Improving gladiolus growth, flower keeping quality by using some vitamins application

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Abstract: Response of growth, flowering quality and active chemical constituents of gladiolus plants by using some vitamins such as, thiamin, ascorbic acid and their combination during two seasons were studied .Plant which received the combined treatments of both vitamins recorded the highest growth, flowers quality and cormelets induction. Thiamine treatments had the lowest effect on photosynthetic pigments, while 200 ppm, thiamin+ 200ppm ascorbic acid, improved growth, delayed flowering opening of vase life , stimulated accumulation of carbohydrate and increased photosynthetic pigments and macronutrients status. Photosynthetic pigments and macronutrients.

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1. Introduction

Gladiolus flowers is considered a main exportable ornamental plants in Egypt, and the flower can be available the year around, the foreign markets demand Egyptian gladiolus with higher quality. Ascorbic acid (vitamins C) is a product of D-glucose metabolism in higher plants which affect on plant growth and development, and play a role in electron transport system (**El-Kobisy et al., (1**). Also, **Smirnoff et al., (2)** proposed a biosynthetic pathway and identified novel some enzymes. They also reported that ascorbate is synthesized from L-galactose via GDP-mannose and GDP- L galactose. Ascorbic acid also has been associated with several types of biological activities in plants such as in enzyme co factors, antioxidant, and as a donor / acceptor in electron transport at the plasma membrane or in the chloroplast (**Conklin (3**). A high level of endogenous ascorbate is essential effectively to maintain the antioxidant system that protects plants from oxidative damage **Cheruth (4**). Further, **Farahat et al., (5)** on *Cupressus sempervirn* L. reported that foliar application of ascorbic acid caused pronounced increases in vegetative growth and chemical constituents as well as essential oil percent.

Thiamine (vitamin B1) is a necessary ingredient for the biosynthesis of co- enzyme. Thiamine pyrophosphates , so it plays an important role in carbohydrate metabolism in plant. It synthesis in leaves and it transported to roots where it controls growth. Thiamine is an important factor for the translocation reactions of the pentose phosphate cycle which provides pentose phosphate for nucleotide synthesis and for the reduction of NADP required for various synthetic. **Youssef and Talaat (6)** found that pronounced increases in vegetative growth and chemical constituents of rosemary plants by foliar application of thiamine were observed.

The objective of the present study was to investigate the effect of ascorbic acid and thiamine on improving the flower characters and chemical constituents of gladiolus plant.

2. Materials and methods

A two pot experiment were carried out at the greenhouse of National Research Centre, Dokki, Egypt, during the two successive growing seasons of 2007/ 2008 and 2008/2009 to investigate the effect of ascorbic acid and thiamine on improving growth, flower characters and chemical constituents of gladiolus plant .The effect of gladiolus corms were kindly supplied by ornamental plants research, Ministry of Agriculture, Giza, Egypt. Ninform corms where sown on the first week of December in two seasons using plastic pots (30 cm diameter) that were filled with loamy sand soil, physical and chemical properties are illustrated in Table (1) which determined according to **Jackson (7).** Gladiolus corms were irrigated regularly with tap water to reach 80 % of water holding capacity of the soil by weighting the pots daily and the needed amount of water was added. All the normal culture practices of growing gladiolus corms were applied as usual manner. The experiment including 8 treatments in addition to the control which were two concentrations of thiamine (Th)and ascorbic acid (ASC)100 and 200 ppm applied each separately and/or in combination at concentrations of(100Th+100 ASC) ,(100 Th 100 + 200ASC), (200Th **+ 100** ASC)and (200 Th + 200 Th) , to the respectively in addition untreated plants.

Table (1): Some physical and chemical properties of the used soils.

Properties	Value	Nutrient content		Value
Clay %	13.1	P	mg/100 g soil	13.4
Silt %	24.1	K		59.2
Sand %	62.8	Mg		17.4
Texture	Loamy sand	Fe	ppm	5.8
EC dSm^{-1}	1.98	Mn		8.3
pH	7.73	Zn		0.84
$CaCO_3$ %	2.15	Cu		0.96

Foliar application of thiamine and ascorbic acid were carried out two times of 30 days intervals, starting at the first week of February at both seasons. The treatments were arranged in a complete randomized block design with six replicates each replicate contained four pots and each pot contained four plants.

