



Article

Characterization of Petal Senescent Types in Cut Dahlia and Extension of Their Vase Life by Treatment with Silver Thiosulfate Complex Followed by Glucose Plus Germicides

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Abstract: The vase life of cut dahlia (*Dahlia × hortensis* Guillaumin) flowers is generally short. Petal senescence is known to be divided into petal wilting, withering, and abscission. Petals with ongoing wilting were isolated from the inflorescence and supplied with water using 10 cultivars to clarify the type of petal senescence of dahlia. Based on the recovery of fresh weight and appearance of petals, senescent types could be categorized into three types: petal senescence in two, six, and two cultivars were categorized into abscission, abscission with withering, and wilting types, respectively. The pulse treatment with silver thiosulfate complex (STS) significantly extended the vase life of 7 out of 10 cultivars. Moreover, pulse treatment with STS followed by continuous treatment with GLA, which was comprised of glucose, isothiazolinone germicide, and aluminum sulfate, increased the relative fresh weight of the five dahlia cultivars more than pulse treatment with STS. Therefore, this combined treatment is available to extend the vase life of cut dahlia, irrespective of the senescent types.

Keywords: abscission; cut flowers; ethylene inhibitors; wilting; withering



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1. Introduction

Postharvest performance is very important for cut flowers. The vase life of cut flowers has been associated with petal longevity. There are two types of petal senescence: petal wilting and abscission. Moreover, van Doorn and Woltering [1] stated that petal senescence could be categorized as wilting, withering, and abscission. Withering means color changes and dehydration of petals.

Dahlia (*Dahlia × hortensis* Guillaumin) belongs to Asteraceae and originated in the highlands of Central and South America. There is marked variation in the size, color, and floral shape in the flowers of the dahlia cultivars. The dahlia has mainly been used as a bedding plant. Recently, dahlia flowers have become used for cut flowers in Japan, but the vase life of cut dahlia flowers is significantly short [2,3].

It has been recognized that the vase life of cut dahlia flowers is terminated with petal wilting or discoloration [2,4]. However, Azuma et al. [5] reported that petal abscission was observed in some cultivars, such as 'Carnelian' and 'Namahage Cute', when cut dahlia flowers are held at 100% relative humidity. Moreover, when cut dahlia flowers are held at 70% relative humidity, petals exhibit wilting without a visible drop, but petals are abscised with a gentle touch of petals. This suggests that the abscission layer was formed in these cultivars, but the presence of other petals inhibited the petal drop. However, it is challenging to completely eliminate the possibility that the petals have been artificially abscised by touching them.

Pulse treatments with ethylene inhibitors, such as silver thiosulfate complex (STS), aminoethoxyvinylglycine, and aminoxyacetic acid, extend the vase life of ethylene-sensitive flowers, including carnation [6,7] and *Delphinium* [8]. Dahlia flowers are sensitive

to ethylene [4], although there are differences in their sensitivity to ethylene among the cultivars [5]. Continuous treatment with 1-methylcyclopropene (1-MCP), an inhibitor of ethylene action, extends the vase life of cut dahlia flowers [5]. These findings suggest that endogenous ethylene is involved in the senescence of dahlia petals. Since 1-MCP is a volatile compound, flowers should be kept sealed to be fully effective for the continuous treatment of 1-MCP. Therefore, continuous treatment with 1-MCP is challenging for practical applications. However, pulse treatment with 1-MCP did not significantly extend the vase life of cut dahlia flowers [5]. STS has been widely used in ethylene-sensitive cut flowers to extend vase life. However, pulse treatment with STS did not extend the vase life of the dahlia ‘Karma Thalia’ and ‘Kokucho’ cultivars [4,9]. Azuma et al. [5] recently reported that pulse treatment with STS extended the vase life of cut ‘Carnelian’ exhibiting petal abscission, but did not significantly extend that of cut ‘Port Light Pair Beauty’ exhibiting petal wilting. These results suggest that STS treatment might be effective in cultivars exhibiting petal abscission, but might not be in cultivars exhibiting petal wilting.

The water relations of cut flowers are often impaired [10]. Bacterial proliferation impairs the water relations of cut flowers. The vase life of some cut flowers, including rose and gerbera, is extended by treatment with antimicrobial compounds [11,12]. Continuous treatment with antimicrobial compounds extends the vase life of cut dahlia flowers in 3 out of 10 cultivars [2].

Treatment with glucose or sucrose promotes flower opening and extends the vase life of cut flowers in many species, including rose [12], carnation [13], and *Eustoma grandiflorum* [14]. The senescence of petals progresses from the outer whorl to the inner whorl in dahlia inflorescence. When dahlia flowers are harvested at standard harvest maturity, the petals of the outer whorl mature, but those of the inner whorl are immature. The vase life of cut dahlia flowers is extended by treatment with sugars combined with antimicrobial compounds [2,15]. This effect is mainly due to the promotion of petal growth at the inner whorl [2]. GLA comprises glucose, isothiazolinone germicide, and aluminum sulfate [3,16]. Continuous treatment with GLA extends the vase life of cut flowers in many plant species including dahlia flowers [3].

In ethylene-sensitive flowers with many floral buds, such as *Gypsophila paniculate* [17], *E. grandiflorum* [14], snapdragon [18], and sweet pea [19], their vase life is extended by pulse treatments with STS in combination with sugars plus germicides. Moreover, the vase life of cut carnation [20] and snapdragon [21] was extended by pulse treatment with STS, followed by continuous treatment with sucrose plus germicides.

Here, we first investigated the postharvest characteristics of cut dahlia flowers using isolated petals to clarify whether petal senescence is due to wilting or abscission. Moreover, we investigated the effect of STS treatment on the vase life of cut dahlia flowers using 10 cultivars to clarify whether STS treatment generally extends the vase life of cut dahlias. Additionally, we evaluated the effect of STS in combination with GLA.

2. Materials and Methods

2.1. Plant Material

Cut dahlia (*Dahlia × hortensis* Guillaumin) flowers of the following 10 cultivars were used: ‘Agitate’, ‘Carnelian’, ‘Garnet’, ‘Kamakura’, ‘LaLaLa’, ‘Micchan’, ‘Moon Waltz’, ‘Namahage Cute’, ‘Port Light Pair Beauty’, and ‘Rose Quartz’. These cut flowers were obtained from growers in Asahi, Chiba Prefecture, Japan, in March, 2020. The flowers were harvested when the first row of ray petals on the outside of the flowers opened. Asteraceae flowers have two types of florets: ray and tubular ones. Most of the florets in the dahlia flowers are ray florets. Hereafter, the term “petal” means ray petal.

