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SCREENING OF LOCAL ACCESSIONS OF *PANICUM MALIACEUM* L. FOR SALT TOLERANCE AT THE SEEDLING STAGE USING BIOMASS PRODUCTION AND ION ACCUMULATION AS SELECTION CRITERIA

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Abstract

In order to assess inter-cultivar variation for salt tolerance in *Panicum maliaceum* L., 18 local accessions were screened at the seedling stage by growing them at 0 and 150 m*M* NaCl in sand culture. A great magnitude of variation for salt tolerance was observed in this set of germplasm. Accessions 008211, 008214, 008220 and 008226 were found to be salt tolerant on the basis of their growth performance at the seedling stage. These tolerant lines accumulated lower amount of Na^+ and maintained higher K^+/Na^+ in their shoots as compared to the sensitive lines. Although a great magnitude of variation for salt tolerance was observed while screening the 18 accessions at the seedling stage, it is not certain whether such variation is detectable at the later growth stages. This needs to be further investigated.

Introduction

Salinity is an ever increasing threat to agriculture as it can substantially reduce plant growth and crop productivity (Ashraf, 1994; 2004; Flowers, 2004; Munns *et al*., 2006). However, different perspective strategies have been listed by various plant scientists to improve crop salt tolerance that will result into enhanced productivity on salt affected soils. Improving salt tolerance of crop and grass species requires access to new genetic diversity and efficient selection criteria (Munns & James, 2003; Ashraf, 2004; Ashraf & Harris, 2004; Munns *et al*., 2006). In this regard, the availability of genetic variation at inter and intra-varietal levels is a pre-requisite (Ashraf, 1994).

Seed germination and seedling growth are known to be more sensitive to salt stress compared with later growth stages in most plant species (Ashraf *et al*., 1986; 1994). Thus, screening of different accessions of a species at the germination stage may lead to find out salt tolerant individuals at early growth stages provided if the degree of salt tolerance of a crop remains consistent throughout its life cycle. Furthermore, glasshouse based screening at germination stage or seedling stage would be efficient in exploring genetic variation. However, its implication to identify salt tolerant germplasm on the basis of germination percentage and survival rate is not feasible (Munns & James, 2003), particularly when the reason for salt-induced reduction in germination percentage or seedling growth is not clearly known, because it could be due to salt-induced osmotic stress (Neumann, 1997), Na^+ toxicity, or nutritional imbalance (Ashraf, 1994; 2004). In addition, the degree of salt tolerance varies with the change in growth stages in most crops e.g., rice (Akbar & Yabuno, 1974), wheat (Kingsbury & Epstein, 1984; Ashraf & Khanum, 1997) and tomato (Foolad & Lin, 1997). Thus, it is necessary to assess salinity tolerance of a crop at different growth stages (Ashraf, 1994; Munns *et al*., 2006; Munns, 2007).

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In long-term experiments to assess genotypic variation for salt tolerance in crops, different scientists use agronomic, biochemical or physiological selection criteria (Ashraf, 1994; 2004; Munns & James, 2003; Munns, 2007), e.g., root length (Ashraf *et al*., 1986), biomass production (Ashraf, 1994), accumulation of proline or glycinebetaine, osmotic adjustment, photosynthetic capacity etc., (Ashraf, 2004; Ashraf & Harris, 2004). The genus *Panicum* is one the largest genera of grasses, including more than 400 species (Roshevits, 1980). Within this large genus, two species viz., *Panicum maliaceum* and to a lesser extent *Panicum miliare* are of economic importance. In view of its great importance as fodder, human feed, seedbird (Martin *et al*., 1976), the present study was conducted to assess the variation for salt tolerance in a set of diverse accessions of proso-millet (*Panicum maliaceum* L.) at the seedling stage using biomass production and ion accumulation as selection criteria.

Material and Method

The seed of 18 accessions of *Panicum miliaceum* was obtained from the National Agriculture Research Centre, Islamabad.

Name and origin of 18 accessions of *Panicum maliaceum* L.

Accession no.	Origin
008208	Droshp, Chitral
008210	Parwak, Chitral
008211	Sloaspur, Chitral
008213	Pengal, Gilgit
008214	Damalgand, Gilgit
008215	Drach, Gilgit
008216	Yangal, Gilgit
008217	Sakandar Abad, Gilgit
008218	Murtazabad, Gilgit
008220	Rando Bagicha, Skardu
008221	Karis, Skardu
008222	Goon, Skardu
008223	Kunis, Skardu
008225	Barah, Skardu
008226	Nasir Abad, Gilgit
008230	Quetta, Quetta
008236	Keris, Skardu
008242	Pishin, Pinhin

This experiment was conducted in a wire house in the Botanic Garden of the Department of Botany, University of Agriculture, Faisalabad. The experiment was arranged in a completely randomized (CRD) factorial design, with two NaCl treatments (0 and 120 m*M*) and 18 accessions of proso millet in four replicates. The seeds were surface sterilized for 10 min, with sodium hypochlorite before sowing.

