Effect of Sucrose on Ethylene Biosynthesis in Cut Spray Carnation Flowers

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Abstract

Various concentrations of sucrose (0, 2.5, 5.0, 7.5, 10.0 or 15.0%) were fed to cut spray carnation flowers cv. Barbara. Vase life, ethylene production, ACC production and ACO activity were measured. Sucrose at 5.0% extended flower vase life the most. Its effect on vase life was associated with a delay in the petal climacteric ethylene peak. The same delay was found in gynoecium ethylene production. It is concluded that sucrose apparently inhibits climacteric ethylene production by inhibiting ACO activity. This effect may be indirect, by affecting gene expression.

INTRODUCTION

The longevity of a number of cut flowers has been associated with their sugar content (Aarts, 1957). The beneficial effect of sugars was attributed to supply of substrate for respiration, structural materials and osmoticum (Halevy and Mayak, 1979). Biocides have to be included in the vase solution if sugars are given, as the sugars themselves will promote bacterial growth, which results in xylem occlusion (net loss of water because rate of water uptake becomes lower than rate of transpiration). The increase in vase life, after sugar treatment, has also been associated with suppression of ethylene biosynthesis, in cut flowers such as snapdragon (Ichimura and Hisamatsu, 1999), rose (Liao et al., 2000), and/or reduced sensitivity to ethylene, as in carnation: Mayak and Dilley, 1976) and delphinium (Ichimura et al., 2000). It is not known how sugars suppress ethylene biosynthesis (O'Donoghue et al., 2002).

This research attempts to test the hypothesis that sugars may extend the vase life of spray carnation by suppressing ethylene biosynthesis, more specifically suppressing ACC production or ACO activity or both.

MATERIALS AND METHODS

Plant Material

Spray carnation cv. Barbara (*Dianthus caryophyllus* L.) flowers were harvested at the stage where the outer petals were just horizontal. Harvest occurred in the morning on the day of the experiment. Flowers were harvested from the institute`s glasshouse and transported dry immediately to the evaluation room. Stems were recut to 4 cm and subjected to treatment within an hour after harvest.

The vase life of flower was determined as the time from dipping the stem in test solution to wilting of the petals or appearance of petal necrosis. The day the flowers were put in the test solution was considered day 0 and 24 h later day 1. Flowers were held at 23°C, 70% relative humidity with a photoperiod of 12h at 10 µmol.m^{-2}.sec^{-1} light intensity from cool-white fluorescent lamps.

Sucrose (Wako, Japan) was applied at various concentrations (0, 2.5, 5.0, 7.5,10.0, 15%), together with 200 mg L-1 hydroxyquinoline sulphate (HQS). From time to time distilled water or sucrose solution was added to replenish uptake by the flowers. For subsequent experiments, sucrose at a concentration of 5.0% was used.

Ethylene Production

Individual flowers were enclosed in glass bottle (150.0 ml volume) for 1 hour at

23°C. One ml gas sample was withdrawn with a hypodermic syringe and injected into a gas chromatograph (Shimadzu GC-7AM) equipped with an alumina column and flame ionization detector. Separate petals (conical flask; vol. 64.5 ml) and gynoecia (conical flask; vol. 24.5ml) were also enclosed for 1 hour at 23°C and ethylene was measured as described.

ACC Assay and ACC Oxidase Activity

ACC was extracted and assayed as described by Lizada and Yang (1979), with modifications. Petals (0.5 g) were weighed, dissected into small pieces and put into test tubes. 5 ml of ethanol 80% (v/v) was added and the solution was placed in a boiling water bath for 20 min. After 20 minutes, the test tubes were removed from the bath and allowed to cool. The sample was homogenized, centrifuged and evaporated in a centrifuge evaporator. The dry residue remaining after evaporation was dissolved in 3 ml of distilled water, and an aliquot of 0.5 ml was assayed for ACC according to Lizada and Yang, 1979.

