Pre- and Postharvest Practices for Optimizing the Postharvest Quality of Cut Delphinium, Dahlia, and Sunflower.

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Abstract

The primary objective of this research was to identify practices by which specialty cut flower growers can extend the vase life of cut delphinium, sunflower, and dahlia. Experiments investigated the effects of nitrogen fertilization rate on delphinium 'Guardian Mix' and the effects of deficit irrigation on delphinium 'Guardian Blue.' 'Guardian Mix' plants produced marketable cut stems at nitrogen rates as low as 50 $mg \cdot L^{-1}$. Deficit irrigation did not change vase life, stomatal conductance, or transpiration rates of delphinium 'Guardian Blue.' Studies tested the effects of foliar calcium applications or benzyladenine application on sunflower 'Moulin Rouge' and 'Procut Lemon.' Calcium did not change the vase life, stomatal conductance, or transpiration rates of either sunflower cultivar. Benzyladenine applied as a preharvest spray or a postharvest dip did not alter vase life of sunflower 'Moulin Rouge' or 'Procut Lemon.' Transpiration rate and conductance rates of sunflowers significantly decreased in the first three days after harvest. In both sunflower experiments, vase life of 'Moulin Rouge' was shorter than vase life of 'Procut Lemon.' Benzyladenine was also applied to dahlia 'Park Princess' and 'Karma Yin Yang' cut flowers. Benzyladenine did not change dahlia vase life. Dahlia 'Park Princess,' 'Bride to Be,' 'Cherish,' and 'Lollipop' cut flowers were not sensitive to exogenous ethylene. Further experiments tested the effect of flower stage at harvest, vase water temperature, or preharvest fungicide application on dahlia 'Park Princess' and 'Karma Yin Yang' cut flowers. Vase life of 'Park Princess' flowers was extended when flowers were

harvested before fully open, but ‘Park Princess’ flowers harvested at budbreak failed to open completely after harvest. ‘Karma Yin Yang’ cut flower vase life did not differ when flowers were harvested at different stages. Placing cut dahlias in hot vase water had varied effects, but did not extend vase life of either cultivar. Fungicide applications extended vase life of ‘Park Princess’ flowers. However, the use of fungicide is not necessary to prevent postharvest fungal infection in cut dahlias. The results of all experiments indicate that optimal handling practices vary between cut flower taxa and that factors determining cut flower vase life are complex.

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General Audience Abstract

The primary objective of this research was to identify practices by which specialty cut flower growers can extend the vase life of cut delphinium, sunflower, and dahlia. Experiments investigated the effects of nitrogen fertilization rate on delphinium 'Guardian Mix' and the effects of decreased irrigation on delphinium 'Guardian Blue.' 'Guardian Mix' plants produced marketable cut stems at low nitrogen rates. Reducing irrigation did not change vase life of delphinium 'Guardian Blue.' Studies tested the effects of foliar calcium applications or the use of a plant growth regulator on sunflower 'Moulin Rouge' and 'Procut Lemon.' Calcium did not change the vase life of either sunflower cultivar. A plant growth regulator applied as a preharvest spray or a postharvest dip did not alter vase life of sunflower 'Moulin Rouge' or 'Procut Lemon.' Gas exchange controlled by leaves of sunflowers significantly decreased in the first three days after harvest. In both sunflower experiments, vase life of 'Moulin Rouge' was shorter than vase life of 'Procut Lemon.' A plant growth regulator was also applied to dahlia 'Park Princess' and 'Karma Yin Yang' cut flowers. The plant growth regulator did not change dahlia vase life. Dahlia 'Park Princess,' 'Bride to Be,' 'Cherish,' and 'Lollipop' cut flowers were not sensitive to exogenous ethylene gas. Further experiments tested the effect of flower stage at harvest, vase water temperature, or preharvest fungicide application on dahlia 'Park Princess' and 'Karma Yin Yang' cut flowers. Vase life of 'Park Princess' flowers was extended when flowers were harvested before fully open, but 'Park Princess' flowers harvested at a very early flower stage failed to open completely after harvest. 'Karma Yin Yang' cut flower vase

life did not differ when flowers were harvested at different flower stages. Placing cut dahlias in hot vase water had varied effects, but did not extend vase life of either cultivar. Fungicide applications extended vase life of 'Park Princess' flowers. However, fungicide was not needed to control postharvest fungal infection in cut dahlias. The results of all experiments indicate that optimal handling practices vary between cut flower taxa and that factors determining cut flower vase life are complex.

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Chapter 1: Literature Review

Cut flower industry

The majority of the cut flowers sold in the U.S. are grown overseas, where labor is cheaper and climates may be more favorable to flower production. In 2014, the wholesale value of U.S.-grown cut flowers was $354 million (USDA-NASS, 2015). The value of imported cut flowers during the same year was $669.8 million (Global Agricultural Trade System, 2016). To be profitable, American cut flower growers specialize in growing crops that are difficult to ship or that are short-lived after harvest; these flower crops are generally referred to as specialty cut flowers.

Market research indicates that consumers of cut flowers are willing to pay extra for flower arrangements with longer vase life and arrangements with a longevity guarantee (Rihn et al., 2014). Consumers, especially those in younger generations, are dissatisfied with the short longevity of flowers (Rihn et al., 2011). Sales of cut flowers may be increased by improvements in postharvest longevity.

Postharvest handling

Flower growers tend to harvest cut flowers in the morning, but flowers harvested later in the day may have a more favorable carbohydrate status and greater postharvest longevity (Ahmad et al., 2014). Much of the existing research on cut flowers focuses on treatment and handling of flower stems after they have been removed from plants. Experiments usually investigate transport and storage of cut stems. Cut flowers are held either wet (in water) or dry (usually in boxes) and are typically stored in a dark cooler (Ahmad and Dole, 2014; Bosma and

Dole, 2002; Dole et al., 2009). After transport and storage, some studies apply a short-term "pulse" treatment by placing stems in a liquid solution. Pulsing treatments are often applied for 4 hours but can last for up to 24 hours. Pulsing solutions vary and include commercially available hydrating solutions, ethylene inhibitors, sugars, biocides, and mineral nutrients (Ahmad and Dole, 2014; Bosma and Dole, 2002; Perik et al., 2014). Finally, after the pulse treatment, flower stems are placed in a vase solution, which can be deionized (DI) water, a commercial holding solution, or a solution containing sucrose and/or mineral nutrients (Dan and Griffith, 1990; Dole et al., 2009; Torre et al., 1999). Sometimes, to simulate florists' use of the cut stems, the impact of floral foams in the vase is also evaluated (Ahmad and Dole, 2014; Bosma and Dole, 2002).

Optimal postharvest handling protocols differ by plant species and even among cultivars, so cultivar-specific recommendations should be researched (Ahmad and Dole, 2014; Barbosa et al., 2015; Clark et al., 2010). For a more extensive review of the impact of postharvest treatments on cut flower vase life, see van Meeteren (2009).

Production environment

While the handling of cut flowers after harvest can alter the vase life of cut flowers, the production environment also plays a role in the postharvest quality and longevity of cut flowers. Temperature, relative humidity, light intensity, and vapor pressure deficit have all been shown to have an effect on the vase life of cut roses (*Rosa hybrida* L.; Marissen, 2005) and freesia (*Freesia hybrida*; Slootweg, 2005). Fertilization can improve the yield and postharvest life of chrysanthemum (*Chrysanthemum morifolium* R.) flowers (Barbosa et al., 2015; Fan et al., 2015). Seasonal variations in temperature, relative humidity, and vapor pressure deficit (VPD) of the

growing environment also contribute to longevity differences. Roses grown in winter tend to have shorter vase life than roses grown at other times of the year (Grossi et al., 2003; In et al., 2016a). Irrigation regimes can also alter cut flower quality and vase life; mild drought stress can improve flower quality but severe drought may inhibit flower production altogether (Chuang and Chang, 2012). Research can help growers understand optimal conditions for producing cut flowers of the highest quality.

