Evaluation of morphological traits of wheat varieties at germination stage under salinity stress

Fatemeh Gholizadeh¹, Ghader Mirzaghaderi¹, Subhan Danish²*, Mohammad Farsi³, Seyed Hasan Marashi³

1 Department of Plant Production and Genetics, Faculty of Agriculture, University of Kurdistan, Sanandaj, Iran, 2 Department of Soil Science, Faculty of Agricultural Sciences and Technology, Bahauddin Zakariya University Multan, Multan, Punjab, Pakistan, 3 Department of Crop Biotechnology and Breeding, Faculty of Agriculture, Ferdowsi University of Mashhad, Mashhad, Iran

* sd96850@gmail.com

Abstract
Salinity stress is one of the major plant growth-limiting factors in agriculture. It causes ionic imbalance, thus decrease the growth and yield attributes of crops especially wheat. Seedling stage is considered as one of the most sensitive stages under salinity stress. Survival of seeds at seedling stage can overcome the adverse impacts of salinity stress to some extent. Selection of salt tolerant varieties in seedling stage is considered as an effective strategy. Hence, current study was conducted to examine the seed germination responses of four wheat varieties under different levels of salinity. The wheat varieties such as ‘Rakhshan’, ‘Sirvan’, ‘Pishgam’ and ‘Heidari’ were grown and four salinity levels of 0, 4, 8 and 12 dS/m were applied under completely randomized design. The varieties such as ‘Sirvan’, ‘Rakhshan’ and ‘Heidari’ showed significant response for germination compared to ‘Pishgam’ at 12 dS/m salinity. Furthermore, the variety ‘Rakhshan’ showed significantly higher germination rate (20.3%), higher root length (33.4%) and higher shoot length (84.3%) than ‘Pishgam’, ‘Sirvan’ and ‘Sirvan’ respectively. However, contrasting results were obtained for dry weight of seedlings where 12.2% increase was observed in ‘Pishgam’ over ‘Rakhshan’ at 12 dS/m salinity that might be due to higher capability to uptake of Na and Cl ions. In conclusion, ‘Rakhshan’ wheat variety proved to be the most salinity tolerant as it grew better under saline soil conditions. More investigations at field level are recommended to declare ‘Rakhshan’ as salinity tolerant cultivar.

Introduction
Arable (6%) and irrigated arable land (20%) of world is becoming unproductive because of high salinity problem. Every year, salinity converts 2000 ha of arable land into barren uncultivable area [1]. Out of total land, 23.8 million hectares soils in Iran are salts affected [2]. These saline soils i.e., 18 million hectares are irrigated for crops cultivation where optimum achievement of wheat growth is extremely difficult. Saline soil is characterized as soils having excessive...
soluble salts in rhizosphere [3]. Continuous use of brackish irrigation predominantly increase
the chances of salinity development in soil [4]. In soil, increase in alkalinity, hydraulic conduc-
tivity, imbalance ionic concentration and specific ion toxicity development are some of major
drawbacks which is induced by salinity development [5].

Plants remain susceptible towards salinity from stage of germination to seed productions
[6, 7]. However, germination of seeds is key factor that played an imperative role in failure or
survival of crops establishment. It also obstructs water and nutrients uptake in crops [8]. Sur-
vival of seedlings at early stage of development under salt stress increases the chance of plant
population survival to reach up to maturity stage [9]. High salt concentration limits root devel-
opment by impairing physiological and metabolic balance and significantly reducing seed ger-
mination rate [10].

Accumulation of stress endogenous ethylene is another major indicator of salinity stress
[11, 12]. Several plant hormones i.e., brassinosteroids, auxins, cytokinins, gibberellins and
abscisic acid respond to salt stress [13, 14]. For such soils selection of salinity-tolerant cultivars
can be proved an effective and economical tool for plants to survive with salinity stress [15]. A
significant increase in the soil salinity decrease the potential of water with ultimately minimize
its optimum uptake into the plants [16]. Furthermore, specific ionic toxicity, over synthesis of
reactive oxygen species and osmotic stresses because of salinity also caused significant decrease
in plants growth indices [17].

Among different toxicity generated specific ions, sodium is most notorious one. It hampers
the growth of roots which adversely affect the development of seedlings after germination [18].
Furthermore, high Na uptake creates ionic imbalance in the plants by disturbing the potassium
(K) concentration. This imbalance of Na and K negatively affect the turgidity of stomata, thus
disturbed the gas exchange attributes in the plants [19–21].

Wheat (Triticum aestivum L.) is the main cereal crop and world’s second important grains
crop, with 730.9, 763.93 and 772.64 million metric tons harvest in 2018, 2019 and 2020 respective-
ly [22]. It is grown for high yield and due to its nutritional benefits, i.e., having carbohy-
drates, protein, minerals, fiber, B-group vitamins [23]. In humans nutrition, wheat share 55%
carbohydrates and 8–12% protein [24]. However, salinity stress adversely affect its growth and
productivity [25]. Therefore, the present investigation was carried out with aim to screen the
salt tolerant wheat variety at initial germination stage. Current study will help the farmers to
choose salt tolerant variety for the achievement of better productivity of crop. Limited litera-
ture work is documented so far on the selected varieties as salinity tolerant which is novelty
aspect of this study. It is hypothesized that selection of salt tolerant varieties can be helpful in
achieving the optimum wheat growth attributes under saline condition.

