Effects of pre- and postharvest applications of salicylic acid on the vase life of cut Alstroemeria flowers (Alstroemeria hybrida)

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ABSTRACT

Purpose: In this study, the effects of different concentrations of salicylic acid on the physiological traits of Alstroemeria (Alstroemeria hybrida cv. ‘Summer Sky’) cut flowers during pre-harvest and postharvest applications are investigated. The aim of this study was to identify the best treatments and methods affecting the vase life of cut Alstroemeria flowers.

Research Method: The study was performed as a factorial experiment based on a completely randomized design using three replications in each combination/mixed treatment. The factors comprised of the pre-harvest foliar and post-harvest (pulsing 24h and continuous) applications of salicylic acid at two concentrations of 30, 200 ppm (SA1) and 50, 300 ppm (SA2), respectively.

Findings: The vase life, chlorophylls a, b, and total, relative fresh weight, SOD activity, and the percentages of dry matter and the absorption of preservative solution were evaluated. The results showed that using high concentrations of salicylic acid (SA2) significantly increased the vase life of Alstroemeria. The pre-harvest foliar application of a 300 ppm of salicylic acid would be useful in prolonging the vase life of cut Alstroemeria flowers and thus would help to reduce the losses caused by the rapid senescence of petals which will lead to an increase in the quality of appearance and economic value.

Research limitations: There was no significant limitation to be reported.

Originality/Value: The positive impact of salicylic acid (50 ppm) in the pre-harvest method (B) was found, and its application improved the vase life of cut Alstroemeria flowers, which should be taken into consideration for commercial uses.
INTRODUCTION

Alstroemeria (*Alstroemeria hybrida*), as an important and popular herbaceous cut flower, belongs to Alstroemeriaeae family. Recently, the popularity of Alstroemeria plants has increased in many countries, including Canada, Japan, UK and USA. This popularity is due to the fact that alstroemeria flowers have beautiful colors and long-vase life (Park et al., 2010). As with other cut flowers, the abscission and senescence of petals are the two major constraints that reduce the ornamental value of cut alstroemeria flowers (Reid, 2004). The presence of sugar in the vase solution effectively delays petal wilting and abscission and prolongs the longevity of many cut flowers. The exogenous sugars provide substrates for respiration and structural support and improve water balance in cut flowers (Yeat et al., 2012). Factors affecting the senescence of petals of cut flowers can be categorized into two groups depending on the involvement of ethylene (Shibuya, 2018). Taking into account this classification, the cut alstroemeria flower falls in the ethylene-sensitive group (Chanasut et al., 2003). As reported by Galati et al. (2017), the average post-harvest life of cut alstroemeria flower is around 8 days.

This study examines the effects of different concentrations of pre-harvest and postharvest salicylic acid on the vase life and life quality of alstroemeria cut flowers. Salicylic acid is a simple phenolic compound with different properties. Nowadays, it is known as an effective herbal hormone in regulating growth and developmental processes in plants (Raskin, 1992). It prevents the degradation of chlorophyll via inhibiting ACC oxidase activity, suppressing ethylene production, and regulating ABA hormone function (Zamani et al., 2011). Kazemi and Ameri (2012) stated that adding 1.5 mM salicylic acid to the gerbera cut flower vase solution increased the absorption rate through reducing bacteria population in the solution. Jalili Marandi et al. (2011) reported that a concentration of 5.1 mM of salicylic acid increased the vase life of gladiolus cut flower from 18 to 21 days in the control group.

Salicylic acid could be considered as an endogenous plant hormone involved in the regulation of plant growth, development, and disease resistance mechanisms (Hayat et al., 2010; Luo et al., 2011). The study aims to identify the best treatments and methods influencing the vase life of cut Alstroemeria flowers.

