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ABA Improves Postharvest Quality of Cut *Lilium* 'Sorbonne' Harvested in Late Period

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With the delay of harvest stage, the carbohydrate content of flower petals and the diameter of flower buds increased, but the vase life was shortened. Effects of abscisic acid (ABA) and sucrose on preservation of cut lilies were related to the harvest stage. Due to the lower level of soluble sugars and other energy source in early harvested cut lilies, the sucrose pretreatment turned to be particularly effective for extending the vase life and increasing the flower fresh weight and bud diameter. For the cut flowers harvested at middle stage, pretreatment with sucrose or ABA separately enhanced the preservation effect, which was not observed when pretreated with the combination of sucrose and ABA. The preservation effect of the harvested flowers at the late development stage was increased by the ABA pretreatments including ABA alone or ABA and sucrose (ABA+Suc). ABA improved sucrose uptake from the pretreatment solution and increased soluble sugar contents in cut flower petals. Combined with sucrose pretreatment, addition of ABA alleviated leaf chlorosis symptom of cut flowers harvested in early period.

Key words: abscisic acid, cut lily, flower preservation, postharvest stage, sucrose

INTRODUCTION

Lily 'Sorbonne' (*Lilium* Oriental 'Sorbonne') has straight flower stems, big flowers with bright colors and long flowering period, therefore it is one of the most popular cut lily varieties (Gill *et al.*, 2006). As one of the basic preservative ingredients, sucrose is widely used in the fresh–keeping of cut lilies to extend the vase life, increase flower size, and improve the flower colors (Han, 2003; Van Doorn and Han, 2011). Carbohydrate content in cut flowers at harvest time was significantly related to flower opening status and the vase life (Van der Meulen–Muisers *et al.*, 2001).

As a senescence phytohormone, abscisic acid (ABA) is related to flower petal senescence of rose (Mayak and Halevy, 1972) and carnation (Nowak and Veen, 1982). ABA improves the synthesis of ethylene, so the flowers, which are sensitive to ethylene, display the senescence effect (Ronen and Mayak, 1981; Nowak and Veen, 1982; Onoue *et al.*, 2000). Exogenous application of ABA makes the aging–related synthesis occur in daffodil petals in the earlier time (Hunter, 2004). The preservation effect of ABA, however, has been reported by regulating water balance of cut flowers through controlling stomata opening (Halevy *et al.*, 1974; Pompodakis and Joyce, 2003). Borohov *et al.* (1976) reported that for the rose petals containing no stomata, ABA regulates water equilibrium through the ways other than stomata opening regulation. Appropriate ABA treatment improves the quality of the vase flowers by inhibiting the sucrose–induced leaf chlorosis (Pompodakis and Joyce, 2003; Shimizu–Yumoto and Ichimura, 2009). Some researchers indicated that ABA might be the main factor affecting senescence of cut flowers (Yang *et al.*, 1996), while others suggested that ABA has dual effects on senescence of cut lilies: low concentrations of ABA show certain anti–aging effect and high concentrations lead to aging enhancement (Li *et al.*, 2012). When a lower concentration of ABA is applied to cut flowers with sucrose, the leaf chlorosis process may delay and the flower fresh weight may increase notably. However, sometimes ABA plus sucrose (ABA+Suc) treatment inhibits the fresh weight of cut flowers, which might be related to the harvest time of cut flowers and the nutritional status of the flowers at harvest time.

In the present study, the preservation effects of sucrose and ABA pretreatments on cut flowers of *Lilium* Oriental 'Sorbonne' at different developmental stages were investigated. Soluble sugar contents in the flower petals prior to and after the pretreatment were determined to understand the preservation mechanism of sucrose and ABA for lily cut flowers.

