



Effect of Salt Stress in the Field on Performance, Correlations, Heritability and Selection Gain of Wheat Doubled Haploids

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Authors' contributions

This work was carried out in collaboration between all authors. Author AMMA-N designed the study, wrote the protocol, and wrote the first draft of the manuscript. Authors SRSS and MMMA managed the literature searches and analyses of the study. Author OMAE-A managed the experimental process. All authors read and approved the final manuscript.

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ABSTRACT

The objective of the present investigation was to study the effect of salt affected soils in different locations in Egypt on performance, variability, correlations, heritability, and genetic advance from selection of 117 bread wheat doubled haploids and 4 checks. The experiments were conducted at the locations (seasons) Serw (2011/12), Sakha (2011/12), Sakha (2013/14) and Gemmeiza (2013/14); where soil EC_e was 9.4, 5.7, 5.5 and 2.4 dSm^{-1} , respectively and water EC_w was 0.5 -0.6 dSm^{-1} . The reduction in grain yield/plant (GYPP) in the most affected environment by salt (Serw) as compared with the lowest affected environment by salt (Gemmeiza) (64.25%) was associated with significant ($P \leq 0.01$) reductions in most studied traits and a slight increase (delay) in days to heading (DTH). Heritability percentages in the broad-sense were at maximum at Sakha (2013/14) for PH, DTM, GFP and GFR and Sakha (2011/12) for NSPP, NGPS, TGW and GYPP. However, the highest expected genetic advance (GA%) from selection in the field was observed at the most affected environment by salt (Serw) for most studied traits. Traits NSPP, GFR, GFP, TGW and GYPP could be recommended as selection criteria for salinity tolerance in the field.

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Keywords: *Triticum aestivum*; heritability; genetic advance; selection criteria; salinity tolerance; doubled haploids.

1. INTRODUCTION

Wheat is one of the oldest and most important cereal crop in Egypt. Although wheat productivity in Egypt has increased during the past years, wheat production supplies only 45% of its annual domestic demand. Egypt needs to make a great effort to increase wheat production *via* extending its growing area outside the Delta and Nile Valley. However, soil of this area is suffering from salinity. Increasing salt tolerance of Egyptian wheat cultivars is one of the cheap methods to spread growing it in this area.

Classical breeding in Egypt was able to develop few bread wheat cultivars such as Sakha 8 and Sakha 93 of higher salinity tolerance than other commercial cultivars. Genetic diversity in wheat has been reduced mainly due to narrow genetic base of the wheat germplasm [1]. Therefore, wheat breeders are always looking for new methods to enrich breeding material of better tolerance to salinity stress. Modern biotechnological techniques in plant breeding, could contribute, to a great extent, to the induction of novel genetic variation, which does not exist in the gene pool, such as somaclonal and/or gametoclonal variation [2]. The anther culture technique helps in developing doubled haploids, in a short time from wheat crosses that show new genetic variation amenable for efficient selection for salinity tolerant genotypes [3]. One hundred seventeen bread wheat doubled haploid (DH) lines derived from the cross Sakha 8 X Line 25 *via* anther culture technique [4] were used in the present investigation; the first parent (Sakha 8) of the cross was an Egyptian salinity tolerant cultivar and the second parent (Line 25) was a high yielding promising breeding line. This set of DH lines is expected to include line(s) that accumulated favorable genes for both high-yield and salinity tolerance.

Salinity reduced vegetative growth more than grain yield. Salam et al. [5] reported that number of tillers and grains, 100 seed weight and grain yield were adversely affected by salinity. Kingsbury and Epstein [6] evaluated 5000 wheat genotypes in 50% seawater and identified 29 accessions that produced seed. Jafari-Shabestari et al. [7] evaluated 400 wheat genotypes in field at California and recognized numerous genotypes that always gave high yield under low and high salinity levels. Ahmad et al.

[8] studied six wheat varieties in salt affected soils and narrated that salt tolerant varieties produced greater yield than salt susceptible ones due to higher dry weight of shoot and spike and better grain development.

Kamboj [9] studied twelve wheat genotypes under salinity conditions and reported that grain yield per plot and harvest index recorded the highest phenotypic and genotypic variations and that high heritability was recorded by grain yield per plot thus, suggesting direct selection for yield would be effective under saline conditions; maximum genetic advance as per cent of mean was also recorded by grain yield and harvest index. Information on heritability, expected selection gain and correlations from such DH population under salt stress conditions are necessary for plant breeders. Changes in heritability and genetic variance components have been predicted as stress level increases [10,11]; such changes were reported by Hoffmann and Parsons [12]. Changes in genetic variation (increase or decrease), would seem likely to arise with increased stress, because different genes may contribute to similar character under diverse environments [13,14]. The objective of the present investigation was to study the response of performance, heritability, genetic advance from selection and correlations in 117 DH lines of wheat to high salt stress conditions in the field at different salt stressed locations in Egypt.

2. MATERIALS AND METHODS

2.1 Plant Materials

Seeds of 121 bread wheat genotypes, including 117 doubled haploid lines, their two parents (Sakha-8 and Line-25) of the cross from which they were produced *via* anther culture technique [4] and the 2 check cultivars Sids-1 and Sakha-93 were obtained from Wheat Research Department, Field Crop Research Institute (FCRI), Agricultural Research Center, (ARC), Egypt. The reason of using this large number of lines in this study is to exploit the maximum variation resulting from the crossing between a salt tolerant cultivar (Sakha-8) and a high yielding promising (Line-25) in an attempt to find new line(s) that assemble more tolerance to salinity and higher yielding ability as compared to

their parents and the check cultivars, i.e., new sources of salinity tolerance.

