

Increasing Reproductive Stage Tolerance to Salinity Stress in Soybean

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ABSTRACT: Salinity seriously threatens crop production in arid and semi-arid regions. So, the study was carried out to assess whether exogenously applied glycine betaine or proline with combining compost has any role in reducing the adverse effects of saline stress and might enhance the salt tolerance in soybean. Salinity stress treatment (Control, 15mM NaCl), compost (Control, 25 ton ha⁻¹) and exogenous proline and glycine betaine at the rate of (Control, 25 mM) arranging in spilt-spilt plot. Results indicated that salinity stress induced reduction in yield traits and quality of seeds compared with those of the unstressed plants. These reductions were mainly attributed to the short duration of protein and oil accumulation and grain yield per plant under saline conditions. Special attention was paid to the application proline and glycine betaine alone or amended with compost showed increased seed yield and quality under all salinities as compare to control. Consequently, application of organic fertilizer to soil and exogenous proline or glycine betaine are a good strategy to improve salinity tolerance in soybean cultivation.

Keywords: Compost; Glycine bataine; Proline; quality; Salinity tolerance; Soybean; Yield.

INTRODUCTION

Soybean seed is a major source of high-quality protein and oil for human consumption (Katerji et al., 2001). In soybean, salinity stress inhibits seed germination and seedling growth, reduces nodulation, and decreases biomass accumulation and yield (Essa, 2002). Soybean is classified as a moderately salt-tolerant crop and the final yield of soybean will be reduced when soil salinity exceeds 5 dS m⁻¹ (Ashraf, 1994). Yield of soybean is the result of genetic potential interacting with environment. Minimizing environmental stress will optimize seed yield (Mc Williams et al., 2004).

Salinity is a worldwide problem, affecting about 95 million hectares worldwide. Salinity is one of the major factor which causes inhibitory effects on plant growth. The reduction in growth under saline conditions is more severe in arid and semi-arid regions (Rengasamy, 2006) due to adverse effects on metabolic and physiological processes (Krishnamurthy et al., 2007). Salinity causes a number of changes in plant metabolism, through ion toxicity and osmotic stress (Mittler, 2002).

To reciprocate environmental stress, plants store multiple groups of compatible solutes such as sugars, free amino acids like glycinebetaine, proline and polyols (Hoque et al., 2007). Betaine and proline are the most popular compatible solutes that contribute to osmotic adjustment (Ashraf and Foolad, 2007), and stabilization and protection of proteins, enzymes and membranes from the damaging effects of salt stress (Okuma et al., 2000 and Ashraf and Foolad, 2007). During osmotic adjustment, many plants accumulate proline in response to salt stress widely believed to function as a protector against salt damage (Wang et al., 2007). Glycine betaine is one of several such compatible solutes that has an osmoprotective function and is known to improve salt stress tolerance in most crop plants (Hossain and Fujita, 2010).

Spreading of organic matter on the soil surface is a known practice. It is a good technique to reduce capillary rise of salts and maintain soil moisture in arid regions. The productivity of irrigated crops with saline water or crops grown under saline stress can be enhanced by using compost as an amendment (Lakhdar et al., 2008). Organic

matter application to saline is an useful remediation method, in terms of the physical, chemical and biological properties of the soil (Wong et al., 2009). When compost is applied to the soil, it can support plant growth and enhance plant yield as well as improve the physical, chemical and biological properties of soils (Convertini et al., 2004). Pang et al (2010) and Saeed and Ahmad (2009) found that using of mulch helped to improve plant growth by lowering down the salinity of rhizosphere. This requires a major focus on improving productivity and sustainability of rice-based farming systems. The aim of present study is to evaluate the effectiveness of proline, glycine betaine alone and with compost as supplement for preventing loss seed yield and maintaining oil and protein content in soybean plants grown under saline conditions.

MATERIAL AND METHODS

Plant Material and Culture Conditions

The experiment was conducted at the Graduate School of Biosphere Science Laboratory of Hiroshima University, Japan in 2011. The seeds of soybean, cultivar (Giza 111) were obtained from ARC, Egypt. The seeds were sown into wood made basin (length 10 meter, width 50 cm, height 50 cm and soil depth 35 cm). The basin was filled with a mixture of soil, perlite and peat moss at the ratio of 4:2:1 (v/v/v). Each basin was fertilized at a rate of 40 kg N ha⁻¹, 120 kg P₂O₅ ha⁻¹ and 100 kg K₂O ha⁻¹ using fertilizer mixture and calcium carbonate (300 kg ha⁻¹). The experiment was designed as a completely randomized block design with arranged a split split plot arrangement with four replication. The trial materials could be summarized as follows: (i) Salinity treatment (Control and 15 mM NaCl), were applied in beginning bloom (R1). (ii) Compost treatments (Control and 24 ton ha⁻¹) were manufactured using wood poop, chicken poop and palm. Chemical analysis of compost was N: 9.1%, phosphorous: 9.0%, potassium: 5.0% and C/N: 2.4. (iii) Exogenous proline and glycine betaine treatments (Control, 25mM prolin or glycine betain) were applied in beginning bloom (R1).

