

## Differential Growth Response of Various Crop Species to Arbuscular Mycorrhizal Inoculation

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To investigate the growth response of various crop species to mycorrhizal inoculation, arbuscular mycorrhizal fungi were applied to *Glycine max*, *Vigna angularis*, *Senna tora*, *Hordeum vulgare* var. *hexastichon*, *Zea mays*, *Sorghum bicolor*, *Allium tuberosum*, *Solanum melongena*, and *Capsicum annuum*. The biomass of the inoculated crops was measured every two weeks for the 12-week growth period. By measuring biomass, we calculated the mycorrhizal responsiveness of the nine crop species. Among the nine crop species, four species showed a significant response to mycorrhizal inoculation. The shoot biomasses of *V. angularis*, *C. annuum*, *A. tuberosum*, and *S. tora* significantly increased with mycorrhizal inoculation.

**KEYWORDS** : AM fungi, Mycorrhizal dependency, Relative growth rate

Arbuscular mycorrhizas (AM) are mutualistic symbioses between fungi in phylum glomeromycota and most terrestrial plant roots (Schuessler *et al.*, 2001; Smith and Read, 2008). There is increasing evidence that arbuscular mycorrhizal fungi (AMF) promote plant growth by improving plant uptake of water and inorganic nutrients, especially phosphorus (P). Additional benefits include increased tolerance to environmental stresses such as nutrient deficient soil, drought conditions, salinity, and pathogens (Kurle and Pflieger, 1996). The potential use of AMF in agriculture has received much attention in the past decades because they reduce the use of chemical fertilizers and pesticide (Harrier and Watson, 2004; Sharma *et al.*, 1997).

Most plants have an association with at least one type of mycorrhiza (Smith and Read, 2008). For some plant species, the association with mycorrhizal fungi is indispensable, while some plants show no significant response or negative growth response to mycorrhizal inoculation. Safir (1987) characterizes plants as independent and facultatively or obligately dependent on AMF for mineral nutrient uptake; the degree of dependence varies with plant species, and particularly with root morphology and soil conditions.

There is little information on the response of crop plants to mycorrhizal inoculation in Korea. In this study, the growth responses to mycorrhizal inoculation of several important crop species are reported. Nine species of crops were selected for this study: three species in Leguminosae (*Glycine max* (L.) Merr., *Vigna angularis* (Willd.) Ohwi & H. Ohashi, and *Senna tora* (L.) Roxb.), three species in Gramineae (*Hordeum vulgare* var. *hexastichon* (L.)

Asch., *Zea mays* L., and *Sorghum bicolor* (L.) Moench), one species in Liliaceae (*Allium tuberosum* Rottler ex Spreng.), and two species in Solanaceae (*Solanum melongena* L. and *Capsicum annuum* L.).

Soil collected from a tobacco arable field site in Chungbuk was used as the inoculum source for mycorrhizal fungi. Seeds for each host plant species were sowed in 15 cm × 17 cm pots, with each pot containing 20 g of inoculum and equal parts, by volume, of autoclaved sand/soil mixture. The plants were watered with distilled water as needed and supplemented with 200 ml of quarter-strength Hoagland solution (2.8 g H<sub>3</sub>BO<sub>3</sub>, 3.4 g MnSO<sub>4</sub>·H<sub>2</sub>O, 0.1 g CuSO<sub>4</sub>·5H<sub>2</sub>O, 16.22 g ZnSO<sub>4</sub>·7H<sub>2</sub>O, 0.1 g (NH<sub>4</sub>)<sub>6</sub>MO<sub>7</sub>O<sub>24</sub>·4H<sub>2</sub>O, 5 ml H<sub>2</sub>SO<sub>4</sub>, 6.72 g Na<sub>2</sub>EDTA, 5.58 g FeSO<sub>4</sub>, 0.94 g Ca(NO<sub>3</sub>)<sub>2</sub>·4H<sub>2</sub>O, 0.52 g MgSO<sub>4</sub>·7H<sub>2</sub>O, 0.66 g KNO<sub>3</sub>, 0.06 g HN<sub>4</sub>H<sub>2</sub>PO<sub>4</sub>) weekly. The average temperatures, light intensity and relative humidity in the greenhouse during the experiments were maintained as 35°C, 85% and above 50%, respectively. The heights of the plants were measured every two weeks. After 12 weeks of growth, roots and shoots were harvested and dry weights were measured after being dried at 60°C for 48 h. The mycorrhizal responsiveness of each plant species was expressed and the formula is like this (Hetrick *et al.*, 1996):

$$\begin{aligned} \text{Mycorrhizal responsiveness (\%)} \\ &= [\text{mean biomass mycorrhizal plant} \\ &\quad - \text{mean biomass non-mycorrhizal plant}] / \\ &\quad - \text{mean biomass mycorrhizal plant}] \times 100. \end{aligned}$$

