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Research Article (Araştırma Makalesi)

Seed Priming with Salicylic Acid Improves Germination and Growth of Lathyrus *sativus* L. under Salinity Stress

Sina Siavash MOGHADDAM[*1], Amir RAHIMI [2], Latifeh POURAKBAR [3], Faegheh JANGJOO[4]

[1,2,4]Urmia University, Agriculture Faculty, Agronomy Department, Urmia, Iran
[3]Urmia University, Science Faculty, Biology Department, Urmia, Iran
[1]https://orcid.org/0000-0001-8643-8991 [2]https://orcid.org/0000-0002-8200-3103 [3]https://orcid.org/0000-0002-2623-7394
[4]https://orcid.org/0000-0002-7308-1425
*Corresponding author e-mail; ss.moghaddam@urmia.ac.ir

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Abstract: Increasing the ability of crops to maintain growth and productivity under saline conditions is of paramount importance. The effect of salicylic acid (SA) priming on germination and physiological traits of *Lathyrus sativus* under salinity stress was studied in a factorial experiment based on a completely randomized design. The experimental treatments composed of SA at three levels (0, 0.1, and 0.2 mM) and NaCl salinity at three levels (0, 50, and 100 mM). The effect of salinity level and SA priming was significant on all recorded factors, except from final germination percentage. As salinity level increased, all germination and physiological traits declined compared with control, whereas the mean germination time and percentage of cell death were increased. Moreover, as salinity was intensified, the Hill reaction was decreased significantly. Salinity exhibited the strongest effects at NaCl rate of 100 mM. Seed priming with SA increased germination speed index, reduced mean germination time, and increased leaf relative water content, seedling fresh and dry weight compared with plants from non-primed seeds. Seedling vigor index was increased by 23.4% in primed seeds with 0.2 mM SA. SA priming especially at 0.2 mM rate increased the Hill reaction rate and reduced percentage of cell death. SA priming could be regarded as a practical approach to improve germination traits, seedling growth, and physiological traits of *Lathyrus sativus* L. under salinity stress conditions.

Tuzluluk Stresi altında *Lathyrus sativus* L.'nin Çimlenme ve Büyümesini İyileştirmede, Salisilik Asit ile Tohum Priming Uygulaması

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Anahtar kelimeler

Hücre ölümü,
Çimlenme parametreleri,
Hill reaksiyonu,
Salisilik asit,
Tuzluluk

Öz: Ürünlerin tuzlu koşullar altında büyümeyi ve verimliliği sürdürme kabiliyetinin arttırılması, büyük önem taşır. Salisilik asit (SA) ile priming, tuzluluk stresi altındaki *Lathyrus sativus*'un çimlenme ve fizyolojik özellikleri üzerine etkisi, tamamen tesadüfi bir faktöriyel deneme deseninde incelenmiştir. Deneme, üç seviyede SA (0, 0.1 ve 0.2 mM) ve üç seviyede (0, 50 ve 100 mM) NaCl'den oluşmuştur. Tuzluluk seviyesinin ve SA priming etkisi, nihai çimlenme yüzdesi hariç, kaydedilen tüm faktörler üzerinde önemli bulunmuştur. Tuzluluk seviyesi arttıkça, tüm çimlenme ve fizyolojik özellikler kontrole göre azalırken, ortalama çimlenme süresi ve hücre ölümü yüzdesi artmıştır. Ayrıca, tuzluluk derecesi arttıkça, Hill reaksiyonu önemli ölçüde azalmıştır. Tuzluluk, 100 mM NaCl uygulamasında en güçlü etkilerini göstermiştir. SA ile tohum priming, çimlenme hız endeksini arttırmış, ortalama çimlenme süresini azaltmış ve yaprak nispi su içeriğini arttırmış; priming yapılmayan tohumlardan elde edilen bitkilere kıyasla taze ve kuru ağırlığı

artırmıştır. 0.2 mM SA içeren priming yapılmış tohumlarda fide canlılığı indeksi % 23.4 artmıştır. Özellikle 0.2 mM SA primingi, Hill reaksiyon hızını arttırmış ve hücre ölümü yüzdesini düşürmüştür. SA priming, tuzluluk stresi koşulları altında *Lathyrus sativus* L.'nin çimlenme özelliklerini, fide büyümesini ve fizyolojik özelliklerini iyileştirmek için pratik bir yaklaşım olarak kabul edilebilir.

1. Introduction

Lathyrus sativus L., commonly known as grass pea, is an annual species of Fabaceae family that is highly adapted to adverse environments. *L. sativus* is resistant to environmental stresses and produces good grain yields under adverse climates (Cocks et al., 2000). Therefore, it is commonly grown for human consumption and livestock feed in Asia and East Africa. This species is also an excellent candidate for green manure owing to its fast vegetative growth, succulent organs, dense foliage, low C/N ratio, low water requirement (Lazanyi, 2000).

