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In-vitro Screening of Finger Millet (*Eleusine Coracana* (L.) *Gaertn*.) Genotypes for Salinity Tolerance

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Finger millet is commonly called "nutritious millet" as the grains are nutritiously superior to many cereals providing fair amount of protein, minerals, calcium and vitamins. The protein of finger millet is considered to be "biologically complete" as in the case of milk. For the utilization of saline lands, it is essential to manage the salinity or to grow crops and their varieties resistant to salinity. Reclamation of saline soils is a time taking and costly affair. Hence, it is urgently needed to identify crops and their genotypes which can resist salinity. Considering the importance of finger millet as food and its general capacity to withstand salinity, forty genotypes of finger millet were screened for salinity tolerance at seedling stage with ½ MS basal medium having four levels of salinity *viz.,* 3000, 6000, 9000 and 12000 ppm along with control. Germination percentage and seedling parameters *viz.,* fresh weight, shoot length, root length, seedling length and dry weight were recorded on 20^{h}and 30 thday after inoculation. Considering all the seedling characters together, the genotype AF 459 showed superiority for all the five characters studied. Germination of this genotype was also found to be least affected at 3000 and 6000 ppm salt concentration. Besides at 9000 ppm, it showed less than 15% reduction in germination compared to control. The seedlings of genotype AF 269 showed higher fresh and dry weight at 3000 ppm salt concentration compared to control, while the genotypes TNAU 1008 and GS 159 had a higher root length both at 3000 and 6000ppm salt concentrations. Besides the genotypes AF 269, TNAU 1008 and GS 159 also showed lesser population reduction at 3000 and 6000 ppm salt concentration.

Keywords: Finger millet, genotypes, *in-vitro* screening, seedling parameters, salinity tolerance.

Finger millet or Ragi (*Eleusine coracana* (L.) Gaertn.) 2n = 4x = 36, is a poor man's crop, originated in Ethiopia (Vavilov, 1951). Finger millet is commonly called "nutritious millet" as the grains are nutritiously superior to many cereals providing fair amount of protein, minerals, calcium and vitamins. The Consultative Group on International Agricultural Research (CGIAR) has estimated that 10 per cent of the area under millets is with finger millet. Globally it is produced in 3.38 million hectares with the production of 3.76 million tonnes (FAO, 2007). In the widest context, the stress environment is one of the major factors responsible for the current global environmental problems. In India, the problem soils occupy 10 million hectares, which include saline soils of 7.2 million hectares and alkali soils of 2.8 million hectares (Biswas and Mukherjee, 1997). The problem soils are the highest in Uttar Pradesh, Bihar, Rajasthan and Madhya Pradesh; followed by Orissa, Andhra Pradesh, Tamil Nadu and Karnataka. In Tamil Nadu, salt affected soils are wide spread in almost all the districts and nearly 3 lakh hectares of land are affected by the soil salinization and sodicity (Vadivel *et al.,* 2001). Reclamation of saline soils is a time taking and costly. Salinity is one of the most widespread stress hazards and occurs mostly in arid and semi-arid region (Abrol, 1986), though it also exists in some sub-humid areas and coastal lands. Hence, it is urgently needed to identify crops and varieties process, which can resist salinity. Finger millet, with many cultivars grown in semi arid areas has attracted research interest to test its level of resistance to salt stress. Thus, some Indian cultivars of finger millet have been a subject of salt stress studies on germination, growth and grain yield (Onkware, 1993). Considering the importance of finger millet as food and its capacity to withstand salinity, the present investigation was carried out with forty genotypes to evaluate their tolerance to salinity under *in-vitro* condition.

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Materials and Methods

Forty genotypes of finger millet (*Eleusine coracana* (L.) Gaertn.) obtained from the germplasm collection of the Department of Millets, Centre for Plant Breeding and Genetics, Tamil Nadu Agricultural University, Coimbatore were chosen for the study. A slightly modified Murashige and Skoog medium with vitamins of B_5medium was used throughout the experiment. The composition of MS basal medium used in this study was similar to hormone free MS medium used by Murashige and Skoog (1962) with B_5vitamins. Four levels of salinity stress *viz.,* 3000, 6000, 9000 and 12000 ppm were imposed by adding sodium chloride (NaCl) to ½ MS medium separately

for screening salinity tolerance along with a control without NaCl. The pH was adjusted to 5.8 using 0.1 N NaOH or 0.1 N HCl. Characters like fresh weight, seedling of shoot length, root length, mean seedling length and dry weight were studied. The observations were recorded individually at 20 and 30 days of seedling growth at each salinity level maintained in three replication for each genotype. Germination was recorded on 15 days. The experimental design was FRBD and the data collected was converted to angular transformation before statistical analysis. (Panse and Sukhatme, 1964).

