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ORIGINAL ARTICLE

AMF and *Bacillus megaterium* Neutralize the Harmful Effects of Salt Stress On Bean Plants

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Abstract

Two pot experiments were directed under open field conditions where green bean (*Phaseolus vulgaris* L.) plants cv. Valentino were irrigated with four levels of salinity (1000, 2000, 3000 and 4000 ppm) combined with two anti-salinity agents (Arbuscular Mycorrhizal fungi [AMF] *Glomus irradicans* 10% w/w, *Bacillus megaterium* [10 ml/pot] and non-inoculated plants as control) to counteract the negative effect of salt stress, improve the growth, yield, enzymes activity and chemical composition of green bean plants during 2017and 2018 growing seasons. All salinity amelioration treatments (AMF and *Bacillus megaterium*) significantly improved vegetative growth, shoots biomass (total fresh and dry weight per plant), chlorophyll and antioxidant enzymatic activity at all verified salinity levels compared with non-inoculated plants (control) which showed severe growth retardation especially under the higher salt concentration (4000 ppm). The lowest values of membrane permeability and maximum leaf relative water content were significantly obtained with AMF and *B. megaterium*. Plants irrigated with lower concentrated saline water (1000 ppm) significantly accumulated lower Na and Cl and higher K than plants irrigated with higher concentrated salinity irrigation water (4000 ppm). The anti-salinity application increased green bean pod yield under all salinity stress levels particularly with AMF followed by *B. megaterium* compared with non-inoculated plants.

Keywords Salt stress · Arbuscular mycorrhizal fungi · *Bacillus megaterium* · Antioxidant enzymes · Membrane permeability

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AMF und *Bacillus megaterium* neutralisieren die schädlichen Auswirkungen von Salzstress auf Bohnenpflanzen

Zusammenfassung

In zwei Topfexperimenten unter Freilandbedingungen wurden grüne Bohnen (*Phaseolus vulgaris* L.) cv. Valentino mit salzhaltigem Wasser bewässert (vier Salinitätsstufen: 1000, 2000, 3000 und 4000 ppm), kombiniert mit zwei unterschiedlichen Behandlungsmethoden (arbuskuläre Mykorrhizapilze [AMF] *Glomus besticansans* 10 % w/w, *Bacillus megaterium* [10 ml/Topf], nicht beimpfte Pflanzen als Kontrolle). Ziel war es, der negativen Auswirkung von Salzstress entgegenzuwirken sowie das Wachstum, den Ertrag, die Enzymaktivität und die chemische Zusammensetzung von grünen Bohnenpflanzen in den Vegetationsperioden 2017 und 2018 zu verbessern. Beide Behandlungen verbesserten signifikant bei allen Salzgehalten das vegetative Wachstum, die Biomasse der Triebe (Gesamt-Frisch- und -Trockengewicht pro Pflanze), die enzymatische Chlorophyll- und antioxidative Aktivität im Vergleich zu nicht beimpften Pflanzen. Die Kontrollpflanzen zeigten eine starke Wachstumsverzögerung, insbesondere bei der höheren Salzkonzentration (4000 ppm). Die geringste Membranpermeabilität und der maximale relative Wassergehalt der Blätter wurden durch die Behandlung mit AMF und *B. megaterium* erreicht. Pflanzen, die mit niedriger konzentriertem Salzwasser (1000 ppm) bewässert wurden, akkumulierten signifikant weniger Na und Cl und mehr K als Pflanzen, die mit höher konzentriertem Salzwasser (4000 ppm) bewässert wurden. Die Behandlungen erhöhten die Ausbeute der grünen Bohnenschoten, insbesondere durch AMF, gefolgt von *B. megaterium,* verglichen mit nicht beimpften Pflanzen.

Schlüsselwörter Salzstress · Arbuskuläre Mykorrhizapilze · *Bacillus megaterium* · Antioxidative Enzyme · Membranpermeabilität

