CHAPTER 4

SAPROBES ON ZINGIBERACEAE

4.1 Introduction

Recent intensive studies of microfungi on plants in the tropics have included those on bamboo (Hyde *et al*., 2002a, b), grasses (Wong and Hyde, 2001), palms (Fröhlich and Hyde, 1999; Hyde and Alias, 2000), sedges (Wong and Hyde, 2001), Pandanaceae (McKenzie *et al*., 2002) and Musaceae (Photita *et al*., 2001a, 2003b) and these provide important data for the "fungal estimates" debate (Hawksworth, 1991, 2001). Although several taxonomic studies have been carried out of fungi on Zingiberaceae (Rao, 1962; Muthappa, 1966; Doi, 1977; Khurana, 1980; Rathaiah, 1980, 1981; Vittal, 1981; Pavgi and Upadhyay, 1986; Samuels, 1989; Hyde, 1997; Bussaban *et al*., 2001a, c, 2003a, b) including endophytic fungi (Bussaban *et al*., 2001b, d), there have been no previous detailed studies of sabrobic fungi on the family. Bussaban *et al*. (2002) provided an annotated checklist of all fungi described from the Zingiberaceae. The present study was initiated in order to study saprobic fungi in detail by examining and comparing the fungi on leaf and pseudostem tissues of four wild and two cultivated species of Zingiberaceae to establish whether the saprobes differ between zingiberaceous hosts, especially between the wild and cultivated species, and whether the fungi in gingers differ from those in other tropical hosts.

4.2 Materials and methods

4.2.1 Sample collection

Dead leaves and pseudostems were collected from the same host in the same sites as that of the endophytic fungi study (Chapter 3). The collection details are listed in Table 4.1. At each collection of *Alpinia malaccensis*, *A. siamense* and *Etlinger littoralis*, ten leaves and ten pseudostems were randomly collected from dead plant material lying on the forest ground, while a further ten leaves and ten pseudostems were collected from standing dead plants (Figure 4.1). For *A. galanga*, *E. elatior* and *Zingiber officinale* ten leaves and ten pseudostems were collected only from the standing dead plants. All samples (20 cm long) were placed in separate snap-locked plastic bags and returned to the laboratory.

Table 4.1 Collection details of saprobic fungi study.

Host	Site	Collection date
Wild species		
Alpinia malaccensis	Huay Kok Ma	15 October 2000
	Doi Pui	15 October 2000
Amomum siamense	Huay Kok Ma	20 May 2000
		15 October 2000
	Medicinal Plant Garden	20 May 2000
		15 October 2000
Etlingera elatior	Queen Sirikit Botanic Garden	21 September 2003
	Chiang Mai University area	28 September 2003
Etlingera littoralis	Huay Kok Ma	12 October 2003
	Medicinal Plant Garden	12 October 2003
Cultivated species		
Alpinia galanga	Chiang Mai Province (Hangdong)	19 October 2002
	Lampang province (Muang)	26 October 2002
Zingiber officinale	Phayao (commercial)	3 September 2001
	Phayao (backyard)	3 September 2001

Figure 4.1 Sampling leaves and pseudostems from fallen (left) and standing (right) dead plants.

4.2.2 Examination of samples

Moist tissue paper was added to the plastic bags to create humid conditions. The samples were then incubated at room temperature and examined for fungi, within 2 weeks, using a dissecting microscope. The fungi present on the samples were identified to genus or species using standard literature (e.g., Ellis, 1971, 1976; Carmichael *et al*., 1980; Sutton, 1980; von Arx, 1981; Hyde *et al*., 2000). Saprobic fungi were isolated by single spore methods and grown on PDA (Choi *et al.,* 1999). Cultures were maintained in the culture collection at Microbial Diversity Laboratory, Faculty of Science, Chiang Mai University and deposited in BIOTEC Culture Collection (BCC), International Collection of Microorganisms from Plants (ICMP) or The University of Hong Kong Culture Collection (HKUCC).

For herbarium specimens, a piece of the substrate containing fungal fruiting structures was cut and placed in an envelope or small cardboard box. Collecting information (e.g., collection date, locality, substrate, collector) was written on each herbarium packet. The herbarium specimens were then dried in a hot air oven (35–40 °C) for 2–3 days and stored at Microbial Diversity Laboratory, Faculty of Science, Chiang Mai University, or deposited in Landcare Research herbarium (PDD).

4.2.3 Statistical analysis

A fungal species list with frequency of occurrence was established for each plant species. The percentage occurrences of fungi were calculated according to the following formula:

$$\text{Percentage occurrence} = \frac{\text{number of samples on which fungus was observed}}{\text{total number of samples examined}} \times 100$$

A data matrix consisting of the numbers of colonised tissue pieces from each sample unit (i.e., leaf or pseudostem, hosts and locality) in rows, and fungi in columns was subjected to ordination using correspondence analysis. The data from different hosts was analysed separately to determine whether the fungal community recovered from different tissues and localities varied. Sample unit and fungal species ordinations were obtained simultaneously, therefore the fungal compositions of different zingiberaceous species at different sites were also compared using 3-dimensional correspondence analysis. Similar communities are positioned closely together in the same ordination space (Anonymous, 1995). Sorenson index was also used to measure similarity between species diversity on different hosts.

Similarity index = 2c/a+b

a: the number of species in host 1
b: the number of species in host 2
c: the number of species in common in both hosts

Species area curves were used to determine the adequacy of the sampling size, Shannon index (H') was used to express species diversity of a community (Shannon and Weaver, 1949).

Shannon index (H') = $-\sum p_i \ln p_i$

p_i: the proportion of occurrence of taxon i^{th} and occurrence of all taxa

4.3 Results

4.3.1 Determination of adequacy of sample size

Species area curves for each collection of zingiberaceous plants almost reached asymptote because the slopes of the curves were declining with the increase of sample size. At about 10 samples, the slopes were close to zero (Figure 4.2). Although the curve did not completely level off, the number of samples was large enough to obtain a highly representative result.

