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Journal of Ornamental Plants
Available online on: www.jornamental.iaurasht.ac.ir
ISSN (Print): 2251-6433 ISSN (Online): 2251-6441

Effect of Pulsing and Wet Cold Storage on the Quality and Vase Life of Cut Gladiolus (*Gladiolus grandiflorus* L.) 'Fado'

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Received: 24 October 2018 Accepted: 31 January 2019
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Abstract

Floriculture is among Kenya's top foreign exchange earners. *Gladiolus* is one of the four famous cut flowers in the world whose popularity in Kenya is attributed to its heat tolerance, many spike forms and color combinations. However, the perishable nature of the flower renders it vulnerable to huge post-harvest losses. The study aimed to evaluate pulsing effect of cut *Gladiolus grandiflorus* 'Fado' with 600 ppm 8-hydroxyquinoline sulfate plus 5 % sucrose prior to wet cold storage duration (0–5 days) on quality and vase life against the control (distilled water). The *Gladiolus* were grown in the open field from corms at the Horticulture Research and Teaching Field, in the Department of Crops, Horticulture and Soils, Egerton University, Kenya, during two successive seasons. A two by six factorial experiment embedded in a completely randomized design with four replicates was adopted. Pro GLM model in two way ANOVA was used to determine differences using Tukey's test at 5 % level of significance. Pulsing treatment had significant effects on the *Gladiolus* quality parameters including: Fresh weight ($P = 0.0031$; 82.214 ± 0.7934 g) as compared to the control; dry weight ($P = 0.0272$); interactive effect of the pulsing and cold storage duration treatments ($P = 0.0004$); maximum vase life (11.5 ± 0.287 days) and opened florets (11 ± 0.15). The highest number of unopened buds (5.18 ± 0.212) were recorded in the control which also had least mean water uptake (23.87 ± 0.26 ml) as compared with the pulsed and cold stored spikes (31.98 ± 0.193 ml). Pulsed cold stored *Gladiolus* preserved quality up to 4 days. The knowledge gained from this study will augment existing technologies in improving quality and market value of this cut flower.

Keywords: Duration, Losses, Sucrose, 8-hydroxyquinoline sulfate.

INTRODUCTION

Gladiolus is a popular cut flower known as queen of the bulbous plants in terms of its market value (Shaukat *et al.*, 2013). In Kenya, it has the potential for production and marketing by small scale growers (Wambani *et al.*, 2003). The fact that *Gladiolus* has many spike forms, colors and color combination attributes makes it a favorite by both florists and consumers (Wambani *et al.*, 2003). All the modern cultivars of gladiolus are of complex hybrid origin raised from the sexual wild South African species and their morphology differs from one genotype to the other (Mar *et al.*, 2011; Parveen, 2017). *Gladiolus* is an ethylene insensitive flower whose exogenous ethylene and ethylene inhibitors have no effect on the petal senescence process (Reid and Jiang, 2012). But ethylene affects the rate and quality of unopened florets on the spikes. *Gladiolus grandiflorus* 'Fado' is one of the exotic cultivars evaluated for suitability for commercialization (Saleem *et al.*, 2013).

The demand for Kenyan cut flowers has increased in both the domestic and international markets over the last two decades (Riisgaard, 2009). This is due to a change in lifestyle among Kenya where flowers are sort for during occasions such as weddings, birthday parties, religious festivals and office decorations (Kargbo *et al.*, 2010). Improved infrastructure and favorable climate in the country are some of the factors that have fostered the growth of floriculture (Mekonnen *et al.*, 2012). Floriculture crops not only have higher profit margins in terms of sales but they take a relatively shorter time to mature compared to conventional crops. According to Horticultural Crop Directorate, floriculture the industry earned Kenya shillings 82.25 billion, which translated into 1.1 % of the national GDP (Melese, 2018). To be able to meet the global competitiveness in the flower industry, Keny farmers have adopted a number of technologies. These include drip irrigation, fertigation systems, greenhouse ventilation systems, net shading, pre-cooling, cold storage facilities, and refrigerated trucks (Asia, 2016).