MATERIALS AND METHODS

This experiment was carried out on *Alstroemeria hybrida* in April 2017 ‘Summer Sky’ cut flowers were obtained from local commercial greenhouse Mahallat is located in Markazi Province in Iran (longitude 50°49’ E and latitude 33°87’ N) and is 1747 m above sea level. Plants were grown under standard greenhouse conditions at 22°C and 16°C day and night temperatures, respectively. The soil used was composed of 70% clay, 25% sand and 5% manure. Then, with the appearance of the first flower bud on the highest branch, the plants were exposed to a number of two different treatments in a pre-harvest way. In the pre-harvest method, by the appearance of the first flower bud on the highest branch, Salicylic acid solutions were sprayed on alstroemeria plants at the rate of 500 ml per plant once a week. About three weeks later, the cut flowers along with all plants were harvested in the early morning and immediately transported to the laboratory of the horticulture department with an appropriate cover (in plastic packages). After removing the leaves at the end of their stem, all flowers were weighted and then uniformly recut with a height of 50 cm and put in the prepared solutions. An aqueous solution of SA (200 and 300 ppm) and distilled water were sprayed to run-off (approximately 500 mL per plant) about three weeks before flowers harvest. Next, both treated (sprayed) and untreated cut flowers were harvested. The cut
flowers treated pre-harvest (B) were kept in vase solution containing sucrose 3% with distilled water. Vase solution concentrations (pulsing 24h: A1) were distilled water with 20% sucrose. Vase solution concentrations (continuous: A2) were SA (30 and 50 ppm) and distilled water with 3% sucrose. Vase life was defined as the time/interval between the start of the treatment and the senescence of flowers. The vase life of cut flowers was evaluated in a 12h photoperiod, the light intensity of 12 μmol m⁻² s⁻¹, the relative humidity of 60 to 70%, and the temperature of 20 ± 2°C. The treatments used in this study are shown in Table 1. In the post-harvest pulse method, the harvested cut flowers were soaked in treatments for 24 hours and then transferred to a 3% sucrose solution until the end of the experiment. Finally, in the post-harvest method, the harvested cut flowers were permanently placed in treatment solutions until the end of the experiment. This study was performed in a completely randomized factorial design using three replications. The data were analyzed using SAS software and treatment means were compared through the LSD test.

**Vase Life**

Alstroemeria flowers longevity was considered as long as 50 percent of the loss flowers. To this end, flowers were daily checked and recorded (Ferrante et al., 2002).

**Destruction of the chlorophyll a, b and total**

To measure the amount of carotenoids in petals, Mazumdar and Majumdar’s (2003) method was used. To measure a, b, and total chlorophyll content of the plants, sampling was performed on the first and last days of the test. The process of measuring chlorophyll was performed using and finally, the leaf chlorophyll content was suggested in mg g⁻¹ of fresh weight. The difference between the amount of chlorophyll of the first and last days represents the degradation of chlorophyll, which is calculated using the following formula (1):

\[
\text{Chlorophyll a} = 9.93 (A_{660}) - 0.777 (A_{642.5})
\]

\[
\text{Chlorophyll b} = 17.6 (A_{642.5}) - 2.81 (A_{660})
\]

Total chlorophyll = 7.12 (A_{660}) - 16.8 (A_{642.5})

Degradation of chlorophyll a, b, total = chlorophyll a, b, and total (first day) - chlorophyll a, b, and total (last day) \hspace{1cm} (1)

**Percentage of the dry matter**

Cut alstroemeria flowers were weighed and placed in the oven at 60°C for 72h and were re-weighed via a digital scale, and the percentage of the dry matter was calculated using the following formula (2):

\[
\text{Percentage of dry matter} = \left( \frac{\text{Fresh weight}}{\text{Dry weight}} \right) \times 100
\]  \hspace{1cm} (2)

<table>
<thead>
<tr>
<th>Treatments</th>
<th>Level</th>
<th>Pre-harvest</th>
<th>Post-harvest, Pulse</th>
<th>Post-harvest, continuous</th>
</tr>
</thead>
<tbody>
<tr>
<td>Salicylic acid</td>
<td>SA1</td>
<td>200 distilled water +3%</td>
<td>200 distilled water +20% sucrose</td>
<td>30 distilled water +3% sucrose</td>
</tr>
<tr>
<td></td>
<td>SA2</td>
<td>300 distilled water +3%</td>
<td>300 distilled water +20% sucrose</td>
<td>50 distilled water +3% sucrose</td>
</tr>
<tr>
<td>Control</td>
<td>DW</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Table 1. Treatments and their application methods studied on vase life and post-harvest physiological characteristics of cut alstroemeria flower.