ACC oxidase was extracted and assayed as described by Vriezen et al. (1999), with modifications. Petals (0.5 g) were weighed, grounded with liquid nitrogen in a chilled mortar and pestle, which was kept in ice. Extraction was done with 1.5 ml of buffer (w/v) containing 100 mM Tris-HCl (pH 7.5) plus 10% glycerol, 30 mM isoascorbic acid and 5 mM DTT. The solution was centrifuged. For ACO assay, 0.2 ml of sample was taken in a test tube. 1.7 ml of buffer, 0.1 ml each of Fe SO_4 and 0.1 M Na HCO_3 was added. To a duplicate sample 0.05 ml ACC (20 mM) was added, and the vial was immediately closed with a silicon cap. The tubes were incubated at 30°C for 30 min with gentle shaking. At the end of the incubation period, 1 ml of headspace air was injected into the GC for ethylene analysis.

RESULTS AND DISCUSSION

Sucrose at 5.0% was found to be the most effective concentration ($P< 0.001$) in extending the vase life of the spray carnation tested (Fig. 1). The increase in vase life was associated with a delay of the climacteric ethylene peak in petals (Fig. 2). Increased ethylene production in the gynoecium showed a similar delay. There was no difference in ethylen production between the petals and the gynoecium, both in controls and in the sucrose treatment (data not shown).

Sucrose treatment increased ACC production (Fig. 3) but delayed and suppressed ACO activity (Fig. 4). The results indicate that sugars do not act by inhibiting ACS activity. In contrast the sucrose effect on ACO activity seems to fully explain the delay in the ethylene climacteric peak after sucrose treatment. This effect may be indirect, by affecting gene expression. The regulation of the induction of the ethylene climacteric may already be repressed, resulting in ACO gene expression.

ACKNOWLEDGEMENTS

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Figures

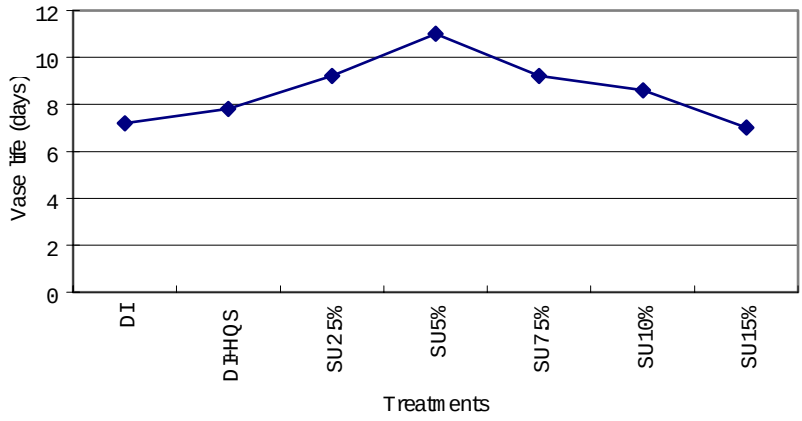

Fig. 1. Effect of sucrose on vase life of spray carnation flowers cv. Barbara.

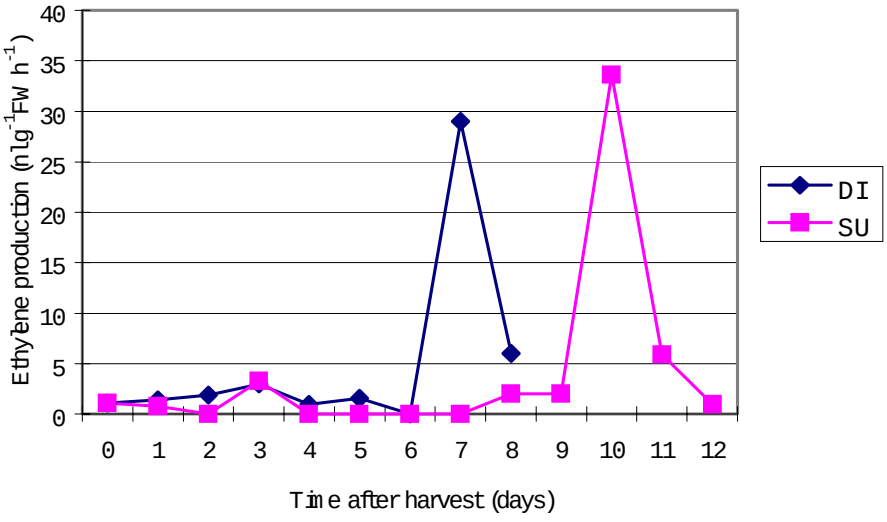

Fig. 2. Effect of sucrose on ethylene production of spray carnation flowers cv. Barbara.

