



Use of methyl jasmonate in bird of paradise¹

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ABSTRACT

The MeJA has been shown to be promising in reducing the damages caused by cold and maintaining the quality in several products. Nevertheless, few studies are conducted with flowers and there are no studies of its use in bird of paradise. The objective of this study was to determine whether the pulsing application with MeJA submitted to cold storage promotes physiological changes of the bird of paradise. Flowers were placed in solutions of 200 g L⁻¹ sucrose added with 100, 250 and 400 µmol L⁻¹ MeJA and control without MeJA for 24 h. The stems were transferred to containers with water at 5 °C for 28 days. During storage, the water uptake rate, transpiration and fresh mass were reduced, while the electrolyte leakage and phenolic compounds increased. The application of MeJA did not reduce the loss of fresh mass and electrolyte leakage. The application of 400 µmol L⁻¹ MeJA has increased peroxidase activity since day 14 of storage. It is concluded that the application of MeJA with pulsing under refrigeration does not promote positive physiological changes for the maintenance of the quality in bird of paradise flowers.

Keywords: oxidative enzymes; quality; *Strelitzia reginae*; water absorption

INTRODUCTION

Bird of paradise is highly valued in floral arrangements due to its shape, color and rusticity, however, as all cut flowers have a reduced commercialization period. Conditioning the flowers at low temperatures is an alternative to maintain quality and prolong durability. However, flowers such as orquídea anthurium, bird of paradise, heliconia, etc., cannot be stored at commercial storage and transportation temperatures (0–4 °C) due to the occurrence of cold injury (Darras, 2019).

In bird of paradise, cold injuries are already observed at temperatures below 13°C (Jaroenkit & Paull, 2003), and this susceptibility varies with the genotype, time of exposure to cold and temperature (Pompodakis *et al.*, 2010).

The lesions are variable, however, the discoloration of the petals and the appearance of necrotic spots are common, as observed in heliconia (Costa *et al.*, 2011) and orquídea (Mapeli *et al.*, 2011).

The cause of these injuries is the production of oxygen-reactive species (ROS) in response to stress, promoting the peroxidation of membrane lipids, which can be measured by electrolyte leakage (EL). Additionally, ROS acts as a signal by activating enzymes in the plant's oxidative system (Suzuki & Mittler, 2006), such as peroxidase (POD), in order to overcome the damage caused.

A study with salicylic acid has shown promise in reducing EL and increasing POD activity in bird of paradise (Pereira *et al.*, 2018) and studies with various vegetables with methyl jasmonate (MeJA) have shown that this hormone can be effective in reducing injuries caused by cold. As observed in studies on guava (González-Aguilar *et al.*, 2004), mango (González-Aguilar *et al.*, 2000), lemon (Siboza *et al.*, 2014), tomato (Ding *et al.*, 2001), grape and orange (Grasemnezhad *et al.*, 2008). However, a little study is verified in cut flowers (Darras, 2019), which is a promising and practical treatment for flower producers and wholesalers.

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In addition to reducing chilling, MeJA can improve floral quality, acting positively on senescence, flowering, stoma closure and regulation of floral opening (Avanci *et al.*, 2010) which would have a positive effect on marketing.

The interruption of water absorption with the cut of the flowers associated with the transpiration rate, causes the reduction of the fresh mass of the flowers, leading to wilting which is one of the main quality factors evaluated by the consumer.

The treatment of roses at room temperature with MeJA improved the color and loss of fresh mass flower (Pietro *et al.*, 2012) and this hormone reduced the incidence of Mofo cinzento (*Botrytis cinerea*) in cut peony flowers (Gast, 2001). The incidence of the disease is one of the reasons for post-harvest losses.

Therefore, the objective of this study was to determine whether the pulsing application with MeJA submitted to cold storage promotes physiological changes of the bird of paradise.

MATERIAL AND METHODS

The flower stems were harvested in the morning at the commercial harvest point with an open floret. Then the stems were standardized to 25 cm and placed in pulsing solutions with 200 g L⁻¹ sucrose added with concentrations of 100, 250 and 400 µmol L⁻¹ MeJA (Methyl Jasmonate, Sigma-Aldrich, 95%), and controlled only with sucrose for 24 h. They were then transferred to containers containing water and stored at 5 °C. The water was exchanged daily. The temperature was determined by datalogger instrument (HTR-157, Instrutherm). The analyzes of water uptake rate, transpiration rate, fresh mass, electrolyte leakage, peroxidase activity (POD), phenolic compounds and floral opening were performed weekly until the appearance of necrotic spots in the sepals, petals and ends of the bracts, which was at 28 days, making the product unsuitable for marketing.

