

Screening sorghum genotypes for salinity tolerant biomass production

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Abstract Genetic improvement of salt tolerance is of high importance due to the extent and the constant increase in salt affected areas. Sorghum [*Sorghum bicolor* (L.) Moench] has been considered relatively more salt tolerant than maize and has the potential as a grain and fodder crop for salt affected areas. One hundred sorghum genotypes were screened for salinity tolerance in pots containing Alfisol and initially irrigated with a 250-mM NaCl solution in a randomized block design with three replications. Subsequently 46 selected genotypes were assessed in a second trial to confirm their responses to salinity. Substantial variation in shoot biomass ratio was identified among the genotypes. The performance of genotypes was consistent across experiments. Seven salinity tolerant and ten salinity sensitive geno-

types are reported. Relative shoot lengths of seedlings were genetically correlated to the shoot biomass ratios at all stages of sampling though the relationships were not close enough to use the trait as a selection criterion. In general, the whole-plant tolerance to salinity resulted in reduced shoot Na⁺ concentration. The K⁺/Na⁺ and Ca²⁺/Na⁺ ratios were also positively related to tolerance but with a lesser r^2 . Therefore, it is concluded that genotypic diversity exists for salt tolerance biomass production and that Na⁺ exclusion from the shoot may be a major mechanism involved in that tolerance.

Keywords Salinity tolerance · *Sorghum bicolor* · Total biomass · Ionic distribution · Shoot Na⁺ content · K⁺/Na⁺ ratio

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Introduction

Salinization is the increase in concentration of total dissolved solids in the soil. Saline soils are estimated to cover about 5–10% of the world's arable land (Szabolcs 1994; Tanji 1990), and the area affected by salinity is increasing steadily, in part due largely to mismanaged irrigation (Ghassemi et al. 1995; Iyengar and Reddy 1994). Soil salinity drastically reduces the productivity of most crops although to a varying extent across species (Francois and Maas 1994; Munns et al.

2002). Besides improving water management practices to reduce the salt accumulation in the root zone, there is a need to improve salinity tolerance of important crops.

Sorghum [*Sorghum bicolor* (L.) Moench], a major grain and forage crop, was previously characterized as moderately tolerant to salinity (Maas 1985; Igartua et al. 1995). It is considered relatively more salt tolerant than maize, the cereal crop ranking first in productivity globally (Maas 1985), and so sorghum has the potential as a crop for salt affected areas (Ayers and Westcott 1985; Igartua et al. 1994). The presence of large genotypic variation for tolerance to salinity reported in sorghum (Taylor et al. 1975; Hassan-ein 1985; Azhar and McNeilly 1987, 1988; Maiti et al. 1994) offers a good scope for integrating tolerance characteristics into appropriate breeding programs to improve crop productivity on saline soils.

Several workers have shown that plant tolerance to high concentrations of salt (salinity) in their rooting medium is under genetic control (Epstein and Jeffries 1964; Epstein 1985; Epstein and Rains 1987; McNeilly 1990; Shannon 1990; Munns et al. 2000). Both additive and dominance effects appear to influence salinity tolerance measured as relative root length (RRL) in sorghum (Azhar and McNeilly 1988) and Na/K ratio in rice (Gregorio and Senadhira 1993) and the heritability values were reported to be low. In fact, QTLs for salt tolerance have been described in several cereals including rice (Flowers et al. 2000; Koyama et al. 2001), barley (Ellis et al. 1997; Mano and Takeda 1997) and bread wheat (Munns et al. 2000; Semikhodiskii et al. 1997) although markers were not robust enough to be used across a range of germplasm. However, the limited success of previous studies was also related to the limited diversity available within the modern cultivars that were used as parents (Munns et al. 2002).

