

Salicylic acid-enhanced morphological and physiological responses in chickpea (*Cicer arietinum*) under water deficit stress

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Abstract

In order to evaluate the effect of foliar application of salicylic acid (SA) on morphological and physiological responses in chickpea under water deficit stress, a field experiment with four chickpea genotypes at two different irrigation regimes were carried out. Foliar spraying of the plants with distilled water (control) and salicylic acid treatments (0.01, 0.1 and 1 mM) were performed four times at 20, 30, 40 and 50 days after sowing. Water deficit stress significantly reduced yield and yield components. Nevertheless, exogenous SA application significantly improved these attributes under water stress conditions. However, drought stress increased leaf proline and soluble sugar concentration and it was further increased by exogenous application of SA. Water stress significantly reduced leaf chlorophyll a, chlorophyll b and carotenoid concentrations as compared to well-watered conditions and these were further increased by exogenous application of SA. Exogenously applied SA inhibited or promoted morphological and physiological changes in plants. SA at a concentration of 0.01 mM negatively affected seed yield and its components, while most efficient doses of SA for improving physiological these attributes were 0.1 and 1 mM. The results suggest that application of exogenous SA could help to reduce the adverse effects of drought stress and might have a key role in providing tolerance to stress by promoting growth and accumulation of proline, soluble sugars and photosynthetic pigments in plant leaves.

Key words: chickpea, chlorophyll, drought, morphology, salicylic acid, physiology, yield.

Abbreviations: SA, salicylic acid.

Introduction

Chickpea (*Cicer arietinum* L.) is one of the most important pulse crops in world, it is valued for nutritive seeds with high protein content (25.3 to 28.9%; Mafakheri et al. 2011). In Iran, chickpea production areas occur only in arid and semi-arid zones and usually chickpea plants are exposed to increasing water deficit during flowering and maturity stages (Talebi et al. 2013). In general, drought is one of the main environmental stresses that adversely affect plant growth, metabolism and yield. Water is a scarce resource in Iran due to the high variability of rainfall. The effects of water stress on plants depend on the timing, duration and magnitude of the deficits. Plants have developed wide morphological, physiological and biochemical responses for environmental stresses such as drought (Talebi et al. 2013). Important mechanisms involved in adaptation of plants to drought stress are changes in leaf water potential and photosynthetic activity, pigment concentration, as well as osmotic adjustment (Hayat et al. 2010; Talebi et al. 2013).

Different methods are practiced in agriculture to enable plants to cope with abiotic and biotic stresses. Seed priming and exogenous foliar application of compatible solutes like sugar polyols, amino acids, proline, glycinebetaine as

well as salicylic acid have been considered in recent years (Hussain et al. 2008; Hayat et al. 2012). Salicylic acid (SA) is an endogenous plant growth regulator that can play an important role in abiotic stress tolerance. Considerable interests have focused on SA due to its ability to induce a protective effect on plants under stress (Hayat et al. 2008; Hussain et al. 2008; Hayat et al. 2010). Several studies have supported the SA-induced increase in the resistance of plants to drought stress (Hayat et al. 2008; Hussain et al. 2008; Pal et al. 2014). A possible mechanism for the role of SA in enhancing plant responses to abiotic stress is that SA participates in the development of stress symptoms, but it is also needed for the acclimation process and the induction of stress tolerance (Horvath et al. 2007). The exact mechanism of effect of SA in the case of drought tolerance is still unclear. SA potentiates the generation of reactive oxygen species in photosynthetic tissues of *Arabidopsis thaliana* during salt and osmotic stress (Borsani et al. 2001), SA is involved in activation of synthesis of carotenoids, xanthophylls, stimulation of net photosynthetic rate, increase in internal CO₂ concentration, water use efficiency, stomatal conductance, transpiration, and also can enhance the rate of deepoxidation with a concomitant decrease in chlorophyll pigments and chlorophyll *a/b* ratio under water

stress condition in different plant species (Fariduddin et al. 2003; Khan et al. 2003).

