

Response of Pea (*Pisum sativum* L.) to Foliar Application of ABA and vitamin C and Interaction of them on some Physiological Characters of Plant

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Article Info	Abstract
Received 8/May/2017 Accepted 5/Dec./2017	<p>The field experimental was conducted during the growth winter season of 2016-2017 at Botanical garden of Department of Biology, College of Education for Pure Science (Ibn Al-Haitham), and University of Baghdad. The experimental aimed to study the effect of different of foliar application (0, 10, 20) mg.L⁻¹ and vitamin C (0, 50, 100, 150) mg.L⁻¹ and interaction of them on some physiological characters of pea (<i>Pisum sativum</i> L.). The results showed that ABA increased the proline and increased the vitamin C content at 10 mg.L⁻¹. The vitamin C increased absolute growth rate (AGR), biomass duration, proline, vitamin C content compared with control plants.</p> <p>Keywords: Pea (<i>Pisum sativum</i>), ABA, Vitamin C.</p> <p style="text-align: right;">الخلاصة</p> <p>أجريت تجربة حقلية للموسم الشتوي 2016-2017 في الحديقة النباتية لقسم علوم الحياة، كلية التربية للعلوم الصرفة (ابن الهيثم)، جامعة بغداد بهدف دراسة تأثير الرش الورقي بحامض الابسيسيك (ABA) بالتراكيز (0، 10، 20) ملغم لتر⁻¹ وحامض الاسكوربيك (فيتامين C) بالتراكيز (0، 50، 100، 150) ملغم لتر⁻¹ وتداخلهما في بعض الخصائص الفسلجية لنبات البازلاء <i>Pisum sativum</i> L. أظهرت النتائج ان حامض الابسيسيك عمل على زيادة حامض البرولين وزيادة محتوى فيتامين C وخاصة عند التركيز 10 ملغم لتر⁻¹ اما فيتامين C فقد عمل على زيادة معدل النمو المطلق واستدامة الكتلة الحيوية وحامض البرولين ومحتوى فيتامين C في النباتات المعاملة مقارنة بمعاملة السيطرة.</p>

Introduction

The pea plant (*Pisum sativum*) belong to Fabaceae family [1]. Green peas contains protein, fiber, carbohydrates, fats, vitamins A, B6 and C, as well as thymine, folic acid, minerals, phosphorus, zinc and calcium [2].

ABA is a growth regulator that plays a major role in influencing the plant's physiological processes, especially after drought or other stress. It reduces the permeability of the membranes and causes the closure of the pores [3].

Also, reduces vegetative growth of plants, dormant seeds and buds and inhibits cell division and expands [4]. ABA is synthesized in the chloroplast and increased their concentration at stress conditions where it converts the sugars into starch, resulting closure of the pore

[5]. Vitamin C (ascorbic acid) is a water-soluble vitamin that stimulates growth by activating many enzymatic reactions [6].

Vitamin C is a non-enzymatic antioxidant, the first line of defense, inhibits cell membrane oxidation, acts on the ROS snipe from cell [7], and plays an important role in controlling of oxidation and reduction processes, particularly stress resistance [8].

The present study aims to know the response of pea plant to the foliar application of both the ABA and vitamin C and their intraction in some parameters or physiological characteristics of the plant.

Materials and methods

A field experiment was carried out for the agricultural season 2016-2017 in the Botanical

Garden of the Department of Biology at College of Education for Pure Sciences (Ibn Al-Haytham) / University of Baghdad. The experiment was designed according to RBCD. The experiment consisted of 12 experimental units, 1 m × 1 m, at three replicates, seed planted in lines, 4 holes, and fertilized by urea (46% N) at average 60 kg. Ha-1 and super-triphosphate (46% P₂O₅) [9]. The treatments were as follows:

1. ABA 0, 10 and 20 mg.L⁻¹.
2. Vitamin C 0, 50, 100 and 150 mg.L⁻¹.

Seeds were planted on 15/10/2016 and took the first harvested on 8/1/2017 and the second harvested on 20/2/2017. The physiological characteristics of three replicates were studied to calculate the mean:

1. Crop growth rate (gm.day⁻¹) of the according to the following equation [10]:

$$CGR = (W_2 - W_1) / (T_2 - T_1)$$

where:

W₁ = dry weight of the vegetative group at the first period or harvesting of measurement.

