EFFECT OF TRYPTOPHAN AND ASCORBIC ACID ON GROWTH AND CHEMICAL CONSTITUENTS OF *KALANCHOE BLOSSFELDIANA* PLANTS.

Journal

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ABSTRACT

A pot experiment was carried out in the nursery of the Ornamental Horticulture Department, Faculty of Agriculture, Cairo University, during the two years (2006 and 2007) with the aim of studying the effect of foliar applications of tryptophan or ascorbic acid at different rates (100, 200 or 300 ppm) applied separately or in combinations with different concentrations plus the untreated control plants on growth behaviour and chemical constituents of *Kalanchoe blossfeldiana* plants. The most important results can be summarized as follows:

- Both tryptophan and ascorbic acids treatments significantly increased plant growth (in terms of plant height, number of leaves/plant, as well as fresh and dry weights of the plants and the photosynthetic pigments (chlorophyll A and B and carotenoids), total soluble sugars and total free amino acids.
- The effect of tryptophan was superior to that of ascorbic acid on increasing plant growth and chemical constituents. Tryptophan at the rate of 300 ppm or ascorbic acid at the rate of 200 ppm was the most effective treatments compared to the untreated plants and the other treatments.
- Tryptophan plus ascorbic acid at the rate of (300:100 ppm) respectively was the most effective treatment compared to the other combinations in producing the tallest plants, and the heaviest fresh and dry weights/plant.

Key words: Tryptophan, Ascorbic acid, Amino acids, *Kalanchoe blossfeldiana.*

INTRODUCTION

Most growers consider the kalanchoe plant a minor crop as flowering pot plants, with its variation in leaf shape, flower colours, and good keeping quality in the home. The kalanchoe has a good potential as a flowering potted plants for many growers. *Kalanchoe blossfeldiana* Poelin (family, *Crassulaceae*) is a native of Madgascar. Other related species originated from arid regions of tropical Africa. The kalanchoe is a succulent plants with fleshy leaves. The leaves are arranged along the stems in pairs, each pair at right angles to the pair above or below. The small, star-shaped florets are produced under short-day conditions. The inflorescence is a dichasial cyme that terminates in cincinni (Graf, 1992).

The role of tryptophan is well known it has an indirect role on the growth via its influence in auxin synthesis. Phillips (1971) reported that alternative routes of IAA synthesis exist in plants, all stating from tryptophan. Thus, when tryptophan was supplied to some plants tissues IAA was formed. Moreover, there have been reports that foliar application of amino acid tryptophan enhanced the vegetative growth and chemical constituents (Talaat and Youssef, 2002) in basil plants, Talaat (2005) on *Pelargonium graveolens* L. and Talaat *et al.,* (2005) on *Catharanthus roseus.*

Abou Dahab and Abd El-Aziz (2006) on *Philodendron erubescens* plants found that tryptophan significantly increased plant growth and the content of carotenoids, total soluble sugars and total free amino acids in the leaves. Also, they found that amino acids had no significant effect on chlorophyll A and B contents.

Ascorbic acid is currently considered to be regulators on plant growth and development owing to their effect on cell division and differentiation. Moreover, this changes in the level of ascorbic acid in response to ionic stress might be important in the regulation of ionic environment within the cell (Hanafy, 1996) on lettuce. Tarraf *et al.,* (1999) on lemongrass plants found that foliar application of ascorbic acid had positive effect on growth parameters. Abd-El-Aziz *et al.,* (2006) reported that ascorbic acid increased growth parameters of *Khaya senegalensis* plants. Abd-El-Aziz *et al.,* (2007) on *Syngonium podophyllus* plants found that ascorbic acid treatment significantly

promoted all growth parameters compared to the untreated plants. Golan-Goldhirsh *et al.,* (1995) on soybean indicated that, plants treated with ascorbic acid were showed increases in photosynthesis processes. Biacs *et al.,* (1988) on tomato stated that sugar content was increased by foliar spray of ascorbic acid. Singh *et al.,* (2001) on *Cassia angustifolia* and Talaat (2008) on sweet pepper detected that ascorbic acid foliar application increased the content of macronutrients (N, P and K).

