

Effect of Some Preservative Solutions on Vase Life of Gladiolus Cut Flowers

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ABSTRACT: Keeping quality and length of vase life are important factors for evaluation of cut flowers quality, for both domestic and export markets. These investigations proposed to determine the effectiveness of some preservative solutions as glutamic acid (100, 200 and 300 mg/l), salicylic acid (200, 400 and 600 mg/l), calcium chloride (1000, 2000 and 3000 mg/l) and aluminum sulphate (100, 200 and 300 mg/l) on quality parameters of (*Gladiolus grandiflorus* L. cv. 'Rose Supreme') flowers. Results showed that all treatments significantly increased the vase life, fresh weight, water uptake and chlorophyll content with decreasing number of bacteria and proline accumulation compared to control. The highest increase in fresh weight, water uptake and chlorophyll index was obtained by glutamic acid (300 mg/l), salicylic acid (400 mg/l), calcium chloride (3000 mg/l) and aluminum sulphate (300 mg/l). Whilst, glutamic acid (300 mg/l) and salicylic acid (400) resulted in the maximum vase life in both experiments, in addition, glutamic acid (200 mg/l) and aluminum sulphate (200 and 300 mg/l) in second experiment. Moreover, glutamic acid (300 mg/l) recorded the most effect on decreasing number of bacteria and proline accumulation in both experiments, furthermore salicylic (400 and 600 mg/l), calcium chloride (3000 mg/l) and aluminum sulphate (100, 200 and 300 mg/l) on number of bacteria and salicylic acid (200 and 400 mg/l), calcium chloride (3000 mg/l) and aluminum sulphate (100, 200 and 300 mg/l) on proline accumulation in first experiment compared to control.

Key words: Gladiolus, vase life, preservative solutions, cut flowers

INTRODUCTION

Gladiolus (*Gladiolus grandiflorus* L.) is an ornamental plant native to South Africa. It belongs to family Iridaceae, having approximately one hundred and fifty known species. This plant is commercially used for cut flowers and occasionally used for landscape purpose. Gladiolus is one of the few plants which produce pleasant cut flowers with long spikes. It is cultivated in most of the tropical and subtropical countries of the world (Adil *et al.*, 2013). Gladiolus flowers are considered main exportable ornamental plants in Egypt, and the flower can be available year-around, the foreign markets demand Egyptian Gladiolus with higher quality (Abo-Leila and Eid, 2011). In addition, there is high demand of Gladiolus in the world as cut flower. In USA, 60 million Gladiolus spikes were sold in the market having worth of 16 million dollars which is 4.5% of total produced cut flowers in 2011 (USDA, 2012). Gladiolus is a popular cut flower in the world, but its longevity is very short. The vase life of individual florets is 4 to 6 days. Life of cut flowers is mainly affected by two main factors, namely ethylene which accelerates the senescence of many flowers and by microorganisms which cause vascular blockage and thus reduces the vase life of cut flowers (Mohammadi *et al.*, 2014). Several chemicals solutions were used as pulsing or preservative solutions for increasing the longevity of cut flowers. Those chemicals are very expensive and most harmful preservative for human causing irritating to skin, eyes and respiratory tract as well as using natural products did not have large attentions as safe materials in vase

solutions (Mohamed, 2015). The maintenance of vase life is an important quality attribute in these economically significant cut flowers. A suitable method for vase life extension, which is easy to use, natural, safe and inexpensive compounds is always crucial in this respect for large-scale applications (Soleimany-Fard *et al.*, 2013). Glutamine, a multifaceted amino acid used as an energy substrate for most cells. Glutamine plays an important role in the nitrogen and carbon skeleton exchange among different tissues, where this amino acid fulfils many different physiological functions (Zamani *et al.*, 2011). Salicylic acid (SA) is considered to be a potent plant hormone because of its diverse regulatory roles in plant metabolism. SA has been found to play a key role in the regulation of plant growth, development and in responses to environmental stresses. Further, its role is evident in ion uptake and transport, photosynthetic rate, stomatal conductance and transpiration (Tehranifar *et al.*, 2013). Calcium chloride is widely used as preservative and firming agent in the fruits and vegetables industry for whole and fresh-cut commodities (Martin-Diana *et al.*, 2007). Aluminum sulphate is used as an antimicrobial compound in commercial preservative solutions (Zadeh and Mirzakhani, 2012). Aluminum sulphate acidifies vase solution, diminishes bacterial proliferation and enhances water uptake (Liao *et al.*, 2001). The aims of this study were to determine the effectiveness of some materials (glutamic acid, salicylic acid, calcium chloride and aluminum sulphate) as preservation treatments to reduce *Gladiolus* cut flowers senescence and increase vase life. Moreover, to find out the most effective concentration among materials used to produce the best quality cut flower with longer vase life.

MATERIALS AND METHODS

Two separated experiments were conducted in the Plant Production Department, Faculty of Agriculture, Saba Basha, Alexandria University in (April and November, 2015) on *Gladiolus* cut flowers. This study was carried out to study the effect of glutamic acid, salicylic acid, calcium chloride and aluminum sulphate treatments on vase life of *Gladiolus* cut flowers (Rose Supreme variety). Cut flowers were obtained from a well-known commercial nursery in Cairo. Cut spikes were cut from the field in early morning, wrapped with polyethylene sheet, and then quickly moved to the laboratory, of an average temperature of (18° C ±1) and (50- 60 %) relative humidity and light from a white fluorescent lamp. Each stem was recut to a length of 60cm before postharvest treatments. Leaves of the lower third part of the stem were removed to avoid contamination in the vase solution as recommended by Khimani *et al.* (2005). After that, flower stems were pulsed in freshly solutions which prepared at the start of experiments from (concentrations of glutamic acid, salicylic acid, calcium chloride, aluminum sulphate) in plastic container for 24 hours. Then the flowers were moved to glass containers (vases) which contained 300 ml of tap water to calculate the vase life and the tested parameters.

