

## 原著論文

A preservative composed of glucose, isothiazolinonic germicide, citric acid, and aluminum sulphate (GLCA) extends the vase life of cut 'Rote Rose' rose flowers under various conditions

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### Summary

In a previous study, a formulation composed of glucose, isothiazolinonic germicide, citric acid, and aluminum sulphate (GLCA) extended the vase life of cut rose flowers. In the present study, we investigated the effect of GLCA on the vase life of cut 'Rote Rose' rose flowers under various conditions. Application of GLCA markedly extended the vase life of flowers held at a high temperature (30°C). GLCA treatment also extended the vase life of flowers harvested earlier than the usual harvesting stage. To examine whether GLCA extended vase life under more typical conditions, we stored cut rose flowers at 10°C overnight, then maintained them at 8°C for 3 days. We then held them at 28°C and began treatment with GLCA. The vase life of these flowers was markedly extended by GLCA treatment. When cut rose flowers were pre-treated with GLCA for 4 days as a preservative for growers, vase life was not significantly extended, but this treatment increased flower diameter and inhibited the problem of bent neck.

**Key Words:** aluminum sulphate, cut flower, glucose, isothiazolinonic germicide, rose, vase life

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## Introduction

The longevity of cut flowers is an important quality for consumers (Imanishi et al., 1992). The vase life of cut rose flowers is generally short (Ichimura et al., 1999), and this short vase life has been attributed to a shortage of soluble carbohydrates (Ichimura et al., 2003) and to vascular occlusion, which restricts the water supply to the flowers (Mayak et al., 1974). Although large amounts of soluble carbohydrates are required for flower opening, roses are harvested at the bud stage, which leads to a shortage of soluble carbohydrates. Occlusions are thought to develop due to various factors, such as the growth of bacteria (van Doorn et al., 1989; Zagory and Reid, 1986), the development of air emboli (Durkin, 1979; van Doorn, 1990), and physiological responses of the stems to harvesting (Marousky, 1969).

To extend the vase life of cut roses, many formulations or chemicals have been developed. There are basically three types of preservatives: those designed for growers, for wet transport, and for consumers. Preservatives for growers are applied to the cut flowers for short-term before shipment. This type of preservative includes RNA-Ag-tris (Ohkawa et al., 1999) and the polymer 2-hydroxy-3-ionene-chloride (Ueyama and Ichimura, 1998). However, these preservatives are not widely used by Japanese growers, probably because of their low effectiveness and high cost. Many preservatives for consumers have also been developed. These preservatives include sugars and antimicrobial compounds that inhibit vascular occlusion. Sucrose plus an 8-hydroxyquinoline compound, such as 8-hydroxyquinoline sulphate (HQS) or 8-hydroxyquinoline citrate, is known to extend the vase life of cut rose flowers (Marousky, 1969; Kaltaler and Steponkus, 1976; Ichimura et al., 1999). However, Ichimura et al. (2002) previously reported that in a study of 10 rose cultivars, sucrose plus HQS did not extend the vase life of 5 cultivars, including 'Rote Rose' and 'Noblesse', which are currently the leading cultivars in Japan. In addition, this solution shortened the vase life of one cultivar. In contrast, continuous treatment with a formulation known as GLCA, which is composed of glucose, Legend MK (a mixture of antimicrobial compounds), citric acid, and aluminum sulphate, markedly extended the vase life of cut rose flowers when they were held at a constant temperature of 23°C (Ichimura et al., 2006). To clarify whether GLCA can be applied in practice, it is necessary to investigate its performance under a range of conditions.

In the present study, we investigated the effectiveness of GLCA in prolonging the vase life of cut 'Rote Rose' rose flowers under various conditions.

