

## Salicylic Acid Seed Priming Enhances Lentil Germination and Seedling Vigor under Salinity Stress

Tuz Stresi Altında Salisilik Asit Tohum Ön Uygulaması ile Mercimekte Çimlenme ve Fide Canlılığının Arttırılması

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### Abstract

Salicylic acid (SA), a plant-derived phenolic compound, acts as a signaling molecule that regulates controlling diverse plant responses and plays a role in plant defense under stress conditions. This study aimed to assess the potential of SA seed priming in mitigating the negative effects of salt stress on lentil germination and early seedling development. The germination and vigor of seeds primed with five different doses of SA (0, 0.25, 0.5, 0.75, and 1 mM) were tested at five salinity levels (0, 50, 100, 150 and 200 mM) in peat-perlite. The research was conducted using a completely randomized design with factorial arrangement under greenhouse conditions. Results indicated that low concentrations of SA (0.25 mM) had positive effects on days to seedling emergence, emergence energy (EE), emergence percentage (EP), emergence rate index (ERI), mean emergence time (MET), peak value (PV) and seedling vigor index suggesting that SA can enhance germination and seedling emergence under high salinity. However, as SA concentration increased, a decrease in emergence rate was observed, indicating that higher SA levels may have inhibitory effects on seed performance after sowing. The results also show that SA pre-treatment can significantly improve shoot length, root length, fresh shoot weight, and fresh root weight of lentil seedlings exposed to NaCl stress. Adverse effects of increasing NaCl concentrations were noted for all measured traits, with shoot length, root length, fresh shoot weight, and fresh root weight decreasing, and, no growth observed at the highest NaCl concentration (200 mM). In conclusion, SA can be effectively used in seed priming to mitigate salinity stress during early lentil growth, but the effectiveness of this treatment depends on the salinity level.

**Keywords:** Abiotic stress, Emergence, Growth, Hormone, *Lens culinaris* L.

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## Öz

Bitkisel kaynaklı bir fenolik bileşik olan salisilik asit (SA), çeşitli bitki tepkilerini kontrol eden bir sinyal molekülü olup stres şartlarında bitki savunmasında rol almaktadır. Bu çalışma, tuz stresinin mercimek çimlenmesi ve erken fide gelişimi üzerindeki olumsuz etkilerini azaltmada salisilik asit ile gerçekleştirilen tohum ön işlem uygulamasının etkisini değerlendirmeyi amaçlamaktadır. Beş farklı salisilik asit dozu (0, 0.25, 0.5, 0.75 ve 1 mM) ile ön işlem uygulanan tohumların çimlenme ve canlılığı 5 farklı tuz seviyesinde (0, 50, 100, 150 ve 200 mM) torf-perlit ortamında test edilmiştir. Deneme faktöriyel düzende tesadüf parselleri deneme deseninde sera koşullarında yürütülmüştür. Sonuçlar, düşük konsantrasyonlardaki salisilik asidin (0.25 mM), fide çıkış zamanı, çıkış enerjisi (EE), çıkış yüzdesi (EP), çıkış oran indeksi (ERI), ortalama çıkış süresi (MET), pik değer (PV) ve fide canlılık indeksi gibi parametreler üzerinde olumlu etkilerini ve SA'ın yüksek tuzlu koşullar altında çimlenmeyi ve fide çıkışını artırabileceğini göstermiştir. Ancak, SA konsantrasyonu arttıkça, çıkış oranında azalma tespit edilmiş olup, yüksek SA seviyesinin ekimden sonra tohum performansı üzerinde engelleyici etkilere sahip olabileceğini düşündürmüştür. Sonuçlar, SA ile ön işlem uygulamasının NaCl stresine maruz kalan mercimek fidelerinin sürgün uzunluğunu, kök uzunluğunu, taze sürgün ağırlığını ve taze kök ağırlığını önemli ölçüde iyileştirebileceğini göstermektedir. Artan NaCl konsantrasyonlarında sürgün uzunluğu, kök uzunluğu, taze sürgün ağırlığı ve taze kök ağırlığı dahil olmak üzere gözlenen tüm özellikler için olumsuz etkiler tespit edilmiş ve en yüksek tuz konsantrasyonu 200 mM seviyesinde büyüme gözlemlenmemiştir. Sonuç olarak, salisilik asit ile ön işlem uygulamasının tuzluluk düzeyine bağlı olarak etkinliğinin değişiklik göstermesine rağmen mercimeğin erken büyüme aşamalarında tuzluluk stresini azaltmada etkili bir araç olarak kullanılabileceğini göstermektedir.

**Anahtar Kelimeler:** Abiyotik stres, Çıkış, Büyüme, Hormon, *Lens culinaris* L.

## 1. Introduction

Lentil (*Lens culinaris* Medik.) is a globally important food legume, cultivated for its edible seeds. According to FAO (2023), the global lentil harvested area in 2023 was 5.6 million hectares, and total production was 7.0 million tons.

Lentil plays a vital role in many areas, including ensuring food security, reducing poverty, revitalizing soil nutrients, and promoting national economic development. Lentil has contributed significantly to human nutrition since its cultivation in Southwest Asia 11,000 years ago (Coyne and McGee, 2013). It contains approximately 25% protein, making it a valuable source of plant-based protein. Additionally, lentil is a good source of carbohydrates (~50%) and dietary fiber (~8%).

