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Succession and natural occurrence of saprobic fungi on leaves of *Magnolia liliifera* in a tropical forest

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Abstract – Leaves of *Magnolia liliifera* were selected to evaluate fungal diversity, and succession of fungi during leaf decomposition, and the effect of baiting on fungal diversity. The leaf samples were from Doi Suthep-Pui National Park, Chiang Mai, Thailand. Twenty-three taxa were identified on senescent leaves of *M. liliifera* during the decay process. Distinct fungal communities were observed with the dominant species on the leaves being different at each succession stage. The most abundant fungal species were *Hyponectria* sp. 1 (on 60% of leaves), *Volutella* sp. 1 (60%), *Gliocladium* sp. 3 (37.1%), *Corynespora cassiicola* (34.3%), *Bionectria ochroleuca* (25.7%), *Cylindrocladium floridanum* (22.8%), *Phaeosphaeria* sp. (17.1%), *Dactylaria longidentata* (11.4%) and *Lasiosphaeria* sp. (11.4%). Leaf bait trials did not show any noticeable effect on fungal diversity when either the upper or lower leaf surface was adjacent to the forest floor. Highest fungal diversity on leaves of *M. liliifera* occurred between day 4 and 40, with most species being present on day 40. On day 56, leaves were found to be skeletonized, and the fungal communities had decreased in number.

Fungal diversity / fungal ecology / fungal succession / leaf baiting / leaf decomposition

INTRODUCTION

Fungal succession on decaying plant material has been observed in both terrestrial and aquatic ecosystems of tropical and temperate regions (Hering, 1965; McKenzie & Hudson, 1976; Gessner *et al.*, 1993; Ho *et al.*, 2002; Somrithipol *et al.*, 2002; Suzuki *et al.*, 2002; Yanna *et al.*, 2002; Handa & Harada, 2005; Paulus *et al.*,

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2006; Kodsueb *et al.*, 2015). Most studies focused on the change in composition of fungal communities with changes of plant species, following different types of disturbance, such as animal, fire, land slide and deforestation (McMullan-Fisher *et al.*, 2002; Suzuki *et al.*, 2002). There have been numerous studies on substratum succession involving a variety of substrata, for example, cellulose film (Tribe, 1957; 1961), other cellulosic substrates (Gorska, 1982), plant litter (Promputtha *et al.*, 2002; Tokumasu & Aoiki, 2002; Yanna *et al.*, 2002; Zhou & Hyde, 2002; Handa & Harada, 2005; Paulus *et al.*, 2006), rust-infected and non-infected litter (McKenzie & Hudson, 1976), and wool (Ghawana *et al.*, 1997).

There have been few studies on fungal succession in Thailand, a tropical region. Somrithipol *et al.* (2002) investigated the succession of fungi on fruits and seeds of *Delonix regia* exposed on the forest floor. Sivichai *et al.* (2002) studied fungal colonization on two timbers, *Dipterocarpus alatus* and *Xylia dolabriformis*, exposed in a freshwater stream. Promputtha *et al.* (2002) investigated fungal succession on senescent leaves of *Manglietia garrettii* (later corrected to *Magnolia liliifera*) on the forest floor.

In this paper, we examined fungal succession on freshly fallen leaves of *Magnolia liliifera* and evaluated the fungal diversity, over the period of leaf decomposition. The fungal communities which were found through the succession were compared with naturally occurring leaf samples (Promputtha *et al.* 2002). The effect of either the upper or lower leaf surface touching the forest floor on fungal communities was also examined.

MATERIALS AND METHODS

A forest area of Doi Suthep-Pui National Park, Chiang Mai, Thailand, was chosen for this study. One-hundred and ten senescent leaves of *Magnolia liliifera* were collected from the park in July (wet season). The selected senescent leaves had just fallen, and were yellow-green with a fresh abscission scar. Ten leaves were randomly selected to represent day 0.

The remaining 100 leaves were randomly divided into two groups of 50 leaves. Leaves from each group were marked separately by tying either bright red wool or bright orange wool to the petioles. The leaves were then randomly placed under *M. liliifera* trees over an area of about 10,000 square meters. Fifty leaves with red wool were arbitrarily placed under the trees, with their lower surface touching the forest floor and the other 50 leaves with orange wool were placed with the upper surface touching the forest floor. At each sampling, five marked senescent leaves with their lower surface touching the forest floor and five marked senescent leaves with their upper surface touching the forest floor were randomly collected. It was planned to collect marked senescent leaves at day 4, 8, 16, 24, 40, 56, 72 and 88 (summarized experimental design of the trial is shown in Fig. 1). However, at day 56, leaves were highly skeletonised comprising vascular tissue with attached remnants of non-vascular tissue. Therefore day 72 and day 88 collections were not made.

