Improvement in vase life of cut rose cv. "Dolce Vita" by preharvest foliar application of calcium chloride and salicylic acid

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Abstract

Rosa hybrida L. is an important commercial cut flower. Salicylic acid (SA) and calcium chloride (CaCl₂) act as endogenous signal molecules responsible for growth parameters in plants. The aim of this study was to evaluate the effects of preharvest SA and CaCl₂ treatments in extending the vase life of cut rose flowers. Therefore, a factorial experiment based on completely randomized design with SA (0, 150, 300, and 450 mg L^{-1}) and $CaCl_2$ (0, 0.75, 1.5, and 2.25%) with 4 replicates and 2 samples (individual flowers) in each replicate, was conducted. Changes in growth, macronutrient concentration, chlorophyll content, leaf relative water content (LRWC), flower quality, vase life, and membrane stability index were investigated in R. hybrida cv. "Dolce Vita." Exogenously applied SA and CaCl₂ increased plant growth (such as shoots and flower buds). Foliar application of SA and CaCl₂ also increased macronutrient concentration (N, K, Ca, and Mg), chlorophyll content, LRWC, flower quality, and vase life; however membrane stability index was decreased with increasing levels of SA and CaCl₂. These results suggest that SA and CaCl₂ could be used as potential growth promoters to improve postharvest life of roses. According to the results of this experiment, SA and CaCl₂ as natural, cheap, safe, and biodegradable compounds are suitable alternatives for conventional chemical treatments in order to prolong vase life of cut rose flowers. Commercialization of these compounds for optimum formulations needs further experiments.

Keywords: chlorophyll content, membrane stability, plant growth, postharvest life.

Abbreviations: CaCl₂, calcium chloride; **cv.**, cultivar(s); **RH**, relative humidity; **SA**, salicylic acid; **LRWC**, leaf relative water content.

Introduction

Rose is ranked as the first economically important cut flower in the world (Van Doorn, 1997). Maintaining good quality of cut flowers and prolonging their vase life are considered important and practical for supplying acceptable products for the markets. Therefore, a large body of literature has been focused on this purpose (Redman *et al.*, 2002; Macnish *et al.*, 2008; Solgi *et al.*, 2009; Zencirkiran, 2010). Vase life of cut flowers is mainly affected by three main factors: 1. preharvest growth conditions, 2. the presence ethylene which accelerates the senescence of many flowers, and 3. microorganisms causing vascular blockage and thus reduce the vase life of cut flowers (Van Doorn, 1994; Zencirkiran, 2005; Zencirkiran, 2010). Worldwide, development of methods for increasing vase life and flower quality in rose plants is vital and

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receives considerable attention. Salicylic acid (SA) plays an important role in abiotic stress tolerance, and considerable interests have been focused on SA due to its ability to induce a protective effect on plants under stress. SA is qualified as a plant hormone due to its physiological and biological roles in plants (Raskin, 1992), and also, it has been suggested as a signal transducer or messenger under stress conditions (Klessig and Malamy, 1994). It has been shown that postharvest treatment of various cut flowers by SA could improve their vase life (Jalili Marandi et al., 2011; Kazemi et al., 2011; Mansouri, 2012); however probable effects of preharvest application of SA on vase life of cut roses have not yet been fully studied (Alaey et al., 2011). Plants sense stress by the synthesis of signaling molecules, such as calcium (Ca²⁺), jasmonic acid, ethylene, and SA, which activate a range of signal transduction pathways. The role of SA as a defense-inducing signal molecule has been well established in plants (Klessig and Malamy, 1994; Ganesan and Thomas, 2001).

