Complete Report

Project "Utilizing Thailand's Biodiversity: ascomycetes, taxonomy, phylogeny and screening for insecticides"

By Dr. Kevin D Hyde and Researchers

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Contact No.BRG5280002

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Project "Utilizing Thailand's Biodiversity: ascomycetes, taxonomy, phylogeny and screening for insecticides"

Researchers

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สนับสนุนโดยสำนักงานกองทุนสนับสนุนการวิจัย

(ความเห็นในรายงานนี้เป็นของผู้วิจัย สกว. ไม่จำเป็นต้องเห็นด้วยเสมอไป)

Contract No. BRG5280002
Project "Utilizing Thailand's Biodiversity: ascomycetes, taxonomy, phylogeny and screening for insecticides"
Fianl report.

Abstract

จากการศึกษาความหลากหลายทางชีวภาพของเชื้อรากลุ่มแอสโคมัยซีสในประเทศไทย ในระยะเวลาสามปีพบเชื้อราที่มีความแตกต่างกันมากกว่า 500 ชนิด และเชื้อราจำนวน 173 ชนิดได้ถูกตีพิมพ์ในวารสารวิชาการระดับนานาชาติ ในด้านที่เกี่ยวข้องกับการแยกเชื้อ การศึกษาลักษณะสัณฐานทางวิทยา และการจัดจำแนกโดยใช้เทคนิคทางอณูชีววิทยา ผลงานวิจัยที่ได้ตีพิมพ์ในวารสารวิชาการระดับนานาชาติมีทั้งหมด 13 ฉบับ โดยมุ่งเน้นการศึกษาเชื้อราในกลุ่มต่างๆ ได้แก่*Aquasubmersa, Astrosphaeriella, Bambusicola, Botryobambusa, Cophinforma, Curvularia, Deniquelata, Diaporthe, Pestalotiopsis, Botryosphaeriaceae, Capnodiaceae, Chaetothyriaceae, Trichomeriaceae, Tubeufiaceae, Kirschsteiniotheliaceae* และมีการรายงานเชื้อราชนิดใหม่ ประกอบด้วย 2 วงศ์, 9 สกุล และ 75 ชนิด นอกจากนี้ได้ศึกษาและวิเคราะห์เชื้อราที่มีบทบาทในการย่อยสลายใบไม้ร่วง สรุปคือ เชื้อราทั้งหมดมากกว่า 500 ชนิด ได้ถูกจัดเก็บเป็นตัวอย่างแห้งที่หน่วยเก็บรักษาเชื้อราแห้ง มหาวิทยาลัยแม่ฟ้าหลวง และเชื้อราจำนวน 173 ไอโซเลทได้ถูกเก็บรักษาไว้ที่หน่วยเก็บรักษาสายพันธุ์เชื้อรา มหาวิทยาลัยแม่ฟ้าหลวง เชื้อราจำนวน 10 ชนิดที่ได้จากการศึกษาความหลากหลายทางชีวภาพของเชื้อราข้างต้น นำมาศึกษาและตรวจสอบหาความสามารถในการผลิตสารออกฤทธิ์ทางชีวภาพ ในโครงการนี้มีนักศึกษาร่วมวิจัยจำนวน 4 คน เป็นนักศึกษาระดับปริญญาโท 1 คน และระดับปริญญาเอก 3 คน จนถึงปัจจุบันนี้ ผลงานที่ได้รับการตีพิมพ์หรืออยู่ในระหว่างการตีพิมพ์มีจำนวน 13 ฉบับ และอีกจำนวน 9 ฉบับอยู่ในขั้นตอนการเตรียมสำหรับตีพิมพ์ ซึ่งผลงานทั้งหมดได้กล่าวถึงการได้รับทุนวิจัยนี้ในกิตติกรรมประกาศ

Abstract

The biodiversity of Thailand ascomycetes has been studied over three years and in this time we collected more than 500 taxa and have published on more than 173 of these. This has involved isolation, and morphological and molecular characterization and we have published thirteen papers with major works on *Aquasubmersa Astrosphaeriella, Bambusicola, Botryobambusa, Cophinforma, Curvularia, Deniquelata, Diaporthe, Pestalotiopsis, Botryosphaeriaceae. Capnodiaceae, Chaetothyriaceae, Trichomeriaceae, Tubeufiaceae* and *Kirschsteiniotheliaceae*. This has resulted in the introduction of two new familes, nine genera and 75 new species. We have also analysed the fungi involved in the decay of leaves. We have deposited more than 500 herbarium specimens in MFLU and 173 cultures in MFLUCC. We have also studied the biochemistry of 10 selected species. Four students were trained, one at the MS and three at the PhD level. To date, thirteen papers are published or in press and a further night publications are in preparation and all acknowledge the grant.

1. Executive summary

1.1 Objective

1.1.1 To explore and identify the poorly studied ascomycetes in Thailand.

The ascomycetes on leaves and woody litter were explored and we have studied more than 500 taxa were identified and 173 isolates made.

1.1.2. To describe new or rare species found.

We have three novel familes, 9 new genera and 75 new species. New and interesting taxa were described and the publications of these fungi have been published or are in preparation (Please see appendix C).

1.1.3. To establish whether the fungal saprobes of leafy and woody litter are generalists, host-specific or host-recurrent.

The diversity of saprobic fungi on leaf litter from two hosts, *Magnolia liliifera* and *Cinnamomum iners*, in northern Thailand were studied. The results of these studies are being published and will discuss whether the fungal saprobes of leafy and woody litter are generalists, host specific or host recurrent (Please see appendix A).

1.1.4. To prepare herbarium specimens, isolate single spore cultures and deposit these in MFLU and BIOTEC.

All identified taxa (more than 500) were deposited as herbarium specimens in MFLU. All cultures 173 from single spore isolation were deposited in MFLU Culture Collection (MFLUCC).

1.1.5. To apply DNA sequencing strategies in the identification and phylogeny of novel species and other ill-defined or taxonomically confused taxa.

The new species and other ill-defined selected taxa were used for DNA extraction, PCR amplification and sequencing. Phylogenetic analyses of these fungi were carried out. Novel species and other ill-defined or taxonomically confused taxa were identified and combined with molecular phylogeny have been written up for publications (please see appendix C).

1.1.6 Assays against insects will be developed and used to establish potential natural metabolites that can be used in agriculture.

Ten cultures were extracted for fungal metabolites by solid phase extraction method. Crude extracts were tested for brine shrimp lethality bioassay to screen insecticidal activity. However, the bioassay was not successful. So, we change the plan to test the fungal metabolites for antimicrobial activity. This part was done in cooperation with Institute of chemical technology, Prague, Czech Republic. Fungal metabolites were extracted and isolated as crude extracts. All fractions of crude extracts were analyzed by FTIR. The antimicrobial activities of fungal metabolites were carried out by disc diffusion methods. Some extracts showed interesting compounds and were effective inhibitors against some pathogenic microorganisms (Please see appendix A).

1.2 Conclusion

1.2.1 Biodiversity of fungi on leaves and woody litter in northern Thailand

The ascomycetes on leaves and woody litter were explored and more than 500 taxa were identified and isolated. All identified taxa are deposited as herbarium specimens in MFLU. All cultures are also deposited in MFLU Culture Collection (MFLUCC). A paper entitled Diversity of fungi on leaves and woody litter has been written and some paper were published (please see appendix A-D)

1.2.2 Phylogenetic and molecular study

Molecular analysis of new species and other ill-defined taxa and groups of fungi have been carried out. In summary, two families, 8 genera and 65 species had been published or are in press. One genera and 10 species are submitted or are in preparation. New and interesting taxa were described with molecular data and phylogenies of other taxa have been refined and published. (please see appendix C)

1.2.3 Study with the type specimens

The families *Planistromellaceae, Patellariaceae Tubeufiaceae, Capnodiaceae* and *Botryosphaeriaceae* were selected for studying type specimens. By studying type specimens we could understand the family and collect species in Thailand. Type specimens were loaned from various herbaria. Samples were examined and made the descriptions. Publications on all families have been written.

1.2.4 Screening antimicrobial activities from fungi

Ten cultures were selected for screening insecticidal activity of fungal metabolites. Fungal metabolites were extracted by solid phase extraction method. Crude extracts were used for brine shrimp lethality bioassay to screen insecticidal activity. However, the bioassay was not successful. So, we change the plan to test the fungal metabolites for antimicrobial activity.

Ten isolates were selected for preliminary screening of antimicrobial activities. This part was done in cooperation with Institute of chemical technology, Prague, Czech Republic. Fungal metabolites were extracted and isolated into crude extracts. All fractions of crude extracts were analyzed by FTIR. The antimicrobial activities of fungal metabolites was carried out by disc diffusion methods. Some extracts showed interesting compounds and were effective inhibitors against some pathogenic microorganisms.

2. Results

Two main students were employed for this research project, but the project also contributed towards other students studies.

2.1 Ms. Jutamart Monkai, M.Sc. (Bioscience) School of science, Mae Fah Luang University. June 2009

2.2 Ms. Supalak Yacharone, Ph.D. (Bioscience) Mae Fah Luang University. October 2009

2.3 Ms. Putarak Chomnunti, Ph.D. (Bioscience) Mae Fah Luang University. October 2009

2.4 Ms.Saranyapat Boonmee, Ph.D. (Bioscience) Mae Fah Luang University. October 2009

3. Project output

Please see table of project output.

4. Any activities

4.1 Published in international journal (Please see appendix D)

We have 22 published / accepted/ in press and in prep papers as the result of this grant

4.2 Poster (Please see appendex E)

4.2.1 The poster was presented at the "10th Conference and meeting between new researcher and senior researcher" at Holiday Inn Resort Regent Beach Cha Am, Phetchabury Province. 14-16 October 2010. In title "Diversity of fungi on leaf and woody litter from some selected trees in northern Thailand and screening for insecticide production.

4.2.2 The posters was presented at the International Symposium on "Fungal Biodiversity and Resources" at Wangcam Hotel, Chiag Rai" 11-13 November 2010. Title "Biodiversity of saprobic fungi on woody litter of northern Thailand and screening for some insecticidal activity".

4.3 An oral presentation (Please see appendex E)

4.3.1 Prof. Dr. Kevin D Hyde gave a talk entitled "Exploration of the biodiversity of Asian microfungi and basidiomycetes" at international Symposium on "Explotation of fungi in the era of molecular phylogenetics and genomics, with special emphasis to their biodiversity, chemical ecology and secondary metabolites"at Faculty of Life Sciences Technical, University of Braunschweig, Germany" 6-11 April 2012

4.3.2 Prof. Dr. Kevin D Hyde was kenote speaker entitled "The value of epitypification" at international Mycological Ascociation on "One Fungus = Which Name?"at "Trippenhuis, Royal Netherland Academiy of Arts and Sciences (KNAW), Amsterdam, Netherlands" 12-14 Apirl 2012

4.3.3 Prof. Dr. Kevin D Hyde was given a talk entitled "Epitypication" at "Guizhuou Acadmy of Agricultural Sciences, China" 1 Apirl 2012

4.3.4 Jutamart Monkai was given an oral paper entitled "Diversity of fungi on leaf litter of *Magnolia lillifera* and *Cinnamomum iners* from Doi Suthep-Pui National Park, Thailand" was given by Mae Fah Luang University at international Symposium on "Fungal Biodiversity and Resources"at Wangcam Hotel, Chiag Rai Province 11-13 November 2010

5. Co-operated with other inter-institute and in Thailand

5.1 Rampai Kodsueb, lecturer from Faculty of Science and Technology, Pibulsongkram Rajabhat University, Phisanulok

6. Co-operated with other faculty in the same university

6.1 Itthayakorn Promputtha, lecturer from School of Cosmetic Science, Mae Fah Luang University, Muang, Chinag Rai

7. Award had been received (Please see appendex F)

7.1 For outstanding professional achievements and contributions to the Asian Mycological Committee by the Asian Mycology Committee Dr Hyde was awarded the Eminent Mycologist Award on August 7, 2011

Table of project output

Expected output	Output indicator	1st year		2nd year		3rd year		Comments
		1	2	1	2	1	2	
1. Discoveries	New fungal strains and species. New data on Thailand's fungal diversity	√	√	√	√	√	√	Yes – please see appendix C
2. Phylogenetics	Discover placement of species and new phylogenies		√	√	√	√	√	The papers published- see appendix C
2. Technology	Bioinsecticide assays		√	√	√	√	√	One student went to Czech to repeat this- please see appendix A
3. Knowledge	Publications		√	√	√	√	√	Twenty-two others publications –please see appendix D
4. Human resource development √ Ph.D.		√	√	√	√	√	√	Two students registered for higher degrees and partial funding of two other students.
5. Publication (s) √ Thai journal √ International journal	Several and both in local and mostly international					√	√	Twenty-two publications please see appendix D. Ten are published and accepted. Three are submitted and 9 other publications being written up.
6. Research presentation √ within Thailand √ Overseas	Several and both in and outside Thailand			√			√	Please see some abstract at appendix E

ลงนาม.....
(หัวหน้าโครงการวิจัยผู้รับทุน)

วันที่ 31 July 2012

BRG5280002

Appendix A

Final Report
Part 1

Diversity of saprobic fungi on dead leaves from *Magnolia liliifera* and *Cinnamomum iners* in northern Thailand and screening for bioactive compounds of some selected fungi.

Reported by

Miss Jutamart Monkai
Master of Science (Bioscience)

Advised by

Assoc. Prof. Dr. Kevin D Hyde
Assist. Prof. Dr. Ekachai Chukeatirote
Prof. Dr. Eric E.C. McKenzie
Dr. Rampai Kodsueb
Dr. Itthayakorn Promputtha
Dr. Sunita Chamyuang

School of science
Mae Fah Luang University

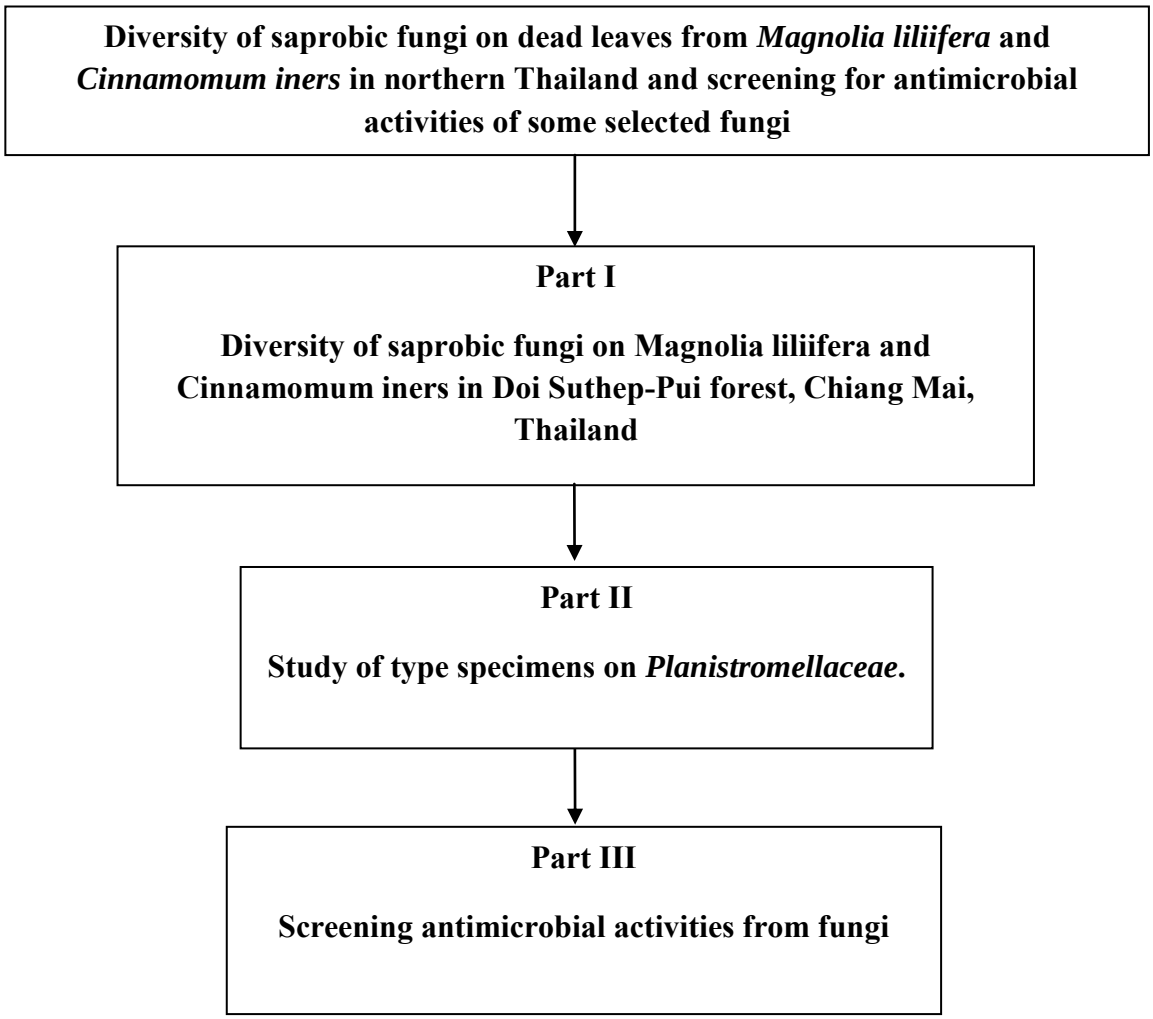
BRG5280002

Diversity of saprobic fungi on dead leaves from *Magnolia liliifera* and *Cinnamomum iners* in northern Thailand and screening for antimicrobial activities of some selected fungi.

Objectives

1. To study the diversity of saprobic fungi on leaves of selected tree species in the forests of northern Thailand.
2. To study type specimens in *Planistromellaceae*.
3. To screen antimicrobial activities of saprobic fungi.

Research contents



Part I

(Draft manuscript)
Part I (Draft manuscript)

Diversity of saprobic fungi on dead leaves from *Magnolia liliifera* and *Cinnamomum iners* in northern Thailand

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[5]*Landcare Research, Private Bag 92170, Auckland, New Zealand*
[6]*King Saud University, College of Science, Botany and Microbiology Department, P.O. Box 2455, Riyadh 1145, Saudi Arabia*

Abstract:

Keywords: fungal diversity, saprobic fungi, *Magnolia liliifera, Cinnamomum iners*.

Introduction

Saprobic fungi function as decomposers of organic material and recycle nutrients to other organisms (Cooke and Rayner, 1984). The diversity of saprobic fungi on various hosts have generally been poorly studied in both temperate and tropical regions (see Polishook *et al*., 1996; Hyde *et al*., 2001; Photita *et al*., 2001; Wong and Hyde, 2001; Parungao *et al*., 2002; Paulus *et al*., 2003; Lee *et al*., 2004; Santana *et al*., 2005; Paulus *et al.*, 2006; Kannangara *et al*., 2007; Przybył *et al.*, 2007). To estimate numbers of fungal species, a greater number studies on fungal diversity are needed, especially in unexplored habitats, hosts or poorly studied countries and especially in tropical regions (Hawksworth and Rossman, 1997; Hyde, 2001).

Forests of the northern parts of Thailand have great plant diversity (Gardner *et al.*, 2000) and several studies on their fungal diversity have been carried out in recent years. These include studies of fungi on monocotyledons (Photita *et al.*, 2003; Bussaban *et al*, 2004; Thongkantha *et al*., 2008; Bhilabutra *et al*, 2010), on *Shorea obtusa* (*Dipterocarpaceae*) (Osono *et al.*, 2009), on *Ficus* species (Wang *et al*., 2008), on leaf and woody litter of *Magnoliaceae* (Promputtha *et al.*, 2002, 2004; Kodsueb *et al.*, 2008), on *Castanopsis diversifolia* (Duong *et al.*, 2008)), on pods of *Delonix regia* (Somrithipol *et al*., 2002), on para rubber (*Hevea brasiliensis*) (Seephueak *et al*., 2010, 2011) and several studies on basidiomycetes (Sanmee, 2008, Sysouphanthong *et al*., 2010, Van de Putte *et al*., 2010; Zhao *et al*., 2010). This has resulted in several taxonomic advances (see Fournier *et al*., 2010, Boonmee *et al*., 2011a,b, Liu *et al*.,

2011). Studies of saprobic fungi have provided further biodiversity data and many new taxa have been also described (Duong *et al*., 2004). However, only few published studies have established the diversity of fungal communities and overlapping fungi on litter types in the tropics (Dulymamode *et al*., 2001; Yanna *et al*., 2001; Ananda and Sridhar, 2004; Paulus *et al*., 2006; Yule and Gomez., 2008). *Magnolia liliifera* (L.) Baill.**,** family *Magnoliaceae*, is an evergreen and in northern Thailand, is commonly located in Doi Suthep-Pui National Park (Gardner *et al*., 2000). *Magnolia liliifera* leaves are large and thick and this host is a good source for saprobic fungi (Promputtha *et al*, 2004). Promputtha *et al.* (2002, 2004) studied saprobic fungi on *Magnolia liliifera* leaves on the forest of Doi Suthep-Pui National Park during the in wet season of 2001. They found that many diverse fungal communities and undescribed species (e.g. *Anthostomella monthadoia*, *Dokmaia monthadangii*, *Hyponectria manglietiae*, *Pseudohalonectria suthepensis*) . Saprobic fungi on decaying woody litter of *Magnolia liliifera* on the forest of Doi Suthep-Pui National Park during the wet and dry seasons were reported by Kodsueb *et al*. (2008). The results show that samples collected in the dry season provided greater species richness than the samples collected in the wet season.

Cinnamomum iners Reinw. ex Blume (Lauraceae) has essential oils in the leaves which are used for flavoring sweets and confectionery (Jantan *et al*., 1995). It has been reported that leaf extracts can produced some biological and pharmaceutical activity (Jantan *et al*., 1992 and Zaridah *et al*., 2006). In Thailand, there have been some studies concerning endophytic fungi from *Cinnamomum* species (Worapong *et al*., 2001, 2003; Lumyong *et al*., 2002; Suwannarach *et al*., 2010). Worapong *et al*. (200-3) and Suwannarach *et al*. (2010) studied the endophytic fungi from *Cinnamomum* species and discovered new - species and new strains that can produce volatile compounds. However, there has been no study on the fungal diversity on decaying *Cinnamomum iners* leaves. It is therefore interesting to study these fungi to understand the relationship between fungi and host.

Previous studies on saprobic fungi on the above hosts were only based in the wet season when the number of fungi on the forest floor is abundant (Hyde *et al*., 2001; Parungao *et al*., 2002; Paulus et *al*., 2006;). In this study we focused on saprobic fungi on *Magnolia liliifera* and *Cinnamomum iners* in dry season at Doi Suthep-Pui, Chiang Mai, Thailand. The purposes of the present study were to examine fungal communities and compare fungal species between two hosts and to establish whether fungal saprobe are host-specific or host-recurrent. In addition, the saprobic fungal communities and patterns on each host were evaluated for the effect of different stages of decomposition, incubation periods or decaying times and parts of leaf (leaf lamina, midrib and petiole) or tissue specificity.

Materials and Methods

Study site and sample collection

The study site was located in an evergreen forest in Doi Suthep-Pui, Chiang Mai, northern Thailand (N 18 48’ 18.73”, E 98 54’ 47.28”, elev., 107 m) and samples were collected in the dry season between November and April ****, when there was low humidity; the forest floor was damp but not wet. Ten decaying leaves were randomly collected from each tree *Magnolia liliifera* and *Cinnamomum iners* in 100^2 meter plots and returned to the laboratory.

Experimental design and examination

Leaves were divided into two stages of decomposition, Stage I were recently green or yellow fallen leaves and stage II were mostly decaying brown leaves (Figure 1). Five leaves of each stage was observed. Samples were incubated in 15 mm diameter sterile Petri dishes with a tissue paper moistened by sterile distilled water at room temperature (Manoch, 2004). All fungi were examined after one week, three weeks and six weeks of incubation. During examination, fungi occurring on leaf lamina, midrib and petiole were recorded. Fungi were identified and described based on morphological characters using relevant references (e.g. Ellis, 1971; 1976; Carmichael *et al*., 1980; Sutton, 1980; Sivanesan, 1984; Hanlin, 1990; 1998a; 1998b; Nag Raj, 1993; Seifert *et al.*,2011). Single spore isolation method was used in fungal isolation (Choi *et al*., 1999; Chomnunti *et al*. 2011). All cultures were grown on potato dextrose agar (PDA) and malt extract agar (MEA) and deposited in MFLU Culture Collection (MFLUCC) and BIOTEC Culture Collection (BCC). Type specimens were also deposited in MFLU Herbarium, Mae Fah Luang University, Thailand.

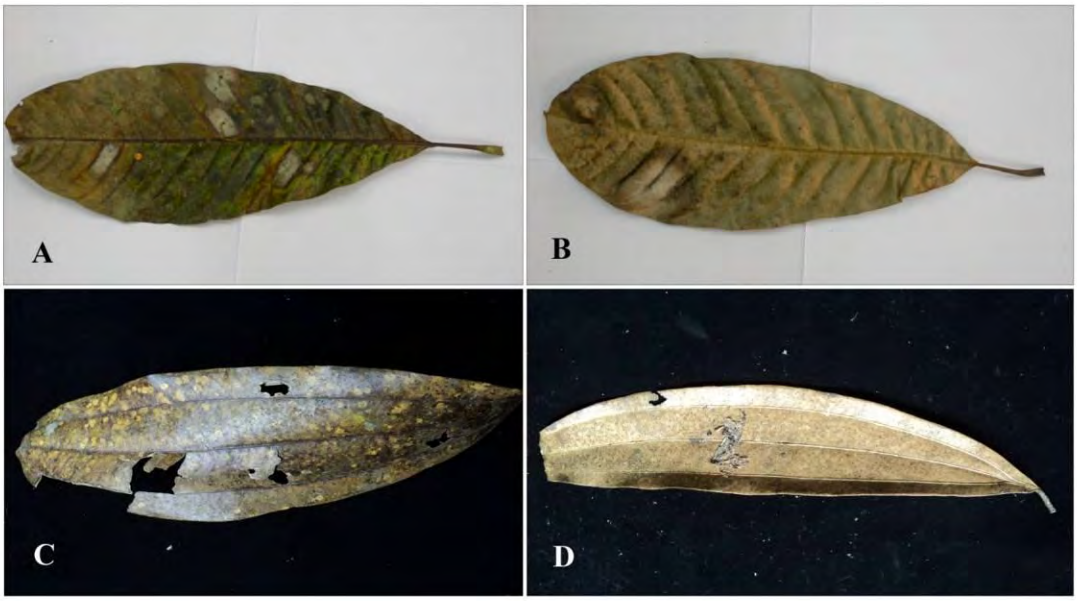

Figure 1 *Magnolia liliifera* and *Cinnamomum iners* leaves at each stage of decomposition. A. Stage I decaying leaf of *Magnolia liliifera*. B. Stage II decaying leaf of *Magnolia liliifera* . C. Stage I decaying leaf of *Cinnamomum iner*s. D. Stage II decaying leaf of *Cinnamomum iners*.

Statistical analysis

The presented occurrence of fungi were calculated and fungal taxa with a percentage occurrence higher than 10 are common species in this study. The formula of fungal percentage occurrence was measured by using the following formula.

$$\text{Percentage occurrence} = \frac{\text{Number of leaves which fungus was detected}}{\text{Total number of leaf samples examined}} \times 100$$

For the species diversity were compared in each stage of decomposition (stage I and II), 3 incubation periods (week I, II and III) and different part of leaves (leaf lamina, midrib and petiole) by using Shannon indices (H') to demonstrate the result of species diversity of a community (Shannon and Weaver, 1949).

$$H' = -\sum_{i=1}^{n} P_i \log_e P_i, \text{ and } P_i = \frac{N_i}{N}$$

N_i is individual number of *i* species
N is individual number of all species
P_i is the proportion of *i* species
n is the number of species.

The similarities of fungal assemblages from leaves at different weeks in both stages were identified by using cluster analysis PC-ORD version 2.12 (Hammer et al., 2001).

Results

1. *Magnolia liliifera*

1.1 Number and percentage of occurrence of fungal species

A total of 141 fungal collections were made and 36 taxa were identified from decaying leaves of *Magnolia liliifera*. This comprised nine ascomycetes (representing 25% of all taxa) and 27 anamorphic fungi (75%) including 11 coelomycetes (31%) and 16 hyphomycetes (44%). The number of fungal collections and their percentage occurrence are shown in Table 1. The most abundant species were *Ellisembia* sp. 1 (80%), *Stachybotrys* sp. 1 (70%), *Colletotrichum* sp. 1 (60%), *Dicyma* sp. 1 (60%), *Lasiosphaeria*-like sp. 1 (60%) and *Volutella* sp. 1 (60%).

1.2 Effect of stage of decomposition at different incubation periods on fungal communities

The percentage occurrence of fungi at each stage of decomposition is given in Table 1. Dominant species at Stage I of composition were *Dicyma* sp. 1 (100%), *Ellisembia* sp. 1 (100%) and *Volutella* sp. 1. (100%). The most abundant species at Stage II of composition were Anamorph of *Eutypa* sp. 1 (60%), *Colletotrichum* sp. 1 (60%), *Ellisembia* sp. 1 (60%), *Ophioceras* sp. 1 (60%) and *Stachybotrys* sp. 1 (60%). The lists of fungi present at each stage of decomposition and their overlap are listed in Table 2.

Diversity indices which in term of Shannon diversity index (H) and Simpson diversity index (D) are given in Table 3. These diversity indices were used to compare fungal communities at each stage of decomposition at different incubation periods. The diversity of fungal species was similar at both stages of decomposition. This is indicated by Shannon indices in Table 3. Whereas, the Shannon indices showed respectively highest to lowest diversity from three weeks, six weeks and one week of incubation. However, species richness at both stages of decomposition was distinctly different. The number of species at Stage I of decomposition was higher than that at Stage II of decomposition.

The dendogram of cluster analysis showed that the fungal communities were divided into three groups. The first group, fungal communities at Stage II, Week 3 and Stage II, Week 6 were more similar to each other than to those on other two groups. The second group, the fungal community at Stage I, Week 1 was close to Stage 2, Week1 and this group was also close to first group. The third group, the fungal community on Stage 1, Week 3 and Stage 1, Week 6 clustered together, and this group was distant from the other two groups (Fig. 2).

1.3 Effect of leaf tissue types on fungal communities

Percentage occurrence of fungi on different part of leaves are given in Table 1. The most abundant species in the leaf material were *Stachybotrys* sp. 1 (70%),

Dicyma sp. 1 (60%), *Lasiosphaeria-like* sp. 1 (60%), *Ellisembia* sp. 1 (40%) and *Phoma* sp. 1 (40%). The most abundant species in the midrib were *Ellisembia* sp. 1 (60%), *Volutella* sp. 1 (40%), anamorph of *Eutypa* sp. 1 (30%), *Colletotrichum* sp. 1 (30%) and *Stachybotrys* sp. 1 (30%). The most abundant species on the petioles were *Ellisembia* sp. 1 (40%), *Fusicoccum* sp. 1 (30%) and *Ophioceras* sp. 1 (30%) (Table 1).

Diversity indices are shown in Table 3. The highest number and diversity of fungi were found on midrib, while the second and third number and diversity were found on leaf lamina and petiole, respectively. But, on midrib and leaf lamina have similarly in number of fungi and more than on petiole.

2. *Cinnamomum iners*

2.1 Number and percentage of occurrence of fungal species

A total of 58 fungal collections and 18 taxa were identified from decaying leaves of *Cinamomum iners*. This comprised two ascomycetes (representing 11.1% of all taxa) and 16 anamorphic fungi (88.9%) including six coelomycetes (33.3%) and ten hyphomycetes (55.6%). The number of fungal collections and their percentage occurrence are shown in Table 4. The most abundant species were Anamorph of *Eutypa* sp. 1 (60%), *Pleurophragmium* sp. 1 (60%), *Acremonium*-like sp. 1 (40%), *Colletotrichum* sp. 1 (40%) and *Sporodochium* sp. 1 (40%).

2.2 Effect of stage of decomposition at different incubation periods on fungal communities

The percentage occurrence of fungi at each stage of decomposition are given in Table 4. The most abundant species at Stage I were *Acromonium*-like sp. *1* (60%), *Pleurophragmium* sp. 1 (60%), anamorph of *Eutypa* sp. 1 (40%), Coelomycetes sp. 3 (40%), *Colletotrichum* sp. 1 (40%), *Diplodia* sp. 1 (40%), Hyphomycetes sp. 1 (40%), *Pyricularia* sp. 1(40%) and Sporodochium sp. 1 (40%). The most abundant species in Stage II were anamorph of *Eutypa* sp. 1 (80%), *Pleurophragmium* sp. 1 (60%), *Colletotrichum* sp. 1 (40%), Hyphomycetes sp. 2 (40%) and Sporodochium sp. 1 (40%). The lists of fungi that were present at each and both stages of decomposition are shown in Table 5.

Diversity indices which in term of Shannon diversity index (H) and Simpson diversity index (D) are given in Table 6. Leaves in Stage I of decomposition supported a greater diversity of fungi than leaves in stage II of decomposition and this was also indicated by the greater Shannon diversity index (Table 6). The number of species in stage I of decomposition was higher than the number of species in stage II of decomposition. In comparison to fungal diversity from different weeks of incubation, Stage I of decomposition showed the highest diversity of fungi after 3 weeks of incubation, the second was 1 week of incubation and the third was 6 weeks of incubation. Stage II of composition showed respectively highest to lowest diversity from 1 week, 3 weeks and 6 weeks of incubation.

The one dendogram of cluster analysis showed that the fungal communities were divided into three groups. The first group, fungal communities on stage I, week 1 and Stage II, week 1 were more similar to each other than to those on other two groups. The second group, the fungal community on stage I, week 6 was close to stage II, week6 and was also close to fungal community on stage II, week 3. The third group was the fungal community on Stage 1, week 3 and this group was distant from the other two groups (Fig. 6).

2.3 Effect of leaf tissue types on fungal communities

Percentage occurrence of fungi on different part of leaves are given in Table 4.

The most abundant species in different parts of leaf are presented. Most abundant species in part of leaf lamina were *Pleurophragmium* sp. 1 (60%), Hyphomycetes sp. 1 (30%) and Sporodochium sp. 1(30%). The most abundant species in a part of midrib were Coelomycetes sp. 3 (20%). The most abundant species in petioles were anamorph of *Eutypa* sp. 1 (60%), *Acremonium*-like sp. 1 (30%)and *Colletotrichum* sp. 1 (30%).

Diversity indices on parts of leaf during observation are shown in Table 5. The highest number and diversity of fungi was found on leaf lamina, while the second and third highest diversity were found on midrib and petiole, respectively. In addition, midrib and petiole provided the same number of fungi and this was lower than the leaf lamina.

3. Similarity of fungi between *Magnolia liliifera* and *Cinnamomum iners*

Fungal communities of each hosts at different stage of decomposition and different incubation periods were analyzed by three-dimensional correspondence analysis (Fig.).

Overlapping genera on both hosts are *Ophioceras* sp., anamorph of *Eutypa* sp., *Colletotrichum* sp. 1, *Diplodia* sp. and *Acremonium-like* sp.

Table 1. Number of decaying leaves of *M. liliifera* and percentage occurrence of fungi occurring on two stages of decomposition during three incubation periods and different parts of leaf.

	Stage I					Stage II							Part of leaf					
Fungal species	Week 1	Week 3	Week 6	TL	PO	Week 1	Week 3	Week 6	TL	PO	Overall TL	Overall PO	L	PO-L	M	PO-M	P	PO-P
Acremonium-like sp.1							1	1	1	20	1	10			1	10	1	10
Anamorph of *Eutypa* sp.1						1	2		3	60	3	30	1	10	3	30		
Ascomycetes sp.1							1		1	20	1	10			1	10	1	10
Ascomycetes sp.2							1		1	20	1	10	1	10				
Beltrania sp.1		1	1	2	40						2	20			1	10	2	20
Botryosphearia sp.1		1		1	20		1		1	20	2	20			1	10	1	10
Canalisporium caribense							1		1	20	1	10	1	10				
Ceolomycetes sp.1		1		1	20						1	10	1	10	1	10	1	10
Ceolomycetes sp.2							1	1	1	20	1	10					1	10
Ceolomycetes sp.3			1	1	20						1	10	1	10				
Ceolomycetes sp.4								1	1	20	1	10			1	10		
Cladosporium sp.1	1			1	20						1	10	1	10				
Clanostachy sp.1		1		1	20						1	10	1	10	1	10	1	10
Colletotrichum sp.1	3	1		3	60	3		1	3	60	6	60	3	30	3	30	2	20
Dicyma sp.1	5	1		5	100	1			1	20	6	60	6	60				
Diplodia sp.1		1	2	3	60						3	30			2	20	1	10
Ellisembia sp.1		4	5	5	100	1	3	1	3	60	8	80	4	40	6	60	4	40
Fusicoccum sp.1		2	2	2	40		1	1	1	20	3	30			2	20	3	30
Glomerella sp.1	2			2	40						2	20			2	20		

Table 1 (continued). Number of decaying leaves of *M. liliifera* and percentage occurrence of fungi occurring on two stages of decomposition during three incubation periods and different parts of leaf.

	Stage I					Stage II							Part of leaf					
Fungal species	Week 1	Week 3	Week 6	TL	PO	Week 1	Week 3	Week 6	TL	PO	Overall TL	Overall PO	L	PO-L	M	PO-M	P	PO-P
Hyphomycetes sp.1							1		1	20	1	10					1	10
Hyphomycetes sp.2		1		1	20						1	10	1	10				
Hyphomycetes sp.3		1		1	20						1	10	1	10				
Hyphomycetes sp.5							1		1	20	1	10			1	10		
Lasiosphaeria-like sp.1	4	1		4	80	2			2	40	6	60	6	60				
Montagonla sp.1							1	1	2	40	2	20	2	20				
Ophioceras sp.1		1	1	1	20		2	2	3	60	4	40			1	10	3	30
Pestalotiopsis sp.1	1			1	20	1					1	10	1	10				
Phaeosphaeria sp.1	2	1		2	40						2	20	2	20				
Phoma sp.1	1	1	1	3	60	2			2	40	5	50	4	40	1	10	1	10
Phomatospora sp.1			1	1	20						1	10	1	10	1	10		
Phomopsis sp.1			1	1	20						1	10					1	10
Stachybotrys sp.1	2	2	3	4	80	2	2	1	3	60	7	70	7	70	3	30	2	20
Stachylidium sp.1							1		1	20	1	10					1	10
Verticillium sp.1		1		1	20						1	10	1	10	1	10	1	10
Volutella sp.1		3	3	5	100		1		1	20	6	60	1	10	4	40	2	20
Zygosporium sp.1						2			1	20	1	10	1	10	1	10		

L = Leaf lamina, M = Midrib, P = Petiole, TL = Total number of leaves, PO = Percentage occurrence

Table 2. Fungal species appearing at each stage of decomposition on *M.liliifera*.

Fungal species appearing in both stage of decomposition	Fungal species appearing in stage of decomposition I	Fungal species appearing in stage of decomposition II
Botryosphearia sp.1	*Beltrania* sp.1	*Acremonium*-like sp.
Colletotrichum sp.1	Ceolomycetes sp.1	Anamorph of *Eutypa* sp.1
Dicyma sp.1	Ceolomycetes sp.3	Ascomycetes sp.1
Ellisembia sp.1	*Cladosporium* sp.1	Ascomycetes sp.2
Fusicoccum sp.1	*Clanostachy* sp.1	*Canalisporium caribense*
Lasiosphaeria-like sp.1	*Diplodia* sp.1	Ceolomycetes sp.2
Ophioceras sp.1	*Glomerella* sp.1	Ceolomycetes sp.4
Phoma sp.1	Hyphomycetes sp.2	Hyphomycetes sp.1
Stachybotrys sp.1	Hyphomycetes sp.3	Hyphomycetes sp.5
Volutella sp.1	*Pestalotiopsis* sp.1	*Montagonla* sp.1
	Phaeosphaeria sp.1	*Stachylidium* sp.1
	Phomatospora sp.1	*Zygosporium* sp.1
	Phomopsis sp.1	
	Verticillium sp.1	
Number species = 10	**Number species = 14**	**Number species = 12**

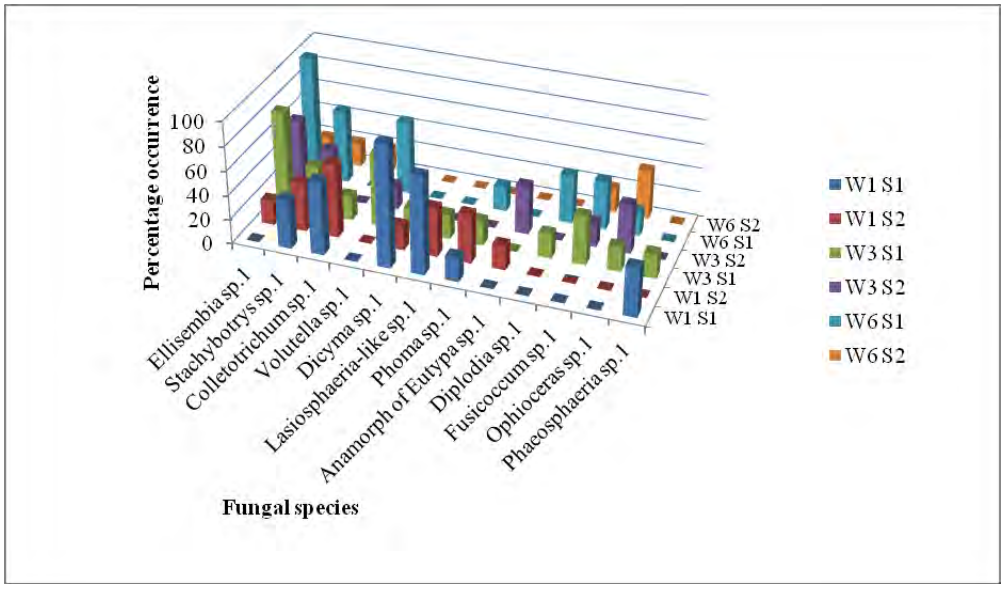

Fig. 2 Most abundant of fungal species occurred on *M. liliifera* distribution at different incubation periods and each stage of decomposition (S1 = stage I, S2 = stage II, W1 = week1, W3 = week 3, W6 = week 6). Species are ordered with most abundant on the left to the least abundant on the right.