Treatments and design

The treatments were arranged in a factorial experiment with Randomized Complete Block Design (RCBD) in two factors (Materials and Concentrations) with 3 replications. All data obtained throughout the course of this study were statistically analyzed by the analysis of variance as described by Steel and Torrie (1980), all analysis were done by Average of SAS (2002) statistical software. Cut flowers were pulsed in concentrations of glutamic acid (100, 200 and 300 mg/l), salicylic acid (200, 400 and 600 mg/l), calcium chloride (1000, 2000 and 3000 mg/l) and aluminum sulphate (100, 200 and 300 mg/l) with Litter tap water and 1% sucrose in the same time, control cut flowers pulsed in 2liter tap water and 1% sucrose for 24 hours.

Vase Life (days):

Was determined when the seventh floret in the spike wilted as recommended by Badr *et al.* (2008).

Total Fresh Weight (g):

The average fresh weight of fresh stems carrying leaves and the flowers were calculated at the full opening stage (Barakat, 2013).

Water Uptake (g):

The volume of water uptake was calculated by subtracting the volume of water evaporated from a control vase without cut flowers and the amount of water decreased in vases containing flowers (Zamani *et al.*, 2011).

Chlorophyll Index (SPAD):

Chlorophyll index was measured by chlorophyll meter (SPAD- 502, Minolta Co. Japan). Average of 3 measurements from different spots of a single leaves was considered (Yadava, 1986).

Number of Bacteria (CFU/ml):

Bacterial contamination was determined in the keeping solution at the end of experiment. The samples of the preservative solutions were taken (1 ml of each) and diluted using sterilized distilled water. One ml of each diluted solution was streaked on nutrient agar into Petri dishes. Cultures were incubated 2 days at 28°C and the colonies appearing on the plates were counted. This experiment was repeated two times with 3 replicates in each treatment at the laboratory of Microbiology Department, Faculty of Agriculture, Saba Basha, Alexandria University (Gendy and Mahmoud, 2012).

Determination of Proline Content in Leaves (µg proline/g):

Proline colorimetric determination proceeded according to Bates *et al.* (1973), Marin *et al.* (2009) based on proline's reaction with ninhydrin. For proline colorimetric determinations, a 1:1:1 solution of proline, ninhydrin acid and glacial acetic acid was incubated at 100°C for 1 hour. The reaction was arrested in an iced bath and the chromophore was extracted with 4 ml toluene and its absorbance at 520 nm was determined in a Bio Mate spectrophotometer (Thermo Spectronic).

RESULTS AND DISCUSSION

Total Fresh Weight (g)

Data in Tables 1 and 2 generally, revealed that, all treatments significantly increased fresh weight compared with the control in first and second experiment (April and November, 2015). In addition, the statistically analyzed data indicated that glutamic acid (300 mg/l), salicylic acid (400 mg/l), calcium chloride (3000 mg/l) and aluminum sulphate (300 mg/l) were more effective on increasing fresh weight than other treatments. The change in fresh weight of *Gladiolus* cut flowers was increased with increasing vase life periods.

Results are similar to those of Zamani *et al.* (2011) on Rose cut flower and Mazher *et al.* (2011) on *Codiaeum variegatum* L. plants, they observed that treatments with glutamic acids had a significant effect on fresh weight. These results seemed to be due to the reduction of MDA (malonyldialdehyde) accumulation, the microbial populations on vase solution of cut flower and ACC oxidase activity (Aminocyclo propane carboxylate oxidase) and improved membrane stability confirmed by Aran *et al.* (2011) similarly Kazemi *et al.* (2012c).

With regard to salicylic acid treatments, the findings proved to be in accordance to results of Mashhadian *et al.* (2012) on *Chrysanthemum* and Marandi *et al.* (2011) on *Gladiolus*, they reported that salicylic acid enhanced the fresh weight also, Sabzi *et al.* (2012) and Ashtari *et al.* (2013) on Rose cut flowers. We could return this increase of fresh weight by treatments of salicylic acid to its antimicrobial activity (inhibiting vascular blockage), it increases water uptake and decrease transpiration rate, thereby enhancing water balance of cut flowers which might be because of the possibility of salicylic acid to decrease pH of vase solution and consequently, the growth and proliferation of bacteria is reduce, which increase water uptake as proved by Soleimany-Fard *et al.* (2013).

Similarly, Sardoei (2014) and Ibrahim *et al.* (2011) on *Narcissus tazetta* and *Gerbera jamesonii*; respectively indicated that the effect of calcium chloride on fresh weight of cut flowers was significant. These results may be referred to the role of calcium in maintenance and modulation of various cell functions as the main role of integrated biocide in floral preservatives is to sustain clarity in vase solution and to avoid blockage of xylem elements by microorganisms as confirmed by Sardoei (2014). Moreover, Cortes *et al.* (2011) indicated that calcium increases tissue resistance by slowing senescence because it inhibits the synthesis or action of ethylene.

On other hand, Viradia *et al.* (2015) and Seyf *et al.* (2012) observed that aluminum sulphate significantly enhanced the fresh weight for longer period in Tuberose and Rose cut flowers compared to control treatments. In general, aluminum sulfate had significant effect on fresh weight loss, this might be related to solution uptake enhancement, improved water relations and prevent vascular blockage by microorganisms which finally resulted extension in vase life as indicated by Mohammadi *et al.* (2012) and Hussien and Yassin (2013).

Regarding the effect of vase life periods on the change in fresh weight of *Gladiolus* cut flowers it was found that fresh weight increased with increasing vase life periods and the differences among all tested vase life periods were statistically significant, except for the last sampling date (20 days) where the difference was significantly decreased in two separated experiment compared with initial time. The increase in fresh weight might be due to the improvements in water balance which is a major factor determines quality and longevity of cut flowers. It is influenced by water uptake and transpiration, being balance between these two processes. Low water uptake is often due to occlusions located mainly in the basal stem end and microbes are common cans of stem end blockage as described by Hajizadeh *et al.* (2012) and Sardoei (2014) who observed that obstruction of the xylem by bacteria, therefore, inability of water absorption by flower steams is one of the current problems that lead to decrease in flowers postharvest longevity and also early welter of them.

