Indian Journal of Experimental Biology
Vol. 60, January 2022, pp. 49-58

Relative performance of wheat genotypes under individual and combined water deficit and salinity stress

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Received 14 October 2019; *revised 12 March 2021*

Ascertaining the genetic variability and its relationships among valuable genetic resources is important for crop improvement programme. Here, we assessed the response of eleven wheat (*Triticum aestivum* L.) genotypes using cluster and principal component analysis (PCA) based on morphophysiological data and yield under nine different environments. Wheat genotype WH 1080 maintained higher photosynthetic efficiency under individual stress of 50% water deficit (drought) and 100 mM NaCl (salt), whereas under interactive stresses KRL 370 and KRL 283 were found to be the best genotypes. The highest value of Na^+/K^+ ratio in shoots was recorded for WH 1080 (1.167) and lowest in KRL 283 (0.612) under combined stresses. Proline accumulation was maximum in KRL 330 (3.17 mg g^{-1} FW) and minimum in KRL 283 (2.8 mg g^{-1} FW). Significantly higher reduction (73.4%) was observed in HD 2009 for grain weight/plant at 100 mM NaCl + 50% WD stress treatment whereas minimum reduction of 39.18% was recorded in KRL 370 in comparison to the control treatment. The PCA showed that the first three components comprising about 91% of the total variation for which the variables were analyzed. AMMI model revealed KRL 210 to be stable genotype as being close to center on biplot. E_5 environment (100 mM NaCl) was most stable followed by E_9 (50% WD + 100 mM NaCl). HD 2888, C-306, HD 2851 and HD 2009 were having positive interaction with E_1 (Control) whereas WH 1080 had positive interaction with water deficit environments i.e. E_2 and E_3 (25 and 50% WD) while KRL 433 had highest positive interaction with combined water deficit and salt stress environments E_6, E_7, E_8 and E_9, followed by KRL 370. Similarly, KRL 283, KRL 330, KRL 210 and Kharchia 65 had high positive interaction with saline environments E_4 and E_5. Findings of the experiment would be beneficial to wheat breeders, specifically the location-specific promising genotypes could possibly be used to develop/breed MAGIC populations to tag genes/alleles conferring drought and salinity tolerance.

Keywords: Abiotic stress, Drought, GGE biplot analysis, *Triticum aestivum*

Soil salinity is one of the notable constraints that have been affecting agriculture in more than 100 countries, worldwide. In recent years, scarcity of freshwater and the secondary salinization of agricultural lands are becoming bigger challenges worldwide. Presently, 6.74 million ha of land is prone to salinity and sodicity in India which will likely to increase to 16.2 million ha by 2050[1]. Soil salinity associated stresses particularly drought can be more pronounced and more detrimental to crop production in years to come as salinized plants experience, initially osmotic stress and subsequently specific ion effects[2,3]. Osmotic stress (inhibits water uptake) is first experienced by roots[4], which have an effective mechanism to sense low water potential arise due to low soil moisture and increased salt concentration. In both these situation, plants are unable to take water from the soil that is necessary for their growth and development, which ultimately leads to the activation of signal transduction pathway common to water deficit and salinity stresses[5-7].

Salt tolerance is a multifarious phenomenon that necessitates alterations in developmental, morpho-logical, physiological and biochemical processes, including reduction in growth and water uptake, modification in stomatal behaviour and reduced photosynthetic efficiency, increased osmolyte accumulation, disturbed ion balance and stress induced gene expression[2,8-10]. Under salt stress conditions, accumulation of Na^+ and Cl^- ions in plant tissues is harmful and is the focus of research on salinity to date[11]. Globally, wheat grown in 220.83 million ha areas, which produced ~769.31 MT of wheat grain. After sharing 107.18 MT grains in

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Abbreviations: AMMI, Additive main effects and multiplicative interactions; ; DAS, days after sowing; dS/m, desi sieman per meter; EC, electrical conductivityt; E, environment; FW, fresh weight; MAGIC, multi-parent advanced generation intercross population; PCA, principal component analysis; Pn, photosynthetic rate; WD, water deficit

national food basket from 30.55 million ha areas, consistently India secure their second position after China in wheat production, with the average productivity of 3508 kg/ha. Salinity stress coupled with drought stress negatively affects the wheat productivity[12] and wheat yields start declining when ECe value exceeds 6 dS m^{-1} in the soil solution[13]. The problem of salinity coupled with drought is widespread in dry land regions and this problem is aggravated further due to extensive exploitation of water resources.

To overcome the adverse effects of salinity and drought stresses, we need to identify tolerant cultivars which will perform better under these situations as well as the mechanism or the traits responsible for their tolerance. Hence, in the present study, we tried to evaluate wheat genotypes in terms of relative physiological, biochemical and agronomic traits related to stress tolerance.

