



Vase Life and Postharvest Attributes of *Alstroemeria aurea* 'Orange Queen' as Influenced by the Application of Sodium Nitroprusside Before and After Harvest

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ABSTRACT

Alstroemeria, with its high yields, stunning blooms, and a diverse color palette, has become a major player in the cut flower industry. However, its postharvest life presents a major hurdle: limited vase life. This, coupled with early leaf yellowing and flower drop (perianth abscission), significantly reduced the economic value of Alstroemeria due to accelerated senescence. We conducted an experiment to address this challenge and improve the vase life and other postharvest qualities of Alstroemeria 'Orange Queen' cut flowers. We investigated the application of sodium nitroprusside (SNP) at varying concentrations (0, 50, 100, and 200 μM) through two methods: preharvest foliar application (spraying leaves) and a short-term postharvest pulse treatment. A completely randomized factorial design was employed for the experiment. The experiment was conducted with three replications, and each replication consisted of three pots of flowers. We measured various factors to assess the effectiveness of SNP treatment, including vase life, relative solution uptake, relative fresh weight, relative water content, malondialdehyde (MDA) content (an indicator of membrane damage), and catalase enzyme activity (an antioxidant enzyme). The results demonstrated that SNP treatment significantly improved these postharvest characteristics. The most effective treatment involved a two-step approach: applying a foliar spray of 100 μM SNP before harvest, followed by a pulse treatment of 50 μM SNP during the vase life. This combination significantly extended the vase life to 16 d. The beneficial effects of SNP were dependent on both the dose applied and the timing of application. In conclusion, our findings suggest that SNP plays a multifaceted role in extending the vase life of cut Alstroemeria flowers. It appears to work by maintaining flower weight and water content, scavenging free radicals through enhanced antioxidant enzyme activity, and inhibiting lipid peroxidation, a process that damages cell membranes. This research provides valuable insights for growers and postharvest handlers seeking to improve the vase life and marketability of Alstroemeria cut flowers.

Abbreviations: Sodium nitroprusside (SNP), Nitric oxide (NO), Relative solution uptake (RSU), Relative fresh weight (RFW), Relative water content (RWC), Malondialdehyde (MDA), Catalase (CAT)

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Introduction

Alstroemeria hybrida L. from the family Alstroemeriaceae, is a recent introduction into the world's floriculture scene and has become a major cut flower. Flowers are simple or compound. Flower colors range from white to dark yellow to various pinks, violets, purples and reds. Yellow throats and black dots at the base of the petals and throats are a trademark of alstroemeria. It is a perennial herbaceous rhizomatous plant (Dhiman and Kashyap, 2021). Nitric Oxide (NO) is classified as a lipophilic free radical (Andrabi et al., 2023). This means it has an affinity for lipids and can easily travel through both the watery interior (cytoplasm) and fatty membranes of cells (Hayat et al., 2010 b). Significantly, NO is also a bioactive molecule, acting as a cellular messenger that influences various processes within the plant (Azizi et al., 2021). This multifunctional plant signal molecule plays a role in various plant processes including senescence (Hussain et al., 2022), regulation of stomatal movement (Lau et al., 2021), chlorophyll production, and anthocyanin biosynthesis (Camprubi et al., 2017), plant growth and development (Kolbert et al., 2019), plant defense signal against pathogen influence (Hussain et al., 2022), plant death, regulation of ion channels of guard cells, mitochondrial and chloroplast functionality (Kolbert et al., 2019), seed germination (Nabi et al., 2019), hypocotyl elongation, xylem differentiation, root development and regulation of photosynthesis (Camprubi et al., 2017).

Nitric oxide (NO) production plays a crucial role in plant resilience. It allows plants to adapt to changing environmental conditions, mediate defense responses against various abiotic stresses (Napieraj et al., 2020), and even prevent chlorophyll degradation under stress (Procházková et al., 2013). Nitric oxide can stimulate the production of many antioxidants in postharvest stages (Asghari, 2015). NO has some interactions with phytohormones. The interaction between NO and ethylene during the maturation and senescence of plant tissues suggested an antagonistic excitement of two gases (Kolbert et al., 2019). Ethylene is detrimental to the longevity of cut flowers by accelerating senescence. Research suggests NO may inhibit ethylene action and synthesis in plants (Seyf et al., 2012; Liu et al., 2022). Due to its eco-friendly nature, SNP emerges as a highly promising and novel substitute for hazardous silver thiosulphate (STS) in postharvest flower studies (Naing et al., 2017). Furthermore, research has shown that adding a nitric oxide

(NO)-releasing a chemical called 2,2-(hydroxyhynitrosodrazino)-bisethanamine (DETA/NO) to vase solutions improves the vase life of various commercially important flowers. This effect is observed across a range of flowers with varying ethylene sensitivity, including chrysanthemums (*Dendranthema grandiflora* RAM.), snapdragons (*Antirrhinum majus* L.), larkspurs (*Delphinium ajacis* L.), Asiatic lilies (*Lilium asiaticum* L.), and gerberas (*Gerbera jamesonii* L.) (Badiyan et al., 2004). SNP effectively reduces ethylene production, a gas known to trigger senescence (aging) and shorten the display life of ornamental flowers (Liao et al., 2013). Even in flowers with lower ethylene sensitivity, SNP can delay senescence by minimizing the production of reactive oxygen species (ROS) (Lone et al., 2021). Additionally, SNP is believed to enhance the activity of antioxidant enzymes within the flower. These enzymes help combat oxidative stress, a major contributor to flower aging, ultimately maximizing plant health and longevity (Allakhverdiev, 2020).

Given its ability to counteract ethylene action, NO emerges as a promising candidate for extending the vase life of various cut flowers, including Alstroemeria. Research reinforces this concept by demonstrating that applying sodium nitroprusside (SNP), a substance that releases NO, to cut roses, gladiolus, and cut lilies improved several quality factors (Dwivedi et al., 2016; Seyf et al., 2012; Dhiman and Parkash, 2013).

