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# Ameliorative effects of nitric oxide on growth, physiology and biochemistry of chickpea plants under salinity stress

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**Abstract:** Salinity is one of the major environmental constraints affecting agriculture in major regions of the world. This study was conducted to evaluate the effect of exogenous nitric oxide (NO) treatments on two chickpea cultivars (Cagatay and Inci) under salt stress conditions. Different NO doses (0. 75 and 100 µM sodium nitroprusside (SNP)) as an NO source were applied to chickpea plants grown under saline (presence of 50 and 100 mM of NaCl) and nonsaline conditions. In this study, plant shoot fresh and dry weight, root fresh and dry weight, chlorophyll a, b, total chlorophyll, chlorophyll reading value (CRV), relative water content (RWC), electrical conductivity (EC), malondialdehyde (MDA), hydrogen peroxide ($H_2O_2$), antioxidant enzyme activity [superoxide dismutase (SOD), peroxidase (POD), ascorbate peroxidase (APX)], K/Na and Ca/Na ratio were examined. Plant growth, RWC, and chlorophyll content were negatively affected by salinity conditions but exogenous NO treatment improved the parameters. EC, $H_2O_2$, and MDA content were increased with salinity conditions while exogenous NO treatment decreased the searched parameters. K/Na and Ca/Na ratios were decreased with 50 and 100 mM of NaCl treatments. Although the cultivars response to salt stress is similar in general, Inci cultivars were found to be more sensitive to salt stress than Cağatay cultivar. The present study revealed that the exogenous NO treatment supported chickpea seedlings against salinity stress by regulating uptaking mineral elements, the antioxidant enzyme activity, and RWC.

**Key words:** Chickpea, physiology, nitric oxide, plant growth, salt stress

## 1. Introduction

Plants are often exposed to environmental stresses that negatively affect their growth, development, and productivity (Bolat et al., 2014; Khan et al., 2015; Aysin et al., 2020; Chang et al., 2020). The steady increase in abiotic stress factors is due to increased pressure on limited farmland for higher productivity to compensate the growth rate of the human population, which is expected to exceed 9 billion by 2050 (Zia-Ul-Haq et al., 2013; Sehar et al., 2019).

Salt stress is one of the most important environmental factors that plants encounter. Soil salinity is the most important abiotic stress factor which directly limits agricultural production areas worldwide (Tiryaki, 2018). Salinity, in food legumes, can limit plant growth. Sodium chloride (NaCl) is the prevailing salt in the soil, causes osmotic and ionic stress effects, of which the osmotic stress minimizes the ability of plants to take up water, minerals, and reduces symbiotic performance, etc. (Rout and Shaw, 2001; Rao et al., 2002; Khan et al., 2012; Ahmad et al., 2016).

Nitric oxide (NO) has now taken an important role in plant development (Kaya et al., 2020). NO plays an important role in resistance to salt, temperature, drought, and heavy metal stress (Siddiqui et al., 2011). Also, it has a key function in various processes of plant growth and development, including seed germination, seed dormancy, root growth, flowering, photosynthesis, plant metabolism, and cell death (Siddiqui et al., 2011; Manai et al., 2014; Mostofa et al., 2015). Nitric oxide (NO) is a gaseous intracellular and intercellular signal molecule with a wide variety of regulatory functions various physiological processes in mammals (Meilhoc et al., 2011). It has been recognized as a potential plant hormone related to plant defense mechanism (Fatma et al., 2016a; 2016b, Per et al., 2017a; 2017b).

Legumes are rich in proteins, recovery of marginal lands, and very important components of the human diet (Jukanti et al., 2012). Among the edible legumes, chickpea (*Cicer arietinum* L.) is a very popular crop around the world as it can supply a rich source of proteins, fats, carbonhydrates for humans and animals and increase the input of combined $N_2$ into the soil through symbiotic association with *Rhizobium* (Rasool et al., 2015; Garg and Singla, 2016). Chickpea normally grows under rainfed and irrigated conditions (Rasool et al., 2015) and this crop is very sensitive to salinity stress.

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Limited studies have been carried out on the application of NO to reduce the effect of salt stress in chickpeas. In this study, we investigated the effect of NO on plant growth, nutrient uptake, physiological and biochemical properties of chickpea seedlings under salt stress.