Materials and Methods

The experiment was designed in a randomized complete block design to evaluate eleven wheat genotypes (differing in their tolerance) for salinity and drought (water deficit; WD) stress responses during 2016-17 and 2017-18 in net house of Crop Improvement Division, ICAR-Central Soil Salinity Research Institute (CSSRI), Karnal, Haryana, India. For this, different treatments of individual and interactive water deficit and salinity stresses *viz*. Control (E1), 25 and 50% water deficit alone (E2 and E3), 50 mM and 100 mM NaCl alone (E4 and E5), 25% WD + 50 mM NaCl (E6), 50% WD + 50 mM NaCl (E7), 25% WD + 100 mM NaCl (E8), and 50% WD +100 mM NaCl (E9) were imposed in 20 kg capacity clay/porcelain pots filled with sandy loam soil in 5 replications. Surface decontaminated seeds of Kharchia 65, KRL 210, KRL 283, KRL 330, KRL 370 (Salinity tolerant), KRL 433 (Salinity and Drought tolerant genotype), HD 2888, WH 1080 and C 306 (Drought tolerant), HD 2009 and HD 2851 (Salt sensitive) were sown in the 2nd week of November in pots. Prior to imposition of stresses, nutrients were supplied through Hoagland nutrient solution. After the initial early growth, salinity and drought stresses (21 DAS) were applied in the pots through a standard methodology. Water deficit stress was given by withholding irrigation supply on the basis of field capacity and salt stress was applied through the application of 50 and 100 mM concentration of sodium chloride (NaCl). The net house was covered with superior quality polythene sheet to evade the entry of rainwater and retain the desired salinity and water deficit stress levels in the pots as per treatments.

Clay/porcelain pots (20 kg capacity) packed with 16 kg soil (field capacity 28% v/v; bulk density of 1.45 g/cc and porosity approximately 40%) were saturated by 100 % first and thereafter depletion of water to 25 and 50% in soil (25 and 50% water scarcity) was made on the basis of field capacity by withholding irrigation supply. For this, 6.5 L water (up to field capacity) was applied in the pots at the weekly interval and evaporation was recorded through pan. During the whole study period, pan evaporation was 2-3 mm day^{-1} *i.e.* 21 mm $week^{-1}$. On this basis, 25 and 50% water deficit treatments were created. Salinity treatment was given as 50 and 100 mM NaCl, applied to pots at regular weekly interval. For taking observations, five plants of each varieties and each treatment were tagged and data were recorded at reproductive stage. Plant height of all the five tagged plants was measured with the help of meter scale rod from the ground surface to the tip of the upper most fully opened leaf. Fully expanded flag leaves were sampled to quantify the chlorophyll content using DMSO (Dimethyl sulphoxide) as described by Hiscox and Israelstam[14]. Photosynthetic rate (Pn) was measured with an infrared open gas exchange system (LI-6400, LICOR Inc., Lincoln, NE, USA) between 10:00 AM and 12:00 PM. Fresh samples were grinded in 3% sulphosalicylic acid to estimate proline content with the method of Bates *et al*.[15] using acid ninhydrin reagents and quantified at 520 nm against blank toluene. For ionic (Na^+ and K^+) contents, collected samples were sundried initially and thereafter shifted in the oven to dry at 65±5°C till a constant weight was achieved. These dried plant sample were grinded and a known quantity of sample (about 0.1 g) was taken in 50 mL flask and digested with 10 mL of di-acid mixture (HNO_3:$HClO_4$ 3:1) by heating smoothly on a hot plate till the solution turns out colourless. After digestion, the contents were cooled and volume was made to 50 mL with DDW and ionic content was measured on flame-photometer (Flame Photometer 128, Systronics) and subsequently, the ratio of Na^+/K^+ was calculated. Treatment/genotype wise five tagged plants were used to record the plant yield in terms of g/plant. All the data were subjected to statistical analysis using statistical programme SAS Version 9.3

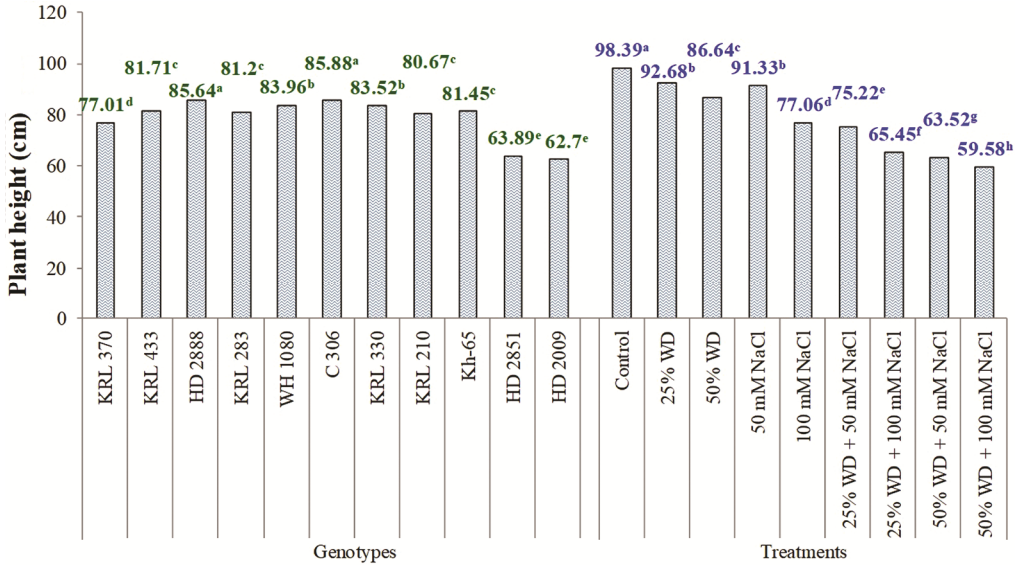

Fig. 1 — Variation in plant height w.r.t. water, salinity and combined stress in wheat

(SAS Institute Inc., Cary, NC, USA) using Duncan's multiple range test.