Plant Sampling and Measurements

At harvest, ten guarded plants were randomly sampled from the two inner rows of each sub-plot to determine number of branches per plant, number of pods per plant, 100 seed weight and seed yield per plant. Seed protein (%), total nitrogen determined by Kjeldahl method according to AOAC (1980). The crude protein was calculated by multiplying nitrogen percentage by converting factor (6.25) (Robinson, 1975). Oil content (%), oil percentage of seed was estimated according to AOCS (1980) using soxhelt apparatus and petroleum ether (40-60°C) as a solvent. Electrical conductivity (EC), soil samples were collected, air-dried, ground and passed through a 2 mm sieve and by particle size distribution was prepared for the determination of the electrical conductivity by EC-meter (Richards, 1954).

Statistical methods, the analysis of variance was carried out according to Gomez and Gomez (1984). Treatment means were compared by Duncan's multiple range test (Duncan, 1955).

RESULTS AND DISCUSSION

Number of branches per plant

Salinity stress led to a significant reduction in number of branch per plant (Fig.1). The reduction in number of internodes and number of branches could severely be affected by salinity (Change et al., 1994). In other hand, application of proline gave the best number of branch per plant with saline treated plants as compared to the other treatments (Fig.1). Proline as osmoprotectants promotes plant growth and yield under normal or stress conditions due to its osmoprotective effect on photosynthetic machinery and regulation of ion homeostasis (Raza et al., 2007). Similarly, exogenous glycine betaine increased significantly the number of branches plant under saline stress (Fig. 1). Yield traits increased of stressed-plants after application of Glycine betaine has been proposed to be partly located on the increased net photosynthesis, decreased rate of photorespiration, stomatal conductance, induced more efficient gas exchange (Makela et al .,1998b). Regarding to compost application increased plant branch number under saline stress as compared to the untreated plants Figure (1). Organic matter (OM) can act as salt ion binding agents which detoxify the toxic ions, particularly Na⁺ and Cl⁻ (Eletr et al., 2013).

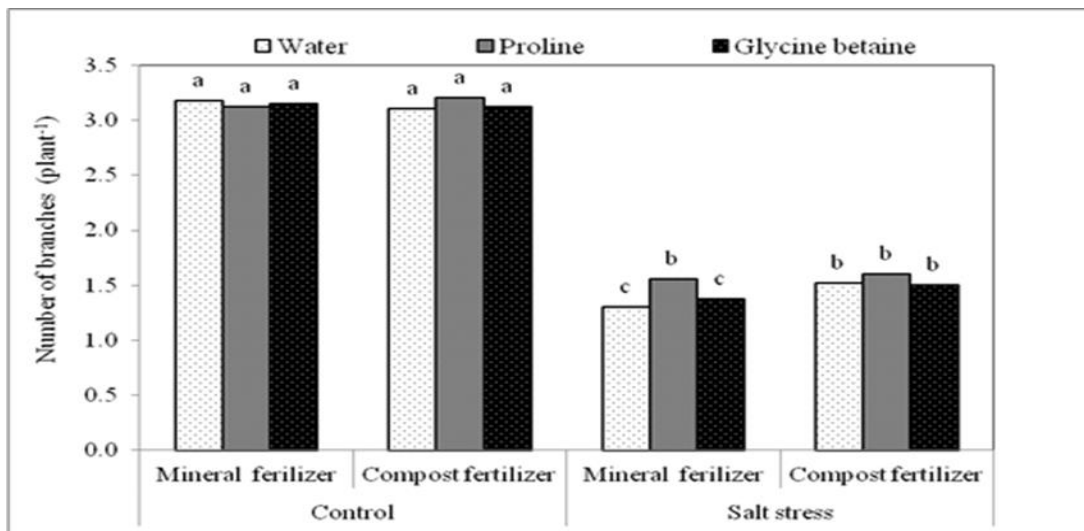


Figure 1. The effects of salinity on number of branches per plant in soybean as affected by compost application and exogenous proline and glycine betaine. Means followed by the same letters in each trait are not significantly different at 5% level, according to Duncan's test.