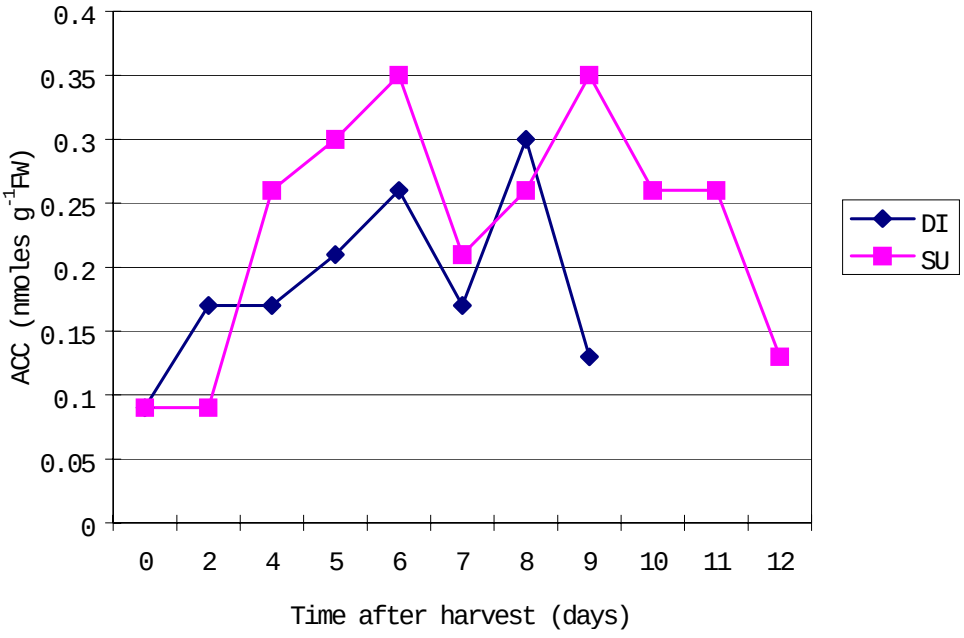

Fig. 3. Effect of sucrose on ACC production of spray carnation flowers cv. Barbara.

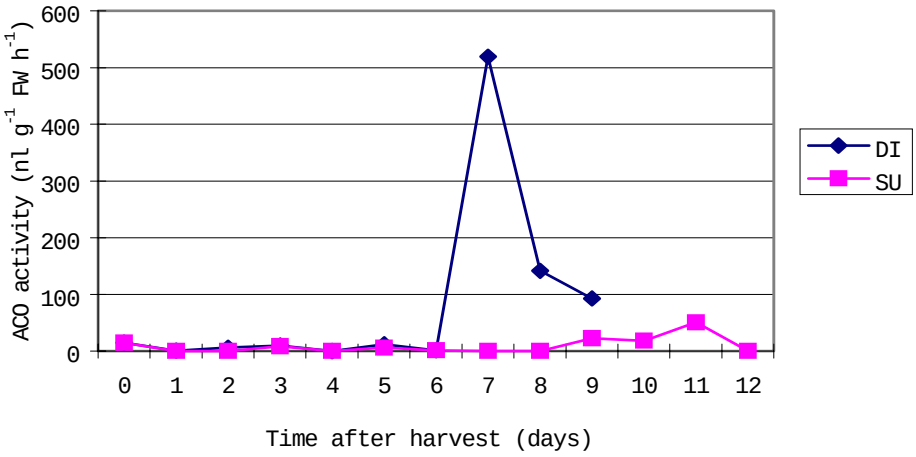

Fig. 4. Effect of sucrose on ACO activity of spray carnation flowers cv. Barbara.