



## Salt stress affects germination and seedling establishment in different wheat (*Triticum aestivum* L.) varieties

Hina Nazeer<sup>1</sup>, Mamoona Rauf<sup>1</sup>, Humaira Gul<sup>1</sup>, Tabassum Yaseen<sup>2</sup>, Adnan Ali Shah<sup>3</sup> and Khushnood Ur Rehman<sup>4\*</sup>

<sup>1</sup>Department of Botany, Abdul Wali Khan University, Mardan, Pakistan (AWKUM)

<sup>2</sup>Department of Botany, Bacha Khan University, Charsadda, Pakistan

<sup>3</sup>Department of Botany, University of Peshawar, Pakistan

<sup>4</sup>Department of Botany, Islamia College Peshawar, Pakistan

\*Corresponding author: Khushnood Ur Rehman (drkhushnood@icp.edu.pk)

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**Key Message:** The present study evaluated the effect of salt stress in wheat. The percentages of all parameters were reduced, while the root shoot ratio increased in *Triticum aestivum* under salt stress. Lalma and Insaf were found to be more salt-tolerant varieties.

**Abstract:** Salt stress is a very important factor worldwide as it reduces the agricultural production of many crops. This project was aimed to determine the effect of salt stress on the process of germination and seedling establishment of six *Triticum aestivum* varieties (Fakhr e Sarhad, Insaf, Lalma, Tatora, Bathoor, and Barsat). Five seeds of six *Triticum aestivum* cultivars were placed in each Petri-plate. Seeds were treated with different concentrations (0, 50 mM, and 150 mM) of NaCl. After 8 days, the experiment was terminated and germination percentage, seedling length, seedlings biomass were recorded, while relative water content (RWC), vigor index (VI), root shoot ratio (RSR), stem weight ratio (SWR), root weight ratio (RWR), and stress tolerance index of different parameters were calculated. Analyzing results, it was observed that salt stress damage plant at observed stages in the form of the

drastic and significant reduction in germination percentage, shoot length, root length, fresh weight, dry weight, RWC, VI, plant height stress tolerance index (PHSI), root length stress tolerance index (RLSI), stem fresh weight tolerance index (SFTI), root fresh weight stress tolerance index (RFTI), stem dry weight stress tolerance index (SDTI), root dry weight stress tolerance index (RDTI), SWR, and RWR while RSR. Reduction percentages of different parameters were increased as salt stress levels. In this study, it was concluded, among six cultivars, variety Lalma showed higher salt-tolerance (@150mM NaCl) at germination-stage (0% reduction) as well as seedling-establishment stage as compared to the other cultivars based on measured parameters shoot length (57% reduction), root length (32% reduction), fresh weight (38% reduction), dry weight (40% reduction), RWC (21% reduction), VI (35% reduction), SWR (69% reduction), RWR (63% reduction), RSR (297% increase). This variety can be used for cultivation on soils suffering from salinity stress in Pakistan. © 2020 Department of Agricultural Sciences, AIOU

**Keywords:** Germination, Relative water contents, Seedling growth, Stress tolerance index, Vigor index, Wheat

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### Introduction

The abiotic stress is known as salinity all over the world and especially in arid and semi-arid areas harms plant growth and yields (Abbasi et al., 2016; Rani et al., 2019). According to Okorogbona et al. (2018) these elements mark nearly one-third of the world's irrigated land. When a plant is exposed to a salt stress condition, this factor disturbs the normal metabolism of the plant, as a result, the plant's growth and its productivity are reduced (Abbasi et al., 2014). When seeds are sown in a saline environment, these seeds with low osmotic-potential don't absorb water in a saline medium, accumulation process of different toxic ions (such as Na<sup>+</sup> and Cl<sup>-</sup>) increases, finally, the process of seed-germination first delayed, reduced, and disrupted.

This feature also triggered a negative impression on the germination process, its percentage, and seedling-growth (Agnihotri et al., 2006).

Wheat is an important crop worldwide and grown in the spring and winter seasons, but the winter-seasoned crop is more effective than the spring-seasoned crop (Tsegaye et al., 2012). Common-wheat which is also known as bread-wheat, these cultivars have white/red bran-color, winter/spring habit with hard/soft endosperm (Chachaiya et al., 2017). As the problem of salinity is quite important world-wide, so different strategies used to overcome harmful effects of this problem on plant growth and metabolism, e.g. salt-tolerant cultivars production using the breeding program, foliar application of different metabolites, etc. plant breeding-program is very useful for the production of salt-tolerant- and more yield

producing lines, while its use is limited as a result of multi-genic nature of salt-tolerance as well as bulky genetic deviation in crops (Turan et al., 2014). For improvement of salt tolerance, this breeding program needs effective screening techniques for the selection and then evaluation of specified traits. Another requirement is the recognition of genetic-variability with information of tolerance-traits inheritance and mechanism of tolerance mechanism in the biological system on different stages of development of plants. It is also necessary to use reliable and suitable design and indirect and direct selection criteria, most appropriate breeding methodology for the transfer of tolerance characters to better genetic-background (Ashraf & Foolad, 2013).

The salinity in the world is increasing day by day so it's important to investigate different wheat germplasm for detection of salt tolerance as well as for high yield production under salt stress which is different for different genotypes that show diverse salt tolerance capacities (Khan & Panda, 2008). Therefore, different crop cultivars have been produced to get more yield of the specific-crop. So the current study was designated to show how much the local wheat varieties are affected by applying salt stress at different stages of development (germination and seedling growth stage).

