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Full Length Research Paper

Role of gibberellic acid (GA_3) in improving salt stress tolerance of two wheat cultivars

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Abstract

Several environmental factors adversely affect plant growth and development and final yield performance of a crop. Drought, salinity, nutrient imbalances and extremes of temperature are among the major environmental constraints to crop productivity worldwide. Gibberellic acid (GA_3) treatment has alleviated the drastic effect of salinity in growth parameters (leaf area, dry weight of grains and photosynthetic pigments) and chemical constituents (carbohydrates, proteins, amino acids and proline content in two wheat cultivars (Sohag 3 and Giza 168). The effect of GA_3 on alleviation of damaging effects of different levels of salinity was studied in view of, wheat grains (*Triticum aestivum* L. Sohag 3 and Giza 168) were screened for germination and growth responses to different NaCl concentrations (50, 100, 150 and 200 mM). After four weeks, two groups; each of pots were sprayed with GA_3 (100 mg L^{-1} synthetic plant growth regulators). The result in this report reveals that the wheat cv. Sohag 3 was the most sensitive to salinity, while cv. Giza 168 was the most tolerant. Thus salinity stress had no effect on leaf area, photosynthetic pigment and consequently crop yield at mild salinity (100 mM), while in cv. Sohag 3, there was a marked and progressive reduction in these parameters by increasing the salinity stress even at the lowest salinity (50 mM) level used. Carbohydrate and proline content increased significantly by salinity stress in the different organs of the two wheat cultivars except for Giza 168 stem where carbohydrates were significant declined by salinity stress. Soluble protein content varied not only between the two wheat cultivars but also between the different organs. While salinity stress induced a significant increase in the soluble protein content in root and leaf, on the other hand it declined the soluble protein in stem of Giza 168. In cv. Sohag 3, the soluble protein content in root and stem decreased slightly by increasing salinity in the soil, this reduction was obvious only at higher salinization. While in leaves the soluble protein content increased markedly by salinity stress. Proline concentration in root, stem and leaf of both cultivars significantly increased with increasing salinity in soil. The accumulation was greater in the salt sensitive cv. Sohag 3 compared to the salt tolerant cv. Giza 168 especially at higher salinity concentration. GA_3 treatments (100 ppm) improved the growth criteria, photosynthetic pigments and consequently the crop yield of two wheats cultivars. This was injudged with the observable increase in protein content in the different organs of the two wheat cultivars.

Key word: Leaf area, carbohydrate, protein, proline, salinity, wheat cultivars, GA_3.

INTRODUCTION

Several environmental factors adversely affect plant growth and development and final yield performance of a crop. Drought, salinity, nutrient imbalances (including mi-neral toxicities and deficiencies) and extremes of temperature are among the major environmental constraints to crop productivity worldwide.

Development of crop plants with stress tolerance, however, requires, among others, knowledge of the

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physiological mechanisms and genetic controls of the contributing traits at different plant developmental stages. In the past two decades, biotechnology research has provided considerable insights into the mechanism of biotic stress tolerance in plants at the molecular level. Furthermore, different biotic stress factors may provoke osmotic stress, oxidative stress and protein denaturation in plants, which lead to similar cellular adaptive responses such as accumulation of compatible solutes, induction of stress proteins, and acceleration of reactive oxygen species scavenging systems. Recently, the authors try to improve plant tolerance to salinity injury through either chemical treatments as plant hormones, Handa et al. (1983) using cultured tomato cells adapted to water stress, found that the concentration of reducing sugars in the cells increased with the degree of adaptation to salinity concentration as high as 600 mM in the cells. It is a common knowledge that salt stress has an adverse effect on the growth and the related metabolites and consequently on the final yield (Abdel-Lateef, 2005). These alterations in plant growth and related metabolites could be due to decrease in natural growth hormones in plant tissues (Fusun et al., 2004).

Exogenous hormonal treatments were found to have adverse effect of stress conditions. Abd-El-Samad (1998), Abd-El-Samad and El-Komy (1998) and Azooz et al. (2004) reported that Indole acetic acida (IAA) and Gibberellic acid (GA_3) stimulate growth in sorghum under stress conditions.

