

Effect of ammonium nitrate on dry weight, sodium and potassium levels of French marigold (*Tageta patula*) in salinity stress condition

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ABSTRACT: In order to evaluate the effect of ammonium nitrate on dry weight, sodium and potassium concentrations of French marigold in salinity stress conditions was conducted an experiment as factorial arrangement in completely randomized design with salinity factor (0.49, 2.0, 4.0, 6.0 and 8.0 mmos/cm) and ammonium nitrate (AN) factor (0, 15 and 30 g/L) at four replicates. Results demonstrated that different levels of ammonium nitrate had significant influence on root dry weight and amount of potassium and sodium so that increasing ammonium nitrate led to reduction root dry weight and potassium and enhancement sodium concentration in the shoot. By increasing salinity concentration, sodium and potassium amounts increased and root and dry weight decreased. Enhancement potassium ion in marigold flower, after salinity stress is indicative to resistance against salinity but in the all salinity levels was not observed impressive changes in relation to this ion.

Keywords: French marigold, Salinity, Ammonium nitrate, Sodium, Potassium.

INTRODUCTION

French marigold (*Tageta patula*) belongs to Asteraceae family (Hassani, 1998), which is used as cut or pot flower and margining in landscape. Nowadays, marigold are grown as commercial for extraction of Carotene pigments especially Xanthophylls. The leaves of marigold are containing especial aromatic oils. Marigold oil has therapeutic applications as well as is insect repulsive. Marigold cultivation is useful to reduction of nematode population in the soil. Application of marigold in cultivation rotation can be a replacement for nematicides to reduce population of scabies-agent nematode (*Meloidogyne* spp.). Regards to the utilization of marigold as summer-flower in the parks and cities landscape especially in southern regions with saline soil, investigation on reaction of this plant to salinity can be useful. Physiological reaction of the plants against stressor agents are two cases: in the one case the plant resist and with this mechanism to encounter with slight stresses maintain itself metabolic activities in high level (similar when there is no stress) and encounter with sever stresses decrease itself metabolic activities. Other case is avoidance, which the plant is avoiding from stress by reducing itself metabolic activities and dormancy. In agriculture by using plant breeding methods, mechanism of avoidance from stress has been encouraged. In the nature, result of both mechanisms is same between different plants. Salinity and drought have especial station between stresses and have been considered more than others (Alizadeh, 2005). Salinity in the agriculture sciences is one of the soil or water properties, which is due to excessive presence of the ions. Saline soils are the soils having $EC > 4.0$ mmos/cm, exchangeable sodium percentage (ESP) less than 15 and $pH < 8.5$. These soils are entitled 'White alkali' and in Russia 'Solonchak' and according to the new American taxonomy procedure 'Solarthia' (Baybordi and Koohestani, 2004). Salinity stress lead to reduction of transpiration trend and consequently delaying to plant growth and yield reduction. Generally, salinity stress could be defined as the general effects of salts on plant growth (Hasheminia *et al.*, 1997). On the whole, the results of investigations show that the influence of salinity on plant growth and metabolism is resulting from reduction of osmotic potential arising from salt accumulation (Levitt, 1980) and ions toxicity (Winggans and Gardner, 1989). Salinity stress affects

various aspects of plant growth and metabolism. In this connection, photosynthesis (Staples and Toenniessen, 1984), nitrate reduction (Polard and Wynjones, 1979) and internal unbalancing of plant growth regulators (Shah and Loomis, 1965) are the most important cases that have been evaluated by researchers. Also in the saline media, high concentration of the ions in root medium affects nutrient elements uptake by the root (Birendra *et al.*, 1996), which these effects have been attributed to presence of high concentrations of chloride and sodium (Abo-Kaseam *et al.*, 1995). Reduction of potassium uptake (Nasseri, 1979), reduction of calcium and magnesium uptake (Al-Harbi, 1995) and reduction of ammonium and nitrate uptake (Soliman *et al.*, 1994) also are among examples of negative effects of salinity stress in relation to nutrient elements uptake. Potassium has the role in stomata conductivity and enzymes activity and transportation within plant. Study on the yield of especial crops has been indicated that different plant can be revealed very different tolerance to salinity. Marigold is very tolerant to salinity based on relative height, relative dry weight and relative flower size but according to the relative flower number is very sensitive (Decital and Morris, 1987). The presence ions in soil or water act as simulator, inhibitor or counteractive in germination stage. Salinity stress affects on the plant in all growth stages but in a majority of cases one stage is more sensitive than other stage. In the saline soils, uptake of some elements such as calcium, magnesium and potassium reduce due to sodium competition (Yeo and Flowers, 1986). Increasing sodium is leading to reduce other cations and cation unbalancing in the plant. This enhancement also is causing to reduce calcium, magnesium and potassium in the plant (Lauchi and Epsetin, 1984). In barley, excessive NaCl is leading to prevent calcium uptake and in fact sodium is causing to deficit calcium (Brown and Hayward, 1970). It has been reported that in salinity conditions, nutritive inhibitors such as sodium chloride is competing to nutritive ions and growth reduction is occurring. So even when osmotic stress has been resolved, the growth of bean, pea and sweet orange has been reduced arising salinity stress, which this reduction has been obtained due to prevalence on potassium (Hassan-shahi *et al.*, 2008). Potassium deficiency has been observed in below parts of squash and sweet clover plants by influenced salinity derived NaCl. Therefore, the main reason of growth prevention by NaCl refers to non-uptake of other minerals in competition to sodium. Low concentrations of sodium may be increase potassium uptake, although high concentrations of sodium decrease potassium uptake. In high concentrations of sodium, calcium uptake decrease (Mormohammadi-Meibodi and Ghareyazi, 2002).

