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Effect of Arbuscular Mycorrhizal Fungus (*Glomus Mosseae*) and Soil Yeasts Interaction on Root Nodulation, N-Fixation and Growth of Faba Bean (*Vichia faba)*

Mohamed, H.M.*

[1]*Department of Soils & Water, Faculty of Agriculture, University of Assiut, 71526, Assiut, Egypt.*

ABSTRACT

Interactions between the arbuscular mycorrhizal fungus (AMF) *Glomus mosseae* and two soil yeasts (*Saccharomyces cerevisiae* and *Candida sake*) and their effects on faba bean plants were studied in a pot experiment in sterile, phosphorus (P) deficient soil. These organisms interacted synergistically when added consecutively at 2-week intervals, where sporulation, root infection with *G. mosseae* and the populations of either soil yeast species were significant with dual inoculation, especially when soil yeast species were inoculated for two weeks prior to sowing. Plant shoot dry weight, uptake of nitrogen (N) and P by the shoots, as well as nodulation and nitrogenase activity of faba bean roots were improved by inoculation with either *G. mosseae* or the two soil yeast species. Soil yeast species *S. cerevisiae* was more effective than *C. sake*. Dual inoculation was more effective on growth, nutrition nodulation and nitrogenase activity than individual inoculation.

Keywords: ***Glomus Mosseae, Candida sake, Saccharomyces cerevisiae,* glasshouse experiment, inoculation, nodulation, *Vichia faba***

INTRODUCTION

Arbuscular mycorrhizal fungi (AMF) are widespread in nature and are a fundamental component of the agro-ecosystem. They are stable, form mutually beneficial plant-fungus associations, in which the fungus is partly inside and partly outside the host and form a living link between root and soil (Bethlenfalvay *et al.*, 1997). One of the most dramatic effects of infection by mycorrhizal fungi on the host plant is the increase in phosphorus (P) uptake by the plant (Koide, 1991), mainly due to the capacity of mycorrhizal fungi to absorb phosphate from soil and transfer it to the roots of the host (Asimi *et al*., 1980). Additionally, mycorrhizal infection results in an increase in uptake of copper (Gildon and Tinker, 1983), zinc (Lambert *et al.*, 1979), and sulphate (Buwalda *et al*., 1983). Sharma (2003) argued that the resistance against biotic and abiotic stresses is due to the effects of AMF inducing the production of plant hormones.

Yeasts are a common component of the rhizosphere in all geographic zones (Slávikova and Vadkertiova, 2003) but little is known about their function in

*Corresponding author : E-mail: hashembefoul@yahoo.com

nutrient cycling (Slávikova *et al.,* 2002) and their interactions with other soil microorganisms. Yeasts have been found in different soils and rhizosphere of various plants (Morais *et al.*, 1995; Ganter, 2006). Although the numbers of yeasts are low in comparison with other microorganisms, many investigators claimed that this group of organisms appear to play an important role in soil fertility and are capable of producing certain growth promoting substances such as hormones, amino acids, vitamins, proteins, organic acids, and soluble and volatile exudates (Sampedro et al., 2004; Boby *et al*., 2007). Nonetheless, despite the known ability of yeasts to produce organic acids, there have been very few reports on their ability to solubilise inorganic phosphate (Kanti and Sudiana, 2002; Vassileva *et al*., 2000; Hesham and Mohamed, 2011). Only a few studies have investigated AMF interactions with soil yeasts (Fracchia *et al.*, 2003; Sampedro *et al*., 2004; Gollner *et al.*, 2006).

Interactions between mycorrhizal fungi and other soil microorganisms may occur widely. Shifts in the presence or abundance of microbial species occuring in the rhizosphere of mycorrhizal plants (Linderman and Paulitz, 1992) and interaction between mycorrhizal fungi and other rhizosphere inhabitants can be detrimental to the mycorrhizal fungi and certain rhizosphere microorganisms (Posta *et al.*, 1994). Hence, this study was conducted to investigate the interactions between the AMF and yeast species, and their effects on faba bean plants.

MATERIALS AND METHODS

Preparation of Microbial Inoculums

Rhizobium leguminosarum, locally isolated from root nodules of common bean plants, was used in this study. The bacterium was cultured on yeast extract-mannitol broth (Trinick and Parker, 1982), at 28ºC on a rotary shaker at a speed of 150 RPM. Bacterial cells were harvested at the late logarithmic growth phase, centrifuged, washed with sterile distilled water, and diluted with sterile distilled water to a cell density of approximately 105 cells per ml. During seeding, 10 ml of the bacterial suspension was inoculated into each pot.