**Material and methods**

**Experimental site and design**

A laboratory experiment was conducted in the Faculty of Agriculture, Ferdowsi University of
Mashhad. The design of experiment was completely randomized design (CRD) with four
replications.

**Salinity development**

The salinity levels were (0, 4, 8 and 12 dS/m). Initially deionized water was taken, and sodium
chloride (NaCl) was added in it. During the addition of salt, magnetic shaker was shaking the
solution and electrical conductivity sensor was also placed in it. Pre-calibrated electrical con-
ductivity meter was used to monitor the salinity level [26]. After achievement of desired salin-
ity level, solution was stored in the clean water bottles for experiment purpose.
Seeds collection
Wheat varieties i.e., 'Rakhshan', 'Sirvan', 'Pishgam' and 'Heidari' were procured from Agricultural Research Center, Mashhad, Khorasan, Iran. Seeds were surface sterilized in 2% sodium hypochlorite solution for 5 minutes and washed with deionized water [27].

Experimental setup and germination conditions
Twenty sterilized seeds were placed in petri dishes with a diameter of 9 cm. Seeds were in sterilized filter paper and saline solution which was made by adding NaCl in sterilized water was applied in petri dishes as per treatment plan. Each treatment received 7 ml of solution. Same amount (7 ml) of sterilized water was added in control treatment petri dishes. The petri dishes were placed in a growth chamber at 25˚C temperature and 50% relative humidity under 16h light and 8h dark photoperiod arranged in a completely randomized design (CRD) with 4 replicates.

Harvesting and data collection
Seedlings were harvested after seven days of germination. The sprouted and germinated seeds were counted on daily basis. Data were collected at 7th days after germination of seeds. The degree of injury was greater in the highest salt concentration (12 dS/m). Seedling dry weight, germination percentage, root and shoot length were taken after seven days. After the final count, germination percentage (GP) [28] and germination rate (GR) was calculated by the following formulae

\[
GP = \left(\frac{NG}{NT}\right) \times 100
\]

\[
NG: \text{number of germinated seeds}; NT: \text{total number of seeds}
\]

\[
GR = \left(\frac{NG}{\text{Day of first count} + \text{Number of GerminatedSeeds} / \text{Day of Final Count}}\right) \times 100
\]

Tolerance indices
To assess the tolerance to salinity of each genotype, we adopted shoot length stress tolerance index (SLSI), root length stress tolerance index (RLSI), fresh weight stress tolerance index (FWSI), dry weight stress tolerance index (DWSI), germination stress tolerance index (GSI) [29].

\[
SLSI = \left(\frac{\text{Shoot length under salinity stress}}{\text{Shoot length under no salinity stress}}\right) \times 100
\]

\[
RLSI = \left(\frac{\text{Root length under salinity stress}}{\text{Root length under no salinity stress}}\right) \times 100
\]

\[
FWSI = \left(\frac{\text{Fresh weight under salinity stress}}{\text{Fresh weight under no salinity stress}}\right) \times 100
\]

\[
DWSI = \left(\frac{\text{Dry weight under salinity stress}}{\text{Dry weight under no salinity stress}}\right) \times 100
\]
\[
\text{GSI} = \left( \frac{\text{Seeds germinated under salinity stress}}{\text{Seeds germinated under no salinity stress}} \right) \times 100
\]

**Statistical analyses**

Data analysis of variance was executed using two-way Analysis of variance (ANOVA) [30]. The mean of treatments was compared using LSD’s test at 5% probability level. Correlation analyses and principal component analysis were assessed to determine the relationships between the traits using origin version 2021 software [31].

**Results**

**Germination**

Results showed that ‘Rakhshan’, ‘Pishgam’ and ‘Heidari’ remained statistically alike to each other but differed significantly over ‘Sirvan’ at 0 dS/m salinity for germination. No significant change was noted in all the varieties at 4 and 8 dS/m for germination. Varieties ‘Rakhshan’, ‘Heidari’ and ‘Sirvan’ showed significantly higher germination at highest level of salinity than ‘Pishgam’ (Fig 1A and 1B). Maximum increase of 15.6% in germination was observed in ‘Rakhshan’ and ‘Sirvan’ than ‘Pishgam’ at highest salinity level, i.e., 12 dS/m. ‘Pishgam’ was most susceptible variety against salinity (12 dS/m) while ‘Rakhshan’ was most susceptible at 4 and 8 dS/m for germination (Table 1). ‘Sirvan’ was resistant variety for germination at all levels of salinity i.e., 4, 8 and 12 dS/m.

