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Full Length Research Paper

Intercropping peanut with traditional Chinese medicinal plants improves soil microcosm environment and peanut production in subtropical China

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Many environmental issues affect peanut production in continuous monocropping system. Deterioration of soil microbial communities, especially for decrease in fungal diversity and increase in fungal pathogens, is widely hypothesized as a key factor for decreasing peanut production. In this study, a pot experiment was conducted to investigate the changes in soil microbial communities and peanut yield under the scheme of peanut intercropped with traditional Chinese medicinal plants, including *Atractylodes lancea*, *Dioscorea zingiberebsis*, *Euphorbia pekinensis*, *Ophiopogon platyphyllum* and *Pinellia ternate*. The results showed that soil microcosm environment was improved, and the fungal diversity and fungal pathogens such as *Fusarium* sp. and *Verticillium* sp. were restrained when peanut intercropped with *A. lancea* and *E. pekinensis*. The DGGE analysis of 18S-rRNA from DNA of the total soil communities showed obvious transferring of species of fungi between peanut monocropping and the intercropping systems. Compared with the control, the superoxide dismutase activity of peanut was increased by 43% in *A. lancea* and 37% in *E. pekinensis* intercropping systems, along with 37 and 16% yield improvement of peanut, respectively. Based on the result of the pot experiment, *A. lancea* and *E. pekinensis* were intercropped with peanuts in a field experiment. A considerable agreement was found between the results obtained from the field and pot experiments. Compared with peanut monocropping system, colony form unit of mould decreased by 31% in *A. lancea* and 18% in *E. pekinensis* intercropping treatments, where peanut yields were respectively increased by 39 and 35%. Further research should include integrated PCR-DGGE analysis to determine the transfer of peanut soil-born pathogens and its mechanism, and the optimization of intercropping system and planting density of medicinal plants to obtain the best benefits, and the understanding of the long-term effect of the intercropping systems.

Key words: Peanut, soil microbial community, intercrop, medicinal plants, succession monocropping obstacles.

INTRODUCTION

Peanut (*Arachis hypogaea* L.) is cultivated in tropical, subtropical and warm temperate climate regions around the world, and China is one of the largest peanut producers (Stalker, 1997). Long-term continual peanut cropping results in continuous decline in peanut yield were probably because of the deterioration of soil microbial community (Sun et al., 2001; Wang and Chen, 2005). Similar phenomena have been found in other crops (Hu et al., 2006; Larkin, 2003; Li et al., 2005). Generally, continuous monocropping may reduce the diversity of bacteria in both species and quantity, lower the number of fungi species and increase mould quantity diversity (Hu et al., 2006; Li et al., 2005; Ryszkowski et al., 1998; Xie et al., 2007; Xu and Wang, 2003). Moreover, the reduction of antagonistic strains and accumulation of plant residues provide a favorable niche for root pathogens,

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causing serious peanut disease and decrease in soil quality (Li et al., 2005; Nannipieri et al., 2003). The composition of soil microorganisms could also be strongly affected by plant species (Wieland et al., 2001). Intercropping has been practiced widely as a traditional way to increase peanut production and make use of micronutrients and other minerals in the rhizosphere (Acosta-Martínez et al., 2004; Chu et al., 2004; Inal et al., 2007; Xu and Wang, 2003). However, few reports have been focused on the influences on soil microbial community (Sun et al., 2001). Crops such as wheat, maize and grain sorghum have been intercropped with peanut for many years. However their shortages in growth period, soil conditions, and relatively lower economic profits impede their application in many areas of China (Connolly et al., 2001).

China has abundant medicinal plant resources, which contain plentiful active compounds, such as alkaloids, terpenoids, phenols, and glycosides. Medicinal plants have widely been used in traditional Chinese medicine, and great attention has been generated in the research on the extraction of active compounds or components from the plants. It has been found that many medicinal plants contain active compounds that have antifungal and antibacterial activities. For example, terpenoid compounds have great effects on protecting roots and stem cortex against fungal infection (Farzaneh et al., 2006). Unfortunately, there is almost no report on the intercropping between peanut and medicinal plants and the influence on the microbial communities in soil.

