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Effects of Application Method and Level of Salicylic acid on some Morphological Characteristics of *Ocimum basilicum* L. Leaves under Sodium Chloride Salinity Stress

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ABSTRACT: Salinity is one of the most important agricultural production barriers in arid and semiarid regions which lead to limitations for plants growth and production. Salicylic acid (SA) is a hormone-like substance which causes plant resistance to environmental stresses (heat, coldness, salinity and dryness).To examine interaction between salinity, foliar application and application method of SA on some morphological indexes of Basil (*Ocimum basilicum* L.) leaf, a factorial experiment was done. Factor A included application method of SA, factor B was four different levels of SA and factor C included the levels of salinity. The collected data were analyzed by SAS statistical software. The results showed that application method of SA, its concentration and the concentration of salinity influenced leaf length, width and dry weight. However, for the rest of the indexes the effect was not significant. Also the interaction between application method of SA and salinity concentration had significant effect on the fresh weight of the leaf and the percentage of its dry weight. The interaction between application method of SA and its concentration influenced the number of leaves. Moreover, the simple effect of SA concentration affects the leaf fresh and dry weight was significant.

Key words: Salicylic Acid, salinity, *Ocimum basilicum*, application method.

INTRODUCTION

Aromatic plants represent a renewable source of flavoring substances, which can be used in the food, perfumery and pharmaceutical industries (Gharib, 2006). Basil (*Ocimum basilicum* L.) is one of the important kinds of lamiaceae family which is used as a kind of spice, a medical plant or fresh vegetable (Farzaneh *et al.,* 2011). It has been introduced as a medical plant in most of the pharmacopeias. *Ocimum basilicum* contains essence and is used as a treatment for some diseases and in food, cosmetics and perfume industry. Its essence has antibacterial and antifungal characteristics and controls insects. (Hassani *et al.*, 2004). Salinity is the main osmotic tension which leads to plants growth and production limitations. Depending on its concentration, the performance and growth of the plant decrease. The more the salinity concentration, the more remarkable the reduction in growth will be. Research has shown that salinity results in a decrease in performance and the dry substance of the plant. One of the reasons for this decrease is photosynthesis (Fatemi and Abdolhosseini, 2012). Salicylic acid (SA) called Hydroxybenzoic Acid belongs to a group of Phenol compounds in plants which has an important role in plants growth as a hormone like substance (Kang *et al.*, 1973). It can have a pivotal role in raising the resistance to diseases in plants especially during systemic acquired resistance (Maddah *et al.,* 2007). Research has shown that applying 0.5 mmolL-1 SA has been very effective in reducing the consequences of salinity stress (Shoa and Miri, 2013). SA regulates the expansion, cell division and death and makes a balance between growth and age (Mazaheri Tirani *et al.,* 2008). The production of SA is a reaction to stress. SA, as a group of Phenol compounds is an effective inducer in the expression of genes involved in resistance which encodes the stress related proteins after being added to the external surface of most of the plants (Moradi and Rezvani, 2012).

MATERIAL AND METHODS

This research was conducted in four replications in a completely randomized factorial design in horticultural Science libratory (Islamic Azad University - Miyaneh branch) to study the effects of SA in inducing *Ocimum basilicum* resistance against salinity stress in pot condition. SA treatment was done one day before the treatment of salinity and early in the morning. Factor A included application method of SA (foliar application and irrigation), factor B the treatment of SA in four levels (0, 0.1, 0.5 and 1 $mmolL^{-1}$) and factor C the treatment of Sodium Chloride (NaCl) in four levels (0, 40, 80 and 120 $mmolL^{-1}$).

These were applied on the plants which had 6 to 8 leaves by irrigation or foliar application (Esfinifarahani *et al.,* 2013). In the end the collected data were analyzed by SAS (ver. 9.1) software and the averages were compared by Duncan's multiple range test method at the level of 0.05 (P< 0.05).

RESULTS

The triple interaction of SA application method, its concentration and the concentration of NaCl was significant on leaf length and width (P< 0.05) and on leaf dry weight (P< 0.01). So was significant the interaction between SA application method and concentration of NaCl on leaf fresh and dry weight (P< 0.05). The interaction effect of SA application method and its concentration on the leaf number and leaf dry weight (P< 0.05) and on leaf dry weight (P< 0.01) was significant. The simple effect of the concentration of SA on leaf fresh weight and leaf dry weight (P< 0.01) was significant. The simple effect of salinity on longest leaf length and leaf fresh weight in (P< 0.05) was significant. Simple effect of application method on leaf dry weight (P<0.05) was significant.