Table 3. Diversity indices of fungi from *M. liliifera* on two stages of decomposition , three incubation periods and different parts of leaf.

	Stage I				Stage II				L	M	P
	Week 1	**Week 3**	**Week 6**	**Total**	**Week 1**	**Week 3**	**Week 6**	**Total**			
Species richness	9	18	12	**24**	8	14	9	**20**	22	23	16
Species evenness	0.85	0.77	0.73	**0.72**	0.95	0.87	0.94	**0.81**	0.64	0.73	0.80
Shannon indices	2.03	2.62	2.17	**2.84**	2.03	2.5	2.14	**2.79**	2.65	2.83	2.63
Simpson indices	0.85	0.90	0.85	**0.92**	0.86	0.9	0.88	**0.93**	0.9	0.92	0.90

L = Leaf lamina, M = Midrib, P = Petio

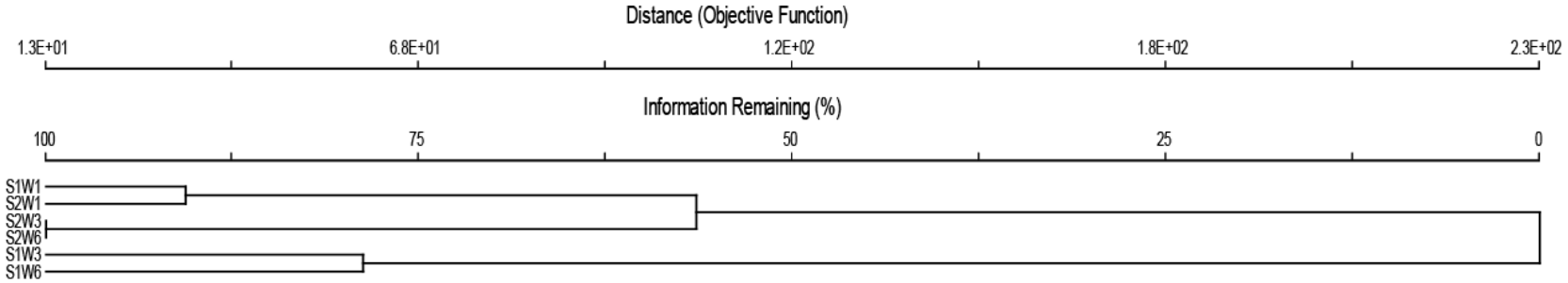

Fig. 3. Cluster analysis of saprobic fungi on *M. liliifera* based on Sørensen distance and the group average method (S1 = stage I, S2 = stage II, W1 = week1, W3 = week 3, W6 = week 6).

Table 4. Number of decaying leaves of *C. iners* and percentage occurrence of fungi occurring on two stages of decomposition during three incubation periods and different parts of leaf.

	Stage I					Stage II					Overall	Overall	Part of leaf					
Fungal species	**Week 1**	**Week 3**	**Week 6**	**TL**	**PO**	**Week 1**	**Week 3**	**Week 6**	**TL**	**PO**	**TL**	**PO**	**L**	**PO-L**	**M**	**PO-M**	**P**	**PO-P**
Acromonium-like sp.1		2	2	3	60			1	1	20	4	40	1	10			3	30
Anamorph *of Eutypa* sp.1	1	2		2	40	1	3	1	4	80	6	60	1	10			6	60
Chaetomium sp.1		1		1	20						1	10	1	10				
Coelomycete *sp.1*	1			1	20						1	10	1	10				
Coelomycetes sp.2		1	1	1	20						1	10	1	10	1	10		
Coelomycetes sp.3		2		2	40						2	20			2	20		
Colletotrichum sp.1	1	1	1	2	40		2		2	40	4	40			1	10	3	30
Crinula sp.1		1	1	1	20						1	10	1	10				
Diplodia sp.1	1	1	1	2	40	1			1	20	3	30	2	20			1	10
Ellislopsis sp.1	1			1	20						1	10			1	10		
Gliocladium-like sp.1		1		1	20						1	10			1	10		
Hyphomycetes sp.1		1	1	2	40		1	1	1	20	3	30	3	30			1	10
Hyphomycetes sp.2							1	2	2	40	2	20	2	20				
Ophioceras sp.1						1			1	20	1	10	1	10				
Pleurophragmium sp.1	3			3	60	3			3	60	6	60	6	60				
Pyricularia sp.1	2	1		2	40		1				2	20	1	10			1	10
Sporodochium sp.1	2			2	40	2			2	40	4	40	3	30	1	10		
Sporodochium sp.2						1	1		1	20	1	10	1	10				

W 1 = Week 1, W3 = Week 3, W6 = Week 6, L= Leaf lamina, M=Midrib, P= Petiole, TL= Total number of leaves

Table 5. Fungal species that appearing at each stage of decomposition on *C.iners*.

Fungal species appearing in both stage of decomposition	Fungal species appearing in stage of decomposition I	Fungal species appearing in stage of decomposition II
Acromonium-like sp.1	*Chaetomium* sp.1	Hyphomycetes sp.2
Anamorph *of Eutypa* sp.1	Coelomycete *sp.1*	*Ophioceras* sp.1
Colletotrichum sp.1	Coelomycetes sp.2	Sporodochium sp.2
Diplodia sp.1	Coelomycetes sp.3	
Hyphomycetes sp.1	*Crinula* sp.1	
Pleurophragmium sp.1	*Ellislopsis* sp.1	
Sporodochium sp.1	*Gliocladium-like* sp.1	
	Pyricularia sp.1	
Number species = 7	**Number species = 8**	**Number species = 3**

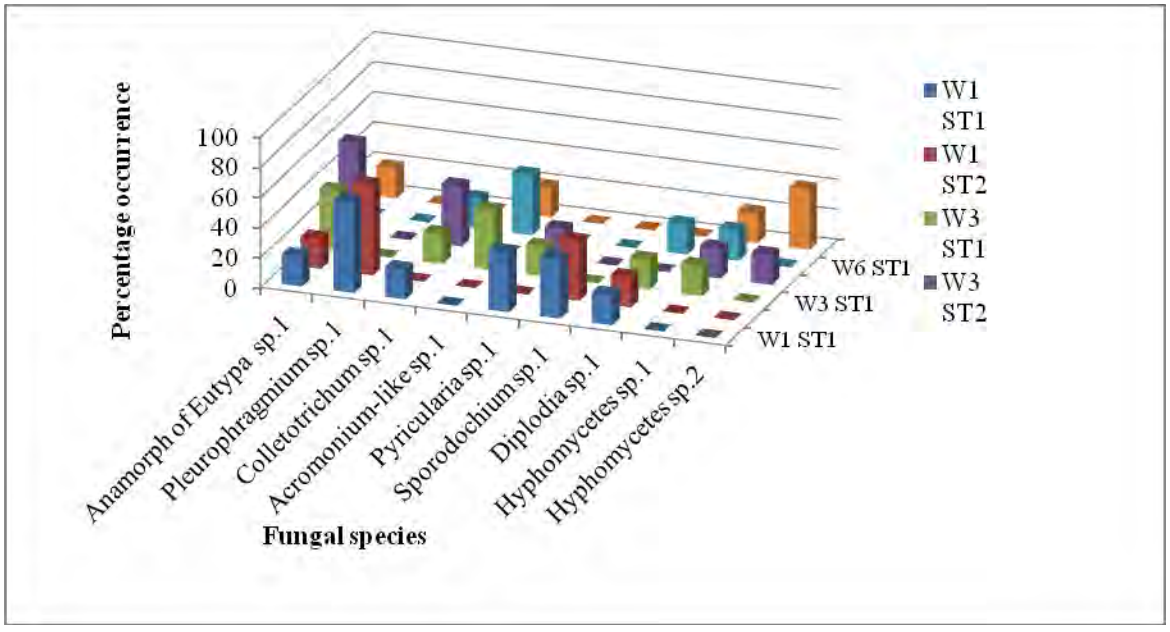

Fig. 4 Most abundant of fungal species occurred on *C. iners* distribution at different incubation periods and each stage of decomposition (S1 = stage I, S2 = stage II, W1 = week1, W3 = week 3, W6 = week 6). Species are ordered with most abundant on the left to the least abundant on the right.

Table 6. Diversity indices of fungi from *C. iners* on two stages of decomposition , three incubation periods and different parts of leaf.

	Stage I				Stage II				L	V	P
	Week 1	**Week 3**	**Week 6**	**Total**	**Week 1**	**Week 3**	**Week 6**	**Total**			
Species richness	8	12	6	**14**	6	5	4	**10**	16	6	6
Species evenness	0.92	0.85	0.96	**0.81**	0.90	0.92	0.95	**0.89**	074	0.96	0.82
Shannon indices	2.00	2.32	1.75	**2.43**	1.67	1.52	1.33	**2.18**	2.50	1.75	1.60
Simpson indices	0.85	0.89	0.82	**0.91**	0.80	0.77	0.72	**0.88**	0.90	0.982	0.80

L = Leaf lamina, M = Midrib, P = Petiole

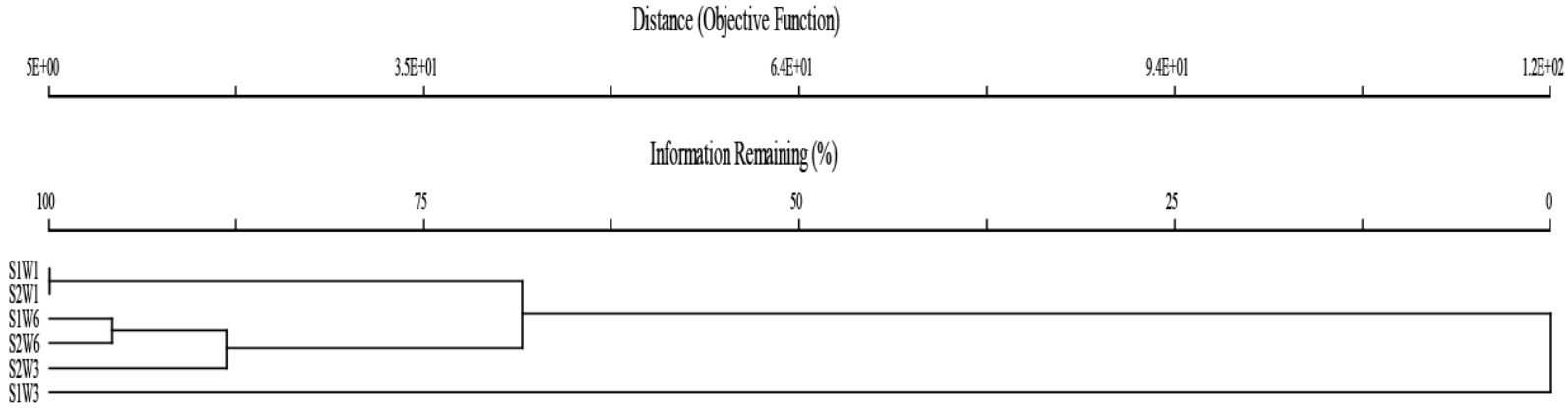

Fig. 5 Cluster analysis of saprobic fungi on *C. iners* based on Sørensen distance and the group average method (S1W1= stage I, week1, S1W3= Stage I, week 3, S1W6= stage I, week6, S2W1= stage II, week1, S2W3= Stage II, week 3, S2W6= stage II, week6).

Discussion

1. Diversity of fungi on decaying leaves in the dry season

In this study, we investigated fungal diversity on decaying leaves of *Magnolia liliifera* and *Cinamonium iners* in the dry season in northern Thailand. Fungal diversity of *M. liliifera* in the dry season can be compared with the previous study in the wet season (Promputtha *et al*., 2002, 2004). Number of fungi recorded on succession study were 22 taxa from 110 leaf samples (Promputtha *et al*., 2002) and recorded on naturally occurring decaying leaves were 37 taxa from 90 leaf samples (Promputtha *et al*., 2004). For this study, 36 fungal taxa were identified from 10 leaf samples. It seems that fungal communities of *M. liliifera* in the dry season is supported greater fungal taxa than in the wet season. The similar results were done by Kodsueb *et al*., 2008; Seephueak *et al*., 2010 and Seephueak *et al*., 2011. These studies reported that samples collected in the dry season had greater species richness and higher Shannon diversity index than samples collected in the rainy season. There are no seasonal effects of fungal communities on palms in Hong Kong (Yanna *et al*., 2001) and on *Pandanus penetrans* in Thailand (Thongkantha *et al*., 2008). The reason is still unclear. Pinnoi *et al.*, 2006 showed that the spore germination and reproduction of fungi were required quite high humidity. Otherwise, the wettest period had an unsuitable ratio between moisture content and aeration of wood with relative high moisture and low aeration (Rayner and Todd, 1979). However, it's difficult to compare this study with the previous study (promputtha *et al*., 2002, 2004). This may be because of the differences in the number of samples and the collecting years, which varied in temperature, humidity and rainfall. In addition, fungal species and pattern were different. The most of fungi in this study is anamorphic fungi, but in previous study is ascomycetes.

In the other hands, only 18 fungal taxa were identified from *C.iners* leaves and most of fungi are anamorphic fungi. Lumyong *et al*. (2002) isolated endophytic fungi of *C.iners* collected from Doi Suthep-Pui National Park, Thailand. The genera of fungi from endophyte (Lumyong *et al*., 2002) was not same as our study excepting only one fungi that is *Colletotrichum* sp. It showed that fungal diversity of *M.liliifera* provides greater species richness and Shannon diversity indices than fungi on *C.iners* leaves (Table 3 and 6). A reason for this may be caused by leaf area differing on each host as leaves of *M.liliifera* are larger than leaves of *C.iners*. The larger leaves are likely to provide higher species diversity than the smaller leaves (Wong and Hyde, 2001) This is consistent with previous studies on hosts such as banana, palm and *Magnolia liliifera* which supported higher diversity and more diverse taxa (Photita *et al*., 2001, 2003; Yanna *et al*., 2001; Promputtha *et al*., 2002, 2004). Other reason that effect to lower diversity and number of fungi on *C.iners*. may be because of the chemical composition of leaves. Leaves of *C.iners* are composed of essential oil such as eugenol and cinnamaldehyde as major bioactive compounds (Jantan *et al*., 1992). There have been many reports showing antimicrobial activities of essential oil on *Cinnamomum* species (Jantan *et al*., 1992; Ranasinghe *et al*., 2002; Mustaffa *et a*l., 2011). Antifungal tests demonstrated that the leaf essential oils of cinnamaldehyde and eugenol had strong inhibitory effect against wood decay fungi (Wang *et al*., 2005; Cheng *et al*., 2006). Yen and Chang (2008) concluded that the synergy of cinnamaldehyde with eugenol could be effected to fungi on the alteration of cell wall structure, reducing of cell wall synthesis, and the addition of radical scavenging.

2.Effect of decomposing stages and incubation periods

The decomposition process of saprobic fungi proceeded following three succession stages: the pioneer (early) stage, mature (middle) stage and the impoverished (later) stage (Dix and Webster, 1985; Yanna and Hyde, 2002). Pioneer communities are generally low fungal diversity and have few species occurring at high percentage occurrence (Dix and Webster, 1985). Mature communities are high fungal diversity and have many species

occurring at high percentage occurrence which some species become obviously dominant species (Dix and Webster, 1985; Promputtha *et al*, 2002). Eventually, the species diversity and the number of species are declined in impoverished communities (Dix and Webster, 1985). In early stage, decomposer fungi might switch their roles from endophytic and pathogen (Lodge and Cantrell 1995; Duong *et al*, 2008). The evidences of endophytic fungi change to be saprobes have been reported in previous studies (Osono *et al.*, 2004; 2009; Tang *et al*., 2005; Koide *et al.*, 2005; Promputtha *et al*., 2010; Purahong and Hyde, 2011). Promputtha *et al*., 2010 also provided that endophytes can produce various degrading enzymes in succession process which is an important activity for their adaptation to saprobic lifestyle. The primary enzymes apply for degrading the small soluble carbon-based molecules, such as hemicelluloses and the most complex; cellulose and lignin are then degraded at the late stage of decomposition (McClaugherty and Berg, 1987; Promputtha *et al*., 2010). In addition to soilborne and airborne fungi can colonize into fallen leaves as well (Duong *et al*, 2008). So, leaves which had touched on the forest floor for a long time might have higher fungal diversity than leaves which had recently fallen from the tree.

In this study, leaves from these two stages were observed. Decaying leaves of *M.liliifera* and *C.iners* in stage of decomposition I supported greater fungal diversity than in stage of decomposition II. Fungal communities on each stage of decomposition were distinct with low percent of overlap. *M.liliifera* and *C.iners* have overlap percentage of fungal species 27.7%, 38.8%, repetitively. In *M.liliifera*, among 24 species occurring on stage of decomposition I, 10 species continued to grow on stage of decomposition II along with additional 12 species. In C.iners, among 15 species occurring on stage of decomposition I, 7 species continued to grow on stage of decomposition II along with additional 3 species. Shanthi and Vitthal., 2010 studied leaf litter fungi of *Pavetta indica* on freshly fallen senescent leaves (grade 1) and on leaves already undergoing active decomposition (grade 2) and found that more taxa were found on grade 1 than on grade 2 litter. According to Seephueak *et al*., 2010 and Seephueak *et al*., 2011, fungi on different stages of decaying leaf and branch litter of the rubber tree were studied. The results showed that the number of taxa on middle stage decaying branches was higher than new and old decaying fallen branches.
The replacement of fungal species composition sequentially throughout decomposing process relies on the capability of decomposers to utilize organic matter and nutrients which are particular on each substrate or host (Frankland, 1992. Tang *et al*., 2005). This hypothesis is supported by our results showing different fungal communities between two hosts on each stage of decomposition and incubation time (fig 2 and 4). On *M.liliifera* in both stage of decomposition, the highest number and diversity of fungi were found in 3 weeks after incubated. This result is similar to previous succession studies that are provided above. On the other hand, on *C.iners*, number and diversity of fungi were distinct on each stage of decomposition. Stage of decomposition I showed the trend of abundant fungi on different incubation periods same as in *M.liliifera.* However, on stage of decomposition II, the highest number and diversity of fungi were found in 1 week after incubated and decline after 3 and 6 weeks of incubation respectively. Furthermore, it can assume that decaying process of *C.iners* leaves might slower than *M.liliifera* leaves. However, this study uses moist chamber method for examination of fungal diversity which does not come from natural habitat like in succession studies. So, in forest ecosystem, saprobic fungi also need environmental factors, such as pH, temperature and moisture for growing.(Osono *et a*l., 2003)

3. Effect of tissue types (Part of leaf)

Abundant of fungi on different part of leaf were distinct on each host. Leaf lamina is the largest surface area may support the greater number of fungal species than other parts of leaf (Promputtha *et al*., 2002). Surprisingly, on *M.liliifera*, midrib provided the highest number and diversity of fungi. In addition, percentage of fungi occurring on each incubation

periods were different among part of leaf (fig. 6). On 1 week of incubation, leaves lamina provided the highest number of fungi, but midribs and petioles still had low number of fungi. On the controversy, number of fungi on leaf lamina were decreased, but number of fungi on midrib and petioles were increased during 3 and 6 weeks of incubation. Especially, petioles provided the highest number of fungi after 6 weeks of incubation (fig. 6).

On the other hand, the highest number and diversity of fungi was found on leaves lamina of *C.iners* (fig. 8). On 1 week of incubation, leaves lamina provided the highest number of fungi, but midribs and petioles still had low number of fungi. Petioles had the highest number of fungi after 3 weeks of incubation. Interestingly, number of fungi on leaves lamina were decreased after 3 weeks of incubation, but were increased after 6 weeks of incubation. Fungi on the part of midribs were not occurred after 6 weeks of incubation (fig. 8) This may assume that primary fungal saprobes prefer to colonize in leaf lamina. Then, fungi are able to encounter on part of midrib and petiole. Structure and moisture content of tissue type may influence the presence of saprobic fungi. Leaves are thin-walled composing most of parenchymatous cells and less moisture. Midrib and petiole have more sclerenchyma cells that are thick-walled supporting more nutrients for fungal growth (Pinnoi *et al*., 2006) Beside, petioles contain vascular bundles which may keep moisture for a longer time (Fisher *et al.*, 2002). This may cause fungi on midrib and petiole are greater number of fungi in the later period. Some fungi can grow in every part of leaf. Some fungi grow in leaf lamina and/or vein and/or petioles. Some fungi strictly grow on one tissue type. For example on *M.liliifera*, Ceolomycetes sp.4, *Glomerella* sp.1 and Hyphomycetes sp.5 only were found on midrib. Hyphomycetes sp.1, *Phomopsis* sp.1 and *Stachylidium* sp.1 only were found on petioles. On *C.iners*, Coelomycetes sp.3, *Ellislopsis* sp.1 and *Gliocladium*-like sp.1 only were found on midrib. This result indicates that saprobic fungi are specific to tissue type. The recurrence of saprobic fungi on certain tissues has been also observed with other hosts (Photita *et al*., 2001, 2003; Yanna and Hyde, 2001; Promputtha *et al*., 2002,2004; Pinnoi *et al*., 2006; Duong *et al*., 2008). Yanna and Hyde, (2001) found that different frond parts of palms supported distinct fungal communities on most samples. The tissue specificity of saprobic fungi may be due to differing nutritional requirements and enzymatic capabilities to utilize different substrate (Adaskaveg *et al,* 1991; Yanna *et al*., 2001; Photita *et al*., 2003)

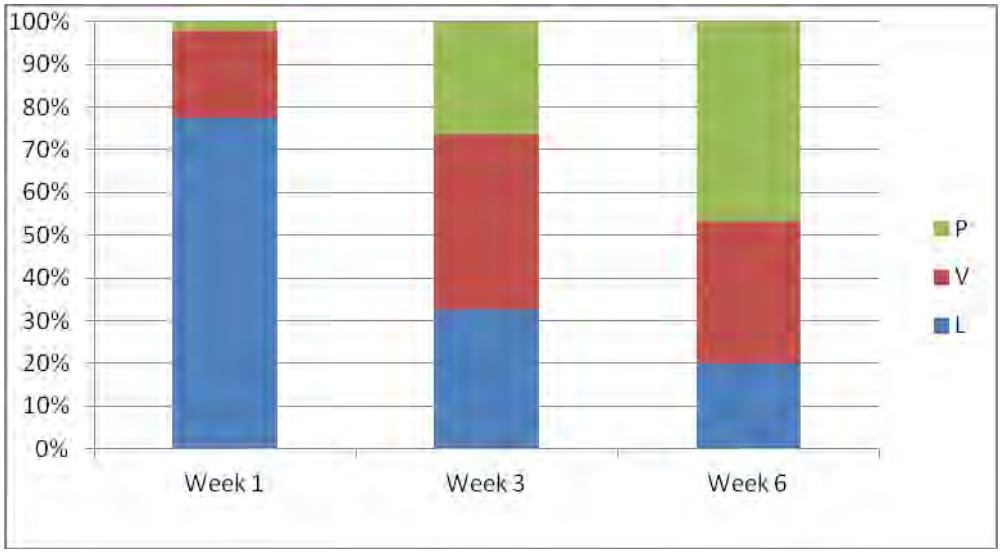

Fig 6. Percentage of fungi occurring on *M.liliifera* on different parts of leaf compare between each incubation periods (P=Petioles, V=Vein, L=Leaf lamina).

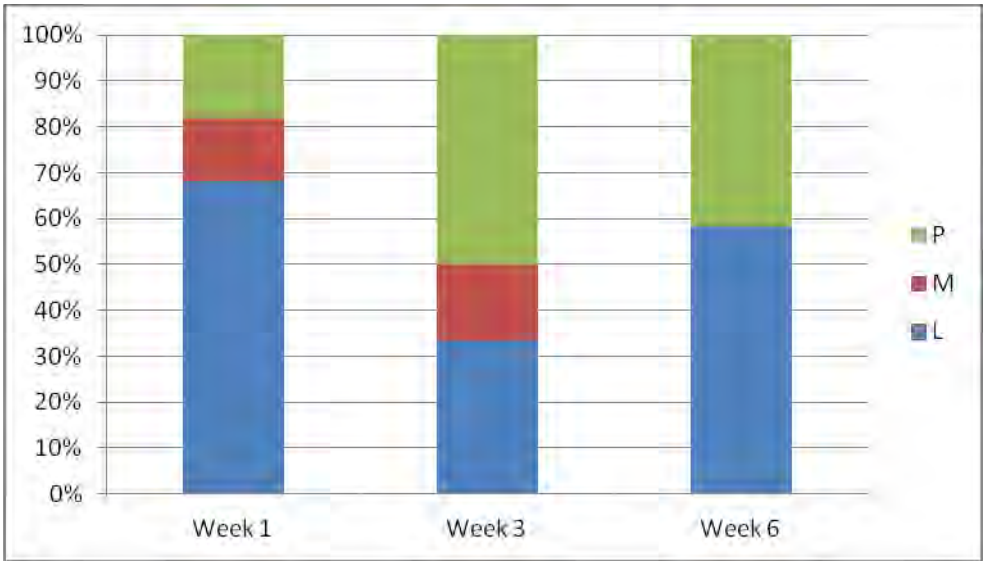

Fig 7. Percentage of fungi occurring on *C.iners* on different parts of leaf compare between each incubation periods (P=Petioles, V=Vein, L=Leaf lamina).

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Part II
Herbarium study on type species in *Planistromellaceae*

Planistromellaceae

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Abstract
The family *Planistromellaceae sensu* Lumbsch and Huhndorf 2010 often are found as saprobes and pathogens on various plants with characterized by ascostromata, interthecial tissues and schizogenously formed and periphysate ostioles. In this paper, we re-examined and described all generic type specimens including *Comminutispora, Eruptio*, *Loratospora*, *Microcyclus*, *Mycosphaerellopsis, Planistroma* and *Planistromella.* Molecular phylogenetic studies of these fungi were concluded and discussed with morphological character from re-examination. Some genera are transferred from *Planistromellaceae* to other family. The generic concept of *Planistromellaceae* is elucidated.

Key words
Planistromellaceae, type specimens, taxonomy

Introduction

The class *Dothideomycetes* comprises the largest fungal species and most phylogenetically diverse group in phylum *Ascomycota* with ascolocular development and bitunicate asci (Kirk *et al.* 2008). Previously, the classification of these fungi determined using morphological character including ascomal character, the presence of pseudoparaphyses and anamorphic states (Luttrell, 1955; von Arx and Müller, 1975; Eriksson, 1981; Barr, 1987). Several studies have been focused on molecular phylogenies of *Dothideomycetes* to elucidate the confusing classification combined with morphological character (Berbee, 1996; Silva-Hanlin and Hanlin, 1999; Lindemuth *et al*., 2001; Lumbsch and Lindemuth, 2001). These early studies supported that *Dothideomycetes* is not monophyletic. The recent phylogenetic analyses using multigenes data have been provided several lineages among a class wide context (Schoch *et al.*, 2006, 2009).

The *Planistromellaceae* was introduced by Barr (1996) with the generic type *Planistromella*. Presently, members of this family are 7 genera including *Comminutispora, Eruptio*, *Loratospora*, *Microcyclus*, *Mycosphaerellopsis, Planistroma* and *Planistromella* (Lumbsch and Huhndorf, 2010). They usually grow in living or dead leaves or stem of various plants, mostly are saprobes but only some species are pathogens (Evan, 1984; Ramaley, 1991, 1992, 1993, 1995, 1996, 1998; Kohlm. and Volkm.-Kohlm, 1993; Barr, 1996; Sivanesan and Shivas, 2002; Lieberei, 2007). The important morphological characters of *Planistromellaceae* are ascostromata, interthecial tissues and schizogenously formed and periphysate ostioles. These fungi are similar to members of the *Pseudosphaeriaceae* in having multiloculate or uniloculate ascostromata and in lacking any true peridial structure. But in those taxa, the locules open by a simple, lysigenous pore or by dehiscence of a caplike

structure. The *Mycosphaerellaceae* also resembles with these species in the characteristic of asci, ascospore and anamorphs excepting that in those taxa, the interthecial tissues are less extensive and the apical pore is formed lysigenously (Barr, 1996). The classification of some genera (*Planistroma*, *Loratospora*, *Eruptio* and *Microcyclus*) are confused by morphological characters which they are quite similar to other related genus. Recently, molecular phylogeny has been done to prove the validation of the members of the family. However, only few genera have been shown the results of molecular data that are included *Loratospora* (Suetrong *et al*., 2009), *Eruptio* (Crous *et al*., 2001, 2009a,b; Verkley *et al*., 2004), *Microcyclus* (Chee and Holiday ,1986; Le Guen, 2004). In the present work, we have been reexamined the type specimens for fully description on the combination with phylogenetic molecular to clarify the generic concept of the *Planistromellaceae* family.

Materials and methods

Type specimens of genera were obtain from Herbarium BPI, DAOM, IMI, K, S and UC. The herbarium specimens were rehydrated in 5% KOH prior to examination. Ascomata were sectioned by free-hand under a Motic SMZ 168 Series microscope. Morphological characters were studied using a Nikon ECLIPSE 80i microscope with a Canon 450D digital camera. The measurements were made using Tarosoft (R) Image Frame Work program.

Results

Planistromellaceae M.E. Barr, *Mycotaxon* 60: 437 (1996)
MycoBank: MB81919

Biotrophic, hemibiotrophic or *saprobic* in leaves and stem of various plants. Ascostromata multiloculate or uniloculate, immersed to erumpent, compose of brown, pseudoparenchymatous cells; locules small, opening by schizogenously formed, periphysate ostioles. Asci bitunicate, fissitunicate, oblong or saccate, in basal layer, often interspersed with and overtopped by cellular remnants of interthecial tissues. Ascospores hyaline or lightly pigmented, yellowish to brownish, aseptate or one to several transversely septate; wall thin, smooth, occasionally surrounded by gel coating; contents guttulate. Anamorphs where known forming pycnidia, locules or acervuli in stroma or bearing conidia over stroma surface prior to locule formation; conidiogenous cells short, conidiogenesis holoblastic; conidia hyaline to brown, aseptate or one to several transversely septate; wall smooth or verruculose, bearing one or more apical appendages at times. Spermatial state developing in the same or separate locules; conidiogenesis phialidic; spermatia minute, hyaline, aseptate.

Notes: The *Planistromellaceae* was introduced by Barr (1996) with characters of ascostromata, interthecial tissues and schizogenously formed, periphysate ostioles. These fungi are similar to members of the *Pseudosphaeriaceae* in having multiloculate or uniloculate ascostromata and in lacking any true peridial structure. But in those taxa, the locules open by a simple, lysigenous pore or by dehiscence of a caplike structure. The *Mycosphaerellaceae* also resembles with these species in the characteristic of asci, ascospore and anamorphs excepting that in those taxa, the interthecial tissues are less extensive and the apical pore is formed lysigenously (Barr, 1996). Members of the family are species of *Planistromella*, *Planistroma*, *Loratospora*, *Eruptio*, *Microcyclus* and *Mycosphaerellopsis*. *Planistromella* is the generic type of *Planistromellaceae*. The type species, *Planistromella yuccifoliorum* is characterized by subepidermal immersed, multilocular ascostromata, periphysate ostiole, bitunicate, slightly clavate or nearly cylindric asci, smooth, hyaline, septate ascospores (Ramaley, 1993).

Family type:

Botryosphaeriales

Planistromella A.W. Ramaley, *Mycotaxon* 47: 260 (1993).
MycoBank: MB22437 Fig. 1

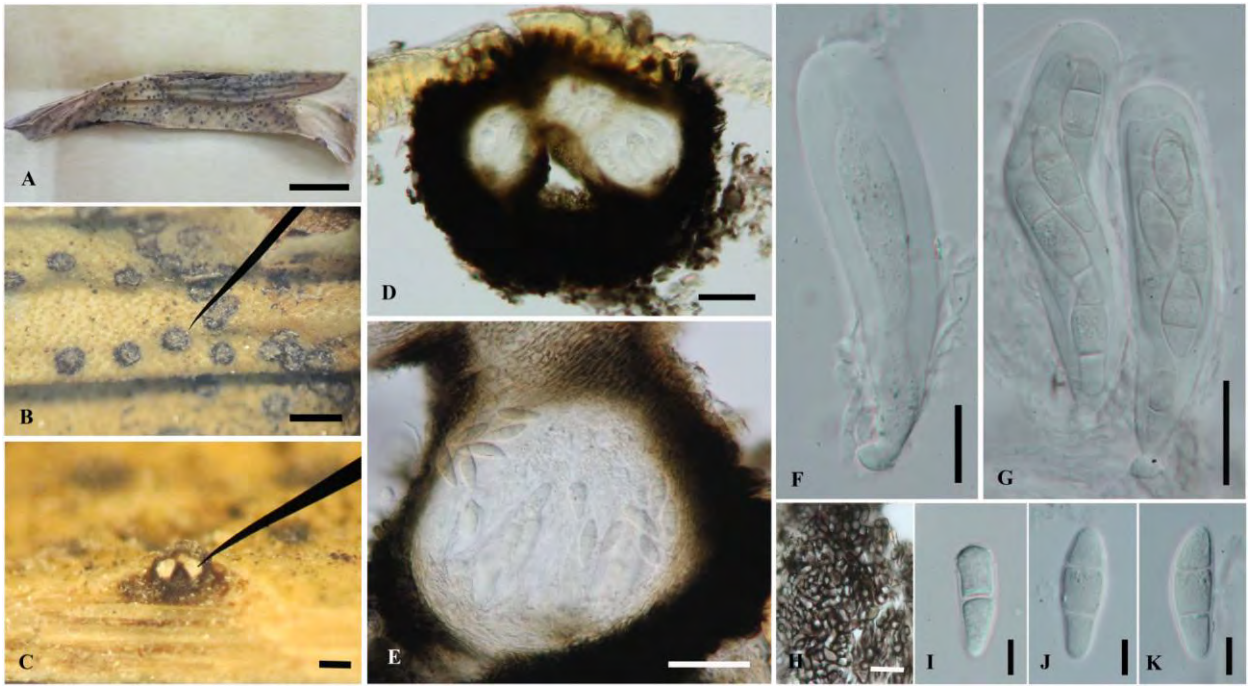


Fig. 1 ***Planistromella yuccifoliorum*** (holotype) on leaves of *Yucca brevifolia.* **A-B.** Ascostromata on the host surface. **C-D.** Section of ascostroma. **E.** Ascoma. **F.** Immature ascus. **G.** Mature asci. **H.** Peridium. **I.** Immature ascospore. **J-K.** Mature ascospores. **Scale bars:** A = 1 cm, **B** = 1000 µm, **C**= 200 µm**, D** = 100 µm**, E-G**= 50 µm**, H =** 20 µm, **I-K** = 10 µm

Ascostromata subepidermal, immersed, becoming erumpent, solitary to gregarious, multilocular, cup-shaped, dark, thick-walled, the upper part attached with the host epidermis. *Locules* ovoid to globose, in a single layer, filled with abundant interthecial tissue, each with a periphysate ostiole, separated by columns of elongated cells. Ascogenous locules developing in the same stroma as the conidial or spermatial locules or both. *Peridium* composed of several layers of dark-walled cells, on the top of a stroma composed of column of elongated cells. *Hamathecium* not seen. *Asci* bitunicate, slightly clavate or nearly cylindric. *Ascospores* smooth, hyaline, septate.

Anamorph of these species belong to *Kellermania* Ellis & Everh., 1885. *Conidiomata* pycnidial, immersed, glabrous, ostiolate; wall thick, of textura angularis, cells thick-walled, dark brown to brown in the outer layers, and of textura prismatica, cells thin-walled, colourless to almost colourless in the inner layers, with columnar, thin-walled, colourless cells surrounding the ostiole; ostiole circular or oval, non-papillate. *Conidiophores* lining the cavity of the conidioma, reduced to conidiogenous cells, invested in mucus. *Conidiogenous cells* discrete, often of two kinds: those producing macroconidia, lining most of the conidiomatal cavity, cylindrical to subcylindrical, colourless, smooth; those producing micro-conidia confined to the area of inner wall around the ostiole, ampulliform to broadly ampulliform, colourless, smooth. *Conidiogenesis*: ontogeny holoblastic by apical wall-building in the first conidium and by replacement wall-building in subsequent conidia; maturation by diffuse wall-building synchronous with conidium ontogeny; delimitation by a

transverse septum; secession schizolytic; proliferation usually absent, when present enteroblastic-percurrent to produce an additional conidium at a higher level; periclinal thickenings on conidiogenous cells absent, but an occasional annellation may be present; regeneration of conidiogenous cells absent. *Macroconidia* cylindrical to narrowly clavate with a truncate base, unicellular or euseptate, coIourless, thick-walled, smooth, bearing apical appendages of type A1; appendages arising initially as tubular extensions of the conidium body, with the lumen ultimately reduced or eliminated by centripetal thickening of the wall, single, stout and unbranched, or up to six, attenuated, flexuous. *Microconidia* cylindrical to ellipsoidal or irregular with a rounded or blunt apex and a truncate base bearing minute marginal frills, unicellular, colourless, smooth.

Notes: Planistromella species are known on Agavaceae; *Agave*, *Furcraea* and *Yucca*. Recently, new species *P. opuntiae* has been found on *Opuntia* in the family *Cactaceae* (Sivanesan and Shivas, 2002). There are 10 species recorded in Index Fungorum (http://www.indexfungorum.org, access on 26/06/12). Five species have *Kellermania* anamorph (Ramaley, 1993, 1995, 1998; Barr, 1996). In addition, Aptroot (2006) proposed the new name of five *Mycosphaerella* species to *Planistromella* including *P. cattleyae*, *P. conglomeratiformis*, *P. masjuscula*, *P. operculata*, and *P. zonata*. *Mycosphaerella valgourgensis* was introduced on leaves of *Yucca* species (Crous *et al.*, 2011). The characteristic of *M. valgourgensis* is similar to *Planistromella acervata* which is represent a species complex (Barr, 1996). Nevertheless, *Planistromella yuccifoliorum* and *Kellermania* anamorph were required for further collections to resolve that *M. valgourgensis* is cluster apart from *Planistromellaceae* (Crous *et al*, 2011).

Generic type: *Planistromella yuccifoliorum* A.W. Ramaley

Planistromella yuccifoliorum A.W. Ramaley, *Mycotaxon* 47: 261 (1993). Plate 1a-l.
MycoBank: MB360149

Ascostromata 0.4-0.6 mm diam., up to 0.5 mm high, multilocular, solitary to gregarious, subepidermal, immersed, becoming erumpent, subglobose to ovoid, dark brown to black, with 1-5 locules, thick-walled, the upper part attached with the host epidermis (Plate 1b-e). *Locules* 150 µm wide × 90.4-244.4 µm high, ovoid to globose, the collapsed locule producing conidia or spermatia or both, periphysate ostiole (Plate 1e, f). *Peridium* 84.5-116.1 µm wide, composed brown pseudoparenchymatous cells of *textura globulosa*, dark brown (Plate 1g). *Hamathecium* not seen. *Asci* 93-153.3 × 25-34.6 µm ($\bar{x}$ = 120.8 × 29.3 µm, n = 13), 8-spored, bitunicate, fissitunicate, clavate to saccate, with a pedicel up to 9 µm wide × 7 µm high, and with an ocular up to 2.5 µm wide × 5 µm high (Plate 1h-i). *Ascospores* 32.1-40.2 × 10.8-14.1 µm ($\bar{x}$ = 36.3 × 12.6 µm, n = 15), overlapping 1-2-seriate, ellipsoid and slightly curved with bluntly rounded ends, hyaline, 2-septate, young ascospore with 1-septate, distoseptate, small guttulate, granulate ornamentation (Plate 1j-l).

Anamorph: *Kellermania yuccifoliorum* A.W. Ramaley, *Mycotaxon* 47: 261 (1993).

Conidiomata subepidermal, dark, immersed, erumpent by remaining at the rim cover by epidermis, solitary to gregarious, 250-600(-800) µm diam., up to 500 µm thick, unilocular, ostiolate. Wall cup-shaped, thick-celled, composed 6-12 layers of dark and 2-3 layers of hyaline cells. Conidiogenesis holoblastic. Conidiophores absent. Macroconidiogenous cells short cylindric, hyaline, smooth, each forming acrogenous holoblastic conidium. Macroconidia narrowly ellipsoid-cylindric, the base bluntly rounded, the apex more pointed and often surmounted by an appendage up to 5 µm long, mostly 2-septate, 50-100 × (8-)13-14(-16) µm. Microconidiogenous cells arising on the upper wall of conidioma and in ostiolar

channel. Microconidia more or less cylindric, aseptate, smooth-walled, hyaline 5-10 × 2.5-4 μm. Spermatia formed in the central locule of a stroma or in the locule in the vertical column of the lateral walls of some conidiomata. Spermatogenous cells discrete or integrated on one-celled conidiophores, phialidic, cylindric to elongate-conical, 8-16 × 2-3.5 μm, Spermatia bacillary, hyaline, smooth, 3-7 × 1.5-2.5 μm.

Cultures: none

Sequence data: none.

Material examined: (10 pt) USA: Califonia, San Bernardino County, Roadside 20 miles east of Baker (Hwy. 91/466), on leaves of *Yucca brevifolia* Engelmann, 14 April 1960, Isabelle Tavares No.466 (UC 1202973, **holotype**).

Notes:

Morphology

Planistromella is the generic type of *Planistromellaceae* (Barr, 1996). The first *Planistromella* species was established by Ramaley (1993) which is the type species and the anamorph *Kellermania yuccifoliorum* is introduced. The genus *Planistromella* is clearly different from other genus in Planistromellaceae forming biseptate ascospores. *Stanogospora gigantea* which formerly was described by Heald and Wolf (1911) has been reexamined by Ramaley (1992). Ramaley designed to accommodate new fungi *Piptarthron pluriloculare. Piptarthron* is the anamorph of *Planistroma* and different from *S. gigantea* in size and shape of conidia. *Kellermania* has apically appendaged conidia but these are lacking in *Piptarthron.*

Phylogenetic study: *Mycosphaerella valgourgensis* was introduced on leaves of *Yucca* species (Crous *et al*, 2011). The characteristic of *M. valgourgensis* is similar to *Planistromella acervata* which is represent a species complex (Barr, 1996). Nevertheless, *Planistromella yuccifoliorum* and *Kellermania* anamorph were required for further collections to resolve that *M. valgourgensis* is cluster apart from *Planistromellaceae* (Crous *et al*, 2011).

