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Effects of pretreatment on the cut flowers quarantined by EB radiation

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Abstract: Fresh cut flowers including yellow and white chrysanthemum (*chrysanthemum* sp) and red carnation (*Dianthus caryophyllus* L) were electron beam irradiated as quarantine treatment. The results showed that the irradiated flowers could meet the phytosanitary requirements in the international trading. In this study, the cut flowers were pretreated with the commercial preservative and sugar solutions in order to increase their radio-tolerance and expand their vase-life. The pretreatment has also reduced the weight loss, browning rate of leaves, and brightness of the irradiated flowers. The results revealed that the commercial quality of the irradiated cut flowers pretreated with 2% glucose solution 2 hours, then 0.024% silver thiosulphate (STS) solution for further 2 hours was remained after storage at 4-6^{o}C. Pre-treatment with 2% glucose and 0.024% STS before irradiation at 400 Gy and 600 Gy was chosen as the best way for improving the raditain tolerance of the cut flowers. The vase-lifes of the irradiated cut flowers are 6 days for yellow chrysanthemum*;* 8 days for white *chrysanthemum* and 8-10 days for red carnation similar to non-irradition ones.

Keywords*:* *Cut flower, electron beam, irradiation, phytosanitary, pretreatment.*

I. INTRODUCTION

Chrysanthemums's and carnation's cut flowers are commodities that require quarantine treatment for export or import. The disinfestation treatment of fresh cut flowers is usually abused chemicals like methyl bromide or other fumigants. However, methyl bromide will be phased out because it is ozone depleting substance and unsafe for workers [1]. And establishing alternative measures such as radiation quarantine technique would be highly desirable.

Many papers were reported for the irradiation effects as quarantine treatment on cut flowers as well as other fresh vegetables. It has been recognized that the radiation sensitivity depends on the kind of flowers [2]. For example, while carnation are relative radiation tolerant flower that can withstand to gamma irradiation up to 700 Gy irrespective of their maturity [3]. Chrysanthemums are sensitive to radiation. Therefore, some pretreatment techniques have been studied and applied for the cut flowers in order to improve their radiation tolerance. The pre-treatment with silver thiosulphate (STS) solution has commercialized for expanding the vase-life of the irradiated cut flowers, especially when they are kept in ethylene-contaminated environments like in supermarkets. It has been reported that the vase-life of many cut flowers such as carnation, matthiola, consolida, chrysanthemum, zingiber, anthirrinum and delphinium can be prolonged by exposure to 1-methylcyclopropene (1-MCP), a volatile and non-toxic compound that can be used as inhibitor for ethylene [4, 5]. Pre-treatment with sugar solution can reduce the radiation-induced damages in some kinds of chrysanthemum [6]. In this studies, the cut flowers of chrysanthemum and carnation were pretreated with sugar, 1-MCP, silver nano and STS, then EB irradiated at 400 and 600 Gy then their quality and radio-tolerance were investigated.

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II. MATERIALS AND METHODS

Cut flower preparation

Chrysanthemum and carnation used for all the tests were collected from some flower exportation companies in Da Lat City. These flowers were harvested at commercial standards in the afternoon, immediately transported to laboratory in the next early morning. Each stem of flower was recut to 1-2 cm and then pre-treated with different chemical solutions in 2h and then irradiated by electron beam accelerator (UERL-10-15S2, 10 MeV, 15 kW, CORAD Co. Ltd., Russia) at the Research and Development Center for Radiation Technology with the same dose of 400 Gy as following:

Table I. Some commercial preservative and sugar solutions were used to pretreatment irradiated cut flowers.

Sample	Pretreatment	Dose (Gy)	Storage Conditions	Measure
Control	Water	400	T = 27^{o}C, RH = 70-80%	- Weight loss - Flower color - Rate of yellowed leaves
Glucose	2% glucose	t.s[a]	t.s	t.s
Glucose-Nano	2% glucose + 50ppm silver Nano	t.s	t.s	t.s
Glucose-STS	2% glucose + 0.024% silver thiosulfate	t.s	t.s	t.s
Glucose-1MCP	2% Glucose + 4nL/L 1-MCP	t.s	t.s	t.s

[a]t.s: the same

To assess the effects of glucose-STS on trading value of the irradiated cut flowers, the flowers was pretreated by soaking in glucose+STS and irradiated at dose 400 Gy and 600 Gy by electron beam. Non treated flowers were also studies as control sample. After 6 days storage, the flowers were moved to room temperature and placing in vase to measure their weight loss, color change, and rate of yellowed leaves.