Efforts to enhance crop yields under salinity stress have also had a limited success because available knowledge of the mechanisms of salt tolerance has not been turned into useful selection criteria to evaluate a wide range of genotypes within and across species. Attempts have been made to evaluate salt tolerance at germination

and emergence stages in grain sorghum (Igartua et al. 1994), and large genotypic differences were reported, but this early evaluation appears to have little relation with overall performance under saline conditions (Munns et al. 2002). Though Na⁺ exclusion and grain K⁺/Na⁺ ratios have been suggested to be reliable traits for selecting salt tolerant crops (Munns and James 2003; Munns et al. 2002; Poustini and Siosemardeh 2004; Netondo et al. 2004), the value of that trait has not been used in a large scale. Therefore, there is a need to identify traits associated with salinity tolerance, and simple, high throughput, repeatable screening methods to evaluate large number of genotypes. In fact, the variation in whole-plant biomass responses to salinity was considered to provide the best means of initial selection of salinity tolerant genotypes (Shannon 1984; Ashraf and McNeilly 1987), prior to the evaluation on the basis of specific traits.

In the present study, we first evaluated the genotypic variation for salinity tolerance during the early vegetative stage among a variety of sorghum entries, including currently used breeding lines, based on the proportion of shoot biomass production under saline condition as that of non-saline control in the first 35 days. Then, we investigated possible physiological traits that could be used later on as screening criteria. We also evaluated seed germination and seedling growth as possible indicators of salinity tolerance, and compared these responses to whole-plant reaction to salinity close to anthesis.

Materials and methods

Pot culture screening

In the first pot experiment, 100 entries of sorghum comprising large number of hybrid parents (for grain and forage values) popular and improved varieties, populations and two hybrids hereafter called genotypes, were exposed to NaCl salinity using a randomized block design. Pots of 12.5 cm diameter were filled with 1.2 kg of Alfisol mixed with di-ammonium phosphate at the equivalent rate of 200 kg ha⁻¹ on 29 March 2003, and sealed at the bottom, to avoid salt loss. Two levels of

salinity were applied prior to sowing through a one-time application of deionized water with and without 250 mM NaCl. The amount of water added to bring the soil to field capacity was determined on a soil weight basis (23.2%, w/w). The resulting solution electrical conductivity (EC) was 23.4 dS m^{-1} and the NaCl treated soil EC_{e} was $18.1 \pm 0.19 \text{ dS m}^{-1}$, compared to 2.9 ± 0.26 without NaCl. Irrigation was provided on alternate days up to 20 days after sowing (DAS) and every day at later stages of growth to replace evapotranspirational losses and bring soil moisture levels to field capacity. The water needed for these subsequent irrigations was determined by daily weighing of ten representative pots, to avoid water logging or deficit in the pots. Sixteen seeds of each genotype were sown in each pot in four equally spaced hills. A maximum of four plants per pot were retained after thinning at 10 DAS. One plant per pot was sampled at 18, 25, 32 and 39 DAS. In case a pot had less than four plants, the plants were reserved for the later sampling stage(s), and earlier sampling was skipped. The harvested plants were separated into root (extractable) and shoot, dried in hot air draught oven at 60°C for 3 days and the dry weights were recorded. A ratio of shoot biomass measured under salinity to that of control was calculated replicate-wise for each sample and these ratios were subjected to statistical analysis. This ratio was used as a proxy for estimating the salinity tolerance for biomass production at vegetative stage.

A second pot experiment was conducted with 43 genotypes, including 40 tolerant and 3 sensitive genotypes selected from the first experiment. Experiment 2 was sown on 17 September 2003. The experimental procedure was the same as in experiment 1, except that the pot size was 15 cm diameter and contained 2 kg Alfisol, and that all plants were harvested at the same time at 35 DAS.

Soil and plant assessment

Ionic contents were estimated using the sample harvested at 39 DAS from experiment 1. The pooled shoots (stem + leaves) of all the three replications were used for the determination of N,

P, K, Na and Ca. One hundred and fifty milligrams of finely ground shoot sample was digested in 4 ml of concentrated sulfuric acid with 0.5% selenium powder at 360°C for 75 min on a block digester and the digest was diluted to 75 ml. Using this digest, total N was estimated using SKALAR Auto Analyser, the Netherlands (Krom 1980) to determine whether N absorption has any role in reducing plant growth under saline conditions. Exchangeable K, Na and Ca were estimated (Sahrawat et al. 2002) using an atomic absorption spectrophotometer (Varion model 1200, Australia).

The EC of the NaCl solutions was measured directly using a conductivity meter (Model 1481-50, Cole-Parmer Instrument Company, Chicago). The soil EC was measured using a 1:2 (soil:water, w/v) extract.