Yeast species *Saccharomyces cerevisiae* and *Candida sake* were previously isolated from a composite sample of clay soil (Hesham and Mohamed, 2011). The strains were maintained on malt-yeast-glucose-peptone agar (YM) slants at 4ºC. One-week old YM slant were scraped into sterile water to give a suspension of 106 cells per ml, 10 ml of which was added per treated pot. Each soil yeast species (*S. cerevisiae* and *C. sake*) were inoculated at 2-week intervals before or after sowing.

The spores of the AMF *Glomus mosseae* were isolated by wet sieving the soil (Gerdeman and Nicolson, 1963) from a stock culture where onion was the host plant. This species was reproduced by onion pot culture within 4 months in sterilised clay loam soil, by autoclaving twice at 121ºC for one hour. The amount of inoculum was adjusted to give about 10^4 spores per pot at sowing. The number

of spores in the soil sample was determined by Gerdeman and Nicolson's (1963) wet sieving method.

Greenhouse Experiment

The trial was carried out in the greenhouse of the Soils and Water Department, Faculty of Agriculture, Assiut University, Egypt. A pot experiment was conducted in the 2013 season to study the interactions between the AMF, *G. mosseae* and two soil yeasts (*S. cerevisiae* and *C. sake*) and their effects on faba bean plants in calcareous soil collected from the El-Gorahib Experimental Farm of Assiut University. The physical and chemical properties of the soil used in this study are presented in Table 1. The experimental design used was a complete randomised block design with six replicates of each treatment. The experiment was established with 6 treatments and one control: uninoculated control (C); inoculation with AM fungus (*G. mosseae*) (Gm); inoculation with *S. cerevisiae* (Sc); inoculation with *C. sake* (Cs); *G. mosseae* and *S. cerevisiae* (Gm + Sc); *G. mosseae* and *C. sake* (Gm + Cs). Yeast species (*S. cerevisiae* and *C. sake*) were inoculated at 2-week intervals before or after sowing.

TABLE 1
Physical and chemical characteristics of soil used.

Properties	Values
Clay (%)	3.9
Silt (%)	30.5
Sand (%)	60.2
Texture grade	Sand loam
Total $CaCO_3$ (%)	16.18
EC dS cm^{-1} (1:1)	1.12
pH (1:1 suspension)	8.3
Total nitrogen (%)	0.005
Organic matter (%)	0.28
Available (P mg g^{-1} soil)	5.60
Exchangeable cation ($cmol_c$ kg^{-1}):	
Ca^{2+}	0.51
Mg^{2+}	0.26
Na^{+}	0.33

Faba bean seeds (*Vichia faba*) cv. Assiut-115 were surface sterilised by shaking in 7% calcium hypochlorite for 10 minutes, rinsed with sterile distilled water and sown (4 seeds per pot) in 30-cm diameter plastic pots containing 5 kg sieved calcareous soil. The soil was autoclaved twice at 1.2 kg cm^2 pressure and 121ºC for one hour at a time. The pots were irrigated to field capacity (47%) during the experimental period under greenhouse conditions. After emergence, the seedlings were thinned to two uniform plants per pot. Plants were harvested 45 days after sowing. The root systems of each six pot replicates per treatment were divided into three batches. In the first batch, mycorrhizal root infection

was measured after clearing and staining 1 cm root segments with tryphan blue (Philips and Hayman, 1970). In the second batch, the number, fresh and dry weights of the nodules and the dry weight of the roots were determined. In the third batch, nitrogenase activity of the nodulating faba bean root was assayed by the acetylene reduction method (Hardy *et al.*, 1973). Unwashed plant roots were placed immediately in a canning jar fitted with a serum stopper for gas sampling. To prevent drying, the roots in the sampling jars were covered with some of the soil that had been removed from them earlier. Also, control jars contained only soil from each treatment. Ten percent of the gaseous atmosphere in the jar was removed and replaced by acetylene (C_2H_2). The jars were then tightly sealed with Parafilm M® and incubated at 30ºC for 24 hours. A volume of 0.1 ml gas sample from each jar was removed and injected into a Pye Unicome 104-inch gas chromatograph containing a flame ionisation detector and a 5 ft. x 118-inch glass column of activated alumina (80-100 mesh). The oven temperature was set at 150º C, and the carrier gas was nitrogen (N) at a flow rate of 30 ml/min. Dried shoots were ground and submitted to the acid-digestion using a 2:1 HNO_3: $HClO_4$ acid mixture for determination of N and P uptake. The population of each soil yeast (*S. cerevisiae* and *C. sake*) in the rhizosphere soil was measured by dilution plate count on malt-yeast-glucose-peptone agar medium at 2, 4 and 6 weeks after sowing, The number of *G. mosseae* spores in the soil was determined using the wet sieving and decanting method (Gerdeman and Nicolson, 1963).

