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Combining Ability and Heterosis for Yield, Fiber Quality and Physio-Chemical Parameters in Cotton Under Salt Stress

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ABSTRACT

Background: There are several abiotic stresses that can limit the yield of cotton crops. Among those stresses, salinity is one that significantly reduces crop productivity. The primary purpose of this research was to explore the breeding potential of cotton genotypes for salinity tolerance. **Results:** For this purpose, the salt-tolerant genotypes, NIAB-545, CIM-595, Coker-307, FH-113, FH-942, DNH-40 and salt sensitive genotypes Cyto-178, VH-363, FH-326 were grown under field conditions. These accessions were hybridized in a Line × Tester mating design to identify the genetic components of yield-contributing traits under control, 10 dSm^{-1}, 15 dSm^{-1} and 20 dSm^{-1} salt stress conditions. Significant levels of variation were found among the genotypes for root and shoot length, fresh shoot and root weight, dry root and shoot weight, Na^+, K^+ and K^+/Na^+, proline, H_2O_2, POD and CAT. Analysis of variance indicated that there were significant differences among genotypes for these traits. Salt stress caused a reduction in all traits except Na^+, GOT, H_2O_2, and proline. Non-additive gene action was found for yield, fiber, ionic, and biochemical traits except for Na^+ ions. The proportional contribution to total variance for all traits was found to be maximum in lines and lower for testers. Among the lines, FH-942 was found to be the best general combiner for plant height, number of bolls, and seed cotton yield under control, 10 dSm^{-1},15 dSm^{-1} and 20 dSm^{-1} conditions. NIAB-545 was the best general combiner for fiber fineness, fiber length, fiber strength,

GOT and catalase under control, 10 dSm^{-1} and 15 dSm^{-1} and 20 dSm^{-1} salt stress. The FH-942 genotype also showed greater general combining ability (GCA) effects for K^+, proline and CAT under control and salt stress conditions. Among the testers, FH-326 was best general combiner for most traits, while Cyto-178 had positive and significant GCA effects for fiber fineness, fiber length, fiber strength, POD and catalase under control and salt stress conditions. **Conclusion:** The cross combination FH-942×FH-326 showed positive and significant specific combining ability effects for most of the traits and exhibited significant heterosis for most of studied traits. The selected hybrid can be effectively utilized in breeding programs aimed at improving salt tolerance in cotton.

Introduction:

Salinity is an important factor that limits growth and development by altering the morphological, physiological, and biochemical attributes of cotton (Rani *et al.,* 2019). The risk of soil salinization to agricultural productivity and food security is already evident and is potentially much greater in the future (Corwin, 2021). Salt stress exerts detrimental effects on primary and secondary plant processes, namely germination, photosynthetic pigments, chlorophyll concentration, antioxidant levels, and seed cotton yield (Tian *et al.,* 2020). It imposes limitations on the level of chlorophyll concentration because it damages photosynthetic pigments (Sharif *et al.,* 2019). High levels of sodium chloride (NaCl) concentrations reduce water potential and making it difficult for the root to extract water from soil (Pessarakli, 2021). Meanwhile, high levels of Na^+ and Cl^- ions in plant cells leads to the inhibition of several enzymatic pathways. Likewise, salinity disturbs the biochemical processes within the cellular organelles of cotton, causing a reduction in antioxidant levels which leads to oxidative damage (Wang *et al.,* 2017c). Salt stress leads to the overproduction of reactive oxygen species (ROS) such as superoxide anions, hydrogen peroxide (H_2O_2), and hydroxyl radicals (Sharif et al. 2019; Chaudhary *et al.,* 2024). Crop plants have an effective and complex antioxidant defense system, including enzymatic antioxidants like peroxidase (POD) and catalase (CAT), and non- enzymatic antioxidants like proline, to protect cells from the harmful impact of ROS. SOD is the first antioxidant to act when ROS begins to produce converting O^{-2} into H_2O_2 and water in the cytosol, mitochondria, and chloroplasts (Foyer and Noctor, 2000). POD, the next antioxidant, acts as barrier by converting H_2O_2 into H_2O and O_2. H_2O_2 is also scavenged by CAT which further dismutates it into water and oxygen (Ashraf, 2009). Considering the aforementioned losses and effects of salinity, it is necessary to develop cotton accessions that exhibits enhanced levels of tolerance to saline conditions.

The degree of tolerance to salt stress varies without significantly affecting the yield of various field crops. For instance, barley (Patterson *et al.,* 2009) crop is considered salt tolerant, while sorghum (De Souza Miranda *et al.,* 2016), sunflower (Khan *et al.,* 2014), and rye (Volkov, 2015) are regarded as moderately salt tolerant. Cotton is also a moderately salt tolerant (Ashraf, 2002), however the ability to tolerate stress varies depending on the growth stage of the crop. It has been reported that the vegetative stage is more tolerant to salt stress, whereas the seedling stage is significantly affected by salt stress. To tackle this problem, sustainable measures are required to enhance and stabilize the production of quality cotton so that it can help to alleviate the burden on the country's economy.

The Line × Tester analysis is one of the suitable analyses to evaluate the combining ability of plants in various combinations (Sprague and Tatum, 1942). One of the advantages of this mating design is that it does not require the assumptions needed in diallel mating designs. . However, the presence of genetic diversity in cotton genotypes is necessary. (Turk, 2017). General combining ability is due to additive gene action, and specific combining ability is due to non-additive gene actions (dominant or epistatic). Various plant researchers have observed additive × additive, additive × dominance, and dominance × dominance gene action for various traits in the cotton crop breeding program for salinity tolerance (Ayaz and Khan, 2015; Dave *et al.,* 2015; Murthy *et al.,* 2018).

Taking into consideration the impact of climate change and the contribution of the cotton crop to the global economy, the present research was designed to evaluate a large number of cotton genotypes for salinity tolerance using various morphological, physiological, and biochemical attributes. The research was conducted with objectives to determine the gene action controlling these traits and to conduct heterotic analysis to assess the performance of hybrids under different saline conditions. Identifying heritable variability in existing accessions is a prerequisite for developing climate resilient germplasm

(Sezener *et al.,* 2015). Moreover, understanding the genetic basis of salt stress, yield, and fibre quality related traits is essential for initiating a cotton breeding program (Meredith and Bridge, 1972; Ahmad *et al.,* 2003; Mubarik *et al.,* 2020).

Materials and Methods:

Development of F1 seed

The nine parents included six salt tolerant genotypes-namely, DNH-40, CIM-595, Coker-307, NIAB-545, FH-942, FH-113 which were used as female parents (lines), and three salt-sensitive genotypes-namely, Cyto-178, VH-363, FH-326 which were utilized as male parents (testers) during hybridization. The seeds of the salt-tolerant and salt-sensitive parents were sown in the field in April, 2013. All recommended plant production and protection practices were followed to ensure a healthy crop stand. At the onset of flowering, the salt-tolerant and salt-sensitive genotypes were crossed in a Line × Tester design, resulting in 18 crosses. Intensive crossing was carried out to generate enough F_0 seeds for evaluation. Moreover, some floral buds on the parent plants were kept covered at all times to prevent cross pollination and to obtain selfed seeds.

Evaluation of parents and crosses

Parents and crossed seeds were sown in four different containers (2.43 m × 1.82 m) placed outside the glass house of the Department of Plant Breeding and Genetics, University of Agriculture, Faisalabad (31.4504˚N, 73.1350˚E). The growing media used in the containers was soil with a pH of 6.7. In each container, s five genotypes including six plants of each genotype were sown during march (plant-to-plant spacing was 30 cm and line-to-line spacing was also 30 cm). The plants were grown in a randomized complete block design (RCBD) in a split arrangement with two factors and three replications. When the plants reached the four-leaf stage, a mixture of salt and water was applied. NaCl was dissolved in water, and the electrical conductivity (EC) of the solution was measured. The EC of the solution was increased to 10 dSm^{-1}, 15 dSm^{-1}, and 20 dSm^{-1} by calculating the required amount of salt to be dissolved in water. The first dose of salt stress was applied when the plants were two weeks old to develop a salinity stress level up to 7.5 dSm^{-1}. The second dose was applied when the seedlings were four weeks old to raise the salinity levels to develop 10 dSm^{-1}, 15 dSm^{-1}, and 20 dSm^{-1}. Salt stress was increased gradually in two stages to avoid shock to the seedlings. . The salt stress treatment continued for 70 days before the first picking. The containers were irrigated with normal water (EC = 1.6) after achieving the desired level of salt stress.

Measurement of seed cotton yield related traits

At harvest, data were recorded for several yield-related traits. Plant height was measured from the cotyledonary node to the apical bud once elongation had ceased. The number of bolls per plant was counted for each of the six plants grown per genotype and then averaged to obtain the number of bolls for each genotype. Seed cotton was picked from each of the six plants of every genotype under each salinity level, weighed using an electronic balance, and averaged per plant. The weight of an individual boll was determined by dividing the seed cotton weight sample by the total number of bolls picked, and then average boll weight was used for statistical analysis.

Measurement of fiber quality traits

Samples of seed cotton were ginned using a single roller-ginning machine. The ginned samples were then processed to determine fiber quality parameters such as fiber length, fiber fineness, and fiber strength, using a High Volume Instrument (HVI-900) system available in the laboratory of the Department of Fibre and Textile Technology, University of Agriculture, Faisalabad.

Estimation of Sodium and Potassium:

The concentrations of potassium (K^+) and sodium (Na^+) were analysed in sub-samples of dried plant leaves obtained after crop harvest. The samples were finely ground using a mill grinder. Approximately 0.5 g of the ground plant leaf samples were placed into digesting tubes, and 10 mL of concentrated nitric acid (HNO_3) and 3 mL perchloric acid ($HClO_4$) were added. The K^+ content was assessed using an atomic absorption spectrophotometer (TAS-986; Persee; China). For the estimation of Na^+ content, leaf samples (0.1 g) were extracted in 10 mL of distilled water by heating at 80°C for 3 h. The Na^+ content in the extracts was analysed using ion chromatography (DX-300; Sunnyvale, CA, USA) according to the method proposed by Munter *et al.* (1984).

Biochemical parameters

For all biochemical analyses, one fully expanded fresh leaf was collected after 30 days from each genotype and immediately preserved at -80°C until analysis.

Hydrogen peroxide

H_2O_2 content was estimated according to the method of Majeed et al., 2019. For the estimation of this trait, 1 g frozen leaf tissue was ground with 5 mL pre-chilled acetone and centrifuged at 3200 × g for 9 min at 4°C by using normal speed micro-centrifuge (SCILOGEX D2012). A NanoDrop Spectrophotometer (Model No. ND-8000 Thermo- Scientific) was used to find absorbance at 410 nm for estimation of H2O2 concentration by using a standard curve.

Proline

Proline content was determined by following the protocol proposed by Chaudhary *et al.* (2020). One gram of ground leaf tissue was homogenized with 5 mL of 3% sulfosalicylic acid. The extract was centrifuged at 11,000 rpm for 10 min, and the supernatant was transferred to microcentrifuge tubes. Separately, a 3% Ninhydrin solution containing equal volume of 6 M ortho-phosphoric acid and glacial acetic acid was prepared. Then, 1 mL from each component *i.e.*, glacial acetic acid, ninhydrin solution, and the supernatant of leaf extract were poured into cuvettes and incubated at 100°C for 50 min. Following incubation, the organic layer of each sample was transferred to an ELISA plate and absorbance was recorded at 520 nm by using toluene as a blank for the standard curve.

