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Influence of Arbuscular Mycorrhizal Fungus and Kinetin on the Response of Mungbean Plants to Irrigation by Seawater

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An experiment was carried out to investigate the effects of pre-inoculation with the mycorrhizal fungus *Glomus clarum* and foliar application of kinetin on the growth of mungbean plant irrigated wht different dilutions of seawater. Arbuscular-mycorrhizal (AM) infection significantly increased dry weight, height, chlorophyll, sugar and protein content, nitrogen and phosphorus-use efficiencies, leaf conductivity, transpiration rate, nitrogenase, acid and alkaline phosphates activities of all salinized mungbean plants in comparison with control and non-mycorrhizal plants irrespective of the presence or absence of kinetin. Mycorrrhizal plants showed higher concentrations of N, P, K, Ca and Mg and lower Na/N, Na/P, Na/K, Na/Ca and Na/Mg ratios than non-mycorrhizal plants when irrigated with certain dilutions of seawater. Mungbean plants showed 597% and 770% dependency on AM fungus *G. clarum* in absence and presence of kinetin, respectively, for biomass production under a level of 30% of seawater. The average value of tolerance index for mycorrhizal plants accounted 267% and 364% in absence and presence kinetin respectively. This study provides evidence for the benefits of kinetin which are actually known for mycorrhizal than non-mycorrhzal plants. AM fungus and kinetin protected the host plants against the detrimental effects of salt. However, mycorrhizal infection was much more effective than kinetin applications. Thus management applications of this arbuscular mycorrhizal endophyte (*G. clarum*) with kinetin could be of importance in using seawater in certain dilutions for irrigation in agriculture.

KEYWORDS **Arbuscular mycorrhizal fungus, Kinetin, Mungbean, Seawater irrigation**

Agricultural practices demand more water than any other activity on this plants. Currently, about 65% of the water removed from all sources world-wide is used for irrigation (Postal, 1997). Current supplies of good quality surface and ground water for crop production have not kept pace with the rapid increase in water demand associated with increased cropping intensities (Pimentel *et al.*, 1999) and/or expansion in irrigated agricultural on marginal land. Water demands exceed supplies in nearly 80% countries which have 40% population of the world.

Saline water is being used in situations when there is insufficient good-quality water for agriculture. Salinity and sodicity are the principal water quality concerns when irrigate with saline water (Ayars and Tanji, 1999). Soil salinity may also result from the nearness of semi-arid sites to the sea or due to the rise of saline ground water into the root zone and concentrating there when evaporation becomes excessive. In Egypt, about 96% of the land is desert and rises saline ground water into root zone in most cultivated lands.

Salinity limits on crop productivity and quality. The majorities of crops are salt-sensitive and can't survive under condition of salinity or can survive only with decreased yields. Plants are stressed in three ways by salinity; (1) low water potential of the root medium leads to water chlorine, (2) toxic effects of ions mainly sodium and chlorine, and (3) nutrient imbalance caused by depression in uptake and/or shoot transport (Marschner, 1995; Adiku *et al.*, 2001). To alleviate the deleterious effects of salinity some rehabilitations such as reclamation of salinized lands, improving of irrigation saline water, chemical amendments to soil and special cultural techniques are applied. Correcting salinity problems is expensive and representing only a temporary solution (Dasgan *et al.*, 2002). Development of cultivars that can produce economic yields under saline conditions as well as phytoremidation involves cultivation of certain symbiont microorganisms, which help more permanently and complementarily plant tolerance to saline (Fooland, 1996; Qadir *et al.*, 2001).

Arbuscular mycorrhizal (AM) fungi are associated with the roots of over 80% of terrestrial plant species (Heijden *et al.*, 1998). The symbiosis between the AM-fungi and the roots improves water use and nutrient uptake, expecially elements with low soil mobility, such as phosphorus, zinc, and copper. AM-fungi associations increase plant tolerance to various biotic and a biotic factors (Smith and Read, 1997).

