

Conservation of *Alstroemeria* cut flowers stored under refrigeration

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ABSTRACT. *Alstroemeria* flowers have shown great importance in the world trade of cut flowers due mainly to its beauty and wide variety of colors. However, the durability of its inflorescences is usually hampered by the rapid yellowing of the leaves, which impairs their decorative quality. Cut flowers require the use of technologies to improve postharvest quality and floral longevity. This research aimed to study the postharvest conservation of inflorescences of *Alstroemeria* cv. Ajax at different storage temperatures. Floral stems were placed in containers with distilled water and stored at four temperatures (4, 8, 12, and 22°C) for 12 days. The following analyses were performed: fresh mass variation, respiratory activity, relative water content, soluble and reducing carbohydrate contents, polyphenol-oxidase and peroxidase enzymes, pigments (anthocyanin and carotenoids), and longevity. The experiment was conducted in a completely randomized design, the results were submitted to analysis of variance (ANOVA), and the effect of treatments submitted to F-test. Significant differences were compared using the least significant difference (LSD) at 95% confidence interval ($p \leq 0.05$). The temperatures of 8 and 12°C were effective in maintaining the postharvest quality of inflorescences during storage period, as they remained turgid due to transpiration reduction caused by low temperatures, and longevity reaching 46 and 22 days, respectively.

Keywords: longevity; pigments; storage.

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Introduction

Alstroemerias flowers (*Alstroemeria hybrid* L.) have shown great importance in the world trade of cut flowers due mainly to its beauty and wide variety of colors (Ferrante, Hunter, Hackett, & Reid, 2002). However, the durability of its inflorescences is usually hampered by the rapid yellowing of the leaves, which impairs their decorative quality.

Cut flowers deteriorate quickly and need technologies to prolong its lifespan. Although several methods have been employed to increase the durability of cut flowers, low-temperature storage is still the most widely used because it enables maintenance of flower quality (Chitarra & Chitarra, 2005). This technique reduces plant metabolism, directly reducing transpiration, respiratory rate, and ethylene production by slowing degradation of sugar reserves, thus extending the durability of flower stems (Nowak, Goszcznska, & Rudnicki, 1991). Cold storage helps maintain the longevity of cut flowers, allowing them to be shipped and received in perfect quality. Many studies have shown that low temperatures are effective in maintaining the postharvest quality of cut flowers, with significant results in extending the useful life thereof. Bellé, Mainardi, Mello, and Zachet (2004) said that storage at 2°C reduced the symptoms of senescence in *Dendranthema grandiflora*. Inflorescences of gerbera cv. Suzanne stored at 2, 4, and 6°C showed longer useful life than those stored at 20°C (Durigan & Mattiuz, 2009).

Sardoei, Mohammadi, and Shahdadneghad (2014) observed a positive and significant effect on increasing the life of narcissus stored at 4°C, delaying the senescence of these flowers. The temperature of 4°C significantly increased the life of inflorescences of gerbera cv. Intenza, which was three times longer than that of inflorescences maintained at 22°C (14.8 days vs. 5.0 days).

Due to the above and the lack of information related to the postharvest conservation of alstroemerias, this study aimed to find the best storage temperature for inflorescences of red *Alstroemeria* cv. Ajax, considering the factors associated with floral senescence and loss of decorative life.

Material and methods

This experiment was carried out using inflorescences of red *Alstroemeria* cv. Ajax from a commercial cultivation in Andradas city, Minas Gerais, Brazil (22° 4' 19" S, 46° 34' 20" W, 913 m altitude). The flowers were harvested early in the morning and then transported in an air-conditioned vehicle, for about 4 hours, to the Laboratory of Postharvest Technology at the Unesp Campus of Jaboticabal, São Paulo State (Brazil).

Upon arriving at the laboratory, stems were standardized to 70 cm in length and the leaves of the base that would be in contact with the maintenance solution were dropped. Afterwards, damaged stems and those that did not present homogeneity for commercial harvest (primary flowers closed but showing the color) were discarded.