Salinity is a limiting factor of crop growth and yield in over 800 million ha of arable land (FAO, 2008) accounting for about 6% of the total global land. Salinity stress causes osmotic, ionic, and oxidative stresses, thereby generating several morphological, physiological, and chemical changes in plants and affecting photosynthesis, protein synthesis, lipid metabolism, and energy generation (Parida et al., 2005). Salinity stress can affect all physiological processes from germination to plant development. Photosynthesis is a key pathway in physiology of plants that is severely influenced by salinity. Previous research showed that NaCl significantly reduced growth parameters, Rubisco activity, photosynthetic efficiency and pigments, as well as sugar contents in maize while the effects of NaCl on the previous parameters were increased with NaCl concentrations (Khodary, 2004). Reduced activity of the Hill reaction was also observed in salt-stressed chloroplasts in wheat (El-Shintinawy, 2000), in cowpea (*Vigna sinensis*) (El-Shahaby et al., 2003), and in maize (El-Shahaby et al., 2003; Zeid, 2009).

Salinity stress can also affect other plant physiological processes. Abscisic acid built in response to salinity induces stomatal closure and, thereby, limits CO_2 inflow to the plant (Leung et al., 1994). Salinity can decrease plant protein content by impairing protein synthesis and increasing the activity of protein hydrolyzing enzymes or may increase plant protein content by synthesizing new proteins and/or reducing proteolytic enzymes (Dubey, 1999). The harmful impacts of salinity stress on plants can be caused by osmotic and ionic stresses (Munns and Tester, 2008) that result in metabolic disorders in cells manifested in the build-up of reactive oxygen species (ROS), namely superoxide (O_2), hydrogen peroxide (H_2O_2), and hydroxyl radical (OH^-) (Halliwell, 2006). The main site of ROS generation in leaves under salinity stress conditions are chloroplasts (Cavalcanti et al., 2007). Under the influence of environmental stresses, the closure of the stomata, and the reduction of gas exchanges, as well as the continued absorption of light energy, result in disturbances in the electron transport chain in the reaction centers and the reduction of the quantum yield of photosystem II (Maxwell and Johnson, 2000). The Hill reaction is described as the photoreduction of an electron acceptor by the hydrogens of water, with the evolution of oxygen. The ultimate electron acceptor is NADP+. The Hill reaction can be measured in isolated chloroplasts. One of the most perceptible responses of plants to environmental stresses is the loss of photosynthesis due to impaired photosystem II activity (Andrews et al., 1995).

One of the approaches to alleviate the impacts of salinity stress is seed priming. Priming refers to the pre-sowing treatment of seeds, by which seeds pass through initial stages of germination, but radicles do not emerge due to the low amount of imbibed water (Nascimento and Aragao, 2004; Ibrahim, 2016). This technique has a lot of advantages, such as fast and uniform emergence of seedlings, maturity progress, wider thermal range for germination, regeneration of damaged cells, reduction of barriers to embryo growth, quantitative and qualitative improvement of protein synthesis, seed dormancy breaking, improvement of environmental stress resistance during sowing, and eventually enhancement of plant growth and development (Ghasemi-Golazani et al., 2010; Ibrahim, 2016).

Salicylic acid (SA) is a plant growth regulator that may have desirable effects on seedling growth and development (Kerantev et al., 2008; Khan et al., 2015). SA, or ortho hydroxyl benzoic acid, belongs to a group of phenolic compounds and is known as an important molecule that regulates plant reaction to environmental stresses (Senarajna et al., 2000). The significance of salicylic acid (SA) has been increasingly recognized in improved plant abiotic stress-tolerance via SA-mediated control of

major plant-metabolic processes (Khan et al., 2015). In fact, SA alleviates salinity effects via increasing the level of hormones like auxins and cytokinins, e.g., it prevented any decrease in indoleacetic acid (IAA) and cytokinin contents and thus reduced stress-induced inhibition of plant growth (Shakirova et al., 2003), reducing the uptake of toxic ions, and contributing to membrane stability (El-Tayeb,2005; Samea-Andabjadid et al., 2018). The exogenous application of salicylic acid prevented the lowering of IAA and cytokinin levels in salinity stressed wheat plants resulting in the betterment of cell division in root apical meristem, thereby increasing growth and productivity of plants (Shakirova et al., 2003). When the seed is subjected to salt stress, the adsorbed SA (during the preparation) rapidly attaches to the glucose and converts to salicylic acid ß-glucoside (SAG).