## Materials and Methods

Different varieties of *Triticum aestivum* L. (Fakhr e Sarhad, Insaf, Lalma, Tatora, Bathoor, and Barsat) were collected from Agriculture Research Institute, Tarnab Peshawar, and in the plant physiology lab Department of

Vigor index (VI) = (Mean root length + Mean shoot length) × Germination percentage

### Root shoot ratio, shoot weight ratio and root weight ratio

Different ratios in experimental seedlings were calculated through different formulae described by Hunt (1982):

$$\text{Root shoot ratio (RSR)} = \frac{\text{Root dry weight}}{\text{Shoot dry weight}}$$

$$\text{Shoot weight ratio (SWR)} = \frac{\text{Shoot dry weight}}{\text{Total dry weight}}$$

$$\text{Plant height stress tolerance index (PHSI)} = \frac{\text{Plant height of stressed plants}}{\text{Plant height of control plants}} \times 100$$

$$\text{Root length stress tolerance index (RLSI)} = \frac{\text{Stressed plants radicle extension}}{\text{Control plants radicle extension}} \times 100$$

$$\text{Shoot fresh weight stress tolerance index (SFSI)} = \frac{\text{Stressed plants sprout fresh mass}}{\text{Control plants sprout fresh mass}} \times 100$$

$$\text{Root fresh weight stress tolerance index (RFSI)} = \frac{\text{Stressed plants root fresh mass}}{\text{Control plants root fresh mass}} \times 100$$

$$\text{Shoot dry weight stress tolerance index (SDSI)} = \frac{\text{Stressed plants shoot dry mass}}{\text{Control plants shoot dry mass}} \times 100$$

Botany AWKUM. The 0.1% HgCl<sub>2</sub> (mercuric-chloride) solution was taken and the wheat varieties seeds were surface-sterilized for 1- minute and then washed three times with D.H<sub>2</sub>O. Sterilized plates lined with two layers of filter paper. Five seeds of six *Triticum aestivum* cultivars were placed in each sterilized Petri plate. Plates were applied with 5 ml distilled water/ 50 mM, 150 mM NaCl concentrations as per the design, and each treatment was replicated three times. A total replicate was placed in an incubator at a temperature of 25 °C for germination for 24 hours. After one day the data was collected from germinated seeds. After one week the observation was concluded and various parameters of seedling were determined (germination percentage, seedling growth, seedling length, seedlings biomass relative water content, vigor index, RSR, SWR, and RWR). Average values of the shoot and root length were noted in cm, the seedling was taken and their fresh weight was noted, and for the determination of dry weight the plant's samples were kept in the oven for 2 days at 50 °C to make it dry.

### Relative water contents

Relative water contents (RWC) were measured by process as defined by Epstein & Grant (1973):

$$\text{RWC (\%)} = \frac{\text{Fresh weight} - \text{Dry weight}}{\text{Turgid weight} - \text{Dry weight}} \times 100$$

### Vigor index

Seedling vigor index (VI) was designed in experimental seedling through technique as pronounced by Abdul-Baki & Anderson (1973):

$$\text{Root weight ratio (RWR)} = \frac{\text{Root dry weight}}{\text{Total dry weight}}$$

### Stress tolerance index

Stress tolerance index (STI) of the parameters of experimental seedling were considered through diverse formulae as defined by Pour-Aboughadareh et al. (2017):

$$\text{Root dry weight stress tolerance index (RDSI)} = \frac{\text{Stressed plants sprout dry mass}}{\text{Control plants root dry mass}} \times 100$$

### Reduction percentages

Reduction percentages of different parameters of experimental seedlings were calculated through different formulae as described by Berova et al. (2002):

$$\text{Shoot length reduction percentage (SLRP)} = \frac{1 - (\text{Shoot length at salt stress})}{\text{Shoot length control}} \times 100$$

$$\text{Root length reduction percentage (RLRP)} = \frac{1 - (\text{Root length at salt stress})}{\text{Root length control}} \times 100$$

$$\text{Fresh weight reduction percentage (FWRP)} = \frac{1 - (\text{Fresh weight at salt stress})}{\text{Fresh weight control}} \times 100$$

$$\text{Dry weight reduction percentage (DWRP)} = \frac{1 - (\text{Dry weight at salt stress})}{\text{Dry weight control}} \times 100$$

### Statistical analysis

SPSS (Version 21) statistical software was used to analyze obtained data. One-way ANOVA (analysis of variance) was made to find probability levels, while diverse mean values were measured through Duncan's Multiple-Range-Test (DMRT) at a 5% level of probability.

## Results and Discussion

### Germination percentage

In-plant life cycle, germination-process, and seedling-establishment is a key and important phase and this phase exhibited susceptibility to stress conditions (e.g. salinity) present in the environment by different species (Liu et al., 2018). Sholi (2012) performed experiments on different crops under salinity stress and observed that the germination-process with seedling-growth was severely affected through the stress-environment. During the present study, results confirmed that salt stress application @ 50 and 150 mM had an inhibitory effect on germination percentage on Lalma and Tatora, while these concentrations enhanced this parameter in Insaf and Barsat as compared to control. Further, two cultivars named Bathoor and Fakhr e Sarhad exhibited equal germination percentages in both salinity levels as compared to control (Table 1). (Riaz et al., 2019) Observed reduced germination percentage in plants after growing in salt stress and stated that this phenomenon was caused as a result of osmotic-stress and toxicity of specific ions. Muhammad and Hussain (2010) observed the same results of reduced germination percentage when grown *Vigna mungo* and *Brassica juncea* under salt stress. Zapata et al. (2003) worked on *Lactuca sativa* and pepper plants respectively and observed a reduction in germination

percentage when grown in different salinity stress. Rolly et al. (2020) reported the reduced germination percentage after NaCl application and argued that salt application causes inhibition of enzyme activities which had a critical role in the germination process.