The aim of this work was to study the effect of amino acid tryptophan as well as ascorbic acid and their combination on growth and chemical constituents of Kalanchoe.

MATERIALS AND METHODS

Pot experiment was carried out during the years (2006 and 2007) at the nursery of the Ornamental Horticulture Department, Faculty of Agriculture, Cairo University, with the aim of studying the effect of foliar applications of amino acid tryptophan and ascorbic acid on the growth behaviour and chemical constituents of *Kalanchoe blossfeldiana* plants.

Plant materials: Seedlings from *Klanchoe blossfeldiana* (4 months old with 6-8 leaves/plant and an average plant height of 10 cm.) were transplanted in plastic pots (16 cm diameter) filled with a mixture of sand plus peatmoss (1:1 v/v) on 15th of March 2006 and 2007. The seedlings were placed in a sunny area in the nursery. Each pot was supplied with 2 gm. from NPK (15:15:15) fertilizer after one month from transplanting and was repeated again after one month from the first application. Other common cultural practices were performed as needed. The plants were sprayed with amino acid tryptophan and ascorbic acid at the rates of (0, 100, 200 or 300 ppm) applied either separately, or in combinations at the rates of (100:100, 100:200, 100:300, 200:100, 200:300, 300:100 or 300:200 ppm) of tryptophan and ascorbic acid respectively in addition to the untreated plants (control). The treatments were applied one month after transplanting (on 15th of April in both years) and sprayed 5 times at one month intervals after first application.

The pots were arranged in a randomized complete blocks design, with 14 treatments replicated three times, each replicate contained of 7 plants.

At the termination date of the experiment (on 15th of November in both years), the following data were recorded:

Measurements of growth parameters:

- Plant height (cm)
- Number of leaves/plant
- Number of branches/plant
- Fresh and dry weights of leaves, branches and stems (g/plant).

Chemical analysis:

Fresh leaf samples were collected from the plants which were received the different treatments to determine the content of photosynthetic pigments (chlorophyll A, chlorophyll B and carotenoids) using the method described by Von Wettstein (1957).

Leaf samples were dried and the contents of total soluble sugars (carbohydrates) were determined according to Dubois *et al.,* (1956).

The content of total free amino acids in leaves was determined according to Rosein (1957).

Statistical analysis: Data obtained were subjected to analysis of variance and the means were analysed using the Duncan's Multiple Range Test (at the 0.05 level), as recommended by Snedecor and Cochran (1980).

RESULTS AND DISCUSSION

Effect of amino acids on plant growth:

1- Plant height (cm.):

Treated kalanchoe plants with tryptophan or ascorbic acid had a significant effect on plant height (Table 1), in the first season, the plant height ranged from 16.12 to 37.53 cm., whereas, in the second year, the recorded values ranged from 15.90 to 35.10 cm. The percentages of increase (compared to the control plants) ranged from 12.1 to 132.8% in the first year and from 15.3 to 120.7% in the second year. The highest percentage of increases was found in plants which were treated by the combination between tryptophan and ascorbic acid at the rate of 300:200ppm), this treatment gave the highest value in both years respectively.

From the above mentioned data one can observe that foliar spray with tryptophan alone at different rates 100, 200 or 300 ppm significantly increased plant height compared to the control and the percentage of increases were 18.3, 29.4 and 11.6% respectively. This means that tryptophan at the highest rate 300 ppm produced the longest plants compared to the control plants. This treatment was the most effective treatment in producing the longest plants in case of tryptophan treatments.

Table (1): Effect of tryptophan (T) and ascorbic acid (A) treatments on plant height (cm), number of leaves/plant, fresh and dry weights (g./plant) of kalanchoe plants.