Lentil is well-suited to semi-arid environments (Farooq et al., 2020; Muehlbauer and McPhee, 2005). High salt levels in the soil (salinity) pose a constant threat to crop yields in arid and semi-arid areas (Manchanda and Garg, 2008; Singh et al., 2017). Evaporation and plant water use (evapotranspiration) concentrate salts in the root zone, which harms plant growth (Ashraf and Waheed, 1993).

Soil salinity, the second-leading abiotic stress after drought for lentil, significantly limits plant growth and seed yield by 20% to 100% (Soren et al., 2020; Kumawat et al., 2017). Addressing salinity, an escalating global issue, is crucial for sustainable agricultural production. Genetic strategies will play a basic role in enhancing crop resilience to salinity (Vadez et al., 2012).

Regulation of salinity stress is not confined to the early growth stages of plants. Analyzing salt tolerance at only one growth stage is insufficient, as plants continuously adapt throughout their life periods (Foolad, 2004). It is essential to evaluate a crop's ability to resist salt at each growth stage to assess its overall tolerance (Ashraf and Waheed, 1993).

SA is a small phenolic molecule native to plants that functions as a signaling molecule, regulating plant responses to a wide range of stimuli. It is also important in responding to both biotic and abiotic stress (Al-Fraihat et al., 2023). Under abiotic stress, exogenous SA enhances the internal glutathione cycle, thereby boosting antioxidant and metal detoxification systems in plants (Wani et al., 2017). The effectiveness of SA in plants is influenced by several factors, including the applied dosage, application method, plant developmental stage, and acclimation (Yang et al., 2023). Priming is a technique that has been effectively applied to improve seed germination, seedling establishment, and early vegetative growth in various field crops like sorghum, millet, triticale, and wheat under salt stress conditions (Beckers and Conrath, 2007; Nimir et al., 2015; Demirbaş and Balkan, 2018; Yadav et al., 2020).

Lentil, like many legumes, has limited tolerance to salinity (Singh et al., 2017). Even mild salt levels in the soil can be detrimental, limiting their cultivation to non-saline environments (Ashraf and Waheed, 1993). Recent studies indicate that salt application and priming in lentils significantly affect germination and emergence (Mushtaq et al., 2017; Alsaeedi et al., 2017; Çakır and Ceyhan, 2021; Sarkar et al., 2024). Sarkar et al. (2024) reported that NaCl stress caused noticeable reduction in the seedlings' shoot and root length. Hossain et al. (2017) reported that NaCl treatment reduced seedling fresh weight by 44% compared to the control. Çakır and Ceyhan (2021) reported that the germination rate decreased as the salt concentration increased, from the control group to the 120 mM NaCl treatment. Similarly, Kayıs and Ceyhan (2015) reported that shoot length decreased with increasing salt doses at 30, 60, 90, and 120 mM NaCl. Foti et al. (2019) noted that differences in germination percentage and other observations were not statistically significant during the entire period at low salt stress (50 mM NaCl). However, they found that high salt stress (100 and 200 mM NaCl) resulted in substantial differences in germination percentage, with the most important effects observed at 200 mM NaCl. Özkorkmaz and Öner (2022) reported that 0.5 SA seed priming reduced the negative effects of salt under high salt concentration in barley. Mushtaq et al. (2017) and Alsaeedi et al. (2017) reported using silica nanoparticles to decrease the effects of salinity stress and improve both shoot and root length. However, there is limited information on the positive effects of SA seed priming on lentil seedling growth.

This study aimed to investigate the effects of seed emergence, seedling and root traits of lentil during the early seedling period at low and high salt concentrations (50-200 mM NaCl), as well as the effects of seed priming with SA on salt stress

## 2. Materials and Methods

The research was conducted under greenhouse conditions at the University of Dicle, Faculty of Agriculture Department of Field Crops. The average temperature and relative humidity were 24 °C and 60% during the day, and 18 °C and 80% at night and photoperiod of 16 h and 8 h (light-dark).

Five SA doses (0, 0.25, 0.50, 0.75, and 1.0 mM/L), five salt (NaCl) concentrations (0, 50, 100, 150, and 200 mM NaCl/L), and their combinations were applied. The red lentil variety Ilke, developed by Dicle University, was used in the study.

Plastic pots, 0.5-liter, with top and bottom diameters of 10 cm and height of 12 cm, were filled with a 1:1 mix of peat and perlite. Seeds were sterilized before treatment by sinking in a 5% sodium hypochlorite solution for 3 min. Seeds were then washed with distilled water and dried with sterile filter paper.

Seeds were soaked for 24 h in 0 (control), 0.25, 0.50, 0.75, and 1.0 mM SA concentrations for seed priming. After priming, 20 seeds were sown in plastic pots. The experiment was arranged in a completely randomized design in a factorial arrangement with two replications.

Different doses of NaCl were applied for 10 days after sowing to maintain the desired salt concentrations. Plants were harvested 11 days after emergence. The shoots and roots were separated and washed under running tap water.

