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Alleviative Effects of Zinc on Biomass Yield and Antioxidative Enzymes Activity in Leaves of Soybean (*Glycine max* L.) Under Salt Stress

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Abstract: The present study was conducted to determine the interaction effects of zinc availability and salt stress in Bangladeshi soybean cultivar (cv. Shohag) whether zinc can alleviate the hazardous effects of salt stress or not. In this study, the plants are grown in zinc treated soil and also exposed to increasing (0, 50, 100, 150, 200, and 250 mM NaCl) levels of salinity. The results showed that the dry weight of root, stem, leaves, petioles and total dry weight were significantly reduced by salinity. The activities of antioxidant enzymes, lipid peroxidation, proline content were significantly affected by salt stress. Zinc supplementation helped the plants to cope with the salinity stress by improving the total dry weight. The antioxidant enzyme activities including catalase (CAT) and ascorbate peroxidase (APX) and proline content increased in response to salinity. The extent of lipid peroxidation noticed in salt stressed plants. However, zinc application enhanced catalase and ascorbate peroxidase activity as well as proline content in growing plants at different salt concentrations. The interaction between zinc and salinity significantly reduced lipid peroxidation. Application of zinc to salt-stressed plants ameliorates the salinity induced hazardous effects by enhancing the activities of antioxidant enzymes such as CAT and APX and Proline content.

Keywords: Soybean, Salinity, Zinc, Biomass, Antioxidant Enzyme, Lipid Peroxidation

1. Introduction

Among other natural disasters, salinity is one of the most pernicious abiotic stresses which reduces crop production worldwide. Salinity causes ion toxicities, osmotic stress, ionic imbalance, oxidative stress, genotoxicity which limits normal growth and development of plants by diminishing cell division and expansion, reducing photosynthetic efficiency, modifying metabolic processes [1–7]. High level of salinity induces reactive oxygen species (ROS) generation in plant cell [8, 9]. ROS formation takes place in chloroplasts, mitochondria, peroxisomes, plasma membrane of plant cells. [10–12]. These ROS can act as signaling molecules, mediating many key physiological processes. But overproduction of ROS causing damages to proteins, lipids, and eventually leading to cell death [5, 13, 14]. Therefore, plants possess various defense mechanisms including enzymatic and non-enzymatic defense systems to combat the harmful effects of ROS. Protection against oxidative stress is mediated by an enzymatic antioxidant mechanism that includes catalase (CAT), superoxide dismutase (SOD), peroxidase (POX) and enzymes of the ascorbate-glutathione cycle as ascorbate peroxidase (APX), monodehydroascorbate reductase (MDHAR), dehydroascorbate reductase (DHAR) [15–17] and non-enzymatic antioxidants such as phenolics, flavonoids, tocopherol, ASC and GSH [18–20]. In plants, superoxide dismutase scavenges superoxide anions and converts them to hydrogen peroxide [21, 22]. Catalase is the second line of defense which converts lethal hydrogen peroxide to water and molecular oxygen [23]. Another versatile antioxidant enzyme is ascorbate peroxidase which makes use of ascorbate (AsA) as electron donor and scavenges H_2O_2. Ascorbate peroxidase and catalase both are H_2O_2 scavengers, where increase APX activity can overcome

salinity-induced reduction in CAT activity [13, 24]. The combined activities of the multiple forms of these enzymes in different subcellular compartments adjust ROS production rates and elimination rates, while maintaining H_2O_2 at the levels required for cell signaling [23]. It was reported that an overaccumulation of H_2O_2 enhance malondialdehyde (MDA) content assuring salinity induced oxidative stress condition in plant [25–27]. Evidence suggested that when plants are exposed to abiotic stress, the oxidative cell damage in plants is controlled by the ability of the antioxidative systems to function [28].