Treatments	Plant height (cm)		Number of leaves/plant		Fresh weight (g.)		Dry weight (g.)	
	First season	Second season	First season	Second season	First season	Second season	First season	Second season
control.	16.12 J	15.90 I	19.00 H	18.69 G	63.08 H	61.23 I	6.91 H	6.30 F
Tryptophan (T) 100 ppm	19.08 I	18.34 H	22.00 C	21.22 DE	73.33 E	70.44 E	7.31 G	6.90 E
T. 200 ppm	20.87 H	19.74 G	21.20 D	20.93 E	76.63 D	73.11 D	7.83 EF	7.05 E
T. 300 ppm	35.08 B	32.11 B	25.00 A	23.50 A	89.32 BC	85.00 BC	9.27 B	8.90 A
Ascorbic acid (A) 100 ppm	27.58 E	25.90 D	20.00 G	19.90 F	67.02 FG	64.22 GH	7.37 G	7.00 E
A. 200 ppm	29.42 D	28.32 C	20.25 FG	19.80 F	71.55 E	68.11 F	7.27 G	6.99 E
A. 300 ppm	25.42 F	24.44 E	23.00 B	22.50 B	90.80 B	86.99 B	7.94 DEF	7.54 D
T. + A. (100:100)	29.25 D	28.65 C	22.75 B	21.75 CD	68.87 F	65.16 G	7.03 H	6.93 E
T. + A. (100:200)	34.00 C	32.11 B	20.60 EF	19.78 F	66.48 G	63.12 GHI	8.40 C	7.95 C
T. + A. (100:300)	21.58 GH	20.68 F	20.25 FG	19.66 F	87.48 C	84.00 C	9.67 A	8.65 B
T. + A. (200:100)	22.28 G	21.38 F	19.80 G	19.40 F	65.07 GH	62.08 HI	8.13 D	7.82 C
T. + A. (300:200)	21.67 GH	20.94 F	23.08 B	22.01 BC	91.23 B	87.11 B	7.78 F	7.00 E
T. + A. (200:300)	27.33 E	25.99 D	20.00 G	19.93 F	87.52 C	84.22 C	8.01 DE	8.45 B
T. + A. (300:100)	37.53 A	35.10 A	21.13 DE	20.78 E	99.96 A	96.10 A	9.480 AB	9.05 A

However, ascorbic acid treatments with the rates of 100, 200 or 300 ppm significantly increased plant height and the percentage of increases were 71,12, 82.5 and 57.6% respectively in the first season compared to control. This means that the most effective treatment in case of ascorbic acid was 200 ppm concentration.

In case of using both tryptophan plus ascorbic acid in the same treatment with the different rates. The data also indicated that these treatments had a significant effect on plant height and the percentages of increases in plant height ranged from 33.8 to 132.8% and the most

effective treatment was the combination of tryptophan and ascorbic acid at the rates of (300 : 200 ppm).

In general, the average percentages of increases in plant height due to tryptophan, ascorbic acid or both tryptophan plus ascorbic acid were 55.0, 70.4 and 77.7% respectively compared to the control. This indicated that the treatment with both tryptophan and ascorbic acid was the most effective treatment in general and (300:200 ppm) from tryptophan and ascorbic acid was the most effective treatment and can be recommend to be used in order to increase plant height in Kalanchoe plant.

The role of amino acids is stimulating growth of several plant species was studied by Phillips (1971) who indicated that several alternative routes of IAA synthesis exist in plants, all starting from amino acids. Russell (1982) reported that the increase in growth as a result of application of amino acids may be due to their conversion into IAA. Attoa *et al.,* (2002) reported that spraying Iberis plants with the amino acid tryptophan increased plant growth. Regarding the effect of amino acids on plant height, the results of this study are in agreement with those obtained by Salonen (1980) on *Atropa belladonna*, Moursy *et al.,* (1988) on *Datora stramonium*, El-Bahar *et al.,* (1990) on *Datora metal*, Gamal El-Din (1992) on *Hyosyeemus muticus,* Talaat and Youssef (2002) on *Ocimum basillicum* and Abo Dahab and Abd El-Aziz (2006) on *Philodendron erubescens* plants, they reported that foliar applications of amino acids significantly promoted plant growth. The data in the second season showed the same trend as in the first season.

