

SCREENING OF COUNTRY BEAN (*LABLAB PURPUREUS* L.) GENOTYPES FOR SALINITY TOLERANCE

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Abstract

Thirty-five country bean (*Lablab purpureus* L.) genotypes collected from coastal belts of Bangladesh were screened for tolerance to a high level of salinity (12 ds m⁻¹ NaCl) in Hogland nutrient solution in a hydroponic system at vegetative growth stage. The experiment was conducted in a complete randomized design with three replications at Genetics and Plant Breeding lab, BSMRAU during January-February, 2016. The genotypes were varied in biochemical properties (proline, carbohydrates, chlorophyll contents etc.), dry root: shoot ratio, relative shoot dry weight and salt susceptibility index (SSI). Wide variation among the genotypes was noticed for root and shoot ratio ($p < 0.05$). Relative shoot length varied from 29.87- 126.04 with an average of 79.29 whereas relative root length showed wide variation with a range of 20.00-300.00. Relative root dry weight (RRDW) varied from 22.06-211.43 with a mean of 79.40 and relative shoot dry weight (RSDW) ranged from 26.57-192.66 with an average of 69.29. Relative root : shoot ratio showed a range of 0.59 to 2.51. On the basis of relative performance of root-shoot dry matter, dry leaves weight, root-shoot ratio, six genotypes were found highly tolerant, seven were moderately tolerant, sixteen were moderately susceptible and six were highly susceptible to salinity stress. Based on visual observation of plants, relative shoot dry weight (RSDW), proline content, total sugar and soil plant analysis development (SPAD) value the genotypes CB031, CB035, CB003, CB023, CB026, CB028, CB013, CB030, CB014, CB024, CB019, CB020 and CB002 were selected as tolerant for further evaluation and use in breeding work.

Keywords: Lablab bean, hyacinth bean, hydroponics, salt stress, coastal regions, yield.

Introduction

Salinity limits the productivity of agricultural crops worldwide which adversely affect on seed germination, plant vigour and crop yield (Majeed and Muhammad, 2019). Soil salinity is jeopardizing the capacity of agriculture to sustain the flourishing human population increase (Dehnavi *et al.*, 2020). Salinity affects plant growth by facilitating intake of toxic ions

and hindering many metabolic, physiological and enzymatic activities. Besides, salinity also increases osmotic potential which results in decreasing absorption of water (Tesfaye *et al.*, 2014).

Globally, every year land becomes non-productive at alarming rate due to accumulation of salt, and so is the case in Bangladesh too. Salinity is regularly affecting

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about one million hectares of lands only in coastal areas of south Bangladesh. These vast areas must be brought under cultivation for increasing total production to attain food security. Two ways are available such as reclamation of salinity and cultivation of salt tolerant crops. The rectification of saline soil is a difficult, complex and expensive process. The other option is possible and feasible to bring salt affected soil into cultivation through the introduction of salt-tolerant species and cultivars capable of tolerating the higher salinity levels (Mannan *et al.*, 2010; Shibli *et al.*, 2021). Screening of available landraces or germplasm of a crop for salt tolerance helps to identify a tolerant cultivar which may, in turn sustain a reasonable yield on salt affected soils (Rasel *et al.*, 2021) and may be used in developing new salt tolerant crops for saline belts of Bangladesh.

Different cultivars of country bean are used for different purpose. Some cultivars are grown for green vegetables while others are preferred as seed. It has been estimated that, country bean seed contain 19-31% protein, 2% fat, 61% carbohydrate (includes 5% fibres) as well as adequate levels of vitamins and minerals (Kumar *et al.*, 2014). Malnutrition is a common phenomenon of the people of Bangladesh due to developing economy in nature. Thus leguminous crops can play an important role to meet up the deficiency of protein. The protein content of lablab bean is much higher which is nearly three folds of cereals. Beside rice, county bean (*Lablab purpureus* L.) is also growing widely in the coastal areas. Hence, screening of the local germplasm of county bean followed by using them through suitable breeding methods for developing salt tolerant cultivar with

high yield potential may increase the total production in the winter (rabi) season in the coastal belt.

