



Impact of water deficit (salt and drought) stress on physiological, biochemical and yield attributes on wheat (*Triticum aestivum*) varieties

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ABSTRACT

This investigation was carried out to assess the combined effect of salt stress and drought on physiological and biochemical traits in wheat (*Triticum aestivum* L.) varieties so as to deduce the probable mechanism of stress tolerance in these varieties. Two salt tolerant (Kharchia 65 and KRL 210) and two salt sensitive (HD 2851 and HD 2009) wheat varieties were subjected to salt (50 mM and 100 mM NaCl) and drought (25% and 50% water deficit) stresses alone as well as in combination to study the interactive effects of salt × drought stresses under pot house conditions at seedling stage. The data recorded on physiological (membrane injury, relative water content, chlorophyll content, Na⁺, K⁺, Cl⁻ and K⁺/Na⁺), biochemical (total soluble sugars and proline) and yield (number of tillers, productive tillers, biomass, and 1000 seed weight) attributes indicated the effect of salt and drought stresses in the studied varieties. These stresses caused a significant decline in chlorophyll content, K⁺ content, number of tillers, number of productive tillers, biomass and yield in sensitive varieties, while the tolerant ones were least affected. The sensitive varieties HD 2851 and HD 2009 accumulated the highest Na⁺ and Cl⁻ content, whereas Kharchia 65 (tolerant) accumulated the lowest amount of these ions. All the varieties, however, showed increased accumulation of compatible solutes (total soluble sugars and proline) with inclined stress which helped in osmotic adjustment. Consistent and significant reductions were observed in number of tillers as well as in productive tillers with every successive increase in salt and drought stresses. Yield in terms of 1000 seed weight followed the trend: 34.51 g in Kharchia 65 > 28.52 g in KRL 210 > 24.89 g in HD 2009 > 24.55 g in HD 2851.

Key words: Drought, Salinity, Wheat varieties, Yield attributes

Global food security of the expanding world population is increasingly threatened by environmental stresses that limit the growth and yield of major food crops. Wheat (*Triticum aestivum* L.; family Poaceae) is the most important cereal crop for the majority of world's population and is the staple food for about two billion people (36% of the world population). Globally, wheat is the second most produced cereal crop after maize (Datta *et al.* 2009). Soil water deficit and salinization are the two most limiting abiotic stresses that adversely affect the production of the world's major agricultural crops (Munns 2011). This situation highlights the need for focussed research to develop multiple stress tolerant crop genotypes that can grow and yield better with

less water, particularly in regions with limited water, land resources and insufficient water use. This situation is more critical in many arid and semi-arid regions of the world where serious water shortages are affecting irrigated crop production (Feres and Soriano 2007).

Water deficit caused by drought and osmotic stresses induces changes in morphology, water status, gas exchange and chlorophyll content which are connected with the onset of protective mechanisms in the plant. In order to stabilize crop production under salt and water stress conditions, an elaborate understanding of crop's physiological and biochemical responses is required which would be helpful in devising appropriate agronomic manipulations.

Salt and drought stresses are known to suppress plant growth and development, and cause membrane leakage, ion imbalance or disequilibrium in different crops (Kumar *et al.* 2016). Generally, the plants accumulate different organic and inorganic solutes in the cytosol to raise osmotic pressure and thereby maintain both turgor and the driving gradient for water uptake (Patade *et al.* 2008). Osmotic regulation through accumulation of low molecular weight organic solutes is an important strategy to avoid osmotic stress. The accumulation of metabolites such as glycine

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betaine, proline, mannitol and soluble sugars maintains the inflammation pressure and cell volume (Houimli *et al.* 2010). Proline and soluble sugars have been shown to increase under water stress and are potentially important contributors to osmotic adjustment. Proline, which is widely found in higher plants, accumulates in stressed plants in larger amounts than other amino acids (Kumar *et al.* 2015). Proline is a strong source to store carbon, nitrogen and a purifier of free radicals. Proline also maintains the structure of cell membrane and proteins and contributes to membrane stability. It may also act as a signalling regulatory molecule able to activate multiple responses that are components of the adaptation process (Maggio *et al.* 2002). So, to identify the genotypes/varieties performing well even under stress conditions as well as understanding the physiological and biochemical traits influencing plant's growth in stress environments (drought/salinity) is still a major challenge for plant biologists to identify target traits in early stages of crop growth.

MATERIALS AND METHODS

The present study was carried out at Crop Improvement Division, ICAR - Central Soil Salinity Research Institute (CSSRI), Karnal, Haryana, India. To evaluate the response of wheat varieties to salt stress coupled with drought (water deficit; WD), a randomized complete block design experiment with 5 replications was conducted in 20 kg capacity clay/porcelain pots during 2013-14 and 2014-15. The net house was covered with a high quality transparent polythene sheet to avoid the rain water entry and to maintain the desired salinity and water deficit stress in the pots as per treatments. Surface sterilized seeds of Kharchia 65, KRL 210, HD 2851 and HD 2009 were sown in pots. Plants were supplied with Hoagland nutrient solution before imposition of treatments (Hoagland and Arnon 1950). Osmotic stress was imposed 3 weeks after sowing of seeds by application of 50 and 100 mM concentration of sodium chloride (NaCl) solution in water and drought stress by withholding water. Drought stress was created by gradually decreasing the water supply to the pots. Drought treatments were imposed by 100% saturation of soil in pots first and then withholding irrigation supply till the depletion of water to 25 and 50% of water in soil (25 and 50% water scarcity).

Water deficit: 20 kg capacity clay/porcelain pots filled with 16 kg soil (field capacity 28% v/v) at bulk density of 1.45 g/cc which have porosity approximately 40%. Initially 6.5 l water (up to field capacity) was given in the pots at weekly interval and evaporation was noted through pan. During the entire study period pan evaporation was 2-3 mm/day, i.e. 21 mm/week. On this basis, 25 and 50% water deficit treatments were induced.