2.2 Sowing Method

The present investigation was carried out in the fields of following experimental research station of ARC: 1. Sakha (Kafr El-Shekh governorate) at season 2011/2012, 2. Serw (Domiati governorate) at season 2011/2012, 3. Sakha (Kafr El-Shekh governorate) at season 2013/2014 and 4. Gemmeiza (El-Gharbia governorate) at season 2013/2014. The stations are located at Sakha (31° 5' 20" N, 30° 57' 3" E and Altitude = 2 m asl); Serw (31° 7' 42" N, 30° 38' 40" E and Altitude = 6 m asl) and Gemmeiza (30° 58' 20" N, 31° 23' 20" E and Altitude = 20 m asl). Sowing date was 29/11/2011 at Sakha, 22/11/2011 at Serw, 27/11/2013 at Sakha and 25/11/2013 at Gemmeiza Station. A simple lattice design (11 X 11) with two replications was used; the seeds were sown in individual hills in

rows. Each row was of 2.5 meter length and row to row distance of 30 cm and hill to hill distance of 10 cm. The irrigation and fertilization were done as recommended by ARC, for commercial production at the three locations. Flooding irrigation was given four times, the first irrigation was given after 21 days from planting and the succeeding ones were given at 20 to 25 days intervals according to the weather conditions. The fertilization was applied using 15 kg P₂O₅ (100 Kg Mono Super Phosphate 15.5%) + 70 kg Nitrogen (210 kg Ammonium Nitrate 33.5%) per feddan (one feddan=4200 m²) split in three parts, first 20% with seeds, second 40% with first irrigation and third time 40% with the second irrigation. Soil and water analyses of the three locations were done in Analysis and Studies Unit (ASU), Soil, Water and Environment Res. Inst. (SWER) of ARC, Egypt and the data are presented in Table 1. The meteorological data at each location were recorded by Meteorological Station of each location in seasons (Table 2).

Table 1. Soil and water analyses at Serw, Sakha and Gemmeiza locations

Analysis	Serw		Sakha		Gemmeiza				
	(2011/2012)		(2011/2012)		(2013/2014)				
	Soil	Water	Soil	Water	Soil	Water	Soil	Water	
Clay %	6.3		55.6		51.9		54.6		
Silt %	40.0		30.1		32.3		35.5		
Fine sand	30.4		8.8		10.8		6.4		
Coarse sand	23.3		5.5		5.0		3.5		
Soil type	Sandy Loam		Clay		Clay		Clay		
pH	8.4	6.8	7.8	7.1	8.1	7.1	7.9	7.4	
EC (dSm ⁻¹)	9.4	0.5	5.7	0.5	5.5	0.5	2.4	0.6	
Salt Conc. (ppm)	7520	294	4568	333	4400	333	1536	384	
Soluble Cations mEq/l	Ca ⁺²	11.2	0.0	3.0	0.0	3.9	0.0	3.0	0.0
	Mg ⁺²	12.6	0.0	3.4	1.0	3.2	0.9	2.4	0.0
	Na ⁺	48.2	2.6	20.0	4.0	19.0	4.5	11.6	3.7
	K ⁺	1.1	0.0	0.6	0.0	0.7	0.0	1.5	0.0
	Ca	6.0	0.0	3.0	0.4	3.0	0.5	1.9	0.4
Soluble Anions mEq/l	Cl ⁻	24.0	1.4	15.0	2.0	14.0	2.2	10.0	2.3
	SO ₄ ⁻	15.0	0.0	9.0	1.0	8.5	1.8	7.0	1.2
	CO ₃ ⁻	2.0	0.0	0.0	0.0	0.0	0.1	1.0	0.1
	NO ₃ ⁻	9.6	0.6	3.0	0.1	2.4	0.3	1.5	0.2
Macro elements ppm	N	20.0	0.0	50.0	0.0	42.0	0.0	55.0	0.0
	P	3.0	0.0	15.0	0.0	16.0	0.0	14.0	0.0
	K	728.0	0.0	820.0	0.0	840.0	0.0	960.0	0.0
Micro elements ppm	Fe	23.0	0.0	40.0	0.0	43.0	0.0	48.0	0.0
	Cu	6.4	0.0	8.2	0.0	9.0	0.0	9.5	0.0
	Zn	0.9	0.0	1.9	0.0	2.1	0.0	2.0	0.0
	Mn	13.2	0.0	18.8	0.0	19.8	0.0	19.9	0.0

Source: analysis and studies unit, soil, water and environment Res. Inst. (SWER), ARC, Egypt

Table 2. Meteorological data during seasons of wheat growing at Serw, Sakha and Gemmeiza locations

Month	Temp. max. (°C)	Temp. min. (°C)	Temp. mean (°C)	RH mean %	Precipitation (mm)
Sakha (2011/2012)					
Nov.	22.8	10.3	16.5	67.5	0
Dec.	21.3	6.8	13.6	72.8	14.6
Jan.	19	13.8	16.3	77.4	0
Feb.	11.4	9.6	10.5	69	32.7
Mar.	14	12.1	13.6	68	42.8
Apr.	19	17.1	18	63	0
Total					90
Sakha (2013/2014)					
Nov.	22.8	10.3	16.4	68.5	0
Dec.	20.7	6.8	12.5	71.6	21.4
Jan.	10.2	8.8	8.9	68.6	32.5
Feb.	11.4	8.8	9.7	74.1	50.4
Mar.	12.5	10.5	11.2	66.5	44.9
Apr.	19.4	16.8	17.9	63	0
Total					149.2
Serw (2011/2012)					
Nov.	21.2	9.6	15.3	63.7	0
Dec.	20.2	7.2	12.8	65.9	16.9
Jan.	14.1	7.2	10.6	64.8	36.6
Feb.	9.6	8	8.3	65.7	58.7
Mar.	10.6	8.3	9	68.7	81.7
Apr.	11.6	9.8	10.4	61.8	41.8
Total					235.8
Gemmeiza (2013/2014)					
Nov.	22.7	10.7	16.6	66	0
Dec.	22.2	9.2	14.5	66.4	17.3
Jan.	20.3	7.5	13.5	69.8	37.3
Feb.	11.7	8.8	11.6	66	59.9
Mar.	11.1	9.5	9.7	68.5	83.4
Apr.	11.8	9.5	10.2	71.1	42.6
Total					240.5