Vase life

The length of vase life for cut flowers is extremely variable. Research reports vase life ranging from 1 day to over 50 days for different cut flower taxa (Clark et al., 2010; Elibox and Umaharan, 2010). The external appearance of flowers at the time of harvest is not always an accurate indicator of postharvest longevity (In et al., 2016b).

The criteria for terminating vase life vary in the literature (Halevy and Mayak, 1981). Many recent studies consider vase life to end when senescence symptoms appear on 50% or more of the flower parts on a cut stem (Ahmad and Dole, 2014). Symptoms signaling the end of vase life vary (Fanourakis et al., 2013). Senescence symptoms include: wilting or discoloration of petals and/or leaves, petal abscission, bending of stems, or infection by pathogens such as *Botrytis cinerea* (Ahmad and Dole, 2014; Perik et al., 2014; Slootweg and Körner, 2009; Tata and Wien, 2014; van Doorn, 2002).

Cut flower senescence

After a flower is cut, it undergoes many physiological changes as it senesces. To better interpret the differences in vase life of cut flowers, it is important to understand the senescence processes that occur while the flower is in the vase.

At a molecular level, signaling pathways that regulate flower senescence are not completely understood (van Doorn, 2015; van Doorn and Woltering, 2008). The triggers for programmed cell death in cut flowers are still unknown (van Doorn, 2015). Plant cells will undergo changes in enzyme activity during senescence (Halevy and Mayak, 1981). In freesia cut flowers, electrolyte leakage and malondialdehyde (MDA) content of stems increased throughout vase life, indicating the breakdown of cells (Shu et al., 2010). Fan et al. (2015) saw a similar rise in MDA content of cut chrysanthemum flowers during vase life. For flowers that lose their petals, petal abscission occurs after formation of cells in the abscission zone. Rapidly maturing cells in the abscission zone lead to early loss of petals during vase life of sunflowers (*Helianthus annuus* L.; Tata and Wien, 2014).

Other senescence processes occur on a larger scale. As a flower senesces, mineral nutrients are remobilized and transported to different flower parts (Jones, 2013). Over their vase life, freesia cut flowers showed declines in both soluble sugars and proteins in the stems (Shu et al., 2010). Metabolic processes in cut stems decline during senescence. Cut freesia respiration rates peaked just after flower buds opened in the vase, then steadily declined thereafter (Shu et al., 2010). Similar patterns in respiration were observed in cut flowers of lily (*Lilium* hybrids; Kim et al., 2005). Many cut flowers produce ethylene during senescence, often in increasing amounts. Roses, snapdragons (*Antirrhinum majus* L.), sunflower, delphinium (*Delphinium hybrida*), and peony (*Paeonia suffruticosa*) all showed increased ethylene production during postharvest vase life (Mensauli-Sodi and Ferrante, 2005; Nabigol, 2012; Philosoph-Hadas et al.,

1996a; Wang et al., 2014). Dahlia (*Dahlia ×hybrida*) flowers produced ethylene at a steady rate during postharvest evaluation (Shimizu-Yumoto and Ichimura, 2013).

Water relations

For cut flowers, water stress contributes to senescence and shortened vase life. Usually, changes in water content of cut flowers are reflected in the weight of the cut stem itself; stem fresh weight of roses and lilies increases at the start of the vase life, and then slowly declines thereafter (Lü et al., 2011). Declining water content can lead to wilting and abscission of flower petals and leaves.

Water content of cut stems is generally a balance between water loss via transpiration and water uptake from the vase solution. Plants lose water vapor through transpiration. Stomata open to allow gas exchange, most importantly the exchange of carbon dioxide and water vapor. The loss of water vapor through stomata creates negative water potential and drives water uptake through the stem. In the case of cut flowers, water will tend to move from the vase solution where water potentials are higher and travel up the stem to areas of lower water potential. Leaves account for the majority of water uptake in cut stems, as stomata tend to be concentrated on leaves; however, the flower and stem also drive some water uptake, especially in species such as carnation (*Dianthus caryophyllus*) where stomata are also located on stems (Carpenter and Rasmussen, 1974). Shortened vase life in cut rose cultivars has been correlated with high rates of transpiration (In et al., 2016a; Mayak et al., 1974); in this case water uptake is not sufficient to replace water lost through the stomata. Chemical means can be used to induce stomatal closure and decrease transpiration rates. Tea-seed saponins and nano-silver are two treatments that can

induce stomatal closure in cut roses, leading to extended vase life (Ichimura et al., 2005; Lü et al., 2010).

Water uptake in cut flowers tends to follow a diurnal pattern where maximum uptake occurs during the day and minimum uptake occurs at night (Lü et al., 2011). Generally, cut flowers exhibit a pattern of decreasing water uptake throughout their vase life (Barrs and Weatherley, 1962; Lü et al., 2011; Mayak et al., 1974). In a comparison of *Anthurium* cultivars, the longest-lived cultivars maintained above average water uptake rates for longer time periods than shorter-lived cultivars (Elibox and Umaharan, 2010).

Often, decreasing water uptake is the result of stem cavitation: the water flow in the xylem is disrupted so that the flower can no longer take up water. In freshly cut rose stems, about 50% of xylem conduits were functional, but after 6 days the amount of functional xylem conduits decreased to less than 10% (Dixon and Peterson, 1989). Stem cavitation can be a result of air embolisms in the stem, microbial buildup, or physical blockages from particles in the vase solution (Dixon et al., 1988; Williamson and Milburn, 1995). Blockages within the stem tend to be concentrated near the cut end of the stem. In roses, all xylem blockages occurred in the 200 mm of stem closest to the cut end (Dixon and Peterson, 1989). Recutting flower stems can be a simple way to overcome cavitation issues at the end of the stem.

Postharvest water relations of cut flowers may be influenced by the status of plants before flower harvest. Roses grown in a springtime greenhouse environment tended to develop stomata that were more responsive to environmental changes, leading them to have decreased postharvest transpiration rates and longer vase life than roses grown in other seasons (In et al., 2016a). Irrigation regimes can also alter the responsiveness of stomata. Growing lisianthus (*Eustoma*

grandiflorum) under moderate water deficit reduced the transpiration rates and extended vase life of 'Piccolo Lime Green' flowers, but did not alter these parameters in 'Ex Rosa Green' flowers (Chuang and Chang, 2012). However, when imposing deficit irrigation, growers should beware of declines in flower yield from plants experiencing drought stress.

Sucrose and carbohydrates

In some cases, stored sugars in cut flowers correlate with the length of vase life (Locke, 2010; van Doorn and Han, 2011). Arrom and Munné-Bosch (2012) found that endogenous sugar levels in lilies were important in maintaining stems after harvest, and vase life was extended when sucrose in the vase solution was used to supplement endogenous sugars. Mehran et al. (2008b) applied sucrose as a spray application during rose production and found that sucrose treatments delayed browning and extended vase life. Adding sucrose to the vase solution can extend the vase life of chrysanthemum, gerbera (*Gerbera jamesonii*), and delphinium (Fan et al., 2015; Ichimura et al., 2000; Perik et al., 2014), but this is not the case for all cut flowers. For instance, sunflowers do not benefit from the addition of sucrose to the vase solution (Mensauli-Sodi and Ferrante, 2005).

The mechanism by which sucrose supplementation can extend vase life is unclear. Sucrose may act to decrease ethylene sensitivity in cut chrysanthemum and *Dendrobium* flowers (Ichimura et al., 2000; Pattaravayo et al., 2013). Sucrose may alter hormone levels within lily flowers (Arrom and Munné-Bosch, 2012). In general, ethylene-sensitive cut flowers benefit more from sugar supplementation, but ethylene-insensitive species can also benefit from exogenous sugars (van Doorn, 2004).

Plant nutrition

Mineral nutrition of a plant can also impact senescence processes. Research indicates that plant nutrients delivered during production impact the quality and longevity of flowering plants after they leave the production facility (Roude et al., 1991; Starkey and Pedersen, 1997) and may also impact postharvest longevity of cut flowers (Barbosa et al., 2015; Steduto et al., 2000; Torre et al., 1999). Osorio et al. (2009) found that increasing the nutrient supply to tulips (*Tulipa* spp.) during production increased the vase life of the flowers.

