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Effect of foliarly applied potassium on *Capsicum Annuum* L. grown under sodium chloride stress

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Abstract

Salinity is one of the environmental factors that has a critical influence on the germination and plant establishment. In present study, two independent experiments were conducted to investigate the effects of salinity and potassium on germination, seedling establishment and growth of *Capsicum annuum* L. A range of sodium chloride concentrations control (non-saline), 60mM NaCl (EC=8.5mS/cm), 100mM (EC=11.73mS/cm) showed reduction in germination percentage and different parameters of seedling stage (plumule length, root length, fresh and dry biomass). Application of potassium (400 ppm and 800 ppm KNO_3) enhances the germination percentage and seedling growth. To study the vegetative growth, some biochemical aspects and ionic composition of different plant parts of *Capsicum annuum* L., a pot experiment was conducted in Botanical Garden, Abdul Wali Khan University Mardan, Pakistan. The experiment was laid out in a completely randomized factorial design with three replicates. Potassium nitrate was used as the potassium source. The rate of potassium treatment was 400 and 800 ppm. Plants were subjected to 60mM (EC=8.5mS/cm), 100mM (EC=11.73mS/cm) levels through addition of NaCl to irrigation water. Results showed that by increasing salt concentration different growth parameters (Plant height, root length, number of leaves, fresh and dry biomass) exhibited decrease. Electrolyte leakage, chlorophyll content, carotenoids, total proteins showed increase under different salt treatments. It has been concluded that application of both concentration of KNO_3 (400 & 800 ppm) through leaves exhibited enhancement effect on growth parameters under normal as well as saline condition but 800 ppm KNO_3 concentration exhibited more pronounced alleviating effect under NaCl stress.

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Introduction

A plant's first line of defense against abiotic stress is in its roots. If the soil holding the plant is healthy and biologically diverse, the plant will have a higher chance of surviving stressful conditions (Brussaard *et al.,* 2007). The most obvious detriment concerning abiotic stress involves farming. It has been claimed by one study that abiotic stress causes the most crop loss of any other factor and that most major crops are reduced in their yield by more than 50% from their potential yield (Wang *et al.,* 2007). It has also been speculated that this yield reduction will only worsen with the dramatic climate changes expected in the future (Lane and Jarvis 2007).

The problem of salinity existed long before the human beings and start of agricultural practices. A progressive increase in salinity has caused degradation of arable land over many hundred years period cultivated land could be degraded due to salinity during less than 100 years (Lewis, 1984). At present, its extent throughout the world is increasing regularly (Schwabe *et al*., 2006) and it has now become a very serious problem for crop production (Munns and Tester, 2008), particularly in arid and semi-arid regions. Soil salinity causes adverse effects on different physiological processes which are responsible for the reduction of growth of plants (Ashraf, 1994; 2004; Munns *et al*., 2006).

Various strategies can be adopted to cope with salinity stress. In recent years there has been much interest in the development of salt tolerant crop varieties. For this purpose, genetic improvement of salinity tolerance in the cultivated genotypes has been proposed as the most effective strategy to solve salinity problems. Salt tolerance should be evaluated at germination, seedling and adult stages (Ashraf, 2004).While establishing appropriate salinity screening techniques, it is also important to understand which of the physiological or biochemical processes is more sensitive to salt stress that can be used as effective selection criterion (Ashraf 2004; Ashraf and Harris, 2004).

Development of crop plants tolerant to salt stress is very important to meet the growing food demand. It has been suggested to exploit naturally occurring inter and intra specific genetic variability by hybridization of selected salt tolerant genotypes with high yielding genotypes adapted with target environment (Munns *et al.,* 2006).

Chilies (*Capsicum annuum* L.) belong to the nightshade family, Solanaceae and originates from South America. The main production areas of pepper are in the northeast of Thailand. The area for growing peppers in Thailand is approximate 1,000 – 1,200 acres, with important partner countries; the U.S., Australia, English, Philippines and Japan (Saterungsri, 2009). Peppers are an important agricultural crop, not only for their economic importance but also for the nutritional value of their fruit, mainly because they are an excellent source of natural pigments and antioxidant compounds in addition to their excellent flavor and pungency (Navarro *et al.,* 2002). Chilies are very rich in vitamin C and pro-vitamin A, particularly the red chilies. In addition, peppers are a good source of most B vitamins, and vitamin B_6 in particular. They are very high in potassium and high in magnesium and iron. Their high vitamin C content can also substantially increase the uptake of non-heme iron from other ingredients in a meal, such as beans and grains (Sparkyby, 2006). Aim of this study was to observe the effect of different NaCl concentrations (60 and 100mM) on *Capsicum annuum* growth and how different concentrations of KNO_3 ameliorate the inhibitory effect of NaCl by studying their germination, growth, biochemistry and ionic composition of different parts.

