



Effect of Salinity on Growth and Protein Content of Rice Genotypes

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Abstract

Rice, the highest producing cereal crops in Bangladesh. Saline induced changes in growth was screened at germination to find out the salt tolerant traditional rice genotypes and chemical tests were carried out to detect the protein-content of rice genotypes. Four traditional rice genotypes (Kalijira, Sakkor khana, Chinigura, Ghoto balam) with one improved variety BRRI dhan28 were used in this experiment. The germination experiment was conducted in petri dish at 0, 8, 12, 16 dS/m salinity with three replications each. Based on the data obtained, the genotypes were scored and categorized as highly tolerant (score 1), tolerant (score 3), moderately tolerant (score 5), susceptible (score 7), and highly susceptible (score 9) to salinity. Salinity caused reduction in germination and declines of shoot length, root length, shoot wet weight and shoot dry weight. Mean salinity tolerance score (MSTS) was computed. Based on MSTS, two genotypes named Kalijira and Chinigura were selected as tolerant, two genotypes named Sakkor khana and BRRI dhan28 as susceptible, and the rest one genotype named Ghoto balam as highly susceptible to salinity in germination stage. Total protein content of rice genotypes were determined by Kjeldahl method. Among 5 varieties studied, Chinigura contains the highest amount of protein (8.855%). The overall data estimated that, Chinigura is more salt tolerant and highly protein rich genotype than other rice genotypes.

Key words : Salinity, Growth, Protein content, Rice , Genotype

Introduction

Rice is one of the most widely grown crops in coastal areas frequently inundated with saline sea water during high tidal period. Agricultural land use in those areas is very poor, which is much lower than country's average cropping intensity (Petersen and Shireen, 2001). The coastal areas of Bangladesh cover more than 30% of the cultivable lands of the country. About 53% of the coastal areas are affected by different degrees of soil salinity. The USDA Salinity Laboratory defines a saline soil as having EC of 4 dS/m or more (US Salinity Lab. Staff, 1954)

Khan and Abdullah (2003) reported that rice crop identified as salt-susceptible both in seedling and reproductive stages. In the most commonly cultivated rice cvs., young seedlings were very sensitive to salinity (Flowers & Yeo, 1981; Lutts *et al.*, 1995). The literature indicates that rice is sensitive to salinity, particularly during the seedling stage (Maas and Hoffman 1977)

Salinity is a major abiotic stress for all stages of rice due to anthropogenic contributions global warming, the rate of sea-level rise is expected to increase and possess dramatic effect on rice production especially in coastal areas (Hakim *et al.*, 2013).

Rice generally tolerates salinities ranging between 1.9-3.0 dS/m (Grattan, 2002). High salinity causes both hyperionic and hyperosmotic stresses and can lead to plant death (Hasegawa *et al.* 2000). Salinity reduces the growth of plant through osmotic effects, reduces the ability of plants to take up water and this causes reduction in growth. Na^+ and Cl^- are the principal ions in majority of salt affected soils, which mainly affect plants growth. The roots of rice plants readily absorb Na^+ which are distributed in all plant organs to pose ion damage, osmotic stress and imbalance nutrition (Siringam *et al.*, 2011). If excessive amount of salt enters the plant, the concentration of salt will eventually rise to a toxic level in older transpiring leaves causing premature senescence and reduce the photosynthetic leaf area of a plant to a level that cannot sustain growth (Munns, 2002; Shereen *et al.*, 2005). Melo *et al.*, (2004) revealed significant variability among 93 rice genotypes in terms of salt tolerance during the germination phase . Agronomical measures alone can't be able to solve the problem.

Development of salt tolerant varieties has been considered as one of the strategies to increase rice production in saline prone coastal areas. As rice is mainly used in Asian countries for major energy source because of its higher starch content but both the quantity and the quality of protein are matters of importance to everyone, particularly to dietitians, physicians, housewives, school and institutional workers, food and feed manufacturers, animal husbandry men. Unless the diet contains enough protein of satisfactory quality, proper nutrition will not result. The total food protein and energy production per hectare were found higher in rice than in wheat. In the developing and under developed countries, cereals supply more than 50 per cent of protein and in the ensuing years, it will, probably, remain the dominant source of protein for nearly two third of world (Jamuna, 1996).