## 2. Material and methods

The experiment was conducted in greenhouse, the temperature ranged from 19 °C to 32 °C, and the average humidity was 65%. Two chickpea cultivars (Cagatay and Inci) were used as plant materials. Different NO doses (0, 75, and 100 µM sodium nitroprusside (SNP as an NO donor)) were applied to chickpea plants grown under saline (50 and 100 mM of NaCl) and nonsaline conditions. NO treatments were prepared at 3 levels of 0 (control), 75, and 100 µM SNP in ultrapure water with Tween 20. These solutions were applied to the seeds (for 24 h at 20 °C) in petri dishes and then seeds were taken out and air-dried. The NO treated seeds were planted in pots (mixture of peat: perlite (3:1, v: v) (Sahin et al., 2002) and seedlings have been obtained under greenhouse conditions. Ten days after emerging seedlings were foliarly treated with NO at 3 levels of 0 (control), 75, and 100 µM SNP. The foliar NO treatments were repeated three times at one weekly interval, as the solutions were sprayed on leaves.

Salinity treatments were initiated ten days after seedling emergence with an increase of 25 mM NaCl to avoid an osmotic shock for plants. The irrigation water was applicated at three levels of NaCl (0, 50, and 100 mM) and their electrical conductivities were measured as 0.54, 5.23, and 7.61 ds $m^{-1}$ respectively. Plants were irrigated with a halfstrength Hoagland solution with ten days intervals.

The relative water content (RWC), electrical conductivity (EC), and chlorophyll reading value (CRV) were measured after 52 days of sowing from each pot before harvest. During harvest, plants were cut from the soil level, and above-ground biomass (shoot) and roots were separated for measurements and analysis. In this study, plant shoot fresh and dry weight, root fresh and dry weight, chlorophyll a, b, total chlorophyll, malondialdehyde (MDA), hydrogen peroxide $H_2O_2$, antioxidant enzyme activity (superoxide dismutase (SOD), peroxidase (POD), ascorbate peroxidase (APx), K/Na and Ca/Na ratio were examined.

RWC and EC were determined according to Yildirim et al. (2015).

CRV was determined as leaf chlorophyll reading value (SPAD) with a chlorophyll meter (Konica Minolta SPAD-502). The measurements were carried out at ten different spot on leaves of each pot, and then the average was used for analyses.

For leaf chlorophyll content (chlorophyll a, chlorophyll b, and total chlorohyll (a+b) of plants; samples were cut at 10 mm diameter from the middle leaves and shaken with 0.2 mL 80% acetone. Then samples were centrifuged at 10,000 rpm at 5 °C by brought to the final volume with 80% acetone. The absorbance values were measured at 663 and 645 nm and calculated (Khan et al., 2003; Ekinci et al., 2020).

Assays of MDA, $H_2O_2$, and antioxidant enzyme activity (POD, APX, SOD) were performed by UV/vis spectrophotometer according to Sarafi et al. (2018).

For the determination of mineral nutrition content, the leaves of the chickpea cultivars were dried at 68 °C for 48 h and K, Ca and Na contents were analyzed by a coupled plasma spectrophotometer (Optima 2100 DV; Perkin-Elmer, Shelton, CT) (Helrich, 1990).

A completely randomized design with three replications was used in the current experiment and we had 12 plants per replicate. Data were subjected to analysis of variance (ANOVA). Means were separated by Duncan's multiple range tests ($p < 0.05$).

## 3. Results

### 3.1. Plant growth

Stress conditions negatively affected the growth parameters of two chickpea varieties. 100 mM of NaCl decreased shoot fresh and dry weight, and the root fresh and dry weight by 47, 37, 54 and 50%, respectively in Cagatay cultivar compared to control. In Inci, the shoot fresh and dry weight, and the root fresh and dry weight were reduced by 62%, 49%, 72%, and 68%, respectively compared to untreated one (Table 1). NO treatments alleviated the deleterious effects of salinity stress on the growth, physiological and biochemical properties of chickpea cultivars (Table 1).

### 3.2. Chlorophyll content (Chl a, Chl b, total Chl)

The Chl a, Chl b, and total Chl contents were significantly decreased by salt stress in two cultivars. The maximum decrease for Chl b compared to the control was recorded at 100 mM NaCl respectively in Inci (53%) and Cagatay (23%) cultivars (Figure 1). This decrease in Chl content in chickpea leaves might partially cause a decrease in growth. Supplementation of NO (100 mM NaCl + 75 µM NO) was increased (23%) comparison with (100 mM NaCl) (for Chl b parameters) as control in Inci cultivar. In Cağatay cultivar the best result (nearly equal control) was obtained 100 mM NaCl + 75 µM NO (for Chl b parameters) (Figure 1).