In our previous study, five traditional medicinal plants were screened to intercrop with peanuts in primary (Xie et al., 2007). In the present study, the five medicinal plants were further studied to examine the changes in soil microbial diversity, peanut physiology status, and yield. As the main pathogens species, the fungi communities in the soil of different treatments were assessed by polymerase chain reaction-denaturing gradient gel electrophoresis (PCR-DGGE). In combination with culture-dependent techniques and morphology identification, the relatively complete knowledge of this crucial microbial group can be obtained. Based on the results of pot experiment, some medicinal plants were introduced to the field experiment. The objectives of this study were to (i) determine the impact of peanut intercropped with medicinal plants on the soil microbial diversity, (ii) analyze the influence of these intercropping systems on peanut growth and yield, and (iii) determine whether the results of field experiment were consistent with pot experiment, and implications for further application in agro-ecosystem to overcome succession monoculture obstacles in peanut production.

MATERIALS AND METHODS

Soil sampling and study site

The soil used in the pot experiment was collected from the surface layer (0 - 20 cm in depth) of a 15-year-old continuous peanut cropping upland at the Ecological Experimental Station of Red Soil, Chinese Academy of Sciences, located in the central subtropical China (N28º13', E116º55' with an altitude of 45 m asl). The soil was classified as an Udic Ferrosol (Alumi-Orthic Acrisol in FAO taxonomy; Udic Kandiudults in USDA taxonomy), which is generally called red soil in China. The soil contained 13.1 g kg^{-1} organic matter, 0.76 g kg^{-1} total N, 2.48 mg kg^{-1} hrdrolyzable N, 14.65 mg kg^{-1} rapidly available P , 258.40 mg kg^{-1} rapidly available K, 10.34 cmol kg^{-1} CEC, and pH 4.96 (1: 2.5H_2O) .

The field experiment was conducted on the soil at the Ecological Experimental Station of Red Soil, Chinese Academy of Sciences. The mean annual precipitation amounts to 1750 mm (from 50 years), and the rainfall generally concentrates from April to later of June. The monthly average temperature varies from minimum (5.9℃) in January and Maximum (30.0℃) in July.

Pot experimental design

Pot experiment was conducted in the greenhouse in 2006 under natural day/night regime and watered as necessary. There were six treatments and each with five replications. Peanut monocropping was the control, and the peanuts intercropped with five Chinese medicinal plants were the treatments. The five traditional medicinal plant species were *Atractylodes lancea*, *Euphorbia pekinensis*, *Diocorea zingiberebsis*, *Ophiopogon platyphyllum,* and *Pinellia ternate*. Plastic pots (23 cm in diameter and 23 cm in height) were used, and each contained 8.0 kg soil sample. One plant of medicinal plants with three seeds of peanut was planted in each pot. All medicinal plants (3-year-old) were transplanted along the edge of pots in February 2005, which sprouted in early March of 2006. Peanut seeds were sown on May 3, 2006, in the center of the pot. Each pot was fertilized with 2 g of $Ca_3(PO_3)_2$, 1 g of urea, and 2 g of K_2SO_4. During the critical period of peanut growth, such as flowering-pegging and pod-filling stages, soil samples were taken by sampling 3 points of the soil at the depth of 0 - 20 cm around the peanut in each pot using an auger. The second leaf from top of each peanut during bloom stage was also sampled to analyze activity of peanut antioxidant enzyme.

Field experimental design

Based on the result of pot study in 2006, *A. lancea* and *E. pekinensis* were introduced to the field experiment in 2007. The field experiment had randomized 3 treatments with 3 replications for each treatment. Each plot was 5.0 × 4.0 m in size. Eight rows of peanut plants were grown in the control plot. There are a row of peanut every 50 cm. Six rows of peanut plants were grown along with two rows of transplanted medicinal plants in each intercropping treatment plot. The layout was two rows of peanut plants intercropped with one row of medicinal plants (PPMPPMPP, where P represents peanut rows and M represents medicinal plant rows). Both peanuts and medicinal plants were planted with 50 cm row width and 20 cm plant width. Peanut seeds were sown on April 8, 2007, and meanwhile *A. lancea* and *E. pekinensis* of about 2-year-old were transplanted from their own habitat in Jiangsu, China. The peanuts were fertilized and managed in the same way as local farmers' practice.