Salt treatment: Based on the water requirement of the pots, 50 mM NaCl and 100 mM NaCl concentration was given to pots at regular weekly interval. At final harvest, 50 mM NaCl concentration pots have mean salinity level about 4.2 d/Sm and 100 mM NaCl pots have 9.1 dS/m.

Growth, physiological and biochemical parameters were

studied 15 days after the imposition of stress treatments. Fully expanded (flag leaf) leaves were sampled for measurement. Membrane injury was estimated according to the method of Dionisio-Sese and Tobita (1998). Relative water content (RWC) was measured following the procedure described by Weatherley (1950). The chlorophyll content was determined using DMSO (Dimethyl sulphoxide) as described by Hiscox and Israelstam (1979). Freshly harvested plants were weighed and analyzed for total soluble sugars (Yemm and Wills 1954) and proline (Bates *et al.* 1973). For Na⁺ and K⁺ content, 100 mg of oven dried and well ground plant material was digested with 10 ml of HNO₃ : HClO₄ (3:1) di-acid mixture and readings were taken with flame photometer using standard NaCl and KCl. Chloride content of leaves was determined volumetrically by modified method of Chhabra (1973).

All the data were subjected to variance analysis using the SAS (Version 9.3, SAS Institute Inc., Cary, NC, USA). Least significant difference test was applied at 5 per cent probability level to compare the mean differences. Correlation analysis was performed to determine the relationship between the traits using the Pearson's correlation coefficient.

RESULTS AND DISCUSSION

The present investigation was carried with four varieties of wheat (Kharchia 65, KRL 210, HD 2851 and HD 2009) differing in salt tolerance for estimating their response to NaCl treatment and its interaction with water stress. Varieties were selected on their comparative tolerance towards salt stress. Analysis of variance showed significant differences among varieties and treatments in 1% probability for membrane injury, relative water content, total chlorophyll, total soluble sugars, proline content, Na⁺, K⁺, K⁺/Na⁺, Cl⁻ content, number of tillers, number of productive tillers, biomass and 1000 seed weight which demonstrated existence of variation among different treatments.

Perusal of the physiological and biochemical data (Table 1) indicated significant differences among the varieties and among the treatments with respect to different parameters. The changes in membrane injury, relative water content and chlorophyll content were proportional to the increase in magnitude of stress conditions. The highest rate of electrolyte leakage (an indicator of membrane damage) was observed in variety HD 2009 (61.47%) at 100 mM NaCl + 50%WD (Table 2) while the minimum leakage occurred in Kharchia 65 (49.22%). The changes in plasma membrane permeability (electrolyte leakage) are primarily controlled by the membrane transport proteins (Serrano *et al.* 1999, Jacobs *et al.* 2011). These changes in permeability and leakage under stress conditions are therefore linked with the modifications in protein and lipid matrix of the plasma membrane. It has been shown that the extent of membrane injury is more pronounced in salt sensitive cultivars as compared to the salt tolerant ones and it might be maintained *via* inherited or induced membrane protection mechanisms under salt stress (Mansour 2013). The decrease in relative

water content (RWC) could be attributed towards early symptoms of stress conditions. Table 1 showed the maximum (66.85%) reduction in RWC was recorded in HD 2009 followed by HD 2851 (64.32%), KRL 210 (53.42%) and the minimum in Kharchia 65 (51.37%). This reduction of

RWC in stressed plants might be associated with a decrease in plant vigour (Halder and Burrage 2003). Mationn *et al.* (1989) reported a similar reduction in RWC in tolerant and sensitive cultivars of barley.

Increasing salt and water deficit stresses significantly

Table 1 Morpho-physiological characterization of different wheat varieties using membrane injury, relative water content, total chlorophyll, yield attributes subjected to drought and salt stress

