Postharvest Biology and Technology 137 (2018) 1–8

Contents lists available at ScienceDirect

Postharvest Biology and Technology

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

Physiological and biochemical modifications by postharvest treatment with sodium nitroprusside extend vase life of cut flowers of two gerbera cultivars

Soheila Shabanian[a], Maryam Nasr Esfahani[a,*], Roya Karamian[b], Lam-Son Phan Tran[c,d,**]

[a] Department of Biology, Faculty of Science, Lorestan University, Khorramabad, 68137-17133, Iran
[b] Department of Biology, Faculty of Science, Bu-Ali Sina University, P. O. Box 65175/4161, Hamedan, Iran
[c] Institute of Research and Development, Duy Tan University, 03 Quang Trung, Da Nang, Viet Nam
[d] Signaling Pathway Research Unit, RIKEN Center for Sustainable Resource Science, 1-7-22, Suehiro-cho, Tsurumi, Yokohama 230-0045, Japan

ARTICLE INFO

Keywords:
Floral senescence
Gerbera jamesonii
Nitric oxide
Oxidative stress
Postharvest life
Water relation

ABSTRACT

Senescence is a major problem of gerbera (*Gerbera jamesonii*) cut flowers limiting their long-distance transportation and subsequent marketing. This study was designed to evaluate whether external application of nitric oxide (NO), provided through 150 μM sodium nitroprusside (SNP), could extend the vase life of gerbera cut flowers, as well as the potential physiological and biochemical mechanisms involved. We used two gerbera cultivars 'Bayadère' and 'Sunway'; watered 'Bayadère' cut flowers have a better performance than watered 'Sunway' cut flowers. NO extended the vase life of cut flowers of both cultivars as compared with their respective control treated with water alone, with 'Sunway' showing better postharvest performance than 'Bayadère'. Application of SNP in vase solution resulted in a decrease in proline content in the stems of cut flowers of both cultivars, providing evidence for alleviation of water deficit in SNP-supplied cut flowers. Improved postharvest performance of SNP-treated gerbera cultivars could be attributed to increases in total phenol and flavonoid contents, which resulted from decreased polyphenol oxidase activity and increased phenylalanine ammonia-lyase activity. A decline in malondialdehyde accumulation in the stems of SNP-treated cut flowers was greater in 'Sunway' flowers than in 'Bayadère' flowers, which was ascribed to the better performance of antioxidant systems in SNP-treated 'Sunway' flowers to reduce the adverse effect of oxidative stress. Taken together, exogenous NO might be promising approaches to improve postharvest performance of flowers.

1. Introduction

Flower longevity and quality are economically important factors for the marketing of ornamental cut flowers because their postharvest short vase life and poor quality lead to difficulties for long-distance transportation, and decreased market value (Gebremedhin et al., 2013; Hegazi, 2016). Senescence is the main reason for the short vase life and poor quality of cut flowers (Mansouri, 2012; Saeed et al., 2016), which is associated with ultrastructural modifications, increase in lipid peroxidation and membrane leakage, increased respiration rate, enhanced activities of hydrolytic enzymes, changes in various cell organelles and degradation of macromolecules (Mansouri, 2012; Rani and Singh, 2014). In addition, it has been well-established that various factors are involved in induction of senescence of cut flowers (Reid and Jiang, 2012). These factors include genotype, environmental factors (e.g. light, temperature, humidity, water relations and nutritional status), microbial activities, production and sensitivity to ethylene and oxidative stress (Ebrahimzadeh et al., 2008; Reid and Jiang, 2012). Therefore, to maintain the quality of cut flowers and to extent their vase life, convenient and economical techniques to slow senescence are needed.