The enzyme that converts SA into SAG is acid salicylic glucosyl transferase. The produced compounds play an important role in expressing the genes associated with increasing seed resistance to salt stress. Among the RS20-RS19-RS17-RSS (genes that increase the tolerance to salinity by counteracting the toxicity of salt ions) and the PST1 gene increases salinity tolerance by scavenging the active oxygen species that causes oxidative stress (El-Tayeb, 2005).Exogenous application of SA to soybeans and corn (Khan et al., 2003) increased plant growth and caused positive changes under salinity conditions, such as increased stomatal conductance, transpiration, and photosynthetic rates in both soybean and corn. However, research data on the effect of seed priming with SA on seed germination and growth of *L. sativus* seedlings do not exist.

Thus, the research question of this study was: can seed priming with SA improve germination and early growth of *L. sativus* under salt stress? Regarding the importance of *Lathyrus sativus* L in the field of medicine, soil improvement and green manure, this research was conducted to investigate the positive effect of salicylic acid for mitigating the salinity stress on germination, physiological traits and growth parameters of *Lathyrus sativus* L. Since salinity has been a major agriculture problem in West Azarbaijan Province in the region of Urmia Lake, the present study outcomes can be useful in combating with abiotic stresses, including salinity, towards the production of *Lathyrus sativus* L in this region.

2. Material and Methods

2.1. Germination and growth parameters

The study was carried out in the laboratory of Agriculture and Biology Department of Urmia University in 2016 as a factorial experiment based on a completely randomized design with three replications. The treatments composed of seed priming with salicylic acid (SA) at three levels (0, 0.1, and 0.2 mM) and salinity with sodium chloride (NaCl) at three levels (0, 50, and 100 mM). The seeds of *L. sativus* were first disinfected with sodium hypochlorite 5% for 2 min, and then rinsed with distilled water. For priming, seeds were soaked in SA solution at pre-determined concentrations (0.1 and 0.2 mM) in darkness at 25°C for 8 h. Then, they were air-dried at room temperature for 24 h to reduce surplus moisture. For the germination assay, 100 seeds were placed between two filter papers in Petri dishes with a diameter of 9 cm. The Petri dishes were placed in a germinator at 25°C.

In order to obtain the necessary moisture, seeds in Petri dish were watered every other day with distilled water and sodium chloride solution at concentrations of 0, 50 and 100 mM. To assess germination parameters (final germination percentage, mean germination time and germination speed index: as showed in formula), the seeds were counted at a certain hour every day until the number of germinated seeds reached a plateau for three consecutive days. The criterion for seed germination was the emergence of radicle at a length > 2 mm. On day seven, five seedlings were selected from each replication to measure the length of seedling, radicle, and plumule as well as seedling fresh and dry weight. After seedling fresh weight was recorded, the samples were oven-dried at 72°C for 48 h and the average of five samples was determined.

The remaining seedlings in the Petri dishes were used to explore the physiological parameters of *L. sativus,* including leaf relative water content, Hill reaction, cell death, as well as post-harvest length and weight parameters. Four seedlings were selected from each replication. Seedlings were planted in perlite-containing pots and were placed in a growth chamber at light/dark regime of 16/8 h. To provide the required moisture for seedlings in the pot, also every other day, distilled water and Hoagland solution containing, 0, 50 and 100 mM sodium chloride was used. To measure the post-harvest length and weight

parameters, root and stem length, root and stem fresh weight, and root and stem dry weight (oven-dried at 72°C for 48 hours) were recorded after 15 days.

2.2. Relative water content

To determine the relative water content of leaves, a quantity of 0.2 g was taken from a developed leaf in each replication, 1 cm^2 was detached from the middle part of its lamina, and then leaf discs were placed in a capped Petri dish containing distilled water, which was placed in darkness at 4°C for 16 h. Then, the leaves were taken out of the distilled water and after letting surplus moisture go, they were placed between two filter papers and their saturated weight was measured immediately. Next, leaves were oven-dried at 70°C for 48 hours to determine dry weight. Leaf relative water content (RWC) was calculated according to the following Formula 5.

2.3. Hill reaction

Hill reaction was measured according to Patsikka et al. (2001). A quantity of 0.2 g of fresh leaf tissue was weighed. Then, it was crushed with 3 mL of 50 mM phosphate buffer with pH 7 cooled down by freezing. The filtrated extract was centrifuged at 10,000 rpm for 2 min and the supernatant was removed. Then, 3 mL of cold phosphate buffer (50 mM, pH 7) was added to the deposit of the centrifuge and the deposit was suspended with a paintbrush. After that, 0.5 mL of the solution was taken and was added with 2 mL of cold phosphate buffer (50 mM, pH 7) and 0.2 mL of dichlorophenolindophenol (DCPIP). Immediately, its absorption was measured at 550 nm with a spectrophotometer. Then, the tubes were exposed to a 150-W lamp for 20 seconds and absorption was measured again. This practice was reiterated for 5 min until we had T = 100. The extent of DCPIP reduction was measured as a percentage of the non-SA-treated control.

