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Effects of arbuscular mycorrhizal fungi inoculation on arsenic accumulation by tobacco (*Nicotiana tabacum* L.)

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Abstract

A pot experiment was conducted to study the effects of arbuscular mycorrhizal (AM) fungi (from contaminated or uncontaminated soils) on arsenic (As) uptake of tobacco (*Nicotiana tabacum* L.) in As-contaminated soil. Mycorrhizal colonization rate, dry weight, As and P uptake by plants, concentrations of water-extractable As and As fractions were determined. A low mycorrhizal colonization rate (< 25%) was detected. Our research indicated that AM fungi isolated from polluted soils were no more effective than those from unpolluted soils when grown in symbiosis with tobacco. No significant differences were observed in roots and stalks dry weights among all treatments. Leaves and total plant dry weights were much higher in *Glomus versiforme* treatment than that in control treatment. As contents in roots and stalks from mycorrhizal treatments were much lower than that from control treatment. Total plant As content exhibited the same trend. P concentrations in tobacco were not affected by colonization, nor were stalks, leaves and total plant P contents. Roots P contents were remarkably lower in HN treatments than in other treatments. Meanwhile, decreased soil pH and lower water-extractable As concentrations and higher levels of As fraction bound to well-crystallized hydrous oxides of Fe and Al were found in mycorrhizal treatments than in controls. The protective effect of mycorrhiza against plant As uptake may be associated with changes in As solubility mediated by changing soil pH. These results indicated that under As stress, proper mechanisms employed by AM fungi can protect tobacco against As uptake. Results confirmed that AM fungi can play an important role in food quality and safety.

Key words: arbuscular mycorrhizal fungi; arsenic; tobacco; soil pH
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Introduction

Arsenic (As) naturally occurs in association with sulfides in lead (Pb), nickel (Ni) and copper (Cu), and some ores (Matschullat, 2000). This non-essential element is biologically toxic and a threat to human health. Low level exposures to As can lead to many pathologies such as carcinogenesis. Around some industrial areas, soil As is detected in great amounts through atmospheric deposition and As-contaminated water due to mining and smelting activity (Matschullat, 2000; Smith *et al*., 1998). Bai *et al*. (2008) reported an As concentration of 287.0 mg/kg in soil near an As mine in Hunan Province, China. Crops, including maize and tobacco, are able to extract toxic ions from the soil and concentrate them in their tissues. This process can enhance concentrations of heavy metals/metalloids in plant biomass, and lead to a serious health risk as increased amounts of these toxic elements enter the food chain (Janoušková *et al*., 2005a). Food is the major source of heavy metals/metalloids exposure for the general non-smoking population in most areas. Heavy smoking may represent another important source of exposure to these elements (Janoušková *et al*., 2007; Smith *et al*., 1997). Nine chemical agents classified as "Group I carcinogens" by IARC have been reported to occur in mainstream cigarette smoke, including As and cadmium (Cd) (IARC, 1993; Smith *et al*., 1997). These toxic elements endanger a smoker well-being, as they are directly inhaled. Thereby, methods to reduce heavy metals/metalloids concentrations in tobacco leaves need to be developed urgently.

Several strategies have been employed to reduce concentrations of heavy metals/metalloids in plants. For example, amendments or genetic modifications of plants can immobilize heavy metals/metalloids in soil thus restricting plant uptake (Janoušková *et al*., 2005a, 2005b; Lugon-Moulin *et al*., 2004).