Providing plants with optimal nutrition via fertilization could allow growers to produce higher quality cut flowers. For instance, nitrogen plays many important roles in plant cells, including as a constituent of proteins and chlorophyll (Hawkesford et al., 2012). Thus, nitrogen fertilization can greatly increase the ability of a plant to perform photosynthesis, thereby producing and storing sugars. Marchese et al. (2005) saw increased foliar chlorophyll and nitrogen, as well as higher photosynthetic rates in lisianthus receiving higher doses of nitrogen fertilizer during production. Nitrogen fertilization has been shown to improve the qualities of individual flower stems by increasing stem length and reducing leaf yellowing (Ahmad et al., 2012). Fertilizing with nitrogen can also improve the overall flower yield from a single plant (Ahmad et al., 2012; Smith et al., 1998).

In addition to nitrogen, other plant nutrients play a role in flower quality. Supplying both macronutrients and micronutrients has been shown to improve quality and longevity of a variety of cut flowers (Barbosa et al., 2015; Burchi et al., 2010; Khoshgoftarmanesh et al., 2008; Saeed et al., 2013). Calcium may be of particular interest for delaying flower senescence. Calcium is an essential plant nutrient with many functions within the plant. Calcium is an important

constituent of compounds that stabilize plant cell walls (Hawkesford et al., 2012). Calcium is involved in the stomatal signaling pathway, which controls the opening and closing of stomata, and thus the loss of water vapor via the leaves (Hetherington and Woodward, 2003). Calcium also acts as a secondary messenger in plants, and may play a role in hormone-driven senescence processes (Philosoph-Hadas et al., 1996a).

When applied during production, calcium can extend vase life, prevent stem bending, and strengthen stems of cut flowers (Gerasopoulos and Chebli, 1999; Li et al., 2012; Mehran et al., 2008a; Nabigol, 2012; Starkey and Pedersen, 1997). These benefits may rely on correct timing of calcium applications during production (Deljou and Gholipour, 2014). Adding calcium to postharvest vase solutions can delay senescence symptoms such as stem bending in gerbera (Gerasopoulos and Chebli, 1999). Similar to the other treatments discussed, responses to calcium treatment will vary between species and cultivars of cut flowers (Deljou and Gholipour, 2014; Gerasopoulos and Chebli, 1999).

Identifying appropriate fertilization rates is important, as nutrient levels that are too high can be detrimental to plant quality. In *Sandersonia*, high nitrogen rates led to shorter stems with lower dry weights (Clark and Burge, 1999). In potted chrysanthemum, several cultivars exhibited decreasing longevity with increasing rates of nitrogen fertilization (Roude et al., 1991). When growers applied high levels of potassium to chrysanthemums, the flower stem quality was compromised by chlorosis, a symptom of the resulting magnesium deficiency (Branson et al., 1968).

With increasing pressure to conserve water, a growing body of research investigates irrigation with recycled wastewater that has a higher concentration of salts. Increasing the salinity of irrigation water can lead to shorter stems, stems with decreased fresh weight, smaller

leaf area, and necrotic edges on leaves (Baas et al., 1995; Cabrera and Perdomo, 2003; Carter and Grieve, 2010; Steduto et al., 2000). However, many cut flower taxa still produce marketable stems when irrigated with high salinity solutions (Grieve, 2011).

Ethylene

Ethylene is a plant hormone that is often associated with senescence. Many cut flower species produce ethylene during senescence (Reid and Wu, 1992; Wang et al., 2014). Some flowers produce increasing amounts of ethylene as they senesce, but this is not true for all taxa (Kumar et al., 2008; Wu et al., 1991a). In ethylene-sensitive species, exposure to ethylene hastens senescence. Petal abscission is a common senescence symptom in ethylene-sensitive species (Brown, 1997; van Doorn, 2002). Levels of ethylene sensitivity vary between different flower species and even cultivars (Woltering and van Doorn, 1988; Wu et al., 1991b). The ethylene sensitivity of a flower can also change as the flower develops (Brown, 1997). Senescence processes may vary between species that are sensitive to ethylene and those that are ethylene-insensitive (Jones, 2013; van Doorn and Woltering, 2008).

In species that are sensitive to ethylene, the use of ethylene inhibitors can protect against the ill effects of ethylene exposure. 1-methylcyclopropene (1-MCP) and silver thiosulfate (STS) inhibit ethylene action and are commonly added to pulsing solutions or vase solutions (Reid and Jiang, 2012).

Gibberellins

In the floriculture industry, plant growth regulators (PGRs) are often used in order to manage the growth and development of ornamental crops. PGRs can also be useful in producing

cut flowers with optimum vase life. Many PGRs have been tested as pulse solutions or in the vase solution for cut flowers. When added to the vase solution, gibberellic acid (GA_3) can extend vase life by delaying leaf yellowing. This is of particular interest for lily (Rabiza-Świder et al., 2012) and calla lily (*Zantedeschia* Spreng.; Janowska and Stanecka, 2011). Gibberelic acid in the vase solution also extended the vase life of gerbera (Danaee et al., 2011). Products that are a combination of gibberellins and benzyladenine (BA) can also improve the vase life of some lilies (Kim et al., 2005; Whitman et al., 2001). Combining GA_3, BA, ethephon, and calcium ions in the vase solution extended the vase life of tulips (van Doorn et al., 2011).

Cytokinins

Postharvest applications of cytokinins are another option for delaying senescence. Benzyladenine (BA) has been tested on a variety of species using several application methods. Benzyladenine included in the vase solution extended the vase life of gerbera (Danaee et al., 2011) and gladiolus (*Gladiolus hortulanus* L.) (Singh et al., 2008), but did not affect flower opening of iris (Van Doorn et al., 2013). As a pulsing solution, BA helped alstroemeria (*Alstroemeria hybrida* L.) flowers maintain their color after harvest (Hicklenton, 1991), and extended vase life of calla lily flowers (Janowska and Stanecka, 2011). Adding BA to other chemicals in a pulsing solution for tulips protected against tepal abscission (van Doorn et al., 2011). However, using BA as a pulse solution for lily negatively affected the vase life (Rabiza-Świder et al., 2012). Imsabai and van Doorn (2013) found that including BA or thidiazuron (TDZ), another synthetic cytokinin, in either the vase solution or pulsing solution delayed petal blackening of lotus (*Nelumbo nucifera* Gaertn.) flowers. TDZ pulse also extended the flower life of iris (*Iris ×hollandica*; Macnish et al., 2010). Including TDZ in the vase solution delayed

chlorophyll loss in leaves of stock (*Matthiola incana* L.) cut flowers and helped maintain gas exchange by the leaves (Ferrante et al., 2009).

In postharvest experiments, BA has also been applied as a spray or a dip. Benzyladenine sprays applied after harvest extended the vase life of lisianthus (Asil and Karimi, 2010) and improved quality of anthurium (*Anthurium andraeanum*) flowers (Favero et al., 2015). Philosoph-Hadas et al. (1996b) found that BA was not effective as a pulsing solution for *Solidago* flowers, but delayed leaf and flower senescence when applied as a spray. Paull and Chantrachit (2001) tested both spraying and dipping tropical cut flowers in BA, with results varying by flower taxa. Dipping *Grevillea* 'Sylvia' stems in BA extended vase life of the flowers, whereas including BA in the vase solution had no effect on this species (Setyadjit et al., 2004). Spraying or dipping dahlia flowers in BA extended their vase life and was more effective than ethylene inhibitors (Shimizu-Yumoto and Ichimura, 2013). Sunflower heads that had been dipped in BA held their petals more strongly than untreated sunflowers (Tata and Wien, 2014).

PGR application method and timing

While not as widely researched, PGRs applied prior to harvest may also impact the vase life of cut flowers. Whitman et al. (2001) applied products combining GA_{4+7} with benzyladenine to Easter lily (*Lilium longiflorum*) and found that treated stems showed delayed chlorosis in the vase. Preharvest foliar sprays of GA_3 improved water relations and vase life of tulip (Khan et al., 2007). Applying PGRs before harvesting cut flowers may be easier and safer than postharvest applications and deserves further research.