Data regarding the dry weights of plants were analyzed using SPSS for Windows, version 10 (SPSS Inc., Chicago, IL, USA). Comparisons of the growth responses of crop plants to AMF were conducted using t-test of the

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**Table 1.** Means and standard errors of plant dry weights in mycorrhizal and non-mycorrhizal treated plants<sup>1)</sup>

Plant species		Non-mycorrhizal		Mycorrhizal		
		Mean	SE	Mean	SE	
<i>Glycine max</i>	Shoot	10.13	1.31	12.72	1.29	
	Root	2.94	0.46	3.81	0.55	
	Total	13.07	1.79	16.53	1.76	
<i>Vigna angularis</i>	Shoot	2.71	0.51	7.80	1.17	**
	Root	1.26	0.21	1.46	0.23	
	Total	3.96	0.69	9.26	1.38	**
<i>Senna tora</i>	Shoot	0.55	0.07	5.04	1.58	*
	Root	0.13	0.03	1.86	0.78	
	Total	0.68	0.09	6.90	2.33	
<i>Hordeum vulgare</i> var. <i>hexastichon</i>	Shoot	5.80	3.35	3.58	0.89	
	Root	3.86	2.52	0.73	0.19	
	Total	9.66	5.86	4.31	1.05	
<i>Zea mays</i>	Shoot	10.44	2.87	14.64	1.40	
	Root	4.58	1.57	6.84	1.17	
	Total	15.02	4.43	21.47	2.44	
<i>Sorghum bicolor</i>	Shoot	4.06	1.27	8.70	1.71	
	Root	2.01	0.59	4.57	0.90	
	Total	6.06	1.85	13.27	2.61	
<i>Allium tuberosum</i>	Shoot	0.07	0.02	1.45	0.39	**
	Root	0.13	0.03	0.59	0.13	*
	Total	0.20	0.05	2.04	0.52	**
<i>Solanum melongena</i>	Shoot	11.80	1.07	10.21	1.35	
	Root	4.86	0.52	4.08	0.76	
	Total	16.66	1.59	14.29	2.08	
<i>Capsicum annuum</i>	Shoot	0.49	0.08	8.27	0.70	***
	Root	0.26	0.05	2.26	0.25	***
	Total	0.75	0.13	10.53	0.92	***

<sup>1)</sup>Asterisks on the bars indicate that mean biomass of inoculated plant was significantly different from non-inoculated control as determined by two-sample student t-test at  $P < 0.05$  (\*),  $< 0.01$  (\*\*),  $< 0.001$  (\*\*\*)

relative growth rates of plants inoculated with AMF.

The growth responses of 9 species of crop plants were investigated after inoculation with AMF in greenhouse cultivation. Staining roots with Trypan blue (Koske and Gemma, 1989) confirmed mycorrhizal colonization of crop plants inoculated with AMF. Also, mycorrhizal colonization was not observed in the root of the plants not inoculated with AMF, indicating no contamination with AMF during the experiment. The biomasses of plants were measured every two weeks during a 12-week growth period. A total of four of the 9 plant species used in this study significantly responded to mycorrhizal inoculation (Table 1, Fig.