Arbuscular mycorrhiza (AM) is formed with the majority of plants growing under natural conditions (Smith and Read, 1997). Some AM fungi genotypes are able to

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survive in polluted soil due to their adaptation to heavy metals/metalloids stress. Lots of studies have focused on the effects of AM fungi on the uptake of heavy metals/metalloids by plants (Bai *et al*., 2008; Chen *et al*., 2004; Heggo *et al*., 1990; Leung *et al*., 2006). Multiple mechanisms are involved in the changes of heavy metals/metalloids concentrations in plants, such as stimulation of growth and nutrient uptake of host plants (Smith and Read, 1997), changes of pH in rhizosphere soil (Li *et al*., 1991; Li and Christie, 2001), and inhibition of heavy metals/metalloids translocation ratio from root to shoot (Chen *et al*., 2004; Weissenhorn *et al*., 1995; Dong *et al*., 2008). Additionally, mycorrhizal effects may depend on plant species, heavy metals/metalloids concentrations and plant growth conditions (Bai *et al*., 2008; Jankong and Visoottiviseth, 2008; Janoušková *et al*., 2007; Leyval *et al*., 1997). Different populations or geographical isolates of AM fungi show great variability in their tolerance to heavy metals/metalloids and associated stress (Bai *et al*., 2008; Waaland and Allen, 1987; Weissenhorn *et al*., 1994). These results, however, were not confirmed, e.g., by Enkhtuya *et al*. (2000) and Weissenhorn *et al*. (1995).

Presently, minimal research has focused on the effects of AM fungi on heavy metals/metalloids uptake by tobacco, especially As (Janoušková *et al*., 2005a, 2005b). Consequently, a pot experiment was designed to test the effects of AM fungi (from contaminated or uncontaminated soils) on As uptake by tobacco in As-containing soil. We hypothesize that isolates reduce As concentration in tobacco leaves. Because As is similar to phosphorus (P) in physicochemical properties and arsenate is thought to be taken up via the phosphate uptake system which may interact with plant P nutrition (Smith and Read, 1997), thus P concentration was also determined.

In contrast to most other pot experiments, which focused on the interactions between heavy metals/metalloids and AM fungi, in this pot experiment metals/metalloids were not added as soluble salt since artificially polluted soil creates an artificial environment where bound and free metals/metalloids are not in equilibrium (Heggo *et al*., 1990), and the effects of metals/metalloids on the tested organism would be overestimated (Chaney *et al*., 1978). Thus, more representative approach using the contaminated soil near the arsenic sulphide mine was chosen in our study. The mine possesses the largest As sulphide ore (As_4S_4) deposits in Asia. Although it has been mined for over 1500 years, mass exploitation and smelting have began in 1958. The waste gases and tailings discharged from mining and smelting activities have caused long-term and severe contamination to local air, water, soil, livestock, vegetables and crops.

1 Materials and methods

1.1 Soil preparation

Soil (0–20 cm) was collected from an agricultural field near an As sulphide mine, in Shimen County, Hunan Province, China. This area has a North-Asia subtropics maritime monsoon climate, with a mean temperature of 16.7°C and an average rainfall of 1500 mm.

The soil is classified as Ali-Perudic Argosols, and has the following properties: pH 6.57 (soil:water, 1:2.5; *m/V*), 2.73% organic matter, 0.146% Kjeldahl-N and 50 mg/kg of 0.5 mol/L $NaHCO_3$-extractable P, 36.12 mg/kg total As. The total As concentration of the soil was extracted by concentrated HNO_3 and HCl at 1:3 (*V/V*), and determined using atomic fluorescence spectrometer (AFS-230E, Haiguang Instrumental Co., China). A standard reference material: GBW07406 from the Institute of Geophysical & Geochemical Exploration, Chinese Academy of Geoscience, was used to verify the accuracy of soil As measurements. Soil was air-dried and sieved with nylon mesh (4 mm) and then sterilized by autoclaving for 30 min at 121°C on two consecutive days prior to the pot experiment.

1.2 AM fungal inocula

Tobacco (*Nicotiana tabacum* L. cv. Yunyan 85), was either left uninoculated (CK) or inoculated with one of the following four AM fungal isolates: a combined inoculum *Glomus* spp. and *Acaulospora* spp. (referred as HN), wet-sieved from an As-polluted soil in Shimen County, Hunan Province (Bai *et al*., 2008); *Acaulospora mellea* (referred as 40), collected from a coalmine in Shandong Province; *Glomus versiforme* (referred as 90001), selected from an uncontaminated site in Shandong Province; *Glomus caledonium* (referred as 90036), originated from an unpolluted agricultural soil in Henan Province. These AM fungal species were identified morphologically according to current taxonomic criteria (Schenck and Perez, 1990) and internet information by INVAM (http://invam.caf.wvu.edu). They were deposited in the greenhouse of the Institute of Soil Science, Chinese Academy of Sciences. The fungal inoculums were propagated on clover grown in an autoclaved substrate successive propagation cycles for 4 months. The inocula were air-dried and sieved ($\leqslant$ 2 mm), and consisted of a mixture of rhizospheric soil from pure pot culture containing spores, hyphae and mycorrhizal root fragments.