**Table 1** Effect of salinity on germination percentage of different wheat varieties

Varieties	Germination Percentage (%)		
	Control	50 mM NaCl	150 mM NaCl
F.S.	93.33±6.66 <sup>b</sup>	93.33±6.66 <sup>a</sup>	93.33±6.66 <sup>ab</sup>
Insaf	86.66±6.66 <sup>b</sup>	100±0 <sup>a</sup> (+15.384)	93.33±6.66 <sup>ab</sup> (+7.692)
Lalma	100±0 <sup>b</sup>	93.33±6.66 <sup>a</sup> (-6.666)	100±0 <sup>b</sup>
Tatora	100±0 <sup>b</sup>	100±0 <sup>a</sup>	93.33±6.66 <sup>ab</sup> (-6.666)
Bathoor	100±0 <sup>b</sup>	100±0 <sup>a</sup>	100±0 <sup>b</sup>
Barsat	66.66±6.66 <sup>a</sup>	93.33±6.66 <sup>a</sup> (+40)	80±0 <sup>a</sup> (+20)

F.S. = Fakhr e Sarhad; In columns values are treatment means with ± SD; In a column, treatment means with different letters are statistically significant at 5% level of probability; Figures in parentheses indicate % promotion (+) and reduction (-) over control.

### Shoot and root length

When salt stress is applied to plants, it creates osmotic pressure which further reduces the water absorption process through roots, which results in the reduced division of cells and differentiation process hence causing the decreased length of

radicle and plumule. After analyzing the results of the present study it was seen that shoot-length and root-length of seedlings in different varieties were significantly reduced after application of salt. According to results regarding shoot length, at 50 mM NaCl concentration, shoot length showed 9%, 30%, 3%, 49%, 15%, 30% reduction, while at 150 mM NaCl level same parameter exhibited 50%, 61%, 57%, 79%, 48% and 45% reduction in Fakhr e Sarhad, Insaf, Lalma, Tatora, Bathoor and Barsat, respectively as compare to control seedlings (Table 2). Lalma and Bathoor showed more tolerance in shoot length as compared to other varieties. Results indicate that, at 50 mM NaCl concentration, root-extension exhibited 26.1%, 27.2%, 13.5%, 21.6%, 22.1% and 26.7% reduction, while at 150 mM NaCl level same parameter exhibited 54.9%, 45.8%, 33.1%, 48.7%, 45.1% and 45.4% reduction in Fakhr e Sarhad, Insaf, Lalma, Tatora, Bathoor and Barsat, respectively (Table 3). Insaf and Lalma showed more tolerance in root-length as compared to other varieties. Ratnakar and Rai (2013) observed a reduction in root length and shoot length which is caused by toxicity of different ions, reduced absorption, and disproportion of nutrients by seedlings when raised under salt stress. The application of high salt levels on plants caused sensitivity of seedling length which was due to low nutrients transition from cotyledons to caulicle under saline condition (Hasaneen et al., 2009). Further, this stress also disturbed the uptake of water through seeds which results in reduced production of enzymes and phytohormones which further leads to reduced seedling growth (Hasaneen et al., 2009). Hewage et al. (2020) suggested that the presence of salt in the growth medium initially worked on the water absorption process and decreased this process through reduction of cell turgor and shoot and root elongation. This reduction in root and shoot length also causes reduced absorption of essential nutrients from the soil (Abbasi et al., 2016) with the enhanced intra-cellular level of both chloride and sodium which in-turn inhibit metabolic activity of expanding and dividing cells and reduce germination.

**Table 2** Effect of salinity on shoot length (cm) of different wheat varieties

Varieties	Shoot length (cm)		
	Control	50 mM NaCl	150 mM NaCl
F.S.	5.8±0.62 <sup>a</sup>	5.28±0.88 <sup>ab</sup> (-8.989)	2.88±0.79 <sup>ab</sup> (-50.287)
Insaf	9.24±0.59 <sup>b</sup>	6.48±0.26 <sup>ab</sup> (-29.920)	3.64±0.41 <sup>b</sup> (-60.598)
Lalma	8.17±0.49 <sup>b</sup>	7.97±0.83 <sup>b</sup> (-2.508)	3.49±0.41 <sup>ab</sup> (-57.259)
Tatora	7.69±1.04 <sup>ab</sup>	3.9 ± 1.33 <sup>a</sup> (-49.262)	1.61±0.65 <sup>a</sup> (-79.011)
Bathoor	8.35±0.59 <sup>b</sup>	7.13±1.21 <sup>b</sup> (-14.536)	4.35±0.80 <sup>b</sup> (-47.923)
Barsat	8.77±0.37 <sup>b</sup>	6.16±0.62 <sup>ab</sup> (-29.833)	4.87±0.39 <sup>b</sup> (-44.520)

F.S. = Fakhr e Sarhad; In columns values are treatment means

with ± SD; In a column, treatment means with different letters are statistically significant at 5% level of probability; Figures in parentheses indicate % promotion (+) and reduction (-) over control.

