HYDROPONIC SCREENING OF SORGHUM (*SORGHUM BICOLOR* L. MOENCH) CULTIVARS FOR SALINITY TOLERANCE

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ABSTRACT

Sorghum *(Sorghum bicolor* L. Moench*)* crop has been considered relatively more salt tolerant than other cereals and has the potential as a grain and fodder crop in saline soils. However, only a few of the cultivars can thrive under relatively high levels of salinity. Genetic improvement of *Sorghum bicolor* for salt tolerance is of importance due to limited arable land and increasing salinity coupled with population pressure. The objective of this study was to evaluate the salinity tolerance of four selected Kenyan sorghum cultivars (Mtama1, El-gadam, Seredo and Serena) obtained from KARI-Katumani. Seeds of the named cultivars were pregerminated in petri dishes lined with moistened 12.5 mm diameter Whatman filter paper in a germination chamber at 27^0C for 3 days prior to transfer into the hydroponics system using the Shive and Robbin's nutrient solution {constituted of Macronutrients contained in the following salts namely: KH_2PO_4, $Ca(NO_3)_2$, $MgSO_4.7H_2O$, NH_4SO_2 and micronutrients in the following salts: $FeSO_4$, $MnCl_2.4H_2O$, H_3BO_3, $(NH_4)_6$ MO_7 $O_{24}.4H_2O$, $ZnSO_4.7H_2O$, $CuSO_4.5H_2O$} for testing of seedling salinity tolerance. The hydroponics was placed at controlled environmental conditions with supplemental lighting of 4750 lux for twelve hours of day and twelve hours of darkness in the biotron. Four salinity levels were established using different NaCl concentrations corresponding to a nutrient solution electrical conductivity (EC) of 5, 10 and 15dS/m and a control of Shive and Robbin's nutrient solution (0.22dS/m). Shoot length, root length, fresh and dry weights of the seedlings were recorded in order to quantify seedling growth under salinity pressure. The factorial experiment was set up in a CRD. There were significant intercultivar differences in shoot growth (pd"0.01), where Serena had the highest growth at high NaCl concentrations (10 and 15 dS/m) while Mtama1 had the least shoot growth among the four cultivars. Even though increment of salinity level, continued to contribute to growth inhibition at an electrical conductivity above 5dS/m Seredo and Serena showed adaptation to the high levels of salinity as compared to Mtama 1 and El-gadam. Results further indicated that root development (presence of root hairs and root length) was significantly inhibited at 10 and 15 dS/m for both Mtama 1 and El-gadam while Seredo and Serena were less affected.

The sensitivity and tolerance levels in the cultivars suggest that there were two classes of tolerance levels: those that were tolerant and not inhibited in shoot and root growth and those that were sensitive. Based on this study it was concluded that, *Sorghum bicolor* L. Moench cultivars differ in their ability to grow under different levels of salinity during the early seedling growth stages. This is an important characteristic to be taken into account when selecting cultivars that can survive in saline soils.

Key words: Salt tolerance, seedling nutrient screening, electrical, conductivity, sorghum cultivars.

1.0 INTRODUCTION

Sorghum (*Sorghum bicolor* (L) Moench) has been classified into five major and ten intermediate races based on grain and glumes morphology (Harlan and de Wet *et al*., 1972). It has been ranked the fifth in importance among the cereal crops namely wheat, rice, maize and barley. Together with pearl millet (*Pennisetum americanum* (L) and finger millet (*Eleusine coracana* (L) Gaertn, it represents Africa's main contribution to the world food supply (de Vries and Toenniessen, 2001) hence the need to search for its hardy local varieties of crops to match the harsh environmental conditions in arid areas.

