# MOLECULAR TAXONOMIC STUDIES OF SELECTED MEMBERS OF THE XYLARIACEAE (FUNGI)

Nuttika Suwannasai

A Thesis Submitted in Partial Fulfillment of the Requirements for the

Degree of Doctor of Philosophy in Microbiology

**Suranaree University of Technology** 

Academic Year 2005

ISBN 974-533442-1

# การศึกษาอนุกรมวิชานเชิงโมเลกุลของเชื้อรากลุ่ม XYLARIACEAE

นางสาวณัฏฐิกา สุวรรณาศรัย

วิทยานิพนธ์นี้เป็นส่วนหนึ่งของการศึกษาตามหลักสูตรปริญญาวิทยาศาสตรดุษฎีบัณฑิต สาขาวิชาจุลชีววิทยา มหาวิทยาลัยเทคโนโลยีสุรนารี ปีการศึกษา 2548 ISBN 974-533442-1

# **MOLECULAR TAXONOMIC STUDIES OF SELECTED MEMBERS OF THE XYLARIACEAE (FUNGI)**

Suranaree University of Technology has approved this thesis submitted in

partial fulfillment of the requirements for the Degree of Doctor of Philosophy.

Thesis Examining Committee

Nothant Than

(Dr. Nathawut Thanee)

Chairperson

(Asst. Prof. Dr. Sureelak Rodtong)

Member (Thesis Advisor)

(Prof. Dr. Anthony J.S. Whallev)

Member

<u>G. Lumhirun</u> (Dr. Surang Thienhirun)

Member

SD Hickobe Sangsoda

(Asst. Prof. Dr. Siddhichoke Sangsoda)

Member Komson Enapatungsuniy (Dr. Komson Pirapatrungsuriya)

Member

S. Rattushi

Thammathaver

(Assoc. Prof. Dr. Saowanee Rattanaphani) (Assoc. Prof. Dr. Sompong Thammathaworn) Acting Vice Rector for Academic Affairs Acting Dean of Institute of Science

ณัฏฐิกา สุวรรณาศรัย : การศึกษาอนุกรมวิชานเชิง โมเลกุลของเชื้อรากลุ่ม XYLARIACEAE (MOLECULAR TAXONOMIC STUDIES OF SELECTED MEMBERS OF THE XYLARIACEAE (FUNGI)) อาจารย์ที่ปรึกษา : ผู้ช่วยศาสตราจารย์ คร. สุรีลักษณ์ รอดทอง, 352 หน้า. ISBN 974-533442-1

การศึกษาเพื่อการระบุและจัดจำแนกชนิดของเชื้อราในกลุ่ม Xylariaceae โดยใช้ข้อมูลทาง ชีววิทยาโมเลกุลนี้เพื่อช่วยแก้ปัญหาที่ไม่สามารถระบุและจัดจำแนกชนิดของเชื้อราที่มีลักษณะทาง สัณฐานที่ใกล้เคียงกันมากและชนิคที่ไม่สามารถเพาะเลี้ยงได้ โดยได้ศึกษาตัวอย่างเชื้อราจากทั้งแหล่ง อ้างอิงจำนวน 31 ตัวอย่าง และแหล่งธรรมชาติจำนวน 338 ตัวอย่าง จาก 14 พื้นที่ใน 11 จังหวัดของ ประเทศไทย ซึ่งจากการศึกษาลักษณะทางสัณฐานและทางเคมีเพื่อระบุชนิดของเชื้อราจากแหล่ง ธรรมชาติพบว่ามีความผันแปรสูงของลักษณะทางสัณฐานและมีข้อจำกัดในการศึกษาแผนภูมิของสาร ทุติยภูมิ ทำให้ไม่สามารถระบุและจัดจำแนกชนิดของตัวอย่างประมาณร้อยละ 30 (จาก 338 ตัวอย่าง) จากนั้นได้สึกษาโดยใช้เทคนิคทางชีววิทยาโมเลกุลโดยหาลำดับนิวคลีโอไทด์ของ 18S rDNA ซึ่ง พบว่ามีขนาดประมาณ 2,000 ถึง 2,200 คู่เบส และ/หรือส่วน Internal transcribed spacer (ITS) 1 และ 2 รวมทั้ง 5.8S rDNA (ITS1-5.8S-ITS2) ซึ่งพบว่ามีขนาดประมาณ 500 ถึง 900 คู่เบส เมื่อ เปรียบเทียบลำดับนิวคลีโอไทค์ภายในกลุ่มของเชื้อราที่ศึกษาและจากฐานข้อมูล GenBank พบว่า ้ลำดับนิวคลีโอไทค์ของเชื้อราที่ศึกษาแต่ละชนิคมีความแตกต่างกันและสามารถระบุชนิดของ ้ตัวอย่างเชื้อราที่มีปัญหาได้อย่างชัดเจน และเมื่อนำลำดับนิวกลีโอไทด์ที่ได้มาจัดแนวความสัมพันธ์ที่ เหมาะสม พบความผันแปรสูงที่สุดในส่วน ITS1 ซึ่งเป็นประโยชน์ในการออกแบบ primers และ probes ที่จำเพาะต่อเชื้อ จากการศึกษาความสัมพันธ์ทางพันธุกรรมในรูปของ Phylogenetic tree ของเชื้อกลุ่มที่ศึกษา พบว่าสามารถอธิบายความสัมพันธ์ของเชื้อราแต่ละชนิดได้และสามารถยืนยัน ้ผลของการพบเชื้อราชนิดใหม่ได้อย่างชัดเจน ทั้งนี้พบว่าเชื้อราที่ได้จากแหล่งธรรมชาติทั้งสิ้นมี 9 สกุล (Astrocystis, Biscogniauxia, Camillea, Daldinia, Hypoxylon, Kretzschmaria, Nemania, Rosellinia และ Xylaria) 59 ชนิด ซึ่งรวมชนิคใหม่ 9 ชนิค คือ Biscogniauxia 1 ชนิด Hypoxylon 5 ชนิด และ Xylaria 3 ชนิด ลำคับนิวคลีโอไทด์ที่ได้ยังเป็นข้อมูลสำคัญในการ สร้างฐานข้อมูลของลำคับนิวกลีโอไทค์ของเชื้อรากลุ่ม Xylariaceae ในประเทศไทย

สาขาวิชาจุลชีววิทยา ปีการศึกษา 2548 ลายมือชื่อนักศึกษา <u>ณิรูรู้ ก</u> *พอรร กาอโร* ลายมือชื่ออาจารย์ที่ปรึกษา <u>borner</u> ลายมือชื่ออาจารย์ที่ปรึกษาร่วม **AVFW cler** ลายมือชื่ออาจารย์ที่ปรึกษาร่วม <u>AVFW cler</u>

# NUTTIKA SUWANNASAI : MOLECULAR TAXONOMIC STUDIES OF SELECTED MEMBERS OF THE XYLARIACEAE (FUNGI). THESIS ADVISOR : ASST. PROF. SUREELAK RODTONG, Ph.D. 352 PP. ISBN 974-533442-1

### XYLARIACEAE/NUCLEOTIDE SEQUENCE/PHYLOGENY/INTERNAL TRANSCRIBED SPACER REGIONS

Species identification and classification of the fungi in the family Xylariaceae based on their molecular data were studied for resolving undescribed species, which were closely related in their morphological characteristics, and some were uncultured specimens. In this study, thirty one specimens from reference sources and three hundred and thirty eight specimens from natural habitats of 14 localities in different 11 provinces of Thailand were examined. Morphological and chemical characterisation results showed high morphological variations and limitations in their secondary metabolite profiles. Approximately 30 % of all collected specimens could not be identified. The molecular technique was then performed. Nucleotide sequences of 18S rDNA having approximately 2,000 to 2,200 bp, and/or the internal transcribed spacer (ITS) 1 and 2 regions including 5.8S rDNA (ITS1-5.8S-ITS2) having approximately 500 to 900 bp, were achieved. The comparison of these nucleotide sequences within specimens examined and sequences from GenBank database exhibited clearly separations among xylariaceous species and these sequences can be used to identify the problem fungi. When the whole ITS sequences were aligned, they revealed the greatest variation in ITS1 region, which was suitable to design specific primers and probes for these particular strains. The phylogenetic trees showed clear relationships within xylariaceous species and also could be used to confirm results of the finding of new species. From this study, the xylariaceous fungi were identified as belonging to nine genera; *Astrocystis, Biscogniauxia, Camillea, Daldinia, Hypoxylon, Kretzschmaria, Nemania, Rosellinia* and *Xylaria*, and were represented by fifty nine species, including nine new species, which one, five, and three species belonged to *Biscogniauxia, Hypoxylon,* and *Xylaria* respectively. In addition, these molecular data are valuable for the creation of the DNA sequence database of the xylariaceous fungi found in Thailand.

School of Microbiology

Academic Year 2005

Student's Signature <u>Nutlika Suvannasa</u> i	
Advisor's Signature Suradak Raditary	
Co-advisor's Signature Arthulalley.x	
Co-advisor's Signature S. Thimhirun	

#### ACKNOWLEDGEMENTS

I would like to thank the Thailand Research Fund through the Royal Golden Jubilee Ph.D. Program (Grant No. PHD/0096/2544) for financial support during my study. Laboratory facilities were supported by Suranaree University of Technology, Nakhon Ratchasima, Thailand, Liverpool John Moores University, Liverpool, U.K., and the Royal Forest Department, Bangkok, Thailand.

I would like to thank my thesis advisor, Asst. Prof. Dr. Sureelak Rodtong for giving me the opportunity to study on this thesis, and for her valuable guidance and suggestions throughout my study. My sincere thank is extended to my co-advisor, Prof. Dr. Anthony J.S. Whalley for supporting the laboratory facilities in Liverpool John Moores University, and for his valuable comments and suggestions. My warmest thanks to my co-advisor, Dr. Surang Thienhirun for her kind guidance of xylariaceous identification and sample collection in the field survey, and also provided several xylariaceous reference strains used in this study.

I would like to thank Dr. Ray Edwards (University of Bradford, U.K.) for helping to analyze secondary metabolite profiles. My sincere thank is also given to Prof. Dr. Colin Reynolds, Dr. George P. Sharples, and all staff at School of Biomolecular Sciences, Liverpool John Moores University, for their helps and continuous support throughout my stayed in Liverpool. My special thanks to Dr. Margaret Ann Whalley, Dr. Yu-Ming Ju (University of Taiwan, Taiwan), Dr. Nuttaporn Ruchikachorn, and Dr. Lianne Nugent for providing xylariaceous reference strains and endophytes.

I am very grateful to thank Mrs. Hadthairat Urairong and all staff at the Biotechnology and Development Office, Department of Agriculture, Prathumthani, Thailand for laboratory facility supporting on DNA sequencing. I would like to thank Dr. Jittra Pipeukaew (Chulalongkorn University, Thailand), Mr. Pravech Ajawatanawong (National Center for Genetic Engineering and Biotechnology, Thailand), and Dr. Eric Rivals (Université de Mons Hainaut, France) for supporting all phylogenetic software.

My sincere thanks is also due to Asst. Prof. Dr. Saowalak Pongpaijit (Prince of Songkla University, Thailand), Dr. Cherdchai Phosri (Pibulsongkram Rajabhat University, Thailand), and Miss Kunthika Vechklang for their help to collect specimens. I would like to thank Miss Korawan Ratanachai for her help on scanning electron microscope technique.

My great appreciations are extended to all staff and friends in the Schools of Biology and Microbiology, Suranaree University of Technology, Nakhon Ratchasima, Thailand, and friends in Liverpool for their help and friendship.

Most of all, my deepest gratitude is extended to my parents and cousins for their infinite love, understanding and constant encouragement throughout my study.

Nuttika Suwannasai

### CONTENTS

#### Page

ABSTRACT IN THAI	Ι
ABSTRACT IN ENGLISH	II
ACKNOWLEDGEMENTS	IV
CONTENTS	VI
LIST OF TABLES	Х
LIST OF FIGURES	XIII
LIST OF ABBREVIATIONS	XX
CHAPTER	
I INTRODUCTION	
1.1 Significance of the study	1
1.2 Research objectives	2
1.3 Scope and limitations of the study	2
1.4 Expected results	3
II LITERATURE REVIEW	
2.1 The Xylariaceae	4
2.1.1 Ecology and host preference of the xylariaceous fungi	4
2.1.2 Xylariaceous fungi as phytopathogens	6
2.1.3 Xylariaceous fungi as endophytes	9
2.2 Taxonomy of the Xylariaceae	13
2.2.1 Morphological taxonomy	16

## **CONTENTS** (Continued)

Page
------

2.2.1.1 Teleomorphic characteristics	16
2.2.1.2 Anamorphic characteristics	26
2.2.2 Chemical taxonomy	30
2.2.3 Molecular taxonomy	33
2.2.3.1 Ribosomal DNA	33
2.2.3.2 Phylogenetic study	38
2.2.3.3 Molecular studies of the Xylariaceae	42
2.3 Problematic groups of xylariaceous fungi	46
2.3.1 Group I: Astrocystis and Rosellinia	46
2.3.2 Group II: Camillea	48
2.3.3 Group III: Daldinia	50
2.3.4 Group IV: <i>Hypoxylon</i>	54
2.3.5 Group V: Xylariaceous endophytes	55
III MATERIALS AND METHODS	
3.1 Chemicals, reagents, and media	57
3.2 Instrumentation	58
3.3 Collection of xylariaceous fungi for taxonomic studies	59
3.4 Morphological taxonomic studies of the problematic groups in	
xylariaceous fungi	60
3.5 Isolation and cultivation of the selected xylariaceous fungi	61
3.6 Chemotaxonomic study of the selected xylariaceous fungi	61

#### **CONTENTS** (Continued)

	3.7 Nucleic acid studies of the selected xylariaceous fungi	63
	3.7.1 Extraction of genomic DNA	63
	3.7.2 Amplification of the ribosomal RNA genes	64
	3.7.3 Detection of PCR-amplified products by agarose gel	
	electrophoresis	66
	3.7.4 Purification of DNA-amplified products	67
	3.7.5 Sequencing of ribosomal DNA	67
	3.7.6 Alignment of DNA sequences	68
	3.7.7 Construction of phylogenetic tree	69
IV	<b>RESULTS AND DISCUSSION</b>	
	4.1 Collection of xylariaceous fungi for taxonomic studies	70
	4.2 Morphological taxonomic studies of the problematic groups in	
	xylariaceous fungi	71
	4.2.1 Group I: Astrocystis and Rosellinia	72
	4.2.2 Group II: Camillea	77
	4.2.3 Group III: Daldinia	81
	4.2.4 Group IV: <i>Hypoxylon</i>	84
	4.2.4.1 Hypoxylon section Annulata	86
	1212 Hunomlan section Hunomlan	101

- 4.2.4.2 *Hypoxylon* section *Hypoxylon* ..... 101

Page

## **CONTENTS** (Continued)

#### Page

4.3.1 TLC analysis of secondary metabolites from agar plugs	191
4.3.2 TLC analysis of secondary metabolites from cultural broth	191
4.4 Nucleic acid studies of the selected xylariaceous fungi	196
4.4.1 Group I: Astrocystis and Rosellinia	196
4.4.2 Group II: <i>Camillea</i>	207
4.4.3 Group III: Daldinia	214
4.4.4 Group IV: <i>Hypoxylon</i>	218
4.4.4.1 Hypoxylon section Annulata	219
4.4.4.2 Hypoxylon section Hypoxylon	231
4.4.5 Group V: Xylariaceous endophytes	242
4.4.6 Phylogenetic analysis of xylariaceous fungi based on ITS1-5.8S-	
ITS2 sequence	249
V CONCLUSION AND FUTURE PERSPECTIVE	252
REFERENCES	256
APPENDICES	282
Appendix A Fungal media and reagents	284
Appendix B Locations and details of xylariaceous collections	287
Appendix C Nucleotide sequences data	295
Appendix D Phylogenetic tree construction	321
Appendix E List of presentations	332
CURRICULUM VITAE	352

### LIST OF TABLES

Table		Page
1	The categorization of the xylariaceous fungi based on the part or	
	position of invading host	5
2	Examples of xylariaceous phytopathogens	8
3	The fungal genera within the Xylariaceae family	14
4	The anamorph-teleomorph relationship within genera of	
	the Xylariaceae	28
5	The applicable categorical levels of each molecular marker or gene	
	region in molecular taxonomic study	38
6	Locations and time of specimen collection in this study	59
7	Nucleotide sequences of PCR primers used in this study	65
8	Xylariaceous collections from 14 locations of Thailand in years	
	2002 and 2003	70
9	Numbers of genera and species of xylariaceous collections in	
	this study in years 2002 and 2003	72
10	Species comparison of Astrocystis and Rosellinia found in this study	73
11	Morphological characteristics of Camillea tinctor found in this study,	
	C. selangorensis, and C. leprieurii	78
12	Morphological characteristics of Daldinia eschscholzii found in this	
	study and the reference specimen of <i>D. concentrica</i>	82

## LIST OF TABLES (Continued)

Table		Page
13	Species of Hypoxylon sect. Annulata, and sect. Hypoxylon	
	found in this study	84
14	Morphological characteristics of Hypoxylon sect. Annulata	
	found in this study	86
15	Morphological characteristics of Hypoxylon sect. Hypoxylon	
	found in this study	103
16	Species of Xylaria, Kretzschmaria, Nemania, and Biscogniauxia	
	found in this study	144
17	Morphological characteristics of Xylaria, Kretzschmaria, Nemania,	
	and <i>Biscogniauxia</i> found in this study	147
18	Nucleotide sequences of NS4 and SR8R primers	197
19	The length of 18S rDNA sequences of A. mirabilis SUT051, SUT056,	
	and Rosellinia sp. ST2310 obtained from DNA sequence analysis	199
20	The length of ITS1-5.8S-ITS2 sequences of A. mirabilis and	
	Rosellinia sp. ST2310 obtained from DNA sequence analysis	205
21	The length of ITS1-5.8S-ITS2 sequences of Camillea tinctor and	
	C. selangorensis obtained from DNA sequence analysis	209
22	The identity matrix of ITS1-5.8S-ITS2 sequence comparison of	
	Camillea tinctor and C. selangorensis	211
23	STR motifs found in the isolates of Camillea tinctor and	
	C. selangorensis	212

## LIST OF TABLES (Continued)

Table		Page
24	The length of ITS1-5.8S-ITS2 sequences of Daldinia eschscholzii and	
	D. concentrica obtained from DNA sequence analysis	216
25	The length of ITS1-5.8S-ITS2 sequences of Hypoxylon sect. Annulata	
	obtained from DNA sequence analysis	221
26	The length of ITS1-5.8S-ITS2 sequences of Hypoxylon sect. Hypoxylon	
	obtained from DNA sequence analysis	232
27	The length of ITS1-5.8S-ITS2 sequences of different species of	
	Xylaria, Kretzschmaria, Nemania, and Biscogniauxia found in this	
	study	244

### LIST OF FIGURES

Fig	Figure	
1	Stromata of the xylariaceous fungi	18
2	Perithecia of the xylariaceous fungi	19
3	Ostioles of the xylariaceous fungi	21
4	The apical apparatus forms of the xylariaceous fungi	23
5	Germ slit characters of the xylariaceous fungi	26
6	Schematic diagram of a tandem repeat unit of rDNA	35
7	Phylogenetic tree styles	40
8	The map of oligonucleotide primers for 18S rDNA and ITS region	
	amplication	65
9	Astrocystis mirabilis Berk. & Broome (SUT051)	74
10	Rosellinia procera Syd. (SUT113)	76
11	Camillea tinctor (Berk.) Læssøe, J.D. Rogers & Whalley. (SUT260)	79
12	Camillea selangorensis M.A. Whalley, A.J.S. Whalley & E.B.G. Jones,	
	(KS15) and <i>C. leprieurii</i> (Mont.) Mont.	80
13	Daldinia eschscholzii (Ehrenb.: Fr.) Rehm (SUT039)	83
14	<i>Hypoxylon</i> cf. <i>archeri</i> (SUT105)	91
15	Hypoxylon atroroseum J.D. Rogers (SUT009)	92
16	Hypoxylon bovei Speg. var. microspora J.H. Miller (SUT025)	93
17	Hypoxylon moriforme Henn. (SUT220)	94
18	Hypoxylon purpureonitens YM. Ju & J.D. Rogers (SUT004)	97

Fig	ure	Page
19	Hypoxylon stygium (Lév.) Sacc. (SUT058)	98
20	Hypoxylon urceolatum (Rehm) YM. Ju & J.D. Rogers (SUT098)	99
21	Hypoxylon taxonomic species 1 sp. nov. (SUT236)	100
22	Hypoxylon anthochroum Berk. & Broome (SUT233)	112
23	Hypoxylon brevisporum YM. Ju & J.D. Rogers (SUT256)	113
24	Hypoxylon duranii J.D. Rogers (SUT223)	114
25	Hypoxylon fendleri Berk. ex Cooke (SUT162)	115
26	<i>Hypoxylon</i> cf. <i>ferrugineum</i> (SUT017)	116
27	<i>Hypoxylon</i> cf. <i>ferrugineum</i> (SUT070)	117
28	<i>Hypoxylon</i> cf. <i>ferrugineum</i> (SUT237)	118
29	Hypoxylon haematostroma Mont. (SUT164)	119
30	Hypoxylon hypomiltum Mont. (SUT166)	121
31	Hypoxylon investiens (Schwein.) M.A. Curtis (SUT063)	122
32	Hypoxylon lenormandii Berk. & M.A. Curtis (SUT065)	123
33	Hypoxylon lenormandii var. microspora (SUT022)	124
34	<i>Hypoxylon macrocarpum</i> Pouzar (SUT045)	125
35	Hypoxylon monticulosum Mont. (SUT116)	127
36	Hypoxylon cf. perforatum (SUT020)	128
37	Hypoxylon cf. perforatum (SUT224)	129
38	Hypoxylon cf. perforatum (SUT294)	130
39	Hypoxylon rubiginosum (Pers.: Fr.) Fr. (SUT215)	131

Fig	Figure	
40	Hypoxylon subgilvum Berk. & Broome var. microsporum (Abe) YM. Ju	
	& J.D. Rogers (SUT234)	135
41	Hypoxylon trogodes Berk. & Broome (SUT187)	136
42	Hypoxylon trogodes Berk. & Broome (SUT154)	137
43	Hypoxylon kanchanapisekii sp. nov. (SUT069)	138
44	Hypoxylon sublenormandii sp. nov. (SUT282)	139
45	Hypoxylon suranareei sp. nov. (SUT182)	140
46	Hypoxylon taxonomic species 2 (SUT082)	142
47	Hypoxylon taxonomic species3 (SUT158)	143
48	Xylaria anisopleura (Mont.) Fr. (SUT205)	157
49	Xylaria badia Pat (SUT076)	158
50	Xylaria beccari Lloyd (SUT092)	160
51	Xylaria brachiata Sacc. (SUT078)	161
52	Xylaria cubensis (Mont.) Fr. (SUT089)	162
53	Xylaria inthino-velutina (Mont.) Fr. (SUT123)	163
54	<i>Xylaria</i> cf. <i>juruensis</i> (SUT035)	165
55	<i>Xylaria</i> cf. <i>juruensis</i> (SUT088)	166
56	<i>Xylaria</i> cf. <i>juruensis</i> (SUT140)	167
57	<i>Xylaria</i> cf. <i>juruensis</i> (SUT170)	168
58	Xylaria juruensis var. microspora (SUT129, SUT138, and SUT139)	169
59	Xylaria maitlandii (Dennis) D. Hawksw (SUT177)	170

----

Fig	ure	Page
60	Xylaria mellisii (Berk.) Cooke (SUT192)	171
61	<i>Xylaria</i> cf. <i>multiplex</i> (SUT028)	172
62	Xylaria muscula Lloyd (SUT029)	175
63	Xylaria psidii J.D. Rogers & Hemmes (SUT125)	176
64	Xylaria schweinitzii Berk. & M.A. Curtis (SUT201)	177
65	Xylaria scruposa (Fr.) Fr. (SUT117)	178
66	Xylaria species 2 (SUT155)	179
67	Xylaria sp. nov. (SUT195)	180
68	Xylaria telfairii (Berk.) Fr. (SUT206)	181
69	<i>Xylaria</i> taxonomic species 1 (SUT075)	183
70	<i>Xylaria</i> taxonomic species 2 (SUT203)	184
71	<i>Xylaria</i> taxonomic species 3 (SUT204)	185
72	<i>Xylaria</i> taxonomic species 4 (SUT207)	186
73	Kretzschmaria species (SUT101)	187
74	Nemania species (SUT258)	188
75	Biscogniauxia capnodes (Berk.) YM. Ju & J.D. Rogers (SUT212)	189
76	Biscogniauxia sp. nov. (SUT290)	190
77	Secondary metabolite profiles of Xylaria isolates compared to those of	
	Xylaria endophytes extracted from 100-mL cultural broth and analyzed	
	by TLC method under visible light	192

Fig	Figure	
78	The pattern of secondary metabolites extracted from X. cubensis and other	
	fungal endophytes by TLC method	194
79	Schematic of secondary metabolites extracted from Xylaria cubensis and	
	other endophytes by TLC method	195
80	Gel electrophoresis of partial 18S rDNA fragments of Astrocystis and	
	Rosellinia	198
81	Sequence alignment of 18S rDNA Rosellinia sp. (ST2310),	
	Rosellinia necatrix (AB014044), and A. mirabilis (SUT056)	200
82	Gel electrophoresis of ITS1-5.8S-ITS2 fragments of Astrocystis and	
	Rosellinia using ITS5 and ITS4 primers	204
83	Phylogenetic tree of Astrocystis and Rosellinia based on ITS1-5.8S-ITS2	
	sequences constructed by using the neighbour-joining method	206
84	Gel electrophoresis of ITS1-5.8S-ITS2 fragments of Camillea using ITS5	
	and ITS4 primers	208
85	ITS1 sequence alignment of Camillea by using ClustalX and BioEdit	
	programs	210
86	Phylogenetic tree of Camillea based on ITS1-5.8S-ITS2 sequences	
	constructed by using the neighbour-joining method	213
87	Gel electrophoresis of ITS1-5.8S-ITS2 fragments of Daldinia using ITS5	
	and ITS4 primers	215

Fig	ure	Page
88	Phylogenetic tree of Daldinia species based on ITS1-5.8S-ITS2 sequences	
	using the neighbour-joining method	217
89	Gel electrophoresis of ITS1-5.8S-ITS2 fragments of Hypoxylon sect.	
	Annulata using ITS5 and ITS4 primers	220
90	Phylogenetic tree of Hypoxylon sect. Annulata containing extremely long	
	ITS1 region (398 bp to 588 bp) based on ITS1-5.8S-ITS2 sequences using	
	the neighbour-joining method	224
91	Phylogenetic tree of Hypoxylon sect. Annulata based on ITS1-5.8S-ITS2	
	sequences using the neighbour-joining method	227
92	Phylogenetic tree of Hypoxylon sect. Annulata based on ITS2 sequences	
	using the neighbour-joining method	229
93	Gel electrophoresis of ITS1-5.8S-ITS2 fragments of Hypoxylon sect.	
	Hypoxylon using ITS5 and ITS4 primers	232
94	Phylogenetic tree of <i>Hypoxylon</i> sect. <i>Hypoxylon</i> based on ITS1-5.8S-ITS2	
	sequences using the neighbour-joining method	227
95	Phylogenetic tree of Hypoxylon based on ITS2 sequences using the	
	neighbour-joining method	241
96	Gel electrophoresis of ITS1-5.8S-ITS2 fragments of Xylaria using ITS5	
	and ITS4 primers	243
97	Phylogenetic tree of Xylaria based on ITS1-5.8S-ITS2 sequences using the	
	neighbour-joining method	247

Figure		Page
98	Phylogenetic tree of xylariaceous fungi based on ITS1-5.8S-ITS2	
	sequences using the neighbour-joining method	250

### LIST OF ABBREVIATIONS

BLAST	Basic Local Alignment Search Tool
bp	Base pair
°C	Degree Celsius
dATP	Deoxyadenosine triphosphate
dCTP	Deoxycytidine triphosphate
dGTP	Deoxyguanosine triphosphate
dNTPs	Deoxynucleoside triphosphate (dATP, dCTP, dGTP, dTTP)
dTTP	Deoxythymidine triphosphate
DNA	Deoxyribonucleic acid
et al.	et alia (and others)
(m, µ) g	(milli, micro) Gram
h	Hour
(m, µ) L	(milli, micro) Litre
(m, µ) M	(milli, micro) Molar
(c, m) m	(centri, milli) Metre
min	Minute
(m, µ) mol	(milli, micro) Mole
%	Percent
PAUP	Phylogenetic analysis using parsimony
PCR	Polymerase chain reaction

## LIST OF ABBREVIATIONS (Continued)

PDA	Potato dextrose agar
rDNA	Ribosomal deoxyribonucleic acid
rpm	Round per minute
sp.	Species
TLC	Thin layer chromatography
v/v	Volume by volume
UV	Ultraviolet

#### **CHAPTER I**

#### **INTRODUCTION**

#### **1.1** Significance of the study

The Xylariaceae is a large and relatively well-known fungal family which is represented in most countries of the world especially in the tropics and subtropics. The fungi in this group play an important role in the natural functions of forest ecosystems. They are wood-decay fungi that are able to break down the major components of wood, and play a role in nutrient cycling in the forest. In addition, the Xylariaceae is known to contain phytopathogens and also endophytes. It has been well investigated for secondary metabolite production (Whalley and Edwards, 1995; Andersen *et al.*, 2001; Stadler *et al.*, 2001; Mühlbauer *et al.*, 2002; Quang *et al.*, 2002; Stadler *et al.*, 2004).

In taxonomic studies of xylariaceous fungi, conventional methods, including morphological, cultural, and chemical features, have been used. They are still frequently used although these methods have limitations regarding very closely related species. Thus, conventional methods are unable to resolve the problem and the confusion in some areas of investigation of the family. Many species are cosmopolitan, and have been frequently reported from different localities and at different stages of development. The xylariaceous fungi also show a great variation in their morphology, and some do not form a teleomorph stage, which causes a difficulty in identification and classification. Therefore, molecular techniques have been chosen to resolve these problems. The nucleic acid sequence data have been successfully applied for the study of evolutionary patterns and phylogeny in fungi. The aim of this study is to apply nucleic acid data based on ribosomal DNA sequences to resolve and clarify the situation regarding selected xylariaceous fungi, where conventional methods have been unsuccessful.

#### **1.2 Research objectives**

This study was undertaken to resolve the selected members of xylariaceous fungi, which are difficult to identify. Therefore, three specific objectives were investigated as follows:

1) to investigate species boundaries in problem species complexes, where traditional taxonomic methodology has failed to resolve the problems,

2) to develop a database for identification of anamorphic isolates of endophytic Xylariaceae, which can not be identified by conventional methods, and

3) to apply molecular techniques to clarify taxonomic relationships in certain genera.

#### **1.3** Scope and limitations of the study

The species complex of selected xylariaceous fungi were studied based on the 18S ribosomal DNA sequence and/or the internal transcribed spacers (ITS) 1 and 2 including 5.8S ribosomal DNA sequence. The nucleotide sequence results were compared to the morphological results. Xylariaceous fungal specimens were collected from forests in Thailand whilst the reference species were obtained from the Royal Forest Department, Thailand, the Liverpool John Moore University, U.K., and the University of Taiwan, Taiwan. The morphological characteristics of the selected xylariaceous fungi were observed. Their chemical characteristics were analyzed by secondary metabolite profiles and compared to the xylariaceous endophytes. Then, this study attempted to resolve and analyze the genetic relationships of selected xylariaceous fungi using different techniques suitable for each genus and species.

#### **1.4 Expected results**

From this study, the nucleotide sequences of the xylariaceous fungi could clearly explain the identification and classification among the problematic genera and/or species. The phylogenetic analysis could help for better understanding of taxonomic and evolutionary relationships among xylariaceous fungi by means of ribosomal DNA sequence analysis. The information of DNA sequences could also be used to design specific primers as well as probes for the detection of specific xylariaceous species in further application. Moreover, the nucleotide sequence database of xylariaceous fungi collected in Thailand would then be developed.

#### **CHAPTER II**

#### LITERATURE REVIEW

#### 2.1 The Xylariaceae

The Xylariaceae is a fungus family belonging to Phylum Ascomycota, and is commonly found throughout the temperate and tropical regions of the world (Ju and Rogers, 1996; Whalley, 1996; Rogers, 2000). The xylariaceous fungus generally has a paraphysate hamathecium and an ascus with the apical apparatus containing 4 to 8 ascospores, one-celled ascospores with a germination slit in each spore, and (Rogers, 1994; Ju and Rogers, 1996; Whalley, 1996). Their habitats are mostly on wood, litter, leaves, seeds, dung, and soil. Some are associated with insect nests. Many species exhibit strong host selectivity and in some cases are host specific (Whalley, 1996).

#### 2.1.1 Ecology and host preference of the xylariaceous fungi

A major role of the Xylariaceae is wood decomposition, and most are reported as white-rot fungi which can produce enzymes to degrade all the major wood components (cellulose and lignin) (Nilsson *et al.*, 1989; Rogers, 2000). The wooddecay fungi in this family are similar to basidiomycete white-rot fungi (Sutherland and Crawford, 1981; Rogers *et al.*, 1997) but they decompose more slowly as found for *Daldinia concentrica* (Bolt.: Fr.) Ces. & De Not. (Merrill *et al.*, 1964; Rogers *et al.*, 1997).

Some xylariaceous species occur on a wide host range such as Hypoxylon rubiginosum Pers.: Fr. and Nemania bipapillata (Berk.) Pouzar, which have been found from several kinds of plants whilst some show a strong host specificity. For example, *Rosellinia buxi* Fabre has only been found on *Buxus sempervirens* L. (Whalley and Hammelev, 1988; Petrini, 1992; Whalley, 1996). *Hypoxylon fraxinophilum* (Bull.: Fr.) Kuntze is always found on *Fraxinus* (Pouzar, 1972) and *Biscogniauxia nummularia* (Bull.: Fr.) Küntze appears restricted to *Fagus* (Whalley and Edwards, 1987). Rogers (2000) categorized the Xylariaceae by the part or position and the invasion time of a host or substrate, which they invade as shown in Table 1.

**Table 1.** The categorization of the xylariaceous fungi based on the part or position of invading host.

Position of invading host	Genera and/or species
To invade living leaves and stems, and	Many species of Anthostomella
often found fruiting on the living host	
material	
To invade living stems and remain dormant	Many species of Daldinia, Biscogniauxia,
until the host is stressed	Camillea, and Hypoxylon
To decay living roots and wood then move	Kretzschmaria clavas (Fr.) Sacc., Rosellinia
to living material from dead material	necatrix and Xylaria spp.
To form fruiting bodies on decayed	Most species of Xylaria and Nemania
material, but to be isolated as endophytes	
from living hosts	
To form fruiting bodies on seed and fruits,	Xylaria magnolia J.D. Rogers found on
and have specific and discrete	Magnolia fruits, Xylaria ianthino-velutina
relationships with their hosts	(Mont.) Fr. found on leguminous pods,
	Xylaria carpophila (Pers.) Fr. found on
	Fagus fruits, and Xylaria persicaria
	(Schwein.: Fr) Berk. & M.A. Curtis found
	on Liquidambar fruits

#### Table 1. (Continued).

Position of invading host	Genera and/or species	
To inhabit dung, and found to be special	Most species of Hypocopra, Podosordaria,	
relationships with animals. Many taxa	and Poronia	
have dormant ascospores, that seem to be		
achieved via passage through a		
mammalian digestive tract.		
To associate with ant and termite nests	Most species of Xylaria including Xylaria	
	melanaxis Ces. and X. nigripes (Kl.) Sacc.	
To inhabit litter and organic soils	Xylariaceous anamorphs such as	
	Nodulisporium and Geniculosporium	
To damage host as pathogens	Camillea tinctor (Berk.) Læssøe, J.D. Rogers	
	& Whalley, Biscogniauxia capnodes	
	(Berk.) YM. Ju & J.D. Rogers, and B.	
	mediterranea (De Not.) Kuntze	

Source: Rogers (2000).

#### 2.1.2 Xylariaceous fungi as phytopathogens

Some xylariaceous fungi are considered to be weak plant pathogens causing canker disease, root rot disease, and needle blight disease (Whalley, 1996; Edwards *et al.*, 2003). Although they are not often considered to be a major cause of plant diseases, an increasing number of pathogenic species is now recognised which lead to economic loss in national ecosystems or under agricultural conditions (Rogers, 1979; Whalley, 1985; Whalley, 1996; Edwards *et al.*, 2003). Rogers (1979) and Rogers *et al.* (1997) reported that xylariaceous fungi are primarily parasites and saprophytes of angiosperm plants. Rogers believed that early angiosperms might have evolved in open areas with regular periods of drought. Therefore, one of the major factors in directing evolution of fungi associated with angiosperms might have been the capacity to survive through dry periods. If xylariaceous fungi co-evolved with

early angiosperms in exploiting the dry sites, they would have evolved to tolerate periods of drought. Rogers (1979) pointed out several properties of the Xylariaceae which might has been derived from co-evolving with their hosts on dry sites: a relatively long period of ascospore maturation and discharge, a rapid germination of ascospores in water, the discharge of ascospores when water is available, and the ability of perithecial stromata and ascospores to withstand severe desiccation (Rogers, 1979). These fungi may weaken the host by absorbing nutrients from it, blocking the vascular tissue, and preventing translocation of photosynthetic, water, and nutrients, or actually destroying cells. In some cases enzymes or toxins are produced (Alexopoulos *et al.*, 1996).

Some species of *Hypoxylon*, *Biscogniauxia*, *Camillea*, and *Xylaria* cause canker diseases (Whalley, 1996; Edwards *et al.*, 2003). Canker diseases contribute to the premature death of trees which have been stressed by drought, construction damage, or other problems. Examples of xylariaceous phytopathogens are shown in Table 2. In addition, these fungi have been investigated for phytotoxin production that may cause the disease (Bodo *et al.*, 1987; Pinon and Manion, 1991; Whalley, 1996; Edwards *et al.*, 2003).

Some species of *Rosellinia*, *Kretzschmaria*, and *Xylaria* cause root rot diseases as shown in Table 2 (Whalley, 1996; Edwards *et al.*, 2003). The symptoms of these diseases are similar to those of other root diseases, leaf yellowing, smaller leaves and premature leaf fall; some branches exhibit dieback. *Rosellinia necatrix* has been reported to produce rosellinic acid (Chen, 1964; Whalley, 1996), cytochalasin E (Aldridge, Burrows, and Turner, 1972; Whalley and Edwards, 1995; Whalley, 1996), rosellichalasin (Kimura, Nakajima, and Hamasaki, 1989; Whalley, 1996) and

rosnecatrone (Edwards *et al.*, 2001; 2003), which might have a significant role in causing the disease symptoms.

Members of xylariaceous fungi that cause needle blight diseases belong to species of *Rosellinia* (Whalley, 1996; Edwards *et al.*, 2003). Examples are noted in Table 2.

Species	Plant
Canker diseases	
Entoleuca mammata (Wahlenberg:	Acer, Alnus, Betula, Carpinus, Fagus, Picea,
F.) J.D. Rogers & YM. Ju	Pyrus, Salix, Sorbus, and Ulnus (Manion and
	Griffin, 1986; Whalley, 1996; Edwards <i>et al.</i> , 2003)
Biscogniauxia mediterranea	Oak (Macara, 1975; Whalley, 1996)
Biscogniauxia nothofagi Whalley,	Nothofagus cunninghamii (Whalley, Læssøe,
Læssøe & Kile	and Kile, 1990; Whalley, 1996)
Camillea punctulata (Berk. & Rev.)	Quercus (Barnett, 1957; Whalley, 1996;
Læssøe, J.D. Rogers & Whalley	Edwards et al., 2003)
Root rot diseases	
Rosellinia necatrix Prill.	Apple, grape wive, pear, plum, sweet cherry,
	poplar, jasmine and scented geranium
	(Cellerino, 1973; Cellerino and Anselmi,
	1980; Guillaumin, Mercier, and Dubois,
	1982; Teixeira de Sousa, 1985; Cellerino,
	Anselmi, and Giorcelli, 1988; Teixeira de
	Sousa et al., 1995; Whalley, 1996)
Rosellinia bunodes (Berk. &	Cacao (Theobroma cacao), quinine (Cinchona
Broome) Sacc.	spp.), coffee (Coffea spp.), rubber (Hevea
	brasiliensis), and tea (Camellia sinesis)
	(Sivanesan and Holliday, 1972; Whalley,
	1996; Edwards et al., 2003)
Kretzschmaria deusta (Hoffm.: Fr.)	Various tree species (Wilkins, 1934; Whalley,
P. Martin	1996; Edwards et al., 2003)
<i>Xylaria arbuscula</i> Fr.	Macadamia (Ko and Kunimoto, 1991)

**Table 2.** Examples of xylariaceous phytopathogens.

 Table 2. (Continued).

Species	Plant
Xylaria mali Fromme and Xylaria	Apple (Clayton, Julis, and Sutton, 1976;
polymorpha (Pers.: Fr.) Grev.	Whalley, 1996; Edwards et al., 2003), and
	Acer rubrum (Sivanesan and Holliday, 1972;
	Whalley, 1996)
Needle blight diseases	
Rosellinia herpotrichioides Hepting	Douglas fir (Pseudotsuga menziesii) in forest
& Davidson	nurseries (Salisbury and Long, 1956; Smith,
	1966; Whalley, 1996; Edwards et al., 2003)
Rosellinia minor (Höhn.) Francis	Young conifer seedlings (Francis, 1986)

#### 2.1.3 Xylariaceous fungi as endophytes

Endophytes are microorganisms that live inside the plant tissue for at least part of their life cycle without causing any disease symptom in the host (Petrini, 1992). Endophytes can be isolated from surface-sterilized plant tissues and cultivated on suitable nutrient agars. The grass or clavicipitaceous endophytes colonize inside of plant tissues and are believed to obtain their nutrition and some degrees of protection from the host plants. In turn, they can confer enhanced fitness to the host plants by producing certain functional metabolites. They are also implicated in improving the ecological adaptability of hosts by enhancing their tolerance to environmental stresses and resistance to phytopathogens and/or herbivores including some insects feeding on the host plant. Endophyte-infected grasses usually possess an increased tolerance to drought (Arachevaleta *et al.*, 1989; Ravel *et al.*, 1997), and aluminium toxicity (Malinowski and Belesky, 1999). Furthermore, some endophytes are able to provide the host plant with protection against some nematodes (Kimmons, Gwinn, and Bernard, 1990; Hallmann and Sikora, 1996), mammalian animals (Bacon *et al.*, 1977), and insect herbivores (Preazler, Gaylord, and Boecklen, 1996; Wilkinson *et* 

*al.*, 2000) as well as bacterial and fungal pathogens (Christensen, 1996; Sturz *et al.*, 1999). The non-clavicipitaceous endophytes occur in a wide range of non-grass hosts, are worldwide in their distribution, and are the source of many bioactive compounds (Schulz *et al.*, 2002; Strobel, 2002).

Over the past two decades, members of the Xylariaceae have been found to be widely and commonly occurring endophytic fungi being especially common and diverse in tropical plants (Petrini and Petrini, 1985; Whalley, 1996; Rodrigues and Petrini, 1997; Rogers, 2000). To date, eight genera of the Xylariaceae have been recorded as endophytes including Anthostomella, Biscogniauxia, Daldinia, Hypoxylon, Kretzschmaria, Nemania, Rosellinia, and Xylaria (Whalley, 1996). Endophytes have been widely investigated because of their ability to produce new or interesting metabolites, which can be used for natural, pharmaceutical, and biological controls of pests and diseases (Azevedo et al., 2000; Schulz et al., 2002; Strobel, 2002). An increasing numbers of studies show that individual xylariaceous species from a dominant part of the endophytes in certain tropical plant leaves (Rodrigues, 1994; Mekkamol, 1998; Photita et al., 2001). Studies on metabolites from xylariaceous fungi, including endophytic isolates, indicate that the family is a rich source of novel and often produces bioactive compounds (Whalley and Edwards, 1999; Isaka et al., 2000; Boonphong et al., 2001; Chinworrungsee et al., 2001; 2002). Xylaria cubensis (Mont.) Fr. was reported as the second most frequent species isolated from leaves of Licuala ramsayi (Muell.) Domin. (Rodrigues and Samuels, 1990) and an unidentified species of Xylaria was a frequent inhabitant of Stylosanthes guianensis Sw. leave (Pereira, Azevedo, and Petrini, 1993). Consequently, Rodrigues et al. (1993) demonstrated that xylariaceous fungi were the most frequent endophytes

isolated from *Euterpe oleracea*, especially *Xylaria cubensis*. Chapela (1989) isolated endophytic fungi from *Fagus grandifolia* and *Populus tremuloides* by using nonselective methods and found 32% and 41% to be xylariaceous fungi. In the study of leaf endophytes from a tropical palm, *Xylaria*, *Anthostomella*, *Daldinia*, and *Hypoxylon* were represented, and the most frequent species was *Xylaria cubensis* (Rodrigues, 1992). In most culture studies of leaf endophytes from tropical plants, *Xylaria* is abundant in plants tissue (Rodrigues, 1994).

Endophytic Xylaria species have been isolated from a wide range of plants including Euterpe, Trachycapus, and Livistona (Rodrigues, 1994; Taylor et al., 1999; Guo et al., 2000); Quercus and Fagus (Fagaceae); Betula, Corylus, and Alnus (Betulaceae); Acer (Sapindaceae); Fraxinus (Oleaceae); Rhizophora and Bruguiera (Rhizophoraceae); Avicennia (Avicenniaceae); Pinus and Picea (Pinaceae); and Nicotiana (Solanaceae) (Brunner and Petrini, 1992); Manilkara (Sapotaceae) (Lodge et al., 1996; Bayman et al., 1998); Tectona grandis L.f. (Mekkamol, 1998; Charesprasert 2001); Samanea saman Merr. (Charessprasert, 2001); Musa acuminate (Photita et al., 2001); Amomum siamense (Bussaban et al., 2001); bamboo (Lumyong et al., 2001); Lepanthes (Orchidaceae; Bayman et al., 1997); Casuarina (Casuarinaceae; Bayman et al., 1998); Schefflera (Araliaceae) (Læssøe and Lodge, 1994); Heisteria (Olaceae) and Ouratea (Ochnaceae) (Arnold et al., 2000); and liverworts (Davis et al., 2003). Endophytic Xylaria species have also been isolated from vascular plants in Europe (Brunner and Petrini, 1992; Taylor et al., 1999), Malaysia (Brunner and Petrini, 1992), the Brazilian Amazon (Rodrigues, 1994), Puerto Rico (Læssøe and Lodge, 1994; Lodge et al., 1996; Bayman et al., 1997; 1998), China (Taylor et al., 1999; Guo et al., 2000), Japan (Brunner and Petrini,

1992), Panama (Arnold *et al.*, 2000), and Thailand (Mekkamol, 1998; Charesprasert, 2001; Lumyong *et al.*, 2001; Ruchichakhon, 2004). In addition, there is some evidence that endophytic *Xylaria* species can be vertically transmitted through seeds of *Casuarina* as in mutualistic endophytes (Clavicipitales) (Bayman *et al.*, 1998). However, given their global range, the horizontal transmission of conidia or spores must also be very effective.

The production of secondary metabolites that are toxic to herbivores or pathogens is a common characteristic of many endophytic mutualisms and also provides the basis for selection favoring the symbiosis in the host plant (Carroll, 1988). *In vitro* studies of endophytic *Xylaria* species have shown that they actively produce secondary metabolites (Brunner and Petrini, 1992), and these may also be produced when the fungus inhabits living plant tissues. Such metabolites include antifungal and antibiotic compounds (Brunner and Petrini, 1992; Petrini *et al.*, 1995). The secondary compounds of the xylariaceous endophyte, *Muscodor albus* Worapong, Strobel & W.M. Hess, were experimentally shown to inhibit the growth of a broad range of plant and human pathogenic bacteria and fungi (Strobel *et al.*, 2001). There has been no research on how these important compounds may affect host ecology.

Accumulating evidence suggests that relationships between endophytic *Xylaria* and their hosts are complex. The further study of endophytic *Xylaria* species is needed to fully understand their ecology. Transplant and inoculation experiments are also needed to address the question of whether *Xylaria* is a mutualistic, antagonistic, or commensalistic endophyte.
# 2.2 Taxonomy of the Xylariaceae

The Xylariaceae is classified in Phylum Ascomycota, Class Pyrenomycetes, and Order Xylariales (Alexopoulos *et al.*, 1996). In the key to genera of Xylariaceae the number of genera is opened to discuss by mycologists depending on the criteria used in the taxonomy. Eriksson and Hawksworth (1993) recognised 35 genera whereas Læssøe (1994) proposed 37 genera with a few uncertain genera. Later, Whalley (1996) reviewed the family and listed 41 genera with Ju and Rogers (1996) accepting 39 genera. Recently four more new genera have been proposed. *Jumillera* J.D. Rogers, Y.-M. Ju & San Martín and *Whalleya* J.D. Rogers, Y.-M. Ju & San Martín have been separated from *Biscogniauxia* Kuntze (Rogers *et al.*, 1997). *Poroleprieuria* M.C. González, Hanlin, Ulloa et E. Aguirre, has been erected for a collection from Mexico (González *et al.*, 2004) and this is closely related to *Leprieuria* Læssøe, J.D. Rogers & Whalley. *Emarcea* Duong, R. Jeewon & K.D. Hyde has very recently been described as a new genus from Thailand containing a single species, *Emarcea castanopsidicola* (Duong *et al.*, 2004). Although there are different opinions, at least 42 genera can be assigned to the family (Table 3).

Thienhirun (1997) reported seventeen xylariaceous genera from Thailand, which were *Anthostomella*, *Astrocystis*, *Biscogniauxia*, *Camillea*, *Daldinia*, *Entonaema*, *Holttumia*, *Hypoxylon*, *Kretzschmaria*, *Kretzschmariella*, *Nemania*, *Podosordaria*, *Poronia*, *Rophalostroma*, *Rosellinia*, *Sacoxylon*, and *Xylaria*. Consequently, three more genera, *Jumillera*, *Stilbohypoxylon*, and *Whalleya* have been included (Thienhirun and Whalley, 2001) and now *Emarcea* was added (Doung *et al.*, 2004).

Eriksson and Hawksworth (1993)	Læssøe (1994)	Whalley (1996)	Ju and Rogers (1996)	Others
Anthostomella Sacc.	Anthostomella	Anthostomella	Anthostomella	
			Areolospora S.C. Jong &	
			E.E. Davis	
Ascotricha Berk.		? Ascotricha	Ascotricha	
? Ascotrichella Valldos.&Guarro		? Ascotrichella		
? Astrocystis Berk. & Broome	Astrocystis	Astrocystis		
Biscogniauxia Kuntze	Biscogniauxia	Biscogniauxia	Biscogniauxia	
Calceomyces Udagawa & S. Ueda	Calceomyces	Calceomyces	Calceomyces	
<i>Camillea</i> Fr.	Camillea	Camillea	Camillea	
	Chaenocarpus Fr.	? Chaenocarpus		
	<i>Collodiscula</i> I.Hino & Katum.	? Collodiscula	Collodiscula	
	Creosphaeria Theiss.	Creosphaeria	Creosphaeria	
Daldinia Ces. & De Not.		Daldinia	Daldinia	
			<i>Discoxylaria</i> Lindquist & J. Wright	
			-	Emarcea Duong, R.
				Jeewon & K.D. Hyde
				(2004)
Engleromyces Henn.	Engleromyces	Engleromyces	Engleromyces	
			Entoleuca Syd.	
Entonaema A. Møller	Entonaema	Entonaema	Entonaema	
	Euepixylon Füisting	? Euepixylon	Euepixylon	
Fassia Dennis				
Helicogermslita Lodha & D. Hawksw.	Helicogermslita	Helicogermslita		
	Holttumia Lloyd	? Holttumia		
Hypocopra (Fr.) J. Kickx f.	Hypocopra	Hypocopra	Hypocopra	
Hypoxylon Bull.	Hypoxylon	Hypoxylon	Hypoxylon	
Induratia Samuels, E. Mull. & Petrini	Induratia	Induratia	Induratia	
				Jumillera J.D. Rogers, Y
				M. Ju & San Martin
	V ( I ·	V I ·		(1997)
Kreizschmaria Fr.	Kretzschmarta	Kretzschmaria	Kreizschmaria Kustansku suislla Viézaz	
Lanviauria I magaa ID Bagara kWhallow	Lapriqueia	Lonviguria	<i>Kreizschmariella</i> viegas	
Longdostoma (Nitsohko) Travorse	Leprieuria	Leprieuria	Leprieuriu	
Lopadosioma (INIISCIKE) Traverso	2 Myzonacsia Virsehst	Lopaaosioma	Lopaaosioma	
	<i>i myconeesia</i> Kirschst.			

**Table 3.** The fungal genera within the Xylariaceae family.

Source: Whalley (1996); Ju and Rogers (1996); Rogers, Ju and San Martín (1997); Duong et al. (2004); González et al. (2004).

# Table 3. (Continued).

Eriksson and Hawksworth (1993)	Læssøe (1994)	Whalley (1996)	Ju and Rogers (1996)	Others
	Nemania Gray emend. Pouzar	Nemania	Nemania	
	Obolarina Pouzar	Obolarina	Obolarina	
? Paucithecium Lloyd				
Penzigia Sacc.		? Penzigia		
Phaeosporis Clem.	Phaeosporis	Phaeosporis		
Phylacia Léy.	Phylacia	Phylacia	Phylacia	
Podosordaria Ellis & Holw.	Podosordaria	Podosordaria	Podosordaria	
Poroconiochaeta Udagawa & Furuva				
				Poroleprieuria M.C.
				González, Hanlin, Ulloa et
				E. Aguirre, (2004)
Poronia Willd.	Poronia	Poronia	Poronia	0
Pulveria Malloch & Rogerson	(as <i>Pvrenomvxa</i> Morgan)	Pulveria	Pulveria	
Rhopalostroma D. Hawksw.	Rhopalostroma	Rhopalostroma	Rhopalostroma	
Rosellinia De Not	Rosellinia	Rosellinia	Rosellinia	
Sarcoxvlon Cooke	Sarcoxvlon	Sarcoxvlon	Sarcoxvlon	
	? Sevnesia Sacc.			
Stilbohvpoxvlon Henn.		? Stilbohvpoxvlon	Stilbohvpoxvlon	
Stromatoneurospora S.C. Jong & E.E. Davis	Stromatoneurospora	Stromatoneurospora	Stromatoneurospora	
Thamnomyces Ehrenb.	Thamnomyces	Thamnomyces	Thamnomyces	
Theissenia Maubl.	Theissenia	Theissenia	Theissenia	
Thuemenella Penz. & Sacc.	Thuemenella	Thuemenella	Thuemenella	
Ustulina Tul. & C. Tul.			Ustulina	
Versiomvces Whalley & Watling		Versiomyces	Versiomvces	
y y c		2	Vivantia J.D. Rogers, YM. Ju,	
			& Cand.	
Wawelia Namysl.	? Wawelia	Wawelia	Wawelia	
2				Whalleya J.D. Rogers, Y
				M. Ju & San Martín
				(1997)
Xylaria Hill ex Schrank	Xylaria	Xylaria	Xylaria	. ,

Source: Whalley (1996); Ju and Rogers (1996); Rogers, Ju and San Martín (1997); Duong et al. (2004); González et al. (2004).

#### 2.2.1 Morphological taxonomy

Principally, the xylariaceous fungi have been characterised mainly on conventional methods regarding teleomorphic and anamorphic characteristics by using macroscopy and microscopy (Eriksson and Hawksworth, 1993; Læssøe, 1994; Rogers, 1994; Ju and Rogers, 1996; Whalley, 1996). Additionally, chemical characteristics have been accepted or widely used in fungal taxonomy (Whalley and Edwards, 1987; Whalley and Edwards, 1995; Stadler *et al.*, 2001; Stadler, Ju, and Rogers, 2004).

#### 2.2.1.1 Teleomorphic characteristics

#### A) Stromata

The stromatal characters of Xylariaceae are extremely variable in shape, size, and colour. They range from applanate, erumpent, effused, subglobose to globose, uniperitheciate, and upright forms. The flattened applanate and erumpent forms are found in the genera *Biscogniauxia*, *Jumillera*, *Whalleya*, and a few *Camillea* species (Figure 1). The superficial and widely effused types occur in *Nemania* and many taxa belonging to the genus *Hypoxylon*. Whereas subglobose to globose forms have been found in *Daldinia* and some species of *Hypoxylon*, and the uniperitheciate stroma is generally restricted to *Rosellinia* and *Astrocystis* (Figure 1). In *Xylaria*, *Kretzschmaria*, and *Rhopalostroma*, stromata are upright but some *Camillea* species, e.g. *C. leprieurii* Mont., have dimorphic forms, which are applanate or erect. The texture of stromata has also been emphasized being defined as hard, fairly hard, woody, and soft. However, these variations of features could result from environmental influences. In addition, moisture and light might affect pigmentation or degree of branching whilst host types or surface shapes of the substratum may

influence growth form (Miller, 1961; Rogers, 1979; Ju and Rogers, 1996; Whalley, 1996).

The colour of the stromatal surface is also an important feature in many species. Their coloration can, however, vary with age and environmental conditions (Miller, 1961). Thus, some species exhibit different stromatal colour depending on the stage and locality of the fungal growth. However, the stromatal colour has been proven to be more useful in the delimitation of taxa above species level with the application of KOH or ethyl acetate extractable pigments in *Daldinia* and *Hypoxylon* proving to be of taxonomic value (Martin, 1968; Greenhalgh and Whalley, 1970; Whalley and Greenhalgh, 1973; Whalley and Whalley, 1977; Ju and Rogers, 1996; Ju *et al.*, 1997; Stadler *et al.*, 2001; Stadler, Ju, and Rogers, 2004).

#### **B)** Perithecia

The perithecial characters of xylariaceous fungi are usually described as globose, ovoid, and obovoid to tubular (Figure 2). Their degree of protruding may be recorded as completely immersed, partially immersed or almost free. Their arrangement may be monostichous or polystichous, and they vary considerably in their dimensions (Luttrell, 1951; Rogers and Berbee, 1964; Mai, 1977; Rogers, 1967). The size of perithecia has been considered in combination with other characters. The characters of perithecia vary in detail at the species level, and might provide useful additional taxonomic information in the family (Jensen, 1985).



Figure 1. Stromata of the xylariaceous fungi; (a) *Camillea heterostromata* (Mont.) Læssøe, J.D. Rogers & Whalley (applanate form), (b) *Biscogniauxia schweinitzii* Y.-M. Ju & J.D. Rogers (applanate form), (c) *Hypoxylon bovei* Speg. (subglobose to globose form), (d) *Hypoxylon fusoideosporum* Y.-M. Ju & J.D. Rogers (subglobose to globose form), (e) *Rosellinia corticium* (Schwein.: Fr.) Sacc. (uniperitheciate form), (f) *Daldinia concentrica* (Bolt.: Fr.) Ces. & De Not. (subglobose to globose form), (g) *Kretzschmaria clavus* (Fr.: Fr.) Sacc. (upright form), (h) *Camillea leprieurii* Mont. (upright form), and (i) *Xylaria schweinitzii* (Berk. & M.A. Curtis) (upright form).

Source: Ju and Rogers (1997).



Figure 2. Perithecia of the xylariaceous fungi; (a) *Biscogniauxia schweinitzii* Y.-M. Ju & J.D. Rogers (tubular), (b) *B. dennisii* (Pouzar) Piuzar (tubular), (c) *Camillea leprieurii* Mont. (long spherical), (d) *C. bilabiata* Speg. (tubular), (e) *Hypoxylon chathamense* Y.-M. Ju & J.D. Rogers (spherical), (f) *H. hypomiltum* Mont. (obovoid to tubular), (g) *Nemania aenea* (Nitschke) Pouzar var. *macrospora* (J.H. Miller) Y.-M. Ju & J.D. Rogers (obovoid), (h) *N. serpens* (Pers.: Fr.) S.F. Gray var. *colliculosa* (Schwein.: Fr.) Y.-M. Ju & J.D. Rogers (obovoid), and (i) *H. fuscum* (Pers.: Fr.) Fr. (spherical).

Source: Ju and Rogers (1997).

#### C) Ostioles

The ostiole characters of xylariaceous fungi consist of two types, the umbilicate ostiolum and the papillate ostiolum. The umbilicate ostiolum is characterised by small circular depressions in the stroma which appear flush with the stromal surface. Umbilicate ostioles are found mainly in species belonging to Hypoxylon section Hypoxylon (Miller, 1961; Ju and Rogers, 1996) and in representatives of many other genera (Figure 3). An umbilicate ostiolum occurring sunken as in Biscogniauxia and some Camillea species is often termed punctate. In contrast, the papillate ostiolum is elevated above the surface of the stroma and, thus, appears as a small nipple-like projection. Papillate ostioles are found in most species of Nemania, Kretzschmaria, many species of Xylaria, and some species of Biscogniauxia (Figure 3). In several members of the section Annulata of Hypoxylon (Ju and Rogers, 1996) the papillate ostiolum is surrounded by a circular depression or disk which has been found to occur as the result of the sloughing off of the stromal surface in this region (Abe, 1986). Ju and Rogers (1996) recognised a bovei-type where the whole disk area is shed in one piece and the *truncatum*-type where the surface is gradually worn away to form the disk. The annulate ostiolum is also found in some species of Nemania, Kretzschmaria, and Xylaria. Therefore, the ostiolar type is an important taxonomic character in *Hypoxylon* and other xylariaceous genera.



Figure 3. Ostioles of the xylariaceous fungi; (a) *Biscogniauxia dennisii* (Pouzar)
Piuzar (papillate), (b) *B. reticulospora* Y.-M. Ju & J.D. Rogers (papillate),
(c) *Hypoxylon bovei* Speg. (papillate with disk), (d) *H. kretzschmariodes*Y.-M. Ju & J.D. Rogers (umbilicate), (e) *H. hypomiltum* Mont. (umbilicate with white substance), and (f) *Xylaria melanura* (Lév.) Sacc. (papillate).
Source: Ju and Rogers (1997).

#### **D)** Ascus and apical apparatus

Asci of most xylariaceous fungi usually contain eight spores expect *Wawelia* which has four spores (Minter and Webster, 1983; Lundqvist, 1992) and *Thuemenella* which has six spores (Samuels and Rossman, 1992). In general, the xylariaceous ascus is unitunicate, cylindrical, and terminates below in a short or long stipe. The ratio of the spore bearing part to the stipe is sometimes taxonomically useful such as in *Biscogniauxia* where the stipes are typically short. In *Xylaria*, *Kretzschmaria*, and *Nemania*, the stipes are invariably long.

The apical tip is usually rounded, and encloses an apical apparatus, which is usually amyloid stained blue in Melzer's iodine reagent (Figure 4). Some are occasionally reddish (dextrinoid) and some do not react visibly with iodine. The significance of the iodine reaction on the apical apparatus has been discussed by Eriksson (1966), Kohn and Korf (1975), and Nannfeldt (1976). The shape and size of the apical apparatus are one of the more important taxonomic features in the Xylariaceae (Munk, 1957; Carroll, 1963; Martin, 1969; Krug and Cain, 1974; Francis, 1975; Rogers, 1979; Læssøe et al., 1989; Ju and Rogers, 1996; Whalley, 1996). There are at least five types of apical apparatus which can be recognised. Firstly, an apical apparatus is constructed from stacks of smaller rings found in Hypocopra and Poronia (Krug and Cain, 1974; Jong and Rogers, 1969). Secondly, it is flattened and appears broader than high as in most species of Hypoxylon s. str. and Daldinia (Ju and Rogers, 1996; Ju, Rogers, and Martin, 1997) (Figure 4). Thirdly, it is discoid as found in *Biscogniauxia* (Ju and Rogers, 1996; Ju, Rogers, and González, 1997; Martin, 1967). Fourthly, it is rhomboid or diamondshaped but only in Camillea (Læssøe et al., 1989). Finally, it is higher than broad, often constricted sub-apically to appear uniform or inverted hat-shaped and is generally characteristic for Xylaria, Rosellinia, Kretzschmaria, and Nemania (Martin, 1967; Rogers, 1979; Van der Gucht, 1995; Whalley, 1996).



Figure 4. The apical apparatus forms of the xylariaceous fungi; (a) Nemania aenea (Nitschke) Pouzar var. macrospora (J.H. Miller) Y.-M. Ju & J.D. Rogers (higher than broad), (b) Hypoxylon rubiginosum Pers.: Fr. (broader than high), and (c) Camillea tinctor (Berk.) Læssøe, J.D. Rogers & Whalley (higher than broad).

Source: Ju and Rogers (1997).

#### E) Ascospores and germination slit

Ascospores of most xylariaceous fungi are usually described as single cell, with a smooth wall, light to dark brown in colour, with a conspicuous full-length germ slit (Rogers, 1979). In general, the ascospores are arranged in a single row within the ascus being uniseriate or obliquely uniseriate.

Most spores are subglobose, ellipsoid, oblong, fusiform, inequilaterally ellipsoid (where one side is flat to slightly concave and the other side is curved) to broadly crescentric, with ends either narrowly or broadly rounded, attenuated or apiculate. Subglobose, ellipsoid, oblong or fusiform ascospores are most common in the genera *Biscogniauxia* and *Camillea* whilst inequilateral ellipsoid spores are characteristics of the genera *Daldinia* and *Hypoxylon* s. str. Broadly crescentric spores are usual found among members of the genera *Kretzschmaria* and

#### Xylaria.

The colour of ascospores ranges from light brown to medium brown to dark brown, sometimes appearing almost black. Ascospores of *Camillea* are pale yellow or almost colourless. They lack germ slits or pores but they are characterised by ornamented spore walls readily seen by scanning electron microscope (SEM). Although most ascospores are mainly smooth, some are ornamented. The ornamentation in the genus *Camillea* varies from poroid, reticulate, ribbed, and echinulate-verrucose. Striate ascospore ornamentation has been found in members of the genus *Stromatoneurospora* (Jong and Davis, 1973), *Nemania chestersii* (Rogers & Whalley) Pöuzar (Rogers and Whalley, 1978), and *Biscogniauxia weldenii* (J.D. Rogers) Whalley & Læssøe (Rogers, 1977). There are also faint transverse striations oriented perpendicular to the long axis of the spores in some species of *Hypoxylon* s. str. section *Hypoxylon* (Rogers and Candoussau, 1982; Rogers, 1985; Van der Gucht and Van der Veken, 1992) and *Daldinia* (Van der Gucht, 1993). Thus, the spore ornamentation has been proven to be very useful in some xylariaceous species but SEM is required to observe it in most taxa.

Some species have a conspicuous hyaline outermost wall layer called the exospore (Child, 1932; Pouzar, 1979) or perispore (Rogers, 1965; 1969). These outer walls are commonly characteristic among *Hypoxylon* s. str. and *Daldinia* (Rogers, 1965; Beckett, 1976; Ju and Rogers, 1996). Whether the perispore is dehiscent or indehiscent in 10% KOH, smooth or ornamented is considered very useful taxonomic data at the species level in *Hypoxylon* s. str. (Ju and Rogers, 1996).

Most xylariaceous fungi have ascospores with germ slits, and germ slit forms have been recognised by most investigators (Vincens, 1918; Carrol, 1963; Krung and Cain, 1974; Martin, 1967; Rogers, 1979; Whalley, 1996). The germ slit is a constant and diagnostic feature of many well-circumscribed species. The important characters in describing the germ slit are shape, position on the spore, orientation along the long axis of the spore, and length. The germ slit can be straight, curved, spiraling, and even dotted. In Hypoxylon germ slits are usually straight but sometimes sigmoid (Figure 5). A few species of Hypoxylon exhibit peculiar germ slits. Some ascospores appear lacking germ slits or pores such as species of Camillea (Rogers, 1977b; Læssøe et al., 1989), Stromatoneurospora (Jong and Davis, 1978) and Nemania chestersii (Rogers and Whalley, 1978). The germ slit can be found on the ventral (concave) side such as the members of the genera Nemania, Kretzschmaria, and Xylaria, or the dorsal (convex) side such as in Daldinia and Hypoxylon s. str. The orientation can be oblique or parallel to the long axis of the spore. The length is short (less than spore length), or long (essentially the length of the spore). The germ slit, when present, is assumed to act as a site for germination providing an easy exit point for the germ tube and it may also facilitate uptake of water and nutrients. Ascospores lacking germ slits may have less elaborate wall structure or, alternatively, might have germination sites in the wall that are not obvious. The germ slit is a fissure in several of the inner wall layers which remain covered by the outermost layers until germination begins (Beckett, 1976).



Figure 5. Germ slit characters of the xylariaceous fungi. (a) *Biscogniauxia anceps* (Sacc.) J.D. Rogers, Y.-M. Ju & Cand (straight form) from Ju and Rogers (1997), (b) *Hypoxylon purpureonitens* Y.-M. Ju & J.D. Rogers (SUT004) (straight form), and (c) *Xylaria* sp. SUT155 (spiral form).

### 2.2.1.2 Anamorphic characteristics

Most species of xylariaceous fungi form anamorphs or an asexual stage. They are characterised by conidia which are holoblastically. They are usually pigmented, and have a broad, circular, flat to truncate base. The anamorphic characteristics of xylariaceous species have been proven valuable in closely related species, and were first reported by Chesters and Greenhalgh (1964). However, the major problem is the inability to obtain anamorphic cultures because the teleomorphic material might not be fresh and in good condition. The anamorphs can develop on the external surfaces of immature or maturing stromata or in close association with them. They are four major characteristics to their growth form.

Firstly, the anamorph develops on immature or mature stromata, or on the wood lying in close proximity to the stromata. The anamorph usually appears as a powdery layer, yellowish gray, gray or brown. The conidiophores develop either monematously (freely) or in a few cases they develop on synnemata. This type of growth form is commonly found in members of the genera *Biscogniauxia*, *Daldinia*, *Hypoxylon*, and *Nemania* (Chesters and Greenhalgh, 1964; Greenhalgh and Chesters, 1968; Jong and Rogers, 1972; Petrini and Müller, 1986).

Secondly, the anamorph develops on immature stromata covering the whole or a part of the surface or developing on specialized structures of the immature stromata. The conidiophores are organized in a dense regular palisade layer. This is the common form found in *Xylaria* (Rogers, 1985) and *Kretzschmaria* (Van der Gucht, 1995).

Thirdly, the anamorph develops separately from the stromata usually on distinctive structures. It is always produced earlier in the growing season than the stromata. The conidiophores form dense palisade layers. This growth form is commonly found in certain *Xylaria* species such as *X. cubensis* with its accompanying *Xylocoremium flabelliforme* (Schwein.: Fr.) J.D. Rogers state (Rogers, 1984; 1985) and *X. poitei* (Lév.) Fr. (Rogers and Callan, 1986).

Fourthly, the anamorph develops superficially on bamboo culms, and consists of a central cone of hyaline thin walled conidiophores. They are arranged in a compact palisade layer, which terminates apically in denticulate conidiogenous cells and are surrounded by sterile carbonaceous tissue. The perithcia develop beneath the conidiome and grow through it. And frequently the remnants of the conidiome persist as a rough ring on the perithecial stromata giving a stellate appearance as in *Astrocystis*. This type of anamorph also occurs in *Collodiscula* and the anamorphs have been assigned to the form-genus *Acanthodochium* Samuels, J.D. Rogers & Nagasawa (Samuels, Rogers, and Nagasawa, 1987; Ju and Rogers, 1990).

All xylariaceous anamorphs have hyaline to light brown conidiophores, and vary in the type of branching and development of the conidiogenous cells. The conidiophores are characterised by the manner of branching and the position of the conidiogenous cells. The relationship between anamorph and teleomorph is shown in Table 4.

Teleomorph	Anamorph
Anthostomella	Geniculosporium Chesters & Greenh. (Martin 1969,=Nodulisporium
	type 2a), Nodulisporium Preuss and Virgariella S. Hughes (Francis,
	Minter, and Caine, 1980)
?Ascotricha	Dicyma Boulanger (Hawksworth, 1971)
?Ascotrichella	?Humicola-like (Valdosera and Guarro, 1988)
Astrocystis	Acanthodochium Samuels, J.D. Rogers & Nagas. (Samuels, Rogers,
	and Nagasawa, 1987; Ju and Rogers, 1990)
Biscogniauxia	Geniculosporium (Eckblad and Granmo, 1978; Whalley and Edwards,
	1985), Nodulisporium (Greenhalgh and Chesters, 1968; Callan and
	Rogers, 1986; González and Rogers, 1993), Periconiella (Petrini and
	Müller, 1986)
Calceomyces	Nodulisporium (Udagawa and Ueda, 1988)
Camillea	Xylocladium Syd. (Crane and Dumont, 1975; Læssøe, Rogers, and
	Whalley, 1989; González and Rogers, 1993)
?Chaeocarpus	Unknown
Collodiscula	Acanthodocium (Samuels, Rogers, and Nagasawa, 1987)
Daldinia	Nodulisporium (Chesters and Greenhalgh, 1964; Petrini and Müller,
	1986)
Engleromyces	Unknown
Entonaema	Nodulisporium (Rogers, 1982)
Euepixylon	Geniculosporium (Whalley, 1976)
Helicogermslita	Unknown
Holttumia	Unknown
Hypocopra	Unknown
Hypoxylon	Nodulisporium, Virgariella, Hadrotrichum Fuckel, Rhinocladiella
	Nannf. (Martin, 1967; Greenhalgh and Chesters, 1968; Jong and
	Rogers, 1972; Petrini and Müller, 1986)
Induratia	Nodulisporium (Samuels, Müller, and Petrini, 1987)
Jumillera	Unknown

**Table 4.** The anamorph-teleomorph relationship within genera of the Xylariaceae.

Table 4. (Continued).

Teleomorph	Anamorph	
Kretzschmaria	Hadrotrichum (Petrini and Müller, 1986)	
Leprieuria	Geniculosporium (Samuels and Müller, 1980)	
Lopadostroma	Scolecosporous anamorph, Libertella-like (Ju, González, and	
	Rogers, 1993)	
Nemania	Geniculosporium (Chesters and Greenhalgh, 1964; Petrini and	
	Müller, 1986)	
Obolarina	Rhinocladiella-like (Candoussau and Rogers, 1990)	
?Penzigia	Unknown	
Phaeosporis	Sporothrix Hektoen & C.F. Perkins (Jong and Davis, 1974)	
Phaeosporis	Sporothrix Hektoen & C.F. Perkins (Jong and Davis, 1974)	
Phylacia	Geniculosporium (Rodrigues and Samuels, 1989)	
Podosordaria	Lindquistia Subram. & Chandrash.(Subramanian and	
	Chandrashekara, 1977; Rogers and Læssøe, 1992)	
Poroleprieuria	Unknown	
Poronia	Lindquistia (Subramanian and Chandrashekara, 1977; Stiers,	
	Rogers, and Russell, 1973)	
Pulveria	Unknown	
Rhopalostroma	Nodulisporium (Hawksworth and Whalley, 1985)	
Rosellinia	Geniculosporium, Dematophora R. Hartig, Nodulisporium (Petrini,	
	1992)	
Sarcoxylon	Unknown	
Seynesia	Acanthodochium (Hyde, 1995)	
Stilbohypoxylon	Unknown	
Stromatoneurospora	Unknown	
Thamnomyces	Nodulisporium (Samuels and Müller, 1980)	
Theissenia	Unknown	
Theumenella	Nodulisporium (Samuels, 1989; Samuels and Rossman, 1992)	
Theumenella	Nodulisporium (Samuels, 1989; Samuels and Rossman, 1992)	
Versiomyces	Unknown	
Wawelia	Anamorph described by Minter & Webster (1983) as being	
	geniculate but not assigned to a form genus	
Whalleya	Unknown	
Xylaria	Typically produced on developing stromata but no form genus yet	
	assigned Xylocoremium flabelliforme (Schwein.: Fr.) J.D. Rogers is	
	associated with X. cubensis (Rogers, 1984, 1985)	

Source: Whalley (1996); Ju and Rogers (1996); Rogers, Ju, and San Martín (1997); Duong *et al.* (2004); González *et al.* (2004).

Conidia of most xylariaceous fungi are all morphologically similar, and exhibit little variation except for moderate differences in overall shape and size. They are unicellular, subglobose, obovoid to ellipsoid, hyaline to light brown, and usually smooth. Since they are produced holoblastically, they all possess a basal scar indicating the former site of attachment to the conidiogenous cell (Greenhalgh, 1967; Stiers *et al.*, 1973; Koehn and Cole, 1975).

#### 2.2.2 Chemical taxonomy

Although secondary metabolites have not been accepted or widely used in fungal taxonomy, they are now known to be useful in the taxonomy of *Penicillium* (Frisvad and Samson, 1991; Lund and Frisvad, 1994; Frisvad *et al.*, 1998), *Aspergillus* (Kozakiewicz, 1994), *Fusarium* (Onji, Aoki, and Tani, 1994), and lichens (Culberson and Culberson, 1994). Fungal secondary metabolites have a great diversity of molecular structures, and frequently show taxonomic specificity in their production which usually occurs during the stationary phase of growth or the idiophase (Bulock, 1980; Whalley and Edwards, 1999).

In the Xylariaceae, there are several genera reported to produce pigments or other secondary metabolites in their stromata and cultures. Ju and Rogers (1996) characterised many species of genera of Xylariaceae with *Nodulisporium*-like anamorphs by conspicuous colours of their fruit bodies such as in *Hypoxylon* and *Daldinia* which extracted stromatal pigment colours in 10% KOH and employed as key features. Van der Gucht (1994) also used colours of organic extracts made with solvents such as acetone to include in species descriptions. These colours of extracted pigments are determined by comparison with a standard chart (Rayner, 1970). The concentration of colour-extracted pigments may vary with age and stages of preservation, however, they usually still contain the same metabolites found in young and fresh specimens, albeit at lower concentrations. In any case, the Xylariaceae is quite creative when it comes to the production of chemical diversity, hence their stromatal pigment colours usually result from the presence of a mixture of several metabolites. Pigments and other secondary metabolites can be separated according to their polarity, and detected as single component. The profile of secondary metabolites can be investigated using chromatographic methods such as thin layer chromatography (TLC) on *Hypoxylon* (Whalley and Whalley, 1977) and ultra-violet light or high performance liquid chromatography and diode array detection (HPLC-DAD) on *Daldinia, Entonaema, Rhopalostroma*, and other xylariaceous fungi (Andersen *et al.*, 2001; Stadler *et al.*, 2001; Mühlbauer *et al.*, 2002; Quang *et al.*, 2002; Stadler *et al.*, 2004).

The Xylariaceae has been shown to produce a large number of secondary metabolites which can be grouped as butyrolactones, dihydroisocoumarins, succinic acid, cytochalasins, and other compounds. These metabolites have been used to demonstrate the possible phylogenetic relationships (Whalley and Edwards, 1987). *Daldinia concentrica* was found to contain 4, 9-dihydroxyperylene quinone in its ascocarps (Allport and Bu'lock, 1958) whilst 1, 8-dimethyoxynaphthalene and its corresponding ether were produced in culture broth (Allport and Bu'lock, 1960). During the same period, Chen (1960; 1964) isolated rosellinic acid and diketopiperazine from cultures of phytopathogenic *Rosellinia necatrix*, and subsequently it was found to produce cytochalasin E (Aldridge *et al.*, 1972). Engleromycin, an epoxide of cytochalasin D, was later isolated from the xylariaceous

taxon, Engleromyces goetzii P. Henn. (Pedersen et al., 1980). Hypoxylon fragiforme was found to owe its orange to brick red stromatal colour to mitorubrin and its derivatives (Steglich et al., 1974) whilst Xylaria polymorpha Pers. produces a hydroxyphthalide derivative, xylaral, which develops a violet purple colour reaction with aqueous ammonia (Gunawan et al., 1990). Extensive studies have resulted in the characterisation of many secondary metabolites from a range of representatives of the family, and have demonstrated a remarkable diversity of chemical compounds produced. A considerable number of these metabolites have proven to be new (Whalley and Edwards, 1995). Most of metabolites produced by the representatives investigated can be grouped as dihydroisocoumarins and derivatives (Anderson et al., 1983), succinic acid and derivatives (Anderson et al., 1985), butyrolactones (Edwards and Whalley, 1979; Anderson et al., 1982), cytochalasins (Edwards et al., 1989), sesquiterpene alcohols (punctaporonins) (Edwards et al., 1988; Edwards et al., 1989), griseofulvin and griseofulvin derivatives (Whalley and Edwards, 1995), naphthalene derivatives (Whalley and Edwards, 1995), and long chain fatty acids (Adeboya et al., 1995).

Generally, the presence of these compounds can be seen to be closely related to systematic position, and the chemical data has proved invaluable in recognising associations between species and genera (Whalley and Edwards, 1995; Whalley, 1996). The dihydroisocoumarins are widely distributed throughout the family but they are probably more representatives of *Hypoxylon*, *Biscogniauxia* and *Camillea* (Whalley and Edwards, 1995). Butyrolactones, so far, appear to be restricted to *Nemania serpens* (Pers.: Fr.) Pouzar whilst cytochalasins are frequently encountered in species of *Xylaria*, *Rosellinia*, and members of the defunct section Primocinerea of Hypoxylon (Whalley, 1996).

Dreyfuss (1986) reported new cytochalasins in endophytic *Xylaria* species from tropic plants. A relationship between the production of some secondary metabolites, e.g. cytochalasins, and the phytopathogenicity of the isolates cannot be excluded (Whalley and Edwards, 1999). On the other hand, the production of secondary metabolites is increasingly used to clarify the taxonomic position of fungal taxa.

#### 2.2.3 Molecular taxonomy

Since the morphological characteristics of several fungal genera are frequently too limited to allow its identification, the molecular techniques are applied. The group of genes, which is most frequently targeted for phylogenetic analysis, is the ribosomal RNA genes (or rDNA). In addition, introns of several protein-encoding genes, such as the  $\beta$ -tubulin (O'Donnell, 1992; Tsai *et al.*, 1994), actin (Cox *et al.*, 1995), chitin synthase (Bowen *et al.*, 1992; Szaniszlo and Momany, 1993), acetyl coenzyme A synthase (Birch, Sims, and Broda, 1992), glyceraldehydes-3-phosphate dehydrogenase (Harmsen *et al.*, 1992), or orotidine 5'-monophosphate decarboxylase genes (Radford, 1993), can also be applied, and can provide the valuable information of molecular taxonomy.

#### 2.2.3.1 Ribosomal DNA

Ribosomal DNA (rDNA) is widely used for the inference of phylogenetic relationships because it is present in all living organisms, and different rates of evolution in different regions. Therefore, it makes rDNA useful for studies at different taxonomic levels (Bruns *et al.*, 1991). The sequences coding for nuclear ribosomal RNA (rDNA) have been chosen in many studies of phylogenetic systematics and evolutionary patterns of fungi (Okada et al., 1997). The fungi and most eukaryotes contain 80S ribosomes, which consist of two subunits, the large (60S) and small (40S) subunits. Each subunit consists of rRNA as a structural molecule and a number of associated proteins. The large subunit contains 28S, 5.8S and 5S rRNA molecules and the small subunit contains 18S rRNA molecule (Figure 6). Genes coding for rRNA are suitable signal molecules as the synthesis of ribosomes has been strongly conserved over evolution, due to the central role of ribosomes in gene expression. The rRNA genes for the rRNA subunits, although not varying greatly in length, contain both strongly conserved and variable regions within their sequences (Van de Peer, Chapelle, and Wachter, 1996). The genes for these rRNA molecules are also separated by the two external transcribed spacers (ETS) and the nontranscribed spacer regions (NTS), which contain the signals for rDNA expression (Figure 6). Both spacers are mainly called the intergenic spacer (IGS). The regions that lie between these RNAs are the two noncoding internal transcribed spacers (ITS1 and ITS2) (Hwang and Kim, 1999). The nucleotide sequences of the rDNA repeat unit have been detected by designed primers according to the highly conserved 18S and 28S regions (White et al., 1990). The most detailed information can be obtained by direct sequencing of the PCR products, which detect every single base-pair difference of the amplified fragment between different samples. Phylogenetic analysis using sequence data combined with mating compatibility studies has shown more promise for resolving phylogenetic relationships and understanding speciation for problematic species complexes in fungi (Bruns et al., 1991).



**Figure 6.** Schematic diagram of a tandem repeat unit of rDNA. Source: Hwang and Kim (1999).

#### A) Small-subunit ribosomal DNA (SSU rDNA)

The nuclear SSU rDNA (18S rRNA gene in eukaryote) is one of the most highly conserved DNA regions, and the size is approximately 1,800 bp (White *et al.*, 1990). The sequence analysis of 18S in most filamentous fungi has been used completely or over 600 bp in subunit. In particularly, the SSU has been studied to reconstruct deep phylogenetic branches that include kingdoms, phyla, classes, or orders (Field *et al.*, 1988; Abele *et al.*, 1989; Friedrich and Tautz, 1995; Aguinaldo *et al.*, 1997; Whiting, 1998).

#### B) 5.8S ribosomal DNA

The degree of nucleotide conservation of 5.8S rDNA, which is the smallest nuclear rDNA of the cluster, is similar to that of SSU rDNA, but its length (approximately 150 bp) is too short to contain enough phylogenetic information. Due to the short length in DNA sequence, it is not advisable to use the 5.8S rDNA region for phylogenetic reconstruction (Hwang and Kim, 1999).

#### C) Large-subunit ribosomal DNA (LSU rDNA)

Nuclear LSU rDNA is much larger than SSU rDNA approximately > 4,000 bp, and shows more variation in the rate of evolution of its different domains compared to the SSU rDNA. It has many divergent domains or expansion segments, so the size of the gene varies considerably among phyla. Nuclear LSU rDNA is known to be useful for examining phylogenetic relationships in slightly low categorical levels such as orders or families (Friedrich and Tautz, 1997; Hwang *et al.*, 1998; Whiting, 1998).

# D) The intergenic sequence (IGS) and the internal transcribed spacer (ITS) regions

Ribosomal DNA spacer regions, IGS and ITS, have been employed to resolve phylogenetic problems in lower categorical levels among genera, species, or populations (Morgen and Blair, 1998; Navajas *et al.*, 1998; Perera *et al.*, 1998). The size of IGS (approximately 4-5 kb) is far larger than those of ITS region (approximately 1 kb). In fungi, the ITS region is often between 600 and 800 bp in length. The ITS region, as well as the intergenic NTS repeat, shows much evolutionary change. Differences in these regions occur between species within a genus (Goosen and Debets, 1996). Several studies have demonstrated that the ITS region is often highly variable among morphologically distinct fungal species, but the intraspecific variation is low in most cases (Gardes and Bruns, 1991; Lee and Taylor, 1992). Due to the large size of the IGS, the ITS regions have been preferred to IGS in phylogenetic approach. However, the IGS has been used in restriction fragment length polymorphism (RFLP) of entire rDNA arrays (Wheeler, 1989). In ITS region from several distantly related evolutionary groups, the variation often consists of tandem arrays of repeat motifs of up to 10-bp length (Gonzalez *et al.*, 1990; Lee and Taylor, 1992; Vogler and DeSalle, 1994). These short repeat motifs are believed to be caused by slipped-strand mispairing or replication slippage (Levinson and Gutman, 1987; Li and Graur, 1991). The processes involve intra-helical mispairing during DNA replication, which results insertion or deletion of bases. The short repeat motifs derived from this process have also been observed in rpoC2, a plastid gene encoding the  $\beta$ " subunit of RNA polymerase in grasses (Cummings *et al.*, 1994). Once an array of repeat motifs has been established, it becomes increasingly prone to additional slipped-strand mispairing events and, thus, accumulation of repeats.

The appropriate region for phylogenetic analysis is very important. Most of such misuses are caused by the lack of understanding of properties of molecular markers or gene regions by the negligence in the categorical levels examined. The selection of molecular markers or gene regions is necessary because the selection of inappropriate molecular makers or gene regions can not explain the correctly phylogenetic relationships. For instance, for studies of relationships among closely related species, the use of nuclear rRNA coding regions (such as nuclear SSU, LSU, 5.8S rDNA) can be problematic, whereas nuclear rDNA spacers such as IGS or ITS appear to have fewer problems because of their higher variation. On the other hand, for deep levels of divergence, the proteins coding genes are saturated at the amino acid level, and highly conserved regions of rDNA are useful. Hwang and Kim (1999) summarized the appropriate categorical levels of commonly used molecular markers or gene regions in rDNA (Table 5). **Table 5.** The applicable categorical levels of each molecular marker or gene region

 in molecular taxonomic study. The bold lines indicate mainly applicable

 categorical levels of each molecular marker or gene region while the dot

 lines indicate less frequently applicable categorical levels



Source: Hwang and Kim (1999).

#### 2.2.3.2 Phylogenetic study

Phylogenetics is the area of research concerned with finding the genetic relationships between species (Baldauf, 2003). The basic idea is to compare specific features of the species, under the natural assumption that similar species (i.e. species with similar characters) are genetically close. The classic phylogenetics used mainly with physical or morphological features, whilst the modern phylogeny uses information extracted from genetic material mainly DNA and protein sequences. Phylogenetics is sometimes called cladistics because the word "clade", a set of descendants from a single ancestor, is derived from the Greek word for branch. Intuitively, the phylogenetic trees are drawn from the ground up like real trees (Figure 7a). However, as these trees get larger and more complex, they can become cluttered and difficult to read. As an alternative, the nodes can be expanded (Figure 7b), and turned the tree on its side (Figure 7c). Then, the tree grows left to right, and all the labels are horizontal. This makes the tree easier to read and to annotate. Thus, the widths of the nodes have no meaning. They are simply adjusted to give even spacing to the branches. To make things slightly more complicated, all branches can rotate freely about the plane of their nodes, then, all trees in Figure 7 are identical (Baldauf, 2003). Molecular phylogenetic trees are usually drawn with proportional branch lengths, which is the lengths of the branches correspond to the amount of evolution (roughly, percent sequence difference) between the two nodes they connect (Figure 7a-f). Thus, the long branches are more divergent than the one attached to short branches. Alternatively, trees can be drawn to display branching patterns only (cladograms), in which case the lengths of the branches have no meaning (Figure 7g), but this is rarely done with molecular sequence trees (Baldauf, 2003).

In the tree construction from nucleotide sequences, the first step is building the dataset. This means finding and retrieving sequences from the public domain. The main repository for these data is the public nucleotide database such as GenBank (U.S.A.), EMBL (Europe), and DDBJ (Japan) (Baldauf, 2003). Then, the next step is sequence alignment, which is the heart of the matter. The role of sequence alignment is to organize sequences so that homologous residues appear in the same column of the alignment. This is a relatively straightforward task for regions that have a highly conserved sequence. Regions of sequences that cannot be unambiguously aligned are normally not included in phylogenetic analyses.



**Figure 7.** Phylogenetic tree styles. All these trees have identical branching patterns. Source: Baldauf (2003).

The common program that has been widely used in multiple sequence alignment is CLUSTAL, which is freely available for use on all major computer platforms (Higgins *et al.*, 1998). This program takes an input set of sequences, and calculates a series of pairwise alignments, comparing each sequence to every other sequence, one at a time.

For phylogenetic tree construction, methods for calculating the trees fall into two general categories (Page and Holmes, 1998). These are distance-matrix methods, also known as clustering or algorithmic methods (e.g. the unweighted pair group method using arithmetric averages (UPGMA), neighbourjoining, Fitch-Margoliash), and discrete data methods, also known as tree searching methods (e.g. parsimony, maximum likelihood, Bayesian methods) (Page and Holmes, 1998; Graur and Li, 1999; Nei and Kumar, 2000; Baldauf, 2003). Distance is relatively simple and straightforward. The distance (roughly, the percent sequence difference) is calculated for all pairwise combinations of OTUs (operational taxonomic units), and then the distances are assembled into a tree. Discrete data methods examine each column of the alignment separately and look for the tree that best accommodates all of this information. The programs for phylogenetic construction are examples of PHYLIP, Mega, and PAUP\*, which are the most comprehensive and widely used (Felsenstein, 1985; Hall, 2000; Swofford, 1999).

However, the methods of phylogenetic tree construction may assign organisms incorrectly to positions along a phylogenetic tree as a result of "false identity" in sequence positions. The extent of this problem varies from one method to another. Thus, the next step in constructing a sequence phylogeny is to assess the reliability of the inferred branching pattern. This is often accomplished by a bootstrap analysis (Felsenstein, 1985). Bootstrap procedures involve construction of new sequence sets by resampling with replacement sites (columns) of the original set, building a tree for each new set, and calculating the percentage of times. A cluster reappears in the bootstrap replications. This percentage is called the bootstrap value, and clusters with a bootstrap value >95% are widely considered to reflect correct relationships (Felsenstein, 1985).

#### 2.2.3.3 Molecular studies of the Xylariaceae

Since the morphological and biochemical characteristics of the Xylariaceae are frequently too limited, molecular techniques have, therefore, been applied.

Lee *et al.* (2000) analyzed 18 species of *Xylaria* and related genera by using nuclear ribosomal ITS1-5.8S-ITS2 sequences. Species of selected *Xylaria* were divided into three groups, and phylogenetic analysis of these was also supported by a set of signature nucleotides of ITS1-5.8S-ITS2 sequences. Group A consisted of *Xylaria arbuscula*, *Xylaria mali*, and *Xylaria apiculata* Cooke, whereas group B consisted of *Xylaria cornu-damae* (Schw.) Fr., *Xylaria longipes* Nitschke, *Xylaria acuta* Peck, *Xylaria castorea* Berkeley, *Xylaria enteroleuca* (Spegazzini) Martin, and *Xylaria fioriana* Saccardo. Group C included *Xylaria polymorpha* and *Xylaria hypoxylon* (L.: Fr.) Greville. In contrast, *Xylaria cubensis* appeared to be separated from other *Xylaria* species. The results showed that a few characteristics based on ascospores, perithecia and stromata, support grouping of *Xylaria* inferred from the molecular data. But there seems to be no character of universal significance that can justify the present phylogenetic results. It may indicate that convergent

evolution of characters occurred many times within *Xylaria* species. Such possible changes in convergent evolution along with variations associated with developmental stages of stromata, might have caused confusions in identifying and classifying *Xylaria* species. Phylogenetic analysis based on molecular data such as ITS sequences of the present study proved to be very practical for taxonomic investigations at specific or generic levels in identification or classification of fungi of highly variable morphology like *Xylaria*.

Molecular and morphological investigations of Daldinia in Northern Europe have also been undertaken by Johannesson et al. (2000). Since the study of Daldinia was undertaken by Ju et al. (1997), which was based on morphological and cultural characteristics, it has proven difficult to name collections from Northern Europe. The confusion over the typification of especially the type species of the genus has also created problems. Therefore, five taxonomic entities of Daldinia concentrica, Daldinia cf. fissa, Daldinia grandis, Daldinia loculata, and Daldinia cf. petriniae, that were found exclusively on burnt wood, were defined based on both morphotaxonomical and ITS-sequence criteria. The results showed that at least five different taxa of Daldinia are present in Northern Europe, and the preference for burnt hosts has either been gained or lost more than once in the history of the genus. Later, Stadler et al. (2001) studied the secondary metabolite profiles coupled with DNA fingerprints of Daldinia. They selected 18S rDNA to amplify and digested DNA fragments with three different restriction enzymes, HpaII, HaeIII, and TaqI. Then the DNA restriction patterns were used to construct the phylogenetic tree according to the unweighted pair group method using arithmetric averages (UPGMA). The results showed clearly within species.

# For the genus Hypoxylon, Sanchez-Ballesteros et al. (2000)

studied the phylogenetic relationships of Hypoxylon and its allies, the complete DNA sequences of the ITS regions (including the 5.8S rRNA gene) from 41 isolates were determined, then aligned and processed for phylogenetic reconstruction, and critically compared to the available taxonomic information. Their results generally agreed with the current concepts and limits established for the genus by Ju and Rogers (1996). The species and varieties of Hypoxylon in the sense of modern authors appear to be a monophyletic group within the Xylariaceae. However, the recent infrageneric division of Hypoxylon into sections Hypoxylon and Annulata (Ju and Rogers, 1996) was not supported by this limited molecular phylogenetic analysis. In another study, Mazzaglia et al. (2001b) confirmed the efficacy of the 5.8S-ITS2 sequence analysis in phylogenetic studies of Hypoxylon fragiforme, Hypoxylon multiforme and related genera. The analysis confirmed that Hypoxylon is a taxonomically and phylogenetically separated taxon from Biscogniauxia and Entoleuca. Moreover, Hypoxylon fragiforme isolates formed a group separated from the single isolate of Hypoxylon multiforme. Although clearly belonging to the same genus, they were once recognised as being very closely related (Miller, 1961). However in the revision of Ju and Rogers (1996), these two species were separated with H. fragiforme being placed in section Hypoxylon and H. multiforme in section Annulata. This was on the basis of absence (Hypoxylon) or presence (Annulata) of a layer of carbonaceous stromatal tissue enclosing the perithecia (Ju and Rogers, 1996).

For *Biscogniauxia*, Mazzaglia *et al.* (2001a) developed a polymerase chain reaction (PCR) assay to detect *B. mediterranea* in asymptomatic tissues of *Quercus cerris*. They designed two specific primers (MED1 and MED2) by

comparison of sequences of ITS1 and ITS4 of 21 isolates of *B. mediterranea* and related species. Both primers were able to detect *B. mediterranea* DNA in the host tissues at picogram quantity of target DNA. The reliability of the results was confirmed by Southern blot analysis.

In addition, Platas *et al.* (2001) found a simple tandem repeat sequence in the ITS1 region of the rDNA of members of order Xylariales. The number of repetitions detected ranged from one to six, and they could be found in pure tandem or interspersed. These replications could have been generated by slipped strand mispairing. The presence of this sequence increases the normal rate of divergence in the ITS1 of the Xylariales.

On the basic of published data to date, molecular taxonomy may be applied and prove to be valuable as a standard technique for identification of members of the Xylariaceae. Therefore, ribosomal DNA subunit sequence analysis of selected xylariaceous fungi and their comparison with the available sequences on databases will greatly help in their identification especially in the absence of a teleomorph or where morphological characteristics are insufficient to clearly separate closely related species. However, it will be necessary to greatly enlarge the available data by including more genera and by increasing the number of isolates examined for each species. The reliable identification of teleomorphic materials using the conventional taxonomy will be an important prerequisite to ensure validity of molecular data deposited in databases.

# **2.3** Problematic groups in the systematic of the xylariaceous fungi

Since the xylariaceous fungi are cosmopolitan fungi and often exhibit high variation in morphology depending on localities of collection, stage of development, and criteria of identification, there are problems in recognizing and delimiting some of the genera and species.

#### 2.3.1 Group I: Astrocystis and Rosellinia

According to the broadly accepted current concept of the genus, *Rosellinia* is delimited within the Xylariaceae by five main characters: the stromata are uniperitheciate (rosellinioid), superficial, subglobose, associated with a hyphal mat usually called subiculum, and associated with a *Geniculosporium*-like anamorph (including *Dematophora* R. Hartig and *Geniculosporium* Chesters & Greenhalgh). The delimitation of *Rosellinia* led L. Petrini (1992) to move taxa excluded from this genus to *Amphisphaerella*, *Anthostromella*, *Astrocystis*, *Coniochaeta*, *Xylaria*, and other sordariaceous or xylariaceous genera.

Astrocystis Berk. & Broome is based on Astrocystis mirabilis Berk. & Broome as a type species, which occurs on bamboo and features a skirt or volva on the perithecial stroma (Berkeley and Broom, 1875). The stellate aspect of the volva led the authors of the species name to provide a somewhat fanciful illustration that gives the impression that *A. mirabilis* looks exactly like a minute earth star (Geaster) (Berkeley and Broome, 1875). Penzig and Saccardo (1904) recognised the strong relationship of *Astrocystis* with *Rosellinia* De Not., and noted that the illustrations provided by Berkeley and Broome are "strongly fictitious". Diehl (1925) published a detailed account of *A. mirabilis*, including its nomenclatural and taxonomic history. He had a broad concept of ascospores as "…acuminate to rounded, elliptical, narrow to broad, light brown becoming dark brown and subopaque when mature 10-21 x 4-2  $\mu$ m, chiefly 11-13 x 5-7  $\mu$ m..." Diehl's expanded concept of the species resulted, in part, from his acceptance of *Rosellinia bambusae* P. Henn. as a synonym of *A. mirabilis*.

Some disagreement over the status of the genus *Astrocystis* Berk. & Br., which accommodates *Rosellinia*-like fungi devoid of subiculum but with stromata splitting the host surface or with a carbonaceous extension at the base, associated with an *Acanthodochium* anamorph, persists between different authors. *Astrocystis* is recognised by Petrini (1993), and Læssøe and Spooner (1994), but synonymized with *Rosellinia* by Ju and Rogers (1990) and San Martín and Rogers (1994). Ju and Rogers (1990) have examined type and other materials identified as *Astrocystis mirabilis*. Using ascospore and stromatal features, these collections can mostly be divided into two distinct groups corresponding to *A. mirabilis* in the original sense and *R. bambusae*. The division is strongly reinforced by data from cultures obtained from recent field collections. Cultural characters differ between representatives of these groups. Each produces a distinctive anamorph in nature and in culture that is referable to *Acanthodochium* Samuels. They, thus, consider *R. bambusae* and *A. mirabilis* (as *Rosellinia*) to be distinct species.

The genus *Rosellinia* appears to be poorly represented in Thailand. Only two species, *R. necatrix* and *R.* cf. *procera*, were reported by Thienhirun (1997) and for *Astrocystis* only one species, *A. mirabilis*, has been reported (Thienhirun, 1997). It appears to be more common in peninsular Malaysia (Whalley, 2001). Since there are not clear separation of these genera and since their anamorphs are entirely different the molecular examination could determine their true relationship.

#### 2.3.2 Group II: Camillea

The genus *Camillea* was erected from other xylariaceous fungi, which possess erect cylindrical or short discoid black stromata and have a hard carbonaceous crust (Fries, 1849). Camillea was reviewed by Læssøe et al. (1989) when the genus was considerably enlarged with many species formerly placed in the genera Nummularia and Hypoxylon section Applanata (Miller, 1961). Camillea is characterised by applanate or cylindric stroma, erumpent through bark, and perithecia completely immersed. The apical apparatus of the ascus is vase- to urn-shaped, domeshaped or somewhat diamond-shaped. Ascospores are light-coloured, ornamented, lacking germ slits, and without a loosening perispore (Læssøe et al., 1989). The ascospores of most *Camillea* species appear smooth by light microscopy but they appear characteristically ornamented by SEM with warts, spines, pits, reticulations or to be longitudinally ribbed (Læssøe et al., 1989; Rogers et al., 1991; San Martín, Gonzales, and Rogers, 1993; Whalley, 1995; Whalley, 1996; Whalley et al., 1999). Læssøe et al. (1989) recognised 28 species and varieties of Camillea of which C. obularia (Fr.) Læssøe, J.D. Rogers & Lodge (as C. broomeiana and C. tinctor were the only species known from outside the New World, with C. tinctor exhibiting a widespread distribution. The discovery of three new species of Camillea from Mexico (San Martín, González, and Rogers, 1993) maintained this pattern until the recent discoveries of C. selangorensis (Whalley et al., 1996) and C. malaysianensis described from Kuala Selangor, Malaysia. Camillea selangorensis was later reported from Thailand (Whalley et al., 1999).

*Camillea leprieurii* Mont. has a dimorphic form, erect (camilleoid) form and applanate (expanded, "hypoxyloid" form), which had been recognised by
Patouillard (1888). He stated that the applanate *H. melanaspis* Mont. was the "forme étalée" or expanded state of C. leprieurii (Mont.) Mont. The examination of ascospores of C. leprieurii and H. melanaspis by SEM revealed an intricate ornamented ascospore wall composed of anastomosing ridges overlying a regular ribbed substructure in which the ribs are orientated perpendicular to the ridges (Rogers, 1977; 1979). Although he was not convinced that these two forms were the same species, he concluded that they have a close relationship (Rogers, 1979). Læssøe et al. (1989) explained the characteristics of both dimorphic forms of C. leprieurii. In erect form, the stromata are erumpent through bark, cylindrical, seated on slightly broader disc, which remains after broken off stromata. The stromata are also apex discoid-depressed with narrow rounded margin, brittle, black or with thin, flaky white ectostroma, initially with brown fungus and host covering, sometimes with felt-like dark brown subiculum of old *Xylocladium* anamorph. For applanate form, the stromata is erumpent through bark, plano-convex with plane ostiolar part, margin without rim, circular, orbicular or confluent, shiny black ('polished') or with flaky white ectostroma, initially with brown fungus (Læssøe et al., 1989). Both forms of C. leprieurii are distributed in Bolivia, Brazil, Colombia, Ecuador, Franch Guiana, Guyana, Nicaragua, Panama, Peru, Puerto Rico, Surinam, and Venezuela, but they have not been recorded in Thailand or Southeast Asia.

*Camillea tinctor* is characterised by stromata which is orbicular to elongate, and applanate with a slightly convex centre. Ascospores are smooth by light microscopy with distinct poroid ornamentation by SEM (Læssøe *et al.*, 1989; Rogers *et al.*, 1991; San Martín, Gonzales, and Rogers, 1993; Whalley, 1995; Whalley, 1996; Whalley *et al.*, 1999). The stromata of *C. tinctor* is usually accompanied by yellow

staining of the wood immediately beneath. In Thailand, *C. tinctor* has been first recorded by Thienhirun (1997), and exhibits a wide distribution from North to South. In addition, *Camillea tinctor* occurs in neighbouring countries.

*Camillea selangorensis* is characterised by stromata which is circular to orbicular, slightly elevated, and 2-3 thick with a slightly raised rim. Ascospores are minutely warted by light microscopy, strongly verrucose by scanning electron microscopy, and the type locality is lowland forest bordering on mangrove (Whalley, 1995; Whalley *et al.*, 1999). The discovery of *C. selangorensis* (Whalley *et al.*, 1995) provided clear evidence that *Camillea*, once considered to be a New World genus, has greater world wide representation than is generally believed. In Thailand, *C. selangorensis* was discovered from a similar ecological situation in Phuket Island (Whalley *et al.*, 1999). This suggested that this species might be expected in similar areas elsewhere in the region.

#### 2.3.3 Group III: Daldinia

The genus *Daldinia* is characterised by conspicuous internal alternating ring zones, which presently comprises of about 25 species (Stadler *et al.*, 2004). The type of the genus is *D. concentrica* which was firstly described by Bolton (1789) in Great Britain. Lloyd (1924) and Child (1932) were among the first to study the biology of *Daldinia*, and gave evidence on the existence of several species within the genus. Tropical species of the genus were studied as well. The surveys of *Daldinia* are now available from countries such as Papua New Guinea (Van der Gucht, 1994; 1995), Mexico (San Martín, 1992), and Thailand (Thienhirun, 1997). The current taxonomy of *Daldinia* is outlined in the latter monograph and subsequent additions by

Rogers *et al.* (1999), Ju, Vasilyeva, and Rogers (1999), Stadler *et al.* (2001) and Stadler, Baumgartner, and Wollweber (2001). Species of *Daldinia* are segregated by the combination of anamorphic and teleomorphic characters and by their colours of stromatal pigments in 10% KOH. The anamorphic states of *Daldinia* spp. and allies are morphologically rather similar, ranging from *Nodulisporium*-like to *Sporothrix* and *Virgariella*-like and further branching patterns (Stadler *et al.*, 2001).

Daldinia concentrica (Rogers et al., 1999; Johannesson, Læssøe, and Stenlid, 2000; Stadler et al., 2001) is now generally accepted to occur primarily in Western and Northern Europe in temperate regions, and its stromata is preferentially encountered on *Fraxinus* whereas *Daldinia eschscholzii* (Ehrenb.: Fr.) Rehm is a widely distributed species of subtropical and tropical climates (Van der Gucht, 1994; Ju et al., 1997). The stromata of *D. eschscholzii* has extractable pigments in KOH, whose colours and intensities were weaker than of *D. loculata* and *D. fissa*, but similar to those observed in *D. concentrica*. According to the current definition of *D. concentrica*, this species cannot be easily distinguished from *D. eschscholzii* by the colour of stromatal pigments alone. However, both species differ in several morphological features. Culture of *D. eschscholzii* generally produces rather small conidia, never exceeding 6  $\mu$ m in length and 3.5  $\mu$ m in width. Moreover, *D. eschscholzii* is characterised by having smaller ascospores, and its stromatal surface is not crackled into a fine network in mature specimens (Ju et al., 1997; Stadler et al., 2004).

Recently, Stadler *et al.* (2004) proposed five new species separated from *D. concentrica* and *D. eschscholzii*. The new species are *D. macaronesica* M. Stadler, Wollweber & J. Castro, *D. martinii* M. Stadler, Venturella & Wollweber, D. raimundi M. Stadler, Venturella & Wollweber, D. palmensis M. Stadler,Wollweber & H-.V. Tichy, and D. vanderguchtiae M. Stadler, Wollweber & Briegert.

*Daldinia macaronesica* differs from *D. eschscholzii*, *D. palmensis*, and *D. vanderguchtiae* in size, morphology and ornamentation of its ascospores and in its anamorphic characters. Stadler *et al.*, (2004) reported *D. macaronesica* is a close relative of *D. concentrica* but it differs in ascospores. They are more slender, show a wider range in size and bear a more conspicuous ornamentation than those of *D. concentrica*. In addition, *D. macaronesica* shows apparent host specificity for *Ocotea foetens*, a plant endemic to the Macaronesian Islands (Kunkel, 1993).

Daldinia martinii differs from D. concentrica and D. eschscholzii in anamorph form. Daldinia martinii has Sporothrix to Virgariella and Nodulisporiumlike forms.

*Daldinia raimundi*, which was originally reported as *D. concentrica pro parte* (Venturella *et al.*, 2001), differs in more conspicuous ornamentation of perispore by SEM. Moreover, the ascospore size of *D. raimundi* is in the range of *D. eschscholzii* (Ju *et al.*, 1997) rather than in the one typically found in *D. concentrica*.

Daldinia palmensis was originally identified as *D. eschscholzii*. The SEM characteristics of *D. palmemsis* ascospores were found in agreement with *D. eschscholzii* found from around the world. Notably, in contrast to the former species, the germ slit of the ascospores of *D. palmensis* may either be straight or slightly undulate. However, only a detailed study of the anamorph revealed significant differences to the former species (Stadler *et al.*, 2004). The conidiophores of *D. palmensis* are *Nodulisporium*-like or *Sporothrix*-like forms whilst those of *D. eschscholzii* are *Nodulisporium*-like in form.

*Daldinia vanderguchtiae* is peculiar among the concentricol-containing taxa of *Daldinia* in having smooth ascospores by SEM. Otherwise its ascospores resemble those of *D. eschscholzii* in size range as well as in shape. However, conidiophores approaching a *Virgariella*-like branching pattern as defined in Ju and Rogers (1996) were hitherto only seen in a culture of *D. grandis* Child 11932 from New Zealand (Ju *et al.*, 1997), and in *D. martini*, but they have never seen in *D. eschscholzii*.

In Thailand, three species of *Daldinia* had been reported since 1963. Carrol (1963) recorded *D. eschscholzii* from Chiang Mai Province and *D. concentrica* was recorded by Phanichapol (1968), Cansrikul (1977) and Schumacher (1982) whilst Ju, Rogers and San Martin (1997) described *D. bambusicola* for a distinctive taxon associated with bamboo and having a Southeast Asian distribution. Although intensive collection has been undertaken by Thienhirun (1997) in the Doi Chiang Dao area and similar forests in Chiang Mai Province, where Schumacher (1982) reported *D. concentrica* as occurring, it was *D. eschscholzii* not *D. concentrica* which was found there. Carroll's record of *D. eschscholzii* is from the nearby Doi Suthep (Carroll, 1963). Thienhirun (1997) believed that the former recorded of *D. concentrica* in fact represent *D. eschscholzii*. Certainly *D. concentrica* is more frequently associated with temperate regions and *D. eschscholzii* with tropics and subtropics (Ju *et al.*, 1997). Thienhirun (1997) reported that five species, *D. cf. caldariorum, D. loculata, D. eschscholzii, D. bambusicola*, and *Daldinia* taxonomic species 1, were found in Thailand.

#### 2.3.4 Group IV: Hypoxylon

The genus *Hypoxylon* delimitation has been rearranged several times among mycologists (Miller, 1961; Martin, 1968; Ju and Rogers, 1996). Miller's monograph of Hypoxylon was divided into four sections, Hypoxylon, Annulata, Applanata and Papillata (Miller, 1961). His monograph was strongly relied on stromatal form, texture, and nature of the ostiole. Consequently, this monograph failed to recognise the relationships between groups of species. The section Applanata sensu Miller has since been redistributed between Camillea and Biscogniauxia (Læssøe, Rogers, and Whalley, 1989; Whalley, Læssøe, and Kile 1990; González and Rogers, 1993) whereas member of the section Papillata subsection Primocinerea (Miller, 1961) have been allocated to a range of genera including Nemania (Pouzar, 1985), Rosellinia (Petrini, 1992), and Euepixylon (Læssøe and Spooner, 1994). The genus Hypoxylon was revised by Ju & Rogers (1996) using four major criteria to define the genus; Nodulisporium-like anamorphs having stromata unipartite, never erect, with a solid and homogenous basal tissue below the perithecial layer. They divided Hypoxylon in two sections, section Hypoxylon and section Annulata, containing at least 130 accepted species and varieties. Ju and Rogers (1996) were able to utilize data, which absent from the monograph of Miller (1961) such as ascospore ornamentation using SEM, form of the apical apparatus of the ascus, germination slit morphology and the colour of stromatal pigments extracted with 10% potassium hydroxide (10% KOH). However, the revision of the genus identification of certain Hypoxylon species remains problematic resulting from considerable variation in species characteristics. This has proved to be most pronounced when considering tropical species of the Annulata especially Hypoxylon nitens, H. moriforme, H. bovei

var. *microspora*, *H. purpureonitens*, *H. stygium*, and *H. stygium* var. *annulata*, when there are variations and overlap in their morphological features.

#### 2.3.5 Group V: Xylariaceous endophytes

There are eight xylariaceous genera recorded as endophytes, Anthostomella (Petrini and Petrini, 1985; Petrini et al., 1987), Biscogniauxia (Petrini and Müller, 1986), Daldinia (Petrini and Petrini, 1985; Petrini and Müller, 1986), Hypoxylon (Petrini and Müller, 1986), Kretzschmaria (Petrini and Petrini, 1985; Petrini and Müller, 1986), Nemania (Petrini and Petrini, 1985; Petrini and Rogers, 1986), Rosellinia (Petrini and Petrini, 1985; Petrini, 1992), and Xylaria (Petrini and Petrini, 1985). The identification of xylariaceous endophytes is often difficult since they fail to produce suitable diagnostic features. And it is very infrequently to form their teleomorph in culture. The situation regarding tropical endophytes is much more complex as a result of their abundance and impressive diversity (Rodrigues and Samuels, 1990; Whalley, 1993; Whalley, 1996). It is doubtful whether differentiation of species on the basis of cultural and anamorphic features alone will ever be possible since differences between individual species are often insufficient to allow for absolute identifications to be made (Petrini, Petrini and Rodrigues, 1995). However, studies of Xvlaria indicate that a combination of morphological characters and biochemical analyses might enable satisfactory identifications to be made (Brunner and Petrini, 1992; Rodrigues, 1992; Rodrigues, Leuchtmann, and Petrini, 1993). There are also indications that secondary metabolite profiles from endophytic isolates might be matched with those obtained from cultures derived from teleomorphic material thus enabling identity to be established (Whalley and Edwards, 1995; 1999).