Introduction

At recent days Egypt is severely suffering from a lack of fresh irrigation water particularly after the building of the Grand Ethiopian Renaissance Dam (Hegazi et al. 2015). Such situation pushed the farmers to utilize semi-saline underground water or reuse of poor-quality drainage water (Hegazi et al. 2017). The effects of salinity on plants include ion toxicity, osmotic stress, impaired growth, mineral deficiencies, photosynthetic imbalance, and combinations of these effects (Galmés et al. 2011). A lot of studies worked to reduce salinity negative effects on the growth and production of various vegetable crops (Ashraf and Foolad 2007; Gupta and Huang 2014; Hegazi et al. 2015, 2017). Green bean is a low-cost and nutritious food from the fabaceae family (Farooq et al. 2017) and also it is a major protein source for Egyptian families (Abdel-Mawgoud 2006). Arbuscular mycorrhizal fungi (AMF) show a positive effect on abiotic stress (Augé 2001). The alleviation of salt stress in plants by AMF is mediated by growth hormones (Barker and Tagu 2000). Various studies revealed that AMF improve plant growth and yield under salt stress conditions (Al-Karaki et al. 2001). Plant growth-promoting rhizobacteria (PGPR) improve plant growth and yield (Noel et al. 1996). It can also protect plants from the deleterious effects of environmental stresses, including flooding (Grichko and Glick 2001), drought (Mayak et al. 2004a), salt (Mayak et al. 2004b). The *B. megaterium* strain grows in both the rhizosphere and the roots. This bacterial strain promotes clover growth under drought conditions. Also, this *B. megaterium* strain increases its own levels of proline and indole acetic acid (an auxin) when grown in vitro under osmotic stress conditions without affecting its own growth (Marulanda et al. 2009). Thus, it is approved that a specific *B. megaterium* strain is capable to amend the plant response to numerous abiotic stresses in different plant species. For this reasons, this study is an attempt to discover an applied technique to alleviate the harmful effect of salinity on growth, yield and the quality of green bean plants using *B. megaterium* (PGPR) and AMF.

Materials and Methods

Location and Growth Conditions

Two pot experiments were proposed out at the Experimental Station Farm, Faculty of Agriculture, Ain Shams University, Cairo, Egypt (30° 4′ 37.28N, 31° 17′ 6.06″ E), during the two seasons (2017 and 2018). Green bean plants (*Phaseolus vulgaris* L.) cv. Valentino (a bush bean of the fine type group) seeds were seeded on the 1st of September in both seasons in black polyethylene bags weighted 15 kg of washed sand (soil physical properties were 89.4% sand, 6.9% silt, and 3.7% clay with a pH of 7.8, EC of 1.68 dS/m). The experimental location has a prevalent arid weather, with cool winters and a warm dry summer.

Plant Material and Treatments

The present trial incorporated four NaCl treatments (1000, 2000, 3000 and 4000 ppm) with two anti-salinity applications (*B. megaterium*, AMF and a control without treatment). Normal farming practices were followed for green bean production, and disease and pest control were followed referring to the recommendation of the Egyptian Ministry of Agriculture. Seeds were seeded in four pits per pot. Ten days after seed germination, seedlings were thinned to five plants per pot/replicate, and after 21 days, NaCl treatments were started. Each experimental plot involved 15 pots arranged in 3 rows, with 5 pots in each row. Two litres of Hoagland's nutrient solution (full strength) was applied weekly to each pot. The experiment was organized in a completely randomized design with three replicates.

Inoculation with *Bacillus megaterium* and AMF

The AMF treatments used in this study were a non-mycorrhizal inoculum as a control and a mycorrhizal inoculum (a mixture of stock cultures of *Glomus* spp. isolate) created by adding 100 g of a mycorrhizal inoculum per bag at sowing time. Inocula consisted of spores, extra-radical mycelium, and mycorrhizal roots, which were achieved from the Department of Agricultural Microbiology, College of Agriculture, Ain Shams University. The active strain of *B. megaterium* (108 CFU/ml) was added at 10 ml/pot. The soil inoculation with *B. megaterium* and AMF was repeated three times at 15-day intervals.

Sampling and Data Recording

Vegetative Growth Parameters

A random sample of three plants/replicate was collected 45 days after sowing to evaluate vegetative growth elements, namely plant length, shoot fresh and dry weight, and root fresh and dry weight. The plants were weighed to record the plant fresh weight and were placed in an oven at 70 ºC until a constant weight was achieved, which was recorded as the plant dry weight.

Determination of Photosynthetic Pigments

At 45 days after sowing, a portable chlorophyll meter (SPAD-502; Konica Minolta Sensing, Inc., Japan) was used to determine the leaf greenness of the plants. For each plant, measurements were taken at four locations on each leaf (two on each side of the midrib on all fully expanded leaves) and then averaged (Khan et al. 2003).

Determination of Membrane Permeability (MP)

To measure membrane permeability (MP), 10 leaf discs (10 mm in diameter) from the young, fully expanded leaves from two plants per replicate were placed in 50-ml glass vials and washed with distilled water to remove electrolytes released during leaf disc excision. Electrolyte leakage was calculated as a percentage of EC1/EC2 (Shi et al. 2006).

Determination of Leaf Relative Water Content (LRWC)

Leaf samples were taken from two plants per replicate (the sixth leaf from the top) to determine fresh weight (FW), dry weight (DW) and turgid weight (TW). FW, TW, and DW values were used to calculate LRWC using the following equation (Kaya et al. 2003): $\text{LRWC}\ (\%) = [(\text{FW} - \text{DW})/(\text{TW} - \text{DW})] \times 100$.

