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Fungicolous xylariaceous fungi in coralloid basidiomata

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ABSTRACT

Fifteen species of xylariaceous fungi were isolated from more than 100 *Scytinopogon* sp. basidiomata in three forests and identified by rDNA ITS sequencing. Xylariaceous fungi were present in the lower part of the basidiomata, occurred less often in the upper part and were absent in the tips of its coralloid basidiomata. These results indicated that xylariaceous fungi grow continuously in the basidiomata and that the apical growth of *Scytinopogon* sp. basidiomata was faster than the growth of xylariaceous fungi in its host. The results suggested that these xylariaceous fungi established a stable coexistence with *Scytinopogon* sp. in the forest. Five species of xylariaceous fungi were endophytes in nearby plants. Our study suggested that these endophytic xylariaceous fungi were sources of fungicolous fungi in *Scytinopogon* sp. in the forest.

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1. Introduction

Fungicolous fungi represent species that regularly accompany other fungi (Jeffries, 1995). This term can be applied to any inter-fungal relationship, including parasites, commensals or saprobionts (Kirk et al., 2008). Most studies report fungicolous fungi growing on sporocarps of other fungi, but not inside them. Only two reports have described the presence of basidiomycete hyphae in the ascocarp of truffles (Ceruti, 1988; Pacioni et al., 2007), and one study isolated *Cryptococcus victoriae* from the inner tissue of *Paxillus* (Yurkov et al., 2012). These fungi are present in healthy fruit bodies, suggesting that they may play significant roles by interacting with the developing host sporocarp.

Recently, most research on fungicolous fungi has focused on biochemical innovation (Shim et al., 2011; Hwang et al., 2015) and the interactions between fungicolous fungi and their host (He et al., 2006; Pacioni et al., 2007). For example, some mycelia have been observed in the sections of truffle ascomata, and they grow attached to the hyphal wall of the mycelium of *Tuber borchii* when in dual culture (Pacioni et al., 2007). Most of the fungicolous fungi reported were microfungi. Few studies have investigated the diversity of a coexisting fungicolous fungi in a host species in forest ecosystems nor the source of these fungicolous fungi. In this study, we examined the association of a coralloid basidiomata and ascomycetous xylariaceous fungi and explored the above questions.

2. Materials and methods

2.1. Study site

The study was conducted in three separate areas: two located in central Taiwan and one in northern Taiwan. The Zen-Len area spans the region from 23°28′ N to 23°55′ N latitude and from 120°48′ E to 121°09′ E longitude. The elevation ranges between 1300–1500 m. The average annual temperature and rainfall are 15.78 °C and 2628 mm, respectively. Most rainfall at this site occurs during the summer, from June to September. Some rainfall also occurs during the spring, from March to May; there is no obvious dry season. The main vegetation in this area is Japanese cedar, *Cryptomeria japonica*. The Lienhuachih area ranges from 500–900 m in altitude, and the main type of flora is the lauro-fagaceous forest (Su, 1984). The mean annual temperature and rainfall is 20.8 °C and 2285 mm with seasonality (Lu et al., 2008). More than half of the rain falls between May and September. Guanwu (121°07′E, 24°31′N) is a subtropical montane forest and ranges from 2000–2250 m in altitude. It is dominated by *C. japonica* with a few *Taiwania cryptomerioides*, *Chamaecyparis formosensis* and *Cunninghamia konishii*.

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2.2. Isolation of xylariaceous fungi

In our previous study, an undescribed *Scytinopogon* species was found to be widely distributed in Japanese cedar plantations of central Taiwan and exhibited a long fruiting season from April to October (Lin et al., 2015). *Scytinopogon* sp. basidiomata were collected from 2006 to 2012 and xylariaceous fungi were isolated from more than 100 *Scytinopogon* sp. sporocarps (data not shown). Mature and healthy basidiomata of *Scytinopogon* sp. were collected from the three study areas in 2010. Basidiomata were cut into 1 cm pieces. The process of sterilization was modified from the procedures of Guo et al. (2001). The sporocarp segments were surface sterilized with 1.05% sodium hypochlorite for 1 min and finally rinsed in sterile water for 45 s. The segments were dried with sterilized tissue paper and placed onto potato dextrose agar. When hyphae grew from the cross sections of the segments, the hyphal tip was isolated and purified on a potato dextrose agar (difco) plate. The cultures were incubated at room temperature.

Elatostema lineolatum, *Diplazium dilatatum* and *C. japonica* were dominant plants in the Zen-Len area. Fresh leaves of *E. lineolatum* and *D. dilatatum*, root, bark and dead leaves of *C. japonica*, and soil samples were collected from the Zen-Len area with *Scytinopogon* sp. basidiomata. Xylariaceous fungi from these samples were also isolated, as described above.

