Genetic options for soil salinity-stress management in sorghum

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ABSTARCT

Attempts to breed sorghum for salinity tolerance are limited owing to the complexity in screening for and inheritance of salinity tolerance. Salinity tolerance at germination/seedling emergence and establishment is not correlated with that in later stages. Combining germination/seedling tolerance with that of adult-plant tolerance is necessary to enhance whole-crop tolerance to salinity stress in sorghum. In this article we have reviewed and discussed the screening methods and selection criteria used to breed sorghum for salinity tolerance using conventional tools and the future strategies for genetic improvement of sorghum for salinity tolerance are outlined.

Key Words: sorghum; salinity; screening; genetics; breeding.

INTRODUCTION

Soil salinity among others is an important abiotic constraint for crop productivity in semi-arid tropics (SAT) of the world, where sorghum is cultivated in vast areas for food/feed/fodder uses. Saline soils are those soils with higher levels of soluble salts, such as sulfates (SO_4), carbonates (CO_3) and chlorides (Cl). These soils often exhibit a whitish surface crust when dry (www.cahe.nmsu.edu). The increased demand for sorghum, especially for feed uses driven by

enhanced demand for poultry and meat products in SAT regions (Klieih et al. 2000) imposes extension of sorghum cultivation in saline soils. Though sorghum is known to be relatively more tolerant to soil salinity than maize (Igartua et al., 1994), genetic enhancement of sorghum for salinity tolerance would further increase sorghum productivity in such soils. In this article, we present a short review on the screening methods, inheritance, selection criteria and the limited attempts to breed sorghum for soil-salinity tolerance through conventional tools and to outline the future strategies to genetic improvement of sorghum for salinity tolerance.

MEASUREMENT OF SOIL SALINITY

Usually, salinity is measured in units of electrical conductivity (EC_e) of a standard soil paste extract taken from the root zone of the plant. Electrical conductivities are measured on the saturated extracts or filtered water extracts from these samples in units of deciSemiens per meter (dS m^{-1}) (Shannon, 1997). The soils with ECe<4 dS m^{-1} are considered as non-saline; those with ECe=4 to 16 dS m^{-1} are considered as in saline-phase and those with ECe>16 dS m^{-1} are considered as saline (www.cahe.nmsu.edu).

CROP RESPONSES TO SALINITY

Sorghum crop responses to salinity depend on several factors such as soil, environment and genetic and their interrelationships. Plants grown in high saline soils experience low water potential and mineral toxicity due to high Na^+ concentration in external solution of plant cells. Plants challenged by this water potential develop a large soil-to-leaf gradient of potentials and are unable to meet transpiration demand, and ultimately may wilt and desiccate (www.cahe.nmsu.edu). Water stress is therefore considered a component of salinity injury. Na^+ toxicity results in disturbances in the mineral nutrition of the plant. The cell metabolism is

seriously affected when cytoplasm Na^+ concentration reaches 50 to 100 Mm (Munns et al., 2002). However, under natural conditions, such high salinity level is not experienced by plants at once during the crop growth period. Instead, salinity level increases progressively due to soil drying, which provides ample time for osmotic adjustment of plants to regulate their water uptake. In controlled conditions also, plants adjust osmotically in response to salinity stress, and therefore may overcome the osmotic effect of salt within a few days and symptoms of wilting disappear (Maas and Nieman, 1978).

The water stress and mineral toxicity induced by soil salinity reduces photosynthesis per unit leaf area indirectly through stomatal closure, and to a smaller extent through direct interference with the photosynthetic apparatus (Netondo et al., 2004). At whole plant level, soil salinity stress results in delayed flowering, reduced plant height and grain and fodder yields in sorghum, although the degree of these responses varies with the genetic background of the lines (Francois et al., 1984, Krishnamurthy et al., 2003; Ramesh et al., 2005). However, grain yield may not decrease until a 'threshold' salinity level is reached. The comprehensive survey of responses of different crop species to salinity stress indicated that 'threshold' level of grain sorghum is about 6.8 dS m^{-1} and that grain yield starts declining at 6.8 dS m^{-1} and the reduction are up to 25% at 7 dS m^{-1} and 50% at 10 dS m^{-1} (Maas and Hoffman, 1977).

