

EFFECTS OF SODIUM CHLORIDE SALINITY ON WATER RELATIONS AND ION ACCUMULATION IN TWO MUNGBEAN VARIETIES DIFFERING IN SALINITY TOLERANCE

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Abstract

Salt tolerance in relation to water status and plant nutrients of two mungbean varieties, BARImung 2 (salinity sensitive) and BUMung 2 (salinity tolerant) was evaluated. The seeds were grown in pots and treated with NaCl levels of 0 (control), 100 and 200 mM. Different parameters related to water relations as well as mineral nutrients were measured. The exudation rate and relative water content were decreased but water saturation deficit was increased by salinity in both the varieties. In BARImung 2 plants, the exudation rate and relative water content were lower but water saturation deficit was higher than those in BUMung 2 at both 100 and 200 mM NaCl levels. Salinity also influenced the accumulation of Na, K, Ca and Mg in leaves, stems and roots of the two said mungbean varieties. Sodium accumulation was increased in all the plant-parts of both the varieties in the order of stem > root > leaf but in BUMung 2 the accumulation was lower than that of BARImung 2 except in root. Potassium accumulation decreased in all parts of both the mungbean varieties but that was lower in BUMung 2 than that of BARImung 2. The contents of Ca and Mg in all the plant-parts increased more in BUMung 2 than those of BARImung 2 with the increase of salinity levels. All these results indicated that high salt tolerance in BUMung 2 was associated with its better water status, more or less uniform mineral nutrient (Ca and Mg) distribution in different plant-parts than that in BARImung 2.

Key words: NaCl salinity, Water relation, Ion accumulation, Mungbean varieties, Salinity tolerance

Introduction

Salinity is one of the major environmental stresses, which affects plant growth and development by disturbing water relations, creating imbalance in plant nutrition and affecting plant physiological and biochemical processes (Karim *et al.* 1993, Munns 2005). Under saline conditions plant suffers from osmotic shock due to lower osmotic potential in the soil solution (Orcutt and Nilsen 2000, Rahnema *et al.* 2010). Islam (2001) pointed out that relative water content (RWC), water saturation deficit (WSD), water retention capacity (WRC) and water uptake capacity (WUC) were affected by salinity in blackgram and mungbean.

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At the whole plant level, salinity stress frequently induces an increase in Na and Cl contents as well as a decrease in K, Ca, N and Pi concentrations (Martinez and Lauchli 1994, Ahmad and Prasad 2012). Generally, Ca and K ions are decreased in plants under saline conditions (Al-Harbi 1995, James *et al.* 2011). In contrast, Ashraf and Rauf (2001) reported that under saline conditions concentrations of Na, K and Ca increased significantly in all parts of the maize seedlings. Usually salinity tolerant variety of a crop shows better water relations, low accumulation of Na, imbalance in the accumulation of nutrient elements, higher photosynthesis and finally better growth than those in salt sensitive ones. In glycophytes, salinity resistance is associated with a restriction of toxic ion absorption at the root level (Schachtman and Munns 1992) vis a vis minimum accumulation of Na and Cl in the shoot of the plant (Akita and Cabsulay 1990).

Leguminous crops are generally sensitive to salinity though there is considerable difference in salt tolerance between legume species (Maas and Hoffman 1977). Mungbean is an important glycophyte legume crop in Bangladesh. Despite the availability of a substantial number of reports on the effect of salinity on mungbean (Raptan *et al.* 2001, Kabir *et al.* 2005), the salt tolerance behavior and growth mechanisms of this crop remain unclear. To clarify salinity tolerance behavior in the present study a sensitive and a tolerant variety of mungbean were treated with NaCl salinity. The objectives of this study were to analyze the influence of salinity on water relations and mineral ion accumulation in different plant parts of two mungbean varieties differing in their salinity tolerance.

Materials and Methods

Two mungbean varieties, BARI mung 2 (salinity sensitive) and BUMung 2 (salinity tolerant) were used for this experiment (Sultana *et al.* 2007). The plants were grown in pots in the research field of Botanical Garden of Jahangirnagar University. The physiological and biochemical analyses were carried out at the Plant Physiology and Plant Biochemistry laboratory of Botany Department, Jahangirnagar University and the Chemistry and Soil Science laboratory of Bangabandhu Sheikh Mujibur Rahman Agricultural University, Gazipur, Bangladesh. Each pot was filled with 12 kg of soil along with compost (made of grass, leaves and cowdung), one fourth of the soil by volume. Basal dose of fertilizers, 40 kg N, 60 kg P and 40 kg K per hectare in the form of urea, triple super phosphate and muriate of potash, respectively were thoroughly mixed with the soil. The total amount of urea was applied in two splits. The soil samples were air dried, crushed and passed through a 3 mm-pore-diameter sieve. The pots were kept under

natural sunshine till harvesting (up to pre-flowering stage). There were six treatment combinations, which were comprised of two mungbean varieties and three levels of salinity. The three salinity levels were non-saline, control and 0 mM, and two salinity treatments, 100 and 200 mM NaCl. The experiment was laid out in a completely randomized design (CRD) with three replications.

