

## The effects of arbuscular mycorrhizal fungi on the seedling growth of four hybrid cucumber (*Cucumis sativus* L.) cultivars

Şefik TÜFENKÇİ<sup>1</sup>, Semra DEMİR<sup>2</sup>, Suat ŞENSOY<sup>3\*</sup>, Hüsameddin ÜNSAL<sup>1</sup>, Emre DEMİRER<sup>2</sup>,

Çeknas ERDİNÇ<sup>3</sup>, Şeyhmus BİÇER<sup>3</sup>, Aytekin EKİNCİALP<sup>3</sup>

<sup>1</sup>Department of Soil Science and Plant Nutrition, Agricultural Faculty, Yüzüncü Yıl University, 65080 Van - TURKEY

<sup>2</sup>Department of Plant Protection, Agricultural Faculty, Yüzüncü Yıl University, 65080 Van - TURKEY

<sup>3</sup>Department of Horticulture, Agricultural Faculty, Yüzüncü Yıl University, 65080 Van - TURKEY

Received: 28.12.2010

**Abstract:** The effectiveness of different arbuscular mycorrhizal fungi (AMF) on different hybrid cucumber cultivars has not been well documented, even under normal seedling conditions. The present study therefore aimed to evaluate colonization, nutrient uptake, dependency, and other seedling traits of 4 cucumber hybrids (Ceren F<sub>1</sub>, Beta F<sub>1</sub>, Silyon F<sub>1</sub> and Maraton F<sub>1</sub>) inoculated by 3 different AMF [*Glomus intraradices* (Gi), *Glomus etunicatum* (Ge) and *Gigaspora margarita* (Gm)]. Traits were evaluated in a growth chamber experiment consisting of a 4 × 4 factorial design (4 cucumber hybrids, 3 AMF plus 1 control) with 3 random replications. AMF-inoculated cucumber seedlings had shorter hypocotyledons and wider and longer cotyledons than non-inoculated seedlings. *Gm*-inoculated seedlings had the narrowest stem diameter and lowest leaf number. AMF-inoculated seedlings had shorter shoots and longer roots than noninoculated ones. There was significant mycorrhizal effect on the iron (Fe) content of shoots and the mycorrhizal colonization rate in roots. Relative mycorrhizal dependency (RMD) varied widely among the hybrid cucumber cultivars tested. It is thought that the determination of high-RMD cultivars could lead to improvements in cucumber seedling production in the future.

**Key words:** AMF, colonization, cucumber, dependency, nutrient uptake, symbiosis

### Arbüsküler mikorhizal fungusların dört hibrit hıyar (*Cucumis sativus* L.) çeşidinin fide gelişimine etkisi

**Özet:** Farklı arbüsküler mikorhizal fungusların (AMF) farklı hibrit hıyar çeşitlerindeki etkinlikleri normal fide koşullarında bile yeterince ortaya konulmamıştır. Bu nedenle, mevcut çalışma 3 farklı AMF [*Glomus intraradices* (Gi), *Glomus etunicatum* (Ge) ve *Gigaspora margarita* (Gm)] türü ile inokule edilen 4 farklı hibrit hıyar çeşidindeki (Ceren F<sub>1</sub>, Beta F<sub>1</sub>, Silyon F<sub>1</sub> ve Maraton F<sub>1</sub>) kolonizasyon, besin maddesi alınımı, bağımlılık ve bazı fide özelliklerine etkilerini belirlemek amacıyla yürütülmüştür. Özellikler, iklim odası koşulları altında yürütülen denemede 4 × 4 faktöriyel deneme deseninde (4 hibrit hıyar çeşidi, 3 AMF ve 1 kontrol) 3 tekerrürlü olarak incelenmiştir. AMF ile inokule edilmiş hıyar fideleri inokule edilmemiş olanlara oranla daha kısa hipokotillere ve daha geniş ve uzun kotiledonlara sahip olmuştur. Genel olarak AMF ile inokule edilmiş fideler, inokule edilmemiş olanlara oranla daha kısa sürgünlere ve daha uzun köklere sahip olurken, *Gm* ile inokule edilmiş fideler en dar gövde çapına ve en az yaprak sayısına sahip olmuşlardır. Sürgün demir (Fe) içeriği ve köklerde kolonizasyon oranı üzerine mikorhizal uygulamaların etkisi önemli bulunmuştur. Oransal mikorizal bağımlılık (OMB) test edilen hibrit hıyar çeşitleri arasında büyük oranda değişmiştir. Yüksek OMB'ye sahip çeşitlerin belirlenmesinin gelecekte hıyar fide yetiştiriciliğinde olumlu gelişmelere yol açacağı düşünülmektedir.

**Anahtar sözcükler:** AMF, bağımlılık, besin alımı, hıyar, kolonizasyon, simbiyosis

\* E-mail: suatsensoy@yyu.edu.tr

## Introduction

Arbuscular mycorrhizal fungi (AMF) are the most widespread root fungal symbionts and are associated with the vast majority of higher plants. Mycorrhizal symbiosis plays an important role in enhancing the growth and health of the host plants. AMF have been shown to improve soil structure (Miller and Jastrow 2000), and their high capacity to increase plant growth and yield by improving plant nutrient uptake gives them great importance (Smith and Read 2008). AMF also enable plants to cope with both biotic and abiotic stresses; they may help fight off some soil-borne diseases (Garmendia et al. 2004; Hu et al. 2010), alleviate certain nutrient deficiencies, improve drought tolerance, overcome the detrimental effects of salinity, and enhance tolerance to pollution (Brundrett 1991; Declerck et al. 1995; Türkmen et al. 2005; Sensoy et al. 2007; Turkmen et al. 2008).

Cucumber (*Cucumis sativus* L.) is an important vegetable in Turkey and throughout the world (FAOSTAT 2009). So far, limited research has been done to reveal the effects of AMF on the growth and yield of cucumbers. Recent studies have suggested that AMF-inoculated cucumbers could benefit from association with AMF (Demir 2002; Wang et al. 2008; Hu et al. 2010; Rouphael et al. 2010; Yıldız 2010).

