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Full Length Research Paper

Pre-harvest calcium sulfate application improves postharvest quality of cut rose flowers

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The quality of cut rose flowers at the postharvest stage is affected by pre-harvest conditions. The present study was conducted to examine the possible involvement of calcium sulfate ($CaSO_4$) in regulation of rose flower senescence. Roses (*Rosa hybrida* L.) cultivars 'Cool Water' and 'Pretty Blinda', were treated with either water or $CaSO_4$ at 200 mg L^{-1} containing 0.01% Tween 20, three times (1, 3 and 5 days) pre-harvest. $CaSO_4$ treatment promoted bud opening and delayed senescence. The treated flowers stayed turgid and continued their initial postharvest growth for longer periods of time. $CaSO_4$ spray increased calcium (Ca) content of stems, leaves and petals of flowers and suppressed ethylene production and ion leakage with age.

Key words: $CaSO_4$, ethylene, ion leakage, longevity, *Rosa hybrida*

INTRODUCTION

The length of vase life is one of the most important factors for quality of cut flowers. A marked variation in vase life among cut rose cultivars has been reported (Mortensen and Gislerod, 1999; Ichimura et al., 2002). The vase life of cut rose flowers is often short. The cut flowers wilt and the floral axis becomes bent just below the flower head, which is called "bent neck". Development of such symptoms is considered to be caused by vascular occlusion, which inhibits water supply to the flowers (Mayak et al., 1974; De Strigter, 1980). 'Cool Water' and 'Pretty Blinda' cultivars are susceptible to bent neck and their flower quality decreased with time by increasing in bent neck (Unpublished data). Nikbakht et al. (2008) reported that calcium (Ca) accumulation in scapes of gerbera flowers could prevent and delay in bent neck incidence.

Ca content in the tissue affects many processes during plant growth, at all stage of development (Ferguson and Drobak, 1988). The use of Ca in vase solutions increases water flow through the stems by association with pectin in the xylem cell walls (Van Ieperen and Van Gelder, 2006). The role of intra and extracellular Ca in altering cell metabolism is often attributed to its influence on cell wall and membrane structure and function (Ferguson, 1984; Konno et al., 1984). Ca is also involved in the regulation of an array of intracellular events as a second messenger. Its signaling role is important for normal cellular function and for the regulation of ongoing metabolic processes in the cytosol and organelles (Bush, 1995)

The recommended Ca concentration in the leaves for adequate growth of roses is at least 10 mg g^{-1} of dry matter (De kreij et al., 1992). Ca addition in the postharvest stage has been reported to promote longevity and bud opening in cut roses (Michalczuk et al., 1989). Gerasopoulus and Chebli (1999) reported that pre-harvest spray of calcium chloride ($CaCl_2$) delayed scape bending in gerbera by 2-3 days over the control. The most common and visibly apparent senescence symptom in rose petals is loss of cell turgor, resulting in wilting and death. Petal senescence is also associated with characteristic biochemical and biophysical events in the tissues, mostly because of alterations in membrane properties that are thought to play a primary role in the senescence process (Marangoni et al., 1996). In addition, many substances have been tested in pre-harvest sprays of many crops (Gerasopoulus and Chebli, 1999). Among them, the Ca ion has been promising to control *Botrytis cinerea* when used in various forms such as sulfate, chlorate, or nitrate, since this ion affects the physiology of both host and the fungus (Abeles et al., 1992).

Ethylene, a gaseous phytohormone, is involved in the senescence of many flowers. However, in roses, ethylene appears to be of minor importance in the natural senescence of most cultivars; ethylene production is low,

Table 1. Longevity and diameter (after 6 days) of cut flowers treated pre-harvest with water (control) and $CaSO_4$.

