

Genetic Analysis of Salinity Tolerance in Wheat (*Triticum aestivum* L.)

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Abstract

Background: The understanding of the genetics of salt tolerance is of utmost need to combat the rising prevalence of soil salinity through employing tolerant cultivars. The current study was carried out to investigate the quantitative genetic basis of agronomical and physiological-related traits of salinity-stressed plants using the seven generations (parents, F₁, F₂, F₃, and backcrosses) of wheat grown in the field under normal and saline conditions.

Results: The combined analysis of variance showed highly significant effects of salinity and genotypes (generations) on all the traits. The scaling tests did not support the three-parameter model (additive-dominance model); hence, the six-parameter model was used to assess the genetic effects governing the traits in this study. The epistatic gene effects were crucial, as were additive and dominance gene effects for plant height, K/Na, and yield in salinity stress conditions. The highest heritability was observed for total chlorophyll, carotenoid, SPAD chlorophyll, and K/Na ratio in saline conditions. The additive genetic variance was more important than the dominance variance for grain weight, K, K/Na in salinity conditions.

Conclusions: The findings of the current study may have important implications in the quantitative genetics of salinity tolerance and the development of cultivars tolerant to salinity in wheat.

Background

Common wheat (*Triticum aestivum* L.) is a crucial staple food with global production of over 700 million tones, contributes significantly to the diet of the world population by supplying 20% of protein and daily calories to 4.5 billion people worldwide [1]. Saline soils are one of the abiotic factors that have adversely affected plant production, especially in the arid and semi-arid regions of the world [2]. Iran is a region where the far-most western side of the Fertile Crescent, as the center of origin of wheat (*Triticum* spp.) [1], is one of the countries with an estimated 18 to 27 million hectares of saline lands has been affected by salinity to various degrees.

The adverse effects of salinity on plant growth and development can be implemented in two phases according to the classical view [2; 3]. In the first phase, osmotic stress happens immediately after exposure to salinity stress and inhibits plant growth. At the second phase, ionic stress (toxicity) happens when plant is exposed to salt for several days or weeks depending on the severity of salinity stress and when toxic ions (e.g. Na⁺ and Cl⁻) accumulate to high concentrations beyond the plant-specific thresholds in the leaves. Plants that tolerate osmotic stress can be maintained their growth rate in the initial stage of salinity exposure [4]. Therefore, in addition to osmoregulation/osmotic adjustment, enhancing osmotic stress tolerance may take place by two opposing strategies. The selection of plants with low leaf area represents the first approach to improve stomata performance. Conversely, selecting of plants with more leaf area and capacity to intercept light is the second strategy to improve the required energy for water uptake by the roots and water flow through the plant [3].

Salinity tolerance is a complex polygenic trait highly influenced by environmental factors and genetic-environment interaction, but we do not yet fully understand the genetic architecture [2]. Generation mean analysis is a reliable biometrical genetic approach in dissecting gene effects in a quantitative trait [5] by having an additional benefit of subtracting the variances of digenic genetic interactions (additive \times additive [i], additive \times dominance [j], and dominance \times dominance [l]) [6]. It is not only critical to exploit natural variations in wheat for saline adaptation but also crucial to understand in terms of their genetic mode of action and implementation in a breeding program. Although there are some studies on the inheritance of salinity tolerance in crop plants, most of them were done with young plants cultivated in pots under greenhouse conditions (see [7]). Koch et al. [8] investigated the inheritance of yield in salinity tolerance in ryegrass using a diallel design with six parental clones. They found relatively high narrow-sense heritability estimates for salinity tolerance. Shamaya et al. [9] used F_2 populations of durum wheat to assess the broad-sense heritability of leaf Na content and the K/Na ratio of hydroponically-grown plants exposed to 100 mM NaCl for ten days at the seedling stage under greenhouse conditions. Another study showed that salinity tolerant plant maintains a low concentration of Na in the cytosols of root and shoot cells through Na exclusion, extrusion or compartmentation under saline conditions [10]. In addition, maintenance of a high concentration of K in the cytosols help plant to minimize salinity damage under salinity stress. Therefore, the K/Na ratio in the root and shoot is an important variable when studying salinity tolerance in many plant species, including wheat [11] and barley [12]; [13].