Germination studies

The germination of all 100 genotypes included in the first pot experiment was investigated in presence and absence of salinity. Twenty seeds of each genotype were surface sterilized with 1% sodium hypochlorite solution for 10 min, and germinated on filter paper in closed Petri dishes for 6 days in 15 ml deionized water (control) or in 15 ml of a 250-mM NaCl solution in a randomized block design with three replications in a growth chamber at 28/25°C day/night temperature with 12 h light. Five representative seedlings from each Petri dish were used for the measurement of root and shoot length. Relative seed germination (RSG) was calculated as the ratio of the number of seeds germinated under saline conditions to the mean number of those germinated in control, RRL as the ratio of root length under saline conditions to the mean RL of control and relative shoot length (RSL) as the ratio of shoot length under saline conditions to the mean SL of control. These variables were subjected to statistical analysis and the best linear unbiased predictors for each trait were estimated. Correlations and regressions of RSG, RRL and RSL against the shoot biomass ratio observed under different stages of vegetative growth were performed on the best linear unbiased predictors.

Statistical analysis

The data from any individual experiment were analyzed using the following linear additive mixed effects model

$$Y_{ik} = \mu + r_i + g_k + e_{ik}$$

where y_{ik} is the observation recorded on genotype k in replicate i , μ is the general mean, r_i is the effect of replicate i , g_k is the effect of genotype k and e_{ik} is the effect of the error term. The general mean μ and replicate effect r_i were considered as fixed effects. The genotype effect g_k and the error term e_{ik} , were assumed as random effects each with mean 0 and constant variances σ_g^2 and σ_e^2 , respectively. Using the above model, the statistical procedure of residual maximum likelihood was employed to obtain the unbiased estimates of the variance components σ_g^2 and σ_e^2 , and the best linear unbiased predictions (BLUPs) of the performance of the 100 genotypes in the first and 43 genotypes in the second experiment. Heritability was estimated as $h^2 = \sigma_g^2 / (\sigma_g^2 + \sigma_e^2)$. The significance of genetic variability among genotypes was assessed from the standard error of the estimate of genetic variance σ_g^2 , assuming the ratio $\sigma_g^2 / SE(\sigma_g^2)$ to follow normal distribution asymptotically.

The above model was extended for over-experiment analysis of the ratios of 43 genotypes that were common in both experiments, assuming experiment effect as fixed, with genotype \times environment interaction (GEI) effect being a random effect assumed to have a mean

of 0 and constant variance σ_{gE}^2 . The significance of GEI was assessed in a manner similar to that of σ_g^2 . The significance of the fixed effect of the year was assessed using the Wald statistic that asymptotically follows a χ^2 distribution and is akin to the F -test in the traditional ANOVA.

One geometric mean (n th root of the product of n observations) of the shoot biomass ratios was calculated out of the four sample BLUPs for each genotype for the first experiment. This geometric mean of the shoot biomass ratio and the shoot biomass ratio of the second experiment were used for grouping the 43 genotypes into representative groups by a hierarchical cluster analysis (using Ward's ISS method). All the statistical analyses were carried out using Genstat, Release 6.1 (Payne 2002).

Results

Pot culture screening

The genotypic variability for salinity tolerance was assessed in the current study, based on the ratio of shoot (stem + leaf) biomass produced under salinity as that of control. Large genotypic variation was found for the shoot biomass ratio at all stages of crop growth in experiment 1 and at 35 DAS in experiment 2 (Table 1). The heritability values observed for the four samples ranged from 0.36 to 0.46 and there was a trend of increase in these values with increasing age of the plants sampled.

