ISSN 1684-5315 @ 2011 Academic Journals

Full Length Research Paper

Evaluation of drought and salinity stress effects on germination and early growth of two cultivars of maize (*Zea mays* L.)

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Accepted 21 September, 2011

To study the effect of polyethylene glycol (PEG) and NaCl stress on germination and early seedling stages on two cultivars of maize, two separated experiment were laid out at seed laboratory in Iran in 2011. This investigation was performed as factorial experiment under completely randomized design (CRD) with three replications. Cultivar factor contains two cultivars (SC666 and SC704) and six levels of stress (0, -2, -4, -6, -8 and -10 bar). The principal aim of this current study was to compare the two cultivars of maize relative to the stress conditions. Results indicate that significant decrease was observed in germination percentage, germination rate, length of radicle and plumule and length of seedling traits in both conditions. We conclude that at each level of osmotic potentials, the inhibitory effect of salinity stress on germination was more than that of drought stress. SC666 cultivar had more tolerant than SC704 cultivar in both stress conditions.

Key words: Maize, early growth, germination, PEG and NaCl stress.

INTRODUCTION

Maize (Zea mays L.) is the sole cultivated member of genus Zea and tribe Maydeae, ranks as one of the three important cereal crops in the world after wheat and rice (Wattoo et al., 2009). Maize being nutritionally an important crop has multiple functions in the traditional farming system, being used as food and fuel for human being and feed for livestock and poultry (Wattoo et al., 2009). Several abiotic stresses affect the crop productivity. One of the most important abiotic factors limiting plant germination and early seedling stages is water stress brought about by drought and salinity (Almansouri et al., 2001), which are widespread problems around the world (Soltani et al., 2006). Maize is being increasingly cultivated in Iran. Its cultivation area is expanding to areas having high potential for accumulation of salts in the soil profile, such as Khuzestan. It is therefore, important to develop new maize varieties with high genetic capacity to tolerate salt stress. Salinity and drought affect the plants in a similar way (Katerji et

al., 2004). Reduced water potential is a common consequence of both salinity and drought (Legocka and Kluk. 2005). Water stress acts by decreasing the percentage and rate of germination and seedling growth (Delachiave and De Pinho, 2003). Germination of seeds, one of the most critical phases of plant life, is greatly influenced by salinity (Misra and Dwivedi, 2004). NaCl and Polyethylene glycol (PEG) compounds have been used to simulate osmotic stress effects in Petri dish (in vitro) for plants to maintain uniform water potential through out the experimental period (Kulkarni and Deshpande, 2007). PEG is the best solute that we are aware of for imposing a low water stress that is reflective of the type of stress imposed by a drying soil (Verslues and Bray, 2004; Verslues et al., 1998; Van der Weele et al., 2000). On the basis of the results many research, NaCl as compared with PEG had more effect on germination and early seedling stage (Gholamin and Khayatnezhad, 2010; Farsiani and Ghobadi, 2009; Mohammadkhani and heidari, 2008). The principal aim of the present study was to compare the effects of drought and salt stress induced on germination and early seedling stages of two cultivars of maize.

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Stress level	PEG (g/lit)	NaCl (g/lit)
-2	114	2.34
-4	158	4.68
-6	189	7.01
-8	212	9.35
-10	232	11.69

Table 1. Value PEG and Nacl for stress levels.

Table 2. Analysis of variance on mean of square germination and seedling growth in drought and salinity stresses.

Source of variance	Df	Germination (%)	Mean germination time	Germination rate	Length of radicle (cm)	Length of plumule (cm)	Length of seedling (cm)
Drought levels	5	80.005*	0.539ns	0.007ns	57.042**	21.333**	129.312**
Cultivar	1	662.891**	4.121ns	0.029*	125.627**	0.030*	154.878**
Drought levels× cultivar	5	98.202*	0.244ns	0.003ns	7.437**	1.280*	7.149*
Error	24	42.603	0.419	0.004	0.935	0.377	2.512
Salinity levels	4	1554.05**	10.92**	0.165**	104.59**	27.54**	224.56**
Cultivar	1	2447.12**	2.75*	0.12**	15.08*	0.12*	14.76*
Salinity levels× Cultivar	4	252.8*	2.06*	0.04**	3.67*	1.18*	4.34*
Error	20	66.64	2.14	0.007	3.22	0.84	4.53

ns, * and **:non significant, significant at 5 and 1% probability levels, respectively.