For the observations, the number of seedling emergences was recorded daily from the first to the seventh day. Emergence energy (EE) (Eq.1), emergence percentage (EP) (Eq.2), emergence rate index (ERI) (Eq.3), mean emergence time (MET) (Eq.4), peak value (PV) (Eq.5), and seedling vigor index (VI) (Eq.6), mortality (Eq.7), seedling shoot and root lengths, seedling fresh shoot and root weights were measured. The seedling shoot and root length were measured in centimeters, and fresh shoot and root weight were determined using precision scales on the 11th day after emergence.

$$\text{Emergence energy (EE)} = \frac{\text{No.of seeds emerged on day (4.day)}}{\text{Total number of seeds tested}} \times 100 \text{ (Maguire,1962)} \quad (\text{Eq.1})$$

$$\text{Emergence percentage (EP)} = \frac{\text{Number of seeds emerged}}{\text{Total number of seeds tested}} \times 100 \text{ (Czabator, 1962; Erbach, 1982)} \quad (\text{Eq. 2})$$

$$\text{Emergence Rate Index (ERI)} = \left(\frac{E1}{1}\right) + \left(\frac{E2}{2}\right) + \left(\frac{E3}{3}\right) + \dots + \left(\frac{Ei}{i}\right) \text{ (AOSA, 1983)} \quad (\text{Eq. 3})$$

E1: Number of first day emergence, E2: Number of second day emergence

$$\text{Mean emergence time (MET)} = \frac{\sum Dn}{\sum n} \text{ (Ellis and Roberts, 1981)} \quad (\text{Eq. 4})$$

Where n is the number of seeds, which were emergence on day D, and D is the number of days counted from the beginning of emergence

$$\text{Peak Value (PV)} = \frac{\text{Highest seed emerged}}{\text{Number of days}} \text{ (Czabator, 1962)} \quad (\text{Eq. 5})$$

$$\text{Seedling Vigour index (VI)} = (\text{Seedling length}) * GP \text{ (Abdul-Baki and Anderson 1973)} \quad (\text{Eq. 6})$$

$$\text{Mortality (\%)} = \frac{\text{number of died seedlings}}{\text{number of total seedlings}} \times 100 \text{ (Abbott, 1925)} \quad (\text{Eq. 7})$$

### 2.1 Statistical Analysis

The data were analyzed using one-way ANOVA in JMP 14 PRO (SAS Institute, 2002), a statistical program. Significant differences among mean values were compared by Duncan's test ( $P < 0.05$ ). To determine the LC<sub>50</sub> of the NaCl dose, probit analysis was performed using the online tool OPSTAT (<http://14.139.232.166/opstat/>). The LC<sub>50</sub> represents the concentration at which 50% of the plants were killed. Probit analysis is a specialized regression model for binomial response variables (Finney and Stevens, 1948).

### 3. Results

The first emergence and the highest number of emergences were observed on the second day after sowing in both the control (10.5 seeds) and 0.25 SA + 0.00 NaCl (10 seeds) treatments. The maximum number of seed emergences (20 seeds) was recorded on the seventh day after sowing in both the control and 0.25 SA + 0.00 NaCl treatments. Seedling emergence occurred in all NaCl treatments except for 200 mM NaCl. The 50 mM NaCl concentration resulted in the highest number of seedlings among the NaCl treatments. All SA treatments significantly increased seed emergence under 50 mM NaCl stress except for 0.75 SA. A sharp decline in seedling emergence was observed under 100 mM NaCl stress. At 150 mM NaCl, seed germination was severely limited, with only 10 % emergence (2 seedlings). No emergence was observed under 200 mM NaCl stress alone, but the combination with SA significantly increased emergence under 150 mM NaCl (11 seedlings with 0.5 SA) and 200 mM NaCl (4 seedlings with 0.75 SA) (Figure 1).