At the flowering stage sample was taken from representative three replicates randomly for each treatment and other three replicates for vase life, and the following parameters were determined the parameters included the following data , plant height (cm), no. of leaves per plant, as well as fresh and dry weights of leaves (g/plant), no. of florets/spike, No. of cormlets, and fresh and dry weight of cormlets (g/plant).

The flower spikes were cuts in the 7.00 am, wrapped in Kraft paper in groups inside treatments and translocated to the laboratory in cool water for 3 hours. The experiment design was a completely randomized with three replicates with five spikes per treatments.

The spikes were placed in glass bottles containing of 500 ml of water and kept into laboratory at room temperature for 15 days and the following data were recorded

Percentage of flowers wilted (on 6 days) total water uptake by the spikes and vase-life (days)

Water uptake (ml/l) by the cut spikes was estimated by subtracting the amount of water at the end of experiment from the initial volume.

Flower opening percentage after 24 h of placing the spikes in glass bottle congaing of water.

Chemical analysis

Photosynthesis pigments (chlorophyll a , chlorophyll b)and total chlorophyll (a+b) as well as carotenoids content were determined in fresh leaves as mg/g FW according to **Saric et al., (8),** Total carbohydrates were determined using colorimetric method as described by **Herbert et al., (9).** Total nitrogen was determined by the methods of **Chapman and Pratt (10)** while phosphorus determination was carried out calorimetrically according to **King (11)**. Potassium was determined photo metrically by flame photometer method as described by **Brown and Lilland (12).**

Statistical analysis:

The recorded data (mean of the two seasons) were statistically analyzed on complete randomized design according to the procedure of **Snedcor and Cochran (13) Means were** compared by least significant differences(LSD 5% levels of probability).

3. Results

Effect of thiamine and ascorbic acid and their combination

Growth parameters:

It is evident from data in Table (2) that there are significant differences among the treatments and control in plant height, leaves number, fresh and dry weights (g/plant). The combined treatment of 200ppm thiamine +200 ascorbic acid followed by 200 thiamine + 100 ppm ascorbic acid were superior to the other treatments and control in previous mentioned parameters. The superiority of the combined treatment over the control was by about 63.41% for plant height, 53.98 % for number of leaves and 17.24 and 62.57 % for fresh and dry weights.

Table (2): Growth parameters of gladiolus plants as affected by some vitamins (mean of the two seasons).

Treatments	Plant height (cm)	Number of leaves /pl.	Leaves fresh weight	Leaves dry weight	FW /plant	DW/ plant
			(g/plant)			
Control	33.4	5.2	5.81	1.95	25.63	4.31
100 ppm Th	42.2	6.5	7.52	1.54	28.35	5.08
200 ppm Th	51.6	6.9	8.21	1.74	34.83	6.17
100 ppm ASC	47.8	8.4	10.54	2.07	36.74	6.97
200 ppm ASC	58.7	10.5	13.83	2.92	53.20	8.14
100 ppm Th+ 100 ppm ASC	49.4	7.7	9.35	1.84	56.21	9.16
100 ppm Th+ 200 ppm ASC	83.2	9.8	12.86	2.76	58.43	10.21
200 ppm Th+ 100 ppm ASC	88.0	10.7	14.58	3.62	59.09	11.34
200 ppm Th+ 200 ppm ASC	91.3	11.3	15.85	3.81	59.61	11.52
LSD (0.05)	2.12	0.23	0.43	0.11	1.12	0.21

Th=Thiamine ASC=ascorbic acid

Florets:

Foliar application of ascorbic acid, thiamine and their combination improved florets characters of gladiolus compared with the control plants. Combined treatment of 200 ppm ascorbic acid + 200 ppm thiamine was more affective than each treatment alone and all combined treatments. However, single treatments of ascorbic acid showed intermediate effect (Table 3).

Table (3): Flower characters of gladiolus plants as affected by some vitamins (mean of the two seasons).