2. 4. Cell death measurement

Cell death is a criterion showing the extent of damage to the cell membrane and was measured according to Baker and Monck (1994) using the absorption of the Evans blue reagent. To measure cell death, three 1-cm pieces of root ends were placed in Evans blue reagent 0.025% in water for 30 min. Then, the pieces were rinsed with water for 15 min. After that, the samples were crushed in 1 mL of 50% methanol solution. The extract was placed in a bain-marie at 50 °C for 15 min, and then, it was centrifuged at 14,000 rpm for 15 min. Afterwards, the absorption was measured at 600 nm with a spectrophotometer and finally, cell death was stated as a percentage of control.

$$\text{Final germination percentage} = \frac{\text{Number of germinated seeds}}{\text{Total number of seeds}} \quad (1)$$

Germination speed index was calculated according to AOSA (1983) formula as follow:

$$\text{GSI} = \frac{\text{No. germinated seeds at first counting}}{\text{First day of counting}} + \cdots + \frac{\text{No. germinated seeds at final counting}}{\text{Final day of counting}} \quad (2)$$

Mean germination time was obtained according to Ellis and Roberts (1981) formula as follow:

$$\text{MGT} = \frac{\sum D_n}{\sum n} \quad (3)$$

Seedling vigor index was calculated according to Brancalion et al. (2008) formula as follow:

$$\text{SVI} = \frac{\text{Seedling length (cm)} \times \text{germination percentage}}{100} \quad (4)$$

Relative water content was estimated by Weatherley (1950) formula as follow:

$$RWC = \frac{\text{Fresh weight - dry weight}}{\text{Saturated weight - dry weight}} \times 100 \quad (5)$$

Data were analyzed with the MSTAT-C software package. Treatment means were compared using Duncan's multiple range test at $P < 0.05$. Also, all graphs were drawn in MS-Excel software package.

3. Results

3.1. Germination and growth parameters

Final germination percentage (FGP) was not affected by salinity level and SA priming. However, germination speed index (GSI) was significantly influenced by salinity level, SA, and their interaction. The salinity levels of 50 and 100 mM reduced GSI significantly (16.65 and 18.55 (% /day) respectively) compared with control (28.00 % /day) (Figure 1). Priming with 0.2 mM SA improved GSI both under control and salinity conditions, but with 0.1 mM SA only under stress conditions.

Salt stress effect led to the increase of MGT from 2.173 (day) in control to 3.229 and 2.773 (day) in 100 and 50 mM of NaCl respectively. The priming with both SA concentrations reduced this parameter significantly, but in a similar extent (Figure 2).

Salinity and SA priming affected radicle, plumule, and seedling length, without significant interaction. Both levels of salinity resulted in reduction of radicle, plumule, and seedling growth. The reduction of plumule and seedling growth was higher at higher salinity levels. However, growth reduction of radicle was the same at both salinity levels. SA-primed seeds had longer radicles, plumules, and seedlings than non-primed seeds. However, the observed effect did not depend of SA concentration (Table 3).

Salinity stress reduced seedling vigor significantly in a dose dependent manner. Higher loss in seedling vigor occurred at higher salinity level (100 mM). Seeds primed with both SA concentrations caused similar improvement of seedling vigor (Table 3).

Both salinity level reduced seedling fresh weight as compared with control. Higher reduction was shown at 100 mM than 50 mM NaCl. In contrast, SA priming enhanced seedling fresh weight as compared with non-primed seeds. The highest effect was observed when 0.2 mM SA was applied (Table 3). However, the low rates of SA tested in this study were not sufficient to improve fresh weight under salinity stress. Salinity stress and SA priming influenced seedling dry weight significantly, without significant interaction. With the increase in salinity level, seedling dry weight was reduced. NaCl level of 100 mM resulted in loss of seedling dry weight from 16.2 mg in control to 12.76 mg in stress combination. The application of SA (0.2 and 0.1 mM) improved seedling dry weight (15.67 and 14.96 mg, respectively) versus control (Table 3), but the effect was significant only for SA at 0.2 mM.

3.2. Relative water content

Analysis of variance revealed the significant effect of salinity, SA priming, and their interaction on leaf relative water content (RWC) at $P < 0.01$ (Table 1). Under control and under stress conditions both SA concentrations caused the increase of RWC (Figure 3).

3.3. Hill reaction and cell death

There were significant differences in the Hill reaction between salinity stress levels and SA rates. As salinity was intensified, the Hill reaction was decreased significantly. The lowest rate of 54.84% was obtained at 100 mM of salinity. The application of SA improved the Hill reaction rate. The highest effect of improvement was shown in plants grown from seeds primed with 0.2 mM SA (Figure 4). Increased salinity level resulted in a significant increase of cell death percentage. The application of SA resulted in a decrease of cell death percentage (Figure 5).

