GENETIC BASIS OF VARIATION FOR SALINITY TOLERANCE IN OKRA (ABELMOSCHUS ESCULENTUS L.)

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Abstract

The development of salt tolerant plants through selection and breeding depends on the presence of the genetic variability within the crop species in response to salt stress, which must have significant genetic component. Such information is not extensively available in vegetable crops. The present study was carried out to gain some information on the genetic basis of variation for salinity tolerance in okra. North Carolina Mating Design II (NCM II) was used for the estimation of genetic components of variation in the traits affecting salinity tolerance.

The inheritance of the traits affecting salinity tolerance at the seedling stage appeared to be controlled by both additive and non-additive effects (dominance and epistasis). The narrow sense heritability estimates ranged from 40 to 65% and 7 to 70% and the estimates of broad sense heritability ranged from 65 to 99% and 20 to 99% for absolute and relative values. The additive effects were relatively more prominent and narrow sense heritability was moderate. The high additive component for absolute Na⁺ and K⁺/Na⁺ ratio at 60 and 80 m*M* NaCl, relative Na⁺ at 80 m*M* NaCl suggested that improvement for salinity tolerance in okra would be possible on the basis of these characteristics through selection and breeding.

The genetic variation for tolerance to NaCl salinity existed among the okra genotypes, which had considerable heritable component and, therefore, genetic improvement of okra genotypes for salinity tolerance through recurrent selection method is possible.

Introduction

The effects of salinity are devastating in arid and semi arid environments (Khan *et al.*, 2003; Azhar *et al.*, 2007). About 5% of cultivated land in the world is salinized (Flowers *et al.*, 1997), primarily due to insufficient drainage and low quality irrigation water (Binzel & Reuveni, 1994). To feed growing populations, marginal lands are to be brought under cultivation, which are not cropped due to their high degree of natural salinity or other toxicities (Flowers & Yeo, 1995). Pakistan is situated within the subtropical region with semi-arid to arid climate. According to a recent survey, of the 16.795 million ha irrigated area in Pakistan, 73% is categorized as non saline, 10% as slightly saline, 4% as moderately saline, 7% as strongly saline and 6% as miscellaneous type area (Anon., 2007). The saline soils contain mixture of different salts (Sandhu & Qureshi, 1986) but in Pakistan more than 60% soils are sodic and salinity stress is mostly due to Na⁺ salts (Plaut, 1993).

To tone down the salinity problem, soil amendments, water management and adjustment in the genetic architecture of the crops have been suggested. Among them, the evolution of salt tolerant varieties and development of transgenics are considered as more sustainable and economical choices to deal with the salinity problem (Epstien *et al.*, 1980; Rush & Epstein, 1981; Flowers & Yeo, 1995; Hollington, 2000; Khan *et al.*, 2001). Consequently, during the last few decades there has been marked increase in the attempts to tailor plants suitable for production on saline soils. Manipulations through somaclonal

variation and mutagenesis, marker assisted selection, somatic hybridization and genetic engineering can provide additional germplasm resources for breeding salt tolerant varieties (Saxena *et al.*, 1993; Mohan *et al.*, 1997; Maggio *et al.*, 2002; Ashraf, 2002). Some wild species in the family Malvaceae, had shown tolerance to drought, salinity and heat stresses (Gorham (1994). The use of such wild species having genes for tolerance to abiotic stresses would be useful for breeding commercially important members of this family i.e., cotton and okra.

Plants growing in saline soils face two problems, high salt concentrations in the soil solution (i.e., high osmotic pressure and correspondingly low soil water potential) and high concentrations of potentially toxic ions such as Cl- and Na+. Salt exclusion minimizes ion toxicity but accelerates water deficit in plants, whereas salt absorption facilitates osmotic adjustment but can lead to ion toxicity and nutritional imbalance (Günes *et al.*, 1996). Characteristics of a salt-tolerant variety include Na⁺ 'exclusion', K⁺/Na⁺ discrimination, retention of ions in the leaf sheath, tissue tolerance, ion partitioning into different-aged leaves, osmotic adjustment, transpiration efficiency, early vigour and early flowering leading to a shorter growing season and the increased water use efficiency (Colmer *et al.*, 2005).

The choice of breeding procedure depends upon the availability of information about the inheritance pattern, the number of genes with major effects, and the nature of gene action controlling salt tolerance in a species. The information on existence of variability for salinity tolerance is well documented in different crop species (Shereen *et al.*, 2001) but to a relatively lesser extent on the genetic basis of the variation for tolerance. The genetic studies reported on grasses (Ashraf *et al.*, 1986b), sorghum (Azhar & McNeilly, 1989), lucerne (Al-Khatib *et al.*, 1994), tomato (Foolad, 1996a, b) and maize (Rao & McNeilly, 1999; Khan *et al.*, 2003; Khan & McNeilly, 2005) provided evidence that salinity tolerance was genetically controlled in these crops.