Samples were placed in separate plastic bags in the forest and taken back to the laboratory. To induce fungal sporulation the samples were incubated individually in the plastic bags with the addition of tissue paper moistened with sterilized water. Samples collected at day 0 were examined for the presence of

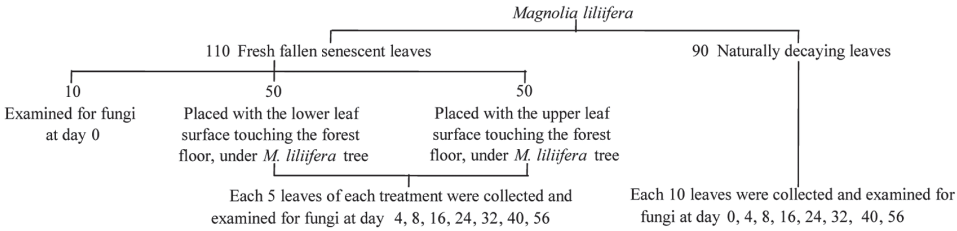

Fig. 1. Experimental design of leaf bait trials for fungal diversity examination throughout decomposition period of *Magnolia liliifera* leaves.

microfungi on the same day of collection. All other samples were examined under a microscope for the presence of microfungi immediately after collection and then periodically for up to two weeks. Mounts of sporulating fungi were made in water for examination with differential interference contrast microscopy, and where feasible fungi were isolated by single spore isolation (Chomnunti *et al.*, 2014). Herbarium specimens were prepared by air-drying in an oven at 37°C, for one week.

During the experimental period, 90 naturally fallen decaying leaves of *M. liliifera* at various stages of decay were also collected for comparison of fungal communities with those from the succession study. The results were published by Promputtha *et al.* (2004) and are used for comparison here.

Statistical analysis and sample calculation

The fungi found in this study are presented in terms of percentage occurrence. Fungal taxa with an overall percentage occurrence equal to or higher than 10 are regarded as dominant species. These taxa are plotted to illustrate changes in the dominant species throughout the experimental period.

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

Percentage similarity = 2c/a+b

a: the number of species in upper surface touching the floor

b: the number of species in lower surface touching the floor

c: the number of species in common in upper and lower surface

A 3-dimensional correspondence analysis was performed to examine the differences in fungal communities at different times of decay, and to see the effect of which surface of the leaf was touching the forest floor.

RESULTS

Fungal diversity

Twenty-three taxa were identified on leaves of *M. liliifera* during the decay process and their percentage occurrences are listed in Table 1. These results are separated into those fungi on leaves with their upper surface touching the forest floor

Table 1. Number of decaying leaves of *Magnolia liliifera* on which fungi occurred during the succession process

Fungus	*Upper leaf surface touching forest floor*									*Lower leaf surface touching forest floor*									*Overall percentage occurrence*
	Day							*TL**	*%*	*Day*							*TL**	*%*	
	0	*4*	*8*	*16*	*24*	*40*	*56*			*0*	*4*	*8*	*16*	*24*	*40*	*56*			
Phyllosticta capitaliensis	1	1	1					3	8.6		1							2.9	5.7
Leptosphaeria sp.	1	1	2					4	11.4	1	4	1					6	17.1	14.3
Phomopsis sp. 1	1	1						2	5.7	1	1						2	5.7	5.7
Volutella sp. 1	2	2	5	5	5	2		21	60	1	3	3	4	5	2		18	51.4	55.7
Cylindrocladium floridanum		3	1	1	2	1		8	22.9		3	4	4	2	2		15	42.9	32.9
Fusarium sp. 1		1						1	2.9									0	1.4
Gliocladium sp. 1		1						1	2.9		2						2	5.7	4.3
Gliocladium sp. 2		2						2	5.7		3						3	8.6	7.1
Hyponectria sp. 1		2	2	3	4	5	5	21	60		1	4	4	4	5	5	23	65.7	62.9
Hyponectria sp. 3		1						1	2.9		1						1	2.9	2.9
Bionectria ochroleuca			2	2	2	3		9	25.7				2	2	5	1	10	28.6	27.1
Fusarium sp. 2									0			1					1	2.9	1.4
Gliocladium sp. 3			3	4	5	1		13	37.1			5	4	5			14	40	38.6
Ijuhya parilis			1					1	2.9									0	1.4
Phaeosphaeria sp.			1	1	1	3		6	17.1			2					2	5.7	11.4

Table 1. (*Continued*)

Taxa	*Upper leaf surface touching the forest floor*									*Lower leaf surface touching the forest floor*									*Overall percentage occurrence*
	Day							*TL*	*%*	*Day*							*TL*	*%*	
	0	*4*	*8*	*16*	*24*	*40*	*56*			*0*	*4*	*8*	*16*	*24*	*40*	*56*			
Corynespora cassiicola			1	2	2	2	5	12	34.3			1	2	2	2	5	12	34.3	34.3
Albonectria rigidiuscula				1				1	2.9									0	1.4
Dactylaria longidentata				1	3			4	11.4				2	3			5	14.3	12.9
Hyponectria sp. 2					1			1	2.9									0	1.4
Anthostomella tenacis						2		2	5.7									0	2.9
Colletotrichum gloeosporioides						1	1	2	5.7						1	2	3	8.6	7.1
Hypoxylon sp.						2	1	3	8.6									0	4.3
Pseudohalonectria suthepensis						1	3	4	11.4						1	1	2	5.7	8.6

*TL: Total number of leaves

and those fungi on leaves with their lower surface touching the forest floor. The overall most common species were *Hyponectria* sp. 1 (62.9%), *Volutella* sp. 1 (55.7%), *Gliocladium* sp. 3 (38.6%), *Corynespora cassiicola* (34.3%), and *Cylindrocladium floridanum* (32.9%).

