92,94,95
13.282	Acetyl atractyloside	44,88,89,90,91,94,95,97,98
13.282	9,19-Cyclolanostan-3-ol	53,89
13,878	8,14-Seco-3,19-epoxyandrostane	88
13,886	D-Homo-24-nor	96
14.192	Ergosta-5,22	47,48,50,52,62

The Ergosterol and Ergosta-7,22-dien-3-ol, (3B, 22E) were the most abundant compounds (fig. 2) in all samples. These compounds served to locate the rest of the peaks in the spectrum and to identify the compounds with more precision. Ergosterol and Ergosta-7,22-dien-3-ol, (3B, 22E) are very common in the majority of the *Basidiomycota*, being the first a component of the cell-wall.



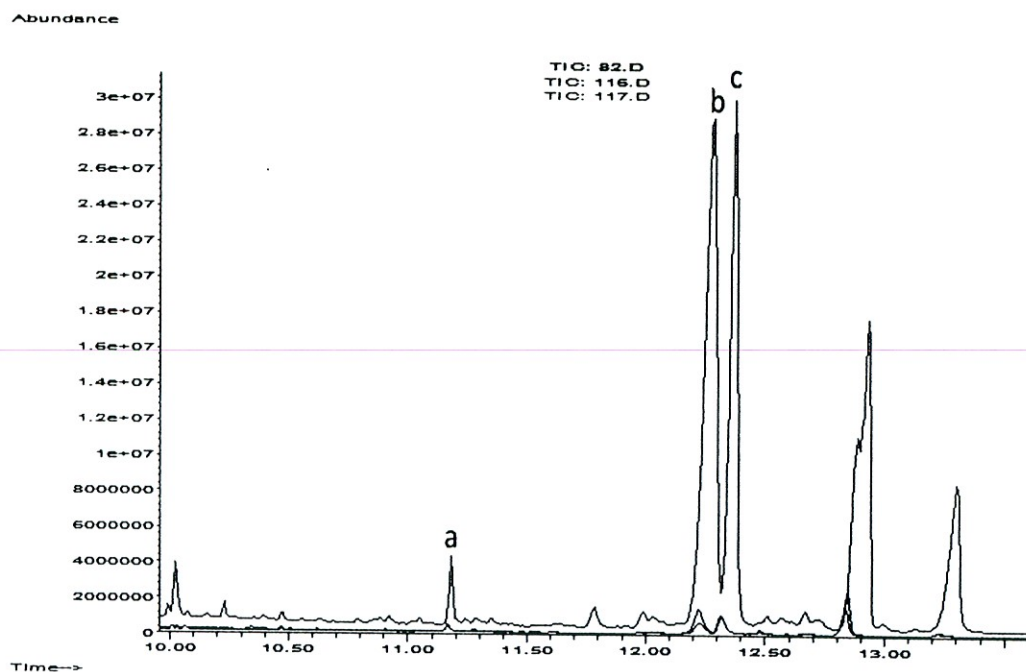


Figure 2. The most common compounds in species of *Ganoderma*: Dehydroergosterol 3,5-dinitrobenzoate (a), 9 (11)-dehydroergosterol (a), Ergosterol (b) and Ergosta-7,22-dien-3-ol, (3B, 22E) (c).

Many of the compounds have been identified in species of *Ganoderma* from Asia and Europe (Keller *et al.* 1997); nevertheless other compounds previously reported were not identified in this study (Luo *et al.* 2002, Silva *et al.* 2006). According with this result, we considered that the triterpenes (ganoderic acids, lucidenic acids, ganodermic acids, ganoderals and ganoderols) are abundant in the species of *Ganoderma*. It is very probable that the triterpenoids composition of the species from America will be different of the species from Asia and Europe. The GC-MS is a tool easy to use to determinate the type and number of compounds present in a sample.

## ACKNOWLEDGMENTS

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## CAPÍTULO III, PARTE B

### **Estabilidad de metabolitos secundarios en *Ganoderma* (*Fungi*, *Basidiomycota*) como fuente potencial de caracteres taxonómicos**

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#### **RESUMEN**

En *Ganoderma* se han aislado e identificado más de 130 compuestos lanostanos tipo triterpenoides, cuyo principal interés ha sido su actividad biológica. Los trabajos previos han revelado una posible especificidad de estos lanostanos en *Ganoderma*; sin embargo, hay muy pocos estudios de su aplicación en el campo quimiotaxonómico y filogenético. Se analizaron 41 muestras de siete especies de *Ganoderma* por GC-MS para determinar la estabilidad de estos compuestos y su posible uso en el estudio de relaciones filogenéticas. Se incluyeron muestras de cuerpos fructíferos de diferentes edades, grado de madurez y provenientes de varias regiones. Se usaron dos métodos de extracción: mecánica y con ultrasonido. Aunque se observó desplazamiento en los tiempos de retención de los compuestos y su concentración en especímenes viejos es baja, aun así los triterpenos son constantes y pueden ser usados en taxonomía.

Palabras claves. Filogenia, lanostanos, taxonomía, triterpenos

#### **INTRODUCCIÓN**

La mayoría de los metabolitos secundarios aparecen como repuestas a condiciones medioambientales, de crecimiento, estrés, entre otros. Por otro lado, dichos metabolitos pueden ser una fuente potencial de caracteres novedosos para los estudios taxonómicos y los análisis filogenéticos en hongos, ya que algunos compuestos son propios de familias, géneros o especies (Frisvad *et al.* 1998). En el caso de los *Basidiomycota*, en ocasiones es complicado contar con material fresco para la extracción de los metabolitos secundarios y esto limita su uso en el estudio sistemático. Una fuente alternativa para la extracción de metabolitos secundarios son los materiales herborizados, pero la conservación de ellos en los herbarios, involucra procesos nocivos para algunos metabolitos, como son el secado y fumigado. Muchas sustancias volátiles e inestables son eliminadas en el proceso de secado o tratamiento, o modificadas después de que los organismos son cortados; pero también muchas de ellas se mantienen estables a través del tiempo.