Source: Meteorological stations of Agric. Res. centre at Serw, Sakha and Gemmeiza. R.H. = relative humidity, Temp. = temperature

2.3 Data Recorded

Data were recorded on the following traits: Days to heading (DTH): It was estimated as the number of days from sowing date to the date at which 50% of main spike awns/plot had completely emerged from the flag leaf. Days to maturity (DTM): It was recorded as the number of days from sowing to the date at which 50% of main peduncles/plot have turned to yellow color (physiological maturity). Grain filling period (GFP): Number of days from 50% anthesis to 50% physiological maturity (on a per plot basis). Grain filling rate (GFR) in g/day: It was calculated as the weight (g) of grain dry matter accumulated per day, as follows: $GFR = GYPP / GFP$. Plant

height (PH) in cm: It was measured as the height of plant at maturity, measured from the soil surface level to the tip of spike, excluding awns. Number of spikes/plant (NSPP): It was measured as the total number of fertile spikes per plant as an average of five plants. Number of grains/spike (NGPP): It was measured as the total number of grains per main spike as an average of five spikes. Thousand grain weight (TGW) in g: It was measured as the weight of 1000 grains using an electronic scale. Grain yield/plant (GYPP) in g: It was measured as the dry matter (biomass) allocated to the grains per plant as an average of five plants. Straw yield/plant (SYPP) in g: It was measured as the dry matter (biomass) allocated to the straw (the above ground parts of the plant, except grains) as an average of five plants.

Biological yield/plant (BYPP) in g: It was measured as the dry matter (biomass) allocated to the grain to the whole plant except root, as an average of five plants. It was estimated as follows: $BYPP = GYPP + SYPP$. Harvest index % (HI): It was estimated as follows: $HI\% = 100 (GYPP / BYPP)$.

2.4 Statistical Analysis

All data were subjected to analysis of variance (ANOVA) of Lattice design (11X11) using GENSTAT 10th EDITION FOR WINDOWS and comparisons of means were made using the least significant difference (LSD) test at $P < 0.05$ and 0.01 levels of confidence, according to Snedecor and Cochran [15]. Combined analyses of variance across locations and/or seasons were performed after making the homogeneity test. Expected mean squares were estimated from ANOVA table (Table 3) according to Hallauer and Miranda [16]. Genotypic (σ_g^2) and phenotypic (σ_{ph}^2) variances were computed as follows: $\sigma_g^2 = (M_2 - M_1) / r$, $\sigma_{ph}^2 = \sigma_g^2 + (\sigma_e^2 / r)$, where r = number of replications.

The following equations (proposed by Burton, [17]) were used to estimate genotypic (GCV) and phenotypic (PCV) coefficients of variations: $GCV = (\sigma_g / \bar{x}) \times 100$, $PCV = (\sigma_{ph} / \bar{x}) \times 100$, Where: \bar{x} = Mean of the trait at the same salinity level.

Table 3. Analysis of variance and expected mean squares (EMS)

SOV	df	MS	EMS
Genotypes (G)	(g-1)	M_2	$\sigma_e^2 + r \sigma_g^2$
Error	(r-1)(g-1)	M_1	σ_e^2

Heritability (%) in the broad sense (h_b^2) was estimated according to Singh and Chaudhary [18] by using the following formula: $h_b^2 \% = 100 \times (\sigma_g^2 / \sigma_{ph}^2)$. Expected genetic advance from selection for all studied traits as a percent of the mean was calculated according to Singh and Chaudhary [18] as follows: $GA (\%) = 100 K h^2 \sigma_{ph} / \bar{x}$, Where: \bar{x} = General mean, σ_{ph} = Square root of the denominator of the appropriate heritability, h^2 = The applied heritability, K = Selection differential ($k = 1.76$, for 10% selection intensity, used in this study). Coefficients of simple and rank correlations between attributes and their significance were calculated according to Steel et al. [19] by using SPSS 7 computer software.

3. RESULTS AND DISCUSSION

3.1 Analysis of Variance

Combined analysis of variance across the four environments (Table 4) revealed that genotypes differed significantly ($P \leq 0.01$) for all studied traits at all environments. Variance due to genotypes was the main contributor to the total variance in the present investigation; this was measured by the highest percentage of genotypes sum of squares to the total sum of squares (Table 4). Mean squares due to genotypes x environments interaction were significant ($P \leq 0.05$ or 0.01), indicating that selection is possible under each specific environment, and therefore, the proper genotype could be identified for each environment of specific salt affected soil. This conclusion is in agreement with that reported by previous investigators, e.g. Al-Naggar et al. [20]. Our results revealed that wheat genotypes responded differently to salinity stress at the three locations in terms of yield and yield components. Similar findings were reported by Richards et al. [21] and Slavich et al. [22].