These isolates were selected to represent different AM fungal species and origin to obtain the information on isolate-dependent variability in the reaction of the tobacco to inoculation.

1.3 Experimental design

Tobacco seeds were surface-sterilized with 0.5% NaClO, washed several times with deionized water and germinated at 28°C for 48 h before sowing. Subsequently, they were seeded into sterilized peat-based seeding substrate on 8 March, 2007. At cross stage (with four leaves), the plantlets were transplanted into seedling pots (4 cm in width $\times$ 4 cm in length $\times$ 5 cm in depth). At the same time, each inoculated plant received 3 g AM fungi inocula and all uninoculated plants were treated with the same amount of inocula that had been autoclaved twice at 121°C for 30 min. After four weeks of growth, tobacco plants with eight leaves were carefully transplanted into

3 L plastic pots (one plant per pot) filled with 2.8 kg of dry soil. Pots were arranged in randomized complete block design with three replicates per treatment. A total of 15 plants (pots) were involved. Plants were grown in a sunlit greenhouse with natural light, a day/night temperature 33°C/22°C, and a relative humidity 40%–60%. Plants were watered to maintain soil moisture at 60%–70% of water holding capacity by adding deionized water during the experimental period. No fertilizers were applied during the study period.

Roots, stalks and leaves were harvested separately after 13 weeks of growth. The taproot and fibrous roots of tobacco extended throughout the soil of the whole pots. The soil adhering to the root segment after a gentle shake was considered to be rhizosphere soil. The soil was thoroughly mixed after roots were removed at harvest and 100 g fresh soil was taken from each pot to air-dry for chemical analyses.

1.4 Plant biomass

Subsamples of fresh roots were taken to assess mycorrhizal colonization. Fresh weights of total roots and of sub-samples were measured. Stalks, leaves and remaining roots were rinsed with tap water and then with deionized water. Tissues were weighed after oven drying at 60°C for 72 h and then ground to < 0.25 mm in a stainless mill. The percentage of water content in remaining roots and total root fresh weight were used to estimate total root dry weight.

1.5 Mycorrhizal colonization rate

Root mycorrhizal colonization was estimated after clearing and staining (Koske and Gemma, 1989), using the grid-line intersect method (Giovannetti and Mosse, 1980). The stained roots were then mounted on glass slides (10 pieces of root per slide) for examination with an eyepiece cross-hair. Colonization percentage of mycorrhiza was estimated for each sample by examination of one hundred 1-cm long pieces of roots.

1.6 Chemical analyses

Dried sub-samples of roots, stalks and leaves were digested by concentrated HNO_3 and HCl at 3:1 (*V/V*) to analyze As. To determine P concentration, samples were digested by concentrated H_2SO_4 and 30% H_2O_2 and measured by inductively coupled plasma atomic emission spectrometry (ICP-AES, IRIS Advantage, Thermo, USA). The accuracy of the analyses was estimated by comparison with reference material GBW07603 from the Institute of Geophysical & Geochemical Exploration, Chinese Academy of Geosciences, and blanks were introduced regularly.

