Efficiency Evaluation of Pulsing Solutions and Cold Storage on Prolonging the Vase Life of *Helianthus annuus* L. Cut Flowers

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ABSTRACT: Preserve the vase life is an important factor for the evaluation of cut flowers quality, for both domestic and export markets. This study aimed to check the effect of cold storage durations (0, 1 and 2 weeks) and various combinations of pulsing solutions T1(tap water) as a control, T2 (Hydroxyquinoline sulfate (8-HQS) 200 mg/L +Sucrose 2%),T3 (Olive leaf extract 10 ml/L +Sucrose 2%), T4 (Lupine extract 10 ml/L +Sucrose 2%), T5 (Olive leaf extract 10 ml/L+ Coriander oil 300 ppm+25mg/L Kinetin +Sucrose 2%) , T6 (Olive leaf extract 10 ml/L + Dill oil 300 ppm+25mg/L Kinetin +Sucrose 2%), T7 (Lupine extract 10 ml/L+ Coriander oil 300 ppm +25mg/L Kinetin +Sucrose 2%), T8 (Lupine extract 10 ml/L + Dill oil 300 ppm +25mg/L Kinetin +Sucrose 2%), T9 (Salicylic acid 200 mg/L+ 25 mg/L Kinetin +Sucrose 2%), T10 (Citric acid 150 mg/L +25 mg/L Kinetin +Sucrose 2%) on quality parameters of *Helianthus annuus L.* cultivar "Sunbright Supreme". A significant decrease in chlorophyll index, water balance, carotenoids content and vase life was obtained by increasing the cold storage duration compared to control. The maximum vase life was recorded by pulsing solutions T5 (10.18 and 11.11days) in both experiments, further T8 and T3 (10.44 and 10.30 respectively) in the second experiment.

Keywords: *Helianthus annuus*, Sunflower, cold storage, vase life, pulsing solutions, cut flowers**,** essential oils**,** Plant extracts

INTRODUCTION

Helianthus annuus L. is the annual cultivated sunflower, that belongs to family Asteraceae which have 67 species in the Helianthus genus, (Parmeshwar 2010). New varieties for use in flower markets of ornamental sunflowers were produced from breeding programs. These varieties have made the sunflower a renowned product in flower arrangements, due to its odd shape and the different colors of flowers, that add life and dynamism to the environments. Since, it is a novel product in the floriculture markets, there is little research on postharvest technology for this species when produced for ornamental purposes, (Nascimento et al., 2019). The sunflower crops are characterized by a short cycle, thus provide a quick return for an investment. Additionally, it is easy for propagation, management, and photosynthetic efficiency are high. Generally, the sunflower is among the most global crops and highly proper to different environments, (Da Silva et al., 2018). People are yet buying flowers and the flower trade is increasing rapidly all over the world, despite all the problems and obstacles that facing cut flowers after harvest. So, we are now required to devise new methods to improve the quality of the flower for the farmer and the consumer. Thus, the objectives of this study, were to define the impact duration of dry cold storage and the best combinations of different compounds that used as pulsing solutions before cold storage on the chlorophyll index and water balance, carotenoids content and vase life of sunflower cut flowers.

MATERIALS AND METHODS

Two separate experiments were achieved at the Department of Plant Production, Faculty of Agriculture "Saba Basha", Alexandria University in June, and January (2019-2020) on Helianthus (sunflower) cut flowers. This study was carried out to evaluate the effect of dry cold storage and different combinations of the pulsing solutions control (tap water), hydroxyquinoline sulfate, sucrose, olive leaf extract, lupine extract, coriander oil, dill oil, kinetin, salicylic acid and citric acid on the vase life of *Helianthus annuus L.* cultivar "Sunbright Supreme" that was gained from a famous commercial nursery at uniform length and weight in the early morning and then covered with a polyethylene sheet and be moved quickly to the laboratory at room temperature, normal relative humidity, and light from a white fluorescent lamp. All stems were cut back even to 40 cm and the leaves of the lower third part of stems were removed in order to avoid contamination in solutions of postharvest treatments as recommended by Khimani *et al.*, (2005). The cut flowers were placed in plastic containers containing pulsing solutions consisting of various combinations of the previous compounds for 24 hours before refrigerated storage for 0, 1, and 2 weeks, under dry conditions wrapped in cellophane paper at 2°C and 90-95 % relative humidity. The flowers were moved to glass containers that have 400 ml of tap water to determine the vase life and other tested data at 18±2°C and about 50-60%relative humidity with a white fluorescent lamp to the light.