In conclusion, it can be stated that tryptophan at the rate of 300 ppm or ascorbic acid at the rate of 200 ppm or the combination of tryptophan and ascorbic acid at the rate of (300:200 ppm) were the most effective treatments in producing a significantly increases in plant height of Kalanchoe plant.

Number of leaves/plant (Table 1):

It is also quite obvious that foliar application with tryptophan or ascorbic acids significantly increased the number of leaves/plant compared to the control plant. The number of leaves/plant ranged from 19.0 to 25.00 leaves/plant in the first season, whereas in the second year the average number of leaves ranged from 18.69 to 23.50 leaves/plant. The lowest value resulted from the control plant,

whereas, the highest one resulted from plants received tryptophan at the rate of 300 ppm.

Foliar application of tryptophan at the rate of 100, 200 or 300 ppm significantly increased the number of leaves/plant by 5.8, 11.5 or 31.55% over the untreated plants.

In case of ascorbic acid the percentages of increase due to the different concentrations were 3.5, 6.5 or 21.0% as a result of 100, 200 or 300 ppm application respectively.

The effect of the combination of tryptophan and ascorbic acid at different rates the data showed that these treatments significantly increased the number of leaves/plant and the percentages of increases ranged from 4.2 to 19.7% compared to the control in the first year. The highest percentage resulted from tryptophan plus ascorbic acid at the rate of (100 :100ppm). This means that this treatment was the most effective treatment in producing the greatest number of leaves/plant.

These results are in harmony with those obtained by Youssef and Talaat (2003). They found that thiamine and ascorbic acid significantly promoted vegetative growth of rosemary plants.

The data in the second season showed the same trend as in the first season. Abd-El-Aziz *et al.,* (2007) on *Syngonium podophyllum* plants reported that foliar application of thiamine, ascorbic acid or kinetin significantly promoted all growth parameters (plant height, number of leaves/plant) compared to the untreated plants.

Fresh and dry weights (g):

Data presented in Table (1) show that foliar application of tryptophan or ascorbic acid with different rates had a highly significant effect on increasing the fresh and dry weights of plants in both seasons. In the first season the fresh weight of the plants varied from 63.08 to 99.96 gm/plant. The lowest value resulted from the control plants, however, the highest value was obtained from plants treated by tryptophan or ascorbic acid alone with significant effect. The highest percentage of increase over the control reached 43 % at the rate of 300 ppm for tryptophan treatment and 44% in case of ascorbic acid at the rate of 200 ppm. This means that these two treatments were the most effective treatments plus the treatment of tryptophan and ascorbic acid at the rate of (300:100 ppm). This is the case in the first season, however, in the second season similar trend was observed.

Similar results were obtained by Abdel Halim (1995) on tomato plants Snirnoff (1996) and Taraaf *et al* (1999) mentioned that ascorbic acid has been implicated as a regulator of cell division.

Shaddad *et al.,* (1990) assumed that the effect of ascorbic acid on plant growth may be due to substantial role of ascorbic acid in many metabolic and physiological processes.

Abd El-Aziz *et al.,* (2007) reported that foliar application of thiamine ascorbic acid and kinetin to Syngonium plants significantly promoted plant growth and fresh and dry weights of plants.