3.4. Post germination test

Salinity level and SA priming influenced root and stem length, root fresh and dry weight significantly ($P < 0.01$), while the interaction of salinity with SA was insignificant for these traits (Table 2). The smallest root length (22.22 cm) and stem length (31.46 cm) were obtained from 100 mM salinity, while the greatest root length (34.35 cm) and the greatest stem length (52.82 cm) was obtained from SA 0.2 mM (Table 3). Different SA levels varied significantly in their effects, so that SA concentration of 0.2 mM had the greatest impact considering both these traits (root and stem length) (Table 3). Salinity resulted in significant loss of root fresh weight, and consequently its dry weight, as compared with control (Table 3). Salinity level of 100 mM caused root fresh weight to decrease from 0.1848 g to 0.1087 g in control. Also, root dry weight at this salinity level was decreased from 0.04956 g to 0.01844 g in control. On the other hand, SA primed seeds favored root fresh and dry weight. Root fresh weight differed significantly in plants treated with 0.1 mM SA from those treated with 0.2 mM SA. Indeed, plants treated with 0.2 mM SA exhibited the greatest root fresh weight. Plants treated with 0.1 mM SA and those treated with 0.2 mM SA had similar root dry weights (0.03333 and 0.04467 g, respectively) (Table 3).

Stem fresh and dry weights were influenced significantly by salinity level, SA priming, and their interaction (Table 2). When there was no salinity stress, SA application improved stem fresh and dry weight. This increase was the greatest at SA rate of 0.2 mM, resulting in stem fresh and dry weights of 1.207 and 0.1750 g, respectively. At salinity level of 50 mM NaCl, while 0.2 mM SA improved these traits (stem fresh and dry weight), it did not significantly differ from SA concentration of 0.1 mM. As salinity level was increased to 100 mM NaCl, only the higher SA concentration (0.2 mM) improved stem fresh and dry weight to 0.4067 and 0.03867 g, respectively (Figure 6 & 7).

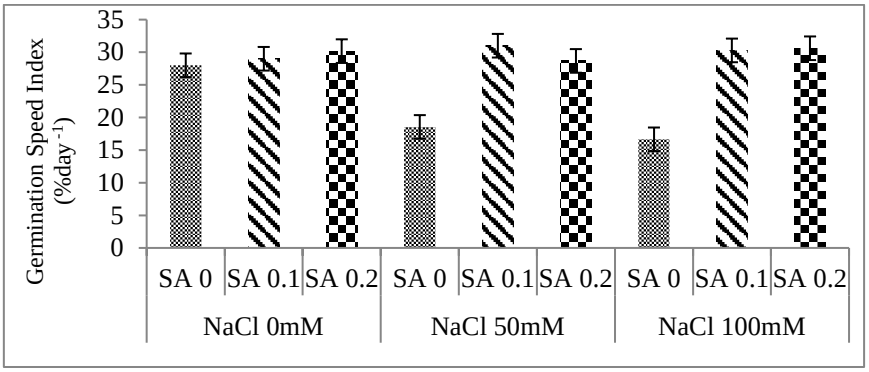

Figure 1. Values of germination speed index among treatments (different letters indicate significant differences at $P < 0.05$).

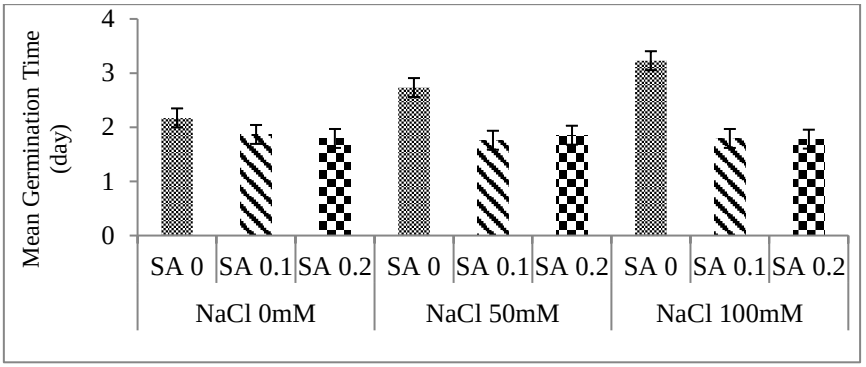

Figure 2. Values of mean germination time among treatments (different letters indicate significant differences at $P < 0.05$).

Table 1. Analysis of variance for the effect of salinity and salicylic acid on germination and physiological traits of *L. sativus*.

