

APPENDIX A

Table A.1 List of fungal records on *Castanopsis* (Farr *et al.*, 2006).

Fungal species	Host species	Locations
<i>Acantharia chaetomoides</i>	<i>Castanopsis</i> sp.	Taiwan
<i>Acantharia sinensis</i>	<i>Castanopsis indica</i>	India
<i>Amazonia balakrishnanii</i>	<i>Castanopsis armata</i>	India
<i>Anthostomella castanopsis</i>	<i>Castanopsis carlesii</i>	Taiwan
<i>Aporhytisma urticae</i>	<i>Castanopsis acuminatissima</i>	Papua New Guinea
<i>Armillaria mellea</i>	<i>Castanopsis chrysophylla</i>	(Root rot.) Oregon,
<i>Aschersonia</i> sp.	<i>Castanopsis</i> sp.	Nepal
<i>Asterina castanopsis</i>	<i>Castanopsis kawakamii</i>	China
<i>Asterina pasaniae</i>	<i>Castanopsis cuspidata</i>	Japan
<i>Astrosporina angustifolia</i>	<i>Castanopsis</i> sp.	Indonesia; Papua New Guinea
<i>Astrosporina bresadolae</i>	<i>Castanopsis</i> sp.	Papua New Guinea
<i>Astrosporina echinosimilis</i>	<i>Castanopsis</i> sp.	Papua New Guinea
<i>Astrosporina gemina</i>	<i>Castanopsis</i> sp.	Papua New Guinea
<i>Astrosporina granulosiceps</i>	<i>Castanopsis</i> sp.	Papua New Guinea
<i>Astrosporina incognita</i>	<i>Castanopsis</i> sp.	Papua New Guinea
<i>Astrosporina lasserooides</i>	<i>Castanopsis</i> sp.	Papua New Guinea
<i>Astrosporina lutea</i>	<i>Castanopsis</i> sp.	Japan; Papua New Guinea
<i>Astrosporina neoumbrina</i>	<i>Castanopsis</i> sp.	Japan; Papua New Guinea
<i>Astrosporina olivaceonigra</i>	<i>Castanopsis</i> sp.	Papua New Guinea
<i>Astrosporina papillata</i>	<i>Castanopsis</i> sp.	Papua New Guinea
<i>Astrosporina petchii</i>	<i>Castanopsis</i> sp.	Indonesia; Malaysia; Papua New Guinea
<i>Astrosporina punctatosquamosa</i>	<i>Castanopsis</i> sp.	Papua New Guinea
<i>Astrosporina titibuensis</i>	<i>Castanopsis</i> sp.	Indonesia; Japan
<i>Bagcheea albomaculans</i>	<i>Castanopsis cuspidata</i> var. <i>sieboldii</i>	Japan
	<i>Castanopsis tribuloides</i> var. <i>ferox</i>	India; Japan
<i>Beltrania rhombica</i>	<i>Castanopsis cuspidate</i>	Japan
	<i>Castanopsis cuspidata</i> var. <i>sieboldii</i>	Japan
<i>Beltraniella buloloensis</i>	<i>Castanopsis</i> sp.	Papua New Guinea
<i>Blastophorum truncatum</i>	<i>Castanopsis cuspidata</i> var. <i>sieboldii</i>	Japan
	<i>Castanopsis</i> sp.	Papua New Guinea
<i>Botryosphaeria</i> sp.	<i>Castanopsis</i> sp.	(On twigs.) Georgia
<i>Ceratosporella lambdaepta</i>	<i>Castanopsis</i> sp.	Papua New Guinea
<i>Chaetendophragma triangularis</i>	<i>Castanopsis</i> sp.	Papua New Guinea
<i>Chaetendophragma triseptata</i>	<i>Castanopsis cuspidata</i> var. <i>sieboldii</i>	Japan
<i>Chaetopsina ramifera</i>	<i>Castanopsis</i> sp.	Papua New Guinea
<i>Chaetothyrium sinense</i>	<i>Castanopsis sclerophylla</i>	China
<i>Chalara montellica</i>	<i>Castanopsis</i> sp.	Papua New Guinea
<i>Ciboria americana</i>	<i>Castanopsis</i> sp.	China
<i>Ciboria glaucescens</i>	<i>Castanopsis argentea</i>	Indonesia
	<i>Castanopsis javanica</i>	Indonesia
	<i>Castanopsis</i> sp.	Indonesia
<i>Ciliospora gelatinosa</i>	<i>Castanopsis cuspidata</i> var. <i>sieboldii</i>	Japan
<i>Cladosporium elatum</i>	<i>Castanopsis cuspidate</i>	Japan

Table A.1 (Continued)

Fungal species	Host species	Locations
<i>Coccomyces castanopsidis</i>	<i>Castanopsis chrysophylla</i>	California,
<i>Coccomyces mucronatus</i>	<i>Castanopsis</i> sp.	China
<i>Codinaea gonytrichoides</i>	<i>Castanopsis cuspidate</i>	Japan
<i>Codinaea simplex</i>	<i>Castanopsis</i> sp.	Papua New Guinea
<i>Coriolus versicolor</i>	<i>Castanopsis chrysophylla</i>	Oregon; Washington
<i>Coronellaria castanopsidis</i>	<i>Castanopsis chrysophylla</i>	Oregon
<i>Cronartium orientale</i>	<i>Castanopsis cuspidata</i> var. <i>sieboldii</i>	Japan
<i>Cronartium quercuum</i>	<i>Castanopsis cuspidate</i>	Japan
	<i>Castanopsis cuspidata</i> var. <i>sieboldii</i>	Japan
<i>Cryphonectria macrospora</i>	<i>Castanopsis cuspidate</i>	Japan
<i>Cryptophiale guadalcanalensis</i>	<i>Castanopsis cuspidata</i> var. <i>sieboldii</i>	Japan
<i>Cystotheca lanestris</i>	<i>Castanopsis delavayi</i>	China
	<i>Castanopsis tibetana</i>	China
<i>Cystotheca tijbodensis</i>	<i>Castanopsis argentea</i>	Indonesia
<i>Cystotheca wrightii</i>	<i>Castanopsis cuspidate</i>	Japan
<i>Dactylaria clavata</i>	<i>Castanopsis cuspidata</i> var. <i>sieboldii</i>	Japan
<i>Dactylaria obtriangularia</i>	<i>Castanopsis cuspidata</i> var. <i>sieboldii</i>	Japan
<i>Dasyscyphus echinophilus</i>	<i>Castanopsis sempervirens</i>	California
<i>Denticularia hachijoensis</i>	<i>Castanopsis cuspidate</i>	Japan
<i>Diplocradiella scalaroides</i>	<i>Castanopsis cuspidate</i>	Japan
	<i>Castanopsis cuspidata</i> var. <i>sieboldii</i>	Japan
<i>Dothidella castanicola</i>	<i>Castanopsis chrysophylla</i>	California,
	<i>Castanopsis sempervirens</i>	California
<i>Dothidella castanopsidis</i>	<i>Castanopsis chrysophylla</i>	California; Oregon
	<i>Castanopsis</i> sp.	(Leaf spot.) California
<i>Dothidella castanopsii</i>	<i>Castanopsis tribuloides</i>	Nepal
<i>Dothidella janus</i>	<i>Castanopsis chrysophylla</i>	California; Oregon;
		Washington
<i>Dothidella janus</i>	<i>Castanopsis sempervirens</i>	California
<i>Elaphomyces reticulosporus</i>	<i>Castanopsis chinensis</i>	China
<i>Ellisiopsis gallesiae</i>	<i>Castanopsis cuspidata</i> var. <i>sieboldii</i>	Japan
<i>Endophragmia</i> sp.	<i>Castanopsis cuspidata</i> var. <i>sieboldii</i>	Japan
<i>Endothia havanensis</i>	<i>Castanopsis cuspidata</i> var. <i>sieboldii</i>	Japan
<i>Endothia macrospora</i>	<i>Castanopsis cuspidata</i> var. <i>sieboldii</i>	Japan
<i>Endothia parasitica</i>	<i>Castanopsis</i> sp.	E states
<i>Endothia singularis</i>	<i>Castanopsis cuspidata</i> var. <i>sieboldii</i>	Japan
<i>Erysiphe fagacearum</i>	<i>Castanopsis concolor</i>	China
	<i>Castanopsis delavayi</i>	China,
	<i>Castanopsis</i> sp.	China,
<i>Erysiphe graminis</i>	<i>Castanopsis</i> sp.	China
<i>Erysiphe polygoni</i>	<i>Castanopsis concolor</i>	China
	<i>Castanopsis delavayi</i>	China,
<i>Erysiphe sikkimensis</i>	<i>Castanopsis concolor</i>	China
	<i>Castanopsis delavayi</i>	China
	<i>Castanopsis indica</i>	India
	<i>Castanopsis</i> sp.	China
	<i>Castanopsis tribuloides</i>	India
<i>Fomes igniarius</i>	<i>Castanopsis chrysophylla</i>	(Heart rot.) Oregon,
<i>Fomes korthalsii</i>	<i>Castanopsis argentea</i>	Indonesia
<i>Fomes truncatosporus</i>	<i>Castanopsis</i> sp.	China
<i>Ganoderma lucidum</i>	<i>Castanopsis chrysophylla</i>	Oregon
<i>Ganoderma oregonense</i>	<i>Castanopsis chrysophylla</i>	(Root rot.) Oregon
<i>Gloeosporium castanopsidis</i>	<i>Castanopsis chrysophylla</i>	(On leaves.) California,

Table A.1 (Continued)

Fungal species	Host species	Locations
<i>Godronia castanopsidis</i>	<i>Castanopsis chrysophylla</i> <i>Castanopsis sempervirens</i> <i>Castanopsis</i> sp.	California California Taiwan
<i>Helotium</i> sp.	<i>Castanopsis sempervirens</i>	(On dead limbs.) California,
<i>Hericium erinaceus</i>	<i>Castanopsis cuspidate</i>	Japan
<i>Hymenochaete tabacina</i>	<i>Castanopsis chrysophylla</i>	(On dead limbs.) California
<i>Hypocrea albocornea</i>	<i>Castanopsis</i> sp.	China; Japan
<i>Hypocrea aurantia</i>	<i>Castanopsis</i> sp.	China
<i>Hypocrea lactea</i>	<i>Castanopsis</i> sp.	Japan
<i>Hypocrea pezizoides</i>	<i>Castanopsis</i> sp.	Japan
<i>Hypocrea tropicosinensis</i>	<i>Castanopsis</i> sp.	China
<i>Hypocrella raciborskii</i>	<i>Castanopsis acuminatissima</i>	Papua New Guinea
<i>Idriella fertilis</i>	<i>Castanopsis cuspidate</i>	Japan
<i>Irenina castanopsidis</i>	<i>Castanopsis tibetana</i>	China, ,
<i>Isthmolongispora minima</i>	<i>Castanopsis cuspidate</i> <i>Castanopsis cuspidata</i> var. <i>sieboldii</i>	New Guinea Japan
<i>Isthmotricladia</i> sp.	<i>Castanopsis</i> sp.	Papua New Guinea
<i>Lachnella bicolor</i>	<i>Castanopsis sempervirens</i> <i>Castanopsis chrysophylla</i>	California, California
<i>Lambertella obpyriformis</i>	<i>Castanopsis</i> sp.	China
<i>Lanzia castanopsis</i>	<i>Castanopsis argentea</i> <i>Castanopsis</i> sp.	Indonesia Indonesia
<i>Lanzia huangshanica</i> f. <i>aurantiaca</i>	<i>Castanopsis</i> sp. <i>Castanopsis cuspidata</i> var. <i>sieboldii</i> <i>Castanopsis sieboldii</i> <i>Castanopsis</i> sp.	China Japan Japan China; Japan,
<i>Laschia agaricina</i>	<i>Castanopsis</i> sp.	China
<i>Lentinula edodes</i>	<i>Castanopsis</i> sp.	Papua New Guinea
<i>Marasmius copelandii</i>	<i>Castanopsis</i> sp.	California
<i>Melanconium cinctum</i>	<i>Castanopsis</i> sp.	Georgia
<i>Meliola castanopsidis</i>	<i>Castanopsis tibetana</i>	China, ,
<i>Meliola castanopsina</i>	<i>Castanopsis carlesii</i> var. <i>sessilis</i> <i>Castanopsis formosana</i> <i>Castanopsis hystrix</i> <i>Castanopsis stipitata</i> <i>Castanopsis tribuloides</i> var. <i>formosana</i>	Taiwan China China; Taiwan China; Taiwan Taiwan
<i>Meliola fusispora</i>	<i>Castanopsis argyrophylla</i> <i>Castanopsis carlesii</i> var. <i>sessilis</i> <i>Castanopsis stipitata</i>	China Taiwan China; Taiwan
<i>Meliola hystrix</i>	<i>Castanopsis hystrix</i>	India
<i>Meliola kawakamii</i>	<i>Castanopsis fabri</i>	China
<i>Meliola mannii</i>	<i>Castanopsis armata</i>	India
<i>Meliola quercina</i>	<i>Castanopsis hystrix</i>	China
<i>Meliola shiiae</i>	<i>Castanopsis argyrophylla</i> <i>Castanopsis borneensis</i> <i>Castanopsis carlesii</i> var. <i>sessilis</i> <i>Castanopsis fabri</i> <i>Castanopsis hystrix</i> <i>Castanopsis kawakamii</i> <i>Castanopsis</i> sp. <i>Castanopsis stipitata</i>	China Taiwan Taiwan China China China; Taiwan China China China; Taiwan China China; Taiwan
<i>Meliola subacuminata</i>	<i>Castanopsis subacuminata</i>	China; Taiwan
<i>Meliola taiyuensis</i>	<i>Castanopsis fissa</i>	China

Table A.1 (Continued)

Fungal species	Host species	Locations
<i>Meliola taiwaniana</i>	<i>Castanopsis hystrix</i> <i>Castanopsis indica</i> <i>Castanopsis taiwaniana</i>	Taiwan China China; Taiwan,
<i>Merismodes fasciculata</i> var. <i>quercina</i>	<i>Castanopsis pubinervis</i>	Japan
<i>Microsphaera alni</i>	<i>Castanopsis chrysophylla</i> <i>Castanopsis</i> sp.	(Powdery mildew.) North Carolina (Powdery mildew.) North Carolina
<i>Microsphaera alphitoides</i>	<i>Castanopsis cuspidate</i> <i>Castanopsis cuspidata</i> var. <i>sieboldii</i>	Korea Japan
<i>Microsphaera sinensis</i>	<i>Castanopsis delavayi</i>	China,
<i>Mirandina typica</i>	<i>Castanopsis</i> sp.	Argentina
<i>Monochaetia nodosporella</i>	<i>Castanopsis delavayi</i>	China
<i>Mycosphaerella janus</i>	<i>Castanopsis chrysophylla</i> <i>Castanopsis sempervirens</i> <i>Castanopsis chrysophylla</i>	California California (On fallen leaves.) Oregon
<i>Myriophacidium</i> <i>aphyophyllicum</i>		
<i>Nakataea fusispora</i>	<i>Castanopsis cuspidata</i> var. <i>sieboldii</i>	Japan
<i>Nectria</i> sp.	<i>Castanopsis acuminatissima</i>	Papua New Guinea
<i>Nematostigma himalayana</i>	<i>Castanopsis tribuloides</i>	Nepal
<i>Oidium</i> sp.	<i>Castanopsis</i> sp.	China
<i>Palawaniella castanopsidis</i>	<i>Castanopsis tribuloides</i>	India,
<i>Peniophora sanguinea</i>	<i>Castanopsis chrysophylla</i>	Oregon,
<i>Pestalotia distincta</i>	<i>Castanopsis cuspidate</i>	Japan
<i>Pestalotia</i> sp.	<i>Castanopsis cuspidata</i> var. <i>sieboldii</i>	Korea
<i>Pestalotiopsis distincta</i>	<i>Castanopsis cuspidate</i>	Japan
<i>Pezicula cinnamomea</i>	<i>Castanopsis chrysophylla</i>	(On dead twigs.) Oregon
<i>Phanerochaete sordida</i>	<i>Castanopsis cuspidate</i>	Japan
<i>Phellinus ferreus</i>	<i>Castanopsis chrysophylla</i>	California
<i>Phellinus illiciicola</i>	<i>Castanopsis</i> sp.	China
<i>Phellinus robustus</i>	<i>Castanopsis hystrix</i>	China
<i>Phlebiella tulasnelloidea</i>	<i>Castanopsis cuspidata</i>	Japan
<i>Phlebiopsis flavidoalba</i>	<i>Castanopsis cuspidate</i>	Japan
<i>Phyllactinia roboris</i>	<i>Castanopsis</i> sp.	China
<i>Phyllosticta castaneae</i>	<i>Castanopsis sclerophylla</i>	China
<i>Phyllosticta castanicola</i>	<i>Castanopsis sempervirens</i>	California
<i>Phyllosticta castenopsidis</i>	<i>Castanopsis stellatospina</i>	Taiwan
<i>Phytophthora</i> sp.	<i>Castanopsis sempervirens</i>	California
<i>Plenotrichum castanopsidis</i>	<i>Castanopsis tribuloides</i>	India,
<i>Polyporus hirsutus</i>	<i>Castanopsis chrysophylla</i>	Oregon; Washington
<i>Polyporus picipes</i>	<i>Castanopsis chrysophylla</i>	Oregon
<i>Polyporus versicolor</i>	<i>Castanopsis chrysophylla</i>	Oregon
<i>Poria ferrea</i>	<i>Castanopsis chrysophylla</i>	California
<i>Prataprajella fusispora</i>	<i>Castanopsis</i> <i>Castanopsis</i> <i>subacuminata stipitata</i>	China; Taiwan
<i>Prataprajella subacuminata</i>	<i>Castanopsis</i> <i>Castanopsis</i> <i>subacuminata stipitata</i>	China; Taiwan
<i>Pseuderospora castanopsidis</i>	<i>Castanopsis tibetana</i>	China,
<i>Pucciniastrum castaneae</i>	<i>Castanopsis javanica</i> <i>Castanopsis</i> sp.	India Pakistan
<i>Pyropolyporus pusillus</i> var. <i>pusillus</i>	<i>Castanopsis</i> sp.	China
<i>Radulodon copelandii</i>	<i>Castanopsis cuspidate</i>	Japan
<i>Ramularia fusisaprophytica</i>	<i>Castanopsis</i> sp.	Papua New Guinea,

Table A.1 (Continued)

Fungal species	Host species	Locations
<i>Ramularia rhombica</i>	<i>Castanopsis cuspidate</i>	Japan
	<i>Castanopsis cuspidata</i> var. <i>sieboldii</i>	Japan
<i>Rhinocladiella cristaspora</i>	<i>Castanopsis cuspidata</i> var. <i>sieboldii</i>	Japan
<i>Rhinocladiella</i> sp.	<i>Castanopsis cuspidata</i>	Japan
<i>Rutstroemia</i> sp.	<i>Castanopsis sempervirens</i>	(On dead limbs.) California
<i>Scolecobasidium tricladiatum</i>	<i>Castanopsis cuspidata</i> var. <i>sieboldii</i>	Japan
<i>Septobasidium pseudopedicellatum</i>	<i>Castanopsis</i> sp.	Georgia
<i>Sistotrema oblongisporum</i>	<i>Castanopsis</i> sp.	Japan
<i>Solosympodiella clavata</i>	<i>Castanopsis cuspidata</i> var. <i>sieboldii</i>	Japan
<i>Speirospis pedatospora</i>	<i>Castanopsis cuspidata</i> var. <i>sieboldii</i>	Japan
<i>Sphaerella weiriana</i>	<i>Castanopsis chrysophylla</i>	California
<i>Sphaerulina myriadea</i>	<i>Castanopsis chrysophylla</i>	(On leaves.) California
	<i>Castanopsis sempervirens</i>	(On leaves.) California
<i>Spiropes harunganae</i>	<i>Castanopsis</i> sp.	China,
<i>Sporidesmiella parva</i>	<i>Castanopsis cuspidata</i> var. <i>sieboldii</i>	Japan
<i>Stachybotrys atra</i> var. <i>microspora</i>	<i>Castanopsis cuspidata</i> var. <i>sieboldii</i>	Japan
<i>Stachybotrys nephrospora</i>	<i>Castanopsis cuspidata</i> var. <i>sieboldii</i>	Japan
<i>Stereum</i> sp.	<i>Castanopsis chrysophylla</i>	California
<i>Strumella macrospora</i>	<i>Castanopsis cuspidate</i>	Japan
<i>Subulispora procurvata</i>	<i>Castanopsis cuspidata</i>	Japan
	<i>Castanopsis cuspidata</i> var. <i>sieboldii</i>	Japan
<i>Sympodiella multiseptata</i>	<i>Castanopsis</i> sp.	Papua New Guinea
<i>Taphrina castanopsidis</i>	<i>Castanopsis chrysophylla</i>	(Leaf blister.) California; Oregon
	<i>Castanopsis sempervirens</i>	California
<i>Taphrina castanopsis</i>	<i>Castanopsis chrysophylla</i>	California
<i>Taphrina kusanoi</i>	<i>Castanopsis cuspidate</i>	Japan
<i>Thozetella cristata</i>	<i>Castanopsis cuspidata</i> var. <i>sieboldii</i>	Japan
<i>Trametes kusanoana</i>	<i>Castanopsis</i> sp.	China
<i>Trechispora farinacea</i>	<i>Castanopsis cuspidate</i>	Japan
<i>Trechispora microspora</i>	<i>Castanopsis cuspidate</i>	Japan
<i>Trechispora mollusca</i>	<i>Castanopsis cuspidata</i>	Japan
<i>Tubulicrinis vinaceus</i>	<i>Castanopsis</i> sp.	Japan
<i>Typhulochaeta japonica</i>	<i>Castanopsis sclerophylla</i>	China
	<i>Castanopsis</i> sp.	China, ,
<i>Valsa coronata</i>	<i>Castanopsis chrysophylla</i>	(On twigs.) Oregon
<i>Volutella ciliata</i>	<i>Castanopsis</i> sp.	Papua New Guinea
<i>Wiesneriomyces javanicus</i>	<i>Castanopsis cuspidate</i>	Japan
	<i>Castanopsis cuspidata</i> var. <i>sieboldii</i>	Japan
<i>Zygosporium parasiticum</i>	<i>Castanopsis</i> sp.	Papua New Guinea

Table A.2 List of fungal records on *Lithocarpus* (Farr *et al.*, 2006).

Fungal species	Host species	Locations
<i>Aleurocorticium candidum</i>	<i>Lithocarpus densiflora</i>	California
<i>Aleurodiscus aurantius</i>	<i>Lithocarpus densiflora</i>	(On dead stems and twigs.) California
<i>Aphanostigme flabellariae</i>	<i>Lithocarpus dealbatus</i>	China
<i>Appendiculella arisanensis</i>	<i>Lithocarpus</i> sp.	China
<i>Appendiculella castanopsifoliae</i>	<i>Lithocarpus castanopsifolius</i>	Taiwan
<i>Appendiculella lithocarpicola</i>	<i>Lithocarpus corneus</i>	China
<i>Appendiculella sinsuiensis</i>	<i>Lithocarpus shinsuiensis</i>	Taiwan
<i>Armillaria gallica</i>	<i>Lithocarpus densiflorus</i>	California
<i>Armillaria mellea</i>	<i>Lithocarpus densiflorus</i>	California
<i>Asteridiella cyclobalanopsidicola</i>	<i>Lithocarpus glaber</i>	China
	<i>Lithocarpus grandifolius</i>	China
	<i>Lithocarpus haipinii</i>	China
	<i>Lithocarpus hancei</i>	China
	<i>Lithocarpus litseifolius</i>	China
	<i>Lithocarpus</i> sp.	China
<i>Asteridiella quercina</i>	<i>Lithocarpus glaber</i>	China
	<i>Lithocarpus hancei</i>	China
	<i>Lithocarpus litseifolius</i>	China
<i>Astrosporina angustifolia</i>	<i>Lithocarpus</i> sp.	Indonesia; Papua New Guinea
<i>Astrosporina bresadolae</i>	<i>Lithocarpus</i> sp.	Papua New Guinea
<i>Astrosporina echinosimilis</i>	<i>Lithocarpus</i> sp.	Papua New Guinea
<i>Astrosporina gemina</i>	<i>Lithocarpus</i> sp.	Papua New Guinea
<i>Astrosporina granulosiceps</i>	<i>Lithocarpus</i> sp.	Papua New Guinea
<i>Astrosporina lasseroides</i>	<i>Lithocarpus</i> sp.	Papua New Guinea
<i>Astrosporina lutea</i>	<i>Lithocarpus</i> sp.	Japan; Papua New Guinea
<i>Astrosporina neoumbrina</i>	<i>Lithocarpus</i> sp.	Japan; Papua New Guinea
<i>Astrosporina olivaceonigra</i>	<i>Lithocarpus</i> sp.	Papua New Guinea
<i>Astrosporina papillata</i>	<i>Lithocarpus</i> sp.	Papua New Guinea
<i>Astrosporina petchii</i>	<i>Lithocarpus</i> sp.	Indonesia; Malaysia; Papua New Guinea
<i>Astrosporina punctatosquamosa</i>	<i>Lithocarpus</i> sp.	Papua New Guinea
<i>Astrosporina titibuensis</i>	<i>Lithocarpus</i> sp.	Indonesia; Japan
<i>Auerswaldiella lithocarpicola</i>	<i>Lithocarpus</i> sp.	Taiwan,
<i>Auricularia auricula</i>	<i>Lithocarpus</i> sp.	Hong Kong
<i>Beltraniella odinae</i>	<i>Lithocarpus edulis</i>	Japan
<i>Bonaria lithocarpi</i>	<i>Lithocarpus densiflorus</i>	California
<i>Bulgaria inquinans</i>	<i>Lithocarpus densiflora</i>	(On wood.) California,
<i>Ceuthocarpon conflictum</i>	<i>Lithocarpus densiflora</i>	(Leaf spot.) California
<i>Chaetasbolisia falcata</i>	<i>Lithocarpus densiflora</i>	California,
<i>Chlorociboria aeruginosa</i>	<i>Lithocarpus densiflora</i>	(Sapwood stain.) California,
<i>Cladosporium microspermum</i>	<i>Lithocarpus viridis</i>	China,
<i>Coccomyces dentatus</i>	<i>Lithocarpus densiflora</i>	(On leaves.) California
<i>Cronartium quercuum</i>	<i>Lithocarpus densiflora</i>	California,
<i>Cystotheca lanestris</i>	<i>Lithocarpus dealbatus</i>	China, ; India
	<i>Lithocarpus henryi</i>	India
<i>Dendrothele candida</i>	<i>Lithocarpus densiflora</i>	(On bark.) California
<i>Diatrype stigma</i>	<i>Lithocarpus densiflora</i>	California
<i>Dimerium elegans</i>	<i>Lithocarpus cuspidata</i>	Japan
	<i>Lithocarpus variolosus</i>	China
<i>Erysiphe trina</i>	<i>Lithocarpus densiflora</i>	(Powdery mildew.) California
<i>Exidiopsis plumbescens</i>	<i>Lithocarpus densiflora</i>	California
<i>Fusarium solani</i>	<i>Lithocarpus densiflora</i>	California

Table A.2 (Continued)