Effect of amino acids on chemical constituents:

Photosynthetic pigments: The data presented in Table (2) show that the amino acid treatments which were used in this study had a significant effect on chlorophyll A contents. The untreated plants gave the lowest value (0.028 mg/100 g F.W.). However, the highest value (0.088 mg/100 g F.W.) resulted from tryptophan treatment at the rate of 300 ppm. The most effective treatments were tryptophan at the rate of 300 ppm and the combination for tryptophan at the rate of 300 and ascorbic acid at the rate of 100 ppm. These treatments significantly produced the highest value compared to the other treatments.

Table (2): Effect of tryptophan and ascorbic acid treatments on photosynthetic pigments of kalanchoe plants.

Treatments	Chlorophyll A (mg/100g F.W.)	Chlorophyll B (mg/100g F.W.)	Carotenoids (mg/100g F.W.)
control.	0.028 H	0.028 F	0.007 E
Tryptophan (T) 100 ppm	0.036 FGH	0.035 EF	0.009 DE
T. 200 ppm	0.047 CDEF	0.034 EF	0.027 A
T. 300 ppm	0.088 A	0.108 A	0.011 CDE
Ascorbic acid (A) 100 ppm	0.057 BC	0.093 AB	0.011 CDE
A. 200 ppm	0.034 GH	0.040 DEF	0.013 BCD
A. 300 ppm	0.053 BCD	0.061 CD	0.015 BC
T. + A. (100:100)	0.059 B	0.047 DEF	0.017 B
T. + A. (100:200)	0.033 GH	0.028 F	0.012 CDE
T. + A. (100:300)	0.042 DEFG	0.046 DEF	0.013 BCD
T. + A. (200:100)	0.080 A	0.058 CDE	0.011 CDE
T. + A. (300:200)	0.033 GH	0.034 F	0.018 B
T. + A. (200:300)	0.041 EFG	0.035 EF	0.014 BCD
T. + A. (300:100)	0.052 BCDE	0.075 BC	0.010 CDE

In general tryptophan treatment increased chlorophyll A contents compared to the untreated plants. This mean that amino acid treatment caused a significant increase in chlorophyll A contents compared to the untreated plants except the following treatments which had no significant effect (tryptophan at the rate of 100 ppm, ascorbic acid at the rate of 200 ppm; tryptophan plus ascorbic acid at the rates of 100:200 ppm and 200:100 ppm).

The same result was also obtained in case of chlorophyll B, however carotenoids contents showed that, tryptophan at the rate of 200 ppm and tryptophan plus ascorbic acid at the rate of 100:100 ppm were the most effective treatments in increasing the carotenoids contents compared to the other treatments.

The present data are in agreement with the findings of Milad (1998) on *Mentha virids*; Shoala (2000) on *Lavendula multifolia*, Hassanein (2003) on *Foeniculum vulgare* and Abo Dahab and Abd El-Aziz (2006) on *Philodendron erubescens* plants. They reported that foliar application of amino acids (tryptophan), caused an increase in the content of photpsynthetic pigments (chlorophyll A, B and carotenoids contents).

Total soluble sugars: Data in Table (3) indicate that the applications of either tryptophan or ascorbic acid as a foliar spray at different rates caused significant increase in the contents of total soluble sugars in the leaves. The most effective treatment in this respect was tryptophan at the rate of 300 ppm, this treatment produced the highest contents (27.04 mg/100g D.W.). However the lowest content (15.55 mg/100g D.W.) was found in the untreated plants.

In general we can observe that the different treatments which were used in this study increased the total soluble sugar contents in the leaves. The same results as in photosynthetic pigments.

These results are in agreement with those obtained by Talaat and Youssef (2002) on *Ocimum basillicum* and Wahba *et al.,* (2002) on *Antholyza aethiopica* plants.

Table (3): Effect of tryptophan and ascorbic acid treatments on total soluble sugars, and total free amino acids of kalanchoe plants.

