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Effects of salt stress on cucumber: Seed germination, vegetative growth and fruit yield

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ABSTRACT

Seventeen cucumber genotypes were subject to four levels of salt stress (control, 2 and 4 dSm^{-1}) in polythene sheet lined earthen pots using completely randomized design with three replications to assess the effects of salt stress on germination, vegetative growth and fruit yield. Combination of NaCl, Na_2CO_3 and K_2SO_4 salts were used to induce salt stress artificially. ANOVA revealed significant differences amongst genotypes and genotype × salt stress level interaction for all the traits indicating differential response of the genotypes. Increased levels of salt concentration resulted in progressive reduction in germination (78.47, 43.67 and 24.90%), number of leaves (19.26, 10.56 and 6.30), survivability (93.77, 59.79 and 39.25%), vine length (88.99, 49.07 and 28.92 cm) and fruit yield per vine (1.10, 0.62 and 0.29 kg); while increased the affected leaves (28.73, 68.92 and 82.59%) and defoliation (13.39, 59.74 and 74.83%), respectively in control, 2 and 4 dSm^{-1}. Thus, these above phenotypic traits appeared to be promising as selection criteria for salt tolerance at morphological level. As a result, genotypes CRC-8 and CHC-2 observed salt tolerant, whereas DC-1, CH-20 and Pusa Uday were salt sensitive.

Key words: Cucumber, fruit yield, salt stress, seed germination, vegetative growth.

INTRODUCTION

Salt stress is one of the major factors reducing plant growth and productivity worldwide and affects about 7% of the world's total land area (Flowers *et al.*, 5). Addition of salts into water lowers its osmotic potential, resulting in decreased availability of water to roots and thus exposes plants to secondary osmotic stress. Adverse effects of salinity on plant growth may be due to ion cytotoxicity (mainly due to Na^+, Cl^-, SO_4^{2-}) and osmotic stress (Zhu, 16). At higher salt levels, the crop yields are reduced so drastically that crop cultivation is not economical without soil amendments. One of the most effective approaches to overcome salt stress problems is to identify and cultivate salt tolerant varieties in such soils to bring about soil reclamation. In irrigated agriculture, improved salt tolerance of crops can lessen the leaching requirement, and so lessen the costs of irrigation. Examining plant growth during growing season provides information about crop's salt tolerance over different stages of growth. Hence, evaluation at different growth stages assumes importance. Extensive research has been done on effects of salt stress on cereals, leguminous crops and some field grown vegetable crops. Though, intervarietal differences are pronounced with respect to salt tolerance in *Cucumis* (Botia *et al.*, 1). However, systematic studies on consequences of salt stress on vegetative growth of cucumber are limited.

The understanding on effects of salt stress and mechanism of tolerance are highly essential to breed for salt tolerant cucumber. The best criterion to breed for salt tolerance is to select for higher yield. Compared to conventional techniques that score and rank salt tolerant genotypes based on single parameter, some success has already been realized in rice by simultaneous use of multiple agronomic parameters and at different growth stages (Shannon, 10). However, this approach is long drawn and requires lot of time and labour. Hence, a reliable and quick method of screening would be necessary for the rapid progress in breeding for salt stress tolerance. Identifying selection criteria during early growth and vegetative stages is an alternative to reduce time required for screening large number of germplasm. Hence, in the present study, the consequences of salt stress on germination and vegetative growth in cucumber were investigated to identify the trait/ criteria important for salt tolerance during early growth and vegetative stages of cucumber.

MATERIALS AND METHODS

The present investigation was carried out at Division of Vegetable Science, IARI, New Delhi. Seventeen cucumber genotypes, namely, CHC-1 and CH-20 (HARP, Ranchi); CRC-8 (Saharanpur); Himangi (Pune); CHC-2 (HARP, Ranchi); G-319, G-338, Long Green, Poinsette (Delhi); ACC-9 (Raipur); CRC-5 (Panipat); WBC-27 (Hoogly); WBC-31 (South 24 Parganas); Pusa Uday (Rai Barelly/ Delhi); DC-3

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(Moradabad); DC-1 (Muzaffarpur) and WBC-28 (Hoogly), previously collected from different parts of India, were taken for study.