A preliminary study of *X. cubensis* comparing secondary metabolites e.g. cubensic acid (Adeboya *et al.*, 1995) obtained from teleomorphic derived cultures with those produced by endophytic *Xylaria* isolates from *Euterpe oleracea* leaves confirmed the finding of Rodrigues that they belonged to *X. cubensis* (Whalley, 1996; Rodrigues, 1992). Ongoing research on secondary metabolites of the Xylariaceae was included endophytic isolates to determine the suitability of this approach for the identification of endophytic members of the family.

# **CHAPTER III**

# **MATERIALS AND METHODS**

#### 3.1 Chemicals, reagents, and media

#### 3.1.1 Morphological taxonomic study

Reagents used for teleomorphic characteristics of xylariaceous fungi were Melzer's solution (Appendix 1.2A) to determine whether or not the ascus apical ring blued (the amyloid iodine reaction), and 10% potassium hydroxide (KOH) (Fluka, Sigma-Aldrich Chemical Company, U.S.A.) to determine whether or not the perispore, and to extract colour pigments of stromata.

The medium used for cultural characteristic study was potato dextrose agar (PDA) (Appendix 1.1A).

#### 3.1.2 Chemical taxonomic study

The medium used for fungal growth in metabolite extraction was 2% malt extract broth (MEB) (Difco, Difco Laboratories, U.S.A.) containing 6% glucose (Merck, Merck KGaA, Germany).

Reagent used for fungal metabolite extraction was ethyl acetate (Sigma, Aldrich Chemical Company, U.S.A.). The components of mobile phase in thin layer chromatography (TLC) were toluene, ethyl acetate, and acetic acid (Sigma). Reagents used for chemical detection of TLC plate were p-nitroaniline and anisaldehyde (Sigma).

#### 3.1.3 Nucleic acid study

Reagents used for genomic DNA extraction were lysis buffer (Appendix 3.1A); phenol, chloroform, isoamyl alcohol (Merck) to purify genomic DNA; isopropyl alcohol (Merck) to precipitate genomic DNA; 70% ethanol to wash genomic DNA pellet. Agarose (Promega, Promega Coporation, U.S.A.) was used to detect DNA by agarose gel electrophoresis.

Reagents used for Polymerase Chain Reaction (PCR) amplification were the 10X PCR buffer (Sigma), dNTPs (dATP, dCTP, dGTP, and dTTP) (Invitrogen, Invitrogen life technologies, U.S.A.), and *Taq* DNA polymerase (Sigma). The oligonucleotide primers were ordered from the Science Pacific Company, Ltd., Thailand. The QIA-quick PCR purification kit (Qiagen, Qiagen Coporation, U.S.A.) was used in PCR purification. The BigDye Terminator Ready Reaction kit (Perkin Elmer, Applied Biosystems Inc., U.S.A.) was used for the nucleotide sequencing reaction.

#### 3.2 Instrumentation

Instruments required for morphological taxonomic and nucleic acid studies of xylariaceous fungi were located at the Instrument Buildings of the Centre for Scientific and Technological Equipment, Suranaree University of Technology, Nakhon Ratchasima, and specimen comparison of collected fungi with reference collections were performed at the Royal Forest Department, Bangkok, Thailand.

Instruments required for chemical taxonomic studies were located at the School of Biomolecular Sciences, Liverpool John Moores University, Liverpool, U.K.

Instruments required for DNA sequencing were located at the Biotechnology

and Development Office, Department of Agriculture, Pathumthani, Thailand.

## 3.3 Collection of xylariaceous fungi for taxonomic studies

The teleomorph stage of xylariaceous fungi were collected during rainy season, June to December in years 2002 and 2003, from 14 different locations in Thailand as described in Table 6. All specimens were recorded for their collection dates, locations, and habitats. The collections were kept as herbarium by freezing at -20°C for one week to destroy insects and/or mites and then drying at 37°C for approximately 7 days before keeping in sealed plastic bags.

Year	Location
2002	Chiang Mai Province
	Nakhon Ratchasima Province
2003	Phu Luang, Nakhon Ratchasima Province
	Nong Rawieng, Nakhon Ratchasima Province
	Burirum Province
	Chaiyaphum Province
	Plant Nursery of the Royal Forest Department, Ratchaburi Province
	Suranaree University of Technology, Nakhon Ratchasima Province
	Kanchanaburi Province
	Petchaboon Province
	Songkhla Province
	Trad Province
	Yasothon Province
	Chiang Rai Province

**Table 6.** Locations and time of specimen collection in this study.

# 3.4 Morphological taxonomic studies of the problematic groups in xylariaceous fungi

#### 3.4.1 Macroscopic study

The teleomorph of the collected xylariaceous specimens were observed for shape, size, colour of their stromatal surface, perithecia, and ostioles using the Olympus Stereomicroscope SZX fitted with the Olympus Digital Camera DP11 (Olympus, Olympus Optical Co., Ltd. Japan). Colours of stromatal surface were determined comparing to the Rayner Mycological Colour Chart (Rayner, 1970). The stromatal pigments of *Hypoxylon* and *Daldinia* were extracted in 10% potassium hydroxide (KOH), leaved for one minute, and observed the colour compared to the colour chart (Ju and Rogers, 1996; Ju, Rogers, and San Martín, 1997).

#### 3.4.2 Microscopic study

The collected xylariaceous fungi were observed for colour, shape, and size of ascospores by mounting with distilled water and using the Olympus Compound Microscope BX51 fitted with the Olympus Digital Camera DP11 (Olympus). The ornamentation of perispore and epispore were observed in 10% KOH to determine dehiscence or indehiscence. The apical apparatus of ascus was examined for amyloid reaction including shape and sized by using Melzer's iodine reagent. The type of germ slit was included.

Ascospore ornamentation of some xylariaceous isolates was also observed using SEM. Dried xylariaceous stromata were attached to aluminium stubs with Dag metallic paint, coated with gold, and examined using JEOL-6400 SEM (JEOL, Japan).

#### **3.5** Isolation and cultivation of the selected xylariaceous fungi

The ascospores of selected xylariaceous specimens were isolated for culture by the method as described by Ju and Rogers (1996) and Thienhirun (1997). A portion of the stromatal surface including the upper parts of perithecia was removed with a sterile razor blade. The contents of the exposed perithecia were scooped out and spotted with a fine-tipped sterile needle in Petri dishes containing PDA medium (Appendix 1.1A). Hyphal tips emerging from the perithecial contents were then cut and transferred to fresh media. All isolates were routinely incubated at 25°C, and subcultured every two months. The anamorph form was also observed. Their mycelia were maintained in 15% glycerol at -20°C as stock cultures.

#### **3.6** Chemotaxonomic study of the selected xylariaceous fungi

Since *Xylaria* species are common endophytes isolated from several plants as described previously and most of them could not form mature teleomorph stage in their cultures, the representatives of *Xylaria* isolates and xylariaceous endophytes were selected to study on secondary metabolite profiles by using TLC technique.

#### 3.6.1 TLC analysis of secondary metabolites from agar plugs

Selected *Xylaria* isolates were grown on yeast extract sucrose (YES) agar (Appendix 2.1A) in 9-cm Petri dishes at 25°C for 4 weeks. The extracellular metabolite analysis was performed according to the standard method (Lund and Frisvad, 1994). Small agar plugs were cut from the fungal colony using a 4-mm flamed cork borer. The plugs were wetted by a drop of chloroform : methanol (2 : 1, v/v) and immediately applied onto a TLC plate (Silica gel 60, Merk Kieselgel

GF254), 2.5 cm from the bottom line. The eluent system composed of toluene : ethyl acetate : 90% formic acid, 5 : 4 : 1, v/v/v). The TLC plate was inspected in daylight and under ultra-violet (UV) transilluminator (366 nm and 265 nm), and all spots were noted. Each detected spot was calculated for retention factor ( $R_f$ ) as follows:

$$R_{\rm f} = \frac{\text{Distance of each compound}}{\text{Distance of solvent}}$$

#### **3.6.2** TLC analysis of secondary metabolites from cultural broth

# 3.6.2.1 Secondary metabolite extraction from 100 mL of cultural broth

Selected *Xylaria* isolates were grown in 100 mL of 2% malt extract broth (Difco) containing 6% glucose for 8 weeks (Pittayakhajonwut, 2000). The broth medium was filtrated from fungal mycelium, and extracted with equal volume of ethyl acetate (Sigma). After extraction, the ethyl acetate layer was transferred to a volume metric flask and the extracted solution was concentrated by evaporating until the solution was changed to powder. Then, the extracted powder was dissolved with 5 mL ethyl acetate. The extracted solution was spotted onto the TLC plate (Silica gel 60, Merk Kieselgel GF254) using capillary tube. The eluent system was toluene : ethyl acetate : acetic acid; 50 : 49 : 1 (v/v/v). The detection systems were p-nitroaniline (Sigma) spray agent and anisaldehyde (Sigma) spray agent.

#### **3.6.2.2** Secondary metabolite extraction from 1 L of cultural broth

An isolate of *Xylaria* was selected to culture in 1 L of 2% malt extract broth (Difco) containing 6% glucose for 8 weeks. The cultural broth was separated and extracted for secondary metabolites using the same procedures as described in section 3.6.2.1.

#### **3.7** Nucleic acid studies of the selected xylariaceous fungi

DNA of the selected xylariaceous fungi and the reference strains were studied.

#### 3.7.1 Extraction of genomic DNA

Genomic DNA of the xylariaceous specimens selected as representatives was extracted from their cultural mycelia and stromatal herbarium, in case of uncultured specimens, using the method of Lee and Taylor (1990) with some modifications. The fungal mycelium was harvested and rinsed with TE buffer (Appendix 3.2A). The washed mycelium was squeezed, placed in a microcentrifuge tube, and stored at -20°C overnight. The frozen mycelium was ground, added lysis buffer (Appendix 3.1A), and incubated at 65°C for an hour. The equal volume of phenol : chloroform : isoamyl alcohol (25 : 24 : 1, v/v/v) was added, gently mixed, and centrifuged at 12,000 rpm (Labofuge 400R, Heraeus Instruments, Heraeus Instruments GmbH, Germany) at 4°C for 20 min. The top supernatant was transferred to a fresh microcentrifuge tube. An equal volume of isopropanol (Merck) and onetenth volume of 3 M ammonium acetate (pH 5.2, Appendix 3.3A) (BDH, BDH Laboratory Supplies Poole, England) were then added, gently mixed, and placed in an icebox for 10-30 min to precipitate genomic DNA. The tube was centrifuged at 12,000 rpm for 30 min at 4°C. The DNA pellet was washed with 400 µL of 70%

ethanol (Merck), air dried, and then resuspended in 50 µL TE buffer. RNA was removed by adding Ribonuclease A (1 mg/mL) (Invitrogen) (Appendix 3.4A) to give a concentration of 10 µg RNase/mL sample and the tube was incubated at 37°C for 30 min. Genomic DNA was detected in 1% agarose gel electrophoresis, stained with ethidium bromide (1 mg/mL) (BioRad, BioRad Laboratories, Italy), and examined under UV transilluminator (BioRad). The concentration of DNA was measured by SmartSpec <sup>TM</sup> 3000 spectrophotometer at 260 nm (BioRad) and the purity of DNA was calculated from the ratio of optical density at 260/280. The conversion factor for determination of DNA concentration is 1 OD<sub>260</sub> = 50 µg/mL of double stranded DNA. Then, DNA solution was maintained at -20°C until use.

#### 3.7.2 Amplification of the ribosomal RNA genes

The 18S ribosomal RNA gene and the internal transcribed spacer (ITS) regions 1 and 2 including 5.8S ribosomal RNA gene of xylariaceous fungi were amplified using PCR.

#### 3.7.2.1 Amplification of 18S ribosomal RNA gene

The 18S ribosomal RNA gene amplification was performed using NS1 and NS8 primers as forward and reverse to obtain the whole gene (Figure 8 and Table 7). The PCR amplification reaction was performed in 50  $\mu$ L mixture containing 50 ng of fungal DNA, 5  $\mu$ L of 10X reaction buffer (10 mM KCL, 20 mM Tris-HCl pH 8.8, 10 mM (NH<sub>4</sub>)<sub>2</sub>SO<sub>4</sub>, 2 mM MgSO<sub>4</sub>, 0.1% Triton X-100), 200  $\mu$ M of each dATP, dCTP, dGTP, and dTTP, 1  $\mu$ M of each primer, 1.0 unit of *Taq* DNA polymerase (Sigma), and adjusted volume to 50  $\mu$ L with deionized water. The program of amplification consisted of 1 cycle of 95°C for 5 min; 35 cycles of 95°C for 1 min, 53°C for 2 min, 72°C for 2 min; and the final cycle of 72°C for 10 min. The PCR reactions were carried out in the automated thermal cycle (i-cycle, BioRad, U.S.A.).





 Table 7. Nucleotide sequences of PCR primers used in this study.

Name	Sequence (5'- 3')	Target region <sup>a</sup>	Reference
NS1	GTAGTCATATGCTTGTCTC	SSU 20-38	White et al. (1990)
NS8	TCCGCAGGTTCACCTACGGA	SSU 1788-1769	White et al. (1990)
ITS4	TCCTCCGCTTATTGATATGC	LSU 60-41	White et al. (1990)
ITS5	GGAAGTAAAAGTCGTAACAAGG	SSU 1744-1763	White <i>et al.</i> (1990)

<sup>a</sup> Saccharomyces cerevisiae numbering

#### 3.7.2.2 Amplification of internal transcribed spacer (ITS) 1 and 2

#### including 5.8S ribosomal RNA gene

The ITS1-5.8S-ITS2 region was amplified using ITS4 and

ITS5 primers (Table 7). The PCR amplification reaction was performed in 50  $\mu$ L mixture containing 10 ng of fungal DNA, 5  $\mu$ L of 10X reaction buffer (10 mM KCL,

20 mM Tris-HCl pH 8.8, 10 mM (NH<sub>4</sub>)<sub>2</sub>SO<sub>4</sub>, 2 mM MgSO<sub>4</sub>, 0.1% Triton X-100), 200  $\mu$ M of each dATP, dCTP, dGTP, dTTP, 2.5  $\mu$ M of each primer, 1.0 unit of *Taq* DNA polymerase, and adjusted volume to 50  $\mu$ L with deionized water. The program of ITS1-5.8S-ITS2 region amplification consisted of 1 cycle of 95°C for 5 min; 35 cycles of 95°C for 30 sec, 53°C for 1 min, 72°C for 1 min; and final cycle of 72°C for 10 min. The PCR reactions were carried out in the automated thermal cycle (BioRad).

#### **3.7.3 Detection of PCR-amplified products by agarose gel electrophoresis**

Agarose gel electrophoresis is a standard method used to separate, identify, and purify DNA fraction. Agarose gel was prepared at a concentration of 1.5% (w/v) in 1X TBE buffer (Appendix 3.5A), melted in microwave oven until completely dissolved, and then poured into gel box with an appropriate comb.

Five microliters of PCR-amplified product was thoroughly mixed with 6X loading buffer (Appendix 3.6A). The mixture was loaded into the submarine 1.5% agarose gel, and electrophoresis was carried out at constant 100 volts until the bromphenol blue dye reached about 2 cm from the lower edge of the gel, then the electrophoresis was stopped. One hundred base pair DNA ladder (Invitrogen) was used as standard markers to determine the molecular size of DNA fragments.

After electrophoresis, the agarose gel was stained with ethidium bromide by soaking the gel in a solution containing 10  $\mu$ g/mL of ethidium bromide, and visualized under UV transilluminator (BioRad). The agarose gel was photographed for being reference.

#### **3.7.4** Purification of DNA-amplified products

The single band of the DNA-amplified product as estimated size was purified throughout the QIAquick purification kit (Qiagen) according the manufacturer's instruction. The DNA-amplified product that contained primer dimer band, approximately 50 bp, was purified by low melting point (LMP) agarose gel purification (BIO 101, Inc., U.S.A.). The DNA was mixed with 6X loading buffer and then loaded into the 1.5% LMP agarose gel. Electrophoresis was carried out at the constant 100 volts until the bromophenol blue dye reached about 2 cm from the lower edge of the gel then electrophoresis was stopped. The gel was stained and viewed as previously described in section 3.7.3.

The DNA band of the expected size visualized under the UV light was cut from the gel by a clean blade and placed into a new 1.5 mL microcentrifuge tube. The gel matrix that did not contain DNA material was trimmed off to obtain the minimum volume of the gel. DNA was eluted from a slice of gel using QIAquick gel purification kit (Qiagen) according the manufacturer's instruction.

#### 3.7.5 Sequencing of ribosomal DNA

#### **3.7.5.1** Preparation of DNA for sequencing

The purified ribosomal DNA amplicons were sequenced using the BigDye Terminator Ready Reaction kit version 2.0 (Perkin Elmer) according to the manufacturer's protocol. The 10  $\mu$ L cycle sequencing reaction mixture contained 80-200 ng DNA, 4  $\mu$ L BigDye, and 5 pM primer. Primers used for the sequencing of 18S rDNA and ITS fragments, were the same as in PCR amplification of each PCR fragment. The thermal profile consisted of 25 cycles of 10 sec at 96°C, 5 sec at 50°C, and 4 min at 60°C. The cycle sequencing was performed in the thermal cycle (BioRad). The PCR mixtures were spun down briefly before DNA precipitation. The sequencing DNA fragments were precipitated by adding 16  $\mu$ L of deionized water and 64  $\mu$ L of 95% ethanol. The tube was vortexed briefly, incubated at 4°C for 15 min, and then spun at 12,000 rpm for 20 min at 4°C. The DNA pellet was washed with 300  $\mu$ L of 70% ethanol, centrifuged at 12,000 rpm for 20 min at 4°C, and discarded the supernatant. The DNA pellet was dried at room temperature in the dark.

#### 3.7.5.2 DNA sequencing

The sequencing gel used for an ABI 377 automated DNA sequencer (Perkin Elmer) was prepared as described in the manufacturer's protocol. The 6% polyacrylamide gel was casted in slab gel glass plates. The sequencing pellet was dissolved in 3  $\mu$ L of loading buffer, and loaded onto the gel. Electrophoresis was carried out at constant 750 volts for 8 h. Fluorescent signals were detected with ABI Collection software. Base calling was performed using sequencing analysis software, and nucleotide sequence determination was performed using sequence navigator software. The resulting sequences were assembled and manually corrected by using Chromas 1.56 program (Technelysium Pty. Ltd).

#### 3.7.6 Alignment of DNA sequences

Completed DNA sequences were aligned using Clustual X software package (Thompson *et al.*, 1994). All alignments were examined and manually optimized with the BioEdit program (North Carolina State University, U.S.A.). In addition, the available xylariaceous sequences of 18S rDNA and ITS1-5.8S-ITS2 from GenBank database were downloaded, and imported to xylariaceous database examined for DNA sequence alignment.

#### **3.7.7** Construction of phylogenetic tree

Phylogenetic trees were constructed with different methods and software packages.

#### 3.7.7.1 Neighbour-joining (NJ) method

Phylogenetic trees were constructed based on genetic distances using neighbour-joining method. The conditional clustering, Kimura 2 parameter distances (Kimura, 1980), was computed with the Dnadist module of the PHYLIP software package version 3.6 (Felsenstein, 1995). Strengths of internal branches of resulting trees were statistically tested by the bootstrap analysis of 1,000 replications.

#### 3.7.7.2 Maximum parsimony (MP) method

Phylogenetic trees were constructed using PAUP\* version 4.0b10 (Swofford, 2000) for the maximum parsimony method. They were analyzed by heuristic searches. The MAXTREES set to 10,000 and TBR branch swapping. All characters were assessed as independent, unordered and equally weighted. Bootstrapping in these analyses was performed using 1,000 replicates.

### **CHAPTER IV**

# **RESULTS AND DISCUSSION**

#### 4.1 Collection of the xylariaceous fungi for taxonomic studies

Three hundreds and thirty eight xylariaceous specimens were collected from 14 different locations in Thailand. The different locations, period of collection, number and type of specimens were recorded. The number of collected specimens is shown in Table 8. The collection locations were selected following consultation with Dr. Surang Thienhirun as being proven as good forest sites for the Xylariaceae. Although most of the collections are from the Northeastern Thailand sites from the North (Chiang Mai Province) to the South (Songkhla Province) were also surveyed. Thus, a range of different forest types was included in this study.

Year	Location	No. of
		specimens
2002	Chiang Mai Province	11
	Nakhon Ratchasima Province	7
2003	Phu Luang, Nakhon Ratchasima Province	7
	Nong Rawieng, Nakhon Ratchasima Province	11
	Burirum Province	9
	Chaiyaphum Province	4

**Table 8.** Xylariaceous collections from 14 locations of Thailand in years 2002 and2003.

Table 8.(Continued).

Year	Location	No. of
		specimens
2003	Ratchaburi Province	51
	Suranaree University of Technology, Nakhon Ratchasima Province	51
	Kanchanaburi Province	49
	Petchaboon Province	9
	Songkhla Province	30
	Trad Province	77
	Yasothon Province	19
	Chiang Rai Province	3

The majority of collections were from Trad Province where the forest was classified as the mixed forest (Thienhirun, 1997).

# 4.2 Morphological taxonomic studies of the problematic groups in xylariaceous fungi

All collected xylariaceous specimens were identified and classified into nine genera and 59 species according to their morphological characteristics as shown in Table 9.

The high numbers of collected specimens belonged to *Hypoxylon* and *Xylaria* respectively. There was also wide distribution and variation of both genera whereas the other xylariaceous genera were rarely represented especially *Astrocystis*, which occurs only on bamboo. Unfortunately *Daldinia bambusicola* occurring on bamboo was not found during the two years of survey. The details for each genus and species are described as follows:

Genus	No. of specimens	No. of species
Astrocystis Berk. & Broome	8	1
Biscogniauxia Kuntze	6	2
Camillea Fr.	5	1
Daldinia Ces. & De Not.	13	1
Hypoxylon Bull.	196	29
Kretzschmaria Fr.	2	1
Nemania S.F. Gray	2	1
Rosellinia De Not.	4	1
Xylaria Hill ex Schrank	102	22

**Table 9.**Numbers of genera and species of xylariaceous collections in this study in<br/>years 2002 and 2003.

#### 4.2.1 Group I: Astrocystis and Rosellinia

Since the genera *Astrocystis* and *Rosellinia* are poorly represented in Thailand, which agreed with Thienhirun (1997), with only two species having been found in this study, *A. mirabilis* Berk. & Broome and *R. procera* Syd. Comparison of the two species is given in Table 10.

Astrocystis mirabilis SUT047, SUT048, SUT049, SUT051, SUT052, SUT054, SUT055, and SUT056 (Figure 9), which was reported as the type species of the genus (Berkeley and Broom, 1875), were found to be very similar to specimens previously described by Thienhirun (1997) collected from Surat Thani Province except for ascospore size, 10-13.8 x 3.8-5  $\mu$ m cf. (10-)10.6-12.5 x 5.6-6.3  $\mu$ m (Thienhirun, 1997). However other characters were well matched *A. mirabilis* as described by Ju and Rogers (1990).

Character	A. mirabilis*	R. procera*
Stromata		
Shape	Subglobose to hemispherical,	Subglobose to hemispherical,
	blackish, each stroma encircled	blackish, embedded on a brown
	with a more or less stellate to	cottony subiculum
	irregular ring of mixed host and	
	stromatic material at the base to	
	midportion	
Color	Black	Black
Perithecia		
Shape	Obovoid	Obovoid
Size	1-1.5 mm diameter	1-1.5 mm diameter
Ostiole	Papillate	Conico-papillate
Asci	Cylindrical	Not observed
Ascospores		
Color	Brown to dark brown	Brown to dark brown
Shape	Ellipsoid	Fusiform with tapering ends
Size	10-13.8 x 3.8-5 μm	(65-)95-125 x 10-15 μm
Apical apparatus	Inverted hat, 2-3(-4) µm high x	Inverted hat, 2-3 µm high x 3.5-
	3-4 μm broad	5 μm broad
Germ slit	Straight slightly less than spore	Straight longitudinal germ slit
	length	spore length
Culture	White radiate strands with	Uncultured
	fimbriate margins, velutinous or	
	floccose, and faintly zonate	
Habitat	On bamboo	On wood
Location	Ratchaburi	Ratchaburi, Nakhon Ratchasima
Specimen	SUT047, SUT048, SUT049,	SUT102, SUT109, SUT113,
examined	SUT051, SUT052, SUT054,	SUT114
	SUT055, SUT056	

**Table 10.** Species comparison of Astrocystis and Rosellinia found in this study.

\* More details on collections are given in Appendix B.



**Figure 9.** Astrocystis mirabilis Berk. & Broome (SUT051); (a) stromatal form (Bar = 1 mm), (b) ascospores with ellipsoid equilateral (Bar = 10  $\mu$ m), (c) apical apparatus bluing in Melzer's iodine reagent (Bar = 4  $\mu$ m), (d) germ slit straight nearly spore length (arrowed) (Bar = 2  $\mu$ m), and (e) cultural characteristics on PDA cultured at 25°C after 3 weeks (Bar = 1 cm).

Four collections of the *Rosellinia* (SUT102, SUT109, SUT113, and SUT114) examined were in close agreement with *R. procera* Syd. (Figure 10) as described by Petrini (1990) except for small differences in ascospore size (65-)95-125 x 10-15  $\mu$ m cf. 75-130 x 15-18  $\mu$ m (Petrini, 1990). These might be because of variation within the species and the different collection areas. Unfortunately, the *Astrocystis* found in this study did not form its anamorph in culture, and also the *Rosellinia* examined could not be cultured. Therefore, there was no information of anamorph characteristics for both taxa. The genus *Astrocystis* has been separated from *Rosellinia* on the basis of host specificity on bamboo and also on features of the stromata splitting the host surface or the presence of a carbonaceous extension at the base (Berkeley and Broome, 1887; Petrini, 1993; 2003; Læssøe and Spooner, 1994). However, there is some disagreement with Ju and Rogers (1990; 1995) and San Martín and Rogers (1994) considering *Astrocystis* to be congeneric with *Rosellinia*.



Figure 10. *Rosellinia procera* Syd. (SUT113); (a) stromatal form with conico-papillate of ostiole (arrowed) (Bar = 2 mm), (b) perithecia (Bar = 3 mm), (c) ascospore (Bar = 10 μm), and (d) apical apparatus (Bar = 5 μm).

#### 4.2.2 Group II: Camillea

One species of *C. tinctor* (Berk.) Læssøe, J.D. Rogers & Whalley (Figure 11) was recorded from this study. *Camillea selangorensis* M.A. Whalley, A.J.S. Whalley & E.B.G. Jones., which is another tropical species firstly described in Malaysia by Whalley (1995), has since been reported to be found in Thailand (Whalley *et al.*, 1999). Unfortunately, it was not found during this study. Therefore, *C. selangorensis* and *C. leprieurii* (Figure 12), provided by Dr. Margaret A. Whalley were used to compare with the *C. tinctor* collected in Thailand, and their morphological characteristics are described in Table 11.

Four collections (SUT099, SUT161, SUT211, and SUT260) examined collected from different localities in Chiang Mai, Trad, Songkhla, and Yasothorn Provinces closely matched *C. tinctor* (Berk.) Læssøe, J.D. Rogers & Whalley as described by Læssøe *et al.* (1989), and also the previously described specimens from Thailand by Thienhirun (1997). The comparison of all collections, which was collected from different locations, suggested that they were identical. The present of orange color staining on the substratum was also observed from some specimens. Since the ascospores of most *Camillea* species appear smooth by light microscopy but appear characteristically ornamented by SEM (Læssøe *et al.*, 1989; Rogers *et al.*, 1991; San Matín, Gonzáles, and Rogers, 1993; Whalley, 1995; Whalley, 1996; Whalley *et al.*, 1999). The collected *C. tinctor* specimens were, therefore, observed by SEM, and found to exhibit the reticulate ornamentation which was the distinctive character of this species as shown in Figure 11f. However, *C. selangorensis* and *C. leprieurii* revealed strongly verrucose or intricately ornamented ascospore walls as described by Whalley *et al.* (1996) and Læssøe *et al.* (1989) respectively.

Character	C. tinctor (Berk.) Læssøe, J.D. Rogers & Whallev *	C. selangorensis M.A. Whalley, A.J.S. Whalley & E.B.G. Jones.	C. leprieurii Mont.
Stromata			
Shape	Applanate with a slightly raised center, elongate elliptic	Circular to orbicular, or elongated, with applanate to convex apex, surrounded by a slightly raised rim	Erumpent through bark, cylindrical, seated on slightly broader disc
Color	Externally black, mat or shiny, internally dark brown, surface smooth	Black	Black
Perithecia			
Shape	Deeply immersed, cylindrical to slightly elongate	Deeply immersed, brittle entostroma, basally seated, cylindrical, individually erumpent	Elongate ovoid
Size	0.3-1 mm high x 0.2-0.5 mm diameter	0.5-0.8 mm diameter	0.2-0.4 mm diameter
Ostiole	Punctiform, slightly raised	Finely papillate becoming punctate in age	
Asci	Cylindrical	Cylindrical	Cylindrical
Ascospores			
Color	Pale yellow	Pale yellow	Colorless to dilute yellow
Shape	Ellipsoid to fusiform, reticulate-poroid by SEM	Ellipsoid inequilateral, minutely warted by light microscopy, strongly verrucose by SEM	Elongate with upper end acute wedge shaped and lower end draw into very long tail, wavy longitudinal rib-structure with ladder-like transverse substructure by SEM
Size	(12.5-)13.8-21.3 x (5.6-)6.3-8.8 μm	10.0-13.8 x 3.8-6.3 μm	(26.3-)29.1-37.6(-38.5) x (5.3-)6.1-7.5 µm
Apical apparatus	Urniform, 2-3 (-4) $\mu m$ high x 3-4 $\mu m$ broad	Rhomboid, 2.5-3.8 $\mu m$ high x 3-3.8 $\mu m$ broad	Dome or thimble-shaped, 3.3-8.5 (-9.5) high x 4.4-7.3 μm broad
Germ slit	No	No	No
Habitat	On wood	On wood	On wood
Location Specimen examined	Songkhla, Yasothorn, Trad SUT099, SUT161, SUT211, and SUT260	Malaysia (provided by M.A. Whalley) KS15	Malaysia (M.A. Whalley)

**Table 11.** Morphological characteristics of *Camillea tinctor* found in this study, *C. selangorensis*, and *C. leprieurii*.

\* More details on collections are given in Appendix B.



Figure 11. *Camillea tinctor* (Berk.) Læssøe, J.D. Rogers & Whalley (SUT260); (a) stromatal form (Bar = 1 mm), (b) perithecia (Bar = 0.5 mm), (c) ascospores (Bar = 15  $\mu$ m), (d) apical apparatus bluing in Melzer's iodine reagent (Bar = 4  $\mu$ m), (e) cultural characteristics on PDA cultured at 25°C after 4 weeks (Bar = 1 cm), and (f) the reticulate ornamentation of ascospore by SEM (Bar = 2  $\mu$ m).



Figure 12. Camillea selangorensis M.A. Whalley, A.J.S. Whalley & E.B.G. Jones (KS15), and C. leprieurii (Mont.) Mont; (a) and (b) stromatal form of C. leprieurii (Bar = 0.5 cm and 0.2 cm respectively), (c) ascospores (Bar = 6 μm), and (d) stromatal form of C. selangorensis (Bar = 1 cm).

#### 4.2.3 Group III: Daldinia

One species of *D. eschscholzii* (Ehrenb.: Fr.) Rehm was recorded from thirteen collections (SUT013, SUT037, SUT038, SUT039, SUT084, SUT085, SUT086, SUT168, SUT169, SUT178, SUT209, SUT268, and SUT278) of the genus (Figure 13). The type species, *D. concentrica* (Bolton: Fr.) Ces. & De Not. which is frequently found in temperate region, was provided by Prof. Anthony J.S Whalley to compare with collected *D. eschscholzii* specimens. Unfortunately, *D. bambusicola*, which occurs on bamboo, has only been found twice in Thailand and it was not found during this study. The description of the *D. eschscholzii* examined is given in Table 12.

Thirteen collections collected from eight provinces were matched *D.* eschscholzii (Ehrenb.: Fr.) Rehm as described by Ju, Rogers, and San Martín (1997). This is a widely distributed species, and it is the most common xylariaceous species found throughout Thailand as previously reported by Whalley (1996), Thienhirun (1997), and Thienhirun and Whalley (2004). From SEM observation, the ascospore wall of *D. eschscholzii* was ornamented with conspicuous transversely oriented fibrils (Figure 13). However, only teleomorphic characteristics, including perispore ornamentation by SEM, were insufficient to identify species of *D. eschscholzii* or *D. concentrica*. Recently Stadler *et al.* (2004) rearranged the two species of *Daldinia* into five new species according to their anamorph characteristics, shape of ascospores, and chemical characteristics. One out of five species belonging to *D. palmensis* M. Stadler, Wollweber & H-.V. Tichy. is closed to *D. eschscholzii*. The conidiophore of *D. palmensis* is *Nodulisporium*-like or *Sporothrix*-like forms whilst *D. eschscholzii* is a *Nodulisporium*-like form. Unfortunately, only four of the collected *D. eschscholzii*  isolates could be cultured, and all of them were *Nodulisporium*-like in form. The remainder of collections was still unable to confirm to be either *D. eschscholzii* or *D. palmensis*.

 Table 12. Morphological characteristics of Daldinia eschscholzii found in this study

 1.1
 6

Character	D. eschscholzii *	D. concentrica (Ju, Rogers, and San Martín, 1997)**
Stromata		
Shape	Turbinate to placentiform, sessile or with	Spherical, sessile, solitary to
	short, stout stipe, solitary to infrequently	aggregated, smooth or with
	aggregated, smooth	inconspicuous perithecial mounds
Color	Surface brown vinaceous, dark brick, sepia,	Surface brown vinaceous, chestnut, or
	grayish sepia, or vinaceous grey, blackened	sepia, blackened and varnished in age
	and varnished in age	
KOH-extractable	Livid purple, dark livid, or vinaceous purple	Livid purple or dark purple
pigments		
Perithecia		
Shape	Tubular	Tubular
Size	0.8-1.5 mm x 0.3-0.4 mm diameter	0.3-0.5 mm diameter x 1-2 mm high
Ostiole	Obsolete or slightly papillate	Slightly papillate
Asci	Cylindrical	Cylindrical
Ascospores		
Color	Brown to dark brown	Brown to dark brown
Shape	Ellipsoid-inequilateral with narrowly rounded	Ellipsoid-inequilateral with narrowly
	ends	rounded ends
Size	11.3-13.8 x 5-6.3 μm	13-17 x 6-7.5 μm
Apical apparatus	Discoid, 0.5 µm high x 2-2.5 µm broad	Discoid, 0.5-1 $\mu$ m high x 3-3.5 $\mu$ m
		broad
Germ slit	Straight full spore-length on convex side	Slightly sigmoid germ slit spore-
		length on convex side
Perispore	Dehiscent, conspicuous coil-like	Dehiscent, smooth
	ornamentation	
Culture	White at first, becoming brownish grey,	-
	fluffy, rapidly grow	
Location	Bangkok, Burirum, Chiang Rai, Nakhon	-
	Ratchasima, Ratchaburi, Trad, and Yasothorn	
Specimen examined	SUT013, SUT037, SUT038, SUT039,	L1 and L2
	SUT084, SUT085, SUT086, SUT168,	
	SUT169, SUT178, SUT209, SUT268,	
	SUT278	

and the reference specimen of *D. concentrica*.

\* More details on collections are given in Appendix B.

\*\* Typical specimens with anamorphic culture from the U.K.



Figure 13. *Daldinia eschscholzii* (Ehrenb.: Fr.) Rehm (SUT039); (a) stromatal form (Bar = 1 cm), (b) perithecia (Bar = 0.5 mm), (c) ascus containing eight ascospores (Bar = 10  $\mu$ m), (d) apical apparatus bluing in Melzer's iodine reagent (arrowed), (e) germ slit straight nearly spore length (arrowed) (Bar = 2  $\mu$ m), (f) ascospore dehiscent in 10% KOH (arrowed), (g) cultural characteristics on PDA cultured at 25°C after 4 weeks (Bar = 1 cm), and (h) the perispore with conspicuous coil-like ornamentation by SEM (Bar = 2  $\mu$ m).

#### 4.2.4 Group IV: Hypoxylon

Twenty nine species of *Hypoxylon* were recorded including 4 new species. Although *Hypoxylon* sect. *Annulata* was focused in this study, other species of *Hypoxylon* sect. *Hypoxylon* also were examined for species differentiation and DNA database construction. The *Hypoxylon* species found and investigated are listed in Table 13.

 Table 13. Species of Hypoxylon sect. Annulata, and sect. Hypoxylon found in this study.

Species	No.*	Remark
Hypoxylon sect. Annulata		
H. cf. archeri	6	Hazel in KOH-extracted pigments,
		white substance on the stromatal
		surface
H. atroroseum J.D. Rogers	10	Ju and Rogers (1996)
H. bovei Speg. var. microspora J.H. Miller	1	Ju and Rogers (1996)
H. moriforme Henn.	5	Ju and Rogers (1996)
H. purpureonitens YM. Ju & J.D., Rogers	12	Ju and Rogers (1996)
H. stygium (Lév.) Sacc.	13	Ju and Rogers (1996)
H. urceolatum (Rehm) YM. Ju & J.D. Rogers	1	Ju and Rogers (1996)
Hypoxylon taxonomic species 1 sp. nov.	15	Green in KOH-extracted pigments,
		truncatum-type in ostiolar form,
		0.3-0.4 mm diameter of ostiolar disc
Hypoxylon sect. Hypoxylon		
H. anthochroum Berk. & Broome	7	Ju and Rogers (1996)
H. brevisporum YM. Ju & J.D. Rogers	1	Ju and Rogers (1996)
H. duranii J.D. Rogers	11	Ju and Rogers (1996)
H. fendleri Berk. ex Cooke	20	Ju and Rogers (1996)
H. cf. ferrugineum (SUT017)	1	Small in ascospore size
H. cf. ferrugineum (SUT070)	1	Brown vinaceous in stromatal
		surface colour, orange in granule
		colour
H. cf. ferrugineum (SUT237)	4	Close to H. ferrugineum except for
		stromatal surface color, KOH-
		extractable pigment, and ascospore
		size
* Normaliser of II and a straight		

\* Number of *Hypoxylon* collections.
Table 13. (Continued)

Species	No.*	Remark
Hypoxylon sect. Hypoxylon		
H. haematostroma Mont. apud Sagra	7	Ju and Rogers (1996)
H. hypomiltum Mont.	1	Ju and Rogers (1996)
H. investiens (Schwein.) M.A. Curtis	7	Ju and Rogers (1996)
H. lenormandii Berk. & M.A. Curtis apud Berk	11	Ju and Rogers (1996)
H. lenormandii var. microspora	1	Thienhirun (1997)
H. macrocarpum Pouzar	1	Ju and Rogers (1996)
H. monticulosum Mont.	28	Ju and Rogers (1996)
H. cf. perforatum (SUT020)	1	Grayish sepia in stromatal surface colour, dark brown to black in granule colour, straight full length in
		germ slit
H. cf. perforatum (SUT224)	1	Brown vinaceous in stromatal surface colour, brown vinaceous in granule colour, straight full length in germ slit
H. cf. perforatum (SUT294)	1	Reddish brown in stromatal surface colour, reddish brown to black in granule colour, straight full length in germ slit
H. rubiginosum (Pers.: Fr.) Fr., Summa Veg	4	Ju and Rogers (1996)
H. subgilvum Berk. & Broome var. microsporum (Abe) YM. Ju & J.D. Rogers	3	Ju and Rogers (1996)
H. trugodes Berk. & Broome	6	Ju and Rogers (1996)
H. sublenormandii sp. nov.	3	Closed to <i>H. lenormandii</i> except for reddish brown in stromatal colour, smaller in ascospore size, and straight germ slit
H. kanchanapisekii sp. nov.	5	Close to <i>H. lenormandii</i> except for stromatal surface color of dull reddish brown not grayish sepia, small ascospores, and having a straight germ slit
H. suranareei sp. nov.	5	Conspicuous perithecial mounds, orange brown in stromatal surface colour, yellowish orange in KOH- extractable pigments, ostioles same or lower than the stromatal surface with white substance

\* Number of *Hypoxylon* collections.

 Table 13. (Continued)

Species	No.*	Remark
Hypoxylon sect. Hypoxylon		
Hypoxylon taxonomic species 2 (SUT082)	1	Brownish yellow in KOH- extractable pigment, ascospore size, and inconspicuous coil-like ornamentation
Hypoxylon taxonomic species 3 (SUT158)	1	Dark brick or brown vinaceous in stromatal surface colour, brown vinaceous in granule colour, amber or yellowish brown in KOH- extracted pigments

\* Number of *Hypoxylon* collections.

#### 4.2.4.1 Hypoxylon section Annulata

Eight species of *Hypoxylon* sect. *Annulata* were observed including a new species, *Hypoxylon* taxonomic species 1 sp. nov. The results are given in Table 14. *Hypoxylon* cf. *archeri* (SUT079, SUT103, SUT105, and SUT112) (Figure 14) closely agreed with *H. archeri* Berk. *apud* J.D. Hook. as described by Ju and Rogers (1996) except for its KOH-extractable pigments having greenish olivaceous according to Ju and Rogers (1996) but they were hazel in the Thai collections. In addition, two specimens related to this taxon provided by Dr. Surang Thienhirun (ST2333 and ST2527) were used as the reference strains. Initially, the specimens examined looked like *H. michelianum* in having a layer of white substance on the stromatal surface, which was striking and has not been observed in other taxa of section *Annulata* (Ju and Rogers, 1996) but the other characters were different.

Character	H. cf. archeri*	H. atroroseum J.D. Rogers*	H. bovei Speg. var. microspora J.H.
			Miller*
Stromata			
Shape	Effused-pulvinate	Effused-pulvinate	Hemispherical to effused-pulvinate
Color	Blackish brown	Vinaceous gray	Black
Granules beneath	Black	Dull reddish brown	Black
surface			
KOH pigments	Hazel	Greenish olivaceous	Greenish olivaceous
Perithecia			
Shape	Spherical	Obovoid	Spherical
Size	0.3-0.4 mm diameter	0.2-0.3 mm diameter x 0.3-0.5 mm high	0.6-1 mm diameter
Ostiole	Coarsely papillate	Papillate	Papillate
Disc			
Туре	Truncatum-type	Truncatum-type	<i>Bovei</i> -type
Size	0.1-0.2 mm diameter	0.1-0.2 mm diameter	0.3-0.7 mm diameter
Apical apparatus	Discoid, 0.5 µm high x 1-1.5 µm broad	Discoid, 0.5 µm high x 1 µm broad	Discoid, 1-1.5 µm high x 2 µm broad
Ascospores			
Color	Brown to dark brown	Light brown	Brown to dark brown
Shape	Ellipsoid-inequilateral, with narrowly	Ellipsoid-equilateral, with narrowly rounded	Ellipsoid-inequilateral, with narrowly
	rounded ends	ends	rounded ends
Size	8.8-10(-11.5) x 3.8-5 μm	6.3-8.8 x 2.5-3.8 μm	7.5-10 x 3.8-5 μm
Germ slit	Straight full length	Straight full length	Straight full length
Perispore	Smooth	Smooth	Smooth
Habitat	On wood	On wood	On wood
Location	Songkhla Province	Nakhon Ratchasima, Trad	Chaiyaphum
Specimen examined	SUT079, SUT103, SUT105, and	SUT009, SUT010, SUT214, and SUT219	SUT025 and SUT242
-	SUT112		

**Table 14.** Morphological characteristics of *Hypoxylon* sect. *Annulata* found in this study.

 Table 14. (Continued).

Character	H. moriforme Henn.*	H. purpureonitens YM. Ju & J.D., Rogers*	H. stygium (Lév.) Sacc.*
Stromata			
Shape	Glomerate, hemispherical to effused-	Glomerate, hemispherical to effused-pulvinate	Effused-pulvinate
	pulvinate		
Color	Blackish with reddish brown tone, some	Blackish with reddish brown tone, some shiny	Blackish with reddish brown tone
	shiny black	black	
Granules beneath	Black	Black	Dull reddish brown
surface			
KOH pigments	Greenish olivaceous	Vinaceous purple	Greenish olivaceous
Perithecia			
Shape	Spherical	Spherical	Obovoid
Size	(0.4-)0.5-1(-1.2) mm diameter	(0.3-)0.5-1 mm diameter	0.2-0.3 mm diameter x 0.3-0.5 mm high
Ostiole	Conical-papillate	Conical-papillate	Papillate
Disc			
Туре	<i>Bovei</i> -type	<i>Bovei</i> -type	Truncatum-type
Size	0.2-0.3 mm diameter	0.2-0.3 mm diameter	0.1-0.2 mm diameter
Apical apparatus	Discoid, 0.5 µm high x 1-1.5 µm broad	Discoid, 0.5 µm high x 1-1.5 µm broad	Discoid, 0.5 µm high x 1µm broad
Ascospores			
Color	Brown	Brown	Light brown
Shape	Ellipsoid-inequilateral, with narrowly	Ellipsoid-inequilateral, with narrowly rounded	Ellipsoid-equilateral, with narrowly
	rounded ends	ends	rounded ends
Size	7.5-9 x 2.8-4.2 μm	7.5-10 x 3.8-5 μm	3.8-6.3 x 2.5-3.8 μm
Germ slit	Straight full length	Straight full length	Straight full length
Perispore	Smooth	Smooth	Smooth
Habitat	On wood	On wood	On wood
Location	Chaiyaphum, Kanchanaburi, Nakhon	Nakhon Ratchasima, Songkhla, Trad, Yasothorn	Ratchaburi, Trad
	Ratchasima, Trad		
Specimen examined	SUT216, SUT220, SUT231, SUT249,	SUT001, SUT004, SUT005, SUT100, SUT160,	SUT058, SUT222, SUT226, SUT229,
-	SUT285, and SUT288	SUT167, and SUT262	SUT230, SUT243, SUT245, SUT247,
	·		SUT253, and SUT257

Table 14.	(Continued).
-----------	--------------

Character	H. urceolatum (Rehm) YM. Ju & J.D. Rogers *	Hypoxylon taxonomic species 1 sp. nov.*
Stromata		
Shape	Effused-pulvinate	Glomerate, hemispherical to effused-pulvinate
Color	Black	Blackish with reddish brown tone, some shiny black
Granules beneath surface	Black	Black
KOH pigments	Vinaceous purple	Green
Perithecia		
Shape	Obovoid to tubular	Spherical
Size	0.2-0.4 mm diameter x 0.4-1 mm high	0.5-0.8 mm diameter
Ostiole	Conical-papillate	Conical-papillate
Disc		
Туре	Truncatum-type	Truncatum-type
Size	0.2-0.3 mm diameter	0.2-0.3 mm diameter
Apical apparatus	Not observed	Discoid, 0.5 µm high x 1-1.5 µm broad
Ascospores		
Color	Pale brown	Brown
Shape	Ellipsoid to fusoid, slightly inequilateral to equilateral,	Ellipsoid-inequilateral, with narrowly rounded ends
	with narrowly rounded ends	
Size	10-12.5 x 2.5-5 μm	7.5-9 x 2.8-4.2 μm
Germ slit	Straight less than spore-length and originating from one	Straight full length
	end	
Perispore	Smooth	Smooth
Habitat	On wood	On wood
Location	Songkhla	Chaiyaphum, Nakhon Ratchasima, Trad, Kanchanaburi
Specimen examined	SUT098	SUT081, SUT238, SUT241, SUT244, SUT246, SUT251, and
		SUT255

*Hypoxylon atroroseum* SUT009, SUT010, SUT214, and SUT219 (Figure 15) examined were similar to *Hypoxylon atroroseum* J.D. Rogers as described by Ju and Rogers (1996) except for its ascospore size, which were 6.3-8.8 x 2.5-3.8  $\mu$ m and 5-7 x 2-3  $\mu$ m respectively. The teleomorphic characteristics of *H. atroroseum* were similar to *H. stygium* except the stromata of *H. atroroseum* often have rosy surface tones.

*Hypoxylon bovei* var. *microspora* SUT242 (Figure 16) examined was similar to *Hypoxylon bovei* Speg. var. *microspora* J.H. Miller. as described by Ju and Rogers (1996).

*Hypoxylon moriforme* SUT216, SUT220, SUT231, SUT249, SUT285, and SUT288 (Figure 17) were similar to *Hypoxylon moriforme* Henn. (Ju and Rogers, 1996). This taxon closely resembles *H. nitens* and *H. bovei* var. *microspora* in KOH-extractable pigments and size of perithecia, disc, and ascospores but they are different in ostiolar disc type. *Hypoxylon moriforme* has a *truncatum*-type disc whilst the other species have a *bovei*-type disc (Ju and Rogers, 1996). For the *truncatum*-type, the outermost layer of stroma around ostioles is flaked off gradually from the ostiole outwards, whereas in the *bovei*-type the outermost layer of stroma dehisces abruptly. However, in the case of mature specimens lacking these outer layers, the ostiolar discs of *bovei*-type look like the *truncatum*-type disc and as a result were difficult to identify. The collected specimens examined were placed in this taxon because of the lack of these outer layers and as their ostiolar discs were identified as belonging to the *truncatum*-type. They also mainly formed glomerate stromata.



Figure 14. *Hypoxylon* cf. *archeri* (SUT105); (a) stromatal form when immature (Bar = 0.2 mm), (b) KOH-extractable pigment, (c) stromata with white fringe surrounding ostiolar disc (Bar = 0.1 mm), (d) ascospores (Bar = 10 μm), (e) straight germ slit spore length (arrowed) (Bar = 2 μm), (f) the thickening on perispore (arrowed) (Bar = 1 μm), (g) SEM micrograph of ascospore (Bar = 1 μm), and (h) cultural characteristics on PDA cultured at 25°C after 3 weeks (Bar = 1 cm).



**Figure 15.** *Hypoxylon atroroseum* J.D. Rogers (SUT009); (a) stromatal form (Bar = 0.1 mm), (b) ascospores (Bar = 10  $\mu$ m), (c) KOH-extractable pigment greenish olivaceous, (d) straight germ slit spore length (arrowed) (Bar = 2  $\mu$ m), (e) the thickening on perispore (arrowed) (Bar = 2  $\mu$ m), and (f) cultural characteristics on PDA cultured at 25°C after 4 weeks (Bar = 1 cm).



**Figure 16.** *Hypoxylon bovei* Speg. var. *microspora* J.H. Miller (SUT025); (a) and (c) stromatal form (Bars = 1 cm and 0.5 mm respectively), (b) KOH-extractable pigment, (d) ascospores (Bar =  $10 \ \mu m$ ), (e) the thickening on perispore (arrowed) (Bar =  $5 \ \mu m$ ), and (f) cultural characteristics on PDA cultured at  $25^{\circ}$ C after 3 weeks (Bar = 1 cm).



**Figure 17.** *Hypoxylon moriforme* Henn. (SUT220); (a) and (c) stromatal form (Bars = 1 cm and 0.3 mm respectively), (b) KOH-extractable pigment greenish olivaceous, (d) ascospores (Bar = 10  $\mu$ m), (e) straight germ slit spore length (arrowed) (Bar = 2  $\mu$ m), and (f) cultural characteristics on PDA cultured at 25°C after 4 weeks (Bar = 1 cm).

*Hypoxylon purpureonitens* SUT001, SUT004, SUT005, SUT100, SUT160, SUT167, and SUT262 (Figure 18) matched *Hypoxylon purpureonitens* Y.-M. Ju & J.D. Rogers (Ju and Rogers, 1996). This taxon is similar to *H. nitens* except its KOH-extractable pigments are purplish (Ju and Rogers, 1996).

*Hypoxylon stygium* SUT058, SUT222, SUT226, SUT229, SUT230, SUT243, SUT245, SUT247, SUT253, and SUT257 (Figure 19) from Thailand fitted *Hypoxylon stygium* (Lév.) Sacc. (Ju and Rogers, 1996). The ascospore size of collected specimens was 3.8-6.3 x 2.5-3.8 μm but *Hypoxylon stygium* (Lév.) Sacc. was 5-7 x 2-3 μm.

*Hypoxylon urceolatum* SUT098 (Figure 20) matched *Hypoxylon urceolatum* (Rehm) Y.-M. Ju & J.D. Rogers as described by Ju and Rogers (1996) except the ascospore size (10-12.5 x 2.5-5  $\mu$ m), which was smaller than specimens recorded by Ju and Rogers (1996) (9-14(-17) x 3.5-4.5  $\mu$ m) but it was close to Thai specimens reported by Thienhirun (1997) (8.8-10 x 3-3.8  $\mu$ m). The cultural characteristics of this taxon have never been observed. In this study, the specimen was cultured on PDA and hypha covered a 9-cm Petri dish in 3 weeks at 25°C. At first, the mycelium was white. Then, it became dull green, floccose, azonate, with diffuse margins, with scattered black patches as shown in Figure 20f. No anamorph was observed.

*Hypoxylon* taxonomic species 1 sp. nov. (Figure 21). Characteristics of this taxon are as follows: stromata glomerate, hemispherical to effused-pulvinate, with perithecial mounds; surface blackish, with reddish brown tone; blackish granules beneath surface, with KOH-extractable pigments greenish olivaceous (90); perithecia spherical, 0.5-0.8 mm diameter, ostioles papillate, encircled with a flattened *truncatum*-type disc 0.3-0.5 mm diameter; asci 100-130  $\mu$ m total length x 3.8-5  $\mu$ m broad, the spore bearing parts 40-65  $\mu$ m long with stipes 30-55  $\mu$ m; ascospores brown to dark brown, unicellular, ellipsoid-inequilateral, with narrowly rounded ends, 7.5-9 x 2.8-4.2  $\mu$ m, with straight-germ slit spore length; perispore dehiscent in 10% KOH, smooth; epispore smooth.

Specimens examined: Thailand, Trad Province, 14 December 2003, Suwannasai, N. (Holotype SUT236), SUT238, SUT241, SUT244, SUT246, SUT251, and SUT255; Chaiyaphum Province (SUT025); Nakhon Ratchasima Province (SUT081).

Colonies on PDA covering 9 cm Petri dish in two weeks at room temperature, 23-28°C, at first white then dull green, floccose, azonate, with diffuse margins, with scattered black patches. Anamorph not formed.

This species was close to *Hypoxylon nitens* (Ces.) Y.-M. Ju & J.D. Rogers. (Ju and Rogers, 1996). Some specimens examined however were shiny black but some were matt. The type of ostiolar disc was *truncatum*-type but *Hypoxylon nitens* (Ces.) Y.-M. Ju & J.D. Rogers was *bovei*-type.



**Figure 18.** *Hypoxylon purpureonitens* Y.-M. Ju & J.D. Rogers (SUT004); (a) stromatal form (Bar = 0.3 mm), (b) ascospores (Bar = 5  $\mu$ m), (c) KOH-extractable pigment vinaceous purple, (d) ascospore dehiscent in 10% KOH (arrowed) (Bar = 2  $\mu$ m), (e) straight germ slit spore length (arrowed) (Bar = 2  $\mu$ m), and (f) cultural characteristics on PDA cultured at 25°C after 4 weeks (Bar = 1 cm).



**Figure 19.** *Hypoxylon stygium* (Lév.) Sacc. (SUT058); (a) stromatal form (Bar = 0.2 mm), (b) KOH-extractable pigment greenish olivaceous, (c) ascospores dehiscent in 10% KOH (arrowed) (Bar = 2  $\mu$ m), (d) ascospores (Bar = 5  $\mu$ m), and (e) cultural characteristics on PDA cultured at 25°C after 4 weeks (Bar = 1 cm).



Figure 20. Hypoxylon urceolatum (Rehm) Y.-M. Ju & J.D. Rogers (SUT098); (a) stromatal form (Bar = 1 cm), (b) KOH-extractable pigment vinaceous purple, (c) stromatal form (Bar = 0.3 mm), (d) perithecia (Bar = 0.4 mm), (e) ascospores (Bar = 10 μm), and (f) cultural characteristics on PDA cultured at 25°C after 4 weeks (Bar = 1 cm).



**Figure 21.** *Hypoxylon* taxonomic species 1 sp. nov. (SUT236); (a) stromatal form (Bar = 0.4 mm), (b) KOH-extractable pigment greenish olivaceous, (c) apical apparatus (arrowed) (Bar = 1  $\mu$ m), (d) ascospore dehiscent in 10% KOH (arrowed) (Bar = 2  $\mu$ m), (e) straight germ slit spore length (arrowed) (Bar = 2  $\mu$ m), (f) ascospores (Bar = 10  $\mu$ m), and (g) cultural characteristics on PDA cultured at 25°C after 4 weeks (Bar = 1 cm).

#### 4.2.4.2 Hypoxylon section Hypoxylon

Twenty one species of *Hypoxylon* sect. *Hypoxylon* (Table 15) were observed. Three of them were described as new species, *Hypoxylon* sublenormandii sp. nov., *Hypoxylon kanchanapisekii* sp. nov., and *Hypoxylon* suranareei sp. nov.

*Hypoxylon anthochroum* SUT233, SUT240, and SUT263 (Figure 22) examined were virtually identical to *Hypoxylon anthochroum* Berk. & Broome as described by Ju and Rogers (1996).

*Hypoxylon brevisporum* SUT256 (Figure 23) examined closely resembled the species *Hypoxylon brevisporum* Y.-M. Ju & J.D. Rogers as described by Ju and Rogers (1996) except for the KOH-extractable pigments, which were hazel or blackish brown and olivaceous grey or greenish olivaceous respectively.

*Hypoxylon duranii* SUT223, SUT239, SUT248, SUT252, SUT254, SUT259, and SUT284 (Figure 24) examined fitted *Hypoxylon duranii* J.D. Rogers (Ju and Rogers, 1996) except the ascospores which were 8.8-10(-11.3) x 2.8-5  $\mu$ m and 9.5-13(-14.5) x 4.5-6.5  $\mu$ m respectively. However, the ascospore size of these collections was similar to those of specimens found in Thailand (Thienhirun, 1997).

*Hypoxylon fendleri* SUT040, SUT061, SUT120, SUT145, SUT159, SUT162, SUT163, SUT165, and SUT280 (Figure 25) examined closely resembled *Hypoxylon fendleri* Berk. *ex* Cooke as described by Ju and Rogers (1996) except the germ slit form which was sigmoid. Initially, one of specimens, SUT120, was placed to *H. retpela* because the germ slit form was straight to slightly sigmoid. After observing perispore ornamentation by SEM, the specimen exhibited inconspicuous coil-like ornamentation, which was a character of *H. fendleri*. Therefore, the specimen SUT120 was considered to be *H. fendleri*. This taxon is similar to *H. retpela*, and these are the only two *Hypoxylon* taxa with a vinaceous stromatal surface among the *Hypoxylon* taxa with orange or orange red granules inside the stromata. These two species differ mainly in the conspicuousness of the ornamentation on the perispore (Ju and Rogers, 1996).

Hypoxylon cf. ferrugineum SUT017 (Figure 26), H. cf. ferrugineum SUT070 (Figure 27), and H. cf. ferrugineum SUT237 (Figure 28) were similar to H. ferrugineum Otth. (Ju and Rogers, 1996). Hypoxylon cf. ferrugineum SUT070 differed in stromatal surface color (dark brick or hazel), and in granule color (rusty brown or ochraceous brown) whereas H. cf. ferrugineum SUT017 differed in ascospore size 12.5-15(-17.5) x 5-7.5  $\mu$ m cf. (13.5-)14-17 x 6.5-8(-8.5)  $\mu$ m (Ju and Rogers, 1996). Hypoxylon cf. ferrugineum (SUT237) differed in stromatal surface color. In addition, this taxon was different from H. cf. ferrugineum (SUT070) in stromatal form and KOH-extractable pigment colour. Although, H. ferrugineum was placed as a variety of H. rubiginosum by Miller (1961), it was recognised as a different species based on habitat of stromata, distribution of granules, colors of the tissue below the perithecial layer, and ascospore size range, (8-)9-12 x 4-5.5  $\mu$ m (Ju and Rogers, 1996). Nevertheless, H. ferrugineum has been found in Swiss and U.S.A. It has never been reported in Southeast Asia.

Hypoxylon haematostroma SUT062, SUT064, SUT164, SUT292, and SUT293 (Figure 29) examined fitted Hypoxylon haematostroma Mont. apud Sagra as described by Ju and Rogers (1996) but they differed from H. haematostroma as reported by Thienhirun (1997), which had smaller ascospores 13-17.9 x 6.3-8.6  $\mu$ m cf. 12.5-13.8 x 6.3-7.5  $\mu$ m (Thienhirun, 1997).

Character	H. anthochroum Berk. & Broome*	H. brevisporum YM. Ju & J.D. Rogers*	H. duranii J.D. Rogers*
Stromata			
Shape	Effused-pulvinate	Effused-pulvinate	Glomerate, restricted-pulvinate to effused-
			pulvinate
Color	Chestnut or brown vinaceous	Brown vinaceous	Brown vinaceous
Granules beneath	Brown to blackish	Black	Reddish brown
surface			
KOH pigments	Olivaceous	Hazel or blackish brown	Isabelline or yellowish brown
Perithecia			
Shape	Obovoid	Obovoid to tubular	Spherical to obovoid
Size	0.2-0.3(-0.4) mm diameter x 0.3-0.6	0.2 mm diameter x 0.3-0.7 mm high	0.1-0.3 mm diameter x 0.2-0.5 mm high
	mm high		
Ostiole	Lower than the stromatal surface	Lower than the stromatal surface, with white	Lower than the stromatal surface
		substance	
Apical apparatus	Discoid, 0.5 µm high x 2-2.5 µm broad	Not observed	Discoid, 0.8-1.5 µm high x 2-3µm broad
Ascospores			
Color	Brown to dark brown	Light brown to brown	Brown to dark brown
Shape	Ellipsoid-inequilateral, with narrowly	Ellipsoid-equilateral, with narrowly rounded	Ellipsoid-inequilateral, with narrowly rounded
	rounded ends	ends	ends
Size	10.8-13(-14) x 4-6 μm	6.1-7.2 x 2.7-3.7 μm	8.8-10(-11.3) x 3.8-5 μm
Germ slit	Straight full length	Straight full length	Straight full length
Perispore	Dehiscent, with inconspicuous coil-like	Smooth	Dehiscent, with very conspicuous coil-like
	ornamentation		ornamentation
Habitat	On wood	On wood	On wood
Location	Trad	Nakhon Ratchasima, Trad	Kanchanaburi, Trad
Specimen examined	SUT233, SUT240, and SUT263	SUT256	SUT223, SUT239, SUT248, SUT252,
			SUT254, SUT259, and SUT284

**Table 15.** Morphological characteristics of *Hypoxylon* sect. *Hypoxylon* found in this study.

#### Table 15. (Continued).

Character	H. fendleri Berk. ex Cooke*	H. cf. ferrugineum (SUT017)*	H. cf. ferrugineum (SUT070)*
Stromata			
Shape	Effused-pulvinate	Hemispherical, pulvinate to effused- pulvinate	Effused-pulvinate
Color	Brown vinaceous or dark brick	Hazel	Brown vinaceous
Granules beneath surface	Orange red	Yellowish orange	Brown vinaceous
KOH pigments Perithecia	Orange	Orange	Orange
Shape	Obovoid	Obovoid	Obovoid
Size	0.2-0.4 mm diameter x 0.3-0.6 mm high	0.2-0.4 mm diameter x 0.3-0.5 mm high	0.2-0.4 mm diameter x 0.3-0.5 mm high
Ostiole	Lower than the stromatal surface	Lower than the stromatal surface, usually with white substance	Lower than the stromatal surface
Apical apparatus	Discoid, 0.5-1.2 µm high x 1.8-2.5 µm broad	Not observed	Not observed
Ascospores			
Color	Brown to dark brown	Brown to dark brown	Dark brown
Shape	Ellipsoid-inequilateral, with narrowly rounded ends	Ellipsoid-inequilateral, with narrowly rounded ends	Ellipsoid-inequilateral, with narrowly rounded ends, infrequently with one or two ends pinched
Size	8.75-11.25(-12.5) x 3.75-5 μm	12.5-15(-17.5) x 5-7.5 μm	(15-)16.3-17.5 x 7.5 μm
Germ slit	Slightly sigmoid full length	Straight full length	Straight full length
Perispore	Dehiscent, with inconspicuous coil-like ornamentation	Dehiscent, with inconspicuous coil-like ornamentation	Dehiscent, with conspicuous coil-like ornamentation
Habitat	On wood	On wood	On wood
Location	Kanchanaburi, Nakhon Ratchasima, Ratchaburi, Yasothorn	Burirum	Ratchaburi
Specimen examined	SUT040, SUT061, SUT120, SUT145, SUT159, SUT162, SUT163, SUT165, and SUT280	SUT017	SUT070

Lable 13. (Continued
----------------------

Character	H. cf. ferrugineum (SUT237)*	H. haematostroma Mont. apud Sagra*	H. hypomiltum Mont.*
Stromata			
Shape	Glomerate	Hemispherical to effused-pulvinate	Effused-pulvinate
Color	Brown vinaceous or rusty brown	Orange red or rust	Dark brick
Granules beneath surface	Brown vinaceous	Reddish brown	Dull rusty brown
KOH pigments	Orange	Orange red	Amber or yellowish brown
Perithecia	-		
Shape	Obovoid	Long tubular	Obovoid
Size	0.2-0.4 mm diameter x 0.3-0.5 mm high	0.2-0.5 mm diameter x 1.8-2.2 mm high	0.3-0.5 mm diameter x 0.5-0.7 mm high
Ostiole	Lower than the stromatal surface	Lower than the stromatal surface	Lower than the stromatal surface
Apical apparatus	Discoid, 0.5 µm high x 2.7-3.4 µm broad	Discoid, 2.5-3 $\mu$ m high x 1.3-1.5 $\mu$ m broad	Discoid, 0.3-0.6 $\mu$ m high x 1.2-1.5 $\mu$ m broad
Ascospores			
Color	Brown to dark brown	Brown to dark brown	Light brown to brown
Shape	Ellipsoid-inequilateral, with narrowly rounded ends	Ellipsoid-inequilateral, with narrowly rounded ends	Nearly equilateral, with nearly acute ends
Size	(12.2)-13.4-17.8 x 5.3-8.3 μm	13-17.9 x 6.3-8.6 μm	7.5-8 x 2.5-3.8 μm
Germ slit	Straight full length	Slightly sigmoid full length	Straight full length
Perispore	Dehiscent, with conspicuous coil-like ornamentation	Dehiscent, smooth	Dehiscent, smooth
Habitat	On wood	On wood	On wood
Location	Trad	Kanchanaburi, Ratchaburi, Yasothorn	Yasothorn
Specimen examined	SUT237	SUT062, SUT064, SUT164, SUT292, and SUT293	SUT166

Character	H. investiens (Schwein.) M.A. Curtis*	H. lenormandii Berk. & M.A. Curtis apud Berk.*	H. lenormandii var. microspora (Thienhirun, 1997)*
Stromata			
Shape	Effused-pulvinate	Glomerate to effused-pulvinate with the tendency to be perithecioid	Effused-pulvinate, with the tendency to be perithecioid
Color	Brown vinaceous or chestnut	Grayish sepia	Blackish brown
Granules beneath surface	Black	Dull orange brown to dark brown	Black
KOH pigments	Dull green	Red	Reddish brown
Perithecia			
Shape	Obovoid to tubular	Spherical	Spherical
Size	0.3-0.4 mm diameter x 0.5-1 mm high	0.3-0.5 (-0.6) mm diameter	0.5-0.8 mm diameter
Ostiole	Lower than the stromatal surface	Slightly higher than the stromatal surface	Coarsely papillate
Apical apparatus	Not observed	Discoid, 0.7-1.5 µm high x 2-3 µm broad	Discoid, 0.5 µm high x 1-1.5 µm broad.
Ascospores			
Color	Light brown to brown	Brown to dark brown	Brown to dark brown
Shape	Ellipsoid, nearly equilateral with broadly rounded ends	Ellipsoid-inequilateral to equilateral, with narrowly rounded ends	Ellipsoid-inequilateral, with narrowly rounded ends
Size	7.3-8.8 x 2.5-3.8 μm	10-12.5 x 3.8-5 μm	5-6.3 x 2.5-3.8 μm
Germ slit	Straight less than length	Slightly sigmoid full length	Straight full length
Perispore	Indehiscent	Dehiscent, with inconspicuous coil-like ornamentation	Dehiscent, smooth
Habitat	On wood	On wood	On wood
Location	Nakhon Ratchasima, Ratchasima	Burirum, Kanchanaburi, Nakhon Ratchasima	Chaiyaphum
Specimen examined	SUT041 and SUT063	SUT016, SUT065, SUT144, SUT147, SUT151, SUT180, SUT181, and SUT283	SUT022

 Table 15. (Continued).

# Table 15. (Continued).

Character	H. macrocarpum Pouzar*	H. monticulosum Mont.*	H. cf. perforatum (SUT020)*
Stromata			
Shape	Effused-pulvinate	Pulvinate to effused-pulvinate	Hemispherical, pulvinate to effused- pulvinate
Color	Brown vinaceous	Rust, brown vinaceous then blackish when mature	Grayish sepia
Granules	Brown vinaceous	Black	Dark brown or black
KOH pigments	Hazel or yellowish brown	Colorless or purple	Amber or yellowish brown
Perithecia			
Shape	Obovoid	Obovoid	Spherical
Size	0.17-0.2 mm diameter x 0.6-0.9 mm high	0.2-0.5 mm diameter x 0.3-0.5 mm high	0.1-0.3 mm diameter
Ostiole	Slightly higher than the stromatal surface	Higher than the stromatal surface and minutely papillate	Lower than the stromatal surface
Apical apparatus	Not observed	Discoid, 1 $\mu$ m high x 2 $\mu$ m broad	Discoid, 0.5-1.8 µm high x 2-2.8 µm broad
Ascospores			
Color	Brown to dark brown	Brown to dark brown	Brown to dark brown
Shape	Ellipsoid-inequilateral to equilateral, with narrowly rounded ends	Ellipsoid-inequilateral, with narrowly rounded ends	Ellipsoid-inequilateral to equilateral, with narrowly rounded to end pinched
Size	8.8-11.3 x 3.8-5 μm	(6.3-)7.5-8.8(-11.3) x 3.8-5µm	(7.5-)8.8-10 x 5-6.3 μm
Germ slit	Straight full length	Slightly sigmoid full length	Straight full length
Perispore	Dehiscent, with inconspicuous coil-like ornamentation	Dehiscent, with smooth to inconspicuous coil-like ornamentation	Dehiscent, with inconspicuous coil-like ornamentation
Habitat	On wood	On wood	On wood
Location	Ratchaburi	Kanchanaburi, Nakhon Ratchasima, Songkhla, Trad	Burirum
Specimen examined	SUT045	SUT042, SUT059, SUT060, SUT073, SUT080,	SUT020
		SUT094, SUT106, SUT115, SUT116, SUT179,	
		SUT185, SUT189, SUT225, SUT227, SUT232,	
		SUT235, SUT264, SUT265, SUT266, SUT287, and SUT295	

<b>Table 15.</b> (	Continued).
--------------------	-------------

Character	H. cf. perforatum (SUT224)*	H. cf. perforatum (SUT294)*	H. rubiginosum (Pers.: Fr.) Fr., Summa Veg*
Stromata			ž
Shape	Hemispherical, pulvinate to effused- pulvinate	Pulvinate to effused-pulvinate	Effused-pulvinate and sometimes pulvinate or even hemispherical
Color	Brown vinaceous	Reddish brown	Brown vinaceous
Granules beneath surface	Brown vinaceous	Reddish brown	Dark brown
KOH pigments Perithecia	Amber or yellowish brown	Amber or honey	Rust
Shape	Obovoid	Spherical	Obovoid
Size	0.1-0.3 mm diameter x 0.3-0.5 mm high	0.1-0.3 mm diameter	0.2-0.5 mm diameter x 0.3-0.6 mm high
Ostiole	Lower than the stromatal surface, usually overlay with conspicuous white substance	Lower than the stromatal surface	Lower than the stromatal surface
Apical apparatus	Discoid, 0.5 μm high x 1-1.5 μm broad.	Discoid, 0.5-1 $\mu$ m high x 2.5 $\mu$ m broad	Discoid, 0.8-1.5 $\mu$ m high x 2-3 $\mu$ m broad
Ascospores			
Color	Brown to dark brown	Brown to dark brown	Brown to dark brown
Shape	Ellipsoid-inequilateral, with narrowly rounded ends	Ellipsoid-inequilateral to equilateral, with narrowly rounded ends	Ellipsoid-inequilateral, with narrowly rounded ends
Size	8.8-10 x 3.8-5(-6.3) μm	8.8-11.3 x 3.8-5 μm	(7.5-)8.8-10 x 3.8-5 μm
Germ slit	Straight full length	Straight full length	Straight full length
Perispore	Dehiscent, with conspicuous coil-like ornamentation	Dehiscent, with inconspicuous coil-like ornamentation	Dehiscent, with smooth to inconspicuous coil-like ornamentation
Habitat	On wood	On wood	On wood
Location	Trad	Kanchanaburi	Trad
Specimen examined	SUT224	SUT294	SUT215 and SUT221

Character	H. subgilvum Berk. & Broome var. microsporum (Abe) YM. Ju & J.D. Rogers *	H. trugodes (SUT154)*	H. trogodes (SUT187)*
Stromata			
Shape	Effused-pulvinate	Effused-pulvinate	Effused-pulvinate
Color	Hazel or dark brick	Brown vinaceous	Sepia
Granules beneath surface	Yellowish orange	Brown vinaceous	Brownish yellow
KOH pigments	Orange	Amber or yellowish brown	Amber or yellow
Perithecia			
Shape	Obovoid	Obovoid	Obovoid
Size	0.2-0.5 mm diameter x 0.3-0.6 mm high	0.2-0.4 mm diameter x 0.3-1.2 mm high	0.2-0.4 mm diameter x 0.3-1.2 mm high
Ostiole	Lower than the stromatal surface	Lower than the stromatal surface, inconspicuous, sometimes on flattened area	Lower than the stromatal surface
Apical apparatus	Not observed	Discoid, 0.3-0.8 µm high x 1.5-2 µm broad	Discoid, 0.3-0.8 µm high x 1.5-2 µm broad
Ascospores			
Color	Brown to dark brown	Dark brown	Brown to dark brown
Shape	Ellipsoid-inequilateral to equilateral, with narrowly rounded ends	Ellipsoid-inequilateral, with narrowly rounded ends	Ellipsoid-inequilateral, with narrowly rounded ends
Size	(3.8-)5-7.5 x 2.5-3.8 μm	10-12.5 x 3.8-5 μm	10-11.3(-12.5) x 3.8-5(-6.3) μm
Germ slit	Straight to slightly sigmoid full spore- length	Straight full length	Straight full length
Perispore	Dehiscent, with inconspicuous coil-like ornamentation	Dehiscent, with inconspicuous coil-like ornamentation	Dehiscent, with inconspicuous coil-like ornamentation
Location	Songkhla	Nakhon Ratchasima	Nakhon Ratchasima, Trad
Specimen examined	SUT095, SUT104, and SUT108	SUT154	SUT187

# Table 15. (Continued).

Table 13. (Commuted)	Table 15.	(Continued)
----------------------	-----------	-------------

Character	H. kanchanapisekii N. Suwannasai, S.	H. sublenormandii N. Suwannasai, S.	H. suranareei N. Suwannasai, S. Rodtong,
	Rodtong, S. Thienhirun & A.J.S.	Rodtong, S. Thienhirun & A.J.S.	S. Thienhirun & A.J.S. Whalley. sp.
	Whalley. sp. nov. *	Whalley. sp. nov. *	nov. *
Stromata			
Shape	Glomerate to pulvinate	Effused-pulvinate	Glomerate to effused-pulvinate with the tendency to be perithecioid
Color	Dull reddish brown	Dark brick or brown vinaceous	Orange brown
Granules beneath surface	Reddish brown	Brown vinaceous	Orange
KOH pigments	Reddish brown	Amber or yellowish brown	Yellowish orange
Perithecia			
Shape	Spherical	Spherical	Obovoid
Size	0.1-0.2 mm diameter	0.3-0.5 (-0.6) mm diameter	0.2-0.4 mm diameter x 0.3-0.5 mm high
Ostiole	Slightly higher or the same as the stromatal surface	Higher than the stromatal surface	Same or lower than the stromatal surface, with white substance
Apical apparatus	Discoid, 1.25 μm high x 2.5 μm broad	Discoid, 0.7-1.5 µm high x 2-3µm broad	Discoid, 0.7-1.5 µm high x 2-3µm broad
Ascospores			
Color	Brown to dark brown	Brown to dark brown	Brown to dark brown
Shape	Ellipsoid-inequilateral to equilateral, with narrowly rounded ends	Ellipsoid-inequilateral to equilateral, with narrowly rounded ends	Ellipsoid-inequilateral, with narrowly rounded ends
Size	(7.5-)10-11.3(-12.5) x 3.8-5 μm	9-12 x 3.8-5 μm	(10-)12.5-13.8 x 5-6.3 μm
Germ slit	Straight full length	Straight full length	Straight full length
Perispore	Indehiscent, smooth	Dehiscent, with inconspicuous coil-like ornamentation	Dehiscent, with inconspicuous coil-like ornamentation
Location	Ratchaburi	Kanchanaburi, Nakhon Ratchasima, Trad	Nakhon Ratchasima
Specimen examined	SUT066, SUT067, SUT068, and SUT069	SUT250 and SUT282	SUT182, SUT183, and SUT184

# Table 15. (Continued).

Character	Hypoxylon taxonomic species 2 *	Hypoxylon taxonomic species 3 *
Stromata		
Shape	Effused-pulvinate and sometimes pulvinate or even hemispherical	Effused-pulvinate
Color	Brown vinaceous	Dark brick or brown vinaceous
Granule beneath surface	Brown vinaceous	Brown vinaceous
KOH pigments	Yellowish brown	Amber or yellowish brown
Perithecia		
Shape	Obovoid	Obovoid
Size	0.2-0.4 mm diameter x 0.3-0.5 mm high	0.2-0.5 mm diameter x 0.3-0.6 mm high
Ostiole	Lower than the stromatal surface	Lower than the stromatal surface, overlay with white substance
Apical apparatus	Discoid, 0.5-1.5 µm high x 2-3 µm broad	Discoid, 0.8-1.5 µm high x 2-3 µm broad
Ascospores		
Color	Brown to dark brown	Brown to dark brown
Shape	Ellipsoid-inequilateral, with narrowly rounded ends	Ellipsoid-inequilateral, with narrowly rounded ends
Size	(8.8)11.3-12.5(17.5) x 5-7.5 μm	10-11.3 x 3.8-5 μm
Germ slit	Straight full length	Straight full length
Perispore	Dehiscent, smooth	Dehiscent, smooth
Habitat	On wood	On wood
Location	Nakhon Ratchasima	Yasothorn
Specimen examined	SUT082	SUT158



**Figure 22.** *Hypoxylon anthochroum* Berk. & Broome (SUT233); (a) stromatal form (Bar = 0.3 mm), (b) ascospores (Bar = 12  $\mu$ m), (c) KOH-extractable pigment olivaceous, (d) ascospore dehiscent in 10% KOH (arrowed) (Bar = 4  $\mu$ m), (e) straight germ slit spore length (Bar = 2  $\mu$ m), and (f) cultural characteristics on PDA cultured at 25°C after 4 weeks (Bar = 1 cm).



**Figure 23.** *Hypoxylon brevisporum* Y.-M. Ju & J.D. Rogers (SUT256); (a) stromatal form (Bar = 1 cm), (b) KOH-extractable pigment hazel, (c) stromatal with white substance on the ostioles (Bar = 0.2 mm), (d) ascospores (Bar =  $6 \mu m$ ), (e) ascospore dehiscent in 10% KOH (arrowed) (Bar =  $4 \mu m$ ), and (f) straight germ slit spore length (Bar =  $2 \mu m$ ).



**Figure 24.** *Hypoxylon duranii* J.D. Rogers (SUT223); (a) stromatal form (Bar = 1 cm), (b) KOH-extractable pigment yellowish brown, (c) stromatal form (Bar = 0.3 mm), (d) ascospores (Bar =  $10 \mu \text{m}$ ), (e) apical apparatus (Bar =  $2 \mu \text{m}$ ), (f) ascospore dehiscent in 10% KOH (arrowed) (Bar =  $2 \mu \text{m}$ ), and (g) straight germ slit spore length (Bar =  $2 \mu \text{m}$ ).



**Figure 25.** *Hypoxylon fendleri* Berk. *ex* Cooke (SUT162); (a) stromatal form (Bar = 0.3 mm), (b) ascospores (Bar = 10  $\mu$ m), (c) KOH-extractable pigment orange, (d) slightly sigmoid germ slit spore length (arrowed) (Bar = 1  $\mu$ m), (e) ascospore dehiscent in 10% KOH (arrowed) (Bar = 1  $\mu$ m), and (f) cultural characteristics on PDA cultured at 25°C after 4 weeks (Bar = 1 cm).



**Figure 26.** *Hypoxylon* cf. *ferrugineum* (SUT017); (a) and (c) stromatal form (Bars = 0.5 cm and 0.3 mm respectively), (b) KOH-extractable pigment orange, and (d) ascospores (Bar =  $10 \mu \text{m}$ ).



**Figure 27.** *Hypoxylon* cf. *ferrugineum* (SUT070); (a) stromatal form (Bar = 0.2 mm), (b) perithecia (Bar = 0.4  $\mu$ m), (c) KOH-extractable pigment yellowish orange, (d) ascospores (Bar = 10  $\mu$ m), (e) ascospore dehiscent in 10% KOH (arrowed) (Bar = 2  $\mu$ m), and (f) cultural characteristics on PDA cultured at 25°C after 2 weeks (Bar = 1 cm).



**Figure 28.** *Hypoxylon* cf. *ferrugineum* (SUT237); (a) stromatal form (Bar = 1 cm), (b) KOH-extractable pigment brownish yellow, (c) stromatal form (Bar =  $0.4 \mu m$ ), (d) ascospores (Bar =  $10 \mu m$ ), (e) cultural characteristics on PDA cultured at 25°C after 4 weeks (Bar = 1 cm), (f) ascospore dehiscent in 10% KOH (arrowed) (Bar =  $5 \mu m$ ), and (g) straight germ slit spore length (arrowed) (Bar =  $2 \mu m$ ).



**Figure 29.** *Hypoxylon haematostroma* Mont. (SUT164); (a) stromatal form (Bar = 1 cm), (b) KOH-extractable pigment orange, (c) stromatal form (Bar = 0.3 mm), (d) ascospores (Bar =  $10 \ \mu$ m), (e) apical apparatus (arrowed) (Bar =  $3 \ \mu$ m), (f) cultural characteristics on PDA cultured at 25°C after 4 weeks (Bar = 1 cm), and (g) ascospore dehiscent in 10% KOH (arrowed) (Bar =  $2 \ \mu$ m).

*Hypoxylon hypomiltum* SUT166 (Figure 30) examined was similar to *Hypoxylon hypomiltum* Mont. described by Ju and Rogers (1996).

*Hypoxylon investiens* SUT041 and SUT063 (Figure 31) matched *Hypoxylon investiens* (Schwein.) M.A. Curtis described by Ju and Rogers (1996) except for a small difference in ascospore size (7.3-8.8 x 2.5-3.8  $\mu$ m cf. (6-)6.5-9.5(-10) x 3-4.5  $\mu$ m. (Ju and Rogers, 1996)).

*Hypoxylon lenormandii* SUT016, SUT065, SUT144, SUT147, SUT151, SUT180, SUT181, and SUT283 (Figure 32) examined were very similar to *Hypoxylon lenormandii* Berk. & M.A. Curtis *apud* Berk. described by Ju and Rogers (1996) except for slightly smaller ascospores (10-12.5 x 3.8-5 μm cf. 9.5-15(-16) x 4-6.5(-7) μm (Ju and Rogers, 1996)).

*Hypoxylon lenormandii* var. *microspora* SUT022 (Figure 33) examined was similar to the species firstly reported by Thienhirun (1997) except for their ascospores, which were 5-6.3 x 2.5-3.8  $\mu$ m and 3.8-5 x 2.5-3  $\mu$ m respectively. This taxon was different from *H. lenormandii* in ascospore size, germ slit form, and its smooth perispore.

*Hypoxylon macrocarpum* SUT045 (Figure 34) closely fitted *Hypoxylon macrocarpum* Pouzar (Ju and Rogers, 1996) except for slightly differences in ascospore size, 8.8-11.3 x 3.8-5  $\mu$ m cf. 9-12.5(-13) x 4-5.5  $\mu$ m, and type of perithecia, obovoid cf. obovoid to tubular (Ju and Rogers, 1996). Although *H. macrocapum* is similar to *H. rubiginosum* but they differ in stromatal pigments (Ju and Rogers, 1996).


**Figure 30.** *Hypoxylon hypomiltum* Mont. (SUT166); (a) stromatal form (Bar = 1 cm), (b) KOH-extractable pigment amber, (c) stromatal form (Bar = 0.2 mm), (d) ascospore (Bar = 8  $\mu$ m), (e) ascospore dehiscent in 10% KOH (arrowed) (Bar = 2  $\mu$ m), (f) straight germ slit spore length (arrowed) (Bar = 2  $\mu$ m), and (g) cultural characteristics on PDA cultured at 25°C after 4 weeks (Bar = 1 cm).



Figure 31. *Hypoxylon investiens* (Schwein.) M.A. Curtis. (SUT063); (a) stromatal form (Bar = 0.4 mm), (b) Perithecia (Bar = 0.2 mm), (c) KOH-extractable pigment dull green, (d) ascospore dehiscent in 10% KOH (arrowed) (Bar =  $2 \mu m$ ), (e) ascospore (Bar =  $8 \mu m$ ), (f) cultural characteristics on PDA cultured at 25°C after 4 weeks (Bar = 1 cm), and (g) straight germ slit spore length (arrowed) (Bar =  $2 \mu m$ ).



**Figure 32.** *Hypoxylon lenormandii* Berk. & M.A. Curtis. (SUT065); (a) stromatal form (Bar =  $0.5 \ \mu$ m), (b) ascospores (Bar =  $10 \ \mu$ m), (c) ascospore dehiscent in 10% KOH (arrowed) (Bar =  $2 \ \mu$ m), (d) KOH-extractable pigment of red, (e) slightly sigmoid germ slit spore length (arrowed) (Bar =  $2 \ \mu$ m), and (f) cultural characteristics on PDA cultured at 25°C after 4 weeks (Bar = 1 cm).



**Figure 33.** *Hypoxylon lenormandii* var. *microspora* (SUT022) (Thienhirun, 1997); (a) stromatal form (Bar = 0.5  $\mu$ m), (b) perithecia (Bar = 0.5  $\mu$ m), (c) KOH-extractable pigment of red, (d) apical apparatus (arrowed) (Bar = 1  $\mu$ m), (e) ascospore dehiscent in 10% KOH (arrowed) (Bar = 2  $\mu$ m), (f) ascospores (Bar = 5  $\mu$ m), and (g) cultural characteristics on PDA cultured at 25°C after 4 weeks (Bar = 1 cm).



**Figure 34.** *Hypoxylon macrocarpum* Pouzar (SUT045); (a) stromatal form (Bar = 1 cm), (b) KOH-extractable pigment hazel, (c) stromatal form (Bar = 0.2 mm), (d) perithecia (Bar = 0.2 mm), (e) straight germ slit spore length (Bar = 2  $\mu$ m), (f) ascospore dehiscent in 10% KOH (arrowed) (Bar = 2  $\mu$ m), (g) ascospores (Bar = 10  $\mu$ m), and (h) cultural characteristics on PDA cultured at 25°C after 4 weeks (Bar = 1 cm).

## Hypoxylon monticulum SUT042, SUT059, SUT060, SUT073,

SUT080, SUT094, SUT106, SUT115, SUT116, SUT179, SUT185, SUT189, SUT225, SUT227, SUT232, SUT235, SUT264, SUT265, SUT266, SUT287, and SUT295 (Figure 35) closely fitted *Hypoxylon monticulum* Mont. (Ju and Rogers, 1996). The KOH-extractable pigments of the examined samples varied from colorless to purplish in color.

*Hypoxylon* cf. *perforatum* SUT020 (Figure 36), *H*. cf. *perforatum* SUT224 (Figure 37), and *H*. cf. *perforatum* SUT294 (Figure 38) collected from different locations were closed to *H. perforatum* (Schwein.: Fr.) Fr. as reported by Ju and Rogers (1996). All of specimens differed from *H. perforatum* in KOHextractable pigments, ascospore size, (8-)9-12(-13) x 4-6 µm, and germ slit form (slightly sigmoid) as shown in Table 15. *Hypoxylon perforatum* was considered to be a variety of *H. rubiginosum* by Petrini and Müller (1986). However, they were separated from each other by color of stromatal granules, color of KOH-extractable pigments, color of their stromata, and their anamorph (Ju and Rogers, 1996).

*Hypoxylon rubiginosum* SUT215 and SUT221 (Figure 39) examined matched *Hypoxylon rubiginosum* (Pers.: Fr.) Fr. as described by Ju and Rogers (1996).



**Figure 35.** *Hypoxylon monticulosum* Mont. (SUT116); (a), (b), and (c) stromatal form (Bars = 1 cm, 1 cm, and 0.5 mm respectively), (d) ascospore (Bar = 10  $\mu$ m), (e) KOH-extract colorless, (f) slightly sigmoid curve germ slit spore length (Bar = 2  $\mu$ m), (g) cultural characteristics on PDA cultured at 25°C after 4 weeks (Bar = 1 cm), and (h) ascospore dehiscent in 10% KOH (arrowed) (Bar = 2  $\mu$ m).



Figure 36. Hypoxylon cf. perforatum (SUT020); (a) stromatal form (Bar = 0.5 cm),
(b) KOH-extractable pigment amber, (c) stromatal form (Bar = 0.3 mm),
(d) ascospores (Bar = 5 μm), (e) straight germ slit spore length (Bar = 2 μm), (f) ascospore dehiscent in 10% KOH (arrowed) (Bar = 2 μm), and
(g) cultural characteristics on PDA cultured at 25°C after 4 weeks (Bar = 1 cm).



**Figure 37.** *Hypoxylon* cf. *perforatum* (SUT224); (a) stromatal form (Bar = 1 cm), (b) KOH-extractable pigment amber, (c) stromatal form (Bar = 0.4 mm), (d) perithecia (Bar = 0.2 mm), (e) ascospores (Bar = 10  $\mu$ m), (f) straight germ slit spore length (Bar = 2  $\mu$ m), and (g) ascospore dehiscent in 10% KOH (arrowed) (Bar = 5  $\mu$ m).



**Figure 38.** *Hypoxylon* cf. *perforatum* (SUT294); (a) stromatal form (Bar = 1 cm), (b) KOH-extractable pigment amber, (c) stromatal form (Bar = 0.3 mm), (d) ascospores (Bar = 10  $\mu$ m), (e) straight germ slit spore length (Bar = 2  $\mu$ m), (f) ascospore dehiscent in 10% KOH (arrowed) (Bar = 2  $\mu$ m), and (g) cultural characteristics on PDA cultured at 25°C after 4 weeks (Bar = 1 cm).



**Figure 39.** *Hypoxylon rubiginosum* (SUT215); (a) stromatal form (Bar = 1 cm), (b) KOH-extractable pigment rust, (c) stromatal form (Bar = 0.2 mm), (d) ascospores (Bar = 10  $\mu$ m), (e) ascospore dehiscent in 10% KOH (arrowed) (Bar = 2  $\mu$ m), and (f) cultural characteristics on PDA cultured at 25°C after 4 weeks (Bar = 1 cm).

*Hypoxylon subgilvum* var. *microsporum* SUT095, SUT104, and SUT108 (Figure 40) examined fitted *Hypoxylon subgilvum* Berk. & Broome var. *microsporum* (Abe) Y.-M. Ju & J.D. Rogers as described by Ju and Rogers (1996).

*Hypoxylon trogodes* SUT187 and SUT154 (Figures 41 and 42) fitted *Hypoxylon trogodes* Berk. & Broome as described by Ju and Rogers (1996). *Hypoxylon trogodes* SUT187 and SUT154 differed slightly from *H. trogodes* in ascospore size.

*Hypoxylon kanchanapisekii* N. Suwannasai, S. Rodtong, S. Thienhirun & A.J.S. Whalley, sp. nov. (Figure 43). Characteristics of this taxon are as follows: stromata glomerate to pulvinate, restricted and usually containing less than 20 perithecia, perithecia occasionally almost free, 0.5-2 mm x 0.1-0.2 mm thick, with perithecial mounds inconspicuous to 1/3 exposed, surface dull reddish brown with KOH extractable pigments brown vinaceous (84), umber (9); perithecia spherical 0.1-0.2 mm diameter, ostioles slightly higher or the same as the stromatal surface; asci 105-120  $\mu$ m total length x 3.8-5  $\mu$ m broad, the spore bearing parts 75-85  $\mu$ m long with stipes 12.5-45  $\mu$ m; ascospores brown, unicellular, equilateral, with narrowly rounded ends, 10-11.25(-12.5) x (0.5-)3.75-5  $\mu$ m, with straight-germ slit less than to nearly spore length; perispore indehiscent in 10% KOH, smooth, epispore smooth.

Specimens examined: Thailand, Plant Nursery of the Royal Forest Department, Ratchaburi Province, the branch of the Royal Forest Department, on bamboo, 28 August 2003, Suwannasai, N. (Holotype SUT069); SUT066; SUT068.

Colonies on PDA covering 9-cm Petri dish in two weeks at room temperature, 23-28°C, at first creamy white then buff, velvety to felty, with concentric zones where aerial hyphal tufts develop. Anamorph not formed. This taxon was close to *H. lenormandii* but it differed in stromatal surface color of dull reddish brown not grayish sepia, in small ascospores  $(7.5-)10-11.3(-12.5) \ge 3.8-5 \ \mu m cf. 9.5-15(-16) \ge 4-6.5(-7) \ \mu m$ , Ju and Rogers (1996), and in having a straight rather than slightly sigmoid germ slit. This taxon was only found on bamboo.

*Hypoxylon sublenormandii* N. Suwannasai, S. Rodtong, S. Thienhirun & A.J.S. Whalley, sp. nov. (Figure 44). Characteristics of this taxon are as follows: stromata glomerate to effused-pulvinate, often appearing almost rosellinioid but joined by thin stromal tissue, conspicuous perithecial mounds, surface reddish brown; reddish brown granules immediately beneath surface and between perithecia, with KOH-extractable pigments brown vinaceous (84), umber (9); perithecia spherical, 0.2-0.4 mm diameter, ostioles slightly higher than the stromatal surface; asci 95-110  $\mu$ m total length x 3.8-5  $\mu$ m broad, the spore bearing parts 65-75  $\mu$ m long with stipes 30-42.5  $\mu$ m; ascospores brown, unicellular, equilateral, with narrowly rounded ends, 9-12 x 4-5  $\mu$ m, with straight-germ slit spore length; perispore dehiscent in 10% KOH, with inconspicuous coil-like ornamentation; epispore smooth.

Specimens examined: Thailand, Kanchanaburi Province, Chong Kho Neab Forest, on bamboo, 14 December 2003, Suwannasai, N. (Holotype SUT282); Trad Province, Ta Gum Forest, on bamboo, 19 September 2003, Phosri, C. SUT250.

Colonies on PDA covering 9-cm Petri dish in two weeks at room temperature, 23-28°C, at first creamy white then brown, felty, azonate, with diffuse margins. Anamorph not formed. This species was similar to *H. lenormandii* and differed mainly in its ascospore size 8-10 x 3.8-5  $\mu$ m cf. 9.5-15(-16) x 4-6.5(-7)  $\mu$ m, Ju and Rogers (1996) and in its straight germ slit of spore length.

*Hypoxylon suranareei* N. Suwannasai, S. Rodtong, S. Thienhirun & A.J.S. Whalley, sp. nov. (Figure 45). Characteristics of this taxon are as follows: stromata glomerate to effused-pulvinate, often appearing almost rosellinioid but joined by thin stromal tissue, conspicuous perithecial mounds, surface orange brown; orange granules immediately beneath surface and between perithecia, with KOH-extractable pigments yellowish orange; perithecia obovoid, 0.2-0.4 mm diameter, ostioles same or lower than the stromatal surface, with white substance; asci 90-120  $\mu$ m total length x 3.8-5  $\mu$ m broad, the spore bearing parts 70-85  $\mu$ m long with stipes 30-50  $\mu$ m; ascospores brown to dark brown, unicellular, equilateral, with narrowly rounded ends (10-)12.5-13.8 x 5-6.3  $\mu$ m, with straight-germ slit spore length; perispore dehiscent in 10% KOH, with inconspicuous coil-like ornamentation; epispore smooth.

Specimens examined: Thailand, Suranaree University of Technology, Nakhon Ratchasima, on wood, 17 November 2003, Suwannasai, N. (Holotype SUT182), SUT183, and SUT184.

Colonies on PDA covering 9-cm Petri dish in two weeks at room temperature, 25°C, at first creamy white then brown, felty, azonate, with diffuse margins. Anamorph not formed.

This taxon was similar to *H. lenormandii* Berk. & M.A. Curtis *apund* Berk. in stromatal form but it was different in stromatal surface color, ascospore size  $(9.5-15(-16) \times 4-6.5(-7) \mu m)$ , germ slit form, and KOH pigment.



**Figure 40.** *Hypoxylon subgilvum* Berk. & Broome var. *microsporum* (Abe) Y.-M. Ju & J.D. Rogers. (SUT234); (a) stromatal form (Bar = 1 cm), (b) KOH-extractable pigment orange, (c) stromatal form (Bar = 0.5 mm), (d) perithecia (Bar =  $0.5 \mu m$ ), (e) ascospores (Bar =  $7 \mu m$ ), (f) straight to slightly sigmoid germ slit spore length (arrowed) (Bar =  $2 \mu m$ ), (g) apical apparatus (arrowed) (Bar =  $1 \mu m$ ), (h) ascospore dehiscent in 10% KOH (arrowed) (Bar =  $1 \mu m$ ), and (i) cultural characteristics on PDA cultured at  $25^{\circ}C$  after 4 weeks (Bar = 1 cm).



**Figure 41.** *Hypoxylon trogodes* Berk. & Broome (SUT187); (a) stromatal form (Bar = 1 cm), (b) perithecia (Bar = 0.2  $\mu$ m), (c) KOH-extractable pigment yellow, (d) straight germ slit spore length (arrowed) (Bar = 2  $\mu$ m), (e) ascospore dehiscent in 10% KOH (arrowed) (Bar = 2  $\mu$ m), (f) ascospores (Bar = 10  $\mu$ m), and (g) cultural characteristics on PDA cultured at 25°C after 4 weeks (Bar = 1 cm).



**Figure 42.** *Hypoxylon trugodes* Berk. & Broome (SUT154); (a) stromatal form (Bar = 1 cm), (b) perithecia (Bar = 0.4  $\mu$ m), (c) KOH-extractable pigment yellowish brown, (d) ascospore dehiscent in 10% KOH (arrowed) (Bar = 2  $\mu$ m), (e) ascospores (Bar = 10  $\mu$ m), and (f) cultural characteristics on PDA cultured at 25°C after 4 weeks (Bar = 1 cm).



**Figure 43.** *Hypoxylon kanchanapisekii* N. Suwannasai, S. Rodtong, S. Thienhirun & A.J.S. Whalley, sp. nov. (SUT069); (a) stromatal form (Bar = 1 cm), (b) KOH-extractable pigment reddish brown, (c) stromatal form (Bar = 0.2 mm), (d) perithecia (Bar =  $0.2 \ \mu m$ ), (e) ascospores (Bar =  $10 \ \mu m$ ), (f) apical apparatus (arrowed) (Bar =  $1 \ \mu m$ ), (g) straight germ slit less than spore length (arrowed) (Bar =  $2 \ \mu m$ ), and (h) cultural characteristics on PDA cultured at 25°C after 4 weeks (Bar =  $1 \ cm$ ).



**Figure 44.** *Hypoxylon sublenormandii* N. Suwannasai, S. Rodtong, S. Thienhirun & A.J.S. Whalley, sp. nov. (SUT282); (a) stromatal form (Bar = 0.5 cm), (b) ascospores (Bar = 10  $\mu$ m), (c) KOH-extractable pigment of reddish brown, (d) apical apparatus (arrowed) (Bar = 1  $\mu$ m), (e) straight germ slit spore length (arrowed) (Bar = 2  $\mu$ m), (f) ascospore dehiscent in 10% KOH (arrowed) (Bar = 2  $\mu$ m), (g) cultural characteristics on PDA cultured at 25°C after 4 weeks (Bar = 1 cm), and (h) SEM micrograph of coil-like ornamentation of perispore (Bar = 1  $\mu$ m).



Figure 45. *Hypoxylon suranareei* N. Suwannasai, S. Rodtong, S. Thienhirun & A.J.S. Whalley, sp. nov. (SUT182); (a) stromatal form (Bar = 1 cm), (b) KOH-extractable pigment yellowish orange, (c) stromatal form (Bar = 0.5 μm), (d) straight germ slit spore length (arrowed) (Bar = 5 μm), (e) ascospore dehiscent in 10% KOH (arrowed) (Bar = 5 μm), (f) ascospores (Bar = 10 μm), and (g) cultural characteristics on PDA cultured at 25°C after 2 weeks (Bar = 1 cm).

*Hypoxylon* taxonomic species 2 (SUT082) (Figure 46) was close to *Hypoxylon rubiginosum* (Pers.: Fr.) Fr. as described by Ju and Rogers (1996) except that KOH-extractable pigment colour was yellowish brown and the range of ascospore size was broader ((8.8-)11.3-12.5(-17.5) x 5-7.5  $\mu$ m) than *Hypoxylon rubiginosum* (Pers.: Fr.) Fr. ((8-)9-12 x 4-4.5  $\mu$ m). In addition, perispore of this taxon was inconspicuous coil-like ornamentation whereas *H. rubiginosum* was smooth (Ju and Rogers, 1996).

*Hypoxylon* taxonomic species 3 (SUT158) (Figure 47) was close to *Hypoxylon rubiginosum* (Pers.: Fr.) Fr. as described by Ju and Rogers (1996) except for KOH-extractable pigment colour. *Hypoxylon* taxonomic species 3 had the same colour of KOH-extractable pigment as *Hypoxylon* taxonomic species 2 but ascospore size of *Hypoxylon* taxonomic species 2 was slightly smaller than *Hypoxylon* taxonomic species 3, (8.8-)11.3-12.5(-17.5) x 5-7.5  $\mu$ m and 10-11.3 x 3.8-5  $\mu$ m respectively.



**Figure 46.** *Hypoxylon* taxonomic species 2 (SUT082); (a) stromatal form (Bar = 1 cm), (b) KOH-extractable pigment rust, (c) stromatal form (Bar = 0.2 mm), (d) ascospores (Bar = 10  $\mu$ m), (e) ascospore dehiscent in 10% KOH (arrowed) (Bar = 2  $\mu$ m), and (f) cultural characteristics on PDA cultured at 25°C after 4 weeks (Bar = 1 cm).



**Figure 47.** *Hypoxylon* taxonomic species 3 (SUT158); (a) stromatal form (Bar = 0.5 cm), (b) ascospores (Bar = 10  $\mu$ m), (c) straight germ slit spore length (arrowed) (Bar = 2  $\mu$ m), (d) KOH-extractable pigment amber, (e) ascospore dehiscent in 10% KOH (arrowed) (Bar = 2  $\mu$ m), and (f) cultural characteristics on PDA cultured at 25°C after 4 weeks (Bar = 1 cm).

## 4.2.5 Group V: Xylariaceous endophytes

Although eight xylariaceous genera have been reported as endophytes, Anthostomella, Biscogniauxia, Daldinia, Hypoxylon, Kretzschmaria, Nemania, Rosellinia, and Xylaria, the common endophytic genus is Xylaria. In this study, twenty two species of Xylaria were recorded (Table 16). Other xylariaceous genera, Kretzschmaria, Nemania, and Biscogniauxia, were also included (Table 16).

 Table 16.
 Species of Xylaria, Kretzschmaria, Nemania, and Biscogniauxia found in this study.

Species	No.*	Remark
Xylaria		
X. anisopleura (Mont.) Fr.	3	According to Rogers and Samuels (1986), Rogers (1988), González and Rogers (1989), and Thienhirun (1997)
X. badia Pat.	15	According to Van der Gucht (1995), and Thienhirun (1997)
X. beccari Lloyd	1	Lloyd (1924)
X. brachiata Sacc.	1	Lloyd (1919)
X. cubensis (Mont.) Fr.	6	According to Rogers and Samuels (1986), Rogers <i>et al.</i> (1988), González and Rogers (1989), and Thienhirun (1997)
X. ianthino-velutina (Mont.) Fr.	2	According to Dennis (1957), González and Rogers (1989), and Thienhirun (1997)
X. cf. juruensis (SUT035)	1	Ascospore size overlaps between <i>X. juruensis</i> and <i>X multiplex</i> .
X. juruensis var. microspora	8	Thienhirun, 1997
X. maitlandii (Dennis) D. Hawksw.	2	According to González and Rogers (1989)
X. mellisii (Berk.) Cooke	2	Van der Gucht (1995)
X. multiplex (Kunze) Fr.	1	Dennis (1957; 1961), González and Rogers (1989), and Thienhirun (1997)

\* Number of collections

Table 16.(Continued).

Species	No.*	Remark
Xylaria		
X. muscula Lloyd	2	Dennis (1957)
X. psidii J.D. Rogers & Hemmes	5	According to Rogers, Ju and
		Hemmes (1992), and Thienhirun
		(1997)
X. schweinitzii Berk. & M.A. Curtis	1	According to Rogers et al. (1988),
		González and Rogers (1989), and
		Thienhirun (1997)
X. scruposa (Fr.) Fr.	1	Van der Gucht (1995)
X. telfairii (Berk.) Fr.	1	According to Dennis (1961),
		Rogers et al. (1987 and 1988),
		Callan and Rogers (1990), and
		Thienhirun (1997)
<i>Xylaria</i> species 2	17	Thienhirun, 1997
<i>Xylaria</i> sp. nov.	27	Rough stromatal surface, finely
		reticulately cracked into small
		angular closely spaced scales so as
		to outline the individual perithecia
<i>Xylaria</i> taxonomic species 1 (SUT075)	1	Smooth stromatal surface except for
	_	peeling layer, externally blackish
		with dark brown outer peeling layer,
		internally creamy white
<i>Xylaria</i> taxonomic species 2 (SUT203)	1	Smooth stromatal surface except for
		peeling layer, externally blackish
		brown to black with dull black
		peeling of outer later, internally
Vulnuis tononomia anagias 2 (SUT204)	1	white
<i>Xytarta</i> taxonomic species 5 (SU1204)	1	stromatal surface by warts external
		black and internally white
<i>Xylaria</i> taxonomic species 4 (SUT207)	1	Smooth stromatal surface except for
	1	ostiolar slightly raised, externally
		copper- to bronze-colored to brown
		with black of ostioles, internally
		creamy white
Kretzschmaria		
Kretzschmaria species (SUT101)	2	Stromata clustered subglobose fertile
		head with short stalk, not branched,
		externally blackish, internally white
Nemania		
Nemania species (SUT258)	2	Erumpent to superficial stromata.
• • • •		smooth with slightly papillate surfac

\* Number of collections

Table 16.(Continued).

Species	No.*	Remark
Biscogniauxia		
Biscogniauxia capnodes (Berk.) YM.	4	Ju and Rogers (1998)
Ju & J.D. Rogers		
Biscogniauxia sp. nov. (SUT290)	2	Applanate stromata, smooth
		surface, externally black and
		internally yellow which
		distinguished from the other known
		species

\* Number of collections

The main characteristics for each species examined are described in Table 17. *Xylaria anisopleura* SUT196, SUT205, and SUT208 (Figure 48) matched *Xylaria anisopleura* (Mont.) Fr. as described by Rogers and Samuels (1986), Rogers (1988), and González and Rogers (1989), but the collections from this study were different from the specimens described by Thienhirun (1997) in ascospore size, (20-)23.8-25(-27.5)  $\mu$ m cf. 17.5-22.5 x 10-11.3  $\mu$ m (Thienhirun, 1997). In addition, the specimens examined had two different stromatal types. The stromata of SUT208 and SUT209 were boarder than high but SUT196 was higher than board. However, both of them had the same cultural characteristics on PDA. The species that is related to *X. anisopleura* is *X. polymorpha* (Pers.: Fr.) Grev., but they are different in the smaller size and moriform shape of stromata and the short, oblique to somewhat spiraling ascospore germ slit (Rogers and Samuels, 1986).

*Xylaria badia* SUT026, SUT032, SUT076, SUT142, SUT309, and SUT310 (Figure 49) were close to *Xylaria badia* Pat. as described by Van der Gucht (1995), and Thienhirun (1997). This taxon was specific to bamboo, and was widely distributed throughout Thailand.

Character	X. anisopleura (Mont.) Fr.*	X. badia Pat.*	X. beccari Lloyd. *
Stromata			
Shape	Clavate to cylindrical, or subglobose,	Short cylindrical to clavate, with rounded	Cylindrical to clavate from short
	with rounded fertile apices	fertile apices	concolorous stipe
Color	Externally dark brown to dull black,	Externally silvery brown and became to	Externally brownish black, internally white
	internally white	grayish brown with age, internally brownish	
TT (	XX7 1	orange	XX7 1
Texture	Woody	Hard	Woody
Surface	Cracked into minute angular scales,	Smooth and shinning	Roughened due to little cracks and small
	rough due to wrinkles, warts and		excrescences
D 11 1	tomentum		
Perithecia			
Shape	Partially immersed, subglobose	Completely immersed, subglobose	Subglobose
Size	0.5-1 mm diameter	0.2-0.3 mm diameter	0.2-0.3 mm diameter
Ostiole	Inconspicuous, papillate, appearing as small hemispherical black discs	Finely papillate and black	Slightly papillate
Apical apparatus	Rectangular, constricted sub-apically, 7-	Discoid, 0.6 µm high x 1.2	Rectangular, 1-1.5 µm high x 1-1.5µm broad
	8(-9) μm high x 4-5 μm broad	μm broad	
Ascospores			
Color	Brown to dark brown	Light brown	Brown
Shape	Ellipsoid-inequilateral to crescentic, with	Ellipsoid, with narrowly rounded ends	Ellipsoid-inequilateral with broadly rounded
	narrowly rounded apiculate ends		ends
Size	(20-)23.8-25(-27.5) x 7.5-8.8µm	7.5-8.8 x 3.8-4.4 μm	(5-)6.3-7.5 x 2.5-3 μm
Germ slit	Straight to curving, oriented obliquely to	Straight less than spore length	Straight full length
	the long axis of the spore		
Habitat	On wood	On wood	On wood
Location	Trad	Kanchanaburi, Nakhon Ratchasima,	Songkhla
		Ratchaburi	
Specimen examined	SUT196, SUT205, and SUT208	SUT026, SUT032, SUT076, SUT142,	SUT092
		SUT309, and SUT310	

**Table 17.** Morphological characteristics of *Xylaria*, *Kretzschmaria*, *Nemania*, and *Biscogniauxia* found in this study.

 Table 17. (Continued).

Character	X. brachiata Sacc.*	X. cubensis (Mont.) Fr.*	X. ianthino-velutina (Mont.) Fr.*
Stromata			
Shape	Upright or prostrate, the fertile part short cylindrical to fusoid with perithecia immersed or with evident perithecial contours, with acute sterile apices	Cylindric-allantoid to clavate, occasionally flattened, with rounded fertile apices, short stipes, arising from tomentose discoid bases	Cylindrical, long conical, or flattened, the fertile parts bearing more or less naked perithecia, grading into ill-defined stipes
Color	Externally brown outer peeling layer, internally white	Externally bronze, becoming dark with age, internally white to cream	Externally blackish, internally white
Texture	Fairly hard	Hard	Soft
Surface	Smooth to roughened with perithecial contours	Smooth, sometimes very faintly, reticulately cracked around the ostioles, or surface conspicuously cracked into small polygonal surface scales	Roughened with perithecia and tomentose except the stromatal apices
Perithecia			
Shape	Subglobose	Completely immersed, subglobose	Subglobose
Size	0.3-0.5 mm diameter	0.3-0.8 mm diameter	0.2-0.3 mm diameter
Ostiole	Inconspicuous to papillate	Finely papillate to annulate	Minutely papillate
Apical apparatus	Rectangular, 3.5-4 µm high x 1.5-2.5 µm broad	Cubic to rectangular, 1.6-2.4 µm high x 1.4- 1.8(-2) µm broad	Cubic to rectangular, 1.3 µm high x 1.3 µm broad
Ascospores			
Color	Brown to dark brown	Light brown	Brown
Shape	Ellipsoid-inequilateral, with broadly to narrowly rounded ends	Ellipsoid-inequilateral, with rounded ends	Ellipsoid-inequilateral with rounded ends
Size	(8-)10-11.3(-12.5) x 3.8-5 μm	(6.3-)7.5-8.8 x 3.8-5 μm	(7.5-)8.8-10(-12.5) x 3.8-4 μm
Germ slit	Straight full length	Straight less than spore length	Straight less than spore length
Habitat	On wood	On wood	On legume pod
Location	Nakhon Ratchasima	Ratchaburi, Trad	Songkhla
Specimen examined	SUT078 and SUT175	SUT089, SUT090, SUT193, SUT194, and SUT199	SUT091 and SUT123

 Table 17. (Continued).

Character	X. cf. juruensis (SUT035)*	X. cf. iuruensis (SUT088)*	X. cf. juruensis (SUT140)*
Stromata			
Shape	Short cylindrical to irregular with acute sterile apices (1 mm long), with short hair	Cylindrical to irregular with acute sterile apices, with thin stalk and no hair	Short cylindrical to irregular with acute sterile apices, short stalk with short hair
Color	Externally blackish with brownish gray peeling layer, internally white	Externally blackish with brownish gray peeling layer, internally white	Externally blackish with brownish gray peeling layer, internally white
Texture	Fairly hard	Fairly hard	Fairly hard
Surface	Roughened with perithecial contours	Roughened with perithecial contours	Roughened with perithecial contours
Perithecia			
Shape	Subglobose	Subglobose	Subglobose
Size	0.2-0.6 mm diameter	0.2-0.4 mm diameter	0.2-0.5 mm diameter
Ostiole	Umbilicate to slightly raise	Umbilicate to slightly raise	Umbilicate to slightly raise
Apical apparatus	Rectangular, 5-7 μm high x 2-4 μm broad	Rectangular, 4.5-7 μm high x 2-3.8 μm broad	Rectangular, 5-7 µm high x 2-3.8 µm broad
Ascospores			
Color	Brown	Brown	Brown
Shape	Ellipsoid-inequilateral, with rounded ends	Ellipsoid-inequilateral, with rounded ends	Ellipsoid-inequilateral, with rounded ends
Size	(10-)11.3-13.8 x 3.8-5 μm	12.5-15 x 3.8-5 μm	12.5-14(-15) x 3.8-5 μm
Germ slit	Straight slightly spore length	Straight slightly spore length	Straight slightly spore length
Habitat	On wood	On wood	On wood
Location	Ratchaburi	Songkhla	Nakhon Ratchasima
Specimen examined	SUT035	SUT088	SUT140

 Table 17. (Continued).