Concluding remarks:

Planistromella species are known on Agavaceae; *Agave*, *Furcraea* and *Yucca*. Recently, new species *P. opuntiae* has been found on *Opuntia* in the family *Cactaceae* (Sivanesan and Shivas, 2002). There are 10 species recorded in Index Fungorum (http://www.indexfungorum.org, access on 26/06/12). Five species have *Kellermania* anamorph (Ramaley, 1993, 1995, 1998; Barr, 1996). In addition, Aptroot (2006) proposed the new name of five *Mycosphaerella* species to *Planistromella* including *P. cattleyae*, *P. conglomeratiformis*, *P. masjuscula*, *P. operculata*, and *P. zonata*.

Planistroma A.W. Ramaley, *Mycotaxon* 42: 69 (1991)
MycoBank: MB25358 Fig. 2

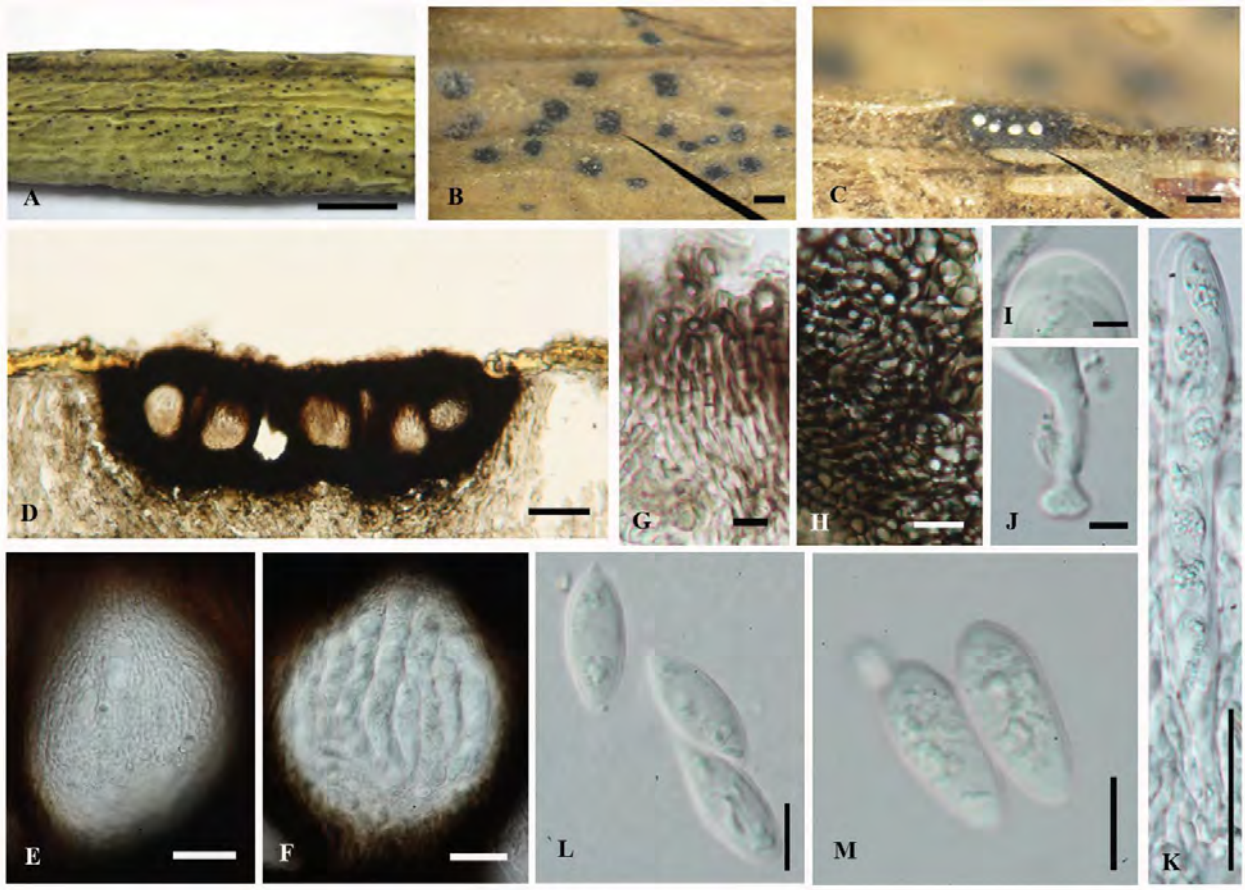


Fig. 2 ***Planistroma yuccigenum*** (holotype) on leaves of *Yucca baccata.* A-**B.**Ascostromata on the host surface. C-**D.** Section of ascostroma. **E.** Young ascoma. **F.** Mature ascoma. **G**. Peridium at the side of ascoma. **H.** Peridium at the base of ascoma. **I.** Ocular chamber **J.** Pedicel. **K.** Asci. **L-M.** ascospore. **Scale bars: A** = 1 cm, **B** = 1 mm, **C** = 0.5 mm, **D** = 200 µm**, E-F** = 40 µm**, G-H =** 10 µm, **I-J**= 5 µm, **K** = 20 µm,**L-M** = 10 µm

Ascostromata subepidermal, immersed, partially erumpent remaining at the rim cover by epidermis, solitary to gregarious, multilocular, hemispherical, thick-walled. *Locules* ovoid to globose, developing in the same stroma of the conidiogenous and/or spermatogenous locules, callapsing with the empty locule which previously producing conidia or spermatia or both, periphysate ostiole. *Peridium* composed of columns of cells at the top layer of the stromata, the subdividing the stromata into locules. *Hamathecium* none. *Asci* bitunicate, cylindrical. *Ascospores* ellipsoid, sometimes surrounded by a slime layer.

Anamorph of these species belong to *Piptarthron* and *Kellermania.* Most species have anamorphs of *Piptarthron* Mont. ex Höhn., 1918. *Mycelium* immersed, dark brown, branched, septate. *Conidiomata* pycnidial, amphigenous, separate or aggregated, globose, dark brown, immersed, unilocular or multilocular, thick-walled; wall composed of loosely aggregated, dark brown, thick-walled textura angularis. *Ostiole* central, papillate or depressed, circular, wall tissue often elongated. *Conidiophores* absent. *Conidiogenous cells* holoblastic, determinate, discrete, doliiform, hyaline, smooth, formed from the inner cells of the pycnidial wall. *Conidia* hyaline, 0-4 euseptate, often constricted, base truncate with a distinct marginal frill, thick-walled, smooth, often guttulate, cylindrical, fusiform, straight or curved.

Notes: Planistroma species are on Agavaceae (Barr, 1996). Nowadays, there are 4 recorded species including *P. kellermania, P. nolinae, P. obtusilunatum* and *P. yuccigenum* (Ramaley, 1991, 1992, 1995, 1998). All species produced *Piptarthron* as anamorph excepting *P. kellermania* has *Kellermania* anamorph (Ramaley, 1991, 1992, 1995, 1998). No *Planistroma* species have been obtained in culture and no sequence data is deposited in GenBank.

Generic type: *Planistroma yuccigenum* A.W. Ramaley

Planistroma yuccigenum A.W. Ramaley, *Mycotaxon* 42: 69 (1991). Plate 2a-n.
MycoBank: MB358836

Ascostromata black ellipsoid to subcircular on surface of leaves, subepidermal, immersed, partially erumpent remaining at the rim cover by epidermis, solitary to gregarious, multilocular, hemispherical, 0.5-0.7 mm diam., forming elongate stroma up to 1 mm long, up to 0.5 mm thick, dark brown to black, 2-10 locules, variable arrangement of locules, thick-walled (Plate 2b-e). *Locules* 43-153.4 µm wide × 55.7-160.9 µm high, ovoid to globose, developing in the same stroma of the conidiogenous and/or spermatogenous locules, callapsing with the empty locule which previously producing conidia or spermatia or both, periphysate ostiole (Plate 2f, g). *Peridium* 48.1-127.4 µm wide at the side, composed of column of elongate cells, reaching from the base to the top, brown, 81.5-196.8 µm wide at the base, composed pseudoparenchymatous cells of *textura globulosa*, dark brown, cup-shaped wall (Plate 2h, i). *Hamathecium* none. *Asci* 72.9-110.9 × 13.8-18.2 µm ($\bar{x}$ = 89.3 × 16 µm, n = 30), 8-spored, bitunicate, fissitunicate, cylindrical, with a long pedicel 7-8 µm up to 15 µm high, at the stipe 5.5 µm wide, and with an ocular up to 2.6 µm wide × 2.9 µm high (Plate 2j-l). *Ascospores* 15.5-21.7 × 7-10.5 µm ($\bar{x}$ = 18.8 × 8.5 µm, n = 30), overlapping 1-2-seriate, ellipsoid with broadly rounded ends at the apex and narrowly rounded ends or tapering toward the base, hyaline, aseptate, irregularly many guttulates, rough (Plate 2m, n).

Anamorph: *Piptarthron pluriloculare* A.W. Ramaley, *Mycotaxon* 42: 69 (1991).

Stromata subepidermal, black, immersed, erumpent by remaining at the rim cover by epidermis, solitary to gregarious, hemispherical, 0.4-1 mm diam, up to 0.5 mm thick, multilocular, 4-30 or more locules in stromata. Wall cup-shaped, 70-100 µm thick, composed of textura angularis, thick-walled, dark-brown, one ostiole per locule, 8-15 µm wide. Conidiophores absent. Microconidiogenous cells form on the wall of locules, short cylindric, hyaline, smooth, 6.5-14.5 × 3.5-5.5 µm, each forming acrogenous holoblastic conidium. Macroconidia fusiform, curved or bent, tapering toward the apex or the base, base truncate, mostly aseptate, smooth, hyaline, (48-)59-76(-98) × (4-)5.5-7(-8) ($\bar{x}$ = 67.8× 6.5) µm. Microconidia cylindric, irregularly swelled or bent, aseptate, smooth-walled, hyaline 5.5-25 × 2.5-3.5 µm. Spermatia formed on a part of walls of a macroconidiogenous locule or in one or more separate locules in a stroma. Spermatogenous cells phialidic, cylindric to elongate-conical, ca 8-16 × 1.5-3 µm, discrete or integrated on a one-celled conidiophores, determinate, hyaline, smooth, forming acrogenous spermatia, Spermatia bacillary, hyaline, aseptate, smooth, 3.5-5.5 × 1.5 µm.

Cultures: none.

Sequence data: none.

Material examined: (10 pt) USA: Colorado, La Plata County, Durango, below Fort Lewis College, Roadside, on leaves of *Yucca baccata*, 26 September 1990, Annette W. Ramaley, (UC 1475061, **holotype**).

Notes:

Morphology

Planistroma was established by Ramaley (1991) with *Planistroma yuccigenum* as the type species. *Planistroma* is characterized by sub-epidermal, semi-erumpent, ostiolate ascomata, epidermis remove, not differentiable perithecial wall and hyaline aseptate ascospores (Ramaley, 1991). This genera is similar to *Planistromella* in stromata character and type of substrate but *Planistromella* has biseptate ascospores. *Bagnisiella* is also resembled to *Planistroma* in having bitunicate asci, hyaline aseptate ascospores and stroma contain old coelomycete locules. However in *Bagnisiella,* the stroma are erumpent and the ascospores are released through rupture of stroma, not through an ostiole (Ramaley, 1991).

Ramaley (1991) was erected *Piptarthron pluriloculare* as the anamorph of *Planistroma yuccigenum* by confirmation of the anamorph-teleomorph connection. *Piptarthron* has thick-walled ostiolate conidiomata, holoblastic conidiogenous cell and conidia lacking appendages. *Piptarthron* is resembled with *Kellermania* which some species are anamorph of *Planistromella,* but *Kellermania* has apically appendaged conidia (Ramaley, 1995). Unexpectedly, *Planistroma kellermania* produced conidia of *Kellermania nolinae* (Ramaley, 1998).

Phylogenetic study: none

Concluding remarks:

Planistroma species are on Agavaceae (Barr, 1996). Nowadays, there are 4 recorded species including *P.kellermania, P.nolinae, P.obtusilunatum* and *P.yuccigenum* (Ramaley, 1991, 1992, 1995, 1998). All species produced *Piptarthron* as anamorph excepting *P.kellermania* has *Kellermania* anamorph (Ramaley, 1991, 1992, 1995, 1998).

Mycosphaerellopsis Höhn., Annales Mycologici 16(1/2): 157 (1918)
MycoBank: MB3345 Fig. 3

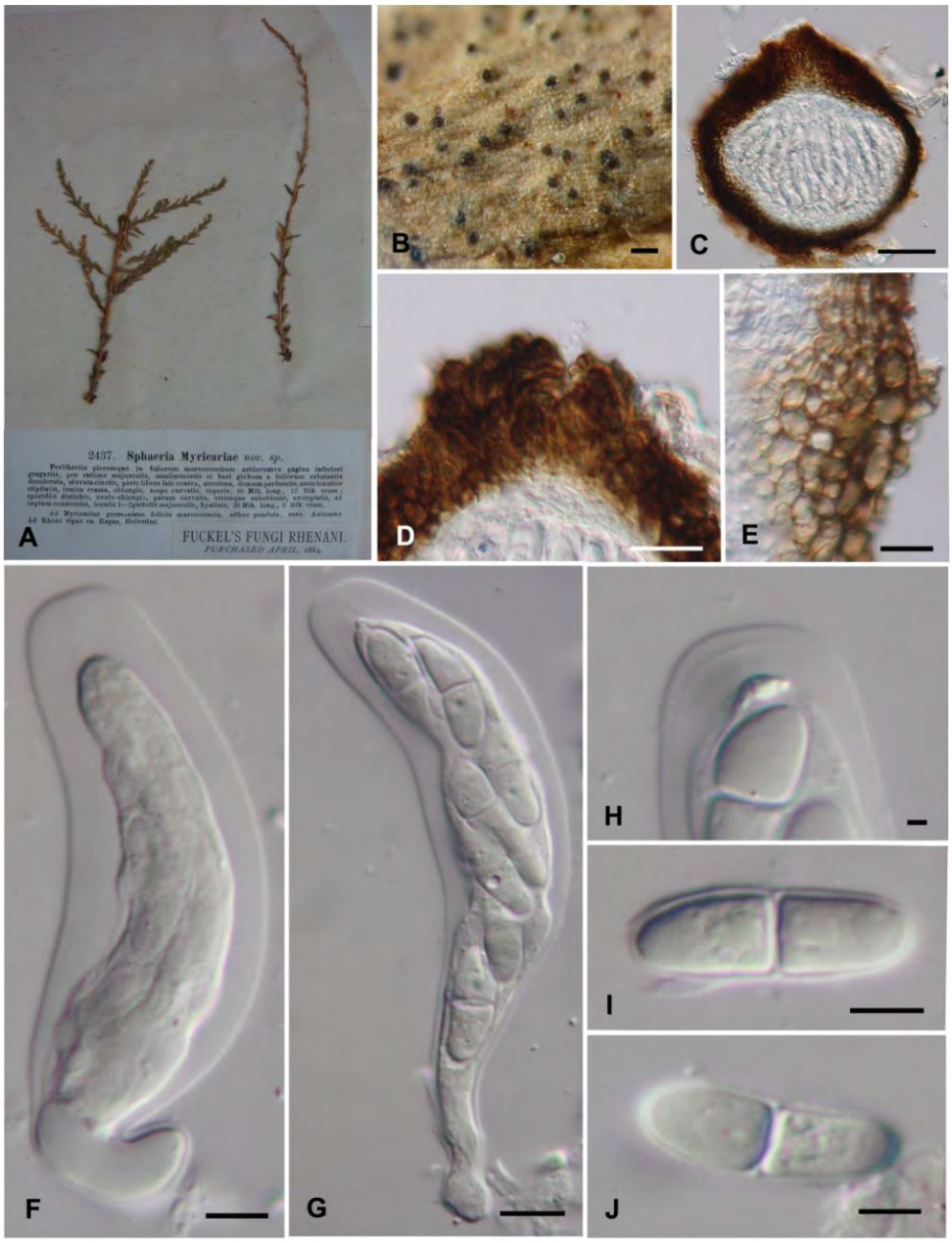


Fig. 3 ***Sphaeria myricariae*** Fuckel, on dead leaves of *Myricariae germanicae*. **A-B.** Ascomata on the host surface. **C.** Section of an ascoma. **D.** Close up of the ostiole. Note periphysate. **E.** Section of the peridium. **F.** Immature ascus. **G.** Mature ascus. **H.** Close up of ocular chamber. **I-J.** Ascospores. **Scale bars: B** = 200 µm, **C** = 30 µm, **D** = 10 µm**, E** = 5 µm, **F** = 3 µm**, G =** 5 µm, **H** = 1 µm, **I-J** = 3 µm.

Ascostromata uniloculate, immersed, periphysate ostioles. Hemathecium not seen. *Asci*,bitunicate, oblong to saccate. *Ascospores* broadly obovoid, light yellowish, didymosporae.
Anamorph: present in the *Myricariae* leaves ; pycnidial conidiomata, the short conidiogenous cells are holoblastic, producing hyaline, oblong, uniseptate conidia.

Notes: There are 2 species recorded in *Mycosphaerellopsis*; *M. myricariae* and *M. moravica* (http://www.indexfungorum.org, access on 26/06/12). No *Mycosphaerellopsis* species have been obtained in culture and no sequence data is deposited in GenBank.

Generic type: *Mycosphaerellopsis myricariae* (Fuckel) Höhn.

Sphaeria myricariae Fuckel, Jb. nassau. Ver. Naturk. 27-28: 22 (1874) [1873-74]
Mycosphaerellopsis myricariae (Fuckel) Höhn., Annales Mycologici 16(1/2): 157 (1918)
Plate 3A-J.

MycoBank: MB499606

≡ *Sphaeria myricariae* Fuckel, Jb. nassau. Ver. Naturk. 27-28: 22 (1874) [1873-74]**.**

Ascomata 76.8-145.7 µm high ($\bar{x}$ = 110 µm, n = 25) × 93.4-156.4 µm ($\bar{x}$ = 117.3 µm, n = 25) diam., uniloculate, solitary, gregarious, immersed to semi-immersed, globose to subglobose, brown to dark brown (Plate 3B, C). *Ostioles* periphysate (Plate 3D). *Peridium* 12-22 µm wide, composed of 3-6 layers, *textura globulosa*, brown (Plate 3E). No pseudoparaphyses. *Asci* 45.5-64.3 × 9.9-12.6 µm ($\bar{x}$ = 53.7 × 11.5 µm, n = 25), 8-spored with 1-3 seriate partially overlapping, bitunicate, fissitunicate, oblong to saccate, with an ocular chamber 1-3 µm wide × 0.5-1.7 µm high, and with a pedicel, 3.6-4.9 µm long (Plate 3F-H). *Ascospores* 10.1-11.8 × 3.8-4.8 µm ($\bar{x}$ = 10.9 × 4.2 µm, n = 25), ellipsoid to broadly obovoid with broadly rounded end and narrowly rounded on another end, hyaline, didymosporae, distoseptate, two-celled with one guttulate in each cell, slightly roughened (Plate 3I-J).

Anamorph: present in the *Myricariae* leaves ; pycnidial conidiomata, the short conidiogenous cells are holoblastic, producing hyaline, oblong, uniseptate conidia, 22.5-29 × 5-6.4 µm (Barr, 1996).

Cultures: none.

Sequence data: none.

Material examined: UK: England, KEW, Royal Botanic Gardens; on *Myricariae germanicae*, April 1884, Fuckel's fungi rhenani (2437, **holotype**).

Notes:
Morphology

Mycosphaerellopsis was a member of the Pseudosphaeriaceae (Höhnel, 1918). Barr (1996), erected the family Planistromellaceae including the genus *Mycosphaerellopsis* distinguished from Pseudosphaeriaceae in the formation of locules open schizogenously by periphysate ostioles. Characteristic of *Mycosphaerellopsis* is different from other genus in *Planistromellaceae* in having uniloculate ascomata, 1-septate and broadly obovoid ascospores (Barr, 1996).

Phylogenetic study: none.
Concluding remarks: There are 2 species recorded in *Mycosphaerellopsis*; *M. myricariae* and *M. moravica* (http://www.indexfungorum.org, access on 26/06/12).

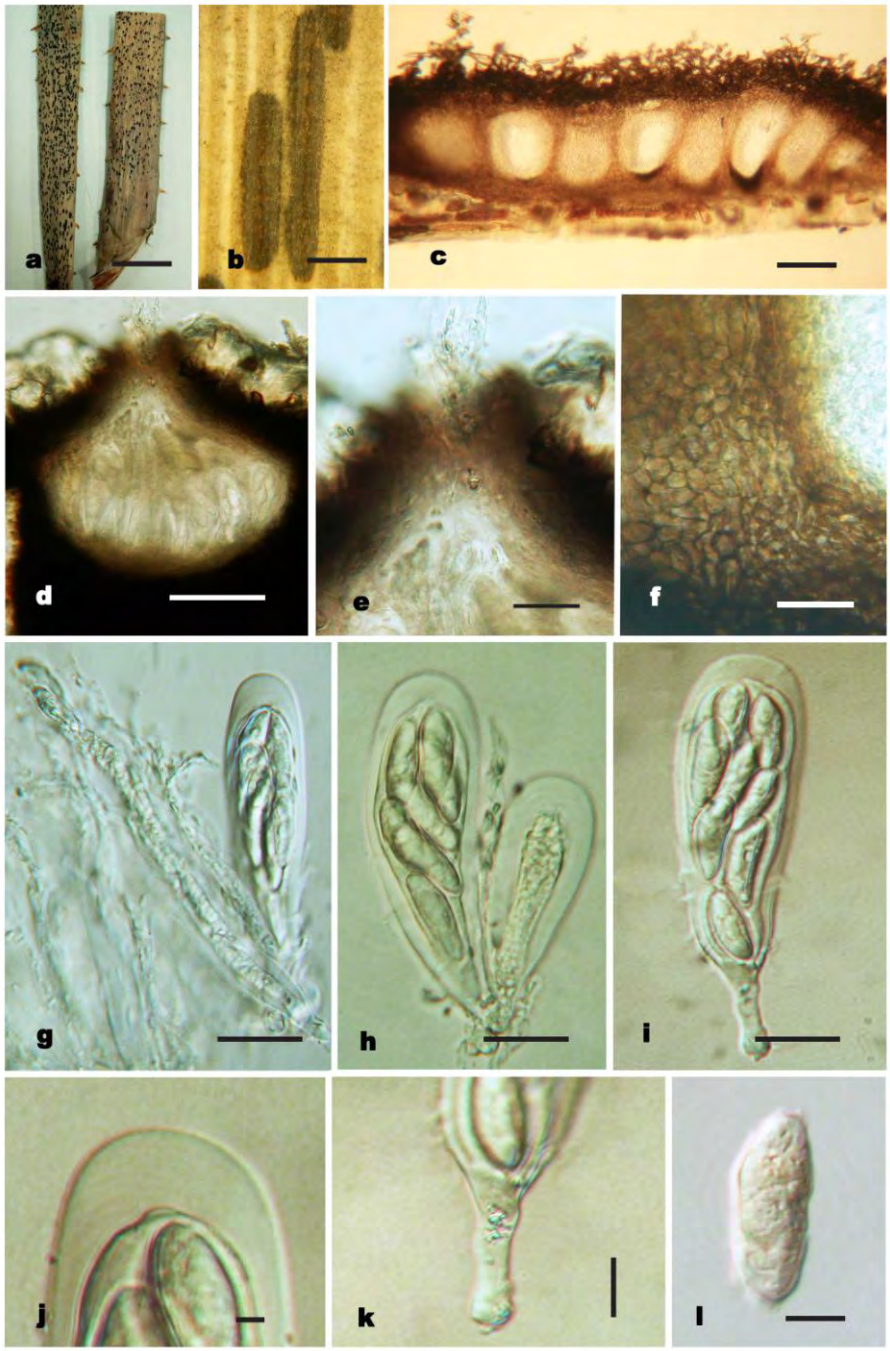


Fig 4. *Comminutispora agavacearum* (holotype) on dead leaves of *Dasylirion leiophyllum*. a-b. Stromata on host surface. c. ascostromata. d-e. Close-up of ascoma and ostiolar. f. Close-up of peridium. g-i. Ascus with ascospores, composed of paraphyses. j-k. Close-up of ocular chamber, thickened apex, and with stalk. l. Mature ascospores. Scale bars = a-c = 100 µm, d-f = 10 µm, g-i = 25 µm, j-k = 5 µm, i = 10 µm.

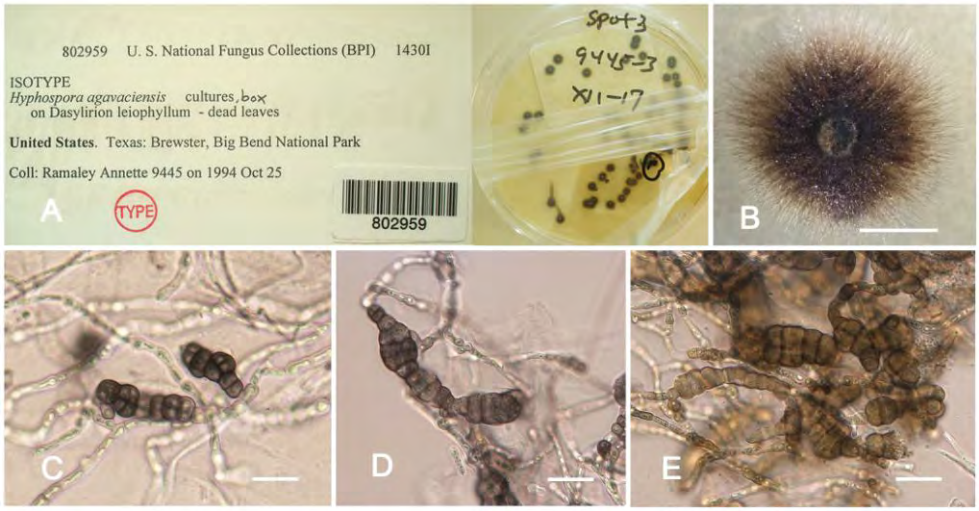


Fig 5. *Hyphospora agavaciensis* (on dried cultures at 1 week). A. Material on dried cultures. B. Colonies on MA. C-E. Dictyochlamydospores. Scale bars: B = 500 µm, C-E = 20 µm.

Ascomata immersed, separate, uniloculate, a amall papilla penetrating the epidermis. *Asci* bitunicate, basal, 8-spored. Pseudoparaphyses lacking, interascal cells plentiful even at maturity, ostiolar canal periphysate. *Ascospores* transversely and longitudinally septate, ultimate segments forming secondary ascospores within the ascospores which are liberated into the ascus.

Anamorph of *Comminutispora* is Hyphospora *Colonies* composed of a somewhat depressed hemi-spheric central part surrounded by a halo of hyaline hyphae in the agar. *Conidiogenous* initials much divided by longitudinal and transverse septa, the small conidiogenous cells formed producing endoconidia. *Endoconidia* unicellular, liberated by breakdown of a part of the cell walls.

Generic type: *Comminutispora agavacearum* A.W. Ramaley [as 'agavaciensis']

Comminutispora agavacearum A.W. Ramaley [as 'agavaciensis'], Mycologia 88(1): 133 (1996). Plate 4a-g.

MycoBank: MB414805

Ascostromata 642.6-1,999.2 µm length × 285.6-428.4 µm width, measured at the surface of host, with 300-600 µm high × 250-300 µm diameters within host tissue, apothecium and elongate hysterothecium, scattered to loosely, immersed, dark brown to black, rather dull, carbonaceous, thick-walled, composed of textura angularis, opening by a sunken longitudinal slite (Plate 4a-f). *Asci* 105-135×27.5-35 µm ($\bar{x}$ = 20.3×29.3 µm, n = 10), bitunicate, saccate, clavate or cylindrical, 8-spored, apex wall 5-7.5 µm thick, with ocular chamber up to 5 µm width × 2.5 µm high, and with pedicel 5-22.5 µm long (Plate 1g-k). *Ascospores* 22.5-35.5×7.5-12.5 µm ($\bar{x}$ = 27.5×9.1 µm, n = 20), amerosporae, biseriate and overlapping, obovoid, ellipsoid to fusoid with broadly to narrowly rounded ends, one-celled, non-septate, roughened, with thin gelatinous coat (Plate 4l).

Anamorph: *Hyphospora agavacearum* A.W. Ramaley [as '*agavaciensis*'], *Mycologia* 133 (1996). Plate 5a-g.

Colonies growing on MA (Malt Agar), reaching 500 µm diam. in one week at 23-25 °C, flat to slightly effuse, radiating, edge fimbriate, hyphae generally dark brown on surface view, outward hyphae pale brownish to hyaline, darkened interior, slightly raised hairy, partly

superficial and immersed (Plate 2a-b). The small conidiogenous cells differentiated from vegetative hyphae to developing endoconidia of dictyochlamydospores, elongation or long chains, variable shape, with (36.5-)93-150(-167) µm long × 15-23(-32.5) µm wide, with transverse and longitudinally septation, dark brown pigmented, constricted at the septa, smooth or slightly verrucose (Plate 5c-e).

Cultures: CBS 619.95 (**unverified**, SSU – Y18699), (**unverified**, LSU –EU981286).

Sequence data: Genbank Y18699, Genbank EU981286.

Material examined: ***Comminutispora agavacearum*** U.S.A.: Texas, National Fungus Collections, on dead leaves of *Dasylirion leiophyllum*, October 1994, (BPI 802958, **holotype**).

Hyphospora agavacearum U.S.A.: Texas, Brewster, Big Bend National Park, on dead leaves of *Dasylirion leiophyllum*, 25 October 1994, Ramaley Annette 9445,(BPI 802959, **holotype**).

Notes: Morphological character of the type specimen seems with *Botryosphaeria* sp.

Morphology

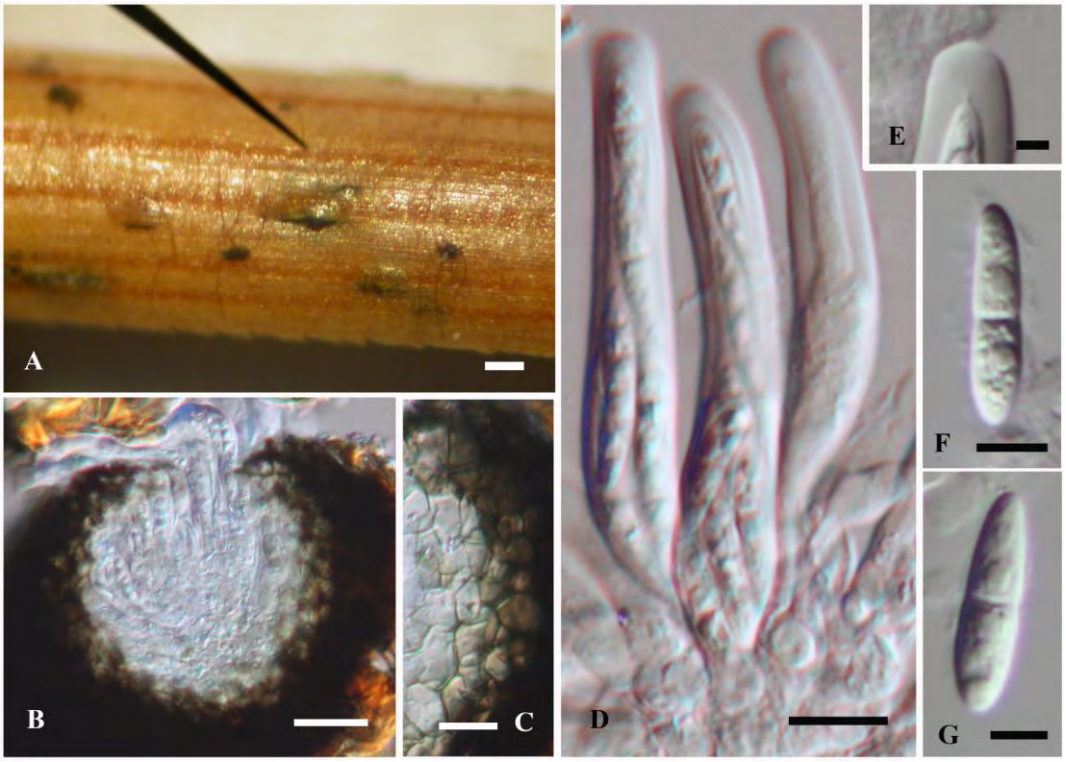
Comminutispora was established by Ramaley (1996) as a monotypic genus based on *Comminutispora agavacearum* and *Hyphospora agavaciensis* as an anamorph. *Comminutispora agavacearum* is characterized by unilocular ascomata, immersed, ostiolar canal periphysate and forming transversely and longitudinally septate ascospores (Ramaley, 1996). So, Ramaley (1996) placed the genus *Comminutispora* in the Dothidiales *sensu.* However, following the classification by Lumbsch and Huhndorf (2010) *Comminutispora* is a member of the family *Planistromellaceae*. *H. agavaciensis* was produces melanized endoconidia are flattened, with colour and texture of colonies are difference, which are depend on media (Zalar *et al*.; 1998), while Tsuneda *et al*. (2004) reexamined between *H. agavaciensis* and *Endoconidioma populi* because both species are similar with endoconidiogenesis for comfirmation, *H. agavaciensis* was transverse and longitudinal divisions of hyphal cell.

Phylogenetic study: Molecular analysis on of the type species; *Comminutispora agavacearum* strain CBS 619.95 was studied by Hambleton *et al.*, 2003 and Schoch *et al.*, (2009) on the basis of SSU and LSU genes. DNA sequence data for SSU showed that *C. agavacearum* presented in a large clade of Capnodiales (Hambleton *et al.*, 2003). In addition, *C. agavacearum* is unrelated with *Planistromellaceae* base on SSU and LSU genes (Schoch *et al.*, 2009).

Concluding remarks:-

Mycosphaerellaceae insertae cedis

Eruptio (Dearn.) M.E. Barr, Mycotaxon 60: 437 (1996)
MycoBank: MB27768 Fig.6



Figs 6. *Eruptio acicola* (holotype) on withered needles of *Yucca brevifolia*. A. Ascostromata on the host surface. B. Section of ascostroma. C. Peridium. D. Asci. E. Ocular chamber. F-G. Ascospore. Scale bars: A = 50 µm, B = 25 µm, C = 20 µm, D = 10 µm, E = 3 µm, F-G = 5 µm.

Ascostromata multiloculate or uniloculate, elongate, erumpent from substrate, pseudoparenchymatous cells, reddish or dark brown, hyphae abundant in leaf tissues. *Locules* small, ostioles schizogenously formed, peri-physate. *Asci* functionally bitunicate, oblong to saccate, basal, pushing into and compressing interthecial cells. *Ascospore* hyaline, oblong fusoid, one septate; wall smooth; minutely guttulate.
Anamorph state are recorded as *Lecanosticta* and *Dothistroma. Lecanosticta* Syd., 1922: *Mycelium* immersed, branched, septate, pale brown. *Conidiomata* acervular, subepidermal, separate, formed of brown, thin- or thick-walled textura angularis. Dehiscence by pushing back a flap of epidermis that remains attached. *Conidiophores* hyaline to pale brown, branched, septate, smooth, formed from the upper cells of the pseudoparenchyma. *Conidiogenous cells* holoblastic, integrated or discrete, indeterminate, cylindrical, hyaline, with 1-2 often widely spaced percurrent proliferations. *Conidia* acrogenous, straight or curved, fusiform, tapered to the rounded apex and truncate base, l-3 euseptate, continuous, pale brown, verrucose.
Notes: Presently, *Eruptio* has three species; *E. acicola*, *E. pini* and *E. gaubae* (http://www.indexfungorum.org, access on 15/07/12). The classification of these fungi is confused by morphological characters which they are very similar to *Mycosphaerella.* Molecular analysis on of the type species; *Eruptio acicola* was studied by Crous *et al*., (2001) based on ITS rDNA sequence. The results showed that *Eruptio* was in the same clade with

Mycosphaerella. This result has also been supported by several studies ; Verkley *et al.*, (2004) based on LSU rRNA gene, Crous *et al*., 2009a based on SSU, ITS, LSU rRNA genes and Crous *et al*., 2009b based on LSU rRNA gene. All of results have been showed that *Lecanosticta* clustered with other anamorphs of *Mycosphaerella.* So, the genus *Eruptio* might be placed to *Mycosphaerellaceae.*

Generic type: *Eruptio acicola* (Dearn.) M.E. Barr

Eruptio acicola (Dearn.) M.E. Barr, *Mycotaxon* 60: 438 (1996). Plate6a-g.

MycoBank: MB436296

≡ ***Oligostroma acicola*** Dearn, *Mycologia* 18(5): 251 (1926).
≡***Dothidea acicola*** (Dearn.) M. Morelet, *Annales de la Société des Sciences Naturelles et d'Archéologie de Toulon et du Var* 177: 9 (1968).
≡ ***Mycosphaerella dearnessii*** M.E. Barr, *Contr. Univ. Mich. Herb.* 9: 587 (1972).
≡ ***Scirrhia acicola*** (Dearn.) Sigg., *Phytopathology* 29: 1076 (1939).
≡ ***Systremma acicola*** (Dearn.) F.A. Wolf & Barbour, *Phytopathology* 31: 70 (1941).

Ascostromata 0.1-0.4 mm diam., up to 1.5 mm. high, uniloculate or multiloculate, linear, scattered to gregarious, immersed, erumpent, globose to subglobose, black (Plate 6a, b). *Locules* 87.8-98.2 µm high × 61.3-83.5 µm diam., small, ovoid to globose, ostioles schizogenously formed, periphysate (Plate 6b). *Peridium* 24.4-28.1 µm thick, reddish brown to dark brown, composed pseudoparenchymatous cells of *textura globulosa* (Plate 6c). *Hamathecium* not seen. *Asci* 40.1-58.6 × 8.6-14.1 µm ($\bar{x}$ = 50.9 × 10.4 µm), obclavate, usually wider in the base and narrowed towards the apex, 8-spored, pedicellate, ocular chamber 0.5 µm wide (Plate 6d, e). *Ascospores* 10.8-15.4 × 2.2-4.4 µm ($\bar{x}$ = 13.8 × 3.5 µm), 1-septate, oblong to cuneate, bluntly rounded at one end, tapering fusiform at other, hyaline, uniseriate on the top, biseriate on the middle and triseriate on the base of asci with partially overlapping, rough, 4-guttulate (Plate 6f, g).

Anamorph: *Lecanosticta acicola* (Thüm.) Syd., in Sydow & Petrak, *Annls mycol.* 22(3/6): 400 (1924).
≡ *Cryptosporium acicola* Thüm. (1878)
≡ *Septoria acicola* (Thüm.) Sacc. (1884)
≡ *Lecanosticta pini* H. Sydow apud Sydow & Petrak (1922)

Stromata elongate, erumpent, the locules opening widely, the bases lined with short hyaline conidiophores; conidia 20-28 X 2.5-3 µm, brown, elongate, tapered at apex, blunt at base, 3-septate, bent, the wall roughened. Microconidial state: locules in immature stromata contain microconidia, these hyaline, 2-3 X 1 µm, 1-celled.

Cultures: CBS: 322.33 (**unverified**, LSU - AY152618), STE-U 3391 (**unverified**, ITS), CBS 871.95; MPFN 314 (**unverified**, LSU - GQ852598), (**unverified**, SSU - GU214663), (**unverified**, ITS - GU214663), (**unverified**, LSU - GU214663).

Sequence data: GenBank AY152618, GenBank GQ852598 , GenBank GU214663, GenBank GU214663, GenBank GU214663.

Material examined: CANADA: Ontario, Ottawa; on withered needles of *Pinus Palustris*, 27 February 1919, G. G. Hedgcock: 32146 (D:5831, **holotype**).

Notes:

Morphology

This fungus has a complicated history with many synonyms for many years. *Oligostroma acicola* was established by Dearness (1926) on needles of *Pinus palustris*.

