

EFFECTS OF SALICYLIC ACID ON SOME GROWTH AND PHYSIOLOGICAL CHARACTERISTICS OF LETTUCE (*LACTUCA SATIVA* L.) UNDER CADMIUM STRESS CONDITIONS

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(nazhad.talabany@gmail.com; ikbal.tahir@koyauniversity.org)**Received:** 07 Aug., 2022 / **Accepted:** 28 Oct., 2022 / **Published:** 01 Jan., 2023 <https://doi.org/10.25271/sjuoz.2022.10.4.987>**ABSTRACT:**

Lettuce (*Lactuca sativa* L.) is considered as the most cultivated and consumed leafy vegetable all over the world. In Erbil Governorate, most farmers used sewage water to irrigate lettuce, which caused health risks especially heavy metal pollutants including cadmium. This research was applied as a factorial experiment to investigate the effects of foliar spraying of salicylic acid (SA) (0, 0.5, 1, 1.5 or 2mM) followed by spraying cadmium chloride (Cd) (0, 1, 2, or 4mM) on the growth, and some physiological and biochemical characteristics of this plant. The results show that most vegetative growth characteristics responded inversely to increasing SA concentrations, whereas the response of root performance improved with using SA application. A low concentration of Cd (1mM) improved root and shoot performance, whereas increasing the concentration to 4 mM decreased these characteristics significantly compared to no Cd added plants, except for the percent of shoot dry matter. Most of photosynthetic pigments decreased significantly by foliar spraying with SA and Cd. Applications of SA increased catalase enzyme activity significantly compared to no SA treated plants. Cadmium foliar application increased peroxidase enzyme activity, ascorbic acid, proline, and the percent of total carbohydrate content and decreased catalase enzyme activity and the percent of infection with watery soft rot significantly compared to no Cd treated plants. It is concluded that lettuce tolerance to cadmium stress was increased by pretreatment foliar application of SA.

KEYWORDS: Cadmium stress, Catalase enzyme, Lettuce, Proline, Salicylic acid.**1. INTRODUCTION**

Lettuce (*Lactuca sativa* L.) is considered as the most consumed and cultivated leafy vegetable all over the world, with yearly continuing production increase. *Lactuca sativa* belongs to the Asteraceae family (Kadereit, 2007). In Iraq, the area culture reached 13266 donum, and the production rate reached 26507 tons yearly (CSO-Iraq, 2018). Lettuce is cultivated especially in the middle region of Iraq, where the crop in markets depends initially on the production from this region, where the seed plant starts in the late summer to give a crop during winter, while in the northern region it begins during late autumn to give a crop during the spring season (Matlub, et al., 1989).

Most Iraqi farmers used sewage water to irrigate leafy vegetables including lettuce, which is polluted and considered as causing health risks to the community. In Erbil Governorate, around 225 hectares areas are irrigated by non-pretreated sewage water for the production of many uncooked vegetables which are consumed by near two millions of people as an important part of their diet (Matraszek, et al., 2016). There are many potential sources of pollution in Erbil Governorate, as the most populous city in the Kurdistan Region. This condition has produced significant soil pollution, especially with copper (Cu), zinc (Zn), cadmium (Cd), and lead (Pb), and the assembly of polycyclic aromatic hydrocarbons on roadsides, industrial and commercial areas (Amjadian, et al., 2016). The most globally known sources of released Cd to the environment are cement plants, phosphate fertilizers, urban traffic, waste incinerators, heating systems, metalworking industries, and power plants. David and Eric (2000) found that about 660 metric tons of cadmium globally are used annually in soils through the application of phosphate fertilizers (Jibril, et al.,