Materials and Methods

The experiment on screening of country bean genotypes against salinity was conducted in the laboratory of Genetics and Plant Breeding of BSMRAU during January-February, 2016. Thirty-six genotypes (denoted as CB001 to CB036), previously collected from coastal regions of Bangladesh under BAS-USAD-PALS project which preserved and maintained at the department were used in the experiment. The experiment was laid-out in a complete randomized design (CRD) with three replications. The seed were sown in a perforated plastic glass containing perlite at the bottom which filled with pit moss up and watered. The seeds of CB018 were not germinated. Ten days old seedlings in plastic bags were transferred to buckets contained Hogland solution (7L/bucket) as nutrient solution medium (Table 1) in two sets following Mannan *et al.* (2010) and Dsouza and Devaraj (2015). The plastic glass put on the bucket in such a way that each bag dived in the solution by an inch so that upper portion remains aerated sufficiently (non-circulating hydroponic systems). One set was treated as control (without NaCl) and the other set was having salt stress. For initial setup of the seedlings under hydroponic condition, no salt was added to the Hogland solution for first four days in both sets. After that, the salinity was maintained at 4 ds m⁻¹ for 5-11 days, then 8 ds m⁻¹ for 12-18 days and finally 12 ds m⁻¹ for 19-25 days by adding NaCl to the Hogland solution in the second sets maintaining pH

Table 1. Composition of Hogland Solution

Name of chemicals	Formulae	For 135 Liter
Calcium Nitrate	$\text{Ca}(\text{NO}_3)_2, 4\text{H}_2\text{O}$	135.00 g
Potassium dihydrogen monophosphate	KH_2PO_4	36.45 g
Ethylene diamine tetra acetic acid Ferric Sodium	$\text{C}_{10}\text{H}_{12}\text{FeN}_2\text{NaO}_8$	10.80 g
Magnesium Sulphate	$\text{MgSO}_4, 7\text{H}_2\text{O}$	71.40 g
Potassium Nitrate	KNO_3	78.30 g
Copper Sulphate	$\text{CuSO}_4, 5\text{H}_2\text{O}$	0.054 g (54.0mg)
Zinc Sulphate	$\text{ZnSO}_4, 7\text{H}_2\text{O}$	0.0594 g (59.4mg)
Manganese Sulphate	MnSO_4	0.8235 g (823.5mg)
Ammonium heptaMolybdate	$(\text{NH}_4)_6\text{Mo}_7\text{O}_{24}, 4\text{H}_2\text{O}$	0.0513 g (51.3 mg)
Boric Acid	H_3BO_3	0.243 g (243mg)

The relative tolerance was calculated using the following formula: Relative tolerance = (variable measured in salt treated plant) / (variable measured in control plant) x 100.

6.5 to 7.0. The buckets were watered carefully to maintain the water levels to seven liters and salt was added if necessary to maintain the respective salinity levels. Leaf SPAD (Soil Plant Analysis Development) values were measured with a portable chlorophyll meter (Minolta SPAD 502) after 18-day after treatment imposition. The pH and salinity (EC, electrical conductivity ds m^{-1}) was measured by a portable water conductivity and soil activity meter (Model HI 99310, Hanna Instrument). The salinity and pH were checked every day morning. The plants were harvested after thirty days of salt treatment imposition. Morphological parameters like root length and shoot length were estimated from fresh samples. An electronic balance (Model-Citizen XK3190-A7M) was used to measure fresh and dry weights of root and shoot.