Barr (1972) placed the fungus to the genus *Mycosphaerella* using locule and ascus development as generic concepts rather than the position of ascocarp. Barr (1996), later erected the family *Planistromellaceae* including the new genus *Eruptio* based on multilocular pseudothecia, open via schizogenously form, periphysate ostioles. *Eruptio* is different from other genus in *Planistromellaceae* on character of ascospores having 1-septate, narrow, oblong to fusoid. Their anamorphs cause the important diseases of pine needles blight: *Lecanosticta acicola* (*E. acicola*) and *Dothistroma pini* (*E. pini*) (Evan, 1984). These anamorphs form acervuli in the stromata, conidia hyaline to brown, septate, cylindrical. Whereas, *Lecanosticta* has 1-3 septate conidia and produces microconidia but *Dothistroma* has 1-5 septate conidia (Barr, 1972 and Barnes *et al*., 2004). Other species that has an anamorph of *Lecanosticta* is *L. gaubae* (*E. gaubae*) which has been transferred from *Mycosphaerella* by Crous (1999) because of the different ascomata character and no *Lecanosticta* species are known to have anamorphs of *Mycosphaerella.*

Phylogenetic study: Molecular analysis on of the type species; *Eruptio acicola* was studied by Crous *et al*., (2001) based on ITS rDNA sequence. The results showed that *Eruptio* was in the same clade with *Mycosphaerella.* This result has also been supported by several studies ; Verkley *et al.*, (2004) based on LSU rRNA gene, Crous *et al*., 2009a based on SSU, ITS, LSU rRNA genes and Crous *et al*., 2009b based on LSU rRNA gene. All of results have been showed that *Lecanosticta* clustered with other anamorphs of *Mycosphaerella.*

Concluding remarks

Presently, *Eruptio* has three species; *E. acicola*, *E. pini* and *E. gaubae* (http://www.indexfungorum.org, access on 15/07/12). The classification of these fungi is confused by morphological characters which they are very similar to *Mycosphaerella.* Recently, many phylogenetic studies have strongly supported that *Eruptio* is a synonym of *Mycosphaerella.* So, the genus *Eruptio* might be placed to *Mycosphaerellaceae.*

Microcyclus *Sacc*., Syd. & P. Syd., in Sydow & Sydow, Annales Mycologici 2(2): 165 (1904)
MycoBank: MB3160 Fig. 7

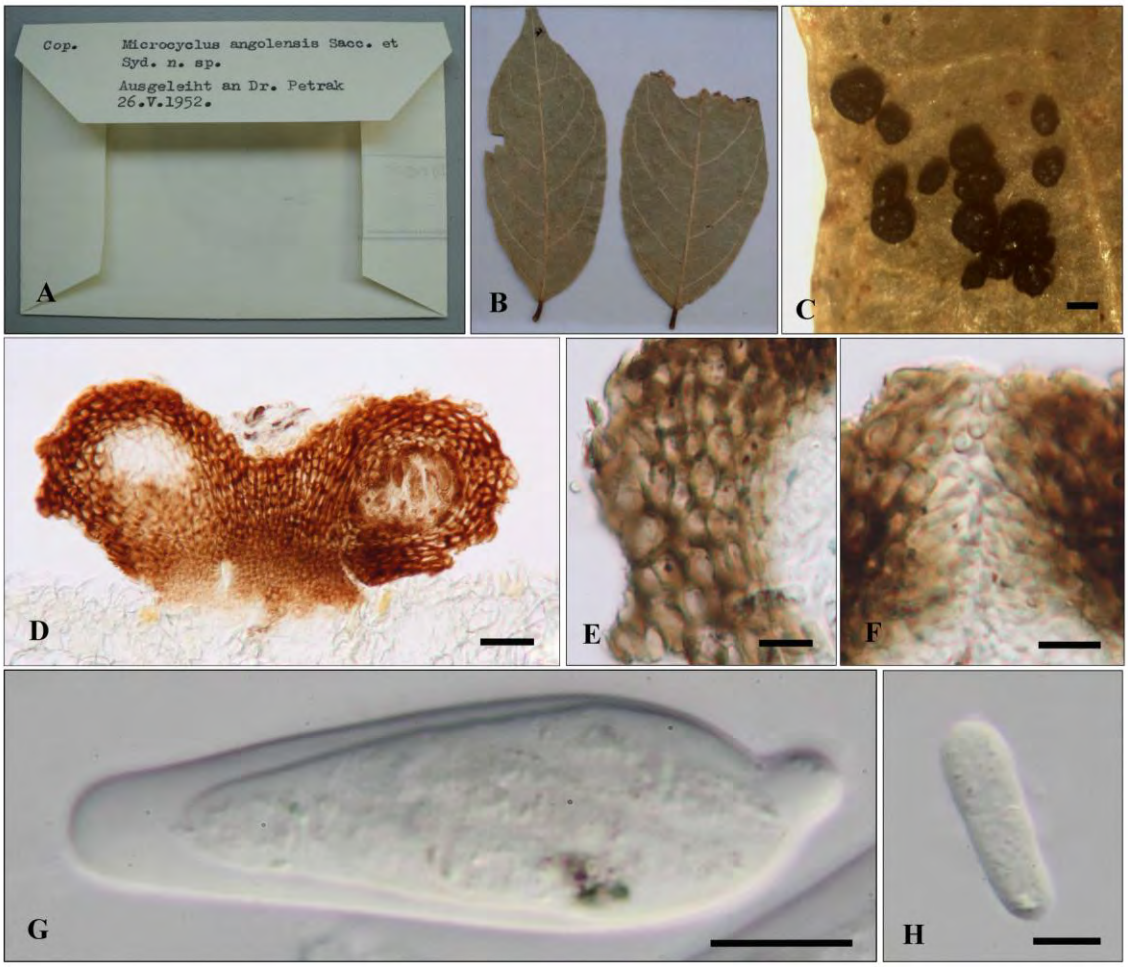


Fig. 7 ***Microcyclus angolensis*** (holotype) on living leaves of *Millettiae thonningii*. **A.** Herbaria packet. **B-C.** Ascostromata on the host surface. **D.** Section of ascostromata. **E.** Section of peridium. **F.** Periphysate ostiole. **G.** Mature ascus. **H.** Ascospore. **Scale bars: C** = 200 µm, **D** = 30 µm**, E-G =** 10 µm, **H** = 5 µm.

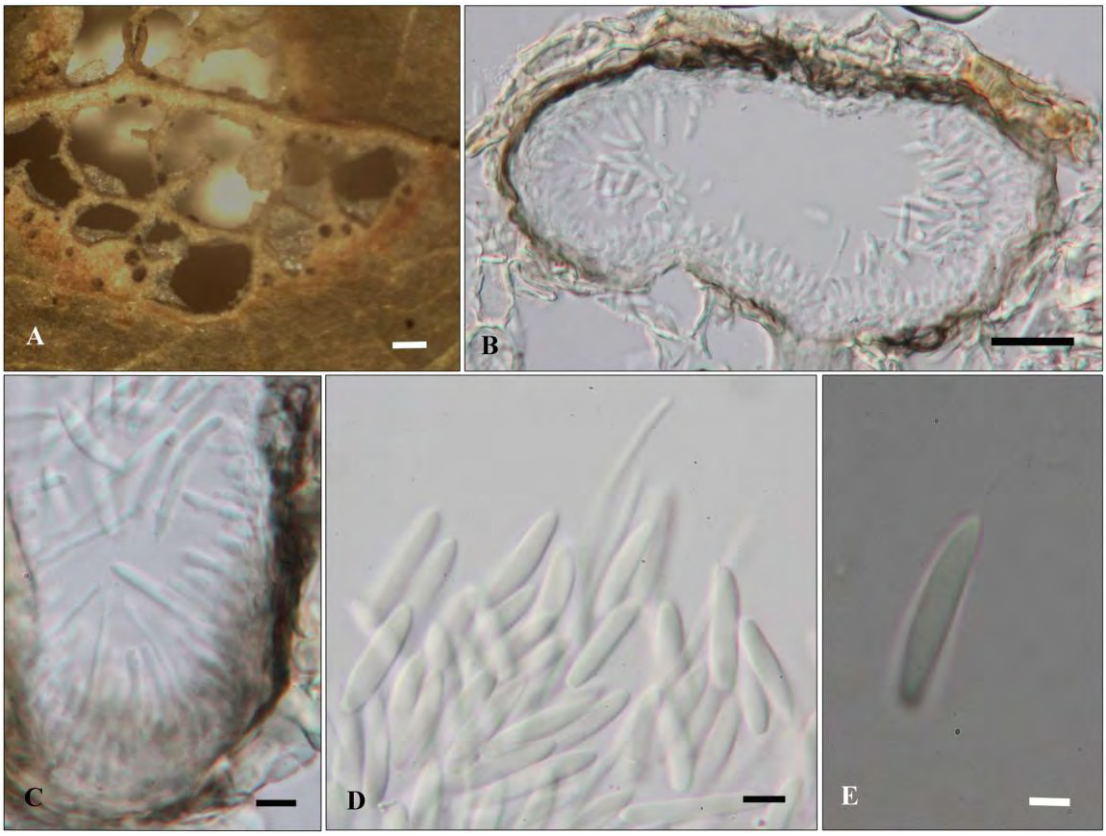


Fig. 8 Anamorph of *Microcyclus angolensis* (holotype), on living leaves of *Millettiae thonningii*. **A.** Conidiomata on the host surface. **B.** Section of conidioma. **C.** Conidiogenous cells. **D.** Conidia and paraphyses. **E.** Conidia with appendage. **Scale bars: A**, **B** = 100 µm, **C, D** = 5 µm**, E** = 3 µm.

Ascostromata variable, pulvinate, crust-like or knob-shaped developing on a basally to centrally attached tapered foot-like hypostroma on leaves, rarely on petioles, erumpent to superficial, multilocular with each locule provided with an apical papillate ostiole. The stroma is composed of irregularly rounded to angular, thick-walled, dark brown cells forming a *textura angularis* or *prismatica* and the cells of the hypostroma are usually elongated, compressed, or they may form a hyphal mat in the mesophyll tissue. *Asci* thick-walled, 8 spored, clavate to clavate-cylindric. *Ascospores* 1-septate usually slightly unequally or rarely in the middle, not or slightly constricted at the septum, mostly hyaline, rarely very pale brown, sometimes mature ascospores may become up to 3-septate. *Pseudoparaphysoids* filiform, hyaline, usually disappearing in mature stromata.

There are two anamorphs of *Microcyclus* species including *Fusicladium* Bonorden, 1851 and *Pazschkeella* H. & P. Sydow, 1901. *Fusicladium* Bonorden, 1851 is described by *Mycelium* immersed, sometimes subcuticular. *Stroma* often present, sometimes subcuticular. *Setae* absent. *Hyphopodia* absent. *Conidiophores* macronematous, mononematous, simple or occasionally once branched, often olivaceous brown, septate, usually fasciculate, bursting through the cuticle of the host plant. *Conidiogenous cells* integrated, terminal, polyblastic, sympodial, cicatrized; old conidial scars usually thickened, conspicuous and prominent, sometimes situated at the end of short lateral projections, numerous and often crowded, giving the conidiophore a nodular appearance. *Conidia* solitary or occasionally in short chains, dry, variable in shape but often tending to be broadly fusiform, truncate at the base and pointed at the apex, 0-3- (often 0- or 1-) septate, pale to mid olive or olivaceous brown, frequently minutely verruculose. *Pazschkeella* is characterized by forming pycnidial cavities

in the stroma and conidiogenous cells that line the cavity form aseptate, hyaline conidia holoblastically.

Notes: There are 36 species recorded in Index Fungorum (http://www.indexfungorum.org access on 18/12/11). *Microcyclus ulei* (Henn.) von Arx is an economically important pathogen that shows as leaf blight on rubber trees in South America (Lieberei, 2007). The taxonomic characterization of this species was described by Chee and Holiday (1986). Le Guen (2004) described microsatellite markers of fungal isolates from Brazil and from French Guiana and evaluated with respect to their variability within the isolates. This approach provides a step to group the highly variable complex isolate groups on a genetic information basis.

Generic type: *Microcyclus angolensis* (Sacc.) Syd. & P. Syd.

Microcyclus angolensis (Sacc.) Syd. & P. Syd. , Annales Mycologici 2(2): 165 (1904) Plate 7A-H.

MycoBank: MB152201

Ascostromata 0.2-0.3 mm wide, *Ascomata* 66.5-116.1 µm high × 57.7-108.8 µm diam., pulvinate, irregularly shaped, developing from central basal hypostroma, superficial, multilocular, composed of pseudoparenchymatous cells; *textura angularis*, thick-walled, dark brown (Plate 7A-E). Ostiole papillate, periphyses (Plate 7F). *Asci* 44.9-69.9 × 13.3-18.6 µm ($\bar{x}$ = 54.5 × 15.8 µm, n = 25), thick-walled, 8-spored with 1-3 seriate partially overlapping, bitunicate, fissitunicate ,cylindrical to clavate ,with an ocular chamber 0.9-1.5 µm wide × 0.5-0.9 µm high, with a pedicel, 3.7-5.9 µm long (Plate 7G). *Ascospores* 14.2-18.4 × 4.4-6.1 µm ($\bar{x}$ = 16.4 × 5.1 µm, n = 25), 1-septate, obovoid, upper cell shorter and wider than lower, not or slightly constricted at the septum, smooth wall, granular, hyaline (Plate 7H).

Anamorph: *Fusicladium* Bonorden and *Pazschkeella* H. & P. Sydow. Plate 8A-E.

Conidiomata 21-42.5 µm high × 48.7-76.4 µm diam., pycnidial, solitary to gregarious , immersed to semi-immersed, becoming erumpent, thin dark brown to black wall surrounding with host tissue (Plate 8A-B). *Conidiophores* hyaline, septate, cylindrical, smooth. *Conidiogenous cells* 1.2-2.9 µm wide, holoblastic, integrated, hyaline, cylindrical, producing a single apical conidium (Plate 8C). *Paraphyses* 1.2-1.4 µm wide, non-septate, unbranched, not-anastomosed (Plate 8D). *Conidia* 12.6-15.1 × 2.3-3.3 µm ($\bar{x}$ = 14 × 2.8 µm, n = 25), hyaline, smooth, thin walled, aseptate, fusiform to cylindrical, sometimes irregular cylindrical, base obtuse, tapering toward apex attached with one appendage (6.6-8 µm long) (Plate 8D-E)**.**

Cultures: *Microcyclus ulei* grows best on potato sucrose agar (PSA) (Chee, 1978). and can produce abundant conidia on PSA containing 0.2% sucrose (Holiday, 1970).

Sequence data: Genetic polymorphism of *Microcyclus ulei* has also been carried out by Le Guen (2004) using microsatellite Mu13 sequence (AY228721.1) and Mu14 sequence (AY228720.1).

Material examined: Angola, Africa; on living leaves of *Millettiae thonningii*, Welwitsch (F8592, F8593, **holotype**).

Notes:

Morphology

Previously, the genus was described to *Dothidella*, but in 1962, Müller and von Arx transferred it to *Microcyclus*. Barr (1996) arranged *Microcyclus* in the new family Planistromellaceae and distinguished this genus from other genus in the family in having

widely erumpent, multiloculate ascomata, 1-septate and oblong ascospores. Many species in this genus are biotrophic in leaves and stem of various plants in tropical and subtropical region (Barr, 1996 and Cannon *et. al.*, 1995). The type species; *M. angolensis* (Sacc.) Syd. & P. Syd. was described the periphyses ostioles by Theissen and Sydow (1915) and Müller and Sanwal (1954). Cannon *et. al.* (1995) placed one *Dothidea* species as *M. porlieriae* (Rehm) P. K. Cannon, Carmarán and A. I. Romero and gave a key to South American species of *Microcyclus*.

Phylogenetic study: *Microcyclus ulei* (Henn.) von Arx is an economically important pathogen that shows as leaf blight on rubber trees in South America (Lieberei, 2007). The taxonomic characterization of this species was described by Chee and Holiday (1986). Le Guen (2004) described microsatellite markers of fungal isolates from Brazil and from French Guiana and evaluated with respect to their variability within the isolates. This approach provides a step to group the highly variable complex isolate groups on a genetic information basis.

Concluding remarks:

Phaeosphaeriaceae

Loratospora Kohlm. & Volkm.-Kohlm., Syst. Ascom. 12(1-2): 10 (1993)
MycoBank: MB26473 Fig. 9

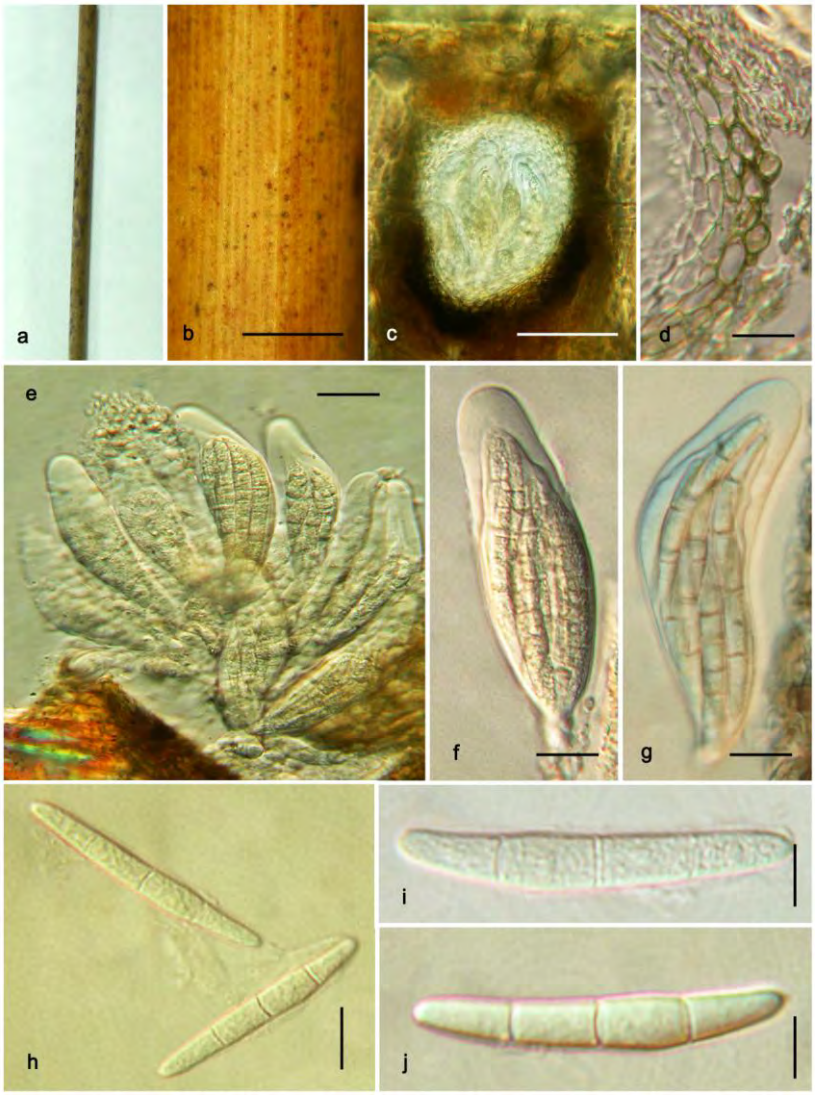


Fig 9. *Loratospora aestuarii Kohlm. & Volkm.-Kohlm* from North Caralina, Broad Creek, on dead culms of *Juncus roemerianus*. **a-b.** Ascomata forming in host tissue. **c.** Vertical section through the ascoma with asci. **d.** Close-up of the peridium at the ascoma side. Note that the wall is not divided into distinct layers. **e-g.** Asci with 8-spored. **h-j.** Ascospores 3-septate or 4 - celled. Scale bars: b-c = 100 µm, d-g = 25 µm, h-j = 5 µm.

Ascomata black immersed in the culms, carbonaceous, ostiolate, neck with periphyses. *Asci* clavate, thick-walled, fissitunicate without an apical apparatus, with an ocular chamber, J-. *Ascospores* hyaline, 3-septate, surrounded by a thin mucilaginous sheath. No species have been known for asexual state.

Notes: Loratospora is a genus recording only one species: *L. aestuarii* (http://www.indexfungorum.org, access on 15/07/12). Molecular analysis on of the type species; *Loratospora aestuarii* strain JK 5535D was studied by Suetrong *et al.*, (2009) based

on multigenes SSU, LSU and RPB2. The results showed that *L. aestuarii* is grouped in *Phaeosphaeriaceae.*
Generic type: *Loratospora aestuarii* Kohlm. & Volkm.-Kohlm.

Loratospora aestuarii Kohlm. & Volkm.-Kohlm., Systema Ascomycetum. 12(1-2): 10 (1993) Plate 9a-j.
MycoBank: MB360815

Ascostromata immersed below in host tissue a slightly raised darkened (Plate 1a, b). Ascostromata in section 97.5-125 µm diam. × 117.5-175 µm high, uniloculate, subglobose, solitary, gregarious, periphysate ostioles (Plate 9c). *Peridium* 17.5-18.8 µm thick ($\bar{x}$ = 17 µm, n = 5), composed of 4-5 layers of peridium, brown thick-wall, cuboid or angular cells (Plate 1d). *Asci* 75-137.5 µm × 20-32.5 µm ($\bar{x}$ = 95.5 × 26.3 µm, n = 20), bitunicate, fissitunicate, clavate to ovoid, apically rounded, apex 10-22.5 µm thick, with an ocular chamber 0.5-1 µm wide, short knob-like pedicel, 8-spored with 3-4 seriate overlapping (Plate 9f-g). *Ascospores* hyaline, narrowly obovoid, fusoid or clavate, 42.5-55-(-57.5) µm × 5-5.7 µm ($\bar{x}$ = 53.5 × 6.8 µm, n = 20), 3-septate, 4-celled, surrounded by a thin mucilaginous sheath, slightly constricted septum, smooth-walled (Plate 9h-j).

Anamorph: Unknown.

Cultures: JK 5535D (**unverified**, SSU - GU296168), (**unverified**, LSU - GU301838), (**unverified**, RBP2 - GU371760)

Sequence data: Genbank GU296168, Genbank GU301838, Genbank GU371760.

Material examined: North Caralina, Broad Creek, on dead culms of *Juncus roemerianus*, 31 Dec 1993, hand section and squash (Type species) and J.K. 5505; *Loratospora aestuarii*, gen. *et* sp. Nov, *Juncus roemerianus*, Broad Creek, NC, 6 April 1993, mature ascoma (neck) / Dusect (**Holotype**), (IMI 357990, **Isotype**).

Notes:
Morphology
Loratospora is a genus recording only one species: *L. aestuarii* (http://www.indexfungorum.org, access on 15/07/12). This species is an obligately and facultatively marine fungus and is characterized by unilocular ascostromata that are immersed in host tissue, ascospores; 3-septate, elongate (Kohlm. and Volkm.-Kohlm, 1993 and Jone *et al*., 2009). Barr (1996) placed *L. aestuarii* in *Planistromellaceae* by locules open via schizogenously form and periphysate ostioles. **Phylogenetic study:** Molecular analysis on of the type species; *Loratospora aestuarii* strain JK 5535D was studied by Suetrong *et al.*, (2009) based on multigenes SSU, LSU and RPB2. The results showed that *L. aestuarii* is grouped in *Phaeosphaeriaceae.*

Concluding remarks:

Acknowledgments
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Part III
Screening antimicrobial activity of secondary metabolites from saprobic fungi

1. Objective

To screen saprobic fungi for the production of bioactive compounds.

2. Methodology

2.1 Preliminary screening of antimicrobial activity

2.1.1 Fungal metabolites extraction

Ten isolates of total fifty-five fungi were selected for preliminary screening. Fungal isolates were selected from the genus which have been reported on producing fungal biological control agents and/or were marked as probably new species. Solid phase extraction method was used for fungal metabolites production (Chamyuang, S. 2010). Inoculum was prepared by cutting agar of pure culture plate into discs, 4 mm. diameter, from the margin of a colony. The inoculum disc was inverted onto the centre of an 85 mm petri dish containing 20 ml of sabouraud dextrose agar. Inoculated plates were incubated for 30 days at 28 °C. For each isolate, five replicate experiments were carried out. Fungal cultures with agar from each isolate were macerated with 30 ml ethyl acetate (EtOAc) and blended with a sterile stainless blender. This extraction process was carried out three times. The agar slurry was left sitting overnight for the first EtOAc extraction, and eight hours each for the second and third extractions. All three EtOAc extracts were combined and transferred to a pre-weighed vial and air dried to yield a crude extract. Crude extracts were prepared to a concentration of 1 g/ml in methanol and stored in 4 °C in airtight bottles.

2.1.2 Test microorganisms

Five pathogenic bacteria and four pathogenic fungi were used as test microorganisms. *Bacillus cereus* (TISTR no.687), *Escherichia coli* (TISTR no.780), *Pseudomonas aeruginosa* (ATCC no.27853), *Salmonella typhimurium* (TISTR no.292), *Staphylococcus aureus* and *Candida albicans* were obtained from Biology and Biotechnology Laboratory of the Scientific and Technological Instruments Center, Mae Fah Luang University, Chiang Rai. *Colletotrichum fructicola*, *Fusarium* sp. (MFU11-0219) and *Alternaria* sp. (MFU11-0123) were obtained from Mae Fah Luang University Culture Collection (MFUCC), Mae Fah Luang University, Chiang Rai.

2.1.3 Antimicrobial activity assay

The antimicrobial screening of the extracts was carried out using a modified paper disk method (Gu, 2009 and Wang, 2008). Bacteria were grown in nutrient broth at 37 °C for 18 hr and yeast was grown in potato dextrose broth at 30 °C for 18 hr. After that, they were adjusted to approximately 10^8 colony-forming unit per milliliter (CFU/ml). Agar plates were swabbed uniformly by test bacteria on nutrient agar (NA) and yeast on potato dextrose agar (PDA). Whereas, other fungi were grown in PDA at 28 °C for 2-3 days until radius of colony are reached to approximately 20 mm.. Sterile paper disks (6 mm diameter) each containing 20 µl of sample solution (0.1 g/ml) were dried thoroughly and placed on the surface of medium. The test plates were then incubated at 37 °C for 24 hr for bacteria and 28 °C for 1-4 days for fungi. Methanol was used as negative control. Streptomycin sulphate (10 µg/ml) and

bacitracin (10 μg/ml) were used as a positive control. For each test, five replicates were performed. The diameter (mm) of the growth inhibition are examined and measured.

2.2 Isolation of fungal metabolites and antimicrobial activity (This part was done in Institute of chemical technology, Prague, Czech Republic)

2.2.1 Fungal metabolites extraction

Fungi were inoculated on plates of sabouraud dextrose agar and incubated for 7-30 days at 28 °C. For each isolate, five replicate experiments were carried out. After incubation, mycelium was maximally scraped out from the surface of agar. Both mycelium and agar were mixed together using grinder. The extraction process was carried out three times. Ethanol was added into the mixtures of agar and mycelium with equal volume (1:1) at the first and the second extraction. At the third extraction, ethyl acetate was used with equal volume (1:1) to the mixture. The extraction was continued by shaking at 150 rpm for 24 hours. After the extraction had been finished, all extracts were combined and evaporated by rotary evaporator. To yield a crude extract, all extracts were air dried and collected into vial (see Diagram 1).

2.2.2 Isolation of crude extracts

Crude extracts were separated into four fractions by their hydrophilic and hydrophobic properties. The solvents of petroleum ether, acetone, ethanol and water were used to dissolve crude extracts respectively. To clean each fraction, insoluble solvent was used in a volume as much as possible by the centrifugation. Then all fractions were air dried, weighed and kept in small vial (see Diagram 2).

Diagram 1 : Fungal metabolites extraction

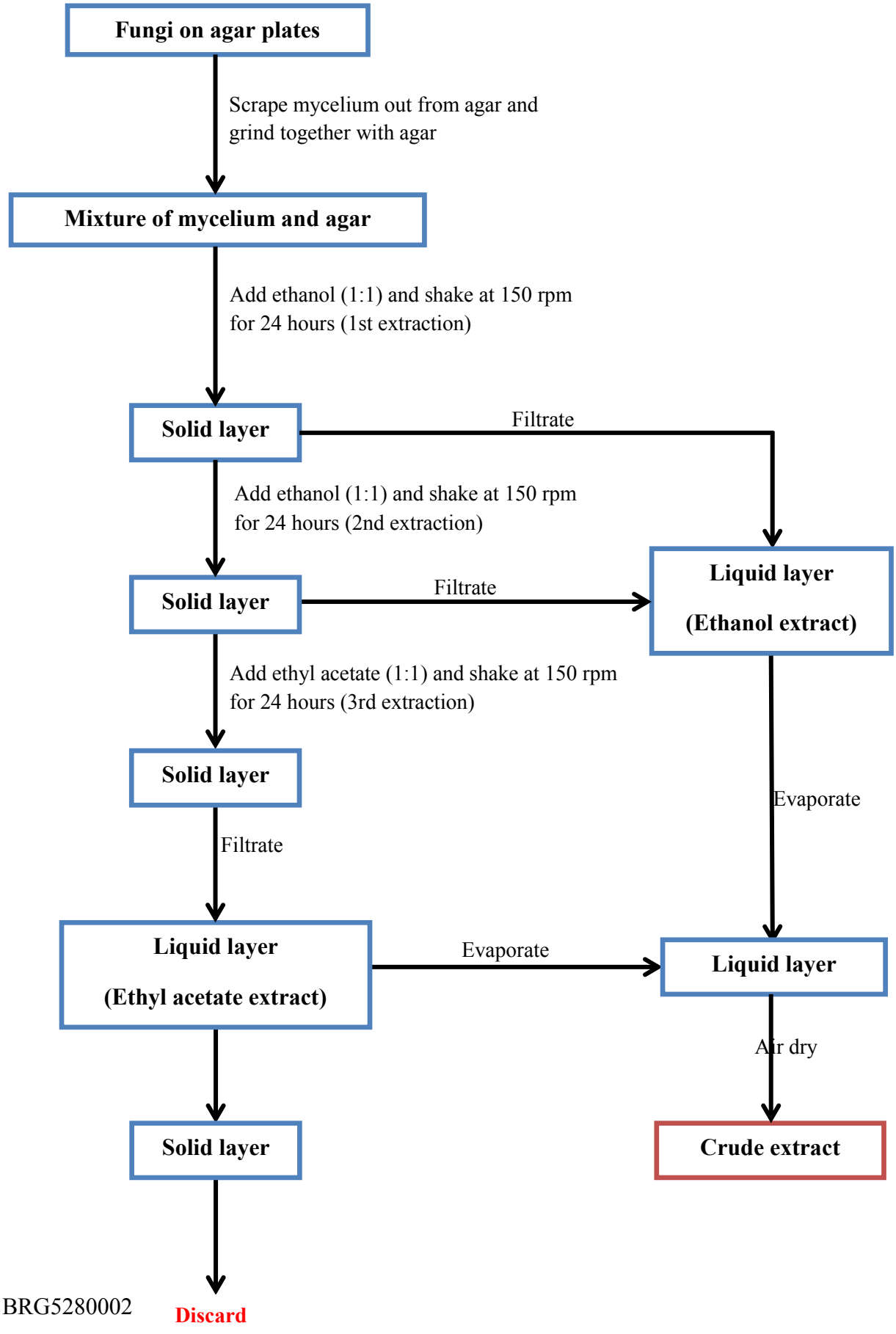

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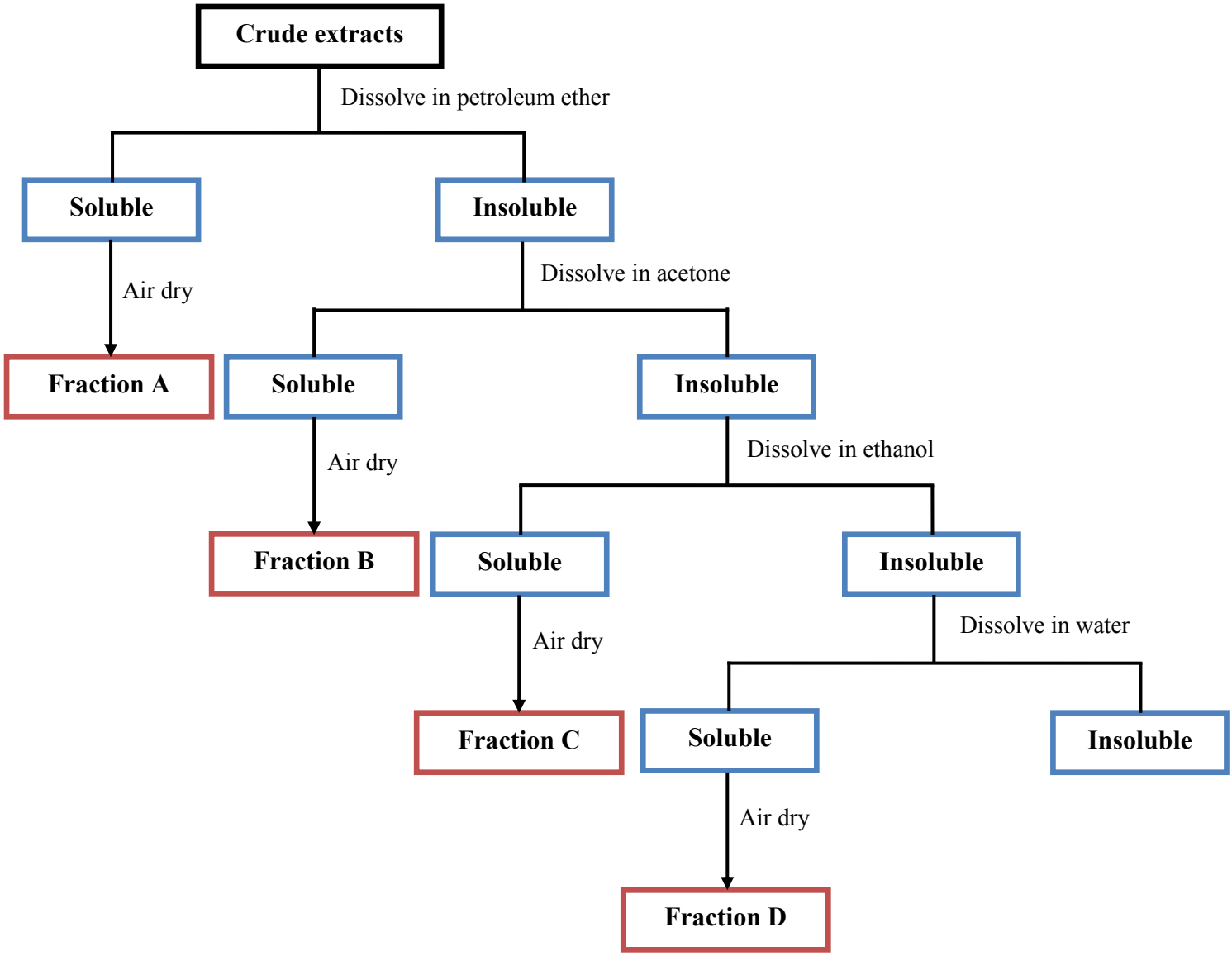
Diagram 2 : Isolation of crude extracts



2.2.3 Analytical spectroscopy of fungal metabolites.

Samples were prepared into a thin tablet by mixing crude extracts with KBr. Tablet samples were measured on FTIR (Nicolet 6700, spectral range 4000-400 cm^{-1} ,resolution 2 cm^{-1} and number of scan 64) using the Omnic 7.3 software.

2.2.4 Antimicrobial activities test.

The antimicrobial activity of fungal metabolites was carried out by disc diffusion methods against 6 species of various microorganisms (Staphylococcus aureus, Bacillus cereus, Enterococcus faecalis, Pseudomonas aeruginosa, Enterobacter cloacae and Saccharomyces cerevisiae).

3.Results

3.1 Preliminary screening of antimicrobial activity

The antimicrobial activity of ten fungal metabolites was assayed in vitro by modified disc diffusion methods against nine pathogenic microorganisms. The result of microbial growth inhibition testing by fungal extracts was shown in Table 1. For antibacterial test, it was found that four fungal species inhibited the growth of at least one test bacteria. Among them, Ophioceras sp.1 (JM018) showed the best antibacterial activity against S.aureus and followed by B.cereus (Figure 1A). Fusicoccum sp.2 (JM024) and Chaetomium sp.1 (CI18) also exhibited antimicrobial activity against Ps.aeruginosa. Another fungi, Verticillium sp.1 (JM013) inhibited the growth of B.cereus. In antifungal test, only two fungal species Chaetomium sp.1 (CI18) and Verticillium sp.1 (JM013) showed the inhibition of growth to C.fructicola ,respectively (Figure 1B).

Table 1. Antimicrobial activitiy of fungal extracts against some pathogenic microorganisms.

Fungal extracts	Zone of inhibition (mm)[1]								
	B.cereus	*E.coli*	*Ps.aeruginosa*	*S.typhimurium*	*S.aureus*	*C.albicans*	*C.fructicola*	*Fusarium* sp.	*Alternaria* sp.
Stachybotrys sp.1 (JM009)	-	-	-	-	-	-	-	-	-
Verticillium sp.1 (JM013)	8	-	-	-	-	-	8	-	-
Beltrania sp.1 (JM014)	-	-	-	-	-	-	-	-	-
Ophioceras sp.1 (JM018)	8	-	-	-	15	-	-	-	-
Fusicoccum sp.1 (JM019)	-	-	-	-	-	10	-	-	-
Fusicoccum sp.2 (JM024)	-	-	9	-	-	11	-	-	-
Ascomycetes sp.1 (JM025)	-	-	-	-	-	-	-	-	-
Sporodochium sp.1 (CI03)	-	-	-	-	-	-	-	-	-
Sporodochium sp.2 (CI09)	-	-	-	-	-	-	-	-	-
Chaetomium sp.1 (CI18)	-	-	8	-	-	-	11	-	-
Methanol[2]	-	-	-	-	-	-	-	-	-
Streptomycin sulphate[2]	20	20	15	20	14	-	-	-	-
Bacitracin[2]	ND	ND	ND	ND	ND	ND	13	ND	ND

- No activity, ND = Not done

1: Inhibition zones are the mean including disc (6 mm) diameter

2: Controls (Streptomycin sulphate 10 µg/ml, Bacitracin 10 µg/ml)

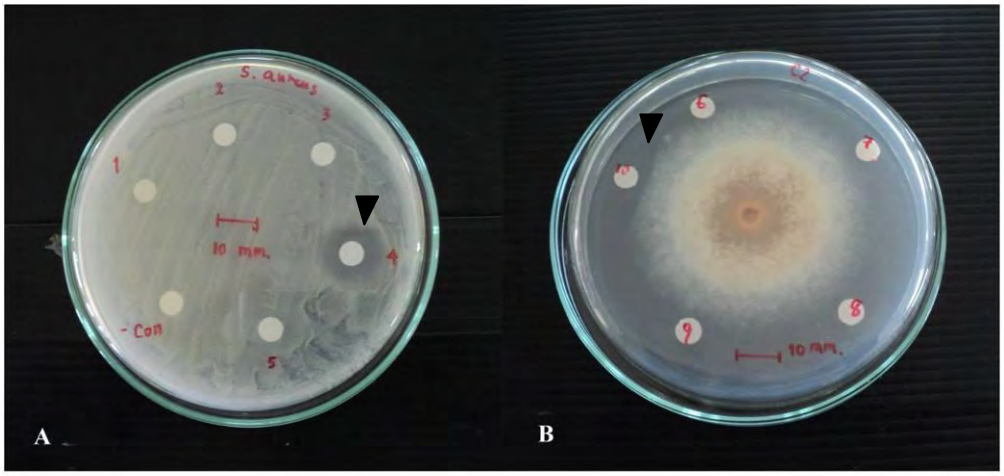


Figure 1. Disc diffusion assay showing antimicrobial activity of fungal extracts A. Inhibition zone of *Ophioceras* sp.1 (JM018) on *S.aureus* plate (arrow point). B. Inhibition zone of *Chaetomium* sp.1 (CI18) with *C.fructicola* (arrow point).

3.2 Isolation of fungal metabolites and antimicrobial activity

3.2.1 Fungal metabolites extraction and isolation of crude extracts

Five fungi from Institute of chemical technology, Prague, Czech Republic and nine fungi from MFUCC, Mae Fah Luang University, Thailand were cultivated. All fungi were extracted and isolated for metabolites. The results of weigh in each mycelium and fractions are shown in table 2.

3.2.2 Analytical spectroscopy of fungal metabolites.

All fractions of crude extracts were analyzed by FTIR. Obtained spectra are shown in Figures 2 – 5. The spectra of fractions A contain characteristic bands of lipids (fats, fatty acids). According to FTIR, two obtained fractions B contain phenolics. Fractions C and D are mixture of various compounds (small molecules) including sugars, glycosides, organic acids, phenolics etc. The presence of proteins, or rather shorter peptides, is possible in some cases (no.1 C). The fractions with some exceptions (mainly A) were highly colored that is an evidence of the presence of various pigments.

3.2.3 Antimicrobial activities test.

The results of microbial growth inhibition testing by fungal extracts were shown in Table 3 and 4. From the results, it can conclude that Trichoderma sp. (no.1), Aspergillus oryzae (no.2), Fusicoccum sp.1 (no.5), Chaetomium sp.1 (no.7) and Verticillium sp.1 (no.9) were effective inhibitors against some test microorganism (figure 6).