A study by Ashouri Vajari and Nalouisi (2013) demonstrated that applying NO to cut carnations (*Dianthus caryophyllus* L. "Tempo") significantly extended their vase life to 16.9 d. NO treatment also delayed petal wilting, maintained water balance within the flower, and enhanced the activity of antioxidant enzymes. Shabanian et al. (2018) investigated the effect of SNP in a vase solution for two gerbera species. Their findings showed that a concentration of 150 μ M SNP improved several quality aspects. Deng et al. (2019) studied the combined effects of NO (applied as sodium nitroprusside) and abscisic acid on cut roses. Their findings showed that both treatments increased the activity of antioxidant enzymes (superoxide dismutase, peroxidase, and ascorbate enzymes), which help combat oxidative stress and contribute to flower longevity. Additionally, Naziri Moghaddam et al. (2021) investigated the effects of SNP on various cut flowers, including roses (*Rosa hybrida* L.), lisianthus (*Eustoma grandiflorum*), and sunflowers (*Helianthus annuus* L.).

One of the most significant challenges faced by cut Alstroemeria flowers is the rapid yellowing of

leaves on the stem before petal abscission (dropping) (Seyed Hajizadeh et al., 2024). This yellowing can develop over several days and worsen rapidly (Ferrant et al., 2009). This is primarily due to the flower's high sensitivity to ethylene gas (Skutnik et al., 2021). Ethylene production increases rapidly after harvest, reducing flower quality, marketability, and overall economic value (Chamani et al., 2012). Sodium nitroprusside (SNP) is a compound that releases NO and has emerged as a promising and potentially eco-friendly approach to maintaining the quality and vase life of cut flowers. By delaying senescence and deterioration, SNP could significantly benefit the cut flower industry. This study aims to evaluate the effectiveness of applying SNP before and after harvest on the vase life and quality of cut *Alstroemeria* flowers and determine the optimal concentration of SNP for this application. By investigating these aspects, this study can contribute valuable insights into the potential use of NO, via SNP, for extending the vase life and improving the commercial value of cut *Alstroemeria* flowers.

Material and Methods

Plant materials and growing conditions

This research, conducted at Urmia University's Department of Horticultural Science (2020-2021), investigated the effects of pre- and postharvest SNP application on the vase life and quality of cut *Alstroemeria* flowers. *Alstroemeria aurea* 'Orange Queen' (Royal Van Zan Ten-Netherlands), known for its vibrant orange blooms with dark speckles (Faust and Dole, 2021), was used in the experiment. Rhizomes, obtained from a commercial greenhouse, were planted in 24 cm diameter by 19 cm tall plastic pots filled with a soilless mixture of perlite and cocopeat (1:3 ratio by volume). The greenhouse environment was maintained at a temperature max/min of 18-21 °C/10-13 °C, respectively, a light duration of 10-12 h, and a light intensity of 400-500 $\mu\text{mol m}^{-2} \text{s}^{-1}$. Plants were fertilized three times a week using a specific nutrient solution detailed in Table 1.

Treatment with sodium nitroprusside (SNP)

In this experiment preharvest foliar application of SNP (Fluka company- Switzerland) at concentrations of 0, 50, 100 and 200 μM was applied jointly with post-harvest application of SNP (as pulsing) with the same concentrations, 0, 50, 100 and 200 μM .

For preharvest application, about one month after planting and greenhouse establishment, foliar spraying of plants commenced. SNP solutions were sprayed at two-week intervals for four months. For postharvest application, cut flowers underwent a 24 h pulse treatment with SNP solutions. Cut flowers were harvested early in the morning when one to two florets were open. Harvested flowers were immediately placed in buckets containing tap water. Flowers were then directly transported to the laboratory. Upon arrival at the lab, the stems of the cut flowers were recut to a uniform length of 40 cm under distilled water. Finally, the cut flowers were treated with SNP solutions (the NO donor) at various concentrations. The red solid compound was dissolved in distilled water to create solutions at three concentrations: 50, 100, and 200 μM . Distilled water alone served as the control treatment (0 μM). Due to the compound's short half-life, the solutions were applied immediately after preparation. Four cut *Alstroemeria* stems were placed in each 500 mL flask containing the prepared SNP solutions. The flowers were held in these solutions for 24 h in the laboratory to allow for sufficient uptake of the compound by the stems. Following the 24 h exposure period, the flower stems were transferred to flasks containing only distilled water. The flowers remained in this distilled water environment until the end of their vase life (wilting). Throughout the experiment, the environment was maintained at a constant temperature of 22 °C \pm 1 °C, with a relative humidity of around 70%. Artificial lighting was provided using fluorescent lamps set to a 12 h photoperiod, with a light intensity of 13 $\mu\text{mol m}^{-2} \text{s}^{-1}$.

Table 1. Nutritional program used for *Alstroemeria* for 100 L nutrient solution.

Mg(SO ₄) ₂	KNO ₃	NH ₄ NO ₃	Na ₂ MoO ₄	Na ₂ [B ₄ O ₅] (OH) ₄	MA P	MnSO ₄	ZnSO ₄	K ₂ SO ₄	Fe chelate 6%	5Ca (NO ₃) ₂ - NH ₄ NO ₃ . 10H ₂ O
10 g	32 g	4 g	0.035 g	0.03 g	5 g	0.2 g	0.15 g	8 g	5 g	10g

Relative fresh weight (RFW) of flowers

The experiment employed relative fresh weight (RFW) to quantify changes in flower weight over time. This measurement is based on the method described by Joyce and Jones (1992).

The RFW is calculated using the following formula:

$$RFW = FW_i / FW_0$$

FW_i: This represents the weight of the flower stem (g) at a specific d during the experiment.

FW₀: This represents the initial weight of the same flower stem (g), measured on the first d of the experiment.

Vase life

The vase life of the cut *Alstroemeria* flowers was determined by daily observation, following the methodology established by Ferrant et al. (2002) and Mutui et al. (2006).

