

Contributions of arbuscular mycorrhizal fungi to growth, photosynthesis, root morphology and ionic balance of citrus seedlings under salt stress

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Received: 3 March 2009 / Revised: 22 July 2009 / Accepted: 20 October 2009 / Published online: 4 November 2009
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Abstract A pot study was conducted to determine the effects of arbuscular mycorrhizal (AM) fungi (*Glomus mosseae* and *Paraglomus occultum*) and salt (NaCl) stress on growth, photosynthesis, root morphology and ionic balance of citrus (*Citrus tangerine* Hort. ex Tanaka) seedlings. Eighty-five-day-old seedlings were exposed to 100 mM NaCl for 60 days to induce salt stress. Mycorrhizal colonization of citrus seedlings was not affected by salinity when associated with *P. occultum*, but significantly decreased when with *G. mosseae*. Compared with the non-mycorrhizal controls, mycorrhizal seedlings generally had greater plant height, stem diameter, shoot, root and total plant biomass, photosynthetic rate, transpiration rate and stomatal conductance under the 0 and 100 mM NaCl stresses. Root length, root projected area and root surface area were also higher in the mycorrhizal than in the non-mycorrhizal seedlings, but higher root volume in seedlings with *G. mosseae*. Leaf Na^+ concentrations were significantly decreased, but leaf K^+ and Mg^{2+} concentrations and the K^+/Na^+ ratio were increased when seedlings with both *G. mosseae* and *P. occultum*. Under the salt stress, Na^+ concentrations were increased but K^+ concentrations decreased in the mycorrhizal seedlings. Under the salt stress, Ca^{2+} concentrations were increased in the seedlings

with *P. occultum* or without AM fungi (AMF), but decreased with *G. mosseae*. Ratios of both $\text{Ca}^{2+}/\text{Na}^+$ and $\text{Mg}^{2+}/\text{Na}^+$ were also increased in seedlings with *G. mosseae* under the non-salinity stress, while only the $\text{Mg}^{2+}/\text{Na}^+$ ratio was increased in seedlings with *P. occultum* under the salt stress. Our results suggested that salt tolerance of citrus seedlings could be enhanced by associated AMF with better plant growth, root morphology, photosynthesis and ionic balance.

Keywords Arbuscular mycorrhiza · Citrus · Ionic balance · Photosynthesis · Root morphology · Salt stress

Abbreviations

AM	Arbuscular mycorrhiza
AMF	Arbuscular mycorrhizal fungi
<i>E</i>	Transpiration rate
g_s	Stomatal conductance
Pn	Photosynthetic rate

Introduction

Salinity adversely affects plant water-holding capacity and ionic imbalance by osmotic stress and/or ion cytotoxicity (mainly Na^+ and Cl^-) to plant cell and functioning, leading to significant decreases of plant productivity and quality (van Hoorn et al. 2001; Chinnusamy et al. 2005; Teakle et al. 2006). It is known that arbuscular mycorrhiza (AM), a mutualistic association between AM fungi (AMF) and roots of most terrestrial plants, could enhance plant growth and alleviate salt stress (Ruiz-Lozano et al. 1996;

Communicated by W. Filek.

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Al-Karaki 2000; Asghari et al. 2005; Asghari 2008). The possible mechanisms of mycorrhizal plants to salt tolerance include: (1) improving plant nutrient uptake, especially P (Al-Karaki 2000; Al-Karaki et al. 2001; Asghari et al. 2005; Asghari 2008), (2) altering root plasticity (Echeverria et al. 2008), (3) providing higher accumulation of soluble sugars in mycorrhizal roots (Feng et al. 2002), (4) elevating K/Na ratio (Giri et al. 2007; Asghari 2008), and (5) maintaining higher antioxidant enzymatic activities (He et al. 2007). AMF have been thus considered as important bio-ameliorators for saline soils (Rabie and Almadini 2005).

Citrus trees are relatively sensitive to salinity stress (Al-Yassin 2004; Levy and Syvertsen 2004). Their growth and fruit yield were impaired under soil salinity without any obviously visible symptoms in leaves (Cerdeira et al. 1990). *Glomus intraradices*-inoculated Carrizo citrange [*Citrus sinensis* (L.) Osb. × *Poncirus trifoliata* (L.) Raf.] had higher dry weight and lower proline content than the uninoculated controls under either 25, 50 or 100 mM NaCl stress (Duke et al. 1986). However, AM colonization was decreased from 62 to 67% in the control to 31–40% at 150 mM NaCl stress, though no significant physiological effects were derived from such colonization decreases to 3-month-old Karna Khatta (*C. karna*) and Troyer Citrange (*Poncirus trifoliata* × *C. sinensis*) seedlings (Murkute et al. 2006). As a result, further study on interactions between citrus and AMF under saline conditions is required.