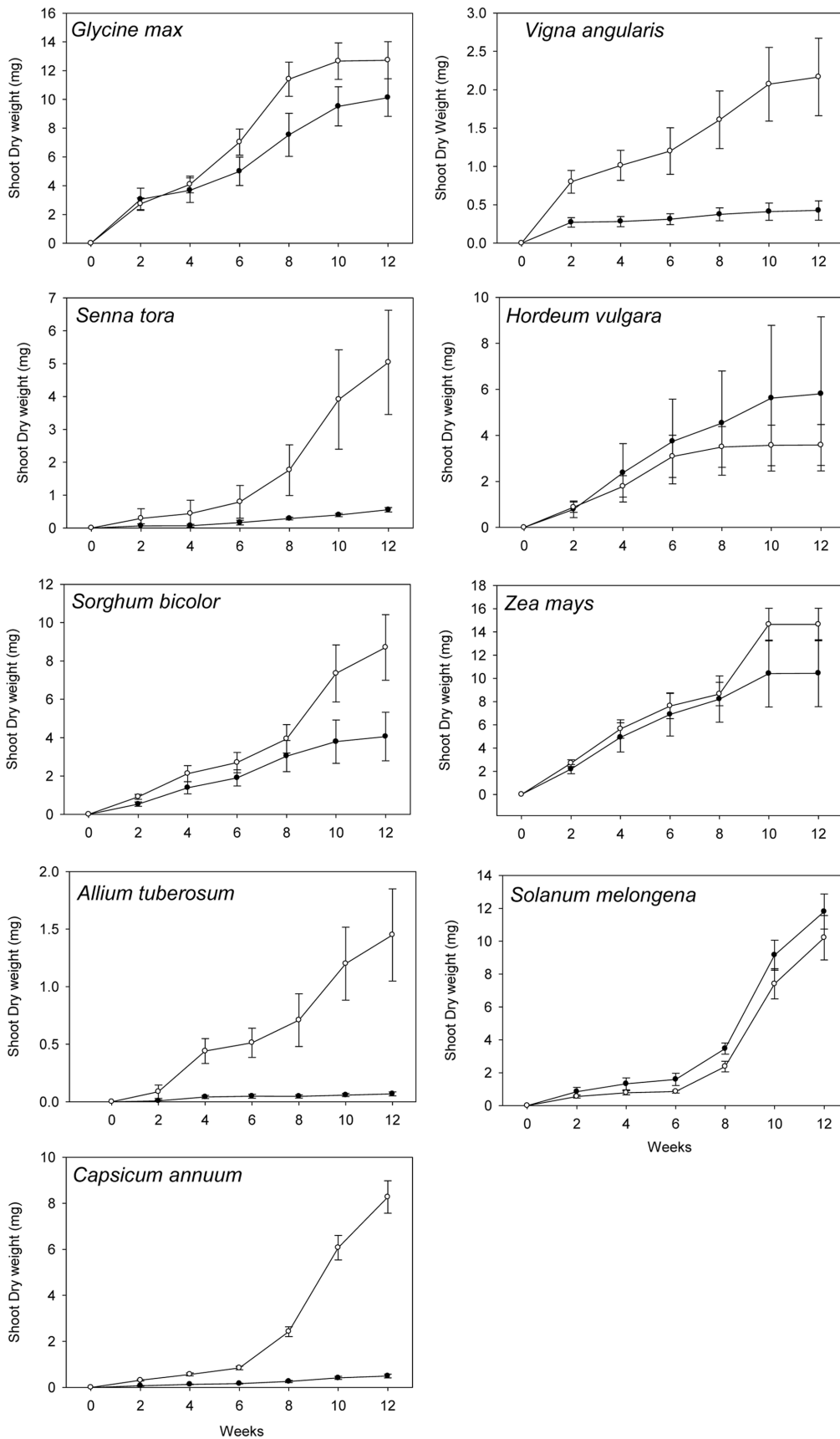
1). Growth of both roots and shoots in *A. tuberosum* and *C. annuum* was enhanced significantly by inoculation with AMF. Only the shoot dry weights of *V. angularis*, *S. tora*, *C. annuum*, and *A. tuberosum* inoculated with AMF were significantly higher than those without AMF. The total biomasses of the plant species *V. angularis*, *C. annuum*, and *A. tuberosum* showed significantly high mycorrhizal responsiveness. Five species of crop plants-*G. max*, *H. vulgare* var. *hexastichon*, *Z. mays*, *S. bicolor*, and *S. melongena*-showed no significant response to mycorrhizal inoculation in this study.

In this study, the extent of the plants' responses to AMF inoculation ranged from highly positive to highly negative (Fig. 2). The results agree with the hypothesis of Johnson *et al.* (1997), that mycorrhizal associations considered symbiotic range functionally along a parasitism-mutualism continuum, and various environmental conditions determine the position of any one case of AMF symbiosis along that continuum. Growth of crop species belonging to Leguminosae tested in this study, except *G. max*, was enhanced by mycorrhizal inoculation. *G. max* is one of the most important crop species in the world and it has shown mycorrhizal dependency in most previous reports (Eom *et al.*, 1994; Khalil *et al.*, 1994). The result for *G. max* in this study could be due to the mycorrhizal community composition of the soil used as inoculum in this study. Soil factors, such as P levels, should be considered in future studies.