Numbers inside the table represent the concentration corresponding to each method-treatment level (ppm).
Superoxide dismutase enzyme (SOD) activity
Superoxide dismutase activity in the petals of cut Alstroemeria flowers was measured 1, 4, and 12 days after the harvest based on the method proposed by Giannopolitis and Ries (1977). The reaction solution (1 mL) contained 50 mM phosphate buffer (pH= 7), 12 mM riboflavin, 13 mM methionine, 0.1 mM EDTA, 7 mM nitro blue tetrazolium (NBT), and 10 μl of extracted enzyme solution. A solution with no enzyme was used as the control. Test tubes were irradiated under fluorescent lights at 100 mmol m^{-2}s^{-1} for 20 min. The absorbance of each solution was measured at 560 nm using a spectrophotometer. One unit of enzyme activity was defined as the amount of enzyme that would inhibit 50% of NBT photo reduction. The results are expressed in μmol g^{-1} FW.

Relative fresh weight (RFW)
The stems of Alstroemeria cut flowers were initially weighed at the beginning of the experiment. The fresh weight of cut flower was measured on the 1st, 4th, 8th, 12th, and last days using a digital scale.

\[ RFW = \frac{FW_t}{FW_{t0}} \times 100 \]
\[ RFW = \text{Relative Fresh Weight (\%)} \]
\[ FW_t = \text{Fresh Weight on days 1, 4, 8, 12} \]
\[ FW_{t0} = \text{Fresh Weight at day 0} \]

Absorption of the vase solution
The solution absorption was calculated through obtaining the weight difference of the vase solution containing preservative solution (without flower), subtracting the evaporation rate, and dividing it by the initial weight of the flowers (3). Furthermore, it was expressed as the solution absorption in mLg^{-1} FW.

\[ S_t = \text{Weight of the solution inside the vase on days 1, 4, 8, 12 and the last day} \]
\[ S_{t-1} = \text{Weight of the solution inside the vase in the previous day} \]

\[ \text{absorption of the solution (mL day}^{-1} \text{ g}^{-1} \text{ FW} ) (S_t - S_{t-1}) / W_{t0} \] (3)

RESULTS AND DISCUSSION

Vase Life
The effect of the interaction among the treatments (T) in different methods (M) on the vase life of Alstroemeria was significant at 5% level. The maximum vase life of Alstroemeria was related to the interaction of Salicylic acid treatment with a concentration of 300 mg/l (SA2) as well as the pre-harvest methods (B) with an average of 17.66 days. Furthermore, the minimum vase life of Alstroemeria was related to the interaction of the DW treatment (distilled water and 20% sucrose) and the pulse method (A1) with an average of 11.33 days (Fig. 1). In another study, it was found that a concentration of 1.5 mM salicylic acid increased the vase life of cut gladiolus flower (Jalili Marandi et al., 2011).
Numerous authors have found that the application of salicylic acid at different concentrations extended the vase life in cut flowers of rose (Zamani et al., 2011), gerbera and lily (Kazemi and Ameri, 2012), and chrysanthemum (Vahdati et al., 2012).

Destruction of the chlorophyll a, b and total
Variance analysis of the data shows that the amount of chlorophyll a, b, and the total is statistically significant at 1% level of likelihood. As shown in Figures 2, 3, and 4, the pre-
harvest application of salicylic acid treatment (300 ppm) is found to be more effective in decreasing chlorophyll degradation. Among the studied treatments, salicylic acid (SA2) and control (DW) treatments had the highest and lowest chlorophyll contents, respectively.

The decrease in chlorophyll is considered to be a symptom of oxidative stress condition, which might be due to the generation of reactive oxygen species (ROS) after infection, causing damage to chlorophyll a and the plant’s failure to capture the light. Thus, in such a case, photosynthesis decreases or stops (Ali & Al-quarainy, 2006). This reduction may be due to chlorophyll degradation, reduced chlorophyll synthesis, and the stability of thylakoid membrane. Besides, it may be associated with the increased activity of chlorophyllase enzyme (El-Shanhouey et al., 2014). It seems that the above anti-ethylene treatments cause the preservation of chlorophyll through inhibiting the activity of chlorophyllase enzyme and developing chloroplasts. Leaf yellowing is a common phenomenon in many sensitive species such as alstroemeria, lilium, chrysanthemum, and stock. In many species, leaf yellowing may be caused via placing plants in a dark place for a long time during storage or transport (Ferrante et al., 2002; Reid, 2004).