It has been reported that SA can provide protection against some types of abiotic stress such as heat in mustard seedlings (Dat et al. 1998), chilling damage in different plants (Kang, Saltveit 2002), heavy metal stress in barley seedlings (Metwally et al. 2003), and drought stress in garlic (Bideshki, Arvin 2010) and wheat (Bezrukova et al. 2001). Most of the previous studies have been conducted in controlled conditions and there have been no reports on the effect of SA application in field experiments. Therefore, the objective of present study was to evaluate effect of different concentrations of SA applied as foliar spray on morphological responses in long-term drought stress in chickpea under field conditions.

Materials and methods

Plants and cultivation conditions

In order to evaluate the effects of salicylic acid (SA) application and irrigation regimes on morphological and physiological responses in chickpea (*Cicer arietinum* L.), a field experiment was conducted at the experimental farm of agriculture faculty, Islamic Azad University, Sanandaj Branch, Iran (35°10'N, 46°59'E; 1393 m above sea level) in spring 2013. Sanandaj is located in north-west of Iran and has a mean annual temperature 12 °C and annual rainfall of 512 mm. Pattern of monthly rainfall (mm) and temperature (°C) during the crop season is presented in Fig 1. Some of the soil physicochemical characteristics were: sand 25.2%, silt 29.6%, clay 45.2%, pH 7.6, organic carbon 0.61%, electrical conductivity 0.49 dS m⁻¹, and available P and K concentrations 8.04 and 299 mg L⁻¹, respectively. The certified seeds of two Kabuli type chickpea cultivars ('ILC482' and 'Jam') and two Desi type cultivars ('Pirouz' and 'Kaka') were purchased from Seed and Plant Improvement Institute, Karaj, Iran.

The experiment was arranged in a split-split-plot arrangement with randomized complete block design and three replications. Two different irrigation regimes including irrigation every 7th day (a1) and rain-fall sowing without irrigation (a2) were compared in the main plots. Four levels of SA were assigned in sub-plots at 0 (distilled water), 0.01, 0.1 and 1 mM SA (as b1, b2, b3 and b4,

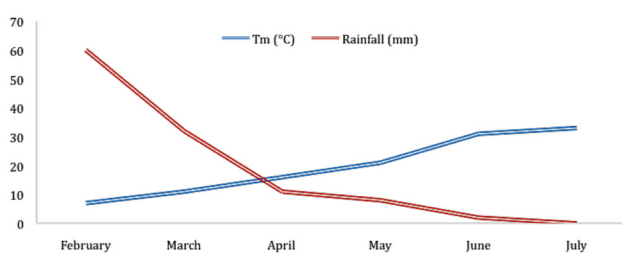


Fig. 1. Pattern of monthly rainfall and temperature recorded during the vegetation season of 2013.

respectively). Four chickpea cultivars were assigned in sub-sub-plots. Each plot contained three sowing rows 3 m in length from each cultivar. Inter- and intra-row spacing was 30 and 10 cm respectively. Sowing of chickpea seeds was performed by hand on 1 March 2013. Foliar spraying of the plants with distilled water (control) and salicylic acid treatments were performed four times at 20, 30, 40 and 50 days after sowing in the amount of 2 L per plot.

Morphological measurements and canopy temperature

At harvest maturity seed yield, number of seeds per plant, plant height, plant biomass and 100-seed weight was determined based on five randomly selected plants from each genotype in sub-plots. Canopy temperature measurements were made during the flowering period. A hand-held infrared thermometer (Teletemp model AG-42, Fullerton, CA) was used to monitor the canopy temperature. The instrument was held so as to view the crop at an angle of 30° from the horizontal at right angles to the rows at a distance of 45 cm from the sample row. This procedure was followed to minimize the influence of exposed soil. Each canopy temperature was the average of three readings recorded from different points in each plant.

Chlorophyll and carotenoid concentration

For measuring the leaf biochemical traits five leaves from different parts of canopy from each plant (five plants from each plot) on day 90th of chickpea grown under two different environments (after flowering and during the pod filling) were harvested. Samples for chlorophyll and carotenoid determination were taken from chickpea leaves using a 0.1 cm diameter cork borer, weighed quickly in pre-weighed clean glass vials and 5 cm³ of 80% acetone was added to these samples. The leaf material was bleached and decanted off. The optical density was read at $\lambda = 663, 646$ and 470 nm using 80% acetone as a blank by a spectrophotometer (Spectronic Genesys-5, Milton Roy). Concentration of chlorophyll *a*, chlorophyll *b* and carotenoids ($\mu\text{g g}^{-1}$) was calculated according to Lichtenthaler and Wellburn (1983).