SOIL SALINITY TOLERANCE

The crop salinity tolerance can be defined as plant growth and economic product (grain/fodder) producing ability in saline soils relative to that in non-saline soils in comparable environments. From an agronomic context, salinity tolerance is described as a complex function of yield decline across a range of salt concentrations (Maas and Hoffman, 1977). Shannon (1997)

has empirically described salinity tolerance (S) as a reduction in yield at a given salinity level (Y_s) with respect to a measured yield under non-saline conditions (Y_e): $S = Y_s/Y_e$. This index may change with the severity and duration of the salinity stress that is imposed.

Mechanisms of tolerance: While osmoregulation, which allows maintenance of turgor, is an important mechanism for tolerance to salinity-induced water stress, Na^+ exclusion from aerial plant parts, restricting Na^+ accumulation to its roots (Weinberg et al., 1984; Grieve & 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). Exclusion of Na^+ from the xylem and compartmentalization in non-vital parts of the plant (apoplast or vacuoles) are other mechanisms to cope with excess salts. The concentration of proline (de la Ibarra and Maiti, 1994) and HCN (Maiti et al., 1994) increases with an increase in salinity levels in tolerant genotypes but not in the sensitive genotypes of sorghum at the seedling stage. Maiti et al. (1994) have reported that presence of a specific protein of 22 Kda molecular weight might confer tolerance to salinity in sorghum. More complete characterization of such a specific protein associated with salinity tolerance would aid in mapping and/or cloning of the genes involved, which could then be deployed in elite agronomic background through marker-assisted selection (MAS) or genetic engineering approaches.

Screening methods: The selection for tolerance under field conditions is usually cumbersome and lack repeatability from season to season. Further, because of the difficulty in assessing the physical and temporal variability in salinity in soil solutions proximal to the root zone, resulting in a large portion of the observed variation in plant growth and yield being due to micro-environmental variation, the selection will be ineffective. It may be possible to compensate for

these problems to some extent by using very large numbers of populations, and replications, as space is not a major limiting factor and by making a judicious selection of breeding strategy and experimental design, and precisely controlling irrigation in artificially salinized plots (Shannon, 1997). A good alternative combination would be to do an early evaluation in controlled conditions and then extend the evaluation of promising lines to field conditions, in large and replicated plots to account for the within-field variability. Two *in vivo* methods have been suggested by Montemurro et al. (1994) for early screening for salinity stress tolerant sweet sorghum genotypes. The methods are: (1) growing the seedlings on sand in polystyrene containers, and (2) growing the seedlings in spectrophotometer cuvettes. Genotypic response to salinity stress was similar in both the methods and classification of sweet sorghum genotypes for salinity tolerance was effectively demonstrated. However, seedling tolerance to salinity may not be translated in adult plant tolerance (Krishnamurthy et al., in preparation, Munns et al., 2002). Considering this, a laboratory screening method is standardized at International Crops Research Institute for the Semi-Arid Tropics (ICRISAT), Patancheru, India. The method allows evaluation of sorghum lines for salinity tolerance at all stages from germination to adult plant stage. In this method, plants are grown in large pots containing 9 kg Alfisol. The salt is applied at a rate of 22.22 g NaCl pot^{-1} basis, which would correspond to a NaCl solution of 250 mM concentration, in sufficient amount to saturate that Alfisol at field capacity. However, to avoid a rapid build up of salt, the quantity of salt is applied in three split doses of 7.40 g NaCl pot^{-1} each time, dissolved at first application in 1.8 L, and subsequently in a sufficient amount to avoid water logging. This method is being followed at International Biosaline Agriculture (ICBA) Dubai and is similar to the one reported by Munns et al. (2002). Two experiments on screening diverse sorghum

genotypes using this method at ICRISAT during 2003 confirmed a rather poor relationship between shoot biomass ratio (biomass under salinity/biomass under control) at 18, 25, 32 and 39 days after sowing (r=0.25, 0.11, 0.06 and -0.03, respectively) and relative germination% (germination under salinity/germination under control) (after discarding the lines with <80% germination) and between shoot biomass yield ratio (yield under salinity/yield under control) assessed at anthesis and stover/grain yield ratio (stover and grain yield under salinity/ stover and grain yield under control) (R^2 = 0.02 to 0.06) (unpublished data). However, a high degree of correspondence between control environment screenings with that of field screening is necessary for effective and routine use of this control environment screening method for genetic enhancement of sorghum for salinity tolerance.

