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Morphological and physiological responses of Tanzania rice genotypes under saline condition and evaluation of traits associated with stress tolerance

S. Kashenge-Killenga [1*], P. Tongoona [2], J. Derera [2]

[1] *Agriculture Research Institute (ARI) KATRIN, Private bag Ifakara, Morogoro –Tanzania*
[2] *African Centre for Crop Improvement (ACCI), School of Agricultural Science and Agribusiness University of Kwazulu Natal (UKZN), Box X01, Scottsville 3209, PMB, South Africa*

Abstract

Rice genotypes including local farmer preferred cultivars were tested against saline stress to evaluate their performance and develop selection criteria for salt tolerance. A hydroponics mass screening technique was used to test the 10 genotypes in NaCl- saline treated and non-treated solutions. Significant variation between genotypes and significant interactions between genotype and salt treatment ($P<0.001$) was observed for all characters. Saline stress reduced plant growth of all tested genotypes. Plant vigour and shoot K^+ concentration were relatively more affected than other traits. On the basis of less than 50% reduction in different growth variables, Pokkali displayed significantly superior performance followed by IR67076-2B-21-2 and IR 56 whereas Nerica1 showed the poorest performance. Tolerant rice genotypes were successful in maintaining low Na^+ and high K^+ uptake and low Na^+/K^+ ratio. The study therefore established that, all the local farmer preferred cultivars except IR 56 performed poorly under saline stress environments. High seedling vigour, less leaf injury, less Na^+, high K^+ accumulation in leaves and low Na^+/K^+ ratio of ion uptake should be considered as desired characteristics while screening the lines for salt tolerance under saline environment.

Keywords: Rice; Oryza sativa; Genotypes screening; Saline; Stress tolerance; Morphological Traits.

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[*] Corresponding author. *E-mail address:* sophykashenge@yahoo.com

1. Introduction

Rapidly increasing soil salinity has multifarious effects on plant growth and productivity; and an increase in this menace is posing a serious threat particularly to irrigated agriculture in arid and semiarid areas. According to Cassman (2004), ten million ha of irrigated land are abandoned every year because of increasing soil salinity and mismanagement of irrigation principles and farming practices have made a huge contribution to the problem.

Generally, rice is grown in almost all regions of Tanzania but at various levels of importance. However, yields are low and still dwindling compared to the increasing consumption (Kafiriti, 2004). Due to increasing demand for rice consumption, about 10 to 25 % of the total consumption is imported annually to augment the shortfall (Kafiriti, 2004; FAO, 2005). In addition the cultivable land is declining due to increasing drought and salinity among other causes of soil degradation. There are less new lands available especially for irrigated cultivation, the available land which can be used as arable irrigated land need high initial investments cost to develop irrigation infrastructures therefore unaffordable to poor resource farmers. An important aspect of this is gainful utilization of these salt affected areas (Araki et al., 2001). To achieve optimal food production in these areas, the most appropriate and logical choice is to grow salt tolerant crops/cultivars (Khan and Abdullah, 2003).

Studies have been done in support of the predominance of saline stress tolerance in explaining yield improvement in rice (Singh, 2001; Moradi et al., 2003). It has been reported further that salt tolerance varies considerably with the developmental stages in a number of species including rice (Flowers, 2004). Rice is comparatively tolerant of saline stress during germination, active tillering, and towards maturity and is sensitive during early seedling and reproductive stages, however genetic variation exists between genotypes (Mahmood et al., 2004). Bayuelo-Jimenez et al. (2002) found that tolerance at emergence followed by seedling survival and establishment are important in the maintenance of optimal crop stand in the field, and ultimately the economic yield.

Improving a given crop or cultivated species for salt tolerance involve reliable screening procedures to search amongst natural diversity within the species, or closely related and inter - fertile species. However, the quantification of salt tolerance poses difficulties, for example, in the field, first, the salt stress may be experimentally uncontrollable due to climatic effects upon rainfall, temperature and water tables; secondly, field heterogeneity for salt is high (Munns et al., 2002) and thirdly, salt uptake and so sensitivity, is modulated by environmental conditions and may affect each variety differently: any parameter which affects the transpiration rate (such as light intensity, temperature and humidity) can change a plant's susceptibility to salinity therefore genotype x environment interactions are highly expected (Yeo et al., 1990; Yeo and Flowers 1989). For that reason, genotype assessments under controlled conditions are highly advocated. In addition, screening for a trait associated with a specific mechanism is preferable to screening for salt tolerance itself (Chaubey and Senadhira, 1994). A range of screening methods have been reported, including field evaluation method, whereby a barren salt affected land was used (Mishra et al., 2006), laboratory techniques as well as pot and solution culture methods. Laboratory techniques and solution culture methods are common greenhouse techniques for screening genotypes under controlled conditions (Singh and Chatrath, (2001) .

Studies on the effect of salt tolerance in plants have indicated that salt damage and consequently adaptation to salt is complex, and no single process can account for the variation in the plant's response to salt stress (Yeo et al., 1990). Varietal variation has been observed within small (ten or fewer) samples of genotypes in characteristics that mitigate the consequences of salt uptake (Krishnamuthy et al., 2007). Based on several studies in salinity stress over the years, such studies have never been done to evaluate rice materials grown under Sub-Saharan Africa environments including Tanzania. It has been reported that rice varieties vary in their tolerance to type and salt levels. Significant variations in phenology, morphology and physiology across environments have also been observed among genotypes (Munns et al., 2006). Therefore, a study was established using a hydroponic screening method under controlled condition to assess the salinity tolerance of some Tanzania farmers' preferred rice cultivars and evaluate the putative traits in the rice materials that contribute to the performance of a genotype under saline condition. Specifically the objectives were to i) determine the performance of rice genotypes commonly gown in irrigation schemes of North-eastern Tanzania under saline conditions, and ii) determine morphological and physiological traits associated with tolerance to saline conditions.

