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The Effects of Salinity and Methyl Jasmonate on Some Morphological Traits of Chamomile (*Matricaria chamomilla*)

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ABSTRACT

Salinity stress is among the major barriers to plant production in many parts of the world, especially in arid and semi-arid areas. Jasmonates are one of the newest plant growth regulators that reduce the damages caused by environmental stresses. In this study, the effects of salinity and methyl jasmonate on some morphological traits of chamomile including root and shoot lengths, fresh weights and dry weights were evaluated. Seeds were planted in green house in pots as a completely randomized design with three pots as replications for each treatment. Plants were treated with 100 and 200 mM concentrations of salt at the four-leaf phase for six days alternately. Methyl jasmonate was sprayed on plants at concentrations of 0.01-0.1 µm/l along with salinity treatments which was continued for 6 hours. Salinity and methyl jasmonate reduced stem length. Salinity reduced dry and fresh weights in proportion to control group. But methyl jasmonate improved the effects of salinity. Growth study of root and shoot in seedlings showed salinity effects and reduction in weights. Increased dry and fresh weights of treated plants in proportion to control group showed the protective effect of methyl jasmonate**.**

Keywords: Chamomile, Salinity, Methyl jasmonate, Morphological traits

INTRODUCTION

Salinity stress is among the major barriers to plant production in many parts of the world, especially in arid and semi-arid areas. Man has been involved in salinity problem for thousands of years.

Jasmonates are one of the newest plant growth regulators that reduce damages caused by environmental stresses. Jasmonic acid, pentylcyclopentan-1-acetic acid and its methyl ester (methyl jasmonate) are derived compounds from cyclopentan linolenic acid with a structure similar to eicosanoids in mammals (Creelman and mullet, 1995). They are generally known as jasmonates with an important role in regulating development and morphogenesis processes. Jasmonic acid methyl ester was reported as an aromatic substance of *Jasmonium grandiflorum* oil in Demole *et al.* study in 1962 (Shi and Sheng, 2005). Jasmonates were identified in 206 plant species with 150 families including ferns, mosses and fungi which show their global spread. Jasmonic acid is a linolenic acid derived from cyclopentanol with a regulatory role in development and responses of plants to environmental stresses like mechanical injuries or chemical diseases (pathogenic attacks). The function of jasmonate in regulation of plant growth and reactions to stress often needs a promotion of inner level by an artificial compound or de novo synthesis. The synthesis of jasmonate in response to chemical diseases has been studied well (Creelman and mullet, 1995).

Jasmonic acid is produced as a signaling molecule in response to external stimulators such as wounding, mechanical force, pathogen attack and osmotic stress (Molina *et al.,* 2002). For example, at the wound site, cysteine with 18 amino acids is transported by vessels to different parts of plant and causes induction of jasmonic acid via polypeptides hydrolysis.

The genes responsible for Jasmonate acid production are expressed in apical part of plant. Increase in jasmonic acid controls the biosynthesis of compounds such as proline and putrescine in environmental stresses and induces the responsible genes of these compounds. In fungal attacks, jasmonate activities encode the proteases, and reduce the damages. Furthermore, it adjusts the proteins of wall such as PRP which may be necessary for barrier synthesis against infection (Gao *et al.,* 2004).

Chamomile is one of the therapeutic annual plants growing highly in the southern areas of Iran with many applications in industry. This plant shows an anti-inflation property because of extant Kamazolen. Also, it is a tranquilizer. Chamomile can store 10 mg of NaCl per 1 gram of tissues. In many reports, it is known as a halophyte plant.

Chamomile is grown in hot and arid areas and is widely cultivated because of its therapeutic essential oil. Considering that salinity stress as an agricultural problem in these geographical areas, researches on the mechanisms of plant resistance with lower cost and higher performance are important. Therefore, this study was performed to investigate the effects of methyl jasmonate on traits of chemical and biochemical growth of chamomile in salinity under green-house condition.

MATERIALS AND METHOD

Chamomile seeds were prepared from Isfahan Research Center of Agriculture and Natural Resources, Iran (2013).

Cultivation and Treatments

Seeds were planted in pots (15 cm diameter) filled with vermiculite. They were intact, equal in size, and without diseases or damages. Seeds were planted in pots in green house in a completely randomized design with three pots as replications for each treatment. Pots were irrigated using distilled water daily and treated with Hoagland solution (half concentration) weekly.