GENETIC VARIABILITY, INHERITANCE AND BREEDING

Genetic variability: Significant genetic variability for salinity tolerance at different stages of crop growth in sorghum have been reported (Igartua et al., 1994; de la Ibarra and Maiti, 1994; Maiti et al.,1994; Peng et al., 1994; de la Ibarra and Maiti, 1995; Krishnamurthy et al., 2003). The research at ICRISAT indicated genetic variability for salinity tolerance at all the growth stages, starting from germination to grain formation in sorghum. For instance, taking 80% germination under non-saline control as a minimum acceptable germination standard for good plant stand required for satisfactory crop productivity, a laboratory study at ICRISAT showed that about half of the test sorghum genotypes with >80% germination under control had <80% germination under induced saline condition (23.4 dSm^{-1}).

Inheritance: Although both additive and dominance gene effects are important in controlling the expression of salinity tolerance at germination and seedling emergence stages, dominance gene

effects appear to be predominant (Igartua et al., 1994). However, relatively high heritability for salinity tolerance (Azhar and McNeilly, 1989) suggests the possibility of rapid improvement in salinity tolerance using high selection pressures and replicated progenies in a hydroponics-based screening system combined with field screening.

Breeding: Reports of attempts to breed sorghum for salinity tolerance are rather limited. A practical problem when breeding sorghum for environments prone to abiotic stresses such as salinity is the choice of optimum selection environment(s) (Igartua, 1995). The evaluation of large numbers of sorghum genotypes across a broad range of salinity levels showed large and significant genotype × salinity level interactions (Azhar and McNeilly, 1987; Krishnamurthy et al., 2003). When confronted with this situation, the plant breeder must decide whether to work over the whole target environment (breeding for wide adaptation), or subdivide it into more homogeneous sub-environments (breeding for specific adaptation) (Igartua, 1995), or work with physiologists to understand some of the reasons for such high genotype × salinity level interactions. In theory, this decision depends mainly on the relative sizes of the genotype × year and genotype × location interactions (Austin, 1993). Unfortunately, the option of breeding for specific adaptation does not exist when working in areas with saline soils, since salinity levels varies concurrently in the same field, and also in the same spot at different times because of increasing salinity levels due to receding moisture. Thus, breeding for saline areas should follow the approach of breeding for wide adaptation (Igartua 1995).

Calhoun et al. (1994) summarized three strategies that address the issue of choice of selection environment for the situations discussed so far: (1) make selection in a stressful environment, (2) select under optimum production conditions, and (3) use a combination of both

the approaches, i.e., select materials that perform well under both stress and non-stress conditions. Igartua (1995) is of the opinion that option (3) is the best. The more environments used, the better will be the results obtained. However, as testing resources are limited, a sensible compromise seems likely to be combination of a non-stress location and at least one stress environment (within the range of target stress environments) between which there is crossover-type genotype × salinity level interaction. Based on the three-component linear equation Maas and Hoffman (1977) and Igartua (1995) conclusively demonstrated the usefulness of the option (3) to select for improved yield in grain sorghum in saline soils.