Leaf proline and soluble sugar concentration

The proline concentration in fresh leaves was determined by adopting the method of Bates et al. (1973). A sample was extracted in sulphosalicylic acid. To the extract, equal volumes of glacial acetic acid and ninhydrin solutions were added. The sample was heated at 100 °C, to which 5 mL of toluene was then added. The absorbance of toluene layer was read at 528 nm, on a spectrophotometer.

Concentrations of total soluble sugars were extracted and analyzed according to Ci et al. (2009). The leaf samples (0.5 g) were homogenized in 2 ml of 80% (v/v) alcohol, and the mortar was washed three times with 3 mL of 80% alcohol. The homogenates were placed at room temperature for 30 min and then centrifuged at 4 000 g for 20 min. The supernatant was stored at 4 °C. The supernatant (0.5 mL)

was mixed with 3 mL of anthrone and the mixtures were incubated at 95 °C for 10 min. The absorbance at 620 nm was then recorded.

The concentration of proline and soluble sugar was expressed in units of $\mu\text{mol g}^{-1}$ fresh weight.

Data analysis

Data were analyzed using one-way analysis of variance (ANOVA) and the means were separated using Duncan's multiple-range tests at the 5% level of significance.

Results

The field experiment was conducted for evaluation of possible effects of exogenous SA application on growth

and physiological parameters of chickpea under water shortage. Four Iranian chickpea cultivars ('Kaka', 'Pirouz', 'Jam' and 'ILC482') were treated with four different SA concentrations (0, 0.01, 0.1 and 1 mM) under well-watered and rainfed-only conditions. Various growth indices and physiological parameters were studied.

Drought stress had significant effect on seed yield and morphological traits. Seed yield and its components decreased from normal irrigation regime to rainfall-only environment (Table 1). Different doses of SA showed different effects of morphological traits, except for plant height where no significant differences were observed by SA application (Table 1). For seed yield and its components, maximum response was generated by foliar application of 0.1 mM SA (Table 1).

Table 1. Seed yield and morphological parameters of four chickpea cultivars grown under four salicylic foliar application and two irrigation regimes. Data are means from three replications. Means followed by same letters in a group of a column are not significantly different at $P \leq 0.05$ according to LSD test

