

## Original Research Article

### PP packaged 10 days cold stored rose cut flowers cv. Bordeaux influenced by different pulsing solution

#### Abstract

The study included different pulsing solution *viz.*, water, HQC 200mg/l, sucrose 50g/l + HQC 200 mg/l, citric acid 200mg/l,  $\alpha$ -lipoic acid 200mg/l, GA<sub>3</sub> 100 mg/l and BA 100mg/l to study the effect on flower quality and vase life of PP (polypropylene) packaged cold storage (at 2°C) rose cv. Bordeaux, for the period of 10 days. Pre-storage pulsing of GA<sub>3</sub> 100mg/l solution treatment showed highly promising results in extending flowers vase life along with maintained flower quality just after storage and at 4<sup>th</sup> DAS in vase life. Further, GA<sub>3</sub> 100mg/l solution treatment recorded significantly higher water up take (175.65 ml), change in fresh weight (10.50%) at 4<sup>th</sup> DAS, total dissolve solids (8.85 °brix, 8.05 °brix and 6.75 °brix) and membrane stability index (81.97 %, 65.97 % and 37.88 %) at 2<sup>nd</sup>, 4<sup>th</sup> and 6<sup>th</sup> DAS. Moreover, from table 2, it was evident that same treatment recorded significantly higher total soluble sugar (59.65 mg/g fresh weight), Estimated protein (18.79 mg/g fresh weight), catalase activity (20.42  $\mu$  mol H<sub>2</sub>O<sub>2</sub> min<sup>-1</sup> g<sup>-1</sup> protein), peroxidation activity (40.30  $\mu$  mol H<sub>2</sub>O<sub>2</sub> min<sup>-1</sup> g<sup>-1</sup> protein), pigment content (15.82 mg/g fresh weight) anthocyanin content in the petals at 4<sup>th</sup> DAS. Whereas, GA<sub>3</sub> 100 mg/l pulsing solution treated PP packaged cold stored rose cut flowers cv. Bordeaux recorded significantly higher vase life (6.88 days) which was at par with treatment P<sub>2</sub> (Sucrose 50 g/l + HQC 200 mg/l).

Key words :Rose, Polypropylene, low temperature storage, Pulsing, GA<sub>3</sub>, sucrose

#### Introduction

Roses are the most popular cut flowers around the world. Increasing popularity and demand in the world trade rose has become highly lucrative business amongst the flower growers. However, it is important to understand that the harvesting stages, postharvest handling and marketing techniques significantly affect the quality and the longevity of the flowers. It is very important to establish a well-planned post-harvest handling protocol to have best quality flowers in the market when it is high in demand. Makwana *et al.* (2024a) and Makwana *et al.* (2015a) observed that PP (Polypropylene) packaging along with cold storage at 2°C temperature provide better result comparing to different wet storage technique which is been used by most of the conventional flower growers and traders. Modified atmosphere packaging along with cold storage is known to create modified internal gaseous components passively (Farber *et al.*, 2003) that minimizing metabolic activities during storage period and retain good flower quality after storage of 10 days (Makwana *et al.*, 2024b). Similar results with seal packaging of cut flowers with polyfilms at low temperature been found beneficial in retaining flower quality, improving opening ability, reducing water loss during post storage phase in stored flowers like rose (Singh *et al.*, 2012 and Makwana *et al.*, 2015b) gladiolus (Grover *et al.*, 2005, Singh *et al.*, 2007), Solidago Canadensis (Zeltzer *et al.*, 2001) and in Lisianthus (Akbulak *et al.*, 2005). Makwana *et al.* (2024b) stated that higher CO<sub>2</sub> and lower O<sub>2</sub> during storage period decrease the production of ethylene and extended the life of fresh produce rose. Such beneficial conditions in orchid flowers showed promising results and

maintained flower quality after completing of storage duration (Poonsri2017 and Poonsri2021).

