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ORIGINAL ARTICLE

RESPONSE OF WHEAT TRITICUM AESTIVUM L. TO (ABA) ACID UNDER THE INFLUENCE OF SALINITY STRESS

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Abstract: Field experiment was conducted into season 2018 at the Al- Thi -Qar governorate. The study included two factors: The first factor is the cultivation of wheat in five different types of soils in terms of salt concentration (3.24, 4.23, 6.73, 7.21 and 9.32) milimos.cm⁻¹. These plants were sprinkled with six levels of abscisic acid ABA (0, 20, 40, 60, 80, 100 and 120) micromole.L⁻¹ to determine the effect of these factors and their interactions on the different characteristics such as total chlorophyll and other characteristics such as proline content in the root and in the root, glutathione reductase (GR), malondialdehyde (MDA), sucrose, glucose, starch, soluble sugar and non-structural carbohydrates (NSC). The results of the experiment showed significant differences in all traits except for MDA in the root and leaves, the third saline concentration 6.73 milimose.cm⁻¹ and the level of the fourth ABA 60 micromole.L⁻¹ were given the highest range of total chlorophyll rate, Proline content in the root and leaves, sucrose content, glucose content, starch content, soluble sugar in the leaves and NSC in the root of the wheat plant, plants those planted at the fifth saline concentration 9.23 milimose.cm⁻¹ and the level of the (0) ABA were superior to the highest rates of the antioxidant enzyme (GR) and proline in the root and leaves of the wheat plant, while the lowest of these enzymes and proline were at the first of saline stress 3.24 milimose. cm⁻¹. In current study, the ABA acid has a high role in reducing the harmful effects of salt stresses, the increasing concentrations of ABA and to a certain extent can protect the wheat plants and increases its resistance to saline stresses.

Keywords: Antioxidant enzyme, Abscisic acid, Saline stress. Wheat.

Cite this article

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1. Introduction

Wheat is the most widely grown crops in the world and provides 20% of the daily protein and of the food calories for 4.5 billion people [Goyal and Verma (2015), Flister and Galushko (2016)]. Many experiments have shown that an early exposure to stressor to certain limit can prepare the plants to respond to subsequent stress. Abscisic acid (ABA) mediated signaling also plays an important part in plant responses to environmental stress and plant pathogens. Abscisic acid is created within the roots in response to cut soil, water potential and different things within which the plant is also underneath stress. ABA also appears to be a key player in mediating the acclimation of plant to abiotic stress, ABA

accumulation helps to maintain high cytosolic K⁺ and Na⁺ homeostasis and better water status in maize exposed to salt stress, ABA performs a protective function in the plant by scavenging free radicals in plants under salt stresses. Many studies explained that the ABA mediates activation of its biosynthesis and inactivation of its degradation pathways to accumulate proline. A close linear correlation was found between the ABA content of the leaves and the stomatal resistance on a leaf area basis. Photosynthesis of wheat plants is a highly sensitive to changes in environmental factors such as salinity and the chlorophyll analysis is a sensitive method for the detection of changes in the photosynthetic apparatus. A better accumulation of

ABA in drought-tolerant wheat vascular plant C 306 and HW twenty-four than inclined Hira in response to water stress. Exogenous application of ABA has been reported to significantly increase the activities of SOD, CAT, APOX and GR and the contents of ascorbate reduced glutathione, α -tocopherol and carotenoids. Adaptation of plants to salt stress (*i.e.* resumption of growth after exposure to high soil salinity) requires cellular ion homeostasis involving net intracellular Na^+ and Cl^- uptake and subsequent vascular compartmentalization without toxic ion accumulation in the cytosol [Datta *et al.* (2009)]. The effect of salinity on plants may cause disturbances in plant metabolism, a common consequence of most abiotic associate degree organic phenomenon stresses is an accrued production of reactive atomic number 8 species (ROS). Oxidative stress in numerous crop plants has been reportable in response to salinity, this noxious ROS causes injury to polymer, proteins, lipids, pigment. Therefore, the aim of this study is to assess the influence of sprinkling ABA on the various biochemical and morphological changes associated with the wheat plant that is cultivated under the different concentration of salinity stress.

2. Materials and Methods

Preparation of spraying a solution with ABA acid

The first factor is the cultivation of the wheat plant in different five types of soils in terms of salt concentration (3.24, 4.23, 6.73, 7.21 and 9.32) milimos/cm. These plants were sprinkled with six levels of abscisic acid ABA (0, 20, 40, 60, 80, 100 and 120) micromole, the ABA acid was sprayed in the early morning on the vegetative part of the wheat plant at the completion of the emergence of the sixth plant leaf until the wetness and the fall of the first drop of the solution from the plant to determine the effect of these factors and their interactions on the different growth characteristics and the specific qualities of wheat grain. The five different plots were plowed into two times orthogonal, each plot was then divided into 3 replicates, each replicate has 7 experimental units with an area of 1 m² each for them. Urea fertilizer 46% N was added by 50 kg in four equal instalments, the first at the time of cultivation and the second at the stage of completion of the fourth leaf and the third when the second node appeared on the stem, while the fourth at the booting stage. 25 kg of Super Phosphate triple 46% P₂O₅ was added as a single batch when preparing the soil of plot.