![Fig. 1](image1.png)

**Fig. 1.** The effects of different treatments and methods on the vase life *Alstroemeria hybrida*
Method: (A1: Pulsing 24 h, A2: Continuous, B: Pre-harvested)
Treatment: (SA1: Salicylic acid 200, 30, 200 ppm, SA2: Salicylic acid 300, 50, 300 ppm, DW: Distilled water)

![Fig. 2](image2.png)

**Fig. 2.** The effects of different treatments and methods on the degradation of chlorophyll a, *Alstroemeria hybrida*
Method: (A1: Pulsing 24 h, A2: Continuous, B: Pre-harvested)
Treatment: (SA1: Salicylic acid 200, 30, 200 ppm, SA2: Salicylic acid 300, 50, 300 ppm, DW: Distilled water)
showed that SA reduces the oxidative damage through maintaining superoxide dismutase (SOD) via ROS in plants and seeds and inhibit the reductions in plant quality. Previous studies showed that sucrose in cucumber enhanced leaf’s area, the total number of leaves, and plant’s height and dry matter. The analysis of variance results showed that the effect of the interaction treatments (T) in different methods (M) on the percentage of dry matter of alstroemeria was not significant. The maximum percentage of the dry matter was related to the salicylic acid treatment with a concentration of 300 mg/l (SA2), 3% sucrose, and a pre-harvest spraying method (B) with an average of 23.73%. On the other hand, the minimum percentage was related to the DW treatment (distilled water and sucrose 20%) and the pulse method (A1) with an average of 10.50%. Comparing the mean percentage of the dry matter in different methods showed the positive effects of the used treatments compared to that of the control treatment. Hence, the use of treatments at different concentrations increased the content of the dry matter compared to the one in control (Fig. 5).

Orabi et al. (2010) and Khodary (2004) reported that foliar application of salicylic acid in cucumber enhanced leaf’s area, the total number of leaves, and plant’s height and dry matter.

**Fig. 3.** The effects of different treatments and methods on the degradation of Chlorophyll b, *Alstroemeria hybrida*

Method: (A1: Pulsing 24 h, A2: Continuous, B: Pre-harvested)
Treatment: (SA1: Salicylic acid 200, 30, 200 ppm, SA2: Salicylic acid 300, 50, 300 ppm, DW: Distilled water)

**Fig. 4.** Effect of different treatments and methods on the degradation of total Chlorophyll of *Alstroemeria hybrida*

Method: (A1: Pulsing 24 h, A2: Continuous, B: Pre-harvested)
Treatment: (SA1: Salicylic acid 200, 30, 200 ppm, SA2: Salicylic acid 300, 50, 300 ppm, DW: Distilled water)

**Percentage of the dry matter**

The analysis of variance results showed that the effect of the interaction treatments (T) in different methods (M) on the percentage of dry matter of alstroemeria was not significant. The maximum percentage of the dry matter was related to the salicylic acid treatment with a concentration of 300 mg/l (SA2), 3% sucrose, and a pre-harvest spraying method (B) with an average of 23.73%. On the other hand, the minimum percentage was related to the DW treatment (distilled water and sucrose 20%) and the pulse method (A1) with an average of 10.50%. Comparing the mean percentage of the dry matter in different methods showed the positive effects of the used treatments compared to that of the control treatment. Hence, the use of treatments at different concentrations increased the content of the dry matter compared to the one in control (Fig. 5).