Treatments	No. of florets/ spike.	Spike length	Diameter of florets	Length of florets	FW of florets/ spike	DW of florets/ spike
		(cm)			g/plant	
Control	3.52	10.1	2.45	4.21	14.51	2.18
100 ppm Th	3.84	14.8	3.83	5.83	14.83	2.37
200 ppm Th	5.71	18.5	4.74	6.32	19.77	3.22
100 ppm ASC	6.83	17.3	5.03	6.84	20.42	3.47
200 ppm ASC	8.51	20.4	6.08	7.75	33.40	6.02
100 ppm Th+ 100 ppm ASC	10.3	17.5	6.24	8.00	35.21	7.08
100 ppm Th+ 200 ppm ASC	11.0	22.7	7.00	8.52	36.78	7.11
200 ppm Th+ 100 ppm ASC	12.2	23.8	6.90	9.00	37.42	8.10
200 ppm Th+ 200 ppm ASC	13.8	25.7	7.42	9.21	38.93	8.23
LSD at 5%	0.58	0.81	0.12	0.05	0.11	0.04

Th=Thiamine ASC=ascorbic acid

Cormelets:

Table (4) show that application of ascorbic acid, thiamine and their combination had promotive effect on cormelets production of gladiolus compared with the control plants.

The low values were obtained by using 100 and/or 200 ppm thiamine while the combined treatment of 200 ppm thiamine + 100 ppm ascorbic acid induced the highest values for number of cormelets, fresh and dry weights (g/plant) . The increments amounted by 77.11 % for number of cormelets, 73.12 and 55.76 % for fresh and dry weights respectively over the control.

Table (4): Induction of gladiolus cormlets as affected by some vitamins (mean of the two seasons).

Treatments	No. of cormlets	Cormlets FW	Cormlets DW
		(g)	
Control	4.43	7.22	1.02
100 ppm Th	5.81	9.10	1.21
200 ppm Th	7.22	12.43	1.34
100 ppm ASC	8.42	13.63	1.95
200 ppm ASC	12.53	18.74	2.09
100 ppm T h+ 100 ppm ASC	14.22	22.11	2.00
100 ppm T h+ 200 ppm ASC	16.71	25.42	2.14
200 ppm T h+ 100 ppm ASC	18.11	26.83	2.29
200 ppm T h+ 200 ppm ASC	15.27	25.11	2.10
LSD (0.05)	0.56	0.31	0.04

Th=Thiamine ASC=ascorbic acid

Photosynthetic pigments:

Results in Table (5) indicated that spraying ascorbic acid, thiamine and/or their combination induced gladiolus leaves with intensive Chl a, Chl b and carotenoids compared with control plants. Single treatments of thiamine gave the lowest favorable effect on photosynthetic pigments Chl a, Chl b and carotenoids, while the combined treatment of 200 ppm thiamine + 100 ppm and/or 200 ppm ascorbic acid gave the greatest promoting effect.

Table (5): Photosynthetic pigments and carotenoids content as affected by some vitamins treatments(mean of the two seasons).

Treatments	Chl (a)	Chl (b)	a+b	Carotenoids
	mg/g/fw			
Control	0.513	0.216	0.729	0.483
100 ppm Th	0.587	0.312	0.899	0.593
200 ppm Th	0.583	0.311	0.894	0.611
100 ppm ASC	0.634	0.345	0.979	0.573
200 ppm ASC	0.687	0.367	1.054	0.613
100 ppm T h+ 100 ppm ASC	0.693	0.389	1.082	0.634
100 ppm T h+ 200 ppm ASC	0.745	0.422	1.167	0.678
200 ppm T h+ 100 ppm ASC	0.814	0.432	1.246	0.719
200 ppm T h+ 200 ppm ASC	0.810	0.430	1.240	0.713
LSD.5%	0.002	0.001	0.005	0.040

Th=Thiamine ASC=ascorbic acid

Macronutrients:

Table (6) indicated that spraying gladiolus plants with ascorbic acid, thiamine and /or their combination, gradually stimulated the concentration of N, P and K in gladiolus leaves, consequently single application of 200 ppm ascorbic acid increased N, P and K by about 74.16, 61.44 and 56.22 % over the control plants, respectively. While when 200 ppm was combined with 100 and /or 200 ppm thiamine , the highest P and K concentration were observed. However, it also clear that the combination between ascorbic acid and thiamine were more effective than each single treatment on N, P and K concentration.