All the genotypes were categorized into four different salinity tolerant groups based on their percent relative shoot dry weight (% RSDW) according to Ashraf and Waheed (1990) and Mannan *et al.* (2010) as follows:

Scale	(%) RSDW	Tolerance group
1	> 80%	Salinity tolerant
2	60 - 80%	Moderately tolerant
3	40 - 60%	Moderately susceptible
4	< 40%	Salinity susceptible

Salinity Susceptibility Index (SSI) for shoot, root and total dry weight of each genotype was calculated as follows: $\text{SSI} = (1 - Y_{ss}/Y_{ns})/ \text{SII}$, where Y_{ss} and Y_{ns} are mean dry weight of a given genotype in salinity stressed (ss) and non-stressed (ns) environment, respectively. SII (Salinity Intensity Index) = $1 - X_{ss}/X_{ns}$, where X_{ss} and X_{ns} are the mean of all genotypes under salinity stressed (ss) and non-stressed environments (Fisher and Maurer, 1978).

Determination of proline, total sugar and photosynthetic pigments

Total sugar was determined from leaf extracts (Irigoyen *et al.*, 1992, Somogyi, 1952). Free proline was estimated in leaf samples (Bates *et al.*, 1973), which were homogenized in 5 ml sulphosalicylic acid (3%) using mortar

and pestle. With about 2 ml of extract in a test tube, 2 ml of glacial acetic acid and 2 ml of ninhydrin reagent were added. In a water bath at 100°C, the mixture was boiled for 30 min and allowed to cool. Six (6) ml of toluene was added in cool reaction mixture and the combination transferred to a separating funnel. After thorough mixing, the chromophore containing toluene was separated and the absorbance read at 520nm in a spectrophotometer against a toluene blank. A portable SPAD meter (Minolta SPAD 502, Japan) was used to measure chlorophyll content as SPAD value.

Statistical analyses

All data were analyzed using MSTAT-C the SAS Institute Inc. Version 6.12 Software.

Results and Discussion

Relative growth parameters

The relative values of range, mean, standard error, standard deviation, variance, CV (%) of some relative plant characteristics of the 35 country bean genotypes are shown in Table 2. Wide variations among the genotypes

for different relative plant parameters were observed. Relative shoot length varied from 29.87- 126.04 with an average of 79.29 having CV 30.71%. Relative root length showed wide variation with the range 20.00-300.00 had CV%, 42.16. Dry leaves weight (RLDW) had also great variation like shoot and root length. Relative root dry weight (RRDW) varied from 22.06-211.43 with a mean of 79.40. The relative shoot dry weight (RSDW) ranged from 26.57-192.66 with an average of 69.29 having CV, 58.67%. Relative root: shoot ratio showed a range of 0.59 to 2.51 with CV value of 33.59%. Relative SPAD value varied from 70.43-149.74 with a mean of 103.01 with 19.39% CV. The root and shoot growth reduced abruptly in salt sensitive plants and this effect did not appear to depend on salt concentration in the growing tissues, it was rather a response to the osmolarity of the external solution as opined by Munns (2005).

The relative performance of the country bean genotypes subjected to salt stress showed a wide range of variation. In most of the genotypes, salinity increased for most of the parameters value considerably. Most of the

Table 2. Relative plant characteristics of 35 country bean genotypes subjected to salinity stress

Relative characters	Range	Mean	Std Error	Std Dev	Variance	F-value	CV%
Shoot length	29.87-126.04	79.29	4.48	24.35	592.81	**	30.71
Root length	59.38-290.91	129.76	10.07	54.70	2992.25	**	42.16
Dry wt. of leaves (RLDW)	20.00-300.00	92.93	11.71	63.60	4044.41	**	68.43
Dry shoot weight (RSDW)	26.57-192.66	62.29	6.73	36.54	1335.48	**	58.67
Dry root weight (RRDW)	22.06-211.43	79.40	8.03	43.64	1904.04	*	54.96
Root:shoot ratio	0.59-2.51	1.34	0.08	0.45	0.20	*	33.59
SPAD Value-2	70.43-149.74	103.01	3.68	19.97	398.79	*	19.39