After standardization, nine inflorescences were randomly selected for an initial assessment, and other 216 inflorescences were randomly distributed into containers with distilled water and stored under four conditions ($4 \pm 1^\circ\text{C}$ and $89 \pm 2\%$ RH, $8 \pm 1^\circ\text{C}$ and $90 \pm 1\%$ RH; $12 \pm 1^\circ\text{C}$ and $88 \pm 2\%$ RH and $22 \pm 2^\circ\text{C}$ and $85 \pm 4\%$ RH).

The experiment was conducted in a completely randomized design with three repetitions of three inflorescences each. Every three days, inflorescences were evaluated for 'variation of fresh mass', in which stems were weighed in a precision scale (0.01 g). In this analysis, negative values indicate gain of fresh mass and positive ones point to loss of fresh mass. Then, the 'amount of CO₂ produced' (respiratory activity) was quantified using a gas chromatograph (Trace GC Ultra, Thermo Scientific), in which air samples were removed from hermetically sealed containers where flowers were maintained for one hour. 'The relative water content (RWC) of petals' was evaluated using 10 petal disks (10 mm diameter) from each repetition and calculated by the following equation (Kramer, 1983): $RWC = (\text{fresh mass} - \text{dry mass}) / (\text{turgid mass} - \text{dry mass}) \times 100$. 'The levels of soluble and reducing carbohydrates' were estimated using petals and quantified following Mattiuz, Rodrigues, Mattiuz, Pietro, and Martins (2010). 'The polyphenol-oxidase (PPO) activity of petals' was determined according to the method described by Adnan, Augustin, and Ghazali (1986). 'Peroxidase (POD) activity' was measured according to Matsuno and Uritani (1972). Lastly, contents of anthocyanins (Francis, 1982) and carotenoids (Lichtenthaler, 1987) were determined using samples of alstroemeria petals, and results expressed as 100 mg g⁻¹ fresh mass.

For 'floral longevity', a set of stems were separated from each treatment to evaluate the following decorative parameters: floral opening, leaf color, falling anthers, and flower turgidity. We used as core scale ranging from 5 to 1, wherein, score 5 (maximum): 100 closed and turgid flowers, 100 green leaves, and 100% attached anthers; score 4: 75 opened and turgid flowers, 75 green leaves, and 75% attached anthers; score 3: 50 opened and turgid flowers, 50 green leaves, and 50% attached anthers; score 2: 25 opened and turgid flowers, 25 green leaves, and 25% attached anthers; score 1: 100% opened and wilted flowers, and fully yellowed leaves, and absence of anthers. This method was described by Galati et al. (2015), who considered a score 3 as trade limiting.

Data were submitted to analysis of variance (ANOVA), and when treatment effect was significant, they were submitted to F-test. Means were compared by the Tukey's test ($p \leq 0.05$). When differences between treatments were greater than the sum of standard deviations, the result was considered significant (Steel & Torrie, 1980).

Results and discussion

Alstroemeria stems stored at 8 and 12°C gained mass until the 12th day of storage (Table 1). This was probably due to a positive water balance, wherein inflorescences maintained at such temperatures absorbed more water than they lost. It is because lower temperatures reduce transpiration rate, thereby reducing the loss of water (Vilas Boas, 2000). These findings are following Santos et al. (2012), who observed a gain of fresh mass in *Epidendrum ibaguense* flowers placed in water and kept at 10°C.

For the stems which were stored at 4°C, the loss of fresh mass started from the 6th day after storage (Table 1). This water loss may be related to stress due to low storage temperature, causing water loss in tissues (Finger, Moraes, Barbosa, & Grossi, 2003).

Table 1. Variation of fresh mass (%) and relative water content (%) in stems of *Alstroemeria* 'Ajax' stored at four temperatures (4, 8, 12, and 22°C).

Treatment	Day of Storage				
	0	3	6	9	12
Variation of fresh mass (%)					
4°C	0 A	-0.69 A	-3.45A	0.46 AB	6.19 AB
8°C	0 A	-9.78 A	-10.89 A	-12.70 B	-17.62C
12°C	0 A	-3.20 A	-6.86 A	-8.47 B	-6.26 BC
22°C	0 A	-3.90 A	6.59A	13.84 A	19.38 A
Relative Water Content (%)					
4°C	86.83 A	82.40 B	78.77 A	78.77 B	81.27 A
8°C	86.83 A	84.47 AB	85.87 A	89.80 A	88.07 A
12°C	86.83 A	87.90 AB	88.03 A	86.23 AB	86.17 A
22°C	86.83 A	92.43 A	87.43 A	78.03 B	66.83 B

*Means followed by the same letter in the column are not significantly different from each other by the Tukey's test at 5% probability.