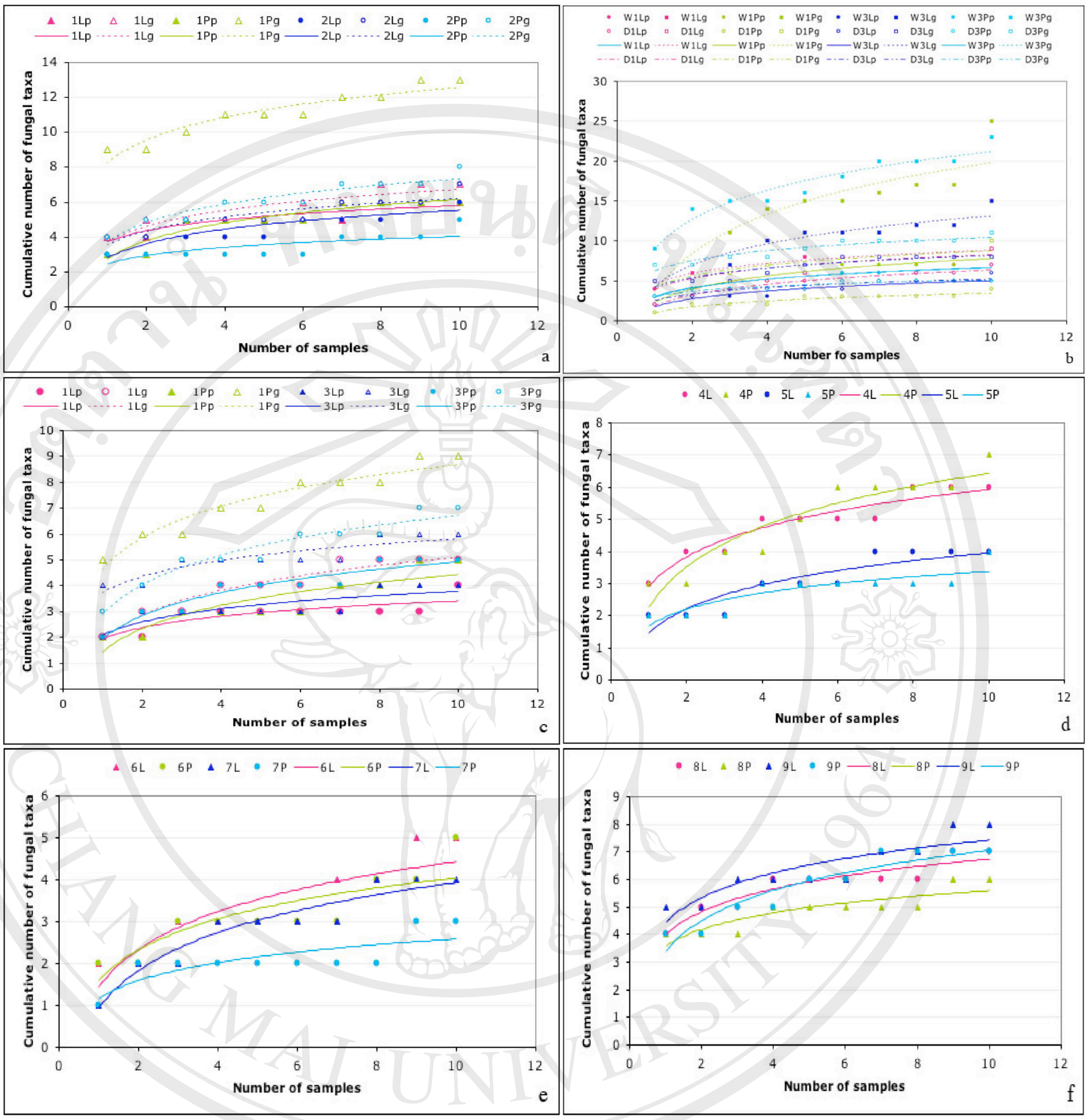

Figure 4.2 Species area curves for saprobic fungi collected from Zingiberaceae: a. *Alpinia malaccensis*, b. *Amomum siamense*, c. *Etlingera littoralis*, d. *E. elatior*, e. *Alpinia galanga*, f. *Zingiber officinale*. W. wet season, D. dry season, 1. Huay Kok Ma, 2. Doi Pui, 3. Medicinal Plant Garden, 4. Queen Sirikit Botanic Garden, 5. Chiang Mai University area, 6. Chiang Mai Province (Hangdong), 7. Lampang Province (Muang), 8. Phayao Province (commercial), 9. Phayao Province (backyard), L. leaf, P. pseudostem, g. collection from forest ground, p. collection from standing dead plants.

4.3.2 Fungal taxonomic composition

Of the 2,485 fungal collections, 163 fungal species were identified comprising 20 basidiomycetes, 26 ascomycetes and 117 anamorphic fungi (12 coelomycetes and 105 hyphomycetes) (Table 4.2). The fungi occurring on leaf and pseudostem of the four wild and two cultivated species in the different sampling sites are listed in Tables 4.3–4.8. *Acremonium* sp. 1 was the most common fungus found on the wild zingiberaceous species, *Alpinia malaccensis*, *Amomum siamense*, *Etlingera elatior* and *E. littoralis*, being found on 31%, 41%, 37% and 27% of samples, respectively. In the cultivated species, *Phomopsis* sp. (35%) was the most common species on *Alpinia galanga*, while *Phaeosphaeria* sp. (80%) was the most commonly encountered species from *Zingiber officinale*.

In each wild species two–seven fungi were recorded with percentage occurrence higher than 19% (Tables 4.3–4.6). Fungi with over 19% percentage occurrence on samples of the wild species included *Acremonium* sp. 1, *Chloridium* sp. and *Verticillium* sp. 3 on *A. malaccensis*; *Acremonium* sp. 1, *Canalisporium caribense*, *Chloridium* sp., *Dactylaria hemibeltranioidea*, *Dactylaria* sp. 2, *Dactylella ellipsospora* and *Periconia digitata* on *A. siamense*; *Acremonium* sp. 1, *Cladosporium cladosporioides*, *Dactylaria hemibeltranioidea*, *Dictyoarthrinium sacchari* and *P. digitata* on *E. elatior* and *Acremonium* sp. 1 and *Chloridium* sp. on *E. littoralis*. Four fungi (*C. cladosporioides*, *Colletotrichum gloeosporioides*, *Curvularia lunata* and *Phomopsis* sp.) with a percentage occurrence higher than 19% were recorded from *A. galanga* while 11 fungi (Ascomycete 3, *Aspergillus niger*, *Cladosporium cladosporioides*, *C. oxysporum*, *Curvularia lunata*, *Fusarium oxysporum*, Hyphomycete 7, *Paecilomyces* sp., *Phaeosphaeria* sp., *Phomopsis* sp. and

Phyllosticta capitalensis) with percentage occurrence higher than 19% were recorded from *Z. officinale* (Tables 4.7, 4.8).

The different zingiberaceous species, supported different assemblages and numbers of fungal taxa. In term of the numbers of taxa recovered fungi were more diverse in wild zingiberaceous species than in cultivated species (Table 4.2) and this is also indicated by the significantly higher Shannon indices (Figure 4.3).

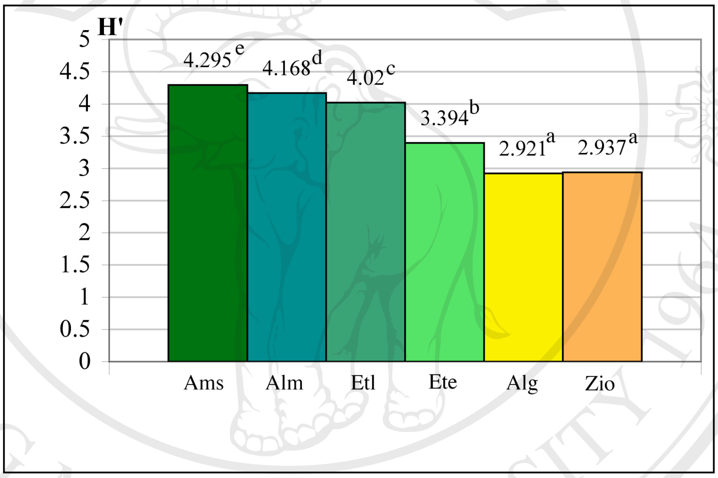

Figure 4.3 Shannon indices (H´) comparing the species diversity of zingiberaceous plants: Ams. *Amomum siamense*, Alm. *Alpinia malaccensis*, Etl. *Etlingera littoralis*, Ete. *Etlingera elatior*, Alg. *Alpinia galanga*, Zio. *Zingiber officinale*. [a–e] values above the bars with no common superscripts are significantly different ($P < 0.05$).

Table 4.2 A comparison of the total fungal taxa recovered from zingiberaceous species.

Taxa	***Alpinia malaccensis***	***Amomum siamense***	***Etlingera littoralis***	***Etlingera elatior***	***Alpinia galanga***	***Zingiber offinale***
Acremonium sp. 1	+	+	+	+		
Acremonium sp. 2	+	+	+	+		
Acremonium sp. 3		+				
Acrodictys sp.	+	+				
Agaricus sp.			+			
Alternaria alternata					+	
Anthostomella sp.	+		+			
Anthracophyllum nigritum			+			
Ascomycete 1	+	+				
Ascomycete 2	+	+	+	+		
Ascomycete 3						+
Aspergillus niger					+	+
Bactrodesmella sp.		+				
Bahusandhika sundarum	+	+				
Basidiomycete 1	+		+			
Basidiomycete 2			+			
Basidiomycete 3	+		+			
Basidiomycete 4			+			
Basidiomycete 5		+				
Basidiomycete 6			+			
Berkleasmium nigroapicale#	+	+				
Berkleasmium sp.*		+				
Berkleasmium sutheppuiense#	+	+				
Bipolaris sp.						+
Botryodiplodia sp.	+	+	+	+		
Brachysporiella sp.	+	+	+	+		
Canalisporium caribense	+	+	+			
Canalisporium exiguum	+	+	+			
Cephalosporiopsis sp.	+		+	+		
Cercophora sp.		+	+			
Cercospora amomi		+		+	+	

Table 4.2 (Continued).

Taxa	***Alpinia malaccensis***	***Amomum siamense***	***Etlingera littoralis***	***Etlingera elatior***	***Alpinia galanga***	***Zingiber offinale***
Cercospora sp.			+			
Cercospora zingibericola						+
Chaetomium sp.		+				
Chalara sp.	+	+	+	+		
Chloridium botryoideum	+	+	+			
Chloridium sp.	+	+	+	+		
Cladorrhinum foecundissinum						+
Cladosporium cladosporioides	+	+	+	+	+	+
Cladosporium oxysporum						+
Cladosporium uredinicola	+	+		+		
Clavaria sp.			+			
Coelomycete 1		+				
Coelomycete 2		+				
Colletotrichum gloeosporioides	+	+	+	+	+	+
Colletotrichum sp.		+			+	
Cordana sp. 1	+	+				
Cordana sp. 2		+				
Corpinus disseminatus			+			
Crepidotus sp. 1			+			
Crepidotus sp. 2		+	+			
Curvularia lunata				+	+	+
Cyathus sp.			+			
Cylindrocladium sp.			+			
Dactylaria hemibeltranioidea	+	+	+	+		+
Dactylaria intermedia	+	+			+	
Dactylaria sp. 1*		+	+	+		
Dactylaria sp. 2*	+	+	+	+		
Dactylella ellipsospora	+	+	+	+		
Dactylella sp.	+	+	+	+		
Dictyoarthrinium sacchari	+	+	+	+		
Dictyosporium heptasporum	+	+	+			
Dictyosporium sp. 1*		+				

Table 4.2 (Continued).