A. Leaf dry weight

The results showed that the effect of SA concentration on leaf dry weight was significant at 0.01(P< 0.01) (Fig. 1). Comparing SA concentration shows a decrease at 0.1 mmolL^{-1} which does not confirm the findings of other researchers. But the difference between first and second level in percent dry weight of the leaf is significant which shows an increase confirming the findings of previous studies. Using SA shows even more increase which is important for more expansion of the surface of the leaf and the increase of fresh and dry weight in colza leaves (Miar Sadeghi *et al.*, 2008).

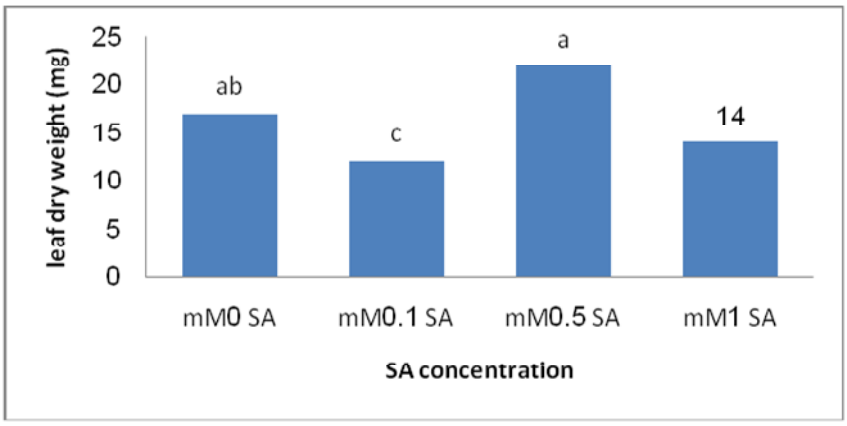

Fig.1. Effect of SA concentration on leaf dry weight.

B. Leaf fresh weight

The results showed that the effect of SA concentration on leaf fresh weight is significant at 0.01level (P< 0.01), but the rest is not. Salicylic Acid shows the largest average of wet weight of the leaf in the concentration of 0.1 mmolL^{-1} and the smallest average belongs to the concentration of 0.5 mmolL^{-1} and shows a decrease compared to optimum treatment (Fig. 2). These findings do not confirm the findings of other researchers.

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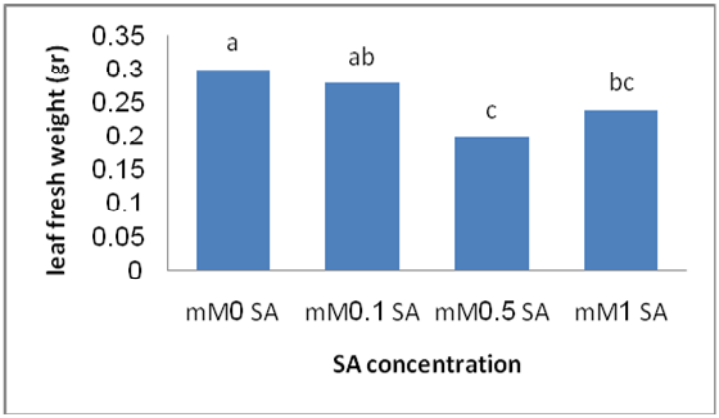

Fig. 2. Effect of SA concentration on leaf fresh weight.

C. Leaf number

The results showed that interactions between application method and concentration of SA on the number of leaves are significant in (P< 0.05). Comparing the averages shows that the biggest average of number of leaves belong to irrigation concentration of 0 mmolL^{-1} SA and the smallest one to foliar application concentration of 0 mmolL^{-1} SA (Fig. 3).

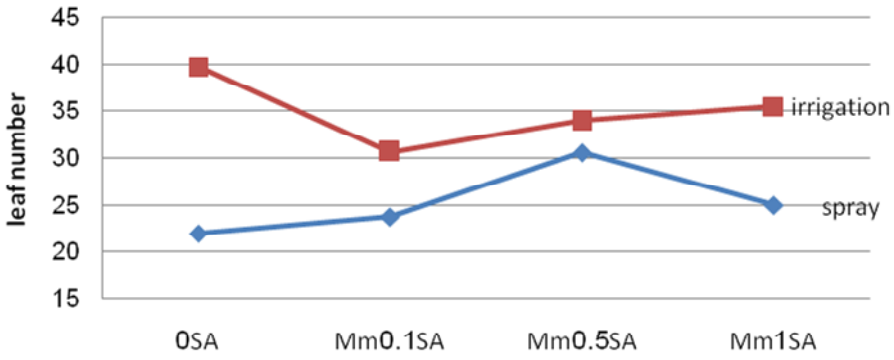

Fig. 3. Effect of SA concentration on leaf number.