MATERIALS AND METHODS

Plant materials

Bud–cut flowers of Oriental lily 'Sorbonne' were harvested in a cut flower production center located in Donghai, Jiangsu Province, China on November 25, 2010. Branches with single top flower buds were selected. The harvested bud–cut flowers were immediately immersed in water and carried to the laboratory for preservation pretreatment. Based on the diameters and colors of the buds, they were divided into three different develop-

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mental stages: (Ⅰ) tight and dark green buds with the diameter of 3.19 cm, (Ⅱ) loose and yellow–green buds with the diameter of 3.40 cm, and (Ⅲ) starting to open and turning red buds with the diameter of 3.62 cm. Flowering branches were retained 30 cm in length and the top four leaves were maintained.

Preservation treatment

The bud–cut flowers of all groups with different developmental stages were subjected to pulsing pretreatments at three conditions with varied combination of 8–hydroxyquinoline (8–HQ, 200 mg·L^{-1}), sucrose (Suc, 30 g·L^{-1}) and ABA (0.5 mg·L^{-1}) as shown in Table 1. Distilled (DI) water was used for the control (CK). The pulsing treatment was carried out for 18h at normal temperature (indoor natural light 9 h + darkness 9h), then the bud–cut flowers were placed in vases containing distilled water and kept in an incubator (Model: RP–435D) at temperatures of 21°C/14°C (day/night), humidity of 60%, and light intensity of 6000 LX/0 LX (day/night). The pulsing and vase solutions were all 500 ml in volume and 12 cm in depth. The solutions in the vases were replaced with distilled water every other day. To ensure the same environmental conditions for different cut flowers for reducing experimental deviations, the vase locations in the incubator were placed randomly and exchanged every day.

Table 1. Compositions of different pretreatment solutions

Pretreatment	Composition of pretreatment solutions
CK	DI water
Suc	sucrose + 8–HQ
ABA	ABA + 8–HQ
ABA+Suc	ABA + sucrose + 8–HQ

Notes: Concentrations of 8–hydroxyquinoline (8–HQ), sucrose and ABA were 200 mg·L^{-1}, 30 g·L^{-1} and 0.5 mg·L^{-1}, respectively.

Measurement of morphological indicator

Leaf color, bud diameter, and fresh weight of 12 bud–cut flowers in each treatment were recorded every day until the flower colors became brown and the petal edge reversely curled, which were regarded as the end of the vase life. Score of leaves was recorded by dividing leaf colors into four degrees: 1, dark green; 2, light green; 3, yellow–green; and 4, yellow–brown. Full bloom was considered when the flower diameter reached maximum. The change rate of fresh weight was calculated as the following:

$$\text{Change rate of fresh weight} = (W_n - W_1)/W_1 \times 100\% \quad \text{.......... (1)}$$

where W_n is the weight of branch on the n day of vase days, and W_1 is the fresh weight on the first vase day.

Determination of petal soluble sugar contents

For measuring the soluble sugar contents in the petals, three flowers were selected randomly prior to and after the pretreatments, at full–bloom and before senescence, respectively. One gram of petals was placed in 20 mL distilled water. After boiling for 30 min followed by cooling to room temperature and filtration with filter paper, the sample filtrates were collected. Glucose was used to prepare a standard solution to make a calibration curve. Dilute anthrone sulfuric acid reagent (5 mL) was mixed with each standard or sample solution (0.1 mL), immediately placed in a boiling water bath for 1min, and then cooled to room temperature. The absorbance at 630nm was measured.

Determination of flower sucrose uptake after the pretreatments.

To determine the flower sucrose uptake, the following formula was used:

$$\text{Sucrose uptake (g)} = 15 - V \times C \quad \text{..........................(2)}$$

where 15 – sugar in the pulsing solution prior to the treatment, g; V – the remaining volume of solution after pretreatment, L; C – the sugar content determined by the anthrone–sulfuric acid method, g/L.

RESULTS

Effects of sucrose and ABA on the vase life and postharvest quality of cut lilies

Compared to the control at Stage I, pretreatment with three solutions all resulted in a slight increase in vase life and flower diameter, but sucrose alone and ABA+Suc treatments exhibited more significant effect than ABA alone (Table 2). However, the pretreatment with ABA+ Suc delayed the opening of the cut flowers than either sucrose alone or ABA alone.