Pulsing is a short term treatment given to cut flowers whose effect lasts the entire shelf life of the flower, even when they are put in water (Asghari *et al.*, 2014). Different pulsing solutions have been used including dimethyl sulphoxide (2%), 8-hydroxyquinolene sulphate, silver thiosulphate, quaternary ammonium salts, slow release chlorine, aluminum sulphate, citric acid, nitric oxide and 8-hydroxyquinoline citrate among others (Lazar *et al.*, 2010). The term 'pulsing' refers to placing freshly harvested flowers for duration of time (ranging from 12 hours to 24 hours) in a solution formulated to extend their vase life. The pretreatment solutions contain sucrose which allows the flower to continue its normal metabolism, a biocide, a weak acid and an anti- ethylene agent and some may also have plant growth regulators. It has been proved that some pulsing solutions significantly increased vase life and florets opening percentage, decreased contamination in vase solution and improved water balance for cut flower spikes and also maintained flower quality i.e. anthocyanin content in petals (Rani and Singh, 2014). Combining pulsing of cut *Gladiolus* with cold storage improved flower quality due to control of bacteria growth.

The low temperature storage of flowers involves both wet and dry storage. In wet storage, flowers are stored with their bases dipped in water or preservative solution. Wet storage is used to hold the flowers for short duration and for their day to day handling. There has been a search for more effective and safer treatment of cut flowers before low temperature storage to enhance their vase life. Indeed different cut flower species requirements for the amount of carbohydrates necessary to keep vitality of postharvest flowers vary (Yamada *et al.*, 2003). Flowers are strongly depend on the carbohydrate status and the acceptable amounts of metabolic sugars affect on the rate of senescence (Kazuo *et al.*, 2005) It has been established that there is variable responses of cut flower species, and even cultivars to preservative solutions (Commission, 2003). This necessitated the study on dual effects of pulsing with 8-HQS in combination with 5 % sucrose solution and cold storage duration on the quality and vase life of cut *Gladiolus grandiflorus* 'Fado'. A similar study on this cultivar of *Gladiolus* has not been reported before.

MATERIALS AND METHODS

The *Gladiolus* flowers were grown in the open field from corms of uniform grade (2.0 – 2.5 cm diameter) at the Horticulture Research and Teaching Field, in the Department of Horticulture, Egerton University, Kenya in two years: July to December, 2013 and 2014. The field lies at a latitude of $0^{o}23'$ S and longitude 35^{o} 35' E in the Lower Highland III Agro-Ecological Zone (LH3) at an altitude of 2238 m above sea level (Fig. 1). Average maximum and minimum temperatures range from 19 ^{o}C to 22 ^{o}C day temperature and 5 ^{o}C to 8 ^{o}C night temperature, respectively, with a total annual rainfall of 1200 to 1400 mm. Soils were predominantly mollic / andosols with a pH of 6.0 to 6.5 (Jeptoo *et al.*, 2012).