When the seedlings reached to four-leaf phase (the appearance of the fifth leaf), treatments were started. Methyl jasmonate concentrations were prepared at concentrations of 0.01-0.1µm/l and the leaves were sprayed with them for six days, alternately. After that, for enforcing the salinity stress, 100 and 200 mM concentrations of salt were used in irrigation water for six days, alternately. Alone salinity treatment was performed for six days, alternately. Environment temperature was 16 °C and 16:8 hours of photoperiod was enforced.

Seedlings Culture and Treatment

After separating intact and equal-sized seeds, 25 seeds were placed in petri dishes furnished with filter paper. For each treatment, four petri dishes were considered as 4 repeats. For salinity treatments, NaCl solution was added into petri dishes at concentrations of 50, 100 and 200 mM. For methyl jasmonate treatments, the seeds were transferred into petri dishes after remaining in their different concentrations of 0.01-0.1 µm/l for 6 hours. For dual treatments, the seeds were transferred into petri dishes after remaining in the mentioned concentrations of methyl jasmonate and irrigation with mentioned concentrations of NaCl.

Different traits including dry and fresh weights of root, aerial parts of plant and seedlings as well as the length of aerial parts, root and seedlings were measured.

Statistical Analysis

In each stage of experiment, each treatment was repeated three times. In each repeat, six plants were considered. Obtained data were analyzed using SAS program-version 9.0 (complete randomized design) and LSD test at 5% probability level and graphs were drawn using Excel program.

RESULTS

Root Length

The effect of methyl jasmonate on root length was significant and both concentrations reduced it in relative to control group. Salinity affected this trait also and the longest roots were belonged to concentration of 100 mM and 200 mM treatment. Assessing the interaction of salinity and methyl jasmonate showed that the different concentrations of methyl jasmonate reduced roots length in both concentrations of NaCl. Figure 1 shows mean comparison results of root length ($p<0.05$).

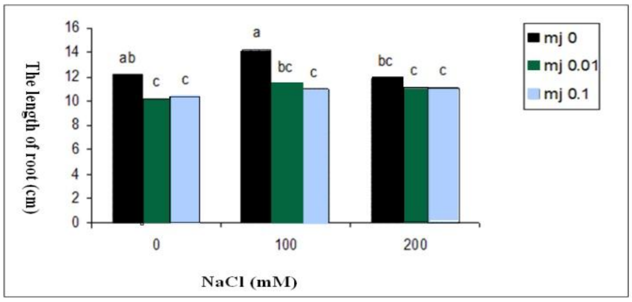

Figure 1. Interaction of methyl jasmonate and salinity stress on root length

Stem Length

The measurements of stem length showed that different concentrations of salinity and methyl jasmonate affected this trait and decreased it significantly ($p<0.05$). High concentrations of both treatments produced shorter stems. The interaction of salinity and methyl jasmonate caused significant decreases in stem length and the least length of stem was related to 200 mM of salinity and 0.1 µM/l of methyl jasmonate, whereas control group produced the highest stem length.

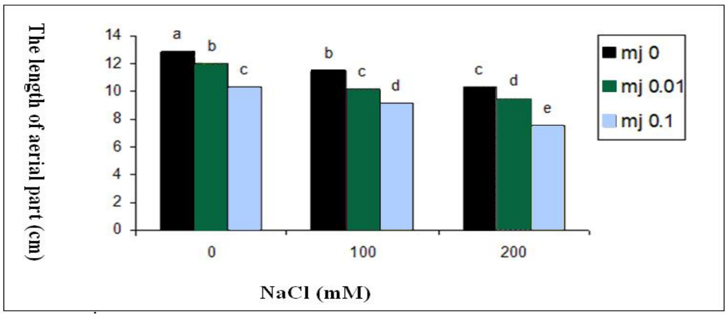

Figure 2. Interaction of methyl jasmonate and salinity stress on stem length

Aerial Parts Fresh Weight

Methyl jasmonate at 0.01 µM/l level showed the highest fresh weight of aerial parts whereas the other concentrations of salinity and methyl jasmonate showed lower fresh weights (Figure 3). Interaction between salinity and methyl jasmonate did not have positive effect in improvement of environmental stress ($p< 0.05$).