Plant salt tolerance is related to its ability to regulate ionic balance, particularly Na^+ , K^+ , Ca^{2+} and Mg^{2+} (Munns et al. 2006). Roots are the site of carbohydrate and nutrient exchange between AMF and host plants, while their function of absorption is closely related to their morphology (Yi et al. 2007). To date, little is known about the interaction between AMF and salinity on ionic balance and root morphology of plants. The objective of this research was to evaluate the effects of two AMF on growth performance, ionic balance, photosynthesis and root morphology of red tangerine (*C. tangerine* Hort. ex Tanaka) seedlings grown under salinity.

Materials and methods

Experiment design

This experiment was conducted inside a greenhouse between March and July 2008 at Yangtze University, Jingzhou City, Hubei Province, China. The experiment consisted of a randomized block design with two factors: three mycorrhizal treatments [*G. mosseae* (Nicol. & Gerd.) Gerdemann & Trappe, *Paraglomus occulta* (Walker

Morton & Redecker and non-AMF control] and two salinity levels of 0 and 100 mM NaCl. Each treatment had three replicates (pots) for a total of 18 pots (three seedlings per pot).

Plant culture and salt stress induction

Seeds of red tangerine were surface sterilized with 70% alcohol for 5 min, rinsed four times with distilled water and sowed into plastic pots (16 cm in depth and 20 cm in mouth diameter) that were filled with 3.4 kg of autoclaved soil mixture. Either 15 g (dry wt) mycorrhizal (*G. mosseae* or *P. occulta*) inoculum (soil, spores, hyphae and root material) or autoclaved inoculum was placed at 5 cm depth of soil mixture. These mycorrhizal inocula were from the Institute of Plant Nutrition and Resources, Beijing Academy of Agriculture and Forestry Sciences, China. Originally, *G. mosseae* was isolated from *Incarvillea yonghusbandii* in a non-saline rhizosphere soil in Sitsang, and *P. occulta* from *Prunus* L. in a saline rhizosphere soil in Beijing, China.

Salinity stress was induced by adding 300 ml of 100 mM NaCl solution after 85 days of sowing, and the control (0 mM NaCl) seedlings were irrigated with 300 ml of distilled water. To avoid osmotic shock, the soil was gradually salinized by 25 mM NaCl per day. Seedlings were harvested 60 days after salinity stress.

Growth and ion characters

Plant height and stem diameter were measured after 145 days of growth. Shoot and root systems were oven-dried at 75°C, and ground into 0.5 mm powder. Approximately 0.1 g dry powder of leaf or root was incubated in 25 ml deionized water at 100°C for 2 h, cooled at room temperature for 30 min and then filtered. Concentrations of K^+ , Na^+ , Ca^{2+} or Mg^{2+} were determined directly with an Atomic Absorption Spectrometer (AI 1200, Aurora Instruments Limited, Canada).

Photosynthesis measurement

Photosynthetic rate (P_n), transpiration rate (E) and stomatal conductance (g_s) were measured by an infrared gas analyzer (Li-6400, Li-Cor, Lincoln, NE, USA) on three replications per treatment from 9:30 to 10:40 am at a sunny day before harvest. Measurements were recorded when the total coefficient of variation was less than 0.5%.

Symbiotic development

One-cm root pieces were taken from the middle part of the root systems, were cleaned with 10% (w/v) KOH and

stained with 0.05% (w/v) trypan blue in lactophenol (Phillips and Hayman 1970). The AM colonization was quantified according to the following formula (Wu et al. 2008):

$$\text{AM colonization (\%)} = \frac{\text{root length infected}}{\text{root length observed}} \times 100$$

Root morphology analysis

The fresh root systems were carefully washed by tap water after harvest and were directly placed on the Regent's water-proof trays. The image of the root system was acquired using an extra optimized Epson Expression/STD 4800 scanner and analyzed with the WinRHIZO software using an automatic global thresholding method (Regent Instruments Inc., Quebec, QC, Canada). It is an automatic and interactive image analysis system specifically designed for root morphological traits including root length, projected area, surface area, average diameter and volume. Roots of six seedlings (two roots/pot) were scanned for each treatment and data were the average of these six samples.

Statistical analysis

Data (means) were analyzed using ANOVA (SAS, Version 8.1) at $P < 0.05$ (Fisher's protected least significant difference). Percentage values were arcsine transformed before statistical analyses.