## Materials and Methods

### *Plant Materials*

Roses (*Rosa hybrida* L.) cv. Rote Rose, harvested at normal harvest maturity (stage 2, as described by Ichimura and Ueyama, 1998) when the petals started to unfold, were obtained from a commercial grower in Ise, Mie Prefecture, Japan, in 2001. These flowers had not undergone any preservation treatment before we obtained them for use in our experiments. To examine the effect of harvest stage, flowers whose petals were still tightly furled (stage 1, Ichimura and Ueyama, 1998) were also harvested. After harvesting, the cut ends of the flower stems were immersed in tap water and stored at 5°C overnight. Next day, the cut flowers were transported to the laboratory and used within 2 h for the experiments described in the remainder of this section. Before proceeding, all flower stems were trimmed to 45 cm, and all leaves except for the upper three were removed.

### ***GLCA treatment of cut rose flowers***

The GLCA solution was composed of 10g L<sup>-1</sup> glucose, 0.5 ml L<sup>-1</sup> CMI/MI (Legend MK; Rohm and Haas Japan K.K., Tokyo, Japan), which contained 11.3g L<sup>-1</sup> 5-chloro-2-methyl-4-isothiazolin-3-one and 3.9 g L<sup>-1</sup> 2-methyl-4-isothiazolin-3-one as active ingredients, and aluminum sulphate at 50 mg L<sup>-1</sup> supplemented with 30 mg L<sup>-1</sup> citric acid to acidify the solution (Ichimura et al. 2006). In all experiments, two cut flowers were placed in each of four 500-ml beakers containing 500 ml of distilled water (DW) or GLCA solution, for a total of eight flowers per treatment. Unless otherwise stated, cut flowers were held at 23°C under 70% relative humidity, and a 12-h photoperiod with 10 μmol m<sup>-2</sup> sec<sup>-1</sup> irradiance from cool-white fluorescent lamps.

### ***Evaluation of vase life***

The fresh weights and flower diameter of the cut flowers were measured daily. Unless otherwise stated, vase life was defined as the period from the start of GLCA treatment to the time when the petals wilted.

### ***Effect of temperature on vase life***

To examine the effect of temperature, the flowers were held at 23°C or 30°C. All other environmental conditions were the same as those described above.

### ***Treatment with GLCA under environmental conditions simulating typical storage and transport conditions***

The cut rose flowers were held in DW at 10°C in darkness for 1 day to simulate wet transport, which is commonly used for roses. The flowers were then cut to a length of 43 cm and held in DW at 8°C in darkness for 3 days to simulate preservation. The flowers were then transferred to a temperature-controlled chamber at 28°C under 70% relative humidity and 10 μmol m<sup>-2</sup> sec<sup>-1</sup> irradiance with a 12-h photoperiod and the application of GLCA was begun.

### ***Time of GLCA treatments***

Cut rose flowers were held at 8°C in darkness during the first 4 days, then transferred to the temperature-controlled chamber at 23°C under 70% relative humidity and 10 μmol m<sup>-2</sup> sec<sup>-1</sup> irradiance with a 12-h photoperiod. The flowers were treated with GLCA either during the first 4 days while they were held at 8°C (pre-treatment), from the 4th day onwards at 23°C (post-treatment), or continuously throughout the experimental period (continuous treatment). Vase life was evaluated from the time when the flowers were transferred to 23°C to the time when the petals wilted.

## **Results**

### ***Effects of GLCA and temperature on vase life***

The fresh weight of flowers held in DW at 23 and 30°C increased for only 3 and 1 day, respectively (Fig. 1), and the flowers did not open fully at 30°C in DW (Table 1). Treatment with GLCA increased fresh weight even when the flowers were held at 30°C. In the GLCA-treated flowers, there was little difference in flower diameter between 23 and 30°C (Table 1). The vase life was significantly shorter at 30°C than at 23°C. Treatment with GLCA markedly extended

