Pak. J. Bot., 44(5): 1533-1541, 2012.

DETECTING DIFFERENCES IN WHEAT FOR SALT TOLERANCE THROUGH MULTI PARAMETERS EVALUATION-II: PHYSIOLOGICAL PARAMETERS

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Abstract

A pot experiment was conducted in a randomized complete block design using 3 wheat genotypes comprising salt sensitive cultivar Inqlab (V1), a known salt tolerant line N-1073 (V2), a newly developed wheat genotype N-9760 (V3) and 4 salinity levels of 1.5 (control), 5, 10, and 15 dS m^{-1} developed in irrigation water. The objectives were to examine i) variation in accumulation of different ions and different attributes of chlorophyll fluorescence on fully developed flag leaf and ii) possible relationship of quantum yield of chlorophyll fluorescence (F_v/F_m) with various ions and grain yield at maturity stage. Concentrations of Ca^{2+} in different genotypes differed significantly under control and various salinity levels in both root and shoot especially under 15 dS m^{-1} ,Cl^- concentrations in roots and Na^+ in shoots and roots increased while K^+ reduced significantly in both shoots and roots under increasing salinity levels. Line N-1073 restricted the movement of Cl^- from root to shoot but not of Na^+ under 15 dS m^{-1} .Salt stress significantly reduced K^+/Na^+ ratio in the shoot and root with a maximum reduction observed in N-1073 (V2) under EC 15 dS m^{-1}. Ca^{2+}/Na^+ ratio also reduced significantly under salt stress especially in Inqlab (V1). All the four chlorophyll fluorescence parameters did not show any effect of salinity on N-1073 (V2), however, F_0 and F_v in V3 and V1 and F_v/F_m in V3 were significantly affected. In V2, the value of F_v/F_m was 0.82 at EC 15 dS m^{-1} but for V3, it was only was 0.28. The relationship of F_v/F_m with various ion concentrations and grain yield was different for all the three genotypes. Categorization of germplasm on the basis of increasing tolerance was of the order of V1, V2 and V3. Possible underlying reasons have been discussed in detail.

Introduction

Soil salinity is a major cause of reducing crop productivity and damaging the land beyond economic repair (Munns *et al.*, 2006; Ashraf, 2009; Saleem *et al*., 2011). The most devastating effect of high salinity is imbalance in uptake of ions especially of K^+, Na^+, Ca^{2+}, and Cl^- which in turn alters osmotic potential, enzyme activation, membrane permeability and electrochemical potential of the plants (Epstein & Jefferies 1964; Grattan & Grieve 1999). Plant acquisition and utilization of necessary nutrients particularly K^+ and Ca^{2+} may impair under saline conditions which can cause ion deficiency. Increased Na^+ and Cl^- and decreased K^+ concentration in both roots and shoots have already been reported to affect the wheat crop significantly (Qureshi *et al.,* 1991; Akhtar *et al.,* 1994; Rashid *et al.,* 1999). High salinity can also cause severe ion toxicity and/or interactions of salts with mineral nutrition which may result in nutrient deficiencies (Viegas *et al.,* 2001; Ashraf, 2004; Munns *et al.,* 2006). It can also cause changes in ratios of $K^{+/}Na^+$ and Ca^{2+}/Na^+ which can further affect the growth and productivity of crop plants (Zhu *et al.,* 2001; Ashraf, 2004). Competition between Na^+ and Cl^- can induce nutrient deficiency which is one of the major factors responsible for reduced crop productivity under saline conditions (Hu & Schmidhalter 2005).

Documented evidence suggests that plants differ widely in the extent to which they accumulate ions, and tolerate various levels of salinity (Ashraf & McNeilly 1988; Glenn *et al.,* 1996). Thus, tolerance itself describes the adaptive ability of plants towards unfavorable environmental factors which in turn depends upon the capability and adaptive ability of the photosynthetic apparatus of crops growing under stressed environment (Ball & Anderson 1986; Shahbaz *et al*., 2011). Chlorophyll fluorescence reflects the photochemical activities of the photosynthetic apparatus which has been used for detecting tolerance to chilling, freezing, drought, and air pollution and for salinity tolerance screening (Netondo *et al.,* 2004). Since to cope with the salinity and increasing crop productivity, new and diverse salt tolerant genotypes especially of staple crops are being produced continuously, so it is imperative to test each and every newly developed salt tolerant genotype in order to have in-depth knowledge about their nutrient status, adaptive ability, and the factor playing important role in that adaptability so that the newly developed genotypes can be used effectively and their differences from the existing genotypes established clearly. In the present study, three different wheat genotypes were assessed under various salinity levels through the use of various physiological parameters including variability in ion uptake by root and shoot, various ions ratios, and chlorophyll fluorescence parameter. The objectives were to examine i) variation in accumulation of different ions and different attributes of chlorophyll fluorescence on fully developed flag leaf and ii) possible relationship of quantum yield of chlorophyll fluorescence (F_v/F_m) with various ions and grain yield at maturity stage. It was hoped that mechanism of salt tolerance of newly developed wheat genotype N-9760 (V3) and the evidence to discriminate it from V1 and V2 could be assessed accurately.