W₂ = dry weight of the vegetative group at the second period or harvesting of measurement.

T₁ = the age of the plant when taking the first period or harvesting of the plant

T₂ = the age of the plant when taking the second period or harvesting of the plant

2. Biomass Duration (gm. Day⁻¹)

Calculated according to the following formula [11]:

$$\text{Biomass Duration} = (W_2 - W_1) (T_2 - T_1) / 2$$

W₁ = dry weight of the vegetative group during the first period or harvesting.

W₂ = dry weight of the vegetative group during the second period or harvesting

T₁ = the age of the plant when taking the first period or harvesting

T₂ = the age of the plant when taking the second period or harvesting.

3. Estimate the concentration of vitamin C (ascorbic acid) mg.100gm⁻¹ wet weight: 1 g of wet leaves were taken and added them 10 ml of oxalic acid 10.05 M. The samples were placed in shade for a day and then filtered and then taken the suspension at volume 2.5 ml and added 2.5 ml of oxalic acid and 0.5 ml of solution Ammonium molybdate (5%) and complements the volume to 25 ml with distilled water.

The samples are read along a 760 nm wavelength in Spectrum lab 22 and the correlation between vitamin C concentration and absorption values for each concentration was determined by using the standard curve to calculate vitamin C concentrations for all treatments [12].

4. Determination of the concentration of amino acid proline (µg. gm⁻¹): 0.5 g of wet leaves was taken and 10 mL of sulfosalicylic acid 3% was added and then transferred to the centrifuge. 2 mL of suspension was added to 2 ml of acetic acid and 2 ml of ninhydrin solution and then placed in a water bath until the appearance of red color. After that separated by the mixture device and then calculated the wavelength at 520 nm by compared with the standard curve of pure proline to determine the concentration of proline in the various treatments [13].

Statistical analysis:

The results were statistically analyzed according to the SAS program [14] to compare the calculation averages by using the least significant difference (L.S.D.) at the probability level of 0.05.

Results and discussion

The results of Table 1, indicate a decrease in crop growth rate (gm.Day⁻¹) of pea plant when treated with ABA, but not significant. For vitamin C, this was significantly increased (68.96, 65.51 and 82.75%) for concentrations 100 and 150 mg. L⁻¹ compared with the control plants either interaction was not significant.

The results of Table 2, show no significant differences between the treatments when application of the plant with ABA at concentrations 10 and 20 mg. L⁻¹. While vitamin C has a significant effect and increased biomass duration with rate 32.98% for 50 mg.L⁻¹ concentration and with an increase rate of 48.07% for 100 mg.L⁻¹ concentration. Also, concentration 150 mg. L⁻¹ increased with rate 52.28% compared to control plants. As for the interaction, it was not significant between the different treatments.

Table 1: Effect of foliar application with ABA and vitamin C on the crop growth rate (gm. Day⁻¹) for pea plant.

Concentration of ABA (mg. L ⁻¹)	Concentration of Vitamin C (mg. L ⁻¹)				Mean of concentration of ABA
	0	50	100	150	
0	0.032	0.051	0.061	0.083	0.057
10	0.027	0.057	0.050	0.045	0.045
20	0.028	0.040	0.048	0.056	0.043
Mean of concentration of Vitamin C	0.029	0.049	0.053	0.061	
L.S.D. (0.05)	For ABA	For vitamin C	For interaction		
	N.S	0.016	N.S		

Table 2: Effect of foliar application with ABA and vitamin C on Biomass Duration (gm. Day⁻¹) for pea plant.