2.3. DNA isolation and ITS amplification

Mycelia of xylariaceous fungi were harvested from the plates and stored at −20 °C. Total DNA from the mycelium was extracted by the CTAB method (Doyle and Doyle, 1990). The PCR amplification of the rDNA ITS region was undertaken using the universal primers ITS5/ITS4 or primers ITS1/ITS4 (White et al., 1990). PCR products were directly sequenced in an ABI PRISM 3730 Genetic Analyzer (PE Applied Biosystems, Foster City, CA, USA).

2.4. Sequence analysis

Sequence data for the ITS regions were analyzed together with the outgroup *Sordaria fimicola* (GenBank accession no. AY681188). Moreover, several species of *Xylaria*, 2 species of *Eutypella*, and one species each of *Hypoxylon* and *Nemania* were used in ITS analysis. These species from GenBank were analyzed with the isolates in this study (Table 1). Sequence data for the ITS region were initially aligned and subsequent manual adjustments were made using the BioEdit Sequence Alignment Editor (Hall, 2011) and Clustal X 1.83 (Thompson et al., 1997). Using the same aligned datasets, parsimony analysis was performed with the default settings and parsimony bootstrap values were generated with 1000 replicate heuristic searches to estimate support for the clade stability of the consensus tree, with 1000 replicates in PAUP (Phylogenetic Analysis Using Parsimony) v4.0b10 (Swofford, 2003).

2.5. Detection and isolation of Xylaria spp. from Scytinopogon sp. basidiomata

To determine the distribution of *Xylaria*, 20 basidiomata of *Scytinopogon* sp. were cut into 1 cm segments from top to bottom and were individually placed into 1.5 ml microcentrifuge tubes. Ten basidiomata were analyzed by isolation and 10 underwent PCR detection. The isolation of *Xylaria* was performed as described previously (modified from Guo et al., 2001). The presence of xylariaceous fungi was recorded. Isolation rate was defined as the number of fragments that xylariaceous fungi isolated, divided by the total number of fragments isolated.

Xylaria-specific primers were designed for PCR detection. Sequences of *Xylaria* spp. from GenBank were aligned with the same regions of xylariaceous fungal isolates and related species using Clustal X 1.83 (Thompson et al., 1997). Primers were designed from the variable regions of the ITS. To examine the specificity of the primers, Basic Local Alignment Search Tool (BLAST) from the National Center for Biotechnology Information was used to search for species with sequences homologous to the primers from GenBank. One oligonucleotide primer pair Xf1 (5′-GGGACATTCTGGGATGGGACATCC-3′) and Xr2 (5′-ACACACAACACGGCCAGGGGAC-3′), which targeted the appropriate *Xylaria* spp. rDNA sequences, were designed, and these primers shared little or no homology with *Scytinopogon* sp. rDNA sequences. Based on the sequencing information, the primer pair was *Xylaria* genus-specific, and the predicted amplification size of the *Xylaria* spp. product was approximately 400–500 bp. Primers were synthesized by Mission Biotech (Taipei, Taiwan). DNA from specimens of 1 cm tissues was extracted by the CTAB method (Doyle and Doyle, 1990) and PCR amplification with *Xylaria*-specific primer pairs was used to detect the presence of *Xylaria*. Detection rate was defined as the number of fragments that *Xylaria* detected, divided by the total number of fragments detected.

3. Results

3.1. Isolation

A total of 53 xylariaceous fungal isolates were isolated from fresh, healthy and intact basidiomata of *Scytinopogon* sp (Table 2). Xylariaceous fungal hyphae grew from tissues of the *Scytinopogon* sp. sample on a PDA plate in 24–48 h (Fig. 1). Pure cultures of these fungi formed white colonies with radial hyphal strands and black pigmentation. After 2–4 weeks, stromata were produced in rays. A total of 7 xylariaceous isolates belonging to 5 *Xylaria* spp. were isolated from plants which located near to *Scytinopogon* sp. in plots. *Scytinopogon* sp. was unculturable on 7 types of culture media (data not shown).

3.2. Diversity of xylariaceous fungi in basidiomata of Scytinopogon sp. and other substrata

The internal transcribed spacer (ITS) rDNA sequences of approximately 600 bp were obtained from 53 xylariaceous fungal isolates. Twenty-two reference sequences representing 14 fungal species were obtained by BLAST (Table 1).

According to ecological studies of endophytic fungi (Arnold et al., 2009; Okane et al., 2012), 90–95% ITS sequence similarity is often used as a species boundary in fungi. The ITS-5.8S rDNA gene dataset contained 85 taxa with 754 characteristics. One hundred and ninety-two base pairs of ambiguous aligned regions were excluded from parsimony. The phylogenetic tree from the maximum parsimony analysis is shown in Fig. 2. The newly isolated xylariaceous fungi were clustered into four clades. Clade A included 5 isolates of *Xylaria* sp. with high branch support (MP = 100%). In Clade B, isolates ZLX7-3 and ZLX7-4 from *Scytinopogon* sp. basidiomata clustered with *Nemania bipapillata* with strong bootstrap values (MP = 100%) and ITS sequence similarity (93%). Clade C included two clusters; GWX-1 was identified as *Hypoxylon monticulosum* with 97% ITS sequence similarity; isolates ZLX7-1 and ZLX7-2 had 95% ITS similarity to *Eutypella* sp., and they were different from the reference sequence of *Eutypella cerviculate*. Clade D consisted of several species of *Xylaria* supported by strong bootstrap values, including *Xylaria* spp., *Xylaria apoda*, *Xylaria adscendens*, *Xylaria bambusicola*, *Xylaria curta*, *Xylaria feejeensis*, *Xylaria grammica*, *Xylaria laevis, Xylaria multiplex*, and *Xylaria papulis*.