Materials and Methods

Plant material: Three wheat (*Triticum aestivum* L) genotypes used in this experiment consisted of a commercial and salt sensitive cultivar (Inqlab: V1), a salt tolerant line (N-1073: V2) produced earlier (Farooq *et al.,* 1992) through wide-hybridization and N 9760 (a new salt tolerant breeding line: V3). Uniform sized seeds were dusted with fungicide (Vitavax) and sown in plastic pots (22 cm internal diameter) containing 5kg soil. Four salinity levels comprising electrical conductivity (EC) of 1.5 (control) and 5, 10, and 15dSm^{-1} prepared by mixing appropriate levels of NaCl in irrigation water were used throughout the study. The experiment was conducted in randomized complete design with three replications: genotypes as main plots and salinity levels as subplots.

Ion uptake analysis: The dried and grounded material (0.1 g) of shoot and root was digested with sulfuric acid and hydrogen per oxide according to the method of Wolf (1982). Concentration of Na^+, K^+ and Ca^{2+} were determined with flame photometer (Jenway, PFP-7) and expressed on mg g^{-1} dry weight basis. A graded series of standards for Na^+, K^+ and Ca^{2+} were prepared and standard curves were drawn. The values of Na^+, K^+ and Ca^{2+} were compared with standard curves and total quantities were calculated and computed. For determining the Cl^- concentration, dried and grounded material (0.1 g) of shoot and root samples were extracted in 10 ml of deionized water and heated at 90^{o}C till the volume became half. The volume was maintained again with 10 ml of de-ionized water. Concentrations of Cl^- were determined with chloride meter (Jenway, PCLM-3).

Chlorophyll fluorescence measurement: The chlorophyll fluorescence parameters were measured through the use of Plant Efficiency Analyzer: PEA (Hansatech Instruments Ltd, Kings Lynn, U.K.). PEA is a compact portable instrument designed for measurement in the field and analysis of chlorophyll fluorescence induction by the high time resolution continuous excitation principle. These are time-dependent changes in fluorescence emission, which occur when a dark-adapted leaf is exposed to light. The selected leaf sample (flag leaf) was dark adapted over night with a clip provided with the instrument. Before taking measurement, PEA sensor unit was held over the clip followed by opening the shutter. A single button press activates the high intensity LED array within the sensor head providing a maximum light intensity of 3000 µmol m^{-2} s^{-1}, sufficient to achieve F_m in most of the samples. The parameters like F_0, F_v, and F_v/F_m were measured accordingly.

Data analysis

Data for various observations were analyzed statistically by adapting analysis of variance technique based on completely randomized design according to Steel & Torrie (1980).

Results

Statistical analysis of the data for the concentrations of different ions in shoot and root is presented in Tables 1 and 2, respectively. It appeared that in both shoots and roots, there existed a significant difference in the three wheat genotypes regarding Ca^{2+}, K^+ Cl^-, Ca^{2+}/Na^+ ratio while difference due to varieties in Na^+ (only in shoots), and K^+/Na^+ ratio was highly ($p< 0.01$) significant. Contrary to this, differences in all the ions due to various salinity levels were highly ($p< 0.001$) significant while interaction between wheat varieties and various salinity levels was only significant for Ca^{2+}/Na^+ ratio in shoots (Table 1) and not in roots (Table2).

Table 1. Analysis of variance of data for different ions in the shoots of three wheat genotypes after 73 days of growth in varying levels of salinity.

SOV	DF	Ca^{2+}	K^+	Na^+	Cl^-	Ca^{2+}/Na^+	K^+/Na^+
Main Effects							
Variety (V)	2	0.111*	25.1*	45.5 **	448.8 *	0.011 *	0.529 **
Salt (S)	3	21.9***	70.4***	142.9***	5417.2***	0.193 ***	2.2 ***
Interaction							
V x S	6	1.3 NS	4.1NS	5.6 NS	81.3 NS	0.007 *	0.069 NS
Error	24	2.27	7.80	8.02	108.45	.002	0.06
Total	35						

NS = Non-significant, * Significant at 5%, ** = Significant at 1% and *** = Significant at 0.1%

Table 2. Analysis of variance for different ions in the roots of three wheat genotypes after 73 days of growth in varying levels of salinity.

SOV	DF	Ca^{2+}	K^+	Na^+	Cl^-	Ca^{2+}/Na^+	K^+/Na^+
Main Effects							
Variety (V)	2	1.33 *	8.69 *	12.69 *	59.69 *	0.009 *	0.457 **
Salt (S)	3	4.91 ***	22.63 ***	70.546***	489.80 ***	0.44 ***	3.65 ***
Interaction							
V x S	6	4.74 NS	0.54 NS	1.21 NS	18.92 NS	0.013NS	0.158NS
Error	24	0.55	1.77	2.55	24.25	0.015	0.073
Total	35						

NS = Non-significant, * Significant at 5%, ** = Significant at 1%, and *** = Significant at 0.1%

In the roots of the genotypes growing under control conditions, concentration of Ca^{2+} was maximum in V3 and minimum in V1. Both the values reduced progressively and significantly with increasing salinity levels and almost in an identical pattern (Fig. 1a). However, in shoots of the control plants, maximum Ca^{2+} concentration appeared in V1 and minimum in V2. All salinity levels caused reduction in shoot Ca^{2+} concentration but the reduction was only significant for V1 (Fig. 1b).