**Rate of germination**

For rate of germination, ‘Rakhshan’ showed significantly better results over ‘Sirvan’, ‘Pishgam’ and ‘Heidari’ at 0 dS/m. No significant difference was noted in rate of germination of ‘Sirvan’, ‘Pishgam’ and ‘Heidari’ at 0 dS/m. ‘Sirvan’ and ‘Heidari’ remained statistically alike but remained significantly better over ‘Rakhshan’ and ‘Pishgam’ for rate of germination at 4 dS/m

![Fig 1. Effects of salinity on germination percentage in wheat varieties.](https://doi.org/10.1371/journal.pone.0258703.g001)
Salinity stress adversely affects wheat seeds germination. Table 1 presents the percentage change in germination of different wheat varieties as influenced by variable salinity levels.

| Wheat Varieties | Germination (%) | Change (%) over Control (0 dS/m) |
|-----------------|-----------------|----------------------------------|
|                 | 4 (dS/m) | 8 (dS/m) | 12 (dS/m) |
| Sirvan          | 5.41     | -2.70    | 0.00      |
| Rakhshan        | -5.00    | -7.50    | -7.50     |
| Heidari         | -2.56    | -5.13    | -7.69     |
| Pishgam         | -1.28    | -5.13    | -17.95    |

Negative sign indicates the decrease due to salinity.

https://doi.org/10.1371/journal.pone.0258703.t001

Salinity (Fig 2A and 2B). 'Rakhshan' also showed significantly higher rate of germination over 'Pishgam' at 4 dS/m. 'Sirvan' and 'Heidari' were non-significant with each other but only 'Heidari' remained significantly better over 'Rakhshan' and 'Pishgam' for rate of germination at 8 dS/m salinity. 'Sirvan' also gave significantly higher rate of germination over 'Pishgam' at 8 dS/m. However, at highest salinity levels (12 dS/m), 'Rakhshan', and 'Sirvan' showed significantly better rate of germination than 'Pishgam'. Maximum increase of 20.3% was observed 'Rakhshan' over 'Pishgam' at 12 dS/m salinity for rate of germination. 'Pishgam' was most susceptible variety against salinity (12 dS/m) while 'Rakhshan' was most susceptible at 4 and 8 dS/m for rate of germination (Table 2). 'Heidari' was resistant variety for rate of germination at 4 and 8 dS/m while 'Sirvan' was more resistant at 12 dS/m.

Root length

In case of root length, 'Sirvan' and 'Heidari' differed significantly over 'Rakhshan' and 'Pishgam' at 0 dS/m. No significant difference was noted in root length of 'Rakhshan', 'Sirvan', 'Pishgam' and 'Heidari' at 4 dS/m. Pishgam remained significantly better over 'Rakhshan', 'Sirvan' and 'Heidari' for root length at 8 dS/m salinity (Fig 3A and 3B). 'Rakhshan' also showed significantly higher root length over 'Heidari' at 8 dS/m. 'Sirvan' and 'Heidari' were non-
Table 2. Percentage change in rate of germination of different wheat varieties as influenced by variable salinity levels.

| Wheat Varieties | Rate of germination change (%) over Control (0 dS/m) |
|-----------------|---------------------------------------------------|
|                 | 4 (dS/m) | 8 (dS/m) | 12 (dS/m) |
| Sirvan          | 5.17     | -15.89   | -35.11    |
| Rakhshan        | -18.87   | -32.97   | -41.75    |
| Heidari         | 13.35    | 2.02     | -37.68    |
| Pishgam         | -16.99   | -20.42   | -42.77    |

Negative sign indicates the decrease due to salinity.

significant with each other for root length at 8 dS/m salinity. However, ‘Rakhshan’ gave significantly higher root length over ‘Sirvan’, ‘Heidari’ at 12 dS/m. Maximum increase of 33.4% was observed ‘Rakhshan’ over ‘Sirvan’ at 12 dS/m salinity for root length. ‘Sirvan’ was most susceptible variety against salinity (4 and 12 dS/m) while ‘Heidari’ and was most susceptible at 8 dS/m for root length (Table 3). ‘Pishgam’ was resistant variety for root length at 4 and 8 dS/m while ‘Rakhshan’ was more resistant at 12 dS/m.

Shoot length

For shoot length, ‘Rakhshan’ showed significantly higher results over ‘Sirvan’, ‘Pishgam’ and ‘Heidari’ at 0 dS/m. No significant difference was noted in shoot length ‘Sirvan’, ‘Pishgam’, ‘Rakhshan’ and ‘Heidari’ at 4 dS/m. ‘Sirvan’ was significantly better for shoot length over ‘Rakhshan’, ‘Pishgam’ and ‘Heidari’ at 8 dS/m. Similarly, ‘Rakhshan’ also gave significantly higher shoot length over ‘Pishgam’ at 8 dS/m. However, at 12 dS/m, ‘Rakhshan’ and ‘Pishgam’ showed significantly better shoot length than ‘Sirvan’ and ‘Heidari’ (Fig 4A and 4B). Maximum increase of 84.3% was observed ‘Rakhshan’ over ‘Sirvan’ at 12 dS/m salinity for shoot length. ‘Rakhshan’ was most susceptible variety against salinity (4 and 8 dS/m) while ‘Sirvan’ and was most susceptible at 8 dS/m for root length (Table 4). ‘Heidari’ was resistant variety for shoot length at 4, ‘Sirvan’ at 8 dS/m while ‘Pishgam’ was more resistant at 12 dS/m.

