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The effect of mycorrhizal inoculation on forage and non-forage plant growth and nutrient uptake under field conditions

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SUMMARY – The effect of mycorrhizal inoculation on nitrogen fixing and non-fixing plant growth and macro- and micro-nutrient uptake were observed under field conditions. Several field experiments were set up on the research farm of Çukurova University (Eastern Mediterranean region). In one experiment clover, lentil, onion, garlic, chickpea and horse-bean plants were used. Cocktail mycorrhiza was used as a mycorrhizal strain. The results have shown, that in mycorrhizal plots, the yields of onion, garlic, chickpea, clover, lentil and horse-bean plants were higher than in non-mycorrhizal plants. Mycorrhizal inoculation also increased Cu and Zn content in the shoot. In another experiment carried out under field conditions for three successful years on horse-bean, chickpea and soybean plants, mycorrhizal inoculation was successfully applied in sterile and non-sterile soil conditions with and without phosphorous application. After a three-year evaluation it was found that under field conditions mycorrhizal inoculation significantly increased horse-bean, chickpea and soybean. Likewise, applying more P resulted in increased yield. Garlic, chickpea and horse-bean did not grow very well in fumigated plots compared to the non-fumigated ones. It was found that in fumigated plots mycorrhizal inoculation significantly increased growth of horse-bean and garlic. Mycorrhizal inoculation also increased nutrient uptake of both plants. Inoculation of the soil with mycorrhizae strains significantly increased plant growth and P uptake of plants, especially under low P supply. In low P application, plant roots were strongly infected and consequently increased plant growth, but with high P level applications there was a slight reduction in root infection. The results show that mycorrhizal inoculation is an effective practice for improving crop production in P-deficient soils.

Key words: Mycorrhizal inoculation, dry matter yield, nutrient uptake, root colonization.

***RESUME –** "Effet de l'inoculation mycorhizienne sur la croissance et sur l'absorption de nutriments des plantes fourragères et non fourragères en conditions de terrain". L'effet de l'inoculation mycorhizienne sur la croissance des plantes fixatrices et non fixatrices d'azote et sur l'absorption de macro- et micro-nutriments a été observé en conditions de terrain. Plusieurs expérimentations de terrain ont été mises en place dans la ferme de recherches de l'Université de Çukurova (region méditerranéenne orientale). Dans une expérience, on a utilisé des plantes de trèfle, lentille, oignon, ail, pois chiche et fèverole. On a utilisé un mélange de mycorhizes comme souche mycorhizienne. Les réultats ont montré que, dans les parcelles mycorhiziennes, les rendements de l'oignon, ail, pois chiche, trèfle, lentille et fèverole, ont été supérieurs par rapport aux plantes non mycorhiziennes. L'inoculation mycorhizienne a également augmenté la teneur en Cu et Zn des pousses. Dans une autre expérience menée en conditions de terrain pendant trois années sur fèverole, pois chiche et soja, l'inoculation mycorhizienne a été appliquée avec succès en conditions de sol stériles et non stériles avec ou sans application de phosphore. Après une évaluation sur trois années, on a trouvé qu'en conditions de terrain l'inoculation mycorhizienne a donné une augmentation significative pour la fèverole, le pois chiche et le soja. De même, l'application de davantage de P a augmenté les résultats pour l'ail. L'ail, le pois chiche et la fèverole ne grandissaient pas très bien dans les parcelles sous fumigation par rapport aux parcelles non soumises à fumigation. On a trouvé que dans les parcelles sous fumigation l'inoculation mycorhizienne augmentait significativement la croissance de la fèverole et de l'ail. L'inoculation mycorhizienne augmentait également l'absorption de nutriments de ces deux plantes. L'inoculation du sol par des souches de mycorhizes augmentait significativement la croissance des plantes et l'absorption de P des plantes, en particulier sous faible apport de P. Avec une faible application de P, les racines des plantes étaient fortement infectées et par conséquent la croissance des plantes augmentait, mais avec des niveaux d'application de P élevés il y avait une lègère réduction de l'infection racinaire. Les résultats montrent que l'inoculation mycorhizienne est une pratique pour améliorer la production des cultures dans les sols présentant un déficit en P.*

***Mots-clés :** Inoculation mycorhizienne, rendement en matière sèche, absorption de nutriments, colonisation par les racines.*

Introduction

South part of Turkey is a centre of domestication of many legume species. The soils of the