Treatment	Membrane injury (%)	RWC (%)	Chlorophyll concentration ($\mu\text{g/ml}$)	NT/5 plant	NPT/5 plant	Biomass (g/plant)	1000 seed wt (g)
<i>Kharchia 65 (Salt tolerant wheat variety)</i>							
Control	16.59 ^h	88.85 ^a	37.38 ^a	17.67 ^a	17.08 ^a	25.00 ^a	42.16 ^a
25% Water deficit (WD)	25.93 ^f	70.46 ^c	31.28 ^b	15.42 ^c	14.83 ^c	20.80 ^{bc}	38.17 ^c
50% Water deficit (WD)	34.80 ^c	54.21 ^e	28.71 ^c	14.17 ^d	13.42 ^d	19.15 ^d	30.34 ^g
50 mM NaCl	19.38 ^g	74.44 ^b	29.75 ^{bc}	16.21 ^b	15.79 ^b	23.80 ^a	40.55 ^b
100 mM NaCl	29.69 ^e	62.47 ^d	26.76 ^d	14.25 ^d	11.75 ^e	19.65 ^{cd}	34.11 ^e
50 mM NaCl + 25% WD	31.51 ^d	59.93 ^d	24.87 ^e	11.42 ^e	10.42 ^f	21.70 ^b	35.28 ^d
100 mM NaCl + 25% WD	36.06 ^c	50.30 ^f	21.31 ^f	9.79 ^f	9.25 ^g	19.10 ^d	31.93 ^f
50 mM NaCl + 50% WD	41.38 ^b	54.14 ^e	19.60 ^g	9.00 ^g	7.17 ^h	17.45 ^e	29.40 ^h
100 mM NaCl + 50% WD	49.22 ^a	43.21 ^g	16.92 ^h	8.58 ^g	6.42 ⁱ	16.46 ^e	28.63 ⁱ
General mean	31.62	62.00	26.29	12.94	11.79	20.35	34.51
LSD (P=0.05)	1.8129	2.7998	1.692	0.5129	0.6428	1.3104	0.6539
<i>KRL 210 (Salt tolerant wheat variety)</i>							
Control	19.79 ⁱ	86.04 ^a	37.44 ^a	17.50 ^a	17.50 ^a	24.50 ^a	35.23 ^a
25% Water deficit (WD)	27.69 ^g	69.81 ^b	32.97 ^b	14.58 ^b	13.58 ^b	19.90 ^b	31.93 ^b
50% Water deficit (WD)	35.66 ^e	48.01 ^e	28.62 ^c	13.50 ^c	11.83 ^c	16.65 ^d	29.58 ^c
50 mM NaCl	25.13 ^h	70.03 ^b	32.06 ^b	14.42 ^b	13.58 ^b	24.80 ^a	29.30 ^c
100 mM NaCl	33.72 ^f	58.82 ^c	27.35 ^c	11.83 ^d	10.42 ^d	18.45 ^{bc}	27.43 ^d
50 mM NaCl + 25% WD	40.01 ^d	53.89 ^d	23.05 ^d	10.42 ^e	9.83 ^e	19.50 ^b	30.38 ^c
100 mM NaCl + 25% WD	43.23 ^c	49.04 ^e	19.74 ^e	8.83 ^f	8.83 ^f	16.90 ^{cd}	25.35 ^e
50 mM NaCl + 50% WD	49.37 ^b	48.00 ^e	16.60 ^f	7.83 ^g	5.83 ^g	14.80 ^e	25.05 ^e
100 mM NaCl + 50% WD	53.91 ^a	40.08 ^f	13.36 ^g	7.42 ^g	5.42 ^g	13.70 ^e	22.45 ^f
General mean	36.50	58.19	25.69	11.81	10.76	18.80	28.52
LSD (P=0.05)	1.5499	3.0101	1.4003	0.6341	0.5621	1.6349	1.084
<i>HD 2851 (Salt sensitive wheat variety)</i>							
Control	19.35 ^h	86.58 ^a	37.75 ^a	16.17 ^a	15.92 ^a	24.60 ^a	37.45 ^a
25% Water deficit (WD)	31.32 ^g	71.27 ^b	31.15 ^b	13.50 ^c	12.25 ^c	18.10 ^c	28.68 ^c
50% Water deficit (WD)	44.68 ^d	50.86 ^d	29.13 ^b	13.08 ^c	11.75 ^c	16.05 ^d	21.68 ^e
50 mM NaCl	19.55 ^h	69.05 ^b	30.35 ^b	14.83 ^b	13.50 ^b	21.90 ^b	30.68 ^b
100 mM NaCl	35.75 ^f	55.49 ^c	24.28 ^c	10.67 ^d	9.83 ^d	16.30 ^d	21.70 ^e
50 mM NaCl + 25% WD	42.34 ^e	50.97 ^d	22.18 ^c	9.42 ^e	8.42 ^e	16.10 ^d	23.04 ^d
100 mM NaCl + 25% WD	47.75 ^c	44.06 ^e	16.78 ^d	8.17 ^f	7.17 ^f	13.20 ^e	21.18 ^e
50 mM NaCl + 50% WD	54.08 ^b	35.21 ^f	13.55 ^e	6.58 ^g	6.06 ^g	11.80 ^{ef}	19.83 ^f
100 mM NaCl + 50% WD	59.84 ^a	30.89 ^g	10.76 ^f	5.33 ^h	4.75 ^h	10.60 ^f	16.73 ^g
General mean	39.41	54.93	23.99	10.86	9.96	16.52	24.55
LSD (P=0.05)	1.3376	3.5905	2.2001	0.9512	0.9457	1.4319	0.728
<i>HD 2009 (Salt sensitive wheat variety)</i>							
Control	17.85 ^h	86.99 ^a	36.73 ^a	16.25 ^a	16.08 ^a	24.20 ^a	44.38 ^a

Contd.

Table 1. (Concluded)

Treatment	Membrane injury (%)	RWC (%)	Chlorophyll concentration (µg/ml)	NT/5 plant	NPT/5 plant	Biomass (g/plant)	1000 seed wt (g)
25% Water deficit (WD)	32.74 ^f	61.99 ^b	31.31 ^b	12.96 ^b	12.67 ^b	16.95 ^c	34.55 ^b
50% Water deficit (WD)	44.62 ^d	45.96 ^d	28.42 ^c	11.83 ^c	11.42 ^c	13.20 ^d	28.83 ^c
50 mM NaCl	23.84 ^g	63.70 ^b	29.52 ^c	13.42 ^b	12.58 ^b	20.05 ^b	28.18 ^c
100 mM NaCl	38.07 ^e	52.27 ^c	25.16 ^d	9.58 ^d	8.83 ^d	15.60 ^c	21.68 ^d
50 mM NaCl + 25% WD	45.29 ^{cd}	49.77 ^c	21.19 ^e	8.75 ^d	7.58 ^e	16.10 ^c	28.75 ^c
100 mM NaCl + 25% WD	49.15 ^c	42.84 ^d	16.80 ^f	7.58 ^e	6.71 ^f	12.60 ^d	15.20 ^e
50 mM NaCl + 50% WD	55.27 ^b	35.04 ^e	13.57 ^g	6.33 ^f	5.54 ^g	11.85 ^{de}	11.83 ^f
100 mM NaCl + 50% WD	61.47 ^a	28.84 ^f	9.89 ^h	5.24 ^g	4.42 ^h	10.50 ^e	10.63 ^f
General mean	40.92	51.93	23.62	10.22	9.54	15.67	24.89
LSD (P=0.05)	3.9828	3.2746	1.356	0.9014	0.6228	1.6926	1.5974