Relative water content (RWC) of petals

The RWC from florets at different stages was determined by Anderson et al. (2004). The fresh weight (FW) of detached florets was recorded by placing them in pre-weighed test tubes with distilled water and again recording the weight of test tubes. The increase in weight of the test tube was FW of florets. The tubes were again weighed after 4 h and now the increase in weight was turgid weight (TW). The florets were then oven dried at 72 °C for 48 h and weight was recorded as dry weight (DW). The RWC was calculated as:

$$RWC = \frac{FW - DW}{TW - DW} \times 100$$

Relative solution uptake (RSU)

The experiment assessed the rate at which the cut *Alstroemeria* flowers absorbed the SNP solutions. This measurement is based on the method established by Aelaei et al. (2017) and was expressed in ml per gram of initial fresh weight by the following formula:

$$RSU = WU_i / FW_0$$

RSU= Relative solution uptake

WU_i= vase solution weight (g) on t = d 5 and 10.

FW₀= Primary fresh weight

Malondialdehyde (MDA)

Petal samples were weighed (0.2 g) and homogenized in 5 mL of a 1% trichloroacetic acid (TCA) solution. The homogenized mixture was centrifuged at 8,000 g for 10 min. 1 mL of the supernatant was mixed with 4 mL of a solution containing 0.5% thiobarbituric acid (TBA) in 20% TCA. The mixture was heated at 95 °C for 30 min. The mixture was then cooled down in an ice bath.

Another centrifugation step was performed at 10,000 g for 10 min to separate any remaining solid material. The absorbance of the final supernatant was measured at two wavelengths: 532 and 600 nm. The MDA content was measured when the extinction coefficient of the sample was 155 mM cm⁻¹ (Horst and Cakmak, 1991).

$$MDA (\mu\text{mol g}^{-1} \text{FW}) = \left(\frac{A_{532} - A_{600}}{155} \right) \times 100$$

Catalase (CAT) activity assay

For the catalase enzyme assay, 500 mg of tissue was extracted in 2.5 mL of 50 mM phosphate buffer (pH 7) at 0 °C by centrifuging at 10,000 g for 15 min and then collecting supernatant to make the final volume to 100 mL. Catalase activity was determined by Aebi (1984). The absorbance of the supernatant was calculated at 240 nm. The CAT activity was measured when the extinction coefficient of the sample was 43.6 mM cm⁻¹.

$$\begin{aligned} & \text{unit } \frac{\text{mM}}{\text{min}} \\ & = \frac{do D \text{ min}^{-1} (\text{slope}) \times \text{vol. of assay}}{\text{Extinction coefficient (43.6)}} \end{aligned}$$

Statistical analysis of data and software used

The researchers used a factorial design, where they investigated the combined effects of three factors on the *Alstroemeria* flowers. The first factor involved pre-treating the plants with different concentrations of SNP (0, 50, 100 and 200 μM) before harvest. The second factor involved applying SNP solutions (0, 50, 100 and 200 μM) to the cut flowers after harvest. Finally, the flowers were monitored and measured at three different time points (0th, 5th and 10th d) during their vase life, as the third factor. The experiment was conducted with three replications, and each replication consisted of three pots of flowers. Statistical software (SAS ver. 9.2) was used to analyze the collected data. To identify significant differences between treatment groups, Tukey's multi-domain test was applied with a very strict threshold for significance (p-value less than 0.01).

Results**Water relations**

Analysis of mean data revealed a treatment with a statistically significant effect on relative fresh weight at both measurement points. This treatment effectively protected against flower weight loss compared to the control and other treatments throughout the experiment. Flowers pretreated with a 100 μM SNP solution before

harvest followed by a short 50 μM SNP pulse treatment after harvest exhibited the most effective maintenance of relative fresh weight

(RFW) throughout their vase life compared to other treatment combinations (Fig. 1).

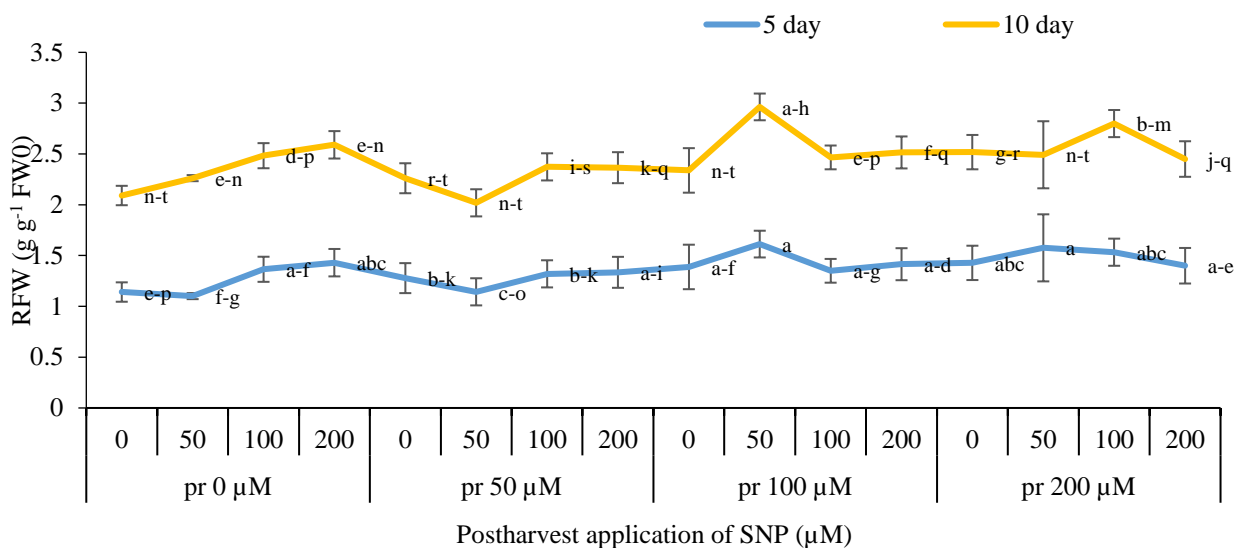


Fig. 1. Effect of pre- and post-harvest application with different concentrations of SNP over vase life period on relative fresh weight of *Alstroemeria* 'Orange Queen' (pr: preharvest application of SNP (μM)). (Error bars represent SE from three replicates). Different letters indicate statistically significant differences between the treatments and control in ($P \leq 0.01$).