To compare the effect of locations, that differ in salinity of their soils, with the genotypes effect, and their interaction, two combined analyses of variance (ANOVA) were performed; one across Serw and Sakha locations in 2011/12 season (Table 5) and another one across Gemmeiza and Sakha locations in 2013/14 season (Table 6). The first contributor to the total variance in the combined ANOVA across Serw and Sakha locations in 2011/12 season was genotype for seven out of nine studied traits, but was location for one trait (number of grains/spike) and genotype x location interaction for one trait (number of spikes/plant) (Table 5). Mean squares due to genotypes x location interaction were significant for all traits, except for plant height trait in 2011/12 season, indicating that the rank of genotypes differs from Serw to Sakha location for eight out of nine traits.

For the combined ANOVA across Sakha and Gemmeiza locations in 2013/14 seasons (Table 6), mean squares due to locations were significant ($P \leq 0.05$ or 0.01) for all the 12 studied traits, except for PH and GYPP traits, indicating that variation of these locations affected on all studied traits except plant height and grain yield/plant. Mean squares due to genotypes and genotypes X locations interaction were significant ($P \leq 0.05$ or 0.01) for all studied traits, indicating

that genotypes differed significantly and their ranks differed from one location to another. Significance of genotype X location interaction variances in both combined analyses (Tables 5 and 6) indicated that selection in each location (specific soil EC_e) could be efficient in isolating the best genotype(s) for each specific location. Thus, wheat genotypes behaved differently under different salt affected soil conditions and the superior genotype under one location might be different under another location, and a specific genotype should be identified as the best one in each location of a specific salt affected soil. Similar findings were also reported by

Richards et al. [21], Slavich et al.[22] and Munir et al. [23].

3.2 Effects of Salt Stressed Environment on Mean Performance

The effects of locations were mainly due to EC_e of the location. The most salt stress was at Serw (EC_e = 9.4) and least stress was at Gemmeiza (EC_e = 2.4); that is why Gemmeiza location was considered as control environment in this experiment, since EC_e in this location is not considered too high for wheat crop. Means of studied traits for the four environments across all genotypes are illustrated in Figs. 1 and 2.

Table 4. Combined analysis of variance of studied traits of 121 genotypes across four environments (Serw 11/12, Sakha 11/12, Sakha 13/14 and Gemmeiza 13/14)

SOV	df	Sum of squares %				
		DTH	DTM	GFP	GFR	PH
Environments (E)	3	2.56	3.25	2.89	5.07	9.58
Error (a)	6	0.39	0.25	0.87	0.98	0.52
Genotypes (G)	120	53.2**	51.3**	49.2**	64.0**	62.2**
G X E	360	20.2**	30.3**	37.0**	26.5**	22.4**
Error (b)	720	23.59	14.97	9.99	3.50	5.31
Total SS		598764	213589	76982	8191	169785
		NSPP	NGPS	TGW	GYPP	
Environments (E)	3	11.25	15.58	12.36	8.69	
Error (a)	6	1.48	1.98	2.05	5.25	
Genotypes (G)	120	52.2**	48.2**	60.2**	54.1**	
G X E	360	32.9**	29.9**	22.4*	31.1**	
Error (b)	720	2.2	4.2	3.0	0.9	
Total SS		64236	512037	457820	26543	

* and ** indicate significant at 0.05, 0.01 probability levels, respectively

Table 5. Combined analysis of variance of studied traits of genotypes across two locations Serw and Sakha in 2011/2012 season

SOV	df	Sum of squares %				
		DTH	DTM	GFP	GFR	PH
Locations (L)	1	9.2*	7.4	4.41	3.78	3.11
Error (a)	2	2.3	5.1	4.29	5.91	3.79
Genotypes (G)	120	51.0**	47.0**	59.3**	52.6**	81.0**
G X L	120	32.1**	32.0**	19.1**	34.6**	5.65
Error (b)	240	6.4	8.5	12.95	3.11	6.48
Total SS		524879.5	495479.3	150782.0	5365	181344.5
		NSPP	NGPS	TGW	GYPP	
Locations (L)	1	9.5	57.66 **	0.056	0.34	
Error (a)	2	7.3	0.31	1.23	0.4	
Genotypes (G)	120	25.2**	19.7**	46.6**	66.4**	
G X L	120	51.3**	19.4**	41.3**	29.8**	
Error (b)	240	6.7	2.92	10.85	3.1	
Total SS		458723.9	359357.4	161836.4	79199	

* and ** indicate significant at 0.05 , 0.01 probability levels, respectively

Table 6. Combined analysis of variance of studied traits of genotypes across two locations Sakha and Gemmeiza in 2013/2014 season

SOV	df	Sum of squares %					
		DTH	DTM	GFP	GFR	PH	NSPP
Location (L)	1	39.9**	20.4**	32.99	22.8**	1.99	19.5**
Error (a)	2	0.096	0.66	2.19	1.93	2.19	2.95
Genotypes (G)	120	47.9**	45.2**	46.5**	56.9**	66.5**	25.5**
G X L	120	9.1*	26.0**	10.7*	15.6**	16.7*	18.8**
Error (b)	240	2.99	7.79	12.58	3.78	12.58	33.3**
Total SS		10856	3093	5847	2499	42234	82624
		NGPS	TGW	GYPP	SYPP	BYPP	HI
Location (L)	1	75.2**	9.18*	0.26	65.6**	53.5**	59.2**
Error (a)	2	0.01	4.62	0.0038	1.38	1.13	0.19
Genotypes (G)	120	8.0*	28.9**	31.5**	9.8*	13.9*	12.8*
G X L	120	6.5*	21.5**	27.7**	7.0*	10.0*	10.4*
Error (b)	240	10.3*	35.9**	40.5**	16.3**	21.4**	17.4**
Total SS		175682	140931	66929	7366990	868982	52869

* and ** indicate significant at 0.05, 0.01 probability levels, respectively

The percentage of change of means of each environment to the control (Gemmeiza environment) showed that mean grain yield/plant of Serw and Sakha at 2011/12 season was significantly decreased by 64.25 and 46.69% as compared to that of Gemmeiza location at 2013/14 season. However, mean grain yield at Sakha in 2013/14 season ($EC_e = 5.5$) slightly and significantly increased by 2.54% than that at Gemmeiza in 2013/14 season ($EC_e = 2.4$).