AM-fungi associations may increase plant tolerance to salinity (Nielsen *et al.*, 1999; Al-Karaki, 2000; Diallo *et al.*, 2001; Yano-Melo *et al.*, 2002). AM-colonized plants have optional and alternative mechanisms available to satisfy their nutritive requirements and to maintain their physiological statues in stress situations and in disturbed ecosys-

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tems (Copeman *et al.*, 1996; Rilling and Steinberg, 2002). Improved salt tolerance of mycorrhizal plants can be related to enhanced mineral nutrition, improvements in physiological process like photosynthetic activity or water use efficiency and production of osmoregulators (Auge and Stodola, 1990; Ruiz-Lozano and Azcon, 2000).

Growth regulators such as gibberellic acid (Khan and Rizvi, 1994), kinetin (Ungar, 1991) and fusicossin (Pyler and Proseus, 1996) are known to alleviate the inhibitory effects of salinity on plants. Khan *et al.* (2000) reported that both gibberellic acid and kinetin alleviated some of inhibitory effects of salinity at 800-mole m^{-3} NaCl on shoot growth of *Suaeda fruticosa* while root growth was promoted by kinetin. Nemat-Alla *et al.* (2002) proved that kinetin alleviates the stress symptoms and regulates the changes in phenolic metabolism of water logged or salinity treated *Vigna sinensis* and *Zea majs*.

Mungbean (*Vigna Radiata* L) is a fast maturing-N_2-fixing leguminous crop (70~90 days) with relatively low fertilizer requirements and good drought tolerance. Mungbean is widely grown, particularly in developing countries, where it is consumed directly or used as a source of starch. Mungbean seeds surpass lentil and broad bean in calcium, iron and vitamin A contents and contain nearly the same percentage of protein (a bout 23 to 24%). Introducing of new high yielding fast matural food crops may help narrow the food gab in Egypt and other developing countries.

The objective of this work was to study the effects of pre-inoculation with the mycorrhizal fungus *Glomus clarum* and foliar application of kinetin on the growth and physiology of mungbean plant irrigated with different concentrations of seawater.

Materials and Methods

The experiment examined factorial combinations of 1) two mycorrhizal treatments a) non-inoculated control and b) inoculated with *G. clarum*, 2) two kinetin treatments a) non-treated and b) treated, and 3) four seawater concentrations 0, 10, 20 and 30% of seawater. Four replications per treatment were used to give a total of 64 pots.

A reclaimed calcareous soil was obtained from a surface layer (0~20 cm) in Burg El-Arab region, Egypt. The soil was dried and then ground to pass through a 2.0 mm sieve. This soil contained 63.9% sand, 2.9% silt and 34.2% clay, 34.9% CaCo3, 0.6% total nitrogen, 2.1 ppm available phosphorus, 0.45-mg/100 gm available potassium and 1.8 m moh/m E.C.

The mycorrhizal inocula consisted of soil, spores, mycelium and infected root fragments obtained from open pot culture of onion. AM species used, *G. clarum* was provided from Faculty of Science, Mansoura University, Egypt.

Growth conditions. The experiment was conducted between June and August 1998 in Botany Department, Faculty of Science, Zagazig University. Seeds of mungbean (V2010) obtained from Agronomy Department, Agric. Res. Center, Giza, Egypt were surface sterilized with 7% calcium hypochlorite for 20 min and subsequently washed with distilled water. Seeds were distributed at a rate of 4 grains/pot (30 cm diameter containing 2 kg of soil), and then thin to 1 seedling/pot 7 days after sowing. The pots were pre-inoculated with *G. clarum*. The inoculum was placed 3 cm below the surface of the soil before sowing date. The non-inoculated pots were supplied with filtered washing of inoculum's to supply the same micro flora other than mycorrhizal fungus. Half of the plants were sprayed with 25 *ml*/plant of 0.5 mM kinetin solution one and two weeks after sowing. Plants for each treatment were watered with equal amounts of the suggested concentrations of the seawater to maintain soil moisture near field capacity. The plants were harvested 8 weeks after planting from sowing date.