Based on the results obtained from the mean data comparison, it was found that all pre-harvest and postharvest sodium nitroprusside (SNP) applications increased relative solution uptake (RSU) compared to the control group. However, RSU gradually decreased over time. As shown in Figure 2, the treatment with 100 μM SNP before harvest and 200 μM SNP after harvest exhibited the highest RSU on d 5, but it declined sharply by

d 10. Therefore, pre-harvest application of a moderate SNP concentration (100 μM) followed by a moderate post-harvest pulse treatment (either 50 or 100 μM) yielded the most consistent improvement in *Alstroemeria*'s solution uptake throughout their vase life (Fig. 2). This suggests this treatment combination might be ideal for maintaining flower hydration.

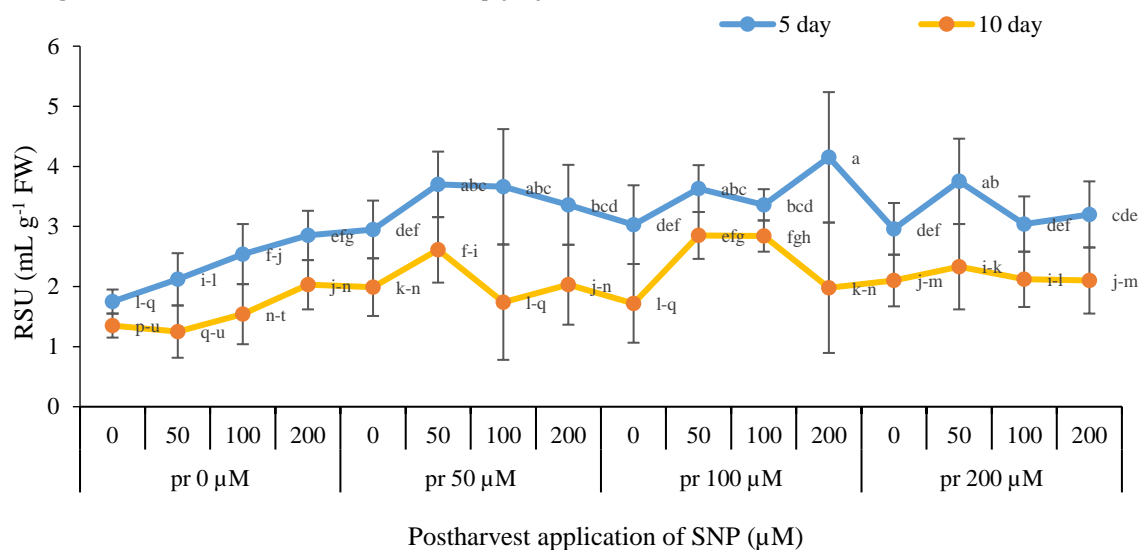


Fig. 2. Effect of pre- and post-harvest application with different concentrations of SNP over vase life period on relative solution uptake of *Alstroemeria* 'Orange Queen' (pr: preharvest application of SNP (μM)). (Error bars represent SE from three replicates). Different letters indicate statistically significant differences between the treatments and control in ($P \leq 0.01$).

Pre-harvest application of either 50 or 100 μM SNP solution, followed by a brief post-harvest pulse treatment with 50 μM SNP solution, significantly improved water retention in flower petals throughout the vase life compared to the

control group (untreated flowers). This finding, as illustrated in Figure 3, suggests that these treatment combinations may be beneficial in extending flower freshness.