Treatments	Total soluble sugars (mg/100g D.W.)	Total free amino acids (mg/g D.W.)
control.	15.55 H	0.020 H
Tryptophan (T) 100 ppm	25.00 B	0.038 ABCD
T. 200 ppm	25.18 B	0.038 ABCD
T. 300 ppm	27.04 A	0.043 AB
Ascorbic acid (A) 100 ppm	19.29 F	0.034 DEF
A. 200 ppm	23.22 C	0.042 ABC
A. 300 ppm	25.02 B	0.027 FG
T. + A. (100:100)	19.70 F	0.029 EFG
T. + A. (100:200)	25.14 B	0.024 GH
T. + A. (100:300)	19.14 F	0.044 A
T. + A. (200:100)	17.15 G	0.038 ABCD
T. + A. (300:200)	16.25 GH	0.042 ABC
T. + A. (200:300)	21.93 D	0.036 CDE
T. + A. (300:100)	20.88 E	0.031 DEFG

Total free amino acids:

The results recorded in Table (3) show that spraying Kalanchoe plants with either tryptophan or ascorbic acid caused a significant increase in the content of total amino acids in the leaves. All tryptophan rates (100, 200 or 300 ppm) significantly increased the total amino acids compared to the untreated plants. Also tryptophan plus ascorbic acid with different rates significantly increased the total amino acid in the leaves compared to the control plants. The most effective treatment in this concern was tryptophan plus ascorbic acid at the rate of 100:300 ppm.

These results are in agreement with the findings of Harridy (1986) on *Catharanthus roseus* and Abo Dahab and Abd El Aziz (2006) on *Philodendron erubescens* plants.

In conclusion, it can be stated that treatment of Kalanchoe plants with tryptophan (especially at the concentration of 300 ppm) was the most effective treatment and had a beneficial effect on plant growth and chemical constituents, followed by the treatment with tryptophan plus ascorbic acid at the rate of 300:100ppm for improving vegetative

growth and increasing fresh and dry weights. Whereas for chemical constituents the treatments of (300:200 ppm) and (100:300 ppm) were the best treatments for increasing photosynthetic pigments and total amino acids content.

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تأثير التربتوفان و حمض الأسكوربيك على النمو و المكونات الكيماوية لنبات الكلانشوا

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أجريت تجربة أصص في مشتل قسم بساتين الزينة بكلية الزراعة جامعة القاهرة خلال عامين متتاليين (٢٠٠٦ ، ٢٠٠٧) بهدف دراسة تأثير الرش بالتربتوفان و حمض الأسكوربيك بتركيزات مختلفة (١٠٠ ، ٢٠٠ ، ٣٠٠ جزء في المليون) على النمو و المكونات الكيماوية لنباتات الكلانشوا.

و يمكن تلخيص أهم النتائج التي تم التوصل إليها في الآتي:

- رش النباتات بالتربتوفان أو حمض الأسكوربيك بالتركيزات المستعملة (١٠٠، ٢٠٠، ٣٠٠ جزء في المليون) أدى إلى زيادة معنوية في نمو النباتات (ارتفاع النبات، عدد الأوراق على النبات، الوزن الطازج و الجاف للنبات) و كذلك الصبغات النباتية (كلوروفيل A ، B و الكاروتينويدات) و كذلك السكريات الكلية الذائبة و الأحماض الأمينية الحرة.
- تأثير استعمال التربتوفان كان أفضل من استعمال حمض الأسكوربيك في زيادة النمو الخضري و كذلك المحتويات الكيماوية.
- استعمال التربتوفان بتركيز ٣٠٠ جزء في المليون و كذلك حمض الأسكوربيك بتركيز ٢٠٠ جزء في المليون كانت أفضل المعاملات مقارنة بالتركيزات الأخرى.
- الرش بمخلوط حمض التربتوفان مع حمض الأسكوربيك بتركيز ١٠٠:٣٠٠ جزء في المليون على التوالي كانت أفضل المعاملات بالمقارنة باستعمال المخاليط بالتركيزات الأخرى في إنتاج أطول النباتات و كذلك أفضل وزن طازج و جاف.