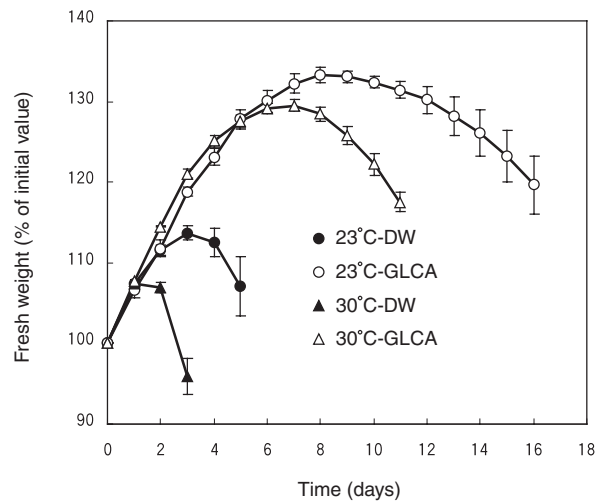


Fig. 1

Changes in the fresh weight of cut rose flowers treated with distilled water (DW) or GLCA at temperatures of 23 and 30°C. Values represent the means of 3 replications  $\pm$  SE.

Table 1 Effects of GLCA and temperature on the vase life and flower diameter of cut rose flowers

Temperature (°C)	Solution	Vase life (days)	Flower diameter <sup>z</sup> (mm)
23	DW	5.1 $\pm$ 0.4 <sup>y</sup>	104 $\pm$ 6
	GLCA	15.1 $\pm$ 0.2	125 $\pm$ 2
30	DW	3.0 $\pm$ 0.0	78 $\pm$ 1
	GLCA	10.9 $\pm$ 0.1	130 $\pm$ 1
Significance ( <i>P</i> value)			
Temperature		<0.0001	0.0099
Solution		<0.0001	<0.0001
Temperature $\times$ Solution		0.0012	0.0005

<sup>z</sup> Maximum diameter.

<sup>y</sup> Values represent means of 3 replications  $\pm$  SE.

the vase life of cut roses at both temperatures.

#### *Effects of GLCA and harvest stage on vase life*

Without GLCA treatment, cut flowers harvested at stage 1 did not open fully (Table 2). Treatment with GLCA markedly increased the fresh weights of cut flowers regardless of the harvesting stage (Fig. 2). GLCA treatment also increased the diameter of flowers harvested at stage 1 to the same size as those harvested at stage 2 and extended the vase life of cut roses harvested at the both stages to a similar degree (Table 2).

#### *Effect of GLCA on vase life under conditions simulating typical storage and transport conditions*

In DW, the fresh weight of cut flowers increased slightly for about the first 3 days, then decreased sharply thereafter

Table 2 Effects of GLCA and harvest stage on the vase life and flower diameter of cut rose flowers

Stage	Solution	Vase life (days)	Flower diameter <sup>z</sup> (mm)
2	DW	5.3±0.3 <sup>y</sup>	108± 6
	GLCA	15.0±0.2	123± 1
1	DW	4.5±0.3	75±12
	GLCA	15.4±0.4	128± 1
Significance ( <i>P</i> value)			
Stage		0.5490	0.0596
Solution		<0.0001	0.0003
Stage × Solution		0.0891	0.0123

<sup>z</sup> Maximum diameter.

<sup>y</sup> Values represent means of 3 replications ± SE.

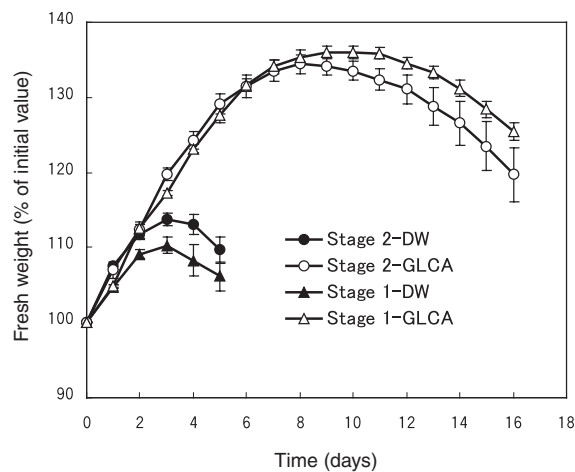


Fig. 2

Changes in the fresh weight of cut rose flowers harvested at stages 1 (tightly furled petals) and 2 (petals beginning to open) and treated with distilled water (DW) or GLCA. Values represent the means of 3 replications ± SE.