Fungal species	Host species	Locations
<i>Fusicolla foliicola</i>	<i>Lithocarpus densiflora</i>	(On leaves.) California
<i>Fusidium griseum</i>	<i>Lithocarpus densiflora</i>	(On dead leaves.) California
<i>Gnomonia lithocarpicola</i>	<i>Lithocarpus rhombocarpus</i>	Taiwan
<i>Gnomoniella lithocarpicola</i>	<i>Lithocarpus ternaticupula</i>	Taiwan
<i>Helminthosporium dorycarpum</i>	<i>Lithocarpus polystachya</i>	China
<i>Hymenochaete tabacina</i>	<i>Lithocarpus densiflora</i>	California,
<i>Hypocrea albocornea</i>	<i>Lithocarpus</i> sp.	China
<i>Hypocrea aurantia</i>	<i>Lithocarpus</i> sp.	China
<i>Hypoxylon mediterraneum</i>	<i>Lithocarpus densiflora</i>	California
<i>Irene castanopsifoliae</i>	<i>Lithocarpus castanopsifolius</i>	China
<i>Irene lithocarpicola</i>	<i>Lithocarpus amygdalifolius</i>	China; Taiwan
	<i>Lithocarpus lepodocarpa</i>	Taiwan
<i>Irpex tabacinoides</i>	<i>Lithocarpus cuspidata</i>	Japan
<i>Lambertella aurantiaca</i>	<i>Lithocarpus microspermus</i>	China
<i>Limacinia lithocarpi</i>	<i>Lithocarpus densiflora</i>	(On living leaves.) California
<i>Marasmius copelandii</i>	<i>Lithocarpus densiflora</i>	California
<i>Marasmius quercophilus</i>	<i>Lithocarpus densiflora</i>	California
<i>Marasmius thiersii</i>	<i>Lithocarpus</i> sp.	California
<i>Melanopsamma verrucosa</i>	<i>Lithocarpus</i> sp.	California
<i>Meliola kawakamii</i>	<i>Lithocarpus elizabethae</i>	China
	<i>Lithocarpus hancei</i>	China
	<i>Lithocarpus kawakamii</i>	China; Taiwan
<i>Meliola kiraiensis</i>	<i>Lithocarpus castanopsifolius</i>	China
	<i>Lithocarpus lepodocarpa</i>	Taiwan
<i>Meliola kodaihoensis</i>	<i>Lithocarpus kodaihoensis</i>	Taiwan
<i>Meliola lithocarpina</i>	<i>Lithocarpus brevicaudatus</i>	China
<i>Meliola lithocarpina</i> var. <i>mengyangensis</i>	<i>Lithocarpus</i> sp.	China
<i>Meliola quercina</i>	<i>Lithocarpus glaber</i>	China
	<i>Lithocarpus brachystachyus</i>	China
	<i>Lithocarpus fordianus</i>	China
	<i>Lithocarpus grandifolius</i>	China
	<i>Lithocarpus hancei</i>	China
	<i>Lithocarpus handelianus</i>	China
	<i>Lithocarpus</i> sp.	China
	<i>Lithocarpus spicata</i>	China
<i>Meliola ramulicola</i>	<i>Lithocarpus brevicaudatus</i>	China; Taiwan
<i>Meliola shiiae</i>	<i>Lithocarpus</i> sp.	China
<i>Meliola subpellucida</i>	<i>Lithocarpus castanopsifolius</i>	China; Taiwan
<i>Meliola taiyuensis</i>	<i>Lithocarpus corneus</i>	China
<i>Menispora minuta</i>	<i>Lithocarpus edulis</i>	Japan
<i>Microsphaera alni</i>	<i>Lithocarpus densiflora</i>	California
<i>Mollisia lithocarpi</i>	<i>Lithocarpus densiflora</i>	(On dead leaves.) California,
<i>Monochaetia hysteriiformis</i>	<i>Lithocarpus densiflora</i>	(On dead leaves.) California
<i>Odontia fimbriata</i>	<i>Lithocarpus densiflora</i>	California
<i>Odontia subabrupta</i>	<i>Lithocarpus densiflora</i>	(On wood.) California
<i>Omphalotus olivascens</i>	<i>Lithocarpus densiflora</i>	California
<i>Pestalotia castagnei</i>	<i>Lithocarpus densiflora</i>	California
<i>Pestalotia glandicola</i>	<i>Lithocarpus brevicaudatus</i>	Korea
<i>Pestalotia montellica</i>	<i>Lithocarpus densiflora</i>	(Leaf blight.) California,
<i>Pestalotiopsis pseudomontellica</i>	<i>Lithocarpus densiflora</i>	California
<i>Phaeobulgaria inquinans</i>	<i>Lithocarpus densiflora</i>	Oregon
<i>Phaeosaccardinula anomala</i>	<i>Lithocarpus densiflora</i>	(On leaves.) California
<i>Phyllosticta castaneae</i>	<i>Lithocarpus</i> sp.	China,
<i>Phyllosticta roboris</i>	<i>Lithocarpus</i> sp.	China

Table A.2 (Continued)

Fungal species	Host species	Locations
<i>Phyllosticta</i> spp.	<i>Lithocarpus</i> sp.	China
<i>Phytophthora cinnamomi</i>	<i>Lithocarpus densiflora</i>	SE states
<i>Phytophthora nemorosa</i>	<i>Lithocarpus densiflorus</i>	California, ; Oregon
<i>Phytophthora ramorum</i>	<i>Lithocarpus densiflorus</i>	California, ; Oregon,
<i>Polyporus patouillardii</i>	<i>Lithocarpus cuspidata</i>	Japan
<i>Polyporus sulphureus</i>	<i>Lithocarpus sieboldii</i>	Japan
<i>Poria eupora</i>	<i>Lithocarpus densiflora</i>	Oregon
<i>Poria ferrea</i>	<i>Lithocarpus densiflora</i>	California
<i>Poria</i> sp.	<i>Lithocarpus densiflora</i>	California
<i>Poria versipora</i>	<i>Lithocarpus densiflora</i>	California; Oregon
<i>Poria viridans</i>	<i>Lithocarpus densiflora</i>	California
<i>Porothelium fimbriatum</i>	<i>Lithocarpus densiflora</i>	California,
<i>Protopeltis lithocarpi</i>	<i>Lithocarpus densiflora</i>	(On leaves.) California
<i>Schizophyllum commune</i>	<i>Lithocarpus glaber</i>	Hong Kong
<i>Scolecobonaria lithocarpi</i>	<i>Lithocarpus densiflorus</i>	California
<i>Septobasidium bogoriense</i>	<i>Lithocarpus uraianus</i>	China
<i>Sphaerotheca lanestris</i>	<i>Lithocarpus dealbatus</i>	China,
<i>Sphaerotheca lanestris</i>	<i>Lithocarpus densiflora</i>	(Powdery mildew.) California,
<i>Sphaerulina conflicta</i>	<i>Lithocarpus densiflora</i>	California
<i>Steccherinum fimbriatum</i>	<i>Lithocarpus densiflora</i>	California
<i>Stereum gausapatum</i>	<i>Lithocarpus densiflora</i>	California
<i>Stereum hirsutum</i>	<i>Lithocarpus densiflora</i>	California,
<i>Trametes dickinsii</i>	<i>Lithocarpus</i> sp.	Japan
<i>Uleomyces decipiens</i>	<i>Lithocarpus amygdalifolius</i>	Taiwan
	<i>Lithocarpus uraianus</i>	China; Taiwan

Table A.3 List of fungi occurring on *Schima* (after Farr *et al.*, 2006).

Fungal species	Host species	Locations
<i>Coriolus versicolor</i>	<i>Schima</i> sp.	China
<i>Corticium salmonicolor</i>	<i>Schima wallichii</i>	Nepal
<i>Daedaleopsis confragosa</i>	<i>Schima</i> sp.	China
<i>Meliola schimicola</i>	<i>Schima</i> sp.	China
	<i>Schima superba</i>	China; Taiwan
<i>Meliola theacearum</i>	<i>Schima khasiana</i>	China
	<i>Schima parviflora</i>	China
	<i>Schima</i> sp.	China
	<i>Schima superba</i>	China
<i>Polystictus versicolor</i>	<i>Schima</i> sp.	China

Table A.4 List of fungi occurring on *Syzygium* (after Farr *et al.*, 2006).

Fungal species	Host species	Locations
<i>Amazonia syzygii</i>	<i>Syzygium cumini</i>	India,
	<i>Syzygium</i> sp.	India
<i>Anungitea antillana</i>	<i>Syzygium jambos</i>	Cuba
<i>Anungitea caespitosa</i>	<i>Syzygium cordatum</i>	South Africa
<i>Anungitea syzygii</i>	<i>Syzygium cordatum</i>	South Africa
<i>Armillaria mellea</i>	<i>Syzygium gerrardii</i>	South Africa; Southern Africa
<i>Armillariella mellea</i>	<i>Syzygium gerrardii</i>	South Africa
<i>Armillariella tabescens</i>	<i>Syzygium cumini</i>	(Root rot.) Florida
	<i>Syzygium jambos</i>	(Root rot.) Florida
<i>Arthrobotryum</i> sp.	<i>Syzygium cumini</i>	China
<i>Aspergillus fumigatus</i>	<i>Syzygium aromaticum</i>	Malaysia
<i>Aspergillus niger</i>	<i>Syzygium aromaticum</i>	Malaysia
<i>Asteridiella hughesii</i>	<i>Syzygium guineense</i>	Ghana
<i>Asteridiella ohiana</i> var. <i>major</i>	<i>Syzygium claviflorum</i>	China; India, ,
<i>Asteridiella syzygii</i>	<i>Syzygium bullockii</i>	China
	<i>Syzygium</i> sp.	China
<i>Asterina claviflora</i>	<i>Syzygium claviflorum</i>	India,
<i>Asterina gopalkrishnanii</i>	<i>Syzygium cumini</i>	India
<i>Asterina pemphidioides</i>	<i>Syzygium gerrardii</i>	Southern Africa
<i>Asterina quarta</i>	<i>Syzygium jambolanum</i>	Indonesia
<i>Asterina syzygii</i>	<i>Syzygium cordatum</i>	Uganda
	<i>Syzygium gerrardii</i>	Africa; South Africa, ; Southern Africa
<i>Asterina syzygii</i> var. <i>microspora</i>	<i>Syzygium guineense</i>	Sierra Leone
<i>Asterina syzygiicola</i>	<i>Syzygium cordatum</i>	Uganda
<i>Asterinella puiggarii</i>	<i>Syzygium cumini</i>	(Leaf spot.) Florida
	<i>Syzygium jambos</i>	(Anthracnose.) Florida
<i>Atichia millardetii</i>	<i>Syzygium</i> sp.	Hawaii
<i>Basidioidendron eyrei</i>	<i>Syzygium jambos</i>	Hawaii
<i>Beltrania mundkurii</i>	<i>Syzygium cumini</i>	India
<i>Botryodiplodia theobromae</i>	<i>Syzygium aromaticum</i>	Indonesia; Malaysia
	<i>Syzygium samarangense</i>	China
<i>Botryodiplodia variispora</i>	<i>Syzygium cumini</i>	India
<i>Botryosphaeria ribis</i>	<i>Syzygium cumini</i>	Florida
<i>Bourdotia kipukaensis</i>	<i>Syzygium jambos</i>	Hawaii
	<i>Syzygium malaccense</i>	Hawaii
<i>Brooksia tropicalis</i>	<i>Syzygium</i> sp.	New Guinea; Papua New Guinea
<i>Calonectria colhounii</i>	<i>Syzygium aromaticum</i>	Florida; Hawaii; Louisiana; North Carolina Oregon; South Carolina; Virginia
<i>Calonectria kytensis</i>	<i>Syzygium aromaticum</i>	Malaysia
<i>Calonectria leguminum</i>	<i>Syzygium aromaticum</i>	Brazil
<i>Calonectria reteaudii</i>	<i>Syzygium aromaticum</i>	Indonesia
<i>Canalisporium caribense</i>	<i>Syzygium jambos</i>	Cuba
<i>Capnodium annonae</i>	<i>Syzygium floribundum</i>	Australia
<i>Capnodium</i> sp.	<i>Syzygium jambos</i>	Brazil; Cuba; Venezuela
<i>Capnodium</i> sp.	<i>Syzygium malaccense</i>	Cuba
<i>Cercospora eugeniae</i>	<i>Syzygium samarangense</i>	China
<i>Cercospora</i> sp.	<i>Syzygium cumini</i>	(Leaf spot.) Florida -
<i>Cercospora syzygii</i>	<i>Syzygium cumini</i>	India
	<i>Syzygium jambos</i>	India
<i>Ceriporia alachuana</i>	<i>Syzygium cumini</i>	Hawaii

Table A.4 (Continued)

Fungal species	Host species	Locations
<i>Ceriporia xylostromatoides</i>	<i>Syzygium jambos</i>	Hawaii
<i>Chaetomella circinoseta</i>	<i>Syzygium jambos</i>	India,
<i>Chaetomella raphigera</i>	<i>Syzygium jambos</i>	India
<i>Chaetospermum chaetosporum</i>	<i>Syzygium cordatum</i>	South Africa
<i>Chaetothyrium syzygii</i>	<i>Syzygium cordatum</i>	Southern Africa
<i>Chaetothyrium wormersleyi</i>	<i>Syzygium</i> sp.	Papua New Guinea
<i>Ciferriusia wormersleyi</i>	<i>Syzygium</i> sp.	New Guinea
<i>Ciliochorella eucalypti</i>	<i>Syzygium cumini</i>	India
<i>Ciliochorella indica</i>	<i>Syzygium cumini</i>	India
<i>Ciliochorella mangiferae</i> ;	<i>Syzygium cumini</i>	Hawaii; India
<i>Cladosporium myrticola</i>	<i>Syzygium jambos</i>	Cuba
<i>Coccomyces urceoloides</i>	<i>Syzygium</i> sp.	Malaysia
<i>Cochliobolus lunatus</i>	<i>Syzygium aromaticum</i>	Malaysia
<i>Colletotrichum gloeosporioides</i>	<i>Syzygium aromaticum</i>	Indonesia
	<i>Syzygium cumini</i>	(Anthracnose.) Florida
	<i>Syzygium jambos</i>	(Anthracnose.) Florida
	<i>Syzygium samarangense</i>	China
<i>Coltricia laeta</i>	<i>Syzygium maire</i>	New Zealand
<i>Coniella castaneicola</i>	<i>Syzygium aromaticum</i>	Indonesia
<i>Coniothyrium fuckelii</i>	<i>Syzygium malaccense</i>	Venezuela
<i>Corynespora cassiicola</i>	<i>Syzygium jambolanum</i>	India
<i>Corynesporopsis rionensis</i>	<i>Syzygium jambos</i>	Cuba
<i>Crepidotus alabamensis</i>	<i>Syzygium jambos</i>	Hawaii
<i>Crepidotus amygdalosporus</i>	<i>Syzygium malaccense</i>	Hawaii
<i>Crepidotus roseus</i> var.	<i>Syzygium cumini</i>	Hawaii
<i>boninensis</i>		
<i>Cryphonectria cubensis</i>	<i>Syzygium aromaticum</i>	Zanzibar
<i>Curvularia lunata</i>	<i>Syzygium aromaticum</i>	Malaysia
<i>Cylindrocladium floridanum</i>	<i>Syzygium aromaticum</i>	Malaysia
<i>Cylindrocladium</i>	<i>Syzygium aromaticum</i>	Indonesia
<i>quinqueseptatum</i>		
<i>Cytospora acaciae</i>	<i>Syzygium cumini</i>	India
<i>Dacrymyces deliquescens</i> var.	<i>Syzygium jambos</i>	Hawaii
<i>deliquescens</i>		
<i>Dictyochaeta assamica</i>	<i>Syzygium jambos</i>	Cuba
<i>Diplodia natalensis</i>	<i>Syzygium jambos</i>	China
<i>Diplodia variispora</i>	<i>Syzygium cordatum</i>	Southern Africa
<i>Discosia artocreas</i>	<i>Syzygium cumini</i>	India
<i>Discosia bombycina</i>	<i>Syzygium cumini</i>	India,
<i>Discosia syzygii</i>	<i>Syzygium cumini</i>	India,
<i>Echidnodella syzygimae</i>	<i>Syzygium cumini</i>	India
<i>Ellisembia adscendens</i>	<i>Syzygium jambos</i>	Cuba
<i>Ellisembia vaga</i>	<i>Syzygium jambos</i>	Cuba
<i>Endothia eugeniae</i>	<i>Syzygium aromaticum</i>	Indonesia
<i>Euryachora syzygii</i>	<i>Syzygium</i> sp.	Congo
<i>Exidiopsis calcea</i>	<i>Syzygium cumini</i>	Hawaii
<i>Flavodon cervinogilvum</i>	<i>Syzygium jambos</i>	Hawaii
<i>Fomes rimosus</i>	<i>Syzygium gerrardii</i>	South Africa; Southern Africa
<i>Fomes senex</i>	<i>Syzygium gerrardii</i>	South Africa, ; Southern Africa
<i>Fomitopsis nivosa</i>	<i>Syzygium cumini</i>	Hawaii
<i>Fusarium</i> sp.	<i>Syzygium jambos</i>	(Root rot.) Florida
<i>Ganoderma australe</i>	<i>Syzygium malaccense</i>	Hawaii
<i>Glomerella cingulata</i>	<i>Syzygium cumini</i>	(Anthracnose.) Florida
	<i>Syzygium jambos</i>	(Anthracnose.) Florida; Hong Kong

Table A.4 (Continued)

Fungal species	Host species	Locations
<i>Grallomyces portoricensis</i>	<i>Syzygium</i> sp.	New Guinea; Papua New Guinea
<i>Hadronema verrucosum</i>	<i>Syzygium cumini</i>	India
<i>Haplotrichum curtisii</i>	<i>Syzygium cumini</i>	Hawaii,
<i>Harknessia syzygii</i>	<i>Syzygium cordatum</i>	South Africa,
<i>Helicoma ambiens</i>	<i>Syzygium jambos</i>	Cuba
<i>Helicoma palmigenum</i>	<i>Syzygium jambos</i>	Cuba
<i>Helicomycetes roseus</i>	<i>Syzygium jambos</i>	Cuba
<i>Hendersonia syzygii</i>	<i>Syzygium cumini</i>	India,
<i>Henningsomyces candidus</i>	<i>Syzygium jambos</i>	Hawaii,
<i>Hermatomyces sphaericum</i>	<i>Syzygium jambos</i>	Cuba
<i>Hymenochaete anomala</i>	<i>Syzygium jambos</i>	Hawaii
<i>Hyphoderma argillaceum</i>	<i>Syzygium jambos</i>	Hawaii,
<i>Hyphoderma nudicephalum</i>	<i>Syzygium cumini</i>	Hawaii,
	<i>Syzygium jambos</i>	Hawaii,
<i>Hyphoderma pallidum</i>	<i>Syzygium jambos</i>	Hawaii,
<i>Hyphoderma praetermissum</i>	<i>Syzygium jambos</i>	Hawaii,
<i>Hyphodontia nesporei</i>	<i>Syzygium jambolanum</i>	Hawaii
<i>Hyphopolynema stilboideum</i>	<i>Syzygium</i> sp.	India
<i>Hypochnicium lundellii</i>	<i>Syzygium malaccense</i>	Hawaii
<i>Hypochnicium punctulatum</i>	<i>Syzygium jambos</i>	Hawaii
<i>Idiocercus indica</i>	<i>Syzygium cumini</i>	India
<i>Idiocercus longipedicellata</i>	<i>Syzygium montagnum</i>	India
<i>Idiocercus variispora</i>	<i>Syzygium cumini</i>	India
<i>Intercalarispora nigra</i>	<i>Syzygium jambos</i>	Cuba
<i>Lachnella conferta</i>	<i>Syzygium jambos</i>	Hawaii,
<i>Lecanostictopsis syzygii</i>	<i>Syzygium cordatum</i>	South Africa
<i>Lembosia syzygiicola</i>	<i>Syzygium cordatum</i>	Uganda
	<i>Syzygium guineense</i>	Sierra Leone; Tanzania
	<i>Syzygium</i> sp.	Sudan
<i>Lentinus bertieri</i>	<i>Syzygium jambos</i>	Hawaii
<i>Leptopeltis</i> sp.	<i>Syzygium jambos</i>	Cuba
<i>Leptosporomyces ovoideus</i>	<i>Syzygium jambos</i>	Hawaii
<i>Limacinula samoensis</i>	<i>Syzygium aromaticum</i>	Indonesia
<i>Macrophoma</i> sp.	<i>Syzygium caryophyllatum</i>	India
<i>Meliola densa</i>	<i>Syzygium munroni</i>	India
<i>Meliola eugeniae-jamboloidis</i>	<i>Syzygium jambos</i>	China
	<i>Syzygium munroni</i>	India
<i>Meliola eugeniae-jamboloidis</i>	<i>Syzygium jambos</i>	India
var. <i>amphigena</i>		
<i>Meliola eugeniae-jamboloidis</i>	<i>Syzygium claviflorum</i>	China
var. <i>eugeniae-jamboloidis</i>	<i>Syzygium jambos</i>	China
	<i>Syzygium myrsinifolium</i>	China
<i>Meliola maduraiensis</i>	<i>Syzygium lanceolatum</i>	India
<i>Meliola myrtacearum</i>	<i>Syzygium hancei</i>	China
<i>Meliola pulchella</i> var. <i>syzygii</i>	<i>Syzygium laetum</i>	India
<i>Meliola singaporensis</i>	<i>Syzygium cumini</i>	China
<i>Meliolina africana</i>	<i>Syzygium gerrardii</i>	South Africa
	<i>Syzygium legatii</i>	South Africa
<i>Meliolina arborescens</i>	<i>Syzygium cumini</i>	India
	<i>Syzygium gerrardii</i>	South Africa; Southern Africa,
	<i>Syzygium legatii</i>	Southern Africa

Table A.4 (Continued)

Fungal species	Host species	Locations
<i>Meliolina mollis</i>	<i>Syzygium cumini</i>	India
<i>Meliolina pulcherrima</i>	<i>Syzygium jambolanum</i>	India
<i>Metasphaeria mahabaleshwarensis</i>	<i>Syzygium cumini</i>	India
<i>Micronectria syzygii</i>	<i>Syzygium guineense</i>	East Africa
<i>Mycena alphetophora</i>	<i>Syzygium cumini</i>	Hawaii
<i>Mycohypallage congesta</i>	<i>Syzygium cumini</i>	India
	<i>Syzygium jambolanum</i>	India
<i>Mycosphaerella marasasii</i>	<i>Syzygium cordatum</i>	South Africa,
<i>Mycosphaerella syzygii</i>	<i>Syzygium cordatum</i>	South Africa
<i>Mycotribulus mirabilis</i>	<i>Syzygium cordatum</i>	South Africa
<i>Neobarclaya congesta</i>	<i>Syzygium cordatum</i>	Southern Africa, ,
<i>Neocapnodium tanakae</i>	<i>Syzygium samarangense</i>	China
<i>Oramasia hirsuta var. cubensis</i>	<i>Syzygium jambos</i>	Cuba
<i>Paecilomyces variotii</i>	<i>Syzygium aromaticum</i>	Malaysia
<i>Paraulocladium angulosporum</i>	<i>Syzygium jambos</i>	Cuba
<i>Passalora syzygii</i>	<i>Syzygium heyneanum</i>	India
<i>Pellicularia koleroga</i>	<i>Syzygium aromaticum</i>	Brazil
<i>Peniophora cinerea</i>	<i>Syzygium jambos</i>	Hawaii
<i>Penzigomyces parvus</i>	<i>Syzygium jambos</i>	Cuba
<i>Perenniporia tephropora</i>	<i>Syzygium cumini</i>	Hawaii
<i>Periconia byssoides</i>	<i>Syzygium jambos</i>	Cuba
<i>Periconia doidgeae</i>	<i>Syzygium gerrardii</i>	Southern Africa
<i>Perisporiopsis quinqueseptata</i>	<i>Syzygium buxifolii</i>	Japan
<i>Pestalospaeria hansenii</i>	<i>Syzygium cordatum</i>	South Africa -
<i>Pestalotia carissae</i>	<i>Syzygium cumini</i>	India
<i>Pestalotia congensis</i>	<i>Syzygium jambolanum</i>	Sri Lanka
<i>Pestalotia congesta</i>	<i>Syzygium jambolanum</i>	Sri Lanka
<i>Pestalotia eugeniae</i>	<i>Syzygium cumini</i>	India
<i>Pestalotia sp.</i>	<i>Syzygium cumini</i>	India
<i>Pestalotia versicolor</i>	<i>Syzygium aromaticum</i>	Brazil
<i>Pestalotiopsis disseminata</i>	<i>Syzygium cumini</i>	India
<i>Pestalotiopsis guepinii</i>	<i>Syzygium jambolanum</i>	India
<i>Pestalotiopsis palmarum</i>	<i>Syzygium samarangense</i>	China
<i>Pestalotiopsis versicolor</i>	<i>Syzygium cumini</i>	India
<i>Phaeochaetia syzygii</i>	<i>Syzygium sp.</i>	South Africa
<i>Phaeocliospora eugeniae</i>	<i>Syzygium cumini</i>	India
<i>Phaeophragmeriella transvaalensis</i>	<i>Syzygium gerrardii</i>	Southern Africa
<i>Phanerochaete australis</i>	<i>Syzygium cumini</i>	Hawaii
	<i>Syzygium jambos</i>	Hawaii
<i>Phanerochaete maiuensis</i>	<i>Syzygium jambos</i>	Hawaii
<i>Phellinus noxius</i>	<i>Syzygium samarangense</i>	Taiwan
<i>Phellinus robiniae</i>	<i>Syzygium gerrardii</i>	South Africa
<i>Phlebia acanthocystis</i>	<i>Syzygium cumini</i>	Hawaii
	<i>Syzygium jambos</i>	Hawaii
<i>Phlebia coccineofulva</i>	<i>Syzygium jambos</i>	Hawaii
<i>Phlebiella tulasnelloidea</i>	<i>Syzygium jambos</i>	Hawaii
<i>Phlebiopsis peniophoroides</i>	<i>Syzygium jambos</i>	Hawaii
<i>Phlebiopsis ravenelii</i>	<i>Syzygium jambos</i>	Hawaii
<i>Phomopsis eugeniae</i>	<i>Syzygium cumini</i>	India
<i>Phomopsis pustulata</i>	<i>Syzygium cumini</i>	India
<i>Phomopsis syzygii</i>	<i>Syzygium samarangense</i>	China
<i>Phyllachora ambigua</i>	<i>Syzygium cumini</i>	India,
<i>Phyllachora bella</i>	<i>Syzygium paniculatum</i>	Australia

Table A.4 (Continued)

Fungal species	Host species	Locations
<i>Phyllosticta eugeniae</i>	<i>Syzygium cumini</i>	(Leaf spot.) Florida
	<i>Syzygium jambos</i>	(Leaf spot.) Florida
<i>Phyllosticta</i> sp.	<i>Syzygium jambos</i>	Cuba
<i>Phyllosticta syzygii</i>	<i>Syzygium aromaticum</i>	Indonesia
<i>Physalidium carbonis</i>	<i>Syzygium jambos</i>	Cuba
<i>Phytophthora palmivora</i>	<i>Syzygium paniculatum</i>	Florida
<i>Polyporus arcularius</i>	<i>Syzygium jambos</i>	Hawaii
<i>Polyporus gilvus</i>	<i>Syzygium guineense</i>	Southern Africa
<i>Pseudocercospora marasasii</i>	<i>Syzygium cordatum</i>	South Africa,
<i>Pseudocercospora myrtacearum</i>	<i>Syzygium cumini</i>	Nepal
	<i>Syzygium jambolanum</i>	India
<i>Pseudocercospora syzygiicola</i>	<i>Syzygium cordatum</i>	South Africa
<i>Pseudocercospora syzygiigena</i>	<i>Syzygium guineense</i>	Zambia
<i>Pseudocercospora myrtacearum</i>	<i>Syzygium heyneanum</i>	India
<i>Puccinia psidii</i>	<i>Syzygium jambos</i>	Brazil ; Colombia, , ,
	<i>Syzygium</i> sp.	Costa Rica
<i>Pucciniopsis</i> sp.	<i>Syzygium cordatum</i>	Southern Africa
<i>Pycnoporus sanguineus</i>	<i>Syzygium jambos</i>	Hawaii
<i>Pythium vexans</i>	<i>Syzygium aromaticum</i>	Malaysia
<i>Rigidoporus vinctus</i>	<i>Syzygium jambos</i>	Hawaii
<i>Robillardia sessilis</i>	<i>Syzygium cumini</i>	India
<i>Sclerotinia</i> sp.	<i>Syzygium paniculatum</i>	California
<i>Scoliosporium syzygii</i>	<i>Syzygium guineense</i>	Africa; East Africa
<i>Scopuloides rimosa</i>	<i>Syzygium cumini</i>	Hawaii
	<i>Syzygium malaccense</i>	Hawaii
<i>Sebacina podlachica</i>	<i>Syzygium malaccense</i>	Hawaii
<i>Selenophoma eugeniae</i>	<i>Syzygium cerasoideum</i>	India
<i>Selenophoma kamatii</i>	<i>Syzygium cumini</i>	India
<i>Septogloeum punctatum</i>	<i>Syzygium cordatum</i>	Southern Africa
<i>Septoria syzygii</i>	<i>Syzygium guineense</i>	Ethiopia
<i>Sphaereloma</i> sp.	<i>Syzygium paniculatum</i>	Florida
<i>Sphaerognomonia syzygii</i>	<i>Syzygium jambolanum</i>	Brazil
<i>Spiropes</i> sp.	<i>Syzygium cumini</i>	China
<i>Sporidesmium tropicale</i>	<i>Syzygium jambos</i>	Cuba
<i>Stachybotrys nilagirica</i>	<i>Syzygium jambos</i>	Cuba
<i>Stagonospora atriplicis</i>	<i>Syzygium guineense</i>	Southern Africa; Zimbabwe
<i>Steccherinum rawakense</i>	<i>Syzygium cumini</i>	Hawaii
<i>Stemphyliomma heterochromum</i>	<i>Syzygium jambos</i>	China
<i>Stenella</i> sp.	<i>Syzygium heyneanum</i>	India
<i>Stereum hirsutum</i>	<i>Syzygium cumini</i>	Hawaii
<i>Subulicystidium longisporum</i>	<i>Syzygium jambos</i>	Hawaii
<i>Tetraploa aristata</i>	<i>Syzygium cordatum</i>	Southern Africa
	<i>Syzygium jambos</i>	Cuba
<i>Thanatephorus cucumeris</i>	<i>Syzygium aromaticum</i>	Malaysia
<i>Thaxteriella eugeniae</i>	<i>Syzygium cumini</i>	India
<i>Tinctoporellus epimiltinus</i>	<i>Syzygium jambos</i>	Hawaii
<i>Tomentella chlorina</i>	<i>Syzygium jambos</i>	Hawaii
<i>Torula herbarum f. quaternella</i>	<i>Syzygium jambos</i>	Cuba
<i>Trametes cervina</i>	<i>Syzygium cumini</i>	Hawaii
<i>Trechispora farinacea</i>	<i>Syzygium jambos</i>	Hawaii
<i>Tretospeira ugandensis</i>	<i>Syzygium jambos</i>	Cuba
<i>Tripaspermum juglandis</i>	<i>Syzygium jambos</i>	Cuba
<i>Tripaspermum roupalae</i>	<i>Syzygium jambos</i>	Venezuela
<i>Tripaspermum</i> sp.	<i>Syzygium aromaticum</i>	Malaysia

