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Evaluation of Wheat (*triticum aestivum* L.) Accession for Salt Tolerance at Different Growth Stages

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Abstract: Wheat is the major staple food crop worldwide and frequently cultivated on saline soil. Cereals are grown in almost every region of the world and are exposed to a variety of environmental stresses that severely affect their growth and grain yield. As a result, breeding for enhanced salinity tolerance in crop species has received considerable attention as it is an economic and efficient alternative. Characters such as germination, survival and seedling growth or biomass accumulation, have been the most commonly used criteria for identifying salinity tolerance in plants. The present studies was carried out with the major objective of providing information on the extent and basis of genetic variation for salinity tolerance in the wheat varieties at the germination, seedling and production stage. During laboratory screening out of 200 wheat genotypes having higher total germination percentage at different salinity stress levels 18 were selected and promoted to seedling stage screening for further evaluation. Germination percentage was showed significant (P< 0.05) difference among tested wheat genotypes under all salt level. At seedling stage when the salt stress increase the time required for wheat germination was increased. Based on yield data from tested genotypes SANDALL-3, ETBW 5954 and VEE7 showed superior grain yield and the inferior was from ANGL. Ultimately, the results of this research will be a helpful and starting point for working on the validation and registration on future for salt tolerant wheat varieties.

Keywords: wheat, salinity, tolerance.

1. INTRODUCTION

Wheat is the major staple food crop worldwide and frequently cultivated on saline soil. Salinity is the serious factor hampering wheat productivity with adverse effect on germination, growth and final economic yield (Feroz et al., 2017). The continuous salinization of arable land is a threat to global food security. Over 800 Mha of land are affected by salinity, which equates to more than 6% of the world's total land area (FAO 2010) and affects more than 20% of present-day agriculture (Mickelbart et al. 2015). Cereals are grown in almost every region of the world and are exposed to a variety of environmental stresses that severely affect their growth and grain yield (Shahbaz and Ashraf, 2013). As a result, breeding for enhanced salinity tolerance in crop species has received considerable attention as it is an economic and efficient alternative (Ashraf, 2009).

In glycophytes plant species, biochemical, physiological and morphological characteristics are negatively affected, leading to abnormal growth and development and eventual plant death (Nishimura *et al.*, 2011; Beakal *et al.*, 2016; Bethel et al., 2019). Moreover, the highest salinity concentration inhabits percentage of seed germination and emergence of roots due to osmotic effect, which is harmful and prevents the plant in maintaining their appropriate nutritional necessities for their fit development (Kalhoro et al., 2016; Bethel et al., 2019). Most of the crops tolerate salinity to a threshold level and above, where yield decreases as the salinity increases (Khan et al., 2006). Plants growing in saline environments suffer from low osmotic potential of soil solution due to increased concentrations of mineral ions. Eventually, these ions enter into cellular sap, causing ionic imbalance. The negative effects of this disturbance are reflected on plant growth (Perez-Alfocea et al., 1994).



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Plant scientists to overcome the salinity have adopted various strategies. Screening is an effective tool to exploit genetic variation among wheat genotypes (Bahmani et al., 2015; Sajid *et al.*, 2017). Characters such as germination, survival and seedling growth or biomass accumulation, have been the most commonly used criteria for identifying salinity tolerance in plants (Khan et al., 2006). The present study was carried out with the major objective of providing information on the extent and basis of genetic variation for salinity tolerance in the wheat varieties at the germination, seedling and production stage.

2. MATERIALS AND METHODS

2.1. Description of study site

The experiment was conducted at Werer Agricultural Research Center, at three growth stages; germination, seedling and reproductive at laboratory, lath house and field condition, respectively through exposing to varying salt stress condition while selecting relatively tolerant at each growth stages and advancing to next growth stage based on their germination percentage and other parameters.

2.2. Experimental materials and procedures

Germination stage screening

At germination stage a total of 200 wheat genotypes which were collected from Werer and Kulumsa Agricultural Research Centers were studied. The experiment was undertaken through subjecting each wheat genotypes to different salt concentration. Saline solution with an EC value of control (distal water), 15 and 25 dS/m were prepared from NaCl. The experimental design was complete randomized design (CRD) with three replications. Twenty seeds of each genotypes were placed in Petri dish and prepared salt concentration was applied to each experimental unites. Seeds that produce full radical were considered as germinated. First germination count was made at 7th day after treatment application. Because saline environment often cause delay in seed germination, counting was also done once again at 10th day after application of the treatment to take into account late germinated seeds. Eighteen relatively tolerant genotypes with higher germination percentage were selected and advanced to seedling stage screening.