Biochemical Composition

Determination of Catalase Activity (CAT) CAT activity was measured following a published method (Montavon et al. 2007). Fresh tissues were homogenized in 50 mmol sodium phosphate buffer (pH 7.0, 1/10, w/v) including 150 mmol NaCl and 0.5 mmol EDTA. One unit of CAT activity was expressed as the amount of enzyme needed to reduce 1 µmol of H_2O_2 per min CAT activity and was expressed as unit $min^{-1} mg^{-1}$ protein.

Determination of Superoxide Dismutase Activity (SOD) The activity of superoxide dismutase (EC1.15.1.1) was analysed using the method of Beauchamp and Fridovich (1971) by measuring the ability of the enzyme to inhibit the photochemical reduction of nitro blue tetrazolium (NBT). A reaction mixture (3 ml) containing 40 mM phosphate buffer (pH 7.8), 13 mM methionine, 75 µM NBT, 2 µM riboflavin, 0.1 mM EDTA and 100 µl of crude enzyme extract was shaken and placed 30 cm below a 15 W fluorescent lamp as a light source. The absorbance was recorded at 650 nm. One unit of SOD activity is the amount of protein required to inhibit 50% of the initial reduction of NBT under light. The activity of SOD was expressed as unit $min^{-1} mg^{-1}$ protein.

Determination of Peroxidase Activity (POD) The activity of peroxidase (EC1.11.1.7) was assayed by the method of Hammmerschmidt et al. (1982). The reaction mixture (2.9 ml) consisted of 0.25% (v/v) guaiacol in 10 mM sodium phosphate buffer (pH 6 containing 10 mM H_2O_2). One hundred microlitres of crude enzyme extract was added to initiate the reaction, which was measured spectrophotometrically (CT200 spectrophotometer) at 470 nm per min. One international unit (IU) of enzyme activity was expressed

as $\Delta OD = 0.01$, and POX activity was expressed as unit $min^{-1} mg^{-1}$ protein.

Determination of Na, K, Cl and P Concentrations The extraction and determination of Na and K concentrations were conducted according to (Xu et al. 2006). Na and K concentrations were measured using an atomic absorption spectrophotometer (Varian spectra AA 220, Varian, Palo Alto, CA, USA). Chloride was measured using an ion chromatography analyser (Model 926, Sherwood Scientific Ltd., Cambridge, UK). Total phosphorus was determined using a spectrophotometer according to the method of Jackson (1973).

Pod Yield Parameters

Green pods were collected 55–60 days after sowing, and the total number of pods per plant, pod length and pod yield (kg/m^2) were calculated according to AOAC (1990).

Experimental Design and Statistical Analysis

Data for all experiments were investigated by analysis of variance (ANOVA) using the general linear model procedure of CoState. The significance of differences between means was tested using an "F" test, and the LSD ($p = 0.05$) was calculated (Snedecor and Cochran 1967).

Results and Discussion

Vegetative Growth

The results in Table 1 show that plants grown in 4000 ppm were significantly harmfully affected, as validated by the vegetative growth traits of green bean plants (e.g., plant length, shoot fresh and dry weight, and root fresh and dry weight). The best growth significantly occurred with 1000 ppm. Salinity amelioration treatments (Mycorrhiza and *B. megaterium*) compared to uninoculated plants (control), upgraded all mentioned plant vegetative growth characteristics affected by salt stress. *B. megaterium* was significantly best in mitigating the salt stress effects at low level (1000 ppm) but it was moderately neutralising the destructive effects of high-salt stress (4000 ppm) for all vegetative growth parameters, followed by Mycorrhiza application. Similar trend was detected in the two seasons. The obtained results confirmed previous findings of Ferri et al. (2000) who observed that salt stress reduced the growth of common bean (*P. vulgaris* L). However, AMF was more effective in improving growth of Chickpea and Zucchini plants (Garg and Bhandari 2016; and Colla et al. 2008). AMF gave a progressive general impact on plant

Table 1 Effect of anti-salinity treatments on vegetative growth parameters of green bean plants irrigated with four levels of salinity