Calcium plays various important roles such as in forming cross-bridges as an essential element, regulating growth and development in plants, as well as a being a constituent of the cell wall (Hepler, 2005). It affects cell wall integrity and is regarded as the last barrier before cell separation (Fry, 2004). Exogenous application of calcium stabilizes the plant cell wall and protects it from cell wall degrading enzymes (White and Broadley, 2003). In addition, calcium participates in crosslinking negative charges, especially on the carboxylic residues of pectin, imparting significant structural rigidity to the cell wall (Hepler, 2010); over 60% of calcium in plants is associated with pectin (Lara et al., 2004). Increase in various growth parameters has been reported after the application of SA (Gunes et al., 2005; Szepesi et al., 2005; Yildirim et al. 2008) and CaCl₂ (Poovaiah and Leopold,1973; Tan et al., 2011; Nasir Khan et al., 2012). However, the role of SA or CaCl₂ in

plant's growth is well studied, whereas information about their interaction on vase life via improvement of physiological parameters, especially their preharvest usage is rare. Therefore, the main objective of this study was to evaluate the interactive effects of SA and CaCl₂ on growth, chlorophyll content, mineral uptake, and also to examine changes in membrane stability, possible change of vase life, relative water content and flower quality of Vita" rose cultivar "Dolce under greenhouse hydroponic conditions.

Materials and Methods

Plant materials, treatments, and growth conditions

R. hybrida L. "Dolce Vita" plants were hydroponically grown in a perlite medium under commercial greenhouse conditions in Shiraz, Iran. The latitude and longitude Shiraz city in Fars province is of 29°36′54″N and 52°32′17″E and its altitude is about 1,600 m above sea level. Day and night temperatures of the greenhouse were 25±1 and 15±1°C, respectively, and RH was maintained at approximately 70%. Average midday photosynthetically active radiation was up to 960 μ mol m⁻² s⁻¹. The nutrient solution composition was (in kg. $1,000 \text{ L}^{-1}$ water): 42 KNO₃, 40 Ca (NO₃)₂·4H₂O, 4 NH₄H₂PO₄, 16 KH₂PO₄, 8 MgSO₄·7H₂O for macronutrients and (in g. $1,000 \text{ L}^{-1}$ water): 100 MnSO₄·7H₂O; 120 ZnSO₄·7H₂O; 2 CuSO₄·5H₂O, 2 NaMoO₄, 3 H₃BO₃ 120 NaFe-EDTA concentrations for micronutrients. The pH of the nutrient solution was adjusted to 5.2 to 5.7 and EC was 1.75 to 1.85 dS m^{-1} . The media were leached weekly prevent to ion accumulation. Topping in the "Dolce Vita" cultivar was performed (on December 1, 2012) from the upper section of the third bud. One week after topping, the first SA and CaCl₂ sprays were carried out on the plant leaves (approximately 500 mL per plant). The second and third foliar applications were conducted with 2 weeks intervals. Treatments included SA (Merck) at 0 (control), 150, 300, and 450 mg L⁻¹ and CaCl₂.2H₂O (Merck) at 0 (control), 0.75, 1.5, and 2.25% concentrations. Control plants were sprayed with distilled water. Tween-20 (2 ml L⁻¹) was added as surfactant to all solutions to improve absorption of chemicals. These sprays were applied in the morning hours (6 to 9 a.m.).

Measured parameters

Flowers were harvested 7 to 10 days after the last treatment date and transferred to the laboratory. As well, leaf samples were taken 1 day before harvest for the determination of macronutrient concentration. An index for the developmental stages of flower bud opening and senescence in rose flowers is described as follows: stage 0, unopened bud; stage I, partially opened bud and sepals ends started to separate from each other; stage II, completely opened bud and sepals ends completely separated from each other: stage III, partially opened flower with their outer petal whorl just unfurled (commercial stage); stage IV, fully opened flower with sepals completely opened; stage V, fully opened flower with anther appearance; stage VI, petals completely unfolded and starting to wilt; stage VII, flower completely senesced; stage VIII, dead flower. All flower stems were cut (at commercial stage) to 35 cm diagonally. Except three or four upper leaves of flower stem, all other leaves were removed. The flowers were put into vase solution (deionized water) and kept in a room with average day and night temperatures of 20±2°C and a RH of 70% under a daily light period of 12 h and 12 μ mol m⁻² s⁻¹ light intensity provided by cool white florescent lamps.