2017). Like other nonessential elements for plants, cadmium is not recognized to have a benefit to plants (Verma, 2008). It is highly toxic to crops (Meng, et al., 2009; Rizwan, et al., 2016). Plants take Cd easily and accumulate it in many parts of plants (Liu, et al., 2016). Cadmium can go in and harm crops by encouraging oxidative stress such as overproduction of reactive oxygen species decreasing the activities of antioxidant enzymes, damaged cell membranes, and reduced biomass, thus reducing the quality of wrinkled crops (Gao, et al., 2020). To decrease the adverse effects of cadmium stress on plants, many strategies were applied including; several direct methods (mycosis fungoid, genetically modified plants, grafting) that are used to overcome heavy metal contamination problems (Edelstein and Ben-Hur, 2018). In latest years, several researches have reported that foliar application of salicylic acid results in a significant effect on plant adaptation to stress factors including heavy metals, as SA actions as a natural signaling particle targeting different plant species to increase their tolerance to abiotic pressures and living (Hayat, et al., 2010a; Hayat, et al., 2010b; Khan, et al., 2015) including protection against toxicity of Cd (Asgher, et al., 2014a). The aims of this study are to investigate salicylic acid and cadmium effects on some growth characteristics, development, and biochemical and physiological properties of lettuce plants, and to know about the ability of salicylic acid to reduce the inhibitory effects of cadmium on this plant.

2. MATERIALS AND METHODS**2.1. Plant Materials, Cultivation, and Treatments**

A factorial experiment was conducted in a vegetable field in the Taq-Taq district (35.8857°N, 44.5932°E), Erbil, Kurdistan

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Region- Iraq, during winter 2021-2022. Lettuce seed (*Lactuca sativa* L.) variety Romaine, produced by NADER company, Netherlands, were planted on October 9th by 2.5 g seeds/ m² in a silty loam soil with 6.3pH and the open field. Transplanting started when seedlings were 10-12 cm in length. Each experimental unit consists of 36 plants/ 2m² as 6 lines, spacing 30-35cm between lines, and surface flood irrigation was used (Figure 1). The average environmental conditions throughout the growing season varied from (8-24.8 C^o) for temperature, 26 – 71.2% for relative humidity, and 0-139.3mm for precipitation. Field agricultural services were done, including weed control (done three times), fertilization, manuring, and using the fungicide CURENOX 50WP.



Figure 1. Transplanting growth and field design (A) Transplanting growth in the seedling bed, (B) Field plotting, (C) one of the experimental units, and (D), an overall photo of the field.

The experiment consisted of two factors, the first was five concentrations of salicylic acid (2-Hydroxybenzoic acid) (0, 0.5, 1, 1.5, and 2mM) mentioned as SA0, SA0.5, SA1, and SA2, and the second factor was four concentrations of cadmium chloride CdCl₂.H₂O (0, 1, 2, and 4mM) which were mentioned as Cd0, Cd1, Cd2, and Cd4 respectively. Ordinary water was used as the control treatment. 0.1% detergent was added to the treatment solutions as a surfactant chemical (Saini, et al., 2014). The solutions were used as foliar spraying above and under the leaves until complete wetting according to the design. The first spray was applied on the 20th of December; and repeated four times 10 days intervals (Guo, et al., 2019).

2.2. Studied Characteristics

Five plants for each experimental unit were used to determine all studied characteristics as follows: for many leaves which comprise all leaves except those less than 5cm (El-Habar and Al-Saabei, 2008), plant leaf area (cm²) by taking leaves pictures and converting them to a binary system with Image J application program (<http://imagej.nih.gov/ij/>, java 1.8.0, 172 (64 bit)) application), leaf thickness (mm) by using caliper micrometer (J0006, size 0-25mm, China), length of plant shoot and root (cm) by standard metric tapeline, wet weight and the percent of shoot and root dry weight as it is mentioned by Tudela, et al., (2017). Chlorophyll A (Chl.A), chlorophyll B (Chl.B), and total carotenoids (total Cars.) were estimated as it is mentioned by Lichtenthaler, and Wellburn, (1983) who found the ratio of 50 ml 80% acetone: 1 g sample. For some enzymatic and non-enzymatic antioxidant content; peroxidase enzyme activity (POD) in leaves (absorbing units. gram⁻¹ fresh weight) is calculated according to the procedure of (Müftügil, 1985) using guaicaol and H₂O₂ spectrophotometrically at 420 nm, catalase enzyme activity (CAT) in leaves (absorbing units. gram⁻¹ fresh weight) using spectrophotometer according to (Aebi, 1974) which depends on the change in absorbance at 240 nm for 30mM H₂O₂ and 50mM phosphate buffer at PH=7

solutions, ascorbic acid (AA) in leaves (mg AA.100g⁻¹ dry weight) by the method of Elbsheer, (2018) spectrophotometrically which determine the absorption at 665 nm using methylene blue solution, proline content (mM.g⁻¹ fresh weight) determined using sulphosalicylic acid and ninhydrin using the spectrophotometer at 520nm which is calibrated with the standard curve of proline according to the work of Bates, et al., (1973), percent of total carbohydrate determined using concentrated sulfuric acid (H₂SO₄) and phenol spectrophotometrically at 488 nm as it is mentioned by Herbert, et al., (1971).