Table (1). Effect of some preservative solutions on fresh weight (g) of *Gladiolus* cut flowers "Rose Supreme" in the first experiment (April, 2015).

Total Fresh Weight (g) (April, 2015)						
Treatments	Vase Life (Days)					Average
	Initial Time	5	10	15	20	
Control	31.87e	53.04g	51.81e	37.13 d	14.07f	37.58f
Glutamic acid 100 mg/l	33.15d	55.49cde	53.55bcd	39.33bc	16.36cde	39.58de
Glutamic acid 200 mg/l	33.43c	55.72cd	54.25b	39.59bc	16.31cde	39.86cd
Glutamic acid 300 mg/l	33.81a	56.52a	56.41a	41.58a	18.25a	41.31a
Salicylic acid 200 mg/l	33.62b	55.34def	52.97cde	39.42bc	16.24cde	39.52de
Salicylic acid 400 mg/l	33.9a	56.25ab	55.77a	41.43a	18.03a	41.08a
Salicylic acid 600 mg/l	33.36c	55.67cd	52.50de	39.21bc	15.37e	39.22e
Calcium chloride 1000mg/l	33.12d	55.14ef	53.33bcd	39.26bc	15.71de	39.31e
Calcium chloride 2000mg/l	33.39c	55.73cd	53.69bcd	39.38bc	16.76bcd	39.79cd
Calcium chloride 3000mg/l	33.64b	56.21ab	55.64a	39.85b	17.65ab	40.60b
Aluminum sulphate 100mg/l	33.05d	55.00f	52.99cde	38.57c	16.20de	39.16e
Aluminum sulphate 200mg/l	33.34c	55.36def	53.37bcd	39.58bc	16.50cd	39.63de
Aluminum sulphate 300mg/l	33.58b	55.85bc	54.11bc	40.16b	17.34abc	40.21bc
Average	33.33d	55.49 a	53.88 b	39.58c	16.52e	39.76
LSD at 0.05	T: 0.47		D: 0.29		T×D: 1.07	
	T: Treatments	D: Vase Life	T×D: Interaction			

Table (2). Effect of some preservative solutions on fresh weight (g) of Gladiolus cut flowers "Rose Supreme" the in second experiment (November, 2015).

Total Fresh Weight (g) (November, 2015)						
Treatments	Vase Life (Days)					
	Initial Time	5	10	15	20	Average
Control	31.19c	55.77g	51.05e	31.98c	22.42g	38.48d
Glutamic acid 100 mg/l	31.24c	58.31edf	52.72cde	36.06ab	25.18cd	40.70bc
Glutamic acid 200 mg/l	31.54b	59.40cde	53.25cd	36.09ab	25.04cde	41.06bc
Glutamic acid 300 mg/l	31.89a	63.00a	57.67a	37.34a	28.83a	43.74a
Salicylic acid 200 mg/l	31.35bc	58.51def	53.18cd	36.18ab	23.84ef	40.61bc
Salicylic acid 400 mg/l	31.84a	61.28ab	56.93a	37.26a	27.52b	42.97a
Salicylic acid 600 mg/l	31.55b	58.57def	52.28de	33.95bc	23.25fg	39.92c
Calcium chloride 1000 mg/l	31.21c	57.01fg	54.48bc	36.15ab	23.95ef	40.56bc
Calcium chloride 2000 mg/l	31.39bc	58.02ef	54.48cd	34.84ab	25.28cd	40.66bc
Calcium chloride 3000 mg/l	31.57b	59.92bcd	56.18ab	34.11bc	25.96c	41.55b
Aluminum sulphate 100 mg/l	31.35bc	57.01fg	53.40cd	35.73ab	24.56de	40.41bc
Aluminum sulphate 200 mg/l	31.51b	58.47def	53.49cd	33.93bc	24.17def	40.31bc
Aluminum sulphate 300 mg/l	31.39bc	60.61bc	53.8cd	34.80ab	24.1def	40.94bc
Average	31.46d	58.91a	54.02b	35.26c	24.93e	40.92
LSD at 0.05		T: 1.34	D: 0.83	T×D: 3.05		
	T: Treatments	D: Vase Life	T×D: Interaction			

Water Uptake (g)

Data in Tables 3 and 4 revealed that all used materials significantly enhanced water uptake compared with the control in first and second experiment (April and November, 2015). Data also showed that glutamic acid (300 mg/l), salicylic acid (400 mg/l), calcium chloride (3000 mg/l) and aluminum sulphate (300 mg/l) caused the greatest water uptake. The increase of water uptake in glutamic acid pulsed stems might be due to decreasing accumulation of bacteria in vase solution which increased water absorption and ACC-oxidase activity (Aminocyclo propane carboxylate oxidase) which relatively has affected on the senescence process as proved by Aran *et al.* (2011). These results are compatible with the findings of Kazemi *et al.* (2012a) on Gerbera, who indicated that glutamine treatments increased cut flowers water absorption.

The enhancing effect of salicylic acid on water uptake may be related to the role of salicylic acid in reducing the microbial population in vase solution of cut flowers and/or positive regulatory role of SA on stomatal closure which regulates the rates of transpiration and increases the water-retaining capacity of leaves and petals as demonstrated by Kazemi *et al.* (2011a, b, and c) and

Khenizy *et al.* (2013). In addition, the role of salicylic acid is evident in ion uptake and transport and also photosynthetic rate, stomatal conductance and transpiration Khan *et al.* (2003). Similar results were obtained by Kazemi *et al.* (2011c) on Gerbera, Zadeh and Mirzakhani (2012) on Carnation and Soleimany-Fard *et al.* (2013) on Alstroemeria cut flower, they revealed that salicylic acid increased water absorption compared to control.