Results and Discussion

Recorded traits

Flag leaves were used for taking observation on physiological and biochemical traits at reproductive stage after seeing the visible effect of stresses (tip burning/yellowing of leaves). The observations were averaged to work out the mean plant height per pot and observed that plant height decreased under stress conditions *i.e.* 5.8% under water deficit stress, 21.7% under salinity stress but severe effects (39.45%) were noted under combined stresses (Fig. 1). Among genotypes, the minimum reduction was found in KRL 370 (24.7%); KRL 433 (29.9%) and maximum in HD 2851 (64.9%) followed by HD 2009 (63.9%) at stress level of 50% WD in combination with 100 mM NaCl than their respective control. Decreased turgor pressure due to reduced uptake of water from the soil, reduced nutrient availability and higher accumulation of toxic ions that ultimately lead to inhibition of cell division and cell expansion could be the possible reason for decrease in plant height and this response is further aggravated by the interaction of both the stresses[16].

Salt toxicity is accountable for the burning of the leaves and other sensitive parts and it resulted in the deprivation of several pigments contained within the plant including chlorophyll that acts as a biochemical marker for stress tolerance. Similarly, these genotypes showed less reduction under individual stresses (8.2%

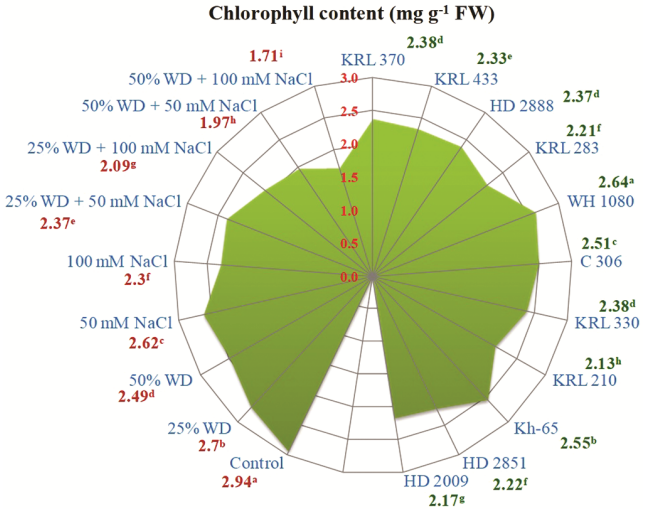

Fig. 2 — Association of genotypic variation in wheat with treatment effect for chlorophyll content

at 50% WD; 21.8% at 100 mM NaCl) rather than combined stresses (41.8% at 50% WD + 100 mM NaCl) than the respective control (Fig. 2). This might be due to the fact that stresses inhibit the activity of ALA synthase enzyme that is responsible for the synthesis of chlorophyll pigments or due to reduced uptake of minerals particularly magnesium, required for the biosynthesis of chlorophyll pigments. Among different wheat genotypes, KRL 283 showed minimum reduction under individual stress *i.e.* 1.57% reduction at 50% WD and 12.99% at 100 mM NaCl whereas, under combined stresses, KRL 370 is the best one (30.99% reduction) followed by KRL 283 (31.5%). Sensitive genotypes showed much higher decrease under individual and combined stresses

because of increased chlorophyllase enzyme activity or due to photoinhibition/ROS formation[17,18].

The photosynthesis process is the backbone for producing biomass by means of source activity, therefore if any change occurs in this attribute due to stress hampered the crop yield. Photosynthetic rate (Pn) in wheat genotypes decreased with increasing levels of stresses in all the genotypes and showed overall 7.79% reduction under water deficit stress; 17.44% reduction under salinity stress and 23.45% under combined stress (Table 1). In nutshell, WH 1080 showed higher photosynthesis efficiency (30.28 µmol CO_2 m^{-2} s^{-1}) over all the treatments but the reduction was lowest in Kharchia 65 (3.85%) at 50% WD, WH 1080 (10.66%) at 100 mM NaCl and KRL 210 (17.3%) at 50% WD + 100 mM NaCl. The possible reason for decreased photosynthesis includes reduced activity of Rubisco due to stomata closing and feedback inhibition through reduced sink size[19] or salts directly reduced turgor pressure in guard cells, thus inhibiting stomatal conductance[20]. The decrease in photosynthesis was positively correlated with the biomass and observed that biomass significantly decreased with increasing stress intensification *i.e.* 14.05% at 50% WD, 22.58% at 100 mM NaCl and 38.83% at 50% WD + 100 mM NaCl. Reductions in the biomass is a general strategy under stress environment and also an indication of severe growth restrictions as depicted by reduced plant height, number of leaves and shoot/root ratio. Genotypic variability revealed that genotype HD 2888 showed less reduction in biomass under water deficit (0.22% reduction at 50% WD) as well as salinity stresses (10.67% reduction at 100 mM NaCl) whereas under combined stresses Kharchia 65 showed minimum reduction of 29.85% (Table 2). Genotype HD 2851 showed maximum biomass reduction under all the stress conditions (25.44% at 50% WD, 33.71% at 100 mM

Table 1 — Differential rate of photosynthesis under individual and combined stress in wheat genotypes