Statistical Analysis

The data reported in this paper are the mean values based on the six replicates. Differences among treatments were tested by ANOVA and mean values among treatments were compared using Duncan's multiple range test at P = 0.05. Statistical analyses of the data were performed using the statistical computer program (Statsoft, 1995).

RESULTS

Interactions Between G. mosseae and Soil Yeast Species

Soil yeast species *S. cerevisiae* or *C. sake* significantly stimulated sporulation and mycorrhizal infection when inoculated with *G. mosseae*, either pre-sowing or at sowing of faba bean seeds (Table 2). The response was very apparent when inoculation was carried out 2 weeks prior to sowing and prominently so when *S. cerevisiae* was applied. Post-sowing inoculation of yeast species did not affect sporulation or mycorrhizal infection.

Dual inoculation with *G. mosseae* and soil yeast species in the rhizosphere significantly increased the population density when the latter was inoculated 2 weeks prior to sowing (Table 2). This response did not occur with inoculation 2 weeks post-sowing. Irrespective of application time, the population density of either soil yeast species doubled every 2 weeks. The population of *S. cerevisiae* was higher than that of *C. sake*, especially 2 weeks after sowing.

TABLE 2
Interaction between *G. mosseae* and soil yeast strains (*S. cerevisiae* and *C. sake*) in the rhizosphere soil of faba bean plants.

Inoculation time for soil yeasts	Inoculation* treatment	G. *mosseae* spore g^{-1} dry soil	Infection by G. *mosseae* (%)	Soil yeasts population c.f.u. x 10^3 g^{-1} dry soil Weeks after sowing		
				2	4	6
At sowing	C	0^{d}	0^{d}	0^{c}	0^{e}	0^{e}
	G m	27^{c}	46^{c}	0^{c}	0^{e}	0^{e}
	Sc	0^{d}	0^{d}	2.53^{b}	4.56^{d}	5.86^{d}
	Cs	0^{d}	0^{d}	2.70^{b}	5.83^{c}	6.96^{c}
	Gm + Sc	52^{a}	60^{a}	3.56^{a}	7.30^{a}	9.76^{a}
	Gm + Cs	39^{b}	52^{b}	3.32^{a}	6.80^{b}	8.90^{b}
2 weeks pre-sowing	C	0^{c}	0^{d}	0^{d}	0^{d}	0^{e}
	G m	33^{b}	46^{c}	0^{d}	0^{d}	0^{e}
	Sc	0^{c}	0^{d}	3.63^{b}	5.90^{b}	10.26^{b}
	Cs	0^{c}	0^{d}	2.83^{c}	4.63^{c}	7.63^{d}
	Gm + Sc	56^{a}	71^{a}	4.80^{a}	7.70^{a}	12.66^{a}
	Gm + Cs	45^{b}	62^{b}	3.90^{b}	5.80^{b}	8.46^{c}
2 weeks post-sowing	C	0^{c}	0^{c}	0^{c}	0^{d}	0^{d}
	G m	32^{b}	46^{b}	0c	0^{d}	0^{d}
	Sc	0^{c}	0^{c}	0.76^{ab}	2.36^{b}	5.11^{c}
	Cs	0^{c}	0^{c}	0.73^{ab}	2.03^{c}	4.90^{c}
	Gm + Sc	34^{a}	50^{a}	0.90^{a}	3.96^{a}	7.72^{a}
	Gm + Cs	31^{b}	47^{b}	0.56^{b}	3.80^{a}	6.80^{b}

* C: uninoculated soil; Gm: *Glomus mosseae*; Sc: *Saccharomyces cerevisiae*; Cs: *Candida sake*.
The values in columns followed by the same letter (s) are not significant at 5% significance level by Duncan's multiple range test.

Nodulation and Nitrogenase Activity

The effect of inoculation with *G. mosseae* and two soil yeast species on nodulation and nitrogenase activity varied with the treatments (Table 3). Under all inoculation conditions, neither soil yeast species significantly affected the number of nodules, fresh and dry weight per plant, nor nitrogenase activity per unit nodule dry weight. The dual inoculation with *G. mosseae* significantly increased nodulation and weight as well as nitrogenase activity per unit nodule dry weight. G. mosseae alone had no effect on nitrogenase activity, but the coupling of either soil yeast species stimulated nitrogenase activity per unit nodule dry weight, almost to the same extent, regardless of time of application of these organisms.