Concerning calcium chloride, results are similar with Sardoei (2014) findings on Narcissus (*Narcissus tazetta*), Ibrahim *et al.* (2011) on Gerbera, Farahat and Gaber (2009) on *Monestera deliciosa* and Cortes *et al.* (2011) on Rosa hybrid, they revealed that calcium chloride treatments increased water uptake compared to control. Thus seemed to be referred to the important role of calcium in increasing tissue resistance and delaying senility through preventing ethylene synthesis and its processing. It was shown that the use of calcium in vase solutions increases water flow through the stem by association with pectin in the xylem cell walls (Zadeh and Mirzakhani 2012).

Regarding aluminum sulphate, results are consistent with Nader *et al.* (2015), Seyf *et al.* (2012) on Rose cut flowers. These significant effect of aluminum sulphate on water uptake which observed in Tables 3 and 4 might be attributed to the action of aluminum sulphate which inhibited vascular blockage and increased absorption of water, ultimately increased the uptake of water in the spike Viradia *et al.* (2015) and Tsegaw *et al.* (2011).

Results also indicated that water uptake of Gladiolus cut flowers was increased with increasing vase life periods and the differences among all tested vase life periods were statistically significant, except for the last sampling date (20 days) where the difference was significantly decreased in both experiments compared with initial time. The decrease in water uptake of cut flowers during vase period was probably due to growth of microbes and vascular blockage suggesting that adding a suitable germicide in vase solution can prevent the growth of microbes and can increase water uptake as confirmed by Anjum *et al.* (2001). Hashemabadi *et al.* (2015) demonstrated that enhancement of vase life can be described with antimicrobial properties of the mentioned above compounds, so that water absorption improved with prevention of vascular blockage and it delays water deficiency related wilting and reported that anti-ethylene compounds and also antibiotics increase water absorption, significantly.

Table (3). Effect of some preservative solutions on water uptake (g) of Gladiolus cut flowers "Rose Supreme" in the first experiment (April, 2015).

water uptake (g) (April, 2015)						
Treatments	Vase Life (Days)					Average
	Initial Time	5	10	15	20	
Control	30.01e	58.91d	50.66d	40.26e	16.64d	39.30e
Glutamic acid 100 mg/l	32.13d	61.88bc	54.37bc	42.21bcd	18.43bc	41.80cd
Glutamic acid 200 mg/l	32.26cd	62.07bc	54.96bc	42.51bcd	19.25b	42.21cd
Glutamic acid 300 mg/l	33.65a	65.00a	55.98ab	45.53a	20.87a	44.21a
Salicylic acid 200 mg/l	32.31cd	62.93b	57.03a	42.17bcd	19.04bc	42.70cd
Salicylic acid 400 mg/l	33.15abc	65.02a	56.16ab	45.66a	20.60a	44.12ab
Salicylic acid 600 mg/l	32.51bcd	61.37bc	53.36c	40.82de	18.60bc	41.33d
Calcium chloride 1000mg/l	31.96d	62.12bc	53.59c	41.74cde	18.25c	41.5cd
Calcium chloride 2000mg/l	32.15d	61.13bc	54.64bc	41.79cde	18.51bc	41.33cd
Calcium chloride 3000mg/l	33.14abc	62.79b	55.58ab	43.39bc	18.86bc	42.75bcd
Aluminum sulphate 100mg/l	32.06d	60.76cd	54.65bc	42.13bcd	18.29c	41.58cd
Aluminum sulphate 200mg/l	32.02d	61.9bc	53.51c	43.41bc	18.76bc	41.92cd
Aluminum sulphate 300mg/l	33.28ab	62.62bc	55.22abc	43.65b	19.02bc	42.76bc
Average	32.36d	62.19a	54.60b	42.72c	18.86e	42.14
LSD at 0.05		T: 1.42	D: 0.88	T×D: 3.21		
	T: Treatments	D: Vase Life	T×D: Interaction			

Table (4). Effect of some preservative solutions on water uptake (g) of Gladiolus cut flowers "Rose Supreme" in the second experiment (November, 2015).

water uptake (g) (November, 2015)						
Treatments	Vase Life (Days)					Average
	Initial Time	5	10	15	20	
Control	27.07f	64.03d	42.24e	28.10d	18.57e	36.00f
Glutamic acid 100 mg/l	28.24cde	65.78cd	45.83bc	29.71c	19.20de	37.75de
Glutamic acid 200 mg/l	28.48bc	66.15cd	45.47cd	30.00c	19.86cd	37.99cd
Glutamic acid 300 mg/l	29.21a	70.77a	48.62a	32.58a	21.31a	40.49a
Salicylic acid 200 mg/l	28.25cde	67.14bc	45.43cd	29.58c	19.09de	37.90cde
Salicylic acid 400 mg/l	29.21a	71.26a	48.41a	32.6a	21.05ab	40.51a
Salicylic acid 600 mg/l	28.44bcd	65.08cd	43.64de	29.70c	18.84e	37.14e
Calcium chloride 1000 mg/l	27.99e	66.65bc	44.01cde	29.54c	19.01de	37.44de
Calcium chloride 2000 mg/l	28.20cde	66.47c	45.52cd	30.07c	19.45cde	37.94cde
Calcium chloride 3000 mg/l	28.78b	69.01ab	47.55ab	31.25b	19.88cd	39.29b
Aluminum sulphate 100 mg/l	28.04de	65.33cd	44.63cd	29.60c	19.29de	37.38de
Aluminum sulphate 200 mg/l	28.52bc	65.95cd	45.22cd	29.90c	19.82cd	37.88cde
Aluminum sulphate 300 mg/l	28.82ab	67.03bc	45.69bc	31.12b	20.22bc	38.58bc
Average	28.40d	66.97a	45.56b	30.29c	19.66e	38.18
LSD at 0.05		T: 0.81	D: 0.50	T×D: 1.83		
	T: Treatments	D: Vase Life	T×D: Interaction			

Total Chlorophyll (SPAD)

Data of the present investigation, listed in Tables 5 and 6 showed that in both experiments (April and November, 2015), all treatments caused delay in degradation of total chlorophyll and preserved total chlorophyll content

compared with the control. Moreover, statistical analysis of these data proved that the most significant increase in chlorophyll leaf content than other treatments was recorded by treatment of glutamic acid (300 mg/l), salicylic acid (400 mg/l), calcium chloride (3000 mg/l) and aluminum sulphate (300 mg/l). On the other hand, our data in the first experiment (April, 2015) revealed that the change in total chlorophyll content of Gladiolus cut flowers was increased with increasing vase life periods.