Seedling stage screening

Seedling stage screening was conducted using eighteen genotypes promoted from seedling stage, under lath house condition. Bulk surface soil (non-saline and alkaline in reaction) was collected and packed into pot. Three salinity levels (control, 10 and 20 dS/m) of saline solutions were prepared from NaCl. Treatments were arranged in CRD with three replications. Accordingly prepared saline solutions were added to each pot maintaining to field capacity and ten seeds of each selected varieties were sown per pot. Emerged seedlings were counted at 7 and 10 days after planting and expressed as percentage of seeds that emerged under control condition. Number of tillers, survived plants, and plant height was recorded. Wheat was maintained up to tillering stage and plant and root length were recorded. All the collected data were statistically analyzed and based on recorded observations ten relatively tolerant materials were selected and advanced to field stage screening.

Mean germination time (MGT) which was calculated according to the equation of Ellis and Roberts (1981): MGT = $\Sigma Dn / \Sigma n$.

Where: n= number of seeds which were germinated on day D,

D= number of days counted from the beginning of germination.

Germination Index (GI) which was calculated as described by the Association of Official Seed Analysts (AOSA, 1983) as: $GI = \sum (Gt / Tt)$.

GI= [Number of germinated seeds in first count/ Day of first count] +...+ [Number of germinated seeds in final count/Day of final count]

Germination stress tolerance index = Germination index of stressed treatment/germination index of control treatment * 100.

Field stage screening

Screening for salt tolerance and evaluation for yield performance at field condition was undertaken at Werer Agricultural research center. Top soil (0-30 cm) sample was collected from the experimental site, air dried and sieved through

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a 2 mm screen. The soil was analyzed in the laboratory for some physical and chemical characteristics by standard methods. Based on initial soil sample analysis, the textural class of the experimental site was found to be silt clay. The soils of study area had 12.45% ESP and soluble salt concentration in the soil was 14.05 dS/m as measured in electrical conductivity (ECe) which indicates that the soils of the study site was saline. The treatments were laid out in RCBD with three replications in size of 3 m x 3m plot. Agronomic data was recorded on plant height, effective tiller number, spike length, root length and grain yield.

2.3. Statistical Analysis

All collected data was subjected to the analysis of variance using SAS statistical software. Significant difference between and among treatment means were assessed using the least significant difference (LSD) at 0.05 level of probability (Gomez and Gomez, 1984).

3. RESULT AND DISCUSSION

3.1. Germination stage screening

20 10

Total Germination Percentage (TGP)

Total germination percentage (TGP) of 200 wheat genotypes were significantly reduced by the increasing levels of saline solution. Total germination percentage of all wheat genotypes showed a decreasing trend with increasing salt stress levels (Figure 1). At 0 dS/m level of salinity, most of wheat genotypes showed more than 85% germination percentage, at 15 dS/m for most wheat genotypes germination percentage were between 45 and 70% and at 25 dS/m salt concentration total germination percentage was below 45%, this clearly showed that when the salt stress increase total germination percentage dramatically decrease. Reduced level of seed germination may be due to loss of viability at higher salinity level, delaying germination of seeds at salinities that cause some stress. This result is in lined with different authors (Oyiga et al., 2016; Attri et al., 2018; Bethel at al., 2019). Based on total germination percentage, out of 200 genotypes 18 of them were selected and promoted to seedling stage screening for further evaluation of these genotypes in response to different germination parameters through subjecting at different salt stress condition.

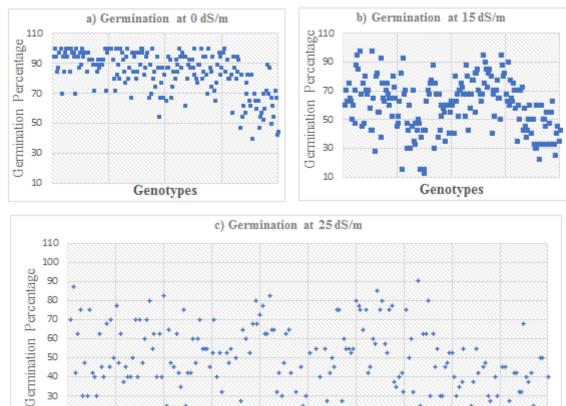


Figure 1: germination percentage of wheat genotypes under different salt level at laboratory

Genotypes

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3.2. Seedling stage Screening

Total Germination Percentage

Analysis of variance (ANOVA) for total germination percentage was showed significant ($P \le 0.05$) difference among tested wheat genotypes at each salt level. The main effect of variety and salt level were highly significant and also the interaction of variety with salt level was significant for total germination percentage. As shown on Table 1 when salt stress increased from 0 to 10 and 20 dS/m germination percentage decreased. Germination percentage did not show consistent trend at germination and seedling stage, because behavior of cultivars varies at both stages. This shows that species /varieties never be selected simply on the basis of higher germination percentage. The same result some varieties appeared to be more sensitive at germination stage. However, it performed quite satisfactorily at seedling stage (Rahman et al., 2008; Homayoun 2011).