Effect of upper or lower leaf surface touching the forest floor on fungal communities

Twenty-two taxa were identified on leaves with their upper surface touching the forest floor during the decay process. This comprised 12 ascomycetes and 10 asexual taxa (9 hyphomycetes and 1 coelomycete). The dominant species were *Hyponectria* sp. 1 (60%), *Volutella* sp. 1 (60%), *Gliocladium* sp. 3 (37.1%), *Corynespora cassiicola* (34.3%), *Bionectria ochroleuca* (25.7%), *Cylindrocladium floridanum* (22.8%), *Phaeosphaeria* sp. (17.1%), *Dactylaria longidentata* (11.4%) and *Lasiosphaeria* sp. (11.4%) (Table 1).

Seventeen fungi were found on leaves with their lower surface touching the forest floor throughout the decay process. This comprised seven ascomycetes and 10 asexual taxa (nine hyphomycetes and one coelomycete). The dominant species were *Hyponectria* sp. 1 (65.7%), *Volutella* sp. 1 (51.4%), *Cylindrocladium floridanum* (42.9%), *Gliocladium* sp. 3 (40%), *Corynespora cassiicola* (34.3%), *Bionectria ochroleuca* (28.6%), *Leptosphaeria* sp. (17.1%) and *Dactylaria longidentata* (14.3%) (Table 1).

Three-dimensional correspondence analysis of fungal communities on senescent leaves of *M. liliifera* showed that the surface of leaves touching the forest floor had no effect on the fungal communities (Fig. 2).

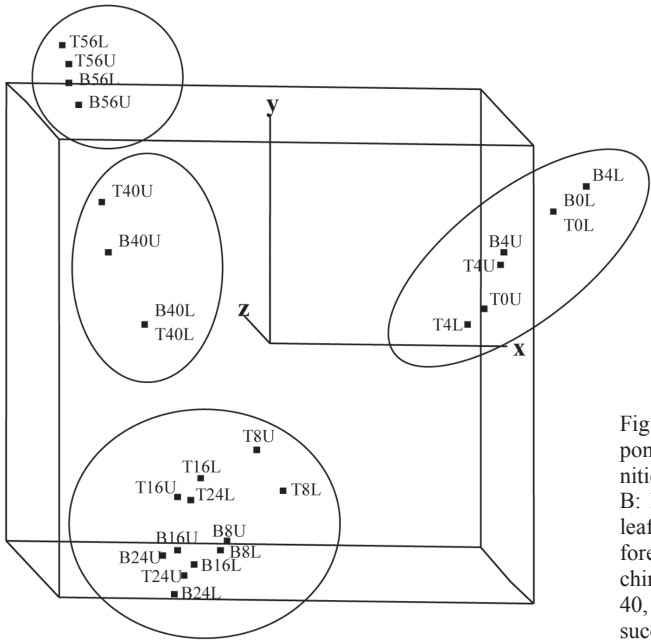

Fig. 2. Three-dimensional correspondence analysis of fungal communities on leaves of *Magnolia liliifera*. B: lower side of leaf. T: top side of leaf. L: lower surface touching the forest floor. U: upper surface touching the forest floor. 0, 4, 8, 16, 24, 40, 56: sampling times and stages of succession (days).

Succession of fungi

Three-dimensional correspondence analysis of fungal communities on senescent leaves of *M. liliifera* showed that there were at least three succession communities, the pioneer community (day 0-4), the mature community (day 8-40) and the impoverished community (day 56 onwards) (Fig. 2). The overall numbers of fungi found at different stages of leaf decay are given in Fig. 3.

The succession patterns of dominant fungi throughout the period of leaf decomposition are shown in Fig. 4. The fungal community composition was distinct at each stage of succession. In the pioneer community stage, fungal communities were low in number and had a low percentage occurrence. The dominant species at this stage was *Volutella* sp. 1. The highest species diversity was present during the mature community stage and the dominant species were *Bionectria ochroleuca*, *Cylindrocladium floridanum*, *Dactylaria longidentata*, *Gliocladium* sp. 3, *Hyponectria* sp. 1, and *Leptosphaeria* sp. In the impoverished community stage, the species diversity and number of species declined. The community was dominated by a few species with relatively high percentage occurrence. Dominant species were *Corynespora cassiicola* and *Hyponectria* sp. 1.