Table 1
Fungal isolates recovered from basidiomata of *Scytinopogon* sp. in Taiwan. GenBank Accession number of the rDNA ITS sequence of isolates used in this study and the query coverage and max identity with their most closely related fungal ITS sequences in GenBank.

Isolates in this study	GenBank sequences			
	Blast result	Accession no.	Query cover	Max identity
[a]ZLX7-1	*Eutypella* sp.	JN637945	98%	99%
ZLX7-2	*Eutypella* sp.	FJ172283	98%	100%
GWX-1	*Hypoxylon monticulosum*	KJ774048	100%	99%
ZLX7-3	*Nemania bipapillata*	GU292818	99%	99%
ZLX7-4	*Nemania bipapillata*	GU292818	94%	97%
ZLX7-5	*Nemania* sp.	JX624281	94%	99%
ZLX10-1	*Xylaria adscendens*	GU322432	99%	93%
ZLX8-1	*Xylaria apoda*	GU322437	98%	99%
ZLX9-1	*Xylaria apoda*	GU322437	97%	99%
ZLX8-2	*Xylaria bambusicola*	JX256820	99%	96%
ZLX11-1	*Xylaria bambusicola*	EF026123	99%	97%
ZLX12-1	*Xylaria bambusicola*	EF026123	100%	96%
ZLX6-1	*Xylaria curta*	GU322444	99%	97%
ZLX6-2	*Xylaria curta*	GU322444	97%	99%
ZLX6-3	*Xylaria curta*	GU322444	97%	97%
ZLX6-4	*Xylaria curta*	GU322444	98%	99%
ZLX6-5	*Xylaria curta*	GU322444	98%	99%
ZLX7-6	*Xylaria curta*	JX256823	99%	97%
ZLX7-7	*Xylaria curta*	JX256823	99%	97%
ZLX7-8	*Xylaria curta*	JX256823	99%	97%
ZLX7-9	*Xylaria curta*	GU322444	100%	97%
ZLX7-10	*Xylaria curta*	GU322444	97%	97%
ZLX7-11	*Xylaria curta*	GU322444	97%	97%
ZLX7-12	*Xylaria curta*	GU322444	97%	97%
ZLX7-13	*Xylaria curta*	GU322444	99%	99%
ZLX9-2	*Xylaria curta*	GU322444	98%	98%
ZLX10-2	*Xylaria curta*	GU322444	98%	97%
ZLX10-3	*Xylaria curta*	GU322444	97%	97%
ZLX11-2	*Xylaria curta*	GU322444	97%	97%
ZLEX-1	*Xylaria curta*	GU322445	100%	99%
ZLEX-2	*Xylaria curta*	GU322444	100%	99%
ZLX10-4	*Xylaria feejeensis*	GU322453	97%	99%
ZLEX-3	*Xylaria feejeensis*	GU322454	100%	100%
ZLX10-5	*Xylaria grammica*	GU300097	100%	100%
ZLX10-6	*Xylaria grammica*	GU300097	100%	100%
ZLX10-7	*Xylaria grammica*	GU300097	100%	97%
ZLX7-14	*Xylaria grammica*	GU300097	99%	100%
ZLX7-15	*Xylaria grammica*	GU300097	95%	99%
ZLEX-4	*Xylaria grammica*	GU300097	100%	99%
LHCX-1	*Xylaria laevis*	GU324747	99%	99%
LHCX-2	*Xylaria laevis*	GU324747	96%	99%
LHCX-3	*Xylaria laevis*	GU324747	100%	99%
ZLX7-16	*Xylaria laevis*	GU324747	98%	99%
ZLDX-1	*Xylaria laevis*	GU324747	100%	99%
ZLDX-2	*Xylaria laevis*	GU324747	100%	99%
GWX-2	*Xylaria multiplex*	GU300099	99%	95%
ZLX6-6	*Xylaria multiplex*	GU300099	99%	95%
ZLX9-3	*Xylaria multiplex*	GU300099	98%	95%
ZLX9-4	*Xylaria multiplex*	GU300099	98%	95%
ZLX7-17	*Xylaria papulis*	GU300100	95%	99%
ZLEX-5	*Xylaria papulis*	GU300100	100%	99%
ZLX10-8	*Xylaria* sp.	HQ435666	98%	97%
ZLX10-9	*Xylaria* sp.	JX436805	98%	98%
ZLX10-10	*Xylaria* sp.	JX436805	99%	98%
ZLX6-7	*Xylaria* sp.	KM066560	94%	97%
ZLX7-18	*Xylaria* sp.	KM066560	93%	99%
ZLX7-19	*Xylaria* sp.	KM066560	95%	99%
ZLX7-20	*Xylaria* sp.	KM066560	95%	99%
ZLX8-3	*Xylaria* sp.	JX436805	100%	99%
ZLX8-4	*Xylaria* sp.	KC507251	99%	99%