Analytical methods.

A-Griwth parameters: At the end of the experiment, plant height, dry matter of the roots, shoots and the whole plant as well as root to shoot ratio (R/S) were measured. Nitrogen was extracted with sulfuric acid using semi-micro Kjeldahl method (Jackson *et al.*, 1973). Phosphorus was extracted by nitric-perchloric acid digestion and measured using the Vanadono-molybdophosphoric colometric method (Jackson, 1967). Potassium and sodium were assayed by using flame spectrophotometer, while calcium and magnesium were determined by atomic absorption (Allen *et al.*, 1984). Sugar and protein contents of plants were estimated according to Naguib (1963) and Bradford (1976), respectively.

At each seawater concentration, the mycorrhizal dependency (M.D.) of the plants was calculated according to Gerdemann (1975) as:

$$\text{M.D.} = (\text{d.w. mycorrhizal plant} \times 100) / \text{d.w. non-mycorrhizal plant}$$

Tolerance indices (Ti) of mycorrhizal and non-mycorrhizal mungbean plants to seawater were determined according to Shetty *et al.* (1995) as:

$$\text{Ti} = \text{D.W. plant at sea water level} \times 100 / \text{D.W. plant at 0.0\% seawater}$$

Some photosynthetic pigments (chlorophylls a, b & carotenoids) of plant leaves were extracted and determined by the method of Harbon (1984). Stomatal conductance, gas exchange (Gs) and transpiration rate (T) of unshaded leaves were measured in one leaf per plant using Li-COR model 1600 diffusion prometer between 11.30 and 1.30 h (Beardsel *et al.*, 1972).

Shoot nitrogen- and phosphorus-use efficiency were

determined as the ratio of the shoot dry weight (milligram) produced per milligram of the total shoot nitrogen or phosphorus (Ruiz-Lozano and Azcon, 2000).

Immediately after harvest, part of the root system was washed carefully in 4°C water to remove the adhering soil particles for quantitative and qualitative assay of soluble acid and alkaline phosphatases (Gianinazzi-Pearson and Gianinazzi, 1976). The remaining part of the root system was cut into 0.5~1.5 cm segments, cleared in 10% KOH and stained with tryptopan blue in lactophenol (Phillips and Hayman, 1970). To estimate the intensity of infection M% and arbuscular development in the infected region of the roots (A%) were estimated in root samples stained for total infectio by the method of Trouvelot *et al.* (1986).

The nitrogenase activity of roots was measured at the end of expriment using the acetylene reduction assay (Hardy *et al.*, 1973). Values of nitrogenase activity were recorded as moles C_2H_4/gm plant/h.

Statistical analysis. The data were statistically analyzed as a complete randomized block design. Means were compared using the least significant difference (L.S.D) procedure (Steel and Torri, 1960) at 0.05.

The effects of mycorrhizal infection, concentration of seawater and kinetin application on measured parameters were subjected to statistical analysis of variance (ANOVA) (Hicks, 1983).

Results and Discussion

As previously reported, increasing uses of saline waters in irrigation markedly changed the physical and chemical properties of the exposed soil. These changes include increase in the soil salinity (Shannon and Grieve, 1999). Current evidence indicates that the level of seawater can be considered as an important factor controlling nutritive or morphological characters of the grown plants (Rao and Take, 2002; Masarrat, 2002). In this connection our results revealed that the growth parameters of salinized non-mycorrhizal mungbean plants irrespective of presence or absence of kinetin were significantly reduced in comparison with the control.

Mycorrhizal symbiosis is a key component in helping plants to cope with adverse environmental conditions. The beneficial effects of mycorrhiza on the growth under saline conditions have been demonstrated in various plant species and families (Diallo *et al.*, 2001; Yano-Melo *et al.*, 2002; Muhsin and Zwiazek, 2002). In this study, the overall growth of mungbean plants infected with *G. clarum* was higher as compared to non-infected plants under irrigation with different levels of seawater (Table 1). However, the presence of kinetin was induced growth improvement in mycorrhizal and non-mycorrhizal plants.