Fig 3. Effects of salinity on root length in wheat varieties. A) Means are average of four replicates. Different values showed significant difference at p < 0.05 compared by LSD. B) Different values at bars are p values obtained during LSD comparison of wheat varieties and salinity levels.
Table 3. Percentage change in root length of different wheat varieties as influenced by variable salinity levels.

| Wheat Varieties | Root Length (cm) | Change (%) over Control (0 dS/m) |
|-----------------|------------------|----------------------------------|
|                 | 4 (dS/m)         | 8 (dS/m)                         | 12 (dS/m)                         |
| Sirvan          | -18.28           | -43.28                           | -65.33                            |
| Rakhsan         | -9.10            | -33.20                           | -49.69                            |
| Heidari         | -14.75           | -48.02                           | -61.76                            |
| Pishgam         | -7.38            | -24.53                           | -52.08                            |

Negative sign indicates the decrease due to salinity.

https://doi.org/10.1371/journal.pone.0258703.t003

Dry weight

Results showed that dry weight was significantly higher in 'Heidari' over 'Sirvan', 'Pishgam' and 'Rakhsan' at 0 dS/m. 'Sirvan' also showed significant increase in dry weight than and 'Pishgam' at 0 dS/m. No significant difference was noted in dry weight of 'Heidari' and 'Pishgam' but significant over 'Rakhsan' and 'Sirvan' at 4 dS/m. 'Pishgam' was significantly better for dry weight over 'Rakhsan', 'Sirvan' and 'Heidari' at 8 dS/m. Similarly, 'Heidari' also gave significantly higher dry weight over 'Rakhsan' at 8 dS/m (Fig 5A and 5B). It was noted that at 12 dS/m, 'Pishgam' showed significantly better dry weight than 'Rakhsan', 'Sirvan' and 'Heidari'. Maximum increase of 12.2% was observed 'Pishgam' over 'Rakhsan' at 12 dS/m salinity for dry weight. 'Heidari' was most susceptible variety against salinity (8 and 12 dS/m) while 'Sirvan' and was most susceptible at 4 dS/m for root length (Table 5). 'Pishgam' was resistant variety for dry weight at 4, 8 and 12 dS/m.

Stress tolerance index

Results showed that RLSI was significantly high in 'Rakhsan' and 'Pishgam' over 'Sirvan' at 4, 8 and 12 dS/m. 'Sirvan' and 'Heidari' showed significantly better SLSI over 'Rakhsan' at 4 dS/
m. 'Sirvan' SLSI was significantly higher at 8 dS/m than Rakhshan', 'Heidari' and 'Pishgam'. However, at 8 dS/m 'Pishgam' showed significantly higher SLSI over Rakhshan', 'Heidari' and 'Sirvan'. 'Rakhshan' and 'Pishgam' were statistically alike but differed significantly over 'Sirvan' and 'Heidari' for DWSI at 4 dS/m. 'Pishgam' DWSI remained significantly better over Sirvan'

| Wheat Varieties | Shoot Length (cm) |
|-----------------|-------------------|
|                 | Change (%) over Control (0 dS/m) |
|                 | 4 (dS/m) | 8 (dS/m) | 12 (dS/m) |
| Sirvan          | -6.25    | -12.28   | -68.89    |
| Rakhshan        | -16.03   | -30.06   | -49.10    |
| Heidari         | -2.31    | -21.53   | -53.63    |
| Pishgam         | -9.64    | -27.09   | -40.74    |

Negative sign indicates the decrease due to salinity.

https://doi.org/10.1371/journal.pone.0258703.t004

Table 4. Percentage change in shoot length of different wheat varieties as influenced by variable salinity levels.

Table 5. Percentage change in dry weight of different wheat varieties as influenced by variable salinity levels.

| Wheat Varieties | Dry Weight (g) |
|-----------------|---------------|
|                 | Change (%) over Control (0 dS/m) |
|                 | 4 (dS/m) | 8 (dS/m) | 12 (dS/m) |
| Sirvan          | -25.95   | -36.76   | -43.78    |
| Rakhshan        | -7.55    | -28.93   | -38.36    |
| Heidari         | -22.64   | -41.98   | -50.94    |
| Pishgam         | -6.25    | -23.86   | -37.50    |

Negative sign indicates the decrease due to salinity.

https://doi.org/10.1371/journal.pone.0258703.t005

Fig 5. Effects of salinity on dry weight in wheat varieties. A) Means are average of four replicates. Different values showed significant difference at p < 0.05 compared by LSD. B) Different values at bars are p values obtained during LSD comparison of wheat varieties and salinity levels.

https://doi.org/10.1371/journal.pone.0258703.g005

m. 'Sirvan' SLSI was significantly higher at 8 dS/m than Rakhshan', 'Heidari' and 'Pishgam'. However, at 8 dS/m 'Pishgam' showed significantly higher SLSI over Rakhshan', 'Heidari' and 'Sirvan'. 'Rakhshan' and 'Pishgam' were statistically alike but differed significantly over 'Sirvan' and 'Heidari' for DWSI at 4 dS/m. 'Pishgam' DWSI remained significantly better over Sirvan'
Table 6. Stress tolerance index of different wheat varieties under variable salinity levels.

| Wheat Varieties | RLSI 4 (dS/m) | RLSI 8 (dS/m) | RLSI 12 (dS/m) | SLSI 4 (dS/m) | SLSI 8 (dS/m) | SLSI 12 (dS/m) |
|-----------------|--------------|--------------|--------------|--------------|--------------|--------------|
| Sirvan          | 81.74b       | 56.66f       | 34.68i       | 93.12a       | 87.75ab      | 31.19g       |
| Rakhshan        | 90.92a       | 66.84e       | 50.24gh      | 84.01bc      | 70.21d       | 50.55f       |
| Heidari         | 85.34bc      | 52.03fg      | 38.17i       | 95.35a       | 77.41cd      | 46.17f       |
| Pishgam         | 86.94ab      | 73.29d       | 46.54h       | 90.73ab      | 72.96d       | 59.78e       |

| Wheat Varieties | DWSI          | FWSI          |
|-----------------|---------------|---------------|
| Sirvan          | 74.09bc       | 63.25d        |
| Rakhshan        | 92.47a        | 71.13c        |
| Heidari         | 77.40b        | 58.07e        |
| Pishgam         | 93.80a        | 76.19b        |