Character	X. cf. juruensis (170)*	X. juruensis var. microspora *	X. maitlandii (Dennis) D. Hawksw. *
Stromata			
Shape	Long cylindrical to irregular with acute sterile apices, with thin stalk and no hair	Cylindrical to irregular, terete to compressed, with hair-like apiculi on stipe	Cylindrical to gregarious, with acute sterile apices (1 mm diam), smooth stipe
Color	Externally blackish with brownish gray peeling layer, internally white	Externally blackish with brown peeling outer layer, internally white	Externally blackish with dark brown peeling outer layer, internally white
Texture	Fairly hard	Fairly hard	Fairly hard
Surface	Roughened with perithecial contours	Roughened by peeling layer	Smooth except for peeling layer
Perithecia			
Shape	Subglobose	Subglobose	Subglobose
Size	0.3-0.5 mm diameter	0.4-0.6 mm diameter	0.3-0.5 mm diameter
Ostiole	Umbilicate to slightly raise	Slightly raised	Slightly raised
Apical apparatus	Rectangular, 5-7 μm high x 2-4 μm broad	Rectangular, 1.4-1.8 μm high x 1.4-1.5 μm broad	Quadrate, 2 $\mu$ m high x 2 $\mu$ m broad
Ascospores			
Color	Brown	Brown	Brown to dark brown
Shape	Ellipsoid-inequilateral, with rounded ends	Ellipsoid-inequilateral with rounded to acute ends	Ellipsoid-inequilateral with broadly rounded ends
Size	11.3-12.5 x 3.8-5 μm	8.8-10 x 3.8-5 μm	11.3-12.5 x 3.8-5 μm
Germ slit	Straight slightly spore length	Straight less than spore length	Straight less than spore length
Habitat	On wood	On wood	On wood
Location	Nakhon Ratchasima	Nakhon Ratchasima	Nakhon Ratchasima
Specimen examined	SUT170	SUT129, SUT138, and SUT139	SUT177

 Table 17. (Continued).

Character	X. mellisii (Berk.) Cooke *	X. multiplex (Kunze) Fr. *	X. muscula Lloyd. *
Stromata			
Shape	Fertile part cylindrical to cylindric- conical, with acute sterile apices, on narrow hirsute stipes	Cylindrical, with acute sterile apices, smooth stipe, arising from enlarged tomentose base	Cylindrical with sterile apices, on short stipes
Color	Externally blackish with grey to brown outer peeling layer, internally white to creamy white	Externally blackish with light brown outer peeling layer, which splited longitudinally into narrow strips, internally white	Externally white with black ostioles, internally creamy white
Texture	Fairly hard	Fairly soft	Woody
Surface	Smooth except for peeling layer and ostiolar discs	Nodulose due to slightly protruding perithecial contours, and smooth	Slightly roughened by ostioles
Perithecia			
Shape	Immersed, subglobose	Partially immersed, subglobose	Subglobose
Size	0.3-0.4 mm diameter	0.5 mm diameter	0.2-0.4 mm diameter
Ostiole	Inconspicuous to finely papillate	Papillate	Umbilicate to slightly raised
Apical apparatus	Rectangular, 3-3.5 µm high x 2-2.5 µm broad	Rectangular, 1.5-2 $\mu$ m high x 1.5 $\mu$ m broad	Quadrate, 1-1.5 µm high x 1-1.5µm broad
Ascospores			
Color	Brown to dark brown	Brown	Light brown
Shape	Ellipsoid-inequilateral with narrowly rounded ends	Ellipsoid-inequilateral, with narrowly rounded ends	Ellipsoid-inequilateral with broadly rounded ends
Size	12.5-15 x 3.8-5 μm	11.3-13.8(-15) x 3.8-5 μm	6-9(-10) x 3-3.5(-4) μm
Germ slit	Straight less than spore length	Straight full length	Straight full length
Habitat	On wood	On wood	On wood
Location	Ratchaburi, Trad	Ratchaburi	Nakhon Ratchasima
Specimen examined	SUT074 and SUT192	SUT028	SUT029
* More details on collect	ctions are given in Appendix B.		

Character	X. psidii J.D. Rogers & Hemmes*	X. schweinitzii Berk. & M.A. Curtis*	X. scruposa (Fr.) Fr.*
Stromata			
Shape	Upright, cylindrical to more or less conical with acute sterile apices, bearing embedded to prominent perithecia	Cylindrical to clavate to irregular, with rounded fertile apices, with long or short stipes or sessile	Cylindrical to clavate to highly irregular, subglobose, on short or long stipes, with rounded or flattened fertile apices, on short to long narrowed smooth stipes
Color	Externally blackish, internally white	Externally brownish black to dull black, internally white	Externally yellowish brown to dark brown, internally white to creamy white
Texture	Fairly soft	Fairly hard	Woody to fairly hard
Surface	Roughened with perithecia	Cracked into minute scales, and rugulose	Rugose and usually roughened by warts
Perithecia			
Shape	Subglobose	Partially immersed, subglobose	Immersed, subglobose
Size	0.2-0.3 mm diameter	0.4-0.6 mm diameter	0.3-0.5 mm diameter
Ostiole	Umbilicate	Inconspicuous, umbilicate, appearing as small hemispherical black disks in between the dark brown scales	Inconspicuous, umbilicate, appearing as hemispherical black discs in between the brown scales
Apical apparatus	Cubic, 2 $\mu$ m high x 2 $\mu$ m broad	Rectangular, 4.5-5 $\mu$ m high x 3.8-5 $\mu$ m broad	Rectangular, constricted subapically, 4-5 µm high x 3-3.5 µm broad
Ascospores			
Color	Brown	Brown to dark brown	Light brown
Shape	Ellipsoid-inequilateral to somewhat fusoid, with rounded to acute ends	Ellipsoid-inequilateral with narrowly rounded ends	Ellipsoid-inequilateral with narrowly rounded to pinched ends
Size	(7.5-)8.8-10(-12.5) x 3.8-4 μm	18.8-21.3 x 6.3-7.5 μm	17.5-21.3(-22.5) x (5-)6.3-7.5 μm
Germ slit	Straight full length	Straight to slightly spiralling, obliquely oriented to the long axis of the spore, less than spore length	Straight to slightly sigmoid, slightly obliquely oriented to the long axis of the spore, less than spore length
Habitat	On Pod	On wood	On wood
Location	Songkhla	Trad	Petchaboon
Specimen examined	SUT124, SUT125, and SUT126	SUT201	SUT117

 Table 17. (Continued).

Character	<i>Xylaria</i> species 2 *	X. telfairii (Berk.) Fr.*	<i>Xylaria</i> sp. nov.*
Stromata		· · · · · · · · · · · · · · · · · · ·	
Shape	Cylindrical, with rounded fertile apices, smooth stipe	Cylindrical to fusiform, not branched or occasionally branched near the base, with rounded fertile apices, smooth stipes, concolorous to the fertile part	Cylindrical, gregarious, with narrowly rounded fertile apices, smooth stipe, which was longitudinally furrowed or wrinkled
Color	Externally blackish with dark brown sloughing scales, internally yellow	Externally pale yellow, clay-colored to orange brown, internally white	Externally dark brown to black, internally white
Texture	Woody	Hard to very hard	Woody
Surface	Rough, cracked into rounded or angular dark brownish scales	Cracked into minute scales	Rough, finely reticulately cracked into small angular closely spaced scales so as to outline the individual perithecia
Perithecia			L
Shape	Immersed, subglobose	Partially immersed, subglobose	Immersed, sometimes vaguely evident in outline, subglobose
Size	0.3- 0.5 mm diameter	0.5-0.7 mm diameter	0.3- 0.5 mm diameter
Ostiole	Slightly raised	Inconspicuous, minute, black, and punctiform	Slightly papillate
Apical apparatus	Rectangular, 1.5-2 µm high x 1.5 µm broad	Rectangular, constricted subapically, 4.5- 5 um high x 3.8-5 um broad	Quadrate to rectangular, 1.5 µm high x 1.5-2 µm broad
Ascospores			
Color	Brown	Dark brown	Light brown to brown to dark brown
Shape	Ellipsoid-equilateral with narrowly rounded ends	Ellipsoid-inequilateral with narrowly rounded to pinched ends	Ellipsoid-equilateral with narrowly rounded ends
Size	(7.5-)10-12.5 x 3.75-5 μm	17.5-20 x 5-6.25 μm	(7.5-)8.8-10 x (2.5-)3.8-5 μm
Germ slit	Slightly sigmoid full spore length	Straight to slightly sigmoid, obliquely oriented to the long axis of the spore, less than spore length	Straight full length
Habitat	On wood	On wood	On wood
Location	Kanchanaburi, Nakhon Ratchasima, Trad	Trad	Chiang Rai, Kanchanaburi, Nakhon Ratchasima, Trad
Specimen examined	SUT127, SUT128, SUT130, SUT132,	SUT206	SUT006, SUT012, SUT027, SUT031, SUT033,
	SUT134, SUT171, SUT195, SUT271,		SUT034, SUT087, SUT093, SUT131, SUT133.
	and SUT274		SUT136, SUT141, SUT143, SUT155, SUT172,
			SUT197, SUT198, SUT272, SUT273, and SUT275

 Table 17. (Continued).

Character	Xylaria taxonomic species 1 *	Xylaria taxonomic species 2 *	Xylaria taxonomic species 3 *
Stromata			
Shape	Cylindrical, with rounded fertile apices, short hair on stromata and stalk	Cylindrical, bearing completely immersed perithecia, with attenuated or acute sterile apices	Prostrate or upright, each stroma consisting of a rachis bearing scattered to crowded, naked perithecia, extended upward into short acute apices
Color	Externally blackish with dark brown outer peeling layer, internally creamy white	Externally blackish brown to black with dull black peeling of outer later, internally white	Externally black, internally white
Texture	Fairly soft	Woody	Fairly soft
Surface	Smooth except for peeling layer	Smooth except for peeling outer layer	Rugose and usually roughened by warts
Perithecia			
Shape	Subglobose	Immersed, subglobose	Subglobose
Size	0.3- 0.5 mm diameter	0.4-0.8 mm diameter	0.4-0.8 mm diameter
Ostiole	Slightly papillate	Umbilicate to slightly raised	Papillate
Apical apparatus	Rectangular, 1-1.5 µm high x 1-1.5µm broad	Not observed	Rectangular, 1-1.5 µm high x 1-1.5µm broad
Ascospores			
Color	Brown	Brown to dark brown	Brown
Shape	Ellipsoid-inequilateral with broadly rounded ends	Ellipsoid-inequilateral with narrowly rounded to pinched ends	Ellipsoid-inequilateral with broadly rounded to pinched ends
Size	12.5-15(-16) x 5-6.3 μm	(7.5-)8.8-10 x 3.8 μm	6.25-7.5 x 2.5-3.8 μm
Germ slit	Straight full length	Straight full length	Straight less than spore length
Habitat	On wood	On wood	On wood
Location	Ratchaburi	Trad	Trad
Specimen examined	SUT075	SUT203	SUT204
* More details on collect	ctions are given in Appendix B.		

	Table 17.	(Continued	).
--	-----------	------------	----

Character	Xylaria taxonomic species4 *	Biscogniauxia capnodes (Berk.) YM. Ju & J.D. Rogers*	Biscogniauxia sp. nov. (SUT290)*
Stromata			
Shape	Clavate to clavate-cylindrical, with rounded fertile apices, on short stipes	Applanate	Applanate
Color	Externally copper- to bronze-colored to brown with black of ostioles, internally creamy white	Black	Externally black and internally yellow
Texture	Very hard	Very hard	Very hard
Surface	Smooth except for ostiolar slightly raised	Smooth	Smooth
Perithecia			
Shape	Completely immersed, subglobose	Obovoid to tubular	Obovoid to tubular
Size	0.5-0.7 mm diameter	0.2-0.3 mm diameter x 0.3-0.5 mm high	0.3-0.4 mm diameter x 0.4-0.5 mm high
Ostiole	Umbilicate to slightly raised	Slightly higher than stromatal surface	Slightly papillate
Apical apparatus	Not observed	Not observed	Discoid, 1.5 µm high x 2.5-3 µm
Ascospores			
Color	Dark brown	Dark brown	Dark brown
Shape	Ellipsoid-inequilateral with narrowly rounded	Ellipsoid, nearly equilateral with narrowly round ends	Ellipsoid, nearly equilateral with narrowly round ends
Size	21.3-25 x 8.8-10 μm	10-12.5 (-13.8) x 6.3-7.5 μm	9.2-11.9 x 5.4-6.7 μm
Germ slit	Straight to curving, oriented obliquely to the long axis of the spore, less than spore length	Straight germ slit spore-length	Straight germ slit spore-length
Habitat	On wood	On wood	On wood
Location	Trad	Trad	Songkhla
Specimen examined	SUT207	SUT212	SUT290

Table 17. (Col
----------------

Character	Kretzschmaria species *	Nemania species *
Stromata		
Shape	Clustered subglobose fertile head with short stalk, not	Erumpent to superficial
Color	Dranched	Dlask
Color	Externally blackish, internally white	Black
Texture	Woody	Very hard
Surface	Cracked into minute angular scales	Smooth with slightly papillate
Perithecia		
Shape	Completely immersed, subglobose	Completely immersed, subglobose
Size	0.2-0.3 mm diameter	0.5-0.6 mm diameter
Ostiole	Inconspicuous papillate	
Apical apparatus	Not observed	Rectangular, 2-3 µm high x 1-1.5µm broad
Ascospores		
Color	Brown to dark brown	Brown to dark brown
Shape	Ellipsoid-inequilateral with rounded ends	Ellipsoid-inequilateral with rounded ends
Size	8.8-10 x 3.8-5 μm	8.9-11.7 x 4.7-6 μm
Germ slit	Straight spore length	Straight spore length
Habitat	On wood	On wood
Location	Songkhla	Trad
Specimen examined	SUT101	SUT258


**Figure 48.** *Xylaria anisopleura* (Mont.) Fr. (SUT205); (a) and (b) stromatal form (Bars = 1 cm and 1 mm respectively), (c) perithecia (Bar = 2 mm), (d) ascospores with germ slits (arrowed) (Bar = 10  $\mu$ m), and (e) cultural characteristics on PDA cultured at 25°C after 4 weeks (Bar = 1 cm).



**Figure 49.** *Xylaria badia* Pat. (SUT076); (a) stromatal form (Bar = 1 cm), (b) perithecia (Bar = 1 mm), (c) stromatal form (Bar = 0.2 mm), (d) ascospores (Bar = 10  $\mu$ m), (e) apical apparatus (arrowed) (Bar = 1  $\mu$ m), (f) straight germ slit less than spore length (Bar = 1  $\mu$ m), and (g) cultural characteristics on PDA cultured at 25°C after 4 weeks (Bar = 1 cm).

*Xylaria beccari* SUT092 (Figure 50) examined closely fitted the species *Xylaria beccari* Lloyd. as described by Lloyd (1924).

*Xylaria brachiata* SUT078 and SUT175 (Figure 51) examined were similar to *X. brachiata* Sacc. described by Lloyd (1919) except for ascospore sizes, which were (8-)10-11.3(-12.5) x  $3.8-5 \mu m$  and (11-14(-16) x  $5-6 \mu m$ ) respectively.

*Xylaria cubensis* SUT089, SUT090, SUT193, SUT194, and SUT199 (Figure 52) examined were similar to *Xylaria cubensis* (Mont.) Fr. described by Rogers and Samuels (1986), Rogers *et al.* (1988), González and Rogers (1989), and Thienhirun (1997). These collections had slightly smaller ascospores ((6.3-)7.5-8.8 x 3.8-5  $\mu$ m) compared to those given by Rogers and Samuels (1986), Rogers *et al.* (1988), and González and Rogers (1989) (8-10.5 x 4-5  $\mu$ m). But these *Xylaria* species were similar to specimens found in Thailand by Thienhirun (1997). *Xylaria cubensis* has been found in various tropical, subtropical, and temperate localities of the world.

*Xylaria inthino-velutina* SUT091 and SUT123 (Figure 53) examined were very similar to *Xylaria inthino-velutina* (Mont.) Fr. (Dennis, 1957; González and Rogers, 1989; and Thienhirun, 1997), but they were slightly different in ascospore size range, which were (7.5-)8.8-10(-12.5) x 3.8-4  $\mu$ m and 10-11.3 x 5-5.6  $\mu$ m respectively. This taxon is also closely related to *X. culleniae* except for ascospore size of *X. culleniae*, which is smaller (7.5-8.8 x 3-3.8  $\mu$ m). *Xylaria inthino-velutina* usually occurs on legume fruits.



**Figure 50.** *Xylaria beccari* Lloyd. (SUT092); (a) and (b) stromatal forms (Bars = 1 and 2 mm respectively), (c) perithecia (Bar = 1 mm), (d) ascospores (Bar = 5  $\mu$ m), (e) apical apparatus (arrowed) (Bar = 1  $\mu$ m), (f) straight germ slit spore length (Bar = 1  $\mu$ m), and (g) cultural characteristics on PDA cultured at 25°C after 4 weeks (Bar = 1 cm).



**Figure 51.** *Xylaria brachiata* Sacc. (SUT078); (a) and (b) stromatal forms (Bars = 1 cm and 0.5 mm respectively), (c) perithecia (Bar = 0.5 mm), (d) ascospores (Bar = 10  $\mu$ m), (e) straight germ slit spore length (arrowed) (Bar = 5  $\mu$ m), (f) apical apparatus (arrowed) (Bar = 5  $\mu$ m), and (g) cultural characteristics on PDA cultured at 25°C after 4 weeks (Bar = 1 cm).



**Figure 52.** *Xylaria cubensis* (Mont.) Fr. (SUT089); (a) and (b) stromatal forms (Bars = 1 cm and 0.5 mm respectively), (c) perithecia (Bar = 0.5 mm), (d) ascospores (Bar  $= 5 \mu$ m), (e) apical apparatus (arrowed) (Bar  $= 2 \mu$ m), and (f) cultural characteristics on PDA cultured at 25°C after 4 weeks (Bar = 1 cm).



**Figure 53.** *Xylaria inthino-velutina* (Mont.) Fr. (SUT123); (a) and (b) stromatal forms (Bars = 1 cm and 0.2 mm respectively), (c) perithecia (Bar = 0.2 mm), (d) ascospores (Bar =  $10 \ \mu m$ ), (e) apical apparatus (arrowed) (Bar =  $1 \ \mu m$ ), (f) straight germ slit spore length (Bar =  $2 \ \mu m$ ), (g) cultural characteristics on PDA cultured at 25°C after 4 weeks (Bar =  $1 \ cm$ ), and (h) exudates from anamorph (Bar =  $0.1 \ mm$ ).

*Xylaria* cf. *juruensis* SUT035 (Figure 54), *Xylaria* cf. *juruensis* SUT088 (Figure 55), *Xylaria* cf. *juruensis* SUT140 (Figure 56), and *Xylaria* cf. *juruensis* SUT170 (Figure 57) examined were similar to both *X. juruensis* P. Henn. and *X. multiplex* (Kunze.) Fr. (Dennis, 1957 and 1961; González and Rogers, 1989; and Thienhirun, 1997). All of them differed slightly in stromatal form, apical apparatus, and ascospore shape and size. Their ascospores were in the same size range and they were between *X. juruensis* (9-11(-12) x (3.5-)4-4.5  $\mu$ m), and *X. multiplex* (14.5-17(-18) x 5-5.5(-6.5)  $\mu$ m).

*Xylaria juruensis* var. *microspora* SUT129, SUT138, and SUT139 (Figure 58) closely matched the species as described by Thienhirun (1997) except that the stipes of the taxon examined were shorter and broader, and the ascospores were smaller than the previous reported species.

*Xylaria maitlandii* SUT177 (Figure 59) was similar to *X. maitlandii* (Dennis) D. Hawksw. as described by González and Rogers (1989). This taxon was different from material described by Dennis (as *Xylosphaera*) from Africa (1958) which the fertile part had hair on.

*Xylaria mellisii* SUT074 and SUT192 (Figure 60) were similar to *Xylaria mellisii* (Berk.) Cooke. as described by Van der Gucht (1995) from Papua New Guinea.

*Xylaria multiplex* SUT028 (Figure 61) was similar to *X. multiplex* (Kunze) Fr. as described by Dennis (1957; 1961), González and Rogers (1989), and Thienhirun (1997) except for the ascospore size range of  $11.3-13.8(-15) \ge 3.8-5 \ \mu\text{m}$  cf. 9-10.5(-11) x 3.5-4  $\mu\text{m}$  (Dennis, 1957; 1961; Thienhirun, 1997; González and Rogers, 1989).



**Figure 54.** *Xylaria* cf. *juruensis* (SUT035); (a) and (b) stromatal forms (Bars = 0.5 cm and 0.5 mm respectively), (c) perithecia (Bar = 0.5 mm), (d) ascospores (Bar = 5  $\mu$ m), (e) straight germ slit spore length (Bar = 2  $\mu$ m), and (f) apical apparatus (arrowed) (Bar = 2  $\mu$ m).



**Figure 55.** *Xylaria* cf. *juruensis* (SUT088); (a) and (b) stromatal forms (Bars = 5 and 1 mm respectively), (c) acute apex (Bar = 1 mm), (d) perithecia (Bar = 0.5 mm), (e) ascospores (Bar = 5  $\mu$ m), (f) straight germ slit spore length (Bar = 5  $\mu$ m), (g) apical apparatus (arrowed) (Bar = 1  $\mu$ m), and (h) cultural characteristics on PDA cultured at 25°C after 4 weeks (Bar = 1 cm).



**Figure 56.** *Xylaria* cf. *juruensis* (SUT140); (a) and (b) stromatal forms (Bars = 1 and 0.1 cm respectively), (c) stalk (Bar = 1 mm), (d) perithecia (Bar = 0.5 mm), (e) ascospores (Bar = 5  $\mu$ m), (f) apical apparatus (arrowed) (Bar = 1  $\mu$ m), and (g) straight germ slit spore length (Bar = 5  $\mu$ m).



Figure 57. *Xylaria* cf. *juruensis* (SUT170); (a) and (b) stromatal forms (Bars = 1 cm and 1 mm respectively), (c) stalk (Bar = 1 mm), (d) perithecia (Bar = 0.5 mm), (e) ascospores (Bar = 5  $\mu$ m), and (f) apical apparatus (arrowed) (Bar = 1  $\mu$ m).



**Figure 58.** *Xylaria juruensis* var. *microspora* (Thienhirun, 1997); (a), (b), (c), and (e) stromatal forms of SUT129, SUT138, SUT139, and SUT129 respectively (Bars = 1, 1, 1, and 0.01 cm), d) perithecia (Bar = 0.5 mm), (f) straight germ slit spore length (Bar = 2  $\mu$ m), and (g) ascospores with apical apparatus (arrowed) (Bar = 10  $\mu$ m).



Figure 59. Xylaria maitlandii (Dennis) D. Hawksw (SUT177); (a), (b) and (c) stromatal forms (Bars = 0.5 cm, 2 mm, and 0.2 mm respectively), (d) perithecia (Bar = 0.5 mm), (e) straight germ slit spore length (Bar = 2  $\mu$ m), and (f) ascospores (Bar = 10  $\mu$ m).



**Figure 60.** *Xylaria mellisii* (Berk.) Cooke. (SUT192); (a) and (c) stromatal forms (Bars = 0.2 cm and 0.4 mm respectively), (b) perithecia (Bar = 0.5 mm), (d) ascospores (Bar = 15  $\mu$ m), (e) straight germ slit spore length (Bar = 5  $\mu$ m), and (f) cultural characteristics on PDA cultured at 25°C after 4 weeks (Bar = 1 cm).



**Figure 61.** *Xylaria* cf. *multiplex* (SUT028); (a) and (c) stromatal forms (Bars = 0.5 cm and 0.5 mm respectively), (b) perithecia (Bar = 0.5 mm), (d) ascospores (Bar = 10  $\mu$ m), (e) apical apparatus (arrowed) (Bar = 1  $\mu$ m), (f) straight germ slit spore length (arrowed) (Bar = 2  $\mu$ m), (g) cultural characteristics on PDA at 25°C after 4 weeks (Bar = 1 cm), and (g) exudates from anamorph (arrowed) (Bar = 0.1 mm).

*Xylaria muscula* SUT029 (Figure 62) appeared identical to the species *Xylaria muscula* Lloyd. described by Dennis (1957).

*Xylaria psidii* SUT124, SUT125, and SUT126 (Figure 63) were very close to the species *Xylaria psidii* J.D. Rogers & Hemmes. described by Rogers, Ju and Hemmes (1992), and Thienhirun (1997).

*Xylaria schweinitzii* SUT201 (Figure 64) fitted very well with the species *Xylaria schweinitzii* Berk. & M.A. Curtis as described by Rogers *et al.* (1988), González and Rogers (1989), and Thienhirun (1997), except that the ascospore size differed from specimens reported by Thienhirun (1997) which were longer, 21-26.3 x 6.5-8  $\mu$ m cf. 18.8-21.3 x 6.3-7.5  $\mu$ m, than the present study.

*Xylaria scruposa* SUT117 (Figure 65) matched very closely the species *Xylaria scruposa* (Fr.) Fr. as reported by Van der Gucht (1995) from Papua New Guinea.

*Xylaria* species 2 (SUT127, SUT128, SUT130, SUT132, SUT134, SUT155, SUT171, SUT195, SUT271 and SUT274) (Figure 66) examined was similar to *Xylaria* species 2 described by Thienhirun (1997). The taxon *Xylaria* species 2 was distinctive with its yellow-colored internal stromatal tissue and spiraling germ slit. Both taxa were widely distributed in Thailand.

*Xylaria* sp. nov. (Figure 67) was a new species. Characteristics of this taxon are as follows: stromata cylindrical, gregarious, with narrowly rounded fertile apices, smooth stipe, which was longitudinally furrowed or wrinkled; external colour dark brown to black, internal colour white; texture woody; surface rough, finely reticulately cracked into small angular closely spaced scales so as to outline the individual perithecia; perithecia immersed, sometimes vaguely evident in outline,

subglobose, 0.3- 0.5 mm diameter; ostiole slightly papillate; apical apparatus quadrate to rectangular, 1.5  $\mu$ m high x 1.5-2  $\mu$ m broad; ascospore light brown to brown to dark brown, ellipsoid-equilateral with narrowly rounded ends, (7.5-)8.8-10 x (2.5-)3.8-5  $\mu$ m; germ slit straight full length.

Specimens examined: Thailand, Suranaree University of Technology, Nakhon Ratchasima, 20 September 2003, Suwannasai, N. (Holotype SUT195), SUT006, SUT012, SUT131, SUT133, SUT136, SUT141, SUT143 SUT172; Ratchaburi Province, SUT027, SUT031, SUT033, SUT034; Songkhla Province, SUT087, SUT093; Trad Province, SUT197, SUT198; Kanchanaburi Province, SUT272, SUT273, and SUT275.

Colonies on PDA covering 9 cm Petri dish in two weeks at room temperature, 23-28°C, at first white, velvety, zonate, becoming overlaid with a grayish brown layer of felty mycelium darkening to brownish black and patchily covered in areas with a thin white mycelial layer; stromata cylindrical, unbranched, developing at periphery of zones, grayish black with whitish interiors.

*Xylaria telfairii* SUT206 (Figure 68) appeared identical the species *Xylaria telfairii* (Berk.) Fr. as described by Dennis (1961), Rogers *et al.* (1987 and 1988), Callan and Rogers (1990), and Thienhirun (1997).



**Figure 62.** *Xylaria muscula* Lloyd. (SUT029); (a) and (b) stromatal forms (Bars = 0.5 cm and 0.2 mm respectively), and (c) perithecia (Bar = 0.2 mm).



**Figure 63.** *Xylaria psidii* J.D. Rogers & Hemmes. (SUT125); (a), (b) and (c) stromatal forms (Bars = 0.5, 0.1, and 0.03 cm respectively), (d) ascospores (Bar = 5  $\mu$ m), (e) straight germ slit spore length (arrowed) (Bar = 2  $\mu$ m), (f) apical apparatus (arrowed) (Bar = 2 $\mu$ m), (g) cultural characteristics on PDA cultured at 25°C after 4 weeks (Bar = 1 cm), and (h) exudates from anamorph (arrowed) (Bar = 0.5 mm).



Figure 64. Xylaria schweinitzii Berk. & M.A. Curtis. (SUT201); (a) and (b) stromatal forms (Bars = 1 cm and 3 mm respectively), (c) ascospores (Bar = 10  $\mu$ m), and (d) straight to slightly spiraling germ slit (arrowed) (Bar = 2  $\mu$ m).



**Figure 65.** *Xylaria scruposa* (Fr.) Fr. (SUT117); (a) and (c) stromatal forms (Bars = 1 cm and 0.5 mm respectively), (b) perithecia (Bar = 0.5  $\mu$ m), (d) ascospores (Bar = 5  $\mu$ m), (e) apical apparatus (arrowed) (Bar = 2 $\mu$ m), (f) straight to slightly sigmoid germ slit slightly obliquely oriented to the long axis of the spore (arrowed) (Bar = 1  $\mu$ m), and (g) cultural characteristics on PDA cultured at 25°C after 4 weeks (Bar = 1 cm).



**Figure 66.** *Xylaria* species 2 (SUT155); (a) and (c) stromatal forms (Bars = 1 cm and 0.5 mm respectively), (b) perithecia (Bar =  $0.5 \mu m$ ), (d) ascospores (Bar =  $10 \mu m$ ), (e) slightly sigmoid germ slit (arrowed) (Bar =  $1 \mu m$ ), and (f) cultural characteristics on PDA cultured at 25°C for 2 weeks (Bar = 1 cm).



Figure 67. *Xylaria* sp. nov.(SUT195); (a) and (c) stromatal forms (Bars = 0.5 cm and 0.3 mm respectively), (b) perithecia (Bar =  $0.5 \mu$ m), (d) ascospores (Bar =  $10 \mu$ m), (e) apical apparatus (Bar =  $1 \mu$ m), (f) straight germ slit spore length (arrowed) (Bar =  $2 \mu$ m), (g) cultural characteristics on PDA cultured at 25°C for 2 weeks (Bar = 1 cm), and (h) exudates from anamorph (Bar = 0.2 mm).



**Figure 68.** *Xylaria telfairii* (Berk.) Fr. (SUT206); (a) and (c) stromatal forms (Bars = 1 cm and 0.5 mm respectively), (b) perithecia (Bar = 0.5  $\mu$ m), (d) ascospores (Bar = 10  $\mu$ m), (e) straight germ slit (arrowed) (Bar = 1  $\mu$ m), and (f) cultural characteristics on PDA cultured at 25°C after 2 weeks (Bar = 1 cm).

Eight collections examined were found to be unidentified species. They were placed into four groups according to their morphological characteristics. Firstly, Xylaria taxonomic species 1 (SUT075) (Figure 69) which had ascospore sizes and germ slit form similar to X. mellisii (Berk.) Cooke as recorded by Van der Gucht (1995) but the stromatal form was different. This taxon was no peeling layer on the external surface, and no apex. Secondly, Xylaria taxonomic species 2 (SUT203) (Figure 70) had peeling layer on the external surface liked X. juruensis var. microspora (Thienhirun, 1997) and X. multiplex (Kunze) Fr. (Dennis, 1961; González and Rogers, 1989; Thienhirun, 1997). Stipes were longer than those species and ascospore size was different. Thirdly, Xylaria taxonomic species 3 (SUT204) (Figure 71) which was close to X. gracillima (Fr.) Fr. in stromatal structure as described by Van der Gucht (1995). It was different in ascospore size and germ slit form, which were 10-12(-13) x 4-5  $\mu$ m and straight germ slit less than spore length for X. gracillima (Fr.) Fr. respectively. The specimen was unculturated. Lastly, Xylaria taxonomic species 4 (Figure 72) had a stroma similar to X. allantoidea (Berk.) Fr. but the ascospore size range was closer X. schweinitzii Berk. & M.A. Curtis as described by Van der Gucht (1995).

*Kretzschmaria* species SUT101 and *Nemania* species SUT258 were shown in Figures 73 and 74 respectively. *Biscogniauxia capnodes* SUT212 (Figure 75) fitted *Biscogniauxia capnodes* (Berk.) Y.-M. Ju & J.D. Rogers as reported by Ju and Rogers (1998) whilst *Biscogniauxia* species (SUT290) sp. nov. was another new species showed in Figure 76.



**Figure 69.** *Xylaria* taxonomic species 1 (SUT075); (a) and (c) stromatal forms (Bars = 0.5 cm and 0.5 mm respectively), (b) perithecia (Bar  $= 0.3 \mu$ m), (d) ascospores (Bar  $= 12 \mu$ m), (e) straight to slightly sigmoid germ slit less than spore length (arrowed) (Bar  $= 2 \mu$ m), and (f) apical apparatus (Bar  $= 2 \mu$ m).



Figure 70. *Xylaria* taxonomic species 2 (SUT203); (a) and (c) stromatal forms (Bars = 1 cm and 0.5 mm respectively), (b) perithecia (Bar  $= 0.5 \mu$ m), (d) ascospores (Bar  $= 10 \mu$ m), and (e) cultural characteristics on PDA cultured at 25°C after 4 weeks (Bar = 1 cm).



Figure 71. *Xylaria* taxonomic species 3 (SUT204); (a) and (b) stromatal forms (Bars = 1 cm and 0.5 mm respectively), (c) straight germ slit spore length (arrowed) (Bar = 2  $\mu$ m), and (d) ascospores (Bar = 8  $\mu$ m).



Figure 72. *Xylaria* taxonomic species 4 (SUT207); (a) and (c) stromatal forms (Bars = 1 cm and 0.5 mm respectively), (b) perithecia (Bar = 0.5  $\mu$ m), (d) ascospores (Bar = 10  $\mu$ m), and (e) sigmoid germ slit (arrowed) (Bar = 5  $\mu$ m).



Figure 73. *Kretzschmaria* species (SUT101); (a) and (c) stromatal forms (Bars = 1 cm and 0.2 mm respectively), (b) perithecia (Bar = 0.3  $\mu$ m), and (d) cultural characteristics on PDA cultured at 25°C after 4 weeks (Bar = 1 cm).



Figure 74. *Nemania* species (SUT258); (a) and (b) stromatal forms (Bars = 1 cm and 0.5 cm respectively), (c) apical apparatus (Bar = 2  $\mu$ m), and (d) ascospores (Bar = 12  $\mu$ m).



Figure 75. Biscogniauxia capnodes (Berk.) Y.-M. Ju & J.D. Rogers (SUT212); (a) stromatal forms (Bar = 1 cm), (b) ascospores (Bar = 0.5 μm), (c) cultural characteristics on PDA cultured at 25°C after 4 weeks (Bar = 1 cm), and (d) straight germ slit spore length (arrowed) (Bar = 2 μm).



**Figure 76.** *Biscogniauxia* sp. nov. (SUT290); (a) and (c) stromatal forms (Bars = 1 cm and 0.5 mm, respectively), (d) ascospores (Bar =  $0.5 \mu$ m), (e) straight germ slit spore length (arrowed) (Bar =  $2 \mu$ m), apical apparatus (arrowed) (Bar =  $2 \mu$ m), and (g) cultural characteristics on PDA cultured at  $25^{\circ}$ C after 4 weeks (Bar = 1 cm).

## 4.3 Chemotaxonomic study of the selected xylariaceous fungi

Since species of *Xylaria* have been reported as endophytes in several plants, the pattern of their secondary metabolites is another possible way to identify species. Therefore, *Xylaria* species which have been recorded as endophyte were selected to study.

## **4.3.1** TLC analysis of secondary metabolites from agar plugs

Four isolates of *X. anisopleura* (ST2329), *Xylaria* species (ST2372), *X. cubensis* (ST2326), and *X. grammica* (ST2348) were selected to study on their secondary metabolite profiles. After growing culture on YES agar, the secondary metabolites were extracted and analyzed by TLC method. There were no spots observed under both visible light and UV light. This might be the result of low concentration of secondary metabolites. This method might not be suitable for those xylariaceous collections, although this technique was frequently used in several fungi such as *Penicillium*, *Chaetomium*, *Fusarium*, *Verticillium*, and *Metarhizium* (Filtenborg and Frisvad, 1980; Filtenborg, Frisvad, and Svendsen, 1983; Lund and Frisvad, 1994).

## 4.3.2 TLC analysis of secondary metabolites from cultural broth

## 4.3.2.1 Secondary metabolite extraction from 100 mL of cultural broth

Four isolates of *Xylaria* were examined for their secondary metabolite profiles by extracting the metabolites from their cultural broths. Two isolates of *Xylaria* endophytes obtained by Dr. Nuttaporn Ruchichakhon (Ruchikachorn, 2005) were included. The TLC pattern of secondary metabolites of all isolates was not clear and they were similar (Figure 77), even though they were different in their species identification by their morphological characteristics.



Figure 77. Secondary metabolite profiles of *Xylaria* isolates compared to those of *Xylaria* endophytes extracted from 100-mL cultural broth and analyzed by TLC method under visible light. UX5 = *Xylaria hypoxylon* endophyte, UX3 = *Xylaria apiculata* endophyte, X1 = *X. cubensis* ST2326, X2 = *X. arbuscula* var. *microspora* ST2372, X3 = *X. anisopleura* ST2329, and X4 = *X. grammica* ST2348.
The patterns of secondary metabolites observed in this analysis might be commonly found in *Xylaria* isolates. However, other different secondary metabolites might be not observed because of their low concentrations.

#### 4.3.2.2 Secondary metabolite extraction from 1 L of cultural broth

An isolate of *Xylaria*, *X. cubensis* ST2326, was selected to culture in 1-L MA broth, then extracted for secondary metabolites and analyzed by TLC. The secondary metabolite profile of *X. cubensis* ST2326 was compared to other *Xylaria* endophytes. The profile exhibited clearly the different patterns of each isolate (Figures 78 and 79). These profiles could be used to identify species especially endophytes. The compound, which had  $R_f$  values around 0.44-0.46, was commonly found in all isolates. *Xylaria cubensis* ST2326 consisted of two different bands which were 0.44 and 0.64  $R_f$  value. The profile of *X. cubensis* ST2326 was not similar to any endophyte isolates.

Although this chemical technique could be used to classify the xylariaceous endophytes, it was time-consuming. Also, it required several steps to analyze and used a large volume of fungal culture for extraction.



Figure 78. The pattern of secondary metabolites extracted from *X. cubensis* and other fungal endophytes by TLC method. Lanes: PD3, *Nodulisporium* sp. endophyte; UX3, *X. apiculata* endophyte; PX1, *X. longipes* endophyte; UX5, *X. hypoxylon* endophyte; N24, *Rosellinia arcuata* endophyte; UD2, *Daldinia concentrica* endophyte; U1, *Hypoxylon rickii* endophyte; 19, *X. apiculata* endophyte; 21, *X. mali* endophyte; and X1, *X. cubensis* ST2326. The number indicated the R<sub>f</sub> value of each compound.



Figure 79. Schematic of secondary metabolites extracted from *Xylaria cubensis* and other endophytes by TLC method. Lanes: PD3, *Nodulisporium* sp. endophyte; UX3, *X. apiculata* endophyte; PX1, *X. longipes* endophyte; UX5, *X. hypoxylon* endophyte; N24, *Rosellinia arcuata* endophyte; UD2, *Daldinia concentrica* endophyte; U1, *Hypoxylon rickii* endophyte; 19, *X. apiculata* endophyte; 21, *X. mali* endophyte; and X1, *X. cubensis* ST2326. The number indicated the R<sub>f</sub> value of each compound.

# 4.4 Nucleic acid studies of the selected xylariaceous fungi

Form the morphological and chemical taxonomic results of xylariaceous specimens showed that approximately 30% of them could not be identified. Therefore, the nucleic acid method of DNA sequencing was then applied to resolve this problem. The results of DNA sequences exhibited clearly relationships between xylariaceous species and could also be used to confirm the results indicating new species.

## 4.4.1 Group I: Astrocystis and Rosellinia

The genera *Astrocystis* and *Rosellinia* are very similar in their morphological characters. There are some disagreements over the status of both genera. Ju and Rogers (1990 and 1995) and San Martín and Rogers (1994) mentioned that *Astrocystis* was accommodated as *Rosellinia*-like fungi whereas Petrini and Whalley (1996) suggested that both genera were different and should be separated from each other. They were different in the stromata character of *Astrocystis*, which spitted from the host surface, and anamorph form, which was *Acanthodochium*. The nucleotide sequences of 18S ribosomal DNA and ITS1-5.8S-ITS2 regions were analyzed to clarify the differentiation of both genera.

## 4.4.1.1 18S rDNA sequence analysis

Genomic DNA of *A. mirabilis* samples (SUT051 and SUT056) were extracted from mycelia and *R. procera* samples (SUT102, SUT109, SUT113, and SUT114) were extracted from ascospores because they could not be cultured. An isolate of *Rosellinia* sp. ST2310 obtained by Dr. Surang Thienhirun was included. The quality and quantity of the DNA solution were measured and adjusted to a suitable concentration for PCR amplification. It was found that no PCR product

was obtained from NS1/NS8 primers, although several modified conditions were performed. Therefore, two new primers, NS4 and SR8R (Table 18), were added to resolve this problem. The amplification of 18S rDNA sequence was divided into two steps by using two sets of primers. The NS1/NS4 primers were used to amplify the fragment at position 20 to 1,131 of 18S rDNA. The SR8R/NS8 primers were used to amplify the fragment at position 732 to 1,769 of 18S rDNA (Figure 8). The program of amplification consisted of 1 cycle of 95°C for 5 min; 35 cycles of 95°C for 1 min, 53°C for 1.30 min, 72°C for 2 min; and the final cycle of 72°C for 10 min. The PCR reactions were carried out in the automated thermal cycle (i-cycle, BioRad, U.S.A.). The amplified 18S rDNA fragments are shown in Figure 80.

 Table 18.
 Nucleotide sequences of NS4 and SR8R primers.

Name	Sequence (5'- 3')	Target region <sup>a</sup>	Reference
NS4	CTTCCGTCAATTCCTTTAAG	SSU 1150-1131	White <i>et al</i> . 1990
SR8R	GAACCAGGACTTTTACCTT	SSU 732-749	Vilgalys, www, 1999

<sup>a</sup> Saccharomyces cerevisiae numbering



Figure 80. Gel electrophoresis of partial 18S rDNA fragments. Lanes: M, DNA marker (100 bp DNA ladder, Invitrogen); 1, 3, and 5, Astrocystis mirabilis (SUT051), A. mirabilis (SUT056) and Rosellinia sp. ST2310 using NS1 and NS4 primers respectively; 2, 4, and 6, Astrocystis mirabilis (SUT051), A. mirabilis (SUT056) and Rosellinia sp. ST2310 using SR8R and NS8 primers respectively.

For 18S rDNA amplification, only genomic DNA extraction from mycelium could be achieved by both set of DNA primers. Although several modified conditions were attempted, no PCR product was obtained from the genomic DNA extraction from ascospores. This might be because of the low concentration of genomic DNA extracted from ascospores, or the degradation of genomic DNA. The size of amplified 18S rDNA fragments of *A. mirabilis* (SUT051 and SUT056) and *Rosellinia* sp. (ST2310) from two primer sets were similar. They were approximately 1,000 bp obtained from the amplification by using NS1/NS4 primers and SR8R/NS8 primers. The both amplified fragments were then combined to obtain the whole 18S rDNA sequence. The length of the DNA sequences is summarized in Table 19.

Table 19.	The	length of	18S	rDNA	sequences	of A.	mirabilis	SU	T051,	SU	Т056,	and
-----------	-----	-----------	-----	------	-----------	-------	-----------	----	-------	----	-------	-----

Species	Location /source	1 <sup>st</sup> fragment (NS1/NS4 primers) (bp)	GenBank accession number	2 <sup>nd</sup> fragment (NS4/NS8 primers) (bp)	GenBank accession number	Total (bp)
A. mirabilis	Ratchaburi	1012	DQ322075	1072	DQ322076	2056
(SUT056)	Province					
A. mirabilis	Ratchaburi	1012	DQ322074	NO	ND	ND
(SUT051)	Province					
<i>Rosellinia</i> sp.	RFD*	1020	DQ322072	1202	DQ322073	2210
(ST2310)						

Rosellinia sp. ST2310 obtained from DNA sequence analysis.

\* The culture was provided by Dr. Surang Thienhirun, The Royal Forest Department, Thailand. NO = Not observed, ND = Not determined

Then, the whole 18S rDNA sequences of *A. mirabilis* (SUT056) and *Rosellinia* sp. (ST2310) including *R. necatrix* (AB014044) from GenBank database were aligned (Figure 81). The result indicated that there was dissimilarity between the genera *Astrocystis* and *Rosellinia*. The percent similarity of *A. mirabilis* (SUT056) to *Rosellinia* sp. (ST2310) and *R. necatrix* (AB014044) was 70.3% and 68.7% respectively (Table 2, Appendix C). Nevertheless, both genera were closely related and showed highly conserved regions in the beginning of 1,000 bp of 18S rDNA sequences, whilst the middle region exhibited the highest variation (Figure 81). Therefore, the partial 18S rDNA sequences of *A. mirabilis* (SUT056) and *Rosellinia* sp. (ST2310) including *R. necatrix* (AY083805) and *A. coccoes* (AY083804) available from GenBank database, which were amplified by using NS1/NS4 primers, were aligned (Figure 2, Appendix C). The result showed high similarity between the genera *Astrocystis* and *Rosellinia* ranging from 97% to 98%

similarity (Table1, Appendix C) and it confirmed the highly conserved region of both genera at the 5' end of 18S rDNA sequence (position of 20 bp to 1,131 bp; Figure 8).

		10	20	30	40	50	60 • • • • •   • • • •	
AB014044	1	CCGCGAAACT	GCGAATGGCT	CATTAAATCA	GTTATCGTTT	ATTTGATAGT	ACCTTACTAC	60
A.mirabilis	1	CGGCGAAACT	GCGAATGGCT	CATTAAATCA	GTTATTGTTT	ATTTGATAGT	ACCTTACTAC	60 60
K. Sp. 312301	T	CGGCGAAACI	GCGAAIGGCI	CATTAAATCA	GITAICGITI	ATTIGATIO	ACCITACIAC	60
		70	80	90	100	) 110	) 120	
	- 4							
ABU14044 A.mirabilis	61 61	ATGGATACCT	GTGGTAATTC	TAGAGCNNNT	ACATGCTGAA	ANATCCCGAC	TCACGGAGGG	120
R. sp. ST2301	61	TTGGATAACC	GTGGTAATTC	TAGAGCTAAT	ACATGCT-AA	AAATCCCGAC	TCACGGAGGG	119
-								
		130	140	) 150	) 160	) 170	180	
AB014044	121	ATGTATTTAT	TAGATTAAAA	ACCAATGCCC	CTCGGGGCTT	TCTGGTGATT	CATAATAACT	180
A.mirabilis	120	ATGTATTTAT	TAGATTAAAA	ACCAATGCCC	CTCGGGGGCTT	TCTGGTGATT	CATAATAACT	179
R.sp.ST2301	120	ATGTATTTAT	TAGATTAAAA	ACCAATGCCC	CTCGGGGGCTT	TCTGGTGATT	CATAATAACT	179
		190	200	) 210	220	) 230 ••••	240	
AB014044	181	TCACGAATCG	CACGGCCTTG	CGCCGGCGAT	GGTTCATTCA	AATTTCTCCC	CTATCAACTT	240
A.mirabilis	180	TCTCGAATCG	CATGGCCTTG	CGCCGGCGAT	GGTTCATTCA	AATTTCTGCC	CTATCAACTT	239
K. Sp. ST2301	190	TUTUGAATUG	CATGGCCTTG	CGCCGGCGAT	GGTTCATTCA	AATTTCTGCC	CTATCAACTT	239
		2.50	2.60	270	1 280	290	300	
AB014044 A mirchilig	241	TCGATGGCAG	GGTCTTGGCC	TGCCATGGTT	TCAACGGGTA	ACGGAGGGTT	AGGGCTCGAC	300
R. sp. ST2301	240 240	TCGATGGCAG	GGTCTTGGCC	TGCCATGGTT	ACAACGGGTA	ACGGAGGGTT	AGGGCTCGAC	299
-								
		310	320	330	340	350	360	
AB014044	301		GAGCCTGAGA	AACGGCTACT	ACATCCAAGG	AAGGCAGCAG	GCGCGCAAAT	360
A.mirabilis	300	CCCGGAGAAG	GAGCCTGAGA	AACGGCTACT	ACATTCAAGG	AAGGCAGCAG	GCGCGCAAAT	359
R.sp.ST2301	300	CCCGGAGAAG	GAGCCTGAGA	AACGGCTACT	ACATCCAAGG	AAGGCAGCAG	GCGCGCAAAT	359
		370	) 380 	) 390	) 400 	) 410 •••• ••••	) 420 ••••	
AB014044	361	TACCCAATCC	CGACACGGGG	AGGTAGTGAC	AATAAATACT	GAT ACAGGGC	TCTTTTGGGT	420
A.mirabilis	360	TACCCAATCC	CGACACGGGG	AGGTAGTGAC	AATAAATACT	GATACAGGGC	TCTTTTGGGT	419
K. Sp. 512301	360	TACCCAATCC	CGACACGGGGG	AGGIAGIGAC	AATAAATACT	GATACAGGGC	ICITIIGGGI	419
		430	440	450	9 460	470	480	
ABU14044 A mirabilis	421	CTTGTAATTG	GAATGAGTAC	AATTTAAATC	CCTTAACGAG	GAACAATTGG	AGGGCAAGTC	480
R. sp. ST2301	420	CTTGTAATTG	GAATGAGT-C	AATTTAAATC	CCTTAACGAG	GAACAATTGG	AGGGCAAGTC	478
		490	. 500	) 510	) 520	) 530	540	
AB014044	481	TGGTGCCAGC	AGCCGCGGTA	ATTCCAGCTC	CAATAGCGTA	TATTAAAGTT	GTTGCAGTTA	540
A.mirabilis	479	TGGTGCCAGC	AGCCGCGGTA	A-TTCAGCTT	CAATAGCGTA	TATTAAAGTT	GGTGCAGTTA	537
R. sp. ST2301	479	TGGTGCCAGC	AGCCGCGGTA	ATTTCAGCTC	CAATAGCGTA	TATTAAAGTT	GGTGCAGGTA	538
		550	560 ۰۰۰۰ ۰۰۰۰	) 570 •••• ••••	) 580 	, 590 	, 600 	
AB014044	541	AAAAGCTCGT	AGTTGAACCT	TGGGCCTGGC	TGGCCGGTCC	GCCTCACCGC	GTGCACTGGT	600
A.mirabilis R en em2301	538	AAA-GCTCGT	AGTTGAACCT	TGGGCCTGGC	TGGCCGGTC-	GCCTCAACGC	GTGCACTGGT	595
R. 39. 012301	507	inn gorogi	19119AAUUI	1333001-90	1330033100	JU TONNOGU	2130001991	593
		610	620	) 630	) 640	) 650	660	
30014044	601							600
A.mirabilis	оот 596	TCGGCCGGGC	CTTTCCCTTT	GGGGGAGCCCT	ATGCCCTTCA	CTGGGTGTAG	TGGGGGAACCA	655 655
R.sp.ST2301	596	TCGGCCGGGC	CTTTTCCTCT	GGGGAGGCCT	ATGCCCTTCA	CTGGGTGG	TAGGGAACCA	653

Figure 81. Sequence alignment of 18S rDNA *Rosellinia* sp. (ST2310), *Rosellinia necatrix* (AB014044), and *A. mirabilis* (SUT056).

670 680 690 700 710 720 661 GGACTTTTAC TGTGAAAAAA TTAGAGTGTT CAAAGCAGGC ATTTGCTCGA ATACATCAGC 720 AB014044 720 A.mirabilis GGACTTTTAC TGTGAAAAAA TTAGAGTGTT CAAAGCAGGC CTATGCTCGA ATACATCAGC 656 715 GGACTTTTAC TGTGAAAAAA TTAGAGTGTT CAAAGCAGGC CTATGCTCGA ATACATCAGC R.sp.ST2301 654 713 770 
 730
 740
 750
 760
 770
 780

 ATGGAATAAT AGAATAGGAC
 GTGTGGTTCT
 ATTTTGTTGG
 TTTCTAGGAC
 CGCCGTAATG
 780 780 AB014044 721 ATGGAATAAT AGAATAGGAC GTGTGGTTCT ATTTTGTTGG TTTCTAGGAC CGCCGTAATG A.mirabilis 716 775 ATGGAATAAT AGAATAGGAC GTGTGGTTCT ATTTTGTTGG TTTCTAGGAC CGCCGTAATG R. sp. ST2301 714 773 
 790
 800
 810
 820
 830
 840

 ATTAATAGGG ACAGTCGGGG GCATCAGTAT TCAATTGTCA GAGGTGAAAT TCTTGGATTT
 AB014044 781 840 A.mirabilis 776 ATTAATAGGG ACAGTCGGGG GCATCAGTAT TCAATTGTCA GAGGTGAAAT TCTTGGATTT 835 R. sp. ST2301 774 ATTAATAGGG ACAGTCGGGG GCATCAGTAT TCAATTGTCA GAGGTGAAAT TCTTGGATTT 833 
 850
 860
 870
 880
 890
 900

 ATTGAAGACT
 AACTACTGCG
 AAAGCATTTG
 CCAAGGATGT
 TTTCATTAAT
 CAGGAACGAA
 900 AB014044 900 841 ATTGAAGACT AACTACTGCG AAAGCATTTG CCAAGGATGT TTTCATTAAT CAGGAACGAA A.mirabilis 895 836 ATTGAAGACT AACTACTGCG AAAGCATTTG CCAAGGATGT TTTCATTAAT CAGGAACGAA R.sp.ST2301 834 893 960 AB014044 901 960 A.mirabilis 896 955 **R.sp.ST2301** 894 AGTTAGGGGA TCGAAGACGA TCAGATACCG TCGTAGTCTT AACCATAAAC TATGCCGACT 953 AB014044 961 1020 A.mirabilis AGGGATCGGA CGATGTTATT TTTTGACTCG TTCGGCACCT TACGAGAAAT CAAAGTC---956 1012 R.sp.ST2301 954 AGGGATCGGA CGATGTTATT TTTTGACTCG TTCGGCACCT TACGAGAAAT CAAAGTCTT-1012 1080 AB014044 1080 A.mirabilis 1025 R. sp. ST2301 1012 --- TCCT--- ----- TT GATCCT---- --- GCGGACG GG-GAGAAGG 1040 1090 1100 1110 1120 1130 1140 AB014044 1139 A.mirabilis 1025 -CCTGGA-TG CTGCGCGCTA --GCCCTATT ATCA--CCTA G---CC--GT -CAAACAAGG R.sp.ST2301 1041 TCCAAGA--- -ACAGGAT-- --TCAACAAT AGCAGGAAGA GGAGGCCTAG ACCCGGAGTT 1073 1092 
 1150
 1160
 1170
 1180
 1190
 120

 ....|....|
 ....|....|
 ....|....|
 ....|....|
 1140
 GGTTGGTGGT
 AGGCCCTCGA
 TATATGCTAG
 TCAGGTGGTA
 ATAATCTGCT
 GTATTAAATA
 1200 AB014044 1199 

 A. mirabilis
 1074
 GCTCAGT-- ---CCTC- -TAG
 TAAGG--- ------A

 R.sp.ST2301
 1093
 GANTAAGGC -ANGACACGA
 TATATAACA CAAGGCG-- ------A

 1093 1126 
 1210
 1220
 1230
 1240
 1250
 1260

 AB014044
 1200
 GTGAGGTTAT
 TCCTGGCGAC
 ATCCTCAAAAT
 TGCGGGGGAAG
 CCCTACAACA
 AAAGCAATGA

 A.mirabilis
 1094
 G----- -----GCTG
 TC-----G
 AAAGCA--- 1259 1112 R. sp. ST2301 1126 ------ CTTAGCCAT GTC----T GACAGAGAGAG ------G GGAGCAGAG-1159 
 1270
 1280
 1290
 1300
 1310
 1320

 1260
 CTACTAAGCG
 CGCCTTGAAA
 AAGAGCGCGT
 GGCGGAGCGT
 AACGGCTCCG
 GTACAGTAAG
 AB014044 1319 A. mirabilis 1113 CCACTA-C- --TCAT--- --AGCACTT CGC---CGC A--G--- ---AGAGTG 1144 R.sp.ST2301 1160 CTACAC--CG GACCAT--- --GAGCAACT TGCGGAAGGT GA----- --ACCATACG 1201

Figure 81. (Continued).

1330 1340 1350 1360 1370 138 ....|....| ....| ....| ....| ....| ....| ....| ....| ....| ....| ....| ....| ....| ....| ....| ....| ....| ....| 1320 AACGTGATTG CCTGGGGTCA TCCGCAGCCA AGCTCCTTAT AGGGATATAT -GAGAGAAGG 1380 AB014044 A.mirabilis 1145 CA-G--ACTC AACG---T-A ---GCTA-TA CGC-AC---- ---ACGTAT -GCC-----R.sp.ST2301 1202 GAAG--GACA CTAAAGGTAA TAAGTCA-CT GGCTCA---- -AGGGTAGGT CGAG------1247 
 1390
 1400
 1410
 1420
 1430
 1440

 1379
 TTCAGAGACT
 TGACGGGGAT
 GGGTGAACTC
 GCAACCAGGT
 TCGCTTAAGA
 TAAAGTCCAT
 1438
 AB014044 A.mirabilis 1177 -TTGGCGA-- -G-CGAGA-- G------ ---ACCAT-- -CGC-AT-GA CA--G-GGA-1209 R. Sp. ST2301 1247 --CGAAGA-- GGATGAGGA- -----ACCG GCCGAGTGGT AACCATAAAC TAT-GAATGC 1295 
 1450
 1460
 1470
 1480
 1490
 150

 1439
 TAAAGGCACG
 AAAGTGTCCT
 TTTAACAACC
 CCCTATAACA
 GGGAGCCTGC
 GGCTTAATTT
 AB014044 1498 A.mirabilis 1209 -- ACAGTACG ----TGCGAA GTT---AAT- T----GAG-C TGAAGACTTC GGTAAGAT--1252 R. SP. ST2301 1296 TAGAGATGCG AAGATCGTTA GTTCTTGACT C-----GTG AGGAACGTAC GAGAAAAT--1347 
 1510
 1520
 1530
 1540
 1550
 156

 1499
 GACTCAACAC
 GGGGAAACTC
 ACCAGGTTAA
 CTGAACAGTT
 ACTGTCTGGG
 CCTGGAATAG
 1560 AB014044 1558 1252 -----GCAT AGAATAGGT- AC----- ---GGCAGCA ACCG-----A.mirabilis 1278 R. sp. ST2301 1347 ---- AAAGT CTTTGGGCTC TT----- --GGGCGAGT A-TGTCTG-- -----1379 
 1570
 1580
 1590
 1600
 1610
 1620

 AB014044
 1559
 TGATTTGTTT
 CGCTAGTGCT
 AGACACTTGT
 CTACGTGGGA
 AAGCTCCCCGA
 TTCGGACGTA
 1618

 A.mirabilis
 1278
 ------ ----TA-CACT
 CGACAGATG ------ ----GAACG-A
 1299

 R.sp. ST2301
 1379
 ----- ----AAGGCT
 -GAAACATAA
 TGAAGCGACA
 TAAGGCACGA
 CCAGAAAGAG
 1424

 1630
 1640
 1650
 1660
 1670
 168

 1619
 GAGCGGTGGC
 CTCGCTACCG
 TTGTCTAGTG
 CACACCAGCT
 GGTACAGGGA
 ACGCTAACCC
 1680 AB014044 1678 1341 R. sp. ST2301 1425 GAGCCATGCT TA----ATTG GAATAGAGGG TGAACTAGCA GGTTCCGGAA GAATGAGGGT 1480 1690 • • • | • • • • ' 
 1690
 1710
 1720
 1730
 174

 1679
 TACATTCGTA GGTATGCCAA TCCTGTGGCG AGCTCAGGTT
 GCGCTGAGCC
 GTTGCAACGC
 1740 AB014044 1738 A. mirabilis 1342 AGGA--CGT- -GAA-----A CCCT-TCGAC AGTGCA---- ----CGAATC GCTC--ATGA 1381 R. Sp. ST2301 1481 AGACAATGTA -GAA----CG CCTTTTAG-- AATTGA---- ----GGCGG GGTCCAAGGC 1524 AB014044 1798 A.mirabilis 1382 GCAGCAATGC TAGG-GC--- -----AGA CGTG-CT--A --TCAG-GCT AAA-----GG R.sp.ST2301 1525 GGGCTAAAGT GGGGAGAAA- -----AAG GTTGGCTTAA ----TGGACC CTT----AA 1419 1567 
 1810
 1820
 1830
 1840
 1850
 186

 1799
 GARATCGTAG
 CCTTCTTGAC
 TAGGTCCGAA
 TGTCCTAATC
 AAGGAGGGCA
 GGGCNCGGCT
 1860 AB014044 1858 1445 R. sp. ST2301 1568 AAAATT---- -----TCCCGA CGCCCC---- ----------CCC----1588 1920 AB014044 1918 1445 -----CACCA GGAG-CGGAG CCTGCG-GTC ------A AATTTGACTC AACACGGGGT A.mirabilis 1489 R. Sp. ST2301 1588 ----CCGCCG TTGCCCCCTC CCGCCTTATT ------ --TTGACCCC AC-CCGGGAA 1631 
 1930
 1940
 1950
 1960
 1970
 1980

 1919
 CGCCCCTCTG
 GAGAATGCAG
 ACACAATGAG
 GA-TTGACAG
 ATTGAGAGACT
 -CTTTCTTGA
 1976
 AB014044 A.mirabilis 1490 AACTCACCAG G----TCCAG ACACAATGAG GA-TTGACAG ATGGGGAGCC -CTTTTTTGA 1543 R. Sp. ST2301 1632 C-CTCCCCAG G----TCCCA GCCCATTAG GATTTCCCAG ATTGACAGCC TCTTTCCTCA 1686

Figure 81. (Continued).

202

AB014044 2029 A.mirabilis 1543 --TTTGGAGG GC-GGAG-GC CATGGCCGAC TTA--GTGGG TGGAGAAATT TGT-CCTGCC 1596 R. sp. ST2301 1687 T-TTTTGTCC GCTGGTGGTG CCTCGCCCGT CTTACGTTGC TCCATTGATT TGT-CTG--C 1742 
 2050
 2060
 2070
 2080
 2090
 2100

 AB014044
 2030
 TTAATTGCGA TAACGAACG- AGACATTTAC
 CTGCTAAATA GCCCGTATTG CTTTGGCAGT

 A.mirabilis
 1597
 TTAATTGCGA TAACCAACG- AGAAATTAC
 CGCCTAAAAA G-CCGTATGC TTTGGGCAGT
 2088 1654 R. Sp. ST2301 1743 TTAATTCCGA TGCCGACCGA AGACATTTAC CTGCTAAATA GCCCGTATTG CTTT-GCCA-1800 211021202130214021502160AB0140442089AC-GCTGGC--TNCTTAG--AGGGACTATCCGCTTAAGC-GGGTGGAAGTTGGA--TGCA.mirabilis1655TCCGCTCGCC-TTCTTTATTAGGCCCTCTCCGCTTACGCCGGGTGCAAGTTGGCATTGCR.sp. ST23011800-CGCTCGCTGCTTCTTAG--AGGGCCTCTCCGCTTCACCCGTTGCACCGT-TCGAT-C 2160 2139 1712 1854 
 2170
 2180
 2190
 2200
 2210
 2222

 2140
 AATAAC--AG
 GTCT-GTGAT
 -GCCCTTAGA
 TGTTCTGGGC
 CGCACGCGCC GTTACA-CTG
 2220 AB014044 2193 A.mirabilis 1713 CACTACCAAG GTCTTGTGTT CGCCCTTAGA TGTTCTGGCC TGCACGCGCC GTTCCT-CTG 1771 R. sp. ST2301 1855 CACTACC-AG GTC-TGTGAT -GCCCTTAGA TGTTCTGGCC CGCACGCG-C GTTCCTACTG 1910 
 2230
 2240
 2250
 2260
 2270
 2280

 AB014044
 2194
 ACAGAGG-AC
 AGCG-AGTA
 C-TTCCTTAG
 TAGAGATACT
 T---GGGTAA
 TCTTGTTAAA
 2245

 A.mirabilis
 1772
 CCA-AGCCGC
 AGCGGCAGTT
 C-TTCCTTAG
 TAGAGATACT
 T---GGGTAA
 TCTTGTTACA
 1826
 R. Sp. ST2301 1911 ACAGAACGAC AGCGCAGGTC CTTTCCTTAG TAGAGATCCT TCGCGCGGTAA TCTTGTTAC-1969 

 2290
 2300
 2310
 2320
 2330
 2340

 2246
 CTCTGT---C
 GGCTGGGGG ATA-GAGCAT
 GCCATTATT
 GCTCTTCAA CG-AG-GAAT

 1827
 CCTCTGGCC
 GGCCTGGGGGC
 ATA-GGAGCAT
 TGCAATTATT
 GCTCTTCAC CG-AG-GAAT

 AB014044 2297 A.mirabilis 1883 R. Sp. ST2301 1970 CCTC-TGT-C GTGCT-GGGC ATAC-AGCAT TGCAATTATT GCTCTTCACA CGTAGCGCCT 2025 2400 AB014044 2345 1884 TCCT-ATGTC -AGCGTAATG TCA-TCCACT -TGCG--TTG AT-TACTGTC CCTGCCC--T A.mirabilis 1934 R. sp. ST2301 2026 TCCTCATGTC TACCGTATAG TCATTCCACT CTGCGCTCTG ATCTCCTGTC CCTGCCCTAT 
 2410
 2420
 2430
 2440
 2450
 2460

 AB014044
 2345
 -TTGT--ACA
 -CCACCCCCC
 GTCGT-ACT
 ACCGA-TTGA
 -ATGGCTCAG
 TGA--G-GC 

 A.mirabilis
 1934
 -TTGT--TCA
 -CA-CCGCCC
 GTCGT-ACT
 CCCCCCTTGA
 TATGGCTCAG
 TCG-AG-GC 2393 1985 R. sp. ST2301 2086 GTCGTGATCA TCACCCCCCC GTCGCTCACT CCCTCCCTGA ATGAGCTCAG TCGTAGCGCT 2145 
 2470
 2480
 2490
 2500
 2510
 252

 2394
 TTTCGGACT ---GGCCCAG
 A-GGAGTC-G
 GCANCGACAC
 TTC--AGGGC
 CGGA-AAGTC
 AB014044 2444 A.mirabilis 1986 TTTCTGGTTC TAGGTCCCAG A-GGTGTCAG GCACCGACAC CTCCTAGGGC CGGCCAAGGT R.sp.ST2301 2146 CTTCTGAGTC TCACTCCAGG ACGGAGTCAC GCATACTGTC TACCTACAG- TGGCC--GTG 2044 2202 2530 ....|....| .... 2445 **ATCCAAACTC GGT** 2457 AB014044 A.mirabilis 2045 CTTGGAACTT T-T 2056 R. sp. ST2301 2203 AC---AACG- -CT 2210

Figure 81. (Continued).

## 4.4.1.2 ITS sequence analysis

Two representatives of *A. mirabilis* (SUT051 and SUT056) isolates and one *Rosellinia* sp. (ST2310) isolate were investigated. The sequences of ITS1-5.8S-ITS2 were analyzed. The amplified ITS1-5.8S-ITS2 fragments of all isolates were similar being approximately 500 bp as shown in Figure 82.



Figure 82. Gel electrophoresis of ITS1-5.8S-ITS2 fragments of Astrocystis and Rosellinia using ITS5 and ITS4 primers. Lanes: M, DNA marker (100 bp DNA ladder, Invitrogen); 1, Rosellinia sp. (ST2310); 2, A. mirabilis (SUT051); 3, A. mirabilis (SUT056).

After sequencing the ITS1-5.8S-ITS2 fragments, the boundaries of the ITS1 and ITS2 regions were determined by comparison to published sequences of the ITS region. The lengths of ITS1-5.8S-ITS2 sequences of *A. mirabilis* and *Rosellinia* sp. ST2310 are summarized in Table 20. The alignment of ITS1-5.8S-ITS2 sequences of *A. mirabilis* (SUT051 and SUT056) and *Rosellinia* sp. ST2310 was

performed including five available ITS1-5.8S-ITS2 sequences from the GenBank database; *R. arcuata* (AB017660), *R. pepo* (AB017659), *R. quercina* (AB017661), and *R. necatrix* (AB017657 and AB017658). The ITS1 region exhibited the highest variation whereas the 5.8S region was highly conserved and constant at 155 bp (Figure 3C in AppendixC). The ITS2 region showed only minor variation. The phylogenetic trees were then constructed using two methods of neighbour-joining method by the PHYLIP program (Felsenstein, 1995), and the maximum parsimony method by PAUP program (Swofford, 2000) as shown in Figure 83 and Appendix 1D respectively.

**Table 20.** The length of ITS1-5.8S-ITS2 sequences of A. mirabilis and Rosellinia sp.ST2310 obtained from DNA sequence analysis.

Species	Location/Source	ITS1	<b>5.8</b> S	ITS2	Total	GenBank
		(bp)	(bp)	(bp)	(bp)	accession
						number
A. mirabilis (SUT051)	Ratchaburi Province	172	155	156	483	DQ322078
A. mirabilis (SUT056)	Ratchaburi Province	172	155	156	483	DQ322076
Rosellinia sp.	RFD*	178	155	160	493	DQ322077
(ST2310)						

\* The culture was provided by Dr. Surang Thienhirun, The Royal Forest Department, Thailand.



**Figure 83.** Phylogenetic tree of *Astrocystis* and *Rosellinia* based on ITS1-5.8S-ITS2 sequences constructed by using the neighbour-joining method. *Daldinia eschscholzii* is the outgroup. Branch lengths are scaled in terms of expected numbers of nucleotide substitution per site. Numbers on branches are bootstrap values from 1,000 replication.

The phylogenetic trees obtained from two methods revealed the same pattern. *Rosellinia* sp. ST2310 was grouped together with other species of *Rosellinia* from GenBank database, *R. arcuata* (AB017660), *R. pepo* (AB017659), *R. quercina* (AB017661), and *R. necatrix* (AB017657 and AB017658), while two isolates of *A. mirabilis* (SUT051 and SUT056) were separated with high 96% bootstrap support. This result indicated the dissimilarity between both genera from a genetic point of view.

In these molecular analyses, both 18S rDNA sequences and ITS1-5.8S-ITS2 sequences demonstrated the dissimilarlity between *Astrocystis* and *Rosellinia*, which was in agreement with the concepts of Petrini (1993, 2003), Læssøe

and Spooner (1994), and Whalley (1996) to separate Astrocystis from Rosellinia.

## 4.4.2 Group II: Camillea

*Camillea tinctor* is the common species of this genus found in Thailand, and is also wide spread in the tropics and subtropics (San Martín, González, and Rogers, 1993; Whalley, 1996). The variation of ITS1-5.8S-ITS2 sequences within these species, *C. tinctor*, *C. selangorensis*, and *C. leprieurii* provided by Dr. Margaret A. Whalley, was investigated.

## 4.4.2.1 Genomic DNA extraction and ITS amplification

Genomic DNA was extracted from mycelia of two collections of *C. tinctor* (SUT161 and SUT260), and also an isolate of reference strain *C. tinctor* (ST2321) obtained by Dr. Surang Thienhirun was included. Two more collections of *C. tinctor* (SUT099 and SUT211) were too old and no ascospore was available for genomic DNA extraction. One specimen of *C. selangorensis* (KS15) obtained from Dr. Margaret A. Whalley was used for DNA extraction from ascospores which had been separated under a stereomicroscope and confirmed before the extraction. A specimen of *C. leprieurii* could not be used for the extraction of genomic DNA because the specimen was covered with other fungal mycelia making it difficult to isolate the pure ascospores without the risk of subsequent DNA contamination. Therefore, only *C. tinctor* and *C. selangorensis* were amplified for their ITS1-5.8S-ITS2 regions by using ITS5 and ITS4 as forward and reverse primers respectively. The results of amplified fragments are shown in Figure 84.



Figure 84. Gel electrophoresis of ITS1-5.8S-ITS2 fragments of *Camillea* using ITS5 and ITS4 primers. Lanes: M, DNA marker (100 bp DNA ladder, Invitrogen); 1, *Camillea tinctor* (ST2321); 2, *C. tinctor* (SUT161); 3, *C. tinctor* (SUT260); and 4, *C. selangorensis* (KS15).

The length of amplified ITS1-5.8S-ITS2 fragments of *C. tinctor* (SUT161, SUT260, and ST2321) and *C. selangorensis* (KS15) were similar in size being approximately 500 bp. Since genomic DNA of *C. selangorensis* (KS15) was extracted from ascospores, the amplified fragment was in low concentration. It was because the only small amount of ascospores was used to extract after observation and isolation under stereomicroscope to make sure they were no contamination from other fungus spores.

#### 4.4.2.2 ITS1-5.8S-ITS2 sequence analysis

The amplified ITS1-5.8S-ITS2 fragments of three *C. tinctor* isolates and one *C. selangorensis* (KS15) specimens were performed. The length of ITS1-5.8S-ITS2 sequences is reported in Table 21.

 Table 21. The length of ITS1-5.8S-ITS2 sequences of Camillea tinctor and C.

 selangorensis obtained from DNA sequence analysis.

Species	Location/Source	ITS1	<b>5.8</b> S	ITS2	Total	GenBank
		(bp)	(bp)	(bp)	(bp)	accession
						number
C. tinctor (ST2321)	RFD <sup>a</sup>	204	155	155	514	DQ322080
C. tinctor (SUT161)	Yasothorn Province	190	155	155	500	DQ322081
C. tinctor (SUT260)	Trad Province	186	155	155	496	DQ322082
C. selangorensis	Liverpool John	175	155	156	486	DQ322083
(KS15)	Moores University <sup>b</sup>					

<sup>a</sup> The culture was provided by Dr. Surang Thienhirun, The Royal Forest Department, Thailand.

<sup>b</sup> The specimen was provided by Dr. M.A. Whalley, Liverpool John Moores University, U.K., collected from Malaysia.

After sequencing the ITS1-5.8S-ITS2 fragments, the boundaries of the ITS1 and ITS2 regions were determined by comparison to published sequences of the ITS regions. The ITS1-5.8S-ITS2 sequences of both species ranged from 496 to 514 bp in length. The ITS1 region was high variable in length ranging from 186 to 204 bp whilst the 5.8S region was quite constant at 155 bp. The length of ITS2 region ranged from 155 to 159 bp. The ITS1-5.8S-ITS2 sequences of *C. tinctor* (SUT161, SUT260, and ST2321) and *C. selangorensis* (KS15), including DNA sequences of *C. tinctor* (AJ390421) available from the GenBank database, were aligned. The result revealed the high variation in ITS1 region as shown in Figure 85.

The complete ITS1-5.8S-ITS2 sequence alignment is given in Appendix 4C. The identity matrix is shown in the Table 22.



Figure 85. ITS1 sequence alignment of Camillea tinctor (AJ390421) from GenBank, C. tinctor (ST2321), C. tinctor (SUT161), C. tinctor (SUT260), and C. selangorensis (KS15) by using ClustalX and BioEdit programs. Blocks indicate the short tandem repeat (STR) sequences.

Species	<i>C. tinctor</i> (AJ390421)	<i>C. tinctor</i> (ST2321)	<i>C. tinctor</i> (SUT161)	C. tinctor (SUT260)	C. selangorensis (KS15)
C. tinctor (AJ390421)	1.000	0.909	0.890	0.881	0.700
C. tinctor (ST2321)		1.000	0.935	0.924	0.711
C. tinctor (SUT161)			1.000	0.986	0.745
C. tinctor (SUT260)				1.000	0.744
C. selangorensis (KS15)					1.000

Table 22. The identity matrix of ITS1-5.8S-ITS2 sequence comparison of Camillea

tinctor and C. selangorensis.

Note: 1.000 means 100% identity.

The ITS1-5.8S-ITS2 sequence comparison of the C. tinctor

(SUT161, SUT260, and ST2321) isolates varied from 88.1% to 98.6% identity which was a result of the insertion and/or deletion of DNA fragments in the ITS1 region. Andersen and Torsten (1997) reported the presence of a DNA motif repeated in tandem or short tandem repeats (STR) of ITS1 sequences, which caused an increased rate of mutation in the ITS1 sequence of these fungi. When using STAR software (Delgrange and Rivals, 2004), ITS1 sequences of different *C. tinctor* isolates examined exhibited eleven nucleotides repeated in tandem from three to five times (Figure 85). There were eight variation patterns of the STR motif found in the isolates of *C. tinctor* and *C. selangorensis* as shown in Table 23. All of these modifications were followed the basic motif 5' CTACCCTGTAG 3' as reported by Platas *et al.* (2001).

**Table 23.** Short tandem repeat motifs found in the isolates of *Camillea tinctor* and *C.selangorensis* (KS15). The gray characters are mutation point changedfrom the basic motif.

No.	STR motif (5' to 3')
1	CTACCCTGTAG
2	CTACCCTGGAG
3	CTACCCGGTAG
4	CTACCCTACAG
5	TTACCCTGGAG
6	TTACCTGTAAG
7	GTACCCCGGAG
8	GCACCCTACAG

The mechanisms of evolution of repetitive sequences are assumed to be shaped by both intra- or inter-strand recombinational effects such as unequal crossing over, or other mechanisms involving failures in the replication of the DNA such as slipped-strand mispairing (SSM) (Levinson and Gutman, 1987) or replication slippage (Pinder *et al.*, 1998). The SSM is a process in which misalignment intermediates are formed during DNA synthesis or recombination, as a result of the slippage of DNA strands in regions containing repeated nucleotides, or repeated sequences. This phenomenon causes short deletions or insertions and duplications (Levinson and Gutman, 1987). These might be the reason for genetic variation among species of *C. tinctor* (SUT161, SUT260, and ST2321) which presented the different numbers and patterns of STR motifs within the ITS1 sequences. Although ITS sequence analysis of *C. tinctor* (SUT161, SUT260, and ST2321) varied from 88.1% to 98.6%, they were placed in the same species. In addition, their morphological characteristics absolutely matched *C. tinctor* (Berk.) Læssøe, J.D. Rogers & Whalley described by Læssøe *et al.* (1989). *Camillea* 

*selangorensis* (KS15) also contained three repeated motifs in subsequently different patterns. The phylogenetic trees of *C. tinctor* and *C. selangorensis* were constructed based on ITS1-5.8S-ITS2 sequences using the neighbour-joining method by the PHYLIP program (Felsenstein, 1995), and the maximum parsimony method by PAUP program (Swofford, 2000) as shown in Figure 86 and Appendix 2D respectively. The trees exhibited the separation of the two species, *C. selangorensis* and *C. tinctor*, from each other with high bootstrap support. The three isolates of *C. tinctor* (SUT161, SUT260, and ST2321) examined were grouped together with the two *C. tinctor* sequences (AJ390421 and AJ390422) from the GenBank database although they exhibited genetic variation within their ITS1 regions.



**Figure 86.** Phylogenetic tree of *Camillea* based on ITS1-5.8S-ITS2 sequences constructed by using the neighbour-joining method. *Daldinia eschscholzii* is the outgroup. Branch lengths are scaled in terms of expected numbers of nucleotide substitution per site. Numbers on branches are bootstrap values from 1,000 replications.

#### 4.4.3 Group III: Daldinia

*Daldinia eschscholzii* and *D. concentrica* have been classified into five new species by Stadler *et al.* (2004) using anamorph characteristics and perispore ornamentation by SEM but there were still some serious limitations for uncultured specimens or failure of anamorph production in culture. The ITS1-5.8S-ITS2 sequence of *Daldinia eschscholzii* and *D. concentrica* were then investigated.

## 4.4.3.1 Genomic DNA extraction and ITS1-5.8S-ITS2 amplification

Genomic DNA of seven representatives of *D. eschscholzii* isolates were extracted from their mycelia. Two cultural isolates of *D. concentrica* (L1 and L2) obtained from Prof. Anthony J.S. Whalley were also included. Initially, the ITS1-5.8S-ITS2 amplification products most *Daldinia* isolates could not be achieved. It might be because of some inhibitors in DNA extracts which had brown to dark brown in colour corresponding to the fungus mycelia. Then, the DNA solutions were diluted into 1:50, 1:100, 1:200, 1:500, and 1:1,000 (v/v) before amplification. The fragments obtained, approximately 500 bp, were successfully amplified from ITS1-5.8S-ITS2 regions as shown in Figure 87.

## 4.4.3.2 ITS1-5.8S-ITS2 sequence analysis

The amplified ITS1-5.8S-ITS2 fragments of *D. eschscholzii* and *D. concentrica* were sequenced. The length of ITS1-5.8S-ITS2 sequences obtained is concluded in Table 24. The length of ITS1-5.8S-ITS2 sequences of all *D. eschscholzii* isolates was similar, ranging from 479 to 482 bp in size, whilst two isolates of *D. concentrica* (L1 and L2) were 499 bp. The comparison of ITS1-5.8S-ITS2 sequences revealed 95.4% to 99.7% identity within isolates of *D. eschscholzii*  examined but there was 100% identity within the *D. concentrica* specimens (Table 4, Appendix C).



Figure 87. Gel electrophoresis of ITS1-5.8S-ITS2 fragments of *Daldinia* using ITS5 and ITS4 primers. Lanes: M, DNA marker (100 bp DNA ladder, Invitrogen); 1, *Daldinia eschscholzii* (SUT038); 2, *D. eschscholzii* (SUT039); 3, *D. eschscholzii* (SUT168); 4, *D. eschscholzii* (SUT169); 5, *D. eschscholzii* (SUT178); 6, *D. eschscholzii* (SUT209); 7, *D. eschscholzii* (SUT278); 8, *D. concentrica* (L1); and 9, *D. concentrica* (L2).

Species	Location/Source	ITS1	<b>5.8</b> S	ITS2	Total	GenBank
		(bp)	(bp)	(bp)	(bp)	accession
						number
D. eschschozii (SUT039)	Ratchaburi Province	164	155	162	481	DQ322084
D. eschschozii (SUT085)	Yasothorn Province	164	155	162	481	DQ322085
D. eschschozii (SUT168)	Yasothorn Province	164	155	163	482	DQ322086
D. eschschozii (SUT178)	Nakhon Ratchasima	164	155	163	482	DQ322087
	Province					
D. eschschozii (SUT209)	Trad Province	163	155	163	481	DQ322088
D. eschschozii (SUT278)	Kanchanaburi	163	155	159	477	DQ322089
	Province					
D. eschschozii (SUT322)	Chiang Rai Province	164	155	164	483	DQ322090
D. concentrica (L1)	Liverpool John	173	155	171	499	DQ322091
	Moores University*					
D. concentica (L2)	Liverpool John	173	155	171	499	DQ322092
	Moores University*					

**Table 24.** The length of ITS1-5.8S-ITS2 sequences of Daldinia eschscholzii and D.

concentrica obtained from DNA sequence analysis.

\* The specimen was provided by Prof. Anthony J.S. Whalley, Liverpool John Moores University, U.K.

The phylogenetic trees of *D. eschscholzii* and *D. concentrica* were constructed based on ITS1-5.8S-ITS2 sequences using the neighbour-joining method by PHYLIP program (Felsenstein, 1995) and the maximum parsimony method by PAUP program (Swofford, 2000) as shown in Figure 88 and Appendix 3D, respectively. The ITS1-5.8S-ITS2 sequences of *Daldinia* species from GenBank database were also included in the phylogenetic tree construction. The trees contained three major clades.



**Figure 88.** Phylogenetic tree of *Daldinia* species based on ITS1-5.8S-ITS2 sequences using the neighbour-joining method. *Camillea tinctor* is the outgroup. Branch lengths are scaled in terms of expected numbers of nucleotide substitution per site. Numbers on branches are bootstrap values from 1,000 replication.

Clade I consisted of D. grandis (AF176982), Daldinia sp.

(AY315403), D. loculata (AF176969, AF176968, AF176967), D. petrinia (AF176975, AF176974, AF176973), D. vericosa (AF163022), D. fissa (AF176981), Daldinia sp. (AF163023), and D. concentrica (AF163021). Clade II consisted of nine D. concentrica (AF176958, AY616681, AY616682, AY616684, AGTS305, AGTS306, AGTS396, L1, and L2). Clade III contained all the D. eschscholzii examined except for only one sequence of D. concentrica (AY616682), which was included. The sequences of D. eschscholzii examined were all grouped together in clade III including D. eschscholzii (AGTS384 and AY616684) from the GenBank database. This result confirmed that D. eschscholzii is a common species found in Thailand. Although D. concentrica (AY616682) was placed in the same clade as D. eschscholzii, it might be caused by genetic variation within the species or belonging to different species because D. concentrica has been separated into different new species as previously described by Stadler et al. (2004). In addition, another sequence of D. concentrica (AF163021) was placed in clade I with high bootstrap support, and this might be the same reasons as mentioned above. Most D. concentrica sequences from database were placed in clade II and these included two specimens examined, D. concentrica (L1 and L2).

### 4.4.4 Group IV: Hypoxylon

Since *Hypoxylon* is a large and complex genus with the high variation in morphological characteristics, several species could not be identified. The nucleic acid methodology was then applied to clarify species problem.

#### 4.4.4.1 Hypoxylon section Annulata

Thirty five collections belonging to nine species of *Hypoxylon* sect. *Annulata* were investigated for their nucleic acid sequences. Twelve isolates provided by Dr. Surang Thienhirun, and two specimens provided by Dr. Ju-Ming Yu as reference strains were also included. Genomic DNA was extracted from either cultural mycelia or ascospores depending on the culturable specimens. In case of coloured DNA solution, it was diluted as previously described in section 4.4.3.1. The length of amplified ITS1-5.8S-ITS2 fragments ranged from 500 to 1,000 bp as shown in Figure 89. The amplified ITS1-5.8S-ITS2 fragments were sequenced. The length of ITS1-5.8S-ITS2 sequences is listed in Table 25.

The ITS1-5.8S-ITS2 sequences of *Hypoxylon* sect. *Annulata* examined varied in length from 525 to 906 bp. This high variation resulted found in the ITS1 region, which ranged from 157 to 588 bp. The ITS2 sequences ranged from 154 to 170 bp whilst 5.8S sequences were highly constant at 155 bp. The extremely long ITS1 regions (716 to 906 bp) were found in four collections of *H. atroroseum* (SUT009 and SUT010), *Hypoxylon* sp. (ST2336), *Hypoxylon* taxonomic species 1 (SUT236, SUT242, SUT251, and SUT285), *H. stygium* (SUT058, SUT231, and SUT243) and *H. urceolatum* (SUT098). The extremely long ITS1 sequences of *H. atroroseum* (SUT009 and SUT010) and *H. stygium* (SUT058, SUT231, and SUT243) were similar to *H. atroroseum* (AJ390397) and *H. stygium* (AJ390409) respectively. Both *H. atroroseum* (AJ390397) and *H. stygium* (AJ390409) were available from the GenBank database as previously reported by Sánchez-Ballesteros *et al.* (2000). Moreover, *Hypoxylon* sp. (ST2336), *Hypoxylon* taxonomic species 1 (SUT236,

SUT242, SUT251, and SUT285), and *H. urceolatum* (SUT098), which had the long ITS1 sequences, were found in this study.



Figure 89. Gel electrophoresis of ITS1-5.8S-ITS2 fragments of Hypoxylon sect. Annulata using ITS5 and ITS4 primers. Lanes: M, DNA marker (100 bp DNA ladder, Invitrogen); 1, Hypoxylon nitens (ST2313); 2, H. stygium;
3, H. purpureonitens (SUT001); 4, H. atroroseum (SUT009); 5, H. purpureonitens (SUT005); 6, H. atroroseum (SUT010); 7, H. nitens (SUT081); 8, H. moriforme (SUT220); 9, H. nitens (244); 10, H. bovei var. microspora (SUT025); 11, H. cf. archeri (SUT103); and 12, H. purpureonitens (SUT262).

# Table 25. The length of ITS1-5.8S-ITS2 sequences of Hypoxylon sect. Annulata

Species	Location/Source	ITS1	<b>5.8</b> S	ITS2	Total	GenBank
		(bp)	(bp)	(bp)	(bp)	accession
	a 111 b 1	• • • •				number
H. cf. archeri (SUT103)	Songkhla Province	209	155	161	525	DQ201121
H. cf. archeri (SUT105)	Songkhla Province	209	155	161	525	DQ201122
<i>H.</i> cf. <i>archeri</i> (ST2333)	RFD <sup>a</sup>	209	155	161	525	DQ201123
<i>H</i> . cf. <i>archeri</i> (ST2527)	RFD <sup>a</sup>	224	155	160	539	DQ201124
H. atroroseum (SUT009)	Nakhon Ratchasima Province	506	155	164	825	DQ223733
H. atroroseum	Nakhon Ratchasima	506	155	164	825	DQ223734
(SUT010)	Province					
<i>H. bovei</i> var.	Chaiyaphum Province	202	155	170	527	DQ322096
microspora (SUT025)						
<i>H. bovei</i> var.	The University of	226	155	167	548	DQ201127
microspora (Ju2)	Taiwan <sup>b</sup>					
H. bovei var.	<b>RFD</b> <sup>a</sup>	226	155	167	548	DQ201129
microspora (ST2579)						
H. bovei var.	$RFD^{a}$	225	155	167	547	DQ201128
microspora (ST2406)						-
H. leptascum var.	<b>RFD</b> <sup>a</sup>	248	155	159	562	DQ322097
macrospora (ST2584)						
H. moriforme (SUT220)	Trad Province	230	155	165	550	DQ322129
H. nitens (Jul)	The University of	158	155	166	479	DO223750
~ ,	Taiwan <sup>b</sup>					
H. nitens (ST2313)	$RFD^{a}$	158	155	166	479	DO223751
H. nitens (ST2332)	$RFD^{a}$	233	155	166	554	DO322098
H. nitens (ST2436)	<b>RFD</b> <sup>a</sup>	158	155	166	479	DQ322099
H. nitens (ST2473)	$RFD^{a}$	158	155	166	479	DO223752
H. purpureonitens	Nakhon Ratchasima	225	155	166	546	DO322100
(SUT001)	Province					
H. purpureonitens	Nakhon Ratchasima	225	155	169	549	DQ223753
(SUT004)	Province					
H. purpureonitens	Nakhon Ratchasima	225	155	165	545	DO322101
(SUT005)	Province					
H. purpureonitens	Yasothorn Province	225	155	169	549	DO223754
(SUT167)						
H. purpureonitens	Trad Province	225	155	169	549	DO223755
(SUT262)						
H. purpureonitens	$RFD^{a}$	225	155	169	549	DO223756
(ST2448)				- • /	• • •	- (
H. purpureonitens	<b>RFD</b> <sup>a</sup>	225	155	169	549	DO223757
(ST2485)		-				
H. stygium (SUT058)	Ratchaburi Province	477	155	164	796	DO223760
H. stygium (SUT231)	Trad Province	588	155	163	906	DQ322102

obtained from DNA sequence analysis.

<sup>a</sup> The culture was provided by Dr. Surang Thienhirun, The Royal Forest Department, Thailand. <sup>b</sup> The specimen was provided by Dr. Ju-Ming Yu, The University of Taiwan, Taiwan.

# Table 25. (Continued).

Species	Location/Source	ITS1 (bp)	5.8S (bp)	ITS2 (bp)	Total (bp)	GenBank accession
						number
H. stygium (SUT243)	Trad Province	477	155	164	796	DQ223761
H. urceolatum (SUT098)	Songkhla Province	398	155	163	716	DQ322103
Hypoxylon sp. (ST2336)	$RFD^{a}$	176	155	154	489	DQ322104
Hypoxylon taxonomic	Nakhon Ratchasima	560	155	155	870	DQ322105
species 1 (SUT081)	Province					
<i>Hypoxylon</i> taxonomic species 1 (SUT236)	Trad Province	566	155	155	876	DQ322106
Hypoxylon taxonomic species 1 (SUT242)	Trad Province	566	155	155	876	DQ322107
Hypoxylon taxonomic species 1 (SUT244)	Trad Province	566	155	154	875	DQ322108
Hypoxylon taxonomic species 1 (SUT251)	Trad Province	566	155	155	876	DQ322109
Hypoxylon taxonomic species 1 (SUT285)	Kanchanaburi Province	566	155	154	875	DQ322110

<sup>a</sup> The culture was provided by Dr. Surang Thienhirun, The Royal Forest Department, Thailand.

<sup>b</sup> The specimen was provided by Dr. Ju-Ming Yu, The University of Taiwan, Taiwan.

#### A) ITS1-5.8S-ITS2 sequence analysis

Since there was a high variation in size of ITS1-5.8S-ITS2 region (476 bp to 906 bp), they could not be unambiguously aligned correctly. For sequence alignment, *Hypoxylon* sect. *Annulata* could be divided into 2 groups according to their ITS1-5.8S-ITS2 fragment sizes, which were 716 bp to 906 bp (group I) and 476 bp to 566 bp (group II) respectively. The group I composed of *H. atroroseum*, *H. stygium*, *H.* cf. *stygium* (SUT231), *Hypoxylon* taxonomic species 1, and *H. urceolatum* (SUT098). After their sequences were aligned with reference strains, *H. stygium* (AJ390409) and *H. atroroseum* (AJ390397), available from the GenBank database, the phylogenetic trees were constructed using both the neighbourjoining method (PHYLIP program; Felsenstein, 1995), and the maximum parsimony (PAUP program; Swofford, 2000). Similar patterns of phylogenetic trees were obtained from the two analysis methods (Figure 90 and Appendix 4D). The trees composed of two clades. Clade I consisted of *H. atroroseum* (SUT009, SUT010, and AJ390397), *H. stygium* (SUT058, SUT243, and AJ390409), *H. cf. stygium* (SUT231), and *H. urceolatum* (SUT098), whilst clade II consisted of only one species, *Hypoxylon* taxonomic species 1 (SUT081, SUT242, SUT244, SUT251, and SUT285).

and *H. atroroseum* (AJ390397) from the GenBank database were grouped together. But the two isolates of *H. stygium* (SUT058 and SUT243) examined were in another group with *H. stygium* (AJ390397) from the GenBank database. *Hypoxylon atroroseum* and *H. stygium* appeared to be closely related as shown by their 94 % identity.

Two isolates of *H. atroroseum* (SUT009 and SUT010)

The ITS1-5.8S-ITS2 sequence alignments of *H. atroroseum* (SUT009, SUT010, and AJ390397) and *H. stygium* (SUT058, SUT243, and AJ390397) revealed the insertion and/or deletion sequences of 28 bp (5' ATCTG CTCGAATAAAATTGCTTCAATAT 3') within the ITS1 region. This sequence fragment might be useful for the designer of a probes or markers for species specific detection. This molecular result was in agreement with their closely related morphological characteristics except that the stromata of *H. atroroseum* often have rosy surface tones and the conidiogenous structure of *H. atroroseum* is *Nodulisporium*-like, whereas that of *H. stygium* is *Periconiella*-like (Ju and Rogers, 1996). However, *H. atroroseum* and *H. stygium* were separated from each other with high 100% bootstrap support based on ITS1-5.8S-ITS2 sequence analysis.



Figure 90. Phylogenetic tree of *Hypoxylon* sect. *Annulata* containing extremely long ITS1 region (398 bp to 588 bp) based on ITS1-5.8S-ITS2 sequences using the neighbour-joining method. *Camillea tinctor* is the outgroup. Branch lengths are scaled in terms of expected numbers of nucleotide substitution per site. Numbers on branches are bootstrap values from 1,000 replications.

In addition H. cf. stygium (SUT231) contained the largest

ITS1 region, 588 bp, and was branched from *H. stygium* but still showed close relationship to both taxa. *Hypoxylon* cf. *stygium* (SUT231) differed from *H. stygium* in stromatal form. *Hypoxylon* cf. *stygium* (SUT231) had conspicuous perithecial mounds and broad size of ostiolar disc (0.1-0.3 mm diameter). This might be genetic variation within the species or it might indicate a distinct taxon.

*Hypoxylon urceolatum* (SUT098) was separately branched from *H. atroroseum* (SUT009, SUT010, and AJ390397), *H. stygium* (SUT058, SUT243, and AJ390397), and *H.* cf. *stygium* (SUT231). In addition, *Hypoxylon urceolatum* has the distinctive characteristic of KOH-extractable pigment in purplish (Ju and Rogers, 1996). However, all taxa in clade I had a convex *truncatum*-type of ostiolar disc and small in the size of their ostiolar disc.

Clade II consisted of only one taxon, *Hypoxylon* taxonomic species 1 (SUT081, SUT242, SUT244, SUT251, and SUT285), which had a *bovei*-type of ostiolar disc. Although its morphological characteristics were close to both *H. nitens* and *H. bovei* var. *microspora*, the size of ITS1-5.8S-ITS2 region was absolutely different from those two taxa.

The group II (476 to 566 bp) composed of *H.* cf. *archeri* (SUT103, SUT105, ST2333, and ST2527), *H. bovei* var. *microspora* (SUT025, ST2406, ST2579, and Ju2), *H. leptascum* var. *macrosporum* (ST2584), *H. moriforme* (SUT220 and ST2332), *H. nitens* (ST2313, ST2436, ST2473, and Ju1), and *H. purpureonitens* (SUT001, SUT004, SUT005, SUT167, SUT262, ST2448, and ST2485). Sequence alignment of ITS1-5.8S-ITS2 was performed. The phylogenetic trees were constructed by using both the neighbour-joining method (PHYLIP)

program, Felsenstein, 1995), and the maximum parsimony method (PAUP program, Swofford, 2000). Both methods also exhibited the similar phylogenetic tree except for bootstrap valves. The trees were divided into three clades (Figure 91 and Appendix 5D).

Clade I consisted of *Hypoxylon* sp. (ST2336), which was

separated from other taxa. Clade II consisted of seven species, *H. annulatum* (AJ390395), *H. bovei* var. *microspora* (SUT025, ST2406, ST2579, and Ju2), *H. cohaerens* (AJ390406), *H. leptascum* var. *macrosporum* (ST2584), *H. moriforme* (SUT220 and ST2332), *H. nitens* (ST2313, ST2436, ST2473, and Ju1), and *H. purpureonitens* (SUT001, SUT004, SUT005, SUT167, SUT262, ST2448, and ST2485), which were clearly separated from each other. These isolates of *H. nitens* (ST2313, ST2436, and ST2473) were grouped together including a reference strain, *H. nitens* (Ju1). *Hypoxylon bovei* var. *microspora* (ST2406, ST2579, and SUT025) isolates were also clustered with a reference strain, *H. bovei* var. *microspora* (Ju2), although some variation within the species appeared to represent a distinctive taxon. *Hypoxylon bovei* var. *microspora* was placed close to *H. moriforme* (SUT220 and ST2332), and they were similar in their morphological characteristics except for the type and size of their ostiolar disc.

All of *H. purpureonitens* isolates (SUT001, SUT004, SUT005, SUT167, SUT262, ST2448, and ST2485) were grouped together and showed slightly variation among collections. Although the morphological characteristics of *H. purpureonitens* and *H. nitens* are very similar except for having purplish KOH-extractable pigments in *H. purpureonitens* (Ju and Rogers, 1996). They were completely different in molecular data.



Figure 91. Phylogenetic tree of *Hypoxylon* sect. *Annulata* based on ITS1-5.8S-ITS2 sequences using the neighbour-joining method. *Camillea tinctor* (AJ394021) is the outgroup. Branch lengths are scaled in terms of expected numbers of nucleotide substitution per site. Numbers on branches are bootstrap values from 1,000 replications.

Clade III contained only one species of *H*. cf. *archari* (SUT103, SUT105, ST2333, and ST2527). The morphological features of this taxon were similar to those of *H*. *archeri* Berk. and *H*. *michelianum* Ces. & De Not. Ascospore dimensions, stromatal form, and coloration were indicative of *H*. *archeri* but the distinctive white fringe surrounding the ostioles was reminiscent of *H*. *michelianum*. This taxon remains unknown.

#### **B) ITS2 sequence analysis**

Although the length of the ITS1 region of *Hypoxylon* sect. *Annulata* differed considerably (ranging from 157 to 588 bp), the length of ITS2 region was not so different (ranging from 147 to 170 bp). Therefore, ITS2 sequences of all *Hypoxylon* sect. *Annulata* taxa were aligned together, and the phylogenetic trees were constructed using both the neighbour-joining method (PHYLIP program, Felsenstein, 1995), and the maximum parsimony method (PAUP program, Swofford, 2000). Both methods exhibited the same pattern of trees except for their bootstrap valves. The trees contained three main clades (Figure 92 and Appendix 6D).

Clade I was a complex clade consisting of seven species, *H. nitens* (ST2313, ST2436, ST2473, and Ju1), *H. atroroseum* (SUT009, SUT010, AJ390397), *H. stygium* (SUT058, SUT243, and AJ390409), *H. cf. stygium* (SUT231), *H. leptascum* var. *microsporum* (ST2548), *H. urceolatum* (SUT098) and *Hypoxylon* taxonomic species 1 (SUT081, SUT242, SUT244, SUT251, and SUT285). Four specimens of *H. nitens* (ST2313, ST2436, ST2473, and Ju1) were grouped together in the same cluster, which was clearly separated from *Hypoxylon* taxonomic species 1 (SUT081, SUT242, SUT244, SUT251, and SUT285).


Figure 92. Phylogenetic tree of *Hypoxylon* sect. *Annulata* based on ITS2 sequences using the neighbour-joining method. *Camillea tinctor* is the outgroup. Branch lengths are scaled in terms of expected numbers of nucleotide substitution per site. Numbers on branches are bootstrap values from 1,000 replications.

*Hypoxylon atroroseum* (SUT009, SUT010, and AJ390397), *H. stygium* (SUT058, SUT243, and AJ390409), and *H. cf. stygium* (SUT231) exhibited a similar pattern on ITS1-5.8S-ITS2 sequence analysis (Figure 90) and ITS2 sequence analysis (Figure 92). Thus, this result confirmed the close relationship among these taxa.

### Hypoxylon leptascum var. microsporum (ST2548) and H.

*urceolatum* (SUT098) were grouped together in the same cluster. Their morphological characters are also similar in convex *truncatum*-type of ostiolar disc, and overlap in ascospore size, but they differ in their KOH-extractable pigments (Ju and Rogers, 1996). *Hypoxylon leptascum* var. *microsporum* (ST2548) was greenish olivaceous whilst *H. urceolatum* (SUT098) was vinaceous purple or vinaceous grey.

Clade II consisted of six species including *H. cohaerens* (AJ390406), *H. bovei* var. *microspora* (SUT025, ST2406, ST2579, and Ju2), *H. purpureonitens* (SUT001, SUT004, SUT005, SUT167, SUT262, ST2448, and ST2485), *Hypoxylon* sp. (ST2336), *H. annulatum* (AJ390395), and *H. moriforme* (SUT220 and ST2332). The pattern of phylogenetic trees in clade II was also similar to the tree constructed from ITS1-5.8S-ITS2 sequence (Figure 91), which confirmed the close relationships within this clade except that *H. moriforme* (ST2336) and *H. annulatum* (AJ390395) placed as sister branch.

*Hypoxylon* sp. (ST2336) was placed in the same cluster as *H. annulatum* (AJ390395), which indicated a close relationship for both species. The morphological characters of *Hypoxylon* sp. (ST2336), which are usually has glomerate stromata, can look quite like *H. annulatum* when the stromata are hemispherical. The usually evident perithecial mounds and less massive stromata are

useful in identifying *H. moriforme*. Moreover, *H. annulatum* is a Northern temperate species, and is highly associated with *Quercus* (Ju and Rogers, 1996).

Clade III contained only *H.* cf. *archari* (SUT103, SUT105, ST2333, and ST2527) isolates (Figure 91).

From molecular analysis of ITS1-5.8S-ITS2 sequences and ITS2 sequence revealed the similarity of phylogenetic relationships based on both methods of the neighbour-joining and maximum parsimony. The trees exhibited a clear separation of the species complex within *Hypoxylon* sect. *Annulata*, and also indicated the range of genetic variation within each species (Figures 90, 91 and 92). Moreover, some taxa might prove to be the new taxa when further collections are made and examined.

### 4.4.4.2 Hypoxylon section Hypoxylon

Forty four representatives of *Hypoxylon* collections belonging to twenty one species were extracted for genomic DNA from either cultural mycelium or ascospores depending on the specimens. The amplified ITS1-5.8S-ITS2 fragments were approximately 500 to 600 bp (Figure 93). They were then sequenced. The sizes of ITS1-5.8S-ITS2 fragments are listed in Table 26.



Figure 93. Gel electrophoresis of ITS1-5.8S-ITS2 fragments of *Hypoxylon* sect. *Hypoxylon* using ITS5 and ITS4 primers. Lanes: M, DNA marker (100 bp DNA ladder, Invitrogen); 1, *Hypoxylon investiens* (SUT041); 2, *H. investiens* (SUT063); 3, *H. lenormandii* (SUT046); 4, *H. lenormandii* (SUT180); 5, *H. hypomiltum* (SUT166); 6, *H. cf. perforatum* (SUT294); 7, *H. rubiginosum* (SUT146); 8, *H. kanchanapisekii* sp. nov. (SUT066); and 9, *H. sublenormandii* sp. nov. (SUT282).

Species	Location/Source	ITS1 (bp)	5.8S	ITS2 (bp)	Total (bp)	GenBank
		(0P)	(0P)	(0P)	(66)	number
H. anthochroum (SUT233)	Trad Province	180	155	162	497	QD201125
H. anthochroum (SUT240)	Trad Province	180	155	162	497	QD201126
H. brevisporum (SUT256)	Trad Province	165	155	161	481	DQ322111
H. duranii (SUT223)	Trad Province	206	155	184	545	DQ322112
H. cf. ferruginuem (SUT070)	Ratchaburi Province	131	155	159	445	DQ322113
H. cf. fendleri (SUT061)	Ratchaburi Province	181	155	163	499	QD201130

 Table 26. The length of ITS1-5.8S-ITS2 sequences of Hypoxylon sect. Hypoxylon obtained from DNA sequence analysis.

\* The culture was provided by Dr. Surang Thienhirun, The Royal Forest Department, Thailand.

## Table 26. (Continued).

Species	Location/Source	ITS1 (bp)	5.8S (bp)	ITS2 (bp)	Total (bp)	GenBank accession
		(~P)	( <b>°</b> P)	(5)	(~P)	number
H. cf. fendleri (SUT120)	Petchaboon Province	185	155	161	501	QD201131
H. cf. fendleri (SUT159)	Yasothorn Province	183	155	164	502	QD201132
H. cf. fendleri (SUT162)	Yasothorn Province	183	155	165	503	QD223735
H. cf. fendleri (SUT165)	Yasothorn Province	182	155	163	500	QD223736
H.cf. fendleri (SUT280)	Kanchanaburi Province	156	155	163	474	QD223737
H. haematostroma (SUT164)	Yasothorn Province	176	155	161	492	QD223738
H. haematostroma (SUT292)	Kanchanaburi Province	176	155	161	492	QD223739
H. haematostroma (SUT293)	Kanchanaburi Province	176	155	161	492	QD223740
H. hypomiltum (SUT166)	Yasothorn Province	213	155	160	528	QD322114
H. investiens (SUT041)	Ratchaburi Province	276	155	153	584	QD322115
H. investiens (SUT063)	Ratchaburi Province	230	155	155	540	QD322116
<i>H. kanchanapisekii</i> (SUT066) sp. nov.	Ratchaburi Province	209	155	162	526	QD223741
<i>H. kanchanapisekii</i> (SUT068) sp. nov.	Ratchaburi Province	209	155	162	526	QD223742
<i>H. kanchanapisekii</i> (SUT069) sp. nov.	Ratchaburi Province	209	155	162	526	QD223743
H. lenormandii (SUT016)	<b>Burirum Province</b>	188	155	160	503	QD223744
H. lenormandii (SUT046)	Ratchaburi Province	208	155	165	528	DQ322117
H. lenormandii (SUT180)	Nakhon Ratchasima Province	188	155	160	503	QD223745
H. lenormandii (ST2324)	RFD*	188	155	160	503	QD223746
H. monticulosum (SUT042)	Ratchaburi Province	171	155	165	491	QD223747
H. monticulosum (SUT080)	Nakhon Ratchasima Province	171	155	165	491	QD223748
H. monticulosum (SUT116)	Songkhla Province	171	155	165	491	QD223749
<i>H</i> . cf. <i>perforatum</i> (SUT020)	Burirum Province	301	155	156	612	QD322118
<i>H</i> . cf. <i>perforatum</i> (SUT218)	Trad Province	206	155	186	547	QD322119
<i>H.</i> cf. <i>perforatum</i> (SUT294)	Kanchanaburi Province	209	155	161	525	QD322120
H. rubiginosum (SUT215)	Trad Province	178	155	164	497	QD223758
H. rubiginosum (SUT221)	Trad Province	178	155	164	497	QD223759
H. subgilvum var. microsporum (SUT104)	Songkhla Province	148	155	164	467	QD322121

\* The culture was provided by Dr. Surang Thienhirun, The Royal Forest Department, Thailand.

## Table 26. (Continued).

Species	Location/Source	ITS1	<b>5.8</b> S	ITS2	Total	GenBank
		(bp)	(bp)	(bp)	(bp)	accession
						number
H. subgilvum var. microsporum (SUT108)	Songkhla Province	149	155	153	457	QD322122
<i>H. sublenormandii</i> sp. nov. (SUT250)	Trad Province	198	155	161	514	QD223762
<i>H. sublenormandii</i> sp. nov. (SUT282)	Kanchanaburi Province	198	155	161	514	QD223763
<i>H. suranareei</i> (SUT183) sp. nov.	Nakhon Ratchasima Province	199	155	162	516	QD223764
H. trugodes(SUT146)	Nakhon Ratchasima Province	182	155	168	505	QD322123
H. trugodes (SUT148)	Nakhon Ratchasima Province	181	155	152	488	QD322124
H. trogodes (SUT187)	Nakhon Ratchasima Province	181	155	158	494	QD322125
<i>Hypoxylon</i> taxonomic species 2 (SUT082)	Nakhon Ratchasima Province	130	155	166	451	QD322126
<i>Hypoxylon</i> taxonomic species 3 (SUT158)	Yasothorn Province	173	155	160	488	QD322127
<i>Hypoxylon</i> taxonomic species 4 (SUT237)	Trad Province	131	155	158	444	QD322128

\* The culture was provided by Dr. Surang Thienhirun, The Royal Forest Department, Thailand.

The length of ITS1-5.8S-ITS2 sequences of *Hypoxylon* sect. *Hypoxylon* ranged from 445 to 612 bp. The ITS1 sequences revealed the highest variation ranging from 131 to 209 bp. The ITS2 sequences ranged from 152 to 184 bp, whilst 5.8S sequences were highly constant at 155 bp. The sequences of two isolates, *H.* cf. *ferrugineum* (SUT017) and *H. macrocapum* (SUT045), could not be achieved. This might be because of either the variation of ITS1-5.8S-ITS2 sequences within the species or the direct sequencing of the amplified ITS1-5.8S-ITS2 fragments, which some fragments might contain a mutation and/or be GC rich. Therefore, their sequences could not be clearly performed.

The ITS1-5.8S-ITS2 sequences of *Hypoxylon* sect. *Hypoxylon* examined were aligned and their phylogenetic trees were constructed by using the neighbour-joining method by the PHYLIP program (Felsenstein, 1995), and the maximum parsimony method by the PAUP program (Swofford, 2000) (Figure 94 and Appendix 7D). The ITS1-5.8S-ITS2 sequences of related *Hypoxylon* species from the GenBank database, *H. fendleri* (AJ390400) and *H. perforatum* (AJ390407), were also included. The phylogenetic trees show four main clades.

Clade I contained five species which were *H. subgivum* var. *microsporum* (SUT108), *H. trugodes* (SUT148 and SUT187), *Hypoxylon* taxonomic species 2 (SUT082), *H.* cf. *ferruginium* (SUT070 and SUT237), *H. brevisporum* (SUT256). The stromatal surface colour of all species in this clade was mostly reddish brown to brownish vinaceous, and the KOH-extractable pigment was of the orange series. The phylogenetic tree showed that *H. subgivum* var. *microsporum* (SUT108) was distinctive and separated from other species in the same clade. Moreover, its ascospores were smaller than other species and it also had very orange pigment in KOH extraction.

Two isolates of *H*. cf. *ferrugineum* (SUT070 and SUT237) were grouped together and were placed as a sister branch of *Hypoxylon* taxonomic species 2 (SUT082). They had the same stromatal colour and KOH-extractable colour but they were different in ascospore size. The tree showed the closely relationship between both species.



**Figure 94.** Phylogenetic tree of *Hypoxylon* sect. *Hypoxylon* based on ITS1-5.8S-ITS2 sequences using the neighbour-joining method. *Camillea tinctor* is the outgroup. Branch lengths are scaled in terms of expected numbers of nucleotide substitution per site. Numbers on branches are bootstrap values from 1,000 replications.

Clade II consisted of four species, *H. monticulosum* (SUT042, SUT080, and SUT116), *H. lenormandii* (SUT016, SUT180, and ST2324), *H. sublenormandii* sp. nov. (SUT046, SUT066, SUT068, and SUT069) and *H.* cf. *fendleri* (SUT061, SUT120, SUT159, SUT162, SUT165, and SUT280).

Hypoxylon monticulosum found in this study could be divided into two different types based on KOH-extractable pigments. Two representatives of *H. monticulosum* isolates, SUT042 and SUT080, were without apparent KOHextractable pigments as detailed by Ju & Rogers (1996) whereas *H. monticulosum* SUT116 had a purple colored extract. This does however agree with Ju & Rogers (1996) who state that "it is noteworthy that the purplish stromatal pigments dark livid to dark vinaceous of *H. monticulosum* and *H. submonticulosum* are easily detected in the young, rusty brown stromata but are hardly so in the mature, blackened stromata". The sequence alignment indicated 99% similarity and it was concluded that they represented the same taxon regardless of extractable pigment in KOH.

Three isolates of *H. lenormandii* (ST2324, SUT016, and SUT180) matched closely the description by Ju & Rogers (1996), and all collections were found on dicotyledonous wood from different forest areas. They were clearly separated from *H. sublenormandii* sp. nov. (SUT250 and SUT282), which occurred on bamboo, with high 100% bootstrap support. They also differed in morphological characters such as spore size, a more reddish brown stromatal surface color, and a straight germ slit (Table 16) and on the basis of this and the sequence data a new species was confirmed.