Cultivar	Irrigation regimes	Salicylic acid (mM)	Weight of 100 seeds (g)	Plant biomass (g per plant)	Number of seeds per plant	Seed yield (g per plant)	Plant height (cm)
'Kaka'	Normal	0	15.01 cd	54.91 a	179 a	26.96 a	35.10 a
		0.01	14.98 cd	55.18 a	180.2 a	26.98 a	34.70 a
		0.1	15.12 cd	59.79 a	183.4 a	27.79 a	35.18 a
		1	15.07 cd	57.09 a	179.5 a	27.73 a	35.20 a
	Rainfed	0	11.02 d	33.12 c	128.6 b	15.63 c	29.72 b
		0.01	12.79 d	34.79 c	130.6 b	16.51 c	29.49 b
		0.1	13.01 d	36.86 c	134.6 a	18.41 c	30.00 b
		1	13.10 d	37.01 c	141.7 a	17.79 c	29.6 b
'Pirouz'	Normal	0	17.63 c	35.16 c	77.18 c d	13.89 cd	36.11 a
		0.01	16.94 c	34.72 c	78.08 c d	13.70 cd	35.67 a
		0.1	18.10 c	36.26 c	80.21 c	14.90 c	36.41 a
		1	18.49 c	35.79 c	79 c	14.70 c	36.20 a
	Rainfed	0	12.76 d	23.92 d	68.17 d	8.78 d	30.98 b
		0.01	11.93 d	24.41 d	68 d	8.68 d	31.18 ab
		0.1	13.72 d	26.98 d	70.19 c d	10.28 d	31.41 ab
		1	14.70 d	25.79 d	70.30 c d	9.97 d	31.12 ab
'Jam'	Normal	0	26.5 a	42.89 b	75.16 c d	19.98 b	33.84 a
		0.01	28.2 a	42.86 b	74.99 c d	19.69 b	33.69 a
		0.1	27.4 a	47.97 ab	75.91 c d	21.52 b	33.91 a
		1	27.54 a	46.54 b	75.62 c d	20.98 b	33.28 a
	Rainfed	0	22.81 b	29.21 d	54.81 e	12.01 cd	30.48 b
		0.01	23.16 b	28.89 d	54.97 e	12.21 cd	30.28 b
		0.1	22.1 b	32.26 cd	56.26 e	13.72 cd	31.44 ab
		1	24.09 b	31.87 cd	55.79 e	13.42 cd	30.97 ab
'ILC482'	Normal	0	24.96 b	41.18 b	78.98 c d	20.69 b	34.97 a
		0.01	27.12 a	42.41 b	79.49 c d	20.94 b	35.11 a
		0.1	25.88 ab	43.49 b	81.18 c	21.18 b	35.19 a
		1	27.75 a	42.98 b	80.37 c	20.78 b	35.18 a
	Rainfed	0	19.77 bc	24.11 d	50.12 e	10.78 d	27.89 b
		0.01	20.81 bc	25.40 d	50.79 e	10.92 d	28.19 b
		0.1	21.82 bc	25.89 d	51.97 e	11.71 d	28.21 b
		1	21.77 bc	25.63 d	51.71 e	11.09 d	28.16 b

Seed yield and most of the measured traits significantly decreased in the rainfall environment as compared to normal irrigation conditions. 'Kaka' showed higher seed yield, number of seeds per plant and plant biomass in both rainfall and irrigated environments, compared to other genotypes, while 100-seed weight of 'Jam' and 'ILC482' was higher than that of 'Kaka' and 'Pirouz' (Table 1). Maximum response was generated by foliar application of 0.1 mM SA, where in the rainfed condition seed yield per plant increased by 64%, seed weight by 40%, number of seeds per plant by 14% and plant biomass by 22.3% over those of the control (Table 1). Plant height under the water stress condition was slightly higher in 1 mM SA-treated plants than in control plants, while in well-watered plants, control plants was slightly higher than SA-treated plants (Table 1).

Water stress significantly increased proline and sugar concentrations in all cultivars (Table 2). Different genotypes and SA application showed different effects on leaf proline and soluble sugar concentration (Table 2). There were no differences for most physiological traits for each cultivar by different SA treatments in well-watered conditions, while SA application resulted in significantly increased leaf proline and soluble sugar concentration in rainfed conditions, than in normal irrigation conditions (Table 2). Among the four concentrations of SA in water stress conditions, 0.01 and 1 mM proved to be the best and they significantly increased proline concentration (42% as compared with the control plants; Table 2). However, in irrigated conditions, 0.1 and 1 mM SA treatment showed the highest effect and significantly increased proline

Table 2. Leaf physiological parameters of four chickpea cultivars grown under four salicylic foliar application and two irrigation regimes. Data are means from three replications. Means followed by same letters in a group of a column are not significantly different at $P \leq 0.05$ according to LSD test