Salt tolerance is a polygenic trait, its expression is affected by various genetic, developmental and physiological interactions within and between the plant, and interaction with the environments (Bernstein & Hayward, 1958, Shannon, 1984). The information obtained from the various species examined for salt tolerance suggested that different genetic setups may be controlling the character, from single major dominant/recessive genes to polygenic control with mainly additive effects, but with some degree of dominance toward tolerance (Gregorio & Senadhira, 1993; Ahsan *et al.*, 1996; Azhar *et al.*, 2007). Moderate to high estimates for narrow sense and broad sense heritabilities for salt tolerance have been recorded in some species indicating that improvement can be made by hybridization followed by stringent selection for salinity tolerance e.g., in alfalfa (Allen *et al.*, 1985), grasses (Ashraf *et al.*, 1986a, 1987), sorghum (Azhar & McNeilly, 1989), wheat (Ahsan *et al.*, 1996) and in tomato (Foolad, 1996a, 1996b).

Okra (*Abelmoschus esculentus* L.) is an annual, often cross pollinated vegetable of the tropical and subtropical areas. In Pakistan the total area under okra cultivation is about 0.0148 million ha and total production is about 0.112 million tones with average yield of about 7.55 tones/ha of green pods (Anon., 2008). Okra like other crops in Pakistan faces a dual threat of biotic and abiotic stresses. The crop is grown in suburbs where soils are saline and drainage water containing heavy metals and salts is used for irrigation which results in lower yields.

Keeping in view the importance of okra as vegetable crop, its nutritional value, area and production in Pakistan and the wide spread of salinity, the present studies were carried out with the major objective of providing information on the extent and basis of genetic variation for salinity tolerance in the okra genotypes at the seedling stage. The information gained would be useful for breeding salt tolerant okra varieties and hybrids in the country.

Materials and Methods

Plant material: The plant material used in this experiment was selected on the basis of its response to NaCl stress. Eighteen okra accessions/ genotypes including 5 most tolerant, 9 moderately tolerant and 4 non-tolerant were selected on the basis of relative ranking for salt tolerance at the seedling stage Table 1 (Ikram, 2009). Of these, 12 accessions/ genotypes Ikra III, Perbhani Kranti, IN-1048, Punjab Selection, Acc.No.019236, Acc.No.019225, Acc.No.015382, Acc.No.019217, Acc.No.019223, Acc.No.019231, Chinese Red and Ikra1 were used as male and six Acc.No.019221, Acc.No.019232, Acc.No.015380-10934, Acc.No.015371, Acc.No.019233 and Acc.No.000010-10237 were used as female for the development of 72 NCM II progenies.

Crossing procedure: The North Carolina Design II (NCM II) mating system of Comstock & Robinson, (1952) was used to make crosses. The selected 18 genotypes were grown in the vegetable experimental area of the Department of Plant Breeding and Genetics, University of Agriculture, Faisalabad. The crop was sown during Spring 2006 in a field on raised beds. The hand emasculation and pollination was carried out as described by Dhankhar & Mishra (2004) and Adeniji (2007). For this purpose fully developed buds were emasculated on the female parent plants. Such buds were fully swollen and pale green or light yellow. Flower buds for emasculation were held gently to avoid stress on the fragile attachments of the buds. A slight ring was made at the base of the flower bud with the help of a blade to facilitate easy removal of the petals and calyx sheath to expose the staminal tubes and the stigma. The under developed anthers were removed using a pair of forceps. Buds after emasculation were bagged to avoid contamination from foreign pollen.

The mature buds on pollen parent were covered with perforated butter paper bags. Pollen was collected in Petri dishes at about 10 a.m. and pollination was carried out by dusting pollen on stigma of emasculated buds with a fine brush. The pollinated buds were labeled and again covered with butter paper bags till pod formation and seed maturity. In this way, 10-15 crosses for each of the NCM II combinations were made to produce sufficient seed. The seed of the crosses was collected separately.

Assessment of NCM II progenies for NaCl tolerance: The response of the 72 NCM II families was assessed in 3 salinity treatments viz., control (0), 60 and 80 mM NaCl prepared in half strength Hoagland's nutrient solution (Hoagland & Arnon, 1950). These three levels were selected on the basis of the results of screening experiments, where 60 and 80 mM NaCl concentrations appeared to discriminate the okra genotypes for NaCl tolerance more evidently. The seed was surface sterilized in 2% bleach for 5 minutes and then washed with distilled water. Seeds were planted in plastic cups of 250 ml capacity filled with washed river sand using 3 replications and 6 NaCl concentrations in randomized complete block design during 2007. There were two cups per replication of each genotype.

Sr. No.	Genotypes	Source/ Origin	Ranks
1.	Acc. No.019232	PGRI, Islamabad	1
2.	Acc. No.000010-10237	PGRI, Islamabad	2
3.	Chinese Red	AARI, Faisalabad	3
4.	Ikra III	AARI, Faisalabad	4
5.	Acc. No.015371	PGRI, Islamabad	5
6.	Punjab Selection	AARI, Faisalabad	6
7.	Parbhani Kranti	AARI, Faisalabad	7
8.	Acc. No.019231	PGRI, Islamabad	8
9.	Ikra 1	AARI, Faisalabad	9
10.	Acc. No.019223	PGRI, Islamabad	10
11.	Acc. No.019236	PGRI, Islamabad	11
12.	Acc. No.019221	PGRI, Islamabad	12
13.	Acc. No.019217	PGRI, Islamabad	13
14.	IN-1048	AARI, Faisalabad	14
15.	Acc. No.019225	PGRI, Islamabad	36
16.	Acc. No.015380-10934	PGRI, Islamabad	37
17.	Acc. No.019233	PGRI, Islamabad	38
18.	Acc. No.015382	PGRI, Islamabad	39

Table 1. Plant material used for genetic study.