### 3.3. RWC, EC and SPAD

RWC is adversely affected by salinity stress in chickpea cultivars. With the effect of salt (at 100 mM NaCl) there was a decrease (24%-Cagatay and 44%-Inci) in two cultivars (Table 2). The decrease in RWC of salt-stressed plants was alleviated by exogenous application of NO, resulting in an enhancement in RWC of 14% in Cağatay and 42% in Inci at 100 mM NaCl + 100 µM NO. Cell

**Table 1**. The effect of salinity and NO treatments on plant growth in chickpea plants.

| Cultivar | NaCl (mM) | NO (µM) | Shoot fresh weight (g plant$^{-1}$) | Shoot dry weight (g plant$^{-1}$) | Root fresh weight (g plant$^{-1}$) | Root dry weight (g plant$^{-1}$) |
|---|---|---|---|---|---|---|
| CAGATAY | **0** | 0 | 42.09 a | 6.5 a | 39.3 a | 2.63 a |
| | | 75 | 42.15 a | 6.7 a | 38.67 a | 2.71 a |
| | | 100 | 37.80 b | 6.06 b | 38.52 a | 2.36 b |
| | **50** | 0 | 32.50 c | 5.06 d | 21.97 c | 1.43 de |
| | | 75 | 37.99 b | 5.76 c | 28.85 b | 1.79 c |
| | | 100 | 29.79 d | 4.63 e | 29.48 b | 1.46 d |
| | **100** | 0 | 22.37 f | 4.08 g | 18.25 d | 1.31 f |
| | | 75 | 26.84 e | 4.33 f | 21.29 c | 1.39 df |
| | | 100 | 20.83 f | 3.27 h | 23.89 c | 1.34 ef |
| | **NaCl** | | *** | *** | *** | *** |
| | **NO** | | *** | *** | *** | *** |
| | **NaCl x NO** | | *** | *** | *** | *** |
| INCI | **0** | **0** | 20.88 a | 2.48 a | 14.52 b | 0.94 ac |
| | | **75** | 13.62 bc | 1.99 ac | 8.69 ce | 0.8 bd |
| | | **100** | 10.14 cd | 1.51 ac | 11.06 c | 0.99 ab |
| | **50** | **0** | 8.47 d | 1.15 c | 6.26 ef | 0.49 de |
| | | **75** | 10.67 cd | 1.49 ac | 14.21 b | 0.71 bd |
| | | **100** | 17.29 ab | 2.30 ab | 18.66 a | 1.15 a |
| | **100** | **0** | 7.92 d | 1.26 bc | 4.03 f | 0.3 e |
| | | **75** | 10.07 cd | 1.58 ac | 8.85 cd | 0.65 cd |
| | | **100** | 7.2 d | 1.21 bc | 7.17 de | 0.57 de |
| | **NaCl** | | *** | **ns** | *** | *** |
| | **NO** | | **ns** | **ns** | *** | ** |
| | **NaCl x NO** | | *** | * | *** | ** |

Data followed by a different letter in column were significantly different according to the DMRT, *: $p < 0.05$; **: $p < 0.01$; ***: $p < 0.001$; ns: $p > 0.05$.

membrane permeability of chickpea cultivars used in the study was determined by the measurement of EC (Table 2). Salinity stress markedly increased the cell membrane permeability in chickpea cultivars but it had a lower effect on Inci cultivar than Cağatay. NaCl treatments of 100 mM damaged the cellular membranes. This damage was reflected in terms of increased electrolyte leakage. Salinity caused a decrease in SPAD values in Cağatay cultivar (Table 2).

### 3.4. $H_2O_2$ and MDA

Increase in $H_2O_2$ contents was observed with the raise of NaCl dose applied to chickpea plants (Table2). $H_2O_2$ content increased by 82% in two cultivars at 100 mM NaCl. Supplementation of exogenous NO to NaCl-stressed plants decreased $H_2O_2$ content by 24% (Cağatay) and 25% (Inci) at 100 mM NaCl + 100 µM NO. As for MDA, an increase by more than 100% (Table 2) in both cultivars under salinity stress.

### 3.5. Antioxidant enzyme activity

The activity of SOD, APX, and POD was increased by salinity stress in two cultivars. Their activities were decreased with exogenous NO application (Figure 2). In Cağatay cultivar, NO applications varied depending on severity of salt stress and the dose of the applications. At 50 mM salt stress, POD, and SOD activity decreased with 100 µM NO application, while there was no significant change in APX activity. At 100 mM salt stress, POD activity decreased significantly with 100 µM NO and 75 µM NO

