Effect of Exogenous Adenosine Triphosphate Supply on the Senescence-related Physiology of Cut Carnation Flowers

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Carnation (*Dianthus caryophyllus* L.) flowers deteriorate rapidly after harvest. Symptoms of deterioration include petal in-rolling and discoloration, which results in reduced vase life (Badiyan et al., 2004; Bowyer et al., 2003; Thompson et al., 1982). The short vase life of cut carnation flowers may limit successful marketing.

Energy metabolism related to physiological disorders and senescence of plant tissues has been investigated in various harvested horticultural crops, such as litchi fruit, pear, and carnation (Duan et al., 2004; Saquet et al., 2003; Solomos and Gross, 1997; Veltman et al., 2003). In carnation and rose, membrane deterioration and senescence are associated with a decrease in respiratory activity and may be delayed when flowers are supplied with respiratory substrates (Fobel et al., 1987; Monteiro et al., 2001; Thompson et al., 1982; Trippi and Paulin, 1984). Trippi and Paulin (1984) showed that an increase in membrane permeability was associated with a decrease of energy production in senescing cut carnation flowers. Vase solutions containing adenosine triphosphate (ATP) can extend the vase life of cut carnation flowers (Song et al., 2006b). Thus, it is suggested that the low-energy status of plant tissues may lead to cut flower senescence and reduced vase life.

This investigation determined the effects of exogenous ATP supply on the senescence-related physiology of cut carnation flowers. With a view to understanding better the role of tissue energy status in the senescence of carnation flowers, the effects of exogenous ATP supply on time to maximum flower expansion, vase life, rates of ethylene production and respiration, and endogenous

ATP and adenosine monophosphate (AMP) concentrations during vase life of cut carnation flowers were investigated.

Carnation cv. 'Master' flowers were obtained from a commercial market in Guangzhou, China. Flowers were transported to the laboratory within 6 h. Flowers were uniform at commercial maturity (outer petals horizontal), as described by Wu et al. (1992). Their stems were recut under water to a length of ≈ 30 cm. Stems were assigned at random to flasks containing either distilled water (control) or 0.1 mmol·L⁻¹ ATP solution until the end of the vase life of the control flowers (up to 10 d). The concentration of 0.1 mmol·L⁻¹ ATP is the most beneficial in extending the vase life of the cut flowers, as identified by Song et al. (2006b). The volume of ATP solution or distilled water was maintained at constant levels by replenishing the flasks daily. There were four flowers per flask and three flasks (replications) per treatment. Flasks were arranged in a completely randomized design. Throughout the experimental period, flowers were held at 25 °C and 70% to 80% relative humidity with a 12-hour light period per day at an irradiance of 12 W/m² at flower level from fluorescent light tubes.

The time (d) from harvest to the maximum flower expansion was calculated. Vase life was evaluated every 12 h on a scale from 1 to 5 pt as described by Song et al. (2006b), where 1 pt is fresh without any deterioration; 2 pt is slight discoloration, mold growth, or wilting; 3 pt is moderate discoloration, mold growth, or wilting; 4 pt is severe discoloration, mold growth, or wilting; and 5 pt is entirely discolored, substantial mold growth, or wilting. All flowers were scored until they had deteriorated to a score of 5 pt.

Stems were weighed every 2 d to enable calculation of relative fresh weight (percent of the initial fresh weight). Respiration and ethylene production rates were measured according to the method of Serrano et al.

(2001). Flower stems were trimmed to about 5 cm in length and sealed in a 500-mL glass jar for 1.5 h. A 1-mL gas sample was then withdrawn from the jar using a syringe. Carbon dioxide and C₂H₄ concentrations were determined by gas chromatography (Shimadzu GC 17A, Tokyo). Rates of ethylene production and respiration were calculated on a fresh weight basis. For determinations of ATP and AMP concentrations, outermost petal tissues (2 g) were homogenized with 15 mL 0.6 mol·L⁻¹ perchloric acid for 1 min in an ice bath as described by Ozogul et al. (2000). The extraction mixture was centrifuged at 3000 g for 10 min at 4 °C. Ten milliliters of supernatant was taken and adjusted to a pH of 6.5 to 6.8 with 1 mol·L⁻¹ KOH. The supernatant was allowed to stand for 30 min in an ice bath to precipitate most of the potassium perchlorate. Precipitate was then removed by centrifugation at 5000 g for 5 min at 4 °C. Adenosine triphosphate and AMP concentrations were determined using a high-performance liquid chromatograph (Waters 2690) equipped with a C₁₈ reverse-phase column [octadecylsilyl (ODS), 4.6 × 250 mm] according to the method of Duan et al. (2004). Adenosine triphosphate and AMP concentrations were expressed on a fresh weight basis.

Experiments were arranged in a completely randomized design. There were three replicates. Data were tested by analysis of variance using SPSS version 7.5. Least significance differences (LSDs) were calculated to compare significant effects at the 5% level.