Results and Discussion

Significant differences among the genotypes with respect to germination at different salinity levels were observed. The interaction of genotypes with different salinity levels was also found to be significant (Table 1). The germination percentage was found to reduce with the increase in salinity level in all the genotypes studied (Table 2). Two genotypes MS 8070 and GS 159 showed same level of germination as that of control at two levels of salt treatment *viz.,* 3000 and 6000 ppm. The genotype TNAU 1008 showed slight increase in germination percentage over (2%) control at 3000 and 6000 ppm. Germination of the genotypes MS 8070, AF 329, GE 2510, MS 9272, TNAU 5, AF 459, GS 69, GS 112, MS 8100, TNAU 21, TNAU 193, TNAU 1008, AF 269, GE 333, GEC 520, GS 159, Malavi 1876, MS 2927, MS 8068 and TNAU 44/2 was least affected at 3000 and 6000 ppm. Germination of AF 329 was not severely affected even at 9000 ppm less than 15% reduction over control. Germination of all the genotypes was severely affected at 12000

Table 1. Analysis of variance for seedling parameters

Source	df	Fresh weight		Shoot length		Root length		Seedling length		Dry weight	
		20 days	30 days	20 days	30 days	20 days	30 days	20 days	30 days	20 days	30 days
Genotypes	39	0.00024**	0.00112**	6.9618**	8.38430**	12.2524**	13.0319**	35.7790**	37.2199**	0.0004**	0.00021**
Treatment	4	0.01271**	0.01266**	238.6546**	306.3044**	365.2662**	609.2545**	1188.4432**	1766.0372**	0.0001**	0.00020**
G X E	156	0.00019**	0.00047**	1.7351**	2.5285**	3.0453**	6.5639**	8.0793**	12.9055**	0.0003**	0.00017**
Error	400	0.00012	0.00026	1.2224	1.4425	1.9143	3.9153	5.4250	8.8725	0.0001	0.00021

** Significant at 1 % level

Table 2. Germination (%) of 40 genotypes at different levels of salt stress on 15th day of culture

Genotypes	Control	Salt concentration				Mean**
		3000 ppm	6000 ppm	9000 ppm	12000 ppm	
Co 9	77.48	77.79	75.56	39.63	28.89	59.76
GE 527	82.79	60.79	68.86	51.89	17.89	58.89
GEC 534	82.79	71.25	60.32	57.12	53.17	59.77
GS 133	82.79	89.89	61.45	28.65	39.75	58.04
GS 261	89.89	89.89	69.32	48.76	15.32	58.53
Malavi 1446	89.89	89.89	51.67	59.17	38.66	61.64
MS 2944	89.89	73.50	75.55	60.52	53.32	65.89
MS 8070	89.89	89.89	89.89	57.12	29.35	60.89
TNAU 30	82.79	75.55	60.79	56.73	29.01	60.76
TNAU 72	89.89	78.68	73.50	57.76	29.12	63.43
AF 329	82.79	75.55	89.89	72.01	39.76	67.02
Co 14	82.79	78.68	75.55	57.76	19.45	60.58
GE 346	82.79	82.79	74.89	39.76	29.45	60.69
GE 2510	81.84	78.68	89.89	39.76	29.12	61.79
GS 431	81.84	74.45	58.89	57.12	19.45	59.69
IE 3297/1	79.32	69.79	58.89	37.89	18.39	58.76
MS 9272	89.89	89.89	78.17	68.00	37.45	67.14
TNAU 5	89.89	78.68	74.89	39.76	37.99	60.00
TNAU 487	79.32	75.55	68.86	37.89	19.45	58.67
TNAU 4833	89.89	73.50	61.45	39.45	51.97	58.75
AF 459	78.17	89.89	82.79	71.17	50.76	72.63
GE 522	77.71	75.55	60.65	39.45	45.64	58.76
GEC 539	82.79	74.68	73.89	61.89	42.54	63.76
GS 69	89.89	89.89	82.84	63.89	49.54	73.77
GS 112	82.89	89.89	82.79	51.50	29.00	63.13
Malavi 2028	79.32	71.25	63.91	55.67	47.54	60.89
MS 8100	81.84	82.79	52.86	61.84	52.67	68.67
TNAU 21	89.89	89.89	82.79	51.50	16.15	62.42
TNAU 193	71.42	89.89	79.32	51.35	16.17	60.76
TNAU 1008	81.84	89.89	89.89	72.02	17.89	65.74
AF 269	89.89	82.79	74.32	65.55	56.89	67.37
AF 3055	89.89	82.79	75.55	65.89	39.78	66..07
GE 333	89.89	82.79	89.89	67.25	50.76	68.73
GEC 417	82.79	51.67	60.79	58.89	52.44	59.07
GEC 520	89.89	89.89	82.79	68.45	52.87	72.79
GS 159	89.89	89.89	89.89	58.89	25.76	63.65
Malavi 1876	89.89	82.79	75.55	45.55	39.76	60.31
MS 2927	89.89	79.32	89.89	29.89	15.63	59.53
MS 8068	82.79	78.68	74.89	19.32	49.54	60.36
TNAU 44/2	82.79	89.89	82.79	55.18	17.89	61.89
Mean**	82.57	77.79	73.42	55.89	29.76	63.32