Means with at least one letter common are not statistically significant (P <0.05) using DUNCAN's Multiple Range Test

reduced chlorophyll concentration in all the varieties as compared to control. The maximum (73.07%) reduction was observed in HD 2009 followed by HD 2851 (71.5%), KRL 210 (64.32%) and Kharchia 65 (54.74%) when plants were exposed to 100 mM NaCl + 50% WD. The salt and water deficit stresses alone caused lesser reductions in chlorophyll concentration but the combined effects of both aggravated stress symptoms with greater reductions noted in chlorophyll concentration (Table 1). At 100 mM NaCl + 50% WD treatment, Kharchia 65 had the maximum (16.92 µg/ml) chlorophyll whereas the minimum (9.89 µg/ml) concentration was recorded in HD 2009. The corresponding decrease in chlorophyll, concentration with increasing stress conditions implies a lower capacity of leaf tissues for light harvesting and production of reactive oxygen species which is mainly driven by excess energy absorption in the photosynthetic apparatus; this might be avoided by degrading the absorbing pigments (Herbinger *et al.* 2002,

Kumar *et al.* 2016).

The accumulation of compatible solutes such as proline, soluble sugars (key osmolytes that contribute to osmotic adjustment) etc. which are non-toxic at higher concentrations to cytoplasmic functions allow additional water uptake from the environment and turgor maintenance (Sairam *et al.* 2002, Misra and Gupta 2005). Data presented in Fig 1 vividly evinced that flag leaves of wheat variety HD 2851 (43.88 mg/g DW) possessed comparatively higher total soluble sugars over the rest of tested varieties. Total soluble sugars significantly increased with the increase in salt and drought stress regardless of the NaCl concentration and water deficit in comparison to unstressed plants. At higher stress condition (100 mM NaCl +50% WD), there was 4.71 fold increased accumulation of total soluble sugars in Kharchia 65, 4.21 fold in KRL 210, 5.15 fold in HD 2851 and 6.23 fold in HD 2009 over their respective control (Fig 1). Soluble sugars act as potential osmoregulators. Elevated

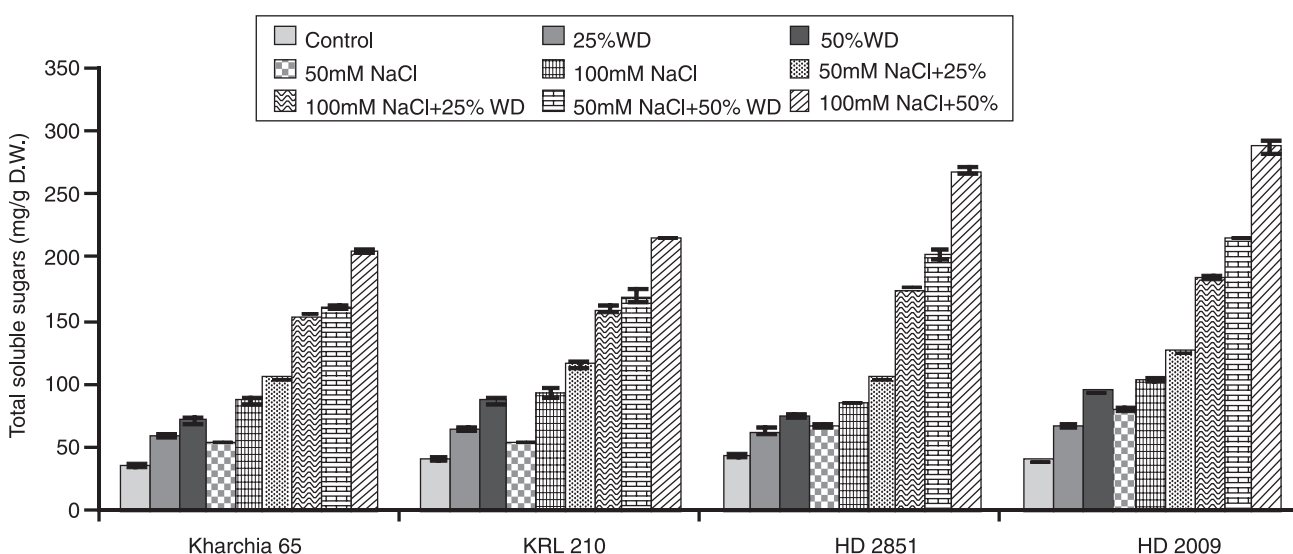


Fig 1 Effect of water deficit (salt and drought) stress on total soluble sugars (mg/g DW) in different wheat varieties.