The current study aimed to investigate the genetic basis of salinity tolerance in wheat using the agronomical and physiological performances of parents, filial generations and backcrosses grown in normal and saline field conditions.

Material And Methods

Plant materials

A salt tolerant ('Barat') and a salt-sensitive ('Nogal') wheat cultivar were crossed to produce the following filial and backcross progenies: F_1 , F_2 , BC_1 , BC_2 , and F_3 , at Research Farm of Isfahan Agricultural and Natural Resources Research and Education Center.

Field experimental and salinity stress conditions

Two parental cultivars and their five derived populations were planted in normal and salt-stress experiments using a randomized complete block design with three replicates. Number of the planting rows were as follows: 2 for parents and F_1 , 4 for each of F_2 , BC_1 , BC_2 , and 1 for each of 176 F_3 families. Thirty plants were grown in each row. The plants in the normal experiment were irrigated with freshwater ($EC_w=0.8 \text{ dSm}^{-1}$), while in the salinity-stress experiment were irrigated with 13 dSm^{-1} starting at four-leaf stage (Zadoks growth stage 14). The electrical conductivity (EC_e) was measured from the 0 to 30 cm soil depth at initial and terminal stages of plant growth. Three soil samples from each experimental plot were used, and the average EC_e were determined for the normal and saline field conditions as 1.8 and 9.6 dS

m⁻¹, respectively. Based on soil sample analysis, 30 kg P ha⁻¹ and 120 kg N ha⁻¹ fertilizers were applied before sowing. Nitrogen fertilizer was splitted into three parts and applied at sowing, tillering, and anthesis stages.

Agronomical traits

The following variables were measured: umber of days to heading date (DH), number of days to pollination date (DP), number of days to maturity date (DM), plant height (PH), peduncle length (PL), single grain weight (GW), number of grains per spike (GN), number of spikes per plant (NS), grain yield per plant (yield).

Physiological traits

Naand K concentration

Leaf samples (100 mg) were incinerated at 550 °C for 4 h. Inorganic ions were then extracted using 10 mL HCl (2 N), and the volume of each sample was standardized to 100 mL. The sodium and potassium contents of the solutions were determined by flame photometry (Jenway PFP7, UK). A standard curve was used to determine the Na and K concentrations [11]. The K/Na ratio was then calculated.

Relative water content (RWC)

A sample of fresh leaves was weighed (FW) immediately after cutting into pieces, placed in 20 mL of distilled water, and left in the dark for 24 h at room temperature. Then the weigh of turgid leaves were used as turgid weight (TW). The oven-dried (70 °C for 48 h) samples were weighted to determine the dry weights (DW). The total RWC was then determined using the below formula [13]:

$$\%RWC = [(FW - DW) / (TW - DW)] \times 100$$

Leaf chlorophyll and carotenoid concentrations

Chlorophyll a, Chlorophyll b and total carotenoid concentrations were measured using the spectrophotometer method (Hitachi F-2500 fluorescence spectrophotometer) with reading absorbance at 646, 663, and 470 nm, respectively. To determine photosynthetic pigments, 0.1 g fresh leaf tissue sampled at early heading stage (Zadoks growth stage 58) were homogenized and extracted in 15 mL acetone 80% using a centrifuge at 5000 rpm for 15 min. Chlorophylls (a and b) and carotenoids (Car) concentrations (mg/g FW) were calculated using below equations [30]:

$$\text{Chlorophyll a} = [(13.36 \times A_{663}) - (5.19 \times A_{646}) \times 8.1] / \text{FW}$$

$$\text{Chlorophyll b} = [(27.43 \times A_{646}) - (8.12 \times A_{663}) \times 8.1] / \text{FW}$$

$$\text{Carotenoids} = [(4.785 \times A_{470}) + (3.657 \times A_{663}) - (12.76 \times A_{646}) \times 8.1] / \text{FW}$$

Total chlorophyll (Tchl) was calculated as the sum of chlorophyll a and b. Only Tchl and Car were subjected to genetic analysis.