Total Carbohydrates:

Data in Table (6) reveal also that vitamins treatments are favourable for increasing carbohydrate % whereas 200 ppm ascorbic acid + 200 ppm thiamine gave the greatest favourable effect, the increment over the control reached 68.18 %.

Table (6): Macronutrient status and total carbohydrates content of gladiolus plant as affected of some vitamins (mean of the two seasons).

Treatments	N	P	K	Total carbohydrates (%)
	mg/g DW			
Control	1.321	0.080	12.34	13.45
100 ppm Th	2.211	0.141	15.21	29.69
200 ppm Th	3.341	0.146	18.32	21.23
100 ppm ASC	4.423	0.152	18.79	30.45

200 ppm ASC	5.112	0.288	20.41	32.34
100 ppm T h+ 100 ppm ASC	5.642	0.219	22.32	31.23
100 ppm T h+ 200 ppm ASC	6.812	0.238	25.43	35.26
200 ppm T h+ 100 ppm ASC	7.234	0.346	28.22	32.84
200 ppm T h+ 200 ppm ASC	7.200	0.368	28.71	35.84
LSD 5%	0.230	0.004	0.82	1.28

Th=Thiamine ASC=ascorbic acid

Vase life:

Table(7) show that spraying gladiolus plants with vitamins induced significant difference in flowers opened percentage, flowers wilted ,water uptake and vase life compared with untreated plants ,the combined treatments of 200ppm thiamine +100 and/or 200ppm ascorbic acid was the most effective treatments for delaying flowers opening% decreasing the wilting flowers and it has also increasing effect on vase life(day).Maximum water uptake by flowers were recorded by 200 ppm thiamine+100 ppm ascorbic acid treatment.

Table (7): Keeping quality of gladiolus cut flowers as affected by some vitamins (mean of the two seasons).

Treatments	Flowers opened%	Flowers wilted%	Water uptake(ml)	Vase life (day)
Control	88.34	9.21	4.60	11.41
100 ppm Th	82.53	88.32	5.30	11.00
200 ppm Th	78.41	75.43	6.22	19.21
100 ppm ASC	71.25	86.24	6.40	19.82
200 ppm ASC	60.43	72.32	7.00	21.21
100 ppm T h+ 100 ppm ASC	57.48	66.43	7.30	20.41
100 ppm T h+ 200 ppm ASC	50.23	64.25	8.10	23.11
200 ppm T h+ 100 ppm ASC	48.41	55.61	9.30	25.64
200 ppm T h+ 200 ppm ASC	45.08	55.72	9.5	25.22
LSD (0.05)	0.230	0.004	0.82	1.28

Th=Thiamine ASC=ascorbic acid

4.Discussion

As expected some vitamins such as ascorbic acid and thiamine used in this experiment led to increase growth parameters of gladiolus plants with regard to plant height (cm), number of leaves, fresh and dry weights(g/plant). The present observation are fully corporate the finding of **El-Fawakhry and El-Tayeb (14)** on chrysanthemum, **Youssef et al (15)** on datura plants, **Mona and Iman (16)** on rose geranium and **Rawia et al**., **(17)** on *Jasminum grandiflorum*, **Nahed et al., (18)** on songonium, **Tarraf et al., (19)** on lemonegrass, **farahate et al**., **(5)** on *Cypressus sempervirent*.The regulatory effect of thiamine ,on meristem plant growth and development indirectly through enhancing the endogenous level of various growth factors as cytokinines and gibberellins **Youssef and Talaat (6).** However, thiamine synthesized in the leaves and transported to root to control growth **Kawasaki (20).**

Regarding ascorbic acid, **Price (21**) reported that it is the most abundant antioxidant which protect plant cell and currently considered to be regulators on plant growth owing to its effect on cell division and differentiation. Our results also shown that thiamine was less effective and has little influence than ascorbic acid in this respect. This could be evident from the work of **Sakr et al., (22)** on canola and **Abdel Aziz et al**., **(23)** on gladiolus. The increment is growth parameters due to ascorbic acid are largely due to stabilize member structures **Blockhina (24),** modulating membrane fluidity in a similar manner to cholesterol and also membrane permeability to small ions and molecules (**Foyer,25**)., implicated in the regulation of cell division by influencing progression from BI to S phase of cell cycle (**Smirnoff (26).**