Assessment of flower quality

Fresh weight loss [7]: Fresh weight loss of tested flowers was measured every 2 days of the vase-life period for treatments.

Color measurement [8]: The color of flower and leaf was measured by colorimeter (CR-200, Minolta Co., Japan). Three stems were chosen from each trial of cut flowers. In each stem chosen 3 flowers and 3 leaves to measured, and the average value of L (luminosity), a (green-red), b (blue-yellow), and $\triangle$E for each stem was collected for analysis. $\triangle$E is the total difference and defined as $(\triangle L^2 + \triangle a^2 + \triangle b^2)^{1/2}$, $\triangle$L, $\triangle$a, $\triangle$b were difference in L, a, and b measurement between three measurements.

Rate of yellowed leaves: Rate of yellowed leaves (%) = (number of yellowed leaves)/(total of leaves in stem) x 100

Data analysis: The experiment data was analyzed of variance (ANOVA) using Statgraphics 15.0 software at the reliability $P \leq 0.05$.

II. RESULTS AND DISCUSSION

A. Effects of chemical for pre-treatment of irradiated chrysanthemums and red carnation

Weight loss (%)

The weight loss of yellow chrysanthemum in control sample was the highest and significant with the others (Figure 1A). The weight loss was found at 7.97% by the pre-treatment with glucose-STS while that of the control sample (without pretreatment by glucose-STS) was determined about 33.64% after 6 days storage. In case of white chrysanthemum, the weight loss was also found at 7.65% for the pre-treatment with glucose-STS and 24.21% for the control sample. It could be concluded that pre-treatment of chrysanthemum by glucose-STS helps to minimize the weight loss after irradiation. However, the mentioned results did not meet for the carnation. There was no significant difference for the weight loss between treatments (Figure 1C). This result can be explained that carnation was tolerant to electron beam at 400 Gy, the same result was reported by Hayashi et al., 1998. Other results reported by Tanabe at al., 1994 also showed that carnation could be able to withstand up to 700 Gy [3].