Four seeds of uniform size were directly sown in the pot and then the pots were watered for easy germination. After seedling establishment, only two better seedlings were allowed to grow in each pot. Then intercultural operation, weeding and pest control measures were taken as and when necessary. The germinated seedlings were watered up to 28th day of seedling emergence. Then watering was stopped. From the 32- to 42nd days after emergence (DAE), which was before flowering stage, the mungbean plants in each treated pot were irrigated with 100 ml of NaCl solutions every day. Salt solutions were prepared artificially by dissolving 5.85 and 11.7 g/l of commercially available NaCl with distilled water to make 100 and 200 mM NaCl solutions, respectively. Tap water was served as non-saline control. To study the physiological parameters, three plants of each treatment of each variety were taken for collecting data at pre-flowering stage.

Measurement of exudation rate: Exudation rate was measured at 5 cm above from the stem base of mungbean plant. At first dry cotton was weighed. A slanting cut on stem was made with a sharp knife. Then the weighed cotton was placed on the cut surface. The exudation of sap was collected from the stem for 1 hr at normal temperature. The final weight of the cotton with sap was taken. The exudation rate was measured by deducting cotton weight from the sap containing cotton weight and expressed in per hour basis.

Measurement of plant water status: The fresh leaves of same sized and same aged of five plants from each treatment were carefully separated. Fresh weights of leaf segments were taken. The collected leaf segments were kept immersed in distilled water for 24 hrs at room temperature in the dark. The turgid weights of those parts were then measured. Afterwards all the leaf materials were oven-dried at 80°C for 72 hrs in order to take dry weight. The fresh, turgid and dry weights of the leaf segments were used to determine the relative water content (RWC) and water saturation deficit (WSD) following Sangakkara *et al.* (1996).

Analyses of mineral ions concentration in different plant-parts: For analysis of Na, K, Ca and Mg, oven dried plant materials (leaves, stems and roots) at harvest were ground with a mortar and pestle. 500 mg ground samples for each ion were taken in a conical flask with 5 ml nitric-per chloric acid (nitric acid + perchloric acid, 5 : 1) dry-ashed at 200 - 220°C for 2 hrs at sand bath for digestion. After digestion distilled water was added,

dissolved and made the digest up to 100 ml. Then 10 ml solution and 2 ml lanthanum chloride ($\text{LaCl}_3 \cdot 7\text{H}_2\text{O}$) were taken in a 100 ml conical flask and by adding distilled water the solution was made up to 100 ml. Then liquid sample was taken in a vial test tube and absorbance of respective ions was measured with Atomic Absorption Spectrophotometer (Model 170-80, Hitachi) following the methods of Yamakawa (1992).

Data were analyzed statistically following randomized complete block design using ANOVA procedure in SAS statistical software.

Results and Discussion

The data on the effect of salinity on water status, exudation rate and water saturation deficit in mungbean varieties are presented in Table 1. Exudation rate in salt treated BARImung 2 plants was much lower than that of BUMung 2 at both 100 and 200 mM NaCl levels. Compared to control plants, exudation rates of BUMung 2 decreased by 39.13 and 86.96%, while those in BARImung 2 were decreased by 74.02 and 96.06% at 100 and 200 mM NaCl, respectively (Table 1). Decreased exudation rate means lower water uptake by plants. Reduction in water uptake by plants due to salt stress has been reported by Islam (2001). Higher exudation rates in BUMung 2 disclosed that BUMung 2 plants could absorb more water than BARImung 2 under saline condition. Similar results were reported by Sangakkara *et al.* (1996) and Faruquei (2002).

Relative water content (RWC) was greater in plants grown at control than the plants grown under salinity stress (Table 1). The leaf of BUMung 2 and BARImung 2 showed identical RWC at control treatment. However, at the high salt concentration the leaf of BARImung 2 showed higher reduction (45.25%) in RWC than that of BUMung 2 (32.63%). Decreased in RWC due to salinity was reported by Kabir *et al.* (2005) in mungbean and Islam (2001) in mungbean and blackgram. It is well known that salinity decreases water potential of soil solution and plant cannot uptake water freely, and consequently RWC decreased (Orcutt and Nilsen 2000). In the present study, results indicated that BARImung 2 was found to suffer more from water stress than BUMung 2. The better water retention in BUMung 2 under saline condition obviously contributed for maintenance of higher plant growth than in BARImung 2. This result is in agreement with the report of White and Izquierdo (1991).