Means are average of 3 replicates. Different letters showed significant difference at p<0.05.

https://doi.org/10.1371/journal.pone.0258703.t006

and ‘Heidari’ at 8 and 12 dS/m. ‘Rakhshan’ also different significantly better over Sirvan’ and ‘Heidari’ at 8 and 12 dS/m. No significant change was noted in FWSI among all the varieties at 4 dS/m. Rakhshan’, ‘Heidari’ and ‘Sirvan’ were significantly higher in FWSI at 8 and 12 dS/m (Table 6).

Fig 6. Person correlation between germination characteristics in wheat varieties.

https://doi.org/10.1371/journal.pone.0258703.g006
Pearson correlation and principal component analysis

Pearson correlation showed that all the growth attributes showed significant positive correlation with each other. Increase in germination, shoot and root length also significantly positively influenced fresh and dry weight of seedlings (Fig 6). Principle component analysis verified that rate of germination and fresh weight of wheat seedlings are closely associated with each other. However, dry weight was more closely dependent on the shoot and root length of wheat seedlings (Figs 7 and 8).

Discussion

In the metabolism of any plant, cell wall components are prime in importance. These metabolic process i.e., nucleic, energy production, respiration and protein metabolism played an imperative role in the enlargement of cell and occupy a significant portion of cell biomass [32–34]. A significant in balance in the chlorophyll content index due to accumulation of sodium ions and potassium ions in the leaf blade is major cause for chlorophyll destruction. Poor chlorophyll content decreases the photosynthesis which resulted in growth of seedlings. it also adversely affects the required photosynthate that are necessity for the survival of any plant under stress condition [35–37]. Increasing stress induced by salinity, decrease the osmotic potential of soil solution which caused a significant decline in its availability to the plants [38–42].

Imbalance in ABA and GA hormones also disturbed the germination of seeds [43]. Higher concentration of ABA under salinity stress induced the dormancy period on the seeds resulted
in poor seeds germination [44–47]. Our findings are also in agreement with above mentioned arguments. Increasing level of salinity significantly decreased the germination and germination rate of different wheat varieties over control. It was also noted that higher salinity level caused a significant decline in shoot and root length of different wheat varieties used in current study.

Poor growth of roots disturbs the optimum uptake of nutrients and water in seedling under salinity stress. When plants suffer from less nutrients and limited water availability, division of cells for the enlargement of shoot also become disturbed. Such conditions resulted in poor development of shoot length [48]. In addition, osmotic stress generated by higher concentration of salts also decreased the rate of seeds germination. Low fresh weight of wheat seedlings at higher level of salinity also validates this fact that less water uptake under salinity stress is dominant adverse effect of higher salts concentration in rhizosphere. Higher osmotic stress stimulated the biosynthesis of reactive oxygen species and ionic toxicity which caused extension in germination time of seeds [49]. Irrigation water uptake is also decreased during imbibition process due to increase in the osmotic potential by higher concentration of salts [50].

Under saline conditions, ionic toxicity i.e., Na⁺ and Cl⁻ on embryo viability also played a key role in poor germination of seeds [51, 52]. Toxic effects of Na⁺ and Cl⁻ resulted in disruption of enzymes and macromolecules, cell organelles and plasma membrane damaged, disturbance in respiration, protein synthesis and photosynthesis [52–54]. It has also been observed that higher concentration of Na and Cl in soil solution disturbs the ionic balance. Competition between essential nutrients and Na restrict the required uptake of macro and micronutrients in the plants which eventually decreased the dry weight of seedlings [55].
In current study, poor dry weight at highest level of salinity might be due to less uptake of nutrients. Impaired shoot and root growth under salinity induced osmotic stress might be due to less cell division and elongation leading to decrease in dry weight of root and shoot [56, 57].

Conclusion
It is concluded that increasing level of salinity adversely affect the germination and seedlings growth attributes in Sirvan', 'Pishgam' and 'Heidari'. Both person correlation and PCA also validated the negative correlation with increasing level of salinity with growth attributes. Based on results 'Rakhshan’ has potential to survive under critical salinity stress. It showed relatively better growth i.e., root and shoot length, fresh and dry weight compared to other varieties in the study. However, deep scientific attention and more experiments at field level are required to declare 'Rakhshan’ as salt tolerant wheat variety.

Author Contributions
Conceptualization: Fatemeh Gholizadeh, Ghader Mirzaghaderi.
Data curation: Fatemeh Gholizadeh, Ghader Mirzaghaderi.
Formal analysis: Mohammad Farsi, Seyed Hasan Marashi.
Investigation: Mohammad Farsi, Seyed Hasan Marashi.
Methodology: Fatemeh Gholizadeh, Ghader Mirzaghaderi.
Resources: Fatemeh Gholizadeh, Ghader Mirzaghaderi.
Software: Fatemeh Gholizadeh, Ghader Mirzaghaderi.
Validation: Mohammad Farsi, Seyed Hasan Marashi.
Visualization: Fatemeh Gholizadeh, Ghader Mirzaghaderi, Subhan Danish.
Writing – original draft: Fatemeh Gholizadeh, Ghader Mirzaghaderi, Subhan Danish.
Writing – review & editing: Fatemeh Gholizadeh, Ghader Mirzaghaderi, Subhan Danish.