Proline accumulation is one of the most common adaptive mechanism in plant under salinity stress. It was stated that elevated salt tolerance is associated with the high level of endogenous proline accumulation in plants [29–32]. This metabolite has a plenty of defensive properties, including stabilizing of cellular structure and decrease in harm of photosynthetic device, redox status balance, cytosolic pH upkeep, stabilization of protein structure [33]. The reason of high proline content is the enhanced activities of ornithine amino transferase (OAT) and pyrroline -5-carboxylate reductase (P-5-CR), the enzymes involved in proline biosynthesis [34] as well as the inhibition of proline oxidase, proline dehydrogenase (PDH), proline catabolizing enzymes [35].

Now-a-days researchers have adopted different strategies to improve crop production and quality. For instance, micronutrient supplementation at required levels maintains plant growth and enhances crop yield by limiting the detrimental effects of stressors [36–38]. However, Zn is considered an essential plant nutrient, which plays an integral part in the formation of chlorophyll, protein, lipid, carbohydrate, enzymes cofactor (DNA and RNA polymerase) and the action of hormones. [39]. Zinc is the main constituents of zinc finger and this protein acts as a structural motif crucial for DNA binding [40].

Soybean is mostly cultivated as oil yielding crop throughout the world that often suffers from salinity problem. It is salt sensitive crop. Global crop production is hampered due to the increase levels of salinity in large arable land. Zinc plays a vital role in protecting critical cellular components such as chlorophyll content by preventing their oxidation [41, 42] and IAA metabolism [43]. Several workers concluded that noxious effects of Na^+ and Cl^- ion can be alleviated by supplementing the soil with zinc [8, 14, 44–48]. Thus, this study was conducted with the aims of evaluating the effects of zinc on growth and the changes in antioxidant enzymes in soybean in response to salinity.

2. Materials and Methods

2.1. Plant Materials and Treatments

The experiments were conducted in 2019 at Plant Physiology and Biochemistry Laboratory of Botany Department, Jahangirnagar University, Savar, Dhaka, Bangladesh. For this experiment certified seeds of soybean (cv. Shohag) were collected from Bangladesh Agricultural Research Institute (BARI). The soil samples were dried and crushed and sufficient water was added to saturate the soil. 10 mg/kg zinc ($ZnSO_4 \cdot 7H_2O$) was mixed thoroughly with soil, and pots were filled with zinc treated soil. The seeds were surface sterilized with 0.1% sodium hypochlorite for 3 minutes followed by washing 3 times with distilled water. The experiment was carried out in randomized block design with three replications. There were five treatments at different concentrations for the experiment viz 50, 100, 150, 200 and 250 mM NaCl, distilled water considered as control and the cultivar were also exposed to salinity stress along with zinc sulfate. Seeds of uniform size were directly sown on 4th February 2019. Distilled water was applied in all pots up to the emergence of seedling. After seedling establishment distilled water in control pots and 12.5 mM NaCl solution were applied in salt treatment. When the first leaf appeared, actual amount of NaCl solution were applied. The salt solutions were applied till maturity. The plants were kept under natural sunshine till harvesting. After 120 days plants were harvested, and leaves, petioles, stems and roots were separated to determine their dry weight.

2.2. Enzyme Assays

For the measurement of the activity of CAT, the leaf samples (100 mg FW) were homogenized to a fine powder with a mortar and pestle. Subsequently, the leaf samples were added to 1ml of extraction buffer containing 50 mM potassium phosphate (pH 7.8) and 1 mM Na-EDTA, and 20 mg of PVPP. The crude homogenate was centrifuged at 15,000 rpm for 10 minutes at 4°C. The supernatant of the sample was used in the assay of the activity of CAT. For the measurement of the activity of CAT the supernatant was applied to PD-10 desalting column filled with sephadex G-25 to desalt the supernatant. The activity of CAT was determined by the method of Aebi [49]. The supernatant (20 µl) was added to 970 µl of 50 mM potassium phosphate buffer (pH 7.8) containing 1 mM Na-EDTA. After the measurement of blank solution at 240 nm for 1 minute with the spectrophotometer (ROTINA 420R), 10 µl 0f 880 mM H_2O_2 was added. Then the absorbance of the sample solution was measured for 2 minutes at 240 nm with the spectrophotometer (ROTINA 420R). The activity of CAT was calculated using the extinction coefficient, 0.04 mM^{-1} cm^{-1}. In the present study, CAT activity was expressed as µkat of H_2O_2 m^{-2}.