Number of pods per plant

Salinity stress induced significant decrease in the number of pods per plant (Fig. 2). These results agree with Taffouo et al. (2009) who reported that, the significant decrease of yield components under salt stress would be partly related to a significant reduction of foliar chlorophyll contents in saline medium. While, exogenous of proline enhanced the yield traits under salinity conditions (Fig. 2). Umar and Bansal (1995) have been found that exogenous application of both osmoregulators lead to stronger stem and roots, improved branching, earlier flowering and greater number of pod due to increasing water use efficiency. Similarly, application of glycine betaine at saline stressed plants increased number of pods per plant (Fig.2). The increase on branch number could be due to increased translocation of photo-assimilates in seed during the seed filling stage (Agboma et al., 1997). Regarding to compost application caused growth enhancement under salinity (Fig.2). Possibly due to the beneficial effect of compost on the physicochemical properties affecting plant growth such as soil structure, available water and soil salinity. By supplying nutrients, particularly OM can improve the mineral nutrient status and growth of plants in saline soils (Walker and Bernal, 2004).

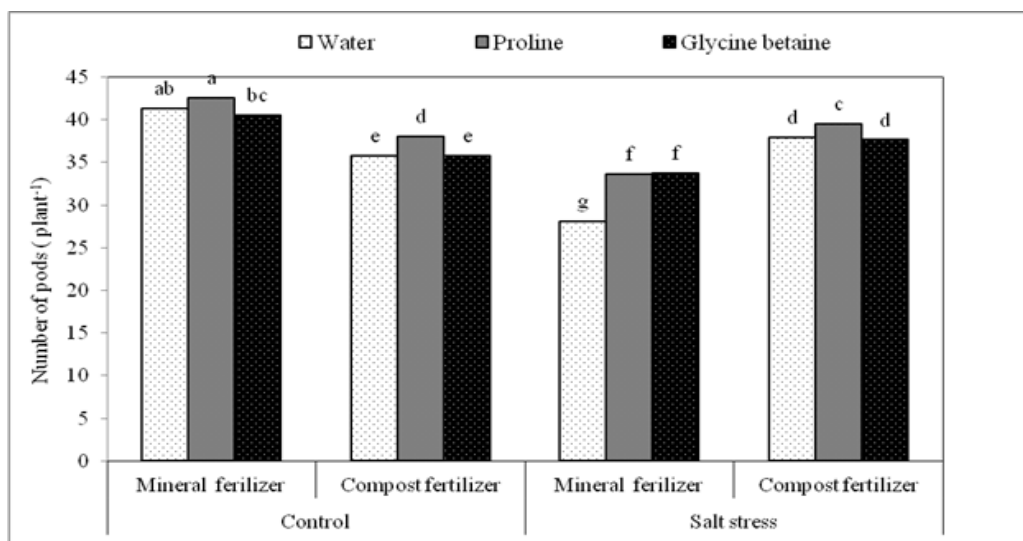


Figure 2. The effects of salinity on pods number per plant in soybean as affected by compost application and exogenous proline and glycine betaine. Means followed by the same letters in each trait are not significantly different at 5% level, according to Duncan's test

100-seed weight

The 100-seed weight decreased significantly under salinity stress (Fig.3). Siddique and Kumar (1985) reported that under saline stress and thus the 1000-seed weight was reduced seriously. In contrast, the 100-seed weight increased significantly with application proline under saline stress as compared to the untreated plants under salinity (Fig.3). Proline as osmoprotectants promotes plant growth and yield traits under normal or stress conditions due to its osmoprotective effect on photosynthetic machinery and regulation of ion homeostasis (Raza et al., 2007). Likewise, the 100-seed weight increased significantly with application glycine betaine under saline stress as compared to the untreated plants under salinity (Fig.3). These increase could be due to increased translocation of photo-assimilates in grains during the grain filling stage. Comparison of earlier reports in the literature with the present study shows that effective and efficient doses of GB application depend on type of species (Agboma et al., 1997).

Moreover, Positive effects of organic compost on 100 seed weight (Fig.3). Might be due to the fact that nitrogen composes protoplasmic protein, which required for increasing the 100 seed weight, similar results were reported by Awad (2004).