D. Wet weight of the leaf

The results showed that the interactions between application method of SA and NaCl concentration on leaf fresh weight are significant at 0.05 levels (P < 0.05). Comparing the averages shows that there is a significant difference in the concentration of 40 mmolL^{-1} so that leaf fresh weight was more in foliar application than irrigation (Fig. 4). It is probably because in foliar application the surface of the leaf is affected directly and shows a more weight compared to irrigation.

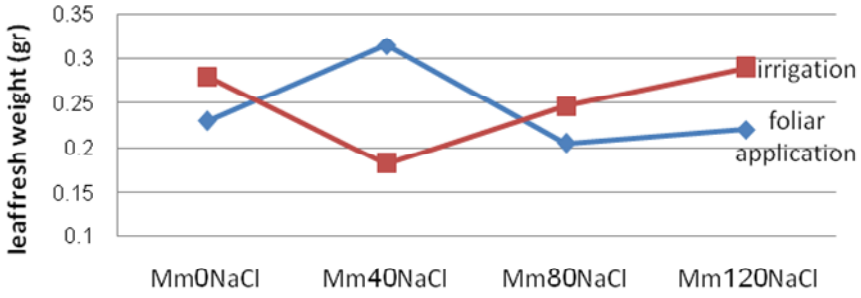

Fig. 4. Effect of interaction between application method of SA and NaCl concentration on leaf fresh weight.

E. Leaf dry weight

The results showed that interactions between application method of SA and NaCl concentration on leaf dry weight are significant in NaCl 80 mmolL^{-1}, there is a significant difference in the application method so that leaf dry weight is more in foliar application than irrigation (Fig. 5).

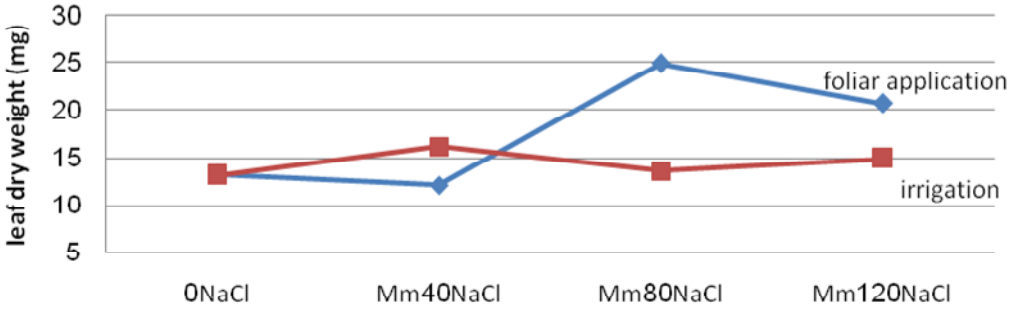

Fig. 5. Effect of interaction between application method of SA and NaCl concentration on leaf dry weight.

F. Leaf width

The results showed that the effect of triple interaction of SA application method, SA concentration and NaCl concentration are significant (P< 0.01). Comparison of means showed that using SA in irrigation, SA with 0 $mmolL^{-1}$ concentrations, and Sodium Chloride with 0 $mmolL^{-1}$ concentrations had the largest mean and consumption of SA in foliar application, SA with 1 $mmolL^{-1}$ concentration and NaCl with 120 $mmolL^{-1}$ concentrations had the smallest mean on width of the leaf (Fig. 6).