Treatment	df	FGP (%)	MGT (day)	GSI (% /day)	RL (cm)	PL (cm)	SL (cm)	SVI	RWC (%)	SFW (mg)	SDW (mg)
Salinity	2	0.037^{ns}	0.236^{**}	28.820^{*}	7.343^{*}	12.305^{**}	37.622^{**}	96814.35^{**}	19.427^{**}	1097.939^{**}	27.898^{**}
SA	2	0.037^{ns}	2.526^{**}	237.081^{**}	3.291^{*}	7.131^{**}	17.83^{**}	46125.17^{**}	265.653^{**}	3285.495^{**}	11.591^{*}
Salinity × SA	4	0.037^{ns}	0.309^{**}	44.062^{**}	0.070^{ns}	0.592^{ns}	0.955^{ns}	2339.492^{ns}	4.304^{**}	174.024^{ns}	3.816^{ns}
Error	18	0.037	0.01	7.115	0.622	0.307	0.838	2062.644	0.513	117.286	2.859
CV%		0.39	4.72	9.88	13.7	12.93	9.13	9.05	0.99	10.28	11.5

** Significant at P < 0.01; * significant at P < 0.05; ns: not significant.
FGP: final germination percentage, MGT: mean germination time, GSI: germination speed index, RL: radicle length, PL: plumule length, SL: seedling length, SVI: seedling vigor index, RWC: relative water content SFW seedling fresh weight, SDW: seedling dry weight)

Table 2. Analysis of variance for the effect of salinity and salicylic acid on germination and physiological traits of *L. sativus*.

Treatment	df	Root LE (cm)	Root FW (g)	Root DW (g)	Stem LE (cm)	Stem FW (g)	Stem DW (g)
Salinity	2	380.205^{**}	0.013^{**}	0.002^{**}	1639.009^{**}	1.126^{**}	0.025^{**}
SA	2	290.145^{**}	0.005^{**}	0.002^{**}	463.742^{**}	0.263^{**}	0.004^{**}
Salinity × SA	4	4.431^{ns}	0^{ns}	0^{ns}	0.924^{ns}	0.022^{*}	0.001^{**}
Error	18	2.837	0	0	5.801	0.006	0
CV%		5.88	8.08	12.48	5.32	14.14	17.04

** Significant at P < 0.01, * significant at P < 0.05, ns: not sginificant.
(Root LE: root length; Stem LE: stem length; Root FW: root fresh weight; Root DW: root dry weight).

Table 3. Mean comparision of the main effect of salinity and SA on germination and physiological parameters of *L. sativus*.

Treatment	RL (cm)	PL (cm)	SL (cm)	SVI	SFW (mg)	SDW (mg)	Root LE (cm)	Stem LE (cm)	Root FW (g)	Root DW (g)
Salinity										
0 mM	6.727 a	5.476 a	12.16 a	610.1 a	117.8 a	16.2 a	35.21 a	58.42 a	0.1848 a	0.04956 a
50 mM	5.602 b	4.253 b	9.856 b	492.8 b	101.5 b	15.11 a	28.52 b	46.04 b	0.1400 b	0.02689 b
100 mM	4.940 b	3.138 c	8.080 c	403.3 c	96.69 b	12.76 b	22.22 c	31.46 c	0.1087 c	0.01844 b
SA										
0 mM	5.162 b	3.264 b	8.427 b	420.6 b	84.42 b	13.44 b	22.99 c	38.52 c	0.1208 c	0.01689 b
0.1 mM	5.736 b	4.873 a	10.61 a	530.4 a	109.6 a	14.96 ab	28.61 b	44.58 b	0.1429 b	0.03333 a
0.2 mM	6.371 a	4.729 a	11.06 a	555.1 a	121.9 a	15.67 a	34.35 a	52.82 a	0.1698 a	0.04467 a

Different letters indicate significant differences at P < 0.05.

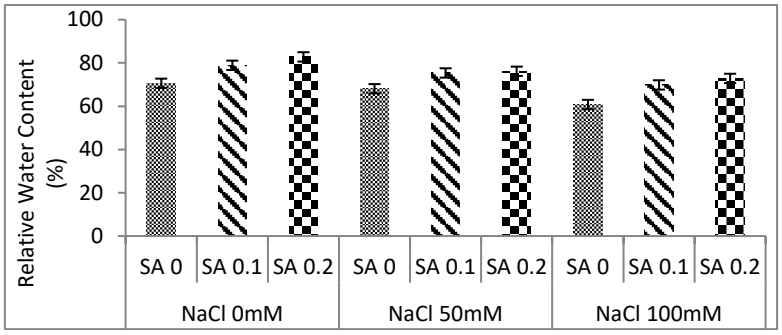

Figure 3. Values of relative water content among treatments (different letters indicate significant differences at $P < 0.05$).

