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Salinity Effects on Germination of Forage Sorghumes

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Abstract: To investigate the effects of osmotic stress on forage sorghum (*Sorghum bicolor* L. Moench) varieties at critical stage of seed germination, i.e., at which salinization is initiated. A study was conducted at the Seed Technology Laboratory at University of Putra Malaysia (UPM- lab.) from October to November 2007. Two forage sorghum varieties (Speedfeed and KFS4) were evaluated in the salinity levels of 0, 5, 10, 15 dS m⁻¹. The experimental design was Completely Random Design (CRD), which was based on factorial with 3 replications. In the experiment, seeds were germinated in covered, sterilized, disposable Petri dishes. Three parameters, namely germination percentage, germination rate, germination index and coefficient velocity of germination were estimated. The results of this study showed that different levels of salinity had significantly affected the germination percentage, germination rate germination index. In addition, a significant difference (1%) was also found in the germination percentage between the two varieties. Meanwhile, the mean comparison of the treatments showed that the germination percentage in distilled water (S1) was the highest (92.5%) and salinity dS m⁻¹ (S4) had the lowest germination percentage. The maximum germination rate (35.58) and germination index (391.67) were retrieved from EC 0 and with the increasing salinity, these parameters were decreased. The evaluation of the two varieties showed that KFS4 (V1) had a higher germination percentage (82.91%) than the speed feed (72.5%). Consequently, the seed germination of KFS4 was better than the speed feed (in salty water and non-stress conditions). In other words, salt stress had been found to delay germination of both varieties.

Key words: Salinity, germination percentage, germination speed, germination index, forage sorghum

INTRODUCTION

Among the stages of the plant life cycle, seed germination is one of the most important key processes in the survival and growth of plants in arid and semi-arid regions (Hadas, 2004). Seed germination and early seedling growth are critical stages for the establishment of plant populations under saline conditions (Khan and Gulzar, 2003). Water stress that occurs during crop establishment and seedling growth is either directly from the low available soil moisture or from osmotic effects associated with salinity. Grasses differ in their upper limit of salinity tolerance and an increase in salinity concentration usually delays and reduces seed germination (Gulzar *et al.*, 2001).

Temperature, which usually increases under salinity and water stress, plays a crucial role in many biological and physiological processes in plants (Al-Ahmadi and Kafi, 2007; Berti and Johnson, 2008). The temperature changes have a major impact on a number of processes which regulate seed germ inability including membrane permeability, activity of membrane-bound as well as

cytosolic enzymes (Tlig *et al.*, 2008) and its interaction with the variable soil water content in surface layers of the soil. Salinity-temperature interaction, in particular, determines seed germination patterns in many salt-affected environments (Al-Khateeb, 2006; Song *et al.*, 2006).

However, abiotic factors, such as water stress brought about by drought and salinity, limiting plant germination and growth during early seedling stages (Mansour, 2000), continues to be a widespread problem around the world (Soltani *et al.*, 2006). Research on responses to salt tolerance of crops at different stages of growth under local climatic, soil and crop conditions is still limited. The degree to which salinity affects germination by an osmotic effect or specific ion toxicity and whether salt tolerance varies in different species is still a subject of study. A good understanding of how salinity affects percentage germination, germination rate, germination index and developmental responses to tolerate salinity will enable the development of new cultivars capable of supporting higher yields under such stress situations. There is therefore, a need for a better

understanding of the effects of salt stress during these important periods of growth. Seed germination has been reported to be the most critical stage influencing crop establishment in areas under saline conditions (Bayuelo-Jimenez *et al.*, 2002) and hence the aim of the current study was to investigate the effects of osmotic stress on two new varieties at this critical stage.

The specific objectives of the study were:

- To compare tolerance level of two forage sorghum varieties to salinity
- To determine the relative salinity and germination responses of the forage sorghum varieties to different levels of salinity

MATERIALS AND METHODS

The experiment was conducted in the seed technology laboratory of Faculty of Agriculture at Universiti Putra Malaysia. The germination experiments were carried out from October to November 2007. The seeds were incubated at laboratory conditions at 23-27°C under light for 12 h daily and were monitored until the end of the germination period of 7 days. The temperature was measured using a thermometer and light intensity was measured with a heavy duty light meter (Extech® Model 407026).