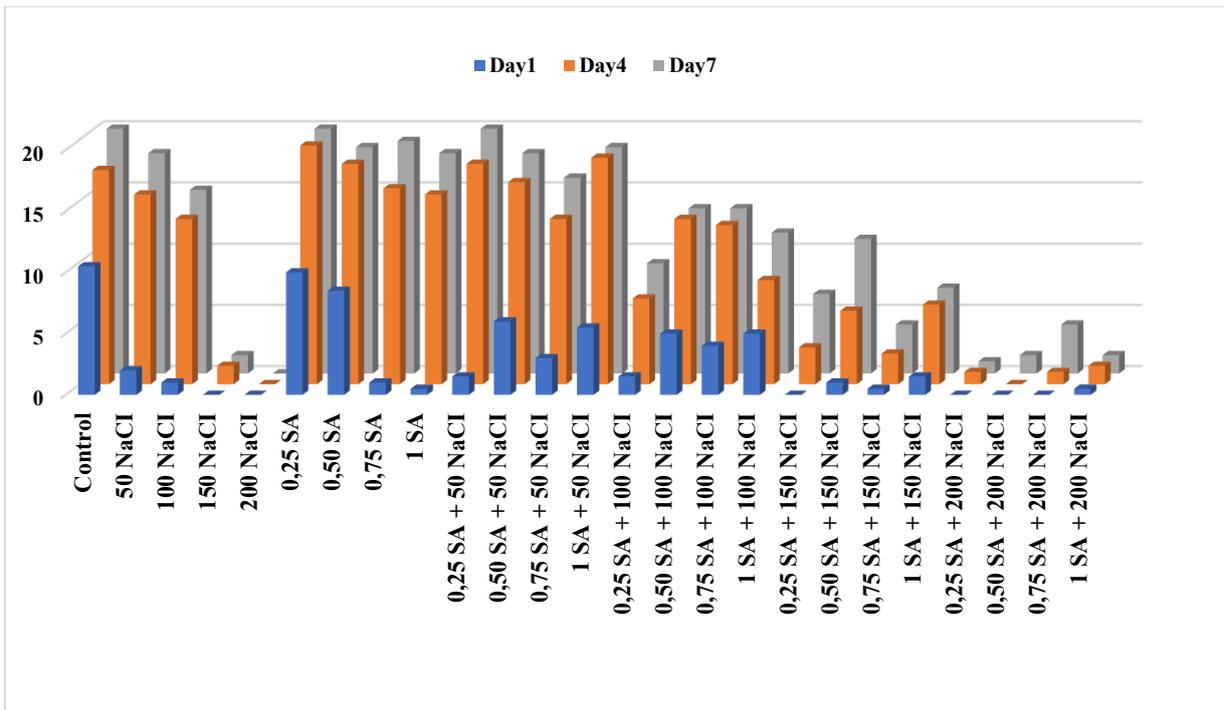


Figure 1. Seedling emergence and response of lentil seeds to NaCl, SA, and their combinations

Probit analysis revealed that the lethal concentration of salt affecting 50% of seed emergence (LC<sub>50</sub>) was 102.38 mM, corresponding to a LogLC<sub>50</sub>=2.01 (Figure 2).

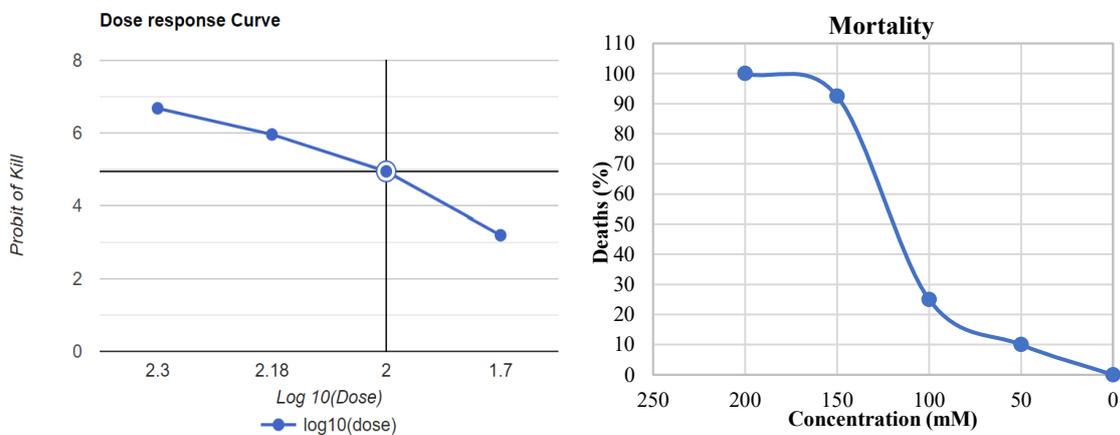


Figure 2. Probit analysis and mortality rates under different salt doses

There were statistically significant differences among the treatments in emergence energy (EE) measured on the fourth day, emergence percentage (EP), and emergence rate index (ERI) (Table 1, Figure 3).

**Table 1. Analysis of variance for EP, EE, ERI, MET, PV, seedling vigor indeks, shoot length, root length, fresh shoot weight and fresh root weight of lentil under SA, NaCl and SA+NaCl treatments**