The research program was undertaken with the following objectives:

- i. To detect salt-tolerant traditional rice genotypes.
- ii. To determine the protein content of traditional rice genotypes.

Materials and Methods

The seeds of 5 rice genotypes were collected from farmers of Sreerampur village of Dumki Upazilla of Patuakhali district. The germination experiment was conducted at the laboratory of the Department of Biochemistry and Food Analysis, PSTU. Chemical analysis of all the rice genotypes was performed in the laboratory of Department of Soil Science, and Central laboratory, PSTU. The details of the salinity stress methods and laboratory experiments are described below.

3.1 Experimental Period and Location

The germination experiment was conducted at the laboratory of the Department of Biochemistry and Food Analysis, PSTU during the period of 04 October 2015 to 15 October 2015. Chemical analysis of those rice genotypes for determining protein content was conducted then in the laboratory of Department of Soil Science, and Central laboratory, PSTU.

3.2 Study Material

Among the 5 rice genotypes, 4 rice genotypes are traditional genotypes cultivated in the coastal region of Bangladesh and the rest 1 variety is improved variety by BRRI. Each of 5 rice genotypes was given a serial number which is same in all the experimental stages. The name of rice genotypes and their serial number is given below.

Table 3.1 List of rice genotypes used in the study

SI No.	Genotype	Type of genotype
1	Kalijira	Landrace
2	Sakkor khana	Landrace
3	Chinigura	Landrace
4	Ghoto Balam	Landrace
5	BRRI dhan28	HYV

3.3 Salinity treatments

Four salinity levels such as (control), 8, 12, and 16 dS/m were maintained in the experiment. The different salinity levels are obtained by dissolving crude salt (collected from seashore) in tap water. During dissolving salt with tap water EC value was frequently measured with EC meter so that prefixed salinity level could be maintained. The control was maintained using tap water.

3.4 Breakdown of seed dormancy

Prior to germination test seeds were subjected to breakdown of dormancy. For this seeds were sun dried for five days.

3.5 Germination Test

Germination test was carried out in Petri dishes of 15 cm diameter. A layer of cotton was set at the bottom of the each Petri dish. Twenty seeds were placed on cotton bed and 15ml of treatment solutions of different salinity levels were poured in each Petri dish to immerse the seeds partially for ensuring proper aeration. The Petri dishes were placed on a table in the laboratory. The seeds were allowed to germinate at room temperature ($25\pm 2^{\circ}\text{C}$). Different salinity water was added to the respective Petri dish everyday as and when necessary. Seeds were considered germinated when radicals measured 2 mm. The number of seeds germinated was recorded at an interval of 24 hours.

3.6 Collection of Data

3.6.1 Percent germination

Number of seeds germinated was recorded at 8 days after sowing and the results were converted into percentage.

3.6.2 Shoot and root length

The young seedlings were up-rooted at 11 days after sowing (DAS). Ten randomly selected plants of each genotype per replication were used for data recording. Root length was measured from base to top of radical whereas shoot length was measured from the base to the tip of the longest leaf of the plumule.

3.6.3 Shoot fresh weight and dry weight

Shoot fresh weight of each up-rooted seedlings was measured and recorded at the same day when up-rooted shoot length was measured. Then the up-rooted seedlings samples were placed to oven at 65°C for 3 days. The dry weight (mg) of shoot of each seedling of genotype was recorded and expressed on the basis of individual treatment.

3.7 Determination of protein

3.7.1 Method: Kjeldahl method was used to determine the nitrogen content and used the conversion factor to determine the protein content from nitrogen. BUCHI digest system K-437 and BUCHI distillation unit K-350 was used for doing this experiment.

Result and Discussion

The experiment was conducted inside the laboratory. Seeds were sown in petri dish. Four salinity levels like 0, 8, 12, and 16 dS/m were maintained. Percent germination was recorded up to 8th days of incubation, whereas observation on other parameters were recorded on 11th day of incubation. The data obtained and derived from the experiment, the rice genotypes were categorized into five groups following the standard evaluation system (IRRI 1996). The groups were named highly tolerant, tolerant, moderately tolerant, susceptible, and highly susceptible. For each group a numerical value (score) in scale of 1 to 9 was assigned.

