Volume 14, Issue 1, Page 65-76, 2025 https://doi.org/10.46810/tdfd.1539065 Research Article

BİNGÖL UNIVERSITY

Türk Doğa ve Fen Dergisi
Turkish Journal of Nature and Science
www.dergipark.gov.tr/tdfd

Effect of K and Si Applications on Plant Development, Na and K Content and Some Antioxidant (SOD, CAT, APX) Activities of Wheat (Triticum aestivum L.) Plant Exposed to Salt Stress

Orhan İNİK[1*], Mehmet Ali Bozkurt[2]

[1] Bingol University, Faculty of Agriculture, Department of Soil Science and Plant Nutrition, Bingöl, Türkiye
[2] Yüzüncü Yıl University, Faculty of Agriculture, Department of Soil Science and Plant Nutrition, Van, Türkiye
Orhan İNİK ORCID No: 0000-0003-1473-1392
Mehmet Ali BOZKURT ORCID No: 0000-0003-3923-857X

**Corresponding author: oinik@bingol.edu.tr*

(Received: 29.08.2024, Accepted: 24.12.2024, Online Publication: 26.03.2025)

Keywords
Salt stress, Wheat, Silicon, Potassium, Antioxidant enzymes

Abstract: The problem of salinity in our soil is increasing day by day. This problem has become even more important with global climate change. Plants are the ones most affected by this problem. Salinity affects many metabolic activities in a very complex way by causing stress in plants. Reactive oxygen species, especially formed by salt stress, cause serious damage to plant cells. They use antioxidative enzymes such as catalase (CAT), ascorbate peroxidase (APX), and superoxide dismutase (SOD) to protect the plant from this stress. The aim of this research is to determine the effects of potassium and silicon applications on some antioxidant enzyme activities in wheat plant in salty conditions. In the experiment conducted under potted conditions, increasing doses of salt (0 and 100 mM NaCl), potassium (0, 150,300 ppm K) and silicon (0, 75, 150 ppm Si) were applied to the wheat plant. According to the results obtained, SOD activity increased with the 100 mM NaCl, 150 mg/kg K, 0 mg/kg Si, and 300 mg/kg Si applications. The effect of salt application on CAT activity was not found to be significant compared to control. APX activity was generally increased with the addition of salt. However, the increase in APX activity due to 100 mM NaCl administration decreased significantly with increasing silicon doses when 150 mg/kg potassium treatment was kept constant. This suggests that K and Si applications can be beneficial in alleviating salt stress.

Tuz Stresindeki Buğday (Triticum aestivum L.) Bitkisinin Bitki Gelişimi, Na ve K İçeriği ve Bazı Antioksidan (SOD, CAT, APX) Aktiviteleri Üzerine K ve Si Uygulamalarının Etkisi

Anahtar Kelimeler
Tuzluluk stresi, Buğday, Silikon, Potasyum, Antioksidan Enzimler

Öz: Topraklarımızdaki tuzluluk sorunu her geçen gün artmaktadır. Küresel iklim değişikliğiyle birlikte bu sorun daha da önemli hale gelmiştir. Bu sorundan en çok etkilenen ise bitkilerdir. Tuzluluk, bitkilerde strese neden olarak birçok metabolik aktiviteyi çok karmaşık bir şekilde etkilemektedir. Özellikle tuz stresinin oluşturduğu reaktif oksijen türleri, bitki hücrelerinde ciddi hasarlara neden olmaktadır. Bitkiler bu stresten korunmak için katalaz (CAT), askorbat peroksidaz (APX) ve süperoksit dismutaz (SOD) gibi antioksidan enzimleri kullanırlar. Bu araştırmanın amacı, tuzlu koşullarda buğday bitkisinde bazı antioksidan enzim aktiviteleri üzerine potasyum ve silisyum uygulamalarının etkilerini belirlemektir. Saksı koşullarında yapılan deneyde buğday bitkisine artan dozlarda tuz (0 ve 100 mM NaCl), potasyum (0, 150,300 ppm K) ve silisyum (0, 75, 150 ppm Si) uygulanmıştır. Elde edilen sonuçlara göre, SOD aktivitesi 100 mM NaCl, 150 mg/kg K, 0 mg/kg Si ve 300 mg/kg Si uygulamalarıyla artmıştır. Tuz uygulamasının CAT aktivitesi üzerine etkisi kontrole göre anlamlı bulunmamıştır. APX aktivitesi genel olarak tuz ilavesiyle artmıştır. Ancak, 100 mM NaCl uygulamasına bağlı APX aktivitesindeki artış, 150 mg/kg potasyum uygulaması sabit tutulduğunda artan silikon dozlarıyla anlamlı şekilde azalmıştır. Bu durum, K ve Si uygulamalarının tuz stresini hafifletmede faydalı olabileceğini düşündürmektedir.

1. INTRODUCTION

Plants show their best development under optimal conditions. Depending on the flexibility of normal metabolism, although they can continue to grow in the face of daily and seasonal changes, their constant or occasional exposure to an unexpected condition may result in diseases, damage or physiological changes that affect their development and survival [1]. The factors that create these unfavorable conditions are referred to as 'stress'. [2-5].

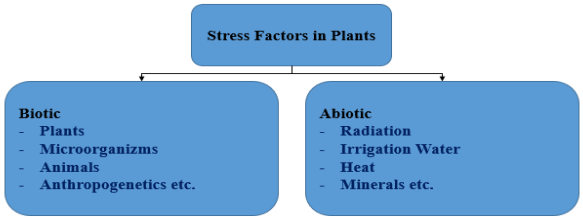


Figure 1. Stress Factors Affecting Plants [6-7]

Stress factors affecting plants are divided into two groups as biotic and abiotic (Figure 1). Of these factors, minerals from abiotic stresses are second with a rate of 20% after the stress factor that most affects usable areas caused by drought. The most important mineral that causes stress is salinity, which occurs in soils [7-8]. Salinity, together with the increasing human population, is one of the stress factors that significantly restrict the production of food products by jeopardizing efficient agriculture in our world. Excessive salinity in the soil affects the development of plants by causing structural, physiological and biochemical changes [9]. Both in the world and in our country, the yield is decreasing day by day in the agricultural lands and some areas are completely out of production due to excessive salinization. There is a salinity problem in 2-2.5 million ha of agricultural area in Turkey [10]. This problem is increasing due to chemical fertilizers used in agricultural areas, pesticides, intensive and unconscious irrigation in agricultural lands, destruction of the natural vegetation of a region and opening it to agricultural lands [11-14].