Table 2. List of fungi show weigh of each mycelium and crude extracts

No	CODE	Fungi	Weigh (g)					Remark
			A:Petroleum ether	B:Acetone	C:Ethanol	D:Water	Mycelium	
1	DBM 4197	*Trichoderma sp.*	0.090	0.015	0.200	-	0.780	From Czech
2	DBM 4336	*Aspergillus oryzae*	0.050	0.015	0.200	-	1.150	From Czech
3	4208	*Botrytis sp.*	0.020	0.010	0.040	-	0.490	From Czech
4	4297	*Stachybotrys sp.*	0.030	0.002	0.130	-	0.400	From Czech
5	JM024	*Fusicoccum sp.1*	0.010	0.009	0.130	-	0.250	From Thailand
6	JM019	*Fusicoccum sp.2*	0.030	0.005	-	-	0.100	From Thailand
7	CI18	*Chaetomium sp.1*	0.120	-	0.850	0.320	-	From Thailand
8	JM025	*Ascomycete sp. 1*	0.210	-	0.070	0.333	-	From Thailand
9	JM013	*Verticillium sp.1*	0.210	-	0.460	0.320	-	From Thailand
10	JM014	*Beltrania sp.1*	0.290	-	0.180	0.280	-	From Thailand
11	JM009	*Stachybotrys sp.1*	0.070	-	0.146	0.075	-	From Thailand
12	CI03	*Sporodochium sp.1*	0.100	-	0.237	0.076	-	From Thailand
13	CI09	*Sporodochium sp.2*	0.100	-	0.285	0.155	-	From Thailand
14	DS77	*Cladosporium sp.*	0.053	-	0.642	0.005	-	From Czech
15	JM019	*Fusicoccum sp.2*	0.263	-	1.774	0.321	-	Repeating
16	CI18	*Chaetomium sp.1*	0.057	-	7.157	5.183	3.045	Large Scale Extraction

- mean no samples

Table 3. Antimicrobial activity of fractions A against some pathogenic microorganisms

No	Fungi	Zone of inhibition (mm)					
		Staphylococcus aureus	*Bacillus cereus*	*Enterococcus faecalis*	*Pseudomonas aeruginosa*	*Enterobacter cloacae*	*Saccharomyces cerevisiae*
1	*Trichoderma* sp.	0	6	0	0	0	0
2	*Aspergillus oryzae*	13	20	3	0	0	0
3	*Botrytis* sp.	0	0	0	0	0	0
4	*Stachybotrys* sp.	6	0	3	0	0	0
5	*Fusicoccum* sp.1	0	0	0	0	0	0
6	*Fusicoccum* sp.2	0	0	0	0	0	0
7	*Chaetomium* sp.1	0	7	0	0	0	0
8	*Ascomycete* sp. 1	0	0	0	0	0	0
9	*Verticillium* sp.1	0	5	0	0	0	0
10	*Beltrania* sp.1	0	0	0	0	0	0
	Water (control)	0	0	0	0	0	0

Table 4. Antimicrobial activity of fractions C against some pathogenic microorganisms

No	Fungi	Zone of inhibition (mm)					
		Staphylococcus aureus	*Bacillus cereus*	*Enterococcus faecalis*	*Pseudomonas aeruginosa*	*Enterobacter cloacae*	*Saccharomyces cerevisiae*
5	***Fusicoccum sp.1***	0	**7**	0	0	**5**	0
7	***Chaetomium sp.1***	0	**6**	0	0	0	0
8	*Ascomycete sp. 1*	0	0	0	0	0	0
9	***Verticillium sp.1***	**4**	**10**	**5**	0	**6**	**4**
	Ethanol (control)	0	2	0	0	4	0

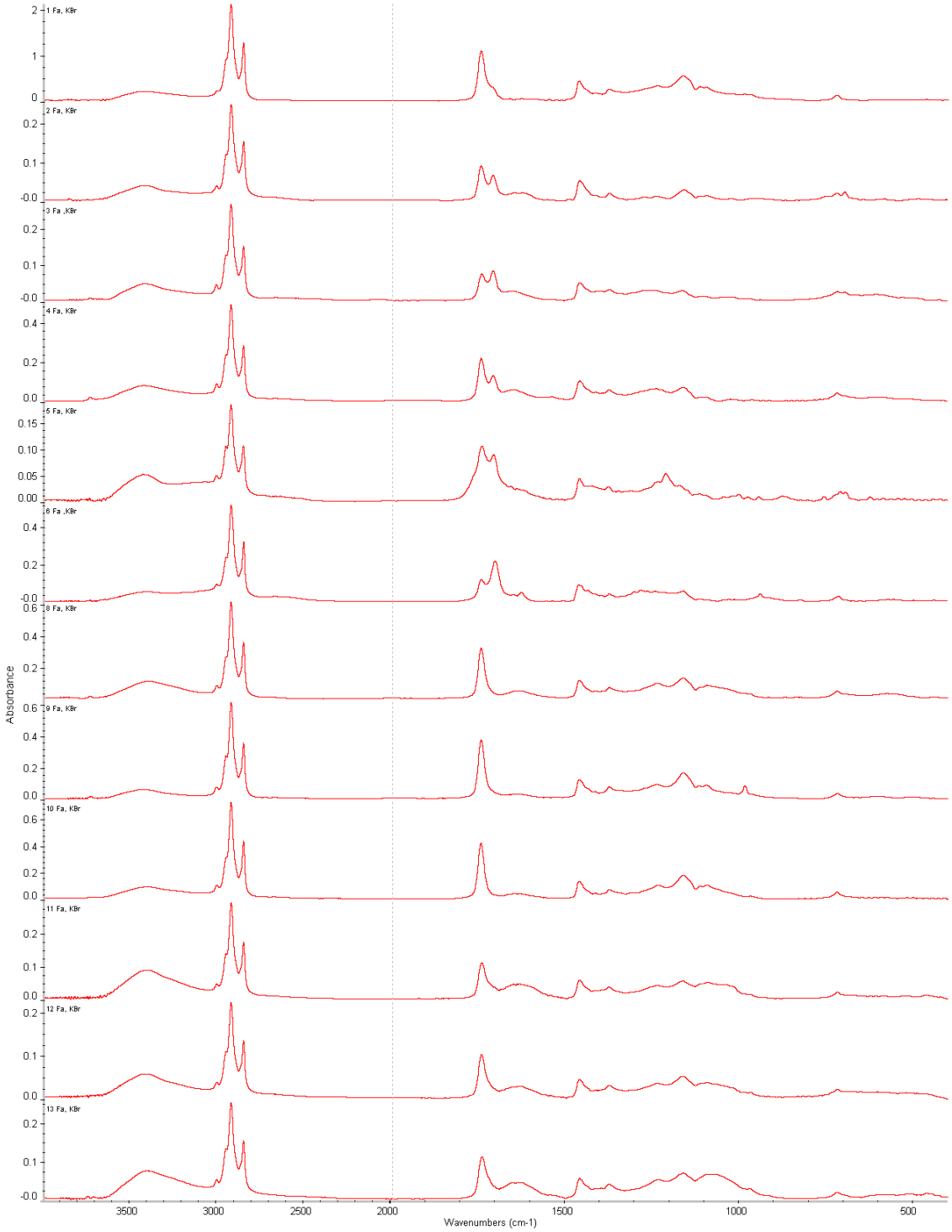


Figure 2. FTIR spectra of fractions A: Petroleum ether soluble

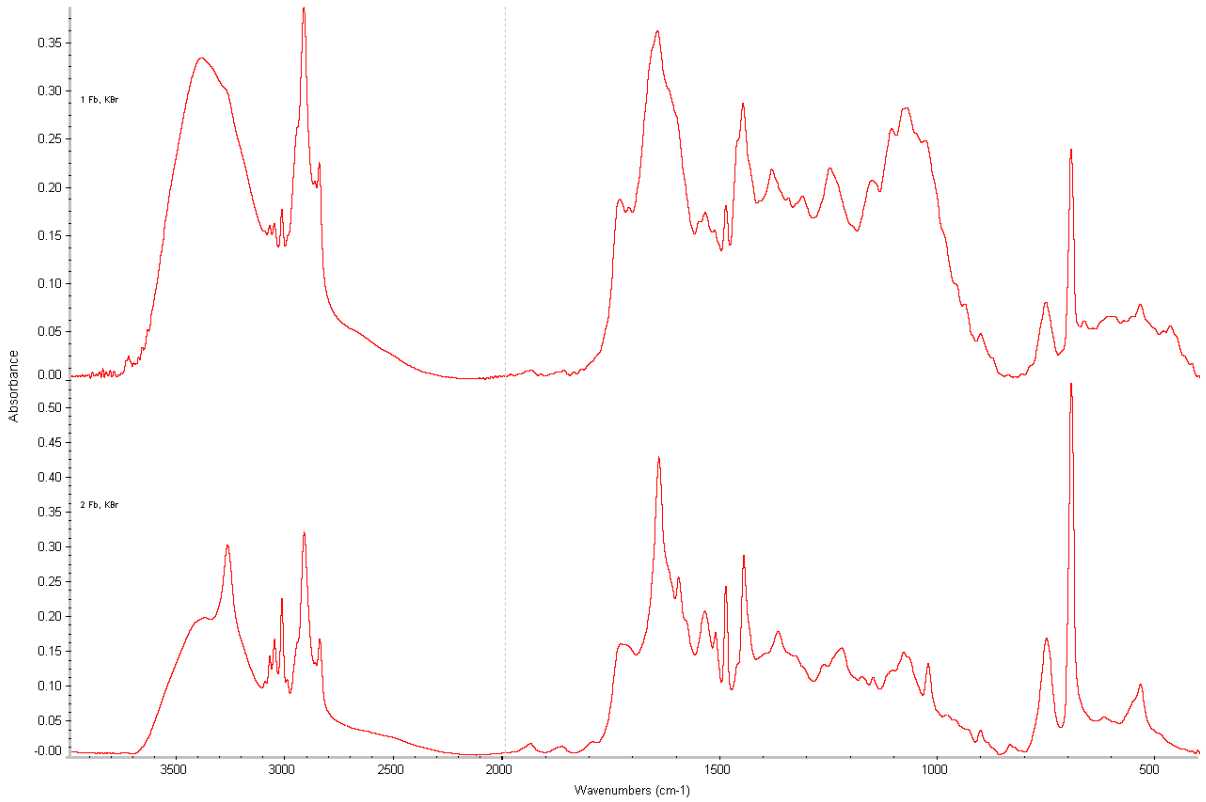


Figure 3. FTIR spectra of fractions B: Acetone soluble

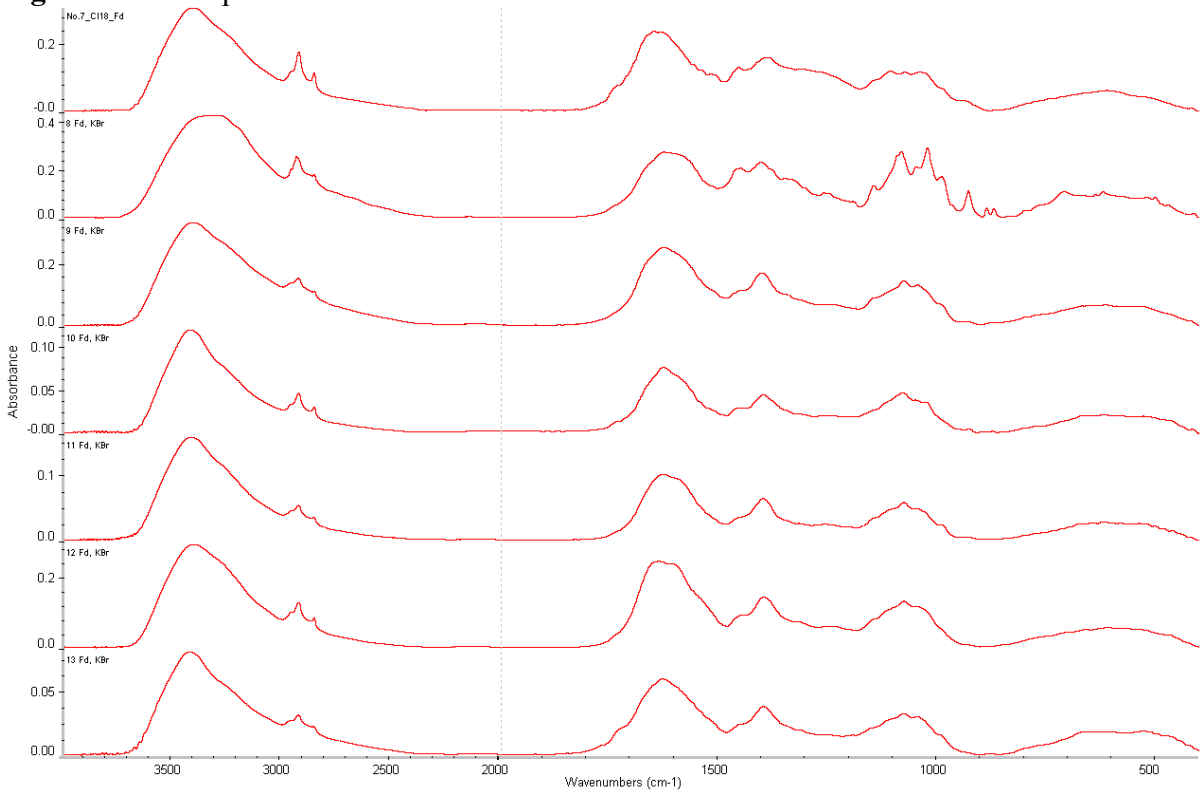


Figure 4. FTIR spectra of fractions D: Water soluble

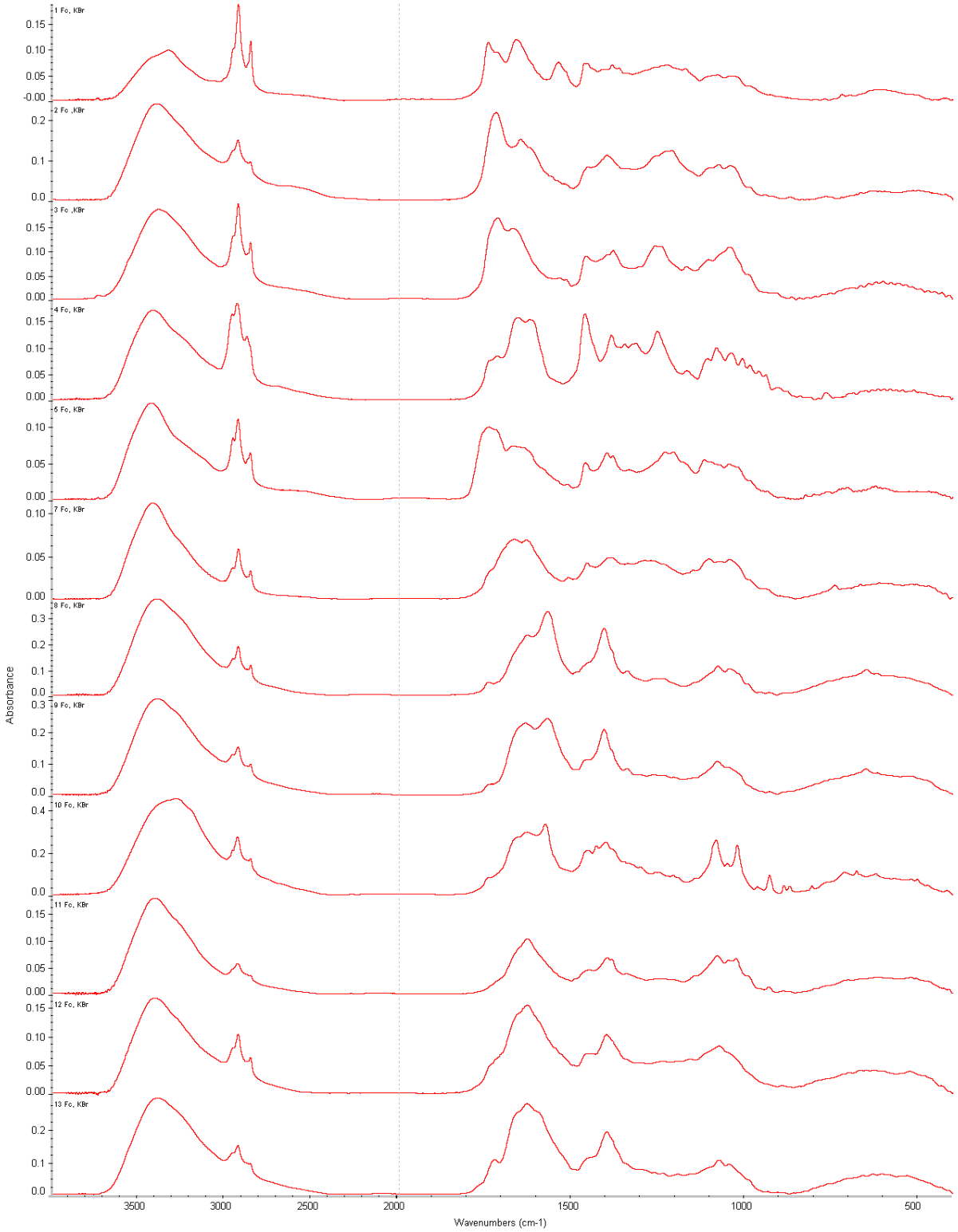


Figure 5. FTIR spectra of fractions C: Ethanol soluble

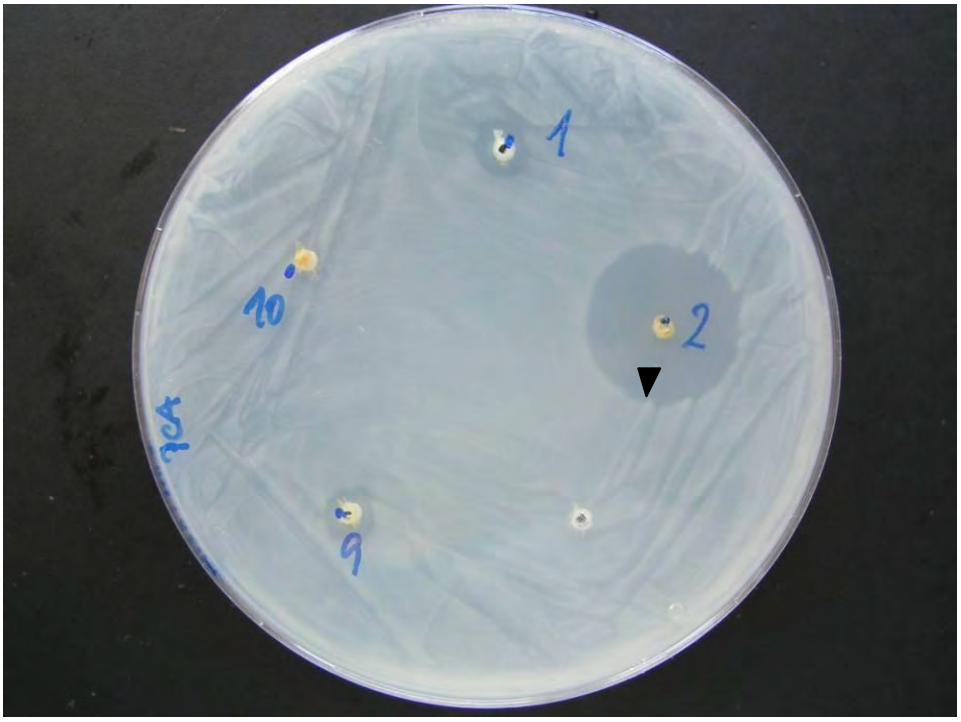


Figure 6. Antimicrobial activity of fungal extracts in Fraction A. (no.1 = *Trichoderma* sp., no.2 = *Aspergillus oryzae*, no.9 = *Verticillium* sp.1 and no.10 = *Beltrania* sp.1)

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Appendix B

The Final Report

Part 2

Biodiversity of saprobic fungi on woody litter in Thailand

Reported by
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Part A
Biodiversity of saprobic fungi on woody litter in Thailand

1. Research Objectives

1. To study biodiversity of saprobic fungi found on woody litter in forests of Thailand
    1.1 Sumary of cultures collection.
    1.2 Interesting fungi
2. To study on herbarium specimens in *Patellariaceae*
3. To collecting and study fresh collection of *Patellariaceae* in Thailand.

2. Research methodology

2.1 Collected samples: Collecting sample will be selected from Northern Thailand, Central of Thailand and Southern Thailand. The collection work will be carried out during the warm wet season over two years (2010-2011). Decaying wood will be targeted. During one collection trip, 30 dead wood samples will be collected randomly and return to laboratory. Each sample will put in separately plastic boxes (Kodsueb et al., 2008). Fungi growing on wood will be identified, isolated by single spore isolation and herbarium specimens will be prepared. Both of living culture and herbarium specimens will be deposited in MFLUCC herbarium.

2.2 Morphological study: Cultures collection of ascomycetes and their anamorphs will be described by traditional morphology (e.g. section of ascomata, texture of peridium, hamathecium, asci and ascospore). Many books and herbarium are loaned to study and use to identify and compare information. (e.g. Carmichael et al., 1980; Sutton, 1980; Hyde et al., 2001; Cai et al., 2006)

2.3 Molecular study

2.3.1 DNA extraction and amplification: Fresh cultures will be used for DNA extraction using CTAB method (Jeewon et al., 2003). DNA samples will be checked for purity and integrity by agarose gel electrophoresis. PCR amplification will be performed for ITS, LSU, SSU genes under the conditions of Guo et al. (2000). PCR products will be sequenced by SinoGenomax company.

2.3.2 Sequence alignment and phylogenetic analyses: Sequences of all strains and the type species sequences will be aligned by Clustal X and optimized manually in BioEdit (Hall, 1999) and PAUP*4.0b10 will be used to obtain the phylogenetic trees (Zhao et al., 2007).

3. Results

3.1 Isolated cultures and interesting fungi

The cultures collection from woody litter specimens in this study have been shown in the table1. Some of interesting fungi were shown the plate

Table1. List of cultures collection

No.	Code	Name	MFLUCC	Habitat	Host	Collection site	Remark
1	KA001	*Thyridaria* sp. nov.	10-0304	Dead wood	Dead wood	DT*	May be new species
2	KA002	*Eutypa* sp1.	-	Dead wood	Dead wood	DT*	-
3	KA003	*Diatrype* sp. like	-	Dead wood	Dead wood	DT*	-
4	KA007	*Rhytidhysteron rufulum*	-	Dead wood	Dead wood	DT*	-
5	KA011	*Dendryphiopsis* sp.	0762	Dead wood	Dead wood	DT*	-
6	KA016	*Endofragmia glanduliformis* like	-	Dead wood	Dead wood	DT*	-
7	KA023	*Monodictys* sp1.	0763	Dead wood	Dead wood	HM**	May be new species
8	KA024	*Dictyopolyshema pirozynskii* like	0764	Dead wood	Dead wood	HM**	-
9	KA026	*Eversia subopaca*	-	Dead wood	Dead wood	HM**	-
10.	KA028	*Hyphomycete* sp.1	-	Dead wood	Dead wood	HM**	-
11	KA029	*Ascomycete* sp.2	-	Dead wood	Dead wood	HM**	-
12	KA036	*Diplodia* sp.1	-	Dead wood	Dead wood	HM**	-
13	KA043	*Pleurothecium recuruatum*	-	Dead wood	Dead wood	HM**	-
14	KA044	*Brachysporiella pulchra*	-	Dead wood	Dead wood	HM**	-
15	KA046	*Monodictys* sp.2	-	Dead wood	Dead wood	HM**	-
16	KA051	*Phaeoisaria clematidis*	-	Dead wood	Dead wood	HM**	-

Table1. List of cultures collection

No.	Code	Name	MFLUCC	Habitat	Host	Collection site	Remark
17	KA055	*Lasiosphaeria* sp.	-	Dead wood	Dead wood	HM**	-
18	KA056	*Monodictys intens*	-	Dead wood	Dead wood	HM**	-
19	KA060	*Diaporthopsis* sp.	-	Dead wood	Dead wood	HM**	-
20	KA061	*Helicosporus* sp.	-	Dead wood	Dead wood	HM**	-
21	KA063	*Veronaea* sp.	-	Dead wood	Dead wood	HM**	-
22	KA064	*Savoryella* sp.	-	Dead wood	Dead wood	HM**	New record
23	KA065	*Thyridium* sp.	-	Dead wood	Dead wood	HM**	-
24	KA066	*Ascomycete* sp.2	-	Dead wood	Dead wood	HM**	-
25	KA067	*Ascomycete* sp.3	-	Dead wood	Dead wood	HM**	-
26	KA068	*Cancellidium* sp.	11-0430	Dead wood	Dead wood	HM**	-
27	KA069	*Stilbella* sp.	-	Dead wood	Dead wood	HM**	-
28	KA070	*Acrogenospora sphaerocephala*	-	Dead wood	Dead wood	HM**	-
29	KA071	*Pestalozziella* sp.	-	Dead wood	Dead wood	HM**	-
30	KA072	*Leptoshaeria* sp.	-	Dead wood	Dead wood	HM**	-
31	KA073	*Linocarpon* sp.	-	Dead wood	Dead wood	HM**	-
32	KA074	*Ascomycete* sp.4	-	Dead wood	Dead wood	HM**	-
33	KA075	*Rhytidhysteron* sp.	-	Dead wood	Dead wood	HM**	-

Table1. List of cultures collection

No.	Code	Name	MFLUCC	Habitat	Host	Collection site	Remark
34	KA076	*Excipularia marsapurensis*	-	Dead wood	Dead wood	HM**	-
35	KA077	*Xylohypho palmicola*	-	Dead wood	Dead wood	HM**	-
36	KA078	*Chaetosphaeris phaeosroma*	-	Dead wood	Dead wood	HM**	-
37	KA079	*Camarosporium* sp.	-	Dead wood	Dead wood	HM**	-
38	KA081	*Myxocyclus polycistis*	-	Dead wood	Dead wood	HM**	-
39	KA082	*Brachysporium* sp.	-	Dead wood	Dead wood	HM**	-
40	KA083	*Gibellula pulchra*	-	Dead wood	Dead wood	HM**	-
41	KA084	*Ascomycete* sp.6	-	Dead wood	Dead wood	HM**	-
42	KA085	*Monodictys sp.*	-	Dead wood	Dead wood	HM**	-
43	KA086	*Ascomycetes sp.*	-	Dead wood	Dead wood	HM**	-
44	KA087	*Coelomycete sp.*	-	Dead wood	Dead wood	SK	-
45	KA088	*Sporidesmium sp.*	-	Dead wood	Dead wood	SK	-
46	KA089	*Leptosphearia sp*	-	Dead wood	Dead wood	DT	-
47	KA090	*Montagnula sp.*	-	Dead wood	Dead wood	DT	Interesting fungi
48	KA091	*Monoharachariella sp.*	-	Dead wood	Dead wood	HK	-
49	KA092	*Endophragmiella sp.*	-	Dead wood	Dead wood	ST	-
50	KA093	*Rhitidhysteron sp.*	-	Dead wood	Dead wood	HK	-
51	KA094	*Stilbella sp.*	-	Dead wood	Dead wood	HS	-

Table1. List of cultures collection

No.	Code	Name	MFLUCC	Habitat	Host	Collection site	Remark
52	KA095	*Leptospearia* sp.	11-0577	Dead wood	Dead wood	CR	-
53	KA096	*Ceolomycete* sp.	-	Dead wood	Dead wood	CR	-
54	KA097	*Antrostromella* sp.	12-0008	Dead wood	Dead wood	CM	Interesting fungi
55	KA098	*Leptosphaeria* sp.	-	Dead wood	Dead wood	CM	-
56	KA099	*Ascomycete* sp.	-	Dead wood	Dead wood	CR	-
57	KA100	*Ascomycete* sp.	12-0011	Dead wood	Dead wood	CR	-
58	KA101	*Ascomycete* sp.	-	Dead wood	Dead wood	CR	-
59	KA102	*Diaporthe* sp.		Dead wood	Dead wood	PY	-
60	KA103	*Pseudopetrakia* sp.(looklike)	12-0009	Dead wood	Dead wood	PY	Interesting fungi
61	KA104	*Sporidesmium* sp.	-	Dead wood	Dead wood	PY	-
62	KA105	*Hysterobrevium* sp. like	12-0010	Dead wood	Dead wood	PY	-
63	KA106	*Diaporthe* sp.	12-0045	Dead wood	Dead wood	PY	-
64	KA107	*Astroshearilla* sp.	12-0046	Dead wood	Dead wood	PY	-
65	KA108	*Ascomycete* sp.	-	Dead wood	Dead wood	PY	-
66	KA109	*Ellisembia adscendens*	12-0047-48	Dead wood	Dead wood	PY	-
67	KA110	*Rhystidhysteron rufulum* like	12-0012	Dead wood	Dead wood	PY	-

Table1. List of cultures collection

No.	Code	Name	MFLUCC	Habitat	Host	Collection site	Remark
68	KA111	*Helicosporus* sp.	12-0049	Dead wood	Dead wood	PY	-
69	KA112	*Rhystidhysteron rufulum* like	12-0013	Dead wood	Dead wood	PY	-
70	KA119	*Hysterographium* sp. like	-	Dead wood	Dead wood	CR	-
71	KA120	*Karschia lignyota like*	-	Dead wood	Dead wood	PY	-
72	KA121	*Rhystidhysteron rufulum* like	-	Dead wood	Dead wood	CR	-

Draft paper

Monodictys appendiculata sp. nov. on decaying wood from northern Thailand

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ABSTRACT- A new species, *Monodictys appendiculata* S. Yacharoen, McKenzie & K.D. Hyde, was found on undetermined dead wood from Chiang Mai, Thailand. The type specimen and a culture derived from the type are deposited, respectively, in the herbarium of Mae Fah Luang University and Mae Fah Luang University culture collection (MFLUCC). *M. appendiculata* differs from similar *Monodictys* species by conidia shape, size, basal cell and presence of an appendage

KEY WORDS- *Monodictys, decaying wood, saprobic fungi, fungi with appendage.*

Introduction

During an investigation into the biodiversity of saprobic fungi on woody litter in Thailand, a new species of *Monodictys* was found on decaying wood collected from Chiang Mai, during the wet season. The genus *Monodictys* was described by Hughes (1958). Seifert et al. (2011) recognized 50 species of *Monodictys*; several additional species have been transferred to other genera (Indexfungorum 2011). The main characteristics of the genus are micro- or semi-macronematous, unbranched or irregularly branched conidiophores with integrated, monoblastic conidiogenous cells. The muriform conidia, which form solitary, are of various shapes (mainly subglobose to elongate, and smooth or verrucose; the basal cell is sometimes thin-walled and paler than the other cells (Ellis, 1971; Hughes, 1958). The new species was distinguished by morphological characters.

Mostly they are found on wood and leaf litter, soil, dung and lichens (*Lecanora, Lepraria*) (Hawksworth, 1979; Seifert et. al, 2011).

Materials and Methods

Sample collection, cultivation and morphology

Dead branches of various trees were collected from Doi Suthep-Pui, Chiang Mai, Thailand in June 2010. Samples were incubated in plastic boxes to which moistened sterilized tissue paper had been added to encourage fungal sporulation under high humidity. Morphological characteristics of fungi were examined under a light microscopy (Nikon ELIPSE 80i). The single spore isolation method outlined by Choi et al. (1999) was used to try to establish colonies of the fungus but, unfortunately, the conidia did not germinate. Species were determined using Ellis (1971; 1976). The type specimen was placed in the herbarium of Mae Fah Luang University (MFLU number).

Taxonomy

***Monodictys* S. Hughes**
(Melanommataceae)
Type species: *Monodictys putredinis* (Wallr.) S. Hughes, Can. J. Bot. 36: 785 (1958)
≡ *Melanconium putredinis* Wallr., Fl. crypt. Germ. (Norimbergae) 2: 181 (1833)

Monodictys is in Melanommataceae family The primary key was used to identify *Monodictys* (Rao & De Hoog, 1986)

***Monodictys appendiculata* S.** Yacharoen, McKenzie and K.D. Hyde, **sp. nov. (Fig. 1a-k)**
Mycobank:****

Stroma none. ***Setae*** and ***hyphopodia*** absent. ***Conidiophores*** micronematous or semi-macronematous, mononematous, unbranched, straight or flexuous, hyaline to brown, smooth, septate, cells sometimes swollen, 16.5-45 × 2.5-7 μm ($\bar{x}$=30 × 5.4; n = 20). ***Conidiogenous cells*** monoblastic, integrated, terminal, determinate, cylindrical, doliform or subspherical. ***Conidia*** solitary, dry, acrogenous, simple, ellipsoidal to oval, often irregular in outline due to constrictions at septate, brown to black, 27-65 × 18.5-46 μm ($\bar{x}$ = 50 × 36; n = 20), basal cell sometimes inflated, paler and thinner-walled than the other cells; with an apical appendage, one-septate, 6.5-17 × 4.5-8.5 μm ($\bar{x}$ = 11 × 6; n = 20).

Substrate: Dead woody litter.

Known distribution: Thailand.

Specimen examined: Thailand, Chiang Mai, Doi Suthep-Pui, Hui Kok Mah, on decaying wood, 18 June 2010, Supalak Yacharoen, MFLUCC0763 (holotype, ****MFLCC).

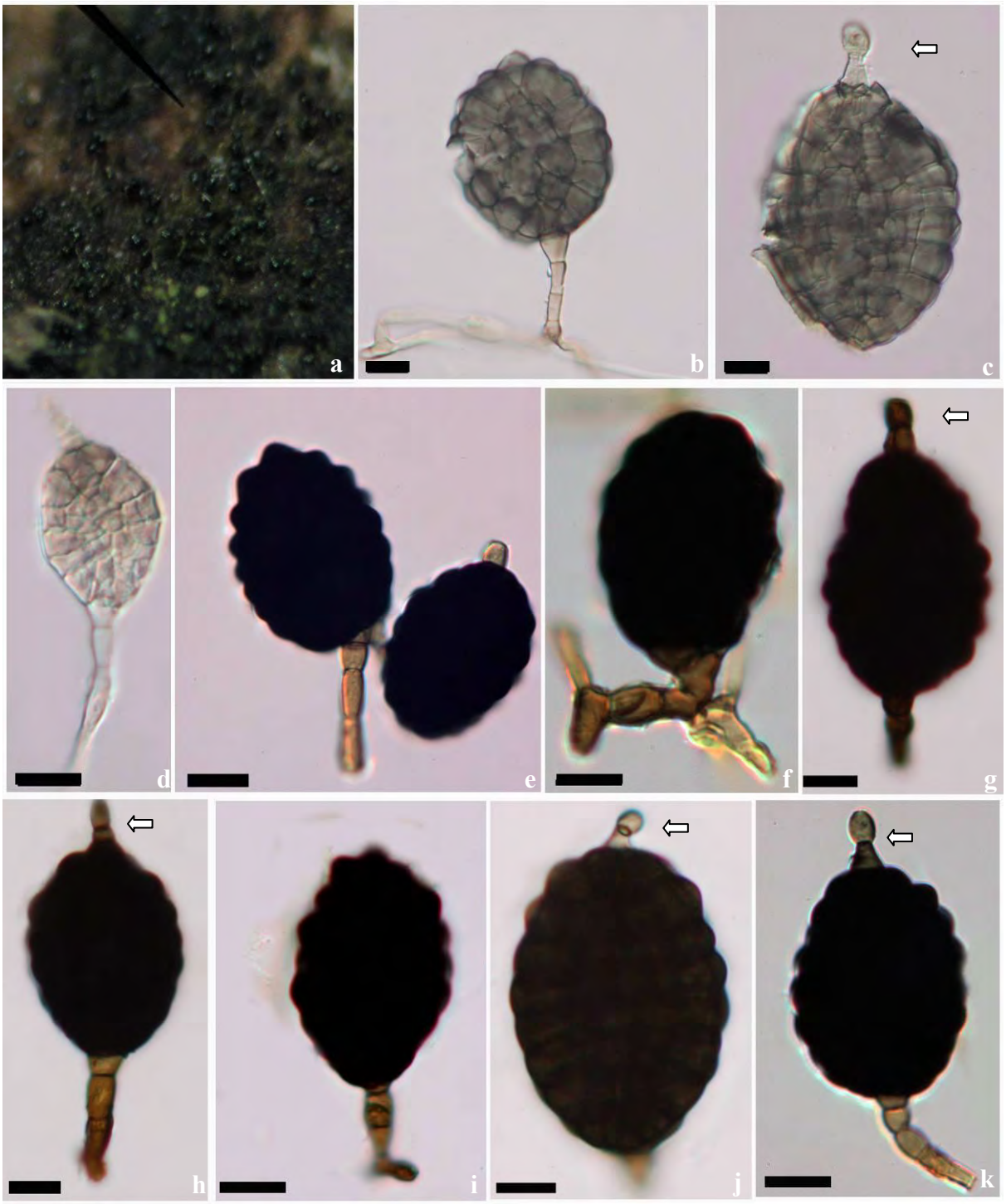

Fig 1 (a-k). *Monodictys appendiculata* (MFLUCC0763). **a.** Colony on host tissue; **b-d,** Young pale grey to grey conidia with visible septation**, e-k** Conidiophores, conidiogenous cells and conidia; apical appendage arrowed. Scale bars = 10 µm.

Table 1. A morphological comparison of *Monodictis appendiculata* and related species

Species	Conidiophores	Conidia			
		Shape	Size (μm)	Basal cell	Appendage
M. appendiculata	Micronematous, straight or flexous, cells sometimes swollen, septate, hyaline to brown	Ellipsoidal to oval, irregular in outline,constricted at septa brown to black	27-65 × 18.5-46	Inflated thinne walled and paler	One septate apical appendage
M. paradoxa	Micronematica, cells inflated	Oblong or ellipsoidal, dark olivaceous or blackish olive	20-43 × 17-30	One or more basal cells paler	Absent
M. nigra	Hyphae laterally or terminally dense, cylindrical, septate, straight or curved, pale brown	Obovoid, muriform, slightly constricted at the septa, brown to black	20-37 × 15-23	Pale brown	Absent
M. monilicellularis	Micronematous, fasciculate, obscure septa, hyaline	Obovoid , muriform, constricted at septa, black, uppep ¾ dark brown, basal ¼ pale brown	33-45× 20-27	Hyaline	Absent

Discussion

Three other species of Monodictys have conidia that are morphologically similar to those of M. appendiculata (Table 1). However, *M. appendiculata* is the only species of Monodictys that has an appendage on the conidia. In addition, the conidia of *M. appendiculata* are overall larger than those of *M. monilicellularis, M. nigra*, or *M. paradoxa. M. monilicellularis* is distinctly pigmented with the basal quarter being pale brown and the top ¾ being dark brown.

The teleomorph of Monodictys has been determined as *Nereiospora* E.B.G. Jones. R.G. Johnson & S.T. Moss 1983, *Ohleria* Funkel 1868, and *Tubeufia* Penz & Sacc. 1898 (Hyde et al, 2011).

M. appendiculata from *M. paradoxa* (Corda) Hughes *M. nigra* Matsush and *M. monilicellularis* base on the morphological feature. There are differenced several characters; e.g. *M. appendiculata* has bigger size than those species. Conidium shape of this species is ellipsoidal and irregular on outline while *M. paradoxa* is oblong and obovoid to muriform are present on *M. nigra.* and *M. monilicellularis* In the conidia color of *M. appendiculata* is similarly with *M. nigra* in black to brown but somewhat deferent to dark olivaceous or blackish olive color from *M. paradoxa*. *M. appendiculata* also different from *M.*

monilicellularis becases *M. monilicellularis* has hyaline color However, one important part that found only From these data, it was distinguished and called new species.

Acknowledgments

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Part B
Study herbarium *Patellariaceae*

The Study on herbarium will help us to understand the biodiversity and natural conservation of fungi. Because of herbarium specimens were shown the typical character of each species. The information were useful for taxonomy and identification. So, herbarium specimens were loan from many herbaria. They were studied on morphological, taken a photo and description. Nine from 15 genus from this family were studied and the list have been shown in table1. Some of them were shown the plate (Fig 1-10).

Table 1 List of herbarium in *Patellariaceae* have been studied.

No.	Herbarium species	Herbaria name	Process
1	*Rhizodiscina lignyota* (Fr.) Hafellner Synonymy: *Karschia ligniyota*	Botanische Staatssammlung München, Germany (M)	Plate & description
2.	*Patellaria atrata* (Hedw) Fr.	Royal Botanic Gardens, U.K. (K)	Plate & description
3.	*Lecanidiella contortae* Sherwood	Department of Botany Naturhistorisches Museum Wien, Austria (W)	Plate & description
4.	*Lirellodisca pyrenulispora* Aptroot [as 'pyrenulaspora'], in Aptroot & Iperen	Centraalbureau voor Schimmelcultures, Utrecht, Netherlands	Plate & description
5	*Pseudoparodia pseudopeziza* (Pat.) Theiss. & Syd.	Farlow Reference Library and Herbarium of Cryptogamic Botany Harvard University, U.S.A. (FH)	Plate & description
6.	*Baggea pachyascus* Auersw.	Garten und Botanisches Museum Berlin Dahlem, Germany (B)	Plate & description
7.	*Endotryblidium insculptum* (Cooke) Petr.	Royal Botanic Gardens, U.K. (K)	Plate & description
8.	*Murangium sequoia* (Plowr. ex W. Phillips) Seaver	The New York botanical Garden, U.S.A (NY)	Plate & description
9.	*Poetschia buellioides* Körb.	National Herbarium Nederland, Leiden University branch, Netherlands (L)	Plate & description
10.	*Stratisporella episemoides* (Nyl.) Hafellner	Botanical Museum, Finnish Museum of Natural History University of Helsinki, Finland (H)	Studying
11.	*Banhegyia setispora* L. Zeller & Tóth	Botanical Department Hungarian, Natural History Museum, Hungary (BP)	Requested
12.	*Holmiella Sabina* (De Not.) Petrini, Samuels & E. Müll.	Royal Botanic Gardens, U.K. (IMI)	Requested
13.	*Schrakia crassula (Starbäck) Hafellner*	Swedish Museum of Natural History, Sweden (S)	Requested
14.	*Rhytidhysteron brasiliense Speg.*	Swedish Museum of Natural, History, Sweden (S)	Requested
15.	*Tryblidaria breutelii Rehm*	Swedish Museum of Natural History, Sweden (S)	Requested

Rhizodiscina

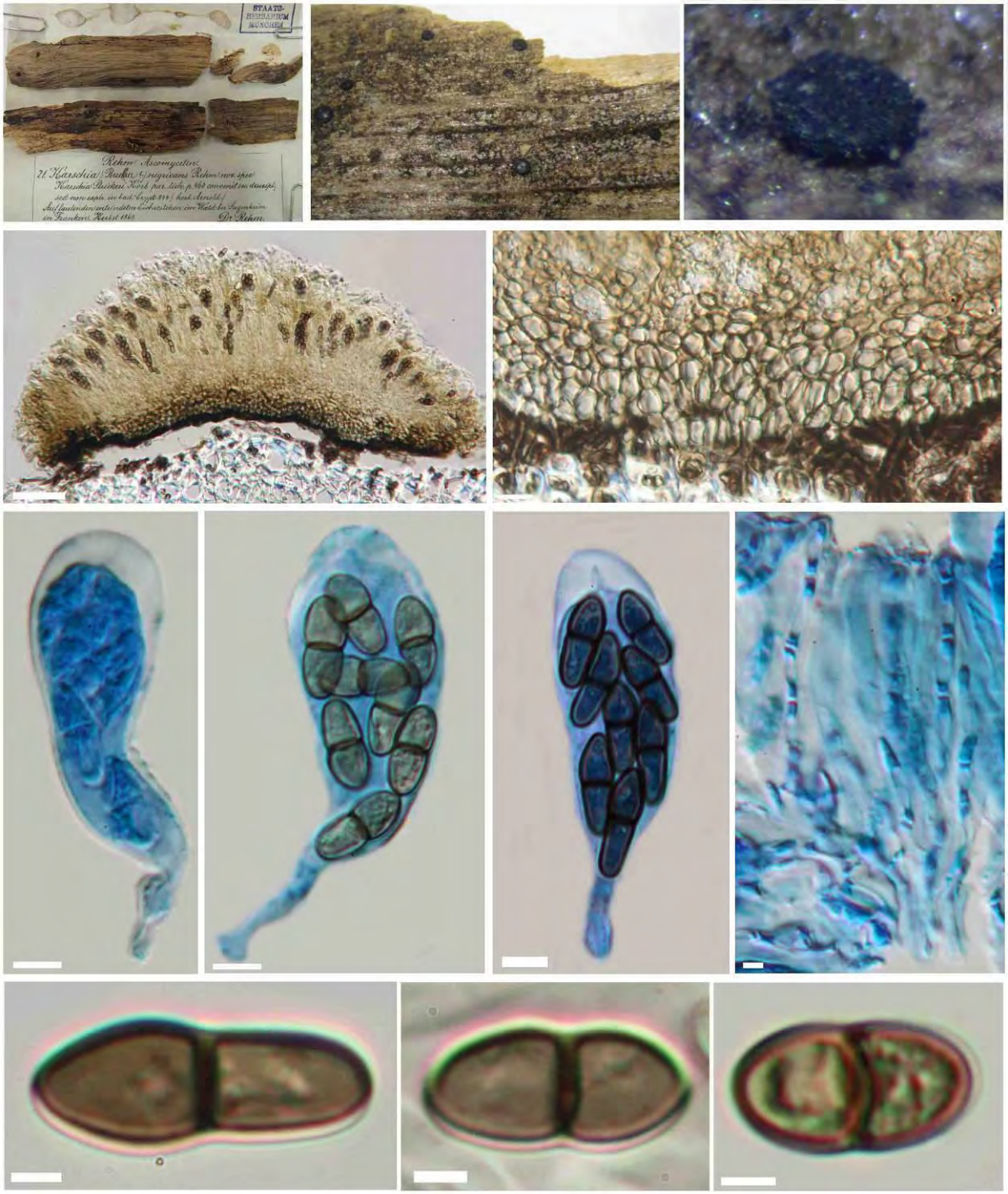

Plate1 (**A-L**) *Rhizodiscina* Hafellner, **A.** Herbarium specimen, **B-C.** Fruiting body on host tissue, **C.** Vertical section of ascoma, **D.**Section of exciple, **F.** immature asci after strained by cotton blue, **G-H.** Mature with bitunicate and 8-spores inside, **I.** Hamathecium with septate, **J-L.** Several kinds of mature spore. Scale bar: D = 50µm; E = 10 µm; F-H = 5 µm and I-L = 2 µm.