Differences in mycorrhizal responsiveness between different crops and between different genotypes within the same crop have also been demonstrated (Declerck et al. 1995; Parke and Kaeppler 2000; Linderman and Davis 2004; Sensoy et al. 2007; Miyauchi et al. 2008). Linderman and Davis (2004) have shown this clearly in their study of different marigold genotypes and AMF, as have Declerck et al. (1995) in their work with bananas and AMF. Sensoy et al. (2007) observed a great variation in mycorrhizal colonization dependency among pepper genotypes, leading them to state that appropriate cultivar-AMF combinations need to be identified in order to derive the utmost benefit from symbiosis.

The effectiveness of AMF appears to have been better on sterilized growth medium, which provided the AMF with a healthier environment in which to perform their beneficial functions (Wang et al. 2008). AMF inoculation could also enhance the transplant performance in some vegetables (Temperini et al.

2009). The benefits of AMF inoculation depend on the genotypic host-fungus combinations and also the type of inocula used (Rouphael et al. 2010). Inadequate attention has been given to mycorrhizal responsiveness by modern breeding activities, especially hybrid cultivar improvement. Khalil et al. (1994) reported that improved cultivars in soybeans showed a lower growth response to colonization than older cultivars.

The effectiveness of different AMF on different cucumber hybrids has not been well documented, even under normal seedling conditions. Therefore, the present study aimed to evaluate colonization, nutrient uptake, responsiveness, and other seedling traits of 4 cucumber hybrids inoculated with 3 different AMF.

## Materials and methods

A total of 4 cucumber hybrids used in protected cultivation in Turkey were examined in the present study: Ceren F<sub>1</sub> (Hazera Seed Company), Beta 309 F<sub>1</sub> (Beta Ziraat Seed Company), Silyon F<sub>1</sub> (Rito Seed Company), Maraton F<sub>1</sub> (Biar Seed Company).

In addition, 3 AMF inocula were tested in the study [*Glomus intraradices* (Gi), *Glomus etunicatum* (Ge), and *Gigaspora margarita* (Gm)], all of which were supplied by the original stocks of Yüzüncü Yıl University's Agriculture Faculty, Department of Plant Protection. The inocula consisted of spores, extraradical mycelium, and mycorrhizal roots.

Growth medium consisted of an autoclaved mixture of perlite and peat moss (1:1 v/v) in seedling trays covered by vermiculite. Each individual plant cell measured approximately 4.5 cm L × 5.5 cm W × 5.5 cm D. The experiment used a 4 × 4 factorial design (4 cucumber hybrids, 3 AMF plus 1 control) with 3 random replications of 15 cells each, for a total of 720 cells. One seed was sown per cell, each of which contained 80 cm<sup>3</sup> of sterilized growth medium. In the AMF-inoculated samples, 5 g (25 spores g<sup>-1</sup>) of inocula, counted by the wet sieving method (Gerdemann and Nicholson 1963), were placed in the growth medium before the seeds were sown (Demir and Onoğur 1999). Seedling trays were placed in a growth chamber at a temperature of 22 ± 2 °C with 12 h of fluorescent illumination (8000

lux light intensity), and were irrigated with distilled water. Each seedling was fertilized twice with 5 mL of nutrient solution (for 1 L: 720 mg  $MgSO_4 \cdot 7H_2O$ , 12.2 mg  $KH_2PO_4$ , 295 mg  $Ca(NO_3)_2 \cdot 4H_2O$ , 240 mg  $KNO_3$ , 0.75 mg  $MnCl_2 \cdot 4H_2O$ , 0.75 mg KI, 0.75 mg  $ZnSO_4 \cdot H_2O$ , 1.5 mg  $H_3BO_3$ , 0.001 mg  $CuSO_4 \cdot 5H_2O$ , 4.3 mg FeNaEDTA, and 0.00017 mg  $Na_2MoO_4 \cdot 2H_2O$ ) as modified by Vosatka and Gryndler (1999). Plants were harvested 9 weeks after seed sowing and inoculation.

Seedling emergence percentage, seedling emergence time, and true leaf emergence time were determined. Hypocotyledon height, cotyledon height, cotyledon length, and stem diameter were measured. Shoot heights, root lengths, stem diameters, leaf numbers, shoot dry weights, and root dry weights of seedlings were determined after harvesting. Samples were then oven-dried at 68 °C for 48 h and ground, and the nitrogen (N) contents of the shoots were measured using the Kjeldahl method; the potassium (K), calcium (Ca), magnesium (Mg), iron (Fe), copper (Cu), zinc (Zn), and manganese (Mn) contents of the shoots were measured using an atomic absorption spectrophotometer; and the phosphorous (P) contents of the shoots were measured using the vanadate-molybdate-yellow procedure with a spectrophotometer (Kacar and İnal 2008).

A modified method of Phillips and Hayman (1970) was used for the detection of AMF presence in dyed cucumber roots and the percentage and intensity of mycorrhizal colonization was estimated using the gridline intersect method (Giovanetti and Mosse 1980).

The relative mycorrhizal dependency (RMD) of cucumber hybrids was expressed as the difference between the total dry weight of the mycorrhizal plant and the total dry weight of the nonmycorrhizal plant as a percentage of the total dry weight of the mycorrhizal plant (Declerck et al. 1995; Sensoy et al. 2007).

Data were analyzed using the SAS statistical program (SAS Software 1997), with variance analysis conducted for all data. Differences between treatments were determined using Duncan's multiple range test.

