Correlation Studies on Effect of Salinity Stress on Phenology, Biochemical, and Seed Yield in Chickpea (*Cicerarietinum* L.) Genotypes.

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Abstract:- The present study was carried out with ten chickpea genotypes sown under three salinity concentration (0 dSm^{-1}, 3 dSm^{-1} and 6 dSm^{-1}). Days to 50 per cent flowering, days to physiological maturity, proline content, membrane injury index and total chlorophyll content at 60 days after sowing showed a strong positive association among each other. However, the degree of association decreased as the salinity levels increased from 0 dSm^{-1} to 6 dSm^{-1}. Further, seed yield and yield related traits was strongly associated with salinity concentrations. The results indicated that higher salt concentrations were associated with increase in duration of phenological parameters. The proline content and membrane injury index were significantly increased under salt stress conditions in all genotypes. Higher chlorophyll content and seed yields were recorded under 0dSm^{-1} (C_1) than 3 dSm^{-1} and 6 dSm^{-1} (C_2 and C_3, respectively). Genotype ICCV96029 showed highest membrane injury index (14.51) and lowest seed yield value (3.28 g) at 6 dSm^{-1}. While, the genotypes JG11 recorded highest proline content (30.27 mg g^{-1} fr. wt) at 6 dSm^{-1} and chlorophyll content at 0 dSm^{-1} (2.979 mg g^{-1} fr. wt). This study indicated that developing genetic variability by identifying salt tolerant in diverse chickpea lines is one of the appropriate strategies used to overcome salinity problem in arid and semi-arid areas.

Keywords:- *Salinity, proline, salt tolerance.*

I. INTRODUCTION

Chickpea (*Cicer arietinum* Linnaeus), a member of family Fabaceae, is an ancient self pollinated leguminous crop, diploid annual (2N=16 chromosomes) grown since 7000 BC, in different area of the world (Tekeoglu, *et al*., 2000) but its cultivation is mainly concentrated in semi-arid environments such as South asia, West Asia, North Africa, East Africa, Southern Europe, North and South America and Australia (Arefian, *et al.*, 2014). The species is grouped into *desi* and *kabuli* type: *desi* generally have small, darker coloured seeds, where as Kabuli is usually producing large, cream-coloured ones. India is the largest producer of chickpea accounting for about 68 per cent of the world chickpea production. It is cultivated in an area of 9.6 million hectares (a record in the last 50 year) with a production of 9.37 million tonnes and productivity of 974 kg per ha (Anon, 2016-17).

Soil salinity affects about 80 million ha of arable lands worldwide (Munns and Tester, 2008); 2.95 million hectare in India and 49.2 thousand hectare in Haryana and this area is expanding (Ali, 2009). Soil in which concentration of salts is so high as to adversely affect plant growth and crop productivity are called salt affected soils from agriculture point of view. Salt-affected soils is a broader term which includes both saline soils (soils having chlorides and sulphates of sodium) and alkaline soils (soils having carbonates and bicarbonates of sodium). Saline soil is formed when chlorides and sulphates of sodium, calcium, magnesium and potassium are abundant in the soil and the process is known as salinization. Salinity causes not only physiological dehydration (water stress) in plants, but also nutrient ion imbalance (Toker, *et al.*, 2007). Salinity stress adversely affects several morphological features and physiological processes like reduction in growth, decrease in chlorophyll, ion balance, water status, photosynthesis, increase in hydrogen peroxide, which causes lipid per oxidation and consequently membrane injury, nodulation and N_2 fixation (Zhu, 2001). When plants are subjected to salinity, reactive oxygen species (ROS) are also generated in response to stress conditions which cause chlorophyll degradation; lipid peroxidation and electrolyte leakage are considered to be indicators of oxidative damage.

II. MATERIALS AND METHODS

The experiment was conducted in pot-culture conditions with the same ten chickpea genotypes

A. Sowing and salinity treatments

Earthen pots of uniform size (30x30 cm) were filled with 10 kg of air-dried soil and farmyard manure in 6:1 ratio. Each pot received a fertilizer dose of 120, 60 and 60 kg/ha of N, P and K, respectively. Before sowing pots were irrigated with 2.5 liters of water (control) or salt solutions of different concentrations. The plants were subjected to three conditions *viz.* control (C1) and three salinity treatments (C2 and C3). Salt solutions were prepared by using Nacl salt. The salt concentrations of different solutions are given below.

C1= 5 gram of NaCl salt dissolved in 1 liter of water for preparing 3 EC

C2= 10 gram of NaCl salt dissolved in 1 liter of water for preparing 6 EC

Actual salinity values are expressed as EC determined at 3 stages and mean of these was taken as salinity at C1, C2 and C3 levels.

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B. Soil electrical conductivity estimation

Twenty gram of air dried soil was mixed with 50 ml of distilled water in a beaker, stirred well and then allowed to settle down for 2 hours. Electrical conductivity values were recorded with Conductivity Bridge.

