

## EFFECTS OF MYCORRHIZATION AND PHOSPHORUS NUTRITION ON NUTRIENT UPTAKE, GROWTH AND FLOWERING OF CHINA ASTER (*Callistephus chinensis* (L.) NEES) CULTIVATED ON EBB-AND-FLOW BENCHES

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### Abstract

The effects were investigated of phosphorus nutrition and AMF inoculation on nutritional status, growth, and flowering of China aster (*Callistephus chinensis* (L.) Nees) 'Milady' during cultivation on ebb-and-flow benches. Two P treatments of 8.68 and 43.40 mg x dm<sup>-3</sup> were applied. One month after inoculation the roots of inoculated plants were infected by mycorrhizal fungi. The control plants had no root infection. The mycorrhizal fungal colonization in plants that were fertilized at 8.68 and 43.40 mg x dm<sup>-3</sup> P was 67% and 60 %, respectively. Slightly increased P content was detected in leaves of mycorrhizal plants grown under low P level. Mycorrhization did not affect leaf P content of plants grown in high P level. Increased Mg content was measured in leaves of mycorrhizal plants grown under both P levels. Mycorrhizal and nonmycorrhizal plants did not differ with regard to leaf N, K, and Ca contents. P nutrition did not also affect the contents of these elements in leaf tissue. Mycorrhization decreased the pH and lowered salt accumulation in growing media. Significantly lower shoot biomass, plant height, shoot number were recorded in all plants inoculated with AMF. Mycorrhization also delayed flowering of China aster; the high P level slightly accelerated it. Mycorrhizal plants had fewer flower buds and flowers than nonmycorrhizal ones. The high P level increased the number of flowers of nonmycorrhizal plants only.

**Key words:** *Callistephus chinensis*, mycorrhization, phosphorus, nutrient uptake, growth, flowering

### INTRODUCTION

Bedding plants cultivated in greenhouses are often grown in disinfected substrates to lower risk of contamination. Disinfection treatment eliminates arbuscular mycorrhizal fungi (AMF) in growing medium. The mycorrhizal inoculation has the potential to enhance growth and flowering of ornamental plants, particularly in nutrient-deficient soils (Gaur and Adholeya,

2005) or in drought conditions (Augz, 2000). Inoculation with AMF during cultivation in greenhouses may be beneficial for further plant growth in outdoor conditions. It was shown earlier that mycorrhization of some bedding plants is significantly reduced by high P concentration (Koide et al. 1999). Plant growth responses to AM fungi are found to vary with the host plant and soil (Entry et al. 2002).

These studies were designed to test the ability of *Glomus* species to colonize the root system of China aster during greenhouse cultivation under different P levels, and to evaluate the effect of mycorrhization and P level on plant growth, flowering and mineral content of leaves and growing medium.

### MATERIALS AND METHODS

Seeds of China aster (*Callistephus chinensis* (L.) Nees), dwarf cultivar 'Milady' used as bedding plants, were sown in February. The seedlings (7-9 cm high, with 6-8 leaves) were planted into sphagnum peat: perlite substrate (3 : 1, v/v), pH 6.1, EC 0.4 mS x cm<sup>-1</sup>. The substrate was inoculated with Endorize – TA AMF inoculum, containing a mixture of different *Glomus* species (Biorize Sarl, France), as described earlier (Nowak, 2004 a). The substrate was not sterilized. The peat used in this experiment was devoid of AMF, as confirmed by the absence of colonization with the non-inoculated treatments. Mycorrhizal infection was estimated after staining the roots with trypan blue (Phillips and Hayman, 1970).

Plants were fertigated by subirrigation (ebb-and-flow benches, Clauhan Project A/S, Denmark). The P treatments were as follows: 8.68, and 43.40 (mg x dm<sup>-3</sup>). The nutrient solutions contained also: N-NO<sub>3</sub> 169.1, K 214.1, Ca 124.0, Mg 18.5, S 35.2, and B 0.24, Fe 1.23, Mn 0.55, Cu 0.03, Mo 0.09, Zn 0.20 (mg x dm<sup>-3</sup>), EC (electrical conductivity) be-

ing  $1.8 \text{ mS} \times \text{cm}^{-1}$ . Potassium nitrate, monobasic potassium phosphate, calcium nitrate, magnesium sulphate, boric acid, iron EDTA, manganese EDTA, copper EDTA, ammonium molybdate, zinc EDTA were used for preparation of nutrient solutions. The nutrient solutions were adjusted to pH 6.0. There were four sections of benches fertigated separately at each P level and mycorrhization treatment. The plants were cultivated under glass from the beginning of April to the end of June. All measurements were conducted at the end of the experiment, after 11 weeks of growth.