There have been numerous studies of the effects of salinity on plants (Duan et al., 2008). Recently, investigations have focused more on the mechanisms of salt tolerance in plants (Munns and Tester, 2008). Some researchers have used plant growth regulators (PGRs) for reducing or eradicating the negative effects of salinity (Mutlu and Bozcuk, 2000). Phytohormones suggested playing important roles in stress responses and adaptation (Sharma et al., 2005). It is jgrowth could be related to a decline in endogenous levels of phytohormones (Debez et al., 2001). Wang et al. (2001) clearly defined that abassic acid (ABA) and jasmonic acid (JA) will be increased in response to salinity, whereas indole-3-acetic acid (IAA) and salicylic acid (SA) are declined. For example, the exogenous application of PGRs, auxins (Khan et al., 2004), gibberellins (Afzal et al., 2005), cytokinins (Gul et al., 2000) produces some benefit in alleviating the adverse effects of salt stress and also improves germination, growth, develop-ment and seed yields and yield quality (Egamberdieva, 2009; Majid et al., 2011).

The aim of this study was to evaluate the possible mode of interaction between salinity and exogenous application of GA_3 on growth (leaf and pigment content) as well as some physiological parameters of two wheat cultivars.

MATERIALS AND METHODS

Two wheat grains (*Triticum aestivum* L.) Sohag 3and Giza 168 were obtained from the breeding program of Seeds Center, Beni Suef, Egypt which were screened for germination responses to different NaCl concentrations (50, 100, 150 and 200 mM) and control (0.0). Three replicats from each concentration were used. Wheat grains were surface sterilized by immersion in a mixture of ethanol 96% and H_2O_2 (1:1) for 3 min, followed by several washings with sterile distilled water. Ten (10) seeds were sown in pots. Each pot contained 2 kg of clay soil. All pots were irrigated with different concentrations of NaCl solutions, corresponding to osmotic potential 50, 100, 150 and 200 mM. In order to maintain the water content of soil (moisture content) was kept near the field capacity using the corresponding salt solution (660 ml). After two weeks from the beginning of vegetative stage, one group each of pots were sprayed two times intervals with GA_3 (10 ml of 100 ppm).

After six months, plants were harvested and dried in an oven at 105°C to evaluate dry matter. Pigments were prepared from fresh leaves of plant. The contents of chlorophyll a, b and carotenoids were determined by using the spectrophotometer method of Metzner et al. (1965). The leaf area was determined by the methods of Norman and Campbell (1994), soluble carbohydrate was determined by the anthrone - sulphuric acid method (Fales, 1951), soluble protein was determined according to the method of Lowry et al. (1951) and proline according to the method of Bates et al. (1973).

Experimental design

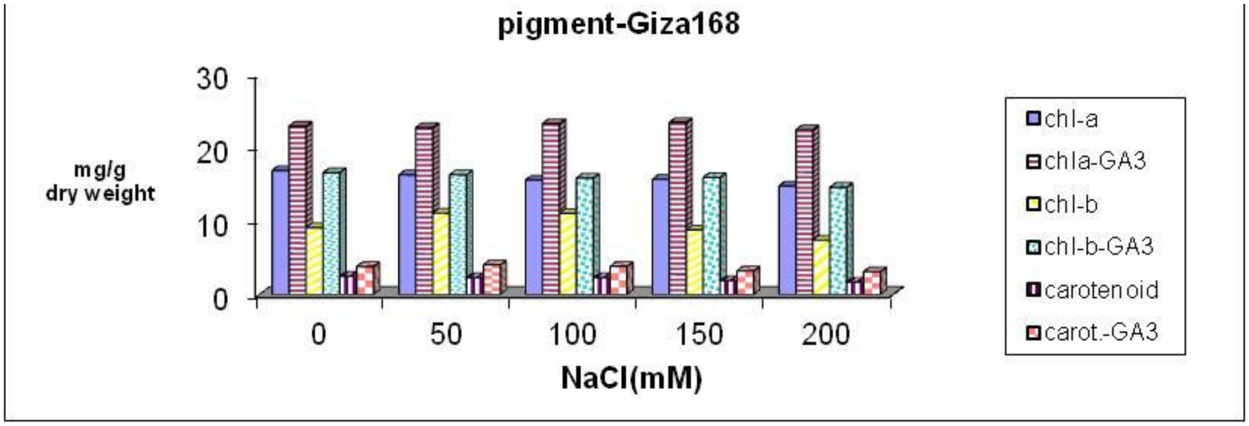
The pots were in four groups, each consists of 15 pots, group 1 and 2 specific for Sohag 3 plants while group 3 and 4 specific for Giza 168. Groups 1 and 3 represented the plants which grow under control and different salinization levels (0.0, 50, 100, 150, 200 mM), three replicats for each treatment. Groups 2 and 4 represented the plants which grow under previous salinization levels and spraying with 10 ml of 100 ppm GA_3.