ABA

Other plant hormones may play a role in senescence. Abscisic acid (ABA) is often associated with flower senescence. High endogenous levels of ABA and exogenous applications of ABA have both been linked to accelerated senescence in cut flowers (Halevy and Mayak, 1981). However, ABA also induces closure of stomata, which could help cut stems maintain their water content by limiting water loss by transpiration. When ABA was added to the vase solution, rose stems with leaves that were held in light showed increased longevity (Halevy et al., 1974). However, in the same study roses without leaves or held in a dark environment had a shorter vase life when the vase solution contained ABA (Halevy et al., 1974).

Auxins

The role of auxins in the senescence of cut flowers remains unclear. Research conducted by Rungruchkanont et al. (2007) suggests that auxins may regulate ethylene sensitivity in *Dendrobium* cut flowers. Auxin treatments induced flower opening in iris (Çelikel and van Doorn, 2015). In lotus flowers, auxins led to earlier blackening of petals and stem bending (Imsabai and van Doorn, 2013).

Summary

Flower senescence is a complicated process. Thus, vase life of cut flowers varies depending on many factors. The preharvest environment can determine plant quality, while the postharvest handling procedures and postharvest environment can be altered to manipulate vase life. Unique recommendations should be made for taxa, as variations in vase life and optimal postharvest handling procedures can vary between species and cultivars of cut flowers.

Producing flowers with longer vase life may help to increase the market for cut flowers, since many consumers express dissatisfaction with flowers that have short postharvest longevity.

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Chapter 2: Evaluating the effects of nitrogen rate and irrigation deficits on the postharvest quality and longevity of delphinium cut flowers

Abstract

Proper fertilization and irrigation practices may improve the production and postharvest longevity of cut flowers. Delphinium 'Guardian Mix', a color series containing blue-, lavender-, and white-flowered cultivars, was grown in soilless substrate with nitrogen delivered as constant liquid feed at 50, 150, 250, or 350 $mg \cdot L^{-1}$. Nitrogen rate did not affect postharvest vase life of 'Guardian Mix' cut flowers. Leaf yellowing was not observed in this experiment, suggesting that delphinium 'Guardian Mix' cut flowers can be produced under nitrogen fertilization at rates as low as 50 $mg \cdot L^{-1}$. Cultivars in the 'Guardian Mix' series exhibited differences in vase life, postharvest water uptake, plant height, and chlorophyll index as measured by a SPAD meter. For all 'Guardian Mix' cut stems, postharvest water uptake was positively correlated with vase life. Delphinium 'Guardian Blue' flowers were grown in soilless substrate and subjected to normal irrigation, moderate or severe water deficits from flower initiation until harvest. Irrigation deficits did not alter the vase life of 'Guardian Blue' cut stems. Transpiration and stomatal conductance rates did not change with water stress. Water stress occurring after flower initiation is not detrimental to the postharvest longevity of delphinium 'Guardian Blue.'

Introduction

The short longevity of cut flowers causes consumer dissatisfaction (Rihn et al., 2011). Vase life of cut flowers is variable and is not always indicated by the exterior quality of flowers

at harvest (In et al., 2016). Production factors, including fertilization and irrigation, can alter the quality of cut flowers and may have an effect on postharvest longevity.

Nutritional requirements of cut flowers differ from flowers produced for landscape use. Ahmad et al. (2012) recommended tissue nutrient levels for several specialty cut flower taxa that were lower than recommendations for similar cultivars grown as bedding plants. *Ranunculus asiaticus* had increased flower yields and vase life when grown at the lowest tested nitrogen rate of 50 $mg \cdot L^{-1}$ (Bernstein et al., 2005). Conversely, *Alstroemeria* hybrids produced for cut flowers showed increased flower yields and more rapid flower development with increasing nitrogen rates (Smith et al., 1998).

Handheld chlorophyll meters offer a nondestructive method of assessing plant nitrogen status (Basyouni et al., 2015; Wood et al., 1993). Chlorophyll meters, including SPAD meters, measure the intensity of green color in leaves by measuring light reflectance at red and infrared wavelengths (Takebe et al., 1990). Green coloring in leaves comes from chlorophyll molecules that contain four N atoms; chlorophyll content in leaves has been shown to correlate with tissue N content (Takebe et al., 1990; Wood et al., 1993). Readings taken with a SPAD chlorophyll meter have been shown to correlate with leaf nitrogen content of poinsettia (*Euphorbia pulcherrima* L. (Willd ex. Klotzsch)) and also with leaf chlorophyll content in poinsettia (Schuch et al., 1995) and a variety of agronomic crops (Zhu et al., 2012). Thus, chlorophyll index readings taken using a SPAD meter offer a nondestructive means of assessing leaf greenness and, indirectly, chlorophyll and nitrogen status of the leaves measured. In lisianthus plants grown for cut flowers, increasing nitrogen fertilization rate led to increases in chlorophyll index as measured with a SPAD meter (Marchese et al., 2005).

Irrigation practices can alter vase life of cut flowers. Plants experiencing drought stress often close stomata to minimize water loss from leaves via transpiration. Reductions in transpiration rate (*E*) have been correlated with extended vase life roses (*Rosa hybrida*; Fanourakis et al., 2012; Mayak et al., 1974) and lisianthus (*Eustoma grandiflorum*; Chuang and Chang, 2012). Deficit irrigation has been shown to extend the vase life of zinnia (*Zinnia elegans*; Twumasi et al., 2005) and lisianthus 'Piccolo Lime Green' (Chuang and Chang, 2012). However water deficits led to decreased production of marketable flower stems in cut rose (*Rosa hybrida*; Bolla et al., 2010) and severe water deficit inhibited flower production of lisianthus (Chuang and Chang, 2012).

Delphinium (*Delphinium elatum* L.) is grown as a cut flower. The wholesale value of delphinium flowers in the United States exceeded $6 million in both 2013 and 2014 (USDA-NASS, 2015). Cut delphinium flowers often exhibit yellowing of leaves after harvest (J.M. Dole, personal communication). Optimizing nutrition and water status of cut stems of delphinium may improve vase life and limit leaf yellowing. The objectives of this study were to identify nitrogen fertilization rate and irrigation practices that optimize the vase life and postharvest quality of delphinium cut flowers. In addition, the impact of deficit irrigation on g_s and *E* of delphinium cut flowers was assessed.

Materials and Methods

Fertilization Experiment

Delphinium elatum 'Guardian Mix' containing blue, white, and lavender cultivars, was received as Big Burly® vernalized plugs (Gro'n Sell, Chalfont, PA). After breaking dormancy, plugs were transplanted on February 12, 2015, into gallon pots (3.8 L) containing soilless

substrate (Fafard 52 mix; Sun Gro Horticulture, Agawam, MA). The substrate contained a starter nutrient charge. Plants were grown in a glass greenhouse in Blacksburg, VA. Average temperature was 21.7±0.2°C during the day and 18.9±0.1 at night, average relative humidity was 45.8±0.3%, and average daily light integral was 11.9±0.9 $mol \cdot m^{-2} \cdot d^{-1}$. At transplant, data was collected on plant height, width in two directions, number of branches, and leaf chlorophyll index in SPAD units (SPAD-502; Spectrum Technologies, Aurora, IL).

Beginning after transplant, fertilizer was applied at four different nitrogen levels as constant liquid feed (CLF): 50 $mg \cdot L^{-1}$, 150 $mg \cdot L^{-1}$, 250 $mg \cdot L^{-1}$, 350 $mg \cdot L^{-1}$. This was a split plot experiment with blocking for greenhouse environmental variability (Figure 2.1). In each of four blocks, nitrogen fertilizer rate was applied as a whole plot treatment to allow for easy application. Cultivar (distinguished at flowering by flower color) was applied to split plots. Postharvest data were collected for four to six split plots within each whole plot.