At pod-filling stage, soil samples were sampled from 5 points around the peanut at the depth of 0 - 20 cm in each plot using an auger. After removing visible root debris, soil samples were stored at 4℃ till analysis.

Soil cultivable microbial diversity analysis

The total numbers of cultivable bacteria, fungi and actinomyces

were determined according to colony forming units (CFUs) on agar plates using dilution plate methods (Fan and Li, 1982). The media used for the cultivation of bacteria, actionmyces and fungi were beef-extract peptone medium, Cause's No. 1 synthetic medium, and Czapek's medium, respectively (Institute of Soil Science, Chinese Academy of Sciences, 1985). Cultivable mould was identified using optical microscope (Barnett and Hunter, 1977; Wei, 1979).

PCR-DGGE

In pot study, total community DNA was extracted from each of the 12 soil samples (0.5 g) using the soil DNA isolation kit (Genmed Scientifics Inc. USA). PCR amplifications of the 18S rRNA fragments were carried out using fungi specific primer set EF4 (5'-GGA AGG GAT GTA TTT ATT AG-3') and Fung5r (5'-GTA AAA GTC CTG GTT CCC C-3'), as described by Borneman and Hartin (2000). The GC clamp (CGC CCG CGC CGC GCG GCG GGC GGG GCG GGG GCA CGG GGG G) was attached to the 5'-end of the forward primer (Muyzer et al., 1993). Each PCR mixture contained 50 ng extracted DNA as template, 10 × reaction buffer, 2U Taq (TaKaRa), 0.4 µM of each primer, 2 mM of $MgCl_2$ and 200 µM of each dNTP, in a final volume of 50 µl. The PCR protocol included a 5 min initial denaturation at 94°C, 35 cycles of 94°C for 1 min, 55°C for 1 min and 72°C for 1.5 min and a final extension at 72°C for 10 min. Final PCR products were checked on a 1% agarose gel stained with ethidium bromide.

DGGE was performed on a DGGE Electrophoresis System (DGGE 2001, CBS, USA). For each treatment, about 400 ng of amplified 18S rRNA gene product was loaded into each lane on an 8% (w/v) acryl-bisacrylamide gel (W/W = 37.5:1 Sangon, Shanghai) with a 30 - 55% denaturing gradient (100% denaturant contained 40% formamide and 7 M urea). Electrophoresis was performed in 1×TAE buffer at 60°C, with 60 V for 1 h, followed by a constant voltage of 100 V for 14 h. After that, gels were stained in 0.01% SYBR Green I (Molecular Probes USA) in 1×TAE solution for 30 min. The gels were later photographed by gel explorer (Bio-Rad Inc., USA).

Anti-oxidative enzymes assays

To evaluate peanuts physiological condition, activities of antioxidant enzymes, including superoxide dismutase (SOD), peroxidase (POD) and catalase (CAT) of peanuts were determined. Fresh leaves (0.5 g) collected from the second leaf from the top of each peanut during the bloom stage were well homogenized with extraction buffers (phosphate acid buffer, pH 7.8), and centrifuged for 10 min (8,000 rev min^{-1}, 4°C). The supernatant was measured by a UV-vis spectrophotometer (2802S, UNIC, USA) at 560 nm for SOD, 470 nm for POD, and 240 nm for CAT (in the control tube, corresponding buffer was added instead of enzyme extract). One unit of SOD activity was defined as the amount of enzyme that caused 50% inhibition of photochemical reduction of NBT. One unit of POD activity was defined as an absorbance change of 0.01 unit per minute, and one unit of CAT activity defined as the amount of 1 µM H_2O_2 decomposed by 1 mg tissue protein per second (Chance and Maehly, 1955; Li, 2000).