Seeds of the two forage sorghum (*Sorghum bicolor* L. Moench) varieties namely, Speedfeed and KFS4 were supplied by Iran Agriculture Research Organization. The seeds were germinated in covered, sterilized, disposable petri dishes lined with Whatman No. 2 filter paper and moistened with distilled water (control), or 5, 10 and 15 dS m⁻¹ salinity (NaCl concentration) treatments. Three replicates of 20 seeds were used for each treatment. Higher rates of NaCl were included to ensure a range of germination reactions.

Four salt water concentrations namely 0, 5, 10 and 15 dS m⁻¹ were evaluated in this study. The salinity of the salt water solutions were determined using an EC meter (HANNA® Model HI 8733). Untreated checks were irrigated with distilled water. On the 1st day, 3 mL of the respective treatments were applied to each Petri dish and on the 3rd day, another 3 mL were added to maintain adequate moisture.

This experiment consisted of 8 treatment combinations with 2 varieties and 4 salinity levels arranged in a Completely Randomized Design (CRD). There were 3 replicates per treatment.

Four parameters, namely germination percentage, germination rate, germination index coefficient of velocity of germination were estimated. For germination

percentage, only the total number of seeds germinated at the end of experiment was considered. Germination rate was calculated using the following formula:

$$\sum GT_1/T_1 + \dots + GT_n/T_n$$

where, GT is seeds germinated each day and T refers to the day during the trial. The Germination Index (GI) was calculated based on the following formula:

$$GI = (5 \times n_1) + (4 \times n_2) + \dots + (1 \times n_5)$$

where, n1, n2, n3, n4 and n5 are the number of seeds germinated on days 5, 4, 3, 2 and 1, respectively. The Coefficient of Velocity of Germination (CVG) was estimated from the formula:

$$CVG = (N_1 + N_2 + \dots + N_x) / 100 \times (N_1 T_1 + \dots + N_x T_x)$$

Where:

N = No. of seeds germinated each day

T = No. of days from seeding corresponding to N.
(Kader and Jutzi, 2004)

Data were subjected to statistical analysis using Proc ANOVA in the SAS Statistical Software (SAS Institute, 2004). The treatment means were compared by Least Significance Differences (LSD) at the 5% probability level.

RESULTS

The results of this study showed that different levels of salinity significantly affected the germination percentage, germination speed and germination index and coefficient of velocity of germination. Variety and salinity effects on seed germination were significant (Table 1). The germination percentage was significantly reduced with increase in salinity levels. The germination percentage in the distilled water control was 25.5% higher than that of seeds treated at a salinity of 15 dS m⁻¹ (Fig. 1a, b). Comparison between varieties showed that KFS4 had 10.4% higher germination percentage than Speedfeed. The germination rate and germination velocity coefficient decreased with increasing salinity (Fig. 2a, b). The highest germination percentage (92.5%) was in distilled water and the lowest was in the 15 dS m⁻¹ salinity treatment. Evaluation between varieties showed that KFS4 had a higher germination percentage (82.91%) than Speedfeed (72.5%). The seed germination of KFS4 was better than Speedfeed in both salty water and non-stress conditions. Salt stress was found to delay germination in both varieties (Fig. 3, 4a, b).

Table 1: ANOVA on some germination characters of different forage sorghum varieties

Treatment	df	Germination (%)	Germination speed	Germination index	Coefficient variability of germination
Block	2	16.666	0.0749	54.166	23.255
Salinity	3	951.041**	121.420**	14620.48**	0.402**
Variety	1	651.041**	0.296 ^{ns}	2501.04 ^{ns}	4.069**
Variety × salinity	3	48.263 ^{ns}	9.586 ^{ns}	887.152 ^{ns}	0.108 ^{ns}
CV (%)		6.120	8.723	7.405	7.405

ns: No significant, *significant on 5% level and **significant on 1% level

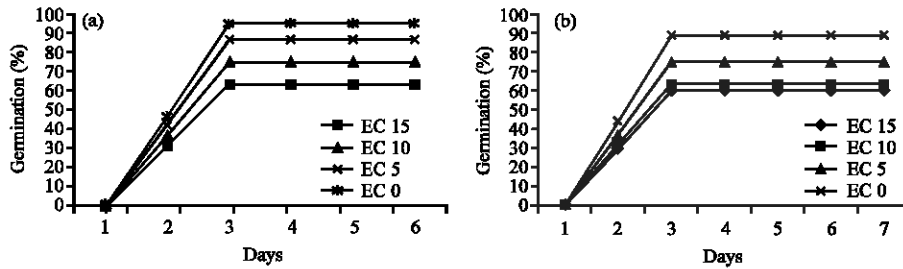


Fig. 1: Effect of salinity on seed germination percentage of forage sorghum varieties. (a) KFS4 and (b) speed feed