(Fig. 3). In contrast, treatment with GLCA increased the fresh weights of the flowers and maintained fresh weight at a high level for about 1 week. Flowers in DW did not open fully, but those in GLCA opened almost completely. GLCA treatment markedly extended the vase life of the flowers under these experimental conditions (Table 3, Fig. 4).

#### ***Effect of time of GLCA treatment on vase life***

Fig.5 shows the changes in fresh weights of the cut flowers. In the control, fresh weight increased during the first 6 days, then decreased thereafter. Pre-treatment with GLCA for 4 days increased the fresh weight more than in the control, but the time before flowers began to decline was similar to that in the control flowers. Post-treatment and continuous treatment with GLCA both markedly increased fresh weights and delayed the time before flowers began to

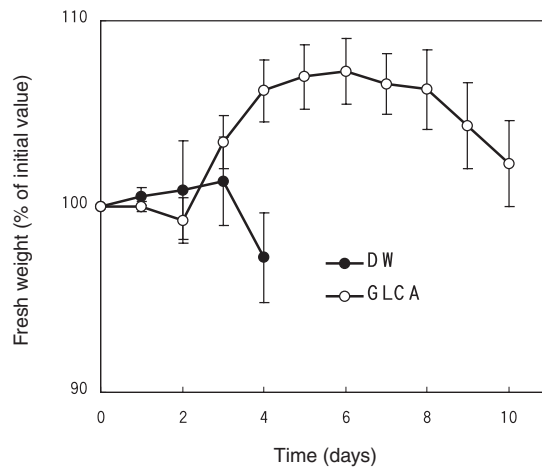


Fig. 3  
Changes in the fresh weight of cut rose flowers. Cut roses were kept at 10°C for 1 day, then at 8°C for 3 days. They were then placed in distilled water (DW) or GLCA and kept at 28°C. Values represent the means of 4 replications  $\pm$  SE.

Table 3 Effect of GLCA on the vase life of cut rose flowers under conditions that simulated typical storage and transport conditions

Solution	Vase life (days)	Flower diameter <sup>z</sup> (mm)
DW	3.8 $\pm$ 0.1 <sup>y</sup>	93 $\pm$ 4
GLCA	10.0 $\pm$ 0.4	125 $\pm$ 2
Significance ( <i>P</i> value)	<0.0001	0.0230

<sup>z</sup> Maximum diameter.

<sup>y</sup> Values represent means of 4 replications  $\pm$  SE.



Fig. 4  
Effect of GLCA on the vase life of cut rose flowers held under conditions simulating typical storage and transport. Left, distilled water (DW); right, GLCA. The photograph was taken 9 days after the start of GLCA treatment.

decline. The vase life was not significantly extended by pre-treatment with GLCA (Table 4). However, pre-treatment with GLCA for 4 days suppressed the problem of bent neck and promoted flower opening (Fig. 6). Flower diameter was significantly increased by the pre-treatment with GLCA compared with that in the control (Table 4). Post-treatment and continuous treatment both significantly extended vase life and increased flower diameter compared with the control (Table 4).