For the measurement of the activity of APX, leaf samples (50 mg FW) were homogenized to a fine powder with a mortar and pestle. Subsequently, leaf samples were added to 0.5 ml of extraction buffer containing 50 mM potassium phosphate (pH 7.0), 1 mM Na-EDTA and 5 mM L-ascorbic acid, and 10 mg of PVPP. The crude homogenate was centrifuged at 15000 rpm for 10 minutes at 4°C. The supernatant of the sample solution was used in the assay of the activity of APX. The activity of APX was determined by the method of Nakano and Asada [50]. The supernatant (20 µl) was added to 910 µl of 50 mM potassium phosphate buffer (pH 7.0) and 50 µl of 5 mM L-ascorbic acid solution. After the measurement of blank solution at 290 nm for 1 minute with the spectrophotometer (ROTINA 420R), 20 µl of 50 mM H_2O_2 was added. Then the

absorbance of the sample solution was measured for 2 minutes at 290 nm with the spectrophotometer (ROTINA 420R). The activity of APX was calculated using the extinction coefficient, 2.8 mM^{-1} cm^{-1}. In the present study, APX activity was expressed as nkat of ascorbate m^{-2}.

2.3. Lipid Peroxidation

Lipid peroxidation was determined by estimating the malondialdehyde (MDA) content according to Health and Packer [51]. Leaf samples (50 mg FW) was homogenized in 0.1ml of 0.05% BHT and 0.9 ml of 20% TCA and 10 mg of PVPP. The crude homogenate was centrifuged at 15,000 g for 10 minutes at 4°C. The supernatant of the sample (400 µl) was added to 1.6 ml of 0.5% TBA and was incubated at 95°C for 30 minutes. The reaction was stopped by placing the test tubes in an ice bath. The samples were then centrifuged at 15,000 g for 5 minutes at 4°C. The absorbance of the sample solution was measured at 600 nm and 532 nm with the spectrophotometer (ROTINA 420R). The amount of MDA was calculated using the extraction coefficient of 155 mM^{-1} cm^{-1}.

2.4. Proline Content

Leaf samples were collected from the three soybean plants of each treatment in each of the three replications for the experiment. Extraction procedure and colorimetric determination with acidic ninhydrin reagent (2.5 gm ninhydrin/100 ml of solution containing glacial acetic acid, distilled water and orthophosphoric acid 85% at a ratio of 6: 3: 1) were carried out. Samples of 0.25 gm leaf fresh weight were grinded in a morter after the addition of a small amount (5ml) of quartz sand and lome of 3% (w/v) aqueous sulfosalicylic acid solution. The homogenate was filtered through 2 layers of glass fiber filter and the clear filtrate was then used in the assay. Glacial acetic acid and ninhydrin reagent (1ml each) were added to 1ml of the filtrate. The closed test tubes with the reaction mixture were kept in a boiling water bath for 1hour and the reaction was terminated in a water bath of room temperature (21°C) for 5 min. Reading were taken immediately at a wave length of 546 nm. The proline concentration was determined from a standard curve and calculated on a fresh weight basis.

2.5. Statistical Analysis

The experiment was arranged out in randomized block design with three replications. Statistical analysis of the collected data were performed using Analysis of Variance (ANOVA). All statistical analysis were performed with the SPSS statistical package (SPSS program 16.00). Data were presented as means for each treatment and means were compared using the Duncan test at the 5% probability level.