Peroxidase activity

The peroxidase activity was assessed by following the method reported by Kaur et al. 2024. The leaf tissues were ground in a mortar and pestle with 0.05 M sodium phosphate buffer, centrifuged at 10,000 rpm for 20 min, and the supernatant was transferred to a microcentrifuge tube. Then 3 mL of the reaction mixture was prepared by mixing equal amounts of guaicol and H_2O_2 which was then poured into the enzyme extract. Absorbance was measured at 470 nm using a Nano Drop Spectrophotometer (Model No. ND-8000 Thermo Scientific).

Catalase

Catalase activity was assayed according to the method described by Chance and Maehly (1955). Leaf tissues were ground with sodium phosphate buffer to prepare a 0.1 mL enzyme extract. The CAT reaction solution was then prepared by combining 40 mM phosphate buffer, 15 mM H_2O_2, and 0.1 mL of enzyme extract. Absorbance was recorded every 20 sec at 240 nm using a Nano Drop Spectrophotometer (Model No. ND-8000 Thermo Scientific).

Statistical Analysis

The analysis of variance was performed using STATISTIX 8.1 software. The estimation of genetic components (GCA and SCA) and heterosis was computed using OPSTAT software.

Results:

Analysis of variance for yield related traits in cotton under control and salt stress conditions

Analysis of variance was carried out for the screened plant material following Line × Tester design for yield related attributes under control and salt stress conditions. The mean squares from this analysis are presented in Table 1. Under control conditions, significant differences were observed for the studied traits. Mean squares due to GCA effects (parents), SCA effects, and lines x testers showed significant results for plant height, number of bolls, boll weight, and seed cotton yield. Lines differed significantly from each other for plant height, number of bolls, and seed cotton yield, and it was non-significant for boll weight. The analysis also revealed that testers are significantly different from each other for boll weight, seed cotton yield, and number of bolls and non-significant for plant height. The interaction of cross vs parent exhibited significant results for seed cotton yield, plant height, and boll weight, and non-significant for number of bolls under control conditions. The GCA effects were significant for fiber fineness, GOT, Na^+ concentration, K^+ concentration, and K^+/Na^+ ratio except fiber strength and fiber length. GCA effects

(parents) were significant for peroxidise dismutase, proline, catalase, and hydrogen peroxide. The analysis further revealed that the SCA effects (crosses) were significant for CAT, H_2O_2, and POD and non-significant for proline.

Under 10 dSm^{-1} level of salt stress, the analysis of variance indicated highly significant differences for the observed traits. Mean square values due to the GCA effects (parents) showed significant difference for number of bolls, plant height, seed cotton yield while boll weight showed non-significant differences. The interaction of Lines × Tester was found to be significant for plant height, seed cotton yield, boll weight, and number of bolls. The crosses due to specific combining ability were also found to be significant for the number of bolls, boll weight, plant height, and seed cotton yield. For plant height, number of bolls and seed cotton yield under 10 dSm^{-1} lines showed a significant contribution, whereas their contribution was non-significant for the remaining agronomic traits. Testers differed significantly from each other for all four agronomic traits. The interaction of cross vs parent was significant for all the observed traits except boll weight, which was non-significant. The Lines × Testers interaction revealed significant differences for fiber fineness, fiber length K^+ concentration and K^+/Na^+ ratio whereas it was non-significant for sodium. Similarly, the Lines × Testers interaction exhibited significant differences forPOD, CAT, and proline but it was non-significant for H_2O_2.

Under 15 dSm^{-1} salinity level, analysis of variance showed that there were significant differences among genotypes for the given traits. The interaction of Lines × Testers was found to be significant for plant height and boll weight, while it was non-significant for number of bolls and seed cotton yield. Mean square values due to GCA effect (parents) were found to be significant for number of bolls, whereas it was found non-significant for the seed cotton yield, boll weight, and plant height. The crosses due to specific combining ability showed highly significantly differences for number of bolls, seed cotton yield, boll weight, and plant height. Significant differences were observed among lines for plant height, number of bolls, and seed cotton yield whereas it boll weight was non significant under 15 dSm^{-1} salt stress. Testers were significantly different from each other for seed cotton yield, number of bolls, and plant height while non-significant for boll weight. The interaction of cross vs parents was found to be significantly different for plant height and boll weight, while non-significant for number of bolls and seed cotton yield. The SCA effects due to crosses were significantly different for all traits. The GCA effects for parents were significant for fiber fineness, fiber length, and ginning turn out, Na^+ concentration, and K^+ concentration, while non-significant for fiber strength and K^+/Na^+ ratio. Tester results showed significant differences for proline and non-significant for H_2O_2, CAT, and POD. The interaction of Lines × Testers exhibited significant differences for all the observed biochemical parameters.

Under 20 dSm^{-1}salt stress level, the analysis of variance indicated highly significant differences for the studied traits. Mean square values due to GCA effects (parents) indicated significant difference for seed cotton yield, number of bolls, plant height, and boll weight. The interaction of Lines × Tester was found to be significant for boll weight, plant height, seed cotton yield, and number of bolls. The crosses for SCA were found to be significant for seed cotton yield, number of bolls, boll weight, and plant height. GCA estimates for parents were significant for fiber fineness, fiber strength, fiber length, GOT, Na^+ concentration, K^+ concentration, and non-significant for K^+/Na^+ ratio. The interaction of line × tester revealed significant differences for fiber fineness, fiber length, fiber strength, Na^+ and K^+ concentration, K^+/Na^+ ratio and GOT. The analysis further showed that lines exhibited significant differences for proline, POD, CAT, and H_2O_2, whilst testers showed significant differences for proline and H_2O_2 and non-significant for CAT and POD. The interaction of lines × testers also exhibited significant differences for POD, proline, and H_2O_2 and non-significant for CAT.

Table 1. Mean square values from Line × Tester analysis for yield, fiber quality and biochemical traits under control and salt stress conditions.

SOV	Trt	DF	PH	N.B.	S.C.Y	B.W.	F.F.	F.S.	F.L.	GOT	Na^+	K^+	K^+/ Na^+	Proline	POD	CAT	H_2O_2
Rep	N	2	19.64	0.53	13.4	0.37	11.44	0.52	13.4	0.27	41.7**	381.4*	0.52	0.003	0.47	42.64	0.03
	S1	2	20.67	5.71*	11.92	5.92	0.03	0.53	0.61	12.73	21.61*	19.62*	0.44	0.28	18.47	12.84	0.056
	S2	2	46.54	0.82	54.45	0.35	0.07	2.53	13.4	0.37	22.62*	271.7**	0.18	0.47	3.29	12.76	0.038
	S3	2	20.67	0.58	14.2	0.31	0.09	3.22	3.34	0.83	38.02*	132.7**	0.22	0.45	3.58	12.85	0.037**
Gen	N	26	331.51**	8.54**	101.5**	0.41**	331**	8.54**	101.4**	0.43**	523.3**	649.4**	0.59**	0.008*	2.84**	103.6*	0.87*
	S1	26	347.21**	7.28**	26.41**	1.48**	0.37**	8.62**	7.06**	37.41**	10.62**	61.62**	0.78**	0.49**	24.28**	29.98	0.043**
	S2	26	326.47**	6.41**	51.56**	0.44**	0.72**	22.4**	101.45*	0.41**	193.45**	432.7**	0.49**	0.76**	13.29**	15.89*	0.067**
	S3	26	332.52**	8.64**	105.5**	0.48**	12.3**	4.83**	23.73**	3.73**	105.87**	323.8**	0.62**	0.56**	18.95**	42.87*	0.013**
Cross	N	17	244.27**	8.67**	62.48**	0.34**	28.27**	8.67	62.4**	0.74**	629.6**	826.7**	0.55**	0.006	2.86**	123.8*	0.78**
	S1	17	288.62**	11.6**	27.61**	1.92**	0.57**	14.2**	9.38**	35.09**	23.7*	123.5**	0.78**	0.58**	26.94**	78.49*	0.056**
	S2	17	261.31**	6.31**	65.45**	0.47**	2.56**	27.7**	62.8**	0.34**	281.4**	721.8**	0.15**	0.84**	16.49**	83.76*	0.076**
	S3	17	245.33*	8.56**	67.34	0.31*	7.93*	22.7*	7.38**		34.83*	243.62		0.67*	24.76	37.76	0.046

			*		**	*	*	*		0.83*		*	0.83*	*	**	*	**
Line	N	5	357.21**	5.37*	58.5*	0.12	51.1**	5.37*	58.5**	0.78**	19.5	921.3**	0.81**	0.002	3.78**	112.8*	0.69**
	S1	5	752.27**	8.45*	26.08**	0.11	1.59**	31.83*	19.29**	91.92**	4.7	271.4**	0.73**	0.76**	25.49**	173.5*	0.075**
	S2	5	487.57**	6.82*	63.12**	0.21	0.81**	15.03*	58.5**	0.12	451.6**	342.5**	0.52**	0.69**	21.84**	46.87*	0.008
	S3	5	365.27**	5.83*	59.28**	0.15	0.98**	14.9**	48.3**	0.66**	283.73**	322.8**	0.32**	0.52**	13.67**	57.87*	0.064**
Tester	N	2	76	17.1**	121.8**	0.56*	23.6	17.21**	121.2**	0.51*	482.3**	781.3**	0.31	0.004	0.148	237.6*	0.81**
	S1	2	124.45**	28.7**	42.32**	2.49*	0.25**	4.3**	6.28**	7.4	385.2**	672.6*	0.53	0.59**	21.94**	48.39*	0.059**
	S2	2	371.89**	23.4**	93.32**	0.29	0.61	14.26**	121.28**	54.56**	322.81**	828.6**	0.72**	0.77**	2.38	2.12	0.02
	S3	2	81	18.1**	125.2**	0.49*	0.12	22.56**	122.67**	32.73**	322.39**	434.8**	0.44**	0.49**	3.87	2.19	0.04*
L × T	N	10	216.19*	8.52**	64.31**	0.34**	18.11*	1.52	64.31**	0.13	532.4**	391.4**	1.52**	0.003	2.79**	101.7*	0.49*
	S1	10	176.51**	7.45**	27.38**	3.32*	0.2*	3.81**	5.07**	12.34**	13.62	502.3**	1.73**	0.57**	23.74**	72.05*	0.029
	S2	10	118.21*	0.43	59.02	0.76**	0.19*	8.52**	64.31**	0.34**	28.8**	671.4**	0.49**	0.84**	14.78*	12.74*	0.03*
	S3	10	218.22**	8.73**	61.42	0.37*	0.53*	6.92*	72.81**	0.73*	18.39*	432.4*	0.33*	0.58*	15.87		0.067**