Soil salinity is one of the main problems of world agriculture. In the arid and semiarid regions the salinisation process occurs because of incomplete lixiviation and intensive soil evaporation (Ahloowalia *et al*., 2004). This process is characterised as the main factor of world soil degradation (Grieve and Maas, 1984) that affects around 60 to 80 million ha (Grieve and Shannon, 2001). Each year, more and more land becomes non-productive owing to salt accumulation. At least 25% of currently cultivated land throughout the world suffers from excess salinity principally from sodium chloride (NaCl). Crops grown in salt affected soils may suffer from osmotic stress, ion toxicity, and mineral deficiency leading to reduced growth and productivity (Munns, 2002; Netondo *et al*., 2004) and cellular dehydration is a general consequence of osmotic stresses, including water deficit at high salinity levels (Incharoensakdi *et al*., 1986; Robinson and Jones, 1986).

The use of irrigation in attempting to improve productivity in arid areas has posed two problems, i.e., soils slowly becoming oxygen deficient and there's a gradual increase in salinity (Anon., 1982). The major source of increased salinity is irrigation water and high salinity causes osmotic drought which strongly influences water potential and causes toxicity in plants (Munns, 2002).

The present increasing pressure of population growth on food supplies and resources, marginal land and the available water can be utilised by growing crops such as drought and saline-tolerant cultivars of sorghum which fills a unique and highly significant place in drought and salinity tolerance in arid zones compared to maize, wheat and rice (Ayana and Bekele, 2000).

Therefore, as the population increases and more of the dry areas are brought into use, sorghum is bound to become increasingly important. It is often grown in areas of relatively low rainfall, high temperatures and saline soils (Boursier and Lauchli, 1990) but salinity is a threat in arid and semi arid regions (Manju *et al*., 2006).

The quantitative nature of stress tolerance and the problems associated with developing appropriate and replicable testing environments make it difficult to distinguish stress-tolerant lines from sensitive lines. One approach to a better understanding of plant stress tolerance is to isolate those morphological characteristics that are proposed to contribute to stress tolerance and to determine their relative importance (Munns, 2002).

Hydroponics is growing plants with their roots submerged in aerated water and is therefore the best way to separate the effects of aeration, effects due to supply of water and nutrients and those due to mechanical resistance because plant roots need both air and water to survive which are provided in this system.

Plants absorb virtually all of the essential elements through their roots since the macro and micronutrients are dissolved in the water to supply the plants (David, 2004). During growth in hydroponics system accumulation of solutes, either actively or passively, is an important adaptation mechanism for plants in response to osmotic stress hence the objective of this study was to evaluate the salinity tolerance levels of four selected Kenyan sorghum cultivars obtained from the Kenya Agricultural Research Institute (KARI) namely: Mtama1, El-gadam, Seredo and Serena and during seedling growth at 0.22, 5, 10 and 15 dS/m salinity levels.

2.0 MATERIALS AND METHODS

Four cultivars of *Sorghum bicolor* L. Moench (Mtama1, El gardam, Seredo and Serena) obtained from Kenya Agricultural Research Institute (KARI)-Katumani, Machakos were grown in Shive and Robbins oratiry solution culture (Boursier Lauchli, 1990) in a growth chamber. The experiments were conducted at the JKUAT plant physiology laboratory. Seeds were washed in running tap water for 30 min, and germination conducted in the laboratory under controlled conditions at 27^0C. The seeds were grown in 90 mm petri dishes lined with moistened 12.5 mm diameter Whatman filter paper with each petri dish containing 35 uniform seeds of a particular cultivar and placed in a germination chamber at room temperature. The Petri dishes were covered with a polyethylene sheet to avoid the loss of moisture through evaporation. After 3 days, the seedlings chosen for uniformity of both the plumule and radicle length of 0.5 cm were transferred to 4L containers (30 seedlings per container) containing 3litres of Shive and Robbins nutrient solution culture. A hydroponics system in the laboratory was set for the growth of seedlings with provision for aeration whereby the nutrient solution was kept circulating to provide enough gaseous exchange for plant growth. The different levels of salinity were imposed in Shive and Robbins nutrient hydroponics solution using NaCl corresponding to the following Electrical conductivities; 5, 10 and 15 dS/m against a control, i.e 0.22dS/m. Treatment application was done immediately after transfer of the seedlings and the nutrient solution was replaced after every two days. pH adjustment (by use of HCl for lowering pH and NaOH for pH increment) through out the growing season was carried out to maintain suitable root growth environment at pH5.5 . Supplemental lighting of 4750 lux for 12 hours a day was provided by use of fluorescent tubes through out the growing season. A temperature of 28^0C was maintained for the day and 25^0C for the night. The seedlings were allowed to grow in this condition for seven days.