Cultivar	Irrigation regimes	Salicylic acid (mM)	Canopy temperature (°C)	Soluble sugars ($\mu\text{mol g}^{-1}$)	Carotenoids ($\mu\text{g g}^{-1}$)	Proline ($\mu\text{mol g}^{-1}$)	Chllorophyll a ($\mu\text{g g}^{-1}$)	Chllorophyll b ($\mu\text{g g}^{-1}$)	
'Kaka'	Normal	0	23.85 b	0.26 c	0.037 a	0.27 b	0.047 a	0.056 a	
		0.01	24.85 b	0.28 c	0.036 a	0.30 b	0.048 a	0.060 a	
		0.1	24.86 b	0.28 c	0.039 a	0.32 b	0.053 a	0.059 a	
		1	24.99 b	0.29 c	0.038 a	0.27 b	0.052 a	0.062 a	
	Rainfed	0	26.61 a	0.38 b	0.026 c	0.35 b	0.025 b	0.021 d	
		0.01	28.12 a	0.47 a	0.028 c	0.40 b	0.027 b	0.025 cd	
		0.1	28.66 a	0.46 a	0.032 b	0.40 b	0.030 b	0.026 cd	
		1	29.22 a	0.46 a	0.030 bc	0.37 b	0.030 b	0.024 cd	
	'Pirouz'	Normal	0	22.49 b	0.27 c	0.038 ab	0.20 c	0.044 a	0.048 ab
			0.01	25.79 ab	0.30 c	0.040 a	0.22 c	0.046 a	0.052 ab
			0.1	26.28 ab	0.29 c	0.044 a	0.19 c	0.049 a	0.053 ab
			1	26.98 ab	0.30 c	0.042a	0.23 c	0.049 a	0.051 ab
Rainfed		0	25.79 ab	0.40 b	0.030 bc	0.36 b	0.024 b	0.030 c	
		0.01	27.41 a	0.48 a	0.032 b	0.39 b	0.026 b	0.033 c	
		0.1	27.89 a	0.46 a	0.030 bc	0.40 ab	0.029 b	0.033 c	
		1	28.49 a	0.46 a	0.032 b	0.37 b	0.029 b	0.032 c	
'Jam'		Normal	0	23.22 b	0.28	0.035 b	0.43 ab	0.050 a	0.040 b
			0.01	24.34 b	0.36 b	0.038 a	0.47 a	0.053 a	0.047 b
			0.1	25.69 ab	0.32 c	0.039 a	0.46 a	0.054 a	0.048 b
			1	25.79 ab	0.32 c	0.036 b	0.44 a	0.055 a	0.045 b
	Rainfed	0	26.88 a	0.42 b	0.033 b	0.43 ab	0.024 b	0.017 d	
		0.01	27.47 a	0.52 a	0.032 b	0.47 a	0.027 b	0.020 d	
		0.1	27.91 a	0.50 a	0.036 b	0.46 a	0.029 b	0.020 d	
		1	28.56 a	0.48 a	0.035 b	0.44 a	0.028 b	0.019 d	
	'ILC482'	Normal	0	24.17 b	0.24 c	0.040 a	0.31 b	0.042 a	0.054 a
			0.01	24.89 b	0.28 c	0.042a	0.36 b	0.046 a	0.059 a
			0.1	25.90 ab	0.26 c	0.044a	0.35 b	0.045 a	0.061 a
			1	25.91 ab	0.26 c	0.042a	0.34 b	0.047 a	0.058 a
Rainfed		0	27.69 a	0.40 b	0.032 b	0.43 a	0.021 b	0.019 d	
		0.01	28.81 a	0.49 a	0.033 b	0.49 a	0.025 b	0.023 d	
		0.1	29.60 a	0.46 a	0.036 b	0.51 a	0.026 b	0.024 d	
		1	30.41 a	0.45 a	0.039 ab	0.49 a	0.024 b	0.022 d	

concentration in comparison to control plants (Table 2). Similarly, soluble sugar concentration in water stress conditions increased by exogenous application of SA at 0.1 and 1 mM, while in well-watered conditions, treatment with different concentrations of SA had no effect on soluble sugar concentration (Table 2).

In general, water stress significantly reduced leaf chlorophyll *a*, chlorophyll *b* and carotenoid concentration as compared to the levels in well-watered conditions (Table 2). SA application showed different effects on leaf photosynthetic pigments depending on genotype. Interactive effects of genotype and irrigation regime for leaf photosynthetic pigments significantly decreased when plants were subjected to water stress (Table 2). It seems that 'Kaka' was able to keep or accumulate more chlorophyll under stress conditions when compared to other genotypes, while it showed higher seed yield and other yield components than in the other genotypes (Table 2). In both well-watered and rainfed conditions, carotenoid and chlorophyll *a* and *b* concentrations were increased significantly by exogenous application of SA. Maximum responses in all cultivars were generated by the foliar application of 0.1 and 1 mM of SA (Table 2). In well-watered conditions higher concentration of carotenoids occurred in 0.1 mM SA treated plants (Table 2).