El análisis de metabolitos secundarios con fines quimiotaxonómicos ha sido desarrollado principalmente en plantas. En hongos, éstos han sido realizados más frecuentemente en *Ascomycota* (Stadler *et al.* 2001a, b, Stadler & Rogers 2004), en donde han demostrado que existen perfiles de metabolitos secundarios específicos de especies y por otro lado, que existen compuestos estables a pesar del tiempo de almacenamiento y tratamiento al que han sido sometidos los hongos. En *Basidiomycota* hay poco trabajos sobre el tema, uno de ellos fue el realizado por Høiland (1983), el cual utilizó especímenes de herbario de *Agaricales*. En *Ganoderma* el análisis de triterpenos por HPLC permitió discriminar entre *G. lucidum* (Curtis) P. Karst. y *G. tsugae* Murrill; además de redeterminar especímenes de *G. resinaceum* mal nominadas con base en caracteres macromorfológicos (Su *et al.* 2002).

En *Ganoderma* se han aislado e identificado más de 130 compuestos lanostanos tipo triterpenoides (Choi & Sa 2000, Mothana *et al.* 2000, Bao *et al.* 2002, González *et al.* 2002, Luo *et al.* 2002, Wang *et al.* 2002, Iwatsuki *et al.* 2003, Li-Zhen & Zhi-Bin 2003, Lu *et al.* 2004, Lindequist *et al.* 2005, Yang *et al.* 2005, de Silva *et al.* 2006) con un interés principalmente farmacológico, ya que se ha demostrado su actividad biológica. Muchos de ellos parecen ser específicos del género, por lo que podrían ser considerados como marcadores químicos; sin embargo, no se ha realizado un estudio que demuestre la estabilidad de estos compuestos en las diferentes especies y tampoco se ha demostrado su estabilidad en materiales herborizados.

Frecuentemente un problema en el análisis de los extractos crudos es la complejidad de la mezcla, ya que ésta posee un sinnúmero de sustancias, lo cual hace difícil su evaluación. Fiehn (2002) y Jonsson *et al.* (2004) discutieron acerca de métodos estadísticos para discriminar muestras complejas en el caso de plantas, que podrían ser aplicados para las muestras de hongos.

El objetivo de este trabajo fue evaluar la estabilidad de los metabolitos secundarios, en particular triterpenos, en siete especies: *G. applanatum* (Pers.) Pat., *G. australe* (Fr.) Pat., *G. curtisii* (Berk.) Murrill, *G. lobatum* (Schwein.) G.F. Atk., *G. oerstedii* (Fr.) Torrend, *G. perturbatum* (Lloyd) Torrend y *G. weberianum* (Bres. & Henn. ex. Sacc.) Steyaert. Los metabolitos estables pueden ser propuestos como una fuente de caracteres para estudios taxonómicos y filogenéticos.

## MATERIALES Y MÉTODOS

**Materiales estudiados.** Los materiales fueron recientemente recolectados y depositados en el herbario IBUG o estaban depositados en los herbarios ENCB, HAJB, IBUG, INBIO o XAL (Holmgren *et al.* 1990). Se estudiaron 41 muestras de siete especies diferentes: *G. applanatum*, *G. australe*, *G. curtisii*, *G. lobatum*, *G. oerstedii*, *G. perturbatum* y *G. weberianum* (Cuadro 1). Para los datos completos (colector y número) de los especímenes ver capítulo III, parte A. Las muestras fueron recolectadas entre 1971 y 2006; se extrajeron por dos métodos diferentes y se analizaron en el Espectrómetro de Gases acoplado a Masas (GC-

MS). Algunas muestras fueron reanalizadas posteriormente para valorar la estabilidad de los compuestos.

**Extracción.** Un gramo de material seco y molido se sometió a extracción mecánica o con ultrasonido en baño maría. En el primer caso, se añadieron 30 ml de metanol, se puso en agitación constante a temperatura ambiente por 4 días, se filtró al vacío y se concentró a 3 ml. Posteriormente, se adicionaron 30 ml de diclorometano (CH<sub>2</sub>Cl<sub>2</sub>) por agitación constante durante 4 días, se filtró al vacío y se concentró a 3 ml. En el método con ultrasonido, un gramo de material seco se extrajo con 30 ml de diclorometano mediante sonicación por 3 horas, se filtró al vacío y se concentró a 3 ml. Las muestras se almacenaron a 4°C hasta su uso.

**Análisis de metabolitos secundarios en GC-MS.** Se utilizó un cromatógrafo de gases acoplado a un detector de masas marca Agilent Technologies modelo 6890N/5973i. Método: Modo de adquisición SCAN de 60.0 a 800.0 uma. Cantidad de muestra inyectada, 2 µl. Columna HP-5MS, 0.25 mm x 30.0 m x 0.25 µm, flujo 1ml/min. Puerto de inyección modo Splitless, gas Helio, flujo total 24.3 ml/min. Horno; temperatura inicial 70 °C (se mantiene por 2 min), rampa de 33 °C/min, temperatura final de 320 °C (mantenida por 6 min). Se analizó 1 ml de muestra previamente filtrado en una membrana de nylon de 13 mm de diámetro y 0.2 µm de diámetro de poro; se utilizó una jeringa de vidrio de 5 ml a la cual se acopló un suinex de acero inoxidable. Se usó un control de lavado de jeringa, el cual fue analizado por GC-MS cada diez muestras.

Cuadro 1. Especies de *Ganoderma* analizadas por GC-MS.

Especies	Código	Fecha de recolecta, herbario	Localidad
<i>G. applanatum</i> (Pers.) Pat.	93	1971 (XAL)	México
	90	2004 (IBUG)	México
<i>G. australe</i> (Fr.) Pat.	84	2005 (IBUG)	México
	91	1995 (IBUG)	México
	95	s. fecha (HAJB)	Cuba
<i>G. curtisii</i> (Berk.) Murrill	44	1986 (IBUG)	México
	48	1971 (IBUG)	México
	49	1998 (IBUG)	México
	50	1988 (IBUG)	México
	54	2005 (IBUG)	México
	61	2005 (IBUG)	México
	77	2004 (INBIO)	Costa Rica
	86	2001 (IBUG)	México
	87	1998 (IBUG)	México
	88	1988 (IBUG)	México
	89	s. fecha (ENCB)	México
<i>G. lobatum</i> (Schwein.) G.F. Atk.	101	1984 (IBUG)	México
	102	1971 (IBUG)	México
	56	1971 (XAL)	México
	68	s. fecha (IBUG)	México