Flag leaf SPAD value and area

Flag leaf blade chlorophyll (i.e. SPAD reading) was estimated using three randomly selected flag leaves from each plot by a SPAD chlorophyll meter (SPAD-502, Konica Minolta, Japan). The measurements were conducted at the early heading stage (Zadoks growth stage 58). Flag leaf area (FLA) was measured by a leaf area meter. The measurements were carried out at the full heading stage (Zadoks growth stage 60).

Statistical analysis

The data were initially checked for normality of distribution and homogeneity of variance using Kolmogorov–Smirnov and Bartlett's tests, respectively. A combined analysis of variance (ANOVA) was used to test the effects of the environment (normal and salinity), generation, and generation by environment interaction. The statistical analyses were carried out using the SAS software package (SAS, Institute, Cary, NC, USA) and SPSS (version 26). Tukey's Honest Significant Difference (HSD) test was used to compare trait means obtained from each set of the seven genotypic groups (two parents and five progenies).

Genetic analysis

Generation mean analysis was conducted following Mather and Jinks [5] model as below:

$$Y = m + a[d] + \beta[h] + a^2[i] + 2a\beta[j] + \beta^2[l]$$

Where Y = mean of generation; m = mean of all generations; [d] = sum of additive effects; [h] = sum of dominance effect; [i] = sum of additive × additive (complementary); [j] = sum of additive × dominant (duplicate); and [l] = sum of dominant × dominant interactions; and α and β = coefficients of genetic parameters.

Mather's scaling test A, B, C and D [5] was initially conducted and then followed by the Joint scaling test of Cavalli [31] to examine the goodness-of-fit of the additive-dominance model. The standard error of the calculated scales (A, B, C and D) was t-tested for significance. When A and B scales are significant, all types of epistasis (non-allelic gene interactions) can play role in controlling the trait in question. The dominant × dominant [dd] type of epistasis is important when the C scale is significant. The significance of the D scale shows additive × additive [aa] type of epistasis [19]. On the other hand, the absence of epistasis can be confirmed by chi-square (χ^2). The best fit model is the one with a non-significant chi-square and significant estimates of the scales. If the results point out the role of epistasis, then a six-parameter genetic model (m, d, h, i, j, and l) should be fitted to the generation means [32]. This was the case with our data (see results section). Broad-sense and narrow-sense heritability were estimated following the procedures outlined by Warner [33]:

$$h^2_b = 100 \times [\sigma^2_{F_2} - (\sigma^2_{P_1} + \sigma^2_{P_2} + 2\sigma^2_{F_1})/4] / \sigma^2_{F_2}$$

$$h^2_n = 100 \times [2 - \sigma^2_{F_2} - (\sigma^2_{BC_1} + \sigma^2_{BC_2})] / \sigma^2_{F_2}$$

Components of genetic variance were calculated based on the data of six generations using the relationships suggested by Mather and Jinks [5] as follows:

$$E_w = (V_{P_1} + V_{P_2} + 2V_{F_1})/4$$

$$F = V_{BC_1} - V_{BC_2}$$

$$D = 4V_{F_2} - 2(V_{BC_1} + V_{BC_2})$$

$$H = 4(V_{BC_1} + V_{BC_2} - V_{F_2} - V_E),$$

Where, E_w = environmental variance; D = additive variance; H = dominance variance; V_{F_2} , V_{F_1} , V_{BC_1} , V_{BC_2} , V_{P_1} and V_{P_2} are variance of F_2 , F_1 , BC_1 , BC_2 , P_1 and P_2 , respectively; and F = joint contribution on all loci.

Results

Agronomical traits

The results of combined-ANOVA for the traits studied in both normal and salinity stress conditions are presented in Table 1. There were significant effects of both salinity and genotypes (families/generations) on all the traits. The genotype \times environment interaction for most of the traits except for PH, PL, FLA, DH, DP, DM was significant. Given the existence of genetic variation in the tolerance to salinity, genetic analysis can be undertaken to estimate different genetic parameters such as components of genetic variance and the heritability of salinity tolerance.

