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Impact of ascorbic acid on the concentration of Carotene and Prolin in the legume (*Vicia faba*) Plants grown with Soil treated with Pb and Ni

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ABSTRACT

This research was conducted at the university of Mosul / college of education for pure science / biology department, to detect the effect of ascorbic acid on the legume plant (*Vicia faba*) grown in a soil polluted with deferent concentration of heavy elements. The results showed that spraying the vegetative parts of the plant with ascorbic acid at 600 mg/l concentration and for once leads to an increase in carotene concentration and a decrease in proline concentration in the tissues of the leaves of legume plant in comparison with the control treatment as they reached (0.198) mg/g. Fresh weight and (0.058)µ / g fresh weight respectively. As it was noted that treatment of soil with element Ni at a concentration of 50 mg/kg soil leads to a decrease in the concentration of carotene in comparison to the control treatment as it reached (0.097) mg/g. Fresh weight, and an increase in proline when treated with element Pb at a concentration of 550 mg/kg soil as it reached (0.157) µ/g in comparison to with the control and other treatments.

Key words : *Ascorbic acid, Carotene and Prolin*

Introduction

Environment pollution has become an international problem as it is of great importance and has a direct relation with the lives of humans, animals, and plants. The pollution with heavy metals has many sources such as the industry and agriculture. In agriculture the addition of fertilizers (Iheanacho *et al.*, 2017) also using the insecticides (Chibuike and Obiora, 2014) has a great role in contaminating the soil with heavy metals. As in industry it's noticed that the human activity in the manufacturing processes also the metals extraction operations (Khalaf, 2017) caused a great increase in the contamination of environment with heavy metals. These metals do not only accumulate in the soil and plants but also it migrate across the food chain (Srivastava and Shukla, 2016) causing damage for the long term in human health (Rajeswari and Sailaja, 2014). Ascorbic acid has an effective role in preserving cells and their components from the harmful effects of reactive oxygen species that are formed when plants are exposed to inappropriate environmental conditions (Luma *et al.*, 2020). As it is one of the compounds that has the ability to defend plants as a cellular molecule that works with high potential as an antioxidant through its ability to assign electrons to a large number of non-enzymatic and enzymatic reactions (Gill and Tuteja, 2010). The aim of the study is to demonstrate the role of ascorbic acid in preserving legumes plants growing in soils treated with heavy elements through its effect on the concentration of carotene and proline in the leaf tissues of the plant.

Materials and Methods

Legumes (*Vicia faba*) seeds were obtained from the local markets in Nineveh / Iraq. They were planted in plastic pots with 5 kg volume in the green house at the department of biology / college of pure science / University of Mosul in the 10th /Oct /2020. The soil were treated with various amounts, the Pb treated with 450 and 550 mg/kg soil concentrations and Ni, with 25 and 50 mg/kg soil concentrations before plantation, after one month of growth the plants were sprayed with two concentrations 300 and 600 mg/l of ascorbic acid, then after another month the a group of the plants were sprayed again with 300 and 600 mg/l of ascorbic acid so that there would be two groups one that has been sprayed once and the other that been sprayed twice. After three months the leafs from the legumes were taken and underwent the following measures:

1. Measuring the concentration of carotene in leaf tissues according to Abdul Jallel *et al*., 2009
2. Measuring the concentration of proline in leaf tissues according to Bates *et al*., 1973.

Results and Discussion

Carotene concentration

Table 1 shows that the treatment of soil with Ni at 50 mg/kg soil concentration caused a decrease in the concentration of carotene in the leafs of legume in comparison to the control and other treatments as it reached (0.097) mg/g fresh weight. The same table showed that spraying the vegetative parts of legume with different concentrations at different occasions with ascorbic acid caused an increase in the concentration of carotene in the tissues of the leafs of legume especially when spraying them with the concentrations 300 and 600 mg/l one time only, the concentration caused significant effects it reached (0.176 and 0.198) mg/gm fresh weight respectively. And it was noticed that when spraying with 300 and 600 mg/l concentrations for two times the carotene concentration increases but didn't cause significant effects in comparision with the spraying them with the concentrations 300 and 600 mg/l one time only. As for the ratios of spraying to treatment with elements it was noted that the best outcome occurred when the legumes were sprayed twice with 600 mg/l concentration in a soil treated with the element Ni with 25 mg/kg soil concentration as it was (0.132) mg/g fresh weight.