In order to test the water-extractable As concentrations in the rhizosphere, soil was air dried and ground to the maximum particle size of 2 mm and shaken in plastic flasks with deionised water (1:5, *m/V*) for 2 h at 20°C. Then, extracts were filtered through ash-free paper and As concentration was determined. Arsenic fractions in rhizosphere soils were extracted through the following sequential extraction procedure (SEP) (Wenzel *et al*., 2001): F1, 0.05 mol/L $(NH_4)_2SO_4$, 20°C, 4 h shaking; F2, 0.05 mol/L $NH_4H_2PO_4$, 20°C, 16 h shaking; F3, 0.2 mol/L NH_4^+-oxalate buffer, pH 3.25, 20°C, 4 h shaking in the dark; F4, 0.2 mol/L NH_4^+-oxalate buffer + ascorbic acid, pH 3.25, 0.5 h in a water basin at (96 ± 3)°C in the light. These As fractions appear to be F1, nonspecifically sorbed; F2, specifically-sorbed; F3, bound to amorphous and poorly-crystalline hydrous oxides of Fe and Al; F4, bound to well-crystallized hydrous oxides of Fe and Al. Soil pH was examined using a microprocessor pH meter (model pH211, Hanan Instruments, Padua, Italy).

1.7 Data analysis

All data were analysed by a one-way analysis of variance (ANOVA) with treatment as factor. Mean separation was conducted based on Duncan's multiple range test, and differences at $p < 0.05$ were considered statistically significant. All statistical analyses were performed by SPSS 12.0 for Windows.

2 Results

2.1 Mycorrhizal colonization rate

No root infection was found in control plants, and the mean proportion of root length colonized in inoculated plants ranged from 5% to 23%. Isolate 40 resulted in the highest ($p < 0.05$) mycorrhizal colonization rate (Fig. 1).

2.2 Plant biomass

There were no significant differences in roots and stalks dry weights among all treatments. While, leaves and total plant dry weights were much higher ($p < 0.05$) in treatments inoculated with isolate 90001 than in control treatments (Table 1).

2.3 pH, water-extractable As and As fractionation concentrations in rhizosphere soil

Soil pH and water-extractable As concentrations were significantly higher ($p < 0.05$) in uninoculated treatment than in inoculated treatments. No marked differences in the As amount of F1, F2 and F3 were observed. For F4, the concentration was significantly higher in mycorrhizal than in control treatment (Table 2).

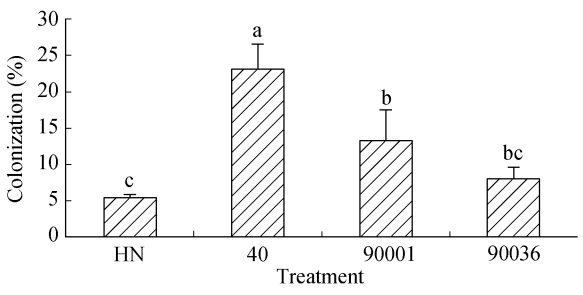

Fig. 1 Mycorrhizal colonization rate of tobacco plants in mycorrhizal treatments. HN: *Glomus* spp. and *Acaulospora* spp.; 40: *Acaulospora mellea*; 90001: *Glomus versiforme*; 90036: *Glomus caledonium*. Bars represent standard deviation and different letters indicate significant differences by Duncan's multiple range test at $p < 0.05$, $n = 3$.

Table 1 Root, stalk and leaf dry weights of tobacco plants in response to different treatments

Treatment	Dry weight (g)			
	Root	Stalk	Leaf	Total
CK	3.21 ± 0.27 a	7.19 ± 0.22 a	13.18 ± 0.71 b	23.58 ± 0.36 b
HN	2.79 ± 0.08 a	7.68 ± 0.24 a	13.64 ± 0.88 b	24.10 ± 1.01 ab
40	2.85 ± 0.21 a	7.17 ± 0.59 a	13.90 ± 0.30 ab	23.92 ± 0.84 ab
90001	3.09 ± 0.27 a	6.61 ± 0.36 a	15.57 ± 0.67 a	25.27 ± 0.43 a
90036	3.01 ± 0.44 a	7.48 ± 0.40 a	14.08 ± 1.14 ab	24.57 ± 1.10 ab

CK: without inoculation.
HN, 40, 90001, and 90036 are the same as in Fig. 1.
Data are presented in mean ± standard deviation.
Different letters in the same column show significant differences by Duncan's multiple range test at $p < 0.05$, $n = 3$.