'Barat' exhibited higher PL and FLA than 'Nogal' but had lower yield and RWC in both environmental conditions (Tables 2 and 3). In addition, 'Barat' performed better than another parent in terms of GW and GN in salinity stress conditions (Table 3). At the same time, the highest mean values of the traits mentioned above were obtained by 'Nogal' in normal conditions (Tables 2). 'Barat' had a lower rate of Na accumulation and a higher K/Na discrimination compared with 'Nogal' in both treatments. The GW, NS, and yield in F_1 were higher than those of the two parents in normal and salinity stress conditions. The F_1 values for PL, DH, DM, SPAD were almost intermediate between those of the parents in both treatments. The mean of trait values in the F_2 generation was lower than the corresponding values for the F_1 generation (Tables 2 and 3). Furthermore, generations differed in GW, GN, and Car within each environment and did respond differently to the two environmental conditions (Tables 2 and 3).

Goodness-of-fit of the genetic model

The results of scaling tests can be used to test the hypothesis of the goodness of fit of the three-parameter model (additive-dominance model) or non-significant effect of epistasis (non-allelic interaction). The results of both individual and joint scaling tests summarized in Table 4 did not support the hypothesis of the additive-dominance model for all the traits in both environmental conditions. In addition, the majority of the traits showed more than one significant scale, indicating the combined effects of epistasis. The six-parameter model was subsequently utilized to assess the genetic effects governing the traits in this study.

Gene action for the inheritance of agronomical and physiological traits

The estimates of additive, dominance, and epistatic genetic effects controlling the traits evaluated in normal and salinity-stress field conditions are given in Tables 5 and 6, respectively. In both normal and salinity stress treatments, the mean genetic effect (m) was significant for all the traits. The six-parameter model showed that additive gene effect was the major contributing gene action in the inheritance of PH in the normal conditions. In salinity stress conditions, the additive, additive \times additive, and dominance \times dominance effects were the significant contributing factors in the expression of PH. In addition, there was a negative and significant dominant effect [h] contributing in PH, represents a shortening effect on the height in both conditions. Additive, dominance, and additive \times additive gene effects were significant for PL in normal conditions (Table 5). In contrast, the additive and additive \times additive gene interaction played a significant role in governing PL in salinity stress conditions (Table 6).

Additive and additive \times additive were the main types of gene actions related to the inheritance of leaf K and Na concentrations in both environmental conditions. At the same time, additive, dominance, additive \times additive, and dominance \times dominance effects were the likely mode of gene actions in the inheritance of K/Na ratio under both conditions (Tables 5 and 6). Additive, additive \times additive, and dominance \times dominance effects were the major contributors to the inheritance of NS in normal conditions, while it was predominantly controlled by additive and additive \times additive types of gene action in salinity treatment (Table 6). Under normal conditions, NS and GN were coordinately inherited with grain yield, while GN and yield were found to have homogeneous genetics in salinity stress conditions. The mode of inheritance of grain yield was dependent upon dominance and dominance \times dominance actions of the genes in both environments.

Heritability and components of genetic variance

The components of genetic variance (additive, dominance, and direction of dominance), average degree of dominance, broad-sense heritability (h^2_b), narrow-sense heritability (h^2_n), and for the traits in normal and salinity stress conditions are presented in Table 7 and 8, respectively. The overall comparison between additive (D) and dominance (H) variances showed that additive genetic variance has a higher contribution than dominance variance for PH, DM, PL, FLA, GW, NS, GN, RWC, SPAD, K/Na, Car traits in normal conditions. In contrast, dominance variance was larger than the additive variance for yield, DH, DP,

K, Tchl. In salinity stress conditions, additive genetic variance was more significant than dominance variance for DH, DM, PL, FLA, GW, NS, K, K/Na traits. In contrast, dominance variance was more important for yield, RWC, SPAD, GN, PH, and DP. As expected, the h^2_b estimates were higher than the h^2_n estimates for all the traits in both environments. Moreover, both the h^2_b and h^2_n estimates were higher for normal (Table 7) than stress (Table 8) conditions. The h^2_b ranged from 30 to 94 percent in normal and 21 to 75 percent in stress conditions. The range of h^2_n was from 14 to 41 percent in normal and 1 to 38 percent in stress conditions. The h^2_n estimates were higher for FLA (38%), and NS (33%) intermediate for K/Na (29%), and lower for SPAD (13%), DP (12%), PH (10%), and yield (1%) under stress conditions. The low h^2_n estimates in normal conditions were 16% for K, 15% for DP, and 14% for yield. The highest h^2_b values were recorded for Tchl (94%) followed by Car (83%), DP (66%), Na (60%), and K/Na (50%) in normal conditions, whereas the highest values were observed for Car (75%), Tchl (63%), Na (60%), and K/Na (48%) in saline conditions.