During the growth of the plants in the field, infections with watery soft rot pathogen (*Sclerotinia sclerotiorum*) were observed; and diagnosed by Dr. Qasim Abdulla Omer Marzani and Dr. Kamaladin Muhamad Fatah, Department of Plant Protection, College of Agricultural Engineering Sciences, University of Salahaddin.

2.3. Statistical Analysis and Experimental Design

A Factorial experiment with randomized complete block design with three replications was used to conduct this study. Data were submitted for analysis of variance; Duncan's multiple range test at a probability level of %5 was used for comparing between experiment means using the SAS program (Al-Mohammadi and Al-Mohammadi, 2002).

3. RESULTS AND DISCUSSIONS

Results in Table 1 show that the characteristics of vegetative growth, exemplified by the leaf area, shoot length, and the percent of shoot dry matter responded inversely to the increase of SA concentration, except for the leaf thickness and shoot fresh weight which increased with increasing SA concentration, similarly to the response of root performance which improved with using SA application. Regarding the effects of cadmium on the performance of shoot and roots, it is clear that a low concentration of Cd increased these characteristics compared to the plants that were not sprayed with SA (SA0) except shoot length and fresh weight which decreased with increasing the Cd concentration. Increasing the concentration to 4mM decreased these characteristics values significantly compared to SA0 treatment, except the percent of shoot dry matter which increased significantly when 4mM of Cd were used. The significant decrease in the plant leaf area caused by high SA concentration (Table 1) is similar to that of Belkadihi, et al., (2012) who found a significant decrease in flax leaf area with increasing SA concentration, and opposite to that of Youssef, et al., (2017a) and Youssef, et al., (2017b) who obtained positive effects of foliar application of SA up to 0.5mM on the vegetative growth parameters of lettuce and strawberry plants (for the leaves area). Whereas their results regarding strawberry support the results we obtained about root dry weight which increased significantly with increasing the SA concentration (Table 1). This increment may be due to increasing the elements content in roots, the reason that gained by (Jamali, et al., 2015) who found that SA at 0.5 or 1mM increased N, P, K, Ca, Mg, Fe, and Zn in shoot and root of strawberry plants. Increasing leaf thickness significantly with increasing SA coincides with that of Youssef, et al., (2017a) who found that spraying SA at 4mM increased the thickness of the cells of both upper and lower epidermis compared with those of the water-sprayed plants. Whereas Cárcamo, et al., (2012) mentioned non-significant differences in leaf thickness between plants that pretreated with SA before Cd and the control plants under saline stress. The significant decrease in lettuce number of leaves, leaf area and thickness, shoot length, and the length of root with Cd application (Table 1) was parallel with the results of Zhao, et al., (2021) who found a decrease in net growth in plants with increasing concentration of cadmium, which may be due to lower chlorophyll and

chlorophyll b content. The inhibition effect of Cd on lettuce vegetative and root growth are due to the imbalance of some plant growth hormones, especially the auxins (Hasenstein, et al., 1988), or to the disturbance of the balance of minerals that are necessary for the growth of plants (Das, et al., 1997), or to the detrimental effect of cadmium on the cell wall formation (Chaoui and El Feejani, 2005), or to the disturbances of the photosynthetic mechanism, especially the ultrastructure of chloroplasts and chlorophylls biosynthesis (Mobin and Khan, 2007; Ebbs and Uchil, 2008). Although the interaction coefficients between SA and Cd resulted in a significant decrease in stem length, some results were obtained.