Statistical analysis

The data of all experiments were subjected to one way analysis variance and means were compared using the least significant difference test (L.S.D.) using statistical program (sta.base.exe.) on computer (Steel and Torrie, 1960).

RESULTS

The data in Figure 1C and B show that leaf area and photosynthetic pigments declined significantly with increasing salinity stress in cv. Sohage 3. The rate of decline in these parameters was greater under 200 mM of NaCl. Salinity had no effect on leaf area and photosynthetic pigment at 50 mM salinity level for cv. Giza 168, and a significant reduction was obtained at higher salinization levels (Figure 1C and A). GA_3 treatments in most cases alleviated the inhibitory effect of salinity in leaf area and photosynthetic pigments of two wheat cultivars especially at 50 mM salinity levels. Moreover the values of leaf area in cv. Giza 168 were markedly higher than those of the control plants even at highest salinity level used as a result of GA_3 treatment. The results in Figure 4A show that soluble carbohydrate content increased significantly by salinity stress in the



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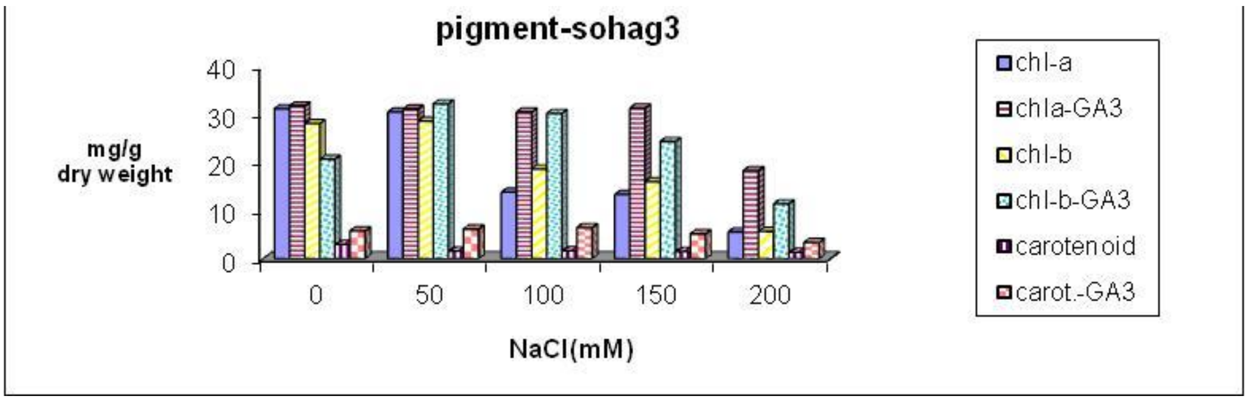