**Table 3** Effect of salinity on root length (cm) of different wheat varieties

Varieties	Root length (cm)		
	Control	50 mM NaCl	150 mM NaCl
F.S.	15.67±1.11 <sup>ab</sup>	11.55±0.73 <sup>ab</sup> (-26.332)	7.02±1.01 <sup>abc</sup> (-55.237)
Insaf	17.32±0.89 <sup>b</sup>	12.55±0.56 <sup>ab</sup> (-27.521)	9.32±0.05 <sup>bcd</sup> (-46.179)
Lalma	17.31±0.17 <sup>b</sup>	15.01±1.07 <sup>b</sup> (-13.260)	11.69±0.81 <sup>d</sup> (-32.473)
Tatora	13.21±2.14 <sup>a</sup>	10.37±2.25 <sup>ab</sup> (-21.493)	6.69±0.75 <sup>ab</sup> (-49.344)
Bathoor	17.05±0.69 <sup>b</sup>	13.17±2.43 <sup>ab</sup> (-22.752)	9.52±1.36 <sup>cd</sup> (-44.175)
Barsat	11.96±1.13 <sup>a</sup>	8.82±0.31 <sup>a</sup> (-26.269)	6.4±0.41 <sup>a</sup> (-46.479)

F.S. = Fakhr e Sarhad; In columns values are treatment means with ± SD; In a column, treatment means with different letters are statistically significant at 5% level of probability; Figures in parentheses indicate % promotion (+) and reduction (-) over control.

### Root shoot ratio

Different ratios are applied to evaluate the segregating of dry stuff amongst root and shoot, among these ratios root to shoot ratio is considered as one of the good indicators for measuring tolerance of any plant to particular environmental stress conditions (Boutraa et al., 2010). Applying various doses of salt (50 mM and 150 mM) on wheat cultivars revealed escalation in root/shoot ratio. In present study, at 50 mM NaCl concentration, root shoot ratio showed 75%, 161%, 167%, 46%, 90% and 55% promotion while at 150 mM NaCl level same parameter exhibited 187%, 279%, 297%, 118%, 156% and 139% elevation in Fakhr e Sarhad, Insaf, Lalma, Tatora, Bathoor and Barsat individually (Table 4). Lalma exhibited supplementary tolerance in root shoot ratio as compared to other varieties. According to the studies of (Acosta-Motos et al., 2017), they considered this ratio (root to shoot ratio) was a common comeback of diverse plant species to diverse environmental stresses. When a large root area is exposed to a salt stress environment, toxic ions retain in this organ more and control the translocation of these ions into the shoot. This process creates a typical resistance/survival mechanism in the plant under any applied environmental stress condition (Acosta-Motos et al., 2017). Haque et al. (2016) also explained it after working on melon genotypes under salt stress as a priority of a plant is to allocate photoassimilate to the root portion in detriment to the above-ground portion, so, helps the plant to improve plant tolerance to applied salt in the growth medium. Partitioning of assimilates is a complex process and mostly controlled by sinks and sources in plants.



**Table 4** Effect of salinity on root shoot ratio (RSR) of different wheat varieties

Varieties	Root Shoot Ratio (RSR)		
	Control	50 mM NaCl	150 mM NaCl
F.S.	0.26±0.01 <sup>a</sup>	0.46±0.01 <sup>a</sup> (+74.802)	0.76±0.02 <sup>a</sup> (+186.903)
Insaf	0.21±0.08 <sup>a</sup>	0.54±0.07 <sup>a</sup> (+161.448)	0.78±0.08 <sup>a</sup> (+279.408)
Lalma	0.23±0.03 <sup>a</sup>	0.61±0.02 <sup>a</sup> (+166.720)	0.9±0.01 <sup>a</sup> (+297.468)
Tatora	0.38±0.05 <sup>a</sup>	0.55±0.05 <sup>a</sup> (+45.694)	0.83±0.05 <sup>a</sup> (+118.067)
Bathoor	0.33±0.04 <sup>a</sup>	0.62±0.03 <sup>a</sup> (+90.464)	0.84±0.05 <sup>a</sup> (+156.416)
Barsat	0.35±0.07 <sup>a</sup>	0.54±0.05 <sup>a</sup> (+54.668)	0.83±0.09 <sup>a</sup> (+138.607)

F.S. = Fakhr e Sarhad; In columns values are treatment means with  $\pm$  SD; In a column, treatment means with different letters are statistically significant at 5% level of probability; Figures in parentheses indicate % promotion (+) and reduction (-) over control.