Taxa	***Alpinia malaccensis***	***Amomum siamense***	***Etlingera littoralis***	***Etlingera elatior***	***Alpinia galanga***	***Zingiber offinale***
Dictyosporium sp. 2*		+				
Didymella sp.		+				
Discomycete	+	+	+			
Endophragmia cesatii	+					
Fusarium sp. 1	+	+	+	+		
Fusarium sp. 2	+	+		+	+	
Fusarium oxysporum					+	+
Gaeumannomyces amomi#		+				
Gelasinospora sp.						+
Gliocladium sp.						+
Gliomastix sp.		+	+	+	+	
Glomerella sp.						+
Graphium sp.	+	+				
Haematonectria sp.		+				
Helicodendron sp.		+				
Helicomyces bellus	+	+	+	+		
Helicomyces sp.	+	+	+	+		
Helicosporium sp.	+	+	+	+		
Hermatomyces sp.		+				
Hormomyces sp.	+	+	+			
Hyphomycete 1		+				
Hyphomycete 2	+	+				
Hyphomycete 3	+	+				
Hyphomycete 4		+				
Hyphomycete 5	+	+	+			
Hyphomycete 6		+				
Hyphomycete 7						+
Hypocrea rufa		+	+			
Hypocrea sp.	+	+	+			
Hyponectria sp.	+	+	+			
Japhneadelphus sp.			+			
Leptosphaeria alpiniae					+	

Table 4.2 (Continued).

Taxa	***Alpinia malaccensis***	***Amomum siamense***	***Etlingera littoralis***	***Etlingera elatior***	***Alpinia galanga***	***Zingiber offinale***
Leptosphaeria sp.		+				
Marasmiellus inoderma			+			
Marasmiellus sp.		+				
Marasmius sp. 1		+	+			
Marasmius sp. 2			+			
Marasmius sp. 3			+			
Marasmius sp. 4	+					
Memnoniella sp.		+			+	
Memnoniella subsimplex	+	+		+	+	
Microsphaeropsis sp.	+	+	+			
Monodictys sp.	+	+	+			
Mycena sp.		+				
Myrothecium cinctum		+	+			+
Myrothecium inundatum						+
Nakataea fusispora			+			
Nectria sp. 1			+			
Nectria sp. 2	+		+			
Nectriopsis sp.	+	+				
Nigrospora oryzae	+	+		+	+	+
Orbilia sp.	+	+	+			
Paecilomyces sp.	+	+	+	+		+
Penicillium sp.	+	+	+	+		
Periconia digitata	+	+	+	+		
Periconia minutissima		+	+	+	+	
Periconia sp. 1	+	+				
Periconia sp. 2	+	+				
Pestalotiopsis sp.	+		+	+		
Phaeosphaeria sp.						+
Phialocephala humicola	+	+	+			
Phialophora sp.	+					
Phoma sp.	+	+	+	+	+	
Phomopsis sp.	+	+	+		+	+

Table 4.2 (Continued).

Taxa	***Alpinia malaccensis***	***Amomum siamense***	***Etlingera littoralis***	***Etlingera elatior***	***Alpinia galanga***	***Zingiber offinale***
Phyllosticta capitalensis	+	+	+		+	+
Pithomyces chatarum					+	
Pleospora sp.		+				
Pyricularia costina	+	+	+			
Pyricularia longispora[#]	+	+				
Pyricularia sp.*		+	+			
Pyriculariopsis sp.	+	+				
Ramichloridium musae	+	+	+			
Septonema sp.	+	+				
Septoria sp.				+	+	
Solosympodiella sp.	+	+				
Sporidesmium leonense	+	+				
Sporidesmium sp.	+	+				
Stachybotrys echinata	+	+	+	+	+	
Stachybotrys sp.	+	+	+	+		+
Stachylidium sp. 1	+					
Stachylidium sp. 2	+	+	+	+		
Staurophoma sp.		+				
Stilbella sp.		+				
Stilbohypoxylon sp.	+	+				
Subulispora sp.	+	+	+			
Synematous sp.	+	+				
Torula sp. 1	+	+				
Torula sp. 2	+	+				
Trichoderma sp.	+	+	+	+	+	+
Vermiculariopsiella sp.						+
Veronaea musae	+	+				
Verticillium sp. 1	+	+	+	+	+	
Verticillium sp. 2	+	+				
Verticillium sp. 3	+	+				
Verticimonosporium sp.		+				
Xenosporium amomi[#]	+	+				

Table 4.2 (Continued).

Taxa	***Alpinia malaccensis***	***Amomum siamense***	***Etlingera littoralis***	***Etlingera elatior***	***Alpinia galanga***	***Zingiber offinale***
Xenosporium intermedium	+	+	+			
Xenosporium sp.*		+				
Xenosporium thaxeri	+	+	+			
Xylaria sp.		+				
Basidiomycetes (20)	3	5	16			
Ascomycetes (26)	10	17	11	1	1	4
Anamorphic fungi (117)	75	95	52	38	23	21
Total taxa (163)	**88**	**117**	**79**	**39**	**24**	**25**
Total collections (2485)	**461**	**1164**	**356**	**159**	**115**	**230**

*Probably undescribed species
#Described during the present study

Table 4.3 Overall percentage occurrence and frequency of fungal taxa recovered from *Amomum siamense*.

Taxa	Wet season								Dry season								Overall percentage occurrence
	Huay Kok Ma				Medicinal Plant Garden				Huay Kok Ma				Medicinal Plant Garden				
	Lp	Lg	Pp	Pg	Lp	Lg	Pp	Pg	Lp	Lg	Pp	Pg	Lp	Lg	Pp	Pg	
Acremonium sp. 1	2	4	2	8	6	4	3	7	2	3	2	5	5	4	4	5	41.3
Acremonium sp. 2		3				2	1	1		3		1		1	1	1	8.8
Acremonium sp. 3								2									1.3
Acrodictys sp.				2			1	1				1			1	2	5.0
Ascomycete 1				1			1					2				1	3.1
Ascomycete 2			1	2				1			1					1	3.8
Bactrodesmella sp.								1									0.6
Bahusandhika sundarum				2													1.3
Basidiomycete 5				1													0.6
Berkleasmium nigroapicale		1		1				3				1				1	4.4
Berkleasmium sp.				1													0.6
Berkleasmium sutheppuiense				2				1									1.9
Botryodiplodia sp.	2	2			3	7			1	2			1	2			12.5
Brachysporiella sp.			1	1				2				1					3.1
Canalisporium caribense	1	1	2	3	1	3	3	9		1	2	2	1	3	4	7	26.9
Canalisporium exiguum			1	4			1	2				2			1	2	8.1
Cercophora sp.				4				2				1				1	5.0
Cercospora amomi		1		2			1									1	3.1
Chaetomium sp.				1		1	2							1			3.1
Chalara sp.			2	2		1		1				2				1	5.6
Chloridium botryoideum		1		4		2		2				2		1		2	8.8
Chloridium sp.		2	4	8		4	1	4	2	2	3	4	3	5	1	2	28.1
Cladosporium cladosporioides	3	2		1	1	2		2	4	2			3	2	1	1	15.0
Cladosporium uredinicola		2				1	2		1	2			1	2	1		7.5
Coelomycete 1				2		1		1									2.5
Coelomycete 2						1											0.6
Colletotrichum gloeosporioides			2	1			1	3		2	1	1	1		1	1	8.8
Colletotrichum sp.								1	1	1							1.9
Cordana sp. 1		1				1		3		1				1		1	5.0
Cordana sp. 2	1				1		1			1							2.5

Table 4.3 (Continued).

Taxa	Wet season								Dry season								Overall percentage occurrence
	Huay Kok Ma				Medicinal Plant Garden				Huay Kok Ma				Medicinal Plant Garden				
	Lp	Lg	Pp	Pg	Lp	Lg	Pp	Pg	Lp	Lg	Pp	Pg	Lp	Lg	Pp	Pg	
Crepidotus sp. 2			1														0.6
Dactylaria hemibeltranioidea	3	1		2	4	1	2	6	3	2		1	3	1	2	2	20.6
Dactylaria intermedia						1		2					2	1		2	5.0
Dactylaria sp. 1						1		2						1			2.5
Dactylaria sp. 2		4	1	3	1	3	2	1	2	4	1	1	4	3	2	2	21.3
Dactylella ellipsospora	2	2	1	5	1	1	2	4		2		5		2	1	4	20.0
Dactylella sp.					2	3		3				2		4		3	10.6
Dictyoarthrinium sacchari							3	8							1	6	11.3
Dictyosporium heptasporum				1			1	2			1	3			2	1	6.9
Dictyosporium sp. 1				2													1.3
Dictyosporium sp. 2				1													0.6
Didymella sp.							1										0.6
Discomycete	1								1								1.3
Fusarium sp. 1				1				1		1		1			1	1	3.8
Fusarium sp. 2		2	1							1	1						3.1
Gaeumannomyces amomi				1													0.6
Gliomastix sp.				2													1.3
Graphium sp.			2	3		1		2								3	6.9
Haematonectria sp.							1	3								1	3.1
Helicodendron sp.		1				3			3	1			2	3			8.1
Helicomyces bellus			1			1		3			1					1	4.4
Helicomyces sp.						1	1	5				1		1		1	6.3
Helicosporium sp.	1		2	3						1	1	2					6.3
Hermatomyces sp.						1											0.6
Hormomyces sp.		1	1	2		3				1		2				1	6.9
Hyphomycete 1		2															1.3
Hyphomycete 2						1											0.6
Hyphomycete 3								1						2	1	2	3.8
Hyphomycete 4			1				1										1.3
Hyphomycete 5		1	1	5													4.4
Hyphomycete 6			1	2						2		2					4.4

Table 4.3 (Continued).