Peanut yield

The peanuts in the pot experiment were harvested on August 25, 2006, while peanuts in the field experiment harvested on August 3, 2007. After harvest, the pods, straws and roots were cleaned, dried by an oven at 80°C, and weighed separately.

Statistics analysis

DGGE gels were clustering analyzed using Bionumerics 4.6 software. Variance analysis of other results was performed by SPSS 14.0 professional software (SPSS Inc.).

RESULTS

Peanut yield

In pot study, the biomass and yield of peanut were significantly increased when peanut plants were grown with *A. lancea* and *E. pekinensis* as compared to control (monocropped). Particularly, in the intercropping with *A. lancea*, the increase percentages were up to 12% in biomass and 37% in yield of peanuts. In contrast, the peanut yield was significantly decreased when peanut plants were grown with *D. zingiberebsis* and *O. platyphyllum*, while, the peanut intercropped with *P. ternate* exhibited no significant difference from the control (Table 1). This might partially attribute to spatial limitation and competition for nutrients in the pots, which could be mitigated in field trials by adjusting the appropriate planting density and fertilization. These results indicate that peanut and medicine plants intercropping, if used appropriately, can increase peanut yield.

In the field study, the peanuts in the intercropping treatments exhibited a better growth condition and scarcely any disease occurrences, with comparison of serious disease in peanut monocropping system. Compared with the control (monocropped) in rows, peanut pod yield in intercropping treatments was increased by 39% in *A. lancea* and 35% in *E. pekinensis*, and the straw yields was increased by 48 and 52%, respectively. If compared with plots, the straw and pod yield also increased but not significantly for there were 8 rows of peanuts in CK but 6 rows peanut in intercropping plots (Table 2).

Culturable soil microbial diversity

In pot study, except for *P. ternate* intercropping, the number of colony forming units (CFU) of mould in the intercropping treatments decreased significantly, as compared with the control at both stages, while CFU of bacteria increased in all intercropping treatments at the pod-filling stage (Table 3). For actinomycete, no significant difference was found in most intercropping treatments as compared to the control. Only in *A. lancea* and *E. pekinensis* treatments, yeast was slightly increased at both stages. These results indicated that some medicinal plants might restrain the growth of soil borne pathogens, and increase bacteria and yeast, which were beneficial to a healthy soil environment for peanuts. Among medicinal plants chosen in this study, *A. lancea* and *E. pekinensis* treatments significantly improved soil

Table 1. Effects of medicinal plants intercropped with peanut on the biological and pod yield of peanut in pot study.

Treatment	Total biomass (g plant^{-1})	Pod yield (g plant^{-1})
CK	27.97 ± 1.44^{ab}	15.29 ± 0.89^{b}
A. lancea	31.35 ± 0.98^{c}	20.89 ± 1.47^{d}
E. pekinensis	30.14 ± 0.63^{a}	17.70 ± 0.58^{c}
D. zingiberebsis	27.49 ± 0.52^{bc}	13.80 ± 0.32^{a}
O. platyphyllum	26.89 ± 0.47^{c}	13.66 ± 0.43^{a}
P. ternate	29.19 ± 1.35^{bc}	15.70 ± 0.59^{b}

Table 2. Effects of medicinal plants intercropped with peanut on peanut yield in field study.

Treatment	Yield (t hm^{-2})	
	Pod	Straw
Peanut monocropping	2.03±0.07	1.64±0.02
Peanut intercropped with *A. lancea*	2.11±0.22	1.82±0.20
Peanut intercropped with *E.pekinensis*	2.06±0.09	1.87±0.12

Table 3. Effects of medicinal plants intercropped with peanut on microbial communities in the pot soil.

