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AN INTEGRATED APPROACH FOR SCREENING OF CHICKPEA GENOTYPES FOR SALINITY TOLERANCE

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SUMMARY

An experiment was conducted with eight genotypes of chickpea to provide useful parameters for screening against salinity stress. Plants were raised under 0, 4, 6 and 8dSm^{-1} of chloride predominant salinity. Salinity affected membrane injury, chlorophyll stability index (CSI) and K/Na ratio of fully expanded leaves of 8 week old plants and yield adversely and the extent of reduction was found to vary with the genotype. Computation of the results on the basis of double ranking system i.e 'absolute value rank' and 'per cent change rank' revealed the tolerance of chickpea genotypes in the decreasing order; KC-1>HC-3>C235>IPC94-94>Pusa256>HC-1>CSG9505>HC-5. Spearman's rank correlation (rs) and coefficient of concordance of these parameters with yield (w) has revealed positive and significant results on the basis of 'per cent change rank' than 'absolute value rank'.

***Key words*: Chickpea, chlorophyll stability index, K/Na ratio, membrane injury, salinity tolerance**

INTRODUCTION

Chickpea is the third most important pulse crop in the world after dry beans and dry peas. Chickpea seeds are rich source of proteins that ranges between 12.6 and 30.5% (Singh *et al*., 1997). India is the leading country with the highest area among the 34 chickpea growing countries under diverse geographical conditions (Ali and Kumar 2000). It occupies about 7.54 million ha area with an average productivity of 812 kg ha^{-1} amounting to 6.13 million tonnes of grain production annually (GOI, 2000). In Haryana, chickpea occupies first position in terms of acreage and production and is grown in an area of 1.0 x 10^5ha with total production 5.8 x 10^4 tonnes (Anonymous 2000).

Soil salinity is one of the major environmental constraint in agriculture crop production and legumes are by and large sensitive to salinity. With increasing irrigation facilities and poor water management practices the problem of salinity is intensifying with time. One of the options for the optimal utilization of these soils is to have plant species that tolerate high salt levels. As improvement of salt tolerance of legumes assumes significance, it has become imperative to work out for an integrated approach for screening of crop genotypes with emphasis on parameters like membrane injury, chlorophyll stability index and K^+/Na^+ ratio which may serve as an index of anticipated yield and present study is an integrated approach in this direction.

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MATERIALS AND METHODS

Plants of eight genotypes of chickpea (*Cicer arietinum* L.) namely CSG 8962 (KC-1), HC-3, C 235, Pusa 256, IPC 94-94, HC-1, CSG 9505 and HC-5 differing in their salinity tolerance were raised under sand culture conditions using the seeds inoculated with *Rhizobium leguminosarum* strain Ca 181. The plants were raised in polythene bags filled with 8 kg of acid treated and thoroughly washed river sand in a screen house under naturally lit conditions. Each bag was having a drainage hole at one of the corners of sealed end. The cut portion was plugged with a wad of glass wool from inner side to allow free drainage of solution from the bag without any outflow of sand and to avoid water logging. Plants were nourished by nitrogen free nutrient solution (Wilson and Reisenauer 1963) at 10 days interval with the following compositions, except the starter dose of NO_3-N(45 mg/pot).Chloride salinity(control, 4.0, 6.0 and 8.0 dS m^{-1}) was created by adding appropriate amounts of different salts viz., NaCl, $CaCl_2$, $MgCl_2$ and $MgSO_4$ to N-free nutrient solution. The salts were used in such a way that Na^+: Ca^{+2}+ Mg^{+2}; Ca^{+2}: Mg^{+2} and Cl^-: SO_4^{-2} ratios were 1:1; 1: 3 and 7:3, respectively on milliequivalent basis. Salinity treatment was administered at sowing time and maintained throughout the course of crop growth. Whenever needed, the pots were irrigated to field capacity with tap water. For each treatment, fifteen pots were used.

Young fully expanded leaves (3rd from the top) from 8 week old plants were used for the assessment of membrane stability in terms of electrolyte leakage (Sullivan 1972). For chlorophyll stability index (CSI) chlorophyll from the leaves was extracted using dimethyl sulphoxide (DMSO) (Hiscox and Israelstam 1979) and calculated by the method as described by Chhabra *et al*.(1980). Sodium and potassium were quantified from young fully expanded leaves by flame photometer (Elico, India). At maturity, number and weight of the seeds per plant were also recorded.