The seeds of proso millet (*Panicum miliaceum*) were directly sown into plastic pots of 28 cm diameter containing 12.50 kg of well washed sand. All pots were irrigated with full strength Hoagland's nutrient solution for 14 days after which time two NaCl treatments (0 or 120 m*M*) in full strength Hoagland's nutrient solution were started. Salt solution was applied in aliquots of 40 m*M* every day. Two liters of the treatment solution were applied to each pot after every week. Plants were harvested after four weeks of the initiation of treatment and data for different parameters were recorded during the course of the experiment.

For the determination of K^+ and Na^+, the dried ground leaf or root material (0.1 g) was digested with sulphuric acid and hydrogen peroxide (Merck) and Na^+, and K^+ in the digests were determined with a flame photometer (Jenway, PFP-7).

Statistical analysis of data: Analysis of variance of all parameters was computed using the COSTAT computer package (CoHort software, Berkeley, USA). Mean values of each attribute were compared using least significance difference test (LSD) at 5% probability following Snedecor & Cochran (1980).

Results and Discussion

Data for shoot and root fresh and dry weights of 18 accessions of *Panicum miliaceum* (Table 1) showed that NaCl treatment had a significant inhibitory effect on the growth attributes of all accessions of *P. miliaceum*. However, different accessions differed in their response to the NaCl treatment. Some showed greater reduction in their biomass production, while the other showed little reduction.

The existence of the genetic variation for salt tolerance is a pre-requisite for the development of salt tolerant cultivars through selection and breeding. Inter and intra-specific variation for salt tolerance provides scope for selecting for its improvement. If sufficient amount of genetic variation for salt tolerance exists in a species, then considerable improvement in tolerance could be expected from selection (Ashraf, 1994). It is argued that selection for salinity tolerance at the seedling stage may not produce tolerant adult plants (Kingsbury & Epstein, 1984; Shannon, 1997). To explore the variation for salt tolerance in proso millet, 18 accessions were screened at the early growth stage i.e., seedling stage, as the salt tolerance at this stage is crucial for yielding vigorous plants for tolerating salt stress at later stages of growth. It is clear from the growth data that there is a decrease in the biomass production under saline conditions but the rate of decrease in biomass under saline conditions was different in different accessions of *P. miliaceum,* so the accessions were classified as tolerant, moderately tolerant and sensitive to salt stress on the basis of their biomass production. Those accessions which produced high biomass were salt tolerant, while those produced less were salt sensitive.

Classes	Range of wt. (g/plant)	Accessions no.
Tolerant	>20	008211, 008214, 008220, 008226
Moderately tolerant	20-15	008210, 008213, 008215, 008216, 008217, 008218, 008222, 008221, 008223, 008225, 008230
Sensitive	<15	008208, 008236, 008242

Table 1. Fresh and dry weights of shoot and root and shoot/root ratio of 18 accessions of proso millet, when grown under non-saline (0 m*M* NaCl) or saline (120 m*M* NaCl) conditions for 28 days.

Accession No.	Shoot fresh wt.		Root fresh wt.		Shoot dry wt.		Root dry wt.		Shoot root ratio	
	0 m*M*	120 m*M*	0 m*M*	120 m*M*	0 m*M*	120 m*M*	0 m*M*	120 m*M*	0 m*M*	120 m*M*
008208	29.99	5.72	2.59	0.79	3.25	1.31	0.46	0.18	11.55	7.22
008210	27.26	17.79	1.61	1.12	4.14	2.48	0.32	0.28	16.82	15.86
008211	21.26	21.14	1.48	1.28	2.73	2.92	0.32	0.36	14.28	16.45
008213	33.89	19.17	1.72	1.20	4.91	2.73	0.43	0.26	19.61	15.86
008214	25.59	24.41	1.64	1.34	4.07	3.68	0.51	0.35	15.52	18.20
008215	22.32	14.15	1.51	1.09	3.27	2.5	0.35	0.3	14.76	12.91
008216	20.56	19.75	1.92	1.72	2.83	2.79	0.27	0.32	10.70	11.42
008217	28.99	19.37	2.64	2.05	3.07	2.59	0.47	0.40	10.97	9.41
008218	33.77	17.06	3.07	1.96	4.32	1.95	0.53	0.38	10.99	8.67
008220	31.70	20.04	2.91	2.23	4.72	2.84	0.56	0.34	10.88	8.96
008221	26.80	18.02	2.63	1.25	3.16	2.08	0.44	0.22	10.18	14.36
008222	28.59	23.73	3.10	3.05	3.38	2.74	0.46	0.64	9.22	7.76
008223	21.68	20.63	1.93	1.88	2.51	2.46	0.52	0.34	11.20	10.93
008225	23.53	16.85	2.69	2.61	2.83	2.03	0.42	0.49	8.74	6.43
008226	18.35	20.49	1.59	2.7	2.77	2.3	0.26	0.52	11.49	6.45
008230	28.45	15.26	1.38	1.13	2.96	1.85	0.36	0.22	20.58	13.49
008236	28.99	12.63	2.03	1.05	3.01	1.32	0.36	0.17	14.23	12.00
008242	28.30	15.41	1.79	1.63	2.65	1.58	0.35	0.27	15.73	9.44
LSD 5% (Salt stress x Accessions)	**8.84**		**0.886**		**1.622**		**0.214**		**ns**	