Plant growth regulators plays and important role in quality of flower and extending storage life of rose cut flowers (Sharma, 2023). Pulsing is a technique is very important for extending post-harvest life of cut flowers. Pulsing treatments comprising of germicides and sugar are mostly used to improve flower diameter, flower size, shape, colour and life of cut flowers (Singh *et al.*, 2007 and Vidhya Sankar and Bhattacharjee, 2002). The 8-HQ has been known to possess strong anti-microbial properties that eliminate vascular blockage and enhance water up take in flowers (Burdett, 1970 and Makwana *et al.* 2015a). Rose cut flowers kept in pulsing of GA<sub>3</sub> solution showed promising results from harvest to senescence (Bhattacharjee, 2000 and Makwana *et al.*, 2024c). Hence this experiment was laid out to evaluate best pulsing solution treatment to the 10 days cold stored rose cut flowers packed with PP material to get the better quality and post-harvest life of rose cut flowers cv. Bordeaux.

### **Material and methods**

Freshly harvest rose cut flowers of cv. Bordeaux were moved from greenhouse and brought to the Floriculture Laboratory, ASPEE College of Horticulture and Forestry, NAU Navsari at an ambient room temperature (18-21°C). The experiment was laid down in completely randomized block design. Total six Pulsing treatments and a control were taken under study along with each treatment replicated three times. Cut flowers of cv. Bordeaux having uniform bud size, shape and fresh weight (10±2 g) and stem length (50±5 cm) were selected and divided in to seven groups each having 30 flowers (10 flowers in each replication) and are provided with different pulsing solution treatment *viz.*, P<sub>1</sub> - HQC 200 mg/l, P<sub>2</sub> - Sucrose 50 g/l + HQC 200 mg/l, P<sub>3</sub> - citric acid 200 mg/l, P<sub>4</sub> - α lipoic acid 200 mg/l, P<sub>5</sub> - GA<sub>3</sub> 100 mg/l, P<sub>6</sub> - BA 100 mg/l and P<sub>0</sub> - water for 3 hour before storage. After pulsing treatment all the bunches were packed with PP packaging uniformly and moved to 2°C cold storage for the duration of 10 days. On completion of 10 day of storage duration, all the treated bunches were shifted from cold storage to laboratory and packaging were removed and stems were re cut 2 cm from the basal end and kept in distilled water at room temperature. The experiment was conducted for two times for two consecutive years represented as Year 1 and Year 2 for taking observations and recording data.

Data were collected regarding different postharvest parameters effecting quality of flowers at different intervals during vase life. Observations on post-harvest parameters like total water uptake (ml), change in fresh weight (%) at 4<sup>th</sup> DAS, total dissolve solids (TDS) at 2<sup>nd</sup>, 4<sup>th</sup> and 6<sup>th</sup> DAS, membrane stability index (MSI) at 2<sup>nd</sup>, 4<sup>th</sup> and 6<sup>th</sup> DAS, total soluble sugar (TSS), Estimated protein (mg/g fresh weight), catalase activity (μ mol H<sub>2</sub>O<sub>2</sub> min<sup>-1</sup> g<sup>-1</sup> protein), peroxidaiton activity (μ mol H<sub>2</sub>O<sub>2</sub> min<sup>-1</sup> g<sup>-1</sup> protein), pigment content (anthocyanin in the petals) were recorded 4<sup>th</sup> day after storage during vase life. TDS was measured with the help of digital refractometer in °Brix. Other parameters were recorded followed by methods listed below.

### **Membrane Stability Index (MSI) in petals**

MSI was calculated on the basis of electrolyte leakage (ion leakage) of petals. The electrolyte leakage was measured by taking five petal discs (1 cm<sup>2</sup>) of flower from the centre of the bud at 4<sup>th</sup> DAT. The petal discs were rinsed well in deionized water prior to incubation in 5 ml of deionized water for 3 h at room temperature. After incubation, conductivity of the bathing solution was measured with the conductivity meter (value A). The petal discs were boiled with bathing solution for 10 minutes to kill tissue. After cooling to room temperature, the conductivity of bathing solution was again measured (value B).