### Fresh and dry biomass

Salt application on plants causes water deficiency in plants and as a result, it alters different physiological processes and causes damage to membranes which in-turn cause reduced fresh and dry biomass of seedlings (Ali et al., 2020). According to the present investigation application of different salt, concentrations caused a continuous and significant reduction in fresh and dry biomass as compared to the control set. Presently, at 50 mM NaCl concentration,

fresh mass displayed 41.2%, 3.7%, 7.8%, 23.4%, 25.2% and 32.3% reduction while at 150 mM NaCl level same parameter exhibited 60.9%, 40.2%, 37.9%, 68.3%, 43.8% and 41.9% reduction in Fakhr e Sarhad, Insaf, Lalma, Tatora, Bathoor and Barsat respectively. Insaf showed more tolerance in fresh biomass as compared to other varieties. In present study, at 50 mM NaCl concentration, dry mass revealed 49.9%, 0.98%, 14.6%, 46.2%, 5.7% and 79.4% reduction, while at 150 mM NaCl level same parameter exhibited 58.2%, 31.1%, 40.2%, 64.3%, 28.5% and 80.9% reduction in Fakhr e Sarhad, Insaf, Lalma, Tatora, Bathoor and Barsat, respectively (Table 5). Barsat and Lalma revealed supplementary tolerance in dry biomass as compared to other varieties. (Hussain et al., 2013) grew black seeds under different salinity levels and observed a significant decrease in fresh and dry biomass. (Hussain et al., 2013) supported the same results in their studies and explained that reduction in biomass in response to salt might be caused due to osmotic stress and accretion of specific ions (e.g. sodium and chloride). Zafar et al. (2015) also observed reduced shoot length and fresh and dry biomass due to salt stress in different genotypes used in their study. Further, Nadeem et al. (2020) grew *Oryza sativa* L. under salt stress and observed different salt-induced changes in the germination process, fresh and dry biomass, and different physiological characters which further cause a reduction in the germination process and early seedling growth. In another study, (Amiri et al., 2015) worked on *Cynara scolymus* and *Echinacea purpurea* and grew these medicinal plants under salt stress and observed a reduction in germination percentage and seedling growth.

**Table 5** Effect of salinity on fresh and dry weight (g) of different wheat varieties

Varieties	Fresh weight (g)			Dry weight (g)		
	Control	50 mM NaCl	150 mM NaCl	Control	50 mM NaCl	150 mM NaCl
F.S.	0.79±0.03 <sup>a</sup>	0.47±0.03 <sup>a</sup> (-40.928)	0.31±0.01 <sup>a</sup> (-61.101)	0.09±0.006 <sup>a</sup>	0.05±0.004 <sup>a</sup> (-50.017)	0.04±0.001 <sup>a</sup> (-57.667)
Insaf	1.14±0.09 <sup>b</sup>	1.09±0.09 <sup>c</sup> (-3.519)	0.69±0.05 <sup>b</sup> (-39.589)	0.07±0.006 <sup>a</sup>	0.07±0.002 <sup>ab</sup> (-0.920)	0.05±0.006 <sup>a</sup> (-30.939)
Lalma	1.05±0.02 <sup>b</sup>	0.97±0.03 <sup>bc</sup> (-7.594)	0.65±0.03 <sup>b</sup> (-38.291)	0.09±0.001 <sup>a</sup>	0.07±0.002 <sup>b</sup> (-14.350)	0.05±0.004 <sup>a</sup> (-39.863)
Tatora	0.86±0.04 <sup>d</sup>	0.65±0.02 <sup>ab</sup> (-24.418)	0.28±0.07 <sup>a</sup> (-67.728)	0.13±0.001 <sup>ab</sup>	0.07±0.001 <sup>ab</sup> (-45.724)	0.05±0.001 <sup>a</sup> (-63.825)
Bathoor	1.08±0.01 <sup>b</sup>	0.82±0.01 <sup>abc</sup> (-24.092)	0.6±0.03 <sup>b</sup> (-44.307)	0.07±0.001 <sup>a</sup>	0.07±0.004 <sup>ab</sup> (-5.393)	0.05±0.004 <sup>a</sup> (-27.908)
Barsat	0.84±0.06 <sup>a</sup>	0.58±0.03 <sup>ab</sup> (-31.620)	0.49±0.01 <sup>ab</sup> (-42.292)	0.23±0.08 <sup>b</sup>	0.05±0.001 <sup>a</sup> (-78.704)	0.04±0.009 <sup>a</sup> (-81.263)

F.S. = Fakhr e Sarhad; In columns values are treatment means with  $\pm$  SD; In a column, treatment means with different letters are statistically significant at 5% level of probability; Figures in parentheses indicate % promotion (+) and reduction (-) over control.

### Relative water contents

Plant growth and physiology depend on relative water content present in it, so the relative water content of any plant exhibits hydration level of different tissues within the plant while a high relative water content level is essential for plant better growth (Rao et al., 2012). In a current study, NaCl was applied in different concentrations (50 mM and 150 mM) on wheat cultivars and they exhibited an inhibitory effect on relative water contents when it was

linked with standard or control. Currently, at 50 mM NaCl concentration, RWC showed 4.5%, 11.2%, 10.9%, 7.25%, 5.8% and 3.39% reduction while at 150 mM NaCl level same parameter exhibited 16.2%, 15.5%, 18.2%, 23%, 25.2% and 20.2% reduction in Fakhr e Sarhad, Insaf, Lalma, Tatora, Bathoor and Barsat correspondingly (Table 6). Insaf and Barsat showed more tolerance in relative water content as compared to other varieties. (Haque et al., 2016) studied mung beans under salt stress and observed a reduction in relative water content and water holding capacity with an increase in water

saturation deficit and water uptake capacity. In another study, (Aydi et al., 2008) observed reduced relative water content (RWC) as a result of the direct response of salt stress on ions accumulation, reduced osmotic adjustment, and water potential.