Treatment	Salinity concentration (ppm)	Plant length (cm)	Shoot FW (g/plant)	Shoot DW (g/plant)	Root FW (g/plant)	Root DW (g/plant)	Plant length (cm)	Shoot FW (g/plant)	Shoot DW (g/plant)	Root FW (g/plant)	Root DW (g/plant)
		2017					2018				
Control	1000 ppm(control)	35.50b	5.36b	1.23bc	9.63ab	0.70ab	47.00b	17.44c	1.85b	8.21b	2.45b
	2000 ppm	28.00d	4.76b	0.96c	6.26bc	0.56ab	34.87d	13.52de	1.51c	7.53bc	0.81cde
	3000 ppm	22.55e	3.70b	0.70bc	4.01bc	0.46ab	24.40fg	9.10fg	1.09de	6.102c	0.57de
	4000 ppm	18.00 f	2.07b	0.36 c	2.10bc	0.33b	23.75g	7.26g	0.45d	2.90d	0.39e
Mycorrhiza	1000 ppm(control)	15.00a	9.21a	2.10ab	13.28a	0.77ab	15.50b	20.19b	2.36a	12.26a	4.66a
	2000 ppm	31.50 c	5.12 b	1.20bc	7.88abc	0.60ab	40.00c	15.11cd	1.82b	8.08b	1.31cd
	3000 ppm	23.30e	3.74 b	0.741c	4.41bc	0.50ab	26.55ef	11.64f	1.17d	6.25c	0.59de
	4000 ppm	18.50f	2.10b	0.40c	2.50bc	0.40b	24.70fg	9.05g	0.85e	3.69d	0.41e
Bacillus	1000 ppm(control)	53.55a	5.63a	2.60a	13.59a	0.90a	56.80a	60.76a	2.58a	12.73a	4.76a
	2000 ppm	33.67bc	5.06 a	1.21c	9.17abc	0.65ab	41.00c	15.58cd	1.82b	8.25b	1.51c
	3000 ppm	24.28e	4.60 a	0.80c	4.50bc	0.47ab	27.30de	11.75ef	1.15d	6.55c	0.59de
	4000 ppm	18.00f	3.36ab	0.38 c	2.50bc	0.36b	24.70fg	9.28fg	0.97de	4.14d	0.48e
LSD at 5%		2.56	3.55	0.84	6.32	0.42	4.71	2.68	0.28	1.36	0.71

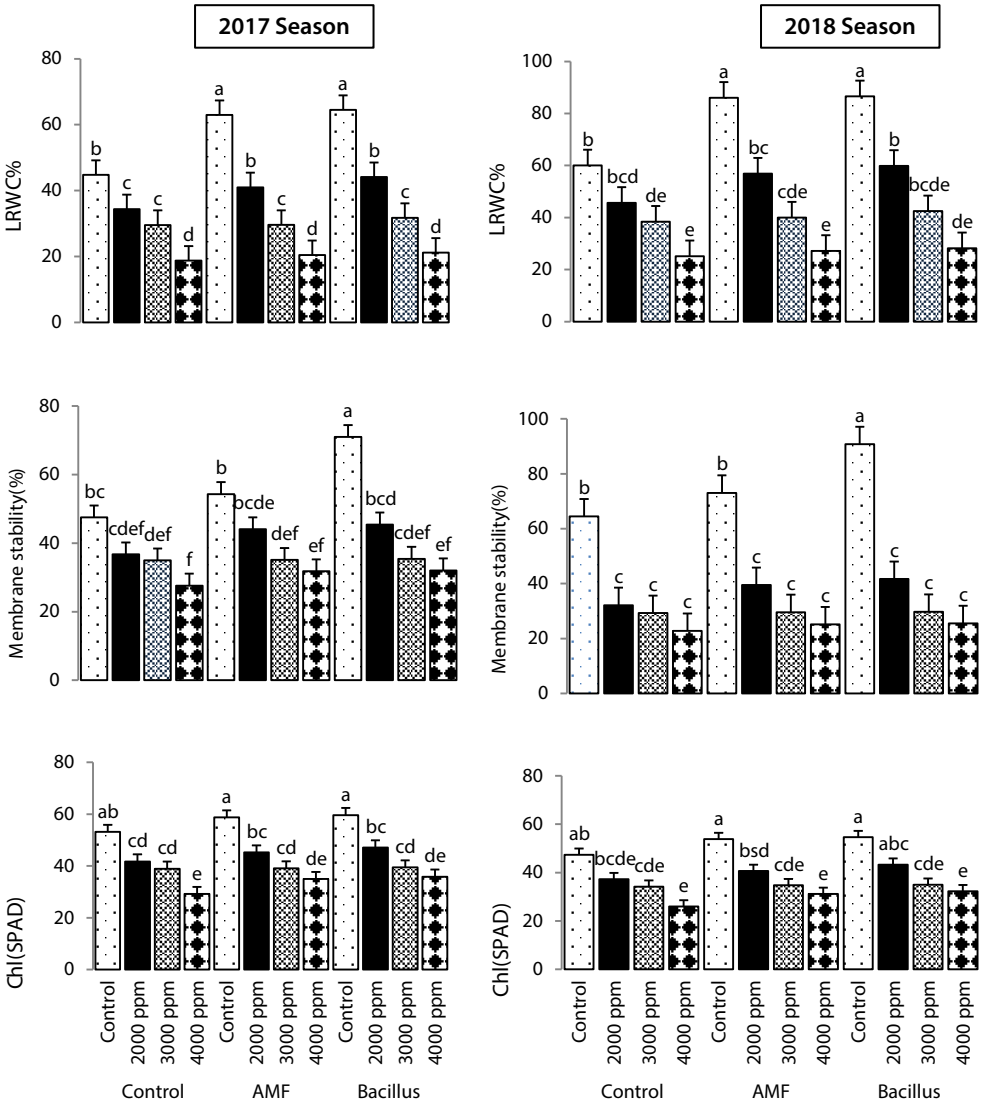

Fig. 1 Effect of different salinity concentrations and anti-salinity treatments on leaf relative water content (LRWC), membrane permeability (MP) and chlorophyll by SPAD in green bean plants (season 2017–2018)