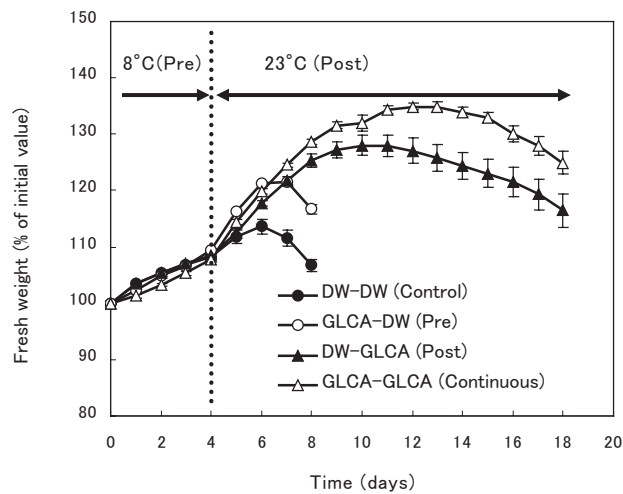


Fig. 5 Changes in the fresh weight of cut rose flowers treated with GLCA at different times. Control, no GLCA treatment; Pre, pre-treatment with GLCA followed by treatment with distilled water (DW); Post, post-treatment with GLCA; Continuous, treatment with GLCA throughout the study period. Values represent the means of 4 replications  $\pm$  SE.

Table 4 Effect of time of GLCA treatment on the vase life of cut rose flowers

Treatment time	Vase life (days)	Flower diameter <sup>z</sup> (mm)
Control (DW)	3.5 $\pm$ 0.2a <sup>y</sup>	105 $\pm$ 4a
Pre-treatment	4.1 $\pm$ 0.1a	117 $\pm$ 3b
Post-treatment	13.4 $\pm$ 0.6b	126 $\pm$ 1bc
Continuous treatment	14.3 $\pm$ 0.3b	131 $\pm$ 2c

<sup>z</sup> Maximum diameter.

<sup>y</sup> Values represent means of 4 replications  $\pm$  SE. Values in a column followed by the same letters do not differ significantly ( $P < 0.05$ , Tukey-Kramer multiple range test).

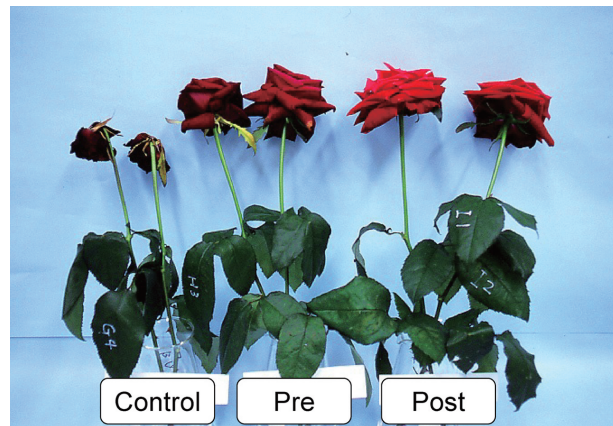


Fig. 6  
Effect of time of GLCA treatment on the vase life of cut rose flowers. Left, distilled water (DW, control); middle, pre-treatment with GLCA; right, post-treatment with GLCA. The photograph was taken 13 days after transfer of the cut roses to 23°C.

## Discussion

Ichimura et al. (2006) reported that GLCA extended the vase life of cut roses held at a constant temperature of 23°C. In the present study, GLCA extended the vase life of cut roses under various conditions. This effect is likely to be due to an increased supply of soluble carbohydrate combined with inhibition of vascular occlusion (Ichimura et al., 2003). In a previous study, a combined treatment with glucose and HQS did not extend the vase life of cut roses in some cultivars (Ichimura et al., 2002). However, GLCA significantly extended the vase life of roses in all the cultivars tested in a previous study (Ichimura et al., 2006). Thus, CMI/MI, antimicrobial compounds contained in GLCA, seems to have low toxicity to cut roses. Knee (2000) also reported that CMI/MI was suitable for extending the vase life of some cut flowers.

The higher the temperature, the shorter was the vase life of cut rose flowers (Ichimura and Ueyama, 1998; Ichimura et al., 1999). Furthermore, the flowers of cut roses maintained at high temperature were smaller than those maintained at low temperature (Ichimura et al., 1999). We confirmed that flowers maintained at 30°C were significantly smaller than those maintained at 23°C without GLCA treatment (Table 1). However, there was little difference in flower size between 23 and 30°C in the GLCA treatments. Furthermore, the vase life of cut roses was significantly extended by GLCA treatment at 30°C (Table 1). Thus, GLCA treatment might improve the vase life of cut roses kept at a high temperature, such as during the summer.