## Results

AMF application had no significant effects on the seedling emergence percentage (Table 1). On the other hand, AMF had significant effects on seedling emergence time (Table 1). *Gm*-inoculated cucumber seedlings emerged the earliest, followed by *Ge*-inoculated ones. *Gi*-inoculated and noninoculated cucumber seedlings emerged the latest. Hypocotyledon height, cotyledon height, and cotyledon length were all significantly affected by genotype and AMF (Table 1). *Gi*-, *Gm*-, and *Ge*-inoculated seedlings had 1.6%, 18.4%, and 23.7% shorter hypocotyledons than those of noninoculated seedlings, respectively. On the other hand, *Ge*-, *Gm*-, and *Gi*-inoculated seedlings had 6.4%, 5.5%, and 1.7% wider cotyledons than those of noninoculated seedlings, respectively. *Gm*-, *Gi*-, and *Ge*-inoculated seedlings also had 9.7%, 5.3%, and 2.0% longer cotyledons than those of noninoculated seedlings, respectively.

AMF application had significant effects on seedling stem diameter, leaf number, shoot height, and root length (Table 2). *Ge*-inoculated cucumber seedlings had the widest stem diameter, followed by *Gi*-inoculated ones. *Gm*-inoculated cucumber seedlings had the narrowest stem diameter. While noninoculated and *Gi*-inoculated cucumber seedlings had the highest leaf number, *Gm*-inoculated cucumber seedlings had the lowest leaf number. *Gi*-, *Ge*-, and *Gm*-inoculated seedlings had 5.0%, 14.2%, and 18.8% shorter shoots than those of noninoculated seedlings, respectively. *Ge*-, *Gm*-, and *Gi*-inoculated seedlings had 22.6%, 17.2%, and 7.1% longer roots than those of noninoculated seedlings, respectively.

AMF application had no significant effects on the fresh or dry weights of cucumber seedling roots (Table 3). There were, however, significant genotypic effects on the dry and fresh weights of shoots from cucumber seedlings. While Beta F<sub>1</sub> had the highest fresh shoot weight, Marathon F<sub>1</sub> had the lowest shoot dry weight.

Although AMF application had no significant effects on N, P, Ca, or Mg contents, there was a significant genotypic effect on K content (Table 4). While Beta F<sub>1</sub> had the highest K content among the shoots studied, Ceren F<sub>1</sub> had the lowest K content.

Table 1. The effects of AMF [*Glomus intraradices* (*Gi*), *Glomus etunicatum* (*Ge*), and *Gigaspora margarita* (*Gm*)] and genotype on the seedling emergence percentage, seedling emergence time, hypocotyledon height, cotyledon length, and cotyledon length of cucumber plants.

Applications	Seedling emergence (%)	Seedling emergence time (day)	Hypocotyledon height (mm)	Cotyledon width (mm)	Cotyledon length (mm)	
Cultivars	Ceren F <sub>1</sub>	93.9 NS	11.3 NS	40.9 a***	24.9 b***	44.9 b***
	Beta F <sub>1</sub>	95.0	11.0	41.1 a	27.8 a	49.6 a
	Silyon F <sub>1</sub>	95.0	10.8	36.0 b	25.0 b	45.9 b
	Maraton F <sub>1</sub>	91.6	11.5	36.7 b	23.7 c	41.2 c
AMF	-AMF	99.4 NS	12.4 a*	43.51 a***	24.5 c**	43.6 c***
	<i>Ge</i>	90.5	10.8 ab	33.18 b	26.1 a	45.9 ab
	<i>Gi</i>	95.5	11.6 a	42.83 a	25.0 bc	44.4 bc
	<i>Gm</i>	90.0	9.7 b	35.48 b	25.9 ab	47.8 a
Noninoculated Ceren F <sub>1</sub>	97.8 NS	10.9 NS	47.3 NS	24.2 NS	44.2 NS	
<i>Ge</i> -inoculated Ceren F <sub>1</sub>	88.9	11.9	34.4	25.8	45.2	
<i>Gi</i> -inoculated Ceren F <sub>1</sub>	95.5	11.4	44.1	24.1	41.7	
<i>Gm</i> -inoculated Ceren F <sub>1</sub>	93.3	10.9	37.9	25.8	48.6	
Noninoculated Beta F <sub>1</sub>	100.0	12.5	44.9	26.6	47.6	
<i>Ge</i> -inoculated Beta F <sub>1</sub>	93.3	10.4	36.3	28.9	50.4	
<i>Gi</i> -inoculated Beta F <sub>1</sub>	97.8	11.5	44.5	27.4	47.4	
<i>Gm</i> -inoculated Beta F <sub>1</sub>	88.9	9.7	39.1	28.7	53.2	
Noninoculated Silyon F <sub>1</sub>	100.0	13.0	40.9	24.3	43.6	
<i>Ge</i> -inoculated Silyon F <sub>1</sub>	91.1	10.4	30.9	25.6	46.7	
<i>Gi</i> -inoculated Silyon F <sub>1</sub>	95.5	11.0	41.4	25.0	47.5	
<i>Gm</i> -inoculated Silyon F <sub>1</sub>	93.3	8.8	31.1	25.2	45.8	
Noninoculated Maraton F <sub>1</sub>	100.0	13.3	40.9	23.2	38.8	
<i>Ge</i> -inoculated Maraton F <sub>1</sub>	88.9	10.6	31.1	24.3	41.2	
<i>Gi</i> -inoculated Maraton F <sub>1</sub>	93.3	12.6	41.3	23.5	41.1	
<i>Gm</i> -inoculated Maraton F <sub>1</sub>	84.4	9.4	33.8	24.0	43.7	

Values are the means of 3 replicate samples. AMF: arbuscular mycorrhizal fungi. -AMF: nonmycorrhizal plants. NS: nonsignificant. \*: P < 0.05; \*\*: P < 0.01; \*\*\*: P < 0.001 (significant).

Table 2. The effects of AMF [*Glomus intraradices* (*Gi*), *Glomus etunicatum* (*Ge*) and *Gigaspora margarita* (*Gm*)] and genotype on the seedling stem diameter, leaf number, shoot height and root length of cucumber plants.