Table 1 Trial means, range of best linear unbiased predicted means and analysis of variance for shoot biomass ratio (shoot biomass under salinity/shoot biomass under

control) for sorghum genotypes sampled at 18, 25, 32 and 39 days after sowing (DAS) in experiment 1 and shoot biomass ratio at 35 DAS in experiment 2

Trait	Trial mean	Range of predicted means	σ_g^2 (SE)	Heritability (h^2)
Ratio of shoot biomass				
<i>Experiment 1 (n = 100)</i>				
18 DAS	0.247	0.091–0.450	0.0111 (0.0026)	0.36
25 DAS	0.314	0.100–0.569	0.0155 (0.0034)	0.42
32 DAS	0.323	0.090–0.706	0.0194 (0.0039)	0.46
39 DAS	0.620	0.302–1.000	0.0317 (0.0072)	0.38
<i>Experiment 2 (n = 43)</i>				
35 DAS	0.090	0.036–0.147	0.0011 (0.0004)	0.36

The hierarchical cluster analysis had yielded five distinct groups at a similarity index of 0.90 and the genotypes in groups with the highest and the lowest shoot biomass ratios were presented in Table 2.

Ion distribution

Shoot Na^+ content under saline conditions was negatively related to the shoot biomass ratio (Fig. 1a; $r^2 = 0.29$, $p \leq 0.001$). This relationship improved further with the mean shoot biomass observed under salinity (Fig. 1b; $r^2 = 0.42$, $p \leq 0.001$). Shoot Na^+ content under control did not show any such relationship either with the shoot biomass ratio or actual shoot biomass under control (data not shown). The overall average shoot Na^+ content under salinity (0.51%) was about two times higher than that under control conditions (0.29%) and ranged from 0.26 to 0.92%.

Shoot K^+ content under saline conditions was not significantly related to the shoot biomass ratio

Table 2 The shoot biomass ratio of sorghum genotypes that clustered under tolerant and sensitive groups based on hierarchical cluster analysis (Ward's ISS method) using the data of experiment 1 (geometric mean of 18, 25, 32 and 39 day ratios) and the 35 day ratio of experiment 2

Genotype	Mean shoot biomass ratio	
	Experiment 1	Experiment 2
Highly tolerant		
CSV 15	0.539	0.098
ICSB 766	0.518	0.087
NTJ 2	0.505	0.109
ICSV 95030	0.536	0.106
S 35	0.524	0.147
ICSB 589	0.501	0.125
ICSB 676	0.502	0.116
Highly sensitive		
GD 65008 Brown	0.352	0.077
ICSB 700	0.356	0.079
PSH 1	0.348	0.082
ICSB 699	0.361	0.072
ICSR 93024-2	0.317	0.086
ICSV 96020	0.337	0.070
ICSV 90017	0.320	0.088
ICSB 405	0.327	0.082
ICSR 170	0.331	0.038
ICSR 56	0.330	0.047

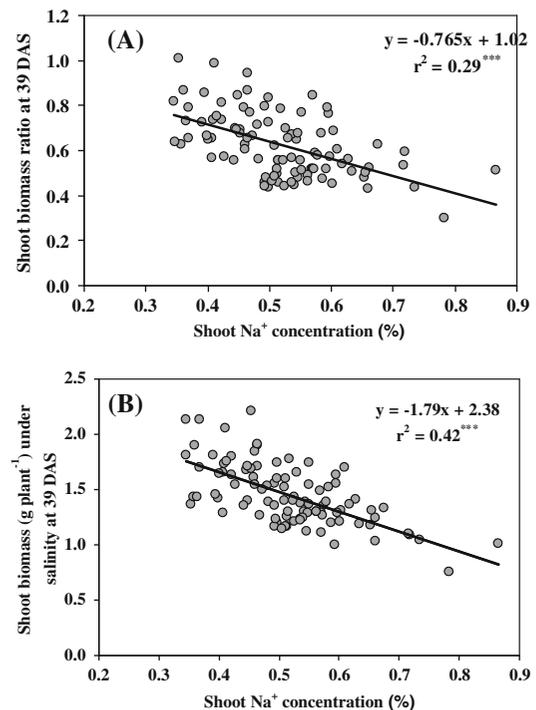


Fig. 1 Relationship of shoot Na^+ concentration (%) with (a) shoot biomass ratio at 39 DAS and (b) shoot biomass under salinity at 39 DAS (triple asterisks significant at 0.001)

($r^2 = 0.03$) whereas it was positively related with the shoot biomass under salinity ($r^2 = 0.13$, $p \leq 0.001$) (data not shown). Unlike the Na^+ content, the mean change in overall mean K^+ content under salinity (1.13%) was not that different from that of the one under control (0.98%). The K^+/Na^+ ratio was significantly and positively associated with the shoot biomass ratio at 39 DAS ($p \leq 0.001$; Fig. 2a). Also this relationship was much closer with the shoot biomass under salinity ($r^2 = 0.31$, $p \leq 0.001$) (data not shown). The overall mean of K^+/Na^+ ratio was about 2.4 under saline conditions, substantially lower than that under the non-saline control (about 3.5).