Gerbera (*Gerbera jameonii* Bolus), belonging to the Asteraceae family, is one of the ten most important popular commercial cut flowers in the world (Cardoso and Teixeira da Silva, 2013), securing the number fourth rank among cut flowers according to the global trends in floriculture (Nair et al., 2006). There is a considerable demand for gerbera cut flowers in both domestic and export markets because they are attractive flowers with large size, and are available in different shades and hues (Jamshidi et al., 2014). However, short vase life and poor quality are major problems for export and trade of gerbera, limiting their marketing (Balestra et al., 2005; Liu et al., 2009). Maintenance of the quality and extending the vase life of gerbera cut flowers are required to meet the demands of growers, wholesalers and consumers. Silver nitrate, UV-C irradiation, S-carvacrol, thymol, geranyl

* Corresponding author at: Department of Biology, Faculty of Science, Lorestan University, Khorramabad, 68137-17133, Iran.
** Corresponding author at: Duy Tan University, Institute of Research and Development, 03 Quang Trung, Da Nang, Japan.
E-mail addresses: esfahani.m@lu.ac.ir (M. Nasr Esfahani), son.tran@riken.jp (L.-S.P. Tran).

https://doi.org/10.1016/j.postharvbio.2017.11.009
Received 2 September 2017; Received in revised form 7 November 2017; Accepted 7 November 2017

diphosphate, 8-hydroxyquinoline citrate, sucrose, calcium, nano-silver and essential oils can be used to improve performance of gerbera cut flowers (Ardebili et al., 2013; Darras et al., 2012; De Witte et al., 2014; Lü et al., 2010; Perik et al., 2012; Solgi et al., 2009). However, there is little information with regard to how gerbera cut flowers can extend their vase life in response to these preservative solutions. Studies of chemical preservatives-induced physiological and biochemical mechanisms that improve the vase life of gerbera cut flowers can provide a mechanistic insight into the process, thereby paving the way to breeding or engineering cut flowers with enhanced performance through biotechnological strategies.

Nitric oxide (NO) is a critical signaling molecule that plays a key role in many processes of plant growth and development, as well as plant responses to environmental stresses (Ahmad et al., 2016). It has been well documented that NO acts as a negative regulator during plant senescence (Prochazkova and Wilhelmova, 2011); however, there is very scant information regarding the beneficial effects of exogenously applied NO in delaying senescence of gerbera cut flowers. In the present study, we examined the efficiency of NO, provided using sodium nitroprusside (SNP) (Prochazkova and Wilhelmova, 2011), in extending the vase life of cut flowers of two gerbera cultivars 'Bayadère' and 'Sunway' in a comparative manner using various physiological and biochemical approaches. More specifically, we examined the effects of SNP postharvest treatments on vase life, fresh weight and relative water uptake (RWU), and also on the key biochemical and physiological parameters in gerbera stems, including accumulation of malondialdehyde (MDA), contents of proline, total phenols and flavonoids and total soluble proteins, as well as the activities of important antioxidant enzymes that included catalase (CAT), ascorbate peroxidase (APX), peroxidase (POD), polyphenol oxidase (PPO), superoxide dismutase (SOD) and phenylalanine ammonia-lyase (PAL).

2. Materials and methods

2.1. Flowers and treatments

We initially examined postharvest potential of flowers of ten gerbera cultivars 'Bayadère', 'Double Dutch', 'Picobella', 'Rosalin', 'Dalanco', 'Dune', 'Stansa', 'Tropic Blend', 'Sunway' and 'Intense', which were obtained from a local commercial greenhouse (Pakdasht, Tehran). Among the examined gerbera cultivars, 'Bayadère' and 'Sunway' scored the highest and lowest postharvest performance, respectively, which were then selected for further studies.

'Bayadère' and 'Sunway' flowers were cut at commercial maturity stage, i.e. when the two outer whorls in the floral head showed mature statement and uniformity in size and quality. After harvest, the flowers were immediately placed upright in buckets filled with water and transferred to the laboratory, where the flower stem ends were re-cut under water to remove air emboli to prevent vascular blockage. After re-cutting, all flower stems were in similar length of 40 cm. Subsequently, cut flowers of each cultivar were separately divided into three groups for the following treatments:

(i) SNP-24 h treatment: the first group of cut flowers were kept in vase solution containing 150 μM SNP that was identified as the optimal SNP concentration for both cultivars in a preliminary experiment. Twenty-four hours after SNP treatment, cut flowers were placed in distilled water until the end of the experiment.
(ii) SNP-48 h treatment: the second group of cut flowers were kept in vase solution containing 150 μM SNP. Forty-eight hours after SNP treatment, cut flowers were placed in distilled water until the end of the experiment.
(iii) Control: the third group of cut flowers were kept in distilled water and used as control.