Leaf area

Leaf area was measured with a Delta-T Devices (Cambridge, UK) area measurement system.

Growth parameters

Plant growth parameters such as fresh and

dry weights and length of stem of shoot and flower bud were recorded one day before harvest. The plant materials were dried at 70 °C for 2 days to determine their dry weight.

Chlorophyll content

Chlorophylls a and b were determined according to the method of Arnon (1949). Fresh leaves (0.2 g) were cut and extracted overnight with 80% acetone at $0-4^{\circ}$ C. The extracts were centrifuged at $10,000 \times g$ for 5 min. Absorbance of the supernatant was read at 645, 663, and 480 nm using a spectrophotometer (Hitachi-220, Japan).

Nitrogen (N) content

The N content was measured by a Kjeldahl digestion method (Pooviah, 1986).

Potassium (K^{+}) content

The K content in leaves was determined in the digested leaf material using specific filters in the flame-photometer (Model, C150, AIMIL, India).

Calcium (Ca^{2+}) and magnesium (Mg^{2+}) content

Leaf samples were dried in an oven at 105°C for 15 min, and thereafter at 80°C up to a constant weight. Approximately 150 mg of dried leaves was burned to ashes in an oven at 550°C. The ashes were dissolved in 65–68% nitric acid (HNO₃) solution and diluted with 0.1 M HNO₃ to 20 mL. Ca²⁺ and Mg²⁺ were measured by atomic absorption spectrometry (Hitachi Z-8000, Hitachi Ltd., Tokyo, Japan).

Vase life

The period from the first day when cut flowers were placed in vase solutions until they lost their ornamental value, mainly defined by wilting, petal browning, and discoloration was considered as vase life and expressed in days.

Flower quality

The scoring system was used to determine the quality of the flowers as follows (Hassan, 2005): points 4: when the petals were completely fresh (no bending, no observable softening in the buds, not bent, and blackened petals). Points 3: start of neck bending, wilting, and staining of petal sup to 20%. Points 2: increase in neck bending and petals wilting up to 20–50%. Point 1: petals wilting up to 50 to 100% and complete neck bending. The following formula was used to express the quality point of flowers:

Flower quality (points) = \sum each replicate daily flower quality point up to the end of the vase life/replicate number.

Leaf relative water content (LRWC)

The samples were weighed immediately for their fresh weight (FW), then floated on distilled water for 4 h, then weighed to obtain turgid weight (TW). The leaf samples were dried in the oven at 70°C for 24 h and then their dry weight (DW) was obtained. The LRWC was calculated using the formula (Gonzalez and Gonzalez-Vilar, 2003),

 $LRWC = (FW-DW)/(TW-DW) \times 100$

Membrane stability index

On 15th day of experimental period, membrane stability of petals was determined by recording the electrical conductivity of leachates in double distilled water at 40 and 100°C (Ezhilmathi et al., 2007). Two similar leaf disks (0.1 g) were cut in uniform sizes and placed in test tubes containing 10 ml of double distilled water to separate. One disk was kept at 40 °C for 30 min and the other at 100 °C in boiling water bath for 15 min, and their respective electric conductivities, C1 and C2, were measured with a conductivity meter. Measurement of membrane stability was performed by the following formula:

Membrane stability index = $[1-(C1/C2)] \times 100$

Experimental design and statistical analysis

The treatments were conducted as factorial in a completely randomized design with four replications and two observations in each replication. The recorded data were analyzed (ANOVA) using SAS v.9.1 software. Means were compared using the least significant difference (LSD) test at $P \le 0.05$.

Results

Leaf area

Different concentrations of SA and $CaCl_2$ increased the leaf area of studied rose cultivar (Table 2). The maximum leaf area for "Dolce Vita" was 79.5 cm² when they were sprayed with SA at 150 mg L⁻¹ plus CaCl₂ at 0.75% concentration. Also, minimum of leaf area was 62.5 cm² when they were sprayed with SA at 450 mg L⁻¹ plus CaCl₂ at 2.25% concentration (Table 1).