	75	1999 (INBIO)	Costa Rica
	76	2002 (INBIO)	Costa Rica
	85	s. fecha (IBUG)	México
	92	1980 (ENCB)	México
	94	1971, (IBUG)	México
	118	s. fecha (IBUG)	México
<i>G. oerstedii</i> (Fr.) Torrend	67	2006 (IBUG)	México
	82	2006 (IBUG)	México
	96	2004 (IBUG)	México
	97	1971 (ENCB)	México
	98	s. fecha (HAJB)	Cuba
	99	s. fecha (IBUG)	México
	116	1988 (IBUG)	México
	117	2006 (IBUG)	México
<i>G. aff. perturbatum</i> (Lloyd) Torrend	69	2006 (IBUG)	China
<i>G. aff. perturbatum</i>	70	2006 (IBUG)	China
<i>G. weberianum</i> (Bres. & Henn. ex. Sacc.) Steyaert	52	2004 (ENCB)	México
	60	2005 (IBUG)	México
	62	2004 (IBUG)	México
	80	2005 (INBIO)	Costa Rica

## RESULTADOS Y DISCUSIÓN

Se identificaron más de 70 compuestos lanostanos tipo triterpenoides (ver Capítulo III, Parte A) por sus espectros de masas, los cuales fueron contrastados con la librería NIST; con base en las probabilidades e iones característicos que reportaba el software del equipo se obtuvieron los perfiles de compuestos. Aquí se reportaron solamente los picos con una abundancia por arriba de 5000, los picos con abundancias menores fueron generalmente muy variables (y con probabilidades muy bajas), por lo que su presencia es poco confiable. Los compuestos de interés tuvieron tiempos de retención entre 11.089 y 13.282. Se observaron pequeños desplazamientos de los picos de una muestra a otra (Fig. 1); sin embargo, se establecieron intervalos donde el compuesto podía ser encontrado. De esta manera, se determinó que esos picos correspondían al mismo compuesto.

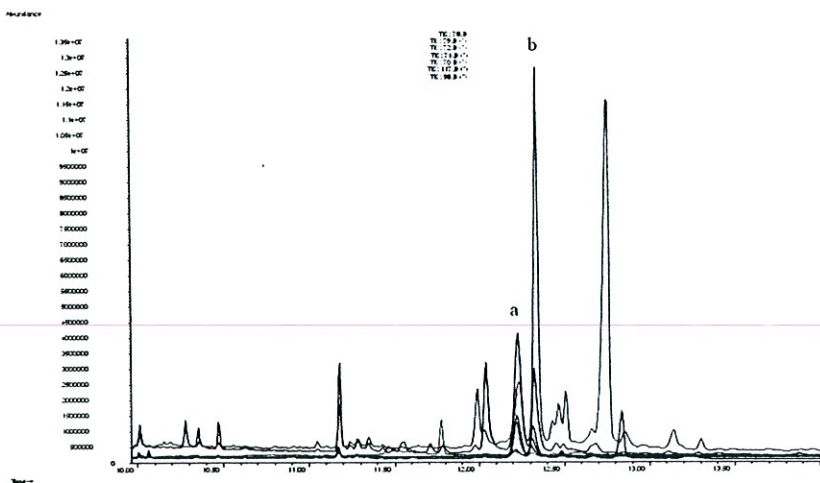


Figura 1. Perfil cromatográfico de siete especímenes de *Ganoderma* en donde se observa el desplazamiento de los tiempos de retención. Nótese los picos más estables y abundantes: ergosterol (a) y ergosta-7,22-dien-3-ol,(3B,22E) (b)

Al comparar muestras procesadas por los dos diferentes métodos de extracción, se observó que los compuestos presentaron generalmente una concentración mayor (expresada como área del pico) cuando la extracción se realizó con ultrasonido. Por otro lado, picos muy pequeños en la extracción mecánica, en donde la identificación del compuesto fue muy variable y no podía se podía asignar un nombre, se detectaron e identificaron en las extracciones con ultrasonido (Figuras 2, 3, 4). De acuerdo con estos resultados, la lisis celular es más efectiva mediante el método ultrasonido, lo cual expone de manera más eficiente los metabolitos secundarios aumentando así la disponibilidad de las sustancias.

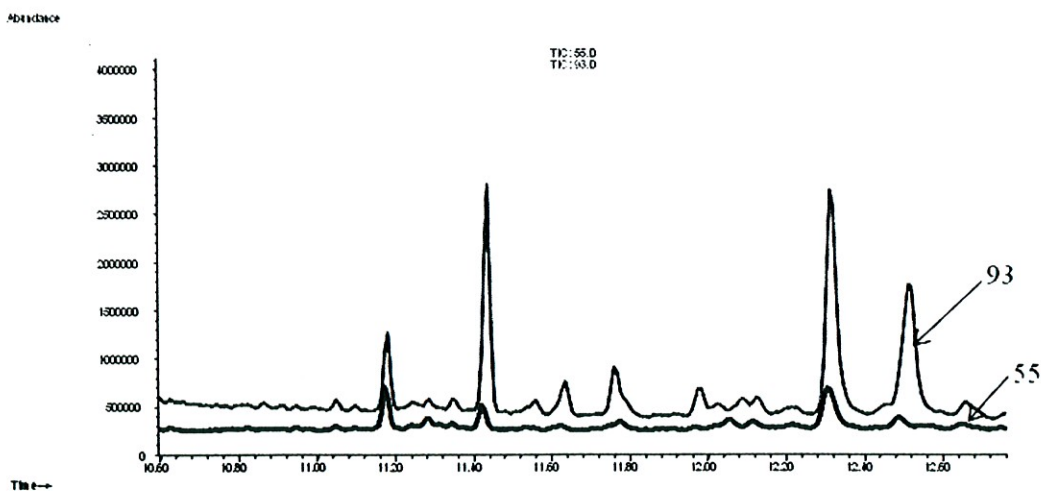


Figura 2. Perfil cromatográfico de *Ganoderma applanatum*. La muestra 93 fue extraída por ultrasonido y la 55 por extracción mecánica.