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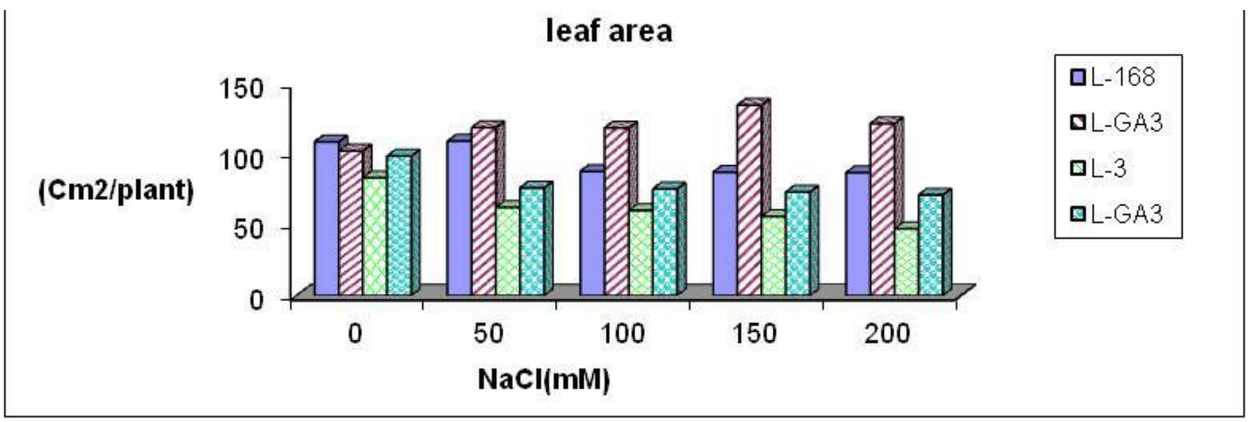

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Figure 1. Effect of salt stress and GA3 on leaf area (Cm2/plant) and pigments (mg/g dry weight) in two wheat cultivars Giza 168 and Sohag3.

different organs of the two wheat cultivars except for Giza 168 stem. Pronounced additional increase insoluble saccharides was recorded in the different organs of the two wheat cultivars as a result of interaction effect of salinity and GA_3 treatments as compared with the corresponding salinized plants.

Soluble protein content varied not only between the two wheat cultivars but also between the different organs