Germination mean time

One of the most common problem in saline soil is delay of seed germination. The main effect of application of salt level highly significantly ($P \le 0.001$) affected germination mean time. However, neither the main effect of variety nor the interaction effect of varieties with salt level influenced this parameter (Table 1). In this experiment when the salt stress increase the time required for wheat germination was increased. Average mean germination time (MGT) at control (0), 10 and 20 ds/m salt concentrations were 8.58, 9.72 and 9.97 days, respectively (Table 2). The highest MGT was observed at control in " DEBEIR " cultivar (8.78 days), and the lowest MGT was recorded at control with " VEE7" cultivar (8.50 days). At 20 ds/m salt concentration, all tested genotypes except "PAVON 76" (9.50 days) mean germination time was 10 days. Decrease and delay in germination in saline medium has also been reported by Rahman et al., (2000) and Mirza and Mahmood (1986). Similar result was also observed by Redondo-Gomez *et al.*, (2008) and Hassen *et al.*, (2014). Salinity induced oxidative stress could be a reason for germination inhibition (Amor et al, 2005).

No.		Germination Percentage			Germination Mean Time		
	Treatments	0	10		0	10	20
	(Genotypes)	dS/m	dS/m	20 dS/m	dS/m	dS/m	dS/m
1	PAUON 76	80.00^{abcd}	43.33 ^{abcdef}	23.33 ^{ab}	8.53	9.78	9.50
2	AZAKIA-5	80.00^{abcd}	26.67 ^{def}	10.00^{b}	8.61	10.00	10.00
3	ANGL	83.33 ^{abcd}	26.67 ^{def}	20.00^{ab}	8.56	9.80	10.00
4	ETBW 5954	100.00^{a}	40.00 ^{abcdef}	10.00^{b}	8.50	10.00	10.00
5	ETBW 5510	83.33 ^{abcd}	56.67 ^{abc}	13.33 ^b	8.57	9.26	10.00
6	DEBEIRA	96.67 ^{ab}	23.67 ^{def}	10.00^{b}	8.53	9.80	10.00
7	SANDALL-3	63.33 ^{de}	23.33 ^{def}	10.00^{b}	8.55	10.00	10.00
8	PBW343	76.67 ^{bcd}	20.00^{ef}	10.00^{b}	8.75	10.00	10.00
9	VEE7	53.33 ^e	33.33 ^{bcdef}	10.00^{b}	8.50	10.00	10.00
10	NQALAB 91*2	86.67^{abc}	30.00 ^{cdef}	46.67^{a}	8.54	9.22	10.00
11	JNRB.5/PIFED	70.00^{cde}	63.33 ^a	20.00^{ab}	8.62	9.34	10.00
12	HUBARA-3*2	96.67 ^{ab}	43.33 ^{abcdef}	10.00^{b}	8.61	9.64	10.00
13	WBLL1	86.67^{abc}	60.00^{ab}	46.67^{a}	8.55	9.80	10.00
14	DEBEIRA//TEUEE	86.67 ^{abc}	40.00^{abcdef}	13.33 ^b	8.67	9.41	10.00
15	DEBEIR/	73.33 ^{cd}	50.00 ^{abcd}	46.67^{a}	8.78	9.75	10.00
16	FLORKWA-1	83.33 ^{abcd}	46.67 ^{abcde}	23.33 ^{ab}	8.60	9.80	10.00
17	KAUZ/RAYON//3//	63.33 ^{de}	30.00 ^{cdef}	20.00^{ab}	8.58	9.55	10.00
18	KAUZ'/SERI/4/CHEN	90.00 ^{abc}	16.67^{f}	10.00^{b}	8.53	9.75	10.00
	LSD(0.05)	22.44	28.61	29.36	NS	NS	NS
	CV (%)	16.75	46.09	90.12	1.56	3.39	2.05
Salt		6.20**			0.10**		
Variety		15.19**			NS		
Salt >	Variety	26.91*			NS		