Cultivar	Treatment	Longevity (day)	Flower diameter (mm)
'Cool Water'	Calcium	10.6 ± 0.3a[z]	82 ± 2a
	Control	7.6 ± 0.6b	72 ± 3b
'Pretty Blinda'	Calcium	10.3 ± 0.3a	87 ± 3a
	Control	8.6 ± 0.3b	79 ± 2b

[z] Data are mean ± standard errors (n = 3). Values with different letters in the same list are statistically significant by Duncan's multiple range test at 5% level.

and the application of ethylene inhibitors or antagonist only slightly improves postharvest life. Nevertheless, with age most rose flower petals exhibit a small but clear-cut rise in ethylene production (Mayak et al., 1972). Ethylene enhances membrane senescence symptoms and has been suggested to initiate this chain of events. However, in a recent study (Nabigol et al., 2009), ethylene was shown to be a mediator of flower senescence rather than its starter. Because exogenous ethylene in onset of postharvest period could not accelerate senescence of cut rose petals. Rose flowers show a climacteric increase in ethylene production (Müller et al., 1998). Nevertheless, Mayak et al. (1972) reported that the climacteric ethylene production increase in a short-lived cultivar. Since *B. cinerea* infects preferably, senescent tissues (Elad and Volpin, 1988), researchers have emphasized the utilization of calcium to increase the resistance of tissues and delay senescence by inhibition of ethylene synthesis or action. In the present study, we investigated the possible relationship between tissue Ca content and senescence processes in rose flowers.

MATERIALS AND METHODS

Roses (*Rosa hybrida* L.) cultivars 'Cool Water' and 'Pretty Blinda', were grown in the Tehran university greenhouse in Karaj, Iran. Flowers were sprayed with either water plus 0.01% Tween 20 (control) or calcium sulfate ($CaSO_4$) at 200 mg L^{-1} containing 0.01% Tween 20. Solutions were sprayed to run-off (approximately 500 ml per plant), three times (1, 3 and 5 days) pre-harvest. Flowers were harvested at normal harvest maturity. After harvest, the cut ends of flower stems were immersed in tap water within 1 h. The cut flowers were then transported to the laboratory and used for experiments.

Vase life determination

Flower stems were trimmed to 40 cm, and all five-leaflet leaves except for the upper three were removed. Three cut flowers were placed in each 500 mL beaker filled with 300 mL, 20 g L^{-1} sucrose plus 200 mg L^{-1} 8-hydroxyquinoline citrate (HQC) as preservative solution. The cut flowers were kept at 23°C under 70% relative humidity. A 12 h photoperiod was maintained with 10 µmol m^{-2} s^{-1} irradiance from cool-white fluorescence lamps. Vase life was the period between harvest and the time when either the petals lost turgor or at least one petal had abscised. The diameter of each cut flower and fresh weight of whole flowering stalk including stem, leaves and flower head, were measured daily.

Leakage of electrolytes

One disc (1 cm^2) was punched from each petal and five discs were immersed for 10 min in 50 ml water. The solution was discarded to remove the ions from the cell-free space and after a further addition of 15 ml H_2O, the discs were shaken (120 rpm) for 90 min on an orbital shaker. The electrical conductivity of the solution was measured and then the petal discs were autoclaved for 30 min at 121°C. The leakage rate was expressed as the percentage of total conductance following tissue destruction.

Ethylene measurement

Each flower was sealed in a 3 L glass vessel and kept at 20°C. After 2 h, a 1 ml gas sample was withdrawn into a syringe and the ethylene concentration determined by a Shimadzu gas-chromatograph equipped with an activated alumina column fitted with a flame ionization detector. Ethylene concentrations were calculated and expressed in nL g^{-1} h^{-1}.

Statistical analyses

Data were taken from three independent experiments, and mean and standard error (SE) values were determined. Data were analysed by Duncan's multiple range test using SAS software.

RESULTS AND DISCUSSION

Ca treatments significantly increased the postharvest life of both 'Cool Water' and 'Pretty Blinda' flowers (Table 1). In general, $CaSO_4$ application was more effective in delaying senescence in 'Cool Water' flowers than in 'Pretty Blinda'; 28% for the former and only 15% for the latter when the treatments were given to whole flowers. Ca application promoted flower opening, and thus increased flower diameter in both cultivars (Table 1).

$CaSO_4$ enhanced the initial fresh weight increment and delayed the later reduction in relative fresh weight of cut flowers (Figure 1). The initial period of growth extended rapidly in 'Pretty Blinda' flowers than in 'Cool Water'. After 8 days, the relative fresh weight of the untreated flowers was below that measured at harvest and wilting symptoms were visible. In contrast, the treated flowers were still turgid and attractive.