We could attribute the delay of chlorophyll degradation in flowers which pulsed in glutamic acid to its action on inhibiting ACC-oxidase activity (Aminocyclo propane carboxylate oxidase) that is the direct precursor of ethylene and decrease ROS (reactive oxygen species) with increase enzyme antioxidant activity decrease number of bacteria and ACC-oxidase activity (Kazemi and Ameri, 2012a). These results were found in agreement with those of Kazemi *et al.* (2012a) and are in accordance with those of Kazemi and Ameri (2012a) on Carnation. Similarly, Zamani *et al.* (2011) on Rose cut flowers.

Likewise, results of salicylic acid were found in accordance to those of Bayat *et al.* (2011) and Kazemi *et al.* (2012c) on Carnation cut flowers and Mohammadi *et al.* (2014) on Gladiolus cut flowers. The effect of salicylic acid might be due to the effect of salicylic acid that affect postharvest life of cut flowers probably via the declining bacterial growth, reducing vascular blockage, reducing transpiration, preventing ethylene formation and inducing antioxidant system in treated cut flowers thereby delaying the senescence process as reported by Danaee *et al.* (2013).

With respect to calcium chloride, results were found in conformity with those of Abdolmaleki *et al.* (2015) on Rose and Zadeh and Mirzakhani (2012) on Carnation, they indicated that calcium chloride have significantly increased chlorophyll content in leaves. These results may due to the important role in increasing tissue resistance and delaying senility through preventing ethylene synthesis and its processing as found by Zadeh and Mirzakhani (2012) and Cortes *et al.* (2011). On the other hand, this salt is already known to lower the respiration as confirmed by Anjum *et al.* (2001). In addition, the calcium ion also seems to affect ethylene action on cell membranes by inhibiting ion leakage and reducing the effect of ethylene on senescence as proved by Asfanani *et al.* (2008).

Effectiveness of aluminum sulphate on delaying the degradation of chlorophyll might be referred to the role of aluminum sulphate as a microbial inhibitor, reducing the transpiration rate and stimulate the minimum ethylene production as reported by Hashemabadi *et al.* (2015), Mohammadi *et al.* (2012) and Tsegaw *et al.* (2011). They are in harmony with findings of Jowker *et al.* (2012) and Hajizadeh *et al.* (2012) on Rose, they stated that aluminum sulphate lead to a considerable delay in degradation of chlorophyll compared to control.

On the other hand, present data in the first experiment (April, 2015) revealed that the change in total chlorophyll content of Gladiolus cut flowers was increased with increasing vase life periods and the differences among all

tested vase life periods were statistically significant, except for the last three dates of sampling (10,15 and 20 days) where the difference was significantly decreased compared with initial time, while the data showed a significant increase in chlorophyll content with increasing periods of the life of flowers of *Gladiolus* cut flowers in the second experiment (November, 2015). Moreover, there were significant differences between all tested stages of vase life, except for the latest stage (20 days) which decreased compared with initial time. The greatest value in chlorophyll content was obtained by 5th day compared to 0 day and 20th day in both experiments of *Gladiolus* (10th and 15th day in first experiment). Leaf yellowing is a form of senescence caused by an internal hormone imbalance, such as a lack of cytokinins as confirmed by Ferrante *et al.* (2004). The maintenance of green color in the leaves is an important quality property in these economically significant ornamental plants. Previous study had revealed that the leaf yellowing of cut flowers is associated with chlorophyll breakdown and loss, thereby decreasing significant vase life Jahanbazi *et al.* (2014).

Table (5). Effect of some preservative solutions on chlorophyll index (SPAD) of *Gladiolus* cut flowers "Rose Supreme" in the first experiment (April, 2015).

Chlorophyll index (SPAD) (April, 2015)						
Treatments	Vase Life (Days)					Average
	Initial Time	5	10	15	20	
Control	46.63c	48.43f	46.01d	42.91c	40.01e	44.80f
Glutamic acid 100 mg/l	47.96b	50.43cde	48.22c	44.73bc	42.89cd	46.85de
Glutamic acid 200 mg/l	48.58b	50.67bcde	48.58c	45.04ab	43.41cd	47.26de
Glutamic acid 300 mg/l	51.56a	52.63a	50.17a	48.54a	45.9a	49.75a
Salicylic acid 200 mg/l	48.08b	50.96bc	48.49c	44.82bc	43.49cd	47.17de
Salicylic acid 400 mg/l	51.19a	52.93a	49.88ab	48.49a	46.12	49.72a
Salicylic acid 600 mg/l	50.38a	50.81bcd	48.59c	44.39bc	42.58cd	47.35cde
Calcium chloride 1000 mg/l	48.26b	49.97e	48.35c	43.83bc	42.92cd	46.67de
Calcium chloride 2000 mg/l	48.58b	50.68bcde	48.36c	43.98bc	43.36cd	46.99de
Calcium chloride 3000 mg/l	50.83a	51.27b	48.83c	45.02bc	44.17c	48.02bc
Aluminum sulphate 100 mg/l	48.03b	50.07de	48.51c	44.62bc	41.97d	46.64e
Aluminum sulphate 200 mg/l	48.62b	50.97bc	48.86bc	45.72b	42.80cd	47.39cd
Aluminum sulphate 300 mg/l	50.36a	51.45b	49.02bc	46.04ab	44.22bc	48.22b
Average	49.16b	50.87a	48.60c	45.24d	43.37e	47.45
LSD at 0.05	T: 0.74		D: 0.46		T×D: 1.67	
	T: Treatments	D: Vase Life	T×D: Interaction			

Table (6). Effect of some preservative solutions on chlorophyll index (SPAD) of Gladiolus cut flowers "Rose Supreme" in the second experiment (November, 2015).