[a] ZLX indicated that the isolate was isolated from sporocarp of *Scytinopogon* sp. in the Zen-Len area; GWX indicated that the isolate was isolated from sporocarp of *Scytinopogon* sp. in Guanwu; LHCX indicated that the isolate was isolated from sporocarp of *Scytinopogon* sp. in Lienhuachih; ZLEX indicated that the isolate was isolated from leaves of *E. lineolatum* in the Zen-Len area; ZLDX indicated that the isolate was isolated from leaves of *D. dilatatum* in the Zen-Len area.

Two *X. apoda* isolates, 3 *X. bambusicola* isolates, 6 *X. grammica* isolates, 2 *X. papulis* isolates, 5 *X. multiplex* isolates, 2 *X. feejeensis* isolates, 6 *X. laevis* isolates and 19 *X. curta* isolates were identified. Isolates ZLX10-1 were similar to *X. adscendens* with 85% ITS sequence similarity and were grouped with *X. adscendens* with 77% branch support. This species was similar to *X. adscendens*.

According to the BLAST results and phylogenetic analysis, 48 isolates of 14 species associated with *Scytinopogon* sp. basidiomata were found in the Zen-Len area, including *Eutypella* sp., *Nemania* sp., *N. bipapillata*, *X. adscendens*, *X. apoda*, *X. bambusicola*, *X. curta*, *X. feejeensis*, *X. grammica*, *X. laevis*, *X. multiplex*, *X. papulis*, and 2 *Xylaria* sp (Table 1). Isolates of *H. monticulosum* and *X. multiplex*

Table 2
Spatial distribution of xylariaceous fungi associated with *Scytinopogon* sp. basidiomata.

Species/isolates	Zen-Len (n = 44)	Guanwu (n = 3)	Lienhuachih (n = 4)	Total
Eutypella sp.	1/2			1/2
Hypoxylon monticulosum		1/1		1/1
Nemania bipapillata	1/2			1/2
Nemania sp.	1/1			1/1
Xylaria adscendens	1/1			1/1
Xylaria apoda	1/2			1/2
Xylaria bambusicola	1/3			1/3
Xylaria curta	1/17			1/17
Xylaria feejeensis	1/1			1/1
Xylaria grammica	1/5			1/5
Xylaria laevis	1/1		1/3	1/4
Xylaria multiplex	1/4	1/1		1/5
Xylaria papulis	1/1			1/1
Xylaria sp.	2/8			2/8
Total	14/48	2/2	1/3	15/53

were found in the Guanwu area; 3 isolates of *X. laevis* were found in the Lienhuachih area (Table 2). In the same area, *X. curta*, *X. feejeensis*, *X. papulis* and *X. grammica* were isolated from leaves of *E. lineolatum*, and *X. laevis* was isolated from leaves of *D. dilatatum* (Table 1). All *Xylaria* spp. isolated from plants were found in the basidiomata of *Scytinopogon* sp. No *Xylaria* spp. were isolated from 102 root, 42 bark and 42 dead leaf samples of *C. japonica* or from 45 soil samples.

3.3. Distribution of Xylaria spp. in Scytinopogon sp. basidiomata

The distribution of *Xylaria* in the basidiomata of *Scytinopogon* sp. was investigated by isolation and PCR detection from the sporocarp segments. Species of *Xylaria* were isolated or detected in 70% basidiomata (N = 20). Here we defined that tip, bottom and middle as the topmost fragment, basal fragment and the other fragments of the coralloid basidiomata, respectively. Both isolation rate and PCR detection rate were 10%, 36% and 70% in the tip, middle and bottom parts of the basidiomata (Table 3).

4. Discussion

We observed that xylariaceous fungal hyphae grew from cross sections of *Scytinopogon* basidiomata and confirmed xylariaceous fungi to be fungicolous fungi of the *Scytinopogon* basidiomata by PCR detection and isolation. Basidiomata of *Scytinopogon* sp. hosted 15 species of xylariaceous fungi. No xylariaceous species were obtained from the other 33 coralloid basidiomycetes isolated (data not shown). It seemed that these xylariaceous species established special associations with the *Scytinopogon* sp., and these associations were unique.

Most fungicolous fungi or mycophilic fungi grow on other fungi (Gams et al., 2004), but do not exist inside the host tissue. Only two reports described the presence of fungicolous hyphae in the ascomata of truffles (Ceruti, 1998; Pacioni et al., 2007). However, they did not discuss the distribution pattern of fungicolous fungi in the host and the environment. In this study, we used isolation and PCR to detect their distribution and found that fungicolous fungi existed in the lower part of the coralloid basidiomata of *Scytinopogon* sp. more frequently than in other parts. They coexisted stably in forests.