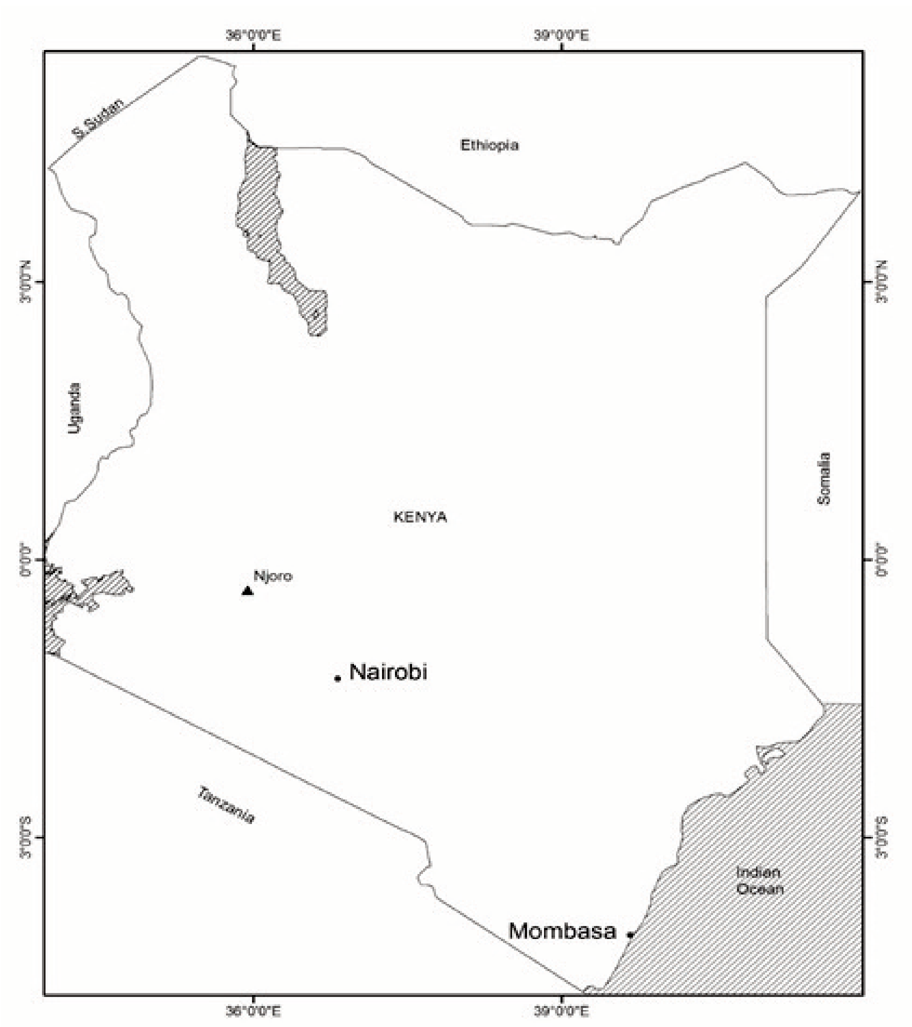

Fig. 1. The location of the study area and station.

The experiment was laid in a Completely Randomized Design with four replications in a laboratory. The flower spikes were harvested at a cut stage when the basal two florets in the spike had shown color. The flower spikes were harvested in the morning hours and brought to the laboratory in a clean plastic bucket. The stem ends were then recut at 2 cm from the base with sharp and clean secateurs. Treatments employed to fresh cut flower were (i) pulsing with sucrose (5 %) + 8-HQC (600 ppm) and (ii) without pulsing (in distilled water) at room temperature for 24 hours

(Fig. 2). These flowers were then transferred immediately to a bucket containing distilled water and kept in a refrigerated cold storage chamber maintained at 3 ± 1°C, in two different sets in separate buckets. After 0, 1, 2, 3, 4 and 5 days of storage, each of the two sets of flower treatments were brought to room temperature and compared with freshly harvested flowers for their keeping quality in tap water in the ambient temperature.

Fig. 2. Vase study of Gladiolus flowers pulsed with 600 ppm 8-HQS plus 5 % sucrose and distilled water respectively.

Study of vase life and quality of flowers

Vase life studies and quality of previously non – pulsed and pulsed gladiolus flowers were recorded after cold storage duration of 0, 1, 2, 3, 4 and 5th days. Each treatment and respective cold storage interval consisted of quadruplets of five flowers. The total number of flowers in the pulsed and non – pulsed cut gladiolus were 240. These flowers were slightly recut at the base and transferred to 1000 ml plastic cylindrical vases containing tap water to study the vase life and quality of flowers at ambient temperature.