Concentration of ABA (mg. L ⁻¹)	Concentration of Vitamin C (mg. L ⁻¹)				Mean of concentration of ABA
	0	50	100	150	
0	31.20	38.00	43.70	46.30	39.80
10	28.50	37.80	39.20	41.00	36.60
20	25.70	38.00	43.80	42.80	37.60
Mean of concentration of Vitamin C	28.50	37.90	42.20	43.40	
L.S.D. (0.05)	For ABA	For vitamin C	For interaction		
	N.S	5.98	N.S		

Table 3: Effect of foliar application with ABA and vitamin C on content of proline acid (mg. gm⁻¹ wet weight) for pea plant.

Concentration of ABA (mg. L ⁻¹)	Concentration of Vitamin C (mg. L ⁻¹)				Mean of concentration of ABA
	0	50	100	150	
0	9.20	11.72	12.25	9.39	10.66
10	8.78	13.9	8.33	12.80	10.77
20	13.24	10.12	12.19	11.68	11.81
Mean of concentration of Vitamin C	10.44	11.68	10.92	11.29	
L.S.D. (0.05)	For ABA	For vitamin C	For interaction		
	0.637	0.735	1.273		

The results of Table 3 show the amount of proline increase when sprayed with ABA significantly at rate 10.78% for 20 mg. L⁻¹ concentrations. While there is no significant increase of 10 mg. L⁻¹ concentrations was observed compared to control plants? When treated with vitamin C, proline significantly increased in concentrations 50 and 150 mg. L⁻¹ (11.87 and 8.14)% respectively compared to control plants, while there was no significant increase in the concentration of 100 mg. L⁻¹ of vitamin C compared to the control plants. For the interaction was the highest value and amounted to 13.24 µg.gm⁻¹ wet weight in the concentration of 20 mg. L⁻¹ for ABA and zero concentration

of vitamin C, and the lowest rate was 9.20 µg.gm⁻¹ wet weight of zero concentration for both ABA and vitamin C. The results of Table 4 indicates a significant increase in vitamin C content in leaves by rate 11.38% for the concentration mg. L⁻¹. While no change in concentration 20 mg. L⁻¹ of ABA compared to control plants. Vitamin C was significantly increased when spraying pea plants at rate (40.62, 9.78 and 59.72)% for concentrations 50, 100 and 150 mg.L⁻¹ respectively compared to the control plants. As for the interaction, the highest value was at concentration 50 mg. L⁻¹ for vitamin C and a concentration 10 mg. L⁻¹ in ABA, while the lowest value was for control plants.

Table 4: Effect of foliar application with ABA and vitamin C on content of vitamin C (mg. gm⁻¹ wet weight) for pea plant.

Concentration of ABA (mg. L ⁻¹)	Concentration of Vitamin C (mg. L ⁻¹)				Mean of concentration of ABA
	0	50	100	150	
0	76.00	116.00	105.70	127.70	106.30
10	97.30	150.00	85.80	140.30	118.40
20	87.30	100.70	94.70	84.30	107.80
Mean of concentration of Vitamin C	86.90	122.20	95.40	138.80	
L.S.D. (0.05)	For ABA	For vitamin C	For interaction		
	8.20	7.11	14.23		