Moreover, the highest improvement in dry matter content were 780% and 777% in mycorrhizal plants treated with kinetin at 20% and 30% respectively. These findings agree with previous results (Mathur and Vyas, 1999; Jalaluddin, 1993; Nemat-Alla, 2002), suggesting that management of AM symbiosis in presence of kinetin could not only improve growth but also help in overcoming the detrimental growth effects of salt stress induced by sequential irrigation with various levels of seawater.

Table 1. Effect of different dilutions of seawater on dry weight, root shoot ratios, plant height and tolerance index of salinity of mycorrhizal and nonmycorrhizal mungbean plants with and without foliar application of kinetin

	Dry weight gm/plant	R/s	Plant height cm	Tolerance index
Control				
Non-mycorrhizal plant	3.627	0.024	40.1	
Mycorrhizal plant	10.896	0.014	51.3	
Non-mycorrhizal plant + kinetin	3.811	0.024	35.5	
Mycorrhizal plant + kinetin	11.587	0.015	56.6	
10% seawater				
Non-mycorrhizal plant	2.667	0.035	25.3	73.5%
Mycorrhizal plant	13.266	0.195	50.5	365%
Non-mycorrhizal plant + kinetin	6.176	0.012	28.8	170%
Mycorrhizal plant + kinetin	16.206	0.279	46.6	446.8%
20% seawater				
Non-mycorrhizal plant	1.865	0.041	24.5	51%
Mycorrhizal plant	10.091	0.167	38.6	278%
Non-mycorrhizal plant + kinetin	3.018	0.014	18.1	89.2%
Mycorrhizal plant + kinetin	14.562	0.24	39.2	407%
30% seawater				
Non-mycorrhizal plant	1.123	0.007	21.8	30%
Mycorrhizal plant	6.710	0.142	27.3	185%
Non-mycorrhizal plant + kinetin	2.163	0.017	16.9	59%
Mycorrhizal plant + kinetin	8.734	0.215	39.2	240%
L. S. D. $p \leq 0.01$	2.67	0.013	6.31	

Root:shoot (R/S) dry weight ratio was increased in mycorrhizal plants with increasing seawater level in irrigation water, irrespective of presence or absence of kinetin. Based on this data it is conceivable to conclude that AM symbiosis may protect the mungbean plant from salt stress by increasing water surface area. This finding has been also proved by the previous workers (Kothari *et al.*, 1990; Ruiz-Lozano and Azcon, 2000).

The average height of the mungbean plants was decreased with increasing levels of seawater. The plant height of mycorrhizal plants was greater as compared to the non-mycorrhizal plant, irrespective of the presence or absence of kinetin. This may be attributed to the role of mycorrhizal symbiosis for increasing root length and branches of plants (Coperman *et al.*, 1996; Rao and Tak, 2002).

The tolerance index of non-mycorrhizal and non-kinetin plants was 30% at a level of 30% seawater (Table 1). On the other hand, tolerance index of non-mycorrhizal mungbean plants treated with kinetin was decreased from 170% at level of 10% seawater to 59% at level 30% of seawater. The role of kinetin to protect plants from salinity was previously proven by Khen and Ungar (1997) and Sawan *et al.* (2000). Thus, mycorrhizal infection improved tolerance index of mungbean plants growing under salt stress compared to non-mycorrhizal plants. The average value of tolerance index for mycorrhizal plants was 276% in absence of kinetin, while in the presence of kinetin it was 364%, indicating that the benefits of kinetin are actually known for mycorrhizal than non-mycorrhizal plants. According to aforementioned results, it is conceivable to suggest that AM-fungi may play an important role in using saline water for irrigation.