2.4 As and P uptake

As concentrations were greater in roots than in leaves and stalks for all treatments (Fig. 2). Inoculation with all four isolates resulted in significantly lower ($p < 0.05$) As concentrations of roots and leaves, except stalks. Roots, leaves and total plant As content exhibited the same trend (Table 3).

Moreover, there were no difference in As uptake between plants inoculated with AM fungi from polluted soils and those inoculated with isolates from unpolluted soils.

The P concentrations in tobacco decreased in the following order: leaves > stalks > roots. No significant difference was found in P concentrations of roots, stalks and leaves among all treatments (Fig. 2), so did in P content of stalks, leaves and total plant (Table 3). However, roots P content was remarkably lower in HN treatments than in other treatments (Table 3).

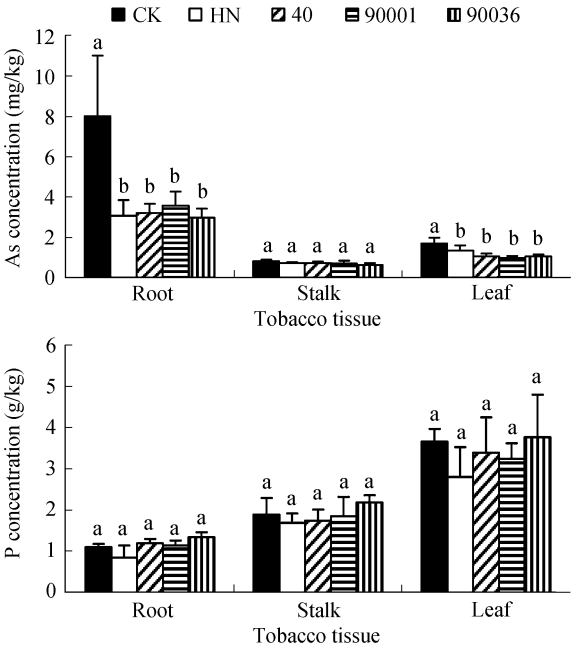

Fig. 2 As and P concentrations in tobacco plants in response to different treatments. Bars represent standard deviation and different letters indicate significant differences by Duncan's multiple range test at $p < 0.05$, $n = 3$.

3 Discussion

Recently, comparison of AM fungi from contaminated soils versus those from unpolluted soils has received increased attentions. Several publications reported the great efficacy of AM fungi from contaminated soils and high

Table 2 pH and water-extractable As and As fractionation concentrations in rhizosphere soil in response to different treatments

Treatment	Soil pH	Water-extractable As concentrations (mg/kg)	F1 (mg/kg)	F2 (mg/kg)	F3 (mg/kg)	F4 (mg/kg)
CK	6.68 ± 0.12 a	0.30 ± 0.02 a	0.04 ± 0.00 a	3.51 ± 0.09 a	6.63 ± 0.10 a	10.38 ± 0.20 c
HN	6.30 ± 0.07 b	0.24 ± 0.00 b	0.05 ± 0.01 a	3.80 ± 0.18 a	6.30 ± 0.29 a	12.72 ± 1.32 b
40	6.30 ± 0.09 b	0.24 ± 0.01 b	0.04 ± 0.00 a	3.58 ± 0.21 a	6.31 ± 0.14 a	14.79 ± 0.37 a
90001	6.23 ± 0.07 b	0.21 ± 0.02 c	0.03 ± 0.00 a	3.51 ± 0.14 a	6.40 ± 0.36 a	15.59 ± 1.73 a
90036	6.33 ± 0.04 b	0.24 ± 0.00 b	0.04 ± 0.00 a	4.01 ± 0.63 a	6.72 ± 0.09 a	13.71 ± 0.51 ab

F1: non-specifically sorbed; F2: specifically sorbed; F3: bound to amorphous and poorly-crystalline hydrous oxides of Fe and Al; F4: bound to well-crystallized hydrous oxides of Fe and Al.
Data are presented in mean ± standard deviation.
Different letters in the same column show significant differences by Duncan's multiple range test at $p < 0.05$, $n = 3$.