Results

AM colonization with *G. mosseae*, but not with *P. occultum*, was significantly decreased under 100 mM NaCl salinity (Fig. 1). Salinity reduced stem diameter, plant height and biomass of citrus seedlings, irrespective of the inoculation with *G. mosseae* or *P. occultum* (Table 1). Under the non-salinity stress, compared to the non-AM control seedlings, stem diameter, plant height, shoot, root and total dry weights were increased by 15, 11, 24, 20 and 25% in *G. mosseae*-inoculated seedlings, and by 7, 7, 28, 27 and 28% in *P. occultum*-inoculated seedlings, respectively. Under the salinity stress, compared to the non-AM control seedlings, stem diameter, plant height, shoot, root and total dry weight in *G. mosseae*-inoculated seedlings were increased by 14, 6, 37, 33 and 31%, and in *P. occultum*-inoculated seedlings by 15, 9, 37, 33 and 34%, respectively.

Soil salinity markedly decreased their P_n and E in both the *G. mosseae*-inoculated and the non-AM seedlings, but not in *P. occultum*-inoculated seedlings (Table 2). Soil salinity decreased their g_s in all seedlings.

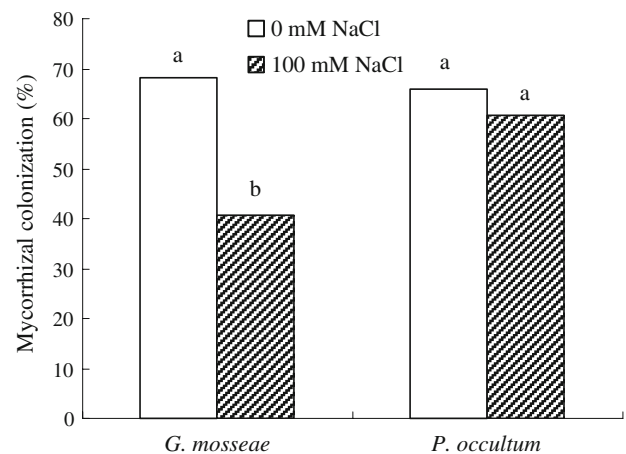


Fig. 1 Root AM colonization of *Citrus tangerine* seedlings under 0 and 100 mM NaCl

Mycorrhizal seedlings had higher P_n , E and g_s than the un-inoculated seedlings under both 0 and 100 mM NaCl salinity levels.

Salinity decreased root length, root projected area, root surface area and root volume in the AM and the non-AM seedlings (Table 3). Root average diameter was not affected by both salinity and AMF inoculation. Plants inoculated with AMF had significantly higher root length, root projected area and root surface area than the control seedlings grown under both 0 and 100 mM NaCl levels, but no differences in these parameters between *G. mosseae*- and *P. occultum*-inoculated seedlings. At both NaCl levels, root volume was significantly increased with *G. mosseae*-inoculated seedlings, but not with *P. occultum*-inoculated seedlings.

Compared to the non-salinity 0 mM NaCl control, Na^+ but not K^+ concentrations were significantly increased under 100 mM NaCl (Table 4). Compared to the non-salinity level, Ca^{2+} concentration was significantly decreased under 100 mM NaCl salinity level in the *G. mosseae*-colonized seedlings, but not in the *P. occultum*-colonized and the non-AM seedlings. Mg^{2+} concentrations were increased by salinity only in the *P. occultum*-inoculated seedlings. Under the non-salinity and salinity conditions, AM symbiosis notably decreased the Na^+ concentration but increased the K^+ and Mg^{2+} concentrations. A significant difference was only observed in the Ca^{2+} concentration between the *G. mosseae*-colonized and the non-AM seedlings under the 100 mM NaCl level.

Salinity significantly decreased the ratio of K^+/Na^+ , $\text{Ca}^{2+}/\text{Na}^+$ or $\text{Mg}^{2+}/\text{Na}^+$ (Table 4). Leaves of AM seedlings had higher K^+/Na^+ and $\text{Mg}^{2+}/\text{Na}^+$ ratios than the non-AM-inoculated control seedlings at both salinity levels, except the *G. mosseae*-colonized seedlings grown under 100 mM NaCl. In addition, only *G. mosseae*

Table 1 Effects of salinity and mycorrhizal associations on growth of *Citrus tangerine* seedlings