Ca^{2+} content was not significantly correlated either to the shoot biomass ratio or to the shoot biomass under salinity. In contrast, the $\text{Ca}^{2+}/\text{Na}^+$ ratio was positively related to both shoot biomass ratio ($r^2 = 0.16$, $p < 0.001$; Fig. 2b) as well as the shoot biomass under salinity ($r^2 = 0.33$, $p < 0.001$).

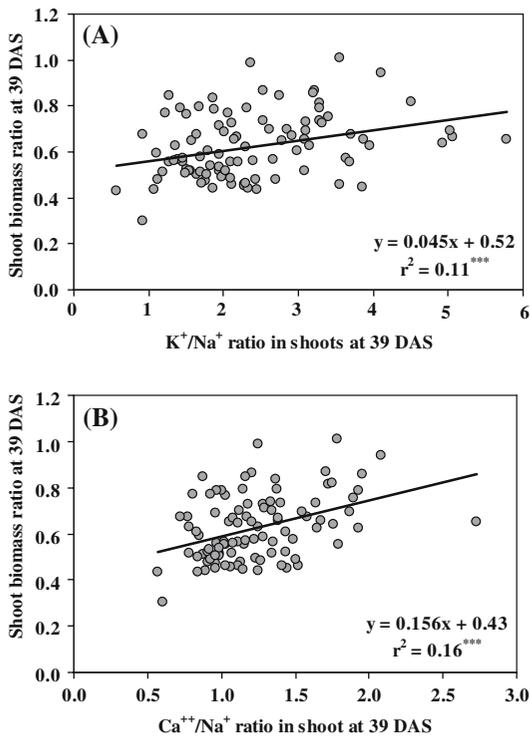


Fig. 2 Relationship between the (a) shoot K^+/Na^+ ratio and the shoot biomass ratio at 39 DAS and (b) shoot Ca^{2+}/Na^+ ratio and the shoot biomass ratio at 39 DAS (*triple asterisks* significant at 0.001)

Under saline conditions, the N concentration of shoots was negatively correlated with the shoot biomass ratio ($r^2 = 0.29$, $p \leq 0.001$; Fig. 3) as well as the shoot biomass under salinity ($r^2 = 0.48$, $p \leq 0.001$) whereas under control

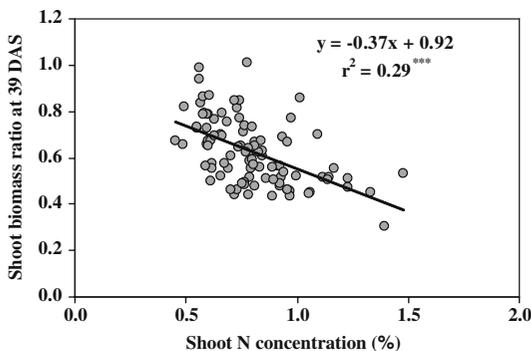


Fig. 3 Relationship of shoot N concentration (%) with the shoot biomass ratio at 39 DAS under saline conditions

conditions this correlation was not significant. This result also indicated that the salinity tolerant genotypes had relatively lower N concentration, varying from 0.5 to 0.9% (Fig. 3). This was likely due to the fact that tolerant plants maintained relatively higher growth rates and thus “diluting” the amount of N taken up, while reduced growth in sensitive genotypes resulted in higher N concentrations in the shoot. In general, N acquisition by plants seems to have been affected under salinity, as indicated by the overall environmental means. The overall mean N concentration under saline conditions was 0.80%, compared to 0.95% in the non-saline control.