biomass under salt stress conditions in sweet pepper plants (Tian et al. 2004). Other studies conveyed similar growth promotion in various leguminous vegetable crops with AM inoculation. These crops included green beans (Neeraj and Singh 2005; Salim and Abou El-Yazied 2015) and pea (Estaún et al. 1987). This effect could be related to a sufficient supply of nutrients (principally phosphorus), with the support of AMF in the host plant (Marschner 1986; Al-Karaki 2000; and Ghoname et al. 2012). *B. megaterium* retained a better root growth, which is considered a salt tolerance trait (Farooq et al. 2017). The *B. megaterium* strain supposed to produce indole-3-acetic acid (an auxin) (Marulanda et al. 2009), which might lead to better root growth and development under saline conditions (Wang et al. 2009). A lot of studies show that the PGPR led to increase root fresh weight and dry weight and shoot fresh weight and dry weight in okra (Habib et al. 2016) and strawberry (Karlidag et al. 2013). PGPR isolates can lessen salt stress effect in plants and enhance shoot/root length, dry matter production in numerous plants (Egamberdieva and Lugtenberg 2014). The progressive effects of PGPR treatments on growth of plants can be attributed to the release of phytohormones, such as indole-3-acetic acid and cytokinins as well as N_2 fixation, phosphate-solubilizing, and production of antimicrobial materials., amino acids, and enzymes, (Gunes et al. 2015).

Photosynthetic Pigments

Data in Fig. 1 show that chlorophyll concentrations were significantly better with lower salinity level (1000 ppm) and were considerably lower with the high salinity level (4000 ppm) which gave the lowest chlorophyll concentration. Anti-salinity applications positively minimized the harmful effect of salt stress and generated a stimulatory effect on all plant photosynthetic pigment concentrations