					**	*		*		*		*	*	*	**	12.67	
Parent	N S1 S2 S3	8 8 8 8	526.22** 371.61** 5.21 532.23**	9.82** 1.36** 8.36** 9.36**	166.8** 151.5** 32.71 172.5**	0.42** 0.48 0.12 0.48**	25.02* 0.08** 0.22** 0.63**	1.23 1.47** 0.82 1.72**	16.28 1.79* 166.28** 177.82**	0.42** 41.45** 0.42** 0.83**	371.7** 33.76* 345.8** 228.3**	829.6** 707.2** 271.4** 523.2**	0.67** 0.09** 0.07 0.3	0.013** 0.47** 0.59** 0.46**	2.59* 22.74** 5.87 7.47	73.05* 28.94* 35.52 12.05	0.73** 0.018 0.07* 0.087**
Cross Vs Parent	N S1 S2 S3	1 1 1 1	22.34* 218.74* 143.54* 231.52*	2.56 8.21** 4.23 2.18	273.6** 29.45** 0.028 291.4**	1.49** 0.08 1.31** 1.38**	21.34* 0.15* 0.34* S3	2.56 7.09** 2.56 1.72	273.26** 2.71* 234.89** 211.82**	1.29** 6.12 1.49** 2.93**	472.5** 0.37 26.87** 33.81**	71.5 661.5** 685.5** 342.8**	0.26 3.89 0.61 0.73**	0.045* 0.12 0.12 0.12	0.06 25.84** 45.78** 49.39**	51.27 11.84* 15.78 11.63	0.12 0.78** 0.04* 0.079**
Error	N S1 S2 S3	52 52 52 52	89.52 37.21 113.67 91.54	1.74 1.61 1.52 1.87	12.23 6.28 17.42 13.28	0.11 0.12 0.16 0.12	88.52 0.04 89.52 3.22	1.74 1.72 1.74 1.82	12.23 0.92 12.23 1.88	0.11 4.61 0.11 0.66	14.87 36.7 29.53 18.39	18.39 34.2 17.34 13.82	0.28 0.63 0.03 0.08	0.005 0.26 0.43 0.37	1.16 17.39 4.57 4.65	23.36 10.08 11.07 14.98	0.34 0.008 0.004 0.004

Where N=control, S1=10 dSm-1, S2= 15 dSm-1, S3= 20 dSm-1, Rep= Replication, Gen= Genotype, L × T= Line × Tester, DF= Degree of freedom, Trt= Treatment, P.H.= Plant height, N.B.= Number of bolls, S.C.Y.= Seed cotton yield, B.W.= Boll weight, FF=Fiber fineness, F.S.=Fiber strength, F.L.=Fiber length, GOT=Ginning turnout, Na^+= Sodium concentration, K^+=Potassium concentration, K^+/Na^+=Potassium to sodium ratio *= significant, **= highly significant

Estimation of genetic components for yield, fiber, ionic and biochemical traits under control and various level of salt stress

Genetic components of variation for yield, fiber, ionic, and biochemical traits were assessed under control and various levels of salt stress, with results presented in Tables 2, 3, and 4, respectively. Under control conditions, the values of specific combining ability were found to be higher than those of GCA for plant height, number of bolls, boll weight, and seed cotton yield, indicating the preponderance of non-additive gene action. Variance due to SCA was also greater than GCA variance for fiber fineness, fiber strength, fiber length, GOT, Na^+ concentration, K^+ concentration, and K^+/Na^+ ratio. Additionally, variance due to SCA was higher for POD, CAT, and proline, suggesting that non-additive gene action plays a major role in the inheritance of these traits. The negative sign associated with H_2O_2 indicated that the direction of dominance is towards the lower value parent.

Under 10 dSm^{-1} salinity level, higher SCA values compared to GCA for the observed traits showed the preponderance of non-additive genes in trait expression. Dominance variance was positive and higher than additive variance for plant height, number of bolls, seed cotton yield, and boll weight. The SCA effects were also greater than GCA for fiber fineness, fiber strength, fiber length, GOT, Na^+ concentration, K^+ concentration, and K^+/Na^+ ratio. SCA was greater for proline, POD, and H_2O_2; However, GCA was greater for the CAT biochemical trait indicating larger additive type of gene action responsible for this trait.

Genetic components at the 15 dSm^{-1} salinity level revealed that the estimates for SCA were higher than those for GCA estimates. Additive variance was found to be positive for seed cotton yield, plant height, and number of bolls, while boll weight showed negative additive variance. All fiber and ionic traits exhibited greater SCA effects except fiber length and K^+/Na^+ ratio. SCA effects were also greater for proline, POD, and CAT indicating the dominant role of non-additive genes in controlling these traits. Furthermore, the analysis revealed that catalase had a greater value for dominance variance confirming the presence of dominant gene action.

Under the 20 dSm^{-1} level of salt stress, SCA was higher than GCA for all yield related traits suggesting the positive contribution of non-additive gene action to their expression. For fiber and ionic traits, SCA was generally higher, except for K^+ concentration and the K^+/Na^+ ratio, where GCA variance was higher. SCA effects showed greater values in all biochemical traits indicating the preponderance of non-additive gene action under the highest level of salt stress.

Estimation of proportional contribution of lines, testers, and their interaction under control and increasing levels of salt stress

The influence of lines, testers, and their interaction on the phenotypic expression of yield, fiber, ionic, as well as biochemical traits is presented in Tables 5, 6, and 7, respectively. Under control conditions, the contribution of lines was found to be maximum for plant height (42.73%) and seed cotton yield (24.82%). The contribution of testers was moderate across most observed parameters. Lines showed a major contribution to fiber fineness (62.49%), fiber strength (74.29%), fiber length (82.34%), GOT (72.41%), Na^+ concentration (52.47%), K^+ concentration (18.82%) and K^+/Na^+ ratio (48.23%). Among testers, the highest contribution was observed for K^+ concentration (12.49%). Line × tester interactions contributed most to the total variance for biochemical traits, including proline (63.48%), POD (58.48%), CAT (46.59%), and H_2O_2 (29.28%). For these traits, lines were a noticeably large contributor to the variance due to POD (42.59%) followed by H_2O_2 (27.28%), CAT (21.06%), and proline (7.38%).

Under the 10 dSm^{-1} level of salt stress, the line variation was the most for plant height (53.62%) followed by number of bolls (32.51%). The variance contribution of testers was highest for the number of bolls (29.02%). L×T variance was maximum for the biochemical traits including H_2O_2 (123.28%), proline (76.81%), POD (54.82%), and CAT (54.78%).

Under the 15 dSm^{-1} salt stress condition, the proportional contribution of lines was greatest towards plant height (63.72%), while the contribution towards the rest of the traits was low. The lines contributed the most for fiber related traits whereas the L×T interaction contributed the most for ionic and biochemical parameters. The lines contributed to fiber fineness (54.52%), fiber strength (64.62%), fiber length (62.35%), and GOT (65.38%). Variance due to L×T contributed the most to the biochemical traits including POD (98.38%), proline (78.98%), and CAT (23.48%).

Under the highest level of salt stress (20 dSm^{-1}), lines contributed the most towards plant height (72.91) and the least towards boll weight (38.08%). Testers contributed the maximum towards seed cotton yield (45.07%), while the minimum contribution was for plant height (16.63%). On the other hand, the contribution of line × tester interaction was maximum for plant height (53.61%) and minimum for seed cotton yield (26.71%). The lines contributed more to the total variance for fiber strength (64.39%), fiber fineness (54.82%), fiber length (61.29%), and GOT (67.29%). The L×T interaction had more variance than testers or lines for Na^+ concentration (73.29%), K^+ concentration (72.47%), and K^+/Na^+ ratio (79.47%). The contribution of L×T was maximum for all

biochemical parameters. The lines themselves contributed towards H_2O_2 (45.39%) followed by CAT (34.59%), POD (23.09%), and proline (6.24%).

Table 2 Estimation of genetic components of variations for yield under normal and salt stress conditions in upland cotton

	Control				10dSm^{-1}				15dSm^{-1}				20dSm^{-1}			
SOV	**GCA σ2**	**SCA σ2**	**Additive σ2**	**Dominance σ2**	**GCA σ2**	**SCA σ2**	**Additive σ2**	**Dominance σ2**	**GCA σ2**	**SCA σ2**	**Additive σ2**	**Dominance σ2**	**GCA σ2**	**SCA σ2**	**Additive σ2**	**Dominance σ2**
P.H.	0.76	45.34	1.36	45.34	5.03	16.38	7.51	16.38	4.51	53.21	7.51	53.21	5.81	62.31	9.18	62.31
N.B	0.016	3.04	0.05	3.04	0.03	0.73	0.31	0.73	0.6	1.65	0.11	1.65	0.93	2.04	0.37	2.04
S.C.Y.	0.3	14.63	0.3	14.62	0.12	12.63	0.27	12.63	0.07	5.72	0.16	5.72	1.56	7.62	0.21	7.62
B.W.	0.002	0.02	0.02	0.02	-0.34	0.4	-0.12	0.4	0.03	0.48	-0.04	0.48	-0.08	0.73	0.05	0.73

GCA=General combining ability, SCA=Specific combining ability, P.H.=Plant height, N.B.=Number of bolls, S.C.Y.=Seed cotton yield, B.W.=Boll weight

Table 3 Estimation of genetic components of variations for fiber and ionic traits under normal and 10dSm^{-1}, 15dSm^{-1}and 20dSm^{-1}salt stress

	Control				10dSm^{-1}				15dSm^{-1}				20dSm^{-1}			
SOV	**GCA σ2**	**SCA σ2**	**Additive σ2**	**Dominance σ2**	**GCA σ2**	**SCA σ2**	**Additive σ2**	**Dominance σ2**	**GCA σ2**	**SCA σ2**	**Additive σ2**	**Dominance σ2**	**GCA σ2**	**SCA σ2**	**Additive σ2**	**Dominance σ2**
F.L.	0.15	0.3	0.4	0.3	0.03	0.07	0.02	0.07	0.07	0.02	0.04	0.02	0.02	0.34	0.05	0.34
F.S.	0.28	1.27	1.32	1.27	0.14	0.26	0.37	0.26	0.15	0.59	0.31	0.59	0.38	0.47	0.72	0.47
F.F.	0.16	1.17	1.16	1.17	0.12	1.42	0.21	1.42	0.19	1.69	0.41	1.69	1.28	1.49	0.21	1.49
GOT	0.74	2.6	1.38	2.6	0.49	1.62	0.72	1.62	0.28	0.41	1.51	0.41	0.47	0.92	0.49	0.92
Na^+	0.27	15.42	2.7	15.42	0.73	16.38	1.67	16.38	0.17	23.71	0.48	23.71	0.51	1.72	1.72	1.72

K^+	1.62	156.3	14.38	156.3	4.71	214.71	9.51	214.71	7.29	312.4	1.67	312.4	34.81	13.8	14.23	13.8
K^+/Na^+	0.01	0.15	0.61	0.15	0.19	0.34	0.01	0.34	0.28	0.14	12.83	0.14	25.2	0.39	0.38	0.39

SOV=Source of variation, GCA=General combining ability, SCA=Specific combining ability, FF=Fiber fineness, F.S.=Fiber strength, F.L.=Fiber length, GOT=Ginning turnout, Na^+= Sodium concentration ,K^+=Potassium concentration, K^+/Na^+=Potassium to sodium ratio

Table 4 Estimationof genetic components of variations of biochemical traits under normal, 10dSm^{-1},15 dSm^{-1} and 20 dSm^{-1} salt stress.