Microbial colony		CK	*A. lancea*	*E. pekinensis*	*D. zingiberebsis*	*O. platyphyllum*	*P. ternate*
Bacteria	f	20.56±2.44^{b}	21.88±2.86^{b}	19.27±3.08^{b}	27.56±3.96^{c}	12.51±1.43^{a}	29.27±2.81^{c}
(×10^{6}CFU·g^{-1} DM)	p	9.86 ±1.21^{a}	15.18±1.17^{bc}	15.18±1.17^{bc}	10.82±2.07^{a}	14.16±1.55^{b}	16.22±1.88^{c}
Actinomycete	f	0.20±0.03^{a}	0.41±0.04^{c}	0.26±0.04^{b}	0.39±0.06^{c}	0.21±0.04^{ab}	0.23±0.05^{ab}
(×10^{6}CFU·g^{-1} DM)	p	1.92±0.35^{a}	2.58±0.38^{b}	2.57±0.32^{b}	2.36±0.43^{b}	2.58±0.38^{b}	2.39±0.45^{b}
Mould	f	14.65±2.27^{c}	10.94±1.54^{b}	6.68±0.65^{a}	5.63±0.61^{a}	6.89±0.83^{a}	13.62±1.99^{c}
(×10^{4}CFU·g^{-1} DM)	p	18.08±2.07^{c}	16.27±1.89^{bc}	14.09±1.37^{b}	8.69±0.90^{a}	14.79±2.21^{b}	15.75±1.53^{b}
Yeast	f	11.27±1.58^{b}	13.82±1.82^{c}	11.21±1.85^{b}	6.22±0.73^{a}	7.46±1.29^{a}	19.13±2.08^{d}
(×10^{4}CFU·g^{-1} DM)	p	8.22±1.52^{bc}	12.57±1.99^{d}	9.12±1.68^{c}	7.29±1.04^{ab}	6.85±1.14^{ab}	5.62±0.52^{a}

'f' stands for abbreviation of flowering-pegging stage, 'p' for pod-filling stage.

Table 4. Effects of medicinal plants intercropped with peanut on microbial communities of the soils in the field study.

Treatment	Bacteria (×10^{6} CFU·g^{-1} DM)	Actinomycete (×10^{6} CFU·g^{-1} DM)	Mould (×10^{4} CFU·g^{-1} DM)	Yeast (×10^{4} CFU·g^{-1} DM)
CK	3.14±0.30^{a}	0.21±0.03^{a}	5.42±0.14^{c}	0.41±0.13^{a}
A. lancea	8.13±0.93^{c}	0.33±0.03^{b}	3.72±0.48^{a}	0.87±0.19^{c}
E. pekinensis	6.19±0.28^{b}	0.19±0.01^{a}	4.44±0.37^{b}	0.74±0.18^{bc}

*Significance at 0.05 level, **significance at 0.01 level.

microcosm environment, and this corresponded with increases in peanuts yields (Tables 1 and 2).

In field study, the CFUs of bacteria and yeast were greatly increased ($p < 0.05$), whereas the CFU of the mould was decreased by 31 and 18% for *A. lancea* and *E. pekinensis* treatments, respectively, suggesting that a positive regulation among different microbial groups (Table 4). Intercropping treatments demonstrated greater total microbial quantity diversity and a higher ratio of bacteria to fungi than the monocropping. Moreover, there were much more observable variations of microbial communities in field than in the pot soil, probably because the open environment of soil in field might have strengthened these effects.