plant components were affected by salinity and consequently reduced shoot dry weight as well as root : shoot ratio of the genotypes. Such negative or deleterious effects of salinity on plant characters were also reported earlier in many crop species e.g., in soyabean (Mannan *et al.*, 2010 and 2013), in mungbean (Sultana *et al.*, 2009), in pepper (Chookhampaeng, 2011), in sugerbeet (Jamil *et al.*, 2007). Salt stress caused the reduction in the growth and development of country bean plants might be due to an increased uptake of toxic sodium (Khan *et al.*, 2013). The NaCl is instantly dissolved in water solvent yielded toxic Na⁺ which is easily absorbed into root tissues. These ions transport throughout plant organs, leading to toxic ion damage, osmotic stress and nutritional imbalance (Chaum *et al.*, 2007) resulting retardation in vegetative growth. Tester and Davenport (2003) noticed that leaves were more vulnerable than roots to Na⁺ simply because Na⁺ and Cl⁻ accumulated to higher levels in shoots than in roots.

Photosynthesis is heavily affected in plants growing under saline conditions. Reduced photosynthesis under salinity causes stomata closure which leads to a reduction of intercellular CO₂ concentration. Salt affects photosynthetic enzymes, chlorophyll and carotenoids reported by Stepien and Klobus (2006). Generally, chlorophyll and carotenoid pigments are reduced in susceptible genotypes but increased in tolerant genotypes under salt stress. Country bean genotypes varied significantly among themselves for leaf chlorophyll content (SPAD values). Relative chlorophyll content (SPAD value) varied from 70.43 (CB001) to 149.74 (CB022) with a mean of 103.01 having CV (19.39%) (Table

2). In our experiments, 14 genotypes (CB008, CB013, CB015, CB019, CB022, CB024, CB025, CB026, CB027, CB028, CB029, CB030, CB032, CB033, CB034, CB035 and CB036) had SPAD value greater than overall mean (103.01) to be considered as salt tolerant. Sodium chloride at high concentrations usually cause osmotic stress by decreasing water potential within the cells, and ionic stress due to specific inhibition of metabolic processes. Reduced photosynthesis under salinity can also be attributed to a decrease in chlorophyll content. Heideri (2012) reported that salinity reduced the chlorophyll content in salt susceptible plants and increased it in salt tolerant plants. The genotypes CB001, CB002 and CB003 contained relative SPAD values around 75.0 (70.43, 76.44 and 73.59, respectively) indicated that they were the most susceptible to salt stress. Photosynthesis pigments (a, b, carotenene) were reduced by increasing salinity levels from 0 to 6 ds/m in basil was reported by Heideri (2012). The chlorophyll a and chlorophyll b content decreased 38, 27% and 32, 32% respectively as a result of the 5 dSm⁻¹ salt application as compare to control in walnut (Akca and Samsunlu, 2012).

Visual observations on leaves after 4 weeks of salt stress were recorded (Table 3). Upon saline stress, necrotic symptoms on leaves were observed in susceptible genotypes. Chlorotic symptoms in leaves were observed in 23 genotypes (CB001, CB002, CB004, CB005, CB006, CB007, CB 008, CB009, CB011, CB012, CB014, CB017, CB020, CB021, CB022, CB023, CB026, CB027, CB028, CB029, CB033, CB034 and CB036) while no chlorosis or leaves remained normal were noticed in 12 (CB003, CB010,

Table 3. Relative SPAD (chlorophyll content) and visual observation of leaves (4WAS) in country bean genotypes under 12.0 dsm⁻¹ salinity stress