### Vigor index

The vigor index of the seedling can be defined as the measure of damaging ability accumulation that causes viability decline of any seed or seed that is unable to germinate/die (Marcos Filho, 2015). This parameter has been used as a tool for measuring the tolerance index of any plant species under any environmental stress condition. In the case of seed, this parameter is used for germination rate and growth rate (Buriro et al., 2011). In a current study, NaCl was applied in different concentrations (50 mM and 150 mM) on wheat cultivars and they

exhibited a reduction in vigor index when it was linked with standard or control (Table 6). Presently, at 50 mM NaCl concentration, vigor index showed 27.2%, 19.9%, 18.8%, 48.9%, 22.9% and 33.1% reduction while at 150 mM NaCl level same parameter exhibited 49.3%, 46.2%, 36.3%, 54.2%, 47.1% and 55.9% reduction in Fakhr e sarhad, Insaf, Lalma, Tatora, Bathoor and Barsat individually. Lalma revealed supplementary tolerance in vigor index as compared to other varieties (Table 6). El Harfi et al. (2021) explained that the lower level of vigor index of any seed is a result of reduced water potential and the negative impact of specific ions. Studies by Saeidnia et al. (2014) observed reduced length of seedlings as a result of the low level of seed's vigor index. Many studies reported a decrease in the vigor index of seedlings in response to environmental stress especially salinity which arose as a result of inhibition of seedling's initial growth by applied stress factor (Borlu et al., 2018).

**Table 6** Effect of salinity on relative water content and vigor index of different wheat varieties

Varieties	Relative water content (RWC)			Vigor index (VI)		
	Control	50 mM NaCl	150 mM NaCl	Control	50 mM NaCl	150 mM NaCl
F.S.	86.49±2.65 <sup>a</sup>	76.01±3.6 <sup>a</sup> (-12.121)	58.89±3.66 <sup>a</sup> (-31.921)	14.86±0.92 <sup>a</sup>	10.86±2.97 <sup>a</sup> (-26.923)	7.58±1.18 <sup>ab</sup> (-48.979)
Insaf	83.59±0.57 <sup>a</sup>	73.38±5.56 <sup>a</sup> (-12.217)	66.08±6.18 <sup>a</sup> (-20.942)	16.61±1.36 <sup>a</sup>	13.36±0.59 <sup>a</sup> (-19.582)	9.03±0.60 <sup>abc</sup> (-45.666)
Lalma	78.33±3.36 <sup>a</sup>	71.27±1.14 <sup>a</sup> (-9.006)	61.88±5.25 <sup>a</sup> (-21.002)	18.71±0.25 <sup>a</sup>	15.08±0.34 <sup>a</sup> (-19.415)	12.09±0.88 <sup>c</sup> (-35.358)
Tatora	78.76±5.25 <sup>a</sup>	69.26±6.15 <sup>a</sup> (-12.063)	59.9±6.82 <sup>a</sup> (-23.940)	14.27±2.41 <sup>a</sup>	10.83±2.45 <sup>a</sup> (-24.083)	6.34±0.82 <sup>b</sup> (-55.594)
Bathoor	83.44±0.14 <sup>a</sup>	74.76±2.19 <sup>a</sup> (-10.405)	57.08±3.98 <sup>a</sup> (-31.591)	18.46±0.66 <sup>a</sup>	14.16±2.73 <sup>a</sup> (-23.271)	9.95±1.50 <sup>bc</sup> (-46.109)
Barsat	82.01±0.27 <sup>a</sup>	75.74±3.34 <sup>a</sup> (-7.642)	57.09±3.18 <sup>a</sup> (-30.389)	13.76±4.91 <sup>a</sup>	9.34±1.37 <sup>a</sup> (-32.081)	6.05±0.57 <sup>a</sup> (-56.045)

F.S. = Fakhr e Sarhad; In columns values are treatment means with ± SD; In a column, treatment means with different letters are statistically significant at 5% level of probability; Figures in parentheses indicate % promotion (+) and reduction (-) over control.

### Shoot/stem weight ratio

In a current study, NaCl was applied in different concentrations (50mM and 150mM) on wheat cultivars and they exhibited a reduction in shoot weight ratio when it was linked with standard or control. Currently, at 50 mM NaCl concentration, shoot weight ratio showed 21%, 24%, 43%, 26%, 32% and 27% reduction while at 150 mM NaCl level same parameter exhibited 45%, 59%, 69%, 54%, 47% and 51% reduction in Fakhr e Sarhad, Insaf, Lalma, Tatoora, Barsaat and Bathoor, respectively. Fakhr e Sarhad and Tatora showed more tolerance in stem weight ratio as compared to other varieties (Table 7). Tewari & Arora, 2016 studied sunflower under different NaCl concentrations and observed reduced vegetative growth of this plant. In another study, Noreen et al. (2020) observed plants under different salinity levels and reported a reduction in root shoot ratio on a dry weight basis.

### Root weight ratio

Different salt stress levels cause a drop in root growth and this organ is considered the most sensitive organ. Low level of oxygen availability under salt stress conditions deprives plants of energy source while amassing of a great ethylene level reduced growth of roots (Abid et al., 2018). This phenomenon of reduced plant growth and root weight ratio, under salt stress, was also observed by different researchers on different crops (García Morales et al., 2012). In a current study, NaCl was applied in different concentrations (50 mM and 150 mM) on wheat cultivars and they exhibited a reduction in root weight ratio when it was linked with standard or control. In present project, at 50 mM NaCl concentration, root weight ratio showed 22%, 19%, 32%, 29%, 43% and 35% reduction while at 150 mM NaCl level same parameter exhibited 45%, 50%, 63%, 66%, 62% and 57% reduction in Fakhr e Sarhad, Insaf, Lalma, Tatoora, Barsaat and Bathoor respectively (Table 7).