* Transformed values are given in parenthesis

		SED	CD (0.05)	CD (0.01)	* Significant at 5%
A	Genotypes	11.56	22.72	29.91	** Significant at 1%
B	Salt concentration	4.09	8.03	10.58	
A x B	Genotypes x Salt concentration	12.84	25.80	33.89	

Plate 1. Variation in seedling characters of AF 269 and TNAU 1008 under different salt stress
A – control B – 3000 ppm C – 6000 ppm
D – 9000 ppm E – 12000 ppm

ppm, and among these, AF 459 recorded the least reduction for germination (47.22%). Responses of genotypes to different salt concentration are given in Table 3. The differential response of finger millet genotypes for germination at different salinity levels had been earlier reported by Panigarh *et al.* (1978)

Table 3. Response of genotypes to different salt concentration

Character	Range of values					
	3000 ppm		6000 ppm		9000 ppm	
	20 days	30 days	20 days	30 days	20 days	30 days
Fresh weight (g)	0.0030-0.0403	0.0053-0.0517	0.0030-0.0410	0.0030-0.0520	0.0030-0.0153	0.0030-0.041
Shoot length (cm)	0.060-4.500	0.830-5.760	0.00-5.300	0.00-5.660	0.00-2.930	0.00-3.2600
Root length (cm)	0.030-7.030	0.200-7.830	0.000-6.430	0.0-7.030	0.00-3.830	0.00-3.8300
Seedling length (cm)	0.100-11.100	1.100-12.400	0.00-11.73	1.700-10.660	0.00-6.360	0.00-7.1000
Dry weight (g)	0.0016-0.0047	0.0015-0.0075	0.0016-0.0070	0.0017-0.0060	0.0016-0.0055	0.0011-0.004

and Onkware (1993). The drastic effect of higher salinity levels on the germination of finger millet had also been reported by Sarma *et al.* (1983) and Uma *et al.* (1995).

Genotypes superior for salt tolerance at 3000 and 6000 ppm were observed for different seedling characters (Table 4). The top ranking genotypes for increased/lesser reduction in fresh weight of seedling over control at 3000 ppm on 20 thand 30 thday of culture were MS 8100, AF 269, AF 459, MS 9272 and Co 14. At 6000 ppm of salt concentration, the genotypes AF 269, AF 329 and AF 459 were found to be superior both on 20 thand 30 thday of culture. Two genotypes *viz.,* AF 269 and AF 459 performed well both at 3000 ppm and 6000 ppm with respect to fresh weight of seedling.