The significance of dominance effects relating to the additive deviations of genes is determined by the average degree of dominance $[(H/D)^{1/2}]$. Accordingly, in this study, the ratio of $(H/D)^{1/2}$ for some of the traits was lower than one, indicating the likely importance of the overdominance type of gene action in the inheritance of these traits. The direction of dominance (F) was positive for NS, RWC, Na, Tchl, Car, and yield in both environments, DP and GN in normal, and PH and SPAD in stress environment. These show the existence of more dominant alleles than recessive alleles in parents for the traits mentioned above, that showed positive F values. The negative 'F' value of K and K/Na showed the predominance of recessive alleles governing these traits in the parents grown under saline conditions. In contrast, it was reversed in the case of Na.

Discussion

Knowledge of genetic components of tolerance would be vital for developing salinity tolerant cultivars in wheat through its implications for the design and deployment of appropriate tools and strategies [7]. Despite the large body of data on the genetic studies carried out in non-saline conditions [14]; [15], little has been known about the inheritance of salinity tolerance in wheat. For the traits studied in the current study, (i) the assumption of significant difference ($p < 0.01$) among parental, F_1 , F_2 , BC_1 , and BC_2 generations allows us to conduct the genetic analysis; (ii) the rejection of null-hypothesis of the additive-dominance model (three-parameter) guides us to consider an alternative model (six-parameter); and (iii) the significant mean effect "m" revealed by the six-parameter model for all the traits, supports the quantitative inheritance of the traits studied. One of the central findings from our study was that different gene actions governing the traits in the two environmental conditions (normal and salinity). Does this imply that breeding strategy for each trait should be developed per a basic gene action in the trait of interest in each of the environments? In general, the answer is "yes", though we should expand an identical strategy to those are similar in gene action.

All the four 'A', 'B', 'C', and 'D' scales were significant for DM and yield in stress conditions. These shows that not only additive and dominance but also other types of gene action such as epistasis may likely contribute to the genetics of DM and yield in wheat when grown under saline conditions. These results are in close agreement with the recent report of Attri et al. [16] on DM and yield in three bread wheat crosses based on normal growth conditions. Yield is the most crucial agronomic trait for selecting to abiotic stress tolerance, including salinity tolerance [2]. Though a high and stable crop yield is the ultimate goal of plant breeders, selection for a complex quantitative trait such as salinity tolerance is a difficult task. The stability of such trait is affected strongly by the interaction between genotype and environmental factors [17]. Thus, we took appropriate account of environmental impacts in the using a two-year field study. In the current study, salinity stress caused a significant decrease in yield and significant genotypic variation for salinity tolerance, findings that are consistent with previous research [7]; [13]; [18].

Additive gene action was found to be one of the positive and significant genetic components in the inheritance of Na, K, and K/Na in salinity stress conditions. Therefore, recurrent selection-based breeding schemes are effective in improving salinity tolerance since we deal with a fixable component of genetic variance [19]. Similar results were found for PH, PL, and NS. These results are somehow consistent with the observations by Yao et al. [20], who showed significant additive gene effect for the PH and PL in wheat under normal environment. Alternatively, given that wheat is a self-fertilized crop, the substantial contribution of additive gene effects in governing the agronomical traits, especially in normal growth conditions. A significant dominance effect for yield and GN under both conditions may suggest that heterosis is an outcome of development of hybrids by crossing contrasting pure lines [5]; [21]. In contrast, the negative dominance effect obtained for PH under both normal and salinity stress treatments showed that the alleles responsible for short height were dominant over the alleles representing tall plant status [22]. The dominance effect alone [h] and dominance by dominance effect [I] components showed values in the opposite direction for some of the traits like K/Na in both normal and stress conditions, indicating the possibility of duplicate epistasis. A disruption of the selection process through reducing diversity in the segregation generations would be resulted from this type of epistasis. Hence, the selection process should be postponed till reaching an acceptable level of the genes with fixed effects. On the other hand, the presence of predominantly duplicate epistasis would point to the preference of the development of hybrid cultivars. In addition, a higher possibility of transgressive segregants leads to the selection of superior recombinant inbred lines through hybridization procedure. Indeed, the opposite signs of h and I types of gene mode actions counterbalance each other, thus resulting in decreased heterosis [16]; [22]. Our data obtained from salinity-stress conditions are consistent with a recent study using a six-parameter model in wheat that reported significant duplicate epistasis for PH and yield in normal conditions [16]. Such complementary interactions could be increased the variation between the generations. Indeed, the differences in gene action types between normal and stress conditions indicate that a distinct set of genes are activated to protect the plant from stress when a plant is subjected to salinity stress [2].