The cut ends of the stems were immersed in tap water after the harvest and stored at approximately 10 °C to investigate senescent types of the dahlia cultivars. The flowers placed in a bucket with tap water were transported to our institute, which took approximately 2 h. After arrival, all leaves of the cut flowers were removed, and the stems were recut to 30 cm and used for experiments.

Flowers were not supplied with water after the harvest to investigate the effect of STS and GLA. The flowers were placed in buckets covered with polyethylene bags to prevent water loss and transported to our institute. The time required to reach our institute and the treatment of the cut flowers were as described above.

2.2. Fresh Weight and Longevity in Isolated Petals and Petals Remaining in Inflorescence

Twelve flowers were used in each cultivar. The four cut flowers were placed in a vessel containing 0.2 mL L^{-1} CMIT/MIT solution (Khason CG; Rohm and Haas Japan, Tokyo, Japan) to inhibit bacterial proliferation. A total of three vessels were used. CMIT/MIT solution contains 11.3 g L^{-1} 5-chloro-2-methyl-4-isothiazolin-3-one and 3.9 g L^{-1} 2-methyl-4-isothiazolin-3-one as active ingredients. The six petals were isolated from the first whorl of different inflorescences and put in 0.2 mL L^{-1} CMIT/MIT solution to measure the fresh weight of petals. The fresh weight of the petals was measured daily. Six petals were detached daily from different inflorescences to measure the fresh weight of petals remaining in the inflorescence. The petal was detached every other day from one flower to avoid adverse effects on petal fresh weight and longevity. The petals and inflorescences were kept at $23 \text{ }^{\circ}\text{C}$, 70% relative humidity, and $10 \text{ } \mu\text{mol m}^{-2} \text{ s}^{-1}$ irradiance using cool-white fluorescent lamps under a 12-h photoperiod.

2.3. Effect of Water Supply on the Recovery of Wilting Petals Isolated from Inflorescence

The cut flowers of the 10 cultivars were kept as described in Section 2.2. When petal wilting was observed as shown in Figure 1, the petals at the outermost whorl were isolated and placed in CMIT/MIT solutions. Then, the fresh weight of the isolated petals was measured daily. The number of cut flowers and repetitions, the fresh petal weight measurement, and the environmental conditions for the detached petals and cut flowers were the same as described in Section 2.2.



Figure 1. Inflorescence with petals in the wilting process. (a) ‘Carnelian’; (b) ‘Kamakura’.

2.4. STS Treatment

STS solution at 0.2 mM was prepared by mixing equal volumes of AgNO_3 and $\text{Na}_2\text{S}_2\text{O}_3 \cdot 5\text{H}_2\text{O}$ at a molar ratio of 1:8. The above 10 cultivars were used for the experiment. Twelve flowers in each cultivar were used for the experiment, and the six cut flowers were put into 300 mL of 0.2 mM STS solution. The other flowers were held in distilled water, and used for the control. The flowers were held at $23 \text{ }^{\circ}\text{C}$, 70% relative humidity in the dark for 16 h. Then, flowers were placed individually in test tubes containing 100 mL of distilled water and kept at $23 \text{ }^{\circ}\text{C}$, 70% relative humidity, and $10 \text{ } \mu\text{mol m}^{-2} \text{ s}^{-1}$ irradiance using cool-white fluorescent lamps under a 12-h photoperiod. The fresh weight of the cut flowers was measured daily. The relative fresh weight was calculated as the percentage of the daily fresh weight to the initial fresh weight. The vase life of the flower was determined as the period from the start of the experiment to the time when petal wilting in the florets of three whorls or the abscission of more than 10 petals was observed.

2.5. Combined Treatment with STS and GLA

Dahlia ‘Garnet’, ‘Kamakura’, ‘LaLaLa’, ‘Micchan’, and ‘Moon Waltz’ flowers were cut into 30 cm length. Twenty flowers in each cultivar were used. Ten flowers in each cultivar were held in 0.2 mM STS solution, prepared as described above. The other 10 flowers were held in distilled water. The flowers were held at 23 °C in the dark for 16 h. The five flowers treated with STS and the five flowers held in distilled water were individually transferred to a GLA solution containing 10 g L⁻¹ glucose, 0.5 mL L⁻¹ CMIT/MIT, and 50 mg L⁻¹ aluminum sulfate [3,16]. GLA was designated from the first letters of glucose, CMIT/MIT whose former commercial name was Legend MK, and aluminum sulfate. The other 10 flowers were transferred to 0.1 mL L⁻¹ CMIT/MIT solution. The measurement of relative fresh weight and the evaluation of vase life were performed as described in Section 2.5.

2.6. Statistical Analysis

Data were analyzed using a two-way analysis of variance (ANOVA), *t*-test, and Tukey–Kramer multiple range test using SigmaPlot software (v.12.5; Systat Software, San Jose, CA, USA).

3. Results

3.1. The Fresh Weight of Petals Remaining in Inflorescence and Isolated Petals

The fresh weight of petals remaining in the inflorescence in ‘Micchan’ was almost constant throughout the experimental period (Figure 2). The fresh weight of petals remaining in the inflorescence in the other nine cultivars slightly increased during the first 1–3 days and decreased thereafter.

The fresh weight of the isolated petals in ‘Agitate’, ‘Kamakura’, and ‘Micchan’ was almost constant throughout the experimental period. The fresh weight of isolated petals in ‘Carnelian’ gradually decreased with time. In the other six cultivars, the fresh weight of isolated petals increased slightly during the first 3–4 days and decreased afterwards.

Longevity was significantly longer in the isolated petals than in the petals remaining in inflorescence in eight cultivars (Table 1). Remarkably, the longevity of isolated petals was two times longer than that of un-isolated petals in ‘Carnelian’, ‘LaLaLa’, and ‘Namahage Cute’.

Table 1. Longevity of isolated petals in 10 dahlia cultivars.