Mohammadi, et al. (2020) found under salinity stress a reduction in plant stem length with application of SA. According to Table 1, the low concentration of SA (0.5mM) led to an increase in shoot dry weight significantly. Some results were obtained by Shehata, et al., (2020) in which foliar application from 100-300 PPM caused an increase of shoot. Although pretreatment with SA before Cd application led to a significant increase in dry weight of roots, whereas SA had a nonsignificant effect on root fresh weight. At the same time, SA combination with Cd significantly reduced root wet weight. Some results were obtained by Shehata, et al. (2020) where the wet weight of roots was significantly increased with SA pretreatment under drought stress. Similarly, Hou, et al., (2007) demonstrated that the inhibition of growth may be the result of cadmium interfering with key metabolic procedures such as photosynthesis and transport of photosynthetic products and nutrients.

The results shown in Table 2 illustrate that the foliar spraying of SA led to an increase in Chl. A significantly. While it led to a decrease in Chl. B and total Cars. significantly with increasing SA concentrations. In the same way increasing Cd concentration led to a decrease in leaf content of Chl. A, Chl. B, and total cars. significantly. The increase in chlorophylls with increasing SA had the same results found by Kalaki, et al., (2014) and Sultan, et al., (2016). However, the significant decrease in each of Chl. B and total Cars. with increasing SA concentration was opposite to that of (Shehata, et al., 2020) who found an increase of total cars. as a result of foliar spray with SA (Kalaki, et al., 2014). These significant decreases in Chl. A, B, and total Cars. as a result of Cd application (Table 2) was also reported by Guo, et al., (2016) who illustrated that chlorophylls contents were decreased significantly by increasing the concentrations of Cd in lettuce (Loi, et al., 2018). The reduction of chlorophyll under Cd stress in leaves of lettuce plant. This may be due to the oxidation of chlorophyll and to damage the chloroplast ultrastructure's under cadmium stress (Chen, et al., 2010), the disturbances of chlorophotosynthesis by displacement of the Cd²⁺ ion with Mg²⁺ ion in the chlorophyll molecule, or inhibition of chlorophyll biosynthesis (Hsu and Kao, 2003), or the degradation of chlorophyll B (Liu, et al., 2014).

Table 3 shows that SA had nonsignificant effects on POD enzyme activity except for the lowest concentration (0.5mM) which increased POD enzyme activity significantly compared to other treatments. The same results were demonstrated by Krantev, et al., (2008) who reported that POD enzyme activity was not affected by SA pretreatment. From the same results in Table 3, it was found that foliar application of Cd at all concentrations and most interactions between SA and Cd increased the enzyme activity significantly compared to the control treatment. SA has been identified to function as a signaling particle essential to generate, tolerate, or increase stress caused by cadmium (Zawoznik, et al., 2007). In this study, pretreatment with SA reduced cadmium-induced oxidative stress as indicated by decreased POD activity. The results were increased in consistency with those mentioned by Guo, et al. (2009).

Regarding CAT enzyme activity, it is significantly increased by foliar spraying with SA application, which was not similar to the results indicating that SA actions as a signaling molecule, changing the antioxidant system Liu, et al., (2016), inhibiting CAT and stimulating peroxides. SA can indirectly reduce ROS accumulation by exciting CAT activity.

Exist of SA and Cd caused an increase in POD enzyme activity. The result was parallel with that obtained by Shi, et al., (2009) that showed that pretreatment with SA reduces the toxicity of cadmium in the seedlings of cannabis. It results from reducing the uptake of cadmium, enhancing POD activity, and the activity of POD increased with cadmium levels and was elevated significantly when the plants exposed to Cd element and presoaked with SA, compared with control treatment. In addition, this CAT enzyme activity decreased under cadmium stress. These were observed by Xu, et al., (2013) in which cadmium exposure significantly decreased the activity of CAT enzyme. Cd also increased significantly vitamin C content. This result was opposite to Xu, et al., (2013). Cadmium exposure causes a significant decrease in the content of vitamin C in lettuce seedlings. Although SA pretreatment before Cd increases both POD and CAT enzyme activities, these results are gained in several observations. The application of SA exogenously increases POD and CAT enzyme activity under the stress of Cd in many plants, like in maize (Krantev, et al., 2008), in rice plant (Guo, et al., 2009), and wheat plant (Kang, et al., 2013). SA can reduce cadmium stress by affecting a variety of diverse procedures, including decreasing the uptake of cadmium and their accumulation in plant organs. Scavenging ROS and strengthening the system of antioxidant defense (Agami and Mohamed, 2013; Li, et al., 2014), and improving the photosynthetic capacity (Krantev, et al., 2008; Janda, et al., 2014)