2. Material and methods

2.1. Germplasm

The study used 10 rice genotypes comprising Pokalli, IR67076-2B-21-2 (abbreviated as IR67076), CSR 27 (Tolerant checks), IR 29 (a susceptible check), TXD 306, IR 64, IR 56, Thailand, (Farmer's preferred cultivated varieties), Gigante and Nerica 1. These cultivars had not been previously evaluated for tolerance to saline stress under local environments. Table 1 shows characteristics of all collections involved in the study.

2.2. Screening of rice genotypes for salinity tolerance (hydroponics based method)

The salt tolerance of rice genotypes were studied in a green house without much environmental control. The screenhouse had well covered roof, but sides were open having only wire gauze and insect proof net. There was no problem of sunlight. The weather data were monitored at Mlingano weather station located 30m from the greenhouse. Seeds of 10 selected genotypes (Table 1) were germinated in sterile mixture of soil, sand and manure at 1:1:1 ratio and seedlings were grown for 14 days. Plastic containers of 40 × 25 × 20 cm were prepared for the screening purpose. A styrofoam sheet was cut to fit the top of each container. Five rows with five holes each were made on each styrofoam sheet and a nylon net was placed at the bottom of each styrofoam sheet to prevent the seedling from falling into the solution as described by Gregorio *et al.* (1997). Each styrofoam sheet was floated in a container filled with 4L of distilled water as indicated in Figure 1.

Table 1. Characteristics of thirteen genotypes used in the study

Genotype	Source[1]	Characteristics
Pokkali (IR – 4595-4-1-1-3)	IRRI	- A traditional donor for saline tolerance – developed by pedigree breeding method. Saline tolerant, local pureline, early maturing (Chaubey and Senadhira, 1994).
IR67076-2B-21-2	IRRI	Moderately salt tolerant & aromatic pureline
CSR 27 (CSR 88IR-7)	IRRI	A fine grained high yielding variety developed from Nona Bokra/IR 5657-33-2, tolerant to saline and sodic soil environment, and known to tolerate pH 9.6 – 9.9 and adaptation of ECe(dSm^{-1}) < 10. Released variety (Chaubey and Senadhira, 1994).
TXD 306 (SARO 5)	NRI	Semi-aromatic cultivated variety, resistant to lodging, medium maturing, high growth vigour, good milling and cooking qualities, released in Tanzania,
IR 64	NRI	Preferred cultivar (highly grown in Mombo irrigation schemes), tolerant to P and Zn deficiency, iron and boron toxicity, Early maturing, high yielding. Originate from IRRI
Nerica 1	NRI	- Upland aromatic, early maturing variety, high growth vigour, strong calms tolerant to lodge, blast and insects
Thailand	NRI	- Local preferred cultivar (highly grown in Mkomazi and Mombo) irrigation schemes, high yielding, heavy grain weight, medium maturing.
IR 56	NRI	- Preferred cultivar (highly grown in Mkomazi and Moshi) irrigation schemes. High yielding, medium maturing variety. Originate from IRRI
Gigante	NRI	- Landrace known to have resistance to rice yellow mottle virus
IR 29	IRRI	Salt sensitive cultivar used as a susceptible check

[1]*(IRRI-International Rice Research Institute, NRI- National Rice Research Institute)*

(a) Plant growth in nutrient solution

After two weeks, the seedlings (at two to three leaf stages) were uprooted, rinsed with sterilized deionised water to remove the soil and were transferred to the prepared containers. Each container had five rows consisting of five genotypes (one genotype per row), and each hole had three seedlings (Figure 1). Control genotypes were grown after every one container for comparison. The seedlings were grown in distilled water for three consecutive days and then distilled water was replaced by a nutrient solution prepared using 1ml/L of working solution (Gregorio et al., 1997).

The working solution was prepared using the following stocks: NH_4NO_3 (91.4 g/L), Na_2HPO_4 (35.6 g/L), $CaCl_2$ (117.4 g/L), $MgSO_4$ (324 g/L) and KSO_4 (70.65mg/L) for macronutrient stocks and a combination of $MnCl_2$ (1.5 g/L), H_3BO_3 (0.934g/L) $ZnSO_4$ (0.035g/L), $FeSO_4$ (7.7g/L), CuSO4 (0.031g/L) $[(NH_4)6Mo_7O_{24}]$ (0.13g/L) and $H_3C_6H_8O_7$ (11.9 g/L) was used to make stock solution for required micronutrients (Gregorio et al., 1997; Yoshida et al., 1976). Seedlings were cultured in the nutrient solution for 10 days prior to salinization to allow proper establishment. The nutrient solution was renewed every 8 days and the pH of 5.0 was maintained daily by adding either NaOH or HCl.

Figure 1. Seedlings growth in a styrofoam sheets floated on modified Yoshinda solution (Yoshida et al., 1976)

(b) Salinisation test

The first experiment was conducted at EC = 60m*M* NaCl (about 6 dS/m), three days later salinity was increased to 120m*M* NaCl (12dS/m) (Gregorio *et al.,* 1997). This was done by adding NaCl to the nutrient solution and pH in nutrient solution was always maintained at 5 daily. All the genotypes including checks died within 10 days. This was due to the high temperatures coupled with a dry environment (low relative humidity), which increased stress to the rice genotypes. A preliminary test experiment was therefore established to generate a screening procedure which would allow proper genotype discrimination under the prevailed condition.