The ABA effects on the metabolic activity and transfers in different parts of the plant and may responsible for the state of stunted growth and acts contrary to proline, oxycyn and cytokine [15]. ABA acts on a slow transformation of starches into sugars, accelerates the aging of leaves and their fall and increases the formation of ethylene due to the environmental stresses on the plant [16]. It was found that the treatment of wheat plant with ABA at in the elongation stage results a decrease in dry weight for vegetative group of the plant [17]. Proline is an important amino acid that has a role in protecting the plant from free radicals (ROS) and thus improves the plant's susceptibility to stress [18]. It was found that the treatment of leaves of plant wheat with ABA resulted in increased leaf content of proline [19]. Ascorbic acid (vitamin C) is catalyst and has a role in oxidation and reduction reactions and it is a non-enzymatic antioxidant [8]. It is a defense system that protects metabolic processes and acts as an adjunct to many plant hormones such as gibberellin, ethylene and salicylic acid and induces to formation of these hormones under stress conditions [20]. The crop growth rate and mass duration were significantly increased because of increase of the dry weight of the vegetative group. This finding is consistent with the researchers when application of the ascorbic acid with different concentrations on wheat plants [21, 28]. The treatment of plants with ABA puts the plant in a state of chemical stress. This stress promotes the accumulation of proline because it is a non-enzymatic antioxidant antagonistic to free radicals [23]. Proline

works to prepare the plant to tolerance different stresses and induce the responsible genes to endurance and the stability of cellular membranes. Vitamin C increases when treated with various concentrations of this vitamin because of this vitamin has the ability to snip free radicals directly and indirectly, which is conceder as the first line of defense of non-enzymatic antioxidants in plant cells. Also, inhibits oxidation of cellular membranes and maintain the metabolic processes in the plant and these results are consistent with Researchers on barley [25] and on wheat [26].

We conclude from this study that ABA has been shown to increase the proline content of plant from vitamin C and has not significantly affected on the crop growth rate and the duration of biomass. Vitamin C has increased the crop growth rate and the duration of biomass and proline and vitamin C content in treated plants compared to control plants.

Conclusions

The foregoing concluded that the ABA had no significant effect in the absolute growth rate and biomass duration of the pea plant. While the addition of an increased concentration of this acid had a positive effect on the leaves content of proline and vitamin C as well as that vitamin C had a significant effect on the absolute growth rate and the biomass duration and leaf content of proline acid and vitamin C.