Taxa	Wet season								Dry season								Overall percentage occurrence
	Huay Kok Ma				Medicinal Plant Garden				Huay Kok Ma				Medicinal Plant Garden				
	Lp	Lg	Pp	Pg	Lp	Lg	Pp	Pg	Lp	Lg	Pp	Pg	Lp	Lg	Pp	Pg	
Hypocrea rufa				1													0.6
Hypocrea sp.			3				2				1	1				1	5.0
Hyponectria sp.			1	4													3.1
Leptosphaeria sp.							1										0.6
Marasmiellus sp.				2													1.3
Marasmius sp. 1				1													0.6
Memnoniella sp.	2	2				2			3								5.6
Memnoniella subsimplex	1	1		3	1	2			3	3		1	2	3			12.5
Microsphaeropsis sp.		1		1	1	5	2	5		1			1	2		2	13.1
Monodictys sp.	1	2	3	4	1	1	1	1		2		1		1	1	1	12.5
Mycena sp.	2								1								1.9
Myrothecium cinctum							1	2									1.9
Nectriopsis sp.								2									1.3
Nigrospora oryzae				1					2	2		1					3.8
Orbilia sp.	1		1														1.3
Paecilomyces sp.	1	1	1	3	2	5	1	2	1	1		1		1	1	1	13.8
Penicillium sp.						1		2		1			1	1	2	2	6.3
Periconia digitata	5	1	4	3	2	4	1	5	1			1		2	2	1	20.0
Periconia minutissima						1		3			1	1		1			4.4
Periconia sp. 1				1	1	3	1	2				1		1	1	1	7.5
Periconia sp. 2	2	2			1	5				1			2				8.1
Phialocephala humicola								1									0.6
Phoma sp.			2			2	1	4								1	6.3
Phomopsis sp.	2	1			1	5				1			2	1			8.1
Phyllosticta capitalensis								2									1.3
Pleospora sp.				1													0.6
Pyricularia costina	3	1			2	2			2	1			3	2			10.0
Pyricularia longispora	2	4		7		1		2	3	3		3	2	1		1	18.1
Pyricularia sp.				2				1									1.9
Pyriculariopsis sp.					1	1		2									2.5
Ramichloridium musae						1		2								1	2.5

Table 4.3 (Continued).

Taxa	Wet season								Dry season								Overall percentage occurrence
	Huay Kok Ma				Medicinal Plant Garden				Huay Kok Ma				Medicinal Plant Garden				
	Lp	**Lg**	**Pp**	**Pg**	**Lp**	**Lg**	**Pp**	**Pg**	**Lp**	**Lg**	**Pp**	**Pg**	**Lp**	**Lg**	**Pp**	**Pg**	
Septonema sp.	3	3		1	2	1		2	2	4		1	3	1		1	15.0
Solosympodiella sp.			3	5			1	3				2			1		9.4
Sporidesmium leonense					1		1	1									1.9
Sporidesmium sp.							1	1								1	1.9
Stachybotrys echinata	5	2	1	3	2	2		1	1	4	1	1	1	1	1		16.3
Stachybotrys sp.		1		2	1	1	1	1			2	1		1	1	2	8.8
Stachylidium sp. 2	5	4	1	3	1	2			1	1	1	2	1	1		1	15.0
Staurophoma sp.		2					1										1.9
Stilbella sp.								1									0.6
Stilbohypoxylon sp.				2				4				1					4.4
Subulispora sp.								1									0.6
Synematous sp.			2	3													3.1
Torula sp. 1			2					2			2	1					4.4
Torula sp. 2			1	1				1									1.9
Trichoderma sp.				3			2	1				1		2		1	6.3
Veronaea musae		1	1			1		3									3.8
Verticillium sp. 1	2	4		2		2	1	1	3	3	1	2	1	2	1	2	16.9
Verticillium sp. 2	1	4	1	1		2	1	1	1	1		1		1		1	10.0
Verticillium sp. 3	2	3	1	1		2		4	4	3	3			2	2	2	18.1
Verticimonosporium sp.			1														0.6
Xenosporium amomi			1				1	3						2	2	4	8.1
Xenosporium intermedium			1				2	4		1		2			2	1	8.1
Xenosporium sp.			2														1.3
Xenosporium thaxeri								2							1	2	3.1
Xylaria sp.								1									0.6
Basidiomycetes (5)	1		1	3					1								
Ascomycetes (17)	2		4	9		1	6	6	1		2	4		1		5	
Anamorphic fungi (95)	23	38	34	49	24	49	34	64	22	37	16	39	22	38	29	46	
Total taxa (117)	**26**	**38**	**39**	**61**	**24**	**50**	**40**	**70**	**24**	**37**	**18**	**43**	**22**	**39**	**29**	**51**	

L = leaf, P = pseudostem, p = samples collected from standing dead plant, g = samples collected from dead plant laying on the forest ground

Table 4.4 Overall percentage occurrence and frequency of fungal taxa recovered from *Alpinia malaccensis*.

Taxa	Huay Kok Ma				Doi Pui				Overall percentage occurrence
	Lp	**Lg**	**Pp**	**Pg**	**Lp**	**Lg**	**Pp**	**Pg**	
Acremonium sp. 1	1	2	4	4	2	5	3	4	31.25
Acremonium sp. 2		1		2		1	1		6.25
Acrodictys sp.			1	1		1			3.75
Anthostomella sp.			1	3					5.00
Ascomycete 1				1					1.25
Ascomycete 2			1	1			1	2	6.25
Bahusandhika sundarum				1					1.25
Basidiomycete 1	1								1.25
Basidiomycete 3					2				2.50
Berkleasmium nigroapicale				1					1.25
Berkleasmium sutheppuiense				1					1.25
Botryodiplodia sp.	2	1			1	3			8.75
Brachysporiella sp.		1	2	5		1		1	12.50
Canalisporium caribense		1	1	4	1	2	2	3	17.50
Canalisporium exiguum				1					1.25
Cephalosporiopsis sp.			1	3					5.00
Chalara sp.			2	1				1	5.00
Chloridium botryoideum		1		1		1	1	2	7.50
Chloridium sp.		4	1	5	1	3	2	2	22.50
Cladosporium cladosporioides	4	1	1		2	2			12.50
Cladosporium uredinicola	1	1	1						3.75
Colletotrichum gloeosporioides		2		1			1	1	6.25
Cordana sp. 1	1	2		1					5.00
Dactylaria hemibeltranioidea		2		2	4	4			15.00
Dactylaria intermedia		1		1	1				3.75
Dactylaria sp. 2	2	1		1					5.00
Dactylella ellipsospora	1	2		2		1	1	2	11.25
Dactylella sp.				1	1	1	1	2	7.50
Dictyoarthrinium sacchari		1	2	3					7.50
Dictyosporium heptasporum		1	1	2	1	1		1	8.75
Discomycete	2	1							3.75
Endophragmia cesatii				1					1.25
Fusarium sp. 1				2			1	1	5.00
Fusarium sp. 2		1	1	1					3.75
Graphium sp.		1	1	2	2	1		1	10.00
Helicomyces bellus	1			1					2.50
Helicomyces sp.		1	1	2	2		1	1	10.00
Helicosporium sp.		1	1	1		1	1	2	8.75
Hormomyces sp.				1					1.25
Hyphomycete 2				1					1.25
Hyphomycete 3		1		1				1	3.75
Hyphomycete 5			1	1					2.50
Hypocrea sp.			1				1	1	3.75
Hyponectria sp.				1					1.25
Marasmius sp. 4		1							1.25
Memnoniella subsimplex		1	1	1	2	1	1	2	11.25
Microsphaeropsis sp.		1		1		1	1	1	6.25

Table 4.4 (Continued).