Treatment /Varieties	Photosynthetic rate (µmol CO_2 m^{-2} s^{-1})									
	Control	Water deficit stress		Salt stress		Water deficit + Salt stress				Mean
		25% WD	50% WD	50 mM NaCl	100 mM NaCl	25% WD + 50 mM NaCl	25% WD + 100 mM NaCl	50% WD + 50 mM NaCl	50% WD + 100 mM NaCl	
KRL 370	34.71	32.47	29.78	32.68	30.07	30.13	27.75	26.49	24.36	29.83^{abc}
KRL 433	34.05	30.60	27.50	31.16	27.80	28.62	26.40	25.24	22.77	28.24^{cd}
HD 2888	37.37	32.21	27.90	33.18	27.69	28.73	26.03	24.77	19.99	28.65^{bcd}
KRL 283	34.96	32.32	29.54	32.41	30.03	30.12	28.22	28.01	23.76	29.93^{ab}
WH 1080	34.98	32.44	30.64	33.03	31.25	31.63	29.33	27.15	22.05	30.28^{a}
C 306	34.43	31.43	29.49	31.11	28.93	29.68	26.53	25.22	21.42	28.69^{abc}
KRL 330	35.78	33.15	30.34	32.87	30.43	30.31	26.22	27.32	22.45	29.87^{ab}
KRL 210	36.62	33.71	29.44	31.06	28.14	29.91	26.74	25.94	22.41	29.33^{abc}
Kh-65	31.62	29.73	26.73	29.55	26.99	27.10	24.81	24.90	22.32	27.08^{d}
HD 2851	29.10	27.98	24.72	24.51	23.20	23.94	20.75	19.79	17.39	23.49^{e}
HD 2009	29.13	27.68	24.13	25.84	23.22	22.55	20.69	19.31	16.69	23.25^{e}
General Mean	33.89^{a}	31.25^{b}	28.2^{c}	30.67^{b}	27.98^{c}	28.43^{c}	25.77^{d}	24.92^{d}	21.42^{e}	
LSD @ 5%	Varieties: 1.61; Treatment: 1.17; Treatment means at same level of varieties: NS; and Varieties means at same or different level of Treatment: NS									

Table 2 — Differential biomass accumulation under individual and combined stress in wheat genotypes

Treatment/Varieties	Control	T1	T2	T3	T4	T5	T6	T7	T8	Mean
KRL 370	20.67	16.99	15.72	18.44	15.36	15.52	14.59	14.23	12.90	16.04^{B}
KRL 433	20.51	16.05	15.58	18.27	16.38	15.87	14.86	14.22	13.65	16.15^{B}
HD 2888	18.19	18.15	16.29	18.14	16.25	16.09	13.97	13.51	12.42	15.89^{B}
KRL 283	20.00	18.05	15.57	17.83	15.69	15.43	15.17	14.00	13.01	16.08^{B}
WH 1080	19.78	18.15	16.79	18.10	15.96	15.59	13.55	13.23	12.76	15.99^{B}
C 306	22.13	16.50	15.00	18.87	14.67	12.73	12.50	12.20	10.77	15.04^{C}
KRL 330	21.68	17.49	16.17	17.28	16.02	15.70	13.72	13.91	12.95	16.1^{B}
KRL 210	20.73	17.98	15.91	17.36	15.68	16.25	14.08	13.47	12.70	16.02^{B}
Kh-65	19.13	17.85	15.42	17.82	16.10	15.69	14.32	13.62	13.42	15.93^{B}
HD 2851	22.30	19.60	16.67	21.03	17.20	15.21	14.89	13.87	12.40	17.02^{A}
HD 2009	19.52	16.23	15.25	16.13	14.63	12.17	12.67	11.67	10.36	14.29^{D}
General Mean	20.42^{A}	17.55^{C}	15.85^{D}	18.12^{B}	15.81^{D}	15.11^{E}	14.03^{F}	13.45^{G}	12.49^{H}	
LSD @ 5%	Varieties: 0.52; Treatment: 0.41; Treatment means at same level of varieties: 1.37; and Varieties means at same or different level of Treatment: 1.39									

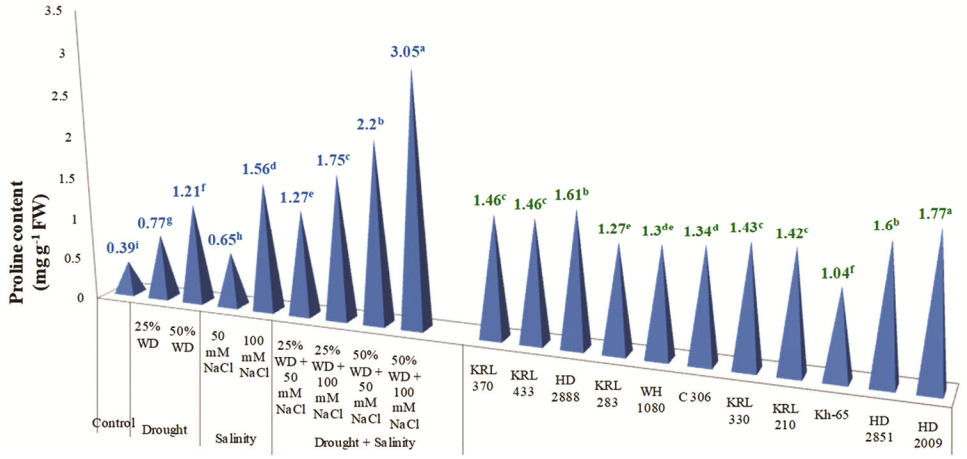

Fig. 3 — Proline accumulation in wheat genotypes w.r.t water, salinity and combined stress

NaCl and 51.33% at 50% WD + 100 mM NaCl). Due to osmotic and ionic stresses caused by water deficit and salt stresses, plants showed nutrient imbalances which adversely affects the photosynthetic efficiency, as well as the transportation of total assimilates to young leaves which ultimately hampers the total biomass production[16,21].