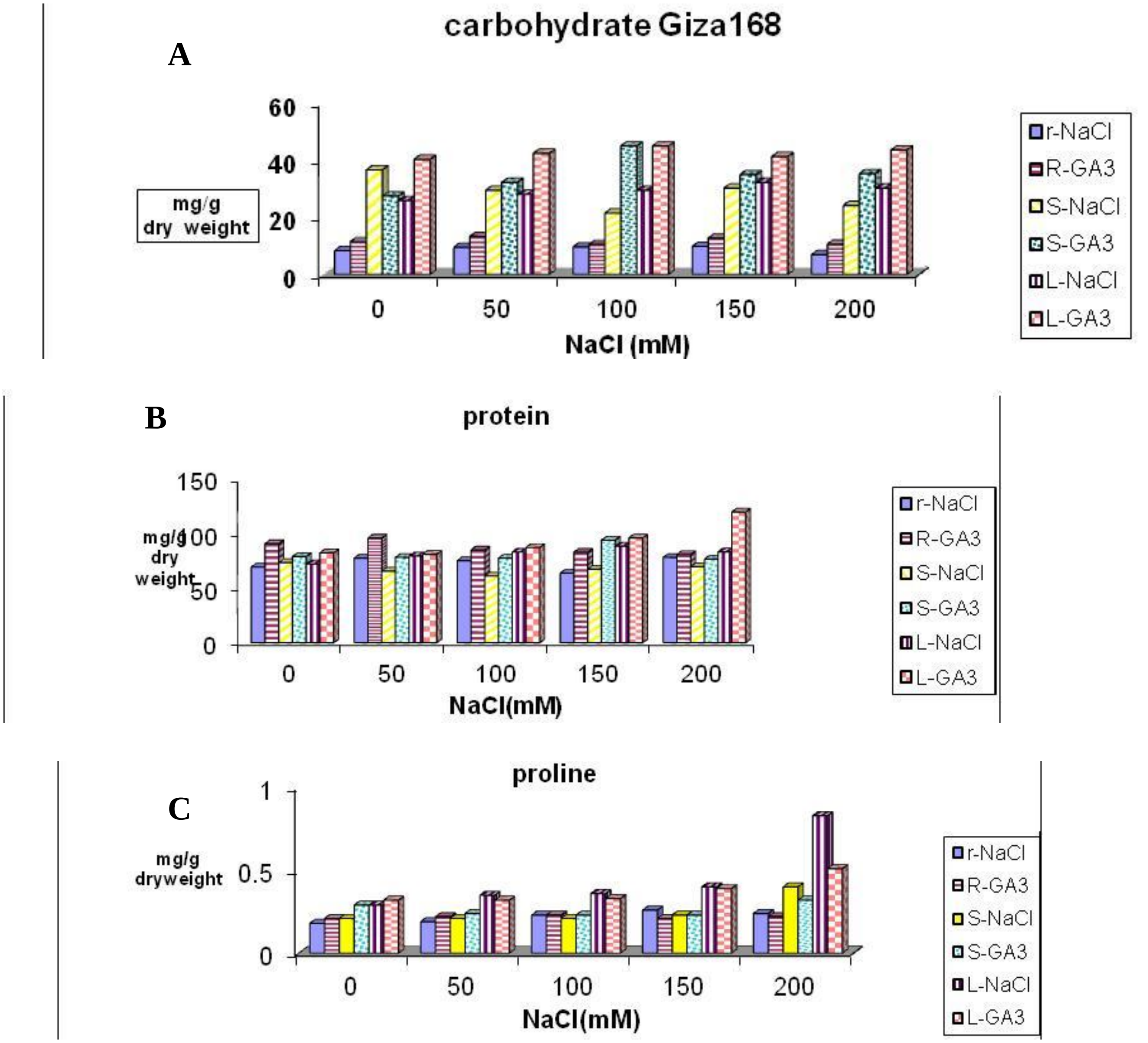
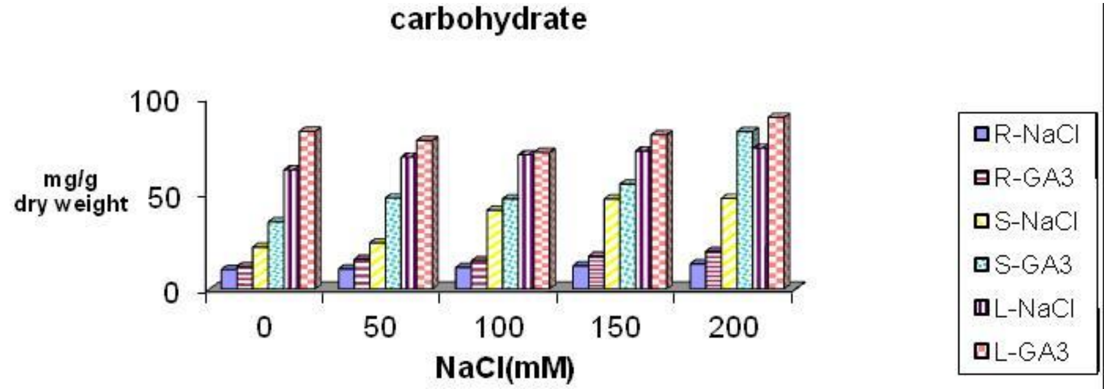


Figure 2. Effect of salt stress and GA_3 on carbohydrate, protein and proline (mg/g dry weight) in root, stem and leaf of Giza168

(Figures 3 and 4B). While salinity stress induced a significant increase in the soluble protein content in root and leaf, and on the other hand it declined the soluble protein in stem of Giza 168 (Figure 4B). In cv. Sohag 3 the soluble protein content in root and stem decreased slightly by increasing salinity in the soil, this reduction was obvious only at higher salinization (Figure 3B). While in leaves the soluble protein content increased markedly by salinity stress. GA_3 treatments activated in most cases the production of soluble protein in the different organs of the two wheat cultivars as compared with the corresponding salinized plants. Proline concentration in root, stem and leaf of both cultivars significantly increased with increasing salinity in the soil (Figure 2). The accumulation was greater in the salt sensitive cv. Sohag 3 compared to the salt tolerant cv. Giza 168 especially at higher salinity concentration. Moreover, proline content remained more or less unchanged in cv. Giza 168 at mild in root, stem and leaf of both cultivars especially at higher salinity levels (Figures 3 and 4C).