The results of non-destructive daily shoot length measurements during the rapid phase of leaf elongation (five consecutive days starting on the second day after transferring the seedlings into the nutrient solution) were obtained. After 7 days, overall shoot length and root length were measured using a ruler while fresh weight and dry weight of all the seedlings from each treatment were measured by electronic precision weighing balance. Dry weight was obtained after drying the seedlings in the oven at 65^0C for 24 hours and cooled before weighing

3.0 RESULTS AND DISCUSSION

3.1 Root Development

The differences in root development were based on the area within which roots are distributed in the nutrient solution. The cultivars differed significantly in root branching as shown by variations in the number of root hairs and root distribution. The cultivars with greater branching such as cultivar Serena were associated with greater root dry matter, longer roots and smaller shoot: root ratios. The opposite was true for the lesser branched cultivars, e.g. cultivar El-gadam whose roots remained short and thin up to the end of the growth period. Serena and Seredo cultivars which showed adaptation to saline growth conditions had more partitioning of dry matter to roots (relatively high root dry matter) and were found to be more saline-tolerant than Mtama 1 and El-gadam (Table 1). The root hairs were found primarily in the upper portion of the root profile with other characteristics of being deep rooted in the hydroponics solution and greater branching with (Plate 1). These characteristics are associated with the ability of the cultivars to cope with temporal and spatial variability in water status within their environment. As a result, the root systems of cultivars Serena and Seredo cultivars growing in 5 and 10dS/m were simpler in form meaning that, the roots were less branched and had fewer root hairs (Plate 2) than the ones growing in 15dS/m which were able to cope with adverse effects of the high NaCl content in the nutrient solution. In general, as the root system grows and becomes larger (greater total length or mass), branching increases and the root architecture becomes more complex in order to acquire resources. This study indicated that sorghum cultivars differed significantly in rooting characteristics. Based on the results obtained in this experiment it was clear that Serena was the most tolerant cultivar among the four cultivars screened for salinity tolerance while El-gadam was most sensitive to salinity (Plate 2) followed by Mtama 1(Table 1). Salt-induced changes in the plant contributed to growth suppression.

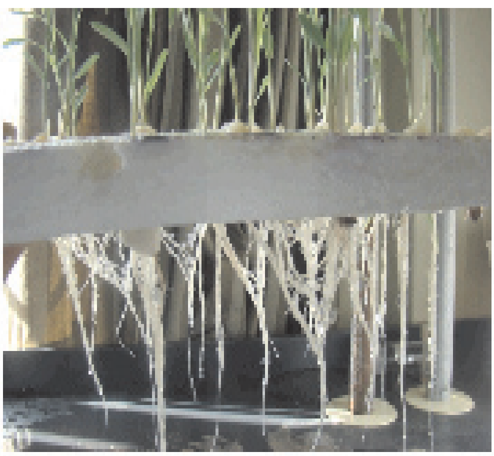

Plate 1: Root morphology of cultivar Serena at 15dS/m

Plants have many adaptive strategies to cope with high salt, soil water deficits and osmotic stress (Masoud *et al*., 2008). Root development (presence of root hairs and root length) as a function of NaCl concentrations provides a useful guideline for salt tolerance (Khan and Gul, 2000). Reduced root elongation was recorded and absence of root hairs was evident at high levels of salinity (10 and 15 dS/m)

Plate 2: Root morphology of cultivar El-gadam at 15dS/m showing inhibition of root hair development at high salinity levels

grown in Shive and Robbin's nutrient solution preceding the root and shoot dry weight comparison, that is, the root: shoot ratio (Table 1).