Proline accumulation was increased in general with stress intensification and maximum accumulation was observed in HD 2009 (3.95 mg g^{-1} FW) and minimum accumulation in Kharchia 65 (2.18 mg g^{-1} FW) under combined stresses of 50% WD + 100 mM NaCl. It was also noted that proline content increased to 3.1 fold under water deficit stress of 50% WD, 4 fold at 100 mM NaCl and 7.82 fold at interactive stress of 50% WD + 100 mM NaCl than control (Fig. 3). In addition to this, it was also observed that salinity stress of 50 Mm NaCl in combination with 25 and 50% WD resulted in lesser proline accumulation with respect to 100 mM NaCl. Such increase in proline content in response to combined stresses seem to be associated with the better ability of plants to endure such stressful conditions[22] and also facilitated to enhance osmotic potential[23,24] by taking up additional water from the environment.

Na/K ratio is also one of the critical factors in determining the genotypic ability to tolerate salinity stress[2,25]. The nutrient imbalance created by ion toxicity is mainly because of substitution of K^+ by Na^+ as both ions strive to enter into the plant root cells. From the recorded observations, no significant increase was seen for Na/K under drought stress (1.8 fold) while at 100 mM NaCl, it increased by 4.4 fold (Fig. 4). An abrupt increased ratio of Na/K was noted

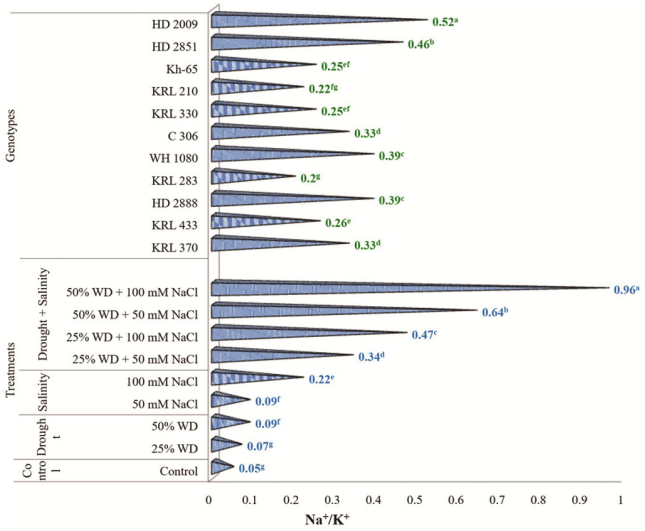

Fig. 4 — Response of wheat genotypic variation with treatment effect on Na^+/K^+ ratio

under combined stresses *i.e.* 10.67 fold and 19.2 fold increase at 25% WD + 100 mM NaCl and 50% WD + 100 mM NaCl, respectively in comparison to control. Genotype KRL 283 showed minimum Na/K ratio under individual salinity and water deficit stress as well as under its interaction with water deficit *i.e.* 1.4 at 50% WD, 2.6 at 100 mM NaCl and 12.6 at 50% WD + 100 mM NaCl compared with its control (Fig. 4).

It was also noted from the results that sensitive genotypes are highly affected by the presence of higher Na^+ which might displace the K^+ and Ca^{2+} due to the undeviating competitiveness between them at plasma membrane level that could also change the composition, integrity, and permeability of plasma membrane[26]. These results are in similarity with the