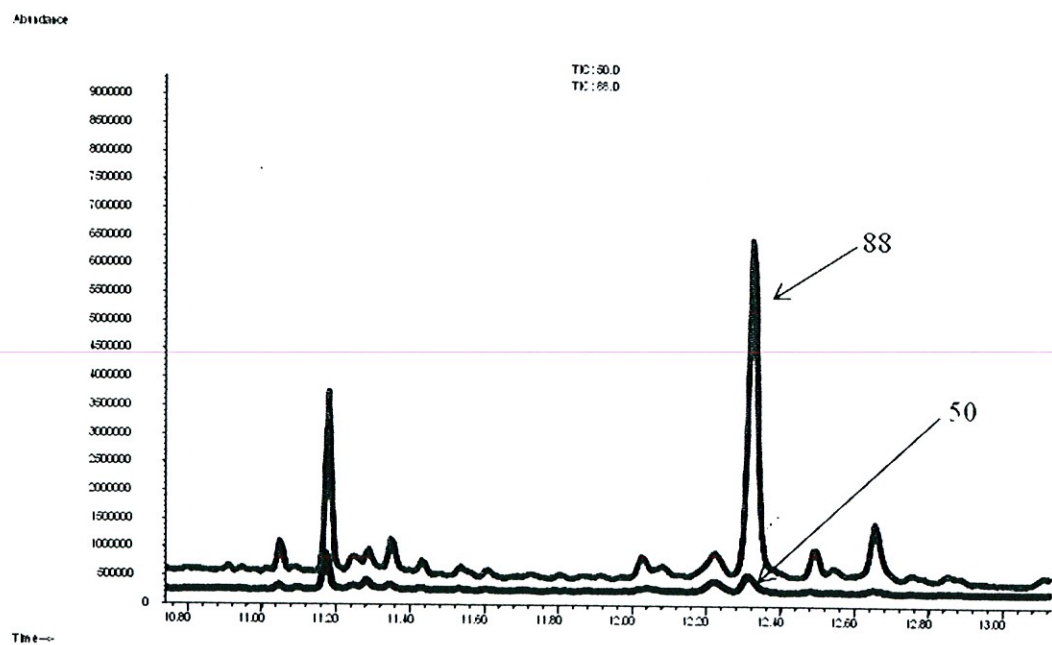


Figura 3. Perfil cromatográfico de *Ganoderma curtisii*. La muestra 88 fue extraída por ultrasonido y la 50 por extracción mecánica.

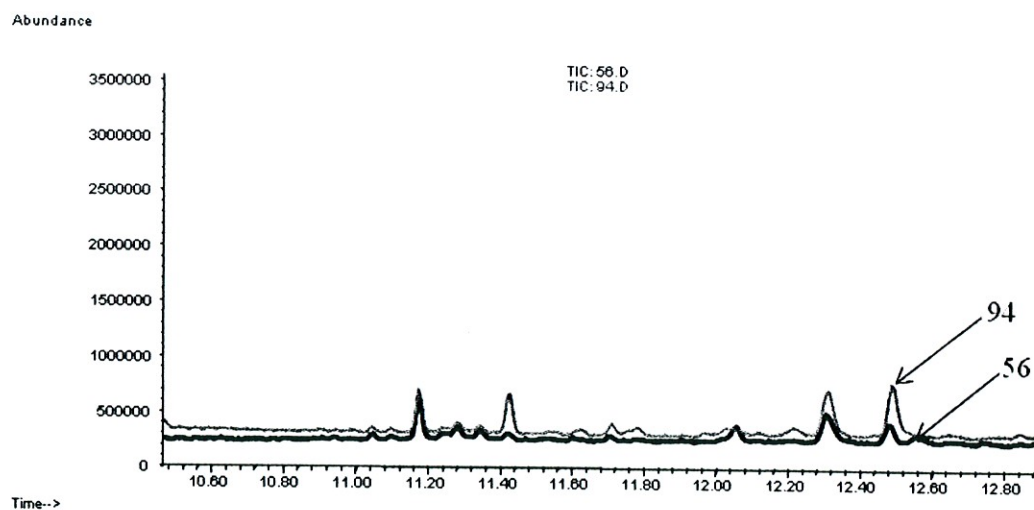


Figura 4. Perfil cromatográfico de *Ganoderma lobatum*. La muestra 94 fue extraída por ultrasonido y la 56 por extracción mecánica.

En los extractos con ultrasonido, se detectaron algunos compuestos que estuvieron presentes independientemente de la edad del espécimen, pero la concentración fue considerablemente más baja en especímenes más viejos (Fig. 5); es el caso de los picos con tiempo de retención (tr) 11.246, el cual corresponde a Anthiaergostan-5,7,9,16,22-penten. Por otro lado, algunos compuestos sólo estuvieron presentes en especímenes recién recolectados; lo cual significa que muy probablemente éstos fueron eliminados mediante el proceso de secado o que su concentración es tan baja que no pudieron ser detectados.



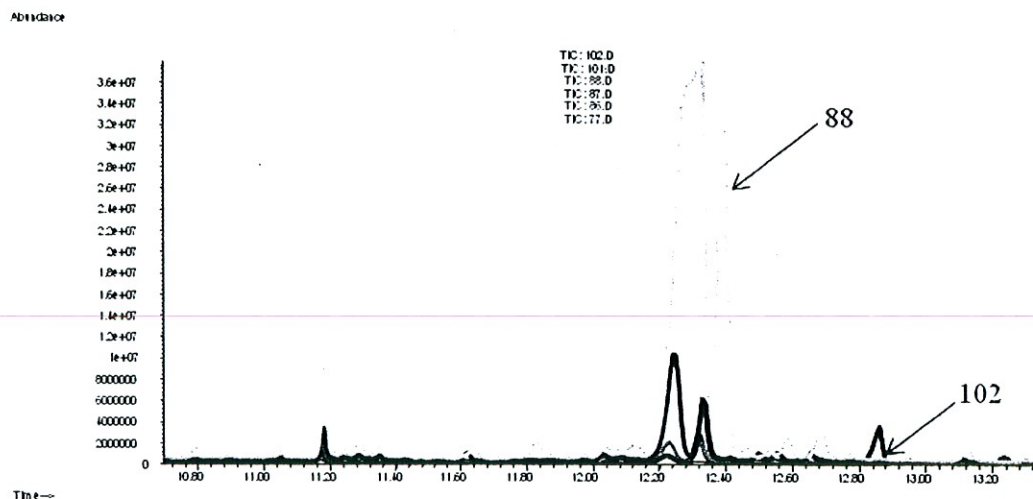


Figura 5. Especímenes de *Ganoderma curtisii*. La muestra 88 es la muestra más reciente y la 102 la más vieja; notése que la concentración de los compuestos es mayor en la muestra más nueva.

