Research Article

THE EFFECT OF SALINITY STRESS ON GERMINATION COMPONENTS OF GRAIN SORGHUM CULTIVARS

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ABSTRACT
In order to investigate the effect of salinity stress on germination characteristics of grain sorghum cultivars, this research was conducted in 2013 in the laboratory. The controlled treatment included three varieties of grain sorghum (Sepideh, Kimia, Payam) as the first treatment and salinity stress using NaCl solution in four levels (0, 4, 8, 12 dS/m) as the second treatment. It was a factorial experiment in the form of randomized complete block design with four replications. The results of the experiment showed that at all levels of salinity stress of 8 and 12 dS/m, growth traits such as germination percentage, mean time to germination, coefficient of velocity of germination, rootlet length, stemlet length, and seed vigor index decreased and a descending trend was observed in them. The highest reduction was related to the salinity level of 12 dS/m and among the sorghum varieties, the best cultivars in terms of germination traits were Kimia, Sepideh, and Payam, respectively. In general, the effect of salinity stress on most of germination related traits of grain sorghum cultivars was significant at 1% level and salinity stress reduced the germination related traits. It can be concluded that salinity stress had an adverse effect on most of the evaluated traits.

Keywords: Germination, Cultivar, Grain Sorghum, Salinity Stress

INTERUCTION
Salinity of water and soil is an important environmental factor restricting the growth and production of plants in arid and semiarid areas (Hajilooei et al., 2009). By the next 25 years 30% and by the mid 21st century, 50% of agricultural lands will be destroyed by salinity which will have a negative impact on agricultural production (Mahajan et al., 2005).

Lack of plants germination in saline soils is often due to high concentration of salt in seed planting area resulting from the upward movement of the soil solution, and consequently the salt accumulation in soil surface (Song et al., 2008). Salts affect the germination process by restricting the amount of available water (osmotic effect) or due to the damage caused by the entrance of ions (ionic effect) into germination metabolic process. Salinity stress is one of the factors preventing germination and establishment of seedlings (Al Mansour et al., 2001). Most salt stresses in nature are associated with sodium salts (Levitt, 1980).

Sorghum is a crop belonging to the millet family (Poaceae) which is suitable for producing grain and forage in the summers in arid and semiarid areas and it can be used for reclamation of saline soils in irrigated agriculture (Evans, 2006).

Compared to the other salt tolerant plants sorghum is placed after atriplex, barley, and sugar beet (Fooman et al., 1992). Chauhan et al., (2012) investigated the salt tolerance of 13 sorghum varieties and reported that as the salinity concentration increased, germination and growth of seedling decreased in all cultivars. All cultivars germinated at all levels of salinity but the highest and the lowest germination percentage respectively belonged to CSV-15 and Pant-1 cultivars at salinity levels of 10 and 12 EC. Salinity stress significantly reduced the rootlet length, stemlet length, and seedling dry weight in sorghum cultivars.

Ramdan et al., (2001) reported that as the salinity increased from 3.6 to 11.6 mmhos /cm, germination percentage decreased from 80% to 12% in maize, from 62% to 2% in soybean, and from 87% to 22% in sorghum. As the salinity level increased, the dry weight of rootlet and stemlet reduced significantly. Examining the effect of salinity on germination rate and percentage and also on the growth of rootlet and stemlet in many crops has shown that salinity stress at germination stage is a reliable test for evaluating
the salinity tolerance of many species because salinity causes the decrease of germination rate and percentage and also the reduction of rootlet and stemlet growth (Munoz, 2002). Due to excessive increase of the world population and the increasing demands for food and the forage supply for the cattle, finding some varieties of grain sorghum which have short growth period and are resistant to salinity and also provide a part of human, livestock, and poultry food is a good way to increase its production in arid and salty lands and environments (Imam, 2004). Since the common and classic evaluations of salinity tolerance in field conditions and based on final performance assessments are time consuming on one hand, and influenced by various uncontrollable factors such as soil, climate, and agricultural operations on the other hand, it is necessary to provide conditions for the quick and relatively accurate evaluation of plants response to stress using a laboratory approach under controlled conditions (Netonad et al., 2004).