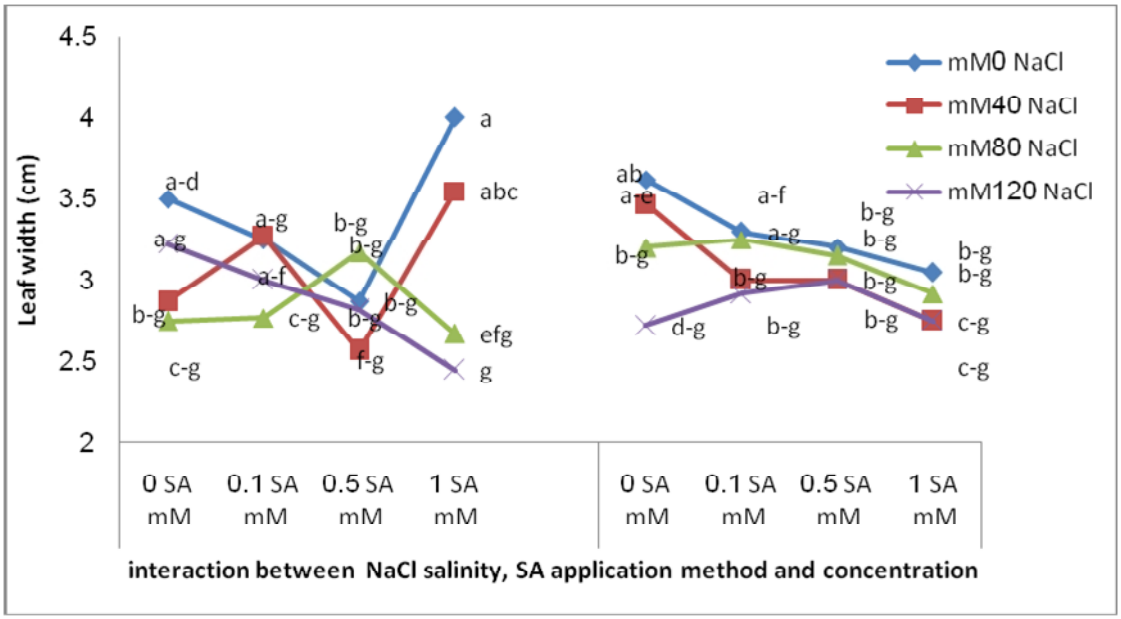

Fig. 6. Effect of interaction between application method of SA, SA concentration and NaCl concentration on leaf. Width.

G. Leaf length

The results showed that the effect of triple interaction of SA application method, SA concentration and NaCl concentration was significant (P< 0.05). Comparison of the means showed that consumption of SA in foliar application, SA with 1 $mmolL^{-1}$, and NaCl with 0 $mmolL^{-1}$ had the heights mean and using SA in foliar application, SA 1 $mmolL^{-1}$ and NaCl 80 $mmolL^{-1}$ had the lowest mean on length of the leaf (Fig. 7).

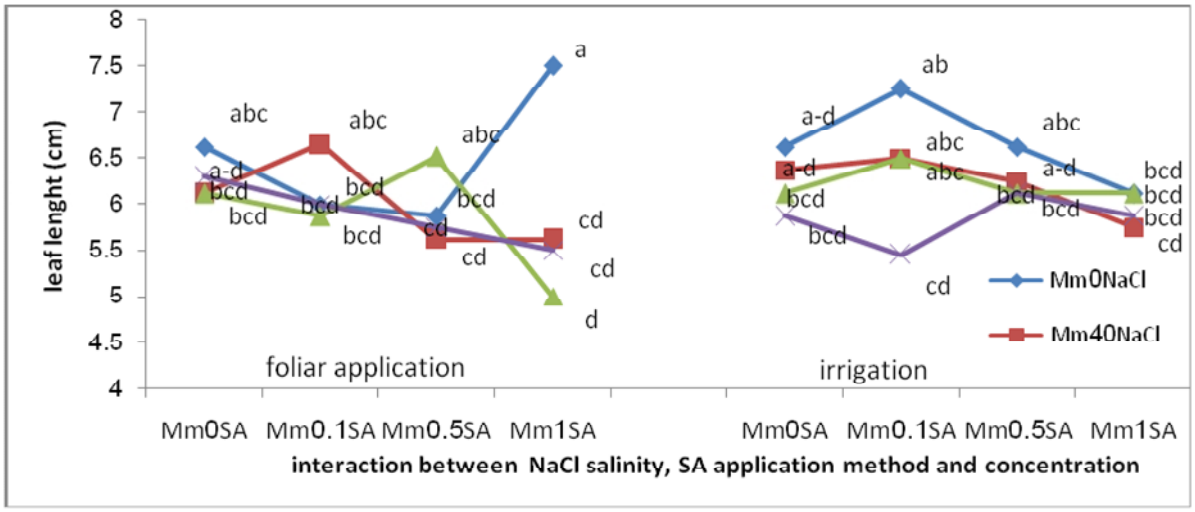

Fig. 7. Effect of interaction between application method of SA, SA concentration and NaCl concentration on leaf length.

