Postharvest Biology and Technology 86 (2013) 479–486

Contents lists available at ScienceDirect

Postharvest Biology and Technology

journal homepage: www.elsevier.com/locate/postharvbio

Postharvest characteristics of cut dahlia flowers with a focus on ethylene and effectiveness of 6-benzylaminopurine treatments in extending vase life

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ARTICLE INFO

Article history:
Received 9 April 2013
Accepted 22 July 2013

Keywords:
6-Benzylaminopurine
Dahlia
Dip
Ethylene
Spray

ABSTRACT

With the aim of extending vase life of cut dahlia flowers, we investigated the postharvest characteristics of the flowers. Our focus was on the role of ethylene on senescence and on treatments that have extended vase life of other flowers. Continuous exposure to ethylene at 2 or 10 $\mu L\,L^{-1}$ significantly accelerated petal abscission in cut flowers. Flowers continuously immersed in 1 or 10 $\mu L\,L^{-1}$ 2-chloroethylphosphonic acid (CEPA) solution wilted earlier than those treated with distilled water (DW) or 0.15 $g\,L^{-1}$ citric acid. Ethylene production from the ovary and ray petal was relatively high (4.5 and 0.9 $nL\,g^{-1}$ fresh weight h^{-1}, respectively) at harvest, but decreased gradually over 5 days. No remarkable increase in ethylene production was observed during senescence. Silver thiosulfate complex (STS), an inhibitor of ethylene action, did not extend the vase life of cut flowers, although a high silver concentration was detected in flower organs. In contrast, pulse treatment with 1-methylcyclopropene (1-MCP) and dip treatment with 6-benzylaminopurine (BA) extended the vase life of florets, and BA was more effective than 1-MCP when the flowers were held in both DW and CEPA. BA spray treatment extended vase life of cut 'Kokucho,' 'Kamakura' and 'Michan' flowers. These results suggest that dahlia flower senescence is partially regulated by ethylene, and BA is more effective in delaying the senescence of cut dahlia flowers than ethylene action inhibitors.

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1. Introduction

Dahlia is a bulbous plant originating from Central and South America. Dahlia flowers are varied in color, shape and size. There are a great many horticultural cultivars in the world. In Japan, cut dahlia flowers have been popular in recent years because a reddish-black cultivar, 'Kokucho,' stimulated a dahlia boom among retailers. However, the short vase life (5–7 days) of cut dahlia flowers has curtailed the expansion of demand.

Ethylene controls the progression of senescence in many flowers, such as carnation (Nichols, 1966; Wu et al., 1991), petunia (Whitehead et al., 1984a), *Eustoma* (Ichimura and Goto, 2000) and sweet pea (Mor et al., 1984). These flowers are sensitive to ethylene, and ethylene production by these flowers increases during flower senescence (Veen, 1979; Mor et al., 1984; Whitehead et al., 1984a; Porat et al., 1993; Ichimura and Goto, 2000; Shimizu-Yumoto and Ichimura, 2009). Woltering and van Doorn (1988) and van Doorn (2001) classified sensitivity to ethylene into five categories based on the degree of acceleration of flower senescence by exposure to 3 $\mu L\,L^{-1}$ ethylene for 24 h.

In some flowers, the length of exposure to ethylene appears to be important for their ethylene responsiveness. For example, in *Campanula medium*, the vase life of flowers is unaffected by exposure to ethylene for 16 h, whereas all flowers wilt at the end of a 48 h ethylene treatment (Kato et al., 2002). Onozaki et al. (2004, 2008) evaluated the sensitivity to ethylene of carnation strains by continuous exposure to ethylene and found marked variations in sensitivity to ethylene among strains. In cut dahlia 'Karma Thalia,' treatment with 1 $\mu L\,L^{-1}$ ethylene for 16 h does not accelerate flower senescence (Dole et al., 2009). However, cut dahlia flowers might wilt on exposure to ethylene for a long period.

A plant growth regulator, 2-chloroethylphosphonic acid (CEPA), is hydrolyzed to ethylene, phosphoric acid and hydrochloric acid at pH values greater than 5. In *Alstroemeria*, tepal abscission is accelerated by continuous absorption of CEPA, but exposure to 2 $\mu L\,L^{-1}$ ethylene for 15 h has no effect on hastening tepal abscission (Wagstaff et al., 2005). Therefore, the response of dahlia flowers to CEPA was also evaluated.

There are many chemical treatments for extending the vase life of cut flowers (Halevy and Mayak, 1981). In cut flowers where senescence is related to ethylene, silver thiosulfate complex (STS),

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http://dx.doi.org/10.1016/j.postharvbio.2013.07.036

an inhibitor of ethylene action, is effective in extending vase life (Veen, 1979; Mor et al., 1984; Whitehead et al., 1984a; Ichimura et al., 1998; Shimizu and Ichimura, 2005). Another ethylene action inhibitor, 1-methylcyclopropene (1-MCP), can extend the longevity of cut carnation, *Delphinium* and sweet pea flowers in air (Serek et al., 1995; Ichimura et al., 2002). Cytokinins delay senescence of leaves (Richmond and Lang, 1957; Gan and Amasino, 1995) and cut flowers (Halevy and Mayak, 1981). In cut carnation flowers, vase life is extended by about 5 days by treatment with kinetin, a synthetic cytokinin (Eisinger, 1977). In addition, kinetin reduces both ethylene production and sensitivity to ethylene in cut carnations (Eisinger, 1977). On the other hand, thidiazuron, a synthetic cytokinin, is effective in extending iris vase life by up to 1.5 days relative to controls (Macnish et al., 2010); iris is classified as an ethylene insensitive flower (van Doorn, 2001). These results suggest that cytokinins delay petal senescence in both ethylene sensitive and insensitive flowers.