The three genotypes growing under control conditions differed significantly ($p<0.001$) with respect to Cl^- concentrations in roots which is maximum in V3 and the minimum in V2. All salinity levels increased Cl^- concentrations but it is only significant in V1 and V2. A maximum increase appeared in V1 while in V2 and V3, the increase was comparatively less but the pattern of increase was nearly identical at EC 15 dS m^{-1} (Fig. 1c). Under control conditions, the concentration of Cl^- was significantly less in shoots of V1 and increased significantly under EC 15 dS m^{-1}. Compared to V1, concentration of Cl^- was significantly higher in V2 and V3 especially under EC 10 and 15 dS m^{-1}(Fig. 1d).

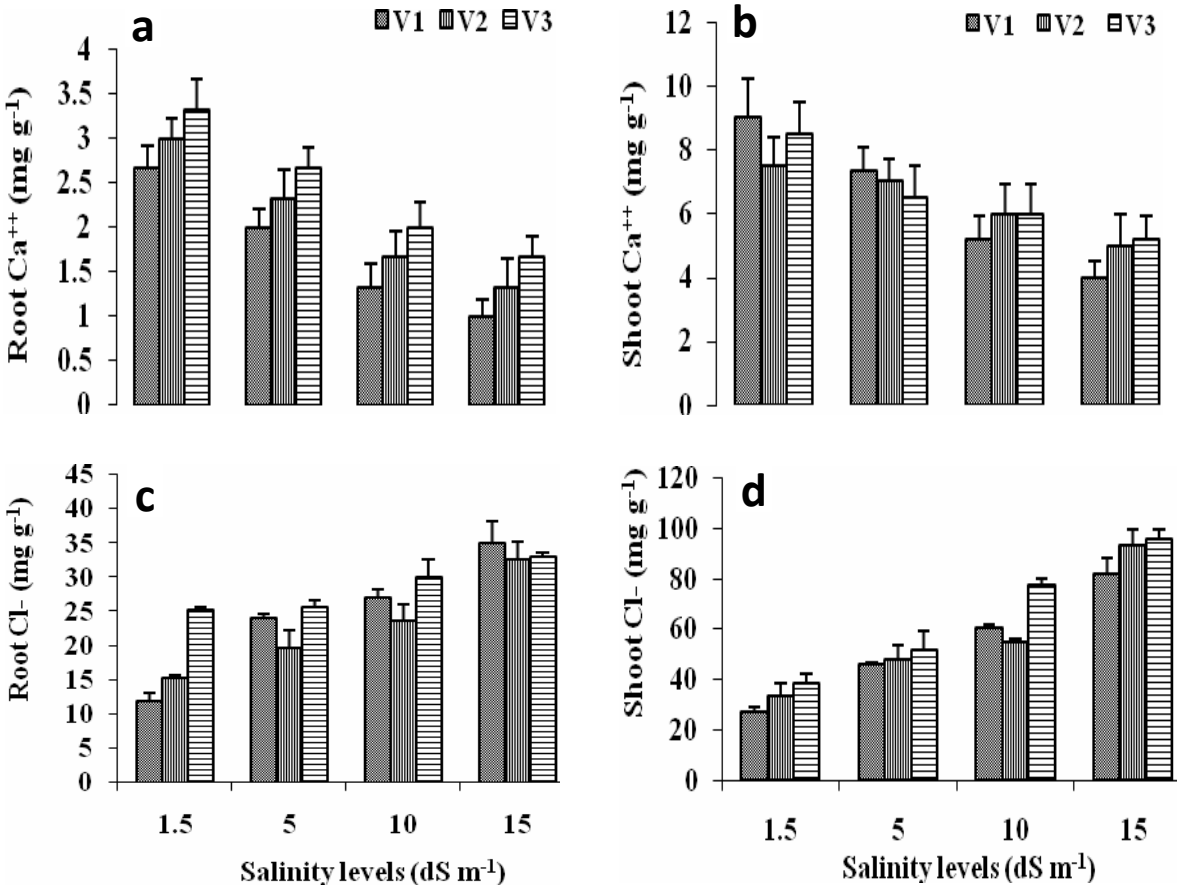

Fig. 1. Concentrations of Ca^{2+} (mg g^{-1} dry weight) in roots (a) and shoots (b) and concentration of Cl^- (mg g^{-1} dry weight) in roots (c) and in shoots (d) of the three wheat genotypes growing under varying salinity levels.

Concentration of Na^+ in roots of the plants growing under control conditions was minimum in V1 and maximum in V3. All salinity levels increased Na^+ concentration in roots significantly with a maximum observed in V3 under EC 10 and 15 dS m^{-1} (Fig. 2a). In shoots, however, difference in Na^+ concentration was not significant under control but salinity induced increase was significantly higher in V1 than that in V3 especially at EC 10 and 15 dS m^{-1} (Fig. 2b).