As for the stems stored at 22°C, a severe loss of fresh mass was observed after the 3rd day of storage (Table 1), possibly due to the higher water loss through transpiration. High transpiration rate combined with a limited water absorption, due to solution composition or high strength and low hydraulic conductance of xylem vessels, are the main factors negatively influencing maintenance of postharvest quality of cut flowers (Vieira, Mendes, Finger, & Barbosa, 2012; Saleem, Khan, Ahmad, & Ahmad, 2014; Costa et al., 2015). Therefore, a positive water balance should be maintained, which, in this study, was kept at 8 and 12°C. Liu et al. (2009) reported that low temperatures (8 to 10°C) decreased water loss and inhibit microbial growth in lilies, hence less obstruction of xylem vessels and better water absorption, as was found in this experiment.

Regarding the relative water content, stems stored at 22°C showed a decrease from the 6th day of storage, which is consistent with the changes of fresh mass (Table 1). For the stems stored at 8 and 12°C, relative water content remained constant. Similar behavior was observed in studies with *Oncidium varicosum* (Mattiuz et al., 2010) and roses (Pietro, Mattiuz, Mattiuz, & Rodrigues, 2012), in which relative water content reductions were noted when plants were stored at room temperature.

A batch of flowers was evaluated separately for longevity. We observed that all buds remained closed for stems stored at 4°C (score 5) until 17 days after the beginning of storage, opening fully only from the 30th day onward. As for leaf color, we noted that stems stored at 4°C remained green up to 39 days of storage. However, after 35 days, a few leaves began to show dark parts, what may be considered a cold disorder symptom since cell-membrane deterioration promotes oxidation, inducing tissue darkening (Jiang, Duan, Joyce, Zhang, & Li, 2004), thereby impairing the decorative life of flowers. Stems stored at 4°C remained turgid until the 25th day of storage (Figure 1A, B, C, and D).

Stems stored at 8°C showed more than 50% of their flower buds closed until 12nd day of storage, which remained turgid until the 39th day. Leaves remained green to the 30th day of storage and anthers intact until the 17th day. At this temperature, flower life was extended until the 46th day of storage, without appearance of chilling injuries.

According to Muniz, Galati, Marques, Mattiuz, and Mattiuz (2016), lower temperatures slow the ageing and deterioration of gerbera flower tissue, as its metabolism decreases while maintaining the quality of flowers for a longer time.

As for respiratory rate, flowers stored at 4 and 8°C presented a reduction from the 9th day of storage, and for those stored at 12°C, such reduction occurred from the 6th day onward. On the 12th day of storage, stems at 22°C showed greater respiratory activity when compared to the other treatments. At the end of storage, respiration was two times lower in stems kept at 4°C compared with those stored at 22°C (Figure 2).

In the specific case of roses, Kuc and Workman (1964) concluded that respiratory rate is six times lower when flowers are kept at 5°C, compared with those stored at 25°C. According to Kerbauy (2004), for most plant tissues, a 10°C increase, within a range of 5 to 25°C, doubles respiratory rate due to increased enzyme activity. Cevallos and Reid (2000) suggested that temperature-dependent changes in respiration have effects on the vase life of daffodil and chrysanthemum cut flowers.

Soluble carbohydrate content in petals of alstroemeria flowers increased during storage period, except for those kept at 4°C, which reduced from the 6th day onward (Figure 3A). Reducing carbohydrate content increased until the 6th day of storage for stems stored at 4, 8, and 12°C, keeping constant thereafter. Conversely, those stored at room temperature (22°C) maintained carbohydrate content unchanged during the entire storage period (Figure 3B).

Reduction in reducing carbohydrate content from the 9th day of storage in stems at 22°C may be related to decreased respiration rates as a function of sugar use as respiration substrate. Nabigol, Naderi, and Kafi (2006) reported that the final stage of flower development is associated with a reduction of carbohydrate content and the lifespan of cut flowers. This fact is related to flower senescence since carbohydrates are used as a breathing substrate, thus depleting over time.