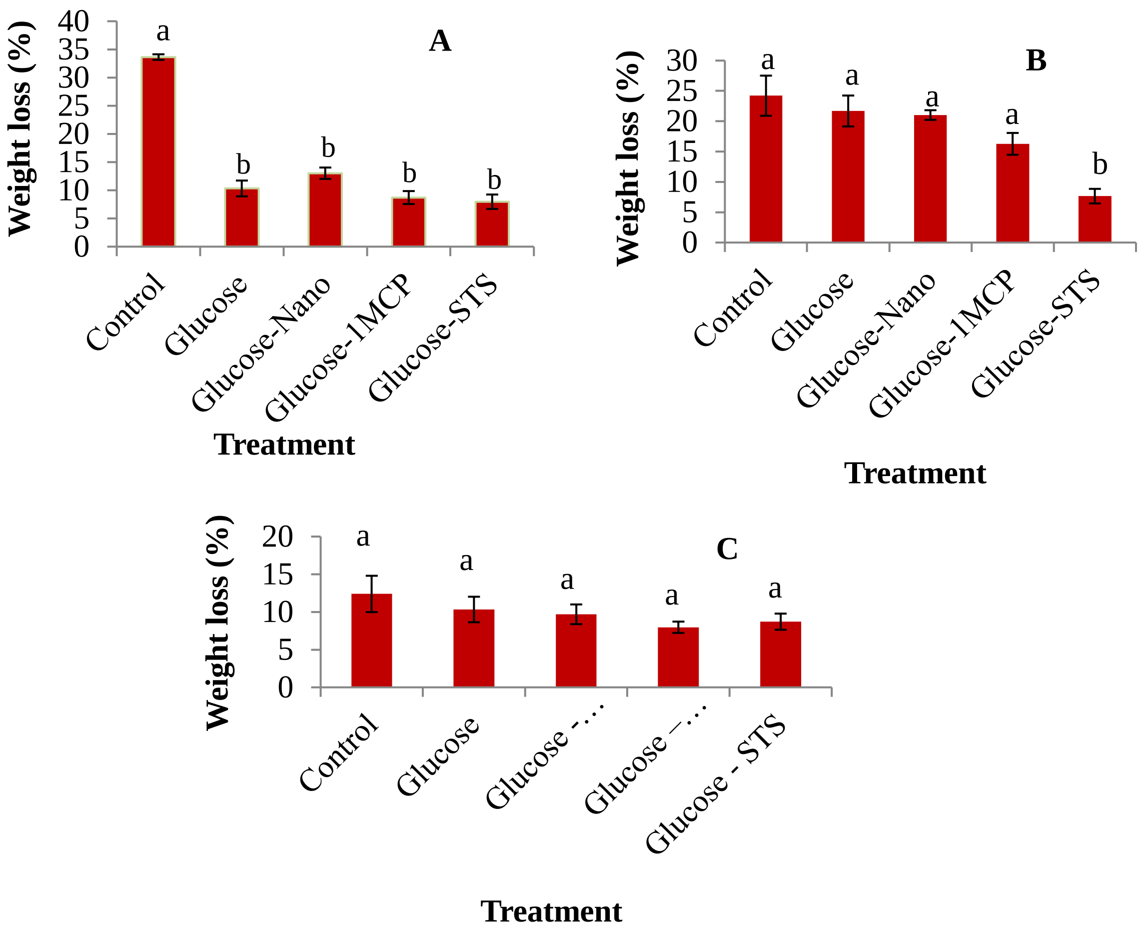

Fig. 1. Effect of commercial treatment on % weight loss of chrysanthemum and carnation after 6days storage at 27ºC, RH ~ 70-80% (A: Yellow Chrysanthemum, B: White chrysanthemum and C: carnation).

Flower color

The flowers of chrysanthemums were damaged by EB irradiation. The brightness of white and yellow chrysanthemum decreased after irradiation at a dose of 400 Gy (34.64% and 48.06% for yellow and white chrysanthemum respectively). The L value were increased in about 59.77% for yellow and 62.27% for white chrysanthemum when placing in 2% glucose + 0.024% STS, but this treatment did not affect to L value of carnation (Figure 2).