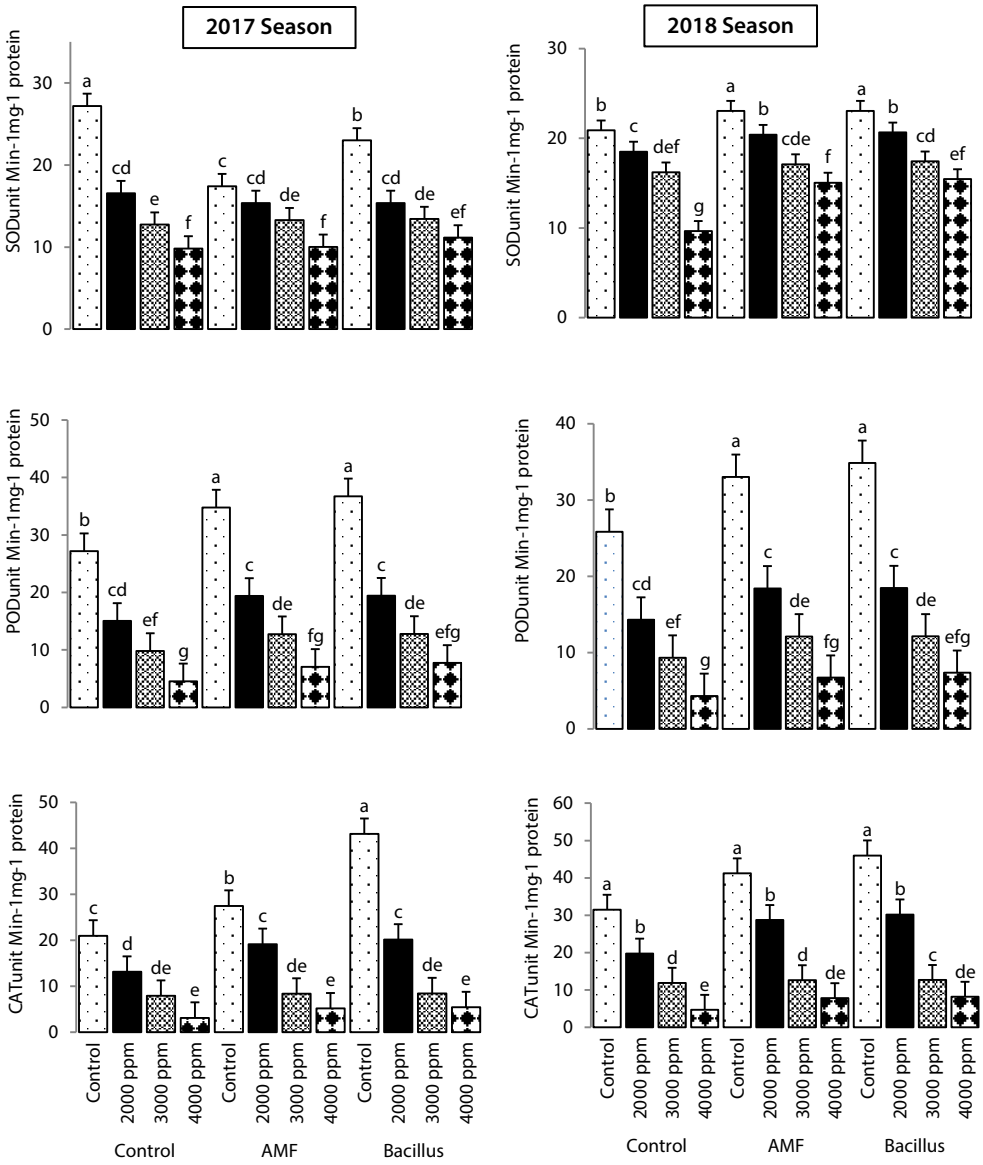

Fig. 2 Effect of different salinity concentrations and anti-salinity treatments on antioxidant enzyme activity of green bean plants (season 2017–2018)

when compared to uninoculated plants. Generally at low salinity level (1000 ppm), *B. megaterium* enhanced chlorophyll content followed by AMF, with no significant differences between them. Higher chlorophyll content in the leaves of mycorrhiza-inoculated plants under salt stress conditions was conveyed by numerous investigators (Giri and Mukerji 2004; Shekoofeh and Sepideh 2011; and Elhindi et al. 2017). The chlorophyll decline under salt stress can be accredited to the antagonistic effects of Na^+ ions on Mg^{2+} absorption. AMF improved the absorption of Mg^{2+} ions, which in turn can intensify chlorophyll synthesis in salt-stressed plants (Giri and Mukerji 2004). The improved photosynthetic pigmentation due to mycorrhizal colonization in plants might be attributed to the inhibition of Na^+ transport, which leads to better function of the photosynthetic mechanism (Borde et al. 2010; García-Garrido and Ocampo 2002). Similar improvement in chlorophyll content were stated after mycorrhizal inoculation in cowpea (Arumugam et al. 2010), fava bean (Ismaiel et al. 2014), snap bean (Salim and Abou El-Yazied 2015), pea (Shinde and Thakur 2015), and lentil (Yaseen et al. 2016). In this respect, Yildirim et al. (2008) reported that PGPR improved the chlorophyll contents of radish leaves under salt stress. PGPR inoculation alleviated the damaging effects of salinity which improved the chlorophyll synthesis, thus enhancing salt tolerance in strawberry plants (Karlidag et al. 2013).

Leaf Relative Water Content (LRWC) and Membrane Permeability (MP)

Results shown in Fig. 2 pointed that high salinity stress level (4000 ppm) deleteriously affected leaf relative water content (LRWC) and membrane permeability (MP), with