Rhizodiscina Hafellner

Rhizodiscina Hafellner, Nova Hedwigia, Beih. 62: 195 (1979)

Synonyms:

Peziza lignyota Fr., Syst. mycol. (Lundae) 2(1): 150 (1822)

Patellaria lignyota (Fr.) Fr., Summa veg. Scand., Section Post. (Stockholm): 366 (1849)

Karschia lignyota (Fr.) Sacc., Syll. fung. (Abellini) 8: 779 (1889)

≡*Peziza lignyota* Fr., Syst. mycol. (Lundae) 2(1): 150 (1822)

Buellia lignyota (Fr.) E. Müll., in Müller & von Arx, Beitr. Kryptfl. Schweiz 11(no. 2): 257 (1962)

Generic description: *Ascomata* superficial, subglobose and non-open when young, exposed when mature, apotechia, black. *Hamathecium* with septate, branched, forming on the top of asci. *Asci* clavated, broadly on the top, bitunicate, 8-spored. *Ascospores* obovoid or oblong, 1-septate, dark brown.

Known anamorphs: None

Literature: Hafellner & Poelt, 1976; Hafellner, 1979; Erikson & Hawksworth, 1993.

Type species: *Rhizodiscina lignyota* (Fr.) Hafellner

Other representative species: -

Rhizodiscina lignyota (Fr.) Hafellner, Beih. Nova Hedwigia 62: 195 (1979) Plate 1A-L.

≡*Karschia lignyota* (Fr.) Sacc., Syll. fung. (Abellini) 8: 779 (1889)

Ascomata 204-208µm high × 580-625µm diam., solitary, scattered, subglobose, closed when young and becoming exposed like disc, apothecia, black, subglobose to ovoid, margin slightly over than the centre, a bit rough at rim (Plate 1B-C).*Hypothecium* 8-25 µm wide, yellowish or light brown, comprising with textular angularis tissue, thick on the base, thin at beside (Plate 1D-E). *Hamathecium* of dense, paraphyses, upper 1 µm, septate, branch, swollen on the top, forming on above the asci (Plate 1I). *Asci* 45-50 × 13-16 µm ($\bar{x}$ = 47 × 14 µm, n = 10), 8-spored, bitunicate with ocular chamber, clavate, broadly on the top, pedicel (Plate 1A-L). *Ascospores* 9-14 × 4-5.25 µm ($\bar{x}$ = 11.5 × 4.8 µm, n = 10), irregularly or clowned, obovoid to fusiform, two-celled,1-septate and constricted at the septum, smooth(Plate 1J-L).

Anamorph: None

Cultures: None

Material examined: Geramane, Sugenheim Franken, on Auf faulenden entrindeten Eichaestchen im Wald, 1869, leg. Dr. Rehm, M 0177903 (Neotype)

Notes:

Morphology

Karschia was introduced to patellariaceous fungi by Hafellner&Poelt (1976) by used the character as follow "Maskierte' bitunicate Ascomycetum mit amyoiden Kappen und amyloider Hymenialgaerte, die vorläufig am ehesten zu den *Patellariaceae* zu stellen wären". This fungi was studied again by Hafellner (1979) and give the name *Rhizodiscina*.They was included in *Patellariaceae* by Eriksson &Hawksworth (1993) until now. There are many characters of *R. lignyota* that similar with *Poetschia andicola*; hamathecium shape and size, asci and spores, epithecium and hyphae. But one thing that used for distinguishes both of genera is the massive exciple of *R. lignyota* nearly a haft of ascoma than *P.andicola.*

Phylogenetic study: Not reported

Concluding remarks:

Recently, *Rhizodiscina* is using as current name of *Karschia*. There are 2 species within this genera; *R. lignyota* and *R. proteae* (index Fungorum, 2008); This genus still lack molecular, phylogeny information.

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Patellaria

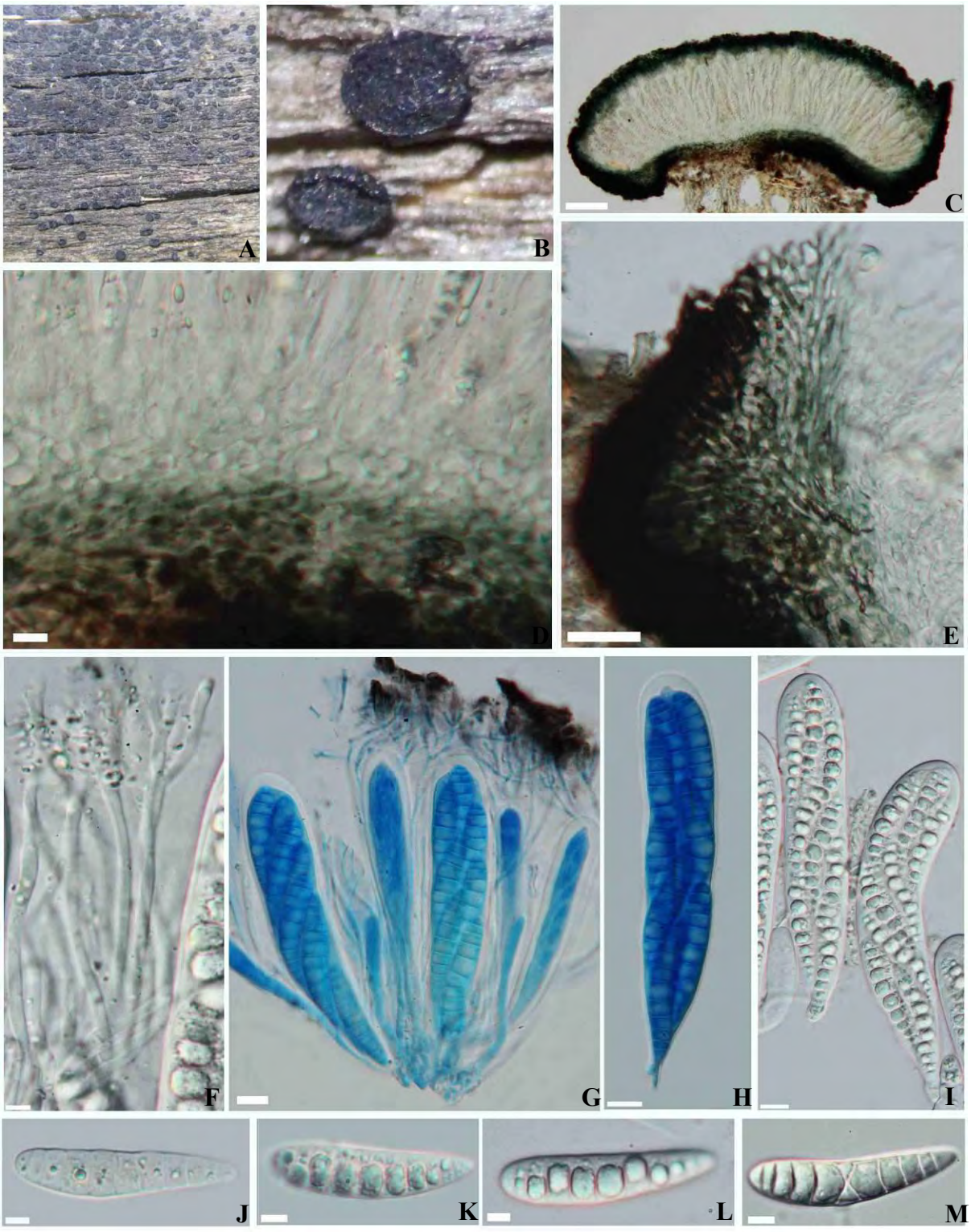

Fig1 (A-M). *Patellaria atrata* (Hedw.) Fr.Endl (IMI 32777) [*Patellaria*]. **A-B.** Ascomata on host tissue, **C.** Hand section of ascoma, **D.** Peridium with texular angularis, **E.** Exciple with elongated and radiating cells, **F.** Hamathecial tissue, **G.** Yong asci and mature asci with occular chamber after applied by cotton blue, **I.** Mature asci with 8-spored, **J-M.** Ascospores. Scale bars: C=50 µm; D, G-I=10 µm; E=20 µm; F, J-M=5µm.

Patellaria Fr.

Patellaria Fr., Syst. mycol. (Lundae) 2(1): 158 (1822)

Synonyms:

Lecanidion Endl., Fl. poson.: 46 (1830)

Generic description: *Ascomata* scatter, closed at first and opening and wide at longitudinal, apothecium, superficial, circulate or a bit elongated, composed with carbonaceous at rim, exposed flat, dark. *Exciple* multi layer were composed, dark brown cell were formed at outer, thick, isodiametric was shown, lighter color cell at inner layer was shown. *Hamathecium* was paraphysoid when too young, paraphysis- like when mature, septate, slender, hyaline, forming greenish brown epithecium at above of asci. *Asci* clavate to cylindrict or slightly fusiform, fussitunicate, 4-8 spored. *Ascospore* clavate, slightly curved, distoseptate, hyaline

Known anamorphs: Not reported

Literature: Hedwig, 1788; Rehm, 1896; Nannfeldt, 1932; Betler, 1939: 1940; Dennis, 1968: 1978; Luttrell, 1973; von Arx & Müller; 1975; Eriksson, 1981 and Bellemére et al. 1986

Type species: *Lecanidion* ***atratum*** (Hedw.) Endl.

Other representative species: -

***Patellaria atrata* (Hedw.) Fr., Syst. mycol. (Lundae) 2(1): 158 (1822)**

≡*Bacidia sublubens* (Paulson) Zahlbr., Cat. Lich. Univers. 8: 409 (1932)
≡*Bilimbia sublubens* Paulson, Trans. Br. mycol. Soc. 12(2-3): 88 (1927)
≡*Cyciedium atratum* (Hedw.) Wallr. [as'Cycledum atrum'], Fl. Crypt. Germ. (Norimbergae) 2: 511 (1833)
≡*Lecanidion atratum* (Hedw.) Endl., Flora Pason 1: 46(1830)
≡ *Lichen atratus* Hedw., Descr. micr.-anal. musc. frond. 2: 61 (1798)
≡*Lecanidion atratum* (Hedw.) Endl., Flora Pason 1: 46(1830) f. atratum
≡*Lecanidion atratum* (Hedw.) Endl., Flora Pason 1: 46(1830) var. atratum
=*Lichen atratus* Hedw., Desc. Micr-anal. Musc. Fround. 2: 61(1798)
≡*Patellaria maura* Massee, Bull. Misc. Inf., Kew: 131 (1898)
≡*Peziza atrata* (Hedw.) Schumach., Enum. Pl. (Kjbenhavn) 2: 417 (1803)
=*Lichen atratus* Hedw., Descr. micr.-anal. musc. frond. 2: 61 (1798)
≡*Peziza patellaria* Pers., Syn. meth. fung. (Göttingen) 2: 670 (1801

Ascomata 40-302 µm high × 59-1160 µm diam. ($\bar{x}$ = 194 × 600 µm, n = 10), solitary, scattered, superficial, closed when young, opening as disc or flatten at middle when mature, swollen or slightly folded at the margin, globose, apothecia, slightly gelatinous when add more water, black (Fig 1A-B). *Exciple* 46-76 µm wide($\bar{x}$=66 µm, n=10)composed with 2 layers, black and thick cell at out site, inner layer formed of isodiametric cell, greenish black (Fig 1E). *Hypothecium* of textular angularis, greenish or gray, pseudoparenchymatous cells at lower (Fig 1D). *Hamathecium* as gelatinous, filamentous like, 1.8-2.7 µm wide ($\bar{x}$ = 2.4µm, n = 10), slightly branched, hyaline, formed network at above of asci, carbornoues cell were formed, black (Fig1 E,G). *Asci* 98-135 × 15-29 µm ($\bar{x}$ = 120 × 22µm, n = 10), 8-spored, with bitunicate, cylindrical to clavate, a distinct pedicel, ocular chamber (Fig 1G-I). *Ascospores* 30-45 × 7-10 µm ($\bar{x}$ = 36.8 × 8.6 µm, n = 10), irregularly and biseriate, fusiform to obovoid, rounded on the top, young ascospore , show the 8-9 euseptate nature clearly whereas mature spore, 6-7 transverse distosepta, cell lumen is condense, hyaline (Fig 1J-M).

Anamorph: not reported

Cultures: VKM: WDCM133, CBS: 958.97(unverified, LSU-GU301855, SSU-GU296181)

Material examined: U.S.A: South Dakota, Northville; on wood *Acer negundo*., October 1925, J.F. Brenckle [Petrak Myc.g.exs 1556; ex IMI 32777, type).

Notes:

Morphology

The main characters of fungi in this species are apothecioid, superficial, black, greenish black of epithecium were formed by a branched and swollen paraphyses, bitunicate or fissitunicate asci, clavate to cylindrict, clavate and hyaline ascospores, there are numerous in asci and spore size, number of spore in asci, host. Those are important information and need to study in the future. Because all information can be used to taxonomy when species related the genus.

Phylogenetic study: There have been few molecular investigations of *Patellaria atratum* when compared to the morphological studies. Only 5 sequence is available in GenBank such as LLS and SSU etc. Moreover, for clarifying and correct identification this species. Both information of morphology and phylogenetic study are very important.

Concluding remarks:

Recently, the species under this genus still not clear yet. Ten species were approximated by Hawksworth et al.(1995). In 1940, five species were reported by Butler. But Dennis (1964: 1978) transferred *P. clavispora* Berk.&Br.(1854) to *Patellatopsis* Dennis in the *Dermateaceae*. Also 1954, Dennis reported 4 species from West Indies which included with new species of *P. jamaicensis* Dennis. Seven species were reported again from India (Tilak&Srinivasulu, 1974). *P.desertorum* Kravtzev and *P. schwarzmanniana* Kazhieva were maintioned from Kazakhstan in addition to *Patellaria atrata* by Schwarzman&Kazhieva (1976). Later, new species of *Lecanidion albizziae* A.K. Pande from India was described by Pande (1977). Until present, the reassessment of species needed to studying for clarifying and not delimited yet.

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Lecanidiella

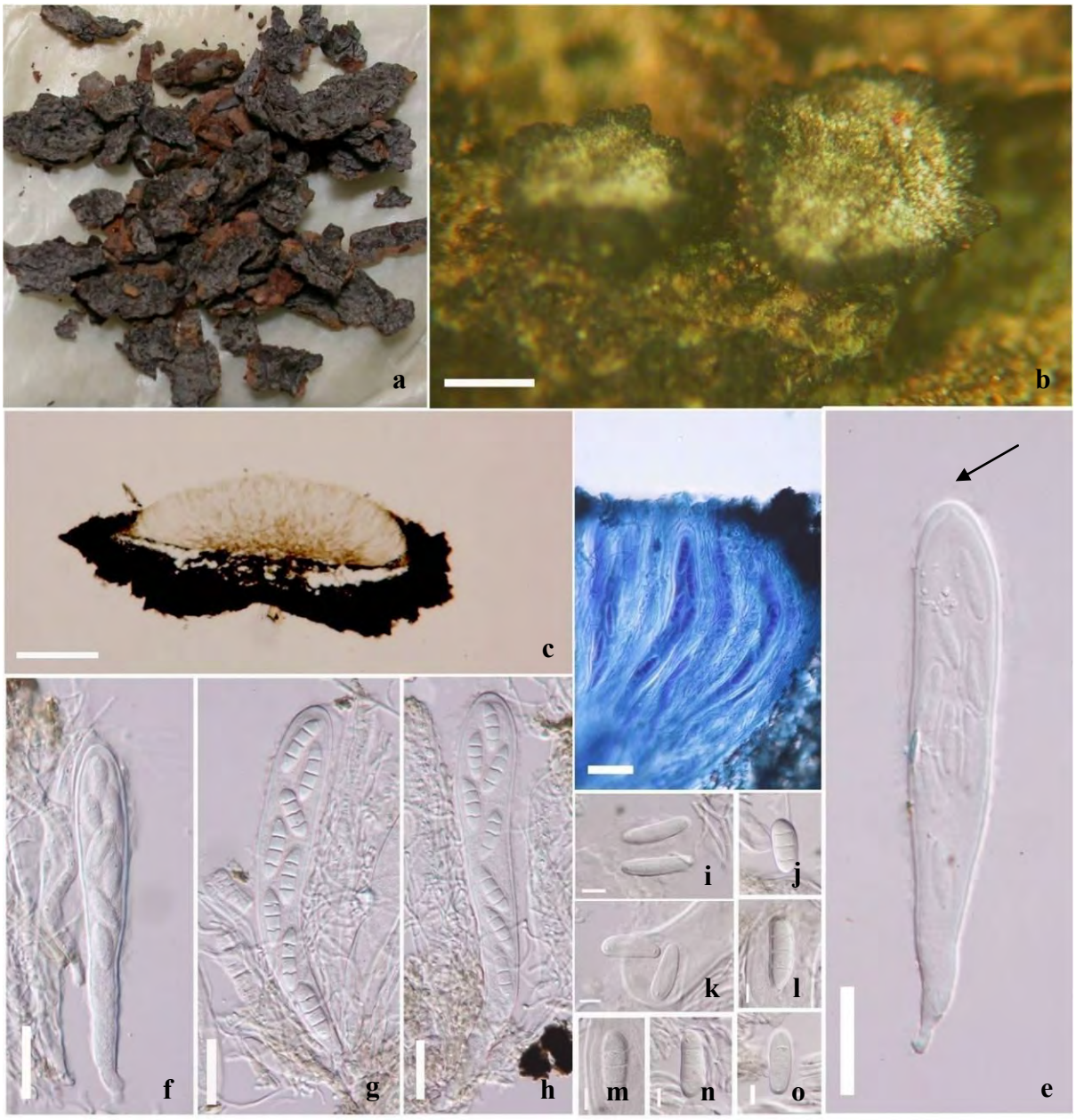

Fig 1 (a-o). ***Lecanidiella contortae*** **Sherwood. a.** Herbarium specimen; **b.** Appearance of hysterothecium on bark of *Pinus contorta*; **c,d** Vertical section of ascocarp; **e,f.** Young Asci with ocular chamber (arrowed); **g,h.** Mature asci with 2 layers; **i-o.** Ascospores. Scale bars: b, d, e-h = 20 µm., c = 150 µm and i-o = 5 µm.

Lecanidiella

Lecanidiella **Sherwood,** Sydowia 38: 272 (1986) [1985]
Generic description: ***Ascomata*** At early state closed and immersed, it became erumpent and superficial through host surface latter, apothecium, tooth-like at the rim, rought, becoming to carbonaceous, black. ***Exciple*** compose of pseudoparengchymatous, thick-walled.

Hypothecium comprising with textular intricata, brown hyphae, branced. Hamathecium is paraphysoid, slender, hyaline, quit enlarged on the tip, connected like network to forming the brown yarn or hair- like epithecium. ***Asci*** cylindrical to clavate shape, bitunicate, 8-spored**.** ***Ascospore*** oblong to fusiform, hyaline, 4-cells and 3 septates, no sheathed.

Known anamorphs: It had not been reported.

Literature: Pirozynski and Reid (1966), Funk (1967), Sherwood (1977; 1980; 1997), Pretini et al. (1979), Eriksson (1981).

Type species: *Lecanidiella contortae Sherwood*

Lecanidiella contortae Sherwood, Sydowia 38: 274 (1986) [1985] Fig 1a-o.

Ascomata 4-7 mm high×12-19 mm diam.($\bar{x}$ = 6×16 µm; n=10), scatter on the bark, closed and immersed on host surface when young, erumpent trough host surface, opening and expose, superficial, apothecium, irregular at the rim, rough, slightly zigzag, black, outer of receptacle composed with carbonaceous, black. ***Exciple*** dark brown to black, thick-walled, cracking. ***Hypothecium*** 70-155 µm wide(average 100 µm; n=10)**,** attached on host surface at the base, composed of textular intricate. ***Hamathecium*** branched and weaved of paraphysoids, cylindrical, hyaline. ***Asci*** 80-115 µm×12-17 µm ($\bar{x}$ = 105 × 15 µm, n = 10), 8-spores cylindrical, clavate and slightly broadly on the top, ocular chamber present, bitunicate. ***Ascospores*** 14-15.5×(5.5-)6-7.5 µm ($\bar{x}$ = 6.9×14.8 µm, n = 10), oblong, fusiform with round off the end, uniseriate, broad fusiform to fusiform with broadly to narrowly rounded ends, hyaline, constriction at septa, 4-cells, 3 septate, smooth, uniseriate at first haft and biseriate at the second haft.

Anamorph: It had not been reported.

Cultures: none

Material examined**:** U.S.A: Origon, Linn County, barren area near Santiam Summit, elev. *Ca* 4500 ft.; on bark of *Pinus contorta*, 12 June 1983; M. Sherwood and L. Pike (BPI 674929-holotype).

Notes:

Morphology: *Lecanidiella contortae* is distinguished or characterized by apothecia ascomata, erumpent on host surface, teeth like at margin, powder like on the middle, hypothecium with textular intritica, bitunicate asci and ascospore with 3-septate, hyaline.

Phylogenetic study: Not reported

Concluding remarks:

The *Lecanidiella* genus had been recorded that there is only one species in this genera. They were distributed on mountain area of western North America. In 1986, Sherwood had been reported and confirm the previously observed (Pirozynski & Reid, 1966; Petrini et al., 1979) that both of *Lecanidiella* and *Homienalia* have the ascomata as closed the parenchyma tissue, crack, irregular when opening to exposed hymenium. But different in spores characters. This genus was related to *Patellaria* of *Petellariaceae* and *Melittosporiella* of *Rhytismatales* by Sherwood (1986). Finally, it is promoting and calling that *Lecanidiella*. Because it can be present on unitunicate or bitunicate. So, somewhat it was similarly and closely with *Melittosporiella* too much. However, *Lecanidiella* still is in *Petellariaceae* family until we can get knowledge about asci structure more.

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Lirellodisca

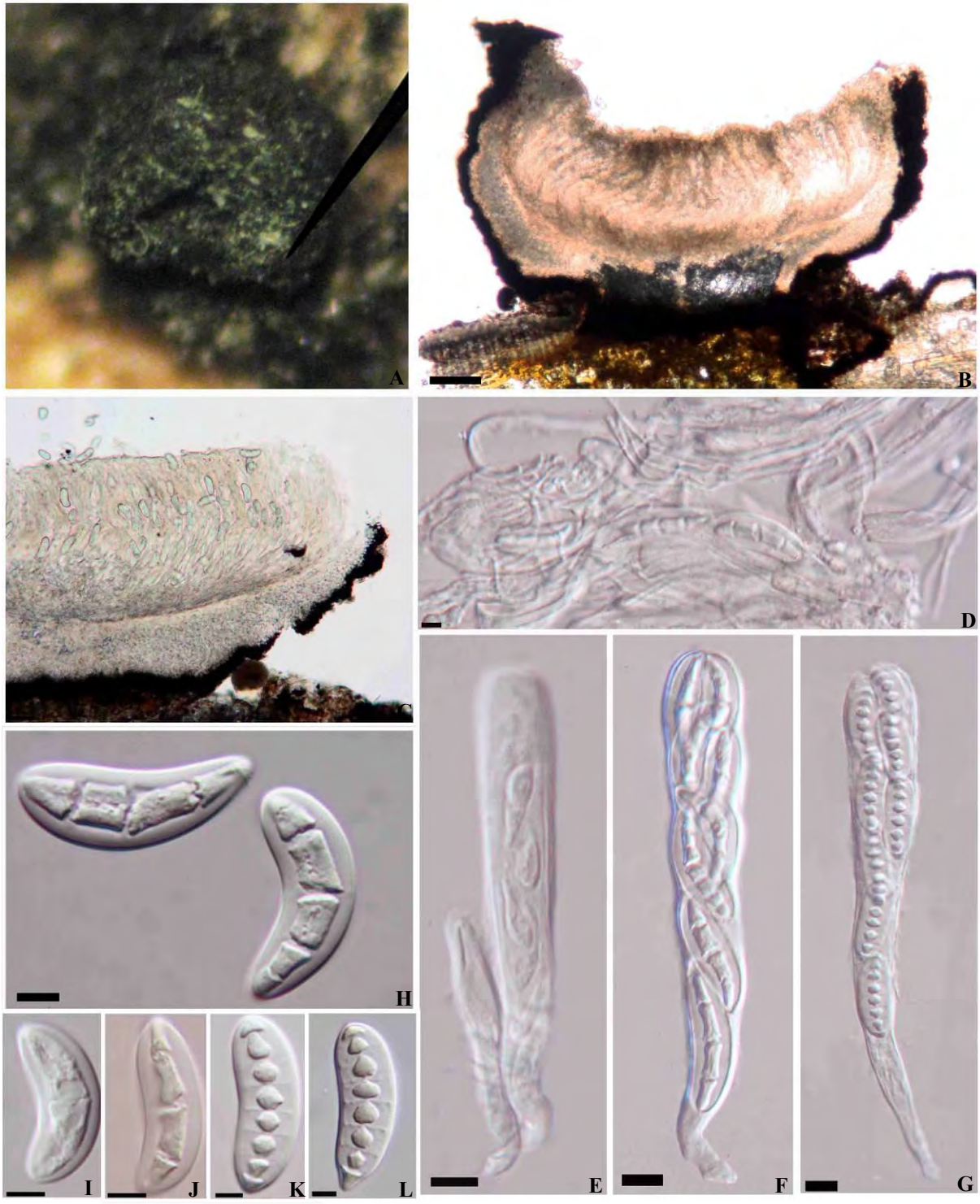

Plate 1(A-G) ***Lirellodisca pyrenulispora*, A.** Ascoma on host tissue, **B.** Hand section of ascoma, **C.** Peridium, **D.** Pseudoparaphyset, **E-F.** Immature asci, **G.** Unituicate with 8-spores, **H-J.** Young spore, **K-L.** Mature spore. Scale bars: B=120 µm; C=100 µm; D=3 µm; E=7 µm; F-G=10 µm; I-L=5 µm.

Lirellodisca

Lirellodisca Aptroot, in Aptroot & Iperen, *Nova Hedwigia* 67(3-4): 485 (1998)
Generic description: *Ascomata* superficial, subglobose, black, unsmooth at the rim. *Peridium* thick, light brown. *Hamathecium* un-brached, non-septate, hyaline. *Asci* oblong to clavated with pedicle, bitunicate?, 8-spores. *Ascospores* ellipsoidal with curved at middle, 1-5 transverse eusept and 6-7 transverse distosepta, hyaline.
Known anamorphs: Not reported.
Literature: Aptroop & Iperen, 1998.
Type species: *Lirellodisca pyrenulispora* Aptroot [as '*pyrenulaspora*'], in Aptroot & Iperen.
Other representative species:-
Lirellodisca pyrenulispora Aptroot [as '*pyrenulaspora*'], in Aptroot & Iperen, *Nova Hedwigia* 67(3-4): 485 (1998) Plate 1A-L.

Ascomata 380-455 µm high ×1,125 -2,350 µm diam., single, scattered, superficial, erumpent on the bark, a bit round, margin is slightly over than the centre and thick, rough at rim (Plate 1A). *Hypothecium* 80-120 µm wide, two-layered, comprising with dark cell of pseudoparenchymatous outside and next is textular of epidermoidea, near the basal is thick layer than beside (Plate 1B-C). *Hamathecium* of pseudoparaphyses in gelatines, up to 1 µm wide, filamentous like (Plate 1D). *Asci* 100-175×15-25 µm ($\bar{x}$ = 135×20 µm, n = 10), 8-spored, bitunicate, cylindrical to oblong, slightly broadly on the top (Plate 1E-G). *Ascospores* 26-36×10-12 µm ($\bar{x}$ = 12×26 µm, n = 10), biseriate, ellipsoidal with curved at the middle, round at the ends, 6 transverse euseptate and mostly 6 transverse of distoseptate, hyaline (Plate 1H-L).

Anamorph: None
Cultures: None
Material examined: Northern province, Owen Stanley Range, Myola, along trail from gueshouse to Naduri., On twigs of *Elaeocarpus* in primary montane forest., 17 October 1995., leg. A. Aptroot (Aptroot no. 38022, holotype).

Notes:
Morphology

The genera of *Lirellodisca* was explained and resembling to *Pyrenula* Acharius which unrelated with them. By used the character of unusual distoseptate as a key. Then it was revision again and grouping by ascoma apothecial like with unrelated *Durella* Tulasne & C. Tulasne. Exactly, it might more related with *Arthoniales*. And very closest to *Phyneromyces* Spegazzini & Hariot. However, this genera was accepted to *Patellariaceae* by Kutorga & Hawksworth (1997) event it not related with any species in here. Because it has cup shape and thick distoseptate which is different to *Arthoniales* and *Phyneromyces* Spegazzini & Hariot respectively (Aptroop, 1998).

Phylogenetic study: None

Concluding remarks:

Recently, there are 2 species in this genera. Including to *Lirellodisca pyrenulispora* and *Lirellodisca pyrenulospora* were listed in index Fungorum (2008).In the future molecular study usefull for confirm the morphological study and clarify the classification of this genera as well

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Pseudoparodia

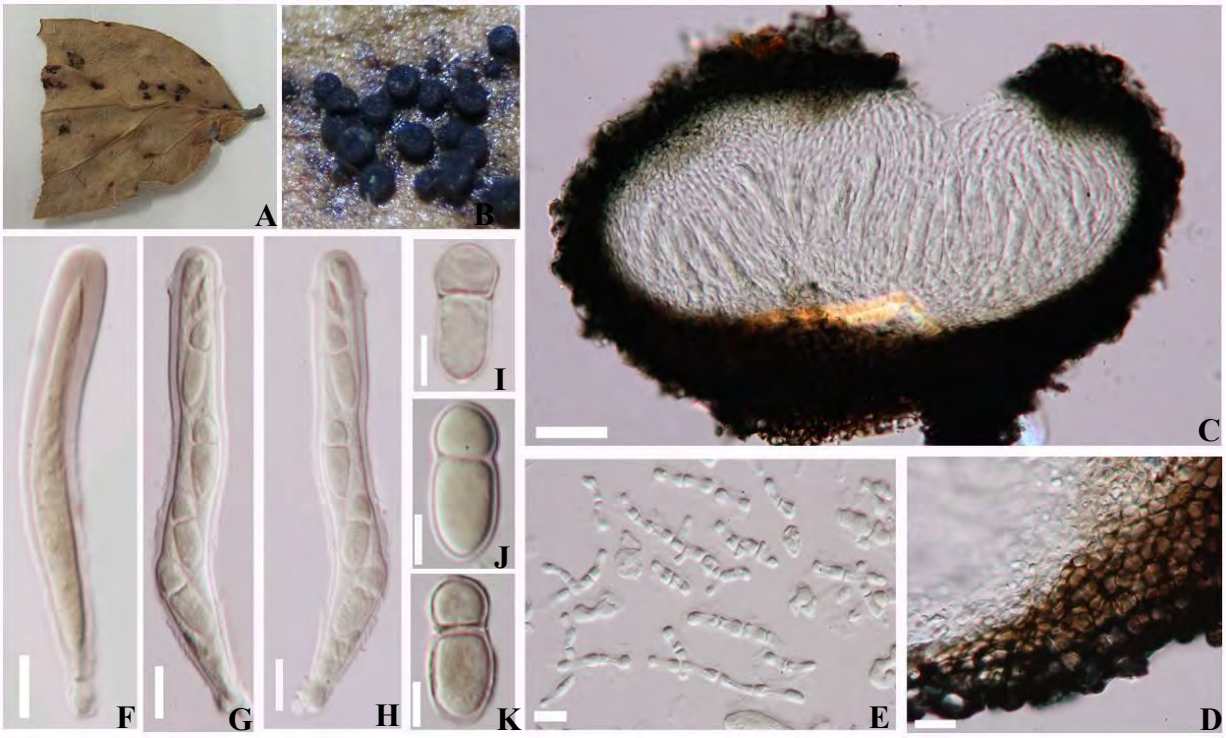

Fig5 (a-k). ***Parodiella pseudopeziza*** (FH 7735, holotype)A. Herbarium specimen, B. Fruiting body on host tissue**,** C. Vertical section on ascoma, D. Peridium of *textura angularis*, E. Hamathecium with constriction at septate, F. Young asci was strained by Maelzer), G-H. Bitunicate asci with 4 ascospores, I-K. Ascospores with a septate**.** Scale bars: C = 50µm; D, E-G = 10µm; H = 7µm;I-K = 5µm.

Pseudoparodia

Pseudoparodia Theiss. & Syd., Annales Mycologici **15** (1-2): 1917.

Synonyms:

Parodiella pseudopeziza Pat., Bull. Herb. Boissier 3(1): 67 (1895)

Generic description: *Ascomata* perithecium, globose and periticoid, superficial. *Peridium* thick walled, black. *Hamathecium* of paraphysis septate, broadly and dense above the asci. *Asci* cylindrical, bitunicate, hyaline, 4-spored. *Ascospore* ellipsoidal, 1-septate, 2-cells not equal, round of the end, hyaline

Known anamorphs: Not reported

Literature: Saccardo(1884); Müller and Arx (1975); Petrak (1947); Zhang and Hyde (2009).

Type species: *Parodiella pseudopeziza* Pat. 1895

Other representative species: None

Parodiella pseudopeziza Pat., *Bull. Herb. Boissier* 3(1): 67 (1895) Plate1(a-k)

≡ *Parodiella pseudopeziza Pat.* 1895, Saccardo's Syll. fung. 24: 1144 .

Ascomata 145-240 µm high×140-245 µm diam.($\bar{x}$=180×190; n=10) agglutinated and scattered, superficial, perithecium-like then a piece of vertical section become to convex after

applied by 5%KOH and slightly turned green-blue, subglobose, stroma bearing perithecial and supported them on the host surface, black. ***Peridium*** composed of *textular angularis*, thick and dense, dark brown to black (20-45 µm wide, $\bar{x}$ = 30; n=10). ***Hamathecium*** comprising cylindrical, slightly swollen, separated to small pieces, unbranched, dense and firmly above asci, very constriction at septum, a bit rough when applied by cotton blue (2-3 µm wide, $\bar{x}$ = 2.5; n=10). ***Asci*** 4-spored oblong to cylindrical, slightly curved**,** bitunicate with a short pedicle, (70-100 µm high, 9-11 µm diam., $\bar{x}$ = 90× 10; n=10). ***Ascospores*** elliptical, round at the end, very constriction at the septum, 1-septate, 2-cells slightly different, lower cell was longer and smaller than upper cell, uniseriate, hyaline to light brown (14-17 µm high, 5-9 µm diam., $\bar{x}$ = 16× 7; n=10)

Anamorph: None reported.

Cultures:

Sequence data: None

Material examined: ECUADOR: San Jorge; on leaves of *Vaccinium* sp., July 1892, G. Lagerh (FH 7735, holotype).

Notes:

Morphology

Pseudoparodia was introduced by Petrak (1947). It was mentioned as Discomycete and related with Venturiaceae. Zhang and Hyde (2009) examimed it that their ascomata are apothecium, which were superficial, paraphysoid with swollen, very dense and forming epithecia on asci. Those characters quit different from Venturiaceae family while their characters more closely to the concept of Patellareaceae. One genera in this family that can confirmed morphological relationship of this genus with Patellareaceae is *Stratisporella episemoides* (Nyl) Hafellner. Consequently, Ying and Hyde (2009) tried to transfer *Pseudoparodia* that has only one species, *P. pseudopeziza* into Patellareaceae.

Phylogenetic study: -

Concluding remarks: This genera has only one spcies which is *P. pseudopeziza* (Pat.) Theiss. & Syd and it still reported as the member in *Venturiaceae* (Index Fungorum, 2008). However, some information still are not correct and need to prove it in the future.

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Baggea

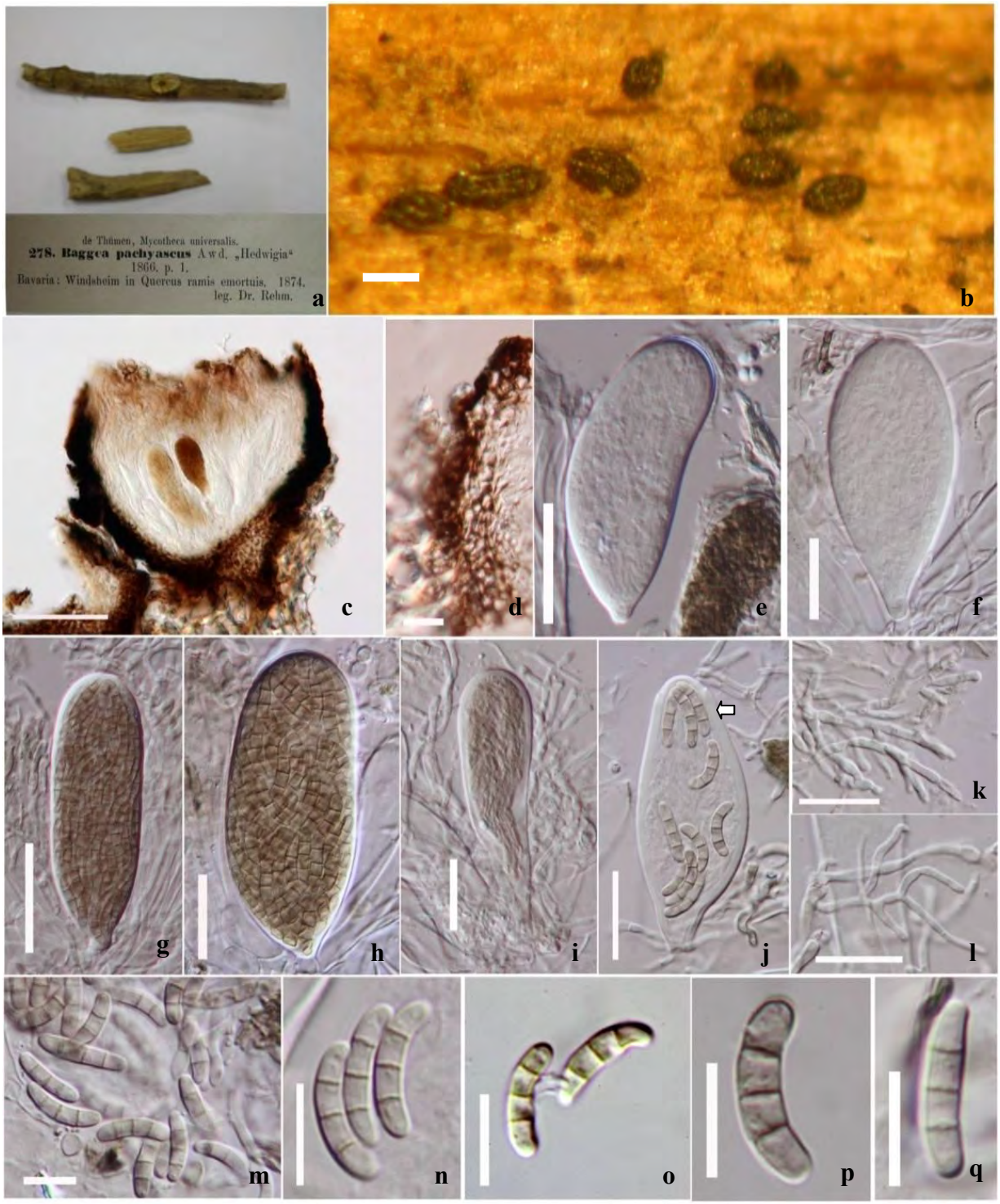

Fig. 1. *Baggea pachyascus* Auersw. a. Hebrarium specimen; **b.** Appearance of hysterothecium on host tissue; **c.** Vertical section of ascoma; **d.** *Hypothecium* ; **e-j.** Asci with 2 layers (arrowed); **k-l.** *Hamathecium*; **m-q.** Ascospores. Scale bars: b, m-p = 10 µm., c-l = 20 µm**.**

Baggea Auersw.