Concentration of K^+ in roots differed significantly both under control and saline conditions. In roots, it was the maximum in V3 and minimum in V2 and decreased significantly in V2 under EC 15 dS m^{-1} (Fig. 2c). Compared to roots, reduction in K^+ concentration in shoot was comparatively less and difference among varieties was also not significant except in V2 where the magnitude of reduction was significantly low. All three genotypes possessed almost the same K^+ concentrations in shoots at EC 15 dS m^{-1} (Fig. 2d).

Under control conditions, K^+/Na^+ ratio in roots was significantly different in all the three genotypes with the maximum in V1 and minimum in V3. Reduction in K^+/Na^+ ratio due to salinity was the minimum in V3 as even at EC 15 dS m^{-1}, K^+/Na^+ ratio was significantly

higher than in V1 and V2 (Fig. 3b). Like K^+/Na^+ ratio, Ca^{2+}/Na^+ ratio in roots was also significantly different in different genotypes with minimum in V3 and maximum in V1. All salinity levels significantly decreased Ca^{2+}/Na^+ ratio more promising being under EC 10 and 15 dS m^{-1} (Fig. 3c). In shoots however, it was maximum in both V1 and V3 under control and decreased nearly to zero in V1 under EC 15 dS m^{-1} (Fig. 3d).

Statistical analysis of the data for chlorophyll fluorescence of the three wheat genotypes is presented in Table 3. All the parameters differed significantly in different genotypes. Difference due to salinity and genotype x salinity interaction is also highly significant ($p<0.001$) except for F_0 where genotypes x environment interaction is non- significant (Table 3).

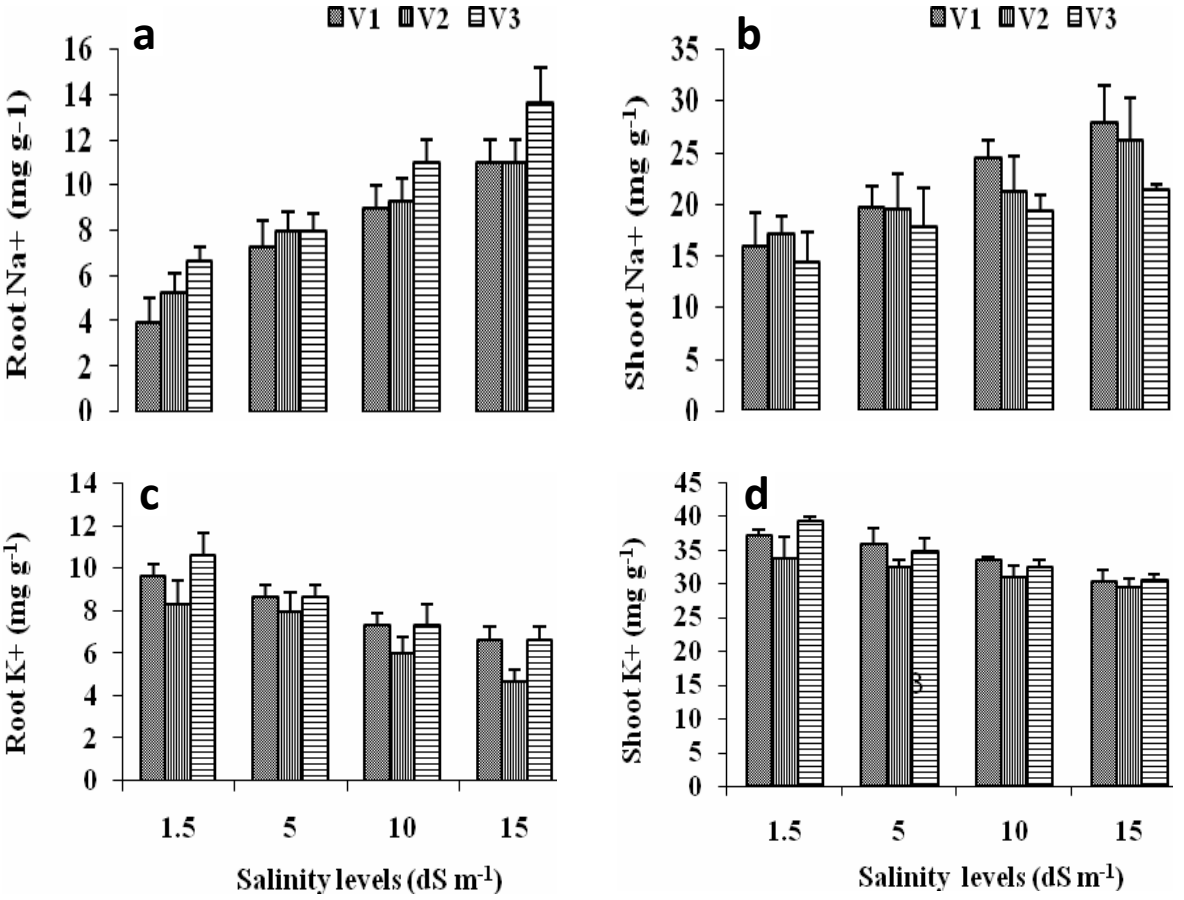

Fig. 2. Concentrations of Na^+ (mg g^{-1} dry weight) in roots (a) and shoots (b) and concentration of K^+ (mg g^{-1} dry weight) in roots (c) and in shoots (d) of the three wheat genotypes growing under varying salinity levels.