RESULTS AND DISCUSSION

Effect of pulsing with 600 ppm of 8-HQS + 5% sucrose solution and wet cold storage duration on the changes in flower weight during post-harvest life of cut *Gladiolus grandiflorus* 'Fado'

Pulsing with 600 ppm 8-HQS + 5 % sucrose for 24 h and wet storage the cut *Gladiolus* at 3 ±1 °C showed a gain in fresh weight just after storage of both pulsed and non-pulsed flowers (Table 1). After the pulsing treatment of 600 ppm 8-HQS plus 5 % sucrose solution for 24 hours, there was significant difference (P = 0. 0031) in the spike mean fresh weight (82.77 ± 0.7934 g) as compared to the control whose mean was 79.214 ± 0.7934 g (Table. 1). The two way interactive effects were significant (0.0376; 0.0004 and 0.0016) on the gladiolus weight after pulsing, after storage and the dry weight at senescence, respectively.

It is evident from Table 1, that the highest increase in fresh weight of the *Gladiolus* spikes just after storage duration was recorded in the treatment of flowers pulsed with 600 ppm HQS + 5% sucrose and wet stored for 3 days followed by flowers pulsed with 600 ppm HQS + 5% sucrose

5% sucrose and wet stored for 3 days followed by flowers pulsed with 600 ppm HQS + 5% sucrose and wet stored for 2 days. However, they were not significantly different from each other. In the case of non-pulsed flowers the highest increase in fresh weight was recorded with the flowers wet stored for 4 days which was significantly different from all the other treatments of storage duration (Table 1). Incidentally the least mean weight after cold storage was 83.43 ± 0.645 g which was observed in spikes that had been subjected to five days cold storage duration and also pulsed with 8-HQS + 5 % sucrose. Prolonged storage of cut flowers has been found to have adverse effect on cut flowers, probably due to microbial proliferation. However, the significant interactive effect (P = 0.0004) of pulsing and cold storage on weight after cold storage period is in agreement with work done by Makwana *et al.* (2015). In their study, cold storage without pulsing treatment led to decrease in vase life and floret opening of cut tuberose inflorescence. Pulsing with 600 ppm 8-HQS + 5 % sucrose solution and cold storage durations also significantly affected the rate of water uptake (P < 0.0001).

Treating cut flowers with chemical preservatives before storage is generally the acceptable procedure adopted to extend the vase life and quality in floriculture (Business, 2016). However, there are exceptions to this rule according to studies done on yarrow 'Cassis' in which the longest vase life was attained by placing the cut stems in water (Clark *et al.*, 2010). Lowering storage temperature is the easiest option adopted to increase the relative humidity to within 90 to 95 percent so as to prevent water loss in cut flowers (Reid and Jiang, 2012). Cold storage of cut flowers facilitates the adjustment of flowers and other planting material supplies against market demand and also enables accumulation of large quantities (Senapati *et al.*, 2016). It has been established that long periods of storage ultimately have negative effects on the ultimate vase life of the product (Bayleyegn *et al.*, 2012). Pulsing treatment has been proved to control xylem vessel blockage that occurs due to air embolism, tissue injury or microbial proliferation (Kwon and Kim, 2000). 8-HQS is an antimicrobial agent that improves water uptake in cut flowers. According to this study, there was no significant difference in the fresh weight of cut *Gladiolus* flowers on the third day in vase after the cold storage duration.

At senescence, the fresh weight was less than the initial weight after pulsing. The maximum fresh weight at senescence was recorded in pulsed flowers and wet stored for 3 days (64.36 g) which was not significantly different from the pulsed flowers and wet stored for 4 days which also gave the maximum vase life (11.50 days). The decrease in fresh weight increased as the storage duration increased from 4 to 5 days. With the non-pulsed flowers, maximum fresh weight was recorded with flowers wet stored for 2 days (64.48 g) which was not significantly different from flowers wet stored for 3 and 4 days.