Xylariaceous fungi are common endophytes and saprotrophs, which inhabit conifers, monocots, dicots, and ferns (Brunner and Petrini, 1992; Jiang et al., 2011; Okane et al., 2012; Chen et al., 2013; Govinda Rajulu et al., 2013). Some xylariaceous fungi are associated with termite nests (Rogers et al., 2005; Ju and Hsieh, 2007); interestingly, all of the fungicolous *Xylaria* species found in this study were not. *Scytinopogon*'s fungicolous fungi *X. curta*, *X. laevis*, *X. feejeensis*, *X. papulis* and *X. grammica* were isolated from ferns and monocots near *Scytinopogon* basidiomata in the Zen-Len area. They were endophytic xylariaceous fungi. Endophytic xylariaceous fungi of nearby plants seemed to be the source of xylariaceous fungi in basidiomata of *Scytinopogon* sp. Xylariaceous fungi not only established endophytic associations with plants but also established a stable coexistence with *Scytinopogon* sp.

The mycoparasitism between fungicolous fungi and their hosts has been widely studied (Gams et al., 2004). Mycoparasites use penetration pegs to invade host hyphae (Deacon, 1976) or secrete antibiotics and lytic enzymes, such as chitinase and glucanase, on

Fig. 1. *Xylaria* hyphae grew from the cross-sections of *Scytinopogon* sp. basidiomata in (A) 24–48 h after isolation and (B) 3–4 d after isolation.

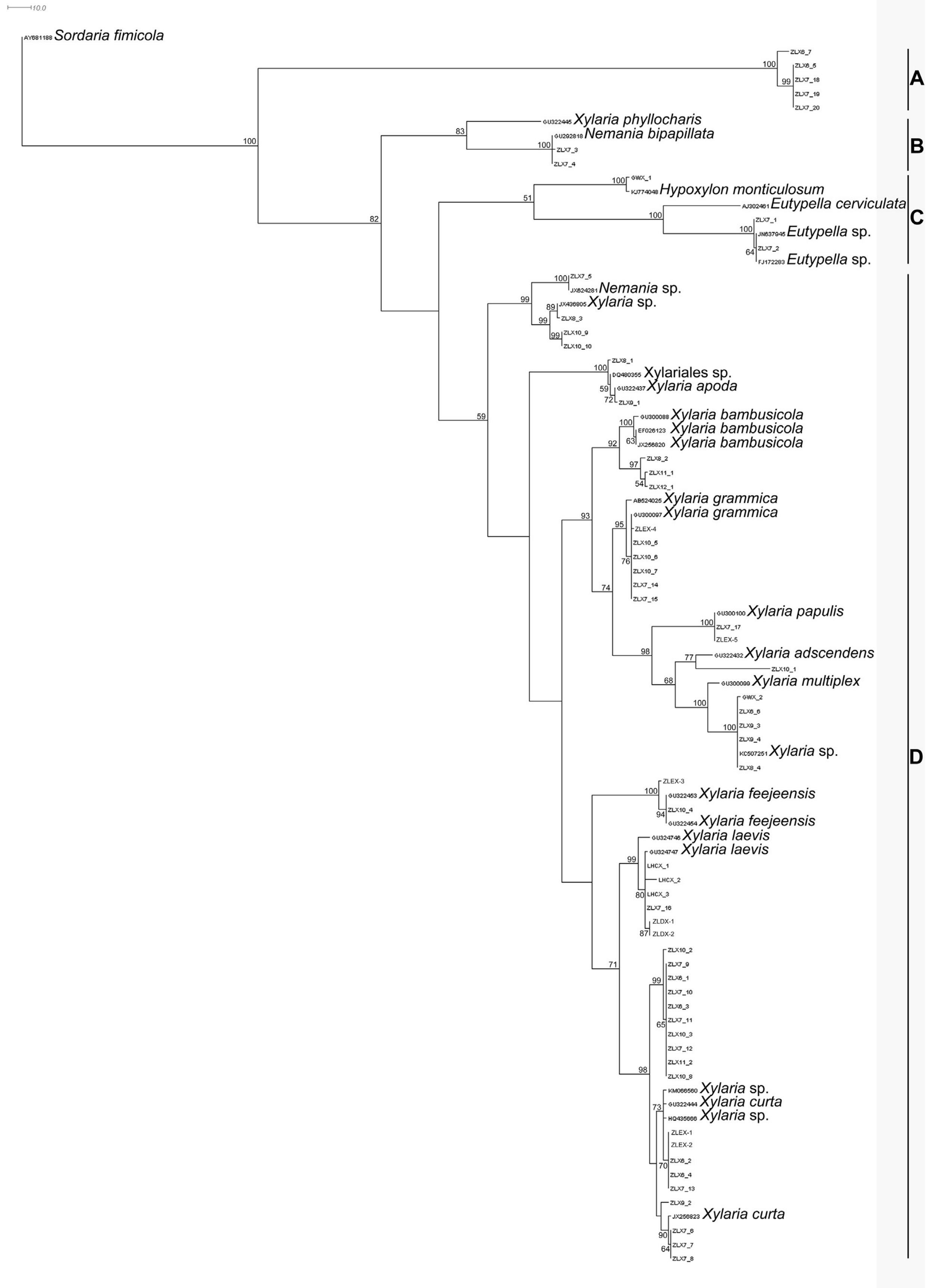

Fig. 2. The tree generated from maximum parsimony analysis based on the ITS-5.8S rDNA gene dataset. Bootstrap values greater than 50% are shown above or below branches.