Chlorophyll index (SPAD) (November, 2015)						
Treatments	Vase Life (Days)					
	Initial Time	5	10	15	20	Average
Control	48.94f	61.04f	57.93c	51.04c	48.18d	53.43g
Glutamic acid 100 mg/l	49.85de	62.87def	60.48b	54.11b	48.74d	55.21f
Glutamic acid 200 mg/l	50.21bcd	63.44de	60.54b	55.65ab	49.09bcd	55.79def
Glutamic acid 300 mg/l	51.00a	67.30a	64.33a	56.81a	51.51a	58.19a
Salicylic acid 200 mg/l	50.02cde	63.66cde	60.21bc	55.3ab	49.01bcd	55.64def
Salicylic acid 400 mg/l	50.97a	66.99a	64.19a	55.12ab	51.13a	57.68ab
Salicylic acid 600 mg/l	50.10cd	63.11de	60.12bc	55.55ab	49.14bcd	55.60def
Calcium chloride 1000mg/l	49.51e	63.81cde	60.33b	53.90b	48.34d	55.18f
Calcium chloride 2000mg/l	49.68de	65.52abc	60.93b	55.75ab	48.98cd	56.17cde
Calcium chloride 3000mg/l	50.50abc	66.12ab	61.81b	56.66a	49.87bc	56.99bc
Aluminum sulphate 100mg/l	49.91de	62.58ef	59.92bc	55.58ab	48.82d	55.36ef
Aluminum sulphate 200mg/l	49.95de	64.46bcde	60.79b	55.78ab	48.78d	55.95def
Aluminum sulphate 300mg/l	50.73ab	64.54bcd	59.67bc	56.52a	50.00b	56.29cd
Average	50.11d	64.27a	60.87b	55.21c	49.35e	55.96
LSD at 0.05		T: 0.88	D: 0.55	T×D: 1.99		
	T: Treatments	D: Vase Life	T×D: Interaction			

Determination of Proline (μg proline/g)

The effect of some preservative solutions on accumulation of proline in leaves of Gladiolus cut flowers in both experiments (April and November, 2015) are presented in Table (7). It has been found that all studied materials had remarkable significant effect on decreasing accumulation of proline compared to control in both experiments. Except for glutamic acid at (100 and 200 mg/l), the last concentration of salicylic acid (600 mg/l) and calcium chloride at (1000 and 2000 mg/l) which did not show significant differences in comparison with control in the first experiment. Moreover, there were no significant differences between the other treatments, except the differences between intermediate level of aluminum sulphate (200 mg/l) compared to the first level of aluminum sulphate (100 mg/l) in the first experiment. While, the greatest effect on decreasing accumulation of proline was recorded by the higher concentration of glutamic acid (300 mg/l) in the second experiment.

Similar results were found by Kazemi *et al.* (2012c) on Carnation, they showed that glutamic acid decrease proline accumulation significantly. These effect of glutamic acid might be referred to the decreases in ACO activity (Aminocyclo propane carboxylate oxidase that is the direct precursor of ethylene as confirmed by Kazemi *et al.* (2012a). We could return the effect of

salicylic acid to improving the antioxidant system and reducing oxidative stress damages during flower senescence as observed by Gerallio and Ghasemnezhad (2011). In addition the inhibition of ethylene biosynthesis and prolong vase life (Marandi *et al.*, 2011). These result are in conformity with those of Kazemi and Shokri (2011) on Lisianthus flowers.

Furthermore, the significant effect of calcium chloride is in agreement with Ibrahim *et al.* (2011) on Gerbera and Zadeh and Mirzakhani (2012) on Carnation. They showed that calcium chloride improved vase life and leaf quality. These effectiveness might be related to calcium chloride ability to inhibit the synthesis or action of ethylene. In addition, its role of increase tissue resistance and lower the rate of respiration as proved by Cortes *et al.* (2011) and Anjum *et al.* (2001). Regarding the effect of aluminum sulphate, results are similar to those of El-Quesni *et al.* (2012) on Schefflera, they found that aluminum sulphate increased vase life and quality. May be this results due to its action as an antimicrobial agent in the solution Hussien and Yassin (2013) and Mohammadi *et al.* (2012) Jowker *et al.* (2012) they found that aluminum sulphate improved postharvest visual quality by retaining leave freshness even at the end of vase life.

Number of Bacteria (CFU /ml)

The effect of some preservative solutions on the number of bacteria of Gladiolus cut flowers "Rose Supreme" in first and second experiment (April and November, 2015), are shown in Table (7), proved that number of bacteria in vase solution decreased significantly by using all studied materials compared to control. Moreover, the best effect on decreasing number of bacteria was obtained by glutamic acid at (300 mg/l), salicylic acid at (400 and 600 mg/l), calcium chloride at (3000 mg/l) and all concentrations of aluminum sulphate (100, 200 and 300 mg/l) in the first experiment. Whilst glutamic acid at 300 mg/l resulted in the greatest effect on decrease number of bacteria in vase solution of Gladiolus cut flowers in the second experiment.

The positive effect of glutamic acid may be that glutamic acid is readily metabolized by plants but not by many microorganisms, so it considered to using it as a possible substitute for sucrose according to Aran *et al.* (2011) and Kazemi and Ameri (2012a). Similar results were obtained by Kazemi *et al.* (2012c) on Carnation cut flowers, who found that glutamic acid decreased microbial population on vase solution significantly. As for main effect of salicylic acid, results are in accordance with those of Kazemi and Ameri (2012a) and Kazemi *et al.* (2012c) on Carnation. They observed a significant effect of salicylic acid on bacterial population. Effectiveness of salicylic acid might be referred to its ability to decrease pH of vase solution which reduce the growth and proliferation of bacteria as confirmed by Soleimany-Fard *et al.* (2013).