Cucumber seeds were sown in earthen pots (~30 cm dia) size lined with polyethylene sheet and each pot filled with 5 kg soil. Plants were exposed to three levels of salt stress, *viz*., 0 (control), 2, 4 and 6 dS m^{-1} which was developed by combination of NaCl, Na_2CO_3 and K_2SO_4 salts (Table 1) following the procedures described by Dubey *et al.* (4). Experiment was laid out in a completely randomized design (CRD) with three replications and five pots per replication per treatment were maintained. In a pot, ten seeds of a cucumber genotype were sown one week after the salt treatment. After germination, one plant was retained in each pot. The EC of the soil in pots was tested at regular intervals and final salt stress level was recorded to be 0.43, 1.71 and 3.82 dS m^{-1} in pots with control, 2 and 4 dS m^{-1} salt stress treatments, respectively at the end of the experiment (60 DAS) when majority of the genotypes started showing wilt symptoms.

The first observation of germination was recorded after 14 days of sowing. The other observations such as number of leaves, percentage of affected leaves, defoliation percentage and survival percentage were recorded at 60 days after sowing. The percentage of affected leaves was calculated by recording the number of leaves showing burning symptoms on their tips. Defoliation percentage was calculated by recording number of defoliated leaves out of total leaves. Survival percentage was recorded by counting the total number of survived plants out of total germination, and vine length of survived plants was measured in cm. Fruit yield per vine was taken by averaging the total weight of fruits from all the pickings from the surviving vines in each treatment. In order to allow comparisons among genotypes, scoring and ranking on a 1-9 scale procedures was followed as described by Zeng *et al.* (15). Accordingly, a salt sensitive genotype DC-1 was chosen susceptible check based on morphological traits and salt tolerance index was estimated and score were given to the genotypes.

Table 1. The quantity of different salts (g/ 5 kg soil) required for the development of desired level of salt stress (dS m^{-1}).

EC level	NaCl	Na_2CO_3	K_2SO_4
0	0.00	0.00	0.00
2	8.45	3.25	5.66
4	16.25	6.63	10.88
6	24.90	9.70	15.99

$$\text{Salt tolerance index} = \frac{\text{Mean of a genotype for a trait over salt stress treatments}}{\text{Mean of susceptible check for the trait over salt stress treatments}}$$

Data analysis for factorial CRD was carried out using Windostat software (Ameerpet, Hyderabad). Levene's Test for homogeneity of variance and Kolmogorov Smirnov's Test for normalcy of percentage data was checked after square root transformation using the software and CD values, were estimated for comparing genotypes (Gomez and Gomez, 6).

RESULTS AND DISCUSSION

In the present investigation of salt stress, observations were recorded up to 4 dS m^{-1}, since none of the genotypes survived beyond at 6 dS m^{-1}. Analysis of variance of the normally distributed data for the germination percentage, parameters of vegetative growth and fruit yield per vine revealed significant differences among genotypes and genotype × salt stress level interaction indicating the existence of considerable genetic variability among the genotypes.

The salt stress had adverse effect on seed germination of all cucumber genotypes and therefore differential response was observed (Tables 2 & 3). Five genotypes (DC-1, ACC-9, Pusa Uday, CRC-5 and CH-20) observed with less than 15% germination at 4 dS m^{-1} of which DC-1 recorded the lowest (9.67%). The average germination was recorded highest in the genotype CRC-8 (56.70%) followed by CHC-2 (53.73%) and G-338 (48.15%), which were significantly superior in comparison to other genotypes (Table 2). The genotypes CRC-8, CHC-2 and G-338 recorded high salt tolerance index (3.14, 2.98 and 2.67, respectively) and score (1, 2 and 3, respectively) (Table 3). The lowest salt tolerance index was recorded in DC-1, ACC-9, Pusa Uday, CRC-5 and WBC-28 (score 9), as the average germination in these genotypes was either less than or close to 20%. The reduction in seed germination at higher salt concentration may be associated with osmotic effect (reduced rate of imbibition), specific ion effect or altered enzyme activities are in agreement with previous results on melon (Torres and Marcos, 12), and *Cucurbita* sp. (Wang *et al*., 14).