Table 3
Xylaria spp. were cultured/detected by PCR with its genus-specific primers from *Scytinopogon* sp. sporocarp samples. Xylariaceous fungi were present in the bottoms and the middle parts of the basidiomata, but were not present in the tips.

Axis-based positions	Presence of xylariaceous fungi																			
	Samples for culture isolation										Samples for DNA detection									
	1	2	3	4	5	6	7	8	9	10	1	2	3	4	5	6	7	8	9	10
a			[a]–		–	–	–	–	–				–		–	–	–	–	–	
b		–	–	–	–	+	+	–	+	+		–	–	–	–	–	–	–	+	+
c	–	–	–	–	–	+	+	–	–	+	–	–	–	–	–	+	+	–	–	+
d	–	–	–	–	–	–	+	+	+	–	–	–	–	–	–	–	+	+	+	–
e	–	–	–	–	–	+	+	–	+	+	–	–	–	–	–	+	+	–	+	+
f	–	–	–	–	+	+	+	–	+	+	–	–	–	–	+	+	+	–	+	+
g	–	–	–	+	nd	nd	nd	+	+	+	–	–	–	+	+	+	+	+	+	+

+: positive result; –: negative result; nd: not detected; blank: no samples.
[a] Light gray panes represent the tips of the coralloid basidiomata; Dark gray panes represent the bottoms of the coralloid basidiomata; Other panes represent the middle parts of the coralloid basidiomata.

their hosts (Jeffries, 1995). Leaf endophytic xylariaceous fungi produce chitinases (Rajulu et al., 2011), and fungicolous xylariaceous fungi may produce chitinases as well. However, no obvious damage or symptom from on *Xylaria*-inhabited basidiomata was observed. According to our observations, the longevity between host and non-host basidiomata were not different. In addition, basidiomata with xylariaceous fungi and without xylariaceous fungi both produced spores normally. These guest xylariaceous fungi were not parasites.

Truffle-hosted fungi and *T. borchii* grew closely when co-cultured, and these truffle-hosted fungi might play a positive or protective role in the basidiomata (Pacioni et al., 2007). Members of endophytic Xylariaceae are known to produce various secondary compounds (Whalley and Edwards, 1995; Stadler and Hellwig, 2005; Richardson et al., 2014). These compounds exhibit antifungal and antibacterial properties (Daferner et al., 1999; Ratnaweera et al., 2014). Endophytes confer these compounds to their host, and then increase the fitness of the host plant (Sanchez-Azofeifa et al., 2012; Gundel et al., 2013). Basidiomata of *Scytinopogon* had a long lifespan and lasted for 5 months. These associated xylariaceous fungi might provide basidiomata compounds which resist invasion or decomposition from other microorganisms. Xylariaceous fungi in *Scytinopogon* basidiomata play similar roles as endophytic fungi in plants. The xylariaceous fungi isolated in this study provide substances for biochemical innovation.

Scytinopogon sp. is a soil-inhabiting fungus. There were no ascomata of xylariaceous fungi in the niche of *Scytinopogon*. According to Rogers (2000), the xylariaceous fungi in our study mainly dwelt in plant substrata and decomposed lignin and cellulose. Their niche occupancy were different. They might not compete for the same ecological niche directly.

Another possibility is commensalism. Xylariaceous fungi were present in 70% of the base and in 20% of the top of *Scytinopogon* sp. basidiomata according to PCR detection. This implied that xylariaceous fungi grew upwards from the basal parts of *Scytinopogon* sp. basidiomata. They were non-detectable in the apical 1 cm segments of the tips of 585 basidiomata (data not shown); only very few tips of the basidiomata were colonized by xylariaceous fungi, which indicated that the apical growth of *Scytinopogon* sp. basidiomata was faster than that of the xylariaceous fungi, and xylariaceous fungi grew continuously in the basidiomata. Xylariaceous fungi presumably obtained the nutrients from their host. Commensalism between xylariaceous fungi and *Scytinopogon* sp. was inferred. Latent saprotrophism or latent parasitism can be triggered by host age or seasonal change for many fungal endophyte in plant hosts (Van Bael et al., 2005; Herre et al., 2007), and presumably for fungi as well. To know the ecological role in full requires more characterization in the field and under laboratory conditions.