Applications		Stem diameter (mm)	Leaf number	Shoot height (cm)	Root length (cm)
Cultivars	Ceren F <sub>1</sub>	3.18 NS	3.03 NS	5.62 NS	22.46 NS
	Beta F <sub>1</sub>	3.38	3.08	5.82	21.55
	Silyon F <sub>1</sub>	3.32	3.26	5.15	21.02
	Maraton F <sub>1</sub>	3.43	3.23	5.70	22.53
AMF	-AMF	3.30 ab*	3.30 a**	6.16 a**	19.59 b**
	<i>Ge</i>	3.43 a	3.12 ab	5.28 bc	24.03 a
	<i>Gi</i>	3.42 a	3.28 a	5.85 ab	20.98
	<i>Gm</i>	3.17 b	2.91 b	5.00 c	22.97
Noninoculated Ceren F <sub>1</sub>		3.23 NS	3.17 NS	6.50 NS	20.53 NS
<i>Ge</i> -inoculated Ceren F <sub>1</sub>		3.30	3.03	5.07	25.20
<i>Gi</i> -inoculated Ceren F <sub>1</sub>		3.17	3.17	5.83	24.13
<i>Gm</i> -inoculated Ceren F <sub>1</sub>		3.03	2.77	5.07	19.97
Noninoculated Beta F <sub>1</sub>		3.33	3.27	6.63	17.30
<i>Ge</i> -inoculated Beta F <sub>1</sub>		3.47	3.03	5.70	24.90
<i>Gi</i> -inoculated Beta F <sub>1</sub>		3.57	3.20	5.73	1.47
<i>Gm</i> -inoculated Beta F <sub>1</sub>		3.13	2.83	5.20	25.57
Noninoculated Silyon F <sub>1</sub>		3.33	3.40	5.37	21.43
<i>Ge</i> -inoculated Silyon F <sub>1</sub>		3.27	3.23	4.73	21.87
<i>Gi</i> -inoculated Silyon F <sub>1</sub>		3.40	3.40	5.70	17.60
<i>Gm</i> -inoculated Silyon F <sub>1</sub>		3.27	3.00	4.80	23.17
Noninoculated Maraton F <sub>1</sub>		3.30	3.37	6.13	19.10
<i>Ge</i> -inoculated Maraton F <sub>1</sub>		3.67	3.17	5.63	24.13
<i>Gi</i> -inoculated Maraton F <sub>1</sub>		3.53	3.33	6.13	23.73
<i>Gm</i> -inoculated Maraton F <sub>1</sub>		3.23	3.03	4.93	23.17

Values are the means of 3 replicate samples. AMF: arbuscular mycorrhizal fungi. -AMF: nonmycorrhizal plants. NS: nonsignificant. \*: P < 0.05; \*\*: P < 0.01 (significant).

Table 3. The effects of AMF [*Glomus intraradices* (*Gi*), *Glomus etunicatum* (*Ge*) and *Gigaspora margarita* (*Gm*)] and cucumber genotypes on the fresh and dry weights of shoots and roots.

Applications		Shoot fresh weight (g plant <sup>-1</sup> )	Root fresh weight (g plant <sup>-1</sup> )	Shoot dry weight (mg plant <sup>-1</sup> )	Root dry weight (mg plant <sup>-1</sup> )
Cultivars	Ceren F <sub>1</sub>	2.64 b***	1.27 NS	338 a**	50.9 NS
	Beta F <sub>1</sub>	2.94 a	1.26	350 a	53.3
	Silyon F <sub>1</sub>	2.62 b	1.15	335 a	48.3
	Maraton F <sub>1</sub>	2.54 b	1.16	264 b	45.0
AMF	-AMF	2.70 NS	1.12 NS	329 NS	49.2 NS
	<i>Ge</i>	2.78	1.28	330	52.7
	<i>Gi</i>	2.74	1.20	324	48.3
	<i>Gm</i>	2.56	1.25	304	47.5
Noninoculated Ceren F <sub>1</sub>		2.77 NS	1.23 NS	330 NS	53.3 NS
<i>Ge</i> -inoculated Ceren F <sub>1</sub>		2.69	1.48	373	60.0
<i>Gi</i> -inoculated Ceren F <sub>1</sub>		2.46	1.29	307	46.7
<i>Gm</i> -inoculated Ceren F <sub>1</sub>		2.64	1.13	343	46.7
Noninoculated Beta F <sub>1</sub>		3.02	1.16	363	56.7
<i>Ge</i> -inoculated Beta F <sub>1</sub>		2.90	1.13	350	50.0
<i>Gi</i> -inoculated Beta F <sub>1</sub>		3.11	1.23	353	50.0
<i>Gm</i> -inoculated Beta F <sub>1</sub>		2.74	1.50	333	56.7
Noninoculated Silyon F <sub>1</sub>		2.58	1.12	353	46.7
<i>Ge</i> -inoculated Silyon F <sub>1</sub>		2.77	1.13	340	50.0
<i>Gi</i> -inoculated Silyon F <sub>1</sub>		2.72	1.23	316	46.7
<i>Gm</i> -inoculated Silyon F <sub>1</sub>		2.55	1.13	333	50.0
Noninoculated Maraton F <sub>1</sub>		2.44	0.96	270	40.0
<i>Ge</i> -inoculated Maraton F <sub>1</sub>		2.77	1.44	260	53.3
<i>Gi</i> -inoculated Maraton F <sub>1</sub>		2.66	1.04	320	50.0
<i>Gm</i> -inoculated Maraton F <sub>1</sub>		2.30	1.20	207	36.7

Values are the means of 3 replicate samples. AMF: arbuscular mycorrhizal fungi. -AMF: nonmycorrhizal plants. NS: nonsignificant. \*\*: P < 0.01; \*\*\*: P < 0.001 (significant).