Growth parameters

DW and FW of shoot and flower bud and length of stem were significantly increased along with increase in SA and CaCl₂ concentrations (Table 1). Highest length of stem (82 cm) of "Dolce Vita" cultivar was obtained when plants were treated with SA at 150 mg L^{-1} plus CaCl₂at 0.75% concentration (Table 1). The lowest length of stem (53.87 cm) was observed in plants treated with 2.25% CaCl₂. Highest FWs of shoot and flower bud (61.25 g) of "Dolce Vita" cultivar was obtained when plants were treated with SA at 150 mg L⁻¹plus CaCl₂ at 0.75% concentration (Table 1). The lowest FWs of shoot and flower bud (24.7 g) were observed in plants treated with 2.25% CaCl₂. The highest DWs of shoot and flower bud (22.18 g) were obtained from plants treated with SA (450 mg L^{-1}) plus CaCl₂ (at 0.75%). Minimum DW of shoot and flower bud (6.15 g) of cultivar belonged to the control plants.

SA	CaCl ₂	Chlorophyll	Leaf area per	Shoot and	Shoot and flower	Stem length
$(mg.l^{-1})$	(%)	content	stem	flower bud	bud dry weights	(cm)
	()	$(mg g^{-1} FW)$	(cm ²)	fresh weights (g)	(g)	
	0	0.69 g	77.50 с-е	33.57 f-h	6.15 d	57.50 gh
0	0.75	0.77 f	71.75 d-f	36.40 fg	10 b-d	62.70 d-h
(control)	1.5	0.81 ef	76.50 с-е	45.25 с-е	11.13 a-d	59.50 f-h
	2.25	0.80 ef	68 ef	24.7 i	6.28 d	53.87 h
150	0	0.90 b	80.25 cd	55 ab	12.43 a-d	78.50 ab
	0.75	0.96 a	97.50 a	61.25 a	14.36 a-d	82 a
	1.5	0.89 b	88.75 ab	46.21 cd	19.67 a-c	71.25 b-e
	2.25	0.79 ef	76 с-е	38.80 ef	16.97 a-d	61.50 e-h
300	0	0.82 с-е	97 a	27.95 hi	10.47 a-d	74.25 a-c
	0.75	0.88 b	82.75 b-d	40.17 d-f	11.17 a-d	74.50 a-c
	1.5	0.87 bc	71.75 d-f	35.82 fg	20.25 ab	72.75 a-d
	2.25	0.82 de	75.50 с-е	38.37 ef	10.40 b-c	75 а-с
450	0	0.819 e	76.25 с-е	33.37 f-h	8.38 cd	66 c-g
	0.75	0.813 ef	83.25 b-c	51.06 bc	22.18 a	68.70 b-f
	1.5	0.86 b-d	73 c-f	39 ef	10.12 b-d	63.50 d-h
	2.25	0.80 ef	62.50 f	30.07 g-i	9.47 b-d	61.12 e-h
LSD ≤ 0.05		0.04	11.13	7.03	2.74	10.71

Table 1. Effects of preharvest spray of SA and CaCl₂ treatments on chlorophyll content, leaf area, shoot and flower bud fresh and dry weights, and stem length of cut rose flowers cv. 'Dolce Vita'

Means followed by the same letters in each column are not significantly different at 5% level of probability using LSD test.