Materials and methods

Germination studies

Seeds of *Capsicum annuum* were obtained from Nursery of Mardan (KPK) and germinated using NaCl and different concentrations of KNO_3. A range of concentrations of sodium chloride control (non-saline), 60mM NaCl (EC=8.5mS/cm), 100mM (EC=11.73mS/cm) NaCl were selected as the NaCl

stress concentration. Seeds were surface sterilized with 0.1% (w/v) mercuric chloride solution and washed thoroughly with several changes of sterile distilled water. They were soaked for 2h either in (i) distilled water (control) (ii) 400 ppm KNO_3 or (iii) 800 ppm KNO_3. Ten seeds from each treatment were distributed in separate petriplates (15 cm diameter) provided with moist Whatman No. 1 filter papers with each concentration of NaCl. The seeds were allowed to germinate in the dark at 28 ± 1◦C. The number of seeds germinated was recorded at the end of 24h and 36 h after transfer into petriplates. On the 9th day, seedling growth in terms of length, fresh and dry weights was recorded. For dry weight measurement, seedlings were dried at 70 ◦C for 24 h.

Growth experiment

The seeds of *Capsicum annuum* were obtained from Nursery of Mardan (KPK). Concentrations of sodium chloride control (non-saline), 60mM NaCl (EC=8.5mS/cm), 100mM (EC=11.73mS/cm) NaCl were selected as the NaCl stress concentration. Seeds of uniform size were washed with distilled water after surface sterilizing with 0.1% mercuric chloride solution. The treatments were applied in three sets as follows.

Set I= Different NaCl concentrations without KNO_3 (Control)
Set II= Different NaCl concentrations with 400 ppm KNO_3.
Set III = Different NaCl concentrations with 800 ppm KNO_3.

4Kg soil was taken in earthen pots of uniform size having basal hole for leaching purpose. Three seeds were sown in each pot and at three leaf stage the pots were irrigated with Hoagland (Nutritive) solution. Each Set has 4 pots/treatment, and each pot was irrigated with 1L of tap water / salt solution twice a week. The plants were harvested at mature plant stage (90 days after sowing) to analyze various morpho-physiological, biochemical and yield attributes.

Measurement of growth attributes

Three plants were harvested randomly from four replicates at mature stage (90 days after sowing). Plant height, Root length, number of leaves, leaf area, number of fruits, fresh and dry biomass (g) were recorded in harvested plants.

Electrolyte leakage (EL)

EL was measured as described by Lutts *et al.* (2004) with a few modifications. Plant material 0.3g was washed with deionized water. Place in tubes with 15ml of deionized water and incubated for 2 hrs. at 25°C. Electrical conductivity of the solution (L_1) was determined. Samples were then autoclaved at 120°c for 20 min and the final electrical conductivity (L_2) was measured after equilibrium at 25°C. EL was measured using the following formula;
EL (%) =$L_1/L2*100$.

Mineral estimation of vegetative parts

Samples of leaf, stem and root were taken at grand period of growth for the analysis of different cations (Na^+, K^+). Samples were dried and 0.5gm of each dry sample was taken for ash weight. Then solution of ash was made in 50ml of de-ionized water, and then dilutions were made in de-ionized water for mineral analysis. Concentration of cations in samples was measured using PFP 1 Flame Photometer.

Chlorophyll content

Chlorophyll concentration (Chl) was estimated following the protocol of Maclachlam, and Zalik, (1963). 0.1g of fresh weight of leaves was taken and macerated with a little of acid washed sand with 3 ml. of 80% acetone, centrifuged at 1000 rpm. For five minutes at 15-30 °C. The debris was then washed 3 times using 1 ml. of 80% acetone each time. The supernatant was then pooled and mad the volume upto7 ml. with acetone. Optical density of these solutions was then recorded at 663nm and 645nm, on photospectrometer. Chlorophyll a, b and Carotenoid were calculated using the following formula.

$$Chlorophyll\ "a"\ (mg/g\ fresh\ weight) = \frac{12.3\ D_{663} - 0.86\ D_{645}}{d \times 1000 \times W} \times V$$

$$Chlorophyll\ "b"\ (mg/g\ fresh\ weight) = \frac{12.3\ D_{645} - 0.86\ D_{663}}{d \times 1000 \times W} \times V$$

$$Carotenoid\ (mg/g\ fresh\ weight) = \frac{7.6\ D_{480} - 1.49\ D_{510}}{d \times 1000 \times W} \times V$$

Protein extraction

Protein was determined by method described by Bradford (1976) and Bollag and Edelstein (2001), using bovine serum albumin as standard. Leaf samples (100 mg) were homogenized with 3ml extraction buffer (50mM Tris-HCl (pH: 7.5), 2mM EDTA, 1mM 2-Mercaptoethanol, 1mM DTT). Samples then were centrifuged at 14000 rpm for 25 min at 4◦˚c. and supernatants were isolated and used for protein assay.

Estimation

Take 0.5 ml of supernatant and adjust volume to 1.0 ml with phosphate buffer and to this 50mL of Bradford reagent was added and absorbance recorded at 595 nm against reagent blank. A standard calibration curve is drawn by using bovine serum albumin as standard. From the standard curve, the concentrations of proteins in samples were obtained.

Experimental design and statistical analysis

The experimental design was completely randomized Design (CRD) with three salt levels and three replicates. Collected data was analyzed statistically by using SPSS v. 21.0 to analysis of variance (ANOVA) and the means compared by Duncan's multiple range test ($P < 0.05$).