Table A.4 (Continued)

Fungal species	Host species	Locations
<i>Tubeufia aciculospora</i>	<i>Syzygium buxifolii</i>	Japan
<i>Uredo neurophila</i>	<i>Syzygium jambos</i>	Colombia
<i>Vesiculomyces epitheloides</i>	<i>Syzygium cumini</i>	Hawaii
	<i>Syzygium jambos</i>	Hawaii
<i>Xenasma rimicola</i>	<i>Syzygium jambos</i>	Hawaii
<i>Xenosporium berkeleyi</i>	<i>Syzygium jambos</i>	Cuba
<i>Zygosporium oscheoides</i>	<i>Syzygium jambos</i>	Cuba
<i>Ophiodothella syzygii</i>	<i>Syzygium suborbiculare</i>	Australia
<i>Phomopsis syzygiicola</i>	<i>Syzygium szemaoense</i>	China

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Table B.1 (Continued)

Code	Castanopsis acuminatissima						Castanopsis diversifolia						Lithocarpus polystachyus						Schima wallitchii						Syzygium albiflorum					
	P	TS	TM	DS	DM	P	P	TS	TM	DS	DM	P	P	TS	TM	DS	DM	P	P	TS	TM	DS	DM	P	P	TS	TM	DS	DM	
<i>Dictyochaeta simplex</i>	1			3	1		3	1		6	4		4	2	1		2		1			2								
<i>Dictyochaeta</i> sp.	1	2		4			2	1	2	2	2		4	2	5															
<i>Dictyoarthrinium africanum</i>									1																					
<i>Didymella</i> sp.																														
<i>Emarcea castanopsidicola</i>							8		8																2					
<i>Gaeumannomyces</i> sp.	3		3																											
<i>Gliocladium cylindrosporium</i>									1	2	2																			
<i>Gnomonia amoena</i>							2	1	3	2	4																			
<i>Gnomonia gromoni</i>																														
<i>Hansfordia pulvinata</i>																														
<i>Helicospora big spores</i>							1		1																					
<i>Helicosporium talbotii</i>																														
<i>Idiella</i> sp.	2	3		4	3		6	4	6				4												1					
<i>Idriella fertilis</i>							1		1																					
<i>Iodosphaeria</i> sp.																														
<i>Kionochaeta spissa</i>	5			9	1		2		5																					
<i>Kramasamuha sibirica</i>									1																					
<i>Lauriomyces</i> sp.	1			1	2																									
<i>Lecanicillium lecanii</i>																														
<i>Linocarpon</i> sp. 1208	2																													
<i>Linocarpon verminosum</i>							5		1																					
<i>Linocarpori</i> sp.																														
<i>Lophodermium australiensis</i>																														
<i>Lophodermium</i> sp.																														
<i>Marasmius</i> sp.																														
<i>Menisporopsis nova-zealandae</i>																														

Table B.1 (Continued)

Code	Castanopsis acuminatissima				Castanopsis diversifolia				Lithocarpus polystachyus				Schima wallichii				Syzygium albiflorum			
	P	TS	TM	DS	P	TS	TM	DS	P	TS	TM	DS	P	TS	TM	DS	P	TS	TM	DS
<i>Microdochium phragmitis</i>				3																
<i>Microthyrium</i> sp.	3																			
<i>Microthyrium</i> sp.		1																		
<i>Mycena</i> sp. 2		2																		
<i>Mycena</i> sp.1.				1																
<i>Neolinocarpon</i> sp.								1												
<i>Oidiodendron</i> sp.								2												
<i>Ophioceras commune</i>	6	6			9	6			2	2										1
<i>Periconia cookei</i>						1														
<i>Periconia dolusa</i>					1															
<i>Phialocephala</i> sp						1														
<i>Pseudohatonectria</i> sp.	2																			
<i>Pseudomassarina</i> sp.					1															
<i>Pyricularia</i> sp.						1														
<i>Rhinocladiella selenoides</i>																				1
<i>Setenosporella falcate</i>	1																			2
<i>Setenosporella</i> sp. 1								2												3
<i>Speiropsis pedatospora</i>					1															
<i>Stemonitis</i> sp.						1														
<i>Stictis</i> sp.																				3
<i>Stictis</i> sp1=A062	2								1	1										
<i>Subramaniomyces fusisaprophyticus</i>	4	5			3	2	4		2	1										1
<i>Subulisporium procurvata</i>	8	3	4	3	11	3	12	5	5	3										3
<i>Thozetella</i> sp.																				
<i>Volutella</i> sp.					2				2	3	1									

Table B.1 (Continued)

Code	<i>Castanopsis acuminatissima</i>				<i>Castanopsis diversifolia</i>				<i>Lithocarpus polystachyus</i>				<i>Schima wallichii</i>				<i>Syzygium albiflorum</i>											
	P	TS	TM	DS	DM	P	TS	TM	DS	DM	P	TS	TM	DS	DM	P	TS	TM	DS	DM	P	TS	TM	DS	DM			
<i>Wiesneromyces javanicus</i>																												
MRC0115																												
<i>Xenocyliodrocladium</i> sp.	1																											
MRC0085																												

Table B.2 List of fungi found on studied hosts in 2004-2005.

Code	<i>Castanopsis acuminatissima</i>				<i>Castanopsis diversifolia</i>				<i>Lithocarpus polystachyus</i>				<i>Schima wallichii</i>				<i>Syzygium albiflorum</i>											
	P	TS	TM	DS	DM	P	TS	TM	DS	DM	P	TS	TM	DS	DM	P	TS	TM	DS	DM	P	TS	TM	DS	DM			
<i>Acremonium zonatum</i>																												
MRC0104																												
<i>Albionectria albosuccinae</i>																												
MRC0015																												
<i>Anthostomella</i> sp.																												
MRC0124																												
<i>Arhachandra cristaspora</i>																												
MRC0021																												
<i>Arthriniium</i> sp.																												
MRC0003																												
<i>Arthrowallemia</i> sp. nov.																												
MRC0166																												
<i>Asco</i> 1308 01																												
MRC0140																												
<i>Asco</i> 1308 02																												
MRC0139																												
<i>Beltrania mangifera</i>																												
MRC0051																												
<i>Beltrania rhombica</i>																												
MRC0020																												
<i>Beltraniella ondinae</i>																												
MRC0106																												
<i>Beltraniella portoricensis</i>																												
MRC0023																												
<i>Cercosporula</i> sp.																												
MRC0162																												
<i>Chalara pteridina</i>																												
MRC0167																												
<i>Choetosphaeria</i>																												
MRC0136																												
<i>Circinotricum</i> sp.																												
MRC0151																												

Table B.2 (Continued)

Code	<i>Castanopsis acuminatissima</i>				<i>Castanopsis diversifolia</i>				<i>Lithocarpus polystachyus</i>				<i>Schima wallichii</i>				<i>Syzygium albiflorum</i>			
	P	TS	TM	DS	P	TS	TM	DS	P	TS	TM	DS	P	TS	TM	DS	P	TS	TM	DS
<i>Cladosporium</i> sp.								2												
<i>Coelomycete</i> sp. 10																1				
<i>Coelomycete</i> sp. 2					2	1	1													
<i>Coelomycete</i> sp. 3									2	1	1	1								
<i>Coelomycete</i> sp. 4																1				
<i>Coelomycete</i> sp. 5																1				
<i>Coelomycete</i> sp. 6					1															
<i>Coelomycete</i> sp. 7																				
<i>Coelomycete</i> sp. 8																				
<i>Coelomycete</i> sp. 9																				
<i>Coelomycetes</i> sp. 11					1															
<i>Coelomycetes</i> sp. 12																				
<i>Cryptophiale adagawae</i>					1															
<i>Cylindrocladium gracile</i>																				
<i>Cylindrocladium pseudogracile</i>																				
<i>Cylindrum griseum</i>																				
<i>Diaporthe</i> sp.																				
<i>Dictyochaeta cylindrospora</i>					1															
<i>Dictyochaeta heteroderae</i>																				
<i>Dictyochaeta pakhalensis</i>																				
<i>Dictyochaeta simplex</i>					1	1	1													
<i>Dictyochaeta</i> sp. nov. 1					2	2	4													
<i>Dictyochaeta</i> sp. nov. 3																				
<i>Dictyochaeta stiptocolla</i>																				
<i>Diademella</i> sp.																				
<i>Discomycete</i> sp. 1																				
<i>Discomycete</i> sp. 2																				

Table B.2 (Continued)

Code	Castanopsis acuminatissima						Castanopsis diversifolia						Lithocarpus polystachyus						Schima wallichii						Syzygium albiflorum									
	P	TS	TM	DS	DM	P	P	TS	TM	DS	DM	P	P	TS	TM	DS	DM	P	P	TS	TM	DS	DM	P	P	TS	TM	DS	DM					
<i>Drechslera cynodontis</i>																																		
<i>Emareea castanopsidicola</i>																																		
<i>Gaeumannomyces</i> sp.	2					3																												
<i>Geotrichum candidum</i>																																		
<i>Gliocladium cylindrosporium</i>																																		
<i>Gnomonia amoena</i>																																		
<i>Gnomonia gromon</i>																																		
<i>Helicospora big spores</i>																																		
<i>Helicospora</i> sp.																																		
<i>Helicosporium talbotii</i>																																		
<i>Hypomyceete</i> sp.																																		
<i>Idriella</i> sp.	2	1																																
<i>Idriella fertilis</i>																																		
<i>Kionochaeta spissa</i>																																		
<i>Kionochaeta spissa</i>																																		
<i>Lasiosphaeria</i> sp.																																		
<i>Lasiosphaeria</i> sp.																																		
<i>Lasiosphaeria</i> sp.																																		
<i>Lasiosphaeria</i> sp.																																		
<i>Lauromyces</i> sp.	1																																	
<i>Lecanicillium lecanii</i>																																		
<i>Lichenopeltella salicis</i>																																		
<i>Linocarpon</i> sp.	1																																	
<i>Linocarpon verminosum</i>																																		
<i>Linocarpon</i> sp.																																		
<i>Lophodermium australiensis</i>																																		
<i>Lophodermium</i> sp.																																		

Table B.2 (Continued)

Code	Castanopsis acuminatissima				Castanopsis diversifolia				Lithocarpus polystachyus				Schima wallichii				Syzygium albiflorum			
	P	TS	TM	DS	P	TS	TM	DS	P	TS	TM	DS	P	TS	TM	DS	P	TS	TM	DS
<i>Marasmius</i> sp.				1																
<i>Menisporopsis nova-zealandae</i>																				
<i>Microdochium phragmitis</i>								1												
<i>Microthyrium</i> sp.				1																
<i>Microthyrium</i> sp.								1												
<i>Mycena</i> sp. 2				2																
<i>Mycena</i> sp. 1.								1												
<i>Neolinocarpon</i> sp.												2								
<i>Niesslia</i> sp.								1												
<i>Otidodendron</i> sp.																				1
<i>Ophioceras commune</i>								6				1				1				
<i>Periconia cookii</i>				6				2				1								
<i>Phialocephala</i> sp.												1								
<i>Pseudohalonestria</i> sp.				2																
<i>Rhinocladiella selenoides</i>																				1
<i>Selenosporella falcate</i>																				1
<i>Selenosporella</i> sp. 1																				5
<i>Selenosporella</i> sp. 3												1								6
<i>Speiropsis pedatospora</i>								1												
<i>Stictis</i> sp.				1																2
<i>Stictis</i> sp. 1																				
<i>Subramaniomyces fusisaprophyticus</i>																				
<i>Subulispora hareae</i>																				
<i>Subulisporium procurvata</i>								7				2				13				5
<i>Terriera</i> sp.																				
<i>Thozetella</i> sp.																				

APPENDIX C

Plates of some fungi found on leaf litters in the study

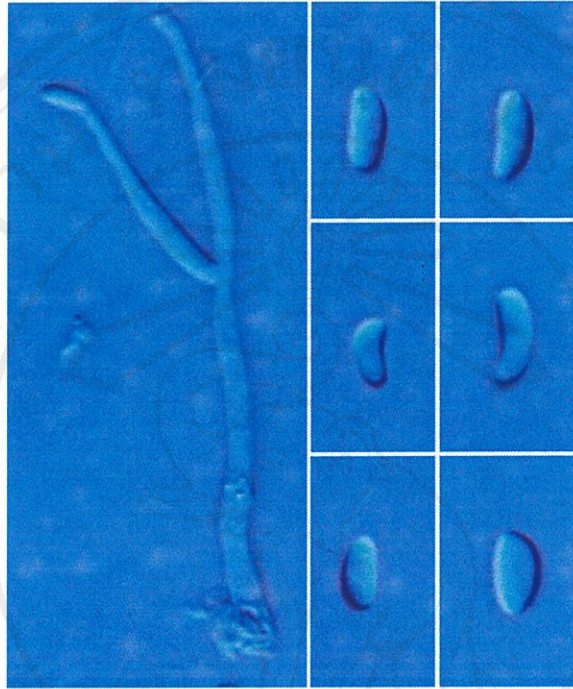


Figure C.1 *Acremonium zonatum*.

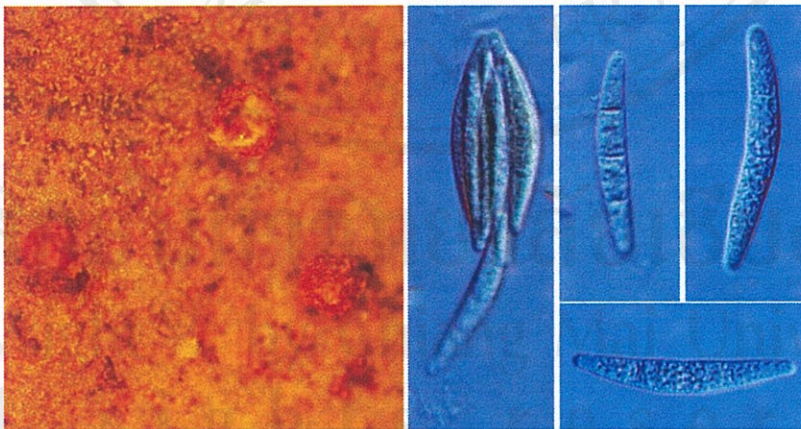


Figure C.2 *Albonectrina albosuccinae*.

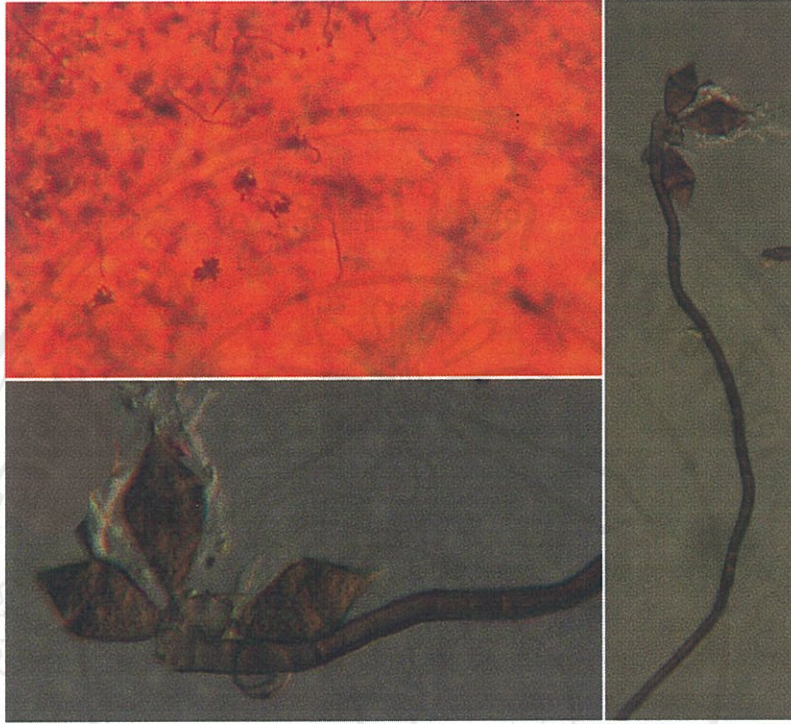


Figure C.3 *Beltrania mangiferae*.

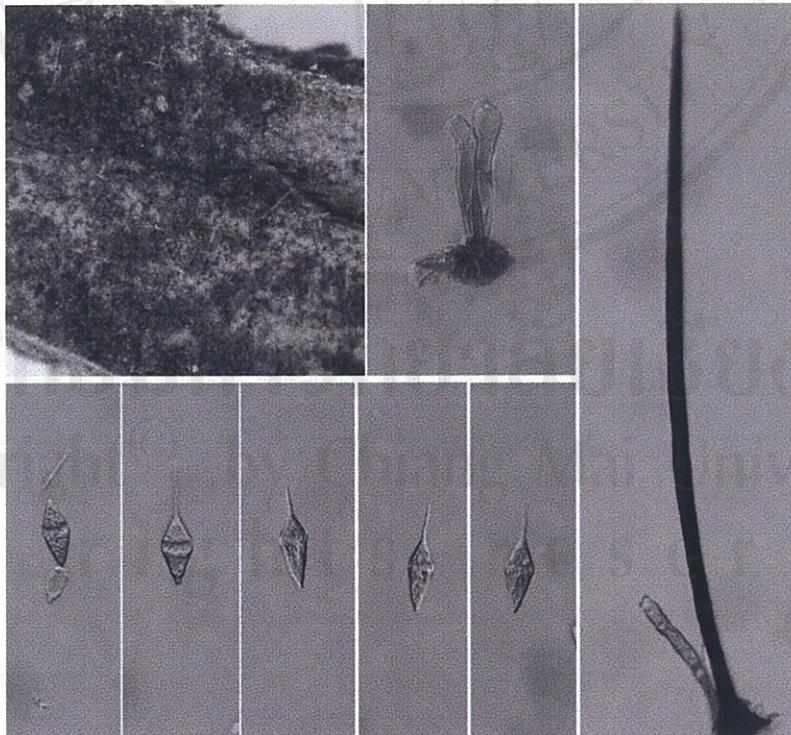


Figure C.4 *Beltrania rhombica*.



Figure C.5 *Beltraniella odinae*.

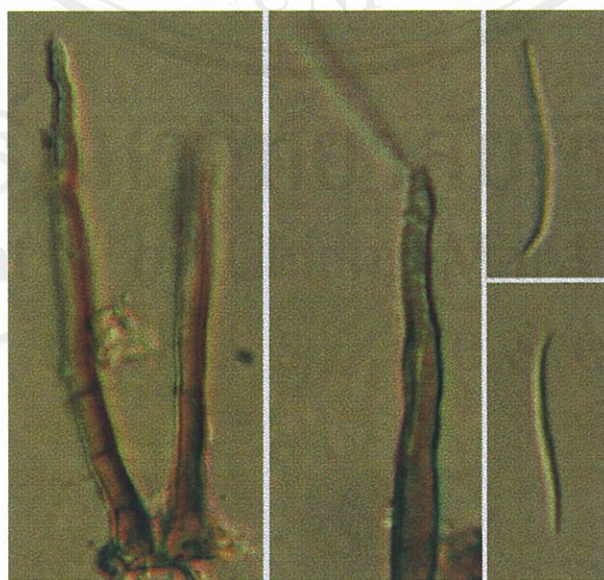


Figure C.6 *Cercosporula* sp.

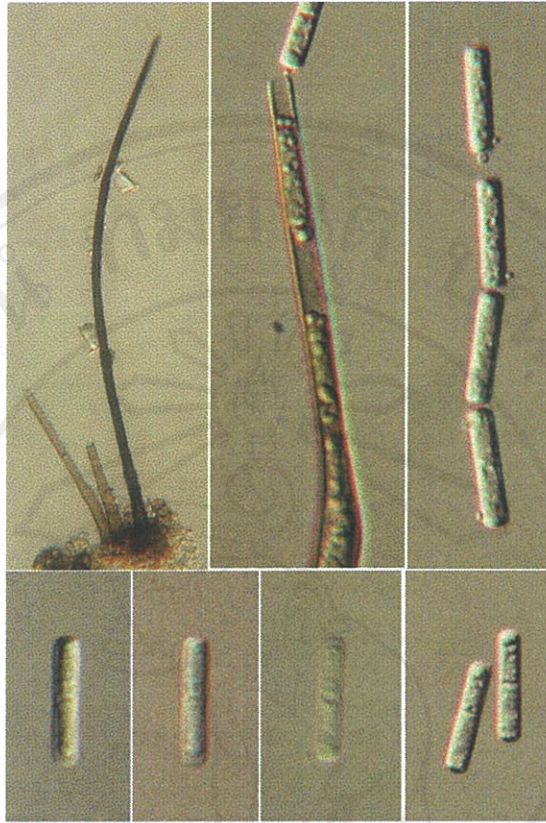


Figure C.7 *Chalara pteridina*.

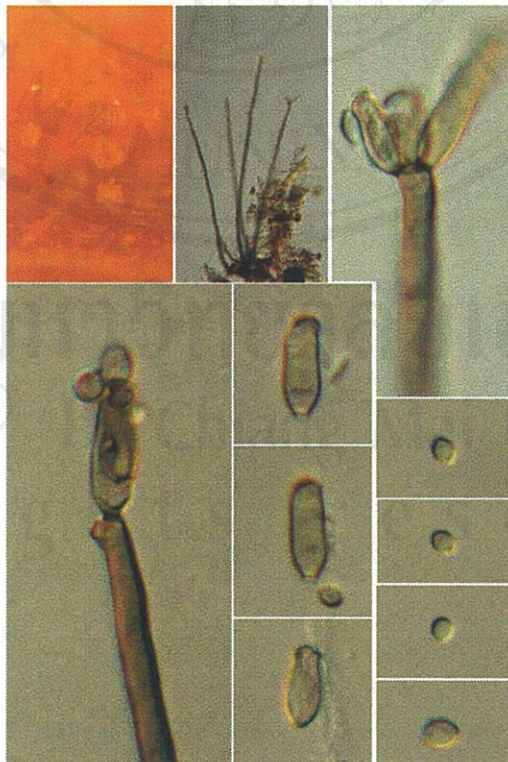


Figure C.8 *Cladosporium sphaerospermum*.

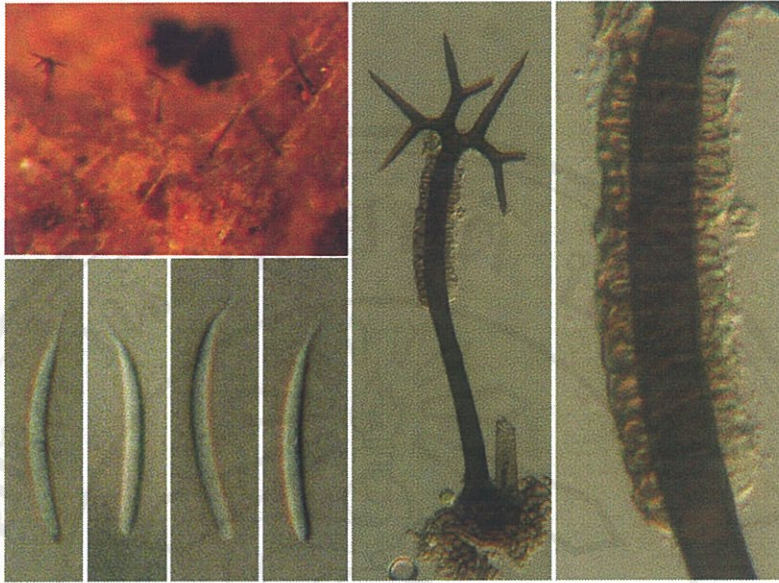


Figure C.9 *Cryptophiale udagawae*.



Figure C.10 *Dictyoarthrinium africanum*.



Figure C.11 *Dictyochaeta cylindrospora*.

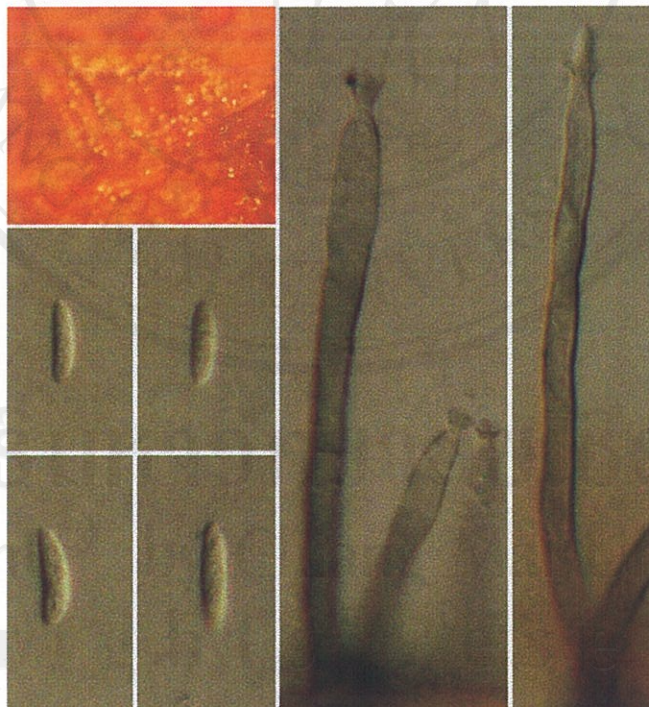


Figure C. 12 *Dictyochaeta heteroderae*.



Figure C.13 *Dictyochaeta simplex*.

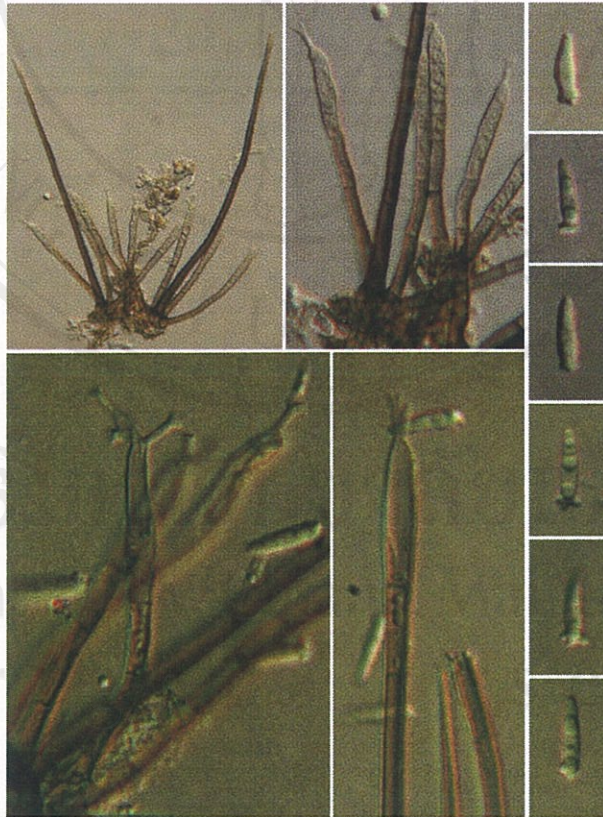


Figure C.14 *Dictyochaeta stipitocolla*.