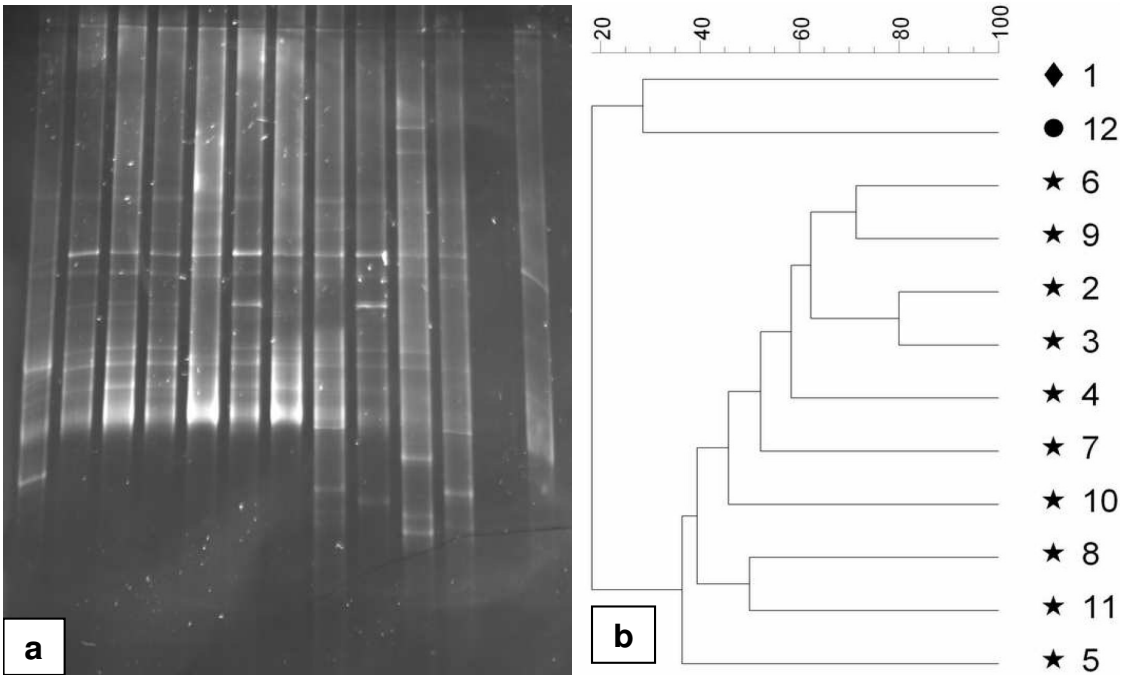

Figure 1. DGGE analysis of 18S-rRNA from total soil microbial communities DNA in pot study. **(a)**: 18S rRNA DGGE profiles. **(b)**: Cluster analyses generated from the above DGGE profiles using Bionumerics 4.6 software. Codes of 1-6 stand for the treatments of peanut monocropping (CK) and peanut intercropped with *A. lancea*, *E. pekinensis*, *D. zingiberebsis*, *O. platyphyllum* and *P. ternate*, respectively, at peanut pod-filling stage. Codes of 7-12 stand for the corresponding treatments at peanut flowering-pegging stage.

Table 5. Effects of medicinal plants intercropped with peanut on species of mould in the soils by morphology identification.

Microbia	CK	*A. lancea*	*E. pekinensis*	*D. zingiberebsis*	*O. platyphyllum*	*P. ternate*
Aspergillus		+		+	+	+
Penicillium	+	+	+	+	+	+
Paecilomyces	+	+	+	+	+	+
Trichoderma				+	+	
Monscus		+	+		+	
Fusarium	+				+	+
Verticillium	+					+
Gliocladium		+			+	
Stibum			+	+	+	+

"+" means checked out in the corresponding soil.

Communities of peanut soils based on morphology identification

Species of culturable fungi separated from the pot soils were identified (Table 5), and the results showed that medicinal plants treatments increased the soil fungi diversity. Among the fungal species, *Fusarium* sp. and *Verticillium* sp. were common pathogens of peanuts that can cause root rotting and chlorosis in peanuts (Berg et al., 2005; Rojo et al., 2007). Both pathogens had been detected in the control. However, neither of them was presented in *E. pekinensis*, *A. lancea* and *D. zingiberebsis* treatments, implying that these three intercropping groups could control those soil-borne pathogens and enrich soil fungal diversity effectively.

Fungal microbial communities studied by 18S-rRNA DGGE

In the pot study, about 20 DNA bands were detected in flowering-pegging and pod-filling stages (Figure 1a). Many bands existed in all samples with minor differences, while some were specific, lost, or with various intensities

Table 6. Effects of intercropping system on peanuts antioxidant enzyme.