Table 3. Analysis of variance for different chlorophyll fluorescence parameters of three wheat genotypes after 73 days of growth in varying levels of salinity.

Source of variation	DF	Initial fluorescence (F_0)	Variable fluorescence (F_v)	Maximum fluorescence (F_m)	Quantum yield (F_v/F_m)
			Main Effects		
Variety (V)	2	201456.7 **	4924586.1 ***	3142195.2 ***	0.146 ***
Salt (S)	3	232700.5 ***	2307619.6 ***	2531300.9 ***	0.082 ***
			Interaction		
V x S	6	57229.5 NS	28.7 ***	1585045.4***	0.045 ***
Error	24	25422.4	47909.3	17708.9	0.006
Total	35				

NS = Non-significant, ** = Significant at 5%, and *** = Significant at 0.1%

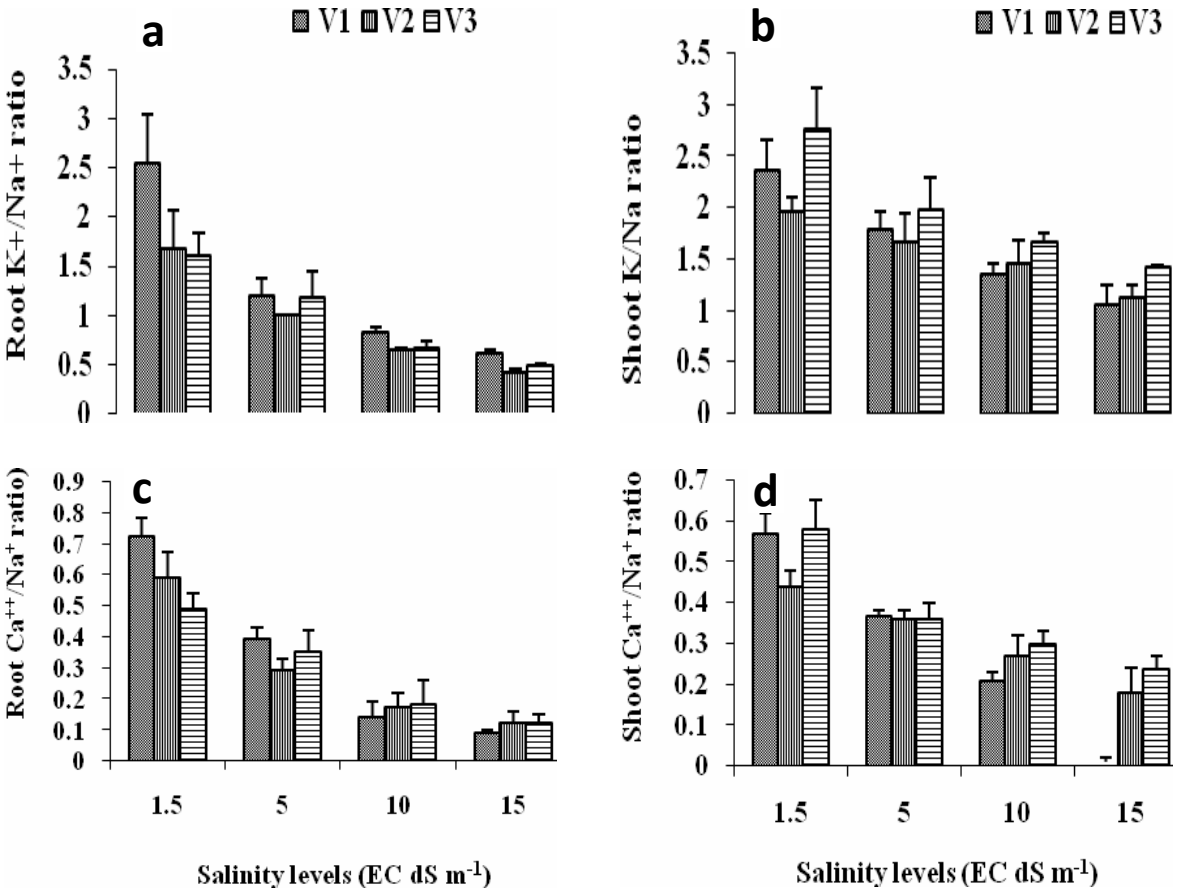

Fig. 3. K^+/Na^+ ratio in roots (a) and shoots (b) and Ca^{2+}/Na^+ ratio in roots (c) and in shoots (d) of the three wheat genotypes growing under varying salinity levels.

Table 4. Correlation values of Fv/Fm and grain yield with various ions in three wheat genotypes growing under non-saline (control) and salinity levels of EC 5, 10 and 15 dS m^{-1}.