Salinity (mM)	AMF status	Stem diameter (mm)	Plant height (cm)	Dry weight (g/plant)		
				Shoot	Root	Total
0	<i>G. mosseae</i>	0.240a	11.9a	0.31ab	0.18a	0.49a
	<i>P. occultum</i>	0.223b	11.5a	0.32a	0.19a	0.51a
	Non-AMF	0.208c	10.7b	0.25c	0.15b	0.40b
100	<i>G. mosseae</i>	0.207c	10.4b	0.26c	0.12b	0.38b
	<i>P. occultum</i>	0.208c	10.7b	0.26bc	0.12b	0.39b
	Non-AMF	0.181d	9.8c	0.19d	0.09c	0.28c
Significance						
AMF		**	**	**	**	**
Salt stress		**	**	**	**	**
AMF \times salt stress		NS	NS	NS	NS	NS

Same letter within each column indicates no significant difference among treatments ($P < 0.05$)

NS not significant

** $P < 0.01$

Table 2 Effects of salinity and mycorrhizal associations on photosynthesis of *Citrus tangerine* seedlings

Salinity (mM)	AMF status	Pn ($\mu\text{mol m}^{-2} \text{s}^{-1}$)	E ($\text{mmol m}^{-2} \text{s}^{-1}$)	g_s ($\text{mmol m}^{-2} \text{s}^{-1}$)
0	<i>G. mosseae</i>	18.16a	1.69a	0.0435a
	<i>P. occultum</i>	16.58ab	1.60ab	0.0415a
	Non-AMF	12.36c	1.30c	0.0355bc
100	<i>G. mosseae</i>	13.83c	1.38bc	0.0333c
	<i>P. occultum</i>	14.51bc	1.44bc	0.0365b
	Non-AMF	4.79d	0.25d	0.0061d
Significance				
AMF		**	**	**
Salt stress		**	**	**
AMF \times salt stress		*	**	**

Same letter within each column indicates no significant difference among treatments ($P < 0.05$)

NS not significant

* $P < 0.05$, ** $P < 0.01$

inoculation notably increased $\text{Ca}^{2+}/\text{Na}^{+}$ ratio, compared with the non-inoculation and non-stressed condition.

Discussion

Our results showed that salt stress reduced colonization of *G. mosseae*-inoculated red tangerine seedlings. This is consistent with other observations in citrus (Duke et al. 1986; Murkute et al. 2006). The decline in AM colonization was possibly from the strong inhibition of salt to hyphal growth (Juniper and Abbott 2006). However, root colonization by *P. occultum* was not affected by soil salinity in our study. Hartmond et al. (1987) also showed

that salinity stress did not affect colonization by *G. intraradices* in the sweet orange and the sour orange. Different responses of AMF to salinity stress in this experiment may derive from different fungal origin, as the spore of *P. occultum* was originated from a saline soil, which was more tolerant to salinity than the spore of *G. mosseae* from a non-saline soil (Carvalho et al. 2004).

Salinity generally inhibits plant growth due to water-deficit and salt-excess effects (Munns et al. 2006). Salt tolerance has usually been assessed as the biomass production (da Silva et al. 2008). Citrus seedlings inoculated with *G. mosseae* or *P. occultum* in this study had higher stem diameter, plant height, shoot, root and total dry weights than the non-AM seedlings under 100 mM NaCl

Table 3 Effects of salinity and mycorrhizal associations on root morphology of *Citrus tangerine* seedlings

Salinity (mM)	AMF status	Root length (cm)	Root projected area (cm ²)	Root surface area (cm ²)	Root average diameter (mm)	Root volume (cm ³)
0	<i>G. mosseae</i>	220.91a	12.07a	37.92a	0.55a	0.52a
	<i>P. occultum</i>	217.02ab	10.95ab	34.39ab	0.51a	0.44b
	Non-AMF	175.31c	9.59c	30.13c	0.55a	0.41bc
100	<i>G. mosseae</i>	187.80c	9.71bc	30.50bc	0.53a	0.40bc
	<i>P. occultum</i>	189.52bc	9.12c	28.64c	0.48a	0.35 cd
	Non-AMF	124.63d	6.58d	20.69d	0.54a	0.28d
Significance						
AMF		**	**	**	NS	**
Salt stress		**	**	**	NS	**
AMF × salt stress		NS	NS	NS	NS	NS

Same letter within each column indicates no significant difference among treatments ($P < 0.05$)

NS not significant

** $P < 0.01$

Table 4 Effects of salinity and mycorrhizal associations on ionic status of leaves of *Citrus tangerine* seedlings