	Control				**10dSm^{-1}**				**15dSm^{-1}**				**20dSm^{-1}**			
SOV	**GCA σ2**	**SCA σ2**	**Additive σ2**	**Dominance σ2**	**GCA σ2**	**SCA σ2**	**Additive σ2**	**Dominance σ2**	**GCA σ2**	**SCA σ2**	**Additive σ2**	**Dominance σ2**	**GCA σ2**	**SCA σ2**	**Additive σ2**	**Dominance σ2**
Proline	0.23	3.43	23.52	3.43	1.09	23.52	3.56	23.52	12.34	25.56	3.09	25.56	2.04	3.01	12.45	3.01
POD	0.11	7.76	0.45	7.76	0.04	0.12	0.034	0.12	0.32	8.87	0.48	8.87	2.09	12.48	3.09	12.48
CAT	2.45	13.56	23.12	13.56	32.45	2.19	12.98	2.19	3.98	15.87	11.09	15.87	0.89	45.87	2.34	45.87
H_2O_2	0.12	-0.11	0.54	0.03	0.01	0.03	0.09	0.02	0.03	0.01	0.01	0.01	0.03	0.03	0.11	0.03

GCA=General combining ability, SCA=Specific combining ability, POD=Peroxidase, CAT=Catalase, H_2O_2=Hydrogen peroxide

Table 5 Proportional contribution of lines, testers and their interaction to total variance for yield traits under normal and various levels of salt stress

	Control			**10dSm^{-1}**			**15dSm^{-1}**			**20dSm^{-1}**		
Trait	**Line**	**Testers**	**L×T**	**Line**	**Testers**	**L×T**	**Line**	**Testers**	**L×T**	**Line**	**Testers**	**L×T**
P.H.	42.73	4.62	57.42	53.62	15.05	29.05	63.72	5.29	33.72	72.91	16.63	53.61
N.B.	17.62	25.61	51.83	32.51	29.02	34.61	21.83	28.63	42.61	41.63	29.06	41.06

S.C.Y.	24.82	22.19	52.03	28.39	16.28	49.05	25.52	17.63	53.07	51.62	45.07	26.71
B.W.	15.62	16.42	67.19	14.62	7.92	76.66	5.29	16.62	75.39	38.08	39.71	47.06

P.H.= Plant height, N.B.= Number of bolls, S.C.Y.= Seed cotton yield, B.W.= Boll weight

Table 6 Proportional contribution of lines, testers and their interaction to total variance for fiber and ionic related traits under normal and various levels of salt stress.

	Control			**10dSm^{-1}**			**15dSm^{-1}**			**20dSm^{-1}**		
Trait	**Line**	**Testers**	**L×T**	**Line**	**Testers**	**L×T**	**Line**	**Testers**	**L×T**	**Line**	**Testers**	**L×T**
F.L.	82.34	4.39	12.42	65.39	6.37	25.82	62.35	12.48	24.73	61.29	16.28	24.71
F.S.	74.29	6.82	17.28	76.27	7.38	21.39	64.62	8.45	21.29	64.39	8.41	22.88
F.F.	62.49	7.34	26.39	54.29	6.23	31.83	54.52	13.28	31.47	54.82	12.37	30.61
GOT	72.41	1.64	21.39	47.32	8.28	82.45	65.38	1.56	82.19	67.29	19.94	25.39
Na^+	52.47	7.39	37.41	31.09	1.06	72.38	16.28	6.39	74.28	17.29	2.38	73.29
K^+	18.82	12.49	63.42	63.43	11.48	36.45	13.56	7.28	72.29	14.29	9.31	72.47
K^+/Na^+	48.23	8.39	41.37	41.83	4.19	85.38	11.38	5.04	74.29	11.37	8.31	79.47

FF= Fiber fineness, F.S.= Fiber strength, F.L.= Fiber length, GOT=Ginning turn out, Na^+= Sodium concentration, K^+= Potassium concentration, K^+/Na^+= Potassium to sodium ratio

Table 7 Proportional contribution of lines, testers and their interaction to total variance for biochemical traits under normal and various levels of salt stress.

	Control			10dSm^{-1}			15dSm^{-1}			20dSm^{-1}		
Trait	**Line**	**Testers**	**L×T**	**Line**	**Testers**	**L×T**	**Line**	**Testers**	**L×T**	**Line**	**Testers**	**L×T**
Proline	7.38	24.39	63.48	21.58	1.87	76.81	9.39	4.59	78.98	6.24	5.08	83.29
POD	42.59	8.76	58.48	46.49	4.29	54.82	1.09	34.78	98.38	23.09	14.74	76.48
CAT	21.06	22.78	46.59	19.45	37.83	54.78	63.29	0.67	23.48	34.59	14.39	38.58
H2O2	27.28	0.38	29.28	56.39	11.83	123.28	1.05	0.49	1.39	45.39	20.05	89.48

L×T=Line ×Tester, POD=Peroxidase, CAT=Catalase,H2O2= Hydrogen peroxide

Heterosis effects for yield, fiber, ionic, as well as biochemical parameters under control and salt stress conditions

Under control conditions, the combinations NIAB-545×Cyto-178, CIM-595×Cyto-178, FH-113×Cyto-178, FH-113×FH-326, and FH-942×FH-326 showed positive and significant heterosis for plant height (Table 8). CIM-595×FH-326, FH-113×Cyto-178, FH-942×Cyto-178, DNH-40×VH-363 and DNH-40×FH-326 displayed significant positive heterosis estimates for the number of bolls per plant. Seed cotton yield is the most important attribute in any cotton breeding program. Coker-307×FH-326, FH-113×FH-326, FH-942×Cyto-178, and FH-942×FH-326 exhibited significant and positive values for seed cotton yield heterosis. FH-113×FH-326 and DNH-40×Cyto-178 displayed positive and significant heterosis for boll weight, whereas, NIAB-545×FH-326 showed significant and negative heterosis. The hybrids NIAB-545×Cyto-178, NIAB-545×VH-363, NIAB-545×FH-326, CIM-595×Cyto-178, CIM-595×VH-363, CIM-595×FH-326, and Coker-307×VH-363 exhibited positive and significant heterosis estimates for fiber fineness. The hybrids NIAB-545×VH-363, NIAB-545×FH-326, CIM-595×VH-363, Coker-307×Cyto-178, FH-942×VH-363, and FH-942×FH-326 indicated positive and significant estimate for heterosis for fiber strength. Coker-307×VH-363, FH-942×VH-363, DNH-40×Cyto-178, DNH-40×FH-326, CIM-595×FH-326, and FH-113×Cyto-178 indicated positive and significant estimates of heterosis for fiber length. The hybrids NIAB-545×FH-326, CIM-595×Cyto-178, and CIM-595×VH-363 displayed positive and significant heterosis for ginning turn out. Coker-307×VH-363 and Coker-307×FH-326 manifested positive and significant heterosis for sodium concentration. Under normal conditions, Coker-307×Cyto-178, FH-113×Cyto-178, and FH-942×Cyto-178 displayed positive and significant heterosis for H_2O_2. NIAB-545×Cyto-178, CIM-595×VH-363 and NIAB-545×FH-326, FH-942×Cyto-178 and DNH-40×Cyto-178 revealed positive and significant heterosis for POD. CIM-595×Cyto-178, CIM-595×VH-363, FH-942×FH-326, DNH-40×Cyto-178, and Coker-307×FH-326 displayed positive and significant heterosis for proline.

Under 10 dSm^{-1} salt stress, FH-942×FH-326 displayed positive and significant heterosis for plant height. Only cross FH-942×FH-326 showed significant and positive heterosis for seed cotton yield. FH-113×FH-326 displayed positive and significant heterosis effects for boll weight. The hybrid FH-942×VH-363 displayed positive and significant heterosis for fiber length, while, CIM-595×Cyto-178, CIM-595×VH-363, CIM-595×FH-326 showed positive and significant heterosis for fiber fineness. NIAB-545×VH-363 displayed positive and significant heterosis for fiber strength. NIAB-545×VH-363, CIM-595×Cyto-178, and DNH-40×VH-363 showed positive and significant heterosis for ginning turn out. FH-942×Cyto-178 displayed positive and significant heterosis under 10 dSm^{-1} for POD. None of the crosses showed significant and positive heterosis for H_2O_2.

Under 15 dSm^{-1} salt stress, the combinations DNH-40×Cyto-178 and DNH-40×Cyto-178 exhibited positive and significant heterosis for plant height, whereas NIAB-545×VH-363 showed negative and significant heterosis. CIM-595×Cyto-178 manifested positive heterosis for number of bolls per plant. DNH-40×Cyto178 and DNH-40×VH-363 displayed positive and significant heterosis, whereas no other cross combination showed significant heterosis for seed cotton yield. The analysis further revealed that Coker-307×FH-326, displayed positive and significant heterosis for boll weight. The heterotic analysis further revealed that the hybrid FH-113×VH-363 displayed positive and significant heterosis for fiber fineness. The results of heterotic analysis further revealed that none of the crosses exhibited positive and significant heterosis for fiber strength and fiber length. CIM-595×FH-326 and FH-113×VH-363 showed positive and significant estimates for heterosis for ginning turn out. None of the crosses showed positive and significant heterosis for catalase.

Under 20 dSm^{-1} salt stress, none of the crosses manifested positive and significant heterosis for plant height. DNH-40×Cyto-178 exhibited significantly positive heterosis estimates for number of bolls per plant. NIAB-545×FH-326 exhibited positive and significant heterosis for seed cotton yield. FH-113×FH-326 showed significantly positive heterosis for boll weight. FH-113×FH-326 displayed positive and significant heterosis for fiber fineness. The hybrids NIAB-545×VH-363 and NIAB-545×FH-326 showed significantly positive estimates for heterosis for fiber strength. FH-113×FH-326 showed significant and positive heterosis for fiber length. NIAB-545×Cyto-178 and FH-942×Cyto-178 manifested positive and significant heterosis for ginning turn out. No significant heterosis was observed for ionic nor biochemical traits under the highest level of salt stress.