During the preliminary test experiment, three setups were tested to standardize the screening system using:

i) application 2 dSm^{-1} after every 24 hours up to a maximum of 6 dSm^{-1} thereafter, maintained until the sensitive check died,

ii) 5 dSm^{-1} was applied and maintained throughout until sensitive check died,

iii) 4 dSm^{-1} was applied and maintained for 3 days and thereafter the concentration was increase to 8 dSm^{-1} and maintained until the sensitive check died.

Result from this preliminary experiment showed that a developed salinisation standard using 2 dSm^{-1} after every 24 hours up to a maximum of 6 dSm^{-1} indicated better differences among genotypes and therefore used for screening the material in the study. The study was in a split plot experiment arranged in randomized complete block design with four replications, where the main factors were salt levels and sub factors were genotypes.

(c) Data collection

Data collection was done 10 days after the maximum desired stress level was achieved. Plant growth (vigour), injury symptoms, shoot fresh and dry weight, shoot Na^+, K^+ and Na^+/K^+ were determined in both control and salt stressed plants. Scoring for salt tolerance on the basis of seedling vigour and salt injury was done on a scale at a respective growth stage of the plant as per standards (IRRI, 1988).

Plant shoots were harvested for the determination of Na^+ and K^+ concentrations at 20d after the start of salt stress treatments. Few seedlings were left in the treated solution to understand the maximum survival days for each of the tested genotypes. Harvested plant samples were dried and ground to a fine powder and about 0.1 g was transferred to a test tube containing 10 mL of 0.1 N acetic acid, and heated in a water bath at 80^oC for 2 h as describe by Ansari and Flowers (1986).

(d) Data treatment and statistical analysis

All the data were tested for normality and the parameters were submitted for analysis of variance (ANOVA) with two levels of classification (salinity and genotype). Data were analyzed using GENSTAT release 11 (Payne et al*.*, 2008) computer package. Analysis of variance (ANOVA) was performed to determine statistically significant differences among genotypes, salinity levels, and their interactions. Simple linear correlation coefficients were calculated to establish relationships between physiological attributes of genotypes under salinity.

3. Results and discussion

3.1. Genotype performance in hydroponics screening method - under saline environment

Loss of plant turgidity was observed immediately after salt imposition in treated solution. Less turgidity loss was observed in tolerant checks Pokkali and IR 67076-2B-21-2 and slightly less turgidity loss was observed in a local cultivar TXD 306; this was probably due to its highly vigorous growth therefore less salt accumulation and effects in plant tissues.

Previously, Munns (2002) reported that saline solutions impose both ionic and osmotic stresses on plants. Yeo and Flowers (1989) reported the importance of vigorous growth at early stages of plant development. They reported that vigorous growth provides a dilution effect of the salts concentration in plant tissues therefore increases survival of the plant under saline condition.

The genotypic variability for salinity tolerance was assessed and expressed as less salt injury symptoms, low Na^+ accumulation and Na^+/K^+ ratio in plant tissues and high biomass accumulation (fresh weight and dry weight). The analysis of variance (ANOVA) indicated that genotypes and salinity effects and their interactions were significant ($P < 0.001$) for all plant characteristics (Table 3). At both salt levels, mean performance of all the traits measured also showed significant ($P < 0.001$) difference among genotypes. Salinity x genotype interaction indicated that genotypes responded differently to saline stress. Rice cultivar seedlings responses were different among genotypes in both salt levels.

Table 3. Mean squares for various traits of rice genotypes under saline and non-saline nutrient solution

Source of variation	d.f.	Mean Squares %K	 %Na	 Na/K	 Survival	 Plant Vigour	 Fresh weight	 Dry weight
Rep	3	0.23	0.24	0.004	0.73	0.09	0.03	0.006
Salt Level (S)	1	165.4***	25.7***	0.20***	351.5***	311.6***	26.6***	1.36***
Error (A)	*3*	*0.28*	*0.18*	*0.009*	*0.73*	*0.09*	*0.08*	*0.005*
Genotype (G)	9	3.4***	1.18***	0.034***	14.7***	15.8***	2.06***	0.11***
G x S	9	3.6***	1.14***	0.011*	14.7***	15.8***	0.23*	0.03***
Error (B)	*54*	*0.32*	*0.10*	*0.003*	*0.4*	*0.34*	*0.09*	*0.005*

****, * =highly significant at $P< 0.001$ and significant at $p<0.05$, respectively*

In general saline stress significantly increased Na^+ accumulation in plant tissues, decreased K^+ absorption resulted in narrower Na^+/K^+ ratio, increased leaf injury symptoms and reduced seedling biomass accumulation in almost all the tested genotypes as compared to the non-saline environment (Table 4 and 5). However, a significant interaction observed for all the traits indicates that there was differential performance between genotypes under different salt environments for all the traits measured. These results indicated that genetic variability exists among the genotypes in terms of the parameters measured and genotypes behaved differently in treated and non-treated solutions. The study therefore, highlights that the traits could be used as selection criteria for salt tolerance at early growth stages of rice plant, however selection should be done under individual salt condition.

Studies have reported similar decreasing trend on measured plant characteristic from control to saline treated media in rice (Singh, 2001; Moradi et al., 2003; Mahmood et al., 2004), in wheat (Munns et al., 2006) in sorghum (Krishnamurthy et al., 2007), in brassicaceae (Ashraf and Sarwar, 2002), in grapevine (Shani and Ben-Gal, 2005).