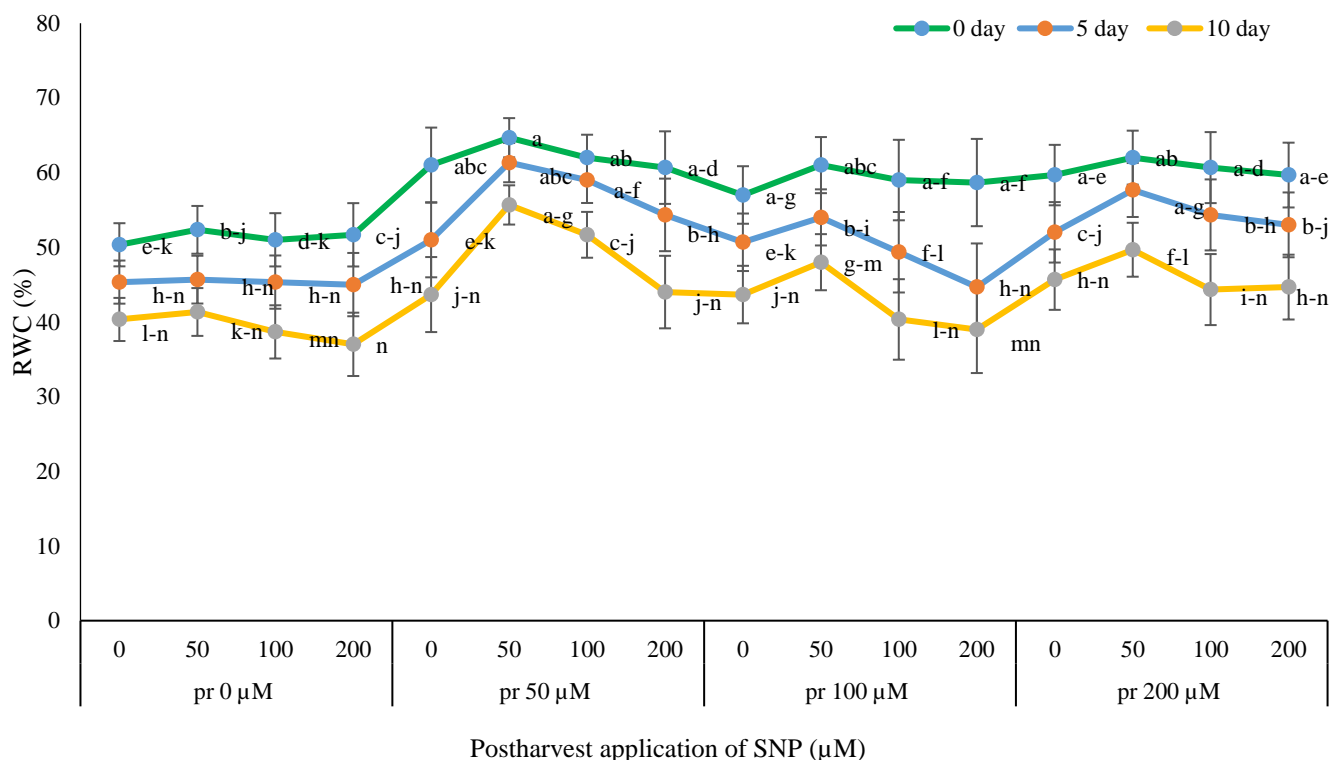


Fig. 3. Effect of pre- and post-harvest application with different concentrations of SNP over vase life period on relative water content of *Alstroemeria* 'Orange Queen' (pr: preharvest application of SNP (μM)). (Error bars represent SE from three replicates). Different letters indicate statistically significant differences between the treatments and control in ($P \leq 0.01$).

Oxidative damage

Sodium nitroprusside (SNP) application in both pre-harvest and post-harvest stages significantly reduced malondialdehyde (MDA) levels compared to the control group across all three measurement times. Only the 200 μM SNP treatment (both pre-harvest and post-harvest) showed no significant difference from the control on d 5 and 10. Notably, MDA content increased over time in all treatments, although these levels remained lower compared to the control. In line with previous findings, pre-harvest application of a moderate SNP concentration (100 μM) followed by a moderate post-harvest pulse treatment (50 μM) resulted in the most substantial reduction in malondialdehyde (MDA) content, a well-known marker of floral stress (Fig. 4). This suggests that this treatment combination might be the most effective in mitigating oxidative stress and potentially prolonging flower lifespan.

Pre-harvest treatment with a 100 μM SNP solution followed by a brief 50 μM SNP pulse after harvest resulted in the most significant increase in catalase activity compared to the control group (untreated flowers). This aligns with the previously observed reduction in malondialdehyde (MDA) content, a marker of stress, as catalase is an enzyme that helps alleviate oxidative stress (Fig. 5). The overall rise in catalase activity over time likely represents a natural response of the flowers to the aging process (senescence).

Vase life

Pre-harvest application of a moderate sodium nitroprusside (SNP) concentration (100 μM) followed by a moderate post-harvest pulse treatment (50 μM) significantly extended the vase life of *Alstroemeria* flowers compared to the control group (untreated flowers) (Fig. 6). This

suggests that this specific treatment combination might be ideal for maximizing the vase life of cut Alstroemeria. Notably, this treatment resulted in a remarkable increase in vase life. This treatment

resulted in a 2.3-fold increase in the vase life of Alstroemeria flowers compared to the control (Fig. 6).

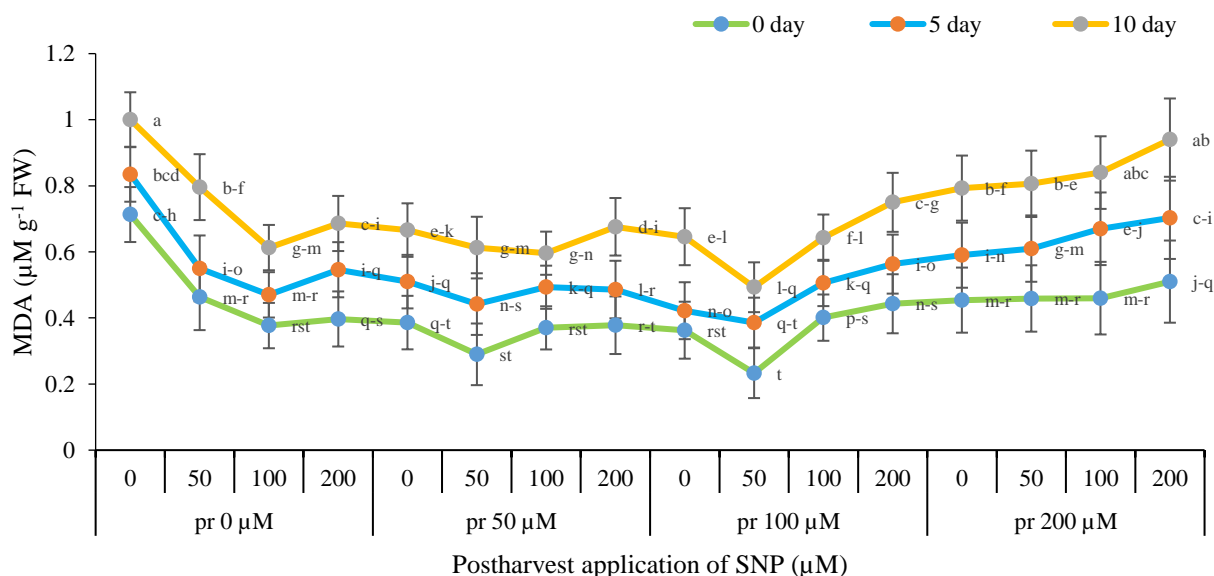


Fig. 4. Effect of pre- and post-harvest application with different concentrations of SNP over vase life period on malondialdehyde content of Alstroemeria ‘Orange Queen’ (pr: preharvest application of SNP (µM)). (Error bars represent SE from three replicates). Different letters indicate statistically significant differences between the treatments and control in (P ≤ 0.05).