Significant additive \times additive effect for DP in normal conditions suggests that epistasis between alleles of different loci contributes to decreasing the DP expression [21]. The yield and GN traits were

predominantly affected by dominance and epistatic gene effects. The significant effects of dominance and epistatic types of gene action in the expression of grain yield in wheat has been reported [23]. These results are also consistent with those obtained by Sharma et al. [24].

The significant effects of the additive and additive \times additive that were higher than the dominance effect for NS in salinity stress conditions may reflect an appreciation of enhanced response to selection. Negative additive \times additive interaction for some of the studied traits, such as DP in normal treatment, shows the potential for reducing these traits along with fixation of additive effects in the subsequent generations [25]. The slightly larger dominance genetic variance than the additive one for PH in saline conditions indicates the comparable role of both types of gene action in determining plant status.

Higher narrow-sense heritability estimates were found for K/Na ratio than either K or Na in saline conditions. In contrast, higher broad-sense heritability estimates were found for Na in both stress and normal field conditions. Together, these may imply the distinct mode of gene action in saline field environment and that K/Na is the most efficient screening criterion for salinity tolerance in wheat, as emphasized previously [11]; [26]. Therefore, these findings and their twin implications strongly support that inference that selection for salinity tolerance should be considered a distinct territory for wheat breeding. One possible reason for the higher heritability of K/Na is not related to quantitative genetics per se but is rather due to the involvement of major genes. This inference has been supported in experimental research by Dashti et al. [26], and the same general point has been made from other perspectives. The new finding combines the importance of maintaining a high K^+/Na^+ ratio in the leaves with the established knowledge of mechanism underlying the substantial contribution of the *Kna1* locus in salinity tolerance via shoot Na^+ exclusion in bread wheat ([2]; [3]; [27]). The *Kna1* locus is located in chromosome 4DL [28], and thus, the selective accumulation of K over Na in the shoot appears to be more specific to bread wheat and less relevant in durum wheat lacking D genome. The higher heritability of K/Na than its counterparts provides further support for the crucial role of K/Na in the success of wheat breeding program for salt tolerance in terms of response to selection [26]. These findings are in line with the review by Benito et al. [29] who highlighted the significant of the additive effects of the genes controlling K/Na ratio and its high h^2_n . The difference between h^2_n and h^2_b is an estimate for the extent of contribution of the dominance effect in the inheritance of the trait in question. Though there were some inconsistencies between the results of generation mean analysis and heritability estimates, the data of generation mean analysis are more reliable as based on a systematized quantitative genetic design [15].

Conclusions

Over the past decades, although the knowledge of genetics and genomics has improved substantially, the additional insights would help the breeder to plan more accurate breeding tools and selection strategies. This study may provide a better understanding of the genetic architecture underlying quantitative trait variation related to salinity tolerance in wheat. Though K/Na possessed higher narrow-sense heritability than either K or Na, additive gene action is a significant contributor to the genetic components controlling

Na, K, and K/Na in salinity stress conditions. The yield and GN were predominantly governed by dominance and epistatic modes of gene action. Thus, any breeding strategy for improving these traits should depend on hybrid development especially in saline environment. The findings might also have broader implications for those interested in the genetic mechanisms of salinity tolerance and in improving cultivars for tolerance to salinity in wheat.