Table 4. Superior genotypes with salt tolerance at 3000 ppm and 6000 ppm for different seedling characters

Character	3000 ppm			6000 ppm			Best performing genotypes
	Genotypes	20thday	30thday	Genotypes	20thday	30thday	
Fresh weight	MS 8100	+83	+21	AF 269	+33	+46	AF 269 AF 459
	AF 269	+48	+68	AF 329	-2	+41	
	AF 459	+29	+13	AF 459	-4	+65	
	MS 9272	-5	+26				
	Co 14	-10	+90				
Shoot length	MS 8100	+70	+7	AF 459	+14	+15	AF 459
	GS 69	+2	+11				
	AF 459	-3	+17				
	MS 9272	-8	0				
Root length	GS 69	+46	+18	AF 459 TNAU 487 TNAU 1008 GS 159	+25 +17 +4 -2	+23 +98 -4 -3	AF 459 TNAU 1008 GS 159
	TNAU 1008	+45	+27				
	GS 133	+35	+4				
	AF 3055	+32	+1				
	AF 459	+14	+5				
	GS 159	+11	+7				
	GEC 520	-5	+16				
	AF 329	-10	+62				
	TNAU 4833	-10	-2				
	TNAU 193	-11	+38				
Seedling length	GS 133	+22	-10	AF 459 TNAU 487	+20 -4	+19 +45	AF 459
	TNAU 1008	+19	-1				
	GE 346	+14	+20				
	AF 459	+6	+11				
Dry weight	GE 522	+79	-6	MS 8068 AF 269 MS 2927 TNAU 44/2 GS 112 AF 459	+28 +16 0 0 -6 -8	+91 +9 +15 +8 0 +21	AF 459 AF 269
	MS 8100	+33	+13				
	AF 459	+18	+52				
	AF 269	+16	-3				
	TNAU 193	-7	-6				

+ values indicate per cent increase in the character over control and- values indicate per cent reduction in the character over control

The genotypes MS 8100, GS 69, AF 459 and MS 9272 had increased/lesser reduction in shoot length of seedling over control at 3000 ppm both on 20 days and 30 days of culture. At 6000 ppm, AF 459 was found to be the best performing genotype. Considering the two salt concentrations, shoot length of seedling was least affected at 3000 ppm and 6000 ppm in the genotype AF 459. Shoot length of seedling significantly decreased with the increase in salinity stress in finger millet. The reduction of shoot length was found to accompany with increased salt stress in several crops (Torres and Echevarria, 1994; Begam *et al*., 1996; Singh *et al*., 1998 and Singh and Singh, 1999).

Plate 2. Variation in seedling characters of TNAU 72 and GEC 520 under different salt stress
A – control B – 3000 ppm C – 6000 ppm D – 9000 ppm E – 12000 ppm

The genotypes that recorded increased/lesser reduction in root length of seedling over control at 3000 ppm both after 20 days and 30 days of culture were GS 69, TNAU 1008, GS 133, AF 3055, AF 459, GS 159, GEC 520, AF 329, TNAU 4833 and TNAU 193. At 6000 ppm salt concentration, the genotypes AF

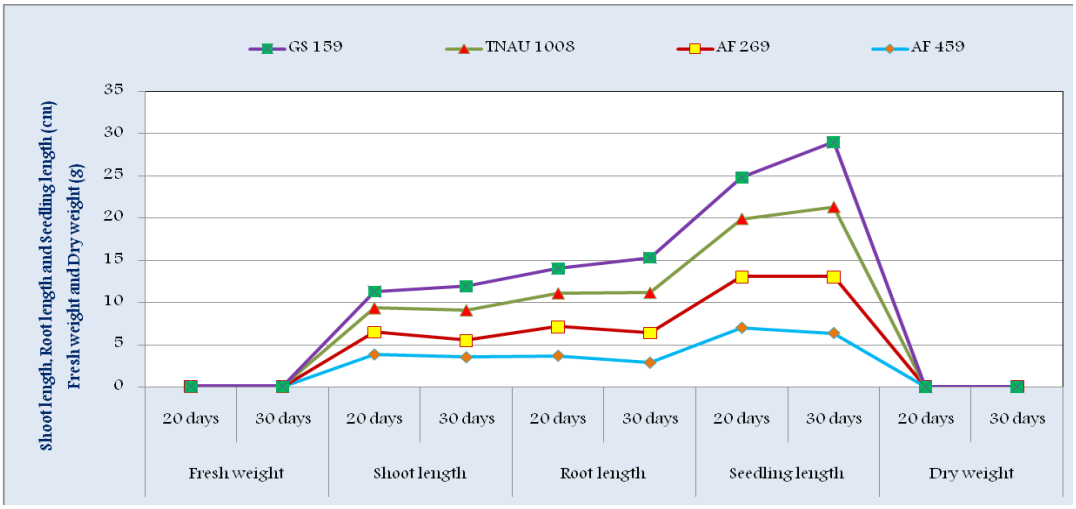

Fig. 1. Overall mean performance of four best performing genotypes for various seedling characters at different levels of salt stress on 20 thand 30 thday of culture

459, TNAU 487, TNAU 1008 and GS 159 had given an increased/lesser reduction in fresh weight of seedling over control. Three genotypes *viz.,* AF 459, TNAU 1008 and GS 159 performed well both at 3000 ppm and 6000 ppm. The reduction in root length of seedling due to increased salinity had been reported by Panigarh *et al.* (1978) in finger millet.