Salt stress inhibits plant growth and development by causing osmotic and ionic stress [15]. The increase in the amount of salt in the root rhizosphere first causes osmotic stress, which leads to a decrease in the amount of usable water, which is also called "physiological drought" [16]. The decrease in the amount of available water leads to reduced cell expansion and a slowdown in shoot development.

During the ion stress phase that occurs in the continuation of osmotic stress, nutrient deficiency or nutrient imbalance occurs in plants when Na and Cl ions increasing in the environment cause an antagonistic effect with the necessary nutrients such as K^+, Ca^{+2} and NO_3^- [17-18]. In particular, this effect significantly inhibits the plant's potassium (K^+) uptake [9,19].
One of the reasons for the stress observed in plants in salty conditions is the inhibition of the uptake, transportation and use of nutrients by plants. In one study, it was determined that the growth decline observed in the barley plant grown in a salty environment was largely due to salt-induced Mn deficiency [20]. In addition, P uptake and movement in soil-grown plants decreased as salt concentration increased [21].

Many studies have been done and are still being done to eliminate the damage of salinity in the soil. In addition to growing salinity-resistant halophyte plants, it can prevent the damage caused by salt stress in the soil in some applications. For example, it is believed that the salt tolerance of plants is related to the limitation in Na intake and that K plays an important role in this limitation. In a study conducted by Litifi et al. (1992) [22], it was seen that potassium regulation, increased K/Na ratio in the plant and salt tolerance increased. It has been determined that salt-tolerant varieties absorb more K than sensitive varieties, and that K plays an important role in drought and salt stress tolerance [23]. Under saline conditions, increasing the turgor pressure in plant tissues, which is a natural defense mechanism, and protecting turgor come to the forefront. Likewise, studies conducted to date have shown that silicone (Si) has beneficial effects on healthy plant growth and development. Silicon has been proven to increase plant tolerance to salt stress and drought [24-25]. It is stated that if Si is added to saline soils, silicon reacts with Na and significantly reduces Na uptake and transport, as well as increases K uptake and reduces the Na/K ratio in the plant. In addition, many studies report that Si reduces oxidative stress in plants [26-34].

Although there are various studies in which K and Si applications are discussed individually in the prevention of salt stress, there are almost no studies investigating K×Si interaction. The aim of this study is to determine the effect of K×Si interaction on some antioxidant enzyme activity in the plant in order to eliminate the damages caused by salinity in the wheat plant.

2. MATERIAL AND METHOD

The trial was carried out in pots under the conditions of the climate room at Yüzüncü Yıl University Faculty of Agriculture. Alparslan Wheat variety plant was used as the plant in the research. The experiment was carried out in randomized plots according to the factorial design with 4 replications. Plants are grown in pots that can weigh up to 1 kg. In the study, a total of 144 pots consisting of two pots were used each repeatedly. The climate room is set to be 14 hours day, 10 hours at night, relative humidity 60%, temperature 25-27 ^{0}C.

The mortar used in the trial was prepared by mixing soil, sand and peat in a ratio of 2: 2:1. In the experiment, 50 ppm P was given to all pots in the form of P (triple superphosphate) as basic fertilization (Figure 2.a).

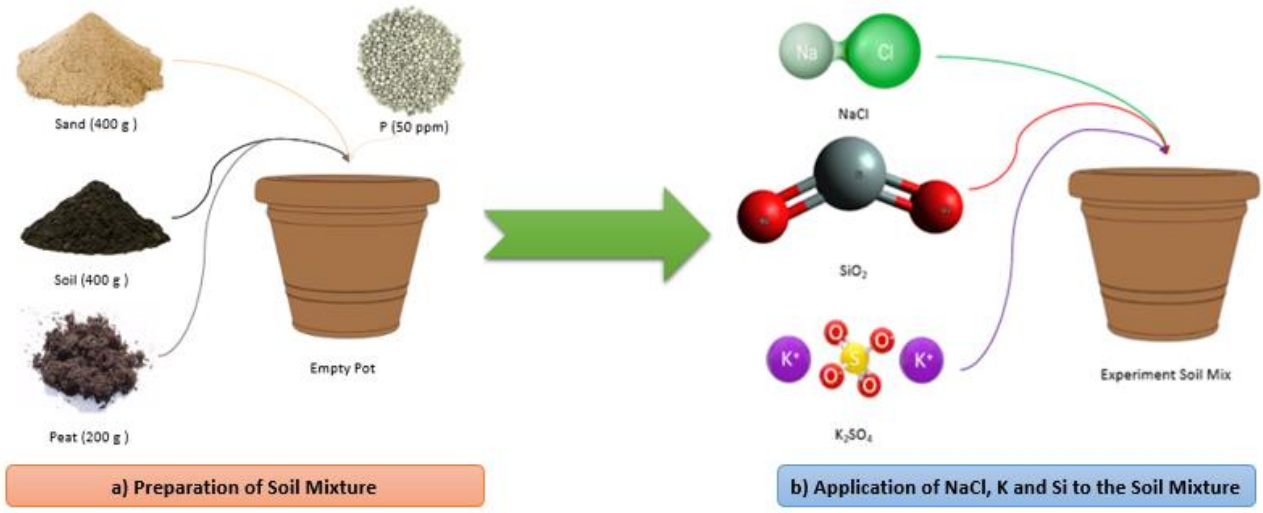


Figure 2: Preparation of soil mixture and application of NaCl, K and Si before planting seeds in pots

Six seeds are planted in a 1 kg plastic pot. After germination is completed three plants are left in each pot. After six weeks of development, the plants were harvested by cutting from the soil surface. Samples for enzyme analysis were taken from the youngest leaves of the plant before harvest and stored in a -82 ^{0}C freezer.

Salt was applied in two levels (0 and 100 mM) as NaCI. Potassium was applied at 3 levels (0, 150, 300 ppm K) with control as K_2SO_4. Silicon application was applied at 3 levels (0, 75, 150 ppm Si) with control as SiO_2 (Figure 2.b; Table 1).