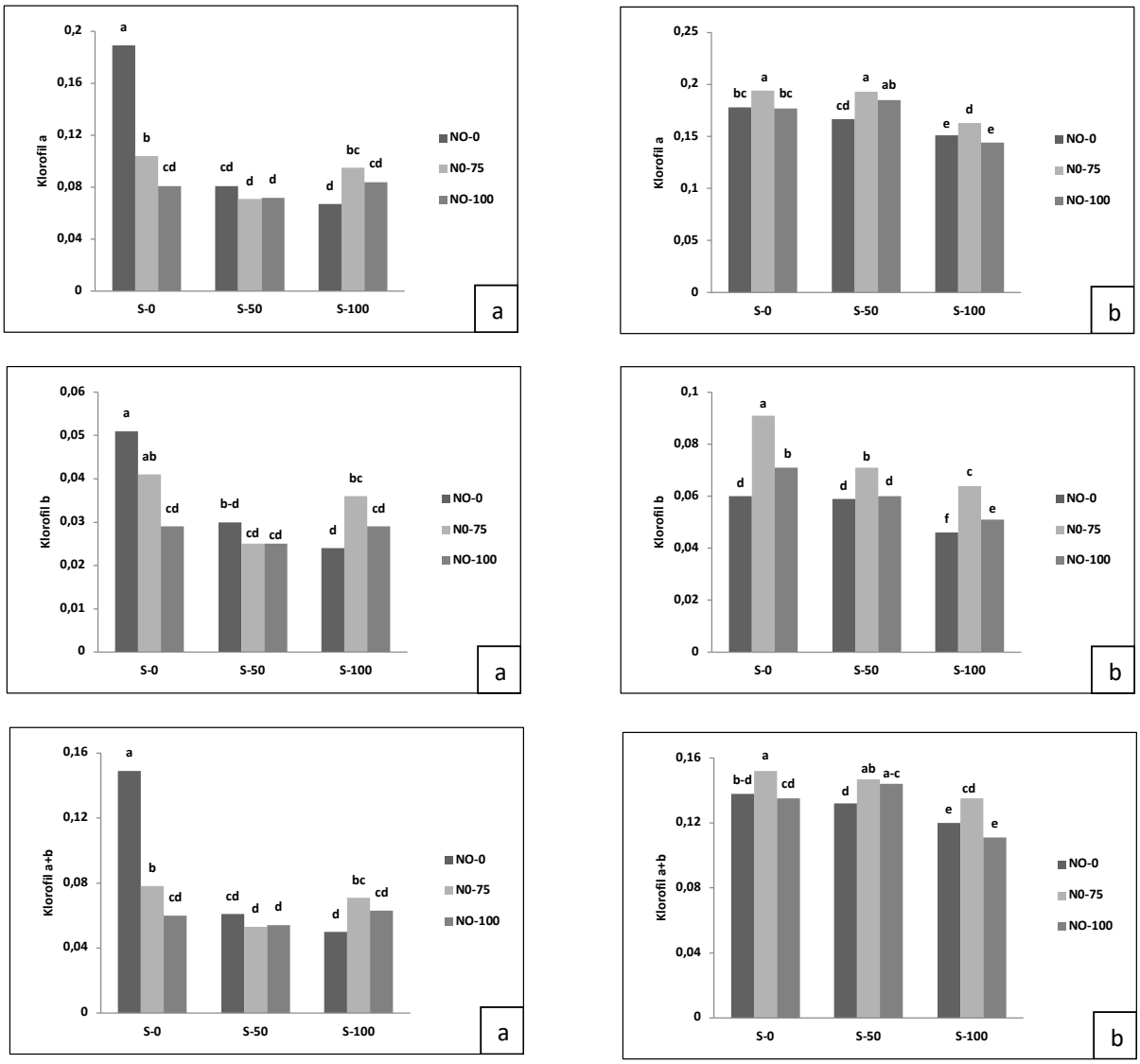


**Figure 1.** Effects of NO on chlorophyll content of chickpea plants under salt stress (Inci-a, Cagatay-b). Different letters indicate significant difference ($p < 0.05$) among the treatments.

application. It occurred in POD, APX, and SOD activities in both salt strengths with NO application in pearl variety.

### 3.6. Mineral uptake

Salinity stress was significantly affected $Na^+$, $K^+$, and $Ca^{2+}$ uptake and the ratio of $K^+/Na^+$ and $Ca^{2+}/Na^+$ significantly decreased in chickpea plants but exogenous NO treatment increased (Figure 3). When evaluated in terms of NO applications, there was no statistically significant difference between applications in salt stresses.