The maximum dry weight at senescence was recorded with pulsed flowers and wet stored for 2 days which also gave a vase life of 11.25 days and was statistically similar to pulsed flowers and wet stored for 4 days. There was significant difference between the dry weight of flower spikes pulsed with 8-HQS + 5 % sucrose compared with the ones that had been placed in distilled water (7.202 ± 0.164; 6.669 ± 0.164, P = 0.0272, respectively) with a strong interactive index between the treatments and the storage durations (P = 0.0016) (Table 1). There was also significant difference (P = 0.0118) in the dry weight of the gladiolus spikes depending on the storage duration. This study has also indicated a two-way interactive index (P = 0.0016) between the pulsing treatment and the period of cold storage and their effect on the dry weight of the cut flower

Effect of pulsing with 600 ppm HQS plus 5 % sucrose and wet cold storage on the number of open florets and unopened buds in *Gladiolus grandiflorus* 'Fado'

There was significant effect of period of cold storage on the number of open florets and unopened buds (Table 2). Highest number of open florets was recorded in the treatment of cut

Table 1. Effect of pulsing with 600 ppm 8-HQS + 5 % sucrose solution versus distilled water (P) and wet cold storage duration on weight (g) changes during post-harvest life of cut *Gladiolus grandiflorus* 'Fado'.

Days of storage	Flower weight (g)												Vase life (days)	
	Just after pre-storage treatment		Just after storage		On third day in vase				On senescence					
					Fresh weight (g)		Dry weight (g)		Fresh weight (g)		Dry weight (g)			
	Pulsing	Nopulsing	Pulsing	No pulsing	Pulsing	No Pulsing	Pulsing	Nopulsing	Pulsing	Nopulsing	Pulsing	Nopulsing	Pulsing	Nopulsing
0	82.96 ^{b}	77.36 ^{b}	-	-	85.93^{a}	85.89^{a}	7.70^{ab}	6.69^{abc}	57.49^{abc}	58.38^{abc}	6.89^{bc}	6.78 ^{bc}	8.75 ^{b}	4.75 ^{c}
1	76.58 ^{b}	79.08 ^{b}	84.43 ^{b}	85.95 ^{b}	82.69^{a}	87.62^{a}	7.13^{abc}	7.19^{abc}	62.53 ^{ab}	63.90^{a}	9.00 ^{a}	6.80^{bcbc}	10.25 ^{ab}	9.50 ^{ab}
2	83.39^{ab}	80.52 ^{b}	92.37^{ab}	88.86 ^{b}	85.99^{a}	78.05^{a}	7.17^{abc}	7.05^{abc}	63.00 ^{ab}	64.48^{a}	6.56^{bc}	6.69^{bc}	10.00 ^{ab}	9.50 ^{ab}
3	90.34 ^{a}	79.44 ^{b}	100.20^{a}	86.63 ^{b}	71.48^{a}	83.54^{a}	7.65^{abc}	7.38^{abc}	64.36 ^{a}	58.13^{abc}	6.87^{bc}	6.67^{bc}	11.50 ^{a}	9.75 ^{ab}
4	83.19^{ab}	81.95^{ab}	89.00 ^{b}	99.76 ^{a}	97.37^{a}	91.69^{a}	8.75^{a}	7.12^{abc}	55.70^{abc}	59.81^{ab}	8.12 ^{ab}	6.18^{bc}	11.25 ^{ab}	11.00^{ab}
5	80.23 ^{b}	76.94 ^{b}	83.43 ^{b}	86.35 ^{b}	72.93^{a}	80.23^{a}	5.15^{bc}	5.75^{c}	49.95 ^{bc}	53.45^{bc}	5.97 ^{c}	6.90^{bc}	10.25ab	10.2^{5ab}
Mean	82.77^{a}	79.21^{b}	89.86^{a}	89.91^{a}	82.72^{a}	84.46^{a}	7.4^{a}	6.8^{b}	59.41^{a}	59.10^{a}	7.20^{a}	6.66^{b}	10.33^{a}	9.13^{b}
C.D at 5 %	0.79	0.79	0.99	0.99	2.7	2.7	0.16	0.16	0.81	0.81	0.163	0.163	0.148	0.148
Pulsing (P)	0.0031		0.9690		0.6521		0.0188		0.7879		0.0272		0.0003	
Period of storage (S)	0.0082		< 0.0001		0.1035		0.0006		< 0.0001		0.0118		< 0.0001	
P× S	0.0376		0.0004		0.6440		0.2754		0.1311		0.0016		0.0041	

* In each column, means with the similar letter(s) are not significantly different ($P < 0.05$) using LSD test.