Los compuestos con tr más estables fueron seleccionados para el análisis (Cuadro 2). El Ergosterol (tr 12.223) y Ergosta-7,22-dien-3-ol, (3 $\beta$ , 22E)- (tr 12.314) fueron los compuestos más abundantes y que estuvieron en la mayoría de las muestras. Estos han sido reportados para *Ganoderma* (Yuan *et al.* 2006); sin embargo, aunque son estables no son buenos marcadores porque al estar presentes en todas las especies no permiten discriminar grupos. El Ergosterol es parte de la pared celular de los hongos; en este trabajo fue detectado hasta en los especímenes más viejos, aunque su concentración fue muy variable.

Algunos compuestos pueden ser considerados con valor taxonómico; a continuación se dan algunos ejemplos. El Ergosta-5,22-dien-3-ol,acetato,(3 $\beta$ e,22E) (tr 11.089-11.115), Ergosta-5,22-dien-3-ol,acetate,(3 $\beta$ e,22E) (tr 11.089-11.115) fueron detectados sólo en *G. australe*, *G. curtisii*, *G. lobatum* y *G. oerstedii*. El Dehydroergosterol 3,5-dinitrobenzoato (tr 11.172) no se detectó en muestras de *G. perturbatum* y *G. weberianum*. El 9(11)-dehidroergosterol (tr 11.172) se presentó en todas las especies, menos en *G. weberianum*. El Antraergosta-5,7,9,22-tetren-ol hexahidrobenzoate (tr 11.246) es un marcador de *G. applanatum*, *G. australe* y *G. curtisii*. El B(9a)-Homo-19-norpregna-9(11),9a,16-trien-20-one,3-(dimetilamina)-4,4,14-trimetil-(5Be5Al) (tr 11.428) sólo está presente en las especies del subgénero *Elfvigia*: *G. applanatum*, *G. australe* y *G. lobatum*. El Antiaergostan-5,7,9,16,22-penten (tr 11.345) fue detectado en todas las especies, excepto en *G. perturbatum* y *G. weberianum*. El Antraergostatetraenol (tr 11.849) fue encontrado sólo en *G. oerstedii* y *G. weberianum*. El Ergosta-4,6,8(14), 22-tetraen-3-one (tr 13.299) no fue detectado en *G. weberianum* y *G. oerstedii*. Otros compuestos de interés fueron: 4,22-Colestadien-3-one (tr 12.479), 7,22-Ergostadienone (tr 12.488), Ergost-7-en-3-ol (tr 12.545), Ergost-8(14)-en-ol,(3  $\beta$ ) (tr 12.661),  $\zeta$ -Ergostenol (tr 12.76) y Acetil atractiloside (tr 13.282); los cuales fueron también marcadores importantes (Fig. 6). Keller *et al.* (1997) consideraron la posibilidad de que existan marcadores químicos para la identificación de especies de *Ganoderma*; sin embargo, sólo analizaron una especie.





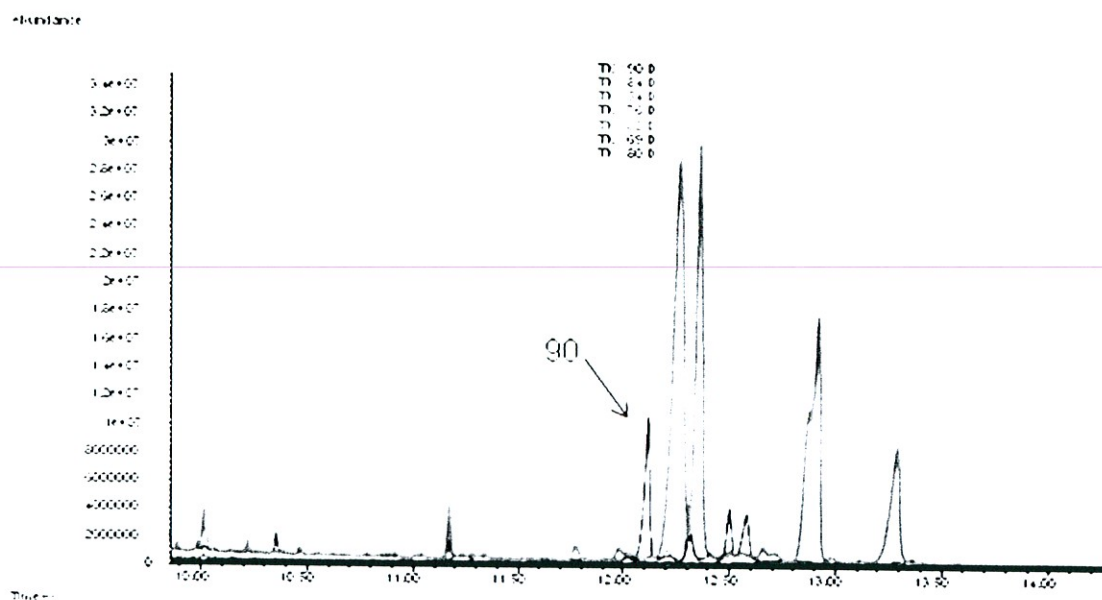


Figura 6. Varias especies de *Ganoderma*, donde se muestran las diferencias en el perfil cromatográfico. Por ejemplo, Bufo-20,22-dienoide, 3,14-dihydroxy-, (3á,5á) (tr 12.132) es característico de la muestra 90.

Las estructuras químicas encontradas sugieren que los compuestos son derivados del Ergosterol, en el cual la cadena lateral cambia, originando los compuestos conocidos comúnmente como ácidos ganodéricos, ácidos lucidénicos, ganodérmico, ganoderales y ganoderoles (Figs. 7) (Mothana *et al.* 2000, Bao *et al.* 2002, González *et al.* 2002, Luo *et al.* 2002, Wang *et al.* 2002, Iwatsuki *et al.* 2003, Lu *et al.* 2004, Lindequist *et al.* 2005, Yang *et al.* 2005, de Silva *et al.* 2006).