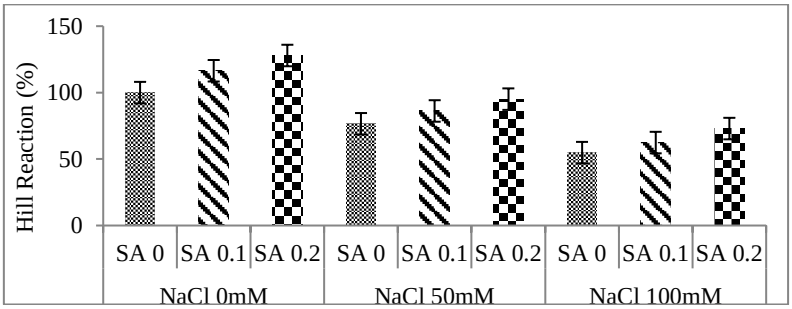

Figure 4. Values of Hill reaction speed (%) among treatments (different letters indicate significant differences at $P < 0.05$).

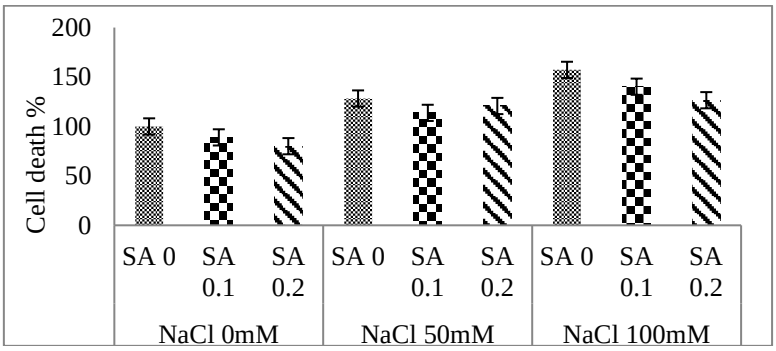

Figure 5. Values of cell death (%) among treatments (different letters indicate significant differences at $P < 0.05$).

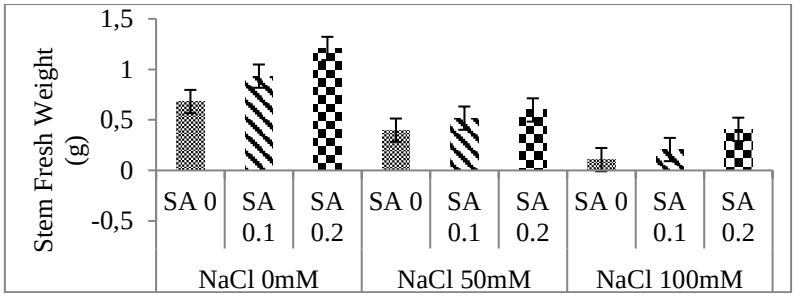

Figure 6. Stem fresh weight among treatments (different letters indicate significant differences at $P < 0.05$).

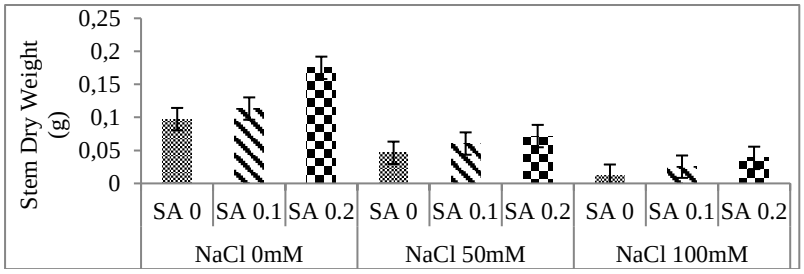

Figure 7. Stem dry weight among treatments (different letters indicate significant differences at $P < 0.05$).