Insaf and Tatora showed more tolerance in root weight ratio as compared to other varieties. Experiments of Khan et al. (2020) on different plant species revealed that shoot and root growth showed a reduction in response to salt stress and was considered a sensitive species. In another experiment by (Rahman et al., 2017), they grown rice plants under salinity and observed a reduction in root weight ratio and shoot weight ratio on a dry weight basis of the plant.

### Salt tolerance index

It is a well-known and common phenomenon that salt application on plants causes a reduction in plumule and radicle length, biomass, and plant height. When different varieties of wheat were grown under salt stress, genotypic-variation was evaluated after the estimation of different physiological parameters with the help of different growth parameters. All physiological indices (PHSI, RLSI, SFSI, RFSI, SDSI, and RDSI) were diminished because of salinity viewing that the growth was suggestively affected

by salt stress. At 50 mM NaCl concentration Lalma showed the highest values of PHSI, RLSI, RFSI, Bathoor showed a high value of SDSI, Insaf showed high values in SFSI, FBSI, RDSI, and DBSI. At 150mM NaCl concentration, Bathoor showed a high value of PHSI, Insaf showed a high value of RDSI, Lalma showed high values of RLSI, RFSI, FBSI, Barsaat showed high values of SFSI, SDSI, DBSI. The varieties Insaf and Lalma had higher tolerance indices reflecting greater salt tolerance (Table 8). Fahad et al. (2017) worked on different crops under stress conditions and observed reduced seed germination and seedling growth with a huge variation of genotypes in response to applied stress. Munns (2005) observed a reduction in the growth of varieties under stress conditions, could be possible as a result of two reasons. Firstly, under stress conditions plants are unable to absorb water properly through the soil as a result of water deficit or osmotic stress created after applied stress. Secondly, absorption of toxic ions as chlorides and sodium which are transported and entered the transpiration stream injure cells in leaves and ultimately reduce photosynthetic rate and growth.

**Table 7** Effect of salinity on stem weight ratio and root weight ratio of different wheat varieties

Varieties	Stem weight ratio (SWR)			Root weight ratio (RWR)		
	Control	50 mM NaCl	150 mM NaCl	Control	50 mM NaCl	150 mM NaCl
F.Sarhad	0.86±0.04 <sup>a</sup>	0.68±0.06 <sup>b</sup> (-20.814)	0.48±0.04 <sup>a</sup> (-44.818)	0.41±0.04 <sup>a</sup>	0.32±0.06 <sup>a</sup> (-22.387)	0.23±0.02 <sup>a</sup> (-44.778)
Insaf	0.8±0.08 <sup>a</sup>	0.61±0.01 <sup>ab</sup> (-23.720)	0.33±0.01 <sup>a</sup> (-59.053)	0.6±0.01 <sup>ab</sup>	0.49±0.09 <sup>a</sup> (-18.554)	0.3±0.05 <sup>a</sup> (-49.859)
Lalma	0.77±0.02 <sup>a</sup>	0.44±0.09 <sup>a</sup> (-43.333)	0.24±0.03 <sup>a</sup> (-69.246)	0.83±0.06 <sup>b</sup>	0.56±0.09 <sup>a</sup> (-32.249)	0.31±0.08 <sup>a</sup> (-62.823)
Tatora	0.77±0.02 <sup>a</sup>	0.57±0.01 <sup>ab</sup> (-26.356)	0.36 ± 0.06 <sup>a</sup> (-52.443)	0.8±0.07 <sup>b</sup>	0.57±0.05 <sup>a</sup> (-29.097)	0.27±0.02 <sup>a</sup> (-66.048)
Bathoor	0.91±0.04 <sup>a</sup>	0.62±0.01 <sup>ab</sup> (-31.993)	0.49 ± 0.06 <sup>a</sup> (-46.610)	0.77±0.04 <sup>b</sup>	0.44±0.04 <sup>a</sup> (-43.033)	0.29±0.04 <sup>a</sup> (-62.336)
Barsat	0.85±0.08 <sup>a</sup>	0.62±0.03 <sup>ab</sup> (-27.472)	0.41 ± 0.08 <sup>a</sup> (-51.391)	0.84±0.04 <sup>b</sup>	0.55±0.01 <sup>a</sup> (-35.181)	0.36 ± 0.02 <sup>a</sup> (-57.465)

F.S. = Fakhr e Sarhad; In columns values are treatment means with ± SD; In a column, treatment means with different letters are statistically significant at 5% level of probability; Figures in parentheses indicate % promotion (+) and reduction (-) over control.