Ions	Correlation values for genotypes growing under							
	A: Non-saline control		B: Saline conditions					
	Fv/Fm	Grain Yield	Fv/Fm			Grain Yield		
	All genotypes		V1	V2	V3	V1	V2	V3
R. Ca	-0.655	-0.507	0.871*	0.988**	0.834*	0.999***	0.974**	0.965**
S. Ca	-0.500	-0.649	0.917**	0.980**	0.768	0.993**	0.889*	0.887*
R. Cl	-0.737	-0.602	-0.870*	-0.940**	-0.842*	-0.974**	-0.868*	-0.961**
S. Cl	-0.575	-0.417	-0.948**	-0.881*	-0.917**	-0.974**	-0.809*	-0.997**
R. Na	-0.655	-0.507	-0.883*	-0.960**	-0.941**	-0.992**	-0.976**	-0.998**
S. Na	-0.993**	-0.998**	-0.936**	-0.925**	-0.838*	-0.988**	-0.863*	-0.942**
R. K	0.963**	0.996**	0.914**	0.980**	0.786	0.993**	0.853*	0.933**
S. K	0.952**	0.992**	0.991**	0.984**	0.802*	0.928**	0.923**	0.926**
R. K/Na	0.981**	0.090**	0.735	0.952**	0.832*	0.966**	0.996**	0.967**
S. K/Na	0.981**	1.000***	0.883*	0.963**	0.758	0.999***	0.928**	0.900**
R.Ca/Na	0.583	0.426	0.810*	0.919**	0.822*	0.993**	0.999***	0.965**
S.Ca/Na	-0.796	-0.893	0.856*	0.980**	0.726	1.000***	0.932**	0.865*
Yield	0.984**		0.871*	0.930**	0.930**			

*Significant at 5%, **Significant at 1%, ***Significant at 0.1%

It is interesting to note that F_0 (Fig. 4a), F_v (Fig. 4b), F_m (Fig. 4c) and F_v/F_m (Fig. 4d) both under control and saline conditions was least affected in V2. However, effect of salinity on F_0 and F_m was significant especially in V1 and V3 (Fig. 4a & b). F_m reduced significantly and F_v/F_m highly significantly ($p<0.001$) in V3 at EC 15dS m^{-1} (Fig. 4d) while in V2 it was absolutely not affected. In V1, it was though reduced under EC 15 dS m^{-1} but reduction was not significant (Fig. 4d). Looking at the relationship of F_v/F_m with concentrations of various ions and grain yield, it appeared that under control conditions, root and shoot Ca^{2+}, root and shoot Cl^- and root and shoot Na^+ had a negative relationship with F_v/F_m and among them relationship of F_v/F_m with concentration of Na^+ in shoot exhibited highly significant value. Relationship of shoot Ca^{2+}/Na^+ ratio was also negatively correlated with F_v/F_m but it was not significant. F_v/F_m exhibited a positive and highly significant relationship with root and shoot K^+, root and shoot K^+/ Na^+ ratio and grain yield. The relationship of grain yield with various ions also exhibited absolutely identical trend (Table 4).