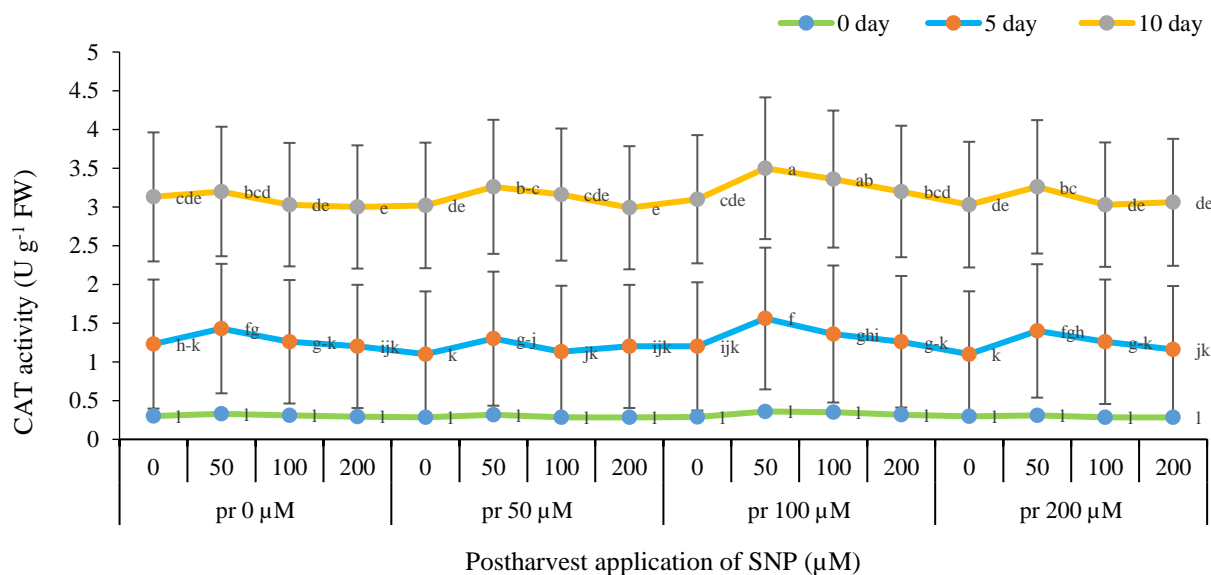


Fig. 5. Effect of pre- and post-harvest application with different concentrations of SNP over vase life period on CAT activity of Alstroemeria ‘Orange Queen’ (pr: preharvest application of SNP (µM)). (Error bars represent SE from three replicates). Different letters indicate statistically significant differences between the treatments and control in (P ≤ 0.01).

Discussion

Water relations

Postharvest senescence and quality deterioration

are major hurdles for the cut flower trade and export sector. This highlights the importance of research into effective postharvest treatments to

maintain flower freshness, a goal of immense value to the floriculture industry. This study investigated the effect of applying sodium nitroprusside in two stages (before and after harvest) on mitigating flower weight reduction during the vase life period. Compared to the control group, flowers treated with sodium nitroprusside exhibited a significantly smaller decrease in weight. Fresh weight retention is

known to be closely linked to the vase life of cut flowers. Maintaining fresh weight throughout the vase life period is therefore crucial for evaluating both vase life and postharvest flower quality. Studies, such as one by Liao et al. (2013), have shown a positive correlation between flower weight and vase life, with heavier flowers typically lasting longer.

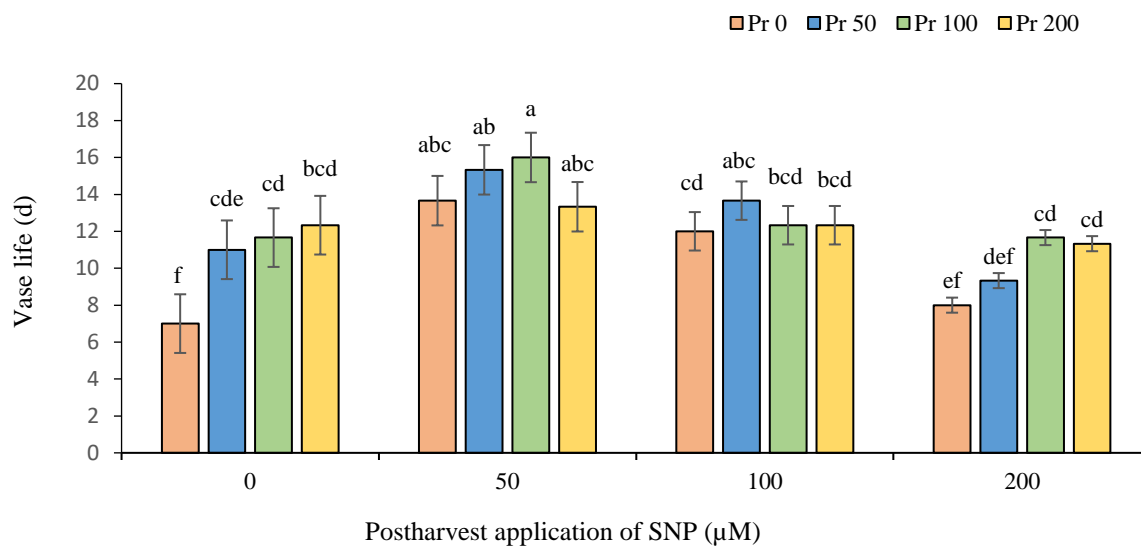


Fig. 6. Effect of pre- and post-harvest application with different concentrations of SNP on vase life of *Istroemeria* 'Orange Queen' (pr: preharvest application of SNP (µM)). (Error bars represent SE from three replicates). Different letters indicate statistically significant differences between the treatments and control in ($P \leq 0.01$).

While nitric oxide (NO) is known to promote water uptake in cut flowers, leading to increased relative fresh weight (RFW), its role in plant stomata is more complex. Recent research (Van Meeteren et al., 2020) suggests that NO is not the key factor in a specific type of stomatal closure triggered by abscisic acid (ABA). However, it might act independently, influencing stomatal closure under other circumstances, particularly when mesophyll tissue is involved. This finding points to an alternative mechanism, plant may utilize to restrict water loss during stress by regulating stomatal aperture. This closure consequently leads to reduced transpiration, ultimately lowering the flower's osmotic pressure.