Maintaining the freshness of cut flowers is the most outstanding merit of wet transport. However, when cut flowers are wet-transported at relatively high temperature, flower opening is accelerated compared with dry transport (Hu et al., 1998). Harvesting flowers at an earlier stage might solve this problem. Thus, we investigated the effect of GLCA on the vase life of cut roses harvested earlier than the conventional stage. Treatment with GLCA extended the vase life of roses harvested at stage 1 to levels similar to those for roses harvested at stage 2. Furthermore, there was little difference in flower diameter between the two stages (Table 2). Thus, GLCA can be used to preserve cut rose flowers harvested earlier than usual.

Guaranteed sale for cut flowers has increased the demand for cut flowers in England (Ogawa, 2004). In Japan, guaranteed sale has been carried out as in European countries. In the present study, the vase life of cut rose flowers



held at 28°C was extended to about 10 days by the application of GLCA under conditions that simulated typical storage and transport conditions (Table 3). As these conditions seem to have a severe effect on the vase life of cut roses, using preservatives such as GLCA might produce cut roses with a guaranteed vase life of at least 1 week.

In Japan, wet transport has become popular recently. Hu et al. (1998) reported that the vase life of cut 'Bridal Pink' rose flowers was longer in wet transport than in dry transport. In addition, the vase life of cut roses was extended by treatment with sugars during transportation (Hu et al., 1998). Thus, we investigated the effect of GLCA pre-treatment for 4 days on vase life, which simulated the application during wet transport. Pre-treatment with GLCA did not significantly extend vase life, but the treatment promoted flower opening and suppressed bent neck (Table 4, Fig. 6). In cut sweet pea flowers, the vase life was extended more by increasing the duration of sucrose treatment (Ichimura and Suto, 1999). Thus, the reduced effectiveness of a pulse treatment with GLCA can be attributed to the decreased amount of glucose absorbed by the cut rose flowers. As GLCA has not been developed as a preservative for use by growers, other formulations will be needed.

In conclusion, continuous treatment with GLCA markedly extended the vase life of cut rose flowers even when they were held at 30°C or harvested at an earlier stage than usual. In addition, GLCA treatment extended the vase life of cut roses under simulated typical storage and transport conditions. Furthermore, treatment with GLCA extended the vase life of all cultivars in a previous study (Ichimura et al., 2002). These findings suggest that GLCA will be widely useful for preserving cut rose flowers under various conditions.

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## グルコース，イソチアゾリン系抗菌剤，クエン酸および硫酸アルミニウムから構成される処方（GLCA）は多様な条件下でバラ‘ローテローゼ’切り花の品質保持期間を延長する

市村 一雄・田口 誠

### 和文摘要

前報でグルコース，イソチアゾリン系抗菌剤，クエン酸および硫酸アルミニウムから構成される処方（GLCA）はバラ切り花の品質保持期間延長に効果があることを見出した。そこで，GLCAが多様な条件下でバラ‘ローテローゼ’切り花の品質保持期間延長にどの程度効果があるか調べた。バラ切り花を30℃の高温条件下で保持した場合も，GLCA処理により品質保持期間は著しく延長した。バラ切り花を通常よりも早期に収穫した場合も，GLCA処理により品質保持期間は延長した。GLCAが実際の流通段階を想定した条件下で品質保持期間延長に効果があるか明らかにするため，10℃で1日間保存した切り花をさらに8℃で3日間保管した後，28℃に移しGLCA処理を開始した。その結果，GLCA処理により品質保持期間は延長した。生産者段階でのGLCA処理がどの程度品質保持効果があるか明らかにするため，GLCAの4日間前処理が品質保持に及ぼす影響を調べた。GLCA前処理は品質保持期間を有意には延長しなかったが，バントネックの発生を抑制し，開花径を増大した。