References
1. Tammam AA, Alhamd MFA, Hemeda MMJ AJ O. CS. Study of salt tolerance in wheat (Triticum aestium L.) cultivar Banysoif 1. A eut J Crop Sci. 2008; 1: 115–125.
2. Khodapanah L, Sulaiman WNA, Khodapanah N. Groundwater quality assessment for different purposes in Estehard district, Tehran, Iran. Eur J Sci Res. 2009; 36: 543–553.
3. Al-Abdoulr Hadi IA, Dinar HA, Ebert G, Büttner C. Effect of salinity on leaf growth, leaf injury and biomass production in date palm (Phoenix dactylifera L.) cultivars. Indian J Sci Technol. 2011; 4: 1542–1546. https://doi.org/10.17485/ijst/2011/v4i11/30283
4. Mishra BK, Meena KK, Dubey PN, Ashwath OP, Kant K, Sorty AM, et al. Influence on yield and quality of fennel (Foeniculum vulgare Mill.) grown under semi-arid saline soil, due to application of native phosphatase solubilizing rhizobacterial isolates. Ecol Eng. 2016; 97: 327–333. https://doi.org/10.1016/j.ecoleng.2016.10.034
5. Machado R, Serralheiro R. Soil Salinity: Effect on Vegetable Crop Growth. Management Practices to Prevent and Mitigate Soil Salinization. Horticulturae. 2017; 3: 30. https://doi.org/10.3390/horticulturae3020030
6. Jeevan Kumar SP, Rajendra Prasad S, Banerjee R, Thammineni C. Seed birth to death: Dual functions of reactive oxygen species in seed physiology. Ann Bot. 2015; 116: 663–668. https://doi.org/10.1093/aob/mcv098 PMID: 26271119
7. Sorty AM, Meena KK, Choudhary K, Bitla UM, Minhas PS, Krishnani KK. Effect of Plant Growth Promoting Bacteria Associated with Halophytic Weed (Psoralea corylifolia L) on Germination and Seedling
Growth of Wheat Under Saline Conditions. Appl Biochem Biotechnol. 2016; 180: 872–882. https://doi.org/10.1007/s12010-016-2139-z PMID: 27215915

8. Gong DH, Wang GZ, Si WT, Zhou Y, Liu Z, Jia J. Effects of Salt Stress on Photosynthetic Pigments and Activity of Ribulose-1,5-bisphosphate Carboxylase/Oxygenase in Kalidium foliatum. Russ J Plant Physiol. 2018; 65: 98–103. https://doi.org/10.1134/S1021443718010144

9. Kader MA, Jutzi SC. Effects of Thermal and Salt Treatments during Imbibition on Germination and Seedling Growth of Sorghum at 42/19°C. J Agron Crop Sci. 2004; 190: 35–38. https://doi.org/10.1046/j.0931-2250.2003.00071.x

10. Munns R, James RA, Làuchli A. Approaches to increasing the salt tolerance of wheat and other cereals. J Experimental Botany. 2006; 57(5): 1025–43. https://doi.org/10.1093/jxb/erj100 PMID: 16510517

11. Wang CY, Adams DO. Chilling-Induced Ethylene Production in Cucumbers (Cucumis sativus L.). Plant Physiol. 1982; 69: 424–427. https://doi.org/10.1104/pp.69.2.424 PMID: 16662222

12. El-Beltagy AS, Khalifa MM, Hall MA. Salinity in relation to ethylene. Egypt J Hort. 1979; 6: 269–271.

13. Ryu H, Cho YG. Plant hormones in salt stress tolerance. Journal of Plant Biology. 2015; 58(3):147–155. https://doi.org/10.1007/s12374-015-0103-z

14. Aloni R, Aloni E, Langhans M, Ullrich CI. Role of cytokinin and auxin in shaping root architecture: Regulating vascular differentiation, lateral root initiation, root apical dominance and root gravitropism. Annals of Botany. 2006; 97(5): 883–893. https://doi.org/10.1093/aob/mol027 PMID: 1647866

15. Arzani A. Improving salinity tolerance in crop plants: A biotechnological view. In Vitro Cellular and Developmental Biology—Plant. 2008; 44:373–383. https://doi.org/10.1007/s11627-008-9157-7

16. Tester M, Davenport R. Na+ tolerance and Na+ transport in higher plants. Ann Bot. 2003; 91: 503–527. https://doi.org/10.1038/aobmc058 PMID: 12646496

17. Marschner H. Mineral Nutrition of Higher Plants. 2nd ed. Mineral Nutrition of Higher Plants. San Diego, USA: Academic Press; 1995. https://doi.org/10.1016/B978-012473542-2/50017-1

18. Jana GA, Al Kharusi L, Sunkar R, Al-Yahyai R, Yaish MW. Metabolic analysis of date palm seedlings exposed to salinity and silicon treatments. Plant Signal Behav. 2019; 14: 1663112. https://doi.org/10.1080/15592424.2019.1663112 PMID: 31505979

19. Ahmad P, Abass Ahanger M, Nasser Alyemeni M, Wijaya L, Alam P, Ashraf M. Mitigation of sodium chloride toxicity in Solanum lycopersicum L. by supplantation of jasmonic acid and nitric oxide. J Plant Interact. 2018; 13: 64–72.