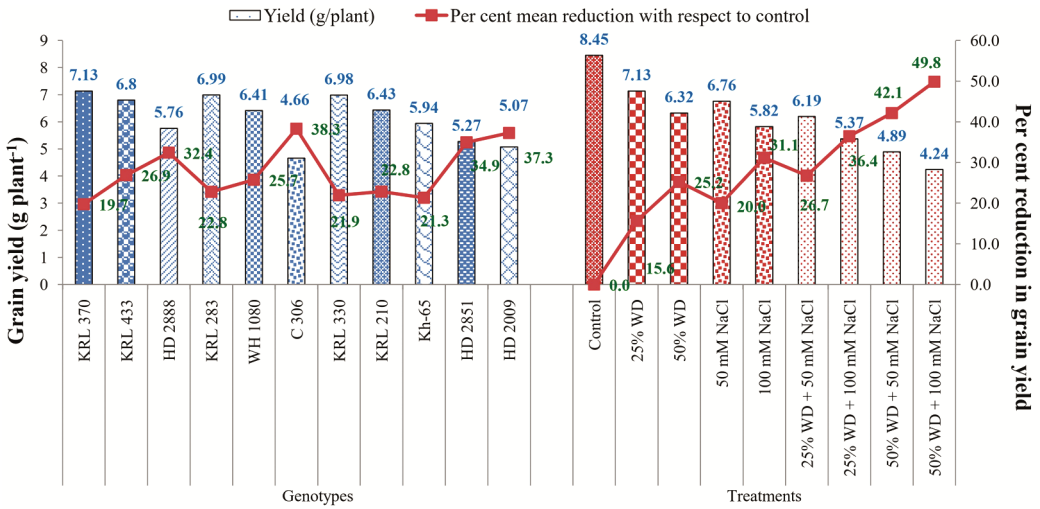

Fig. 5 — Grain yield of wheat genotypes under water, saline and combined stress

earlier finding of Chippa & Lal[27]; Sharma & Gill[28] who also observed that tolerant crops varieties manifested lesser K reduction with less buildup of Na as compared to sensitive ones that resulted in low Na:K or high K:Na ratio[2].

Yield depends on the capability of the crops to assimilate and exploit the available resources and, thus, it is the interaction of many components contributing to final harvest. Reduction in photosynthesis sources including plant leaf area and shoot length disturb the source - sink ratio due to stress occurrence before flowering and hence producing lesser grains. In the present study also these individual stresses declined the mean grain yield by 15.62% under 50% water deficit and 31.12% under saline stress of 100 mM NaCl whereas plant yield reduced drastically under combined stress of 50% WD + 100 mM NaCl (Fig. 5).

Reduction in grain yield might possibly be due to decreased pollen viability and stigma receptivity leading to poor seed setting, chaffy grains and reduced seed weight under stress conditions ultimately culminating in lower crop yields[29,30]. Among the genotypes (Fig. 5), Kharchia 65 showed minimum reduction of 5.43% at 50% WD, KRL 370 at 100 mM NaCl and 50% WD + 100 mM NaCl (19.03 and 36.49%), respectively. Maximum reduction was noted in KRL 433 (26.56%) under water deficit stress and in HD 2009 at 100 mM NaCl (46.53%) and 50% WD + 100 mM NaCl (68.94%). The result obtained depicted that genotypes which have tolerance to one stress, could also tolerate the other stress. Interestingly, significantly higher grain yield was recorded in KRL 370 *i.e.* 7.13 g/plant followed by KRL 283 and KRL 330 (6.98 g/plant) and lowest was recorded in C-306 (4.66 g/plant) over all the treatments. Higher reduction in sensitive genotypes might be due to inadequate photosynthetic source or early maturity (shrivelled grain).

Table 3 — Coefficients associated with the first three principal components

Particulars	PC1	PC2	PC3
Eigen value	5.74	0.38	0.26
Variance (%)	82.00	5.00	4.00
Cumulative variance (%)	0.82	0.87	0.91
Vector Coefficient			
Plant height	00.39	00.03	00.03
Chlorophyll content	00.38	0.43	00.17
Photosynthetic rate	00.36	00.73	0.20
Na/K	0.38	00.21	00.65
Proline content	0.39	00.25	00.26
Biomass	00.38	0.18	00.58
Grain yield	00.37	00.37	00.32

Principal component analysis (PCA)

In reflecting the discrepancy patterns among the genotypes, PCA analysis revealed that the first three principal components are most suitable and constructive in discriminating the variation among different genotypes. First three components comprising about 91% of total variation (Table 3), that provides a clear understanding of the elementary structure for which the variables analyzed. The selection of coefficients of the proper vectors made on cut-off limit *i.e.* greater than 0.3 (positive or negative value as per desired traits) had an adequate outcome to be adjudged significant[31].

Out of the three principal components, the first component accounted for 82.0% of total variance that

might indicate that higher proline content and lower plant height were the variables that contributed towards stress tolerance which were also related with high yield component values (Table 3). The second component represented 5% of total variance which ascertained the role of high chlorophyll content and biomass accorded positively for stress tolerance in wheat. The third principal component signified for 4% and was allied with low Na/K and high photosynthetic rates which might have played some role in stress tolerance (Table 3). Results of earlier researchers[32-34] are corroborative with our findings regarding importance of these traits for abiotic stress tolerance.

Genotype × Environment Interaction (GGE) analysis of Genotype-by-Environment Data (AMMI analysis)

In our results, significant yield differences were observed among wheat genotypes using AMMI analysis of G×E data. The G×E component was again chunked and described by two interaction principal component axes (IPCA) namely IPCA1 and IPCA2. The outcomes of AMMI1 (AMMI model with first IPCA axis) and AMMI2 (IPCA1 with IPCA2) analysis is presented with the help of biplot in Fig. 6 A and B, respectively.