The crop yield represented as gram of dry weight per one spike of Giza 168 remained unchanged up to 100 mM, then a slight reduction was exhibited (Figure 2). While in cv. Sohag 3 a gradual reduction was obtained as



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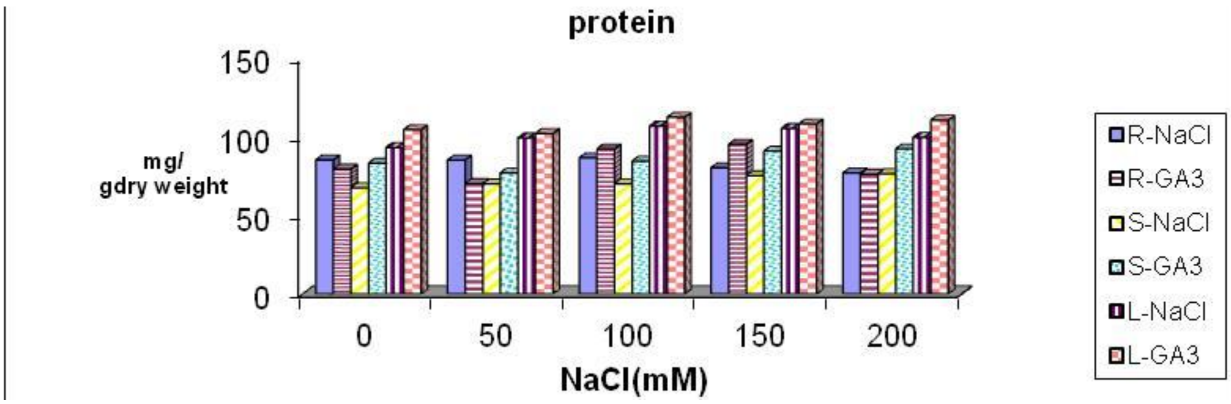

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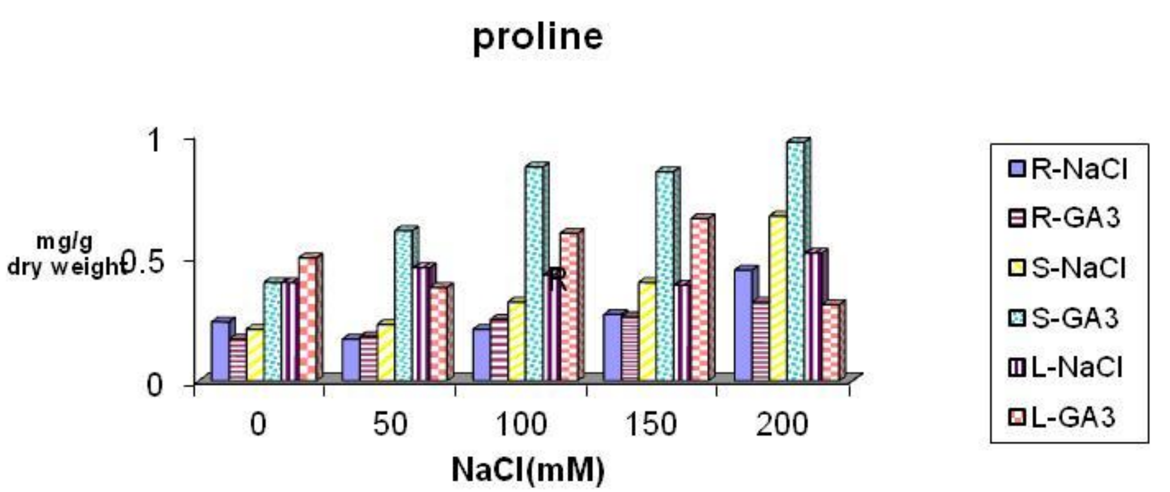

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Figure 3. Effect of salt stress and GA_3 on carbohydrate, protein and proline (mg/g dry weight) in root, stem and leaf of Sohag3.

a result of salinity stress, which was more obvious at the higher salinization level used .