3.2. Shoot Na+ and K+ concentration and Shoot Na+ / K+ ratio

The differences among the genotypes and salinity levels and their interactions were significant for shoot Na^+ and K^+ concentration and shoot Na^+/K^+ (Table 3). Analysis of shoot ion content showed that all the genotypes had increased levels of Na^+ and differential performance was observed in some genotypes under saline and non-saline conditions. Genotypes IR 56, Pokkali and IR67076-2B-21-2 performed well under treated solution by having minimum Na^+ accumulation in plant tissue. The rest of genotypes, for example, Gigante, IR 64 and Thailand performed well under control condition by having less shoot Na^+ concentration but performed poorer under saline condition (Figure 2). Genotype CSR 27 performed poorly than the rest of genotypes. On the basis of less than 50% reduction in shoot Na^+ concentration, better performing genotypes were the tolerant checks (Pokkali, IR67076-2B-21-2) and a local cultivar IR 56 (Table 4).

Table 4. Mean shoot Na^+, K^+ concentrations and Na^+/K^+ ratio in rice genotypes under salinized (6 dSm^{-1}) condition

Genotypes	%Na^+ conc.			%K^+ conc.			Na^+ /K^+		
	Treated	Control	% Increase	Treated	Control	% decrease	Treated	Control	% Increase
CSR 27	2.74	1.57	42.7	5.70	6.48	12.1	0.30	0.18	41.5
Gigante	2.53	0.53	79.1	4.66	7.96	41.4	0.33	0.11	65.3
IR 29	2.57	0.65	74.7	4.23	9.35	54.7	0.37	0.15	60.8
IR 56	0.93	0.65	30.1	4.61	7.14	35.5	0.11	0.08	25.6
IR 64	2.59	0.52	79.9	4.20	9.81	57.2	0.34	0.12	63.4
IR67076-2B-21-2	1.36	0.71	47.8	5.80	9.73	40.4	0.21	0.12	40.9
Nerica 1	2.47	0.97	60.7	4.55	7.40	38.5	0.46	0.20	55.8
Pokkali	1.10	0.76	30.9	4.89	7.39	33.8	0.12	0.09	20.9
Thailand	2.73	0.61	77.6	3.49	7.93	56.0	0.49	0.20	59.7
TXD 306	2.41	1.01	58.9	4.70	6.38	26.3	0.37	0.17	54.6
Sed	*0.23*			*0.41*			*0.04*		
Lsd $_{0.05}$	*0.46*			*0.82*			*0.08*		

Note: Decrease or increase (%) = (Value of control plant -value of treated)/ value of control) x 100)

Table 5. Mean plant injury, vigor and biomass in rice genotypes under salinized (6 dSm^{-1}) condition.

Genotype	Plant Injury (Rated 1-9)[1]			Plant vigour (Rated 1-9)[2]			Fresh wt (g)			Dry wt (g)		
	Treated	Control	% change	Treated	Control	% change	Treated	Control	% change	Treated	Control	% change
CSR 27	4.5	1	77.8	2.3	1	56.5	0.87	1.65	47.2	0.22	0.52	57.7
Gigante	6.9	1	82.9	6.3	1	84.1	0.85	2.32	63.3	0.30	0.67	55.2
IR 29	8.5	1	88.2	8.5	1	88.2	0.35	1.34	73.8	0.10	0.55	81.8
IR 56	3.9	1	74.0	7.9	1	87.3	0.72	1.31	45.0	0.28	0.51	45.0
IR 64	6.2	1	83.7	5.3	1	81.1	0.53	1.38	61.6	0.15	0.41	63.4
IR67076-2B-21-2	2.0	1	49.2	2.3	1	56.5	0.86	1.66	48.2	0.21	0.48	56.2
Nerica 1	8.7	1	88.5	7.3	1	86.3	0.72	2.04	64.7	0.20	0.69	71.2
Pokkali	1.2	1	13.0	1.2	1	16.7	1.83	2.70	32.2	0.60	0.76	21.1
Thailand	5.7	1	82.3	6.6	1	84.8	0.69	1.53	54.9	0.29	0.49	40.8
TXD 306	5.7	1	82.4	1.8	1	44.4	0.98	1.95	49.8	0.29	0.58	50.0
Sed	0.43			0.40			0.22			0.23		
Lsd $_{0.05}$	0.87			0.82			0.44			0.09		

Note: Decrease or increase (%) = (Value of control plant -value of treated)/ value of control) x 100)

[1] Salt injury rating:1= Growth and tillering nearly normal; 9 = Almost all plants dead or dying

[2] Seedling vigour:1 = Extra vigorous (very fast growing; plants at 5-6 leaf stage, have 2 or more tillers in majority of the population); 9 = Very weak (stunted growth; yellowing of leaves).

All the tolerant genotypes had comparatively less sodium uptake than the sensitive ones. Except for a local cultivar IR 56 which also showed good performance even more than the checks, all other local cultivars performed poorly in comparison to susceptible check (IR 29). The highest Na^+ uptake was recorded in Gigante and IR 64 which was closely followed by Thailand and IR 29. The rest of the genotypes formed the medium group regarding Na^+ uptake. In their studies Mishra et al. (2001) and Munns (2002) also reported similar results and showed that Na^+ is a highly damaging element in plant tissue and absorbed more under salt stress environment.