Whole plants were harvested. Plant height, leaf number, fresh and dry weights of leaves, numbers of flower buds and flowers, mineral nutrient content of leaves and substrates, root colonization by AM fungi were determined.

Leaf nutrient contents were determined for 3 plants per replicate. There were 3 replications per treatment. All mature leaves were sampled. The leaf tissues were oven-dried at  $78^{\circ}\text{C}$ , milled to homogeneous samples, and then treated with the mixture of 65%  $\text{HNO}_3$  and 60%  $\text{HClO}_4$  (3.5 : 1, v : v). The concentrations of  $\text{K}^+$ ,  $\text{Ca}^{2+}$  and  $\text{Mg}^{2+}$  were measured using atomic absorption spectrophotometry (PU 9100X; Philips, Holland), P was determined colorimetrically by using vanadium-molybdate complex, N was determined using the Kjeldahl method with an automatic distillation system with boric acid (Kieltec, Tecator, Sweden).

Available macroelements contained in the medium were extracted with acetic acid using the modified Spurway method (Nowosielski, 1989). The concentrations of K, Ca, Mg, and P were determined

as described above for leaf samples.  $\text{N-NO}_3$  was measured with a nitrate-specific electrode, EC was determined by a conductivity meter (Type OK-1021, Badelkis, Budapest, Hungary).

The experiment employed a split-plot two factorial design. There were 20 pots per treatment, each plant was treated as a replication. The treatments were statistically analyzed by analysis of variance and means were compared with Duncan's multiple range test at 95% level of significance.

## RESULTS AND DISCUSSION

The mycorrhizal root colonization percentage at the end of the experiment, 11 weeks after inoculation, was rather high in both P treatments, 67% for plants that were fertilized at  $8.68 \text{ mg} \times \text{dm}^{-3} \text{P}$ , while for those fertilized at  $43.40 \text{ mg} \times \text{dm}^{-3} \text{P}$  was 60%. All noninoculated roots remained free of colonization by AMF.

Many studies have shown that mycorrhizal plants have higher shoot P concentrations than plants with no or reduced AMF colonization when these plants are grown in soils with low P content. Information on acquisition of the other macronutrients N, K, Ca, Mg by mycorrhizal plants has been inconsistent, in that increases, no effects, and decreases having been reported (Clark and Zeto, 2000; George, 2000). In China aster slightly increased P content was detected in leaves of mycorrhizal plants grown under low P level (Tab. 1). Mycorrhization did not affect leaf P content of plants grown in high P level. Increased Mg content was measured in leaves of mycorrhizal plants grown

Table 1  
The effects of mycorrhization with AMF and P level on mineral element content of leaves of *Callistephus chinensis* 'Milady' cultivated on ebb-and-flow benches.

Mycorrhization	P level $\text{mg} \times \text{dm}^{-3}$	Mineral element content ( $\text{g} \times \text{kg}^{-1}$ of dry matter)				
		N	P	K	Ca	Mg
No AMF	8.68	4.58a	0.067 a	5.4 a	1.1 a	0.2 a
	43.40	4.56a	0.165 c	5.7 a	1.2 a	0.3 a
AMF	8.68	4.61a	0.078 b	5.2 a	1.3 a	0.4 b
	43.40	4.67a	0.168 c	5.7 a	1.2 a	0.4 b
Significance (main effects)						
Mycorrhization		n.s.	***	n.s.	n.s.	***
P level		n.s.	***	n.s.	n.s.	n.s.
Myc. x P level		n.s.	***	n.s.	n.s.	n.s.