Table 3 As and P content in tobacco plants in response to different treatments

Treatment	As content (μg)				P content (mg)			
	Root	Stalk	Leaf	Total	Root	Stalk	Leaf	Total
CK	25.55 ± 8.68 a	5.75 ± 0.67 a	22.12 ± 2.94 a	53.42 ± 6.38 a	3.53 ± 0.34 a	13.52 ± 2.49 a	48.39 ± 6.63 a	65.45 ± 8.22 a
HN	8.49 ± 2.20 b	5.48 ± 0.53 a	18.03 ± 3.26 b	32.01 ± 1.77 b	2.34 ± 0.85 b	13.02 ± 1.93 a	38.55 ± 11.84 a	53.91 ± 13.79 a
40	9.12 ± 1.65 b	5.07 ± 0.92 a	14.33 ± 1.87 b	28.52 ± 3.27 b	3.39 ± 0.17 a	12.55 ± 2.87 a	47.21 ± 12.75 a	63.16 ± 15.31 a
90001	11.07 ± 2.85 b	4.49 ± 0.87 a	15.26 ± 0.85 b	30.82 ± 2.79 b	3.52 ± 0.24 a	12.30 ± 3.33 a	50.27 ± 4.03 a	66.09 ± 7.56 a
90036	8.84 ± 1.17 b	4.72 ± 0.84 a	14.60 ± 0.13 b	28.15 ± 1.02 b	4.01 ± 0.59 a	16.38 ± 1.61 a	52.42 ± 11.31 a	72.82 ± 12.26 a

Data are presented in mean ± standard deviation. Different letters in the same column show significant differences by Duncan's multiple range test at $p < 0.05$, $n = 3$.

sensitivity of isolates from unpolluted soils not adapted to stress of contaminated soils (Bai *et al*., 2008; Wang *et al*., 2005; Weissenhorn *et al*., 1994). However, there were no obvious difference in the effect of an isolate on the tobacco growth and As uptake among all tested AM fungi observed in this experiment. Inoculation with all four AM fungi resulted in lower As concentrations in leaves. Similarly, Koomen *et al*. (1987) found that indigenous AM fungi did not have a significant effect based on mycorrhizal growth response in their original soils. Janoušková *et al*. (2007) reported no relationship between the effect of an isolate on Cd uptake by tobacco and its origin. The probable explanation, as stated by Enkhtuya *et al*. (2000), is that isolates from contaminated soils may lose their adaptation and tolerance to the original stress, while those from unpolluted soils presented great plasticity and adaptation to the stress.

Decreased As concentrations in mycorrhizal plants compared with non-mycorrhizal plants have been well documented (Bai *et al*., 2008; Trotta *et al*., 2006; Weissenhorn *et al*., 1995; Xu *et al*., 2008). It has been suggested that the mechanism could be a biomass dilution effect as a result of improved growth due to enhanced P nutrition (Chen *et al*., 2007; Dong *et al*., 2008; Smith and Read, 1997). However, no significant difference was observed in P concentration and dry weights (except leaves biomass of 90001-inoculated plants) between inoculated and uninoculated plants. Thus, dilution effect cannot be assumed in our study. AM fungi are able to increase the surface area and absorption zone of mycorrhizal roots and thus improve nutrient uptake, especially P, thus improving plant growth (Janoušková *et al*., 2005b; Wang *et al*., 2005). The mycorrhizal growth response depends on many factors, such as heavy metals/metalloids and P concentrations in soil, AM fungal and plant species, and soil properties (Bai *et al*., 2008; Janoušková *et al*., 2007; Wang *et al*., 2005; Xia *et al*., 2007). Our results may be explained by high soil available P concentrations. Son and Smith (1988) reported that growth responses of plants to mycorrhizal colonization were found positively on P-deficient soils, but negatively in conditions of high P supply. On one hand, it may be more efficient for plant to take up soil P directly than to take it up via the fungus when there is a ready supply of soil P (Jakobsen *et al*., 2002). On the other hand, as summarized by Xu *et al*. (2008), the more P nutrition supplied, the more strong transcription of the phosphate transporter may be suppressed. Moreover, P and As are chemical analogues. These two elements show similar behavior, e.g., both compete for the same adsorption sites on soil particles (Adriano, 2001), and are transported across the plasma membrane via P transporters systems (Ullrich-Eberius *et al*., 1989). Thus, under the stress of As, AM fungi may regulate P uptake – no more P was accumulated – so as to downregulate As transport into its host. Similarly, Meharg and Macnair (1992) found that suppression of high affinity P uptake through downregulating phosphate/arsenate transporters (Gonzalez-Chavez *et al*., 2002) could assist plants to enhance As tolerance.