Initially, *H*. cf. *fendleri* (SUT061, SUT159, SUT162, SUT165, and SUT280) collections had been identified as *H. fendleri* since their

morphological characteristics matched to H. fendleri Berk. ex Cooke (Ju and Rogers 1996). However Ju and Rogers (1996) pointed out that *H. fendleri* and *H. retpela* Van der Gucht & Van der Veken are very similar stating "These two fungi differ mainly in the conspicuousness of their perispore ornamentation". The ornamentation in H. retpela is described as very conspicuous coil-like. However all the Thai collections had similar coiling, which was not noticeably conspicuous. Thus the description for H. fendleri (Ju & Rogers, 1996) was the nearest match. The phylogenetic result showed that all the Thai isolates (SUT061, SUT159, SUT162, SUT165, and SUT280) grouped together and were placed as a sister branch of *H. fendleri* (AJ390400) based on the GenBank database sequence with high bootstrap support. The percentage similarity of H. fendleri (AJ390400) to SUT061, SUT159, SUT162, SUT165, and SUT280 isolates was 85%, 85%, 85%, 85%, and 80% respectively. They are therefore quite different and as a result the Thai collections were recorded as H. cf. fendleri. This might be the result of a wide range of *H. fendleri* descriptions (morphological) or genetic variation within this taxon found in Thailand. More collections of specimens around the world are required for a better understanding of species delimitation and genetic variation within this taxon.

Clade III was a big clade and consisted of eleven species, *H. haematostroma* (SUT164, SUT292, and SUT293), *H. anthochroum* (SUT233 and SUT240), *H. kanchanapisekii* sp. nov. (SUT046, SUT066, SUT068, and SUT069), *H. rubiginosum* (SUT215 and SUT221), *H. investiens* (SUT041 and SUT063), *H. perforatum* (AJ390407), *H. cf. perforatum* (SUT020), *H. cf. perforatum* (SUT294), *H. suranareei* sp. nov. (SUT182), *H. hypomiltum* (SUT166), *Hypoxylon* taxonomic species 3 (SUT158). Three isolates of *H. haematostroma* (SUT164, SUT292, and

SUT293) were separated from other taxa in the same clade because their distinctive teleomorphic characteristics having red or orange red stromatal granules, constantly long tubular perithecia, and large ascospores (Ju and Rogers, 1996).

*Hypoxylon anthochroum, H. kanchanapisekii* sp. nov. (SUT046, SUT066, SUT068, and SUT069), *H. rubiginosum* (SUT215 and SUT221), and *H. investiens* were separated clearly from each other with high bootstrap support. Although *H. anthochroum* was considered to be a synonym of *H. rubiginosum* by Miller (1961), they are different in colour of KOH-extractable pigments. Two isolates of *H. investiens* (SUT041 and SUT063) exhibited genetic variation within the ITS1 region. These might be the appearance of insertion and/or deletion fragments of short repeated nucleotide sequences.

Hypoxylon cf. perforatum (SUT020), H. cf. perforatum (SUT294), and H. perforatum (AJ390407) from GenBank database grouped in the same branch and included H. suranareei sp. nov. (SUT182). Since H. perforatum described by Ju and Rogers (1996) had a wide range of stromatal surface colour, dark brick, grayish sepia, brown vinaceous, or umber, and also perispore ornamentation varied from smooth to inconspicuous coil-like ornamentation. Two isolates of H. cf. perforatum (SUT020) and H. cf. perforatum (SUT294) were different in stromatal surface colour but they were similar in having a smooth perispore. Importantly, both taxa are lacking white substance deposited around the ostioles which is usually found in this species (Ju and Rogers, 1996). The ITS1-5.8S-ITS2 sequence alignment of H. perforatum (SUT020) and H. cf. perforatum (SUT294) respectively, which might indicate different taxa. However, further investigations of more collections are required.

and SUT218). The ascospore size of *H. duranii* was similar to *H. anthochroum* but it differed in having conspicuous coil-like ornamentation of the perispore (Ju and Rogers, 1996), and the molecular data also revealed the different between both species very clearly.

The relationship between *Hypoxylon* sect. *Annulata* and *Hypoxylon* sect. *Hypoxylon* was then analyzed. Representatives of each species from both sections were aligned and the phylogenetic trees were constructed based on ITS2 sequences by using the neighbour-joining method by the PHYLIP program (Felsenstein, 1995), and the maximum parsimony method by the PAUP program (Swofford, 2000) as shown in Figure 95 and Appendix 8D, respectively. The species of *Hypoxylon* sect. *Annulata* appeared as a paraphylectic group with *Hypoxylon* sect. *Hypoxylon* sect. *Annulata* (H. cf. stygium (SUT231), H. atroroseum (SUT009), H. stygium (SUT058), Hypoxylon taxonomic species 1 (SUT081 and SUT285), *H. leptascum* var. macrosporum (ST2584) and *H. urceolatum* (SUT098)) were grouped together. The phylogenetic tree based on ITS2 sequence analysis did not support the concept of the division of *Hypoxylon* into two sections. The other nucleotide regions such as 28S rDNA or IGS may be more suitable candidates.



**Figure 95.** Phylogenetic tree of *Hypoxylon* based on ITS2 sequences using the neighbour-joining method. *Camillea tinctor* is the outgroup. Branch lengths are scaled in terms of expected numbers of nucleotide substitution per site. Numbers on branches are bootstrap values from 1,000 replications.

## 4.4.5 Group V: Xylariaceous endophytes

Since members of *Xylaria* have been reported as common endophytes in many plants and there are serious limitations in species identification because of the lack of their teleomorph stage in culture, the *Xylaria* collections were selected to investigate for their ITS1-5.8S-ITS2 ribosomal nucleotide sequence analysis.

### 4.4.5.1 Genomic DNA extraction and ITS region amplification

Forty representatives of *Xylaria* specimens were extracted for genomic DNA including ten isolates of *Xylaria* obtained from Dr. Surang Thienhirun. Three more isolates, *Biscogniauxia* sp. nov. (SUT290), *Kretzschmaria* sp. (ST2325), and *Nemania* sp. (SUT258) were included. The amplified ITS1-5.8S-ITS2 fragments were approximately 500 to 600 bp (Figure 96).

### 4.4.5.2 ITS1-5.8S-ITS2 sequence analysis

The amplified ITS1-5.8S-ITS2 fragments were sequenced and the size of each specimen examined (Table 27).



Figure 96. Gel electrophoresis of ITS1-5.8S-ITS2 fragments of *Xylaria* using ITS5 and ITS4 primers. Lanes: M, DNA marker (100 base pair DNA ladder, Invitrogen); 1, *Xylaria* sp. nov. (SUT012); 2, *Xylaria* sp. nov. (SUT014); 3, *X. multiplex* (SUT028); 4, *X. badia* (SUT032); 5, *X. badia* (SUT076); 6, *X. mellisii* (SUT074); 7, *X. cf. juruensis* (SUT088); 8, *X. ianthino-velutina* (SUT091); 9, *X. ianthino-velutina* (SUT123); 10, *X. cubensis* (089); 11, *Xylaria* species 2 (SUT130); and 12, *Xylaria* species 2 (SUT195).

### ITS1 **5.8**S ITS2 Total GenBank **Species** Location/Source accession (bp) (bp) (bp) (bp) number X. anisopleura (SUT196) Trad Province 465 155 155 775 DQ322130 *X. anisopleura* (SUT205) Trad Province 141 155 DQ322131 155 451 RFD\* *X. anisopleura* (ST2329) 465 155 155 775 DQ322132 X. cf. apiculata (SUT203) 181 495 Trad Province 155 159 DQ322133 RFD\* X. arbuscula var. 182 155 162 499 DQ322134 microspora (ST2372) X. badia (SUT032) Ratchaburi Province 181 155 158 494 DQ322135 *X. badia* (SUT076) Ratchaburi Province 179 155 494 DQ322136 160 X. badia (SUT142) Nakhon Ratchasima 181 155 159 495 DQ322137 Province RFD\* 181 496 DQ322138 X. badia (ST2417) 155 160 X. beccari (SUT092) Songkhla Province 273 155 155 583 DQ322139 X. brachiata (SUT078) Ratchaburi Province 181 155 163 499 DQ322140 X. cubensis (SUT090) Songkhla Province 179 155 159 493 DQ322141 X. cubensis (ST2027) RFD\* 171 155 165 491 DO322142 X. cubensis (ST2326) RFD\* 188 155 163 506 DQ322143 209 155 *X. curta* (ST2382) RFD\* 162 526 DQ322144 494 X. grammica (ST2348) RFD\* 180 155 159 DQ322145 X. grammica (ST2363) RFD\* 155 490 DQ322146 180 155 X. ianthino-velutina Nakhon Ratchasima 177 155 156 488 DQ322147 Province (SUT123) X. cf. juruensis (SUT088) Songkhla Province 182 155 156 493 DQ322148 Nakhon Ratchasima 155 DQ322149 X. cf. juruensis (SUT140) 181 162 498 Province X. juruensis var. Nakhon Ratchasima 179 155 159 493 DQ322150 Province microspora (SUT129) X. juruensis var. Nakhon Ratchasima 181 155 159 495 DQ322151 Province microspora (SUT138) X. juruensis var. Nakhon Ratchasima 182 155 159 496 DQ322152 Province microspora (SUT139) X. maitlandii (SUT177) Nakhon Ratchasima 181 155 159 495 DQ322153 Province X. multiplex (SUT028) Ratchaburi Province 178 155 162 495 DQ322154 RFD\* X. multiplex (ST2298) 178 155 161 494 DQ322155 X. mellisii (SUT074) Ratchaburi Province 155 155 167 477 DQ322156 X. mellisii (SUT192) Trad Province 184 155 161 500 DQ322157 X. psidii (SUT124) Nakhon Ratchasima 181 155 159 495 DQ322158 Province

Table 27. The length of ITS1-5.8S-ITS2 sequences of different species of Xylaria,

\* The culture was provided by Dr. Surang Thienhirun, The Royal Forest Department, Thailand.

Species	Location/Source	ITS1 (bp)	5.8S (bp)	ITS2 (bp)	Total (bp)	GenBank accession number
X. psidii (SUT125)	Nakhon Ratchasima Province	180	155	160	495	DQ322159
X. schweinitzii (SUT201)	Trad Province	151	155	156	462	DQ322160
X. schweinitzii (ST2349)	RFD*	151	155	156	462	DQ322161
<i>Xylaria</i> species 2 (SUT127)	Nakhon Ratchasima Province	174	155	158	487	DQ322162
<i>Xylaria</i> species 2 (SUT130)	Nakhon Ratchasima Province	174	155	157	486	DQ322163
<i>Xylaria</i> species 2 (SUT195)	Trad Province	174	155	157	486	DQ322164
<i>Xylaria</i> sp. nov. (SUT027)	Ratchaburi Province	178	155	149	482	DQ322165
Xylaria sp. nov. (SUT155)	Yasothorn Province	178	155	149	482	DQ322166
Xylaria sp. nov. (SUT198)	Trad Province	178	155	149	482	DQ322167
Xylaria sp. nov. (SUT200)	Trad Province	178	155	149	482	DQ322168
<i>Xylaria</i> taxonomic species 1 (SUT207)	Trad Province	176	155	155	486	DQ322169
Kretzschmaria sp. (ST2325)	RFD*	176	155	196	527	DQ322093
Nemania sp. (SUT258)	Trad Province	186	155	156	497	DQ322094
Biscogniauxia sp. nov. (SUT290)	Kanchanaburi Province	215	155	148	518	DQ322095

\* The culture was provided by Dr. Surang Thienhirun, The Royal Forest Department, Thailand.

When the amplified ITS1-5.8S-ITS2 fragments (451 to 775

bp) were sequenced, the highest variation was found in the ITS1 region ranging from 151 to 465 bp. But the 5.8S region was highly constant at 155 bp. The ITS2 region ranging from 148 to 165 bp, was slightly different. Two isolates of *X. anisopleura* (SUT196 and ST2329) exhibited extremely long ITS1 region, 465 bp, as found in some species of *Hypoxylon* sect. *Annulata*, which was described in section 4.4.4.1, whereas one isolate of *X. anisopleura* (SUT205) exhibited only 141 bp for its ITS1

region. Therefore, the ITS1-5.8S-ITS2 sequences of three *X. anisopleura* isolates were searched for repeated sequence motif by using STAR software (Delgrange and Rivals, 2004) and no repeated sequences were observed. However, their morphological characteristics were similar except for the stromatal form as shown in the Figure 47. More collections of specimens were required for a better understanding of genetic variation within this taxon.

The phylogenetic trees of *Xylaria* were constructed based on ITS1-5.8S-ITS2 sequences using the neighbour-joining method by the PHYLIP program (Felsenstein, 1995), and the maximum parsimony method by the PAUP (Swofford, 2000). The ITS1-5.8S-ITS2 sequences of related *Xylaria* species from the GenBank database were also included. The tree contained four major clades (Figure 97 and Appendix 9D). Clade I consisted only one taxon *X. beccari* (SUT092), which was separated from other clades. Clade II contained twelve species *X. curta* (ST2382), *X. enteroteuca* (AF163033), *X. cornu-damae* (AF163031), *X. castorea* (AF163030), *X. acuta* (AF163026), *X. longipes* (AF163039), *X. floriana* (AF163034), *X. ianthino-velutina* (SUT123), *Xylaria* taxonomic species 1 (SUT027), *X. badia* (SUT032, SUT076, SUT142, and ST2417), *Xylaria* sp. nov. (SUT027, SUT155, SUT198, SUT200, and ST2298), and *Xylaria* species 2 (SUT127, SUT130, and SUT195). Each species in this clade was separated clearly from each other.



**Figure 97.** Phylogenetic tree of *Xylaria* based on ITS1-5.8S-ITS2 sequences using the neighbour-joining method. *Camillea tinctor* is the outgroup. Branch lengths are scaled in terms of expected numbers of nucleotide substitution per site. Numbers on branches are bootstrap values from 1,000 replications.

247

Clade III consisted of *X. polymorpha* (AF163042), *X. cubensis* (AY787733, SUT090, ST2027, and ST2326). All species in this clade had large stromata and hard tissue. Although three isolates of *X. cubensis* examined were placed as sister branches to *X. cubensis* (AY787733) from the GenBank database they showed high variation within the species. The ascospore size of *X. cubensis* (SUT090), (6.3-)7.5-8.8 x 3.8-5  $\mu$ m, was smaller than *X. cubensis* (Mont.) Fr., 8-10.5 x 4-5  $\mu$ m, described by Rogers and Samuels (1987). It could be a different taxon, *X. cf. cubensis*. This might also be the variation within species. In addition, *X. cubensis* is found in various tropical, subtropical and temperate localities of the world. Thus, it was possible that the *X. cubensis* isolates collected in Thailand might be different from the temperate zone in genetic data.

Clade IV consisted of *X. apiculata* (AF163027), *X.* cf. *apiculata* (SUT203), *X.* cf. *maitandii* (SUT177), *X. bambusicola* (SUT129, SUT138, and SUT139), *X.* cf. *juruensis* (SUT088 and SUT140), *X. arbuscula* var. *microspora* (SUT192 and ST2372), *X. mali* (AF163040), *X. hypoxylon* (AF163037), *X. grammica* (ST2348 and ST2363), *X. arbuscula* (ST and ST), *X. brachiata* (SUT078), *X.* cf. *multiplex* (SUT028), *X. mellisii* (SUT074), and *X. psidii* (SUT124 and SUT125). Three isolates of *X. bambusicola* examined were identical and very close to *X.* cf. *maitlandii* (SUT177) and *X.* cf. *apiculata* (SUT203).

Moreover, the host preference of *Xylaria* seems to be off limited taxonomic value in this analysis.

# 4.4.6 Phylogenetic analysis of xylariaceous fungi based on ITS1-5.8S-ITS2 sequences

The ITS1-5.8S-ITS2 sequence of each species of the Xylariaceae examined was aligned and the phylogenetic trees were constructed by using the neighbour-joining method by the PHYLIP program (Felsenstein, 1995), and the maximum parsimony method by the PAUP (Swofford, 2000) (Figures 98 and Appendix 10D). The tree constructed by the neighbour-joining was divided into three clades.

The species representatives of genera *Nemania*, *Astrocystis*, *Kretzschmaria*, *Rosellinia* were placed in Clade I, which demonstrated the close relationship between those genera.

Three different species of *Biscogniauxia* (SUT290, AF201706, and AJ390411) and *C. tinctor* isolates were grouped together (Figure 98 and Appendix 9D). The relationship of both genera reflected the nature of their bipartite stromata which differentiated them from species of *Hypoxylon* sensu Miller (Miller, 1960) and supported the current concept of *Hypoxylon* sensu Ju and Rogers (Ju and Rogers, 1996). Surprisingly, one species of *C. selangorensis* was placed to clade I. this might be the result of the presence of short repeated sequences in ITS1 region as described previously in section 4.4.2.2. However, more collections in *C. selangorensis* were required to obtain more molecular data, which could be used to explain the reliable relationship of this taxon.



**Figure 98.** Phylogenetic tree of xylariaceous fungi based on ITS1-5.8S-ITS2 sequences using the neighbour-joining method. *Diatrype disciformis* is the outgroup. Branch lengths are scaled in terms of expected numbers of nucleotide substitution per site. Numbers on branches are bootstrap values from 1,000 replications.

Both species of *D. concentrica* and *D. eschscholzii* were in the same group, and placed as a sister branch of the *Hypoxylon* group in clade II. *Daldinia* and *Hypoxylon* were close as previously indicated by Bull (1791) although *Daldinia* had already been separated from *Hypoxylon* on the basis of alternating different stromatal anatomy of ring zones (Ju, Rogers, and San Martín, 1997; Stadler *et al.*, 2004).

Most species of *Hypoxylon* from both sections *Annulata* and *Hypoxylon* were placed in clade II except for *H. subgilvum* var. *microsporum* (SUT108) which was placed in clade III. This result exhibited the strong relationship within this genus and the similar finding for most species in *Xylaria*. They were mainly placed in clade I except for *X. cubensis* (SUT090) and *X. mellisii* (SUT074) which were placed in clade II and III respectively.

Therefore, all of the ITS1-5.8S-ITS2 sequences results and their relationships analyzed by using the phylogenetic trees proved to be valuable for taxonomic investigation from a molecular point of view as well as for developing a DNA sequence database. Additionally, this molecular data would be useful for the designation of specific primers and for the development of specific probes for the detection of species of certain *Xylaria* in environmental situations.

## **CHAPTER V**

## **CONCLUSION AND FUTURE PERSPECTIVE**

Species identification and classification of selected members of the xylariaceous fungi based on their molecular data were studied for resolving undescribed species relied on morphological and cultural characteristics. Three hundreds and thirty eight xylariaceous specimens were collected from natural habitats of 14 localities in different 11 provinces in Thailand. The specimens were identified and classified into species level. The high numbers of collected specimens belonged to genera *Hypoxylon* and *Xylaria* respectively. Both genera were also wide distribution, and found to reveal high variation in their morphological characteristics whereas the other xylariaceous genera were rarely represented especially *Astrocystis*.

Four xylariaceous isolates, *X. anisopleura* (ST2329), *Xylaria* sp. (ST2372), *X. cubensis* (ST2326), and *X. grammica* (ST2348), were selected for the study of secondary metabolite profiles using TLC comparing to xylariaceous endophytes. The profiles of secondary metabolites extracted from 100 mL cultural broth (2% malt extract broth containing 6% glucose) of the four isolates were similar, and did not exhibit any differences among species. Therefore, an isolate, *Xylaria cubensis* (ST2326), was cultured in 1-L cultural broth. Its secondary metabolites were extracted and analyzed by TLC method then compared to nine isolates of xylariaceous endophytes (Ruchikachorn, 2005). Each isolate had its different secondary metabolite profile analyzed by R<sub>f</sub> values. Although this technique is very useful to classify the

xylariaceous endophytes, it was time-consuming. Also, it needed high concentration of metabolites obtained from large volume of fungal culture for extraction.

One hundred and sixty nine representatives of xylariaceous fungi were investigated in their nucleotide sequences of 18S rDNA and/or the internal transcribed spacer (ITS) 1 and 2 regions including 5.8S rDNA. These nucleotide sequences were then compared to sequences from thirty eight reference specimens. It was found that 18S rDNA sequences of Astrocystis and Rosellinia which are very closely related genera according to their morphological characteristics were approximately 2,056 and 2,210 bp respectively. Nucleotide sequence of A. mirabilis (SUT056) exhibited 70.3% identity to Rosellinia sp. ST2310 and 68.7% identity to R. necatrix from GenBank database accession number AB014044. The results of ITS1-5.8S-ITS2 sequence analysis showed approximately 515 and 493 bp of A. mirabilis (SUT051 and SUT056) and Rosellinia sp. ST2310 respectively. ITS sequence comparison among both species ranged from 65.9% to 70.6% identity. However, molecular data of 18S rDNA and ITS1-5.8S-ITS2 sequences demonstrated the dissimilarity between Astrocystis and Rosellinia, which confirmed the opinion of Petrini (1993) and Whalley (1996) and disagreed with Ju and Rogers (1990) who combined Astrocystis with Rosellinia.

*Camillea tinctor*, which is the common species of *Camillea* found in Thailand were studied and compared to *C. selangorensis*. Three isolates of *C. tinctor* (SUT161, SUT260, and ST2321) and a reference specimen of *C. selangorensis* (KS15) were investigated on ITS1-5.8S-ITS2 sequences and their sizes ranged from 496 to 529 bp. ITS sequence comparison revealed the high variation within ITS1 region and it was found that the nucleotide repeated in tandem from three to five times, which might

caused by unequal crossing over or failures in the replication of the DNA. This result agreed with the previous report of the tandem repeated sequence found in Xylariales (Platas *et al.*, 2001). The phylogenetic tree of *Camillea* exhibited the separation of *C*. *selangorensis* and *C. tinctor* from each other but all *C. tinctor* examined were grouped together with *C. tinctor* sequences from GenBank database accession numbers AJ39041 and AJ39042 respectively.

*Daldinia eschscholzii* and *D. concentrica* were recently examined and five new species recognised by Stadler *et al.* (2004) based on anamorph characteristics and perispore ornamentation. In addition, some *Daldinia* collections could not be cultured and this caused problems in identification. Therefore, ITS1-5.8S-ITS2 sequences of seven *D. eschscholzii* representatives and two anamorphic isolates of *D. concentrica* (L1 and L2) were investigated. The sizes ranging from 479 to 499 bp were found. ITS sequence comparison and phylogenetic analysis of *Daldinia* examined including twenty sequences of *Daldinia* available from GenBank database indicated that all seven isolates of *D. eschscholzii* are the same species, which separated clearly from *D. concentrica* (L1 and L2).

Seventy nine isolates of *Hypoxylon* species from both sect. *Annulata* and sect. *Hypoxylon* were studied on ITS1-5.8S-ITS2 sequences. The sizes of ITS sequences varied from 445 to 906 bp. Most *Hypoxylon* sect. *Annulata*, *H. stygium*, *H. atroroseum*, *H.* cf. *stygium* (SUT231), *H. urceolatum*, and *Hypoxylon* taxonomic species 1, exhibited extremely long sequences in the ITS1 region. The whole ITS sequence alignment revealed the greatest variation in ITS1 region whereas 5.8S and ITS2 regions were more conserved. The phylogenetic tree showed clearly relationships of *Hypoxylon* species and could be used to solve the morphological

taxonomic problems.

Fifty nine isolates of *Xylaria* species and three isolates belonged to *Biscogniauxia* sp. (SUT290), *Kretzschmaria* sp. (ST2325), and *Nemania* sp. (SUT258), were investigated on ITS1-5.8S-ITS2 sequences. The sizes of ITS sequences ranged from 451 to 775 bp. Alignments of the *Xylaria* species sequences exhibited the greatest variation in the ITS1 regions whilst the 5.8S sequences gave approximately 99% similarity for all isolates tested. The phylogenetic tree showed clear separation of each species.

Therefore, these molecular data showed clearly relationships within xylariaceous species examined and also could be used to confirm results of the finding of new species. From this study, the xylariaceous fungi were identified as belonging to nine genera; *Astrocystis, Biscogniauxia, Camillea, Daldinia, Hypoxylon, Kretzschmaria, Nemania, Rosellinia* and *Xylaria*, and were represented by fifty nine species, including seven new species, *Hypoxylon kanchanapisekii* sp. nov., *Hypoxylon sublenormandii* sp. nov., *Hypoxylon suranareei* sp. nov., *Hypoxylon* taxonomic species 1 sp. nov., *Xylaria* species 2, *Xylaria* sp. nov., *Biscogniauxia* sp. nov.

The molecular data results from this study are valuable for the creation of DNA sequence database of the xylariaceous fungi found in Thailand. These nucleotide sequences can be used to design specific primers and DNA probes for certain species especially xylariaceous endophytes, which are difficult to identify. In addition, molecular data will be very useful for explaining the evolutionary and genetic variation of xylariaceous fungi found in Thailand comparing to other fungi form over the world.

## REFERENCES

- Abele, L.G., Kim, W., and Felegenhauer, B.E. (1989). Molecular evidence for inclusion of the phylum Pentastomida in the Crustacea. Molecular Biology Evolution. 6: 685-691.
- Adeboya, M., Edwards, R.L., Læssøe, T., Maitland, D.J., and Whalley, A.J.S. (1995).
  Metabolites of the higher fungi (part 28): globoscinic acid and globoscin, a labile acid-lactone system from *Xylaria globosa* and *Xylaria obovata*.
  Journal of the Chemical Society. 1: 2067-2072.
- Aguinaldo, A.M.A., Turbeville, J.M., and Linford, L.S. (1997). Evidence for a clade of nematodes, arthropods and other moulting animals. **Nature**. 387: 489-493.
- Aldridge, D.C., Burrows, B.F., and Turner, W.B. (1972). Structures of the fungal metabolites cytochalasin E and cytochalasin F (*Rosellinia necatrix*). Journal of the Chemical Society. 3: 148-149.
- Alexopoulos, C.J., Mims, C.W., and Blackwell, M. (1996). **Introductory Mycology** (4<sup>th</sup> ed.). USA: John Wiley and Sons.
- Allport, D.C. and Bulock, J.D. (1958). The pigmentation and cell-wall material of *Daldinia* sp. Journal of the Chemical Society. 1958: 4090-4094.
- Allport, D.C. and Bulock, J.D. (1960). Biosynthetic pathways in *Daldinia concentrica*. Journal of the Chemical Society. 1960: 654-662.
- Anderson, J.R., Edwards, R.L., and Whalley, A.J.S. (1982). Metabolites of the higher fungi (part 19). Serpenone, 3-Methoxy-4-methyl-5 prop-1-enylfuran-2(5H)-

one. A new y-butyrolactone from the fungus *Hypoxylon serpens* (Barron's strain) (Persoon ex Fries) Kickx. Journal of the Chemical Society Perkins Transactions. 1: 215-221.

- Anderson, J.R., Edwards, R.L., and Whalley, A.J.S. (1983). Metabolites of the higher fungi (part 21). 3-Methyl-3,4-dihydroisocoumarins and related compounds from the ascomycete family Xylariaceae. Journal of the Chemical Society Perkins Transactions. 1: 2185-2192.
- Anderson, J.R., Edwards, R.L., and Whalley, A.J.S. (1985). Metabolites of the higher fungi (part 22). 3-Butyl-3-methylsuccinic acid and 2-hexylidene-3-methylsuccinic acid from xylariaceous fungi. Journal of the Chemical Society Perkins Transactions. 1: 1481-1485.
- Andersen, B., Kroger, E., and Roberts, R. (2001). Chemical and morphological segregation of *Alternaria alternata*, *A. gaisen* and *A. longipes*. Mycological Research. 105: 291-299.
- Arnold, A.E., Maynard, Z., Gilbert, G.S., Coley, P.D., and Kursar, T.A. (2000). Are tropical endophytes hyperdiverse? **Ecology Letters**. 3: 267-274.
- Azevedo, J.L., Maccheroni, W., Pereira, J.O., and Araujo, W.L. (2000). Endophytic microorganisms: a review on insect control and recent advances on tropical plants. EJB Electronic Journal of Biotechnology [On-line serial]. Available: http://www.ejb.org/content/vol3/issue1/full/4.
- Baldauf, S.L. (2003). Phylogeny for the faint of heart: a tutorial. **Trends in Genetics**. 19: 345-351.
- Barnett, H.L. (1957). *Hypoxylon punctulatum* and its conidial state on dead oak trees and in culture. **Mycologia**. 49: 588-595.

- Bayman, P., Angulo-Sandoval, P., Baez-Ortiz, Z., and Lodge, D.J. (1998).
  Distribution and dispersal of *Xylaria* endophytes in two tree species in Puerto Rico. Mycological Research. 102: 944-948.
- Bayman, P., Lebron, L.L., Tremblay, R., and Lodge, D.J. (1997). Variation in endophytic fungi from roots and leaves of *Leanthes* (Orchidaceae). New Phytologist. 135: 143-149.
- Berkeley, M.L. and Broome, C.E. (1875). Enumeration of the fungi of Ceylon. Journal of the Linnean Society. 14: 29-140.
- Birch, P.R.J., Sims, P.F.G., and Broda, P. (1992). Nucleotide sequence of a gene from *Phanerochaete chrysosporium* that shows homology to the *facA* gene of *Aspergillus nidulans*. **DNA Sequence**. 2: 319-323.
- Bodo, B., Davoust, D., Lecommandeur, D., Rebuffat, S., Genetet, I., and Pinon, J. (1987). Hymatoxin A, a diterpene sulfate phytotoxin of *Hypoxylon mammatum*, parasite of aspen. Tetrahedron Letters. 28: 2355-2358.
- Boonphong, S., Kittakoop, P., Isaka, M., Pittayakhajonwut, D., Tanticharoen, M., and Thebtaranonth, Y. (2001). Multiplolides A and B, new antifungal 10membered lactones from *Xylaria multiplex*. Journal of National Products. 64: 965-967.
- Bowen, B.H., Taylor, J.W., and White, T.J. (1992). Molecular evolution of the fungi: human pathogens. **Molecular Biological Evolution**. 9: 893-904.
- Brunner, F. and Petrini, O. (1992). Taxonomy of some *Xylaria* species and xylariaceous endophytes by isoenzyme electrophoresis. Mycological Research. 96: 723-733.

Bruns, T.D., White, T.J., and Taylor, J.W. (1991). Fungal molecular systematics.

## Annual Revolution Ecology Systems. 22: 525-564.

- Bulock, J.D. (1980). A study in secondary metabolism. In P.S. Steyn (ed.). The biosynthesis of mycotoxins (pp. 1-16). New York: Academic Press.
- Bussaban, B., Lumyong, S., Lumyong, P., McKenzie, E.H.C., and Hyde, K.D. (2001).
  Endophytic fungi from *Amomum siamense*. Canadian Journal of Microbiology. 47: 1-6.
- Callan, B.E. and Rogers, J.D. (1986). Cultural characters and anamorphs of *Biscogniauxia* (= *Nummularia*) marginata, B. dennisii and B. repanda.
  Canadian Journal of Botany. 64: 842-847.
- Callan, B.E. and Rogers, J.D. (1990). Teleomorph-anamorph connections and correlations in some *Xylaria* species. **Mycotaxon**. 36: 343-369.
- Candoussau, F. and Rogers, J.D. (1990). Notes on *Obolarina dryophila* from France. Mycotaxon. 39: 345-349.
- Cansrikul, A. (1977). Mushrooms in Thailand. Bangkok: Thai Watanaphanich.
- Carroll (1963). Studies on the Flora of Thailand 24. Pyrenomycetes. Dansk Botansk Arkiv. 23: 101-114.
- Carroll, G. (1988). Fungal endophytes in stems and leaves: from latent pathogen to mutualistic symbiont. **Ecology**. 69: 2-9.
- Cellerino, G.P., Anselmi, N., and Giorcelli, A. (1988). Repartition en Italie et conditions favorisant les attaques de *Rosellinia necatrix* Prill. sur Peuplier.
   Annales of the Faculty of Science and Agriculture University of Torino. 15: 165-176.
- Chapela, I.H. and Boddy, L. (1988). Fungal colonization of attached beech branches
  1. Early stages of development of fungal communities. New Phytologist.
  110: 39-45.

- Chen, Y.-S. (1960). Studies on the metabolic products of *Rosellinia necatrix* Berlese
  Pt. 1. Isolation and characterization of several physiologically active neutral substances. Bulletin Agriculture Chemical Society of Japan. 24: 372-381.
- Chen, Y.-S. (1964). Studies on the metabolic products of Rosellinia necatrix Berlese
  II. The structure of rosellinic acid. Agriculture Biology Chemistry. 28: 431435.
- Chester, C.G.G. and Greenhalgh, G.N. (1964). Geniculosporium serpens gen. et sp. nov., the imperfect state of Hypoxylon serpens. Transactions of the British Mycological Society. 47: 393-401.
- Child, M. (1932). The genus *Daldinia*. **Annals of the Missouri Botanical Garden**. 19: 429-496.
- Chinworrungsee, M., Kittakoop, P., Isaka, M., Rungrod, A., Tanticharoen, M., and Thebtaranonth, Y. (2001). Antimalarial halorosellinic acid from the marine fungus *Halorosellinia oceanica*. Bioorganic and Medicinal Chemistry Letters. 11: 1965-1969.
- Chinworrungsee, M., Kittakoop, P., Isaka, M., Chanphen, R., Tanticharoen, M., and Thebtaranonth, Y. (2002). Halorosellins A and B, unique isocoumarin glucosides from the marine fungus *Halorosellinia oceanica*. Journal of the Chemical Society Perkins Transactions. 22: 2473-2476.
- Claydon, N., Grove, J.F., and Pople, M. (1985). Elm bark beetle boring and feeding deterrents from *Phomopsis oblonga*. **Phytochemistry**. 24: 937-943.
- Clayton, C.N., Julis, A.J., and Sutton, T.B. (1976). Root rot diseases of apple in North Carolina. Agricultural Experiment Station. North Carolina State University at Raleigh. **Bulletin**. 455: 3-11.

- Cox, G.M., Rude, T.H., Dykstra, C.C., and Perfect, J.R. (1995). The actin gene from *Cryptococcus neoformans*: structure and phylogenetic analysis. Journal of Medicine Veterinary Mycology. 33: 261-266.
- Crane, J.L. and Dumont, K.P. (1975). Hyphomycetes from the West India and Venezuela. Canadian Journal of Botany. 53: 843-851.
- Cummings, M.P., King, L.M., and Kellog, E.A. (1994). Slipped-strand mispairing in a plastid gene: *rpo*C2 in grasses (Poaceae). **Molecular Biology Evolution**. 11: 1-8.
- Davis, E.C., Franklin, J.B., Shaw, A.J., and Vilgalys, R. (2003). Endophytic *Xylaria* (Xylariaceae) among liverworts and angiosperms: phylogenetics, distribution, and symbiosis. American Journal of Botany. 90: 1660-1667.
- Dennis, R.W.G. (1957). Further notes on tropical American Xylariaceae. Kew Bulletin. 1957: 297-332.

Diehl, W.W. (1925). The genus Astrocystis. Mycologia. 17: 185-190.

- Duong, L.M., Lumyong, S., Hyde, K.D., and Jeewon, R. (2004). *Emarcea castanopsidicola* gen. et sp. nov. from Thailand, a new xylariaceous taxon based on morphology and DNA sequences. **Studies in Mycology**. 50: 253-260.
- Eckblad, F.-E. and Granmo, A. (1978). The genus *Nummularia* (Ascomycetes) in Norway. Norwegian Journal of Botany. 25: 69-75.
- Edwards, R.L., Jonglaekha, N., Kshirsagar, A., Maitland, D.J., Mekkamol, S., Nugent, L.K., Phosri, C., Rodtong, S., Ruchikachorn, N., Sangvichien, E., Sharples, G.P., Sihanonth, P., Suwannasai, N., Thienhirun, S., Whalley, A.J.S., and Whalley, M.A. (2003). The Xylariaceae as phytopathogens. Recent Research Developments in Plant Sciences. 1: 1-19.

- Edwards, R.L., Maitland, D.J., and Whalley, A.J.S. (1989). Metabolites of the higher fungi (part 24). Cytochalasin N, O, P, Q, and R. New cytochalasins from the fungus *Hypoxylon terricola* Mill. **Journal of the Chemical Society Perkins Transactions**. 1: 57-65.
- Edwards, R.L., Maitland, D.J., Scowen, I.J., de Sousa, A.J.T., and Whalley, A.J.S. (2001). Metabolites of the higher fungi (part 32). Rosenectrone, a phytotoxic bicycle (4.1.0) hept-3-en-2-one from the fungus *Rosellinia necatrix* Prill.
  Journal of the Chemical Society Perkins Transactions. 1: 537-542.
- Edwards, R.L., Poyser, J.P., and Whalley, A.J.S. (1988). Metabolites of the higher fungi (part 23). The punctaporonins. Novel bi-, tri-, and tetra-cyclic sesquiterpenes related to caryophyllene from the fungus *Poronia punctata* (Linnaeus: Fries) Fries. Journal of the Chemical Society Perkins Transactions. 1: 823-831.
- Edwards, R.L. and Whalley, A.J.S. (1979). Metabolites of the higher fungi. Part 18. 3-butyl-4-methylfuran-2(5H)-one and 3-butyl-4-methylenefuran-2(5H)-one.
  New y-butyrolactones from the fungus *Hypoxylon serpens* (Persoon ex Fries) Kickx. Journal of the Chemical Society Perkins Transactions. 1: 803-806.
- Eriksson, O.E. and Hawksworth, D.L. (1993). Outline of the ascomycetes-1993. Systema Ascomycetum. 12: 51-257.
- Felsenstein, J. (1985). Confidence limits on phylogenies: an approach using the bootstrap. **Evolution**. 39: 783-791.
- Field, K.G., Olsen, G.J., and Lane, D.L. (1988). Molecular phylogeny of the animal kingdom. Science. 239: 748-753.

- Francis, S.M. (1986). Needle blights of conifers. Transactions of the British Mycological Society. 87: 397-400.
- Francis, S.M., Minter, D.W., and Caine, T.S. (1980). Three new species of Anthostomella. Transactions of the British Mycological Society. 75: 201-206.
- Friedrich, M. and Tautz, D. (1995). Ribosomal DNA phylogeny of the major extant arthropod classes and the evolution of myriapods. **Nature**. 376: 165-167.
- Friedrich, M. and Tautz, D. (1997). Evolution and phylogeny of the Diptera: a molecular phylogenetic analysis using 28S rDNA sequences. Systematic Biology. 46: 674-698.
- Fries, N. (1849). Summa Vegetabilium Scandinaviae (part 2). Stockholm and Leipzig: Bonnier.
- Frisvad, J.C., Bridge, P.D., and Arora, D.K. (1998). Chemical Fungal Taxonomy. New York: Marcel Dekker.
- Frisvad, J.C. and Samson, R.A. (1991). Mycotoxins produced by species of *Penicillium* and *Aspergillus* occuring in cereals. In J. Chelkowski (ed.).
  Cereal Grain. Mycotoxins, Fungi and quality in drying and storage (pp. 441-476). Amsterdam: Elsevier.
- Furuya, K. and Udagawa, S. (1977). Coprophilous pyrenomycetes from Japan. IV.Transactions of the Mycological Society of Japan. 17: 243-261.
- Gardes, M. and Bruns, T.D. (1993). ITS primers with enhanced specificity for basidiomycetes-application to the identification of mycorhizae and rusts. Molecular Ecology. 2: 113-118.
- González, F.S.M. and Rogers, J.D. (1993). *Biscogniauxia* and *Camillea* in Mexico. Mycotaxon. 47: 229-258.

- González, I.L., Sylvester, J.E., Smith, T.F., Stambolian, D., and Schmickel, R.D. (1990). Ribosomal RNA gene sequences and hominoid phylogeny.
   Molecular Biology Evolution. 7: 203-219.
- Goosen, T. and Debets, F. (1996). Molecular genetic analysis. In C.J. Bos (ed.).
  Fungal genetics: Principles and practices (pp. 97-117). New York: Marcel Dekker. Granmo, A., Hammelev, D., Knudsen, H., Læssøe, T., Sara, M., and Whalley, A. (1989). The genera *Biscogniauxia* and *Hypoxylon* (Sphaeriales) in the Nordic countries. Opera Botanica. 100: 59-84.
- Graur, D. and Li, W.-H. (1999). Fundamentals of Molecular Evolution. Sinauer Associates.
- Greenhalgh, G.N. (1967). A note on the conidial scar in the Xylariaceae. New Phytologist. 66: 65-66.
- Greenhalgh, G.N. and Chesters, C.C.G. (1968). Conidiophore morphology in some British members of the Xylariaceae. Transactions of the British Mycological Society. 51: 57-82.
- Greenhalgh, G.N. and Whalley, A.J.S. (1970). Stromal pigments of some species of *Hypoxylon*. Transactions of the British Mycological Society. 55: 89-96.
- Guarro, J., Gene, J., and Stchigel, A.M. (1999). Developments in fungal taxonomy. Clinical Microbiology Reviews. 12: 454-500.
- Guillaumin, J.J., Mercier, S., and Dubos, B. (1982). Les pourridies a Armillariella et Rosellinia en France sur vigne, arbres fruitiers et cultures florales. L.
  Ftiologie et symptomatologie. Agronomie. 2: 71-80.
- Gunawan, S., Steffan, B., and Steglich, W. (1990). Xylaral, ein Hydroxyphthalid-Derivat aus fruchtkorpern von Xylaria polymorpha (Ascomycetes). Liebigs Annual Chemistry: 19: 825-827.
- Guo, L.D., Hyde, K.D., and Liew, E.C.Y. (2000). Identification of endophytic fungi from *Livistona chinensis* based on morphology and rDNA sequences. New Phytologist. 147: 617-630.
- Harmsen, M.C., Schuren, F.H.J., Moukha, S.M., Van Zuilen, C.M., Punt, P.J., and Wessels, J.G.H. (1992). Sequence analysis of the glyceraldehydes-3phosphate dehydrogenase genes from the basidiomycetes *Schizophyllum commune*, *Phanerochaete chrysosporium* and *Agaricus bisporus*. Current Genetics. 22: 447-454.
- Hall, B.G. (2000). Phylogenetic Trees Made Easy: a How-To Manual for Molecular Biologists. Sinauer Associates.
- Hawksworth, D.L. (1971). A revision of the genus Ascotricha Berk. Mycological Papers. 126: 1-28.
- Hawksworth, D.L. and Whalley, A.J.S. (1985). A new species of *Rhopalostroma* witha *Nodulisporium* anamorph from Thailand. Transactions of the BritishMycological Society. 84: 560-562.
- Higgins, D.G. and Sharp, P.M. (1988). CLUSTAL: a package for performing multiple sequence alignment on a microcomputer. **Gene**. 73: 237-244.
- Hwang, U.-W. and Kim, W. (1999). General properties and phylogenetic utilities of nuclear ribosomal DNA and mitochondrial DNA commonly used in molecular systematics. The Korean Journal of Parasitology. 37: 215-228.
- Hwang, U.-W., Kim, W., Tautz, D., and Friedrich, M. (1998). Molecular phylogenetics at the Felsenstein zone: approaching the *Strepsiptera* problem using 5.8S and 28S rDNA sequences. **Molecular Phylogenetic Evolution**. 9: 470-480.

- Hyde, K.D. (1995). Fungi from palms. XXI. The genus Seynesia. Sydowia. 47: 180-198.
- Isaka, M., Jaturapat, A., Rukseree, K., Danwisetkanjana, K., Tanticharoen, M., and Thebtaranonth, Y. (2001). Phomoxanthones A and B, novel xanthone dimers from the endophytic fungus *Phomopsis* species. Journal of National Products. 64: 1015-1018.
- Johannesson, H., Laessoe, T., and Stenlid, J. (2000). Molecular and morphological investigation of *Daldinia* in northern Europe. **Mycological Research**. 104: 275-280.
- Jong, S.C. and Davis, E.E. (1974). *Areolospora*, a new humicolous genus in the Xylariaceae. Norwegian Journal of Botany. 21: 23-30.
- Jong, S.C. and Rogers, J.D. (1972). Illustrations and descriptions of conidial states of some *Hypoxylon* species. Washington State University Agricultural Experiment Station Technical Bulletin. 71: 1-49.
- Ju, Y.-M., González, F.S.M., and Rogers, J.D. (1993). Three xylariaceous fungi with scolecosporous conidia. **Mycotaxon**. 47: 219-228.
- Ju, Y.-M. and Rogers, J.D. (1990). Astrocystis reconsidered. Mycologia. 82: 342-349.
- Ju, Y.-M. and Rogers, J.D. (1996). A revision of the genus Hypoxylon. St Paul. Minnesota. U.S.A.: American Phytopathological Society Press.
- Ju, Y.-M., Rogers, J.D., and San Martin, F. (1997). A revision of the genus *Daldinia*. **Mycotaxon**. 61: 243-293.
- Ju, Y.-M., Vasilyeva, L., and Rogers, J.D. (1999). *Daldinia singularis* sp. nov. from eastern Russia, and notes on some other taxa. **Mycotaxon**. 71: 405-412.
- Kimura, Y., Nakajima, H., and Hamasaki, T. (1989). Structure of rosellichalasin, a new metabolite produced by *Rosellinia necatrix*. Agricultural and

Biological Chemistry. 53: 1699-1701.

- Ko, W.H. and Kunimoto, R.K. (1991). Quick decline of macadamia trees: association with *Xylaria arbuscula*. **Plant Pathology**. 40: 643-644.
- Koehn, R.D. and Cole, G.T. (1975). An ultrastructural comparison of *Podosordaria* leporina and *Poronia oedipus* (Ascomycetes). Canadian Journal of Botany. 53: 2251-2259.
- Læssøe, T. (1994). Index Ascomycetum 1. Xylariaceae. Systema Ascomycetum. 1: 43-112.
- Læssøe, T. and Lodge, D.J. (1994). Three host specific *Xylaria* species. **Mycologia**. 86: 436-446.
- Læssøe, T., Rogers, J.D., and Whalley, A.J.S. (1989). *Camillea, Joniella*, and light-spored species of *Hypoxylon*. **Mycological Research**. 93: 121-155.
- Læssøe, Y. and Spooner, B.M. (1994). *Rosellinia* and *Astrocystis* (Xylariaceae): new species and generic concepts. **Kew Bulletin**. 49: 1-70.
- Lee, J.S., Ko, K.S., and Jung, H.S. (2000). Phylogenetic analysis of *Xylaria* based on nuclear ribosomal ITS1-5.8S-ITS2 sequences. FEMS Microbiology Letters. 187: 89-93.
- Lee, S.B. and Taylor, J.W. (1992). Phylogeny of five fungus-like protoctistan *Phytophthora* species, inferred from the internal transcribed spacers of ribosomal DNA. **Molecular Biology Evolution**. 9: 636-653.
- Levinson, G. and Gutman, G.A. (1987). Slipped-strand mispairing: a major mechanism for DNA sequence evolution. **Molecular Biology Evolution**. 4: 203-221.
- Li, W.-H. and Graur, D. (1991). Fundamentals of Molecular Evolution. Sunderland: Sinauer.

- Lloyd, C.G. (1924). Additional notes on *Camillea*. Mycological Writing. 7: 1280-1282.
- Lodge, D.J., Fistter, P.J., and Sutton, B.C. (1996). Endophytic fungi of *Manilkara bidentata* leaves in Puerto Rico. **Mycologia**. 88: 733-738.
- Lumyong, S., Thongkantha, S., Lumyong, P., and Tomita, F. (2000). Endophytic fungi from 13 bamboo species in Thailand. Biotechnology for Sustainable Utilization of Biological Resources in the Tropics. 14: 96-101.
- Lund, F. and Frisvad, J.C. (1994). Chemotaxonomy of *Penicillium aurantiogriseum* and related species. **Mycological Research**. 98: 481-492.
- Macara, A.M. (1975). Estimative em 1975. Dos prejuizos causados pelas principais doencas do sobreiro num montado da regiao ribatejana. Boletim do Instituto dos Produtos Florestais. **Cortica**. 444: 205-212.
- Manion, P.D. and Griffin, D.M. (1986). Sixty-five years of research on *Hypoxylon* canker of aspen. **Plant Disease**. 70: 803-808.
- Martin, P. (1967). Studies in the Xylariaceae: 1. New and old concepts. Journal of South African Botany. 33: 205-228.
- Martin, P. (1968). Studies in the Xylariaceae IV. *Hypoxylon*. Sections *Papillata* and *Annulata*. **South African Journal of Botany**. 34: 303-330.
- Martin, P. (1969). Studies in the Xylariaceae V. *Euhypoxylon*. Journal of South African Botany. 35: 149-206.
- Mazzaglia, A., Anselmi, N., Vicario, S., and Vannini, A. (2001b). Sequence analysis of the 5.8S rDNA and ITS regions in evaluating genetic relationships among some species of *Hypoxylon* and related genera. **Mycological Research**. 105: 670-675.

- Mekkamol, S. (1998). Endophytic fungi in *Tectona grandis* L. (Teak). Ph.D. Dissertation, Liverpool John Moores University, U.K.
- Merrill, W., French, D.W., and Wood, F.A. (1964). Decay of wood by species of the Xylariaceae. **Phytopathology**. 54: 56-58.
- Miller, J.H. (1961). A monograph of the world species of *Hypoxylon*. Athens, U.S.A.: University of Georgia Press.
- Minter, D.W. and Webster, J. (1983). Wawelia octospora sp. nov., a xerophilous and coprophilous member of the Xylariaceae. Transactions of the British Mycological Society. 80: 370-373.
- Morgan-Jones, G. and Hashmi, M.H. (1973). The conidial state of *Xylaria johorensis*. **Canadian Journal of Botany**. 51: 109-111.
- Morgen, J.A.T. and Blair, D. (1998). Relative merits of nuclear ribosomal internal transcribed spacers and mitochondrial CO1 and ND1 genes for distinguishing among *Echinostoma* species. **Parasitology**. 116: 289-297.
- Mühlbauer, A., triebel, D., Persoh, D., Wollweber, H., Seip, S., and Stadler, M. (2002). Macrocarpones, novel metabolites from stromata of *Hypoxylon* macrocarpum and new evidence on the chemotaxonomy of *Hypoxylon*.
  Mycological Progress. 1: 235-248.
- Navajas, M., Lagnel, J., Gutierrez, J., and Boursot, P. (1998). Species-wide homogeneity of nuclear ribosomal ITS2 sequences in the spider mite *Tetranychus urticae* contrasts with extensive mitochondrial COI polymorphism. **Heredity**. 80: 742-752.
- Nei, M. and Kumar, S. (2000). Molecular Evolution and Phylogenetics. Cambridge University Press.

- Nilsson, T., Daniel, G., Kirk, T.K., and Obst, J.R. (1989). Chemistry and microscopy of wood decay by some higher ascomycetes. **Holzforschung**. 43: 11-18.
- O'Donnell, K. (1992). Ribosomal DNA internal transcribed spacers are highly divergent in the phytopathogenic ascomycetes *Fusarium sambucinum* (*Gibberella pulicaris*). **Current Genetics**. 22: 213-220.
- Okada, G., Takematsu, A., and Takamura, Y. (1997). Phylogenetic relationships of the hyphomycete genera *Chaetopsina* and *Kionochaeta* on 18S rDNA sequences. **Mycoscience**. 38: 409-420.
- Page, R.D.M. and Holmes, E.C. (1998). Molecular Evolution: a Phylogenetic Approach. Blackwell Science.
- Patouillard, N. (1888). Fragments Mycologiques. Le genre *Camillea* et ses allies. Journal de Botanique. 2: 49-53.
- Pedersen, E.J., Larsen, P., and Boll, P.M. (1980). Engleromycin, a new cytochalasin from *Engleromyces goetzei* Hennings. **Tetrahedron Letter**. 21: 5079-5082.
- Penzig, O. and Saccardo, P.A. (1904). Icones Fungorum. Bril Leiden. 124.
- Pereira, J.O., Azevedo, J.L., and Petrini, O. (1993). Endophytic fungi of *Stylosanthes*: a first report. **Mycologia**. 85: 362-364.
- Perera, O.P., Cockburn, A.F., Mitchell, S.E., Conn, J., and Seawright, J.A. (1998).
  Species specific repeat units in the intergenic spacer of the ribosomal RNA cistron of Anopheles aquasalis. Current American Journal of Medicine. 59: 673-678.
- Petrak, F. (1961). Uber das Auftreten von Rosellinia herpotrichioides Hepting and Davidson auf jungen Fichten im Pflanzgarten Rauris der Forstverwaltung Lend in Salzburg. Sydowia. 15: 242-246.

- Petrini, O. (1992). Fungal endophytes of tree leaves. In J.H. Andrews and S.S. Hirano (eds.). Microbial Ecology of Leaves (pp. 179-197). New York: Springer Verlag.
- Petrini, L.E. (1992). Rosellinia species of the temperate zones. Sydowia. 44: 169-281.
- Petrini, L.E. and Müller, E. (1986). Haupt- und Nebenfruchtfomen europaischer *Hypoxylon*-Arten (Xylariaceae, Sphaeriales) und verwandter Pilze.
  Mycologia Helvetica. 1: 501-627.
- Petrini, L.E. and Petrini, O. (1985). Xylariaceous fungi as endophytes. Sydowia. 36: 216-234.
- Petrini, L.E., Petrini, O., and Fisher, P.J. (1987). Anthostomella calligoni, an endophyte of Suaeda fruticosa in Dorset. Transactionss of the British Mycological Society. 89: 387-389.
- Petrini, O., Petrini, L.E., and Rodrigues, K.F. (1995). Xylariaceous endophytes: an exercise in biodiversity. **Fitopatologia Brasileira**. 20: 531-539.
- Petrini, L.E. and Rogers, J.D. (1986). A summary of the *Hypoxylon serpens* complex. Mycotaxon. 26: 409-436.
- Phanichapol, D. (1968). Check-list of fungi in the Forest Herbarium. Natural History bulletin of the Siam Society. 22: 263-269.
- Photita, W., Lumyong, S., Lumyong, P., and Hyde, K.D. (2001). Endophytic fungi of wild banana (*Musa acuminata*) at Doi Suthep Pui National Park, Thailand.
  Mycological Research. 105: 1508-1513.
- Pinon, J. and Manion, P.D. (1991). *Hypoxylon mammatum* and its toxins recent advances in understanding their relationships with canker disease of poplar.
   European Journal of Forest Pathology. 21: 202-209.

- Platas, G., Acero, J., Borkowski, J.A., González, V., Portal, M.A., Rubio, V., Sánchez-Ballesteros, J., Salazar, O., and Peláez, F. (2001). Presence of a simple tandem repeat in the ITS1 region of the Xylariales. Current Microbiology. 43: 43-50.
- Pouzar, Z. (1972). *Hypoxylon fraxinophilum* spec. nov. and *H. moravicium* spec. nov., two interesting species found on *Fraxinus angustifolia*. Česká Mykologie. 26: 129-137.
- Pouzar, Z. (1985). Reassessment of *Hypoxylon serpens* complex I. Céska Mykologie. 39: 15-25.
- Quang, D.N., Hashimoto, T., Tanaka, M., Baumgartner, M., Stadler, M., and Asakawa, Y. (2002). Chemical constituents of the ascomycete *Daldinia concentrica*. Journal of Natural Products. 65: 1869-1874.
- Radford, A. (1993). A fungal phylogeny based upon orotidine-5'-monophosphate decarboxylase. Journal of Molecular Evolution. 36: 389-395.
- Rayner, R.W. (1970). A mycological colour chart. Commonweath Mycological Institute. Kew.
- Rodrigues, K.F. (1992). Endophytic fungi in the tropical palm *Euterpe oleracea* Mart. Ph.D. Dissertation, University of New York, U.S.A.
- Rodrigues, K.F. (1994). The foliar fungal endophytes of the Amazonian palm *Euterpe oleracea* Mart. **Mycologia**. 86: 376-385.
- Rodrigues, K.F., Leuchtmann, A., and Petrini, O. (1993). Endophytic species of *Xylaria*: cultural and isozymic studies. **Sydowia**. 45: 116-138.
- Rodrigues, K.F., and Petrini, O. (1997). Biodiversity of endophytic fungi in a tropical regions. In K.D. Hyde. Biodiversity of Tropical Microfungi (pp. 57-69).
  Hong Kong: Hong Kong University Press.

- Rodrigues, K.F. and Samuels, G.J. (1990). Preliminary study of endophytic fungi in a tropical palm. **Mycological Research**. 94: 827-830.
- Rogers, J.D. (1975). *Xylaria polymorpha* II. Cytology of a form with typical robust stromata. **Canadian Journal of Botany**. 53: 1736-1743.
- Rogers, J.D. (1977). A new *Hypoxylon* species with appendaged, ornamented ascospores. Canadian Journal of Botany. 55: 2394-2398.
- Rogers, J.D. (1979). The Xylariaceae: Systematic, biological and evolutionary aspects. Mycologia. 71: 1-42.
- Rogers, J.D. (1982). *Entonaema liquescens*: description of the anamorph and thoughts on its systematic position. **Mycotaxon**. 23: 429-437.
- Rogers, J.D. (1984). Xylaria cubensis and its anamorph Xylocoremium flabelliforme,
   Xylaria allantoidea and Xylaria poitei in Continental United States.
   Mycologia. 76: 912-923.
- Rogers, J.D. (1985). Anamorphs of *Xylaria*: taxonomic considerations. **Sydowia**. 38: 255-262.
- Rogers, J.D. (1994). Problem genera and family interfaces in the eupyrenomycetes. In
   D.L. Hawksworth. Ascomycete Systematics: Problems and Perspectives
   in the Nineties (pp. 321-331). New York: Plenum Press.
- Rogers, J.D. (2000). Thoughts and musings on tropical Xylariaceae. Mycological Research. 104: 1412-1420.
- Rogers, J.D. and Callan, B.E. (1986). *Xylaria poitei*: stromata, cultural description, and structure of conidia and ascospores. **Mycotaxon**. 26: 287-296.
- Rogers, J.D. and Læssøe, T. (1992). *Podosordaria ingii* sp. nov. and its *Lindquistia* anamorph. **Mycotaxon**. 44: 435-443.

- Rogers, J.D., Læssøe, T., and Lodge, D.J. (1991). *Camillea*: new combinations and a new species. **Mycologia**. 83: 224-227.
- Rogers, J.D., Ju, Y.-M., and Hemmes, D.E. (1997). *Xylaria moelleroclavus* sp. nov. and its *Moelleroclavus* anamorphic state. **Mycological Research**. 100: 669-674.
- Ruchikachorn, N. (2005). Endophytic fungi of *Cassia fistula* L. Ph.D. Dissertation. Liverpool John Moores University. UK.
- Salisbury, P.J. and Long, J.R. (1956). A new needle blight of Douglas fir seedlings caused by *Rosellinia herpotrichioides* Hepting and Davidson. **Proceedings** of the Canadian Phytopathological Society. 23: 19.
- Samuels, G.J. (1989). *Thuemenella cubispora*, a xylariaceous ascomycete and its biogeography. **Mycological Society of America Newsletter**. 40: 46.
- Samuels, G.J. and Müller, E. (1980). Life history studies on Brazilian ascomycetes. 8. *Thamnomyces chordalis* (anam.: *Nodulisporium*) and *Camillea bacillum* (anam.: *Geniculosporium*) with notes on taxonomy of the Xylariaceae.
  Sydowia. 33: 274-281.
- Samuels, G.J., Müller, E., and Petrini, O. (1987). Studies in the Amphisphaeriaceae (sensu lato) 3. New species of *Monographella* and *Pestalosphaeria* and two new genera. Mycotaxon. 28: 473-499.
- Samuels, G.J., Rogers, J.D., and Nagasawa, E. (1987). Studies in the Amphisphaeriaceae (sensu lato). 1. *Collodiscula japonica* and its anamorph, *Acanthodochium collodisculae*. **Mycotaxon**. 28: 453-459.
- Samuels, G.J. and Rossman, A.Y. (1992). *Thuemenella* and *Sarawakus*. Mycologia. 84: 26-40.

- Sanchez-Ballesteros, J., Gonzalez, V., Salazar, O., Acero, J., Portal, M.A., Julian, M., and Rubio, V. (2000). Phylogenetic study of *Hypoxylon* and related genera based on ribosomal ITS sequences. **Mycologia**. 92: 964-977.
- San Maetin González, F. (1992). A mycofloristic and cultural study of the Xylariaceae of Maxico. Ph.D. dissertation. Washington State University.
- Schulz, B., Boyle, C., Draeger, S., Römmert, A., and Krohn, K. (2002). Endophytic fungi: a source of novel biologically active secondary metabolites.
   Mycological Research. 106: 996-1004.
- Schumacher, T. (1982). Ascomycetes from Northern Thailand. Nordic Journal of Botany. 2: 257-263.
- Sivanesan, A. and Holliday, P. (1972). *Rosellinia necatrix*. C.M.I. Descriptions of Pathogenic Fungi and Bacteria. No. 352.
- Smith, R.S. (1966). *Rosellinia* needle blight of Douglas fir (*Pseudotsuga menziesii*) in California. **Plant Disease Reporter**. 50: 249-250.
- Stadler, M., Baumgartner, M., Wollweber, H., Rogers, J.D., and Ju, Y.-M. (2001). *Daldinia decipiens* sp. nov. and notes on some other European *Daldinia* spp. inhabiting Betulaceae. **Mycotaxon**. 80: 167-177.
- Stadler, M., Wollweber, H., Mühlbauer, A., Asakawa, Y., Hashimoto, T., Rogers, J.D., Ju, Y.-M., Wetzstein, H.-G., and Tichy, H.-V. (2001). Molecular chemotaxonomy of *Daldinia* and other Xylariaceae. Mycological Research. 105: 1191-1205.
- Stadler, M., Ju, Y.-M., and Rogers, J.D. (2004). Chemotaxonomy of *Entonaema*, *Rhopalostroma* and other Xylariaceae. Mycological Research. 108: 239-256.

- Steglich, W., Klaar, M., and Furtner, W. (1974). (+)-Mitorubrin derivatives from *Hypoxylon fragiforme*. **Phytochemistry**. 13: 2874-2875.
- Stiers, D.L., Rogers, J.D., and Russell, D.W. (1973). Conidial state of *Poronia* punctata. Canadian Journal of Botany. 51: 481-484.
- Strobel, G.A. and Long, D. (1998). Endophytic microbes embody pharmaceutical potential. ASM News. 64: 263-268.
- Strobel, G.A., Dirksie, E., Sears, J., and Markworth, C. (2001). Volatile antimicrobials from a novel endophytic fungus. Microbiology. 147: 2943-2950.
- Strobel, G.A., Ford, E., Worapong, J., Harper, J.K., Arif, A.M., Grant, D.M., Fung, P., and Chau, R.M.W. (2002). Isopestacin, a unique isobenzofuranone from *Pestalotiopsis microspora* possessing antifungal and antioxidant properties. **Phytochemistry**. 60: 179-183.
- Subramanian, C.V. and Chandrashekara, K.V. (1977). *Lindquistia*, a new hyphomycete genus. **Boletin de la Sociedad Argentina de Botanica**. 18: 145-151.
- Sutherland, J.B. and Crawford, D.L. (1981). Lignin and glucan degradation by species of Xylariaceae. Transactions of the British Mycological Society. 76: 335-337.
- Swofford, D.L. and Olsen, G.L. (1990). Phylogeny reconstruction. In D. Hillis and C. Moritz (eds.). Molecular Systematics (pp. 411-501). Sunderland: Sinauer Associates.
- Szaniszlo, P.J. and Momany, M. (1993). Chitin, chitin synthase and chitin synthase conserved region homologues in *Wangiella dematitidis*. In B. Maresca, G.S.

Kobayashi and H. Yamaguchi (eds.). **Molecular biology and its application to medical mycology** (pp. 229-242). Germany: Springer-Verlag KG.

- Sztejnberg, A. and Madar, Z. (1980). Host range of *Dematophora necatrix*, the cause of white root rot disease in fruit trees. **Plant Disease**. 64: 662-664.
- Taylor, J.E., Hyde, K.D., and Jone, B.G. (1999). Endophytic fungi associated with the temperature palm, *Trachycarpus fortunei*, within and outside its natural geographic range. New Phytologist. 142: 335-346.
- Teixeira de Sousa, A.J. (1985). Lutte contre *Rosellinia necatrix* (Hartig) Berlese, agent du pourridie laineux: sensibilite de quelques especes vegetales et lutte chimique. **European Journal of Forest Pathology**. 15: 323-332.
- Teixeira de Sousa, A.J., Guillaumin, J.J., Sharples, G.P., and Whalley, A.J.S. (1995).
   *Rosellinia necatrix* and white root rot of fruit trees and other plants in Portugal and nearby regions. Mycologist. 9: 31-33.
- Thienhirun, S. (1997). A Preliminary Account of the Xylariaceae of Thailand. Ph.D. Dissertation. Liverpool John Moores University. U.K.
- Thienhirun, S. and Whalley, A.J.S. (2001). Wood decay Xylariaceae. Fungal Diversity of Thailand: Towards a Checklist of Thai Fungi Seminar. National Center for Genetic Engineering and Biotechnology 15-16 November.
- Tsai, H.F., Liu, J.S., Staben, C., Christensen, M.J., Latch, G.C.M., Siegel, M.R., and Schardl, C.L. (1994). Evolutionary diversification of fungal endophytes of tall fescue gress by hybridization with *Epichoë species*. Proc Natl Acad Sci U.S.A. 91: 2542-2546.
- Udagawa, S.-I. and Ueda, S. (1988). *Calceomyces*, a new genus of the Xylariaceae with shoe-shaped ascospores. **Mycotaxon**. 32: 447-455.

- Valldosera, E. and Guarro, G. (1988). Some coprophilous ascomycetes from Chile. Transactions of the British Mycological Society. 90: 601-605.
- Van de Peer, Y., Chapelle, S., and Wachter, R.D. (1996). A quantitative map of nucleotide substitution rates in bacterial rRNA. Nucleic Acids Research. 24: 3381-3391.
- Van der Gucht, K. (1992). Contribution towards a revision of the genera *Camillea* and *Biscogniauxia* (Xylariaceae, Ascomycetes) from Papua New Guinea.
  Mycotaxon. 45: 259-273.
- Van der Gucht, K. (1994). The Xylariaceae of Papua New Guinea. Ph.D Thesis University of Gent. Belgium.
- Van der Gucht, K. (1995). Illustrations and descriptions of xylariaceous fungi collected in Papua New Guinea. Bulletin Jardin Botanique National de Belgique. 64: 219-403.
- Van der Gucht, K., Ju, Y.-M., and Rogers, J.D. (1997). New *Hypoxylon* species from Papua New Guinea. **Mycotaxon**. 55: 547-555.
- Van der Gucht, K. and Whalley, A.J.S. (1992). A new variety and combination for *Hypoxylon citriforme*. **Mycological Research**. 96: 895-896.
- Vogler, A.P. and DeDalle, R. (1994). Evolution and phylogenetic information content of the ITS-1 region in the Tiger Beetle *Cicindela dorsalis*. Molecular Biology Evolution. 11: 393-405.
- Whalley, A.J.S. (1976). Notes on the conidial state of *Hypoxylon udum*. **Transactions** of the British Mycological Society. 67: 515-517.
- Whalley, A.J.S. (1985). The Xylariaceae: some ecological considerations. **Sydowia**. 38: 369-382.

- Whalley, A.J.S. (1993). Tropical Xylariaceae: their distribution and ecological characteristics. In S. Isaac, J.C. Frankland, R. Watling, and A.J.S. Whalley.
  Aspects of Tropical Mycology (pp. 103-119). Cambridge: Cambridge University Press.
- Whalley, A.J.S. (1996). The xylariaceous way of life. Mycological Research. 100: 897-922.
- Whalley, A.J.S. and Edwards, R.L. (1985). Nummulariella marginata: its conidial state, secondary metabolites and taxonomic relationships. Transactions of the British Mycological Society. 85: 385-390.
- Whalley, A.J.S. and Edwards, R.L. (1987). Xylariaceous fungi: use of secondary metabolites. In A.D.M. Rayner, C.M. Brasier, and D. Moore. The Evolutionary Biology of Fungi (pp. 423-434). Cambridge: Cambridge University Press.
- Whalley, A.J.S. and Edwards, R.L. (1995). Secondary metabolites and systematic arrangement within the Xylariaceae. **Canadian Journal of Botany**. 73 (Suppl.1): S802-S810.
- Whalley, A.J.S. and Edwards, R.L. (1999). The Xylariaceae: A case study in biological and chemical diversity [On-line]. Available: http://www.iupac. org/ symposia/ proceedings/phuket97/whalley.html.
- Whalley, A.J.S. and Hammelev, D. (1988). Some Xylariaceae from Georgia and the Russian Soviet Federative Socialist Republic. Agarica. 9: 67-70.
- Whalley, A.J.S., Laessoe, T., and Kile, G.A. (1990). A new species of *Biscogniauxia* with appendaged ascospores from Tasmania. Mycological Research. 94: 27-239.

- Whalley, A.J.S. and Watling, R. (1982). Distribution of *Daldinia concentrica* in the British Isles. **Transactions of the British Mycological Society**. 78: 47-53.
- Whalley, A.J.S. and Whalley, M.A. (1977). Stromal pigments and taxonomy of *Hypoxylon*. Mycopathologia. 61: 99-103.
- Whalley, M.A. (1995). *Camillea fusiformis* sp. nov. from Ecuador. **Sydowia**. 47: 82-88.
- Whalley, M.A., Whalley, A.J.S., and Jones, E.B.G. (1996). *Camillea selangorensis* sp. nov. from Malaysia. **Sydowia**. 48: 145-151.
- Whalley, M.A., Whalley, A.J.S., Thienhirun, S., and Sihanonth, P. (1999). Camillea malaysianensis sp. nov. and the distribution of Camillea in Southeast Asia. Kew Bulletin. 54: 715-722.
- White, T.J., Bruns, T.D., Lee, S., and Taylor, J.W. (1990). Amplification and direct sequencing of fungal ribosomal RNA genes for phylogenetics. In M.A. Innis, D.H. Gelfand, J.S. Sninsky and T.J. White (eds.). PCR Protocols: a guide to methods and applications (pp. 315-322). San Diego: Academic Press.
- Whiting, M.F. (1998). Phylogenetic position of the Strepsiptera: review of molecular and morphological evidence. International Journal of Morphological Embryology. 27: 53-60.
- Wilkins, W.H. (1934). Studies in the genus Ustulina with special reference to parasitism. I. Introduction, survey of previous literature and host index.
  Transactions of the British Mycological Society. 18: 320-346.
- Wilkins, W.H. (1943). Studies in the genus Ustulina with special reference to parasitism. VI. A brief account of heart rot of beech (Fagus sylvatica L.) caused by Ustulina. Transactions of the British Mycological Society. 26: 169-170.

# **APPENDIX** A

# FUNGAL MEDIA AND REAGENTS

## 1. Reagents and medium used for morphological taxonomic study

#### 1.1 Potato Dextrose Agar (PDA)

Potato	300.00	g
Dextrose	40.00	g
Agar	15.00	g

Potato slices were boiled in 1000-mL distilled water for 30 min and filtrated. The potato solution was then mixed with dextrose and agar, and adjusted the volume to 1,000 mL with distilled water. The medium was sterilized by autoclaving for 10 minutes at 121°C, 15 lb/square inches after preparation.

#### 1.2 Melzer's reagent

Chloral hydrate	100.00	g
Potassium iodine	5.00	g
Iodine	5.00	g

The ingredients were dissolved and adjusted the volume to 100 mL with distilled water. The reagent was stored in dark bottle at room temperature.

### 1.3 10% KOH

00	g
U	0

The ingredient was dissolved and adjusted the volume to 100 mL with distilled water.

## 2. Media used for chemical taxonomic study

The media were sterilized by autoclaving for 10 min at 121°C, 15 lb/square inches after preparation.

2.1	2.1 Yeast Extract Sucrose Agar (YES)			
	Yeast Extract	20.00	g	
	MgSO <sub>4</sub> .5H <sub>2</sub> O	0.50	g	
	Agar	15.00	g	

The ingredients were dissolved and adjusted the volume to 1,000 mL with distilled water.

#### 2.2 2% Malt Extract Broth containing 6% glucose

Malt Extract Broth	20.00	g
Glucose	60.00	g

The ingredients were dissolved and adjusted the volume to 1,000 mL with distilled water.

## 3 Chemicals and reagents used for nucleic acid study

#### 3.1 Lysis buffer

Tris Base	6.06	g
EDTA (C <sub>10</sub> H <sub>14</sub> N <sub>2</sub> O <sub>8</sub> Na <sub>2</sub> .2H <sub>2</sub> O)	18.61	g
Sodium dodecylsulfate (SDS)	30.00	g
2-Mercaptoethanol	10.00	mL

The ingredients were dissolved and adjusted the volume to 1,000 mL with deionized water. Then, the solution was sterilized by autoclaving for 10 min at 121°C, 15 lb/square inches after preparation.

#### 3.2 Tris-EDTA (TE) Buffer

Tris Base	1.21	g
		$\mathcal{O}$

EDTA (
$$C_{10}H_{14}N_2O_8Na_2.2H_2O$$
) 0.37 g

The ingredients were dissolved and adjusted the volume to 1,000 mL with deionized water. Then, the solution was sterilized by autoclaving for 10 min at 121°C, 15 lb/square inches after preparation.

#### 3.3 Sodium acetate (3.0 M)

The chemical was dissolved in deionized water, adjusted to pH 5.2 with glacial acetic acid, and adjusted the volume to 100 mL with deionized water. Then, the solution was sterilized by autoclaving for 10 min at 121°C, 15 lb/square inches after preparation.

#### 3.4 RNAase (10 mg/mL)

RNAase 10.00 mg

The RNAase was dissolved in 10 mM Tris-HCl (pH 7.5), 15 mM NaCl and stored at -20°C.

3.5	3.5 Tris-borate (TBE) buffer (5X)			
	Tris Base	54.00	g	
	Boric acid	27.50	g	
	EDTA (C <sub>10</sub> H <sub>14</sub> N <sub>2</sub> O <sub>8</sub> Na <sub>2</sub> .2H <sub>2</sub> O)	0.37	g	

The ingredients were dissolved and adjusted the volume to 1,000 mL with deionized water.

### 3.6 Gel loading buffer (6X)

Bromophenol blue	25.00	g
------------------	-------	---

The dye was dissolved and adjusted the volume to 10 mL with 40% sucrose in water.

#### 3.7 Ethidium bromide (10 mg/mL)

Ethidium bromide (Sigma)	1.00	g
--------------------------	------	---

The chemical was dissolved and adjusted the volume to 10 mL with sterilized deionized water.

# **APPENDIX B**

# LOCATIONS AND DETAILS OF XYLARIACEOUS

# **COLLECTIONS**

Locations of xylariaceous collections and details of their morphological

characteristics were given in Tables 1B and 2B respectively.

Code	Location	Date
SUT001 - SUT007	Phu Luang, Nakhon Ratchasima Province	28 July 2003
SUT008 - SUT012	Nong Rawieng, Nakhon Ratchasima Province	9 August 2003
SUT013 - SUT021	Burirum Province	24 August 2003
SUT022 - SUT025	Chaiyaphum Province	22 August 2003
SUT026 - SUT076	Ratchaburi Province	28 August 2003
SUT077 - SUT082	Nong Rawieng, Nakhon Ratchasima Province	1 September 2003
SUT083 - SUT084	Bangkok	5 September 2003
SUT085 - SUT086	Yasothon Province	6 September 2003
SUT087 - SUT116	Songkhla Province	8 September 2003
SUT117 - SUT122	Petchaboon Province	10 September 2003
SUT123 - SUT152	Suranaree University of Technology, Nakhon	20-25 September 2003
	Ratchasima Province	
SUT153 - SUT169	Yasothon Province	15 November 2003
SUT170 - SUT191	Suranaree University of Technology, Nakhon	17 November 2003
	Ratchasima Province	
SUT192 - SUT268	Trad Province	19 November 2003
SUT269 - SUT271	Chiang Rai Province	10 December 2003
SUT272 - SUT323	Kanchanaburi Province	14 December 2003
SUT324 - SUT327	Chiang Rai Province	25 January 2003
SUT328 - SUT334	Chiang Mai Province	16 June 2002
SUT335 - SUT338	Nakhon Ratchasima Province	20 July 2002

**Table 1B.** Locations and collecting dates of xylariaceous collections.

Code	Species	Stromatal colour	KOH extracted pigments	Ascospore size (µm)
SUT001	H. purpureonitens	Blackish with reddish brown	Purple	8.8-10x2.5-5
SUT002	H. purpureonitens	Blackish with reddish brown	Purple	8.8-11.5x2.5-5
SUT003	H. purpureonitens	Blackish with reddish brown	Purple	NF
SUT004	H. purpureonitens	Blackish with reddish brown	Purple	3.8-5x7.5-10
SUT005	H. purpureonitens	Blackish with reddish brown	Purple	3.8-5x7.5-10
SUT006	<i>Xylaria</i> sp. nov.	Dark brown to black	Colorless	(7.5)8.8-10x3.8-5
SU1007	<i>Xylaria</i> sp. nov.	Dark brown to black	Colorless	8.8-10x3.8-5
SU1008	species 1 sp. nov.	Віаск	Greenish olivaceous	NF
SUT009	H. atroroseum	Brown vinaceous or chestnut	Greenish olivaceous	6.3-8.8x2.5-3.8
SUT010	H. atroroseum	Brown vinaceous or chestnut	Greenish olivaceous	5-6.3x2.5
SUT011	Xylaria sp.	Brownish black	Colorless	NF
SU1012	<i>Xylaria</i> sp. nov.	Dark brown to black	Colorless	(7.5)8.8-10x3.8-5
SUI013	Dalainia eschscholzii Valasia escaso	Brown vinaceous	Purple	11.3-13.8X3-0.3
SU1014	<i>Aylaria</i> sp. nov.	Dark brown to black	Colorless	8.8-10X5.8-5
501015	species 1 sp. nov.		Greenisn onvaceous	INF
SUT016	H. lenormandii	Grayish sepia	Red	10-12.5x5
SUT017	H. cf. ferrugineum	Hazel	Orange	12.5-15(17.5)x5-7.5
SUT018	H. lenormandii	Grayish sepia	Red	12.5-15x5-6.3
SU1019	<i>Eutypa</i> sp.	-	-	-
SU1020	H. cl. perforatum	Grayish sepia	Y ellowish brown	NF (7.5)9.9.105.6.2
SU1021	H. monticulosum	Brownish vinaceous to black	Colorless	(7.5)8.8-10x5-0.5
501022	H. lenormanali Var. microspora		Ked	5-782.5-5.8
SUT023	H. truncatum	Blackish brown with white fringe	Olivaceous	7.5-10x3.8-5
SUT024	H. stygium	Blackish with reddish brown	Greenish olivaceous	3.8-6.3x2.5-3.8
SU1025	H. bovei var. microspora	Black	Greenish olivaceous	7.5-10x3.8-5
SUT026	X. badia	Silvery brown	Colorless	10-12x3.8
SUT027	<i>Xylaria</i> sp. nov.	Dark brown to black	Colorless	8.8-10x3.8-5
SUT028	X. cf. <i>multiplex</i>	Blackish with light brown with peeling layer	Colorless	11.3-13.8x3.8-5
SUT029	X. muscula	White with black ostioles	Colorless	NF
SUT030	<i>Xylaria</i> sp. nov.	Black	Colorless	NF
SUT031	<i>Xylaria</i> sp. nov.	Dark brown to black	Colorless	8.8-10x3.8-5
SU1032	X. badia	Silvery brown	Colorless	9.8-12x3.8-5
SU1033	<i>Xylaria</i> sp. nov.	Dark brown to black	Colorless	(7.5)8.8-10x3.8-5
SU1034	<i>Xylaria</i> sp. nov.	Dark brown to black	Colorless	8.8-10X3.8-5
501055	A. cl. juruensis	peeling layer	Coloriess	(10)11.3-13.8x3.8-5
SUT036	X. ianthino-velutina	Black	Colorless	(7.5)8.8-10x3.8
SUT037	D. eschscholzii	Brown vinaceous	Purple	10-12.5x5-6.3
SU1038	D. eschscholzii	Brown vinaceous	Purple	10-13.8x3.8-5
SU1039	D. eschscholzu	Brown vinaceous	Purple	11.3-12.5x5-6.3
SU1040	H. fenaleri H. immentione	Brown vinaceous	Dull groop	(5)/.5-10X5./5
SU1041	H. monticulosum	Brownish vinaceous to block	Colorless	7.3-0.0X2.3-3.0 6 2 7 5x2 5 2
SUT042	H monticulosum	Brownish black	Durple	6375v7538
SUT043	H monticulosum	Brownish black	Purple	63-75x2.5-3.8
SUT045	H anthochroum	Brown vinaceous or chestnut	Dull green	8 8-11 25x3 8-5
SUT046	H. sublenormandii sp.	Reddish brown	Reddish brown	(8.8)12.5-15x5-6.3
SUT047	nov. A mirabilis	Black	Colorless	10-13-5
SU1047	A. mirabilis	Black	Colorless	10 12 5 2 8 5
SUT048	A mirabilis	Black	Colorless	10-12.3x3.0-3 (8.8)9-12 5v3 8-5
SUT050	A. mirubuis Not Yulariaceae	Black	Colorless	(0.0)9-12.5x5.0-5
SUT051	A mirahilis	Black	Colorless	$(8.8)9_{-13}5_{x3}8_{-5}$
SUT052	Not Xvlariaceae	-	Colorless	-
SUT053	Not Xylariaceae	_	Colorless	_
SUT054	A. mirabilis	Black	Colorless	NF
SUT055	A. mirabilis	Black	Colorless	10-13.8x5
SUT056	A. mirabilis	Black	Colorless	11.2-13.8x5
SUT057	A. mirabilis	Black	Colorless	10-13.8x3.8-5
SUT058	H. stygium	Blackish with reddish brown	Greenish olivaceous	3.8-6.3x2.5-3.8

 Table 2B.
 More details of xylariaceous collections.

 $\overline{NF} = Not found.$ 

Code	Species	Stromatal colour	KOH extracted pigments	Ascospore size (µm)		
SUT059	H. monticulosum	Brownish vinaceous to black	Purple	6.3-7.5x2.5		
SUT060	H. monticulosum	Brownish vinaceous to black	Purple	7.5-8.8x2.5-3		
SUT061	H. cf. fendleri	Brownish vinaceous	Orange	10-12.5x3.8-5		
SUT062	H. haematostroma	Orange red or rust	Orange red	NF		
SUT063	H investions	Brownish vinaceous	Dull green	$(5)75_88x(25)38_4$		
SUT064	H haematostroma	Orange red or rust	Orange red	(5)7.5-0.0X(2.5)5.0-1 NF		
301004	11. ndemalosiroma		Ofalige fed	141		
SUT065	H. lenormandii	Grayish sepia	Red	(8.8)10-12.5x3.8-5		
SUT066	H. kanchanabhisakii sp. nov.	Dull reddish brown	Reddish brown	(8.8)10-11.3x3.8-5		
SUT067	<i>H. kanchanabhisakii</i> sp. nov.	Dull reddish brown	Reddish brown	(7.5)10-11.3x3.8		
SUT068	H. kanchanabhisakii sp. nov.	Dull reddish brown	Reddish brown	10-11.3(13)x(2.5) 3.8-5		
SUT069	H. kanchanabhisakii sp. nov.	Dull reddish brown	Reddish brown	10-11.3x3.8-5		
SUT070	H of ferrugineum	Brown vinaceous	Orange	(5)16 5-17 5x6 6-7 4		
SUT070	H kanchanabhisakii sp	Dull reddish brown	Peddish brown	$(5)10.5 17.5 \times 0.0 7.5$ 10 11 3(12 5) $\times 3.8 5$		
301071	nov.			(12 5)11 2 10 2 2 5		
SU1072	H. kanchanabhisaku sp. nov.	Dull reddish brown	Reddish brown	(12.5)11.3-10x3.8-5		
SUT073	H. monticulosum	Brownish vinaceous to black	Colorless	7.5-8.8x3.8		
SUT074	X. mellisii	Blackish with gray to brown outer peeling layer	Colorless	NF		
SUT075	<i>Xylaria</i> taxonomic species 1	Dark brown to black	Colorless	12.5-15(16.5)x5-6.3		
SUT076	X hadia	Silvery brown	Colorless	9 8-12x3 8-5		
SUT077	V neidii	Black	Colorless	2 2 2 2 2 5		
SU1077	A. psian V. hugohista	Diack Brown outer posling lover	Colorlass	0.0-3.0-3		
501078	x. brachiata	Brown outer peeling layer	Coloriess	(8.8)10- 11.3(12.5)x3.8-5		
SUT079	H. cf. archeri	Blackish brown	Hazel	8.8-10x3.8-5		
SUT080	H. monticulosum	Brownish vinaceous to black	Colorless	7.5-8.8x3.8-5		
SUT081	Hypoxylon taxonomic species 1 sp. nov.	Black with shinny	Greenish olivaceous	7.5-10x3.8-5		
SUT082	Hypoxylon taxonomic species 2	Brown vinaceous	Yellowish brown	(8.8)11.3- 12.5(17.5)x5-7.5		
SUT083	Xvlaria sp. nov.	Dark brown to black	Colorless	7.5-8.8x3.8		
SUT084	D eschscholzii	Brown vinaceous	Purple	10-13 8 x 5-7 5		
SUT085	D. eschscholzii	Brown vinaceous	Purple	10-13 5 x 3 8-6 3		
SUT085	D. eschscholzii	Brown vinaceous	Durple	0514x2862		
501080	D. eschscholzh	Brown vinaceous	Pulple	9.3-14X3.8-0.3		
SUT087	Xylaria sp. nov.	Dark brown to black	Colorless	7.5-8.8(10)x3.8		
SU1088	X. cf. juruensis	Blackish with light brown with peeling layer	Colorless	12.5-15x3.8-5		
SUT089	X. cubensis	Bronze becoming dark with age	Colorless	7.5-8.8x3.8		
SUT090	X. cubensis	Bronze becoming dark with age	Colorless	7.5-8.8x3.8		
SUT091	X. ianthino-velutina	Black	Colorless	(7.5)8.8- 10(12.5)x3.8-5		
SUT092	X. beccari	Brownish black	Colorless	(5)6.3-7.5x2.5		
SUT093	Xylaria sp. nov	Dark brown to black	Colorless	(7.5)8 8-10x3 8-5		
SUT093	H monticulosum	Brownish vinaceous to black	Purnle	7 5_8 8v3 8		
SUT095	H. subgilvum var.	Dark brick	Orange	(5)6.5-8.8x2.5		
SUTOOS	R cannodas	Black	Colorlaga	NE		
SUT098 SUT097	B. capnodes B. capnodes	Black	Colorless	иг 10-12.5(-13.8)х6.3-		
SUT098	H. urceolatum	Black	Colorless	7.5 10-12.5x2.5-5		
SUT099	C tinctor	Black	Colorless	NF		
SUT100	H nurnuraonitans	Blackish with reddish brown	Dumla	75_10+255		
SUT100	II. purpureonnens	Diackish with reduish brown	Colorier	1.J-10A2.J-J 0.0.102.0.5		
SU1101	<i>Kretzschmaria</i> sp.	BIACK	Colorless	8.8-10x3.8-5		
SUT102	K. procera	Black	Colorless	(70)100- 135(162.5)x8.8-15		
SUT103	H. cf. archeri	Blackish brown	Hazel	8.8-10x2.5-5		
SUT104	H. subgilvum var. microsporum	Dark brick	Orange	(3.8)5-7.5x2.5-3.8		
	II of another	Diagleich heaven	Hazel	8 8-10x3 8-5		
SUT105	H CL archeri	DIACKISH DIOWH				

Code	Species	Stromatal colour	KOH extracted pigments	Ascospore size (µm)
SUT107	H. cf. fendleri	Brownish vinaceous	Orange	NF
SUT108	H. subgilvum var. microsporum	Dark brick	Orange	(2.5)6.3-8.8x2.5-3
SUT109	R. procera	Black	Colorless	(77.5)90-117.5x10- 12.5
SUT110	Nemania sp.	Black	Colorless	NF
SUT111	Nemania sp.	Black	Colorless	NF
SUT112	H. cf. archeri	Blackish brown	Hazel	8-10x3.8-5
SUT113	R. procera	Black	Colorless	(70)100-112.5x10- 12.5
SUT114	R. procera	Black	Colorless	(65)95-125x10-15
SUT115	H. monticulosum	Brownish vinaceous to black	Purple	6.3-7.5x2.5-3.8
SUT116	H. monticulosum	Brownish vinaceous to black	Purple	(6.3)7.5- 8.8(11.3)x3.8
SUT117	X. scrupora	Yellowish brown to dark brown	Colorless	17.5- 21.3(22.5)x(5)6.3-7.5
SUT118	Xvlaria sp. nov.	Dark brown to black	Colorless	10-11.5x3.2-3.7
SUT119	X. anisopleura	Dark brown to dull black	Colorless	8.8-10x3.8-5
SUT120	H. cf. fendleri	Brownish vinaceous	Orange	(8.8)10-12.5x3.8-5
SUT121	H. anthochroum	Brown vinaceous or chestnut	Olivaceous	10.8-13(14)x4-6
SUT122	Biscogniauxia sp.	Black	Colorless	9.2-11.9 x 5.4-6.7
SUT123	X. ianthino-velutina	Black	Colorless	7.5-8.8(10)x3.8-5
SUT124	X. psidii	Black	Colorless	(7.5)8.8-10x3.8-5
SUT125	X. psidii	Black	Colorless	(7.5)8.8-10x3.8-5
SUT126	X. psidii	Black	Colorless	7.5-8.8(10)x3.8-5
SUT127	Xylaria species 2	Dark brown to black	Colorless	(8.8)10- 11.3(12.5)x2.5-3.8
SUT128	Xylaria species 2	Dark brown to black	Colorless	8.8-10x3.8-5
SUT129	X. juruensis var.	Blackish with brown peeling outer	Colorless	(7.5)10-2.5x3.8
CUT120	microspora	layer	Calaria	9 9 10-2 9 5
SUT130 SUT131	<i>Xylaria</i> species 2 <i>Xylaria</i> sp. nov.	Dark brown to black	Colorless	8.8-10x3.8-5 10-11.3(13.8)x3.2-
SUT132	Vularia species ?	Dark brown to black	Colorless	5.7 8 8-10x3 8-5
SUT132	<i>Xylaria</i> sp. pov	Dark brown to black	Colorless	8 8-10x3 8-5
SUT134	Xylaria species 2	Dark brown to black	Colorless	$10-11 3x^2 5-3 8$
SUT135	Xylaria species 2 Xylaria sp. (Immature)	Dark brown to black	Colorless	NF
SUT136	Xylaria sp. nov.	Dark brown to black	Colorless	(7.5)8 8-10x3 8-5
SUT137	X. juruensis var.	Blackish with brown peeling outer	Colorless	(7.5)10-11.3x3.8-5
SUT138	X. juruensis var.	Blackish with brown peeling outer	Colorless	8.8-10x3.8-5
SUT139	X. juruensis var.	Blackish with brown peeling outer	Colorless	8.8-10x3.8-5
SUT140	X. cf. juruensis	Blackish with brown peeling outer	Colorless	12.5-13.8(15)x3.8-5
SUT141	Xylaria sp. nov	Dark brown to black	Colorless	(7.5)8.8-10x2 5-3 8
SUT142	X. badia	Silvery brown	Colorless	(8.8)10-11.3x3.8
SUT143	Xylaria sp. nov.	Dark brown to black	Colorless	8.8-10x3.8-5
SUT144	H. lenormandii	Gravish sepia	Red	10-11.3x3.8-5
SUT145	H. cf. fendleri	Brownish vinaceous	Orange	8.8-11.3x3.8-5
SUT146	H. rubiginosum	Brown vinaceous	Yellowish brown	10-11.2x5-6.3
SUT147	H. lenormandii	Grayish sepia	Red	10-12.5x3.8-5
SUT148	H. rubiginosum	Brown vinaceous	Yellowish brown	10-11.3x3.8-5
SUT149	H. rubiginosum	Brown vinaceous	Yellowish brown	11.3-12.5x5-6.3
SUT150	H. sublenormandii sp. nov.	Reddish brown	Orange	8.8-10x3.8
SUT151	H. lenormandii	Grayish sepia	Red	11.3-12.5x3.8-5
SUT152	H. cf. fendleri	Brown	Orange	8.8-10x3.8-5
SUT153	H. anthochroum	Brown vinaceous or chestnut	Dull green	NF
SUT154	H. anthochroum	Brown vinaceous or chestnut	Dull green	10-12.5x3.8-5
SUT155	Xylaria sp. nov.	Dark brown to black	Colorless	7.5-8.8x2.5-3.8

Code	Species	Stromatal colour	KOH extracted pigments	Ascospore size (µm)
SUT156	B. capnodes	Black	Colorless	12.5-15(18.8)x6.3- 7.5
SUT157	H. rubiginosum	Brown vinaceous	Yellowish brown	10-11.3x3.8-5
SUT158	Hypoxylon taxonomic species 2	Dark brick to brown vinaceous	Yellowish brown	10-11.3x3.8-5
SUT159	H. cf. fendleri	Brownish vinaceous	Orange	(8.8)10-11.3(15)x3.8-
SUT160	H. purpureonitens	Blackish with reddish brown	Purple	10-11.3x3.8-5
SUT161	C. tinctor	Black	-	15-17.5x6.3-7.5
SUT162	H. cf. fendleri	Brownish vinaceous	Orange	8.8-10x3.8-5
SUT163	H. cf. fendleri	Brownish vinaceous	Orange	10-12.5x3.8-5
SUT164	H. haematostroma	Orange red or rust	Orange red	15-16.3x6.3-7.5
SUT165	H. cf. fendleri	Brownish vinaceous	Orange	10-12.5x3.8-5
SUT166	H. hypomiltum	Blackish with brown peeling outer layer	Yellowish brown	7.5-2.5x3.8
SUT167	H. purpureonitens	Blackish with reddish brown	Purple	(6.3)7.5-10x3.8
SUT168	D. eschscholzii	Brown vinaceous	Purple	10-12.5x5-6.3
SUT169	D. eschscholzii	Brown vinaceous	Purple	11.3-12.5x5-6.3
SUT170	X. cf. juruensis	Blackish with brown peeling outer layer	Colorless	11.3-12.5x3.8-5
SUT171	Xylaria species 2	Dark brown to black	Colorless	10-11.3x2.5-3.8
SUT172	<i>Xylaria</i> sp. nov.	Dark brown to black	Colorless	(6.3)7.5-8.8x3.8
SUT173	<i>Xylaria</i> sp. nov.	Dark brown to black	Colorless	7.5-10x3.8-5
SUT174	X. cf. apiculata	Black	Colorless	7.5-8.8x3.8
SUT175	X. brachiata	Brown outer peeling layer	Colorless	10-12.5x3.8-5
SUT176	X. cf. apiculata	Black	Colorless	8.8-10x3.8-5
SU11//	X. maitlandii	layer	Colorless	8.8-10x3.8-5
SUT178	D. eschscholzii	Brown vinaceous	Purple	10-12.5(15)x5-6.3
SUT179	H. monticulosum	Brownish vinaceous to black	Colorless	7.5-8.8(10)x2.5
SUT180	H. lenormandii	Grayish sepia	Red	10-12.5x3.8-5
SUI181	H. lenormandu	Grayish sepia	Ked Vallassiah ananaa	10-12.5X3.8-5
SUI182	H. suranarii sp. nov.	Dark brown to black	Yellowish orange	12.5-13.8X5-0.5
SUI185 SUIT184	H. suranarii sp. nov.	Dark brown to black	Vellowish orange	(10)12513.033-0.5
SUT184	H monticulosum	Brownish vinaceous to black	Durple	(10)12.3-13.6x3-0.3 6 3 7 5(8 8) $\times$ 2 5 3 8
SUT185	H cf fendleri	Brownish vinaceous	Orange	10-11 3x5-6 3
SUT187	H trugodes	Senia	Yellow	10-11 3x5-6 3
SUT188	Hypoxylon taxonomic species 1 sp. nov	Black with shinny	Greenish olivaceous	7.5-8.8x2.5-3.8
SUT189	H. monticulosum	Brownish vinaceous to black	Purple	7.5-8.8x2.5-3.8
SUT190	H. cf. fendleri	Brownish vinaceous	Orange	6.3-7.5x2.5
SUT191	H. cf. fendleri	Brownish vinaceous	Orange	15-16.3x6.3-7.5
SUT192	X. mellisii	Black	Colorless	12.5-15x3.8-5
SUT193	X. cubensis	Bronze becoming dark with age	Colorless	7.5-8.8x3.8
SUT194	X. cubensis	Bronze becoming dark with age	Colorless	7.5-8.8x3.8
SUT195	Xylaria species 2	Dark brown to black	Colorless	10-11.3x2.5-3.8
SUT196	X. anisopleura	Dark brown to dull black	Colorless	(20)23.8- 25(27.5)x7.5-8.8
SUT197	Xylaria sp. nov.	Dark brown to black	Colorless	6.3-7.5(8.8)x3.8
SUT198	<i>Xylaria</i> sp. nov.	Dark brown to black	Colorless	(6.5)7.5-8.8(10)x3.8
SUT199	X. cubensis	Bronze becoming dark with age	Colorless	(6.2)7.5-8.8x3.8
SUT200	<i>Xylaria</i> sp. nov.	Dark brown to black	Colorless	7.5-8.8x3.8
SUT201	X. schweinitzii	Brownish black to dull black	Colorless	18.8-21.3x6.3-7.5
SUT202	X. cubensis	Bronze becoming dark with age	Colorless	6.3-7.5x3.8
801203	<i>Xylaria</i> taxonomic species 2	Віаск	Colorless	(7.5)8.8-10x3.8
SUT204	<i>Xylaria</i> taxonomic species 3	Black	Colorless	6.3-7.5x2.5-3.8
SUT205	X. anisopleura	Dark brown to dull black	Colorless	(17.5)18.8-2.3x6.3- 7.5
SUT206	X. telfairii	Pale yellow, clay-colored to orange brown	Colorless	17.5-20x5-6.3
SUT207	<i>Xylaria</i> taxonomic species 4	Copper- to bronze-colored to brown	Colorless	21.3-25x8.8-10

 $\overline{NF} = Not found.$ 

Code	Species	Stromatal colour	KOH extracted	Ascospore size (µm)
CLIT200	V ania antauna	Doub because to dull block	Colorlass	20.21.5.7.5.9
SU1208	A. anisopieura	Dark brown to dull black	Dumlo	20-21.5X7.5-8
SU1209	D. eschscholzh	Brown vinaceous	Purple	NF 11 2 12 5
SU1210	B. caphodes	Black	Colorless	11.5-12.5x0.5-7.5
SU1211	C. IInclor B. campadag	Diack	Colorless	10, 12, 5(12, 9) = 6, 2
501212	B. capnoaes	Бласк	Coloriess	10-12.5(15.8)x6.3- 7.5
SUT213	H. nitens	Black with shinny	Greenish olivaceous	7.5-9.5x3.8-5
SUT214	H. atroroseum	Brown vinaceous or chestnut	Greenish olivaceous	3.8-5x1.5-2.5
SUT215	H. rubiginosum	Reddish brown	Yellowish brown	8.8-10x3.8-5
SUT216	H. moriforme	Black	Greenish olivaceous	7.5-8.8x3.8-5
SUT217	H. rubiginosum	Reddish brown	Yellowish brown	(7.5)8.8-10x3.8-5
SUT218	H. cf. perforatum	Brown vinaceous	Yellowish brown	8.8-10(11.3)x3.8-5
SUT219	H. atroroseum	Blackish brown	Greenish olivaceous	5-6.3x2.5
SU1220	H. moriforme	Black with shinny	Greenish olivaceous	7.5-8.8x3.8
SU1221	H. rubiginosum	Reddish brown	Yellowish brown	7.5-8.8(10)x3.8
SU1222	H. stygium	Blackish with reddish brown	Greenish olivaceous	5-6.3x2.5
SU1223	H. duranii	Brown vinaceous or chestnut	Reddish brown	8.8-10x3.8-5
SU1224	H. auranii	Brown vinaceous or chestnut	Reddish brown	8.8-10x3.8-5(6.3)
SU1225	H. monticulosum	Brownish vinaceous to black	Purple	$0.3 - 7.3 \times 2.3(3.8)$
SU1220	H. siygium	Brownish vinacoous to block	Colorloss	3-0.3X2.3
SU1227	H. monticulosum	Brownish vinaceous to black	Colorless	0.5-7.5(0.0)X2.5-5.0 NE
SUT228	H stygium	Blackish with reddish brown	Greenish olivaçãous	5 6 3x2 5
SUT229	H stygium	Blackish with reddish brown	Greenish olivaceous	5-6 3x2 5
SUT230	H stygium	Blackish with reddish brown	Greenish olivaceous	5-6 3x2 5-3
SUT232	H monticulosum	Brownish vinaceous to black	Colorless	7 5-8 8x2 5-3 8
SUT232	H anthochroum	Brown vinaceous or chestnut	Dull green	10-11 3x3 8-5
SUT234	H suboilvum	Hazel to dark brick	Orange	8 8-10x 3 8-5
SUT235	H monticulosum	Brownish vinaceous to black	Colorless	NF
SUT236	Hyporylon taxonomic	Black	Greenish olivaceous	7 2-9 8x3 1-4 4
501200	species 1 sp. nov.	Ditter		
SUT237	H. cf. ferrugineum	Brown vinaceous to rusty brown	Orange	(12.5)13.4-17.8x5.3- 8 3
SUT238	<i>Hypoxylon</i> taxonomic species 1 sp. nov.	Black with shinny	Greenish olivaceous	7.9-9.1x3.2-4.1
SUT239	H. duranii	Brown vinaceous or chestnut	Reddish brown	8.5-10.8x4-5.6
SUT240	H. anthochroum	Brown vinaceous or chestnut	Dull green	10.8-13(14)x4-6
SUT241	Hypoxylon taxonomic species 1 sp. nov.	Black with shinny	Greenish olivaceous	7.5-8.9x2.8-4
SUT242	H. bovei var.	Black with shinny	Greenish olivaceous	7.3-9x3.1-4
SUT243	H. stygium	Blackish with reddish brown	Greenish olivaceous	4.7-6.5x1.8-2.4
SUT244	Hypoxylon taxonomic	Black with shinny	Greenish olivaceous	7.6-9.1x2.8-4.2
SUT245	H stygium	Blackish with reddish brown	Greenish olivaceous	5.5-6.4x1 7-2.4
SUT246	Hypoxylon taxonomic	Black with shinny	Greenish olivaceous	7.3-8.7x3-4
SUT247	H stygium	Blackish with reddish brown	Greenish olivaçãous	16-59-1876
SUT247	H duranii	Brown vinaceous or chestnut	Reddish brown	$(6.7)8.1_{-9.5x4}3_{-5}$
SUT248	H stygium	Blackish with reddish brown	Greenish olivaceous	7 5-8 8x3 1-4 4
SUT250	H. sublenormandii sp.	Reddish brown	Reddish brown	8.9-11.3x3.4-4.7
SUT251	Hypoxylon taxonomic species 1 sp. nov	Black	Greenish olivaceous	7-8.2x3.4-4
SUT252	H. duranii	Brown vinaceous or chestnut	Reddish brown	8.5-10.4x4.5-5.5
SUT253	H. stygium	Blackish with reddish brown	Greenish olivaceous	4.8-5.9x1.8-2.2
SUT254	H. duranii	Brown vinaceous or chestnut	Reddish brown	8.2-9x(3.9)4.4-5.4
SUT255	Hypoxylon taxonomic species 1 sp. nov	Black with shinny	Greenish olivaceous	6.6-8x3.4-4.6
SUT256	H. brevisporum	Brown vinaceous or chestnut	Hazel	6.1-7.2x2.7-3.7
SUT257	H. stygium	Blackish with reddish brown	Greenish olivaceous	5.6-6.3x2.1-2.8
SUT258	Nemania sp.	Black	Colorless	8.9-11.7x4.7-6
SUT259	H. duranii	Brown vinaceous or chestnut	Reddish brown	9-10.5x4.6-5.5
SUT260	C. tinctor	Black	Colorless	13.3-18.4x5.5-7.3
SUT261	Nemania species	Black	Colorless	11.8-14.9x7.3-8.7

Code	Species	Stromatal colour	KOH extracted pigments	Ascospore size (µm)
SUT262	H. purpureonitens	Blackish with reddish brown	Purple	8.6-12.3x3.6-4.6
SUT263	H. anthochroum	Brown vinaceous or chestnut	Dull green	9.7-12.2x4-5.4
SUT264	H. monticulosum	Brownish vinaceous to black	Purple	5.8-7.2x2.4-4.1
SUT265	H. monticulosum	Brownish vinaceous to black	Purple	6.8-9x2.8-3.9
SUT266	H. monticulosum	Brownish vinaceous to black	Purple	6.9-8.5x3-3.6
SUT267	H. monticulosum	Brownish vinaceous to black	Purple	NF
SUT268	D. eschscholzii	Brown vinaceous	Purple	10.8-13.2x5.8-6.6
SUT269	<i>Xylaria</i> sp. nov.	Dark brown to black	Colorless	8.9-10.3x3.9-5
SUT270	X. cubensis	Bronze becoming dark with age	Colorless	7.5-8.8x3.8-5
SUT271	X. cubensis	Bronze becoming dark with age	Colorless	(6.3)7.5-8.8x3.8-5
SUT272	<i>Xylaria</i> sp. nov.	Dark brown to black	Colorless	7.5-9x4.1-4.9
SUT273	Xylaria sp. nov.	Dark brown to black	Colorless	7.1-9x3.6-4.8
SUT274	Xylaria species 2	Dark brown to black	Colorless	9.5-12.1x3.4-4.5
SUT275	Xylaria sp. nov.	Dark brown to black	Colorless	7.6-8.8x4-5
SUT276	Xylaria taxonomic species 5	Dark brown to black	Colorless	12.5-14.7x4.5-6.2
SUT277	X. cubensis	Bronze becoming dark with age	Colorless	7.5-9x3.8-5
SUT278	D. eschscholzii	Brown vinaceous	Purple	10-12.3x4.7-6.1
SUT279	H. cf. fendleri	Brownish vinaceous	Orange	NF
SUT280	H. cf. fendleri	Brownish vinaceous	Orange	8.4-10.9x3.9-5
SUT281	H. duranii	Brown vinaceous or chestnut	Reddish brown	9.7-11.7x4.5-5.5
SUT282	H. sublenormandii sp.	Reddish brown	Reddish brown	9-11.8x4.5-5.1
SUT283	H. lenormandii	Gravish sepia	Red	9.5-11.9x4.6-5.9
SUT284	H duranii	Brown vinaceous or chestnut	Reddish brown	9-10 9x4-5 3
SUT285	Hypoxylon taxonomic species 1 sp. poy	Black with shinny	Greenish olivaceous	6.1-9x2.3-4
SUT286	H monticulosum	Brownish vinaceous to black	Purple	NF
SUT287	H monticulosum	Brownish vinaceous to black	Purple	6 8-8 4x 3 3-4
SUT288	H nitens	Black with shinny	Greenish olivaceous	7 2-8 6x3 1-4 3
SUT289	H monticulosum	Brownish vinaceous to black	Colorless	NF
SUT290	Riscogniauxia sp	Black	Colorless	9 2-11 9x5 4-6 7
SUT291	H cf. fendleri	Brownish vinaceous	Orange	8 8-10x3 8-5
SUT292	H haematostroma	Orange red or rust	Orange red	13-15 2x6 3-7 9
SUT293	H. haematostroma	Orange red or rust	Orange red	15.6-17.9x7.1-8.6
SUT294	H ruhiginosum	Brown vinaceous	Yellowish brown	8 8-11 3x5
SUT295	H. monticulosum	Brownish vinaceous to black	Purple	6.3-7.5x2.5-3.8
SUT296	H. rubiginosum	Brown vinaceous	Yellowish brown	(8.8)10-11.3x5-6.3
SUT297	H. rubiginosum	Brown vinaceous	Yellowish brown	8.8-10x3.8-5
SUT298	H. nitens	Black with shinny	Greenish olivaceous	10-12.5x3.8-5
SUT299	H. nitens	Black with shinny	Greenish olivaceous	10-11.3(12.5)x3.8- 5(6.3)
SUT300	H. lenormandii	Gravish sepia	Red	8.8-10x2.5-3.8
SUT301	H. monticulosum	Brownish black to black	Colorless	6.3-7.5x3.8-5
SUT302	H. monticulosum	Brownish black to black	Colorless	6.3-7.5x3.8
SUT303	H. sublenormandii sp.	Brownish black to black	Reddish brown	8-10x3.8-5
SUT304	H. sublenormandii sp.	Brownish black to black	Reddish brown	8-10x3.8-5
SUT305	H. sublenormandii sp.	Brownish black to black	Reddish brown	8-10x3.8-5
SUT306	H. duranii	Brown vinaceous or chestnut	Reddish brown	9-10.9x4-5 3
SUT307	H lenormandii	Gravish senia	Red	8 8-10x2 5-3 8
SUT308	H. cf. fendleri	Brownish vinaceous	Orange	8.8-10x3 8-5
SUT309	X badia	Silvery brown	Colorless	8 8-11 3x3 8-5
SUT310	X. badia	Silvery brown	Colorless	8.8-11.3x3 8-5
SUT311	H. sublenormandii sp.	Reddish brown	Reddish brown	(10)11.3-13.8x5-7.5
SUT312	H. lenormandii	Gravish sepia	Red	11.3-12.5x3.8-5
SUT313	H. lenormandii	Gravish sepia	Red	8 8-11 3x3 8-5
SUT314	H fendleri	Brownish vinaceous	Orange	8 8-12 5x5-6 3
SUT315	H. monticulosum	Brownish vinaceous to black	Pumle	6.3-7 5x3 8-5
SUT316	H. nitens	Black	Greenish olivaceous	10-12.5x3 8-5(6 3)
SUT317	H. nitens	Black	Greenish olivaceous	$10-12.5 \times 3.8-5(6.3)$
SUT318	H. purpureonitens	Blackish with reddish brown	Purple	8.8-12.5x3.8-5

Code	Species	Stromatal colour	KOH extracted pigments	Ascospore size (µm)
SUT319	H. purpureonitens	Blackish with reddish brown	Purple	8.8-11.5x2.5-5
SUT320	H. rubiginosum	Brown vinaceous	Yellowish brown	8.8-10x3.8-5
SUT321	<i>Xylaria</i> sp. nov.	Dark brown to black	Colorless	8.9-10.5x3.8-5
SUT322	Xylaria sp. nov.	Dark brown to black	Colorless	8.9-10.5x3.8-5
SUT323	Xylaria species 2	Dark brown to black	Colorless	10-11.3x2.5-3.8
SUT324	X. cubensis	Bronze becoming dark with age	Colorless	7.5-8.8x3.8
SUT325	H. monticulosum	Brownish vinaceous to black	Purple	6.9-8.5x3-3.6
SUT326	D. eschscholzii	Brown vinaceous	Purple	10-13.5 x 3.8-6.3
SUT327	H. stygium	Blackish with reddish brown	Greenish olivaceous	7.5-8.8x3.1-4.4

# **APPENDIX C**

## NUCLEOTIDE SEQUENCE DATA

Nucleotide sequence results of 18S rDNA and ITS1-5.8S-ITS2 regions were presented in dendrogram for example in Figure 1C using ITS5 primer.

TG TCA A CA 1A C CA GA C G T TGC C TC G GCA G GC C GC G C GC C A C C TC TC TC A G G G G C G C G C G C G C G C C A C G C T C 70 C C GC C G GC 140 <sup>T</sup> T G A 200 210 TC TC TTGGTTC TGGCA TCGA 220 230 GAAGAAC GCAGC GAAA 250 260 270 280 290 300 A T G T GA A T T GCA GA A T TCA G T GA A T C GA A T C T T TGA AC GC A C A T T GC GC C C A T T an <mark>n</mark>a T 330 340 350 TC GA GC G TCA T T TCAAC C C T TAA GC C 310 TAGTGGGCA 360 370 380 T T A G T G T T G G G A G C C T A C G G A G A C G T A Gl TGCTGC 410 420 CGGA GTCG GtAC GCAC TC TA 390 CC 400 CAAA G TTA GT G 430 440 450 460 A C G TA G TAA T TA T C TA T C TC G C C T G T G A G C C G G A C C G G t C сста O TA G C T 460 470 GtCCCTG CGtAAA 500 510 520 GTA GAA TA C C GC T G A C T A g C A TATCAA T A G CG 480 CACA TCTAA 490 GAICCITC 0.0 GΤ GA TCA 520 FCAATA GCG

Figure 1C. Sequence electrophenogram of ITS1-5.8S-ITS2 region of Astrocystis mirabilis (SUT051) using ITS5 primer.