Taxa	Huay Kok Ma				Doi Pui				Overall percentage occurrence
	Lp	**Lg**	**Pp**	**Pg**	**Lp**	**Lg**	**Pp**	**Pg**	
Monodictys sp.		2	1	3	1	1			10.00
Nectria sp. 2			1	4			1	2	10.00
Nectriopsis sp.				1					1.25
Nigrospora oryzae	1		1	2				3	8.75
Orbilia sp.				1					1.25
Paecilomyces sp.			1	2	3	1	1	2	12.50
Penicillium sp.		1				1		1	3.75
Periconia digitata	3	1	2	2	1	2	1	2	17.50
Periconia sp. 1	1		2	1		1		1	7.50
Periconia sp. 2	1	1							2.50
Pestalotiopsis sp.	2								2.50
Phialocephala humicola				1				4	6.25
Phialophora sp.		1	2	4		1	3	2	16.25
Phoma sp.	1		1		2	1	1		7.50
Phomopsis sp.	1	2			2				6.25
Phyllosticta capitalensis			1		1				2.50
Pyricularia costina	4	3			2	1			12.50
Pyricularia longispora	2	1		3					7.50
Pyriculariopsis sp.	1	1	2			1			6.25
Ramichloridium musae	2	1		1				1	6.25
Septonema sp.	2	1	1				3		8.75
Solosympodiella sp.	1		1	2		1		4	11.25
Sporidesmium leonense			1		1		2	1	6.25
Sporidesmium sp.						1			1.25
Stachybotrys echinata	2	1	1	1		1	1		8.75
Stachybotrys sp.	1			1	2				5.00
Stachylidium sp. 1				2					2.50
Stachylidium sp. 2	2	1	1		3	3	1		13.75
Stilbohypoxylon sp.				1					1.25
Subulispora sp.				1					1.25
Synematous sp.				1					1.25
Torula sp. 1		1	1					1	3.75
Torula sp. 2			1	1					2.50
Trichoderma sp.				2					2.50
Veronaea musae		1	1				1	2	6.25
Verticillium sp. 1	1	1		1	2	2	1	3	13.75
Verticillium sp. 2	2	1	1	1	1	1			8.75
Verticillium sp. 3	3	2	1	3	1	4	1	1	20.00
Xenosporium amomi				1					1.25
Xenosporium intermedium				1					1.25
Xenosporium thaxeri				1					1.25
Basidiomycetes (3)	1	1			1				
Ascomycetes (10)	1	1	4	8			3	3	
Anamorphic fungi (75)	27	42	36	58	25	33	24	31	
Total taxa (88)	**29**	**44**	**40**	**66**	**26**	**33**	**27**	**34**	

L = leaf, P = pseudostem, p = samples collected from standing dead plant, g = samples collected from dead plant laying on the forest ground

Table 4.5 Overall percentage occurrence and frequency of fungal taxa recovered from *Etlingera littoralis*.

Taxa	**Huay Kok Ma**				**Medicinal Plant Garden**				**Overall percentage occurrence**
	Lp	**Lg**	**Pp**	**Pg**	**Lp**	**Lg**	**Pp**	**Pg**	
Acremonium sp. 1	3	2	4	4	1	2	4	2	27.50
Acremonium sp. 2		1	1	1		1	2	2	10.00
Agaricus sp.								1	1.25
Anthostomella sp.			1	1					2.50
Anthracophyllum nigritum								1	1.25
Ascomtcete 2				1					1.25
Basidiomycete 1					4	2			7.50
Basidiomycete 2					2				2.50
Basidiomycete 3						1			1.25
Basidiomycete 4								1	1.25
Basidiomycete 6								2	2.50
Botryodiplodia sp.	1				1	1			3.75
Brachysporiella sp.				3			1	1	6.25
Canalisporium caribense			1	3		1	2	2	11.25
Canalisporium exiguum		1		1		1			3.75
Cephalosporiopsis		1		1		1	1	1	6.25
Cercophora sp.				2			1	1	5.00
Cercospora sp.	2	1				2			6.25
Chalara sp.		1	3	1		1	1	2	11.25
Chloridium botryoideum		1		1		1		1	5.00
Chloridium sp.		1	3	5	1		2	4	20.00
Cladosporium cladosporioides	2	1	1	1	4	3		3	18.75
Clavaria sp.				3				2	6.25
Colletotrichum gloeosporioides			1					2	3.75
Corpinus disseminatus								1	1.25
Crepidotus sp. 1			1	1					2.50
Crepidotus sp. 2						1			1.25
Cyathus sp.				1					1.25
Cylindrocladium sp.						1	1		2.50
Dactylaria hemibeltranioidea	2	1		4	3	2	1	2	18.75
Dactylaria sp. 1				1	2	2	1	3	11.25
Dactylaria sp. 2	3	3	1	2	1	1	1	1	16.25
Dactylella ellipsospora		6		3		1		1	13.75
Dactylella sp.		2	1	1		4	1	3	15.00
Dictyoarthrinium sacchari				1					1.25
Dictyosporium heptasporum							1	1	2.50
Discomycete	1								1.25
Fusarium sp. 1		1						1	2.50
Gliomastix sp.				2				2	5.00
Helicosmyces bellus			1	1					2.50
Helicomyces sp.			1	3		1		1	7.50
Helicosporium sp.		1		1					2.50
Hormomyces sp.			1	1		2		1	6.25
Hyphomycete 5			1	2					3.75
Hypocrea rufa				2					2.50

Table 4.5 (Continued).

Taxa	**Huay Kok Ma**				**Medicinal Plant Garden**				**Overall percentage occurrence**
	Lp	**Lg**	**Pp**	**Pg**	**Lp**	**Lg**	**Pp**	**Pg**	
Hypocrea sp.			1	1			1	1	5.00
Hyponectria sp.			1	3					5.00
Japhneadelphus sp.						1			1.25
Marasmiellus inoderma						1		1	2.50
Marasmius sp. 1								1	1.25
Marasmius sp. 2		1		1					2.50
Marasmius sp. 3					2	1			3.75
Microsphaeropsis sp.					2	1	3		7.50
Monodictys sp.		1		1			1	1	5.00
Myrothecium cinctum							1	1	2.50
Nakataea fusispora	3								3.75
Nectria sp. 1			5	2			4	1	15.00
Nectria sp. 2				2			1	1	5.00
Orbilia sp.				1					1.25
Paecilomyces sp.		2	1	1	1	1			7.50
Penicillium sp.		1	2	4			2	2	13.75
Periconia digitata	3	1	1		1	2			10.00
Periconia minutissima					1				1.25
Pestalotiopsis sp.	1								1.25
Phialocephala humicola		1				1	1		3.75
Phoma sp.			1						1.25
Phomopsis sp.			1				2		3.75
Phyllosticta capitalensis		1							1.25
Pyricularia costina	2				1	1			5.00
Pyricularia sp.	1	1		1		1		1	6.25
Ramichloridium musae					1	1	1		3.75
Stachybotrys echinata		1			1	1			3.75
Stachybotrys sp.				1				1	2.50
Stachylidium sp. 2	2	1			1	1	1		7.50
Subulispora sp.		1			1	1			3.75
Trichoderma sp.		2							2.50
Verticillium sp. 1	3	2			1	3			11.25
Xenosporium intermedium			1	1		2	1		6.25
Xenosporium thaxeri								1	1.25
Basidiomycetes (16)		1	1	4	3	5		6	
Ascomycetes (11)	1		4	9		1	4	4	
Anamorphic fungi (52)	13	27	20	28	17	30	22	28	
Total taxa (79)	**14**	**28**	**24**	**41**	**20**	**36**	**26**	**38**	

L = leaf, P = pseudostem, p = samples collected from standing dead plant, g = samples collected from dead plant laying on the forest ground

Table 4.6 Overall percentage occurrence and frequency of fungal taxa recovered from *Etlingera elatior*.

Taxa	**Queen Sirikit Botanic Garden**		**Chiang Mai University**		**Overall percentage occurrence**
	Leaf	**Pseudostem**	**Leaf**	**Pseudostem**	
Acremonium sp. 1	2	6	3	4	37.5
Acremonium sp. 2	1	3	1	1	15.0
Ascomycete 2		1			2.5
Botryodiplodia sp.			1		2.5
Brachysporiella sp.		1			2.5
Cephalosporiopsis sp.		2		2	10.0
Cercospora amomi		2		1	7.5
Chalara sp.	1	1	1	1	10.0
Chloridium sp.	1	3	1	2	17.5
Cladosporium cladosporioides	5		3		20.0
Cladosporium uredinicola	1		1	1	7.5
Colletotrichum gloeosporioides	4		3	2	22.5
Curvularia lunata	3	1	1		12.5
Dactylaria hemibeltranioidea	4	4	1	2	27.5
Dactylaria sp. 1	2	3			12.5
Dactylaria sp. 2	2		3	1	15.0
Dactylella ellipsospora	1	2		1	10.0
Dactylella sp.	1	1	1	1	10.0
Dictyoarthrinium sacchari	4	4			20.0
Fusarium sp. 1		1		1	5.0
Fusarium sp. 2	1		1	1	7.5
Gliomastix sp.	1	2			7.5
Helicomyces bellus		1			2.5
Helicomyces sp.		1			2.5
Helicosporium sp.	1				2.5
Memnoniella subsimplex	3	1	1	1	15.0
Nigrospora oryzae				1	2.5
Paecilomyces sp.	2		1		7.5
Penicillium sp.		1			2.5
Periconia digitata	1	4	1	2	20.0
Periconia minutissima	1	2	2	2	17.5
Pestalotiopsis sp.	1		1		5.0
Phoma sp.		2			5.0
Septoria sp.	2				5.0
Stachybotrys echinata			3		7.5
Stachybotrys sp.	1				2.5
Stachylidium sp. 2	2	1			7.5
Trichoderma sp.		1	1	1	7.5
Verticillium sp. 1	1				2.5
Ascomycetes (1)		1			
Anamorphic fungi (38)	26	24	20	19	
Total taxa (39)	**26**	**25**	**20**	**19**	

Table 4.7 Overall percentage occurrence and frequency of fungal taxa recovered from *Alpinia galanga*.