Cultivar	Petal Longevity (Days)				S Significance		
	Inflorescence		Isolated				
Agitate	4.7	±	0.4	5.5	±	0.2	NS
Carnelian	2.5	±	0.2	5.0	±	0.4	***
Garnet	4.2	±	0.2	4.8	±	0.2	*
Kamakura	3.2	±	0.2	4.5	±	0.2	***
LaLaLa	2.7	±	0.2	5.7	±	0.2	***
Micchan	4.7	±	0.2	5.2	±	0.5	NS
Moon Waltz	4.5	±	0.2	5.8	±	0.2	***
Namahage Cute	2.8	±	0.4	7.0	±	0.0	***
Port Light Pair Beauty	3.0	±	0.0	4.0	±	0.0	***
Rose Quartz	3.5	±	0.2	4.7	±	0.2	**
Two-way ANOVA							
Cultivar			<0.001				
Petal type			<0.001				
Cultivar × Petal type			<0.001				

Values are means of six replicates ± SE. NS indicates not significant, and *, **, and *** indicate significant differences at $p < 0.05$, $p < 0.01$, and $p < 0.001$, respectively, by *t*-test.

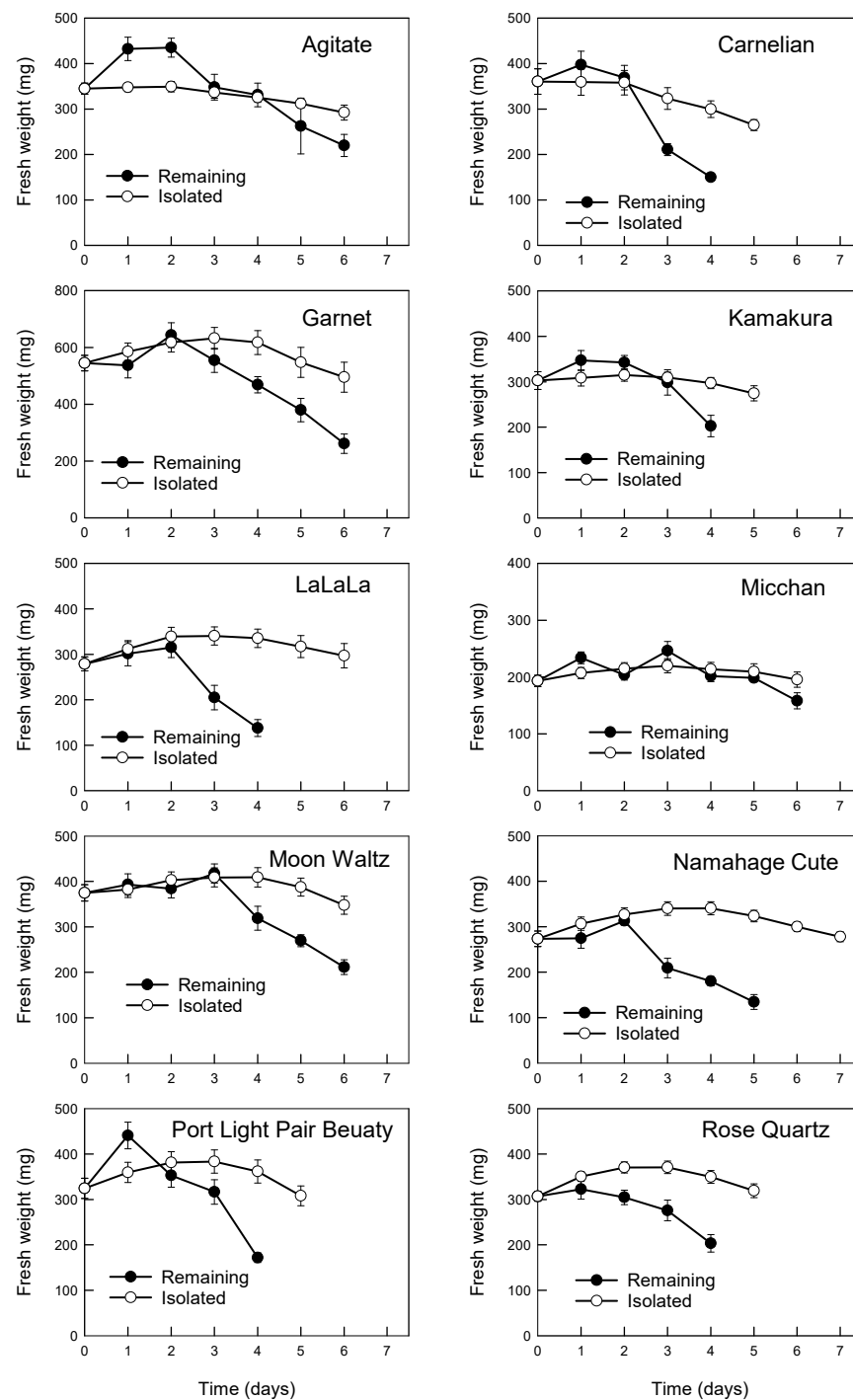


Figure 2. The fresh weight of isolated petals and those remaining in the inflorescence of 10 dahlia cultivars. Values are the means of six replicates \pm SE.

3.2. The Fresh Weight of Wilting Petals Isolated from the Inflorescence and Remaining in the Inflorescence

The wilting of petals in ‘Agitate’, ‘Carnelian’, ‘Garnet’, ‘Kamakura’, ‘LaLaLa’, ‘Micchan’, ‘Moon Waltz’, ‘Namahage Cute’, ‘Port Light Pair Beauty’, and ‘Rose Quartz’ was observed on day 5, 3, 4, 3, 3, 6, 5, 6, 4, and 5, respectively.

The fresh weight of wilting petals in the inflorescence in ‘Agitate’ and ‘Micchan’ decreased moderately on day 1, and it decreased sharply thereafter (Figure 3). In the other eight cultivars, the fresh weight of petals remaining in inflorescence obviously decreased during the first day.