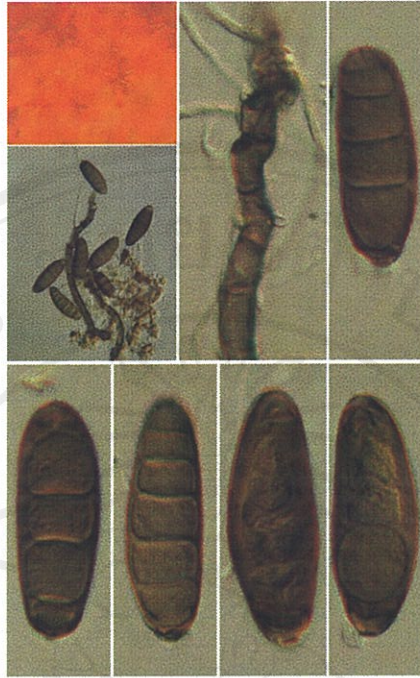


Figure C.15 *Drechslera cynodontis*.

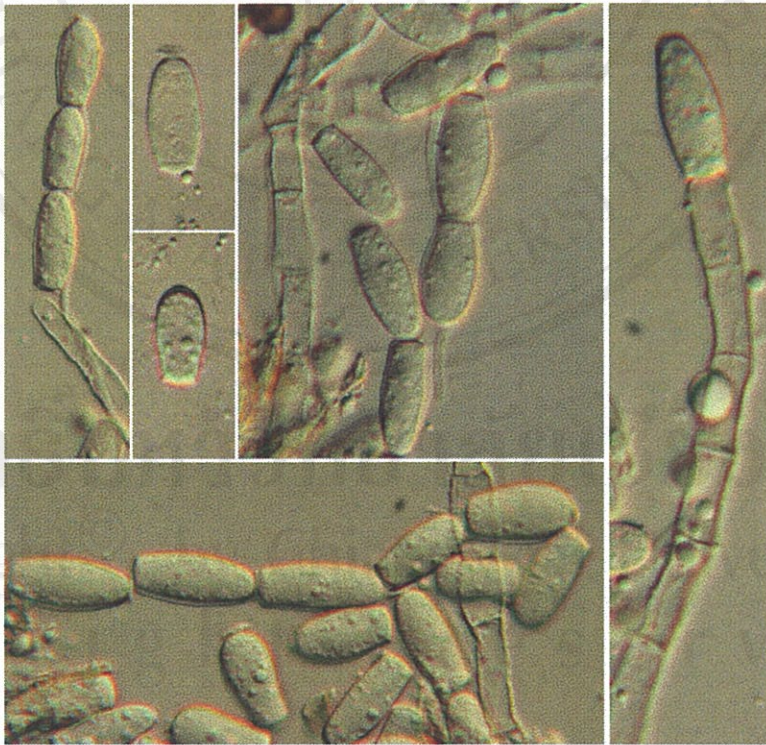


Figure C.16 *Geotrichum candidum*.

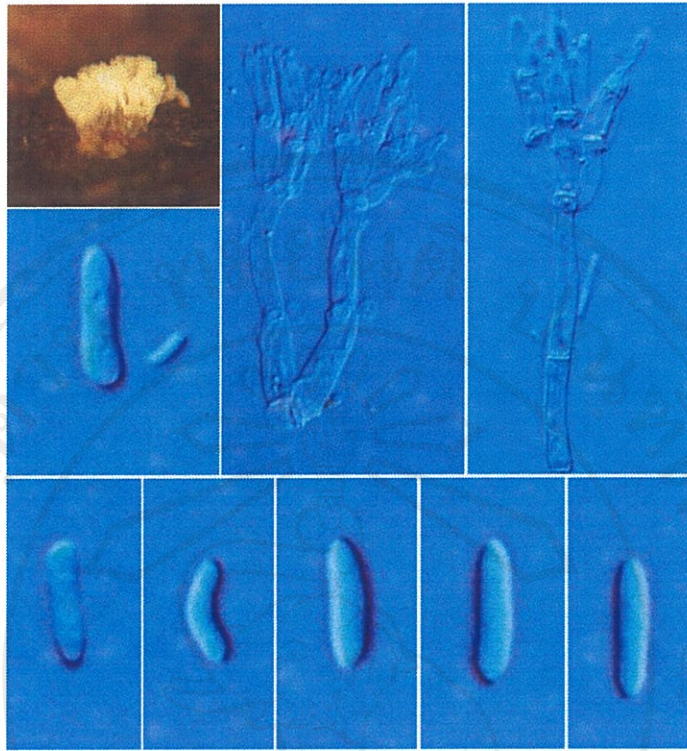


Figure C.17 *Gliocladium cylindrosporum*.

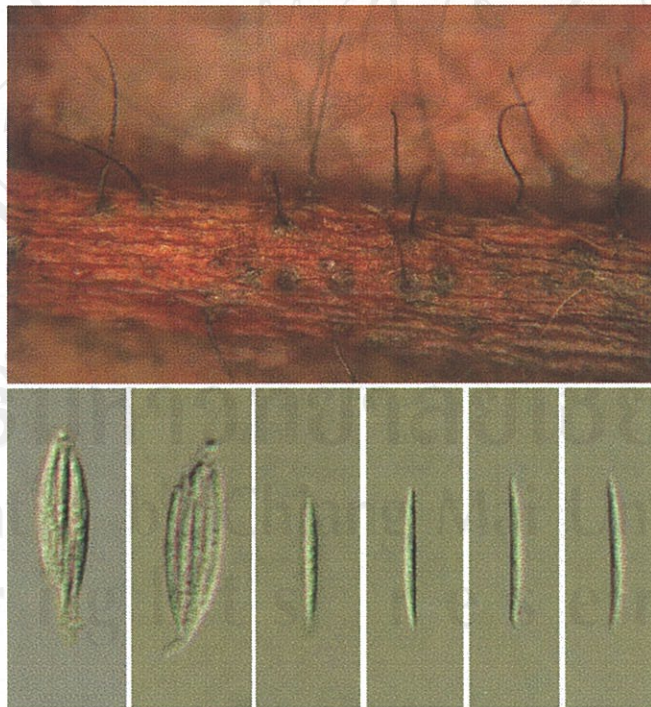


Figure C.18 *Gnomonia amoena*.

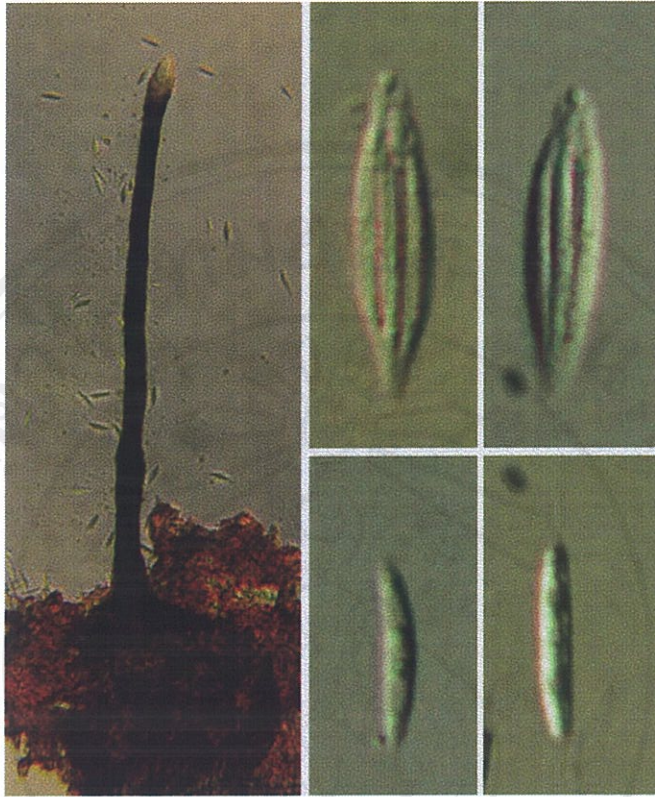


Figure C.19 *Gnomonia gnomon*.

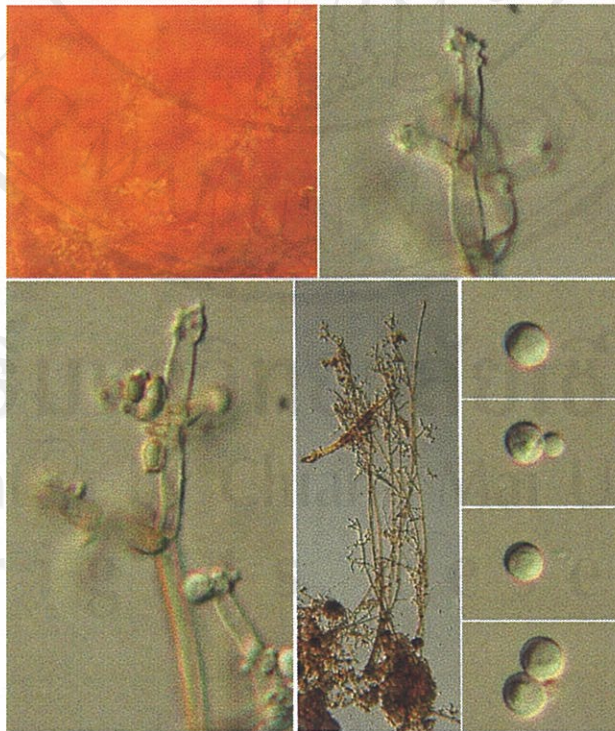


Figure C.20 *Hansfordia pulvinata*.

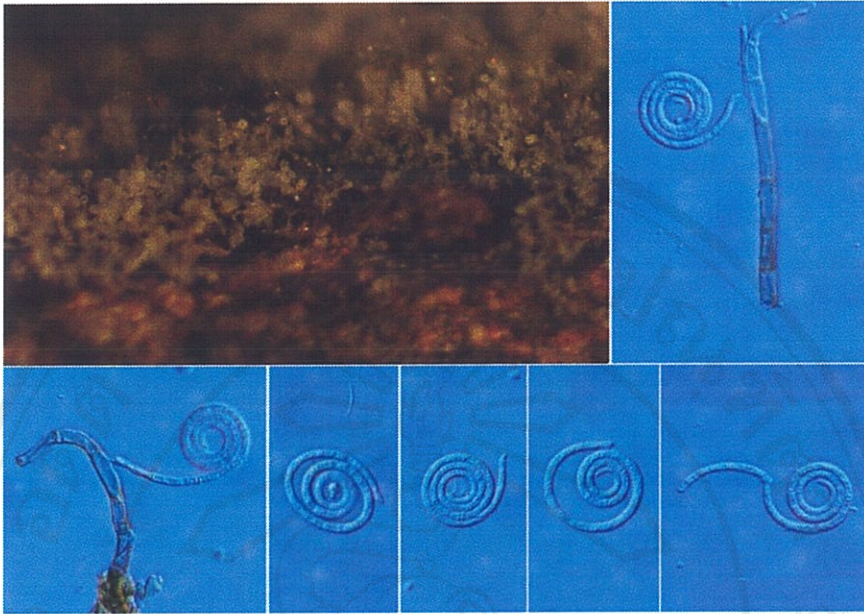


Figure C.21 *Helicosporium talbotii*.



Figure C.22 *Idriella fertilis*.

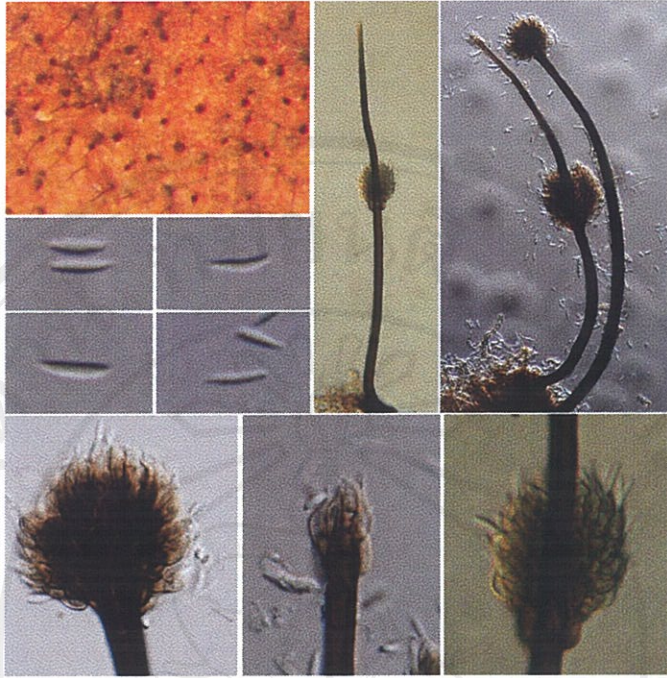


Figure C.23 *Kionochoeta spissa*.

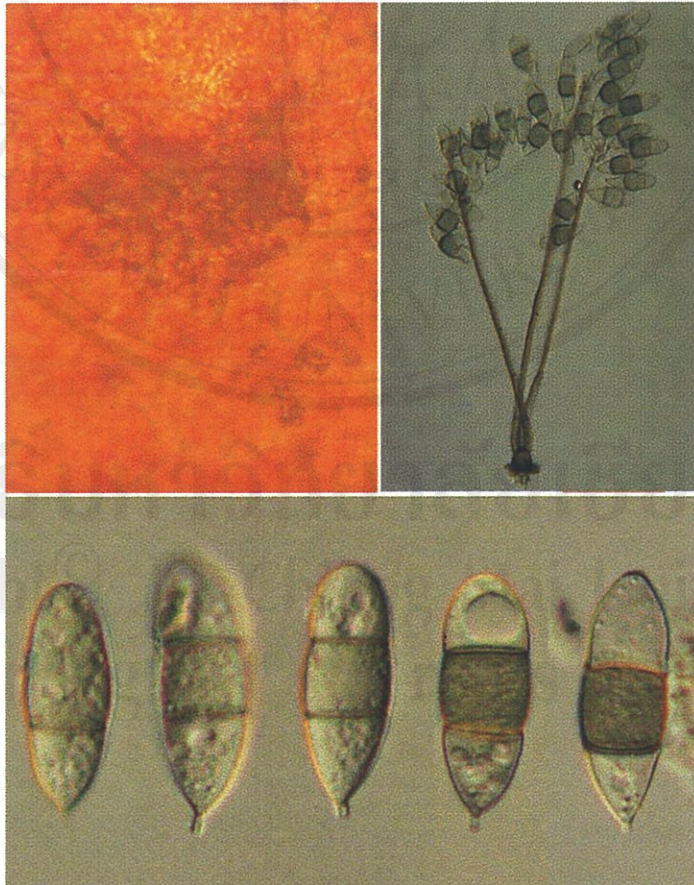


Figure C.24 *Kramasamuha sibika*.

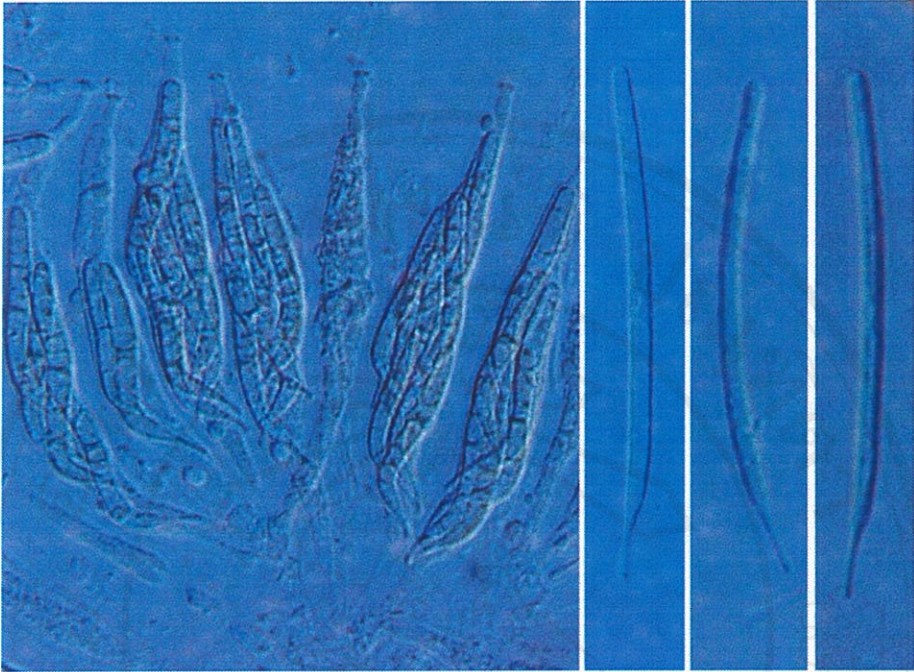


Figure C.25 *Hilberina chapmanii*.

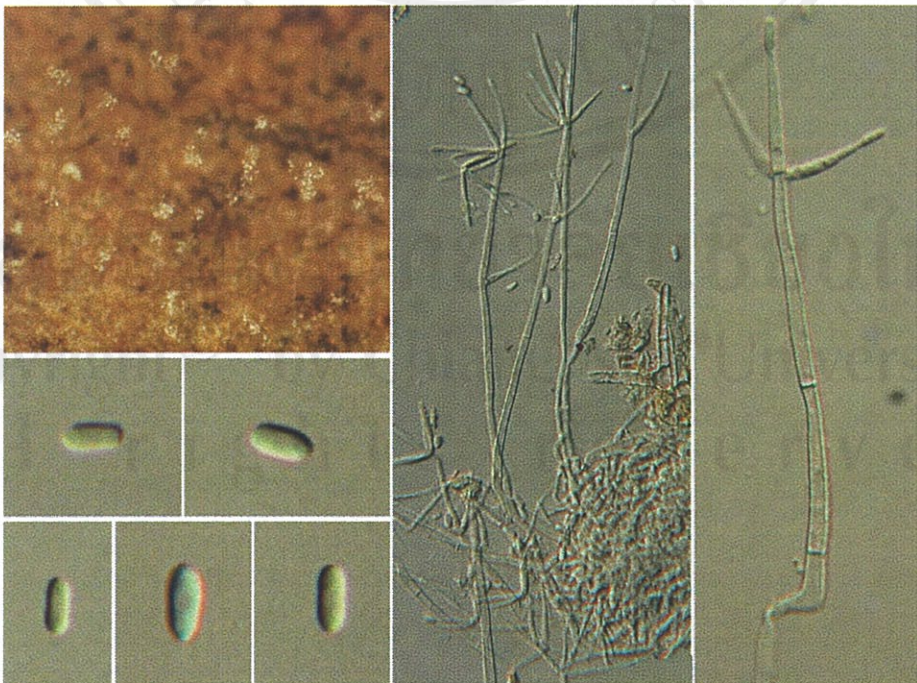


Figure C.26 *Lecanicillium lecanii*.



Figure C.27 *Linocarpus* sp.

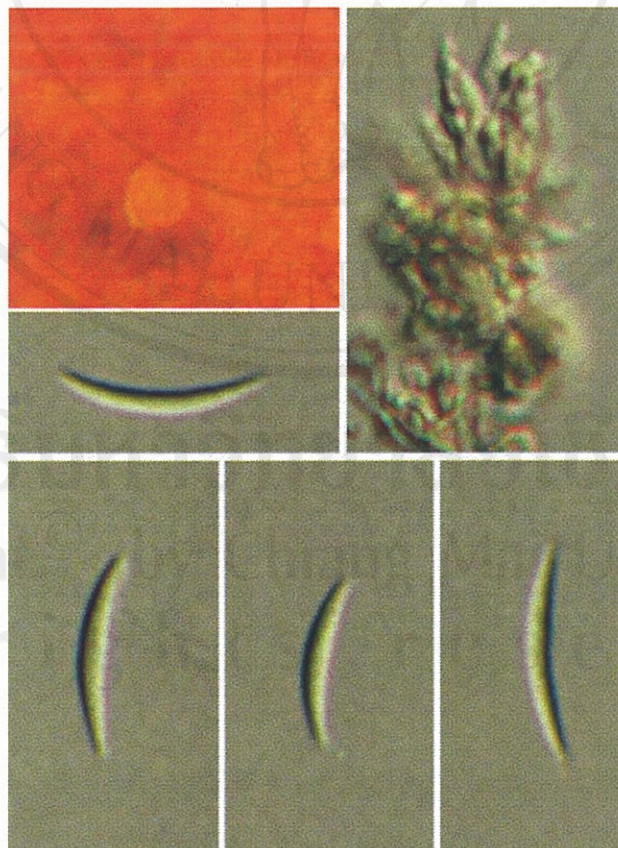


Figure C.28 *Microdochium phragmitis*.



Figure C.29 *Ophioceras commune*.



Figure C.30 *Periconia cookei*



Figure C.31 *Selenosporella falcate*.

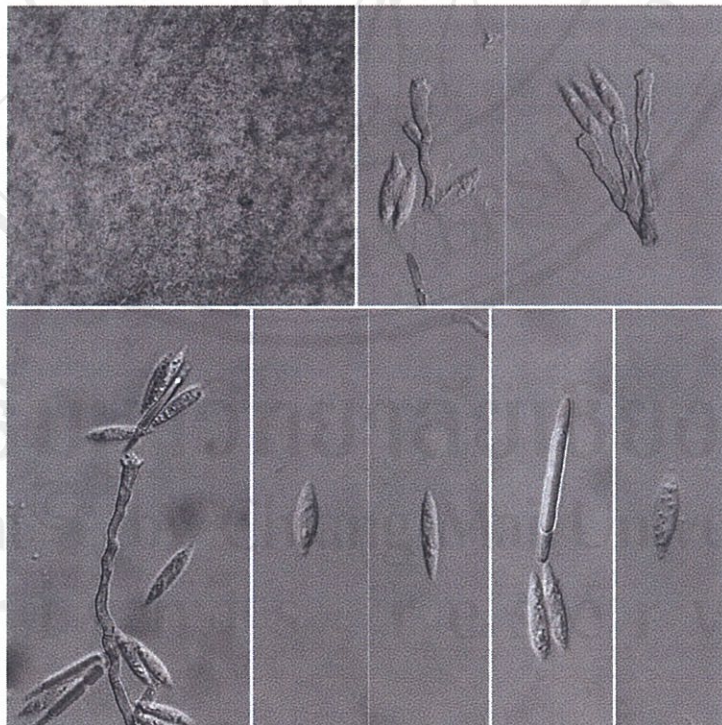


Figure C.32 *Subramaniomyces fuisaprophyticus*.

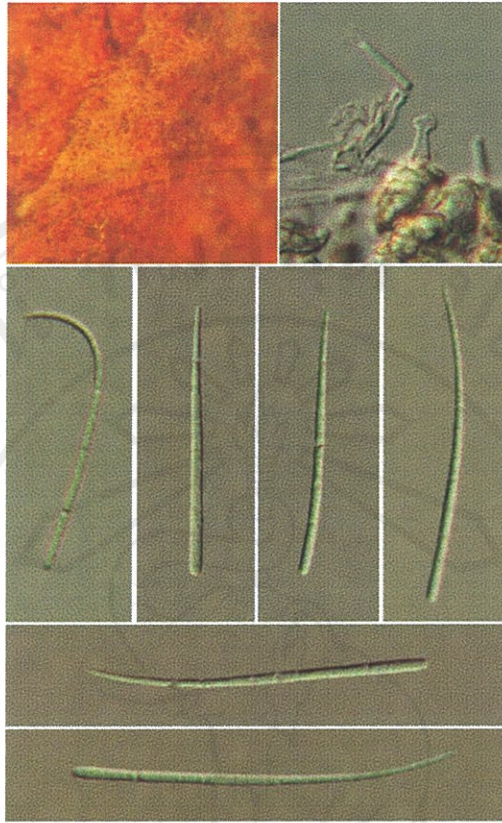


Figure C.33 *Polyscytalum hareae*.

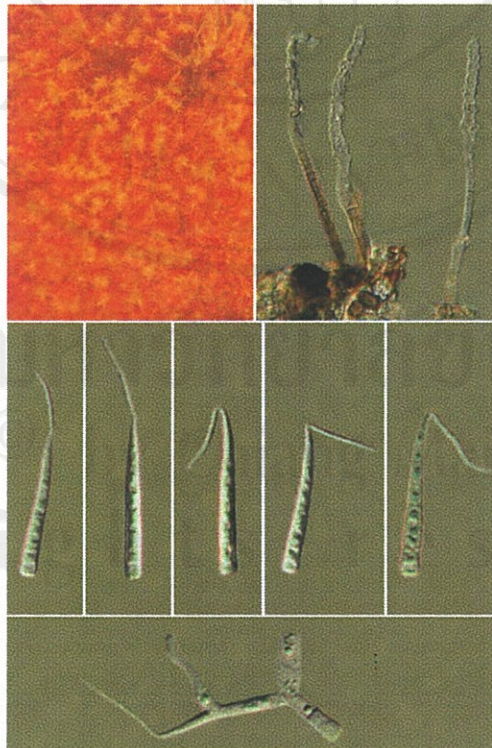


Figure C34 *Subulispora procurvata*.



Figure C.35 *Wiesneriomyces* sp.

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APPENDIX D

Table D.1 List of fungi recorded on sterile hanging leaves of *Castanopsis diversifolia* during succession study (numbers indicates number of times that a fungus encountered).

Species names	Day 7		Day 21			Day 51			Day 86			Day 120			
	P and main vein	Up side up	Up side down	P and main vein	Up side up	Up side down	P and main vein	Up side up	Up side down	P and main vein	Up side up	Up side down	P and main vein	Up side up	Up side down
<i>Acremonium zonatum</i>						1									
<i>Alternaria</i> sp.															1
<i>Aspergillus</i> sp.			1												
<i>Asterina</i> sp.											2				
<i>Bacillispora aquatica</i>	3			2			1								
<i>Beltrania rhombica</i>								1	1						
<i>Beltraniella portoricensis</i>					1	2					1	1		2	1
<i>Cercosporula</i> sp.								1	1						
<i>Chalara pteridina</i>								1	1		3	3			2
<i>Cladosporium cladosporioides</i>															1
<i>Cladosporium oxysporum</i>			6			6			5			6			3
<i>Cladosporium</i> sp. 1															1
<i>Cladosporium sphaerospermum</i>			4			7			5			5			3
<i>Cladosporium tenuissimum</i>								1							
<i>Coelomycetes</i> sp. 2						1		1	1		1				
<i>Coelomycetes</i> sp. 5							2			3				3	
<i>Coelomycetes</i> sp. 6								2			3				
<i>Cylindrocladium gracile</i>				2		1									
<i>Cylindrum griseum</i>		1				2		1			1	1			
<i>Dendrodochium cylindricum</i>	1									1					
<i>Dictyochaeta cylindrospora</i>								2	2						
<i>Dictyochaeta simplex</i>											2	1			
<i>Dictyochaeta stipitocolla</i>										1					
<i>Dictyochaeta</i> -like					1										
<i>Drechslera cynodontis</i>			1												
<i>Emarcea castanopsidicola</i>								4	4		2	2		1	
<i>Endophragmiella</i> sp.											2	2		1	
<i>Fusarium</i> sp.			2							1					
<i>Geotrichum candidum</i>			1												
<i>Gliocladium cylindrosporum</i>		3	1		3	2									
<i>Hansfordia</i> sp.			1												
<i>Haplographium</i> sp.											1				
<i>Idriella</i> sp. 1	1	2	5		2	2			1		1	2		1	1
<i>Idriella fertilis</i>						1									
<i>Idriella</i> sp. 2						1									
<i>Kramasamuha sibika</i>		1									1				

Table D.1 (Continued)

Species names	Day 7			Day 21			Day 51			Day 86			Day 120		
	P and main vain	Up side up	Up side down	P and main vain	Up side up	Up side down	P and main vain	Up side up	Up side down	P and main vain	Up side up	Up side down	P and main vain	Up side up	Up side down
<i>Lauriomyces bellulus</i>				3			1								
<i>Lecanicillium lecanii</i>			1												
<i>Lichenopeltella salicis</i>												1			
<i>Microdochium phragmitis</i>			1									1			
<i>Microthyrium</i> sp.	1														
<i>Monochaetia</i> sp.								1	1		1			1	
<i>Mycosphaeriella</i> sp.			4			2									
<i>Oedocephalum</i> sp.			4												
<i>Arthrallema</i> sp.	1	2	4		2	3			2	1	2	9	1	2	2
<i>Penicillium</i> sp.						2									
<i>Periconia cookei</i>			2			1									
<i>Periconia</i> sp. 1														1	
<i>Pestalospaeria</i> sp.			1		1	1		1							
<i>Pestalotiopsis tecomicola</i>		2	5			3		3	3						
<i>Phomopsis</i> sp.			1												
<i>Pithomyces karoo</i>			2			1									
<i>Pyricularia</i> sp.			1												
<i>Ramularia gei</i>												3			3
<i>Subramaniomyces fusisaprophyticus</i>			1			1		2		2	5		1	2	
<i>Subulispora procurvata</i>			2		3	3		2	3		2		1	1	
<i>Thozetella</i> sp.					1	1									
<i>Thysanophora</i> sp.									1						
<i>Tritirachium bulbophorum</i>	1														
Unidentified hyphomycete			2						2						
Unidentified ascomycete sp. 2									1						
<i>Wiesneriomyces javanieus</i>								1	1						
<i>Xenocylindrocladium</i> sp.	1														
<i>Zygosporium gibbum</i>			7			9			9			8			2
<i>Zygosporium minus</i>						1									

Table D.2 List of fungi recorded on sterile leaves of *Castanopsis diversifolia* on the ground during succession study (numbers indicates number of times that a fungus encountered).