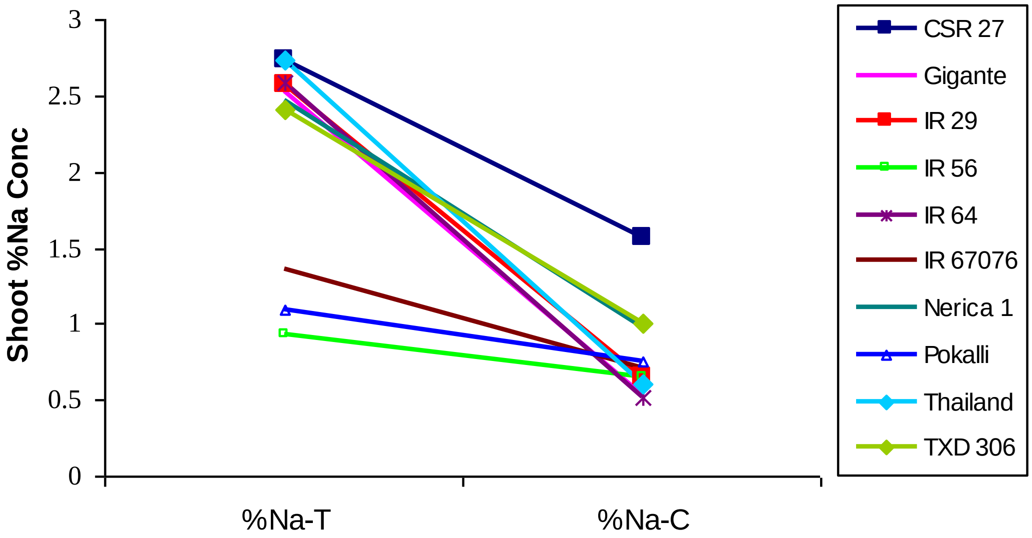

Figure 2. Changes in shoot %Na concentration of rice genotypes under saline treated (%Na-T) and non-treated (%Na-C) solution

The trend for K^+ was the reverse to that of sodium; it decreased with the increase in salinity (Figure 3). The result showed that, increase in shoot Na^+ concentration concomitantly decreased shoot K^+ concentration. Minimum reduction in K^+ uptake under saline condition was noted in CSR 27 followed by TXD 306 and Pokalli, other genotypes having higher reduction in K^+ uptake than the control. However all the rice genotypes (except IR 64, Thailand and a susceptible check IR 29) maintained less than 50 % reduction in K^+ (Table 4). The study also indicated that, increasing uptake of Na^+ causes low K^+ uptake which results in increasing Na^+/K^+ ratio in the plant shoot.

Genotypes with higher Na^+ concentrations also showed a decreased leaf K^+ concentration resulting in an increase in Na^+/K^+ ratio. There is therefore an indication of some inhibition effect on K^+ absorption under saline environments. The check genotype Pokalli had the lowest change in Na^+/K^+ between treated and non treated solution followed by IR 56. A greater change was obsesved in Gigante and the poorest performance was observed in Nerica 1 (Figure 4). Genotypes Pokalli and IR67076-2B-21-2, CSR 27 and a local cultivar IR 56 had maintained the minimum Na^+ tissue concentration, therefore narrower Na^+/K^+ ratio and minimum reduction of less than 50% (Table 4). This suggests that these genotypes are capable of taking up more K^+ under salt stress to maintain superior Na^+/K^+ balance in the shoot.

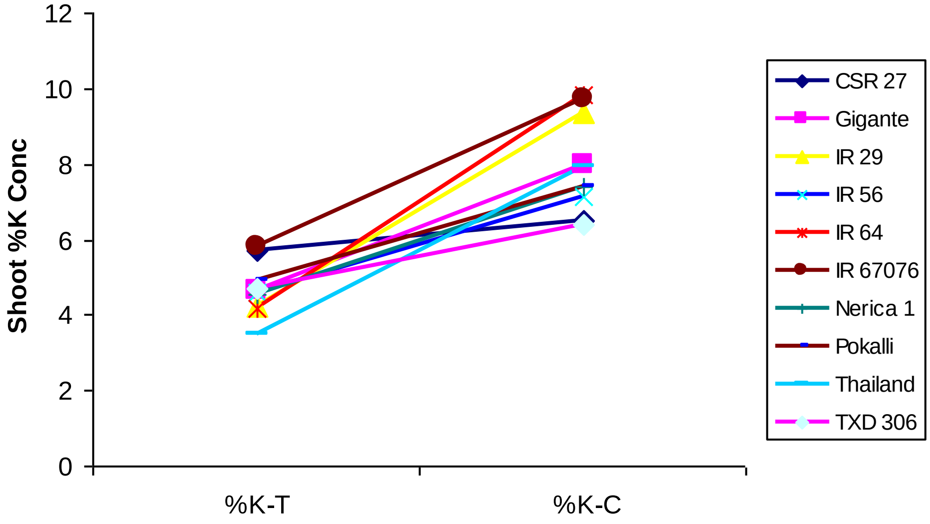

Figure 3. Changes in shoot %K concentration of rice genotypes under saline treated (%K-T) and non-treated (%K-C) solution

Similar observations were reported by Mishra et al. (2001). Blaha et al. (2000) reported that high Na^+/ K^+ ratio can disrupt various enzyme processes in the cytoplasm. Devitt et al. (1981) added that increase in Na^+ concentration in growing plants create Na^+/K^+ imbalance which adversely affects grain yield. According to Bhandal and Malik (1988), metabolic toxicity of Na^+ is largely a result of its ability to compete with K^+ for binding sites essential for cellular function. More than 50 enzymes are activated by K^+, and Na^+ cannot substitute this role. In this case, the use of Na^+ and Na^+/ K^+ ratio was taken into consideration as a useful characteristic while screening the lines.