## 4. Discussion

Chickpea is an important pulse crop grown in the arid and semiarid regions. High salt concentration causes osmotic and ionic stress in plants. It limits the growth and development of plants by affecting several key metabolic processes (Läuchli and Grattan, 2007). In this research, two different chickpea cultivars were used and the effect of NO application on salt stress was investigated. In this purpose, the effect of NO application on plant shoot fresh and dry weight, root fresh and dry weight, chlorophyll a, b, total chlorohyll, CRW (SPAD), RWC, EC, MDA, $H_2O_2$, antioxidant enzyme activity (SOD, POD, APX), K/Na and Ca/Na ratio of two chickpea cultivars under salinity conditions were examined under salt stress.

In study, salt stress significantly decreased the growth of chickpea cultivars. These results are similar to the findings of Kotula et al. (2015), Ahmad et al. (2016) who reported that salinity stress significantly decreased the plant

**Table 2.** The effect of salinity and NO treatments on EC (electrical conductivity), RWC (relative water content), CRV (chlorophyll reading value), MDA (malondialdehyde), and hydrogen peroxide ($H_2O_2$) in chickpea plants.

| Cultivar | NaCl (mM) | NO (µM) | EC (%) | RWC (%) | CRV (SPAD) | MDA | $H_2O_2$ |
|---|---|---|---|---|---|---|---|
| CAGATAY | **0** | **0** | 11.24 f | 74.87 b | 42.14 bc | 11.12 d | 103.91 e |
| | | **75** | 11.86 f | 78.24 a | 46.03 a | 13.14 cd | 113.34 d |
| | | **100** | 20.72 e | 77.63 a | 43.13 bc | 14.47 bc | 103.32 e |
| | **50** | **0** | 27.87 d | 71.20 c | 37.07 ef | 16.26 b | 137.50 c |
| | | **75** | 20.96 e | 65.45 d | 44.27 ab | 15.68 b | 120.11 d |
| | | **100** | 21.83 e | 67.60 d | 41.83 c | 14.06 bc | 117.58 d |
| | **100** | **0** | 60.85 a | 57.09 f | 35.50 f | 23.65 a | 189.47 a |
| | | **75** | 38.80 c | 61.38 e | 40.80 cd | 22.37 a | 164.86 b |
| | | **100** | 50.96 b | 68.14 d | 39.20 de | 23.75 a | 143.96 c |
| | **NaCl** | | *** | *** | *** | *** | *** |
| | **NO** | | *** | *** | *** | *** | *** |
| | **NaCl x NO** | | *** | *** | **ns** | *** | *** |
| INCI | **0** | **0** | 13.80 d | 71.69 b | 32.87 ab | 11.17 e | 108.67 e |
| | | **75** | 23.89 c | 70.95 b | 37.97 a | 10.87 e | 111.00 e |
| | | **100** | 15.05 d | 93.38 a | 31.00 ab | 12.33 de | 111.70 e |
| | **50** | **0** | 46.69 b | 55.36 bc | 31.23 ab | 17.62 c | 144.32 c |
| | | **75** | 39.97 b | 50.82 bc | 30.67 ab | 13.59 d | 142.52 c |
| | | **100** | 46.37 b | 56.40 bc | 33.10 ab | 13.23 d | 136.41 d |
| | **100** | **0** | 56.75 a | 40.00 c | 33.30 ab | 27.29 a | 195.88 a |
| | | **75** | 58.85 a | 55.25 bc | 30.80 ab | 20.60 b | 157.64 b |
| | | **100** | 45.97 b | 62.46 b | 29.17 b | 18.63 c | 146.38 c |
| | **NaCl** | | *** | *** | **ns** | *** | **ns** |
| | **NO** | | * | * | **ns** | *** | *** |
| | **NaCl x NO** | | ** | **ns** | **ns** | *** | *** |

Data followed by a different letter in column were significantly different according to the DMRT *:$p < 0.05$; **:$p < 0.01$; ***: $p < 0.001$; ns: $p > 0.05$.

growth in chickpea. But coapplication of NO and salinity promoted the growth of plants under saline conditions compared to the salinity treatment. These results supported earlier reports on different crops, such as wheat (Kausar and Shahbaz, 2013), rice (Mostofa et al., 2015), pepper (Shams et al., 2019), and pea (Dadasoglu et al., 2021). NO can weaken the cell wall, act on the phospholipids bilayer, increase membrane fluidity and stimulate cell enlargement and plant growth (Leshem and Haramaty, 1996). They reported that NO is involved in increasing the osmotic pressure of the plant cells and improving the cytoplasmic viscosity under high salinity. Nitric oxide is an important signaling molecule involved in amelioration of growth and development of plants under biotic and abiotic stresses (Esim and Atici, 2014; Manai et al., 2014).