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SA mg L^{-1}	CaCl ₂ %	N (%)	K (%)	Ca (%)	Mg (%)			
	0	1.71 j	3.13 с-е	1.05 h	0.18 h			
	0.75	2.63 d-g	3.01 de	1.85 e-g	0.21 fg			
0 (control)	1.5	2.61 e-g	3.31 a-d	2.16 b-e	0.25 de			
	2.25	2.55 gh	3.19 b-e	2.34 а-с	0.22 fg			
	0	2.84 bc	3 28 a-d	2 44 a-c	0.22 fg			
	0.75	3.02 a	3.62 a	2.55 ab	0.22 rg			
150	1.5	2.25 h	3.32 a-d	2.57 ab	0.29 bc			
	2.25	2.52 gh	3.16 с-е	2.48 ab	0.22 fg			
	0	2.84 bc	3.53 ab	1.60 g	0.237 ef			
	0.75	2.73 с-е	3.58 a	1.87 d-g	0.30 ab			
300	1.5	2.86 b	3.14 c-e	2.26 a-d	0.32 a			
	2.25	2.75 b-d	2.91 e	2.51 ab	0.21 fg			
	0	2.53 gh	3 39 a-c	1 68 fg	0.23 f			
	0.75	2.44 h	3.43 a-c	2.05 c-f	0.21 fg			
450	1.5	2.54 gh	3.20 h-e	2.26 a-e	0.20 gh			
	2.25	2.61 fg	3.11 c-e	2.62 a	0.21 fg			
LSD ≤ 0.05		0.12	0.35	0.4	0.02			

Table 2. Effects of preharvest spray of SA and CaCl₂ treatments on mineral content in leaves of cut rose flowers cv. 'Dolce Vita'

Means followed by the same letters in each column are not significantly different at 5% level of probability using LSD test.

Chlorophyll content

The chlorophyll content in leaves increased following exogenous SA and CaCl₂ application in "Dolce Vita" cultivar. The highest chlorophyll content (0.96 mg g⁻¹ FW) was obtained when plants were treated with SA at 150 mg L⁻¹ and CaCl₂ at 0.75% concentration. The lowest chlorophyll content (0.69 mg g⁻¹ FW) was observed in control samples (Table 1).

Macronutrient concentration

Table 2 presents changes of macronutrient concentration following SA and CaCl₂ application. Lowest of N content in leaves of "Dolce Vita" cultivar was related to control samples and highest was obtained in plants sprayed with SA at150 mg L^{-1} SA and CaCl₂ at 0.75% concentration. Highest K content in the leaves was obtained when the roses were treated with SA at 150 mg L^{-1} and CaCl₂ at 0.75% concentration. The lowest K content was obtained in plants treated with 300 mg L^{-1} SA with 2.25% CaCl₂. Along with increasing in CaCl₂ concentration, Κ content in leaves decreased. Ca content in leaves increased progressively along with increase in exogenous CaCl₂ level. The results showed that the highest Ca content in the leaves was obtained when plants were treated with SA at 450 mg L^{-1} and CaCl₂at 2.25% concentration. The lowest content of Ca in leaves was observed in control samples. Highest Mg content in the leaves was obtained from plants treated with 300 mg L^{-1} SA and CaCl₂ at 0.75%, and the lowest Mg content was observed in control leaves.

Vase life

Figure 1 presents changes in vase life following SA and $CaCl_2$ application. Different concentrations of SA and $CaCl_2$ extended the vase life of "Dolce Vita" rose. The longest vase life was 29.25 days when they were sprayed with SA at150 mg L⁻¹ SA plus CaCl₂ at 0.75% concentration. Minimum vase life belonged to the control plants.

Flower quality

The highest flower quality was observed in plants treated with 150 mg L⁻¹ SA and 0.75% CaCl₂ solutions, and lowest flower quality was obtained in control samples (Fig. 2).

LRWC

Figure 3 presents changes of LRWC following SA and CaCl₂ application. Both SA and CaCl₂ caused a significant increase in LRWC of rose cultivar under the study. The highest LRWC for "Dolce Vita" was 79.17% when they were sprayed with SA at 150 mg L^{-1} SA plus CaCl₂ at 0.75% concentration (Fig. 3). Minimum of LRWC belonged to the control plants.

Membrane stability

Maximum of electrolyte leakage in "Dolce Vita" cultivar was related to control samples and minimum was obtained in plants sprayed with SA at 150 mg L^{-1} SA and CaCl₂ at 0.75% concentration (Fig. 4).