Plants were planted in the form of lines with a distance of 20 cm. The seeds of the Baghdad variety were grown by 30 kg/ha on 15/11/2018. Preparing the solution according to the required concentrations and dissolve it in 2.5 ml of ethyl alcohol of 50% concentration, a substance was added to the solution (liquid soap) to reduce surface tension and to ensure complete wetness:

1. Spraying with 0 μM ABA (spraying using distilled water only).
2. Spraying with 20 μM ABA (prepared by dissolving 5.33 mg of ABA acid in 1 liter distilled water).
3. Spraying with 40 μM ABA (prepared by dissolving 10.66 mg of ABA acid in 1 liter distilled water).
4. Spraying with 60 μM ABA (prepared by dissolving 16 mg of ABA acid in 1 liter distilled water).
5. Spraying with 80 μM ABA (prepared by dissolving 21.33 mg of ABA acid in 1 liter distilled water).
6. Spraying with 100 μM ABA (prepared by dissolving 26.66 mg of ABA acid in 1 liter distilled water).
7. Spraying with 120 μM ABA (prepared by dissolving 32 mg of ABA acid in 1 liter distilled water).

Total chlorophyll

Total chlorophyll was estimated by placing the SPAD-502 meter on the fifth leaf from the bottom and avoided placing the meter over major leaf veins. Measurements on wheat mid way between the midrib and the leaf margin about 10 cm from the stem.

Nonstructural carbohydrates (NSC)

According to Newell *et al.* (2002), NSC traits were determined by sieving the dried samples (leaves and roots) of the wheat plant separately, soluble sugars were extracted from 0.05 g ground material in 10 ml 80% (v/v) ethanol, the soluble sugar concentration was determined at 490 nm. The starch contained was hydrolyzed to glucose and determined according to calorimetrically at 490 NM. Amyloglucosidase solution, the soluble sugars and starch are referred to as (NSC)

Prodetermination

According to Bates *et al.* (1973), read

spectrophotometrically at 520 nm, using toluene as blank. At 4°C, the reagent is stable for 24 h, a standard curve was used for concentration from 0 - 512 μL (20-100 $\mu\text{g}/\text{ml}$) of L-proline.

Malondialdehyde (mda)

Absorption reads at 532 NM and the amount of nonspecific absorption at 600 NM read and determined.

Enzyme extraction

According to Esfandiari *et al.* (2007), 0.5 g sample was homogenized in ice-cold 0.1 M phosphate buffer (pH = 7.5) containing 0.5 mM EDTA with pestle and mortar. Each of them was centrifuged at 4°C in a Beckman cold centrifuge for fifteen min at 15000 \times g.

Enzyme activity assay

The protein content of the samples was determined using a Bovine albumin method [Bradford (1976)].

Glutathione reductase (gr)

The increase in absorbance at 412 nm recorded at 25°C over an amount of five min on a photomere.

Statistical analysis

Analysis of variance was allotted on the info mistreatment package of SPSS, version 20 and important variations among treatment suggest that were calculated by Duncan's multiple vary check.

3. Results

The results of the ANOVA are summarized in Figs. 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15 and 16, Variants showed significant difference ($p < 0.05$) for different traits indicating a high variability among them. All of these traits were influenced by different levels of salts and ABA acid except for MDA in the root and leaves and in the interaction between (salts \times ABA), indicating that the MDA in the root and leaves were not influenced by these factors. The maximum average of sucrose of root and leaves, NSC of root, starch of leaves and soluble sugar of leaves was observed at the salt concentration 6.73 milimos. cm^{-1} (76.04 cm^2 , 21.07 $\mu\text{mol.g}^{-1}$ DW, 15.14 $\mu\text{mol.g}^{-1}$ DW, 12.35%, 227.71 $\mu\text{mol.g}^{-1}$ DW and 5.86 $\mu\text{mol.g}^{-1}$ DW, respectively, while the increasing of salinity concentration of soil caused