Germination studies

Sixteen genotypes that showed <80% germination under control conditions were excluded from the study of the variation in seed germination under salinity and a subsequent relationship between root and shoot growth under salinity at seedling stage with the biomass production at 39 DAS, to avoid confusion between poor seed germination and salt effects on early vegetative growth. There was a significant genotypic variation in the response of germination to salinity measured as the variation in the ratio of germination under salinity to that of control (RSG) (Table 3). However, germination was relatively less affected by salinity as shown by the trial mean for RSG (Table 3). RSG was 30–40% in genotypes ICSR 170, ICSR 56 and M 35-1 indicating that these genotypes are highly sensitive to seed

Table 3 Trial means, range of best linear unbiased predicted means and analysis of variance for the ratio of seeds germinated in 250 mM saline solution as that of control (%) (RSG), ratio of root length under salinity as that of control (RRL) and the ratio of shoot length under salinity as that of control (RSL) in 84 sorghum genotypes

Trait	Trial mean	Range of predicted means	σ^2_g (SE)	Heritability (h^2)
RSG	0.87	0.35–1.04	0.0208 (0.0038)	0.66
RRL	0.233	0.080–0.563	0.0092 (0.0016)	0.85
RSL	0.108	0.013–0.274	0.0038 (0.0006)	0.78

germination under salinity. It was 58–70% in genotypes SP 20666B, ICSB 401 and ICSR 90017 indicating that these were moderately sensitive. Seventeen genotypes, other than the six mentioned above, were significantly (71–83%) less in RSG (data not shown).

Following seed germination, the ratio of root and shoot growth of the seedlings, estimated as length under salinity to that of control, was also adversely affected and varied greatly across genotypes (Table 3). Shoot growth was relatively more affected by salinity than root growth as shown by the overall means and the ranges of these two traits (Table 3). The significance pattern of the genetic correlations, while relating RSG, RRL and RSL of the seedlings with the shoot biomass ratio at 18, 25, 32 and 39 DAS, was largely the same as that of the phenotypic correlation (Table 4). Neither RSG nor RRL was genetically correlated with the shoot biomass ratio at any stage except for a significant phenotypic correlation of RSG with shoot biomass ratio at 25 DAS. However, RSL was generally correlated with the shoot biomass ratio observed at different stages with a probability level range of 0.05–0.01 (Table 4). The correlations coefficients obtained with shoot biomass under salinity instead of shoot biomass ratio were also largely of similar magnitude (data not shown).

Discussion

The main purpose of this study was to assess the range of variation for salinity tolerance of biomass production in sorghum, as a first step to future breeding efforts. We have identified genotypes that are contrasting for their relative biomass production at the early vegetative stage. The most tolerant entries included some elite B-lines, such as ICSB 589, ICSB 676 and ICSB 766, that are regularly used in the crossing program for introgressing various other tolerance characteristics, and a few improved varieties, such as CSV 15, NTJ 2, ICSV 95030 and S 35, that are already cultivated in many parts of the world. Similarly, the highly sensitive entries also included B-lines (ICSB 405, ICSB 699 and ICSB 700), restorers (ICSR 93024-2, ICSR 170 and ICSR 56) and improved varieties (ICSV 90017 and ICSV 96020). We have confirmed the poor value of using an early assessment of salinity tolerance at seedling stage. We have showed some potential in the use of shoot Na concentration as an indirect selection criterion.

Measuring the biomass production at 39 DAS following saturation of the soil to field capacity with a 250-mM NaCl solution has provided an accurate screen for tolerance of the relative biomass production in the early vegetative stage

Table 4 Genetic and phenotypic correlations of the shoot biomass ratios (salinity/control) (SBR) observed at 18, 25, 32 and 39 days after sowing (DAS) with the relative seed

germination (%) (RSG), relative root length ratio (RRL) and the relative shoot length ratio (RSL) in 84 sorghum genotypes

	SBR (18 DAS)	SBR (25 DAS)	SBR (32 DAS)	SBR (39 DAS)	RSG	RRL
Genetic correlation						
SBR (25 DAS)	0.463**					
SBR (32 DAS)	0.437**	0.676***				
SBR (39 DAS)	0.404**	0.531***	0.831***			
RSG	0.249	0.106	0.061	-0.032		
RRL	0.117	-0.052	0.051	0.208	0.194	
RSL	0.341*	0.361**	0.260*	0.347**	0.211	0.467***
Phenotypic correlation						
SBR (25 DAS)	0.248***					
SBR (32 DAS)	0.280***	0.409***				
SBR (39 DAS)	0.231***	0.169*	0.392***			
RSG	0.134	0.152*	0.026	-0.093		
RRL	0.057	-0.028	0.017	0.114	0.143	
RSL	0.186**	0.222**	0.164*	0.204**	0.172*	0.462***