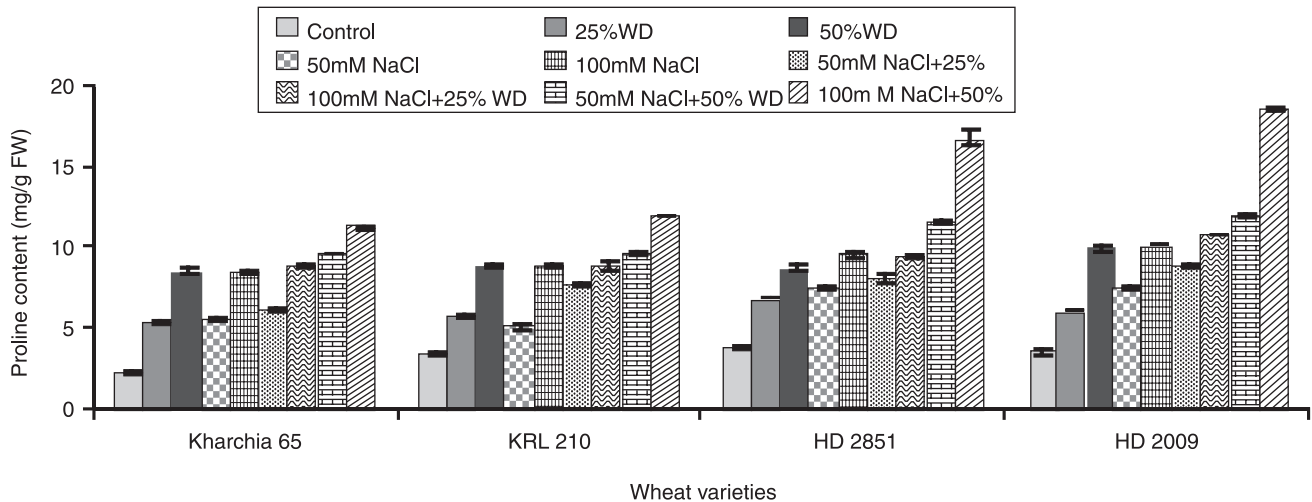


Fig 2 Effect of water deficit (salt and drought) stress on proline content (mg/g FW) in different wheat varieties.

sugar levels under stress conditions might contribute towards turgor maintenance (Kumar *et al.* 2015).

Proline accumulation is an important physiological index for plant response to abiotic stresses. Among different wheat varieties, highest proline content was recorded in HD 2851 and minimum in Kharchia 65 under non-stress conditions (Fig 2). It increased with increasing level of salt and drought stress (either alone or in combination) in all the varieties. At highest level (100 mM NaCl + 50% WD) of stress, the maximum (80.52%) proline accumulation was noted in HD 2009 and minimum (70.13%) in KRL 210 as compared to their control plants. Wheat variety HD 2009 showed the highest (9.81 mg/g FW) while Kharchia 65 showed the lowest (7.46 mg/g FW) mean value for proline content (Fig 2). Proline is a potent osmoregulator molecule and counteracts the adverse effects of toxic salt ions in cell vacuoles, contributes to membrane stability and mitigates the effect of NaCl on cell disruption. Accumulation of these osmolytes can also improve plant tolerance by protecting and stabilizing membranes and enzymes during stress conditions (Ashraf 2004, Lata *et al.* 2017).

A survey of available literature also suggests that in addition to accumulation of compatible organic solutes, an enhancement in specific inorganic ions and K^+/Na^+ discrimination under stress conditions constitutes an important mechanism of plant tolerance (Flowers *et al.* 1977). It is clearly shown from the results that all the varieties showed differing responses to salt stress coupled with drought for the accumulation of ions. Sodium (Na^+) and chloride (Cl^-) content increased in all the varieties. However, the sensitive varieties HD 2851 and HD 2009 accumulated the highest mean Na^+ (0.63 and 0.68%, respectively), whereas the tolerant Kharchia 65 accumulated the lowest (0.4%) amount of Na^+ . At 100 mM NaCl + 50% WD, HD 2851 and HD 2009 had significantly higher Na^+ content (1.51 and 1.55% DW, respectively), whereas Kharchia 65 had significantly lower (1.14% DW) Na^+ content (Fig 3). A similar increase was also noted for Cl^- ions (Fig 4). Kharchia

65 and KRL 210 maintained significantly lower (4.54 and 4.55% DW, respectively) Cl^- over all the treatments. At highest level of salt and water deficit stress, HD 2009 had the lowest (5.5% DW) Cl^- content while KRL 210 accumulated highest (6.72% DW) Cl^- content. Excessive accumulation of Na^+ and Cl^- in the leaves has been considered highly harmful for normal metabolism of plants, and tolerant genotypes have the capacity for successful salt exclusion. Some plants accumulate Na^+ and Cl^- for osmotic adjustment, which is equally damaging to plant growth due to ionic toxicity and nutrient imbalance (Asha 2012). As shown in Fig 5 and 6, the leaf K^+ concentration as well as K^+/Na^+ declined significantly ($P < 0.01$) under NaCl and WD induced osmotic stress. K^+ and Na^+ are known to be the major inorganic components, which provide necessary osmotic potential for water uptake by plant cells (Tejera *et al.* 2006). At the highest stress conditions, K^+ concentration decreased from 1.09% to 0.39% in Kharchia 65, from 1.32% DW to 0.34% DW in KRL 210, from 1.29% DW to 0.34% DW in HD 2851 and from 1.14% DW to 0.28% DW in HD 2009 in comparison to the control (Fig 5). The decrease in K^+ contents was due to the presence of excessive Na^+ in growth medium because high external Na^+ content is known to have an antagonistic effect on K^+ uptake in plants (Sarwar and Ashraf 2003). Jampeetong and Brix (2009) also reported that Na^+ competes with K^+ for uptake into cells, particularly when the external concentrations of Na^+ were substantially higher than that of K^+ , and the ability to maintain Na^+/K^+ homeostasis in the cell was crucial for the tolerance of plants. There was a large decrease in K^+/Na^+ ratio under stress compared to control, however, varieties showed variation in the decrease of K^+/Na^+ ratio in response to applied stress (Fig 6). At higher combined stress (100 mM NaCl + 50% WD), K^+/Na^+ ratio followed the trend as follows; Kharchia 65 (0.34) > KRL 210 (0.26) > HD 2851 (0.23) > HD 2009 (0.18). Regulation of K^+ uptake and prevention of Na^+ entry, efflux of Na^+ from the cell and sequestration of Na^+ into vacuole for osmotic adjustment are common strategies for maintenance of desirable K^+