Taxa	**Chiang Mai**		**Lampang**		**Overall percentage occurrence**
	Leaf	**Pseudostem**	**Leaf**	**Pseudostem**	
Alternaria alternata	4	1	1		15.0
Aspergillus niger		2	1		7.5
Cercospora amomi		2	1	1	10.0
Cladosporium cladosporioides	2	4	1	3	25.0
Colletotrichum gloeosporioides	2	3	4	1	25.0
Colletotrichum sp.	1				2.5
Cuvularia lunata	2	5	3	3	32.5
Dactylaria intermedia	2	1			7.5
Fusarium sp. 2	1	2	1	1	12.5
Fusarium oxysporum	2	1			7.5
Gliomastix sp.				1	2.5
Leptosphaeria alpiniae	3	1	2		15.0
Memnoniella sp.			1		2.5
Memnoniella subsimplex	3		1	1	12.5
Nigrospora oryzae		1	1	2	10.0
Periconia minutissima	1				2.5
Phoma sp.	1		3	1	12.5
Phomopsis sp.	4	2	4	4	35.0
Phyllosticta capitalensis		1	1	1	7.5
Pithomyces chatarum		2			5.0
Septoria sp.			1		2.5
Stachybotrys echinata	2	1			7.5
Trichoderma sp.	1	1	2	1	12.5
Verticillium sp. 1	3	1	1	1	15.0
Ascomycetes (1)	1	1	1		
Anamorphic fungi (23)	15	16	16	12	
Total taxa (24)	**16**	**17**	**17**	**12**	

Table 4.8 Overall percentage occurrence and frequency of fungal taxa recovered from *Zingiber officinale*.

Taxa	**Phayao (commercial)**		**Phayao (backyard)**		**Overall percentage occurrence**
	Leaf	**Pseudostem**	**Leaf**	**Pseudostem**	
Ascomycete 3	3	2	2	4	27.5
Aspergillus niger	1	2	2	5	25.0
Bipolaris sp.	2		1		7.5
Cercospora zingibericola	2		1		7.5
Cladorrhinum foecundissinum				2	5.0
Cladosporium cladosporioides	2	2	4	1	22.5
Cladosporium oxysporum	1	5	3	4	32.5
Colletotrichum gloeosporioides		2	2	1	12.5
Curvularia lunata	1	6	4	5	40.0
Dactylaria hemibetranioidea	2	1	1	1	12.5
Fusarium oxysporum	6	3	1	4	35.0
Gelasinospora sp.		1		2	7.5
Gliocladium sp.				1	2.5
Glomerella sp.	1	2		1	10.0
Hyphomycete 7	5	5	8	6	60.0
Myrothecium cinctum	4	1	2		17.5
Myrothecium inundatum	2		1		7.5
Nigrospora oryzae	2		3		12.5
Paecilomyces sp.	2	3	2	4	27.5
Phaeosphaeria sp.	7	6	10	9	80.0
Phomopsis sp.	8	2	10	2	55.0
Phyllosticta capitalensis	2	2	3	1	20.0
Stachybotrys sp.	1	1	3	2	17.5
Trichoderma sp.	2	1	1	3	17.5
Vermiculariopsiella sp.	3	2			12.5
Ascomycetes (4)	3	3	2	4	
Anamorphic fungi (21)	18	16	18	15	
Total taxa (25)	**21**	**19**	**20**	**19**	

4.3.3 Effect of site, type of sample (forest ground and standing plant) and tissue type (leaf and pseudostem) on fungal communities

Three dimensional correspondence analysis plots of fungal communities on different levels (site, tissue type, season or position of dead samples) of each host are presented in Figure 4.4. Percentages of total variance explained by the models are 48–100%.

Distinct fungal communities were found on the leaves and pseudostems samples of the six zingiberaceous species (Figures 4.4a–f). This is indicated by more or less the separation of the samples from leaves and pseudostems. In *Amomum siamense*, similarity of fungal communities on leaves was higher than on pseudostems. This is indicated by the closer cluster formed by all leaves of fallen and standing samples collected in different sites and seasons (circled in Figure 4.4b). By contrast, in *Etlingera litoralis* and *Zingiber officinale* similarity of fungal communities on pseudostems was higher than on leaves. These are indicated by the closer clusters formed by all samples of pseudostems from each collection (circled in Figure 4.4c, f). The results from these three species showed that the tissue types had the strongest influence on the fungal communities.

The fungal communities on samples of *Etlingera elatior* showed a degree of difference influenced by both tissue type and site, indicated by clearly separated clusters for all collections (Figure 4.4d). Fungal species compositions on *Alpinia malaccensis* were affected by situation of samples, tissue types and sites (Figure 4.4a). This is indicated by the separation of the samples from each other. The samples collected from Doi Pui (circled in Figure 4.4a), however, showed that tissue type was a stronger influence on fungal communities than the situation of samples. This is indicated by the closer distances of the samples from the same tissue type than samples from dead standing plants or plants on the forest ground.

Correspondence analysis of *Alpinia galanga* and *Amomum siamense* showed some evidence of an effect of collecting site on fungal compositions. This is indicated by the closer distances of the samples on leaves and pseudostems of *A. galanga* collected in the same site than in different sites (Figure 4.4e) For *A. siamense*, with the exception of all pseudostem samples from Huay Kok Ma, there are closer distances of the samples on leaves or pseudostems of fallen or standing plants, collected in the same site than in different sites (Figure 4.4b).

For *Amomum siamense*, although the fungal communities on leaf samples collected from different sites in Doi Suthep-Pui National Park were similar (indicated by the closed cluster formed by all samples of leaves from each collection), those leaves collected in Huay Kok Ma, from fallen and standing samples were distinct (indicated by the separation between the samples from the ground: W1Lg and D1Lg and standing plants: W1Lp and D1Lp) (Figure 4.4b). The exclusive presence of *Staurophoma* sp. on leaves of dead plants on the forest ground, and Discomycete and *Mycena* sp. on leaves of standing dead plants attributed to the distinct fungal communities found on these sample situations (Table 4.3).

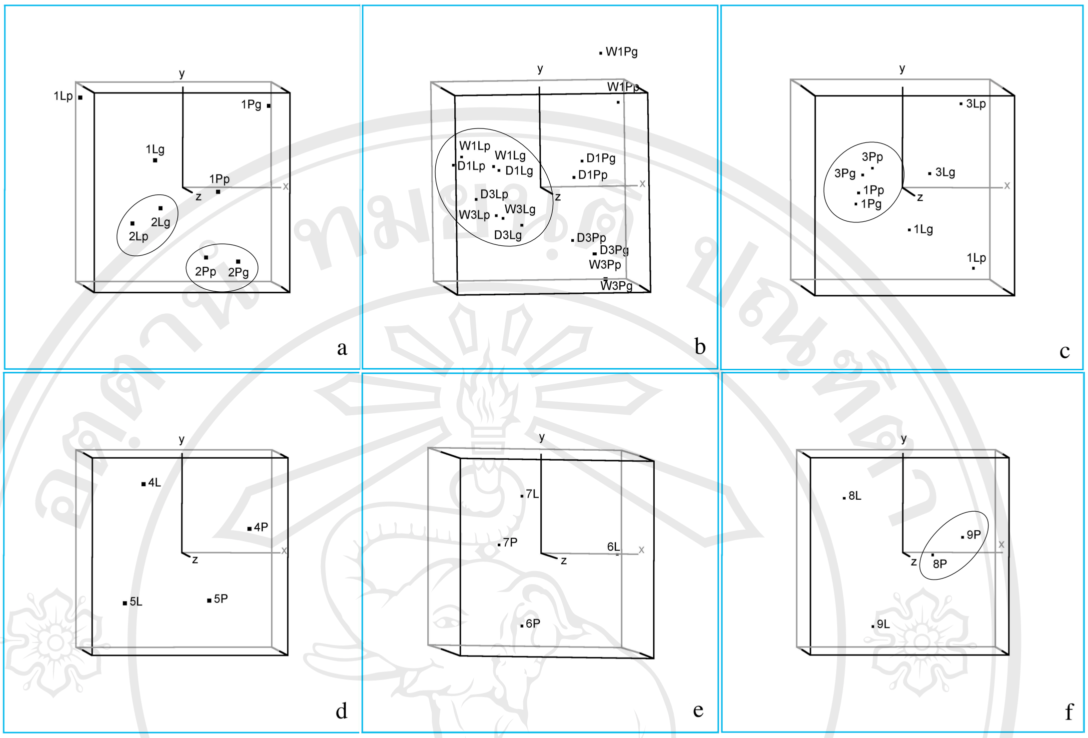

Figure 4.4 Three dimensional correspondence analyses for the fungal compositions of Zingiberaceae: a. *Alpinia malaccensis*, b. *Amomum siamense*, c. *Etlingera littoralis*, d. *E. elatior*, e. *Alpinia galanga*, f. *Zingiber officinale*. W. wet season, D. dry season, 1. Huay Kok Ma, 2. Doi Pui, 3. Medicinal Plant Garden, 4. Queen Sirikit Botanic Garden, 5. Chiang Mai University area, 6. Chiang Mai Province (Hangdong), 7. Lampang Province (Muang), 8. Phayao Province (commercial), 9. Phayao Province (backyard), L. leaf, P. pseudostem, g. collection from forest ground, p. collection from standing dead plant. Percentages of total variance explained by the models (a–f) are 65%, 48%, 61%, 100%, 100% and 100% respectively.