Table 1. Experiment pattern topics

$NaCl_0K_0Si_0$ (kontrol)	10- $NaCl_1K_0Si_0$ (100 mM NaCl)
$NaCl_0K_0Si_1$ (75 ppm Si)	11- $NaCl_1K_0Si_1$ (100 mM NaCl + 75 ppm Si)
$NaCl_0K_0Si_2$ (150 ppm Si)	12- $NaCl_1K_0Si_2$ (100 mM NaCl + 150 ppm Si)
$NaCl_0K_1Si_0$ (150 ppm K)	13- $NaCl_1K_1Si_0$ (100 mM NaCl +150 ppm K)
$NaCl_0K_1Si_1$ (150 ppm K + 75 ppm Si)	14- $NaCl_1K_1Si_1$ (100 mM NaCl +150 ppm K + 75 ppm Si)
$NaCl_0K_1Si_2$ (150 ppm K + 150 ppm Si)	15- $NaCl_1K_1Si_2$ (100 mM NaCl +150 ppm K + 150 ppm Si)
$NaCl_0K_2Si_0$ (300 ppm K)	16- $NaCl_1K_2Si_0$ (100 mM NaCl +300 ppm K)
$NaCl_0K_2Si_1$ (300 ppm K + 75 ppm Si)	17- $NaCl_1K_2Si_1$ (100 mM NaCl +300 ppm K + 75 ppm Si)
$NaCl_0K_2Si_2$ (300 ppm K + 150 ppm Si)	18- $NaCl_1K_2Si_2$ (100 mM NaCl +300 ppm K + 150 ppm Si)

As stated by Jakson (1958), the pH values in the experimental soil were determined by diluting the pH values with pure water in a ratio of 1/2.5 [35]. Lime (%) was detected using Scheibler's calcimeter, as noted by Hızalan and Ünal (1966) [36]. EC (dS/m), using the 1/2.5 method was determined by a handheld electrical constantimeter [37]. Organic matter was determined according to the modified Walkley-Black method [38]. Total nitrogen was determined according to the Kjeldahl method [37] and the available phosphorus was determined according to the Sodium bicarbonate method [39]. Exchangeable potassium (C), calcium (Ca), magnesium (Mg) and sodium (Na) were determined in the Atomic Absorption Spectrophotometer by shaking with 1 N Ammonium acetate according to Thomas (1982) [40]. Available microelements (Fe, Mn, Zn and Cu) in soil samples were determined by reading the prepared solutions with the 0.05 M DTPA method, pH adjusted to 7.3, on an atomic absorption spectrophotometer (AAS). [37]. Some analyses of the trial soil are given in Table 2.

Table 2. Some features of the experiment soil

Properties	**Experimental Soil**
pH (1/2.5)	8.26
EC (dS/m)	0.80
$CaCO_3$ (%)	13.6
Organic Madder (%)	6.27
Total N (%)	0.15
Available P (mg/kg)	13.3
Exchangeable Cations (ppm)	
K	732
Ca	23709
Mg	749
Na	459
Extractable with DTPA (ppm)	
Fe	19.3
Zn	0.46
Cu	0.47
Mn	5.16

Catalase (CAT) activity in plant leaves was determined by measuring the change in absorbance due to H2O2 decrease at a wavelength of 240 nm [41]. Superoxide Dismutase (SOD) Activity was determined by inhibition of nitroblue tetrazolium (NBT) at a wavelength of 560 nm [42]. Ascorbate Peroxidase (APX) Activity was measured

by H_2O_2 reduction due to ascorbic acid [43]. After the statistical analysis of the obtained findings was determined by SPSS package program, the significance of the difference between the averages of the applications was made with Duncan multiple comparison test ($P<0.05$) [44].

3. RESULTS

3.1. Plant Productivity

Some of the images of the experiment are in Figure 3. The effect of salt stress on plant development is clearly visible (Figure 3a).

$NaCl_0K_2Si_1$ application increased plant growth compared to $NaCl_0K_2Si_2$ application (figure 3.b.).

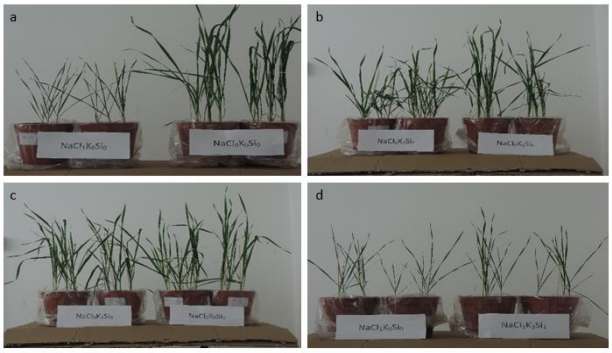


Figure 3. Some images of plant development.

Variance analysis results regarding the effects of NaCl, K, Si and the double and triple interactions of these applications on plant fresh weight, plant dry weight and plant height are shown in Table 3.

Table 3. Variance analysis results regarding the effects of NaCl, K and Si applications on plant fresh weight, dry weight and plant height.

Variation Source	S.D.	Fresh Weight		Dry Weight		Plant Height	
		Mean of Squares	F Value	Mean of Squares	F Value	Mean of Squares	F Value
NaCl	1	1445.3	6512.2***	33.6	3304.7***	4403.6	1356.2***
K	2	1.303	5.872**	0.000	0.03ns.	19.55	6.02**
Si	2	0.554	2.495ns.	0.040	3.94*	8.46	2.60ns.
NaClxK	2	0.564	2.543ns.	0.060	5.93**	29.01	8.94***
NaClxSi	2	0.097	0.439ns.	0.003	0.25ns.	0.25	0.08ns.
KxSi	4	0.232	1.047ns.	0.010	0.95ns.	2.73	0.84ns.
NaClxKxSi	4	0.153	0.687ns.	0.041	4.01**	5.00	1.54ns.
Error	54	0.222		0.010		3.25	
Total	71						

*: Important at p<0.05, **: Important at p<0.01 , ***: Important at p<0.001, ns: non significant

Variance analysis results regarding the effects of NaCl, K, Si and the double and triple interactions of these applications on plant fresh weight, plant dry weight and plant height are shown in Table 3. While the effect of NaCl ($P<0.001$) and K ($P<0.01$) applications on plant fresh weight was found to be statistically significant, the effect of other applications was not found to be significant. The fresh plant weight, which was determined as 10.9 grams in the NaCl0Si0K0 application, decreased significantly to 1.58 grams in the $NaCl_1Si_0K_0$ application. Increasing potassium doses in both salt-free ($NaCl_0$) and salty ($NaCl_1$) conditions caused a decrease in plant fresh weight. The effect of Si doses on plant fresh weight was not found to be significant (Table 4.).