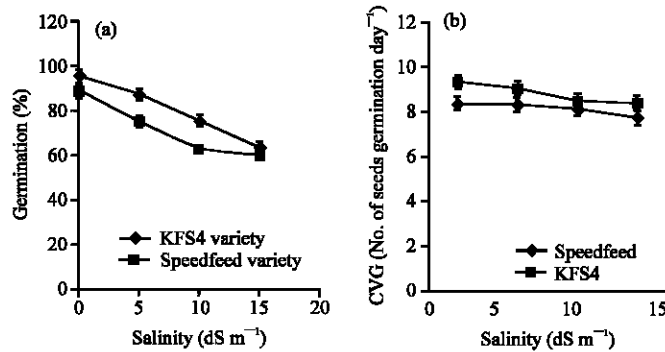


Fig. 2: (a) Effect of salinity on germination percentage and (b) coefficient of velocity of germination of the sorghum varieties tested

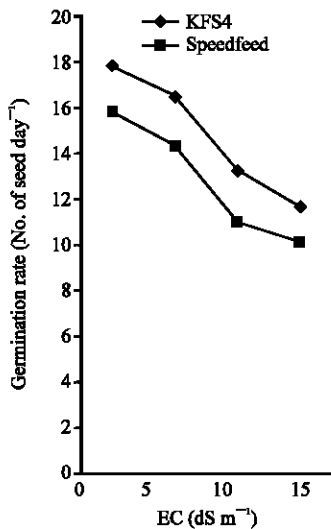


Fig. 3: Effect of salinity on germination rate of KFS4 and speedfeed

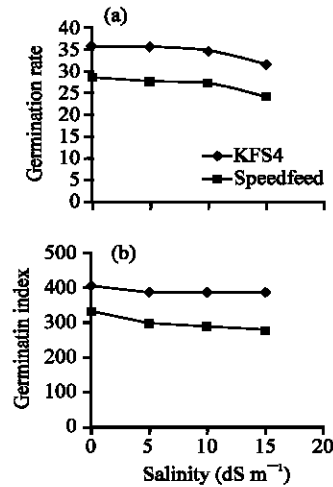


Fig. 4: (a) Effect of salinity on germination rate and (b) germination index of the two forage sorghum varieties

There were no significant differences in germination rate and germination index between the two varieties (Appendix A1). The interaction effects (variety×salinity) were also not significant in all three parameters measured (Appendix A1). The highest germination rate (35.58) and germination index (391.67) were obtained with 0 dS m⁻¹ (distilled water) salinity treatment and with increasing salinity, these parameters were found to decrease (Table 1, Fig. 1-4).

DISCUSSION

Salinity reduced germination percentage and also delayed the germination rate as the salt level was increased. In the field, germination rarely takes place in soils of high salt concentrations. In irrigated agriculture, salt would normally be leached from the surface at sowing and in dry-land agriculture, the crop is normally planted after rain. In salt-affected situations where the crop is sown without rain or leaching irrigation, the soil in the top 10 cm is likely to be sodic as well as saline and the main constraint to emergence might be the hardness of the soil as much as salts in the soil solution. Under such conditions, emergence rate might be a more practical screening criterion than germination rate and seedling vigour may also be a useful screening factor for soils that form hard crusts (Serraj and Sinclair, 2002). However, most crops are capable of germinating at high salinity levels.

In the present study on salinity, the variety effect on percentage germination was found to be highly significant (1%) (Table 2). Similar reductions have been reported on germination and seedling growth in saline soils, but with varying responses between species and cultivars (Hampson and Simpson, 1990). Comparison of treatments

showed that mean percentage germination decreased from 92.5 to 63.0 when salinity increased from 0 to 15 dS m⁻¹. Salinity affects the germination of seeds by creating an external osmotic potential which prevents water uptake. The toxic effects of Na⁺ and Cl⁻ ions on germinating seeds affect uniformity in plant density with negative effects on the yield (Gamze *et al.*, 2004). The results of the present study are in agreement with Munns (2002), where factors that adversely affected seed germination include sensitivity to drought stress and salt tolerance. Both germination and seedling growth are reduced in saline soils (Okcu *et al.*, 2005; Hampson and Simpson, 1990).