More than 8% (PC1 = 60.5; PC2= 27.8) of the total variation was described by the first two IPCA axes, hence AMMI analysis was effectual in the elucidation of G×E interaction component. Graphical representation of IPCA1 with mean grain yield (Fig. 6A) divulged that KRL 370 had the highest attribute significance whereas C 306 showed the utmost positive AMMI1 score. Among different environments, E_1 (Control) was most favorable for analyzed trait (8.63) with high positive interaction with genotypes (0.65). Even though E_2 environment (25% WD) showed the highest positive interaction with genotypes but the mean value (7.31) is less than E_1. Similarly the environment E_3 (50% WD), also manifested the positive interaction (0.72) with genotypes with mean value (6.50) is less than E_2. Remaining other environments *i.e.* E_4, E_5, E_6, E_7, E_8 and E_9 had lower mean values in comparison to E_3 and exhibited negative interaction with genotypes (Table 4). According to the AMMI model, the genotypes which are designated by means greater than grand mean and virtually zero IPCA score are reckoned as generally adaptable to all environments. Hence, on the basis of analyzed data, WH 1080 and KRL 210 were having general adaptability. On the other hand, high mean performance of the genotypes with greater value of IPCA score are judged as specific adaptable to the environments. Genotypes KRL 370, C-306, HD 2851 and HD 2009 owing specific adaptation because of their higher mean and IPCA score. Wheat genotype (WH 1080) possessed positive interaction and showed specifically favored adaptation with E_1, E_2 and E_3 environment. Environment that is virtually noticeable near to the perpendicular line have similar means and the others those visible near to horizontal line have similar interaction pattern. AMMI1 biplot suggested that all

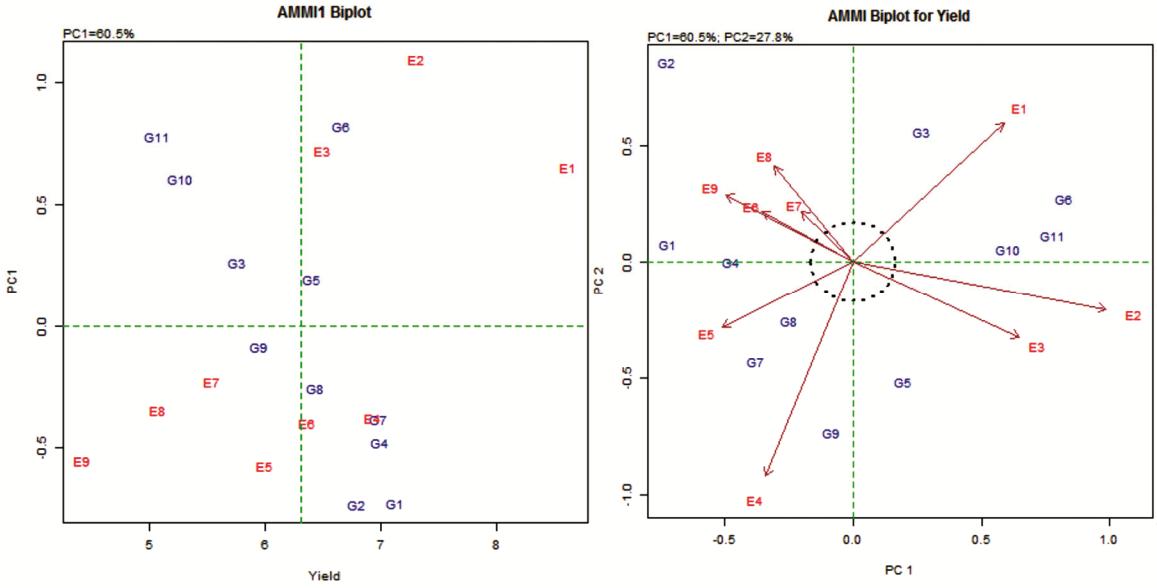

Fig. 6 — Biplot of mean grain yield with IPCA1 (A) and IPCA1 with IPCA2 (B)

Table 4 — Analysis of variance of AMMI model for Yield; and AMMI1 and AMMI2 score for 11 genotypes and nine environments

Source		D.F.	S.S.	M.S.
Genotype		10	135.70	13.57**
Environment		8	415.09	51.89**
Genotype x Environment		80	52.24	0.66**
AMMI 1		17	21.05	1.24**
AMMI 2		15	9.68	0.65**
AMMI Score of Genotypes and Environments				
	Genotypes (Code in Biplot)		AMMI 1	AMMI 2	Mean yield
G1	KRL 370		00.72	0.07	7.13
G2	KRL 433		00.73	0.86	6.80
G3	HD 2888		0.26	0.56	5.76
G4	KRL 283		00.48	0.00	6.99
G5	WH 1080		0.19	00.52	6.41
G6	C-306		0.82	0.27	6.65
G7	KRL 330		00.38	00.43	6.98
G8	KRL 210		00.25	00.26	6.43
G9	Kharchia 65		00.09	00.73	5.94
G10	HD 2851		0.60	0.06	5.27
G11	HD 2009		0.78	0.11	5.07
	Environments				
E_1	Control		0.65	0.66	8.63
E_2	25 % Water deficit		1.10	00.23	7.31
E_3	50 % Water deficit		0.72	00.36	6.50
E_4	50 mM NaCl		00.38	01.02	6.94
E_5	100 mM NaCl		00.57	00.31	6.00
E_6	25 % Water deficit + 50 mM NaCl		00.40	0.24	6.37
E_7	50 % Water deficit + 50 mM NaCl		00.23	0.24	5.55
E_8	25 % Water deficit + 100 mM NaCl		00.34	0.46	5.08
E_9	50 % Water deficit + 100 mM NaCl		00.55	0.32	4.42

three environments are divergent for mean and interaction.