20. Maser P, Mäser P, Gierth M, Schroeder JI. Molecular mechanisms of potassium and sodium uptake in plants. Plant Soil. 2002; 247: 43–54. https://doi.org/10.1023/A:1021159130729

21. Zhang H, Irving LJ, McGill C, Matthew C, Zhou D, Kemp P. The effects of salinity and osmotic stress on barley germination rate: Sodium as an osmotic regulator. Ann Bot. 2010; 106(6): 1027–1035. https://doi.org/10.1093/aob/mcq204 PMID: 20929898

22. Shahbandeh M. Global wheat production from 2011/2012 to 2020/2021 (in million metric tons). Germany; 2021. Available: https://www.statista.com/statistics/267268/production-of-wheat-worldwide-since-1990/

23. Mahajan S, Tuteja N. Cold, salinity and drought stresses: An overview. Archives of Biochemistry and Biophysics. 2005; 444(2):139–158. https://doi.org/10.1016/j.abb.2005.02.008 PMID: 16309626

24. Bos C, Julius B, Fouillet L, Turlan L, Darè S, Luengo C, et al. Postprandial metabolic utilization of wheat protein in humans. Am J Clin Nutr. 2005; 81: 87–94. https://doi.org/10.1093/ajcn/81.1.87 PMID: 15640465

25. Tmrk M, Rötter RP, Ruíz-Ramos M, Kersebaum KC, Olsen JE, Žalud Z, et al. Adverse weather conditions for European wheat production will become more frequent with climate change. Nat Clim Chang. 2014; 4: 637–643. https://doi.org/10.1038/nclimate2242

26. Rhoades JD. Salinity: Electrical Conductivity and Total Dissolved Solids. In: Sparks D.L., Page A.L., Helmke P.A., Loepert R.H., Soltanpour P. N., Tabatabaí M. A., et al., editors. Methods of Soil Analysis, Part 3, Chemical Methods. Madison, WI, USA: Soil Science Society of America; 1996. pp. 417–435. https://doi.org/10.2136/ssasobokser5.3.c14

27. Ahmad I, Akhter MJ, Zahir ZA, Naveed M, Mitter B, Sessitsch A. Cadmium-tolerant bacteria induce metal stress tolerance in cereals. Environ Sci Pollut Res. 2014; 21: 11054–11065. https://doi.org/10.1007/s11356-014-3010-9 PMID: 24849374

28. Shehzad M, Ayub M, Ahmad AUH, Yaseen M. Influence of Priming Techniques on Emergence and Seedling Growth of Forage Sorghum (Sorghum bicolor L.). J Anim Plant Sci. 2012; 22: 154–158.

29. Ashraf MY, Hussain F, Akhter J, Gul A, Ross M, Ebert G. Effect of different sources and rates of nitrogen and supra optimal level of potassium fertilization on growth, yield and nutrient uptake by sugarcane grown under saline conditions. Pakistan J Bot. 2008; 40: 1521–1531.
30. Steel RG, Torrie JH, Dickey DA. Principles and Procedures of Statistics: A Biometrical Approach. 3rd ed. Singapore: McGraw Hill Book International Co.; 1997.

31. OriginLab Corporation. OriginPro. Northampton, MA, USA.: OriginLab; 2021. Available: https://store.originlab.com/store/Default.aspx?CategoryID = 59&ItemID = EF-096N0P-ESTU

32. Zhong H, Läuchli A. Incorporation of [14C] glucose into cell wall polysaccharides of cotton roots: effects of NaCl and CaCl2. Plant Physiol. 1988; 88: 511–514. https://doi.org/10.1104/pp.88.3.511 PMID: 16666336

33. Qin S, Zhang YJ, Yuan B, Xu PY, Xing K, Wang J, et al. Isolation of ACC deaminase-producing habitat-adapted symbiotic bacteria associated with halophyte Limonium sinense (Girard) Kuntze and evaluating their plant growth-promoting activity under salt stress. Plant Soil. 2014; 374: 753–766. https://doi.org/10.1007/s11104-013-1918-3

34. Ashraf M, Harris PJ. Potential biochemical indicators of salinity tolerance in plants. Plant Science. 2004; 166(1):3–16. https://doi.org/10.1016/j.plantsci.2003.10.024

35. Saddiq MS, Iqbal S, Hafeez MB, Ibrahim AMH, Raza A, Fatima EM, et al. Effect of Salinity Stress on Physiological Changes in Winter and Spring Wheat. Agronomy. 2021; 11: 1193.