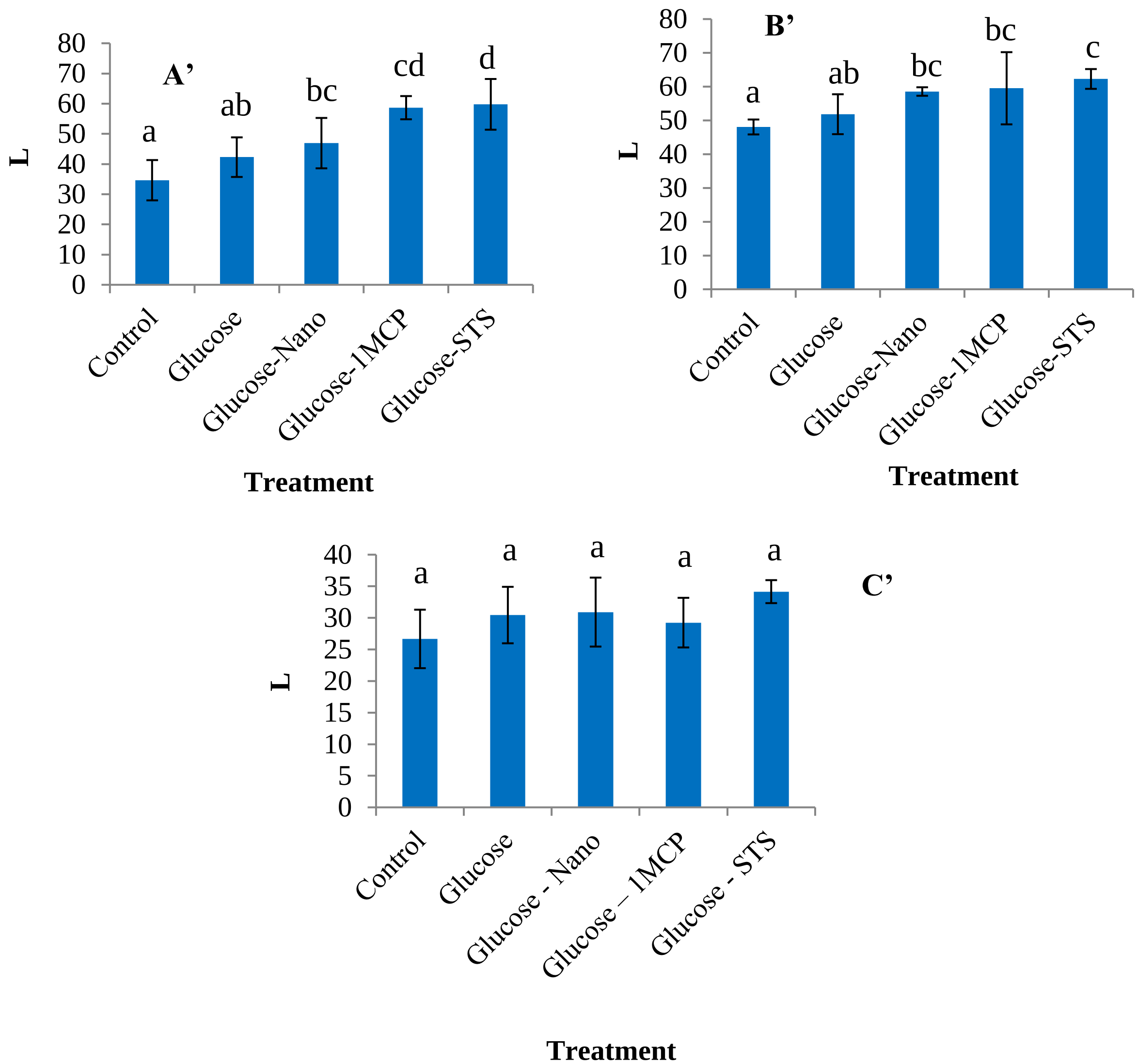

Fig. 2. Effect of commercial treatment on L value of chrysanthemum and carnation after 6 days storage at 27°C, RH = 70-80%
A’: Yellow chrysanthemum, B’: White chrysanthemum and C’: Carnation

Browning rate of leaves

The rate of yellowed leaves showed that pretreating cut flower with glucose - STS was the most effective to compare with the others. This rate was quite different between pre-treatment of glucose - STS (25.61%; 6.56%; 0.67%) and the control (74.52%; 41.5%; 6.36%) for white, yellow chrysanthemum and carnation respectively (Figure 3). While the results in treatment with 2% glucose; 2% glucose + 50ppm silver Nano or 2% glucose + 4nL/L 1-MCP showed insignificantly differences.