MATERIALS AND METHODS
The experimental research was conducted in 2013 in the specialized laboratory of the faculty of agriculture in Ahvaz Azad University. It was a factorial experiment in the form of randomized complete block design with four replications. The first factor included three varieties of grain sorghum (Kimia, Payam, Sepideh). The second factor included four levels of salinity (0, 4, 8, and 12 dS/m) using NaCl solution. In order to experiment the seeds germination, the standard germination method was used (ISTA, 1996). In order to disinfect the seeds they were placed in the solution of sodium hypochlorite 10% for 30 seconds. Then the seeds were washed three to five times with distilled water. Glass Petri dishes (9 cm in diameter) were used for the experiment. In each Petri dish one Whatman filter paper No. 1 was placed and 10 ml of water or the solution of the salt was poured in it. Then 20 disinfected seeds were added to the Petri dishes medium. Petri dishes were transferred to the germinators that were set at 25°C. In order to measure the seeds germination after placing them in Petri dishes and applying salinity stress on them since the second day, germinated seeds were counted every day until the 7th day. A seed is germinated when the length of its rootlet is 2 mm. in this experiment the following traits were evaluated:
The germination percentage was calculated by the following formula (Scott et al., 1984):

\[
\text{Germination percentage} = \frac{\text{number of germinated seeds during the experiment period}}{\text{total number of planted seeds}} \times 100
\]

The rate of germination was calculated via the following formula (Snintezi et al., 2000):

\[
\text{Rate of Germination} = \frac{\sum n \sum (d \times n)}{n \times \sum n}
\]

The mean time for germination (MTG) was calculated through the following formula (Ellis and Robert, 1981):

\[
\text{MTG} = \frac{\sum (n \times d)}{\sum n}
\]

The coefficient of velocity of germination (CVG) was calculated via the following formula (Hagoir, 1962):

\[
\text{CVG} = G_1 + G_2 + G_3 + ... + G_n / (1 \times G_1) + (2 \times G_2) + (3 \times G_3) + ... (n \times G_n)
\]

The seed vigor index (SVI) was measured through the following formula (Hagoir, 1962):

\[
\text{Seed Vigor Index (SVI)} = \frac{\text{number of germinated seeds until the lasting counting day}}{\times \text{seedling length}}
\]

The analysis of variance was done based on factorial experiment and in the form of a randomized complete block design. The Duncan's test at 5% probability level was used to compare the means and data analysis was done using SAS software and the diagrams were drawn by Excel 2007 software.
RESULTS AND DISCUSSION

Germination Rate and Percentage

The ANOVA results (Table 1) show that the effect of salinity stress on germination rate and percentage was significant at 1% probability level. Sarmad (2001) showed in a research that the increase of salinity significantly reduced the rate of sorghum germination. The most important interactive effects of water stress and salinity conditions include the different pattern of proteins synthesis, delay in the emergence of embryonic tissues and reduction of germination rate and germination (Shinha and Gupta, 1982). The decrease of germination of grain sorghum cultivars at the level of 12 dS can be related to the increase of osmotic pressure of the solution and consequently insufficient water absorption for germination on one hand, and the penetration of Na+ and Cl- into the seed tissue and disruption of metabolism and thus the increase of the leakage of intracellular materials out of the cell, on the other hand. The negative effect of salinity on germination percentage of sorghum has been reported in different experiments (Malival and Palival, 2002).

Table 1: The ANOVA results of measured traits including germination percentage, mean time to germination, coefficient of velocity of germination, rootlet length, stemlet length, seed vigor index based on the mean of squares

<table>
<thead>
<tr>
<th>Sources of variations</th>
<th>Degree of freedom</th>
<th>Germination percentage</th>
<th>Mean time to germination</th>
<th>Coefficient Of velocity germination</th>
<th>Rootlet length</th>
<th>Stemlet length</th>
<th>Seed vigor index</th>
</tr>
</thead>
<tbody>
<tr>
<td>Variety (v)</td>
<td>3</td>
<td>183.94**</td>
<td>0.05538**</td>
<td>19.468*</td>
<td>1.1526**</td>
<td>1.5700**</td>
<td>156.9**</td>
</tr>
<tr>
<td>Salinity (s)</td>
<td>2</td>
<td>187.95**</td>
<td>1.07007**</td>
<td>23.271*</td>
<td>14.6361*</td>
<td>5.6912**</td>
<td>7258.2*</td>
</tr>
<tr>
<td>variety x salinity</td>
<td>6</td>
<td>2.01ns</td>
<td>0.04901*</td>
<td>0.565ns</td>
<td>*</td>
<td>0.0285ns</td>
<td>*</td>
</tr>
<tr>
<td>error</td>
<td>47</td>
<td>19.48</td>
<td>0.1560</td>
<td>4.891</td>
<td>0.0388ns</td>
<td>0.04</td>
<td>1.7 ns</td>
</tr>
<tr>
<td>coefficient of variations (cv)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