The increasing levels of salt concentration from control to 2 to 4 dS m^{-1} resulted in progressive reduction in vegetative growth of cucumber (Tables 2 & 3). The highest mean number of leaves was recorded in CHC-2 followed by CRC-8 and also observed highest salt tolerance index and the highest salt tolerance score. The lower mean number of leaves was recorded in DC-1 and CH-20 and had lower salt tolerance index and score. Higher salt stress (4 dS m^{-1}) had injurious effect on leaves, which resulted in very high percentage of affected leaves (82.59)

Table 2. Effect of different salinity levels on vegetative growth characters in cucumber genotypes.

Genotype	Germination (%)				No. of leaves				Affected leaves (%)				Defoliation (%)			
	Salinity (dS m^{-1})			Mean	Salinity (dS m^{-1})			Mean	Salinity (dS m^{-1})			Mean	Salinity (dS m^{-1})			Mean
	Control	2	4		Control	2	4		Control	2	4		Control	2	4	
CHC-1	79.20	40.30	21.53	30.92	20.15	11.22	8.15	9.69	18.63	65.27	75.37	70.32	12.17	58.27	62.20	60.24
CRC-8	81.40	67.23	46.16	56.70	21.15	13.48	8.23	10.86	18.50	51.27	67.67	59.47	11.13	44.87	61.40	53.14
Himangi	75.27	47.27	25.37	36.32	18.72	11.18	5.63	8.40	20.43	63.48	78.43	70.96	13.10	39.00	79.57	59.29
CHC-2	76.10	64.33	43.12	53.73	19.67	15.95	8.36	12.16	21.50	45.87	71.50	58.69	16.67	39.77	67.00	53.39
G-319	78.00	47.23	28.43	37.83	17.73	9.71	5.46	7.59	18.70	68.17	88.83	78.50	11.33	59.43	71.53	65.48
G-338	79.50	56.13	40.17	48.15	19.85	10.66	6.63	8.65	18.43	64.63	73.20	68.92	11.57	55.57	67.10	61.34
Long Green	78.03	47.17	35.17	41.17	19.37	12.26	7.37	9.82	21.40	72.50	81.63	77.07	14.33	61.50	70.87	66.19
ACC-9	78.00	26.37	10.73	18.55	21.41	12.65	5.55	9.10	23.47	74.67	83.57	79.12	15.03	62.33	78.10	70.22
Poinsett	80.93	50.30	25.48	37.89	19.85	11.44	5.59	8.52	19.41	62.37	78.60	70.49	12.40	52.30	68.87	60.59
CRC-5	78.43	30.10	12.26	21.18	18.98	8.54	6.33	7.44	21.13	68.53	82.47	75.50	13.30	58.13	72.10	65.12
WBC-27	81.30	47.60	28.10	37.85	17.20	11.31	8.54	9.93	22.20	70.37	81.07	75.72	14.53	61.37	71.50	66.44
CH-20	79.47	41.10	14.20	27.65	18.29	6.98	2.87	4.93	18.23	83.46	95.53	89.50	11.60	71.86	86.80	79.33
WBC-31	77.56	46.53	28.87	37.70	18.29	9.79	7.37	8.58	21.67	68.46	85.17	76.82	13.13	57.40	75.23	66.32
Pusa Uday	75.07	33.63	11.63	22.63	20.41	7.84	5.68	6.76	22.13	78.80	91.47	85.14	12.90	68.20	81.27	74.74
DC-3	79.97	40.27	25.13	32.70	20.80	10.66	7.37	9.02	18.73	74.40	87.87	81.14	11.57	68.27	85.93	77.10
DC-1	76.23	26.43	9.67	18.05	17.20	5.20	2.04	3.62	22.63	81.13	98.17	89.65	16.47	71.10	91.37	81.24
WBC-28	79.53	30.40	15.17	22.79	18.29	10.70	5.98	8.34	25.68	78.50	98.23	88.37	16.50	66.13	88.38	77.26
Mean	78.47	43.67	24.90		19.26	10.56	6.30		28.73	68.92	82.59		13.39	59.74	74.83	
CD (p = 0.05)				5.11				1.41				9.26				9.11