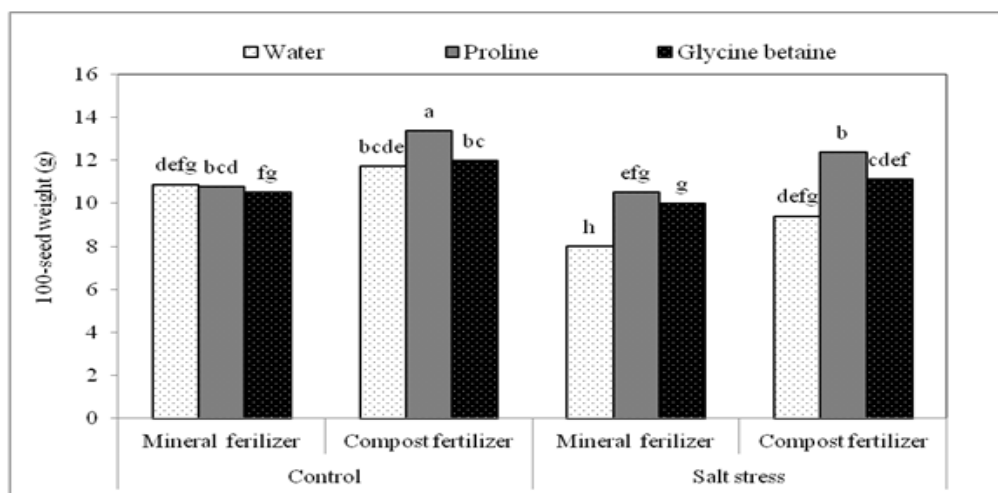


Figure 3. The effects of salinity on 100-seed weight in soybean as affected by compost application and exogenous proline and glycine betaine. Means followed by the same letters in each trait are not significantly different at 5% level, according to Duncan's test

Seed yield

Seed yield decreased significantly under saline stress in soybean (Fig.4). Reduction in seed yield due to salinity stress was also reported for soybean (Ghassemi-Golezani et al., 2010). Disturbed water and nutritional balance of plants may cause reduced crop yield in saline conditions (Muhammad and Hussain, 2010). These results agree with El-Zeiny et al (2007) observed a negative relationship between seed yield and salinity stress. Contrary, application of proline under salinity increased seed yield per plant (Fig.4). The proposed functions of accumulated proline are osmoregulation, maintenance of membrane and protein stability and growth (Wahba et al., 2007). Accumulated proline may supply energy to increase salinity tolerance and may occur through an increase in its synthesis constantly with inhibition of its catabolism (Jaleel et al., 2007) and may be a mechanism for stress tolerance (Demir, 2000). Role of proline during stress and the signalling events that regulate stress-induced accumulation is vital in developing plants for stress tolerance. From the above discussion we conclude that Pro is comparatively more important than GB in salinity tolerance. As well as, application of glycine betaine under salinity increased seed yield per plant (Fig.4). Generally, exogenous application of glycine betaine at both concentrations enhanced the yield and its components under salinity stress. The increase in seed yield due to osmoregulators may be the result of any of these reasons; increasing potassium content (Farouk ,2000). The seed yield increase following the application of GB could be associated with the greater number of filled seeds and more large seeds (Agboma et al., 1997). Similar trends have been obtained with approximate increase in experiments conducted in the greenhouse of commercial vegetable grower in Finland but with unstressed tomato plants (Makela et al., 1998a). Also, the application of compost increase in seed yield (Fig.4). Might be due to improvement of the structure of the soil by increasing the soil water holding capacity which gave rise to good aeration and drainage that encourage better root growth and nutrient absorption (Saleh et al., 2003)

