The Efficiency of Physiological Markers for Screening Salt-Tolerant Stock (Matthiola incana L.) Cultivars

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Abstract

Stock (*Matthiola incana* L.) is one of the most commercial cut-flowers in the world. To evaluate the efficiency of physiological markers for salt tolerance, 10 cultivars of stock were selected and their seedlings were irrigated with salt water (6 dS/m) from four true-leaf stage for six weeks. The dry weight of shoots and roots, and K^+, Na^+ and Ca^{2+} concentrations in shoots were evaluated in the study. Based on the results, dry weights of shoots and roots had positive correlations with K^+/Na^+ selectivity coefficient. This indicates that K^+/Na^+ ratio in shoots has a direct correlation with the amount of plant growth under salinity conditions. Also, a negative correlation was found between Ca^{2+} and K^+ accumulation and dry weights of the shoots. There was a low Ca^{2+} to Na^+ ratio in the shoots, which confirms the importance of the K^+ accumulation rate in the shoots or the K^+/Na^+ selectivity coefficient, as the determinant factor of salt tolerance rate in stock. In conclusion, 'Column Lilac Lavender', 'Column Lavender' and 'Column Cooper' with the highest K^+/Na^+ ratio in shoots, were considered as the most tolerant, and 'Column Mid-Blue' and 'Column Rose' with the lowest K^+/Na^+ ratio, as the most sensitive cultivars to salinity. The physiological markers used in this study can be introduced for large-scale screening of salt-tolerant varieties of the stock.

Introduction

Many plants face significant stressors in their natural environments, including salinity and drought (Iwuala et al. 2023). Of these, salinity stress poses a significant threat to the global ecosystem, crop production, food security, and sustainable agriculture (Pistelli et al. 2023). Salinity is a major abiotic stressor that negatively impacts plant growth and physiological processes (Shahbaz and Ashraf 2013; Yamaguchi-Shinozaki and Shinozaki 2005; Ma et al. 2023), posing a significant obstacle to agricultural production (Kamanga et al. 2020). With salinity affecting 10% of the world's total arable land, it has the potential to become a significant threat to feeding the growing population (Mishra et al. 2023).

High levels of salinity can cause stress in plants in two primary ways: first, it reduces water uptake by roots, resulting in reduced water absorption, which adversely affects plant growth and cellular metabolism. Second, it disrupts tissue and cellular functions in different parts of the plant, particularly in photosynthetic organs, due to ion toxicity, depending on the salt level (Munns and Tester 2008). Strategies to mitigate the deleterious effects of salinity, such as saline land improvement, improvement of saline water, and specific techniques, can be costly and temporary (Lauchli and Epstein 1990, Marschner 1995; Munns 1993). To ensure sustainable production in saline soils, it is crucial to identify and introduce cultivars capable of growing and producing economically acceptable yields under saline conditions. This will help to reduce the adverse effects of salt stress on plants (Epstein et al. 1980; Fooland 1996) and achieve the primary objective of sustainable agriculture (Assaha et al. 2017).

Stock, a plant belonging to the Brassicaceae family, is highly valued for its fragrance, color, and shape of flowers. However, there is limited research on identifying salt-tolerant cultivars of stock flowers that can be grown in arid and semi-arid regions with less than 500mm mean annual precipitation. While many cultivars can tolerate moderately saline soil, high levels of salinity can impair growth and yield. Therefore, it is necessary to introduce salt-tolerant cultivars that can produce economically acceptable yields under saline conditions.

A study was conducted to assess the impact of salinity on the growth and ionic relations of two cultivars of stock flowers, ‘Frolic’ and ‘Cheerful White’. The results indicated that increasing salinity from 2.5 dS/m to 14 dS/m led to a significant decrease in the main stem height of both cultivars when they reached 50% flowering. However, ‘Frolic’ was found to be significantly shorter than ‘Cheerful White’.

Another study evaluated the effects of different levels of salinity (ranging from 1–5 dS/m) on two types of stock flowers, ‘Pan American’ and ‘Cinderella’. Both cultivars experienced significant changes in their morphological, physiological, and biochemical properties when exposed to high levels of salinity stress. However, both cultivars showed similar resistance to salt in low concentrations of NaCl in irrigation water.

To survive salt stress, plants require adequate potassium supply (Lotfi et al. 2009). While K^+ accumulation substantially reduces in shoots of stock flower cultivars, the selectivity coefficient of K^+/Na^+ increases up to 60% when salinity increases from 2.5 dS/m to 14 dS/m. This increase was also assessed in tomato seedlings. K^+/Na^+ ratio is a suitable parameter for early screening of salt-tolerant tomato cultivars.

In conclusion, identifying and introducing salt-tolerant cultivars of stock flowers that can grow and produce economically acceptable yields under saline conditions is crucial for sustainable agriculture. The K^+/Na^+ ratio is a suitable parameter for early screening of salt-tolerant tomato cultivars and a significant linear relationship was reported between K^+/Na^+ ratio and grain yield in rice.