Gladiolus flowers pulsed with 600 ppm HQS + 5% sucrose and wet stored for 4 days (11.0) followed by *Gladiolus* flowers pulsed with 600 ppm HQS + 5% sucrose and wet stored for 3 days (10.5) which also gave a maximum vase life of 11.50 and 11.25 days, respectively. The highest number of unopened buds were found in the spikes pulsed with distilled water and received no cold storage treatment (Table 2). In the same study, 100 mg Al_2 $(SO)_4$ plus 2 % sucrose showed the highest vase life for *Dendrobium* 'Ela Sakur'. In another study a report indicated a two way significant interaction ($P < 0.01$) between pulsing solution and storage period on *Botrytis* incidence (Bayleyegn *et al*., 2012). The keeping quality of cut *Gladiolus grandiflorus* L. cv. 'White' prosperity, pulsing with 20% sucrose + 300 ppm Al_2 $(SO)_4$ + GA_3 ppm gave superior results in terms of days to open basal florets, vase life, number of florets opened, floret size, longevity of open florets and fresh weight/gain/loss (Bhat and Sheikh, 2015).

Table 2. Effect of pulsing with 600 ppm HQS plus 5 % sucrose on the number of open florets and unopened buds in *Gladiolus grandiflorus* L. 'Fado'.

Days of Storage	Opened florets (No.)		Unopened buds (No.)		Vase life (days)	
	Pulsed	Non- pulsed	Pulsed	Non-pulsed	Pulsed	Non–pulsed
0	8.60 [bc]	6.88 [c]	5.00 [b]	8.00 [a]	8.75 [b]	4.75 [c]
1	10.00 [ab]	10.00 [ab]	4.25 [b]	4.50 [b]	10.25 [ab]	9.50 [ab]
2	9.75 [ab]	9.50 [ab]	5.13 [b]	4.50 [b]	10.00 [ab]	9.50 [ab]
3	10.50 [ab]	9.25 [ab]	3.75 [b]	5.08 [b]	11.50 [a]	9.75 [ab]
4	11.00 [a]	10.25[ab]	4.50 [b]	4.50 [b]	11.25 [ab]	11.00[ab]
5	8.75 [bc]	10.00 [ab]	4.25 [b]	4.50 [b]	10.25ab	10.2[5ab]
Mean	9.76 [a]	9.31[a]	4.47	5.18	10.33[a]	9.13[b]
C.D at 5 %	0.159	0.159	0.2129	0.2129	0.148	0.148
Pulsing (P)	0.0512<		0.0256		0.0003<	
Period of storage (S)	0.0001		0.0012		< 0.0001	
P × S	0.0095		0.0259		0.00041	

Means followed by the same letter within evaluation period are not significantly different according to Tukey's test at 5 % level of confidence.

Effect of pulsing and wet cold storage on water uptake and vase life during the post-harvest life of cut *Gladiolus grandiflorus* 'Fado'

There was significant effect on water uptake by cut *Gladiolus* spikes due to the pulsing and wet cold storage treatments ($P<0.0001$). The same trend was observed after pulsing, on the third day and at senescence (Table 3). The mean water uptake for spikes pulsed with 600 ppm plus 5 % sucrose was 21.205 ± 0.16 ml in comparison with those pulsed with distilled water (12.412 ± 0.16 ml) as shown in Table 3. A higher mean water uptake (31.98 ml ± 0.193) was observed on the third day with spikes pulsed with 600 ppm 8-HQC + 5% sucrose and 23.87 ml ± 0.26 at senescence, respectively in contrast with 23.14 ± 0.93 and 17.054 ± 0.26 ml, respectively for the non-pulsed cut *Gladiolus* spikes (Table 3).