Salinity (mM)	AMF status	Na ⁺ (mg/g)	K ⁺ (mg/g)	Ca ²⁺ (mg/g)	Mg ²⁺ (mg/g)	K ⁺ /Na ⁺	Ca ²⁺ /Na ⁺	Mg ²⁺ /Na ⁺
0	<i>G. mosseae</i>	7.96d	22.93a	11.70b	4.69bc	2.88a	1.47a	0.59a
	<i>P. occultum</i>	7.93d	22.39a	10.71bc	4.65bc	2.83a	1.35ab	0.59a
	Non-AMF	8.78c	20.89b	10.77bc	4.30d	2.38b	1.23b	0.49b
100	<i>G. mosseae</i>	15.17b	19.14b	10.09c	4.77b	1.26c	0.67d	0.31cd
	<i>P. occultum</i>	14.98b	20.73b	13.88a	5.28a	1.38c	0.93c	0.35c
	Non-AMF	16.59a	16.93c	13.29a	4.38cd	1.02d	0.80cd	0.26d
Significance								
AMF		**	**	*	**	**	NS	**
Salt stress		**	**	**	*	**	**	**
AMF × salt stress		NS	NS	**	NS	NS	**	NS

Same letter within each column indicates no significant difference among treatments ($P < 0.05$)

NS not significant

* $P < 0.05$, ** $P < 0.01$

level, indicating an enhancement of salt tolerance by mycorrhizal plants. These results are in agreement with previous studies on onion (Hirrel and Gerdemann 1980; Ojala et al. 1983), pepper (Hirrel and Gerdemann 1980), maize (Feng et al. 2002; Sheng et al. 2008), lettuce (Jahromi et al. 2008) and tomato (Al-Karaki 2000). Improved growth of mycorrhizal plants in saline environments is partly related to mycorrhizal-mediated P enhancement in host plants (Hirrel and Gerdemann 1980; Ojala et al. 1983; Al-Karaki 2000).

Photosynthesis is one of the primary processes to be affected by salt stress (Chaves et al. 2009). A significant reduction in photosynthesis had been found in salt-stressed citrus (Iglesias et al. 2004). In the present study, salt stress also reduced the Pn, E and g_s in both non-AM-inoculated and AM-inoculated citrus seedlings. However, mycorrhizal

seedlings had significantly higher Pn, E and g_s than the non-mycorrhizal seedlings under salt stress (Table 2). This is consistent with Ruiz-Lozano et al. (1996) and Sheng et al. (2008) under salt stress. The salt tolerance of mycorrhizal seedlings might be increased, since higher photosynthetic capacity can theoretically increase water use efficiency as more carbon is assimilated per unit water transpired (Nandy et al. 2007). The increase of photosynthetic ability in mycorrhizal seedlings under salt stress could be resulted from a lower intercellular CO₂ concentration in mycorrhizal seedlings (Sheng et al. 2008).

Root system can interact with salt stress, and adjust its morphology and physiology to strengthen its absorption and survival (Yi et al. 2007). Correlation showed that better root morphology meant more salt-tolerant (Ashraf et al. 2005). Colonization by AMF could alter the morphology of

a root system in a structural, spatial, quantitative and temporal manner (Kapoor et al. 2008). In the present experiment, mycorrhizal citrus seedlings had greater root length, root projected area and root surface area than the non-mycorrhizal control seedlings under NaCl stress, which is consistent with studies with cherimoya, grapevine, apple, pepper, maize, zucchini and beach plum (Azcón-Aguilar et al. 1996; Locatelli et al. 2002; Aguin et al. 2004; Padilla and Encina 2005; Schroeder and Janos 2005; Zai et al. 2007). Better root systems in mycorrhizal plants would contribute greater salt tolerance. In addition, the improvement of AM root morphology is often attributed to a modified endogenous balance of growth regulators such as cytokinins and gibberellins (Berta et al. 1993), and a sizeable quantity of polysaccharides in root hairs (Zangaro et al. 2005).

Salt stress affects plant physiological traits through changes of ionic status in the cells (Hasegawa et al. 2000; Munns et al. 2006). Ionic imbalances in plants can occur in plant cells due to salt stress (Lutts et al. 1999). Such an imbalance could come from nutrient availability, competitive uptake, transport or partitioning within the plants (Rabie 2005). Our results showed a significant increase of Na^+ , but a significant decreases of K^+ in both the AM and non-AM seedlings, which are in agreement with Al-Karaki (2000) and Khan et al. (2000). Our study also showed that Na^+ concentrations were reduced, but K^+ and Mg^{2+} increased when citrus seedlings inoculated with AMF. Similarly, Na^+ was reduced but K^+ and Mg^{2+} were increased under salinity when *Sesbania*, *Acacia nilotica*, *Sesbania aegyptiaca* and *Sesbania grandiflora* inoculated with AMF (Giri and Mukerji 2004; Giri et al. 2007). Increased K^+ and Mg^{2+} concentrations in citrus seedlings by AM colonization under salinity would help the seedlings to prevent cellular Na^+ accumulation to a toxic level, thus protecting host plants against salinity stress.