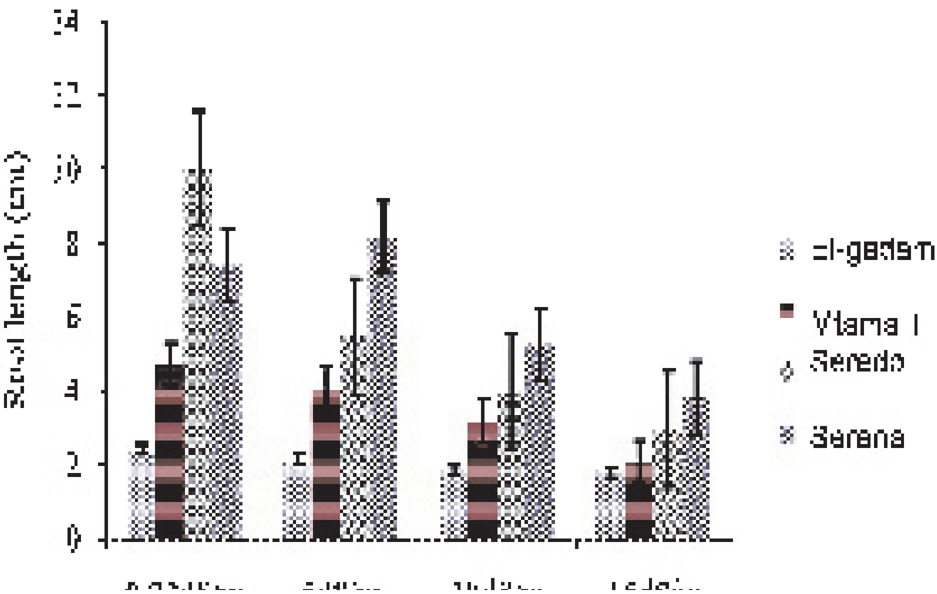

Figure 1: Effect of salinity level on root length of four sorghum cultivars grown in Shive and Robbin's nutrient solution

3.2 Root:Shoot Ratio

The lack of continued seedling growth after seven days in hydroponics solution of highest level of salinity was used to mark the seedling tolerance level at which fresh and dry weights were obtained having observed a significant interaction between cultivars and salinity levels. Seedlings growth rate reduction and damage rate caused to them due to salinity was used as a measure to separate sorghum cultivars that are susceptible or resistant to soil salinity. Salinity level (15dS/m) substantially reduced root length and root hair development (Plate 2). Similarly Lynch and van Beem, (1993) found that roots with an enhanced capacity for growth and branching became long and thick, which enabled greater exploration for water and nutrients. Seven days after growth in the nutrient solution susceptible cultivars had larger root/shoot ratios than tolerant cultivars (Table 1). Studies on the response to salinity have identified an initial phase related to osmotic stress (Phase 1) before the toxic effects of salt reduce growth (Munns, 1993). This is a direct result of the tolerance levels to saline conditions.

Table 1: Shoot dry weight/root dry weight ratio of four sorghum cultivars (the values are means of the three trials)

Cultivar	Treatment 1 Control (0.22dS/m)	Treatment 2 5dS/m	Treatment 3 10dS/m	Treatment 4 15dS/m
Serena	0.9279±0.07	0.8725±0.05	0.9191±0.03	0.8220±0.03
Seredo	0.8784±0.07	0.9608±0.07	0.8868±0.04	0.7340±0.08
Mtama 1	0.9181±0.04	0.9950±0.01	0.8398±0.03	0.8606±0.08
Elgadam	0.9212±0.02	0.9943±0.04	0.9065±0.07	0.9483±0.04
LSD	0.1981	0.723	0.1367	0.1864
CV%	23.1%	12.3%	19.2%	16.2%