Salicylic acid and Calcium chloride treatments

Fig. 1. Effects of preharvest spray of salicylic acid and calcium chloride on vase life of cut rose flowers cv. "Dolce Vita"



Salicylic acid and Calcium chloride treatments

Fig. 2. Effects of preharvest spray of salicylic acid and calcium chloride on flower quality of cut rose flowers cv. "Dolce Vita"



Salicylic acid and Calcium chloride treatments

Fig. 3. Effects of preharvest spray of salicylic acid and calcium chloride on leaf relative water content of cut rose flowers cv. "Dolce Vita"



Salicylic acid and Calcium chloride treatments

Fig. 4. Effects of preharvest spray of salicylic acid and calcium chloride on electrolyte leakage of cut rose flowers cv. "Dolce Vita"

Discussion

Plants treated with SA and CaCl₂ had higher growth parameters than untreated plants. FW and DW of shoot and flower bud and length of stem of "Dolce Vita" cultivar was found to be significantly increased along with augmentation of the SA and CaCl₂ concentrations. These results were in agreement with the findings of Baas et al. (2000) and Abadi (2010) on rose, Karlidag et al. (2009) on strawberry, and Yildirim et al. (2008) on cucumber who showed that SA and Ca²⁺ treatments ameliorated the beneficial effects of hydroponic conditions on FW and DW of plants. Furthermore, it has been reported that SA applications increased carbon dioxide (CO_2) assimilation and photosynthetic rate led to a higher DW (Fariduddin et al., 2003; Khan et al., 2003; Szepsi et al., 2005). This increase in dry matter content might also be attributed to the increased mineral uptake by SA and CaCl₂ treatments. Similar results were reported by Hayat (2012), Nedjimi and Daoud (2009), and Feng et al. (2010) who indicated the beneficial influences of foliar application of SA and CaCl₂ on growth parameters. Singh and Usha (2003) and Pirasteh-Anosheh et al. (2012) reported that SA-treated wheat had higher DW compared to untreated seedlings under water stress.

In this study, SA and CaCl₂-treated plants had higher chlorophyll content in comparison to untreated plants. These results were in accordance with findings of El-Tayeb (2005) and Gunes et al. (2006). They indicated that SA treatment caused increased chlorophyll content of leaves of barley and maize. Khodary (2004) reported increased chlorophylls a and b content following SA application. The results of this investigation were also in agreement with findings of Ghai et al. (2002) on rutabaga, Moharekar et al. (2003), and Pirasteh-Anosheh et al. (2012) on wheat and Yildirim et al. (2006) on cucumber. Furthermore, Shi et al. (2006) indicated

that foliar application of SA at 1.00 mM concentration increased chlorophyll content of the leaves of cucumber seedlings grown under heat stress. Ethylene can induce leaf chlorosis through an enzymatic process that is associated with reduction in the chlorophyll content; however, it has been reported that SA can alleviate ethylene-induced leaf chlorosis by blockage in ethylene biosynthesis (Jalili Marandi et al., 2011). Leaf senescence can be delayed by exogenous application of calcium due to improved maintenance of chlorophyll and proteins content. Moreover, the application of calcium leads to delay in leaf senescence, maintenance of cell integrity, reduction in free spaces of leaf tissues, and encourages water saving that all occur during senescence (Poovaiah and Leopold, 1973). Increase in chlorophyll content after CaCl₂ and SA foliar application have been reported by various authors previously (Yildirim et al., 2008; Yildirim et al., 2009; Feng et al., 2010; Tan et al., 2011; Hayat, 2012; Nasir Khan et al., 2012; Agami and Mohamed, 2013).

In agreement with these results, Yildirim et al. (2008) reported that mineral content of cucumber was improved by SA application. Similar results have also been reported in maize (Gunes et al., 2005) and tomato (Szepesi et al., 2005). Under proper conditions of pH and concentration, SA significantly could affect mineral absorption (Harper and Balke, 1981). Glass (1974) reported that exogenously applied SA on barley and oat roots inhibited potassium (K) absorption in a pH-and concentration-dependent manner.