On the other hand, the best effect on decreasing the number of bacteria by calcium chloride may be attributed to the absence of a carbon source which inhibited bacterial growth as proved by Cortes *et al.* (2011). These results are in harmony with those of Hashemabadi *et al.* (2015) on Carnation cut flowers who reported that anti-ethylene compounds and also antibiotics increase water absorption, significantly. The significant control on number of bacteria that

obtained in aluminum sulphate pulsed flower stems may be related to not only limited to lowering the pH of vase solution but also its effect is based at least in part, on its action as an antimicrobial agent in the solution (Hussen and Yassin 2013). These results are in agreement with those of Jowker *et al.* (2012) on Rose cut flowers who showed that aluminum sulphate inhibited microbial proliferation in vase solution.

Vase Life (Day)

Data presented in Table (7) cleared the effect of some preservative solutions on vase life of Gladiolus cut flowers "Rose Supreme" in first and second experiment (April and November, 2015). It has been proved from our results that, in general the four compounds which used in the two experiments showed positive effect on increasing vase life of Gladiolus cut flowers in comparison with control in both experiments. On the other hand, the last concentration of glutamic acid at (300 mg/l) and the intermediate concentration of salicylic acid (400 mg/l) resulted in the highest increase in the vase life in first experiment. Whereas, the best effect was found in glutamic acid at (200 and 300 mg/l), salicylic acid at (400 mg/l) and aluminum sulphate at (200 and 300 mg/l) in the second experiment.

With respect to glutamic acid effects, results are similar to the findings of Kazemi *et al.* (2012) on Carnation cut flowers, Kazemi *et al.* (2012a) on Gerbera and Zamani *et al.* (2011) on Rose. They showed that treatments of glutamine significantly increased vase life and delayed flower senescence compared distilled water (control). These enhancement on vase life duration may be due to glutamic acid ability to control microbial contamination, so that improved water absorption, prevented vascular blockage and delayed water deficiency related wilting and control ethylene production that extend vase life of cut flowers (Hashemabadi *et al.*, 2015). Similarly, Zamani *et al.* (2011) on Rose. Further, Kazemi and Ameri (2012a) stated that all glutamine concentrations prolonged Carnation vase life, while decreasing accumulation of bacteria in vase solution and ACC- oxidase activity (Aminocyclo propane carboxylate oxidase). Furthermore it has been proved that the observed decrease in ACC activity could be at least one mechanism through which relatively has affected on the senescence process (Aran *et al.*, 2011).

These significant increase in vase life may be due to that salicylic acid (SA), a natural phenolic secondary metabolite, in various aspects of vital processes like ethylene biosynthesis, stomatal conductance, respiration, senescence and the activation of defense systems against different pathogens is well An and Mou (2011). SA treatments were effective to affect postharvest life of cut flowers probably via the declined bacterial growth, reduced vascular blockage, reduced transpiration, prevented ethylene formation and induced antioxidant system in treated cut flowers thereby delaying the senescence process confirmed by Danaee *et al.* (2013). These results are in accordance with those of Roodbaraky *et al.* (2012), Kazemi *et al.* (2012) on Carnation and Soleimany-Fard *et al.* (2013) on Alstroemeria flowers. They indicated that salicylic acid increased the vase life of cut flowers compared to control.

The improving effect of calcium chloride on vase life might be attributed to possibility of calcium to decrease the respiration rate, osmotic adjustment and stability of cell membrane and increase water flow through the stems by association with pectin in the xylem cell walls, also calcium play important role in increasing tissue resistance and delaying senility through preventing ethylene synthesis and it's processing as confirmed by Sardoei (2014). Similar results were obtained by Amiri *et al.* (2009) and Ibrahim *et al.* (2011) on Gerbera, Zadeh and Mirzakhani (2012) on Carnation, Cortes *et al.* (2011), Abdolmaleki *et al.* (2015) on Rose and Seyedi *et al.* (2013) on Liliium cut flowers, they proved that treatments with calcium chloride increased longevity of cut flowers to highest values and delayed senescence.

We could return the effect of aluminum sulphate on increasing vase life of cut flowers to the antimicrobial effect, increase water uptake, inhibit ethylene production and action and reduce transpiration rate as reported by seyf *et al.* (2012); Mohammadi *et al.* (2012); Jowkar *et al.* (2012) and Hashemabadi *et al.* (2015). These results in agreement with those of Jowkar *et al.* (2012); seyf *et al.* (2012), Basaki *et al.* (2013) and Nader *et al.* (2015) on Rose, Hashemabadi *et al.* (2015) on Carnation, Mohammadi *et al.* (2014) on Tuberose and Amiri *et al.* (2009) on Gerbera cut flowers. They showed that the effect of aluminum sulphate on life of flower was significant.

During senescence, marked changes occur in the biochemical and biophysical properties of the cell membranes. Ethylene plays a central role in the senescence of many cut flowers as proved by Reid (1989). Flower vase life is affected by respiration, carbohydrates deterioration, disease inoculation, water uptake etc. During vase life of cut flowers, ethylene synthesis plays a major role in senescence. Similarly carbohydrates and soluble sugars in the petals also help in quality retention of cut flowers for longer period proved by Hussien and Yassin (2013). There was a direct relationship between vase life and, increasing of relative fresh weight and water uptake. Obstruction of the xylem by bacteria, therefore, inability of water absorption by flower steams is one of the current problems that lead to decrease in flowers postharvest longevity and also early wilting of them as observed by Sardoei (2014). Various studies have found that bacterial contamination is one of the most important factors in reducing postharvest life of cut flowers with the negative impact on respiration, photosynthesis and water uptake, also with increasing the evaporation, caused water imbalance and indirectly stimulates ethylene production and shortens postharvest life of cut flowers found by Mohammadi *et al.* (2012). According to the scientific findings, the postharvest life of different ornamental cut flowers could be affected by the application of various chemicals as preservatives as reported by Danaee *et al.* (2013). Anti-ethylene and antimicrobial compounds due to their stem end bacterial contamination control, can stimulate ethylene production indirectly and can control ethylene production and extending vase life of cut flowers confirmed by Hashemabadi *et al.* (2015).