Different wheat and quinoa cultivars were studied to evaluate their tolerance to salt toxicity. The results showed that salt-tolerant cultivars exhibited better growth, gas exchange characteristics, and stronger antioxidant systems, along with less oxidative stress. Furthermore, the salt-tolerant cultivars accumulated higher levels of osmolytes, hydrogen sulfide, and nitric oxide, making these physiological and biochemical markers promising for screening tolerant cultivars under salinity conditions. Among the four quinoa cultivars studied, ‘Red Faro’ and ‘UDEC-5’ showed low levels of Na^+ accumulation and high K/Na selectivity. ‘UDEC-5’ also exhibited high stomatal conductance, leading to high net photosynthesis even at high NaCl concentrations. In addition, Faro red and UDEC-5 showed low levels of lipid peroxidation, high antioxidant activities, and high proline accumulation, indicating strong defense against ROS and efficient osmotic regulation. The study also demonstrated the high diversity and intraspecific genetic differences in stock flowers, which can be used to better understand the mechanisms of salinity tolerance in this plant and evaluate physiological responses using various cultivars. Overall, the study's findings may be useful in identifying salt-tolerant varieties of stock flowers that can withstand climate change conditions.

Materials and Methods

The experiment was conducted in a research greenhouse at the College of Aburaihan, University of Tehran, located in Pakdasht, Iran. It involved 10 improved stock cultivars (Table 1), planted in 20 cm diameter pots filled with a mixture of garden soil, perlite, and peat moss in a ratio of 2:1:1. The seeds of the stock were planted in late February, and prior to seeding, a soil sample was taken to determine pH and electrical conductivity. The greenhouse had an average day/night temperature of 27/20°C, a day length of 10–13 hours, and relative air humidity of 60%.

Salt stress was induced 32 days after planting, when plants had four fully expanded leaves. To create an EC value of 6 ds/m (as described by Heuer and Ravina, 2004), appropriate amounts of $CaCl_2$: NaCl were added to distilled water in a ratio of 2:8.

Plants were harvested six weeks after planting, and their height was measured using metal meters in centimeters. The plants were then divided into two parts: the shoot (including stem, young and old leaves) and the root. After a thorough washing of both parts with distilled water, they were dried at 70°C for 72 hours and weighed.

Following this, the vegetative tissues of the aerial parts were ground and one gram of plant powder was put into a porcelain capsule and placed in an electric oven at 550°C for six hours until the plant organic matter was burned and only the mineral remained as ash. After that, 10ml of 2N hydrochloric acid was added to the ash, and the resulting solution was heated in a water bath until half of the acid evaporated and the solution turned yellow. This solution was used to measure the elements potassium, calcium, and sodium by atomic absorption spectrometer.

The experimental design was a completely randomized design (CRD) with six replications. The data was analyzed using SAS software, and Duncan's multiple range test was applied to compare the means. SPSS software was also used to calculate correlation coefficients between the traits and perform statistical cluster analysis of the cultivars.

Results

Shoot dry weight

‘Column Lavender’ and ‘Excelsior Purple-Heart’ had the highest shoot dry weight, while ‘Column Mid-Blue’ and ‘Column Rose’ had the lowest.

Root dry weight

‘Column Cooper’ cultivar produced the highest root dry weight, while ‘Column Mid-Blue’ and ‘Column Rose’ cultivars had the lowest.

Plant height

Based on the analysis of variance, there was no significant difference in bush height among the cultivars when exposed to water salinity levels of 6 dS/m.

Shoot K^+

Based on the results, the highest amount of potassium accumulation was observed in the 'Lilac-Lavender', 'Crimson', 'Lavender' and 'Cooper' cultivars, while the lowest was in 'Mid-Blue' and 'Rose' cultivars.

Shoot Ca^{2+}

Based on the results, the cultivar with the highest Ca^{2+} accumulation was 'Column Mid-Blue', while the lowest was observed in 'Centum Carmine' and 'Column White' cultivars.

Shoot Na^+

There was no significant difference in Na^+ accumulation in shoots among the different cultivars, according to the results.

Shoot K^+/Na^+

The results indicated that the selectivity coefficient of K^+/Na^+ varied significantly across different cultivars. 'Column Lilac-Lavender' cultivar exhibited the highest rate, while 'Column Mid-Blue' and 'Column Rose' cultivars had the lowest rates.

Shoot Ca^{2+}/Na^+

There was no significant difference between the cultivars in terms of the selectivity coefficient of Ca^{2+} and Na^+.

Cluster Analysis

The results of the cluster analysis, which used the UPGMA clustering method and Euclidean distance coefficient, indicated that the cultivars 'Column Lavender', 'Column Cooper' and 'Column Lilac-Lavender', with the highest K^+/Na^+ ratio, were grouped together in the same cluster (Fig. 6). On the other hand, the cultivar 'Column Mid-Blue', which had the lowest K^+/Na^+ selectivity coefficient and was less similar to other cultivars, did not belong to any group. The cultivar 'Column Rose' had the least similarity with the other cultivars and a low selectivity coefficient, which caused it to stand separately.