Water saturation deficit (WSD) showed an inverse trend of RWC (Table 1). WSD indicates the degree of water deficit in plants. Salinity increased the WSD in both the varieties compared to that of control treatment. However, the higher salt (200 mM)

treated plants of BARImung 2 showed relatively higher WSD (283.25%) than that of BUMung 2 (258.04%) over control. This finding reveals that BARImung 2 suffered more from water deficit especially at high salt concentration than BUMung 2. Similar results were reported by Islam (2001) and Kabir *et al.* (2005).

Table 1. Effect of NaCl salinity on exudation rate, relative water content (RWC) and water saturation deficit (WSD) of two mungbean varieties at pre-flowering stage.

Variety	Salinity level (mM)	Exudation rate (mg/min)	Changes over control (%)	Relative water content (RWC)	Changes over control (%)	Water saturation deficit (WSD)	Changes over control (%)
BARImung 2	0	2.54 a		85.08 a		10.33 d	
	100	0.66 d	-74.02	60.13 c	-29.33	34.46 c	233.59
	200	0.10 e	-96.06	46.58 d	-45.25	39.59 b	283.25
BUMung 2	0	2.07 b		89.67 a		14.92 d	
	100	1.26 c	-39.13	65.54 b	-26.91	39.87 b	167.23
	200	0.27 e	-86.96	60.41 c	-32.63	53.42 a	258.04

In a column followed by common small letters do not differ significantly at 5% level of significance.

Accumulation of mineral ions in different plant-parts: Results of Na accumulation of two mungbean varieties are presented in Table 2. Compared to control, Na accumulation was increased with increasing salinity levels for both the varieties. At high salt concentration however Na accumulation was higher (50 and 455.56%) in leaf and stem parts of BARImung 2 than that in BUMung 2 (31.11 and 316.67%). The tolerant variety, BUMung 2 accumulated less amount of Na in most of the plant parts except root compared to BARImung 2 (Table 2). Blum (1988) also reported that tolerant crop accumulated less amount of Na than susceptible one. The findings of Karim *et al.* (1992) in triticale and Raptan *et al.* (2001) in blackgram and mungbean indicated that tolerant cultivar maintained relatively larger Na in the root and a smaller amount in the shoot compared to the salt-susceptible cultivar. In contrast to shoot, Na concentration was greatly reduced in leaf. The reduction was higher in BARImung 2 compared to that of BUMung 2 (Table 2). This observation indicated that the translocation of Na from shoot to the leaf was regulated efficiently in BUMung 2 compared to BARImung 2. Probably this might be the reason for higher tolerance of BUMung 2 to salinity stress.

Table 2. Effect of NaCl salinity on Na accumulation in two mungbean varieties at pre-flowering stage.

Variety	Salinity level (mM)	Na in leaf (mg/g)	Changes over control (%)	Na in stem (mg/g)	Changes over control (%)	Na in root (mg/g)	Changes over control (%)
BARImung 2	0	0.050 a		0.063 d		0.605 c	
	100	0.068 b	36.00	0.233 b	269.84	0.783 b	29.42
	200	0.075 c	50.00	0.350 a	455.56	0.947 a	56.53
BUmung 2	0	0.045 a		0.060 d		0.220 d	
	100	0.054 a	20.00	0.168 c	180.00	0.663 c	201.36
	200	0.059 b	31.11	0.250 b	316.67	0.795 b	261.36

In a column followed by common small letters do not differ significantly at 5% level of significance.

Results of K accumulation in two mungbean varieties are presented in Table 3. The table shows that K accumulation was decreased by salinity in all parts of both the varieties of mungbean. With the increase in salinity from 0 to 200 mM, the reduction of K in all parts of BARImung 2 was higher compared to those of BUMung 2 (Table 3). At high salt concentration the reduction percentage of K in leaf, stem and root of BARImung 2 were 28.50, 44.41 and 49.19 whereas those were 24.37, 37.90 and 36.78 in BUMung 2, respectively (Table 3). These results indicated that BUMung 2 accumulated higher K ion than that of BARImung 2.

Salinity induced reduction in K accumulation was reported in forage crops (Datta *et al.* 1996), and blackgram and mungbean (Raptan *et al.* 2001). Under saline conditions plant cells utilize K as a metabolite to maintain turgor to escape from osmotic shock (Blum 1988). In fact, the ability to maintain metabolically significant concentration of K may be essential for salt tolerance in glycophytes (Zhang and Blumward 2001, Daşgan *et al.* 2002).