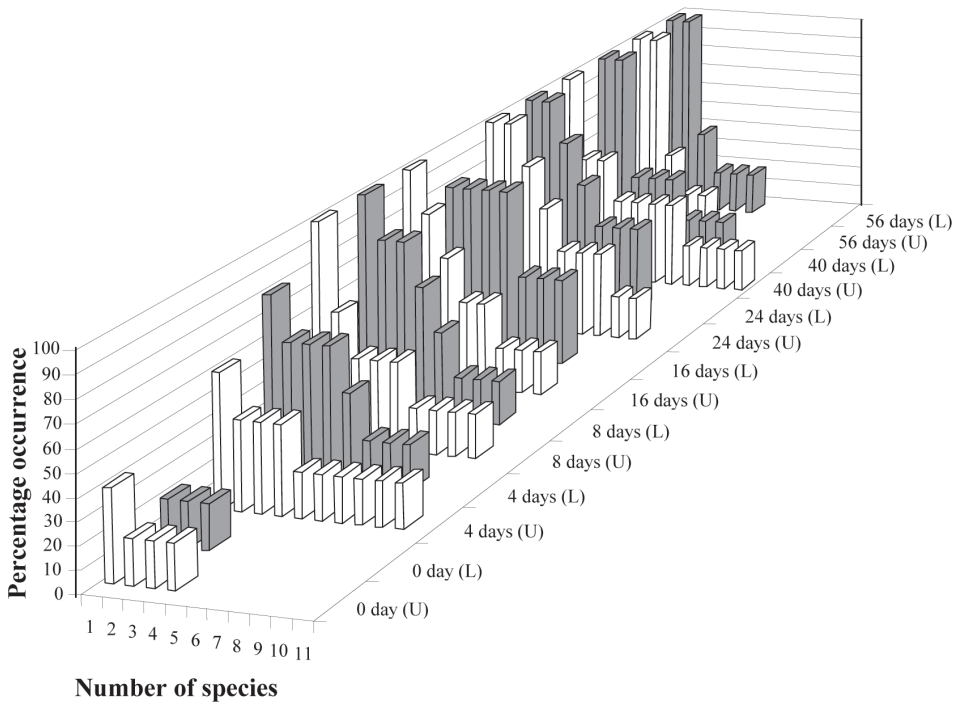

Fig. 3. Number of fungal species occurring at different sampling times. Species in each sampling time are ordered with most abundant on the left to the least abundant on the right. **L**: lower leaf surface touching the forest floor. **U**: upper leaf surface touching the forest floor.

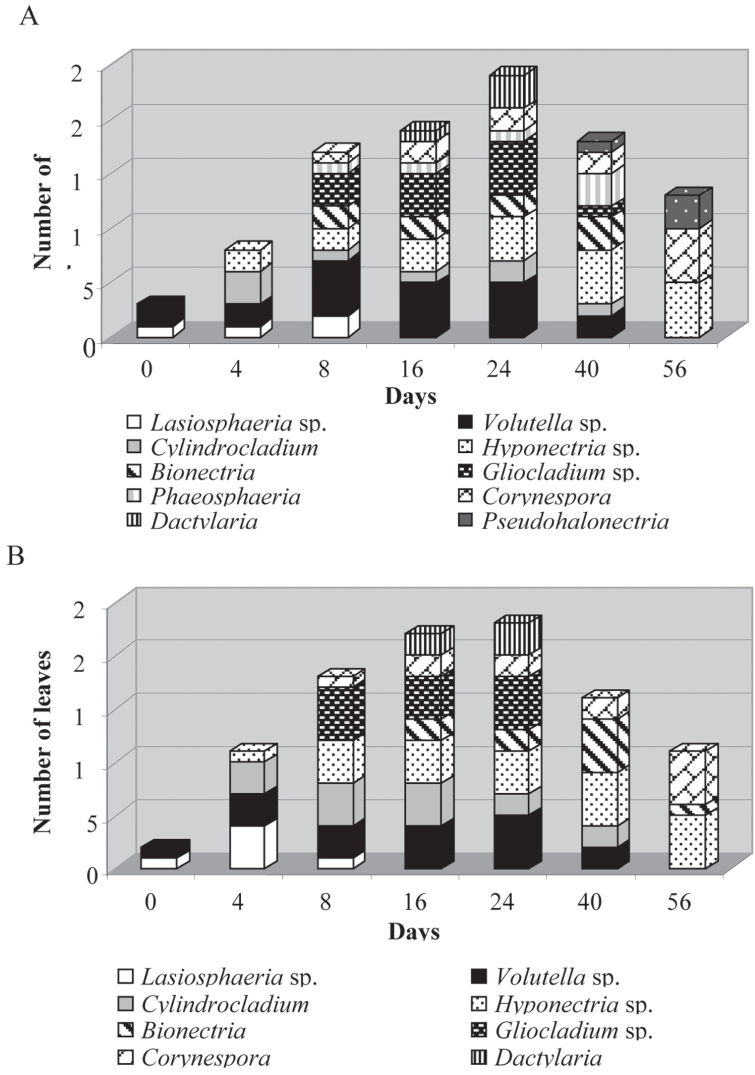

Fig. 4. Succession pattern of dominant fungi on leaves of *Magnolia liliifera* throughout the experimental period. A: upper surface touching the forest floor. B: lower surface touching the forest floor.

Fungal saprobes on naturally occurring dead leaves during the succession period

The taxa recorded on naturally occurring dead leaves of *M. liliifera* are listed in Table 2 with their percentage occurrence. Forty fungal taxa were identified, comprising 21 ascomycetes and 19 asexual fungi (13 hyphomycetes and 6 coelomycetes). Dominant species, found on more than 10% of samples were *Bionectria ochroleuca*, *Corynespora cassiicola*, *Cylindrocladium floridanum*, *Dokmaia monthadangii*, *Gliocladium* sp. 1, *Hyponectria* sp. 1, *Hyponectria* sp. 2, *Hypoxylon* sp., *Leptosphaeria* sp. and *Pseudohalonectria suthepensis* (Table 3).