The Chl a, Chl b, and total Chl contents were significantly decreased by salt stress in the two cultivars. The decrease in Chl content under NaCl stress (Figure 1) might be assigned to the demolition of Chl pigments, decreased Chl syntheses, and the exposure of the pigment-protein complexes (Rasool et al., 2013). Similarly, Ahmad et al. (2016) determined that salinity decreased chlorophyll content in chickpea, but combined the application of NO and salinity increased chlorophyll content compared to the control. NO was found to activate the enhancement of photosynthetic activity in chickpea plants, potentially by protecting the membrane of the cell organelle containing Chl against salt-stimulated ion toxicity (Kausar and Shahbaz, 2013). The increase in photosynthetic activity due to NO application under salt stress, has also been reported plant species including wheat (Ruan et al., 2004), rice (Habib and Ashraf, 2014), chickpea (Ahmad et al., 2016) and pea (Dadasoglu et al., 2021).

RWC is adversely affected by imposition of NaCl, which leads to decrease in water uptake and injury of

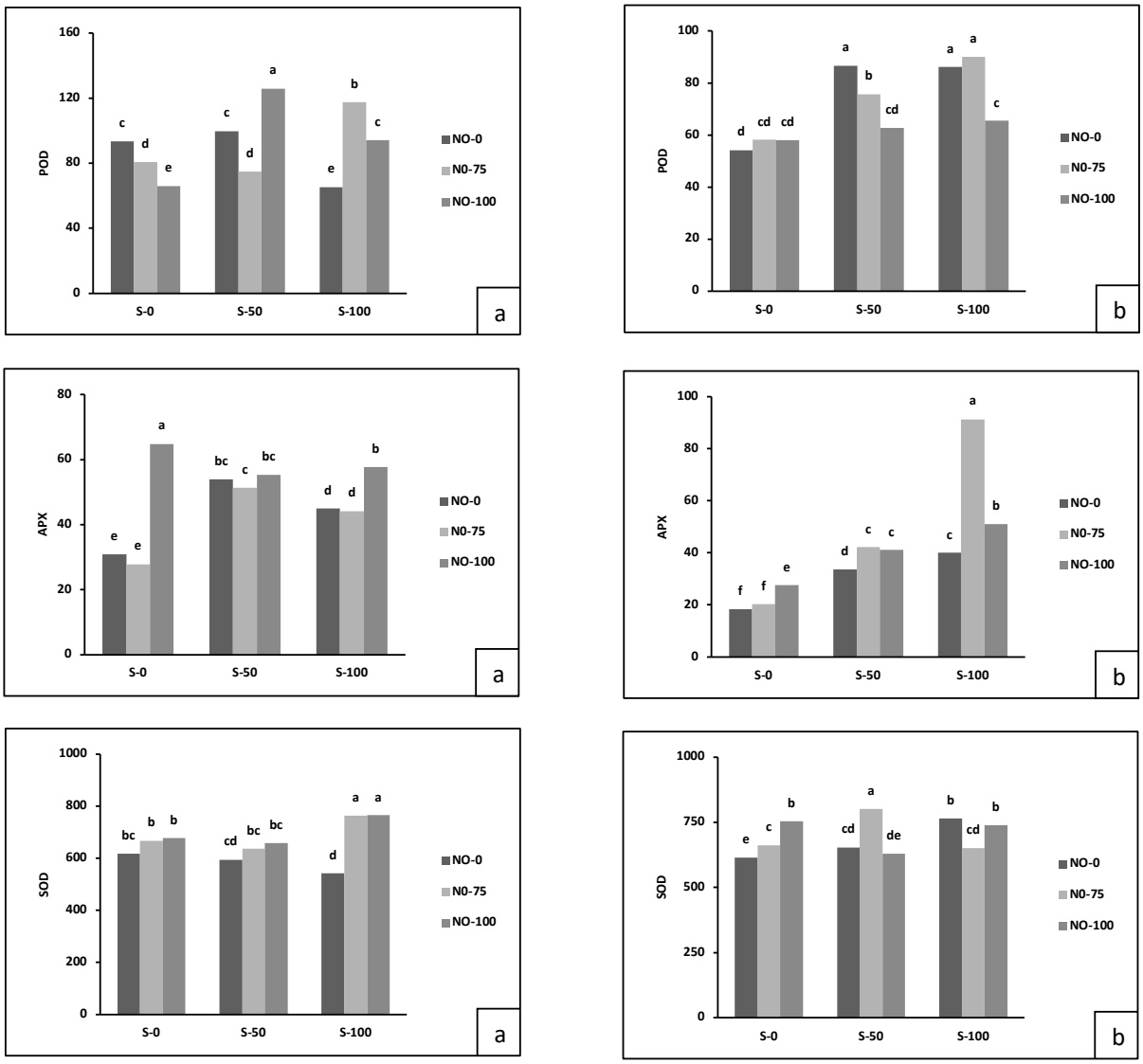


**Figure 2.** Effects of NO on activities of peroxidase (POD), ascorbate peroxidase (APX), and superoxide dismutase (SOD) in leaves of chickpea plants under salt stress (Inci-a, Cagatay-b). Different letters indicate significant difference ($p < 0.05$) among the treatments.