**Table 8** Physiological stress tolerance indices of different wheat varieties grown under different salt treatments

Wheat varieties/ Salinity levels	PHSI	RLSI	SFSI	RFSI	FSTI	SDSI	RDSI	DSTI
50 mM NaCl								
F.S.	89.7±6.3 <sup>a</sup>	74.4±7.1 <sup>a</sup>	65.7±2.6 <sup>a</sup>	54.4±3.6 <sup>a</sup>	59.1±3.4 <sup>a</sup>	47.8±4.5 <sup>a</sup>	60.2±16.2 <sup>a</sup>	51.1±7.9 <sup>ab</sup>
Insaf	70.9±7.2 <sup>a</sup>	72.5±0.9 <sup>a</sup>	99.8±4.5 <sup>a</sup>	95.1±2.4 <sup>a</sup>	96.5±0.3 <sup>a</sup>	105.3±6.7 <sup>c</sup>	93.6±8.5 <sup>a</sup>	100.2± 6.3 <sup>c</sup>
Lalma	98.2±11.6 <sup>a</sup>	86.9±6.9 <sup>a</sup>	87.3±7.3 <sup>a</sup>	98.7±13.9 <sup>a</sup>	92.5±3.7 <sup>a</sup>	94.8±7.7 <sup>bc</sup>	77.9±9.4 <sup>a</sup>	85.6±2.2 <sup>bc</sup>
Tatora	55.9±22.3 <sup>a</sup>	87.1±29.6 <sup>a</sup>	62.3±26.5 <sup>a</sup>	90.1±37.1 <sup>a</sup>	75.2±28.7 <sup>a</sup>	55.9±17.6 <sup>b</sup>	68.2±26 <sup>a</sup>	58.8±19.5 <sup>abc</sup>
Bathoor	88.2±19.3 <sup>a</sup>	76.4±11.6 <sup>a</sup>	81.8±17.2 <sup>a</sup>	72.2±12.1 <sup>a</sup>	75.9±13.5 <sup>a</sup>	107.7±5.7 <sup>c</sup>	80.4±11.9 <sup>a</sup>	95.1±8.8 <sup>c</sup>
Barsat	70.1±6.2 <sup>a</sup>	74.8±6.2 <sup>a</sup>	77.9±21.7 <sup>a</sup>	79.1±22.2 <sup>a</sup>	68.6±1.8 <sup>a</sup>	72.1±2.3 <sup>ab</sup>	36.2±3.1 <sup>a</sup>	37.4±2.4 <sup>a</sup>
150 mM NaCl								
F.S.	52.3±18.4 <sup>a</sup>	45.7±9.1 <sup>a</sup>	44.6 ± 16 <sup>ab</sup>	34.1 ± 14.5 <sup>a</sup>	37.9 ± 13.8 <sup>a</sup>	40.3 ± 13 <sup>a</sup>	49.6 ± 27.2 <sup>a</sup>	40.7 ± 1.7 <sup>a</sup>
Insaf	40.3±7.2 <sup>a</sup>	54.1±3.1 <sup>a</sup>	52.6 ± 9.3 <sup>b</sup>	66.1 ± 5.4 <sup>a</sup>	60.5 ± 1.3 <sup>a</sup>	68.6 ± 12 <sup>a</sup>	74.1 ± 14.8 <sup>a</sup>	70.8 ± 12.7 <sup>a</sup>
Lalma	43.6±7.3 <sup>a</sup>	67.6±5.1 <sup>a</sup>	49.1 ± 4.9 <sup>ab</sup>	72.5 ± 5.6 <sup>a</sup>	61.9 ± 4.5 <sup>a</sup>	60.9 ± 5.8 <sup>a</sup>	61.1 ± 8.7 <sup>a</sup>	60.3 ± 6.1 <sup>a</sup>
Tatora	23.1±9.7 <sup>a</sup>	55.7±16.1 <sup>a</sup>	16.1 ± 6.6 <sup>a</sup>	47.7 ± 13.1 <sup>a</sup>	31.9 ± 8 <sup>a</sup>	41.5 ± 17.3 <sup>a</sup>	38.3 ± 13.6 <sup>a</sup>	40.3 ± 1.2 <sup>a</sup>
Bathoor	51.4±6.4 <sup>a</sup>	56.4±9.9 <sup>a</sup>	59.5 ± 13.9 <sup>b</sup>	53.2 ± 12.1 <sup>a</sup>	55.6 ± 12.6 <sup>a</sup>	72.9 ± 7.6 <sup>a</sup>	71.1 ± 3.1 <sup>a</sup>	71.9 ± 4.7 <sup>a</sup>
Barsat	55.5±4.2 <sup>a</sup>	55.1±8.3 <sup>a</sup>	53.1 ± 7.9 <sup>b</sup>	74.2 ± 22.5 <sup>a</sup>	58.5 ± 5.4 <sup>a</sup>	55.1 ± 17.3 <sup>a</sup>	50.5 ± 4.6 <sup>a</sup>	40.3 ± 2.4 <sup>a</sup>

F.S. = Fakhr e Sarhad; In columns values are treatment means with ± SD; In a column, treatment means with different letters are statistically significant at 5% level of probability; PHSI = Plant Height Stress Tolerance Index; RLSI = Root Length Stress Tolerance Index; SFSI = Shoot Fresh Weight Stress Tolerance Index; RFSI = Root Fresh Weight Stress Tolerance Index; FSTI = Total Fresh Weight Stress Tolerance Index; SDSI = Shoot Dry Weight Stress Tolerance Index; RDSI = Root Dry Weight Stress Tolerance Index; DSTI = Total Dry Weight Stress Tolerance Index.

## Conclusion

From the current result, it was concluded that increasing NaCl concentration showed reduced germination percentage, shoot length, root length, fresh weight, dry weight, relative water content, vigor index, stem weight ratio, root weight ratio while root shoot ratio increased in *Triticum aestivum*. After comparison in different varieties, Lalma and Insaf seem more salt-tolerant than other cultivars at the germination and seedling establishment stage.

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