Table 2. Percentage occurrence of fungal taxa on 90 naturally occurring dead leaves of *Magnolia liliifera*

Fungus	*Number of leaves*	*Percentage occurrence*
Corynespora cassiicola	47	52.2
Hyponectria sp. 1	37	41.1
Gliocladium sp. 1	19	21.1
Cylindrocladium floridanum	16	17.8
Leptosphaeria sp.	16	17.8
Pseudohalonectria suthepensis	14	15.6
Hyponectria sp. 2	13	14.4
Hypoxylon sp.	13	14.4
Bionectria ochroleuca	11	12.2
Dokmaia monthadangii	10	11.1
Volutella sp. 1	8	8.9
Nectria sp. 3	7	7.8
Colletotrichum sp. 2	6	6.7
Hyponectria sp. 3	6	6.7
Phaeosphaeria sp.	6	6.7
Nectria haematococca	5	5.6
Periconia jabalpurensis	5	5.6
Anthostomella monthadoia	4	4.4
Colletotrichum sp. 1	4	4.4
Dactylaria longidentata	4	4.4
Fusarium sp. 1	3	3.3
Stachybotrys parvispora	3	3.3
Anthostomella tenacis	2	2.2
Dactylaria dimorphospora	2	2.2
Gliocladium sp. 2	2	2.2
Albonectria rigidiuscula	1	1.1
Bionectria palmicola	1	1.1
Bionectria sp.	1	1.1
Colletotrichum gloeosporioides	1	1.1
Fusarium sp. 2	1	1.1
Gliocladium sp. 3	1	1.1
Guignardia mangiferae	1	1.1
Phyllosticta capitaliensis	1	1.1
Ijuhya parilis	1	1.1
Munkovalsaria appendiculata	1	1.1
Phomopsis sp. 1	1	1.1
Phomopsis sp. 2	1	1.1
Phomopsis sp. 3	1	1.1
Physalospora sp.	1	1.1
Rhytisma sp.	1	1.1

Table 3. Dominant taxa and percentage occurrence on decaying leaf baits (succession of fungi) and naturally decaying samples (Promputtha *et al*., 2004) of *Magnolia liliifera* (upper and lower leaf combined)

Leaf baits	*Naturally decaying leaves*
Bionectria ochroleuca (27.1%)	*Bionectria ochroleuca* (12.2%)
Corynespora cassiicola (34.3%)	*Corynespora cassiicola* (52.2%)
Cylindrocladium floridanum (32.9%)	*Cylindrocladium floridanum* (17.8%)
Dactylaria longidentata (12.9%)	*Dokmaia monthadangii* (11.1%)
Gliocladium sp. 3 (38.6%)	*Gliocladium* sp. 1 (21.1%)
Hyponectria sp. 1 (62.9%)	*Hyponectria* sp. 1 (41.1%)
Leptosphaeria sp. (14.3%)	*Hyponectria* sp. 2 (14.4%)
Phaeosphaeria sp. (11.4%)	*Hypoxylon* sp. (14.4%)
Volutella sp. 1 (55.7%)	*Leptosphaeria* sp. (17.8%)
	Pseudohalonectria sethepensis (15.6%)

DISCUSSION

Fungal diversity

The highest fungal diversity on leaves of *Magnolia liliifera* occurred between day 4 and 40, with most species being present on day 40 (Fig. 3). On day 56 leaves were found to be skeletonized, and the fungal communities had decreased in number.

The time for fungal communities to reach a peak of species diversity, or fungal activity varies between different studies, depending on the plant substrate. For example, complete decomposition of sugarcane bagasse needed 20 weeks and maximum colony counts were recorded during weeks 6-13 (Sandhu & Sidhu, 1980). In a 2-year study of the decomposition of leaf and root litter of pineapple, the number of species, the number of viable propagules associated with the litter and the rate of weight loss was maximum during the middle phase of the litter decomposition (Tiwari *et al.*, 1994). During one year of complete decomposition process of fronds of *Phoenix hanceeana*, species diversity also peaked at the middle stage of decomposition, 120 days for leaves, 150 days for rachis tips and 200 days for mid-rachides and rachis-bases (Yanna *et al.*, 2002).

The number of fungi on *M. liliifera* leaves was moderately high when compared with other studies using direct examination. Parungao *et al.* (2002) examined ten leaves from different tree species in rainforests in northern Queensland, Australia and found 0-14 taxa on each leaf species. Photita *et al.* (2001) examined the large leaves of *Musa acuminata* in Hong Kong and found 20 taxa on the leaves at Nim Shue Wan and 18 taxa on the leaves in Fung Yuen. In Thailand, Duong *et al.* (2008) investigated 60 senescent leaves of *Castanopsis diversifolia* in Doi Suthep-Pui National Park and found 55 taxa during the 4-month incubation period. Photita *et al.* (2003) recorded between 17 and 27 taxa on *Musa acuminata* leaves at various sites. Polishook *et al.* (1996) identified between 8 and 15 species from leaves of *Manilkara bidentata* and 9 and 11 species from leaves of *Guarea guidonia* in Puerto Rico. Nine taxa occurred on decomposing leaves, two species on freshly fallen

leaves and four species on senescent leaves of *Fagus crenata* (Osono, 2002). Forty-one taxa were found on leaves of the palm *Nypa fruticans* (Hyde & Alias, 2000).