Cormelet and flowers characters tended to increase in the presence of ascorbic acid and/or thiamine comparing with the case of complete absence of vitamin (control). Ascorbic acid was rather than thiamine tended to be more effective and showed the most beneficial effect, and the positive response appeared to be raised with rise in the concentration. The present observations are in agreement with the finding of **Sakr et al., (27**) on wheat. In this respect thiamine is an important factor for the translocation reaction of the pentose phosphate cycle, which provides pentose phosphate for nucleotide synthesis and for the role of NADP required for various synthetic pathway **Kawasaki, (20),** the author also added that, thiamine is a necessary for biosynthesis of Co-enzyme thiamine pro-phosphate, so it plays important role in carbohydrate metabolism which reflected on the increase in cormelets weights and numbers. The stimulatory effect of thiamine on flower characters was demonstrated by El-**Fawakhry and El-Tayeb (14)** on chrysanthemum flowers and **Rawia et al**., **(17)** on Jasmine, **Abdel Aziz (23)** on gladiolus . Similar results were obtained by **El-Quesni et al., (28)** on *Hibiscus rose sinesis* L. Ascorbic acid has been associated with several types of biological activities in plants, such as enzyme Co-factor, as antioxidant and as a donor/or acceptor in electron transport at plasma membrane on the chloroplast. Such increments in cormelets number in our result s may be due to increase in photosynthetic pigments which reflect on carbohydrate content and cormelets weight **Conktin, (29).**

Our results demonstrated that, single application of either ascorbic acid and/or thiamine led to increments in carbohydrates %, photosynthetic pigments and mineral ions contents N, P and K in gladiolus leaves. Apparently these observations may be due to that thiamine is a necessary for the biosynthesis of Co-enzyme thiamine pyrophosphate which plays a role in carbohydrate metabolism (**Kawasaki, 20).** The promotive effect of thiamine on total carbohydrates content may be due to their important role in the biosynthesis of chlorophyll molecule which in turn affected chlorophyll content **Youssef and Talaat,(18).**

Such observation were reported by **Hassanein (30)** on *Foeniculum vulgarire* L. and **Abo Dahab (31)** on *phelodedorn erubescene*, plants, they concluded that thiamine increase photosynthetic pigments in plants. The superiority of ascorbic acid in increasing growth parameters was accompanied by high promotive effect on the previous chemical constituents. Such finding show an analogy with those obtained by **Sakr et al., (22)** on Canola plants. In this connection ascorbic acid occurs in chloroplasts, vacuoles, mitochondria and cell wall (**Andrson et al**., **32, Ravtenkranz et al., 33).** The concentration in chloroplast can be high and probably related to its control photosynthesis **Foyer, (34).** Ascorbate acts as antioxidant (**Asoda, 35).** Co-enzyme co-factor **Davies et al.,(36).** Electrons donor for photosynthetic and mitochondrial electron transport **Asard et al**., **(37). Ascorbate** is produced of D-glucose metabolism which affects some nutritional cycle activity in higher plants **El-Kobisy et al., (1).**

The present data indicating further that supplemental addition of both thiamine + ascorbic acid greatly improved growth and chemical constituents of gladiolus growth, cormelets and flower induction with high quality and long vase life. Such positive increase could be explained by the fact that vitamins considered as a bio-regulators compounds which in little concentration exerted profound influence upon plant growth and production and may be due to the synergetic effect between ascorbic acid and thiamine in increasing plant growth and metabolism. This result hold true with finding of **Rawia et al., (17)** on *jasminum grandflorum*. Our results showed that the combined treatments between both vitamins ascorbic acid or thiamine sprayed to gladiolus plants induced flowers with high quality and long vase life. This observation are harmony with the finding of **Muhammad et al.,(38)** may be due to the pro motive effect of vitamins on most chemical constituents of plants.

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