MATERIALS AND METHODS

In two separated experiments, effect of drought and salt stresses induced by different osmotic potential levels [(distilled water) 0, -2, -4, -6, -8 and -10 bar] of (PEG 6000) and NaCl treatments on germination and early seedling development of maize were studied (Table 1). Two cultivar of maize including SC704 and SC666 were used. This investigation was performed as factorial experiment under CRD with three replications at seed laboratory, Islamic Azad University Shoushtar Branch in Iran in 2011. In each experiment and each level of stress, twenty seeds of any cultivar were selected and sterilized in sodium hypochlorite (1%) and then washed in distilled water for two times. The seeds of both cultivars were germinated in Petri dishes on 2 layers of filter paper in an incubator maintained at 25°C. Daily, germination rate was measured and need to replace the filter papers and add the PEG and NaCl soluble were performed. Seeds were considered germinated when the emergent redicle reached 2 mm length. After 7 days, germination percentage was measured by ISTA (International Seed Testing Association) standard method. At the end of the seventh day, the germination percentage, mean germination time (MGT) (Ellis and Robert, 1981), germination rate, the length of radicle and plumule of seeds and length of seedling were also measured.

Formula 1:
$$GP = \frac{SNG}{SNO} \times 100$$

Where, GP is the germination percentage; SNG is the number of germinated seeds and SN0 is the number of experimental seeds with viability (Scott et al., 1984).

Formula 2:
$$GR = \frac{\sum N}{\sum (n \times g)}$$

Where, GR is the germination rate; n is the number of germinated seed on gth day and g is the number of total germinated seeds (Ellis and Robert, 1981). Osmotic potentials of PEG 6000 were calculated as described by Michael and Kaufman (1973). For statistical analysis the data of germinating pecentage were

transformed to
$$\arcsin \sqrt{\frac{X}{100}}$$
 . It should noted that in this

experiment salinity level -10 bar NaCl salt was also considered, but the data not harvestad at this level and the -10 bar salt level was removed. Analyses were done using the MSTAT-C software (Michigan State University). Differences between means were determined by Duncan's Multiple Range Tests (DMRT) at probability level 5%.

RESULTS

Analysis of variance showed that there were significant difference between genotypes, stress levels and their interaction for both experiments, except for mean germination time and GR traits in drought stress (Table 2). Mean comparison results indicated that for Table 2 all traits except mean germination time in drought stress

Table 3. Comparison of means simple effects of cultivar and drought stress levels on germination and seedling growth.

Treatment	Germination (%)	Mean germination time (day)	Germination rate	Length of radicle (cm)	Length of plumule (cm)	Length of seedling (cm)
SC 704	37.85 b	3.18 a	0.32 ab	3.41 b	2.43 b	5.84 b
SC 666	46.43 a	2.50 a	0.38 a	7.14 a	3.00 a	9.99 a
0	46.93 a	2.62 a	0.397 a	10.84 a	5.75 a	16.26 a
-2 bar	45.33 ab	2.77 a	0.367 ab	6.42 b	3.24 b	9.65 b
-4 bar	42.72 ab	2.80 a	0.365 ab	4.87 c	2.51 c	7.37 c
-6 bar	41.71 ab	3.04 a	0.354 ab	4.03 cd	2.40 c	6.43 c
-8 bar	39.84 ab	2.49 a	0.327 ab	2.15 e	0.88 d	3.93 d
-10 bar	36.82 b	3.32 a	0.302 b	3.33 d	0.49 d	3.82 d

Means with similar letter(s) in each trait is not significantly different at 5% probability level according to Duncan's multiple range test.

Table 4. Comparison of means interaction effects of cultivar and drought stress levels on germination and seedling growth.