Table 1. *Effects of zinc on dry matter production of soybean cv Shohag under salinity.*

Treatments	Root dry weight (gm/plant)	Stem dry weight (gm/plant)	Leaf dry weight (gm/plant)	Petiole dry weight (gm/plant)	Total dry weight (gm/plant)	Root/shoot ratio
0 mM NaCl	2.33 ± 0.22 a	4.92 ± 0.62 a	1.24 ± 0.34 a	0.023 ± 0.012 a	8.51 ± 0.46 a	0.38 ± 0.03 a
50 mM NaCl	1.38 ± 0.05 b (59.23)	2.51 ± 0.77 b (51.02)	1.21 ± 0.20 ab (97.58)	0.016 ± 0.001 ab (69.56)	5.13 ± 0.6 b (60.28)	0.37 ± 0.04 a (97.36)
50 mM NaCl + Zn	1.71 ± 0.08 b (73.39)	3.12 ± 0.59 b (63.41)	1.49 ± 0.22 ab (120.16)	0.024 ± 0.004 ab (104.35)	6.34 ± 0.77 b (74.50)	0.37 ± 0.04 a (97.37)
100 mM NaCl	0.50 ± 0.08 c (21.46)	1.84 ± 0.36 bc (37.39)	0.98 ± 0.13 abc (79.03)	0.014 ± 0.018 ab (63.67)	3.34 ± 0.39 c (39.24)	0.18 ± 0.02 b (47.37)
100 mM NaCl + Zn	0.56 ± 0.05 c (24.03)	2.84 ± 0.76 b (57.72)	1.18 ± 0.51 abc (95.16)	0.016 ± 0.008 abc (69.56)	4.60 ± 0.63 c (54.05)	0.14 ± 0.01 b (36.84)
150 mM NaCl	0.29 ± 0.04 d (12.45)	1.59 ± 0.19 c (32.31)	0.87 ± 0.14 bcd (70.16)	0.008 ± 0.003 ab (35.89)	2.75 ± 0.27 cd (32.31)	0.12 ± 0.03 b (31.58)
150 mM NaCl + Zn	0.39 ± 0.03 d (16.74)	2.19 ± 0.57 bc (44.51)	0.94 ± 0.24 bcd (75.80)	0.013 ± 0.008 bc (56.52)	3.53 ± 0.78 d (41.48)	0.13 ± 0.04 b (34.21)
200 mM NaCl	0.23 ± 0.02 d (9.87)	1.47 ± 0.36 c (29.88)	0.69 ± 0.17 cd (55.64)	0.007 ± 0.004 b (30.77)	2.39 ± 0.39 de (28.08)	0.11 ± 0.02 b (28.15)
200 mM NaCl + Zn	0.25 ± 0.09 e (10.73)	1.49 ± 0.34 c (30.28)	0.79 ± 0.22 cd (63.71)	0.013 ± 0.004 bc (56.52)	2.52 ± 0.40 de (29.61)	0.11 ± 0.07 b (28.94)
250 mM NaCl	0.20 ± 0.06 d (8.58)	1.19 ± 0.21 c (24.19)	0.42 ± 0.20 d (33.87)	0.005 ± 0.001 b (19.66)	1.82 ± 0.36 e (21.39)	0.13 ± 0.07 b (34.21)
250 mM NaCl + Zn	0.22 ± 0.05 e (9.44)	1.25 ± 0.27 c (25.40)	0.47 ± 0.13 d (37.90)	0.007 ± 0.003 c (30.43)	1.93 ± 0.24 e (22.68)	0.13 ± 0.04 b (34.21)

***** Average value of 3 plants in each treatment. ***** Means in a column followed by the same letter do not differ significantly at 5% level and ± means standard deviation ***** Values within parenthesis indicate percentage relative to the control.