Species names	Day 7		Day 21		Day 51		Day 86		Day 120			
	P and main vein	Up side up	Up side down	P and main vein	Up side up	Up side down	P and main vein	Up side up	Up side down	P and main vein	Up side up	Up side down
<i>Beltrania rhombica</i>		1	2		3	2	1	1	2	1		1
<i>Beltraniella ondinae</i>		1	1									
<i>Beltraniella portociensis</i>		1	1		1	1	1	1	2	1	3	2
<i>Chaetendophragma triangularia</i>												1
<i>Chaetosphaeria</i> sp.									1		1	
<i>Chalara pteridina</i>		1	1		1	1	1	1	1	2		
<i>Cladosporium oxysporum</i>			1									1
<i>Cladosporium sphaerospermum</i>											1	
<i>Coelomycetes</i> sp. 2								2		1		
<i>Coelomycetes</i> sp. 6									2			1
<i>Coelomycetes</i> sp. 8							1			1		
<i>Coelomycetes</i> sp. 9							1		1			
<i>Coelomycetes</i> sp. 10							1		1			
<i>Cryptophiale udagawae</i>								1	1	1	2	1
<i>Cylindrocladium gracile</i>			1		1	1	2	1	2	2		
<i>Cylindrum griseum</i>										1		
<i>Dendrodochium cylindricum</i>			1									
<i>Dictyochaeta heteroderae</i>												1
<i>Dictyochaeta pakhalensis</i>									1	1	1	1
<i>Dictyochaeta simplex</i>		1	2		1	1	1	1	1	1		
<i>Dictyochaeta</i> sp.												1
<i>Dictyochaeta stipitocolla</i>							1	1	1		1	1
<i>Drechslera cynodontis</i>					1							
<i>Geotrichum candidum</i>			1		1							
<i>Gilocladium cylidrosporum</i>			1		2	1	1					
<i>Haplographium</i> sp. state of			1									
<i>Hyaloscypha dematiicola</i>												
<i>Helicosporium talbotii</i>										1		2
<i>Hyponectria buxi</i>								1	1	1		
<i>Idriella</i> sp. 1		2	2		2	3	2	1	1	1	2	1
<i>Idriella fertilis</i>		1			1							2
<i>Kionochaeta spissa</i>		1	1		1	1	1	2	1			
<i>Lauriomyces bellulus</i>		1			1		1		1			
<i>Lecanicillium lecanii</i>					1							
<i>Marasmius</i> sp. 1												1
<i>Marasmius</i> sp. 2												1
<i>Menisporopsis nova-zelandiae</i>		2	1		1	2	1		2	2		
<i>Microdochium phragmitis</i>		2	2		1	1	2	1		1		
<i>Mycena</i> sp. 1										1		
<i>Mycena</i> sp. 3							1					
<i>Ophioceras commune</i>					2		1		1			
<i>Penecillium</i> sp.					1			1				

Table D.2 (Continued)

Species names	Day 7		Day 21			Day 51			Day 86			Day 120			
	P and main vain	Up side up	Up side down	P and main vain	Up side up	Up side down	P and main vain	Up side up	Up side down	P and main vain	Up side up	Up side down	P and main vain	Up side up	Up side down
<i>Phaeoisaria</i> sp.			1												1
<i>Phomopsis</i> sp.					1		1								
<i>Pseudobotrytis terrestris</i>															1
<i>Pseudohalonectria</i> sp.							1	1	2	1			1	1	
<i>Sesquicillium candelabrum</i>							1								
<i>Stemonitis</i> sp.							1								
<i>Subramaniomyces fusisaprophyticus</i>	1	1		1	1		1	1	3	1					
<i>Subulisporea procurvata</i>	2	1	1	2	2		1	2	2	2			1	1	
<i>Thozetella</i> sp.					1					1	1		1		
<i>Verticillium</i> sp.									1						
<i>Wiesneriomyces javanieus</i>				1			1	1							
<i>Xenocylindrocladium</i> sp.	2												1		

Table D.3 List of fungi recorded on unsterile leaves of *Castanopsis diversifolia* on the ground during succession study (numbers indicates number of times that a fungus encountered).

	Day 7		Day 21			Day 51			Day 86			Day 120			
	P and main vain	Up side up	Up side down	P and main vain	Up side up	Up side down	P and main vain	Up side up	Up side down	P and main vain	Up side up	Up side down	P and main vain	Up side up	Up side down
<i>Acremonium zonatum</i>				1	1	1	1	2	1	2	1		1	1	
<i>Albonectria albosuccinea</i>								1	1						
Annulatascaceae sp.									2						
<i>Ardhachandra cristaspora</i>						1			1						
<i>Arthrimum</i> sp.				3	1		3	2							
<i>Beltrania mangifera</i>												1			
<i>Beltrania rhombica</i>					4	2		3	5	1	1				1
<i>Beltraniella odinae</i>	2			2	3		3	3	1	2					1
<i>Beltraniella portoricensis</i>				2	1		2	2	1	1					
<i>Beltraniella</i> sp.									1						
<i>Chaetosphaeria</i> sp.															1
<i>Chalara pteridina</i>				3	1										
<i>Cladosporium</i> sp. 2						1			1			1			
<i>Cladosporium sphaerospermum</i>												1			
Coelomycete sp. 2	1	1		3	3		3	2							
Coelomycete sp. 3		1		1	2		2	2							
Coelomycete sp. 4	3				1										
Coelomycete sp. 5				4											
Coelomycete sp. 6	1	1		2	1		2	1		1	1		1	1	
Coelomycete sp. 9							1				3				
Coelomycete sp. 10														2	1
Coelomycete sp. 11								1							

Table D.3 (Continued)

	Day 7			Day 21			Day 51			Day 86			Day 120		
	P and main vain	Up side up	Up side down	P and main vain	Up side up	Up side down	P and main vain	Up side up	Up side down	P and main vain	Up side up	Up side down	P and main vain	Up side up	Up side down
<i>Cryptophiale udagawae</i>								1	2		1	1		2	1
<i>Cylindrocladium pseudogracile</i>				1											
<i>Dendrodochium cylindricum</i>								1	1		1	1			
<i>Dictyochaeta simplex</i>											1	2			
<i>Dictyochaeta</i> sp. 2						1		2			3	1			
<i>Emarcea castanopsidicola*</i>						1			1			1			
<i>Gliocladium cylindrosporum</i>		1		1	1										
<i>Gnomonia petiolorum</i>				1			4			1					
<i>Hansfordia</i> sp.				2											1
<i>Helicosporium talbotii</i>															1
<i>Hyponectria buxi</i>											1				
<i>Idriella</i> sp. 1	2	1		4	5	1	2	3	1	1	1				
<i>Kionochaeta spissa</i>						1		1							
<i>Lachnum</i> sp.						1									
<i>Lecanicillium lecanii</i>				3	1			2	2		2	2		1	2
<i>Lophodermium australiense</i>															2
<i>Lophodermium</i> sp. 3									1		1			1	
<i>Marasmius</i> sp. 1													1		1
<i>Menisporopsis novae-zelandiae</i>					1				1						
<i>Mycena</i> sp. 1														1	
<i>Mycosphaerella</i> sp.	1					1		2							
<i>Ophioceras commune</i>		1		2	2			2	2			2			
<i>Parasymphodiella laxa</i>							1	1	1		1	1	1	1	
<i>Penicillium</i> sp.											2	2		2	3
<i>Periconia paludosa</i>														3	1
<i>Periconia</i> sp. 2								3	3						
<i>Stemonitis</i> sp.									1		1			1	
<i>Stictis</i> sp. 1												1		1	1
<i>Stomiopeltis</i> sp.	1					1									
<i>Subramaniomyces fusisaprophyticus</i>					1	1			1						
<i>Subulispora procurvata</i>				2	2			2	1				1		
Unidentified ascomycete sp. 1					1										
Unidentified hyphomycete sp.												1			