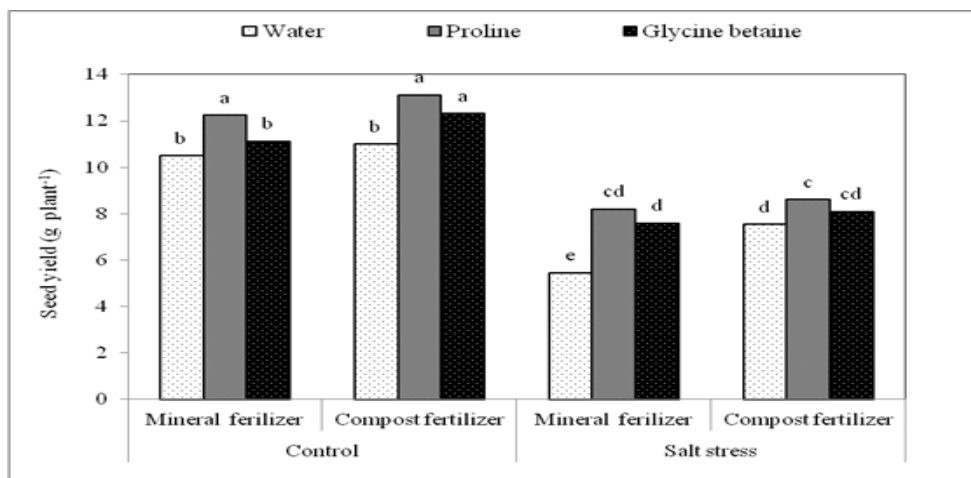


Figure 4. The effects of salinity on seed yield in soybean as affected by compost application and exogenous proline and glycine betaine. Means followed by the same letters in each trait are not significantly different at 5% level, according to Duncan's test

Seed protein

There appears a significant decrease in protein content with salt stress. Data in (Fig.5) these reductions may be attributed to the weakening of salinity to protein - pigment – lipid complex or enzyme activities (Taffouo et al., 2009). Medhat (2002) reported that salinity stress induce changes in the ion content of plant's cells, which intern induce changes in the activity of certain metabolic systems that might have serious consequences for protein. Application of proline induced significant increases in protein percentage (Fig.5). While, the increases in protein percentage of stressed soybean in response to proline might be attributed to the increase in leaf photosynthetic pigments and assimilates (Fig.5). Therefore, translocation of assimilates from stem to seeds is the main source as well as increased factors for growth and development of seeds (Sadak and Dawood ,2014). As well as, glycine betaine improved the seed protein content (Fig.5) due topromotes protein contentunder normal or stress conditions due to its osmoprotective effect on photosynthetic machinery and regulation of ion homeostasis (Ali, 2011). Regarding to application of compost showed slight increase of protein content under non-saline as for nitrogen (Fig.5). Salinity generally reduces in nitrogen content in plant organs. An increase in chloride uptake and accumulation is accompanied by a decrease in shoot nitrogen concentrations due to the competition between chloride and nitrate, which decreases nitrate content (Sakr et al., 2007).

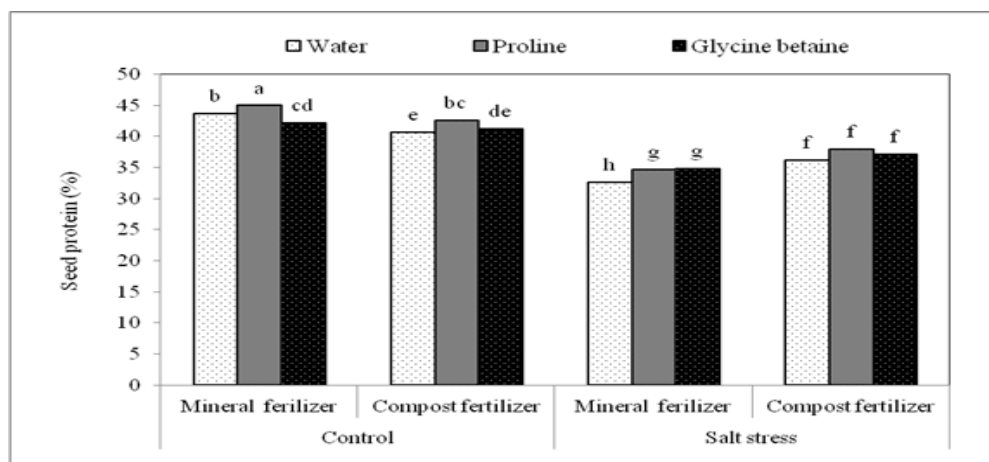


Figure 5. The effects of salinity on protein (%) in soybean as affected by compost application and exogenous proline and glycine betaine. Means followed by the same letters in each trait are not significantly different at 5% level, according to Duncan's test