Treatment	SOD activity (Unit·g^{-1} FW·min^{-1})	POD activity (Unit·g^{-1} FW·min^{-1})	CAT activity (Unit·g^{-1} FW·min^{-1})
CK	203.68±50.41^{a}	1420.60±134.38^{ab}	18.76±2.64^{bc}
A. lancea	291.75±33.48^{b}	1238.80±108.25^{a}	22.82±2.06^{c}
E. pekinensi	278.46±22.20^{b}	1259.60±197.47^{a}	17.58±2.11^{bc}
D. zingiberebsis	249.61±48.65^{ab}	1331.60±243.38^{ab}	9.24±1.74^{a}
O. platyphyllum	247.87±15.97^{ab}	1491.00±121.08^{b}	14.18±2.47^{ab}
P. ternate	217.43±12.60^{a}	1226.20±89.77^{a}	15.18±3.22^{ab}

SOD, superoxide dismutase; POD, peroxidase; CAT, catalase.

in different intercropping treatments at different stages. This suggested that fungal diversity in the soil environment was not inherently stable, and it could be greatly influenced by vegetation, and altered at different growth stages of peanut. Cluster analysis showed that the control and intercropping systems shared low similarity in fungal community structure (Figure 1b), implying that the intercropping with medicinal plants could also influence soil fungi species. In fact, the fungal profile of *A. lancea* treatment was 80% similar to that of *E. pekinensis* treatment at peanut pod-filling stage.

Antioxidant enzyme activities of peanut

In the pot study, SOD activity significantly increased in all the five intercropping treatments compared with the control, and the increase was up to 43% in *A. lancea* treatment and 37% in *E. pekinensis* treatment (Table 6). However, POD and CAT appeared to be less sensitive than SOD to the cropping system change. POD and CAT activities slightly decreased in most intercropping treatments, and CAT activity increased only in *A. lancea* intercropping treatment.

DISSCUSSION

A continuous monocropping system may result in negative impact on crop production, soil microbial diversity, and soil fertility (Monneveux et al., 2006; Ryszkowski et al., 1998; Sun et al., 2001; Wang and Chen, 2005; Xu and Wang, 2003). Diverse intercropping can control crop disease and therefore improve productivity (Govaerts et al., 2006; Zhu et al., 2000). Similarly, data in Tables 1 and 2 showed an increase of 37 - 39% in peanut yield when peanut intercropped with *A. lancea.* Positive effects were also found in *E. pekinensis* and *P. ternate* treatments. Because medicinal plants possess greater economic value than other plants, intercropping peanut with selective medicinal plant species may have both environmental and economic benefits.

Soil microbial communities play a key role to plant growth and metabolic functions of soil, and they also contribute to nutrients recycling and competition against pathogens (Hagn et al., 2003). The microbial community is very sensitive to vegetations, while it could in turn strongly affect the plants growth (Benizri and Amiaud, 2005; Kowalchuk et al., 2002; Lejon et al., 2005; Sall et al., 2006; Yang and Crowley, 2000). The cultivation of continuous monocropping may result in increase of fungi and decrease of bacteria as well as decrease of bacteria to fungi ratio, and therefore accelerate abundance of soil borne pathogens around plant roots (Ryszkowski et al., 1998). Such changes in soil microbial communities have wide impact on soil quality including reduction of crop production. Considering the limitation of single technique, culture-dependent and culture-independent methods should be involved to provide an objective assessment of soil microcosm (Phillips, 2006). In the pot study, CFU of bacteria increased in most intercropping treatments together with decrease in CFU of mould, the ratio of bacteria to fungi was greater than that of monocropping system (Table 3), demonstrating a positive effect of the intercropping systems.