Nitrogen plays key role in plant nutrition. In fact, plant life is not possible without this element. Green plants affect the presence or absence of nitrogen more than each other element. Contrary to other elements, nitrogen not available in mother soil materials and it add to soil via rain water or atmosphere by biological agents such as fixative nitrogen bacteria. In addition to nitrogen uptake by plant, amount of this element lose via leaching, erosion, ammonia sublimation and elemental nitrogen (Ghazan-shahi, 1999). Usually increasing uptake and accumulation of Cl^- is associated with reduction of NO_3^- in plant shoot. Many researchers attribute this subject to antagonism effect of Cl^- on uptake of NO_3^- (Kafkafi *et al.*, 1982). While some others said this reaction is due to influence of salinity on reduction of water absorption (Lehle *et al.*, 1992). It has been reported that in saline condition, uptake of nitrate and ammonium ions reduce and finally the plant involve to nitrogen deficiency. It is causing to reduce amino acids and protein synthesis. Also, under salinity stress, nitrate reduction in the root is confused (Odegbaro and Smith, 1969).

MATERIALS AND METHODS

In order to examine the effect of ammonium nitrate on dry weight, sodium and potassium amount of French marigolds (*Tageta patula*) under salinity stress conditions was done an experiment as factorial arrangement in completely randomized design (CRD) in Jahrom city. The first factor was different concentrations of ammonium nitrate including 0, 15 and 30 g/L and the second factor was different levels of salinity causing by NaCl including the solutions with electrical conductivity 0.49 (ambient water as control treatment), 2.0, 4.0, 6.0 and 8.0 mmos/cm with four replicates. Marigold seed was provided from a seed production center in Esfahan and was disinfected by Benomyl and 3 seeds were cultured in the 10-liters pots containing manure, sand and agricultural soil (1:1:1) and was daily irrigated by above solutions. Then the characteristics of root dry weight and shoot dry weight and amount of sodium and potassium were measured. In order to measure sodium and potassium amounts, the shoot samples were dried in oven 70 °C for 24 hours. Then 0.2 g the powdered sample was placed in 550 °C for 5 hours until whitening samples. Then the plant extracts were prepared by using 2 cc HCl and boiling water and 50 cc distilled water. Sodium and potassium concentration were measured by Flame photometer device (Ghanavati *et al.*, 2006). The obtained data was analyzed by MSTAT-C software and the means were compared by using Duncan's multiple range test (DMRT).

RESULTS AND DISCUSSION

The irrigated plants by solution of 8.0 mmos/cm died after germination stage, so this salinity level was deleted from analysis.

Root dry weight (RDW)

Based on the results of analysis of variance (Table 1), Salinity levels had significant influence ($p < 0.01$) on RDW but there was no significant effect in application of different levels of ammonium nitrate. Interaction between both factors was significant ($p < 0.01$).

Table 1. Data analysis of variance in relation to the evaluated traits

S.V	D.F	Mean Square (MS)			
		Root Dry weight (RDW)	Shoot Dry weight (SDW)	Na	K
Ammonium nitrate (A)	2	0.022 ^{ns}	0.014 ^{ns}	0.431 ^{**}	0.896 ^{**}
Salinity (B)	3	0.589 ^{**}	5.727 ^{**}	4.099 ^{**}	0.025 ^{**}
Interaction AB	6	0.040 ^{**}	0.54 [†]	0.747 ^{**}	0.080 ^{**}
Error	36	0.012	0.098	0.004	0.003
C.V %		15.14	16.61	8.05	3.81

^{ns} not significant; [†] , significant $p < 0.05$ and $p < 0.01$ respectively.

In evaluation salinity effects, the highest RDW was observed in control treatment (0.59 g) and the lowest in salinity 6.0 mmos/cm (0.12 g) (Table 2). In comparison of ammonium nitrate levels, RDW in AN 0 g/L significantly was more than AN 15 or AN 30 g/L. RDW decreased by increasing ammonium nitrate concentration (Table 2).