Water stress significantly increased canopy temperature (Table 2). No significant differences were observed between genotypes in both environments for canopy temperature. Treatment of plants with SA significantly increased canopy temperature in both well-watered and rainfed conditions, so that the maximum response was generated by concentration of 1 mM of SA and was 50% higher than that of the control plants (Table 2).

Discussion

From the results of this study it can be concluded that the drought stress significantly affects all of the studied morphological and physiological traits in the chickpea cultivars. Drought stress significantly decreased seed yield and other morphological traits, while increased soluble sugar and proline. The results showed the role of SA in regulating drought stress response of chickpea, and suggested that SA could be used as a potential growth regulator to improve plant growth under drought stress conditions. SA at a concentration of 0.01 and 0.1 mM decreased the seed yield and its components, while most efficient doses of SA for improving physiological attributes were 0.1 and 1 mM.

Water deficit stress is deleterious for plant growth and yield (Garg et al. 2004; Samarah et al. 2004) and is one of the largest limiting factors in agriculture (Reddy et al. 2004). In chickpea, complex response to drought stress as a result of variation in physiological parameters has been reported (Gunes et al. 2006; Gunes et al. 2008; Hayat et al.

2012). Several reports were published in the last decade demonstrating that SA may play a pivotal role in signaling for plant resistance to environmental stresses (Hayat et al. 2008; Hayat et al. 2012; He et al. 2014).

Drought stress promoted or inhibited a variety of physiological and biochemical changes that can be used as markers against stress. The present investigation suggests positive effects of SA on morphological attributes such as seed weight, plant biomass and seed yield. Increased number of seeds per plant and seed weight seemed to be a direct result of improved seed yield (Table 1). Increases in biomass and consequently the seed yield of drought stressed plants in response to SA may be related to the induction of protective role of membranes that increase the tolerance of plants to damage. Coronado et al. (1998) reported that aqueous solutions of SA as a spray to shoots of soybean significantly increased the growth of shoots and roots in either greenhouse or field conditions. Proline and soluble sugars are important components of the defense system of plants to counteract stress (Hayat et al. 2008). As one of the most important cellular compatible solutes, proline plays an important role in plant adaptation to drought or dehydration (Szabados, Savoure 2010; He et al. 2014). Increasing evidence has revealed that exogenous SA promotes proline accumulation in plants exposed to abiotic or biotic stresses (Mishra, Saxena 2009).

In this study, exogenous application of 0.01 and 0.1 SA in rainfed-only environment resulted in increased endogenous proline concentration (Table 2), whereas higher concentration of SA might have reversed the phenomenon. Similar observation was made in different plant species subjected to exogenous application of SA under drought stress conditions (Hayat et al. 2008; Hayat et al. 2012; Afshari et al. 2013). In this study, when the plants were subjected to drought stress, chlorophyll *a*, chlorophyll *b* and carotenoid concentration was significantly reduced (Table 2). Defense in photosynthetic pigments by water stress seems to be the consequence of the closure of stomata, thereby decreasing CO₂ supply as well as internal CO₂ concentration (Tiwari et al. 2005; Hayat et al. 2008). However, the total carotenoid, chlorophyll *a* and chlorophyll *b* concentration in SA-treated plants was also higher than in control plants during the drought period (Table 2). Increased photosynthetic pigment concentration in plants subjected to SA under drought stress may be the result of enhanced activity of Rubisco and PEP carboxylase under stress (Popova et al. 2003; Singh, Usha 2003).