DISCUSSION

The wheat cv. Sohag 3 was the most sensitive to salinity, while cultivar Giza 168 was the most tolerant. This response was obvious in the leaf area, photosynthetic pigments, crop yields as well as the related metabolic activities. The lack of any response to increasing NaCl concentration for change in leaf area, photosynthetic pigments and consequently crop yield (Spike dry weight) of cv. Giza 168 up to 100 mM NaCl concentration is

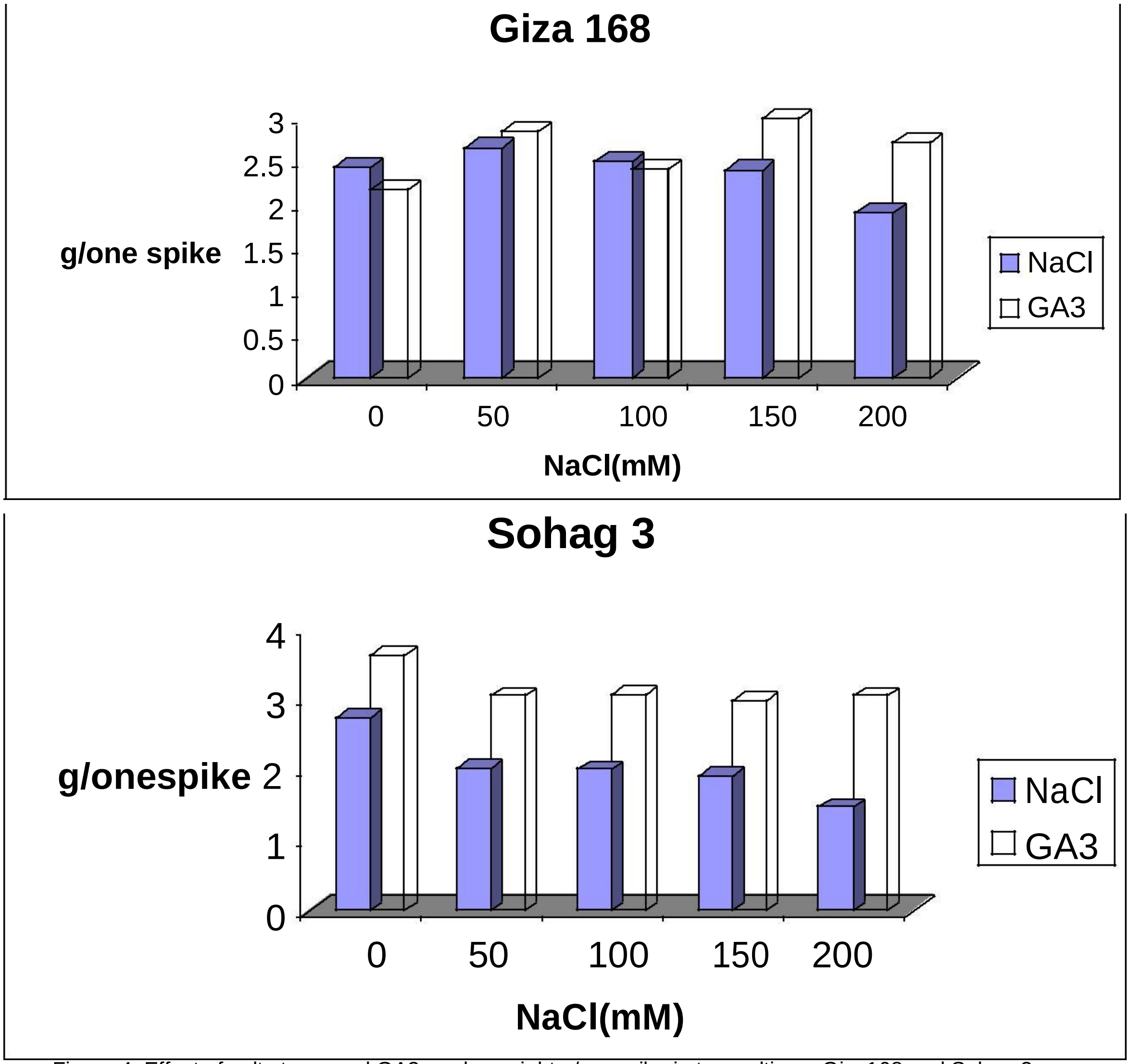

Figure 4. Effect of salt stress and GA3 on dry weight g/ on spike in two cultivars Giza168 and Sohag 3.

interpreted as a sign of slat tolerance. While in cv. Sohag 3 there was an inhibitory effect of salinity in leaf area, photosynthetic pigments and crop yield even at lower salinity. These differences in salt tolerance were accompanied by a large variation in the organic solutes, which consequently promoted differences in the water status (conservation and utilization) of the two wheat cultivars.

In cv. Giza 168, soluble saccharides and soluble protein in both roots and leaves were in most cases increased by salinity stress. This suggests that the salt adapted characteristics of cv. Gize 168 included saccharides and protein involved in tissue growth and osmoregulation (Yordanov et al., 2003; Abd-El-Samad et al., 2004). The relative ability of a plant to increase the concentration of solutes in its tissues (osmotic adjustment) will partially determine its tolerance to stress condition (Richardson and McCree, 1985; Marktester and Davenport, 2003).

The sensitivity of cv. Sohag 3 however was associated with a lowering in soluble protein in both root and stem including to a pronounced reduction in leaf area which consequently resulted to a progressive depletion in photosynthetic pigments and crop yield even at low salinity levels (Marcelis and Van Hooijdank, 1999; Parida et al., 2002; Abd El-Samadet al., 2004). Thus it can be concluded that salt tolerance appears linked with equilibrium and interconversion between saccharides and nitrogen metabolism whereas saline injury leads to metabolic disturbances in both components (Shaddad, 1990; Hasegawa et al., 2000; Abd El-Samad et al., 2004; Gonzalez et al., 2009). The differences in the accumulation of sugar between the two wheat cultivars could be a valid trait to discriminate genotypes of different tolerance to saline and osmotic stresses. Ballbrea et al.,