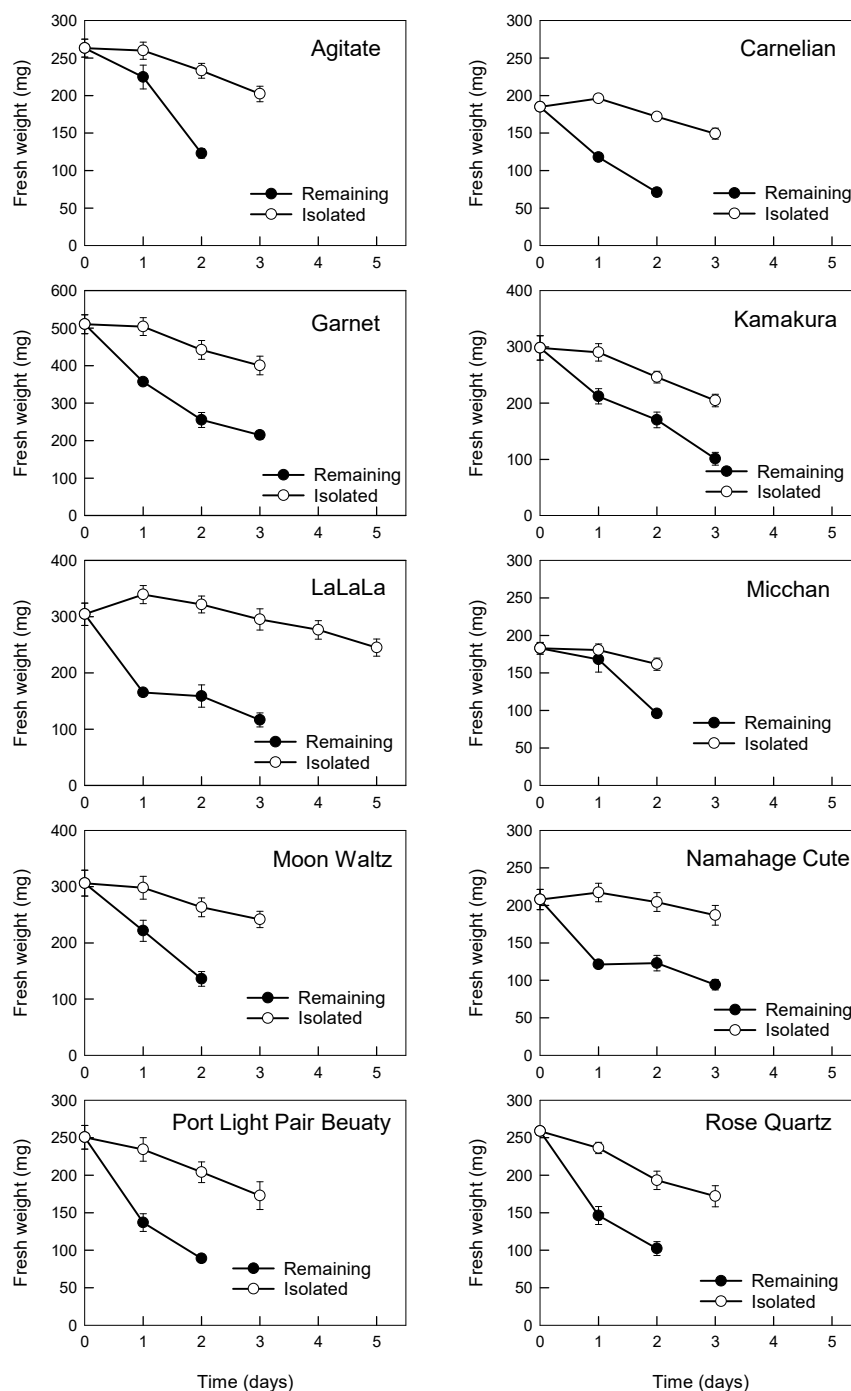


Figure 3. The fresh weight of petals isolated from inflorescence in the wilting process and those remaining in the inflorescences of 10 dahlia cultivars. Values are the means of six replicates \pm SE.

In contrast, the fresh weight of isolated petals in ‘Carnelian’, ‘LaLaLa’, and ‘Namahage Cute’ increased during the first day, and decreased thereafter. Additionally, the fresh weight of the isolated petals in ‘Agitate’, ‘Garnet’, ‘Kamakura’, ‘Micchan’, and ‘Moon Waltz’ was almost constant during the first day, and decreased thereafter. In ‘Port Light Pair Beauty’, and ‘Rose Quartz’, the fresh weight of petals gradually decreased with time. As shown in Figure 4, petals of ‘Agitate’, ‘Carnelian’, ‘Garnet’, ‘Kamakura’, ‘LaLaLa’, ‘Moon Waltz’, and ‘Namahage Cute’ exhibited bending due to petal wilting. The supplement of water induced recovery from wilting in these cultivars.