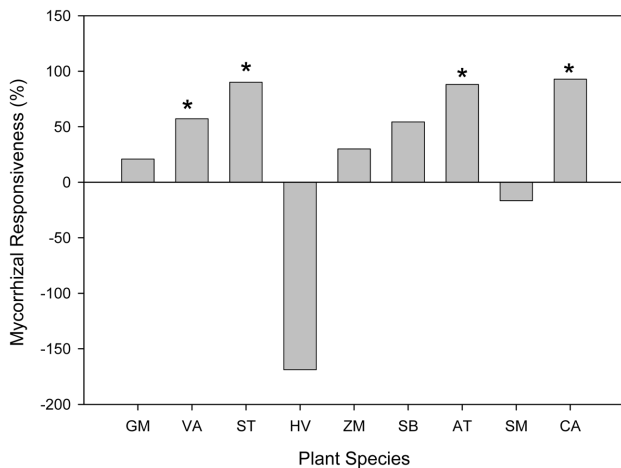
In plant species belonging to Gramineae-such as *H. vulgare* var. *hexastichon*, *Z. mays* and *S. bicolor*-differences in relative growth rates between mycorrhizal and non-mycorrhizal treatments were low and there was no significant response to mycorrhizal inoculation. However, *A. tuberosum* in Liliaceae showed a significant increase in plant growth due to AMF inoculation. Root morphology and architectures, as well as environmental conditions such as soil nutrients, influence the mycorrhizal responsiveness of host plants (Hetrick *et al.*, 1991). Plants with thick, poorly branched roots that possess few root hairs, such as those in Liliaceae, are usually more dependent on mycorrhizae for normal growth and development than plants with thin, highly branched roots, like those in Gramineae.

Both the dry weight and relative growth rate of *C. annuum* inoculated with AMF were significantly higher than those not inoculated with AMF. *C. annuum* is an important vegetable in South Korea and worldwide and it is well known to have a significant positive response to AMF inoculation (Davies Jr. *et al.*, 2002; Park *et al.*, 1999). Results from this study agree with those of previous studies. However, *S. melongena*, which belongs to the same family as *C. annuum* (i.e., Solanaceae), showed no significant growth response to AMF.

The present study aimed to evaluate the responsiveness of important crop species to AMF inoculation in field soil



**Fig. 1.** Growth responses of plants to mycorrhizal inoculation. Mean  $\pm$  SE at 12 weeks after planting. AM (open circle), mycorrhizal plants; NM (dark circle), non-mycorrhizal control plants.



**Fig. 2.** Mycorrhizal responsiveness of plant species used in this study. GM; *Glycine max*, VA; *Vigna angularis*, ST; *Senna tora*, SM; *Solanum melongena*, CA; *Capsicum annum*, AT; *Allium tuberosum*, HV; *Hordeum vulgare* var. *hexastichon*, ZM; *Zea mays*, SB; *Sorghum bicolor*. Asterisks on bars indicate significant responses to mycorrhizal inoculation at  $P = 0.05$ . \* MR = Mycorrhizal responsiveness (%) =  $[\text{mean biomass mycorrhizal plant} - \text{mean biomass non-mycorrhizal plant}] / \text{mean biomass mycorrhizal plant} \times 100$ . Asterisks on the bars indicate that mean biomass of inoculated plant was significantly different from non-inoculated control as determined by two-sample t-test at  $P < 0.05$ .

and under greenhouse conditions. The host species showed differential growth responses to AMF. A number of factors can affect the growth response of host plants to AMF colonization, including the genotypes of host plants and environmental conditions (Sensory *et al.*, 2007). Different levels of responsiveness among different genotypes in the same crop species have been demonstrated in previous studies (Declerck *et al.*, 1995; Linderman and Davis, 2004). This study did not account for the factor of host-plant genotype, but it should be considered in future study.

There have been many reports that different species or genotypes of AMF prompt different levels of growth response (Klironomos, 2003; Sensoy *et al.*, 2007; van der Heijden *et al.*, 1998). The soil used in this study as AMF inoculum included a mixture of several AMF species, but the taxa of the AMF used in the study could have affected the responses of host plant species in ways that other AMF mixtures may not. Also, it has been widely accepted that high P levels in soil reduce mycorrhizal colonization of host plants and negate AMF's potential growth enhancement (Smith and Read, 2008). Conventional agricultural practices involve a high fertilizer input that can cause low mycorrhizal inoculum potential and therefore a loss of effective AMF response. This study did not control the amount of P in the potting medium; instead, it

used soils collected from agricultural field soil as an AMF inoculum. The P level in the pots of soil could be one reason for the insignificant responses in several species of host plants and this factor should be addressed in future study.

The benefits of AMF association to plant species present great opportunities for current agricultural practices; the proper use of these symbiotic associations is important to maintaining sustainable agriculture. The results in this study demonstrate the enhanced growth of several crop species inoculated with AMF; they therefore suggest the possibility that AMF could be applied to these 9 commonly cultivated crop species in Korea.

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