Ranking of okra genotypes for different seedling traits was based on evaluation of each parameter against NaCl stress with number 1 being the most tolerant (Ikram, 2009).

The experiment was carried out in a growth chamber adjusted at 28 ± 1 °C and relative humidity maintained between 75-80% with 16 hours photoperiod. The respective solutions were added at three days interval to compensate the evapo-transpiration loss in the cups. The data were recorded for the seedling traits from 3 week old seedlings.

Germination percentage: Ten surface sterilized seeds of each cross were sown in each cup (250 ml) moistened with respective solution in 3 replications. After 5 days the number of seeds germinated in each cup was counted. The germination percentage was calculated as under:

Germination percentage = $\frac{\text{No. of seeds germinated}}{\text{No. of seeds sown}} \times 100$

Root length (cm): After three weeks of growth, the seedlings were uprooted and washed with distilled water to remove sand. The root length of 6 randomly selected seedlings from each replication was measured in cm from the base of hypocotyl to the tip of longest root.

Shoot length (cm): The shoot length of 6 randomly selected seedlings from each replication was measured in cm from the base of hypocotyl to the shoot tip.

Na⁺ and K⁺ concentration: The leaves of seedlings were collected from each genotype separately, washed with distilled water, placed in 1.5 ml microfuge tube and stored in a freezer at -80°C for one week. The cell sap was extracted by crushing the leaves in microfuge tubes with the help of small iron balls using standard technique of centrifugation as explained by Gorham *et al.*, (1984). The Na⁺ and K⁺ ion contents in the

 \mathbf{K}^+ / \mathbf{Na}^+ ratio: Potassium uptake in relation to sodium (\mathbf{K}^+ / \mathbf{Na}^+ ratio) was estimated from the data of \mathbf{Na}^+ and \mathbf{K}^+ ions.

On the basis of these measurements, the responses of 72 NCM II progenies were compared using absolute values (absolute salt tolerance, Dewey, 1962) and relative values (relative salt tolerance, Maas, 1986). The relative salt tolerance of the okra genotypes was determined as mean value of a character in NaCl divided by mean value of a character in control expressed as percentage.

Statistical analysis: The mean data of the seedling traits in each replication for the control and NaCl concentrations were subjected to analysis of variance, separately as absolute and relative germination percentage, root length, shoot length, Na⁺ concentration, K⁺ concentration and K⁺/Na⁺ ratio. Analyses of variance of the 72 NCM II progenies were carried out following Becker (1992), and genetic components of variation were derived following Kearsey (1965) and Lawrence (1984). The computer package SPSS 12.0 for Windows was used for this purpose.

Results

Absolute tolerance: There were significant ($p \le 0.01$) differences among the male half sib families in control (0), 60 and 80 mM NaCl salinity for germination percentage, root and shoot length, Na⁺, K⁺ concentration and K⁺/Na⁺ ratio (Table 2). There were significant differences among female families for all the traits in control (0), 60 and 80 mM salinity except for K⁺/Na⁺ ratio which indicated non significant (P \ge 0.05) differences in 60 mM NaCl salinity. The interaction male × female was significant for germination percentage, root length, shoot length and K⁺ concentration in control (0), 60 and 80 mM NaCl salinity which indicated the involvement of additive and non-additive effects for controlling these traits (Table 2). The interaction male × female was significant ($p \ge 0.05$) in 60 and 80 mM NaCl salinity which indicated that additive effects were more important for the control of these traits. The female and male mean square ratio was non significant ($p \ge 0.05$) at all the concentrations which suggested that maternal effects were not important in controlling the traits at these NaCl concentrations in the okra genotypes.

The non additive effects (H_R) were greater than additive (D_R) and environmental effects (E) in control and two NaCl concentrations for germination percentage, root length, shoot length and K⁺ concentration (Fig. 1a,b). However, the additive effects (D_R) were smaller than the non additive (H_R) for Na⁺ concentration and K⁺/Na⁺ ratio in non saline condition but greater in 60 and 80 mM NaCl concentrations for both the traits. The potence ratio was about 1 or above in control, 60 and 80 mM NaCl concentration, which suggested nearly complete or overdominance for all the traits, both in non saline and saline growth mediums. However, the potence ratio was less than 1 under saline conditions for Na⁺ concentration and K⁺/Na⁺ ratio indicating the partial dominance of additive gene effects for salinity tolerance. The estimates of narrow sense heritability ranged from 0.395 to 0.653 whereas the corresponding estimates of broad sense heritability ranged from 0.649 to 0.998 in control and in 60 and 80 mM NaCl concentrations.