Genotypes	SPAD value			Visual observation of leaves (4WAS)				
	Relative value	Genotypes	Relative value	Genotypes	Relative value	Genotypes	Relative value	Relative value
CB001	70.43	CB020	84.88	CB001	Chlorotic	CB020	Chlorotic	Chlorotic
CB002	76.44	CB021	93.65	CB002	Chlorotic	CB021	Chlorotic	Chlorotic
CB003	73.59	CB022	149.74	CB003	Normal	CB022	Normal	Chlorotic
CB004	84.88	CB023	101.60	CB004	Chlorotic	CB023	Chlorotic	Chlorotic
CB005	80.66	CB024	114.44	CB005	Chlorotic	CB024	Chlorotic	Normal
CB006	100.40	CB025	124.06	CB006	Chlorotic	CB025	Chlorotic	Normal
CB007	89.66	CB026	102.81	CB007	Chlorotic	CB026	Chlorotic	Chlorotic
CB008	129.09	CB027	112.20	CB008	Chlorotic	CB027	Chlorotic	Chlorotic
CB009	78.26	CB028	104.87	CB009	Chlorotic	CB028	Chlorotic	Chlorotic
CB010	106.35	CB029	134.19	CB010	Normal	CB029	Normal	Chlorotic
CB011	94.96	CB030	101.44	CB011	Chlorotic	CB030	Chlorotic	Normal
CB012	92.17	CB031	88.49	CB012	Chlorotic	CB031	Chlorotic	Normal
CB013	126.21	CB032	112.75	CB013	Normal	CB032	Normal	Normal
CB014	83.91	CB033	104.10	CB014	Chlorotic	CB033	Chlorotic	Chlorotic
CB015	105.69	CB034	110.41	CB015	Normal	CB034	Normal	Chlorotic
CB016	99.68	CB035	121.75	CB016	Normal	CB035	Normal	Normal
CB017	100.32	CB036	115.83	CB017	Chlorotic	CB036	Chlorotic	Chlorotic
CB019	121.27	CB019	Normal	CB019	Normal			
Range		70.43-149.74		Range		-		
Mean		103.01		Mean		-		
SE		3.68		SE		-		
SD		19.97		SD		-		
Variance		398.79		Variance		-		
CV (%)		19.39		CV (%)		-		

CB013, CB015, CB016, CB019, CB 024, CB025, CB030, CB031, CB032 and CB035) genotypes.

Biochemical parameters

The major physiological processes viz. photosynthesis, protein synthesis and energy and lipid metabolisms are affected during the onset and development of salt stress with in a plant (Parviaz and Satyawati, 2008). Plants accumulated an array of metabolites including proline, carbohydrates, chlorophyll contents etc. when exposed to stressful condition (temperature, salinity, alkalinity, drought, cold, pathogen infection etc) (Hayat *et al.*, 2012). Proline is a proteinoous amino acid with an exceptional conformational rigidity which protects the plants from various stresses and also helps plants to recover from stress more rapidly (Hayat *et al.*, 2012). Proline contributes to stabilizing sub-cellular structures (e.g., membranes and proteins), scavenging free radicals and buffering cellular redox potential under stress conditions apart from acting as an osmolyte for osmotic adjustment (Ashraf and Foolad, 2007). It may also function as protein compatible hydrotrope (Singh *et al.*, 2017) beside alleviating cytoplasmic acidosis and maintaining appropriate NADP⁺/NADPH ratios compatible with metabolism (Hare and Cress, 1997). Many researchers opined that proline accumulation under salt stress were correlated with stress tolerance in many plant species, and its concentration were shown to be generally higher in salt tolerant than in salt sensitive plants. Its accumulation normally occurs in cytoplasm where it functions as molecular chaperons stabilizing the structure of proteins and its accumulation

buffers cytosolic pH and maintains cell redox status. It has also been proposed that its accumulation may be part of a stress signal influencing adaptive responses. The relative proline content and total sugar of 35 country bean genotypes under 12 dS m⁻¹ salt stress are presented in Table 4.