Lower temperatures inhibit enzymatic activities of polyphenol-oxidase (PPO) and peroxidase (POD), as evidenced by the absence of flower darkening during storage. On the other side, storage at room temperature (22°C) increases the activity of such enzymes, resulting in a fast darkening at the end (Figure 4A and B). Peroxidase activity increase in flowers stored at 22°C may have occurred once the darkening or discoloration of tissues come from reactions catalyzed by this enzyme (Laurente & Clemente, 2005).

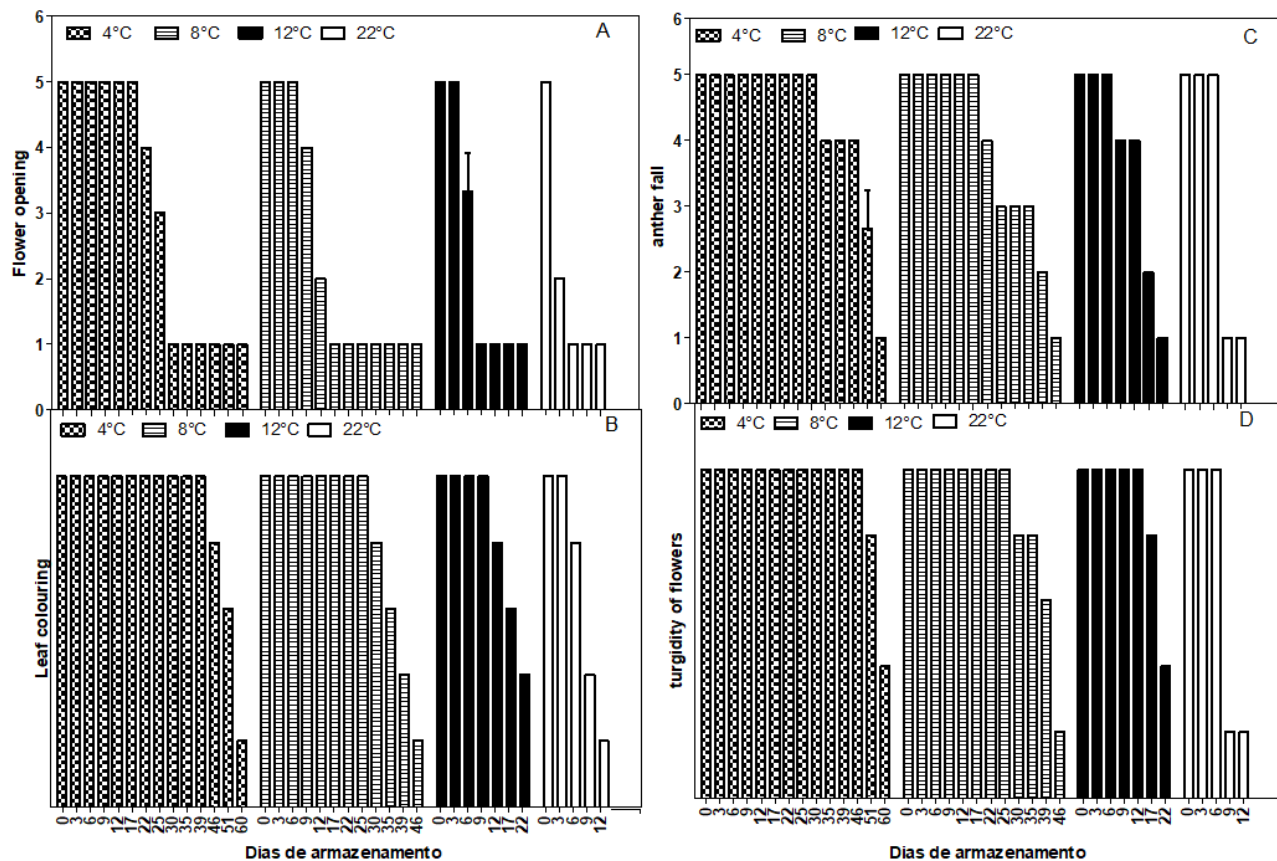


Figure 1. Qualitative characteristics of cut alstroemeria 'Ajax' stored at four temperatures (4, 8, 12, and 22°C). Vertical bars represent the standard error of the mean.