*and ** respectively mean significant at 5% and 1% level; ns means the difference is not difference

Table 2: Mean comparison of the simple measured traits including germination percentage, mean time to germination, coefficient of velocity of germination, rootlet length, stemlet length, seed vigor index based on the mean of squares

<table>
<thead>
<tr>
<th>treatments</th>
<th>Germination percentage</th>
<th>Mean time to germination</th>
<th>Coefficient Of velocity germination</th>
<th>Rootlet length</th>
<th>Stemlet length</th>
<th>Seed vigor index</th>
</tr>
</thead>
<tbody>
<tr>
<td>Salinity</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Control 4</td>
<td>87.0975a</td>
<td>1.745b</td>
<td>40.4708a</td>
<td>3.8658a</td>
<td>3.1058a</td>
<td>78.3883a</td>
</tr>
<tr>
<td>8</td>
<td>76.8733b</td>
<td>1.542c</td>
<td>37.8850ab</td>
<td>3.2242ab</td>
<td>2.6108b</td>
<td>58.7400b</td>
</tr>
<tr>
<td>12</td>
<td>71.5450c</td>
<td>1.792ab</td>
<td>34.0283b</td>
<td>2.1167b</td>
<td>2.0267ab</td>
<td>45.3540c</td>
</tr>
<tr>
<td>variety Kimia</td>
<td>63.3592d</td>
<td>2.249a</td>
<td>29.7233c</td>
<td>1.3983c</td>
<td>1.5233c</td>
<td>19.8025d</td>
</tr>
<tr>
<td>sepideh</td>
<td>77.9663a</td>
<td>1.607b</td>
<td>38.2894a</td>
<td>2.8963a</td>
<td>2.6075a</td>
<td>53.7819a</td>
</tr>
<tr>
<td>payam</td>
<td>74.9888b</td>
<td>1.819b</td>
<td>35.2606b</td>
<td>2.6931ab</td>
<td>2.3575ab</td>
<td>50.4000ab</td>
</tr>
</tbody>
</table>

*: According to Duncan's test the means with similar letters in each column are not significantly different at 5% level
Mean Time to Germination and Coefficient of Velocity of Germination

The effect of different levels of salinity on the mean time to germination was significant at 1% probability level and also different cultivars of grain sorghum were significantly different at 1% level. The highest mean time to germination belonged to salinity level of 12 dS by 2.249. Also, the highest mean time to germination belonged to Payam cultivar by 2.069. The other cultivars (Kimia and Sepideh) were not significantly different from each other at all levels. Esmaeili Poor and Mojadam (2009) reported that high concentrations of salt (150 and 200 mM/l) increased the mean time to germination in sweet sorghum genotypes.

The ANOVA results showed that the effect of different levels of salinity on germination rate was significant at 5% level and different varieties of grain sorghum were significantly different at 5% level. At salinity conditions the highest coefficient of velocity of germination (4708/40 days) belonged to the control treatment. Moreover, the highest CVG belonged to Kimia cultivar by 2894/38 days (Table 4). An important index in evaluating the salinity tolerance of different cultivars is the rate of their germination, so that the cultivars with high germination rate will emerge faster than other cultivars in salinity stress conditions. It seems like that high germination rate in genotypes is due to higher rate of water absorption and their seed inflammation. If the water absorption by the seeds is disrupted or the absorption is done slowly the metabolic activities of germination inside the seeds will be done moderately and thus the time period that root gets out of the seed will increase and consequently, the germination rate will decrease (Abnoos, 2001).

Seed Vigor Index

The ANOVA results showed that the effect of different levels of salinity on seed vigor index of grain sorghum varieties was significant at 1% probability level. The highest seed vigor index by 78/3883 belonged to the control treatment. Moreover, Kimia and Sepideh cultivars had the highest and Payam cultivar had the lowest seed vigor index (Table 3). The decrease of seed vigor under salinity stress condition was reported in sorghum and millet (Khales and Agha, 2007). Low vigor of seed might affect the performance in two ways: first, the percentage of emerged seedlings will be less than the expected level and thus plant density will be lower than the normal extent; second, the seedling growth rate in such plants might be less than the growth rate of plants resulting from strong seeds (Chauhan et al., 2012).