Table 2. Shoot and root Na^+ and K^+ and shoot K^+/Na^+ ratio of 18 accessions of proso millet, when grown under non-saline (0 m*M* NaCl) or saline (120 m*M* NaCl) conditions for 28 days.

Accession No.	Shoot Na^+		Root Na^+		Shoot K^+		Root K^+		Shoot K^+/Na^+ ratio	
	0 m*M*	120 m*M*	0 m*M*	120 m*M*	0 m*M*	120 m*M*	0 m*M*	120 m*M*	0 m*M*	120 m*M*
008208	7.875	11.87	7.87	11.87	8.75	7.37	35.62	11.5	6.19	3.52
008210	6.87	8.37	6.87	8.375	7.62	7.37	23.75	11.12	6.27	5.00
008211	6.125	9.25	6.12	9.25	9.15	9.00	29.17	9.00	7.24	6.01
008213	7.00	11.13	7.00	11.125	8.75	6.12	26.87	7.12	6.96	3.2
008214	6.625	8.25	6.62	8.25	8.75	8.12	30	10.37	7.35	5.53
008215	7.00	8.25	7.00	8.25	8.65	8.12	28.75	7.75	6.87	5.83
008216	7.00	8.00	7.00	8.00	8.25	8.00	25.00	12.00	6.61	5.62
008217	6.00	12.12	6.00	12.125	8.65	8.25	23.75	10.8	7.68	3.73
008218	6.5	8.33	8.33	8.33	9.15	8.5	23.3	9.17	5.77	6.5
008220	6.875	9.75	6.87	9.75	7.75	5.75	27.5	9.00	6.36	3.46
008221	7.5	12.33	7.5	12.33	9.00	8.83	31.5	8.67	6.67	4.32
008222	8.00	9.5	8.00	9.50	9.65	7.87	33.75	12.5	7.19	4.67
008223	6.83	11.33	6.83	11.33	10.25	9.67	24.37	10.00	6.71	5.46
008225	6.67	8.125	6.67	8.12	9.75	8.87	28.33	10.00	8.06	6.08
008226	5.17	6.00	5.17	6.00	9.5	8.75	37.5	9.67	9.99	8.12
008230	7.125	7.75	7.12	7.00	9.33	7.50	19.7	8.50	8.66	5.71
008236	6.125	19.17	6.12	19.17	10.25	9.83	26.25	7.87	8.67	3.69
008242	6.125	8.67	6.12	8.67	11.17	9.50	36.25	8.67	9.99	7.79
LSD 5% (Salt stress x Accessions)	**3.92**		**ns**		**ns**		**7.06**		**ns**	

Data for shoot and root Na^+ showed that the pattern of Na^+ accumulation was different in different accessions of *P. miliaceum*. It was observed that most of the salt tolerant accessions (high biomass producing) accumulated low amount of Na^+ in their shoots and roots under saline conditions but the salt sensitive accessions (low biomass producing) accumulated high amount of Na^+ under saline conditions. The salt tolerant accessions also maintained higher shoot K^+/Na^+ ratio as compared to the salt sensitive accessions (Table 2). The results for Na^+ accumulation and maintenance of K^+/Na^+ ratio are well known phenomena of salt tolerance in a number of mesophytes (Ashraf, 2004; Ahmad & Jabeen, 2005)

Although a considerable magnitude of variation for salt tolerance was observed in a set of 18 accessions while screening them at the seedling stage, it is not sure whether the tolerant accessions found at the seedling stage would maintain their degree of salt tolerance when tested as adult. This needs to be elucidated in a further study.

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