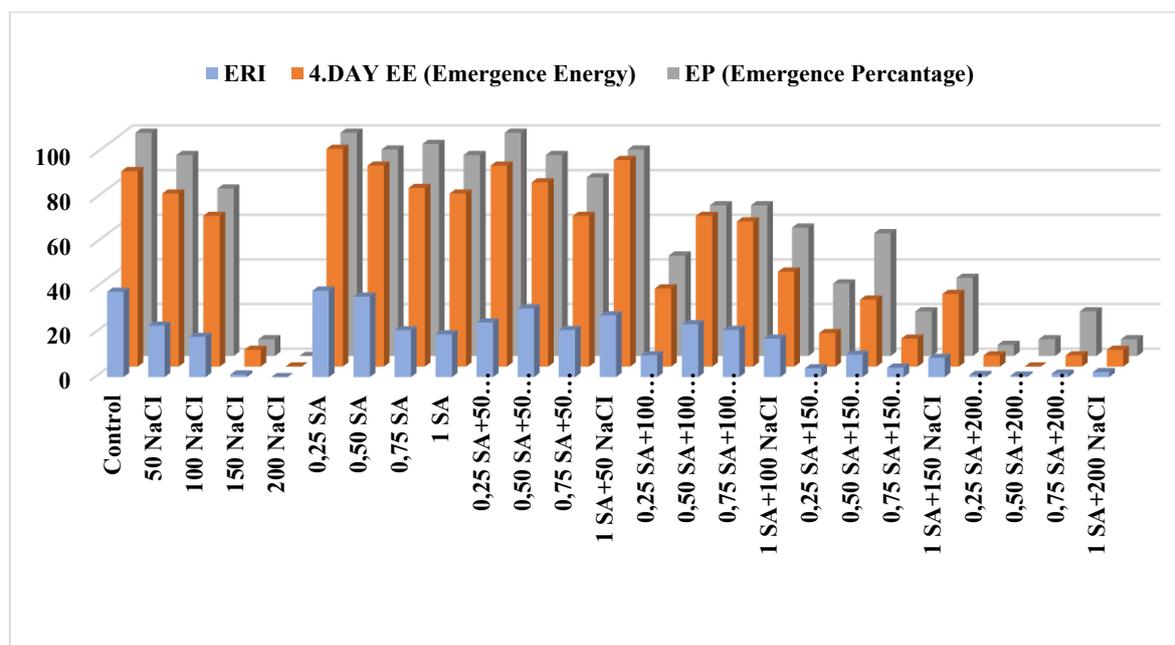
Source	DF	EP	EE	ERI	MET	PV
Treatment	23	2409.04**	2348.82**	305**	1.19 **	1.971**
Error	24	127.08	83.85	12	0.076	0.35
		Seedling vigor indeks	Shoot length	Root length	Fresh shoot weight	Fresh root weight
Treatment	23	1639586**	153.37**	16.852**	6528.47**	845.428**
Error	24	36954	1.57	0.505	154.57	44.686

\*,0,05, \*\*, 0,01 level of significance. **EP:** Emergence Percentage, **EE:** Emergence Energy, **ERI:** Emergence Rate Index, **MET:** Mean Emergence Time, **PV:** Peak Value

The highest EP (100%) was observed in the control, 0.25 SA and 0.25 SA + 50 NaCl treatments. SA seed priming significantly reduced the negative effects of NaCl on emergence. The 0.5 mM SA treatment improved EP by 55% under 150 mM NaCl stress. Furthermore, SA seed priming effectively eliminated the inhibitory effects of 200 mM NaCl on germination as EP was measured for all SA+200 mM NaCl combinations.

The highest EE on the fourth day was observed with 0.25 SA (97.5%). EE was reduced to 77.5% and 67.5% with 50 mM NaCl and 100 mM NaCl, respectively. At 150 mM NaCl, EE decreased significantly to 7.5%, and no EE was observed in 200 mM NaCl dose. All SA doses except 0.75 SA + 50 mM NaCl had a positive effect on EE. The 0.50 and 1.0 mM SA treatments had a significant effect on EE, increasing it by over 30% at 150 mM NaCl. Under 200 mM NaCl, 0.75 and 1.0 mM SA treatments increased EE by 5% and 7.5%, respectively, indicating the effect of SA in reducing salt stress.

The highest emergency rate index (ERI) of 38.71 was observed with 0.25 mM SA followed by the control (38.19) and 0.50 mM SA (36.01). All SA doses combined with 50 mM NaCl, except 0.75 mM SA, increased ERI compared to 50 mM NaCl alone. At 100 mM NaCl, SA seed priming with 0.50 mM SA and 0.75 mM SA treatments significantly increased ERI compared to 100 NaCl alone. The highest ERI under 150 mM NaCl was observed with 0.50 mM SA. Although no seed emergence was observed at 200 mM NaCl, SA seed priming had a positive effect on ERI (Figure 3).



**Figure 3. Effect of NaCl, salicylic acid (SA), and their combinations on emergence rate index**

The effects of NaCl, SA and their combinations on MET and PV of lentil seeds were statistically significant (Table 1, Figure 4). Higher salt concentrations delayed average seed emergence time. In contrast, SA seed priming reduced MET.

**Table 2. Effect of NaCl, SA and their combination treatments on peak value, shoot length, root length, fresh shoot weight, fresh root weight, and seedling vigor index**