Table 2. Single effects and interaction between ammonium nitrate and salinity on root dry weight (g)

Ammonium nitrate	0	15	30	Salinity mean
Salinity (mmos/cm)	g/L			
0.49 (Control)	^ψ 0.74 ^a	0.56 ^b	0.49 ^{bc}	[†] 0.59 ^A
2	0.34 ^{cd}	0.40 ^c	0.31 ^d	0.35 ^B
4	0.25 ^{de}	0.15 ^f	0.14 ^f	0.18 ^C
6	0.20 ^{ef}	0.07 ^g	0.08 ^g	0.12 ^D
Ammonium nitrate mean	[†] 0.38 ^A	0.29 ^B	0.25 ^B	

^ψMeans, having minuscule (interaction), with similar letters are not significantly different ($p < 0.05$) according to DMRT

[†]Means in the right column and below row, having capital (single effects), with similar letters are not significantly different ($p < 0.05$) according to DMRT.

In evaluation interaction between both factors, the highest RDW was observed in 0 g/L ammonium nitrate and control treatment of salinity (0.74). RDW decreased by increasing ammonium nitrate levels in each salinity level. In the all ammonium nitrate concentrations, by enhancement salinity level, RDW decreased (Table 2). Root is an organ that it performs the duty of water and nutrients uptake and salinity stress mainly damages to the plant toward root. Root is the organ that is confronting to salinity stress and regards to osmotic regulation and avoiding mechanism that performs to reduce salinity effect (Carla *et al.*, 2009), a lot of the received energy from shoots is spending to compare salinity stress. This operation leads to reduction of root efficiency to provide water and nutrients for other organs. These agents may be decrease root dry weight (Safarnejad *et al.*, 2005). The obtained results in the present study also are confirmer to above investigations.

Shoot dry weight (SDW)

Salinity levels had significant influence ($p < 0.01$) on SDW but ammonium nitrate had no significant effect on SDW. Interaction between both factors was significant ($p < 0.05$) (Table 1). In comparison salinity effect, the greatest SDW was observed in control treatment (1.79 g). Salinity significantly caused to reduce SDW (Table 3). Evaluation the influence ammonium nitrate on SDW showed the highest SDW was relative to AN 0 and AN 15 g/L. SDW significantly decreased by enhancement ammonium nitrate from 15 to 30 g/L (Table 3). Interaction between salinity and ammonium nitrate indicated that the greatest SDW obtained in control salinity treatment and AN 15 g/L (1.86 g), which had no significant difference to AN 0 and AN 30 g/L in same this salinity level. In application 15 g/L ammonium nitrate, SDW reduction showed slower trend in salinity 2.0 to 6.0 mmos/cm (Table 3). Shanon (1986) reported that salinity in water and soil cause to reduce shoot and stem growth and affect plant yield. It has been reported that in many soils nitrogen deficiency is one of the main restrictive nutrition agents for plant growth. Therefore, apart from soil salinity, nitrogen application is leading to develop plant growth and yield enhancement.

Under salinity condition, reduction nitrogen uptake is one of the decrescent plant growth agents (Al-Rawahy *et al.*, 1992). Chloride has inhibitor effect on uptake and metabolism of nitrate (Bar *et al.*, 1997). Therefore, under salinity stress the need of plant to nitrogen is more than non-saline conditions (Grattan and Grieve, 1999).

Table 3. Single effects and interaction between ammonium nitrate and salinity on shoot dry weight (g)

Ammonium nitrate	0	15	30	Salinity mean
Salinity (mmos/cm)	g/L			
0.49 (Control)	^ψ 1.78 ^a	1.86 ^a	1.72 ^a	†1.79 ^A
2	1.06 ^b	1.00 ^{bc}	1.11 ^b	1.05 ^B
4	0.62 ^c	0.52 ^{cd}	0.46 ^d	0.53 ^C
6	0.23 ^e	0.31 ^{de}	0.13 ^f	0.37 ^D
Ammonium nitrate mean	†0.92 ^A	0.92 ^A	0.85 ^A	

^ψMeans, having minuscule (interaction), with similar letters are not significantly different (p<0.05) according to DMRT

†Means in the right column and below row, having capital (single effects), with similar letters are not significantly different (p<0.05) according to DMRT.

Sodium (Na)

Salinity and ammonium nitrate levels had significant influence (p<0.01) on Na amount. Interaction between both factors was significant (p<0.01) (Table 1). In comparison salinity effect, Na amount was 0.04% in control treatment but by increasing salinity level Na amount significantly increased so that the greatest Na amount was observed in salinity 6.0 mmos/cm (1.4%) (Table 4). Evaluation the effect ammonium nitrate on Na amount indicated Na in the shoot enhanced by increasing ammonium nitrate concentration. The highest amount of Na (0.87%) was observed in 30 g/L ammonium nitrate (Table 4).