Treatment		Germination (%)	Mean germination time (day)	Germination rate	Length of radicle (cm)	Length of plumule (cm)	Length of seedling (cm)
	0	41.01 bc	3.02 ab	0.343 abc	8.85 b	5.67 b	14.52 b
	-2 bar	38.59 bc	2.88 ab	0.347 abc	3.33 de	3.83 bc	7.17 ef
00 704	-4 bar	37.39 bc	3.23 a	0.307 bc	1.90 ef	2.08 c	3.98 gh
SC 704	-6 bar	36.99 c	3.40 a	0.325 bc	2.50 de	2.17 c	4.67 fgh
	-8 bar	36.99 c	3.12 ab	0.327 bc	0.567 f	0.57 d	1.133 i
	-10 bar	36.12 c	3.42 a	0.293 c	3.28 de	0.28 d	3.567 hi
	0	57.74 a	2.22 ab	0.450 a	12.83 a	6.1 a	18.00 a
	-2 bar	49.66 ab	2.65 ab	0.387 abc	9.50 b	3 bc	12.15 bc
SC 666	-4 bar	47.35 abc	2.37 ab	0.423 ab	7.83 b	2.97 bc	10.77 cd
	-6 bar	46.05 abc	2.68 ab	0.383 abc	5.57 c	2.65 c	8.20 de
	-8 bar	42.68 bc	1.87 b	0.327 bc	3.73 d	2.64 c	6.73 efg
	-10 bar	35.10 c	3.22 a	0.310 bc	3.38 de	0.69 d	4.07 gh

Means with similar letter(s) in each trait is not significantly different at 5% probability level according to Duncan's multiple range tests.

and length of plumule in salinity stress, SC666 cultivar had more than SC704 cultivar (Tables 3 and 5). Also, the highest of this trait were in the control level of PEG and NaCl (Tables 3 and 5). The variability trend of germination is more severe and in primary levels of stress, decrease amount is significant. The mean germination time did not change with a decrease in the osmotic potential in PEG experiment (Table 3). In the -10 bar potential of NaCl germination for two cultivar not showed, therefore experiment was deleted. The results of Tables 4 and 6 show that the germination is inversely affected to the NaCl and PEG concentrations, it means that SC666 and SC704 cultivars of maize showed a reduction in germination with an increase in NaCl or PEG concentrations induced water deficit (Tables 3, 4, 5 and 6), but this reduction in NaCl treatment were higher than PEG treatment. At treatment by NaCl, no germination occurred at -10 bar in two cultivars and germination was very low at -6 bar in SC704 cultivar and was very low at - 8 bar in SC666 cultivar (Table 6). Mohammadkhani and Heidari (2008) reported that at treatment by NaCl no germination occurred at -10 bar in SC301 cultivar and -17.6 bar in SC704 cultivar. Farsiani and Ghobadi (2009) showed that germination percentage and germination rate in SC403 cultivar was more than SC704 cultivar and SC704 cultivar no germination occurred at -6 bar by NaCl.

The mean germination time increased with a decrease in the osmotic potential in NaCl solution, but in PEG solution not changed (Tables 3 and 5). In NaCl treatments, the mean germination time was delayed by stress conditions. Compared to PEG, mean germination time for NaCl was higher at an equivalent osmotic potential (Tables 4 and 6). Alebrahim et al. (2008) reported that with a decrease in the osmotic potential in PEG and NaCl solutions, the mean germination time in lines of MO17 and B73 increased. Mostafavi (2011) with study on 7 genotypes of safflower reported that the mean

Table 5. Comparison of means simple effects of cultivar and salinity stress levels on germination and seedling growth.

Treatment	Germination (%)	Mean germination time (day)	Germination rate	Length of radicle (cm)	Length of plumule (cm)	Length of seedling (cm)
SC 704	23.37 b	2.38 b	0.25 b	3.91 b	3.94 a	7.68 b
SC 666	43.16 a	3.04 a	0.38 a	5.47 a	3.73 a	9.21 a
0	46.93 a	1.6 bc	0.448 a	10.84 a	3.05 a	16.26 a
-2 bar	38.04 ab	2.3 ab	0.397 ab	7.13 b	2.6 a	12.6 b
-4 bar	32.78 bc	2.62 ab	0.315 bc	2.05 c	2.75 b	4.57 c
-6 bar	25.25 c	3.13 ab	0.315 bc	2.17 c	2.39 b	5.3 c
-8 bar	23.32 c	3.9 a	0.272 c	2.27 c	2.18 b	3.5 c

Means with similar letter(s) in each trait is not significantly different at 5% probability level according to Duncan's multiple range tests.

Table 6. Comparison of means interaction effects of cultivar and salinity stress levels on germination and seedling growth.