Electrolyte Leakage (%) = (value A/ value B) X 100

MSI (%) = [1-(value A/ value B)] X 100

#### **Total Soluble Sugar (TSS) (mg/g fresh weight)**

Petals (100 mg) were extracted with 5 ml of 80% ethanol and centrifuged at 3000 rpm for 10 minutes. Extraction was repeated 4 times with 80% ethanol and supernatants were collected into 25 ml volumetric flasks. Final volume of the extract was made to 25 ml with 80 % ethanol. The extract (0.3 ml) was pipetted from treatments into separate test tubes and the tubes were placed in a boiling water bath for 3 minutes to evaporate the ethanol. One ml of millipore water and 4 ml of 0.2% anthrone reagent (200 mg in 100 ml H<sub>2</sub>SO<sub>4</sub>) was added in each test tube and placed in ice cold water. Reagent blank was prepared by adding 1 ml of distilled water and 4 ml of anthrone reagent. The intensity of colour was read at 600 nm on spectrophotometer. A standard curve was prepared using 10 mg glucose per 100 ml distilled water (Franscisttet *et al.*, 1971).

Total soluble sugar (mg/g) = Sample O.D. x Standard O.D. x Dilution factor

#### **Estimation of protein**

Protein concentration of each enzyme extract and seed protein was estimated by method of Lowry *et al.* (1951).

Reagents :

- (i) Solution A : 2% Na<sub>2</sub>CO<sub>3</sub> in 0.1 N NaOH
- (ii) Solution B : (a) 1% CuSO<sub>4</sub>.5H<sub>2</sub>O solution  
(b) 2% sodium potassium tartrate solution

Working solution of B : Prepared fresh before use by mixing equal volume of solution B(a) and B(b).

(iii) Solution C : Prepared fresh before use by mixing 50 ml of solution A and 1 ml of working solution of B.

(iv) Solution D : Folin & Ciocalteu reagent (1N) reagent.

Enzymes extract/protein extract (25 µl) were taken in test tube and volume was made up to 1 ml with millipore water. A tube with 1 ml of water served as blank. Five ml of solution C was mixed by vortexing and kept for 10 min. Then 0.5 ml of solution D (Folin & Ciocalteu reagent) was mixed with vortex and kept it room temperature for 30 min. Absorbance was read at 660 nm. A standard curve was prepared with bovine serum albumin in the range of 10-80 µg.

#### **Catalase Activity (CAT)**

Total catalase (EC 1.11.1.6) activity was determined in the homogenates by measuring the decrease in absorption in 3ml mixture at 240nm as H<sub>2</sub>O<sub>2</sub> (ε = 39.4 mM<sup>-1</sup> cm<sup>-1</sup>) was consumed according the method of Aebi (1984) and enzyme activity expressed as µmol

H<sub>2</sub>O<sub>2</sub> oxidized min-1 g-1 protein. The 3 ml mixture containing 50mM sodium phosphate buffer (pH 7.0), 10mM H<sub>2</sub>O<sub>2</sub> and 50µl enzyme extract.

### **Peroxidase (POD) Activity**

POD (EC 1.11.1.7) activity was determined in the homogenates by measuring the increase in absorption at 470nm due to the formation of tetraguaiacol ( $\epsilon = 26.6 \text{ mM}^{-1}\text{cm}^{-1}$ ) in a reaction mixture containing 50mM sodium phosphate buffer pH 7.0, 0.1mM EDTA, 0.1ml enzyme extract, 10mM guaiacol and 10mM H<sub>2</sub>O<sub>2</sub> (Costa *et. al.*, 2002).

Enzyme unit of CAT and POD was calculated as :

$$\text{mM/min/g protein} = \frac{\Delta \text{O.D.}}{\text{Enzyme Conc. (g)} \times \epsilon}$$

Where,

$\epsilon$  = Extinction coefficient

Enzyme conc.(g) = Amount of enzyme in 3 ml reaction mixture

All the data recorded for various observations during the course of investigation were statistically analyzed (Panse and Sukhatme, 1978).