In the present experiment, the relative proline accumulation in country bean genotypes varied from 17.65 (CB002) to 1110.09 (CB019) with as average of 483.32 having CV value of 54%. It is well known that proline accumulation under salt stress has been correlated with stress tolerance in many plant species viz. alfalfa (Hayat *et al.*, 2012) and its concentration has been shown to be generally higher in salt tolerant than in salt sensitive plants. In our study, 18 genotypes (CB001, CB04, CB016, CB017, CB019, CB021, CB022, CB024, CB025, CB027, CB028, CB029, CB031 and CB035) had higher proline accumulation than overall mean (483.32) to be considered as salt tolerant. The genotypes CB002 and CB014 had the low value (17.65 and 73.16, respectively) indicated they were the most susceptible to salt stress. Osmotically proline is very active and contributes to membrane stability and mitigates the effect of NaCl on cell membrane disruption. Hayat *et al.* (2012) also reported that proline was widely occurred in higher plants and accumulated in larger amounts than other amino acids which regulated the accumulation of useable N.

The range, mean, coefficient of variation for relative total sugar content in country bean genotypes were 30.58 (CB024) to 310.78 (CB015), 160.14 and 12.61%, respectively (Table 5). For the various organic osmotica, sugars contribute up to 50% of the total osmotic

Table 4. Relative proline and total sugar content in country bean genotypes under 12dsm⁻¹ salinity stress

Genotypes	Proline content			Total sugar			
	Relative value	Genotypes	Relative value	Genotypes	Relative value	Genotypes	
CB001	567.57	CB020	331.60	CB001	138.82	CB020	302.86
CB002	17.65	CB021	985.71	CB002	153.32	CB021	106.65
CB003	430.91	CB022	608.79	CB003	147.79	CB022	116.74
CB004	505.88	CB023	420.55	CB004	118.65	CB023	137.95
CB005	270.52	CB024	590.98	CB005	272.34	CB024	30.58
CB006	460.14	CB025	710.24	CB006	211.52	CB025	140.14
CB007	464.96	CB026	478.11	CB007	216.49	CB026	83.47
CB008	361.38	CB027	776.58	CB008	156.30	CB027	144.44
CB009	355.06	CB028	617.53	CB009	246.98	CB028	116.75
CB010	255.65	CB029	524.44	CB010	148.55	CB029	107.48
CB011	273.47	CB030	618.05	CB011	171.77	CB030	106.44
CB012	334.93	CB031	625.72	CB012	172.78	CB031	103.23
CB013	241.19	CB032	417.52	CB013	225.53	CB032	160.44
CB014	73.16	CB033	237.62	CB014	163.22	CB033	83.46
CB015	222.32	CB034	272.04	CB015	310.78	CB034	151.67
CB016	786.79	CB035	542.56	CB016	147.65	CB035	263.47
CB017	788.12	CB036	477.13	CB017	121.81	CB036	163.42
CB019	1110.09			CB019	140.26		
Range		17.65-1110.09		Range		30.58-310.78	
Mean		483.32		Mean		160.14	
SE		48.06		SE		12.61	
SD		260.99		SD		68.49	
Variance		68117.16		Variance		4691.20	
CV(%)		54.00		CV(%)		12.61	

Table 5. Classification of country bean genotypes based on percent relative shoot dry weight (% RSDW) under 12.0 dsm⁻¹ salinity stresses

(%RSDW)	>80	60-80	40-60	<40
Tolerance level	Tolerant	Moderately tolerant	Moderately susceptible	Susceptible
Genotypes	CB031, CB035, CB003, CB023, CB026, CB028	CB013, CB030, CB014, CB024, CB019, CB020, CB002,	CB032, CB027, CB006, CB017, CB012, CB008, CB011, CB010, CB021, CB016, CB034, CB009, CB022, CB033, CB007, CB036	CB015, CB025, CB004, CB001, CB029, CB005,
No. of genotypes	6	7	16	6