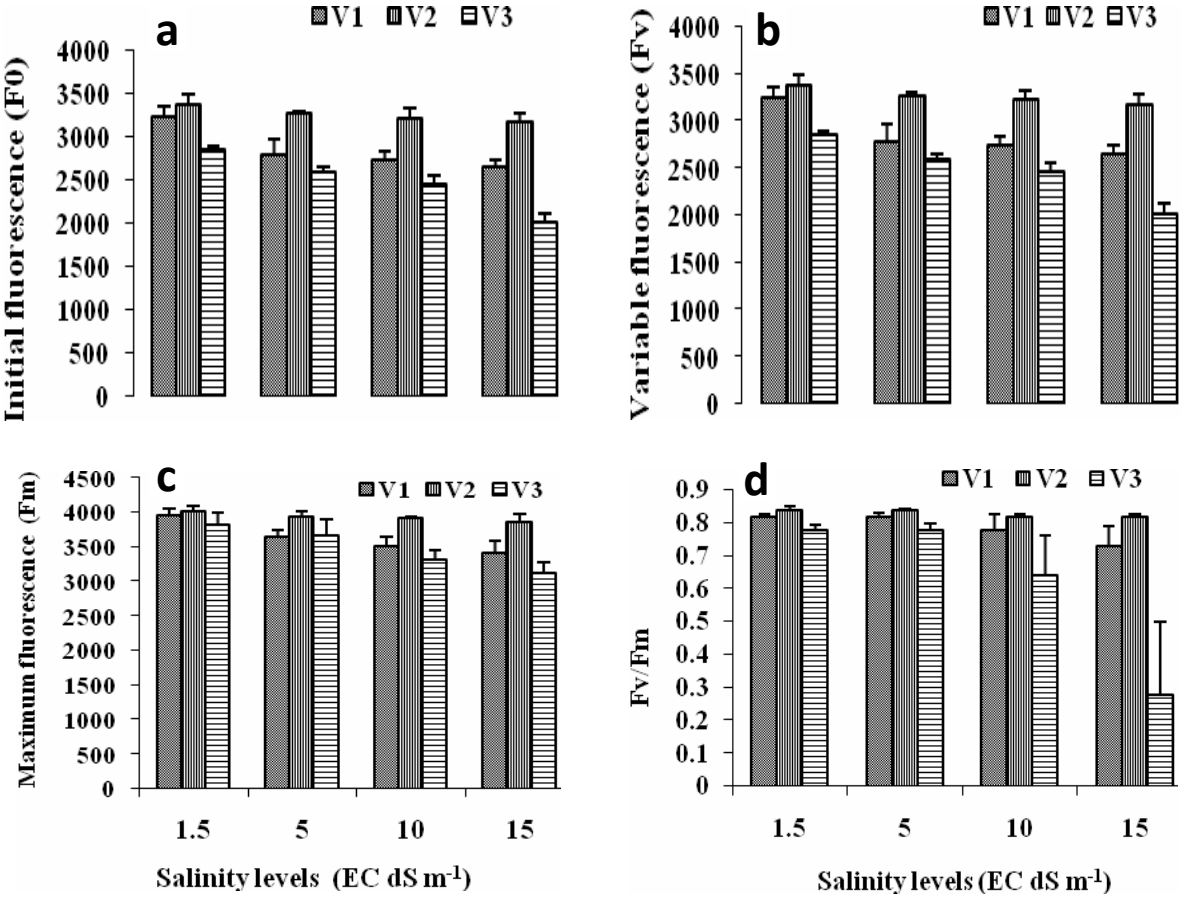

Fig. 4. Chlorophyll fluorescence parameters comprising initial fluorescence: F_0 (a), variable fluorescence: F_v (b), maximum fluorescence: F_m (c) and quantum yield of chlorophyll fluorescence: F_v/F_m (d) of the three wheat genotypes growing under varying salinity levels.

Under saline conditions however, relationship of various ions with F_v/F_m is positive and highly significant except for root and shoot Cl^- and root and shoot Na^+, which exhibited a negative relationship (Table 4a). The relationship appeared much stronger in V2 than in V1 and V3. Interestingly, shoot K^+ was more strongly related with F_v/F_m in V1 than in V3 but shoot K^+/Na^+ ratio was more strongly related with F_v/F_m in V2 compared to V1 and V3. F_v/F_m was also very strongly related with grain yield especially in V2 and V3 growing under saline conditions. Like F_v/F_m, grain yield was also correlated significantly ($p<0.001$) and positively with various ions except for root and shoot Cl^- and root and shoot Na^+. The relationships of shoot K^+/Na^+ and shoot Ca^{2+}/Na^+ ratios appeared much stronger in V1 followed by V2 and V3 (Table 4b).

Discussion

Variations in the uptake of nutrient ions under saline conditions are very well documented (Epstein & Jefferies 1964; Cramer *et al.*, 1987; Mohammad *et al.*, 1987; Martinez & Cerda 1989; Grattan & Grieve 1999 latest reference addede ; Naidu & Rengasamy 1993; Kent & Lauchli 1998; Essa, 2002; Hu & Schmidhalter 2005; Tuna *et al.*, 2007). The tolerant plants are expected to partition

toxic ions such as Na^+, Cl^- and even NO_3^- away from the physiologically active cells (Gorham, 1995); hence differential accumulation of various ions in different parts of a plant is an important indicator of tolerance.

In the present study, we have not observed significant variation in concentrations of Ca^{2+} in roots and shoots under control and saline environment. However, Cl^- concentration under both the condition did exhibited significant variations especially comparatively higher Cl^- concentration in roots compared to shoots in otherwise salt sensitive commercial wheat genotype V1. It appeared that this genotype possesses the ability to absorb comparatively more Cl^- under control condition which increased significantly under EC 10 and 15 dS m^{-1} but its ability to restrict accumulation of higher Cl^- in shoot compare to that in V2 and V3 is unique as it has only been reported earlier (Shannon, 1978) in one of the salt tolerant wild wheat grasses *Elytrigia pontica* (Farooq *et al.,* 1988, Gorham *et al.,* 1985 a, b). Chloride is an essential micronutrient involved in turgor and osmo-regulation (White & Broadley 2001) but its excessive accumulation in the leaves can be toxic for the plants (Hajrasuliha, 1980). Since accumulation of Cl^- is significantly less in shoots of V1 (a known salt sensitive commercial cultivar) compared to salt tolerant V2 and V3 hence, its sensitivity to salinity is probably independent of Cl^- in shoot: information which has not yet been reported in the literature. V3 also accumulated significantly less Na^+ in shoot: a mechanism through which plants protect themselves for NaCl toxicity (Cramer *et al.,* 1985) especially under EC 10 and 15dS m^{-1} compared to V1 and V2 which exhibited the highest Na^+ concentration in shoot. Thus, comparatively high Na^+ in shoots of V1 appeared to be one of the reasons of its sensitivity while ability to accumulate less Na^+ in shoots appeared to have imparted more tolerance to V3 compared to V2. The higher tolerance of V3 is further confirmed from the fact that though concentration of K^+ in all the three genotypes is nearly identical, it is probably the higher K^+ than less Na^+ (K^+/Na^+ ratio of 1.5) in the shoot under higher salinity that makes V3 tolerant while a little higher (not significant) concentration of Na^+ and significantly less concentration of K^+ (K^+/Na^+ ratio of 1.3) in the shoots that makes the V1 (commercial cultivar) less tolerant (not sensitive) compared to V3.