Orabi et al. (2010) and Khodary (2004) reported that foliar application of salicylic acid in cucumber enhanced leaf’s area, the total number of leaves, and plant’s height and dry matter.
**Fig. 5.** The effects of different treatments and methods on dry matter percentage *Alstroemeria hybrida* 
Method: (A1: Pulsing 24 h, A2: Continuous, B: Pre-harvested) 
Treatment: (SA1: Salicylic acid 200, 30, 200 ppm, SA2: Salicylic acid 300, 50, 300 ppm, DW: Distilled water)

**Fig. 6.** The effects of different treatments on SOD of *Alstroemeria hybrida* during vase period in different methods. Method: (A1: Pulsing 24 h, A2: Continuous, B: Pre-harvested) 
Treatment: (SA1: Salicylic acid 200, 30, 200 ppm, SA2: Salicylic acid 300, 50, 300 ppm, DW: Distilled water)

**Superoxide dismutase enzyme (SOD) activity**

The ANOVA test showed that interaction treatments (T) in the methods (M) of the SOD enzyme were significant on cut flowers on different days. The results showed that SOD activity significantly decreased during the experiment. Among all the treatments of salicylic acid, SA2 had the highest SOD activity, while the lowest activity of SOD enzyme was related to the control treatment (DW) in all methods. Overall, the highest amount of SOD enzyme was found as the result of implementing the pre-harvest salicylic acid treatment (300 ppm) with 3% sucrose on the 12th day and an average of 91.8 µ mol/g FW. In contrast, the lowest amount of SOD activity was related to the application of pulsing (distilled water with 20% sucrose) with an average of 22.68 µ mol/g FW on the 12th day (Fig. 6). Superoxide dismutase and peroxidase are antioxidant enzymes. These enzymes interfere during lipid peroxidation via ROS in plants and seeds and inhibit the reductions in plant quality. Previous studies showed that SA reduces the oxidative damage through maintaining superoxide dismutase activity to remove O₂ (Rao et al., 1997).
Relative Fresh Weight
The obtained data shows that the Relative Fresh Weight (RFW) of cut Alstroemeria flowers decreased during the vase period. Additionally, it was found that all treatments significantly increased the RFW during the first 8 days of the experiment in all methods. The RFW for some treatments significantly decreased from the 8th day until the end of the experiment (Fig. 7).

These products commonly take water and other materials from the mother plant; however, when they are cut off, they rapidly move into senescence and death as the result of water loss and weight reduction. This reduction is much higher in stressed conditions. SA can modulate plant responses to a wide range of oxidative stresses and prevent cell wall degradation (Shirasu et al., 1997).

Fig. 7. The effects of different treatments on relative fresh weight of *Alstroemeria hybrida* during vase period in different methods. Method: (A1: Pulsing 24 h, A2: Continuous, B: Pre-harvested) Treatment: (SA1: Salicylic acid 200, 30, 200 ppm, SA2: Salicylic acid 300, 50, 300 ppm, DW: Distilled water)

Fig. 8. The effects of different treatments on the absorption of the preservative solution of *Alstroemeria hybrida* during vase period in different methods. Method: (A1: Pulsing 24 h, A2: Continuous, B: Pre-harvested) Treatment: (SA1: Salicylic acid 200, 30, 200 ppm, SA2: Salicylic acid 300, 50, 300 ppm, DW: Distilled water)
Absorption of preservative solution
The effects the interaction among treatments (T) in different methods (M) on the absorption of preservative solution was significantly different at 5% level. In general, through comparing Figure 8, it can be deduced that the maximum absorption of the preservative solution was related to the salicylic acid treatment with a concentration of 300 mg/l (SA2) with 3% sucrose as well as the pre-harvest spraying method (B) on 8th day with an average of 0.88 mL/g FW. On the other hand, the minimum one was related to the DW treatment (distilled water and 20% sucrose) on the last day and in the pulse method (A1) with an average of 0.12 mL/g FW (Fig. 8).

The presence of bacteria in the vase solution causes their entry along with water into the plant as well as their accumulation on the surface of the cut parts and wooden vessels, which leads to the build-up of bacteria and lack of water absorption in the plant (Bleeksma & Van Doorn, 2003). Therefore, it seems that salicylic acid has increased the solution absorption through reducing bacteria population and preventing blockage of the wooden vessel.

CONCLUSION
The results of the present study suggested that in addition to the suppression of bacterial growth at stem-ends, the studied treatment extended the vase-life, reduced chlorophyll degradation, and enhanced superoxide dismutase activity in cut Alstroemeria flower. The most significant results were found in the case of salicylic acid at the rate of 50 ppm, where the pre-harvest (foliar) application of treatments was more effective than the one in post-harvest continuous or pulse methods.

CONFLICT OF INTEREST
The authors have no conflict of interest to report.

REFERENCES