The experiments were conducted under controlled conditions (12 h photoperiod at a photosynthetically activated radiation of 10 μmol m^{-2} s^{-1} provided by fluorescent lamps, 20 ± 1 °C, and relative humidity of 60 ± 5%). The end of vase life of cut flowers was considered the time point when flowers showed symptoms of petal wilting (Gerasopoulos and Chebli, 1999). RWUs of cut flowers were determined on the basis of initial fresh weight over the first seven days of experiments as previously described by Lü et al. (2010). The whole stems of cut flowers were harvested at days 0, 3 and 7 of vase life in three biological replicates, frozen immediately in liquid nitrogen and stored at −80 °C. Before analysis, all stems of cut flowers in each replicate were ground in liquid nitrogen using a mortar and pestle to obtain homogeneous samples.

2.2. Extraction and determination of the contents of proline, MDA, total phenols and flavonoids

0.5 g homogeneous samples of the stems were mixed with 5 mL of 3% aqueous 5-sulfosalicylic acid. After centrifugation at 11,500 × *g* for 15 min, the supernatant (1 mL) was used to measure proline content at 520 nm (Bates et al., 1973). MDA content, which was used as an indicator for lipid peroxidation of the stems was measured using thiobarbituric acid (TBA) according to Heath and Packer (1968). Total phenol and flavonoid contents of flower stems were determined using the Folin-Ciocalteu procedure (Singleton and Rossi, 1965) and the colorimetric assay (Zhishen et al., 1999), respectively. All the data were expressed on a fresh weight basis.

2.3. Extraction and determination of CAT, APX, PPO, POD, SOD and PAL activities and total soluble protein content

For determination of total soluble proteins and the activities of enzymes, the supernatants were obtained from homogeneous samples of the stems as previously described by Nasr Esfahani and Mostajeran (2011). Total soluble protein content was estimated using the Bradford reagent (Bradford, 1976). The activities of enzymes were determined according to standard methods previously reported for CAT (EC 1.11.1.6) (Aebi, 1984), APX (EC 1.11.1.11) (Nakano and Asada, 1981), PPO (EC 1.10.3.1) (Pütter, 1974), POD (EC 1.11.1.7) (Chance and Maehly, 1955), SOD (EC 1.15.1.1) (Giannopolitis and Ries, 1977) and PAL (EC 4.3.1.24) (Koukol and Conn, 1961). Enzyme activities were expressed in unit on a fresh weight basis.

2.4. Statistical analysis

For determination of vase life and RWU parameters, five biological replicates ($n = 5$ replicates/cultivar; each replicate had five cut flowers) from each treatment (distilled water, SNP-24 and SNP-48 h) were used. For measurements of proline, MDA, total soluble protein, total phenol and flavonoid contents, as well as the activities of antioxidant enzymes, three biological stem replicates ($n = 3$ replicates; each replicate had five stems from five cut flowers) from each treatment (distilled water, SNP-24 and SNP-48 h) for each gerbera cultivar and each harvesting time (days 0, 3 and 7 of vase life) were used. Data were subjected to a three-way ANOVA, and significant differences between treatments, the times after harvest, and the gerbera cultivars were measured by Duncan's multiple range test ($P \leq 0.05$). All statistical analyses were determined using SPSS software package 16.0.

3. Results

3.1. Effects of postharvest SNP treatment on vase life and fresh weight of gerbera cultivars

A comparison of vase life of watered 'Bayadère' and 'Sunway' cultivars revealed that 'Bayadère' had an 82% longer vase life than 'Sunway' cut flowers (Fig. 1). The vase life of cut flowers kept in vase