At Stage Ⅱ, the vase life of the cut flowers pretreated with sucrose and ABA was 12.17 d and 12.08 d, respectively, which was about 1 d later than the control CK (11.08 d). Correspondingly, the time to reach the maximum flower diameter was also delayed. The pretreatment of sucrose or ABA extended the vase life mainly by delaying the opening time. The diameters of the cut flowers in sucrose or ABA pretreatment were not significantly different from those of the control group. At this stage, the pretreatment with ABA+Suc did not result in significant difference in vase life or bud diameter from the control.

At Stage Ⅲ, the sucrose treatment did not affect the vase life, but resulted in slight decrease in the bud diameter. The pretreatment with ABA alone or with ABA+ Suc not only extended vase life, but also increased the flower diameter.

The effects of sucrose, ABA or ABA+Suc on vase life and cut flower diameter varied from the development stage of the harvest time. Along with the delay of postharvest stage from I to Ⅲ, the cut flowers opened quickly and the flower diameter increased, but the vase life was shortened.

Effect of sucrose and ABA pretreatment on leaf chlorosis of cut lilies

Figure 1 shows the scoring values of leaves in the first

Table 2. Effect of Sucrose and ABA pretreatments on vase life and postharvest quality

Postharvest stage	Treatments	Vase life of flowers (days)	Diameter of opened flowers (cm)	Days to anthesis
I	CK	11.75±1.22[z]	18.91 d[y]	7.58±1.73
	Suc	12.42±0.67	19.83 bcd	7.67±1.37
	ABA	12.08±0.90	19.17 cd	7.33±1.61
	ABA+Suc	12.42±0.79	19.67 bcd	8.92±1.72
Ⅱ	CK	11.08±1.00	20.26 ab	6.75±1.14
	Suc	12.17±0.72	20.46 ab	7.17±1.34
	ABA	12.08±0.79	20.23 ab	7.33±0.78
	ABA+Suc	11.70±1.25	20.29 ab	6.90±1.20
Ⅲ	CK	10.60±1.43	20.23 ab	5.80±1.14
	Suc	10.58±1.93	19.67 bcd	5.25±1.22
	ABA	11.27±1.42	20.03 bc	5.73±1.27
	ABA+Suc	11.58±0.90	21.12 a	5.92±1.83

[z] Values ± standard deviation.
[y] Different letters in the same column mean significant differences at the level of P<5%.

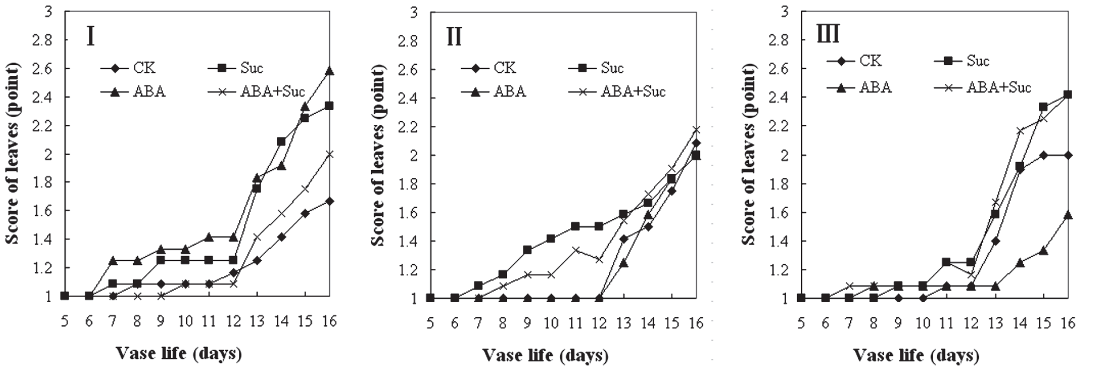

Fig. 1. Effects of sucrose and ABA pretreatments on ornamental quality of leaves of 'Sorbonne' cut branches. I, Ⅱ, and Ⅲ in the figures indicate postharvest stages (I: earliest development status; Ⅲ: latest developmental status; and Ⅱ: the intermediate status).