Pulsing of *Gladiolus* cut spikes with 20% sucrose plus 200 ppm 8-HQS for 20 hrs at 23±2°C increased flower vase life compared to non-pulsed flowers that were stored at 5±1°C (Bhat and Sheikh, 2015). A study done on rose flowers (Bayleyegn *et al*., 2012) showed that storage time significantly (P < 0.001) affected on solution uptake, relative fresh weight and TSS contents of petals. Cold storage of cut flowers facilitates the adjustment of flowers and other planting material supplies against market demand and enable accumulation of quantities of flowers (Senapati *et. al*., 2016).

Table 3. Effect of pulsing with 600 ppm 8-HQS + 5 % sucrose solution and wet cold storage duration on water uptake during postharvest life of cut *Gladiolus grandiflorus* cv. 'Fado'.

Storage duration (days)	Water uptake (ml)							
	After pre-storage		After storage duration		On third day in vase		At senescence	
	Pulsing	No pulsing	Pulsing	No pulsing	Pulsing	No pulsing	Pulsing	No pulsing
0	18.51^{cd}	10.99^{ef}	-	-	44.56^{a}	29.32^{d}	8.75^{b}	4.75^{c}
1	23.93^{a}	11.70^{ef}	36.09^{c}	30.36^{d}	34.69^{c}	23.24^{e}	10.25^{ab}	9.50^{ab}
2	18.89^{cd}	11.57^{ef}	30.90^{d}	30.11^{d}	28.93^{d}	23.62^{e}	10.00^{ab}	9.50^{ab}
3	20.34^{bc}	10.04^{f}	31.65^{d}	26.44^{e}	23.34^{e}	18.35^{g}	11.50^{a}	9.75^{ab}
4	24.31^{a}	12.55^{e}	51.66^{a}	47.25^{b}	39.63^{b}	23.77^{e}	11.25^{ab}	11.00^{ab}
5	21.28^{b}	17.62^{d}	30.61^{d}	29.53^{d}	20.75^{f}	20.56^{fg}	10.25^{ab}	10.25^{hi}
Mean	21.20^{a}	12.41^{b}	36.18^{a}	32.74^{b}	31.98^{a}	23.15^{b}	23.87^{a}	17.05^{b}
C.D at 5 %	0.16	0.16	0.25	0.25	0.19	0.19	0.26	0.26
Pulsing (P)	< 0.0001		< 0.0001		< 0.0001		< 0.0001	
Period of storage (S)	< 0.0001		< 0.0001		< 0.0001		< 0.0001	
P×S	< 0.0001		0.0001		< 0.0001		< 0.0001	

Means followed by the same letter within evaluation period are not significantly different according to Tukey's test at 5 % level of significance.

CONCLUSION

The pulsing treatment coupled with cold storage had significant effect on the vase life and post-harvest quality of cut *Gladiolus grandiflorus* 'Fado'.

The quality parameters of fresh and dry weight, number of open florets per spike and water uptake were superior in *Gladiolus* spikes pulsed with 600 ppm 8-hydroxyquinoline sulphate coupled with 5 % sucrose solution in comparison with the control.

It can be concluded that *Gladiolus* cut flowers can be wet stored at 3 ± 1°C up to a maximum of 4 days without any adverse effect on their subsequent vase life and quality.

These results can augment existing cut flower technologies in enhancing the vase life and quality of *Gladiolus grandiflorus* 'Fado'.

RECOMMENDATIONS

Dry storage of the *Gladiolus* cv. 'Fado' can be adopted after the pulsing treatment to assess the quality and vase life of this cut flower in preference over the wet cold storage.

Variation in the concentration of 8-hydroxyquinoline sulphate can be done to gauge whether this would improve the vase life and quality of the cut *Gladiolus*.

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How to cite this article:

Kavulani Chore, J., Mwangi, M. and Karori Mbuthia, S. 2019. Effect of pulsing and wet cold storage on the quality and vase life of cut gladiolus (*Gladiolus grandiflorus* L.) 'Fado'. *Journal of Ornamental Plants, 9*(1), 13-22.

URL: http://jornamental.iaurasht.ac.ir/article_664261_215e26d5e92924c4d4f200f1dcb9843e.pdf