Results and discussion

Germination

Salinity is considered as a major abiotic stress and significant factor affecting crop production all over the world and especially in arid and semi-arid region (Davidson and Chevalier, 1987). Seed germination was negatively affected by drought (Damirkaya *et al.,* 2006) and salinity stresses (Zhu, 2002). Seeds treated with different NaCl concentrations exhibited significant ($P<0.05$) reduction in germination as compared to their non-saline media. Salts in the medium may have osmotic or specific toxicity effects on germinating seeds and seed germination may be merely retarded or may be reduced. Seed germinability is reduced under salt stressed conditions due to physiological injuries under such conditions and these stressed seeds are desiccation sensitive. Salinity can affect germination and seedling growth either by creating an osmotic pressure that prevents water uptake or by toxic effects of sodium and chloride ions (Hopper *et al.,* 1979). Seeds germinated in saline medium with the amendment of KNO_3 showed significant ($P<0.05$) improvement in both saline as well as in non- saline environment. These results agree with some studies which indicated that priming treatments significantly improved seed performance over the control and KNO_3 primed seeds excelled over all other treatments (Farooq *et al.,* 2005).

Seedling stage

Seeds treated with different NaCl concentrations exhibited non-significant reduction in shoot length in both NaCl concentrations as compared to control. Significantly reduced plumule growth with increasing salt levels in the present study agrees with the findings of Khan *et al.* (1994), Ibrar *et al.* (2003) and Jabeen *et al.* (2003) who also reported significant decline in plumule growth at 10 dS/m and higher salinity levels. Seeds germinated in saline medium with the amendment of KNO_3 showed significant ($P<0.05$) improvement in stem length in both saline media and control. Our findings showed that K^+ application not only stimulated the negative effects of salinity on growth, but also reduced dry matter accumulation particularly at low and medium stress. This is in contrast with the data of some authors who have reported ameliorative effect of K^+ on salinity tolerance (Shirazi *et al.,* 2005; Ottow *et al.*, 2005).

Seeds treated with different NaCl concentrations exhibited non-significant decrease in root length as compared to both NaCl (60and 100mM). Elevated salinity slows down water uptake by seeds, thereby inhibiting their germination and root elongation (Uhvits, 1946; Simon, 1984; Werner and Finkelstein, 1995). Seeds germinated in saline medium with the

amendment of KNO_3 showed improvement in root length in both saline as well as in non- saline environment. Our results confirm the findings of Stofella *et al.* (1992), who reported that priming of the pepper seeds significantly improved root length. Significant improvement in root and shoot length may be attributed to earlier germination induced by primed over unprimed seeds (Farooq *et al.,* 2005), which resulted in vigorous seedlings with more root and shoot length than the seedlings from unprimed seeds.

Table 1. Effect of KNO3 and different NaCl concentrations on electrolyte leakage of *Capsicum annuum*.

Treatment	Without KNO_3	400 ppm KNO_3	800 ppm KNO_3
Control			
Mean	62.584a	95.080a	67.056a
SE	±39.719	±12.406	±22.088
60 mMNaCl			
Mean	132.749a	126.130b	117.719a
SE	±5.606	±3.498	±2.280
	(+112.111)	(+32.656)	(+75.552)
100 mMNaCl			
Mean	81.472a	87.262b	68.901a
SE	±2.587	±1.700	±11.491
	(+30.18)	(-8.222)	(+2.751)
$LSD_{0.05}$	80.309	25.874	49.953

Means followed by different letters in the same column differ significantly
at 95% probability level according to New Duncan's Multiple Range Test.
Figures in parentheses indicate % promotion (+) and reduction (-) over control.

Seeds treated with different NaCl concentrations exhibited significant ($P<0.01$) reduction in biomass in treated plants as compared to control. Seedling growth was suppressed under saline conditions, which is strongly in accordance with Cicek and Cakirlar (2002) who reported that salinity reduced shoot length, fresh and dry weight of maize seedlings. Same results were also studied for Corn (Bar-Tal *et al.,* 1991), tomato (Adams, 1988 and Satti and Al-Yahyi, 1995), cucumber and pepper (Kaya *et al.,* 2001). Seeds germinated in saline medium with the amendment of KNO_3 showed significant ($P<0.01$) improvement in biomass (fresh and dry weight) in control and both salinity levels. Akram *et al.* (2009) observed an improvement in growth of sunflower due to the foliar spray of K2SO4 and KNO3 at 1.25% under saline concentration of 150mM NaCl.