Table 4. The effects of AMF [*Glomus intraradices* (*Gi*), *Glomus etunicatum* (*Ge*), and *Gigaspora margarita* (*Gm*)] and cucumber genotype on nitrogen, phosphorous, potassium, calcium and magnesium contents of shoots.

Applications		Nitrogen (%)	Phosphorous (%)	Potassium (%)	Calcium (%)	Magnesium (%)
Cultivars	Ceren F <sub>1</sub>	1.71 NS	1.07 NS	1.76 b*	4.16 NS	1.13 NS
	Beta F <sub>1</sub>	1.81	1.05	2.30 a	4.85	1.08
	Silyon F <sub>1</sub>	1.67	1.09	2.06 ab	5.01	1.25
	Maraton F <sub>1</sub>	2.19	1.25	2.21 a	3.90	1.41
AMF	-AMF	1.73 NS	1.11 NS	1.91 NS	4.77 NS	1.25 NS
	<i>Ge</i>	1.90	1.13	2.30	4.23	1.13
	<i>Gi</i>	1.79	1.21	2.12	4.30	1.28
	<i>Gm</i>	1.92	1.13	1.98	4.66	1.20
Noninoculated Ceren F <sub>1</sub>		1.45 NS	0.94 NS	1.53 NS	4.38 NS	1.15 NS
<i>Ge</i> -inoculated Ceren F <sub>1</sub>		1.73	1.08	1.95	3.19	0.95
<i>Gi</i> -inoculated Ceren F <sub>1</sub>		1.90	1.23	1.81	4.74	1.21
<i>Gm</i> -inoculated Ceren F <sub>1</sub>		1.73	1.04	1.65	4.00	1.13
Noninoculated Beta F <sub>1</sub>		2.00	1.28	1.94	5.28	1.08
<i>Ge</i> -inoculated Beta F <sub>1</sub>		1.80	0.95	2.57	4.20	1.03
<i>Gi</i> -inoculated Beta F <sub>1</sub>		1.50	1.26	2.57	5.61	1.15
<i>Gm</i> -inoculated Beta F <sub>1</sub>		1.95	1.38	2.00	4.56	1.08
Noninoculated Silyon F <sub>1</sub>		1.57	1.06	1.77	4.74	0.99
<i>Ge</i> -inoculated Silyon F <sub>1</sub>		1.57	1.16	2.66	5.75	1.52
<i>Gi</i> -inoculated Silyon F <sub>1</sub>		2.30	1.29	1.96	4.72	1.23
<i>Gm</i> -inoculated Silyon F <sub>1</sub>		1.47	0.99	1.83	4.82	1.22
Noninoculated Maraton F <sub>1</sub>		1.90	1.20	2.28	4.65	1.75
<i>Ge</i> -inoculated Maraton F <sub>1</sub>		2.53	1.39	2.02	3.41	0.95
<i>Gi</i> -inoculated Maraton F <sub>1</sub>		1.45	0.99	2.13	2.55	1.47
<i>Gm</i> -inoculated Maraton F <sub>1</sub>		2.85	1.29	2.43	5.51	1.46

Values are the means of 3 replicate samples. AMF: arbuscular mycorrhizal fungi. -AMF: nonmycorrhizal plants. NS: nonsignificant. \*: P < 0.05 (significant).

Although AMF application had no significant effects on Cu, Zn, or Mn contents, there was a significant mycorrhizal effect on the Fe content and mycorrhizal colonization, as well as significant genotypic effects on the Zn and Mn contents and mycorrhizal colonization (Table 5). *Gm*-inoculated cucumber seedlings had the highest Fe contents, followed by those inoculated with *Gi*, while *Ge*-inoculated cucumber seedlings had the lowest Fe contents. While Ceren F<sub>1</sub> had the highest Zn content in shoots, Silyon F<sub>1</sub> had the lowest Zn content in

shoots. While Maraton F<sub>1</sub> had the highest Mn content in shoots, Beta F<sub>1</sub> had the lowest Zn content in shoots. The cultivar Ceren F<sub>1</sub> had the highest mycorrhizal colonization compared to the other genotypes.

RMD varied widely among the hybrid cucumber cultivars tested (Figure). *Ge*- and *Gm*-inoculated Ceren F<sub>1</sub> and *Ge*- and *Gi*-inoculated Maraton F<sub>1</sub> had positive RMDs. On the other hand, Beta F<sub>1</sub> and Silyon F<sub>1</sub> had negative RMDs to all of the AMF tested. Moreover, *Gi*-inoculated Ceren F<sub>1</sub> and *Gm*-inoculated Maraton F<sub>1</sub> also had negative RMDs.

Table 5. The effects of AMF [*Glomus intraradices* (*Gi*), *Glomus etunicatum* (*Ge*), and *Gigaspora margarita* (*Gm*)] and cucumber genotype on mycorrhizal colonization and iron, copper, zinc and manganese contents of shoots.