$CaSO_4$ slowed the rate of electrolyte leakage from the

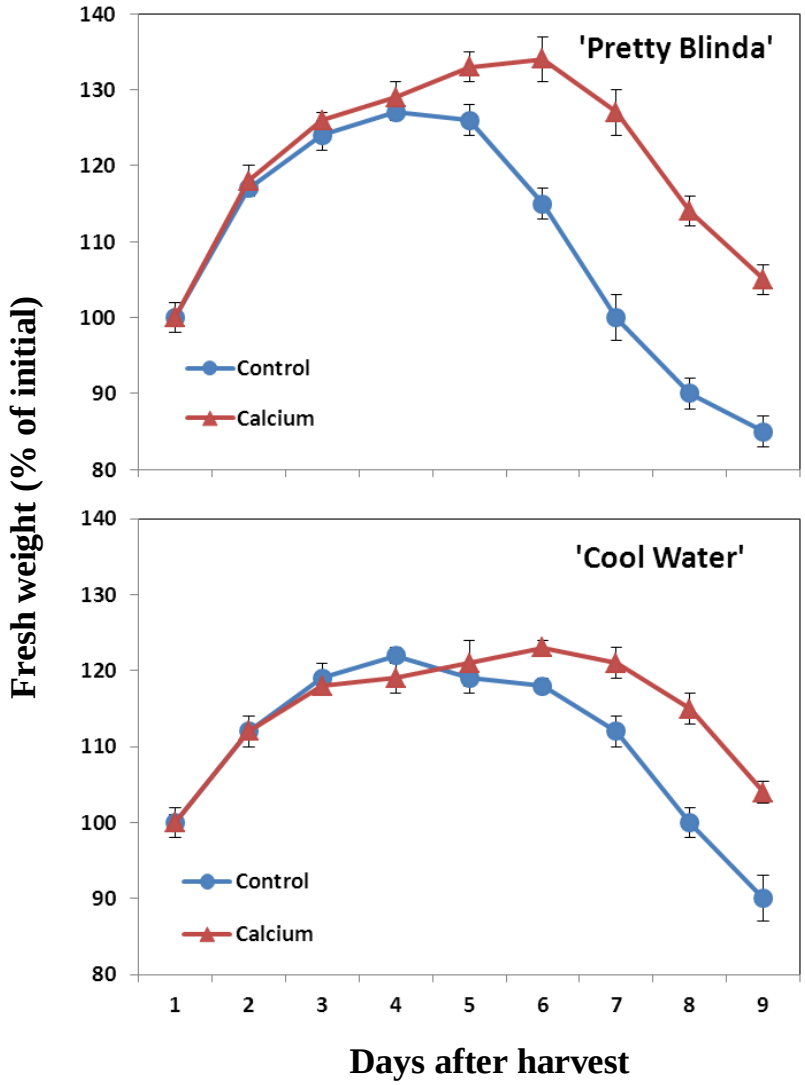

Figure 1. Effects of pre-harvest $CaSO_4$ treatment on fresh weight of cut 'Pretty Blinda' and 'Cool Water' flowers. Vertical bars show standard errors of means (n, 3).

petals (Figure 2). After 3 days for both treated and untreated flowers a similar increase in leakage rate began, with no significant difference between them. However, after 6 and 9 days the leakage rate was 8 and 29% higher, respectively, in untreated versus Ca treated flowers.

In both cultivars, ethylene production increased during the first days after harvest, peaking on after 6 days, and then decreasing to 9 days. However, there was little difference in ethylene production patterns between 'Cool Water' and 'Pretty Blinda'. Ca treated flowers produced less ethylene than untreated ones (Figure 3).