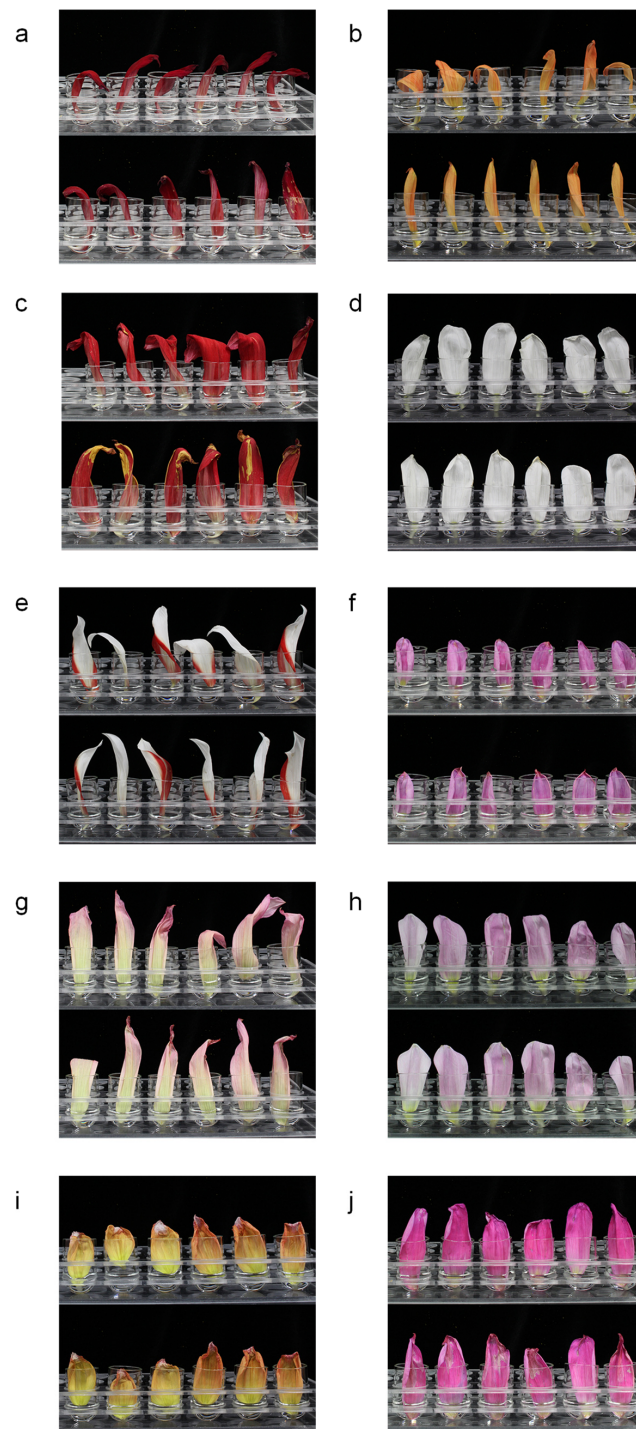


Figure 4. The effect of water supply on recovery from wilting in isolated petals of 10 cultivars. The upper panel shows petals on the day of wilting, and the lower panel shows petals 1 day after the start of water supply in each cultivar. (a) 'Agitate'; (b) 'Carnelian'; (c) 'Garnet'; (d) 'Kamakura'; (e) 'LaLaLa'; (f) 'Micchan'; (g) 'Moon Waltz'; (h) 'Namahage Cute'; (i) 'Port Light Pair Beauty'; (j) 'Rose Quartz'.

3.3. The Effect of STS on the Vase Life of Cut Dahlia

The uptake of STS solutions in 'Agitate', 'Carnelian', 'Garnet', 'Kamakura', 'LaLaLa', 'Micchan', 'Moon Waltz', 'Namahage Cute', 'Port Light Pair Beauty', and 'Rose Quartz' flowers was 10.8, 8.3, 9.2, 12.2, 8.7, 7.5, 8.7, 18.3, 6.7, and 5.8 mL per flower, respectively.

The relative fresh weight of control flowers in these 10 cultivars increased during the first 1–3 days, and decreased thereafter (Figure 5). Treatment with STS suppressed decreases in the relative fresh weight of ‘Agitate’, ‘Carnelian’, ‘Garnet’, and ‘Namahage Cute’. In ‘Moon Waltz’, the STS treatment suppressed an increase in the relative fresh weight but suppressed these decreases. However, STS treatment only slightly affected the relative fresh weight in ‘Kamakura’, ‘Micchan’, ‘Port Light Pair Beauty’, and ‘Rose Quartz’.

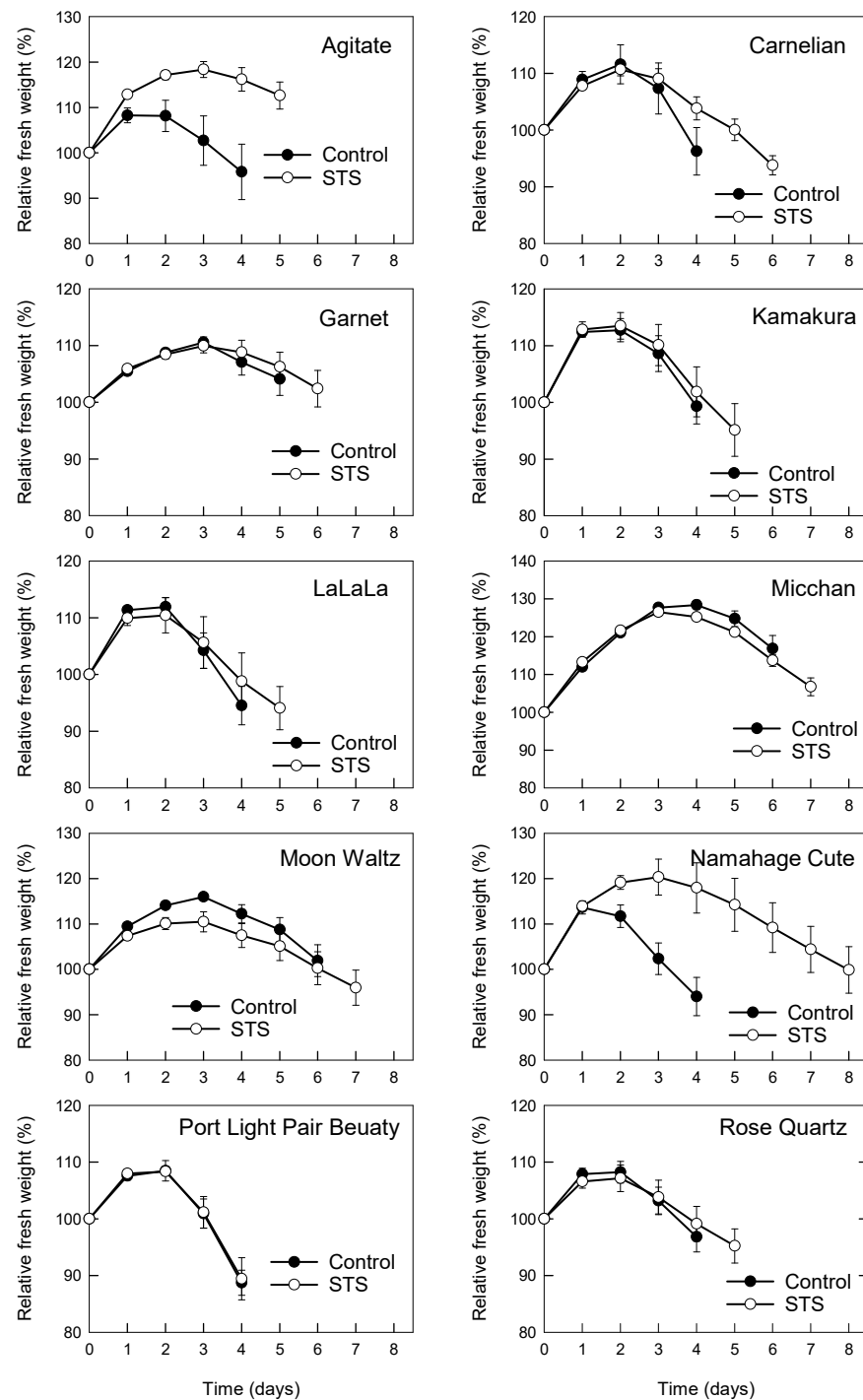


Figure 5. The relative fresh weight of cut dahlia flowers after treatment with STS of 10 dahlia cultivars. Values are the means of six replicates \pm SE.