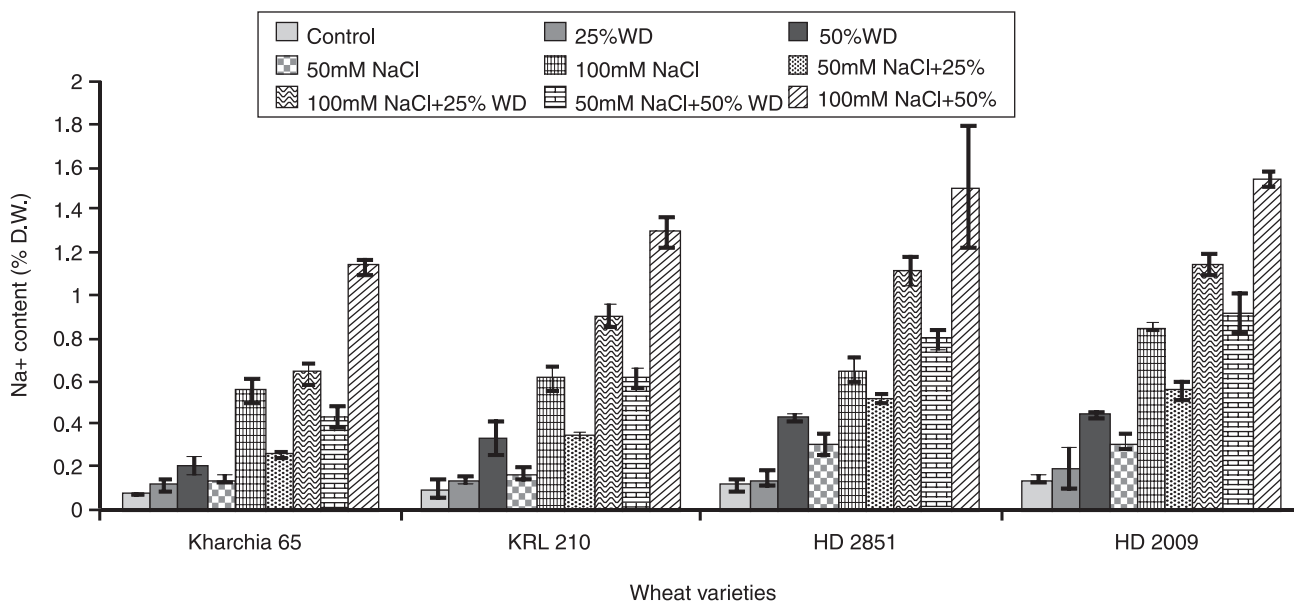


Fig 3 Effect of water deficit (salt and drought) stress on Na⁺ content (% DW) in different wheat varieties.

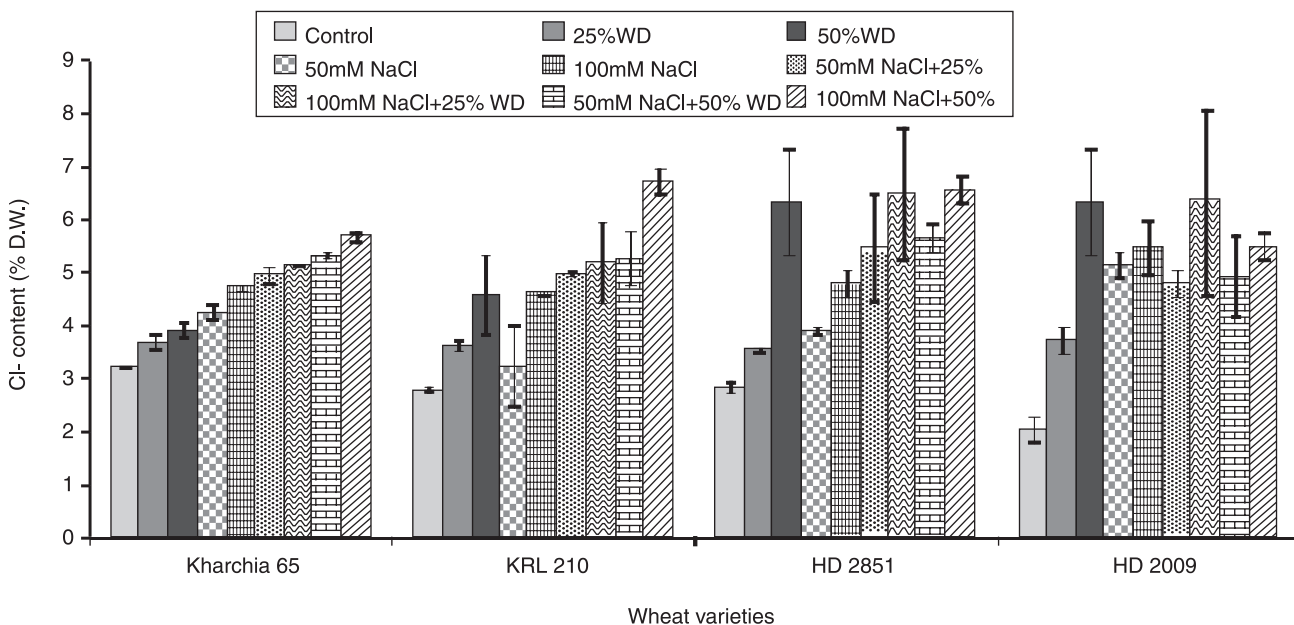


Fig 4 Effect of water deficit (salt and drought) stress on Cl⁻ content (% DW) in different wheat varieties.

Na⁺ ratio in the cytosol (Asha 2012).

There was a consistent and significant reduction ($P < 0.01$) in number of tillers as well as in number of productive tillers with each successive increase in NaCl and WD stresses. Regarding the number of tillers, salt tolerant variety Kharchia 65 showed lesser reduction (51.44%) than other studied varieties while significantly higher reductions were noted for salt sensitive varieties (67.04% in HD 2851 and 67.75% in HD 2009) at 100 mM NaCl + 50% WD in comparison with non stressed plants (Table 1). Contrary to number of tillers, much higher reductions were noted in number of productive tillers. Increasing stress conditions increased reduction in productive tillers, i.e. 62.04% in Kharchia 65, 69.03% in KRL 210, 70.16% in HD 2851 and 72.51% in HD 2009. Stress conditions might inhibit tiller

emergence, formation and could cause their abortion at later stages (Nicolas *et al.* 1993). Number of productive tillers depends on the environmental conditions present during tiller bud initiation and subsequent development stages. It has been suggested that tiller emergence, abortion, or both are affected by limited soil water (Davidson and Chevalier 1990), nutrient deficiencies (Masle 1985), and salt stress (Nicolas *et al.* 1993).