Applications		Iron (mg kg <sup>-1</sup> )	Copper (mg kg <sup>-1</sup> )	Zinc (mg kg <sup>-1</sup> )	Manganese (mg kg <sup>-1</sup> )	Mycorrhizal colonization (%)
Cultivars	Ceren F <sub>1</sub>	61.2 NS	13.85 NS	34.14 b**	74.4 ab*	44.83 a***
	Beta F <sub>1</sub>	62.1	9.94	30.94 b	67.81 b	25.26 b
	Silyon F <sub>1</sub>	66.0	8.88	29.49 b	74.72 ab	24.02 b
	Maraton F <sub>1</sub>	72.9	10.59	41.99 a	84.21 a	21.50 b
AMF	-AMF	59.8 bc*	8.34 NS	33.72 NS	71.49 NS	0.00 b***
	<i>Ge</i>	54.5 c	12.48	31.80	71.82	40.32 a
	<i>Gi</i>	73.6 ab	11.04	35.15	73.85	40.51 a
	<i>Gm</i>	75.9 a	11.70	35.90	82.7	33.86 a
Noninoculated Ceren F <sub>1</sub>		51.5 NS	14.05 NS	34.24 NS	75.57 NS	0.00 NS
<i>Ge</i> -inoculated Ceren F <sub>1</sub>		53.3	15.35	33.12	67.47	64.9
<i>Gi</i> -inoculated Ceren F <sub>1</sub>		71.7	14.02	32.82	67.33	63.8
<i>Gm</i> -inoculated Ceren F <sub>1</sub>		65.4	11.96	36.37	87.22	53.3
Noninoculated Beta F <sub>1</sub>		63.7	7.11	28.54	63.04	0.00
<i>Ge</i> -inoculated Beta F <sub>1</sub>		49.0	10.16	29.31	66.67	34.1
<i>Gi</i> -inoculated Beta F <sub>1</sub>		76.9	7.42	32.37	67.18	34.4
<i>Gm</i> -inoculated Beta F <sub>1</sub>		57.9	15.07	33.56	74.34	32.4
Noninoculated Silyon F <sub>1</sub>		61.3	4.54	30.52	69.55	0.00
<i>Ge</i> -inoculated Silyon F <sub>1</sub>		41.9	12.43	27.81	70.49	37.7
<i>Gi</i> -inoculated Silyon F <sub>1</sub>		87.2	12.19	34.57	81.61	30.5
<i>Gm</i> -inoculated Silyon F <sub>1</sub>		80.6	6.37	25.06	75.82	32.7
Noninoculated Maraton F <sub>1</sub>		62.9	6.43	41.56	77.78	0.00
<i>Ge</i> -inoculated Maraton F <sub>1</sub>		71.6	11.99	36.96	87.42	29.3
<i>Gi</i> -inoculated Maraton F <sub>1</sub>		63.9	10.54	40.84	79.28	33.2
<i>Gm</i> -inoculated Maraton F <sub>1</sub>		93.7	13.40	48.63	93.42	23.3

Values are the means of 3 replicate samples. AMF: arbuscular mycorrhizal fungi. -AMF: nonmycorrhizal plants. NS: nonsignificant. \*: P < 0.05; \*\*: P < 0.01; \*\*\*: P < 0.001(significant).



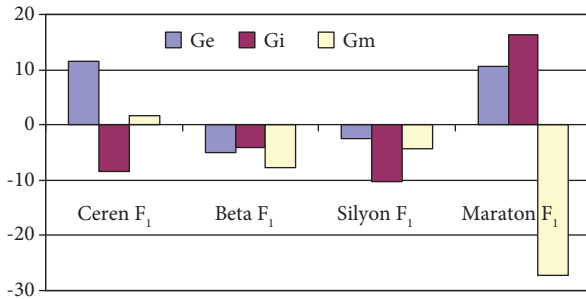


Figure. The relative mycorrhizal dependency of 4 cucumber hybrid cultivars inoculated with 3 AMF [*Glomus intraradices* (Gi), *Glomus etunicatum* (Ge) and *Gigaspora margarita* (Gm)].

## Discussion

AMF are well known to have significant positive effects on many crops. Several studies have demonstrated that the genetic variation in plant responses to AMF is almost universal, regardless of the crop (Declerck et al. 1995; Parke and Kaeppeler 2000; Linderman and Davis 2004; Sensoy et al. 2007). Thus far, a limited amount of research has been done to reveal the effects of AMF on the growth and yield of cucumbers. Recent studies have suggested that AMF-inoculated cucumbers could benefit from association with AMF (Çiğışar et al. 2000; Demir 2002; Abak et al. 2007; Wang et al. 2008; Hu et al. 2010; Roupheal et al. 2010; Yıldız 2010).

Roupheal et al. (2010) studied the effects of AMF inoculation on 2 cucumber genotypes (the hybrid Ekron and the open-pollinated Marketmore) and reported significant effects of genotype on most of the investigated traits. In the present study, there were also significant genotypic effects on many of the traits investigated. Similarly, Wang et al. (2008) reported that the growth of cucumber seedlings was influenced differently by the 3 AMF species examined in that study. Those researchers investigated the effects of AMF (*Glomus intraradices*, *G. mosseae*, and *G. versiforme*) on the growth and yield of cucumber cultivar Jinlu No. 3. Their results indicated that the growth of seedlings was significantly enhanced by *G. mosseae*, inhibited by *G. versiforme*, and not significantly influenced by *G. intraradices*.

Roupheal et al. (2010) reported that mycorrhizal cucumber plants had a higher macronutrient

concentration in the leaf tissue compared to noninoculated ones. These researchers also observed that *Glomus intraradices* enhanced the uptake and translocation of Fe toward the shoot. In the present study, both *Gigaspora margarita*- and *Glomus intraradices*-inoculated cucumber seedlings had higher Fe contents in the shoots. Wang et al. (2008) reported on the effects of 3 AMF species (*Glomus mosseae*, *G. intraradices*, and *G. versiforme*) in cucumber cultivar Jinlu No. 3. These researchers found that the N and P contents in roots and the Mg, Cu, and Zn contents in shoots were increased by inoculation with AMF, while the K and Fe contents in shoots decreased significantly. In the present study, there were significant effects of AMF application on Fe concentration. Wang et al. (2008) observed a decrease in Fe content, but in the present study, there was an increase in Fe contents by some AMF species. Miyauchi et al. (2008) studied the interactions between diazotrophic bacteria and mycorrhizal fungi in maize genotypes including 3 hybrids and 2 inbred lines. These researchers reported that the inbred maize lines showed higher phosphorous concentrations when compared to the hybrids mainly colonized by AMF.