Spraying of cut flowers with $CaSO_4$ delayed senescence, extended flower longevity, and promoted bud opening in both 'Cool Water' and 'Pretty Blinda' flowers (Table 1). The beneficial effects of Ca on postharvest longevity and bud opening have been demonstrated previously in several rose cultivars (Michalczuk et al., 1989). The similar responses of different species and different rose cultivars having distinct genetic backgrounds may reflect calcium's general effect on postharvest life and senescence. Culture treatments, such as incorporation of calcium in the nutrient solution (Starkey and Pedersen, 1997) influence the calcium level in flower tissue. Increased Ca concentrations in the tissue have been shown to result in better postharvest quality, mainly due to reduced gray mold infection and reduced discoloration of petal edges (Starkey and Pedersen, 1997; De capdeville et al., 2005). The main difference between treated and untreated rose flowers in our experiments was that calcium treated flowers stayed turgid for a longer period (Figure 1). Among the elements determining the quality of rose flowers, bud opening is a major factor. Bud opening requires full turgidity, which

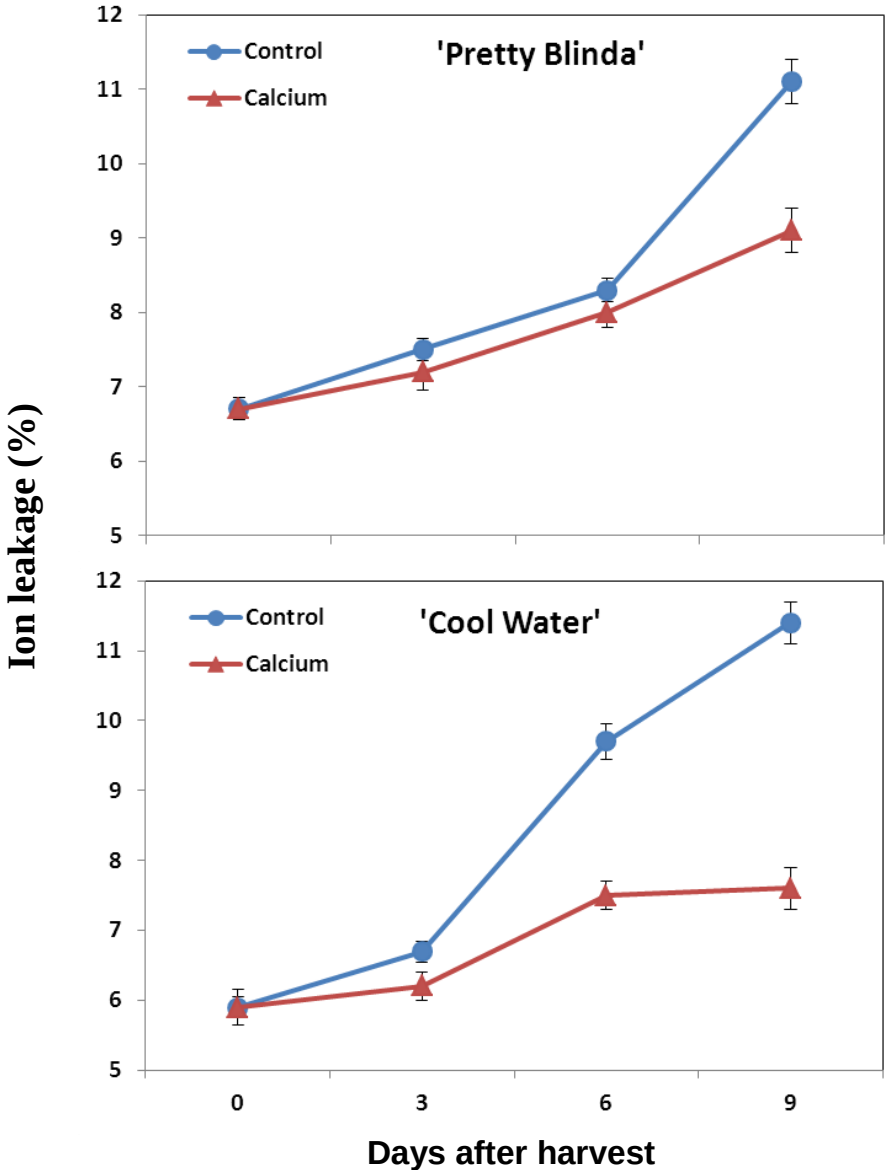

Figure 2. Effects of pre-harvest $CaSO_4$ treatment on ion leakage of 'Pretty Blinda' and 'Cool Water' petals. Vertical bars show standard errors of means (n, 3).

depends on both continued water uptake and diminished leakage from cells. Ca treatment delayed the increase in membrane permeability in flowers (Figure 2), this could explain, at least in part, the observed improvement in the flower's water status, and indicate that a possible mode of Ca action is via maintenance of membrane integrity and function.