Abbreviations

Car: Carotenoid

FLA: Flag leaf area

DH: Number of days to heading date

DP: Number of days to pollination date

DM: Number of days to maturity date

GN: Number of grains per spike

GW: grain weight

K: Potassium

Na: Sodium

NS: Number of spikes per plant

PH: Plant height

PL: Peduncle length

RWC: Relative water content

Tchl: Total chlorophyll

Yield: Grain yield per plant

m: mean

[d]: additive effect

[h]: dominance effect

[i]: additive × additive effect

[j]: dominance × dominance effect

[I]: additive × dominance effect

Declarations

Ethics approval and consent to participate

The seeds of wheat cultivars, 'Barat' and 'Nogal', were procured from Seed and Plant Improvement Institute, Karaj, Iran. In this work, we did not use transgenic material or technology thus it does not require ethical approval.

The experimental research on plants, including sampling of plant materials, was complied with institutional, national, or international guidelines. The field study was conducted in accordance with local legislation.

Consent for publication

Not applicable.

Availability of data

The raw data supporting the conclusions of this article will be made available by the authors, without undue reservation.

Competing interests

The authors declare that they have no competing interests.

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Authors' contributions

SO, AA, MEM participated in the conceived and designed the research. SO conducted the experiments under the supervision of AA, MEM, and MM. SO performed the data analysis, and drafted the manuscript with significant inputs from AA, MEM, and MM. All authors read and approved the final manuscript.

References

1. Arzani A, Ashraf M. Cultivated ancient wheats (*Triticum* spp.): A potential source of health-beneficial food products. *Compr. Rev. Food Sci. Food Saf.* 2017; 16(3): 477-488.
2. Arzani A, Ashraf M. Smart engineering of genetic resources for enhanced salinity tolerance in crop plants. *Crit. Rev. Plant Sci.* 2016; 35(3): 146-189.
3. Munns R, Tester M. Mechanisms of salinity tolerance. *Annu. Rev. Plant Biol.* 2008; 59: 651-681.
4. Maas EV, Hoffman GJ. Crop salt tolerance-current assessment. *J. Irrig. Drain. Div.* 1977; 103: 15-34.
5. Mather K, Jinks JL. *Introduction Biometrical Genetics*. 3rd Edition, Chapman and Hall, London, 1982; 396.
6. Kearsley MJ, Pooni HS. *The genetical analysis of quantitative traits*. Chapman and Hall, London, 1996; 380.
7. Akrami M, Arzani A. Inheritance of fruit yield and quality in melon (*Cucumis melo* L.) grown under field salinity stress. *Sci. Rep.* 2019; 9(1): 1-13.
8. Koch MJ, Meyer WA, Bonos SA. Inheritance of salinity tolerance in perennial ryegrass. *Crop Sci.* 2015; 55(4): 1834-1842.
9. Shamaya NJ, Shavrukov Y, Langridge P, Roy SJ, Tester M. Genetics of Na⁺ exclusion and salinity tolerance in Afghani durum wheat landraces. *BMC Plant Biol.* 2017; 17(1): 1-8.
10. Munns R, Day DA, Fricke W, Watt M, Arsova B, Barkla BJ, Bose J, Byrt CS, Chen ZH, Foster KJ, Gilliam M. Energy costs of salt tolerance in crop plants. *New Phytol.* 2020; 225(3): 1072-1090.
11. Houshmand S, Arzani A, Maibody SAM, Feizi M. Evaluation of salt-tolerant genotypes of durum wheat derived from in vitro and field experiments. *Field Crops Res.* 2005; 91(2-3): 345-354.
12. Mahlooji M, Sharifi RS, Razmjoo J, Sabzalian MR, Sedghi M. Effect of salt stress on photosynthesis and physiological parameters of three contrasting barley genotypes. *Photosynthetica* 2018; 56(2): 549-56.
13. Ebrahim F, Arzani A, Rahimmalek M, Sun D, Peng J. Salinity tolerance of wild barley *Hordeum vulgare* ssp. *spontaneum*. *Plant Breed.* 2020; 139(2): 304-316.
14. Said AA. Generation mean analysis in wheat (*Triticum aestivum* L.) under drought stress conditions. *Ann. Agric. Sci.* 2014; 59(2): 177-184.
15. Saleem S, Kashif M, Hussain M, Khan AS, Saleem MF. Genetic behavior of morpho-physiological traits and their role for breeding drought tolerant wheat. *Pak. J. Bot.* 2016; 48(3): 925-933.