The reduction in grain yield in the most affected environment by salt (Serw) as compared with the lowest affected environment by salt (Gemmeiza) was associated with a significant ($P \leq 0.01$) reduction in 7 out of 8 studied traits, *i.e.* DTM (8.39%), PH (39.25%), GFP (20.0%), GFR (55.25%), NSPP (42.97%), NGPS (24.48%) and TGW (23.91%) and a slight increase (delay) in DTH (4.81%) (Figs. 1 and 2). The direction of increase or decrease was generally from Gemmeiza in 2013/14 to Sakha in 2013/14, Sakha 2011/12 and Serw 2011/12, which is the same gradual direction of EC_e of these environments (2.4, 5.5, 5.7 and 9.4, respectively).

Season (year) effect was shown when comparing means of traits taken from the same location (Sakha) in 2011/12 season with those taken in 2013/14 season. Means in 2013/14 were significantly higher in magnitude than those in 2011/12 season for DTM (later), GFP, GFR, PH, TGW and grain yield/plant and significantly lower for DTH (earlier) trait. The reason for these differences at Sakha could be attributed to the changes in climatic factors such as temperature,

rainfall, ... etc (Table 2) prevailed during the wheat growing seasons 2011/12 and 2013/14 and not to the soil EC_e , which was very close to each other (5.5 vs. 5.7). It seems that climatic conditions prevailed in 2013/14 season were more suitable for wheat crop than those prevailed in 2011/12 season.

Comparing the two locations of Serw and Sakha at the same season (2011/12) showed that means of grain yield and the majority of its components and plant height were higher under Sakha than those at Serw. The reduction in means of traits at Serw as compared with Sakha is probably due to the higher soil EC_e at Serw (9.4) than that at Sakha (5.5) location. The reduction in trait means due to salt stress of soil at Serw compared with Sakha in the same season was much lower than that calculated due to salt stress at Serw in 2011/12 season compared with Sakha and Gemmeiza in another season (2013/14).

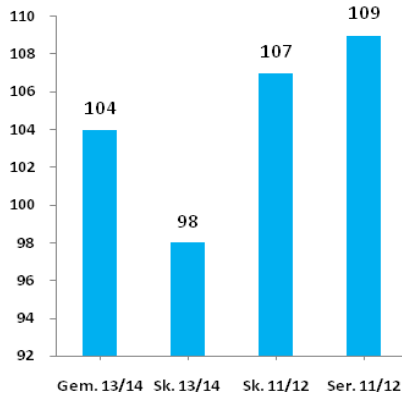
In 2011/2012 season, high salinity stress at Serw caused a significant reduction in GYPP (32.9%), GFR (31.2%), NSPP (14.4%), NGPS (27.0%) and PH (19.8%) as compared with medium salinity stress at Sakha, but caused a slight increase in 1000 grain weight (8.8%), days to maturity and days to heading (delay) (1.4 and 1.9%, respectively).

Field conditions vary from site to site, not only in soil salinity, but also in soil physical and chemical properties such as sodicity, high pH, and boron, and interactions between these stresses can occur. High pH can cause reduced K^+ uptake

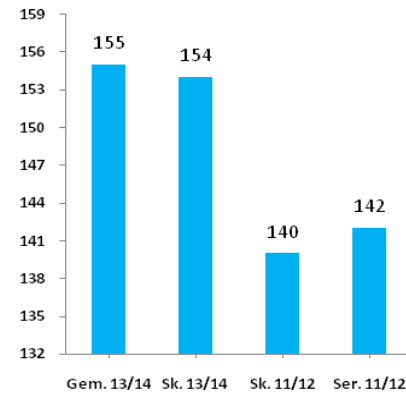
even though it might not affect Na⁺ uptake [24], and boron can affect subcellular distribution of

salt in leaves and hence salt tolerance of the plant [25].

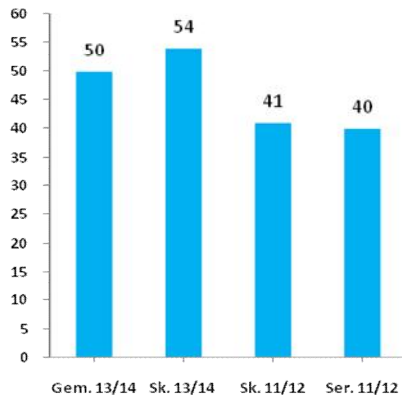
Days to heading (DTH)
L.S.D_{0.05} = 3.40



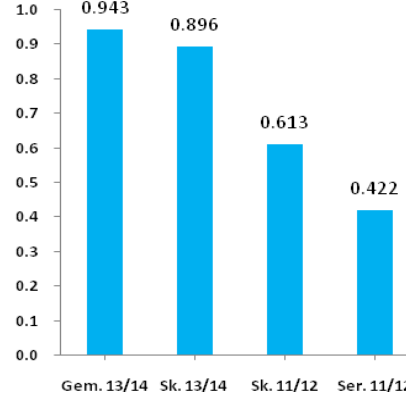
Days to maturity (DTM)
L.S.D_{0.05} = 1.37