Among all treatments, 150 mg L⁻¹ SA solution combined with CaCl₂ (0.75%) was the best in prolonging the vase life of studied rose cultivar. Similar results have been reported in *R. hybrida* L. cultivar "Black Magic" (Alaey *et al.*, 2011) and gladiolus (Ezhilmathi *et al.*, 2007). This treatment enhanced the concentration of calcium, potassium, magnesium, nitrogen

of the leaves, LRWC, chlorophyll content, DW and FW of shoot and flower bud , length of stem, flower quality, and reduced electrolyte leakage; and simultaneously increased sugar, starch, protein, and antioxidant enzymes activities such as peroxidase, and superoxide catalase. dismutase (data not shown). In agreement with our results Tari et al. (2002) and Szepsi et al. (2005) found that exogenous SA treatments increased LRWC of tomato plants. This can be due to increase in leaf diffusive resistance induced by SA and reduced transpiration. Our results were also in accordance with findings of Mortazavi et al. (2007) on rose, Nasir Khan et al. (2012) on Brassica juncea cv. Varuna, and Nedjimi and Daoud (2009) on atriplex that showed a consistently higher LRWC with CaCl₂ application. One of the major factors in the maintenance of membrane integrity and ion-transport regulation is calcium (Cramer et al., 1986). Calcium plays a crucial role in plant membrane stability, cell wall stabilization, and cell integrity (Hirschi, 2004); SA also facilitated the maintenance of membrane functions (El-Tayeb, 2005; Gunes et al., 2006). The senescence process is associated with increasing the membrane leakage. In agreement with our results, Kazemi et al. (2011) reported that membrane permeability was improved by SA. Similar effects on membrane stability were reported when SA was applied on chrvsanthemum (Mansouri. 2012). Reduced value of physiological characteristics in plants treated with SA up to150 mg L^{-1} and 0.75% CaCl₂ might be phytotoxicity due to the (growthinhibiting) effects at higher concentrations of these compounds. Additional research is needed to determine the exact biochemical cause of this effect. Previously, War et al. (2011) found that chickpea plants treated with SA at 2 mM concentration showed symptoms of phytotoxicity. Accordingly, they suggested that SA at 1.5 mM concentration was safe for these plants and

could be utilized as a plant defense inducer. Also, Kovacik et al. (2009) found that Matricaria chamomilla plants treated with SA at 250 µM concentration showed phytotoxicity (growth-inhibiting) and decrease in chlorophylls, water content, and soluble proteins. In terms of phenolic metabolism, it seems that the higher SA had a toxic effect. In conclusion, while SA has been reported to be the inducer of various positive physiological and biochemical effects in plants, it has a negative effect on the growth at a higher concentration as well. It has been reported that the effects of the plant growth substances are dose dependent. It is therefore important to find out the safe concentration of SA. However, due to the application of calcium fertilizer in hydroponic system, we assumed that with increasing concentrations of CaCl₂, the endogenous content of calcium in plant was elevated and led to plant phytotoxicity (growth-inhibiting). Thus, lower concentrations of CaCl₂ can be suggested supplemental application which for supplies enough calcium during growth and development of this cultivar.

Conclusion

Treatment with SA and CaCl₂ extends the postharvest life of rose flowers at relatively low SA and CaCl₂ concentrations required to generate maximum effectiveness. This effect may be exerted by improving the membrane stability and different morphological and physiological attributes. The use of concentrations of 150 mg L^{-1} SA and 0.75% CaCl₂ at preharvest stage (growing and developing stages) could be recommended to improve postharvest vase life of roses and possibly other ornamental crops. In terms of overall performance, SA was the most effective followed by CaCl₂. According to the results of this experiment, we can generally say that SA and CaCl₂ as natural, cheap, safe, and biodegradable compounds can be suitable alternative chemical treatments in order to prolong vase life of cut flowers which is a fact that would be much appreciated by the growers and handlers of cut flowers. Commercialization of these compounds for optimum formulations needs further research and may be recommended for each particular species or cultivars.

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