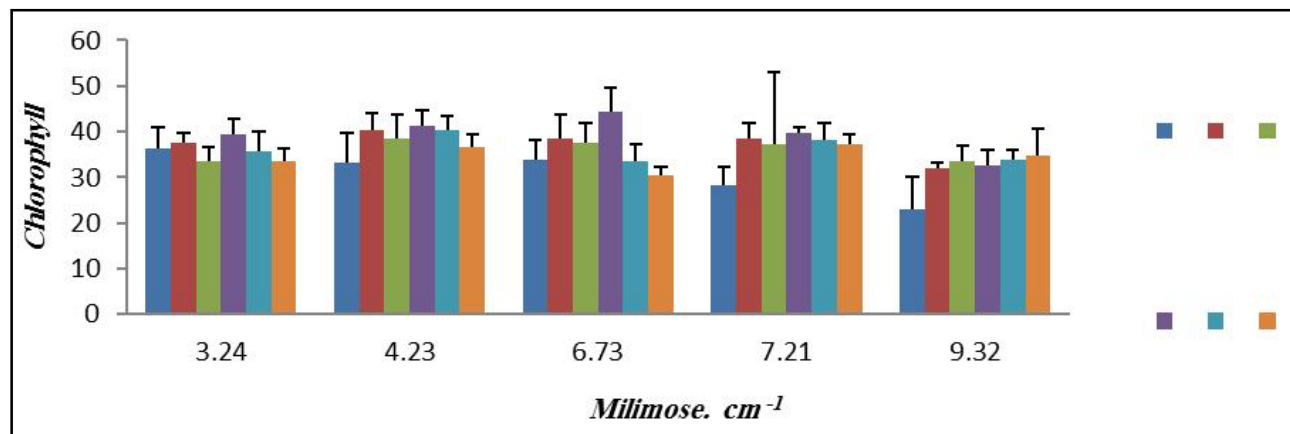


Fig. 1: Chlorophyll content

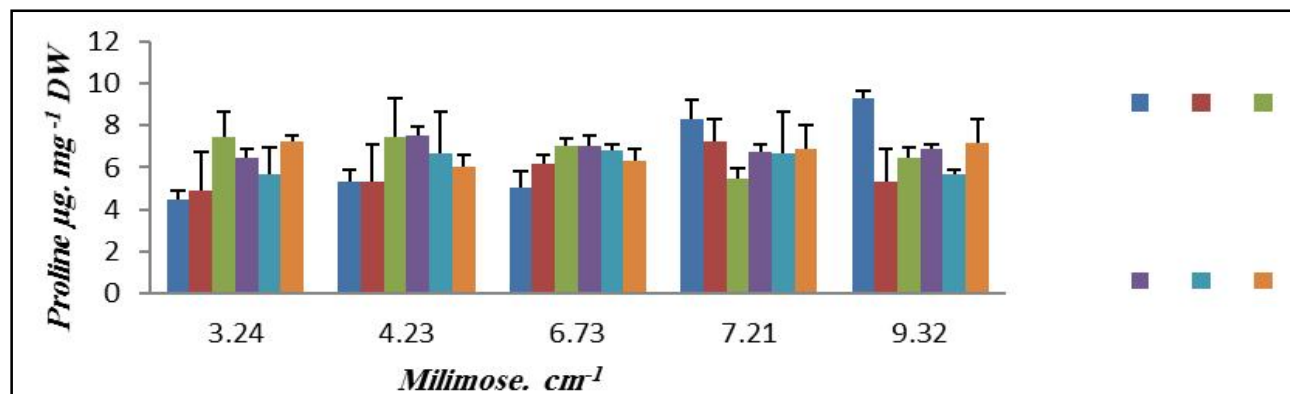


Fig. 2: Proline content

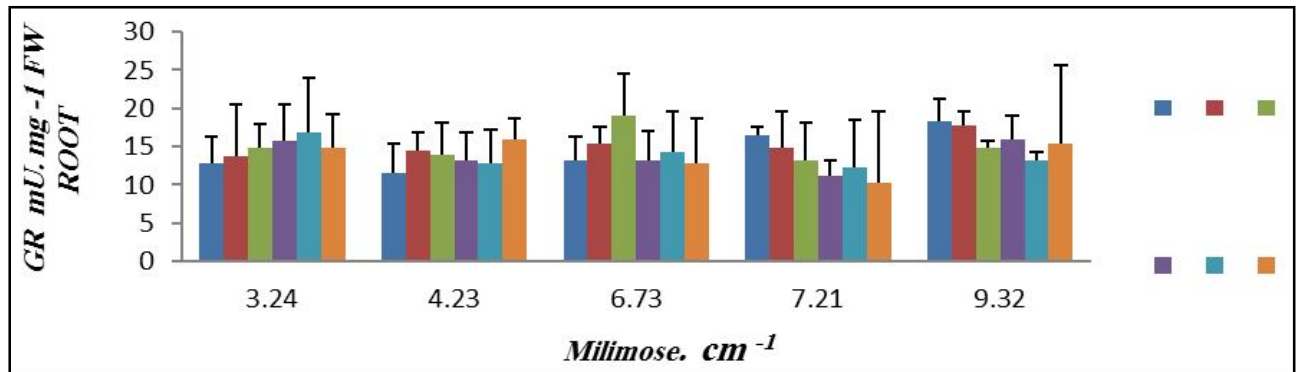


Fig. 3: Glutathione reductase (GR) in the root

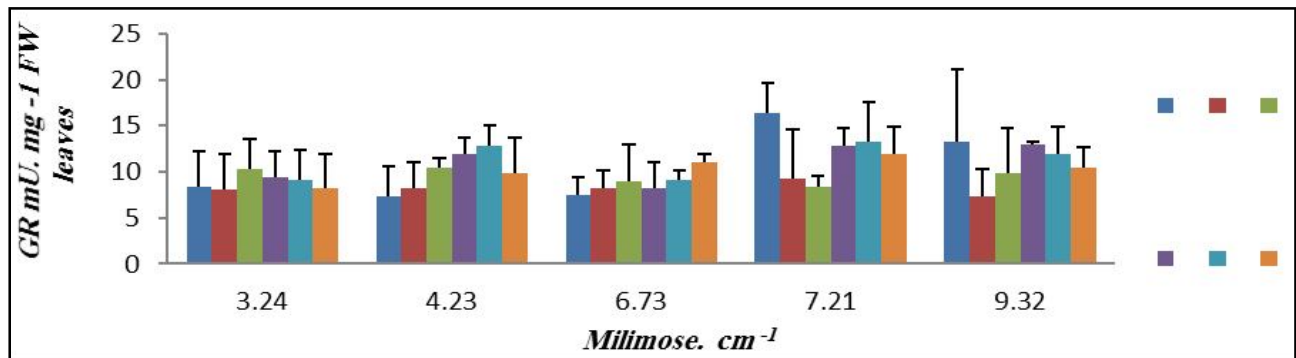


Fig. 4: Glutathione reductase (GR) in the leaves

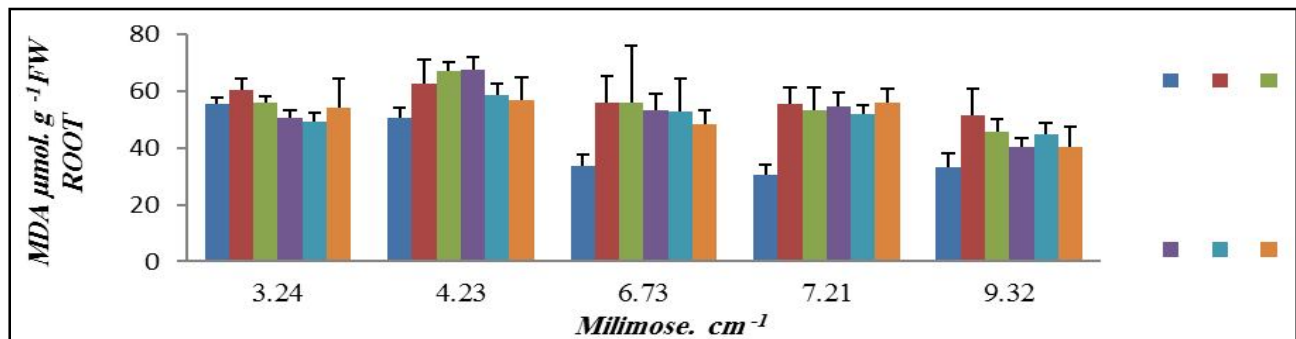


Fig. 5: Malondialdehyde (MDA) in the root

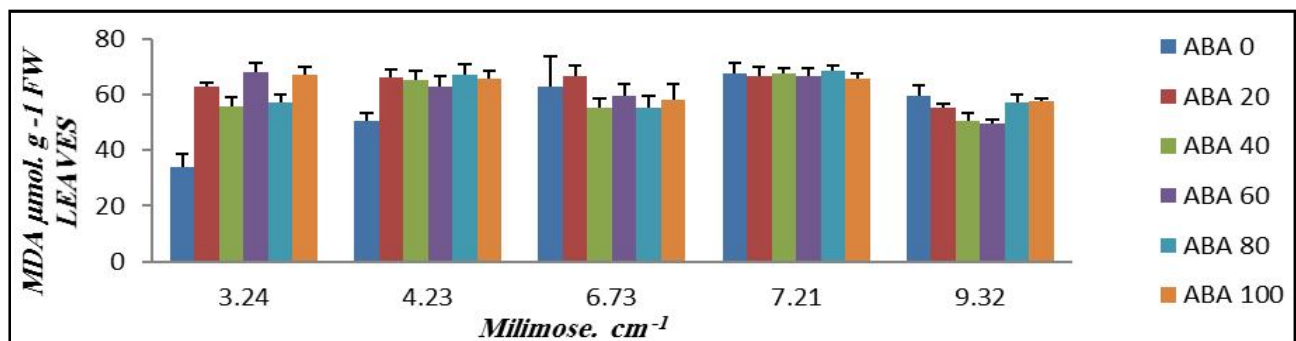


Fig. 6: Malondialdehyde (MDA) in the leaves

decrease in the mains of the traits studied, the high concentration of salt at the level 9.32 milimose. cm⁻¹ recorded the minimum average of chlorophyll, proline, sucrose of root and leaves, glucose of root, starch of root and leaves, soluble sugar of root and leaves, NSC

of root and leaves (58.50 cm², 55.87 cm, 31.59, 6.80, 15.87 mU.Mg-1 FW, 15.39 μmol.g-1 DW, 10.58 μmol.g-1 DW, 13.21 μmol.g-1 DW, 7.60 μmol.g-1 DW, 180.12 μmol.g⁻¹ DW, 3.08%, 5.44%, 8.50% and 6.50%, respectively. The maximum average of glucose of root,