Water uptake and transpiration rates were determined according to the methodology described by Van Doorn *et al.* (2002). The stems were laid in individual tubes previously weighed and contained 200 g of deionized water. The tubes were weighed on days 0 and 7 with and without the stems. To reduce the effects of evaporation, the upper end of the tubes were wrapped with four layers of PVC film. Water uptake and transpiration rate were obtained in mg g⁻¹ of fresh mass day⁻¹. Water uptake was calculated using the equation: $V = (MSi - MSf) / MHf$. In addition, the estimated transpiration rate was calculated using the equation: $T = V - (MHf - MHi)$, where V is absorbed solution volume, MSi is mass of solution on day 0, MSf is mass of solution on day 7, MHf is mass of stems on day 7, T is transpiration rate, and MHi is mass of stems on day 0.

The fresh mass was determined by the weight of the floral stems on each evaluation day and expressed as a percentage.

Electrolyte leakage (EL) was determined with a conductivity meter (LFT 613T, Schott Geratie). Four 10-mm diameter disks were removed from the bracts, soaked in 20 mL of distilled water in a closed container for 6 h, to assess first the free conductivity (FC) reading. Afterward, the containers were placed capped in an oven at 90 °C for 2 h. After cooling, total conductivity (TC) readings were performed, where $EL = (FC/TC) \times 100$ (Lima *et al.*, 2002).

Peroxidase (POD) was determined according to Lagrimini *et al.* (1997), with modifications. In the extraction, 5 g of bracts and sepals were ground with 25 mL of extraction buffer (0.1 mol L⁻¹ phosphate buffer, pH 6.0, added with 0.1% sodium bisulfite and 0.15 mol L⁻¹ of sodium chloride). The homogenate was filtered and centrifuged at 17000 g for 30 min at 4 °C. In the enzymatic activity assay, an aliquot of the filtered homogenate was added to the reaction medium containing 0.5 mL of guaiacol (1.7%), 1.5 mL of extraction buffer (pH 6.0) and 0.5 mL of hydrogen peroxide (1.8%). Readings were performed using a spectrophotometer at 470 nm and enzymatic activity expressed in µmol min⁻¹ mg⁻¹ of protein. The same filtered homogenate used in the enzymatic assay was used for protein quantification by the Bradford (1976), using bovine serum albumin (BSA) as standard.

The phenolic compounds were determined according to Folin-Denis, described by Kubota (1995), with modifications. Fifty grams of bracts and sepals were macerated with 10 mL of methanol. The homogenate was centrifuged, a 0.5 mL aliquot was mixed with 2.5 mL of 1:3 Folin-Denis reagent and two mL of 10% NaCO₃. After one h in the dark, the samples had their absorbance read in a spectrophotometer at 700 nm, using D-catechin as standard.

The experiment was set up in a randomized complete block design, in a split plot scheme. The plots were the doses and the subplots, the days of evaluations. The experiment was composed of four blocks and the experimental unit consisting of two flower stems. The data were submitted to analysis of variance by the System of Statistical Analysis and Genetics of UFV - GENES-UFV (Cruz, 2006) and expressed in response surface.

RESULTS AND DISCUSSION

Water uptake rate, transpiration and fresh mass were reduced with storage time (Figure 1A, 1B and 2). The reduction in water uptake during storage is associated with blockage of conducting vessels (Van Meeteren *et al.*, 2001), which occurs physiologically in the stems of the bird of paradise, involving the activity of the enzymes POD and PPO, resulting in deposition of lignin, obstruc-

ting the pores that connect the xylem vessels (Marques *et al.*, 2011). Despite the reduction of respiration, the restrictions of water absorption minimize the fresh mass 40% from the 14 days of storage. According to Nowak & Rudnicki (1990) a variation of 10 to 15% of fresh mass compromises the quality and durability of the flowers and can result in wilt symptoms.

Greater water uptake and transpiration were observed in flowers treated with 250 and 400 $\mu\text{mol L}^{-1}$ MeJA (Figure 1A; Figure 1B). However, the application of MeJA did not reduce the fresh mass loss of bird of paradise because the increase of transpiration was compensated by elevation of water uptake (Figure 2). In work with rose cv. Vagas in ambient temperature, the application of MeJA had a positive effect water uptake what occasioned less loss of fresh mass (Pietro *et al.*, 2012).

Electrolyte leakage increased with storage time (Figure 3A). The extended exposure to cold caused rupture of the cell membranes, therefore resulting in the leak of ions and metabolites, which can be monitored by determination of electrolyte leakage. Membrane damage occurs due to the generation of ROS at levels above the removal capacity by the antioxidant mechanisms present in the tissues. Lipid molecules pass from the gel state to the crystalline gel state, as the primary response of the cold sensitive tissues. The application of MeJA was not effective in reducing the electrolytes leakage, being not effective in reducing the cold injury in floral stems of bird of paradise.