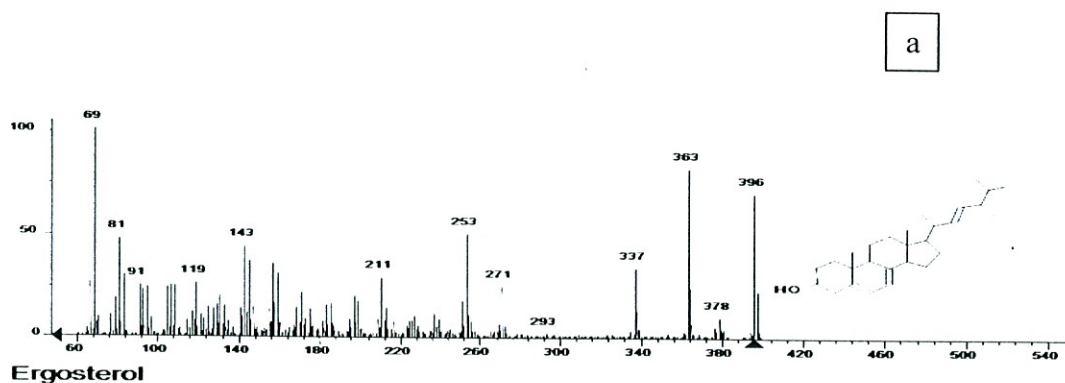


Figura 7a. Estructura de algunos compuestos químicos de interés en *Ganoderma*.

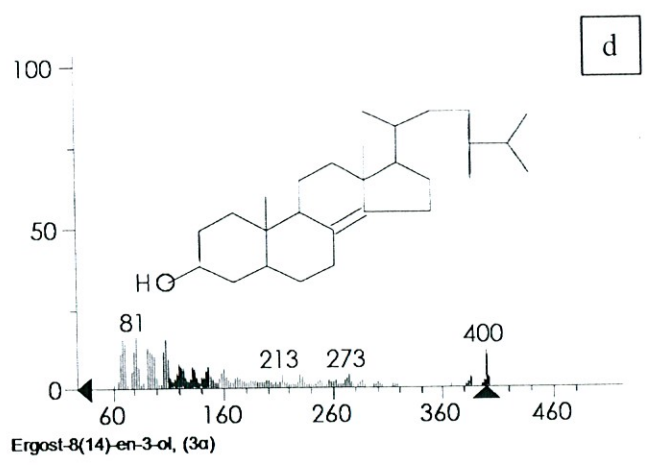
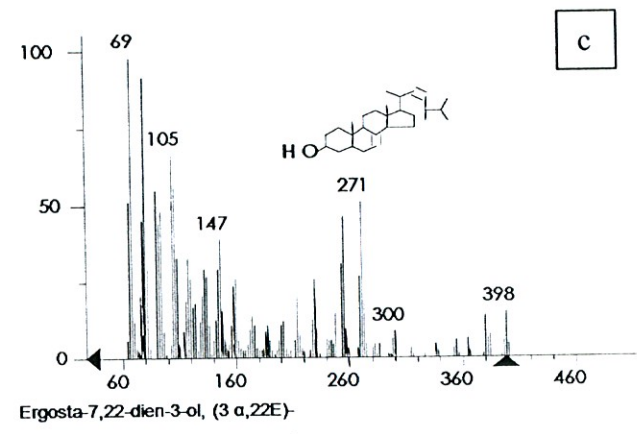
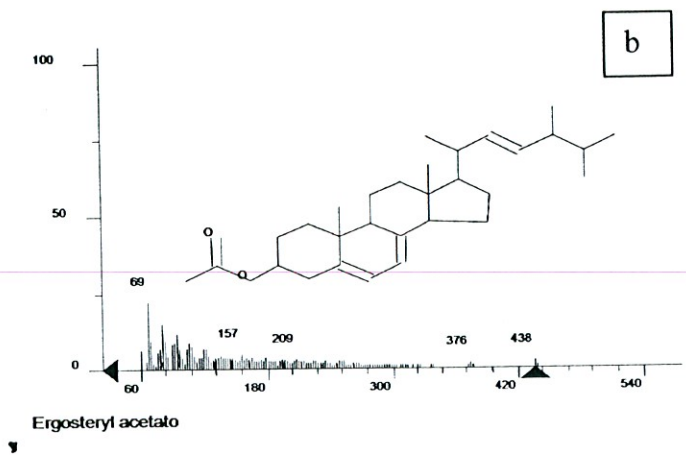
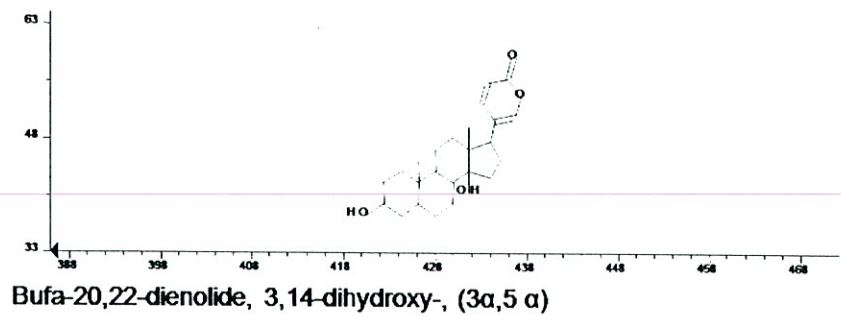
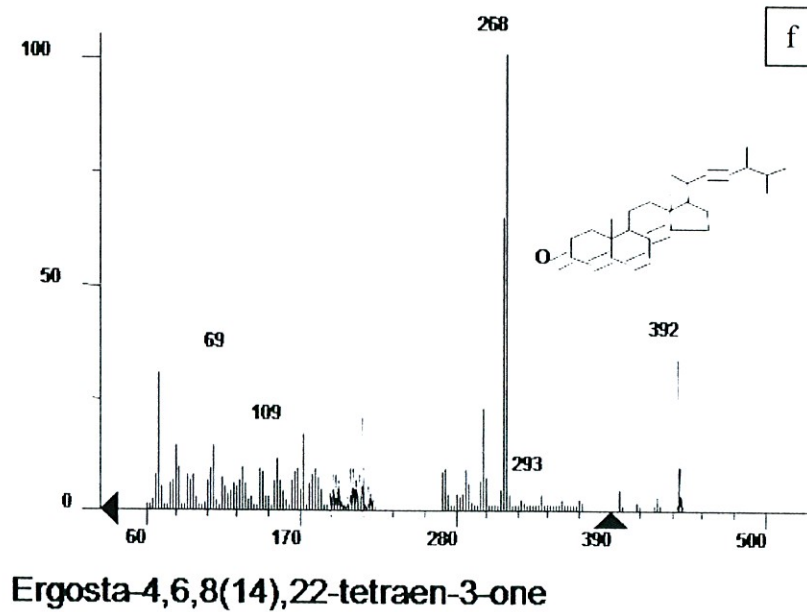