4.1 Germination percentage

Salinity stress significantly reduced the germination percentage of the rice genotypes. Generally in control condition, the percentage of germination was found the highest and it was gradually decreased with the increase of salt concentration. The rate of germination of rice genotypes in control variety from 85-100%, being the lowest in Ghoto balam. At salinity levels 8, 12, and 16 dS/m germination were 85-95%, 70-90%, and 35-60% respectively. Thus, in the present study the lowest germination percentage was observed in 16 ds/m salinity. Table 4.1 shows that, the mean germination percentage after 8 days of incubation at 0,8,12, and 16 dS/m salinity were 94, 91.6, 83.2, and 43.6 % respectively.

For screening, the rice genotypes were scored based on the result of germination percentage where seeds were maintained at 16 dS/m salinity for 8 days. The range of reductions in germination percent were 30.0-36.9%, 37.0-43.9%, 44.0-50.9%, 51.0-57.9%, and 58.0-65.9% denoting them as highly tolerant, tolerant, moderately tolerant, susceptible, and highly susceptible genotypes and corresponding tolerance scores were 1, 3, 5, 7, and 9 respectively. Based on the score value, one genotype was found as highly tolerant (score 1), one genotype was found as moderately tolerant (score 5), three genotypes were found as highly susceptible (score 9) to salinity stress. The result clearly indicates that, germination percentage is mostly affected by salinity.

Table 4.1 Germination percentage of rice genotypes as influenced by different levels of salinity

Sl. No.	Rice Genotypes	0 dS/m	8 dS/m	12 dS/m	16 dS/m	% decrease at 16 dS/m	^a Score at 16 dS/m
1	Kalijira	100	95	88	50	50	5
2	Sakkor khana	100	93	85	38	62	9
3	Chinigura	95	95	90	60	36.8	1
4	Ghoto balam	85	90	83	35	58.8	9
5	BRRRI dhan28	90	85	70	35	61.1	9
	Mean	94	91.6	83.2	43.6	53.74	

^aScore: 30.0-36.9%: 1; 37.0-43.9%: 3; 44.0-50.9%: 5; 51.0-57.9%: 7; 58.0-65.9%: 9

4.2 Shoot length

Shoot length was gradually decreased with the increase of the salt concentration of the growth medium. At 0, 8, 12, and 16 dS/m salinity, the mean value of shoot length were 8.19, 2.67, 1.18, and 0.48 cm respectively (Table 4.2). Under 16 dS/m salinity the reduction of shoot length range from 89.17-97.65% with a mean value of 93.17%. To categorize the rice genotype on the basis of their capability to minimize the decreasing effect (on shoot length), tolerance index table was constructed as (89.0-90.9%: 1; 91.0-92.9%: 3; 93.0-94.9%: 5; 95.0-96.9%: 7; 97.0-98.9%: 9) indicates highly tolerant, tolerant, moderately tolerant, susceptible, and highly susceptible genotypes respectively. Based on the score value at 16 dS/m of salinity it was found that, two genotypes were highly tolerant, two genotypes were moderately tolerant, and one genotype was highly susceptible.

Table 4.2 Effect of salinity on shoot length of rice genotypes in germination stage

Sl. No.	Rice Genotypes	Shoot length (cm/plant)				% decrease over control			^a Score at 16 dS/m
		0 dS/m	8 dS/m	12 dS/m	16 dS/m	8 dS/m	12 dS/m	16 dS/m	
1	Kalijira	5.63	3.74	1.56	0.61	33.57	72.29	89.17	1
2	Sakkor khana	6.88	2.63	1.18	0.38	61.77	82.85	94.48	5
3	Chinigura	7.26	2.28	1.28	0.75	68.60	82.37	89.67	1
4	Ghoto balam	14.92	3.53	1.05	0.35	76.34	92.96	97.65	9
5	BRR1 dhan28	6.27	1.15	0.83	0.32	81.66	86.76	94.90	5
	Mean	8.19	2.67	1.18	0.48	62.59	83.45	93.17	