Values followed by the same letter are not significantly different according to the Duncan test ( $P < 0.05$ )

\*\*\*, \*\*, n.s. Significant at  $P < 0.01$ ,  $P < 0.05$ , not significant, respectively

under both P levels. Mycorrhizal and nonmycorrhizal plants did not differ with regard to leaf N, K, and Ca contents. P nutrition did not also affect the contents of these elements in leaf tissue.

Mycorrhization slightly decreased the pH of growing media (Tab. 2). Similar results concerning the effect of AMF on pH were earlier obtained by other authors (Li et al. 1991). AMF significantly lowered salt accumulation in growing media. P level did not affect the pH of growing media, slightly increased total soluble salt content in noninoculated medium, and slightly lowered it in AMF inoculated medium. The lowest N level was measured in growing medium inoculated with AMF and fertilized with nutrient solution containing less P. A higher P level increased P content of growing medium. Mycorrhization and P level in nu-

trient solution did not affect K, Ca, and Mg contents of growing medium.

Significantly lower shoot biomass, plant height, shoot number were recorded in all plants inoculated with AMF (Tab. 3). Reduced growth as a result of mycorrhization was also observed in some *Tagetes* cultivars (Linderman and Davis, 2004). No effect of root inoculation with *Glomus intraradices* on shoot biomass was earlier recorded in *Callistephus chinensis* grown in P-deficient soil under field conditions (Gaur and Adholeya, 2005). It is well known that mycorrhization can depress plant growth primarily by sink competition for photosynthates (Douds et al. 1988). Some studies have indicated that plant growth depression due to mycorrhizal colonization is attributed to greater carbon expenditure in a colonized

Table 2  
The effects of mycorrhization with AMF and P level on chemical characteristic of growing medium at the end of culture of *Callistephus chinensis* 'Milady' cultivated on ebb-and-flow benches.

Mycorrhization	P level mg × dm <sup>-3</sup>	pH	Total soluble salts (g × dm <sup>-3</sup> )	Macroelement content (mg × dm <sup>-3</sup> )				
				N-NO <sub>3</sub>	P	K	Ca	Mg
No AMF	8.68	7.3b	4.9c	1282b	65a	1494a	1224a	175a
	43.40	7.2b	5.5d	1340b	343b	1497a	1367a	184a
AMF	8.68	6.75a	4.15b	1111a	59a	1415a	1211a	152a
	43.40	6.75a	3.95a	1251b	223b	1502a	1249a	169a
Significance (main effects)								
Mycorrhization		***	***	**	n.s.	n.s.	n.s.	n.s.
P level		n.s.	n.s.	**	***	n.s.	n.s.	n.s.
Myc. x P level		n.s.	***	***	n.s.	n.s.	n.s.	n.s.

Explanations as in Table 1

Table 3  
The effects of mycorrhization with AMF and P level on growth of *Callistephus chinensis* 'Milady' cultivated on ebb-and-flow benches.

Mycorrhization	P level mg × dm <sup>-3</sup>	Shoot f.w. (g)	Shoot d.w. (g)	Plant height (cm)	Shoot number	Leaf number
No AMF	8.68	66.2b	13.1b	28.8b	9.2b	31.0ab
	43.40	71.5c	14.4c	29.0b	12.1c	29.6a
AMF	8.68	41.7a	7.9a	27.8ab	5.8a	32.8b
	43.40	43.8a	8.5a	27.3a	6.4a	32.7b
Significance (main effects)						
Mycorrhization		***	***	***	***	***
P level		**	**	n.s.	***	n.s.
Myc. x P level		n.s.	n.s.	n.s.	***	n.s.

Explanations as in Table 1

Table 4  
The effects of mycorrhization with AMF and P level on flowering and visual evaluation of *Callistephus chinensis* 'Milady' cultivated on ebb-and-flow benches.

Mycorrhization	P level mg × dm <sup>-3</sup>	Number of days from planting to flowering	Flower bud number	Flower number	Visual evaluation <sup>1</sup>
No AMF	8.68	69.6b	5.7b	2.6b	3.8b
	43.40	66.7a	5.4b	4.6c	4.3c
AMF	8.68	76.4d	4.3a	1.2a	3.2a
	43.40	73.7c	4.4a	1.2a	3.4a
Significance (main effects)					
Mycorrhization		***	***	***	***
P level		***	n.s.	***	**
Myc. x P level		n.s.	n.s.	***	n.s.