It was shown with ANOVA that the STS treatment significantly extended the vase life of cut dahlia flowers (Table 2). The vase life of cut flowers in 7 out of 10 cultivars was

significantly extended by the STS treatment. Remarkably, the extension of vase life by STS treatment was pronounced in ‘Agitate’, ‘Carnelian’, ‘Kamakura’, ‘Moon Waltz’, and ‘Namahage Cute’ (Table 2, Figure 6).

Table 2. Effect of pulse treatment with STS on the vase life of 10 dahlia cultivars.

Cultivar	Vase Life (Days)						Significance
	Control			STS			
Agitate	3.2	±	0.3	4.8	±	0.2	***
Carnelian	3.2	±	0.4	5.5	±	0.2	***
Garnet	4.3	±	0.2	5.3	±	0.3	*
Kamakura	3.0	±	0.4	4.3	±	0.3	*
LaLaLa	3.7	±	0.2	4.3	±	0.3	NS
Micchan	5.8	±	0.2	6.6	±	0.4	NS
Moon Waltz	5.8	±	0.4	7.2	±	0.3	*
Namahage Cute	3.3	±	0.3	7.0	±	0.6	**
Port Light Pair Beauty	3.2	±	0.2	3.5	±	0.3	NS
Rose Quartz	3.5	±	0.2	4.7	±	0.2	**
Two-way ANOVA							
Cultivar				<0.001			
STS				<0.001			
Cultivar × STS				<0.001			

Values are the means of six replicates ± SE. NS indicates not significant, and *, **, and *** indicate significant difference at $p < 0.05$, $p < 0.01$, and $p < 0.001$, respectively, by *t*-test.

3.4. The Effect of STS in Combination with Glucose and Antimicrobial Compounds

The uptake of STS solutions in ‘Garnet’, ‘Kamakura’, ‘LaLaLa’, ‘Micchan’, and ‘Moon Waltz’ flowers was 13.8, 17.0, 11.0, 11.0, and 10.5 mL per flower, respectively.

The relative fresh weight of these five cultivars in control increased during the first 1–2 days, and decreased thereafter (Figure 7). The relative fresh weight of ‘Kamakura’ and ‘Micchan’ flowers was increased by the STS treatment more than by the GLA treatment. The increases in the relative fresh weight in these cultivars were promoted more by the combined treatment with STS and GLA than by the STS alone and the GLA alone.

ANOVA results showed that the STS treatment extended the vase life of four cultivars at a significance level of 0.1% (Table 3). It also showed that the GLA treatment extended the vase life of one and two cultivars at significance levels of 0.1 and 5%, respectively. The combined treatment with STS and GLA significantly extended the vase life more than GLA alone in ‘LaLaLa’, ‘Micchan’, and ‘Moon Waltz’. The interaction between STS and GLA in the four cultivars for vase life was not significant, indicating that the effect of STS on the extension of vase life was independent of that of glucose plus germicides.

Table 3. Effect of pulse treatment with STS followed by continuous treatment with GLA on the vase life of five dahlia cultivars.

Treatment	Vase Life (Days)																			
	Garnet			Kamakura			LaLaLa			Micchan			Moon Waltz							
Control	3.8	±	0.4	a	2.8	±	0.4	a	3.3	±	0.4	a	3.8	±	0.2	a	5.0	±	0.4	a
STS	4.8	±	0.7	ab	4.8	±	0.2	b	5.4	±	0.4	b	7.6	±	0.2	c	7.0	±	0.5	bc
GLA	5.0	±	0.3	ab	4.0	±	0.3	ab	3.2	±	0.4	a	6.2	±	0.4	b	5.6	±	0.2	ab
STS + GLA	6.2	±	0.6	b	5.2	±	0.5	b	5.8	±	0.4	b	8.6	±	0.2	c	7.4	±	0.4	c
Two-way ANOVA																				
STS					0.062				<0.001				<0.001				<0.001			<0.001
GLA					0.026				0.041				0.800				<0.001			0.242
STS × GLA					0.856				0.284				0.450				0.021			0.810

Values are the mean of six replicates ± standard errors. Different letters in each column indicate a significantly difference ($p < 0.05$) by the Tukey–Kramer multiple range test.

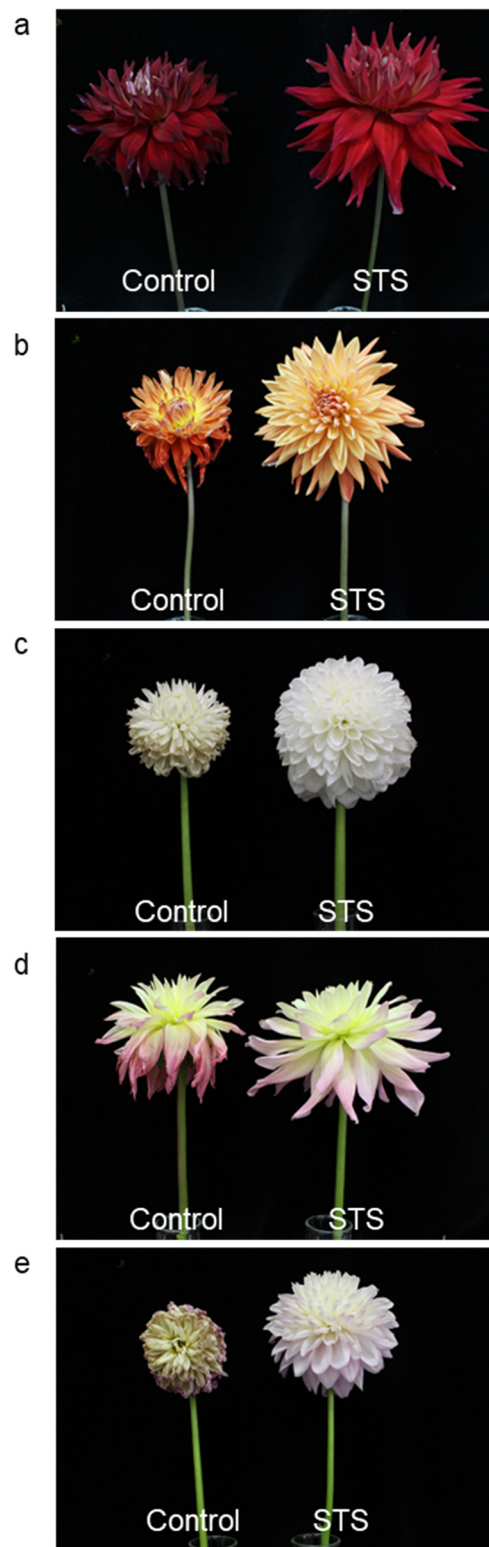


Figure 6. The effect of pulse treatment with STS on the vase life of cut dahlia. (a) 'Agitate' on day 3; (b) 'Carnelian' on day 4; (c) 'Kamakura' on day 5; (d) 'Moon Waltz' on day 5; (e) 'Namahage Cute' on day 7.