The major inhibitory effect of salt stress along with water deficit on the plant growth are due to osmotic shock, ion toxicity and nutritional imbalance leading to reduced photosynthetic activity and other physiological abnormalities (Ali and Awan 2004). These factors might be involved in sodium uptake through root and its subsequent distribution in different vegetative and floral parts. Consequently, it

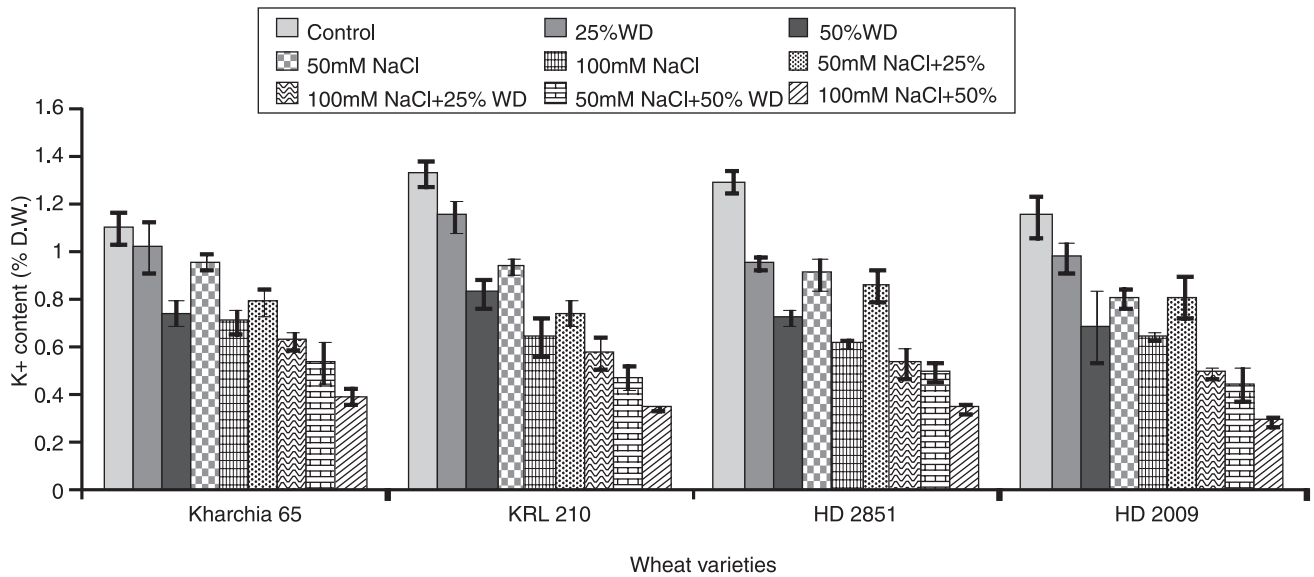


Fig 5 Effect of water deficit (salt and drought) stress on K⁺ content (% DW) in different wheat varieties.

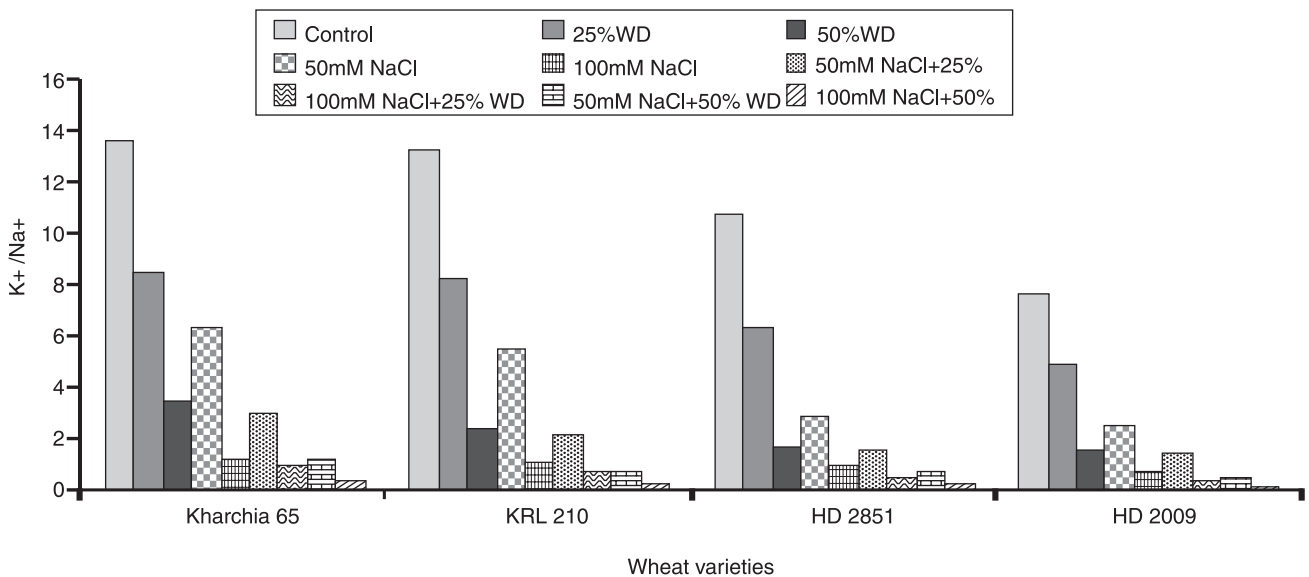


Fig 6 Effect of water deficit (salt and drought) stress on K⁺/Na⁺ in different wheat varieties.