Growth studies

Plants treated with different NaCl concentrations exhibited significant ($P<0.001$) reduction plant height in different concentration of NaCl as compare to control (non-saline). Plants treated with high salt concentration exhibited reduction in plant height. According to Alam *et al.* (2004), it is possible that the decrease in the observed plant height in salinized plants were due to several reasons. One possibility is that salinity reduced photosynthesis, which in turn limited the supply of carbohydrate needed for growth. A second possibility is that salinity reduced shoot and roots growth by reducing turgor in expanding tissues resulting from lowered water potential in root growth medium. Third, a disturbance in mineral supply, either an excess or deficiency, induced by changes in concentrations of specific ions in the growth medium, might have directly affected growth (Lazof and Bernstein, 1998; Zhu, 2002). Plants grown in saline medium and amended with KNO_3 showed reduction

in plant height in saline medium as compare to their respective control (non-saline). Height of both the plant sunflower and safflower growing under different sea salt concentrations without any foliar application was reduced. Height of those plants sprayed with only water showed little betterment. A significant increase in height was observed in plants sprayed by individual macro or micronutrient solutions (i.e. K, B and Fe) but combined effects of their mixture was more significant irrespective to their growth under non saline (control) or saline conditions by El-Kader *et al.* (2006) and Ahmad & Jabeen (2009).

Table 2. Effect of KNO_3 and different NaCl concentrations on chlorophyll a, b, total chlorophyll, carotenoids, total sugars and proteins of *Capsicum annuum*.

Without KNO3

Treatment	Chlorophyll-a	Chlorophyll-b	Total Chlorophyll	a/b Ratio	Carotenoids	Total Protein
	(mg/gm fr.wt)	(mg/gm fr.wt)	(mg/gm fr.wt)		(mg/gm fr.wt)	(mg/gm dry.wt)
Control						
Mean	2.792a	1.457a	4.249a	1.920a	1.216a	0.288a
SE	±0.137	±0.110	±0.2484	±0.051	±0.070	±0.096
60 mM NaCl						
Mean	1.926a	0.952a	2.879a	2.022a	1.0148a	2.564a
SE	±0.006	±0.018	±0.011	±0.046	±0.008	±0.116
	(-31.010)	(-34.660)	(-32.240)	(+5.310)	(-16.610)	(+790.28)
100 mM NaCl						
Mean	4.062a	1.883a	5.946a	2.181a	1.578a	2.517a
SE	±0.583	±0.374	±0.957	±0.123	±0.333	±0.132
	(+45.505)	(+29.240)	(+39.940)	(+13.590)	(+29.770)	(+773.960)
$LSD_{0.05}$	3.593	1.767	5.354	2.363	1.540	2.412

400ppm KNO3

Treatment	Chlorophyll-a	Chlorophyll-b	Total Chlorophyll	a/b Ratio	Carotenoids	Total Protein
	(mg/gm fr.wt)	(mg/gm fr.wt)	(mg/gm fr.wt)		(mg/gm fr.wt)	(mg/gm dry.wt)
Control						
Mean	3.385a	1.802a	5.187a	1.882a	1.513a	0.261a
SE	±0.091	±0.068	±0.119	±0.0842	±0.00093	±0.065
60 mM NaCl						
Mean	3.452a	1.980a	5.432a	1.753a	1.652a	1.060b
SE	±0.472	±0.179	±0.583	±0.203	±0.168	±0.151
	(+1.980)	(+9.880)	(+4.720)	(-6.850)	(+9.190)	(+306.13)
100 mM NaCl						
Mean	1.680b	0.780b	2.461b	2.155a	0.761b	4.789b
SE	±0.179	±0.0807	±0.2563	±0.081	±0.072	±0.550
	(-50.370)	(-56.710)	(-52.55)	(+14.510)	(-49.70)	(+1731.42)
$LSD_{0.05}$	1.027	0.416	1.295	0.470	0.366	1.295

800 ppm KNO3

Treatment	Chlorophyll-a	Chlorophyll-b	Total Chlorophyll	a/b Ratio	Carotenoids	Total Protein
	(mg/gm fr.wt)	(mg/gm fr.wt)	(mg/gm fr.wt)		(mg/gm fr.wt)	(mg/gm dry.wt)
Control						
Mean	3.039a	1.464a	4.504a	2.081a	1.250a	0.058a
SE	±0.252	±0.145	±0.396	±0.0464	±0.343	±4.778
60 mM NaCl						
Mean	2.858a	1.401a	4.260a	2.046a	1.192a	4.859b
SE	±0.198	±0.122	±0.317	±0.0676	±0.130	±0.140
	(-5.960)	(-4.30)	(-20.056)	(+45.418)	(-61.973)	(+293520735276.06)
100 mM NaCl						
Mean	1.501b	0.310b	1.812b	6.795a	0.650a	3.998c
SE	±0.479	±0.135	±0.343	±4.505	±0.1870995	±0.235
	(-50.610)	(-100)	(-100)	(-100)	(-100)	(-100)
$LSD_{0.05}$	0.925	0.473	1.253	5.983	0.881	0.449

Means followed by different letters in the same column differ significantly at 95% probability level according to New Duncan's Multiple Range Test.

Figures in parentheses indicate % promotion (+) and reduction (-) over control.

In this study, root length showed significant ($P<0.001$) reduction under different salinity levels. According to Hartung (2004) that the adverse effects of salinity on plant height and root length may be due to the diverse effects of salinity on meristimatic cell division and elongation as well as root penetration. Gama *et al.* (2007) also reported reduction in root length as a result of salinity.

Table 3. Cation composition of Capsicum annuum grown at different concentrations of KNO3 and NaCl.