4.3.4 Effect of host on fungal communities

The host plants especially between the wild and cultivated species, affected fungal communities. The Sorensen indices showed that fungal taxa occurring on samples between wild species (45–76%) were most similar and relatively higher than samples between the two cultivated species (37%). The similarities of fungi recovered from samples between wild and cultivated species were low (14–44%) (Table 4.9). Correspondence analysis also indicated that the fungal communities on the four wild species were more similar than to cultivated species as expressed by the close cluster formed by both leaf and pseudostem samples of the four wild species collected from each of the various sites (circled in Figure 4.5). One hundred and thirteen fungi (e.g., *Acremonium* sp.1, *Canalisporium caribense*, *Chloridium* sp., *Dactylaria* sp. 2, *Dactylella ellipsospora*, *Dactylella* sp., *Periconia digitata*, *Pyricularia longispora*, *Stachylidium* sp. 2, *Verticillium* sp. 3) were found only on the wild species, while 17 fungi (e.g., *Alternaria alternata*, Ascomycete 3, *Cladosporium oxysporum*, *Fusarium oxysporum*, Hyphomycete 7, *Phaeosphaeria* sp.) were found only the cultivated species (Table 4.2). Fungal communities between the cultivated species were also distinct, indicated by the two separated clusters formed by the samples on *A. galanga* (at top) and *Z. officinale* (at right) (Figure 4.5). Fourteen fungi (e.g., *Alternaria alternata*, *Fusarium* sp. 2, *Leptosphaeria alpiniae*, *Verticillium* sp. 1) were found only on *A. galanga*, while 16 fungi (e.g., Ascomycete 3, *Cladosporium oxysporum*, Hyphomycete 7, *Phaeosphaeria* sp.) were found only on *Z. officinale* (Table 4.2).

Table 4.9 Similarity of fungal taxa compositions between Zingiberaceae.

Sorenson index (%)	*Amomum siamense*	*Etlingera littoralis*	*Etlingera elatior*	*Alpinia galanga*	*Zingiber officinale*
Alpinia malaccensis	76	62	52	21	16
Amomum siamense		57	45	24	14
Etlingera littoralis			54	19	17
Etlingera elatior				44	25
Alpinia galanga					37

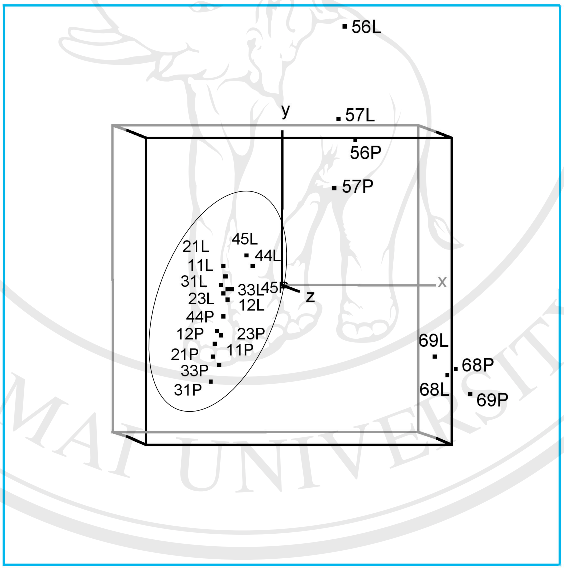

Figure 4.5 Three dimensional correspondence analyses for the fungal compositions of Zingiberaceae. The first digit refers to plant species: 1. *Alpinia malaccensis*, 2. *Amomum siamense*, 3. *Etlingera littoralis*, 4. *E. elatior*, 5. *Alpinia galanga*, 6. *Zingiber officinale*. The second digit refers to sites: 1. Huay Kok Ma, 2. Doi Pui, 3. Medicinal Plant Garden, 4. Queen Sirikit Botanic Garden, 5. Chiang Mai University, 6. Chiang Mai Province (Hangdong), 7. Lampang Province (Muang), 8. Phayao Province (commercial), 9. Phayao Province (backyard), L. leaf, P. pseudostem. Percentage of total variance explained by the models is 41%.

4.3.5 Effect of season on fungal communities

Only samples of *Amomum siamense* were collected in wet and dry season, and there was little effect of season on the pattern of fungal occurrence on all samples collected in Doi Suthep-Pui National Park. In the correspondence analysis, fungal communities on the wet (W) collections do not form a coherent cluster separate from the fungal communities on the dry (D) collections. Only the fungal communities on pseudostems from Huay Kok Ma in wet collections (W1Pg and W1Pp) were markedly different from those on pseudostems from Huay Kok Ma in dry collections (D1Pg and D1Pp), indicated by the distinct separation between wet and dry collections of pseudostems collected from standing plant or the forest ground (Figure 4.2b). The exclusive presence of 16 fungi (*Bahusandhika sundarum*, Basidiomycete 5, *Berkleasmium* sp., *Crepidotus* sp. 2, *Dictyosporium* sp. 1, *Dictyosporium* sp. 2, *Gaeumannomyces amomi*, *Gliomastix* sp., *Hypocrea rufa*, *Hyponectria* sp., *Marasmiellus* sp., *Marasmius* sp. 1, *Pleospora* sp., Synematous sp., *Verticimonosporium* sp. and *Xenosporium* sp.) in wet collections of pseudostem collected from dead standing plant or on the forest ground at Huay Kok Ma attributed to the distinct fungal communities found on these samples in the wet collections (Table 4.3).

The collections from *A. siamense* showed that the effect of tissue type on fungal species composition was greater than the effect of seasons. This is indicated by clusters formed by samples on the leaves and pseudostems with the higher similarity of fungal communities on leaves collected from different sites or seasons (circled in Figure 4.4b).

4.4 Discussion

4.4.1 Biodiversity and host specificity

In this study, high numbers of fungal taxa were found (163 fungi were identified from 440 samples of the four wild and two cultivated zingiberaceous species). Although no previous studies have concentrated on the saprobic fungi on Zingiberaceae, 300 fungal taxa have been reported from members of the Zingiberaceae (Appendix C). Surprisingly, only 18 of these fungal species were found in the present study. These results suggest that the fungi on Zingiberaceae are diverse and could be a source for more undescribed species (Bussaban *et al*., 2001b). Eight new fungi were described during this study—*Berkleasmium nigroapicale*, *B. sutheppuiense*, *Gaeumannomyces amomi*, *Leiosphaerella amomi*, *Pyricularia kookicola*, *P. longispora*, *P. variabilis* and *Xenosporium amomi* (Bussaban *et al*., 2001a, c, 2003a, b) and seven fungi (*Berkleasmium* sp., *Dactylaria* sp. 1, *Dactylaria* sp. 2, *Dictyosporium* sp. 1, *Dictyosporium* sp. 2, *Pyricularia* sp. and *Xenosporium* sp. marked with asterisk in Table 4.3) are probably new species (informally described in Appendix C).