Results of Mg accumulation of two mungbean varieties are presented in Table 4. Mg accumulation was increased almost in all parts except root with the increasing salinity levels for both the varieties. However, Mg accumulation was lower in BARImung 2 than that of BUMung 2. In leaves and stems BUMung 2 accumulated higher (27.37 and 21.39%) Mg contents than those of BARImung 2 (15.32 and 14.51%, respectively) at 200 mM NaCl. The decrease in Mg content in roots of BARImung 2 was higher (46.43%) than that of BUMung 2 (22.63%) over control (Table 4). These results indicated that BUMung 2 accumulated higher Mg than that of BARImung 2. Raptan *et al.* (2001)

reported an increasing pattern of Mg accumulation in blackgram and mungbean under saline conditions, though Patil *et al.* (1995) did not find any influence of salinity on Mg accumulation in greengram. Mg accumulation in plant organs is probably helpful to maintain the osmoregulation to protect the plant cells from the osmotic shock caused by salinity (Greenway and Munns 1980).

Table 3. Effect of NaCl salinity on K accumulation in two mungbean varieties at pre-flowering stage.

Variety	Salinity level (mM)	K in leaf (mg/g)	Changes over control (%)	K in stem (mg/g)	Changes over control (%)	K in root (mg/g)	Changes over control (%)
BARImung 2	0	1.428 c		1.448 c		2.893 b	
	100	1.207 c	-15.48	1.243 c	-14.23	2.122 c	-26.65
	200	1.021 d	-28.50	0.805 d	-44.41	2.033 c	-49.19
BUmung 2	0	2.692 a		3.166 a		3.622 a	
	100	2.442 b	-9.29	2.866 b	-9.48	2.948 b	-18.61
	200	2.036 b	-24.37	1.966 c	-37.90	2.290 c	-36.78

In a column followed by common small letters do not differ significantly at 5% level of significance.

Table 4. Effect of NaCl salinity on Mg accumulation in two mungbean varieties at pre-flowering stage.

Variety	Conc. of NaCl (mM)	Mg in leaf (mg/g)	Changes over control (%)	Mg in stem (mg/g)	Changes over control (%)	Mg in root (mg/g)	Changes over control (%)
BARImung 2	0	0.359 c		0.317 c		0.659 a	
	100	0.398 bc	10.86	0.343 c	8.20	0.577 b	-12.44
	200	0.414 b	15.32	0.363 bc	14.51	0.353 d	-46.43
BUmung 2	0	0.380 bc		0.402 b		0.486 c	
	100	0.468 a	23.16	0.458 a	13.93	0.437 b	-10.08
	200	0.484 a	27.37	0.488 a	21.39	0.376 d	-22.63

In a column followed by common small letters do not differ significantly at 5% level of significance.

Table 5. Effect of NaCl salinity on Ca accumulation in two mungbean varieties at pre-flowering stage.

Variety	Conc. of NaCl (mM)	Ca in leaf (mg/g)	Changes over control (%)	Ca in stem (mg/g)	Changes over control (%)	Ca in root (mg/g)	Changes over control (%)
BARImung 2	0	3.680 c		2.160 c		3.270 a	
	100	4.640 b	26.09	2.620 b	21.30	1.860 b	-43.12
	200	5.240 ab	42.39	3.000 a	38.89	1.466 c	-55.17
BUmung 2	0	3.223 c		0.979 d		1.375 cd	
	100	4.349 b	34.94	1.738 c	77.53	1.225 d	-10.91
	200	5.540 a	71.89	2.560 b	161.49	1.171 d	-14.84

In a column followed by common small letters do not differ significantly at 5% level of significance.

Results of Ca accumulation of two mungbean varieties are presented in Table 5. The table shows that Ca accumulation was increased in leaves and stems with the increasing salinity levels for both the varieties, except roots. At high concentration of NaCl (200 mM), the percentages of Ca ion in leaf and stem of BUMung 2 were 71.89 and 161.49 whereas those in BARImung 2 were 42.39 and 38.89, respectively (Table 5). Similar results were reported by Raptan *et al.* (2001) in blackgram and mungbean. Salinity induced reduction of calcium uptake was reported by Patil *et al.* (1995) in greengram. Blum (1988) found that like some other elements Ca also acts as metabolite to protect plant cells from the osmotic shock in different crops under saline stress. The increase in Ca contents in leaves and stems was higher in BUMung 2 than those of BARImung 2 and the decrease of Ca content in roots was higher in BARImung 2 than that of BUMung 2 over control (Table 5). These results revealed that BUMung 2 accumulated higher Ca than that of BARImung 2 which indicates the high tolerance of BUMung 2 to salinity stress. Lutts *et al.* (1996) reported that Ca content was decreased with salinity in salt-sensitive genotypes while they remained at constant levels in salt-resistant ones.

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