H. Leaf dry weight

The results showed that the effect of triple interaction of SA application method, SA concentration and NaCl concentration was significant (P< 0.01). Comparison of the means showed that using SA in foliar application, SA 0 mmolL^{-1}, and NaCl 80 mmolL^{-1} had the heights mean and using SA in irrigation, SA with 1 mmolL^{-1}, NaCl 80 mmolL^{-1} had the lowest mean on leaf dry weight (Fig. 8).

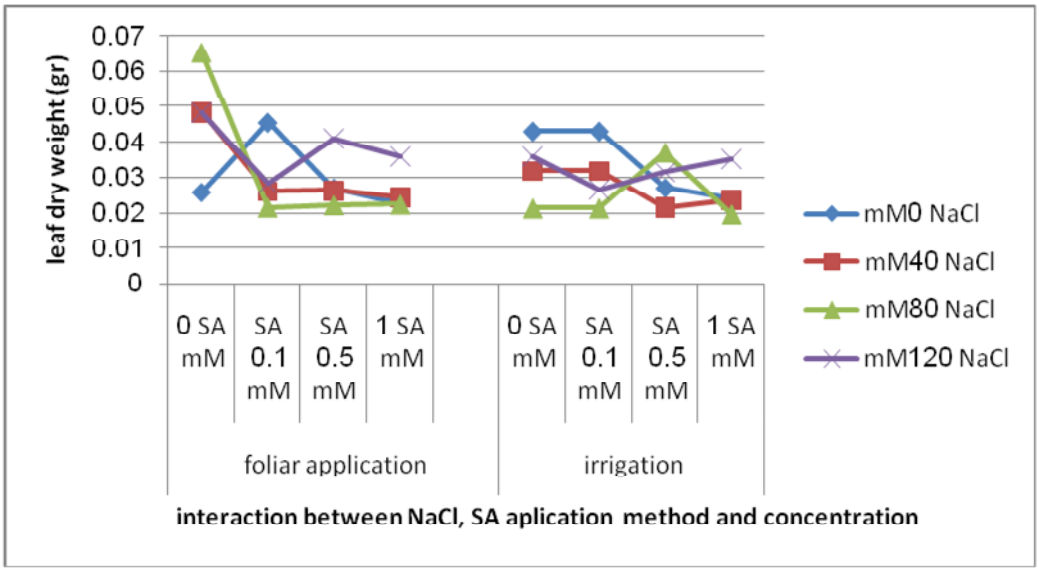

Fig. 8. Effect of interaction between application method of SA, SA concentration and NaCl concentration on leaf dry weight.

DISCUSSION

Currently environmental stresses are one of the main determining factors in distributional patterns of the products in the world. It is estimated that only 10 percent of the cultivatable lands of the world may be without stress. Salinity stress will be effective on the physiological processes of the plant from the level of germination until its complete evolution (Dolatabadian *et al.*, 2009). The production of SA is a reaction to stress. SA, as a group of Phenol compounds is an effective inducer in the expression of genes involved in resistance which encodes the stress related proteins after being added to the external surface of most of the plants (Moradi and Rezvani, 2012).The functional mechanism of SA against stress is related to its role in regulating antioxidant enzymes and other compounds with active oxygen in the plant. An experiment on the effect of salinity on corn showed that under the salinity stress the activity of peroxidase enzyme in Bordbar species increases. Catalases and peroxidases are the enzymes which have an important role in responding to non-biological stresses like salinity. Peroxidases are responsible for removing extra quantities of peroxide hydrogen. And SA leads to the production or activation of these antioxidants directly or indirectly (Nourafcan *et al.*, 2012).

Also they studied the effects of SA on performance and ingredients of green Cuminum. The result showed that treatment through foliar application had a better effect than irrigation (Esfinifarahani *et al.*, 2013). Maddah *et al.* (2007) believe that spraying SA is more effective on plants than irrigation. A research showed that using SA can have a positive effect on leaf length of *Aloe vera*, concentration of its gel, and fresh weight of its branches (Abdollahi, 2011). At the end with regard to the above mentioned results, it can be said that *Ocimum basilicum* L. is generally sensitive to salinity, because most of physiological indices in this experiment have changed in line with resistance mechanism and a decrease in growth has been observed. Applying SA to confront the salinity stress could not have a significant effect. Also using SA solved in water showed a better effect than irrigation.

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