Genotype	Survivability (%)				Vine length (cm)				Fruit yield per vine (kg)				Yield reduction (%) over control		
	Salinity (dS m^{-1})			Mean	Salinity (dS m^{-1})			Mean	Salinity (dS m^{-1})			Mean			
	Control	2	4		Control	2	4		Control	2	4		2 dS m^{-1}	4 dS m^{-1}	Mean
CHC-1	94.50	60.84	41.37	51.11	88.86	57.20	33.46	45.33	1.17	0.64	0.27	0.45	45.60	76.80	61.20
CRC-8	96.70	75.47	56.30	65.89	101.86	72.24	45.14	58.94	1.50	1.08	0.66	0.87	27.87	56.25	42.06
Himangi	90.57	63.27	37.73	50.50	92.34	46.06	28.26	37.16	1.10	0.72	0.34	0.53	34.82	68.85	51.84
CHC-2	91.40	69.37	51.97	60.67	85.00	64.94	40.44	52.69	1.35	0.95	0.55	0.75	29.99	59.00	44.50
G-319	93.30	73.27	40.30	56.79	82.86	53.20	30.94	42.07	1.00	0.65	0.35	0.50	35.00	65.10	50.05
G-338	94.80	68.40	50.67	59.54	90.80	58.40	34.94	46.67	1.20	0.84	0.50	0.67	30.00	58.00	44.00
Long Green	93.33	60.27	40.57	50.42	80.80	51.26	28.86	40.06	1.05	0.64	0.32	0.48	39.24	69.37	54.30

Contd...

Contd...

ACC-9	93.30	54.73	35.13	44.93	83.20	52.46	30.60	41.53	0.90	0.46	0.18	0.32	48.72	80.47	64.59
Poinsett	96.23	66.30	43.13	54.72	84.94	49.46	26.80	38.13	0.85	0.54	0.26	0.40	36.63	69.81	53.22
CRC-5	93.73	45.13	32.00	38.57	84.00	50.80	29.94	40.37	1.04	0.47	0.16	0.32	55.00	84.25	69.63
WBC-27	96.60	60.27	36.30	48.29	94.40	49.14	31.12	40.13	0.95	0.54	0.23	0.39	43.04	75.69	59.37
CH-20	94.77	47.03	31.20	39.12	86.34	26.94	15.34	21.14	1.35	0.47	0.14	0.31	65.00	89.72	77.36
WBC-31	92.86	63.37	36.23	49.80	88.46	42.80	26.00	34.40	0.89	0.55	0.26	0.41	37.65	71.02	54.34
Pusa Uday	90.37	51.47	33.63	42.55	85.46	25.26	12.60	18.93	1.10	0.49	0.16	0.32	55.49	85.60	70.54
DC-3	95.27	55.43	35.67	45.55	88.40	42.00	23.26	32.63	0.83	0.45	0.19	0.32	46.26	77.40	61.83
DC-1	91.53	50.37	30.00	40.19	100.26	29.26	15.40	22.33	1.35	0.47	0.14	0.31	65.00	89.72	77.36
WBC-28	94.83	51.37	35.00	43.19	94.86	62.34	38.54	50.44	1.13	0.54	0.20	0.37	52.65	82.54	67.59
Mean	93.77	59.79	39.25		88.99	49.07	28.92		1.10	0.62	0.29		44.00	74.09	59.05
CD (p = 0.05)				9.12				6.13				0.14			4.23

*Data were analyzed after testing normally distributed and homogeneous using square root transformation in Windostat software. Critical differences at p = 0.05 was presented to categorize genotypes into two broad categories, *i.e.* salt stress tolerant and salt sensitive. Genotypes within the same group (either stress tolerant or salt sensitive) were not compared, since unreliable information. However, genotypes within a group were presented accurate values wise in the results section to conclude into reliable score and index values.