Table 2. Mean squares from analysis of variance of NCM-II progenies of okra for absolute values in 0 (control) 60 and 80 mM NaCl

Source of	Df	Germination	Root	Shoot	Na'	K'	K'/Na'	
variation	Ы	% age	length	length	concentration	concentration	ratio	
0 mM NaCl (Control)								
Replications	2	54.780 ^{NS}	0.304 ^{NS}	5.264 ^{NS}	1.213 ^{NS}	57.456 ^{NS}	0.383 ^{NS}	
Males (m)	11	1042.940**	16.970**	25.075**	77.468 **	422.117**	8.626 **	
Females (f)	5	1250.050**	6.704*	25.037*	81.143 **	260.661**	9.555 **	
Males × Females	55	209.300*	2.398*	7.925**	14.600 *	73.669*	1.760 *	
Error	142	143.700	1.676	4.743	10.199	51.122	1.230	
MS _f /MS _m		1.199 ^{NS}	0.395 ^{NS}	0.998 ^{NS}	1.047^{NS}	0.618 ^{NS}	1.108	
60 mM NaCl								
Replications	2	63.000 ^{NS}	0.877^{NS}	2.332 ^{NS}	262.57 ^{NS}	1.376 ^{NS}	0.051 ^{NS}	
Males (m)	11	1168.300**	13.804**	38.901**	2278.50 **	181.883**	0.398 **	
Females (f)	5	2356.920**	8.433**	15.077*	944.23 *	115.906**	0.039 ^{NS}	
Males × Females	55	324.400*	2.425*	6.286*	347.49 ^{NS}	30.650*	0.070^{NS}	
Error	142	226.050	1.678	4.365	258.85	21.319	0.057	
MS _f /MS _m		2.017 ^{NS}	0.611 ^{NS}	0.388 ^{NS}	0.414 ^{NS}	0.637 ^{NS}	0.098 ^{NS}	
80 mM NaCl								
Replications	2	73.200 ^{NS}	0.304 ^{NS}	11.971 ^{NS}	109.62 ^{NS}	1.485 ^{NS}	0.030 ^{NS}	
Males (m)	11	1277.100**	16.970**	68.203**	6422.64 **	169.115 **	0.241 **	
Females (f)	5	1534.970**	6.704**	39.758**	2372.01 *	181.618 **	0.081 *	
Males × Females	55	242.900*	2.398*	11.178*	782.36 ^{NS}	41.698 *	0.033 ^{NS}	
Error	142	168.610	2.065	7.743	715.93	26.382	0.025	
MS_f/MS_m		1.202 ^{NS}	0.806 ^{NS}	0.583 ^{NS}	0.369 ^{NS}	1.074^{NS}	0.335 ^{NS}	

Relative tolerance: The male half sib families differed significantly at $p \le 0.01$ for relative shoot length, Na⁺ & K⁺ concentration in both NaCl concentrations and for relative germination percentage, K^+/Na^+ ratio in 80 mM NaCl salinity but at p ≤ 0.05 for relative K^+/Na^+ ratio in 60 mM NaCl (Table 3). There were non significant (p \geq 0.05) differences among males in 60 and 80 mM NaCl salinity for relative root length and for relative germination percentage in 60 mM NaCl salinity. There were significant differences among female families for all the traits in 60 and 80 mM salinity except for relative germination percentage, root length and K⁺ concentration which had non significant (p \ge 0.05) differences in 60 mM NaCl salinity. The interaction between male \times female was significant for relative germination percentage in 60 mM NaCl, and for relative root length, shoot length, K⁺/Na⁺ ratio in 60 and 80 mM NaCl salinity which indicated the involvement of additive and non-additive effects for controlling these traits (Table 3). The interaction between male \times female was non significant (p \ge 0.05) for Na⁺ and K⁺ concentration in 60 and 80 mM NaCl salinity which suggested that additive effects were more important for the control of these traits. The maternal effects were not detected in controlling the traits in the okra genotypes because the female and male mean square ratio was non significant ($p \ge 0.05$) at all the NaCl concentrations.

The non additive effects (H_R) were greater than additive (D_R) and environmental effects (E) for relative germination percentage, root length, shoot length and K⁺/Na⁺ ratio in the two NaCl concentrations and relative Na⁺ concentration in 60 mM NaCl (Fig. 2a,b). On the other hand the additive effects (D_R) were greater than the non additive (H_R) for relative Na⁺ concentration in 80 mM and relative K⁺ concentration in saline condition at 60 and 80 mM NaCl concentrations. The potence ratio was above 1 in 60 and 80 mM NaCl concentration for all the traits suggestive of overdominance. However, the potence ratio was less than 1 under saline conditions for relative Na⁺ concentration in 60 mM and relative K⁺ concentration in both 60 and 80 mM demonstrating the partial dominance of additive gene effects for salinity tolerance. The estimates of narrow sense heritability ranged from 0.066 to 0.699 whereas the corresponding estimates of broad sense heritability ranged from 0.202 to 0.994 in 60 and 80 mM NaCl concentrations.

GENETIC BASIS OF SALINITY TOLERANCE IN OKRA



Fig. 1a. Components of variation and heritabilities for NCM-II progenies of okra for absolute values.