The effect of NaCl ($P<0.001$), Si ($P<0.05$), NaClxK ($P<0.01$) and NaClxKxSi ($P<0.01$) applications on plant dry weight was found to be significant (Table 3.). The dry weight of the plant, which was determined as 1.59 grams in the $NaCl_0Si_0K_0$ application, decreased significantly to 0.28 grams in the $NaCl_1Si_0K_0$ application. At constant potassium level (K_1) under salt-free conditions, Si1 application significantly increased plant dry weight compared to other silicon doses.

Table 4. Effect of NaCl, K and Si applications on plant fresh weight (g/pot)

Si Doses (mg/kg)	K Doses (mg/kg)	$NaCl_0$	$NaCl_1$
		Average	Average
Si_0	K_0	A 10.91 a	A 1.580 a *
	K_1 (150)	A 10.05 b	A 1.100 b *
	K_2 (300)	A 10.26 b	A 1.248 b *
Si_1 (75)	K_0	B 10.40 a	B 1.230 a *
	K_1 (150)	A 10.35 a	A 1.110 a *
	K_2 (300)	A 9.79 a	A 1.392 a *
Si_2 (150)	K_0	B 10.53 a	B 1.198 ab *
	K_1 (150)	A 9.85 b	A 1.215 a *
	K_2 (300)	A 9.56 b	B 0.980 b *

a,b, c: The difference between potassium levels that take on different lowercase letters at the same silicon and NaCl level is important ($p<0.05$).
A, B,C: The difference between different capitalized silicon levels at the same potassium and NaCl levels is important ($p<0.05$).
*: The difference between NaCl1 administration and control (NaCl0) is significant ($p<0.05$).

When the K_0 dose was kept constant under saline conditions, Si_1 application significantly increased the plant dry weight compared to other silicon doses. Similarly, when $NaCl_1K_1$ application was kept constant, plant dry weight increased significantly with increasing silicon doses (Table 5.). These results show that the NaClxKxSi interaction is important on plant dry weight.

Table 5. Effect of NaCl, K and Si applications on plant dry weight (g/pot)

Si Doses (mg/kg)	K Doses (mg/kg)	$NaCl_0$	$NaCl_1$
		Average	Average
Si_0	K_0	A 1.59 a	B 0.282 a *
	K_1 (150)	B 1.55 a	B 0.227 b *
	K_2 (300)	A 1.66 a	A 0.240 b *
Si_1 (75)	K_0	A 1.54 b	A 0.452 a *
	K_1 (150)	A 1.83 a	A 0.260 b *
	K_2 (300)	A 1.71 ab	A 0.235 b *
Si_2 (150)	K_0	A 1.63 a	B 0.262 a *
	K_1 (150)	B 1.60 a	A 0.267 a *
	K_2 (300)	A 1.69 a	A 0.252 a *

a,b, c: The difference between potassium levels that take on different lowercase letters at the same silicon and NaCl level is important ($p<0.05$).
A, B,C: The difference between different capitalized silicon levels at the same potassium and NaCl levels is important ($p<0.05$).
*: The difference between NaCl1 administration and control (NaCl0) is significant ($p<0.05$).

The effects of NaCl ($P<0.001$), K ($P<0.01$) and NaClxK ($P<0.001$) applications on plant height were found to be statistically significant (Table 3.). The plant height, which was determined as 40.6 cm in the $NaCl_0Si_0K_0$ application, decreased significantly to 25.2 cm in the $NaCl_1Si_0K_0$ application. In salt-free conditions, when NaCl and Si doses were kept constant, increasing potassium doses caused plant height to decrease.

The effect of increasing potassium doses at constant silicon level under saline conditions on plant height was not found to be significant. The effect of silicon doses at constant potassium level on plant height was found to be insignificant in both salt-free and salty conditions (Table 6.).

Table 6. Effect of NaCl, K and Si applications on plant height (cm)

Si Doses (mg/kg)	K Doses (mg/kg)	$NaCl_0$	$NaCl_1$
		Average	Average
Si_0	K_0	A 40.625 a	A 25.225 a *
	K_1 (150)	B 39.625 ab	A 22.475 a *
	K_2 (300)	A 37.850 b	A 22.775 a *
Si_1 (75)	K_0	A 40.650 a	A 22.650 a *
	K_1 (150)	A 39.875 a	A 23.325 a *
	K_2 (300)	B 36.850 b	A 24.900 a *
Si_2 (150)	K_0	A 40.075 a	A 22.300 a *
	K_1 (150)	A 39.050 a	A 22.475 a *
	K_2 (300)	A 35.325 b	A 23.025 a *

a,b, c: The difference between potassium levels that take on different lowercase letters at the same silicon and NaCl level is important ($p<0.05$).
A, B,C: The difference between different capitalized silicon levels at the same potassium and NaCl levels is important ($p<0.05$).
*: The difference between NaCl1 administration and control (NaCl0) is significant ($p<0.05$).

3.2. Na and K Content in Plants

The effects of NaCl, K, Si and the double and triple interactions of these applications on the potassium (K) and sodium (Na) content in the plant can be seen in Table 7.