ССССААЛАСТ СССАТСАСТ САТТАЛАТСА СТТАТТСТТТ АТТГСАТАСТ АССТТАСТАС АТССАТАСС СТССТАЛАТС ТАСАС СССССАЛАСТ СССАТСАСТ САТТАЛАТСА СТТАТССТТТ АТТГСАТАСТ АССТТАСТАС АТССАТАСС СТССТАЛАТС ТАСАС СССССАЛАСТ СССАТССС САТТАЛАТСА СТТАТССТТТ АТТГСАТАСТ АССТТАСТАС ТГССАТАСС СТССТАЛАСС СТССАТАСС СССССАЛАСТ СССАТССС САТТАЛАТСА СТТАТССТТТ АТТГСАТАСТ АССТТАСТАС ТГССАТАСС СТССАТАСС СССССАЛАСТ СССАТССС САТТАЛАТСА СТТАТССТТТ АТТГСАТАСТ АССТТАСТАС СТССАТАСТ СТССАТАСС СТССАТАСС СССССАЛАСТ СССАТССС САТТАЛАТСА СТАТСССТТА АТТГСАТАСТ АССТТАСТАС СТССАТАСТ СТССАТАСС СТССАТАСТ СТАСАСТ СССССАЛАСТ СССАТСАСТ САТТАЛАТСА СТАТССТТА АТТГСАТАСТ АССТТАСТАС СТССАТАСТ СТССАТАСТ СТССАТАСТ СТССАТАСТ САССТАСТАСТ SUT051 SUT056 ST2301 90 90 90 90 AY083804 1 AY083805 1 TCACGGAGGG ATGTATTTAT TCACGGAGGG ATGTATTTAT TCACGGAGGG ATGTATTTAT TCACGGAGGG ATGTATTTAT TCACGGAGGG ATGTATTTAT ТАСАТТАААА АССАЛОССС СТСССССС ТАСАТТАААА АССАЛОССС СТССССССС CTCCCCCCTT CTCCCCCCCTT CTCCCCCCCTT SUT051 SUT056 ST2301 ACATGCI ACATGCI ACATGCI ACATGCI 91 91 91 91 91 -AA AAATCCCGAC -AA AAATCCCGAC -AA AAATCCCGAC -GA AAATCCCGAC AY083804 AY083805 179 TAT TAGAT 
 SUT051
 180

 SUT056
 180

 ST2301
 180

 AY083804
 180

 AY083805
 181
 280 290 300 310 320 33 ACAACGGTA ACCACGGTT ACCCCCACACA ACCCCTACT ACAACGGTA ACCACGGTT ACCCCTCAC COCCACACA ACCCCTACT ACAACGGTA ACCACGGTT ACCCCTCAC COCCACACA GACCCTACA AACCCCTACT ACAACCGGTA ACCACGGTT ACCCCTCAC COCCACACAG GACCCTCACA AACCCCTACT CAACCCGTA ACCACGGTT ACCCCTCACC COCCACACAG GACCCTCACA AACCCCTACT CAACCCGTA ACCGACGGTT ACCCCTCACC COCCACACAG GACCCTCACA AACCCCTACT 
 SUT051
 270

 SUT056
 270

 ST2301
 270

 AY083804
 270

 AY083805
 271
 370 380 390 400 410 420 430 440 45 TACCAATCC CGACACGGG AGGTAGTGAC AATAAATACT GATACAGGC TCTTTTGGGT CTTGTAATTG GAATGAGTAC AATTAAAT TACCAATCC CGACACGGG AGGTAGTGAC AATAAATACT GATACAGGGC TCTTTTGGGT CTTGTAATTG GAATGAGTAC AATTAAATACT TACCAATCC CGACACGGGG AGGTAGTGAC AATAAATACT GATACAGGGC TCTTTTGGGT CTTGTAATTG GAATGAGTAC AATTAAATC TACCAATCC CGACACGGGG AGGTAGTGAC AATAAATACT GATACAGGGC TCTTTTGGGT CTTGTAATTG GAATGAGTAC AATTAAATC TACCAATCC CGACACGGGG AGGTAGTGAC AATAAATACT GATACAGGGC TCTTTTGGGT CTTGTAATTG GAATGAGTAC AATTAAATC 360 360 360 360 361 507051 SUT056 ST2301 AY083804 AY083805 460 470 480 490 500 510 520 530 54 CCTTAACGAE GAACAATIGE AGGEAAGET TGGTGCCAEC AGCCCCGGTA A-TTCACTT CAATAGCGTA TATTAAGTT GGTGCAGTA CCTTAACGAE GAACAATIGE AGGECAAGET TGGTGCCAEC AGCCCCGGGTA A-TTCACTT CAATAGCGTA TATTAAAGTT GGTGCAGGTA CCTTAACGAE GAACAATIGE AGGECAAGET TGGTGCCAEC AGCCCCGGGTA ATTCCACTC CAATAGCGTA TATTAAAGTT GGTGCAGGTA CCTTAACGAE GAACAATIGE AGGECAAGET TGGTGCCAEC AGCCCCGGGTA ATTCCACTC CAATAGCGTA TATTAAAGTT GGTGCAGGTA CCTTAACGAE GAACAATIGE AGGECAAGET TGGTGCCAEC AGCCCCGGGTA ATTCCACTC CAATAGCGTA TATTAAAGTT GGTGCAGGTA CCTTAACGAE GAACAATIGE AGGECAAGET TGGTGCCAEC AGCCCCGGGTA ATTCCACTC CAATAGCGTA TATTAAAGTT GGTGCAGGTA SUT051 SUT056 ST2301 449 449 449 450 451 AY083804 AY083805 550 560 570 580 590 600 610 620 AMA-GCTOGT AGTTGAACCT TGGGCCTGGC TGGCCGGTC- GCCTCAACGC GTGCACTGGT TCGGCCGGGC CTTTCCCTTT AAA-GCTCGT AGTTGAACCT TGGGCCTGGC TGGCCGGTC- GCCTCAACGC GTGCACTGGT TCGGCCGGGC CTTTCCCTTT AAA-GCTCGT AGTTGAACCT TGGGCCTGGC TGGCCGGGC GCCCCACGC GTGCACTGGT TCGGCCGGGC CTTTCCCTCT AAAA-GCTCGT AGTTGAACCT TGGGCCTGGC TGGCCGGTCC GCCTCACCGC GTGCACTGGT TCGGCCGGGC CTTTCCCTCT AAAAGCTCGT AGTTGAACCT TGGGCCTGGC TGGCCGGTCC GCCTCACCGC GTGCACTGGT TCGGCCGGGC CTTTCCCTCT AAAAGCTCGT AGTTGAACCT TGGGCCTGGC TGGCCGGTCC GCCTCACCGC GTGCACTGGT TCGGCCGGGC CTTTCCCTCT SUT051 538 SUT056 538 ST2301 539 ▲¥083804 540 ▲¥083805 541 640 650 660 670 690 690 700 710 721 ATGCCTTCA CTGGCTGTAC TGGCGAACA GCACTITTAC TGGAAAAA TTAGAGTGTT CAAACAAGC CTATGCTCGA ATACATCAGC ATGCCTTCA CTGGCTGTAC TGGCAACCA GCACTITTAC TGGGAAAAA TTAGAGTGTT CAAAGCAGGC CTATGCTCGA ATACATCAGC ATGCCCTCA CTGGCTG-G TAGGGAACCA GGACTITTAC TGTGAAAAAA TTAGAGTGTT CAAAGCAGGC CTATGCTCGA ATACATCAGC ATGCCCTCA CTGGCTGTAG CGGGGAACCA GGACTITTAC TGTGAAAAAA TTAGAGTGTT CAAAGCAGGC CTATGCTCGA ATACATCAGC ATGCCCTCCA CTGGCTGTAG CGGGGAACCA GGACTITTAC TGTGAAAAAA TTAGAGTGTT CAAAGCAGGC CTATGCTCGA ATACATCAGC ATGCCCTCCA CTGGCTGTAG CGGGGAACCA GGACTITTAC TGTGAAAAAA TTAGAGTGTT CAAAGCAGGC CTATGCTCGA ATACATCAGC SUT051 SUT056 ST2301 626 626 626 630 631 730 740 750 760 770 780 780 810 ATGGATAAT AGATAGGAC GIGTGGTTCT ATTHIGTIGG THICLAGGAC GOCGTAATG ATTAATAGGA ACAGTCOGGG GCATAGTAT ATGGATAAT AGATAGGAC GIGTGGTTCT ATTHIGTIGG THICLAGGAC GOCGTAATG ATTAATAGGA ACAGTCOGGG GCATAGTAT ATGGAATAAT AGAATAGGAC GIGTGGTTCT ATTHIGTIGG THICLAGGAC GOCGTAATG ATTAATAGGA ACAGTCOGGG GCATAGTAT ATGGAATAAT AGAATAGGAC GIGTGGTTCT ATTHIGTIGG THICLAGGAC COCCGTAATG ATTAATAGGA ACAGTCOGGG GCATAGTAT ATGGAATAAT AGAATAGGAC GIGTGGTTCT ATTHIGTIGG THICLAGGAC COCCGTAATG ATTAATAGGA ACAGTCOGGG GCATACAGTAT 
 SUT051
 716

 SUT056
 716

 ST2301
 714

 AY083804
 720

 AY083805
 721
 820 TCANTIGECA GAGGEGAAAT TCTTCGATTT ATTGAAGACT AACTACTCGC AAAGCATTE CCAAGGATGT TTTCATTAAT CAGGAACGAA TCANTIGECA GAGGEGAAAT TCTTCGATTT ATTGAAGACT AACTACTCGC AAAGCATTE CCAAGGATGT TTTCATTAAT CAGGAACGAA TCANTIGECA GAGGEGAAAT TCTTCGATTT ATTGAAGACT AACTACTCGC AAAGCATTEG CCAAGGATGT TTTCATTAAT CAGGAACGAA TCANTIGECA GAGGEGAAAT TCTTCGATTT ATTGAAGACT AACTACTCGC AAAGCATTEG CCAAGGATGT TTTCATTAAT CAGGAACGAA TCANTIGECA GAGGEGAAAT TCTTCGATTT ATTGAAGACT AACTACTCGC AAAGCATTEG CCAAGGATGT TTTCATTAAT CAGGAACGAA TCANTIGECA GAGGEGAAAT TCTTCGATTT ATTGAAGACT AACTACTCGC AAAGCATTEG CCAAGGATGT TTTCATTAAT CAGGAACGAA TCANTIGECA GAGGEGAAAT TCTTCGATTT ATTGAAGACT AACTACTCGC AAAGCATTEG CCAAGGATGT TTTCATTAAT CAGGAACGAA SUT051806SUT056806ST2301804AY083804810AY083805811 TCG TCG TCG TCG TCG 896 896 894 900 901 985 985 ST2301 AY083804 AY083805 1000 TROGCACCT TACCACAAAT CAAAGTC 1012 TROGCACCT TACCACAAAT CAAAGTC 1012 TROGCACCT TACCACAAAT CAAAGTC 1010 TROGCACCT TACGACAAAT CAAAGTC 1010 TROGCACCT TACGACAAAT CAAAGTT 1017 
 SUT051
 986

 SUT056
 986

 ST2301
 984

 AY083804
 990

 AY083805
 991

Figure 2C. Multiple sequence alignment of partial 18S rDNA using NS1 and NS4 primers of Astrocystis mirabilis (SUT051, SUT056) and Rosellinia sp. (ST3201) examined compared to DNA sequences from GenBank database, R. necatrix (AY083805) and A. cocoes (AY083804), by using ClustalX and BioEdit program.

AB017660 AB017657 AB017658 AB017659 AB017661 ST2301 SUT051 SUT056	10 1 ÀÀÀGÀGTTCT 1 ÀÀÀGÀGTTCT 1 ÀÀÀGÀGTTCT 1 ÀAÀGÀGTTTA 1 ÀCÀGÀGCTTA 1 ÀÀÀGÀGTTTA 1 ÀTÀGÀGTTTC 1 ÀTÀGÀGTTTC	20 <b>AT-AACTCCC</b> <b>AT-AACTCCC</b> <b>AT-AACTCCC</b> <b>CA-AGCTCCC</b> <b>CTTAACTCCC</b> <b>CA-ACTCCC</b> <b>CA-ACTCCC</b> <b>CA-ACTCC</b> -	30 ААААСССАТС ААААСССАТС АААСССАТС АААСССАТС АААСССАТС АААСССАТС –АААСССАТС –АААСССАТС	40 ТСААСАТАСС ТСААСАТАСС ТСААСАТАСС ТСААСАТАСС ТСААСАТАСС ТСААСАТАСС ТСААСАТАСС ТСААСАТАСС	50 ACCCCTTCCC ACACCTTCCC ACACCTTCCC TTACCTTCCC TACCTTCCC ACACCTTCCC ACACCTTCCC ACACCTTCCC	60 TCGCLAGETC TCGCLAGETC TCGCCGCGGG TCGCCGCGGGTC TCGCCAGGCC TCGCCAGGCC TCGCCAGGCC	70 GCGTCCTA— GCGTCCTA— GCGTCCTA— GCGTCGTA— GCGTCGTA— GCGTCCTAA— GCGTGCCAAC GCGTGCCAAC	80 CCCCGAAGTG CCCCGAAGTG CCCCGAAGTG ACGG CCCCGGAGGG CCCCGGAGGG CCCCGGAGGG CCCCGGAGGG CCCCGGAGGG CCCCGGAGGG CCCCCGAAGTG CCCCGAAGTG	77 77 79 78 75 76 76
AB017660 AB017657 AB017658 AB017658 AB017661 ST2301 SUT051 SUT056	90 78 CCCTACCCTG 78 CCCTACCCTG 78 CCCTACCCTG 70 CCCGAGA 79 CCCTACCCTG 77 CCCCCCCTG 77 GCCGCCCCCC 77 GCCGCCCCCC	100 TTÁGGÓCTA TTÁGGÓCTA TTÁGGÓCTA TÁGGÓCTA TÁGGÓCTA GCAAGGÓCTG GCAAGGÓCTG	110 CCCGGTGGGC CCCGGTGGGC CCCGGTGGGC CCTGGTGCGC CCCGGTAGGC CCCGGCCGTAC CCGGCCGTAC	CCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCC	130 -CTECCEGC -CTECCEGC -CTECCEGC -CTECCEGC -CTECCEGC -CTECCEGC TCTCECTEGT TCTCECTEGT	140           660         00           660	ACGAAAC ACGAAAC ACGAAAC ACGAAAC CTGAAAC CTGAAAC CTGGAAC CTGGACGGA CCTGGACGGA	160 TCTGT- TCTGT- TCTGT- TCTGT- TCTGT- TCTGT- GCCGTCCCCC GCCGTCCCCCC	142 142 142 143 140 156 156
AB017660 AB017657 AB017658 AB017659 AB017661 ST2301 SUT051 SUT056	170 143 TTAGC—ATT 143 TTAGC—ATT 143 TTAGC—ATT 143 TTAGC—ATT 144 TTTATCATT 141 TTAATT—ACT 157 TTAATTCTTG 157 TTAATTCTTG	180 GA-AT-TCTG GA-AT-TCTG GA-AT-TCTG GA-AT-TCTG GA-AT-TCTG GG-ATATCTG AATACTGTTG AATACTGTTG	190 ÅÅČÅC ÅÅČÅC ÅÅČÅC ÅÅČÅC	— АТААСТАА — АТААСТАА — АТААСТАА — АТААСТАА — АТААСТАА — АТААСТАА — АТААСТАА ТСАТААСТАА ТСАТААСТАА	АТААСТТААА АТААСТТААА АТААСТТААА АТААСТТААА АТААСТТААА АТААСТТААА АТААСТТААА АТАСТТААА АТТАССТААА АТТАССТААА	220 <u>ÀCTITCÀACA</u> <u>ACTITCÀACA</u> <u>ACTITCÀACA</u> <u>ACTITCÀACA</u> <u>ACTITCÀACA</u> <u>ACTITCÀACA</u> <u>ACTITCÀACA</u>	23 ACGATCTCT ACGGATCTCT ACGGATCTCT ACGGATCTCT ACGGATCTCT ACGGATCTCT ACGGATCTCT ACGGATCTCT	TGGTTCTGGC TGGTTCTGGC TGGTTCTGGC TGGTTCTGGC TGGTTCTGGC TGGTTCTGGC TGGTTCTGGC TGGTTCTGGC	211 211 211 191 219 210 236 236
AB017660 AB017657 AB017658 AB017659 AB017661 ST2301 SUT051 SUT055 SUT056	250 212 ATCGATGAAG 212 ATCGATGAAG 212 ATCGATGAAG 214 ATCGATGAAG 216 ATCGATGAAG 211 ATCGATGAAG 237 ATCGATGAAG 237 ATCGATGAAG	260 ÀÀCGCÀGCGA ÀACGCÀGCGA ÀACGCÀGCGA ÀACGCÀGCGA ÀACGCÀGCGA ÀACGCÀGCGA ÀACGCÀGCGA ÀACGCÀGCGA	270 AATGCGATAA AATGCGATAA AATGCGATAA AATGCGATAA AATGCGATAA AATGCGATAA AATGCAATAA AATGCAATAA	CTAATGTGAA GTAATGTGAA GTAATGTGAA GTAATGTGAA GTAATGTGAA GTAATGTGAA GTAATGTGAA GTAATGTGAA	TIGCAGAATT TIGCAGAATT TIGCAGAATT TIGCAGAATT TIGCAGAATT TIGCAGAATT TIGCAGAATT TIGCAGAATT	CAGTGAÁTCA CAGTGAÁTCA CAGTGAÁTCA CAGTGAÁTCA CAGTGAÁTCA CAGTGAÁTCA CAGTGAÁTCA CAGTGAÁTCA	TCGAATCTTT TCGAATCTTT TCGAATCTTT TCGAATCTTT TCGAATCTTT TCGAATCTTT TCGAATCTTT TCGAATCTTT TCGAATCTTT	З20 GÀÁCGCÀCÀT GAACGCÀCAT GAACGCÀCAT GAACGCÀCAT GAACGCÀCAT GAACGCÀCAT GAACGCÀCAA GAACGCÀCAA	291 291 291 271 295 290 316 316
AB017660 AB017657 AB017658 AB017659 AB017661 ST2301 SUT051 SUT056	330 292 TGCGCCATT 292 TGCGCCCATT 292 TGCGCCCATT 296 TGCGCCCATT 291 TGCGCCCATT 317 TGCGCCCATT 317 TGCGCCCAAT	З40 АСТАТТСТАС АСТАТТСТАС АСТАТТСТАС АСТАТТСТАС АСТАТТСТАС АСТАТТСТАС АСТАТСТАС АСТАТСТАС АСТАТСТАС АСТАТСТАС	350 TGGGCATGCC TGGGCATGCC CGGCATGCC TGGGCATGCC TGGGCATGCC TGGGCATGCC TGGGCATGCC	TGTTCAACCG TGTTCAACCG TGTTCGACCG TGTTCGACCG TGTTCGACCG TGTTCGACCG TGGTCGACCG TGGTCGACCG TGGTCGACCG	TCATTICAAC TCATTICAAC TCATTICAAC TCATTICAAC TCATTICAAC TCATTICAAC TCATTICAAC TCATTICAAC TCATTICAAC	CCTTAAGCCC CCTTAAGCCC CCTTAAGCCC CCTTAAGCCC CCTTAAGCCC CCTTAAGCCC CCTTAAGCCC CCTTAAGCCC	CTGTIGCITA CTGTIGCITA CTGTIGCITA CAGCTGCITA CAGCTGCITA CTGTIGCITA -TGCTGGITA -TGCTGGITA	digitication GTGTTGGGGG GTGTTGGGGG GTGTTGGGGG GTGTTGGGGG GCGTTGGGGGG A-GGTGGGGAG A-GGTGGGAG	371 371 351 379 370 393 393
AB017660 AB017657 AB017658 AB017659 AB017661 ST2301 SUT051 SUT056	410 372 CCTGCAGCGC 372 CCTGCAGCGC 372 CCTGCAGCGC 3752 CCCCGCGCC 376 CCTACGCGCA 371 CCTACGCGA 394 CCTACGG-GA	-CTGCT-GCA -CTGCT-GCA -CTGCT-GCA GCTGCCGCG -C-GTA -GCCGCTA -C-GTA	430 GCCCCTCGAA GCCCCTCGAA GCCCCTCGAA GCCCCTCGAA GCTCCCAAA GCTTCCCAAA GCTTCCCAAA	diciáciteces GTCAGTEGCE GTCAGTEGCE GTTAGTEGCE GTTAGTEGCE ATTAGTEGCE GTTAGTEGCE GTTAGTEGCE GTTAGTEGCE	GAGTCGGTCA GAGTCGGTCA GAGTCGGTCA GAGTCGGTCG GAGTCGGTT- GAGTCGGT GAGTCGG-A- GAGTCGG-A-	CACACTCTAG CACACTCTAG CACACTCTAG CACACTCTAG CCCACCCCAG CCCACCCCAG CACACTCTAG CGCACT-TAA CGCACT-TAA	ACGTAGTAGA ACGTAGTAGA ACGTAGTAGA GCGTAGTAGA ACGTAGTAGA ACGTAGTAGA ACGTAGTAAA ACGAAG-AAT ACGAAG-AAT	480 THICTCATCT TTTCTCATCT TTTCTCATCT TTTCTCATCT TTTCTCATCT TTTTCTCATCT TTTTCTCATCT TTTTCTCATCT TTTTCTCATCT TAT-CAAT— TAT-CAAT—	449 449 431 448 444 459
AB017660 AB017657 AB017658 AB017659 AB017661 ST2301 SUT051 SUT056	490 450 CCCTATCGT 450 CCCTATCGT 450 CCCTATCGT 432 CCCTCTATG 449 CCCCTATAG 449 CCCCTATTAG 460 CCCTGCAAG	T-GTGCCGGT T-GTGCCGGT T-GTGCCGGT G-GCGCCGGT GTGTGCCGGT TTGGACCGGT TCGGACCGGT TCGGACCGGT	510 ccccreccer ccccreccer ccccreccer ccccreccea ccccreccea ccccreccea ccccreccea ccccreccea cccrreccea cccrreccea	520 34444C4CCCC 4444C4CCCCC 4444C4CCCCCC 4444CCCCCCC 4444CCCCCCC 4444CCCCCCC 64444CCCCCC G4444CCCCCC	-CCTATÀCCA           -CCTATÀCCA           -CCTATÀCCA           ATCTATACCA           ATTTATCAAA           AATTTATCAAG           AAATTAAAGG	ÀÀ 499 ÀÀ 499 ÀÀ 499 ÀÀ 499 ÀA 499 ÀA 499 ÀA 495 G- 495 506 			

Figure 3C. The ITS1-5.8S-ITS2 sequence alignment of Astrocystis mirabilis (SUT056, SUT051) and Rosellinia sp. (ST2301) examined compared to DNA sequences from GenBank database, R. arcuata (AB017660), R. pepo (AB017659), R. quercina (AB017661), and R. necatrix (AB017657 and AB017658), by using ClustalX and BioEdit programs for phylogenetic tree construction in Figure 83. Arrows indicate the start and the stop of 5.8S rDNA sequences.

ST2301 SUT051 SUT056

297

AJ390421 AJ390422 ST2321 AJ390423 SUT161 SUT260 KS15	1 1 1 1 1	10 GCGAGTTAAT GCGAGTTAAT GCGAGTTAAT ACGAGTTAAT ACGAGTTAAT AAGAGTTA	20 Тасааастос тасааастос тасааастос тасааастос тасааастос тасааастос тасааастос тасааастос тасааастос тасааастос	30 АААСССАТСТ АААСССАТСТ АААСССАССТ АААСССАССТ АААСССАССТ АААСССАССТ АААСССАССТ АААСССАССТ	40 GAACTTACCT GAACTTACCT GAACGTACCT GAACGTACCT GAACGTACCT GAACGTACCT GAACGTACCT	50 GCTGTTGCCT ACTGTTGCCT ATTGTTGCCT ACTGTTGCCT ACTGTCGCCT AACGTTGCCT	60 CGCCAGGTTG CGCCAGGTTG CGCCAGGTTG CGCCAGGTCG CGCCAGGTCG CGCCAGGTCG CGCCGGCCGGTCG	70 CCCTCCCGAG CCCTCCCGAG TCCTCTAGAG TCCTCCCCCG TCCTCTAGAG -CCTGAGAGG TAC	TG   CT     TG   CC     CT   CC     TGAAGTTCTC   CC     TG   CC     CC   CC	74 74 80 74 70 59
AJ390421 AJ390422 ST2321 AJ390423 SUT161 SUT260 KS15	75 75 75 81 75 71 60	90 TÀCCCTGGÀG TÀCCCTGGÀG TÀCCCTGGÀG TÀCCCTGGÀG TÀCCCTGGÀG TÀCCCTGGÀG TÀCCCTGGÀG	100 TGGCCTACCC TGGCCTACCC TA-CTACCC GTAGTTACCC TA-CTTACCC TA-CTTACCC TGCCTTACCT	TGGÅGTÅGCT TGGÅGTÅGCT TGGÅGTÅGGT TGGÅGTÅGGT TGGÅGTÅGCT TGGÅGTÅGCT GTÅÅGTGCCT	ACCCTGTAGT ACCCTGTAGT ACCCTGTAGT ACCCTGGAGA ACCCTGGAGA ACCCTG ACCCTG ACCCTG ACCCTG	GCCTACCCTG GCCTACCCTG GCCTACCCTG GCCTACCCTA AGGGAGGTGC	GAGTAGGCÁC GAGTAGGCÁC GAGTAGGCAC TTAC	) 150 COCCCAGCGC CCCCCAGCGC CCCCCAGCGC CCCCCAGCAC CCCCCAGCAC CAGCGC CAGCGC CAGCGC TAG-GC	160 GCAACCAGAC GCAACCAGAC GCAACCAGAC CCAACAAGAC GCAACCAGAC GCAACCAGAC GCAACCAGAC ACGGGTAAGC	154 154 139 154 125 121 110
AJ390421 AJ390422 ST2321 AJ390423 SUT161 SUT260 KS15	155 155 140 155 126 122 111	171 CTGCCAGAGG CTGCCAGAGG CTGCCGAAGG CTGCCGAAGG CTGCCGAAGG CTGCCGAAGG CCGCCGGCGC	180 АССТСТСВААС АСАТСТВААС АСАТСТВААС АСАТТВААС АСАСТТАААТ АСАСТТАААТ СССАТТАААС	190 TCTTTTTAC TCTTTTCTAC TCTTTTTAC TCTATTTAT TCTTTTTAC TCTTTTTAC TCTGTTTAAT	ACTOGAACTC ACTOGAACTC CCTOGAACTC CCCOGAACTC CCCOGAACTC CCCOGAACTC CCCOGAACTC ACTOGATATC	21 ТСАААСТАТТ ТСАААСТАТТ ТСАААСТАТТ ТСАААСТАТТ ТСАААСТАТТ ТСАААСТАТТ ТСАААСТАТТ ТСАААСТАТТ	22(1 АТАСАААСАА АТАСАААСАА АТАСАААТАА АТАСАААТАА АТАСАААТАА АТАСАААТАА АТАТАААТАА АТАТАААТАА АС-ТАААТАА	230 СТТААААСТТ СТТААААСТТ СТТААААСТТ СТТААААСТТ СТТААААСТТ СТТААААСТТ СТТААААСТТ СТТААААСТТ	TCAACAACGG TCAACAACGG TCAACAACGG TCAACAACGG TCAACAACGG TCAACAACGG TCAACAACGG TCAACAACGG	234 234 219 234 205 201 188
AJ390421 AJ390422 ST2321 AJ390423 SUT161 SUT260 KS15	235 235 220 235 206 202 189	251 ATCTCTTGGT ATCTCTTGGT ATCTCTTGGT ATCTCTTGGT ATCTCTTGGT ATCTCTTGGT ATCTCTTGGT	TCTGCCATCG TCTGCCATCG TCTGCCATCG TCTGCCATCG TCTGCCATCG TCTGCCATCG TCTGCCATCG TCTGCCATCG TCTGCCATCG	271 <b>ÀTGÀAGÀÀCG</b> <b>ATGAAGÀACG</b> <b>ATGAAGÀACG</b> <b>ATGAAGÀACG</b> <b>ATGAAGÀACG</b> <b>ATGAAGÀACG</b> <b>ATGAAGÀACG</b> <b>ATGAAGÀACG</b>	ŻĂĠĊĠĂĂĂŢĠ ĊĂĠĊĠĂĂĂŢĠ CĂĠĊĠĂĂĂŢĠ CĂĠĊĠĂĂĂŢĠ CĂĠĊĠĂĂĂŢĠ CĂĠĊĠĂĂĂŢĠ CĂĠĊĠĂĂĂŢĠ CĂĠĊĠĂĂĂŢĠ	CGATAAGTAA CGATAAGTAA CGATAAGTAA CGATAAGTAA CGATAAGTAA CGATAAGTAA CGATAAGTAA CGATAAGTAA	TGTGAATTGC TGTGAATTGC TGTGAATTGC TGTGAATTGC TGTGAATTGC TGTGAATTGC TGTGAATTGC TGTGAATTGC TGTGAATTGC	31 AGAATTCAGT AGAATTCAGT AGAATTCAGT AGAATTCAGT AGAATTCAGT AGAATTCAGT AGAATTCAGT	GAATCATCGA GAATCATCGA GAATCATCGA GAATCATCGA GAATCATCGA GAATCATCGA GAATCATCGA GAATCATCGA GAATCATCGA	314 314 299 314 285 281 268
AJ390421 AJ390422 ST2321 AJ390423 SUT161 SUT260 KS15	315 315 300 315 286 282 269	331 АТСТТТБААС АТСТТТБААС АТСТТТБААС АТСТТТБААС АТСТТТБААС АТСТТТБААС АТСТТТБААС	GCÁCATTIGCE GCÁCATTIGCE GCACATTIGCE GCACATTIGCE GCACATTIGCE GCACATTIGCE GCACATTIGCE GCACATTIGCE	CCTAACAGTA CCTAACAGTA CCTAACAGTA CCTAATAGTA CCTAATAGTA CCTAATAGTA CCTAATAGTA CCTAATAGTA CCCATTAGTA	TICTGTTAGG TICTGTTAGG TICTGTTAGG TICTGTTAGG TICTGTTAGG TICTGTTAGG TICTGTTAGG TICTGTGTGGGG	CATECCTETT CATECCTETT CATECCTETT CATECCTETT CATECCTETT CATECCTETT CATECCTETT	CGAGCGTCAT CGAGCGTCAT CGAGCGTCAT CGAGCGTCAT CGAGCGTCAT CGAGCGTCAT CGAGCGTCAT	390 TTCAACCCCC TTCAACCCCC TTCAACCCCC TTCAACCCCC TTCAACCCCC TTCAACCCCC TTCAACCCCC	) 400 ÀÀĠĊĊĊTÀTT AAGCCCTÀTT AAGCCCTÀTT AAGCCCTÀTT AAGCCCTÀTT AAGCCCTÀTT AAGCCCTÀTT AAGCCCTÀTT	394 394 379 394 365 361 348
AJ390421 AJ390422 ST2321 AJ390423 SUT161 SUT260 KS15	395 395 380 395 366 362 349	41 TECTTGÀCGT TECTTGÀCGT TECTTGÀCGT TECTTGÀCGT TECTTGÀCGT TECTTGÀCGT TECTTGÀCGT	1 420 TGGGAGTITA TGGGAGTITA TGGGAGTITA TGGGAGTITA TGGGAGTITA TGGGAGTITA TGGGGGCCTA	430 CCGAAACG-T CCGAAACG-T CCGAAACG-T CCGAAACG-T CCGAAACG-T CCGAAACG-T CCGAAACG-T CCGTAGCCGT	О 441 ААТТССТСАА ААТТССТСАА ААТТССТСАА ААТТССТСАА ААТТССТСАА ААТТССТСАА ААТТССТСАА ААТТССТСАА АССССТТАА	ATATAGTEGEC ATATAGTEGEC ATATAGTEGEC ATATAGTEGEC ATATAGTEGEC ATATAGTEGEC ATATAGTEGEC AATTAGTEGEC	GGAGCTAGGT GGAGCTAGGT GGAGCTAGGT GGAGCTAGGT GGAGTTAGGT GGAGTTAGGT GGAGTCGGTT	ÓGTECTCTAA CETECTCTAA CETECTCTAA CETECTCTAA CETECTCTAA CETECTCTAA CETECTCTAA CACACTCTAG	GCGTAGTAAT GCGTAGTAAT GCGTAGTAAC GCGTAGTAAC GCGTAGTAAC GCGTAGTAAC GCGTAGTAAC ACGTAGTAAA	473 473 458 473 444 440 428
AJ390421 AJ390422 ST2321 AJ390423 SUT161 SUT160 KS15	474 474 459 474 445 441 429	491 ТАТАТІСТОС ТАТАТІСТОС ТАТАТТСТОС ТТААТІСТОС ТАТАТІСТОС ТАТАТІСТОС ТАТАТІСТОС ТАТТАТІСТОС	CTTCTGCAGC CTTCTGCAGC CTTCTGCAGC CTTCTGTAGC CTTCTGCAGC CTTCTGCAGC CTTCTGCAGC CCTATTAGTT	CGGTCTAGGT CGGTCTAGGT CGGTTTAGGT CGGCTAAGGT CGGCTTAGGT CGGCTTAGGT GGACCGGTCC	CTIGCOGTAA CTIGCOGTAA CTIGCOGTAA CCTGCOGTAA CCTGCOGTAA CCTGCOGTAA CCTGCOGTAA	) 53 абсостатат абсостатат абсостатат абсостатат абсостатат абсостатат абсостатат абсостатат абсостатат абсостатат абсостатат абсостатат	1111CI         529           1111CI         529           1111CI         529           1111CI         514           1111CI         529           1111CI         529           1111CI         529           1111CI         500           1111CI         496           1111-C-         481			

Figure 4C. The ITS1-5.8S-ITS2 sequence alignment of *Camillea tinctor* (AJ390421, AJ390422), *C. tinctor* (ST2321), *C. obularia* (AJ390423), *C. tinctor* (SUT161), *C. tinctor* (SUT260), and *C. selangorensis* (KS15) by using ClustalX and BioEdit programs for phylogenetic tree construction in Figure 86. Arrows indicate the start and the stop of 5.8S rDNA sequences.

		10	20	30	40	50	60	70	80	90	10	0 110	120	13	140	150	
		····I····I				····I····I					·····				1	· · · · · l · · · · · l	
SUT209	1	AAGAGTTATC	TAAACTCC-A	ACCCTATGTG	AAC-TTACCG	CCGT-GCCTC	GGCGGGGCCGC	GTTCGCCCTG	TAGTTTACTA	CCTGGC	GGCGC	GCTACAGGCC	CGCCGGTGGA	CTGCTAAAC-	TCTGTTATAT	ATACG-TATC	136
SUT178	1	AAGAGTTATC	TAAACTCC-A	ACCCTATGTG	AAC-TTACCG	CCGTTGCCTC	GGCGGGGCCGC	GTTCGCCCTG	TAGTTTACTA	CCTGGC	GGCGC	GCTACAGGCC	CGCCGGTGGA	CTGCTAAAC-	TCTGTTATAT	ATACG-TATC	137
SUT278	1	AAGAGTTAGG	-AAACTCC-A	ACCCTATGTG	AAC-TTACCG	CCGTTGCCTC	GGCGGGCCGC	GTTCGCCCTG	TAGTTTACTA	CCTGGC	GGCGC	GCTACAGGCC	CGCCGGTGGA	CTGCTAAAC-	TCTOTTATAT	ATACG-TATC	136
SUT039	1	GAGTTATC	TAAACTCC-A	ACCCTGTGTG	AAC-TTACCG	CCGTTGCCTC	GGCGGGCCGC	GTTCGCCCTG	TAGTTTACTA	CCTGGC	GGCGC	GCTACAGGCC	CGCCGGTGGA	CTGCTAAAC-	TCTGTTATAT	ATACG-TATC	135
AY616684	1	TTGAGTTATC	TAAACTCC-A	ACCCTATGTG	AAC-TTACCG	CCGTTGCCTC	GGCGGGGCCGC	GTTCGCCCTG	TAGTTTACTA	CCTGGC	GGCGC	GCTACAGGCC	CGCCGGTGGA	CTGCTAAAC-	TCTGTTATAT	ATACG-TATC	137
SUT168	1	CTGAGTTATC	TAAACTCC-A	ACCCTATGTG	AAC-TTACCG	CCGTTGCCTC	GGCGGGGCCGC	GTTCGCCCTG	TAGTTTACTA	CCTGGC	GGCGC	GCTACAGGCC	CGCCGGTGGA	CTGCTAAAC-	TCTGTTATAT	ATACG-TATC	137
SUT322	1	CTGAGTTATC	TAAACTCC-A	ACCCTATGTG	AAC-TGACCG	CCGTTGCCTC	GGCGGGGCCGA	GTTCGCCCTG	AAGACGACTA	CCTGGC	GGCGC	GCTACAGGCC	CGCCGGTGGA	CTGCTAAAC-	TCTGGGATAT	ATACG-TATC	137
SUT085	1	CTGAGTTATC	TAAACTCC-A	ACCCTATGTG	AAC-TTACCG	CCGTTGCCTC	GGCGGGGCCGC	GTTCGCCCTG	TAGTTTACTA	CCTGGC	GGCGC	GCTACAGGCC	CGCCGGTGGA	CTGCTAAAC-	TCTGTTATAT	ATACG-TATC	137
AY616682	1	CTGAGTTATC	TAAACTCCCA	ACCCTATGTG	AACCTTACCG	TCGTTGCCTC	GGCGGGTTGG	G	-GAGCTACCC	TGTAGT	AGCCC	CCCGTAGGCC	CGCCGGTGGA	CTGTAAACT-	CTTGTTTTTT	GTATG-GAAT	129
AY616681	1	CCGAGTTATC	TAAACTCC-A	ACCCTTTGTG	AAACTTACCG	TCGTTGCCTC	GGCGGGGCTGC	GCTTACCCTG	TAG-CTACCC	TGTAGCTACC	CGGTAGGCGC	GCTCCAAGCC	CGCCGGTGGA	CCACTAAAC-	TCTGTTTTAA	TACCG-AATC	146
AY616682	1	CCGAGTTATC	TAAACTCC-A	ACCCTTTGTG	AAACTTACCG	TCGTTGCCTC	GGCGGGCTGC	GCTTACCCTG	TAG-CTACCC	TGTAGCTACC	CGGTAGGCGC	GCTCCAAGCC	CGCCGGTGGA	CCACTAAAC-	TCTGTTTTAA	TACCG-AATC	146
AY616683	1	CCGAGTTATC	TAAACTCC-A	ACCCTTTGTG	AAACTTACCG	TCGTTGCCTC	GGCGGGCTGC	GCTTACCCTG	TAG-CTACCC	TGTAGCTACC	CGGTAGGCGC	GCTCCAAGCC	CGCCGGTGGA	CCACTAAAC-	TCTGTTTTAA	TACCG-AATC	146
AF176955	1	CCGAGTTATC	TAAACTCC-A	ACCCTTTGTG	AAACTTACCG	TCGTTGCCTC	GGCGGGGCTGC	GCTTACCCTG	TAG-CTACCC	TGTAGCTACC	CGGTAGGCGC	GCTCCAAGCC	CGCCGGTGGA	CCACTAAAC-	TCTGTTTTAA	TACCG-AATC	146
AF176958	1	CTGAGTTATC	TAAACTCC-A	ACCCTTTGTG	AAACTTACCG	TCGTTGCCTC	GGCGGGGCTGC	GCTTACCCTG	TAG-CTACCC	TGTAGCTACC	CGGTAGGCGC	GCTCCAAGCC	CGCCGGTGGA	CCACTAAAC-	TCTGTTTTAA	TACCG-AATC	146
AF176954	1	CTGAGTTATC	TAAACTCC-A	ACCCTTTGTG	AAACTTACCG	TCGTTGCCTC	GGCGGGCTGC	GCTTACCCTG	TAG-CTACCC	TGTAGCTACC	CGGTAGGCGC	GCTCCAAGCC	CGCCGGTGGA	CCACTAAAC-	TCTGTTTTAA	TACCG-AATC	146
L1	1	CTGAGTTATC	TAAACTCC-A	ACCCTTTGTG	AAACTTACCG	TCGTTGCCTC	GGCGGGCTGC	GCTTACCCTG	TAG-CTACCC	TGTAGCTACC	CGGTAGGCGC	GCTCCAAGCC	CGCCGGTGGA	CCACTAAAC-	TCTGTTTTAA	TACCG-AATC	146
L2	1	CTGAGTTATC	TAAACTCC-A	ACCCTTTGTG	AAACTTACCG	TCGTTGCCTC	GGCGGGGCTGC	GCTTACCCTG	TAG-CTACCC	TGTAGCTACC	CGGTAGGCGC	GCTCCAAGCC	CGCCGGTGGA	CCACTAAAC-	TCTGTTTTAA	TACCG-AATC	146
AF176957	1	CTGAGTTATC	TAAACTCC-A	ACCCTTTGTG	AAACTTACCG	TCGTTGCCTC	GGCGGGCTGC	GCTTACCCTG	TAG-CTACCC	TGTAGCTACC	CGGTAGGCGC	GCTCCAAGCC	CGCCGGTGGA	CCACTAAAC-	TCTGTTTTAA	TACCG-AATC	146
AF176969	1	CTGAGTTATC	TAAACTCCCA	ACCCTTTGTG	AACCTTACCG	TCGTTGCCTC	GGCGGGCTGT	ACTTACCCTG	TAG-CTACCC	TGTAGCTACC	CGGTAGGTGC	GCTCCAAGCC	CGCCGGTGGA	CCACTAAAT-	TCTATTTTAC	TACTG-TATC	147
AF176968	1	CTGAGTTATC	TAAACTCCCA	ACCCTTTGTG	AACCTTACCG	TCGTTGCCTC	GGCGGGCTGT	ACTTACCCTG	TAG-CTACCC	TGTAGCTACC	CGGTAGGTGC	GCTCCAAGCC	CGCCGGTGGA	CCACTAAAT-	TCTATTTTAC	TACTG-TATC	147
AF176967	1	CTGAGTTATC	TAAACTCCCA	ACCCTTTGTG	AACCTTACCG	TCGTTGCCTC	GGCGGGCTGT	ACTTACCCTG	TAG-CTACCC	TGTAGCTACC	CGGTAGGTGC	GCTCCAAGCC	CGCCGGTGGA	CCACTAAAT-	TCTATTTTAC	TACTG-TATC	147
AY315403	1	CTGAGTTATC	TAAACTCCCA	ACCCTTTGTG	AACCTTACCG	TCGTTGCCTC	GGCGGGCTGT	ACTTACCCTG	TAG-CTACCC	TGTAGCTACC	CGGTAGGTGC	GCTCCAAGCC	CGCCGGTGGA	CCACTAAAT-	TCTATTTTAC	TACTG-TATC	147
AF176982	1	CTGAGTTATC	TAAACTCCCA	ACCCTTTGTG	AACCTTACCG	TCGTTGCCTC	GGCGGGCTGT	ACTTACCCTG	TAG-CTACCC	TGTAGCTACC	CGGTAGGTGC	GCTCCAAGCC	CGCCGGTGGA	CCACTAAAT-	TCTATTTTAC	TACTG-TATC	147
AF176975	1	CTGAGTTATC	TAAACTCCCA	ACCCTATGTG	AACCTTACCG	TCGTTGCCTC	GGCGGGCCGC	GCTTACCCGG	TAG-CTACCC	TGTAGCTACC	CGGTAGGTGC	GCTATAGGCC	CGCCGGTGGA	CTACTCAACT	CTGTTTTTAA	TACTG-TATC	148
AF176974	1	CTGAGTTATC	TAAACTCCCA	ACCCTATGTG	AACCTTACCG	TCGTTGCCTC	GGCGGGGCCGC	GCTTACCCGG	TAG-CTACCC	TGTAGCTACC	CGGTAGGTGC	GCTATAGGCC	CGCCGGTGGA	CTACTCAACT	CTGTTTTTAA	TACTG-TATC	148
AF176973	1	CTGAGTTATC	TAAACTCCCA	ACCCTATGTG	AACCTTACCG	TCGTTGCCTC	GGCGGGCCGC	GCTTACCCGG	TAG-CTACCC	TGTAGCTACC	CGGTAGGTGC	GCTATAGGCC	CGCCGGTGGA	CTACTCAACT	CTGTTTTTAA	TACTG-TATC	148
AF163022	1	CTGAGTTATC	TAAACTCCCA	ACCCTATOTO	AACCTTACCG	TCGTTGCCTC	GGCGGGGCTGT	GCTTACCCGG	TAG-CTACCC	TOTAGCTACC	COGTAGOTAC	GCTGCAAGCC	-GCCGGTGGA	CCACTAAAG-	GGTTTAAT	TACTG-TATC	144
AF163023	1		AACTCCCA	ACCCTATGTG	AACCTTACCG	TNGTTGC-TC	GENGGGCTGT	G-TTACCCGG	TAGTACCC	TG	-GCTAGGTAC	G-TGCAAGCC	-GCCGGTGGA	CCACTAAACC	TCTGTTTAAT	TACTGCTATC	123
AF163021	1		-AAACTCCCA	ACCCTATGTG	AACCTTACCG	TNGTTGC-TC	GENEGETET	G-TTACCCGG	TAGTACCC	TG	-GCTAGGTAC	G-TGCAAGCC	-GCCGGTGGA	CCACTAAACC	TCTGTTTAAT	TACTGCTATC	124
AF176981	1	CTGAGTTATC	TAAACTCCCA	ACCCTATGTG	AACCTTACCG	TCGTTGCCTC	GGCGGGCTGT	GCTTACCCGG	TAG-CTACCC	TGTAGCTACC	CGGTAGGTAC	GCTGCAAGCC	CGCCGGTGGA	CCACTAAAC-	TCTGTTTAAT	TACTG-TATC	147
	-																

Figure 5C. The ITS1-5.8S-ITS2 sequence alignment of *Daldinia eschscholzii* (SUT209, SUT178, SUT278, SUT039, SUT168, SUT322, SUT085, AY616684) and *D. concentrica* (L1 and L2) examined compared to DNA sequences from GenBank database, *D. concentrica* (AY616682, AY616681, AY616683, AY176955, AY176958, AY1769584), *D. grandis* (AF176982), *D. loculata* (AF176969, AF176968, AF176967), *D. petrinia* (AF176975, AF176974, AF176973), *D. vericosa* (AF163022), *D. fissa* (AF176981), *Daldinia* sp. (AF163023, AY315403), by using ClustalX and BioEdit programs for phylogenetic tree construction in Figure 88. Arrows indicate the start and the stop of 5.8S rDNA sequences.

		16	0 17	0 8	0 19	0 200	21	0 220	23	0 24	0 25	26	0 27	28	0 29	0 300	2
																	225
S0T209	137	TCTGAATGCT	TCAACTTAAT	AAGTTAAAAC	TTTCAACAAC	GGATCTCTTG	GTTCTGGCAT	CGATGAAGAA	CGCAGCGAAA	TGCGATAAGT	AATGTGAATT	GCAGAATTCA	GTGAATCATC	GAATCTTTGA	ACGCACATTG	CGCCCATTAG	286
SUT178	138	TCTGAATGCT	TCAACTTAAT	AAGTTAAAAC	TTTCAACAAC	GGATCTCTTG	GTTCTGGCAT	CGATGAAGAA	CGCAGCGAAA	TGCGATAAGT	AATGTGAATT	GCAGAATTCA	GTGAATCATC	GAATCTTTGA	ACGCACATTG	CGCCCATTAG	287
SUT278	137	TCTGAATGCT	TCAACTTAAT	AAGTTAAAAC	TTTCAACAAC	GGATCTCTTG	GTTCTGGCAT	CGATGAAGAA	CGCAGCGAAA	TGCGATAAGG	AATGTGAATT	GCAGAATTCA	GTGAATCATC	GAATCTTTGA	ACGCACATTG	CGCCCATTAG	286
SUT039	136	TCTGAATGCT	TCAACTTAAT	AAGTTAAAAC	TTTCAACAAC	GGATCTCTTG	GTTCTGGCAT	CGATGAAGAA	CGCAGCGAAA	TGCGATAAGT	AATGTGAATT	GCAGAATTCA	GTGAATCATC	GAATCTTTGA	ACGCACATTG	CGCCCATTAG	285
AY616684	138	TCTGAATGCT	TCAACTTAAT	AAGTTAAAAC	TTTCAACAAC	GGATCTCTTG	GTTCTGGCAT	CGATGAAGAA	CGCAGCGAAA	TGCGATAAGT	AATGTGAATT	GCAGAATTCA	GTGAATCATC	GAATCTTTGA	ACGCACATTG	CGCCCATTAG	287
SUT168	138	TCTGAATGCT	TCAACTTAAT	AAGTTAAAAC	TTTCAACAAC	GGATCTCTTG	GTTCTGGCAT	CGATGAAGAA	CGCAGCGAAA	TGCGATAAGT	AATGTGAATT	GCAGAATTCA	GTGAATCATC	GAATCTTTGA	ACGCACATTG	CGCCCATTAG	287
SUT322	138	TATGAATGCT	TCAACTTAAT	AAGTTAAAAC	TTTCAACAAC	GGATCTCTTG	GGCCTGGCAT	CGATGAAGAA	CGCAGCGAAA	TGCGATAAGT	AATGTGAATT	GCAGAATTCA	GTGAATCATA	CAATCTTTGA	ACGCACATTG	CGCCCATTAG	287
SUT085	138	TCTGAATGCT	TCAACTTAAT	AAGTTAAAAC	TTTCAACAAC	GGATCTCTTG	GTTCTGGCAT	CGATGAAGAA	CGCAGCGAAA	TGCGATAAGT	AATGTGAATT	GCAGAATTCA	GTGAATCATC	GAATCTTTGA	ACGCACATTG	CGCCCATTAG	287
AY616682	130	TCTGAATGCT	TCAACTTAAT	AAGTTAAAAC	TTTCAACAAC	GGATCTCTTG	GTTCTGGCAT	CGATGAAGAA	CGCAGCGAAA	TGCGATAAGT	AATGTGAATT	GCAGAATTCA	GTGAATCATC	GAATCTTTGA	ACGCACATTG	CGCCCATTAG	279
AY616681	147	TCTGAATGCT	TCAACTTAAT	AAGTTAAAAC	TTTCAACAAC	GGATCTCTTG	GTTCTGGCAT	CGATGAAGAA	CGCAGCGAAA	TGCGATAAGT	AATGTGAATT	GCAGAATTCA	GTGAATCATC	GAATCTTTGA	ACGCACATTG	CGCCCATTAG	296
AY616682	147	TCTGAATGCT	TCAACTTAAT	AAGTTAAAAC	TTTCAACAAC	GGATCTCTTG	GTTCTGGCAT	CGATGAAGAA	CGCAGCGAAA	TGCGATAAGT	AATGTGAATT	GCAGAATTCA	GTGAATCATC	GAATCTTTGA	ACGCACATTG	CGCCCATTAG	296
AY616683	147	TCTGAATGCT	TCAACTTAGT	AAGTTAAAAC	TTTCAACAAC	GGATCTCTTG	GTTCTGGCAT	CGATGAAGAA	CGCAGCGAAA	TGCGATAAGT	AATGTGAATT	GCAGAATTCA	GTGAATCATC	GAATCTTTGA	ACGCACATTG	CGCCCATTAG	296
AF176955	147	TCTGAATGCT	TCAACTTAAT	AAGTTAAAAC	TTTCAACAAC	GGATCTCTTG	GTTCTGGCAT	CGATGAAGAA	CGCAGCGAAA	TGCGATAAGT	AATGTGAATT	GCAGAATTCA	GTGAATCATC	GAATCTTTGA	ACGCACATTG	CGCCCATTAG	296
AF176958	147	TCTGAATGCT	TCAACTTAAT	AAGTTAAAAAC	TTTCAACAAC	GGATCTCTTG	GTTCTGGCAT	CGATGAAGAA	CGCAGCGAAA	TGCGATAAGT	AATGTGAATT	GCAGAATTCA	GTGAATCATC	GAATCTTTGA	ACGCACATTG	CGCCCATTAG	296
AF176954	147	TCTGAATGCT	TCAACTTAAT	AAGTTAAAAC	TTTCAACAAC	GGATCTCTTG	GTTCTGGCAT	CGATGAAGAA	CGCAGCGAAA	TGCGATAAGT	AATGTGAATT	GCAGAATTCA	GTGAATCATC	GAATCTTTGA	ACGCACATTG	CGCCCATTAG	296
LI	147	TCTGAATGCT	TCAACTTAAT	AAGTTAAAAC	TTTCAACAAC	GGATCTCTTG	GTTCTGGCAT	CGATGAAGAA	CGCAGCGAAA	TGCGATAAGT	AATGTGAATT	GCAGAATTCA	GTGAATCATC	GAATCTTTGA	ACGCACATTG	CGCCCATTAG	296
L2	147	TCTGAATGCT	TCAACTTAAT	AAGTTAAAAAC	TTTCAACAAC	GGATCTCTTG	GTTCTGGCAT	CGATGAAGAA	CGCAGCGAAA	TGCGATAAGT	AATGTGAATT	GCAGAATTCA	GTGAATCATC	GAATCTTTGA	ACGCACATTG	CGCCCATTAG	296
AF176957	147	TCTGAATGCT	TCAACTTAAT	AAGTTAAAAC	TTTCAACAAC	GGATCTCTTG	GTTCTGGCAT	CGATGAAGAA	CGCAGCGAAA	TGCGATAAGT	AATGTGAATT	GCAGAATTCA	GTGAATCATC	GAATCTTTGA	ACGCACATTG	CGCCCATTAG	296
AF176969	148	TCTGAATGCT	TCAACTTAAT	AAGTTAAAAAC	TTTCAACAAC	GGATCTCTTG	GTTCTGGCAT	CGATGAAGAA	CGCAGCGAAA	TGCGATAAGT	AATGTGAATT	GCAGAATTCA	GTGAATCATC	GAATCTTTGA	ACGCACATTG	CGCCCATTAG	297
AF176968	148	TCTGAATGCT	TCAACTTAAT	AAGTTAAAAC	TTTCAACAAC	GGATCTCTTG	GTTCTGGCAT	CGATGAAGAA	CGCAGCGAAA	TGCGATAAGT	AATGTGAATT	GCAGAATTCA	GTGAATCATC	GAATCTTTGA	ACGCACATTG	CGCCCATTAG	297
AF176967	148	TCTGAATGCT	TCAACTTAAT	AAGTTAAAAC	TTTCAACAAC	GGATCTCTTG	GTTCTGGCAT	CGATGAAGAA	CGCAGCGAAA	TGCGATAAGT	AATGTGAATT	GCAGAATTCA	GTGAATCATC	GAATCTTTGA	ACGCACATTG	CGCCCATTAG	297
AY315403	148	TCTGAATGCT	TCAACTTAAT	AAGTTAAAAC	TTTCAACAAC	GGATCTCTTG	GTTCTGGCAT	CGATGAAGAA	CGCAGCGAAA	TGCGATAAGT	AATGTGAATT	GCAGAATTCA	GTGAATCATC	GAATCTTTGA	ACGCACATTG	CGCCCATTAG	297
AF176982	148	TCTGAATGCT	TCAACTTAAT	AAGTTAAAAAC	TTTCAACAAC	GGATCTCTTG	GTTCTGGCAT	CGATGAAGAA	CGCAGCGAAA	TGCGATAAGT	AATGTGAATT	GCAGAATTCA	GTGAATCATC	GAATCTTTGA	ACGCACATTG	CGCCCATTAG	297
AF176975	149	TCTGAATGCT	TCAACTTAAT	AAGTTAAAAC	TTTCAACAAC	GGATCTCTTG	GTTCTGGCAT	CGATGAAGAA	CGCAGCGAAA	TGCGATAACT	AATGTGAATT	GCAGAATTCA	GTGAATCATC	GAATCTTTGA	ACGCACATTG	CGCCCATTAG	298
AF176974	149	TCTGAATGCT	TCAACTTAAT	AAGTTAAAAC	TTTCAACAAC	GGATCTCTTG	GTTCTGGCAT	CGATGAAGAA	CGCAGCGAAA	TGCGATAACT	AATGTGAATT	GCAGAATTCA	GTGAATCATC	GAATCTTTGA	ACGCACATTG	CGCCCATTAG	298
AF176973	149	TCTGAATGCT	TCAACTTAAT	AAGTTAAAAC	TTTCAACAAC	GGATCTCTTG	GTTCTGGCAT	CGATGAAGAA	CGCAGCGAAA	TGCGATAACT	AATGTGAATT	GCAGAATTCA	GTGAATCATC	GAATCTTTGA	ACGCACATTG	CGCCCATTAG	298
AF163022	145	TCTGAATGCT	TCAACTTAAT	AAGTTAAAAAC	TTTCAACAAC	GGATCTCTTG	GTTCTGGCAT	CGATGAAGAA	CGCAGCGAAA	TGCGATAAGT	AATGTGAATT	GCAGAATTCA	GTGAATCATC	GAATCTITGA	ACGCACATTG	CGCCCGCTAG	294
		310	320	330	340	350	360	370	380	390	400	410	420	430	440	450	
		·····		····I····I		····		····					····	· · · · I · · · · I		· · · · · [ · · · · ]	
SUT209	287	TATTCTAGTG	GGCATGCCTG	TTCGAGCGTC	ATTTCAACCC	TTAAGCCCCT	GTTGCTTAGC	GTTGGGAATC	T-AGGTCTCC	AGGGC	CTAGTTCCCC	AAAGTCATCG	GCGGAGTCGG	AGCGTACTCT	CAGCGTAGTA	ATACC-ATTC	429
SUT178	288	TATTCTAGTG	GGCATGCCTG	TTCGAGCGTC	ATTTCAACCC	TTAAGCCCCT	GTTGCTTAGC	GTTGGGAATC	T-AGGTCTCC	AGGGC	CTAGTTCCCC	AAAGTCATCG	GCGGAGTCGG	AGCGTACTCT	CAGCGTAGTA	ATACC-ATTC	430
SUT278	287	TATTCTAGTG	GGCATGCCTG	TTCGAGCGTC	ATTTCAACCC	TTAAGCCCCT	GTTGCTTAGC	GTTGGGAATC	T-AGGTCTCC	AGGGC	CTAGTTCCC-	AAAGTCATCG	GCGGAGTCGG	AGCGTACTCT	CAGCGTAGTA	ATACC-ATTC	428
SUT039	286	TATTCTAGTG	GGCATGCCTG	TTCGAGCGTC	ATTTCAACCC	TTAAGCCCCT	GTTGCTTAGC	GTTGGGAATC	T-AGGTCTCC	AGGGC	CTAGTTCCCC	AAAGTCATCG	GCGGAGTCGG	AGCGTACTCT	CAGCGTAGTA	ATACC-ATTC	428
AY616684	288	TATTCTAGTG	GGCATGCCTG	TTCGAGCGTC	ATTTCAACCC	TTAAGCCCCT	GTTGCTTAGC	GTTGGGAATC	T-AGGTCTCC	AGGC	CTAGTTCCCC	AAAGTCATCG	GCGGAGTCGG	AGCGTACTCT	CAGCGTAGTA	ATACC-ATTC	429
SUT168	288	TATTCTAGTG	GGCATGCCTG	TTCGAGCGTC	ATTTCAACCC	TTAAGCCCCT	GTTGCTTAGC	GTTGGGAATC	T-AGGTCTCC	AGGGC	CTAGTTCCCC	AAAGTCATCG	GCGGAGTCGG	AGCGTACTCT	CAGCGTAGTA	ATACC-ATTC	430
SUT322	200	AATTCTAGTG	GGCATGCCTG	TCCGAGCGTC	ATTTCAACCC	TTAAGCCCCT	GITGCTTAGC	GITGGGAATC	T-AGGTCTCC	AGGGC	CTAGTTCCCC	RAAGTCATCG	GCGGAGTCGG	AGCGTACTCT	CAGCGTAGTA	ATACC-ATTC	430
SUTURS	288	TATTCTAGTG	GACATGCCTG	TTCGAGCGTC	ATTTCAACCC	TTAAGCCCCT	GTTGCTTAAC	GTTGGGAATC	T-AG-TCTTC	AGGC	CTAGTTCC	AAGTTATTG	GCGGAGTCG-	AGCG-ACTCT	CA-CG-AATA	ATACATTC	420
A1010082	280	TATTCTAGTG	GGCATGCCTA	TTCGAGCGTC	ATTTCAACCC	TTAAGCCCTA	GICGCITAGC	GTGGGGGAGTC	TGAGCCCTTC	AGGGGGC	TCAGTTCCTC	AAAGTCATCG	GCGGAGTCGG	ATCGTGCTCT	GAGCGTAGTA	ATTTTCTC	929
A1010001	207	TATICIAGIG	GGCATGCCTA	TICGAGCGIC	ATTTCAACCC	TIRAGCCITA	GITGCTIAGC	GTTGGGAGTC	TGCGCTGTAC	TIGTIACGGC	GCAGITCCTC	AAGIGATIG	GCGGAGTTAG	GGCATACICI	ARGEGIAGIA	ATATTICTIC	440
A1010082	297	TATICIAGIG	GGCATGCCTA	TICGAGCOTC	ATTTCAACCC	TTAAGCCTTA	GITGCTTAGC	GTTGGGAGIC	TGCGCTGTAC	TIGITACGGC	GCAGITCCTC	AAAGIGATIG	CCCCACTTAG	GGCATACICI	ANGCOTAGIA	ATATTICITC	440
AT010005	207	TATICIASIS	GOCATOCCIA	TICGAGCGIC	ATTTCAACCC	TIMAGCCITA	GITGCTIAGC	GINGGGAGIC	TOCOCTOTAC	TIGITACOOC	GCAGIICCIC	AAAGIGATIG	CCCCACTTAG	GGCATACICI	ANGCOTAGIA	ATATICITC	440
AF176955	297	TATTCTAGIG	GGCATGCCTA	TICGAGCGIC	ATTTCAACCC	TTAAGCCTTA	GTTGCTTAGC	GTTGGGAGIC	TOCOCTOTAC	TIGITALGGC	GCAGITCCTC .	AAGTGATIG	CCCCA CTTAG	GGCATACICI	AAGCGTAGTA	ATATTICITC	440
32176954	297	TATTCTAGIG	GOCATOCCTA	TICONOCOTO	ATTTCAACCC	TTANGCCTTA	OTTOCTTAGC	OTTOGOADIC	TOCOCTOTAC	TTOTTACOOC	GCAGITCCTC	AAAGTGATTG	OCOGAGITAG	GOCATACICI	AAGCOTAGTA	ATATTICITC	440
1.1	297	TATTCTACTC	GGCATGCCTA	TTCGAGCGTC	ATTTCAACCC	TTARGCCTTRA	GTTGCTTAGC	GTTGGGAGTC	TOCOCTOTAC	TTOTTACGGC	GCAGTTCCTC	ABGTGBTTG	CCCCACTTAG	GCATACTCT	AAGCOTACTA	ATATTTCTTC	446
1.2	207	TATTCTAGIG	GGCATGCCTA	TTCGAGCGTC	ATTTCAACCC	TTANGCOTTA	GTTGCTTAGC	GTTGGGAGIC	TACACTATAC	TTOTTACOOC	GCAGTTCCTC	AAAGTGATTG	GCGGAGTTAG	GGCATACTOT	AAGCGTAGTA	ATATTOTTO	116
AF176957	297	TATTCTAGTG	GGCATGCCTA	TTCGAGCGTC	ATTTCAACCC	TTANGCOTTA	GTTGCTTAGC	GTTGGGAGTC	TACACTATAC	TTOTTACOOC	GCAGTTCCTC	AAAGTGATTG	GCGGAGTTAG	GGCATACTCT	AAGCGTAGTA	ATATTTCTTC	446
31176969	298	TATTCTAGTG	GGCATGCCTA	TTCGAGCGTC	ATTTCAACCC	TTANGCCT-A	GTTGCTTAGT	GTTGGGAATC	TGCCCTGTAT	TT ATAGG	GCAGTTCCTT	ABAGTGATCG	GCGGAGTTAG	GCATACTCT	AAGCGTAGTA	ATATT-CTTC	442
AF176968	298	TATTCTAGTG	GGCATGCCTA	TTCGAGCGTC	ATTTCAACCC	TTANGCCT-A	GTTGCTTAGT	GTTGGGBATC	TGCCCTGTAT	TT ATAGG	GCAGTTCCTT	ABAGTGATCG	GCGGAGTTAG	GGCATACTCT	ANGCOTAGTA	ATATT-CTTC	442
AF176967	298	TATTCTAGTG	GGCATGCCTA	TTCGAGCGTC	ATTTCAACCC	TTAAGCCT-A	GTTGCTTAGT	GTTGGGAATC	TGCCCTGTAT	TTATAGG	GCAGTTCCTT	ABAGTGATCG	GCGGAGTTAG	GGCATACTCT	AAGCGTAGTA	ATATT-CTTC	442
AY315403	298	TATTCTAGTG	GGCATGCCTA	TTCGAGCGTC	ATTTCAACCC	TTAAGCCTCA	GTTGCTTAGC	GTTGGGAATC	TGCCCTGTAT	TTATAGG	GCAGTTCCTT	AAAGTGATTG	GCGGAGTTAG	GGCATACTCT	AAGCGTAGTA	ATATT-CTTC	443
AF176982	298	TATTCTAGTO	GGCATGCCTA	TTCGAGCGTC	ATTTCAACCC	TTAAGCCT-A	GTTGCTTAGC	GTTGGGAATC	TGCCCTGTAT	TTACAGG	GCAGTTCCTT	AAAGTGATTG	GCGGAGTTAG	GGCATACTCT	AAGCGTAGTA	ATATT-CTTC	442
AF176975	299	TATTCTAGTG	GGCATGCCTA	TTCGAGCGTC	ATTTCAACCC	TTAAGCCT-A	GCTGCTTAGT	GTTGGGAACC	TGCCCCGTAG	CTACGGG	GCAGCTCCCT	AAAGTCATCG	GCGGAGTTAG	GGCATACTCT	AAGCGTAGTA	CTATT-CTTC	443
AF176974	299	TATTCTAGTG	GGCATGCCTA	TTCGAGCGTC	ATTTCAACCC	TTAAGCCT-A	GCTGCTTAGT	GTTGGGAACC	TGCCCCGTAG	CTACGGG	GCAGCTCCCT	AAAGTCATCG	GCGGAGTTAG	GGCATACTCT	AAGCGTAGTA	CTATT-CTTC	443
AF176973						THE REAL PROPERTY AND ADDRESS OF THE PARTY O											440
	299	TATTCTAGTG	GGCATGCCTA	TTCGAGCGTC	ATTTCAACCC	TTAAGCCT-A	GCTGCTTAGT	GTTGGGAACC	TGCCCCGTAG	CTACGGG	GCAGCTCCCT	AAAGTCATCG	GCGGAGTTAG	GGCATACTCT	AAGCGTAGTA	CTATT-CTTC	44.3
AF163022	299	TATTCTAGTG	GGCATGCCTA GGCATGCCTG	TTCGAGCGTC	ATTTCAACCC	TTAAGCCT-A	GCTGCTTAGT GCTGCTTAGT	GTTGGGAACC	TGCCCCGTAG	CTACGGG	GCAGCTCCCT	AAAGTCATCG	GCGGAGTTAG	GGCATACTCT GGCATACTCT	AAGCGTAGTA AAGCGTAGTA	CTATT-CTTC ATATTTCTTC	443
AF163022 AF163023	299 295 274	TATTCTAGTG TATTCTAGCG TATTCTAGCG	GGCATGCCTA GGCATGCCTG GGCATGCCTG	TTCGAGCGTC TTCGAGCGTC TCCGAGCGTC	ATTTCAACCC ATTTCAACCC ATTTCAACCC	TTAAGCCT-A TTAAGCCT-A TTAAGCCT-A	GCTGCTTAGT GCTGCTTAGT GCTGCTTAGT	GTTGGGAACC	TGCCCCGTAG TGCCCTGCAT TGCCCAGTAC	CTACGGG CTGCAGC	GCAGCTCCCT GCAGTTCCTT GCAGTTCCTT	AAAGTCATCG AAAGTAATCG AAAGTAATCG	GCGGAGTTAG GCGGAGTTAG GCGGAGTTAG	GGCATACTCT GGCATACTCT GGCATACTCT	AAGCGTAGTA AAGCGTAGTA AA-CGTAGTA	CTATT-CTTC ATATTTCTTC ATATTTCTTC	443 440 418
AF163022 AF163023 AF163021	299 295 274 275	TATTCTAGTG TATTCTAGCG TATTCTAGCG TATTCTAGCG	GGCATGCCTA GGCATGCCTG GGCATGCCTG GGCATGCCTG	TTCGAGCGTC TTCGAGCGTC TCCGAGCGTC TCCGAGCGTC	ATTTCAACCC ATTTCAACCC ATTTCAACCC ATTTCAACCC	TTAAGCCT-A TTAAGCCT-A TTAAGCCT-A TTAAGCCT-A	GCTGCTTAGT GCTGCTTAGT GCTGCTTAGT GCTGCTTAGT	GTTGGGAACC GTTGGGAATC GTTGGGAATC GTTGGGAATC	TGCCCCGTAG TGCCCTGCAT TGCCCAGTAC TGCCCAGTAC	CTACGGG CTGCAGC CTACAGC CTACAGC	GCAGCTCCCT GCAGTTCCTT GCAGTTCCTT GCAGTTCCTT	AAAGTCATCG AAAGTAATCG AAAGTAATCG AAAGTAATCG	GCGGAGTTAG GCGGAGTTAG GCGGAGTTAG GCGGAGTTAG	GGCATACTCT GGCATACTCT GGCATACTCT GGCATACTCT	AAGCGTAGTA AAGCGTAGTA AA-CGTAGTA AA-CGTAGTA	CTATT-CTTC ATATTTCTTC ATATTTCTTC ATATTTCTTC	443 440 418 419

Figure 5C. (Continued).

		460	470	480	0 490	500	0	
		···· l···· l			···· [ · · · · ]			
SUT209	430	TCGCTTTTGC	AGTAGCCCCG	GCGGCTTGCC	GTAAAACCCC	TATATCT	TTAGTGG	483
SUT178	431	TCGCTTTTGC	AGTAGCCCCG	GCGGCTTGCC	GTAAAACCCC	TATATCT	TTAGTGG	484
SUT278	429	TCGCTTTTGC	AGTAGCCCCG	GCGGCTTGCC	GTAAA-CCCC	TATATCT	TTAGGTT	481
SUT039	429	TCGCTTTTGC	AGTAGCCCCG	GCGGCTTGCC	GTAAA-CCCC	TATATCT	TTAGTGG	481
AY616684	430	TCGCTTTTGC	AGTAGCACCG	GCGGCTTGCC	GTAAAACCCC	T		470
SUT168	431	TCGCTTTTGC	AGTAGCCCCG	GCGGCTTGCC	GTAAAACCCC	TATATCT	TTAGTGG	484
SUT322	431	TCG-TTTTGC	AGTAGCCCCG	GCGGCTTGCC	GTAAAACCCC	TATATCT	TTAGTGG	483
SUT085	421	TTGCTTTTGC	AGTAACCCCG	GCGG-TTGCC	GTAAACC			456
AY616682	425	TCGCTTCTGA	GGCCGTTCCG	GTGACTGGCC	GTAAAACCCC	TATACTT	CTAGTGG	478
AY616681	447	TCGCTTCTGT	AGTTGTCCTG	GCGGCTTGCC	GTTAAACCCC	T		487
AY616682	447	TCGCTTCTGT	AGTTGTCCTG	GCGGCTTGCC	GT-AAACCCC	T		486
AY616683	447	TCGCTTCTGT	AGTTGTCCTG	GCGGCTTGCC	GTTAAACCCC	T		487
AF176955	447	TCGCTTCTGT	AGTTGTCCTG	GCGGCTTGCC	GTTAAACCCC	TATATTTT	CTAGTGG	501
AF176958	447	TCGCTTCTGT	AGTTGTCCTG	GCGGCTTGCC	GTTAAACCCC	TATATTTT	CTAGTGG	501
AF176954	447	TCGCTTCTGT	AGTTGTCCTG	GCGGCTTGCC	GTTAAACCCC	TATATTTT	CTAGTGG	501
L1	447	TCGCTTCTGT	AGTTGTCCTG	GCGGCTTGCC	GTTAAACCCC	TATATTTT	CTAGTGG	501
L2	447	TCGCTTCTGT	AGTTGTCCTG	GCGGCTTGCC	GTTAAACCCC	TATATTTT	CTAGTGG	501
AF176957	447	TCGCTTCTGT	AGTTGTCCTG	GCGGCTTGCC	GTTAAACCCC	TATATTTT	CTAGTGG	501
AF176969	443	TCGCTTCTGT	AGTTGTCCTG	GCGGCTTGCC	GTTAAACCCC	TATATTT	CTAGTGG	496
AF176968	443	TCGCTTCTGT	AGTTGTCCTG	GCGGCTTGCC	GTTAAACCCC	TATATTT	CTAGTGG	496
AF176967	443	TCGCTTCTGT	AGTTGTCCTG	GCGGCTTGCC	GTTAAACCCC	TATATTT	CTAGTGG	496
AY315403	444	TCGCTTCTGT	AGTTGTCCTG	GCGGCTTGCC	GTTAAACCCC	TATATTT	CTAGTGG	497
AF176982	443	TCGCTTCTGT	AGTTGTCCTG	GCGGCTTGCC	GTTAAACCCC	TATATTT	CTAGTGG	496
AF176975	444	TCGCTTCTGT	AGTTGTCCTG	GCGGCTTGCC	GTAAACCCCC	TATATTTTTT	CTAGTGG	500
AF176974	444	TCGCTTCTGT	AGTTGTCCTG	GCGGCTTGCC	GTAAACCCCC	TATATTTTTT	CTAGTGG	500
AF176973	444	TCGCTTCTGT	AGTTGTCCTG	GCGGCTTGCC	GTAAACCCCC	TATATTTTTT	CTAGTGG	500
AF163022	441	TCGCTTCTGT	AGTTGTCCTG	GCGGCTTGCC	GTTAAACCCC	TATATTT	CTAGTGG	494
AF163023	419	TCGCTTCTGT	AGTTGTCCTG	GCGGCTTGCC	GTTAAACCCC	TATATTT	CTAGTGG	472
AF163021	420	TCGCTTCTGT	AGTTGTCCTG	GCGGCTTGCC	GTTAAACCCC	TATATTT	CTAGTGG	473
AF176981	443	TCGCTTCTGT	AGTTGTCCTG	GCGGCTTGCC	GTTAAACCCC	TATATTT	CTAGTGG	496

Figure 5C. (Continued).

		10		20 3	0 40	50	60	70	80	90	100	11	12	0 13	140	150	0 160	
		···· I···· I		1 [		**** [ **** ]				tree [ reset				****	seed as early		11	
SUT242	1	CTGAGTTCAT	AAAA-CTC	A ACCCTATGTO	AACCTTACCT	CTGTTTCCTC	-GGCCAACTG	CAAAGGCCTG	CCCGCGGGGGT	TTACCC	TG	CTGGGAAAAG	GGCCCGACGA	CGCTTTCCAA	CCCTTCGGGG	-AAGGTTAG-	CTGAGGG	14:
SUT244	1	CTGAGTTACC	AAAA-CTC	A ACCCTATGTO	AACCTTACCT	CTGTTTCCTC	-GGCCAACTG	CAAAGGCCTG	CCCACGGGGT	TTACCC	TG	CTGGGAAAAG	GGCCCGACGA	CGCTTTCCAA	CCCTTCGGGG	-AAGGTTAG-	CTGAGGG	14:
SUT251	1	CTGAGTTACC	AAAA-CTC	A ACCCTATGTO	AACCTTACCT	CTGTTTCCTC	-GGCCAACTG	CAAAGGCCTG	CCCGCTGGGT	TTACCC	TG	CTGGGAAAAG	GGCCCGACGA	CGCTTTCCAA	CCCTTCGGGG	-AAGGTTAG-	CTGAGGG	14:
SUT081	1	CTGAGTTACC	AAAA-CTC	A ACCOTATOTO	AACCTTACCT	CCGTTTCCTC	-GGCCAACTG	CAAAGGCCTG	CCCGCGGGGGT	TTACCC	TG	CTGGGAAAAG	GGCCCGACGA	CGCTTTCCAA	CCCTTCGGGG	-AAGGTTAG-	CTGAGGG	14:
SUT285	1	CTGAGTTACC	AAAA-CTC	A ACCOTATOTO	AACCTTACCT	CTGTTTCCTC	-GGCCAACTG	CAAAGGCCTG	CCCCCCCCCCCC	TTACCC	TG	CTGGGAAAAG	GGCCCGACGA	CGCTTTCCAA	CCCTTCGGGG	-AAGGTTAG-	CTGAGGG	14:
SUT231	1	CTGAGTTCTA	AAAAACTCO	A ACCOTTOTO	AACCT-ACCT	CTGTTTCCTC	CGGCGTACCG	CCCTGGCCTA	CCTACAGGGC	CCCCCCTARG	GGGGGGTTCTG	CTAGGGAGGT	GCCTAGCTGA	TAGCACCTAA	TCCTTCGGGC	TCTGGGTAG-	-CTGTGAAGG	15
SUT058	1	CTGAGTTATC	AAAAACTC	A ACCOTTIGTO	AACCT-ACCT	ATGTTTCCTC	CGGCGTTCCG	CTTTAGCCTA	CCCACAGGGC	TCCCCTAAGG	GGGGGGTTCTG	CTGGGGAGGT	GCCTGAGTGC	TACCTA-	TCCTTCGGGG	TACGGTTAGT	GCAGTGAAGG	15
SUT243	1	CTGAGTTATC	AAAAACTCO	A ACCOTTIGTO	AACCT-ACCT	ATGTTTCCTC	CGGCGTTCCG	CTTTAGCCTA	CCCACAGGGC	TCCCCTAAGG	GGGGGGTTCTG	CTGGGGAGGT	GCCTGAGTGC	TACCTA-	TCCTTCGGGG	TACGGTTAGT	GCAGTGAAGG	15
AJ390409	1	CTGAGTTATC	AAAAACTC	A ACCOTTIGTO	AACCT-ACCT	ATGTTTCCTC	CGGCGTACCG	CTTTAGCCTA	CCCACAGGGC	TCCCCTAAGG	GGGGGGTTCTG	CTGGGGGAGGT	GCCTGAGTGC	TACCTA-	TCCTTCGGGG	TACGGTTAGT	GCAGTGAAGG	15!
SUT009	1	CTGAGTTATC	AAAAACTCO	A ACCOTTOTO	AACCT-ACCT	ATGTTTCCTC	CGGCGTACCG	CTGTAGCCTA	CCCGCAGGGC	TCCCCCTTAG	GGGGGGTTTTG	CTGGGGAGGT	GCCTGAGTGC	TACCTA-	TCCTTCGGGG	TACGGTTAGT	GCAGTGAAGG	15
SUT010	1	CTGAGTTATC	AAAAACTC	A ACCOTTIGTO	AACCT-ACCT	ATGTTTCCTC	CGGCGTACCG	CTGTAGCCTA	CCCGCAGGGC	TCCCCCTTAG	GGGGGGTTTTG	CTGGGGAGGT	GCCTGAGTGC	TACCTA-	TCCTTCGGGG	TACGGTTAGT	GCAGTGAAGG	15
AJ390397	1	CTGAGTTATC	AAAAACTCO	A ACCOTTIGTO	AACCT-ACCT	ATGTTTCCTC	CCCCCTACCC	CTGTAGCCTA	CCCGCAGGGC	TCCCCCTTAG	GGGGGGTTTTG	CTGGGGAGGT	GCCTGAGTGC	TACCTA-	TCCTTCGGGG	TACGGTTAGT	GCAGTGAAGG	15!
SUT098 H.urceolatum	1	CTGAGTTTAC	CAAAACTCO	A ACCOTTOTO	AACCT-ACTA	CTGTTTCCTC	CGGCGTAACG	CTTTAGCCTA	CCTACAGGGC	ATTCTTTTGG	GGATGTTCTG	CTAGGGAGGT	GCCCGAAGCA	C	-CCTTC	TTAA-		12:
		17	0	180 19	20	0 21	0 22	0 23	24	0 25	0 26	27	28	29	300	310	0 320	
				1											· · · · [ · · · · ]			
SUT242	142	TGCCG-CTAA	GGCCGGCCG	C AGCGCCG	TAAACTGTTC	CAAAATACTT	TGTCCAACTC	TACCCTATAG	AACCAATCGT	TCGAATATCT	TATCTCGAGG	CTTTTCTTTT	TGCCTTGAGG	CTGAGGCTTT	TCCTTGCCTT	GCCTTTCTTC	CAGTTCAAAT	291
SUT244	142	TGCCG-CTAA	GGCCGGCCG	C AGCGCCG	TAAACTGTTC	CAAAATACTT	TGTCCAACTC	TACCCTATAG	AACTAATCGT	TCGAATCTCT	TATCTCGAGG	CTTTTCTTTT	TGCCTTGAGG	CTGAGGCTTT	TCCTTGCCTT	GCCTTTCTTC	CAGTTCAAAT	291
SUT251	142	TGCCG-CTAA	GGCCGGCCG	C AGCGCCG	TAAACTGTTC	CAAAATACTT	TGTCCAACTC	TACCCTATAG	AACCAATCGT	TCGAATCTCT	TATCTCGAGG	CTTTTCTTTT	TGCCTTGAGG	CTGAGGCTTT	TCCTTGCCTT	GCCTTTCTTC	CAGTTCAAAT	291
SUT081	142	TGCCG-CTAA	GGCCGGCCG	C AGCGCCG	TAAACTGTTC	CAAAATACTT	TGTCCAACTC	TACCCTATAG	AACCAATCOT	TEGAATETET	TATCTCGAGG	CTTTTCTTTT	TGCCTTGAGG	CTGAGGCTTT	TCCTTGCCTT	GCCTTTCTTC	CAGTTCAAAT	294
SUT285	142	TGCCG-CTAA	GGCCGGCC	C AGCGCCG	TAAACTGTTC	CAAAATACTT	TOTCCAACTC	TACCCTATAG	AACTAATCGT	TEGAATCTET	TATCTCGAGG	CTTTTCTTTT	TGCCTTGAGG	CTGAGGCTTT	TCCTTGCCTT	GCCTTTCTTC	CAGTTCAAAT	291
SUT231	158	TGCCGACCAA	GGCCT-CG	T AGCGCCCGAG	AGGACCGCTC	GAAGCTCTTT	TTTCCAACTC	TTGCTAGAAC	AAACCCTTAT	-CGAAGGAAC	TATTCCAATA	AAAACCACTC	GAAAAAATA	TCGAAGAAAA	TTATCGAAGT	AACTATCGAA	GAAATTATCG	31
SUT058	156	TGCTGACCAA	GGCCT-CG	C GGCGCCGAG	AGGACCGCTC	CAAACTT			AAGCACCTAG	-TGCA	TCCAACC	CCGCGT	TGAACAACTA	TCGAAAAT	CTGCTTTTGC	TTTTTTTCTT	TACGCTA	27:
SUT243	156	TGCTGACCAA	GGCCT-CG	C GGCGCCGAG	AGGACCGCTC	CAAACTT			AAGCACCTAG	-TGCA	TCCAACC	CCGCGT	TGAACAACTA	TCGAAAAT	CTGCTTTTGC	TTTTTTTCTT	TACGCTA	27:
AJ390409	156	TGCTGACCAA	GGCCT-CG	C GGCGCCGAG	AGGACCGCTC	CAAACTT			AAGCACCTAG	-TGCA	TCCAACC	CCGCGT	TGAACAACTA	TCGAAAAT	CTGCTTTTGC	TTTTTTTCTT	TACGCTA	27;
SUT009	156	TGCCGACCAA	GGCCT-CG	C GGCGCCGAG	AGGACCGCTC	CAAACTT			AAGCACTTAG	-TGCA	TCCAACC	CCGCGT	TGAACAACTA	TCGAAAAT	TTGCTTTTGC	TTTTTTTCTT	TACGCTA	27.
SUT010	150	BOCCONCENT								-								27.
	130				AGGACCGCTC	CAAACTT			AAGCACTTAG	-TGCA	TCCAACC	CCGCGT	<b>IGAACAACIA</b>	TCGAAAAT	LIGCT I GC	Internation Client	ACGC A	- 64 A
AJ390397	156	TGCCGACCAA	GGCCT-CGG	C GGCGCCGAG	AGGACCGCTC AGGACCGCTC	CAAACTT			AAGCACTTAG	-TGCA	TCCAACC	CCGCGT	ТGААСААСТА ТGААСААСТА	TCGAAAAT	TTGCTTTTGC	TTTTTTTCTT	TACGCTA	27:

Figure 6C. The ITS1-5.8S-ITS2 sequence alignment of *Hypoxylon* sect. Annulata, Hypoxylon cf. nitens (SUT242, SUT244, SUT251, SUT081, SUT285), *H. cf. stygium* (SUT231), *H. stygium* (SUT058, SUT243, AJ390409), *H. atroroseum* (SUT009, SUT010, AJ390397), *H. urceolatum* (SUT098), by using ClustalX and BioEdit programs for phylogenetic tree construction in Figure 90. Arrows indicate the start and the end of 5.8S rDNA sequences.
		330	340	350	36	370	0 38	390	400	410	0 421	430	440	45	460	470	480	
													11		11		11	
SUT242	299	GTGTTTCCCG	GTTGGAATTA	TCTCTCGAAG	TTTACGATGT	TACGACCTTA	TGAAATGCCT	TCGCGTGAAA	TGCTACCCTG	TACTACTTCT	CGCTCGAATG	TGTTTCCCGG	TTGGAATTTT	CGCTCGAATA	TAATTCCTTT	TCTGTACTAA	TACTGTTTTT	458
SUT244	299	GTGTTTCCCG	GTTGGAATTA	TCTCTCGAAG	TTTACGATGT	TACGACCTTA	TGAAATGCCT	TCGCCTGAAA	TGCTACCCTG	TACTACTTCT	CGCTCGAATG	TETTTCCCGG	TTGGAATTTT	CGCTCGAATA	TAATTCTTTT	TCTGTACTAA	TACTGITTTT	458
SUT251	299	GTGTTTCCCG	GTTGGAATTA	TCTCTCGAAG	TTTACGATGT	TACGACCTTA	TGAAATGCCT	TCGCGTGAAA	TGCTACCCTG	TACTACTTCT	CGCTCGAATG	TGTTTCCCGG	TTGGAATTTT	CGCTCGAATA	TAATTCTTTT	TCTGTACTAA	TACTGTTTTT	458
SUTURI	299	GIGITITCCCC	GTTGGAATTA	TETETEGARG	TTTACGACOT	TACGACCTTA	TGARATGCCT	TCGCGTGAAA	TOCTACCCTG	TACTACTTCT	COCTOGAATG	TGTTTCCCCGG	THOGRAPHIT	CGCTCGAATA	TAATTCTTT	TOTOTACTAA	TACTOTTTT	430
501205	233	GIGITICCCG	GITGGAATTA	TETETEGARG	TTTACGATOT	TACGACCTTA	TGAMATGCCT	TEGEGIGAAA	TOCTACCCTG	TACTACTICI	LOCICGANIG	TOTTICCCGG	TIGGAATITT	CGCTCGARTA	TAATTCTTTT	TUTUTACTAR	TACIGITIT	100
SU1231	274	AAGATTC	111	CCCCG	TATAATOAT	TATOACTIAC	AAMAAMMMOM	TCOCTCORRES	GECCERT	THEAMOCTAC	ATTCICOTAG	IGCANTITIT	ACCCCATAGO	COTTCOTOTT	TAGOAG	ATTOTACTOR	CORCEGACO-	202
SUT243	274	ABACGTC	TTT	CCCGG	TTGGAATTAT	TGCTCGARAT	AATAATTTCT	TTACCCTGCA	GTCGTTTGTT	TTCAAGCTAC	ANTAT				TCGAAA	ATTGTTCAAA	GCTCTGAGG-	382
AJ390409	274	AAACGTC	TTT	CCCGG	TTGGAATTAT	TGCTCGAAAT	AATAATTTCT	TTACCCTGCA	GTCGTTTGTT	TTCAAGCTAC	AATAT			CTGC	TCGAAA	ATTGTTCAAA	GCTCTGAGG-	382
SUT009	274	AAACGTC	TTT	CCCCGG	TTGGAATTAT	TGCTCGAAAT	GATAATTTCC	TTACCCTGTA	GTCGTTTGTT	TTCAAGCTAC	AATATATCTG	CTCGAATAAA	ATTGCTTCAA	TATTTGC	TCGAAA	ATTGTTCAAA	GCTCTGAGG-	411
SUT010	274	AAACGTC	TTT	CCCCGG	TTGGAATTAT	TGCTCGAAAT	GATAATTTCC	TTACCCTGTA	GTCGTTTGTT	TTCAAGCTAC	AATATATCTG	CTCGAATAAA	ATTGCTTCAA	TATTTGC	TCGAAA	ATTGTTCAAA	GCTCTGAGG-	411
AJ390397	274	AAACGTC	TTT	CCCCGG	TTGGAATTAT	TGCTCGAAAT	GATAATTTCC	TTACCCTGTA	GTCGTTTGTT	TTCAAGCTAC	AATATATCTG	CTCGAATAAA	ATTGCTTCAA	TATTTGC	TCGAAA	ATTGTTCAAA	GCTCTGAGG-	411
SUT098_H.urceolatum_	214	-TGTCTC	CA	ACTCTAC	CTAGAAT	AGCAACCGAT	CGAATTTAGT	GGGTTTAAAA	TTTGTG	ATCGAATTTT	GTTTTCAAA-			-ACTTAG	TGATCG	AATTTAGTGG	GCTTAAATC-	322
		490	500	510	52	530	0 54	550	560	57	0 59	590	600	61	620	630	640	
	450				!		11			!					1			
SUT242	459	GTTTCTGCTC	GAAAGTCTTT	CCCGGTTGGA	ATTTTTC	GCTCGAGGTT	CTATTTTTT	AGAGTCTGAA	TTGCATCAAA	ACAAATTTTT	GTTAAAAACA	ACTTATATCA	AAACTITCAA	CAACGGATCT	CTTGGTTCTG	GCATCGATGA	AGAACGCAGC	013
501244	409	GITTETGETC	GAAAGTCTTT	CCCGGTTGGA	ATTTTTC	GCTCGAGGTT	CTATTTTTA	AGAGTCTGAA	TIGCATCAAA	ACAAATTTTT	GTTAAAAACA	ACTIVITATICA	AAACTITCAA	CAACGGATCT	CITGGITCIG	GCATCGATGA	AGAACGCAGC	615
SUT251	455	GTT-CTGCTC	GARAGICTTT	CCCGGTTGGA	ATTTTTC	GCTCGAGGTT	CTATTTTTT	AGAGTCTGAA	TIGCATCAAA	ACABATTITT	GTTAAAAAACA	ACTTATATCA	ARACTITCAR	CAACGGATCT	CITCOTTCTG	GCATCGATGA	AGAACGCAGC	600
SUT285	459	GTTTCTGCTC	GAAAGTCTTT	CCCGGTTGGA	ATTTTTC	GCTCGAGGTT	CTATTTTTTT	AGAGTCTGAA	TTGCATCARA	ACAAATTTTT	GTTAAAAACA	ACTTATATCA	ABACTTTCAA	CAACGGATCT	CTTGGTTCTG	GCATCGATCA	AGAACGCAGC	615
SUT231	463	CCTTTAGCTC	AAACGTGTTT	CCCGGTTGGA	ATTAAATCTC	GACAAGAACT	GTTCTAGTTT	AGGGTCTGAA	T-AACCTTAA	ACTTTTTTTG	CATAAAAACA	AATTACTTTA	AAACTTTCAA	CAACGGATCT	CITEGITCIG	GCATCGATGA	AGAACGCAGC	621
SUT058	382	GGTCT	GAATGAATTC	ATAAAATTGG	CAAAAGCCAC	-CTATAAACT	ACCOTT-CTT	ACCCCCTCAT	C-AAACCAAG	GTTTTA		AAT-ACGTTA	AAACTTTCAA	CAACGGATCT	CTTGGTTCTG	GCATCGATGA	AGAACGCAGC	526
SUT243	382	GGTCT	GAATGAATTC	ATAAAATTGG	CAAAAGCCAC	-CTATAAACT	ACGGTT-CTT	AGGGGGTGAT	C-AAACCAAG	GTTTTA	AAAACCA	AAT-ACGTTA	AAACTTTCAA	CAACGGATCT	CTTGGTTCTG	GCATCGATGA	AGAACGCAGC	526
AJ390409	382	GGTCT	GAATGAATTC	ATAAAATTGG	CAAAAGCCAC	-CTATAAACT	ACCOTT-CTT	AGGGGGTGAT	C-AAACCAAG	GTTTTA	AAAACCA	AAT-ACGTTA	AAACTTTCAA	CAACGGATCT	CTTGGTTCTG	GCATCGATGA	AGAACGCAGC	526
SUT009	411	GGTCT	GAATGAATTC	ATAAAATTGG	CAAAAGCCAC	-CTATAAACT	ACGGTT-TTT	AGGGGGTGAT	C-AAACCAAG	GTTTTA	AAAACCA	AAT-ACGTTA	AAACTTTCAA	CAACGGATCT	CTTGGTTCTG	GCATCGATGA	AGAACGCAGC	555
SUT010	411	GGTCT	GAATGAATTC	ATAAAATTGG	CAAAAGCCAC	-CTATAAACT	ACCCTT-TTT	ACCCCCTCAT	C-AAACCAAG	GTTTTA	AAA-CCA	AAT-ACGTTA	AAACTTTCAA	CAACGGATCT	CTTGGTTCTG	GCATCGATGA	AGAACGCAGC	554
AJ390397	411	GGTCT	GAATGAATTC	ATAAAATTGG	CARAAGCCAC	-CTATAAACT	ACGGTT-TTT	AGGGGGGTGAT	C-AAACCAAG	GTTTTA	AAAACCA	AAT-ACGTTA	ARACTITCAR	CAACGGATCT	CITEGITCIE	GCATCGATGA	AGAACGCAGC	555
SUT098_H.urceolatum_	322	T	GAGGGCTATT	CTAGC	GAT	CAGTAGGT	CTGATCGCAT	TAACCCTAAC	TGTTTAA	ATTAAA	CAAATTA	ACTTTTA	AAACTTTCAA	CAACGGATCT	CITEGITETE	GCATCGATGA	AGAACGCAGC	446
SUT242	616	GAAATGCGAT	AAGTAATGTG	AATTGCAGAA	TTCAGTGAAT	CATCGAATCT	TTGAACGCAC	ATTGCGCCCA	TTAGTATTCT	AGTOGGCATG	CCTATTCGAG	COTCATTACA	ACCOTTAAGC	CETGFFGETF	AGCGTTGGGA	ATCTACGGCT	TAGGCG-	771
501244 cim051	616	GARATOCOAT	ANGTANTOTO	AATTOCAGAA	TTCAGTGAAT	CARCGAATCT	TEGAACOCAC	ATTOCOCCCA	TROTATICT	AGEGGGGCATG	CCTATTCGAG	COTCATTACA	ACCOMPANDO	CONCERNICOL	ACCONTICCO	ATCTACCOCT		771
SUT081	610	GAAATGCGAT	AAGTAATGTG	AATTGCAGAA	TTCAGTGAAT	CATCGAATCT	TTGAACGCAC	ATTGCGCCCA	TTAGTATTCT	AGTGGGCATG	CCTATTCGAG	CGTCATTACA	ACCOTTAAGC	CCTGTTGCTT	AGCGTTGGGA	ATCTACGGCT	TAGGCG-	765
SUT285	616	GAAATGCGAT	AAGTAATGTG	AATTGCAGAA	TTCAGTGAAT	CATCGAATCT	TTGAACGCAC	ATTGCGCCCA	TTAGTATTCT	AGTGGGCATG	CCTATTCGAG	CGTCATTACA	ACCCTTAAGC	CCTGTTGCTT	AGCGTTGGGA	ATCTACGGCT	TAGGCG-	771
SUT231	622	GAAATGCGAT	AAGTAATGTG	AATTGCAGAA	TTCAGTGAAT	CATCGAATCT	TTGAACGCAC	ATTGCGCCCA	TTAGTATTCT	AGTGGGCATG	CCTATTCGAG	CGTCATAACA	ACCOTTAAGC	CCTGTAGCTT	AGCGTTGGGA	ACCTACCGCT	TAAGCGG	778
SUT058	527	GAAATGCGAT	AAGTAATGTG	AATTGCAGAA	TTCAGTGAAT	CATCGAATCT	TTGAACGCAC	ATTGCGCCCA	TTAGTATTCT	AGTGGGGCATG	CCTATTCGAG	CGTCAT-ACA	ACCCTTAAGC	CTTGTAGCTT	AGCGTTGGGA	ATCTACCCCT	-ACTGAGGG-	683
SUT243	527	GAAATGCGAT	AAGTAATGTG	AATTGCAGAA	TTCAGTGAAT	CATCGAATCT	TTGAACGCAC	ATTGCGCCCA	TTAGTATTCT	AGTGGGCATG	CCTATTCGAG	CGTCAT-ACA	ACCOTTAAGC	CTTGTAGCTT	AGCGTTGGGA	ATCTACCCCT	-ACTGAGGG-	683
AJ390409	527	GAAATGCGAT	AAGTAATGTG	AATTGCAGAA	TTCAGTGAAT	CATCGAATCT	TTGAACGCAC	ATTGCGCCCA	TTAGTATTCT	AGTGGGGCATG	CCTATTCGAG	CGTCATTACA	ACCCTTAAGC	CTTGTAGCTT	AGCGTTGGGA	ATCTACCCCT	CACTGAGGGG	686
SUT009	556	GAAATGCGAT	AAGTAATGTG	AATTGCAGAA	TTCAGTGAAT	CATCGAATCT	TTGAACGCAC	ATTGCGCCCA	TTAGTATTCT	AGTGGGGCATG	CCTATTCGAG	CGTCATTATA	ACCOTTAAGC	CTTGTTGCTT	AGCGTTGGGA	ATCTACCCCT	CACTAAGGGG	715
SUT010	555	GAAATGCGAT	AAGTAATGTG	AATGGCAGAA	TTCAGTGAAT	CATCGAATCT	TTGAACGCCC	ATTGCGCCCA	ATAAGAATTT	AGTGGGCATG	CCTATTCGAG	CGTCATTATA	ACCCTTAAGC	CTTGTTGCTT	AACCGTGGGA	ATCTACCCCT	CACTGAGGGG	714
AJ390397	556	GAAATGCGAT	AAGTAATGTG	AATTGCAGAA	TTCAGTGAAT	CATCGAATCT	TTGAACGCAC	ATTGCGCCCA	TTAGTATTCT	AGTGGGCATG	CCTATTCGAG	CGTCATTATA	ACCCTTAAGC	CTTGTTGCTT	AGCGTTGGGA	ATCTACCCCT	CACTGAGGGG	715
SUT098_H.urceolatum_	447	GAAATGCGAT	AAGTAATGCG	AATTGCAGAA	TTCAGTGAAT	CATCGAATCT	TTGAACGCAC	ATTGCGCCCG	CTAGCATTCT	AGCGGGGCATG	CCTATTCGAG	CGTCATTACG	ACCCTTAAGC	CTTGTCGTTT	AGCGTTGGGA	ATCTGCGGTT	TAGGCCG	603
				N 922		0) (PA		20 T222					3					
		810	820	830	0 840		0 86	0 870	880	89	0 90	910						
SUT242	772	TAGTTCCTTA	AAATTAGTGG	CGGAGTTATA	GCACTCTT	AGCGTAGTAA	TTTGCCTCGC	TTCTGAGCTG	-CTGTAGCTG	CCTGCCGT	-AAAACCCCT	ATA-CTTCTA	GT 876					
SUT244	772	TAGTTCCTCA	AAATTAGTGG	CGGAGTTATA	GCACTCTC	GGCGTAGTAA	TTTGCCTCGC	TTCTGAGCTG	-CTGTAGCTG	CCTGCCGT	-AAAACCC-T	ATA-CTTCTA	GT 875					
SUT251	772	TAGTTCCTCA	AAATTAGTGG	CGGAGTTATA	GCACTCTT	AGCGTAGTAA	TTTGCCTCGC	TTCTGAGCTG	-CTGTAGCTG	CCTGCCGT	-AAAACCCCT	ATA-CTTCTA	GT 876					
SUT081	766	TAGTTCCTCA	AAATTAGTGG	CGGAGTTATA	GCACTCTC	AGCGTAGTAA	TTTGCCTCGC	TTCTGAGCTG	-CTGTAGCTG	CCTGCCGT	-AAAACCCCT	ATA-CTTCTA	GT 870					
SUT285	772	TAGTTCCTCA	AAATTAGTGG	CGGAGTTATA	GCACTCTC	AGCGTAGTAA	TTTGCCTCGC	TTCTGAGCTG	-CTGTAGCTG	CCTGCCGT	-AAAACCC-T	ATA-CTTCTA	GT 875					
SUT231	779	TAGCTCCTTA	AATTTAGTGG	CGGAGTTACA	GCACCCCCTA	AGCGTAGTAA	ACTACCTCGC	TTTCAGGGAG	CCTGTAGCGG	CCTGCCGTTA	AAAAACCCCT	ATAA-TTCTA	GT 889					
SUT058	684	TAGTTCCTTA	AATT-AGTGG	CGGGGT-ATA	GCACACTCTA	AGCGTAGTAG	TTTAACTCGC	TTTCAGGGAG	GCTGTAGCTG	CTTGCCGT	AAAACCCCCT	ATAACTTATA	GT 791					
SUT243	684	TAGTTCCTTA	AATT-AGTGG	CGGGGT-ATA	GCACACTCTA	AGCGTAGTAG	TTTAACTCGC	TTTCAGGGAG	GCTGTAGCTG	CTTGCCGT	AAAACCCCCT	ATAACTTATA	GT 791					
AJ390409	687	TAGTTCCTTA	AATTTAGTGG	CGGGGTTATA	GCACACTCTA	AGCGTAGTAG	TITAACTCGC	TTTCAGGGAG	GCTGTAGCTG	CTTGCCGT	AAAACCCCCT	ATAACTTATA	GT 796					
501009	710	TAGTTCCTTA	AATGTAGTGG	COGGOTTATA	GCCCACTCTA	AGCGTAGTAG	TITAACTCGC	TITCAGGGAG	GETGTAGETG	CTTGCCGT	AAAACCCCTT	ATAACTTATA	00 825					
3,7390397	715	TAGTTCCTTA	AATOTAGTGG	COCCUTATA	GCACACTOTA	AGCGTAGTAG	TTTAACTOGC	TTTCAGGGAG	GCTGTAGCTG	CTTGCCGT	AAAACCCCCTT	ATAACTTATA	67 825					
SUT098 H urceolatum	604	CAGTTCCTTA	AATTCAGTCC	CGGAGTTATA	GCACACCCTA	AGCGTAGTAS	CTTACATOGC	TCCTGGGGAG	TCTATAGCCC	CCTGCCGTTA	AAAAACCCCT	ATA-TTTCTA	GT 714					
service a. arceviatura	0.04						v. inoni ouo					man travin	A. 173					

Figure 6C. (Continued).

		10	20	30	40	50	60	70	) 80	) 90	10	0 11	0 120	) 130	14	0 150	1
			· · [ · · · · ]											· · · · [ · · · · ]	****[****]		
SUT103	1	CCGAGTT-AA AC	AAAACTCC	AAA-CCCTTT	GTGAACCTTA	CCAAAGTTGC	CTCGGCGTGA	GCT-GCGG-T	TACCCTGTAG	TTACC	CTGGAGGCGT	CTACCCTGTA	GGTG	C	TTACCCTGGA	GC-TACCTTG	125
SUT105	1	CCGAGTT-AA AC	AAAACTCC	AAA-CCCTTT	GTGAACCTTA	CCAAAGTTGC	CTCGGCGTGA	GCT-GCGG-T	TACCCTGTAG	TTACC	CTGGAGGCGT	CTACCCTGTA	GGTG	C	TTACCCTGGA	GC-TACCTTG	125
ST2333	1	ACGAGTT-AA AC	AAAACTCC	AAA-CCCTTT	GTGAACCTTA	CCAAAGTTGC	CTCGGCGTGA	GCT-GCGG-T	TACCCTGTAG	TTACC	CTGAAAGCGT	CTACCCTGTA	GGTG	C	TTACCCTGGA	GC-TACCCTG	125
ST2527	1	ACGAGTT-AA AC	AAAACTCC	AAA-CCCTTT	<b>GTGAACCTTA</b>	CCAAAGTTGC	CTCGGCGTGA	GCT-GCGG-T	TACCCTGTAG	TTACC	CTGGAAGCGT	TTACCCTATA	GGTGTTTACC	<b>CTATAGTAGC</b>	TTACCCTGGA	GC-TACCCTG	140
ST2336	1	AAGAGTAT AA	CAACTCCC	AAACCCAT	<b>GTGAACATAC</b>	CTCATGTTGC	CTCGGCAGGC	CTC-GC		CTC	TCTCGTAGGC	CTTACCCTGT	AAGG	C	ATACCCGGGA	GGCG	103
AJ390395	1	CCGAGTT-AT CA	CAACTCC-	AACCCTTT	<b>GTGAACCTTA</b>	CCGTCGTTTC	CTCGGCGCAC	TGC			TGTGGGAGG-	CTACCCTGTA	GCGGTT	GT	TTACCCTACA	GGACG	101
ST2584	1	CTGAGTCC CC	CAAAACTC	CAA-CCCTTT	GTGAACCT-A	CCACAGTTTC	CTCCGCGCAA	ACGCCCTAG-	CCTAACCTAG	GCCTGGGCGC	CGCCGAGAGG	ACAATGCTCC	AACACTTATA	TCCAAC-CCT	ACTACCTAGG	ACACAACCGA	144
SUT001	1	CTGAGTAT CA	AAAACTTC	CAAACCCTTT	GTGAACCT-A	CCGCCGTTGC	CTCGGCGCGC	GCT-GCGGCT	ACCCGCC	CCGG-A	CAGAAGGGCA	GCTGCCTGTG	AGGGCCGCTG	TAAAC	CCTTCCGTCC	AGGTACCGGC	133
SUT004	1	CTGAGTAT CA	AAAACTTC	CAAACCCTTT	GTGAACCT-A	CCGCCGTTGC	CTCGGCGCGC	GCT-GCGGCT	ACCCGCC	CCGG-A	CAGAAGGGCA	GCTGCCTGTG	AGGGCCGCTG	TAAAC	CCTTCCGTCC	AGGTACCGGC	133
ST2485	1	CTGAGTT-AT CA	AAAACTCC	AAAACCCTTT	GTGAACCT-A	CCGCCGTTGC	CTCGGCGCGC	GCT-GCGGCT	ACCCGCC	CCGG-A	CAGAAGGGCA	GCTGCCTGTG	AGGGCCGCTG	TAAAC	CCTTCCGTCC	AGGTACCGGC	134
SUT005	1	CTGAGTAT CA	AAAACTTC	CAAACCCTTT	GTGAACCT-A	CCGCCGTTGC	CTCGGCGCGC	GCT-GCGGCT	ACCCGCC	CCGG-A	CAGAAGGGCA	GCTGCCTGTG	AAGGCCGCTG	TAAAC	CCTTCCGTCC	AGGTACCGGC	133
SUT262	1	CTGAGTT-AT CA	AAAACTCC	AAAACCCTTT	GTGAACCT-A	CCGCCGTTGC	CTCGGCGCGC	GCT-GCGGCT	ACCCGCC	CCGG-A	CAGAAGGGCA	GCTGCCTGTG	AGGGCCGCTG	TAAAC	CCTTCCGTCC	AGGTACCGGC	134
ST2448	1	CTGAGTT-AT CA	AAAACTCC	AAAACCCTTT	GTGAACCT-A	CCGCCGTTGC	CTCGGCGCGC	GCT-GCGGCT	ACCCGCC	CCGG-A	CAGAAGGGCG	GCTGCCTGTG	AGGGCCGCTG	TAAAC	CCTTCCGTCC	AGGTACCGGC	134
SUT1 67	1	CTGAGTT-AT CA	AAAACTCC	AAAACCCTTT	GTGAACCT-A	CCCCCCTTCC	CTCGGCGCGC	GCT-GCGGCT	ACCCGCC	CCGG-A	CAGAAGGGCA	GCTGCCTGTG	AGGGCCGCTG	TAAAC	COTTCCCTCC	AGGTACCGGC	134
JU2	1	GCGAGTTCAT CA	AAAACTCC	AAAACCCTTT	GTGAACCTTA	CCCCACTTCC	CTCGGCGTGC	GCC-GCGGCC	GTTGGGC		CACCCCAACC	GCCCCCCGAA	ACCCCCCCCC	GTGGG	GTTACC-GGC	AGGCCCC-G-	134
ST2579	ĩ	GCGAGTTCAT CA	AAAACTCC	AAAACCCTTT	GTGAACCTTA	CCGCAGTTGC	CTCGGCGTGC	GCC-GCGGCC	GTTGGGGC	CTGCTG	CAGGCCAACG	GCCCCCCGAA	ACGGGGGGGGGGGG	GTGGG	GTAACC-GGC	AGGCCCCCG-	135
ST2406	1	GCGAGTT-AC CA	AAAACTCC	AAAACCCTTT	GTGAACCTTA	CCGCAGTTGC	CTCGGCGTGC		GTTGGGC		CAGCCCACCG	GCCCCCCGAA	ACGGGGGGGGGGG	GTGGG	GTTACC-GGC	AGGCCCCCG-	134
SITE 100	1	GCGAGTTACC AC	AGAACTCC	AAAACCCTTT	GTGAACCTTA	CCGCAGTTGC	CTCGGCGAGT	GCT-GCGGCT	ATATCCC	CTGTCC	C	-CCGCCCGTC	AGGGCTGCGG	G	GC	AGGCTCT-A-	113
SUT220	1	CACACTTACC CC	TAAAACTC	CATACCCTT	GTGAACCTAC	CTATCOTTCC	CTCGGCGCGC	GCT-GCGGCT	GACGTCCCCA	AGAGCTGCTC	CCCCTCCT	AAGGCCCTCG	AATTCCCCCCC	GGGGC	mmmmcm_mcc	GGGCTTTAG-	140
cm2332	1	CAGAGIIGIC GG		CAMACCOUTT	GIGAACCIAC	CTATCOTICC	CTCGGCGCCCC	GCT GCGGCT	GACGICCGGA	AGAGCTGCTC	CCCCTCCT	ARGGCCCTGG	AATTCCGGGGG	CCCCC	mmmmcm_mcc	ACCCMMMAC-	140
312332	1	CTCACACTOIC GO	CAAAACTC	CATACCCTTT	CTCAACCIAC	CCMMACMMCC	CTCGGCGCCC	CCC	GACGICCGAA	AGAGCIGCIC		CCMACCCCCC	ACCACCCC			AGGCTTTAG=	107
Cm2426	1	CTGAGAGIAA AA	ACAAAACIC	-AA-CCCTTT	GIGAACCITA	CCTIAGTIGC	CTCGGCGTGC	CCC				GUIACCCGGG	COCCO	1499909	GITACCCIGI	AGCC	0.2
ST2430	1	CTGAGTT-TC TA	ACAACTCC	-AA-CCCTTT	GTCGAACCTA	CCACTGTTTC	CTCGGCGTAC	TGCCGCGGC-				CTCTGG	GCCGC		TGC	AG	02
5124/3	1	CTGAGTT-TC TA	ACAACTCC	CAA-CCCTTT	GTCGAACCTA	CCACTGTTTC	CTCGGCGTAC	TGCCGCGGGC-				CTCTGG	GCCGC		TGC	AG	83
JUL	1	CTGAGTT-TT TA	ACAACTCC	-AA-CCCTTT	GTCGAATCTA	CCACTGTTTC	CTCGGCGTAC	TGCCGCGGGC-				CTCTGG	GCCGC		TGC	AG	82
ST2313 H.n	1	CTGAGTT-TC TA	ACAACTCC	-AA-CCCTTT	GTCGAACCTA	CCACTGTTTC	CTCGGCGTAC	TGCCGCGGC-				CTCTGG	GCCGC		TGC	AG	82

Figure 7C. The ITS1-5.8S-ITS2 sequence alignment of *Hypoxylon* sect. *Annulata*, *Hypoxylon* cf. archeri (SUT103, SUT105, ST2333, ST2527), *Hypoxylon* sp. ST2336, *H. annulatum* (AJ390395), *H. leptascum* var. *microsporum* (ST2584), *H. purpureonitens* (SUT001, SUT004, SUT005, SUT167, SUT262, ST2485, ST2448), *H. bovei* var. *microspora* (Ju2, ST2579, ST2406, SUT025), *H. moriforme* (SUT220, ST2332), *H. cohaerens* (AJ390406), *H. nitens* (Ju1, ST2436, ST2473, ST2313) by using ClustalX and BioEdit programs for phylogenetic tree construction in Figure 91. Arrows indicate the start and the end of 5.8S rDNA sequences.