Experimental layout and statistical analysis:

Data in Table (1) was arranged in a Split-Split Plot Design, the duration of cold storage (S) was considered as the main plot, whereas the subplot was the vase life periods (V). On the other hand, the pulsing solutions (P) were the sub-sub plot. Otherwise, data in Table (2) was arranged in a Split Plot Design, the duration of cold storage (S) was considered as the main plot and the pulsing solutions (P) were the subplot. All data were statistically analyzed by the analysis of variance as described by Steel and Torrie (1980); all analyses were done by Average of SAS (2002) statistical software.

Chlorophyll Index (SPAD): Chlorophyll index was measured by chlorophyll meter (SPAD- 502, Minolta Co. Japan). An average of 3 measurements from different spots of a single leaf was considered, (Yadava, 1986).

Water Balance (g/flower): The water balance was calculated as the difference between water uptake through the cut flowers and water loss by flowers at the end of vase life, (Sankat and Mujaffar 1993).

Carotenoids content in Flowers (mg/100g FW): Carotenoids were determined calorimetrically in 5 g of the fresh petals according to the method described by (Alan, 1994).

Vase Life (days): The end of the vase life of sunflower was defined as the number of days on vase till wilting of petals and collapsing of the stem, (Clark et al., 2010).

RESULTS AND DISSCUSSION

Table (1): The effect of duration of cold storage, vase life periods and pulsing solutions on chlorophyll index (SPAD), water balance (g) and carotenoids (m g/100g FW) of *Helianthus annuus L.* cultivar "Sunbright Supreme" in the first and second experiments.

Treatment	Chlorophyll Index		Water Balance (g)		Carotenoids (mg/100g FW)	
	2019	2020	2019	2020	2019	2020
	Duration of cold storage		Duration of cold storage		Duration of cold storage (S)	
S0	38.16	35.97	17.06	25.31	6.48	6.71
S1	35.43	35.87	10.22	18.46	6.12	5.88
S2	30.43	36.4	7.71	13.27	4.68	4.61
L.S.D	1.2	1.98	2.77	9.26	0.3	0.26
	Vase life periods (V)		Vase life periods (V)		Vase life periods (V)	
V1	35.4	36.92	18.23	23.45	6.66	6.59
V2	34.06	36.22	8.86	16.78	5.39	5.09
V3	34.56	35.09	7.9	16.81	5.22	5.52
L.S.D	0.55	1.62	1.53	4.83	0.21	0.33
	Pulsing solutions (P)		Pulsing solutions (P)		pulsing solutions (P)	
P1	36.14	36.41	13.67	20.04	4.41	4.56
P2	32.78	38	12.83	11.98	5.44	5.45
P3	32.99	37.09	10.84	22.42	5.54	5.26
P4	33.1	36.8	12.22	20.68	6.05	5.9
P5	34.72	37.68	14.36	23.23	6.06	5.93
P6	39.45	33.95	14.34	12.59	5.72	5.7
P7	33.03	36.44	9.58	18.61	5.29	5.1
P8	34.09	36.09	10.39	24.32	5.7	5.6
P9	36.03	34.56	8.63	17.72	6.69	7.04
P10	34.39	33.78	9.79	18.55	6.7	6.8
L.S.D	1.86	2.2	3.55	5.16	0.3	0.25
	Interactions		Interactions		Interactions	
S X V	**	NS	**	**	**	**
S X P	**	**	**	**	**	**
V X P	**	*	**	*	**	**
S X V X P	**	NS	**	NS	**	**