root system (Zeng et al., 2011). When plants are exposed to salinity, primarily they face an osmotic challenge that reduces water uptake by roots. Besides ABA-mediated stomatal closure effects, transpiration entails low relative water content in the cell (Blatt & Armstrong 1993). As it reported before studies (Zeng et al., 2011), in the present study, supplementation of NO had a positive impact on RWC of chickpea plants under salt stress too (Table 2). Salinity decreased RWC and NO application increased in some crops, including chickpea (Ahmad et al., 2016), pea (Dadasoglu et al., 2021), and rice (Habib and Ashraf, 2014) which is similar to our findings. The decrease in relative water content could be due to nonavailability of water near the root zone and loss of water by transpiration (Kotagiri and Kolluru, 2017).

Salinity stress causes elevated ROS levels, and these elevated ROS levels play a dual role in the salinity responses of plants; as toxic by products causing oxidative damage or as signaling molecules mediating salt tolerance (Miller et al. 2008; Jiang et al. 2013). Salt stress induces lipid peroxidation through reactive oxygen species (ROS) production (Liang et al., 2003; Verma and Mishra, 2005), thus making the membrane leaky as evinced by increased electrical conductivity. Exogenously applied NO could prevent injury to membranes (Sheokand et al., 2008). In the present investigation also, NO could completely ameliorate

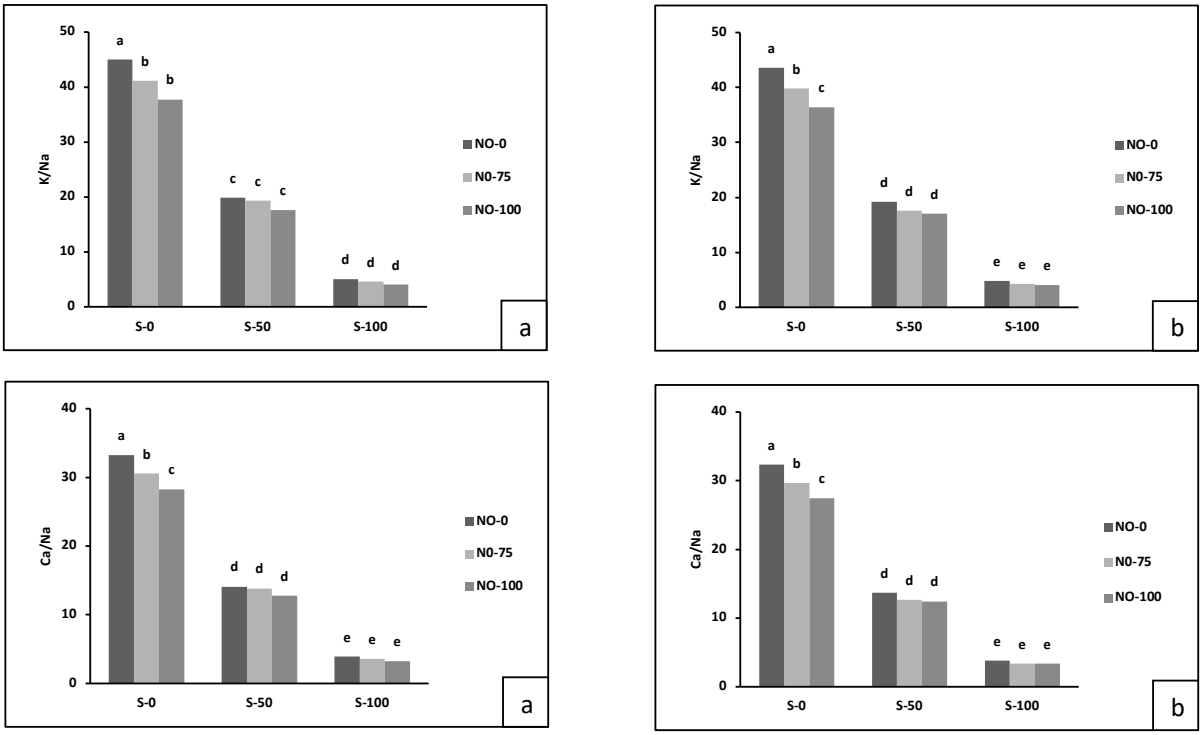


**Figure 3.** Effects of NO on K/Na and Ca/Na ratio on chickpea plants under salt stress (Inci-a, Cagatay-b). Different letters indicate significant difference ($p < 0.05$) among the treatments.