		.60 17	0 18	0 19	0 20	0 210	220	0 23	0 240	D <b>V</b> 25	26	0 27	0 280	) 29	0 300	)
SUT103	126 TAACCGGCT	A ACCCCCC	CCGAAGGA	CCACTAA	АСФСФСФФФФ	TACCCAAGTG	ТАТСТСТСАА	TGCTTCAA	CTAAATAA	GTTAAAACTT	TCAACAACGG	ATCTCTTGGT	TCTGGCATCG	ATGAAGAACG	CAGCGAAATG	264
SUT105	126 TAACCGGCT	A ACGGCCCG	CCGAAGGA	CCACTAA	ACTCTGTTTT	TACCCAAGTG	TATCTCTGAA	TGCTTCAA	CTAAATAA	GTTAAAACTT	TCAACAACGG	ATCTCTTGGT	TCTGGCATCG	ATGAAGAACG	CAGCGAAATG	264
ST2333	126 TAACCGGCT	A ACGGCCCG	CCGAAGGA	CCACTAA	ACTCTGTTTT	TACCCAAGTG	TATCTCTGAA	TGCTTCAA	CTAAATAA	GTTAAAACTT	TCAACAACGG	ATCTCTTGGT	TCTGGCATCG	ATGAAGAACG	CAGCGAAATG	264
ST2527	141 TAACCGGCT	A ACGGCCCG	CCGAAGGA	CCACTAA	ACTCTGTTTT	TACCCAAGTG	TATCTCTGAA	TGCTTCAA	CTAAATAA	GTTAAAACTT	TCAACAACGG	ATCTCTTGGT	TCTGGCATCG	ATGAAGAACG	CAGCGAAATG	279
ST2336	104 CGGGCA	CCCC-TG	CCGGCGGC	CCACGAA	ACTCTGTCTC	-ATCGTT	GAGTTCTGAA	CCCATAAC	-TAAATAA	GTTAAAACTT	TCAACAACGG	ATCTCTTGGT	TCTGGCATCG	ATGAAGAACG	CAGCGAAATG	231
AJ390395	102 CACCCTGCA	G CGGCGCC	GAAAGGA-	CTACCAA	AACTCTTTT-	-ATCCAAGTT	-ACCTCGAAC	AATTTACT	ATAC-AATA-	GTTTAAACTT	TCAACAACGG	ATCTCTTGGT	TCTGGCATCG	ATGAAGAACG	CAGCGAAATG	235
ST2584	145 TCGAGGCCT	G CGGGCTTAAA	TCTTAGGGCT	TCTCTAGCGA	CCAGTAGGGT	CTGAATGGCG	TTAAACCTAA	C-TGTTTAAA	ТТААААСААА	TCAACAACTT	TCAACAACGG	ATCTCTTGGT	TCTGGCATCG	ATGAAGAACG	CAGCGAAATG	293
SUT001	134 TAGCCGGCT	G AACAGCCC-G	CCGAAGGA	CCGCTATAAA	AACTCTCGCT	CCGGCGTGTA	TACCTTCTGA	A-TCTTCCAA	CTTATAATGA	GTTAAAACTT	TCAACAACGG	ATCTCTTGGT	TCTGGCATCG	ATGAAGAACG	CAGCGAAATG	279
SUT004	134 TAGCCGGCT	G AACAGCCC-G	CCGAAGGA	CCGCTATAAA	AACTCTCGCT	CCGGCGTGTA	TACCTTCTGA	ATCCTTCCAA	CTTATAATGA	GTTAAAACTT	TCAACAACGG	ATCTCTTGGT	TCTGGCATCG	ATGAAGAACG	CAGCGAAATG	280
ST2485	135 TAGCCGGCT	G AACAGCCC-G	CCGAAGGA	CCGCTATAAA	AACTCTCGCT	CCGGCGTGTA	TACCTTCTGA	A-TCTTCCAA	CTTATAATGA	GTTAAAACTT	TCAACAACGG	ATCTCTTGGT	TCTGGCATCG	ATGAAGAACG	CAGCGAAATG	280
SUT005	134 TAGCCGGCT	G AACAGCCC-G	CCGAAGGA	CCGCTATAAA	AACTCTCGCT	CCGGCGTGTA	TACCTTCTGA	A-TCTTCCAA	CTTATAATGA	GTTAAAACTT	TCAACAACGG	ATCTCTTGGT	TCTGGCATCG	ATGAAGAACG	CAGCGAAATG	279
SUT262	135 TAGCCGGCT	G AACAGCCC-G	CCGAAGGA	CCGCTATAAA	AACTCTCGCT	CCGGCGTGTA	TACCTTCTGA	A-TCTTCCAA	CTTATAATGA	GTTAAAACTT	TCAACAACGG	ATCTCTTGGT	TCTGGCATCG	ATGAAGAACG	CAGCGAAATG	280
ST2448	135 TAGCCGGCT	G AACAGCCC-G	CCGAAGGA	CCGCTATAAA	AACTOTOGOT	CCGGCGTGTA	TACCTTCTGA	A-TCTTCCAA	CTTATAATGA	GTTAAAACTT	TCAACAACGG	ATCTCTTGGT	TOTGGCATCG	ATGAAGAACG	CAGCGAAATG	280
501167	135 TAGCCGGC1	A AACAGCCC-G	CCGAAGGA	CCCCCAAAC	AACTCTCGCT	T-AACAACCC	TACCTTCTGA	APTOTTCCAA	CTTATAATGA	CCTTARAACTT CCTTARAACTT	TCAACAACGG	ATCTCTTGGT	TCTGGCATCG	ATGAAGAACG	CAGCGAAATG	200
502579	135 GCCGGCA	A AACGGCCC-G	CCAAAGGA-C	CCGCAAACTA	AACTCTAAAT	T-AACAACGG	TACCTTCTCA	AAACTTCAAA	CTTTTATCA	GTTAAAACTT	TCAACAACGG	ATCTCTTGGT	TCTGGCATCG	ATGAAGAACG	CAGCGAAATG	280
ST2406	134 GCCGGCA	A AACGGCCC-G	CCAAAGGA-C	ССССАВАСТА	ААСТСТАААТ	T-AACAACG-	TACCTTCTCA	AAACTTCAAA	Сттттаатса	GTTAAAACTT	TCAACAACGG	ATCTCTTGGT	TCTGGCATCG	ATGAAGAACG	CAGCGAAATG	278
SUT025	113GCCGATC	C ACCAGCCC-G	TCAAAGGA-C	CCGCTATCTA	AACTCTGAAT	T-GACTACG-	TAACTTCTGA	AAACTTCCAA	CTTGAAATAA	GTTAAAACTT	TCAACAACGG	ATCTCTTGGT	TCTGGCATCG	ATGAAGAACG	CAGCGAAATG	257
SUT220	140CCGGCTA	A AACAGCAC-G	CCAGAGGA-C	CCACAAACAA	A-CTCTAATT	T-AACGTCG-	TACCTTCTGA	AAAATATCAA	CTTTAATTA-	-TTAAAACTT	TCAACAACGG	ATCTCTTGGC	TCTGGCATCG	ATGAAGAACG	CAGCGAAATG	281
ST2332	140CCGGCTA	A AACAGCACTG	CCAGATGGAC	CCACAAACTA	G-CTCTAATT	T-AACGTCG-	TACCTTCTGA	AAAATATCAA	CTTTAATTA-	-TTAAAACTT	TCAACAACGG	ATCTCTTGGC	TCTGGCATCG	ATGAAGAACG	CAGCGAAATG	283
AJ390406	107 <b>GG</b>	C CACGGCCC-G	CCGAAGGA	CCACTG	AACTCTTGTT	T-ATCACATT	GCATCTCTGA	TTTAAACTA-	-TTAAAATAA	GTTAAAACTT	TCAACAACGG	ATCTCTTGGT	TCTGGCATCG	ATGAAGAACG	CAGCGAAATG	241
ST2436	82CGCCC	G AGGGACCGTA	CCAAACTC		TTTTATT	TCTCCCCAGT	AAAACTCATA	TAAAATTTAT	<b>ТАСААААТАА</b>	GTTAAAACTT	TCAACAACGG	ATCTCTTGGT	TCTGGCATCG	ATGAAGAACG	CAGCGAAATG	213
ST2473	83CGCCG	G AAGGACCGTA	CCAAACTC		TTTTATT	TCTCCCCAGT	AAAACTCATA	TAAAATTTAT	ТАСААААТАА	GTTAAAACTT	TCAACAACGG	ATCTCTTGGT	TCTGGCATCG	ATGAAGAACG	CAGCGAAATG	214
Ju1	82CGGCG	G AAG-ACCGTA	CCAAACTC		TTTTATT	TCTCCCCAGT	AAAACTCATA	TACAATTTAT	TATAAAATAA	GTTAAAACTT	<b>TCAACAACGG</b>	ATCTCTTGGT	TCTGGCATCG	ATGAAGAACG	CAGCGAAATG	212
ST2313 H.n	82CGCCC	G AAGGACCGTA	CCAAACTC		TTTTATT	TCTCCCCAGT	AAAATTCATA	TAAAATTTAT	ТАСААААТАА	GTTAAAACTT	TCAACAACGG	ATCTCTTGGT	TCTGGCATCG	ATGAAGAACG	CAGCGAAATG	213
	3	10 32	0 33	0 34	0 35	0 360	370	0 38	0 390	0 40	41	0 42	0 430	) 44	0 450	)
	:	10 32   ···· ····	0 33 	0 34 	0 35( 	0 360 	) 37( 	0 38 	0 39( 	0 40 	41 	0 42 	0 43( 	) 44( 	0 450 	)
SUT103	265 CGATAAGTA	10 32     A TGTGAATTGC	0 330	0 34    GAATCATCGA	0 350	0 360    GCACATTGCG	370    CCCATTAGTA	0 38    TTCTAGTGGG	0 390    CATGCCTATT	0 40    CGAGCGTCAT	41    TTCAACCCTT	0 420    AAGCCCT-GT	0 430	440    TGGGAGTCCG	0 450    CTAAT	408
SUT103 SUT105	265 CGATAAGTA 265 CGATAAGTA	10 32     A TGTGAATTGC A TGTGAATTGC	0 330	0 34    GAATCATCGA GAATCATCGA	0 350	0 360    GCACATTGCG GCACATTGCG	CCCATTAGTA	0 38    TTCTAGTGGG TTCTAGTGGG	0 390    CATGCCTATT CATGCCTATT	CGAGCGTCAT	41 TTCAACCCTT TTCAACCCTT	0 420 AAGCCCT-GT AAGCCCT-GT	AGCTTAGCGT	TGGGAGTCCG	0 450    CTAAT CTAAT	408
SUT103 SUT105 ST2333	265 CGATAAGTA 265 CGATAAGTA 265 CGATAAGTA 265 CGATAAGTA	10 32 1     A TGTGAATTGC A TGTGAATTGC A TGTGAATTGC A TGTGAATTGC	0 330    AGAATTCAGT AGAATTCAGT AGAATTCAGT	0 34    GAATCATCGA GAATCATCGA GAATCATCGA	0 350	0 360    GCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG	CCCATTAGTA CCCATTAGTA CCCATTAGTA CCCATTAGTA	0 38    TTCTAGTGGG TTCTAGTGGG TTCTAGTGGG	0 390    CATGCCTATT CATGCCTATT CATGCCTATT	CGAGCGTCAT CGAGCGTCAT CGAGCGTCAT	41 TTCAACCCTT TTCAACCCTT TTCAACCCTT	0 420    AAGCCCT-GT AAGCCCT-GT AAGCCCT-GT	AGCTTAGCGT AGCTTAGCGT AGCTTAGCGC AGCTTAGCGC	TGGGAGTCCG TGGGAGTCCG TGGGAGTCCG	0 450    CTAAT CTAAT CTAAT	408 408 408 408
SUT103 SUT105 ST2333 ST2527 ST2336	265 CGATAAGTA 265 CGATAAGTA 265 CGATAAGTA 280 CGATAAGTA 280 CGATAAGTA	10 32     A TGTGAATTGC A TGTGAATTGC A TGTGAATTGC A TGTGAATTGC A TGTGAATTGC	0 33 AGAATTCAGT AGAATTCAGT AGAATTCAGT AGAATTCAGT AGAATTCAGT	0 34 GAATCATCGA GAATCATCGA GAATCATCGA GAATCATCGA GAATCATCGA	0 350 ATCTTTGAAC ATCTTTGAAC ATCTTTGAAC ATCTTTGAAC ATCTTTGAAC	0 360 GCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG	CCCATTAGTA CCCATTAGTA CCCATTAGTA CCCATTAGTA CCCATTAGTA CCCATTAGTA	0 38 TTCTAGTGGG TTCTAGTGGG TTCTAGTGGG TTCTAGTGGG	0 390 CATGCCTATT CATGCCTATT CATGCCTATT CATGCCTATT CATGCCTATT	CGAGCGTCAT CGAGCGTCAT CGAGCGTCAT CGAGCGTCAT CGAGCGTCAT	41. TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT	0 420 AAGCCCT-GT AAGCCCT-GT AAGCCCT-GT AAGCCCT-GT AAGCCCT-GT	AGCTTAGCGT AGCTTAGCGT AGCTTAGCGC AGCTTAGCGC AGCTTAGCGC	TGGGAGTCCG TGGGAGTCCG TGGGAGTCCG TGGGAGTCCG TGGGAGTCCG	0 450     CTAAT CTAAT CTAAT CTAT	408 408 408 422 374
SUT103 SUT105 ST2333 ST2527 ST2336 A.T300395	265 CGATAAGTA 265 CGATAAGTA 265 CGATAAGTA 260 CGATAAGTA 232 CGATAAGTA 232 CGATAAGTA 236 CGATAAGTA	10         32                                 A         TGTGAATTGC           A         TGTGAATTGC           A         TGTGAATTGC           A         TGTGAATTGC           A         TGTGAATTGC           A         TGTGAATTGC	0 33 AGAATTCAGT AGAATTCAGT AGAATTCAGT AGAATTCAGT AGAATTCAGT AGAATTCAGT	0 34 GAATCATCGA GAATCATCGA GAATCATCGA GAATCATCGA GAATCATCGA GAATCATCGA	0 350 ATCTTTGAAC ATCTTTGAAC ATCTTTGAAC ATCTTTGAAC ATCTTTGAAC	0 360 GCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG	CCCATTAGTA CCCATTAGTA CCCATTAGTA CCCATTAGTA CCCATTAGTA CCCATTAGTA	0 38 <sup>8</sup> TTCTAGTGGG TTCTAGTGGG TTCTAGTGGG TTCTAGTGGG TTCTAGTGGG	0 390 CATGCCTATT CATGCCTATT CATGCCTATT CATGCCTATT CATGCCTGTT CATGCCTGTT	CGACGTCAT CGAGCGTCAT CGAGCGTCAT CGAGCGTCAT CGAGCGTCAT CGAGCGTCAT	41. TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT	0 420 AAGCCCT-GT AAGCCCT-GT AAGCCCT-GT AAGCCCT-GT AAGCCCT-GT AAGCCCTCGT	AGCTTAGCGT AGCTTAGCGT AGCTTAGCGC AGCTTAGCGC AGCTTAGCGC TGCTTAGCGC TGCTTAGCGC	TGGGAGTCCG TGGGAGTCCG TGGGAGTCCG TGGGAGTCCG TGGGAGTCCG TGGGAGCCTA	450           CTAAT           CTAAT           CTAAT           CTAAT           CTAT           CTAT           CGGT           CGGCT	408 408 408 422 374 381
SUT103 SUT105 ST2333 ST2527 ST2336 AJ390395 ST2584	265 CGATAAGTA 265 CGATAAGTA 265 CGATAAGTA 260 CGATAAGTA 280 CGATAAGTA 232 CGATAAGTA 232 CGATAAGTA 234 CGATAAGTA	10 32 1     A TGTGAATTGC A TGTGAATTGC A TGTGAATTGC A TGTGAATTGC A TGTGAATTGC A TGTGAATTGC A TGTGAATTGC	0 330 AGAATTCAGT AGAATTCAGT AGAATTCAGT AGAATTCAGT AGAATTCAGT AGAATTCAGT AGAATTCAGT	0 34 GAATCATCGA GAATCATCGA GAATCATCGA GAATCATCGA GAATCATCGA GAATCATCGA GAATCATCGA	0 350 ATCTTTGAAC ATCTTTGAAC ATCTTTGAAC ATCTTTGAAC ATCTTTGAAC ATCTTTGAAC	0 360 CACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG	0 CCCATTAGTA CCCATTAGTA CCCATTAGTA CCCATTAGTA CCCATTAGTA CCCATTAGTA	0 38 TTCTAGTGGG TTCTAGTGGG TTCTAGTGGG TTCTAGTGGG TTCTAGTGGG TTCTAGTGGG	0 390 CATGCCTATT CATGCCTATT CATGCCTATT CATGCCTATT CATGCCTATT CATGCCTATT CATGCCTATT	CGAGCGTCAT CGAGCGTCAT CGAGCGTCAT CGAGCGTCAT CGAGCGTCAT CGAGCGTCAT CGAGCGTCAT	41 TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT	0 420 AAGCCCT-GT AAGCCCT-GT AAGCCCT-GT AAGCCCT-GT AAGCCCT-GT AAGCCCT-GT	AGCTTAGCGT AGCTTAGCGT AGCTTAGCGC AGCTTAGCGC AGCTTAGCGC TGCTTAGCGC TGCTTAGCGT	TGGGAGTCCG TGGGAGTCCG TGGGAGTCCG TGGGAGTCCG TGGGAGTCCG TGGGAGTCCG TGGGAGTCTG	0 450    CTAAT CTAAT CTAAT CTACT CTACCT CGCCTC	408 408 408 422 374 381 439
SUT103 SUT105 ST2333 ST2527 ST2336 AJ390395 ST2584 SUT001	265 CGATAAGTA 265 CGATAAGTA 265 CGATAAGTA 260 CGATAAGTA 280 CGATAAGTA 232 CGATAAGTA 236 CGATAAGTA 294 CGATAAGTA 280 CGATAAGTA 280 CGATAAGTA	10 32     A TGTGAATTGC A TGTGAATTGC A TGTGAATTGC A TGTGAATTGC A TGTGAATTGC A TGTGAATTGC A TGTGAATTGC A TGTGAATTGC A TGTGAATTGC	0 330 AGAATTCAGT AGAATTCAGT AGAATTCAGT AGAATTCAGT AGAATTCAGT AGAATTCAGT AGAATTCAGT AGAATTCAGT	0 34 GAATCATCGA GAATCATCGA GAATCATCGA GAATCATCGA GAATCATCGA GAATCATCGA GAATCATCGA GAATCATCGA	0 350 ATCTTTGAAC ATCTTTGAAC ATCTTTGAAC ATCTTTGAAC ATCTTTGAAC ATCTTTGAAC ATCTTTGAAC	CACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG	CCCATTAGTA CCCATTAGTA CCCATTAGTA CCCATTAGTA CCCATTAGTA CCCATTAGTA CCCCATTAGTA CCCCCTAGTA	0 38 TTCTAGTGGG TTCTAGTGGG TTCTAGTGGG TTCTAGTGGG TTCTAGTGGG TTCTAGTGGG TTCTGGCGGG	0 390 CATGCCTATT CATGCCTATT CATGCCTATT CATGCCTATT CATGCCTATT CATGCCTATT CATGCCTATT	CGAGCGTCAT CGAGCGTCAT CGAGCGTCAT CGAGCGTCAT CGAGCGTCAT CGAGCGTCAT CGAGCGTCAT CGAGCGTCAT	41 TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TACAACCCTT TTCAACCCTT	0 420 AAGCCCT-GT AAGCCCT-GT AAGCCCT-GT AAGCCCT-GT AAGCCCT-GT AAGCCCT-GT AAGCCCTGT AAGCCCCTGT	AGCTTAGCGT AGCTTAGCGT AGCTTAGCGC AGCTTAGCGC AGCTTAGCGC TGCTTAGCGT CGCTTAGCGT TGCTTAGCGT	TGGGAGTCCG TGGGAGTCCG TGGGAGTCCG TGGGAGTCCG TGGGAGTCCG TGGGAGTCTG TGGGAGTCTG TGGGCGTCTG	0 450    CTAAT CTAAT CTAAT CTACCT CGGCTC CGCCGTGCCC	408 408 408 422 374 381 439 429
SUT103 SUT105 ST2333 ST2527 ST2336 AJ390395 ST2584 SUT001 SUT004	265 CGATAAGTP 265 CGATAAGTP 265 CGATAAGTP 260 CGATAAGTP 220 CGATAAGTP 236 CGATAAGTP 236 CGATAAGTP 280 CGATAAGTP 281 CGATAAGTP	10 32 1     A TOTGAATTGC A TOTGAATTGC A TOTGAATTGC A TOTGAATTGC A TOTGAATTGC A TOTGAATTGC A TOTGAATTGC A TOTGAATTGC	0 33:    AGAATTCAGT AGAATTCAGT AGAATTCAGT AGAATTCAGT AGAATTCAGT AGAATTCAGT AGAATTCAGT AGAATTCAGT	0 34    GAATCATCGA GAATCATCGA GAATCATCGA GAATCATCGA GAATCATCGA GAATCATCGA GAATCATCGA GAATCATCGA	0 350 ATCTTTGAAC ATCTTTGAAC ATCTTTGAAC ATCTTTGAAC ATCTTTGAAC ATCTTTGAAC ATCTTTGAAC ATCTTTGAAC	0 360 GCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG	O 371 CCCATTAGTA CCCATTAGTA CCCATTAGTA CCCATTAGTA CCCATTAGTA CCCATTAGTA CCCACTAGTA CCCACTAGTA	0 38 TTCTAGTGGG TTCTAGTGGG TTCTAGTGGG TTCTAGTGGG TTCTAGTGGG TTCTGGTGGG TTCTGGTGGG TTCTGGTGGG	0 390 CATGCCTATT CATGCCTATT CATGCCTATT CATGCCTATT CATGCCTATT CATGCCTATT CATGCCTATT CATGCCTATT	CGAGCGTCAT CGAGCGTCAT CGAGCGTCAT CGAGCGTCAT CGAGCGTCAT CGAGCGTCAT CGAGCGTCAT CGAGCGTCAT	41 TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT	0 42: AAGCCCT-GT AAGCCCT-GT AAGCCCT-GT AAGCCCT-GT AAGCCCT-GT AAGCCCTGT AAGCCCTGT AAGCCCCTGT	AGCTTAGCGT AGCTTAGCGT AGCTTAGCGC AGCTTAGCGC TGCTTAGCGC TGCTTAGCGT TGCTTAGCGT TGCTTAGCGT	TGGGAGTCCG TGGGAGTCCG TGGGAGTCCG TGGGAGTCCG TGGGAGTCCG TGGGAGTCTG TGGGAGTCTG TGGGCGTCTG	0 450    CTAAT CTAAT CTAT CTAT CTAT CGG CTAGCCT CGGCTC CGCCGTGCCC CGCCGTGCCC	408 408 408 422 374 381 439 429 429
SUT103 SUT105 ST2333 ST2527 ST2336 AJ390395 ST2584 SUT001 SUT004 ST2485	265 CGATAGT 265 CGATAGT 265 CGATAGT 265 CGATAGT 280 CGATAGT 232 CGATAGT 234 CGATAGT 294 CGATAGT 280 CGATAGT 281 CGATAGT	10 32 1     A TGTGAATTGC A TGTGAATTGC A TGTGAATTGC A TGTGAATTGC A TGTGAATTGC A TGTGAATTGC A TGTGAATTGC A TGTGAATTGC A TGTGAATTGC	0 33 AGAATTCAGT AGAATTCAGT AGAATTCAGT AGAATTCAGT AGAATTCAGT AGAATTCAGT AGAATTCAGT AGAATTCAGT AGAATTCAGT	0 34 GAATCATCGA GAATCATCGA GAATCATCGA GAATCATCGA GAATCATCGA GAATCATCGA GAATCATCGA GAATCATCGA GAATCATCGA GAATCATCGA	0 35 ATCTTTGAAC ATCTTTGAAC ATCTTTGAAC ATCTTTGAAC ATCTTTGAAC ATCTTTGAAC ATCTTTGAAC ATCTTTGAAC	0 360 GCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG	CCCATTAGTA CCCATTAGTA CCCATTAGTA CCCATTAGTA CCCATTAGTA CCCATTAGTA CCCATTAGTA CCCACTAGTA CCCACTAGTA	TTCTAGTGGG TTCTAGTGGG TTCTAGTGGG TTCTAGTGGG TTCTAGTGGG TTCTGGTGGGG TTCTGGTGGGG TTCTGGTGGGGG TTCTGGTGGG	0 390 CATGCCTATT CATGCCTATT CATGCCTATT CATGCCTATT CATGCCTATT CATGCCTATT CATGCCTATT CATGCCTATT CATGCCTATT	CGAGCGTCAT CGAGCGTCAT CGAGCGTCAT CGAGCGTCAT CGAGCGTCAT CGAGCGTCAT CGAGCGTCAT CGAGCGTCAT CGAGCGTCAT	41 TTCAACCTT TTCAACCTT TTCAACCTT TTCAACCTT TTCAACCTT TTCAACCTT TTCAACCTT TTCAACCTT TTCAACCTT TTCAACCTT	0 421 AAGCCCT-GT AAGCCCT-GT AAGCCCT-GT AAGCCCT-GT AAGCCCT-GT AAGCCCTGT AAGCCCCTGT AAGCCCCTGT AAGCCCCTGT	AGCTTAGCGT AGCTTAGCGC AGCTTAGCGC AGCTTAGCGC TGCTTAGCGT CGCTTAGCGT TGCTTAGCGT TGCTTAGCGT TGCTTAGCGT	A44 TGGGAGTCCG TGGGAGTCCG TGGGAGTCCG TGGGAGTCCG TGGGAGTCTG TGGGAGTCTG TGGGCGTCTG TGGGCGTCTG	0 450 	408 408 408 422 374 381 439 429 429 430 430
SUT103 SUT105 ST2333 ST2527 ST2336 AJ390395 ST2584 SUT001 SUT004 ST2485 SUT005	265 CGATAGTP 265 CGATAGTP 265 CGATAGTP 280 CGATAGTP 232 CGATAGTP 234 CGATAGTP 280 CGATAGTP 281 CGATAGTP 281 CGATAGTP 281 CGATAGTP 281 CGATAGTP 281 CGATAGTP	10 32 1	0 331 AGAATTCAGT AGAATTCAGT AGAATTCAGT AGAATTCAGT AGAATTCAGT AGAATTCAGT AGAATTCAGT AGAATTCAGT AGAATTCAGT	0 34 GAATCATCGA GAATCATCGA GAATCATCGA GAATCATCGA GAATCATCGA GAATCATCGA GAATCATCGA GAATCATCGA GAATCATCGA GAATCATCGA	0 35 	0 360 GCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG	0 371 	0 38 TTCTAGTGGG TTCTAGTGGG TTCTAGTGGG TTCTAGTGGG TTCTAGTGGG TTCTGGGGGGG TTCTGGTGGG TTCTGGTGGG TTCTGGTGGG	0 399 CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT	CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT	41 TTCAACCTT TTCAACCTT TTCAACCTT TTCAACCTT TTCAACCTT TTCAACCTT TTCAACCTT TTCAACCTT TTCAACCTT TTCAACCTT	0 421 AAGCCCT-GT AAGCCCT-GT AAGCCCT-GT AAGCCCT-GT AAGCCCT-GT AAGCCCTGT AAGCCCCTGT AAGCCCCTGT AAGCCCCTGT	AGCTTACCGT AGCTTACCGC AGCTTACCGC AGCTTACCGC AGCTTACCGC TGCTTACGGT TGCTTACCGT TGCTTACCGT TGCTTACCGT TGCTTACCGT	A44 TGGGAGTCCG TGGGAGTCCG TGGGAGTCCG TGGGAGTCCG TGGGAGCTCG TGGGCGTCTG TGGGCGTCTG TGGGCGTCTG	0 450 CTAAT CTAAT CTAT CTA-CT CGGCTC CGGCCC CGCCGTCCCC CGCCGTCCCC CGCCGTCCCC	408 408 408 422 374 381 439 429 430 430 429
SUT103 SUT105 ST2333 ST2527 ST2336 AJ390395 ST2584 SUT001 SUT004 ST2485 SUT005 SUT005 SUT262	265 CGATAGT 265 CGATAGT 265 CGATAGT 265 CGATAGT 280 CGATAGT 232 CGATAGT 230 CGATAGT 294 CGATAGT 281 CGATAGT 281 CGATAGT 281 CGATAGT 281 CGATAGT	10 32 1	0 331 А СААТТСАСТ АСААТТСАСТ АСААТТСАСТ АСААТТСАСТ АСААТТСАСТ АСААТТСАСТ АСААТТСАСТ АСААТТСАСТ АСААТТСАСТ АСААТТСАСТ АСААТТСАСТ	0 34 	0 351 I I ATCTTTGAAC ATCTTTGAAC ATCTTTGAAC ATCTTTGAAC ATCTTTGAAC ATCTTTGAAC ATCTTTGAAC ATCTTTGAAC ATCTTTGAAC	0 360 CCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG	о 371 СССАТТАСТА СССАТТАСТА СССАТТАСТА СССАТТАСТА СССАТТАСТА СССАТТАСТА СССАСТАСТА СССАСТАСТА СССАСТАСТА СССАСТАСТА	0 38 TTCTAGTGGG TTCTAGTGGG TTCTAGTGGG TTCTAGTGGG TTCTAGTGGG TTCTGGTGGG TTCTGGTGGG TTCTGGTGGG TTCTGGTGGG TTCTGGTGGG	0 390 CATGCCTATT CATGCCTATT CATGCCTATT CATGCCTATT CATGCCTATT CATGCCTATT CATGCCTATT CATGCCTATT CATGCCTATT CATGCCTATT	0 40 CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT	41 TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT	0 421 AAGCCCT-GT AAGCCCT-GT AAGCCCT-GT AAGCCCT-GT AAGCCCTCGT AAGCCCCTGT AAGCCCCTGT AAGCCCCTGT AAGCCCCTGT AAGCCCCTGT	0 43( 	44 GGGAGTCCG TGGGAGTCCG TGGGAGTCCG TGGGAGTCCG TGGGAGTCCG TGGGAGTCTG TGGGCGTCTG TGGGCGTCTG TGGGCGTCTG	0 450 	408 408 408 422 374 381 439 429 430 430 429 430
SUT103 SUT105 ST2333 ST2527 ST2536 AJ390395 ST2584 SUT001 SUT004 ST2465 SUT262 SUT262 SUT262	265 CGATAGTP 265 CGATAGTP 265 CGATAGTP 260 CGATAGTP 280 CGATAGTP 232 CGATAGTP 294 CGATAGTP 294 CGATAGTP 281 CGATAGTP 281 CGATAGTP 281 CGATAGTP 281 CGATAGTP 281 CGATAGTP	10 32 I	0 331 	0 34 	0 351 	0 360 CCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG	0 371 CCCATTAGTA CCCATTAGTA CCCATTAGTA CCCATTAGTA CCCATTAGTA CCCATTAGTA CCCACTAGTA CCCACTAGTA CCCACTAGTA CCCACTAGTA	0 38 TTCTAGTGGG TTCTAGTGGG TTCTAGTGGG TTCTAGTGGG TTCTGGTGGG TTCTGGTGGG TTCTGGTGGG TTCTGGTGGG TTCTGGTGGG	0 390 CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT	0 40 CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT	41 TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT	0 42: 	0 43( 	44 TGGGAGTCCG TGGGAGTCCG TGGGAGTCCG TGGGAGTCCG TGGGAGTCTG TGGGCGTCTG TGGGCGTCTG TGGGCGTCTG TGGGCGTCTG	0 450 	408 408 408 422 374 381 439 429 430 430 429 430 430
SUT103 SVT105 ST2333 ST2527 ST2336 AJ390395 ST2584 SUT001 SUT004 ST2485 SUT005 SUT262 ST2488 SUT167	265 CGATAGTP 265 CGATAGTP 265 CGATAGTP 265 CGATAGTP 280 CGATAGTP 232 CGATAGTP 234 CGATAGTP 281 CGATAGTP 281 CGATAGTP 281 CGATAGTP 281 CGATAGTP 281 CGATAGTP 281 CGATAGTP 281 CGATAGTP	10 32 1	0 331 	0 34 	0 351 	0 360 CCATTGOG GCACATTGOG GCACATTGOG GCACATTGOG GCACATTGOG GCACATTGOG GCACATTGOG GCACATTGOG GCACATTGOG GCACATTGOG GCACATTGOG GCACATTGOG GCACATTGOG	о 371 СССАТТАСТА СССАТТАСТА СССАТТАСТА СССАТТАСТА СССАТТАСТА СССАТТАСТА СССАСТАСТА СССАСТАСТА СССАСТАСТА СССАСТАСТА СССАСТАСТА СССАСТАСТА	0 38 TTCTAGTGGG TTCTAGTGGG TTCTAGTGGG TTCTAGTGGG TTCTAGTGGG TTCTGGTGGG TTCTGGTGGG TTCTGGTGGG TTCTGGTGGG TTCTGGTGGG	0 390 CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT	0 40 CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT	41. TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT	0 42: AAGCCCT-GT AAGCCCT-GT AAGCCCT-GT AAGCCCT-GT AAGCCCTCGT AAGCCCTCGT AAGCCCCTGT AAGCCCCTGT AAGCCCCTGT AAGCCCCTGT AAGCCCCTGT	0 43( 	44 TGGGAGTCCG TGGGAGTCCG TGGGAGTCCG TGGGAGTCCG TGGGAGTCCG TGGGAGTCTG TGGGCGTCTG TGGGCGTCTG TGGGCGTCTG TGGGCGTCTG	0 450 	408 408 408 422 374 381 439 429 430 430 430 430 430
SUT103 SUT105 ST2333 ST2527 ST2336 AJ390395 ST2584 SUT001 SUT004 ST2485 SUT004 SUT005 SUT262 SUT262 SUT262 SUT262 SUT267 JU2	265 CGATAGTP 265 CGATAGTP 265 CGATAGTP 280 CGATAGTP 280 CGATAGTP 280 CGATAGTP 280 CGATAGTP 281 CGATAGTP	10 32 1	0 331 	0 34 	0 351 	0 360 CCATTCCG CCACATTCCG CCACATTCCG CCACATTCCG CCACATTCCG CCACATTCCG CCACATTCCG CCACATTCCG CCACATTCCG CCACATTCCG CCACATTCCG CCACATTCCG CCACATTCCG CCACATTCCG	0 371 	0 38 TTCTACTGG TTCTACTGG TTCTACTGG TTCTACTGGG TTCTACTGGG TTCTACTGGG TTCTGCGGG TTCTGGTGGG TTCTGGTGGG TTCTGGTGGG TTCTGGTGGG	0 390 CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT	0 40 CCACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT	41 TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT	0 42: 	430 AGCTTAGGT AGCTTAGGT AGCTTAGGT GGCTTAGGT TGCTTAGGT TGCTTAGGT TGCTTAGGT TGCTTAGGT TGCTTAGGT TGCTTAGGT TGCTTAGGT TGCTTAGGT	44 TGGGAGTCCG TGGGAGTCCG TGGGAGTCCG TGGGAGTCCG TGGGAGTCTG TGGGCGTCTG TGGGCGTCTG TGGGCGTCTG TGGGCGTCTG TGGGCGTCTG TGGGCGTCTG	0 450 	408 408 408 422 374 381 439 429 430 429 430 429 430 429 430 429 430 429 430 429 430 429
SUT103 SUT105 ST2333 ST2527 ST2336 AJ390395 ST2584 SUT001 SUT004 SUT2045 SUT205 SUT262 SUT262 SUT262 SUT262 SUT167 Ju2 ST2579	265 CGATAGTP 265 CGATAGTP 265 CGATAGTP 260 CGATAGTP 230 CGATAGTP 230 CGATAGTP 230 CGATAGTP 281 CGATAGTP	10 32 1	0 331 	0 34 	0 351 	0 360 CCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG	0 371 	0 38 	0 390 CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT	0 40 CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT	41 TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT	0 42: 	0 43( 	44 TGGGAGTCCG TGGGAGTCCG TGGGAGTCCG TGGGAGTCCG TGGGAGTCCG TGGGAGTCTG TGGGCGTCTG TGGGCGTCTG TGGGCGTCTG TGGGCGTCTG TGGGCGTCTG TGGGCGTCCG TGGGAGTCCG	0 450 	408 408 408 422 374 381 439 429 430 430 429 430 430 430 430 430 424 424
SUT103 ST2105 ST2333 ST2527 ST2336 AJ390395 ST2584 SUT001 SUT004 SUT004 SUT005 SUT262 SUT262 SUT262 SUT262 SUT167 JU2 ST2448 SUT167 JU2	265 CGATAGT 265 CGATAGT 265 CGATAGT 265 CGATAGT 280 CGATAGT 232 CGATAGT 234 CGATAGT 294 CGATAGT 280 CGATAGT 281 CGATAGT 281 CGATAGT 281 CGATAGT 281 CGATAGT 281 CGATAGT 281 CGATAGT 281 CGATAGT 281 CGATAGT 281 CGATAGT	10 32 1	0 331 	0 34 	0 35: 	0 360 	0 371 	0 38 TTCTACTGGG TTCTACTGGG TTCTACTGGG TTCTACTGGG TTCTACTGGG TTCTGGGGGG TTCTGGGGGG TTCTGGTGGG TTCTGGTGGG TTCTGGTGGG TTCTGGTGGG TTCTGGTGGG TTCTGGTGGG TTCTGGTGGG TTCTGGTGGG	0 390 CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT	0 40 CCASCGTCAT CGACGGTCAT CGACGGTCAT CGACGGTCAT CGACGGTCAT CGACGGTCAT CGACGGTCAT CGACGGTCAT CGACGGTCAT CGACGGTCAT CGACGGTCAT CGACGGTCAT	41. TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT	0 42: AAGCCT-GT AAGCCT-GT AAGCCT-GT AAGCCT-GT AAGCCCT-GT AAGCCCTCGT AAGCCCTCGT AAGCCCCTGT AAGCCCCTGT AAGCCCCTGT AAGCCCCTGT AAGCCCTTCG AAGCCCTTCG	0 43( ) 43( AGCTTAGCGT AGCTTAGCGT AGCTTAGCGT TGCTTAGCGT TGCTTAGCGT TGCTTAGCGT TGCTTAGCGT TGCTTAGCGT TGCTTAGCGT TGCTTAGCGT TGCTTAGCGT -GCTTAGCGT -GCTTAGCGT	44 TGGGAGTCCG TGGGAGTCCG TGGGAGTCCG TGGGAGTCCG TGGGAGTCCG TGGGCGTCTG TGGGCGTCTG TGGGCGTCTG TGGGCGTCTG TGGGCGTCTG TGGGCGTCTG TGGGCGTCTG TGGGCGTCTG TGGGCGTCCG TGGGAGTCCG TGGGAGTCCG	0 450 	408 408 408 422 374 381 429 430 430 430 430 430 430 430 430 430 430
SUT103 SUT105 ST2333 ST2527 ST2336 AJ390395 ST2584 SUT001 SUT004 ST2485 SUT005 SUT065 SUT262 ST2448 SUT167 JU2 ST2579 ST2406 SUT259	265 CGATAGTA 265 CGATAGTA 265 CGATAGTA 280 CGATAGTA 280 CGATAGTA 280 CGATAGTA 280 CGATAGTA 281 CGATAGTA 281 CGATAGTA 281 CGATAGTA 281 CGATAGTA 281 CGATAGTA 281 CGATAGTA 281 CGATAGTA 281 CGATAGTA 281 CGATAGTA 280 CGATAGTA 280 CGATAGTA 280 CGATAGTA 280 CGATAGTA 280 CGATAGTA	10 32 1	0 331 	0 34 	0 355 	0 360 	0 371 	0 38 TTCTAGTGGG TTCTAGTGGG TTCTAGTGGG TTCTAGTGGG TTCTAGTGGG TTCTGGGGGGG TTCTGGTGGG TTCTGGTGGG TTCTGGTGGG TTCTGGTGGG TTCTGGTGGG TTCTGGTGGG TTCTGGTGGG TTCTGGTGGG TTCTGGTGGG TTCTGGTGGG TTCTGGTGGG TTCTGGTGGG TTCTGGTGGG	0 390 CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT	0 40 CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT	41. TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT	0 42: AAGCCT-GT AAGCCT-GT AAGCCT-GT AAGCCT-GT AAGCCCT-GT AAGCCCTGT AAGCCCTGT AAGCCCTGT AAGCCCTGT AAGCCCTGT AAGCCCTGT AAGCCCTGT AAGCCCTTG AAGCCCTTG AAGCCCTTG AAGCCCTTG AAGCCCTTG	0 43( 	44 TGGAGATCCG TGGGAGTCCG TGGGAGTCCG TGGGAGTCCG TGGGAGTCCG TGGGAGTCTG TGGGCGTCTG TGGGCGTCTG TGGGCGTCTG TGGGCGTCTG TGGGAGTCCG TGGGAGTCCG TGGGAGTCCG TGGGAGTCCC	0 450     CTAAT CTAAT CTAAT CTACT CTACCT CGCCTC CGCCTCCCC CGCCGTCCCC CGCCGTCCCC CGCCGTCCCC CGCCGTCCCC CGCCGTCCCC CGTCCCC CGTCCCC CGTCCCC CGTCCCC	408 408 408 422 374 381 429 430 429 430 430 430 430 430 430 424 430 430 430 430 430 430 430 430 430 43
SUT103 SUT105 ST2333 ST2527 ST2336 AJ390395 ST2584 SUT001 ST2485 SUT004 ST2485 SUT005 SUT262 ST2448 SUT167 Ju2 ST2579 ST2406 SUT025 SUT025 SUT025	265 CGATAGTP 265 CGATAGTP 265 CGATAGTP 260 CGATAGTP 280 CGATAGTP 280 CGATAGTP 294 CGATAGTP 281 CGATAGTP 282 CGATAGTP 282 CGATAGTP	10 32 1	0 331 	0 34 	0 351 	0 360 CCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG GCACATTGCG	0 371 	0 38 	0 390 CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT	0 40 CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT	41. TTCAACCTT	0 42: 	0 430 AGCTTAGGT AGCTTAGGT AGCTTAGGT GGCTTAGGT TGCTTAGGT TGCTTAGGT TGCTTAGGT TGCTTAGGT TGCTTAGGT TGCTTAGGT GCTTAGGT GCTTAGGT -GCTTAGGT -GCTTAGGT -GCTTAGGT -GCTTAGGT	44 TGGGAGTCCG TGGGAGTCCG TGGGAGTCCG TGGGAGTCCG TGGGAGTCCG TGGGAGTCTG TGGGCGTCTG TGGGCGTCTG TGGGCGTCTG TGGGCGTCTG TGGGCGTCTG TGGGAGTCCG TGGGAGTCCG TGGGAGTCCC TGGGAGTCCC	0 450 	408 408 408 422 374 381 439 429 430 429 430 429 430 429 430 429 430 420 424 420 424 424
SUT103 SUT105 ST2333 ST2527 ST2336 AJ390395 ST2584 SUT001 SUT004 SUT005 SUT262 SUT262 SUT262 SUT262 SUT262 ST2448 SUT167 JU2 ST2579 ST22406 SUT025 SUT220 SUT220 SUT220	265 CGATAGT 265 CGATAGT 265 CGATAGT 265 CGATAGT 280 CGATAGT 232 CGATAGT 232 CGATAGT 294 CGATAGT 294 CGATAGT 281 CGATAGT 282 CGATAGT 282 CGATAGT 282 CGATAGT	10 32 1	0 331 	0 34 	0 351 	0 360 CCACATTGCG	0 371 	0 38 	0 390 CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT CATECCTATT	0 40 CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT	41. TTCAACCTT	0 42: 	0 430 AGCTAGCGT AGCTAGCGT AGCTAGCGT AGCTAGCGT TGCTAGCGT TGCTAGCGT TGCTAGCGT TGCTAGCGT TGCTAGCGT TGCTAGCGT TGCTAGCGT GCTAGCGT GCTAGCGT GCTAGCGT GCTAGCGT GCTAGCGT GCTAGCGT GCTAGCGT GCTAGCGT	44 TGGGAGTCCG TGGGAGTCCG TGGGAGTCCG TGGGAGTCCG TGGGAGTCCG TGGGAGTCTG TGGGCGTCTG TGGGCGTCTG TGGGCGTCTG TGGGCGTCTG TGGGCGTCTG TGGGCGTCTG TGGGCGTCTG TGGGAGTCCG TGGGAGTCCG TGGGAGTCCG TGGGAGTCCG TGGGATCCG TGGGATCCG	0 450 	408 408 408 408 422 374 381 439 429 430 430 430 429 430 429 430 429 420 424 426 424 426 424
SUT103 SUT105 ST2333 ST2527 ST2336 AJ390395 ST2584 SUT001 SUT004 ST2485 SUT005 SUT025 SUT262 ST2448 SUT167 JU2 ST2579 ST2406 SUT025 SUT220 ST2322 AJ390406 ST2322 AJ390406	265 CGATAGTA 265 CGATAGTA 265 CGATAGTA 280 CGATAGTA 280 CGATAGTA 280 CGATAGTA 280 CGATAGTA 281 CGATAGTA 281 CGATAGTA 281 CGATAGTA 281 CGATAGTA 281 CGATAGTA 281 CGATAGTA 281 CGATAGTA 280 C	10 32 1	0 331 	0 34 	0 351 	0 360 CCATTCCG GCACATTCCG	0 371 	0 38 TTCTASTGG TTCTASTGG TTCTASTGGG TTCTASTGGG TTCTASTGGG TTCTGSTGGG TTCTGGTGGG TTCTGGTGGG TTCTGGTGGG TTCTGGTGGG TTCTGGTGGG TTCTGGTGGG TTCTGGTGGG TTCTGGTGGG TTCTGGTGGG TTCTGGTGGG TTCTGGTGGG TTCTGGTGGG TTCTGGCGGG TTCTGGCGGG TTCTGGCGGG TTCTGGCGGG TTCTGGCGGG TTCTGGCGGG TTCTGGCGGG TTCTGCCGG TTCTGCCGG TTCTGCCGG TTCTGCCGG TTCTGCCG TTCTGCCGG TTCTGCCGG TTCTGCCGG TTCTGCCG TTCTGC TTCTGCCG TTCTGCCG TTCTGCCG TTCTGCCG TTCTGCCG TTCTGCCG TTCTGCCG TTCTGCCG TTCTGCCG TTCTGCCG TTCTGCCG TTCTGCCG TTCTGCCG TTCTGCCG TTCTGCCG TTCTGCCG TTCTGCC TTCTGCCG TTCTGCC TTCTGCCC T	0 390 	0 40 CCACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT	41. TTCAACCCTT	0 42: 	0 430 AGCTTAGGT AGCTTAGGT AGCTTAGGT GGCTTAGGT GGCTTAGGT GGCTTAGGT GGCTTAGGT GGCTTAGGT GGCTTAGGT GGCTTAGGT GGCTTAGGT GGCTTAGGT GGCTTAGGT GGCTTAGGT GGCTTAGGT GGCTTAGGT	44 TGGGAGTCCG TGGGAGTCCG TGGGAGTCCG TGGGAGTCCG TGGGAGTCCG TGGGAGTCTG TGGGCGTCTG TGGGCGTCTG TGGGCGTCTG TGGGCGTCTG TGGGCGTCTG TGGGAGTCCG TGGGAGTCCG TGGGAGTCCG TGGGAGTCCG TGGGAATCCG TGGGAATCCG TGGGAATCCG TGGGAATCCG	0 450     CTAAT CTAAT CTAAT CTACT CTACCT CGGCTC CGCCGTGCCC CGCCGTGCCC CGCCGTGCCC CGCCGTGCCC CGTCCCC CGCCCCCCCC CGCCCCCCC CGCCCCCCC CGCCCCCCC CGCCCCCCC CGCCCCCCC CGCCCCCCC CGCCCCCCC CGCCCCCCC CGCCCCCCC CGCCCCCCC CGCCCCCCC CGCCCCCCC CGCCCCCCC CGCCCCCCC CGCCCCCCC CGCCCCCCC CGCCCCCCC CGCCCCCCC CGCCCCCCCC	408 408 408 422 374 439 429 430 430 430 430 424 430 430 424 430 430 430 430 430 430 430 430 430 43
SUT103 SUT105 ST2333 ST2527 ST2336 AJ390395 ST2584 SUT001 SUT004 ST2485 SUT005 SUT262 ST2448 SUT167 JU2 ST2579 ST2406 SUT025 SUT05	265 CGATAAGT2 265 CGATAAGT2 265 CGATAAGT2 260 CGATAAGT2 280 CGATAAGT2 280 CGATAAGT2 294 CGATAAGT2 281 CGATAAGT2 282 CGATAAGT2 282 CGATAAGT2 282 CGATAAGT2 282 CGATAAGT2 284 CGATAAGT2 284 CGATAAGT2 284 CGATAAGT2 284 CGATAAGT2 284 CGATAAGT2 284 CGATAAGT2 285 CGATAAGT2 284 CGATAAGT2 284 CGATAAGT2 285 CGATAAGT2 284 CGATAAGT2 284 CGATAAGT2 285 CGATAAGT2 285 CGATAAGT2 285 CGATAAGT2 284 CGATAAGT2 285 CGATAAGT2 285 CGATAAGT2 285 CGATAAGT2 285 CGATAGT2 285	10 32 1	0 331 	0 34 	0 351 	0 360 CCACATTCCG GCACATTCCG	0 371 	0 38 	0 390 CATECCTATT	0 40 CCACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT	41. TTCAACCTT	0 42: AGCCCT-GT AAGCCCT-GT AAGCCCT-GT AAGCCCT-GT AAGCCCT-GT AAGCCCTGT AAGCCCTGT AAGCCCTGT AAGCCCTGT AAGCCCTGT AAGCCCTGT AAGCCCTGG AAGCCCTGG AAGCCCTGG AAGCCCTGG AAGCCCTGG AAGCCCTGG AAGCCCTGG AAGCCCTGG AAGCCCTGG AAGCCCTGG AAGCCCTGG AAGCCCTGG AAGCCCTGG AAGCCCTGG AAGCCCTGG AAGCCCTGG AAGCCCTGG AAGCCCCGG AAGCCCCGG AAGCCCCGG AAGCCCCGG AAGCCCCGG	0 430 AGCTTAGGT AGCTTAGGT AGCTTAGGT GGCTAGGT TGCTTAGGT TGCTTAGGT TGCTTAGGT TGCTTAGGT TGCTTAGGT TGCTTAGGT GGCTTAGGT -GCTTAGGT -GCTTAGGT -GCTTAGGT -GCTTAGGT -GCTTAGGT -GCTTAGGT -GCTTAGGT	44 TGGGAGTCCG TGGGAGTCCG TGGGAGTCCG TGGGAGTCCG TGGGAGTCCG TGGGAGTCTG TGGGCGTCTG TGGGCGTCTG TGGGCGTCTG TGGGCGTCTG TGGGCGTCTG TGGGAGTCCC TGGGAGTCC TGGGAGTCC TGGGAGCC TGGGAGTCC TGGGAGTCC TGGGAGTCC TGGGAGTCC TGGGAGTCC TGGGAG	0 450     CTAAT CTAAT CTAAT CTACT CTACCT CTACCCC CGCCTCCCC CGCCGTCCCC CGCCGTCCCC CGCCGTCCCC CGCCGTCCCC CGCCGTCCCC CGTCCCC CGTCCCC CGTCT-TCAG CGTCT-CAG CGTCT-CCA CGCCCCC	408 408 408 422 374 439 429 430 430 430 430 424 430 430 424 430 430 424 430 430 430 430 430 432 430 430 432 430 430 430 430 430 430 430 430 430 430
SUT103 SUT105 ST2333 ST2527 ST2336 AJ390395 ST2584 SUT001 SUT004 SUT005 SUT262 SUT262 SUT262 SUT262 SUT262 SUT262 SUT279 ST2406 SUT2579 ST2406 SUT225 SUT220 ST2332 AJ390406 ST2436 ST2436 ST2436	265 CGATAGTP 265 CGATAGTP 265 CGATAGTP 260 CGATAGTP 230 CGATAGTP 230 CGATAGTP 230 CGATAGTP 294 CGATAGTP 281 CGATAGTP 281 CGATAGTP 281 CGATAGTP 281 CGATAGTP 281 CGATAGTP 281 CGATAGTP 281 CGATAGTP 281 CGATAGTP 281 CGATAGTP 282 CGATAGTP 284 C	10 32 1	0 331 	0 34 	0 351 	0 360 CCACATTGCG	0 371 	0 38 	0 390 CATECCTATT	0 CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT CGACCGTCAT	41. TTCAACCTT	0 42: AGCCCT-GT AGCCCT-GT AGCCCT-GT AGCCCT-GT AGCCCT-GT AGCCCT-GT AGCCCT-GT AGCCCTGT AGCCCTGT AGCCCCTGT AGCCCCTGT AGCCCCTGT AGCCCCTGT AGCCCCTGT AGCCCCCGT AGCCCCCGT AGCCCCCGT AGCCCCCGT AGCCCCCGT	0 430 AGCTAGCGT AGCTAGCGT AGCTAGCGT AGCTAGCGT TGCTAGCGT TGCTAGCGT TGCTAGCGT TGCTAGCGT TGCTAGCGT TGCTAGCGT TGCTAGCGT -GCTAGCGT -GCTAGCGT -GCTAGCGT -GCTAGCGT AGCTAGCGT TGCTAGCGT TGCTAGCGT TGCTAGCGT	44 TGGGAGTCCG TGGGAGTCCG TGGGAGTCCG TGGGAGTCCG TGGGAGTCCG TGGGAGTCTG TGGGCGTCTG TGGGCGTCTG TGGGCGTCTG TGGGCGTCTG TGGGAGTCCG TGGGAGTCCG TGGGAGTCCG TGGGAGTCCG TGGGAGTCCG TGGGAGTCTG TGGGAGTCTG	0 450 	408 408 408 422 374 430 429 430 430 430 430 430 430 430 430 430 430
SUT103 SUT105 ST2133 ST2527 ST2336 AJ390395 ST2584 SUT001 SUT004 ST2485 SUT005 SUT262 ST2485 SUT167 Ju2 ST2579 ST2406 SUT220 ST2406 SUT220 ST2322 AJ390406 ST2436 ST2433 Ju1	265 CGATAGT 265 CGATAGT 265 CGATAGT 265 CGATAGT 280 CGATAGT 280 CGATAGT 280 CGATAGT 281 CGATAGT 281 CGATAGT 281 CGATAGT 281 CGATAGT 281 CGATAGT 281 CGATAGT 281 CGATAGT 281 CGATAGT 280 C	10 32 1	0 331 	0 34 	0 351 	0 360 CCATTGG GCACATTGG	0 371 	0 38 TTCTASTGG TTCTASTGG TTCTASTGGG TTCTASTGGG TTCTASTGGG TTCTGTGGG TTCTGGTGGG TTCTGGTGGG TTCTGGTGGG TTCTGGTGGG TTCTGGTGGG TTCTGGTGGG TTCTGGTGGG TTCTGGTGGG TTCTGGTGGG TTCTGGTGGG TTCTGGTGGG TTCTAGTGGG TTCTAGTGGG TTCTAGTGGG TTCTAGTGGG TTCTAGTGGG TTCTAGTGGG TTCTAGTGGG TTCTAGTGGG	0 390 CATECCTATT	0 CGACCGTCAT	41. TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCAACCCTT TTCACCCCTT TTCACCCCTT TTCACCCCTT TTCACCCCTT TTCACCCCTT TTCACCCCTT TTCACCCCTT TTCACCCCTT TTCACCCCTT TTCACCCCTT TTCACCCCTT TTCACCCCTT TTCACCCCTT TTCACCCCTT	0 42: 	0 430 AGCTTAGCGT AGCTTAGCGT AGCTTAGCGT GGCTTAGCGT GGCTTAGCGT GGCTTAGCGT GGCTTAGCGT GGCTTAGCGT GGCTTAGCGT GGCTTAGCGT GGCTTAGCGT GGCTTAGCGT GGCTTAGCGT GGCTTAGCGT GGCTTAGCGT GGCTTAGCGT GGCTTAGCGT GGCTTAGCGT GGCTTAGCGT GGCTTAGCGT GGCTTAGCGT	44 TGGGAGTCCG TGGGAGTCCG TGGGAGTCCG TGGGAGTCCG TGGGAGTCCG TGGGAGTCTG TGGGCGTCTG TGGGCGTCTG TGGGCGTCTG TGGGCGTCTG TGGGAGTCCG TGGGAGTCCG TGGGAGTCCG TGGGAGTCCG TGGGAGTCTG TGGGAGTCTG TGGGAGTCTG	0 450     CTAAT CTAAT CTAAT CTACT CTACCT CGGCCCT CGCCGTCC CGCCGTCCCC CGCCGTCCCC CGCCGTCCCC CGCCGTCCCC CGTCCCC CGCCCCCCCC CGTCCCC CGTCCCC CGCCCCCCCC CGTCCCC CGCCCCCCCC CGTCCCC CGCCCCCCCC CGCCCCCCCC CGCCCCCCC CGCCCCCCCCC CGCCCCCCC CGCCCCCCC CGCCCCCCC CGCCCCCCC CGCCCCCCC CGCCCCCCC CGCCCCCCC CGCCCCCCC CGCCCCCC CGCCCCCCC CGCCCCCCC CGCCCCCCCCC CGCCCCCCC CGCCCCCCCCC CGCCCCCCCCC CGCCCCCCCCC CGCCCCCCCC CGCCCCCCCC CGCCCCCC CGCCCCCC CGCCCCCC CGCCCCCC CGCCCCCC CGCCCCCC CGCCCCCC CGCCCCCC CGCCCCCC CGCCCCCC CGCCCCC CGCCCCCC CGCCCCCC CGCCCCCC CGCCCCCC CGCCCCCC CGCCCCCC CGCCCCCC CGCCCCCC CGCCCCCC CGCCCCC CGCCCCC CGCCCCC CGCCCCC CGCCCA CGCCCA	408 408 408 422 374 439 429 430 429 430 429 430 424 430 424 430 424 430 424 430 424 430 430 424 430 360 361 350 350

Figure 7C. (Continued).

		46	0 470	48	0 490	50	51	0 520	0 53	540	) 55	560	57	3	
SUT103	409	TTTAGCGGCT	CCTTAAAGTT	ATTGGCGGAG	TTATAGCGTA	CTCTAAGCGT	AGTAATTT-T	TATCTCG	CTTCTGTAGT	GGCCCTAAC-	TGTTAGCCAT	AAAACCCCTA	TATTTTTCTA	AT	525
SUT105	409	TTTAGCGGCT	CCTTAAAGTT	ATTGGCGGAG	TTATAGCGTA	CTCTAAGCGT	AGTAATTT-T	TATCTCG	CTTCTGTAGT	GGCCCTAAC-	TGTTAGCCAT	AAAACCCCTA	TATTTTTCTA	AT	525
ST2333	409	TTTAGCGGTT	CCTTAAAGTT	ATTGGCGGAG	TTATAGCGTA	CTCTAAGCGT	AGTAATTT-T	TATCTCG	CTTCTGTAGT	AGCCCTAAC-	TGTTAGCCAT	AAAACCCCTA	TATTTTTCTA	AT	525
ST2527	423	TTTAGCGGTT	CCTTAAAGTT	ATTGGCGGAG	TTATAGCGTA	CTCTAAGCGT	AGTAATTT-T	TATCTCG	CTTCTGTAGT	AGCCCTAAC-	TGTTAGCCAT	AAAACCCCTA	TATTTTTCTA	AT	539
ST2336	375	CACCGTAGCT	CCCCAAAGTC	AGTGGCGGAG	TCGGCTCACA	CTCTAGACGT	AGTAATTT	CTCACCT	CGCCTATAGT	TGGACCGGT-	CCCCTGCCGT	AAAACGCCCA	AGTCTTTAAA	<b>A</b> -	489
AJ390395	382	CGGCGCAGTT	CCTTAAATTC	ATTGGCGGAG	<b>CTGTGGCACA</b>	CTCTAGGCGT	AGTAGTTTAA	CACCTCG	CCTCTAGAGT	GGCCGCGGT-	TACTGGCCGT	AAAACCCCTA	TATTTCTAGT		497
ST2584	440	AGGCCGAGTT	CCTTAAATT-	AGTGGCGGAG	T-ACAGCACA	ACCTAAGCGT	AGTAGGTTA-	CCTCG	CTCCCGGGGA	GTCTGTGGCG	CCTGCGTAAA	AAAAAACCCT	AAACCTTCTA		551
SUT001	430	TGGCGCAGTG	CCCTAAATCT	ATCGGCGGAG	CCGTAGCACA	CTCTGAGCGT	AGTAATT-AC	AGTT-CCTCG	CTCCTGCAGT	GGCCGCGGC-	GGCTGGC-GT	TAACCCCT	ACAACTTCTA	GT	545
SUT004	431	TGGCGCAGTG	CCCTAAATCT	ATCGGCGGAG	CCGTAGCACA	CTCTGAGCGT	AGTAATT-AC	AGTT-CCTCG	CTCCTGCAGT	GGCCGCGGC-	GGCTGGCCGT	TAAACCCCCT	ACAACTTCTA	GT	549
ST2485	431	TGGCGCAGTG	CCCTAAATCT	ATCGGCGGAG	CCGTAGCACA	CTCTGAGCGT	AGTAATT-AC	AGTT-CCTCG	CTCCTGCAGT	GGCCGCGGC-	GGCTGGCCGT	TAAACCCCCT	ACAACTTCTA	GT	549
SUT005	430	TGGCGCAGTG	CCCTAAATCC	ATCGGCGGAG	CCGTAGCACA	CTCTGAGCGT	AGTAATT-AC	AGTT-CCTCG	CTCCTGCAGT	G-CCGCGGC-	GGCTTGGCGT	AACCCCCT	ATA-CTTCTA	GT	544
SUT262	431	TGGCGCAGTG	CCCTAAATCT	ATCGGCGGAG	CCGTAGCACA	CTCTGAGCGT	AGTAATT-AC	AGTT-CCTCG	CTCCTGCAGT	GGCCGCGGC-	GGCTGGCCGT	TAAACCCCCT	ATAACTTCTA	GT	549
ST2448	431	TGGCGCAGTG	CCCTAAATC-	ATCGGCGGAG	CCG-AGCACA	CTCTGAGCGT	AGTAATAC	AGTCCTCG	CTCCTGCAGT	GGCCGCGGC-	GGCT-GCCGT	AACCCCT	ATA-CTTCTA	GT	540
SUT167	431	CGGCGCAGTG	CCCTAAATCC	ATCGGCGGAG	CCGTAGCACA	CTCTGAGCGT	AGTAATT-AC	AGTT-CCTCG	CTCCTGCAGT	GGCCGCGGC-	GGCT-GCCGT	TAAA-CCCCT	ATAACTTCTA	GT	547
Ju2	424	-GGCGCGGT-	CCCCAAAGTC	ATTGGCGGCT	TCGCAGCCCA	CTCTGAGCGT	AGTAATCAAC	TGGT-TCTCG	CTCCTGCAGT	GGCCGGCGG-	AGCCCGCCGT	AAAACCCCCC	СТАТААСТАА	GT	542
ST2579	426	-GGCGCGGTT	CCCCAAAGTC	ATTGGCGGCT	T-GCAGCCAA	CTCTGAGCGT	AGTAATCAAC	TGTT-TCTCG	CTCCTGCAGT	GGCCGGCGG-	AGCC-GCCGT	AAAACCCCCC	СТАТААСТАА	GT	543
ST2406	425	CGGCGCGGTT	CCCCAAAGTC	ATTGGCGGCT	TCGCAGCCCA	CTCTGAGCGT	AGTAATCAAC	TGTT-TCTCG	CTCCTGCAGT	GGCCGCGGC-	AGCCCGCCGT	AAAACCCCCC	<b>CTATAACTTA</b>	GT	544
SUT025	407	GGGCGGGGTT	CCTTAAAGTC	ATTGGCGGCG	TCGCAGCCCA	CTCTGAGCGT	AGTAATCTAC	TGTT-TCTCG	CTCCTGCAGT	GGCCGCGGCT	GGCTTGCCGT	AAAACCCCCT	ATATGTCTGA	G-	526
SUT220	430	GGGCGCGGTT	CCCTAAATTC	ATCGGCGGCG	CCGGGGGCGTC	TTCTGAGCGT	AGTAATTTAT	TATCTCG	C-CCTGAAGC	TAGCCCCGTA	CGCCCGCCGT	AAAACCCCCC	AACTACCTTG	<b>T</b> -	546
ST2332	432	GGGCGCGGTT	CCCTAAATTC	ATCGGCGGCG	CCGGGGGCGTC	TTCTGAGCGT	AGTAATTTAT	TATCTCG	C-CCTGAAGC	TAGCTCCGTA	CGCCCGCCGT	AAAACCCCCC	AACTACCGTA	СТ	549
AJ390406	390	CGGCGCAGCT	CCTCAAAGTC	AGTGGCGGAG	TCGGGTCGTG	CTCTGAGCGT	AGTAGTTAAT	ATCTCG	CTTCTGCGGT	GCCCCCGGC-	TGCCTGCCGT	AAAACCCCCC	CCTATACTTT	CG	506
ST2436	361	GGCCGCAGTT	CCTCAAAGTC	AGTGGCGGAG	TTGTAGCACA	CTCTAAGCGT	AGTAGTTTC	CATTGCCTCG	CATGCAGAGC	GGCCTCAGC-	TGCCAGCCGT	AAAGCCCTAT	ACTTCTTAGT		479
ST2473	362	GGCCGCAGTT	CCTCAAAGTC	AGTGGCGGAG	TTGTAGCACA	CTCTAAGCGT	AGTAGTTTC	CATTGCCTCG	CATGCAGAGC	GGCCTCAGC-	TGCCAGCCGT	AAAGCCCTAT	ACTTCTTAGT		480
Ju1	360	GGCCGCAGTT	CCTCAAAGTC	AGTGGCGGAG	TTGTAGCACA	CTCTAAGCGT	AGTAGTTTTC	CATTGCCTCG	C-TGCAGAGC	GGCCTCAGC-	TGCCAGCCGT	AAAGCCCTAT	ACTTCT-AGT		476
ST2313 H.n	361	GGCCGCAGTT	CCTCAAAGTC	AGTGGCGGAG	TTGTAGCACA	CTCTAAGCGT	AGTAGTTTTC	CATTGCCTCG	C-TGCAGAGC	GGCCTCAGC-	TGCCAGCCGT	AAAGCCCTAT	ACTTCTTAGT		478
_															

Figure 7C. (Continued).

		10	20	30	40	50	60	70	80	90	10	0 11	0 12	0 130	140	150	16	0 17	18	0	
2020	4														1						
Ju2	1	CTATICGAGC	GTRATITCAR	CCCTTARGCC	C-TICGCCTT	ARCCGT-GGG	AGTCCGCGTC	CCC-CCC	CCCCT-CCCC	ARAGTCATTG	GCGGCTTCGC	ACCCCACTCT	GAGCGTAGTA	ATCAACTGGT	TCTCGCTCCT	GCAGTGGCCG	GCGGAGCCCG	CCGT-ARAAC	CCCCCCTATA	ACTARCT	179
ST2579	1	CTATICGAGC	GTAATTTCAA	CCCTTAAGCC	C-TICGGCTT	AAGCGTTGGG	AGTCCGCGTC	CCC-GGC	GCGGTTCCCC	AAAGTCATTG	GCGGCTT-GC	AGCCAACTCT	GAGCGTAGTA	ATCAACTGTT	TCTCGCTCCT	GCAGTGGCCG	GCGGAGCC-G	CCGT-AAAAC	CCCCCCTATA	ACTAAGT	179
ST2406	1	CTATTCGAGC	GTCATITCAA	CCCTTAAGCC	C-TTCGGCTT	A-GCGTTGGG	AGTCCGCGTC	CCCCGGC	GCGGTTCCCC	AAAGTCATTG	GCGGCTTCGC	AGCCCACTCT	GAGCGTAGTA	ATCAACTGTT	TETEGETEET	GCAGTGGCCG	CGGCAGCCCG	CCGT-AAAAC	CCCCCCTATA	ACTTAGT	181
SUT025	1	CTATICGAGC	GTCATITICGA	CCCTTAAGCC	C-TOCCOCTT	A-GCGTIGGG	AGTOCCCCCC	CCCACCCCCC	CECETTCETT	ARAGTCATTG	CCCCCCTCCC	ACCCCACTCT	GAGCGTAGTA	ATCTACTOTT	TCTCGCTCCT	GCAGTGGCCG	CECCECCTTC	CCGT-AAAAC	CCCCTATATC	TCTGAG-	183
ST2333	1	CTATICGAGC	GTCATTTCAA	CCCTTAAGCC	CTGTA-GCTT	A-GCGCTGGG	AGTCCGCTAA	TTITA	GCGGTTCCTT	AAAGTTATTG	GCGGAGTTAT	AGCGTACTCT	AAGCGTAGTA	ATTTITA	TCTCCCTTCT	GTAGTAGCCC	TAACIGTTAG	CCATAAAA	CCCCTATATT	TTTCTAA	175
ST2527	1	CTATICGAGC	GTCATTTCAA	CCCTTAAGCC	CTGTA-GCTT	A-GCGCTGGG	AGTCCGCTA-	TTTTA	GCGGTTCCTT	AAAGTTATTG	GCGGAGTTAT	AGCGTACTCT	AAGCGTAGTA	ATTTTTA	TCTCGCTTCT	GTAGTAGCCC	TAACTGTTAG	CCATAAAA	CCCCTATATT	TTTCTAA	174
SUT105	1	CTATICGAGC	GTCATTICAR	CCCTTAAGCC	CTGTA-GCTT	A-GCGTTGGG	AGTCCGCTAR	TTTTA	GCGGCTCCTT	ARAGTTATIG	GCGGAGTTAT	ACCETACTCT	ARGCGTAGTA	ATTTTTA	TCTCGCTTCT	GTAGTGGCCC	TAACTGTTAG	CCATAAAA	CCCCTATATT	TITCTAA	175
SUT285	1	CTATICGAGC	GTCATTACAA	CCCTTAAGCC	C-TGTTGCTT	A-GCGTIGGG	AATCTACGGC	TTAGGCG	-TAGTICCTC	ARAATTAGTG	GCGGAGTTAT	AGCACTCT	CAGCGTAGTA	ATTTG	CCTCGCTTCT	GRECTE-CTE	TAGCTGCCTG	CCGTAAAA	CCC-TATAC-	TTCTAGT	169
SUT081	1	CTATICGAGC	GTCATTACAA	CCCTTAAGCC	C-TGTTGCTT	A-GCGTTGGG	AATCTACGGC	TTAGGCG	-TAGTTCCTC	AAAATTAGTG	GCGGAGTTAT	AGCACTCT	CAGCGTAGTA	ATTTG	CCTCGCTTCT	GAGCTG-CTG	TAGCTGCCTG	CCGTAAAA	CCCCTATAC-	TTCTAGT	170
SUT244	1	CTATTCGAGC	GTCATTACAR	CCCTTARGCC	C-TGTTGCTT	R-GCGTTGGG	AATCTACGGC	TTAGGCG	-TAGTTCCTC	ARARTTAGTG	GCGGAGTTAT	AGCACTCT	CGGCGTAGTA	ATTTG	CCTCGCTTCT	GAGCTG-CTG	TAGCTGCCTG	CCGTAAAA	CCC-TATAC-	TTCTAGT	169
SUT251	1	CTATICGAGC	<b>GTCATTACAA</b>	CCCTTARGCC	C-TOTTCCTT	A-GCGTIGGG	ARTCTACCCC	TTREGCC	-TAGTTCCTC	ARAATTAGTG	GCGGAGTTAT	ACCAC TCT	TAGCGTAGTA	ATTTG	CCTCGCTTCT	GRECTE-CTC	TACCICCCTC	CCCTAAAA	CCCCTATAC-	TTCTAGT	170
SUT242	1	CTATICGAGC	GTCATTACAA	CCCTTAAGCC	C-TGTTGCTT	A-GCGTTGGG	AATCTACGGC	TTAGGCG	-TAGTICCTT	AAAATTAGTG	GCGGAGTTAT	AGCACTCT	TAGCGTAGTA	ATTTG	CCTCGCTTCT	GAGCTG-CTG	TAGCTGCCTG	CCGTAAAA	CCCCTATAC-	TTCTAGT	170
SUT009	1	CTATICGAGC	<b>GTCATTATAA</b>	CCCTTAAGCC	T-TGTTGCTT	A-GCGTTGGG	AATCTACCCC	TCACTAAGGG	GTAGTICCTT	AAATGTAGTG	GCGGGGGTTAT	AGCCCACTCT	AAGCGTAGTA	GTTTA	ACTCGCTTTC	AGGGAGGCTG	TAGCTGCTTG	CCGT-AAAAC	CCCTTATAAC	TTATAGG	179
SUT010	1	CTATICGAGC	<b>GTCATTATAA</b>	CCCTTARGCC	T-TOTTCOTT	A-ACCGTGGG	AATCTACCCC	TCACTGAGGG	GTAGTTCCTT	ARATCTACTC	GCGGGGGTTAT	ACCCCACTCT	ARCCGTAGTA	GTTTA	ACTCCCTTTC	ACCCACCCTC	TACCTCCTTC	CCGT-AAAAC	CCCTTATAAC	TTATAGG	179
AJ390397	1	CTATTCGAGC	GTCATTATAA	CCCTTAAGCC	T-TGTTGCTT	A-GCGTTGGG	AATCTACCCC	TCACTGAGGG	GTAGTICCTT	AAATGTAGTG	GCGGGGGTTAT	AGCACACTCT	AAGCGTAGTA	GTTTA	ACTCGCTTTC	AGGGAGGCTG	TAGCTGCTTG	CCGT-AAAAC	CCCTTATAAC	TTATAGT	179
SUT058	1	CTATTCGAGC	GTCAT-ACAA	CCCTTAAGCC	T-TGTAGCTT	A-GCGTTGGG	AATCTACCCC	T-ACTGAGGG	-TAGTICCTT	AAATT-AGTG	GCGGGGGT-AT	AGCACACTCT	AAGCGTAGTA	GTTTA	ACTCGCTTTC	AGGGAGGCTG	TAGCTGCTTG	CCGT-AAAAC	CCCCTATAAC	TTATAGT	174
AJ390409	1	CTATTCGAGC	GTCATTACAA	CCCTTAAGCC	T-TGTAGCTT	A-GCGTTGGG	AATCTACCCC	TCACTGAGGG	GTAGTICCTT	ARATTTAGTG	GCGGGGGTTAT	AGCACACTCT	ARGCGTAGTA	GTTTA	ACTOGCTITC	AGGGAGGCTG	TRECTECTTE	CCGT-AAAAC	CCCCTATAAC	TTATAGT	179
SUT243	1	CTATICGAGC	GTCAT-ACAA	CCCTTAAGCC	T-TGTAGCTT	A-GCGTIGGG	AATCTACCCC	T-ACTGAGGG	-TAGTTCCTT	ARATT-AGTG	GCGGGGGT-AT	AGCACACTCT	AAGCGTAGTA	GTTTA	ACTCGCTTTC	AGGGAGGCTG	TAGCTGCTTG	CCGT-AAAAC	CCCCTATAAC	TTATAGT	174
SUT231	1	CTATICGAGC	GTCATAACAA	CCCTTAAGCC	C-TGTAGCTT	A-GCGTTGGG	AACCTACCGC	TTARGCG	GTAGCTCCTT	AAATTTAGTG	GCGGAGTTAC	AGCACCCCCT	AAGCGTAGTA	AAACTA	CCTCGCTTTC	AGGGAGCCTG	TAGCGGCCTG	CCGTTAAAAA	CCCCTATAA-	TTCTAGT	177
ST2584	1	CTATICGAGC	<b>GTCATTACAA</b>	CCCTTAAGCC	CCTGTCGCTT	A-GCGTTGGG	AGTCTGCGGC	TCAGGCC	G-AGTTCCTT	ARATT-AGTG	GCGGAGT-AC	AGCACARCCT	ARCCOTAGTA	GGTTA	CCTCGCTCCC	GGGGAGTCTG	TEGCEGECTE	CGTAAAAAAA	ACCCTARACC	TTCTA	173
SUT098	1	CTATTCGAGC	<b>GTCATTACGA</b>	CCCTTARGCC	TITCTCCTTT	A-GCGTTGGG	ARTCTGCGGT	TTAGGCC	GCAGTTCCTT	ARATTCAGTG	GCGGAGTTAT	ACCACACCCT	ARGCGTRGTA	ACTTA	CATCGCTCCT	GGGGAGTCTA	TACCCCCTC	CCGTTARARA	CCCCTATAT-	TTCTAGT	177
ST2436	1	CTATICGAGC	GTCATTTCGA	CCCTTAAGCC	CCAGTTGCTT	A-GCGTTGGG	AGTCTGCGGC	CCAGGCC	GCAGTTCCTC	AAAGTCAGTG	GCGGAGTTGT	AGCACACTCT	AAGCGTAGTA	GTTTCCATTG	CCTCGCATGC	AGAGCGGCCT	CAGCTGCCAG	CCGTAAA	GCCCTATACT	TCTTAGT	180
ST2473	1	CTATICGAGC	GTCATTICGA	CCCTTAAGCC	CCAGTTGCTT	A-GCGTTGGG	AGTCTGCGGC	CCAGGCC	GCAGTTCCTC	AAAGTCAGTG	GCGGAGTTGT	AGCACACTCT	AAGCGTAGTA	GTTTCCATTG	CCTCGCATGC	AGAGCGGCCT	CAGCTGCCAG	CCGTAAA	GCCCTATACT	TCTTAGT	180
ST2313	1	CTATICGAGC	GTCATTTCGA	CCCTTARGCC	CCRGTTGCTT	A-GCGTTGGG	AGTCTGCGGC	CCRGGCC	GCAGTTCCTC	ARAGTCAGTG	GCGGRGTTGT	RECACACTOT	ARGCGTAGTA	GTTTCCATTG	CCTCGC-TGC	AGAGCGGCCT	CAGCTGCCAG	CCGTAAA	GCCCTATACT	TCTTAGT	179
Jul	1	CTATTCGAGC	GTCATTTCGA	CCCTTAGGCC	CCAGTTGCTT	A-GCGTTGGG	AGTCTGCGGC	CCAGGCC	GCAGTTCCTC	AAAGTCAGTG	GCGGAGTTGT	AGCACACTCT	AAGCGTAGTA	GTITICCATTG	CCTCGC-TGC	AGAGCGGCCT	CAGCIGCCAG	CCGTAAA	GCCCTATACT	TCT-AGT	178
AJ390406	1	CTATTCGAGC	GTCATTTCGA	CCATTAAGCC	CCTGTAGCTT	A-GCGTTGGG	AGCCTACGTC	CTCGCGGC	GCAGCTCCTC	ARAGTCAGTG	GCGGAGTCGG	GTCGTGCTCT	GAGCGTAGTA	GTTAATA	TCTCGCTTCT	GCGGTGCCCC	CGGCTGCCTG	CCGT-AAACC	CCCCCCTATA	CTTTCGT	180
ST2336	1	CTGTTCGAGC	<b>GTCATTTCAA</b>	CCCTTAAGCC	CTCGTTGCTT	A-GTGTTGGG	AGCCTACGG -	CACC	GTAGCTCCCC	ARAGTCAGTG	GCGGAGTCGG	CTCACACTCT	AGACGTAGTA	ATTTCTCA	CCTCGCCTAT	AGT-TGGAC-	CEGTCCCCTE	CCGTAAAA	CGCCCAAGTC	TTTAAAA	173
AJ390395	1	CTATTCGAGC	GTCATTICAA	CCCTTAAGCC	CT-GTTGCTT	A-GCGTTGGG	AATCTGCTAG	CCTCCGC	GCAGTTCCTT	AAATTCATTG	GCGGRGCTGT	GGCACACTCT	AGGCGTAGTA	GTTTRACA	CCTCGCCTCT	AGAGTGGCCG	CEGTTACTES	CCGTAAAA	CCCCTATATT	TCTAGT-	177
SUT001	1	CTATTCGAGC	GTCATTTCAA	CCCTTAAGCC	CCTGTTGCTT	A-GCGTTGGG	CETCTECECC	GTGCCCTGGC	GCAGTGCCCT	AAATCTATCG	GCGGAGCCGT	AGCACACTCT	GAGCGTAGTA	ATT-ACAGTT	CCTCGCTCCT	GCAGTGGCCG	CEGECEGETEG	C-GTT-AA	CCCCTACAAC	TTCTAGT	181
SUT004	1	CTATICGAGC	GTCATTTCAA	CCCTTAAGCC	CCTGTTGCTT	A-GCGTTGGG	CGTCTGCGCC	GTGCCCTGGC	GCAGTGCCCT	AAATCTATCG	GCGGAGCCGT	AGCACACTCT	GAGCGTAGTA	ATT-ACAGTT	CCTCGCTCCT	GCAGTGGCCG	CGGCGGCTGG	CCGTT-AAAC	CCCCTACAAC	TTCTAGT	184
ST2485	1	CTATTCGAGC	GTCATTTCAA.	CCCTTAAGCC	CCTGTTGCTT	A-GCGTTGGG	CETCTECECC	GTGCCCTGGC	GCAGTGCCCT	ARATCTATCS	GCGGAGCCGT	AGCACACTOT	GAGCGTAGTA	ATT-ACAGTT	CCTCGCTCCT	CCACTCCCCC	CCCCCCCCTCC	CCGTT-AAAC	CCCCTACAAC	TTCTAGT	184
SUT262	1	CTATICGAGC	GTCATTICAA	CCCTTAAGCC	CCTGTTGCTT	A-GCGTTGGG	CGTCTGCGCC	GTGCCCTGGC	GCAGTGCCCT	ARATCTATCG	GCGGAGCCGT	AGCACACTCT	GAGCGTAGTA	ATT-ACAGTT	CCTCGCTCCT	GCAGTGGCCG	CGGCGGCTGG	CCGTT-AAAC	CCCCTATAAC	TTCTAGT	184
SUT005	1	CTATTCGAGC	GTCATTTCAA	CCCTTAAGCC	CCTGTTGCTT	A-GCGTTGGG	CGTCTGCGCC	GTGCCCTGGC	GCAGTGCCCT	ARATCCATCG	GCGGAGCCGT	AGCACACTOT	GAGCGTAGTA	ATT-ACAGTT	CCTCGCTCCT	GCAGTG-CCG	CGGCGGCTTG	GCGTAAC	CCCCTATA-C	TTCTAGT	180
ST2448	1	CTATTCGAGC	GTCATTTCAR	CCCTTARGCC	CCTGTTGCTT	A-GCGTTGGG	CGTCTGCGCC	GTGCCCTGGC	GCAGTGCCCT	ARATC-ATCG	GCGGAGCCG-	AGCACACTOT	GAGCGTAGTA	ATACAGT-	CCTCGCTCCT	GCAGTGGCCG	CGGCGGCT-G	CCGTAAC	CCC-TATA-C	TICTAGT	175
SUT167	1	CTATICGAGO	GTCATTICAA	COUTTARGO	CONGINGOIT	A-GCGTTGGG	CGTCTGCGCC	GTGCCCCGGC	GCAGTGCCCT	ABATCCATCG	GOGGAGCOGT	AGCACACTOT	GAGCGTAGTA	ATT-ACAGTT	COTOGOTOOT	GCAGTGGCCG	DESCESCT-S	COGTE-AAA-	CCCCTATAAC	TTCTAGT	182
SUT220	1	CTATCCGAGC	GTCATTACGA	CCCTTAAGCC	CCCGG-GCTT	A-GCGTTGGG	AATCCGCGTC	TT-CAGGGGC	GCGGTTCCCT	ABATTCATCG	GCGGCGCCGG	GGCGTCTTCT	GAGCGTAGTA	ATTTATTA	TCTCGC-CCT	GAAGCTAGCC	CCGTCGCCCG	CCGT-AAAAC	CCCCCAACTA	CCTTGT-	179
572332	1	CTATCCG2GC	GTCATTACCA.	COULTS SCOOL	CCCGG-GCTT	A-GCGTTCGG	aamoogogmo	TT-CAGGGGG	GCGGTTCCCT	ABATTC ATTC	GCGGCGCCCGG	GGCGTCTTCT	GAGCGTACTA	Adabah-Pdates	TOTOGO-COT	GARGCTACCT	COSTOGCOOG	CCGT-3323C	CCCCCAACTA	COGTACT	180
ST2332	1	CTATCCGAGC	GTCATTACGA	CCCTTAAGCC	CCCGG-GCTT	A-GCGTTGGG	AATCCGCGTC	TT-CAGGGGC	GCGGTTCCCT	ARATTCATCG	GCGGCGCCGG	GGCGTCTTCT	GAGCGTAGTA	ATTTATTA	TETEGE-CET	GARGCTAGCT	CCGTCGCCCG	CCGT-AAAAC	CCCCCAACTA	CCGTACT	180

Figure 8C. The ITS2 sequence alignment of Hypoxylon sect. Annulata for phylogenetic tree construction in Figure 95 by using

ClustalX and BioEdit programs. Arrows indicate the start and the end of 5.8S rDNA sequences.

		10	20	) 30	40	50	60	70	80	90	10	) 110	120	) 13	0 140	15	0 160	
		· · · · · [ · · · · ]	$\cdots \cdots 1 \cdots \cdots 1$								· · · · I · · · · I	· · · · I · · · · I	$\cdots \cdots 1 \cdots \cdots 1$			$\cdots \cdots 1 \cdots \cdots 1$		
SUT223	1	CTGAGTTGAC	CATAAAAACT	CCCAA-CCCT	ATGTGAACCT	TTTGTTCTCG	TTGCCTTGGC	ATGCGCCAGG	GGGGCGGCGT	ACCA		C	GTACCCCGGG	ACCTAT			AAAC	104
SUT218	1	<b>CTGAGTTGAC</b>	TATAAAAACT	CCCAA-CCCT	ATGTGAACCT	TTTGTTCTCG	TTGCCTCGGC	ATGCGCCAGG	GGGGCGGCGT	ACCA		C	GTACCCCGCG	ACCTAT			AAAC	104
SUT233	1	CTGAGTTATC	TAAACT	CCAAA-CCCT	TTGTGAAC-T	TACCGTCG	TTTCCTCGGC	GTGTGCTG	CGGCTACCCC	GTAG			CTACC				TACCCGG	86
SUT240	1	CTGAGTTATC	TAAACT	CCAAA-CCCT	TTGTGAAC-T	TACCGTCG	TTTCCTCGGC	GTGTGCTG	CGGCTACCCC	GTAG			CTACC				TACCCGG	86
SUT066	1	CTGAGT	-ATAAAAACT	CTAAAACCCT	TTGTGAACCT	TACCGCCG	TTGCCTCGGC	GTGCGCTG	CGGCTACCCT	GTAG			CTACCCTGTA	GGTGCC	TACCCTGTAG	GTGCC	TACTCGG	113
SUT068	1	CTGAGT	-ATAAAAACT	CTAAAACCCT	TTGTGAACCT	TACCGCCG	TTGCCTCGGC	GTGCGCTG	CGGCTACCCT	GTAG			CTACCCTGTA	GGTGCC	TACCCTGTAG	GTGCC	TACTCGG	113
SUT069	1	CTGAGT	-ATAAAAACT	CTAAAACCCT	TTGTGAACCT	TACCGCCG	TTGCCTCGGC	GTGCGCTG	CGGCTACCCT	GTAG			CTACCCTGTA	GGTGCC	TACCCTGTAG	GTGCC	TACTCGG	113
SUT046	1	CTGAGT	-ATAAAAACT	CTAAAACCCT	TTGTGAACCT	TACCGCCG	TTGCCTCGGC	GTGCGCTG	CGGCTACCCT	GTAG			CTACCCTGTA	GGTGTC	TACCCTGTAG	TTGCC	TACCCGG	113
SUT041	1	ATGAGTT	-ATTAAAACT	CCAAA-CCCT	ATGTGAACCT	TACCGTCG	TTGCCTCGGC	GTGAGCTA	CGGCTACCCT	GTAGTTACCC	TGGAGCT	ACCCTATAG-	CTACCCTGTA	GTTACCCTAT	TACCCTGCAG	CTACC-CTAT	AGGTACCCTG	146
SUT063	1	AAGAGTT	-ATTAAAACT	CCAAA-CCCT	TTGTGAACCT	TACCGTCG	GTGCCTCGGA	GTGAGCTA	CGGCTACCCT	GTAGCTACCC	TGTAGC-	ACCCTATAG-	CTACCCTGTA	GT	-ACCCTACAG	CTACC-CTAT	AGGTACCTTG	136
SUT020	1	CTGAGTTCT-	-ACAAAAACT	CCCAA-CCCT	<b>TTGTGAATCA</b>	TACCTATG	TTGCCTCGGC	GCC-GAGCAG	CAGCTACCCG	GGAGTTACCC	TGTAGGCTAC	CCTGTAGATA	TCTACCCTGT	AGGGAGCTAC	TACCCTGTAG	ATATTACGAT	GTCTACCCTG	154
SUT294	1	CTGAGTTCT-	-AC-AAAACT	CCCAA-CCCT	ATGTGAATCT	TACCACTG	TTGCCTCGGC	GCT-GAGCGG	CAGCTACCCG	GGAG			CTACCCTGGA	GGGACCTACC	TACCCTG			104
SUT256	1	GCGAGTTCTC	GAT-AAAACT	CCGTAACCCT	GTGCGAACCC	TACCACTG	TTGCCTCGGC	GTCGCC	CGCC				CTTGCG	GGAC				77
SUT154	1	CTGAGTTCT-	AAACT	CCAACCCT	ATGTGAACT-	TACCACTG	TTGCCTCGGC	GCTGTGCC	TGCG				AGAGCA	GGCCC				72
SUT070	1	CTGAGTTCT-	ACAACT	CCCACCCT	CTGTGAATA-	TACTACAG	TTGCCTCGGC	GA	-GGC				GCTACA	G				62
SUT237	1	CTGAGTTCT-	ATCAACT	CCCACCCT	ATGTGAACA-	TACTACAG	TTGCCTCGGC	GG	-GGT				GCTACA					62
SUT108	1	CCGAGTT-T-	ACACCT	CCCAAACCCC	-TGTGAACA-	TACCTATACG	TTGCCTCGGC	GGATCAGC	CCGC				GCCCCG	TAAA				74
SUT082	1	CTGAGTTAT-	CACAACT	CCAACCCT	GTGTGAAC-T	TACTACTG	TTGCCTCGGC	GGC					GCTGTG	CGA				63
SUT080	1	<b>CTGAGTTATC</b>	CAAAACT	CCCAA-CCCT	TTGTGAAC-T	TACCACTG	TTGCCTCGGC	GAGTTGTG	CTAC				CCTATA	GCTACC				77
SUT116	1	<b>CTGAGTTATC</b>	CAAAACT	CCCAA-CCCT	TTGTGAAC-T	TACCACTG	TTGCCTCGGC	GAGTTGTG	CTAC				CCTATA	GCTACC				77
SUT042	1	<b>CTGAGTTATC</b>	CAAAACT	CCCAA-CCCT	TTGTGAAC-T	TACCACTG	TTGCCTCGGC	GAGTTGTG	CTAC				CCTATA	GCTACC				77
SUT164	1	CAGAGTTATC	CAACT	CCCAA-CCCT	TTGTGAACCT	TACCGCAG	TTGCCTCGGC	GCGAGCCG	CGGCT				ACCCTGCA	GCTACC				79
SUT293	1	CAGAGTTATC	CAACT	CCCAA-CCCT	TTGTGAACCT	TACCGCAG	TTGCCTCGGC	GCGAGCCG	CGGCT				ACCCTGCA	GCTACC				79
SUT292	1	CAGAGTTATC	CAACT	CCCAA-CCCT	TTGTGAACCT	TACCGCAG	TTGCCTCGGC	GCGAGCCG	CGGCT				ACCCTGCA	GCTACC				79
SUT159	1	CAGAGTTATT	CTAAACT	CCAAA-CCCT	ATGTGAAC-T	TACCACTG	TTGCCTTGGC	GTG-TGCC	GCGAG				CTACCCTGTA	GTGA			CCTAC	84
SUT162	1	CAGAGTTATT	CTAAACT	CCAAA-CCCT	ATGTGAAC-T	TACCACTG	TTGCCTTGGC	GTG-TGCC	GCGAG				CTACCCTGTA	GTGA			CCTAC	84
SUT061	1	CAGAGTTATT	CTAAACT	CCAAA-CCCT	ATGTGAAC-T	TACCACTG	TTGCCTTGGC	GTG-TGCC	GCGAG				CTACACTGTA	GTGA			CCTAC	84
SUT280	1	CAGAGTTATT	CTAAACT	CCAAA-CCCT	ATGTGAAC-T	TACCACTG	GTGCCTTGGC	GTG-TGCC	G									61
SUT165	1	CAGAGTTATT	CTAAACT	CCAAAACCCT	ATGTGAAC-T	TACCACTG	TTGCCTTGGC	GTG-TGCC	GCGAG				CTACACTGTA	GTGA			CCTAC	85
SUT120	1	CAGAGTTATA	CTAAACT	CCAAA-CCCT	ATGTGAACTT	TACCACTG	TTGCCTTGGC	GTG-TGCC	GCTTG				CTACCCTGTA	GCTC			CCTAC	85
SUT016	1	CAGAGTTACC	AAAACT	CCCAA-CCCT	TTGTGTACC-	TACTACCA	TTGCTTCGGC	GGGCTGCG	GCTACCCTGC	AG			CTACCCTGTA	ATTC			AGG	89
SUT180	1	CAGAGTTACC	AAAACT	CCCAA-CCCT	TTGTGTACC-	TACTACCA	TTGCTTCGGC	GGGCTGCG	GCTACCCTGC	AG			CTACCCTGTA	ATTC			AGG	89
ST2324	1	CAGAGTTACC	AAAACT	CCCAA-CCCT	TTGTGTACC-	TACTACCA	TTGCTTCGGC	GGGCTGCG	GCTACCCTGC	AG			CTACCCTGTA	ATTC			AGG	89
SUT250	1	CTGAGTTACT	CAAAACT	CCCAA-CCCT	ATGTGAAC-T	TACCATCG	TTTCTTCGGC	GGGCTGCG	GCTACCCTGT	AC			CTACCCTGAA	GCAA			AAGA	91
SUT282	1	CAGAGTTACT	AAACT	CCCAA-CCCT	ATGTGAAC-T	TACCGTCG	TTGCTTCGGC	GGGCTGCG	GCTACCCTGT	AC			CTACCCTGTA	CCTACCCTGT			AGGGCCC	98
SUT166	1	CAGAGTTACT	ATAAAACT	CCCAA-CCCT	TTGTGAACCT	TACCGTCG	TTGCCTCGGT	GGAAGGTGGT	GTGCGGTGGG	AAG-CTACCC	TGGAG	C	CTACCCTGTA	GATAGC			TAC	109
SUT158	1	CAGAGTTAAT	CCAAACT	CCCAA-CCCT	TTGTGAACCA	TATCGTCG	TTGCCTCGGG	CGT	GAGCGGCGG-				CCACCCAGAG	AGTAGC			TAC	85
SUT148	1	CAGAGTTATT	AAAAACT	CCCAAACCCT	TTGTGAACGT	TACTGTTG	TTGCCTCGGC	GT	GAGCGAGGG-				CTACCC	GGGAGC			TAC	81
SUT187	1	CAGAGTTATT	AAAAACT	CCCAAACCCT	TTGTGAACGT	TACTGTTG	TTGCCTCGGC	GT	GAGCGAGGG-				CTACCC	GGGAGC			TAC	81
SUT182	1	<b>CTGAGTTCTA</b>	CAAAAACT	CCCAA-CCCT	TTGTGAACCC	TACTTCAG	TTGCCTCGGC	GCT-GAGCGG	TAGCTACCCG	GGAA		A	GCTACCCTGG	AGAAATC			C	97
SUT215	1	<b>CTGAGTTATC</b>	CAAACT	CCAAAACCCT	TTGTGAACCT	TACCGTCG	TTGCCTCGGC	GTGAGCTG	CGGCTACCCG	GTAG			CTACCCTGTA	G				87
SUT221	1	<b>CTGAGTTATC</b>	CAAACT	CCAAAACCCT	TTGTGAACCT	TACCGTCG	TTGCCTCGGC	GTGAGCTG	CGGCTACCCG	GTAG			CTACCCTGTA	G				87
AJ390400	1	cagagttaat	ccaaaact	ccaaaaccct	atgtgaac-t	taccgctg	ttgcctcggc	gtg-tgcc	gcgag	ctaccc	tgtagctacc	<b>cagtaggtga</b>	ctaccctgta	gtca			cctac	112

**Figure 9C.** The ITS1-5.8S-ITS2 multiple sequence alignment of *Hypoxylon* sect. *Hypoxylon* for phylogenetic tree construction in Figure 94 by using ClustalX and BioEdit programs. Arrows indicate the start and the end of 5.8S rDNA sequences.

		170	) 18	0 190	200	0 21	0 220	230	240	25	0 26	27	280	290	300	) 31(	320	)
														· · · · I · · · · I	· · · · I · · · · I		· · · · I · · · · I	
SUT223	105	GGTAGGGGTC	TTGGGCCCCC	CGCCCGCATC	CGGCCTGCCG	GTGGACCAAC	CCAACTCTTG	CAAATCTT	GTGAAATTCT	GAAATATA	AAAATAAACG	AATCAAZACT	TTCAACAACG	GATCTCTTGG	TTCTGGCATC	GATGAAGAAC	GCAGCGAAAT	260
SUT218	105	GGCAGGGGTC	TTGGGCGCCC	CGCCCGCACC	CGGCCTGCCG	GTGGACCAAC	CCAACTCTTG	CAAATATT	GTGGAACTCT	GAAATATA	AAAATAAACG	AATCAAAACT	TTCAACAACG	GATCTCTTGG	TTCTGGCATC	GATGAAGAAC	GCAGCGAAAT	260
SUT233	87	TAGCTA	CCCTGTAGCT	G-GCCCA-CG	GCCCGCCG	CAGGACCGCT	AAACTCTTGT	TTTTACCA	CTGTATCTCT	GAATTGTT	AACTGAAATA	AGTTAAAACT	TTCAACAACA	GATCTCTTGG	TTCTGGCATC	GATGAAGAAC	GCAGCGAAAT	234
SUT240	87	TAGCTA	CCCTGTAGCT	G-GCCCA-CG	GCCCGCCG	CAGGACCGCT	AAACTCTTGT	TTTTACCA	CTGTATCTCT	GAATTGTT	AACTGAAATA	AGTTAAAACT	TTCAACAACG	GATCTCTTGG	TTCTGGCATC	GATGAAGAAC	GCAGCGAAAT	234
SUT066	114	CAGCTG	CCCTGTAGCC	G-GACCA-CG	GCCCGCCG	GAGGACTGCT	AAACTCTTGT	TTTTTTACCA	CTGTATCTCT	GAATTCTT	AACTGAAATA	CGTTAAAACT	TTCAACAACG	GATCTCTTGG	TTCTGGCATC	GATGAAGAAC	GCAGCGAAAT	263
SUT068	114	CAGCTG	CCCTGTAGCC	G-GACCA-CG	GCCCGCCG	GAGGACTGCT	AAACTCTTGT	TTTTTTACCA	CTGTATCTCT	GAATTCTT	AACTGAAATA	CGTTAAAACT	TTCAACAACG	GATCTCTTGG	TTCTGGCATC	GATGAAGAAC	GCAGCGAAAT	263
SUT069	114	CAGCTG	CCCTGTAGCC	G-GACCA-CG	GCCCGCCG	GAGGACTGCT	AAACTCTTGT	TTTTTTACCA	CTGTATCTCT	GAATTCTT	AACTGAAATA	CGTTAAAACT	TTCAACAACG	GATCTCTTGG	TTCTGGCATC	GATGAAGAAC	GCAGCGAAAT	263
SUT046	114	TAGCTA	CCCTGTAGCC	G-GACCA-CG	GCCCGCCG	GAGGACTGCT	AAACTCTTGT	TTTTT-ACCA	CTGTATCTCT	GAATTCTT	AACTGAAATA	CGTTAAAACT	TTCAACAACG	GATCTCTTGG	TTCTGGCATC	GATGAAGAAC	GCAGCGAAAT	262
SUT041	146	GAGCTA	CCCTGTAGAC	G-GCTTA-TG	GCCCGCCG	AAGGACCGCT	AAACTCTTGT	TTTTATTG	CTGTTATTCT	GAATTATA	ААСТААААТА	AGTTAAAACT	TTCAACAACG	GATCTCTTGG	TTCTGGCATC	GATGAAGAAC	GCAGCGAAAT	294
SUT063	136	AAGCTA	CCCTGTAGAC	G-GCTTA-TG	GCCCGCCG	AAGGACCGCT	AAACTCTTGT	TTTTATTG	CTGGTATTCT	GAATTATA	AACTAAAATA	AGTTAAAACT	TTCAACAACG	GATCTCTTGG	TTCTGGCATC	GATGAAGAAC	GCAGCGAAAT	284
SUT020	155	TAAGGAGCTA	CCCTGGAGTT	GCACTCA-CG	CTCCGCCG	ATGGACCAGT	AAACTCT-GT	TTTTT-ATAG	-TGTATCTCT	GAATTCTT	TAACAAAATT	CGTTAAAACT	TTCAACAACG	GATCTCTTGG	TTCTGGCATC	GATGAAGAAC	GCAGCGAAAT	30€
SUT294	104	GAGCTA	CCCTGGAGTT	GCGCCTAACG	CTCCGCTG	GCGGACCACC	AAACTCT-GT	TTTAC-A-AG	-TGTATCTCT	GAGTATAT	ААССААААТА	CGTTAAAACT	TTCAACAACG	GATCTCTTGG	TTCTGGCATC	GATTAAGAAC	GCAGCCAAAT	252
SUT256	77	G	CCAGTGGGCC	C-ATG-AACT	GGACTC	-TGTTTTA	GCTGCTCACT	GCAGCCACTG	-TGAATATCT	GAACGGCCTT	AACTGAAATA	CGTTAAAACT	TTCAACAACG	GATCTCTTGG	TTCTGGCATC	GATGAAGAAC	GCAGCGAAAT	218
SUT154	72	G	CCGGTGGACC	ACT	AAACTC	-TGTTATA	CCTACT		GTATCTCT	GAATTTAT	AACTGAAATA	CGTTAAAACT	TTCAACAACG	GATCTCTTGG	TTCTGGCATC	GATGAAGAAC	GCAGCGAAAT	191
SUT070	62		CCT	C-GCC-GGCG	GACCAC	T	AAACTC-TGT	TTTGTCCA	CTGTATCTCT	GAATTGAGTA	ACAAAATA	CGTTAAAACT	TTCAACAACG	GATCTCTTGG	TTCTGGCATC	GATGAAGAAC	GCAGCGAAAT	185
SUT237	62		ccc	C-GCC-GGCG	GACCAC	T	AAACTC-TGT	TTTATCCA	CTGTATCTCT	GAATTTAATA	ACGAAATA	CGTTAAAACT	TTCAACAACG	GATCTCTTGG	TTCTGGCATC	GATGAAGAAC	GCAGCGAAAT	185
SUT108	74	A	CGGGACGGCC	C-GCC-CGAG	GACCCC	T	AAACTC-TGT	TTTTAG	TGGAACTTCT	GAGTAAAACA	AACAAATA	AATCAAAACT	TTCAACAACG	GATCTCTTGG	TTCTGGCATC	GATGAAGAAC	GCAGCAAAAT	203
SUT082	63		GCAGCC	C-GCC-GGCG	GACCAC	TAAAC	TCTGT	TTTTA	CAGCATCTCT	GAATGATA	ACTTAAAT	AGTTAAAACT	TTCAACAACG	GATCTCTTGG	TTCTGGCATC	GATGAAGAAC	GCAGCGAAAT	184
SUT080	77	-CTGTAGCTA	CCCGGGAACA	C-ATT-CCAA	GCTCGC	-CAGAGGACC	TACCAACTCT	GTTTTATACT	GTATCTCT	GAACTTTATG	ACTAAATA	AGTTAAAACT	TTCAACAACG	GATCTCTTGG	TTCTGGCATC	GATGAAGAAC	GCAGCGAAAT	225
SUT116	77	-CTGTAGCTA	CCCGGGAACA	C-ATT-CCAA	GCTCGC	-CAGAGGACC	TACCAACTCT	GTTTTATACT	GTATCTCT	GAACTTTATA	ACTT-AAATA	AGTTAAAACT	TTCAACAACG	GATCTCTTGG	TTCTGGCATC	GATGAAGAAC	GCAGCGAAAT	22€
SUT042	77	-CTGTAGCTA	CCCGGGTACA	C-ATT-CCAA	GCTCGC	-CAGAGGACC	TACCAACTCT	GTTTTATACT	GTATCTCT	AACTTTATAA	CTAAATA	AGTTAAAACT	TTCAACAACG	GATCTCTTGG	TTCTGGCATC	GATGAAGAAC	GCAGCGAAAT	224
SUT164	79	-CTGTAGCTG	CCGCGTAGCA	C-GCA-CATG	GCCCGCCG	GTGGACAG-C	TAAACTCTT-	GTATGTACAC	AAGTATGTCT	GATTGCTT	AAATAAAATA	AGTCAAAACT	TTCAACAACG	GATCTCTTGG	TTCTGGCATC	GATGAAGAAC	GCAGCGAAAT	230
SUT293	79	-CTGTAGCTG	CCGCGTAGCA	C-GCA-CATG	GCCCGCCG	GTGGACAG-C	TAAACTCTT-	GTATGTACAC	AAGTATGTCT	GATTGCTT	AAATAAAATA	AGTCAAAACT	TTCAACAACG	GATCTCTTGG	TTCTGGCATC	GATGAAGAAC	GCAGCGAAAT	230
SUT292	79	-CTGTAGCTG	CCGCGTAGCA	C-GCA-CATG	GCCCGCCG	GTGGACAG-C	TAAACTCTT-	GTATGTACAC	AAGTATGTCT	GATTGCTT	AAATAAAATA	AGTCAAAACT	TTCAACAACG	GATCTCTTGG	TTCTGGCATC	GATGAAGAAC	GCAGCGAAAT	230
SUT159	85	CCGGGAGCTA	CCCTGTAGTG	C-GCA-TACG	GCCCGCCG	AAGGACTAAC	TAAACTCTTT	GTCTT-ACTG	TG-AATATCT	GAATGCTT	CAACTTAATA	AGTTAAAACT	TTCAACAACG	GATCTCTTGG	TTCTGGCATC	GATGAAGAAC	GCAGCGAAAT	23€
SUT162	85	CCGGGAGCTA	CCCTGTAGTG	C-GCA-TACG	GCCCGCCG	AAGGACTAAC	TAAACTCTTT	GTCTT-ACTG	TG-AATATCT	GAATGCTT	CAACTTAATA	AGTTAAAACT	TTCAACAACG	GATCTCTTGG	TTCTGGCATC	GATGAAGAAC	GCAGCGAAAT	236
SUT061	85	CCGGGAGCTA	CCCTGTAGTG	C-GCA-TATG	GCCCGCCG	AAGGACTAAC	TAAACTCTTT	GTCTT-ACTG	TG-AATATCT	GAATGCTT	CAACTTAATA	AGTTAAA-CT	TTCAACAACG	GATCTCTTGG	TTCTGGCATC	GATGAAGAAC	GCAGCGAAAT	235
SUT280	61	GGAGCTA	CCCTGTAGTG	C-GCA-TATG	GCC-GCCG	AAGGACTAAC	TAAACTCTTT	GTCTT-ACTG	TG-AATATCT	GAATGCTT	CAACTTAATA	AGTTAAAACT	TTCAACAACG	GATCTCTTGG	TTCTGGCATC	GATGAAGAAC	GCAGCGAAAT	209
SUT165	86	CCGGGAGCTA	CCCTGTAGTG	C-GCA-TACG	GCCCGCCG	AAGGACTA-C	TAAACTCTTT	GTCTT-ACTG	TG-AATATCT	GAATGCTT	CAACTTAATA	AGTTAAAACT	TTCAACAACG	GATCTCTTGG	TTCTGGCATC	GATGAAGAAC	GCAGCGAAAT	236
SUT120	86	CCTGGAGCTA	CCCTGTAGCA	C-GCA-CACG	GCCCGCCA	AAGGACCA-C	TAAACTCTTT	ATTTTTACTG	TG-AATATCT	GAATGCTT	CAACTTAATA	AGTTAAAACT	TTCAACAACG	GATCTCTTGG	TTCTGGCATC	GATGAAGAAC	GCAGCGAAAT	237
SUT016	89	-GTGGGCTTG	CCTGGTAGCT	C-GCGCGAAG	GCCCGTCA	GAGGACCA-T	TAAACTCTTG	TTACCCTGTA	CGTCATATCT	GAATGCTT	CAACTTAATA	AGTTAAAACT	TTCAACAACG	GATCTCTTGG	TTCTGGCATC	GATGAAGAAC	GCAGCGAAAT	242
SUT180	89	-GTGGGCTTG	CCTGGTAGCT	C-GCGCGAAG	GCCCGTNA	GAGGACCA-T	TAAACTCTTG	TTACCCTGTA	CGTTATATCT	GAATGCTT	CAACTTAATA	AGTTAAAACT	TTCAACAACG	GATCTCTTGG	TTCTGGCATC	GATGAAGAAC	GCAGCGAAAT	242
ST2324	89	-GTGGGCTTG	CCTGGTAGCT	C-GCGCGAAG	GCCCGTCA	GAGGACCA-T	TAAACTCTTG	TTACCCTGTA	CTTAATATCT	GAATGCTT	CAACTTAATA	AGTTAAAACT	TTCAACAACG	GATCTCTTGG	TTCTGGCATC	GATGAAGAAC	GCAGCGAAAT	242
SUT250	92	GGGCGGGCTG	CACGGTAGCT	T-GCCATAAG	GCCCGTCA	GAGGACCA-T	TAAACTCGTG	TTACCCTGTA	CGTAA-ATCT	GAATACTT	CAACTTAATA	AGTTAAAACT	TTCAACAACG	GATCTCTTGG	TTCTGGCATC	GATGAAGAAC	GCAGCGAAAT	244
SUT282	99	GGGCGGGTTT	CCTGGTAGCT	T-GCGCTAAG	GCCCGTCA	GAGGACCA-T	CAAACTCATG	TTACCCTGTA	CGTACTATCT	GAATACTT	CAACTTAATA	AGTTAAAACT	TTCAACAACG	GATCTCTTGG	TTCTGGCATC	GATGAAGAAC	GCAGCGAAAT	252
SUT166	110	CCTGGAGCTA	CCCTGAAAAT	ACGCCCCCCG	CCAGCCGCCG	AAGGACTACT	AAACTCTTGT	TTT-ACTG	-TGTCTCTCT	GAATA-ATGA	AACAAAAATT	CGTTAAAACT	TTCAACAACG	GATCTCTTGG	TTCTGGCATC	GATGAAGAAC	GCAGCGAAAT	264
SUT158	86	CCTGGAGCTG	CAAACT	ACGCCC	GCCG	GAGGACCACT	AAACTCTTGT	TTTTACCA	-TGTATTTCT	GAATG-CTTC	AACTATAAAT	AGTTAAAACT	TTCAACAACG	GATCTCTTGG	TTCTGGCATC	GATGAAGAAC	GCAGCGAAAT	225
SUT148	82	CCTGTAGCTA	CCCT-GTAAC	CCGTTGTAAG	CCCGCCG	GAGGACCACT	AAACTCTGGT	TTATTACTG	-TGTATCTCT	GAATG-CTTC	AACTGAAATC	AGTTAAAACT	TTCAACAACG	GATCTCTTGG	TTCTGGCATC	GATGAAGAAC	GCAGCGAAAT	235
SUT187	82	CCTGTAGCTA	CCCT-GTAAC	CCGTTGTAAG	CCCGCCG	GAGGACCACT	AAACTCTGGT	TTATTTACTG	-TOTATCTCT	GAATG-CTTC	AACTGAAATC	AGTTAAAACT	TTCAACAACG	GATCTCTTGG	TTCTGGCATC	GATGAAGAAC	GCAGCGAAAT	235
SUT182	98	GGGGCGACCT	ACCCTGTAGT	TACACCTAAC	GCT-CCGCCG	GTGGACCACT	AAACTCTCTT	TTTAACCA	CTGTATCTCT	GAAATACTTA	ACGAAATA	CGTTAAAACT	TTCAACAACG	GATCTCTTGG	TTCTGGCATC	GATGAAGAAC	GCAGCGAAAT	252
SUT215	87	СТА	CCCTGTAGCC	GGTTCACG	GCCCGCCG	AAGGACAGCT	AAACTCTTGT	TAATT-ACCA	CTGTATCTCT	GAATTGTC	AACT-AAATA	AGTTAAAACT	TTCAACAACG	GATCTCTTGG	TTCTGGCATC	GATGAAGAAC	GCAGCGAAAT	232
SUT221	87	CTA	CCCTGTAGCC	GGTTCACG	GCCCGCCG	AAGGACAGCT	AAACTCTTGT	TAATT-ACCA	CTGTATCTCT	GAATTGTC	AACT-AAATA	AGTTAAAACT	TTCAACAACG	GATCTCTTGG	TTCTGGCATC	GATGAAGAAC	GCAGCGAAAT	230

Figure 9C. (Continued).