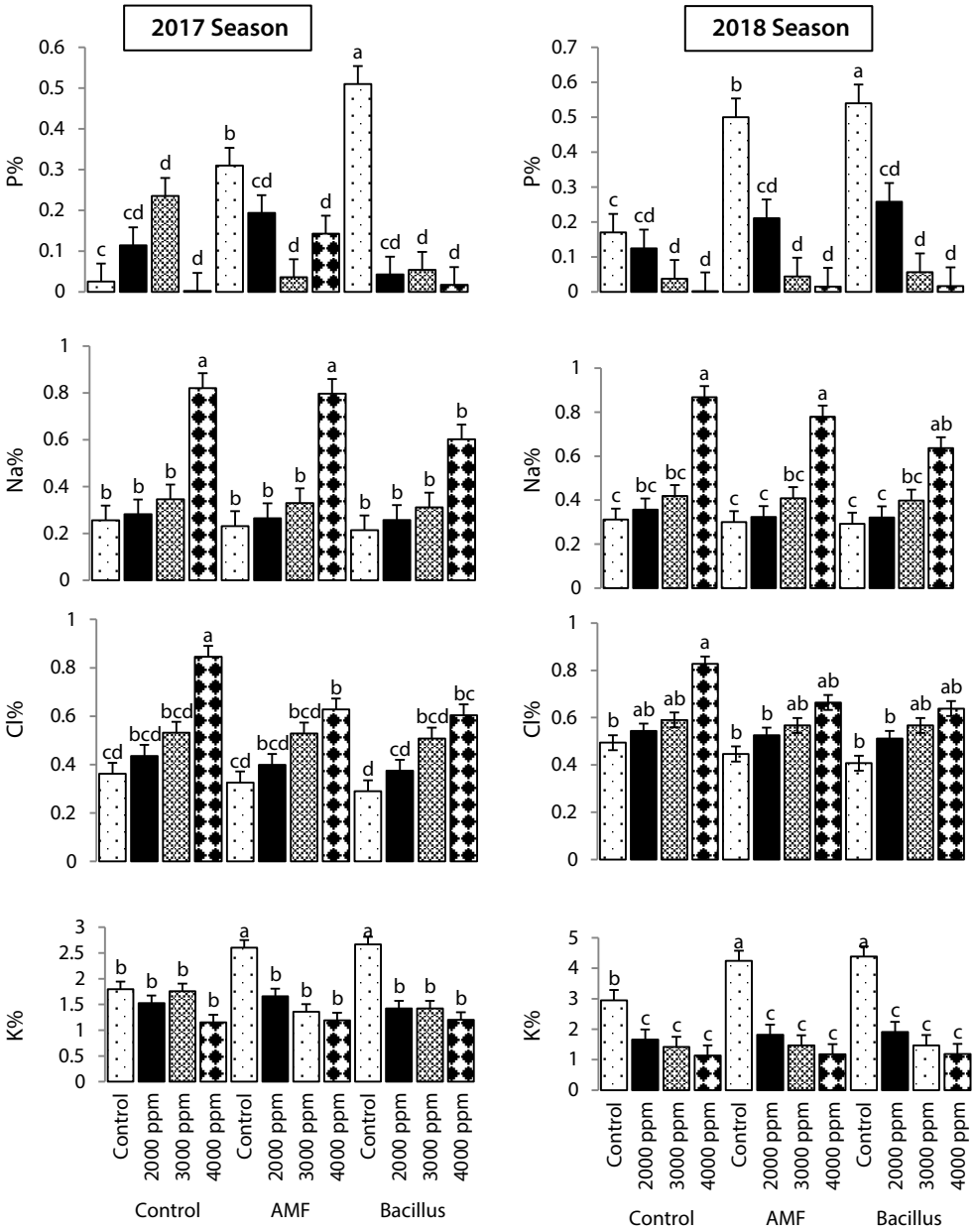

Fig. 3 Effect of different salinity concentrations and anti-salinity treatments on mineral content (P, Na, Cl and K) of green bean plants (season 2017–2018)

significant differences among salinity levels. Salinity amelioration treatments positively improved LRWC and significantly reduced the damaging effect of salinity on MP compared to uninoculated plants, with significant differences between the inoculated plants and uninoculated plants (control). The achieved results confirmed former reports of (Kaya et al. 2009), which revealed that pigeon pea (*Cajanus cajan*) demonstrated higher relative MP when treated with AMF. Moreover, the electrical conductivity of mycorrhizal plants was higher in pigeon pea plant roots (Garg and Manchanda 2008). As well, Colla et al. (2008) conveyed water status enhancement of Zucchini plants colonized by *Glomus intraradices* when exposed to salinity stress. PGRB treatments initiated an increment in LRWC under salt stress (Yildirim et al. 2008). Additional investigation of Yildirim et al. (2008) indicated that PGRB-inoculated plants had better electrolyte leakage than their relevant uninoculated controls.

Antioxidant Enzyme Activity

Antioxidant enzyme activity (CAT, POD and SOD), as demonstrated in Fig. 2, revealed a significant progressive effect with salinity amelioration application. The best results were obtained by *B. megaterium*. The most negative effect of high saline condition (4000 ppm) was recorded in the non-inoculated plants (control) On the other hand, salinity amelioration treatments significantly increased enzyme ac-

Table 2 Effect of anti-salinity treatments on pod yield of green bean plants irrigated with four levels of salinity

Treatment	Salinity concentration (ppm)	Pod weight (g) 2017	Pod length (cm)	Total yield (kg/m^2)	Pod weight (g) 2018	Pod length (cm)	Total yield (kg/m^2)
Control	1000 ppm(control)	3.26ab	9.00ab	3.30abc	5.23ab	15.10a	3.65abc
	2000 ppm	2.40abc	8.17ab	2.70c	2.78bc	10.46bc	2.94c
	3000 ppm	1.50bcd	7.00b	2.29c	1.58bc	8.13bc	2.43c
	4000 ppm	0.00d	5.70b	0.00d	0.40c	5.70d	0.50d
Mycorrhiza (AMF)	1000 ppm(control)	4.60ab	9.50ab	4.22ab	7.70a	16.00a	4.70ab
	2000 ppm	3.10abc	8.67ab	3.11bc	3.50abc	11.55b	3.74bc
	3000 ppm	1.60bcd	7.13b	2.35c	1.70bc	9.90bc	2.56c
	4000 ppm	0.40 cd	6.00b	0.52d	0.40c	7.83 cd	0.51d
Bacillus (PGPR)	1000 ppm(control)	4.90a	12.00a	4.39a	7.70a	15.90a	4.56a
	2000 ppm	3.20abc	9.00a	3.26abc	3.60abc	11.50b	3.40bc
	3000 ppm	1.60bcd	7.28b	2.32c	1.90bc	9.72bc	2.56c
	4000 ppm	0.80 cd	6.15b	0.52d	0.80bc	7.60 cd	0.51d
LSD at 5%		2.57	3.41	1.10	3.91	2.65	1.05