Table (7). Effect of some preservative solutions on proline (μg proline/g), number of bacteria (CFU, Colony Forming Unit) and vase life (days) of Gladiolus cut flowers "Rose Supreme" in the first and the second experiments (April and November, 2015).

Treatments	First experiment (April, 2015)			Second experiment (November, 2015)		
	Proline μg proline/g	Number of bacteria Log 7 CFU/ ml	Vase life (days)	Proline μg proline/g	Number of bacteria Log 7 CFU/ ml	Vase life (days)
Control	943.3a	20.5a	14.92d	426.66a	11a	17.5d
Glutamic acid 100 mg/l	708.5ab	6.33bcd	19.67bc	186.86e	7.67c	21.58b
Glutamic acid 200 mg/l	688.3ab	7.33bc	20.42b	264.72c	5d	22.25ab
Glutamic acid 300 mg/l	190.4cd	5cde	22.17a	54.50i	3e	23.17a
Salicylic acid 200 mg/l	349.3cd	6.33bcd	20.58b	147.93gh	8.33bc	19.42c
Salicylic acid 400 mg/l	209.0cd	3.67def	22.17a	367.49b	5.33d	22.67ab
Salicylic acid 600 mg/l	928.1a	4def	19.75bc	264.72c	6d	19.35c
Calcium chloride 1000mg/l	817.5a	8b	18.5c	362.82b	9.67ab	19.42c
Calcium chloride 2000mg/l	915.5a	7bc	18.83c	266.27c	8.67bc	20.08c
Calcium chloride 3000mg/l	301.5cd	4.67cde	19.42bc	152.60f	8c	20.25c
Aluminum sulphate 100mg/l	479.6bc	2.67ef	19.17bc	214.89d	9bc	19.75c
Aluminum sulphate 200mg/l	129.9d	2.33ef	19.92bc	130.80gh	7.67c	21.5b
Aluminum sulphate 300mg/l	360.2cd	1.33f	20.58b	115.23h	5.67d	22.17ab
LSD at 0.05	303.44	2.83	1.43	17.29	1.40	1.17

CONCLUSION

In conclusion, the present study demonstrates that all treatments showed significant effect on quality parameters and flowers longevity compared to control in Gladiolus experiments. Glutamic acid with last concentration (300 mg/l) and intermediate concentration of salicylic acid (400 mg/l) were more effective on increasing vase life compared to control than other treatments in both experiments, except for treatments of glutamic acid (200 mg/l) and aluminum sulphate (200 and 300 mg/l) in second experiment. Our findings support for wider testing and use of the natural, cheap, safe and biodegradable compounds.

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الملخص العربي

تأثير بعض محاليل الحفظ على عمر أزهار الجلاديولس المقطوفة

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تعتبر المحافظة على جودة وعمر الأزهار من العوامل الهامة لتقييم جودة زهور القطف في كل من الأسواق المحلية وأسواق التصدير. تهدف هذه الدراسة إلى تحديد فعالية بعض محاليل الحفظ مثل حمض الجلوتاميك (١٠٠ و ٢٠٠ و ٣٠٠ مجم/لتر) و حمض الصفصاف (٢٠٠ و ٤٠٠ و ٦٠٠ مجم/لتر) وكلوريد الكالسيوم (١٠٠٠ و ٢٠٠٠ و ٣٠٠٠ مجم/لتر) وكبريتات الألومنيوم (١٠٠ و ٢٠٠ و ٣٠٠ مجم/لتر) على معايير الجودة لأزهار الجلاديولس صنف "Rose Supreme". أظهرت النتائج أن كل المعاملات أدت إلى زيادة معنوية في عمر الأزهار والوزن الطازج و كمية الماء الممتص ومحتوى الكلوروفيل مع خفض عدد البكتيريا وتراكم البرولين مقارنة بالكنترول. تم الحصول على أعلى زيادة في الوزن الطازج وكمية الماء الممتص ومحتوى الكلوروفيل من قبل حمض الجلوتاميك (٣٠٠ مجم/لتر) وحمض الصفصاف (٤٠٠ مجم/لتر) وكلوريد الكالسيوم (٣٠٠٠ مجم/لتر) وكبريتات الألومنيوم (٣٠٠ مجم/لتر)، في حين أسفرت المعاملة بحمض الجلوتاميك (٣٠٠ مجم/لتر) وحمض الساليسك (٤٠٠ مجم/لتر) عن أطول مدة بقاء للأزهار في كلا التجريبتين، بالإضافة إلى حمض الجلوتاميك (٢٠٠ مجم/لتر) وكبريتات الألومنيوم (٢٠٠ و ٣٠٠ مجم/لتر) في التجربة الثانية. علاوة على ذلك، كان لحمض الجلوتاميك (٣٠٠ مجم/لتر) الأثر الأكبر على تقليل عدد البكتيريا وتراكم البرولين في كلا التجريبتين، بالإضافة إلى حمض الساليسك (٤٠٠ و ٦٠٠ مجم/لتر) وكلوريد الكالسيوم (٣٠٠٠ مجم/لتر) وكبريتات الألومنيوم (١٠٠ و ٢٠٠ و ٣٠٠ مجم/لتر) على عدد البكتيريا و حمض الساليسك (٢٠٠ و ٤٠٠ مجم/لتر) وكلوريد الكالسيوم (٣٠٠٠ مجم/لتر) وكبريتات الألومنيوم (١٠٠ و ٢٠٠ و ٣٠٠ مجم/لتر) على تراكم البرولين في التجربة الأولى مقارنة بالكنترول.