Cukurova region are of different origin, most of them are alluvial, very productive and some are calcareous with low organic matter and nitrogen content and low P availability. Some of the soils are very low in micronutrient content. For a better crop production it is necessary to use fertilizer or managing soil indigenous mycorrhizae. Mycorrhizal infection is a very common association between plant roots and micro organisms which is responsible for nutrient uptake especially P and N (Ortas, 1994). Mycorrhizal inoculation also can increase plant resistant against drought and salt effect. Mycorrrhizal infection can reduce excess pesticides and fungicides application due to the protective effective of mycorrhiza against root pathogens (Marschner, 1995). Mycorrhizal fungi are known to increase the P uptake of plants on P-deficient soils. Also it is known that there is strong correlation between mycorrhizal inoculation and N fixation (Barea, 1991; Harley and Smith, 1983). Mycorrhizal inoculation of legume plants depending on biological nitrogen fixation increases plant P and N uptake and nodulation. Low mycorrhizal efficiency and the lower number of nodulation will result a deficiency of P and N nutrition for soybean. Thus there is a strong need to use the natural sources such as *Rhizobium* and mycorrhizal fungi inoculation to growth plant and accordingly reducing N and P fertilizer application.

The role of legume by fixing nitrogen is very important by using less nitrogen, as fertilizer. As a result less nitrogen fertilizer is applied for next crop. If there is a dual inoculation by using rhizobium and mycorrhiza the effect is higher. Also, the effect of legumes species on exudation of organic anions especially in rhizosphere dynamics will have an influence on the availability of phosphorus and zinc and iron mobilization.

Chickpea, lentil, horse bean are coarse-rooted legume species which are an essential source of protein for low income people of Mediterranean part of Turkey. Since legume plants are rarely responded to the P fertilization at same time are considered to be a P efficient plant. Weber (1992) demonstrated in northern Syria that mycorrhizae infected chickpea plant increased P uptake and plant dry matter production. Islam (1985) showed that mycorrhizal under pot and field conditions can increase dry matter production and P uptake of chickpea growth on Northern Syria soils. So it is sound to use dual inoculation of rhizobium and mycorrhiza for better plant nutrition and also for soil fertility development. By using crop rotation in the system with forage crops soil fertility will be more sustainable as well. For developing countries such as Turkey which are spending most of their income for buying fertilizer it is of important for soil fertility. Management of mycorrhizal populations in the field is certainly feasible and requires a clear understanding of the ecology of plant communities and different farming systems which affect the populations of mycorrhizal fungi and their diversity and nutrient uptake and growth of crops. So it is so important to manage the indigenous mycorrhizae when the soil nutrients, especially phosphorus are limited under field conditions. For sustainable food management soil and crop management can help to get maximum benefit from indigenous mycorrhiza (Ortas, 2003).

The aim of the work, determining the potential effect of indigenous and selected mycorrhizal inoculation on several forage and non forage plants growth and nutrient uptake under field conditions.

Materials and methods

Three experiments were carried out for several successive years. The experiments were carried out on the Arık soil series (*Entic Chromoxerert*), located at the Agriculture Faculty Research Farm, University of Çukurova, in the Eastern Mediterranean region of Turkey. Soil chemical and physical properties are presented in Table 1.

Table 1. Chemical and physical properties of soil used (means of three replicates)

Soil Texture (%)			Olsen extractable P (kg P_2O_5ha^{-1})	Extractable K* (mg 100 g^{-1} Soil)	Total N (%)	pH (2:5 in water)	Total Salt (%)	Organic matter (%)	$CaCO_3$ (cmol$_c$ kg^{-1})	CEC (cmol$_c$ kg^{-1})	Number of mycorrhizal spores
Clay	Sand	Silt									
54.1	33.0	12.9	82	900	0.11	7.3	0.06	1.37	15.8	30.4	110-115

*Extracted in 1.0 N NH_4OAc (mg 100 g^{-1} soil).

Experiment 1. Several plant species were tested under field conditions with and without mycorrhizal inoculation and 100 kg P_2O_5 per ha phosphorus applied.

Experiment 2. Horse bean, clover and lentil plant were tested under field conditions with and without mycorrhizal inoculation. Field soil was fertilized with TSP to provide 0 and 100 kg P_2O_5 per ha, as the soil treatments P0 and P1 respectively.

Experiment 3. Horse been plant were used for three successive years under field conditions with and without sterilization, mycorrhizal inoculation were done.