tivity at the 1000 ppm salinity levels. Previous data for other investigations showed that both salt and drought stress could induce oxidative stress, as indicated by the increased level of lipid peroxidation (Hossain et al. 2015). The activities of SOD and POD were increased by salt and drought stress, but activity of CAT was declined (Pan et al. 2006). To resist the stressful environment, plants provides numerous antioxidant enzymes to preserve them from the damaging effects of ROS. So, antioxidative enzymes have a key character as a resistance mechanism in several plant species (Koyro et al. 2012). Improved antioxidant enzymes involved with AMF plants have been verified by many researchers. Mycorrhizal plants had a higher accumulation of antioxidative enzymes and thus improve general plant growth under stress (Miller et al. 2010; Scheibe and Beck 2011). Similar results were conveyed by Han and Lee (2005) for lettuce. Heidari and Golpayegani (2012) indicated that inoculation with *Bacillus lentus* and *Azospirillum rasilens* significantly increased Ascorbate peroxidase and Glutathione peroxidase activity of basil leaves grown under drought stress.

Mineral Content

The data in Fig. 3 show that highest Na^+ and Cl^- content was obtained with the highest salinity stress level (4000 ppm) while, the maximum K^+ and P levels were achieved with the low salinity stress level (1000 ppm). The salinity amelioration treatments led to a significant increases in K^+ and P, level with a premier result obtained with *B. megaterium* treatment. There were a significant differences between salinity amelioration treatments and the control (uninoculated) on Na^+ and Cl^- content. The achieved results confirmed those of a previous study (Juniper and Abbott 1993), who revealed that salinity initiates an imbalances in the K^+/Na^+ ratio, and harmfully affecting plant growth. As mycorrhizal plants retain higher Na^+/K^+ (higher K^+ uptake in shoots), such plants by a dilution effect has the ability to alleviate salt stress. Under saline conditions, higher levels of sodium (Na^+) not only interrupt the uptake of other nutrients but also lead to specific ion toxicity (Ashraf 1994). A high K^+/Na^+ ratio is essential for salinity tolerance and preservation of plant osmotic potential (Hamdia et al. 2004). The promoting effect of mycorrhizal inoculation on snap bean growth is possibly due to the effects of AMF on enhancing soil structure (Miller and Jastrow 2000). Also, salt stressed zucchini plants when colonized by *Glomus intraradices* had a better nutrient content (Colla et al. 2008). Growth improvement of plants inoculated with AMF has been partially correlated to that mycorrhiza enriched nutrient accomplishment, particularly P nutrition (Sharifi et al. 2007). Furthermore, PGPR can improve the uptake of mineral element by motivating root formation and growth (Yildirim et al. 2008).

Pod Yield Parameters

The collected data in Table 2 show that yield (number of pods per plant, total pods yield per plant and average pods weight) was positively superior under low salinity level (1000 ppm). Meanwhile, low salinity level gave the maximum yield compared to the highest salinity stress level (4000 ppm). However, anti-salinity applications positively enhanced the pod yield of green bean plants under saline stress conditions. Hence, the highest pod yields of bean plants were significantly observed with B. megaterium, followed by AMF, compared with control (uninoculated) plants at all salinity stress levels. Similar findings of Khan et al. (2016) showed that salinity stress delayed flow-

ering and lessened flower numbers and pod set in green bean plants. Youssef et al. (2017) showed that AMF inoculation significantly increased pod number and green pods yield of snap bean. Also, Colla et al. (2008) conveyed an improvement in fruit quality of Glomus intraradices colonized zucchini plants grown under salinity stress. Al-Karaki (2000) indicated a higher fresh fruit yield, fruit weight and fruit number in a mycorrhizal inoculated.

Conclusion

This investigation affirmed the significance of both AMF and *Bacillus megaterium* application on ameliorating the negative effect of salinity on growth and pod yield of green beans. Chlorophyll and antioxidant enzymatic activity at all verified salinity levels were markedly enhanced by AMF and *Bacillus megaterium* treatments compared with non-inoculated plants (control) which showed severe growth retardation especially under the higher salt concentration.

Conflict of interest N.A. Abdel Motaleb, S.A. Abd Elhady and A.A. Ghoname declare that they have no competing interests.

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