potential in glycophytes subject to saline conditions (Cram, 1976). Despite a significant decrease in net CO₂ assimilation rate, the accumulation of soluble carbohydrates in plants viz. *Prosopis albahas* (Meloni *et al.*, 2004), *Bruguiera parviflora* (Parida *et al.*, 2002), *Lepidium crassifolium* (Murakeozy *et al.*, 2003) were reported when the plants exposed to salinity or drought. Ashraf and Tufail (1995) found that the salt tolerant lines had generally greater soluble sugars than the salt sensitive ones in sunflower. In our experiments, 13 genotypes (CB005, CB006, CB007, CB 009, CB0011, CB012, CB013, CB014, CB015, CB020, CB032, CB035, and CB036) had higher total sugar accumulation than overall mean (160.14) to be considered as salt tolerant. The genotypes CB024, CB026 and CB033 contained relative values below 100 (30.58, 83.47 and 43.46, respectively) indicated they were the most susceptible to salt stress. Parida *et al.* (2002) observed that carbohydrates such as sugars (glucose, fructose, sucrose, fructans) and starch which were accumulated under salt stress played a leading role towards osmo-protection, osmotic adjustment, carbon storage and radical scavenging. A decrease in starch content and

an increase in both reducing and non-reducing sugars and polyphenol levels had also been reported in leaves of *Bruguiera parviflora* (Parida *et al.*, 2002). The amount and contents of soluble sugars and total saccharides were increased significantly, but the starch content was not affected in tomato leaves.

Thermal growth of country bean genotypes was adversely affected in salt stress as compared to control in early growth stage. The studied genotypes were classified into four groups on the basis of their performance in relative dry shoot biomass production according to Ashraf and Waheed (1990) and Mannan *et al.* (2012) (Table 5). According to the scale and score, six genotypes (CB031, CB035, CB003, CB023, CB026 and CB028) were grouped as tolerant (17.1% genotypes), seven genotypes (CB013, CB030, CB014, CB024, CB019, CB020 and CB002,) were as moderately tolerant (20%), 16 genotypes (CB032, CB027, CB006, CB017, CB012, CB008, CB011, CB010, CB021, CB016, CB034, CB009, CB022, CB033, CB007 and CB036) were as moderately susceptible (45.7%) and six genotypes (CB015, CB025, CB004, CB001, CB029 and CB005) were fall into susceptible category (17.1%).

Salinity susceptibility index (SSI) for shoot and root of the accessions are presented in Table 6. Salinity susceptibility index (SSI) for shoot varied from -2.022 (CB031) to 1.602(CB005) with a mean value 0.823 having CV, 96.92% indicated wide variation among 35 country bean genotypes. Genotypes having negative value and less value indicated they were tolerant to salt stress (Mannan *et al.*, 2012). In the study, 12 genotypes (CB031, CB035, CB003, CB023, CB026, CB028, CB013, CB030, CB014, CB024, CB019 and

CB020) had SSI for shoot less than overall mean ($SSI_{shoot}=0.823$) including two contained negative values (CB031 and CB035). About six genotypes showed SSI_{shoot} value around or below 1.0 and they were CB005, CB002, CB032, CB027, CB006 and CB017.

Salinity susceptibility index (SSI) for root varied from -4.845 (CB031) to 3.389 (CB029) with a mean value 0.999 having CV, 167.7% indicated their existed wide variation among 35 country bean genotypes. Genotypes having

Table 6. Salinity Susceptible Index (SSI) for root and shoot of country bean under 12.0 $ds\ m^{-1}$ salinity stress.