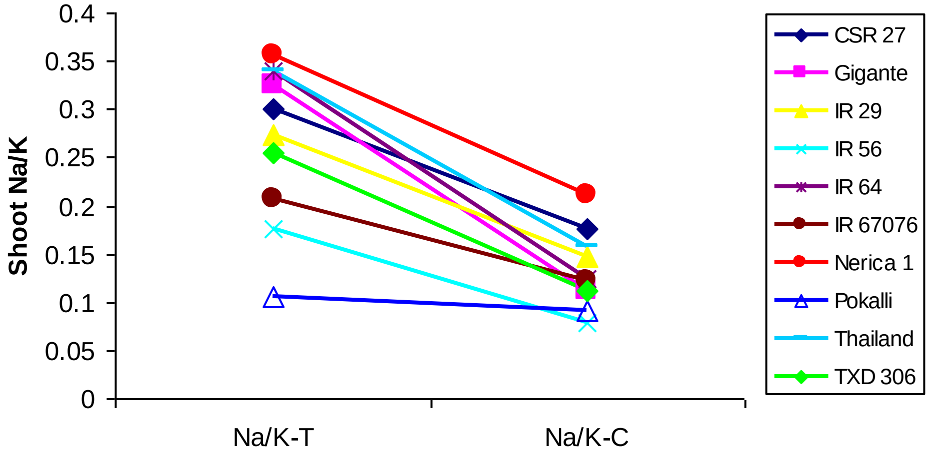

Figure 4. Changes in shoot Na+/K+ ratio of rice genotypes under saline treated (Na/K-T) and non-treated (Na/K-C) solution

3.3. Plant vigour and injury symptoms

The growth observations recoded at seedling stage showed that there was a significant liner decrease and increase in plant vigour and injury symptoms, respectively from non-treated to treated solution (Figure 5). Plants exhibited symptoms of salt injury symptoms 7- 10 days after treatment. Significant variation on plant injury was observed among genotypes whereby the tolerant checks (Pokalli, IR67076-2B-21-2, CSR 27) and a local cultivar TXD 306 showed less injury and nearly normal growth than the rest of genotypes. During the study it has been observed that salt injury started with reduction in effective leaf area (Figure 5 & 6).

Among the measured characters, plant vigour was more affected than the rest of the traits as most of the genotypes had > 50% reduction in vigour (Table 5 and 6). Only genotype Pokalli (a saline tolerant check) and TXD 306 (a local cultivar with high vigour characteristics) performed better by having < 50% reduction in vigour when compared to the control. Maximum reduction was observed in genotypes IR 29 (a susceptible check), Nerica 1 and IR 64.

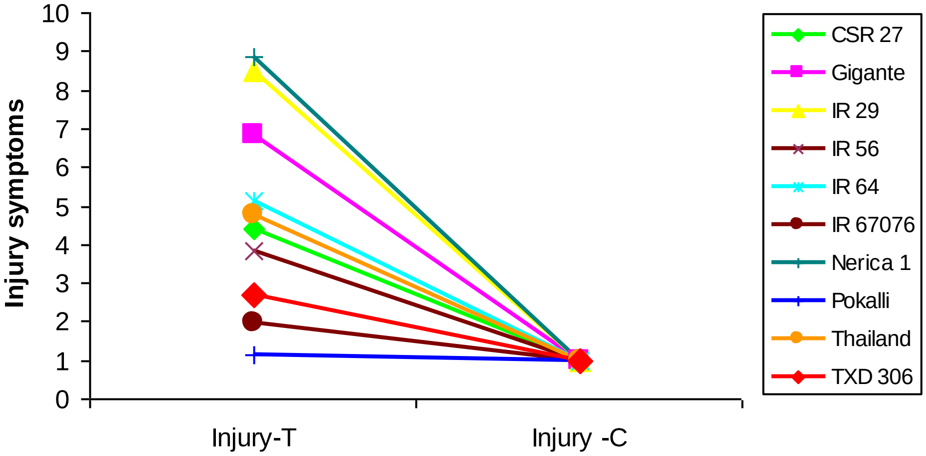

Figure 5. Changes in shoot injury symptoms of rice genotypes under saline treated (Injury-T) and non-treated (Injury-C) solution

Early vegetative vigour is important as it is associated with various combinations of rapid seedling emergence, early development, heavy tillering, and rapid increase in seedling height. Richards (1992) highlighted that breeding for more vigorous plants has been argued to be agronomically the most effective approach for increasing yield in saline soils. Furthermore, Acquaah, (2007) reported that although heritability of early vigour is low, evaluation and selection can definitely result in measurable breeding advance.

Injury symptoms were more intense on older leaves of sensitive genotypes. The older leaves started to roll followed by the next older, and so on. Finally, the survivors had the old leaves losing vitality with the youngest remaining green. All the tolerant genotypes showed minimum Na^+, low Na^+/K^+ ratio and high vigour and had also low leaf injury symptoms. Maximum injury symptoms were observed in Nerica 1 and IR

29 and only genotype Pokalli and IR67076-2B-21-2 (saline tolerant checks) showed less injury symptoms and had <50% reduction on injury symptoms (Table 5).