For the first two weeks of the study, fertilizer was mixed from a base solution of 50 $mg \cdot L^{-1}$-N fertilizer (Jack's Professional 5-12-26 Hydroponic fertilizer, 5N-5.2P-21.6K; J.R. Peters, Allentown, PA) with calcium nitrate (Jack's Professional 15-0-0 Calcium Nitrate, 15N-0P-0K-15Ca; J.R. Peters) added to reach the desired nitrogen rate. To avoid differences in calcium rates among treatments, the fertilizer was altered after the initial two weeks of the study. On February 27, all pots were leached with water. From February 28 until the end of the study, the same base solution of 50 $mg \cdot L^{-1}$-N was used with calcium chloride dihydrate (Fisher Scientific, Pittsburgh, PA) added to supply calcium ions at a calcium:magnesium ratio of 2.5:1 and ammonium nitrate (Fisher Scientific) added to reach the desired N-rates. For all fertilizer formulations, the J.R. Peters Laboratory verified nutrient content.

Plants were watered based on volumetric water content (VWC) of the substrate, as measured daily with a Procheck handheld VWC meter (Decagon Devices, Pullman, WA). Each day, substrate VWC was measured for one randomly selected plant in each whole plot and block. For the first two weeks of the experiment, plants of a single fertilizer treatment were irrigated if mean VWC measured for plants in that treatment on a given day fell below 15%. After two weeks, the VWC threshold value was raised to 17% in order to avoid wilting of plants by the end of the day. Each pot received a measured volume of fertilizer in order to achieve a leaching fraction of 10%. Leaching fractions were measured every two weeks and the volume of fertilizer delivered was adjusted as necessary; the observed leaching fraction was 9.4±1.6%. Substrate pH and electrical conductivity (EC) were monitored every 2 weeks after initiating treatment (WAT) using the PourThru method.

Flower harvests began 6 weeks after transplant. Flower stems were harvested before 11 AM when at least one floret had opened so the throat of the floret was visible, but before half the florets had opened (Figure 2.2). Data were collected for all flower stems with properly formed flowers and a stem length of at least 45 cm. At harvest, plant height, width in two directions, number of branches, number of flower stems, stem caliper at 2 cm below the lowest floret, and whole plant fresh weight were measured. Chlorophyll index was measured using a SPAD meter for a lower leaf located 5 cm from the base of the plant and for the uppermost leaf on the flower stalk. Stems were re-cut to 45 cm in length and placed in tap water before being transported to the postharvest evaluation laboratory. For one plant of each cultivar in each split plot, leaves were dried in an oven at 55°C for at least 48 hours. After drying, leaves were ground and sent to QAL (Panama City, FL) for complete nutritional analysis of macro- and micronutrients.

The postharvest laboratory was intended to simulate a consumer's home. Average daily temperature in the postharvest laboratory was 22.9 ± 0.0°C and average humidity was 34.0 ± 0.3%. Fluorescent bulbs provided light for ~8 $h \cdot day^{-1}$ and average instantaneous light intensity during the day was 11.6 ± 4.3 $\mu mol \cdot m^{-2} \cdot s^{-1}$. Flower stems were held individually in vases filled with 500 mL of distilled water and any leaves below the water line were removed from the cut stems.

In the postharvest laboratory, fresh weight of the cut stems was measured at the start and end of vase life. Chlorophyll index of the uppermost leaf was measured with a SPAD meter at four days after harvest. Water uptake was measured seven days after harvest. Vase life was assessed visually once each day in the morning. Vase life was defined as the number of days until petals had abscised or wilted on least half the florets (Figure 2.3). After vase life ended, flower stems were dried in an oven at 55°C for at least 48 hours before measuring dry weight.

Deficit Irrigation Experiment

This experiment was conducted twice. For the first experiment, *Delphinium elatum* 'Guardian Blue' plugs were obtained from a commercial grower (Tagawa Gardens, Centennial, CO). Delphinium 'Guardian Blue' for the second experiment were grown from seed (Ball Horticultural Company, West Chicago, IL). Plugs were transplanted to 8 cm square pots (0.5 L) containing soilless substrate (Fafard 52 mix; Sun Gro Horticulture) until roots had reached the sides and bottom of the pots. While in these containers, delphinium were fertilized with 150 $mg \cdot L^{-1}$ N of 15N-2.2P-12.5K-4Ca-2Mg (Jack's Professional LX; J.R. Peters Inc.) via Constant Liquid Feed (CLF).

After roots had developed, plants were transplanted to trade gallon (2.8 L) containers. Controlled release fertilizer (Osmocote Plus 15-9-12, 5 to 6 month longevity at 21° C; 15N-3.9P-10.0K; Scotts Miracle-Gro, Marysville, OH) was incorporated into soilless media (Fafard 52; SunGro Horticulture, Agawam, MA) at a rate of 4.7 $kg \cdot m^{-3}$. Plants were irrigated overhead with clear water for 3 weeks until roots had reached the sides and bottom of pots and flower buds began to form. Plants were grown in a greenhouse covered in a double layer of polyethylene film in Blacksburg, VA. Delphinium were grown from November 2015 until February 2016. Average temperature was 19.6 ± 0.01°C during the day and 18.4 ± 0.1 at night, average relative humidity was 28.7 ± 0.2%, and average daily light integral was 6.5 ± 0.3 $mol \cdot m^{-2} \cdot d^{-1}$.

Three weeks after the final transplant, 36 plants were selected for uniformity and arranged in a randomized complete block design (Figure 2.4). Two blocks were included to control for greenhouse environmental variation. Water deficit treatments were applied at three levels within each block: control (sufficient water), moderate water deficit, and severe water deficit. Water deficit treatment was applied by subirrigation to six plants sitting in a single tray in each block; each plant was treated as a subsample. Water deficits were applied by monitoring the volumetric water content (VWC) of the substrate daily with a Procheck handheld VWC meter (Decagon Devices). Each day, substrate VWC was measured for two randomly selected pots within each treatment and block. If mean VWC measured was below 18%, 13%, or 8% for the control, moderate and severe water deficits, respectively, subirrigation was applied to all plants receiving that treatment. Due to more rapid water absorption by dry substrate, duration of subirrigation was adjusted for each treatment. Control plants were subirrigated for 9 minutes, while plants under moderate or severe water deficit were subirrigated for 5 or 3 minutes, respectively.

Stomatal conductance (g_s) and transpiration rates (E) of the youngest fully expanded leaf were measured using a LI-6400XT (LI-COR, Lincoln, NE) with the 6400-02 LED chamber attachment or the 6400-40 leaf chamber fluorometer chamber attachment (Appendix A). Measurements were taken at three times for each plant: prior to applying irrigation treatments, 2 weeks after initiating treatment (WAT), and 1 day after flowers were harvested. Environmental conditions during measurement of stomatal conductance and transpiration are listed in Appendix A.

Flower stems were harvested at the same flower stage as delphinium in the nitrogen experiment (Figure 2.2). Flower harvests for the first experiment occurred from January 24 until February 9 and flower stems were cut to 40 cm in length. Harvests for the second experiment occurred from February 24 until March 6; plants were shorter in this experiment and stems were cut to 30 cm in length. Flowers that did not bloom within these time periods were not included in postharvest evaluations. For both experiments, the two uppermost, fully expanded leaves with a width of at least 1 cm were left on the stem; all leaves below these were removed. Any axillary flower buds emerging 30 cm or more below the main flower stalk were also removed. Each flower stem was placed in an individual vase containing 5000 mL of deionized water. Postharvest evaluation occurred in a shaded area of the greenhouse. Average daily temperature in the postharvest evaluation area was 19.3 ± 0.2°C and average relative humidity was 29.6 ± 0.3%. Average instantaneous light intensity during the day was 57.7 ± 3.4 $\mu mol \cdot m^{-2} \cdot s^{-1}$.

Fresh weight of flower stems was measured at harvest, 1 day after harvest, and at the end of vase life. Flower stems were dried in an oven at 55°C for at least 48 hours before measuring dry weight. Water uptake was measured 1 day after harvest and vase water was changed at this time. A jar containing only deionized water was used to quantify evaporation during the first

day after harvest; the volume of water evaporated from this jar was subtracted from the uptake for all stems. Vase life was determined visually once daily and was terminated based on the same criteria as in the nitrogen study (Figure 2.3).