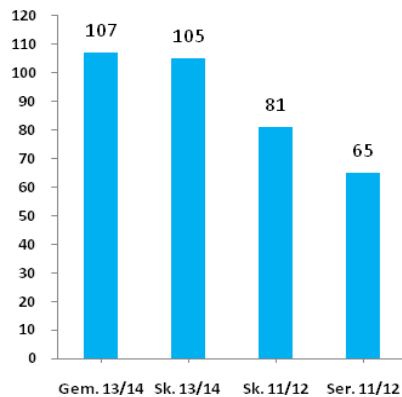
Grain filling period (GFP)
L.S.D_{0.05} = 2.21



Grain filling rate (g/day) (GFR)
L.S.D_{0.05} = 1.31



Plant height (cm) (PH)
L.S.D_{0.05} = 1.61



No. of spikes/plant (NSPP)
L.S.D_{0.05} = 1.03

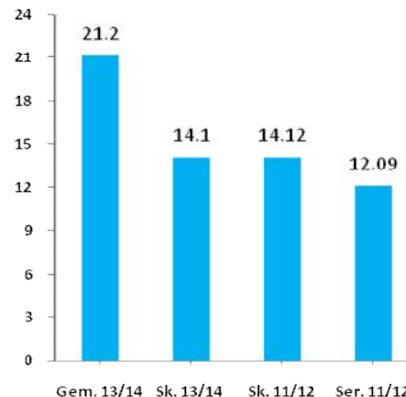


Fig. 1. Means of DTH, DTM, GFP, GFR, PH and NSPP at Gemmeiza (Gem.) 13/14, Sakha (Sk.) 13/14, Sakha (Sk.) 11/12 and Serw (Ser.) 11/12

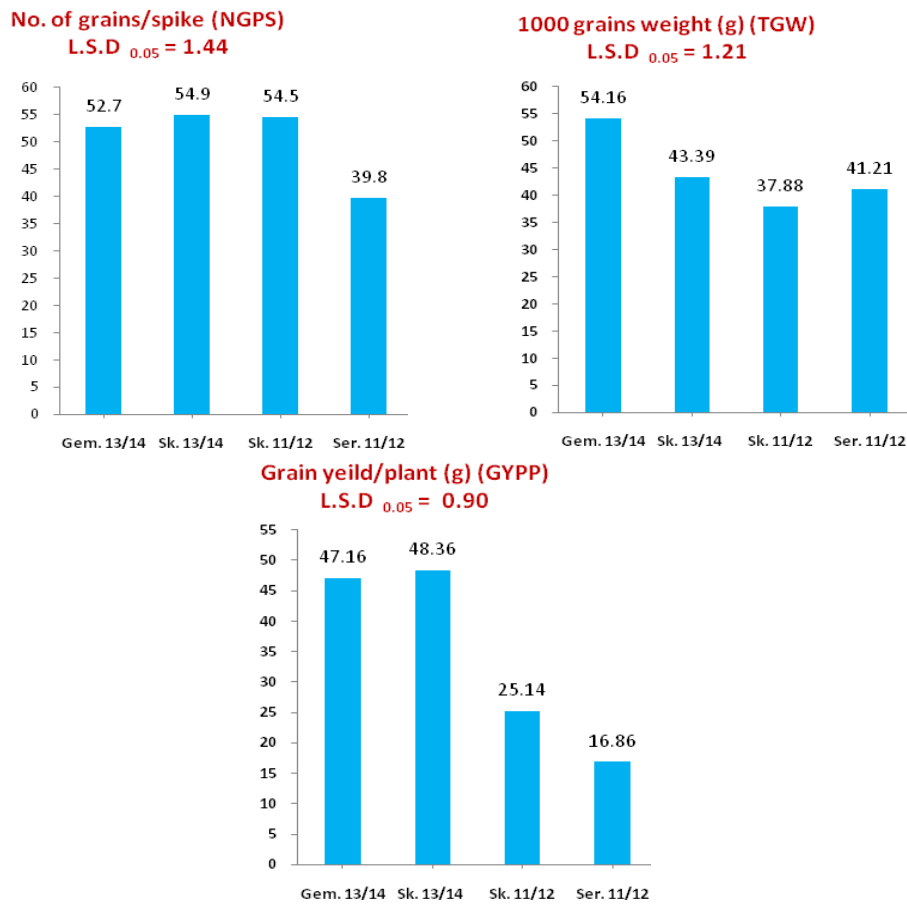


Fig. 2. Means of NGPS, TGW and GYPP at Gemmeiza (Gem.) 13/14, Sakha (Sk.) 13/14, Sakha (Sk.) 11/12 and Serw (Ser.) 11/12

The inhibitory effect of increasing salt in the soil of the fields on phenological, agronomic and yield traits of wheat observed in the present study was previously reported by several investigators. Salinity stress at different phenological stages inhibits photosynthetic activities of the plant because it had a direct inhibitory effect on the Calvin cycle enzymes [26]. Tillers plant-1 is the most salinity sensitive trait in wheat [27]. Therefore, to increase the yield under stress conditions, it is necessary to maintain high plant density. Tiller formation included tiller number and tiller biomass. Salinity reduces tiller number by delaying and reducing tiller emergence at the vegetative stage. After tiller emergence, growth of tillers at all stages is inhibited by salinity due to its damage on the essential metabolic reaction in plants, resulting in low tiller biomass and small tiller size [28]. E_{Ce} >7.5 dSm⁻¹ in soil water could eradicate most of the secondary tillers and greatly reduce the formation of tertiary and lateral tillers. The yield

potential of wheat is greatly dependent on the number of tillers plant-1 that is affected in the early life cycle. Number of tillers regulates grain yield by its prime influence on the number of spikes in wheat [29]. High salinity has been observed to delay the onset of flowering in many plant species [30]. Further study is needed to determine the causes of the delay in flowering due to salt stress

3.3 Effects on Trait Interrelationships

Correlation coefficients between mean grain yield per plant and other studied traits under each environment and combined across environments are presented in Table 7. In general, grain yield showed a positive and significant correlation, with most studied traits at each environment, but in low to medium magnitude. Magnitude of correlation coefficient for combined data across all environments was generally

higher than under separate environments and reached to 0.96 between GYPP and grain filling rate and 0.91 between GYPP and harvest index.