### **Result and discussion**

Pulsing treatment significantly influence Physiological loss in weight, change in fresh weight, water uptake, bud diameter, per cent bud opening, MSI, TDS, dry weight and vase life of the flowers (Table 1). Among various pulsing solutions treated PP packed 10 days cold stored (2°C) rose cut spikes cv. Bordeaux under observations cut flowers treated with GA<sub>3</sub> 100 mg/l solution recorded significantly higher water up take (175.65 ml), change in fresh weight (10.50%) at 4<sup>th</sup> DAS, total dissolve solids (8.85 °brix, 8.05 °brix and 6.75 °brix) and membrane stability index (81.97 %, 65.97 % and 37.88 %) at 2<sup>nd</sup>, 4<sup>th</sup> and 6<sup>th</sup> DAS. Moreover, from table 2, it was evident that same treatment recoded significantly highertotal soluble sugar (59.65 mg/g fresh weight), Estimated protein (18.79 mg/g fresh weight), catalase activity (20.42 µ mol H<sub>2</sub>O<sub>2</sub> min<sup>-1</sup> g<sup>-1</sup> protein), peroxidaiton activity (40.30 µ mol H<sub>2</sub>O<sub>2</sub> min<sup>-1</sup> g<sup>-1</sup> protein), pigment content (15.82 mg/g fresh weight) anthocyanin content in the petals at 4<sup>th</sup> DAS. Whereas, GA<sub>3</sub> 100 mg/l pulsing solution treated PP packaged cold stored rose cut flowers cv. Bordeaux recorded significantly higher vase life (6.88 days) which was at par with treatment P<sub>2</sub> (Sucrose 50 g/l + HQC 200 mg/l).

GA<sub>3</sub> enhance the liposomal permeability of the cell membrane to glucose (Wood and Paleg, 1972), hydrolyse starch, fructans and sucrose into glucose and fructose molecules (Salisbury and Ross, 1992). This might have facilitated the higher intake of the sugar in the cell, which further enhanced water uptake due to osmotic pull (Ho & Nichols, 1997). It also known to be involved in mobilization of stored food (Srivastava, 2005) and further, to increase water uptake and retention of fresh weight (van Doorn, 2004). Further antioxidant property of GA<sub>3</sub> contributed in stabilized cell membrane structure and thus reduced electrolyte leakage in the petal tissue as also reported in gladiolus (Dantuluri *et al.*, 2008), rose (Sabehat and Zeislin, 1994), in chrysanthemum (Elanchezhian and Srivastava, 2001). Further, higher respiratory substrate and increased level of sugar in petal (TSS status, Table 2) facilitated higher rate of respiration necessary of cell division and cell enlargement (Ho

and Nichols, 1977) leading to high TDS as well as high MSI (Makwana *et al.*, 2024a). The enhanced vase life of rose cut flowers pulsed with GA<sub>3</sub> 100 mg/l solution can be attributed to continued and increased water uptake, higher retention of fresh weight and also high maintained sugar level in petal tissue (high TSS), higher retained catalase and peroxidase activity and protein content resulted in delaying electrolytic leakage contributed to optimum continuation of the cell metabolism that facilitated cell growth and development, formation of cellular constituents and the liberation of energy for other cellular functions. Similar effects of enhanced postharvest vase life with GA<sub>3</sub> in rose (Makwana *et al.* 2024a and Makwana *et al.*, 2015a) and gladiolus (Dantuluri *et al.*, 2008; Singh *et al.*, 2008) and delayed leaf and petal abscission in rose (Van Doorn *et al.*, 1994 and Ahmadi and Hassani 2015) has been reported earlier.

### **Conclusion:**

In conclusion, GA<sub>3</sub> 100 mg/l solution as pulsing treatment had a significant effect on increasing vase life of PP packed rose cut flowers cv. Bordeaux cold stored (2°C) for 10 days and improving their quality of flowers by increase in retention of fresh weight, higher total water uptake, total soluble sugar, estimated protein, catalase activity and peroxidase activity and therefore improving flowers quality.

### **Consent**

As per international standards or university standards, respondents' written consent has been collected and preserved by the author(s).

### **Ethical Approval**

All experimental protocols including the involvement of human participants were approved and followed the guidelines of the Department of floriculture and landscaping of Aspee College of Horticulture of the Navsari Agricultural University, Navsari.