Statistical Analysis

Data were analyzed using SAS JMP Pro 11 (SAS Institute, Cary, NC). Analysis of variance (ANOVA) with a significance level of 0.05 was used to compare means. Data for the nitrogen study were analyzed as a split plot experiment with four blocks; nitrogen rate was the whole plot factor and cultivar was the split plot factor. Linear and quadratic regressions were used to analyze the effect of increasing nitrogen rates. Data for the irrigation study were analyzed as a randomized complete block design with subsampling; plants receiving the same irrigation treatment within a single block were treated as subsamples.

Results and Discussion

Fertilization Experiment

At transplant, plants assigned to different nitrogen rates were similar in number of branches ($P = 0.66$) and plant width ($P = 0.23$). The number of branches at transplant was also similar for plants of all cultivars ($P = 0.28$). Plant width at transplant differed among cultivars ($P = 0.0013$), with lavender-flowered plants about 1 cm wider than the blue and white plants. When flowers were harvested, plants of different cultivars showed no differences in time to harvest ($P = 0.16$), plant width ($P = 0.36$), number of branches ($P = 0.26$), number of flower stems ($P = 0.77$), stem caliper ($P = 0.61$), or whole plant fresh weight ($P = 0.14$). Nitrogen rate did not change time to harvest ($P = 0.97$), plant width ($P = 0.65$), number of branches ($P = 0.09$),

number of flower stems ($P = 0.56$), stem caliper ($P = 0.45$), or whole plant fresh weight ($P = 0.41$). The fresh weight of cut stems of each cultivar were similar at harvest ($P = 0.85$) and at the end of vase life ($P = 0.67$). Dry weight of flower stems of all cultivars was the same ($P = 0.61$). Nitrogen rate did not change the fresh weight of flower stems at harvest ($P = 0.23$), fresh weight at the end of vase life ($P = 0.11$), or dry weight ($P = 0.32$).

Nitrogen rate did not significantly impact postharvest parameters of delphinium 'Guardian Mix' (Table 2.1). The end of vase life for all delphinium in this study was due to petal abscission and petal wilting (Figure 2.3). Mean vase life of cut delphinium flowers was 6 days and did not differ with nitrogen rate ($P = 0.09$). Vase life of delphinium 'Guardian Mix' fertilized at 350 mg·L^{-1} N was 0.3 days shorter than flowers receiving lower N rates. Previous research indicates that flower longevity correlates with N rates for other cut flower taxa. The longevity of *Ranunculus* cut flowers and flowers of potted chrysanthemum (*Chrysanthemum ×morifolium* Ramat.) were negatively related to N rate (Bernstein et al., 2005; Roude et al., 1991). Our results suggest a similar response to high N rates in delphinium. However, a 0.3 day reduction in delphinium vase life is unlikely to impact the value of these flowers. Nitrogen rate did not change the postharvest water uptake or height of delphinium plants measured at transplant and harvest (Table 2.1).

Because nitrogen is a mobile plant nutrient, delphinium plants receiving low rates of nitrogen utilized nitrogen stored in older leaves to facilitate growth and development of younger leaves. Chlorophyll index measurements of delphinium reflect this nitrogen mobilization. Chlorophyll index as measured with a SPAD meter have been correlated with the N content of plant tissues (Schuch et al., 1995). The chlorophyll index of older leaves of delphinium at harvest was significantly affected by N rate (Table 2.1, Figure 2.5). Older leaves of plants

receiving 150 mg·L^{-1} N or less had a chlorophyll index that was 14% less than the chlorophyll index measured in plants at the two highest N rates (P = 0.0005). Young leaves showed the opposite pattern at harvest: chlorophyll index of young leaves decreased with increasing N rates (P = 0.051). The young leaves of plants at the lowest N rate had a chlorophyll index 5% greater than young leaves of plants receiving other N rates. In general, plants fertilized at lower nitrogen rates showed the largest difference in chlorophyll index between the old and young leaves (Figure 2.5). Plants receiving 50 mg·L^{-1} N exhibited a 43% increase in chlorophyll index from old to young leaves; this difference was reduced to 19% at 250 and 350 mg·L^{-1} N. This suggests that delphinium receiving low N rates mobilized more nitrogen from older leaves to younger leaves. When used as cut flowers, the older leaves of delphinium are removed, so the measured differences in chlorophyll index would not impact marketability of these flowers.

Young leaves of delphinium showed slight differences in chlorophyll index measured at harvest (P = 0.051), with delphinium receiving the lowest N rate having a chlorophyll index 5% greater than other cut delphinium flowers (Figure 2.5). By 4 days after harvest, chlorophyll index of young leaves differed significantly between plants receiving different N rates (Table 2.1). The chlorophyll index of young leaves of plants receiving 50 or 150 mg·L^{-1} N was 5% greater than that of plants receiving higher N rates (P = 0.02). However, differences in leaf color were not detected during visual assessments of vase life and thus were not considered detrimental to market value of delphinium ‘Guardian Mix’ cut flowers. Previous research on herbaceous landscape plants suggests that acceptable aesthetic quality can be reached before growth parameters, such as chlorophyll index, are optimized (Shurberg et al., 2012a, 2012b).

Mean nitrogen content of delphinium ‘Guardian Mix’ tissues was 4.09 ± 0.16%. Tissue nitrogen levels were not different for plants receiving different N rates (P = 0.12) or plants of

different cultivars ($P = 0.19$). Tissue N content increased with N rate for the lavender-flowered cultivar only; tissue nitrogen content of the blue- and white-flowered cultivars did not consistently increase with nitrogen rate (data not reported). Nitrogen content of delphinium 'Guardian Mix' in this experiment fell above the recommended range of 2.0% to 3.2% for delphinium Pacific hybrids (Bryson et al., 2014). This suggests that fertilization at 50 $mg \cdot L^{-1}$ N is sufficient for growth and flowering of delphinium 'Guardian Mix.' In contrast with these results, Marchese et al. (2005) observed increasing chlorophyll index and nitrogen content in leaves of lisianthus receiving increasing rates of nitrogen.

As expected, increasing N rate increased the media electrical conductivity (EC) beginning 2 WAT and continuing throughout the experiment (Figure 2.6). At 2 WAT, plants fertilized at the lowest N rate had a substrate EC of 1.72 $\pm$ 0.09 $mS \cdot cm^{-1}$, while EC for plants receiving the highest N rate was 2.81 $\pm$ 0.10 $mS \cdot cm^{-1}$. Note that the substrate contained a starter nutrient charge. In addition, all pots were leached after measuring EC and pH at 2 WAT and fertilizer formulation was altered thereafter, hence the non-linear increases in EC over time. Distinct differences in EC for each N rate were especially evident when flower harvests began 6 WAT. By 6 WAT, EC of plants receiving 150 $mg \cdot L^{-1}$ N or higher had reached over 3 $mS \cdot cm^{-1}$, exceeding the highest recommended EC range for greenhouse crops (Cavins et al., 2000). High media EC did not detract from the vase life or quality of delphinium 'Guardian Mix' cut flowers; all stems in postharvest evaluation were considered marketable. Grieve (2011) found that a variety of taxa produce marketable cut flowers when irrigated with high salinity wastewater. It should be noted that the number of branches was slightly decreased in delphinium 'Guardian Mix' plants receiving the highest N rate ($P = 0.09$). Branching increased with N rate up to 250 $mg \cdot L^{-1}$ N, with plants at this rate having 2.3 $\pm$ 0.3 branches. However, branches were reduced to

1.5 ± 0.2 branches in plants receiving 350 mg·L^{-1} N. This suggests that high substrate EC at 350 mg·L^{-1} N may inhibit plant growth and could have a detrimental effect on the production of secondary flower stems in delphinium 'Guardian Mix.'