Treatment	EC (dSm^{-1})
C1	1.4
C2	3.2
C3	6.3

Plants were watered as and when required. Pots were hand weeded from time to time

III. RESULTS AND DISCUSSION

Data on days to 50 per cent flowering and physiological maturity differed significantly with respect to genotypes, salinity levels and their interactions. Among the salinity levels, significantly higher days to 50 per cent flowering and physiological maturity was recorded 6dSm^{-1} (57.77 and 86.37days, respectively). However, least days to 50 per cent flowering and physiological maturity was recorded in control (42.27 and 77.73 days, respectively). Kotula et al., (2015) reported that at 50mM NaCl concentration days to first flowering and pod initiation were significantly increased and pod size and number of pods per plant were significantly decreased under salt stressed condition in salt susceptible genotypes. Among the genotypes, significantly higher membrane injury index was recorded in genotype ICCV96029 (14.51 %) followed by JAKI9218 (13.78 %) which was on par with NBeG47 (13.47%) at 60 days after sowing. Further, genotype MNK 1 and ICC5003 recorded significantly lower value of membrane injury index, (11.66% and 11.72%, respectively). Membrane injury index estimation gives an idea about the sensitivity of cell membrane under given set of stress conditions as it is related to leaf tissues membrane stability. Borzouei et al., (2012) reported that that increased in cell membrane injury and decreased membrane stability index under salt stress showed the extent of lipid peroxidation caused by relative oxygen species.

Proline content differed significantly with genotype, salinity levels and their interactions. Significantly lower proline content was observed in ICCV96029 and NBeG 47 (16.62 and 17.85 mg g^{-1} fresh weight, respectively) followed by GBM2 (17.94 mg g^{-1} fresh weight,) at 30 days after sowing. However, the genotypes ICC1431, ICC5003, Annigeri 1, MNK1 and GBM2 (23.99, 23.10, 23.08, 23.06 and 22.67 mg g^{-1} fresh weight, respectively) were found on par with each other at 60 days after sowing. The increasing salt concentration resulted in higher level of proline content, protein and malondialdehyde content in chickpea genotypes and Proline accumulation increased upon salt stress in tolerant chickpea genotypes and proline accumulation was more delayed in susceptible genotypes during salinity (Arefian *et al*., 2014). Among the interaction effect the genotype JG 11 recorded significantly higher total chlorophyll at 0 dSm^{-1} (2.979 mg g^{-1}fr. wt.) and the genotype ICCV96029 (1.326 mg g^{-1}fr. wt.) recorded significantly lower total chlorophyll content at 6 dSm^{-1} during 60 days after sowing. Similar findings were noticed by Taibi et al., (2016) reported that the lipid peroxidation of chloroplast during salt stress decrease the chlorophyll pigments and in all genotypes the increased salinity levels reduced the dry mass and chlorophyll pigments and increased malondialdehyde content.

The table on seed yield per plant showed a decreasing trend with respect to salinity levels. Generally, 0dSm^{-1} (control) was favoured by higher yield compared to 6dSm^{-1}. Under 0dSm^{-1} significantly higher seed yield was recorded (7.18 g $plant^{-1}$) followed by 3dSm^{-1} (4.37g $plant^{-1}$). 6dSm^{-1} recorded significantly least seed yield (2.95 g $plant^{-1}$). Salt stress typically impact the various physiological process (low water uptake, decreased respiration, decreased photosynthesis) and ultimately yield of crop (Hussain *et al*., 2016). Nevertheless, significant differences among the chickpea genotypes were observed. The genotype JG11 (12.33) recorded significantly higher number of pods followed by BGD-103 and MNK1 (11.22 and 10.78, respectively). Genotype JG11 recorded significantly higher harvest index (59.15 %) under 0dSm^{-1}, which was found on par with the genotype BGD103 and MNK1 (59.10 and 59.02 %, respectively) under same salinity level. Significantly lower harvest index was observed in genotype ICC96029 (11.56%) under 6dSm^{-1} followed by NBeG47 (20.82 %) under same salt concentration. Reduction in yield and yield related traits under elevated salt concentration may be the result of various factors acting simultaneously like the reduction in the photosynthesis and the subsequent decline in leaf area and stomatal conductance, which would result in the reduction of accumulated biomass (Rasool et al ., 2012 ;Samineni et al., 2011; Grewal, 2010; Singla and Garg, 2004).

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	Days to 50% flowering				Days to Physiological mturity			
Genotypes	**C1**	**C2**	**C3**	**Mean**	**C1**	**C2**	**C3**	**Mean**
Annigeri 1	40.00	49.67	57.67	49.11	75.33	78.33	83.33	79.00
JAKI 9218	46.67	50.00	58.33	51.67	81.67	84.67	88.00	84.78
T3 BGD 103	46.00	47.00	53.67	48.89	80.33	84.33	87.67	84.11
MNK 1	40.67	48.00	56.00	48.22	78.67	83.00	85.33	82.33
JG11	45.67	51.67	56.67	51.33	77.00	82.67	87.00	82.22
GBM 2	43.67	51.00	58.67	51.11	75.67	82.00	84.33	80.67
NBeG 47	41.33	51.33	59.67	50.78	78.00	83.33	86.67	82.67
ICC 1431	41.67	52.33	57.00	50.33	81.00	84.67	89.33	85.00
ICC 5003	43.33	52.00	57.33	50.89	79.00	82.67	87.00	82.89
ICCV 96029	33.67	46.33	62.67	47.56	70.67	77.67	85.00	77.78
Mean	42.27	49.93	57.77		77.73	82.33	86.37	
	SEm±		**LSD @5%**		**SEm±**		**LSD @5%**	
TRT	0.31817		0.84643		0.45782		1.21794	
CONC	0.09545		0.25393		0.13735		0.36538	
T*C	0.95452		2.5393		1.37347		3.65382	