Petals treated with Ca produced less ethylene than their untreated counterparts (Figure 3). A similar Ca effect was reported for cut roses by Torre et al. (1999). This Ca effect may be due to its stabilizing effect on cell membranes since, in general, inhibition of phospholipids degradation results in the decreased production of membrane's lipid metabolites and, ultimately lower ethylene production (Borochov et al., 1997).

Conclusion

Ca in the cellular solution is essential to maintaining normal membrane function and to ensuring the selective permeability of the membrane. Therefore, increased Ca concentrations in the petals should lead to lower membrane permeability and thus to better membrane functionality, which may delay disruption of Ca compartmentation and maintain the normal cellular functions for a longer period. Therefore, pre-harvest application of $CaSO_4$ could be effective in extending vase life of cut

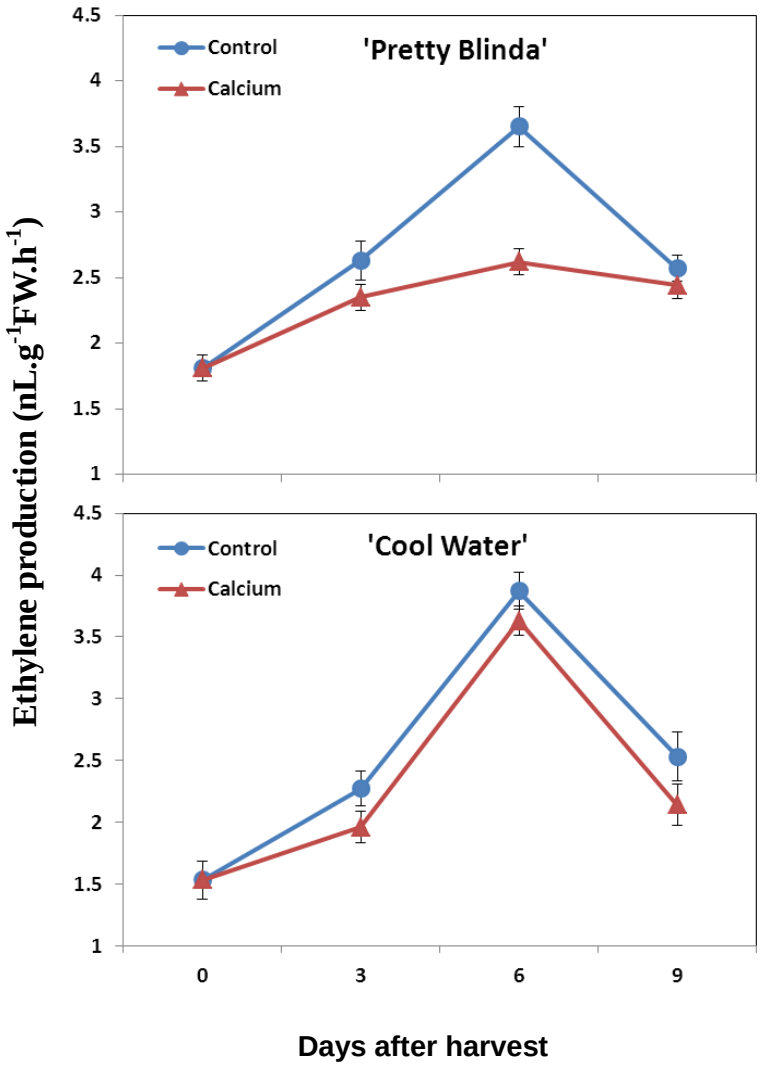

Figure 3. Effects of pre-harvest $CaSO_4$ treatment on ethylene production of cut 'Pretty Blinda' and 'Cool Water' flowers. Vertical bars show standard errors of means (n, 3).

rose flowers.

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