36. Mostofa MG, Rahman MM, Ansary MMU, Keya SS, Abdelrahman M, Miah MG, et al. Silicon in mitigation of abiotic stress-induced oxidative damage in plants. Crit Rev Biotechnol. 2021; 41(6):918–934. https://doi.org/10.1080/07388551.2021.1892582 PMID: 33784900

37. Mansour E, Moustafa ESA, Desoky E-SM, Ali M, Yasin MAT, Attia A, et al. Multidimensional evaluation for detecting salt tolerance of bread wheat genotypes under actual saline field growing conditions. Plants. 2020; 9: 1324. https://doi.org/10.3390/plants9101324 PMID: 33036311

38. Mwando E, Han Y, Angessa TT, Zhou G, Hill CB, Zhang XQ, et al. Genome-Wide Association Study of Salinity Tolerance During Germination in Barley (Hordeum vulgare L.). Front Plant Sci. 2020; 11: 118. https://doi.org/10.3389/fpls.2020.00118 PMID: 32153619

39. Fiaz K, Danish S, Younis U, Malik SA, Raza Shah MH, Niaz S. Drought impact on Pb/Cd toxicity remediated by biochar in Brassica campestris. J Soil Sci Plant Nutr. 2014; 14: 845–854. https://doi.org/10.4067/S0718-95162014005000067

40. Danish S, Zafar-ul-Hye M, Hussain S, Riaz M, Qayyum MF. Mitigation of drought stress in maize through inoculation with drought tolerant ACC deaminase containing PGPR under axenic conditions. Pakistan J Bot. 2020; 52: 49–60. https://doi.org/10.30848/PJB2020.117

41. Danish S, Zafar-ul-Hye M, Mohsin F, Hussain M. ACC-deaminase producing plant growth promoting rhizobacteria and biochar mitigate adverse effects of drought stress on maize growth. PLoS One. 2020; 15: e0230615. Available: https://doi.org/10.1371/journal.pone.0230615 PMID: 32251430

42. Danish S, Zafar-ul-Hye M. Combined role of ACC deaminase producing bacteria and biochar on cereals productivity under drought. Phyton (B Aires). 2020; 89: 217–227. https://doi.org/10.32604/phyton.2020.08523

43. Han C, Yang P. Studies on the molecular mechanisms of seed germination. Proteomics. 2015; 15(10): 1671–1679. https://doi.org/10.1002/pmic.201400375 PMID: 25597791

44. Finkelstein R, Reeves W, Ariizumi T, Steber C. Molecular aspects of seed dormancy. Annu Rev Plant Biol. 2008; 59: 387–415. https://doi.org/10.1146/annurev.arplant.59.032607.092740 PMID: 18257711

45. Miransari M, Smith DL. Plant hormones and seed germination. Environ Exp Bot. 2014; 99: 110–121. https://doi.org/10.1016/j.enveex.2013.11.005

46. Iqbal M, Ashraf M. Gibberellic acid mediated induction of salt tolerance in wheat plants: Growth, ionic partitioning, photosynthesis, yield and hormonal homeostasis. Environ Exp Bot. 2013; 86: 76–85. https://doi.org/10.1016/j.enveex.2010.06.002

47. Yoshida T, Mogami J, Yamaguchi-Shinozaki K. ABA-dependent and ABA-independent signaling in response to osmotic stress in plants. Current Opinion in Plant Biology. 2014, pp. 133–139. https://doi.org/10.1016/j.pbi.2014.07.009 PMID: 25104049

48. Rahman MS, Miyake H, Takeoka Y. Effect of Sodium Chloride Salinity on Seed Germination and Early Seedling Growth of Rice (Oryza sativa L.). Pakistan J Biol Sci. 2001; 4: 351–355. https://doi.org/10.3923/pjbs.2001.351.355

49. Munns R. Comparative physiology of salt and water stress. Plant, Cell Environ. 2002; 25: 239–250. https://doi.org/10.1046/j.1365-3040.2001.00808.x PMID: 11841667

50. Munns R, Tester M. Mechanisms of Salinity Tolerance. Annu Rev Plant Biol. 2008; 59: 651–681. https://doi.org/10.1146/annurev.arplant.59.032607.092911 PMID: 18444910

51. Jahromi F, Aroca R, Porcel R, Ruiz-Lozano JM. Influence of salinity on the in vitro development of G. moso intraradices and on the in vivo physiological and molecular responses of mycorrhizal lettuce plants. Microb Ecol. 2008; 55: 45–53. https://doi.org/10.1007/s00248-007-9249-7 PMID: 17393053
52. Daszkowska-Golec A. Arabidopsis seed germination under abiotic stress as a concert of action of phytohormones. OMICS A Journal of Integrative Biology. 2011. pp. 763–774. https://doi.org/10.1089/omi.2011.0082 PMID: 22011341

53. Parida AK, Das AB. Salt tolerance and salinity effects on plants: A review. Ecotoxicol Environ Saf. 2005; 60: 324–349. https://doi.org/10.1016/j.ecoenv.2004.06.010 PMID: 15590011

54. Panda SK, Khan MH. Growth, oxidative damage and antioxidant responses in green gram (Vigna radiata L.) under short-term salinity stress and its recovery. J Agron Crop Sci. 2009; 195: 442–454. https://doi.org/10.1111/j.1439-037X.2009.00371.x

55. Abari AK, Nasr MH, Hojjati M, Bayat D. Salt effects on seed germination and seedling emergence of two Acacia species. African J Plant Sci. 2011; 5: 52–56.

56. Zeiger E, Taiz L. Plant Physiology. Sunderland, MA, USA: Sinauer Associates Inc., Publishers; 2010.

57. Paul S, Aggarwal C, Manjunatha B., Rathi MS. Characterization of osmotolerant rhizobacteria for plant growth promoting activities in vitro and during plant-microbe association under osmotic stress. Indian J Exp Biol. 2018; 56: 582–589.