RMD varied greatly among the hybrid cucumber cultivars tested in the present study. The RMDs ranged from 16.22% to -27.21%, with no clear correlation between the mycorrhizal response and the AMF colonization level and intensity. The finding of a lack of correlation between AMF colonization and RMD was similar to the findings of a study by Linderman and Davis (2004) that looked at different marigold genotypes and AMF, and the findings of a study by Sensoy et al. (2007) that compared different pepper genotypes and AMF. The present study demonstrated that the variation in responsiveness to AMF among hybrid cucumber cultivars was relatively large. In the present study, there were different AMF colonization levels among the hybrid cucumber cultivars. Ceren F<sub>1</sub> had the highest mycorrhizal colonization compared to the other genotypes. Roupheal et al. (2010) observed higher root colonization in the open-pollinated variety Marketmore (21.8%) than in the hybrid Ekron (12.7%). Demir (2002) determined that *G. intraradices* colonized the roots of cucumber cultivar Toros (41.4%) and established a good symbiotic relationship. Yıldız (2010) studied the effects of

indigenous *Glomus* sp. and commercial BioOrganics (*Glomus aggregatum*, *G. clarum*, *G. deserticola*, *G. intraradices*, *G. monosporus*, *G. mosseae*, *Gigaspora margarita*, and *Paraglomus brasilianum*) on some vegetables including cucumber and found that the indigenous *Glomus* spp. showed positive effects on cucumber seedlings; the colonization rates of *Glomus* spp. and commercial BioOrganics were 71% and 47%, respectively. Çiğşar et al. (2000) demonstrated the beneficial effects of using local isolates in a greenhouse study on cucumber. Abak et al. (2007) investigated the effects of 4 different AMF (*Glomus mosseae*, *G. etunicatum*, *G. fasciculatum*, and *G. caledonium*) on the soilless cucumber Dairus F<sub>1</sub> and found that *G. caledonium* had some positive effects on plant growth and yield.

Azcon et al. (1991) observed that differences in functional compatibility existed among AMF even though the root colonization was similar. AMF species were also different in terms of their carbon demands (Saikkonen et al. 1999). Wang et al. (2008) speculated that the growth response induced by a specific AMF relied on recognition between the AMF species and the host plant; sometimes negative growth response with AMF was induced by the imbalance of carbohydrate distribution between the shoots and roots of the plants. Hodge et al. (2010) stated that the major fluxes in AMF symbiosis appear to be of carbon from plant to fungus and of phosphorous (and possibly nitrogen) from fungus to plant. Miyauchi et al. (2008) reported that mycorrhizae increased shoot and root biomass in maize, as well as root traits such as total length, specific length, total surface, and the incidence of root hairs in all of the genotypes, including 3 hybrids. Sensoy et al. (2007) stated that the fact that the most responsive pepper genotypes had the lowest dry weights appeared to indicate that RMD and genotype dry weight were negatively correlated, with increased plant vigor related to a reduction in the symbiotic effect. Their findings are in line with those of Linderman and Davis (2004) for marigolds.

Parke and Kaeppler (2000) stated that the inadequate attention given to mycorrhizal responsiveness by modern breeding activities could result in increased needs for nutrient inputs and a loss of genes in crop germplasm. Other important benefits of AMF symbiosis, such as drought tolerance, disease

resistance, and soil-structure maintenance, may also be underrecognized in current practices.

Temperini et al. (2009) reported that AMF inoculation showed enhanced transplant performance in local pepper genotypes, and they proposed that the use of AMF in nurseries could provide a benefit to the development of bell pepper transplants, particularly under organic farming conditions. There are many other benefits of AMF in agriculture. AMF may influence crop development, even in phosphorous-rich soils, and the development of cultivars with improved symbiotic qualities would ensure the production of good crop yields while improving agrosystem sustainability (Hamel and Strullu 2006). Hu et al. (2010) reported that an AMF consortium (mainly consisting of *Glomus* spp. and *Acaulospora* spp.) could suppress Fusarium wilt in cucumber cultivar Jinyan 4 and, therefore, that it showed potential as a biological control agent in greenhouse agroecosystems.

In conclusion, AMF present valuable opportunities for current agricultural practice with regard to various biotic and abiotic stress conditions. The effective utilization of these symbiotic soil fungi is indispensable for sustainable agriculture. As seen in the examples demonstrated by the results of these studies, AMF might improve seedling traits in vegetable species. Considering the wide variety of responses of different plant cultivars to AMF, as demonstrated in this and other studies, appropriate cultivar-AMF combinations need to be identified in order to derive the utmost benefit from symbiosis. To date, modern plant breeding methods have paid little attention to such symbiotic relationships. Greater interest in this area, incorporating AMF-related traits from high-RMD genotypes into modern cultivars, could lead to improvements in cucumber production by increasing strength under the various biotic and abiotic stress conditions that might be encountered during the crop production season. The development of cultivars with improved symbiotic qualities would ensure the production of good crop yields while improving agrosystem sustainability. Growth response at seedling nursing could be a good indicator of AMF applications, but the effectiveness of AMF on the full growing period of hybrids should also be investigated in more depth for use in protected cultivation conditions in the future.