S: duration of cold storage**, V:** vase life periods and **P**: pulsing solutions. (*) Significant at 0.05 level of probability. (**) Highly significant at 0.01 level of probability. (NS) not significant at 0.01 level of probability. L.S.D least significant difference at 0.05 level of probability

Chlorophyll Index (SPAD):

Data revealed that zero storage duration had a significant increase in chlorophyll index of *Helianthus annuus* cut flowers (38.16) followed by the first duration of storage (35.43) compared to the second storage duration in the first experiment. Regarding the second experiment, no significant differences were found between storage durations and non-storage treatments. Otherwise, the chlorophyll index had a significant decrease with increasing vase life periods in the first experiment, except the second period of vase life that didn't show a significant difference in comparison with the third vase life period. On the other hand, the biggest chlorophyll index was recorded by pulsing solution 6 in the first experiment. These decrease in the index of chlorophyll with increasing cold storage duration may be due to the process of respiration thus resulted in more consumption of carbohydrates and depletion of stored food in cut flowers then reducing chlorophyll content. Moreover, the low temperature during cold storage resulted in reducing photosynthetic efficiency in the tissue of plants as shown by (Barakat, 2013). That decrease in chlorophyll index during vase life periods may be due to the increase of respiration rate and the action of ethylene which usually causes yellowing leaves as reported by (Marandi *et al.*, 2011). This significant effect of pulsing solution 6 on chlorophyll index may be returned to the effect of olive leaf extract that is rich in polyphenols that has excellent antimicrobial and antioxidant properties. Additionally, the role of dill oil to enhance the vase life of cut flowers by controlling microorganisms. Furthermore, the ability of kinetin and sucrose to keeping the longevity of cut flowers through enhancing water balance and delaying leaf yellowing as stated by (Chen *et al.*, 2001), (Bayat *et al.*, 2013), (Stanojevic *et al.*, 2016) and (Al-Rimawi *et al.*, 2017). A highly significant effect was shown in interactions on chlorophyll index between storage duration and vase life periods, storage duration and pulsing solutions, vase life periods and pulsing solutions and between the three interaction factors in both experiments, except the interaction between vase life periods and pulsing solutions in the second experiment that had a significant effect. Moreover, the non-significant effect of interactions between storage duration and vase life period in the second experiment and between the three interaction factors (S*V*P) in the second experiment.

Water Balance (g):

Data reported that the water balance of *Helianthus annuus* cut flowers significantly decreased with increasing storage duration in both experiments. In addition, the significant effect of vase life periods on the water balance of *Helianthus annuus* cut flowers in both experiments. Whereas, the first period of vase life recorded the best water balance compared to the second and third vase life periods in the first and second experiments. Furthermore, there were significant differences in the water balance of *Helianthus annuus* cut flowers between the different combinations of pulsing solutions in both experiments. The pulsing solutions 5, 8 resulted in the better water balance in the first and second experiments respectively. This decrease in water balance with increasing the cold storage duration could be due to the decrease of the water uptake effectiveness during the long duration of cold storage that increasing water stress as a result of blocking the stem by air. Where the water deficit is increased only when the transpiration rate is higher than the rate of water uptake, hence the delaying of water stress onset is achieved by reducing the transpiration rate as indicated by (Van Meeteren, 1992), (Ferrante *et al.,* 2002) and (van Doorn, 1997). Likewise, the decrease in water uptake during the latest vase life periods might be a result of bacterial proliferation thus increase water deficiency-related wilting and ending vase life as shown by (Anjum *et al.*, 2001) and (Hashemabadi *et al.*, 2015). The positive effect of pulsing solutions 5 and 8 might be returned to the effectiveness of essential oils like dill and coriander to prolong the vase life of cut flowers as antimicrobial agents. Furthermore, the ability of leaf extracts for reducing the microbial proliferation of vase solution where the olive leaf extract and lupine extract are considered as antimicrobial and antioxidant agents that preserving vase life of cut flowers. Additionally, reduction of water stress as a result of using cytokinin and sucrose thus delaying senescence of cut flowers as indicated by (Shanan *et al.*, 2010), (Rahman *et al.*, 2012), (Yateem *et al.*, 2014), (Khan et al., 2015), and (Arrom and Munné-Bosch 2012). Results showed the highly significant effect in interactions between storage duration and vase life periods, additionally, storage duration and pulsing solutions. Moreover, vase life periods and pulsing solutions and the interactions between the three factors on the water balance of *Helianthus* *annuus* cut flowers in both experiments, except the significant effect between the vase life periods and pulsing solutions in the second experiment. Further, the non-significant effect between the three factors of interaction in the second experiment.