Grain yield/ plant showed near perfect positive association with grain filling rate (GFR) with a correlation coefficient of > 0.93 under all environments, and showed significant and positive correlations with BYPP (0.76), HI (0.60), GFP (0.59) and SYPP (0.52) at Gemmeiza 2013/14, HI (0.84), SYPP (0.62) and GFP (0.51) at Sakha 13/14, GFP (0.35) at Sakha 2011/12, NSPP (0.73) and GFP (0.43) at Serw 2011/12 NSPP (0.65) and SYPP (0.64) for combined data across environments. The strongest associations in the present investigation carried out in the field were therefore between GYPP and each of GFR, GFP, NSPP and TGW. In wheat, under salinity conditions several researches reported a strong correlation between grain yield and number of tillers per plant. They indicated that the importance of number of fertile tillers, which in turn results in number of spikes / plant as selection criterion to increase grain yield under salinity conditions [23,27,31].

3.4 Effect on Variability, Heritability and Selection Gain

Phenotypic (PCV) and genotypic (GCV) coefficients of variation for phenological and yield attributes of the 121 wheat genotypes under four environment in the fields of Serw (2011/12), Sakha (2011/12), Sakha (2013/14) and Gemmeiza (2013/14) are presented in Table (8). Maximum PCV and GCV values were recorded by DTM, NSPP and TGW at Serw (2011/12) where salinity was the highest in this experiment. On the contrary, minimum values of PCV and GCV were recorded at Gemmeiza (2013/14) for 6 out of 9 traits, i.e. DTH, DTM, GFP, GFR, PH, NGPS and GYPP, where the soil salinity was the lowest in this experiment.

Heritability percentages in the broad-sense (Table 9) were at maximum at Sakha (2013/14) for PH, DTM, GFP and GFR and Sakha (2011/12) for NSPP, NGPS, TGW and GYPP and at minimum at Gemmeiza (2013/14) in 5 traits and Sakha (2013/14) in two traits (GYPP and TGW). The highest h^2_b estimates were generally shown by DTM, GFP, GFR, PH and NSPP traits.

The highest expected genetic advance from selection (GA%) in the field, based on 10% selection intensity was observed in Serw

(2011/12), i.e. at the highest salt stressed environment in the present study for 6 out of 9 traits, while the lowest GA% estimates were observed at Gemmeiza 13/14, i.e. at the lowest salt stressed environment in this study for 7 out of 9 traits. In general, the highest GA% estimates were given by GFR, NSPP, NGPS and TGW traits in the field.

Traits showing strong correlations with grain yield, high heritability estimates and high genetic advance from selection in the field experiment were number of spikes/plant and grain filling rate across all environments and NSPP, GFR, GFP and TGW under high salinity conditions (Serw) in this experiment. These traits could be recommended as selection criteria for salinity tolerance in the field.

In wheat high estimates of heritability and genetic advance from selection were reported by several investigators for spikes/plant [27,28,32-34] grain filling period and rate [35-39] under abiotic stress conditions such as salinity and drought. Increases in heritability and additive variance components have been predicted as stress level increases [10,11]; such changes were reported by [12]. Such changes in genetic variation (increase or decrease) would seem likely to arise with increased stress, because different genes may contribute to similar character under diverse environments [13,14]. Based on heritability estimates, Ali et al. [40] suggested that prospects of improving the traits that confer salinity tolerance by selection and crossing are significant if genes controlling salinity tolerance have additive effects. High heritability estimates associated with high genetic advance for major quantitative traits in wheat offer better scope of selection of genotypes in early segregating generations [41]. Heritability estimates assess the relationship in parents and progeny; therefore, crosses have been made to incorporate desirable genes in present wheat varieties to increase the crop productivity. Likewise, Rebetzke and Richards [42] and Sial et al. [43] reported the use of semi-dwarfing genes (Rht1 and Rht2) to develop high yielding cultivars which have resistance against lodging. Munir [44] reported that salinity also affected the heritability of wheat genotypes. Heritability values under salinity stress were found to be lower as compared to the controlled conditions during both phases of experiment. Under 200 mM NaCl, heritability estimates for shoot fresh and dry weights and root dry weight were found above 80%. However, under 250 mM salinity stress

heritability estimates were computed between 59% to 78% for all the seven traits studied. This suggested that high salinity level had more negative effects on heritability. Low heritability under stressed environments and high heritability under non-stressed environment was previously reported by many investigators [34,39,45].

The highest predicted genetic advance (GA%) from selection (Table 9) based on 10% selection intensity was generally observed if selection was done under the most salt stressed environment in this study Serw. At this salt affected location, the estimate of heritability, PCV and GCV for grain yield/plant was 95.52, 79.43, and 77.63%, respectively. Many investigators reported that the optimum selection environment for discrimination of salinity tolerance of a large number of wheat genotypes should be of high salinity level [6]. Based on the results of heritability and predicted genetic advance, there are two contrasting conclusions in the literature about the best selection environment for obtaining stress tolerant genotypes for the use in the target

environment. The first group found that the non-stressed selection environment is the best, because of the higher heritability and expected genetic advance from selection for grain yield than the stressed environment [46,47,48]. However, the second group found that stressed environment is better because of its higher heritability and the higher actual genetic gain from selection than non-stressed environment [11,34,36,37,39,49]. Atlin and Frey [50] reported that selection under salinity stressful environment would ensure the preservation of alleles for salinity tolerance.