Ascorbic acid (AA) is considered as one of the non-enzymatic antioxidants, helping as a main donor to the redox state of cells and keeping plants from different oxidative damage (Smirnoff, et al., 2000). Table 3 shows that there was a significant increase of ascorbic acid under the stress of Cd, also with pretreatment of SA, especially for the concentrations SA1.5 and Cd 2mM concentrations. These results confirm those of Choudhury and Panda, (2004). Increasing the amount of ascorbate under stress would lead to removing the species of the activated oxygen by activating the ascorbic cycle (Devlin and Withman, 1988). In otherwise foliar application of SA significantly increased the AA under Cd stress, the same result was found by Hasanuzzaman, et al., (2011). SA alleviates the effects of stress by improving the level of enzymes responsible for AA regeneration (Hasanuzzaman, et al., 2014). SA enhanced ascorbate content which serves as an antioxidant and reduces the levels of H₂O₂ under salt stress (Yan, et al., 2018).

Moreover, the amino acid proline and sugars content both doing as osmolytes and maintain cell swelling (Wael, et al., 2015). The results in Table 3 show that the low concentrations of SA (0.5, 1, 1.5mM) significantly increased proline content, as SA pretreatment before Cd exposure significantly increased proline inclusion (Table 3). These results were supported by the improvement after SA providing during cadmium stress in *Phaseolus vulgaris* plants. Providing SA increased the oligosaccharides and proline to give protection of fertilizer and balance of ion (Farhangi-Abriz and Ghassemi-Golezani, 2018). Increasing the total carbohydrate by SA and Cd application compared to the control treatment (Table 3) agrees with the results observed for beet and potato plants (El-Tayeb, 2005), where drought stress leads to increment of starch resolution and the concentration of solution sugars (sucrose and fructose) which help to maintain the cellular turgidity (Yazdanpanah, et al., 2011).

Infection with *Sclerotinia sclerotiorum* had negative effects on lettuce cultivars (Akbudak, et al., 2022). Results in Table 4 shows the percentages of lettuce plant infection with watery soft rot in the field under salicylic acid and cadmium foliar application, where spraying SA had nonsignificant effect on the percent of infection compared to no SA treated plants (Figure

2). Whereas foliar spraying with Cd decreased the infection significantly to 0.11 and 0.111% for 1 and 2mM and 0.08% for 4mM compared to no Cd treated plants which gave 0.123%. The reduction in the percent of watery soft rot by Cd application coincided with, Ma, et al., (2005) who reported that cadmium is generally collected in the walls of cells and soluble fractions, which is confirmed by our results (no published data). However, high humidity and temperature are other factors that may be sources of increasing disease as mentioned in paragraph 2.1. Climatic and environmental conductions, especially precipitation and temperature, can influence the growth and survival of a variety of pathogenic microorganisms,

which can be maintained for a long time in the crop production environment (Guber, et al., 2006; Parker, et al., 2010).



Figure 2. Lettuce plant (A) symptoms of infection by watery soft rot (B) whole infected plant.

Table 1. Effects of foliar spraying with salicylic acid (SA), cadmium (Cd), and their interactions on vegetative growth of *Lactuca sativa* L. Var. Romaine.