*, **, *** indicates significance at 0.05, 0.01 and 0.001 probability levels, respectively

under saline conditions, and has revealed substantial variation among genotypes. The salt concentration (250 mM NaCl resulting in a soil ECe of $18.1 \pm 0.19 \text{ dS m}^{-1}$) chosen for screening, was similar to that in some previous studies (De La Rosa-Ibarra and Maiti 1995; Yang et al. 1990; Igartua et al. 1994; Netondo et al. 2004). However, few others have also used lower concentrations for sorghum in some other studies (ECe $10\text{--}11 \text{ dS m}^{-1}$) (Maas 1985; Francois et al. 1984; El-Haddad and O'Leary 1994; Igartua et al. 1995). We used this salinity level to cover the salinity-affected soil levels that occur in most sorghum growing areas globally as large number of previous workers has chosen $15\text{--}20 \text{ dS m}^{-1}$ as screening medium for screening large number of sorghum genotypes. The level of salt concentration used in the present study seemed suitable for screening this crop species as only few genotypes could reach a ratio of 0.50 at the maximum productivity stage (39 DAS) in this study under salinity.

The Na^+ concentration in plant shoots under saline conditions appeared to be the trait that was the most closely related to the shoot biomass ratio (29%, $p \leq 0.001$). The use of shoot Na^+ concentration to predict the shoot biomass ratio would certainly deserve more investigation to identify an accurate screen related to the ionic relation in plant under salinity. Sorghum has been considered to be an efficient excluder of Na^+ from aerial plant parts, restricting Na^+ accumulation to its roots (Weinberg et al. 1984; Grieve and Maas 1988). Recent data show that sorghum genotypes accumulate Na^+ in their roots and stems but succeed in excluding most of it from their leaves (Netondo et al. 2004). So, further investigation of the localization of Na in plant part and its possible relation with tolerance is needed. The K^+/Na^+ and $\text{Ca}^{2+}/\text{Na}^+$ ratios were also significantly and positively associated with the shoot biomass ratio, though fairly poorly. In contrast, among various characteristics that was studied only the shoot growth ratio of the seedlings and the shoot biomass ratio at early vegetative stages were related but the magnitude of this relationship was low ($r^2 \leq 0.11$). A germination test would, nonetheless, be useful to discard accessions that are sensitive to salinity at germination stage and thus

would help in limiting the number for the actual screening. Removal of such sensitive material as well as the ones with poor seed viability are expected to decrease the error variances in experimental measurements conducted at later stages of plant growth.

For understanding the genetic control of salinity tolerance as shoot biomass ratio produced under salinity to that of control, the heritability values were estimated for this ratio. These heritability values for salinity tolerance ranged from 0.36 to 0.46 showing that the genetic differences explain a major part of the phenotypic differences. There may be a scope to further improve the screening efficiency for shoot biomass ratio and thereby the operational heritability values by sampling larger numbers of plants at one-time. In relatively more sensitive rice, the heritability values reported were low. The narrow sense (0.198) and broad sense (0.367) heritability values for K^+/Na^+ ratio, at 12 dS m^{-1} culture medium conditions were 0.198 and 0.367, respectively (Akbar et al. 1985).

Conclusions

Overall, it can be concluded that substantial variation in early vegetative stage salinity tolerance among sorghum genotypes was found in this study, and several relatively salinity tolerant and sensitive sorghum genotypes for biomass production were identified. The Na^+ exclusion from the shoot, estimated by shoot Na^+ content or K^+/Na^+ ratio, was well-related to the ratio of shoot biomass, our proxy for salinity tolerance. Further investigation would be needed in that relation to find out a more accurate screen. Seed germination or early seedling growth responses to salinity are not useful as traits for selection of salinity tolerant genotypes as their relationship to shoot biomass ratio were not adequately close.

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