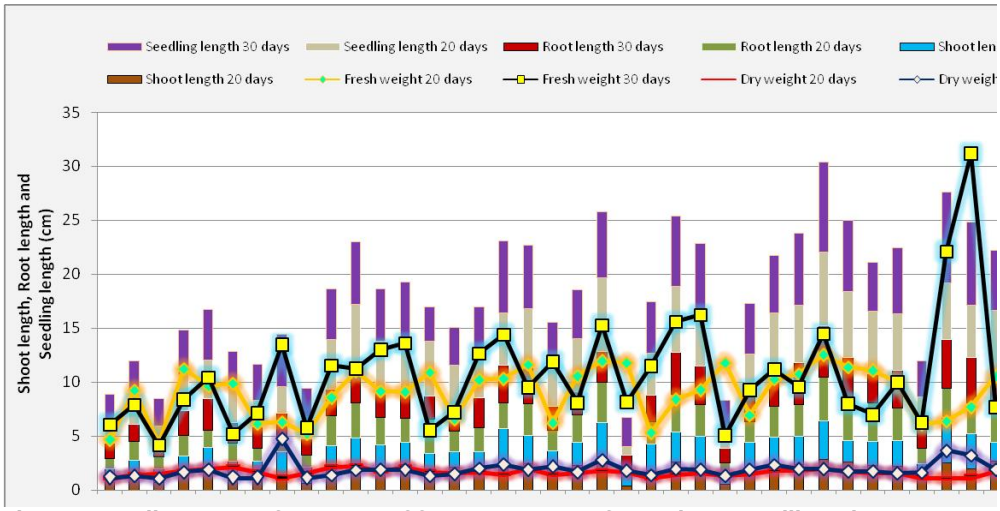

Fig. 2. Overall mean performance of forty genotypes for various seedling characters at different levels of salt stress on 20thand 30 thday of culture

The genotypes GS 133, TNAU 1008, GE 346 and AF 459 showed an increased/lesser reduction at 3000 ppm both on 20 thand 30 thday of culture with respect to seedling length. The genotypes AF 459 and TNAU 487 performed well at 6000 ppm at 20 and 30 days of culture. Considering the two salt concentrations 3000 ppm and 6000 ppm together, seedling length was least affected in the genotype AF 459. The reduction of seedling length due to increased salinity had been reported in several crops by Abul-Baki and Anderson (1973), Torres and Echevarria (1994), Singh *et al.* (1998) and Singh and Singh (1999).

Seedling dry weight in the genotypes GE 522, MS 8100, AF 459, AF 269 and TNAU 193 were recorded with increased/lesser reduction over control at 3000 ppm after 20 and 30 days of culture. At 20 and 30 days of culture, the genotypes MS 8068, AF 269, MS 2927, TNAU 44/2, GS 112 and AF 459 were found to show better performance compared to other genotypes at 6000 ppm. Considering the two salt concentrations (3000 ppm and 6000 ppm) together, the genotypes AF 459 and AF 269 showed a higher level of tolerance with increased seedling dry weight. The variation in seedling characters of four genotypes (AF 269 and TNAU 1008, TNAU 72 and GEC 520) under different levels of salt stress was documented (Plate 1 and Plate 2).

Conclusion

Considering all the seedling characters, the genotype AF 459 showed superiority for all the five seedling characters studied. Germination of this genotype was also least affected at 3000 and 6000 ppm salt concentration. The genotype AF 269 had higher fresh weight and dry weight seedling of at the lowest level of salt concentration used, while the genotypes TNAU 1008 and GS 159 had a higher root length. The genotypes AF 269, TNAU 1008 and GS 159 also showed the least reduction in germination at 3000 and 6000 ppm salt concentration. The overall mean performances of the best performing genotypes for various seedling characters at different levels of salt stress on 20 thand 30 thday of culture are presented in Figure 1 and Figure 2, respectively. Hence, these genotypes can be used as parent in hybridization programme for the improvement of finger millet in salt affected areas.

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