Treatments	Plant Leaves Number	Plant Leaf Area (cm ²)	Leaf Thickness (mm)	Shoot			Root		
				Length (cm)	Fresh Weight (g)	Dry Matter (%)	length (cm)	Fresh Weight (g)	Dry Matter (%)
SA concentration (mM)									
SA 0	51.16 B	103.96 A	0.55 B	36.49 A	956.9 CD	6.18 A	16.49 B	47.40 C	16.01 C
SA 0.5	51.74 B	98.53 C	0.61 AB	35.41 B	1065.7 A	6.09 C	16.58 B	52.67 AB	16.06 C
SA 1	56.16 A	102.53 AB	0.65 A	36.24 AB	1032.0AB	6.04 D	17.66 A	51.02 AB	19.99 A
SA 1.5	52.58 B	100.29 BC	0.66 A	36.58 A	1005.0 BC	6.07 C	17.08AB	53.15 A	19.52 A
SA 2	51.08 B	98.67 C	0.64 A	35.91 AB	948.0 D	6.13 B	16.99AB	49.32 CB	18.16 B
Cd concentration (mM)									
Cd 0	54.86 A	105.06 B	0.62 AB	37.73 A	1158.0 A	6.14 B	17.06 B	53.54 B	17.12 B
Cd 1	56.26 A	98.93 C	0.67 A	36.66 B	1096.6 B	6.12 B	18.19 A	57.64 A	21.41 A
Cd 2	53.99 B	109.93 A	0.62 A B	36.13 B	1081.8 B	5.92 C	16.99 B	52.86 B	17.13 B
Cd 4	45.06 C	89.87 D	0.58 B	33.99 C	669.6 C	6.24 A	15.59 C	38.81 C	16.13 C
Interactions between SA and Cd									
SA0 Cd0	54.33 B-D	115.35 A	0.61 B-E	39.33 A	1084.6D-E	6.41 C	16.33C-G	46.88 E-G	14.01 I-K
SA0 Cd1	58.66 A	95.30 FG	0.56 C-E	37.0 B-E	1196.0 A-C	5.89 I	18.0 A-C	57.76 BC	18.36 FG
SA0 Cd2	55.0 A-C	112.91 AB	0.56 C-E	38.0 A-C	1094.3 B-E	5.92 HI	16.33 C-G	50.49 C-G	16.76 H
SA0 Cd4	36.66 G	92.30 F-H	0.48 E	31.66 H	452.6 H	6.50 B	15.33 FG	34.50 I	14.94 I
SA0.5 Cd0	54.33 B-D	106.20 CD	0.59 B-E	36.33C-F	1211.0 AB	5.91 HI	15.66 E-G	49.95 D-G	13.26 K
SA0.5 Cd1	57.33 AB	97.39 E-G	0.77 A	36.66C-F	1145.0B-D	6.24 D	17.66 A-D	58.01 BC	20.70 CD
SA0.5 Cd2	56.66 AB	106.18 CD	0.62 A-E	34.66 FG	1187.6 A-C	5.41 K	17.0 B-F	59.66 AB	16.64 H
SA0.5 Cd4	38.66 G	84.36 I	0.49 DE	34.0 G	719.3 G	6.81 A	16.0 D-G	43.07 GH	13.65 JK
SA1 Cd0	58.66 A	109.45 A-C	0.59 B-E	37.66A-C	1205.6 A-C	6.40 C	19.33 A	54.36 B-E	16.28 H
SA1 Cd1	58.0 AB	94.32 F-H	0.69 A-C	37.33B-D	1137.0B-D	5.97 GH	18.66 AB	59.88 AB	21.47 C
SA1 Cd2	52.0 C-E	114.56 A	0.64 A-D	34.66 FG	1012.6EF	6.06 EF	17.66 A-D	56.23 B-D	22.63 B
SA1 Cd4	56.0 A-C	91.80 GH	0.67 A-C	35.33D-G	772.6G	5.71 J	15.0 G	33.64 I	19.59 DE
SA1.5 Cd0	59.0 A	98.43 EF	0.62 A-E	39.0 AB	1290.0 A	6.09 E	17.33 B-E	65.58 A	21.53 C
SA1.5 Cd1	56.66 AB	104.85 CD	0.72 AB	36.0C-G	1064.0DE	6.11 E	18.66 AB	55.54 B-D	27.21 A
SA1.5 Cd2	49.66 E	105.64 CD	0.67 A-C	36.33C-F	940.6 F	6.09 E	16.33 C-G	47.31 E-G	14.71 IJ
SA1.5 Cd4	45.0 F	92.25 F-H	0.66 A-C	35.0E-G	725.3 G	6.01 FG	16.0 D-G	44.19 F-H	14.63 IJ
SA2 Cd0	48.0 EF	95.89 FG	0.72 AB	36.33C-F	998.6 EF	5.87 I	16.66C-G	50.97 C-F	20.53 CD
SA2 Cd1	50.66 DE	102.79 DE	0.61 B-E	36.33C-F	941.0 F	6.37 C	18.0 A-C	57.03 B-D	19.34 EF
SA2 Cd2	56.66 AB	107.37 B-D	0.61 B-E	37.0 B-E	1174.0 A-D	6.11 E	17.66 A-D	50.65 C-G	14.90 I
SA2 Cd4	49.0 E	88.66 HI	0.62 A-E	34.0 G	678.3 G	6.19 D	15.66 E-G	38.65 HI	17.87 G

* Means followed by the same letters within columns are not significantly differed at $p \leq 0.05$ according to the test of Duncan's Multiple Range.