Plant Growth and Uptake of Nutrients

There was a significant interaction effect between soil yeast species and mycorrhizal inoculation on plant growth (Table 4). Soil yeast species affected the dry weight of either shoots or roots of faba bean plants, except when *S. cerevisiae* was inoculated 2 weeks before sowing. Whilst inoculation with *G. mosseae* increased the dry weight of either shoots and roots of faba bean plants, dual inoculation with *G. mosseae* and yeast species significantly increased the dry weight above that of the *G. mosseae* treatment alone. Maximum weights were achieved when inoculation was performed 2 weeks before sowing.

TABLE 3
Effect of inoculation with G. mosseae or/and soil yeast strains (*S. cerevisiae* and *C. sake*) on nodulation and nitrogenase activity in faba bean plants.

Inoculation time for soil yeasts	Inoculation* treatment	Nodulation			Nitrogenase activity nm C_2H_2 h^{-1} per plant
		Number per plant	Fresh wt. mg^{-1} plant	Dry wt. mg^{-1} plant	
At sowing	C	36^{e}	476.5^{b}	72.3^{e}	1.63^{b}
	G m	50^{c}	607.4^{ab}	96.0^{c}	$1.90a^{b}$
	Sc	41^{d}	545.2^{b}	82.3^{d}	$1.86a^{b}$
	Cs	55^{b}	596.4^{ab}	80.3^{d}	1.80^{ab}
	Gm + Sc	63^{a}	715.9^{a}	109.4^{a}	2.26^{a}
	Gm + Cs	47^{c}	602.8^{ab}	101.5^{b}	2.00^{ab}
2 weeks pre-sowing	C	36^{e}	542.5^{b}	73.4^{e}	1.76^{b}
	G m	53^{c}	644.7^{ab}	99.8^{c}	1.86^{ab}
	Sc	44^{d}	609.5^{ab}	90.9^{d}	1.96^{ab}
	Cs	43^{d}	680.5^{ab}	97.7^{c}	2.06^{ab}
	Gm + Sc	65^{a}	746.9^{a}	113.9^{a}	2.50^{a}
	Gm + Cs	60^{b}	627.2^{ab}	106.3^{b}	1.90^{ab}
2 weeks post-sowing	C	35^{d}	549.7^{bc}	75.8^{d}	1.66^{b}
	G m	51^{b}	633.6^{ab}	97.4^{b}	1.83^{b}
	Sc	42^{c}	494.3^{c}	74.5^{d}	1.73^{b}
	Cs	38^{cd}	547.5^{bc}	83.0^{c}	1.66^{b}
	Gm + Sc	60^{a}	720.3^{a}	108.8^{a}	2.13^{a}
	Gm + Cs	56^{a}	605.1^{b}	91.5^{b}	1.90^{b}

* C: uninoculated soil; Gm: *Glomus mosseae*; Sc: *Saccharomyces cerevisiae*; Cs: *Candida sake*. The values in columns followed by the same letter (s) are not significant at 5% significance level by Duncan's multiple range test.

TABLE 4
Effect of inoculation with *G. mosseae* or/and soil yeast strains (*S. cerviceae* and *C. sake*) on plant growth, nitrogen and phosphorus uptake of faba bean plants.