^aScore: 89.0-90.9%: 1; 91.0-92.9%: 3; 93.0-94.9%: 5; 95.0-96.9%: 7; 97.0-98.9%: 9

4.3 Root length

Root length was gradually decreased with the increase of the salt concentration of the growth medium. At 0, 8, 12, and 16 dS/m salinity, the mean value of root length were 5.94, 2.97, 1.65, and 0.58 cm respectively (Table 4.3). Under 16 dS/m salinity the reduction of root length range from 77.17-96.29 % with a mean value of 87.25%. To categorize the rice genotype on the basis of their capability to minimize the decreasing effect (on root length), tolerance index table was constructed as (77.0-80.9%: 1; 81.0-84.9%: 3; 85.0-88.9%: 5; 89.0-93.9%: 7; 94.0-97.9%: 9) indicates highly tolerant, tolerant, moderately tolerant, susceptible, and highly susceptible genotypes respectively. Based on the score value at 16 dS/m of salinity it was found that, two genotypes were highly tolerant, two genotypes were susceptible, and one genotype was highly susceptible.

Table 4.3 Effect of salinity on root length of rice genotypes in germination stage

Sl. No.	Rice Genotypes	Root length (cm/plant)				% decrease over control			^a Score at 16 dS/m
		0 dS/m	8 dS/m	12 dS/m	16 dS/m	8 dS/m	12 dS/m	16 dS/m	
1	Kalijira	3.11	2.53	1.65	0.71	18.65	46.95	77.17	1
2	Sakkor khana	4.93	3.34	1.59	0.51	32.25	67.75	89.66	7
3	Chinigura	4.50	2.24	1.39	0.86	50.22	69.11	80.89	1
4	Ghoto balam	12.12	4.28	2.04	0.45	64.69	83.17	96.29	9
5	BRR1 dhan28	5.04	2.44	1.58	0.39	51.59	68.65	92.26	7
	Mean	5.94	2.97	1.65	0.58	43.48	67.01	87.25	

^aScore: 77.0-80.9%: 1; 81.0-84.9%: 3; 85.0-88.9%: 5; 89.0-93.9%: 7; 94.0-97.9%: 9

4.4 Shoot fresh weight

Shoot fresh weight was determined at 11th days after incubation. It was gradually decreased with the increase of the salt concentration of the growth medium. Thus the mean reduction percent of shoot weight was the lowest in 8 dS/m (70.02%) of salinity and was the highest in 16 dS/m (96.42%) (Table 4.1.4). At salinity levels 8, 12, and 16 dS/m, the ranges of shoot fresh weight reductions were (54.76-81.48%), (76.19-91.05%), and (91.63-99.98%) respectively. Percent decrease of shoot fresh weight under different levels of salinity greatly varied among the genotypes. The highly tolerant genotypes had the minimum decrease and that of higher decrease in highly susceptible genotypes (Table 4.4). For screening, tolerance index table was constructed as (90.0-91.9%: 1; 92.0-93.9%: 3; 94.0-95.9%: 5; 96.0-97.9%: 7; 98.0-99.99%: 9) indicates highly tolerant, tolerant, moderately tolerant, susceptible, and highly susceptible genotypes respectively. Based on the score value at 16 dS/m of salinity it was found that, one genotype was highly tolerant, one genotype was susceptible, and three genotypes were highly susceptible.

Table 4.4 Effect of salinity on shoot fresh weight of rice genotypes in germination stage

Sl. No.	Rice Genotypes	Shoot wet weight (mg/plant)				% decrease over control			^a Score at 16 dS/m
		0 dS/m	8 dS/m	12 dS/m	16 dS/m	8 dS/m	12 dS/m	16 dS/m	
1	Kalijira	21.00	9.50	5.00	1.50	54.76	76.19	92.86	9
2	Sakkor khana	22.00	7.50	4.00	0.51	65.91	81.82	97.68	7
3	Chinigura	24.00	6.00	4.50	2.01	75.00	81.25	91.63	1
4	Ghoto balam	47.00	12.72	4.50	0.01	72.94	90.43	99.98	9
5	BRRI dhan28	21.00	3.89	1.88	0.01	81.48	91.05	99.95	9
	Mean	27.00	7.92	3.98	0.81	70.02	84.15	96.42	