Application of 0.1 mmol·L⁻¹ ATP to the vase solution extended the vase life of cut carnation flowers from 12.7 to 16.3 d, an increase of 3.6 d, and delayed the time to maximum flower expansion by 1.7 d, compared with non-ATP-treated flowers held in distilled water (Table 1). Fresh weight of cut carnation flowers increased within the first 2 d of vase life, but declined at day 6 (Fig. 1A). The ATP-treated cut carnation flowers had a relatively high fresh weight compared with the control flowers throughout the vase life evaluation period (Fig. 1A). Similar results were observed in response to provision of respiratory substrates to potted and cut rose flowers (Monteiro et al., 2002; Podd and Van Staden, 2002; Van Doorn and Reid, 1991).

Cut carnation flowers can show a marked climactericlike respiration pattern during senescence (Mayak and Dilley, 1976; Podd and Van Staden, 2002). However, Trippi and Paulin (1984) found generally decreasing

Table 1. Effects of exogenous adenosine triphosphate (ATP) supply on the time from harvest to maximum flower expansion and vase life of cut carnation flower.

ATP	Time to maximum flower expansion(d)	Vase life(d)
Control	7.1 ± 0.2 a	12.7 ± 0.9 a
0.1 mmol·L ⁻¹	8.8 ± 0.4 b	16.3 ± 1.0 c

Data are the mean ± SE of 12 flowers. Means within a column followed by a different letter are significantly different at the 5% level. ATP, adenosine triphosphate.

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Fig. 1. (A–C) Effects of exogenous adenosine triphosphate (ATP) supply on fresh weight (A), respiration (B), and ethylene production rate (C) of the outermost petals of cut carnation flowers in a distilled water vase solution containing either 0 (●) or 0.1 (▲) mmol·L⁻¹ ATP. Data are the mean ± SE of 12 flowers. Where no error bar appears, the SE was smaller than the size of the symbol.

Fig. 2. (A, B) Effects of an exogenous adenosine triphosphate (ATP) supply on ATP (A) and adenosine monophosphate (AMP) (B) contents of the outermost petals of cut carnation flowers in a distilled water vase solution containing either 0 (●) or 0.1 (▲) mmol·L⁻¹ ATP. Data are the mean ± SE of 12 flowers. Where no error bar appears, the SE was smaller than the size of the symbol.

respiratory activity during senescence of cut carnation flowers. Van Doorn and Reid (1991) reported that respiratory patterns of cut carnation flowers during vase life were cultivar dependent. In this study, exogenous supply of 0.1 mmol·L⁻¹ ATP generally enhanced respiratory activity (Fig. 1B). The observed trends in respiration rate during vase life evaluation were only weakly climacteric.

Ethylene production by the cut carnation flowers slowly increased after 4 d and reached a maximum after 8 d of vase life (Fig. 1C). The peak in ethylene production rate was coincident with the appearance of visible senescence symptoms, including petal in-rolling and withering. Provision of 0.1 mmol·L⁻¹ ATP in the vase solution consistently reduced ethylene production rates throughout the vase life evaluation period. Van Doorn and Reid (1991) observed that vase life extension in cut flowers was related to inhibition in ethylene production. Borochoy and Adam (1984) found that ATP supply inhibited ethylene production of detached carnation petals.

Adenosine triphosphate is the primary energy pool in living tissues. Endogenous ATP content in cut carnation flowers increased from day 0 to day 4, and then decreased (Fig. 2A). In contrast, AMP content increased after day 4 (Fig. 2B). Exogenous ATP supply maintained relatively a higher ATP content and reduced the increase in AMP content at the later stages of vase life. Exogenous sucrose to vase life solution increased carbohydrate levels, enhanced respiration, and extended the longevity of potted miniature rose, possibly as a result of maintenance of tissue energy (Monteiro et al., 2002). A correlation between energy metabolism and incidence of core browning of pear fruit or senescence of cut carnation flower has been established (Saquet et al., 2001; Trippi et al., 1988; Veltman et al., 2003). Reduction of the ATP level synthesized in the cell could lead to a loss of cellular integrity, and incomplete terminal oxidation (Pradet and Raymond, 1983). In longan fruit, high ATP content and adenylate energy charge level of pericarp tissues could contribute to maintenance of membrane integrity (Su et al., 2005). Furthermore, exogenous application of ATP increased tissue energy status and exhibited potential for browning control and quality maintenance of harvested litchi fruit (Song et al., 2006a). It is suggested that an exogenous ATP supply could contribute to the maintenance of tissue energy and thereby delay senescence of cut carnation flowers.

In conclusion, an exogenous ATP supply extended the vase life and retarded the senescence of cut carnation flowers mediated through increasing fresh weight, enhancing respiration, reducing ethylene production rates, and maintaining higher endogenous ATP concentration in tissues, and showed the potential for practical horticultural value.

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