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UNDER PEER REVIEW

**Table 1: Effect of different pulsing solution on total water uptake (ml), Change in fresh weight (%) at 4<sup>th</sup> DAS, Total dissolve solids (°brix) and membrane stability index (%) in PP packaged 10 days cold stored rose cut flowers cv. Bordeaux at 2<sup>nd</sup>, 4<sup>th</sup> and 6<sup>th</sup> DAS.**

Treatment	Total water uptake (ml)	Change in fresh weight (%) at 4 <sup>th</sup> DAS	Total dissolve solids (°brix)			Membrane stability index (%)		
			2 <sup>nd</sup> DAS	4 <sup>th</sup> DAS	6 <sup>th</sup> DAS	2 <sup>nd</sup> DAS	4 <sup>th</sup> DAS	6 <sup>th</sup> DAS
P <sub>0</sub> (Water)	159.36	9.51	8.65	7.79	6.50	78.16	62.19	34.71
P <sub>1</sub> (HQC 200 mg/l)	161.45	9.63	8.68	7.88	6.58	78.63	62.60	34.58
P <sub>2</sub> (Sucrose 50 g/l + HQC 200 mg/l)	166.72	9.95	8.74	7.95	6.65	80.07	64.43	36.31
P <sub>3</sub> (Citric acid 200 mg/l)	162.62	9.76	8.69	7.89	6.59	78.34	62.28	34.23
P <sub>4</sub> ( $\alpha$ -lipoic acid 200 mg/l)	165.02	9.75	8.70	7.90	6.60	78.37	62.31	34.31
P <sub>5</sub> (GA <sub>3</sub> 100 mg/l)	175.65	10.50	8.85	8.05	6.75	81.97	65.97	37.88
P <sub>6</sub> (BA 100 mg/l)	166.06	9.73	8.73	7.93	6.63	79.47	63.93	35.82
CD at 5 %	2.23	0.22	0.07	0.08	0.07	1.29	1.18	0.81

**Table 2: Effect of different pulsing solution on Total soluble sugar (mg/g fresh weight), estimated protein (mg/g fresh weight), Catalase activity ( $\mu$  mol H<sub>2</sub>O<sub>2</sub> min<sup>-1</sup> g<sup>-1</sup> protein), Peroxidation activity ( $\mu$  mol H<sub>2</sub>O<sub>2</sub> min<sup>-1</sup> g<sup>-1</sup> protein), Anthocyanin content (mg/g fresh weight) and vase life in PP packaged 10 days cold stored rose cut flowers cv. Bordeaux at 4<sup>th</sup> DAS.**

Treatment	Total soluble sugar (mg/g)	Estimated Protein (mg/g)	Catalase activity ( $\mu$ mol H <sub>2</sub> O <sub>2</sub> min <sup>-1</sup> g <sup>-1</sup> protein)	Peroxidation activity ( $\mu$ mol H <sub>2</sub> O <sub>2</sub> min <sup>-1</sup> g <sup>-1</sup> protein)	Anthocyanin content (mg/g)	Vase life
P <sub>0</sub> (Water)	54.07	17.70	18.82	36.88	14.14	5.28
P <sub>1</sub> (HQC 200 mg/l)	55.20	17.56	18.85	37.09	14.39	6.30
P <sub>2</sub> (Sucrose 50 g/l + HQC 200 mg/l)	56.44	18.12	19.97	39.14	15.71	6.79
P <sub>3</sub> (Citric acid 200 mg/l)	54.85	17.64	19.18	36.37	14.87	6.21
P <sub>4</sub> ( $\alpha$ -lipoic acid 200 mg/l)	55.14	17.71	17.05	37.07	14.71	6.23
P <sub>5</sub> (GA <sub>3</sub> 100 mg/l)	59.65	18.49	20.42	40.30	15.82	6.88
P <sub>6</sub> (BA 100 mg/l)	55.55	17.71	19.41	38.63	15.15	6.45
CD at 5 %	1.30	0.34	0.47	1.20	0.46	0.12