IKRAM-UL-HAQ ET AL.,



Fig. 1b. Components of variation and heritabilities for NCM-II progenies of okra for absolute values.

GENETIC BASIS OF SALINITY TOLERANCE IN OKRA



Fig. 2a.Components of variation and heritabilities for NCM-II progenies of okra for relative values.

1576

IKRAM-UL-HAQ ET AL.,



Fig. 2b.Components of variation and heritabilities for NCM-II progenies of okra for relative values.

Table 3. Mean squares from analysis of variance of NCM-II progenies of

oki a for relative values in 60 and 80 milli NaCi.								
Source of	Df	Germination	Root	Shoot	Na^+	\mathbf{K}^+	K ⁺ /Na ⁺	
variation	וע	% age	length	length	concentration	concentration	ratio	
60 mM NaCl								
Replications	2	75.720 ^{NS}	289.498 ^{NS}	203.94 ^{NS}	73644 ^{NS}	41.730 ^{NS}	0.012 ^{NS}	
Males (m)	11	826.970 ^{NS}	699.575 ^{NS}	1146.12**	439156 **	1850.76**	0.055*	
Females (f)	5	1129.14 ^{NS}	606.310 ^{NS}	1081.67**	247221*	348.420 ^{NS}	0.062*	
Males × Females	55	716.230**	438.936**	425.51*	84233 ^{NS}	515.440 ^{NS}	0.026**	
Error	142	422.450	244.851	274.65	64574	529.710	0.015	
MS _f /MS _m		1.365 ^{NS}	0.867^{NS}	0.944 ^{NS}	0.563 ^{NS}	0.188 ^{NS}	1.12 ^{NS}	
80 mM NaCl								
Replications	2	17.590 ^{NS}	157.69 ^{NS}	763.650 ^{NS}	10934 ^{NS}	109.310 ^{NS}	10.668 ^{NS}	
Males (m)	11	1649.980**	766.93 ^{NS}	2739.120**	1174351 **	2338.640**	118.387**	
Females (f)	5	944.550*	1527.28*	1827.440*	891565 **	1238.300 *	121.046**	
Males × Females	55	393.420 ^{NS}	579.16*	695.470*	155978 ^{NS}	496.940 ^{NS}	21.392*	
Error	142	311.250	372.630	468.280	137109	466.760	14.855	
MS_f/MS_m		0.572 ^{NS}	1.991 ^{NS}	0.667 ^{NS}	0.759 ^{NS}	0.529 ^{NS}	1.022 ^{NS}	

Discussion

For affecting salt tolerance in a crop there must be sufficient genetic variation within the crop in response to salt, and this variation should be genetically controlled, to make selection and breeding possible for a target trait (Epstein & Norlyn, 1977; Shannon, 1978; Epstein et al., 1980). Both additive and non additive genetic effects were important for the control of salinity tolerance in the 72 progenies assessed at the seedling stage of okra. The data indicated that genes with both additive and dominance effects were important for the absolute germination percentage, root length and shoot length in control and both NaCl concentrations and for relative shoot length in saline conditions (Tables 2 & 3). The magnitude of non additive effects (H_R) was greater than additive effects at these concentrations. But for relative germination and relative root length at 60 mM NaCl genes with dominance effects were more important for the control of salinity tolerance. The additive portion can be fixed through selection in a breeding program while the dominance is useful for hybrid production. Therefore, the pattern of gene action exhibited is advantageous in breeding program aimed at improvement of salinity tolerance in okra. In maize both additive and non-additive effects were significant for the expression of salt tolerance, the later was predominant under stress at the seedling stage (Khan & McNeilly, 2005). Similarly, for germination, additive effects were important and heritability values were higher in tomato (Foolad & Jones, 1991). At the seedling stage, both additive and dominance genetic effects were important in rice (Gregoria & Senadhira, 1993).

The potence ratio estimated as $(D_R/H_R)^{\frac{1}{2}}$ indicated overdominance for the absolute and relative germination percentage, root length and shoot length, absolute K⁺ ion concentration and relative K⁺/Na⁺ ratio. But for absolute Na⁺, absolute K⁺/Na⁺ ratio, relative K⁺ concentration at both the levels and relative Na⁺ concentration at 80 mM NaCl revealed additive gene effects for salinity tolerance with partial dominance. These results suggested that selection can be made on the basis of these estimates of ions accumulation for increased salinity tolerance. Nevertheless, potence ratio may not be true estimation of the degree of dominance in the presence of non allelic interaction (epistasis). These estimates should be discussed with care in the present situation because NCM II does not provide information about epistatic effects, and involvement of these effects is expected for the quantitative nature of the traits associated with salinity tolerance (Khan & McNeilly, 2000). Rao & McNeilly (1999) using NCM II mating design found that both additive and dominance properties were important in controlling the expression of salt tolerance in maize. The higher non additive component in the present studies suggested that the pattern of the inheritance of tolerance was complex, as was found in sorghum at higher salinity concentrations (Azhar & McNeilly, 1988).