3. Results

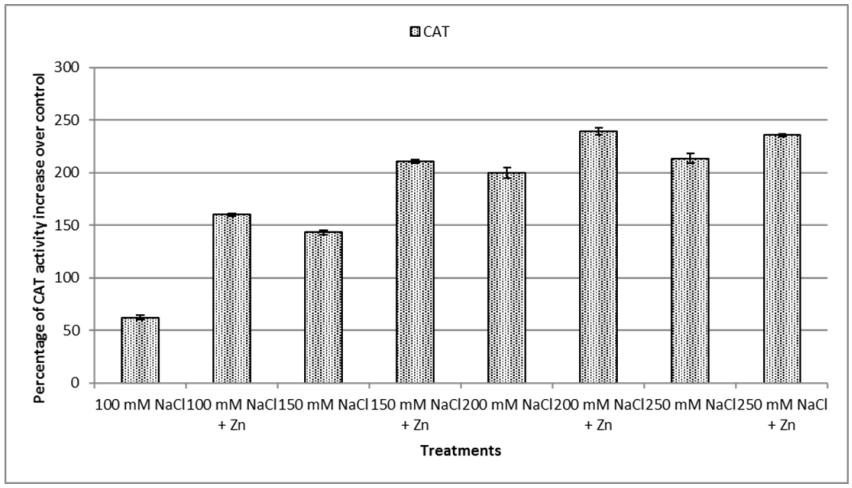
3.1. Biomass

Dry matter production in roots decreased by 40.77%, 78.54%, 87.55%, 90.13% and 91.42% under salinity treatments of 50, 100, 150, 200 and 250 mM NaCl, respectively (Table 1) (significant at <0.05). Zinc application on NaCl stressed soybean plants increased the dry matter production in roots by 23.91%, 12.00%, 34.48%, 8.00% and 10.00% under salinity levels at 50, 100, 150, 200 and 250 mM respectively (Table 1). Dry weight of stems decreased by 48.98%, 62.61%, 67.69%, 70.12% and 75.81% under salinity treatments of 50, 100, 150, 200 and 250 mM NaCl,

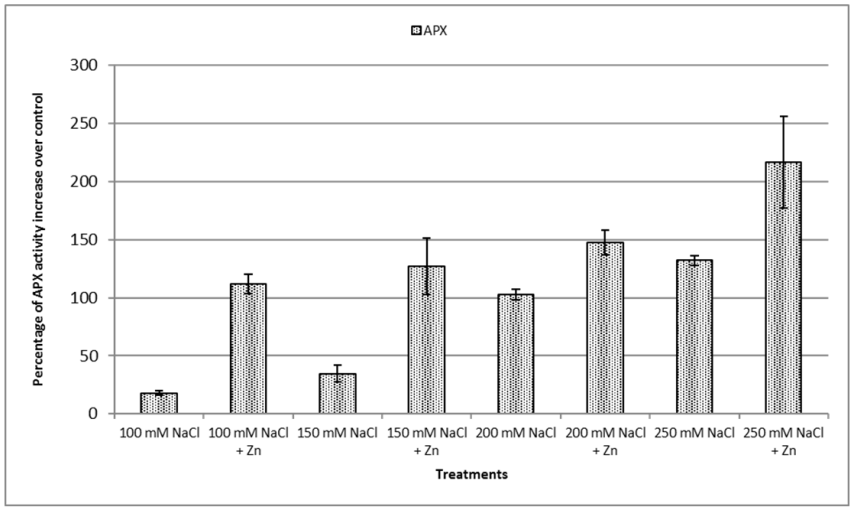
respectively (Table 1) (significant at <0.05). Whereas, zinc application on NaCl stressed soybean plants increased the dry weight of stems by 24.30%, 54.35%, 37.74%, 1.36% and 5.04% under salinity levels at 50, 100, 150, 200 and 250 mM respectively (Table 1). The dry weight of leaves decreased by 2.42%, 20.97%, 29.84%, 44.36% and 66.13% under salinity treatments of 50, 100, 150, 200 and 250 mM NaCl, respectively (Table 1) (significant at <0.05). Zinc application on NaCl stressed soybean plants increased the dry weight of leaves by 28.00%, 20.41%, 8.04%, 14.49% and 11.90% under salinity levels at 50, 100, 150, 200 and 250 mM, respectively (Table 1). The dry weight of petioles decreased by 30.34%, 36.33%, 64.11%, 69.23% and 80.34% under salinity treatments of 50, 100, 150, 200 and 250 mM NaCl, respectively (Table 1) (significant at <0.05). Zinc application on NaCl stressed soybean plants increased the dry weight of petioles by 50.00%, 14.29%, 62.50%, 85.71% and 40.00% under salinity levels at 50, 100, 150, 200 and 250 mM respectively (Table 1). Root/shoot ratio decreased by 2.64%, 52.63%, 68.42%, 71.85% and 65.79% under salinity treatments of 50, 100, 150, 200 and 250 mM NaCl, respectively (Table 1) (significant at <0.05). Results showed that root/shoot ratio was not significantly affected by zinc supplementation (Table 1).