Our study showed that AM colonization altered the ratio of K^+/Na^+ , $\text{Ca}^{2+}/\text{Na}^+$ or $\text{Mg}^{2+}/\text{Na}^+$. Mohammad et al. (2003), Rabie (2005) and Giri et al. (2007) also observed a high K^+/Na^+ ratio under salinity in AM-inoculated mungbean, barley or *A. nilotica*. Maintenance of a higher cytosolic K^+/Na^+ ratio is a key feature of plant salt tolerance (Wu and Zou 2009). Therefore, a higher K^+/Na^+ ratio may be essential for normal cellular functions of mycorrhizal plants in enhancing salt tolerance, since AM associations could help plants to compete with K^+ uptake, or induce high affinity K^+ transporters under salinity (Chinnusamy et al. 2005).

Ca^{2+} is a component of protein phosphorylation/dephosphorylation cascade(s) that couple salt stress perception and signal transduction to physiological mechanisms of adaptation by plants (Pardo et al. 1998). Our results showed that a higher $\text{Ca}^{2+}/\text{Na}^+$ ratio only occurred

in seedlings inoculated with *G. mosseae* in the non-stressed condition, and *P. occultum* inoculation did not affect the $\text{Ca}^{2+}/\text{Na}^+$ ratio under salinity stress. These results suggested that $\text{Ca}^{2+}/\text{Na}^+$ ratio may not be a good indicative index concerning the enhanced salt tolerance by AMF. A field experiment showed that the reduction in Na^+ uptake together with a related increase in Mg^{2+} absorption might be an important salt-alleviation mechanism for AM plants growing in saline soils (Giri and Mukerji 2004). Our results also showed that mycorrhizal colonization, except for *G. mosseae* under 100 mM NaCl, significantly increased the $\text{Mg}^{2+}/\text{Na}^+$ ratio because of the reduce of Na^+ and the increase of Mg^{2+} at the same time. This may potentially reduce the antagonistic effect of Na^+ , enhancing salt tolerance of mycorrhizal plants.

In summary, our results suggested that AM association could improve plant growth and root morphology, increase photosynthetic capacity, and reduce Na^+ concentrations for a better ionic balance in citrus seedlings. All these could lead to an enhanced salt tolerance by mycorrhizal seedlings. As a result, the application of AMF could make an important contribution to citrus production under salinity stress.

Acknowledgments The research was supported by a Doctoral Scientific Research Foundation (39210264) and an Undergraduate Research Program (091048923;02) from Yangtze University.

References

- Aguin O, Mansilla JP, Vilarino A, Sainz MJ (2004) Effects of mycorrhizal inoculation on root morphology and nursery production of three grapevine rootstocks. *Am J Enol Vitic* 55:108–111
- Al-Karaki GN (2000) Growth of mycorrhizal tomato and mineral acquisition under salt stress. *Mycorrhiza* 10:51–54
- Al-Karaki GN, Hammad R, Rusan M (2001) Response of two tomato cultivars differing in salt tolerance to inoculation with mycorrhizal fungi under salt stress. *Mycorrhiza* 11:43–47
- Al-Yassin A (2004) Influence of salinity on citrus: a review paper. *J Cent Eur Agric* 5:263–272
- Asghari HR (2008) Vesicular-arbuscular (VA) mycorrhizae improve salinity tolerance in pre-inoculation subterranean clover (*Trifolium subterraneum*) seedlings. *Int J Plant Product* 2:243–256
- Asghari HR, Marschner P, Smith SE, Smith FA (2005) Growth response of *Atriplex nummularia* to inoculation with arbuscular mycorrhizal fungi at different salinity levels. *Plant Soil* 273: 245–256
- Ashraf MY, Akhtar K, Sarwar G, Ashraf M (2005) Role of the rooting system in salt tolerance potential of different guar accessions. *Agron Sustain Dev* 25:243–249
- Azcón-Aguilar C, Padilla IMG, Encina CL, Azcón R, Barea MG (1996) Arbuscular mycorrhizal inoculation enhances plant growth and changes root system morphology in micropropagated *Annona cherimola* Mill. *Agronomie* 16:647–652
- Berta G, Fusconi A, Trotta A (1993) VA mycorrhizal infection and the morphology and function of root systems. *Environ Exp Bot* 33:159–173