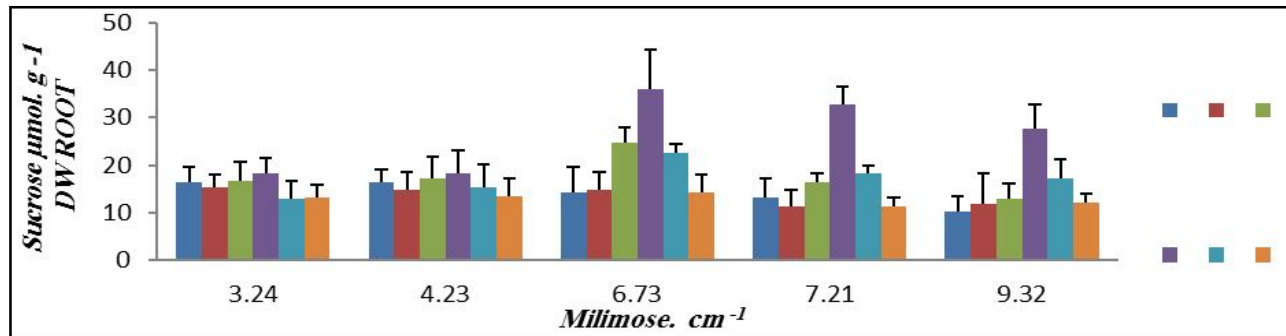


Fig. 7: Sucrose in the root

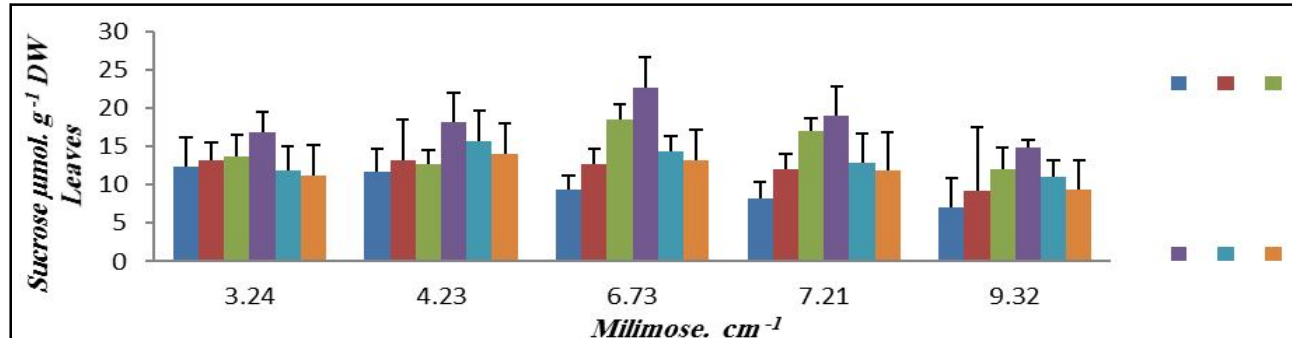


Fig. 8: Sucrose in the leaves

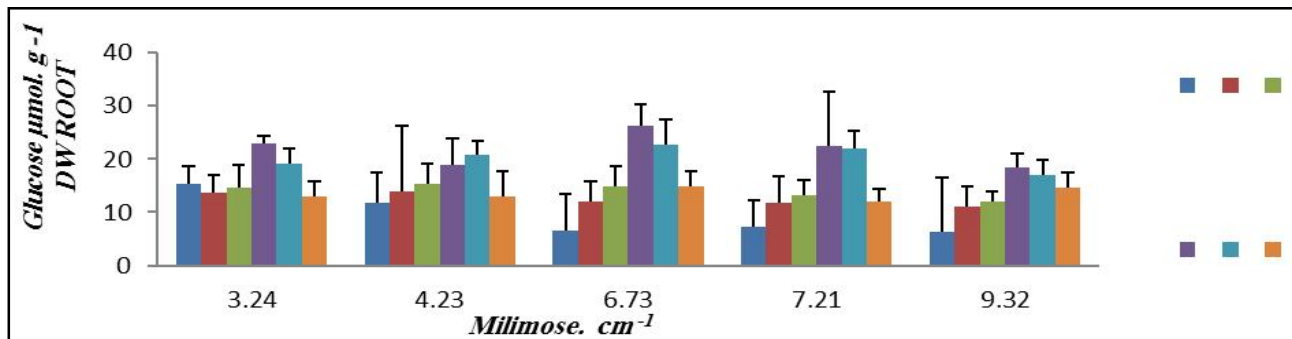


Fig. 9: Glucose in the root

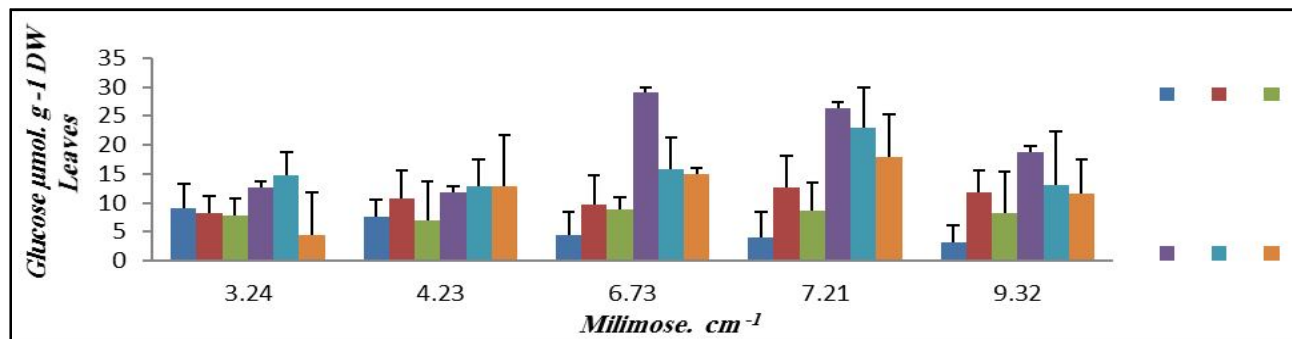


Fig. 10: Glucose in the leaves

starch of root, was observed at the lowest concentration of salts (47.91 cm, 16.49 $\mu\text{mol.g}^{-1}$ DW and 11.13 $\mu\text{mol.g}^{-1}$ DW, respectively. By increasing the concentration of ABA acid lead to the increasing in the average of the most traits at the certain limit 60 micromole.L⁻¹, such as chlorophyll, proline, sucrose of