Soil Preparation and Sowing. The field was ploughed in autumn and planted in December. Seed were sowed in 5-7 cm depth. For chickpea row spacing and plant spacing was 25x10 cm, for lentil 20x2 cm and horse bean 35x10 cm. Cocktail mycorrhize with approximately 1000 spore at 5 cm below the seed bed was used. Non-inoculated pots received the same amount of non-mycorrhizal spore inoculum. Plants were harvested at maturation stage. After drying the seed and biomass data were recorded. Root samples were carefully collected from soil and stained as described by Koske and Gemma (1989), and examined for the presence and degree of mycorrhizal infection according to Giovannetti and Mosse (1980). Statistical analyses were performed using the Statistical Analysis System (SAS, 1989) package.

Results and discussion

Experiment I

Onion, garlic, chickpea, horse bean, clover and lentil were used under field conditions. In this experiment cocktail mycorrhiza was used as mycorrhizae species. The results have shown, that in mycorrhizal plots, the yields of all plant species was higher than in non mycorrhizal plants (Fig. 1).

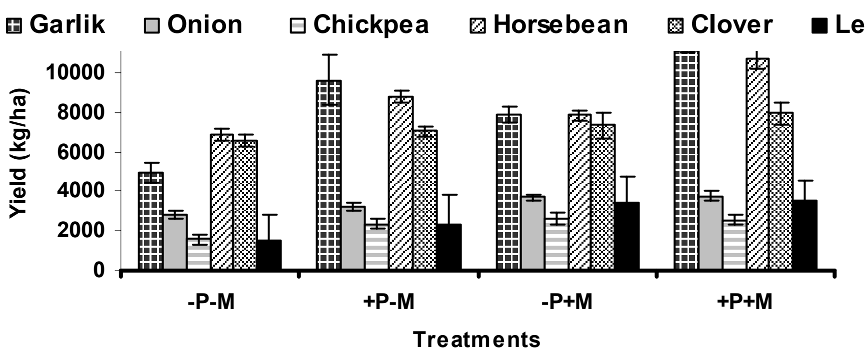

Fig. 1. The effect of mycorrhizal inoculation and P addition on yield.

Mycorrhizal inoculation increased the shoot Cu and Zn content. Horse bean, onion and lentil have given high response to P addition and mycorrhizal inoculation. Since clover, lentil, chickpea and horse bean are nitrogen fixing plants they have taken more macro and micronutrient. Mycorrhiza inoculation significantly increased plant micronutrient uptake as well. This is very important for human and animal feeding.

Mycorrhizal dependency was also calculated. It has been found that under low level of P treatment mycorrhizal dependency is higher than high P treatment (Fig. 2). In horsebean plant even in high P treatment mycorrhizal dependency was higher than low P treatment.

Compared to clover and horse bean, lentil, chickpea, garlic and onion are highly mycorrhizal dependent plants. Baylis (1975) hypothesised that mycorrhization and dependency on mycorrhizae of a plant species depend on root coarseness of host species. Since horse bean has coarser roots would therefore be expected to have more intensive mycorrhization and to depend more on AM. The results were recently derived and shown that there is a high mycorrhizal potential which can successfully infect plant roots. Also it has been shown that there is a positive interaction between *B. japonicum* and mycorrhizal inoculation in term of nutrient uptake (Coskan *et al*., 2003).

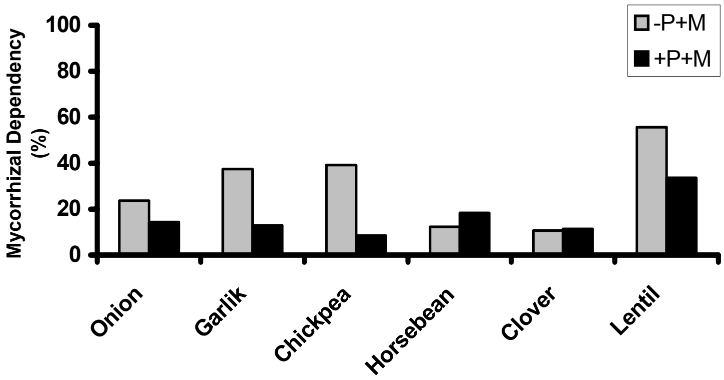

Fig. 2. The effect of P and mycorrhiza inoculation on mycorrhizal dependency of field crops.

The better P uptake by mycorrhizal host plants often leads to secondary indirect effect such as improved host performance under drought, environment stress factors. The results of Weber (1992) in northern Syria shown that mycorrhizae infected chickpea plant get benefit from mycorrhiza most probably due to high water consumption thus increased drought stress under Mediterranean conditions.