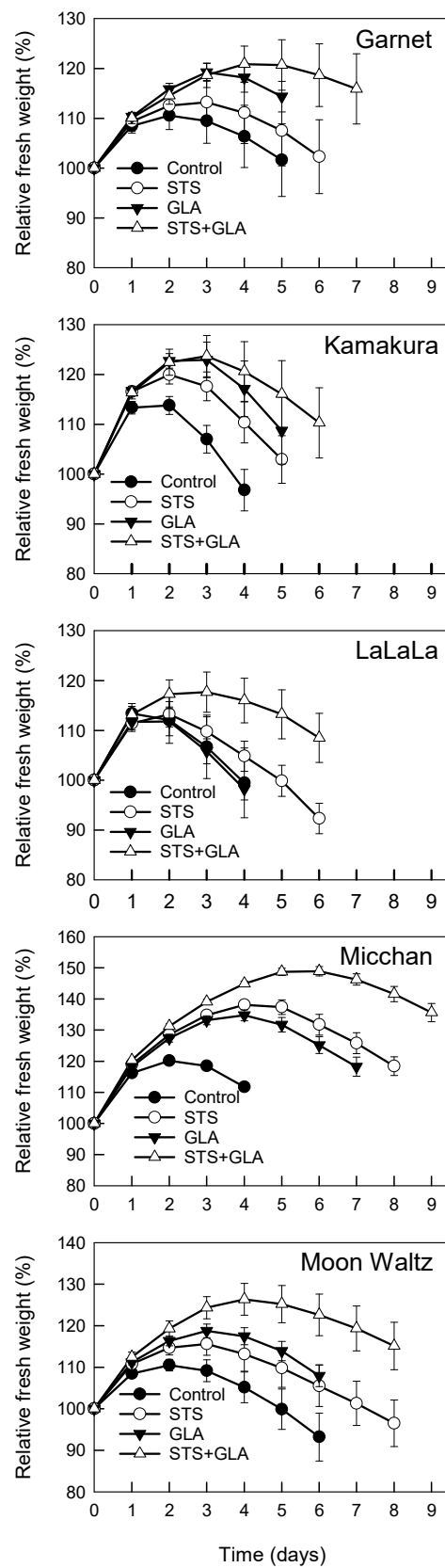


Figure 7. The relative fresh weight of five dahlia cultivars pulse-treated with STS followed by continuous treatment with GLA. Values are the means of five replicates \pm SE.

In ‘Micchan’ and ‘Moon Waltz’, the wilting of the outer petals was suppressed, but the growth of the inner petals was not promoted by the STS treatment. In contrast, the growth

of the inner petals was promoted, but the wilting of the outer petals was not suppressed by GLA treatment. The combined treatment with STS and GLA delayed the wilting of the outer petals and promoted the growth of the inner petals, resulting in the increase in fresh weight and extension of vase life (Figure 8). Similar results were obtained for ‘Kamakura’ and ‘LaLaLa’ cultivars.

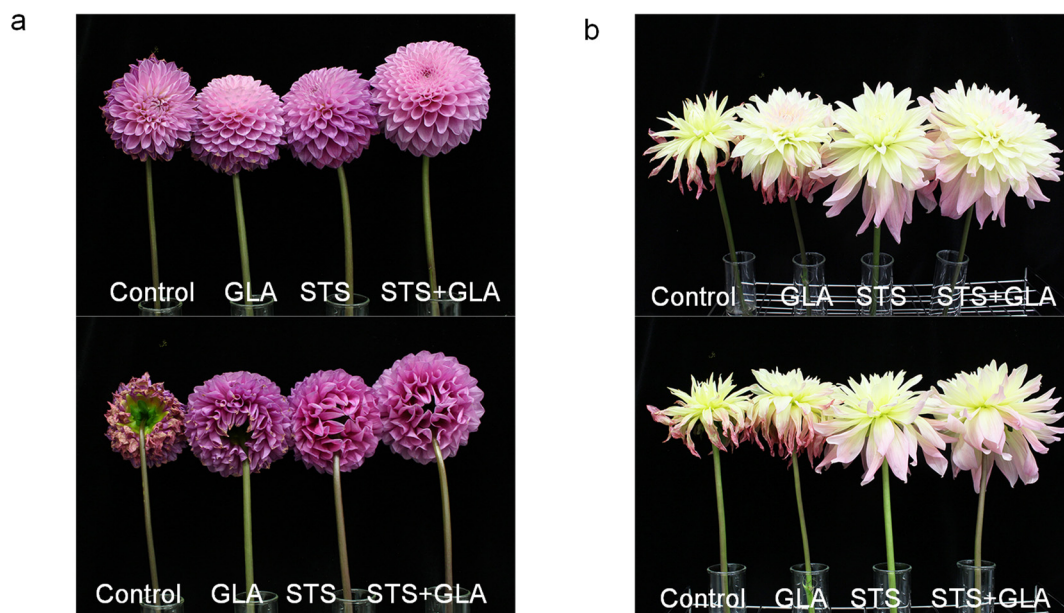


Figure 8. Effect of pulse treatment with STS followed by continuous treatment with GLA on the vase life of cut dahlia. (a) ‘Micchan’ on day 7; (b) ‘Moon Waltz’ on day 7. The upper and lower panels are focused on the inner and outer petals, respectively, in each cultivar.

4. Discussion

Here, we first investigated the postharvest characteristics of cut dahlia flowers using isolated petals to clarify whether petal senescence is due to the dahlia flowers wilting or abscission. Next, we investigated the effect of STS treatment on the vase life of cut dahlia flowers using 10 cultivars to clarify whether STS treatment generally extends the vase life of cut dahlias. Moreover, the effect of STS in combination with GLA on the vase life of cut dahlia flowers was investigated.