over control (28.73). The lowest mean percentage of affected leaves was observed in CHC-2 followed by CRC-8, while highest was recorded in DC-1 followed by CH-20. Table 3 indicated that the genotypes CHC-2 and CRC-8, had top salt tolerance score of 1, while CH-20 and DC-1 recorded the lowest score of 9. Similarly, defoliation percentage (>60%) was severe at 4 dS m^{-1} in all the genotypes (Table 2). Mean defoliation percentage was highest in DC-1 (81.24) followed by CH-20 (79.33), whereas CHC-2 and CRC-8 had lower (<53) defoliation. The salt stress of 4 dS m^{-1} resulted in very low survivability (39.25%) in comparison to control (93.77%). At 4 dS m^{-1} salt stress level, survivability reduced drastically compared to control conditions especially in DC-1 (30%), CH-20 (31.20%), CRC-5 (32.00%) and Pusa Uday (33.63%). High survivability was recorded in CRC-8 followed by CHC-2 and G-338 both at 4 dS m^{-1} (>50%), which also recorded top score and index (except G-338 had score 3). Vine length decreased as salt stress level was increased. Longest mean vine length was observed in CRC-8 (58.94 cm) followed by CHC-2 (52.69 cm), whereas, shortest mean vine length was recorded in Pusa Uday (18.93 cm) followed by CH-20 (21.14 cm) and DC-1 (22.33 cm). Table 3 showed that maximum index of 2.64 was observed in CRC-8 with a score of 1 followed by CHC-2, whereas, the lowest salt tolerance index (0.85) was observed for Pusa Uday. The lowest salt tolerance score of 9 was observed in Pusa Uday, CH-20 and DC-1.

There was a progressive reduction in fruit yield per vine as salt stress increased in all the genotypes (Table 3). Highest fruit yield per vine was observed in CRC-8 (0.87 kg) followed by CHC-2 (0.75 kg), whereas the lowest in CH-20 and DC-1 (0. 31 kg each). It could be noted that under salt free conditions, the fruit yield of CH-20 and DC-1 (1.35 kg) was statistically on par with highest yielding CRC-8 (1.50 kg). Table 3 showed that maximum index of 2.84 was observed in CRC-8 with a score of 1 followed by CHC-2 (2.45 and score 3), whereas, the lowest index (1.0) was observed for CH-20 and DC-1 whose index was on par with those of Pusa Uday, ACC-9, DC-3, WBC-28 and CRC-5 with a score of 9. Excepting CRC-8, CHC-2 and G-338, other genotypes were susceptible or moderately susceptible (score >6). The average fruit yield reduction under salt stress was 44.0% at 2 dS m^{-1} and 74.09% at 4 dS m^{-1} (Table 2). Among the genotypes minimum yield reduction under salt stress was observed in CRC-8 (42.06%) followed by G-338 (44%) and CHC-2 (44.50%). These genotypes had top scores (1-4) based on the salt tolerance index (Table 3). Further, highest reduction under salt stress

Table 3. Salt tolerance index and score among cucumber genotypes at mean value of salt stress treatments.