Genotypes	Shoot dry weight		Root dry weight				
	SSI	Genotypes	SSI	Genotypes	SSI	Genotypes	SSI
CB001	1.483	CB020	0.775	CB001	2.930	CB020	0.714
CB002	0.845	CB021	1.147	CB002	0.483	CB021	2.515
CB003	0.197	CB022	1.303	CB003	-1.359	CB022	1.624
CB004	1.362	CB023	0.236	CB004	2.219	CB023	-0.093
CB005	1.602	CB024	0.608	CB005	1.449	CB024	0.843
CB006	0.998	CB025	1.348	CB006	2.525	CB025	1.545
CB007	1.317	CB026	0.449	CB007	1.911	CB026	-1.102
CB008	1.082	CB027	0.991	CB008	1.932	CB027	1.642
CB009	1.266	CB028	0.451	CB009	0.881	CB028	0.368
CB010	1.114	CB029	1.578	CB010	0.641	CB029	3.389
CB011	1.088	CB030	0.515	CB011	1.889	CB030	-0.833
CB012	1.009	CB031	-2.022	CB012	1.487	CB031	-4.845
CB013	0.494	CB032	0.977	CB013	-0.362	CB032	1.511
CB014	0.589	CB033	1.305	CB014	0.431	CB033	1.571
CB015	1.340	CB034	1.257	CB015	3.351	CB034	1.768
CB016	1.247	CB035	-0.157	CB016	1.943	CB035	-2.435
CB017	1.002	CB036	1.320	CB017	1.101	CB036	0.366
CB019	0.743			CB019	2.597		
Range		-2.022-1.602		Range		-4.845 -3.389	
Mean		0.823		Mean		0.990	
SE		0.1347		SE		0.281	
SD		0.7973		SD		1.660	
Variance		0.6357		Variance		2.756	
CV (%)		96.917		CV(%)		167.69	

negative value and less value for SSI_{root} indicated that they were tolerant to salt stress. In the present study, 15 genotypes (CB036, CB028, CB014, CB002, CB010, CB020, CB024, CB009) had SSI_{root} for root less than overall mean ($SSI_{\text{root}}=0.999$) including seven contained negative values (CB031, CB035, CB003, CB026, CB030, CB013, CB023). A single genotype (CB017) showed SSI_{root} value around 1.0 (1.10).

SSI_{shoot} and SSI_{root} were highly correlated as similar genotypes responded to salinity with same trend. Such type of screening was carried out and grouping was made at the seedling stages in chickpea (AI-Muttawa, 2003), lentil (Ashraf and Wahid, 1990), mungbean (Aziz, 2003) and Soybean (Mannan *et al.*, 2012). Screening of genotypes for salt stress at early stage is also corroborating faithfully the tolerance level at maturity stage too as depicted by Mannan *et al.* (2012) and has a positive relationship (Ashraf and Waheed, 1990). However, Karim *et al.* (2012) suggested that salt tolerance may be measured in terms of absolute growth at a given salt concentration, or in relative terms depending upon the growth potential of a particular genotype under non saline conditions. Not only the growth stages, a plant responds to salt tolerance is species and genotypes dependent and depends on many factors/ processes viz. the length and severity of the salinity, the age and stage of development, the organ and the cell type and the sub-cellular compartment. An example of avoidance at the cellular level is the process of osmotic adjustment, where the osmotic potential of the cell is lowered in order to favour water uptake and maintenance of turgor (Bray, 1997).

Conclusion

Considering morphological (plant growth, leaf discoloration and yellowing), physiological (relative root and shoot growth, relative root and shoot dry weight, salinity susceptibility index for root and shoot), and biochemical parameters (proline content, total sugar and SPAD value) related to salinity stress, the genotypes CB031, CB035, CB003, CB023, CB026, CB028, CB013, CB030, CB014, CB024, CB019, CB020 and CB002 were selected for further study and utilize in improvement of country bean onward.

Acknowledgements

The present project work is financed by the Research Management Wing of Bangabandhu Sheikh Mujibur Rahman Agricultural University. The authors would like to acknowledge their gratitude towards university authority for financial support. The authors also express their gratitude to BAS-USDA-PALS authority for their financial assistance for collecting experimental materials from coastal regions of Bangladesh during 2010-2012.

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