16. Attri H, Dey T, Singh B, Kour A. Genetic estimation of grain yield and its attributes in three wheat (*Triticum aestivum* L.) crosses using six parameter model. *J. Genet.* 2021; 100(2): 1-9.
17. Arzani A. Improving salinity tolerance in crop plants: a biotechnological view. *In Vitro Cell. Dev. Biol. Plant.* 2008; 44(5): 373-383.
18. Sairam RK, Rao KV, Srivastava GC. Differential response of wheat genotypes to long term salinity stress in relation to oxidative stress, antioxidant activity and osmolyte concentration. *Plant Sci.* 2002; 163(5): 1037-1046.
19. Hayman BI. The separation of epistatic from additive and dominance variation in generation means. *Heredity.* 1958; 12: 371-391.
20. Yao JB, Ma HX, Ren LJ, Zhang PP, Yang XM, Yao GC, Zhang P, Zhou MP. Genetic analysis of plant height and its components in diallel crosses of bread wheat (*Triticum aestivum* L.). *Aust. J. Crop Sci.* 2011; 5(11): 1408-1418.
21. Almeida VC, Viana JMS, Risso LA, Ribeiro C, DeLima RO. Generation mean analysis for nitrogen and phosphorus uptake, utilization, and translocation indexes at vegetative stage in tropical popcorn. *Euphytica.* 2018; 214: 103.
22. Amiri R, Bahraminejad S, Cheghamirza K, Arzani, A. Genetic analysis of iron and zinc concentrations in bread wheat grains. *J. Cereal Sci.* 2020; 95: e103077.
23. Raikwar RS. Genetic architecture of yield and quality traits in wheat (*Triticum aestivum* L.). *Indian J. Genet. Plant Breed.* 2019; 79 (1): 100-103.
24. Sharma SN, Sain RS, Sharma RK. Genetic control of quantitative traits in durum wheat under normal and late-sowing environments. *Sabao J. Breed. Genet.* 2002; 34: 35-43.
25. Kere GM, Guo Q, Shen J, Xu J, Chen J. Heritability and gene effects for salinity tolerance in cucumber (*Cucumis sativus* L.) estimated by generation mean analysis. *Sci. Hortic.* 2013; 159: 122-127.
26. Dashti H, Naghavi MR, Tajabadipour A. Genetic analysis of salinity tolerance in a bread wheat cross. *J. Agric. Sci. Technol.* 2010; 12: 347-356.
27. Hussain S, Hussain S, Ali B, Ren X, Chen X, Li Q, Saqib M, Ahmad N. Recent progress in understanding salinity tolerance in plants: Story of Na⁺/K⁺ balance and beyond. *Plant Physiol. Biochem.* 2021; 160: 239-256.
28. Gorham J, Hardy C, Wyn Jones RG, Joppa LR, Law CN. Chromosome location of a K/Na discrimination character in the D genome of wheat. *Theor. Appl. Genet.* 1987; 74:584-588.

29. Benito B, Haro R, Amtmann A, Cuin TA, Dreyer I. The twins K⁺ and Na⁺ in plants. *J. Plant Physiol.* 2014; 171(9): 723-731.
30. Lichtenthaler HK, Buschmann C. Chlorophylls and carotenoides: measurement and characterization by UVVIS spectroscopy. John Wiley and Sons, Inc, New York. 2001.
31. Cavalli LL. An analysis of linkage in quantitative inheritance. An analysis of linkage in quantitative inheritance. In: E. C. R. Reeve & C.H. Waddington (Eds), *Quantitative Inheritance*. 1952; 135-144. HMSO, London.
32. Jinks JL, Jones RM. Estimation of the components of heterosis. *Genetics*. 1958; 43(2): 223-234.
33. Warner JN. A method of estimating heritability. *J. Agron.* 1952; 44: 427-430.

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