Without KNO3

Treatment	STEM			ROOT			LEAVES		
	Na^+	K^+	K^+/Na^+	Na^+	K^+	K^+/Na^+	Na^+	K^+	K^+/Na^+
Control									
Mean	269.56a	174.85a	0.654a	710.47a	130.26a	0.183a	214.743a	229.32a	1.070a
SE	±17.595	±19.37	±0.089	±74.095	±22.637	±0.029	±7.437	±13.532	±0.074
60 mM NaCl									
Mean	358.263a	130.78a	0.373ab	908.116a	84.5a	0.091a	322.92b	267.54ab	0.878a
SE	±13.040	±35.582	±0.113	±34.850	±17.546	±0.0164	±40.480	±20.876	±0.199
	(-32.906)	(-25.204)	(-43.001)	(+27.819)	(-35.129)	(-50.013)	(+50.374)	(+16.666)	(-17.971)
100 mM NaCl									
Mean	371.91b	98.28a	0.270b	903.823b	163.93a	0.185a	483.076c	313.82b	0.648a
SE	±36.663	±8.193	±0.037	±31.601	±48.110	±0.061	±23.624	±28.174	±0.041
	(+37.969)	(-43.791)	(-58.725)	(+27.214)	(+25.848)	(+0.8155)	(+124.955)	(+36.848)	(-39.455)
$LSD_{0.05}$	85.32	82.579	0.297	175.327	111.862	0.141	94.808	75.101	0.433

400ppm KNO3

Treatment	STEM			ROOT			LEAVES		
	Na^+	K^+	K^+/Na^+	Na^+	K^+	K^+/Na^+	Na^+	K^+	K^+/Na^+
Control									
Mean	223.176a	142.22a	0.646a	499.866a	171.73a	0.364a	169.203a	277.55a	1.650a
SE	±14.501	±20.345	±0.117	±70.040	±13.060	±0.075	±15.717	±16.299	±0.054
60 mM NaCl									
Mean	233.22a	150.54a	0.693a	639.783a	151.84a	0.244a	226.55b	285.22a	1.261b
SE	±38.401	±17.320	±0.161	±104.090	±10.902	±0.023	±8.444	±14.56	±0.071
	(+4.500)	(+5.850)	(+7.315)	(+27.990)	(-11.582)	(-32.798)	(+33.892)	(+2.763)	(-23.546)
100 mM NaCl									
Mean	293.25a	66.17b	0.224b	667.306a	251.81a	0.385a	355.58c	332.54a	0.937c
SE	±26.680	±]8.32	±0.008	±138.683	±53.743	±0.075	±11.345	±21.767	±0.071
	(+31.398)	(-53.473)	(-65.341)	(+33.496)	(+46.631)	(+5.688)	(+110.149)	(+19.812)	(-43.165)
$LSD_{0.05}$	97.810	55.910	0.399	373.630	112.625	0.223	42.243	61.627	0.230

800 ppm KNO3

Treatment	STEM			ROOT			LEAVES		
	Na^+	K^+	K^+/Na^+	Na^+	K^+	K^+/Na^+	Na^+	K^+	K^+/Na^+
Control									
Mean	253.23a	165.1a	0.665a	226.55a	217.62a	0.962a	134.243a	277.55a	2.098a
SE	±18.506	±13.533	±0.093	±11.960	±21.827	±0.093	±13.700	±31.151	±0.261
60 mM NaCl									
Mean	266.493a	128.18a	0.489a	428.566a	405.47a	0.972a	225.476ab	300.69a	1.382ab
SE	±16.713	±16.330	±0.083	±60.149	±92.448	±0.226	±32.265	±29.437	±0.225
	(+5.237)	(-22.362)	(-26.395)	(+89.170)	(+86.320)	(+1.024)	(+67.961)	(+8.337)	(-34.110)
100 mM NaCl									
Mean	288.88a	125.19a	0.438a	509.91b	259.74a	0.509a	337.563a	348.53a	1.133b
SE	±17.003	±33.924	±0.119	±0	±49.299	±0.096	±76.232	±12.012	±0.2286
	(-14.078)	(-24.173)	(-34.153)	(+125.076)	(+19.354)	(-47.057)	(+151.456)	(+25.573)	(-45.999)
$LSD_{0.05}$	60.300	79.932	0.347	131.535	228.240	0.526	167.635	88.930	0.827

Means followed by different letters in the same column differ significantly at 95% probability level according to New Duncan's Multiple Range Test.

Figures in parentheses indicate % promotion (+) and reduction (-) over control.

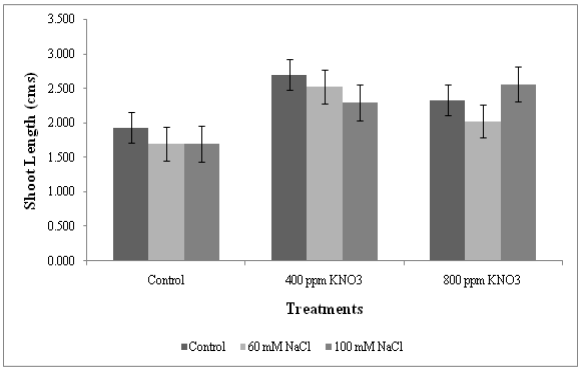

Fig. 1. Effect of KNO_3 and NaCl concentrations on shoot length (cms) of *Capsicum annuum* seedlings.