Interestingly V2 also exhibited K^+/Na^+ ratio nearly similar to V1, Na^+ concentration higher than V3 and K^+ concentration equal to V3 and V1 nevertheless, V2 is reported (Farooq & Azam 2007) to be more tolerant than V3 and V1. Probably, it is the minimum concentration of Ca^{2+} in shoot and almost negligible (0.1) Ca^{2+}/Na^+ ratio that makes V1 the salt sensitive genotype. High Ca^{2+} concentration is reported to be effective in maintaining of K^+/Na^+ selectivity (Cramer *et al.,* 1987; Kent & Lauchli 1989) under saline environment. It appeared that despite accumulating less Cl^- in shoot, V1 could not restrict movement of Na^+ from root to shoot and thus its Ca^{2+} was compromised (Hu & Schmidhalter, 2005) which probably made V1 sensitive (less tolerant) to salinity compared to V2 and V3.

The photochemical efficiency of PS-II or quantum yield of chlorophyll fluorescence (F_v/F_m) is a very power full and widely used but sensitive technique (Maxwell & Johnson 2000; Baker & Rosenqvist, 2004) for determining the condition of plants growing under any kind of environmental stresses like freezing, elevated ozone, heat, heavy metals, and excess water (Meinander *et al.,* 1996; Lazar *et al.,* 1997; Lu & Zhang 1998). For a healthy plant, its value under stress should range between 0.8 and 0.83 (Zarco-Tejada *et al.,* 2001). Reading below 0.8 indicates that the specific plant is growing under stress. In the present study, we observed that F_v/F_m value for V2 is 0.82 at the highest levels of salinity which finally confirms that V2 (N-1073) is the most salt tolerant among the three genotypes.

In most of the genotypes, leaf chlorophyll fluorescence responses to increasing salinity appeared as change in F_v/F_m and F_0. Difference in F_0 responses among genotypes provides an insight into effects on the leaf photosynthetic apparatus. For example, increase in F_0 is associated with dissociation of the light harvesting chlorophyll a & b complexes from the reaction center complex of photosystem-II (Yamane *et al.,* 2000; Kanwal *et al*., 2011). A decrease in F_0 may reflect alterations in the xanthophyll-cycle-dependent, non-radiative energy dissipation process (Hong & Xu 1999). This indicates one of the initial forms of salt damage within the plant that is impairment of the photo-protective process that facilitates the dissipation of excess energy within the leaf (i.e., the xanthophyll cycle). In the present study, F_0 did not show any variation in V2 which indicated that its light harvesting chlorophyll a & b did not dissociated from the reaction center complex of photosystem-II and hence V1 is functioning normally even under EC 15 dS m^{-1}. Since F_0 is not affected hence F_0/F_m and F_v/F_m also did not affected which again confirms the tolerance of this genotype. The value of F_v/F_m of V1 is less than the standard value of 0.82 which indicated that the genotype is growing under stress. Interestingly, V3 which otherwise appeared the most salt tolerant genotype exhibited F_v/F_m value of 0.28 which is significantly ($p<0.001$) less than that of standard value of 0.80. It appeared that F_v/F_m is not related to salt stress in V3 thereby confirming its salt tolerance mechanism totally different from V1 and V2. Significant decrease in F_v/F_m suggests that though the plant is facing extreme stress conditions (Adams & Adams 2004; Kanwal *et al*., 2011) its nature and possible underlying causes needs further investigations.

The relationships between F_v/F_m and concentrations of various ions and grain yield of the three genotypes growing under normal and saline conditions was also studied. The status of these relationships is almost similar for V1 and V2 with one exception that relationship of F_v/F_m with K^+/Na^+ ratio in root is not significant in V1 where as it is highly significant in V2 and V3. V3 exhibited entirely different pattern as in this genotype relationships of F_v/F_m with concentrations of Ca^{++} in shoot, K^+ in root, K^+/Na^+ and Ca^{2+}/Na^+ ratios in shoot are not significant which is contrary to the V2 were all these relationships are highly significant. However, when relationships of grain yield with various ions were studied, we observed certain values of relationship significantly less in V2 compared to those in both V1 and V3. All these variations confirm that the three genotypes used in the present study were significantly different from each other with the order of increasing tolerance as V1, V2 and V3.

Acknowledgment

This work is a part of M. Phil thesis of the first author who conducted this research on the Agriculture Linkage Program (ALP) of the USAID research project titled "Enhancement and evolution of germplasm for stressed lands through the use of agro-biodiversity" given to Dr. Shafqat Farooq as Principal Investigator (PI).

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(Received for publication 14 October 2011)