 Table 1: The main and interaction effect of different level of salt on wheat germination percentage and germination mean time

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The same letter within each column for each salt concentration are not significantly different at α = 0.05, based on LSD test. Where: NS (Non-significant); LSD (Least Significant Difference); CV (coefficient of Variance); ** (Significant at α = 0.001) and * (Significant at α = 0.05)

3.3. Field Stage screening

Performance of wheat varieties to Plant height, Effective tiller, Spike length and root length

The result presented on Table 2 showed that plant height, effective tiller number, spike length and grain yield were significantly affected due to variation in tested wheat genotypes under saline soil condition. Maximum plant height (83.51cm) was recorded at ETBW 5954 while minimum value (71.92 cm) was obtained from PBW-343 wheat genotype. Variation in plant height indicate presence of genetic variability among tested genotypes. Effective tiller number was statistically different (Table 2). From the entire genotypes considered the maximum effective tiller numbers were gained by AZAKIA-5 (6.82). However, minimum value (4.98) was recorded at ANGL. Highest spike length was recorded from ANGL (9.09 cm), while lowest spike length (8.16 cm) was obtained from DEBEIRA. From the result root length was not significantly affected by accessions.

Table 2: The response of wheat varieties for plant height, effective tiller number, spike length, root length and grain yield under saline soil condition.

No.		Plant		Spike	Root	Grain
	Wheat	height	Effective	length	length	Yield
	Genotypes	(cm)	tiller (#)	(cm)	(cm)	(kg/ha)
1	PAVON76	78.18 ^{abcd}	5.07 ^{bc}	8.51 ^{ab}	10.18	3112.6 ^{abc}
2	AZAKIA-5	73.09 ^{cd}	6.82 ^a	8.09 ^b	9.78	3104.2 ^{abc}
3	ANGL	81.04^{ab}	4.98 ^c	9.09 ^a	10.22	2728.3 ^c
4	ETBW 5954	83.51 ^a	5.11 ^{bc}	8.97^{a}	11.04	3466.5 ^{ab}
5	DEBEIRA	76.39 ^{bcd}	5.04 ^{bc}	8.16 ^b	10.08	3097.1 ^{abc}
6	SANDALL-3	81.78^{ab}	5.47 ^{bc}	8.54^{ab}	10.51	3709.2 ^a
7	PBW-343	71.92 ^d	6.02^{abc}	8.45^{ab}	11.15	3017.2 ^{bc}
8	VEE7	76.59 ^{bcd}	6.20^{ab}	8.24 ^b	11.5	3327.6 ^{abc}
9	INRB	76.51 ^{bcd}	5.91 ^{abc}	8.99 ^a	9.81	3075.9 ^{abc}
10	GAMBOO	78.82 ^{abc}	6.00 ^{abc}	8.29 ^b	9.4	3230.4 ^{abc}
	LSD(0.05)	6.55	1.19	0.68	NS	691.28
	CV (%)	8.96	22.3	8.45	24.49	23.02

Grain yield

Grain yield showed significant difference ($P \le 0.05$) among tested wheat genotypes, indicating existence of genetic variability among tested genotypes in response to salinity stress tolerance. Highest grain yield of 3709.2 kg/ha was recorded from SANDALL-3 genotype while minimum grain yield of 2728.3 kg/ha was obtained from ANGL genotype. The result clearly suggest genetic variability for salinity response and grain yield significantly affected by salt. High concentrations of sodium (Na+), chloride (Cl-), magnesium (Mg2+), calcium (Ca2+), sulphate (SO4 2-) and bicarbonate (HCO3 -) in soil disturb plant growth and development, ultimately leading to loss in yield (Shahbaz and Ashraf, 2007; Shahbaz et al., 2011; et al., 2012; Perveen Shahbaz et al., 2012; bethel et al., 2019).

4. CONCLUSION AND RECOMMENDATION

During laboratory screening out of 200 wheat genotypes having higher total germination percentage at different salinity stress levels 18 were selected and promoted to seedling stage screening for further evaluation. Germination percentage was showed significant ($P \le 0.05$) difference among tested wheat genotypes under all salt level. At seedling stage when the salt stress increase the time required for wheat germination was increased. Based on yield data from tested genotypes SANDALL-3, ETBW 5954 and VEE7 showed superior grain yield and the inferior was from ANGL. Ultimately, the results of this research will be a helpful and starting point for working on the validation and registration on future for salt tolerant wheat varieties.

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