Appreciably greater numbers of fungi have been identified using indirect methods (pulverization-washing-plating of particle suspensions) to detect fungal diversity in litter (e.g. Polishook *et al.*, 1996; Paulus *et al.*, 2003), however, this method will also detect dormant spores and may therefore not be a good reflection of the fungi involved in decaying leaves. Previous studies have not used molecular data to resolve the taxa and therefore names may be wrong. It will be interesting to establish if the species identified from different hosts using morphology are actually the same taxa when resolved by molecular data.

Effect of leaf surface touching the forest floor

Fungal communities on leaves of *Magnolia liliifera* with their upper surface touching the forest floor were similar to those on leaves with their lower surface touching the forest floor. The dominant species and time of their occurrence were similar (percentage similarity 77%). *Corynespora cassiicola*, *Cylindrocladium floridanum*, *Hyponectria* sp. 1, and *Volutella* sp. 1 were common on leaves with either their upper or lower surface touching the forest floor. When leaves fall from a tree they can land with either surface touching the ground and there would appear to be little apparent advantage for any fungus to restrict itself to either surface. While the surface of leaves touching the ground had no significant effect on fungal communities developing on baited decaying leaves of *Magnolia liliifera*, studies of other leaves would suggest that fungal colonies are often either restricted to a particular surface, or more common on one surface than the other. Plant pathogenic fungi often more commonly fruit on one surface than on the other.

Fungal communities on baits versus those on naturally decaying samples

Examination of fungi on naturally occurring samples is likely to result in finding fungi that only occur during certain stages of decay. For example, fungal communities on naturally occurring decaying leaves in this study were similar to those found on mature community stage leaf baits (Table 2). Fungal communities which change rapidly, or taxa that are present for a relatively short time, may easily be missed on collections of naturally occurring samples, especially those that appear only during the pioneer or impoverished stages. For example, *Gliocladium* sp. 2 on leaves of *Magnolia liliifera* was dominant (occurrence up to 60%) for a short period only during the early succession study. It was not dominant on the naturally occurring sample. In addition, *Dokmaia monthadangii* on *Magnolia liliifera* was dominant (occurrence up to 11%) on naturally occurring leaves, but was not found on the leaf baits (Table 3).

Factors supporting higher diversity

The reasons for higher diversity of fungi in this study compared to some previous studies are unclear. Large leaves, with a greater surface area may provide more substrata for fungal growth and thus support a larger number of species (Promputtha *et al.*, 2002). Photita *et al.* (2001) reported that the large leaves of *Musa acuminata* supported a higher diversity, than the smaller leaves of other tree species. The three tree species studied had different sized leaves. Forty fungal taxa were

recorded from leaves of *M. liliifera* (leaves large, 18-30 × 8-12 cm). In this study, leaf size was not a factor that affected fungal diversity. Leaf biochemistry, such as chemical content, lignocellulose or pH may be factors that lead to a higher species diversity (Promputtha *et al.*, 2002). More slowly decaying leaves may provide substrata over a longer period allowing the development of more fungal species.

Fungal succession

There have been several studies of the fungal succession on various materials, including submerged wood in fresh water (Ho *et al.*, 2002; Kane *et al.*, 2002; Sivichai *et al.*, 2002), leaf litter (Osono, 2002; Promputtha *et al.*, 2002; Yanna *et al.*, 2002), pods of *Dolenix regia* (Somrithipol *et al.*, 2002), rust-infected litter (McKenzie & Hudson, 1976), and dung (Richardson, 2001; 2002). These studies have established similar patterns in occurrence with time there being early, intermediate or late colonizers (Jones & Hyde, 2002). Changes of species composition throughout the decay process have been observed and classified by several authors (Dix & Webster, 1985; Promputtha *et al.*, 2002; Yanna *et al.*, 2002; Zhou & Hyde, 2002). In the present study, the fungal communities grouped into three succession stages, the pioneer stage, mature stage and impoverished stage (Dix & Webster, 1985; Promputtha *et al.*, 2002; Yanna *et al.*, 2002). Leung (1998) divided fungi from bamboo into two groups, early colonizers and regular inhabitants, while Zhou & Hyde (2002) divided fungi identified on bamboo into five groups, early colonizers, middle stage colonizers, later colonizers, regular inhabitants and sporadic inhabitants.

The fungal communities on leaves of *Magnolia liliifera* during succession period grouped into three succession stages, (1) pioneer communities, those fungi occurring during day 0-4 and disappearing thereafter, (2) mature communities, those fungi occurring during an intermediate period of decay and disappearing thereafter, and (3) impoverished communities, those fungi occurring in the latter stages of decay and present until the end of the study.

Decomposition rate of substrate

The time taken for decomposition of leaf litter varies enormously. For instance, in cool temperate pine forest, it may take 10 years to full decomposition of pine needles, and one year for decomposition of leaves in ash and sycamore woods. In a tropical forest leaves may decompose in only a few weeks (Hudson, 1980). The time taken for decomposition of monocotyledons in tropical regions is relatively short, e.g. 14 months for leaves of sugarcane (Hudson, 1962), 19 months for stems of couch grass (Hudson & Webster, 1958), two years for litter of pineapple (Tiwari *et al.*, 1994), one year for leaves and rachis-tips of *Phoenix hanceana* and 18 months for mid-rachides and rachis-bases of *Phoenix hanceana* (Yanna *et al.*, 2001). The decomposition of senescent leaves of *Magnolia liliifera* and of *Meliosma simplicifolia* and *Berchemia floribunda* (results not reported) were rapid, leaves were completely decayed within two months, more than 72 days and within 24 days, respectively.