There were differences in growth and postharvest quality of the three cultivars in the 'Guardian Mix' (Table 2.2). Plants of different cultivars had different width at transplant, heights at both transplant and harvest, number of florets, vase life, and postharvest water uptake. Cultivars were distinguished by flower color and were unknown until time of flowering. However, initial size of the plants was different for the cultivars. The width of plants with white and blue flowers was about 1 cm less than the width of plants with lavender flowers. In addition, plants with white flowers were significantly shorter at the time of transplant, a difference that may have been reflected in the slightly longer time to harvest for this cultivar; on average, white flowers were harvested about 4 days later than blue and lavender flowers ($P = 0.16$). At harvest, plants with white flowers were over 10 cm taller and had up to 7 more florets than other cultivars. Postharvest longevity of white flowers was 1 day shorter and water uptake was lower than that of other cultivars. Similarly, a study of anthurium (*Anthurium andraeanum* Hort.) showed wide variations in vase life and water uptake between cultivars (Elibox and Umaharan, 2010).

Vase life of delphinium 'Guardian Mix' in this experiment was correlated with water uptake ($r = 0.57$); flower stems with higher uptake tended to have a longer vase life (Figure 2.7). Similarly, *Anthurium* flowers that maintained high levels of water uptake after harvest exhibited extended vase life (Elibox and Umaharan, 2010). Optimizing water relations of delphinium 'Guardian Mix' during production and after harvest may be important for extending flower vase life.

Delphinium 'Guardian Mix' plants produced marketable, high quality flowers at nitrogen rates as low as 50 $mg \cdot L^{-1}$. For cut flower production, the youngest leaves are of main concern for quality of the cut stems. Chlorophyll index of leaves, as measured with a SPAD meter, indicated that plants receiving lower N rates mobilized N from older leaves to younger leaves. This mobilization of nitrogen allowed plants at all N rates to produce marketable cut stems. Cultivars in the 'Guardian Mix' exhibited differences in plant and flower size, vase life, and water uptake. Water uptake for all stems was correlated with vase life, suggesting that water relations may be the key to extending vase life of delphinium 'Guardian Mix.'

Deficit Irrigation Experiment

Substrate of plants receiving water deficit treatments was allowed to dry more than controls (Figure 2.8). Plants under water deficit required less frequent irrigation. In the first repetition of this experiment with harvests in January, control plants received irrigated on 9 dates, while moderate and severe water deficit plants were irrigated on 5 and 4 dates, respectively. In the second experiment that was harvested in February, irrigation occurred on 8, 6, and 4 dates for the control, moderate deficit, and severe deficit treatments, respectively. Observed mean of daily VWC measurements for all plants in the control, moderate deficit, and severe deficit treatments were 20.0 ± 0.4%, 18.2 ± 0.4%, and 14.5 ± 0.5%, respectively.

Water deficits did not alter vase life, water uptake, or fresh or dry weights of delphinium 'Guardian Blue' flowers (Table 2.3). Fresh weights of the flowers at harvest seemed to reflect the water deficit with stems grown under higher levels of water stress weighing less. However, the trends observed in stem fresh weights were not significant at the $P = 0.05$ level ($P = 0.051$ for the January experiment, $P = 0.058$ for the February experiment). One day after harvest, fresh

weight of all cut delphinium increased and the weight of all stems was similar ($P = 0.34$ in the January experiment, $P = 0.23$ in the February experiment). Similar to the results seen for delphinium 'Guardian Blue,' the postharvest quality and longevity of lisianthus 'Ex Rosa Green' flowers were not altered by drought stress (Chuang and Chang, 2012). However, in the same study, moderate drought stress improved vase life, lowered transpiration and water uptake of lisianthus 'Piccolo Lime Green.'

Stomatal conductance (g_s) and transpiration rates (E) of delphinium 'Guardian Blue' plants grown with water deficits were not reduced when compared to control plants (Figure 2.9). In general, g_s and E showed less variation after drought stress treatments had begun. This was true for all irrigation treatments, suggesting an effect of plant age rather than irrigation treatment. Postharvest g_s and E of cut flowers showed the greatest variation. In previous studies, deficit irrigation led to decreased g_s of carnation (Álvarez et al., 2009) and reduced E in lisianthus 'Piccolo Lime Green' (Chuang and Chang, 2012). Reduced transpiration led to extended vase life in lisianthus (Chuang and Chang, 2012), a relationship that has also been observed in cut roses (Fanourakis et al., 2012; Mayak et al., 1974). In et al. (2016) found that stomatal function in cut roses was correlated with preharvest environmental conditions, including relative humidity, temperature, and vapor pressure deficit. In our study, substrate VWC of delphinium 'Guardian Blue' was the only environmental factor that was manipulated. It is possible that the greenhouse environmental factors such as temperature, relative humidity, and vapor pressure deficit, allowed similar stomatal development on leaves of plants in all irrigation treatments.

In this study, drought stress was applied after flower buds had initiated. After flower buds were visible, delphinium 'Guardian Blue' plants developed few new leaves. Stomatal size and density are determined by both genetic and environmental factors at the time of stomatal

development (Hetherington and Woodward, 2003). Applying drought stress earlier in delphinium production may have provided more time for stomata to develop differences between treatments. However, genetics may play a larger role in determining E and g_s of delphinium 'Guardian Blue.' In roses, seasonal variation occurs in stomatal size, transpiration rates, and vase life, with spring-grown roses showing the lowest transpiration rates and longest vase life (In et al., 2016). Environments with lower relative humidity typically produce roses with lower E and longer vase life (Fanourakis et al., 2012; In et al, 2016). Altering the water content in the media may not have the same effect on transpiration as the relative humidity of the greenhouse environment.

Conclusions

The results of these studies suggest that high quality delphinium cut flowers can be produced at low nitrogen rates and under drought stress. Delphinium 'Guardian Mix' flowers produced marketable cut flowers when fertilized at rates as low as 50 $mg \cdot L^{-1}$ N. In addition, high substrate EC was not detrimental to delphinium 'Guardian Mix' flower quality. Delphinium 'Guardian Blue' plants produced marketable cut flowers under both moderate and severe irrigation deficits. Stomatal conductance, transpiration rate, and vase life of 'Guardian Blue' cut flowers were not altered by deficit irrigation.

These experiments investigated the first cut flower collected from a delphinium plant. However, delphinium plants will produce additional flower stems after the first cut and growers can harvest two to three times in a single season. Delphinium can also be grown as a perennial, with cut stems harvested from a single plant over multiple seasons. Leaf yellowing may not be an issue with the first cut, but may become an issue with successive harvests. Supplying

adequate nitrogen and irrigation may be important in establishing healthy, productive plants. Further research should investigate the effects of nitrogen fertilization and deficit irrigation on successive cuts from delphinium 'Guardian Mix' plants.

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Tables and Figures

Table 2.1. Analysis of variance of fertilizer rate and cultivar (distinguished by flower color) on vase life, water uptake, plant height (including flower stems if present) and chlorophyll index (as measured with a SPAD meter) for Delphinium 'Guardian Mix' cut flowers.

		Mean square						
Source	df	Vase life	Water uptake	Plant height at transplant	Plant height at harvest	SPAD of old leaves at harvest	SPAD of young leaves at harvest	SPAD of young leaves 4 DAH
Nitrogen rate	3	1.42^{NS}	79^{NS}	5.5^{NS}	109^{NS}	79***	36.9^{NS}	79.1*
Cultivar	2	4.84***	9422***	11.6***	719**	141***	61.4^{NS}	8.2^{NS}
N rate × cultivar	6	1.29*	562^{NS}	1.5^{NS}	108^{NS}	12^{NS}	28.0^{NS}	49.1^{NS}

^{NS},*,**,*** Nonsignificant or significant at $P \leq 0.05, 0.01$, or 0.001, respectively.

Table 2.2. Plant width at transplant, plant height (including flower stem if present), number of florets, vase life, and water uptake of Delphinium 'Guardian Mix' flowers. 'Guardian Mix' contains three cultivars distinguished by flower color. Plant width is the average of one measurement taken at the widest part of the plant and a second measurement perpendicular to this. Plant width, height, number of florets, vase life, and water uptake were not significantly affected by nitrogen rate.