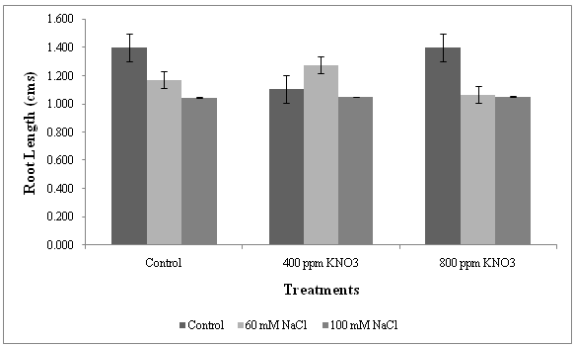

Fig. 2. Effect of KNO_3 and NaCl concentrations on root length (cms) of *Capsicum annuum* seedlings.

Plants treated with different NaCl concentrations exhibited significant ($P<0.05$) reduction number of leaves in NaCl stress. Salinity generally affects the growth rate and results in plants with smaller leaves. Additionally, this type of stress produces changes in leaf color (chlorosis) and even necrosis, which leads to the deterioration of the leaves and thus to inhibition of photosynthesis. In general, the reduction in growth of plant under salt stress can lead to death of the plant (Munns *et al.,* 2005). One of the main problems related to salinity in *C. annuum* manifests in the aerial region of the plant, particularly in the leaves. In sensitive genotypes, chlorosis and necrosis of the leaves was observed (Aktas *et al.,* 2006), as well as a reduction in leaf area (Chartzoulakis and Klapaki, 2000). This effect was accompanied by a reduction in chlorophyll content, a relatively low rate of photosynthesis and net assimilation of CO_2 and a low conductance (Martinez-Ballesta*et al.,* 2004). At high concentrations of NaCl (100 to 150 mM), the photosynthetic rate was reduced to approximately 85%, which led to decreased plant growth and to ultimately death of the plants. Amendment of different concentrations of KNO_3 exhibited increase in this parameter. Foliar supply of KNO_3 to the salt treated plants may reduce toxic ions uptake as well improve K and N status of salt treated plants. The role of potassium in ionic balance is reflected in nitrate metabolism (Jeschke and Wolf, 1985). Nitrogen being an active participant of chlorophyll and protein is an essential element for plant growth. Spray with potassium result an increase in leaf potassium content which was accompanied by increased rates of photosynthesis, photorespiration and RuBP carboxylase activity. Hence there was considerable improvement in growth even under saline strata in present Investigation.

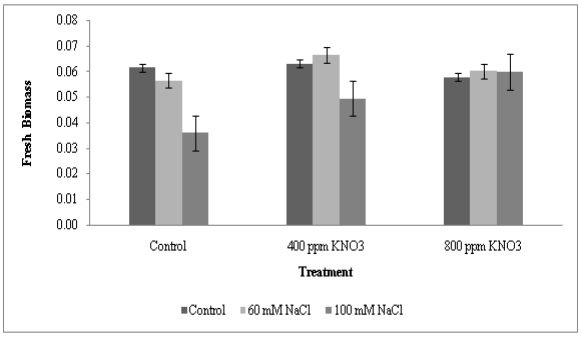

Fig. 3. Effect of KNO_3 and NaCl concentrations on fresh biomass (gms) of *Capsicum annuum* seedlings.

Plants treated with high salt concentration exhibited significant ($P<0.001$) reduction in fresh and dry biomass. The observed reduction in dry weight under high salt concentration might be attributed to the combination effects of osmotic and specific ions of Cl^- and Na^+ (Basal, 2010; Hajer *et al.,* 2006). Several researchers reported that fresh and dry weights of cotton plants might be affected under saline conditions (Ahmad *et al.,* 2002; Akhtar *et al.,* 2010; Basal, 2010). Similar observation has also been recorded in tomato (Hajer*et al.,* 2006) and Maize (khatoon *et al.,* 2010). Reduction in total biomass under different salinity levels was also reported by Khan *et al. (1989),* Tahir & Mehdi (2001) and Bassil *et al.* (2002). Reduction in biomass with increasing concentration of salinity may be due to reduction in water content both in roots and shoots which indicates that plants were not adjusted osmotically under various levels of salinity.

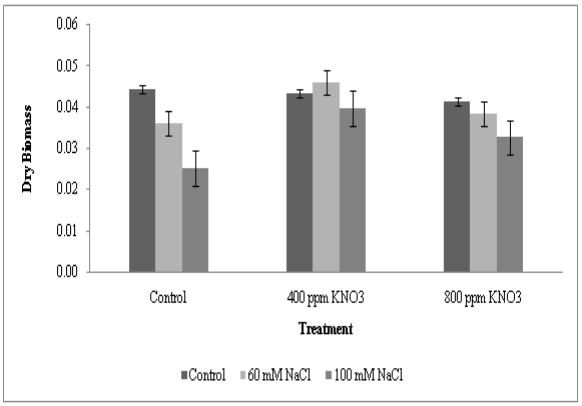

Fig. 4. Effect of KNO_3 and NaCl concentrations on dry biomass (gms) of *Capsicum annuum* seedlings.