3.2. Antioxidant Enzyme Activities

NaCl treatments increased CAT activity over the control with a maximal increase of 162.04%, 243.06%, 299.80% and 313.47% under salinity levels at 100, 150, 200 and 250 mM, respectively (Figure 1A) (significant at <0.05). Zn application further increased CAT activity in salt stressed plants by 60.49%, 27.75%, 13.12% and 7.09% under salinity levels at 100, 150, 200 and 250 mM, respectively (Figure 1A). NaCl treatments increased APX activity over the control with a maximal increase of 117.99%, 134.67%, 202.64% and 232.19% under salinity levels at 100, 150, 200 and 250 mM respectively (Figure 1B) (significant at <0.05). Zn application further increased APX activity in salt stressed plants by 79.36%, 68.61%, 14.59% and 36.45% under salinity levels at 100, 150, 200 and 250 mM respectively (Figure 1B).



A



B

Figure 1. *Changes in catalase (CAT) (A) and ascorbate peroxidase (APX) (B) activities in leaves of soybean cultivar Shohag as affected by salinity and zinc.*

3.3. Lipid Peroxidation

The oxidative damage was observed as malondialdehyde (MDA) content, which is a product of lipid peroxidation. Results showed that lipid peroxidation was significantly influenced by salt stress. MDA content was higher under saline conditions, compared with control. Salinity levels at 100, 150, 200, and 250 mM NaCl, caused 151.96%, 208.14%, 224.76% and 280.94% increase in leaf MDA content in non-zinc application plants, respectively (Figure 2) (significant at <0.05). However, lipid peroxidation significantly reduced by 18.83%, 10.44%, 9.08% and 12.62% at 100, 150, 200, and 250 mM NaCl, compared with NaCl treatments without zinc application, respectively (Figure 2).

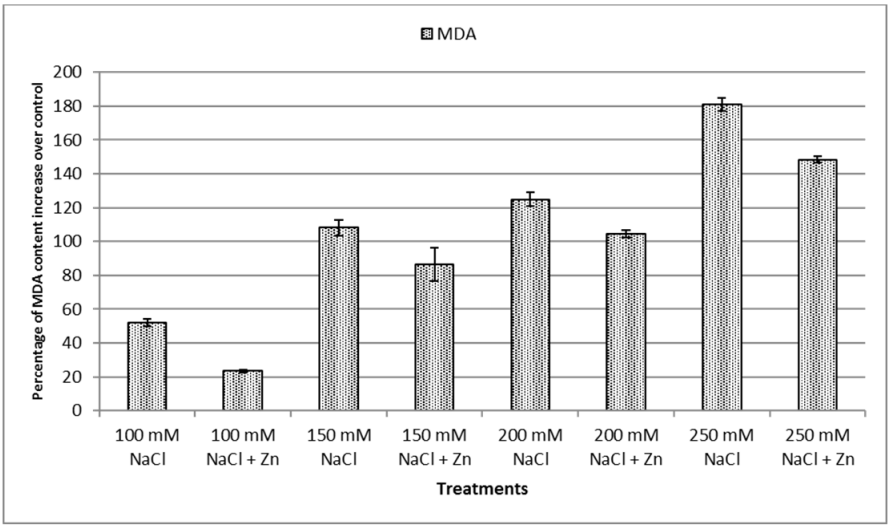


Figure 2. *Changes in MDA content in leaves of soybean cultivar Shohag as affected by salinity and zinc.*

3.4. Proline Content

The proline content in leaf tissue was significantly increased with increasing salinity. The free proline content was significantly enhanced in the stressed plants over control plants in both conditions (with and without zinc application). NaCl treatments increased proline content over the control with a maximal increase of 315.02%, 404.05%, 461.81% and 568.68% under salinity levels at 100, 150, 200 and 250 mM respectively (Figure 3) (significant at <0.05). However, with zinc application proline content significantly increased at 100, 150, 200 and 250 mM NaCl by 7.87%, 10.02%, 7.26% and 6.47% in leaves (Figure 3).