APPENDIX E

Phylotypes sequences revealed from DGGE

```
>Phylotype_DLL1
TAAACAAGTCTGTACGTGAGACTGCGAATGTCTCATAAAATCAGTTATAGTTTATTTGATGATACCTTG
CTACATGGATAACTGTGGTAATTCTAGAGCTAATACATGCATTTCAAGCCCCGACTTCTGGAAGGGGTGT
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>Phylotype_DLU1
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GAAGGGGTGTATTTATTAGATAAAAAACCAACGCGGCTCGCCGCTCCTTTGGTGATTTCATAATAACTTC
TCGAATCGCATGGCCTTGTGCCGGCGATGCTTCATTCAAATATCTGCCCTATCAACTTTTCGATGGTAGG
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>Phylotype_DLU3
CATGCCTGTCTAGTATAAAACAAAGTTTGTACTGTGAACTGCGAATGGCTCATTAAATCAGTTATAGTTT
ATTTGATGATTCCTTGTACATGGATAACTGTGGTAATTCTAGAGCTAATACATGCATTTTCAGCCCCGA
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>Phylotype_DLL3
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>Phylotype_LU
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ACAATGGTTTCAACGGGTAACGGGGAATAGATTCCGACTC
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>Phylotype_SU5

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>Phylotype_SU4

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>Phylotype_SU1

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>Phylotype_LR1

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>Phylotype_SL2

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GGCCTTGTGCCGGCGATGGTTCATTCAAATTTCTGCCCTATCAACTTTCGATGGCTGGGTCTTGCCA
GCCATGGTGACAACGGGTAACGGGGATTAGGGCTCGACCCCGGAGA

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Special project: Study on cellulose degradation activities of *Aspergillus* and *Penicillium* genera isolated from rubbishes in Hanoi area (Bachelor Major in French)

Scholarships and Awards

1998: French linguistic awards (Two-months trip to Montpellier Universite III, Montpellier)

1999: Francophone scholarship for students presenting major in French (three-month period at Department of Molecular Evolotion, The University du Maine, Le Mans).

2004: 2-month exchange student to the Department of University of Hong Kong (February- April).

2004-2005: 6-month exchange student to Hong Kong University (December 2004- May 2005).

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2006: 3-month exchange student to centraalbureau voor schimmels cultures (CBS), Utrecht, Netherlands.

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Emarcea castanopsidicola gen. et sp. nov. from Thailand, a new xylariaceous taxon based on morphology and DNA sequences

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Abstract: We describe a unique ascomycete genus occurring on leaf litter of *Castanopsis diversifolia* from monsoonal forests of northern Thailand. *Emarcea castanopsidicola* gen. et sp. nov. is typical of Xylariales as ascospores develop beneath a blackened clypeus, ostioles are papillate and asci are unitunicate with a J⁺ subapical ring. The ascospores in *Emarcea castanopsidicola* are, however, 1-septate, hyaline and long fusiform, which distinguishes it from other genera in the Xylariaceae. In order to substantiate these morphological findings, we analysed three sets of sequence data generated from ribosomal DNA gene (18S, 28S and ITS) under different optimality criteria. We analysed this data to provide further information on the phylogeny and taxonomic position of this new taxon. All phylogenies were essentially similar and there is conclusive molecular evidence to support the establishment of *Emarcea castanopsidicola* within the Xylariales. Results indicate that this taxon bears close phylogenetic affinities to *Muscodor* (anamorphic Xylariaceae) and *Xylaria* species and therefore this genus is best accommodated in the Xylariaceae.

Taxonomic novelties: *Emarcea* Duong, R. Jeewon & K.D. Hyde gen. nov., *Emarcea castanopsidicola* Duong, R. Jeewon & K.D. Hyde sp. nov.

Key words: *Castanopsis*, phylogeny, rDNA, systematics, Xylariaceae.

INTRODUCTION

We are studying the microfungi occurring on leaf litter in northern forests of Thailand. This substrate, which has a high fungal biodiversity, has resulted in the description of several new species from *Magnolia liliifera* (e.g. Promputtha *et al.* 2002). We are now studying the fungi on leaf litter of several other hosts including *Castanopsis diversifolia*. Fungal diversity from host species of *Castanopsis* is quite well documented. So far, a total of 175 fungal species from 108 genera have been recorded from *Castanopsis* spp. (EMBL fungal databases 2004). *Anthostomella castanopsis* is the only record from Xylariaceae. Preliminary fungal succession studies on *Castanopsis fissa* in Hong Kong have not yielded any xylariaceous fungi (Tang *et al.* unpubl. data) but in this paper, we introduce a new ascomycete in the Xylariaceae which was found on *Castanopsis diversifolia*. The aim of this paper is to describe *Emarcea castanopsidicola* as a new taxon and to establish its ordinal and familial placement using morphological characteristics and sequence data.

MATERIALS AND METHODS

Sampling

Dead leaves of *Castanopsis diversifolia* were randomly collected from the forest floor, placed in sterile plastic bags and returned to the laboratory. They were then incubated separately in plastic boxes lined with moistened tissue and examined periodically using a grid method (Paulus & Hyde 2004). Located ascospores were mounted in water to look for asci, ascospores and paraphyses. Melzer's reagent (Dickinson & Lucas 1983) was used to check for iodine reactions and India ink was used to establish if appendages or sheaths occurred on the ascospores. All morphological measurements are in sterile water, with a mean from 25 measurements for each character. Single-spore isolation was carried out from fresh samples, using a hand-made glass needle (Goh 1999). Single-spore cultures grown on artificial medium for 3 weeks were used for further molecular studies.

Molecular methods

DNA extraction was carried out using CTAB lysis buffer and phenol chloroform as outlined by Jeewon *et al.* (2002, 2004). Partial sequences from three different regions of the rDNA molecule (characterised by different rates of evolution) were amplified.

Table 1. Fungi used in the study with their GenBank accession numbers

.Ingroups (18S, 28S)	18S rDNA	28S rDNA	Ingroups (ITS)	ITS, 5.8S
<i>Ambrosiella macrospora</i>		AF282873	<i>Muscodor albus</i>	AY555731
<i>Ambrosiella sulfurea</i>	AF348149		<i>Muscodor albus</i>	AF324336
<i>Aniptodera chesapeakensis</i>	U46870		<i>Muscodor albus</i>	AY527045
<i>Apiosporopsis carpinea</i>	AF277110		<i>Muscodor albus</i>	AY527048
<i>Arecophila</i> sp.		AF452039	<i>Muscodor azulenus</i>	AY244622
<i>Ascovaginospora stellipala</i>	U85087		<i>Muscodor</i> sp. A35	AY034665
<i>Bartalinia robillardoides</i>		AF382366	<i>Muscodor vitigenus</i>	AY100022
<i>Cainia graminis</i>		AF452033	<i>Xylaria arbuscula</i>	AY183369
<i>Chaetomium globosum</i>	AY545725	AY545729	<i>Xylaria arbuscula</i>	AY183369
<i>Clohesia corticola</i>		AF132329	<i>Xylaria enteroleuca</i>	AF163033
<i>Coniochaeta ligniaria</i>	AY198389		<i>Xylaria hypoxylon</i>	AJ309350
<i>Cryphonectria havanensis</i>		AF408339	<i>Xylaria hypoxylon</i>	AF194027
<i>Cryphonectria parasitica</i>	AF277116		<i>Xylaria mali</i>	AF163040
<i>Diaporthe pustulata</i>		AF408358	<i>Xylaria</i> sp. F19	AY315404
<i>Discosia</i> sp.		AF382381	<i>Xylaria</i> sp. F4	AY315405
<i>Discostroma fuscillum</i>	AF346548		<i>Emarcea castanopsidicola</i>	AY603496
<i>Discostroma</i> sp.		AF382380		
<i>Discostroma tricellulare</i>	AF346546		Outgroup ITS	
<i>Discula fraxinea</i>	AF277106		<i>Diatrype flavovirens</i>	AJ302428
<i>Discula quercina</i>	AF277108		<i>Cryptosphaeria ligniota</i>	AJ302418
<i>Emarcea castanopsidicola</i>	AY603494	AY603495		
<i>Halosphaeria appendiculata</i>		U46885	Outgroup 28S dataset	
<i>Kretzschmaria clavus</i>		AJ390434	<i>Dothidea sambuci</i>	AF382387
<i>Lepteutypa cupressi</i>		AF382379	<i>Pleospora herbarum</i>	AF382386
<i>Leucostoma auerswaldii</i>		AF408384		
<i>Lignincola laevis</i>		U46890	Outgroup 18S dataset	
<i>Linocarpon pandanicola</i>		AF452041	<i>Dothidea insculpta</i>	U42474
<i>Linocarpon</i> sp.		AF452042	<i>Pleospora betae</i>	U43466
<i>Lollipopaia minuta</i>	AF301534			
<i>Muscodor albus</i>	AF324337			
<i>Muscodor</i> sp. A3 5	AY034664			
<i>Nais inornata</i>	AF050482			
<i>Neurospora crassa</i>		AF286411		
<i>Nimbospora effusa</i>	U46877			
<i>Nohea umiumi</i>	U46878	U46893		
<i>Ophiodeira monosemeia</i>		U46894		
<i>Ophiostoma africanum</i>		AF221015		
<i>Ophiostoma piliferum</i>		AF221625		
<i>Ophiostoma piliferum</i>	AF136961			
<i>Ophiostoma torulosum</i>	AY497517			
<i>Pestalospaeria hansenii</i>	AF242846			
<i>Pestalospaeria</i> sp.		AF452031		
<i>Plagiostoma euphorbiae</i>	AF277114			
<i>Seimatosporium leptospermi</i>		AF382373		
<i>Seynesia erumpens</i>		AF279410		
<i>Sordaria fimicola</i>	AY545724	AY545728		
<i>Thielavia cephalothecoides</i>		AF286413		
<i>Truncatella angustata</i>	AF346560	AF382383		
<i>Umbrinosphaeria caesia</i>		AF261069		
<i>Valsella salicis</i>		AF408389		
<i>Xylaria acuta</i>	AY544719	AY544676		
<i>Xylaria hypoxylon</i>	AY544760	AY544648		

Primer pairs NS1 (5'-GTA GTC ATA TGC TTG TCT C-3') & NS4 (5'-CTT CCG TCA ATT CCT TTA AG-3') as defined by White *et al.* (1990) were used to amplify a region spanning approximately 1200 nucleotides from the small subunit (18S) of the rDNA. LROR (5'-ACCCGCTGAACTTAAGC-3') and LR5 (5'-TCCTGAGGGAACTTCG-3') primer pairs as defined by Vilgalys & Hester (1990) were used to

amplify a segment of the large 28S subunit (about 950 nucleotides). In addition, primer pairs ITS 4 (5'-TCC TCC GCT TAT TGA TAT GC-3') and ITS 5 (5'-GGA AGT AAA AGT CGT AAC AAG G-3') as defined by White *et al.* (1990) were used to generate about 600 nucleotides from the complete ITS (including 5.8S) regions. The amplification conditions were performed in a 50 µL reaction volume as follows: 1×

PCR buffer, 0.2 mM each dNTP, 0.3 μ M of each primer; 1.5 mM MgCl₂, 0.8 units Taq Polymerase and 10 ng DNA. PCR parameters for all the regions were as follows: Initial denaturation 94 °C for 3 min, 30 cycles of 94 °C for 1 min, 52 °C for 50 s, 72 °C for 1 min, final extension of 72 °C for 10 min. Characterisation of PCR products was done via agarose gel electrophoresis on a 1 % agarose gel containing ethidium bromide as the staining agent. DNA sequencing was performed using primers as mentioned above in an Applied Biosystem 3730 DNA Analyzer at the Genome Research Centre (University of Hong Kong).

Partial sequences generated from the different primers from *Emarcea castanopsidicola* rDNA were assembled using BioEdit (Hall 1999). Once consensus DNA sequences were obtained from the different rDNA regions under investigation, a BLAST search was performed in GenBank. DNA sequences were also submitted in GenBank. Based on the BLAST search results, putative taxa were selected as sister groups for further analyses. In addition, fungal members from *Halosphaeriales*, *Ophiostomatales*, *Sordariales* and the *Xylariales* (*Amphisphaeriaceae*) were also included in the 18S and 28S datasets, while species from *Dothidea* and *Pleospora* were used as outgroups. In the ITS dataset, however, only species from *Xylariaceae* were used as ingroups while *Diatrype flavovirens* and *Cryptosphaeria ligniota* (*Diatrypaceae*) were used as outgroup based on their close taxonomic affinities with *Xylariaceae*. Taxa used and their GenBank accession numbers are shown in Table 1. Multiple alignment was done in BioEdit (Hall 1999) and Clustal X (Thompson *et al.* 1997).

Phylogenetic analyses were conducted in PAUP v. 4.0b10 (Swofford 2002). Prior to phylogenetic analysis, ambiguous sequences at the start and the end were deleted and gaps manually adjusted to optimise alignment. Analyses were done under different optimality criteria. Gaps were treated as missing data in all analyses but the characters were also reweighted at different transition transversion ratios to examine the effect of weighting. Maximum Parsimony (MP) analyses were conducted using heuristic searches as implemented in PAUP, with the default options. One thousand pseudo-resamplings were performed, each with 10 replicates of random stepwise addition of taxa, to determine bootstrap support levels. Phenetic and Maximum Likelihood (ML) analyses were also run under a variety of assumptions as described by Jeewon *et al.* (2002, 2003a, b). A strict parsimonious tree generated from a MP analysis was used as starting tree in the ML search. Transition-transversion ratios, shape parameter and base frequencies were initially estimated. Different models of nucleotide substitutions were tested with rates assumed to follow a gamma distribution with no enforcement of a molecular clock. These estimated parameters were used in subsequent ML searches. Descriptive tree statistics (tree length

[TL], consistency index [CI], retention index [RI], rescaled consistency index [RC], homoplasy index [HI], and Log Likelihood [-Ln L]) were calculated for all trees generated under different optimality criteria. Kishino-Hasegawa tests (Kishino & Hasegawa 1989) and Templeton tests (Templeton 1983), as implemented in PAUP*, were performed in order to determine whether trees were significantly different. Trees were figured in Treeview (Page 1996).

RESULTS

Taxonomy

Emarcea Duong, R. Jeewon & K.D. Hyde, gen. nov. MycoBank MB500070.

Etymology: *Emarcea* is derived from MRC (Mushroom Research Centre) where the taxon was described and identified; *castanopsidicola* is from the name of the host *Castanopsis diversifolia*.

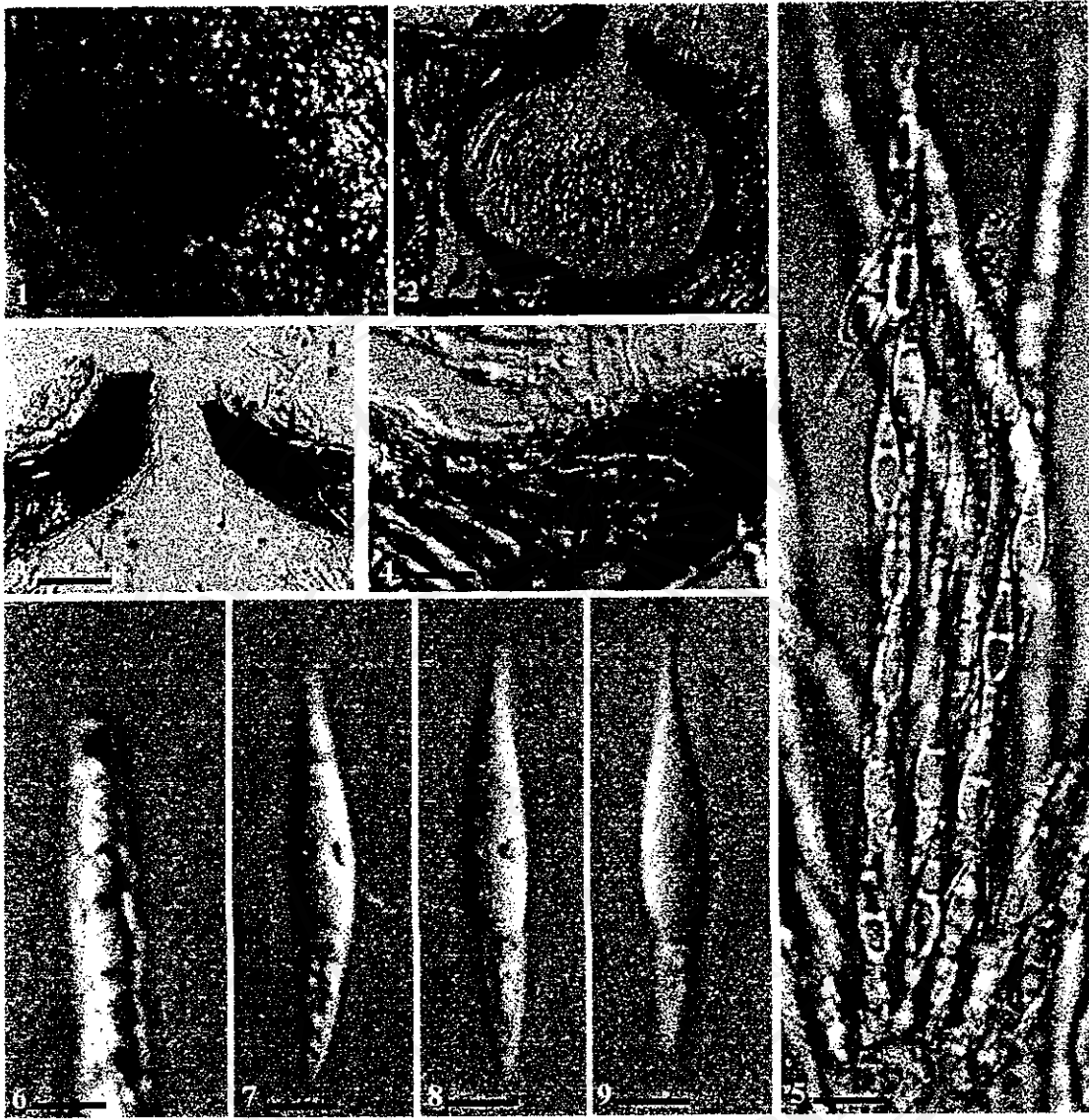
Ascomata sub clypeo immersa, globosa vel subglobosa, coriacea, ostiolo papillato, periphysato. Paraphyses hyalinae septatae. Asci octospori, cylindrici, pedicellati, unitunicati, hyalini, persistentes, rotundati ad apicem, ambitus medius, apparatus apicali J⁺. Ascospores uniseriatae, ellipsoideae, fusiformes, angustatae ad apicem, hyalinae, bicellulares, inaequaliter euseptatae, et aliquando, appendicibus mucilaginosi ad basim praeditae.

Ascomata immersed under a blackened clypeus, subglobose to globose, coriaceous, solitary; ostiole, papillate, periphysate. *Peridium* thin, comprising several layers of flattened ellipsoidal cells, brown outwardly, hyaline inwardly, and often dark brown around at the base. *Paraphyses* hyaline, septate, slightly constricted at septa, tapering at apex, longer than asci. *Asci* 8-spored, unitunicate, cylindrical, pedicellate, persistent, rounded at the apex, with a cylindrical, subapical J⁺ ring. *Ascospores* overlapping uniseriate, long fusiform, hyaline, 2-celled; apical cell obclavate, guttulate, tapering to a point; basal cell shorter than the apical cell and usually with mucilage material at the base.

Type species: *Emarcea castanopsidicola* Duong, R. Jeewon & K.D. Hyde, sp. nov.

Emarcea castanopsidicola Duong, R. Jeewon & K.D. Hyde, sp. nov. MycoBank MB500071.

Ascomata 196–250 μ m alta, 215–280 μ m diam, sub clypeo immersa, globosa vel subglobosa, coriacea, ostiolo papillato, periphysato. *Peridium* e 4–6 stratis cellularum



Figs 1–9. Interference contrast micrographs of *Emarcea castanopsidicola*. 1. Surface view of ascoma immersed as seen on the host. 2. A section of an ascoma with hyaline and septate paraphyses. 3. Ostiole and periphyses. 4. Peridium. 5. Asci containing ascospores. 6. Ascus apical ring (J^+ ring) coloured by Melzer's iodine reagent. 7–9. Ascospores squeezed from ascus. Scale bars: 1–2 = 70 μm ; 3 = 25 μm ; 4 = 12 μm ; 5 = 6 μm ; 6–9 = 4 μm .

compositum, extus brunnearum, intus hyalinarum, fuscioribus in regione ostiolarum. Paraphyses hyalinae, ramosae vel simplices, septatae, paulo constrictae ad septa, apice attenuato, longiores quam asci. Asci 90–119 \times 4–5 μm (in medio 106 \times 4.5 μm), octospori, cylindrici, pedicellati, unitunicati, hyalini, persistentes, sursum rotundati, apparatu apicali J^+ , 1.5–2.2 \times 1–1.5 μm (in medio 1.9 \times 1.25 μm). Ascospores 15.5–20.5 \times 3–4.5 μm (in medio 18.5 \times 3.5 μm), uniseriatae, ellipsoideae, fusiformes, apice acuminato, hyalinae, bicellulares, inaequaliter euseptatae, aliquando appendicibus mucilaginosi ad basim praeditae.

Ascomata 196–250 μm high, 215–280 μm diam, immersed under a blackened clypeus, globose to subglobose, coriaceous, solitary; ostiole papillate, periphysate. *Peridium* thin (6.5–8.5 μm), comprising 4–6 cell layers, brown outwardly, hyaline inwardly,

and often dark brown around the base. *Paraphyses* 5–8 μm wide at base, septate, slightly constricted at septa, tapering at apex, longer than asci. *Asci* 90–119 \times 4–5 μm (av. 106 \times 4.6 μm , $n = 15$), 8-spored, unitunicate, cylindrical, pedicellate, persistent, rounded at the apex, with a cylindrical, subapical J^+ ring, 1.5–2.2 high, 1–1.5 μm diam (av. 1.92 \times 1.25 μm ; $n = 25$). *Ascospores* 15.5–20.5 \times 3–4.5 μm (av. 18.64 \times 3.46 μm ; $n = 25$), overlapping uniseriate, long fusiform hyaline, 2-celled, apical cell long obclavate (13–16 \times 3–4.5 μm), guttulate, tapering to a point, basal cell cylindrical, tapering to a rounded end (6–8 \times 2–2.5 μm), usually with mucilage at the end.

Mode of life: Saprobic on dead leaves of *Castanopsis diversifolia* (Fagaceae).

Known distribution: Thailand (Doi Suthep, Chiang Mai).

Holotype: Thailand, Chiang Mai Province, Doi Suthep Pui National Park, altitude 1146 m, 18°48.402' North, 98°54.617' East, on dead leaves of *Castanopsis diversifolia* (Fagaceae), 14 Aug. 2003, Duong Minh Lam (holotype at Mushroom Research Centre, Chiang Mai, Thailand; isotypes in MRC DLA 008, HKUM 17498, CMU H224410, and PDD 78748; ex-type living cultures in CBS and HKUCC 10344).

Additional specimens examined: Thailand, Chiang Mai Province, Doi Suthep Pui National Park, altitude 1146 m, 18°48.402' North, 98°54.617' East, on dead leaves of *Castanopsis diversifolia* (Fagaceae), 15 Aug. 2003, Duong Minh Lam (Mushroom Research Centre, Chiang Mai, Thailand); *ibid.*, 4 Oct. 2003 (Mushroom Research Centre, Chiang Mai, Thailand); 20 Nov. 2003 (Mushroom Research Centre, Chiang Mai, Thailand); and 25 Jun. 2004 (Mushroom Research Centre, Chiang Mai, Thailand).

DNA analyses

Small subunit (18S) dataset: This DNA matrix consisted of 28 taxa with *Pleospora betae* and *Dothidea insculpta* as outgroups. The dataset was aligned without problems, but ambiguous taxa from other amphispheariaceous and xylariaceous genera, which would not properly align, were excluded from the alignment and analyses. The final aligned dataset was 968 characters, out of which 159 were parsimony informative, 58 parsimony uninformative and 751 constant characters. Parsimony analysis treating gaps as missing state and unequal weighting generated two trees, which were similar in topology and not significantly different from each other (based on KH and Templeton tests as implemented in PAUP). Tree length was 410 with a $-\text{LnL}$ of 3858.135. Weighted parsimony with a transition transversion of 1.5 to 1

resulted in two trees which are topologically identical to each other except that the *Xylariaceae* clade was more resolved when the dataset was bootstrapped. The strict consensus tree generated from this weighted parsimony was not significantly different from the strict consensus tree generated from the unweighted parsimony (Table 2). A transition transversion (TT) ratio of 1.5 to 1 was used for subsequent analyses, as 1.56 was the estimated value from ML. Figure 10 shows the relationships of *Emarcea castanopsidicola* with other amphispheariaceous and xylariaceous members. Clearly, this new taxon fits in the *Xylariales* and has 60 % bootstrap support. ML analyses of the same dataset under different models of nucleotide substitution resulted in identical tree topologies (results not shown). Treating gaps as fifth state did not affect tree topologies regarding the taxonomic placement of the ingroup under investigation.

Large subunit (28S) dataset: The 34 taxa formed an aligned data matrix of 900 characters in length and consisted of 31.5 % of parsimony informative characters. *Dothidea sambuci* and *Pleospora herbarum* were used as outgroups. Tree indices for different trees obtained have been summarised in Table 2. Unweighted parsimony resulted in 3 trees with a tree length of 1204 and a $-\text{LnL}$ of 7723.008, whereas weighted parsimony with a TT ratio of 1.5 : 1 (as estimated from ML analyses) resulted in a single most parsimonious tree with a tree length of 1446 and a $-\text{LnL}$ of 7708.490. This tree is shown in Fig. 11 with bootstrap values from 1000 replicates. Bootstrap values support the position of *Emarcea castanopsidicola* in the *Xylariales* (74 %) and it forms a putative monophyletic group with *Xylaria* species and other members from the *Amphisphaeriaceae* and *Cainiaceae* with 99 % bootstrap confidence.

Table 2. Summary of the tree indices, Kishino Hasegawa & Templeton Tests on trees obtained under different criteria.

	TL	CI	RI	RC	HI	No of trees (PIC)	$-\text{Ln L}$	KH/Templeton Tests of strict consensus ^a
18S								
TTr = 1:1	410	0.649	0.782	0.508	0.351	2 (159) *	3858.135	Best tree
TTr = 1.5:1	490.5	0.650	0.789	0.513	0.350	2 (159) *	3858.135	P = 1.00 / P = 1.00
TTr = 2:1	571	0.651	0.793	0.517	0.349	8 (159) *	3885.104	P = 0.0081 / P = 0.0156
28S								
TTr = 1:1	1204	0.515	0.699	0.360	0.485	3 (283) *	7723.008	P = 0.0004 / P = 0.0020
TTr = 1.5:1	1446	0.516	0.700	0.361	0.484	1 (287) *	7708.490	Best tree
TTr = 2:1	1690	0.516	0.699	0.361	0.484	1 (287) *	7711.363	P = 0.6951 / P = 0.8238
ITS								
TTr = 1:1	530	0.745	0.810	0.604	0.255	26 (172) *	3266.192	P = 0.0066 / P = 0.0078
TTr = 1.5:1	635.5	0.741	0.816	0.605	0.259	13 (177) *	3262.711	Best tree
TTr = 2:1	777	0.739	0.820	0.606	0.261	26 (177) *	2277.557	P = 0.0022 / P = 0.0020

TTr = Transition Transversion ratio; TL = tree length; CI = Consistency Index; RI = Retention Index; RC = Rescaled Consistency Index; HI = Homoplasy Index; $-\text{Ln L}$ = - Log Likelihood; KH = Kishino and Hasegawa test; PIC = Parsimony informative characters. *Subsequent values for $-\text{Ln L}$, KH and templeton tests were done with the strict consensus trees. ^aProbability of getting a more extreme T-value under the null hypothesis of no difference between the two trees (two tailed test) with significance at $P < 0.05$.

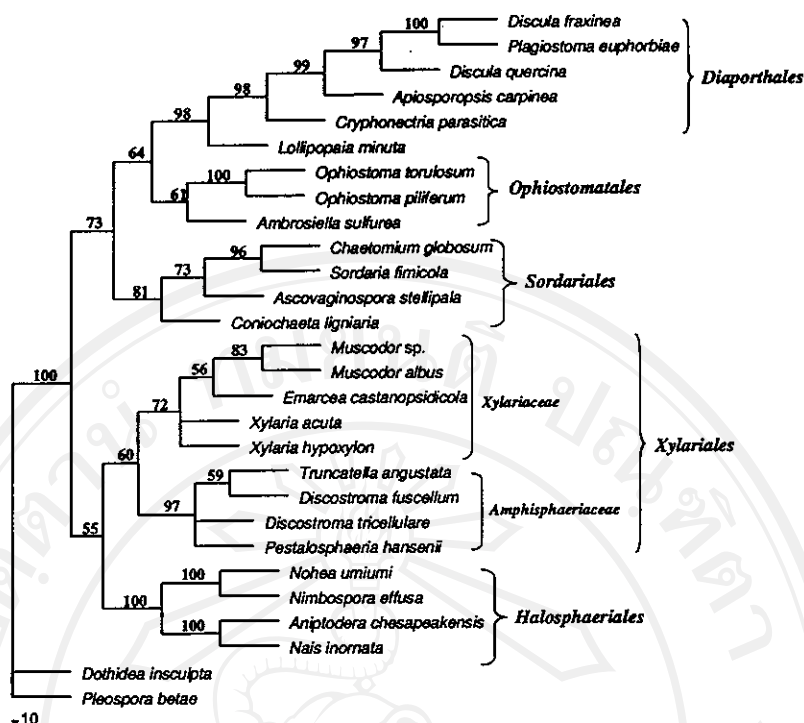


Fig. 10. Phylogenetic tree based on partial 18S DNA sequences. The tree was rooted with *Dothidea insculpta* and *Pleospora betae* and constructed under the Maximum Parsimony criterion with a transition transversion ratio of 1.5 : 1. The number at each branch point represents percentage bootstrap support calculated from 1000 replicates. Branch lengths are proportional to the numbers of nucleotide substitutions and are measured by scale bar (Bar, 10 % sequence divergence).

Trees from unweighted and weighted parsimony were significantly different from each other (Table 2). ML analyses under the HKY model with an estimated TT ratio of 1.6 and estimated shape parameter of 0.3 gave similar tree topologies (results not shown). Estimated base frequencies were as follows: A = 0.205; C = 0.261; G = 0.268; T = 0.263.

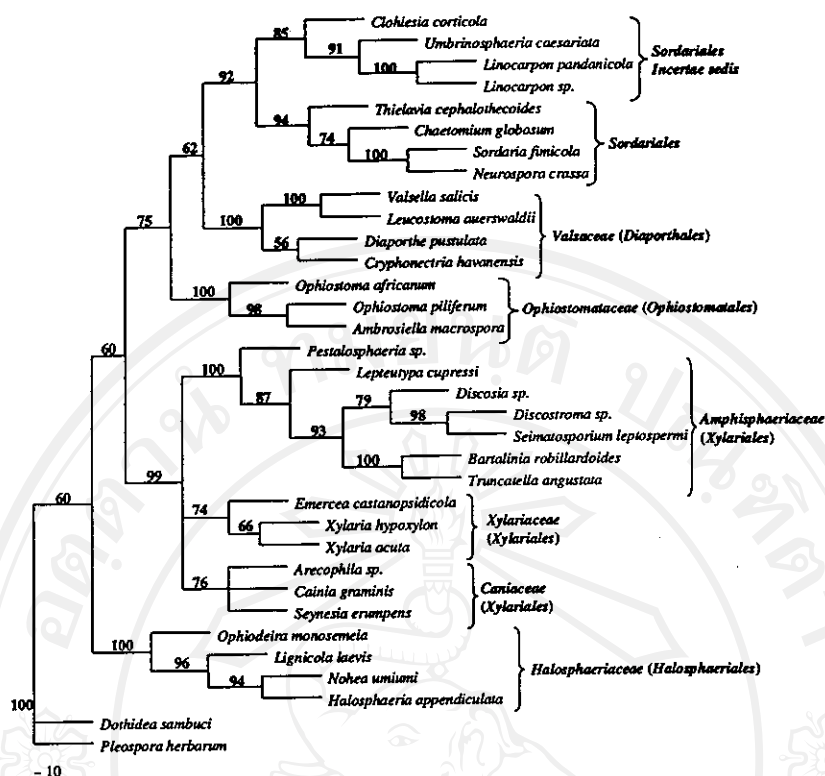
ITS (+5.8S) dataset: A dataset consisting of 19 taxa from *Xylaria* and *Muscodora* (*Xylariaceae*) and two species from *Diatrypaceae* (*Diatrype flavovirens* and *Cryptosphaeria ligniota*) included as outgroups were used in MP analysis. This dataset contained 600 characters (177 [29.5 %] parsimony-informative; 320 constant and 103 parsimony-uninformative). As shown in Table 2, weighted parsimony gave better tree topologies and *E. castanopsidicola* was found to have close phylogenetic affinities with other *Muscodora* species (results not shown). Other taxa were not included in the analysis as the sequences appeared to be divergent and hence could not be properly aligned.

DISCUSSION

Morphological characters, such as the ascomata being immersed beneath a clypeus, papillate ostioles and unitunicate asci with a J⁺ subapical ring, indicate that *E. castanopsidicola* should be placed in *Xylariales*