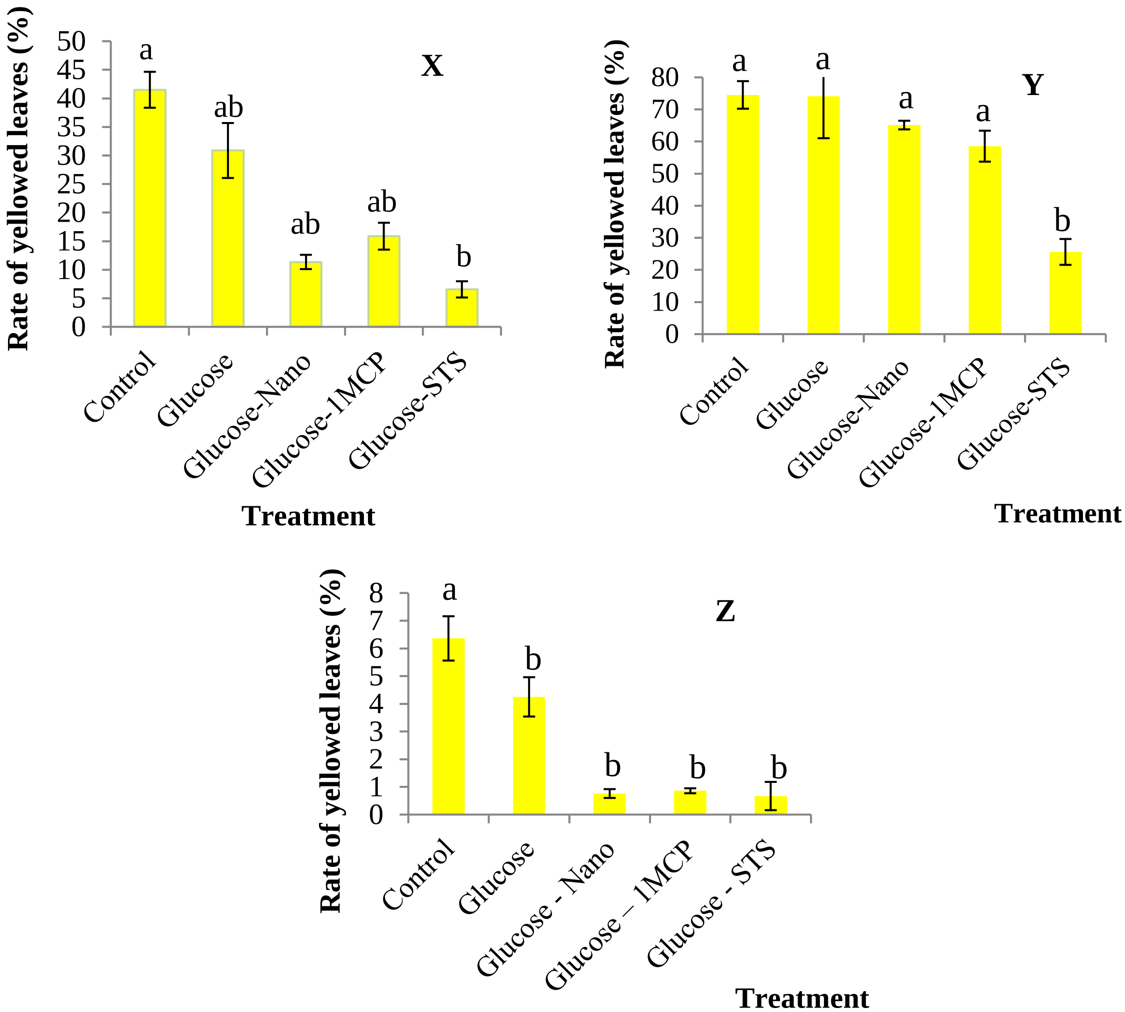

Fig. 3. Rate of yellowed leaves of chrysanthemum and carnation between treatments after 6 days storage at 27ºC, RH ~ 70-80%

X: Yellow chrysanthemum, Y: White chrysanthemum and Z: carnation

B. Effects of glucose-STS for pre-treatment cut flowers before irradiation at trade conditions

Yellow chrysanthemum

The results in this experience showed that shortening flowers shelf life has many causes. Using Glucose-STS to treat cut flowers before irradiation increased their radio-tolerance when keeping them under the trade conditions (5 days at 4-6ºC in transportation by air to destination). The weight loss, color of flower, leaf color and rate of yellowed leaves for yellow chrysanthemum were showed at Table II and III, for white chrysanthemum at Table IV and V in all treatments. In the Table II, means of L value did not significantly between G-S-400 Gy and control (non-treatment, and unirradiated). Meanwhile, there was the significant difference between the G-S-0 Gy and G-S-600 Gy (66.13% and 69.23%). The weight loss was not appeared during 6 days for all application. Sample G-S-0 Gy was the lowest weight loss (0.62%) while the control was 0.82% and can be kept until by eleventh day in vase. △E value was not significantly in other treatments. As a result of these application, the limitation in weight loss leads to increase of the vase life. Similar result was reported by Zencirkiran (2010), fresh weight loss decreased with application 2mM STS and extended the vase life of floret and spikes of cut Freesia "Cordura" [9].

Leaf color and rate of yellowed leaves showed at Table III. The differences in the color of leaves (L value and △E) were not significantly for all treatments. There was insignificant difference in the rate of yellowed leaves between the control sample and G-S-400 Gy; G-S-600 Gy. However, this parameter of the sample G-S-0 Gy (0.46%) was significantly in comparison with sample G-S-600 Gy (0.65%). Quality of yellow chrysanthemum in all application was not changed after eleventh day of storage and the vase life of yellow chrysanthemum was 6 days. The results indicated that glucose reduced injury to cut flowers. In addition, STS seemed to inhibit the ethylene production during storage indicating by the low rate of yellowed leaves of treated cut flowers.

Table II. Weight loss and flower color change of yellow chrysanthemum during time storage