Experiment II

Mycorrhizal inoculum significantly increased the plant seed and biomass yield. Also P application increased yield. It has been found that mycorrihzal inoculation increased seed yield up to 19 % and biomass yield increased up to 21% (Table 2). Contribution of mycorrhizal inoculation on yield in P0 treatment is higher than P100 treatments. In general, mycorrhizal contribution on biomass production was higher than seed production. This needs to be researched for further.

Table 2. Effect of phosphorus and mycorrhizal inoculation on forage crop seed and biomass yield

P treatment (kg/ha)	Yield (kg ha^{-1}				Contribution of Mycorrhiza (%)	
	-M		+M			
	Seed	Biomass	Seed	Biomass	Seed	Biomass
Lentil						
0	1105	2482	1317	2934	19	18
100	1421	2717	1607	3161	13	16
Chickpea						
0	964	2108	1103	2548	14	21
100	1265	2471	1398	2952	11	19
Horse bean						
0	2286	4952	2618	5868	15	18
100	3196	5632	3592	6522	12	16

Since mycorrhiza is much more dependent on P treatment, with P addition the mycorrhizal contribution is decreased. Excessive P fertilization not only reduced mycorrhizal efficiency, but was frequently reported to decrease mycorrhization (Hayman, 1983) which is probably plant-mediated

(Menge *et al.*, 1978). P deficiency increases the efflux of root exudates which might stimulate fungal growth (Graham *et al.,* 1981). Koide and Li (1990) suggested that the infection is controlled by the status of the root resulting from local P availability. But in some cases AM are tolerant to excessive fertilizer P applications (Sieverding and Howeler 1985). In present experiment, under high P addition, mycorrhizal inoculation has a contribution. Weber (1992), reported although chickpea, lentil and horse bean plant under field conditions have high root infection and get benefit but there was no apparent evidence for a differing degree of mycorrhizal dependency.

All three plants were significantly inoculated with myocorhizal inoculation. As expected with P addition plant root colonization was relatively less than non P addition treatment. Since there is indigenous mycorrhiza in non-sterile soil there is nearly 29 percentages of root colonization (Fig. 3).

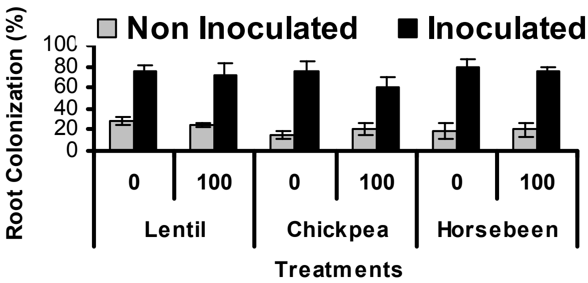

Fig. 3. The effect of mycorrhizal inoculation on root colonization.

Experiment III

Another experiment was carried out under field conditions for three successful years during 1997 to 1999 with horse bean plant. In plane of Cukurova there is a serious problem with soil born disease such as plant parasitic nematodes, soil-borne plant pathogens, root-rod and some weed pest. Soil fumigation may have a dual effect on plant growth, such as increased growth by elimination of soil-borne pathogens, or conversely, stunted growth by exacerbation of existing P deficiency as a result of reduction of mycorrhizal colonization in low-P soils. Since soil fumigation reduced useful organisms such as mycorrhizae it is necessary to inoculate again. Especially mycorrhizal depended plants need more mycorrhizal inoculation. Mycorrhizal inoculation successfully was applied in sterile and non sterile soil conditions with and without phosphorous application. The field experiment was set up on Menzilat soil series (Typic Xerofluvent) which is located at Çukurova Research Farm. In these experiments 0 and 100 kg P_2O_5/ha as triple superphosphate was applied. Mycorrhizal inoculum was applied (by hand) 5 cm under the seeds.

After three years evaluation it has been found that under field condition mycorrhizal inoculation significantly increased horse bean yield. Also increasing P application increased yield. Plants have not been grown very well in fumigated plots than non-fumigated one. Dry matter productions of plants grown in non-fumigated soils were much higher than fumigated one (Fig. 4). However in fumigated plots plant grown better than in non fumigated plots. It has been found that in fumigated plots mycorrhizal inoculation significantly increased growth of horse bean and garlic. Also mycorrhizal inoculation increased nutrient uptake of both plants.