4. Discussion and Conclusion

The study showed SA has significant effect on germination indices, growth and physiological parameters under salinity stress. SA increased GSI and decreased MGT on salt stress. The loss of GSI under salt stress could be related to the negative impact of low water potential on water uptake as well as toxic effect of ions (Na and Cl) on biochemical processes and catabolic (enzymatic hydrolysis of seed storage materials) and anabolic (generation of new tissues by materials hydrolyzed at the first step) stages of germination (Shamsadin Saeid et al., 2008). However, the positive effect of SA on GSI could be due to reducing oxidative damage under high salinity (Lee et al., 2010). It is possible that SA stimulates the seed germination via bio-synthesis of gibberellic acid and acts as thermogene inducers (Shah, 2003). The reduced MGT in SA primed seeds may be attributed to the increased water uptake and promotion of the biological processes during germination provoked by SA in those seeds (Debez et al., 2018), and the reduced accumulation of Na^+ and Cl^- ions by SA application (Jini and Joseph, 2017). Entesari et al. (2012) reported the same effect on the mung bean grown under salinity stress and primed with SA. Hamid et al. (2010) reported that SA priming of wheat seeds under salinity stress resulted in the production of more vigorous and larger seedlings and enhanced chlorophyll, dissolved sugars and proteins content of the plant. The positive effect of SA treatment on seedling growth under salinity stress could probably be caused by the involvement of this growth regulator in cell division (Shakirova et al., 2003; Dolatabadian et al., 2009). Increased cell division of apical meristem of initial roots, which in turn resulted in an increased level of elongation was shown in SA treated wheat (Shakirova et al., 2003). In this study priming with SA was decreased negative effects of salinity on root and shoot length, fresh and dry weight. Delavari et al. (2014) founded germination, length of root and shoot, fresh and dry weight decreased under salinity but pre-treatment with SA improved them.SA treatment alleviates osmotic stress and allows better water uptake. The mechanism by which SA increases the growth of the root and shoots in some plants are still unknown, but it is possible that SA adjusts elongation and cell division with other substances such as auxin (Nourafcan, 2015). The mechanism by which SA improves root and shoot growth of some plants is not well-understood, but SA is likely to regulate cell elongation and division through the aid of other compounds, like auxin (Shakirova et al., 2003). It was also shown that SA hinders the oxidation of auxin (Farkhonded et al., 2012). So, it seems that the increased level of seedling dry weight is related to the SA-induced increase in root and stem length through different ways.

It has been reported that the desirable effect of SA on seedling vigor under no stress conditions is accompanied with increased level of IAA and ABA (Shakirova et al., 2003). As mentioned above, the low effect of SA on seedling vigor under salinity stress could be attributed to the low rates of SA tested in this study. The mechanism by which SA improves root and shoot growth is not well-understood. However, Fariduddin et al. (2003) stated that SA inhibits auxin oxidation, whose elevated content increases net photosynthetic rate in leaf. Since salinity stress reduces cell division, it seems that the increase in seedling fresh weight associated with increasing root and shoots length that is affected by salicylic acid.

According to our results salinity reduced relative water content and SA improved that. Likewise, Agarwal et al. (2005) concluded that foliar application of SA improved RWC of leaves in wheat plants. Singh et al. (2015) reported the exogenous application of SA reduced the growth inhibition of plants caused by NaCl, and increased leaf relative water contents. The increased level of leaf RWC by SA can be related to the role of SA in increasing the capability of the antioxidant defensive system, alleviating stress, and increasing membrane stability and cohesion as well as osmotic adjustment through the

increased level of potassium as a crucial element in maintaining cell turgor (Bandurska and Stroinski, 2005; Korkmaz et al., 2007).

Reduced activity of the Hill reaction was also observed in salt-stressed chloroplasts of various plant species (El-Shintinawy, 2000; El-Shahaby et al., 2003; Zeid, 2009). Proteins D1 and D2 of the photosystem II are also damaged under stress conditions. These proteins are the main components of the photosystem II and their degradation results in photoinhibition (Bissati et al., 2000; Kruk et al., 2005). SA by retention and accelerating the repair of protein D1 and D2, also induction of protein kinases and reversible phosphorylation of proteins, reduce the severity of damage under stress conditions (Hui-Jie et al., 2011). The enhancement of Hill reaction activity with SA priming may be due to increased synthesis of chlorophyll content along with acceleration of photosynthesis performance and carbohydrate metabolism (Khodary, 2004). Ervin et al. (2005) Observed increased activity of superoxide dismutase after treatment of plants with SA and argued that SA activates the antioxidant system and transmits the message to enhance the efficiency of the photosystems II. Bhattacharjee and Mukherjee (2002) reported that salinity induces oxidative stress causing membrane degradation. Salinity stress induces oxidative stress and, thereby, escalates the generation and accumulation of active radicals. This effect, in turn, oxidizes proteins and lipids and ruins membrane structure (Molassiotis et al., 2006). SA contributes to membrane maintenance by influencing polyamines, like putrescine, spermine, and spermidine, and generating membrane-stable complexes (Nemeth et al., 2002). This response may act through reducing the amount of hydrogen peroxide (Borsani et al., 2001), which is a toxic molecule in germinating seeds (Wojtyla et al., 2016).

This study assessed the effect of salinity stress and SA seed priming on *L. sativus* germination parameters and early growth, for which research data do not exist in the literature. Salinity stress (50 mM and 100 mM NaCl) resulted in significant decline of germination parameters, seedling vigor, and seedling growth, whereas the application of SA priming alleviated some negative effects of salinity on germination and related traits and improved most growth and physiological traits of *L. sativus*. According to the results, it can be concluded that the priming of *L. sativus* seeds with SA can alleviate the effect of salinity and improve the resistance of seedlings to salinity stress.

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