Parameter	Sample	Time (days)					Mean of sample
		0	6	8	11	13	
L	Control	65.02±2.41	65.49±6.03	67.59±1.53	61.97±0.48	70.50±5.30	66.11AB
	G-S-0 Gy	65.64±2.52	62.83±2.63	66.51±1.69	66.40±3.26	68.69±2.83	66.01A
	G-S-400 Gy	68.49±2.11	64.57±5.47	66.21±4.31	69.78±2.79	72.74±9.74	68.36AB
	G-S-600 Gy	66.26±2.62	60.68±2.51	71.12±1.21	69.39±4.20	75.83±3.10	68.66B
Mean of Time		66.35ab	63.39a	67.86bc	66.88ab	71.94d	
Delta E	Control	55.87±3.39	55.54±7.10	57.03±2.91	50.73±2.57	57.57±3.66	55.35A
	G-S-0 Gy	56.30±2.47	59.36±2.01	57.03±3.24	56.26±1.90	53.47±2.72	56.48A
	G-S-400 Gy	60.64±2.31	60.19±6.39	55.67±4.74	57.54±7.54	56.62±7.42	58.13A
	G-S-600 Gy	58.03±4.80	55.36±3.54	59.67±3.26	53.81±4.95	55.20±6.45	56.38A
Mean of Time		57.71a	57.61a	57.35a	54.58a	55.71a	
Weight loss, gr/sprig (%)	Control	0.005	0.005	0.26±0.01	0.4±0.03	0.82±0.07	0.3B
	G-S-0 Gy	0.005	0.005	0.005	0.005	0.62±0.00	0.13A
	G-S-400 Gy	0.005	0.005	0.33±0.01	0.6±0.06	0.91±0.05	0.37C
	G-S-600 Gy	0.005	0.005	0.36±0.02	0.63±0.03	0.96±0.02	0.39C
Mean of Time		0.005a	0.005a	0.24b	0.41c	0.83c	

Mean values within same stage followed by the same letter are not significant different at P=0.05.
The weight loss (%) was translated to arcsin√%

Table III. Change in leaf color and the rate of yellowed leaves of yellow chrysanthemum during storage time.

Parameter	Sample	Time (days)					Mean of sample
		0	6	8	11	13	
L	Control	28.70±1.02	42.53±1.20	29.60±6.40	35.38±6.58	45.34±3.00	36.31A
	G-S-0 Gy	29.68±2.72	35.79±1.10	32.83±2.01	40.65±1.27	39.44±4.85	35.68A
	G-S-400 Gy	31.77±1.85	35.61±0.95	35.57±1.27	35.89±3.65	35.55±3.10	34.88A
	G-S-600 Gy	36.50±5.09	38.16±1.59	32.23±2.48	37.62±0.25	38.94±1.03	36.69A
Mean of Time		31.66a	38.02bc	32.55ab	37.39bc	39.82c	

Delta E	Control	24.29±0.43	18.40±4.14	23.66±5.08	19.14±4.79	11.26±0.73	19.35A
	G-S-0 Gy	23.13±1.75	17.86±0.39	20.14±1.94	13.73±0.28	13.98±2.93	17.77A
	G-S-400 Gy	21.14±1.75	17.69±1.01	18.47±5.13	15.43±2.46	17.63±2.74	18.07A
	G-S-600 Gy	17.81±3.09	15.92±1.00	20.31±2.20	14.88±0.16	13.35±0.68	16.45A
Mean of Time		21.59a	17.47b	20.64a	15.79bc	14.05c	
Yellowed rate of leaves, %	Control	0.005	0.005	0.62±0.06	1.03±0.06	1.57	0.64AB
	G-S-0 Gy	0.005	0.005	0.29±0.01	0.44±0.03	1.57	0.46B
	G-S-400 Gy	0.005	0.005	0.64±0.06	0.92±0.08	1.57	0.63AB
	G-S-600 Gy	0.005	0.005	0.51±0.02	1.18±0.05	1.57	0.65A
Mean of Time		0.005a	0.005a	0.52b	0.89c	1.57d	

Mean values within same stage followed by the same letter are not significant different at P=0.05.
The Yellowed rate of leaves (%) was translated to arcsin√%

White chrysanthemum

Using glucose + STS had a stronger effect on flower color and weight loss of white chrysanthemum. After 11 days in storage, the weight loss of sample G-S-400 Gy and G-S-600 Gy decreased slower than that of the control (Table IV). High water content of those flowers can be one indication to the radio-sensitivity due to the water radiolysis occurrence caused by the radiation [9]. L values of treated flower did not showed any significantly in all application. The sample G-S-0 Gy had the least weight loss (0.005%) while the control was 0.34%.

The leaf of chrysanthemum deteriorated first on the treatment by irradiation. The rate of yellowed leaves was 0.61% and 0.68% at sample G-S-400 Gy and G-S-600 Gy respectively. But they did not significantly with the control 0.45% (Table IV). The vase life of them was the same about 8 days. This considering the free radical that normally are produced during the irradiation of biological systems could be responsible for the senescence acceleration observed in the cut flowers, and free radicals of oxygen can damage lipids, proteins, carbohydrates and nucleic acids, whose injuriuos effects may be minimized by scavengers [10]. Glucose+STS is a good energy supply and probaly contributed to equilibrate the cell metabolism, increasing the defence against damages. This result is also agreement with the results explained by Zencirkiran (2010) [9].