There is a third group of investigators that believes that selection should be practiced across a variety of stressed and non-stressed environments to select stress tolerant and high-yielding genotypes for the use in the stressed target environment e.g. [20,39]. It is observed in the present study that the estimates of expected genetic advance for selection are generally high in most studied traits, especially under the

Table 7. Simple correlation coefficients between Grain yield/plant and studied traits across all studied genotypes in 2011/2012 and 2013/2014 seasons at Serw, Sakha and Gemmeiza

Traits	Serw	Sakha	Sakha	Gemmeiza	Combined
	2011/2012	2011/2012	2013/2014	2013/2014	
DTH	0.03	-0.03	-0.052	-0.04	0.16
DTM	0.03	-0.12	0.140	-0.01	0.12
GFP	0.43**	0.35**	0.51**	0.59**	0.68**
GFR	0.99**	0.98**	0.93**	0.95**	0.96**
PH	0.31**	0.08	-0.119	0.04	0.38**
NSPP	0.73**	0.06	0.44**	0.28**	0.65**
NGPP	-0.03	0.28**	0.39**	0.14	0.32**
TGW	0.31**	0.30**	0.36**	0.37**	0.78**
BYPP	-----	-----	0.20**	0.76**	0.29**
SYPP	-----	-----	0.62**	0.52**	0.64**
HI	-----	-----	0.84**	0.60**	0.91**

* and ** indicate significant at 0.05 , 0.01 probability levels, respectively

Table 8. Phenotypic (PCV) and genotypic (GCV) coefficient of variation for selected agronomic traits in DH lines evaluated in the field at Serw, Sakha and Gemmeiza locations

Traits	PCV (%)				GCV (%)			
	Serw	Sakha	Sakha	Gemmeiza	Serw	Sakha	Sakha	Gemmeiza
	11/12	11/12	13/14	13/14	11/12	11/12	13/14	13/14
DTH	13.22	9.40	3.66	3.45	11.96	8.86	3.64	3.29
DTM	10.39	31.20	1.56	1.19	9.43	30.26	1.53	1.05
GFP	41.13	33.06	8.30	5.92	38.76	30.08	8.01	5.59
GFR	1327	716	646	551	1250	667	623	509
PH	28.94	21.41	8.29	7.86	27.95	20.49	8.15	6.68
NSPP	63.15	171.33	26.72	55.47	60.41	164.82	23.36	24.55
NGPS	118.71	81.08	18.38	3.86	108.18	75.05	45.17	1.21
TGW	31.39	54.43	13.61	29.81	26.48	52.81	5.92	16.35
GYPP	79.43	46.73	15.50	22.13	77.63	46.35	5.92	14.05

Table 9. Heritability in the broad-sense (h^2_b %) and expected genetic advance from selection (GA %) for nine agronomic traits in DH lines evaluated in the field at Serw, Sakha and Gemmeiza locations

Traits	h^2_b (%)				GA (%)			
	Serw	Sakha	Sakha	Gemmeiza	Serw	Sakha	Sakha	Gemmeiza
	11/12	11/12	13/14	13/14	11/12	11/12	13/14	13/14
DTH	81.95	88.72	85.03	90.91	19.06	14.68	6.36	5.52
DTM	82.30	94.11	95.32	78.32	15.06	51.67	2.62	1.64
GFP	88.79	82.76	93.07	89.01	64.27	48.16	13.59	9.28
GFR	88.61	86.90	93.08	85.36	2070	1094	1058	827
PH	93.28	91.64	96.72	72.21	47.51	34.53	14.11	9.99
NSPP	91.50	92.54	76.40	19.60	101.70	279.05	35.93	19.13
NGPS	83.04	85.67	46.18	9.83	173.50	122.26	54.03	0.67
TGW	71.18	94.13	18.94	30.09	39.32	90.18	4.54	15.79
GYPP	95.52	98.40	14.60	40.30	133.53	80.92	3.98	15.70

stressed environment (Serw). Over estimation of GA% in the present study in some cases might be attributed to using high estimates of heritability in broad sense, which include both types of genetic variance, i.e. additive, dominance and epistatic genetic variances (heritable and non-heritable components). It is therefore expected that such GA% estimates would be lower in case we used narrow-sense heritability, which depends only on the additive and additive X additive variances (heritable components only). The sufficient variability at both phenotypic and genotypic levels coupled with the high estimates of h^2_b and GA% even under the most salt stressed environment would allow us to conclude that selection for high GYPP, NSPP, GFR, GFP and TGW would result in great progress in such traits and thus improving wheat salinity tolerance. These traits are therefore recommended as selection criteria for salinity tolerance under elevated level of salinity conditions in field experiments.

4. CONCLUSION

Results of the present study concluded that performance, correlations, phenotypic and genotypic variability, heritability and predicted genetic advance from selection for traits of doubled haploids changed from the most salt affected location to the least salt affected location. The highest expected genetic advance (GA%) from selection in the field was observed at the most affected location with salt (Serw) for 6 out of 9 traits, while the lowest GA% estimates were observed at the least affected location with salt (Gemmeiza) for 7 traits. Traits showing strong correlations with grain yield, high heritability estimates and high genetic advance from selection in the field were NSPP, GFR, GFP and TGW under high salinity conditions (Serw);

these traits together with GYPP are recommended as selection criteria for salinity tolerance under field conditions.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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