AM has been reported to increase stomatal conductance and transpiration rate (Auge *et al.*, 1995; Ruiz-Lozano and Azcon, 1995; Muhsin and Zwiazek, 2002). It appears from the present study that leaf conductivity and transpiration rate of mungbean plants were increased by rising level of seawater in irrigation water. However, these measurements were still higher in mycorrhizal plant than non-mycorrhizal one irrespective of presence or absence of kinetin (Table 2). This result indicates on *G. clarum* act as salt tolerance inducers for plants that subjected to salt stress. In this connection, evidence from the previous study (Ruiz-Lozano and Azcon, 2000; Diallo *et al.*, 2001) indicates that the effect of VAM fungi on salt tolerance seemed to be based on increased gas exchange (increased photosynthetic rate, transpiration, stomatal conductance and water use efficiency) rather than on nutrient uptake. Meroguihae *et al.* (2002) found that transpiration rate, stomatal conductivity, the demand for water and concentration of Na^+ and/or Cl^- ions were increased in mycorrhizal plants subjected to salt stress. Based on these data and on existing literatures, the greater salt tolerance of mycorrhizal plants may be the result of increasing stomatal conductance and transpiration rate.

The total chlorophyll content of the plants was not reduced significantly with increasing seawater concentra-

Table 2. Effect of different dilutions of seawater on leaf conductivity, transpiration rate, chlorophyll and sugar contents of mycorrhizal and nonmycorrhizal mungbean plants with and without foliar application of kinetin

	Leaf conductivity mmho/m	Transpiration rate	Chlorohpyll content ug/gm				Sugar content gm/plant
			Total	a	b	c	
Control							
Non-mycorrhizal plant	27.2	1.297	2.44	1.29	0.79	0.36	1.56
Mycorrhizal plant	10.6	0.316	5.133	2.98	2.03	0.123	4.72
Non-mycorrhizal plant + kinetin	37.2	1.11	3.72	1.71	1.8	0.21	1.64
Mycorrhizal plant + kinetin	10.2	0.411	5.185	2.83	2.23	0.129	4.51
10% seawater							
Non-mycorrhizal plant	38.2	1.301	2.16	1.1	0.67	0.39	0.72
Mycorrhizal plant	49.6	1.436	4.85	2.74	2.06	0.09	5.0
Non-mycorrhizal plant + kinetin	40.1	1.352	2.39	1.13	0.97	0.29	0.94
Mycorrhizal plant + kinetin	66.8	1.972	3.82	1.42	2.17	0.23	3.05
20% seawater							
Non-mycorrhizal plant	47.5	1.386	1.75	0.96	0.45	0.34	0.65
Mycorrhizal plant	69.3	2.013	4.3	2.32	1.83	0.15	5.25
Non-mycorrhizal plant + kinetin	53.6	1.62	1.84	1.12	0.53	0.19	0.78
Mycorrhizal plant + kinetin	74.0	2.415	2.75	1.16	1.36	0.25	3.85
30% seawater							
Non-mycorrhizal plant	48.2	1.422	1.66	0.89	0.43	0.34	0.63
Mycorrhizal plant	76.0	2.592	3.62	2.05	1.55	0.02	6.59
Non-mycorrhizal plant + kinetin	56.0	1.792	1.74	1.1	0.45	0.19	0.66
Mycorrhizal plant + kinetin	74.0	2.415	2.75	1.16	1.36	0.25	3.32
L. S. D. $p \leq 0.01$	7.83	0.093	0.43				3.26

tion as compared to control. However, total chlorophyll content of mycorrhizal plants exhibited higher values than that corresponding non-mycorrhizal plants. Moreover, similar trend was observed for chlorophyll a and chlorophyll b.

The total sugar content in AM plants was increased as the level of seawater increased. On the other hand, non-infected plants had lower sugar content with higher levels of seawater. However, it was observed that the presence of kinetin may suppress the sugar accumulation in mycorrhizal plants and the maximum decrease was recorded at a concentration of 30% seawater. Similar observations were previously reported by Chopra *et al.* (1998).