- Carvalho LM, Correia PM, Martins-Loução MA (2004) Arbuscular mycorrhizal fungal propagules in a salt marsh. *Mycorrhiza* 14:165–170
- Cerda A, Nieves M, Guillen MG (1990) Salt tolerance of lemon trees as affected by rootstock. *Irrig Sci* 11:245–249
- Chaves MM, Flexas J, Pinheiro C (2009) Photosynthesis under drought and salt stress: regulation mechanisms from whole plant to cell. *Ann Bot* 103:551–560
- Chinnusamy V, Jagendorf A, Zhu J-K (2005) Understanding and improving salt tolerance in plants. *Crop Sci* 45:437–448
- da Silva EC, Nogueira RJMC, de Araújo FP, de Melo NF, de Azevedo Neto AD (2008) Physiological responses to salt stress in young umbu plants. *Environ Exp Bot* 63:147–157
- Duke ER, Johnson CR, Koch KE (1986) Accumulation of phosphorus, dry matter and betaine during NaCl stress of split-root citrus seedlings colonized with vesicular-arbuscular mycorrhizal fungi on zero, one or two halves. *New Phytol* 104:583–590
- Echeverria M, Scambato AA, Sannazzaro AI, Maiale S, Ruiz OA, Menéndez AB (2008) Phenotypic plasticity with respect to salt stress response by *Lotus glaber*: the role of its AM fungal and rhizobial symbionts. *Mycorrhiza* 18:317–329
- Feng G, Zhang FS, Li XL, Tian CY, Tang C, Rengel Z (2002) Improved tolerance of maize plants to salt stress by arbuscular mycorrhiza is related to higher accumulation of soluble sugars in roots. *Mycorrhiza* 12:185–190
- Giri B, Mukerji KG (2004) Mycorrhizal inoculant alleviates salt stress in *Sesbania aegyptiaca* and *Sesbania grandiflora* under field conditions: evidence for reduced sodium and improved magnesium uptake. *Mycorrhiza* 14:307–312
- Giri B, Kapoor R, Mukerji KG (2007) Improved tolerance of *Acacia nilotica* to salt stress by arbuscular mycorrhiza, *Glomus fasciculatum* may be partly related to elevated K/Na ratios in root and shoot tissues. *Microb Ecol* 54:753–760
- Hartmond U, Schaesberg NV, Graham JH, Syvertsen JP (1987) Salinity and flooding stress effects on mycorrhizal and non-mycorrhizal citrus rootstock seedlings. *Plant Soil* 104:37–43
- Hasegawa P, Bressan RA, Zhu JK, Bohnert HJ (2000) Plant cellular and molecular responses to high salinity. *Annu Rev Plant Physiol Plant Mol Biol* 51:463–499
- He ZQ, He CX, Zhang ZB, Zou ZR, Wang HS (2007) Changes in antioxidative enzymes and cell membrane osmosis in tomato colonized by arbuscular mycorrhizae under NaCl stress. *Colloids Surf B Biointerfaces* 59:128–133
- Hirrel MC, Gerdemann JW (1980) Improved growth of onion and bell pepper in saline soils by two vesicular-arbuscular mycorrhizal fungi. *Soil Sci Soc Am J* 44:654–658
- Iglesias DJ, Levy Y, Gómez-Cadenas A, Tadeo FR, Primo-Millo E, Talon M (2004) Nitrate improves growth in salt-stressed citrus seedlings through effects on photosynthetic activity and chloride accumulation. *Tree Physiol* 24:1027–1034
- Jahromi F, Aroca R, Porcel R, Ruiz-Lozano JM (2008) Influence of salinity on the in vitro development of *Glomus intraradices* and on the in vivo physiological and molecular responses of mycorrhizal lettuce plants. *Microb Ecol* 55:45–53
- Juniper S, Abbott LK (2006) Soil salinity delays germination and limits growth of hyphae from propagules of arbuscular mycorrhizal fungi. *Mycorrhiza* 15:371–379
- Kapoor R, Sharma D, Bhatnagar AK (2008) Arbuscular mycorrhizae in micropropagation systems and their potential applications. *Sci Hortic* 116:227–239
- Khan M, Ungar I, Showalters A (2000) Effects of salinity on growth water relation and ion accumulation of the subtropical perennial halophyte, *Atriplex griffithii* var. *stocksii*. *Ann Bot* 85:225–232
- Levy Y, Syvertsen J (2004) Irrigation water quality and salinity effects in citrus trees. *Hortic Rev* 30:37–82
- Locatelli LM, Vitovski CA, Lovato PE (2002) Root architecture of apple rootstocks inoculated with arbuscular mycorrhizal fungi. *Pesq Agropec Bras* 37:1239–1245
- Lutts S, Kinet JM, Bouharmont J (1999) Improvement of rice callus regeneration in the presence of NaCl. *Plant Cell Tissue Organ Cult* 57:3–11