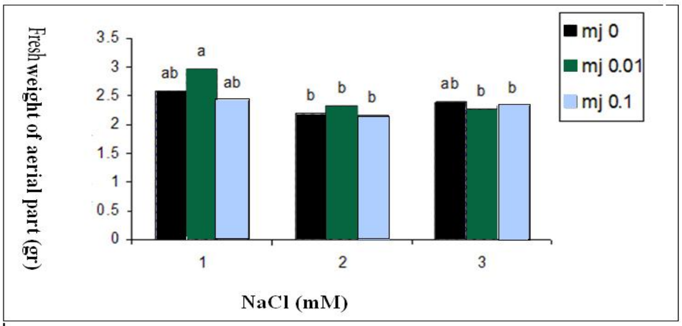

Figure 3. Interaction of methyl jasmonate and salinity stress on fresh weight of aerial Parts

Root Fresh Weight

Fresh weight of root was affected by methyl jasmonate spraying. The least fresh weight was obtained in 0.1 µM/l of methyl jasmonate but 0.01 µM/l treatment increased fresh weight in proportion to the control group. Salinity showed a significant effect on fresh weight of root and fresh weight was decreased by increasing NaCl amount. Interaction of salinity and methyl jasmonate showed significant effect on root fresh weight ($p<0.05$) so that 0.1 µM/l level of methyl jasmonate showed an increased fresh weight interaction which indicates an improvement in salinity stress effects.

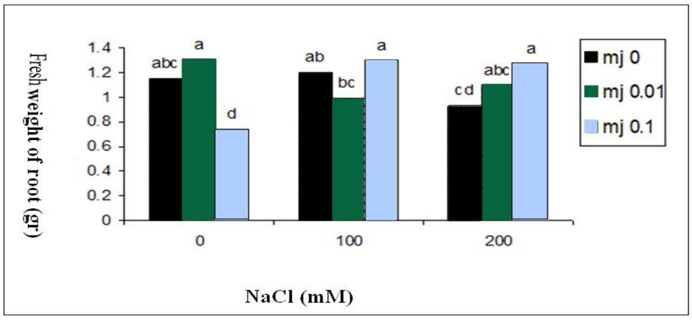

Figure 4. Interaction of methyl jasmonate and salinity stress on root fresh weight

Stemlet Length

Methyl jasmonate in both levels caused a significant decrease is stemlet in proportion to control group ($p<0.05$). Salinity also decreased this trait significantly ($p<0.05$) and the least length of stemlet was related to 200 mM treatment. Interaction of salinity and methyl jasmonate was significant ($p<0.01$) and methyl jasmonate could improve the negative effects of salinity stress. The highest improvement was observed in 0.1 µM/l level. The tallest stemlets were observed in concentration of 50 mM/l of salinity and 0.1 µM/l of methyl jasmonate.

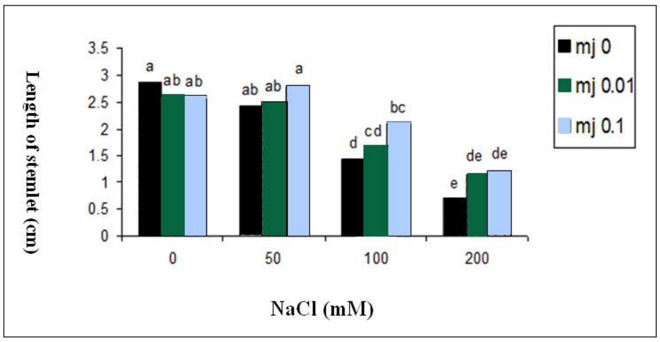

Figure 5. Interaction of methyl jasmonate and salinity stress on length of stemlet

Rootlet Length

Methyl jasmonate caused no significant differences in length of rootlet ($p<0.01$) but salinity reduced it and the length of rootlet was decreased by increasing NaCl concentration ($p<0.05$) so that the least length was observed in the highest salinity. Interaction of salinity and methyl jasmonate produced the longest rootlets in combination of 50 mM salinity and 0.1 µM/l of methyl jasmonate. Methyl jasmonate was really effective and 0.1 µM/l treatment improved this trait (Figure 6).