Based on these observations, we can infer that the selectivity coefficient of K^+/Na^+ is a crucial physiological indicator that is used to estimate the efficiency of the ionic selectivity mechanism in stock flower cultivars. According to this physiological parameter, the cultivars 'Column Lavender', 'Column Lilac-Lavender', and 'Column Cooper' with the highest K^+/Na^+ ratio, along with 'Column Mid-Blue' and

‘Column Rose’, with the lowest K^+/Na^+ ratio, could be considered as the most and least tolerant cultivars, respectively, to salinity in the current research.

Morphological and physiological traits, along with their correlation coefficients, were measured to screen for salt-tolerant stock flower cultivars.

Table 2
Physiological traits measured in stock flower cultivars under salinity stress

Cultivars	Shoot DW	Root DW	Shoot Ca^{2+}	Shoot K^+	Shoot K^+/Na^+
Centum Carmine	1.72 ± 0.22abc	0.53 ± 0.07ab	1.91 ± 0.09c	2.66 ± 0.14abc	1.00 ± 0.18ab
Column Lilac- Lavender	1.89 ± 0.22ab	0.56 ± 0.07ab	2.14 ± 0.09bc	3.22 ± 0.14a	1.44 ± 0.18a
Column Rose	0.9 ± 0.22bc	0.31 ± 0.07b	2.99 ± 0.09a	1.09 ± 0.14e	0.42 ± 0.18bc
Column Crimson	1.71 ± 0.22abc	0.59 ± 0.07ab	2.42 ± 0.09b	3.09 ± 0.14a	1.05 ± 0.18ab
Column Mid-Blue	0.79 ± 0.22c	0.29 ± 0.07b	3.17 ± 0.09a	1.19 ± 0.14e	0.21 ± 0.18c
Column Gold	1.90 ± 0.22ab	0.50 ± 0.07ab	2.06 ± 0.09bc	2.39 ± 0.14bdc	0.84 ± 0.18abc
Column Lavender	2.31 ± 0.22a	0.69 ± 0.07a	2.39 ± 0.09b	3.02 ± 0.14ab	1.34 ± 0.18a
Excelsior Purple- Heart	2.14 ± 0.22a	0.54 ± 0.07ab	2.38 ± 0.09b	1.89 ± 0.14d	0.54 ± 0.18bc
Column White	1.55 ± 0.22ab	0.47 ± 0.07ab	1.97 ± 0.09bc	2.32 ± 0.14dc	0.52 ± 0.18bc
Column Cooper	1.95 ± 0.22ab	0.75 ± 0.07a	2.28 ± 0.09bc	3.02 ± 0.14ab	1.24 ± 0.18a

Table 3
Correlation coefficients among the traits measured for screening of salt-tolerant cultivars

Traits	Shoot DW	Root DW	Shoot Ca^{2+}	Shoot K^+	Shoot K^+/Na^+	Shoot Ca^{2+}/Na^+	Plant height	Shoot Na^+
Shoot DW	1							
Root DW	0.86**	1						
Shoot Ca^{2+}	-0.69*	-0.57	1					
Shoot K^+	0.76**	0.86**	-0.71*	1				
Shoot K^+/Na^+	0.71*	0.83**	-0.54	0.92**	1			
Shoot Ca^{2+}/Na^+	0.17	0.32	0.19	0.25	0.56	1		
Plant height	0.56	0.4	-0.61*	0.72*	0.68*	0.19	1	
Shoot Na^+	-0.68*	-0.74*	0.53	-0.78**	-0.91**	-0.68*	-0.62*	1

*, **, ^{ns}: Statistically significant at *P < 0.01*, *0.05*, and non-significant, respectively

Discussion

The research analyzed the response of different stock flower cultivars to water salinity, with a particular focus on the accumulation of K^+, Ca^{2+}, and Na^+ in the plant shoots. The results showed that while there was no significant difference among cultivars in response to water salinity at 6 dS/m, the dry weight of root and shoot did show a statistical difference. These findings are consistent with a previous study by Grieve et al. (2006), which reported a reduction in stem length of certain cultivars when exposed to higher salt concentrations. Additionally, there were highly significant differences among cultivars in terms of K^+ and Ca^{2+} accumulation, however, Na^+ quantities did not differ considerably among the cultivars.

Of the selectivity coefficients of K^+/Na^+ and Ca^{2+}/Na^+ measured, only K^+/Na^+ was statistically significant. The cultivars ‘Column Cooper’, ‘Column Lavender’ and ‘Column Lilac-Lavender’ had the highest selectivity coefficient of K^+/Na^+ and the highest shoot dry mass, while ‘Column Mid-Blue’ had the lowest K^+/Na^+ and therefore the lowest shoot dry weight.

The correlation results among the parameters revealed significant and positive correlations between K^+ accumulation in shoots and dry mass of root and shoot. However, the amount of Ca^{2+} accumulated in shoots was negatively correlated with shoot dry weight, and stored K^+ in shoots. A significant and negative correlation was also found between the amount of Na^+ and root dry weight, shoot dry weight, accumulation of K^+, K^+/Na^+ and Ca^{2+}/Na^+ selectivity coefficients, and plant height. The highest correlation was observed between the quantity of Na^+ accumulation and the selectivity coefficient of K^+/Na^+.