adversely affects the transportation of total assimilates to the growing regions (Munns 2002). Data presented in Table 1 showed that Kharchia 65 accumulated the highest (25.0 g) biomass followed by variety HD 2851 (24.6 g), KRL 210 (24.5 g) while the least biomass accumulation was noted in variety HD 2009 (24.2 g) under non-stress conditions. With increasing stress, reductions were noted in biomass accumulation for all the studied varieties. Significantly higher (56.91%) reductions were observed in HD 2851 followed by HD 2009 (56.61%), KRL 210 (44.08%) and Kharchia 65 (34.16%). The plants produce maximum biomass under adequate water supply, whereas water deficit causes a marked decrease in plant biomass production (Clarke *et al.* 1991 and Ashraf 1998). Yield depends on the ability of the crops to assimilate and utilize the resources and, thus, is the interplay of many components contributing

towards final harvest. Significant variation in yield (1000 seed weight) of wheat varieties was noticed in evaluated crop growing seasons (Table 1). Mean 1000 seed weight of wheat varieties followed the trend: 34.51 g in Kharchia 65 > 28.52 g in KRL 210 > 24.89 g in HD 2009 > 24.55 g in HD 2851. Stress treatments had significant effect on 1000 seed weight. Stresses reduced yield by reducing the number of filled grains per panicle. Reduction in seed weight may possibly be due to decreased pollen viability or decreased receptivity of the stigmatic surface or both (Abdullah *et al.* 2001). In comparison to control, at 100 mM NaCl + 50% WD stress treatment 1000 seed weight varied significantly, i.e. from 42.16 g to 28.63 g in Kh 65, from 35.23 g to 22.45 g in KRL 210, from 37.45 g to 16.73 g in HD 2851 and from 44.38 g to 10.63 g in HD 2009 (Table 2). Under salt stress and water deficit conditions, reduction in yield and its components might be attributed to low production,

Table 2 Pearson's correlation coefficients for association of different wheat varieties among membrane injury, relative water content, total chlorophyll, total soluble sugars, proline, Na⁺, K⁺, K⁺/Na⁺, Cl⁻ content, Yield attributes subjected to drought and salt stress

Trait	Membrane Injury	RWC	Chlorophyll content	Total soluble sugars	Proline content	Na ⁺ content	K ⁺ content	K ⁺ /Na ⁺	Cl ⁻ content	NT	NPT	Biomass	1000 seed wt
Membrane injury (%)	1	-0.948**	-0.920**	0.903**	0.892**	0.852**	-0.871**	-0.769**	0.812**	-0.936**	-0.934**	-0.934**	-0.848**
RWC (%)		1	0.916**	-0.868**	-0.914**	-0.825**	0.907**	0.873**	-0.850**	0.915**	0.916**	0.922**	0.846**
Chl content (µg/ml)			1	-0.959**	-0.884**	-0.887**	0.921**	0.805**	-0.791**	0.962**	0.965**	0.840**	0.796**
TSS (mg/g)				1	0.899**	0.917**	-0.877**	-0.690**	0.722**	-0.927**	-0.921**	-0.811**	-0.770**
Proline (mg/g)					1	0.891**	-0.887**	-0.785**	0.756**	-0.867**	-0.867**	-0.878**	-0.848**
Na ⁺ content (%)						1	-0.864**	-0.710**	0.760**	-0.868**	-0.867**	-0.808**	-0.799**
K ⁺ content (%)							1	0.879**	-0.831**	0.884**	0.909**	0.818**	0.767**
K ⁺ /Na ⁺								1	-0.811**	0.817**	0.847**	0.764**	0.713**
Cl ⁻ content (%)									1	-0.782**	-0.800**	-0.804**	-0.716**
NT										1	0.987**	0.876**	0.833**
NPT											1	0.867**	0.822**
Biomass (g)												1	0.862**
1000 seed wt (g)													1

P* < 0.05; P** < 0.01; ns: Non - significant, RWC: relative water content; NT: number of tillers; NPT: number of productive tillers.

senescence and physiologically less active green foliage (Wahid *et al.* 1997) and the reduction in photosynthetic rate might be a supplementary effect.

Results from correlation study showed significant interaction among different traits in all the varieties. Highest significant and positive correlation was observed between number of tillers and productive tillers (r = 0.987**). Significant and positive correlation with these traits also exhibited by chlorophyll content (r = 0.962** and 0.965**), RWC (r = 0.915** and 0.916**), K⁺ content (r = 0.881** and 0.909**) and K⁺/Na⁺ (r = 0.817** and 0.847**) showing that at higher stress, decrease in chlorophyll content, RWC, K⁺ content and K⁺/Na⁺ cause breakdown of photosynthetic machinery, which leads to decrease in number of tillers and productive tillers and ultimately yield. Significant and negative correlation with number of tillers and productive tillers showed by membrane injury (r = -0.936** and -0.934**), TSS (r = -0.927** and -0.921**), proline content (r = -0.867**), Na⁺ (r = -0.982** and -0.977**) and Cl⁻ (r = -0.782** and -0.800**) revealed that, these biochemical parameters increased with increasing salt and drought stress to compensate yield loss due to decrease in number of tillers and productive tillers (Table 1).

Conclusions

Water deficit causes many adverse effects on plant growth, which is due to low osmotic potential of soil solution (osmotic stress), specific ion effects (salt stress), nutritional imbalance or a combination of these factors. All these factors affect plant growth and development by impairing physiological and biochemical activities which ultimately leads to reduction in grain yield. In present experiment, salt stress coupled with drought caused more damage in salt sensitive (HD 2009) wheat varieties as evidenced by the lower RWC, chlorophyll content, K⁺/Na⁺, NT, NPT and yield.

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