(1997) reported that it is important to know how sink-source relationships are affected in plants growing under stress condition because the efficient use of assimilate may be a limiting factor to plant growth under drought and salinity.

The physiological significance of proline accumulation is controversial. While some characters have reported that as a sign of stress (Rai et al., 2003; Claussen, 2005), others suggest that at high concentration it act as a soluble solute for inter cellular osmotic adjustment (Silveira et al., 2003). According to our results which may concluded that the reduction in leaf area and proline accumulation was higher and detected in the salt sensitive cv. Sohag 3 compared to the salt tolerant cv. Giza 168. On the other hand, proline content was higher in the stem of cv. Sohage 3 (salt sensitive cultivars) than stem of cv. Giza 168 at any concentration of NaCl. Therefore the physiological significance of proline is still complicated.

Synthetic plant growth regulators treatments (that is GA_3) increased adaptation of two wheat cultivars to salinity to some extent. Salt tolerance of these two wheat cultivars may be achieved through osmoregulation which in turn increased water flow and water status (conservation and utilization) using the organic solutes (saccharides and proteins), which in turn increased the photosynthetic area and crop yield of the two wheat cultivars. This activation was concomitant with increasing the photosynthetic area and crop yield of two wheat cultivars. This confirms the view of many authors (Manetas, 1990; Silveira et al., 2003; Abd-El-Samad et al., 2004; Hassanein et al., 2009).

GA_3 treatment not only alleviated the inhibitory effect of salt stress on crop yield of the two wheat cultivars, but also increased the crop yield of the two wheat cultivars.

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