(*sensu* Kirk *et al.* 2001), where it could be included in the *Amphisphaeriaceae* or the *Xylariaceae*. Most amphisphaeriaceous species however, have brown two-celled ascospores and *Pestalotiopsis*-like anamorphs (Barr 1994, Kang *et al.* 1998, 1999, Jeewon *et al.* 2003c). On the other hand, most taxa in the *Xylariaceae* have a well-developed stroma and unicellular brown ascospores, invariably with a germ slit and produce mostly hyphomycetous anamorphs in culture (*sensu* Kirk *et al.* 2001). In some genera, however, the stroma is reduced to a clypeus (e.g. *Anthostomella*, *Fasciatispora*) and a sporodochial *Geniculosporium* anamorph has only been determined in a handful of species (Hyde & Goh 1998, Lu & Hyde 2000). *Emarcea* has hyaline bicellular ascospores and did not produce a *Pestalotiopsis* anamorph (or any anamorph) in culture. Morphological characteristics do not clearly indicate whether *Emarcea* should be placed in *Amphisphaeriaceae* or *Xylariaceae* and therefore we decided to use rDNA sequence analysis.

In the *Xylariaceae*, *Emarcea* should be compared with *Anthostomella*. *Anthostomella* species often have ascospores with one large brown cell and one dwarf cell, e.g. *A. clypeata*, *A. clypeoides*, *A. foveolaris*, *A. rostrispora*, *A. triangularis* and *A. unguiculata* (Lu & Hyde 2000). *Anthostomella* species, however, always have brown ascospores, usually with germ slits, and this has not been observed in mature or old material of *Emarcea*.



- 10

Fig. 11. Evolutionary relatedness of *Emarcea castanopsidicola* based on a Maximum Parsimony analysis of the 28S rDNA partial gene sequence with no molecular clock enforced. This tree is topologically identical to the Maximum Likelihood tree. Designated outgroups are *Pleospora herbarum* and *Dothidea sambuci*. Transition was weighted 1.5 times. Bootstrap support based on 1000 replicates for each clade shown on the branches.

Phylogenies generated using Maximum Parsimony and Maximum Likelihood from the three different datasets (18S, 28S and ITS) support the monophyly of the xylariaceous species under investigation. The small subunit dataset (18S) shows that *Emarcea castanopsidicola* is nested in a clade with *Muscodor* and *Xylaria* species. The *Xylariaceae* clade is connected to the *Amphisphaeriaceae* clade with a bootstrap support of 60 % (Fig. 10). Together these two clades constitute the *Xylariales*. This gene region (18S) evolves rather slowly (White *et al.* 1990) and provides valuable insights into the systematics of *Emarcea castanopsidicola* and its familial placement at the ordinal and familial level. Similar molecular findings were obtained from phylogenies derived from the large sub-unit (28S). *Emarcea castanopsidicola* is phylogenetically related to *Xylaria curta* and *X. hypoxylon* in the *Xylariales* (Fig. 11). The ITS dataset provides further sequence-based evidence to elucidate relationships of *Emarcea castanopsidicola* with other members of the *Xylariaceae*. *Emarcea castanopsidicola* forms a sister group to *Muscodor* species with 100 % bootstrap confidence. This is not unexpected as *Muscodor*, a recently described new anamorphic genus, bears close phylogenetic affinities to *Xylaria* (Worapong *et al.* 2001). Worapong *et al.* (2001) analysed genetic sequences of this endophytic fungus and found that partial 18S rDNA sequences and the entire ITS sequences (including 5.8S) share a high

degree of homology with other *Xylaria* species. Phylogenies based on Maximum Parsimony from their study also revealed that *Muscodor* is more closely related to the *Xylariaceae* than to the *Amphisphaeriaceae*. Similar findings are reported here, and given that *Emarcea castanopsidicola* forms a sister group to *Muscodor* in all our analyses, there is no doubt that our new taxon should be accommodated in the *Xylariaceae*.

In this study, we have been quite selective in the ingroups that we used. Several xylariaceous species, however, could not be included (e.g. *Anthostomella*, which appears to have close morphological affinities to *Emarcea castanopsidicola*) because of ambiguous sequence alignment and secondly due to the high degree of polytomies that were encountered in the phylogenetic analyses, especially when the molecular dataset was subjected to bootstrap analyses. Similar results were reported by Smith *et al.* (2003). Morphological observations are therefore still very important in establishing familial placement in this group of fungi. Based on molecular results and morphological examination, there is conclusive evidence to conclude that our new taxon, *Emarcea castanopsidicola*, belongs in the *Xylariaceae*.

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15

Fungi on leaf litter

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Fungal communities occurring on leaf litter have been studied for a relatively long time in temperate regions but few studies have been undertaken in the tropics. In Thailand there have been some studies of fungi on decaying leaves in tropical forests. Species composition in leaf litter differs from area to area and between tree species. Different study methods result in very different species composition. A very large number of rare taxa and sterile mycelium are usually isolated by the particle filtration method. Fewer, but sporulating species are identified by direct examination. The most common species on decaying leaves are listed. Relationships between fungi, host and geographical distribution of the fungi are discussed.

Key words: decaying leaves, diversity, leaf fungi, particle filtration

Introduction

Leaves are one of the three indispensable structures of a tree. Fallen leaves create an enormous biomass that provides a huge carbon and nitrogen source (Lodge, 1993) for microorganisms. Follicolous fungi occur on leaves and can include taxa living within the tree canopy on living leaves and also taxa in leaf litter on the forest floor (Kirk *et al.*, 2001). They include biotrophic taxa such as pathogens and endophytes. This chapter deals with saprobic fungi occurring on leaves in forest leaf litter.

There have been several studies of fungi occurring on leaves in both temperate and tropical regions (Aoki *et al.*, 1990; Bills and Polishook, 1994a,b; Castaneda Ruiz *et al.*, 1998; Osono, 2002; Tokumasu *et al.*, 1994). Particle filtration or direct observations are the methods generally used to study leaf litter fungi. Particle filtration generally results in larger numbers of isolates, with numerous rare species (Polishook *et al.*, 1996; Paulus and Hyde, 2003, 2004). Many of the fungi obtained by particle filtration are, however, sterile mycelia. Dick (1990) detailed pathogenic microfungi on leaves of *Eucalyptus* in New Zealand. Bills *et al.* (1994a) used particle filtration to estimate fungal diversity on mixed leaf litter in lowland forest of Costa Rica and 177 species were recorded. These authors also used the same method for studies on fungal diversity in a single tree of *Heliconia mariae* (*Heliconiaceae*) and 137 species were identified (Bills *et al.*, 1994b). Bills *et al.* (1994a,b) isolated a large number of rare species and few common species by particle filtration from leaf litter in Costa Rica. Paulus and Hyde (2003) studied diversity of microfungi on decaying leaves of *Ficus pleurocarpa* by using the same method as Bills *et al.* (1994) and more than 100 species were identified. It is not clear whether all fungi isolated via the particle filtration method are involved in

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Fungi on leaf litter

litter decay or whether dormant spores are isolated. Therefore the direct observation method is preferred in identifying fungal communities on leaf litter, although it results in fewer species. Parungao *et al.* (2002) directly studied fungi on decaying leaf litter of 13 tree species in Australia resulting in 57 species. Paulus and Hyde (2003) used particle filtration and direct observations to estimate fungal diversity on leaf litter of *Neolitsea dealbata* (Lauraceae) and 86 species were recorded by the first method and 28 species by the latter, with 14 species overlapping. Both methods however, indicate that there is a high diversity of microfungi on leaf litter.

Diversity of leaf litter fungi

There were few abundant species and a high proportion of rare species (Bills *et al.* 1994a,b; Paulus and Hyde, 2003). In another study of fungal diversity on leaf litter of *Ficus pleurocarpa* (Moraceae) 104 species were recorded by using the direct method and 153 species by using the particle filtration method (Paulus and Hyde, 2004). Kirk (1983) studied the microfungi on leaf litter of *Laurus nobilis* (Lauraceae) by using direct observation and 126 species were recorded. The above studies obtained higher numbers of fungi than the estimated ratio of 6 unique fungi per tree species (Hawksworth, 1991).

Some fungi appear to be common on leaf litter in many studies. They include: *Beltrania rhombica*, *Beltraniella portoricensis*, *Chaetopsina fulva*, *Hansfordia pulvinata*, *Hypocreales* spp., *Idriella lunata*, *Selenosporella curvispora* and *Zygosporium echinosporium* (Heredia, 1993; Bills and Polishook 1994a,b; Parungao *et al.*, 2002; Paulus and Hyde, 2003; Promputtha *et al.*, 2002). In addition many new fungal taxa have been described from decaying leaves (Kirk, 1983; Castaneda Ruiz *et al.*, 1998; Ono, 2001; Promputtha *et al.*, 2003, 2004a,c,d; Duong *et al.*, 2004).

Studies on fungal diversity on leaf litter from various hosts have revealed different fungal communities, often with little overlap (Parungao *et al.*, 2002; Polishook *et al.*, 1996). Parungao *et al.* (2002) found that only 36 (63%) of the 57 taxa identified on leaves of 13 tree species in North Queensland occurred only on one leaf type and no fungi were present on all. Similarly there was only 25-28% overlap between fungi on *Guarea guidonia* and *Manilkara bidentata* in the same forest location in Puerto Rico (Polishook *et al.*, 1996). The reasons for the low overlap are not clear, but may be related to host or leaf chemistry. Paulus *et al.* (2004) found the presence of small amounts of chemicals such as glycol, chlorohydrin and bromohydrin in leaves may effect fungal presence. Ghimire and Hyde (2004), however, suggested that endophytes may become saprobes once leaves senesce and this may account for differences in fungal communities on different hosts. Studies to date have concentrated on fungal communities on litter of unrelated plant species and this may account for the lack of overlap of fungi on the various hosts. Therefore to understand the diversity of fungal communities on leaf fungi we need to investigate a greater number of hosts (both closely and distantly related) as well as different geographic regions. In this way we may resolve the question as to whether leaf fungi are host-specific, host-recurrent and/or dependent on geographical location.

Host- and geographical- specificity of leaf fungi

Photita *et al.* (2001) studied fungal diversity on *Musa acuminata* (Musaceae) and came to the conclusion that species composition on one leaf was different between petiole and blade areas. Percentage overlap between these two regions was 49%. This

indicates that microhabitats can influence the distribution of microfungi on leaf litter. Some species are only known from *Musa* spp., however, some of these species also occur on leaf litter of *Heliconia mariae* (Bills *et al.*, 1994b). This may be because *Heliconia* and *Musa* are structurally similar. Typical primary saprobic species in the tropics are *Curvularia* spp. (Tokumasu *et al.*, 1997; Ellis, 1971, 1976). In contrast, some members of the common primary saprobes in temperate zones are absent or very rare in the tropics. The conclusion that microfungi are heavily influenced by climatic and geographical factors have been noted elsewhere (Van Maanen *et al.*, 2000; Tokumasu and Aoki, 2002).

Table 1. List of common microfungi on decaying leaves using the particle filtration method (Queensland, Australia: Paulus *et al.*, 2003, 2004; Costa Rica: Bills *et al.*, 1994a,b; Polishook *et al.*, 1996; Germany: Tokumasu *et al.*, 1994; Japan: Osono, 2002., Tokumasu and Aoki, 2002. and Thai forests: Tokumasu *et al.*, 1997).

Species	
<i>Acremonium</i> spp.	<i>Fusarium</i> spp.
<i>Alysidiopsis</i> sp.	<i>Idriella lunata</i>
<i>Aspergillus</i> spp.	<i>Idriella</i> spp.
<i>Aureobasidium pullulans</i>	<i>Mucor</i> spp.
<i>Beltrania rhombica</i>	<i>Penicillium</i> spp.
<i>Beltraniella portoricensis</i>	<i>Phoma</i> spp.
<i>Chaetopsina fulva</i>	<i>Phomopsis</i> spp.
<i>Cladosporium cladosporioides</i>	<i>Rhinochadiella</i> sp.
<i>Cladosporium</i> spp.	<i>Stachybotrys parvispora</i>
<i>Colletotrichum</i> spp.	<i>Thozetella radicata</i>
<i>Cyphellophora taiwanensis</i>	<i>Trichoderma harzianum</i>
<i>Dactylaria</i> spp.	<i>Trichoderma</i> spp.
<i>Dictyochoaeta</i> spp.	<i>Verticillium</i> spp.
<i>Fusarium solani</i>	<i>Volutella</i> spp.

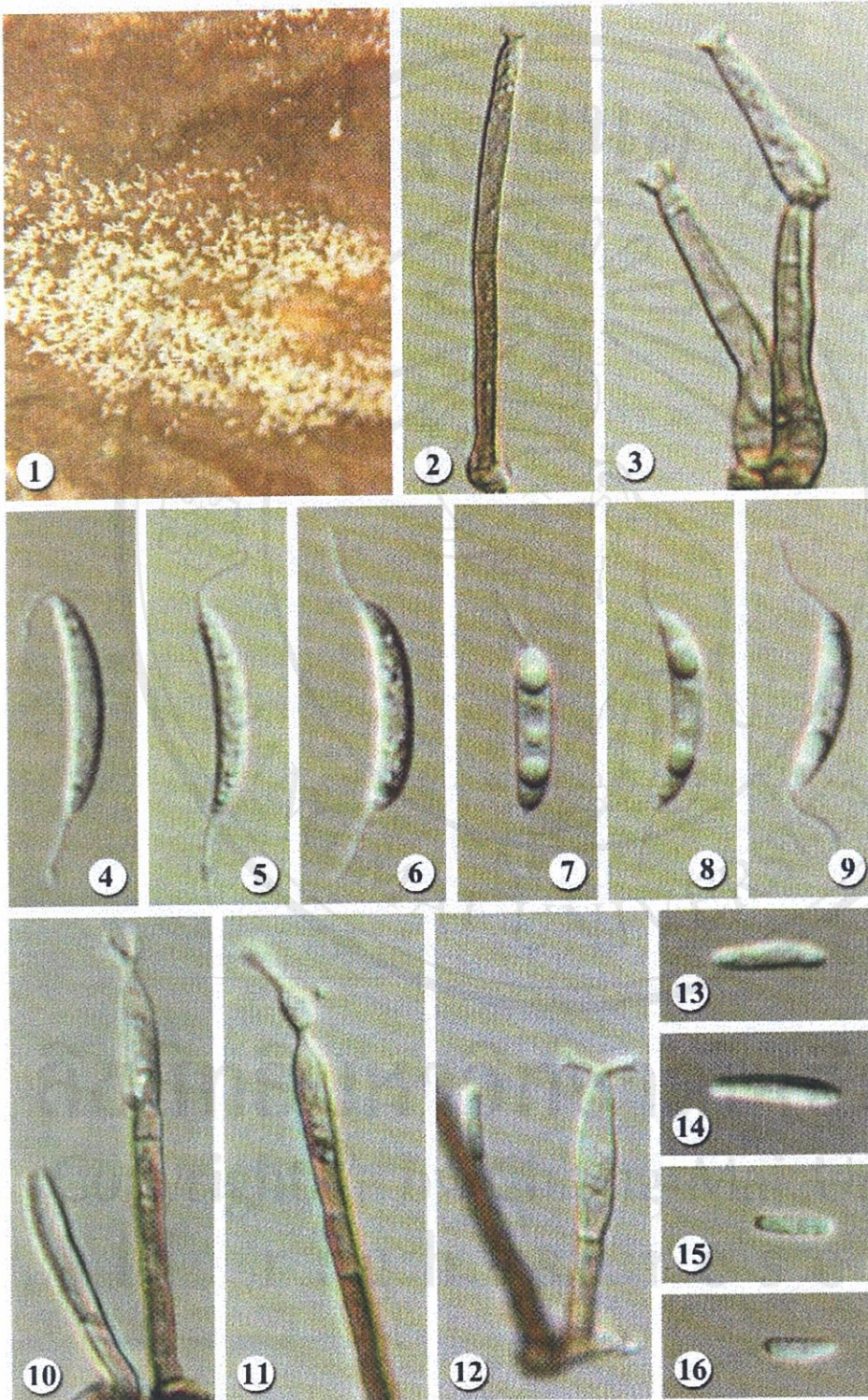
Table 2. List of common fungi on decaying leaves using the direct method (from Parungao *et al.*, 2002; Promputtha *et al.*, 2002, 2004b; Paulus *et al.*, 2004 and present results from Thailand).

Species	
<i>Acremonium</i> spp.	<i>Hypocreales</i> spp.
<i>Beltrania rhombica</i>	<i>Idriella lunata</i>
<i>Beltraniella portoricensis</i>	<i>Lophodermium</i> sp.
<i>Chaetopsina fulva</i>	<i>Ophioceras</i> spp.
<i>Cladosporium</i> spp.	<i>Selenosporella curvispora</i>
<i>Dictyochoaeta</i> spp.	<i>Zygosporium echinosporium</i>

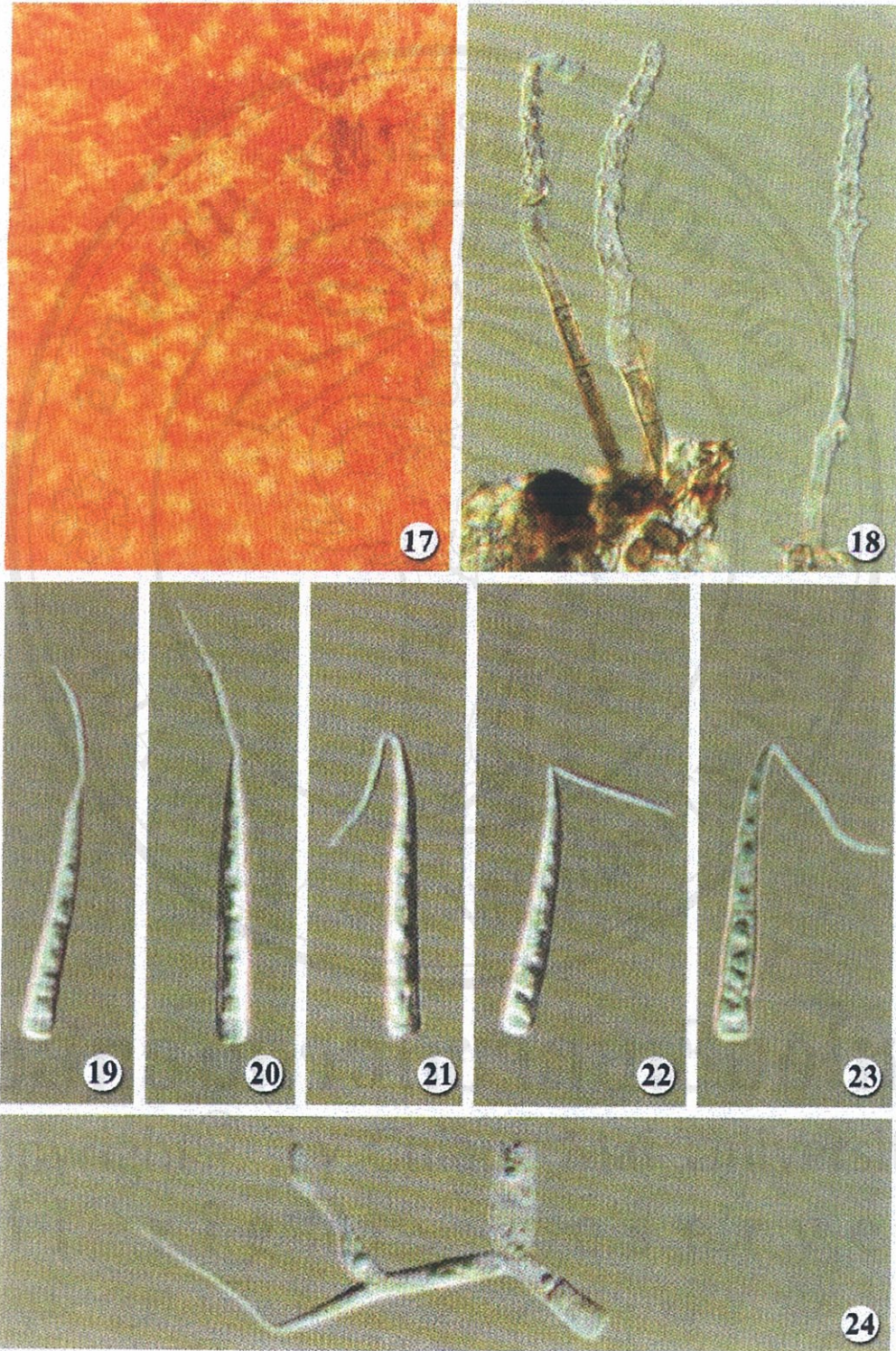
Foliicolous fungi in Thailand

There have been several studies on the diversity of fungi on leaf litter in Northern Thailand (Tokumasu *et al.*, 1997; Promputtha *et al.*, 2002, 2003; Bussaban *et al.*, 2003). Microfungal communities on decaying pine needles were studied by Tokumasu *et al.* (1997) and 127 taxa were recorded, and the most common species on pine leaf litter in all 5 locations in Thailand were: *Alysidiopsis* sp., *Cladosporium cladosporioides*, *C. musae*, *C. tenuissimum* and *Mucor* spp. Other common species were *Aureobasidium pullulans*, *Chalara* spp., *Pestalotiopsis* sp. and *Scolecobasidium humicola*, while

Fungi on leaf litter

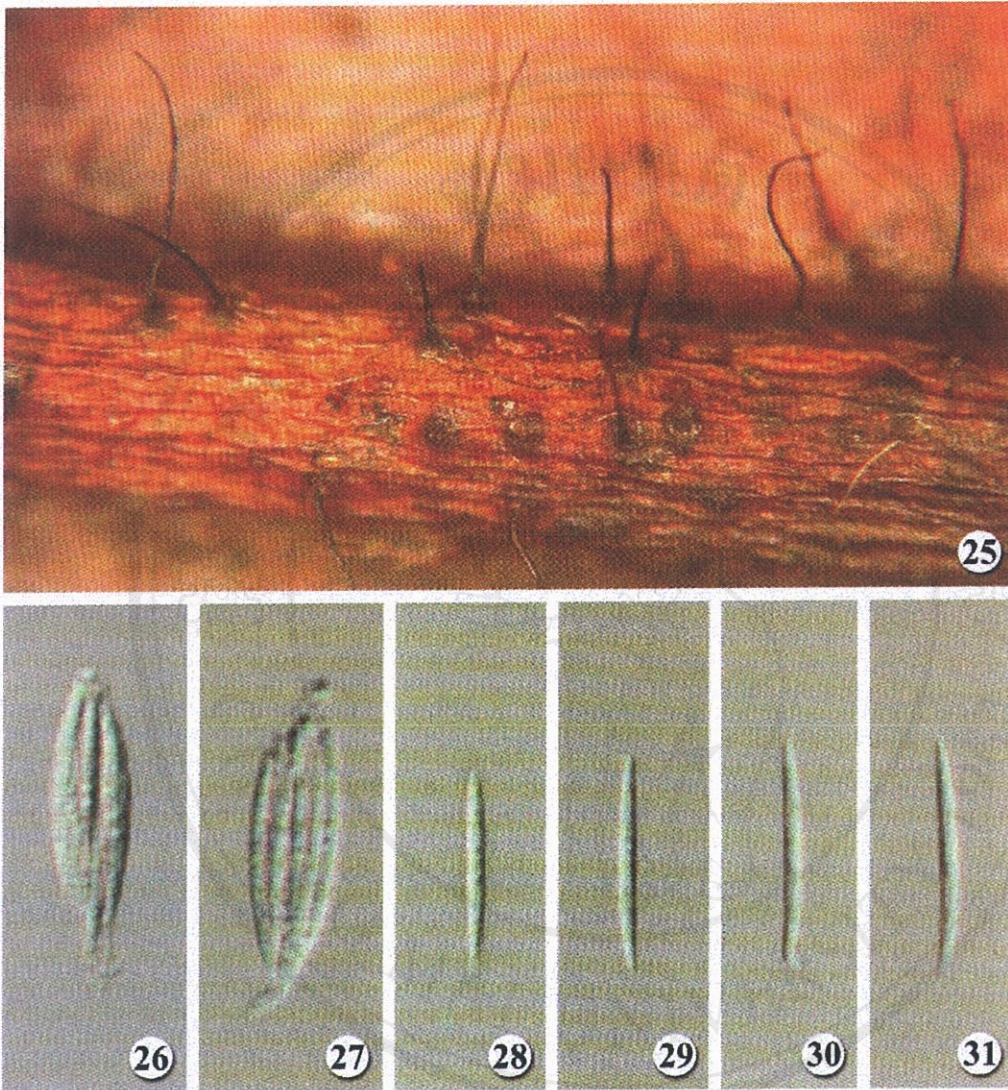


Figs 1-16. *Dictyochaeta* spp. 1-9. *Dictyochaeta simplex*. 10-16. *Dictyochaeta cylindrospora*. 1. Colonies on leaf surface. 2, 3. Conidiophores. 4-9. Conidia.



Figs 17-24. *Subulispora procurvata*. 17. Colonies on leaf surface. 18. Conidiophores. 19-23. Conidia. 24. Germinated conidium.

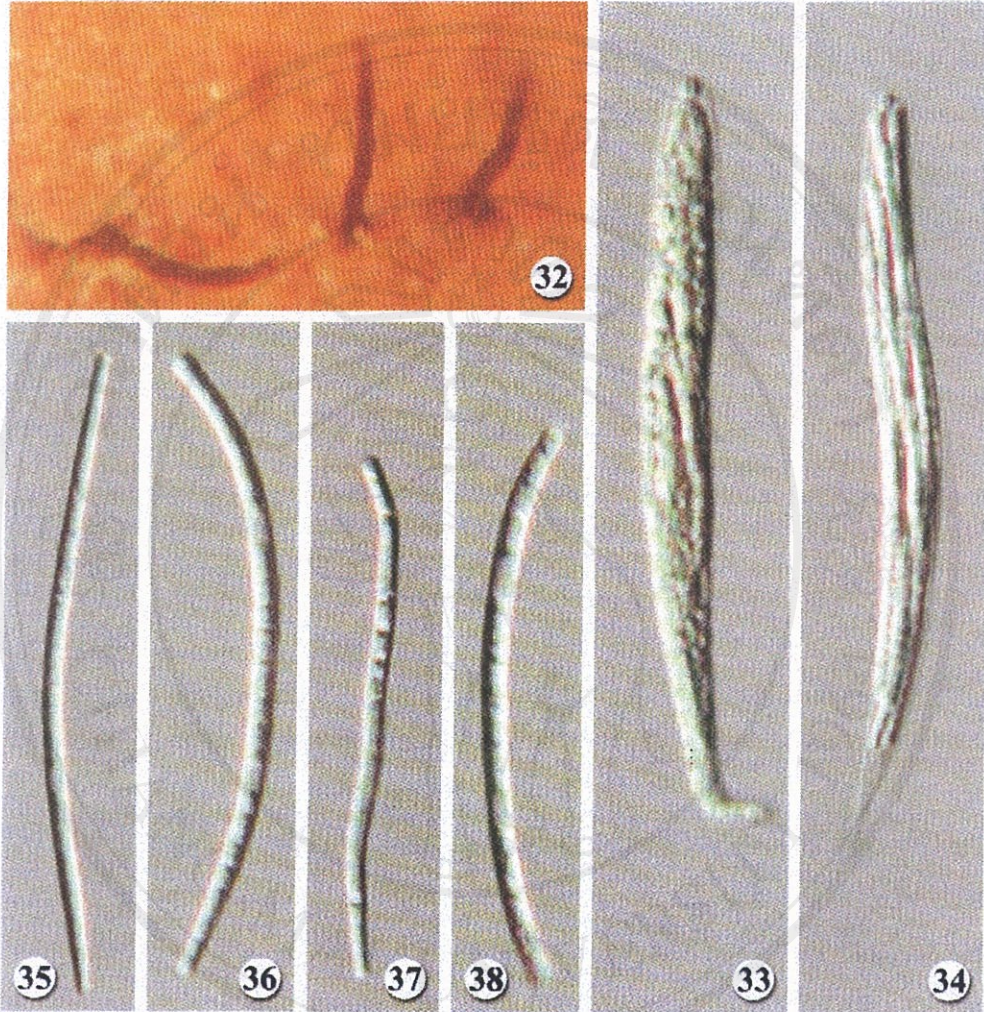
Fungi on leaf litter



Figs 25-31. *Gnomonia amoena*. 25. Immersed ascomata with long necks. 26, 27. Asci. 28-31. Ascospores.

Dictyochaeta spp. and *Scolecobasidium humicola* appeared to be restricted to partly discolored needles (*Pinaceae*). Common taxa on pine needles in Germany and Japan were *Aureobasidium pullulans*, *Chalara* spp., *Cladosporium* spp., *Dactylaria* spp., *Pestalotiopsis* spp. and *Scolecobasidium humicola* (Aoki *et al.*, 1990). Some taxa with high abundance in Japan, eg. *Dactylaria fusiformis*, *Thozetella crista* and *D. naviculiformis*, were however, not abundant on pine needles in Thailand. Several fungal taxa involved in the succession of pine needles in the tropics also differ from those in temperate regions (e.g. *Cladosporium tenuissimum* and *C. macrocarpum* are common in the tropics, but rare in temperate forests (Tokumasu *et al.*, 1997).

The fungi recorded from pine needles differ from those on monocotyledon and angiosperm litter and this is probably because the pine litter studies utilized agar for their sampling, while other studies involved direct observation.



Figs 32-38. *Ophioceras commune*. 32. Ascomata with long necks. 33, 34. Asci. 35-38. Filiform ascospores.

Promptuttha *et al.* (2002) studied the succession of fungi on *Magnolia lillifera* (*Magnoliaceae*) baited leaf litter in Chiang Mai, Thailand and 22 microfungi were recorded. They also studied natural litter of the same host including 37 fungi, comprising 20 ascomycetes and 17 anamorphic fungi (Promptuttha *et al.*, 2004). Common species included *Bionectria ochroleuca*, *Cylindrocladium floridanum*, *Gliocladium* sp., *Dokmaia monthadangii*, *Gliocladium* sp. 1, *Hyponectria manglietiae* sp. nov, *Hyponectria manglietiagarrettii* sp. nov, *Hypoxyylon* sp., *Lasiosphaeria* sp., *Pseudohalonectria suthepensis* and *Sporodesmium crassisporium* being the most common species. These species are common fungi on leaf litter in other studies (Paulus and Hyde, 2004). These studies conclude that fungal diversity is higher during the early decay process, but differs from fungal succession in temperate forests (Kendrick and Burges, 1962). Most of recorded fungi from natural litter of *Magnolia lillifera* (Promptuttha *et al.*, 2004b) were ascomycetes. This is different from all previous studies

Fungi on leaf litter

on foliicolous fungi. Of the 37 recorded fungi, 7 species (19%) were new to science (*Anthostomella monthadoia*, *Dokmaia monthadangii*, *Hyponectria manglietiae*^{*}, *Hyponectria manglietiagarrettii*^{*}, *Hyponectria suthepensis*^{*}, *Munkovalsaria bipolaris*^{*} and *Pseudohalonectria suthepensis*). These results indicate possible host-specificity of microfungi on leaves in the tropics and a high potential for discovering new taxa in Thailand. Common species in these studies differ indicating that microfungi in the tropics are as diverse as their hosts.

We are studying the fungi on litter of various tree species in Chiang Mai Province and are finding numerous fungi including several new species (e.g. Duong *et al.*, 2004). Common hyphomycetes are *Dicryochaeta simplex* (Figs 1-9), *D. cylindrospora* (Figs 10-16), *Subuldispora procurvata* (Figs 17-24), while common ascomycetes are *Gnomonia amoena* (Figs 25-31) and *Ophioceras commune* (Figs 32-38).

Role and importance of Thai litter fungi

Fungi play an essential role in the decay of organic material in tropical forests, recycling inorganic nutrients for other organisms. Many insects also rely on their mycelium as an important food source. Many fungi produce inhibitory compounds in defense of territory and capture resource. They may therefore be potentially an excellent biological resource in the search for new antibiotics and novel compounds (Hyde, 2001).

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^{*}These taxa are presently being prepared for publication.

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DGGE coupled with ribosomal DNA gene phylogenies reveal uncharacterized fungal phylotypes

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Most fungal diversity studies have previously been based on morphological examination and cultivation methods. In this study we use a molecular method based on DGGE coupled with sequence analysis of 18S rRNA gene to assess fungal diversity on leaves of *Magnolia liliifera*. To achieve this, we extracted total genomic DNA and used fungal specific primers (NS1 and GCFung) to obtain fungal sequences. PCR-DGGE analysis recovered 14 operational taxonomic units (OTU) from different parts of the studied leaves. Phylogenetically, 8 OTUs belonged to the order *Pleosporales* and other bitunicate ascomycetes; 2 and 3 were related to the *Xylariaceae*, (*Xylariales*) and *Hypocreales*, respectively; 1 OTU was phylogenetically affiliated with the *Rhytismatales*. While this molecular approach identified taxa that were not recovered from morphological or cultural studies, it did not detect other taxa that were predominantly isolated using traditional methods. The three different parts of one leaf tested (petioles and midribs, leaf blade lower and upper parts) yielded different fungal taxa that possible indicate tissue-recurrence. The findings are compared with previous studies on the same host where endophytes were investigated using traditional culturing techniques.

Key words: DGGE, endophytes, fungal diversity, leaf fungi, phylogeny, rDNA, unculturable fungi

Introduction

Plant associated fungi are highly diverse, with saprobes, endophytes and pathogens occurring in all plant species examined, e.g *Proteaceae* (Lee *et al.*, 2005), *Magnolia liliifera* (Promputtha *et al.*, 2005a), grasses (Bacon and White, 1994), palms (Fröhlich *et al.*, 2000; Rodrigues, 1996; Taylor *et al.*, 1999), banana (Brown *et al.*, 1998; Photita *et al.*, 2004a,b) and mangroves (Suryanarayanan and Kumaresan, 2000). Fungal endophytes live inside plants for all or part of their life cycle without causing any disease symptoms or tissue

damage (Wilson, 1995). The endophytes may also become saprobes when the leaves senesce (Boddy and Griffith, 1989; Petrini, 1991; Photita *et al.*, 2004a,b). De Bary (1866) was the first to observe endophytes and ever since then they have been widely studied (Petrini, 1991; Photita *et al.*, 2004a,b; Suryanarayanan and Thennarasan, 2004; Vettraino *et al.*, 2005). Endophytic fungi play important roles in plant life, forming symbiotic associations, facilitating nutrient cycling between plant and fungus, enhancing plant growth, increasing resistance and producing toxins that protect plants from animal and insect herbivores (Bultman and Murphy, 2000; Clay, 1987).

Endophytic fungi have previously been identified based on morphological characters from sporulating isolates on artificial media (Guo *et al.*, 1998, 2003; Taylor *et al.*, 1999). The fungi that do not sporulate on media have been termed mycelia sterilia and often been grouped as morphospecies (Guo *et al.*, 2000, 2003; Promptuttha *et al.*, 2005a). Methods to promote sporulation in mycelia sterilia have been developed (Fröhlich *et al.*, 2000; Guo *et al.*, 1998; Taylor *et al.*, 1999) and proportions of non-sporulating endophytes range from 11-54% (Fisher *et al.*, 1994, Fröhlich *et al.*, 2000; Guo *et al.*, 2000; Kumar *et al.*, 2004). To resolve the problem of identifying non-sporulating isolates, DNA sequence-based methodologies have been successfully used for the phylogenetic placement and classification of morphospecies obtained as endophytes (Guo *et al.*, 2000, 2001, 2003; Promptuttha *et al.*, 2005a; Wang *et al.*, 2005).

Despite the advances in identifying endophytes by enhanced cultural techniques and molecular identification of morphospecies, studies are still flawed by the fact that fast-growing fungi will be isolated preferentially while unculturable fungi and slow-growing fungi will escape detection. Culture-independent methods for screening fungal diversity from natural samples are therefore necessary (Guo *et al.*, 2001; Kemp, 1994). With this in mind, Guo *et al.* (2001) developed a technique using direct amplification of rDNA gene extracted from frond tissue of *Livistona chinensis* followed by cloning, sequencing and phylogenetic analysis to identify endophytic fungi. They successfully recovered some endophytic fungi that had not previously been isolated from cultural studies. However, only 6 phylotypes were recovered: one plant, one basidiomycete and four ascomycetes. The most common endophytic fungi occurring on *Livistona chinensis*, such as *Guignardia*, *Pseudospiropes* and *Xylaria* species (Guo *et al.*, 2000) however, were not encountered.

Advances in technology now provide additional molecular tools to evaluate diversity, and to study ecology and phylogeny (Countway *et al.*, 2005; De Hoog *et al.*, 2005; Green *et al.*, 2004; Iotti *et al.*, 2005; Jeewon *et al.*, 2004; Le Bourhis *et al.*, 2005; Lim *et al.*, 2005). Denaturing gradient gel

electrophoresis (DGGE) is a technique that has effectively been used to estimate the diversity of prokaryotes and eukaryotes in natural samples (Anderson *et al.*, 2004; Countway *et al.*, 2005; Dar *et al.*, 2005; Díez *et al.*, 2001; Jeewon and Hyde, 2006).