Treatment	Germination (%)		Mean germination time (day)	Germination rate	Length of radicle (cm)	Length of plumule (cm)	Length of seedling (cm)
	0	36.12 b	3.02 ab	0.34 abc	8.85 b	6.33 a	14.52 ab
	-2 bar	34.52 b	2.53 abc	0.41 ab	5.98 b	4.92 ab	11.07 b
SC 704	-4 bar	28.25 b	3.33 ab	0.35 abc	1.83 c	3.13 c	4.8 c
	-6 bar	9.51 c	1 bc	0.08 d	1.67 c	2.67 c	4.5 c
	-8 bar	8.47 c	2 abc	0.06 d	1.23 c	2.67 c	3.5 cd
	0	57.74 a	2.22 abc	0.45 a	12.83 a	5.17 a	18 a
SC 666	-2 bar	41.57 b	2.07 abc	0.49 a	8.27 b	5.87 a	14.13 b
	-4 bar	41 b	4.45 a	0.46 a	2.27 c	2.07 c	4.33 c
	-6 bar	38.8 b	2.2 abc	0.28 bc	2.67 c	3.43 bc	6.10 c
	-8 bar	37.31 b	4.27 a	0.24 c	1.3 c	2.1 c	3.5 cd

Means with similar letter(s) in each trait is not significantly different at 5% probability level according to Duncan's multiple range tests.

germination time increased with a decrease in the osmotic potential in NaCl solution. The germination rate were decreased by increasing NaCl concentrations (Tables 5 and 6), but germination rate in the experiment by PEG in two cultivars had no significant difference and by decrease in osmotic potential had very low change (Tables 3 and 4). This agrees with the results of Farsiani and Ghobadi (2009) and Khayatnezhad et al. (2010) in maize. Gholamin and Khavatnezhad (2010) in wheat and Mostafavi (2011) in safflower. At the control level in both stress, length of radicle and plumule and length of seedling reached their highest values. For length of radicle and plumule and length of seedling in both experiments except length of plumule trait in NaCl experiment, SC666 cultivar was higher than SC704 cultivar. All other treatments gradually reduced the seedling growing (Tables 3, 4, 5 and 6). The results is in agreement with many researches (Gholamin and Khayatnezhad, 2010; Farsiani and Ghobadi, 2009; Mohammadkhani and heidari, 2008; Jajarmi, 2009; Khayatnezhad et al., 2010).

DISCUSSION

This study concludes that PEG and NaCl adversely affected the germination and seedling growth of maize. In low water stress, NaCl had a greater inhibitory effect in germination and seedling growth than PEG. Distinct genetic differences were found among the cultivars with respect to germination and seedling growth subjected to NaCl and PEG. It seems that SC666 cultivar in drought and salinity stresses condition had more tolerant than SC704 cultivar and had more yield potential. Some studied referred that stress can contribute to improve germination rate and seedling emergence in different plant species by increasing the expression of aquaporins (Gao et al., 1999), enhancement of ATPase activity, ribonucleic acid (RNA) and acid phosphathase synthesis (Fu et al., 1988), also by increase of amylases, proteases or lipases activity (Ashraf and Foolad, 2005). Kramer (1974) reported that the first effect measurable due to water deficit was the growth reduction, caused by the declining in the cellular expansion. The cellular

elongation process and the carbohydrates wall synthesis were very susceptible to water deficit (Wenkert et al., 1978) and the growing decrease was a consequence of the turgescence laying down of those cells (Shalhevet et al., 1995). Water stress due to drought is probably the most significant abiotic factor limiting plant and also crop growth and development (Hartmann et al., 2005). Drought stresses is physiologically related, because induce osmotic stress and most of the metabolic responses of the affected plants are similar to some extent (Djibril et al., 2005). Water deficit affects the germination of seed and the growth of seedlings negatively (Van Den Berg and Zeng, 2006). According to the results of the present study, it is suggested that more experiments were carried out on the similar cultivars and further investigation be done on SC666 cultivar. Results of the current study are in agreement with other experiments in different plants including Farsiani and Ghobadi (2009) and Khavatnezhad et al. (2010) in maize. Almansouri et al. (2001), Soltani et al. (2006) and Gholamin and Khayatnezhad (2010) in wheat and Mostafavi (2011) in safflower.

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