Table 2. Effects of foliar spraying with salicylic acid (SA), cadmium (Cd), and their interactions on photosynthetic pigments of *Lactuca sativa* L. Var. Romaine.

Treatments	Chlorophyll A	Chlorophyll B	Total Carotenoids
	Mg.g ⁻¹ Fresh Weight		
SA concentration (mM)			
SA 0	0.566 C	0.213 A	0.274 A
SA 0.5	0.542 D	0.197 B	0.247 C
SA 1	0.570 C	0.195 B	0.258 B
SA 1.5	0.580 B	0.197 B	0.262 B
SA 2	0.586 A	0.209 A	0.266 AB
Cd concentration (mM)			
Cd 0	0.684 A	0.262 A	0.308 A
Cd 1	0.558 B	0.199 B	0.255 B
Cd 2	0.555 B	0.184 C	0.263 B
Cd 4	0.478 C	0.164 D	0.219 C
Interactions between SA and Cd			
SA0 Cd0	0.748 A	0.329 A	0.336 A
SA0 Cd1	0.573 G	0.197 F	0.278 D-F
SA0 Cd2	0.585 F	0.203 F	0.302 B-D

SA0 Cd4	0.361 L	0.126 J	0.184 K
SA0.5 Cd0	0.727 B	0.296 B	0.324 AB
SA0.5 Cd1	0.401 K	0.129 J	0.186 K
SA0.5 Cd2	0.634 D	0.224 CD	0.286 C-E
SA0.5 Cd4	0.406 K	0.139 J	0.195 K
SA1 Cd0	0.635 D	0.222 C-E	0.286 C-E
SA1 Cd1	0.632 D	0.234 C	0.285 C-E
SA1 Cd2	0.499 J	0.159 I	0.241 H-J
SA1 Cd4	0.515 I	0.168 H-G	0.220 J
SA1.5 Cd0	0.673 C	0.227 C	0.306 BC
SA1.5 Cd1	0.591 EF	0.205 EF	0.266 EG
SA1.5 Cd2	0.518 I	0.177 GH	0.233 IJ
SA1.5 Cd4	0.540 H	0.180 G	0.245 G-J
SA2 Cd0	0.639 D	0.237 C	0.291 CD
SA2 Cd1	0.597 E	0.233 C	0.264 E-H
SA2 Cd2	0.541 H	0.160 HI	0.256 F-I
SA2 Cd4	0.567 G	0.207 D-F	0.254 G-I

* Means followed by the same letters within columns are not significantly differed at $p \leq 0.05$ according to the test of Duncan's Multiple Range.

Table 3. Effects of foliar spraying with salicylic acid (SA), cadmium (Cd), and their interactions on some antioxidants compounds of *Lactuca sativa* L. Var. Romaine.