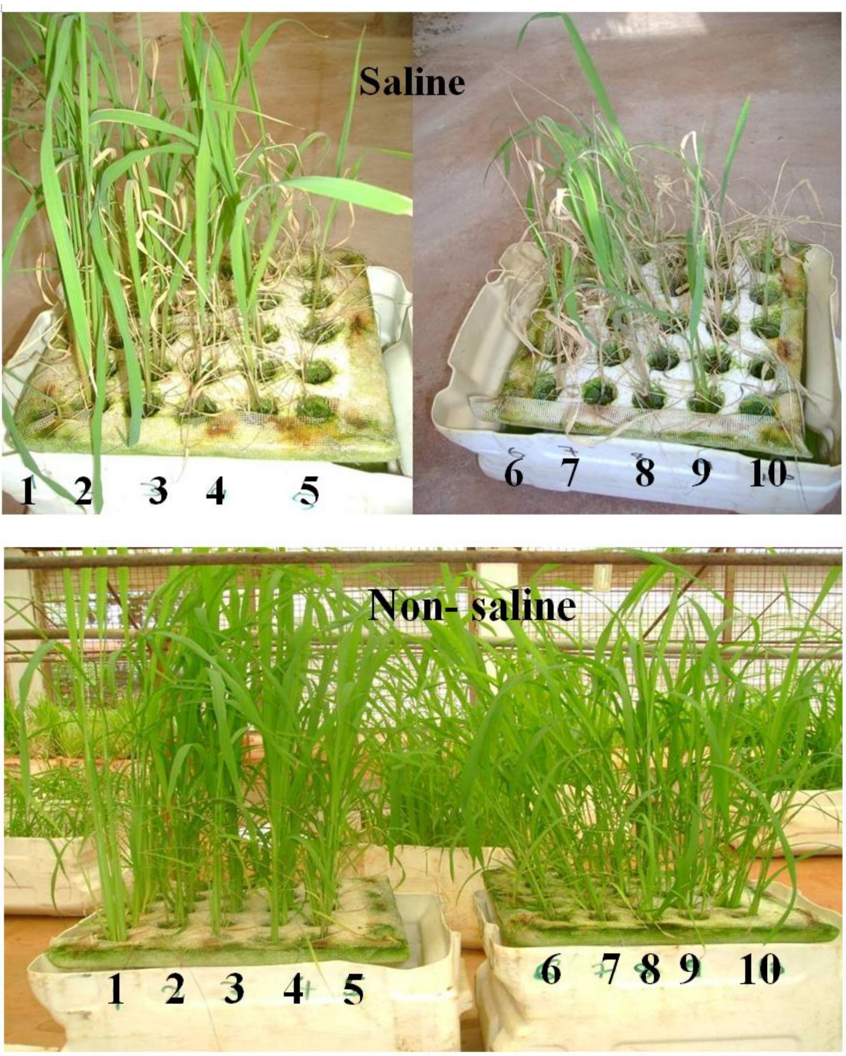

Figure 6. Plant injury symptoms of genotypes under saline and non-saline nutrient solution

3.4. Shoot fresh and dry weights

A significant variability among rice genotypes was observed in terms of shoot fresh and dry weight. A significant decrease in shoot fresh weights was observed under saline stress compared to the control. Analysis of shoot dry weight also showed a significant linear decrease and differential performance was observed among some genotype under saline and non-saline condition (Figure 7). On the basis of less than 50% reduction in shoot fresh weight, superior performances were observed in Pokkali followed by IR 56, CSR 27, IR67076-2B-21-2 and TXD 306. Minimum shoot fresh weights accumulations were observed in IR 29 and Nerica 1 (Table 5). Reduction in biomass accumulation under saline conditions has also been reported in earlier studies (Mishra et al., 2001; Blaha et al., 2000; Farshadfar et al., 2007).

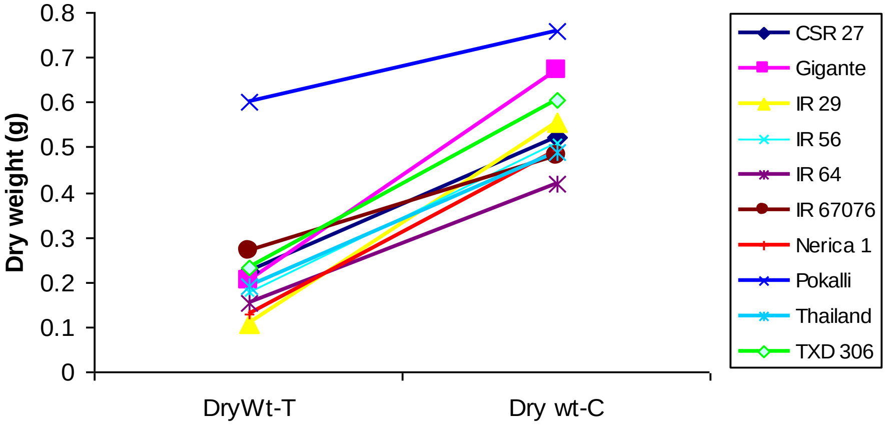

Figure 7. Changes in shoot dry weight of rice genotypes under saline treated (DryWt-T) and non-treated (DryWt-C) solution

The comparison regarding shoot ion concentration and growth parameters among the genotypes (Table 6) clearly indicated that genotype Pokkali maintained all the seven recorded parameters by having less than 50 % reduction over the control, therefore rated highly tolerant. This genotype survived longer (37d) under salinized conditions compared to all the tested materials. Pokkali was followed by IR67076-2B-21-2 which maintained five out of seven parameters and rated tolerant. Genotype IR67076-2B-21-2 had equal survival rate, injury symptoms and vigour similar to those of Pokkali (Figure 6), but had less survival days and relatively low dry matter accumulation.