Inoculation time for soil yeasts	Inoculation* treatment	Dry weight g per plant		N-uptake mg per plant	P-uptake mg per plant
		Shoots	Roots		
At sowing	C	0.97^{e}	0.68^{e}	27.87^{c}	1.59^{bc}
	G m	1.41^{bc}	0.89^{c}	28.56^{abc}	2.12^{a}
	Sc	1.23^{cd}	0.79^{d}	28.80^{abc}	1.60^{c}
	Cs	1.07^{de}	0.76^{d}	29.60^{a}	1.63^{b}
	Gm + Sc	1.98^{a}	1.09^{a}	29.33^{ab}	2.09^{a}
	Gm + Cs	1.62^{b}	0.99^{b}	28.30^{cb}	2.15^{a}
2 weeks pre-sowing	C	0.98^{e}	0.67^{e}	28.36^{b}	1.53^{e}
	G m	1.44^{c}	0.90^{c}	28.64^{b}	2.14^{a}
	Sc	1.26^{d}	0.80^{d}	30.23^{b}	1.71^{c}
	Cs	1.08^{e}	0.69^{e}	29.26^{b}	1.61^{d}
	Gm + Sc	2.31^{a}	1.16^{a}	34.40^{a}	2.20^{b}
	Gm + Cs	1.89^{b}	1.05^{b}	30.06^{b}	2.18^{b}
2 weeks post-sowing	C	1.00^{cd}	0.68^{b}	28.3^{a}	1.53^{d}
	G m	1.47^{b}	0.85^{b}	28.7^{a}	2.12^{b}
	Sc	1.13^{c}	0.74^{b}	27.5^{a}	1.55^{d}
	Cs	0.95^{d}	0.76^{b}	29.3^{a}	1.65^{c}
	Gm + Sc	2.04^{a}	0.97^{a}	32.3^{a}	1.97^{a}
	Gm + Cs	1.37^{b}	0.90^{b}	29.8^{a}	1.93^{ab}

* C: uninoculated soil; Gm: *Glomus mosseae*; Sc: *Saccharomyces cerevisiae*; Cs: *Candida sake*. The values in column followed by the same letter (s) are not significant at 5% significance level by Duncan's multiple range test.

With regard to nutrient-uptake, the inoculation of soil yeast species, alone or in the presence of *G. mosseae*, did not affect faba bean shoot N-uptake (Table 4). Whereas, the P-uptake was significantly affected by the presence of either soil yeast species except for *S. cerevisiae* inoculated 2 weeks before sowing. *G. mosseae*, alone or coupled with soil yeasts, increased P accumulation in faba bean shoots.

DISCUSSION

This study showed a significant increase in infection and spore production of *G. mosseae* as a result of its association with either soil yeast species (*S. cerevisiae* or *C. sake*), with the former being more effective than the latter. The results obtained in the present study were consistent with the findings of Boby *et al*. (2008) who investigated all the yeasts (*Rhodotorula mucilaginosa, Metschnikowia pulcherrima, Trichosporon cutaneum* var. *cutaneum*, *S. cerevisiae, Cryptococcus laurentii*, and *Debaryomyces occidentalis* var. *occidentalis*) to show a synergistic interaction with the *G. mosseae* with significant increases in mycorrhizal root colonisation and spore numbers. Singh *et al*. (1991) reported an increase in the production of vesicles, arbuscular and spores of native AMF due to inoculation with S. cerevisiae in legumes. Furthermore, Fracchia *et al.* (2003) reported the enhancement of arbuscular mycorrhizal colonisation of soybean and red clover with the application of the yeast *R. mucilaginosa* to the soil. In contrast, Gollner *et al*. (2006) showed that presence of soil yeasts did not significantly affect mycorrhizal colonisation of maize roots, but negatively affected the length of AMF extraradical mycelium. The yeasts may enhance AMF development by supplying vitamin B12 to the rhizosphere, as AMF have been shown to be stimulated by this vitamin (Singh *et al*., 1991). Thus, vitamin B12 produced by the soil yeasts might have resulted in better plant growth and yield in plants treated with both *G. mosseae* and soil yeasts. The observations of Boby *et al*. (2008) show the effect of inoculation with *S. cerevisiae* on non-mycorrhizal plants to be negligible, while it did increase the root colonisation and spore count of mycorrhizal plants. This suggests that the yeasts specifically stimulate arbuscular mycorrhizal development rather than the host plant, which upholds the observation made by Larsen and Jacobsen (1996). It is quite possible that Vitamin B12 production by soil yeasts could be the main reason for the stimulation of mycorrhizal development observed in this study and needs further investigation. Time of soil yeasts inoculation played a role in sporulation and infection of mycorrhizal fungus. Two weeks post sowing seemed to have a lower effect. This might be attributed to the age of soil yeasts (being only 4 weeks old) compared with inoculation 2 weeks pre-sowing (8 weeks old). Under the latter condition, the soil yeast count was double that under the former condition. Soil yeasts seemed to secrete moderately harmful metabolites which hinder, to a certain extent, spore formation by *G. mosseae.*