root and leaves, glucose of root and leaves, starch of leaves, soluble sugar of root and leaves%, NSC of root and leaves and then the rates gradually decreased after that concentration of ABA (76.47 cm², 76.91 cm, 39.48, 6.94 $\mu\text{g.mg}^{-1}$ DW, 26.61 $\mu\text{mol.g}^{-1}$ DW, 18.28 $\mu\text{mol.g}^{-1}$ DW, 21.78 $\mu\text{mol.g}^{-1}$ DW, 19.72 $\mu\text{mol.g}^{-1}$ DW, 247.63%,

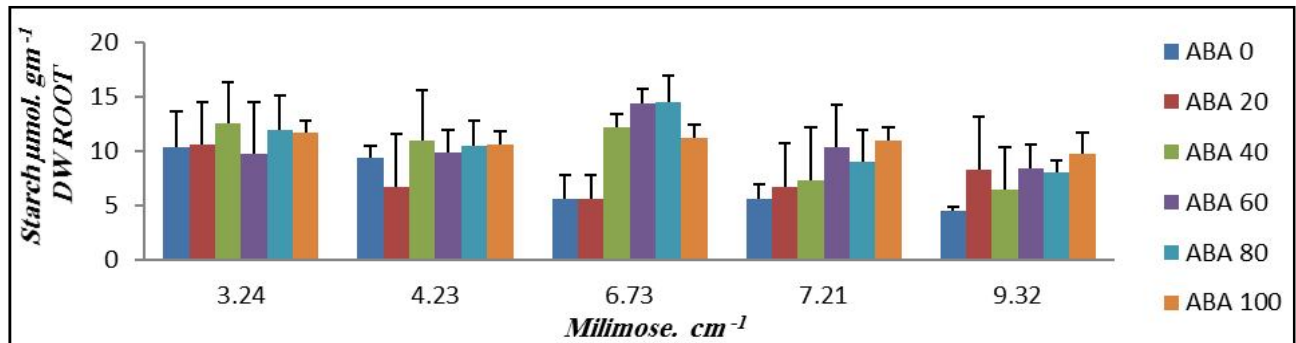


Fig. 11: Starch in the root

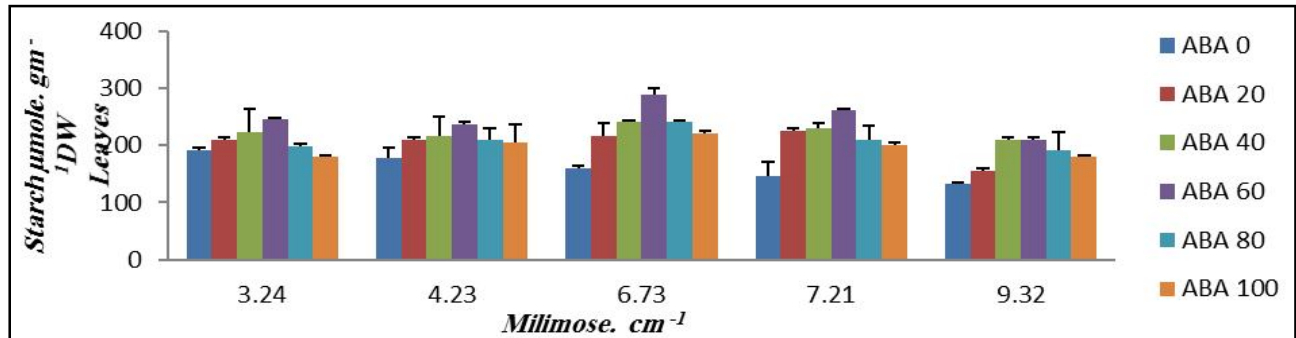


Fig. 12: starch in the leaves

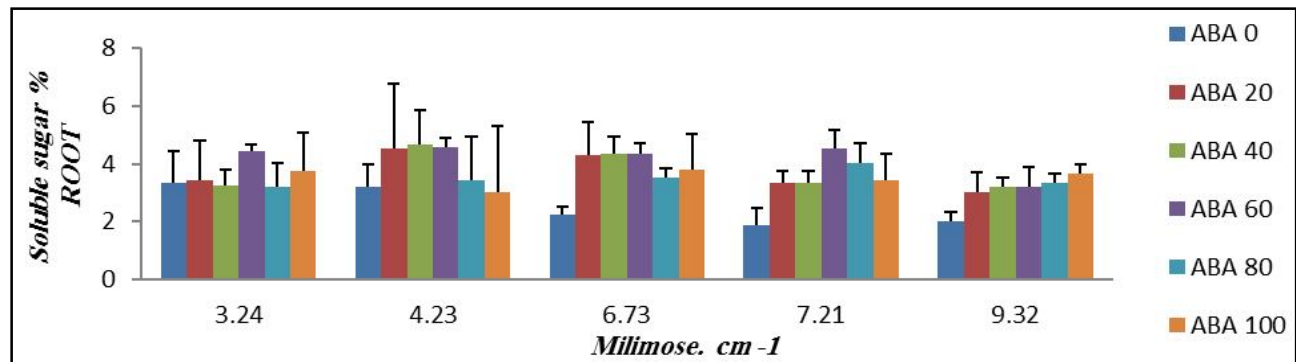


Fig. 13: Soluble sugar% in the root

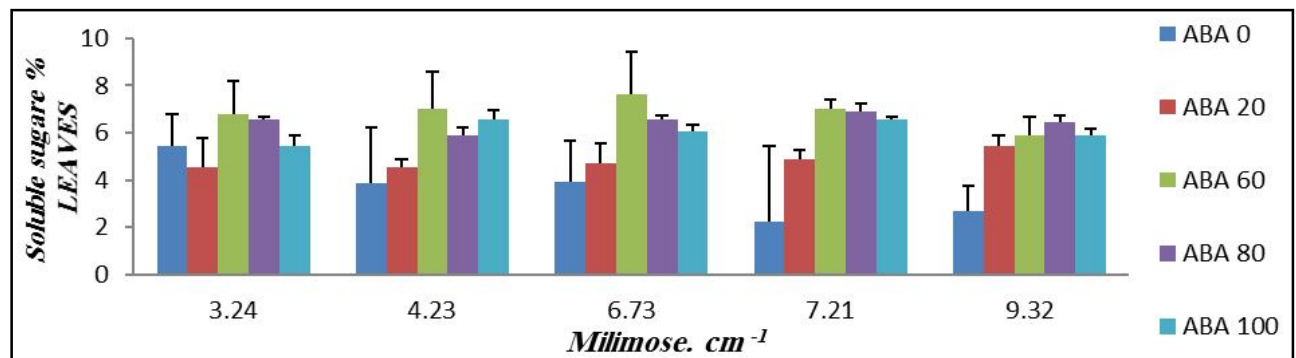


Fig. 14: Soluble sugar% in the leaves

4.22%, 6.87%, 12.85% and 9.76%, respectively. The minimum values of chlorophyll, sucrose of leaves, glucose of root and leaves, starch of root and leaves, soluble sugar of root and leaves, NSC of root and leaves was recorded at the level of (0) ABA (58.90 cm, 30.93, 9.71 µmol.g-1 DW, 9.44 µmol.g-1 DW, 5.66 µmol.g-1

DW, 7.11 µmol.g-1 DW, 161.30 µmol.g-1 DW, 2.53%, 3.62%, 6.93% and 5.81%, respectively. The interaction of (salts×ABA) showed significant differences ($p < 0.5$) for all traits except MDA in root and leaves, indicating that the MDA was not influenced by the factors of this study, the high values of chlorophyll, GR