Table 7. Variance analysis results of the effects of NaCl, K and Si applications on K and Na content in the plant

Variation Source	S.D.	K		Na	
		Mean of Squares	F Value	Mean of Squares	F Value
NaCl	1	17.0	111.3 ***	12.69	1815.8***
K	2	0.695	9.02**	0.85	120.3 ***
Si	2	0.177	6.61 ns.	0.42	60.14***
NaClxK	2	0.436	26.3 *	1.07	152.9 ***
NaClxSi	2	0.001	3.21ns.	0.40	56.47***
KxSi	4	0.518	26.0 **	0.62	89.20***
NaClxKxSi	4	0.271	4.21*	0.62	88.33***
Error	54	0.093		0.01	
Total	71				

*: Important at $p<0.05$, **: Important at $p<0.01$, ***: Important at $p<0.001$, ns: non significan

The effect of NaCl ($P<0.001$), K ($P<0.01$), NaClxK ($P<0.05$), KxSi ($P<0.01$) and NaClxKxSi ($P<0.05$) applications on the potassium content in the plant is statistically significant. was found (Table 7.). Under salt-free conditions, potassium doses at a fixed dose of Si_0 caused a decrease in plant potassium content compared to the control, while increasing potassium doses, keeping the Si_2 dose constant, significantly increased the potassium content in the plant compared to the control. In saline conditions, when the silicon dose was generally kept

constant, increasing potassium doses increased the plant potassium content compared to the control.
This increase was found to be statistically significant at Si_0 and Si_1 doses compared to the control (K_0) (Table 8.). The effect of all variation sources on sodium in the plant was found to be significant (P<0.001) (Table 7.). Increasing potassium applications at a constant silicon dose under salt-free conditions significantly reduced plant sodium content. Application of Si_1 at a fixed K_0 dose significantly increased the sodium content in the plant compared to other silicon doses. Increasing potassium applications at a fixed silicon dose under saline conditions generally significantly increased the sodium content of the plant.

Table 8. Effect of NaCl, K and Si applications on potassium content in plants (%)

Si Doses (mg/kg)	K Doses (mg/kg)	$NaCl_0$ Average	$NaCl_1$ Average
Si_0	K_0	A 2.38 a	A 1.15 b *
	K_1 (150)	A 2.12 b	B 0.79 c *
	K_2 (300)	A 2.18 b	A 1.80 a *
Si_1 (75)	K_0	A 2.17 a	A 0.80 b *
	K_1 (150)	A 1.95 a	B 0.86 b *
	K_2 (300)	A 2.05 a	B 1.56 a *
Si_2 (150)	K_0	B 1.75 b	A 0.91 a *
	K_1 (150)	A 2.41 a	A 1.50 a *
	K_2 (300)	A 2.32 a	C 1.20 a *

a,b, c: The difference between potassium levels that take on different lowercase letters at the same silicon and NaCl level is important (p<0.05).
A, B,C: The difference between different capitalized silicon levels at the same potassium and NaCl levels is important (p<0.05).
*: The difference between NaCl1 administration and control (NaCl0) is significant (p<0.05).

When the K_0 dose was kept constant, the highest sodium content was determined in the Si_1 dose, while when the K_2 dose was kept constant, the highest sodium content was obtained with the Si_2 application (Table 9.).

Table 9. Effect of NaCl, K and Si applications on sodium content in plants (%)

Si Doses (mg/kg)	K Doses (mg/kg)	$NaCl_0$ Average	$NaCl_1$ Average
Si_0	K_0	B 0.088 a	B 0.642 a *
	K_1 (150)	A 0.087 a	A 0.783 a *
	K_2 (300)	A 0.053 b	B 0.749 a *
Si_1 (75)	K_0	A 0.120 a	A 0.827 b *
	K_1 (150)	A 0.089 b	B 0.661 c *
	K_2 (300)	A 0.055 c	B 0.992 a *
Si_2 (150)	K_0	B 0.101 a	B 0.581 b *
	K_1 (150)	A 0.100 a	B 0.666 b *
	K_2 (300)	A 0.061 b	A 0.407 a *

a,b, c: The difference between potassium levels that take on different lowercase letters at the same silicon and NaCl level is important (p<0.05).
A, B,C: The difference between different capitalized silicon levels at the same potassium and NaCl levels is important (p<0.05).
*: The difference between NaCl1 administration and control (NaCl0) is significant (p<0.05).

3.3. Antioxidant Enzyme Activity

The effect of NaCl, K, Si and the double and triple interactions of these applications on SOD, CAT and APX enzymes in the plant is seen in Table 3.

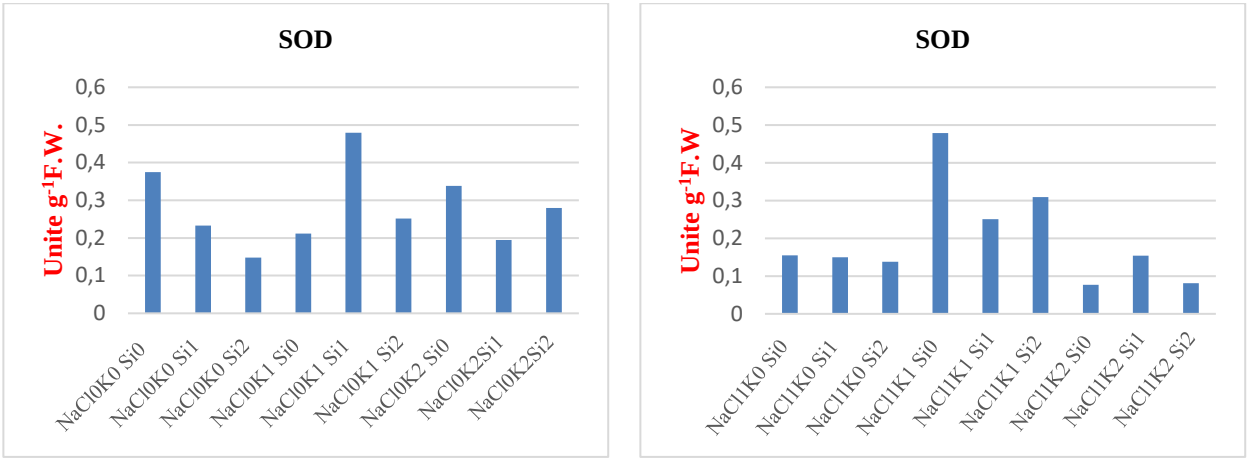
The effect of NaCl (P<0.01), K (P<0.001), Si (P<0.01), NaClxK (P<0.01) and NaClxKxSi (P<0.001) applications on SOD enzyme in the plant was found to be statistically significant (Table 10).