Genotype	Germination (%)		No. of leaves/vine		Affected leaves (%)		Defoliation (%)		Survivability (%)		Vine length (cm)		Yield per vine (kg)	
	Index	Score	Index	Score	Index	Score	Index	Score	Index	Score	Index	Score	Index	Score
CHC-1	1.71	7	2.68	3	0.78	4	0.74	3	1.27	6	2.03	4	1.49	7
CRC-8	3.14	1	3.00	2	0.66	1	0.65	1	1.64	1	2.64	1	2.84	1
Himangi	2.01	6	2.32	5	0.79	4	0.73	2	1.26	6	1.66	5	1.73	6
CHC-2	2.98	2	3.36	1	0.65	1	0.66	1	1.51	3	2.36	2	2.45	3
G-319	2.10	5	2.10	5	0.88	6	0.81	5	1.41	4	1.88	4	1.63	7
G-338	2.67	3	2.39	4	0.77	3	0.76	3	1.48	3	2.09	3	2.20	4
Long Green	2.28	5	2.71	3	0.86	6	0.81	5	1.25	6	1.79	5	1.57	7
ACC-9	1.03	9	2.51	4	0.88	6	0.86	6	1.12	7	1.86	5	1.04	9
Poinsett	2.10	5	2.35	4	0.79	4	0.75	3	1.36	4	1.71	5	1.30	8
CRC-5	1.17	9	2.05	6	0.84	5	0.80	4	0.96	9	1.81	5	1.03	9
WBC-27	2.10	5	2.74	3	0.84	5	0.82	5	1.20	6	1.80	5	1.26	8
CH-20	1.53	7	1.36	8	1.00	9	0.98	9	0.97	9	0.95	9	1.00	9
WBC-31	2.09	5	2.37	4	0.86	6	0.82	5	1.24	6	1.54	6	1.33	8
Pusa Uday	1.25	9	1.87	6	0.95	8	0.92	7	1.06	8	0.85	9	1.06	9
DC-3	1.81	6	2.49	4	0.91	7	0.95	8	1.13	7	1.46	6	1.04	9
DC-1	1.00	9	1.00	9	1.00	9	1.00	9	1.00	9	1.00	9	1.00	9
WBC-28	1.26	9	2.30	5	0.99	9	0.95	8	1.07	8	2.26	3	1.20	9
Range	1.00 - 3.14		1.00 - 3.36		0.65 - 1.00		0.65 - 1.00		0.96 - 1.64		0.85 - 2.64		1.00 - 2.84	
CD (p = 0.05)	0.215		0.224		0.035		0.033		0.071		0.182		0.185	
							Score (Range)							
1	3.10 - 3.36		3.13 - 3.40		0.65 - 0.69		0.65 - 0.69		1.59 - 1.67		2.47 - 2.67		2.74 - 2.96	
2	2.84 - 3.10		2.86 - 3.13		0.69 - 0.73		0.69 - 0.73		1.51 - 1.59		2.27 - 2.47		2.52 - 2.74	
3	2.58 - 2.84		2.60 - 2.86		0.73 - 0.77		0.73 - 0.77		1.43 - 1.51		2.06 - 2.27		2.31 - 2.52	
4	2.31 - 2.58		2.33 - 2.60		0.77 - 0.81		0.77 - 0.81		1.35 - 1.43		1.86 - 2.06		2.09 - 2.31	
5	2.05 - 2.31		2.06 - 2.33		0.81 - 0.85		0.81 - 0.84		1.27 - 1.35		1.66 - 1.86		1.87 - 2.09	
6	1.79 - 2.05		1.80 - 2.06		0.85 - 0.89		0.84 - 0.88		1.20 - 1.27		1.46 - 1.66		1.65 - 1.87	
7	1.53 - 1.79		1.53 - 1.80		0.89 - 0.93		0.88 - 0.92		1.12 - 1.20		1.25 - 1.46		1.44 - 1.65	
8	1.26 - 1.53		1.27 - 1.53		0.93 - 0.97		0.92 - 0.96		1.04 - 1.12		1.05 - 1.25		1.22 - 1.44	
9	1.00 - 1.26		1.00 - 1.27		0.97 - 1.01		0.96 - 1.00		0.96 - 1.04		0.85 - 1.05		1.00 - 1.22	

was seen in CH-20 and DC-1 (77.36%) followed by Pusa Uday (70.54%), which also had lowest score of 9 based on the salt tolerance index. Results of the present investigation are in agreement with previous findings on muskmelon (Del-Amor *et al.*, 3), zucchini squash (Rouphael *et al.*, 9), melon (Bustan *et al.*, 2), and cucumber (Trajkava *et al.*, 13). The presence of excess ions in soil environment led to accumulation of ions in plant cells affecting directly the protein hydration. It may be due to the Na^+ and particularly Cl^- which are preferentially accumulated in basal (older) leaves rather than apical (younger) leaves, as a result, leaves with severe chlorosis were shed (Sykes, 11).

Thus, in the present investigation, multiple traits such as germination, defoliation, survival percentages and vine length were idendified promising as selection criteria for salt tolerance at morphological level in cucumber. The genotypes CRC-8 and CHC-2 appeared salt tolerant as high mean values of salt tolerance index and score (1.1 and 1.8) were recorded, which may be included as one of the parents in cucumber breeding programmes for salt tolerance.

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