5 days to 16 days, the higher score showed more serious leaf chlorosis symptom. Compared to the control, the cut flowers at Stage I pretreated with ABA, sucrose or ABA+Suc all displayed more serious leaf chlorosis symptom although ABA+Suc pretreatment resulted in less leaf chlorosis symptom than ABA or sucrose pretreatment. For the cut flowers at Stage Ⅱ, when sucrose or ABA+Suc pretreatment was applied, the leaf ornamental quality decreased in the first 12 days being placed in the vase solution indicated by the increased score of leaves. The chlorosis of the leaves without sucrose pretreatment was not investigated in the first 12 days. After 15 days in the vase solution, there was no significant difference in leaf chlorosis among the treatments. For the cut flowers at Stage Ⅲ, leaf chlorosis symptom developed fastest due to the exogenous addition of sucrose and extra addition of ABA did not reduce the degree of leaf chlorosis. When ABA was pretreated alone, however, the leaves during postharvest evaluation kept bright green, and the ornamental value was higher compared to the control.

Effect of sucrose and ABA pretreatment on the fresh weight of cut lilies

As shown in Fig. 2, the change rates of fresh weight of the cut flowers at the three harvest stages all increased in the first 5–8 days followed by a decrease. The weight of cut flowers at Stage I increased most tremendously and the decrease started later than the cut flowers at other two stages. On the 12th day, the fresh weight of the cut flowers at Stages I and Ⅱ was still increasing, but that of the flowers at Stage Ⅲ began to decline.

For the cut flowers at Stage I, Suc, ABA and ABA+Suc pretreatments led to a significant increase in their fresh weight, especially Suc and ABA+Suc pretreatments compared to the control CK. For those at Stage Ⅱ, all pretreatments resulted in an increase in cut flower fresh weight. The Suc pretreatment had the most significant effect followed by ABA and ABA+Suc. At Stage Ⅲ, All of the four pretreatments exhibited very similar impact and they did not affect the fresh weight as significant as found for those at Stages I and Ⅱ.

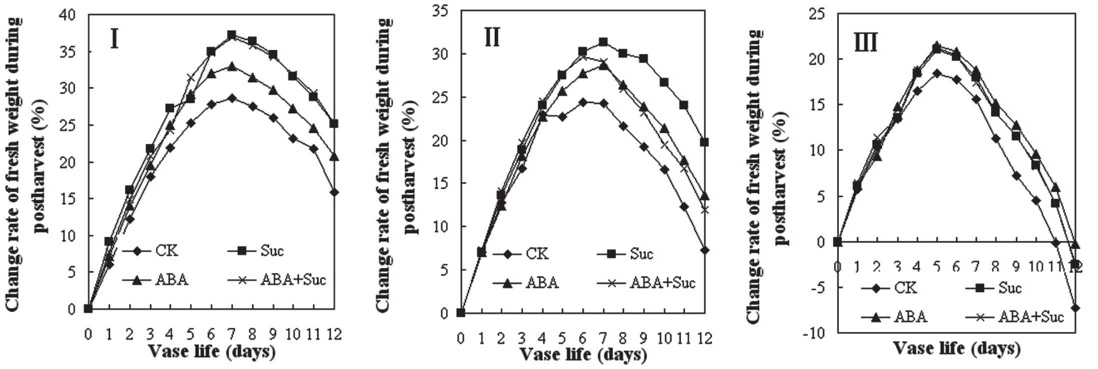

Fig. 2. Effects of sucrose and ABA pretreatments on fresh weight of 'Sorbonne' cut flowers. I, Ⅱ and Ⅲ in the figures indicate postharvest stages (I: earliest development status; Ⅲ: latest developmental status; and II: the intermediate status).