^aScore: 90.0-91.9%: 1; 92.0-93.9%: 3; 94.0-95.9%: 5; 96.0-97.9%: 7; 98.0-99.99%: 9

4.5 Shoot dry weight

Shoot dry weight was determined at 14th days after incubation. It was gradually decreased with the increase of the salt concentration of the growth medium. Thus the mean reduction percent of shoot weight was the lowest in 8 dS/m (65.33%) of salinity and was the highest in 16 dS/m (97.90%) (Table 4.5). At salinity levels 8, 12, and 16 dS/m, the ranges of shoot dry weight reductions were (50.00-85.37%), (75.00-93.90%), and (92.50-99.99%) respectively. Percent decrease of shoot dry weight under different levels of salinity greatly varied among the genotypes. The highly tolerant genotypes had the minimum decrease and that of higher decrease in highly susceptible genotypes (Table 4.5). For screening, tolerance index table was constructed as (92.0-93.5%: 1; 93.6-95.0%: 3; 95.1-96.6%: 5; 96.7-98.2%: 7; 98.3-100%: 9) indicates highly tolerant, tolerant, moderately tolerant, susceptible, and highly susceptible genotypes respectively. Based on the score value at 16 dS/m of salinity it was found that, one genotype was highly tolerant, one genotype was susceptible, and three genotypes were highly susceptible.

Table 4.5 Effect of salinity on shoot dry weight of rice genotypes in germination stage

Sl. No.	Rice Genotypes	Shoot dry weight (mg/plant)				% decrease over control			^a Score at 16 dS/m
		0 dS/m	8 dS/m	12 dS/m	16 dS/m	8 dS/m	12 dS/m	16 dS/m	
1	Kalijira	4.00	2.00	1.00	0.30	50.00	75.00	92.50	1
2	Sakkor khana	3.00	1.45	0.30	0.001	51.67	90.00	99.97	9
3	Chinigura	3.40	0.90	0.65	0.10	73.53	80.88	97.06	7
4	Ghoto balam	7.70	2.61	0.80	0.001	66.10	89.61	99.99	9
5	BRRI dhan28	4.10	0.60	0.25	0.001	85.37	93.90	99.98	9
	Mean	4.44	1.51	0.60	0.08	65.33	85.88	97.90	

^aScore: 92.0-93.5%: 1; 93.6-95.0%: 3; 95.1-96.6%: 5; 96.7-98.2%: 7; 98.3-100%: 9

4.6 Mean salinity tolerance score (MSTS)

An attempt was undertaken to compute the mean of score values of all the parameters studied in germination stage. The score values and mean salinity tolerance score (MSTS) values are presented in Table 4.6. To stay away from the fraction in figures, mean salinity tolerance score values from 1-2, 2-4, 4-6, 6-8 and above 8 were acknowledged as 1, 3, 5, 7, and 9 respectively.

It appeared from the table that, no genotypes were found as highly tolerant, two genotypes scored 3 (tolerant), no genotypes were found as moderately tolerant, two genotypes scored 7 (susceptible), and only one genotype scored 9 (highly susceptible) to salinity. The genotypes found as tolerant to high degree of salinity are Kalijira and Chinigura.

Table 4.6 Mean salinity tolerance score of growth parameters of rice genotypes at germination stage

Sl. No.	Rice genotypes	% Germination	Shoot length	Root length	Shoot fresh weight	Shoot dry weight	Mean score
1	Kalijira	5	1	1	9	1	3 (3.4)
2	Sakkor khana	9	5	7	7	9	7 (7.4)
3	Chinigura	1	1	1	1	7	3 (2.2)
4	Ghoto balam	9	9	9	9	9	9 (9.0)
5	BRRRI dhan28	9	5	7	9	9	7 (7.8)

4.7 Protein Content of rice genotypes

Total protein content of rice grains were determined by Kjeldahl method. From the Table 4.7 it is cleared that, among 5 varieties studied, Chinigura contains the highest amount of protein (8.855%) and Sakkokr khana, and BRRRI dhan28 contains the same lowest amount of protein (6.682%).