Table 10. Variance analysis results of the effect of NaCl, K and Si applications on SOD, CAT and APX enzyme activations in the plant

Variation Source	S.D.	SOD		CAT		APX	
		Mean of Squares	F Value	Mean of Squares	F Value	Mean of Squares	F Value
NaCl Doses	1	0.09	12.78^{**}	0.001	6.40^{*}	4.84	73.51^{***}
K Doses	2	0.17	22.91^{***}	0.003	24.15	0.81	12.31^{***}
Si Doses	2	0.04	$5.87^{**.}$	0.001	6.90^{**}	0.62	9.39^{***}
NaClxK	2	0.05	6.99^{**}	0.001	11.14^{***}	0.61	9.28^{***}
NaClxSi	2	0.01	$1.73^{ns.}$	1.60	$0.13^{ns.}$	0.50	7.63^{**}
KxSi	4	0.01	$0.75^{ns.}$	0.00	2.84^{*}	0.93	14.12^{***}
NaClxKxSi	4	0.08	10.90^{***}	9.30	$0.80^{ns.}$	0.38	5.76^{**}
Error	54	0.01		0.00		0.07	
Total	71						

*: Important at p<0.05, **: Important at p<0.01 , ***: Important at p<0.001, ns: non significant

When the silicon doses are kept constant separately in salty conditions, the difference created by the K_1 dose in potassium applications is found to be statistically significantly higher than other doses. The highest SOD activity at a constant dose of K_1 in saline conditions was determined at the dose of no silicon (Si_0). When the K_2 dose was kept constant, the difference between the silicon doses and the Si_1 dose was found to be significant and high (Figure 4 and Table 11).



a b

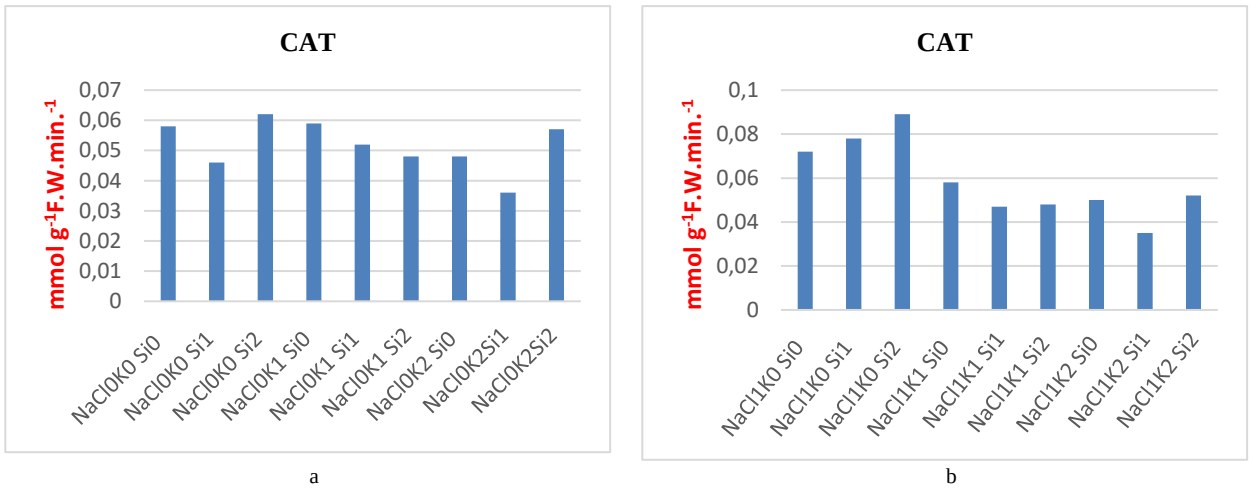
Figure 4. SOD enzyme activity in the plant; a. Effect of unsalted KxSi interactions, b. Effect of 100 mM NaCl saline KxSi interactions

Table 11. Effect of NaCl, K and Si applications on superoxide dismutase (SOD) enzyme activity in the plant (Unite g-1F.W.)

Si Doses (mg/kg)	K Doses (mg/kg)	$NaCl_0$ Average	$NaCl_1$ Average
Si_0	K_0	A 0.375 a	A0.155 b *
	K_1 (150)	B 0.212 a	A 0.479 a *
	K_2 (300)	A 0.338 a	B 0.077 c *
Si_1 (75)	K_0	AB 0.233 b	A 0.150 b *
	K_1 (150)	A 0.480 a	B 0.251 a *
	K_2 (300)	B 0.195 b	A 0.154 b
Si_2 (150)	K_0	B 0.148 a	A 0.138 b
	K_1 (150)	B 0.252 a	B 0.309 a
	K_2 (300)	B 0.208 a	B 0.081 b *

a,b, c: The difference between potassium levels that take on different lowercase letters at the same silicon and NaCl level is important ($p<0.05$).
A, B,C: The difference between different capitalized silicon levels at the same potassium and NaCl levels is important ($p<0.05$).
*: The difference between NaCl1 administration and control (NaCl0) is significant ($p<0.05$).

The effect of NaCl ($P<0.05$), K ($P<0.01$), Si ($P<0.01$), NaClxK ($P<0.001$) and KxSi ($P<0.05$) applications on CAT enzyme in the plant was found to be important (Table 10). While salt application did not affect CAT activity in general, the addition of salt to only the K_0-Si_1 application group significantly increased CAT activity. The CAT activity obtained by K_2-Si_1 application under salt-free conditions was found to be statistically significantly different from other applications.



a b

Figure 5. CAT enzyme activity in the plant; a. Effect of unsalted KxSi interactions, b. Effect of 100 mM NaCl saline KxSi interactions

When the level of silicon (Si_0, Si_1 and Si_2) was kept constant under saline conditions, increased potassium doses significantly reduced CAT Catalase activity decreased significantly from 0.078 mmol g^{-1}F.W.$min.^{-1}$ in the K_0-Si_1 interaction group to 0.035 mmol g^{-1}F.W.$min.^{-1}$ in the K_2-Si_1 interaction group. Increased doses of silicon in pots where potassium was applied under the same conditions significantly increased CAT activity (Figure 5 and Table 12).