Both additive and non additive genetic effects were considerable for absolute K^+ in control and salinity and for relative K^+/Na^+ ratio in both NaCl concentrations. Absolute Na⁺ in control and relative Na⁺ in 60 m*M* exhibited both additive and non additive effects but in increased NaCl, the genes with additive effects were more important. For absolute K^+/Na^+ ratio under non stress conditions, additive and dominance effects were important, but at higher salinity, the genes with additive effects appeared to control the salt tolerance. Thus, the high additive component for absolute Na⁺ and K⁺/Na⁺ ratio at 60 and 80 m*M*, relative Na⁺ at 80 m*M* NaCl suggested that improvement for salinity tolerance in okra would be possible on the basis of these characteristics through selection and breeding. The additive genetic effects with partial dominance were reported for the traits like Na⁺, K⁺, Cl⁻ contents , K⁺/Na⁺ ratio, osmotic pressure, plant height and grain yield per plant in wheat (Salam, 1993).

The maternal effects were either absent or too small to be detected by the NCM II analysis, similar findings for Mn in maize were observed by Khan & McNeilly (2000) but Rao & McNeilly (1999) suggested considerable maternal effects for affecting NaCl stress in maize. These contradictions in the results may be due to the use of different crop and environment.

Heritability is an effective tool with the plant breeders to separate the heritable variability from the total phenotypic variation. The high heritability estimate suggested that genetic improvement in the material under study is possible. The narrow sense heritability estimates in the present study ranged from 0.395 to 0.653 and 0.066 to 0.699 for absolute and relative values respectively. The estimates of broad sense heritability were relatively greater than narrow sense and ranged from 0.649 to 0.998 and 0.202 to 0.994 for absolute and relative values respectively.. The considerably high broad sense heritability estimates suggested that salinity tolerance in these okra genotypes was genetically determined. The moderate narrow sense heritability estimates indicated the importance of genes with additive effects in the character expression and suggested that breeding and selection could be used to improve salinity tolerance in okra. Narrow sense heritability ranged between 0.19 to 0.72 and 0.50 to 0.98 in 7 grass and 4 forage species (Ashraf et al., 1986a, 1987) and 0.70–0.95 for ion contents in spring wheat (Ahsan et al., 1996). Naveed et al., (2009) found that narrow sense heritability and genetic advance were high for fruit yield under non-stress conditions than under drought stress and suggested that direct selection would only be possible under non stress conditions. Foolad (1996a) found heritability estimates in F₂ and F₃ tomato generations for rapid germination under various salt concentrations ranging from 0.67 to 0.76, and moderate estimates of narrow sense heritability (0.49) were obtained for tomato shoot dry weight (Foolad, 1996b). Broad sense heritability in sorghum for root length grown in saline nutrient solution, a character used to quantify tolerance, ranged from 0.38 to 0.73 (Azhar & McNeilly, 1989), and in alfalfa 0.50 for seed germination after five cycles of selection in saline conditions (Allen et al., 1985).

It has been argued that the heritability and additive variance components increased with an increase in the salinity (Mather, 1973; Blum, 1988; Hoffman & Parsons, 1991). The estimates of narrow sense heritability and additive variance for absolute shoot length

increased with increase in NaCl concentrations. For absolute germination percentage the additive variance decreased from 60 to 80 m*M*, for absolute root length, K^+ and K^+/Na^+ ratio from 0 to 60 m*M* NaCl concentrations. Such changes are natural because different genes may affect the same trait under different environments (Richards, 1978; Rumbaugh *et al.*, 1984). Whilst in sorghum the additive variance was decreased with increase in NaCl concentration from 100 to 150 m*M* (Azhar & McNeilly, 1988), and in maize from 0 to 60 mM (Rao & McNeilly, 1999) and in cotton from 0 to 250 m*M* (Azhar *et al.*, 2007). Thus the difference in magnitude of additive variance noted here is not unusual with earlier findings and suggested that the pattern of inheritance of salinity tolerance in okra is complex.

The information gathered from the NCM II analysis, suggested that variation for salinity tolerance in okra at seedling stage was governed by both additive and non additive genetic effects. The additive effects were relatively more prominent and narrow sense heritability was moderate. It is concluded that the genetic variation for tolerance to NaCl salinity existed among the okra genotypes, which had considerable heritable component and, therefore, genetic improvement of okra genotypes for salinity tolerance through breeding and recurrent selection is possible.