Baggea Auersw**.**, *Hedwigia* **5**: 1 (1866)
Generic description: *Ascomata* fusiform to ellipsoidal, superficial, black, opening by a longitudinal slit, margin slightly raised above flat and black slit *Peridium* composed of outer black to dark brown walled and inner paler brown walled, *texura globulosa-angularis*. *Hamathecium* comprising relatively wide, septate pseudoparaphyses, branching especially in upper part, occasionally anastomosed in the lower part, hyaline, tips slightly swollen and agglutinated. *Asci* polysporus, bitunicate, broadly clavate or clavate, with short stipes, an ocular chamber in the apical dome, asci not maturing simultaneously. *Ascospores* numerous in asci, allantoid, with rounded ends, with transverse septa, curved, brown, thin-walled.
Known anamorphs: None
Literature: Saccardo, 1883; Ark and Müller, 1975; Farr *et. al,* 1979; Kutorga and Hawksworth, 1997; Magnes *et.al*., 1998.
Type species: *Baggea pachyascus* Auersw.

Baggea pachyascus Auersw*., Hedwigia* **5**: 1 (1866). Plate 1a-q.

Ascomata 150-205 µm. in diameter X 175-250 µm. high ($\bar{x}$ = 180 X 215 µm, n = 10) fusiform to ellipsoidal, superficial, black, opening by a longitudinal slit, margin slightly raised above flat and black slit which widens on wetting, texture unclear when cross-section,(sometime circular, triangular or irregularly shaped) (Fig 1, c). *Hypothecium* medium, 23-54 µm. ($\bar{x}$ = 37.82 µm, n = 10), composed of outer walled black to dark brown and inner walled paler brown, *texura globulosa* to *angularis* (Fig 1, d). *Hamathecium* 2.9-4.3 µm wide ($\bar{x}$ = 3.63 µm, n = 10), comprising relatively wide, septate pseudoparaphyses 3-5 µm wide ($\bar{x}$ = 3.75 µm, n = 10), branched, especially in upper part, occasionally anastomosed in the lower part, hyaline, tips slightly swollen and agglutinated (Fig 1. k-l). *Asci* 99-144 µm X 29-51µm. high ($\bar{x}$ = 122X 41 µm, n = 10)**,** polysporus, bitunicate, broadly clavate or clavate, with short stipes andapical dome, the walled 1-2 µm (n = 10) thick in asci, asci not maturing simultaneously. *Ascospores* 12-15 µm X 2.3-4.2 µm ($\bar{x}$ = 13 X 3 µm, n = 10), numberous, allantoid, with rounded ends, usually with 3 transverse septa, only rarely with up to 6 septa, curved, brown, thin-walled, smooth.

Anamorph: not reported.

Cultures: none

Material examined: Botanischer garten UND, Botaniched museum, Berlin-Dahlem R. On *Quercus ramis emotuis*, at Windsheim, Bravaria, Dr. Rehm. 1874

Notes:
Morphology

The first time Aueswald(1866) introduced this genus in *Hysteriaceae*. ThenSaccardo(1883) and Rehm (1912) also agree with the first auther. When Nannfeld(1932) and Zogg(1962) studied again at characteristic of ascomata and paraphysoid. they moved this genus to *Lecanorales*. However Rehm(1896), Clements&Shear (1931) and von Arx & Müller (1975) assigned it to the *Patellariaceae*. but still not clear about the relationship of this genus and family. But *Baggea* has two characters that other genus didn't have: polyporus asci, cylindric and slightly curved ascospores. Now aday we know that there is only one species in this genus.
Phylogenetic study: none

Concluding remarks

Baggea Auersw. has to be included in the family *Patellariaceae*. This genera belong with apothecioid or histeriform**,** usually dark ascomata that are usually closed at the beginning of development, a pseudoparenchymatous excipulum with a crust-like outer surface, a pseudoparenchymatous or prosenchymatous hypothecium, a hamathecium consisting of paraphysoids forming a (pseudo-) epithecium above the asci (occasionally paraphyses are added in a later stage of development), bitunicate asci (fissitunicate where know), and septate, brown spores. A new description and illustrations for *Baggea* Auersw is *Baggea pachyascus* Auersw., there is one species of the genus.

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Endotryblidium Petr.

Endotryblidium

Endotryblidium Petr**.**, *Sydowia* **13**(1-6): 244 (1959)
Generic description: *Ascomata* apothecium, at first immersed, erumpent but still under host surface, irregular, black. *Peridium* composed of pseudoparenchymatous cells, thick. *Hamathecium* of paraphyses broad, anastomosing with carbonaceous cell at above. *Asci* clavated to oblong, bitunicate, 8-spored, with a short stipitate. *Ascospores* broad fusiform to oblong, rounded ends, crowed, hyaline when young, latter brown, two-celled, constricted at the septum.
Known anamorphs: none reported
Literature: Petrak, 1959

BRG5280002

Type species: *Endotryblidium insculptum* (Cooke) Petr.
Other representative species:-

Endotryblidium insculptum (Cooke) Petr., *Sydowia* 13(1-6): 245 (1959). Plate 1a-i.

≡ *Triblidium insculptum Cooke* [as *'Tryblidium'*] 1876

Ascomata 450-930 µm diam × 200-390 µm high ($\bar{x}$ = 260 × 660; n=10), immersed, scattered, erumpent but still under host surface, broad, hysterothecia look like to irregular, black. *Peridium* thin, attached with pseudoparenchymatous cells, cell wall 90-145 µm wide ($\bar{x}$=120; n = 10) thick, hard, near the base composed of hyaline hyphae mass producing asci. *Hamathecium* of paraphyses, long, 2.0-3.5 µm broad, anastomosing with carbonaceous cell at above. *Asci* 40-60 µm × 100-120 µm ($\bar{x}$=50×115 µm, n=10), 8-spored, bitunicate, clavate to oblong, with a 5-13 µm long, ($\bar{x}$=9 µm; n=10)short pedicel,. *Ascospores* 20-30 µm diam.×50-60 µm. long ($\bar{x}$=25×50 µm; n=10), crowded, broad fusiform to oblong with rounded ends, hyaline was young, brown on mature stage, two-celled, constricted at the septum.

Anamorph: not recorded.

Cultures:

Material examined: USA, New Jersey, Newfield; on dead branches of hickory., May 1874, Ellis (No. 2111)

Notes:
Morphology

Phylogenetic study: Not reported

Concluding remarks:

Reference
Petrak, F. 1959. Sydowia 13(1-6): 244.

Murangium Seaver

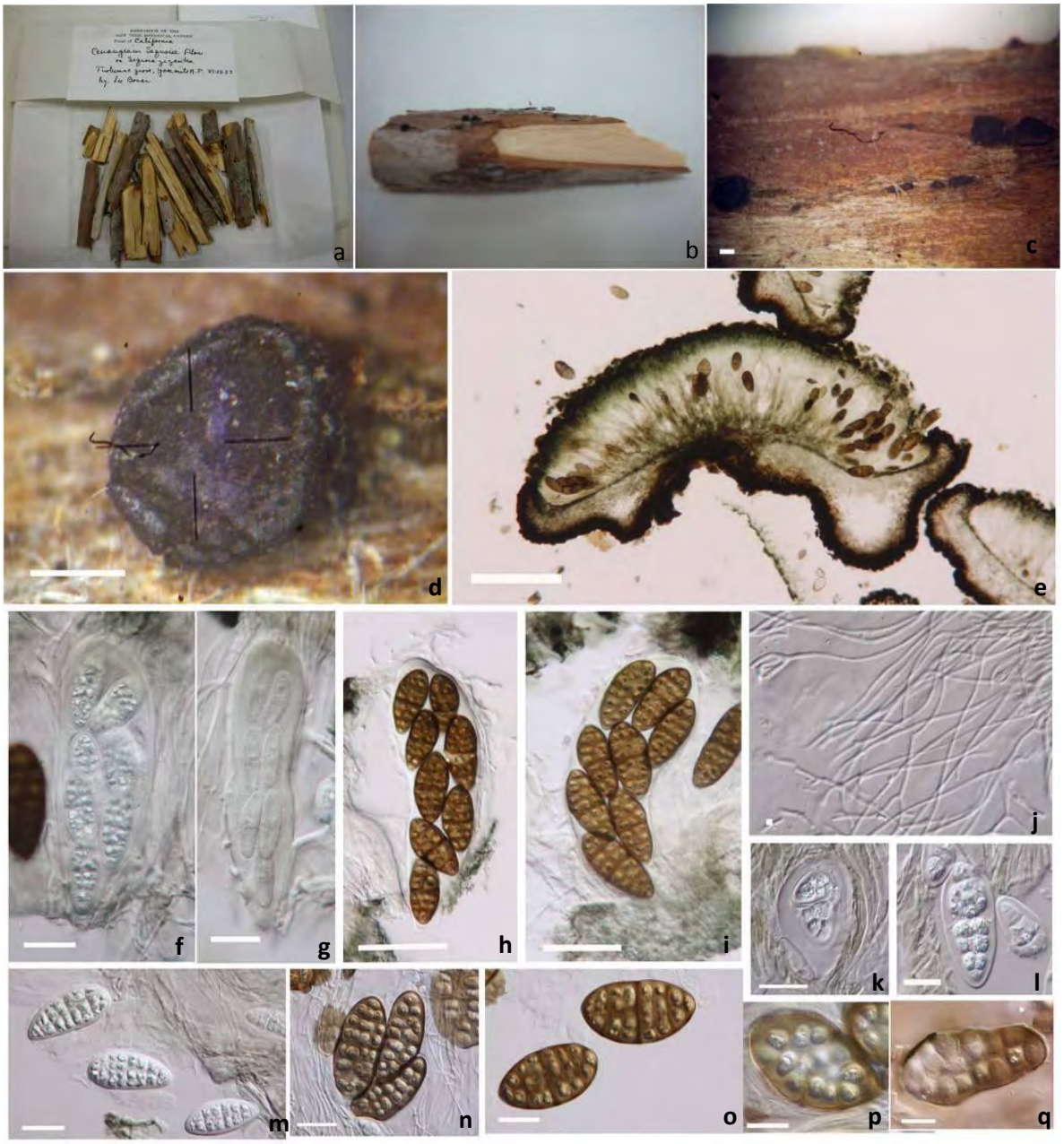

Plate1 ***Murangium sequoia*** **(a-q).**, **a-b.** Herbarium specimen, **c.** Ascomata on host surface, **d.** Close up of ascomata, **e.** Section of ascomata**, f-g.** Young asci, **h-i**. Asci, **j.** Paraphysoid, **k-m**. young ascospores, **n-q**. mature ascospores, Scale bar: c = 5 µm, j = 10 µm, l, p-q = 30 µm, d, f-g, k, m-o = 50 µm, e =300 µm,

Murangium

***Murangium* Seaver** *North American Cup-fungi*, (Inoperculates) (New York): 368 (1951).
Generic description: *Ascomata* apothecia, cup-shaped, closed and immersed when immature, became opening and exposed when mature, black. *Asci* bitunicate, broad-clavate, 8-spored. *Ascospores* obovoid-oblong, rough, brown and large.
Anamorphs reported for genus: None
Literature: Seaver (1951), Korf & Zuang (in Eriksson & Hawksworth 1987) Kutorga & Hawksworth (1997).
Type species: *Murangium sequoia* (Plowr. Ex W. Phillips) Seaver.
Other representative species: None
Murangium sequoia (Plowr. Ex W. Phillips) Seaver, North American Cup fungi (Inoperculates) (New York): 368 (1951). Plate 1a-q
=*Cenangium sequoiae* Plowr., (1878)

Ascomata apothecia, 490-562 µm. high ($\bar{x}$ = 530) and 1,318-1,546 µm. diameter, ($\bar{x}$ = 1,430), occurring in linear groups or single, closed and immersed at first, exposed and opening with an irregular aperture when mature, black, cup-like, and tapering, there is the folded margin over disc with lobes. *Hypothecium* with irregularly shaped cells with thick gelatinized walls, colourless. *Hamathecium* of paraphysoids, hyaline, septatesand branched like network on the top of them. *Asci* 218-443 µm. lengh ($\bar{x}$ = 334) and 62-143 µm. wide ($\bar{x}$ = 100), clavate and broadly, short base, bitunicate and apical domed. 8-spored inside. *Ascospore* ellipsoid or obvoid and oblong, rather rounded and the end, 2-3 seriate, 4-7 transverse and 1-2 longitudinal septa, all most constrict at the middle, hyaline when immature and became brown when mature, rough, 37-75 µm.wide ($\bar{x}$ = 49) and 85-153 µm. length ($\bar{x}$ = 106).

Anamorph: None.
Cultures and DNA sequences: None
Material examined: California, Tuolumne grove, in Yosemite n.P. v1.23.33, on a bark of *Sequoia gigautea***,** leg. Lee Bonar (NY).

Note:
Morphology: *Murangium* genus was in *Patellariaceae* and introduced by Seaver. Their ascospore inoperate in *Cenangiaceae* Rehm.Because their asci of *M. sequoiae* were fissitunicate.(Eriksson & Hawksworth, 1987). In addition, character of apothecioid, hamathecium of paraphysoids and bitunicate asci made them belong with *Patellariaceae* .

Phylogenetic study: not report

Concluding remarks:-There are variation in size of asci and ascospore that depend on author's work and material examined at different stage.

Reference:
Seaver F.J. 1951 The North American Cup-fungi (Inopercolates).- New York: Seaver.
Eriksson O.E. & Hawksworth D.L.1987. Nots on ascomycete systematic Nos 225-463.- Systema Ascomycetum 6; 111-165.
Kutorga. E& Hawksworth. D.L. 1997. A reassessment of the genera referred to the family *Patellariaceae* (Ascomycota) .-Systema Ascomycetum 15; 54-58.

Poetschia

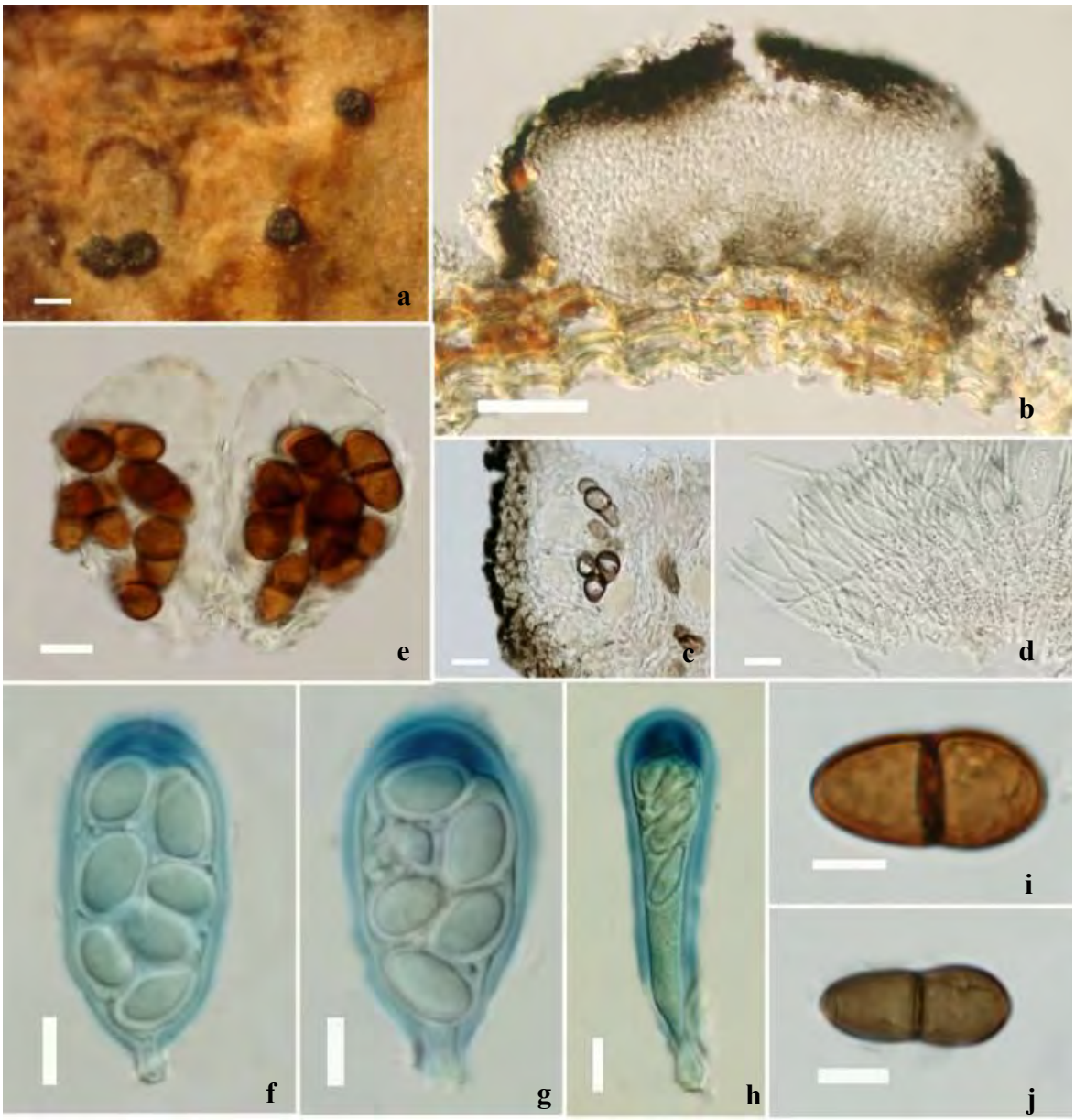

Fig1. *Poetschia buellioides* Körb (a-j). **a.** Apothecium on host tissue, **b.** Cross section with carbonaceous cell, **c.** Peridium comprising *textura angularis*, **d.** Elongated of hamathecail, **e.** Bitunicate after applied by cotton blue, **f-h.** Young asci, **i-j.** brown spore with 1 septate. Scale bare: a = 100 µm; b = 30 µm; c-d = 10 µm; e-j = 5 µm.

Poetschia

***Poetschia* Körb.,** Parerga lichenol. (Breslau): 280 (1861)
Generic description: Ascomata closed when young. Then become to open like cup shape when mature state, dark color and superficial, sessile, carbonaceous tissue appear at the outer surface receptacle. **Exciple** it was camefrom dark brown of pseudoparenchyma cell, thick walled. **Hypothecium** hyaline to pale brown of pseudoparenchyma**. Hamathecium** epithecium was show mid brown from the top of asci. **Asci** cylindrical to clavate, broadly on the top, bitunicate. **Ascospore** obovoid or broadly of truncate, 2 cells, smooth, brown.
Known anamorphs: It had not been reported
Literature: Saccardo 1889, Hafellner 1979 and Kutorga & Hawksworth 1997
Type species: *Poetschia buellioides* Körb
Other representative species: *P. andicola* (Speg.) Hafellner, *P. caerulescens* (Hafellner) Kutorga & D. Hawksw. and *P. caerulescens* (Hafellner) Kutorga & D. Hawksw.

Poetschia buellioides Körb., Parerga lichenol. (Breslau): 280 (1861). Plate1a-j.

Ascomata scattered, superficial, apothechium-like, ascoma closed when young and become opening when mature, sub-globose on out line, black, slightly convex after rehydrated on pieces of vertical section (100-105 µm high × 208-211 µm diam. $\bar{x}$ = 100× 210; n=10) *Exciple* composed of pseudoparenchyma cells, thick walled, brown to dark brown. *Hypothecium* pseudoparengchyma cells present, hyaline to light brown. *Hamathecium* were branched and septate on paraphysoid, hyaline, sticky, slightly swollen on the top, cylindrical (1.5-2 µm wide, $\bar{x}$=1.8 µm; n=10). *Asci* clavated with broadly on the top, or obovoid and broadly at the base, bituncate, thin layer, ocular chamber apparent and dense when young, 6-8 spored (36-45 µm high × 8-20 µm diam. $\bar{x}$ = 42× 12; n=10). *Ascospore* crowded in asci, ellipsoidal, constriction at the septate , 2-cells with 1 septate, upper part of cell larger than the lower part, smooth, brown to dark brown (17-19 µm high × 7.5-8.6 µm diam. $\bar{x}$ = 18× 8; n=10)

Anamorph: It had not been reported
Cultures: Not reported
***Material examined*:** Bei Gresten in Niederösterreich, auf Apfelbaumrinden (L 910.204-614).

Notes:
Morphology

At first, the genus of *Poetschia* has only one species which was *P. buellioides*. And it also was promoted *Karschia* Körb as the synonym of *Poetschia* by Saccardo (1889). The extendation of the species's number was occurred again by Hafellner (1979). The genus of *Poetschia* was associated to *Patellariaceae* family by Kutorka & Hawksworth (1997) They designed it by used some characters as their basis. Apotheciod shape, receptacle with pseudoparenchymatous and bitunicate asci.

Phylogenetic study: Not reported

Concluding remarks: Recently, there are 4 species belong to this genera and were reported as the member in *Patellariaceae* family (Kutorka & Hawksworth,1997). There are *P. andicola* (Speg.) Hafellner, *P.buellioides* Körb., *P. caaerulescens* (Hafellner) Kutorka & D. Hawksw. And *P. cratincola* (Rehm) Hafellner.

References

Körber. GW. 1861. Ergänzungen zum Systema lichenum Germaiae. Parerga Lichenol (Breslau): 280.
Hafellner J. 1979. *Karschia* revision einer Sammelattung an der Grenze von lichenisienlen unf nichtlichenisierten Ascomyceten. Nova Hedigia Beih 62: 188.
Kutorga. E and Hawksworth. D.L. 1997. A reassessment of the genera referred to the family *Patellariaceae* (Ascomycota) .Systema Ascomycetum 15: 63-66

Part C

Collecting and study fresh collection of *Patellariaceae* in Thailand.

The objective of this study is comparison fresh specimens from Thailand which have the character similary with *Patellariaceae*

3.1. Methodology.

3.1.1 Sample collection and examination.

Collection of woody litter samples were randomly from Chiang Rai, Phetchaboon, Phayao Province, Thailand. Starting during wet-dry season of 2011-2012. Samples were returned to the laboratory. Each sample was separately incubated in plastic boxes or zip lock plastic bag with paper and sterilize distilled water. The fungi present on the samples were examined. The fungi were identified, recorded, photographed and fully described. Single spore isolation methods were used to isolate the fungi (Ho and Ko, 1996). Cultures were maintained on Sabouraud's dextrose agar (SDA)/Potato dextrose agar (PDA) at 25ºC (Chamyuang, 2010) and deposited in MFLU culture collection. Dry specimens also were deposited in MFLU herbarium.

3.1.2 Molecular phylogenetics DNA extraction

DNA extraction: from cultures were carried out using CTAB lysis buffer and phenol chloroform as outlined by Jeewon *et al*. (2003). DNA samples were checked for purity and integrity by gel electrophoresis.

PCR Amplification: PCR amplification was performed in Thermal controllable Cycler DNA amplification using 3 different genes. PCR profiles were optimized for each gene amplified. Products were purified using the QIAquick DNA purification Kit (QIAGEN, LTD.)

Sequencing: Both strands of the DNA were sequenced in an automated sequencer following manufacturer's protocols.

Alignment: All sequences are being aligned using Clustal X with default settings (Thomson *et al*., 1997). Then they will be manually adjusted in BioEdit.

Phylogenetic analysis: Different DNA analyzing methods, Bayesian, MP, ML and NJ, will be used in Paup (Swofford, 2004). Support values for NJ and MP will be established by bootstrapping with 1000 replicates. The above work has routinely been carried out by Hyde and coworkers (e.g. Jeewon *et al*., 2002, 2003, 2004; Zhao *et al*., 2007).

Analysis of data: ClustalX will be used for sequence alignment and PAUP*4.0b10 for phylogenetic analysis. The above work has routinely been carried out by Hyde and coworkers (Zhao *et al*., 2007).

3.2 Results

3.2.1 Isolated cultures and interesting fungi

The cultures collection from woody litter specimens in this study have been shown in the table3. Some fungi were shown in the plate (Fig 1-6)

Table 3. Some fresh collection list from Thailand and were isolated, culture and sequence

No.	Code	*Fungal Species*	Habitat	MFLUCC& Sequence (MFLU culture collection)
1.	KA100	*Rhystidhysteron rufulum* like	Phetchaboon	12-0011
2.	KA105	*Hysterobrevium sp. like*	Phayao	12-0010
3.	KA110	*Hysterographium* sp. Like	Phetchaboon	12-0012
4	KA112	*Rhystidhysteron rufulum* like	Phayao	12-0013
5.	KA119	*Hysterographium* sp. like	Chiang Rai	Prepare for deposit and send to sequence
6.	KA120	*Karschia lignyota like*	Phayao	Prepare for deposit and send to sequence
7.	KA121	*Rhystidhysteron rufulum* like	Chiang Rai	Prepare for deposit and send to sequence

KA105

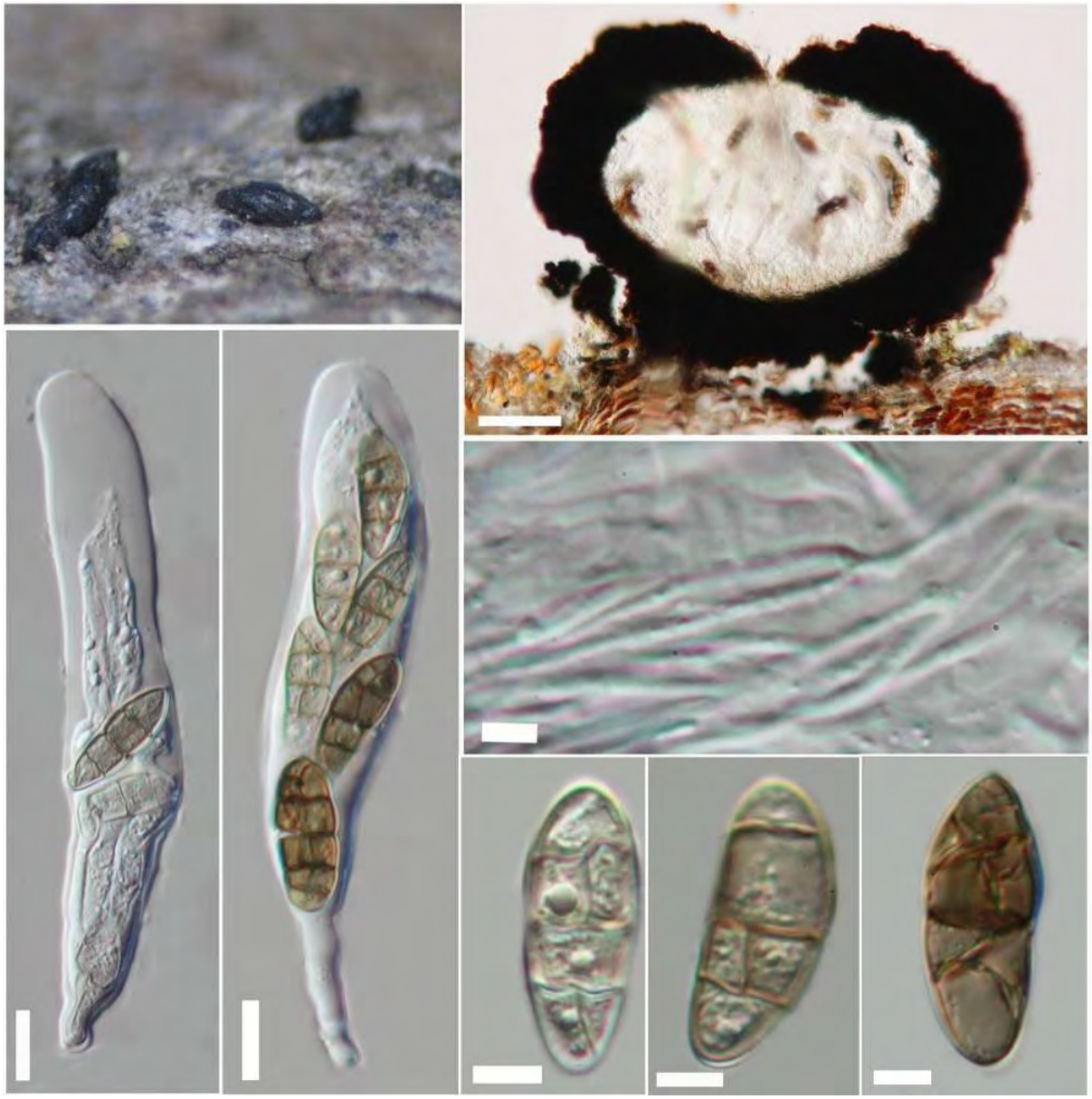

Fig1(a-h). *Hysterobrevium* sp. like KA105 MFLUCC12-0010**, a**. Fruiting body on host surface, **b**. Section of ascoma with slit-slot opening, **c**. Young asci, **d**. Mature asci with spores, **e.** Hamathecium, **f**. Young asco spore, **g-h**. Mature spore in muriform. Scale bar: b= 50 µm; c-d = 15 µm; e-h = 5 µm

KA110

Fig2(a-j). *Hysterographium* sp. like KA110 MFLUCC12-0012**, a**. Superficial of ascomata on dead wood, **b**. Section of ascoma with thick peridium, **c-e.** Young asci with bitunicate, **g.** Mature asci with crowed brown spores, **h**. Young spore, i-j. Mature spore in muriform. Scale bar: b = 50 µm; c-g = 10 µm; h-j = 5 µm.

KA112

Fig3(a-i). ***Rhystidhysteron rufulum*** **like KA112** MFLUCC12-0013**, a.** Ascomata with cub shape on host surface, **b.** Section of ascoma, **c.** Perdium with textular angularis, **d.** Individual cells elongated and branched toward the apices of hamathecium, **e.** Hamathecium with septate, **f-g,** uniseriate of ascospore, **h.** spore shape, **i.** spore germinated. Scale bar: b=120 µm; c=30 µm; d-e, h= 5 µm; f-g,i=10 µm;

KA119

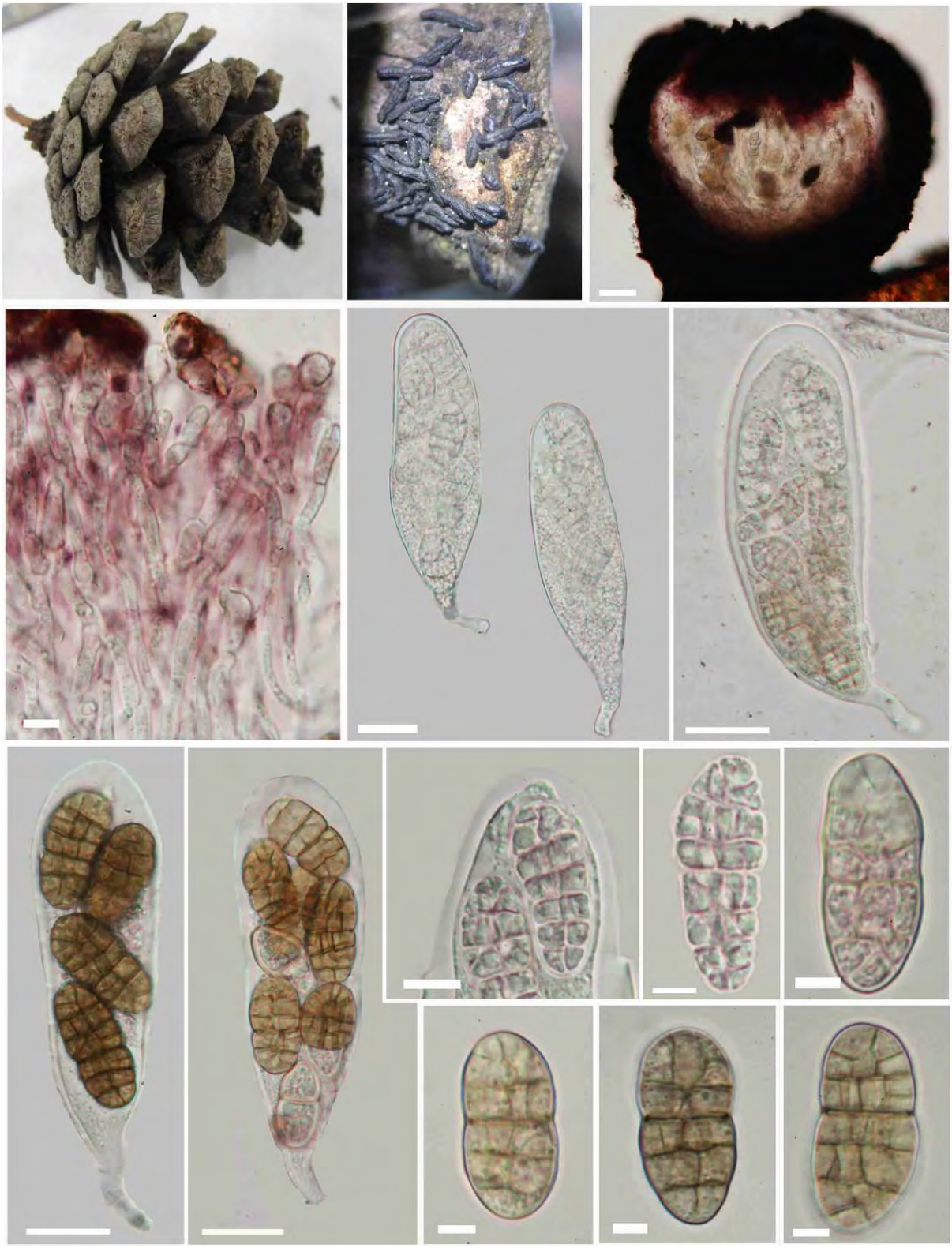

Fig4 (A-N) ***Hysterographium*** **sp. like.** KA119, **A-B** Ascomata on fruit of pine, **C.** Vertical section of ascoma, **D.** Purple hamathecium with septate, **E-F.** Young asci, **G-H.** Bitunicate with 8-spores of mature asci, **I.** Ocular chamber on the tip, **J-N.** Ascospore with muriform shape. Bar line: C=70µm; D, J-N =5µm; E-H=20 µm; I=7 µm.

BRG5280002

Ascomata 504-627 µm high × 475-640 µm diam.($\bar{x}$ = 548×555; n=10), aggregate, grouping, superficial, fusiform to oblong with slit-slot opening, hard, black,. *Peridium* 37-65 µm wide ($\bar{x}$ = 50.11; n=20), one-layered, composed with dark cells of pseudoparenchymatous, (Fig 1C). *Hamathecium* of dense, oblong, 2.7-4.2 µm ($\bar{x}$ = 3.2; n=20), broadly on the top, anastomosing mostly above the asci, septate (Fig 1D). *Asci* 100-120 × 23-34 µm ($\bar{x}$ = 112.5 × 30 µm, n = 10), 8-spored, with bitunicate, obovoid, pedicel, with ocular chamber (Fig 1E-H). *Ascospores* 24.7-34.1 × 11.5-16.2 µm ($\bar{x}$ = 28 × 14. µm, n = 10), biseriate, muriform with broadly on upward to narrowly rounded ends, constricted at middle, light brown (Fig 1J-N).

***Material examined*:** Thailand: Chiang Rai, Doi Mae Sa long; on pine fruit, 20 May 2012, *Sholva* , KA119.

KA120

Fig5 (A-I) *Karschia lignyota* like. KA120 A-B. Apothecia on algae, **C.** Vertical section on superficial ascoma, **D.** Yong asci with 8-spored, **E-F.** Mature asci with bitunicate asci, **G** Hamathecium broadly on the top with septate, **H-I.** ascospore with 1 septate and 2 cells. Bar line: C=30µm; D-F=7µm; **G**=4 µm; **H-I**=5 µm.

Ascomata 60-110 µm high × 180-318 µm diam. ($\bar{x}$=95×252; n=10), scattered, or in small groups of 2-3, apothecium, superficial on algae, folding at the rim, plane at the centre, black, (Fig2 A-B). *Peridium* 17-43 µm wide ($\bar{x}$=31; n=10), one-layered, composed of *textura*

angularis, brown at inner to dark brown at outer. *Hamathecium* of dense, paraphyses 1.7-2.6 µm broad ($\bar{x}$=1.9; n=10), swollen at the top and formed the network above the asci. *Asci* 48-77 × 11-22 µm ($\bar{x}$ = 67 × 16 µm, n = 10), 8-spored, with bitunicate, cylindrical and broadly on the top with ocular chamber, small pedicel, (Fig 2D-F). *Ascospores* 17.5-22 × 6.7-8.8 µm ($\bar{x}$ = 19 × 7.5 µm, n = 20), overlapping biseriate, obovoid to fusiform, broadly at the middle cell to narrowly at the ends, brown, two-celled, constrict at the septum (Fig 1H-I).

Material examined: Thailand, Phayao, Chun, Khoo Phang Lang temple; on algae from dead twing, 30 October 2011, S.Yacharoen

3.2.2 Molecular phylogenetics DNA extraction

Sequenced result of KA 110-112 already received and they are in the processing for analyses and doing phylogenetic tree. Their molecular information will be used to comparison with *Patellariaceae* data**.** Culture of of KA 119-122 will be prepared and send to sequence as soon as possible.

Appendix C

Table of new families, genera and species

Table of now families, genera and species

Published paper			
Paper (SCI impact factor)	**New genera**	**New species**	**Family phylogeny revision other taxa in Thailand studied**
Boonmee *et al*., 2012 (IF = 2.031)	-*Halokirschsteiniothelia* S. Boonmee & K.D. Hyde gen. nov.	-*Halokirschsteiniothelia* (Linder) S. Boonmee& K. D. Hyde comb. nov -*Morosphaeria elaterascus*(Shearer) S. Boonmee & K.D. Hyde comb. nov. -*Kirschsteiniothelia lignicola*S. Boonmee & K.D. Hyde sp. nov. -*Kirschsteiniothelia emarceis*S. Boonmee & K.D Hyde sp. nov.	*Kirschsteiniotheliaceae* S. Boonmee & K.D. Hyde fam. nov.
Boonmee *et al*., 2011 (IF = 4.76)	-*Chlamydotubeufia* Boonmee & K.D. Hyde, gen. nov.	-*Tubeufia khunkornensis* Boonmee & K.D. Hyde, sp. nov. -*Acanthostigma chiangmaiense* Boonmee & K.D. Hyde, sp. nov. -*Chlamydotubeufia huaikangplaensis* Boonmee & K.D. Hyde, sp. Nov. -*Chlamydotubeufia khunkornensis* Boonmee & K.D. Hyde, sp. nov. -*Chlamydotubeufia depressispora* (Matsush.) Boonmee &K.D. Hyde, comb nov. -*Chlamydotubeufia chlamydospora* (Shearer) Boonmee & K.D. Hyde, comb nov. -*Thaxteriella inthanonensis* Boonmee & K.D. Hyde, sp. nov.	*Tubeufiaceae*
Chomnunti *et al*., 2012 (IF = 2.031)		-*Ceramothyrium thailandicum*Chomnunti & K.D.Hyde sp. nov. -*Chaetothyrium brischofiacola*Chomnunti & K.D.Hyde sp. nov. -*Phaeosaccardinula ficus*Chomnunti & K.D. Hyde sp. nov.	*Chaetothyriaceae*
Chomnunti *et al.,* 2012 online (IF = 4.769)		-*Trichomerium foliicola* Chomnunti & K.D. Hyde, sp. nov. -*Trichomerium deniqulatum* Chomnunti & K.D. Hyde, sp. nov. -*Trichomerium gloeosporum* Chomnunti & K.D. Hyde, sp. nov.	*Trichomeriaceae* Chomnunti & K.D. Hyde, fam. nov.
Chomnunti *et al*., 2011 (IF = 4.769)		-*Phragmocapnias asiaticus*Chomnunti & KD Hyde, sp.nov. -*Phragmocapnias longicollus*(Matsush.) Chomnunti & KD Hyde, comb. nov. -*Phragmocapnias penzigii*(Woron.) Chomnunti & KD Hyde, comb. nov. -*Phragmocapnias siamenis* Chomnunti & KD Hyde, sp. nov. -*Leptoxyphium cacuminum*Chomnunti & KD Hyde, sp.nov. -*Capnodium coartatum*Chomnunti & KD Hyde, sp. nov.	*Capnodiaceae*.
Dimuthu *et al*., 2012 online (IF = 4.769)		-*Curvularia hawaiiensis*comb (Bugnic) Manamgoda, L. Cai & K.D. Hyde, comb. nov. -*Curvularia ovariicola* (Alcorn) Manamgoda, L. Cai & K.D Hyde, comb. nov. -*Curvularia perotidis* (Alcorn) Manamgoda, L. Cai, K.D. Hyde, comb. nov. -*Curvularia ravenelii* (M.A. Curtis) Manamgoda, L. Cai, K.D. Hyde, comb. nov.	

Table of new families, genera and species (cont)

Paper (SCI impact factor)	New genera	New species	Family phylogeny revision other taxa in Thailand studied
Dimuthu *et al*., 2012 (cont.) online (IF = 4.769)		-*Curvularia tripogonis* (A.S. Patil&V.G.Rao) Manamgoda, L. Cai and K.D. Hyde, comb. nov. -*Curvularia australiensis* (M.B. Ellis) Manamgoda, L.Cai. & K.D. Hyde, comb. nov.	
Dhanushka *et al*., 2012 online (IF = 4.769)		-*Diaporthe amygdale* (Delacr.) Udayanga, Crous & K.D.Hyde, comb. nov. -*Diaporthe castaneae-mollisimae* (S.X, Jiang & H.B. Ma) Udayanga, Crous & K.D. Hyde, comb. nov. -*Diaporthe cotoneastri* (Punith.) Udayanga, Crous & K.D. Hyde, comb. nov. -*Diaporthe cuppatea* (E. Jansen, Lampr. & Crous) Udayanga, Crous & K.D. Hyde, comb. nov. -*Diaporthe phoenicicola* (Traverso & Spessa) Udayanga, Crous & K.D. Hyde, comb. nov. -*Diaporthe sclerotioides* (Kesteren) Udayanga, Crous & K.D. Hyde, comb. nov. -*Diaporthe neoviticola* Udayanga, Crous & K.D. Hyde, nom. nov.	
Liu *et al*., 2011a (IF = 4.769)	-*Fissuroma* J.K. Liu., R. Phookamsak., E.B.G. Jones & K.D. Hyde, gen. nov. -*Neoastrosphaeriella* J.K. Liu., E.B.G. Jones & K.D. Hyde, gen. nov.	-*Fissuroma aggregata*(I. Hino & Katum) R. Phookamsak., J.K. Liu, E.B.G. Jones & K.D. Hyde, comb. nov. -*Fissuroma maculans*(Rehm) J.K. Liu., E.B.G. Jones & K.D. Hyde, comb. nov. -*Neoastrosphaeriella krabiensis*J.K. Liu., E.B.G. Jones & K.D. Hyde., sp. nov.	
Sajeewa *et.al.,* 2012 online (IF = 4.769)		-*Pestalotiopsis asiatica* Maharachchikumbura & K.D. Hyde, sp. nov. -*Pestalotiopsis chinensis* Maharachchikumbura & K.D. Hyde, sp. nov. - *Pestalotiopsis chrysea* Maharachchikumbura & K.D. Hyde, sp. nov. - *Pestalotiopsis clavata* Maharachchikumbura & K.D. Hyde, sp. nov. - *Pestalotiopsis diversiseta* Maharachchikumbura & K.D. Hyde, sp. nov. - *Pestalotiopsis ellipsospora* Maharachchikumbura & K.D. Hyde, sp. nov. - *Pestalotiopis inflexa* Maharachchikumbura & K.D. Hyde, sp. nov. - *Pestalotiopsis intermedia* Maharachchikumbura & K.D. Hyde, sp. nov. - *Pestalotiopis linearis* Maharachchikumbura & K.D. Hyde, sp. nov.	