Sugars also extend the vase life of cut flowers (Halevy and Mayak, 1981). In cut dahlia 'Purple Gem' flowers, a solution of 10% glucose plus silver nitrate extends the vase life slightly (Lukaszewska, 1986). Commercial preservative solutions also extend the vase life of cut dahlia 'Eveline' or 'Karma Thalia' (Swart, 1986; Dole et al., 2009). Cold storage for 48 h is not harmful to the cut dahlia 'Eveline' and prolongs its vase life (Swart, 1986), but cold storage for one week decreases the vase life of cut dahlia 'Karma Thalia' (Dole et al., 2009).

In this study, to clarify the relationship between ethylene and flower senescence, we studied ethylene production and sensitivity to ethylene by continuous exposure of cut dahlia flowers to ethylene, and investigated the effect of STS, 1-MCP and the cytokinin 6-benzylaminopurine (BA) on vase life. The best method for BA application was also investigated because Mayak and Halevy (1970) reported that application of BA by immersing flower stems is ineffective in extending the vase life of cut roses, but application directly to the flower buds delays senescence.

2. Materials and methods

2.1. Plant materials

The dahlia (*Dahlia × hybrida*) 'Kokucho,' a semi-cactus petal form, was used in experiments unless otherwise stated.

For ethylene measurements, STS treatments for flowers and BA treatments for florets, dahlias were grown under long daylength (16:00–20:00) conditions in a greenhouse (11 °C minimum and 30 °C maximum) in Tsukuba, Japan (E 140°05′, N 36°02′) from December to February. The flowers were harvested at the stage when the first row of petals from the outside had opened. To analyze silver concentration, the dahlia flowers were grown under the same conditions in June. The flowers were harvested at the stage when the first to third rows of petals from the outside had opened.

To evaluate sensitivity to ethylene, cut dahlia 'Kokucho' flowers were wet-transported using tap water from Yamagata Prefecture to the NARO Institute of Floricultural Science (NIFS) in November. It took 1 day from harvest to arrival at NIFS. After transport, the first to third row of petals from the outside had already opened. To evaluate BA treatments of 'Kokucho' flowers, cut dahlia flowers were wet-transported using tap water from Nagano Prefecture to NIFS in February. It took 2 days from harvest to arrival at NIFS. After transport, the first or first to second rows of petals from the outside had already opened. For BA treatments of white 'Kamakura' flowers, a formal decorative petal form, and pink 'Michan' flowers, a pompom petal form, cut dahlia flowers were wet-transported using tap water from Yamagata Prefecture to NIFS in May. It took 1 day from harvest to arrival at NIFS. After transport, the first to third rows of petals from the outside had already opened.

2.2. Evaluation conditions and standard of vase life of dahlia florets or flowers

The florets or flowers were held at 23 °C, 70% relative humidity with a 12 h photoperiod having a 10 μmol m^{-2} s^{-1} irradiance from cool-white fluorescent lamps throughout the experimental period unless otherwise stated. The longevity of cut flowers was defined as the time from treatment to when several petals had abscised, or two-thirds of the petals of whole flowers had wilted or discolored. The vase life of a floret was defined as the time from treatment to when the petals wilted or discolored.

2.3. Evaluation of sensitivity to ethylene

The cut flowers were recut to 15 cm, and leaves were removed. The flowers were placed in individual vessels containing distilled water (DW) and placed in a 70 L transparent acrylic box fitted with a septum through which ethylene was introduced to achieve a concentration of 2 or 10 μL L^{-1}. The control chamber was not treated with ethylene. The box was kept at 23 °C with a 12 h photoperiod having a 10 μmol m^{-2} s^{-1} irradiance from cool-white fluorescent lamps throughout the experimental period. After 24 h, the boxes were opened for 5 min to release the ethylene. Then, the boxes were closed and ethylene was introduced to achieve a concentration of 2 or 10 μL L^{-1}. This process was continued until wilting or abscission of petals. Sensitivity to ethylene was evaluated based on the elapsed time from the start of ethylene treatment to petal wilting or abscission.

The cut end of stems was immersed in DW (control), 0.15 g L^{-1} citric acid monohydrate, or 1 μL L^{-1} or 10 μL L^{-1} CEPA (Nissan Ethrel 10, Nissan Chemical Industries, Tokyo, Japan) continuously until petal wilting or abscission. The response of dahlia flowers to CEPA was evaluated using the elapsed time from the start of CEPA treatment to petal wilting or abscission. The weights of the cut flowers and the vessels with DW were recorded prior to placing the cut flower into the vessel. Weights were recorded daily during the experimental period.

2.4. Measurement of ethylene production

A dahlia flower is a capitulum inflorescence, which is composed of many florets. Therefore, the stage of development of florets is different for each row. We measured ethylene production from the first row of florets from opening to senescence. The six florets in the first row were used for measurement of ethylene production per flower. A floret is composed of a ray petal, a style, an ovary, stamens and a calyx. The floret was divided into calyx, ovary and ray petal with style and stamens because style and stamens were too small to remove from the ray petal without wounding. Ray petals and a receptacle were placed in a vessel (148 mL), whereas ovaries and calyxes were placed in a test-tube (14.8 mL). All vessels were sealed and kept at 23 °C. Two hours later, a 1 mL gas sample was withdrawn into a syringe; ethylene concentration was determined using a model GC-14B gas chromatograph (Shimazu Corporation, Kyoto, Japan), equipped with an alumina column and flame ionization detector. Measurements were made for three replications.

2.5. Treatment with silver thiosulfate complex (STS) for flowers

The cut flowers were recut to 10 cm, and leaves were removed. The cut ends of stems were immersed in 0.2 mM STS for 24 h,

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