Valuable methods to examine fungal succession

Changes in species colonization during decay were observed when leaf baits were periodically retrieved from the forest and examined directly after

incubation in a moist chamber. This method is most widely used in succession studies of plant litter (Webster, 1957; Srivastava *et al.*, 1983; Hyde, 1991). Other useful methods of study include culture plating (Pugh, 1958; Srivastava *et al.*, 1983), leaf disc washing (Sandhu & Sidhu, 1980) and particle filtration (Paulus *et al.*, 2006). Succession patterns can be confirmed by direct and particle filtration methods. Leaf disc washing excludes fungi that grow slowly or cannot grow at all on agar plates. It also encourages fast growing, ubiquitous species such as *Penicillium*, and the results may be unrepresentative (Lee & Hyde, 2002). Employing a raft of different methods may give a more accurate result of fungal succession (Yanna *et al.*, 2001).

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REFERENCES

CHOMNUNTI P., HONGSANAN S., AGUIRRE-HUDSON B., TIAN Q., PERŠOH D., DHAMI M.K., ALIAS A.S., XU J., LIU X., STADLER M. & HYDE K.D., 2014 — The sooty moulds. *Fungal Diversity* 66: 1-36.

DIX N.J. & WEBSTER J., 1985 — Fungal Ecology. Chapman and Hall, London.

DUONG L.M., MCKENZIE E.H.C., LUMYONG, S. & HYDE, K.D., 2008 — Fungal succession on senescent leaves of *Castanopsis diversifolia* in Doi Suthep-Pui National Park, Thailand. *Fungal Diversity* 30: 23-36.

GESSNER M.O., THOMAS M., JEAN-LOUIS A.M. & CHAUVET E., 1993 — Stable successional patterns of aquatic hyphomycetes on leaves decaying in a summer cool stream. *Mycological Research* 97: 163-172.

GHAWANA V.K., SHRIVASTAVA J.N. & KUSHWAHA R.K.S., 1997 — Some observations on fungal succession during decomposition of wool in soil. *Mycoscience* 38: 79-81.

GORSKA B., 1982 — Changes of microfungi during decomposition of plant material in two different woodland associations. Prace Naukowe Uniwersytetu Slaskiego Katowicach, 99-119.

HANDA T. & HARADA Y., 2005 — Succession of microfungal flora on *Rodgersia podophylla* plants at the forest side of *Cryptomeria* plantation. *Mycoscience* 46: 129-136.

HERING T.F., 1965 — Succession of fungi in the litter of a Lake District oakwood. *Transactions of the British Mycological Society* 48: 391-408.

HO W.H., YANNA HYDE KD. & HODGKISS I.J., 2002 — Seasonality and sequential of fungi on wood submerged in Tai Po Kau Forest Stream. Hong Kong. *Fungal Diversity* 10: 21-43.

HUDSON H.J., 1962 — Succession of micro-fungi on aging leaves of *Saccharum officinarum. Transactions of the British Mycological Society* 45: 395-423.

HUDSON H.J., 1980 — *Fungal Saprophytism*. The Camelot Press, London.

HUDSON H.J. & WEBSTER J., 1958 — Succession of fungi on decaying stems of *Agropyron repens. Transactions of the British Mycological Society* 41: 165-177.

HYDE K.D., 1991 — Fungal colonization of *Rhizophora apiculata* and *Xylocarpus granatum* poles in Kampong Kapok mangrove, Brunei. *Sydowia* 43: 31-38.

HYDE K.D. & ALIAS S.A., 2000 — Biodiversity and distribution of fungi associated with decomposing *Nypa fruticans*. *Biodiversity Conservation* 9: 393-402.

JONES E.B.G. & HYDE K.D., 2002 — Succession: where do we go from here? *Fungal Diversity* 10: 241-253.

KANE D.F., TAM W.Y. & JONES E.B.G., 2002 — Fungi colonising and sporulating on submerged wood in the River Severn, UK. *Fungal Diversity* 10: 45-55.

KODSUEB R., LUMYONG S., MCKENZIE E.H.C., BAHKALI A.H. & HYDE K.D., 2015 — Relationships between terrestrial and freshwater lignicolous fungi. *Fungal Ecology* 19: 155-168.

LEE O.H.K. & HYDE K.D., 2002 — Phylloplane fungi in Hong Kong mangroves: evaluation of study methods. *Mycologia* 94: 596-606.

LEUNG S.S., 1998 — A study of saprophytic fungi associated with bamboo culms in terrestrial, freshwater and marine habitats, Ph.D. Thesis, The City University of Hong Kong, Hong Kong.

MCKENZIE E.H.C. & HUDSON H.J., 1976 — Mycoflora of rust-infected and non-infected plant material during decay. *Transactions of the British Mycological Society* 66: 223-238.