A significant (P<0.001) increase in fresh and dry biomass was observed in plants sprayed by KNO_3 irrespective to their growth under non saline control or saline conditions. The obtained results are in agreement with the findings of Asad *et al.* (2003), Basole *et al.* (2003), Kassab (2005) and Thalooth *et al.* (2006). These results suggested that foliar application of nutrient solution partially overcame salt-induced detrimental effects in plant.

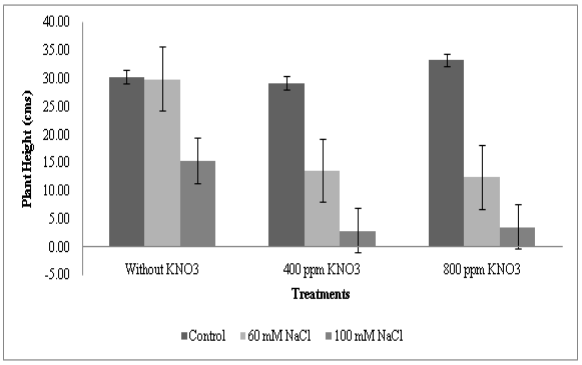

Fig. 5. Effect of KNO_3 and NaCl concentrations on plant height (cms) of *Capsicum annuum*.

Plants treated with different NaCl concentrations exhibited increase in electrolyte leakagein both concentration of NaCl (60 and 100mM) as compare to control. Quan *et al.* (2004) found higher electrolyte leakage in sugar beet plants grown under salt stress than in plants grown under control conditions. Amendment of plants with KNO_3 showed significant (P<0.05) decrease in this parameter which contradicts the result obtained by Kaya *et al.* (2002).

Plants treated with different NaCl concentrations exhibited significant (P<0.05) increase in Chlorophyll a, b, total Chlorophyll and a/b ratios in high concentration of NaCl (100mM) as compare to control. The decreased levels in chlorophyll content under saline stress is commonly reported phenomenon and established that it may be due to different reasons; one of them is related to membrane deterioration (Ashraf & Bhatti, 2000). Plants grown in saline medium with the amendment of KNO_3 (400ppm, 800ppm) showed significant (P<0.05) reduction in both NaCl levels as compared to control. K+ is an ameliorative effect under the salinity stress (Mohammad *et al.,* 2011).

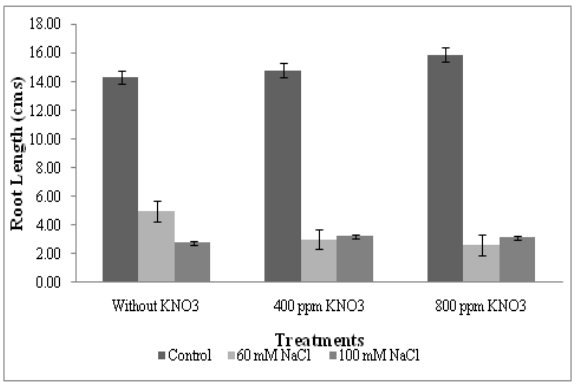

Fig. 6. Effect of KNO_3 and NaCl concentrations on root length (cms) of *Capsicum annuum*.

Plants treated with different NaCl concentrations exhibited significant (P<0.05) increase in carotenoids in high NaCl concentration of (100mM) as compare to 60mM and control (non-saline). A reduction in carotenoids content due to salinity stress has been observed by Ali *et al.* (1992) in *Brassica juncea*, Agastian *et al.* (2000) in mulberry and Parida *et al.* (2005) in *Aegiceros corniculatum*. Plants grown in saline medium with the amendment of KNO_3 (400ppm, 800ppm) showed significant (P 0.05) decrease in Carotenoids at both salinity levels as compare to their respective control. Ammonium phosphate and potassium sulphate had a positive effect on chlorophyll and carotenoids concentrations.

Plants treated with different NaCl concentrations exhibited significant (P<0.05) increase in total proteins in both concentration of NaCl as compare to control. The exogenous application of KNO_3 is related to increased NO_3^- absorption, its reduction and assimilation (Ruiz and Romero, 1999). The soluble protein concentrations significantly (P<0.05) increased with the foliar application of KNO_3

irrespective to the plant growth under non saline or saline conditions. It may be due to the direct involvement of K^+ in several steps of translation process, including the binding of tRNA to ribosomes (Evans and Wildes, 1971).