Table IV. Change in flower color and weight loss of white chrysanthemum during storage time at trade conditions.

Parameter	Sample	Time (days)					Mean of sample
		0	6	8	11	13	
L	Control	65.01±6.11	63.31±3.00	53.33±2.09	66.61±3.32	70.23±8.42	63.69A
	G-S-0 Gy	75.22±0.46	55.69±2.10	52.31±1.25	66.49±0.69	70.77±6.58	64.09A
	G-S-400 Gy	78.10±0.86	56.08±4.39	53.63±5.62	63.90±7.39	70.97±5.03	64.54A
	G-S-600 Gy	81.59±0.47	59.58±7.69	52.35±9.29	57.15±3.70	72.98±4.06	64.73A
Mean of Time		74.98a	58.66c	52.9d	63.54b	71.24a	
Delta E	Control	23.48±6.16	25.74±4.42	16.36±1.50	34.81±2.39	21.83±4.76	24.44A
	G-S-0 Gy	33.41±1.14	22.31±1.24	16.12±0.54	36.17±0.54	23.67±2.09	33.02B
	G-S-400 Gy	35.88±0.92	20.91±1.10	18.35±2.45	17.24±2.08	21.21±4.06	22.72A
	G-S-600 Gy	37.34±0.6	21.97±0.47	20.83±3.60	24.70±0.5	33.79±3.77	27.73AB

Mean of Time		32.53a	22.73c	17.92d	28.23b	25.13c	
Weight loss, gr/sprig (%)	Control	0.005	0.005	0.34±0.02	0.34±0.02	0.46±0.03	0.23A
	G-S-0 Gy	0.005	0.005	0.005	0.005	0.36±0.02	0.076B
	G-S-400 Gy	0.005	0.005	0.005	0.29±0.01	0.49±0.01	0.16AB
	G-S-600 Gy	0.005	0.005	0.005	0.26±0.05	0.5±0.02	0.16AB
Mean of Time		0.005a	0.005a	0.089a	0.22a	0.45b	

Mean values within same stage followed by the same letter are not significant different at P=0.05.
The weight loss (%) was translated to arcsin√%

Table V. Leaf color and yellow leaves of white chrysanthemum during storage time at trade conditions.

Parameter	**Sample**	**Time (days)**					**Mean of sample**
		0	**6**	**8**	**11**	**13**	
L	Control	31.73±1.77	36.01±0.58	34.41±2.27	34.28±0.36	32.53±0.85	33.79AB
	G-S-0 Gy	31.75±1.29	32.07±1.58	30.40±2.29	35.94±0.49	38.25±3.82	33.68AB
	G-S-400 Gy	36.71±1.55	28.39±0.96	31.56±2.56	33.72±0.50	31.28±2.14	32.33B
	G-S-600 Gy	35.90±0.47	31.63±2.53	35.82±4.44	36.38±1.74	38.74±3.39	35.69A
Mean of Time		34.02a	32.03a	33.05a	35.08a	35.20a	
Delta E	Control	20.75±1.21	18.46±0.97	20.19±1.85	17.72±5.03	18.72±3.52	19.17AB
	G-S-0 Gy	17.08±1.29	21.21±1.46	21.33±1.83	15.34±3.26	13.0±3.08	17.59AB
	G-S-400 Gy	18.43±3.64	24.62±3.27	21.19±2.95	16.87±1.14	21.49±2.99	20.52A
	G-S-600 Gy	18.63±2.50	21.42±1.38	19.20±0.18	12.73±1.68	12.36±1.44	16.86B
Mean of Time		18.72ab	21.43bc	20.48b	15.66a	16.39a	
Yellowed rate of leaves, %	Control	0.005	0.005	0.005	0.92±0.08	1.3±0.07	0.45AB
	G-S-0 Gy	0.005	0.005	0.005	0.65±0.02	0.98±0.02	0.33A
	G-S-400 Gy	0.005	0.18±0.03	0.36±0.06	0.92±0.03	1.57	0.61B
	G-S-600 Gy	0.005	0.005	0.24±0.05	1.57	1.57	0.68B
Mean of Time		0.005a	0.049a	0.15a	1.02b	1.36c	