The two checks genotypes were followed by IR 56 which successfully maintained four out of eight parameters and therefore rated slightly tolerant. Among the local genotypes tested, IR 56 showed significantly more superior survival and less injury than the rest. This genotype appears third after Pokkali and IR67076-2B-21-2 in terms of survival rate and injury but had significantly lower survival days, vigour and biomass accumulation than the two checks. The poorest performance was noted in the case of IR 64 and Nerica1 which did not perform well in any of the characters measured therefore rated highly susceptible.

3.5. Association of characters

In the present study association of characters was investigated under both salinized and control solutions, but only the results on association of characters under saline solution were presented in Table 7. Under control solution only significant and positive associations were observed between fresh weight and dry weight (r = 0.39**; P=0.01); between %Na^+ and Na^+/K^+ (r = 0.88***; P<0.001), and significant negative associations was observed between dry weight and Na^+/K^+ ratio (r = -0.34**; P=0.01). The rest of the traits did not show any significant association. Under saline treated solution, there was a strong negative association of all the growth parameters with the Na^+ and Na^+/K^+ ratio, indicating the negative effects of increasing Na^+ concentration and Na^+/K^+ ratio on biomass accumulation under saline conditions. Although associations between K^+ and growth parameters were significant only for plant vigour, a weak positive relationship between K^+ with shoot fresh and dry weight were still observed (Table 7). This indicates the enhancement of K^+ towards biomass accumulation in the plant under such environments.

Table 6. Rice genotypic performance calculated on the basis of less than 50 % reduction in the character measured under saline condition.

Genotype	Characters								
	Injury	Vigour	% Na	% K	Na/K	Fresh wt	Dry wt	No of variables	Rating
CSR 27	-	-	+	-	+	+	-	3	S
Gigante	-	-	-	-	-	-	-	0	HS
IR 29	-	-	-	-	+	-	-	1	HS
IR 56	-	-	+	+	+	+	-	4	ST
IR 64	-	-	-	-	-	-	-	0	HS
IR67076-2B-21-2	+	-	+	-	+	+	+	5	T
Nerica 1	-	-	-	-	-	-	-	0	HS
Pokkali	+	+	+	+	+	+	+	7	HT
Thailand	-	-	-	-	+	-	+	2	S
TXD 306	-	+	-	-	-	+	+	3	S

Note: + = Less than 50 % reduction over control, - = More than 50 % reduction over control
HT = Highly tolerant, T = Tolerant, ST = Slightly tolerant, HS = Highly sensitive ; S = Sensitive

Table 7 Phynotypic correlation coefficients of symptomatic and growth attributes of NaCl grown rice genotypes with shoot ionic content (n=10).

Plant Characteristics	**%K^+**	**%Na^+**	**Na^+/ K^+**	**DW**	**FW**	**PI**
%K^+	1					
%Na^+	-0.34*	1				
Na^+/ K^+	-0.24*	0.88***	1			
Dry weight (DW)	0.14ns	-0.49**	-0.34**	1		
Fresh weight (FW)	0.25ns	-0.37**	-0.23*	0.82***	1	
Plant Injury (PI)	-0.08ns	0.62***	0.35**	-0.54***	-0.60***	1
Plant Vigor (PV)	0.29*	-0.45**	-0.30*	0.55***	0.65***	-0.84***

****,** , *and ns = significant at 0.1%, 1%, 5% and not significant*

Krishnamurthy et al. (2007) reported similar findings on weak association of shoot K^+ concentration with biomass accumulation. Plant injury showed a strong positive relationship with Na^+ and Na^+/K^+ ratio, but a weak and negative one with K^+ and the K^+/Na^+ ratio, as has been observed in rice (Mahmood et al., 2004), green gram (Misra and Dwivedi, 2004) and other crops (Farshadfar et al., 2007). This confirms the detrimental effect of high Na^+ concentrations in the plant tissues. A highly significant positive correlation coefficient was found between plant vigour and shoot fresh weight, shoot dry weight and K^+ concentration in plant. On the other hand significant inverse associations were observed between plant vigour and plant injury, shoot Na^+ concentration and Na^+/K^+ ratio. Studies have reported similar inverse associations between Na^+ and $Na/^+K^+$ on survival and biomass accumulation in the plant, for example in cowpea genotypes (Murillo-Amador et al., 2002) in green gram (Misra and Dwivedi, 2004), in barley (Farshadfar et al., 2007) and in sorghum (Krishnamurthy et al., 2007). This indicates that a more vigorous and less injured the plant, increases the chance to survive, grow and accumulate high biomass under salt stress environment.

4. Conclusion

From these experiments it can be concluded that:

1. Genotypes Pokkali, IR67076-2B-21-2 and IR 56 showed outstanding performance in saline condition, therefore makes them useful donors for salt tolerance.
2. Among the local farmer preferred varieties, IR 56 had superior performance. Based on its ability to grow well, absorbing less Na^+ and maintain high K^+ content under salinity stress, and maintain good tillering ability, IR 56 can be used to breed for tolerance in saline condition.
3. Plant vigour, less injury symptoms, shoot fresh and /or dry weight, low leaf Na^+ and low Na^+/K^+ concentrations are good indicative characteristics of a better performing genotype at the vegetative stages. It can therefore conclude that that, these traits could make good criteria for selection of salt tolerant materials under saline condition.

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