Xylariaceous fungi not only grew in the plant tissues, they established a stable coexistence with *Scytinopogon* sp. basidiomata in the forest. Some of these xylariaceous fungal species were endophytes in nearby plants. Our study suggested that these endophytic xylariaceous fungi were sources of fungicolous fungi in *Scytinopogon* sp. in the forest.

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References

Arnold, A.E., Miadlikowska, J., Higgins, K.L., Sarvate, S.D., Gugger, P., Way, A., Hofstetter, V., Kauff, F., Lutzoni, F., 2009. A phylogenetic estimation of tropic transition networks for ascomycetous fungi: are lichens cradles of symbiotrophic fungal diversification? Syst. Biol. 58, 283–297.
Brunner, F., Petrini, O., 1992. Taxonomy of some *Xylaria* species and xylariaceous endophytes by isozyme electrophoresis. Mycol. Res. 96, 723–733.
Chen, J., Zhang, L.C., Xing, Y.M., Wang, Y.Q., Xing, X.K., Zhang, D.W., Liang, H.Q., Guo, S.X., 2013. Diversity and taxonomy of endophytic xylariaceous fungi from medicinal plants of *Dendrobium* (Orhidaceae). PLoS One 8, e58268.
Ceruti, A., 1988. Evoluzione delle conoscenze biologiche sul genere Tuber. In: Bencivenga, M., Granetti, B. (Eds.), Attidel Secondo Congresso Internazionale Sul Tartufo. Comunitá Montana dei Monti Martani edel Serano, Spoleto, pp. 1–16.
Daferner, M., Mensch, S., Anke, T., Sterner, O., 1999. Hypoxysordarin, a new sordarin derivative from *Hypoxylon croceum*. Z. Naturforsch. C 54, 474–480.
Deacon, J.W., 1976. Studies on *Pythium oligandrum*, an aggressive parasite of other fungi. Trans. Br. Mycol. Soc. 66, 383–391.
Doyle, J.J., Doyle, J.L., 1990. Isolation of plant DNA from fresh tissue. Focus 12, 13–15.
Gams, W., Diederich, P., Pöldmaa, K., 2004. Fungicolous fungi. In: Mueller, G.M., Bills, G.F., Foster, M.S. (Eds.), Biodiversity of Fungi: Inventorying and Monitoring. Elsevier, Amsterdam, pp. 343–392.
Govinda Rajulu, M.B., Thirunavukkarasu, N., Babu, A.G., Aggarwal, A., Suryanarayanan, T.S., Reddy, M.S., 2013. Endophytic Xylariaceae from the forests of Western Ghats, southern India: distribution and biological activities. Mycology 4, 29–37.
Guo, L.D., Hyde, K.D., Liew, E.C., 2001. Detection and taxonomic placement of endophytic fungi within frond tissues of *Livistona chinensis* based on rDNA sequences. Mol. Phylogenetics Evol. 20, 1–13.
Gundel, P.E., Helander, M., Casas, C., Hamilton, C.E., Faeth, S.H., Saikkonen, K., 2013. *Neotyphodium* fungal endophyte in tall fescue (*Schedonorus phoenix*): a comparison of three Northern European wild populations and the cultivar Kentucky-31. Fungal Divers. 60, 15–24.