Seed oil

The effect of salt stress on oil content was significant (Fig.6). Soybean seed oil contents influenced by environmental factors such as salinity (Nakasathien et al., 2000). Mean comparisons showed that the lowest oil percentage was obtained in salt stress these results agree with Zadeh and Naeini (2007) who reported that the

reduction may be attributed also to the weakening of salinity to protein – pigment – lipid complex or enzyme activities. But, the increases in seeds quality of stressed soybean in response to proline (Fig.6). Proline might be attributed to the increase in leaf photosynthetic pigments and assimilates. Therefore, translocation of assimilates from stem to seeds is the main source as well as increased factors for growth and development of seeds (Dawood and Sadak, 2014). As a result, exogenous glycine betaine improved the seed oil content (Fig.6). Glycine betaine showed ameliorating effects on these seed oil and has an antioxidant role in protecting plants from salt-induced oxidative damages. Similar, under saline conditions, Glycine betaine mitigates the inhibition of growth of plant cells and Glycinebetaine as an efficient antioxidant system often correlates with the alleviation of oxidative damage and improved tolerance to salt stress (Malekzadeh et al., 2012). Regarding to application of compost at was more effective on seed oil content than control might be due to the fact that nitrogen composes protoplasmic protein, which required for increasing the growth and then increasing the seed oil content and oil yield (Fig.6). These results may be due to the effect of organic manure by improving the physical structure of the soil and increasing available nitrogen, which reflects the greater growth and, consequently, more absorption of nitrogen and more crude protein synthesis. These results are in line with those obtained by El-Bana (2000).

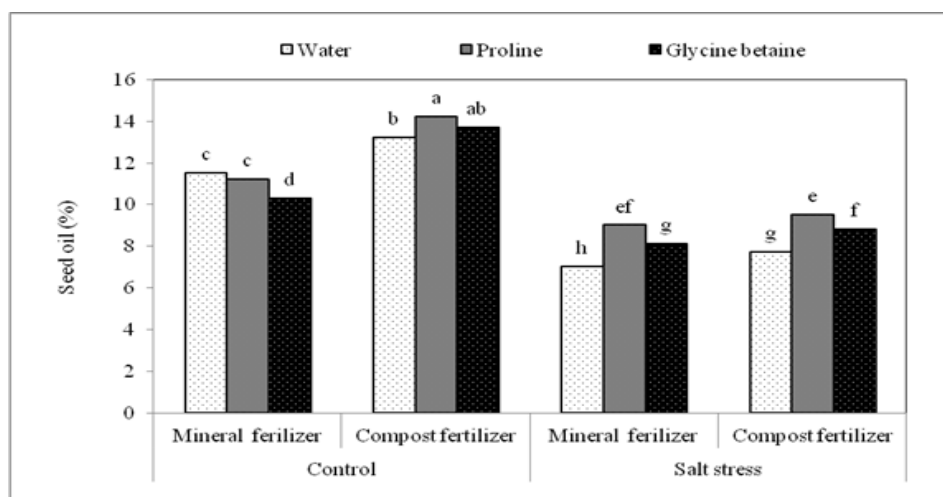


Figure 6. The effects of salinity on seed oil content in soybean as affected by compost application and exogenous proline and glycine betaine. Means followed by the same letters in each trait are not significantly different at 5% level, according to Duncan's test.

Electrical conductivity (EC)

With respect to the values of electrical conductivity (Fig.7) which shows that increase in EC is directly proportional under salinity stress and has decreased EC under compost application. The increment in EC values may be due to the released organic acids from the decomposed compost. These results are agreement with those reported by Saharinen et al. (1996). Addition of organic matter to the surface soil has decreased dispersion and EC down to the subsoil, compared to the addition of gypsum alone (Vance et al., 1998).

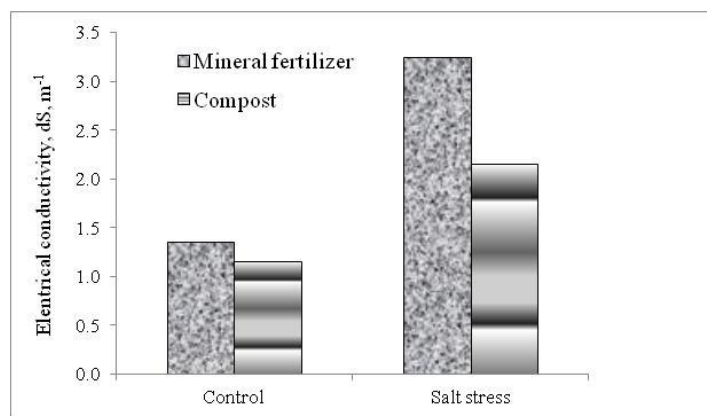


Figure 7. Effects of compost application on EC under salinity.

CONCLUSION

It is concluded that exogenous application of proline and glycine betaine combining with compost application alleviated detrimental effects of salt stress and may be a practical approach to improve salt stress tolerance in soybean.

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