MCMULLAN-FISHER S.J.M., MAY T.W. & KEANE P.J., 2002 — The macrofungal community and fire in a Mountain Ash forest in southern Australia. *Fungal Diversity* 10: 57-76.

OSONO T., 2002 — Phyllosphere fungi on leaf litter of *Fagus crenata*: occurrence, colonization, and succession. *Canadian Journal of Botany* 80: 460-469.

PARUNGAO M.M., FRYAR S.C. & HYDE K.D., 2002 — Diversity of fungi on rainforest litter in north Queensland, Australia. *Biodiversity Conservation* 11: 1185-1194.

PAULUS B., GADEK P. & HYDE, K.D., 2003 — Estimation of microfungal diversity in tropical rainforest leaf litter using particle filtration: the effects of leaf storage and surface treatment. *Mycological Research* 107: 748-756.

PAULUS B., GADEK P. & HYDE K.D., 2006 — Successional patterns of microfungi in fallen leaves of *Ficus pleurocarpa (Moraceae)* in an Australian tropical rainforest. *Biotropica* 38: 42-51.

PHOTITA W., LUMYONG S., LUMYONG P., HO W.H., MCKENZIE E.H.C. & HYDE K.D., 2001 — Fungi on *Musa acuminata* in Hong Kong. *Fungal Diversity* 6: 99-106.

PHOTITA W., LUMYONG S., LUMYONG P., MCKENZIE E.H.C. & HYDE K.D., 2003 — Saprobic fungi on dead wild banana. *Mycotaxon* 85: 345-356.

POLISHOOK J.D., BILLS G.F. & LODGE D.J., 1996 — Microfungi from decaying leaves of two rain forest trees in Puerto Rico. *Journal of Industrial Microbiology & Biotechnology* 17: 284-294.

PROMPUTTHA I., LUMYONG S., LUMYONG P., MCKENZIE E.H.C. & HYDE K.D., 2002 — Fungal succession on senescent leaves of *Manglietia garrettii* in Doi Suthep-Pui National park, northern Thailand. *Fungal Diversity* 10, 89-100.

PROMPUTTHA I., LUMYONG S., LUMYONG P., MCKENZIE E.H.C. & HYDE, K.D., 2004 — Fungal saprobes on dead leaves of *Magnolia liliifera* (Magnoliaceae) in Thailand. *Cryptogamie Mycologie* 25: 315-321.

PUGH G.J.F., 1958 — Leaf litter fungi found on *Carex paniculata* L. *Transactions of the British Mycological Society* 41: 185-195.

RICHARDSON M.J., 2002 — The *coprophilous* succession. *Fungal Diversity* 10: 101-111.

RICHARDSON M.S., 2001 — Diversity occurrence of *coprophilous* fungi. *Mycological Research* 105: 387-402.

SANDHU D.K. & SIDHU M.S., 1980 — Fungal succession on decomposing sugarcane bagasse. *Transactions of the British Mycological Society* 75: 281-286.

SIVICHAI S., JONES E.B.G. & HYWEL-JONES N.L., 2002 — Fungal colonisation of wood in a fresh water stream at Tad Ta Phu, Khao Yai National Park, Thailand. *Fungal Diversity* 10: 113-129.

SOMRITHIPOL S., JONES E.B.G. & HYWEL-JONES N.L., 2002 — Fungal diversity and succession on pods of *Delonix regia* (*Leguminosae*) exposed in a tropical forest in Thailand. *Fungal Diversity* 10: 131-139.

SRIVASTAVA S.K., RAIZADA B.B.S. & GUPTA S.C., 1983 — Succession of microfungi on decaying stems of *Triticum aestivum.* Proceedings of National Academy of Science India Section B (Biological Sciences) 53: 231-246.

SUZUKI A., UCHIDA M. & KITA Y., 2002 — Experimental analyses of successive occurrence of ammonia fungi in the field. *Fungal Diversity* 10: 141-165.

TIWARI S.C., TIWARI B.K. & MISHRA R.R., 1994 — Succession of microfungi associated with the decomposing litters of pineapple (*Ananas comosus*). *Pedobiologia* 38: 185-192.

TOKUMASU S. & AOIKI T., 2002 — A new approach to studying microfungal succession on decaying pine needles in an oceanic subtropical region in Japan. *Fungal Diversity*. 10: 167-183.

TRIBE H.T., 1957 — Ecology of micro-organisms in soils as observed during their development upon buried cellulose film, in: Williams, R.E.O., Spicer, C.C. (Eds.). *Microbial Ecology*. 287-298.

TRIBE H.T., 1961 — Microbiology of cellulose decomposition in soil. *Soil Science* 92: 61-77.

WEBSTER J., 1957 — Succession of fungi on decaying cocksfoot culms. T*he Journal of Ecology* 45: 1-30.

YANNA HO W.H. & HYDE K.D., 2002 — Fungal succession on fronds of *Phoenix hanceana* in Kong Kong. *Fungal Diversity*. 10: 185-211.

YANNA HO W.H., HYDE K.D. & GOH T.K., 2001 — Occurrence of fungi on tissues of *Livistona chinensis*. *Fungal Diversity* 6: 167-180.

ZHOU D.Q. & HYDE K.D., 2002 — Fungal succession on bamboo in Kong Kong. *Fungal Diversity* 10: 213-227.