		33	0 340	) 35(	0 360	370	380	) 390	400	) 410	0 420	43	0 440	) 45(	46	) 470	) 480	)
												· <b>T</b> ·····						
SUT223	261	GCGATAAGTA	ATGTGAATTG	CAGAATTCAG	TGAATCATCG	AATCTTTGAA	CGCATATTGC	GCCCATTAGT	ATTCTAGTGG	GCATGCCTAT	TCGAGCGTCA	TTTCGACCCT	TACGCTCTAG	TAGCGTAGC-	GTTGGGACTC	TAGCCCTGCC	ATAGGCTAGT	419
SUT218	261	GCGATAAGTA	ATGTGAATTG	CAGAATTCAG	TGAATCATCG	AATCTTTGAA	CGCATATTGC	GCCCATTAGT	ATTCTAGTGG	GCATGCCTAT	TCGAGCGTCA	TTTCGAACCA	TACGCTTTAG	TAGCGTAAC-	GTTGGGACTC	TAGCCCTGCC	ATAGGCTAGT	419
SUT233	235	GCGATAAGTA	ATGTGAATTG	CAGAATTCAG	TGAATCATCG	AATCTTTGAA	CGCACATTGC	GCCCATTAGT	ATTCTAGTGG	GCATGCCTAT	TCGAGCGTCA	TTTCAACCCT	TAAGCCT-TG	TTGCTTA-GC	GTTGGGAGTC	TACGGCTT	CGGCGTA-GC	385
SUT240	235	GCGATAAGTA	ATGTGAATTG	CAGAATTCAG	TGAATCATCG	AATCTTTGAA	CGCACATTGC	GCCCATTAGT	ATTCTAGTGG	GCATGCCTAT	TCGAGCGTCA	TTTCAACCCT	TAAGCCT-TG	TTGCTTA-GC	GTTGGGAGTC	TACGGCTT	CGGCGTA-GC	385
SUT066	264	GCGATAAGTA	ATGTGAATTG	CAGAATTCAG	TGAATCATCG	AATCTTTGAA	CCCACATTCC	GCCCACTGGT	ATTCCAGTCG	GCATGCCTAT	TCGAGCGTCA	TTTCAACCCT	TAAGCCCCTG	TTGCTTA-GC	GTTGGGAGTC	TACGTCTT-A	CGGCGTA-GT	420
SUT068	264	GCGATAAGTA	ATGTGAATTG	CAGAATTCAG	TGAATCATCG	AATCTTTGAA	CGCACATTGC	GCCCACTGGT	ATTCCAGTGG	GCATGCCTAT	TCGAGCGTCA	TTTCAACCCT	TAAGCCCCTG	TTGCTTA-GC	GTTGGGAGTC	TACGTCTT-A	CGGCGTA-GT	420
SUT069	264	GCGATAAGTA	ATGTGAATTG	CAGAATTCAG	TGAATCATCG	AATCTTTGAA	CGCACATTGC	GCCCACTGGT	ATTCCAGTGG	GCATGCCTAT	TCGAGCGTCA	TTTCAACCCT	TAAGCCCCTG	TTGCTTA-GC	GTTGGGAGTC	TACGTCTT-A	CGGCGTA-GT	420
SUT046	263	GCGATAAGTA	ATGTGAATTG	CAGAATTCAG	TGAATCATCG	AATCTTTGAA	CGCACATTGC	GCCCACTGGT	ATTCCAGTGG	GCATGCCTAT	TCGAGCGTCA	TTTCAACCCT	TAAGCCCCTG	TTGCTTAAGC	GTTGGGAGTC	TACGTCTT-A	CGGCGTAAGT	421
SUT041	295	GCGATAAGTA	ATGTGAATTG	CAGAATTCAG	TGAATCATCG	AATCTTTGAA	CCCACATTCC	GCCCATTAGT	ATTCTAGTCG	GCATGCCTAT	TCGAGCGTCA	TTTCGACCCC	TAAGCCCTTG	TTGCTTA-GT	GTTGGGAATC	TAC	-GGCGTA-GT	444
SUT063	285	GCGATAAGCA	ATGTGAATTG	CAGAATTCAG	TGAATCATCG	AATCTTTGAA	CGCACATTGC	GCCCATTAGT	ATTCTAGTGG	GCATGCCTAT	TCGAGCGTGA	TTTCGACCCC	TAAGCCCCTG	NTGCTTA-GC	GTTGGGAATC	TAC	-GGCGTA-GT	434
SUT020	307	GCGATAAGTA	ATGTGAATTG	CAGAATTCAG	TGAATCATCG	AATCTTTGAA	CGCATATTGC	GCCCAGTAGT	ATTCTACTGG	GCATGCCTAT	TCGAGCGTCA	TTTCAACCCT	TATGCCC-TG	TAGCATA-GT	GTTGGGGGCTC	TACC-G	AAAGGTA-GT	458
SUT294	253	GCGATACGTA	ATGTGAATTG	CAGAATTCAG	TGAATCATCG	AATCTTTGAA	CGCATATTGC	GCCCATTAGT	ATTCTAGTGG	GCATGCCTAT	TCGAGCGTCA	TTTCAACCCT	TAAGCCC-TG	TAGCTTA-GC	GTTGGGACTC	TACTCCTC-C	GGGTGTA-GT	408
SUT256	219	GCGATAAGTA	ATGTGAATTG	CAGAATTCAG	TGAACCATCG	AATCTTTGAA	CGCACATTGC	GCCCACTAGC	ATTCTAGTGG	GCATGCCTAT	CCGAGCGTCA	TTTCAACCCT	AAGGCCCTTG	CGGCTAACC-	GTTGGAAGCC	TGTGGCTG	CAGCGCAGCT	375
SUT154	192	GCGATAAGTA	ATGTGAATTG	CAGAATTCAG	TGAATCATCG	AATCTTTGAA	CGCACATTGC	GCCCATTAGT	ATTCTAGTGG	GCATGCCTAT	TCGAGCGTCA	TTTCAACCCT	TAAGCCCCTG	TTGCTTAGT-	GTTGGGAATC	TGCGTTA	CGGCGCAG-T	34€
SUT070	186	GCGATAAGTA	ATGTGAATTG	CAGAATTCAG	TGAATCATCG	AATCTTTGAA	CGCACATTGC	GCCCATTAGT	ATTCTAGTGG	GCATGCCTAT	TCGAGCGTCA	TTTCAACCCT	TACGCCCT-G	TAGCGTAGT-	GTTGGGAATC	TACCT	ATAGGTAG	33€
SUT237	186	GCGATAAGTA	ATGTGAATTG	CAGAATTCAG	TGAATCATCG	AATCTTTGAA	CGCACATTGC	GCCCATTAGT	ATTCTAGTGG	GCATGCCTAT	TCGAGCGTCA	TTTCAACCCT	TACGCCCT-G	TAGCGTAGC-	GTTGGGAATC	TACCT	GCGGGTAG	336
SUT108	204	GCGATAAGTA	ATGTGAATTG	CAGAATTCAG	TGAATCATCG	AATCTTTGAA	CGCACATTGC	GCCCGGCAGT	ATTCTGGCGG	GCATGCCTGT	TCGAGCGTCA	TTTCAACCCT	CAAGCTC	-AGCTTGGT-	GTTGGGACTC	GCGGT	AACCCGCG	351
SUT082	185	GCGATAAGTA	ATGTGAATTG	CAGAATTCAG	TGAATCATCG	AATCTTTGAA	CGCACATTGC	GCCCATTAGT	ATTCTAGTGG	GCATGCCTAT	TCGAGCGTCA	TTTCAACCCT	TAAGCCCCTG	TTGCTTAGC-	GTTGGGAATC	TACCTCCTTC	GGGGGCGTAG	343
SUT080	226	GCGATAAGTA	ATGTGAATTG	CAGAATTCAG	TGAATCATCG	AATCTTTGAA	CGCACATTGC	GCCCATTAGT	ATTCTAGTGG	GCATGCCTAT	TCGAGCGTCA	TTTCAACCCT	TAAGCCTCAG	TTGCTTAGC-	GTTGGGACTC	TACGACCTAT	TATAGCGTAG	384
SUT116	227	GCGATAAGTA	ATGTGAATTG	CAGAATTCAG	TGAATCATCG	AATCTTTGAA	CGCACATTGC	GCCCATTAGT	ATTCTAGTGG	GCATGCCTAT	TCGAGCGTCA	TTTCAACCCT	TAAGCCTCAG	TTGCTTAGC-	GTTGGGACTC	TACGACCTAT	TATAGCGTAG	385
SUT042	225	GCGATAAGTA	ATGTGAATTG	CAGAATTCAG	TGAATCATCG	AATCTTTGAA	CGCACATTGC	GCCCATTAGT	ATTCTAGTGG	GCATGCCTAT	TCGAGCGTCA	TTTCAACCCT	TAAGCCTCAG	TTGCTTAGC-	GTTGGGACTC	TACGACCTAT	TATATCGTAG	383
SUT164	231	GCGATAAGTA	ATGTGAATTG	CAGAATTCAG	TGAATCATCG	AATCTTTGAA	CGCACATTGC	GCCCATTAGT	ATTCTAGTGG	GCATGCCTAT	TCGAGCGTCA	TTTCAACCCT	TAAGCCTCTG	TCGCTTAGC-	GTTGGGAGCC	TACGTC	TACAGCGTAT	385
SUT293	231	GCGATAAGTA	ATGTGAATTG	CAGAATTCAG	TGAATCATCG	AATCTTTGAA	CGCACATTGC	GCCCATTAGT	ATTCTAGTGG	GCATGCCTAT	TCGAGCGTCA	TTTCAACCCT	TAAGCCTCTG	TCGCTTAGC-	GTTGGGAGCC	TACGTC	TACAGCGTAT	385
SUT292	231	GCGATAAGTA	ATGTGAATTG	CAGAATTCAG	TGAATCATCG	AATCTTTGAA	CGCACATTGC	GCCCATTAGT	ATTCTAGTGG	GCATGCCTAT	TCGAGCGTCA	TTTCAACCCT	TAAGCCTCTG	TCGCTTAGC-	GTTGGGAGCC	TACGTC	TACAGCGTAT	385
SUT159	237	GCGATAAGTA	ATGTGAATTG	CAGAATTCAG	TGAATCATCG	AATCTTTGAA	CGCACATTGC	GCCCATTAGT	ATTCTAGTGG	GCATGCCTAT	TCGAGCGTCA	TTTCAACCCT	TAAGCCTT-G	TTGCTTAGC-	GTTGGGAATC	AGCGTCTTCA	CGGCGCTG-T	393
SUT162	237	GCGATAAGTA	ATGTGAATTG	CAGAATTCAG	TGAATCATCG	AATCTTTGAA	CCCACATTCC	GCCCATTAGT	ATTCTAGTCG	GCATGCCTAT	TCGAGCGTCA	TTTCAACCCT	TAAGCCTT-G	TTGCTTAGC-	<b>GTTGGGAATC</b>	AGCGTCTTCA	CGGCGCTG-T	393
SUT061	236	GCGATAAGTA	ATGTGAATTG	CAGAATTCAG	TGAATCATCG	AATCTTTGAA	CGCACATTGC	GCCCATTAGT	ATTCTAGTGG	GCATGCCTAT	TCGAGCGTCA	TTTCAACCCT	TAAGCCTT-G	TTGCTTAGC-	GTTGGGAATC	AGCGTCTTCA	CGGCGCTG-T	392
SUT280	210	GCGATAAGTA	ATGTGAATTG	CAGAATTCAG	TGAATCATCG	AATCTTTGAA	CGCACATTGC	GCCCATTAGT	ATTCTAGTGG	GCATGCCTAT	TCGAGCGTCA	TTTCAACCCT	TAAGCCTT-G	TTGCTTAGC-	GTTGGGAATC	AGCGTCTTCA	CGGCGCTG-T	366
SUT165	237	GCGATAAGTA	ATGTGAATTG	CAGAATTCAG	TGAATCATCG	AATCTTTGAA	CGCACATTGC	GCCCATTAGT	ATTCTAGTGG	GCATGCCTAT	TCGAGCGTCA	TTTCAACCCT	TAAGCCTT-G	TTGCTTAGC-	GTTGGGAATC	AGCGTCTTCA	CGGCGCTG-T	393
SUT120	238	GCGATAAGTA	ATGTGAATTG	CAGAATTCAG	TGAATCATCG	AATCTTTGAA	CGCACATTGC	GCCCATTAGT	<b>ATTCTAGTGG</b>	GCATGCCTAT	TCGAGCGTCA	TTTCAACCCT	TAAGCCCC-G	TTGCTTAGC-	GTTGGGAATC	AGCGTCTTCT	CGGCGCTG-T	394
SUT016	243	GCGATAAGTA	ATGTGAATTG	CAGAATTCAG	TGAATCATCG	AATCTTTGAA	CGCACATTGC	GCCCATTAGT	ATTCTAGTGG	GCATGCCTAT	TCGAGCGTCA	TTTCAACCCT	TAAGCCTC-T	CGGCTTAGC-	GTTGGGACCC	TGCGGCGT-A	CGGCGCAG-G	398
SUT180	243	GCGATAAGTA	ATGTGAATTG	CAGAATTCAG	TGAATCATCG	AATCTTTGAA	CGCACATTGC	GCCCATTAGT	ATTCTAGTGG	GCATGCCTAT	TCGAGCGTCA	TTTCAACCCT	TAAGCCCT-T	CGGCTTAGC-	GTTGGGACCC	TGCGGCGT-A	CGGCGCAG-G	398
ST2324	243	GCGATAAGTA	ATGTGAATTG	CAGAATTCAG	TGAATCATCG	AATCTTTGAA	CGCACATTGC	GCCCATTAGT	ATTCTAGTGG	GCATGCCTAT	TCGAGCGTCA	TTTCAACCCT	TAAGCCCT-G	CGGCTTAGC-	GTTGGGACCC	TGCGGCGT-G	CGGCGCAG-G	398
SUT250	245	GCGATAAGTA	ATGTGAATTG	CAGAATTCAG	TGAATCATCG	AATCTTTGAA	CGCACATTGC	GCCCATTAGT	ATTCTAGTGG	GCATGCCTAT	TCGAGCGTCA	TTTCAACCCT	TAAGCACT-G	TAGCTTAGCC	GTTGGGAATC	TACGGCTT-A	CCCTGTAG-T	401
SUT282	253	GCGATAAGTA	ATGTGAATTG	CAGAATTCAG	TGAATCATCG	AATCTTTGAA	CGCACATTGC	GCCCATTAGT	ATTCTAGTGG	GCATGCCTAT	TCGAGCGTCA	TTTCAACCCT	TAAGCCCT-G	TAGCTTAGC-	GTTGGGAATC	TACGGCTT-A	CCCTGTAG-T	408
SUT166	265	GCGATAAGTA	ATGTGAATTG	CAGAATTCAG	TGAATCATCG	AATCTTTGAA	CGCATATTGC	GCCCATTAGT	ATTCTAGTGG	GCATGCCTAT	TCGAGCGTCA	TTTCAACCCT	TAAGCCCTAT	CTGCTTAGC-	GTTGGGAGTC	TGC-GCC	TTGCCGCAGT	419
SUT158	228	GCGATAAGTA	ATGTGAATTG	CAGAATTCAG	TGAATCATCG	AATCTTTGAA	CCCATATTCC	GCCCATTAGT	ATTCTAGTCG	GCATGCCTAT	TCGAGCGTCA	TTTCAACCCT	TAAGC-CTTG	TTGCTTAGT-	GTTGGGAGTC	TACCGCC	TCCCCCTACT	382
SUT148	236	GCGATAAGTA	ATGTGAATTG	CAGAATTCAG	TGAATCATCG	AATCTTTGAA	CGCACATTGC	GCCCATTAGT	ATTCTAGTGG	GCATGCCTAT	TCGAGCGTCA	TTTCAACCCT	TAAGCCCCTG	TTGCTTAAC-	GTTGGGAGCC	TACGTCTG	CGGGCGTAGT	392
SUT187	236	GCGATAAGTA	ATGTGAATTG	CAGAATTCAG	TGAATCATCG	AATCTTTGAA	CGCACATTGC	GCCCATTAGT	ATTCTAGTGG	GCATGCCTAT	TCGAGCGTCA	TTTCAACCCT	TAAGCCCCTG	TTGCTTAGC-	GTTGGGAGTC	TACGTCTG	CGGGCGTAGT	392
SUT182	253	GCGATAAGTA	ATGTGAATTG	CAGAATTCAG	TGAATCATCG	AATCTTTGAA	CGCATATTGC	GCCCATTAGT	ATTCTAGTGG	GCATGCCTAT	TCGAGCGTCA	TTTCAACCCT	TAAGC-CCTG	TTGCTTAGT-	GTTAGGAGCC	TCTTTCTTCA	GAGGGCAGCT	410
SUT215	233	GCGATAAGTA	ATGTGAATTG	CAGAATTCAG	TGAATCATCG	AATCTTTGAA	CGCACATTGC	GCCCATTAGT	ATTCTAGTGG	GCATGCCTAT	TCGAGCGTCA	TTTCAACCCT	TAAGCCAGTG	CTGCTTAGT-	GTTGGGAGCA	TACCCTCCCC	GGGGGGGTATC	391
SUT221	233	GCGATAAGTA	ATGTGAATTG	CAGAATTCAG	TGAATCATCG	AATCTTTGAA	CGCACATTGC	GCCCATTAGT	ATTCTAGTGG	GCATGCCTAT	TCGAGCGTCA	TTT-AACCCT	TAAGCCAGTG	CTGCTTAGT-	GTTGGGAGCA	TACCCTCCCC	GGGGGGGTATC	390
AJ390400	268	gcgataagta	atgtgaattg	cagaattcag	tgaatcatcg	aatctttgaa	cgcacattgc	gcccattagt	attctagtgg	gcatgcctat	tcgagcgtca	tttcaaccct	taagcccctg	tcgcttagc-	gttgggaatc	tacgtt	acggcgtagt	422

Figure 9C. (Continued).

		490	500	510	) 520	530	0 54	0 55	560	) 570	580	) 59	0	
SUT223	420	TCCCGAAGAC	CA-GTGGCAG	ACCTGGGGCC	GTACCTAAGC	GTAGTAAACT	ATCACATCGC	TCTGGCTGGT	ACCCCTAGGC	TTCTAGCCGT	AAAACTGCCT	AGCAGTTGTG	AC	530
SUT218	420	TCCCGAAGAA	CA-GTGGCAG	ACCTGGGNCC	GTACCTAAGC	GTAGTAAACT	ATCACATCGC	TCTGGCTGGT	ACCCCTAGGC	TTCTAGCCGT	AAAACCGCCT	AACAGTTGTG	AC	530
SUT233	390	TCCTGAAAGT	TA-GTGGCGG	AGTTAGGGTA	CACTCTCACC	GTAGTAA	CACTTCTCGC	TCG-TGTGGT	G-GCCTGGC	TGCTGGCCGT	TAAACCCCC-	ATACCTTTTA	GT	494
SUT240	390	TCCTGAAAGT	TA-GTGGCGG	AGTTAGGGTA	CACTCTCAGC	GTAGTAA	CACTTCTCGC	TCG-TGTGGT	G-GCCCTGGC	TGCTGGCCGT	TAAACCCCC-	ATACCTTTTA	GT	494
SUT066	421	TCCTGAAAGT	TA-GTGGCGG	AGTCAGGGTG	CACTCTCAGC	GTAGTAA	TTTCTCTCGC	TTG-TGTGGT	G-TCCCTGGC	TCCTCCCCCT	TAAACCCCCT	ATATTTTCTA	GT	526
SUT068	421	TCCTGAAAGT	TA-GTGGCGG	AGTCAGGGTG	CACTCTCAGC	GTAGTAA	TTTCTCTCGC	TTG-TGTGGT	G-TCCCTGGC	TGCTGGCCGT	TAAACCCCCT	ATATTTTCTA	GT	526
SUT069	421	TCCTGAAAGT	TA-GTGGCGG	AGTCAGGGTG	CACTCTCAGC	GTAGTAA	TTTCTCTCGC	TTG-TGTGGT	G-TCCCTGGC	TGCTGGCCGT	TAAACCCCCT	ATATTTTCTA	$\mathbf{GT}$	526
SUT046	422	TCCTGAAAGT	TAAGTGGCGG	AGTTAGGGTA	CACTCTCAGC	GTAGTAA	TTTCTCTCGC	TCG-TGTGGT	G-TCCCTGGC	TGCCGGCCGT	TAAACCCCCT	ATATTTTCTA	GT	528
SUT041	445	TCCTTAA-GT	TA-GTGGCGG	AGTTAGGGTA	CACTCTCAGC	GTAGTAA-T-	ATCT-CTCGC	TCG-TGTGGT	G-GCCTTGGC	TGCTAGCCGG	TAAAAC-CCT	ATATTTTTA	AT	548
SUT063	435	TCCTTAAAGT	TA-GTGGCGG	AGTTAGGGTA	CACTCCCAGC	GTAGTAAAT-	ATCT-CTCGC	NCG-TGTGGG	G-GCCCTGGC	TGCTGCCGTT	AAAACC-CCT	ATATCTTTTA	GT	540
SUT020	459	CCCCGAAAAC	CA-GTGGCGG	TGTTCGG-TA	CACTCATAGC	GTAGTAATT-	TTCTTCTCGC	TTC-TGACGT	G-GCCTGAAT	CTTCAGCCGT	AAAACCTCCT	ATTTTCTA	GT	563
SUT294	409	TCCCTAAAAC	CA-GTGGCGG	TGTTAGG-TA	CCCTCATAGC	GTAGTAAAT-	CTTTTCTCGC	TTC-TGCAGT	G-TGTCTAGC	TACCTGCCGT	TAAACCCCCC	TATTTTTCTA	GT	515
SUT256	376	TCCTTAAAGT	CAGT-GGCGG	GGTTGGGGCCG	CGCCTTCAGC	GTAGTAGTT-	-CTATGTCGT	TGT-TGCGGC	GGCCGAAC	TTGCGGCCGT	AAAG-CCCGT	GATGCTTTTA	<b>A</b> -	479
SUT154	347	TCCTTAAAGT	GATTTGGCGG	AGCTAGTGCA	TACTCTAGGC	GTAGTAAATA	CCATTCTCGC	TTT-TGTAGT	A-GGCCTGGC	GGCTTGCCGT	AAAA-CCCCT	-ATACTTCTA	GT	454
SUT070	337	TTCCTCAAAT	CGATTGGCGG	AGTTAGCACA	TACTCTAGGC	GTAGTAACA-	CCATTCTCGC	TTC-GGTAGT	AAGTGCTGGC	GGCTAGCCAC	TAAA-CCCCC	TATACTTCTA	GT	445
SUT237	337	TTCCTCAAAT	CGATTGGCGG	AGTTAGCGCA	TACTCTAGGC	GTAGTAATA-	CCATTCTCGC	TTC-TGTAGT	A-GTGCTAGC	GGCTAGCCAT	TAAA-CCCCC	TATATTTCTA	GT	444
SUT108	352	TTCCCCAAAT	CGATTGGCGG	TCACGTCGAG	CTTCCATAGC	GTAGTAATC-	ATACAC-CTC	GTT-ACTGGT	AATCGTCGCG	GCCACGCCGT	TAAA-CCCCA	ACTTCTG	AA	456
SUT082	344	TTCCTGAAAG	TGATTGGCGG	AGTTAGAGCA	TACTCTAGGC	GTAGTAACA-	TACCTCTCGC	TTC-TGCAGT	A-GCCCTGGC	GACCTGCCGT	AAAA-CCCCC	TATACTTCTA	GT	451
SUT080	385	TTCCTTAAAG	GTAGTGGCGG	AGTTATAGCA	CACTCTAAGC	GTAGTAATT-	-CTCTCTCGC	TTCTTGTAGT	G-GCTATAAT	TGCTAGCCAT	AAAA-CACCC	CCTATTTTAA	т-	491
SUT116	386	TTCCTTAAAG	TTAGTGGCGG	AGTTATAGCA	CACTCTAAGC	GTAGTAATT-	-CTCTCTCGC	TTCTTGTAGT	G-GTTATAGT	TGCTAGCCAT	AAAA-CACCC	CCTATTTTAA	<b>T</b> -	492
SUT042	384	TTCCTTAAAG	TTAGTGGCGG	AGTTATAGCA	CACTCTAAGC	GTAGTAATT-	-CTTTCTCGC	TTCATCTAGT	G-GTTATAGT	CGCTAGCCAT	AAAA-CACCC	CTTATTTAA	<b>T</b> -	490
SUT164	386	CTCCTCAAAG	TCAGTGGCGG	TGCTGGAGCA	CACTCTCAGC	GTAGTAAGTT	TTCTTCTCGC	TTC-TGTAGT	G-GCCCTGC	AGCCTGCCGT	AAAA-CCTCC	AACACTTAGT		492
SUT293	386	CTCCTCAAAG	TCAGTGGCGG	TGCTGGAGCA	CACTCTCAGC	GTAGTA-GTT	TTCTTCTCGC	TTC-TGTAGT	G-GCCCTGC	AGCCTGCCGT	AAAA-CCTCC	AACACTTAGT		491
SUT292	386	CTCCTCAAAG	TTAGTGGCGG	TGCTGGAGCA	CACTCTCAGC	GTAGTA-GTT	TTCTTCTCGC	TTC-TGTAGT	GCCCCTGC	AGCCTGCCGT	AAAA-CCTCC	AACACTTAGT		490
SUT159	394	TCCTTAAATT	TAAGTGGCGG	AGTTATAGCA	CACTCTAAGC	GTAGTAAAT-	-CTTCCTCGT	TTC-TGAAGT	T-GCCTTGAT	TCTTAGCCGT	AAAACCCCC-	-TATTTCTA	AT	499
SUT162	394	TCCTTAAATT	TAAGTGGCGG	AGTTATAGCA	CACTCTAAGC	GTAGTAAAT-	-CTTCCTCGT	TTC-TGAAGT	T-GCCTTGAT	TCTTAGCCGT	AAAACCCCCC	-TATTTTCTA	AT	500
SUT061	393	TCCTTAAATT	TA-GTGGCGG	AGTTATAGCA	CACTCTAAGC	GTAGTAAAT-	-CTTTCTCGT	TTC-TGAAGT	T-GCCTTGAT	TCTTAGCCGT	AAAACCCCCC	-TATTTCTA	AT	498
SUT280	367	TCCTTAAATT	TA-GTGGCGG	AGTTATAGCA	CACTCTAAGC	GTAGTAAAT-	-CTTTCTCGT	TTC-TGAAGT	T-GCCTTGAT	TCTTAGCCGT	AAAACCCCCC	-TATTTTCTA	AT	472
SUT165	394	TCCTTAAATT	TA-GTGGCGG	AGTTATAGCA	CACTCTAAGC	GTAGTAAAT-	-CTTTCTCGT	TTC-TGAAGT	T-GCCTTGAT	TCTTAGCCGT	AAAACCCCCC	-TATTATCTA	AT	499
SUT120	395	TCCTTAAATT	TA-GTGGCGG	AGTTATAGCA	CACTCTAAGC	GTAGTAAAT-	-CTT-CTCGC	TTC-TGAGGT	C-GCCCTGAC	TCCTCGCCGT	AAAACCCCCT	-ATTTTTCTA	AT	499
SUT016	399	TCCTTAAATT	TA-GTGGCGG	AGTTGGTGCA	TACTCTTAGC	GTAGTAAAT-	-CTT-CTCGC	TCT-CGTAGT	A-CGCCTAGC	TACCCGCCGT	AAAACCCCCC	-TATTTTTA	AT	503
SUT180	399	TCCTTAAATT	TA-GTGGCGG	AGTTGGTGCA	TACTCTTAGC	GTAGTAAAT-	-CTT-CTCGC	TCA-CGTAGT	A-CGCCTAGC	TACCCGCCGT	AAAACCCCCC	-TATTTTTA	AT	503
ST2324	399	TCCTTAAATC	TA-GTGGCGG	AGTTGGTGCA	TACTCTTAGC	GTAGTAAAT-	-CTT-CTCGC	TCA-CGTAGT	A-CGCCTAGC	TACCCGCCGT	AAAAAACCCC	-TATTCTTTA	AT	503
SUT250	402	TCCTCAAAGT	TA-GTGGCGG	AGTTGGTGCA	TACTCTTAGC	GTAGTAATT-	-ACC-CTCGC	TCA-CGTAGT	A-TGCCTAGC	TGCCTGCCGT	AAAACCCCCT	ATATATTCTA	GT	507
SUT282	409	TCCTCAAAGT	TA-GTGGCGG	AGTTGGTGCA	TACTCTTAGC	GTAGTAATT-	-CTT-CTCGC	TCA-CGCAGT	A-AGCCCGGC	TGCCTGCCGT	AAAACCCCCC	TATATTTCTA	GT	514
SUT166	420	CCCCTAAAGG	TA-GTGGCAG	TGTTAGG-TA	CACTCGTAGC	GTAGTAATT-	TTCTTCTCGC	TTC-TGTAGT	GGTCCTGAAC	GTCTAGCCGT	TAAAACCCCT	ATTTCTCAA-		524
SUT158	383	TCCTAAAAGG	TA-GTGGCGG	TGTCAGGGCA	CACTCGTAGC	GTAGTAATT-	TTCTTCTCGC	TTC-TGTAGT	GGTTCTGG-C	AACCTGCCGT	AAAACCCCCT	ATTTTCTAGT		488
SUT148	393	TCCTGAAAGT	-A-GTG-CGG	AGTTAGGGCG	TACTCTTA-C	GTAGTAAAT-	CACTATTCG-	TAC-TG-AGT	AGT-CTAA-C	TTT-AGCCGG	AAACCCCCAT	ATTTAGT		488
SUT187	393	TCCTGAAAGT	TA-GTGGCGG	AGTTAGGGCG	TACTCTTAAC	GTAGTAAAT-	CACTATTCG-	TAC-TG-AGT	AGTTCTAA-C	TTTCAGCCGT	AAAACCCTAT	ATTTAAGT		494
SUT182	411	CCCTAAAGAT	AGTGCAG	TGTTCGGGTA	CACTCGTAGC	GTAAGTAATT	TCTATCTCGC	TTC-TCCAGT	G-GCCCGAAT	TATTCGCCGT	AAAA-CCCCT	ATTTTCTCAA		514
SUT215	392	TCCTTAAAGT	TA-GTGGCGG	AGTTAGGGTA	CACTCTCAGC	GTAGTAATT-	TCTCTCGC	TCG-GGTAGT	G-GCCTGGC	TGCTTGCCGT	TAAGCCCCTA	TTTTCTTTTA	GT	497
SUT221	391	TCCTTAAAGT	TA-GTGGCGG	AGTTAGGGTA	CACTCTCAGC	GTAGTAATT-	TCTCTCGC	TCG-GGTGGT	GGGGCCTGCT	GCTGCCGT	TAAGCCCT	ATTTCTTCGA	GT	493
AJ390400	423	tcctcaaatt	ta-gtggcgg	agttgtagca	cactctaagc	gtagtagaat	-ctt-ctcgc	ttc-tgtagt	t-actgtgac	ttttgccgt	aaaaccctct	-atttt-cta	gt	527

Figure 9C. (Continued).

		10	20	30	40	50	60	70	80	90	10	110	0 12	130	140	0 15	50 16	170	180	0
			···· [ · · · · ]	···· [ · · · · ]			****1****1			· · · · 1 · · · · 1	****1****1	···· 1 · · · · 1							····1····1	
SUT032	1	CAGAGTTC	TATTACTCCC	AAACCCATGT	GCA-CATACC	GTACGTTGCC	TCGGCAGGCG	GCGC	CTACCC	CGTAGCGCCC	TACACCCGGT	AGGGCCTG	CCCGGTGGAC	GCCGACAAAG	CCTGCCGGCG	GCCCCTO	AAAATTCTGT	TTC-TTACTG	GATCTCTGAA	16.
SUT142	1	CAGAGTTC	TATAACTCCC	AAACCCATGT	GCA-CATACC	GTACGTTGCC	TCGGCAGGCG	GTGC	CTACCC	CGTAGCGCCC	TACACCCGGT	AGGGCCTG	CCCGGTGGAC	GCCGACAAAG	CCTGCCGGCG	GCCCCTC	AAAATTCTGT	TTC-TTACTG	GATCTCTGAA	16
SUT076	1	CAGAGTTC	-AT-ACTCCC	AAACCCATGT	GCA-CATACC	GTACGTTGCC	TCGGCAGGCG	GCGC	CTACCC	CGTAGCGCCC	TACACCCGGT	AGGGCCTG	CCCGGTGGAC	GCCGACAAAG	CCTGCCGGCG	GCCCCTC	AAAATTCTGT	TTC-TTACTG	GATCTCTGAA	15
ST2417	1	CAGAGTTC	TATCACTCCC	AAACCCATGT	GCA-CATACC	GTACGTTGCC	TCGGCAGGCG	GCGC	CTACCC	CGTAGCGCCC	TACACCCGGT	AGGGCCTG	CCCGGTGGAC	GCCGACAAAG	CCTGCCGGCG	GCCCCTC	AAAATTCTGT	TTC-TTACTG	GATCTCTGAA	16
SUT207	1	AAGAGTTC	TATAACTCCC	AAACCCATGT	GAA-CATACC	TTACGTTGCC	TOGGCAGGTC	GTGC	CNACCC	CGTAGCGCCC	TACCCTGT	AGGACCTA	CCCGGTAGAC	GCGGGTAA-G	CTTGCCGGCG	GCCCACC	AAA-CTCTGT	TTA-GTATTG	AAT-TOTGAA	15
AF163033	1		CTCCC	AAACCCATGT	GAA-CATACC	TTACGTTGCC	TCGGCAGGTC	GCGC	CTACCT	AGTAGCAC-C	CTACC-CTGT	AGG-CCTA	CCCGGGAGAC	GCGGGTA-AG	CCTGCCGGCG	-CCCACC	AAACTCT-GT	TTAAT-ATTG	A-ATTCTGAA	14
AF163026	1	AAGAGTTCTA	TAACTCCCTA	AAACCCATGT	GAA-CATACC	TTACGTTGCC	TCGGCAGGTC	GCGC	CTACCC	CGTAACGT-C	CTACC-CTGT	AGG-CACCTA	CCCGGTAGGC	GCGGGTA-AG	CCTGCCGACG	-CC-ACC	AAACTCT-GT	TTAGT-ATTG	A-ATTCTGAA	15
AF163030	1			CCCATGT	GAA-CATACC	TTACGTTGCC	TCGGCAGGTC	GCGC	CTACCC	CGTAGCGC-C	CTACC-CTGT	AGG-A-CCTA	CCCGGAAGAC	GCGGGTA-AG	CCTGCCGGCG	GCCCACC	AAACTCT-GT	TTATT-TTTG	A-ATTCTGAA	13
AF163039	1	AAGAGTTCTA	TAACTCCC	AAACCCATGT	GAA-CATACC	TTACATTGCC	TCGGCAGGTC	GTGC	CTACCC	CGTAGCGT-C	CTACC-CTGT	AGG-A-CCTA	CCCGGTAGAC	GCGGGTA-AG	CCTGCCGGCG	-CCCACC	AAACTCT-GT	TTAGT-ATTG	A-ATTCTGAA	15
AF163031	1	AAGAGTTATT	ATABACTCCC	ABACCCATCT	GAA-CATACC	CTTTGTTGCC	TOGGCAGGTC	GCGC	TTACCC	CGTGAGAC-C	TTACC-CTGT	AAG-GECCEA	CCCGGTAGGC	CCCCCTA-AC	CORRECCESCO	GCCCBTC	ARACTCT-CT	TTACT-ATTC	TTATTCTCAA	16
AF163034	1	AAGAGTTCTA	TAACTCCC	ABACCCATGT	GAA-CATACC	TTACGTTGCC	TCGGCAGGTC	GCGC	CTACCC	CGTAGCGC-C	CTACC-CTGT	AGG-CCTA	CCCGG-AAGT	GCGGGTA-AG	CCTGCCGGCG	-000800	ABACTCTTGT	TTAACCATCT	GCATTCTGAA.	15
5117123	÷.	ABGAGT-GTA	TACTCCC	ANACCONTOT	GAR-CATACC	TRACGTEGOC	TOGGCAGGTC	6060		CGTAGAGC-C	CTACA-CTT	300-00073	CCCGGTGGAC	606662-36		GCTC3C1	ANACTOT-OT		T-ACTOTORA	15
572027	1	CTGAGTTATC	CARACTOCC	AACCOTTTGT	GAACT-TACC	A-CTOTTOCC	TCGGCGAGTT	GTGC	TACCC	TATAG	CTACC-CTOT	AGCTA	CCCGGG-AAC	ACATTOCAAG	CTCGCCAGAG	GACCTAC-CI	ACTCTGT	TTT-ATACTC	TATCTCTCAA	15
Cm2326	-	CT33377-00	C33330-000	ABTCOTTARA	COMPREMICOCC	ACCECTERCOC	CTTACCCCTT	GTGCCCCC		TOTACTORA	07300-0000	Games a GOTA	CCCTCCTACT	GCCC27270CC	CCCCCCCBAG	GACTARC-TR	ALCOCOPPECT	OTT-ACTORS	ANTATOTCAN	16
SIM090	÷	COCLORADO	TABB-CTCCS	AACCOMMCOM	CARCE-TACC	G-RCCRRRCC	TOCCCOTOT	CTCC		CORR.CORC	CTACC-CCCT	CCCTA	CCCCCCTATIN	G-CCCNACCC	TCCCNCCCAT	ChCCCCC-N	Acmommomm	TTT ACTORS	TATCTCTCAA	15
AV787733	-	CACACITATIO	TAAA CICCA	ABBCCCBECE	Chace-macc		TCOCCOCCC	ACCOMACC		maccomomac	mmacc-cooc	ACC-CACCER	cccmom-hoc	CCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCC	CORRECCOCC	concescant	manacommo	mmm-mma cmc	ammamondad	10
AL 707733	÷.	CAGAGITATC	TAACICCC	ANACCOATOT	Chack-Inco	1-ACOTTOCC	100000000	AGCCIACC		INCCCIDING	TTACC-COOO	AUC-OROCIA	CCCTOT-AGC	CEGETOCHOG	COIRCOCOCC	CONTOGRETATE	- IAAACICITO	TTT-TTAGIG	ATTAICIONO	10
501201	1	ANGAGITTAT	TAACICCC	AAACCCATOT	GAACA-TACC	ATACOTTOCC	TCGGCRGGCT	ACAL			CTOCT-C	TA	TTAGAGCGAT	GIGINACCOG	CCTGCCGGCG	GICCATT	AACTCIOT	TTA-TTTTTO	AAT-TCTGAG	10
012040	-	ANGAGITTAT	TAACTCCC	ANACCOATOT	GAACA-TACC	ATACOTTOCC	TCGGCAGGCT	COL	00000000		CIGCI-Case	100.00000	TTAGAGTGAT	GIGIARCCAG	COTOCOGOCO	CROWN C. CI	AACTOTOT	TTA-TTTTTG	MAT-TUTGAG	10.
501203	÷.	ANGAGITATT	A-CAACTCCC	ANACCOATOT	GAACCTTACC	ATTIGITUCC	TUGGCAGGTU	GCA	GETTACCC	TOTOAGAC-C	CTACC-CTGT	AGG-GCCTCA	CUIGGIAGIT	GCGGGTA-AT	CUIGCUGGIG	GICTA-C-C/	AACTCIGT	TTACTAIG	TTATICIGAA	10
SUT177	-	ARGAGITATT	A-CARCTCCC	ARACCCATGT	GAACCTTACC	ATTOTICC	TCGGCAGGTC	-GCA	GCTTACCC	TOTGAGAT-C	CTACC-CTGT	AGG-GCCTCA	CCTGGTAGTT	GCGGGTA-AT	COTOCCOUTO	GTCTA-C-C/	AACTCTGF	TTACTATO	TTATTCTGAA	10
SUTISE	+	AAGAGTTATT	A-CAACTCCC	AAACCCATGT	GAACCTTACC	ATTTOTTOCC	TCGGCAGGTC	-GCA	GCTTACCC	TGTGCGAC-C	CTACC-CTGT	AGG-GCCTCA	CCTGGTAGTT	GCGGGGTA-AT	CETGECGGTG	GTCTA-T-CA	AACTCTGT	TTACTATG	TTATTCTGAA	10
SUT139	1	AAGAGTTATT	A-CAACTCCC	ARACCCATGT	GAACCTTACC	ATTTGTTGCC	TCGGCAGGTC	-GCAA	GCTTACCC	TGTGCGAC-C	CTACC-CTGT	AGG-GCCTCA	CCTGGTAGTT	GCGGGTA-AT	CCTGCCGGTG	GTCTA-C-CA	AACTCTGT	TTACTATG	TTATTCTGAA	10.
SUT129	1	AAGAGTTT	A-CAACTCCC	AAACCCATGT	GAACCTTACC	ATTTGTTGCC	TEGGEAGGTE	-GCA	GCTTACCC	TGTGCGAC-C	CTACC-CTGT	AGG-GCCTCA	CCTGGTAGTT	GCGGGTA-AT	CCIGCCGGIG	GTCTA-T-C/	A AACTCTGT	TTACTATG	TTATTCTGAA	15
ST2372	1	AAGAGTTAAA	A-CAACTCCT	AAACCCATGT	GAA-CCTACC	-TTTGTTGCC	TCGGCAGGTC	TGCA	ACTTACCC	CGAGGGGA-C	CTACC-CTGT	AGGGACCTTA	CCCGGTAGTT	GCGGGGCATAA	CCTGCCGGTG	GTCTA-C-TA	AACTCTGT	TTACTATG	TTATTCTGAA	16
SUT192	1	AAGAGTTAAA	AACAACTCCT	AAACCCATGT	GAAACCTACC	-TTTGTTGCC	TCGGCAGGTC	TGCA	ACTTACCC	TGAGGGGA-C	CTACC-CTGT	AGGTACCTTA	CCCGGTAGTT	GCGGGGCATAA	CCTGCCGGTG	GTCTA-C-TI	AACTCTGT	TTACTATG	TCATTCTGAA	10.
SUTOBB	1	AAGAGTTATA	CAACTCCT	AAACCCATGT	GAA-CCTACC	-TTTGTTGCC	TCGGCAGGTC	TGCA	GCCTACCC	TGTGAGGG-C	CTACC-CTGT	AGG-ATCTTA	CGCGGTAGTT	GCAGGTTCAA	CCTGCCGCTG	GTCTA-C-C?	A AACTCTGT	TTT-ACCATG	TTATTCTGAA	16
SUT140	1	AAGAGTTATA	CAACTCCT	AAACCCATGT	GAA-CCTACC	-TTTGTTGCC	TCGGCAGGCC	TGCA	GCTTACCC	TGTGAGCC-C	CTACC-CTGT	AGGGACCTTA	CCCGGTAGTT	GCGGATACAA	CCTGCCGGTG	GTCTA-C-TA	AACTCTGT	TTACTATG	TTATTCTGAA	16
AF163027	1	AAGAGTTATT	A-CAACTCCC	AAACCCATGT	GAA-CATACC	TTCTGTTGCC	TCGGCAGGTC	TGCA	GCCTACCC	TGTAAGCC-C	CTACC-CTGT	AGGGACCTTA	CCCGGTAGTT	GCGGGTAAAG	CCTGCCGGTG	GTCTA-C-TC	AACTCTGT	TTATTATG	TTATTCTGAA	16:
ST2348	1	AAGAGTT-AT	TACAACTCCC	AAACCCATGT	GAA-CTTACC	TTCTGTTGCC	TCGGCAGGTC	-GCG	ACCTACCC	TGTGAGGC-C	TTACC-CTGT	AGG-GCCCTA	CTTGGTAGTC	GCGGGTA-CG	CCTGCCGGTG	GCCCA-T-GZ	A AACTCTGT	TTATTCT-TG	TTATTCTGAA	16
5T2363	1	AAGAGTAT	TACAACTCCC	AAACCCATGT	GAA-CTTACC	TTCTGTTGCC	TCGGCAGGTC	-GCG	ACCTACCC	TGTGAGGC-C	CTACC-CTGT	AGG-GCCCTA	CCTGGTAGTC	GCGGGTA-CG	CCTGCCGGTG	GCCCA-T-GI	A AACTCTGT	TTATTCT-TG	TTATTCTGAA	15
AF163037	1	AAGAGTT-AT	TACAACTCCC	AAACCCATGT	GAA-CTTACC	TTCTGTTGCC	TCGGCAGGTC	-GTG	ACCTACCC	TGTGAGGC-C	CTACC-CTGT	AGG-GCCCTA	CCCGGTAGTC	GCGGGTA-CG	CCTGCCGGTG	GCCCA-T-GI	A AACTCTGT	TTATTCA-TG	TCATTCTGAA	16
AF163040	1	AAGAGTT-TT	GATAACTCCC	AAACCCATGT	GAA-CTTACC	TTCTGTTGCC	TCGGCAGGTC	-GCG	TCTACCC	TGTG-GCA-C	CTACC-CTGT	AGG-ACCCGA	CCTGGTGGTC	GCGGTCA-TG	CCTGCCGGTG	GCCCT-T-TZ	A AACTTTCTGT	GTATTCTATG	TTATTCTGAG	16
SUT078	1	CAGAGTTTGA	ACGAACTCC-	AAACCCATGT	GAA-CTTACC	TTCTGTTGCC	TCGGCAGGGT	CGCG	CCTACCG	TGTGAGGC-C	CTACCACTGT	AGG-GCCCTA	CGCGGTGCGT	GCGGGGCAGC-	CCTGCCGGCG	GCCCGTC	AAA-TTCTGT	TTG-ACTACG	TTATTCTGAA	16
SUT028	1	AAGAGTTTGA	AC-AACTCC-	AAACCCATGT	GAA-CTN-CC	TTCTGTTGCC	TCGGCAGGGT	CGCG	CCTACCG	TGTGAGGC-C	CTACCACTGT	AGG-GCCCTA	CGCGGTGCGT	GCGGGGCAGC-	CCTGCCGGCG	GCCCGC	AAA-TTCTGT	TTG-ACTACG	TTATTCTGAA	15
SUT124	1	AAGAGTAT	AC-AACTCC-	AAACCCATGT	GAA-CTTACC	GTACGTTGCC	TCGGCAGG-T	CGCG	CTCACCC	CGTAACAC-C	CTACCACG-T	AGG-GGCCTA	CTCGGTGGCC	GCGGACTAAG	CCTGTCGGTG	GCCCAAC-CZ	AACTCTGTCA	GTG-ATTGTG	TCTTCTGAAC	16
SUT125	1	AAGAGTAT	AC-AACTCC-	AAACCCATGT	GAA-CTTACC	GTACGTTGCC	TCGGCAGG-T	CGCG	CTCACCC	CGTAGCAC-C	CTACCACG-T	AGG-GGCCTA	CTCGGTGGCC	GCGGACTAAG	CCTGTCGGTG	GCCCAAC-CZ	A AACTCTGTCA	GTG-ATTGTG	TCTTCTGAAC	16
SUT074	1	CCGAGT-T	AC-AACTCCC	AAACCCATGT	GAA-CATACC	TACTGTTGCT	TCGGCGGGAT	TGCC	CCGGGCG	CCTCGTGT-G	CCCCGGAT-C	AGG-CGCCCG	CCTAG-GAAC	TTGAACT	CTTGTTTT	ATTTTO	AATCTTCTGA	GTA-GTT		13
SUT027	1	AAGAGTT	TATAACTCCC	AAACCCATGT	GAA-CATACC	TAACGTTGCC	TCGGCGGGTC	GTAC	CTACCC	TGTAGTGCAC	TTACCTGT	AAG-TGCCTA	CCCGGTAGGC	ACGGGTA-AG	CCCGCCGGCG	CCCCATTZ	AACTCTGT	TTAATTACTG	GATATCTGAA	15
SUT198	1	AAGAGTT	TATAACTCCC	AAACCCATGT	GAA-CATACC	TAACGTTGCC	TCGGCGGGTC	GTAC	CTACCC	TGTAGTGCAC	TTACCTGT	AAG-TGCCTA	CCCGGTAGGC	ACGGGTA-AG	CCCGCCGGCG	CCCCATTJ	AACTCTGT	TTAATTACTG	GATATCTGAA	15
SUT155	1	AAGAGTT	TATAACTCCC	AAACCCATGT	GAA-CATACC	TAACGTTGCC	TCGGCGGGTC	GTAC	CTACCC	TGTAGTGCAC	TTACCTGT	AAG-TGCCTA	CCCGGTAGGC	ACGGGTA-AG	CCCGCCGGCG	CCCCATTI	AACTCTGT	TTAATTACTG	GATATCTGAA	15
SUT200	1	AAGAGTT	TATAACTCCC	AAACCCATGT	GAA-CATACC	TAACGTTGCC	TCGGCGGGTC	GTAC	CTACCC	TGTAGTGCAC	TTACCTGT	AAG-TGCCTA	CCCGGTAGGC	ACGGGTA-AG	CCCGCCGGCG	CCCCATT	A AACTCTGT	TTAATTACTG	GATATCTGAA	15
572298	1	AAGAGTT	TATAACTCCC	AAACCCATGT	GAA-CATACC	TAACGTTGCC	TCGGCGGGTC	GTAC	CTACCC	TGTAGTGCAC	TTACCTGT	AAG-TGCCTA	CCCGGTAGGC	ACCCCTA-AC	CCCGCCGGCG	CCCCATTI	AACTCTGT	TTAATTACTG	GATATCTGAA	15
SUT127	1	AAGAGTT	CTATACTCCC	AAACCCATGT	GAA-CATACC	GTATGTTGCC	TCGGCAGGTC	GTGT	CTACCC	TGTGGTGCCT	TACCCTGT	AGGGCCTA	CCTGGTAGAT	CCGGATAG	CCTGCCGACG	GCCCCTCZ	AACTCTGT	TTAAT-AGTG	AATCTCTGAA	15
SUT195	1	AAGAGTT	CTATACTCCC	AAACCCATGT	GAA-CATACC	GTATGTTGCC	TCGGCAGGTC	GTGT	CTATCC	TGTGGTGCCC	TACCCTGT	AGGGCCTA	CCTGGTAGAT	CCGGATAG	CCTGCCGACG	GCCCCTCJ	AACTCTGT	TTAAT-AGTG	ANTOTOTGAN	15
SUT130	1	AAGAGTT	CTATACTCCC	AAACCCATGT	GAA-CATACC	GTATGTTGCC	TCGGCAGGTC	GTGT	CTACCC	TGTGGTGTCC	TACCCTGT	AAGGCCTA	CCTGGTAGAT	CCGGATAG	CCTGCCGACG	GCCCCTCA	AACTCTGT	TTAAT-AATG	AATCTCTGAA	15
AF163042	1	AAGAGTTT	TATAACTCCC	AAACCCATGT	GAA-CATACC	GTACGTTGCC	TCGGTGCGTC	TCCCCGTGAG	G-ACCTACCC	TGTAGGAC-G	CTACG-CTGT	AAGGOTTA	TCGGGAAGAT	GCACTAA-AG	CCTGCCGGCG	GCCCATTZ	AACTCTGT	TTA-TTTTTG	AATT-CTGAG	16
572382	1	AAGAGTTC	TATGACTCCC	AAACCCATGT	GAA-CATACC	GTACGTTGCC	TOGGOGGGGGTC	TACCCTGTAG	CACCOTACCO	TGTAAGAC-C	CTACC-CTGT	AGGAGACCTA	CCCGGCAGAC	GNGGGTA-AG	CCTGCCGGCG	GCCCA-CGCZ	AACTCTGT	TTTGGCAATG	TAATTCTGAA	17
SUT092	1	GCAGGTTGCG	CCTACTTCGT	GGCCACATGT	GCGGCCTACA	TTGTAGGA	GCTATGGACT	ATTCCTCCTA	G-ACCTACCC	TOTAGTAGAC	GTACC-TTGT	AGATATCATA	CCTGGTAGAC	GCGGGTA-AG	CCTGCCGGTG	GCCCBTT	AACACTCTCT	TTA-GCGTTG	TOTTCTGAG	17

Figure 10C. The ITS1-5.8S-ITS2 sequence alignment of *Xylaria* specimens for phylogenetic construction in Figure 97 by using ClustalX

and BioEdit programs. Arrows indicate the start and the end of 5.8S rDNA sequences.

	190	200	210	22	0 23	0 24	25	26	0 27	28	29	0 30	31	32	) 330	340	35	360	
				···· I · · · · I							····I····I		****				···· [ · · · · [		
SUT032_Xba	162 C-ATATACCA A	AAATACGTTA	AAACTTTCAA	CAACGGATCT	CTTGGTTCTG	GCATCGATGA	AGAACGCAGC	GAAATGCGAT	AAGTAATGTG	AATTGCAGAA	TTCAGTGAAT	CATCGAATCT	TTGAACGCAC	ATTGCGCCCA	TTAGTATTCT	AGTGGGGCATG	CCTGTTCGAG	CGTCATTTCA	34
SUT142	162 C-ATATACCA A	AAATACGTTA	AAACTTTCAA	CAACGGATCT	CTTGGTTCTG	GCATCGATGA	AGAACGCAGC	GAAATGCGAT	AAGTAATGTG	AATTGCAGAA	TTCAGTGAAT	CATCGAATCT	TTGAACGCAC	ATTGCGCCCA	TTAGTATTCT	AGTGGGCATG	CCTGTTCGAG	CGTCATTTCA	34
SUT076	160 C-ATATACCA A	AAATACGTTA	AAACTTTCAA	CAACGGATCT	CTTGGTTCTG	GCATCGATGA	AGAACGCAGC	GAAATGCGAT	AAGTAATGTG	AATTGCAGAA	TTCAGTGAAT	CATCGAATCT	TTGAACGCAC	ATTGCGCCCA	TTAGTATTCT	AGTGGGCATG	CCTGTTCGAG	CGTCATTTCA	33
ST2417	162 C-ATATACCA A	AAATACGTTA	AAACTTTCAA	CAACGGATCT	CTTGGTTCTG	GCATCGATGA	AGAACGCAGC	GAAATGCGAT	AAGTAATGTG	AATTGCAGAA	TTCAGTGAAT	CATCGAATCT	TTGAACGCAC	ATTGCGCCCA	TTAGTATTCT	AGTGGGGCATG	CCTGTTCGAG	CGTCATTTCA	34
SUT207	157 C-CTGTAACA A	AAATAAGTTA	AAACTTTCAA	CAACGGATCT	CTTGGTTCTG	GCATCGATGA	AGAACGCAGC	GAAATGCGAT	ANGTANTOTO	AATTGCAGAA	TTCAGTGAAT	CATCGAATCT	TTGAACGCAC	ATTGCGCCCA	CTAGTATTCT	AGTGGGGCATG	CCTGTTCGAG	CGTCATTTCA	33
AF163033	142 CA-TATAACT A	AAATAAGTTA	AAACTTTCAA	CAACGGATCT	CTTGGTTCTG	GCATCGATGA	AGAACGCAGC	GAAATGCGAT	AAGTAATGTG	AATTGCAGAA	TTCAGTGAAT	CAT-GAATCT	TTGAACGCAC	ATTGCGCCCA	TTAGTATTCT	AGTGGGGCATG	CCTGTTCGAG	CGTCATTTCA	31
AF163026	158 CC-TATAACT A	AAATAAGTTA	AAACTTTCAA	CAACGGATCT	CTTGGTTCTG	GCATCGAT-A	AGAACGCAGC	GAAATGCGAT	ANGTAATGTG	AATTGCAGAA	TTCAGTGAAT	CATCGAATCT	TTGAACGCAC	ATTGCGCCCA	TTAGTATTCT	AGTGGGTATG	CCTGTTCGAG	CGTCATTTCA	33
AF163030	136 CC-TATAACT A	AAATAAGTTA	AAACTTTCAA	CAACGGATCT	CTTGGTTCTG	GCATCGATGA	AGAACGCAGC	GAAATGCGAT	AAGTAATGTG	AATTGCAGAA	TTCAGTGAAT	CATCGAATCT	TTGAACGCAC	ATTGCGCCCA	TTAGTATTCT	AGTGGGGCATG	CCTGTTCGAG	CGTCATTTCA	31.
AF163039	156 CC-TATAACT A	AAATAAGTTA	AAACTTTCAA	CAACGGATCT	CTTGGTTCTG	GCATCGATGA	AGAACGCAGC	GAAATGCGAT	AAGTAATGTG	AATTGCAGAA	TTCAGTGAAT	CATCGAATCT	TTGAACGCAC	ATTGCGCCCA	TTAGTATTCT	AGTGGGCATG	CCTGTTCGAG	CGTCATTTCA	33.
AF163031	161 CC-TATAACT A	AAATAAGTTA	AAACTTTCAA	CAACGGATCT	CTTGGTTCTG	GCATCGATGA	AGAACGCAGC	GAAATGCGAT	AAGTAATGTG	AATTGCAGAA	TTCAGTGAAT	CATCGAATCT	TTGAACGCAC	ATTGCGCCCA	TTAGTATTCT	AGTEGECATE	CCTGTTCGAG	CGTCATTTCA	33
AF163034	15/ AA-CATAACT A	AAATACGTTA	AAACTTTCAA	CAACGGATCT	CTTGGTTCTG	GCATCGAT-A	AGAACGGAGC	GAAATGCGAT	AAGTAATGTG	AATTGCAGAA	TTCAGTGAAT	CATCGAATCT	TTGAACGCAC	ATTGCGCCCA	TTAGTATTCT	AGTGGGGCATG	CCTGTTCGAG	CGTCATTTCA	33
SUT123	156 CAATATAACT A	AAATAAGTTA	АЛАСТТТСАА	CAACGGATCT	CTTGGTTCTG	GCATCGATGA	AGAACGCAGC	GAAATGCGAT	AAGTAATGTG	AATTGCAGAA	TTCAGTGAAT	CATCGAATCT	TTGAACGCAC	ATTGCGCCCA	TTAGTATTCT	AGTGGGCATG	CCTGTTCGAG	CGTCATTTCA	33
ST2027	151 TTTTATAACT A	AAATAAGTTA	AAACTTTCAA	CAACGGATCT	CTTGGTTCTG	GCATCGATGA	AGAACGCAGC	GAAATGCGAT	AAGTAATGTG	AATTGCAGAA	TTCAGTGAAT	CATCGAATCT	TTGGACGCAC	ATTGCGCCCA	TTAGTATTCT	AGTGGGGCATG	CCTATTCGAG	CGTCATTTCA	33
ST2326	168 TGCTTCAACT T	TAATAAGTTA	AAACTTTCAA	CAACGGATCT	CTTGGTTCTG	GCATCGATGA	AGAACGCAGC	GAAATGCGAT	ANGTANTOTO	AATTGCAGAA	TTCAGTGAAT	CATCGAATCT	TTGAACGCAC	ATTGCGCCCA	TTAGTATTCT	AGTGGGGCATG	CCTATTCGAG	CGTCATTTCA	34
SUT090	160 TTGAAACT C	SAATAAGTTA	AAACTTTCAA	CAACGGATCT	CTTGGTTCTG	GCATCGATGA	AGAACGCAGC	GAAATGCGAT	AAGTAATGTG	AATTGCAGAA	TTCAGTGAAT	CATCGAATCT	TTGAACGCAC	ATTGCGCCCA	TTAGTATTCT	AGTGGGGCATG	CCTATTCGAG	CGTCATTTCA	33
AY787733	168 TOTTTATACT T	TAATAAGTTA	AAACTTTCAA	CAACGGATCT	CTTGGTTCTG	GCATCGAT-A	AGAACGCAGC	GAAATGCGAT	ARGTAATGTG	AATTGCAGAA	TTCAGTGAAT	CATCGAATCT	TTGAACGCAC	ATTGCGCCCA	TTAGTATTCT	AGTGGGCATG	CCTGTTCGAG	CGTCATTTCA	34
SUT201	133 GTTATATA A	AAATAAGTTA	AAACTTTCAA	CAACGGATCT	CTTGGTTCTG	GCATCGATGA	AGAACGCAGC	GAAATGCGAT	ANGTAATGTG	AATTGCAGAA	TTTAGTGAAT	CATCGAATCT	TTGAACGCAC	ATTGCGCCCA	TTAGTATTCT	AGTGGGGCATG	CCTGTTCGAG	CGTCATTTCA	31
ST2349	133 GTTACATA A	AAATAAGTTA	AAACTTTCAA	CAACGGATCT	CTTGGTTCTG	GCATCGATGA	AGAACGCAGC	GAAATGCGAT	AAGTAATGTG	AATTGCAGAA	TTTAGTGAAT	CATCGAATCT	TTGAACGCAC	ATTGCGCCCA	TTAGTATTCT	AGTGGGGCATG	CCTGTTCGAG	CGTCATTTCA	31
SUT203	161 TATTATAACT A	AAATAAGTTA	AAACTTTCAA	CAACGGATCT	CTTGGTTCTG	GCATCGATGA	AGAACGCAGC	GAAATGCGAT	AAGTAATGTG	AATTGCAGAA	TTCAGTGAAT	CATCGAATCT	TTGAACGCAC	ATTGCGCCCA	TTAGTATTCT	AGTGGGGCATG	CCTGTTCGAG	CGTCATTTCA	34
SUT177	161 TATTATAACT A	AAATAAGTTA	AAACTTTCAA	CAACGGATCT	CTTGGTTCTG	GCATCGATGA	AGAACGCAGC	GAAATGCGAT	AAGTAATGTG	AATTGCAGAA	TTCAGTGAAT	CATCGAATCT	TTGAACGCAC	ATTGCGCCCA	TTAGTATTCT	AGTGGGGCATG	CCTGTTCGAG	CGTCATTTCA	34
SUT138	161 TATTATAACT A	AAATAAGTTA	AAACTTTCAA	CAACGGATCT	CTTGGTTCTG	GCATCGATGA	AGAACGCAGC	GAAATGCGAT	AAGTAATGTG	AATTGCAGAA	TTCAGTGAAT	CATCGAATCT	TTGAACGCAC	ATTGCGCCCA	TTAATATTCT	AGTGGGGCATG	CCTGTTCGAG	CGTCATTTCA	34
SUT139	162 TATTATAACT A	AAATAAGTTA	ARACTITCAR	CAACGGATCT	CTTGGTTCTG	GCATCGATGA	AGAACGCAGC	GAAATGCGAT	AAGTAATGTG	AATTGCAGAA	TTCAGTGAAT	CATCGAATCT	TTGAACGCAC	ATTGCGCCCA	TTAGTATTCT	AGTGGGGCATG	CCTGTTCGAG	CGTCATTTCA	34
SUT129	159 TATTATAACT A	AATAAGTTA	AAACTTTCAA	CAACGGATCT	CTTGGTTCTG	GCATCGATGA	AGAACGCAGC	GAAATGCGAT	AAGTAATGTG	AATTGCAGAA	TTCAGTGAAT	CATCGAATCT	TTGAACGCAC	ATTGCGCCCA	TTAGTATTCT	AGTGGGGCATG	CCTGTTCGAG	CGTCATTTCA	33
ST2372	162 TAATATAACT A	AAATAAGTTA	AAACTTTCAA	CAACGGATCT	CTTGGTTCTG	GCATCGATGA	AGAACGCAGC	GAAATGCGAT	AAGTAATGTG	AATTGCAGAA	TTCAGTGAAT	CATCGAATCT	TTGAACGCAC	ATTGCGCCCA	TTAGTATTCT	AGTGGGGCATG	CCTGTTCGAG	CGTCATTTCA	34
SUT192	164 TAATATAACT A	AAATAAGTTA	AAACTTTCAA	CAACGGATCT	CTTGGTTCTG	GCATCGATGA	AGAACGCAGC	GAAATGCGAT	ANGTANTOTO	AATTGCAGAA	TTCAGTGAAT	CATCGAATCT	TTGAACGCAC	ATTGCGCCCA	TTAGTATTCT	AGTEGECATE	CCTGTTCGAG	COTCATTICA	34.
SUTOBB	161 TAATATAACT A	AAATAAGTTA	AAACTTTCAA	CAACGGATCT	CTTGGTTCTG	GCATCGATGA	AGAACGCAGC	GAAATGCGAT	AAGTAATGTG	AATTGCAGAA	TTCAGTGAAT	CATCGAATCT	TTGAACGCAC	ATTGCGCCCA	TTAGTATTCT	AGTGGGCATG	CCTGTTCGAG	CGTCATTTCA	34
SUT140	161 TAGTATAACT A	AAATAAGTTA	AAACTTTCAA	CAACGGATCT	CTTGGTTCTG	GCATCGATGA	AGAACGCAGC	GAAATGCGAT	AAGTAATGTG	AATTGCAGAA	TTCAGTGAAT	CATCGAATCT	TTGAACGCAC	ATTGCGCCCA	TTAGTATTCT	AGTGGGGCATG	CCTGTTCGAG	COTCATTTCA	34
AF163027	163 TACTATAACT A	AAATAAGTTA	AAACTTTCAA	CAACGGATCT	CTTGGTTCTG	GCATCGAGTA	AGAACGCAGC	GAAATGCGAT	AAGTAATGTG	AATTGCAGAA	TTCAGTGAAT	CATCGAATCT	TTGAACGCAC	ATTGCGCCCA	TTAGTATTCT	AGTGGGGCATG	CCTGTTCGAG	CGTCATTTCA	34:
ST2348	161 T-CTATAACT A	AAATAAGTTA	AAACTTTCAA	CAACGGATCT	CTTGGTTCTG	GCATCGATGA	AGAACGCAGC	GAAATGCGAT	AAGTAATGTG	AATTGCAGAA	TTCAGTGAAT	CATCGAATCT	TTGAACGCAC	ATTGCGCCCA	TTAGTATTCT	AGTGGGCATG	CCTGTTCGAG	CGTCATTTCA	33
ST2363	160 T-CTATAACT A	AAATAAGTTA	AAACTTTCAA	CAACGGATCT	CTTGGTTCTG	GCATCGATGA	AGAACGCAGC	GAAATGCGAT	AAGTAATGTG	AATTGCAGAA	TTCAGTGAAT	CATCGAATCT	TTGAACGCAC	ATTGCGCCCA	TTAGTATTCT	AGTGGGGCATG	CCTGTTCGAG	CGTCATTTCA	33
AF163037	161 T-CTATAACT A	AAATAAGTTA	AAACTTTCAA	CAACGGATCT	CTTGGTTCTG	GCATCGATGA	AGAACGCAGC	GAAATGCGAT	AAGTAATGTG	AATTGCAGAA	TTCAGTGAAT	CATCGAATCT	TTGAACGCAC	ATTGCGCCCA	TTAGTATTCT	AGTGGGGCATG	CCTGTTCGAG	CGTCATTTCA	33
AF163040	162 T-TGGCAACT A	AAATAAGTTA	AAACTTTCAA	CAACGGATCT	CTTGGTTCTG	GCATCGATGA	AGAACGCAGC	GAAATGCGAT	AAGTAATGTG	ANTTGCAGAA	TTCAGTGAAT	CATCGAATCT	TTGAACGCAC	ATTGCGCCCA	TTAGTATTCT	AGTGGGGCATG	CCTGTTCGAG	CGTCATTTCA	34
SUT078	162 T-ACATAACA A	AAAGAGTTAA	AAACTTTCAA	CAACGGATCT	CTTGGTTCTG	GCATCGATGA	AGAACGCAGC	GAAATGCGAT	AAGTAATGTG	AATTGCAGAA	TTCAGTGAAT	CATCGAATCT	TTGAACGCAC	ATTGCGCCCA	TTAGTATTCT	AGTGGGGCATG	CCTGTTCGAG	CGTCATTTCA	34
SUT028	159 T-ACATAACA A	AAAGAGTTAA	AAACTTTCAA	CAACGGATCT	CTTGGTTCTG	GCATCGATGA	AGAACGCAGC	GRAATGCGAT	AAGTAATGTG	AATTGCAGAA	TTCAGTGAAT	CATCGAATCT	TTGAACGCAC	ATTGCGCCCA	TTAGTATTCT	AGTGGGGCATG	CCTGTTCGAG	CGTCATTTCA	33.
SUT124	161 T-TGACAACG A	AAATACGTTA	AAACTTTCAA	CAACGGATCT	CTTGGTTCTG	GCATCGATGA	AGAACGCAGC	GAAATGCGAT	AAGTAATGTG	AATTGCAGAA	TTCAGTGAAT	CATCGAATCT	TTGAACGCAC	ATTGCGCCCA	TTAGTATTCT	AGTGGGGCATG	CCTGTTCGAG	CGTCATTTCA	33
SUT125	161 T-TGACAACG A	AAATACGTTA	AAACTTTCAA	CAACGGATCT	CTTGGTTCTG	GCATCGATGA	AGAACGCAGC	GAAATGCGAT	AAGTAATGTG	AATTGCAGAA	TTCAGTGAAT	CATCGAATCT	TTGAACGCAC	ATTGCGCCCA	TTAGTATTCT	AGTGGGCATG	CCTGTTCGAG	CGTCATTTCA	33
SUT074	140 T-TTACAAAT A	AAATAA	AAACTTTCAA	CAACGGATCT	CTTGGTTCTG	GCATCGATGA	AGAACGCAGC	GAAATGCGAT	AAGTAATGTG	AATTGCAGAA	TTCAGTGAAT	CATCGAATCT	TTGAACGCAC	ATTGCGCCCG	CCAGTATTCT	GGCGGGGCATG	CCTGTCTGAG	CGTCATTTCA	31.
SUT027	159 TTACAACT A	AAATAAGTTA	AAACTTTCAA	CAACGGATCT	CTTGGTTCTG	GCATCGATGA	AGAACGCAGC	GAAATGCGAT	AAGTAATGTG	AATTGCAGAA	TTCAGAGAAT	CATCGAATCT	TTGAACGCAC	ATTGCGCCCA	TTAGTATTCT	AGTGGGGCATG	CCTGTTCGAG	CGTCATTTCA	33
SUT198	159 TTACAACT A	AAATAAGTTA	AAACTTTCAA	CAACGGATCT	CTTGGTTCTG	GCATCGATGA	AGAACGCAGC	GAAATGCGAT	AAGTAATGTG	AATTGCAGAA	TTCAGTGAAT	CATCGAATCT	TTGAACGCAC	ATTGCGCCCA	TTAGTATTCT	AGTGGGGCATG	CCTGTTCGAG	CGTCATTTCA	33
SUT155	159 TTACAACT A	AAATAAGTTA	AAACTTTCAA	CAACGGATCT	CTTGGTTCTG	GCATCGATGA	AGAACGCAGC	GAAATGCGAT	AAGTAATGTG	AATTGCAGAA	TTCAGTGAAT	CATCGAATCT	TTGAACGCAC	ATTGCGCCCA	TTAGTATTCT	AGTGGGGCATG	CCTGTTCGAG	CGTCATTTCA	33
SUT200	159 TTACAACT A	AAATAAGTTA	AAACTTTCAA	CAACGGATCT	CTTGGTTCTG	GCATCGATGA	AGAACGCAGC	GAAATGCGAT	AAGTAATGTG	AATTGCAGAA	TTCAGTGAAT	CATCGAATCT	TTGAACGCAC	ATTGCGCCCA	TTAGTATTCT	AGTGGGGCATG	CCTGTTCGAG	CGTCATTTCA	33
ST2298	159 TTACAACT A	AATAAGTTA	AAACTTTCAA	CAACGGATCT	CTTGGTTCTG	GCATCGATGA	AGAACGCAGC	GAAATGCGAT	AAGTAATGTG	AATTGCAGAA	TTCAGTGAAT	CATCGAATCT	TTGAACGCAC	ATTGCGCCCA	TTAGTATTCT	AGTEGECATE	CCTGTTCGAG	CGTCATTTCA	33
SUT127	156 CTATAACT A	AAATAAGTTA	AAACTTTCAA	CAACGGATCT	CTTGGTTCTG	GCATCGATGA	AGAACGCAGC	GAAATGCGAT	AAGTAATGTG	AATTGCAGAA	TTCAGTGAAT	CATCGAATCT	TTGAACGCAC	ATTGCGCCCA	TTAGTATTCT	AGTGGGGCATG	CCTGTTCGAG	CGTCATTTCA	33
SUT195	156 CTATAACT A	AAATAAGTTA	AAACTTTCAA	CAACGGATCT	CTTGGTTCTG	GCATCGATGA	AGAACGCAGC	GAAATGCGAT	AAGTAATGTG	AATTGCAGAA	TTCAGTGAAT	CATCGAATCT	TTGAACGCAC	ATTGCGCCCA	TTAGTATTCT	AGTGGGGCATG	CCTGTTCGAG	CGTCATTTCA	33.
SUT130	156 CTATAACT A	AAATAAGTTA	AAACTTTCAA	CAACGGATCT	CTTGGTTCTG	GCATCGATGA	AGAACGCAGC	GAAATGCGAT	AAGTAATGTG	AATTGCAGAA	TTCAGTGAAT	CATCGAATCT	TTGAACGCAC	ATTGCGCCCA	TTAGTATTCT	AGTGGGGCATG	CCTGTTCGAG	CGTCATTTCA	33
AF163042	166 G-CTATAAT- A	AAATAAGTTA	AAACTTTCAA	CAACGGATCT	CTTGGTTCTG	GCATCGATGA	AGAACGCAGC	GAAATGCGAT	AAGTAATGTG	AATTGCAGAA	TTTAGTGAAT	CATCGAATCT	TTGAACGCAC	ATTGCGCCCA	CTAGTATTCT	AGTGGGGCATG	CCTGTTCGAG	CGTCATTTCA	34
572382	172 TACTATAACT A	AAATCAGTTA	AAACTTTCAA	CAACGGATCT	CTTGGTTCTG	GCATCGATGA	AGAACGCAGC	GAAATGCGAT	AAGTAATGTG	AATTGCAGAA	TTCAGTGAAT	CATCGAATCT	TTGAACGCAC	ATTGCGCCCA	TTAGTATTCT	AGTEGECATE	CCTGTTCGAG	CGTCATTTCA	35
SUT092	172 T-ATACAACG A	AAAGAATTAA	AAACTTTCAA	CAACGGATCT	CTTGGTTCTG	GCATCGATGA	AGAACGCAGC	GAAATGCGAT	AAGTAATGTG	AATTGCAGAA	TTCAGTGAAT	CATCGAATCT	TTGAACGCAC	ATTGCGCCCA	TTAGTACTCT	AGTGGGGCATG	CCTGTTCGAG	CGTCATTTCA	35

Figure 10C. (Continued).

		37	0 38	0 39	0 40	0 410	42	0 43	0. 440	45	0 46	0 4	70 48	0 49	0 50	0 51	0 52	9	
0012-0222-0203	12072												1						inerere.
SUT032_Xba	341	ACCCTTAAGC	CC-CTGTTGC	TTA-CGTTGG	GAGCCTACC-	GTCACA	CGTAGCTCCT	GAAAAGTAGT	GGCGGAGTCG	GT-TCTCACT	CTAGACGT-G	TAAATTC	T ATCTCGCCTA	TCAGTAGGA-	CGCCTCCCTC	GCCGTAAAAC	CCCCCTATAT	TTTAAA	494
SUT142	341	ACCCTTAAGC	CC-CTGTTGC	TTAGCGTTGG	GAGCCTACC-	GTCACA	CGTAGCTCCT	GAAAAGTAGT	GGCGGAGTCG	GT-TCTCACT	CTAGACGTAG	TAAATTC	T ATCTCGC-TA	TCAGTAGGA-	CECCTCCCTC	GCCGTAAAAC	CCCCCTATAT	TTTAAA	495
SUT076	339	ACCOTTAAGC	CC-CTGTTGC	TTAGCGTTGG	GAGCCTACC-	GTCACA	CGTAGCTCCT	GAAAAGTAGT	GGCGGAGTCG	GT-TCTCACT	CTAGACGTAG	TAAATTC	T ATCTCGCCTA	TCAGTAGGA-	CCGGTCCCTC	GCCGTAAAAC	CCCCCTATAT	TTTAAA	494
ST2417	341	ACCCTTAAGC	CC-CTGTTGC	TTAGCGTTGG	GAGCCTACC-	GTCACA	CGTAGCTCCT	GAAAAGTAGT	GGCGGAGTCG	GT-TCTCACT	CTAGACGTAG	TAAATTC	T ATCTCGCCTA	TCAGTAGGA-	CCGGTCCCTC	GCCGTAAAAC	CCCCCTATAT	TTTAAA	496
SUT207	336	ACCOTTAAGC	CT-CTGTTGC	TTAGTGTTGG	GAGCCTACG-	GTCATA	-GTAGCTTCT	CAAA-GTAGT	GGCGGAGCTG	GC-TCACACT	TCAGGCGTAG	TAGATGC	T ATCTCGCTTG	TGAGGTAAG-	CCGGTCCCCA	GCCGTAAAAC	CCCG-TA-AT	TTTAA-	486
AF163033	320	ACCCTTAAGC	CT-CTGTTGC	TTAGCGTTGG	GAGCCTACA-	GCACCTG-	TAGCTCCT	CAAAGTTAGT	GGCGGAGTCG	GT-TCACACT	CTAGACGTAG	TA-ATTT	T ATCTCGCCTA	TTAGTTGGA-	CCGGTCTCCC	GCCGTAAAAC	CCC-TATT	TTTAAA	470
AF163026	336	ACCCTTAAGC	CT-CTGTTGC	TTAGTGTTGG	GAGCCTACG-	GTATA-G-	TAGCTCCT	CAAAGTTAGT	GGCAGAGTCG	GT-TCACACT	CTAGACGTAG	TAGATTT	T ATTTCGCCTA	TTAGTTGGA-	CTGGCCTCTT	GCCATAAAAC	CCCCTAAATT	TTTAAA	489
AF163030	315	ACCCTTAAGC	CT-CTGTTGC	TTAGTGTTAG	GAGCCTACG-	GTACCCG-	TAGCTCCC	CAAAGTTAGC	GGCGGAGTCG	GT-TCACACT	CTAGACGTAG	TA-ATTT	T TTCTCGCCTA	TCAGTTGGA-	CCCCTCCCTT	GCCGTAAAAC	CCCCAATT	TTTAAA	466
AF163039	335	ACCOTTAAGC	CC-CTGTTGC	TTAGTGTTGG	GAGCCTACG-	GTAATAG-	TAGCTCCT	GAAAGTTAGT	GGCGGAGTCG	GT-TCACACT	CTAGACGTAG	TAGATTT	T ATCTCGCCTA	TCAGTTGGA-	CCCCTCCCTT	GCCGTAAAAC	CACCTAA-TT	TCTAAA	488
AF163031	340	ACCCTTAAGC	CT-TTGTTGC	TTAGCGTTGG	GAGCCTACG-	CTTCT-G-	TAGCTCCT	TAAAGTTAGT	GGCGGAGTTA	GTATCACACT	CTAGACGTAG	TA-AATT	T ATCTCGCCTA	T-AGTTGTA-	CTGGTCCCTT	GCCATAAAAC	CCCCTAA-TT	TTTAAA	491
AF163034	335	ACCCTTAAGC	CC-TTGTTGC	TTAGCGTTGG	GAGCCTACG-	CAGAAAGC	GCTAGCTCCT	CAAAACCAGT	GGCGGAGTCG	GT-TCACACT	CTAGACGTAG	TAAATCT	C ATCTCGCCTA	TTAGTTGGA-	CCCCTCCCCT	GCCGTAAAAC	CCCCTAT-TC	TTAAAA	491
SUT123	336	ACCCTTAAGC	CC-CTGTTGC	TTAGCGTTGG	GAGCCTAC	TGCCGG-	TAGCTCCT	CAAAGTTAGT	GGCGGAGTCG	GT-ACGCACC	CTAGACGTAG	TAATTCT	T ATCTCGCCTA	TAGGTCGTG-	CCGGTCCCCT	GCCGTTAAAC	CCCCAATT	TCTAAA	486
ST2027	331	ACCCTTAAGC	CT-CACTCCC	TTAACGTTGG	GACTCTACGA	CCTATTATAG	CGTAGTTCCT	TAAAGTTAGT	GGCGGAGTTA	TAGCCCACT-	CTAAGCGTAG	TA-ATTCTC	T CTCGCTTCTT	GTAGTGGTT-	ATAGTTGCTA	GCCATAAAAC	CCCNCTATTT	T AAT	490
ST2326	348	ACCOTTAAGC	CT-T-GTTGC	TTAGCGTTGG	GAATCAGCGT	CTTTACGG	CGCTGTTCCT	TAAATTTAGT	GGCGGAGTTA	TAGCACACTT	CTAAGCGTAG	TA-AATCTT	T CTCGTTTCTG	G-AGTTGCC-	TTGATTCTTA	GCCGTAAAAC	CCCCCTATTT	TGTAAT	506
SUT090	338	ACCOTTAAGC	CT-T-GTTGC	TTAGCGTTGG	GAGTCTACGG	CTTCGG	CGTAGCTCCT	GAAAGTTAGT	GGCGGAGTTA	GGGTACACT-	CTCAGCGTAG	TA-ACACT-	T CTCGCTCGTG	T-GGTGGCC-	CTGGCTGCTG	GCCGTTAAAC	CCCCATACCT	TTTAGT	492
AY787733	347	ACCCTCAAGC	CC-TAGCTGC	TTGGTATTGG	GAGCTTGT	CTGCGG	-ACAACTCCT	CAAAAGCATT	GGCG-AGTCG	CGGTG-ACC-	CCAAGCGTAG	TA-ATTCT-	T CTCGCTTAGG	TGTGTTAACG	CTGGCGTTCG	GCCACTAA	CCCCCTATTT	TCTAGT	497
SUT201	311	ACCCTTAAGC	CT-TTGTTGC	TTAGCGTTGG	GAGCCTACGG	TAG	CGTAGCTCCT	CAAAATCAGT	GGCGGAGTCG	GTTCACACT-	CTAGACGTAG	TACATTTTA	T CTCGTCTGTG	AGTTGGG-	CTGGTCCCCT	GCCGTAAAA-	CCCCTAATTT	TTAAA-	462
ST2349	311	ACCCTTAAGC	CT-TTGTTGC	TTAGTGTTGG	GAGCCTACGG	TAA	CGTAGCTCCT	TAAAATTAGT	GGCGGAGTCG	GTTCACACT-	CTAGACGTAG	TACATTTTA	T CTCGTCTGTG	AGTTGGG-	CTGGTCCCCT	GCCGTAAAA-	CCCCTAATTT	CTAAA-	462
SUT203	341	ACCOTTAAGC	CC-CTGTTGC	TTAGCGTTGG	GAGCCTAC	AACCCTT	CGTAGCTCCT	CAAAGTTAGT	GGCGGAGTCG	GTTTCACACT	CTAGACGTAG	TAATTTT	T ATCTC-GCCT	ATAGATGAGC	CGGTCCCC-T	GCCGTAAAAC	CCCCT-AATT	TTTAAA	495
SUT177	341	ACCCTTAAGC	CC-CTGTTGC	TTAGCGTTGG	GAGCCTAC	AACCCTC	TGTAGCTCCT	CAAAGTTAGT	GGCGGAGTCG	GTTTCACACT	CTAGACGTAG	TAATTTT	T ATCTC-GCCT	ATAGATGAGC	CGGTCCCC-T	GCCGTAAAAC	CCCCT-AATT	TTTAAA	495
SUT138	341	ACCOTTAAGC	CC-CTGTTGC	TTAGCGTTGG	GAGCCTAC	AACCCTC	TGTAGCTCCT	CAAAGTTAGT	GGCGGAGTCG	GTTTCACACT	CTAGACGTAG	TAATTTT	T ATCTC-GCCT	ATAGATGAGC	CGATCCCC-T	GCCGTAAAAC	CCCCT-AATT	TTTAAA	495
SUT139	342	ACCCTTAAGC	CC-CTGTTGC	TTAGCGTTGG	GAGCCTAC	AACCCTC	TGTAGCTCCT	CAAAGTTAGT	GGCGGAGTCG	GTTTCACACT	CTAGACGTAG	TAATTTT	T ATCTC-GCCT	ATAGATGAGC	CGATCCCC-T	GCCGTAAAAC	CCCCT-AATT	TTTAAA	496
SUT129	339	ACCOTTAAGC	CC-CTGTTGC	TTAGCGTTGG	GAGCCTAC	AACCCTC	TGTAGCTCCT	CAAAGTTAGT	GGCGGAGTCG	GTTTCACACT	CTAGACGTAG	TAATTTT	T ATCTC-GCCT	ATAGATGAGC	CGATCCCC-T	GCCGTAAAAC	CCCCT-AATT	TTTAAA	493
ST2372	342	ACCCTTAAGC	CCTGTTGC	TTAGCGTTGG	GAGCCTAC	AGATACCCTC	TGTAGTTCCT	TAAAGTTAGT	GGCGGAGTCG	GTTTCACACT	CTAGACGTAG	TAAATTT	T ATATC-GCCT	ATAGATGAGC	CGGTCCCT-T	GCCGTAAAAC	CCCCT-AATT	TCTAAA	498
SUT192	344	ACCOTTAAGC	CCTGTTGC	TTAGCGTTGG	GAGCCTAC	AGATACCCTC	TGTAGTTCCT	TAAAGTTAGT	GGCGGAGTCG	GTTTCACACT	CTAGACGTAG	TAAATTT	T ATNTC-GCCT	ATAGATGAGC	CGGTCCCT-T	GCCGTAAAAC	CCCCT-AATT	TCTAAA	500
SUT088	341	ACCOTTAAGC	CC-CTGTTGC	TTAGCGTTGG	GAGCCTAC	AGATACCCTC	TGTAGCTCCC	-AAAGTTAGT	GGCGGAGTCG	GTTTCACACT	CTAGACGTAG	TAATTTT	T ATCTC-GCCT	ATAGATGAGC	CCGGTCCT	GCCGTAAA-C	CCCCAAT-	TCTAA-	492
SUT140	341	ACCOTTAAGO	CC-CTGTTGC	TTAGCGTTGG	GAGCCTAC	AGATACTOTC	TGTAGTTCCT	TAAAGTTAGT	GGCGGAGTCG	GTTTCACACT	CTAGACGTAG	TAAATTT	T ATATC-GCCT	ATAGATGAGC	CGGTCCCCGT	GCCGTAAAAC	CCCCAATA	TCTAAA	498
AF163027	343	ACCCTTAAGC	CC-CTGTTGC	TTAGTGTTGG	GAGCCTACTG	AAGACCCTTC	TGTAGCTCCT	CANAGTTAGT	GGCGGAGTCG	GTTTCACACT	CTAGACGTAG	TAACTTT	T ATCTC-GCCT	ATAGATGCGC	CGGTCCCC-T	GCCGTAAAAC	CCCCT-AATT	TCTAAA	502
ST2348	340	ACCOTTAAGC	CC-CTGTTGC	TTAGCGTTGG	GAGCCTAC	AGCCTTC	TGTAGCTCCC	CAAAGTTAGT	GGCGGAGTCG	GTTT-ACACT	CTAGACGTAG	TAAATTT	T ATCTCAGTCT	GCAGTTAGGC	CGGTCCCT-C	GCCGTAAAAC	CCCCC-AATT	TTTAAA	494
ST2363	339	ACCCTTAAGC	CC-CTGTTGC	TTAGCGTTGG	GAGCCTAC	AGCCTTC	TGTAGCTCCC	CAAAGTTAGT	GGCGGAGTCG	GTTT-ACACT	CTAGACGTAG	TAAATTT	T ATCTC-GTCT	GCAGTTAGGC	CGG-CCCT-C	GCCGTAAAAC	CCCCC-AATT	TTAA	489
AF163037	340	ACCOTTANAC	CC-CTGGTGC	TTAATGTTGG	GAGCCTAC	AGACTTC	TGTAGCTTCC	CARAGTTAGT	GGCAGAGTCG	GTTT-GCACT	CTAGACGTAG	TAATTTC	T ATCTC-GTCT	GCAGTTAGGC	CGGTCCCT-C	GCCGTAAAAC	CCCCCTARTC	TTTAAA	494
AF163040	341	ACCCTTAAGC	CC-CTGTCGC	TTAGTGTTGG	GAGCCTAC	AGTCTTC	TGTAGCTCCC	TAAAGTTAGT	GGCGGAGTCG	GTTCACT	CTAGACGTAG	TAGAT-C	T ATCTC-GTCT	ATAGTTAAGC	CGGTCTCT-T	GCCGTAAAAC	CCCCT-AATT	TCTCAA	491
SUT078	341	ACCOTTAAGO	CC-CTGTTGC	TTACCOTTCG	GAGCCTACA-	GCCCGC	TETAGETCCC	CAAAGATAGT	GCCCCACTCA	GT-TCATACT	CTAGGCGTAG	TAAA-TOTO	T ATCTC-GTCT	ATGGATGCG-	CTGGCGCCTC	GCCGTAAAAC	CCCCCTARTC	TTTTAC	495
SUT028	338	ACCOTTAAGC	CC-CTGTTGC	TTAGCGTTGG	GAGCCTACA-	GCCCGC	TGTAGCTCCC	CAAAGATAGT	GGCNGAGTCA	GT-NCATACT	CTAGGCGTAG	TAAA-TCTG	T ATCTC-NTCT	ATGGATGCG-	CTGGCGCCTC	GCCGTAAAAC	CCCC-TAATC	TTTTAC	492
SUT124	340	ACCOTTAAGO	CC-CTGTTGC	TTACTGTTCG	GAGCCTAC	AGCGAT	G-TAGCTCCT	CAAAGTTAGT	GCCGGAGTCG	GTTACACACT	CTAGGCGCAG	TAAACTOTT	T ATCTC-GTCT	ACCCT-CTCC	CCGGTCCCTT	GCCGTAAAAC	CCCC-CAATT	TCTCAA	494
SUT125	340	ACCCTTAAGC	CC-CTGTTGC	TTAGTGTTGG	GAGCCTAC	AGCGAT	G-TAGCTCCT	CAAAGTTAGT	GGCGGAGTCG	GTTACACACT	CTAGGCGCAG	TAAACTCTT	T ATCTC-GTCT	ACGGTTGTGG	CCGGTCCCTT	GCCGTAAAAC	CCCC-CANTT	TCTCAA	495
SUT074	315	ACCOTCATOO	CC-CTACCCC	GTGGTGTTGG	GAGCCCGCG-	AGGGAC	GGCCGGCCCC	TAAATCTAGT	GGCGGACCCG	TEGTOGECTE	CCCTGCGAAG	TAGTGA	T ATTCC-GCAT	CGGAGAGCGA	CGAG-CCCCT	GCCGTTAAAC	CCCCAACT	TTCCAA	467
SUT027	337	ACCCTTAAGC	CTTCTGTAGC	TTAGCGTTAG	GGGCCTACC-	GTATGGC	GGTAGCCCCT	TAAAATTAGT	GGCGGAGTCG	GTCACACT	CTAGACGTAG	TAATAT	T ATCTCGCCTA	T-AGTTGGAC	CGGTCCT	GCCGTAAA	CCTAA-TT	ATA	481
SUT198	337	ACCOTTAAGO	CTTCTGTTGC	TTACCOTTCC	GEGECTACC-	GTATCCC	GGTAGCCCCT	TAAAATTAGT	GGCGGAGTCG	GTCACACT	CTAGACGTAG	TAATAT	T ATCTCGCCTA	T-AGTTGGAC	CGGTCCT	GCCGTAAA	CCTATT	ATA	480
SUT155	337	ACCOTTANCO	CTTCTGTTGC	TTAGCGTTGG	GGGCCTACC-	GTATGGC	GGTAGCCCCT	TAAAATTAGT	GGCGGAGTCG	GTCACACT	CTAGACGTAG	TAATAT	T ATCTCGCCTA	T-AGTTGGAC	COGTCCT	GCCGTAAA	CCTAA-TT	ATA	481
SUT200	337	ACCOTTANCO	CTTCTCTCTCC	TTACCOTTCG	GGGCCTACC-	GTATCCC	GGTAGCCCCT	TAAAATTAGT	GGCGGAGTCG	GTCACACT	CTAGACGTAG	TABTAT	T ATCTCCCCTA	T-ACTTCCAC	CCCTCCT	GCCGTAAA	CCTAA-TT	ATA	481
ST2298	337	ACCOTTANC	CTTCTGTTGC	TTAGCGTTGG	GGGCCTACC-	GTATGGC	GGTAGCCCCT	TAAAATTAGT	GGCGGAGTCG	GT-TCACACT	CTAGACGTAG	TAAATAT	T ATCTCGCCTA	TTAGTTGGAC	COGTCCCC-T	GCCGTAAAAC	-CCCTABTTT	ATGCAA	493
SUT127	334	ACCOTTAACC	CT-CTGTTGC	TTACTGTTCC	GAGCCTACG-	GTTC	AGTAGCTCCT	CANASTRACT	GCCGGAGTCG	GT-TCACACT	CTAGACGTAG	TAATOTT	T ATCTCGCCTA	TCAGTTGGAC	CGGTCCCCGT	GCCGTAAAAC	-CCCTAATT	CTCAA-	487
SUT195	334	ACCOTTAAGO	CT-CTGTTGC	TTAGTGTTGG	GAGCCTACG-		AGTAGCTCCT	CABAATTAGT	GGCGGAGTCG	GT-TCACACT	CTAGACGTAG	TAATGTT	T ATCTCGCCTA	TCAGTTGGAC	COGTCCCC-T	GCCGTAAAAC	-CCCTAATTT	CTCAA-	486
5177130	334	ACCOTTAAGO	CT-CTGTTGC	TTACTOTTCC	GAGCCTACG-		AGTAGCTCCT	CABASTTAGT	GGCGGAGTCG	GT-TCACACT	CTAGACGTAG	TAATATC	P ATCTOGCCTA	TCAGTTGGAC	CGGTCCCC-T	GCCGTABABAC	-CCCTAATTT	CTCAA-	486
AF163042	344	ACCOTTAAGO	CC-CTGTTGC	TTAGTGTTGG	GAGCCTACG-		CAGCGENECT	CARATGRAGT	GGCGGAGTTG	GT-TCACACT	CTAGAC								427
572382	352	ACCOTTANCO	CT-CTOTTGC	TTACTOTTCC	GAGCCTACG-		CGTAGCTCCT	CABACTCACT	GGCGGAGTCG	GCTCNCAGCT	CTAGACGTAG		* ********	TOTOTOTO-	COGGECCCCT	GCCGTAGAAA	CCCCCCTATT	TTTAAA	506
SUT092	351	ACCOTTAAGO	CC-TTGTTGC	TTAGCGTTGG	GAGCCTACA-	GCGT	TGTAGCTCCT	TAAATTTAGT	GGCGGAACCG	GT-CCGCCTT	CTAGACGTAG	TAAT-TOTT	T ATTTCGCCTA	CA-AGTCGTA	CCGGTCCC-T	GCCGTAAAAA	GCGTTAAGAT		498

Figure 10C. (Continued).

		10	20	30	40	50	0 60	70	80	90	10	0 11	0 12	0 13	10 14	0 15	0 160	0 170	180	3
1000															+++++	11	11			1.22
SUT256	1	GCGAGTTCTC	GATAAAACTC	CGTAACCCTG	TGCGAACCCT	A-CCACTGTT	GCCTCGGCGT		*********	GCCCG	CCCTTGCG		********	********						73
Jul	1	CIGAGITTIT	AACAACTC	CAACCCTT	TGTCGAATCT	A-CCACTGTT	TCCTCGGCGT	ACT	********	GCCGC	GGCCTCTG		********							71
SUT070	1	CIGAGIICIA	CAACT	CCCACCCTC-	TGTGAA-TAT	A-CTACAGTT	GCCTCGGCGA			GG	CGCT									58
SUT237	1	CTGAGTTCTA	TCAACT	CCCACCCTA-	TGTGAA-CAT	A-CTACAGTT	GCCTCGGCGG	*********		GG	TGCT									59
SUT154	1	CTGAGTTCTA	AACT	CCAACCCTA-	TGTGAA-CTT	A-CCACTGTT	GCCTCGGCGC			TG	TGCCTGC		********							60
SUT082	1	CTGAGTTATC	ACAACT	CCAACCCTG-	TOTGAA-CTT	A-CTACTOTT	GCCTCGGCGG			CG	CTGT									59
SUT074	1	CCGAGTT-AC	AACTC	CCAAACCCA-	TOTGAA-CAT	ACCTACTOTT	GCTTCGGCGG	GATGCCCCGG	GCGC	CTCG	TGTGCCCC									78
5117108	1	CCGAGTETAC		CCARACCCC-	TOTOBA-CAT	ACCTATOOTT	0007000000	ATCOCCC	0000		TA-33300									75
0100161	-	ACCACTTANT	TAC	CCARACCCA-	000033-000	ACCTACTOR	0000000000	OTTOTOTOT			TOCCTACCOT	003023-0								109
mm2.60		ACCACITANT.		CONNECCH		heerheiter	CONTRACTOR			1010	CONTRACTOR	COLORA			CONGRATING		CIACCO			105
501260		ACGAGTTAAT	TACAAACT	CCAAACCCA-	COTORA-COT	ACCTACIOIC	GCCTCGGCAG	0100010-		AGAG	-GUCTACCCT	GUADIA-C		TIA	COTGGAGTAG		CTACCC	10		100
512321	1	GCGAGTTAAT	TACAGACT	CCAAACCCA-	COTGAA-COT	ACCTACTOTT	GCCTCGGCAG	OTTOTOCTOT		AGAG	CICCIACCCI	GGAGTA-C		CTAC	CCTGGAGTAG	GTACCCCGGA	GIGGCIACCC	TA		123
AJ390421	1	GCGAGTTAAT	TACAAACT	CCAAACCCA-	TGTGAA-CTT	ACCTGCTGTT	GCCTCGGCAG	GTTGCGCTGC		GGAG	TGCTTACCCT	GGAGTGGC		CTAC	CCTGGAGTAG	CTACCCTGTA	GIGCCIACCC	TGGAGT	AGGCACCC	130
KS15	1	AAGAGTTAT-	AACT	CCARACCCA-	TGTGAA-CAT	ACCTARCOTT	GCCTCGGCGG	GTCG			TACCTACCCT	GTAGTG-CA-		CTTAC	C-TGTAGTGC	CTACCCGGTA	G			98
AJ390411	1	GCGAGTTAAT	TACAAACT	CCAAACCCA-	TGTGAA-CAT	ACCTACTOTT	GCCTCGGCAG	GCCGTGCTGC	GCGG	CTGGAT	COCCTCCCCC	GCGTGGGCAG	********	CTAC	CCTGGAGTAG	CTACCCTG	TAGCG	GGAGCG	GCCCGCCC	137
SUT290	1	ATGAGAGAGT	TAACAAAACT	CCARACCCA-	TGTGAA-CAT	ACCTICIGIT	GCCTCGGCAG	GTCGTGCTGC	GCGG	CCGGAC	CGC-TACCCT	GGAGCAGC		TAC	CCTGGAGTAG	CTACCCTG	GAGTT	GGCGGC	ACCTACCC	135
AF201706	1	GCGAGT	TAAACAAACT	CCAAACCCA-	TOTGAA-CAT	ACCTACTOTT	GCCTCGGCAG	GTCGTGCTGT	GCGG	TOGTGA	TACCTACCCT	GGAGTAGGAA	C	CTAC	CCTGTAGTGA	C	CTACCC	TGGAGTGG	AGCTACCC	132
SUT203	1	AAGAGTTAT-	TACAACTC	CCAAACCCA-	TGTGAACCTT	ACCATTIGTT	GCCTCGGCAG	GTCGCAGCTT	ACCC	TGTGAG	ACCCTACCCT	GTAGG					-GCCTCACCT			100
SUT129	1	AAGAGTT	TACAACTC	CCAAACCCA-	TOTGAACCTT	ACCATTIGTT	GCCTCGGCAG	GTCGCAGCTT	ACCC	TGTGCG	ACCOTACCCT	GTAGG					-GCCTCACCT			98
SUT192	1	AAGAGTTAAA	AACAACTC	CTARACCCA-	TGTGAAACCT	ACC-TTTGTT	GCCTCGGCAG	GTCGCAACTT	ACCC	TGAGGG	GACCTACCCT	GTAGGT					-ACCTTACCC			101
SUT088	1	AAGAGTTA	TACAACTC	CTARACCCA-	TGTGAA-CCT	ACC-TTTGTT	GCCTCGGCAG	GTOGCAGCOT	ACCC	TGTGAG	GGCCTACCCT	GTAGG					-ATCTTACGC			97
572348	1	AAGAGTTAT-	TACBACTC	CCAAACCCA-	TGTGAA-CTT	ACCTTCTGTT	GCCTCGGCAG	GTOGOGACCT	ACCC	TGTGAG	GCCTTACCCT	GTAGG					-GCCCTACTT			9.9
SU7078	1	CAGAGTTTG-	AACGAACT	CCARACCCA-	TGTGAA-CTT	ACCTTCTGTT	GCCTCGGCAG	GGTGC-GCCT	ACCG	TGTGAG	GCCCTACCCT	GTAGG					-GCCCTACGC			98
510028	1	AAGAGTERG-	3303309	CCARACCCA-	TCTC33-CTN		GODTOGODAG	00000-0000	3000		GCCCTRACCCT	CTACC					-600093060			96
0100104	-	AACACER	The-shore	C-111CCC1-		ACCORDO	COCECCOCE	00000-0000	1000		ACCOMACCAC	01100					-000023020			05
3 7300437	÷.	220200000	The	C-AAACCCA-	TOTOMA-CTT	ACCOLACOTT	COORDOCAD	00000000000	ROBROD	1000000100	ACCOMPANY ACCAS	1		00033-00		00010	1000001ACTC	0000		140
10390437	÷.	ANGAGITTOT	TAC-FARCIC	CTARACCCA-	TOTORA-CIT	ACCTICIOTI	GCCTCGGCAG	CONCORCOCA	101100	ACCCTOTAGO	ACCOTATOOT	ATA-GIACCT	TACAG	66640466	TACTA	COGRE	AGACCTACCC	COTA		140
A0390434	÷	ACGAGITTAT	-ATAACTC	CTAAACCCA-	TOTGAA-CTT	ACCTICION	GCCTCGGCAG	GERGATCEGE	ACCTUT	ACCCTGTAGT	ACCTTACCCG	GAACGAACCT	GETATTACAT	AGTAATAGAA	TTCCAAC	COGGATAGAT	AGOCCTACOT	GOTA		133
ST2325	1	AAGAGTTCT-	-ATAACTC	CCAAACCCA-	TGTGAA-CAT	ACCITACOTT	GCCTCGGCAG	GTCGCGCC		ACCTAGTAGC	ACCOTACCCT	GTA					GGGCCTACCC	GG		38
507092	1	CAGAGTTCT-	-GTACTCT	CTCAACCCTG	TOTGAA-CTT	ACTITACOTT	GCCTCGGCAG	GTTGCGCCTA	CTTCGTGGCC	ACTATGTAGC	GGCCTACATT	GTA-GGAGCT	ACCTATTCCT	GGTAGACCTA	CCCTGTATAT	TGTACCTGGT	AGACGTACCT	TGTAGATGGT	GGATCATACC	1/4
ST2382	1	AAGAGTTCT-	-ATGACTC	CCAAACCCA-	TGTGAA-CAT	ACCOTACOTT	GCCTCGGCAG	GTCGCGTCTA	CCC	TGTAGC	ACCCTACCCT	GTA-AGACC-	********	CTA	CCCTGTAGGA	CTTACCCGGT	AGGACCT	*********	ACC	124
ST2310	1	AAGAGTTT	-ATAACTC	CCAAACCCCA	TGTGAA-CAT	ACCTAACGTT	GCCTCGGCGG	GTCGTACCTA	CCC	TGTAGT	GCACTTACCT	GTA					GIGCCIACCC			97
SUT127	1	AAGAGTTCT-	-ATA-CTC	CCAAACCCA-	TGTGAA-CAT	ACCGTATGTT	GCCTCGGCAG	GTCGTGTCTA	CCC	TGTGGT	GCCTTACCCT	GTA					GGGCCTACCT			95
SUT123	1	AAGAGTGT	-ATA-CTC	CCAAACCCA-	TOTGAA-CAT	ACCTTACGTT	GCCTCGGCAG	GTCGCGCCTA	CCC	COTAGA	GCCCTACACT	TTA	********				GGCGCTACCC			95
SUT051	1	TAGAGTTTC-	-CAA-CTC	C-AAACCCA-	TOTGAA-CAT	ACCAGACOTT	GCCTCGGCAG	GCCGCGTGCC	AACC	TCTCTC	AGGG-GCGGC	GCG					-GCAAGGCCT			94
SUT056	1	TAGAGTTTC-	-CAA-CTC	C-ARACCCA-	TGTGAA-CAT	ACCAGACOTT	GCCTCGGCAG	GCCGCGTGCC	AACC	TCTCTC	AGGG-GCGGC	GCG				C	-GCAAGGCCT			94
AY541610	1	GAGAGTCTAA	ACAA-CTC	CCAAACCCAC	TOTGAA-CAT	ACCCCACOTT	GCCTCGGCAG	GC	-GCC	CGGCTC	CGCG-ACCCC	TCG					GGGCT			84
SUT258	1	AAGAGTTCTT	ACAAACTC	CCAAACCCC-	TOTGAA-CAT	ACCTITIGTT	GCCTCGGCAG	GCCCTGGCCT	ACCC	CGTAGT	GGCG-CCCCT	ACG				C	TOTAGGGCCC			99
AJ390436	1	CAGAGTTATC	AAAA-CTC	CCARACCCA-	TGTGAA-CAT	ACCTOGCOTT	GCCTCGGCAG	GTGGCGTCTC	GCCC	CGTAAG	AACCTACCCT	GTA				G	GAACGTACCC			99
SUT201	1	AAGAGTTTAT	-TAA-CTC	CCAAACCCA-	TGTGAA-CAT	ACCATACOTT	GCCTCGGCAG	GCTAC	-ATC	TGCTCT	ATTA-GAGCO	ATG					7			81
SUT207	1	AAGAGTTCTA	-TAA-CTC	CCAAACCCA-	TGTGAA-CAT	ACCTTACGTT	GCCTCGGCAG	GTOGTGC-CN	ACCC	CGTAGC	GCCCTACCCT	GTA				G	-GACCTACCC			96
SUT032	1	CAGAGTTCT-	-ATTACTC	CCARACCCA-	TGTGCA-CAT	ACCOTACOTT	GCCTCGGCAG	GCGGCGCCTA	CCC	CGTAGC	GCCCTACACC	CGG				TA	GGGCCTGCCC			98
SUT090	1	CTGAGTTATC	TAAACTC	CAAA-CCCTT	GGTGAAC-TT	A-CCGTCGTT	TCCTCGGCGT	-GTTGTGGGG	GTA		TAGCTACC						TACCC	GGTGGC		92
SUT233	1	CTGAGTTATC	TAAACTC	CAAA-CCCTT	TOTGAAC-TT	A-CCGTCGTT	TCCTCGGCGT	-GTGCTGCGG	CTA		TAGCTACC						TACCC	GGTAGC	TA	92
SU7066	1	CTGAGT-ATA	AAAACTC	TAAAACCOTT	TOTGAACCTT	A-CCGCCGTT	accreacer	-000000000	C73		TAGCTACCCT	GTAGGTGCCT				CCTGTAG	GTGCCTACTC	000000	76	119
\$117063	÷	ANGAGETATT		CAAA-CCCTT	TOTOLACOTT	A-COGTOGOT	OCCTCGGAGT	-GAGCTACCC	C73		TAGOTACOOT	GRAGCACCCT		ACCTACCCTO	TAGTACCOTA	CAGCERCOCE	ATAGOTACCT	TGALGO		142
010000	-	CREACERATE		CININ COULT		1-000000	concocco	-010000000	C#1		THOUTHOUSE	CERCC-		nouncert	- INGINEGUIN					00
DUILLI	÷	CIGNOTIATO	CAAACIC	CAMAACCOTT	TOTORACCTT	A-CCOICOIT	0001000001	-040010000	CIA		INCOMPOSIT	UINUC				-			IA	30
Juz	÷	COADTICATE	AAAAACTC	CAAAACCCCTT	TOTGAACCTT	A-CCGCAGTT	GCCTCGGCGT	-600006066	000		GCCTG-CTGC	AGGCCAAC	66	CCCCCCGAAA	0000000000	T	GGGG ACCG	GCAGGC		131
SUTZZU	÷	CAGAGTIGIC	GGRAAAACTC	CATAACCOTT	TOTGAACCTA	C-CTATCOTT	GCCTCGGCGC	-CCGCTGCGG	CTGACG	TCCGG	AAGAG-CTGC	TOCCOUTCOT		GGCCCTGGAM	TICCGGGGGGGG		GGCTTTCTT	000000	TT	130
507001	1	CTGAGT-ATC	A-AAAACTTC	CAAAACCCTT	TOTGAACCTA	CCGCCGTT	GCCTCGGCGC	-GCGCTGCGG	CTA	CCCGC	CCCGGACAGA	AGGGCAGC		TOCCTOTORO	GGCCGCTGTA	A	ACCOTTCCOT	CCAGGT	AC	130
SUT218	1	TGAGTTGACT	ATAAAAACTC	C-CAACCCTA	TGTGAACCTT	TGTTCTCGTT	GCCTCGGCAT	-GCGCCAGGG	GGG	CGG	CGTACCACGT	ACCCCGCG		-ACCTATAAA	CGGCAGGGG-		TCTT	GGGCGC	CC	122
SUT223	1	TGAGTTGACC	ATAAAAACTC	C-CAACCCTA	TGTGAACCTT	TGTTCTCGTT	GCCTTGGCAT	-GCGCCAGGG	GGG	CGG	COTACCACOT	ACCCCGGG		-ACCTATAAA	CGGTAGGGG-		TCTT	GGGCGC	cc	122
SUT016	1	CAGAGTTACC	AAAACTC	C-CAACCOTT	TGTGTACCTA	CTACCATT	GCTTCGGCGG	GCTGCGG	C72	CCCTG	CAGCTACCCT	GTAATT		CAGO	GTGGGC		TTGCCT	GGTAGC	TC	109
SUT282	1	CAGAGTTACT		C-CARCCCTA	TOTGAACTTA	CCGTCGTT	GCTTCGGCGG	GCTGCGG	CTA	CCCTG	TACCTACCCT	GTACCTAC		CCTGTAGO	accesseses		GTTTCCT	GGTAGC	TT	119
SUT166	1	CAGAGTTACT	ATAAAACT	CCCAACCCTT	TGTGAACCTT	A-CCGTCGTT	GCCTCGGTGG	ALOGICGICT	GCG	01000	AAGCTACCCT	GGAGTTAC		ACCCTGTAGA	TAGCTACCCT	GG	AGCTACCC	TGAAAA	TA	133
SUT158	1	CAGAGTTAAT	CCAAA-CT	CCCAACCCTT	TGTGAACCAT	A-TCGTCGTT	GCCTCGGGCG	TGA	GCG	GCGG-			CC	ACCCAGAGAG	TAGCTACCCT	GG	AGCTGC		TA	102
SUT294	1	CTGAGTTCTA	CAAAA-CT	CCCAACCCTA	TGTGAATCTT	A-CCACTGTT	GCCTCGGCGC	TGAGCGGCAG	CTA	CCCGG	GAGCTACCCT	GGAGGGAC		ACCCTGTAGA	TEGCTACCCT	GG	AGCTACCC	TGGAGT	TG	132
SUT182	1	CTGAGTTCTA	CAAAAACT	CCCAACCOTT	TGTGAACCCT	A-CTTCAGTT	GCCTCGGCGC	TGAGCGGTAG	CTA	CCCGG	GAA	GAG		ACCCTGGAGA	AATCCGGGGGC	G	ACCTACCC	TGTAGT	TA	120
SUT148	1	CAGAGTTATT	AAAAACTC	CCAAACCOTT	TGTGAACGTT	A-CIGITGIT	GCCTCGGCGT	-GAGCGAGGG	CTA	CCCGG	GAG			ACCCTGTAG-	CTACCCT	GT		GTTG	TA	108
SUT187	1	CAGAGTTATT	A AAAACTC	CCAAACCCTT	TGTGAACGTT	A-CTGTTGTT	GCCTCGGCGT	-GAGCGAGGG	CTA	CCCGG	GAG		CT	ACCCTGTAG-	CTACCCT	GT		GTTG	TA	108
SUT116	1	CTGAGTTATC	CAAAACT	CCCAACCCTT	TOTGAAC-TT	A-CCACTOTT	accreace-	GTTGTG-	CTAC		TAGCTACCCT	GTAG					CTACCC	GGGAAC	A	95
SUT103	1	CCGAGTTANA	CAAAACT	CCAMACCOTT	TOTGAACCTT	A-CCANAGET	GCCTCGGCGT		77AC	0076	TAGTTACCCT	GGAGGCGTCT	A			CCCTGTAG	GTOCTTACCC	TGGAGCT	h	119
51177120	-	CAGAGETATA	CT	CCARACCOTA	TOTOLLOTT	A-CCACTOT	accessoco-				TTOCTACCOT	CTACC						TOGACOT		20
AF616682	-	CCGAGTTATC	Terrestance	CCAACCOURT	TOTGALLO	A-CCGTCCTT	000000000	OCTOCOCTON.			TAGOTACCO	GTAG					CTACCC			89
3.8616691		COCO COMPTONIC	Real Street	00110000	TOTOL STATE	3-00000011	00000000000	COROCOTTA	000			CONC					CERCEC			0.0
A2 010001	-	COMOTIATC	AAACT	CCAACCOTT-	TOTGAAACTT	A-CCOTCOTT	GCCTCGGCGG	SCIGCOCITA	000	<u>7</u> G	INCOMPCCO I	0110					CTACCC	-		83
SUTIOBD	1	CIGAGITATC	TAAACT	CCAACCCTA-	TOTGAA-CTT	A-CCGCCGTT	GCCTCGGCGG	GCCGCGTTCG	CCCT	G7	AUTTACT						ACCT			82
SUTUESD	1	CTGAGTTATC	TAAACT	CCAACCCTA-	TOTGAA-CTT	A-CCGCCGTT	GCCTCGGCGG	GCCGCGTTCG	CCCT	67	AGTTTACT						ACCT	G		52
A1010084	4	TIGAGTTATC	TAAACT	CCAACCCTA-	TOTGAA-CTT	A-COGCOGTT	GUCTCGGCGG	UCCGCGT1CG	CUCT	GT	AUTTACT						ACCT	G		82
SUT164	1	CAGAGTTATC	CYYCLC	CCAACCCTT-	TOTGAACCTT	A-CCGCAGTT	eccacecec	GA-GCCGCGG	CIAC	CCTG	CAGCTACCCT	GING					CIOCC-			90
ST2584		CEGAGECCCC	Ca	CCARCCOTT-	TGTG33-CC7	3-CCACOPPT	CCTCCCCCCCCCC	ABAGCCCTAG	CCTAAC	CTROCCEGO	GCGCCCAG	3003033700					-003303097	ATATOCALCO	CTACTACCTA	1.30

		19	200	210	22	0 23	24	250	260	27	280	29	0 30	310	320	330	34	0 350	360	5
aumore.	24													¥						222
SUT256	73		GG	ACGCCAGTGG	CCCATGAACT	GGACT	CT	GTTTTAGCTG	CTCACTGCAG	CCACTGTGAA	TATCTGACGG	CCTTAACTGA	AATACGTTAA	AACTITCAAC	AACGGATCTC	TIGGTTCIGG	CATCGATGAA	GAACGCAGCG	AAATGCGATA	222
SUT070	50			TOCOCTOCADO	accactates	GENCE		AC-TOTOTT	TOTOCOCKO	TATOTOTOAL	CTCATARCAR	GAGTAACAA	ANTACOTTAN	AACTITCAC	AACGGATCTC	TROUTEROG	CATCONTONN	GAACGCAGCG	BARTOCCATA	101
SUT237	59		CC	CCGCCGGCGG	ACCACTAA			AC-TOTOTT	TA-TCCACTG	TATCTCTGAA	TT	-TAATAACGA	AATACGTTAA	AACTTTCA	AACGGATCTC	TIGGTICIGG	CATCGATGAA	GAACGCAGCG	AAATGCGATA	191
SUT154	60	GAGAGCAG	GC	CCGCCGGTGG	ACCACTAA			AC-TCTGTTA	TA-CCTACTG	TATCTCTGAA	TT	TATAACTGA	AATACGTTAA	AACTITCAAC	AACGGATCTC	TTGGTTCTGG	CATCGATGAA	GAACGCAGCG	AAATGCGATA	197
SUT082	59	GCGAGCA-	GC	CCGCCGGCGG	ACCACTAR			AC-TCTGTTT	TTACAG	CATCTCTGAA		-GATAACTTA	AATA-GTTAA	AACTITCAAC	AACGGATCTC	TIGGTICIGG	CATCGATGAA	GAACGCAGCG	AAATGCGATA	190
SUT074	78	-GGATCAGGC	GC	CCGCCTAGGA	ACTTG			ACTCTTGTTT	TATTTTG	AACTTCTGAG	TA	-GTTTTTACA	AATAAATAAA	AACTITCAAC	AACGGATCTC	TTGGTTCTGG	CATCGATGAA	GAACGCAGCG	AAATGCGATA	213
SUT108	75	-GGACGGCCC	GC	CCGAGGA	CCCCT		A	AACTCTGTTT	TTAGTG	GACTTCTGAG	TA	-AAACAAACA	AATAAATCAA	AACTITCAAC	AACGGATCTC	TIGGTICIGG	CATCGATGAA	GAACGCAGCA	AAATGCGATA	206
SUT161	109	CAGCGCGC	AACCAGAC	CTGCCGAAGG	ACACTTAA	********	********	AT-TCTTTTT	TAC-CCCG	GAACTCTGAA	ACT	ATTATATA	AATAAGTTAA	AACTITCAAC	AACGGATCTC	TIGGTICIGG	CATCGATGAA	GAACGCAGCG	AAATGCGATA	250
SU1260	123	CAGCGCGC	AACCAGAC	CTOCCGAAGG	ACACTTAA			AT-TOTTTT	TAC-CCCG	GAACTCTGAA	AC	ATTATATA	AATAAGTTAA	AACTITCAAC	AACGGATCTC	TIGGTICIGG	CATCGATGAA	GAACGCAGCG	AAATGCGATA	264
AJ390421	137	CGCAGCGCGC	AACCAGAC	CTGCCAGAGG	ACCTCTGA			AC-TCTTTTT	TAC-ACTG	GAACTCTGAA	ACT	ATTATACA	AACAAGTTAA	AACTITCAAC	AACGGATCTC	TIGGTICIGG	CATCGATGAA	GAACGCAGCG	AAATGCGATA	279
K\$15	98	GC	ACGGGTAAGC	CCGCCGGCGC	CCCATTAA			AC-TCTGTTT		GATATCTGAA	TT	ATAACTA	AATAAGTTAA	AACTITCAAC	AACGGATCTC	TTGGTTCTGG	CATCGATGAA	GAACGCAGCG	AAATGCGATA	234
AJ390411	138	CGCGGCGCGC	CAACAGGC	CTGCCGAAGG	ACCCCTAA			AC-TCTGTTT	CGCACCTG	TATCTCTGAG	TTT	ATTATACA	AATAAGTTAA	AACTITCAAC	AACGGATCTC	TTGGTTCTGG	CATCGATGAA	GAACGCAGCG	AAATGCGATA	281
SUT290	136	TGCAGCATAC	AACCAGGC	CTGCCGGAGT	GCTCCTAA			AC-TOTOTTT	TACACCTG	TATCTCTGAG	TCT	ATTATACA	ANTANGTTAN	ARCTITCARC	AACGGATCTC	TTGGTTCTGG	CATCGATGAA	GAACGCAGCG	ARATGCGATA	279
AF201706	133	TGCAGCACGC	AACCAGGC	CTGCCAAAGG	ACCCCTAA			AC-TCTGTTA	T	TACCTCTGAG	ΤСλ	ACTATACA	AATAAGTTAA	AACTITCAAC	AACGGATCTC	TIGGTICIGG	CATCGATGAA	GAACGCAGCG	AAATGCGATA	273
SUT203	100	-GGTAGTTGC	GGGTAATC	CTGCCGGTGG	TCTA-CCA		A	AC-TCTGTT-	T-ACTATG	TTATTCTGAA	TA	-TTATAACTA	AATAAGTTAA	AACTITCAAC	AACGGATCTC	TIGGTICIGG	CATCGATGAA	GAACGCAGCG	AAATGCGATA	241
507129	101	-GGTAGTTGC	CCCCAR-ARC	CTGCCGGTGG	TOTA-TCA			AC-TOTOTT-	ACTATO	TATTCTGAA		-TTATAACTA	AATAAGTTAA	AACTTTCAAC	AACGGATCTC	TIGGTTCTGG	CATCGATGAA	GARCOCAGOG	AAATGCGATA	239
SUTOBB	97	-GGTAGTTGC	AGGTTTCAAC	CTGCCGCTGG	TCTA-CCA			AC-TOTOTT-	TTACCATG	TTATTCTGAA	TA	-ATATAACTA	AATAAGTTAA	AACTITCAAC	AACGGATCTC	TTGGTTCTGG	CATCGATGAA	GAACGCAGCG	AAATGCGATA	241
572348	99	-GGTAGTCGC	GGGTACGC	CTGCCGGTGG	CCCA-TGA			AC-TCTGTT-	TATTCTTG	TTATTCTGAA		-CTATAACTA	AATA-GTTAA	AACTTTCAAC	AACGGATCTC	TIGGTICIGG	CATCGATGAA	GAACGCAGCG	ARATGCGATA	239
SUT078	98	-GGTGCGTGC	GGGCAGCC	CTGCCGGCGG	CCCG-TGA		A	AT-TOTOTT-	TGACTACG	TTATTCTGAA		-ACATAACAA	ANTAGTTAAA	AACTITCAAC	AACGGATCTC	TTGGTTCTGG	CATCGATGAA	GAACGCAGCG	AAATGCGATA	239
SUT028	96	-GGTGCGTGC	GGGCAGCC	CTGCCGGCGG	CCCGGA		A	AT-TCTGTT-	TGACTACG	TTATTCTGAA	T	-ACATAACAA	AATAGTTAAA	AACTITCAAC	AACGGATCTC	TIGGTICIGG	CATCGATGAA	GAACGCAGCG	AAATGCGATA	236
SUT124	95	-GGTGGCCGC	GGACTA-AGC	CTOTCGGTGG	CCCAACCA		X	AC-TCTGTCA	G-TGATTOTG	TC-TTCTGAA	CT	-TGACAACGA	ANTACGTTAN	AACTITCAAC	AACGGATCTC	TIGGTICIGG	CATCGATGAA	GAACGCAGCG	AAATGCGATA	240
AJ390437	140	-GAGAGATGC	GGTGCCAC	CTGCCGGTGG	ACTGAAAAAC		AA	AT-TCTGTTT	AGTAAATG	GTATTCTGAA	TC	ATTATATGGA	AATAAGTTAA	AACTITCAAC	AACGGATCTC	TIGGTICIGG	CATCGATGAA	GAACGCAGCG	AAATGCGATA	288
AJ390434	153	-ATAAAGTGC	GCTCCCGC	CTGCCGGTGG	ACCATGAAAT			AC-TCTGTTT	AATAGCTG	GTATTCTGA-	TA	ATTATATGGA	AATAAGTTAA	AACTITCAAC	AACGGATCTC	TIGGTICIGG	CATCGATGAA	GAACGCAGCG	AAATGCGATA	300
512525	175	TOGTAGACOC	GGGT33GC	CTOCCOGCOG	CCCATTARC-			AC-TOTOTT		TOTTTCTGAG		A-TACAACCA	ANTIATTAN	AACTITCAAC	ARCOGATOTO	TROUTEROG	CATCGATGAA	GAACGCAGCG	ANATOCONTA	317
ST2382	125	CGGCAGACGN	GGGTAAGC	CTGCCGGCGG	CCCACGCAA-			AC-TCTGTTT	TGGCAATG	TAATTCTGAA	T	ACTATAACTA	AATAGTTAAA	A-CTTTCAAC	AACGGATCTC	TIGGTICIGG	CATCGATGAA	GAACGCAGCG	AAATGCGATA	268
ST2310	97	-GGTAGGCAC	GGGTAAGC	ccoccoccoc	CCCATTAA			AC-TCTGTTT	AATTACTG	GATATCTGAA		TATAACTA	AATAAGTTAA	AACTITCAAC	AACGGATCTC	TIGGTICIGG	CATCGATGAA	GAACGCAGCG	AAATGCGATA	238
SUT127	95	-GGTAGATCC	GGATAGC	CTGCCGACGG	CCCCTCAA			AC-TCTGTTT	AATAG-TG	AATCTCTGAA	C	TATAACTA	AATAAGTTAA	AACTITCAAC	AACGGATCTC	TIGGTICIGG	CATCGATGAA	GAACGCAGCG	AAATGCGATA	234
SUT123	95	-GGTGGACGC	GGGCAAGC	ccecceecee	CTCACTAA			AC-TCTGTTT	AGCATTG	TACTTCTGAA	C	алтаталста	AATA-GTTAA	AACTITCAAC	AACGGATCTC	TIGGTICIGG	CATCGATGAA	GAACGCAGCG	AAATGCGATA	236
SUT051	94	-GCCGGCCGT	ACC	TCCCCGGCGT	CTCGCTGGTG	GGGCCGGCCC	CTGGACGGAG	GCGTCCGCCT	TA-ATTCTTG	AA-TACTGTT	GAATTCTAAA	ATCATAACTA	AATTAGTTAA	AACTITCAAC	AACGGATCTC	TIGGTTCIGG	CATCGATGAA	GAACGCAGCG	AAATGCGATA	264
SUT056	94	-GCCGGCCGT	ACC	TCCCCGGCGT	CTCGCTGGTG	GGGCCGGCCC	CTGGACGGAG	GCGTCCGCCT	TA-ATTCTTG	AA-TACTGTT	GAATTCTAAA	ATCATAACTA	AATTAGGTAA	AACTITCAAC	AACGGATCTC	TTGGTTCTGG	CATCGATGAA	GAACGCAGCG	AAATGCAATA	264
A1541610	00	-00000010-	GCC	CIGCCGGCGG	CCCAC	CCCCCCLC	GAA	AC-TOTOTOT	AGCACIG	AA-TICIGAG	CCCGAGAGGGG	COTATAACAA	AATTAGTTAA	AACTITCAAC	AACGGATCTC	TIGGTICIGG	CATCGATGAA	GAACGCAGCG	AAATGCCATA	246
AJ390436	99	-GGTAGACG-	ACC	CTGCCGACGG	CCCCC		GRA	AC-TCTGTTT	TATAGCATTA	AACTTCTG		AACATAACTA	AATAAGTTAA	AACTITCAAC	AACGGATCTC	TIGGTTCTGG	CATCGATGAA	GAACGCAGCG	AAATGCGATA	237
SUT201	81	-GTAACCGG-	C	CTGCCGGCGG	TCCATT		A	AC-TCTGTTT	ATTTTTG	AA-TTCTG	AG	GTTATA-TAA	AATAAGTTAA	AACTTTCAAC	AACGGATCTC	TIGGTICIGG	CATCGATGAA	GAACGCAGCG	AAATGCGATA	211
SUT207	96	-GGTAGACGC	GGGTAAGC	TTGCCGGCGG	CCCAC		GAA	AC-TCTGTTT	AGTATTG	AA-TTCTG	AA	CCTGTAACAA	AATAAGTTAA	AACTTTCAAC	AACGGATCTC	TIGGTICIGG	CATCGATGAA	GAACGCAGCG	ANATGCGATA	236
SUT032	98	-GGTGGACGC	CGACAA-AGC	CTGCCGGCGG	CCCCTGAA			AATTCTGTTT	CTTACTG	GATCTCTGAA	CA	TATACCAA	AATACGTTAA	AACTITCAAC	AACGGATCTC	TTGGTTCTGG	CATCGATGAA	GAACGCAGCG	AAATGCGATA	241
SUT090	93	CCCTGTATNT	GGCCNACGGT	CCGNCCCATG	ACCGGTAAAC	T	T	TGTTTTT	ACCACTG	TATGTCTGAA	TT	GAААСТА	AATAAGTTAA	AACTITCAAC	AACGGATCTC	TIGGTICIGG	CATCGATGAA	GAACGCAGCG	AAATGCGATA	237
SUT233	93	CCCTGTAGCT	GGCCCACGGC	CCGCCGCAGG	ACCGCTAAAC	T	T	TGTTTTT	ACCACTG	TATCTCTGAA	TT	GTTAACTA	AATAAGTTAA	AACTITCAAC	AACAGATCTC	TTGGTTCTGG	CATCGATGAA	GAACGCAGCG	AAATGCGATA	238
SUTUES	142	COORCEAGEC	GGACCACGGC	CCCCCCGGAGG	ACTOCTAAAC		T	monome ma	ACCACTG	CENTERCECAN		ATTAACTA	AATACGTTAA	AACTITCAAC	AACGGATCTC	TREATERS	CATCGATGAA	GAACGCAGCG	AAATGCGATA	287
SUT221	61	CCCTGTAGCC	GGTTCACGGC	CCGCCGAAGG	ACAGCTABAC			TOTTAATT	ACCACTG	TATCTCTGAA		GTCBACTA	AATAAGTTAA	BACTTTCARC	AACGGATCTC	TTGGTTCTGG	CATCGATGAA	GAACGCAGCG	ARATGCGATA	237
Ju2	132	CGGCCGGC-A	AAACGGC	CCGGCAAAGG	ACCCGCAAAC	T	A	ACTCTARATT	AACAACGG	TACTTCTGAA	AA	CTTCAAACTT	AATGAGGTAA	AACTTTCAAC	AACGGATCTC	TTGGTTCTGG	CATCGATGAA	GAACGCAGCG	AAATGCGATA	279
SUT220	139	TAGCCGGCTA	AAACAGC	ACGCCAGAGG	ACCCACAAAC		A	ACTOTAATTT	AACGTCG-	TACTTCTGAA	AA	ATATCAACTA	ATTATTAA	AACTTTCAAC	AACGGATCTC	TTGGCTCTGG	CATCGATGAA	GAACGCAGCG	AAATGCGATA	283
SUT001	131	CGGCTAGCCG	GCTGAACAGC	CCGCCGAAGG	ACCOCTATAA	A	A	ATCTCGCTCC	GGCGTGTA	TACTTCTGAA	T	CTTCCAACTT	AATGAGTTAA	AACTTTCAAC	AACGGATCTC	TIGGTICIGG	CATCGATGAA	GAACGCAGCG	AAATGCGATA	281
SUT218	123	CGCCCGCA	CCCGGC	CICCCCCCCCCCC	ACCAACC	C		ACTCTTG-CA	AATATTOT	GGACTCTGAA	AT	ATA-AAAATA	AACGAATCAA	AACTITCAAC	AACGGATCTC	TIGGTICIGG	CATCGATGAA	GAACGCAGCG	AAATGCGATA	263
SUT223	123	CGCCCGCA	TCCGGC	CTGCCGGTGG	ACCAACC	C		ACTCTTG-CA	AATCTTGT	GAATTCTGAA	λ1	ΑΤΑ-ΑΑΑΑΤΑ	AACGAATCAA	AACTITCAAC	AACGGATCTC	TIGGTICIGG	CATCGATGAA	GAACGCAGCG	AAATGCGATA	263
SUTU16	120	GCGCGAA	GGC	CCGTCAGAGG	ACCATTAAAC			PTGTTACCC-	TOTACG	TCTATCTGAA	TGanana	CTT-CAACTT	AATAAGTTAA	AACTTTCAAC	AACGGATCTC	TIGGTICIGG	CATCGATGAA	GAACGCAGCG	ARATGCGATA	247
SUT166	134	CGCCCCCC	GCCAG	CCGCCGAAGG	ACTACTABAC			TTGTTTT-	ACTG-TG	TCTCTCTGAA		ATGAAACA-A	ATTC-GTTAR	AACTITCAAC	AACGGATCTC	TTGGTTCTGG	CATCGATGAA	GAACGCAGCG	ABATGCGATA	271
SUT158	103	CGCC		-CGCCGGAGG	ACCACTARAC	T	c	TTGTTTTT	ACCA-TG	TATTTCTGAA	TG	CTTCAACT-A	AATA-GTTAA	AACTITCAAC	AACGGATCTC	TIGGTICIGG	CATCGATGAA	GAACGCAGCG	AAATGCGATA	231
SUT294	133	CGCCTAAC	GCT	CCGCTGGCGG	ACCACCARAC	T	C	TOTTTT	ACAAGTG	TATCTCTGAG	TA	TATAACCA-A	AATACGTTAA	AACTTTCAAC	AACGGATCTC	TIGGTICIGG	CATCGATTAA	GAACGCAGCC	AAATGCGATA	269
SUT182	121	CACCTAAC	GCT	CCGCCGGTGG	ACCACTAAAC	T	C	TGTTTTTA	ACCACTG	TATCTCTGAA	AT	ACTTAACG-A	AATACGTTAA	AACTITCAAC	AACGGATCTC	TIGGTICIGG	CATCGATGAA	GAACGCAGCG	AAATGCGATA	259
SUT148	109	AGCC		-CGCCGGAGG	ACCACTARAC		C	TEGTTTATTT	ACTG-TG	TATCTCTGAA	TG	CTTCAACT-A	AATCAGTTAA	AACTITCAAC	AACGGATCTC	TIGGTICIGG	CATCGATGAA	GAACGCAGCG	AAATGCGATA	239
SUT187	109	AGCC		-CGCCGGAGG	ACCACTAAAC		C	TOGTTTATTT	ACTG-TG	TATCTCTGAA	1G	CITCAACIGA	AATCAGTTAA	AACTITCAAC	AACGGATCTC	TIGGTICIGG	CATCGATGAA	GAACGCAGCG	AAATGCGATA	241
SUT116	120	CATTCCAA	GCCTTA ACCCCC	TEGCCAGAGG	ACCTACCA			ACCTGTTTTA	T-ACTG	TATCTCTGAA	C	-ITATAACTA	AATAAGTTAA	AACTITCAAC	AACGGATCTC	TIGGTTCTGG	CATCGATGAA	GAACGCAGCG	AAATGCGATA	229
SUT120	96	CCCTGTAGCA	CGCACACOGC	CCGCCAAAGG	ACCACTAA			ACTOTTATT	T-TTACTOTO	AATATCTCAA	TG	-CTTCAAAAT	AATAAGTTAA	AACTITCAAC	AACGGATCTC	TTGGTTCTGG	CATCGATGAA	GAACGCAGCG	AAATGCGATA	243
AF616682	89	-GGTAGGCGC	GCTCCA-AGC	CCGCCGGTGG	ACCACTAA			AC-TCTGTTT	TAATACCG	AATCTCTGAA	TG	-CTTCAACTT	ANTANGTTAN	AACTITCAAC	AACGGATCTC	TIGGTICIGG	CATCGATGAA	GAACGCAGCG	AAATGCGATA	233
AF616681	89	-GGTAGGCGC	GCTCCA-AGC	CCCCCCGTCG	ACCACTAA			AC-TOTOTT	TAATACCG	AATCTCTGAA	TG	-CTTCAACTT	AGTAAGTTAA	AACTTTCAAC	AACGGATCTC	TTGGTTCTGG	CATCGATGAA	GAACGCAGCG	AAATGCGATA	233
SUT168D	82	-GCGGCGCGC	TACAGGC	CCGCCGGTGG	ACTGCTAA			AC-TCTGTTA	TATATACG	TATCTCTGAA	TG	-CTTCAACTT	AATAAGTTAA	AACTITCAAC	AACGGATCTC	TIGGTICIGG	CATCGATGAA	GAACGCAGCG	AAATGCGATA	224
SUT085D	82	-GCGGCGCGC	TACAGGC	CCGCCGGTGG	ACTGCTAA			AC-TCTGTTA	TATATACG	TATCTCTGAA	TG	-CTTCAACTT	AATAAGTTAA	AACTITCAAC	ARCOGATCTC	TTGGTTCTGG	CATCGATGAA	GAACGCAGCG	ARATGCGATA	224
AY616684	82	-GCGGCGCGC	TACAGGC	CCGCCGGTGG	ACTGCTAA		*********	AC-TCTGTTA	TATATACG	TATCTCTGAA	TG	-CTTCAACTT	AATAAGTTAA	AACTITCAAC	AACGGATCTC	TTGGTTCTGG	CATCGATGAA	GAACGCAGCG	AAATGCGATA	224
572584	131	GGACAAGCAA	CGATCGAGGC	CTOCCOUTGG	ABATCTTAGG	COPPERCENTA	GCGACCAGTA	GOOTCTGATG	GOGTTABACC	TACTOTOTAL	10	TAAATAA	ATCANTTTA	BACTTTCAAC	AACCGATCTC	TREETERS	CATCGATGAA	GRACECAGCG	ANATOCOATA	300

Figure 11C. (Continued).