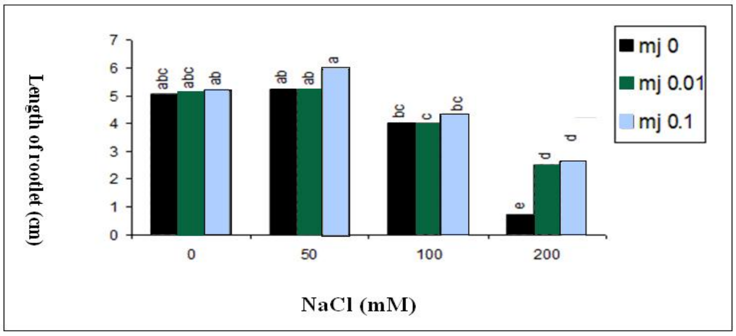

Figure 6. Interaction of methyl jasmonate and salinity stress on length of rootlet

Seedlings Fresh Weight

Salinity and methyl jasmonate reduced fresh weight of seedlings in proportion to the control ($p<0.05$). The least fresh weight of seedlings was related to 200 mM of salinity whereas control treatment had the highest fresh weight. Interaction of salinity and methyl jasmonate was also significant and the highest fresh weight was belonged to 50 mM of salinity and 0.1 µM/l of methyl jasmonate. Application of methyl jasmonate was effective and improved the stress of high salinity (Figure 7).

Seedlings Dry Weight

According to results, methyl jasmonate did not affect seedlings dry weight. Salinity increased this trait relative to control group ($p<0.05$) and the highest dry weight was obtained in 100 mM of salinity. Interaction of salinity and methyl jasmonate affected this trait and applying of 0.1 µM/l of methyl jasmonate increase dry weight in high salinity (Figure 8).

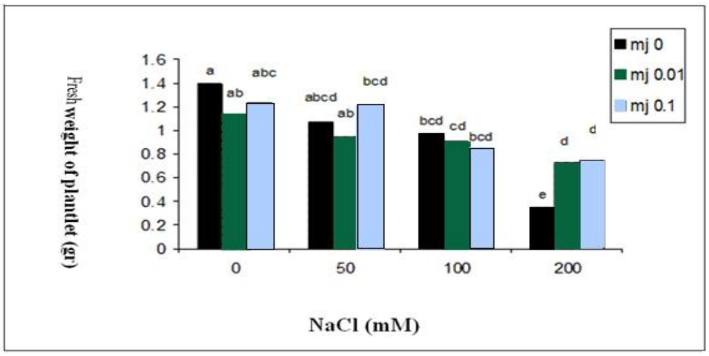

Figure 7. Interaction of methyl jasmonate and salinity stress on fresh weight of seedlings

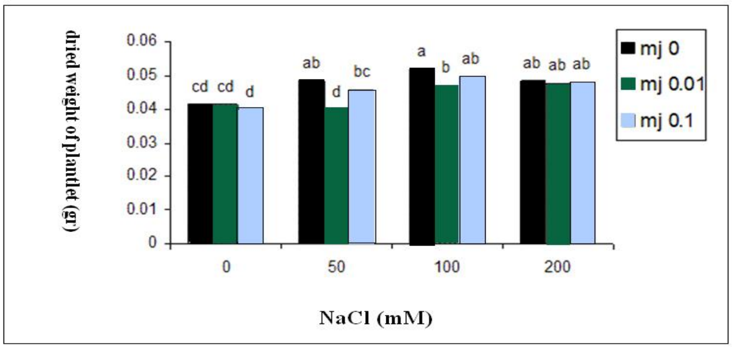

Figure 8. Interaction of methyl jasmonate and salinity stress on dry weight of seedlings

DISCUSSION

Growth is the result of increase in cell division and irreversible increase in cell volume. Abiotic stresses affect cell division and reduce the growth and development (Taiz and Zeiger, 2007).

High salinity causes imbalance of ions in plant cells, oxidative stresses and reduced plant growth (Kepczynski and Bialecka, 1994). Plants can respond to salinity stress via some changes or modifications in cellular processes and morphological structure in order to reduce ion toxicity and keep balance between production and loss of oxygen radicals. In salinity stress, the osmotic pressure is reduced and this decreased turgor is an important factor in growth inhibition so that it stops elongation and division of cells (Poustini, 1995).