Figura 7 b,c,d. Estructura de algunos compuestos químicos de interés en *Ganoderma*.

e



f



g

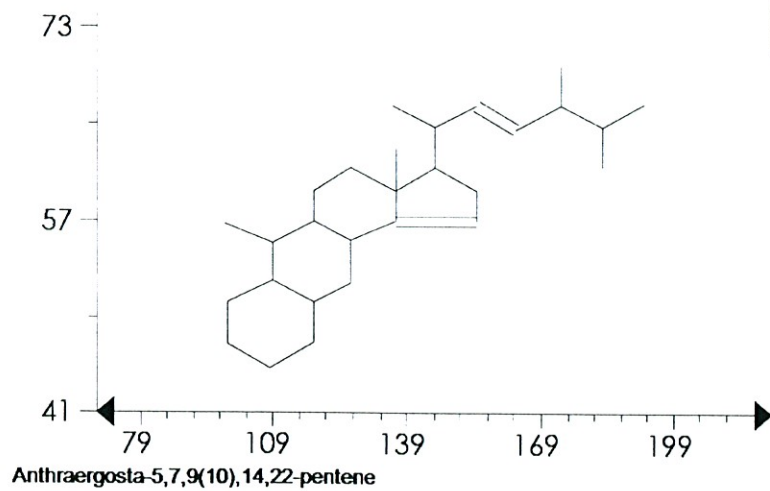
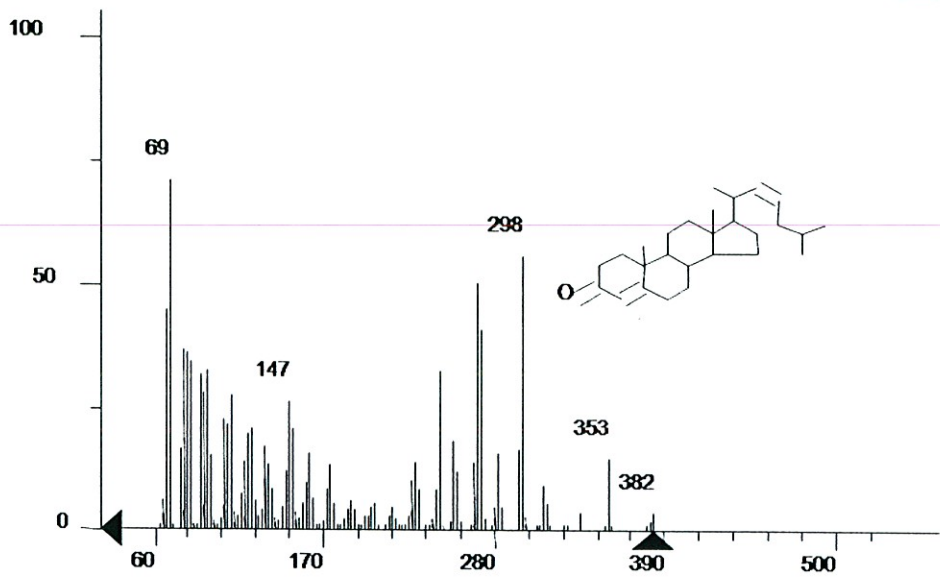


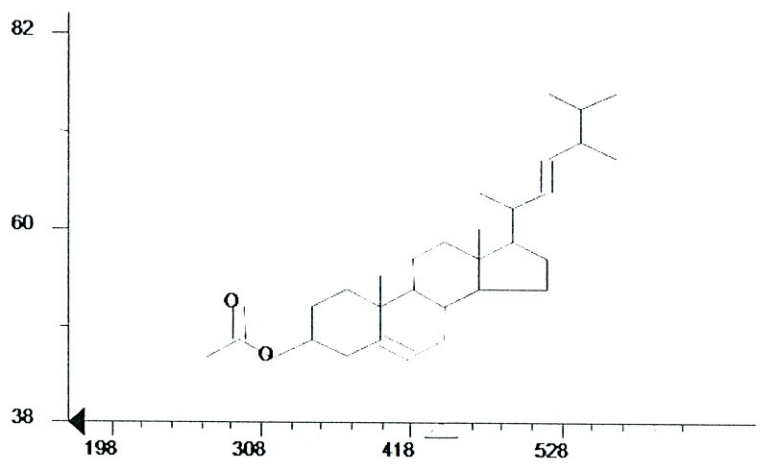
Figura 7 e, f, g. Estructura de algunos compuestos químicos de interés en *Ganoderma*.

h



4,22-Cholestadien-3-one

i



Ergosta-5,22-dien-3-ol, acetate, (3 $\alpha$ ,22E)

Figura 7 h, i. Estructura de algunos compuestos químicos de interés en *Ganoderma*.

De acuerdo con estos resultados, los materiales herborizados presentaron un menor número de compuestos cuando los materiales fueron procesados mecánicamente, ya que sólo se pudieron detectar al procesar la muestra con ultrasonido. Algunos compuestos únicamente están presentes en materiales recientes, pero existe un buen número de compuestos estables, que pueden ser potencialmente usados en taxonomía ya que permanecen en materiales almacenados. La detección de compuestos en el GC-MS es una opción en el análisis de muestras complejas. Se concluye que con la metodología usada se pueden obtener marcadores químicos específicos de especies de *Ganoderma*, que pueden ser usados como caracteres taxonómicos, una vez que la técnica haya sido estandarizada.

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## CAPÍTULO IV

### DISCUSIÓN Y CONCLUSIONES GENERALES

La sistemática a través de los tiempos ha permitido estudiar la taxonomía y las relaciones filogenéticas entre los diferentes grupos de organismos. Cada día son más las herramientas disponibles que permiten resolver preguntas de una manera más convincente y hacer hipótesis más robustas. La taxonomía tradicional ha usado a los caracteres morfológicos como principal fuente de análisis; sin embargo, en algunos casos éstos no son suficientes en la determinación de las especies y en el establecimiento de sus relaciones filogenéticas. Con el advenimiento de la biología molecular se ha logrado un avance importante en el esclarecimiento de relaciones de parentesco, lo cual en muchos casos ha podido ser interpretado a la luz de los datos morfológicos. Otras fuentes menos usadas son los caracteres químicos, los cuales son fuentes potenciales de información poco investigados.