Table 4. Single effects and interaction between ammonium nitrate and salinity on shoot Na amount (%)

Ammonium nitrate	0	15	30	Salinity mean
Salinity (mmos/cm)	g/L			
0.49 (Control)	^ψ 0.04 ^{gh}	0.04 ^{gh}	0.04 ^{gh}	†0.04 ^D
2	0.09 ^g	0.57 ^f	0.80 ^e	0.49 ^C
4	0.66 ^{ef}	0.76 ^e	0.99 ^d	0.77 ^B
6	1.19 ^c	1.37 ^b	1.64 ^a	1.4 ^A
Ammonium nitrate mean	†0.49 ^C	0.68 ^B	0.87 ^A	

^ψMeans, having minuscule (interaction), with similar letters are not significantly different (p<0.05) according to DMRT

†Means in the right column and below row, having capital (single effects), with similar letters are not significantly different (p<0.05) according to DMRT.

Interaction between salinity and ammonium nitrate demonstrated that the greatest Na amount was observed in 6.0 mmos/cm salinity treatment and AN 30 g/L (1.64 g). Application ammonium nitrate had no influence on reduction Na in the shoot (Table 4). Based on above results can be said French marigold easily moves Na from root to shoot. Also application ammonium nitrate led to increasing Na in the shoot.

Potassium (K)

Salinity and ammonium nitrate levels had significant influence (p<0.01) on K amount. Interaction between both factors was significant (p<0.01) (Table 1). In assessment salinity effect, the highest K amount was observed in 6.0 mmos/cm salinity treatment (1.51%). Amount of shoot K in control salinity treatment to salinity of 4.0 mmos/cm were in the same statistical level (Table 5). Evaluation the effect ammonium nitrate on K amount showed the highest K in the shoot was relative to AN 0 g/L (1.62%) and the lowest at AN 15 g/L (1.32%). Amount of K at AN 30 g/L was more than AN 15 g/L but both were lower level than AN 0 g/L (Table 5).

Interaction between salinity and ammonium nitrate demonstrated that the greatest K amount was observed in 6.0 mmos/cm salinity treatment and AN 0 g/L (1.75 g). Amount of K in the shoot decreased by increasing ammonium nitrate levels in control salinity and salinity 4.0 and 6.0 mmos/cm (Table 5). Investigations show in many plants potassium ion concentration decreased when salinity increase by the form Na ion or K/Na ratio (Grainfenberg *et al.*, 1995). In contrast, in some plants were observed that potassium in the leaves enhanced by increasing salinity (Cachorro *et al.*, 1993). In the present study, reduction K amount by increasing salinity levels is not according to the findings of Gregorio *et al.* (1998) but is conformable to the results of Ruiz *et al.* (1997). In fact, ammonium nitrate could not been increased K uptake in high salinity concentration.

Table 5. Single effects and interaction between ammonium nitrate and salinity on shoot K amount (%)

Ammonium nitrate	0	15	30	Salinity mean
Salinity (mmhos/cm)	g/L			
0.49 (Control)	^ψ 1.66 ^b	1.27 [†]	1.28 [†]	[†] 1.40 ^B
2	1.46 ^c	1.31 ^e	1.46 ^c	1.41 ^B
4	1.62 ^{bc}	1.29 ^{ef}	1.39 ^d	1.43 ^{AB}
6	1.75 ^a	1.41 ^{cd}	1.39 ^d	1.51 ^A
Ammonium nitrate mean	[†] 0.62 ^A	1.32 ^B	1.38 ^B	

^ψMeans, having minuscule (interaction), with similar letters are not significantly different ($p < 0.05$) according to DMRT

[†]Means in the right column and below row, having capital (single effects), with similar letters are not significantly different ($p < 0.05$) according to DMRT.

CONCLUSION

Different concentrations of ammonium nitrate had no significant influence on shoot dry weight. Different levels of ammonium nitrate had significant influence on root dry weight and amount of potassium and sodium so that increasing ammonium nitrate led to reduction root dry weight and potassium and enhancement sodium concentration in the shoot. Various salinity concentrations had significant influence on the all evaluated characteristics so that by increasing salinity concentration, sodium and potassium amounts increased and root and dry weight decreased. Enhancement potassium ion in marigold flower, after salinity stress is indicative to resistance against salinity but in the all salinity levels was not observed impressive changes in relation to this ion. In saline soils because of high levels of Na and Cl ions in soil solution and ion unbalancing, in spite of presence micro elements in the soil solution there is not possibility to transfer these elements to shoot and consequently lead to deficiency appearance and damage to plant.

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