Our results are in agreement with observations for tomato and cowpea plants treated with SA under drought stress (Hayat et al. 2008; Afshari et al. 2013). The present results indicate that drought stress retards the growth and metabolic activity of different genotypes of chickpea. SA resulted in higher concentrations of proline, soluble sugars and photosynthetic pigments, both under well-watered and drought stress conditions. The influence of

SA on photosynthetic and osmolyte accumulation was more pronounced under stress, suggesting that the elevated level of these parameters, at least in part, increased the tolerance of chickpea plants to water stress, thus protected the photosynthetic machinery and perhaps plant growth. The present study also confirmed the observations made by Idrees et al. (2010), Hayat et al. (2012) and Bidabadi et al. (2012) that the most effective role of SA in alleviating the hazards of stresses can be observed at lower (up to 1 mM) doses. It is clear that adverse effects of drought stress on chickpea growth and leaf biochemical parameters were significantly improved by exogenous application of SA. The mechanisms by which drought inhibits growth are complex and controversial. Although the causes of drought stress have been characterized, our understanding of the mechanisms by which drought prevents plant growth and alternating way to help the plants keep their potential yield under drought stress is still rather poor. Our results indicated that SA treatment applied as a foliar spray promoted photosynthetic activity, osmolyte accumulation as well as plant growth under drought stress.