Table 4.7 Protein Content of rice genotypes

Sl. No.	Rice Genotypes	% Nitrogen	% Protein
1	Kalijira	1.176	6.762
2	Sakkor khana	1.162	6.682
3	Chinigura	1.540	8.855
4	Ghoto balam	1.260	7.245
5	BRRRI dhan28	1.162	6.682

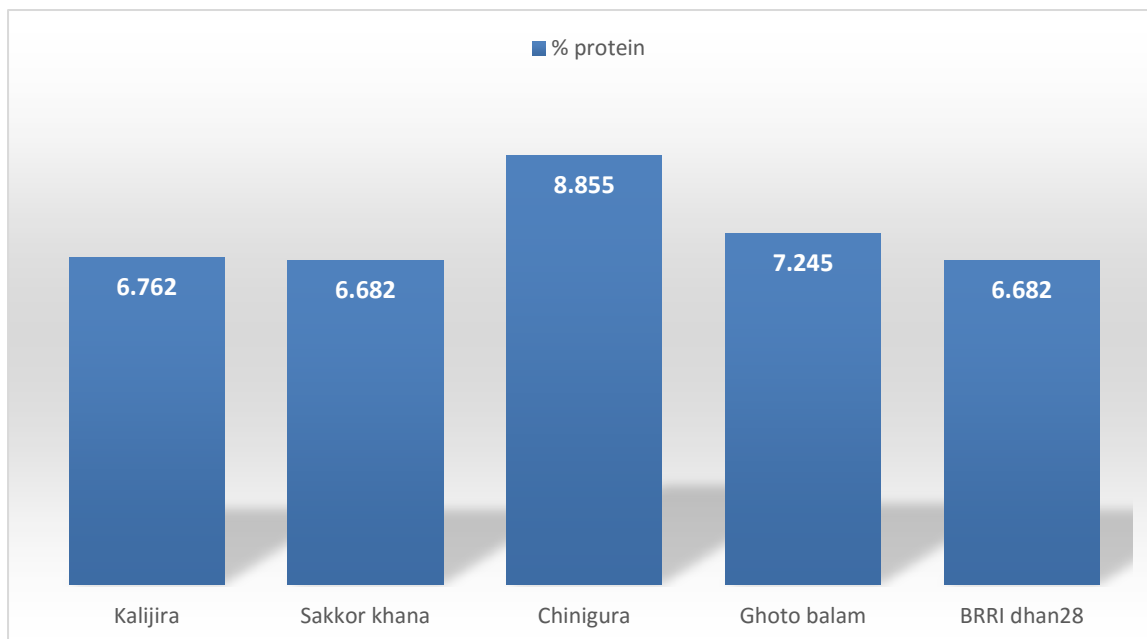


Figure 4.7 Protein Content of rice genotypes

Conclusion

Salinity stress reduced the germination percentage of the rice genotypes. Under lower salt concentration germination percentage reached the maximum value within short time in compare to higher salt concentration. Percent germination was reduced as an average of 53.74% at 16 dS/m. Shoot length, shoot fresh weight, shoot dry weight and root length were also reduced due to salinity stress. The inhibitory effect of salt was more pronounced in shoot than root. Based on mean salinity tolerance score (MSTS), none genotypes were found as highly tolerant, two genotypes named Kalijira and Chinigura were found as tolerant, two genotypes named Sakkor khana and BRRRI dhan28 as susceptible, and the rest one genotype named Ghoto balam as highly susceptible to salinity in germination stage. Total protein content of rice grain were determined by Kjeldahl method. Among 5 varieties studied, Chinigura contains the highest amount of protein



(8.855%). The overall data estimated that, Chinigura is more salt tolerant and highly protein rich genotype than other rice genotypes.

Future research need

- Field performance of the selected rice genotypes in high saline soils of Bangladesh
- Development of highly nutritious and high yielding salt tolerant rice varieties using selected salt tolerant traditional rice genotypes

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