## References

- Abak K, Rehber Y, Yıldız M, Daşgan HY, Ortaş İ (2007) Topraksız hıyar yetiştiriciliğinde Vesiküler arbusküler mikorizaların bitki gelişimi ve meyve verimine etkisi. Türkiye V. Ulusal Bahçe Bitk Kong, Bildiri Kitapları Cilt II, 4-7 September 2007, Erzurum, pp. 258-261.
- Azcon R, Rubio R, Barea JM (1991) Selective interactions between different species of mycorrhizal fungi and *Rhizobium meliloti* strains, and their effects on growth, N<sub>2</sub>-fixation (<sup>15</sup>N) and nutrition of *Medicago sativa* L. New Phytol 117: 399-404.
- Brundrett M (1991) Mycorrhizas in natural ecosystems. Advances in Ecological Research 21: 171-313.
- Çığşar S, Sarı N, Ortaş İ (2000) The effects of vesicular-arbuscular mycorrhizae on the plant growth and nutrient uptake of cucumber. Turk J Agric For 24: 571-578.
- Declerck S, Plenchette C, Strullu DG (1995) Mycorrhizal dependency of banana (*Musa acuminata*, AAA group) cultivar. Plant and Soil 176: 183-187.
- Demir S (2002) Mikorhizal fungus *Glomus intraradices* (Schenck&Smith)'in bazı sebze bitkilerinin köklerinde kolonizasyonu. Yüzyüncü Yıl Üniversitesi J Agr Sci 12: 53-57.
- Demir S, Onoğur E (1999) *Glomus intraradices* Schenck&Smith: a hopeful vesicular-arbuscular mycorrhizal (VAM) fungus determined in soils of Türkiye. J Turk Phytopatol 28: 33-34.
- FAOSTAT (2009) Statistic Database. Available at <http://apps.fao.org/>.
- Garmendia I, Goicoechea N, Aguirolea J (2004) Effectiveness of three *Glomus* species in protecting pepper (*Capsicum annuum* L.) against verticillium wilt. Biological Control 31: 296-305.
- Gerdemann LW, Nicholson TH (1963) Spores of mycorrhizal endogene extracted from soil by wet sieving and decanting. Trans Br Mycol Soc 46: 235-244.
- Giovanetti M, Mosse B (1980) An evaluation of techniques for measuring vesicular-arbuscular mycorrhizal infection in roots. New Phytol 84: 489-500.
- Hamel C, Strullu DG (2006) Arbuscular mycorrhizal fungi in field crop production: potential and new direction. Canadian J Plant Sci 86: 941-950.
- Hodge A, Helgason T, Fitter AH (2010) Nutritional ecology of arbuscular mycorrhizal fungi. Fungal Ecology 3: 267-273.
- Hu J, Lin X, Wang J, Shen W, Wu S, Peng S, Mao T (2010) Arbuscular mycorrhizal fungal inoculation enhances suppression of cucumber fusarium wilt in greenhouse soils. Pedosphere 20: 586-593.
- Kacar B, İnal A (2008) Plant Analysis. Nobel Publications, Ankara.
- Khalil S, Loynachan TE, Tabatabai MA (1994) Mycorrhizal dependency and nutrient uptake by improved and unimproved corn and soybean cultivars. Agron J 86: 949-958.
- Linderman RG, Davis AE (2004) Varied response of marigold (*Tagetes* spp.) genotypes to inoculation with different arbuscular mycorrhizal fungi. Sci Hort 99: 67-78.
- Miller RM, Jastrow JD (2000) Mycorrhizal fungi influence soil structure. In: Arbuscular Mycorrhizas: Physiology and Function (Eds. Y Kapulnik, DD Douds Jr). Kluwer Academic Publications, Dordrecht, pp. 3-18.
- Miyauchi MYH, Lima DS, Nogueira MA, Lovato GM, Murate LS, Cruz MF, Ferreira JM, Andrade G (2008) Interactions between diazotrophic bacteria and mycorrhizal fungus in maize genotypes. Scientia Agricola 65: 525-531.
- Parke JL, Kaepler SW (2000) Effects of genetic differences among crop species and cultivars upon the arbuscular mycorrhizal symbiosis. In: Arbuscular Mycorrhizas: Physiology and Function (Eds. Y Kapulnik, DD Douds Jr). Kluwer Academic Publication, Dordrecht, pp. 131-146.
- Phillips JM, Hayman DS (1970) Improved procedure for cleaning roots and staining parasitic and vesicular-arbuscular mycorrhizal fungi for rapid assessment of infection. Trans Br Mycol Soc 55: 158-161.
- Rouphael Y, Cardarelli M, Di Mattia E, Tullio M, Rea E, Colla G (2010). Enhancement of alkalinity tolerance in two cucumber genotypes inoculated with an arbuscular mycorrhizal biofertilizer containing *Glomus intraradices*. Biol Fertil Soil 46: 499-509.
- Saikkonen K, Ahonen-Jonnarth U, Markkola AM, Helander M, Tuomi J, Roitto M, Ranta H (1999) Defoliation and mycorrhizal symbiosis: a functional balance between carbon sources and below-ground sinks. Ecology Letters 2: 19-26.
- Sensoy S, Demir S, Turkmen O, Erdinc C, Savur OB (2007) Responses of some different pepper (*Capsicum annuum* L.) genotypes to inoculation with two different arbuscular mycorrhizal fungi. Sci Hort 113: 92-95.
- Smith SE, Read DM (2008) Mycorrhizal Symbiosis, 3rd ed. Academic Press, London.
- Temperini O, Rouphael Y, Parrano L, Biagiola E, Colla G, Mariotti R, Rea E, Rivera CM (2009) Nursery inoculation of pepper with arbuscular mycorrhizal fungi: an effective tool to enhance transplant performance. Acta Hort (ISHS) 807: 591-596.
- Türkmen Ö, Demir S, Şensoy S, Dursun A (2005) Effects of arbuscular mycorrhizal fungus and humic acid on the seedling development and nutrient content of pepper grown under saline soil conditions. J Biol Sci 5: 568-574.
- Turkmen O, Sensoy S, Demir S, Erdinc C (2008) Effects of two different AMF species on growth and nutrient content of pepper seedlings grown under moderate salt stress. Afr J Biotech 7: 392-396.
- Vosatka V, Gryndler M (1999) Treatment with culture fractions from *Pseudomonas putida* modifies the development of *Glomus fistulosum* mycorrhiza and the response of potato and maize plants to inoculation. Appl Soil Ecol 11: 245-251.
- Wang C, Li X, Zhou J, Wang G, Dong Y (2008) Effects of arbuscular mycorrhizal fungi on growth and yield of cucumber plants. Commun Soil Sci Plant Anal 39: 499-509.
- Yıldız A (2010) A native *Glomus* sp. from fields in Aydın province and effects of native and commercial mycorrhizal fungi inoculants on the growth of some vegetables. Turk J Biol 34: 447-452.