Treatment	Peak Value	Shoot Length (cm)	Root Length (cm)	Fresh Shoot Weight (mg)	Fresh Root Weight (mg)	Seedling Vigor Index
Control	3.7 <sup>ab</sup>	23.63 <sup>a</sup>	6.91 <sup>ae</sup>	160.04 <sup>a</sup>	52.77 <sup>ab</sup>	2363.6 <sup>a</sup>
0.25 SA	4.2 <sup>ab</sup>	23.99 <sup>a</sup>	8.38 <sup>ab</sup>	163.46 <sup>a</sup>	54.08 <sup>a</sup>	2399.4 <sup>a</sup>
0.50 SA	4.5 <sup>ab</sup>	23.98 <sup>a</sup>	8.16 <sup>abc</sup>	163.76 <sup>a</sup>	43.77 <sup>ab</sup>	2218.5 <sup>a</sup>
0.75 SA	4.0 <sup>ab</sup>	23.07 <sup>a</sup>	7.59 <sup>ad</sup>	157.13 <sup>a</sup>	52.94 <sup>ab</sup>	2199.23 <sup>a</sup>
1 SA	3.3 <sup>b</sup>	22.94 <sup>a</sup>	8.67 <sup>a</sup>	159.50 <sup>a</sup>	54.20 <sup>a</sup>	2068.5 <sup>a</sup>
50 mM NaCl	3.7 <sup>ab</sup>	19.87 <sup>abc</sup>	7.10 <sup>ad</sup>	124.62 <sup>ab</sup>	52.05 <sup>ab</sup>	1777.5 <sup>abc</sup>
100 mM NaCl	4.0 <sup>ab</sup>	14.72 <sup>d</sup>	4.70 <sup>def</sup>	88.03 <sup>bc</sup>	29.47 <sup>ad</sup>	1104.4 <sup>cde</sup>
150 mM NaCl	2.5 <sup>b</sup>	3.35 <sup>g</sup>	1.83 <sup>fgh</sup>	32.00 <sup>d</sup>	14.40 <sup>cd</sup>	24.0 <sup>g</sup>
200 mM NaCl	0.0 <sup>ns</sup>	0.0 <sup>ns</sup>	0.0 <sup>ns</sup>	0.0 <sup>ns</sup>	0.0 <sup>ns</sup>	0.0 <sup>ns</sup>
0.25 SA + 50 mM NaCl	2.9 <sup>b</sup>	17.47 <sup>bcd</sup>	5.95 <sup>ae</sup>	102.53 <sup>bc</sup>	48.36 <sup>ab</sup>	1747.5 <sup>ad</sup>
0.50 SA + 50 mM NaCl	2.9 <sup>b</sup>	20.97 <sup>ab</sup>	7.50 <sup>ad</sup>	126.21 <sup>ab</sup>	55.59 <sup>a</sup>	1895.5 <sup>ab</sup>
0.75 SA + 50 mM NaCl	3.9 <sup>ab</sup>	15.07 <sup>cd</sup>	5.75 <sup>be</sup>	95.99 <sup>bc</sup>	39.29 <sup>abc</sup>	1206.0 <sup>be</sup>
1 SA + 50 mM NaCl	5.8 <sup>a</sup>	20.00 <sup>abc</sup>	7.12 <sup>ad</sup>	155.28 <sup>a</sup>	56.42 <sup>a</sup>	1851.9 <sup>abc</sup>
0.25 SA + 100 mM NaCl	3.1 <sup>b</sup>	9.14 <sup>ef</sup>	4.07 <sup>efg</sup>	56.98 <sup>cd</sup>	25.97 <sup>bcd</sup>	427.5 <sup>efg</sup>
0.50 SA + 100 mM NaCl	5.8 <sup>a</sup>	14.05 <sup>de</sup>	5.85 <sup>ad</sup>	87.85 <sup>bc</sup>	36.40 <sup>abc</sup>	934.124 <sup>ef</sup>
0.75 SA + 100 mM NaCl	4.5 <sup>ab</sup>	14.40 <sup>d</sup>	5.69 <sup>be</sup>	89.37 <sup>bc</sup>	41.72 <sup>ab</sup>	977.9 <sup>de</sup>
1 SA + 100 mM NaCl	3.1 <sup>b</sup>	17.06 <sup>bcd</sup>	5.43 <sup>cde</sup>	103.00 <sup>bc</sup>	36.10 <sup>abc</sup>	976.3 <sup>de</sup>
0.25 SA + 150 mM NaCl	2.9 <sup>b</sup>	3.00 <sup>g</sup>	1.73 <sup>gh</sup>	24.37 <sup>d</sup>	8.34 <sup>d</sup>	96.7 <sup>g</sup>
0.50 SA + 150 mM NaCl	2.9 <sup>b</sup>	3.35 <sup>g</sup>	1.33 <sup>gh</sup>	23.71 <sup>d</sup>	3.80 <sup>d</sup>	182.8 <sup>fg</sup>
0.75 SA + 150 mM NaCl	3.7 <sup>ab</sup>	2.13 <sup>g</sup>	0.82 <sup>h</sup>	22.11 <sup>d</sup>	5.44 <sup>d</sup>	43.3 <sup>g</sup>
1 SA + 150 mM NaCl	3.9 <sup>ab</sup>	4.67 <sup>fg</sup>	1.64 <sup>gh</sup>	30.75 <sup>d</sup>	8.04 <sup>d</sup>	165.3 <sup>fg</sup>
0.25 SA + 200 mM NaCl	2.5 <sup>b</sup>	1.83 <sup>g</sup>	1.33 <sup>gh</sup>	15.00 <sup>d</sup>	7.00 <sup>d</sup>	9.6 <sup>g</sup>
0.50 SA + 200 mM NaCl	3.7 <sup>ab</sup>	1.25 <sup>g</sup>	0.50 <sup>h</sup>	9.00 <sup>d</sup>	5.30 <sup>d</sup>	9.5 <sup>g</sup>
0.75 SA + 200 mM NaCl	2.9 <sup>b</sup>	1.64 <sup>g</sup>	0.85 <sup>h</sup>	23.00 <sup>d</sup>	6.29 <sup>d</sup>	33.0 <sup>g</sup>
1 SA + 200 mM NaCl	5.8 <sup>a</sup>	1.83 <sup>g</sup>	0.50 <sup>h</sup>	15.03 <sup>d</sup>	4.28 <sup>d</sup>	13.6 <sup>g</sup>

Means followed by the same letter are not significantly different ( $p < 0.05$ ), ns: non significant

Control and SA only treatments (SA + 0 NaCl) with high seed emergence, exhibited high PV values. The highest PV was observed in 0.25 SA + 50 mM NaCl and 0.50 SA + 50 mM NaCl. A high PV value was also found in 1.0 SA + 100 mM NaCl (Figure 4, Table 2).