It was concluded that under the current research conditions, the amount of K^+ accumulated in shoots determines salt tolerance in stock flower. The plant is able to accumulate Ca^{2+} at higher levels under salinity stress, similar to other members of the Brassicaceae family. The stock flower tolerates salinity through controlling Na^+ absorption *via* salt excretion mechanism and maintenance of high K^+/Na^+ ratio.

The study suggests that the K^+/Na^+ selectivity coefficient is a good criterion for screening salt-tolerant cultivars of stock flower, which is consistent with previous studies. Using this physiological trait as a pre-selection index, stock flower genotypes can be screened at the seedling stage, which is saving time and resources in breeding programs. The results obtained from this study are comparable to those obtained from screening 54 tomato genotypes for salt tolerance by Yildiz Dasgan et al. (2002).

Declarations

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Author Contributions: M.A. and M.R.R. led, coordinated and designed the project. Z.H.Z.E, M.A., M.R.R., M.L., M.D and K.V. performed the experiment, analyzed the data and wrote the manuscript. All authors commented on the results and the manuscript. All authors have read and agreed to the published version of the manuscript.

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Table 1

Table 1 is available in the Supplementary Files section.

Figures

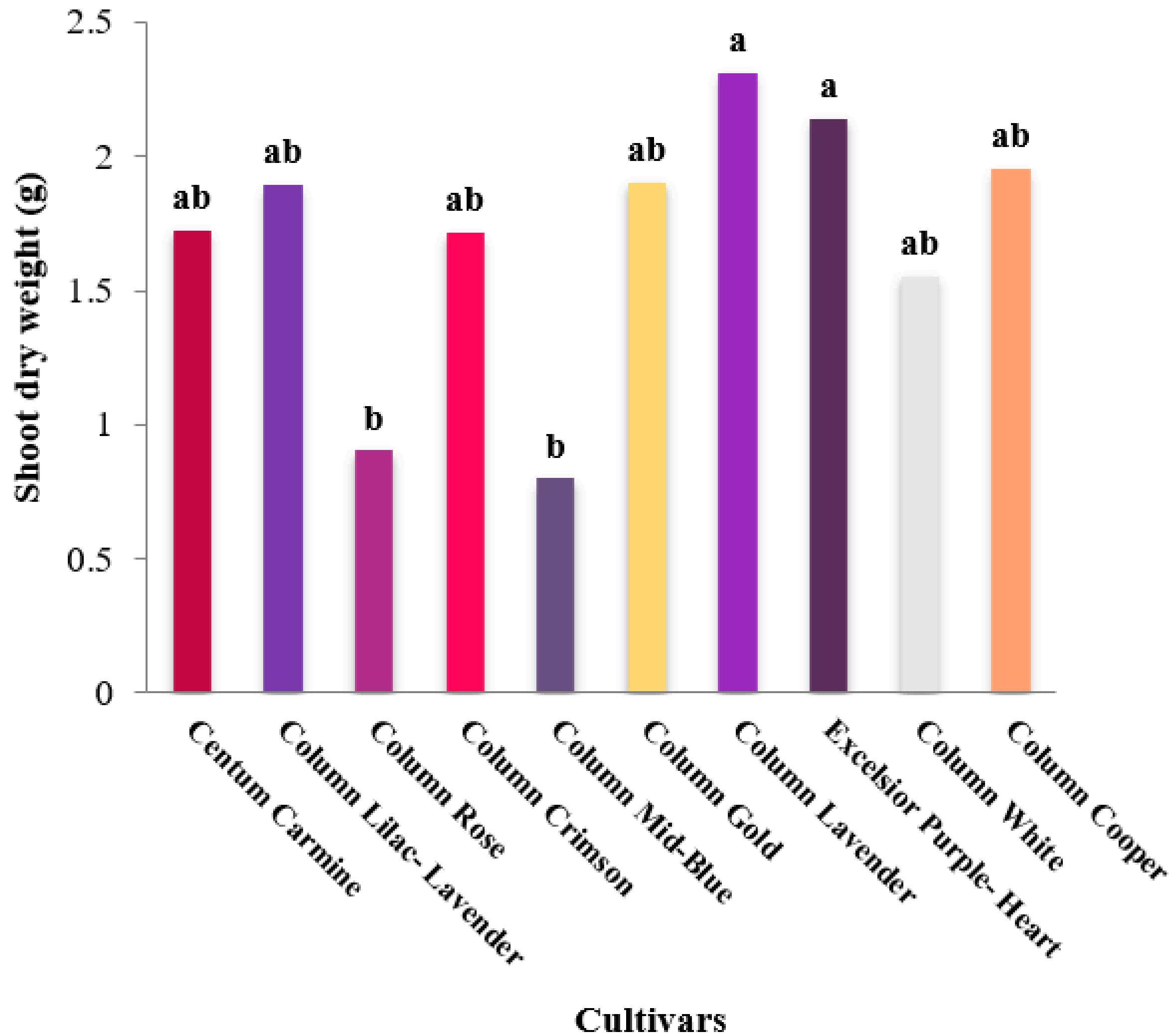

Figure 1

Shoot dry weight in different stock flower cultivars

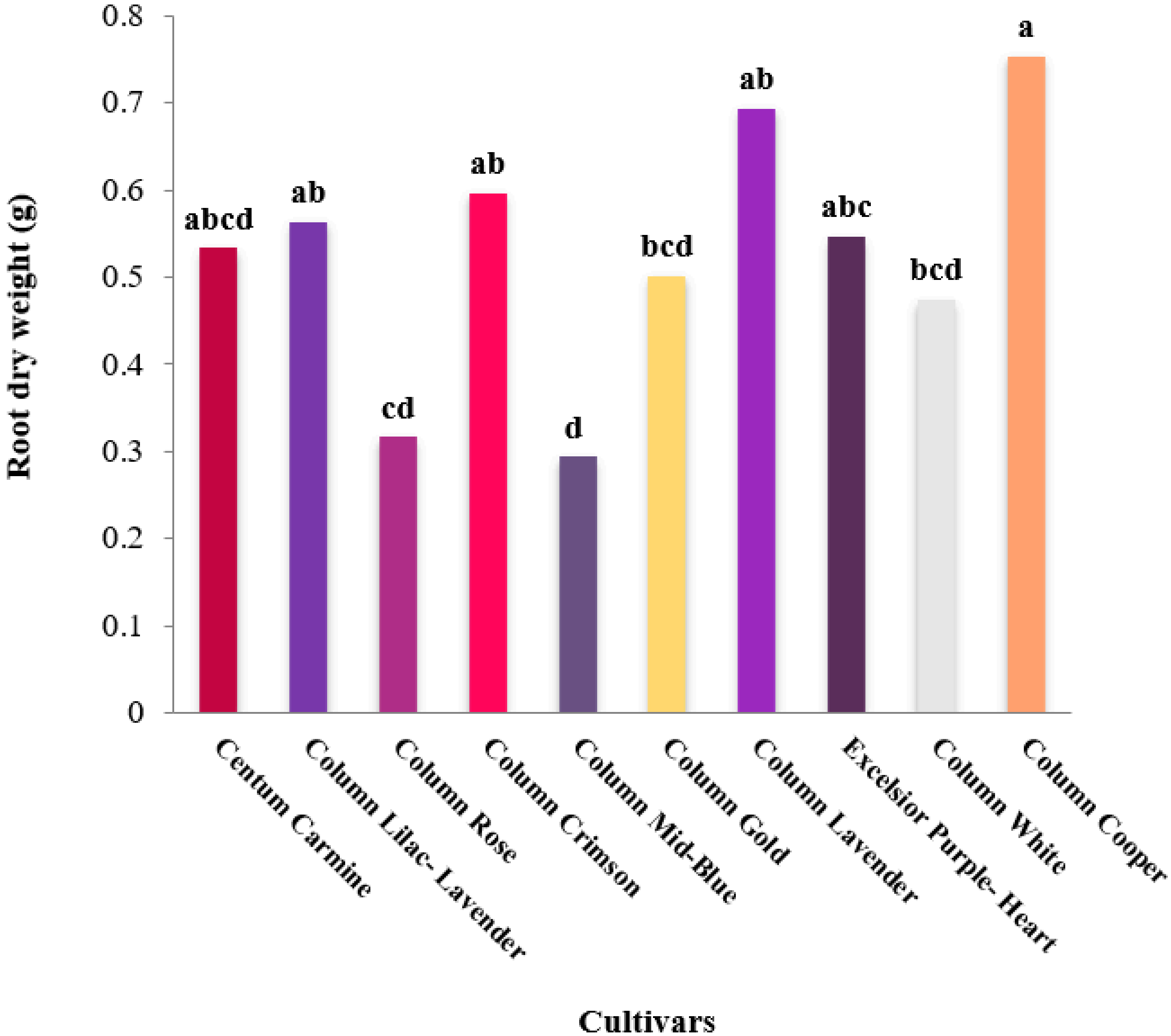

Figure 2

Root dry weight in different stock flower cultivars

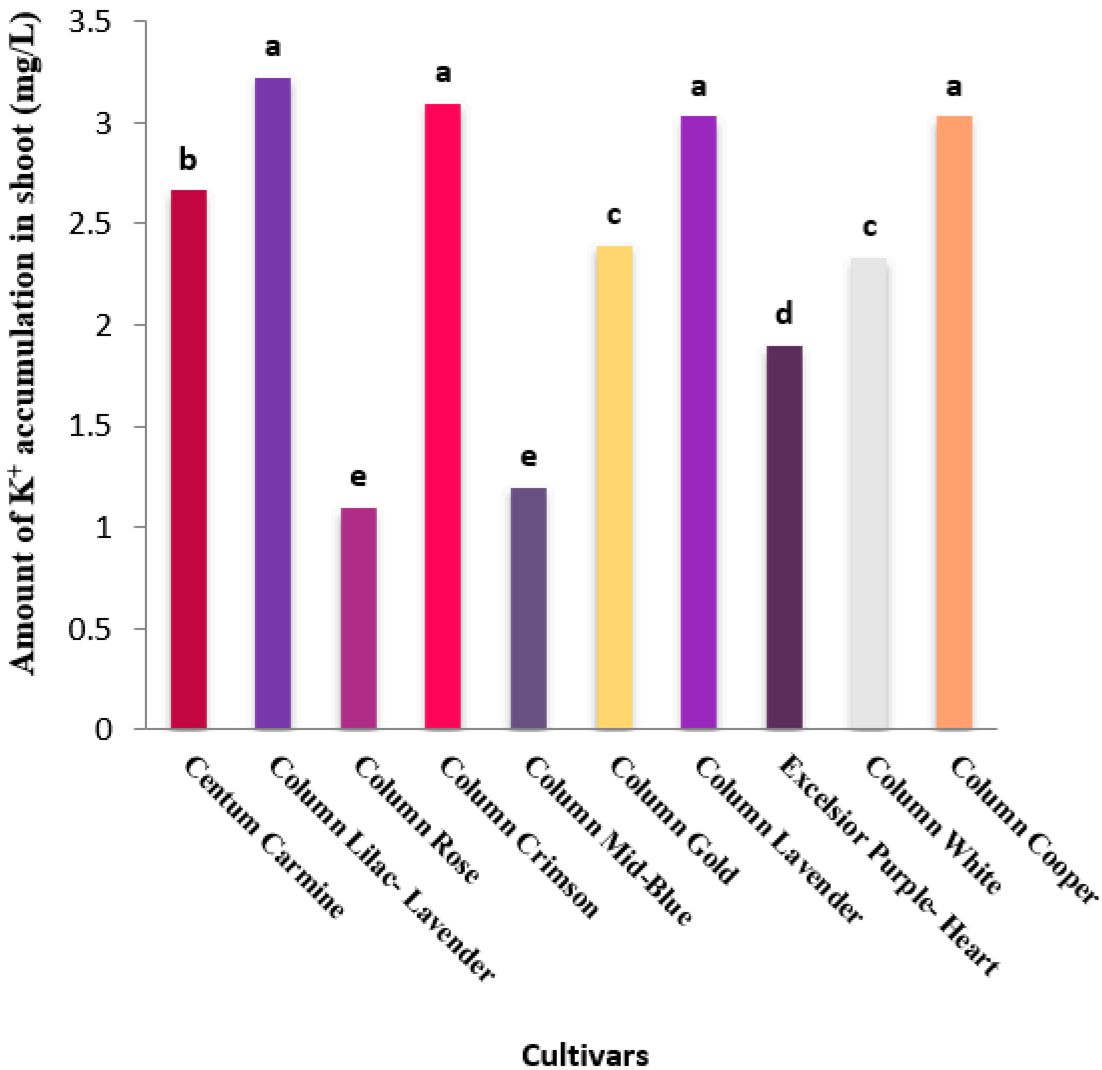

Figure 3

Amount of K^+ accumulation in shoots in different cultivars of stock flower

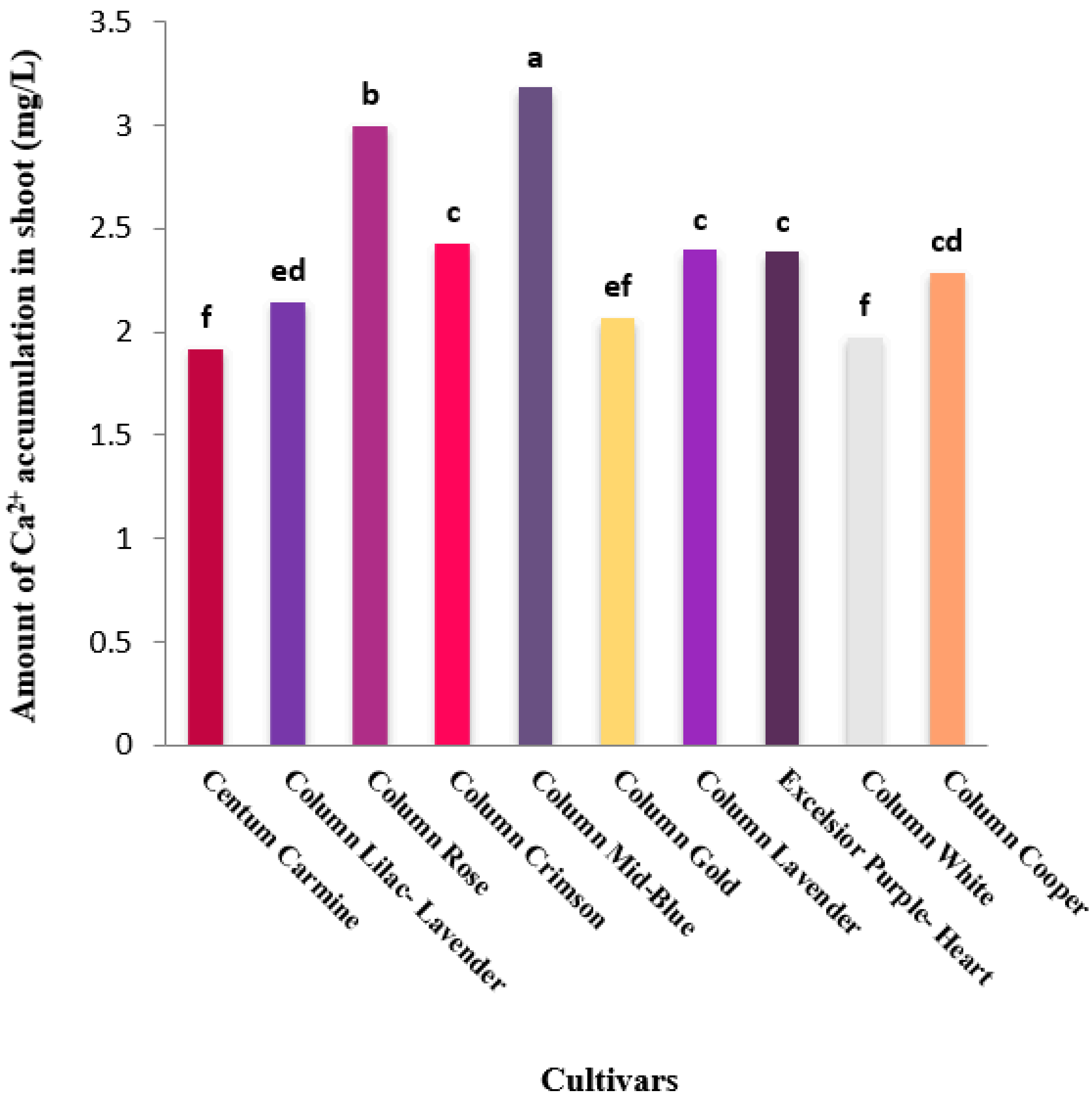