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References

- Adeniji, O.T. 2007. Heritability and number of genes governing pod yield in West African okra (*Abelmoschus caillei* (A. Chev) stevels. *Agric. J.*, 2: 483-486
- Ahsan, M., D. Write and D.S. Virk. 1996. Genetic analysis of salt tolerance in spring wheat (*Triticum aestivum* L.). Cereal Res. Commun., 24: 353-360.
- Al-Khatib, M., T. McNeilly and J.C. Collins. 1994. Between and within cultivar variability in salt tolerance in lucerne (*Medicago sativa* L.). *Genet. Resour. Crop Evol.*, 41: 159-164.
- Allen, S.G., A.K. Dobrenz, M.H. Schonhorst and J.E. Stoner. 1985. Heritability of NaCl tolerance in germinating alfalfa seeds. Agron. J., 77: 99-101.
- Anonymous. 2007. Agricultural Statistics of Pakistan. MINFAL, Islamabad, Pakistan.
- Anonymous. 2008. Fruits, Vegetable and Condiments Statistics, 2006-07. MINFAL, Islamabad, Pakistan.
- Ashraf, M. 2002. Exploitation of genetic variation for improvement of salt tolerance in spring wheat. In: *Prospects for Saline Agriculture*. (Eds.): R. Ahmad and K.A. Malik. Kluwer, Dordrecht. p. 113-121.
- Ashraf, M., T. McNeilly and A.D. Bradshaw. 1986a. The potential for evolution of salt tolerance in seven grass species. *New Phytol.*, 103: 299-309
- Ashraf, M., T. McNeilly and A.D. Bradshaw. 1986b. Tolerance of sodium chloride and its genetic basis in natural populations of four grass species. *New Phytol.*, 103: 725-734.
- Ashraf, M., T. McNeilly and A.D. Bradshaw. 1987. Selection and heritability of tolerance to Sodium chloride in four forage species. *Crop Sci.*, 27: 232-234
- Azhar, F.M. and T. McNeilly. 1988. The genetic basis of variation for salt tolerance in *Sorghum bicolor* (L.) Moench seedlings. *Plant Breed.*, 101: 114-121.
- Azhar, F.M. and T. McNeilly. 1989. Heritability estimates of variation for NaCl tolerance in Sorghum bicolour (L.) Moench seedlings. Euphytica, 43: 69-72

- Azhar, F.M., A.A. Khan and N. Saleem. 2007. Genetic mechanism controlling salt tolerance in Gossypium hirsutum L. seedlings. Pak. J. Bot., 39:115-121
- Becker, W.A. 1992. *Manual of Quantitative Genetics*. 5th ed. Acad. Enterprises, Pullman, Washington, USA.
- Bernstein, L. and H.E. Hayward. 1958. Physiology of salt tolerance. Annu. Rev. Plant Physiol., 9: 25-46.
- Binzel, M.L. and M. Reuveni. 1994. Cellular Mechanism of salt tolerance in plants. *Hort. Rev.*, 16 PT, 38: 33-69.
- Blum, A. 1988. *Plant Breeding for Stress Environments*. CRC. Press. Inc. Boca Raton, Florida. USA.
- Colmer, T.D., R. Munns and T.J. Flowers. 2005. Improving salt tolerance of wheat and barley: future prospects. *Aust. J. Expt. Agric.*, 45: 1425-1443
- Comstock, R.E. and H.F. Robinson, 1952. Estimation of average dominance of genes. Heterosis, Ch 30 Iowa State College Press, USA.
- Dewey, D.R. 1962. Breeding crested wheatgrass for salt tolerance. Crop Sci., 2: 403-407.
- Dhankhar, B. S. and J. P. Mishra. 2004. Objectives of okra breeding. J. New Seeds, 6: 195-209.
- Epstein, E. and J.D. Norlyn.1977. Sea-water based crop production: A feasibility study. *Science*, 197: 249-251.
- Epstein, E., D.W. Rush, R.W. Kingsbury, D.B. Kellery, G.A. Cunningham and A.F. Worna. 1980. Saline culture of crops: A genetic approach. *Science*, 210: 399-404.
- Flowers, T.J. and A.R. Yeo. 1995. Breeding for salinity resistance in crop plants: where next. Aust. J. Plant Physiol., 22: 875-884.
- Flowers, T.J., A. Garcia, M. Koyama and A.R. Yeo. 1997. Breeding for salt tolerance in crop plants-the role of molecular biology. *Acta Physiolo. Plant.*, 19: 427-433.
- Foolad, M.R. 1996a. Response to selection for tolerance during germination in tomato seed derived from P1-174263. J. Amer. Soc. Hort. Sci., 121: 1006-1011.
- Foolad, M.R. 1996b. Genetic analysis of salt tolerance during vegetative growth in tomato, *Lycopersican esculentum* Mill. *Plant Breed.*, 115(4): 245-250.
- Foolad, M.R. and R.A. Jones. 1991. Genetic analysis of salt tolerance during germination in *Lycopersicon. Theor. Appli. Genet.*, 81: 321-326.
- Gorham, J. 1994. Salt tolerance in the Triticeae: K/Na discrimination in some perennial wheat grasses and their amphiploids with wheat. J. Expt. Bot., 273: 441-447.
- Gorham, J., E. McDonnell and R.G. Wyn Jones. 1984. Pinitol and other solutes in salt stressed Sesbania aculeate. Z. Pflanzenphys, 114: 173-178
- Gregorio, G.B. and D. Senadhira. 1993. Genetic analysis of salinity tolerance in rice. *Theor. Appl. Genet.*, 86: 333-338.
- Günes, A., A. Inal and A. Alpaslan. 1996. Effect of salinity on stomatal resistance, proline, and mineral composition of pepper. J. Plant Nutr., 19: 389-396.
- Hoagland, D.R. and D.I. Arnon. 1950. *The water culture method for growing plants without soil*. Agric. Expt. Sta. UC Riverside. Circular No. 347.
- Hoffman, A.A. and P.A. Parsons. 1991. *Evolutionary Genetics and Environmental Stress*. Oxford Univ. Press, New York.
- Hollington, P.A. 2000. Technological breakthroughs in screening/ breeding wheat varieties for salt tolerance. In: *Proceedings of the National Conference 'Salinity Management in Agriculture'*, (Eds.): S.K. Gupta, S.K. Sharma and N.K. Tyagi. December 1998. Central Soil Salinity Res. Inst. Karnal, India, pp. 273-289.
- Ikram, H. 2009. *Genetic basis of variation for salinity tolerance in okra (Abelmoschus esculentus* L). Ph.D. thesis. University of Agriculture, Faisalabad, Pakistan.
- Kearsey, M.J. 1965. Biometrical analysis of a random mating population: Comparison of five experimental designs. *Heredity*, 20: 205-235.
- Khan, A.A. and T. McNeilly. 2000. Genetic analysis of aluminium and manganese tolerance in maize (*Zea mays* L.). J. Genet. Breed., 54: 245-249.