Following the salinity stress, chlorophyll content, growth of root, dry weights of stem and root were reduced in rapeseed (Steppuh *et al.*, 2001).

In this study, salinity affected the growth of root and stem of seedlings and reduced their weights. Salinity decreased the growth of rootlet and stemlet and germination in wheat and the reduction was more in higher salinities (Poustini, 2005).

It was found in a study that germination of eggplant was delayed in concentrations of more than 50 mM of NaCl but the final percentage of germination was not reduced. The germination percentage was significantly reduced only in the highest salinity level (100-150 mM). Increasing salinity up to 100-150 mM reduced root length to 36-47% and the emergence of seedlings was delayed in high salinity treatments (Chartzoulakis and Loupassaki, 1997). Seedlings phase is more sensitive in eggplant than germination phase. Similar results were observed in pepper (Chartzoulakis and Klapaki, 2000).

Reduction in dry weights of stems and roots in barely, sugar beet, corn, wheat and also reduction in root dry weight of safflower have been reported (Kaya and Ipek, 2003).

In current study, salinity reduced the fresh weights of stem and root. Both salinity and methyl jasmonate reduced the length of stem. Reduced length of stem in salinity treatment can be due to decreased turgor pressure and cell elongation.

Also, reduced growth of stem by methyl jasmonate is due to its effect on stomata cells. It can change the properties of guard cells and causes stomatal closure and thereby control the photosynthesis. The obtained results about the reducing effects of 0.1 µM/l methyl jasmonate on stem growth are similar to results of Yeh *et al.* (1995) in rice and Lynch and Lauchli (1998) in *Pharbitis nil* and pepper seedlings.

It has been reported that jasmonate inhibits the elongation of coleoptiles by auxin via blocking the formation of glucose in cell wall polysaccharides. Generally, salinity causes significant decrease in the length of root. Similar results have been reported in two susceptible and resistant cultivars of rice and cowpea (Creelman and Mullet, 1997).

Munnz (2002) stated that salinity tolerant plants showed some transmissions of Na and Cl into leaves and have the ability to separate these ions in vacuoles to prevent their accumulation in cytoplasm or cell walls in order to avoid ion toxicity.

Methyl jasmonate decreased the length of root in treated plants. It has been reported that concentration of 0.1 μM/l of methyl jasmonate caused a reduction in the length of stem and root in *Pharbitis nil* (Maciejewska and kopcewicz, 2002). There are few reports about increasing effect of methyl jasmonate on root length. Treatment of methyl jasmonate in 7-10 μM/l increased root length and probable reason is reformation of root meristems (Maciejewska and kopcewicz, 2002).

The increase in concentration of methyl jasmonate reduced fresh and dry weights of plant aerial parts. Salinity stress decreased dry weights of stem and roots in barely and sugar beet (El Tayeb, 2005) and dry weight of root in safflower (Kaya and Ipek, 2003). In another report, salinity treatments reduced fresh and dry weights of stems and roots in sugar beet (Ghoulam and Fares, 2001). In this study, salinity reduced fresh and dry weights of treated plants relative to the control treatment, but the application of methyl jasmonate improved the effect of salinity. Increased dry and fresh weights of treated plants in relative to control treatment showed the protective effect of methyl jasmonate.

CONCLUSION

Considering that salinity is one of the major obstacles in crop production of arid and semi-arid regions and encountering salinity requires a lot of expenses and time, researches on the mechanisms of plant resistance with lower cost and higher performance are important. Jasmonates as plant growth regulators are really important in biochemical and physiological processes. Methyl jasmonate in deenite concentration and time decreased damages of salinity at germination stage.

In oxidative stress, one way to reduce the osmotic potential is to increase osmotic agents. Osmotic adjustment in chamomile under salinity stress by soluble salts such as sugar and proline confirms this subject. Methyl jasmonate protects macromolecules and cell membrane via increasing sugar and proteins. In present study, increase in malondialdehyde levels indicates the production of free radicals in salinity stress and shows that production of these radicals caused membrane injuries. Salinity could limit plant growth through water shortages, imbalances of ions, and reducing photo-assimilates. On the other hand, plant tried to lose less water via reducing aerial parts.

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