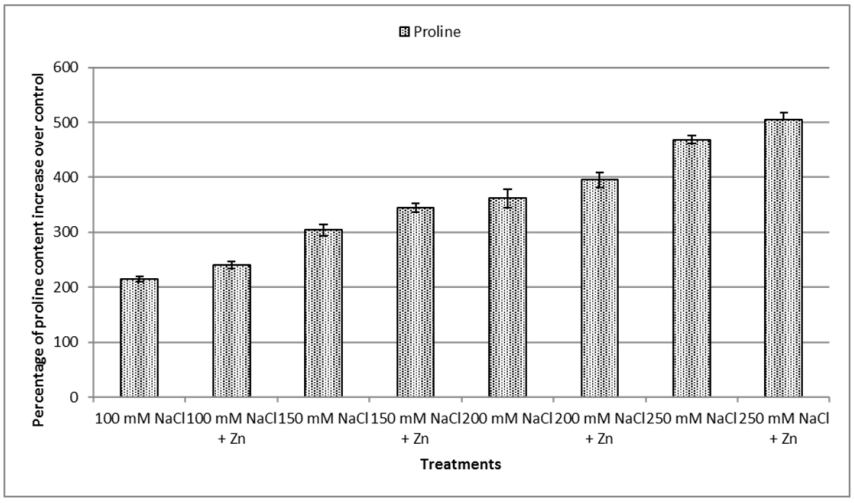


Figure 3. *Changes in proline content in leaves of soybean cultivar Shohag as affected by salinity and zinc.*

4. Discussion

In the present experiment, salinity caused a decrease in total dry weight of plants by reducing root DW, stem DW, petiole DW and leaf DW (Table 1). Khan *et al.* [52] observed a decrease in the growth and biomass yield of soybean due to salinization. Higher salinity levels counteract the normal growth of plants by negatively affecting various physiological and biochemical processes such as photosynthesis, antioxidant enzyme activity, nitrogen

metabolism, ionic homeostasis [53, 54] osmotic stress and low absorption of essential elements [55], proline metabolism [56]. On contrary, the total dry weight of salt-affected soybean plants increased with zinc application. Weisany *et al.* [45] and Jiang *et al.* [48] stated that Zn could increase dry weight of soybean plants under both normal and salinity stress conditions. Zinc has positive effects on the uptake and partitioning of important mineral elements [46, 57]. It plays a protective role by maintaining structural integrity of plasma membrane [46], increasing the natural auxin synthesis, thereby activating the cell division [58], accumulating phospholipid [59], improving protein synthesis [60] and inhibiting excessive Na^+ and Cl^- uptake [57, 61, 62].

Plants activate various antioxidant enzymes to mitigate the harmful effects of oxidative stress [23]. The enhancement of ROS production in saline plants induces an increase in antioxidant enzymes activity [4, 8, 40, 46]. In this study, salinity stress up-regulated the activities of CAT and APX in soybean plants. (Figure 1A and B). These results are analogous with earlier reports on, wheat and barley [63], mung bean [25], soybean [45], mustard [8]. Catalase is a tetramer containing heme group and helps in detoxification of ROS by converting 26 million of H_2O_2 into water within a minute [64]. APX is the only enzyme that can scavenge H_2O_2 in chloroplast while CAT is absent. Moreover, different isoforms of APX such as cytosolic APX (cAPX), mitochondrial APX (mtAPX), peroxisomal APX (pAPX) are H_2O_2 scavengers involved in AsA-GSH cycle [65]. Abbaspour [66] reported that the increase in APX activity could be due to the high concentration of intracellular H_2O_2 induced by salt stress. But zinc supplementation further increased CAT and APX activities under salinity stress (Figure 1A and B). Similar result was found by Weisany *et al.* [45] who suggested that Zn plays an important role in regulating the transmembrane redox homeostasis, thereby neutralizing the hazardous effects of ROS produced by oxidative stress by increasing activity of antioxidant enzymes. In contrast, reduction in catalase activity was observed in many plants species treated with zinc under salinity [8, 59]. Zn is indirectly required for a high activity of enzymes involved in H_2O_2 detoxification such as CAT [2]. The use of zinc is believed to increase the activity of the enzyme APX. This may be due to zinc's ability to participate in the biosynthesis of antioxidant enzymes [67].