There was an increase in electrolyte leakage from 14th day with the application of 250 $\mu\text{mol L}^{-1}$, followed by a small reduction with the dosage of 400 $\mu\text{mol L}^{-1}$ (Figure 3 A). While for the POD activity, the increase occurred from the 14th day onwards only at a dosage of 400 $\mu\text{mol L}^{-1}$ (Figure 3 B). These results indicate that higher dosages of MeJA may have led to increased

ROS, which resulted in increased POD activity. In a study with mamona (*Ricinus communis*), it was verified that MeJA induced the accumulation of hydrogen peroxide shortly after its application (Soares *et al.*, 2010). These data reinforce the idea that MeJA may be related to alterations in oxidative stress, altering the activities of some key enzymes that control this process and promoting the accumulation of ROS in the early stages of stress (Soares *et al.*, 2010).

The POD is a low-temperature-stimulated stress enzyme in species that are sensitive to cold (El-Hilari *et al.*, 2003) to reduce damage caused by ROS, such as hydrogen peroxide. The increase in POD activity under temperature stress conditions was also observed in 'Navelina' orange stored at 10 °C and 'Fortune' tangerine at 4 and 8 °C (El-Hilari *et al.*, 2003).

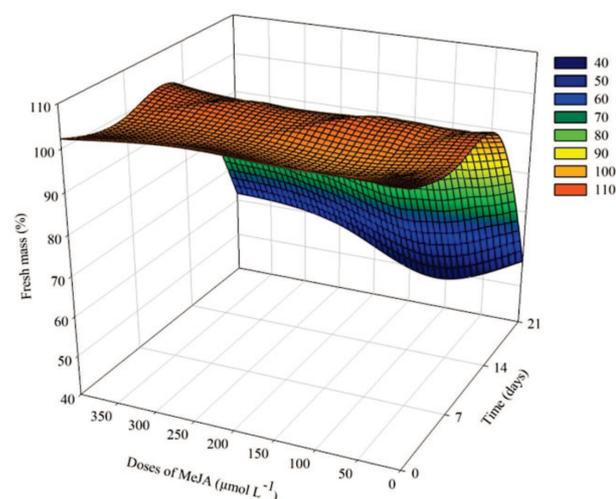


Figure 2: Fresh mass of bird of paradise cut flowers submitted to MeJA (100, 250 and 400 $\mu\text{mol L}^{-1}$) and control (without MeJA) storage during 0, 7, 14 and 21 days at 5 °C.

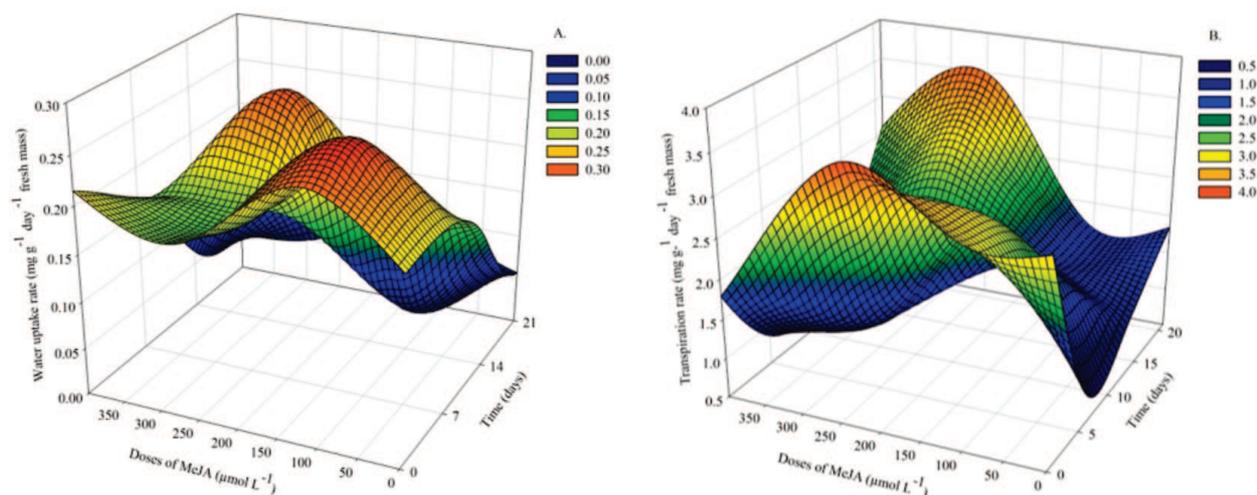


Figure 1: Water uptake rate (A) and transpiration rate (B) bird of paradise cut flowers submitted to MeJA (100, 250 and 400 $\mu\text{mol L}^{-1}$) and control (without MeJA) storage during 0, 7, 14 and 21 days at 5 °C.