As most pathogens of peanuts are fungi (Rojo et al., 2007), the alternation of fungi species is an evident index on evaluating soil microcosm. Plant species were reported to influence rhizosphere-associated fungi antagonistic to pathogens (Berg et al., 2005). In this study, nine species of mould were identified in most treatments, including common soil fungi *Aspergillus* sp., *Penicillium* sp., and *Paecilomyces* sp. (Table 5). While two pathogens of peanut *Fusarium* sp. and *Verticillium* sp. was found in the monocropping soil, those pathogens did not exist in *A. lancea*, *E. pekinensis* and *D. zingiberebsis* intercropping treatments. Combined with 18S-rRNA DGGE analysis results, fungi species was increased in the intercropping systems, but the peanut pathogen species were actually reduced. Large amounts of sesquiterpene and triterpene in *A. lancea* and *E. pekinensis* might be the reasons for the apparent regulation on soil fungi of these two plants, and terpenoid had widely been used to control pathogens (Guo et al., 2006; Kong and Ming, 1996). As strongly active allelopathy materials, terpenoid in low concentration could effectively regulate dynamics of soil microcosm balance, and enhance plant adaptability to environment. *D. zingiberebsis* also had a positive effect on soil microcosm as *A. lancea* and *E. pekinensis*,

possibly owing to the release of saponin from the root (Li et al., 2003). However, peanut yield in *D. zingiberebsis* treatment was less than that of peanut monocropping group. A possible explanation is that the big size of root system of *D. zingiberebsis* might take too much space, and take up too much nutrients in the pot. It reminded us that many factors should be considered in the intercropping systems, and those factors should include growth habit, usage of space and nutrients, life span, climatic condition, and economic benefits.

Autotoxin materials and soil-borne pathogens accumulated during the continuous monocropping of peanut provide an unhealthy environment for the plant root. To obtain a higher yield and quality of peanut, balanced microbial community and high resistance to environmental stress were all indispensable. Antioxidative enzymes, such as SOD, POD and CAT are common and important indices for evaluating the physiological conditions and stress-tolerance of plants. A main role of SOD is to catalyze the dismutation of superoxide anions to dioxygen and hydrogen peroxide (H_2O_2). The increased production of H_2O_2 is subsequently eliminated by POD or CAT detoxification (Garratt et al., 2002; Raza et al., 2007; Shalata et al., 2001). In the intercropping systems, SOD activities of peanuts all increased, especially for *A. lancea* and *E. pekinensis* treatments. It is suggested that soil microbial community, crop physiological condition and productivity might have correlated one another.

A. lancea and *E. pekinensis* intercropping treatments had positive effects on soil microbial diversity and peanut yield, which may attribute to the active materials secreted from these two plants. Therefore, these two medicinal plants can be chosen in the field study. There was considerable agreement between results from the field and those from the pot experiment, and it was unexpected that effects in field study were more apparent than that in the pot study, implying the impacts of the intercropping system might previously be underestimated. It was true that connected environment of soil in field could accelerate the growth of soil-borne pathogens, and result in serious and extensive diseases of peanut, as appeared in the control in field study. However, the peanut intercropped with medicinal plants was free of these diseases. We believed that the open environment of soil in the field also enhanced the effects of terpenoid that was largely contained in *A. lancea* and *E. pekinensis*. On the other hand, the little spatial limitation in the field mitigated the competition in nutrients in the intercropping system.

Compared to the traditional intercropping farming system management, the intercropping systems in this study showed some advantages on soil microcosm and peanut growth, improvement of soil biodiversity, and the restoration of agro-ecosystem of red soil in subtropical China. Conceptually, the intercropping system could be applicable for crops other than peanut, as monocropping system exists extensively in agriculture. Further studies should be conducted to understand the implications of intercropping of crops with variety of plants.

It is important to realize that the effects could differ among different medicinal plants. To select a proper medicinal plant, active components, height, size of root, growth period, and the adaptability to soil and climate of medicinal plant are all needed to take into consideration. As for peanut, medicinal plants belonging to perennial herbage are more appropriate than the arbor, as suggested by Qin et al. (1999). Arbor and frutex might cause great competition in sunlight and water, leading to a reduction in peanut yield (Wang et al., 2003).

This study showed positive effects of the medicinal plants *A. lancea* and *E. pekinensis* intercropped with peanuts on both peanut production and soil microbial community. Future focus should be given on understanding the mechanisms that govern interactions between intercropped species so that the optimized intercropping systems can be proposed. Further research should also use integrated PCR-DGGE analysis to reveal more definite transfer of peanut soil-born pathogens and its mechanism. The long-term effectiveness of the intercropping systems needs to be monitored.

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