Values followed by the same letter are not significantly different according to the Duncan test ( $P < 0.05$ )

\*\*\*, \*\*, n.s. Significant at  $P < 0.01$ ,  $P < 0.05$ , not significant, respectively. <sup>1</sup>5 – very good plants, 1 – unsaleable plants

root system (Peng et al. 1993). In field cultivation mycorrhizal roots can explore more soil volume than non-mycorrhizal ones, due to their extramatrical hyphae (Sawaki and Saito, 2001); it is impossible in pot culture in very limited volume of growing media. Results obtained on *Typha latifolia* (Dunham et al. 2003) also suggest that under greenhouse conditions, AMF act to reduce plant growth despite increased mineral nutrition and photosynthetic activity.

Mycorrhization delayed flowering of China aster, high P level slightly accelerated it (Tab. 2). Mycorrhizal plants had fewer flower buds and flowers than nonmycorrhizal ones. High P level increased the number of flowers of nonmycorrhizal plants only. Mycorrhization decreased visual evaluation scores of plants at the end of production in the greenhouse. A positive effect of mycorrhization on flower initiation time and flower number of China aster grown in field conditions was reported by Gaur and Adholeya (2005). Summarizing, in greenhouse conditions the effect of mycorrhization on China aster growth and flowering was not beneficial, probably due to hyphae development at the expense of the host plant. The negative effect of mycorrhization on plant growth in greenhouses can be diminished or even overcome by CO<sub>2</sub> enrichment (Nowak, 2004 b). One must not discount the beneficial effects AMF may have upon further plant growth and health in outdoor conditions.

## CONCLUSIONS

1. China aster root colonization by AMF during cultivation on ebb-and-flow benches is possible under both low and high P levels in nutrient solution.

2. AMF inoculation decreased growth of China aster and delayed flowering time.
3. Inoculation with AMF increased P content of China aster leaves of plants grown under low P level, and Mg content in leaves of plants grown in both P levels.
4. Inoculation with AMF decreased the pH and slightly lowered salt accumulation in growing media.

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## **Wpływ mikoryzacji i nawożenia fosforem na zawartość składników mineralnych, wzrost i kwitnienie astra chińskiego (*Callistephus chinensis* (L.) Nees) uprawianego na stołach zalewowych**

### **Streszczenie**

Badano wpływ nawożenia fosforem i inokulacji grzybami mikoryzowymi na zawartość składników mineralnych w liściach, wzrost i kwitnienie astra chińskiego (*Callistephus chinensis* (L.) Nees) 'Milady' uprawianego na stołach zalewowych. Zastosowano dwa stężenia P w pożywce: 8,68 i 43,40 mg × dm<sup>-3</sup>. Korzenie roślin zostały zasiedlone przez grzyby mikoryzowe po miesiącu od inokulacji. Korzenie roślin nawożonych pożywką zawierającą 8,68 mg × dm<sup>-3</sup> zostały zasiedlone w 67%, a nawożonych 43,40 mg × dm<sup>-3</sup> w 60%. Korzenie roślin kontrolnych nie zostały zasiedlone. Stwierdzono nieco wyższą zawartość P w liściach roślin zmikoryzowanych nawożonych pożywką zawierającą mniej P. Mikoryzacja nie wpływała na zawartość P w liściach roślin uprawianych przy wyższym poziomie P w pożywce. Rośliny zmikoryzowane miały więcej Mg w liściach, niezależnie od stężenia P w pożywce. Mikoryzacja nie wpływała na zawartość N, K i Ca w liściach, niezależnie od poziomu P. Poziom P nie wpływał także na zawartość tych składników w liściach. Mikoryzacja obniżała pH podłoża i zawartość rozpuszczalnych soli w podłożu, przy obu poziomach P. Rośliny inokulowane miały mniejszą świeżą masę, mniej pędów i były niższe niż nieinokulowane. Mikoryzacja opóźniała kwitnienie, niezależnie od poziomu P. Wyższy poziom P trochę je przyspieszał. Wyższy poziom P zwiększał liczbę kwiatów u roślin niezmikoryzowanych.