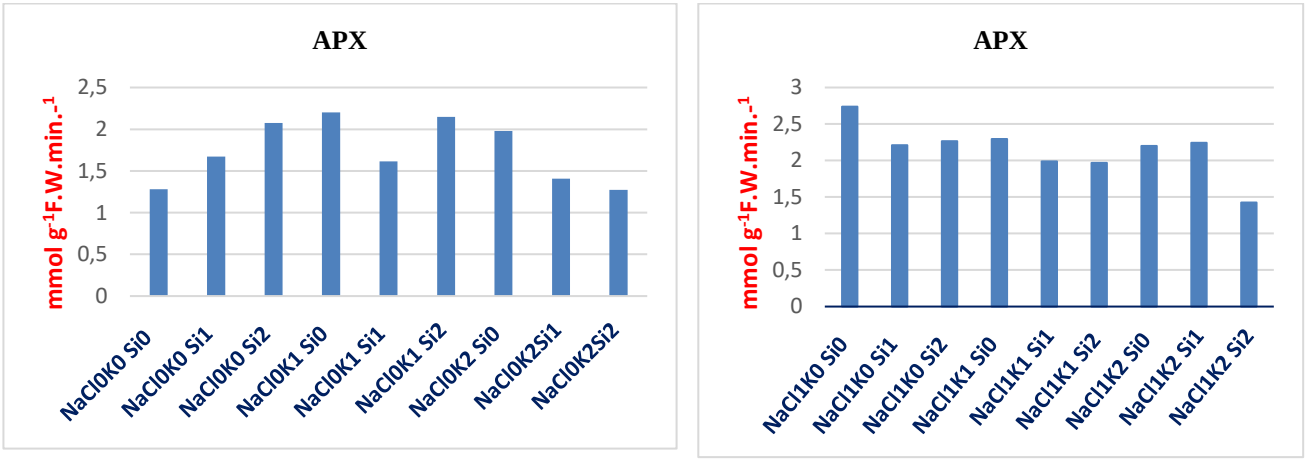
The effect of all variation sources on APX enzyme in the plant was found to be significant (Table 10). When the Si_0 dose from silicon applications was kept constant under unsalted ($NaCl_0$) conditions, increased potassium doses

increased APX activity, while increased potassium levels at constant Si1 and Si_2 doses led to a decrease in APX activity. The difference of Si_2 dose in silicon applications when K_0 dose was kept constant from potassium applications, the difference caused by reducing Si1 activity when K_1 dose was kept constant, and the increase in activity obtained with Si_0 dose when K_2 dose was kept constant were found to be statistically important.

Table 12. Effect of NaCl, K and Si applications on catalase (CAT) enzyme activity in the plant (mmol g^{-1}F.W.$min.^{-1}$)

Si Doses (mg/kg)	K Doses (mg/kg)	$NaCl_0$ Average	$NaCl_1$ Average
Si_0	K_0	A 0.058 a	B 0.072 a
	K_1 (150)	A 0.059 a	A 0.058 ab
	K_2 (300)	A 0.048 a	A 0.050 b
Si_1 (75)	K_0	A 0.046 a	AB 0.078 a *
	K_1 (150)	A 0.052 a	A 0.047 b
	K_2 (300)	A 0.037 b	B 0.035 b
Si_2 (150)	K_0	A 0.063 a	A 0.089 a
	K_1 (150)	A 0.049 a	A 0.048 b
	K_2 (300)	A 0.057 a	A 0.052 b

a,b, c: The difference between potassium levels that take on different lowercase letters at the same silicon and NaCl level is important ($p<0.05$).
A, B,C: The difference between different capitalized silicon levels at the same potassium and NaCl levels is important ($p<0.05$).
*: The difference between NaCl1 administration and control (NaCl0) is significant ($p<0.05$).



a b

Figure 6. APX enzyme activity in the plant; a. Effect of unsalted KxSi interactions, b. Effect of 100 mM NaCl saline KxSi interactions

Considering the effects of these applications in saline conditions, the difference between silicon applications was found to be insignificant when the K_0 dose was kept constant. The difference of Si0 dose when K_1 dose was kept constant and the difference of all silicon doses when K_2 dose was kept constant were found to be statistically significant and the lowest activity was obtained with Si_2 administration (1.425 mmol g^{-1} F.W. min^{-1}). In general, when the silicon dose was kept constant, the increased potassium level caused a decrease in APX activity (Figure 6 and Table 13).

Table 13. Effect of NaCl, K and Si applications on ascorbate peroxidase (APX) enzyme activity in the plant (mmol g^{-1} F.W. min^{-1})

Si Doses (mg/kg)	K Doses (mg/kg)	$NaCl_0$ Average	$NaCl_1$ Average
Si_0	K_0	B 1.282 b	A 2.738 a *
	K_1 (150)	A 2.200 a	A 2.695 a *
	K_2 (300)	A 1.980 a	B 2.200 b
Si_1 (75)	K_0	B 1.673 a	A 2.210 a
	K_1 (150)	B 1.615 ab	B 1.988 a *
	K_2 (300)	B 1.408 b	A 2.433 a *
Si_2 (150)	K_0	A 2.075 a	A 2.665 a *
	K_1 (150)	A 2.148 a	B 1.968 b
	K_2 (300)	B 1.273 b	C 1.425 c

a,b, c: The difference between potassium levels that take on different lowercase letters at the same silicon and NaCl level is important ($p<0.05$).
A, B,C: The difference between different capitalized silicon levels at the same potassium and NaCl levels is important ($p<0.05$).
*: The difference between NaCl1 administration and control (NaCl0) is significant ($p<0.05$).