Deleterious effects of salinity are thought to result from low water potentials, ion toxicities, nutrient deficiencies, or a combination of these factors. In this connection, our results in Table 3 revealed significant decreases of N, P and K contents and a slight increase of Ca^{+2} and Mg^{+2} as well as significant high accumulation of Na^{+} in non-mycorrhizal plants with increasing seawater contents. These results are in agreement with the previous work of Khan *et al.* (2000) who found that nutrient deficiencies can occur in plants when high concentration of Na^{+} reduce the amounts of available K^{+}, Mg^{2+} and Ca^{2+} or Na^{+} displaces membrane-bound Ca^{2+}. They also suggest that Na^{+} may have a direct toxic effect, by interfering with the function of K^{+} as a cofactor in various reactions.

In the present investigation, it is noticeahle that salinity causes a nutrient unbalance in mungbean plants, which show higher concentrations of the macro-elements N, P, K, Ca and Mg in mycorrhized plants under the saline conditions. Moreover, the results also indicate that mycorrhizal mungbean showed lower Na/N, Na/P, Na/k, Na/Ca and Na/Mg, ratios than that of non-mycorrhized one. These results and tolerance index of salinity results emphasize that AM fungi increase salinity tolerance of the mungbean plants in the present study (Table 3). Similar observations have been made from the previous workers (Jarstfer *et al.*, 1998; Al-Karaki, 2000; Yano-Melo *et al.*, 2002; Roa and Tak, 2002). Na^{+} content of mycorrhizal plants was closely similar to that of non-mycorrhizal plants under non-salinized control conditions. Based on these data and on existing literature, it is conceivable to suggest that mycorrhizal plants may be better equipped to withstand the toxic effects of salt stress.

Table 3. Effect of different dilutions of seawater on % of mineral contents of mycorrhizal and non-mycorrhizal mungbean plants with and without foliar application of kinetin

	N	P	K	Ca	Mg	Ng
Control						
Non-mycorrhizal plant	1.75	0.15	1.21	0.81	0.63	0.47
Mycorrhizal plant	1.94	0.19	1.39	0.98	0.79	0.42
Non-mycorrhizal plant + kinetin	1.75	0.16	1.24	0.83	0.75	0.44
Mycorrhizal plant + kinetin	2.13	0.19	2.43	1.56	1.1	0.41
10% seawater						
Non-mycorrhizal plant	1.78	0.16	1.86	0.83	0.65	0.68
Mycorrhizal plant	2.42	0.28	2.29	1.39	1.12	0.44
Non-mycorrhizal plant + kinetin	1.82	0.13	1.52	0.83	0.81	0.62
Mycorrhizal plant + kinetin	2.63	0.30	2.95	2.1	1.87	0.45
20% seawater						
Non-mycorrhizal plant	1.26	0.09	1.15	1.3	0.75	0.98
Mycorrhizal plant	2.98	0.41	2.29	1.96	1.41	0.47
Non-mycorrhizal plant + kinetin	1.8	0.09	1.2	1.28	0.82	0.83
Mycorrhizal plant + kinetin	3.6	0.41	3.1	2.87	2.12	0.47
30% seawater						
Non-mycorrhizal plant	0.98	0.09	1.62	1.35	0.90	1.36
Mycorrhizal plant	2.81	0.33	2.29	2.88	1.61	0.47
Non-mycorrhizal plant + kinetin	1.87	0.08	1.1	1.35	0.93	1.1
Mycorrhizal plant + kinetin	3.12	0.33	2.99	3.24	2.53	45
L. S. D. $p \leq 0.01$	1.04	0.001	0.71	1.06	0.73	0.01