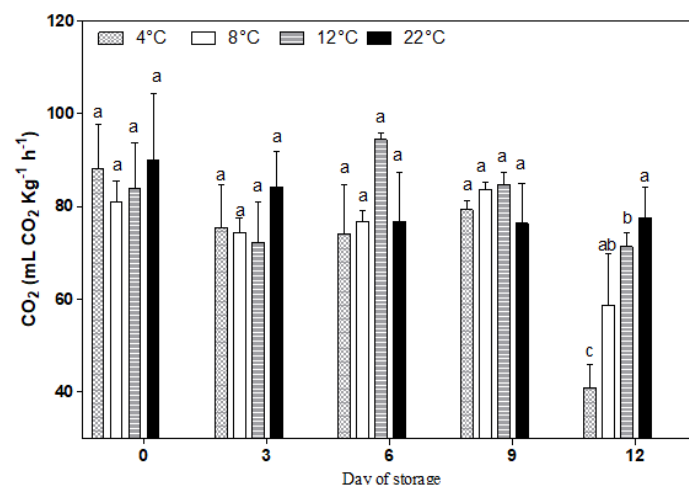


Figure 2. Respiratory rate of cut alstroemeria 'Ajax' stored at four temperatures (4, 8, 12, and 22°C). Vertical bars represent the standard error of the mean. Different letters show significant difference between treatments.

Even though all treatments showed an increase in carotenoids and anthocyanins, stems stored at 22°C had higher contents throughout the storage period (Figure 5A and B). This indicates that high temperatures increase plant metabolism, boosting the synthesis of these pigments. The presence of conjugated double bonds in carotenoid molecules facilitates their oxidation, favoring darkening and intensifying coloration, which is a natural aspect of vegetal senescence (Damodaran, Parkin, & Fennema, 2007).

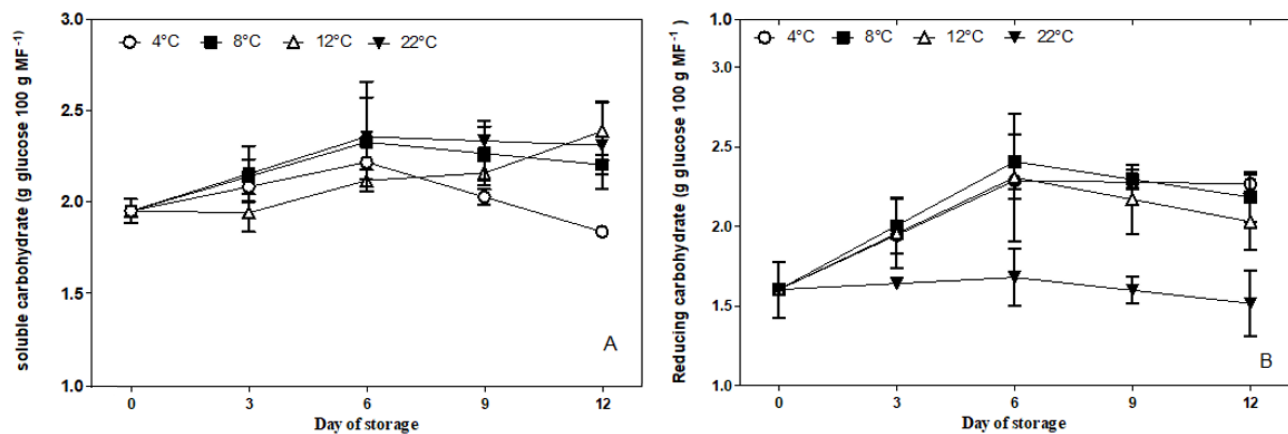


Figure 3. Soluble (A) and reducing (B) carbohydrate contents in cut alstroemeria 'Ajax' stored at four temperatures (4, 8, 12, and 22°C). Vertical bars represent the standard error of the mean.