Considering that good germination (80% is considered minimum acceptable for crop cultivation in non-saline soils), and seedling emergence and establishment are crucial for whole-plant productivity and that these stages are more sensitive to soil salinity toxicity than any later stages (Peng et al., 1994), enhancing the salinity-tolerance at the germination, emergence, and seedling establishment stages should be one of the breeding objectives. Discarding all those genotypes that have <80% germination under saline condition in the laboratory test can routinely be used as the first-stage selection in genetic improvement for salinity tolerance as, such a procedure would help circumvent the problem of confounding effects of inherent poor germination ability with the salt effects on early vegetative growth. However, seedling stage tolerance must be combined with adult plant tolerance to achieve reasonable levels of crop productivity under saline conditions. This is because, normally in farmer's fields, seeds germinate under high moisture, whereby seedlings are exposed to relatively lower salinity stress and as soil moisture depletes in later stages plants are exposed to severe stress. Therefore, the genotypes selected from germination test under salinity stress can then be moved to pot-culture evaluation and field

evaluation for salinity tolerance for grain and fodder yields. Results of pot-culture studies at ICRISAT have shown highly significant positive correlation (r = 0.75**) between total dry matter yield under salinity and salinity tolerance index (ratio of dry matter yield under salinity stress to that under non-salinity) in sorghum, indicating that selection for high dry matter yield under saline conditions, besides improving the yield potential, will also improve salinity tolerance. However, highly significant correlation (r = 0.97**) between the panicle weight ratio (ratio of panicle weight under salinity to that under non-salinity) and grain yield ratio (ratio of grain yield under salinity to that under non-salinity) in sorghum showed panicle weight ratio as a highly effective selection criterion. Research at ICRISAT has also showed that sodium (Na^+) concentration in the shoot was significantly but negatively correlated (r = -0.65**) with total dry matter yield at 39 days after sowing (DAS) in sorghum (unpublished data), indicating that this simple non-destructive ionic concentration measurement can be effectively used to discard those genotypes which are likely to have lower dry matter yield in saline soils.

Igartua and Gracia (1998) assessed the effectiveness of divergent selection for tolerance to salinity at germination-seedling emergence stage in grain sorghum composite population AD11B. A two-cycle recurrent selection for highest and lowest tolerance to salinity for germination and emergence ability based on the laboratory evaluation of S_1 progenies was effective in separating the population (AD11B) into two subpopulations (highly tolerant and least tolerant to salinity). Such selections at seedling and maturity stages are likely to be effective in improving whole plant tolerance to salinity in sorghum. Field emergence in induced saline soils confirmed the effectiveness of this selection procedure to shift the populations in the desired direction.

Krishnamurthy et al. (2003) at ICRISAT have shown that some of the released and popular sorghum varieties that are bred for normal production conditions also exhibited better performance under induced salinity (23.4 dS m^{-1}) relative to non-saline control in a series of pot-culture experiments. Field evaluation of the lines selected from the pot-culture experiments confirmed the superior performance of these cultivars under natural salinity (at 8 dS m^{-1}) in a research station in India and farmers' fields (at 10 dS m^{-1}) in Oman (Ramesh et al., 2005). From the agronomy and plant breeding points of view, cultivars that perform better under both salinity-stress and salinity-free conditions are desirable (Calhoun et al., 1994). Theoretical investigations (Rosielle and Hamblin, 1981) indicating general increase in mean yield in both stress and stress-free environments if selection is practiced for mean productivity (average yield in stress and stress-free environments) lend adequate support to these practical considerations.

CONCLUSIONS

Attempts to breed sorghum for enhanced salinity-tolerance have been limited owing to the complexity in screening for and inheritance of salinity tolerance. Considerable variability in soil salinity levels both spatially and temporally, and within the same field, favors breeding sorghum for wide adaptation. Enhancing the salinity-tolerance of sorghum at the germination-emergence/seedling establishment stages should be combined with adult plant tolerance to achieve reasonable levels of productivity under salinity stress. Considering that *Sorghum halepense* is more tolerant to salinity than *S bicolor* (Yang et al. 1990), *S halepense* may be utilized for genetic enhancement of sorghum for salinity-tolerance. Taking a cue from durum wheat (Munns et al., 2002), it might be worth assessing genetic variability and unravel

inheritance of Na^+ exclusion, an important component trait of salinity tolerance in sorghum (Blum, 1988; Yang et al. 1990), for use in genetic enhancement of sorghum for salinity tolerance.

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