Increasing seawater concentration had a negative effect on protein content of plants (Table 4). In mycorrhizal plants, protein content was decreased with increasing salinity level of seawater but still exhibited higher values than those of control plants. On the other hand, increasing seawater concentration had a negative effect on values of nitrogen and phosphorus-use efficiencies in non-mycorrhizal plants, while these values were unaffected in mycorrhizal plants. These results are in agreement with those of Ruiz-Lozano and Azcon (2000). Moreover, nitrogenase activity was reduced in non-mycorrhizal plants with higher seawater levels and stimulated in mycorrhizal plants irrespective of absence or presence of kinetin. These results are in accordance with those of Rao and Tak (2002). On the other hand, acid and alkaline phosphatase activities increased in mycorrhizal plants than non-mycorrhizal one under salinity conditions. However, raising dilution of seawater in irrigated water lowered acid and alkaline phosphatase activities. These result in harmony with those of Rao and Tak, 2002 and Abdel-Fattah *et al.*, 2002.

The frequency of mycorrhizal root segments (F%), intensity of mycorrhizal colonization in root tissue (M%) and the rate of arbuscular formation in root segments (A%) increased with higher seawater concentrations (Table 5). The highest stimulation rate of F, M, & A were measured at level of 10% of seawater. Evidence from our results in Table 5 indicates that mycorrhizal dependencies for plant dry mass increased by raising seawater during irrigation. The increase varied from 300% to 597% at a level of 30% seawater in absence of kinetin, while in presence of kinetin the increase from 319% to 680% and 777% at a level of 20 and 30% seawater, respectively. Based on these data it is condeivable to conclude that mungbean plant tolerance and its growth depend on mycorrhizal symbiosis when irrigated with water mixed with various levels of seawater. Abd-El-Fattah and Rabie (1995) obtained similar observation.

Table 4. Effect of different dilutions of seawater on % of protein content, nitrogen and phosphorus-use efficiencies, nitrogenase and phosphatase enzymes of mycorrhizal and non-mycorrhizal mungbean plants with and without foliar application of kinetin

	Protien content	N-use effeciency	P-use effeciency	N_2-ase activity in moles C_2H_4/hr/g dry root	Phosphatase activities u/*ml*/min	
					Total	a
Control						
Non-mycorrhizal plant	11.25	41.4	42.6	75.1	325	1220
Mycorrhizal plant	13.4	72.9	84.6	82.7	520	1635
Non-mycorrhizal plant + kinetin	11.38	43.7	42.9	75.0	320	1110
Mycorrhizal plant + kinetin	13.68	73.1	84.3	82.4	515	1610
10% seawater						
Non-mycorrhizal plant	12.4	32.9	35.0	77.9	300	1192
Mycorrhizal plant	21.13	73.6	85.0	116.5	610	1640
Non-mycorrhizal plant + kinetin	13.21	33.0	31.9	80.4	310	1205
Mycorrhizal plant + kinetin	22.63	72.0	82.3	117.9	627	1605
20% seawater						
Non-mycorrhizal plant	9.6	26.4	24.5	64.7	300	1068
Mycorrhizal plant	19.37	69.7	86.1	108.3	570	1585
Non-mycorrhizal plant + kinetin	10.2	27.8	24.7	74.6	302	1042
Mycorrhizal plant + kinetin	18.3	74.7	83.5	107.8	570	1498
30% seawater						
Non-mycorrhizal plant	7.42	25.1	23.0	59.3	290	923
Mycorrhizal plant	16.5	71.8	80.3	108.0	556	1421
Non-mycorrhizal plant + kinetin	9.6	26.4	22.3	63.9	267	936
Mycorrhizal plant + kinetin	16.6	72.4	81.2	105.2	549	1490
L. S. D. p≤0.01	4.31	10.65	13.51	8.3	15.6	10.92

Table 5. Effect of different dilutions of seawater on percentage of mycorrhizal levels (as indicated by non-vital staining techniques) in mungbean plants inoculated with *Glomus clarum* with and without foliar application of kinetin