- Mohammad MJ, Malkawi HI, Shibli R (2003) Effects of arbuscular mycorrhizal fungi and phosphorus fertilization on growth and nutrient uptake of barley grown on soils with different levels of salts. *J Plant Nutr* 26:125–137
- Munns R, James RA, Läuchli A (2006) Approaches to increasing the salt tolerance of wheat and other cereals. *J Exp Bot* 57:1025–1043
- Murkute AA, Sharma S, Singh SK (2006) Studies on salt stress tolerance of citrus rootstock genotypes with arbuscular mycorrhizal fungi. *Hortic Sci* 33:70–76
- Nandy P, Das S, Ghose M, Spooner-Hart R (2007) Effects of salinity on photosynthesis, leaf anatomy, ion accumulation and photosynthetic nitrogen use efficiency in five Indian mangroves. *Wetlands Ecol Manage* 15:347–357
- Ojala JC, Jarrell WM, Menge JA, Johnson ELV (1983) Influence of mycorrhizal fungi on the mineral nutrition and yield of onion in saline soil. *Agron J* 75:255–259
- Padilla IMG, Encina CL (2005) Changes in root morphology accompanying mycorrhizal alleviation of phosphorus deficiency in micropropagated *Annona cherimola* Mill. plants. *Sci Hortic* 106:360–369
- Pardo JM, Reddy MP, Yang S, Maggio A, Huh GH, Matsumoto T, Coca MA, Paino-D'Urzo M, Koiwa H, Yun DJ, Watad AA, Bressan RA, Hasegawa PM (1998) Stress signaling through Ca^{2+} /calmodulin-dependent protein phosphatase calcineurin mediates salt adaptation in plants. *Proc Natl Acad Sci USA* 95:9681–9686
- Phillips JM, Hayman DS (1970) Improved procedures for clearing roots and staining parasitic and vesicular-arbuscular mycorrhizal fungi for rapid assessment of infection. *Trans Br Mycol Soc* 55:158–161
- Rabie GH (2005) Influence of arbuscular mycorrhizal fungi and kinetin on the response of mungbean plants to irrigation with seawater. *Mycorrhiza* 15:225–230
- Rabie GH, Almadini AM (2005) Role of bioinoculants in development of salt-tolerance of *Vicia faba* plants under salinity stress. *Afr J Biotechnol* 4:210–222
- Ruiz-Lozano JM, Azcón R, Gomez M (1996) Alleviation of salt stress by arbuscular mycorrhizal *Glomus* species in *Lactuca sativa* plants. *Physiol Plant* 98:767–772
- Schroeder MS, Janos DP (2005) Plant growth, phosphorus nutrition, and root morphological responses to arbuscular mycorrhizas, phosphorus fertilization, and intraspecific density. *Mycorrhiza* 15:203–216
- Sheng M, Tang M, Chen H, Yang B, Zhang F, Huang Y (2008) Influence of arbuscular mycorrhizae on photosynthesis and water status of maize plants under salt stress. *Mycorrhiza* 18:287–296
- Teakle NL, Real D, Colmer TD (2006) Growth and ion relations in response to combined salinity and waterlogging in the perennial forage legumes *Lotus corniculatus* and *Lotus tenuis*. *Plant Soil* 289:369–383
- van Hoorn JW, Katerji N, Hamdy A, Mastorilli M (2001) Effect of salinity on yield and nitrogen uptake of four grain legumes and on biological nitrogen contribution from the soil. *Agric Water Manage* 51:87–98
- Wu QS, Zou YN (2009) Adaptive responses of birch-leaved pear (*Pyrus betulaeifolia*) seedlings to salinity stress. *Not Bot Hort Agrobot Cluj* 37:133–138
- Wu QS, Xia RX, Zou YN (2008) Improved soil structure and citrus growth after inoculation with three arbuscular mycorrhizal fungi under drought stress. *Eur J Soil Biol* 44:122–128

- Yi LP, Ma J, Li Y (2007) Impact of salt stress on the features and activities of root system for three desert halophyte species in their seedlings stage. *Sci China Ser D Earth Sci* 50(Suppl I): 97–106
- Zai XM, Qin P, Wan SW, Zhao FG, Wang G, Yan DL, Zhou J (2007) Effects of arbuscular mycorrhizal fungi on the rooting and growth of beach plum (*Prunus maritima*) cuttings. *J Hortic Sci Biotechnol* 82:863–866
- Zangaro W, Nishidate FR, Camargo FRS, Romagnoli GG, Vandressen J (2005) Relationships among arbuscular mycorrhizas, root morphology and seedlings growth of tropical native woody species in southern Brazil. *J Trop Ecol* 21:529–540