Table 8 Mid parent-heterosis effects of yield, fiber and ionic as well as biochemical traits under control and salt stress conditions

Crosses	Trt	PH	N.B	S.C.Y.	B.W	FL	FS	FF	GOT	Na+	K+	K+/Na+	Proline	POD	CAT	H_2O_2
NIAB-545×Cyto-178	N	18.38*	12.56	-5.38	21.7*	3.99	3.69	4.38*	-9.2	3.19	14.48	38.9**	-18.38	78.4*	56.4*	-6.38
	S1	6.33	28.38	14.38	11.78	7.37	6.37	-4.3*	-15.7*	6.37	3.29	23.29	-2.48	19.13	-5.38	14.38
	S2	-8.29	12.48	24.39	-4.39	-9.34	-2.34	3.02	3.08	-2.34	22.04	12.53	-6.39	5.62	1.38	24.39
	S3	-7.39	3.07	7.49	-4.30	-4.39	-4.39	2.18	11.8*	-4.39	-4.39	-4.39	-16.49	-3.51	14.32	7.49
NIAB-545×VH-363	N	-4.39	4.39	-23.28	-14.47	16.48	33.7**	12**	-9.39	56.4	7.43	-18.38	24.2	-11.2	38.9*	22.05
	S1	-7.39	6.49	-4.28	4.31	3.29	29.1*	2.11	14.5*	-5.38	14.3	-2.48	9.11	0.67	23.29	-21.94
	S2	-11.7*	-13.38	-12	2.09	22.04	12.34	0.38	0.48	1.38	12.1	-6.39	-3.19	2.09	12.53	-5.49
	S3	6.31	1.38	-15.39	-1.79	-4.39	34.16*	-1.38	-9.3**	14.3	1.39	-16.49	-4.50	-2.19	-4.39	-2.58
NIAB-545×FH-326	N	7.43	-42.4*	-45.6**	-67.8**	-12.51	94.8**	8.38**	56.4*	7.4	3.29	13.28	-7.38	67.**	37.2*	7.43
	S1	14.3	-8.38	-34.2**	-5.38	11.49	23.29	-3.78*	-5.38	14	6.37	-11.48	14.38	-5.38	28.4*	14.3
	S2	12.19	-9.59	12.78	23.22	5.51	3.29	1.33	1.38	1.1	-2.34	5.09	24.39	23.22	15.34	12.19
	S3	1.39	-6.58	38.4**	-13.28	-9.3	34.39*	3.89	14.32	1.3	-4.39	6.48	7.49	13.28	14.56	1.39

CIM-595×Cyto-178	N S1 S2 S3	23.9* 2.19 1.38 2.18	4.38 21.55 34.14* 21.48	16.39 14.38 15.39 -14.39	5.19 4.21 -1.34 -4.51	-11.27 0.67 2.09 -2.19	-10.48 12.29 3.29 -11.3	16** 8.39** 4.23 -2.48	37.2* 28.4* 15.34 14.56	16.48 3.29 22.04 -4.39	3.29 6.37 2.34 4.39	6.19 12.1 15.37 1.89	53.4** -5.38 1.38 14.32	11.48 12.89 3.29 -11.3	16.48 3.29 22.04 -4.39	4.19 6.59 -1.84 -5.49
CIM-595×VH-363	N S1 S2 S3	-16.71 12.14 12.24 10.07	-14.29 12.48 15.38 11.47	-18.38 -2.48 -6.39 -16.49	12.48 3.29 22.04 -4.39	16.48 3.29 22.04 -4.39	21.7* 11.78 -4.39 -4.30	14.8** 9.38** 2.14 -4.39	38.9* 23.29 12.53 -4.39	-6.38 14.3 24.3 7.49	8.23 14.3 12.1 1.39	38.9** 23.29 12.53 -4.39	7.43 14.3 12.19 1.39	56.4* -5.38 1.38 14.32	3.29 6.37 -2.34 -4.39	16.48 3.29 22.04 -4.39
CIM-595×FH-326	N S1 S2 S3	-11.48 12.89 3.29 -11.3	38.9** 23.29 12.53 -4.39	14.39 -14.34 -3.48 -13.28	14.2 12.3 15.3 11.4	2.4 6.2 12.3 1.3	8.82 3.29 2.71 3.8	15.8** 5.49** 0.29 -7.59*	4.89 -7.38* 13.28* 2.16	14.2 12.3 15.3 11.4	21.7* 11.78 -4.39 -4.30	37.2** 28.4* 15.34 14.56	22.05 -21.94 -5.49 -2.58	21.7 11.78 -4.39 -4.30	7.43 14.3 12.19 1.39	13.28 -11.48 5.09 6.48
Coker-307×Cyto-178	N S1	-11.27 -13.28	5.39 7.37	-17.08 12.29	37.9* 23.29	-11.27 0.67	24.7* 13.78	2.4 6.2	12.38 12.89	14.48 3.29	14.2* 12.3	16.48 13.18	-15.28 -2.18	22.05	-11.27 0.67	38.9** 23.29

	S2 S3	4.29 1.78	-2.31 -5.29	2.19 -11.3	12.53 -4.39	2.09 -2.19	-4.39 -4.30	12.3 3.2	3.29 -11.3	22.04 -4.39	15.3* 11.4	-4.33 -1.70	-5.69 -13.29	21.94 -5.49 -2.58	2.09 -2.19	12.53 -4.39
Coker-307×VH-363	N S1 S2 S3	-20.2* -13.28 11.38 -9.49	-15.38 11.28 -21.08 -5.49	-11.32 32.3 -3.43 2.11	5.19 6.27 -3.31 -2.31	66.8** -5.38 24.22 -13.18	-10.48 12.29 3.29 -11.3	58.1** -4.18 21.72 -13.28	12.48 15.38 11.47 5.39	37.2** 28.4* 15.34 14.56	6.53 14.3 12.1 1.39	7.43 14.3 12.19 1.39	3.29 6.37 -2.34 -4.39	3.29 6.37 -2.34 -4.39	38.9* 23.29 12.53 -4.39	-11.27 0.67 2.09 -2.19
Coker-307×FH-326	N S1 S2 S3	5.41 5.39 -4.78 3.82	16.49 -5.38 23.22 -13.34	51.6** -4.15 24.02 -8.28	4.89* -7.38* 13.28* 2.16	-11.27 0.67 2.09 -2.19	16.48 3.29 22.04 -4.39	-11.27 0.67 2.09 -2.19	7.43 14.3 12.19 1.39	25.4** 17..1* 11.34 11.06	3.29 6.37 2.3 4.39	-16.18 14.89 3.19 -12.8	41.7** -9.48 3.72 13.35	7.43 14.3 12.19 1.39	94.8** 23.29 3.29 -4.39	3.29 6.37 -2.34 -4.39
FH-113×Cyto-178	N S1 S2 S3	21.7* 11.78 -4.39 -4.30	53.5** 20.06 -12.48 14.39	13.28 20.06 -12.56 14.39	14.2 12.3 15.3 11.4	25.1* 16.12 -5.31 -6.52	3.29 6.37 -2.34 -4.39	67.8** -6.31 26.12 -11.58	35.1 27.6 12.04 9.39	23.7 15.78 -4.39 -4.30	19.7* 13.18 -5.29 -1.38	22.05 -21.94 -5.49 -2.58	7.43 14.3 12.19 1.39	21.37 22.05 21.28	-6.38 14.38 24.39 7.49	56.4** -5.38 1.38 14.32

														-5.49		
FH-113×VH-363	N S1 S2 S3	-7.49 13.28 -7.59 -11.45	22.05 -21.94 -5.49 -2.58	21.37 22.05 -21.28 -5.49	13.28 11.48 5.09 6.48	15.48 3.29 22.24 -4.59	7.4 14 1.1 1.3	4.89* -7.38* 13.28* 2.16	-6.38 14.38 24.39* 7.49	16.1 13.1 15.7 12.4	7.41 6.27 -2.14 -2.35	11.78 15.38 14.37 6.32	-16.42 11.59 4.09 -15.2	16.48 3.29 22.04 -4.39	38.9* 23.29 12.53 -4.39	3.29 6.37 -2.34 -4.39
FH-113×FH-326	N S1 S2 S3	34.8** 11.45 -11.45 -12.45	12.48 15.38 11.47 5.39	72.3** 12.48 15.38 11.47	33.2** 28.4* 13.34 41.67*	2.4 6.2 12.3 28.76*	24.7* 11.78 -4.39 -4.30	2.4 6.2 12.3 19.73*	0.41 1.67 1.23 2.77	-15.38 11.28 -21.08 -5.49	3.43 24.3 11.1 1.39	7.29 6.37 -2.34 -4.39	22.05 -21.94 -5.49 -2.58	18.72 13.29 -6.36 -2.52	37.2* 28.4* 15.34 14.56	16.48 3.29 22.04 -4.39
FH-942×Cyto-178	N S1 S2 S3	11.34 -16.34 -12.45 13.28	23.29* 3.28 -4.39 6.49	94.8** 23.29 3.29 -4.39	45.8** 32.29 3.19 -5.39	-12.18 17.19 3.19 -10.3	14.48 3.29 22.04 -4.39	12.48 3.29 21.04 -4.37	2.11 11.25 3.48 31.28*	-11.27 0.67 2.09 -2.19	-17.21 0.47 3.09 -3.19	3.29 6.37 -2.34 -4.39	13.28 -11.48 5.09 6.48	45.6* 34.2* 12.78	16.48 3.29 22.04 -4.39	24.8* 14.71 -5.31 -4.20

														38.4*		
FH-942×VH-363	N S1 S2 S3	12.22 -9.48 5.69 8.78	9.23 6.75 -4.33 7.81	16.48 3.29 22.04 -4.39	3.29 6.37 -2.34 -4.39	37.2** 28.4* 15.34 14.56	22.05 -21.94 -5.49 -2.58	21.7* 11.78 -4.39 -4.30	4.49 6.67 -5.12 -6.37	-34.27 0.87 2.19 -2.19	5.29 6.37 -2.34 -4.39	-16.38 -2.78 -7.31 -12.49	-14.18 11.11 4.89 -9.6	3.19 6.37 -2.34 -4.39	-11.27 0.67 2.09 -2.19	-15.38 11.28 -21.08 -5.49
FH-942×FH-326	N S1 S2 S3	31.6** 23.3* 15.11 12.51	11.78 -4.39 -8.04 12.71	37.3** 28.5** 15.34 14.56	21.7* 11.78 -4.39 -4.30	13.43 3.29 22.04 -4.29	3.99 7.37 -9.34 -4.39	45.8** 32.29 3.19 -5.39	6.4 -5.38 1.38 14.32	16.48 3.29 22.04 -4.39	21.7* 11.78 -4.39 -4.30	35.1** 22.4* 15.34 14.56	56.4** -5.38 1.38 14.32	27.37 22.05 21.28 -5.49	3.29 6.37 -2.34 -4.39	18.38 -2.48 -6.39 -16.49
DNH-40×Cyto-178	N S1 S2 S3	-14.38 -3.38 21.4* 12.46	11.78 16.38 3.07 34.1**	-13.79 -2.69 18.3* 12.46	67.8** -5.38 23.22 -13.28	38.9** 23.29 12.53 -4.39	-15.38 11.28 -21.08 -5.49	-11.27 0.67 2.09 -2.19	12.48 15.38 11.47 5.39	3.19 6.87 -2.34 -4.39	19.7* 13.78 -4.39 -4.30	-10.48 12.29 3.29 -11.3	38.9** 23.29 12.53 -4.39	94.8** 23.29 3.29 -4.39	25.7* 14.78 -4.39 -6.30	6.8 -5.38 23.22 -13.28

DNH-40×VH-363	N S1 S2 S3	-17.6* 12.36 23.6* 14.26	53.4** -5.38 1.38 14.32	-15.6* 12.36 23.6* 14.26	-12.48 2.61 5.23 -9.1	3.29 6.37 -2.34 -4.39	62.9** -5.38 21.37 -13.28	3.29 6.37 -2.34 -4.39	2.22 7.61* -4.13 -17.59	-14.42 9.21 5.61 -10.7	-11.27 0.67 2.09 -2.19	56.4** -5.38 1.38 14.32	16.48 3.29 22.04 -4.39	3.99 7.37 -9.34 -4.39	37.2* 28.4* 15.34 14.56	16.48 3.29 22.04 -4.39
DNH-40×FH-326	N S1 S2 S3	-12.27 0.67 2.09 -2.19	69.8** -5.38 17.12 -12.08	-11.27 0.67 2.09 -2.19	4.19 5.37 -2.14 -3.43	27.7* 12.78 -4.39 -4.30	16.48 3.29 22.04 -4.39	3.2 6.37. 34.9 5.98	1.11 2.11 4.39 4.44	21.7 11.78 -4.39 -4.30	37.2** 28.4* 15.34 14.56	12.18 12.36 8.41 6.29	4.29 7.55 -2.28 -5.33	19.3 14.18 -4.76 -5.64	22.37 27.05 -21.28 -5.49	-11.27 0.67 2.09 -2.19