The fungi recorded in the present study can be compared with those recorded on other monocotyledonus plants. Photita *et al*. (2001a, 2003b) identified 46 fungi from *Musa acuminata* in Hong Kong and 80 on the same host in Thailand (Doi Suthep-Pui National Park). Only five of these taxa from Hong Kong and ten from Thailand were found on Zingiberaceous species in the present study (*Alternaria alternata*, *Canalisporium caribense*, *Claodosporium cladosporioides*, *C. oxysporum*, *Colletotrichum gloeosporioides*, *Dictyoarthrinium sacchari*, *Dictyosporium* *heptasporum*, *Memnoniella subsimplex*, *Nigrospora oryzae*, *Periconia digitata* and *Pithomyces chatarum*). *Canalisporium caribense* and *Dictyosporium heptasporum* were also found on Pandanaceae (McKenzie *et al*., 2002). Yanna (2001) found 288 fungi on decaying palm fronds in Australia, Brunei and Hong Kong, but only five of these taxa were found on zingiberaceous species: *Chloridium botryoideum*, *Dactylella ellipsospora*, *Myrothecium cinctum*, *Periconia minutissima* and *Ramichloridium musae*. Fröhlich and Hyde (1999) identified 242 fungi from six individual *Licuala ramsayi* palms from Australia and Brunei, while Pinnoi (2004) found 114 fungi from *Eleiodoxa conferta* in Thailand. However, no fungi from these two studies were found in the present study. Hyde *et al*. (2002a) identified 85 taxa from bamboo (*Bambusa* spp. and *Dendrocalamus* spp.) culms. Three species, *Curvularia lunata*, *Cladosporium cladosporioides* and *Canalisporium caribense*, overlapped with the present study. The overlap of fungi occurring on zingiberaceous species with those recorded from studies of other monocotyledonous hosts is thus low. This may be influenced by climate and geographical distribution, and/or host-specificity and host-recurrence of the fungi (Photita *et al*., 2001a; Yanna, 2001; Zhou and Hyde, 2001). Saprobic fungi are unlikely to be host specific and, therefore, host specificity is unlikely to influence saprobic fungal diversity. An important factor that may influence the biodiversity of saprobic fungi on various hosts is therefore host-recurrence. The reasons as to why fungi may occur recurrently on certain hosts is not understood, but may be related to the presence of these fungi as endophytes. It is unlikely that climatic conditions played a significant part in determining the fungal compositions in the studies mentioned above since all were carried out in the tropics, and some were also conducted in Thailand.

4.4.2 Comparison of fungi on Zingiberaceae

Among the six zingiberaceous species studied, there were differences in terms of fungi and numbers of fungal taxa recovered. More fungal taxa were found in the wild species, indicating that the fungi in the wild species, is more diverse than in the cultivated species. The Shannon indices (Figure 4.3) also indicated more diversity in wild than in cultivated species.

The fungal communities among wild zingiberaceous species also differ, and this may be dictated by the differences in host morphology and anatomy, and differences in habitat. *Etlingera elatior* is morphologically and anatomically similar to *E. littoralis*, thus the difference in fungal diversity and community structure may be explained by environmental conditions of the habitats they are found in. *Etlingera elatior* is a wild species but the plants studied were grown in artificial forest and Chiang Mai University area, with environment conditions usually different from those in the forest (*E. littoralis*) with its high humidity, high rainfall and less human disturbance. *Etlingera littoralis* and *Amomum siamense* were found in the same localities in Doi Suthep-Pui National Park the differences in their fungal communities may result from morphological and physiological differences of the host plants. More basidiomycetes were found on *E. littoralis*, while more ascomycetes and anamorphic fungi were found on *A. siamense*.

The fungal communities on wild zingiberaceous species differs from that on cultivated species in terms of fungi and number of fungal species. Although *Alpinia* *galanga* is morphologically similar to *A. malaccensis*, the difference in fungal diversity and structure can be explained by environmental conditions of the habitats in which they are found. *Alpinia malaccensis* is a wild species, and where it grows environmental conditions such as climate, temperature, rain and humidity are different to those in a cultivated area where *A. galanga* is found.

4.4.3 Tissue specificity

Different fungal communities were found on leaves and pseudostems of dead zingiberaceous species. More fungi occurred on the pseudostem than on the leaf, and on leaves most fungi occurred along the midvien. Tsoumis (1991) reported various components and quantities of cellulose, hemicellulose and pectin in different plant tissue types. This may account for the fungi confined to specific tissues as some fungi may have different enzyme systems that can degrade different substrata. More ascomycetes and basidiomycetes occurred on pseudostems of *Alpinia malaccensis*, *Amomum siamense* and *Etlingera littoralis* (Table 4.3–4.5). Pseudostems contain more concentrated supportive tissue, a higher nutrient content than leaves, and they have spongy cell walls that may take up water and retain moisture for a longer time. This may then allow growth and sporulation of ascomycetes and basidiomycetes fungi which, in general, produce larger fruiting bodies.

More fungi were found on pseudostem and leaf samples collected from the forest ground than from standing samples. The higher moisture level on the forest floor may account for the greater number of fungi in this microhabitat. Moisture retention and humidity in the substratum will help fungi to grow (Dix and Webster, 1995). High availability of moisture increases diffusion of solutes to the fungi and therefore increased enzyme activities (Carroll and Wicklow, 1992; Dix and Webster, 1995). Fungi typically recovered from soil, e.g., *Aspergillus*, *Penicillium*, *Trichoderma* and *Paecilomyces* spp. appear to be active in leaf litter decomposition (Frankland, 1992). These genera were more often found on leaves and pseudostems from the forest ground than on samples from standing plants. This may also account for the greater number of fungi on samples from forest ground.

Fungal tissue recurrence has been reported with other hosts, and it has been shown that the fungi on standing hosts were vertically distributed (Sadaba *et al*., 1995; Poon and Hyde, 1998; Hyde *et al*., 2002b). There were more ascomycetes on the lower culm tissues of *Phragmites australis* comprising sclerenchyma, and more anamorphic taxa on the upper herbaceous tissues (Poon and Hyde, 1998). This may, however, have been influenced by moisture as the bases were submerged. Sadaba *et al*. (1995) found different fungal communities on herbaceous and woody parts of *Acanthus ilicifolius*, more ascomycetes occurring on the lower woody part and more anamorphic taxa on the upper herbaceous parts. Hyde *et al*. (2002b) also found different fungal communities on the basal, middle and apical portions of bamboo culms. The recurrence of certain fungi on different tissue types may be due to differences in nutrition requirements, or the ability of the fungi to utilise different substrates (Adaskaveg *et al*., 1991; Ingold and Hudson, 1993). Soft and woody tissues of *Musa acuminata* and *Magnolia lillifera* have been found to support different groups of saprobic fungi (Photita *et al*., 2001a, 2003b;

Kodseub *et al*., 2004). Different plant tissues and organs may in fact resemble distinct microhabitats (Petrini *et al*., 1992b). Alternatively, it may be related to the distribution of endophytes. If these fungi become saprobes after the death of their hosts, the saprobes may be restricted to the tissues in which they have been living as endophytes. In the present study some fungi, e.g., *Colletotrichum gloeosporioides* were recovered from both leaves and pseudostems, while *Phyllosticta capitalensis* and *Pyricularia costina* were mainly recovered from leaves. This is a similar distribution as found for these fungi in the endophytic study (Chapter 3).

4.4.4 Abundance of anamorphic fungi

Most taxa recovered in the present study are anamorphic fungi (72%) with fewer ascomycetes (16%) and basidiomycetes (12%) recovered. Anamorphic fungi were the most abundant fungi recorded at all collections. Several hyphomycetes encountered in this study have been previously recorded on zingiberaceous species (Appendix C). These fungi can also occur on large woody monocotyledons, e.g. *Pandanaceae* (McKenzie and Hyde, 1997; McKenzie *et al*., 2002). Anamorphic taxa were also the most frequently recorded fungi on other monocotyledonous hosts, e.g., *Heliconia* (Bills and Polishook, 1994b), grasses (Wong and Hyde, 2001), palms (Yanna, 2001), and bamboo (Hyde *et al*., 2001, 2002a, b).

Zingiberaceous plant tissues are herbaceous and decay rather quickly. The fungi on its tissues therefore need to sporulate rapidly in order to disperse their spores before the tissues completely break down. A high proportion of the fungi appear to be anamorphic states of fungi in Hypocreales, e.g., *Fusarium*, *Gliocladium*, *Myrothecium*, *Stachybotrys*, *Stilbella*, *Trichoderma*, *Verticillium*, and probably some *Acremonium* spp.; these anamorphs generally sporulate faster than the teleomorphs. Most anamorphic taxa will usually sporulate in culture within one week to a month, while the teleomorph may take considerably longer, or often fail to sporulate (Hyde *et al.*, 1987). This may account for the dominance of anamorphic taxa found on herbaceous tissues in this study.

4.4.5 Plant pathogens

Costaceae, Musaceae and Zingiberaceae may occupy similar habitats, and the genera have been considered closely related in Zingiberales. Therefore, it might be expected that some fungi originally described on leaves of *Costus* spp. or *Musa* spp. could be isolated from Zingiberaceae. Some widespread facultative pathogens of *Musa* spp., e.g., *Fusarium oxysporum*, were recovered in the present study, although in low frequencies, *Ramichloridium musae*, a fungus generally associated with *Musa* spp. was also found. *Phyllosticta capitalensis* recovered from zingiberaceous plants in the present study is known to be parasitic on Orchidaceae (van der Aa, 1973; Okane *et al.*, 2003). *Cercospora amomi*, *C. zingibericola* and *Curvularia lunata* which are known to be pathogens of Zingiberaceae (Ramakrishnan, 1942; Kar and Mandal, 1969; Farr *et al.*, 1989; Sontirat *et al.*, 1994) were also identified as saprobes in this study.