Building on the previous discussion about stomatal closure, the pre-harvest application of sodium nitroprusside (SNP) might offer an additional benefit. As an NO-releasing compound, SNP could potentially stimulate endogenous β -glucuronase activity within the flower tissue. This enzyme is crucial for breaking down complex

sugars in cell walls, promoting cell wall loosening and facilitating cell development and overall growth. Studies have shown that a lack of nitric oxide (NO) in *Arabidopsis* mutants leads to reduced β -glucuronase activity, hindering plant growth. Since growth is associated with increased turgor pressure (cell wall pressure) and consequently higher water content, NO deficiency can result in decreased turgor pressure and lower fresh weight in plants (Hayat et al., 2010a). This suggests that SNP application might enhance β -glucuronase activity, potentially promoting cell wall loosening and ultimately contributing to improved water relations and flower quality.

Nitric oxide's (NO) influence extends beyond water relations, affecting various plant processes like stem elongation, meristem development, and communication through the vascular system. Its presence in both vascular tissues and epidermal cells further supports this link (Allagulova et al., 2023). In this context, the pre-harvest SNP application in our study could potentially enhance vascular development, ultimately

improving water transport and contributing to the observed reduction in flower weight loss. Furthermore, our findings align with existing research demonstrating the positive effects of NO on flower RFW and the effectiveness of SNP in enhancing RFW across various flower types, including gladiolus (Mittal and Shalini, 2021), roses (Mirzaei Esgandian and Jabbarzadeh, 2020), and gerberas (Karamian et al., 2020).

Alstroemeria flowers treated with the combination of pre- and post-harvest sodium nitroprusside (SNP) application (spray and pulse treatment) exhibited increased solution uptake and superior performance compared to the control group (Fig. 2). This aligns with prior research (Fanourakis et al., 2022) highlighting the critical role of water status, particularly petal turgor, in cut flower vase life. After harvest, water uptake from the parent plant ceases, while transpiration continues, leading to water loss and wilting. Wilted petals become aesthetically unappealing and significantly shorten vase life (Ahmadi-Majd et al., 2021; Fanourakis et al., 2021a, 2020b). Therefore, maintaining optimal water balance is essential for extending vase life. SNP application potentially benefits flowers in two ways: by improving water uptake and possibly by enhancing xylem differentiation, which could facilitate water transport (Lutter et al., 2024). SNP effectiveness is dose-dependent. While low concentrations promote water uptake, high concentrations can disrupt translocation channels within the plant (Zangani et al., 2023). Additionally, high SNP doses might increase microbial growth in the vase solution and on the stem surface, as well as air embolism in xylem tissues – all factors that can hinder water uptake. Beyond its role in water uptake, SNP has also been shown to improve flower quality by increasing stem strength and chlorophyll retention (Zhang et al., 2019), indirectly contributing to longer vase life.

Flowers treated with SNP displayed a greater capacity to retain water, as evidenced by their higher relative water content (RWC) and relative fresh weight (RFW). Maintaining water relations during postharvest storage is crucial for extending vase life. Conversely, disrupted water balance accelerates flower senescence (Hassan and Ali, 2014; Hassan et al., 2014; Fahmy et al., 2020). These studies also suggest that maintaining fresh weight can be attributed to increased water uptake and/or reduced transpiration, ultimately preventing weight loss. Our findings align with previous research indicating that SNP treatment improves water relations and increases RWC throughout the vase life period (Abbasi et al., 2020; Sadeghi Faragheh

et al., 2017; Ghaei, 2018).

Oxidative damage

The application of sodium nitroprusside (SNP) in both pre-harvest and post-harvest stages effectively reduced the level of malondialdehyde (MDA) in the flowers, although MDA content did exhibit a gradual increase over time. MDA is a well-established marker of oxidative stress, as its concentration reflects the level of damage to cell membranes. Therefore, maintaining membrane stability plays a critical role in delaying senescence during cut flower postharvest by inhibiting processes like electrolyte leakage, sugar loss, pigment degradation, and solute leakage (Ghadakchiasl et al., 2017). By reducing MDA content, SNP application helps flowers maintain membrane integrity and minimize stress-related damage, ultimately contributing to extended vase life. Our findings demonstrate that SNP concentrations (50 and 100 μM) effectively mitigated the changes in lipid peroxidation reflected by MDA production. These results are consistent with previous research by Mittal et al. (2021), Kazemzadeh-Beneh et al. (2018), and Mohasseli and Sadeghi (2017), who all reported that SNP treatment extended flower vase life. Senescence in cut flowers is a complex process involving a tightly regulated sequence of physiological and biochemical events. These events include the degradation of proteins and DNA, lipid peroxidation and membrane leakage, macromolecule breakdown, cellular compartmentalization, floral abscission, color changes, leaf yellowing, and weight loss (Naing et al., 2017). By mitigating oxidative stress and maintaining membrane integrity, SNP application can potentially delay the onset of these senescence-related processes, thereby extending the vase life of cut flowers. The observed changes during senescence are often triggered by an increase in reactive oxygen species (ROS) production. This imbalance between ROS and antioxidant enzymes leads to secondary oxidative damage and loss of cell membrane permeability (Hasanuzzaman et al., 2021). Lipid peroxidation, a complex process initiated by the removal of hydrogen atoms from unsaturated fatty acids, ultimately results in the formation of malondialdehyde (MDA). Sodium nitroprusside (SNP), as a nitric oxide (NO) donor, appears to mitigate lipid peroxidation through three potential mechanisms. First, NO directly scavenges reactive lipid species and peroxide radicals, effectively terminating the peroxidation chain and consequently reducing MDA production (Niu et al., 2023). Second, SNP may

inhibit the activity of lipoxygenase, a key enzyme involved in lipid peroxidation, by potentially reducing the iron (Fe^{3+}) in its active site to Fe^{2+} (Naziri Moghaddam et al., 2014). Finally, SNP may stimulate the production of antioxidant enzymes, which scavenge free radicals and prevent cell membrane damage. This indirect antioxidant effect further contributes to the reduction of MDA levels (Mohasseli and Sadeghi, 2017). It's important to acknowledge that, as reported in previous studies (Dwivedi et al., 2016), high concentrations of nitric oxide (NO)-releasing compounds can have adverse effects on plants, with the degree of toxicity varying depending on the plant species. In our study, the 200 μM SNP concentration indeed exhibited a trend of increasing malondialdehyde (MDA) levels. This suggests that this concentration might exceed the optimal level for *Alstroemeria*, potentially causing some degree of cellular stress and consequently leading to higher MDA production.