Carotenoids (m g/100g FW):

Data indicated that carotenoids content in petals of *Helianthus annuus* cut flowers was decreased with increasing storage duration in both experiments. The highest content of carotenoids 6.48 and 6.71 in the first and second experiments respectively was recorded by the control (zero storage duration). Likewise, carotenoid content in petals of *Helianthus annuus* cut flowers was decreased with increasing vase life periods in both experiments. The highest carotenoid content was obtained by the first vase life period 6.66 and 6.59 in the first and second experiments respectively. Otherwise, combinations of pulsing solutions had a significant increase in carotenoid content in petals of *Helianthus annuus* cut flowers compared to control in both experiments. The pulsing solution number 10, 9 recorded the highest content of carotenoids in both experiments. These findings may be due to the high rate of respiration processes during the long storage duration thus, lead to rapid senescence that required metabolic energy to initiate it that obtains through respiration as reported by (Ezhilmathi *et al.,* 2007) and (Halevy and Mayak 1979). Moreover, as a result of the increase in respiration rate with the vase life periods passed on which cause consumption of carbohydrates and food depletion leading to senescence of flower "the terminal phase of developmental processes" thus to the end of its life span as stated by (Rani and Singh, 2014). On the other hand, the positive effect of the pulsing solutions 10 and 9 may be due to the salicylic acid role in prolonging vase life, minimizing respiration rate, and higher stability of the membrane and delaying the rapid senescence. Likewise, the ability of citric acid and sucrose to delaying the senescence of flowers, extending the vase life, and promoting post-harvest quality of cut rose flowers. Additionally, the effective of kinetin and sucrose in keeping the longevity of flower (Peng *et al.,* 2007), (Ezhilmathi *et al.,* 2007), (Ershad Langroudi *et al.,* 2020), (Aryal *et al.,* 2019) and (Chen *et al.,* 2001). There were highly significant effects of interactions between storage duration and vase life periods, between storage duration and pulsing solutions, between vase life periods and pulsing solution and the three interaction factors (S*V*P) on carotenoids content in the petals of *Helianthus annuus* cut flowers.

Vase Life (Days):

Data revealed a significant decrease in vase life of *Helianthus annuus* cut flowers with increasing storage duration in both experiments. The longest vase life was achieved by control, followed by the first duration than the second duration of storage in the first experiment. However, there was an absence of a significant difference between the first and the second duration of storage in the second experiment. Furthermore, the vase life of *Helianthus annuus* cut flowers was increased significantly with using combinations of pulsing solutions compared to control (tap water) in both experiments, except treatment of pulsing solution (10) and (2and 10) in the first and second experiments, respectively where the differences weren't big enough to be significant compared to control. The pulsing solution 5 resulted in the maximum vase life (10.18 and 11.11) in the first and second experiments respectively, in addition, the pulsing solutions 8 and 3 (10.44 and 10.30 respectively) in the second experiment. Decreasing the vase life might be due to the high respiration rate during the long dry cold storage of flowers. Respiration leads to higher rates of carbohydrates consumption and food depletion thence reflected in the number of days of vase life as shown by (Walton *et al.,* 2010). The positive effect of pulsing solutions 5, 8 and 3 may be returned to the olive leaf extract and lupine extract, coriander oil and dill oil as antimicrobial and antioxidant properties and the ability of kinetin and sucrose to delay senescence thus increase the vase life of cut flowers as shown by (Yateem *et al.,* 2014), (Khan et al., 2015), (Silva and Domingues 2017), (Stanojevic *et al.,* 2016) and (Chen *et al.,* 2001). Data reported that there was a highly significant effect of interaction on the vase life of *Helianthus annuus* cut flowers between storage duration and pulsing solution in the first experiment, whilst the effect of interaction was significant in the first experiment.