As for the effect of ascorbic acid concentration, we note that spraying with concentration 600 mg/l compared to spraying with concentration 300 mg/l led to a significant superiority of carotene concentration in the leaf tissues of legume plants, and it was recorded as (0.129) mg/g fresh weight. As for the effect of the number of times spraying with ascorbic acid, no significant differences were found Also, the significant differences were not observed when comparing the effect of lead with nickel as the effect of the element type. The decrease in carotene concentration in the leaves of legume is due to the restriction in carotene formation caused by the effect of pollution metals in the soil that inhibit the absorption of the raw materials responsible for the constitution of carotene (Feng *et al*., 2010) also the heavy metals interfere in the biological processes of carotene. As for the increase in carotene concentration in the tissues of legume leafs when they are sprayed with ascorbic acid with 300 and 600 mg/l concentrations once or twice, it could be due to the fact that the ascorbic acid is one of the antioxidants which work in an opposite way against the free oxygen (Nudrat *et al*., 2017) alongside that it eliminates

Table 1. The effect of spraying the vegetative of legumes plants growing with soils treated with heavy metals with ascorbic acid on the concentration of carotene (mg / g fresh weight)

Effect of elements / Effect of Ascorbic acid		Control 0.0	Pb mg/kg 450	Pb mg/kg 550	Ni mg/kg 25	Ni mg/kg 50	Effect of the Ascorbic acid concentration	Effect of the number of times sprayed
		0.133 c	0.109 g	0.110 g	0.101 gh	0.097 h		
Sprayed once	300	0.176 b	0.112 g	0.111 g	0.117 e	0.114 ef	0.125 b	0.128 a
Mg/l	600	0.198 a	0.126 d	0.110 g	0.119 e	0.099 h	0.129 a	
Sprayed twice	300	0.134 c	0.119 e	0.117 e	0.123 de	0.122 de		0.125 ab
Mg/l	600	0.137 c	0.127 d	0.114 ef	0.132 cd	0.122 de		
Effect of element type			0.116 a		0.115 a			

the poisons and have a great role in the regulation of enzymes and photosynthesis (Xu *et al*., 2015).

Prolin concentration

Table 2 showed a significant increase in prolin concentration in the tissues of legume leafs that are grown in polluted soils with element Pb at 550 mg/kg soil concentration as it reached 0.157 µ/gm fresh weight in comparision to the comparison operator, this agrees with what (Al-Rashedy, 2020) stated; in the case a of a significant increase of prolin concentration in the tissues leafs of mint plant when they are exposed to different concentrations of cobalt and nickel. As there were a significant decrease in prolin concentration in the tissues of leafs of legume when the vegetative parts were sprayed with ascorbic acid only once at 300 and 600 mg/l concentrations as it reached (0.059, 0.058) µ/gm fresh weight. Table 2 also showed that the concentration of prolin decreased to (0.072) µ/gm fresh weight in the plants growing in soil polluted with element Ni at 25 mg / kg fresh weight concentration when they were sprayed with ascorbic acid with 300 and 600 mg/l concentrations twice in comparision to other comparative operators, as for the effect of ascorbic acid concentration, we note from the Table that spraying plants with a concentration of 600 mg/l led to a significant decrease in the concentration of prolin compared to spraying at a concentration of 300 mg/l, which amounted to 0.080 µ/g fresh weight. As for comparing the effect of the number of spraying times, we note that there are no significant differences between spraying plants once and spraying twice in the concentration of proline in the leaf tissues of legume plants. As for the effect of the element type, we notice a significant superiority of prolin concentration in the leaf tissue of legume plants growing with soils treated with lead, and it reached 0.100 µ/g fresh weight compared to its concentration in plants growing with soils treated with nickel. This is consistent with what (El-Beltagi *et al.,* 2020) found, in that treating chickpea plants suffering from stress with ascorbic acid resulted in significant changes in prolin concentration. The increase in the concentration of prolin in the tissues of the leafs of legume growing in polluted soil with heavy metals could be due to the fact that prolin is an antioxidant material that regulates and lowers the risks of active oxygen (ROS). In the plants that are exposed to harsh environmental conditions, prolin acts directly on establishing an equilibrium state between oxidation and reduction (Singh *et al*., 2015).

The decrease in prolin concentration in the tissues of leafs of legume when they are sprayed with ascorbic acid could be due to the great role of ascorbic acid in aiding the antioxidant enzymes in the plants that are exposed to an unappealing environmental conditions (Abdul Hameed *et al*., 2015).

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Table 2. The effect of spraying the vegetative of legumes plants growing with soils treated with heavy metals with ascorbic acid on the concentration of prolin (µ/g fresh weight)

Effect of element / Effect of ascorbic acid		Control 0.0	Pb mg/kg 450	Pb mg/kg 550	Ni mg/kg 25	Ni mg/kg 50	Effect of the Ascorbic acid concentration	Effect of the number of times sprayed
		0.078 ef	0.116 b	0.157 a	0.089 d	0.100 c		
Sprayed once	300	0.059 h	0.083 de	0.102 c	0.078 ef	0.087 d	0.084 a	0.080 a
Mg/l	600	0.058 h	0.082 e	0.089 d	0.079 ef	0.087 d	0.080 b	
Sprayed twice	300	0.076ef	0.077 ef	0.117 b	0.072 f	0.085 de		0.082 a
Mg/l	600	0.069 g	0.087 d	0.093 d	0.072 f	0.078 ef		
Effect of element type			0.100 a		0.083 b			

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