Figure 5 shows that pre- and post-harvest application of sodium nitroprusside (SNP) increased catalase activity, an essential antioxidant enzyme. During the postharvest stage, reactive oxygen species (ROS) production is a major threat to flowers and plants. ROS can damage cell membranes and diminish antioxidant levels, ultimately accelerating senescence. Under normal conditions, plants produce low levels of ROS in various tissues. However, stress conditions and senescence can significantly increase ROS levels. Previous research has shown that excessive ROS can damage cellular components like proteins, RNA, DNA, and membranes, while also triggering lipid peroxidation (Lin et al., 2019). Research has documented that nitric oxide (NO) acts as both an antioxidant and a cytotoxic agent, depending on its interaction with free radicals (Dwivedi et al., 2016). Beyond directly scavenging free radicals, NO can indirectly enhance antioxidant defenses by stimulating the production of antioxidant enzymes. This further suppresses free radicals and delays senescence (Seyf et al., 2012; Lau et al., 2021). NO may achieve this by increasing the activity of specific iron-containing antioxidant enzymes, such as catalase (CAT) and superoxide dismutase (SOD) (Lau et al., 2021). Research suggests that low concentrations of NO can trigger the expression of genes involved in the synthesis of protective antioxidant enzymes, further bolstering the cellular defense system against ROS (Fan et al., 2014). Additionally, NO can react directly with reactive oxygen species (ROS) to form peroxynitrite. At normal cellular pH levels, peroxynitrite can decompose into harmless products or react with damaging hydrogen

peroxide radicals, ultimately reducing overall ROS levels (Fan et al., 2014). The observed increase in catalase activity in our study aligns with the proposed mechanism and supports the role of SNP in enhancing the antioxidant defense system of *Alstroemeria* flowers. These findings are consistent with previous research demonstrating positive effects of NO on antioxidant enzyme activity in gerbera (Naing et al., 2017) and gladiolus (Mittal and Shalini, 2021) treated with SNP. It's crucial to remember that high SNP concentrations (200 μM) can be detrimental, causing a decrease in antioxidant enzyme activity and overall antioxidant capacity. Studies have shown that the negative effects of high SNP on catalase activity are dose-dependent (Seyf et al., 2012). Therefore, to maximize catalase activity in our study, we selected a pre-harvest foliar application of 100 μM SNP combined with a 50 μM post-harvest pulse treatment as the optimal concentrations.

Vase life

This study investigates the use of sodium nitroprusside (SNP) as a way to extend the vase life of *Alstroemeria* cut flowers. Senescence, a major hurdle in maintaining cut flowers, is often triggered by reactive oxygen species (ROS) that overwhelm the plant's natural antioxidant defenses (Howard, 2013). In recent years, nitric oxide (NO) has emerged as a promising antioxidant molecule in plants (Khator et al., 2024). However, its effectiveness is highly dependent on both the concentration used and the specific plant species. High NO concentrations can actually worsen oxidative stress through a process called nitrosative stress, leading to increased cellular damage (Wang et al., 2021). While high NO acts like free radicals and harms cellular components, low concentrations are essential for plant growth and function as part of the plant defense system. This duality highlights NO's potential for both beneficial and harmful effects (Salachna and Byczynska, 2017). Our findings mirrored previous observations regarding the importance of concentration. Pre-harvest application of high concentration SNP (200 μM) resulted in a reduction in vase life. Conversely, the application of 50 μM SNP yielded the longest vase life, demonstrating the positive impact of low SNP concentrations. Sensitivity to ethylene and water stress are major limitations to the postharvest life of cut flowers (Naziri Moghaddam et al., 2021). In our study, the combined pre-harvest (100 μM) and post-harvest (50 μM) application of SNP not only extended vase life but also reduced water loss. Yellowing

leaves and a shortened vase life are significant postharvest concerns for *Alstroemeria*. Our results suggest that SNP application, as a source of exogenous NO, can be a valuable tool for enhancing the postharvest performance of *Alstroemeria* cut flowers. We believe this positive effect is achieved through NO's ability to protect plants from oxidative stress and delay senescence via two mechanisms: its direct antioxidant activity and its modulation of defensive gene expression (Shi et al., 2016). These findings align with previous research demonstrating the effectiveness of SNP in extending the vase life of various cut flowers, including *Eucomis* (Salachna and Byczynska, 2017), *Gerbera* (Ghaei et al., 2018), roses (Mirzaei Esgandian and Jabbarzadeh, 2020), *gladiolus* (Dwivedi et al., 2016; Kazemzadeh-Beneh et al., 2018).

Conclusion

In conclusion, the application of sodium nitroprusside (SNP) as a pre- and post-harvest treatment for *Alstroemeria* 'Orange Queen' cut flowers has demonstrated promising potential to extend vase life and enhance overall postharvest quality. The observed benefits are attributed to SNP's ability to mitigate oxidative stress by scavenging free radicals, enhancing antioxidant enzyme activity, and reducing lipid peroxidation. Several factors, including fresh weight content, water uptake, antioxidant enzyme activity, and lipid peroxidation, were found to be key determinants of vase life. The results demonstrated that both foliar application and supplementation of the vase solution with SNP effectively enhanced relative fresh weight (RFW) and water uptake. The effectiveness of SNP was found to be concentration-dependent, with a pre-harvest application of 100 μ M SNP combined with a post-harvest pulse treatment of 50 μ M SNP yielding the most optimal results. These findings suggest that SNP can be a valuable tool for florists and growers seeking to extend the marketability and consumer satisfaction of *Alstroemeria* cut flowers.

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Conflict of Interest

The authors indicate no conflict of interest in this work.

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