The earlier stages of growth are more sensitive to salinity than subsequent ones. The stand, subsequent growth and final yield of crop plants are decreased when the moisture supply is limited (Ghoulam and Fares, 2001). Jacoby (1994) reported that the initial and primary effect of salinity, particularly at low to moderate concentrations, is due to its osmotic effects. However, there were also differences in terms of tolerance to salinity among species and cultivars, as well as among the different plant growth parameters recorded. This is indicated by the two varieties in the present study where KFS4 had a higher germination percentage (82.91%) than speedfeed (72.5%).

However, no significant differences were observed in terms of germination rate and germination index for both varieties. KFS4 had a germination rate and germination index of 30.74 and 346.67, respectively. In addition, the significant interaction effect (variety×salinity) showed that the germination percentage of KFS4 in Distilled water was highest (96.66%), while speedfeed in 15 dS m⁻¹ Salinity had the lowest germination percentage (61.66%). Similar responses were reported by Hester *et al.* (2001), who indicated crop performance decreased significantly

Table 2: Means for germination characteristics of forage sorghum varieties as affected by salinity

Source of treatment	Germination (%)	Germination speed	Germination index	Coefficient variability of germination
Variety				
KFS 4	82.917a	30.743a	346.67a	9.855a
Speedfeed	72.500b	30.521a	326.25a	9.031b
LSD	4.164	2.340	21.82	0.217
Salinity (dS m⁻¹)				
0	92.50a	35.583a	391.67a	9.698a
5	82.50b	33.056a	360.83a	9.578b
10	72.50c	28.194b	312.50b	9.394c
15	63.00d	25.694b	280.83c	9.101d
LSD	6.35	3.565	33.24	0.331
Variety×Salinity				
S1V1	96.667a	35.611a	400.00a	10.251a
S2V1	90.00a	35.556a	383.33ab	10.011ab
S3V1	88.333a	34.583a	383.33ab	9.623bc
S4V1	80.00b	31.528ab	338.33bc	9.534cd
S1V2	75.00b	28.611bc	328.33bc	9.165d
S2V2	65.00c	27.778bc	296.67cde	9.145d
S3V2	65.00c	27.222bc	286.67de	9.145d
S4V2	61.667c	24.167c	275.00e	8.671e
LSD(5%)	9.408	5.286	49.29	0.491

Means within columns within a source (variety, salinity or interaction) followed by the same letter(s) are not significantly different at p<0.05

with increase in salt concentration, but the threshold concentration and the rate of decrease varied with species and ecotypes. There were marked inter-specific differences in crop tolerance towards salinity and within species ecotypes which can tolerate much higher salt concentrations than normal populations do exist. The effects of salinity on plants include ion toxicity, osmotic stress, mineral deficiencies, physiological and biochemical perturbations and combinations of these stresses (Munns, 2002; Hasegawa *et al.*, 2000).

Vincente *et al.* (2004) demonstrated that the reaction to salt stress varied according to the stage of plant development and that a given cultivar might be tolerant at one stage but sensitive at another. Meanwhile, Bayuelo-Jimenez *et al.* (2002) indicated both significant and non-significant associations between tolerance at the germination stage and adult plant growth and development.

The maximum germination rate and coefficient of velocity of germination were found in the low salinity treatment and decreased with increasing salinity, resulting in an average germination rate of 36.5%. Similar results were reported by Munns (2002) and Okcu *et al.* (2005). Jacoby (1994) had also reported differences in tolerance to salinity among species and cultivars.

The results showed that the different germination parameters were affected by salinity and responses varied with salinity levels and variety. The effect of salinity on germination of sorghum is reflected in a lower germination percentage, germination rate, germination index and coefficient of velocity of germination, which may be a result of a combination of osmotic and specific ion effects of Cl and Na (Al-Rwahi, 1989). Limitations at high salinity may also be due to depletion of energy that is needed for growth and loss of turgor (Marcum, 2006).

CONCLUSION

Salt stress reduced and delayed germination and emergence of the forage sorghums. The germination rate, germination index and coefficient of velocity of germination decreased under salinity treatments. The germination percentage was a maximum in distilled water, but decreased with increasing salinity. Seed germination of KFS4 was higher than Speedfeed. Increase in salinity caused corresponding decline in germination. Critical salinity (75% seed germination) for KFS4 variety was observed at 10 dS m⁻¹ salinity, while for Speedfeed the critical salinity was 5 dS m⁻¹. In this study, the relative salt tolerance and competitiveness between the two forage sorghums were determined. The results indicate availability of salinity tolerant alternatives that can be useful in future planning for saline environments.

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