Table 3. Change of sucrose content in the pulsing solution prior to and post pretreatments

Serial Number	Sucrose Uptake (g)	Serial Number	Sucrose Uptake (g)
Suc I	4.15 d	ABA+ Suc I	4.08 d
Suc Ⅱ	6.31 bc	ABA+ Suc Ⅱ	5.74 c
Suc Ⅲ	6.60 ab	ABA+ Suc Ⅲ	7.16 a

Effect of sucrose and ABA pretreatments on the soluble sugar content in petals

The amount of sucrose uptaken by the cut flowers was obtained (Table 3) by calculating the changes of sucrose content in the pretreatment solution. The order of sucrose uptake of cut flowers from the treatment solution was Stage Ⅲ > Stage Ⅱ > Stage I, indicating that the cut flowers at increased developmental stage was more active in the metabolism and thus requiring more sucrose. Application of ABA with sucrose led to slight inhibition of sucrose absorption at State Ⅱ and promotion at State Ⅲ, but no significant difference in sucrose uptake induced by ABA application was found. With increasing developmental stage of the cut flowers, the soluble sugar content in the flower petals increased and more nutrients were accumulated. Except for the cut flowers at Stage I pretreated with water (CK) and sucrose, the soluble sugar contents in the cut flowers pretreated with other solutions tended to decrease. However, the sucrose contents in the cut flower petals pretreated with sucrose–containing solutions, including Suc and ABA+Suc, were higher than those pretreated with water (CK) and ABA treatment. The soluble sugar contents of the cut flowers pretreated with Suc and ABA+Suc at Stages Ⅱ and Ⅲ increased until the 5th vase day and right after they dropped tremendously as well as in the control and ABA–pretreated cut flowers.

DISCUSSION

Along with the developmental stages of cut flowers changing from I through Ⅲ, cut flowers opened quickly and the flower diameter increased, but the vase life was shortened (Table 2). Sugar and ABA showed different effects on the preservation of cut flowers at the different developmental stages because of varied sugar accumulation at the three stages.

For the cut flowers at earlier developmental stage (I), less carbohydrate accumulated in the petals compared to that in the later stages (Fig. 3). Shortage of energy substances in the cut flowers is the main factors leading to senescence especially for those harvested at the early developmental stages (Van der Meulen–Muisers *et al.*, 2001). As one of the basic component in a preservative

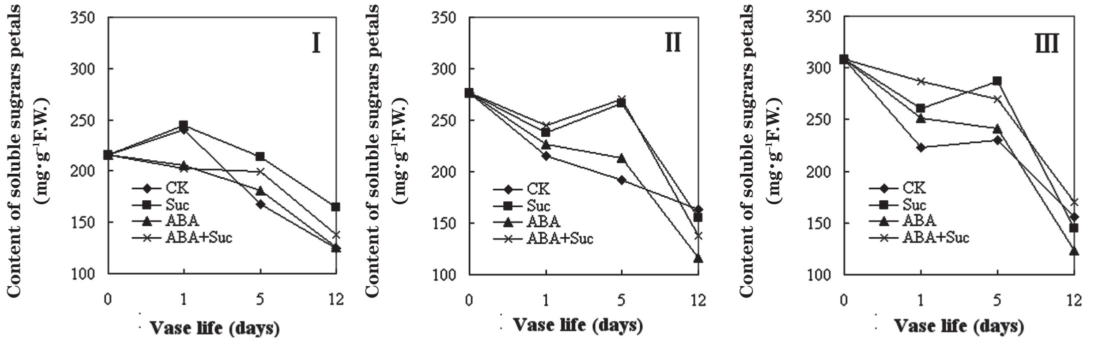

Fig. 3. Effects of sucrose and ABA pretreatments on soluble sugar contents in 'Sorbonne' cut flower petals. I, Ⅱ, and Ⅲ in the figures indicate postharvest stages (I: earliest development status; Ⅲ: latest developmental status; and Ⅱ: the intermediate status).