Table 1: Effect of salinity stress on days to 50% flowering and days to physiological maturity in chickpea genotypes

	Membrane injury index (%)				Proline content (mg g^{-1} fr. wt.)				Total chlorophyll(mg g^{-1}fr.wt.)			
ddd **Genotypes**	**C1**	**C2**	**C3**	**Mean**	**C1**	**C2**	**C3**	**Mean**	**C1**	**C2**	**C3**	**Mean**
Annigeri 1	10.44	13.75	15.80	13.33	19.82	22.31	27.10	23.08	2.617	2.074	1.623	2.105
JAKI 9218	10.75	14.12	16.45	13.78	22.33	25.17	30.01	25.84	2.502	2.036	1.571	2.036
BGD 103	9.43	13.19	15.10	12.57	19.64	24.45	29.25	24.45	2.821	2.248	1.847	2.305
MNK 1	9.38	11.74	13.85	11.66	19.65	23.15	26.38	23.06	2.861	2.222	1.791	2.291
JG11	9.54	13.09	14.94	12.52	21.22	25.67	30.27	25.72	2.979	2.304	1.904	2.396
GBM 2	11.28	12.90	14.92	13.03	19.83	22.49	25.69	22.67	2.426	1.985	1.523	1.978
NBeG 47	12.03	13.17	15.22	13.47	18.87	21.87	24.70	21.81	2.246	1.783	1.364	1.798
ICC 1431	10.62	11.92	13.89	12.14	21.32	23.42	27.24	23.99	2.709	2.169	1.745	2.208
ICC 5003	10.47	11.35	13.33	11.72	19.43	24.01	25.86	23.10	2.591	2.127	1.651	2.123
ICCV 96029	11.72	14.85	16.95	14.51	17.89	20.45	23.68	20.67	2.343	1.742	1.326	1.804
Mean	10.57	13.01	15.04		20.00	23.30	27.02		2.610	2.069	1.634	
	SEm±		**LSD @5%**		**SEm±**		**LSD @5%**		**SEm±**		**LSD @5%**	
TRT	0.01966		0.0523		0.19377		0.51549		0.19896		0.5293	
CONC	0.0059		0.01569		0.05813		0.15465		0.05969		0.15879	
T*C	0.05898		0.1569		0.58132		1.54648		0.59689		1.58789	

Table 2: Effect of salinity stress on membrane injury index, proline content and chlorophyll content in chickpea genotypes.

	Pod number				Seed yield (g/plant)				Harvest index(%)			
Genotypes	**C1**	**C2**	**C3**	**Mean**	**C1**	**C2**	**C3**	**Mean**	**C1**	**C2**	**C3**	**Mean**
Annigeri 1	18.33	9.00	4.00	10.44	7.83	4.19	2.92	4.98	52.91	39.21	28.42	40.18
JAKI 9218	16.00	8.67	3.33	9.33	6.66	3.98	2.61	4.42	50.65	38.22	25.88	38.25
BGD 103	17.33	10.67	5.67	11.22	8.06	5.49	3.88	5.81	59.10	48.35	34.90	47.45
MNK 1	17.00	10.33	5.00	10.78	8.33	5.10	3.67	5.70	59.02	45.71	33.84	46.19
JG11	19.67	11.00	6.33	12.33	9.00	5.93	4.33	6.42	59.15	50.91	38.29	49.45
GBM 2	15.67	8.00	2.67	8.78	6.36	3.65	2.41	4.14	48.73	36.47	24.22	36.47
NBeG 47	14.67	7.67	2.33	8.22	7.50	3.18	2.07	4.25	58.43	32.29	20.82	37.18
ICC 1431	16.33	10.00	4.67	10.33	5.10	4.81	3.43	4.45	38.14	43.88	31.87	37.96
ICC 5003	17.67	9.67	4.33	10.56	6.98	4.47	3.15	4.86	48.96	40.95	30.08	39.99
ICCV 96029	18.00	6.33	1.33	8.56	5.99	2.84	1.00	3.28	45.15	29.41	11.56	28.71
Mean	17.07	9.13	3.97		7.18	4.37	2.95		52.02	40.54	27.99	
	SEm±		**LSD @5%**		**SEm±**		**LSD @5%**		**SEm±**		**LSD @5%**	
TRT	0.17916		0.47662		0.02995		0.07967		0.30264		0.8051	
CONC	0.05375		0.14299		0.00898		0.0239		0.09079		0.24153	
T*C	0.53748		1.42986		0.08985		0.23902		0.90792		2.41531	

Table 3: Effect of Salinity stress on pod number , seed yield and harvest index in chickpea genotypes