Stemlet Length

The ANOVA results showed that the effects of different levels of salinity and cultivars on stemlet length were significant at 1% level, but the interactive effect of salinity and cultivar on it was not significant. The results showed that the stemlet length was significantly affected by salt concentration. As the salt concentration increased at high levels of salinity, the stemlet length decreased in all cultivars. Comparison of the means indicated that in Kimia and Sepideh cultivars, the means of stemlet length by 2.6075 and 2.3575 cm respectively, were not different significantly while the length of stemlet in Payam cultivar by 1.9850 cm was significantly different from the other two cultivars. The length of stemlet under the effect of different levels of salinity was statistically placed in four different categories and the highest and the lowest stemlet length by 3.1058 and 1.5233 cm respectively, belonged to the levels of 0 and 12 dS/m.

The length of stemlet had a descending trend among the salinity treatments. The decrease of stemlet length in high concentrations of salt has been reported in sanifoin (Bagheri, 1991) which results from preventing the transfer of nutrients from cotyledons to the embryo. Furthermore, as the salinity of solution increases, the water absorption by the seed is disrupted, the secretion of hormones and the activity of enzymes are lowered and consequently the seedling growth (including the rootlet and stemlet) is deficient. Rahimian et al., (1991) reported that the decrease of rootlet and stemlet length in NaCl solution was probably due to the toxicity of ions and their negative effects on the membrane cells.

Rootlet Length

The ANOVA results showed that the effect of different levels of salinity and on rootlet length was significant at 1% level; moreover, the effect of different cultivars of grain sorghum on rootlet length was significant at 1% level but the interactive effect of salinity and cultivar on it was not significant.
Comparison of the means showed that the highest and the lowest rootlet length by 3.8658 cm and 1.3983 cm respectively belonged to the salinity treatments of 0 and 12 dS/m. The highest length of rootlet belonged to Kimia cultivar and the lowest length of rootlet belonged to Kimia cultivar.

As the salinity level increased to 8 dS/m the length of rootlet decreased so that by the increase of salt concentration to 12 dS/m the length of rootlet decreased by 50% compared to the control treatment. Since the salinity affects germination via the increase of osmotic pressure and consequently the decrease of water absorption and also through the toxic effects of ions such as Na and Cl, the reduction of the studied germination indices can be attributed to the decrease of rate and speed of primary absorption of water and also the negative effect of osmotic potentials and ions toxicity on biochemical processes of catabolic phases (enzyme hydrolysis of seed storage materials) and anabolic phases (making new tissues by means of hydrolyzed materials) or disruption in absorption of useful ions or all of them at germination stage (Tadayon and Imam, 2007).

The reports made by Weinberg, Markar and Termet showed that as the salt concentration increased in the seed bed, the absorption of calcium and magnesium by the plants root and stem increased. Khales and Agha (2007) investigated the effect of different levels of salinity on germination in forage sorghum seeds and stated that in salinity stress conditions the weight and length of rootlet and stemlet reduced significantly. In salinity stress conditions, the amount of protein in cell membrane which affects the elongation and the growth of the cell decreases, but some pectin compounds that soften the cell wall will increase. The germinated seeds in saline mediums have shorter stemlets and rootlets and NaCl has an inhibitive effect on the emergence of embryonic tissues more than the other saline materials (Cartridge, 1994). The decrease of water potential at germination medium due to salinity increases the rate of toxicity (Grin, 1980).

**Conclusion**

According to the results obtained from the experiments on grain sorghum, the decrease of traits was observed at salinity level of 12 dS/m. Comparison of the means of cultivars showed that Payam cultivar had the lowest rate in most of the studied traits. It was ranked after Sepideh cultivar. It can indicate the higher sensitivity of Payam cultivar than Sepideh cultivar to salinity stress. In Fact, examining traits such as germination, germination percentage, seed vigor index, length of rootlet and stemlet and the significant superiority of Kimia cultivar in such traits at different salinity treatments can indicate the priority of this cultivar under stress conditions on one hand, and can emphasize the applicability of the studied traits in evaluating salinity tolerant grain sorghum, on the other hand.

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