	F %	M %	A %	M. D.
Mycorrhizal plant				
0.0 seawater (control)	68	21	11	300%
10% seawater	78	36	19.2	497%
20% seawater	74	24	16	541%
30% seawater	71	23	17.1	597%
Mycorrhizal plant + kinetin				
0.0 seawater (control)	68	19	11.3	319%
10% seawater	81	40	22	607%
20% seawater	75	26	17.4	780%
30% seawater	73	21	17.2	777%
L. S. D. p≤0.01	3.36		1.2	21.8

Salinity is one of the major limitations on crop productivity and quality in the world. Katerji *et al.* (1998) and Hoorn *et al.* (2002), proved that the negative effects of the salinity are reducing the growth rate, biomass reduction, shorter stature, smaller leaves, osmotic effects, nutritional deficiency as well as mineral disorders. Our results confirmed the highly significant effect of salinity on dry weight, transportation rate, K^+ and Na^+ accumulations as well as acid and alkaline phosphatase activity (Table 6). Moreover, the significant effect of salinity on plant height, leaf conductivity, Ca^+ and protein contents was proved in this investgation.

Combined effects of kinetin and salinity had highly significant positive effects on leaf conductivity and transpiration rate of mungbean. This finding agrees with previous results of Nemat-Alla (2002); they found that kinetin alleviates the salinity stress symptoms *V. sinensis* and *Z. mays*.

From careful examination of Table 6 it was noticeable that, the combined action of mycorrhizal symbiosis with kinetin and with salinity as well as altogether are closely related to the effects of mycorrhizal symbiosis only on growth of mungbean plants in this investigation. Based on these data and existing literature, it is of conceivable that, mycorrhizal symbiosis is a prime factor for growth and survival of myngbean plants under salinity stress in the present investigation. Similar observations have been made before (Cuartero and Munoz, 1999; Auge, 2001; Davies *et al.*, 2002). They reported that vesicular-arbuscular mycorrhizal fungi seemed to increase growth and salt tolerance in some plants such as onions, legumes, wheat, tomato and banana.

The present study, therefore, revealed that by whatever mechanism (increasing leaf conductivity and transpiration, as well as enhanced mineral nutrition by improving nutrient availability, competitive uptake, transport or partitioning within plant), the AM fungus, *G. clarum*, increased most efficiently salinity stress tolerance in mungbean plants. It colonized the roots most effectively and the mungbean plants showed 597% and 780% dependency on

Table 6. ANOVA considering growth parameters of mungbean plants, VAM symbiosis, kinetin treatment and salinity after the experimental period

Variables	VAM (M)	Kinetin (K)	Salinity (S)	MXK	MXS	S x K	SxMxK
Dry weight	**	ns	**	*	**	ns	*
Plant height	*	ns	*	ns	**	ns	ns
Leaf conductivity	*	**	*	**	*	*	*
Transpiration rate	**	*	**	**	*	*	**
Chlorophyll content	*	ns	ns	*	ns	ns	*
Sugar content	ns	ns	ns	ns	*	ns	*
Nitrogen	*	ns	ns	*	*	ns	**
Phosphorus	**	ns	ns	*	**	ns	**
Potassium	*	*	**	*	**	ns	*
Calcium	*	ns	*	*	**	ns	*
Magnesium	*	ns	ns	*	*	ns	*
Sodium	*	ns	**	*	**	ns	**
Protein	**	ns	*	*	*	ns	*
Nitrogenase	**	ns	ns	*	*	ns	*
Alkaline phosphate	**	ns	ns	**	*	ns	*
Acid phosphate	**	ns	ns	**	*	ns	*

VAM vesicular arbuscular mycorrhizal fungi.
**Highly significant $p \leq 0.01$.
*Significant $p \leq 0.05$.
ns Non-significant.

this endophyte in absence and presence of kinetin, respectively, for biomass production under a level of 30% of seawater in irrigated water. Thus, applications of this AM endophyte (*G. clarum*) with kinetin could be of importance in using seawater in certain dilutions for irrigation in agricultural purposes.

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