Treatments	Peroxidase (Unit. Gram ⁻¹)	Catalase (mol. Mint ⁻¹)	Ascorbic Acid (mg AA.100 g ⁻¹ dry sample of leaves)	Proline (Mm. gr ⁻¹ fresh weight)	Total Carbohydrate (%)
SA concentration (mM)					
SA 0	1226.66 B	24.79 B	0.954 AB	61.55 B	0.695 D
SA 0.5	1374.99 A	26.36 A	0.407 C	69.47 A	1.183 B
SA 1	1270.00 B	26.94 A	0.780 B	70.62 A	1.338 A
SA 1.5	1243.33 B	27.22 A	1.044 A	59.12 C	1.160 B
SA 2	1205.00 B	27.14 A	0.162 D	62.76 B	0.907 C
Cd concentration (mM)					
Cd 0	1060.66 B	29.05 A	0.382 C	46.62 D	0.728 C
Cd 1	1345.33 A	29.14 A	0.789 B	60.54 C	1.135 B
Cd 2	1286.66 A	24.32 B	1.266 A	73.39 B	1.224 A
Cd 4	1363.33 A	23.45 B	0.241 C	78.27 A	1.140 B
Interactions between SA and Cd					
SA0 Cd0	900.00 F	27.54 D-G	0.596 D-F	49.97 K	0.253 L
SA0 Cd1	1593.33 A	28.61 B-F	1.630 B	50.1 K	0.233 L
SA0 Cd2	1270.0 B-D	24.00 I-K	1.156 C	65.74 H	1.473 C
SA0 Cd4	1143.33 C-E	19.03 L	0.436 E-G	80.41 A-C	0.824 IJ
SA0.5 Cd0	1100.0 DE	26.83 F-H	0.193 GF	62.12 I	1.235 DE
SA0.5 Cd1	1383.33 B	29.46 B-E	0.246 E-G	67.18 GH	0.772 J
SA0.5 Cd2	1593.33 A	23.05 K	0.980 CD	69.84 FG	1.259 DE
SA0.5 Cd4	1423.33 AB	26.11 F-I	0.210 E-G	78.74 B-D	1.467 C
SA1 Cd0	1126.67 C-E	29.91 B-D	0.176 GF	54.82 J	1.011 GH
SA1 Cd1	1260.0 B-D	34.25 A	1.0 C D	74.74 E	1.265 DE
SA1 Cd2	1270.0 B-D	19.53 L	1.596 B	70.82 F	1.138 FE
SA1 Cd4	1393.33 B	24.100 I-K	0.350 E-G	82.10 AB	1.939 B
SA1.5 Cd0	1126.67C-E	30.60 B	0.703 C-E	35.48 M	0.928 HI
SA1.5 Cd1	1106.67 DE	25.68 G-J	0.930 CD	44.28 L	2.077 A
SA1.5 Cd2	1300.0 BC	27.95 C-F	2.406 A	82.36 A	1.072 FG
SA1.5 Cd4	1440.0 AB	24.65 H-K	0.140 GF	74.38 E	0.564 K
SA2 Cd0	1020.0 EF	30.37 BC	0.246 E-G	30.71 N	0.214 L
SA2 Cd1	1383.33 B	27.70 D-G	0.140 GF	66.41 H	1.329 D
SA2 Cd2	1000.0 EF	27.11 E-H	0.193 FG	78.20 CD	1.179 EF
SA2 Cd4	1416.67 AB	23.40 J K	0.070 G	75.74 DE	0.909 HI

* Means followed by the same letters within columns are not differed significantly at $p \leq 0.05$ according to the test of Duncan's Multiple Range.

Table 4. Effects of foliar spraying with salicylic acid (SA), cadmium (Cd), and their interactions on the percent of watery soft rot infection of lettuce (*Lactuca sativa* L.) var. Romaine.

SA concentration (mM)	Cd concentration (mM)				Average of SA
	Cd 0	Cd 1	Cd 2	Cd 4	
SA 0	0.100 A-D	0.113 A-D	0.130 A-D	0.066 D	0.102 AB
SA 0.5	0.103 A-D	0.083 B-D	0.083 B-D	0.083 B-D	0.088 B
SA 1.0	0.076 CD	0.100 A-D	0.103 A-D	0.066 D	0.086 B
SA 1.5	0.176 A	0.173 AB	0.110 A-D	0.103 A-D	0.140 A
SA 2.0	0.163 A-C	0.083 B-D	0.130 A-D	0.083 B-D	0.114 AB
Average of Cd	0.123 A	0.110 AB	0.111 AB	0.080 B	

Means followed by the same letters for each factor and their interactions are not differed significantly at $p \leq 0.05$ according to the test of Duncan's Multiple Range.

4. CONCLUSIONS AND RECOMMENDATIONS

From this study, we can conclude that foliar application of SA can be used to reduce the inhibitory effects of the stress of cadmium by improving the performance of root, chlorophyll A, both kinds of antioxidants (enzymatic and non-enzymatic) such as; peroxidase, catalase, proline, and total carbohydrates in plant leaves. High concentration of Cd inhibits most vegetative growth parameters and photosynthesis pigments, whereas low concentration increases enzymatic and nonenzymatic constituent content. It is recommended to conduct a study by applying cadmium in the soil and a study to examine the effects of SA and Cd at molecular levels.

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