One possible mechanism for our results could be an induction of changes in the chemical behavior of elements in soil by AM fungi. Apparently, metals/metalloid uptake in plants is highly correlated with their solubility in soil. The possibility was tested by determining the concentrations of water-extractable As and As fractions at the end of this experiment. Overall, significantly lower water-extractable As concentrations were detected in mycorrhizal treatments compared with non-mycorrhizal treatments. Several mechanisms may be involved in the changes of metals/metalloids availability in mycorrhizal treatment. Using solution culture, Denny and Ridge (1995) found that the ericaceous mycorrhizal fungi could produce slimes to bind abundant Zn, and then Zn concentration in solution declined. Additionally, AM fungi seemed to be involved in the transformation of inorganic As into less toxic organic forms which consequently resulted in a lower As uptake by sunflower plants (Ultra *et al*., 2007a, 2007b). Among all soil properties, pH is one of paramount important factors which could influence metals/metalloids solubility in soil (EI-Kherbawy *et al*., 1989; Navarro *et al*., 2006; Xia *et al*., 2007). A small rise of soil pH resulting from the activities of mycorrhizal hyphae could bring about a substantial decline in Zn concentration in the soil solution (Li and Christie, 2001). In the present experiment, pH in rhizosphere was lower in mycorrhizal treatments than in non-mycorrhizal treatments. This was consistent with the report of Li *et al*. (1991), who found decreased soil pH at the root-soil interface, in the hyphal compartment and the hyphae-soil interface. The result may be attributed to the activities of mycorrhizal hyphae, as their presence was the only difference between mycorrhizal and the corresponding non-mycorrhizal treatments. Research has shown that soil pH is changed by the activity of mycorrhizal hyphae through modifying the amount and composition of root exudation (Leyval *et al*., 1997; Li and Christie, 2001; Li *et al*., 1991; Meharg, 2003). The decline in pH corresponds with a decline in solution As concentration in mycorrhizal treatments. As Masscheleyn *et al*. (1991) found, an alkaline pH released substantial proportions of arsenic into solution. Navarro *et al*. (2006) suggested As bioavailability declined when soil pH decreased, likely due to its adsorption onto Fe oxide surfaces (Liu *et al*., 2001). In this study, more As fraction bound to well-crystallized hydrous oxides of Fe and Al (F4) that is significantly less bioavailable (Fitz and Wenzel, 2002), was detected in mycorrhizal treatments than in control. Likewise, increased F4-As with inoculated treatments was observed by Bai *et al*. (2008) at high soil As levels. Our results indicated that the protective effect of mycorrhiza could be linked to the change in As availability resulting from the variation of soil pH that have been induced in turn by the activities of mycrorrhizal hyphae in soil.

4 Conclusions

It seems that when grown in symbiosis with tobacco in the As polluted soil, AM fungi from polluted soils were no more effective than those from uncontaminated soils. The decreased As uptake in mycorrhizal plants could be

associated with the decline of As availability resulting from the decrease in soil pH caused by the AM fungi. All these results indicated that, proper mechanisms would be employed by AM fungi to benefit plants to restrict As uptake. Furthermore, experiments under field conditions should be performed to study the extent to which mycorrhizal fungi can alleviate As plant toxicity.

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