The population of soil yeasts in the root zone soil was stimulated in plants inoculated with *G. mosseae* + soil yeasts. The stimulatory effects of *G. mosseae* on the activity of either test soil yeast species may be attributed to alterations in the

root exudates. AMF can significantly influence the microflora in the rhizosphere directly through fungal exudates or indirectly through altering root exudation patterns (Linderman, 1992). Boby *et al.,* (2008) found that soil yeasts in the root zone soil were stimulated in plants inoculated with *G. mosseae* + soil yeasts. The plants inoculated with *G. mosseae* alone also had a higher yeast population in the root zone soil compared to uninoculated plants. These results contradict those of Sampedro *et al*., (2004) who found no effect due to the presence of the AMF G. mosseae on populations of soil yeasts (*R. mucilaginosa, C. laurentii* and *S. kunashirensis*) in the rhizosphere of soybean. Fracchia *et al.,* (2003) observed a similar population of *Rhodotorula mucilaginosa* in rhizosphere of soybean inoculated with *G. mosseae* and in rhizosphere of red clover inoculated with *Gigaspora rosea.*

The results further showed that nodule number, fresh and dry weights and nitrogenase activity were highly apparent due to treatment with *G. mosseae*. Coupling either yeast species with *G. mosseae,* in the soil under the test condition, increased the efficacy of the latter organism to initiate nodulation, better nitrogenase activity and growth of faba bean plants. Singh *et al.,* (1991) report that the increases in nodule numbers and dry weights of legumes due to inoculation with yeasts were caused by stimulation of the legumes' indigenous microflora. Similarly, the increases in nodulation and other symbiotic parameters of forage legumes (*Trifolium alexandrium* and *Medicago sativa*) due to combined inoculation of yeasts (*S. cerevisiae* and *Candida torpicalis*) and specific *Rhizobium* sp. have been reported earlier (Tuladhur and Sub Rao, 1985) and were also attributed to the stimulatory action of yeasts on the multiplication of native rhizobia (Tuladhur, 1983).

Similarly, *G. mosseae* stimulated growth and the uptake of N and P in the faba bean shoot system, regardless of inoculation time, whereas *C. sake* alone showed no significant effect. The same applied to *S. cerevisiae* when inoculated 2 weeks before sowing indicating the difference in metabolic activity of these two organisms. The former seemed to produce metabolic activators (bio-regulators) that stimulated the activity of faba bean plants. Many investigators claimed that yeasts seemed to play an important role in soil fertility and are capable of producing certain growth promoting substances such as hormones, amino acids, vitamins, proteins, organic acids and soluble and volatile exudates (Sampedro *et al*., 2004; Boby *et al*., 2007; Hesham and Mohamed, 2011). Dual inoculation with *G. mosseae* and two soil yeast species highly stimulated the growth of the faba bean regardless of the type of treatment, indicating the synergistic effect between the two types of organisms. However, the efficacy of these organisms for N-uptake in faba bean shoot was unaffected by either type of treatment, except when inoculated 2 weeks pre-sowing, indicating that the soil yeasts had a significant role in the synergistic efficacy of these organisms. The efficacy of *G. mosseae* for P accumulation was highly apparent when either soil yeast was coupled with *G. mosseae*. AMF are known to improve P nutrition of plants, especially in P

deficient soil, and can translocate phosphate by scavenging a larger volume of soil with extensive hyphae (Kothari *et al.*, 1990; Ortas *et al.*, 2002).

Some studies reveal that interactions between soil microorganisms and AMF are important for plant growth (Azco´n-Aguilar *et al.*, 2002; Barea *et al*., 2002). Gollner *et al.,* (2006) showed that inoculation with soil yeasts and AMF can significantly affect the shoot dry weight of maize. Moreover, this study found specific effects of certain combinations of mycorrhizal inoculation and yeast species on plant biomass. In plants not inoculated with AMF, only *C. sake* increased plant growth compared to plants inoculated with Glomus intraradices where all two yeasts showed positive effects on shoot dry weight. Similar results were obtained by Bhowmik and Singh (2004) when inoculation of *Chloris guyana* with the yeast *S. cerevisiae* alone did not affect plant growth. However, inoculation with the yeast and the AMF *G. mosseae* resulted in a significant increase in plant biomass.

CONCLUSION

In conclusion, the results indicated that the soil yeasts (*S. cerevisiae* and C. *sake*) and AMF (*G. mosseae*), generally exhibit positive mutual relationships. Dual inoculation of faba bean with soil yeast and AMF increased shoot and root biomass, nodulation, nitrogen-fixing activity, and uptake of N and P. Soil inoculation with these microorganisms contributed to improvement in the growth of faba bean plants and they may be applied to other crops for better growth. Future studies therefore should be focused on selecting compatible and efficient combinations of AMF and soil yeasts species for successful use in reestablishing vegetation of disturbed ecosystems.

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