The senescent types of cut dahlia flowers have been categorized into petal abscission and petal wilting [5]. In our study, the fresh weight of isolated petals in the 10 cultivars did not decrease sharply. In contrast, there was an obvious decrease in the fresh weight of petals remaining in the inflorescence in seven cultivars, ‘Carnelian’, ‘Garnet’, ‘Kamakura’, ‘LaLaLa’, ‘Moon Waltz’, ‘Namahage Cute’, and ‘Port Light Pair Beauty’ (Figure 2). Moreover, the petals of some cultivars, including ‘Carnelian’, ‘Moon Waltz’, and ‘Namahage Cute’, dropped with gentle touching [5]. Therefore, petal abscission may occur in the above seven cultivars for which the fresh weight of petals obviously decreased. This explanation is supported by the finding that the fresh weight of cut flowers exhibiting petal wilting, such as carnation [20], *Eustoma grandiflorum* [22], daylily [23], and rose [24], gradually decreases with time. Wilting petals were isolated from the inflorescence and supplied with water to strengthen this assumption. If petals abscise, the fresh weight of petals is expected to be increased by the water supply. The fresh weight of petals in ‘Carnelian’, ‘LaLaLa’, and ‘Namahage Cute’ increased with water supply during the first day, suggesting that apparent petal wilting in these cultivars is caused by the blockage of water flow due to abscission.

In the five cultivars, ‘Garnet’, ‘Kamakura’, ‘Moon Waltz’, ‘Port Light Pair Beauty’, and ‘Rose Quartz’, the browning of petals, a senescent symptom, was observed (Figure 4). Thus, petal senescence appears to progress in these cultivars although incomplete petal abscission may occur. Azuma et al. [5] reported that petal abscission was not observed in ‘Kamakura’

and 'Port Light Pair Beauty'. However, Yang et al. [25] observed that the formation of the abscission layer progressed during senescence in 'Kamakura'. Therefore, we concluded that petal abscission might have occurred in these two cultivars.

The fresh weight of petals remaining in the inflorescence in 'Agitate' and 'Micchan' did not obviously decrease (Figure 2). Additionally, the fresh weight of isolated petals in these cultivars did not increase with the water supply (Figure 3). Moreover, the petals in these cultivars exhibited browning. Petal abscission was not observed in the two cultivars [5]. Therefore, petal senescence in these cultivars advances without petal abscission.

From these results, the petal senescence of cut dahlia flowers could be categorized into three types: petal senescence in two, six, and two cultivars were categorized into abscission, abscission with withering, and wilting types, respectively.

STS treatment extended the vase life of cultivars exhibiting petal abscission, such as 'Namahage Cute' and 'LaLaLa' and cultivars exhibiting petal wilting, such as 'Agitate' and 'Micchan' (Table 3). These results indicate that pulse treatment with STS effectively extends the vase life of cut dahlia flowers irrespective of senescent types. The vase life of 'Agitate' was within 5 days, but its relative fresh weight was higher than 110% on day 5. This discrepancy is apparently due to the evaluation of vase life, which was determined to be the time when the petals at the outer three whorls either wilted or abscised. In this cultivar, the petals at the outer three whorls wilted, but the petals showing expansive growth at the inner whorl did not wilt on day 5.

The STS treatment did not significantly extend the vase life of some cultivars, including 'Port Light Pair Beauty' (Tables 2 and 3), which is consistent with the results of a previous study [5]. However, the continuous treatment with 1-MCP extended the vase life of all cultivars tested, including 'Port Light Pair Beauty' [5], suggesting that endogenous ethylene is involved in petal senescence in the dahlia. The accumulation of silver has been shown to be lower in flowers than in stems and leaves in cut flowers treated with STS in cut carnation [26] and *Delphinium* [27]. Therefore, the lower effectiveness of STS may be due to the low accumulation of silver in the flowers.

The effect of STS treatment on the extension of vase life in 'Kamakura' and 'Micchan' was greater in the results shown in Table 3 than in those shown in Table 2. The uptake of STS solution was greater in the latter experiment than in the former experiment in most cultivars. Thus, the different effectiveness of STS may be attributed to the amount of STS uptake. The uptake of STS by the cut dahlia flowers should be increased as much as possible to extend their vase life in practical applications.

The relative fresh weight of cut flowers in some cultivars, including 'Micchan' and 'Kamakura', did not increase with the STS treatment (Figure 5). The increase in the fresh weight of cut dahlia flowers has been shown to be mainly due to petal growth [2]. However, the petal growth in *E. grandiflorum* [14], snapdragon [18], and sweet pea [19] was slightly affected by the STS treatment.

The combined treatment with STS and GLA significantly extended the vase life of three out of five cultivars more than GLA alone, but did not extend it more than STS alone in the five cultivars (Table 3). As observed in some cultivars, including 'Micchan' and 'Moon Waltz', STS delayed petal wilting in the outer petals but GLA slightly delayed it (Figure 8). Therefore, the effectiveness of STS in extending vase life was partially due to this evaluation method for vase life. It should be noted that GLA promoted petal growth and extended the petal vase life of inner inflorescence (Figure 8). Similarly, Azuma et al. [2] reported that continuous treatment with glucose plus CMIT/MIT extended the petal vase life of inner inflorescence in cut dahlia flowers [2].

For some cut flowers, such as carnation [28], rose [29], and anthurium [30], cytokinins, including 6-benzylaminopurine (BA), extend their vase life. Similarly, treatment with BA extends the vase life of cut dahlia flowers [4]. Combined treatment with BA and GLA extends the vase life of cut dahlia flowers [31,32]. BA treatment increases invertase activity, which may be associated with extending vase life [33]. This suggests that the action mechanism of BA on the extension of the vase life of cut dahlia flowers differs from that of

STS and GLA. Thus, the vase life of cut dahlia flowers is expected to be extended more by combined treatment with STS, BA, and GLA than with STS and GLA.

In conclusion, we categorized the petal senescence of cut dahlia petals into three types. Petal senescence in two, six, and two cultivars was due to abscission, abscission with withering, and wilting, respectively. Pulse treatment with STS significantly extended the vase life of both cultivars exhibiting petal abscission and those exhibiting petal wilting. Pulse treatment with STS, followed by continuous treatment with GLA extended the vase life more than STS alone or GLA alone in the five cultivars, ‘Garnet’, ‘Kamakura’, ‘LaLaLa’, ‘Micchan’, and ‘Moon Waltz’. This treatment is available to extend vase life in cut dahlia flowers.

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