MDA concentrations is an indicator of oxidative stress which determine the extent of lipid peroxidation. H_2O_2 induced membrane damage leads to acceleration of the Haber-Weiss reaction by generating hydroxyl free radicals, leading to increased lipid peroxidation [68]. Our study reported a significant increase in MDA content in leaves of soybean plants under the action of salt (Figure 2). The extent of lipid peroxidation occurs due to salt stress has been reported by several workers [27, 63, 69, 70]. However, consequences of an increase in H_2O_2 are the overproduction of membrane fatty acids, the denaturation of proteins as well as impeding DNA integrity [71, 72]. Zinc plays an indispensable role in controlling the formation and detoxification of free oxygen radicals following oxidation of lipid membranes [73]. Kawano *et al.* [74] concluded that Zn^+ have an inhibitory effects on membrane bound, NADPH oxidase. Results showed a significant decline in concentrations of MDA in leaves of soybean plants exposed to salt and zinc (Figure 2). These results are corresponding with previous research on soybean [45, 68], mustard [8], eggplant [59].

In our study, proline content was significantly enhanced in leaves of salt stressed soybean plants (Figure 3). Proline is considered as an osmolytes which contributes to osmotic adjustment, stabilizing subcellular structure, detoxifying free radicals as well as buffering cellular redox potential under salinity stress [68]. The findings are in accordance with other investigations [4, 33]. Proline's high accumulation aids in the maintenance of tissue water potential under stress conditions, accordingly offering an important resistance strategy [8, 36, 75, 76]. In saline plants, the activity of key enzymes involved in proline synthesis is increased, leading to an increase in proline [4]. In this study, the application of Zn increased the proline content of soybean plants under influence of salinity (Figure 3). Similar results were observed by other authors [8, 59, 77]. Zinc application induces a low availability of solutes, improves the accumulation of proline and allows plant cell to maintain a higher water content. Interestingly, many plants have lower proline content [45, 68]. According to Weisany *et al.* [45] the application of zinc in salted plants results in low proline content may be due to dilution effect.

5. Conclusions

Based on the study, it is concluded that salt stress adversely affects the biomass, antioxidant enzymes activities (CAT and APX), lipid peroxidation (MDA) and proline content in soybean plants. Zinc application to NaCl-stressed plants reduced lipid peroxidation and enhanced proline content and antioxidative enzymes activities. Thereby, zinc promotes plant growth under stress condition. Hence, it can be concluded that zinc could partially alleviate the adverse effects of salinity and provide better tolerance capacity to salt stress through osmotic adjustment as well as modulating the antioxidant enzymes activity and proline content.

Abbreviations

APX- Ascorbate peroxidase; AsC- Ascorbic acid; BHT- Butylated hydroxytoluene; CAT- Catalase; DHAR- Dehydroascorbate reductase; GSH- Reduced glutathione; H_2O_2- Hydrogen peroxide; IAA- Indole-3-acetic acid; MDA- Malondialdehyde; MDHAR- Monodehydroascorbate reductase; POX- Peroxidase; PVPP- Polyvinyl polypyrrolidone; ROS- Reactive oxygen species; SOD- Superoxide dismutase; TCA- Trichloroacetic acid.

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