Table of new families, genera and species (cont)

Paper (SCI impact factor)	New genera	New species	Family phylogeny revision other taxa in Thailand studied
Sajeewa *et.al.,* 2012 (cont.) online (IF = 4.769)		- *Pestalotiopsis rosea* Maharachchikumbura & K.D. Hyde, sp. nov. - *Pestalotiopsis saprophyta* Maharachchikumbura & K.D. Hyde, sp. nov. -*Pestalotiopsis umberspora* Maharachchikumbura & K.D. Hyde, sp. nov. -*Pestalotiopsis unicolor* Maharachchikumbura & K.D. Hyde, sp. nov. -*Pestalotiopsis verruculosa* Maharachchikumbura, & K.D. Hyde, sp. nov.	
Paper inpress (submited paper)			
Dai *et al.,* 2012	-*Bambusicola* D. Q. Dai & K.D. Hyde, gen. nov.	- *Bambusicola massarinia* D.Q. Dai & K.D. Hyde, sp. nov. - *Bambusicola bambusae* D.Q. Dai & K.D. Hyde, sp. nov. - *Bambusicola irregulispora* D.Q. Dai & K.D. Hyde, sp. nov. - *Bambusicola splendida* D.Q. Dai & K.D. Hyde, sp. nov.	
Paper inpress (submited paper)			
Ariyawangsa et.al., 2012 (In press)	-*Deniquelata* Ariyawansa & K.D. Hyde, gen. nov.	-*Deniquelata barringtonia* Ariyawansa & K.D. Hyde sp. nov.	
Liu *et al*., 2012 (In press.)	-*Botryobambusa* R. Phookamsak., J.K. Liu & K.D. Hyde, gen. nov. -*Cophinforma* Doilom., J.K. Liu & K.D. Hyde, gen. nov.	-*Auerswaldia dothiorella* D.Q. Dai., J.K. Liu & K.D. Hyde, sp. nov. -*Auerswaldia lignicola* Ariyawansa, J.K. Liu & K.D. Hyde, sp. nov -*Botryobambusa fusicoccum* R. Phookamsak., J.K. Liu & K.D. Hyde -*Botryosphaeria fusispora* Boonmee., J.K. Liu & K.D. Hyde sp. nov. -*Cophinforma eucalyptus* Doilom., J.K. Liu & K.D. Hyde. -*Phaeobotryosphaeria eucalyptus* Doilom., J.K. Liu & K.D. Hyde, sp. nov.	
Zhang *et.al*., 2012 (In press)		-*Lindgomyces griseosporus* Y. Zhang ter, J. Fourn. & K.D. Hyde, sp. nov.	
Paper in prep.			
Monkai *et al*., 2012 (In prep.)		-*Fusicoccum* sp. nov. -*Ascomycete* sp.1 nov. -*Ascomycete* sp.2 nov. -*Sporodochium* sp.1 nov. -*Sporodochium* sp.2 nov.	
Yacharoen *et al*., 2012 (In prep.)		-*Monodictys appendiculata* S. Yacharoen, McKenzie and K.D. Hyde, sp. nov.	

Table of now families, genera and species (cont)

Paper (SCI impact factor)	New genera	New species	Family phylogeny revision other taxa in Thailand studied
Paper inprep.(cont)			
Yacharoen *et al*., 2012 (In prep.)		-*Thyridaria* sp. nov.	
Zhang *et.al*., 2012 (In prep.)	*Aquasubmersa* K.D. Hyde & H. Zhang ter, gen. nov.	-*Acrocalymma aquatica* H. Zhang ter & K.D. Hyde, sp. nov. -*Aquasubmersa mircensis* H. Zhang ter & K.D. Hyde, sp. nov.	
Zhang *et.al*., 2012 (In prep.)		-*Misturatosphaeria mariae* Yin. Zhang, J. Fourn. & K.D. Hyde, sp. nov.	
Total = 19 papers	**Total = 9 new genera**	**Total = 75 new species**	**Total = 2 new families**

Appendix D

Table of total publications
Published/accepted/in press(submitted)/in prep.

Table of publications

No.	Publication	SCI impact factor
Published		
1	Boonmee S, Ko Ko TW, Chukeatirote E, Hyde KD, Chen H, Cai L, McKenzie EHC, Jones EBG, Kodsueb R, Bahkali AH (2012). Two new *Kirschsteiniothelia* species with a *Dendryphiopsis* anamorph cluster in *Kirschsteiniotheliaceae* fam. nov. Mycologia 104(3): 698-714	IF = 2.031
2.	Boonmee S, Zhang Y, Chomnunti P, Chukeatirote E, Tsui CKM, Bahkali AH, Hyde KD (2011). Revision of lignicolous *Tubeufiaceae* based on morphological reexamination and phylogenetic analysis. Fungal Diversity 51(1): 63-102	IF = 4.76
3.	Chomnunti P, Ko Ko TW, Chukeatirote E, Hyde KD, Cai L, Jones EBG, Kodsueb R, Bahkali AH, Chen H (2012). Phylogeny of *Chaetothyriaceae* in northern Thailand including three new species. Mycologia 104(2): 382-395	IF = 2.031
4.	Chomnunti P., Bhat D.J., Jones E.B.G., Chukeatirote E, Bahkali A.H. and Hyde K.D. (2012). Trichomeriaceae, a new sooty mould family of *Chaetothyriales*. Fungal Diversity(online).	IF = 4.769
5.	Chomnunti P, Schoch CL, Aguirre-Hudson B, Ko Ko TW, Hongsanan S, Jones EBG, Kodsueb R, Phookamsak R., Chukeatirote E, Bahkali AH, Hyde KD (2011). *Capnodiaceae*. Fungal Diversity 51(1):103-134.	IF = 4.769
6.	Dimuthu S. M., Cai L., McKenzie E.H.C., Crous P.W., Madrid H., Chukeatirote E., Shivas R., Tan Y.P. and Hyde K.D. (2012) A Phylogenetic and taxonomic re-evaluation of the *Bipolaris-Cochliobolus-Curvularia* complex. Fungal Diversity (online)	IF = 4.769
7.	Dhanushka U., Liu X., Crous P.W., McKenzie E.H.C., Chukeatirote E. and Hyde K.D. (2012) A multi-locus phylogenetic evaluation of *Diaporthe* (Phomopsis). Fungal Diversity (online)	IF = 4.769
8.	Liu J.K., Chukeatirote E, Phookamsak R, Jones E.B.G., Zhang Y, Ko Ko TW, Hu HL, Boonmee S, Doilom M, Chukeatirote E, Bahkali A.H., Wang Y, Hyde K.D. (2011). *Astrosphaeriella* is polyphyletic, with species in *Fissuroma* gen. nov., and *Neoastrosphaeriella* gen. nov. Fungal Diversity 51(1):135-154	IF = 4.769
9.	Sajeewa S.N.M., Guo L.D., Cai L., Chukeatirote E., Wu W.P., Sun X., Crous P.W., Bhat J., McKenzie E.H.C., Bahkali A.H. and Hyde K.D. (2012). A multi-locus backbone tree for Pestalotiopsis, with a polyphasic characterization of 14 new species. Fungal Diversity (online).	IF = 4.769
Accepted paper		
10.	Dai D.D., Bhat D.J., Liu J.K., ZhaoR. & Kevin D. H. (2012). *Bambusicola*, a new genus from bamboo with asexual and sexual morphs. Cryptogamie mycologie (accepted, waiting proofs).	-
Paper in press(submitted)		
11.	Ariyawangsa H.A., Maharachchikumbura S.S.N., Karunarathne S.C., Chukeatirote E., Bahkali A.H., Kang J.C., Bhat D.J. and Hyde K.D. (2012). *Deniquelata barringtoniae* gen. et sp. nov., associated with leaf spots of Barringtonia asiatica. Mycologia (in press).	-
12.	Liu J.K., Phookamsak R., Doilom M., Wikee S., Li Y.M., Ariyawansha H., Boonmee S., Chomnunti P., Dai D.Q., Hongsanan S., Romero A.I., Monkai J., Bhat J., Chukeatirote E., Jones E.B.G., Ko Ko T.W., Zhou Y.C., Zhuang W.Y. and Hyde K.D. (2012). Towards a natural classification of *Botryosphaeriales*. Fungal Diversity. (in press).	-
13.	Zhang Y, Fournier J, Bakhali AH, Hyde KD.(2012) Lindgomyces in Europe New species and first records of the aquatic ascomycetous genus Lindgomyces from Europe. (in press)	

Table of publications

No.	Publication	SCI impact factor
Paper in prep.		
14.	Monkai J, Boonmee S, Chomnunti P., Chukeatirote E., Crous P.W. and Hyde K.D. (2012). *Planistromellaceae* (in prep).	-
15.	Monkai J, Promputtha I, Kodsueb R, Chukeatirote E, McKenzie, EHC, Bahkali, AH and Hyde, KD (2012). Diversity of saprobic fungi on dead leaves from *Magnolia liliifera* and *Cinnamomum iners* in northern Thailand. Mycoscience (in prep).	-
16.	Monkai J, Chukeatirote E, McKenzie, EHC, Bahkali, AH and Hyde, KD (2013).New saprobic fungi in northern Thailand. (in prep).	-
17.	Yacharoen S, Promputtha I, Kodsueb R, Chukeatirote E, McKenzie EHC, Hyde KD (2012). *Monodictys appendiculata* sp. nov. on decaying wood from northern Thailand. Mycotaxon (in prep).	-
18.	Yacharoen S, Chomnunti P., Chukeatirote E and Hyde KD (2012). Revision of *Patellariaceae* in Thailand (in prep).	-
19.	Yacharoen S, Chukeatirote E, McKenzie EHC, Bhat D.J. and Hyde KD (2013). New and interest saprobic fungi on terrestrial wood from northern Thailand (in prep).	-
20.	Zhang H., Hyde K.D., McKenzie E.H.C., Bahkali A.H. and Zhou D. (2012). Sequence data reveals phylogenetic affinities of *Acrocalymma aquatica* sp. nov, *Aquasubmersa mircensis* gen. et sp. nov. and *Clohesyomyces aquaticus* (freshwater coelomycetes) (in prep).	-
21.	Zhang Y., Fournier J., Bahkali A.H. and Hyde K.D. (2012). *Misturatosphaeria mariae* sp. nov. from France, a first record of *Misturatosphaeri*a in Europe (in prep).	-

Mycologia, 104(3), 2012, pp. 000–000. DOI: 10.3852/11-089

Two new *Kirschsteiniothelia* species with *Dendryphiopsis* anamorphs cluster in *Kirschsteiniotheliaceae* fam. nov.

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Abstract: Two new *Kirschsteiniothelia* species are proposed in this study; both were collected on decaying wood from Chiang Mai and Chiang Rai provinces in northern Thailand. The taxa were isolated and the morphological characters are described and illustrated. ITS, LSU and SSU combined sequence analysis showed taxa of *Kirschsteiniothelia* separating into three lineages: (i) *K. elaterascus* grouped within Morosphaeriaceae (Pleosporales); (ii) *K. maritima* clustered with *Mytilinidion* spp. as a sister group in the Mytilinidiaceae clade; and (iii) the two new *Kirschsteiniothelia* species, which produce *Dendryphiopsis* anamorphs in culture, clustered with *K. aethiops* (the generic type) and the anamorph *D. atra*. The new family Kirschsteiniotheliaceae is introduced to accommodate taxa grouping with *K. aethiops*. *K. elaterascus* is transferred to *Monosphaeria* (Morosphaeriaceae) and a new genus *Halokirschsteiniothelia* is introduced to accommodate *K. maritima* (Mytilinidiaceae).

Key words: Dothideomycetes, Kirschsteiniotheliaceae, new species, phylogeny

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Introduction

Kirschsteiniothelia was introduced by Hawksworth (1985a) and is represented by the type *K. aethiops* based on *Sphaeria aethiops* Berk. & M.A. Curtis. The genus is characterized by superficial to semi-immersed, hemispherical or subglobose, dark brown to black ascomata, cylindrical-clavate asci that develop among numerous pseudoparaphyses, and mostly one-septate (in some species two-septate), ellipsoidal, dark-brown ascospores; there are presently 18 species recorded in *Index Fungorum* and seven estimated species in Kirk et al. (2008).

Kirschsteiniothelia D. Hawksw. is a genus of the Dothideomycetes (Hawksworth 1985a), although its ordinal and familial placements are uncertain and it is currently classified as Dothideomycetes incertae sedis in *Index Fungorum* and in Lumbsch and Huhndorf (2010). In MycoBank (Crous et al. 2004, Robert et al. 2005) this genus is placed in the Pleosporaceae, while its known hyphomycete anamorphs are referred to the Pleomassariaceae (Hyde et al. 2011). Thus placement of the genus is uncertain. Schoch et al. (2006) analyzed molecular data for *K. aethiops*, which did not cluster close to Pleosporaceae, and it was suggested that this genus should be transferred to a separate family. In a recent molecular phylogenetic analysis of the Dothideomycetes *K. elaterascus* Shearer clustered in the same clade as *Monosphaeria* (Morosphaeriaceae) while *K. maritima* (Linder) D. Hawksw. clustered in the Mytilinidiaceae clade, as a sister group of *Mytilinidion* (Schoch et al. 2009, Suetrong et al. 2009).

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Revision of lignicolous *Tubeufiaceae* based on morphological reexamination and phylogenetic analysis

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Received: 18 October 2011 / Accepted: 23 October 2011 / Published online: 17 November 2011

Abstract In this paper we revisit the family *Tubeufiaceae* with notes on genera that we have re-examined where possible. Generic type specimens of *Acanthophiobolus*, *Kamalomyces*, *Podonectria*, *Thaxteriella* and *Thaxteriellopsis* were re-examined, described and illustrated and shown to belong to *Tubeufiaceae*. Notes are provided on *Acanthostigma*, *Chaetosphaerulina*, *Thaxteriina* and *Tubeufia*, which are retained in *Tubeufiaceae*; however, we were unable to locate the types of these genera during the time frame of this study. *Allonecte* is excluded from the *Tubeufiaceae*, as the ascospores are fusiform-ellipsoidal, grey-brown and 1-septate and the asci are cylindrical, all of which are features more typical of *Hazslinszkyomyces*, where it is transferred. *Byssocallis* has yellow to orange ascomata and clavate ascospores which is atypical of *Tubeufiaceae*. Thus its taxonomic status needs to be reevaluated. *Lentoudinaceopsis* has an endophytic habit, cylindro-clavate asci and two-celled ascospores more typical of *Hazslinszkyomyces*, where it is transferred. *Taphrophila* has small ascomata, a thin peridium, branching setae around the apex of the ascomata, clavate to saccate asci and lacks pseudoparaphyses. These are features atypical of the *Tubeufiaceae*, and *Taphrophila* should be placed in the Dothideomycetes *incertae sedis*. Twelve new collections of *Tubeufiaceae* from Thailand were isolated, and their DNA was extracted. The sequence data of LSU, SSU and ITS rDNA were amplified and analyzed using parsimony and likelihood methods. The results of phylogenetic analysis was used to establish the inter-generic relationships in *Tubeufiaceae*. *Thaxteriellopsis lignicola*, epitypified in this investigation, is a sister taxon in the family *Tubeufiaceae* based on phylogenetic analysis of rRNA sequence data. *Chlamydotubeufia* is introduced as a new genus based on the production of dictyochlamydosporous anamorphs, including two new species. Three new species, one each in *Acanthostigma*, *Tubeufia* and *Thaxteriella* are also described and illustrated. The phylogenetic placement of these genera is also discussed.

Keywords Anamorph · *Aquaphila* · Dothideomycetes · Helicosporous · Molecular phylogeny · Woody litter fungi

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Phylogeny of Chaetothyriaceae in northern Thailand including three new species

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Abstract: In a recent study unusual taxa of epiphyllous ascomycota belonging to Chaetothyriaceae (Eurotiomycetes) were collected in northern Thailand. This family is poorly understood due to morphological confusion and lack of phylogenetic studies. This paper deals with three new species, *Ceramothyrium thailandicum*, *Chaetothyrium brischofiicola* and *Phaeoacanthinula ficus*, which are fully described and illustrated. A DNA sequence analyses of LSU and ITS rDNA genes shows that the new species cluster in the Chaetothyriaceae. This paper adds six sequences for Chaetothyriaceae to GenBank, providing much needed data for the family.

Key words: *Ceramothyrium*, Chaetothyriaceae, *Chaetothyrium*, LSU, *Phaeoacanthinula*, phylogeny

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Introduction

The Chaetothyriaceae are typical of capnodiaceous Dothideomycetes because they form on the surface of leaves and resemble typical sooty molds (Batista and Ciferri 1962). Species of Chaetothyriaceae are mostly epiphytes, colonizing the surface of living leaves with mycelium appressed to the host cuticle without penetrating host tissues (Batista and Ciferri 1962, von Arx and Müller 1975). Ascomata are surrounded by a thin pellicle of superficial mycelium forming black sooty mold-like areas on leaves that are easily detached from the cuticle (Batista and Ciferri 1962). However the ecology of many species of Chaetothyriaceae is poorly studied and it is unclear whether they are saprotrophic or biotrophic (Barr 1987). Members of Chaetothyriaceae often are confused with capnodiaceous sooty molds due to their similar morphology and habitat preferences, however these fungi are never associated with insects such as several Capnodiaceae (Hansford 1946). Sooty molds are a general taxonomic term for capnodiaceous and/or chaetothyriaceous fungi; common genera from both these groups often are found growing together in sooty mold complexes in plant exudates or the sugary honeydew secreted by insects, for example *Aithaloderma* (*Leptoxyphium*), *Aureobasidium*, *Capnodium*, *Cladosporium*, *Microxyphium*, *Rhodoxyphium*, *Scorias* and *Trichomerium* (*Triposporium*) (Thaung 2006).

Studies on Chaetothyriaceae were conducted mainly by Hansford (1946), Batista and Ciferri (1962), von Arx and Müller (1975) and Hughes (1976), and few studies have been undertaken since. Members of Chaetothyriaceae are primarily tropical species characterized by dark mycelium forming as a loose net of hyphae over the substrate, and they produce ascomata beneath a mycelial pellicle with or without setae (Batista and Ciferri 1962, Hughes 1976, Pereira et al. 2009). The family is poorly circumscribed and most work comprised brief descriptions with line drawings (e.g. Hansford 1946, Batista and Ciferri 1962). The arrangement of genera often seem subjective because individual authors emphasize certain characters, such as spore septation, presence of ascomata setae and mycelium color (Batista and Ciferri 1962, Hughes 1976). Batista and Ciferri (1962) considered the family Chaetothyriaceae to be the type family in order Chaetothyriales. This group share a number of centrum characters with members of the Dothideomycetes, such as the presence of bitunicate

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Trichomeriaceae, a new sooty mould family of Chaetothyriales

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Received: 17 July 2012 / Accepted: 1 August 2012

Abstract *Trichomerium* is a genus of foliar epiphytes with the appearance of sooty moulds, mostly occurring on the surface of living leaves and apparently gaining their nutrients from insect exudates. Species have ascostromata with setae and develop on a loosely interwoven mycelial mass of dark brown hyphae, while asci have a bitunicate appearance with hyaline ascospores. In this study, we made 16 collections of *Trichomerium* from Thailand. All were isolated, and the LSU and ITS rDNA gene regions sequenced. Phylogenetic analysis indicated that the *Trichomerium* species form a monophyletic clade within *Chaetothyriales* and warrant the introduction of a new family *Trichomeriaceae*. Bootstrap support for the *Chaetothyriales* is 100 % and clearly separates *Trichomeriaceae* from *Capnodiales* which are morphologically very similar. A detailed account of *Trichomerium* is provided and we describe and illustrate three new species based on morphological and molecular data. We propose that *T. foliicola* is adopted as the generic type of *Trichomerium* because it has been impossible to obtain the holotype specimen of *T. confricola* and also no molecular data exists in worldwide databases for this species or genus.

Keywords Foliar epiphytes · Phylogeny · Sooty moulds · *Trichomerium*

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Published online: 26 August 2012

Introduction

The taxonomy of genera of foliar epiphytes is poorly known as they have not been well-studied. No molecular data is available for most genera and therefore an understanding of the higher level classification of these fungi is rather inadequate. We have, therefore, initiated a research program to collect and study these important taxa using morphology and phylogeny. Our initial study (Chomnunti et al. 2012) resulted in the transfer of the genus *Trichomerium*, previously placed in *Capnodiaceae* to *Chaetothyriaceae* in *Chaetothyriales*. We have also provided an account of *Microthyriaceae* (Wu et al. 2011) and are presently studying other genera of foliar epiphytes. Examples of foliar epiphyte genera with a sooty mold-like appearance are *Aithaloderma*, *Capnodium*, *Phragmocapnias* and *Scorias*. Chomnunti et al. (2011) gave an account of the genera in *Capnodiaceae*, while Chomnunti et al. (2012) dealt with species of *Chaetothyriaceae*. The genus *Trichomerium* was placed in *Chaetothyriaceae* but no further data was provided (Chomnunti et al. 2011). Hughes and Seifert (2012) provided notes on the taxonomy and nomenclature of sooty mould names, but further work is required to resolve their interrelationship, especially at the molecular level.

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Capnodiaceae

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Abstract In this paper we revisit the *Capnodiaceae* with notes on selected genera. Type specimens of the ascomycetous genera *Aithaloderma*, *Anopeltis*, *Callebaea*, *Capnodaria*, *Echinothecium*, *Phragmocapnias* and *Scorias* were re-examined, described and illustrated. *Leptoxyphium* is anamorphic *Capnodiaceae* and *Polychaeton* is a legitimate and earlier name for *Capnodium*, but in order to maintain nomenclatural stability we propose that the teleomorphic name should be considered for the approved lists of names currently in preparation for fungi. Notes are provided on the ascomycetous genus *Scoriadopsis*. However, we were unable to locate the type of this genus during the time frame of this study. The ascomycetous genera *Aithaloderma*, *Ceramoclasteropsis*, *Hyaloscoleconroma* and *Trichomerium* are excluded from *Capnodiaceae* on the basis of having ascostromata and trans-septate hyaline ascospores and should be accommodated in *Chaetothyriaceae*. *Callebaea* is excluded as the ascomata are thyriothecia and the genus is placed in *Micropeltidaceae*. *Echinothecium* is excluded as synonym of *Sphaerellothecium* and is transferred to *Mycosphaerellaceae*. The type specimen of *Capniophaeum* is lost and this should be considered as a doubtful genus. The coelomycetous *Microxiphium* is polyphyletic, while the status of *Fumiglobus*, *Polychaetella* and *Tripospermum* is unclear. Fourteen new collections of sooty moulds made in Thailand were isolated and

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Fungal Diversity
DOI 10.1007/s13225-012-0190-9

A multi-locus phylogenetic evaluation of *Diaporthe* (*Phomopsis*)

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Received: 12 June 2012 / Accepted: 12 July 2012

Abstract The genus *Diaporthe* (*Phomopsis*) includes important plant pathogenic fungi with wide host ranges and geographic distributions. In the present study, phylogenetic species recognition in *Diaporthe* is re-evaluated using a multi-locus phylogeny based on a combined data matrix of rDNA ITS, and partial sequences from the translation elongation factor 1–α (EF 1-α), β-tubulin (TUB) and calmodulin (CAL) molecular markers. DNA sequences of available ex-type cultures have been included, providing a multi-locus backbone tree for future studies on *Diaporthe*. Four utilizable loci were analyzed individually and in combination, and ITS, EF 1-α and multi-locus phylogenetic trees are presented. The phylogenetic tree inferred by combined analysis of four loci provided the best resolution for species as compared to single gene analysis. Notes are provided for nine species previously known in *Phomopsis* that are transferred to *Diaporthe* in the present study. The unraveling of cryptic species complexes of *Diaporthe* based on Genealogical Concordance Phylogenetic Species Recognition (GCPSR) is emphasized.

Keywords Ex-type culture · Host diversity · Mating types · Molecular systematics · New combination · Phytopathogen · Species recognition · Taxonomy

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Introduction

The genus *Diaporthe* Nitschke (anamorph *Phomopsis* (Sacc.) Bubák) includes phytopathologically important taxa with wide host ranges and geographic distributions (Uecker 1988; Crous and Groenewald 2005; Rossman et al. 2007). *Diaporthe* species have also been reported as endophytes in healthy leaves and stems, saprobes on decaying wood and leaf litter, and even parasites in humans and other mammals (van Warmelo et al. 1970; Santos et al. 1999; Garcia-Reyne et al. 2011; Iriart et al. 2011; Botella & Diez 2011; Sun et al. 2011; Rocha et al. 2011). The host specificity and geographic distributions of most phytopathogenic species of *Diaporthe* are unknown, hindering the international exchange of agricultural commodities (Udayanga et al. 2011; Cowley et al. 2012; Sun et al. 2012). Studies on phytopathogenic *Diaporthe* species are therefore particularly important to plant pathologists working on wide range of crop diseases (e.g. grapes, sunflower, soybeans and various diseases associated with fruit and ornamental trees). DNA sequence comparisons have made it possible to reliably connect sexual and asexual states of the species of pleomorphic genus *Diaporthe*. Being the older name, *Diaporthe* has priority over *Phomopsis* and should be the generic name adopted for these taxa in future studies (Santos et al. 2010,

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A phylogenetic and taxonomic re-evaluation of the *Bipolaris* - *Cochliobolus* - *Curvularia* Complex

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Abstract Three genera, *Cochliobolus*, *Bipolaris* and *Curvularia* form a complex that contains many plant pathogens, mostly on grasses (*Poaceae*) with a worldwide distribution. The taxonomy of this complex is confusing as frequent nomenclatural changes and refinements have occurred. There is no clear morphological boundary between the asexual genera *Bipolaris* and *Curvularia*, and some species show intermediate morphology. We investigated this complex based on a set of ex-type cultures and collections from northern Thailand. Combined gene analysis of rDNA ITS (internal transcribed spacer), GPDH (glyceraldehyde 3-phosphate dehydrogenase), LSU (large subunit) and EF1-α (translation elongation factor 1-α) shows that this generic complex divides into two groups. *Bipolaris* and *Cochliobolus* species clustered in Group 1 along with their type species, whereas *Curvularia* species (including species named as *Bipolaris*, *Cochliobolus* and *Curvularia*) clustered in Group 2, with its generic type. The nomenclatural conflict in this complex is resolved giving priority to the more commonly used established generic names *Bipolaris* and *Curvularia*. Modern descriptions of the genera *Bipolaris* and *Curvularia* are provided and species resolved in this study are transferred to one of these genera based on their phylogeny.

Keywords Anamorph · Generic complex · Neotype · Nomenclature · Pathogens · *Pseudocochliobolus*

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Introduction

Species of *Cochliobolus* Drechsler (1934), with asexual states in *Bipolaris* Shoemaker (1959) and *Curvularia* Boedijn (1933), are important plant pathogens associated with over 60 host genera (Sivanesan 1987; Manamgoda et al. 2011; Agrios 2005). A few species in these genera are occasionally involved in opportunistic infections of vertebrates (Rinaldi et al. 1987; Dasgupta et al. 2005; Hoog et al. 2005). Accurate identification and precise naming of species are crucial since the name is the key to accessing all accumulated knowledge (Cai et al. 2009, 2011; Hyde et al. 2010; Hawksworth et al. 2011; Ko Ko et al. 2011; Udayanga et al. 2011). Frequent name changes as a result of refinements to the taxonomy in *Bipolaris*, *Curvularia* and *Cochliobolus* have caused confusion

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Astrosphaeriella is polyphyletic, with species in *Fissuroma* gen. nov., and *Neoastrosphaeriella* gen. nov.

Jian-Kui Liu · Rungtiwa Phookamsak · E. B. Gareth Jones · Ying Zhang · Thida W. Ko-Ko · Hong-Li Hu · Saranyaphat Boonmee · Mingkhuan Doilom · Ekachai Chukeatirote · Ali H. Bahkali · Yong Wang · Kevin D. Hyde

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Abstract Collections of fungi from bamboo and palm plants in Thailand resulted in the identification of several species of *Astrosphaeriella*, including the type species *A. fusispora*, which is a synonym of *A. stellata*. Species of *Astrosphaeriella* have been previously circumscribed on the basis of morphology and, to a lesser extent, on host affiliation. In order to obtain a phylogenetic understanding of the genus, eleven strains of *Astrosphaeriella sensu lato* were sequenced in this study. Molecular analyses based on a combined dataset of 18S and 28S nrDNA sequences were carried out to infer the phylogenetic placement of these strains in the *Pleosporales*. The phylogenetic analyses showed that *Astrosphaeriella* is polyphyletic, with *Astrosphaeriella* species clustering in four clades, two clades, including species with slit-like ostioles, clustered in *Aigialaceae*; the clade that includes the generic type clustered together with *Delitschia*; and *A. Africana*, which has striate ascospores, deviated from these three clades and had a basal position in the *Pleosporales*. A new combination in *Fissuroma* gen. nov. and new genus *Neoastrosphaeriella* are introduced in *Aigialaceae* to include the species with slit-like ascomata.

Keywords *Aigialaceae* · Phylogeny · *Pleosporales* · Taxonomy · Type study

Electronic supplementary material The online version of this article (doi:10.1007/s13225-011-0142-9) contains supplementary material, which is available to authorized users.

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A multi-locus backbone tree for *Pestalotiopsis*, with a polyphasic characterization of 14 new species

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Abstract *Pestalotiopsis* is a taxonomically confused, pathogenic and chemically creative genus requiring a critical re-examination using a multi-gene phylogeny based on ex-type and ex-epitype cultures. In this study 40 isolates of *Pestalotiopsis*, comprised of 28 strains collected from living and dead plant material of various host plants from China were studied by means of morphology and analysis of ITS, β-tubulin and *tef1* gene sequence data. Based on molecular and morphological data we describe 14 new species (*Pestalotiopsis asiatica*, *P. chinensis*, *P. chrysea*, *P. clavata*, *P. diversiseta*, *P. ellipsospora*, *P. inflexa*, *P. intermedia*, *P. linearis*, *P. rosea*, *P. saprophytica*, *P. umbrispora*, *P. unicolor* and *P. vaccinicola*) and three species are epitypified (*P. adusta*, *P. clavispora* and *P. foedans*). Of the 10 gene regions (ACT, β-tubulin, CAL, GPDH, GS, ITS, LSU, RPB 1, SSU and *tef1*) utilized to resolve cryptic *Pestalotiopsis* species, ITS, β-tubulin and *tef1* proved to be the better markers. The other gene regions were less useful due to poor success in PCR amplification and/or in their ability to resolve species boundaries. As a single gene *tef1* met the requirements for an ideal candidate and functions well for species delimitation due to its better species resolution and PCR success. Although β-tubulin showed fairly good differences among species, a combination of ITS, β-tubulin and *tef1* gene data gave the best resolution as compared to single gene analysis. This work provides a backbone tree for 22 ex-type/epitypified species of *Pestalotiopsis* and can be used in future studies of the genus.

Keywords β-tubulin · Epitype · ITS · Phylogeny · Saprobe · *tef1*

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Appendix E

Poster and Oral presentation

The posters was presented at the International Symposium on "Fungal Biodiversity and Resources" at Wangcam Hotel, Chiag Rai province" 11-13 November 2010

BRG5280002

Biodiversity of saprobic fungi on woody litter of northern Thailand and screening for some insecticidal activity

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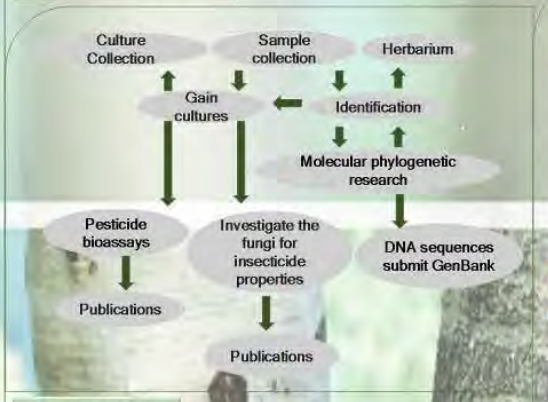
Abstract

Woody litter fungi are one of the major groups that represent an essential component of biodiversity. They are not important only as decomposers in terrestrial ecosystems but have ability to synthesize a diverse range of metabolites which often possess potential biological activity that may inhibit insect's growth. The purpose of this project is to study the diversity of saprobic fungi on woody litter in northern Thailand and screen the fungi for insecticide production. Decaying woody litter of various tree species were selected from Doi Suthep-Pui and Doi Tung in the dry and wet seasons of 2010. A moist chamber method was used for incubation. A total of 69 taxa were identified by morphological characters and comprised 15 ascomycetes, 54 anamorphic taxa (14 coelomycetes and 34 hyphomycetes) and 2 myxomycetes, 2 discomycetes and 2 lichens. Three taxa are probably new to science and will be published following molecular analysis. Thirty-two isolates have been investigated and these will be selected for molecular phylogenetic analysis. All isolates will be assayed for insecticidal activities and selected strains will be used to find out the structure of bioactive secondary metabolites.

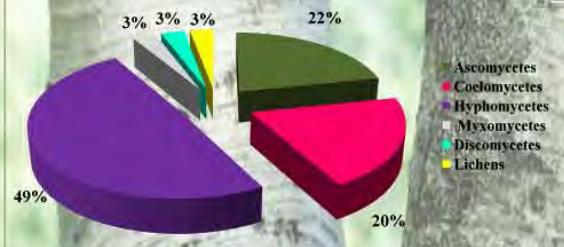
Introduction

There have been few studies of fungi in poorly studied habitats in Thailand and this includes studies on decaying wood in forests of northern Thailand. This research will help to address the lack of knowledge the microfungi on dead wood. We are investigating the insecticidal properties of these fungi using bioassays.

Materials and Methods



Group of fungi



Plates of some interesting fungi

Conclusion

We are in process of identifying fungal species. Now, 69 cultures have been selected for further molecular phylogenetic analysis. New and interesting fungi will be described and will be published. Furthermore, insecticidal bioassays will be developed and tested on all cultures.

Acknowledgements

This project is supported by Thailand Research Fund (BRG 5280002) which is gratefully acknowledged. The authors would like to sincerely thank The School of Science, Scientific and Technological Instruments Center, Mae Fah Luang University for continued support. We also thank Prof. Dr. Lei Cai and Dr. Sunita Chamyuang for assistance

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Kodsueb, R., McKenzie, E.H.C., Lumyong, S. and Hyde, K.D. (2008). Diversity of saprobic fungi on *Magnoliaceae*. Fungal Diversity 30: 37-53.
Promputtha, I., Lumyong, S., Lumyong, P., McKenzie, E.H.C. and Hyde, K.D. (2004). Fungal saprobes

The poster was presented at the "10th Conference and meeting between new researcher and senior researcher" at Holiday Inn Resort Regent Beach Cha Am, Phetchabury Province. 14-16 October 2010

Diversity of fungi on leaf and woody litter from some selected trees in northern Thailand and screening for insecticide production

Jutamart Monkai[1] , Supalak Yacharone[1] , Itthayakorn Promputtha[2] , Rampai Kodsueb[3] , Ekachai Chukeatirote[1] , Eric H. C. McKenzie[4] and Kevin D. Hyde[1,5,*]

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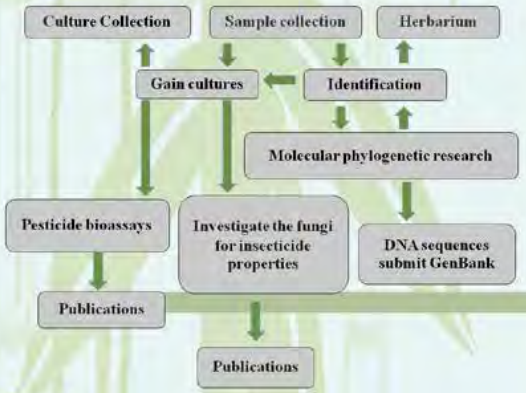
Abstract

Leaf and woody litter fungi are important decomposers in terrestrial ecosystems and these fungi can produce secondary metabolites that may inhibit the growth of insects. This research aims to study the diversity of saprobic fungi on leaves and woody litter in northern Thailand and screen the fungi for insecticide production. Decaying leaves and woody litter of various tree species were selected from Doi Suthep-Pui National Park and Doi Tung National Park in the dry and wet seasons of 2010. A moist chamber method was used for incubation. A total of 94 taxa were identified by morphological character and comprises 18 ascomycetes, 70 anamorphic taxa (26 coelomycetes and 44 hyphomycetes) and 2 myxomycetes. Five taxa are probably new to science and will be published following molecular analysis. Seventy isolates have been obtained and these will be selected for molecular phylogenetic analysis and all isolates will be assayed for insecticidal activities.

Introduction

The most species phylum of fungi in Thailand is the Ascomycota and yet they are relatively poorly studied. This research will help to address the lack of knowledge of ascomycetes and their anamorphs by investigating these microfungi on leaf litter and dead wood. We will are investigating the insecticidal properties of these fungi using bioassays.

Materials and Methods



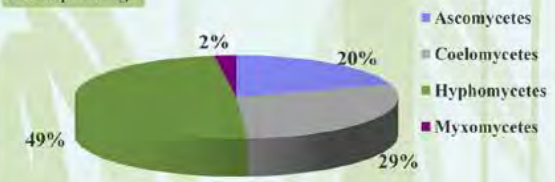
2. Plate of some interesting and probably new species.

Results

1. Number of fungal taxa found on dead leaves and woods from each host in two collection sites.

Collection sites	Host	Type of samples	Number of fungal taxa			
			Ascomycetes	Anamorphic fungi		Others
				Coelomycetes	Hyphomycetes	
1. Medicinal Plant Garden, Doi Suthep-Pui National Park	*Magnolia liliifera*	Dead leaves	9	11	16	0
	Cinnamomum iners	Dead leaves	2	6	10	0
	Unknown	Dead wood	3	4	8	2
	Wood					
2.Houi Kok Mah, Doi Suthep-Pui National Park	Unknown Wood	Dead wood	3	5	10	0
3.Doi Tung National Park	Unknown Wood	Dead wood	1	0	0	0

3. Group of fungi



Discussions and Conclusions

We are in process of identify fungal species. Now, 70 cultures have been selected for further molecular phylogenetic analysis. New and interesting fungi have been described and will be published. We will go to collect more samples from same site next season. Furthermore, insecticidal bioassays will be developed and tested on all cultures.

Acknowledgements

This project is supported by Thailand Research Fund (BRG 5280002) which is gratefully acknowledged. The authors would like to sincerely thank The School of Science, Scientific and Technological Instruments Center, Mae Fah Luang University for continued support. We also thank Prof. Dr. Lei Cai for assistance.

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Sutton, B.C. (1980). The Coelomycetes. Kew, UK. Commonwealth Mycological Institute.

An oral paper entitled “Diversity of fungi on leaf litter of *Magnolia lillifera* and *Cinnamomum iners* from Doi Suthep-Pui National Park, Thailand” was given by Mae Fah Luang University at international Symposium on "Fungal Biodiversity and Resources"at Wangcam Hotel, Chiag Rai province 11-13 November 2010

BRG5280002

Diversity of fungi on leaf litter of *Magnolia lillifera* and *Cinnamomum iners* from Doi Suthep-Pui National Park, Thailand

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Leaf litter fungi play an important role as decomposers in natural ecosystems. One major aim to study these saprobic fungi is to establish their interaction with plant hosts whether they are generalists, host specific or host recurrent. In this study, two tree species namely *Magnolia liliifera* and *Cinnamomum iners* in the forest of Doi Suthep-Pui National Park, Chiang Mai Province were selected. Decaying leaves of each host were collected during dry season and examined for fungal presence. Fungal communities (numbers and species) from different hosts were then recorded and these data were used to assess fungal diversity using statistical analyses. For *M. liliifera*, 36 taxa were identified comprising 9 ascomycetes and 27 anamorphic fungi (11 coelomycetes and 16 hyphomycetes). Dominant species were *Ellisembia* sp.1 (46.7%), *Stachybotrys* sp.1 (40.0%), *Colletotrichum* sp.1 (26.7%) and *Ophioceras* sp.1 (26.7%). Eighteen taxa were identified from *C. iners* including 2 ascomycetes and 16 anamorphic fungi (6 coelomycetes and 10 hyphomycetes). Dominant species were anamorph of *Eutypa* sp.1 (26.7%), *Pleurophragmium* sp.1 (20.0%) and *Colletotrichum* sp.1 (16.7%). The overlapping fungi of both tree species are anamorph of *Eutypa* sp., *Colletotrichum* sp., *Diplodia* sp. and *Ophioceras* sp.
Keywords: fungal diversity, leaf litter fungi, Thailand

Appendix F

Award

Eminent Mycologist Award

Awarded to

Dr. Kevin Hyde

For outstanding professional achievements and contributions to the Asian Mycological Committee

Presented by

Dr. Xingzhong Liu

The President of Asian Mycology Committee

August 7, 2011

BRG5280002