The results showed that at higher rates of P application the effect of mycorrhizae is masked. The potential value of mycorrhizae in natural ecosystems and their importance is diminished under high rate of fertilizer application. At the same experiment mycorrhizal inoculation also increased plant P, Zn and Cu content compared to the control. At the lowest P supply, shoot dry matter production was significantly depressed. This decreasing effect of low P supply was particularly obvious when soils were sterilized and not inoculated with mycorrhizae. Inoculation of soil with mycorrhizae species significantly increased plant growth and P uptake of plants, especially under low P supply. In low P application, plant roots were strongly infected and consequently increased plant growth, but in high P level application there was a slight reduction in root infection. The results show that mycorrhizal inoculation is an effective practice for improving crop production in P deficient soils. Ortas and

Akpinar, (2006) shown that mycorrhizal inoculation significantly increased plant growth and mycorrhizae dependency. Plant grown in fumigated soil has a minimum level of mycorrhizal infection. Plants grown in control plots have higher level of mycorrhizal infection. The differences in dry matter production and nutrient uptake in non-fumigated plot was considered to be related to indigenous mycorrhizal infection.

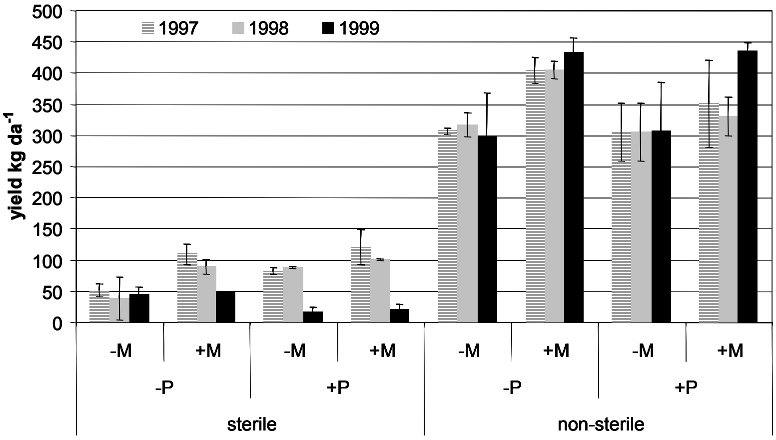

Fig. 4. The effects of treatments on horse bean yield.

Mycorrhizal dependency has been calculated. In sterile soil condition with low P treatment the dependency is much higher than no sterile condition (Fig. 5). Probably, there is an indigenous mycorrhizal spore in soil, as a consequence of indigenous mycorrhizal spore plant getting benefit from mycorrhizae.

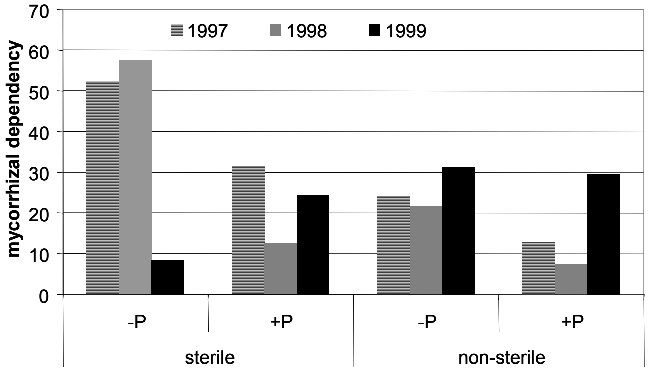

Fig. 5. Mycorrhizal dependency of horse bean.

In the present experiment since soil were fumigated the population of indigenous AM was therefore reduced which provide a low mycorhizal percentages compared to a non fumigated control or to a fumigated but re-inoculated with high mycorrhization plant. Under the fumigation conditions may be the effect of mycorhizal inoculation overestimated, because in non-sterile soils, mycorrhizal activity may be limited by other soil born micro organisms (Hetrick *et al.*, 1989).

The overall results revealed that yields were lower in fumigated plots than in the non-fumigated ones. Conversely, MBr application reduced yield compared with the non-fumigated one whether or not the plants were inoculated. Mycorhizal inoculation may have had some other benefits to the plant such as protecting it against soil-borne pathogens and environmental stress.

It appears that there are some other benefits of mycorrhizae on forage plants such as controlling disease and increasing plant resistance. We conclude that although mycorrhizal inoculation increased some vegetable yield, this increase is not easily explained through better nutrient uptake by AMF plants than by uncolonized plants (Ortas and Akpinar, 2006). For future experiment, forages plant should be tested under several management systems.

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