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References

- [1] Al-Kateb, Y. M. Classification of seed plants, first edition. Dar Al-Kuttab for Printing and Publishing, University of Mosul, pp318, 1988.
- [2] Matlob, A. N. Ezz-El-Din, S. M. and Karim, S. A. Vegetable Production (Part 1). Dar Al-Kuttab for Printing and Publishing, University of Mosul, Iraq, 1980.
- [3] Verma, S. K. and Verma, M. T textbook of Plant Physiology, Biochemistry and Biotechnology. S. Chand and Company LTD. Ram Nager, New Delhi, 2008.
- [4] Jain, V. K. Fundamental of Plant Physiology. Cland & Company, LTD. Ram Nager, New Delhi, 2011.
- [5] Zhu, Y. D. Zhaou, X. Xia, Z. Wen, S. Y. Shen, J. Ma, J. Tu, C. and Fut, J., " The role of abscisic acid in early the development, " Plant Mol. Biol., vol . 2, pp.1-2, 2010.
- [6] Kefelic, V. I., " Vitamins and some other representative of non-hormonal plant growth regulars, Priki. Biochem, " Microbiol., vol. 17, pp. 5-15, 1981.
- [7] Quan, L. J. Zhang, B. Shi, W. W. and Li, H. Y., " Hydrogen peroxide in plant. Versatile Molecule of oxygen species network, " J. Intergr. Plant Biol., vol. 50, no. 1, pp. 2-8, 2008.
- [8] Arrigoni, O. and De Tullio, M. C., "Ascorbic acid much more than just an antioxidant. Biochem, " Et Biochem. Acta, vol . 1509, pp. 1-9, 2002.
- [9] Ali, H. G. Eis, T. A. and Jeda'an, H. M. Legume crops. Department of Field Crops, College of Agriculture, University of Baghdad, Press Higher Education and Scientific Research, Mosul. 1990.
- [10] Mnteith, J. L., " Reassessment of maximum growth rates for C₃ and C₄ crops," Exp. Agric, vol.14, pp.1-5, 1978.
- [11] Kvent, J. Svobal, J. and Fiala, K., "Canopy development in stands of *Typha latifolia* L. and *Phragmites communis*. Trin. South Maravid Hidro, " Biologia, vol.10, pp. 63-75, 1969.
- [12] Hussain, I. Khan, M. A. Khan, F. U. Ayaz, S. and Khan, F. U. UV., " Spectrophotometric analysis profile of ascorbic acid in medical plant of Pakistan. World Appl, " Sci. J., vol. 9, no. 7, pp. 800- 803. 2010.
- [13] Bates, I. S. Waldron, R. P. and Tears, J. D., " Rapid determinates of free proline for water stress studies, " Plant and Soil, vol.39, pp. 205-207, 1973.
- [14] SAS. Statistical Analysis System. Users Guide. Statistical-Version 9.1th ed. SAS. Inst. Inc. Cary. N.C. USA. 2012.
- [15] Magnon, M. Bruzzone, S. Guide, G. Darnonte, E. Searfis, C. Sturia, L. Palomba, D. Deflora, D. and Zocch, A., " Abscisic acid released by human monocytes activates and Vascular smooth muscle cell responses involved in atherogenesis, " J. Biol. Chem., vol. 284, no. 26, 2009.
- [16] Nambara, E. and Marion-Poll, A., " Abscisic acid biosynthesis and catabolism. Ann, " Rev. Plant Biol., vol. 56, pp. 165-185. 2005.
- [17] Al- Fattalawi, S. K. A-A. Effect of application of ABA in growing *Triticum aestivum* L. growing under different levels of water stress. Master Thesis, College of Education for Pure Sciences / Karbala University. 2013.
- [18] Turkan, I. and Demiral, T., " Recent development understanding salinity," Environ. Exp. Bot., vol. 67, pp. 2-9. 2009.
- [19] Bano, A. and Samina, Y., " Role of phytohormones under induced drought stress in a Wheat, " Pak. J. Bot., vol. 42, no. 4, pp. 2579-2587. 2010.
- [20] Khan, T. A. Majid, M. and Mohammed, F., " Ascorbic acid: an enigmatic molecule to developmental and environmental

- stress in plant, " Int. J. Appl. Biol. Pharm. Tech., vol.2, no. 3, pp. 468-483,2011.
- [21] Amin, A. A. Rashad, El-Sh. M. and Gharib, F. A. E., " Chary in morphological, physiology, and reproductive characters of wheat plants as affected by foliar application with salicylic acid and ascorbic acid. Auster, " J. Basic and Appl. Sci., vol. 2, no. 2, pp. 252-261,2008
- [22] El-Awadi, M. F. El-Lethy, S. K. and El-Rokick, K. G., " Effect of the two antioxidant, glutathione and Ascorbic acid on vegetative growth, yield and some biochemical changes I two wheat cultivars, " Plant Sci., vol. 2, no. 5, pp. 215-221, 2014.
- [23] Gupta, S. D., "Reactive oxygen species and antioxidant in higher plant, " J. Stress Physiol. Biochem., vol. 6,no. 3,pp. 64-90. 2011.
- [24] Johari-Pirvattou, M. Qasimov, N. and Maralian, H., "Effect of soil water stress on yield and proline content of four wheat times, " Agri. J. Biotech., vol.9,pp. 36-40, 2010.
- [25] Al-Arkawazi, A. L. A. Effect of foliar application of hydrogen peroxide and vitamin C on growth and yield of two cultivars of (*Hordeum vulgare* L.) planted in the dry farming land. PhD thesis, College of Education for Pure Sciences (Ibn Al-Haitham), Baghdad University. 2016.
- [26] Zhwan, K. H. and Khursheed, M. Q, "Effect of foliar application of ascorbic acid on growth, yield components and some chemical constituents of wheat under water stress conditions, Jord, " J. Agric. Sci., vol. 10, no. 1,pp, 1-15, 2014.