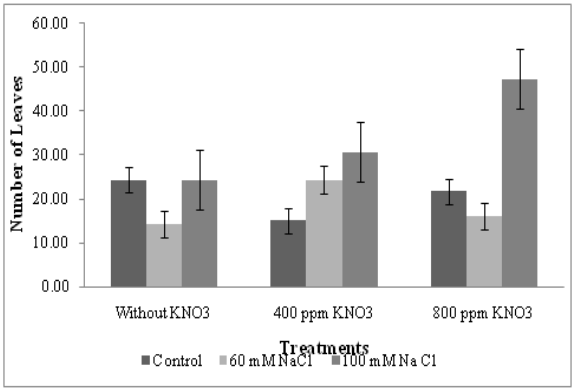

Fig. 7. Effect of KNO_3 and NaCl concentrations on number of leaves of *Capsicum annuum*.

Plants treated with different NaCl concentrations exhibited significant ($P<0.05$) increase in Na^+ concentration as compared to non-saline, One of the most important effects of high concentrations of Na^+ is the displacement of K^+ from target sites within the cell because both ions show great chemical similarity. The first point of interaction between Na^+ and K^+ is the entry of these ions to the root symplast. However, high concentrations of Na^+ inhibit directly the transport systems for K^+. Furthermore, Na^+ causes depolarization of the membrane electrical potential, which decreases the absorption of K^+. Accordingly, in salt stress conditions, K^+ deficiency may occur as has been observed in corn, melon and pepper (Botella *et al.,* 1997; Kaya and Higgs, 2003; Kaya *et al.,* 2007; AlemánGuillén, 2009).

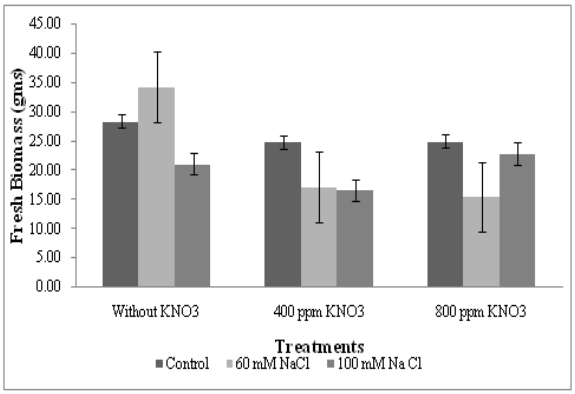

Fig. 8. Effect of KNO_3 and NaCl concentrations on fresh biomass (gms) of *Capsicum annuum*.

The absorption of K^+ plays an important role in the growth and development of plants (Mengel and Kirkby, 1982; Ashley *et al.,* 2006) because of the various functions performed by K^+ in cells. Thus, salt-tolerant plants must maintain a high level of K^+ in the cells (Göl, 2006). Under salt stress, K^+ deficiency occurs because the ionic radius and hydration energy of K^+ and Na^+ are similar, which prevents discrimination between these two ions (Zhang *et al.,* 2010). Because of the competition between K^+ and Na^+, the selectivity and operation of transport systems for $K^+$and Na^+ through various cellular membranes is essential for maintaining a proper cytoplasmic K^+ / Na^+ relationship. High concentrations can inhibit Na+ transport systems in favor of K^+ and K^+ / Na^+ selective systems, which may influence the salt tolerance of plants (Alemán-Guillen, 2009).

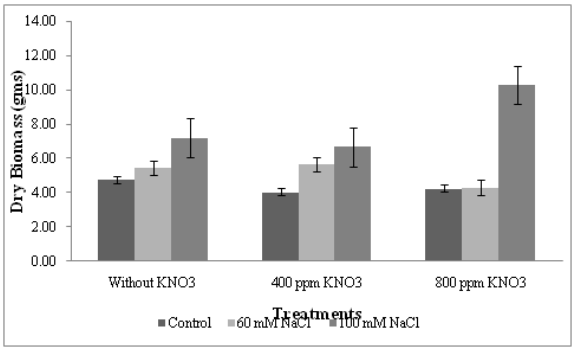

Fig. 9. Effect of KNO_3 and NaCl concentrations on dry biomass (gms) of *Capsicum annuum*.

In salinity, control of homeostasis depends on sodium efflux from the cytoplasm as the K^+ concentration is maintained. Channels and K^+ transporters may control the transport of Na^+ (Jérémie-Diedhiou, 2006). In wheat species that are tolerant to salinity, a minor efflux of K^+ from the roots in the presence of high concentrations of NaCl is correlated with tolerance to salt stress because this lower efflux of K^+ from the roots maintains the K^+ / Na^+ relationship in the plant (Cuin*et al.,* 2008). A high cytosolic K^+ / Na^+ ratio is an essential requirement for plant growth at high salt concentrations (Zhu, 2003).

In present investigation foliar application of KNO_3 alleviate the toxicity of Na^+ by decreasing the chances of its accumulation in plant parts. The results of

present investigation are in agreement with the findings of many workers in different plant species (Sultana *et al.,* 2001; Cha-um *et al.,* 2010) who found that nutrients were absorbed by the leaves when applied onto the shoot. Treated leaves contained higher element concentration compared to non-sprayed plants even under saline condition.

It is concluded from the study that growth of *Capsicum annuum* showed reduction under different NaCl concentrations (60 and 100 mM). Application of KNO_3 (400 & 800 ppm) showed increased growth under normal as well as stressed condition, but 800 ppm KNO_3 concentration exhibited more pronounced alleviating effect under NaCl stress.

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