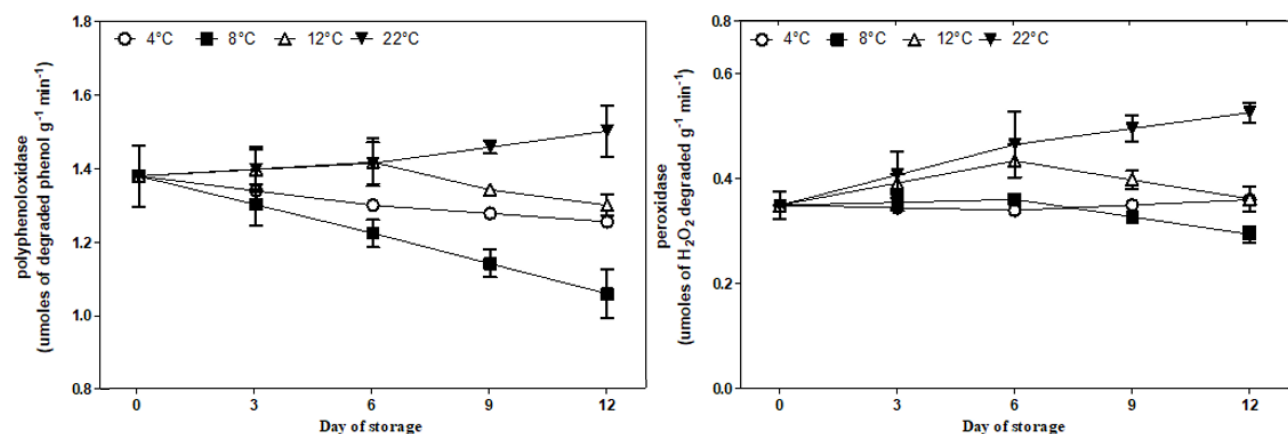


Figure 4. Activities of polyphenol-oxidase (A) and peroxidase (B) enzymes in cut alstroemeria 'Ajax' stored at four temperatures (4, 8, 12, and 22°C). Vertical bars represent the standard error of the mean.

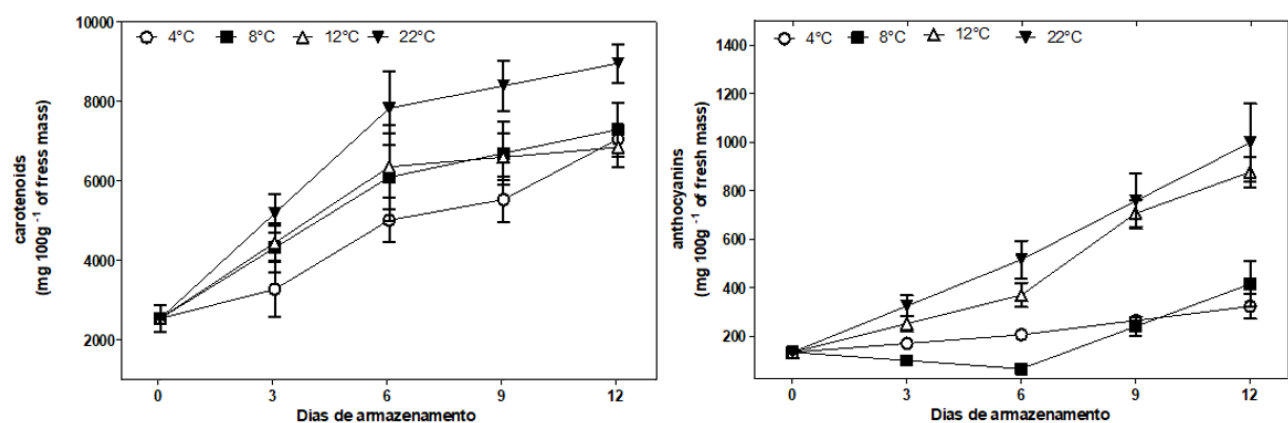


Figure 5. Content of carotenoids (A) and anthocyanins (B) in cut alstroemeria 'Ajax' stored at four temperatures (4, 8, 12, and 22°C). Vertical bars represent the standard error of the mean.

Conclusion

Storage of cut *Alstroemeria* 'Ajax' inflorescences at 8°C maintained their postharvest quality effectively since flowers remained turgid and leaves and stems green for a longer time, extending their longevity until 46 days of storage. Storage at 4°C is not recommended as it causes chilling injury to the leaves.

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