1580

- Khan, A.A. and T. McNeilly. 2005. Triple test cross analysis for salinity tolerance based upon seedling root length in maize (*Zea mays* L.). *Breed. Sci.*, 55: 321-325.
- Khan, A.A., S.A. Rao and T. McNeilly. 2003. Assessment of salinity tolerance based upon seedling root growth response functions in maize (*Zea mays L.*). *Euphytica*, 131: 81-89.
- Khan, A.A., T. McNeilly and F.M. Azhar. 2001. Stress tolerance in crop plants. (Review). Int. J. Agric. Biol., 3: 250-55.
- Lawrence, M.J. 1984. The genetical analysis of ecological traits. In: *Evolutionary Ecology*. (Ed.): B. Shorrocks. pp. 27-63. Blackwell Sci. Publ. Oxford, London, Edinburgh.
- Maas, E.V. 1986. Salt tolerance of plants. Appl. Agric. Res., 1: 12-26.
- Maggio, A., T. Matsumoto, P.M. Hasegawa, J.M. Pardo and R.A. Bressen. 2002. The long and winding road to halotolerance genes. In: *Salinity: Environment-Plants-Molecules*. pp. 505-533. Kluwer Academic Publisher, (Eds.): A. Lauchli and U. Luttge. The Netherlands.
- Mather, K. 1973. Genetic Structure of Populations. Chapman and Hall, London.
- Mohan, M., S. Nair, A. Bhagwat, T.G. Krishna, M. Yano, C.R. Bhatia and T. Sasaki. 1997. Genome mapping, molecular markers and marker assisted selection in crop plants. *Mol. Breed.*, 3: 87-103.
- Naveed, A., A.A. Khan and I.A. Khan. 2009. Generation mean analysis of water stress tolerance in okra (*Abelmoschus esculentus* L). *Pak. J. Bot.*, 41: 195-205.
- Plaut, Z. 1993. Photosynthesis in plant/crops under water and salt stress. In: *Handbook of Plant and Cop Physiology*. (Ed.): M. Pessarakli. Marcel Dekker Inc., New York. pp. 587-602.
- Rao, S.A and T. McNeilly. 1999. Genetic basis of variation for salt tolerance in maize (*Zea mays* L.). *Euphytica*, 108:145-150.
- Richards, R.A. 1978. Genetic analysis of drought stress response in rapeseed (*Brassica campestris* and *Brassica napus*). I. Assessment of environments for maximum selection response in grain yield. *Euphytica*, 27: 609-615.
- Rumbaugh, M.O., K.H. Asay and O.A. Johnson. 1984. Influence of drought stress on genetic variance of alfalfa and wheat grass seedling. *Crop Sci.*, 24: 297-303.
- Rush, D.W. and E. Epstein. 1981. Breeding and selection for salt tolerance by the incorporation of wild germplasm into a domesticated tomato. J. Amer. Soc. Hort. Sci., 106: 669-704.
- Salam, A. 1993. *Physiological/Genetical studies on the aspects of salt tolerance in wheat (Triticum aestivum* L.) Ph.D. thesis, University of Wales, UK
- Sandhu, G.R. and R.H. Qureshi. 1986. Salt affected soils of Pakistan and their utilization. Proc. Rev. Res., 5: 106.
- Saxena, N.P., C. Johansen., M.C. Saxena and S.N. Selim. 1993. Selection for drought and salinity resistance in cool season food legumes. In: *Breeding for Stress Tolerance in Cool-Season Food Legumes*. (Eds.): K.B. Singh and N.P. Saxena. John Wiley and Sons, UK.
- Shannon, M.C. 1978. Testing salt tolerance variability among long wheatgrass lines. *Agron. J.*, 70: 719-722.
- Shannon, M.C. 1984. Breeding, selection and genetics of salt tolerance. In: Salinity Tolerance in Plants-Strategies for Crop Improvement. (Eds.): R.C. Staples, R.C. and G.A. Toenniessen. pp. 231-254. John Wiley & Sons, New York, USA.
- Shereen, A., R. Ansari and A.Q. Soomro. 2001. Salt tolerance index in soybean (*Glycine max* L): effect on growth and ion relations. *Pak. J. Bot.*, 33: 393-402.

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