Hall, T.A., 2011. BioEdit: an important software for molecular biology. GERF Bull. Biosci. 2, 60–61.
He, P., He, X., Zhang, C., 2006. Interactions between *Psilocybe fasciata* and its companion fungus *Acremonium strictim*. Ecol. Res. 21, 387–395.
Herre, E.A., Mejía, L.C., Kyllo, D.A., Rojas, E., Maynard, Z., Butler, A., Van Bael, S.A., 2007. Ecological implications of anti-pathogen effects of tropical fungal endophytes and mycorrhizae. Ecology 88, 550–558.
Hwang, I.H., Swenson, D.C., Gloer, J.B., Wicklow, D.T., 2015. Pestaloporonins: caryophyllene-derived sesquiterpenoids from a fungicolous isolate of *Pestalotiopsis* sp. Org. Lett. 17, 4284–4287.
Jeffries, P., 1995. Biology and ecology of mycoparasitism. Can. J. Bot. 73, 1284–1290.
Jiang, W., Yang, G., Zhang, C., Fu, C., 2011. Species composition and molecular analysis of symbiotic fungi in roots of *Changnienia amoena* (Orchidaceae). Afr. J. Microbiol. Res. 5, 222–228.
Ju, Y.M., Hsieh, H.M., 2007. *Xylaria* species associated with nests of *Odontotermes formosanus* in Taiwan. Mycologia 99, 936–957.
Kirk, P.M., Cannon, P.F., Minter, D.W., Stalpers, J.A., 2008. Ainsworth & Bisby's Dictionary of Fungi, tenth ed. CAB International, Wallingford.
Lin, W.R., Wang, P.H., Chen, M.C., Kuo, Y.L., Chiang, P.N., Wang, M.K., 2015. The impacts of thinning on the fruiting of saprophytic fungi in *Cryptomeria japonica* plantations in central Taiwan. For. Ecol. Manag. 336, 183–193.
Lu, S.Y., Hwang, L.S., Huang, H.H., 2008. Complication of Meteorological Records for the Lienhuachih Station 1997–2007. Taiwan Forestry Research Institute, Taipei (in Chinese with English abstract).
Okane, I., Srikitikulchai, P., Tabuchi, Y., Sivichai, S., Nakagirl, A., 2012. Recognition and characterization of four Thai xylariaceous fungi inhabiting various tropical foliages as endophytes by DNA sequences and host plant preference. Mycoscience 12, 122–132.
Pacioni, G., Leonardi, M., Aimola, P., Ragnelli, A.M., Rubini, A., Paolocci, F., 2007. Isolation and characterization of some mycelia inhabiting *Tuber* ascomata. Mycol. Res. 111, 1450–1460.
Rajulu, M.B.G., Thirunavukkarasu, N., Suryanarayanan, T.S., Ravishankar, J.P., El Gueddari, N.E., Moerschbacher, B.M., 2011. Chitinolytic enzymes from endophytic fungi. Fungal Divers. 47, 43–53.
Ratnaweera, P.B., Williams, D.E., de Silva, E.D., Wijesundera, R.L., Dalisay, D.S., Andersen, R.J., 2014. Helvolic acid, an antibacterial nortriterpenoid from a fungal endophyte, *Xylaria* sp. of orchid *Anoectochilus setaceus* endemic to Sri Lanka. Mycology 5, 23–28.
Richardson, S.N., Walker, A.K., Nsiama, T.K., McFarlane, J., Sumarah, M.W., Ibrahim, A., Miller, J.D., 2014. Griseofulvin-producing *Xylaria* endophytes of *Pinus strobus* and *Vaccinium angustifolium*: evidence for a conifer-understory species endophyte ecology. Fungal Ecol. 11, 107–113.
Rogers, J.D., 2000. Thoughts and musings on tropical Xylariaceae. Mycol. Res. 104, 1412–1420.
Rogers, J.D., Ju, Y.M., Lehmann, J., 2005. Some *Xylaria* species on termite nests. Mycologia 97, 914–923.
Sanchez-Azofeifa, A., Oki, Y., Fernandes, G.W., Ball, R.A., Gamon, J., 2012. Relationships between endophyte diversity and leaf optical properties. Trees 26, 291–299.
Shim, S.H., Baltrusaitis, J., Gloer, J.B., Wicklow, D.T., 2011. Phomalevones A–C: dimeric and pseudodimeric polyketides from a fungicolous Hawaiian isolate of *Phoma* sp. (Cucurbitariaceae). J. Nat. Prod. 74, 395–401.
Stadler, M., Hellwig, V., 2005. Chemotaxonomy of the Xylariaceae and remarkable bioactive compounds from Xylariales and their associated asexual stages. In: Pandalai, S.G. (Ed.), Recent Research Developments in Phytochemistry, Vol. 9. Research Signpost, Trivandrum, pp. 41–93.
Su, H.J., 1984. Studies on the climate and vegetation types of the natural forests in Taiwan (II): altitudinal vegetation zones in relation to temperature gradient. Q. Jounal Chin. For. 17, 57–73.
Swofford, D.L., 2003. PAUP*: Phylogenetic Analysis Using Parsimony (* and Other Methods), Version 4.0b10. Sinauer Associates, Sunderland.
Thompson, J.D., Gibson, T.J., Plewniak, F., Jeanmougin, F., Higgins, D.G., 1997. The Clustal X windows interface: flexible strategies for multiple sequence alignment aided by quality analyse tools. Nucleic Acids Res. 25, 4876–4882.
Van Bael, S.A., Maynard, Z., Rojas, E., Mejia, L.C., Kyllo, D.A., Herre, E.A., Robbins, N., Bischoff, J.F., Arnold, A.E., 2005. Emerging perspectives on the ecological roles of endophytic fungi in tropical plants. In: Dighton, J., White Jr., J.F., White, J., Oudemans, P. (Eds.), The Fungal Community: its Organization and Role in the Ecosystem. Taylor & Francis, London, pp. 181–192.
Whalley, A.J.S., Edwards, R.L., 1995. Secondary metabolites and systematic arrangement within the Xylariaceae. Can. J. Bot. 73, 802–810.
White, T.J., Bruns, T., Lee, S., Taylor, J., 1990. Amplification and direct sequencing of fungal ribosomal RNA genes for phylogenetics. In: Innis, M.A., Gelfand, D.H., Sninsky, J.J., White, T.J. (Eds.), PCR Protocols: a Guide to Methods and Applications. Academic Press, San Diego, pp. 315–322.
Yurkov, A., Kruger, D., Begerow, D., Arnold, N., Tarkka, M.T., 2012. Basidiomycetous yeasts from Boletales fruiting bodies and their interactions with the mycoparasite *Sepedonium chrysospermum* and the host fungus *Paxillus*. Microb. Ecol. 36, 295–303.