Figure 4

Amount of Ca^{2+} accumulation in the shoots of different stock flower cultivars

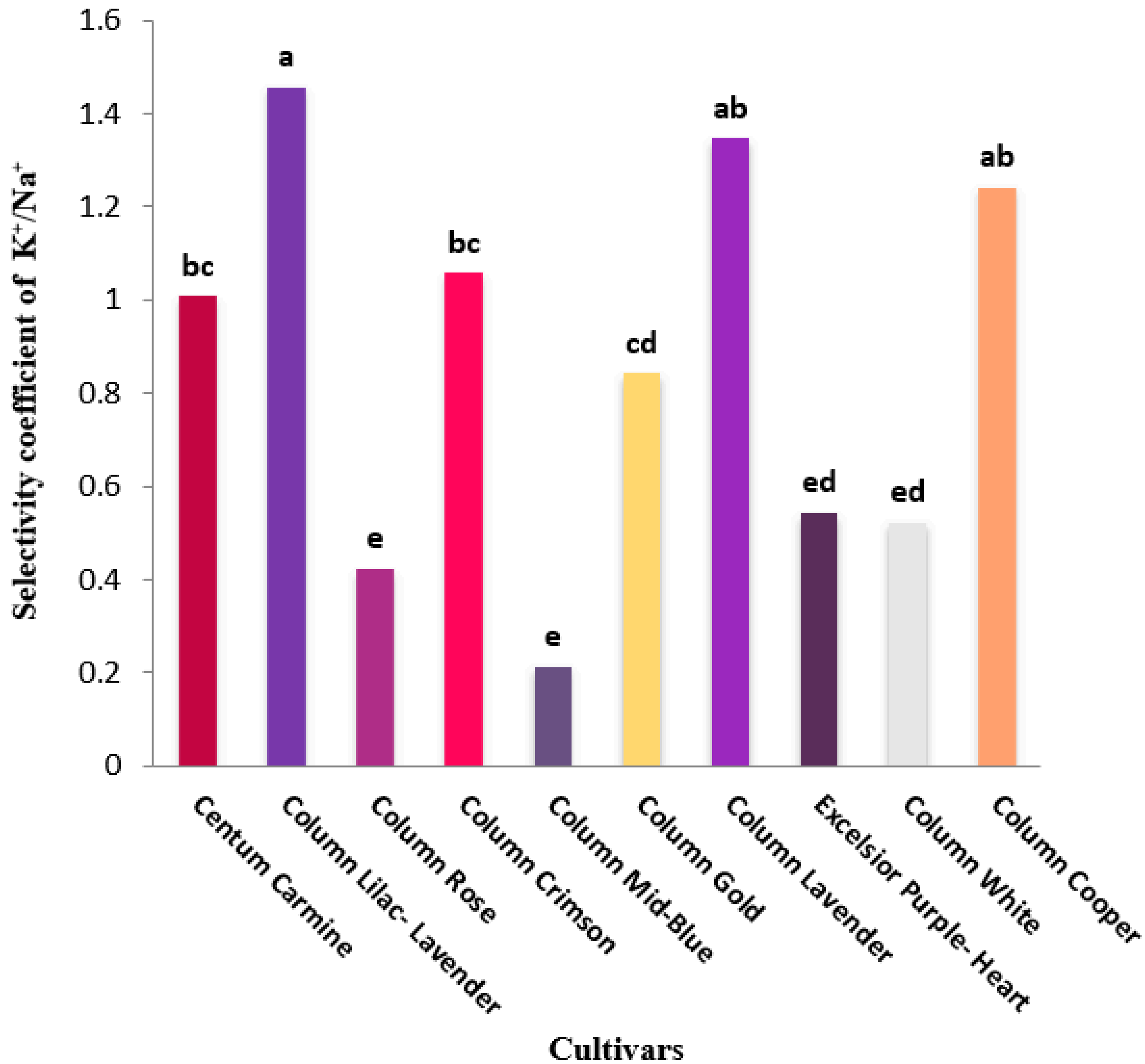

Figure 5

Selectivity coefficient of K/Na in the shoots of different cultivars of stock flower

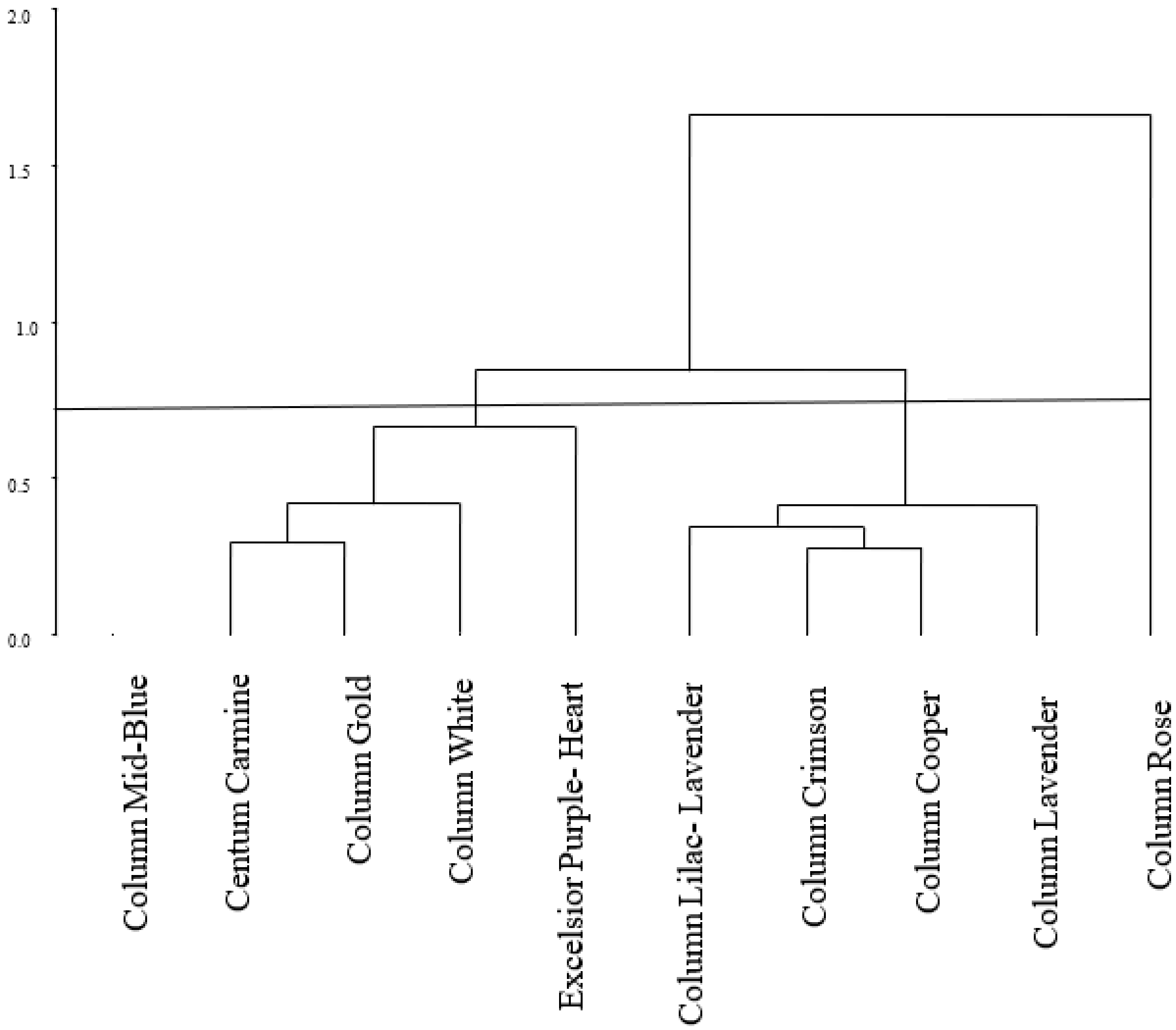

Figure 6

Cluster analysis of stock cultivars based on the studied traits under salinity stress

Supplementary Files

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- Table1.docx