N=control, S1=10 dSm-1, S2= 15 dSm-1, S3= 20 dSm-1, Trt= Treatment, P.H.= Plant height, N.B.= Number of bolls, S.C.Y.= Seed cotton yield, B.W.= Boll weight, FF= Fiber fineness, F.S.=Fiber strength, F.L.=Fiber length, GOT=Ginning turn out, Na^+= Sodium concentration, K^+=Potassium concentration, K^+/Na^+=Potassium to sodium ratio POD= peroxidase and CAT = Catalase

General and specific combining ability estimations for all studied traits under control and various levels of salinity

Plant height is an important parameter in cotton which is greatly reduced under salt stress. Under control conditions, the genotypes FH-942 (12.63), FH-113 (4.71), and NIAB-545 (0.5) showed positive GCA effects for plant height whereas Coker-307 exhibited negative GCA value among female parents (Table 10). Specific combining ability estimates for FH-942×VH-363 (14.61) and FH-942×FH-326 (16.82) were positive and significant for plant height, whereas negative and significant estimates were observed for CIM-595×FH-326 (-13.83) (Table 9). Among lines, FH-942 (1.93) showed significant positive values and among testers FH-326 (1.31) showed significant positive value for GCA effects when measured for number of bolls. FH-942 (4.98) displayed significantly positive GCA estimates for seed cotton yield among female parents, whilst among male parents, FH-326 (3.07) showed positive and significant GCA effects. For boll weight, none of the female parents showed significant GCA effects, while among male parents, VH-363(0.3) exhibited significantly positive results. In addition, under 10 and 15 dSm^{-1} levels of salt stress, no female or male parents displayed significant GCA estimates for boll weight. Under control conditions, the genotypes NIAB-545 (0.43) and CIM-595 (0.51) exhibited positive and significant GCA effects for fiber length. While among the male parents, Cyto-178 (0.16) displayed positive and significant GCA effects. Under normal conditions, fiber strength of the female parents NIAB-545 (1.71), CIM-595 (1.79), and FH 113(0.54) exhibited significantly positive GCA estimates, whereas among testers, Cyto-178(0.42) also revealed significantly positive GCA effects. Coker-307×Cyto-178 (1.59) and FH-942×FH-326 (0.99) hybrids showed positive and significant specific combining ability effects for fiber length. Under normal conditions, the female parents NIAB-545 (1.71), CIM-595 (1.79), and FH 113(0.54) exhibited significantly positive GCA estimates and the testers Cyto-178 (0.16) revealed significantly positive GCA effects for fiber strength. Under the 15 dSm^{-1} level of salt stress, Coker-307×FH-326 (1.43) revealed positive and significant SCA effects for fiber strength. Among female parents, NIAB-545 (0.76), CIM-595 (0.64), FH-942 (0.71) and DNH-40 (0.93) showed positive and significant estimates under control conditions for fiber fineness. GOT is an important trait with relation to lint production. Under normal condition, amongst the female parents, NIAB-545 (2.81) and CIM-595 (6.08) indicated positive and significant GCA effects for GOT while none of the male parents exhibited desirable SCA effects. Under normal, 10 dSm^{-1} and 15 dSm^{-1} salt stress, and among male and female parents, none of the parents indicated positive and significant GCA effects. Under normal conditions, female parents Coker-307 (6.49), FH-942 (5.39), and DNH-40(4.29) showed significantly positive GCA effects for K^+ concentration, whilst among male parents only, FH-326 (6.38) showed positive and significant GCA effects. Under normal conditions, none of the parents showed positive and significant GCA effects for the ratio of potassium to sodium. However, for this trait under all salt stress levels, Coker-307 (0.24, 0.24, 0.34, respectively) had positive and significant GCA estimates and FH-326 (0.13) was significant at 15 dSm^{-1}salt stress. Under normal conditions, among female parents NIAB-545 (0.23) manifested positive and significant results, whilst among testers VH-363 (-0.22) showed negative and significant GCA for proline. Among the crosses, FH-942×Cyto-178 showed positive and significant SCA effects, whereas NIAB-545×FH-326, NIAB-545×FH-326, and FH-113×FH-326 exhibited negative and significant SCA values for proline. Under normal conditions, Coker-307 (3.27) indicated positive and significant values among lines, whilst Cyto-178 (1.48) showed positive and significant results for peroxidise. Under 20 dSm^{-1} salinity level, Coker-307×VH-363, Coker-307×FH-326, FH-113×FH-326, FH-942×Cyto-178, and FH-942×FH 326 exhibited positive and significant estimates of SCA effects for biochemical parameters. However, under 15 dSm^{-1} salt stress level, DNH-40 (6.39) among females showed positive and significant results while no male parent exhibited significant GCA ability for catalase. When evaluating crosses, none of the crosses exhibited positive response with relation to SCA effects for H_2O_2 under control, 10 dSm^{-1} 15 dSm^{-1}, and 20 dSm^{-1} salt stress conditions.

Table 9 Specific combining ability effects of crosses for different traits under normal and various levels of salt stress

Crosses	Trt	PH	N.B	S.C.Y.	B.W	FL	FS	FF	GOT	Na+	K+	$K^+/Na+$	Proline	POD	CAT	H_2O_2
NIAB-545×Cyto-178	N	14.48	-9.2*	3.69	3.19	14.48	56.4*	12.56	-6.38	94.8*	21.7*	38.9**	3.99	3.19	-5.38	18.38*
	S1	3.29	-15.7*	6.37	6.37	3.29	-5.38	28.38	14.38	23.29	11.78	23.29	7.37	6.37	14.38	6.33
	S2	22.04	3.08	-2.34	-2.34	22.04	1.38	12.48	24.39	3.29	-4.39	12.53	-9.34	-2.34	24.39	-8.29
	S3	-4.39	11.8*	-4.39	-4.39	-4.39	14.32	3.07	7.49	-4.39	-4.30	-4.39	-4.39	-4.39	7.49	-7.39
NIAB-545×VH-363	N	7.43	-9.39*	37.2**	56.4	7.43	38.9*	4.39	22.05	-11.2	-11.27	-18.38	16.48	56.4	-23.28	-4.39
	S1	14.3	14.5*	28.4*	-5.38	14.3	23.29	6.49	-21.94	0.67	0.67	-2.48	3.29	-5.38	-4.28	-7.39
	S2	12.1	0.48	15.34	1.38	12.1	12.53	-13.38	-5.49	2.09	2.09	-6.39	22.04	1.38	-12	-11.7*
	S3	1.39	-9.3**	14.56	14.3	1.39	-4.39	1.38	-2.58	-2.19	-2.19	-16.49	-4.39	14.3	-15.39	6.31
NIAB-545×FH-326	N	4.72	56.4*	94.8**	7.23	4.73	37.2*	-42.4*	7.43	67.34**	67.8**	13.28	-10.48	8.39	-45.6**	7.43
	S1	7.17	-5.38	23.29	14.83	6.37	28.4*	-8.38	14.3	-4.72	-5.38	-11.48	12.29	13.61	-34.2**	14.3
	S2	-2.34	1.38	3.29	1.17	-2.34	15.34	-9.59	12.19	23.22	26.38	5.09	3.29	3.27	12.78	12.19
	S3	-4.39	14.32	-4.39	1.43	-4.39	14.56	-6.58	1.39	12.28	-13.28	6.48	-11.3	1.83	38.4**	1.39
CIM-595× Cyto-178	N	9.69	37.2*	-10.48	16.48	4.29	16.48	4.38	3.29	11.48	5.73	7.43	-11.27	16.48	16.39	23.9*
	S1	6.57	28.4*	12.29	3.29	6.37	3.29	21.55	6.37	12.89	4.17	14.3	0.67	3.29	14.38	2.19
	S2	2.34	15.34	3.29	22.04	2.34	22.04	8.39	-2.34	3.29	-3.34	12.19	2.09	22.04	15.39	1.38

	S3	3.32	14.56	-11.3	-4.39	4.39	-4.39	21.48	-4.39	-11.3	-4.78	1.39	-2.19	-4.39	-14.39	2.18
CIM-595×VH-363	N	7.43	38.9*	21.7*	-7.38	8.23	5.39	-14.29	13.38	56.4*	12.48	36.28**	15.29	-6.38	-18.38	-14.29
	S1	14.3	23.29	11.78	12.34	14.3	6.27	12.48	3.29	-5.38	3.29	23.29	3.29	14.38	-2.48	12.34
	S2	12.1	12.53	-4.39	21.36	12.1	-2.14	15.38	22.04	1.38	22.04	12.53	22.04	24.32	-6.39	15.38
	S3	1.39	-4.39	-4.30	7.49	1.39	-4.39	11.47	-4.39	14.32	-4.39	-4.39	-4.39	7.49	-16.49	11.47
CIM-595×FH-326	N	21.7*	4.89*	8.93	16.38	21.7*	7.43	38.9**	13.28	21.7*	13.22	36.23**	2.47	14.26	14.39	-11.48
	S1	11.78	-7.38*	14.52	11.37	11.78	14.37	23.29	-11.48	11.78	11.36	28.41*	6.27	12.32	-14.34	12.89
	S2	-4.39	13.28*	1.12	14.32	-4.39	12.19	12.53	5.09	-4.39	15.36	15.34	12.32	15.37	-3.48	3.29
	S3	-4.30	2.16	1.39	10.48	-4.30	1.39	-4.39	6.48	-4.30	11.41	14.56	1.38	11.41	-13.28	-11.3
Coker-307× Cyto-178	N	13.26*	12.38	24.7*	14.48	14.28*	-11.27	3.29	38.9**	22.05	37.9*	21.7*	-11.27	14.48	-11.48	-11.27
	S1	11.37	12.89	13.78	3.29	12.31	0.67	6.37	23.29	21.94	23.29	11.78	0.67	3.29	12.89	-13.28
	S2	14.39*	3.29	-4.39	22.04	15.39*	2.09	-2.34	12.53	-5.49	12.53	-4.39	2.09	22.04	3.29	4.29
	S3	14.48	-11.3	-4.30	-4.39	11.46	-2.19	-4.39	-4.39	-2.58	-4.39	-4.30	-2.19	-4.39	-11.3	1.78
Coker-307× VH-363	N	7.43	12.48	-11.63	37.2**	6.53	38.9*	-15.38	-12.18	3.29	3.29	7.43	62.1**	37.2**	-11.32	-20.2*
	S1	14.32	15.38	12.24	28.4*	14.36	23.29	11.28	0.67	6.37	6.37	14.37	-5.38	28.4*	32.3	-13.28
	S2	12.17	11.47	4.29	15.34	12.13	12.53	-21.08	2.09	-2.34	-2.34	12.19	24.22	15.34	-3.43	11.38
	S3	1.39	5.39	-16.3	14.56	1.39	-4.39	-5.49	-2.19	-4.39	-4.39	1.39	-13.18	14.56	2.11	-9.49
Coker-307× FH-326	N	4.62	7.43	17.29	37.2**	4.19	92.47**	15.19	3.29	7.43	4.89*	-10.48	-11.27	37.22**	67.8**	4.29