Based on these parameters, chickpea genotypes were ranked on the basis of absolute value (absolute rank) and magnitude of per cent change at 8dS m^{-1} over the respective controls. Rank correlation and coefficient of concordance was worked out (Raghav Rao 1983).

RESULTS AND DISCUSSION

Membrane injury index (MII): Electrolyte leakage was more in leaves as compared to roots and it increased significantly with increasing level of salinity (Table 1). The genotype HC-5 showed maximum increase, over control followed by HC-1 and Pusa 256 while minimum increase in membrane injury was evident in KC-1 followed by HC-3. Dionisio-Sese and Tobita (1998) reported that increasing magnitude of salinity stress induced more electrolyte leakage from leaves of sensitive varieties of rice when compared to tolerant ones. A similar increment in electrolyte leakage from roots as well as shoot of green gram raised under saline conditions has been reported (Panda 2001).

Chlorophyll stability index (CSI): A significant increase in CSI of the leaf was observed at all levels of salinity. Among tested genotypes, KC-1 and HC-3 showed the lowest i.e., 104.56% and 108.80% increase in CSI respectively, at 8.0 dS m^{-1} salinity indicating their tolerance to salinity. On the other hand, increase in CSI was maximum in HC-5 (164.43%) and HC-1 (153.76%) revealing their sensitivity to salinity (Table 1). Kumar *et al.* (2003) however, reported that CSI decreased with increasing concentration of NaCl in both sensitive and tolerant mulberry cultivars and higher CSI was found in tolerant as compared to sensitive cultivars.

Mineral content: Sodium content of the leaf increased significantly with the progressive increase in the level of salinity; increase being minimum in KC-1 (37.18%) followed by HC-3 and maximum in HC-5 (68.22%) followed by HC-1 and C 235.

Under non-saline conditions, K^+ content of the leaf was highest in Pusa 256 and lowest in C 235. Lower salinity did not affect K^+ content in HC-3, C 235, IPC 94-94 and HC-1 but increased in other genotypes. Higher levels of salinity increased K^+ content in all genotypes except CSG 9505 and HC-5 where a decline was evident (Table 1). At 8.0 dSm^{-1}

Table 1. Effect of salinity on membrane injury (%) , chlorophyll stability index and Na/K ratio in the leaf of different chickpea genotypes

Genotype	Control	Salinity level (dSm^{-1}) 4.0	6.0	8.0	Mean
Membrane injury (%) and chlorophyll stability index (in parenthesis)					
KC-1	262.83 (92.87)	323.59(117.68)	392.01(160.18)	470.02(189.98)	362.11(140.17)
HC-3	198.58(105.55)	221.07(140.34)	300.95(183.61)	365.55(220.30)	271.53(162.45)
C 235	353.20(101.21)	452.94(153.49)	543.31(186.09)	707.88(216.68)	514.33(164.36)
Pusa 256	261.86(103.06)	312.68(143.97)	421.98(202.12)	537.00(245.17)	383.38(173.58)
IPC 94-94	301.45(100.58)	371.93(142.14)	467.13(186.03)	607.88(239.04)	437.09(166.94)
HC-1	337.28(91.51)	433.63(140.01)	571.13(183.23)	794.00(232.22)	534.01(161.74)
CSG 9505	284.71(113.85)	343.02(165.13)	452.44(217.60)	571.40(277.10)	412.89(193.42)
HC-5	365.19(116.25)	469.11(178.75)	605.58(242.99)	911.26(307.40)	587.78(211.34)
Mean	295.63(103.11)	365.99(147.69)	469.31(195.23)	620.62(240.98)	
CD at 5%	Genotype(G) = 13.26 (5.08)	Salinity(S) = 9.37 (3.59)	G x S = 26.52 (10.17)		
K^+/Na^+ ratio					
KC-1	92.87	117.68	160.18	189.98	140.17
HC-3	105.55	140.34	183.61	220.30	162.45
C 235	101.21	153.49	186.09	216.68	164.36
Pusa 256	103.06	143.97	202.12	245.17	173.58
IPC 94-94	100.58	142.14	186.03	239.04	166.94
HC-1	91.51	140.01	183.23	232.22	161.74
CSG 9505	113.85	165.13	217.60	277.10	193.42
HC-5	116.25	178.75	242.99	307.40	211.34
Mean	103.11	147.69	195.23	240.98	
CD at 5%	Genotype(G) = 5.08	Salinity(S) = 3.59	G x S = 10.17		

maximum increase in K^+ content was noticed in KC-1 (33.19%) followed by IPC 94-94 and HC-3.