References

- Afshari M., Shekari F., Azimkhani R., Habibi H., Fotokian M.H. 2013. Effects of foliar application of salicylic acid on growth and physiological attributes of cowpea under water stress conditions. *Iranian Agric. Res.* 32: 55–70.
- Bezrukova M.V., Sakhabutdinova R., Fathutdinova R.A., Kyldiarova I., Shakirova F. 2001. The role of hormonal changes in protective action of salicylic acid on growth of wheat seedlings under water deficit. *Agrochimiya* 2: 51–54.
- Bidabadi S.S., Mahmood M., Baninasab B., Ghobadi C. 2012. Influence of salicylic acid on morphological and physiological responses of banana (*Musa acuminata* cv. 'Berangan', AAA) shoot tips to *in vitro* water stress induced by polyethylene glycol. *Plant. Omics J.* 5: 33–39.
- Bideshki A., Arvin M.J. 2010. Effect of salicylic acid (SA) and drought stress on growth, bulb yield and allicin content of garlic (*Allium sativum*) in field. *Plant Ecophysiol.* 2: 73–79.
- Borsani O., Valpuesta V., Botella M.A. 2001. Evidence for a role of salicylic acid in the oxidative damage generated by NaCl and osmotic stress in Arabidopsis seedlings. *Plant Physiol.* 126: 1024–1034.
- Coronado M.A.G., Lopez C.T., Saavedra A.L. 1998. Effects of salicylic acid on the growth of roots and shoots in soybean. *Plant Physiol. Biochem.* 8: 563–565.
- Dat J.F., Foyer C.H., Scott I.M. 1998. Changes in salicylic acid and antioxidants during induced thermotolerance in mustard seedlings. *Plant Physiol.* 118: 1455–1461.
- Fariduddin Q., Hayat S., Ahmad A. 2003. Salicylic acid influence net photosynthesis rate, carboxylation efficiency, nitrate reductase activity and seed yield in *Brassica juncea*. *Photosynthetica* 41: 281–284.
- Garg B.K., Burman U., Kathju S. 2004. The influence of phosphorus nutrition on the physiological response of moth bean genotypes to drought. *J. Plant Nutr. Soil. Sci.* 167: 503–508.
- Gunes A., Cicek N.C., Inal A., Alpaslan M., Eraslan F., Guneri E., Guzelordu T. 2006. Genotypic response of chickpea (*Cicer arietinum* L.) cultivars to drought stress implemented at pre- and post-anthesis stages and its relations with nutrient uptake and efficiency. *Plant Soil. Environ.* 52: 368–376.
- Gunes A., Inal A., Adak M.S., Bagci E.G., Cicek N., Eraslan F. 2008. Effect of drought stress implemented at pre- or post-anthesis stage some physiological as screening criteria in chickpea cultivars. *Rus. J. Plant. Physiol.* 55: 59–67.
- Hayat Q., Hayat S., Alyemeni M.N., Ahmad A. 2012. Salicylic acid mediated changes in growth, photosynthesis, nitrogen metabolism and antioxidant defense system in *Cicer arietinum* L. *Plant Soil. Environ.* 58: 417–423.
- Hayat Q., Hayat S., Irfan M., Ahmad A. 2010. Effect of exogenous salicylic acid under changing environment: a review. *Environ. Exp. Bot.* 68: 14–25.
- Hayat S., Hasan S.A., Fariduddin Q., Ahmad A. 2008. Growth of tomato (*Lycopersicon esculentum*) in response to salicylic acid under water stress. *J. Plant. Interact.* 3: 297–304.
- He Q., Zhao S., Ma Q., Zhang Y., Huang L., Li G., Hao L. 2014. Endogenous salicylic acid levels and signaling positively regulate Arabidopsis response to polyethylene glycol-simulated drought stress. *J. Plant Growth Regul.* 33: 871–880.
- Horváth E., Pál M., Szalai G., Páldi E., Janda T. 2007. Exogenous 4-hydroxybenzoic acid and salicylic acid modulate the effect of short-term drought and freezing stress on wheat plants. *Biol. Plant.* 51: 480–487.
- Hussain M., Malik M.A., Farooq M., Ashraf M.Y., Cheema M.A. 2008. Improving drought tolerance by exogenous application of glycinebetaine and salicylic acid in sunflower. *J. Agron. Crop. Sci.* 194: 193–199.
- Idrees M., Khan M.M.A., Aftab T., Naeem M., Hashmi M. 2010. Salicylic acid-induced physiological and biochemical changes in lemongrass varieties under water stress. *J. Plant. Interact.* 5: 293–303.
- Khan W., Prithiviraj B., Smith D. 2003. Photosynthetic responses of corn and soybean to foliar application of salicylates. *J. Plant. Physiol.* 160: 485–492.
- Lichtenthaler H.K., Wellburn A.R. 1983. Determinations of total carotenoids and chlorophylls *a* and *b* of leaf extracts in different solvents. *Bioch. Soc. Trans.* 11: 591–592.
- Mafakheri A., Siosemardeh A., Bahramejad B., Struik P.C., Sohrabi Y. 2011. Effect of drought stress and subsequent recovery on protein, carbohydrate contents, catalase and peroxidase activities in three chickpea (*Cicer arietinum*) cultivars. *Austr. J. Crop Sci.* 5: 1255–1260.
- Metwally A., Finkemeier I., Georgi M., Dietz K.J. 2003. Salicylic acid alleviates the cadmium toxicity in barley seedlings. *Plant. Physiol.* 132: 272–281.
- Mishra N., Saxena P. 2009. Effect of salicylic acid on proline metabolism in lentil grown under salinity stress. *Plant. Sci.* 177: 181–189.
- Pal M., Kovacs V., Szalai G., Soos V., Ma X., Liu H., Mei H., Janda T. 2014. Salicylic acid and abiotic stress responses in rice. *J. Agron. Crop. Sci.* 200: 1–11.
- Popova L., Ananieva E., Hristova V., Christov K., Georgieva K., Alexieva V., Stoinova Z.H. 2003. Salicylic acid and methyl jasmonate-induced protection on photosynthesis to paraquat oxidative stress. *Bulg. J. Plant. Physiol.* SI: 133–152.
- Reddy A.R., Chaitanya K.V., Vivekanandan M. 2004. Drought-induced responses of photosynthesis and antioxidant metabolism in higher plants. *J. Plant. Physiol.* 161: 1189–1202.
- Samarah N., Mullen R., Cianzio S. 2004. Size distribution and mineral nutrients of soybean seeds in response to drought

- stress. *J. Plant Nutr.* 27: 815–835.
- Singh B., Usha K. 2003. Salicylic acid induced physiological and biochemical changes in wheat seedlings under water stress. *Plant. Growth Regul.* 39: 137–141.
- Szabados L., Savoure A. 2010. Proline: a multifunctional amino acid. *Trends Plant. Sci.* 15:89–97
- Talebi R., Ensafi M.H., Baghebani N., Karami E., Mohammadi K.H. 2013. Physiological responses of chickpea (*Cicer arietinum*) genotypes to drought stress. *Environ. Exp. Biol.* 11: 9–15.
- Tiwari A., Kumar P., Singh S., Ansari S.A. 2005. Carbonic anhydrase in relation to higher plants. *Photosynthetica* 43: 1–11.