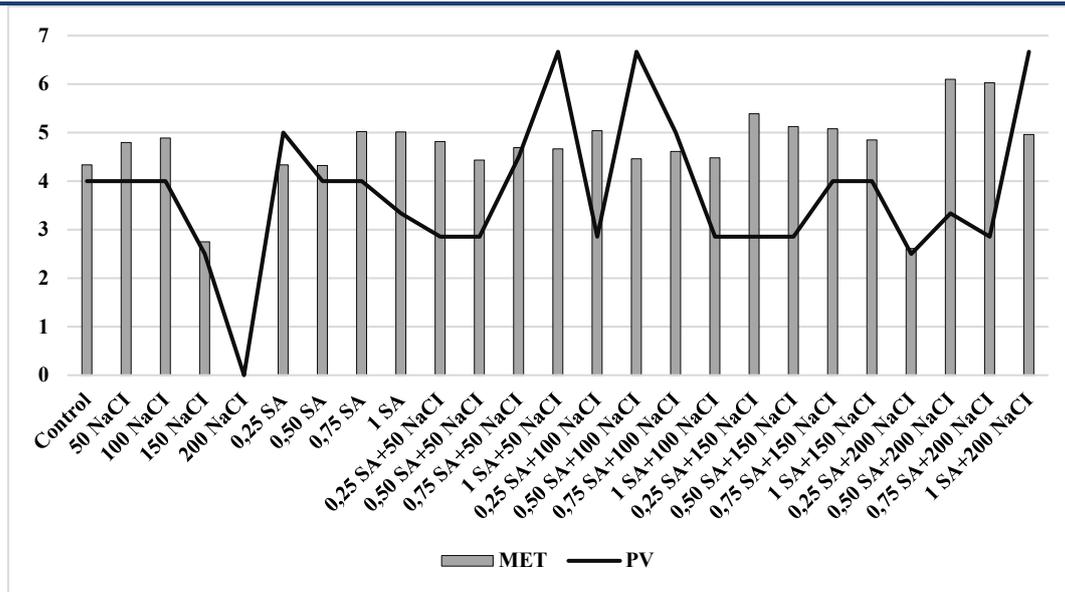


Figure 4. Effect of NaCl, SA and their combination treatments on Mean Emergence Time

The treatments SA, NaCl, and their combinations were statistically significant for shoot and root length, fresh shoot and root weight, and seedling vigor index (Table 1). The mean values were given in Table 2.

Shoot length, root length, fresh shoot weight, and fresh root weight were recorded for all treatments, except 200 mM NaCl. The highest shoot length and shoot weight values were recorded in the control, all SA doses, 50 mM NaCl, 0.50 SA + 50 mM NaCl and 1.0 SA+ 50 mM NaCl treatments, and these were statistically in the same group.

The highest root length was recorded with 1.0 mM SA, followed by all SA doses, 50 mM NaCl, 0.25 SA + 50 mM NaCl, 0.50 SA + 50 mM NaCl, and 1.0 SA+ 50 mM NaCl treatments. High root weight values were observed with 0.25 SA, 1.0 mM SA, 0.50 SA + 50 mM NaCl, and 1.0 SA+ 50 mM NaCl, followed by other SA doses, the control group, 50 mM NaCl and 100 mM NaCl with SA seed priming. In the 150 mM NaCl and 200 mM NaCl treatments, all SA seed priming resulted in lower stem and root lengths, and stem-root weight.

The seedling vigor index was recorded for all treatments except for 200 mM NaCl. High seedling vigor index values were observed in the control group and in all SA doses following 50 mM NaCl, 0.50 SA + 50 mM NaCl, 1.0 SA+ 50 mM NaCl, and 0.25 SA + 50 mM NaCl treatments, and these were statistically in the same group.

#### 4. Discussion

Salinity is a major limiting abiotic factor for the growth and yield of lentil, a salt-sensitive crop species. Salt stress negatively affects plant physiological processes, and only salt tolerant genotypes can survive severe salinity or minimize yield losses. In contrast, SA plays an important role in protecting against stress factors including salt stress, heavy metal stress, bacterial, fungal, and viral diseases (Wani et al., 2017).

Assessing seed germination and early seedling vigor can be a powerful tool to uncover genetic variation in lentil germplasm for salinity tolerance (Foti et al., 2019). In this framework, the present study aimed to determine the effects of SA as a seed priming treatment to reduce salinity stress on lentil germination and early seedling growth stages.

Our results showed that salinity directly caused delayed seedling emergence and reduced seedling growth, emergence energy, emergence percentage, emergence rate index, mean germination time, peak value, and seedling vigor index.