AMMI2 biplot does not manifest the additive main effects, but it is very explanatory on interaction component and the graph is highly applicable when IPCA2 is substantial and consequential. In AMMI2 biplots, the genotypes having score near to the centre of the biplot are considered as more stable since the stability reduces with increased distance from the centre. AMMI 2 biplots also described the nature of interactions of genotypes with the environment by measuring different angles between G and E vectors such as positive for acute angles, negligible for right angles, and negative for obtuse angles. Concomitantly, the correlation is determined through the angle developed between vectors of two different environments. KRL 210 was stable genotypes as being close to centre on biplot. E_5 (100 mM NaCl) was most stable environment followed by E_9 (50% WD + 100 mM NaCl) as suggested by AMMI2 score (Table 4). HD 2888, C-306, HD 2851 and HD 2009 were having positive interaction with E_1 (Control). WH 1080 had positive interaction with water deficit environments *i.e.* E_2 and E_3 (25 % and 50% WD) while KRL 433 had highest positive interaction with coupled stress environments E_6, E_7, E_8 and E_9, followed by KRL 370. Similarly, KRL 283, KRL 330, KRL 210 and Kharchia 65 had high positive interaction with saline environments E_4 and E_5. Similar to our findings, Singh *et al*.[35] and Mackey *et al*.[36] were also reported corroborative results for identification of traits, genotypes and best environmental conditions for abiotic stress tolerance in bread wheat.

Environment analysis

The "which-won-where" GGE biplot analysis is an effectual analytic aid for analyzing bigger environments[41]. The best outcome of the polygon bi-plot analysis is to conceptualize all possible interactions of genotypes within different environments. The perpendicular lines in the biplot have divided the biplot into 5 sectors in which each location fell in either of the sectors (Fig. 7A).

In this study, this 'which won where' feature of the biplot ascertained that KRL 433 was the winning genotype in environment E_1 and E_8. Similarly, KRL 370 was the vertex/winning genotype in environment E_5, E_6, E_7 and E_9 whereas; genotype KRL 330 was the

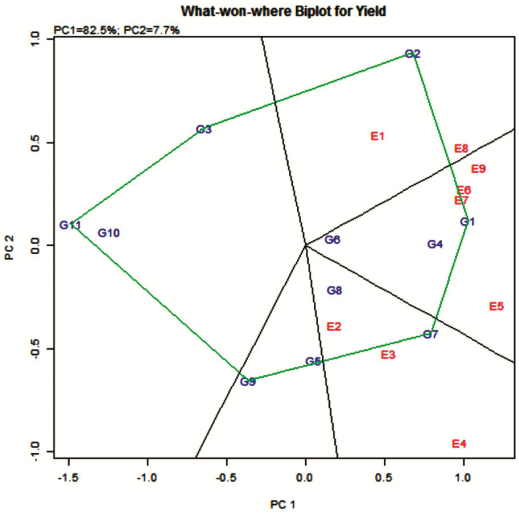

Fig. 7 — (A) G×E data based view of "which-won-where" GGE biplot for plant yield. The genotypes are labeled as G1 to G11 and the environments are labeled as E_1 to $E_{9;}$ (B) The "mean performance and stability of genotype based on a subset of the G×E data; and (C) The discriminating power and representativeness of test environment based on a subset of the G×E data.

winning genotype in environment E_2, E_3 and E_4. The vertex genotypes were identified as the most responsive genotypes being placed farthest from the point of origin[37]. On the other hand, the result also showed some genotypes (HD 2888, WH 1080, Kharchia 65, and HD 2009) which fall in sectors where there were no locations at all; hence these genotypes seem to be poorly adapted to five locations. The present results are in confirmation with the studies of earlier wheat researchers[32,33,35,36].

Genotypes evaluation for stress-environment

The evaluation of genotypes is purposeful specifically where mean performance of the ideal genotypes coincide with maximum stability. As both G+GE contribute for GGE and also the AEC abscissa represents genotype's contributions to G, hence, the AEC ordinate depicting a genotype's stability should specify the genotypes' contributions to GE. In our study also, G1 is the most stable genotype located proximal to the AEC abscissa with a near zero projection onto the AEC ordinate (Fig. 7B). It means this genotype is most consistent within the saline environment (Fig. 7B). Our findings are in conformity with results reported by Mwadzingeni *et al*.[32] and Grzesiak *et al*.[33].

Evaluation of test environment

The "ideal" test environment should discriminate the genotypes representing the adoptable-environment. As AEC abscissa is the "average-environment axis," having small angles with it, hence the test environments with small angles and longer vectors are perfect for selecting superiority of genotypes. When the test environment is close to the origin of biplot, it will not differentiate the genotypes because the genotypes will have similar performance in that test environment. AEC abscissa with long vectors and angles can only be used for culling unstable genotypes but cannot be used for selecting superior genotypes. Our studies also represent E5, E6 and E7 as the most discriminating environment (Fig. 7C). Similar results were also reported by Thokozile *et al*.[38] for identification of the most descriptive location in discriminating the genotypes with most representative environment.

Conclusion

Results obtained from physiological attributes and PCA analysis represented that in wheat crop, for water deficit stress, WH 1080 genotype appears to be best suited; and for salinity stress, KRL 283, KRL 330, KRL 210 and Kharchia 65 are the best. For interactive water deficit nd salinity stress, the genotype KRL 433 proved to be the best which could be further used by the breeders specifically for developing multiple abiotic stress tolerant genotypes conferring tolerance to these stresses.

Conflict of Interest

Authors declare no competing interests.

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