En la época actual, algunos estudios relevantes en *Ganoderma* son los realizados por de Bazzalo & Wright (1982), Gottlieb & Wright (1999a, b), Ryvardeen (2000, 2004) y Decock & Herrera-Figueroa (2007). En éstos, se han precisado caracteres morfológicos importantes en la determinación de las especies, pero muchos otros continúan sin ser explorados a profundidad.

En *Ganodermataceae* Donk los límites taxonómicos no son aún muy claros, especialmente porque el uso inadecuado de los caracteres morfológicos ha originado confusiones, así muchas especies permanecen sin clasificar y otras han sido clasificadas erróneamente. Por ejemplo, *Amauroderma* Murrill y *Ganoderma* P. Karst., los dos grupos con mayor número de especies en *Ganodermataceae* están conformados por especies con características morfológicas muy diferentes, y no es claro si éstos corresponden a grupos naturales. Los análisis filogenéticos realizados hasta el momento por Moncalvo *et al.* (1995a, b), Moncalvo (2000), Smith & Sivasithamparam (2000a), Hong & Jung (2004) con datos moleculares, no son definitivos para contestar la pregunta de las relaciones filogenéticas en *Ganodermataceae*, porque no han incluido la suficiente representatividad para determinar la monofilia de los grupos. Los estudios realizados por Gottlieb *et al.* (1998) y Smith & Sivasithamparam (2000b) con isoenzimas mostraron que existen patrones que permiten establecer grupos naturales, pero sólo se discuten las relaciones fenéticas y se incluyeron pocos representantes.

En este trabajo, se definieron 45 caracteres morfológicos para 52 taxa de *Ganodermataceae* y grupo externo. En el análisis se incluyeron los ocho géneros propuestos en *Ganodermataceae*: *Amauroderma*, *Ganoderma*, *Elfvigia* P. Karst., *Haddowia* Steyaert, *Humphreya* Steyaert, *Magoderma* Steyaert, *Tomophagus* Murrill y *Trachyderma* (Imaz.) Imaz. Se encontró que a algunos caracteres, principalmente del contexto, basidiosporas y células del pileipellis, no se les ha dado el suficiente valor taxonómico. En este trabajo se propone a la consistencia del basidioma, estructura, color y presencia de sustancias resinosas en el contexto como caracteres importantes en la determinación de las especies. Las células del pileipellis presentan una gran número de caracteres potenciales, tal como fue propuesto por Gottlieb & Wright (1999a, b); sin embargo éstos no habían sido

bien definidos. La forma, el número de protuberancias, el grosor de la pared y las granulaciones son caracteres con valor taxonómico que se lograron definir en las células del pileipellis. En las basidiosporas encontramos la mayor fuente de información; proponemos la forma del ápice, disposición y grosor de los pilares como caracteres taxonómicos novedosos en el estudio de las relaciones filogenéticas.

Aunque el análisis con datos morfológicos falló en resolver las relaciones filogenéticas dentro de *Ganoderma*, las relaciones intergenéricas fueron casi resueltas con valores altos de bootstrap. En contraposición con algunos de los análisis realizados hasta ahora (Moncalvo *et al.* 1995b, Smith & Sivasithamparam 2000), en este trabajo *Ganoderma* resultó ser parafilético y los resultados apoyan la clasificación de los ocho géneros previamente propuestos, los cuales son principalmente definidos por el tipo de pileipellis y caracteres de las basidiosporas. Además, se apoya la monofilia de *Ganodermataceae*; así *Haddowiaceae* Jülich no es una familia independiente. En el análisis molecular realizado con secuencias del ITS del DNAr, se incluyeron seis de los géneros propuestos y especies de clasificación incierta. El árbol de consenso estricto obtenido no está completamente resuelto, pero se recuperaron seis clados, los cuales fueron altamente congruentes con los caracteres morfológicos y con los resultados del análisis con datos morfológicos, ya que en general se recuperaron los mismos clados.

Un gran aporte de este estudio fue el de definir caracteres morfológicos que resultaron ser útiles en la determinación de las especies y en establecer los límites entre los géneros. Esto nos permitió describir una nueva especie y aclarar la posición taxonómica de especies que fueron sinonimizadas bajo el mismo nombre. Por ejemplo para *Ganoderma resinaceum* Boud. se habían propuesto más de 12 sinónimos (Index Fungorum), que incluían especies con características morfológicas muy diferentes. Por otro lado, se presentan 10 segundos registros para el mundo y otros 30 para áreas geográficas específicas y se amplió la distribución geográfica de especies en donde sólo se conocía el tipo. *Ganoderma areolatum* Murrill fue sinonimizado con *Navisporus flococossus* (Bres.) Ryvar den, con base principalmente en las diferencias en las basidiosporas.

Se estableció que *Humphreya* [*H. coffeata* (Berk.) Steyaert], *Haddowia* [*H. neurospora* (J.S. Furtado) Teixeira], *Tomophagus* [*T. colossus* (Fr.) Murrill] y *Trachyderma* [*T. tsunodae* (Yasuda ex Lloyd) Imazeki] pueden ser considerados como géneros independientes de *Ganoderma*. Por otro lado, algunas especies con posición taxonómica confusa no fueron resueltas dentro del cladograma (por ejemplo *G. amazonense* Weir, *G. guianensis* Decock & Ryvar den, *G. lignosum* Pat.) y otras quedaron resueltas como clados separados de *Ganoderma*. Se necesita incluir un mayor número de representantes en los análisis para determinar su posición taxonómica. Además se requiere de más estudios morfológicos y análisis con otras regiones del DNA.

A través del análisis de triterpenos por espectrometría de gases acoplado a masas (GC-MS) se pudieron identificar más de 70 triterpenos, de ellos al menos 15 potencialmente útiles en la taxonomía de *Ganoderma*. Los triterpenos pudieron

ser detectados por GC-MS aún en materiales herborizados y especímenes con más de 30 años de haber sido recolectados, lo que permite su uso en taxonomía.

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