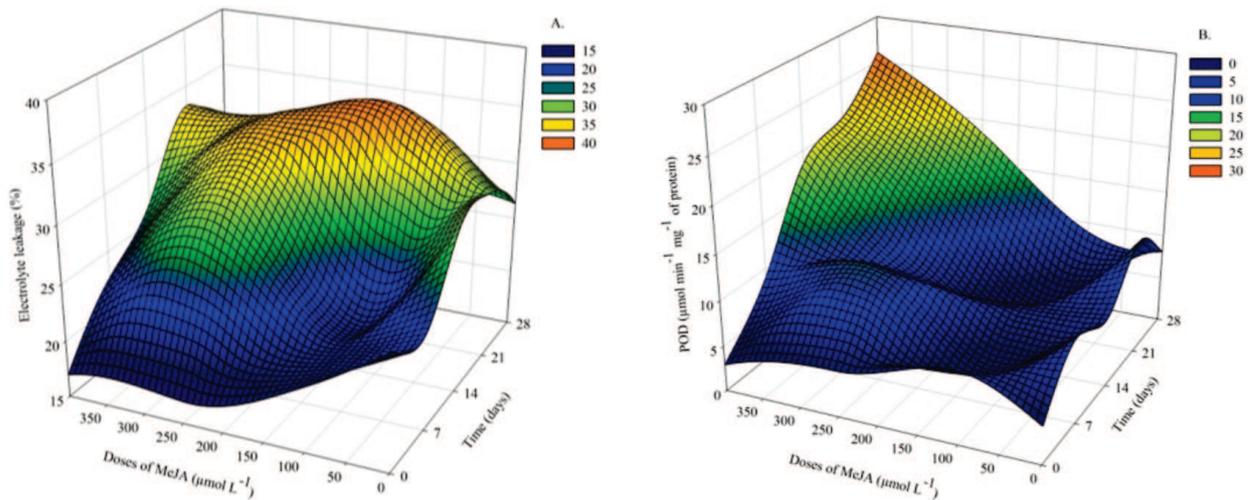


Figure 3: Electrolyte leakage (A) and peroxidase activity (POD) of bird of paradise cut flowers submitted to MeJA (100, 250 and 400 $\mu\text{mol L}^{-1}$) and control (without MeJA) storage during 0, 7, 14 and 21 days at 5 °C.

These results indicate that the application of MeJA was not effective in reducing the damage caused by the cold in flower stems of bird of paradise.

The phenolic compounds increased with the storage time, for the control and dose treatments of 100 $\mu\text{mol L}^{-1}$ of MeJA (Figure 4), however, there was no increase in the phenolic compounds over the storage time, for the highest doses, indicating the effectiveness of MeJA in reducing phenols. Thus, greater darkening in the control flowers and treated with 100 $\mu\text{mol L}^{-1}$ of MeJA would be expected, which was not verified. At 28 days, all treatments showed dark spots on the bracts and sepals.

In orange and tangerine fruits, no relationship was observed between darkening and peroxidase activity (El-Hilari *et al.*, 2003). This was also not verified in this work with bird of paradise. However, in peaches stored for three weeks at 5 °C followed by 3-day exposure at 20 °C, a higher

POD activity and lower phenolic content were observed with MeJA application in relation to the control (without MeJA) (Meng *et al.*, 2009).

CONCLUSION

The application of MeJA with pulsing under refrigeration does not promote positive physiological changes for the maintenance of the quality in bird of paradise flowers.

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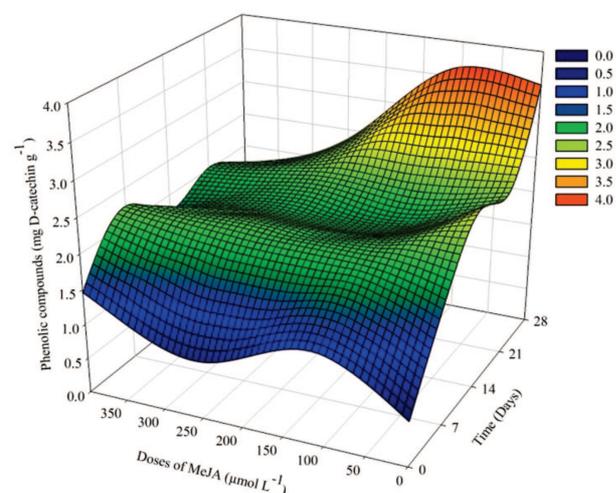


Figure 4: Phenolic compounds of bird of paradise cut flowers submitted to MeJA (100, 250 and 400 $\mu\text{mol L}^{-1}$) and control (without MeJA) storage during 0, 7, 14 and 21 days at 5 °C.

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