4. DISCUSSION

In the pot experiment conducted with wheat plants, application of 100 mM NaCl significantly reduced plant fresh weight, plant dry weight and plant height (Tables 4, 5 and 6). In a similar experiment, it was determined that salt stress negatively affected plant development [45]. Increasing the amount of salt in the root zone creates osmotic stress in the plant and prevents the plant from using water [16]. Decreasing the amount of available water causes cell expansion to decrease and shoot development to slow down. In their study, Hu and Schmidhalter (2005) reported that increased osmotic stress, caused by higher concentrations of Na+ and Cl- ions, reduced the uptake of ions such as K^+, Ca^{2+}, and NO_3^-, leading to nutrient deficiencies or imbalances in the plant [17]. In the experiment conducted, while increasing potassium doses under saline conditions did not cause a significant change in plant height at all silicon levels, plant fresh and dry weights generally decreased with the addition of potassium (Tables 4, 5 and 6). In a similar study, the negative effects of salinity were eliminated by potassium application [46]. Zheng et al. (2008) suggested in their study that potassium application reduced salt stress and increased plant growth [47]. In the experiment, potassium did not show the expected results in eliminating the harmful effects of salinity on plant growth parameters. This may be due to the high salt concentration used in the experiment.

While the effect of silicon applications on plant fresh weight and plant height was not found to be significant, plant dry weight increased significantly with silicon application. The effect of NaClxKxSi interaction on plant dry weight was found to be significant (Table 3). In saline conditions, increasing silicon doses at constant K_0 and K_1 levels significantly increased plant dry weight compared to the control (Table 5). In a similar study, a significant increase in plant growth was observed with the addition of silicon to wheat plants grown under salt stress [48]. Gurmani et al. (2013) suggested in their study with two types of wheat under saline conditions that silicon inhibited the transport of Na^+ and increased plant growth [32]. In a study conducted with wheat plants under salt stress conditions, it was determined that silicon and potassium nitrate applications increased plant weight, grain weight, spike height and photosynthesis rate [49].

Table 6 shows that the interaction of NaCl, K, and Si applications had a statistically significant effect on K and Na levels in the plant . This effect caused the K content to decrease in saline conditions and increased the Na content (Tables 8 and 9). In many plants, salt generally causes the Na^+ level to increase and the K^+ level to decrease [15, 50-51]. Erdal et al. (2000) suggested that other cations should be taken in addition to Na in order to make the plant tolerant to salinity in the presence of Na [52]. In salt stress, application of silicon and potassium together or alone increased the potassium uptake of the plant [49]. Gurmani et al. (2013) suggested that silicon applied to wheat plants under salt stress inhibited Na^+ transport and reduced the Na^+/K^+ ratio [32]. There are generally two basic concepts in studies on salt stress. These are salt stress and salt shock. While salt shock is not very visible in the soil in agricultural areas, salt stress disappears after a certain time and its effect gradually increases. In salt stress studies, NaCl or sea water is applied to plants in a controlled manner under laboratory or greenhouse conditions. These applications are generally applied to the experiment at increasing concentrations of 25, 50 or 100 mM NaCl, and the plant is exposed to stress in this way [11, 53-55]. In this experiment, the decrease in potassium content in salty conditions compared to the control can be explained by the fact that the high salt level applied creates an antagonistic effect between Na and K in the root region of the plant, preventing the uptake of potassium.

In the experiment, the application of 100 mM NaCl inhibited the productivity of the plant and directly caused an increase in the activity of antioxidant enzymes. In particular, it caused a significant increase in the activity of the plant's superoxide dismutase and ascorbate peroxidase enzymes. This indicates that the plant is stressed. NaCl, K and Si applications significantly affected antioxidant enzyme activities in wheat plants (Table 11, 12, 13). In the study conducted by Karanlık (2001), it was explained that free oxygen radicals synthesized at increasing levels in the plant under stress damage the cells [56]. As in all living things under stress, antioxidant enzymes that convert free oxygen radicals into harmless compounds during stress in plants are resistant to damage. Enzymes such as SOD, CAT and APX are the most effective enzymes in destroying free oxygen radicals [41, 57-58]. Çakmak (1997) suggested that potassium deficiency may further increase the tissue damage that can be caused by free oxygen radicals synthesized due to salt stress [59]. Potassium applied in salt-stressed cereal crops improved antioxidant activity [60]. Al-Whaibi et al. (2012) reported that silicon applied in salt stress improved SOD, CAT and APX enzyme activities [61]. In another study, significant improvement in SOD and APX enzyme activities was seen with the application of silicon in salt stress [62-64]

5. CONCLUSION

The amount of saline soils in the world and in our country is increasing day by day. This increase reduces yields, and some areas are completely out of production due to excessive salinization. Only in our country, there is a salinity problem in approximately 2.5 million hectares of agricultural area and it is increasing. In this pot trial, the effect of single or combined applications of potassium and silicon under salt stress conditions on some antioxidative enzyme activity in the plant was investigated.

The first wall of strength of plants when exposed to stress is antioxidant enzymes. Antioxidants, which are a defense mechanism in the plant at the time of stress, help the plant to tolerate stress up to a certain limit. In the experiment, the addition of salt to the wheat plant generally reduced superoxide dysmutase activity, while ascorbate caused an increase in peroxidase activity. This increase is indicative of stress caused by salinity. When the dose of silicon was kept constant under saline conditions, administration of

K_1 (150 ppm K) significantly increased SOD activity (Table 11). This is an indication that potassium and silicon $NaCl_1K_1Si_0$ and $NaCl_1K_1Si_2$ applications increase SOD activity against salt stress. The administration of salt did not cause a significant change in CAT activity. In all applications with salinity, APX activity is increased. However, when Si_0 and Si_2 doses were kept constant, the increased potassium doses caused a significant reduction in increased APX activity due to salt stress.

As a result, salt applications caused the plant to stress. Although the KxSi interactions used to relieve this stress provided the plant with tolerance to salinity up to a certain point, they could not prevent the plant from stressing sufficiently. This may have been due to the high concentration of salt used in the experiment, or it may have been due to the fact that the wheat plant used was overly sensitive to salinity.

Acknowledgement

This work is produced from the second author's master's thesis. We would also like to thank the Van Yüzüncü Yıl University Scientific Research Projects Directorate for supporting the study as a master's thesis project (2014-FBE-YL171).

Statements & Declarations

Funding

This study was supported by Yüzüncüyıl University, Scientific Research Projects Coordination Unit.

Author Contributions

All authors contributed to the conceptualization and design of the study. Material preparation, data collection and analysis were carried out by Orhan İNİK and Mehmet Ali BOZKURT. The first draft of the manuscript was written by Orhan İNİK, and the other author commented on previous versions of the manuscript. The other author finally read and approved the manuscript.

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