316

		37	0 380	390	400	410	0 42	430	440	45	0 46	0 47	0 48	0 490	50	51	0 52	0 530	540	0
CIIII DE C	222	ACTINATION				TCARCORACE.						Chemphone							Nacio Concesso	200
Jul	216	AGTAATOTGA	ATTGCAGAAT	TCAGTGAATC	ATCGAATCTT	TGRACGCACA	TTGCGCCCAT	TAGTATTCTA	GTGGGCATGC	CTATTCGAGC	GTCATTTCGA	CCCTTAGGCC	CC-AGTTGCT	TAGCOTTOOR	AGTCTGCGGC	CCAGGCC	GCAGT	TCCTCAAAGT	CAGTOGCOGO	386
SUT070	192	AGTAATGTGA	ATTGCAGAAT	TCAGTGAATC	ATCGAATCTT	TGAACGCACA	TTGCGCCCAT	TAGTATTCTA	GTGGGCATGC	CTATTCGAGC	GTCATTTCAA	CCCTTACGCC	CTGTAGCG	TAGTGTTGGG	AATCTAC	CTATAG	GTAGT	TCCTCAAATC	GATTGGCGGA	357
SUT237	192	AGTAATGTGA	ATTGCAGAAT	TCAGTGAATC	ATCGAATCTT	TGAACGCACA	TTGCGCCCAT	TAGTATTCTA	GTGGGGCATGC	CTATTCGAGC	GTCATTTCAA	CCCTTACGCC	CTGTAGCG	TAGCOTTOGO	AATCTAC	CTGCGG	GTAGT	TCCTCAAATC	GATTGGCGGA	357
SUT154	198	AGTAATGTGA	ATTGCAGAAT	TCAGTGAATC	ATCGAATCTT	TGAACGCACA	TTGCGCCCAT	TAGTATTCTA	GTGGGCATGC	CTATTCGAGC	GTCATTTCAA	CCCTTAAGCC	CC-TGTTGCT	TAGTGTTGGG	AATCTGCG	TTACGGC	GCAGT	TCCTTAAAGT	GATTGGCGGA	366
SUT082	191	AGTAATGTGA	ATTGCAGAAT	TCAGTGAATC	ATCGAATCTT	TGAACGCACA	TTGCGCCCAT	TAGTATTCTA	GTGGGCATGC	CTATTCGAGC	GTCATTTCAA	CCCTTAAGCC	CC-TOTTOCT	TAGCGTTGGG	AATCTACCTC	CTTCGGGGG	GCGTAGT	TCCTGAAAGT	GATTGGCGGA	364
SUT074	214	AGTAATGTGA	ATTGCAGAAT	TCAGTGAATC	ATCGAATCTT	TGAACGCACA	TTGCGCCCGC	CAGTATTCTG	GCGGGCATGC	CTGTCTGAGC	GTCATTTCAA	CCCTCATGCC	CC-TAGGGCG	TGGTGTTGGG	GATCGGCCAA	AGCCCGCGAG	GGACGGCCGG	CCCCTARATC	TAGTGGCGGA	392
SUT108	207	AGTAATGTGA	ATTGCAGAAT	TCAGTGAATC	ATCGAATCTT	TGRACGCACA	TTGCGCCCGG	CAGTATTCTG	GCGGGGCATGC	CTGTTCGAGC	GTCATTTCAR	CCCTCAAGCT	CAGCT	TGGTGTTGGG	ACTCG-CGGT	AACCCGC	GT	TCCCCAAATC	GATTGGCGGT	369
SUTIBL	231	AGTAATOTGA	ATTGCAGAAT	TCAOTGAATC	ATCGAATCTT	TGAACGCACA	TTGCGCCTAA	TAGTATTCTG	TRAGGCATGC	CTGTTCGAGC	GTCATTTCAA	COCCAAGOO	CT-ATTTGCT	TGACGTTGGG	AGTTTACGGA	AAC	COLAR	TOCTCAAATA	TAGTGGCGGA	417
572321	265	AGTAATOTGA	ATTOCAGAAT	TCAGTGAATC	ATCOMATCHT	TGAACGCACA	TTOCOCCTAN	TAGTATTOTO	TTAGGCATGC	CTGTTCGAGC	GTCATTTCAA	CCCCCAAGCC	CT-ATTTCCT	TGACGTTGGG	AGTTTACCCA	AAC	GTAAT	TCCTCAAATA	TAGTOCCOGA	431
AJ390421	280	AGTAATGTGA	ATTGCAGAAT	TCAGTGAATC	ATCGAATCTT	TGAACGCACA	TTGCGCCTAA	CAGTATTCTG	TTAGGCATGC	CTGTTCGAGC	GTCATTTCAA	CCCCCAAGCC	CT-ATTTGCT	TGACGTTGGG	AGTTTACGGA	AAC	GTAAT	TCCTCAAATA	TAGTGGCGGA	446
KS15	235	AGTAATGTGA	ATTGCAGAAT	TCAGTGAATC	ATCGAATCTT	TGAACGCACA	TTGCGCCCAT	TAGTATTCTA	GTGGGCATGC	CTGTTCGAGC	GTCATTTCAA	CCCTTAAGCC	TTCTGTTGCT	TAGCGTTGGG	GGCCTACCGT	ATG	GCGGTAGC	CCCTTAAAAT	TAGTGGCGGA	405
AJ390411	282	AGTAATGTGA	ATTGCAGAAT	TCAGTGAATC	ATCGAATCTT	TGAACGCACA	TTGCGCCTAA	TAGTATTCTG	TTAGGCATGC	CTGTTCGAGC	GTCATTTCAA	CCCCCAAGCC	CT-ATTTGCT	TGACGTTGGG	AGTTTACGGA	GAC	GTAAT	TCCTCAAATA	TAGTGGCGGA	448
SUT290	280	AGTAATGTGA	ATTGCAGAAT	TCAGTGAATC	ATCGAATCTT	TGAACGCACA	TTGCGCCTAA	TAGTATTCTG	TTAGGCATGC	CTGTTCGAGC	GTCATTTCAA	CCCCCAAGCC	CT-GTCTGCT	TGACGTTGGG	AGCT-ACGGA	GAC	GTAAC	TCCTCAAATA	TAGTGGCGGA	445
AF201706	274	AGTAATGTGA	ATTGCAGAAT	TCAGTGAATC	ATCGAATCTT	TGAACGCACA	TTGCGCCTAA	TAGTATTCTG	TTAGGCATGC	CTGTTCGAGC	GTCATTTCAA	CCCCCAAGCG	CT-ACTTGCT	TGACGTTGGG	AATTTACAGC	TGCT	GTAAT	TCCTCAAATT	TAGTGGCGGA	441
SUT203	242	AGTAATGTGA	ATTGCAGAAT	TCAGTGAATC	ATCGAATCTT	TGAACGCACA	TTGCGCCCAT	TAGTATTCTA	GTGGGCATGC	CTGTTCGAGC	GTCATTTCAA	CCCTTAAGCC	CC-TGTTGCT	TAGCGTTGGG	AGCCTACA	-AC	CCTTCGTAGC	TCCTCAAAGT	TAGTGGCGGA	410
SUT129	240	AGTAATGTGA	ATTGCAGAAT	TCAGTGAATC	ATCGAATCTT	TGAACGCACA	TTGCGCCCAT	TAGTATTCTA	GTGGGGCATGC	CTGTTCGAGC	GTCATTTCAA	CCCTTAAGCC	CC-TGTTGCT	TAGCGTTGGG	AGCCTACA	-AC	CCTCTGTAGC	TCCTCAAAGT	TAGTGGCGGA	408
SUTOBB	242	AGTAATGTGA	ATTGCAGAAT	TCAGTGAATC	ATCGAATCTT	TGAACGCACA	TTGCGCCCAT	TAGTATTOTA	GTGGGGCATGC	CTGTTCGAGC	GTCATTTCAA	CCCTTAAGCC	CC-TOTTGCT	TAGCOTTOGG	AGCCTACAGA	TAC	COTOTOTAGE	TCC-CAAAGT	TAGTOGCOGA	412
ST2348	240	AGTAATGTGA	ATTGCAGAAT	TCAGTGAATC	ATCGAATCTT	TGAACGCACA	TTGCGCCCAT	TAGTATTCTA	GTGGGCATGC	CTGTTCGAGC	GTCATTTCAA	CCCTTAAGCC	CC-TGTTGCT	TAGCGTTGGG	AGCCTACA	-GC	CTTCTGTAGC	TCCCCAAAGT	TAGTGGCGGA	408
SUT078	240	AGTAATGTGA	ATTGCAGAAT	TCAGTGAATC	ATCGAATCTT	TGAACGCACA	TTGCGCCCAT	TAGTATTCTA	GTGGGCATGC	CTGTTCGAGC	GTCATTTCAA	CCCTTAAGCC	CC-TOTTOCT	TAGCGTTGGG	AGCCTACA	-GC	CCGCTGTAGC	TCCCCAAAGA	TAGTGGCGGA	408
SUT028	237	AGTAATGTGA	ATTGCAGAAT	TCAGTGAATC	ATCGAATCTT	TGAACGCACA	TTGCGCCCAT	TAGTATTCTA	GTGGGGCATGC	CTGTTCGAGC	GTCATTTCAA	CCCTTAAGCC	CC-TGTTGCT	TAGCGTTGGG	AGCCTACA	-GC	CCGCTGTAGC	TCCCCAAAGA	TAGTGGCNGA	405
SUT124	241	AGTAATGTGA	ATTGCAGAAT	TCAGTGAATC	ATCGAATCTT	TGAACGCACA	TTGCGCCCAT	TAGTATTCTA	GTGGGCATGC	CTGTTCGAGC	GTCATTTCAA	CCCTTAAGCC	CC-TOTTGCT	TACTOTTCCC	AGCCTACA		GCGATGTAGC	TCCTCAAAGT	TAGTGGCGGA	407
AJ390437	289	AGTAATGTGA	ATTGCAGAAT	TTAGTGAATC	ATCGAATCTT	TGAACGCACA	TTGCGCCCAT	TAGTATTCTA	GTGGGCATGC	CTGTTCGAGC	GTCATTTCAA	CCCTTAAGCC	TC-TGTTGCT	TAGTGTTGGG	AGCCTACCGC	ATTCT	TTGCTGTAGC	TCCTTAAAGT	TAGTGGCGGA	462
AJ390434	301	AGTAATGTGA	ATTGCAGAAT	TTAGTGAATC	ATCGAATCTT	TGAACGCACA	TTGCGCCCAT	TAGTATTCTA	GTGGGGCATGC	CTGTTCGAGC	GTCATTTCAA	CCCTTAAGCC	TT-TGTTGCT	TAGTGTTGGG	AGCCTACGGC	-TTTC	TCGCCGTAGC	TCCTTAAAGT	TAGTGGCGGA	4/3
S12325	237	AGTAATOTGA	ATTOCAGAAT	TCAGTGAATC	ATCOARTCTT	TGAALGCALA	TIGCGCCCAT	TAGTATICTA	GIGGGCATGC	CTOTICGAGE	GTCATTTCAA	COOTTANGCO	CE-TOTTOCT	TAGCOTTOGG	AGCCTACAGC		-ACCTOTAGE	TCCTCAAAGT	TAGTOGCOGA	404
502382	269	AGTAATGTGA	ATTGCAGAAT	TCAGTGAATC	ATCGAATCTT	TGAACGCACA	TTGCGCCCAT	TAGTATTCTA	GTGGGGCATGC	CTGTTCGAGC	GTCATTTCAA	CCCTTAAGCC	TC-TGTTGCT	TAGEGTTGGG	AGCCTACAGC		ACCGTAGC	TCCTCBAAGT	CAGTGGCGGA	435
ST2310	239	AGTAATGTGA	ATTGCAGAAT	TCAGTGAATC	ATCGAATCTT	TGAACGCACA	TTGCGCCCAT	TAGTATTCTA	GTGGGCATGC	CTGTTCGAGC	GTCATTTCAA	CCCTTAAGCC	TTCTGTTGCT	TAGCGTTGGG	GGCCTACCGT	A	TGGCGGTAGC	CCCTTAAAAT	TAGTGGCGGA	409
SUT127	235	AGTAATGTGA	ATTGCAGAAT	TCAGTGAATC	ATCGAATCTT	TGAACGCACA	TTGCGCCCAT	TAGTATTCTA	GTGGGGCATGC	CTGTTCGAGC	GTCATTTCAA	CCCTTAAGCC	TC-TOTTOCT	TAGTGTTGGG	AGCCTACGGT		-TCTAGTAGC	TCCTCAAAAT	TAGTGGCGGA	402
SUT123	237	AGTAATGTGA	ATTGCAGAAT	TCAGTGAATC	ATCGAATCTT	TGAACGCACA	TTGCGCCCAT	TAGTATTCTA	GTGGGGCATGC	CTGTTCGAGC	GTCATTTCAA	CCCTTAAGCC	CC-TGTTGCT	TAGCGTTGGG	AGCCTACTGC		CGGTAGC	TCCTCAAAGT	TAGTGGCGGA	402
SUT051	265	AGTAATGTGA	ATTGCAGAAT	TCAGTGAATC	ATCGAATCTT	TGAACGCACA	TTGCGCCCAT	TAGTATTCTA	GTGGGCATGC	CTGTTCGAGC	GTCATTTCAA	CCCTTAAGCC	CTGCTGCT	TAGTGTTGGG	AGCCTACGGA	GAC	GTAGC	TCCCCAAAGT	TAGTGGCGGA	430
SUT056	265	AGTAATGTGA	ATTGCAGAAT	TCAGTGAATC	ATCGAATCTT	TGAACGCACA	ATGCGCCCAA	TAGTAATCTA	GTGGGCATGC	CTGGTCGAGC	GTCATT-CAA	CCCTTAAGCC	CTGCTGGT	TAAGGT-GGG	AGCCTACGG-	GAC	GTAGC	TTCCCAAAGT	TAGTGGCGGA	427
AY541610	229	AGTAATGTGA	ATTGCAGAAT	TCAGTGAATC	ATCGAATCTT	TGAACGCACA	TTGCGCCCGC	CAGTATTCTG	GCGGGGCATGC	CTGTCCGAGC	GTCATTTCAA	CCCTCAAGCC	TCGTTGCT	TGGTGTTGGG	AGCCTACGG-	-CT	GTAGC	TCCTCAAAGT	CAGTGGCGGG	392
3,7390436	238	AGTANTOTON	ATTOCAGAAT	TCAGTGAATC	ATCGAATCTT	TGRACOCACA	TTGCGCCCAT	TAGTATTOTA	GTGGGGCATGC	CTOTTCGAGC	GTCATTTCAA	COCTTANGEC	CC-TOTTOCT	TAGCOTTOGO	AGCCTACCCC	33070	-TOTOGTAGO	TCCCCAAAGT	CAGTOGCOTO	410
SUT201	212	AGTAATGTGA	ATTGCAGAAT	TTAGTGAATC	ATCGAATCTT	TGAACGCACA	TTGCGCCCAT	TAGTATTCTA	GTGGGCATGC	CTGTTCGAGC	GTCATTTCAA	CCCTTAAGCC	TT-TOTTOCT	TAGCGTTGGG	AGCCTACGGT	AGC	GTAGC	TCCTCAAAAT	CAGTGGCGGA	378
SUT207	237	AGTAATGTGA	ATTGCAGAAT	TCAGTGAATC	ATCGAATCTT	TGAACGCACA	TTGCGCCCAC	TAGTATTCTA	GTGGGCATGC	CTGTTCGAGC	GTCATTTCAR	CCCTTAAGCC	TC-TGTTGCT	TAGTGTTGGG	AGCCTACGGT	CAT	AGTAGC	TTCTCAAAGT	-AGTGGCGGA	403
SUT032	242	AGTAATGTGA	ATTGCAGAAT	TCAGTGAATC	ATCGAATCTT	TGAACGCACA	TTGCGCCCAT	TAGTATTCTA	GTGGGCATGC	CTGTTCGAGC	GTCATTTCAA	CCCTTAAGCC	CC-TGTTGCT	TA-CGTTGGG	AGCCTACCGT	CAC	ACGTAGC	TCCTGAAAAG	TAGTGGCGGA	409
SUT090	238	AGTAATGTGA	ATTGCAGAAT	TCAGTGAATC	ATCGAATCTT	TGAACGCACA	TTGCGCCCAT	TAGTATTCTA	GIGGGCATGC	CTATTCGAGC	GTCATTTCAA	CCCTTAAGCC	T-TGTT-GCT	TAGCGTTGGG	AGTCTACGGC	TT	-CGGCGTAGC	TCCTGAAAGT	TAGTGGCGGA	406
SUT233	239	AGTAATGTGA	ATTGCAGAAT	TCAGTGAATC	ATCGAATCTT	TGAACGCACA	TTGCGCCCAT	TAGTATTCTA	GTGGGCATGC	CTATTCGAGC	GTCATTTCAA	CCCTTAAGCC	T-TGTT-GCT	TAGCGTTGGG	AGTCTACGGC	TT	-CGGCGTAGC	TCCTGAAAGT	TAGTGGCGGA	407
SUT066	268	AGTAATGTGA	ATTGCAGAAT	TCAGTGAATC	ATCGAATCTT	TGAACGCACA	TTGCGCCCAC	TGGTATTCCA	GTGGGCATGC	CTATTCGAGC	GTCATTTCAA	CCCTTAAGCC	CCTGTT-GCT	TAGCGTTGGG	AGTCTACGTC	TTA	-CGGCGTAGT	TCCTGAAAGT	TAGTGGCGGA	438
SUT063	228	AGCAATGTGA	ATTOCAGAAT	TCAGTGAATC	ATCGAATCTT	TGAACGCACA	TTGCGCCCAT	TAGTATTCTA	GEGGGGARGC	CTATTCGAGC	GTGATTTCGA	COOPERAGOO	ACTONT-GCT	TAGEGTTGGG	AGOATAC	0000	GGCGTAGT	TCCTTAAAGT	TAGTGGCGGGA	400
Ju2	280	AGTAATGTGA	ATTGCAGAAT	TCAGTGAATC	ATCGAATCTT	TGAACGCACA	TTGCGCCCAC	CAGTATTCTG	GTGGGCATGC	CTATTCGAGC	GTAATTTCAA	CCCTTAAGCC	CTTCGGCT	TAAGCGTGGG	AGTCCGCGTC	CC-C	GGCGCGGT	CCCC-AAAGT	CATTGGCGGC	447
SUT220	284	AGTAATGCGA	ATTGCAGAAT	TCAGTGAATC	ATCGAATCTT	TGAACGCACA	TTGCGCCCGC	CAGCATTOTO	GCGGGCATGC	CTATCCGAGC	GTCATTACGA	CCCTTAAGCC	CCCGGGCT	TAGCGTTGGG	ANTCOGCOTC	TT-CA	GGGGCGCGGT	TCCCTAAATT	CATCGGCGGC	455
SUT001	282	AGTAATGTGA	ATTGCAGAAT	TCAGTGAATC	ATCGAATCTT	TGAACGCACA	TTGCGCCCAC	TAGTATTCTG	GTGGGGCATGC	CTATTCGAGC	GTCATTTCAA	CCCTTAAGCC	CCTGTT-GCT	TAGCGTTGGG	COTCTGCGCC	GTGCC	CTGGCGCAGT	GCCCTAAATC	TATCGGCGGA	455
SUT218	264	AGTAATGTGA	ATTGCAGAAT	TCAGTGAATC	ATCGAATCTT	TGAACGCATA	TTGCGCCCAT	TAGTATTCTA	GTGGGCATGC	CTATTCGAGC	GTCATTTCGA	ACCATACGCC	TTTAGTAGCG	TAACGTTGGG	ACTCTTAGCC	CTGCC	ATAGGCTAGT	TCCCGAAGAA	CAGTGGCAGA	438
SUT223	264	AGTAATGTGA	ATTGCAGAAT	TCAGTGAATC	ATCGAATCTT	TGAACGCATA	TTGCGCCCAT	TAGTATTCTA	GTGGGCATGC	CTATTCGAGC	GTCATTTCGA	CCCTTACGCC	TCTAGTAGCG	TAGCGTTGGG	ACTCT-AGCC	CTGCC	ATAGGCTAGT	TCCCGAAGAC	CAGTGGCAGA	437
SUT016	248	AGTAATGTGA	ATTGCAGAAT	TCAGTGAATC	ATCGAATCTT	TGAACGCACA	TTGCGCCCAT	TAGTATTCTA	GTGGGGCATGC	CTATTCGAGC	GTCATTTCAA	CCCTTAAGCC	TCTCGGCT	TAGCGTTGGG	ACCCTGCGGC	GTAC	GGCGCAGG	TCCTTAAATT	TAGTGGCGGA	417
SUT202 SUT166	272	AGTAATGTGA	ATTOCAGAAT	TCAGTGAATC	ATCGAATCTT	TGAACGCACA	TTGCGCCCAT	TAGTATTCTA	GIGGGCATGC	CTATTCGAGC	GTCATTTCAA	CCCTTAAGCC	CTGTAGCT	TAGCOTTOGG	AATCTACGGC		TTACCCCC	CCCCCTAAAGT	TAGTGGCGGA	441
SUT158	232	AGTAATGTGA	ATTGCAGAAT	TCAGTGAATC	ATCGAATCTT	TGAACGCATA	TTGCGCCCAT	TAGTATTCTA	GTGGGCATGC	CTATTCGAGC	GTCATTTCAA	CCCTTAAG	CCTTGTTGCT	TAGTGTTGGG	AGTCTACC	-GCC	TGGCGGTAGT	TCCTARARG	TAGTGGCGGT	400
SUT294	270	COTNATOTOA	ATTGCANAAT	TCAGTGAATC	ATCGAATCTT	TGAACGCATA	NTGCGCCCAT	TAGTATTCTA	GTGGGCATGC	CTATTCGAGC	GTCATTTCAA	CCCTTAAG	CCCTGTAGCT	TAGCGTTGGG	ACTCTACT	CCTC	CGGGTGTAGT	TCCCTAAAAC	CANTGGCGGT	439
SUT182	260	AGTAATGTGA	ATTGCAGAAT	TCAGTGAATC	ATCGAATCTT	TGAACGCATA	TTGCGCCCAT	TAGTATTCTA	GTGGGCATGC	CTATTCGAGC	GTCATTTCAA	CCCTTAAG	CCCTGTTGCT	TACTOTTACC	AGCCTGCTTT	CTTC	AGAGGGCAGC	TCCCTAAAGA	TAGTG-CAGT	430
SUT148	240	AGTAATGTGA	ATTGCAGAAT	TCAGTGAATC	ATCGAATCTT	TGAACGCACA	TTGCGCCCAT	TAGTATTCTA	GTGGGGCATGC	CTATTCGAGC	GTCATTTCAA	CCCTTAAGC-	CCCTGTTGCT	TAACGTTGGG	AGCCTACG	TCTG	CGGGCGTAGT	TCCTGAAAGT	-AGTG-CGGA	408
SUT187	242	AGTAATGTGA	ATTGCAGAAT	TCAGTGAATC	ATCGAATCTT	TGAACGCACA	TTGCGCCCAT	TAGTATTCTA	GTGGGCATGC	CTATTCGAGC	GTCATTTCAA	CCCTTAAGC-	CCCTGTTGCT	TAGCGTTGGG	AGTCTACG	GTCA	TAGTAGC	TCCTCAAAGT	TAGTGGCGGA	409
SUT116	230	AGTAATGTGA	ATTGCAGAAT	TCAGTGAATC	ATCGAATCTT	TGAACGCACA	TTGCGCCCAT	TAGTATTCTA	GTGGGCATGC	CTATTCGAGC	GTCATTTCAA	CCCTTAAGCC	TC-AGTTGCT	TAGCGTTGGG	ACTCTACGAC	CTATT	ATAGCGTAGT	TCCTTAAAGT	TAGTGGCGGA	403
SUT103	208	AGTAATGIGA	ATTGCAGAAT	TCAGTGAATC	ATCGAATCTT	TGAACGCACA	TIGCGCCCAT	TAGTATICTA	GTGGGGCATGC	CTATTCGAGC	GICATTICAA	CCCTTAAGCC	-C-TGTAGCT	TAGCGTTGGG	AGTCCGCTA-	ATT	TTAGCGGC	TCCTTAAAGT	TATTGGCGGA	435
AF616682	234	AGTAATGTCA	ATTGCAGAAT	TCAGTGAATC	ATCGAATCTT	TGAACGCACA	TTGCGCCCAT	TAGTATTOTA	GTGGGGCATGC	CTATTCGAGC	GTCATTTCAA	CCCTTAACCC	TT-AGTTOCT	TAGCOTTOGO	AGTOTGCCOT	GTACTTOPT-	ACGGCGCACT	TCCTCAAAGT	GATTGGCCGA	417
AF616681	234	AGTAATGTGA	ATTGCAGAAT	TCAGTGAATC	ATCGAATCTT	TGAACGCACA	TTGCGCCCAT	TAGTATTCTA	GTGGGCATGC	CTATTCGAGC	GTCATTTCAA	CCCTTAAGCC	TT-AGTTGCT	TAGCOTTOGG	AGTCTGCGCT	GTACTTGTT-	ACCOCCACT	TCCTCAAAGT	GATTGGCGGA	411
SUT168D	225	AGTAATGTGA	ATTGCAGAAT	TCAGTGAATC	ATCGAATCTT	TGAACGCACA	TTGCGCCCAT	TAGTATTCTA	GTGGGCATGC	CTGTTCGAGC	GTCATTTCAA	CCCTTAAGCC	CC-TOTTOCT	TAGCGTTGGG	AATCTAGGTC	TCCAG	GGCCTAGT	TCCCCAAAGT	CATCGGCGGA	396
SUT085D	225	AGTAATGTGA	ATTGCAGAAT	TCAGTGAATC	ATCGAATCTT	TGAACGCACA	TTGCGCCCAT	TAGTATTCTA	GTGGACATGC	CTGTTCGAGC	GTCATTTCAA	CCCTTAAGCC	CC-TGTTGCT	TAACGTTGGG	AATCTAGGTC	TTCAG	GGCCTAGT	TCCCCAA-GT	TATTGGCGGA	395
AY616684	225	AGTAATGTGA	ATTGCAGAAT	TCAGTGAATC	ATCGAATCTT	TGAACGCACA	TTGCGCCCAT	TAGTATTCTA	GTGGGGCATGC	CTGTTCGAGC	GTCATTTCAR	CCCTTAAGCC	CC-TOTTGCT	TAGCGTTGGG	AATCTAGGTC	TCCAG	G-CCTAGT	TCCCCAAAGT	CATCGGCGGA	395
SUT164	237	AGTAATGTGA	ATTGCAGAAT	TCAGTGAATC	ATCGAATCTT	TGAACGCACA	TTGCGCCCAT	TAGTATTCTA	GTGGGCATGC	CTATTCGAGC	GTCATTTCAA	CCCTTAAGCC	TC-TGTCGCT	TAGCGTTGGG	AGCCTACGTC	TAC	AGCGTATC	TCCTCAAAGT	CAGTGGCGGT	406
ST2584	301	AGTAATGCGA	ATTGCAGAAT	TCAGTGAATC	ATCGAATCTT	TGAACGCACA	TTGCGCCCGC	TAGTATTCTG	GCGGGGCATGC	CTATTCGAGC	GTCATTACAA	CCCTTAAGCC	CC-TGTCGCT	TAGCGTTGGG	AGTCTGCGGC	TCA	GGCCGAGT	TCCTTAAA-T	TAGTGGCGAG	409

Figure 11C. (Continued).

		550	560	570	580	590	600	610	620	63	2	
ermote	200	comecce-c	cocommence	CARNER	manor	commente	-000000000	A		conce memory		460
SUT256	389	GTTGGGGGC-C	CCCCTTCAGC	GAAMAGITT	TATOT	COTTOTIG	-GGGGGGGCCA	A==TTCCGGC	COGRARGCCC	GTGATCTTNN		409
JUI	387	GTTGTAGC-A	CACTCTAAGC	GTAGTAGTTT	TCCATTGCCT	CGCTGCAG	-AGCGGCCTC	AGCTGCCAGC	CGTAAAGCCCC	-TATACTTCT	AGT	4/4
SUTO 70	350	GTTAGEAC-A	TACTUTAGGC	GTAGTAA-CA	CCATTCT	CGCTTCGGT-	AGTAAGTGCT	GGCGGCTAGC	CACTAAACCC	COTATACTTC	TAGT	440
SUIZ3/	220	GETTAGEGE-A	TACTOTAGGO	GIAGIAA-IA	CCAITCT	CGCITCIGI-	AGTA-GIGGT	AGCGGCTAGC	CATTAAACCC	CONTRACTOR	TAGE	452
501134	307	GUTAGIGC-A	TACTOTAGGE	GIAGIAAAIA	CCAIICI	CGCTTTTGT-	AGTAGGC-CT	GGCGGCTTGC	CG-TARAACC	COTATACTIC	TAGI	455
S01002	202	OCCOCTOCTOC	COROCOCCO	GIAGINA-CA	TACCICI	COCTICIOC-	-CACACOCAC	CACOCORCETOC	COTRARACCE	CONTRACTIC	1401	475
01014	373	CLCGTCGTGG	CERCENTER	GRAGIAGIGA	TATTCCG	CATCG	-GAGAGCOAC	COCCOUNTING	COTTANACCC	CANCELLO		475
501108	410	CACOTCO-AG	C-CREATAGE	GINGINATCA	TACA	COTTACTO-C	-GIAATCOTC	COROCCACOC	COTTANACCC	C-AACTICIG	AA1	500
SUT260	414	GTTAGGTCGT	G-CTCTAAGC	GTAGTAACTA	TATTCT	CGCTTCTG-C	AGCCGGCTTA	GGTCCTGC	CGTABABCCC		PCT	495
ST2321	432	GCTARGTOGT	G-CTCTAAGC	GTAGTAACTA	TATTAT	CGCTTCTG-T	AGCCGGTTTA	GGTCCTGC	CGTAAAACCT	TATATTT	TCT	514
AJ390421	447	GCTAGGTCGT	G-CTCTAAGC	GTAGTAATTA	TATTCT	CGCTTCTG-C	AGCCGGTCTA	GGTCTTGC	CGTAAAGCCC	TATATTTT	TCT	529
KS15	406	GTCGGTTCAC	CTCTAGAC	GTAGTAAATA	TTATCT	CGCCTATT-A	GTTGGACCGG	TCCCCTGC	CGTAAAACCC	C-TAATTTTC		485
AJ390411	449	GCTGGGTCGT	G-CTCTAAGC	GTAGTAACCA	AATTCT	CGCTTCTG-A	GGCCGGCCCG	GGTCCTGC	CGTAAAACCC	CCTATATTTT	TTTTCT-	536
SUT290	446	GCTAGGTCGT	G-CTCTAAGC	GTAGTAACTA	TACCCT	CGCTTCTG-T	AGCCGGCTAR	GGTCCTGC	CGGAAAACCC	C-TATATTTT	CT	528
AF201706	442	GCTAGGTCAT	G-CTCTGAGC	GTAGTAATTC	TGTTCT	CGCTCCTG-A	AGCTGCCCTA	TATCCTGC	CGTAAAACCC	C-TAAATTAT	AATCT	527
SUT203	411	GTCGGTTTCA	CACTCTAGAC	GTAGTAAT-T	TT-TA-TCTC	-GCCTATA	-GATGAGCCG	GTCCCC-TGC	CGTAAAACCC	CC-TAATTTT		491
SUT129	409	GTCGGTTTCA	CACTOTAGAC	GTAGTAAT-T	TT-TA-TCTC	-GCCTATA	-GATGAGCCG	ATCCCC-TGC	CGTAAAACCC	CC-TAATTTT		489
SUT192	415	GTCGGTTTCA	CACTCTAGAC	GTAGTAAA-T	TT-TA-TNTC	-GCCTATA	-GATGAGCCG	GTCCCT-TGC	CGTAAAACCC	CC-TAATTTC		495
SUTO88	413	GTCGGTTTCA	CACTCTAGAC	GTAGTAAT-T	TT-TA-TCTC	-GCCTATA	-GATGAGCCC	GGTCCTGC	CGTAAACC	CC-CAATTCT		490
ST2348	409	GTCGGTTT-A	CACTCTAGAC	GTAGTAAA-T	TT-TA-TCTC	AGTCTGCA	-GTTAGGCCG	GTCCCT-CGC	CGTAAAACCC	CC-CAATTTT		489
SUT078	409	GTCAGTTC-A	TACTCTAGGC	GTAGTAAA-T	CTCTA-TCTC	-GTCTATG	-GATGCGCTG	GCGCCT-CGC	COTAAAACCC	CCCTAATCTT	******	490
SUT028	406	GTCAGTNC-A	TACTCTAGGC	GTAGTAAA-T	CTGTA-TCTC	-NTCTATG	-GATGCGCTG	GCGCCT-CGC	CGTAAAACCC	CC-TAATCTT		486
SUT124	408	GTCGGTTACA	CACTCTAGGC	GCAGTAAACT	CTTTA-TCTC	-GTCTACG	-GTGTGGCCG	GTCCCT-TGC	CGTAAAACCC	CC-CAATTTC		490
AJ390437	463	GTCGGTTCAC	A-CTCTAGGC	GTAGTAAAGA	TTTTA-TTCT	CGCCTGTA-G	AGATGAGCCG	GTCCCC-TGC	CGTAAAACAC	CCCCCTATTT	TT	550
AJ390434	474	GTCGGTTCAC	A-CTCTAGGC	GTAGTAAATA	TTTTCT	CGCCTA-C	GGATGGACCG	GTCCCC-TGC	COTAAAAAA	алсалалтат	ATTTTA-	560
ST2325	405	GTCGGTTCAC	A-CTCTAGAC	GTAGTAATTT	TATCT	CGCCTATT	AGTTGACC	GTCTCC-CGC	CGTAAACCCT	ATTTTAAAGG		483
SUT092	485	ACCGGTCCGC	C-TTCTAGAC	GTAGTAATTC	TTTATTT	CGCCTACA	AGTCGTACCG	GTCCCTGC	CGTAAAAAGG	CGTTARGACA		566
ST2382	436	GTCGGCTCNC	AGCTCTAGAC	GTAGTAATTC	TTTTCT	CGCCTGTG	TGTTGTGCCG	GTCCCC-TGC	CGTAGAAACC	CCCCCTAATT		518
ST2310	410	GTCGGTTCAC	A-CTCTAGAC	GTAGTAAATA	TTATCT	CGCCTATT	AGTTGGACCG	GTCCCC-TGC	CGTAAAACCC	CTAATTTATC	AA	493
SUT127	403	GTCGGTTCAC	A-CTCTAGAC	GTAGTAATGT	TTATCT	CGCCTATC	AGTTGGACCG	GTCCCCGTGC	CGTAAAACCC	CTAATTTCTC		485
SUTI23	421	GTCGGTACGC	ACCCCTAGAC	GTAGTAATTC	TTATCT	CGCCTATA	SGTCGTGCCG	GTCCCC-TGC	COTTANACCC	CCCACIFIC		983
sum056	431	GTCGGIACGC	A-CICIAGAC	GARG-BATTA	TC-1AICI	CCCCCCCC	AGTOGGACCO	GROCCE-IGC	COLANAACCC	CCCASA MER-		501
37541610	202	GCTGGCTCGC	A-COCCAGE.	GRAGTAGTTA	TT-T-CTCT	CACCEGEGEG	-CTCCCCCCC	GTCCCC-TGC	COTABABOOC	CCCAGACTER	TTACTC-	480
SUT258	414	GTCGG-TCBC	A-CTCCAGAC	GTAGTACTCT	TT-CBCCT	CGCCTGTA	-GCTGGBCCG	GTCCCC-TGC	CGGBBBBBCBC	CCCAAAATTT		494
AJ390436	411	GCCGGTTCGC	A-CTCCAGAC	GTAGTAGCTT	TTACACGT	CGCCTGTA	GCGCGGGGCCG	GTCCCC-TGC	CGTAAAACAC	CCCAATTTTT	ATAGGT-	500
SUT201	379	GTCGGTTCAC	A-CTCTAGAC	GTAGTACAT-	TT-TATCT	CGTCTGTG	AGTTGGGCTG	GTCCCC-TGC	CGTAAAACCC	CTAATTTTT-		459
SUT207	404	GCTGGCTCAC	A-CTTCAGGC	GTAGTAGATG	CTATCT	CGCTTGTG	AGGTAAGCCG	GTCCCC-AGC	CGTAAAACCC	CGTAATTT		483
SUT032	410	GTCGGTTCTC	A-CTCTAGAC	GT-GTAAATT	CTATCT	CGCCTATC	AGTAGGACGC	CTCCCT-CGC	CGTAAAACCC	CCCTATATTT		490
SUTO90	407	GTTAGGGT-A	CACTOTCAGO	GTAGTAACA-	CTTCT	CGCTCGTGT-	GGTGGCCCTG	-GCTGCTGGC	COTTABACCC	CC-ATACCTT	TTAGT	491
SUT233	408	GTTAGGGT-A	CACTCTCAGC	GTAGTAACA-	CTTCT	CGCTCGTGT-	GGTGGCCCTG	-GCTGCTGGC	CGTTAAACCC	CC-ATACCTT	TTAGT	492
SUT066	439	GTCAGGGT-G	CACTCTCAGC	GTAGTAATT-	TCTCT	CGCTTGTGT-	GGTGTCCCTG	-GCTGCTGGC	COTTANACCC	CCTATATTTT	CTAGT	524
SUT063	453	GTTAGGGT-A	CACTCCCAGC	<b>GTAGTAAATA</b>	TCTCT	CGCNCGTGT-	GGGGGGCCCTG	-GCTGCTG-C	COTTABAACC	CCTATATCTT	TTAGT	538
SUT221	410	GTTAGGGT-A	CACTCTCAGC	GTAGTAATT-	TCTCT	CGCTCGGGT-	GGTGGGGGCCT	-GCTGCTG-C	CGTTAAGC	CCTATTTCTT	CGAGT	492
Ju2	448	TTCGCAGC-C	CACTCTGAGC	GTAGTAA-TC	AACTGGTTCT	CGCTCCTGC-	AGTGGCCGGC	GGAGCCC-GC	CGTAAAACCC	CCCCTA-TAA	CTAAGT-	538
SUT220	456	ecceseec-e	TETTETGAGE	GTAGTAA-TT	TATTATCT	CGCCCTGAA-	GCTAGCCCCG	TACGCCC-GC	CGTAAAACCC	CCCAAC-TAC	CTTGT	543
SUT001	456	GCCGTAGC-A	CACTCTGAGC	GTAGTAA-TT	-ACAGTTCCT	CGCTCCTGC-	AGTGGCCGCG	-GCGGCT-GG	CGTTAACCCC	TACAAC-T-T	CTAGT	542
SUT218	439	CCTGGGNCCG	TACCCTAAGC	GTAGTAAACT	TATCACAT	CGCTCTGGCT	GGTACCCCTA	GGCTTCTAGC	CGTAAAACCG	GACAAC-CTA	ACAGTTG	532
SUT223	438	CCTGGGGCCG	TACCCTAAGC	OTAGTAAACT	TATCACAT	COCTCIGGCT	GOTACCCCTA	GOCTTCTAGC	CUTAAAACTG	-ACAAC-CTA	GCAGTIG	530
SUTUIS	418	GITGGIGC-A	TACTCTTAGC	GTAGTAAATC	TTCT	CGCTCTCGT-	AGTACGCCTA	GCTACCC-GC	CUTAAAACCC	CUCTAT-TTT	TTAAT	502
501202	440	CTTOOTOC-A	CACTOTTAGE	CIAGINATIC	Terestor	COCTURCOC-	AGTAAGCCCCG	ABOCHOCCT-GC	COTANACCC	Company	Charter	526
SUT166	442	GTCAGGGC-3	CACTCOTAGE	GTAGTAATT	T	CCCTTCTCT-	AGTGGTTCTG	GCALCOTOTAGC	COTABABCCC	C-CTATTTCT	TACT	485
SUT294	440	GTTAGON	CCCTCATACC	GTAGTAAATC	TTTTTCT	CGCTTCTCC-	AGTGNGTCTA	NCTACCN-GC	CGTTABAACCC	CCCTATTTT	CTAGT	525
SUT182	431	GTTCGGGT-3	CACTOGRAGO	GTAGTAATOR	CTATCT	CGCTTCTCC-	AGTGGCCCCA	ATTATTC-CC	CGTAAAACCC	CTAPPTTC	TCAA	515
SUT148	400	GTTAGGGC-G	TACTOTTA-C	GTAGTAAATC	ACTATT	CGT-ACTC	AGTAGT-CTA	ACTTT-A-GC	CG-GAAACCC	CCATATTTAG		486
SUT187	410	GCTGGCTC-A	CACTCCAGGO	GTAGTAGATO	CTATCT	CGCTTGTG	AGTTAGGCCG	GTCCCCA-GC	CGTAAAACCC	CGTAATTTTT	AAAGGTT	498
SUT116	404	GTTATAGC-A	CACTCTAAGC	GTAGTAATTC	TCTCT	CCCTTCTTCT	AGTGGTTATA	GTTGCT-AGC	CATAAAACAC	CCCCTATT	TTAAT	489
SUT103	436	GTTATAGC-G	TACTCTAAGC	GTAGTAATTT	TTATCT	CGCTTCT-GT	AGTGGCCCTA	ACTGTT-AGC	CATAAAACCC	CTATATTTTT	CTANT	523
SUT120	415	GTTATAGC-A	CACTCTAAGC	GTAGTAAATC	TTCT	CGCTTCT-GA	GGTCGCCCTG	ACTCCT-CGC	CGTAAAACCC	CCTA-TTTTT	CTAAT	499
AF616682	412	GTTAGGGC-A	TACTCTAAGC	GTAGTAATAT	TTCTTCT	CGCTTCTGT-	AGTTGTCCTG	GCGGCT-TGC	CGT-AAACCC	CT		486
AF616681	412	GTTAGGGC-A	TACTOTAAGO	GTAGTAATAT	TTCTTCT	CGCTTCTGT-	AGTTGTCCTG	GCGGCT-TGC	COTTANACCO	CT		487
SUT168D	397	GTCGGAGC-G	TACTCTCAGC	GTAGTAATAC	CATTCT	CGCTTTTGC-	AGTAGCCCCG	GCGGCT-TGC	CGTAAAACCC	CTATATCTTT	AGTGGT-	485
SUT085D	396	GTCG-AGC-G	-ACTCTCAGC	G-AATAATAC	CATTCT	TGCTTTTGC-	AGTAACCCCG	GCGG-T-TGC	CGTAAA-CCC	CTATATCTTT	AGT	476
AY616684	396	GTCGGAGC-G	TACTCTCAGC	GTAGTAATAC	CATTCT	CGCTTTTGC-	AGTAGCACCG	GCGGCT-TGC	CGTAAAACCC	CT		470
SUT164	407	GCTGGAGC-A	CACTCTCAGC	GTAGTAAGTT	TTCTTCT	CGCTTCTGT-	AGTGGCCCCT	GCAGCC-TGC	CGTAAAACCT	CCAACACTTA	GT	492
ST2584	470	TACAGCAC	-AACCTAAGC	GTAGTAGGTT	ACCT	CGCTCCCGGG	GAGTCTGTGG	CCCCCCCCCC	AAAAAAAAA	CCTAAACCTT	CTA	553

Figure 11C. (Continued).

# Table 1C. The identity matrix of partial 18S rDNA sequences using NS1 and NS4 primers of Astrocystis mirabilis (SUT051, SUT056), Rosellinia sp. (ST2301), R. necatrix (AY083805), and A. cocoes (AY083804) calculated by the BioEdit program.

Species	A. mirabilis (SUT051)	A. mirabilis (SUT056)	A. cocoes (AY083804)	Rosellinia sp. (ST2301)	<i>R. necatrix</i> (AY083805)
A. mirabilis (SUT051)	1.000	1.000	0.982	0.981	0.974
A. mirabilis (SUT056)		1.000	0.982	0.981	0.974
A. cocoes (AY083804)			1.000	0.977	0.970
<i>Rosellinia</i> sp. (ST2301)				1.000	0.983
<i>R. necatrix</i> (AY083805)					1.000

Note: 1.000 means 100% identity.

Table 2C. The identity matrix of 18S rDNA ranging from NS1 and NS8 primers of

Astrocystis mirabilis (SUT056), Rosellinia sp. (ST2301), and R. necatrix

(AB014044) calculated by the BioEdit program.

Species	A. mirabilis (SUT056)	Rosellinia sp. (ST2301)	<i>R. necatrix</i> (AB014044)
A. mirabilis (SUT056)	1.000	0.703	0.687
Rosellinia sp. (ST2301)		1.000	0.673
<i>R. necatrix</i> (AB014044)			1.000

Note: 1.000 means 100% identity.

**Table 3C.** The identity matrix of ITS1-5.8S-ITS2 sequences of Astrocystis mirabilis (SUT056, SUT051), Rosellinia sp. ST2301, R.arcuata (AB017660), R. pepo (AB017659), R. quercina (AB017661), and R. necatrix (AB017657 and AB017658)calculated by the BioEdit program.

Species	AB017660	AB017657	AB017658	AB017659	AB017661	ST2301	SUT051	SUT056
AB017660	1.000	1.000	0.997	0.824	0.851	0.837	0.689	0.689
AB017657		1.000	0.997	0.824	0.851	0.837	0.689	0.689
AB017658			1.000	0.826	0.853	0.839	0.691	0.691
AB017659				1.000	0.772	0.764	0.659	0.659
AB017661					1.000	0.840	0.706	0.706
ST2301						1.000	0.699	0.699
SUT051							1.000	1.000
SUT056								1.000

Note: 1.000 means 100% identity.

Table 4C. The identity matrix of ITS1-5.8S-ITS2 sequences of Camillea tinctor (AJ390421, AJ390422), C. tinctor (ST2321), C.

obularia (AJ390423), C. tinctor (SUT161), C. tinctor (SUT260), and C. selangorensis (KS15) calculated by the BioEdit

program.

Species	AJ390421	AJ390422	ST2321	AJ390423	SUT161	SUT260	KS15
AJ390421	1.000	0.998	0.909	0.867	0.892	0.879	0.716
AJ390422		1.000	0.911	0.869	0.894	0.880	0.718
ST2321			1.000	0.858	0.937	0.920	0.730
AJ390423				1.000	0.863	0.856	0.709
SUT161					1.000	0.978	0.770
SUT260						1.000	0.766
KS15							1.000

Note: 1.000 means 100% identity.

## **APPENDIX D**

# **PHYLOGENETIC TREE CONSTRUCTIONS**



Figure 1D. Phylogenetic tree of *Rosellinia* and *Astrocystis* constructed by the maximum parsimony method using PAUP\* program based on ITS1-5.8S-ITS2 sequences. Tree length = 246; Consistent index (CI) = 0.8984; Homoplasy index (HI) = 0.1016; Retention index (RI) = 0.8663. Numbers of the branches indicate the bootstrap values resulting from 100 bootstrap replications.



Figure 2D. Phylogenetic tree of *Camillea* constructed by the maximum parsimony method using PAUP\* program based on ITS1-5.8S-ITS2 sequences. Tree length = 181; Consistent index (CI) = 0.9337; Homoplasy index (HI) = 0.0663; Retention index (RI) = 0.7600. Numbers of the branches indicate the bootstrap values resulting from 100 bootstrap replications.



Figure 3D. Phylogenetic tree of *Daldinia* constructed by the maximum parsimony method using PAUP\* program based on ITS1-5.8S-ITS2 sequences.
Tree length = 200; Consistent index (CI) = 0.8400; Homoplasy index (HI) = 0.1600; Retention index (RI) = 0.9489. Numbers of the branches indicate the bootstrap values resulting from 100 bootstrap replications.



Figure 4D. Phylogenetic tree of *Hypoxylon* sect. *Annulata* constructed by the maximum parsimony method using PAUP\* program based ITS1-5.8S-ITS2 sequences. Tree length = 613; Consistent index (CI) = 0.9396; Homoplasy index (HI) = 0.0604; Retention index (RI) = 0.9693. Numbers of the branches indicate the bootstrap values resulting from 100 bootstrap replications.



Figure 5D. Phylogenetic tree of *Hypoxylon* sect. *Annulata* constructed by the maximum parsimony method using PAUP\* program based on ITS1-5.8S-ITS2 sequences. Tree length = 867; Consistent index (CI) = 0.6586; Homoplasy index (HI) = 0.3414; Retention index (RI) = 0.8087. Numbers of the branches indicate the bootstrap values resulting from 100 bootstrap replications.



Figure 6D. Phylogenetic tree of *Hypoxylon* sect. *Annulata* constructed by the maximum parsimony method using PAUP\* program based on ITS2 sequences. Tree length = 330; Consistent index (CI) = 0.6242; Homoplasy index (HI) = 0.3758; Retention index (RI) = 0.8545. Numbers of the branches indicate the bootstrap values resulting from 100 bootstrap replications.



Figure 7D. Phylogenetic tree of *Hypoxylon* sect. *Hypoxylon* constructed by the maximum parsimony method using PAUP\* program based on ITS1-5.8S-ITS2 sequences. Tree length = 1437; Consistent index (CI) = 0.4878; Homoplasy index (HI) = 0.5122; Retention index (RI) = 0.7126. Numbers of the branches indicate the bootstrap values resulting from 100 bootstrap replications.



Figure 8D. Phylogenetic tree of *Hypoxylon* constructed by the maximum parsimony method using PAUP\* program based on ITS2 sequences. *Hypoxylon* sect. *Hypoxylon* is black whilst sect. *Annulata* is blue. Tree length = 798; Consistent index (CI) = 0.3797; Homoplasy index (HI) = 0.6203; Retention index (RI) = 0.4620. Numbers of the branches indicate the bootstrap values resulting from 100 bootstrap replications.



**Figure 9D.** Phylogenetic tree of *Xylaria* constructed by the maximum parsimony method using PAUP\* program based on ITS2 sequences. Tree length = 1230; Consistent index (CI) = 0.4886; Homoplasy index (HI) = 0.5114; Retention index (RI) = 0.6921. Numbers of the branches indicate the bootstrap values resulting from 100 bootstrap replications.



Figure 10D. Phylogenetic tree of xylariaceous fungi constructed by the maximum parsimony method using PAUP\* program based on ITS2 sequences. Tree length = 3244; Consistent index (CI) = 0.3203; Homoplasy index (HI) = 0.6797; Retention index (RI) = 0.5708. Numbers of the branches indicate the bootstrap values resulting from 100 bootstrap replications.



Figure 10D. (Continued).

## **APPENDIX E**

# LIST OF PRESENTATIONS

## **Poster Presentation**

- Suwannasai, N., Rodtong, S., Thienhirun, S., and Whalley, A.J.S. (2002). Taxonomic problems of high morphological variation of *Hypoxylon* spp. in Thailand. RGJ-Ph.D. Congress III, April 25-27, 2002, Jomtien Palm Beach Hotel and Resort, Chonburi, Thailand.
- Thienhirun, S., Rodtong, S., Phukhawan, N., and Suwannasai, N. Xylariaceous Fungus in Phu Hin Rongkra National Park, Thailand 2003. (2003). Technology for Life, July 17-21, 2003, Pattaya Exhibition and Conventional Hall, Thailand.

ณัฏฐิกา สุวรรณาศรัย, สุรีลักษณ์ รอดทอง, สุรางค์ เธียรหิรัญ, และ Whalley, A.J.S. (2547). ข้อ

มูลทางชีววิทยาโมเลกุลเพื่อการอนุกรมวิชานของเชื้อราสกุล Hypoxylon (Molecular and Biology data for the Taxonomy Study of Hypoxylon). การประชุมความหลากหลายทางชีวภาพ "งานวิจัยจากอดีตสู่อนาคต", 30 สิงหาคม —

3 กันยายน 2547, โรงแรมเวียงอินท์ จ.เชียงราย ประเทศไทย

Suwannasai, N., Rodtong, S., Thienhirun, S., and Whalley, A.J.S. (2004). ITS Sequence Heterogeneity of *Xylaria* Species and Some Other Xylariaceous Genera. The IV Asia-Pacific Mycological Congress and the IX International Marine and Freshwater Mycology Symposium, November 14-19, 2004, Lotus Hotel Pang Suan Kaew, Chiang Mai, Thailand.

- Suwannasai, N., Rodtong, S., Thienhirun, S., and Whalley, A.J.S. (2005). **Perispore Ornamentations for the Indication of** *Hypoxylon* **Species**. The 22<sup>nd</sup> Annual Conference of the Microscopy Society of Thailand, Febuary 2-4, 2005, The Tide Resort, Chonburi, Thailand.
- Suwannasai, N., Rodtong, S., Thienhirun, S., and Whalley, A.J.S. (2005). Molecular taxonomic studies of selected members of the Xylariaceae. Annual Scientific Meeting Exploitation of Fungi, September 5-8, 2005, British Mycological Society, Manchester, U.K.

## **Oral Presentation**

- Suwannasai, N., Rodtong, S., Thienhirun, S., and Whalley, A.J.S. (2004). **Trend in the Finding of New Xylariaceous Fungal Species in Thailand**. The 4<sup>th</sup> National Symposium on Graduate Research, August 10-11, 2004, Lotus Hotel Pang Suan Kaew, Chiang Mai, Thailand.
- Suwannasai, N., Rodtong, S., Thienhirun, S., and Whalley, A.J.S. (2004). Relationships within *Hypoxylon* Species Based on Morphological and Molecular Data. (2004). The IV Asia-Pacific Mycological Congress and the IX International Marine and Freshwater Mycology Symposium, November 14-19, 2004, Lotus Hotel Pang Suan Kaew, Chiang Mai, Thailand.
- Suwannasai, N., Rodtong, S., Thienhirun, S., and Whalley, A.J.S. (2005). Nucleotide Sequence Data for the Clarification of Species Complex in

**Xylariaceous Fungi**, RGJ-Ph.D. Congress VI, April 28-30, 2005, Jomtien Palm Beach Hotel and Resort, Chonburi, Thailand.

## Manuscripts

- Edwards, R.L., Jonglaekha, N., Kshirsagar, A., Maitland, D.J., Mekkamol, S., Nugent,
  L.K., Phosri, C., Rodtong, S., Ruchikachorn, N., Sangvichien, E.,
  Sharples, G.P., Sihanonth, P., Suwannasai, N., Thienhirun, S., Whalley,
  A.J.S., and Whalley, M.A. (2003). The Xylariaceae as phytopathogens.
  Recent Research Developments in Plant Sciences. 1: 1-19.
- Suwannasai, N., Rodtong, S., Thienhirun, S., and Whalley, A.J.S. (2005). Perispore Ornamentations for the Indication of *Hypoxylon* Species. Journal of Microscopy Society of Thailand. 19: 207-208.
- Suwannasai, N., Rodtong, S., Thienhirun, S., and Whalley, A.J.S. (2005). New Species and Phylogenetic Relationships of *Hypoxylon* Species from Thailand based on the Internal Transcribed Spacer Regions of Ribosomal Nucleotide Sequences. **Mycotaxon**.

335

#### S3-P7

#### Taxonomic Problems of High Morphological Variation of *Hypoxylon* spp. in Thailand

Nuttika Suwannasai", Sureelak Rodtong", Surang Thienhirun $^{\rm b}$  and AJS Whalley

- " School of Microbiology, Institute of Science, Suranaree University of Technology, Nakorn Ratchasima 30000, Thailand.
- \* Forest Products Research Division, Royal Forest Department, Chatuchak,
- Bangkok 10900, Thailand.
- <sup>c</sup> School of Biomolecular Sciences, Liverpool John Moores University, Liverpool L3 3AF UK.

#### Objective

To identify and classify *Hypoxylon* species using conventional taxonomic methodology.

#### Methods

*Hypoxylon*, Xylariaceae (Ascomycota) specimens, were collected from forest areas as recording by Thienhirun (1). The samples were identified and classified using their macroscopic and microscopic characteristics of teleomorph stage (1,2 and 3). The stromatal and perithecial structures were characterized in size, shape, texture and colour. The colour pigments were extracted using 10% KOH. Asci and ascospores were mounted in Melzer's reagent to detect blue apical rings from amyloid iodine reaction. Cultural features on potato dextrose agar (PDA) were performed. Fungal mycelia were collected for genomic DNA investigations in our future study

#### Results

Form morphological taxonomy, six species of *Hypoxylon: Hypoxylon stygium*, *Hypoxylon nitens*, *Hypoxylon moriforme*, *Hypoxylon purpurconitens*, *Hypoxylon stygium* var. *annulatum* and *Hypoxylon bovei* var. *microspora* were very closely related. Their extracted pigments were green, except *H. purpureonitens* was purple. The stromata, perithecia and disks were also vary in sizes and shapes. They were different from previously reported by Rogers (2). When cultured, they did not form stromatal structure (teleomorph stage) which commonly found in nature. Therefore, other techniques particularly molecular biology techniques (4), for example could help to clarify these problems.

#### Conclusion

Six *Hypoxylon* species were found to be difficult to identify and classify using morphological and culture characteristics, which are frequently used by several investigators. The absence of teleomorphs in their cultures increased the limitation of the conventional method. Nucleic acid techniques will be of our interest for further problem clarification.

Keywords: Hypoxylon, Xylariaceae, taxonomy, culture, teleomorph stage

#### Selected References

- Thienhirun S (1997) A Preliminary account of the Xylariaceae of Thailand. PhD Thesis, Liverpool John Moores University. UK.
- Ju YM, Rogers JD (1996) A revision of the genus Hypoxylon. Minnesota. USA: American Phytopathological Society Press.
- 3. Whalley AJS (1996) The xylariaceous way of life. Mycological Research 100, 897-922.
- Mazzaglia A, Anselmi N, Vicario S and Vannini A (2001) Sequence analysis of the 5.85 rDNA and ITS regions in evaluating genetic relationships among some species of Hypoxylon and related genera.

## ข้อมูลทางชีววิทยาโมเลกุลเพื่อการศึกษาอนุกรมวิธานของเชื้อราสกุล Hypoxylon

Molecular Biology Data for the Taxonomy Study of Hypoxylon (Fungi)

ณัฏฐิกา สุวรรณาศรัย ่ สุรีลักษณ์ รอดทอง ่ สุรางค์ เธียรหิรัญ² และ Anthony Whalley ้

่ สาขาวิชาจุลชีววิทยา สำนักวิชาวิทยาศาสตร์ มหาวิทยาลัยเทคโนโลยีสุรนารี นครราชสีมา 30000

<sup>2</sup> สำนักวิจัยการจัดการป่าไม้และผลิตผลป่าไม้ กรมป่าไม้ จตุจักร กรุงเทพฯ 10900

<sup>3</sup> School of Biomolecular Sciences, Liverpool John Moores University, Liverpool, L3 3AF, UK

#### บทคัดย่อ

Hypoxylon เป็นเชื้อรา Ascomycetes สกุลใหญ่สกุลหนึ่งในวงศ์ Xylariaceae ที่มีบทบาทสำคัญในการย่อยสลาย ซากพืชในระบบนิเวศและบางชนิดยังเป็นสาเหตุของโรคพืช ในการศึกษาอนุกรมวิธานของเชื้อราสกุลนี้อาศัยลักษณะ ทางสัณฐานเป็นหลักได้แก่ ลักษณะรูปร่าง ขนาด และ สี ของ Stroma, Perithecia, Ascospore, Germ slit และ Apical apparatus รวมถึงการเกิดปฏิกิริยาเคมีของ Stroma กับสารละลาย KOH 10% ซึ่งลักษณะทางสัณฐาน ดังกล่าวมีความผันแปรสูงและยากต่อการใช้เพื่อให้ได้ผลการศึกษาอนุกรมวิธานของ *Hypoxylon* ชนิดที่ใกล้เคียงกัน ดังนั้นในการศึกษาครั้งนี้จึงนำเทคนิคทางชีววิทยาโมเลกุลมาศึกษาอนุกรมวิธานของ *Hypoxylon* ชนิดที่ใกล้เคียงกัน ดังนั้นในการศึกษาครั้งนี้จึงนำเทคนิคทางชีววิทยาโมเลกุลมาศึกษาเพื่อให้ได้ข้อมูลลำดับนิวคลีโอไทด์ของดีเอ็นเอ (DNA) ในส่วน Internal transcribed spacer (ITS) regions 1 และ 2 และ 5.8S ribosomal RNA gene ซึ่งพบว่า ITS regions ดังกล่าวมีขนาดประมาณ 500 ถึง 900 คู่เบส และมีลำดับนิวคลีโอไทด์ที่สามารถใช้เพื่อช่วยระบุและจัด จำแนกชนิดของ *Hypoxylon* ได้อย่างชัดเจนและยังเป็นข้อมูลสำคัญที่ใช้เพื่ออธิบายความสัมพันธ์และวิวัฒนาการ ของเชื้อราสกุลนี้ได้ด้วย

#### Abstract

*Hypoxylon*, an Ascomycetes fungus, is one of the large genera of the family Xylariaceae, which plays a major role in wood decomposition in ecosystems, and some are weak plant pathogens. The taxonomy study of *Hypoxylon* species is principally relied on their morphological characteristics of stroma, perithecia, ascospore, germ slit, apical apparatus, and the chemical reaction of stroma with 10% KOH. The high variation of morphological characters among *Hypoxylon* species especially in closely related species is always encountered, which resulted in the taxonomic problems. In this study, the molecular biology technique was applied to obtain data of DNA sequences of internal transcribed spacer (ITS) regions 1 and 2 including 5.8S ribosomal RNA gene. Approximately 500 to 900 base pairs of the ITS sequences were achieved. These sequences could be used for aiding the clear-cut identification and classification of *Hypoxylon* species. The molecular data are also valuable for the explanation of relationships and evolution of the fungus.

คำสำคัญ: Xylariaceae, Hypoxylon, ITS, DNA sequences

#### บทนำ

เชื้อราสกุล *Hypoxylon* เป็นเชื้อราสกุลใหญ่สกุลหนึ่งในวงศ์ Xylariaceae (Ascomycetes) ประกอบด้วยอย่าง น้อย 130 ชนิด (Ju & Rogers, 1996) ซึ่งเป็นที่รู้จักและพบได้ในหลายประเทศโดยเฉพาะในเขตร้อนและเขตอบอุ่น ในประเทศไทยมีรายงานว่าพบเชื้อราสกุลนี้อย่างน้อย 47 ชนิด (Thienhirun, 1997) ซึ่งพบได้บนท่อนไม้และกิ่งไม้ เชื้อราสกุลนี้มีบทบาทสำคัญในการย่อยสลายซึ่งช่วยรักษาสมดุลของระบบนิเวศ และยังพบว่าบางชนิดเป็นสาเหตุ ของโรคพืช เช่น Hypoxylon rubiginosum และ Hypoxylon mammata (คือ Entoleuca mammata ตาม Edward และคณะ (2003) ซึ่งก่อให้เกิดโรค Canker ในการศึกษาอนุกรมวิธานของเชื้อราสกุลนี้อาศัยลักษณะทางสัณฐานเป็น หลักได้แก่ ลักษณะรูปร่าง ขนาด และ สี ของ Stroma, Perithecia, Ascospore, Germ slit และ Apical apparatus รวมถึงการเกิดปฏิกิริยาเคมีของ Stroma กับสารละลาย KOH 10% ซึ่งลักษณะทางสัณฐานดังกล่าวมีความผันแปร สูงโดยเฉพาะอย่างยิ่งชนิดที่ใกล้เคียงกันของ Hypoxylon Section Annulata ซึ่งพบ 1/3 ของเชื้อสกุล Hypoxylon มี ลักษณะเด่นคือ Ostioles อยู่ในระดับที่สูงกว่าผิวของ Stroma และมีลักษณะเป็นแผ่นกลม (Disc) อยู่ข้างบน ซึ่งยาก ต่อการศึกษาเพื่อให้ได้ข้อมูลทางอนุกรมวิธานที่ถูกต้อง ดังนั้นในการศึกษาครั้งนี้จึงได้นำเทคนิคทางชีววิทยาโมเลกุล โดยการหาลำดับนิวคลีโอไทด์ของดีเอ็นเอในส่วน Internal transcribed spacer (ITS) regions 1 และ 2 รวมทั้งส่วน 5.8S ribosomal RNA gene มาช่วยในการระบุและจัดจำแนกชนิดเพื่อเป็นข้อมูลในการศึกษาอนุกรมวิธานของเชื้อ รากลุ่มนี้ได้อย่างเชื่อมั่นและชัดเจนขอไป

### ระเบียบวิธีวิจัย

1. การศึกษาเพื่อการระบุและจัดจำแนกชนิดของเชื้อราสกุล Hypoxylon ที่พบในประเทศไทย

ศึกษาเพื่อการระบุและจัดจำแนกชนิดตามลักษณะทางสัณฐาน (ลักษณะ รูปร่าง ขนาด และ สี ของ Stromata, Perithecia, Ascospores, Germ slit และ Apical apparatus) รวมทั้งการเกิดปฏิกิริยาของ Stromata กับ KOH 10% ของเซื้อราสกุล *Hypoxylon* Section *Annulata* จำนวน 38 ตัวอย่าง ที่รวบรวมได้จากพื้นที่ในประเทศไทย (ตารางที่ 1)

2. การศึกษาเพื่อให้ได้ข้อมูลทางชีววิทยาโมเลกุลของเชื้อราสกุล Hypoxylon

แยกให้ได้เชื้อบริสุทธิ์จากสปอร์ โดยเพาะเลี้ยงบนอาหาร Potato Dextrose Agar (PDA) และ สกัดดีเอ็นเอ จากเส้นใยโดยใช้วิธีที่ประยุกต์จาก White และ คณะ (1990) จากนั้นใช้เทคนิคพีซีอาร์ (Polymerase Chain Reaction, PCR) เพิ่มจำนวนดีเอ็นเอในส่วน Internal Transcribed Spacer (ITS) regions 1 และ 2 รวมทั้งส่วน 5.8S ribosomal RNA gene โดยใช้ Primers ITS4 (5' TCCTCCGCTTATTGATATGC 3') และ ITS5 (5' GGAAGTAAAAGTCGTAACAAGG 3') (White และคณะ, 1990) ตรวจสอบขนาดของขึ้นดีเอ็นเอที่ได้ (PCR Products) โดยใช้เทคนิคเจลอิเล็กโทรโฟรีซีส (Gel electrophoresis) และหาลำดับ นิวคลีโอไทด์ ด้วยเครื่อง ABI370 Automate DNA Sequencer (Perkin Elmer, USA) จากนั้นวิเคราะห์ข้อมูลที่ได้ด้วยโปรแกรม Chromas และ BioEdit เพื่อ สร้างแผนภูมิความสัมพันธ์ทางพันธุกรรม (Phylogenetic tree) โดยใช้โปรแกรม MagAlign (DNASTAR, USA) เปรียบเทียบกับเซื้ออ้างอิงใน GenBank (AJ390397 และ AJ390409)

## ผลการวิจัย อภิปราย และ สรุปผลการวิจัย

1. การระบุและจัดจำแนกชนิดของเชื้อราสกุล Hypoxylon ตามลักษณะทางสัณฐาน

จากการศึกษาลักษณะทางสัณฐานเพื่อการระบุและจัดจำแนกชนิดของเซื้อรา Hypoxylon ใน Section Annulata สามารถจัดจำแนกได้อย่างน้อย 7 ชนิด (Species) คือ Hypoxylon stygium, H. atroroseum, H. nitens, H. moriforme, H. purpureonitens, H. bovei var. microspora, H. urceolatum และ พบว่าบางตัวอย่างไม่ สามารถจัดจำแนกชนิดได้ เนื่องจากลักษณะทางสัณฐานมีความผันแปรสูงคือสีและขนาดของ Stroma, Ostiolar disc, Ascospores รวมทั้งการเกิดปฏิกิริยาของ Stroma กับ KOH 10% ใกล้เคียงกันมากดังตัวอย่างในตารางที่ 1 และรูปที่ 1

## 2. ข้อมูลทางชีววิทยาโมเลกุลของเชื้อราในสกุล Hypoxylon

ในการเพิ่มจำนวนดีเอ็นเอในส่วน ITS regions ของเชื้อรา Hypoxylon พบว่ามีขนาดแตกต่างกันอยู่ในช่วง 500 ถึง 900 คู่เบส (รูปที่ 2) ซึ่งใช้เพื่อหาลำดับนิวคลีโอไทด์และความสัมพันธ์ เมื่อนำลำดับนิวคลีโอไทด์ที่ได้มาจัด แนวความสัมพันธ์ที่เหมาะสม (Alignment) พบความผันแปรสูงที่สุดในส่วน ITS1 โดยเฉพาะ H. stygium และ H. atroroseum (รูปที่ 3) ซึ่งมีลักษณะทางสัณฐานใกล้เคียงกันมาก แต่พบความต่างของลำดับนิวคลีโอไทด์ที่ได้มาจัด (รูปที่ 3) ส่วน H. nitens, H. bovei var. microspora และ H. moriforme ซึ่งมีลักษณะทางสัณฐานที่คาบเกี่ยวกัน และมีจำนวนตัวอย่างที่หลากหลาย พบว่า H. nitens มีขนาดของส่วน ITS1 ประมาณ 564 คู่เบส ขณะที่ H. bovei var. microspora และ H. moriforme มีขนาดประมาณ 207 และ 181 ตามลำดับ รวมทั้งมีลำดับนิวคลีโอไทด์ที่ แตกต่างกัน ทำให้สามารถแยก Hypoxylon แต่ละชนิดออกจากกันได้อย่างชัดเจน (รูปที่ 4) โดยหาความสัมพันธ์ทาง พันธุกรรมในรูปของ Phylogenetic tree (รูปที่ 5) และเชื้อ Hypoxylon บางตัวอย่างที่ยังไม่สามารถระบุและจัด จำแนกชนิดได้เช่น Hypoxylon sp. SUT103, Hypoxylon sp. ST2345 และ Hypoxylon sp. ST2406 พบว่า สามารถระบุแยกเชื้อราดังกล่าวออกมาจากกลุ่มของ Hypoxylon ชนิดอื่น อาจนำมาซึ่งการค้นพบเชื้อราชนิดใหม่ใน Hypoxylon ก็เป็นได้ โดยลำดับนิวคลีโอไทด์ที่ได้จากการศึกษาครั้งนี้สามารถนำมาใช้ออกแบบ Primers และ/หรือ Probes ที่จำเพาะต่อเชื้อแต่ละชนิดเพื่อช่วยในการศึกษาครั้งนี้สามารถนำมาใช้ออกแบบ Primers และ/หรือ ความสัมพันธ์และวิวัฒนาการของเชื้อราสกูดนี้อีก้วย
ตารางที่ 1 ความหลากหลายของลักษณะทางสัณฐานและการเกิดปฏิกิริยาของ Stroma กับ KOH 10% ของเชื้อราสกุล *Hypoxylon* Section *Annulata* ที่พบใน ประเทศไทย

จำนวนตัวอย่าง					10	2	ω	5	9	~	1	3	~	£-	-
พื้นที่ของจังหวัดที่พบเชื้อ					ตราด ราชบุรี	นครราชสีมา	ตราด นครราชสีมา	นครราชสีมา	ตราด นครราชสีมา สงขลา	ଖ୍ୟ୩ରୀ	ж	<b>ଶ</b> ଏ୩ର <b>୮</b>	г	1	
สีจากปฏิกิริยาของ	Stroma ทับ KOH 10%				នៅខ្មាភ	នៃខ្ម	្លឿ២៤	នៅ១១	2124	ะเวง	្លោមឲ	เหลือง-น้ำตาล	នៅខ្មាភ	រឿខ្មារ	91
านาดาคาล Ascospores (µm)					3.75-6.25 x 2.5-3.75	4.6-6.25 x 2.5-3.75	7.6-9.1 x 3.4-4.0	7.5-10.0 x 3.75-5.0	7.5-10.0 x 3.75-5.0	10.0-12.5 x 2.5-5.0	6.0-9.0 x 2.5-4.0	8.75-10.0-(11.5)x2.5-3.75	6.3-7.5 x 3.8	7.5-8.8 x 3.8-5	
ขนาด	୩୧୯	Ostiolar	disc	(mm/Ø)	0.1	0.1	0.3-0.4	0.3-0.6	0.2-0.5	0.2-0.3	0.2-0.5	0.1	0.1	0.3	
ଖି୩୧୦	Stroma				ด้ามัน	ด้า-ชมพู	ด้ามัน	ື່ຄຳ	ด้ามัน	ື່ຄຳ	ື່ຄຳ	ື່ຄ	ື່ຄ່	ື່ຄຳ	
ชนิดของเชื้อรา					Hypoxylon stygium	H. atroroseum	H. nitens	H. bovei var. microspora	H. purpureonitens	H. urceolatum	H. moriforme*	H. sp. SUT103	H. sp. ST2345*	H. sp. ST2406*	a

เชื้อจากสำนักวิจัยการจัดการป่าไม้และผลิตผลป่าไม้ กรมบ่าไม้ กรุงเทพฯ (ดร.สุรางค์ เธียรหิรัญ ผู้เก็บและดูแลตัวอย่างเชื้อรา)



รูปที่ 1 ตัวอย่างลักษณะ Stroma (กำลังขยาย 40 เท่า) ของเชื้อราสกุล Hypoxylon ใน Section Annulata (ก) Hypoxylon stygium, (ข) H. atroroseum, (ค) H. nitens, (ง) H. bovei var. microspora, (จ) H. purpureonitens, (ฉ) H. urceolatum, (ซ) Hypoxylon sp. SUT103, (ซ) Hypoxylon sp. SUT251



รูปที่ 2 ผลผลิตจาก PCR ในส่วนของ ITS1-5.8S-ITS2 ของเชื้อรา *Hypoxylon* Section *Annulata* เมื่อตรวจหาด้วย เทคนิคเจลอิเล็กโทรโฟรีซีส (Agarose 1.5%)

ช่อง 1: M, Molecular weight marker (100 base pairs DNA Ladder, Promaga); 1, 5 และ 8, Hypoxylon purpureonitens; 2 และ 4, H. atroroseum; 3, H. bovei var. microspora; 6, 7 และ 9, H. nitens; 10 และ 12, Hypoxylon sp. SUT103; 11, H. moriforme



รูปที่ 3 การจัดแนวความสัมพันธ์ที่เหมาะสมของลำดับนิวคลีโอไทด์ในส่วน ITS1 ของเชื้อ Hypoxylon atroroseum กับ H. stygium (ลูกศรแสดงช่วงของลำดับนิวคลีโอไทด์ที่ต่างกัน) ซึ่งพบความเหมือน (homology) 90 %



รูปที่ 4 การจัดแนวความสัมพันธ์ที่เหมาะสมของลำดับนิวคลีโอไทด์ในส่วน ITS1 ของเชื้อราในสกุล Hypoxylon 10 ชนิด (Hypoxylon atroroseum, H. stygium, H. nitens, Hypoxylon sp. ST2345, H. urceolatum, H. purpureonitens, Hypoxylon sp. SUT103, Hypoxylon sp. ST2338, H. moriforme และ H. bovei var. microspora)

	410	420	430	440	450	460	470	480
SUT003H.atroroseum.seq SUT05H.stygium.seq SUT05H.nitens.seq SUT06H.mitens.seq SUT06H.mirceolatum.seq SUT006H.purpureooitens.S SUT101H.sp.ST103.seq SUT101H.sp.ST103.seq SUT2406H.spS72338.seq ST2406H.moriforme.SEQ ST245H.moriforme.SEQ ST025H.bovel var.microsp	TTECAAGCI TTECAAGCI TGCTACCCTUTACI TGCTACCCTUTACI TTECAA - NICCCCCCGAA - GSCCCCCGAA - GSCCCCCCGAA - AGCCCETCAAA	асаалалатат ас асптетока осттетнов алеттас	ТОСТОБАЛЯ ОСРАНОТИТ ОТКЛОТОТИ ТОАТОБААТТ	AAATTICTTC TOCATTING CTACG-TO TAITIGGCOT CG CCCCC CCCCC CCCCC CCCCC CCCCC CCCCC CCCC	AATATTTOOT MITATTTOOT AATATTOOT A-ATCTO A-ATCTO A-CC A-CC GGCC A-CC	COMMANTOT COMMANTOT COMMANTATION COMMANTO ACCORTAT 	TOMASCINI TOMASTICI TOTTITICA TOTTITICA TOTA-SOLAI	TGAGOG -GAOOG -TACTAA TAGTOG CAGTAG
	490	5do	slo	520	s30	540	550	560
070098.stroroseum.seq UT0584.stygium.seq UT0581.stygium.seq UT0581.sp.072345.seq UT0988.urceolatum.seq UT0988.urceolatum.seq UT0988.sp.77103.seq T24068.sp.77103.seq T24068.sp.77103.seq T24068.sp.72338.seq T23368.moriforme.s2Q T23368.moriforme.s2Q	CTCTCA-ATO 	AATTCATAAA SURTCATAAA TCATAAA TCATAAA AATTCAAAA TCATAAAA AATTCTOAT AACTCTOAAA AACTCTOAAA	-AATTOSIAA A-TTOSIAA TOTTOSIAA TOSICOJAAT TTAAC CODIC-GHIT TAACCAAA TODIC-GHIT TAACCAAA TOTICTOACTACT TOACT-ACCT	A-GCURINTA AAGCORDIA TTOGRATHIT ATACITIC CORA ATACITIC GIAICITIC A-ARATT A-ACTIVIT	IA—ARCIA IA—ARCIA SCHORAL- STUSIAGAA SCHO XIPT-GA	CITT-T- CHITCTATT CREATTACT T- CITCT	TACOS TACOS TIT-TACAS TACOTOCAAA TAA	GTUA GTUA CTUAAT CUAAT 
	510	560	590					
UT009H.atroroseum.seq DT058H.stygium.seq DT05H.nitems.seq DT05H.urceolatum.seq DT05H.urceolatum.seq DT05H.purpureoniteus.s DT105H.sp.37103.seq	TCAAACCANGG TCAAACCANGG GCATCAAAACAAAT GCTTCAAACAAAT TCAACC TCAACAAC GCTTC-RAC	ТТТТАААА-С ТТТТААААА - ТТАСААААА - ТТАСААААА - АААТ - ТТАТАА - Т. ААА-	CARATACOPT CARATACOPT CARTACTTATAT CARTACTTATAT TACTT TACTT TACTT	а Ха Ха Аа Аа Аа Аа Аа				

รูปที่ 4 (ต่อเนื่อง)



รูปที่ 5 Phylogenetic tree ของ ITS1-5.8S-ITS2 regions ของเชื้อราชนิดต่างๆในสกุล *Hypoxylon* Section Annulata

**กิตติกรรมประกาศ**: งานวิจัยนี้ได้รับทุนสนันสนุนการวิจัยจากโครงการปริญญาเอกกาญจนาภิเษก สำนักงาน กองทุนสนับสนุนการวิจัย และการสนับสนุนสถานที่และเครื่องมือจากมหาวิทยาลัยเทคโนโลยีสุรนารีและกรมป่าไม้

## เอกสารอ้างอิง:

- Ju, Y.-M. and Rogers, J.D. (1996) A revision of the genus *Hypoxylon*. Minnesota: American Phytopathological Society Press.
- (2) Edwards, R.D., Jonglaekha, N., Kshirsagar, A., Maitland, D.J., Mekkamol, S., Nugent, L.K., et al. (2003) The Xylariaceae as phytopathogens. Recent Research Develop Plant Science. 1: 1-19.
- (2) Sánchez-Ballesteros, J.,González, V., Salazar, O., Acero, J., Portal, M.A., Julián, M., et al. (2000) Phylogenetic study of *Hypoxylon* and related genera based on ribosomal ITS sequences. Mycologia. 92: 964-977.
- (3) Thienhirun, S. (1997) A Preliminary Account of the Xylariaceae of Thailand: Liverpool John Moores University.
- (4) Whalley, A.J.S. (1996) The xylariaceous way of life. Mycological Research. 100: 897-922.
- (5) White, T.J., Bruns, T.D., Lee, S. and Taylor, J.W. (1990). Amplification and direct sequencing of fungal ribosomal RNA genes for phylogenetics. In M.A. Innis, D.H. Gelfand, J.S. Sninsky and T.J. White (eds.). PCR Protocols: a guide to methods and applications (pp. 315-322). San Diego: Academic Press.

#### IV ASIA-PACIFIC MYCOLOGICAL CONGRESS

An improved method for the rapid dereplication of impure chemical ITS sequence heterogenecity of *Xylaria* species and some other Xylariaceous genera

## N. Suwannasai<sup>1</sup>, S. Rodtong<sup>1</sup>, S. Thienhirun<sup>2</sup> and A.J.S. Whalley<sup>3</sup>

<sup>1</sup>School of Microbiology, Institute of Sciences, Suranaree University of Technology, Nakhon Ratchasima 30000, Thailand

<sup>2</sup>Forest Management and Forest Products Research Office, Royal Forest Department, Chatuchak, Bangkok 10900, Thailand

<sup>3</sup>School of Biomolecular Sciences, Liverpool John Moores University, Liverpool, L3 3AF, UK

Xylaria was the first described genus of the family Xylariaceae and it is a large and relatively well-known fungus group in most countries in the world. There are also other well-known genera such as Biscogniauxia, Camillea, Daldinia, and Hypoxylon. They occur on wood, leaves, seeds, dung, and soil or in a few cases are associated with insects. Some species are weak phytopathogens and many Xylaria species have been reported as endophytes living inside healthy plant tissue without apparent damage to the host. Recently, endophytes have been widely investigated because of their ability to produce new or interesting secondary metabolites some of which have proved to be bioactive. The conventional taxonomic studies of Xylaria have been based on morphological and cultural characteristics but have in some cases included their metabolite profiles. The major problems concern the high morphological variation among Xylaria species depending on stages of development and localities of collection, and the lack of teleomorph stage in the culture. Therefore, the molecular taxonomic study was undertaken overcome these problems. Nucleotide sequences of ITS1, 5.8S, and ITS2 rDNA of 48 Xylaria isolates as well as some other Xylariaceous genera obtained from different collection locations were analyzed. Alignments of the Xylaria species sequences exhibited the greatest variation in the ITS regions. The 5.8S sequence gave approximately 99% similarity for all isolates tested but ITS sequence comparison results supported a monophyletic group in this genus, which is separated from the genera Biscogniauxia, Camillea, Daldinia, and Hypoxylon. The sequences proved to be valuable for the taxonomic investigation of fungi such as Xylaria with their high morphological variation. In addition, a database of this molecular data would be useful for the designation of specific primers and for the development of species specific probes for the detection of Xylaria in environmental situations.

**AP59** 

22nd MST Annual Conference

Journal of Microscopy Society of Thailand 2005, 19(1): 207-208

Poster Presentation

# Perispore Ornamentations for the Indication of Hypoxylon Species

## Nuttika Suwannasai<sup>1</sup>, Sureelak Rodtong<sup>1</sup>, Surang Thienhirun<sup>2</sup>, Anthony JS Whalley<sup>3</sup>

School of Microbiology, Institute of Science, Suranaree University of Technology, Nakhon Ratchasima 30000, Thailand

<sup>2</sup>Forest Management and Forest Products Research Office, Royal Forest Department,

Chatuchak, Bangkok 10900, Thailand <sup>3</sup>School of Biomolecular Sciences, Liverpool John Moores University, Liverpool L3 3AF, UK

The perispore ornamentation observed by using scanning electron microscope (SEM) has been recently used to indicate new or complex species in the fungal taxonomy. Some species of Hypoxylon have been reported concerning the significance of this characteristic in species indication but many of them have no record. In this study, three complex species of Hypoxylon fendleri, H. retpela, and H. cf. lenormandii collected in Thailand, have been investigated in their morphological characteristics both macroscopic and microscopic methods including perispore ornamentations by SEM. Hypoxylon fendleri and H. retpela are very closely related species. Their SEM micrographs of perispores resulted the same conspicuousness of the coil-like ornamentation which was different from Ju and Rogers (1996) who stated that H. fendleri and H. retpela differ mainly in the conspicuousness of the ornamentation on the perispore. However DNA sequencing results indicated 14 % divergence among both species. In case H. cf. lenormandii SUT065 occurring on bamboo has strange coil-like ornamentation which was different from H. lenormandii occurring on wood that have inconspicuous coil-like ornamentation. This result was supported by DNA sequencing data to separate H. cf. lenormandii to be a new variety.

#### References

- Ju Y-M, Rogers JD. A Revision of the Genus Hypoxylon. Mycologia Memoir No. 20. APS Press 1. 1996.
- Rogers JD, Candoussau F. Hypoxylon gillesii, a new species with ornamented ascospores from 2. Madagascar. Mycotaxon 1982, 15: 507-514.
- 3. Van der Gucht K. Spore ornamentation makes a nice difference: Daldinia eschscholzii and Daldinia concentrica. In: Aspects of Tropical Mycology (Editted by Isaac S, Frankland JC, Watling R, Whalley AJS) Cambridge, Cambridge University Press 1993, 309-310.
- Van der Gucht K, Van der Veken P, Contribution towards a revision of the genus Hypoxylon s. str. (Xylariaceae, Ascomycetes) from Papua New Guinea. Mycotaxon 1992, 44: 275-299.
- Whalley MA, Whalley AJS, Jones EBG. Camillea selangorensis sp. nov. from Malaysia. Sydowia 1996, 48: 145-151.



Figure 1. The fungus genus Hypoxylon. SEM micrographs of coil-like perispore ornamentation of H. cf. lenormandii (a), H. retpela (b), and H. fendleri (c), germ slit of H. cf. lenormandii (arrow) (d); and ostiole of H. cf. lenormandii (arrow) (d); and ostiole of H. cf. lenormandii (arrow) (e). Light microscope micrographs of H. retpela ascus containing eight ascospores (f), stromatal forms of H. fendleri (g), H. retpela (h), and H. cf. lenormandii (i). Bars equal 1 µm for a, b, c, d, e; and 1 mm for g, h, i.

## Annual Scientific Meeting Exploitation of Fungi September 5-8 2005

Gms

British Mycological Society promoting fungal sc

## Abstract Form - Papers and Posters

#### Please submit by email to: <u>bmsmeetings@tiscali.co.uk</u> An electronic copy can be obtained from www.britmycolsoc.org.uk

#### LAST DATE FOR SUBMISSION - JUNE 1 2005

Authors names N.Suwannasai, S.Rodtong, S. Thienhirun and A.J.S. Whalley

Address(es) Institute of Microbiology, Faculty of Science, Suranaree University of Technology, Nakhon Ratchassima 300000; Forest Products Research Division, Royal Forest Department, Bangkok 10900, Thailand & School of Biomolecular Sciences, Liverpool John Moores University, Byrom Street, Liverpool L3 3AF, UK., Thailand, R

Email address of presenting author a.j.whalley@livjm.ac.uk

Paper/Poster title

Molecular taxonomic studies of selected members of the Xylariaceae

Abstract - maximum 200 words

Representative species of a number of genera belonging to the family Xylariaceae (Ascomycotina) were collected from different regions of Thailand. These were identified using traditional morphological characters of both teleomorph and anamorph stages (when available) and together with selected temperate and non-Thai species were subjected to molecular examination. Nucleotide sequences of ITS1, 5.8S and ITS tDNA were analysed. In general the ITS1 region exhibited the greatest variation among the species studied whereas 5.8S and ITS2 regions were more conserved. The molecular data indicated that *Xylaria* is a monophyletic group which is separated from the genera *Biaognianxia, Camillea, Daldinia* and *Hypoxylan.* The data also supported a clear distinction between *Astropystia* and *Rosellinia* which contrasts with some modern authors views. Furthermore it was found that closely related species groups or genera appeared as separate entities but retained a close similarity. In most cases the molecular data supported the traditional taxonomic grouppings.

Please identify which mode of presentation your prefer and the session relevant to your work, by checking the boxes below.

Oral Presentation	Poster presentation	
Session 1	Session 4	
Session 2	Session 5	
Session 3	Howard Eggins Session	

For office use

Date received	Acknowledged	Acceptance as oral paper	Acceptance as poster	Formatted for publication	

D\_ST\_34

## แนวโน้มการพบเชื้อราชนิดใหม่ในกลุ่ม Xylariaceae ในประเทศไทย

TREND IN THE FINDING OF NEW XYLARIACEOUS FUNGAL SPECIES IN THAILAND ณัฏฐิกา สุวรรณาศรัย<sup>1</sup> สุรัลักษณ์ รอดทอง' สุรางค์ เธียรหิรัญ² และ Anthony Whalley<sup>3</sup>

่ สาขาวิชาจุลชีววิทยา สำนักวิชาวิทยาศาสตร์ มหาวิทยาลัยเทคโนโลยีสุรนารี นครราชสีมา 30000, <sup>2</sup> สำนักวิจัยการ จัดการบำไม้และผลิตผลบำไม้ กรมปำไม้ กรุงเทพฯ, <sup>3</sup> School of Biomolecular Sciences, Liverpool John Moores University, Liverpool, UK; e-mail address: d4410126@ccs.sut.ac.th

บทคัดย่อ: เชื้อราวงศ์ Xylariaceae เป็นเชื้อวงศ์ใหญ่ใน Ascomycetes ประกอบด้วย 45 สกุล (genera) พบได้บน ท่อนไม้ ไบ่ไม้ ผลไม้ มูลสัตว์ และบนดิน และมีบทบาทสำคัญในการย่อยสลายซึ่งช่วยรักษาสมดุลของระบบนิเวศ นอกจากนี้ยังพบว่าบางชนิดเป็นสาเหตุของโรคพีซ และบางชนิดสามารถเจริญอยู่ร่วมกับพืชโดยไม่ก่อให้เกิดโรคและ อาจสร้างสารต้านแมลงศัตรูพืชได้อีกด้วย (3) จากการสำรวจและเก็บตัวอย่างเชื้อรากลุ่ม Xylariaceae ในพื้นที่ 9 จังหวัดในประเทศไทยได้จำนวน 320 ตัวอย่าง สามารถจัดจำแนกตามลักษณะทางสัณฐานและการเจริญได้ 69 ชนิด ใน 9 สกุล คือ Hypoxylon, Xylaria, Daldinia, Astrocystis, Camellea, Biscogniauxia, Rosellinia, Kretzschmaria และ Nemania ทั้งนี้พบเชื้อ Hypoxylon 1 ชนิด ที่มีแนวโน้มว่าจะเป็นขนิดใหม่ คือมีวงแหวนสีขาว อยู่บนผิวของ Stroma ซึ่งเป็นลักษณะเด่นในการระบุว่าเป็น Hypoxylon michelianum ที่พบในแถบยุโรปเท่านั้น (1) แต่เสื้อที่พบนี้มีลักษณะดังกล่าวแต่ขนาดของสปอร์เล็กกว่าประมาณ 1 เท่าซึ่งยังไม่เคยมีการรายงานมาก่อน

ระเบียบวิธีวิจัย: รวบรวมตัวอย่างเชื้อราในกลุ่ม Xylariaceae จากพื้นที่ 9 จังหวัดในประเทศไทยมาศึกษาเพื่อการ ระบุและจัดจำแนกชนิดตามลักษณะทางสัณฐาน (ลักษณะ ขนาด รูปร่าง และรูปแบบของ Stromata, Perithecia, Ascus, Ascospores, Apical apparatus) รวมทั้งการเกิดปฏิกิริยาของ Stromata กับ KOH 10% และศึกษา ลักษณะการเจริญของเส้นใยโดยเพาะเลี้ยงในอาหาร Potato Dextrose Agar (PDA)

ผลการวิจัย อธิปราย และสรุปผลการวิจัย: จากตัวอย่างที่รวบรวมได้จำนวน 320 ตัวอย่าง สามารถจัดอยู่ในสกุล

Hypoxylon 35 ขนิด Xylaria 25 ขนิด Daldinia 1 ขนิด Astrocystis 1 ขนิด Carnellea 1 ชนิด Biscogniauxia 3 ชนิด Rosellinia 1 ชนิด Kretzschmaria 1 ชนิด Nemania 1 ขนิด และพบเชื้อ Hypoxylon 1 ชนิด ที่มีแนวโน้มว่าจะเป็นขนิดใหม่คือมีวงแหวนสี ขาวอยู่บนผิวของ Stroma ซึ่งเป็นลักษณะจำเพาะของ Hypoxylon michelianum ที่ พบในแถบยุโรปเท่านั้น (1) แต่เชื้อที่พบนี้มีขนาดสปอร์เล็กกว่าประมาณ 1 เท่า ซึ่งยัง ไม่เคยมีการรายงานมาก่อน ทั้งนี้การศึกษาลักษณะทางสัณฐานมีข้อจำกัดในการระบุ และจัดจำแนกขนิดได้อย่างเชื่อมั่นและชัดเจน ดังนั้นจึงได้ดำเนินการศึกษาต่อเนื่อง โดยการใช้เทคนิคทางชีวโมเลกุลเพื่อการระบุชนิดที่แน่นอน สร้างฐานข้อมูล และ พัฒนาวิธีการตรวจจับเชื้อแต่ละชนิดต่อไป



รูปที1. ตัวอย่างเชื่อราในกลุ่ม Xylariaceae

คำนิยม: งานวิจัยนี้ได้รับทุนวิจัยจากโครงการปริญญาเอกกาญจนาภิเษก สำนักงานกองทุนสนับสนุนการวิจัย และ การสนับสนุนสถานที่และเครื่องมือจากมหาวิทยาลัยเทคโนโลยีสุรนารี

เอกสารอ้างอิง: (1) Ju, Y.-M. and Rogers, J.D. (1996) A revision of the genus *Hypoxylon*. Minnesota: American Phytopathological Society Press.

(2) Thienhirun, S. (1997) A Preliminary Account of the Xylariaceae of Thailand: Liverpool John Moores University. IV ASIA-PACIFIC MYCOLOGICAL CONGRESS

AO3-3

# Relationships within Hypoxylon species based on morphological and molecular data

# N. Suwannasai<sup>1</sup>, S. Rodtong<sup>1</sup>, S. Thienhirun<sup>2</sup> and A.J.S. Whalley<sup>3</sup>

School of Microbiology, Institute of Sciences, Suranaree University of Technology, Nakhon Ratchasima, Thailand 30000.

<sup>2</sup>Forest Management and Forest Products Research Office, Royal Forest Department, Chatuchak, Bangkok, Thailand 10900.

<sup>3</sup>School of Biomolecular Sciences, Liverpool John Moores University, Liverpool, L3 3AF, UK.

Hypoxylon is one of the largest and best known genera of the family Xylariaceae. It has been reported to comprise of at least 130 species and found in most countries but is especially well represented in the tropics and subtropics. Although Hypoxylon species are primarily wood-decay fungi, that play an important role in the natural functions of ecosystems, many species are weak plant pathogens of angiosperms. Some Hypoxylon species seem to be highly host specific but others appear to exhibit wide host ranges. In this study, relationships between Hypoxylon species are revealed based on their morphological and molecular characteristics. One hundred and eighty six Hypoxylon specimens were collected from several forest areas in Thailand and these were then subjected to taxonomic investigation based on their morphological and cultural features and nucleic acid sequences. It was found that there can be considerable variation in morphological characters and furthermore some specimens could not be cultured. This resulted in some identification problems. However, different relationships between the Hypoxylon species were achieved following sequence analysis of ITS1-5.8S-ITS2 rDNA regions. The molecular results showed clearly the relationships of the Hypoxylon species studied and could be used to solve the morphological taxonomic problems. The ITS1 region indicated the highest variation among Hypoxylon species whereas 5.8S and ITS2 regions were more conserved. These molecular data could be applied for distinguishing morphological similar Hypoxylon species which had otherwise proved difficult to separate.

RGJ - Ph.D. Congress VI

#### \$3A-020

## Nucleotide Sequence Data for the Clarification of Species Complex in Xylariaceous Fungi

### Nuttika Suwannasai<sup>a</sup>, Sureelak Rodtong<sup>a</sup>, Surang Thienhirun<sup>b</sup> and Anthony Whalley<sup>c</sup>

<sup>a</sup> School of Microbiology, Institute of Science, Suranaree University of Technology, Nakhon Ratchasima 30000, Thailand,

<sup>b</sup> Forest Management and Forest Products Research Office, Royal Forest Department, Bangkok 10900, Thailand.
<sup>c</sup> School of Biomolecular Sciences, Liverpool John Moores University, Liverpool L3 3AF, U.K.

#### Introduction

Because xylariaceous fungi are high variation in morphological characteristics, an attempt to use the molecular techniques to assist was performed.

#### Objective

To clarify the species complex in xylariaceous fungi using internal transcribed spacer region sequences of rRNA gene.

#### Methods

Two hundreds and fifty one xylariaceous isolates were collected from different forest areas in Thailand and identified to species level based on morphological methods (2). Then, their ascospores were isolated and cultured on potato dextrose agar for DNA extraction (1). The internal transcribed spacer regions (ITS) including 5.8S ribosomal nucleotide sequences was amplified and sequenced. ITS sequences were aligned by ClustalX and manually edited by BioEdit program. The phylogenetic trees were constructed using neighbour joining (NJ) and maximum parsimony (MP) methods by PHYLIP and PAUP software packages respectively.

#### Results

Nine genera of Xylariaceae, Astrocystis, Biscogniauxia. Camillea, Daldinia, Hypoxylon, Kretzschmaria, Nemania, Rosellinia, and Xylaria, were recorded from the whole lot of specimens collected, and identified to fifty species. Hypoxylon and Xylaria were the common genera, and showed high variation in their morphological characters resulting in the difficulty in identification. Their amplification sizes of ITS fragments ranged from 500-900 base pairs, which contained ITS1, ITS2 and 5.8S rDNA regions. Most Hypoxylon section Annulata indicated the extremely long sequences in the ITS1 region, which were the tandem repeat sequences. These repeated sequences could be generated by slipped-strand mispairing or replication slippage. The whole ITS sequence alignments revealed the greatest variation in ITS1 regions, which was suitable to design specific primers and/or probes for these particular strains. The phylogenetic tree showed clearly the relationships of complex species. In this study, at least two new species and one new variety of Hypoxylon were recorded.

#### Conclusion

The nucleotide sequence data based on ITS sequences were proved to be useful for the clarification of species complex in xylariaceous taxonomic investigation. These results are also very useful to create the DNA sequence database of the xylariaceous fungi found in Thailand.

Keywords: Xylariaceae, Nucleotide sequence, Phylogeny, ITS

#### **Selected References**

- Platas, G., Acero, J., Borkowski, J.A., Gonzá, V., Portal, M.A., Rubio. (2001). Presence of a Simple Tandem Repeat in the ITS1 Region of the Xylariales. *Current Microbiology*, 43: 43-50.
- Thienhirun, S. (1997). A Preliminary Account of the Xylariaceae of Thailand: Ph.D Thesis, Liverpool John Moores University, U.K.

161

## **CURRICULUM VITAE**

- 1) NAME: Miss Nuttika Suwannasai
- 2) DATE OF BIRTH: 3 June 1973
- 3) PLACE OF BIRTH: Lopburi, Thailand

## 4) EDUCATION:

- 1991-1994 B.Sc. (Microbiology), Chulalongkorn University, Bangkok, Thailand.
- 1995-1998 M.Sc. (Microbiology, Immunology), Mahidol University, Bangkok, Thailand.

## 5) EXPERIENCE:

- 1998-1999 Research assistant at the Plant Genetics Conservation Project, Chitralada Palace, Bangkok, Thailand.
- 2000-2001 Research assistant at the National Center for Genetic Engineering and Biotechnology (BIOTECH), Bangkok, Thailand.

## 6) SCHOLARSHIP:

2001-2005 The Thailand Research Fund through the Royal Golden Jubilee Ph.D. Program (Grant No. PHD/0096/2544).