DGGE has been successfully applied to document fungal communities (May *et al.*, 2001; Nikolcheva *et al.*, 2003, 2005; Vainio *et al.*, 2000, 2005). Vainio and Hantula (2000) studied wood-inhabiting fungi and found that phylotypes isolated from environmental samples were comparable to fungi isolated from the same substrate at varying depths. Vainio *et al.* (2005) used DGGE to test the effect of sample treatment using a commercial formulation of *Phlebiopsis gigantea* on fungal communities of treated samples. Two different amplification products were observed on average from a single piece of sample (approximately 500 mm³). The conclusion was that treatment of environmental samples did not reduce the overall fungal diversity within the treated plots. May *et al.* (2001) accessed fungal communities associated with whole plant corn silage. Results indicated that one inoculum dramatically influenced the fungal community. This method has, however, not yet been used in studies on fungal communities in living leaves, although Nikolcheva *et al.* (2003) investigated fungi on decaying leaves from freshwater from different hosts (alder, beech, linden, oak and red maple) and found that the highest diversity occurred one week after leaves were submerged. In another study, Nikolcheva *et al.* (2005) studied fungal communities occurring at the initial stage of leaf decaying in three plant species (linden, maple and oak) and found up to 7 operational taxonomic units (OTUs) on the second day, which was the highest biodiversity during the decay process.

In the present study we used DGGE to establish the fungal communities on living leaves of *Magnolia liliifera* collected from Doi Suthep Pui National Park, Chiang Mai, in Thailand. We chose this host because several studies had been published on endophytic and saprobic fungal communities on this host at this location (Promputtha *et al.* 2004, 2005a,b) that would permit comparison. The present work aims 1) to characterise the fungal communities based on DGGE coupled with phylogenetic analysis, 2) to reveal fungi that possibly are not recovered through cultural and microscopy techniques.

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Methods

DNA extraction

Living, healthy leaves of *Magnolia liliifera* were collected at Doi Suthep-Pui National Park (1146 m alt, 18°48.402' North, 98°54.617' East), Chiang Mai, Thailand. Three leaves were used for DNA isolation. The leaves were divided into three parts (petioles and midribs, lower and upper parts of leaf blades) in order to assess whether fungal distribution from different leaf parts is different. Each part was separately ground into powder state with liquid nitrogen. DNA isolations were carried out 3 times from each part of leaves, using a modified CTAB procedure as outlined by Jeewon *et al.* (2002, 2004; Cai *et al.*, 2005). Fifty milligrams of leaf powder was placed in a sterile 1.5 ml tube. 650 µl of pre-heated 2 × CTAB was then added, followed by 1 hour of incubation at 65°C. The same volume of pre-heated (65°C) phenol:chloroform : isoamylalcohol (25:24:1) was added (hot phenol extraction) into the sample tube and mixed gently. The contents were centrifuged at 14000 rpm for 30 min and the upper phase was transferred to another fresh tube. This step was repeated 3 times or until no interphase could be seen. DNA was precipitated from the supernatant (500 µl) by 2 volume of absolute ethanol (4°C) and kept at -20°C overnight. Then, samples were centrifuged at 4°C, 14000 rpm for 30 min. DNA pellets were washed in ice-cold 70% ethanol 2 times with centrifugation at 1000 rpm at 4°C in between and then dried in a vacuum. DNA samples were diluted in 75 µl of TE buffer containing RNase (10 mg.µl⁻¹) and incubated at 37°C for 2 hours and examined on 1% agarose gel containing ethidium bromide.

Polymerase Chain Reaction (PCR)

A partial sequence of 18S rDNA gene (about 320 bp) was amplified using primer pair NS1 (5'-GTAGTCATATGCTTGTCTC-3') (White *et al.*, 1990) and GCfung (5'-CGCCCGCCGCGCCCCGCGCCCCGGCCCCGCCGCCCGCCCCATTCCCCGTTACCCGTTG-3') as described by May *et al.* (2001). Each PCR reaction contained 5 µl of 10 × PCR buffer, 2 µl of MgCl₂ (25 mM), 4 µl of dNTP (2.5 µM) mixture, 1 µl of BSA (1 µg/µl), 0.3 µM of each primer, 0.8 units *Taq* Polymerase, and 10 ng template DNA. The optimized PCR thermal cycles for the primer pair NS1 and GCfung was as follows: initial denaturation at 95°C for 4 min and 35 cycles of 95°C for 1 min, annealing at 50°C for 1 min and 10 sec, extension at 72°C for 2 min, then followed by a last extension at 72°C for 8 min.

Denaturing Gradient Gel Electrophoresis (DGGE)

DGGE gels were prepared with the aid of a Bio-Rad model 475 Gradient Delivery System. The gels contained 7% (wt/vol) of acrylamide (acrolamide/bisacrolamide 37.5:1) and a range of denaturant concentration from 10% to 55% (formamide and urea). DNA concentration of PCR products was estimated by spectrophotometer and 2 µg products were loaded on DGGE gels. The gels were run at 150 V for 7 hours in 1 × TAE buffer (pH 8.0) at 60°C. DGGE gels were stained with ethidium bromide in 1x TAE for 20 min and then destained in 1 × TAE for 15 min. The gel photographs were viewed by the computer program Gel Doc. DNA bands on the DGGE gels were excised under UV trans-illumination using sterile scalpels and then soaked in 30 µl of sterile double-distilled water at 4°C overnight. 0.7 µl of DNA solution was used for re-amplification, using the primer pair described above without GC clamp. Re-amplification was done with the following thermal program: 95°C for 3 min, followed by 36 cycles of 94°C for 1 min, 40 sec at 50°C, 30 sec at 72°C and 8 min of a last extension at 72°C. DNA was then purified by using purification kits (Amersham Biosciences GFX™ PCR DNA and Gel Band Purification Kit). Purified DNA was sequenced, using the NS1 primer, in an automated sequencer (Applied Biosystem 3730 DNA Analyzer) at the Genome Research Centre, The University of Hong Kong.

Phylogenetic analysis

When DNA sequences were obtained from NS1 primer, a blast search was performed in GenBank to find possible sister groups (Table 1). Phylogenetic analyses were conducted in PAUP* 4.0b10 (Swofford, 2004) and multiple alignments were done in Bioedit (Hall, 1999) and Clustal X (Thompson *et al.*, 1997). Parsimony analyses included representatives of members from other orders (sequences available from GenBank) to resolve phylogenetic relationships and to root cladograms. Trees were inferred using the heuristic search option with 1000 random addition sequence additions. Gaps were treated as missing data and characters were unordered and weighted equally and differentially. Clade stability was assessed in a bootstrap analysis with 1000 replicates, random sequence additions with maxtrees set to 2000 and other default parameters as implemented in PAUP*. Further details are outlined in Jeewon *et al.* (2002, 2004).

Bayesian analyses were performed using the MrBayes3.0B4 program (Huelsenbeck and Ronquist, 2001). The Markov Chain-Monte Carlo (MCMC)

method was used to approximate the posterior probabilities of each branch, counting the occurrence of each branch in trees that were visited in the MCMC analysis progress.

Table 1. List of species used in the study and their GenBank accession numbers

Species	Accession number	Species	Accession number
<i>Aureobasidium pullulans</i>	AY137506	<i>Chaetomium globosum</i>	AB048285
<i>Botryosphaeria tsugae</i>	AF271127	<i>Clypeosphaeria uniseptata</i>	AY083812
<i>Cladosporium cladosporioides</i>	AF548071	<i>Daldinia</i> sp.	AY315425
<i>Mycosphaerella latebrosa</i>	AY251114	<i>Discostroma tosta</i>	AY083814
<i>Didymella cucurbitacearum</i>	AY293779	<i>Graphostroma platystoma</i>	AY083808
<i>Dothidea ribesia</i>	AY016343	<i>Haematon haematococca</i>	AY489697
<i>Elytroderma deformans</i>	AF203455	<i>Hypocrea rufa</i>	AY489694
<i>Lophodermium pinastri</i>	AF106014	<i>Hyponectria buxi</i>	AF130976
<i>Massariosphaeria phaeospora</i>	AF164368	<i>Hypoxylon fragiforme</i>	AB014046
<i>Mycocalicium albonigrum</i>	L37736	<i>Leuconectria clusiae</i>	AY489700
<i>Passalora fulva</i>	AY251109	<i>Monographella nivalis</i>	AF064049
<i>Phaeococcomyces nigricans</i>	AY843273	<i>Neocosmospora vasinfecta</i>	U44117
<i>Rhizoscyphus ericae</i>	AY524847	<i>Neurospora crassa</i>	AY046271
<i>Sarcinomyces petricola</i>	Y18702	<i>Seimatoantlerium</i> sp.	AF346555
<i>Septoria rosae</i>	AY251113	<i>Sordaria fimicola</i>	X69851
<i>Sphaerulina oryzina</i>	AY251103	<i>Thielavia terrestris</i>	U43969
<i>Trimmatostroma macowanii</i>	AY251118	<i>Viridispora diparietispora</i>	AY489703
<i>Tryblidiopsis pinastri</i>	AF106013	Outgroup	
<i>Westerdykella cylindrical</i>	AY016355	<i>Didymella cucurbitacearum</i>	AY293779
<i>Mycosphaerella</i> sp.	AY251116	<i>Pleospora</i> sp.	AY392129
Outgroup			
<i>Cosmospora coccinea</i>	AY489702		
<i>Leuconectria clusiae</i>	AY489700		

Results

Among the three leaves used for DNA extraction, two of them did not give any PCR product using the NS1 and FungGC primer pair. The DGGE profiles of the third leaf yielded 14 different bands from the different parts (Fig. 1). Three bands were from the lower parts, five bands from the upper parts of the leaf blades, and six from petioles and midribs. All fourteen bands were excised for sequencing analysis. Although some bands (eg. ML1 and MU1; ML2 and MR2) shared similar electrophoretic mobility the sequence analyses revealed that the fungal taxa were not identical or phylogenetically related.

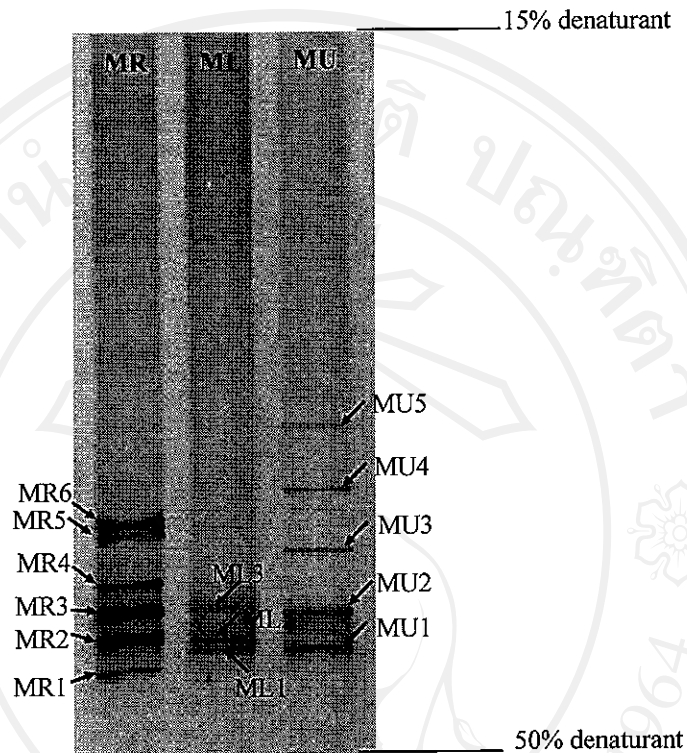


Fig. 1. Denaturant gradient gel electrophoresis profiles of 18S rDNA sequences amplified from DNA extracted directly from living leaf samples of *Magnolia liliifera*. Lane 1: Total genomic DNA extracted from leaf midrib and petiole (MR); Lane 2: Total genomic DNA extracted from upper parts of leaf blade (MU); Lane 3: Total genomic DNA extracted from lower parts of leaf blade (ML).

Table 2 shows the most similar taxa to the different OTUs following blast search results in GenBank. Blast search showed that ML1 and MR1 have high sequence similarity to *Trimmatostroma macowanii*; ML2 was similar to *Mycosphaerella* sp.; MU3 and MU4 were similar to *Botryosphaeria* species; MR2 and MR3 were similar to *Cordyceps* and uncultured *Hypocreales*; MR4 was similar to *Phaeococcomyces nigricans*; MR5 and MR6 were similar to *Hypoxylon fragiforme*; MU1 was similar to *Didymella cucurbitacearum*; MU2 was similar to Fungal sp. (AY843229); and MU3 was similar to *Elytroderma deformans*.

The Maximum Parsimony (MP) and Bayesian analysis of the unitunicate ascomycete dataset comprised 25 taxa with *Pleospora* sp. and *Didymella cucurbitacearum* as outgroups with 5 OTUs from DGGE (MR2, MR3, MR5, MR6 and MU5) resulted in one MP tree (Fig. 2). MR5 and MR6

belong to the *Xylariaceae* with 55% bootstrap and 0.91 Bayesian posterior probabilities. Phylogenetic results showed that MR2, MR3 and MU5 were hypocrealean taxa with of 92% bootstrap support and 1.00 posterior probability (Fig. 2). However, further phylogenetic placement of these OTUs could not be resolved.

Table 2. Percentage sequence similarities of uncultured phylotypes from *Magnolia liliifera* leaf colonizers to other sequences in GenBank.

OTU	Species	Similarity (%)	GenBank No	Family	Order
ML1	<i>Trimmatostroma macowanii</i>	299/300 (99)	AY251118	<i>Mycosphaerellaceae</i>	<i>Mycosphaerellales</i>
	<i>Coccodinium bartschii</i>	299/300 (99)	U77668	Icertae sedis	Icertae sedis
ML2	<i>Mycosphaerella</i> sp.	290/296 (97)	AY251116	<i>Mycosphaerellaceae</i>	<i>Mycosphaerellales</i>
	<i>Mycosphaerella latebrosa</i>	290/296 (97)	AY251114	<i>Mycosphaerellaceae</i>	<i>Mycosphaerellales</i>
ML3	<i>Botryosphaeria tsugae</i>	288/291 (98)	AF271127	<i>Botryosphaeriaceae</i>	<i>Dothideales</i>
	<i>Trimmatostroma macowanii</i>	285/288 (98)	AY251118	<i>Mycosphaerellaceae</i>	<i>Mycosphaerellales</i>
MU1	<i>Didymella cucurbitacearum</i>	313/316 (99)	AY293779	Icertae sedis	Icertae sedis
MU2	Fungal sp.	308/311 (99)	AY843229	Icertae sedis	Icertae sedis
	<i>Fusicoccum dimidiatum</i>	312/316 (98)	AF258605	<i>Botryosphaeriaceae</i>	<i>Dothideales</i>
MU3	<i>Elytroderma deformans</i>	294/295 (99)	AF203455	<i>Rhytismataceae</i>	<i>Rhytismatales</i>
MU4	<i>Botryosphaeria ribis</i>	304/309 (98)	AF271129	<i>Botryosphaeriaceae</i>	<i>Dothideales</i>
MU5	<i>Cordyceps sinensis</i>	294/295 (99)	AB187268	<i>Clavicipitaceae</i>	<i>Hypocreales</i>
MR1	<i>Trimmatostroma macowanii</i>	298/299 (99)	AY251118	<i>Mycosphaerellaceae</i>	<i>Mycosphaerellales</i>
MR2	<i>Cordyceps sinensis</i>	287/289 (99)	AB187268	<i>Clavicipitaceae</i>	<i>Hypocreales</i>
MR3	Uncultured	234/234	AY275188	Icertae sedis	Icertae sedis
	Hypocreales	(100)			
MR4	<i>Phaeococcomyces nigricans</i>	244/245 (99)	AY843273	<i>Herpotrichiellaceae</i>	<i>Chaetothyriales</i>
	<i>Hymenoscyphus ericae</i>	290/309 (93)	AY524847	<i>Helotiaceae</i>	<i>Helotiales</i>
MR5	<i>Hypoxylon fragiforme</i>	288/289 (99)	AY083810	<i>Xylariaceae</i>	<i>Xylariales</i>
MR6	<i>Hypoxylon fragiforme</i>	288/289 (99)	AY083810	<i>Xylariaceae</i>	<i>Xylariales</i>

Parsimony analysis of bitunicate ascomycetes showed that ML2 was related to *Passalora fulva* and clustered with taxa of the family *Mycosphaerellaceae* (*Mycosphaerella latebrosa*, *Mycosphaerella* sp., *Sphaerulina oryzina*, *Septoria rosae*) with 0.74 posterior probabilities (Fig. 3). ML1 and MR1 were sister taxa to *Cladosporium cladosporoides*

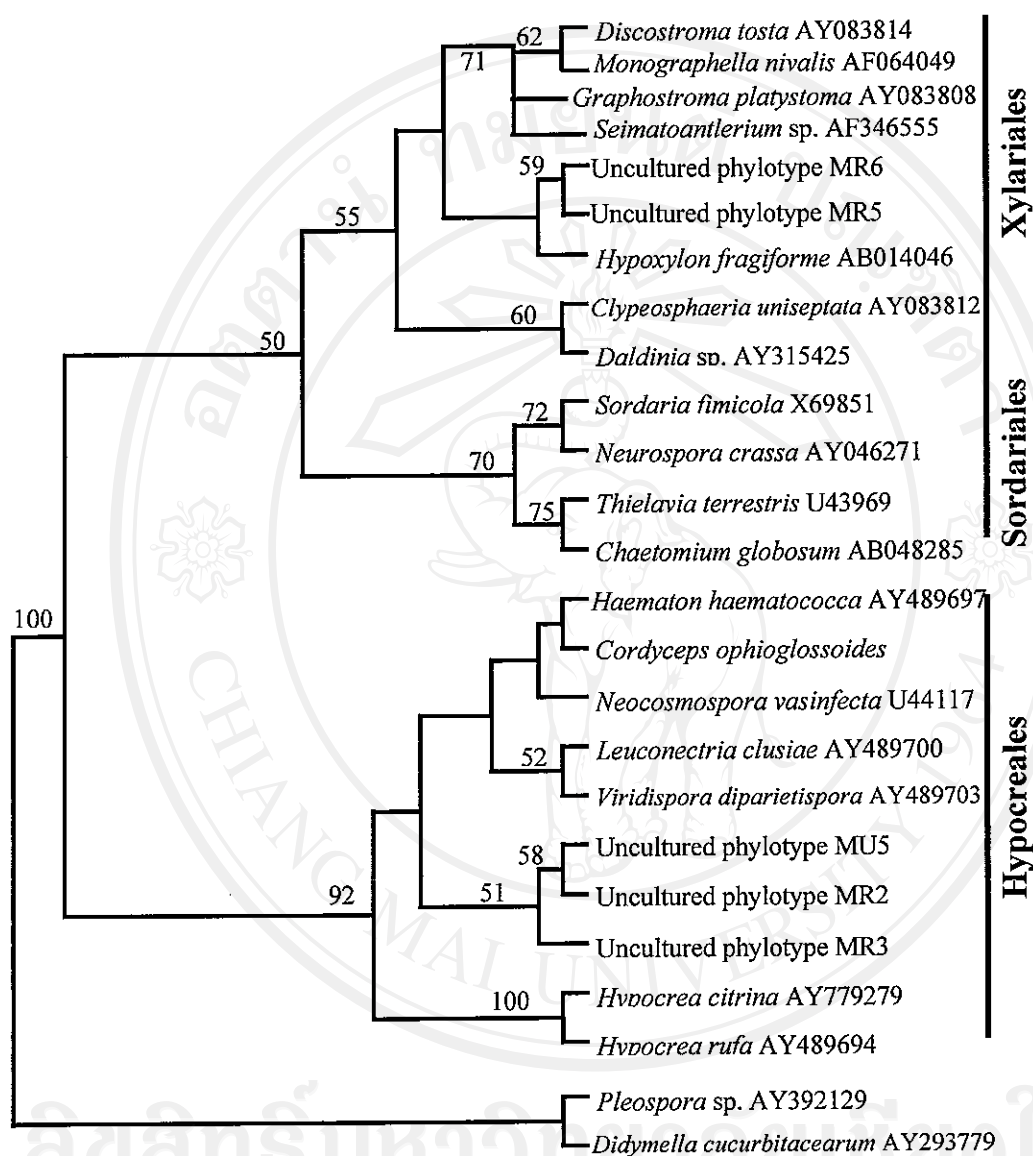


Fig. 2. Maximum-parsimony tree generated from partial 18S rDNA sequences of 25 taxa showing the relationships of MR2, MR3, MR5, MR6 and MU5 with reference taxa. The tree was rooted with *Pleospora* sp. and *Didymella cucurbitacearum* (Tree length = 125, CI = 0.688, RI = 0.851, RC = 0.586, and HI = 0.312). Bootstrap values $\geq 50\%$ (2000 replicates) are shown on the branches.

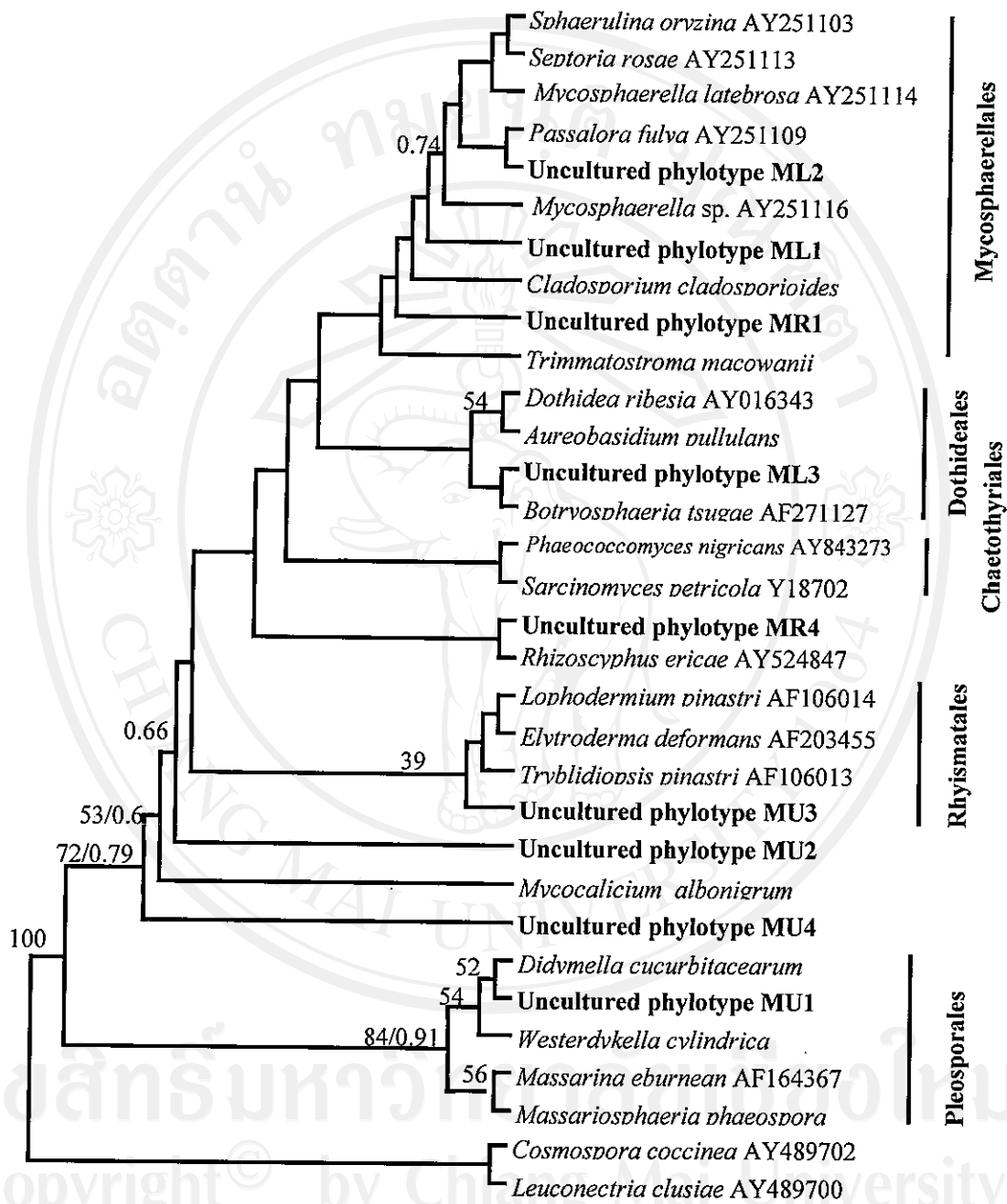


Fig. 3. Maximum-parsimony tree generated from partial 18S sequences of 32 taxa showing the relationships of ML1, ML2, ML3, MR1, MR4, MU1, MU2, MU3 and MU4 with reference taxa. The tree was rooted with *Cosmospora* and *Leuconectria clusiae* (tree length = 117, CI = 0.709, HI = 0.291, RI = 0.806, RC = 0.572). The numbers above branches were bootstrap and posterior probability values.

(*Mycosphaerellaceae*) and *Trimmatostroma macowanii*. ML3 was related to species of *Dothideales* (*Aureobasidium pullulans*, *Botryosphaeria tsugae* and *Dothidea ribesia*). MR4 clustered with *Rhizoscyphus ericae* (*Helotiaceae*, *Helotiales*). MU3 was related to the group comprising *Elytroderma deformans*, *Lophodermium pinastri*, and *Trybliidiopsis pinastri* (*Rhytismataceae*, *Rhytismatales*). However, these relationships did not have any support (Fig. 3). MU1 belongs to the family *Pleosporaceae* and is related to *Didymella cucurbitacearum*, *Massarina eburnea*, *Massariosphaeria phaeospora*, and *Westerdykella cylindrica* with relatively high support (84 bootstrap and 0.91 posterior probabilities) (Fig. 3). The phylogenetic placement of MU2 and MU4 could not be resolved using available DNA sequences.

Discussion

Previous studies on *Magnolia liliifera* have already shown that this host harbours numerous saprobic and endophytic fungal taxa (Promputtha *et al.*, 2004, 2005a,b). There, is however, one major limitation of these previous studies as they targeted fungi that either produce fruiting-bodies (which can be identified upon microscopic examination) or those fungi that can be easily cultured on artificial media. It has already been shown in several other studies that direct morphological examination of fruiting structures on substrates or culture-dependent methods give bias estimates of fungal communities (Duong *et al.*, 2004; Guo *et al.*, 2001; Promputtha *et al.*, 2004). This study targets fungal communities using DGGE with sequence analyses in an attempt to characterize unknown fungal taxa from *Magnolia liliifera*.

Studies on endophytes of Magnolia liliifera

We found fourteen operational taxonomic units (OTUs) on healthy leaf using PCR-DGGE coupled with sequence analyses. Phylogenetic analyses were useful to establish their taxonomic placement and systematic relationships with known fungi.

Fungal communities on *Magnolia liliifera* have been investigated by various methods. Promputtha *et al.* (2005a) identified 77 taxa of endophytic fungi from leaves of *Magnolia liliifera* of which 46 were sporulating and belonged to 8 genera (*Colletotrichum* (10), *Corynespora* (2), *Curvularia* (2), *Fusarium* (5), *Guignardia* (2), *Phomopsis* (11), *Trichoderma* (1), and *Xylaria* (13); 31 were morphospecies. In order to identify those morphospecies at a lower taxonomic level, rDNA gene sequence analyses were performed. The authors successfully identified the morphospecies into 6 ascomycete genera

(*Bionectria* - one morphospecies, *Diaporthe* - 24 morphospecies, *Glomerella* - one morphospecies, *Hypoxylon* - one morphospecies, *Massarina* - one morphospecies, and *Xylaria* - 3 morphospecies). Most of sporulating and non-sporulating fungi were common endophytic fungi that were also found in many other studies (Kumar *et al.*, 2004; Suryanarayanan *et al.*, 2005).

In the present study, 14 OTUs were successfully recovered from one leaf sample only using DGGE, revealing some typical endophytic genera (e.g. xylariaceous, and hypocrealean genera) as well as taxa that appear unable to grow on artificial media. Twelve OTUs were distributed amongst 6 different orders of ascomycetes (*Dothideales*, *Hypocreales*, *Mycosphaerellales*, *Pleosporales*, *Rhytismatales* and *Xylariales*) and 2 bitunicate fungi (MU2 and MU4), whose phylogenetic placement could not be resolved. This is not surprising as O'Brien *et al.* (2005) sequenced all kinds of organisms (soil and litter microorganisms from a mixed deciduous forest in the Southeastern United States) using a universal primer and identified them using the available data from GenBank. They found that 12% of the sequences could not be identified even to phylum level. Most endophytic isolates recovered using traditional methodologies and rDNA gene sequence analyses from *Magnolia liliifera* resulted in mostly unitunicate ascomycetes (Promputtha *et al.*, 2004, 2005a). Among the 77 taxa previously identified from leaves of *Magnolia liliifera*, only one species of *Massarina*, *Corynespora*, *Curvularia* (*Pleosporales*) and *Guignardia* (*Botryosphaeriales*) were isolated. In contrast, most OTUs identified using DGGE in this study are bitunicate ascomycetes. MU1 belongs to the *Pleosporales* and is sister taxon to *Didymella*, a genus without proper familial placement (Fig. 3). Surprisingly no endophytic or saprobic fungi had previously been isolated from *Magnolia liliifera* that were related or similar to the *Mycosphaerellales* and *Rhytismatales*. Therefore, the OTUs (ML1, ML2, MR1 and MU3) recovered from DGGE sequence analyses in this study indicate that these taxa are possibly unculturable or slow growing and have gone undetected in previous studies. In contrast, however, diaporthalean taxa and many others commonly isolated as endophytes (especially 24 *Phomopsis* species of 31 morphospecies from *Magnolia liliifera*) using artificial media were not identified using DGGE.

DGGE analysis coupled with phylogeny revealed that 5 phylotypes were unitunicate ascomycetes. Operational taxonomic units MR2, MR3 and MU5 belong to the order *Hypocreales*. In particular, they have high sequence similarity to those of *Cordyceps* species amongst many available small subunit sequences of other hypocrealean species in the GenBank (Table 2, Fig. 2). *Cordyceps* and its anamorphs are parasites of insects (Liang *et al.*, 2005) and their occurrence as endophytes is surprising. No *Cordyceps* species was

isolated from *Magnolia liliifera*. Promputtha *et al.* (2005a) found other hypocrealean genera isolated from the host such as *Trichoderma* and *Fusarium*. Other fungal diversity studies based on DGGE and sequence analyses have also reported taxa with phylogenetic affinities to the *Hypocreales* (Bougoure and Cainey, 2005, Gomes *et al.*, 2003). In our study, however, sequence analyses revealed that none of the OTUs recovered are *Trichoderma* or *Fusarium*.

OTUs MR5 and MR6 are closely related to *Hypoxylon* species. *Hypoxylon* comprises common endophytes and saprobes of various plant hosts (Petrini, 1991; Suryanarayanan *et al.*, 2005). An endophytic species of this genus was also isolated from *Magnolia liliifera* (Promputtha *et al.*, 2005a). *Xylaria* is another common endophyte that belongs to *Xylariaceae*, a family is known to harbor numerous endophytic species, and it was abundant in *Magnolia liliifera* (Promputtha *et al.*, 2005a). Our molecular approach used here failed to recover any *Xylaria* species.

This study demonstrated that DGGE could be used to detect known and abundant fungi (*Xylariales*, *Hypocreales* and *Pleosporales*) as well as unknown endophytic fungi (*Mycosphaerellales*, *Dothideales*, *Helotiales* and *Rhytismatales*). On the other hand, taxa such as *Diaporthe* and *Xylaria* that are abundant in cultural studies and other commonly found endophytic taxa were not recovered through DGGE. There may be several reasons for this. It has been suggested that some endophytes are not abundant in leaves, while others occupy spaces as small as single cells (Ghimire and Hyde, 2004; Varma *et al.*, 2004). In such cases there may not be enough fungal DNA to allow successful molecular detection. It might be plausible that these fungi present in a small amounts in the leaf tissues are not detected by DGGE, but being fast-growing fungi they are recovered abundantly on artificial media in traditional endophytic studies. The number of fungi identified by DGGE is small when compared to direct observation and traditional culturing of endophytes (Promputtha *et al.*, 2005a). However, when comparing to other studies such as Nikocheva *et al.* (2003, 2005), the number of fungal taxa revealed in this study is relatively large. The number of endophytic fungi isolated from parts of leaves is hard to estimate as parts, rather than the whole leaf is used. Fungal endophytes show various patterns within the leaves; some are confined to single plant cells, others occur internally or externally as single hyphae or they may grow throughout the veins (Varma *et al.*, 2004).

DGGE is a suitable method that can be applied in future studies to estimate fungal diversity, but it has several drawbacks. The primer pair NS1 and GCFung, as described by May *et al.* (2001) amplifies less than 400 nucleotides and it appears to be specific to ascomycetes. In addition, the region

amplified and sequenced (partial 18S rDNA gene) is rather conserved and therefore not appropriate to properly identify taxa at the genus or species level. Further studies should consider primers that are more universal (for fungi) and that give better phylogenetic resolution at generic or species level.

Fungi on different parts of leaves

Tissue specificity and recurrence in fungi have been studied at different host-taxonomic levels (Parungao *et al.*, 2002; Paulus *et al.*, 2006, Photita *et al.*, 2001, Polishook *et al.*, 1996; Varma *et al.*, 2004) as reviewed by Zhou and Hyde (2001). They concluded that many host-specific endophytic fungi become saprobes at leaf senescence. However, it is hard to say whether a given fungus is host-specific or host-recurrent.

In this study we applied DGGE to establish whether the method could detect differences in fungal communities present in different *Magnolia liliifera* leaf parts. We found a different fungal spectrum in different leaf parts. Different numbers of unitunicate and bitunicate ascomycetes were found in midrib and leaf blades. Two of six sequence types (MR1 and MR4) from the midrib were bitunicate ascomycetes and one of 8 sequence types (MU5) from leaf blades was a unitunicate ascomycete.

Fungi have been shown to be tissue-recurrent in several studies (Photita *et al.*, 2001, Promputtha *et al.*, 2004; Van Ryckegem and Verbeken 2005a,b; Wong *et al.*, 2001). Photita *et al.* (2001) studied fungal diversity on *Musa acuminata* in Hong Kong and found that distribution of fungal communities was different in leaf blades and petioles. Of 46 taxa from 2 study sites, only 11 taxa were present in both leaves and petioles. Six taxa were only identified from the petioles. Promputtha *et al.* (2004) documented the saprobic fungal communities on dead leaves of *Magnolia liliifera* with 5 taxa on midribs and petioles, and 33 taxa on leaf blades. The only one species found on both leaf blades and petiole was *Sporidesmium crassisporum*. The OTUs identified in this study illustrate differences in fungal communities between tissue types but given the sparse leaf samples studied, we refrain from making conclusive statements as to whether these taxa are tissue recurrent.

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