	S1	6.37	14.3	3.29	28.45*	6.67	23.29	-5.38	6.37	14.3	-7.38*	12.29	0.67	28.45*	-5.38	5.39
	S2	2.34	12.19	22.04	15.34	2.11	3.29	23.22	-2.34	12.19	13.28*	3.29	2.09	15.34	23.22	-4.78
	S3	4.39	1.39	-4.39	14.56	4.89	-4.39	-13.34	-4.39	1.39	2.16	-11.32	-2.19	14.56	-13.28	4.22
FH-113×Cyto-178	N	23.74*	37.2*	3.29	23.76*	22.63*	-7.38	53.5**	56.4**	21.37	14.23	22.05	21.7*	23.75*	13.28	21.73*
	S1	11.78	28.4*	6.37	15.78	14.52	14.38	20.06	-5.38	22.05	12.35	-21.94	11.78	15.78	20.06	11.78
	S2	-4.39	15.34	-2.34	-4.39	-5.39	24.39	-12.48	1.38	21.28	15.38	-5.49	-4.39	-4.19	-12.56	-4.39
	S3	-4.30	14.56	-4.39	-4.30	-4.20	7.49	14.39	14.32	-5.49	11.42	-2.58	-4.30	-4.30	14.39	-4.30
FH-113×VH-363	N	5.28	-6.38	8.21	14.25	4.29	38.9*	22.05	3.82	16.48	13.28	12.48	15.48	14.21	21.37	-7.49
	S1	6.37	14.38	13.28	12.33	6.37	23.29	-21.94	5.17	3.29	11.48	15.38	3.29	12.32	22.05	13.28
	S2	-2.14	24.39	1.78	15.35	-2.34	12.53	-5.49	-4.14	22.04	5.09	11.47	22.24	15.35	-21.28	-7.59
	S3	-4.39	7.49	1.35	11.48	-4.39	-4.39	-2.58	-4.69	-4.39	6.48	5.39	-4.59	11.41	-5.49	-11.45
FH-113×FH-326	N	7.43	0.41	24.77*	-15.38	3.43	36.83*	12.48	16.48	21.7*	33.2**	7.29	2.45	-15.38	72.3**	34.8**
	S1	14.36	1.67	11.78	11.28	24.36	27.46*	15.38	3.29	11.78	28.4*	6.37	6.24	11.28	12.48	11.45
	S2	12.12	1.23	-4.39	-21.08	11.17	14.34	11.47	22.04	-4.39	15.34	-2.34	12.33	-21.08	15.38	-11.45
	S3	1.39	2.77	-4.30	-5.49	1.39	13.26	5.39	-4.39	-4.30	14.56	-4.39	4.83	-5.49	11.47	-12.45
FH-942×Cyto-178	N	-17.21	2.11	14.48	-11.27	-17.21	16.48	23.29*	21.73*	45.6*	45.8**	3.29	-10.48	-11.27	94.8**	11.34

	S1	0.47	11.25	3.29	0.67	0.47	3.29	3.28	11.78	34.82*	32.29	6.37	12.29	0.67	23.29	-16.34
	S2	3.09	3.48	22.04	2.09	3.09	22.04	-4.39	-4.39	12.78	3.19	-2.34	3.29	2.09	3.29	-12.45
	S3	-3.19	31.28*	-4.39	-2.19	-3.19	-4.39	6.49	-4.30	38.44*	-5.39	-4.39	-11.3	-2.19	-4.39	13.28
FH-942×VH-363	N	5.29	3.29	22.05	-34.27	5.29	-11.27	3.29	-15.38	3.29	4.73	-18.38	37.2**	-34.27	16.48	13.28
	S1	6.37	6.37	-21.94	0.87	6.37	0.67	22.05	11.28	6.37	5.17	-2.48	28.4*	0.87	3.29	-11.48
	S2	-2.34	-2.34	-5.49	2.19	-2.34	2.09	-4.33	-21.08	-2.34	-2.34	-6.39	15.34	2.19	22.04	5.09
	S3	-4.39	-4.39	-2.58	-2.19	-4.39	-2.19	6.41	-5.49	-4.39	-2.69	-16.49	14.56	-2.19	-4.39	6.48
FH-942×FH-326	N	21.7*	56.4*	3.99	16.48	24.8*	3.29	11.78	18.38	27.37	21.7*	37.2**	13.43	16.48	37.3**	37.2**
	S1	11.78	-5.38	7.37	3.29	11.78	6.37	-4.39	-2.48	22.05	11.78	28.4*	3.29	3.29	28.5**	28.4*
	S2	-4.39	1.38	-9.34	22.04	-5.39	-2.34	-8.04	-6.39	21.28	-4.39	15.34	22.04	22.04	14.24	15.34
	S3	-4.30	14.32	-4.39	-4.39	-4.70	-4.39	12.71	-16.49	-5.49	-4.30	14.56	-4.29	-4.39	11.56	14.56
DNH-40×Cyto-178	N	21.7*	12.48	-15.38	3.19	19.7*	25.7*	13.28	64.8**	94.8*	67.8**	-10.48	38.9**	3.19	-13.38	-14.38
	S1	11.78	15.38	11.28	6.87	13.78	14.78	11.28	-5.38	23.29	-5.38	12.29	23.29	6.87	-3.19	-3.38
	S2	-4.39	11.47	-21.08	-2.34	-4.39	-4.39	3.07	23.22	3.29	23.22	3.29	12.53	-2.34	23.4*	21.4*
	S3	-4.30	5.39	-5.49	-4.39	-4.30	-6.30	37.5**	-13.28	-4.39	-13.28	-11.3	-4.39	-4.39	12.26	12.46
DNH-40×VH-363	N	-11.27	2.22	66.82**	-10.48	-11.27	37.2*	53.4**	16.48	3.99	-10.48	55.4**	3.29	-10.48	-15.6*	-17.64*
	S1	0.67	7.61*	-5.37	12.29	0.67	28.4*	-5.38	3.29	7.37	12.29	-5.38	6.37	12.29	12.36	12.36
	S2	2.09	-4.13	22.22	3.29	2.09	15.34	1.38	22.04	-9.34	3.29	1.38	-2.34	3.29	23.6*	23.68*

	S3	-2.19	-17.59	-13.28	-11.3	-2.19	14.56	14.32	-4.39	-4.39	-11.3	14.32	-4.39	-11.3	14.26	14.26
DNH-40×FH-326	N	37.2**	1.11	16.48	21.7*	37.2**	22.37	69.8**	-11.27	21.7*	3.29	12.48	27.7*	21.7*	-11.27	-12.27
	S1	28.45*	2.11	3.29	11.78	28.4*	27.05	-5.38	0.67	11.78	6.37	15.38	12.78	11.78	0.67	0.67
	S2	15.34	4.39	22.04	-4.39	15.34	-21.28	23.22	2.09	-4.39	-2.34	11.47	-4.39	-4.39	2.09	2.09
	S3	14.56	4.44	-4.39	-4.30	14.56	-5.49	-13.28	-2.19	-4.30	-4.39	5.39	-4.30	-4.30	-2.19	-2.19

N=control, S1=10 dSm-1, S2= 15 dSm-1, S3= 20 dSm-1, Trt= Treatment, P.H.= Plant height, N.B.= Number of bolls, S.C.Y.= Seed cotton yield, B.W.= Boll weight, FF= Fiber fineness, F.S.=Fiber strength, F.L.=Fiber length, GOT=Ginning turn out, Na^+= Sodium concentration, K^+=Potassium concentration, K^+/Na^+=Potassium to sodium ratio POD=Peroxidase, CAT=Catalase,H2O2= Hydrogen peroxide

Table 10 General combining ability effects of parents for different traits under normal and various levels of salt stress

Genotype	**Trt**	**PH**	**N.B**	**S.C.Y.**	**B.W**	**FL**	**FS**	**FF**	**GOT**	**Na+**	**K+**	**K^+/Na^+**	**Proline**	**POD**	**CAT**	**H_2O_2**
Lines																
NIAB-545	N	0.5	-0.86	-5.09	-0.23	0.43**	1.71**	0.76*	2.81**	2.41	-9.38**	-0.68	0.23**	-0.68	5.38**	0.03
	S1	-6.21	-0.7	-1.72	0.19	0.32**	0.32**	1.17**	0.59*	2.18*	-1.46	-0.18	0.24*	-0.69	4.39*	-0.05
	S2	8.71**	-0.49	-0.81	-0.09	0.17**	1.18**	1.15**	3.28**	-1.59	-8.49**	-0.03	-0.49	-0.39	1.36	0.04
	S3	8.51**	-0.51	-0.81	-0.04	0.32**	1.17**	-8.49**	-0.03	0.67	-9.48	-0.18	0.23	-0.45	1.34	-0.05
CIM-595	N	-3.71	-0.03	0.62	0.12	0.51**	1.79**	0.64*	6.08**	2.31	-2.81	-0.61	-0.18	0.15	-4.39**	-0.08
	S1	-8.51	-0.22	-0.61	-0.72	0.38**	0.78	0.29	3.29**	-0.38	-0.37	0.04	-0.32*	0.42	-1.29	-0.07
	S2		-0.37	-0.29	-0.26	0.37**	0.79*	-0.41	-2.12*	-0.49	-0.51	-0.2	-0.87**	0.74		0.05

	S3	12.74** 11.62**	-0.27	-0.28	-0.16	0.38**	0.78	-0.51	-0.2	0.32	1.17**	0.04	0.58**	0.34	-0.05 -0.23	0.03
Coker-307	N S1 S2 S3	-7.61** -4.91 -0.31 -0.52	-0.72 -1.91** -0.18 -0.17	-0.59 -1.32 0.15 0.18	0.34 0.08 0.14 0.15	-0.59** 0.49** 0.48** 0.49**	-3.29** -3.82** -2.69** -3.82**	-2.64** -2.71** -3.73** 9.29**	-1.04 -0.58 0.38 0.24**	-1.23 -2.76 -3.71 0.31	6.49** 9.27** 9.29** 0.78	0.38 0.34* 0.24** 0.34*	0.13 -0.45** -0.12 0.68**	3.27** 2.18** 2.85** 2.08	-0.89 -1.22 -0.86 -0.76	0.03 0.04 -0.01 -0.02
FH-113	N S1 S2 S3	4.71 -0.27 2.71 2.61	0.87 1.82** 1.15** 1.27**	0.17 1.52 1.72 1.72	-0.41 -0.3 0.27 0.27	-0.32 -0.28** -0.38** -0.28**	0.54* 0.94* 0.04 0.94*	-0.4 0.41 0.09 1.39**	-2.47** -1.53 -1.04 -0.21	0.82 2.93 1.42 0.49	-6.79** -8.39** 7.39** -3.82**	-0.36 -0.48* -0.21 -0.48*	0.11 0.52** -0.34 -0.21	-5.39** -3.29** 2.34** -2.78**	-0.58 -1.59 -1.76 5.49**	0.01 -0.01 -0.06 0.01
FH-942	N S1 S2 S3	12.63** 12.73** 16.92**	1.93* 0.76 0.93* 0.79*	4.98* 6.91** 2.08* 1.98*	0.06 0.18 -0.3 -0.3	0.02 0.13* 0.16** 0.13*	-0.25 0.15 0.69* 0.15	0.71* 0.62 -0.93** 1.32**	-3.71** -3.38** -2.19* -0.12	-2.71 3.06 3.29 -0.28	5.39** 2.61 8.32** 0.94*	0.74 -0.17 -0.12 -0.17	-0.15 0.24* -0.89 0.35	1.03 0.78 -0.14 0.38	2.16 -0.38 -4.78**	-0.05 0.02 -0.02 0.04