solution, sucrose not only supplies the cut flowers with energy, metabolism substrate, but also promotes the water balance of cut flowers (Halevy and Mayak, 1979). Even though the cut flowers at early stage absorbed less sucrose from pretreatment solutions (Table 3), the soluble sugar content in the cut flower petals significantly increased after the pretreatment (Fig. 3). Sucrose pretreatment including sucrose alone and ABA+Suc resulted in extended vase life (one more day) and increased fresh weight and bud diameter of cut flowers. The preservation effect of sucrose on ethylene–sensitive cut flowers was less significant than that on the ethylene–insensitive cut flowers (Van Doorn, 2004). Han (2003) found that sucrose treatment does not significantly extend the vase life of Oriental lily 'Stargazer' as well as the bud diameter, but increase the content of anthocyanins in the petals.

The cut flowers harvested at late developmental time have accumulated a greater amount of carbohydrate (Fig. 3). The preservation effect of sucrose pretreatment, therefore, was not as significant as for the early–harvested cut flowers and the impact of ABA turned to be more important. ABA treatments including ABA and ABA+Suc significantly promoted the vase quality of the cut flowers harvested at stage Ⅲ (Table 2). It was assumed that the ABA pretreatment could slow down the metabolism of cut flowers to some extent, and thereby delayed the senescence. To the cut flowers of stage Ⅱ, the sugar or ABA pretreatment alone extended the vase life of cut flowers, but the two combined together inhibited the fresh weight and sucrose absorption of cut flowers, resulting less significant preservation effect (Figs. 2 and 3). These findings indicate that the effect of sucrose and ABA treatment on the preservation of cut flower is directly related to the developmental stages of cut flowers at harvest time.

To the cut flowers at different harvest time (varied developmental stage), ABA treatment extended the vase life at varying degrees, balanced the water of cut flowers, and promoted the quality of cut flowers (Table 2 and Fig. 2). Li *et al*. (2012) found that low concentrations of ABA in 8–HQ solution inhibits the promotion effect of 8–HQ on fresh weight of cut flowers without significant effect on the vase life and other ornamental quality of cut flowers. In this study, compared with the control (treated with water), the ABA treatment resulted in the promotion effect on cut flower fresh weight, which might be due to the added 8–HQ. The effect of ABA plus sucrose on vase quality was affected by the developmental stages of the cut flowers. The treatment had a remarkable effect on the cut flowers with the advanced developmental stage time, alleviating the decline of the soluble sugar content in the cut flower. The ABA treatment might slow down the senescence of the cut flowers by adjusting metabolism of the internal substance in the petals, instead of simple stomatal regulation. Borohov *et al*. (1976) indicated that the preservation effect of ABA was not necessarily related to stomatal regulation. Ichimura and Shimizu–Yumoto (2007) also found that ABA treatment could promote sucrose absorption of petals, while its inhibitory effect on transpiration was not observed.

Application of appropriate concentration of ABA increased chlorophyll content in the leaves of cut flowers (Ye and Li, 2008), and postponed the sucrose–induced leaf wilting (Markhart and Harper, 1995; Shimizu–Yumoto and Ichimura, 2009). Similar findings were also reported for lily cut flowers (Li *et al*., 2012). Our study indicates that the effect of ABA treatment (including ABA alone and ABA+Suc) on leaf chlorosis varied from the cut flower developmental stage at harvest time. It was suggested that leaf chlorosis of cut flowers was related to the excessive carbohydrate accumulation in the leaves (Markhart and Harper, 1995; Shimizu–Yumoto, and Ichimura, 2007). In this study, the regulatory role of ABA on leaf chlorosis degree of cut flower was also affected by the developmental stage of the cut flowers at harvest time, so ABA might affect leaf chlorosis through regulating the carbohydrate accumulation in the leaves.

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