Although contents of Na^+ and K^+ increased with progressive increase of salinity, K^+/Na^+ ratio decreased with increasing level of salinity in all genotypes except KC-1, where no change was evident at 4.0 dS m^{-1} salinity. At highest level of salinity decline in K^+/Na^+ ratio was minimum in KC-1 followed by HC-3, IPC 94-94 and Pusa 256 and maximum in genotype HC-5 (39.75%) followed by CSG 9505, C 235 and HC-1 (Table 1). Salinity mediated decline in K^+/Na^+ ratio is widely documented (Dhingra *et al*. 1994, Oczan *et al*. 2000) which inactivates enzymes and inhibits protein synthesis. Moreover at high concentrations, Na^+ can replace Ca^{+2} from plasmamembrane as has been reported for cotton roots (Cramer *et al*. 1985), resulting in a change in membrane permeability. Based on K^+/Na^+ ratio, genotypes KC-1 and HC-3 are designated as salt tolerant while genotypes HC-5 and HC-1 as relatively salt sensitive.

Yield parameters: Low level of salinity did not affect seed number significantly in KC-1 and IPC 94-94 but decreased it in rest of the genotypes. Number of seeds and their weight decreased with the progressive increase in the level of substrate salinity. At highest

Table 2. Effect of salinity on number of seeds and seed weight (g plant^{-1} in parentheses) in different genotypes of chickpea

Genotype	Control	Salinity level (dSm^{-1}) 4.0	6.0	8.0	Mean
KC-1	15.83 (1.95)	15.16 (1.88)	13.33 (1.61)	12.66 (1.38)	14.24 (1.70)
HC-3	15.33 (3.81)	14.00 (3.47)	13.00 (3.00)	11.50 (2.41)	13.45 (3.17)
C 235	16.16 (1.97)	14.50 (1.52)	12.66 (1.32)	11.16 (0.97)	13.62 (1.44)
Pusa 256	15.83 (3.43)	13.83 (2.76)	11.83 (2.00)	8.66 (1.07)	12.53 (2.31)
IPC 94-94	15.50 (2.81)	14.83 (2.44)	11.66 (1.68)	9.00 (1.06)	12.74 (1.99)
HC-1	15.16 (2.10)	12.33 (1.43)	10.00 (0.92)	7.83 (0.64)	11.33 (1.27)
CSG 9505	16.33 (2.29)	9.33 (1.08)	7.16 (0.69)	6.00 (0.49)	9.70 (1.13)
HC-5	16.00 (2.60)	12.73 (1.90)	9.96 (1.20)	7.66 (0.75)	11.59 (1.61)
Mean	15.77 (2.62)	13.34 (2.06)	11.20 (1.55)	9.31 (1.10)	
CD at 5%	Genotype(G) = 0.483 (0.035)	Salinity(S) = 0.342 (0.025)	G x S=0.967 (0.071)		

level of salinity (8.0 dS m^{-1}) maximum reduction in seed number was evident in CSG 9505 (63.25%) followed by HC-5, HC-1 and Pusa 256 and minimum in KC-1 (20.02%) followed by HC-3. The minimum reduction in seed weight per plant was evident in KC-1 (29.33%) followed by HC-3 (36.74%), C 235 (50.76%), while genotype CSG 9505 (78.6%) showed maximum reduction followed by HC-5 (71.15%), HC-1 (69.52%) and Pusa 256 (68.22%, Table 2). Reduction in seed yield in terms of their number and weight may be ascribed to failure of fertilization or to sibling rivalry for photoassimilates.

Ranking of chickpea genotypes: Ranking of chickpea genotypes for their relative tolerance to salinity on the basis of parameters studied by following 'dual ranking system' on the basis of absolute value (absolute value rank) as well as per cent change (per cent change rank) at highest salinity level (8.0 dS m^{-1}) has evinced greater salinity tolerance of KC-1 while HC-5 as the most sensitive genotype. Based on this ranking salinity tolerance of chickpea genotypes decreased in the following order : KC-1 > HC-3 > C 235 > IPC 94-94 > Pusa 256 > HC-1 > CSG 9505 > HC-5. Subbarao *et al.* (1990) catergorized ICPL 227 genotype of pigeonpea as most tolerant and Hy3C as most sensitive to salinity by 'dual ranking system'. However these workers did not work out Spearman's coefficient of rank correlation and coefficient of concordance for different characters with yield. Computation of coefficient of concordance revealed that CSI, MII and K^+/Na^+ ratio of the young fully expanded leaf can be employed to evaluate salinity tolerance status of chickpea genotypes without waiting for the final yield.

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