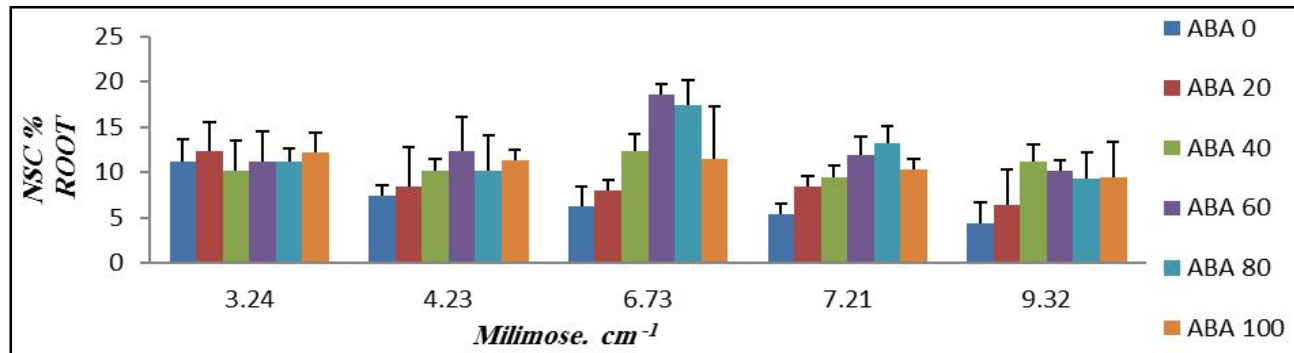


Fig. 15: NSC% in the root

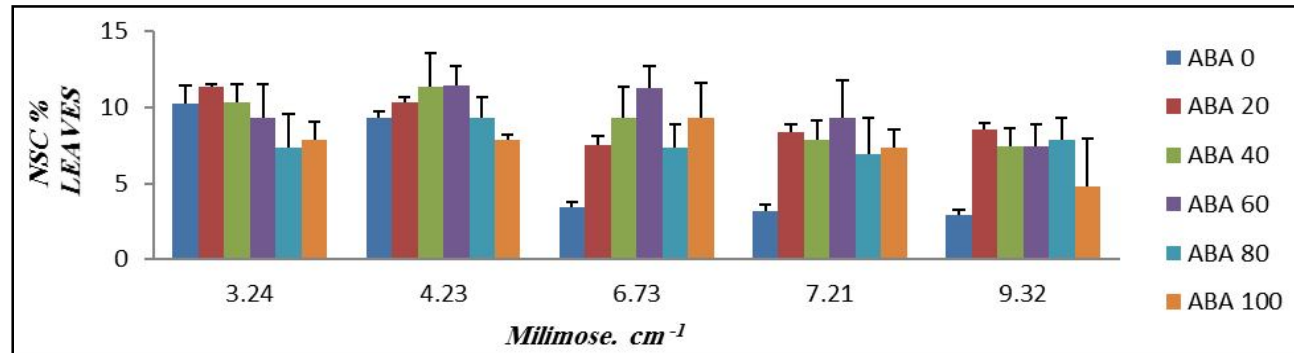


Fig. 16: NSC% in the leaves

of root, sucrose of root and leaves, glucose of root and leaves, starch of root and leaves, NSC of root were observed in the interaction: salt 6.73 milimos. cm⁻¹ × ABA 60 micromole.L⁻¹ (89.13 cm²), 88.54 cm, 44.32, 19.04 mU.Mg⁻¹ FW, 35.87 μmol.g⁻¹ DW, 22.67 μmol.g⁻¹ DW, 26.33 μmol.g⁻¹ DW, 28.97 μmol.g⁻¹ DW, 14.32 μmol.g⁻¹ DW, 288.43 μmol.g⁻¹ DW, 7.65%, respectively, while the lowest values of the most traits was at the interaction (salt 9.32×0 ABA) such as chlorophyll, sucrose of root and leaves, glucose of root and leaves, starch of root and leaves, soluble sugar of root and leaves, NSC of root and leaves (44.87 cm, 22.9, 10.32 μmol.g⁻¹ DW, 7.01 μmol.g⁻¹ DW, 6.21 μmol.g⁻¹ DW, 3.22 μmol.g⁻¹ DW, 4.55 μmol.g⁻¹ DW, 132.98 μmol.g⁻¹ DW, 2.01%, 2.67%, 4.33% and 2.89%), respectively.

4. Discussion

The results of this study indicated that the concentration of salts in the soil has greatly affected the growth rates and we observed a remarkable reduction in leaves area, plant height as well as low rates of chlorophyll and sucrose in the root and leaves of the wheat plant, decrease in the rates of glucose in the roots and leaves as well as, starch in the root and leaves, soluble sugar in the roots and leaves and NSC in the root and leaves. The plants cultivated under the salinity stress are affected through reduced water

potential in system root, toxicity of Na⁺ and Cl⁻ ions, the ions Na⁺ and Cl⁻ causes both hyperionic and hyperosmotic stresses and can lead to reduction of plant growth and then plant death in the advanced stages, the reduction of leaves area, plant height, chlorophyll and the other growth traits in this study may be because of that the increase salt concentration also increase the “osmotic potential” of root zone which prevent the water uptake by any plants. The results are consistent with the findings of Hasegawa *et al.* (2000). The reduction in photosynthetic under the salinity stress is mainly related to a marked inhibition of chemical action and the reduction in water potential in plants. However, the results of this study on the chlorophyll reduction are consistent with the results of Fisarakis *et al.* (2001). In this study, the high chlorophyll rate in the plant was observed at the low salinity levels in the soil, while the chlorophyll rate in the wheat leaves decreased when the salinity rate in the soil increased, our results are consistent with results of Goyal and Verma (2015). They reported that the photosynthesis of plants depends on the salinity concentration which increased gradually to a certain limit, then reduces under high concentration. These results are also consistent with Hasegawa *et al.* (2000). In a recent study, it was observed that there was an increase in glutathione reductase (GR) enzyme on the leaves and roots of the wheat plant as a reaction

to that increase in salt concentration of soil. The results are consistent with the results of Pompella *et al.* (2003). In our current study, it was observed that by increasing the concentration of salts in the soil to a certain extent increased the rate of formation of proline in the plant, whether in the leaves or in the roots and then any increase in the concentration of salts in the soil decreased the formation of proline. This indicates that Proline at certain limits can be a defensive means of plant in the resistance of salt stress and this is consistent with the findings of Varucha *et al.* (2018). The results of recent study with respect to growth traits such as leaves area, plant height, chlorophyll content and other traits such as proline in plant, GR, sucrose, glucose, soluble sugar and NSC, pointed out to increasing in the rate of these traits by increasing the levels of the ABA to a certain extent and then the rates decrease. This indicates that the ABA can increase the resistance of wheat plants to the salt stress of the soil, this was consistent with the result of Babu *et al.* (2012). The significant increase in total soluble carbohydrates must be associated with the over education in photosynthetic rate, so our results are consistent with the findings of Varucha *et al.* (2018) and Sircelj *et al.* (2005).

5. Conclusion

This study revealed several damages promoted by the salts have significant effects on the reduction of plant growth rates in all its forms, as well as the effect of plant production of different compounds such as sugars in the plant and the ABA acid has a significant role in reducing the harmful effects of salts, the increasing concentrations of ABA and to a certain extent can protect the plant and increases its resistance to saline stresses.

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