Increasing NaCl doses led to delayed seedling emergence, and, no emergence was observed at the highest NaCl concentration (200 mM). The results also indicate the detrimental effects of NaCl on seedling emergence. This aligns with the well-documented inhibitory effects of salinity on plant growth and development (Yuan and

Lin 2008; Lee et al., 2010; Altuner et al., 2022). In contrast, seed priming with SA eliminated the negative effects of NaCl and improved seedling emergence under low (0.25 mM SA) to moderate (0.50 mM SA) saline conditions, suggesting that SA can mitigate the negative impacts of salinity on seed germination and early seedling growth. Similar findings were reported in wheat (Dolatabadian et al., 2009) and positive effects of SA in enhancing plant tolerance to various environmental stresses, including salinity, have been documented (Al-Fraihat et al., 2023; Boukari et al., 2019; Lee et al., 2010). The results for total seedling emergence are consistent with probit analysis, which shows %50 that salinity is detrimental up to 102 mM NaCl doses. Employing probit viability analysis, proved effective in predicting germination loss in lentil seeds (Tabti et al., 2018; Tang and Sokhansanj 1993).

The observed increases in emergence rate, emergence energy, emergence percentage and emergence rate index in the control and 0.25 SA treatment groups suggest that low concentrations of SA can promote germination and emergence. However, as SA concentration increased, emergence rate decreased, indicating that higher levels of SA might have inhibitory effects on germination and emergence. These findings confirm previous reports on lentil (Kamçı et al., 2024) and alfaalfa (Boukari et al., 2019).

The findings of this study demonstrate the significant influence of SA on the MET and seedling vigor index of lentil seeds under saline conditions. The observation that the lowest and highest MET values were recorded with SA combinations under 200 NaCl conditions suggests that SA can both promote and inhibit seed germination depending on its concentration and the level of salinity stress. Similar results were reported for faba bean (Anaya et al., 2018). This might be due to the complex interactions between SA and other plant hormones and metabolic pathways involved in seed germination and stress responses (Nun et al., 2003; Vlot et al., 2009).

The study also showed the negative effects of NaCl on MET and PV. As NaCl concentration increased, MET increased and PV decreased, indicating delayed and less vigorous germination.

The significant influence of SA on the PV of lentil seedling emergence was observed under saline conditions. The lowest PV values recorded in the control and 50 mM NaCl treatments suggest that NaCl stress can significantly reduce seed vigor. SA seed priming significantly improved PV under saline conditions, suggesting that SA can mitigate the negative impacts of salinity on seed vigor. At low NaCl concentrations (50 mM), low concentrations of SA (0.25 and 0.50 mM) seed priming significantly improved PV, while higher concentrations of SA (1 mM) did not improve PV under higher NaCl concentrations (100, 150, and 200 mM). Badil et al. (2016) also reported this in sugarcane, noting that higher concentrations of SA have a less positive effect on improving salt tolerance compared to lower concentrations of SA.

As the NaCl concentration increased, shoot length, root length, fresh shoot weight, and fresh root weight decreased, and no growth was observed at the highest NaCl concentration (200 mM). Similar results have been reported, showing that increased salinity stress disrupts morphological traits such as plant height (Pandey et al., 2020). SA treatments can ameliorate the negative effects of salinity on lentil growth, and SA could be used as a potential growth regulator to improve plant salinity tolerance (Misra and Saxena, 2009). Results from this study indicated a significant influence of SA on the growth parameters of lentil seedlings under saline conditions. The results indicate that SA pre-treatment can significantly improve shoot length, root length, fresh shoot weight, and fresh root weight of lentil seedlings exposed to NaCl stress. Similar findings have also been reported in wheat (Apon et al., 2023; Afzal et al., 2006). The highest shoot length achieved with 0.25 and 0.50 SA suggests that SA at low concentrations of SA may effectively promote shoot growth. However, higher concentrations of SA indicated inhibitory effects on shoot growth.

## 5. Conclusions

Germination is the initial and essential stage in the development of healthy, high yielding crops. If germination is negatively affected by any factor, yield losses are inevitable. This study provides evidence that SA might be a useful seed priming option for improving seedling emergence, including rate index, energy, percentage, mean emergence time, peak value, and growth of lentil seeds under saline conditions. While SA priming of lentil shown promise in mitigating salt stress during early growth, further research is needed to fully understand its impact on vegetative and reproductive stages, as well as soil-plant nutrient dynamics. Soil characteristics can significantly influence the effectiveness of SA priming in alleviating salt stress. Despite these considerations, SA priming remains a promising

salinity management technique for improving lentil stand establishment in saline environments.

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#### **Ethical Statement**

There is no need to obtain permission from the ethics committee for this study.

#### **Conflicts of Interest**

We declare that there is no conflict of interest between us as the article authors.

#### **Authorship Contribution Statement**

Concept: Basdemir, F., Kamçı, G., Biçer, T. B.; Design: Basdemir, F., Kamçı, G., Biçer, T. B.; Data Collection or Processing: Basdemir, F., Kamçı, G.; Statistical Analyses Basdemir, F., Kamçı, G., Biçer, T. B.; Literature Search: Basdemir, F., Kamçı, G., Biçer, T. B.; Writing, Review and Editing: Basdemir, F., Kamçı, G., Biçer, T. B.

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