CONCLUSION

There was a significant decrease in chlorophyll index, water balance and carotenoids pigments in *Helianthus annuus* L. cv. "Sunbright Supreme" with increasing of both cold storage duration and vase life periods. Additionally, the pulsing solution 5 was achieved the maximum vase life (10.18 and 11.11) in the first and second experiments respectively, further the pulsing solutions 3 and 8 (10.44 and 10.30 respectively) in the second experiment.

Table (2): The effect of duration of cold storage and pulsing solutions on the vase life of *Helianthus annuus L.* cultivar "Sunbright Supreme" in the first and second experiments.

Treatment	Vase Life (Days)	
	2019	2020
	Duration of cold storage	
S0	11.55	13.61
S1	7.35	8.39
S2	6.22	7.46
L.S.D	0.54	1.14
	pulsing solutions (P)	
P1	7.01	8.70
P2	7.94	9.44
P3	8.66	10.30
P4	8.56	10.04
P5	10.18	11.11
P6	8.48	9.96
P7	8.59	9.74
P8	8.67	10.44
P9	8.07	9.78
P10	7.59	8.67
L.S.D	0.67	0.89
	Interactions	
S X P	**	*

Duration of Cold Storage and P: pulsing solutions. (*) Significant at 0.05 level of probability. (**) Highly significant at 0.01 level of probability. L.S.D least significant difference at 0.05 level of probability.

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الملخص العربى

تقييم كفاءة المحاليل النابضة والتخزين البارد فى إطالة عمر أزهار عبادالشمس المقطوفة

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يعتبر العمر القصير نسبيًا للزهور المقطوفة بعد الحصاد أحد أخطر العوامل التي تؤثر على تسويق الزهور المقطوفة. لذلك ، فإن الحفاظ على عمرالأزهار هو عامل مهم لتقييم جودة زهور القطف ، لكل من الأسواق المحلية وأسواق التصدير. هدفت هذه الدراسة إلى التحقق من تأثير فترات التخزين البارد (0 ، 1 ، 2 أسبوع) ومجموعات مختلفة من المحاليل النابضة P1(ماء الصنبور) كنترول, P2 (كبريتات هيدروكسى الكينولين8-HQS 200 مجم/ لتر + سكروز 2٪), P3 (مستخلص أوراق الزيتون 10 مل / لتر + سكروز 2٪), P4 (مستخلص الترمس 10 مل / لتر + سكروز 2٪), P5 (مستخلص أوراق الزيتون 10 مل / لتر + زيت كزبرة 300 جزء في المليون + 25 مجم / لتر كينتين + سكروز 2٪), P6 (مستخلص أوراق الزيتون 10 مل / لتر + زيت شبت 300 جزء في المليون + 25 ملجم / لتر كينتين + سكروز 2٪), P7 (مستخلص الترمس 10 مل / لتر + زيت كزبرة 300 جزء في المليون + 25 مجم / لتر كينتين + سكروز 2 ٪), P8 (مستخلص الترمس 10 مل / لتر + زيت الشبت 300 جزء في المليون + 25 مجم / لتر كينتين + سكروز 2٪), P9 (حمض الساليسيليك 200 مجم / لتر + 25 مجم / لتر كينتين + سكروز 2٪), P10 (حمض الستريك 150 مجم / لتر + 25 مجم / لتر كينتين + سكروز 2٪). بناءً على معايير جودة أزهار *Helianthus annuus L.* صنف," Sunbright" Supremeأظهرت البيانات انخفاضًا معنويًا في مؤشر الكلوروفيل و التوازن المائى ومحتوى الكاروتينات وفترة حياة الأزهار في أزهارعبادالشمس مع زيادة مدة التخزين البارد في كلا التجربتين. تم تسجيل الحد الأقصى لعمر الأزهار بواسطة T5 (10.18 و 11.11 يومًا) في كلا التجربتين ، وكذلك T8 و T3 (10.44 و 10.30 على التوالي) في التجربة الثانية.