the adverse effect of NaCl treatment on membrane injury and lipid peroxidation. A protective effect of NO on relative membrane injury has been reported under salt stress (Zhao et al., 2004; Zhang et al., 2006; Wu et al., 2011; Ahmad et al., 2016; Shams et al., 2019; Dadasoglu et al., 2021). The findings were similar to those of Shams et al. (2019) who demonstrated that salinity stress decreased RWC and increased EC, but exogenous NO application ameliorated the adverse effect of salinity stress.

As for MDA, an increase was observed in both cultivars under salinity stress. As well $H_2O_2$ supplementation of NO decreased. Thereby increasing tissue electrolyte conductivity chickpea plants accumulated higher $H_2O_2$ and MDA contents under salt stress. This increase in $H_2O_2$ and MDA content may be due to membrane damage caused by ROS-induced oxidative damage. Consistent with the observations of Zheng et al. (2009); Khan et al. (2012) and Ahmad et al. (2016) with agreement. But exogenous application of NO decreased $H_2O_2$ and MDA content (Ahmad et al., 2018). NO application could be an effective way to protect plants against oxidative injury caused by salt stress. NO stimulates mitogen-activated protein kinase (MAPK) which in turn activates transcription factors for induction of stress-related genes (Neill et al., 2008).

These results are similar to earlier studies of Ahmad et al., (2018), Shams et al., (2019) and Dadasoglu et al., (2021) who found that NO treatment increased enzyme activity under salinity stress. Increasing evidence indicates that ROS also function as important signalling molecules in plants and that they are involved in regulating a broad range of processes, such as growth, development, defence, and responses to various abiotic and biotic stresses (Radwan et al., 2010; Sharma et al., 2012; Baxter et al., 2014; Vuleta et al., 2016; Xu et al., 2019). NO, as a signaling molecule and it activates the expression of antioxidant enzymes thereby providing salt tolerance (Hernandez et al., 2000; Ahmad et al., 2016; Gadelha et al., 2017). Therefore, it can be concluded that NO treatment increased the enzyme activity under saline condition. Zhang et al. (2004); Wu et al. (2011); Ahmad et al. (2016); and Dadasoglu et al. (2021) on maize (*Zea mays*), soybean (*Glycine max*), chickpea (*Cicer arietinum*) and pea (*Pisum sativum*) respectively, demonstrated that application of NO increased the plant growth under saline conditions, which might be due to increased activities of antioxidant enzymes. NO has been shown to alleviate the effects of biotic and abiotic stresses on plants by mediating $H_2O_2$ induced mitigation of oxidative damage by regulation of the antioxidant defense mechanism (Mostofa et al., 2015; Singh and Vinayak, 2015). SOD, APX, and POD are known as ROS eliminator (Radwan et al., 2010; Sharma et al., 2012; Vuleta et al., 2016).

As we mentioned in the previous study, the decrease in $K^+/Na^+$ and $Ca^{2+}/Na^+$ ratios can be connected to the over accumulation of $Na^+$ and the decrease in $Ca^{2+}$ and $K^+$ concentration in leaves of pea cultivars under NaCl stress (Dadasoglu et al., 2021). Shams et al. (2019) reported that, by enhancing the absorption of K, Ca and declining the absorption of Na and then it led to mitigate the deleterious effect of salinity. The results are same in this study. Therefore, it can be concluded that exogenous NO treatment increased the ratio $K^+/Na^+$ and $Ca^{2+}/Na^+$.

## 5. Conclusion

Plants expose to a wide range of environmental stresses. Among these, salinity is considered one of the major abiotic stresses that limits crop yield. As known chickpea is highly sensitive to salinity and therefore needs considerable enhancement of salinity tolerance. Salinity decreased the growth and development of chickpea by acting morphologically, physiologically, and biochemically. Taken together, our findings highlighted the importance of NO on plant growth, antioxidant enzyme activity and chlorophyll contents in chickpea plants. Exogenously NO treatment diminished oxidative damage by upregulating antioxidant enzymes and osmolytes, thereby ameliorating a significant decrease in ROS-induced lipid peroxidation and electrolyte leakage. Therefore, NO can be a promising approach for salt stress management.

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