		16.82**													-4.38**	
DNH-40	N	-3.71	-0.71	-1.53	0.02	-0.23	0.14	0.93**	-0.38	-2.04	4.29**	0.61	0.12	0.2	0.42	0.02
	S1	5.81	-0.29	0.69	0.25	0.03	0.12	0.96*	-0.17	-1.04	4.27**	0.11	0.14	1.05	2.45	0.04
	S2	1.62	1.26**	2.98**	-0.03	0.06	-0.06	1.20	1.18	1.56	3.31	0.04	0.14	0.47	6.39**	0.01
	S3	1.52	1.61**	2.59**	-0.03	0.03	0.12	3.31	0.04	0.13	0.15	0.11	0.87**	0.41	1.38	0.03
S.E.	N	2.65	0.51	1.41	0.07	0.05	0.26	0.26	0.73	2.61	1.37	0.48	0.25**	0.83	1.38	0.05
	S1	2.71	0.29	1.55	0.12	0.07	0.38	0.29	1.05	-2.81	1.17	0.18	0.03	0.29	2.49	0.03
	S2	1.95	0.39	0.91	0.29	0.05	0.35	0.36	1.04	1.04	1.38	0.05	0.43	0.67	1.36	0.03
	S3	1.85	0.38	0.71	0.26	0.07	0.38	1.38	0.05	0.03	0.12	0.18	0.15	0.75	1.45	0.01
Testers																
Cyto-178	N	1.52	-0.21	-1.5	-0.35	0.16**	0.42*	0.61**	0.65	0.67	-4.29**	-0.25	0.12	1.48**	3.56**	0.02
	S1	-1.52	0.08	-0.16	-0.08	0.17**	0.46	0.63*	-0.89	0.31	-5.39**	-0.26	0.17	0.48	3.98*	0.03
	S2	-0.71	0.3	0.15	-0.12	0.18**	0.49	0.84	-0.39	2.51	7.12**	-0.17**	-0.34	3.25**	0.28	0.04
	S3	-0.76	0.3	0.17	-0.21	0.16**	0.42*	0.61**	0.65	0.63*	-0.89	0.31	0.45*	-0.89*	0.76	0.09
VH-363	N	-2.51	-0.42	-1.51	0.3**	-0.14**	-0.72**	-0.46*	0.52	0.21	-2.45*	-0.15	-0.13	-0.05	-3.71**	-0.03
	S1	-3.71	-1.27**	0.17	0.17	-0.07*	-0.62*	-0.38	0.76	0.69	-1.97*	-0.18	-0.24*	0.28	-1.49	0.01
	S2	-2.41	1.52**	1.71**	0.41			-0.42	0.46	-1.27	-1.04	0.04	0.12	0.47	-0.38	0.06

	S3	-2.08	1.31**	1.71**	0.37**	-0.15** -0.14**	-0.69* -0.72**	-0.46*	0.52	-0.38	0.76	0.69	0.12	-0.41	-0.12	-0.05
FH-326	N	0.68	1.31*	3.07*	-0.04	-0.05	0.17	-0.16	0.15	-0.93	6.38**	0.35	-0.22**	-1.16*	-0.38	-0.02
	S1	6.28*	1.08	2.61	-0.07	-0.05	0.13	-0.29	0.17	0.67	6.95**	0.27	-0.03	-0.87	-1.75	-0.02
	S2	2.91*	1.18**	1.28	-0.31	-0.18	0.12	-0.32	0.03	-1.31	5.16**	0.13*	0.31	0.72	0.17	0.05
	S3	3.61*	1.08**	1.29	-0.28	-0.05	0.17	-0.16	0.15	-0.29	0.17	0.67	0.26	0.28	-0.67	0.02
S.E.	N	2.14	0.47	0.38	0.09	0.04	0.19	0.31	0.52	1.72	1.18	0.27	0.01	0.52	1.36	0.05
	S1	2.05	0.35	1.08	0.07	0.06	0.25	-0.21	0.68	1.69	0.78	0.04	0.01	0.48	1.45	0.03
	S2	1.61	0.28	0.59	0.18	0.04	0.23	0.23	0.71	1.28	1.52	0.05	0.12	0.52	1.11	0.05
	S3	1.51	0.31	0.69	0.17	0.04	0.19	0.31	0.52	-0.21	0.68	1.69	0.18	0.86	0.45	0.03

Discussion

Saline areas are increasing every year due to several factors, such as low precipitation, high surface evaporation, weathering of native rocks, irrigation with saline water, and poor cultural practices. The adverse effects of salinity highlight the need for plant breeders to develop new germplasm of crop plants. Actually, there are some prerequisites before starting a breeding program such as presence of genetic variation in germplasm for certain traits and choice of reliable criteria for screening of large germplasm (Malhotra, 2015; Singh *et al.,* 2023). Plant breeders rely on genetic diversity in various traits to develop new genotypes that can offer resistance to biotic and abiotic stresses. To overcome the salinity stress in cotton, genetic variation has been assessed for its role in conferring salinity stress. Scientists worldwide have confirmed the presence of genetic variability in cotton plants for salinity tolerance by characterizing physiological, biochemical, and yield related parameters (Cushman *et al.,* 2020; Zhao *et al.,* 2020). Studies have shown that each crop has a specificgrowth stage that is particularly sensitive to salt stress and each stage responds differently (Blum, 2018). Selection for salinity tolerance based on seedling traits conducted in several field crops, including maize (*Zea mays*) (Akram *et al.,* 2010), wheat (*Triticum aestivum*) (Hameed *et al.,* 2008), and rice (*Oryza sativa*) (Aslam *et al.,* 1993). Breeding for salt tolerance in cotton or other crop can be successful when plants show some heritable variations for the traits involved.

High Na^+ concentration in leaf sap is a primary indicator of salinity stress (Meneguzzo *et al.,* 2000), and elevated levels of Na^+ disrupt various metabolic activities in cells (Akram *et al.,* 2007). Genotypes that can exclude Na^+ tend to survive more effectively under stress conditions (Akram *et al.,* 2007; Ramani *et al.,* 2018). Simultaneously, higher K^+/Na^+ ratios have been observed in various parts of salt-tolerant cotton (An *et al*., 2025). Many signaling pathways are triggered when salt enters the cell, contributing to the development of ionic tolerance by reducing the net Na^+ translocation and limiting its influx into the cell. To prevent harmful effects on cytoplasmic functions, dangerous ions are ultimately sequestered into vacuoles (Maryum *et al.,* 2022). Chlorophyll content is also an important criterion for selection, as it is degraded under salt stress, leading to reduced photosynthesis and stunted plant growth. Higher chlorophyll content has been observed in salt-tolerant genotypes compared to salt susceptible rice lines. Elevated chlorophyll concentration is associated with increased photosynthetic rates, greater dry matter production, and higher grain yield in rice (Harinasut *et al.,* 2000; Khan *et al.,* 2009). Similarly, lower chlorophyll content in salt sensitive cultivars has been reported in studies of Ashraf *et al.* (2005); Iqbal *et al.* (2006) and Khan *et al.* (2009). Some genotypes maintained higher level of antioxidants under stress compared to other accessions. Additionally, the ROS-scavenging capacity of CIM-595, Coker-307, and FH-113 was also high. There is positive association between antioxidants and ROS in cotton, as increased antioxidants concentration in response to salinity also elevate ROS generation (Ibrahim *et al.,* 2019; Hasanuzzaman and Fujita, 2022). This indicator has also been successfully used to identify salt tolerant lines of wheat (Kumar *et al.,* 2017), cotton (Sairam *et al.,* 2001), and rice (Vaidyanathan *et al.,* 2003). Similar findings were reported by Zafar et al. (2022b) and Farooq et al. (2022), who described increased SOD, POD, and CAT activity in leaves under salt stress. Dong et al. (2020) reported a 53% increase in POD activity in tolerant cotton cultivars. During fiber development, antioxidant activity increases in response to salt stress (Ju et al., 2023).

These findings are consistent with those of Serrano and Rodriguez-Navarro, (2001). Similarly, salt stress significantly affects boll weight and the number of bolls in cotton plants (Nawaz *et al.,* 2023). The number of bolls was reduced due to poor growth, toxicity caused by excessive uptake of sodium and chloride ions. (Akhtar *et al.,* 2010). In our study, the number of bolls were more in salt tolerant cotton cultivars as compared to salt susceptible parents and crosses. Similar results were found by Sun et al., 2024; Zafar et al., 2025 and Guo et al., 2024. Further, reduction in plant growth can also be referred to as shrinkage of cell walls, hindrance in the differentiation of the tissues, disturbance of essential ions, and injuries to the growing tissues (Dong*et al.,* 2010). The reduction of boll weight reported when photosynthesis processes are affected under salt stress and number of ATPs will produce less quantities (Toyoshima *et al.,* 2011). Many scientists observed reduction in boll weight under salt stress result into poor yield (Farooq et al., 2020, Ma et al., 2021). The fiber quality trait also showed poor performance in salt stress with decrease and negative correlation was found with salt stress (Folkman, 2013; Zhang *et al.,* 2013). The results are in accordance with the findings of Zhao et al., 2025 and Li et al., 2025. Most of fiber quality traits namely fiber strength, fiber fineness, and fiber length were decreasing due to salt stress (Zhang *et al.,* 2024). Cotton growth and lint yield was significantly reduced in salt stress treatment compared to control. A similar finding was observed by Heng *et al.,* (2024). Heterosis breeding involves determining the genetic architecture of crop plants in agricultural research (Wang *et al.,* 2016). The crosses that showed better heterotic values must be considered for hybrid development program suitable for salt affected areas (Turk, 2017). Abdel-Aty et al. (2023) conducted an experiment and observed cross combinations with positive and significant heterotic effects over the better parents for agronomic and yield-related attributes of cotton, which could be utilized for the development of high-yielding cultivars. The occurrence of dominant gene action for recorded traits suggests that the genotypes can be a good option for the hybrid development.

Conclusion

In conclusion, the complex genetic architecture of cotton traits requires a comprehensive understanding for its improvement for salt stress tolerance. The genotypes, NIAB-545 and FH-942 were found to be best general combiner amongst the lines whereas FH-326 was declared as best general combiner from tester series. This indicated parents having high GCA can be used in crossing programs for enhancing salinity tolerance in the genotypes. The hybrid, FH-942 × FH-326 was the best specific combiner and best heterotic hybrid. These results and findings can be employed for more cotton breeding programs planned for the development of new germplasm to be grown in salt affected areas of the country.

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Ethics Declaration

Disclaimer

USDA is an equal opportunity provider and employer.

Availability of data and materials

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Ethics approval and consent to participate

Not applicable.

Consent for publication

Not applicable.

Competing interests

All authors declare no competing interests.