Mean values within same stage followed by the same letter are not significant different at P=0.05.
The Yellowed rate of leaves (%) was translated to arcsin√%

Red carnation

In the Table VI, the result showed that did not significantly about color, weight loss of carnation after 13 day storage. Yellowed rate of leaves increased significantly at the eleventh day. This result can be explained that basic biological processes of flowers such as respiration and transpiration continue after harvest. Sample G-S-600 Gy was significantly in comparison with the sample G-S-0 Gy. ΔE and weight loss were not significantly in all treatment. Same result was exported by Ichimura et al (2002), Celikel and Reid (2002) [11, 12], STS treatment had positively effect on vase life of carnation, matthiola, consolida, anthirrinum and delphinum.

Table VI. Flower color, weight loss and yellowed rate of leaves of carnation during storge time at trade conditions

Parameter	Sample	Storage period (days)					Mean of sample
		0	6	8	11	13	
L	Control	27.71±4.02	28.46±2.51	23.08±2.35	36.21±3.43	34.84±3.09	30.06A
	G-S-0 Gy	29.02±1.32	19.4±1.98	22.90±2.57	22.56±2.67	29.24±2.03	24.62B
	G-S-400 Gy	31.85±2.16	23.06±5.27	19.35±1.56	26.88±2.86	25.85±4.26	25.39B
	G-S-600 Gy	29.76±0.74	21.66±5.18	22.42±5.33	30.53±7.14	40.06±4.10	28.89A
Mean of Time		29.59bc	23.15a	21.94a	29.05b	32.49c	
Delta E	Control	44.37±10.79	51.27±5.31	53.77±1.37	54.51±1.36	52.37±1.27	51.26A
	G-S-0 Gy	52.20±2.50	52.57±1.56	50.65±1.52	48.77±0.95	46.54±5.05	50.15A
	G-S-400 Gy	52.26±0.36	52.97±0.65	52.81±2.76	49.36±1.55	48.63±2.93	51.21A
	G-S-600 Gy	48.07±7.21	51.90±1.40	51.48±0.55	47.36±3.46	51.37±1.82	50.04A
Mean of Time		49.22a	52.18a	52.18a	49.99a	49.73a	
Weight loss, gr/sprig (%)	Control	0.005	0.005	0.005	0.29±0.03	0.29±0.03	0.12A
	G-S-0 Gy	0.005	0.005	0.005	0.005	0.29±0.03	0.06A
	G-S-400 Gy	0.005	0.005	0.005	0.42±0.07	0.42±0.07	0.17A
	G-S-600 Gy	0.005	0.005	0.005	0.29±0.03	0.29±0.03	0.12A
Mean of Time		0.005a	0.005a	0.005a	0.25ab	0.32ab	
Yellow rate of leaves, %	Control	0.005	0.005	0.005	0.1±0.02	0.22±0.06	0.067AB
	G-S-0 Gy	0.005	0.005	0.005	0.005	0.18±0.01	0.04A
	G-S-400 Gy	0.005	0.005	0.005	0.2±0.04	0.21±0.02	0.085AB
	G-S-600 Gy	0.005	0.005	0.005	0.22±0.02	0.22±0.05	0.091B
Mean of Time		0.005a	0.005a	0.005a	0.13b	0.21b	

Mean values within same stage followed by the same letter are not significant different at P=0.05.
The weight loss and yellowed rate of leaves (%) was translated to arcsin√%

Fig. 4. Vase-life of cut flowers after 11 days under trade conditions : from left to right respectively : control, Glucose + STS + 0 Gy, Glucose + STS + 400 Gy and Glucose + STS + 600 Gy.

III. CONCLUSIONS

- The fresh cut flowers were EB irradiated as quarantine treatment. The radiation tolerance of the irradiated flowers was significantly increased and their commercial quality was remained by the pre-treatment with 2% glucose and 0.024% STS solutions.

- Using 2% glucose and 0.024% STS for pre-treatment before irradiation could also improve the quality of cut flowers under the trade conditions (at the temperature of 4-6^{o}C during 5 days for transportation by air).

- The vase-life prolong to 6 days for yellow chrysanthemum, 8 days for white chrysanthemum and 8-10 days for carnation (similar to their no-treated flowers), suggested that this pretreatment method could be applied for improving the radiation tolerance and maintenance the commercial value of radiation quarantined flowers.

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