3.3 Shoot Development

The ANOVA for all the cultivars for the five days growth in Shive and Robbin's nutrient solution as well as treatment by cultivar interaction was insignificant. The development of shoots differed in the selected four cultivars. Seedling shoot length was enhanced in cultivars Seredo and Serena as compared to Mtama 1 and El-gadam at 10 and 15 dS/m. Except for the tolerant cultivars (Seredo and Serena) an increment in salinity level adversely inhibited growth in the salinity sensitive cultivars (Fig.1. 2). Nevertheless, the influence was more pronounced at 10 and 15 dS/m salinity levels than low salinity levels (5 and 0.22 dS/m). Subsequent growth of the salinity sensitive cultivars resulted in unexpanded chlorotic leaves, while the seedlings of tolerant cultivars were green and fully expanded when exposed to similar salinity levels (Plate 1). Strongest suppression of growth was observed at highest salinity level (15 dS/m). Growth inhibition occurred at an average shoot height of 3.98 cm for salinity sensitive cultivars and at 6.7cm and 8.5 cm for Serena and Seredo respectively (Fig.1.2). The fact that at lower salinity levels (5 dS/m) significant differences were observed, this was attributed to varietal differences in morphological characteristics. The optimal salinity level for hydroponic screening was chosen as 15 dS/m at which all the four cultivars were compared to mark their seedling

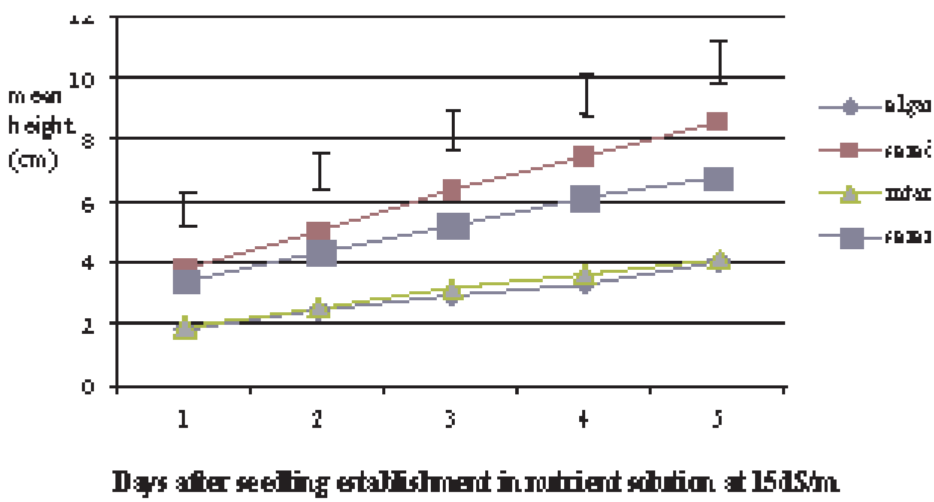

Figure 2: Shoot length of four sorghum cultivars grown in Shive and Robbin's nutrient solution

4.0 CONCLUSION

Water stress is associated with low available water as well as osmotic effects associated with salinity. Seedling root elongation and root dry weight of the susceptible cultivars (Mtama 1 and El-gadam) were adversely affected at high salinity levels (10 and 15 dS/m) and therefore there was very little shoot growth at 10 and 15 dS/m. Seedling root growth was less sensitive to osmotic stress than shoot growth. Seven days exposure to different levels of salinity affected root and shoot growth differently although shoot length was severely affected. Shoot elongation and dry weight were more sensitive to salinity than root growth, with significant reductions in growth occurring at high salinity levels 10 and 15 dS/m. Tolerant genotypes had longer mesocotyls with faster growth rates than the susceptible genotypes. The mechanism involved in salinity tolerance appears to be that of avoidance by fast growth. Plants adapt to water deficits by many different mechanisms including changes in morphology, altered patterns of development as well as a range of physiological and biochemical processes. An increased capacity to maintain growth under osmotic stress could be especially beneficial for high-input crops grown under intermittent irrigation with moderately saline water or under dry land conditions where the level of salinity is low or transient.

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