Cultivar flower color	Plant width at transplant (cm)	Plant height at transplant (cm)	Plant height at harvest (cm)	Number of florets	Vase life (days)	Water uptake (mL)
Blue[z]	5.61a[w]	5.5a	57.5b	11.5a	6.3a	146a
Lavender[y]	6.37b	5.6a	63.1a	10.1a	6.2a	133a
White[x]	4.75a	4.0b	69.3a	17.1b	5.5b	106b
P value	0.0013	0.0005	0.0011	<0.0001	0.0004	<0.0001

[z]n=33 for all columns except uptake n=30.

[y]n=15

[x]n=37 for all columns except uptake n=35.

[w]Means within a column followed by the same letter are not significantly different (student's t test, $P < 0.05$).

Table 2.3. Postharvest parameters of delphinium 'Guardian Blue' cut flowers produced under normal irrigation (control), or moderate or severe water deficit beginning at flower bud initiation. The experiment was conducted twice, with harvests beginning in January and February for the first and second experiments, respectively. Each value represents the mean of measurements from at least 5 cut flowers receiving the specified irrigation treatment.

Water deficit	Vase life (days)	Water uptake 1 day after harvest (mL)	Fresh weight at harvest (g)	Fresh weight 1 day after harvest (g)	Fresh weight at termination (g)	Dry weight (g)
January experiment						
Control	8.1	13.0	19.3	21.0	15.2	2.8
Moderate	7.2	20.0	17.2	19.4	15.4	2.5
Severe	8.7	17.8	14.6	18.2	15.3	2.6
P value	0.557	0.328	0.0506	0.335	0.979	0.500
February experiment						
Control	9.3	8.6	16.9	19.7	14.4	2.4
Moderate	8.7	0.8	17.1	19.8	14.4	2.4
Severe	9.8	8.0	13.5	16.4	11.7	2.1
P value	0.446	0.462	0.0581	0.2289	0.483	0.4684

Figure 2.1. Diagram of experimental design for delphinium 'Guardian Blue' fertilization experiment. Blocks were included to control for variation in the greenhouse environment. Blocks were divided into 4 whole plots; for ease of application, nitrogen rate applied to whole plots by constant liquid feed at four levels: 50, 150, 250, or 350 mg·L^{-1}. Each whole plot contained 6 split plots containing a single plant; cultivar (as distinguished by flower color) was applied to split plots. Postharvest data were collected for 4 to 6 split plots within each whole plot.

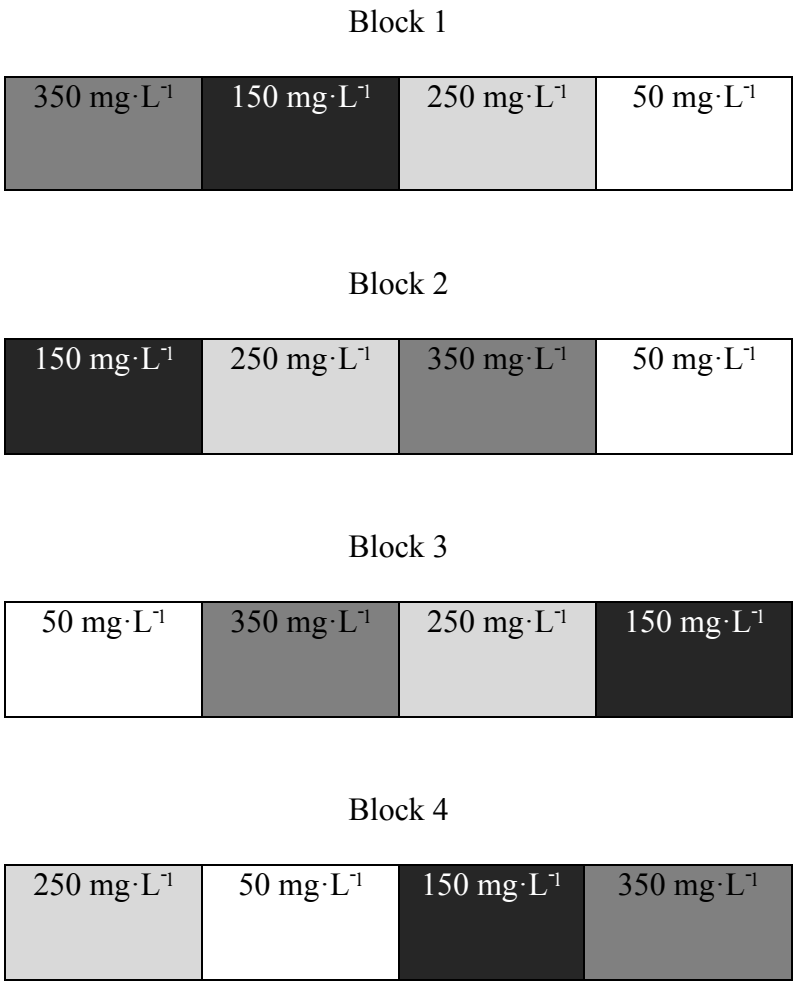


Figure 2.2. Minimum (left) and maximum (right) harvest stages for delphinium ‘Guardian Blue’ cut flowers. Flowers were harvested when at least one floret had opened so that the throat of the floret was visible, but before half of the florets were open.

Figure 2.3. Petal abscission (left) and petal wilt (right) in delphinium ‘Guardian Blue’ cut flowers. Vase life was terminated when at least 50% of the florets exhibited petal abscission or petal wilt.

Figure 2.4. Experimental design for delphinium ‘Guardian Blue’ deficit irrigation experiment. The experiment was divided into two blocks to control for greenhouse environmental variation. Within each block, irrigation treatment was applied at three levels: control (sufficient irrigation), moderate or severe water deficit. Irrigation treatments were applied by subirrigation to six plants (indicated by black circles) sitting in a single tray; each plant was treated as a subsample.

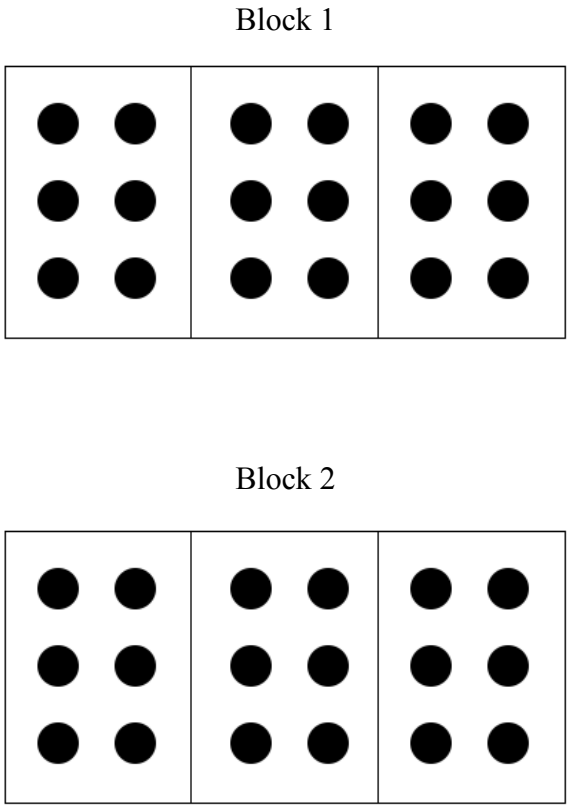


Figure 2.5. Response of leaf chlorophyll index of delphinium 'Guardian Mix' cut flowers to nitrogen rate. Chlorophyll index was measured with a SPAD-502 (Spectrum Technologies, Aurora, IL) at flower harvest (top) and 4 days after harvest (bottom) for the youngest fully expanded leaf and an old leaf located 5 cm for the base of the plant for plants receiving nitrogen at 50, 150, 250, or 350 mg·L^{-1}. Each data point represents the mean chlorophyll content measured on at least 20 cut stems, error bars represent 1 standard error from the mean. Regression curves show a quadratic effect on nitrogen rate; $R^2 = 0.06$ for young leaves at harvest, $R^2 = 0.21$ for old leaves at harvest, $R^2 = 0.03$ for young leaves 4 DAH.

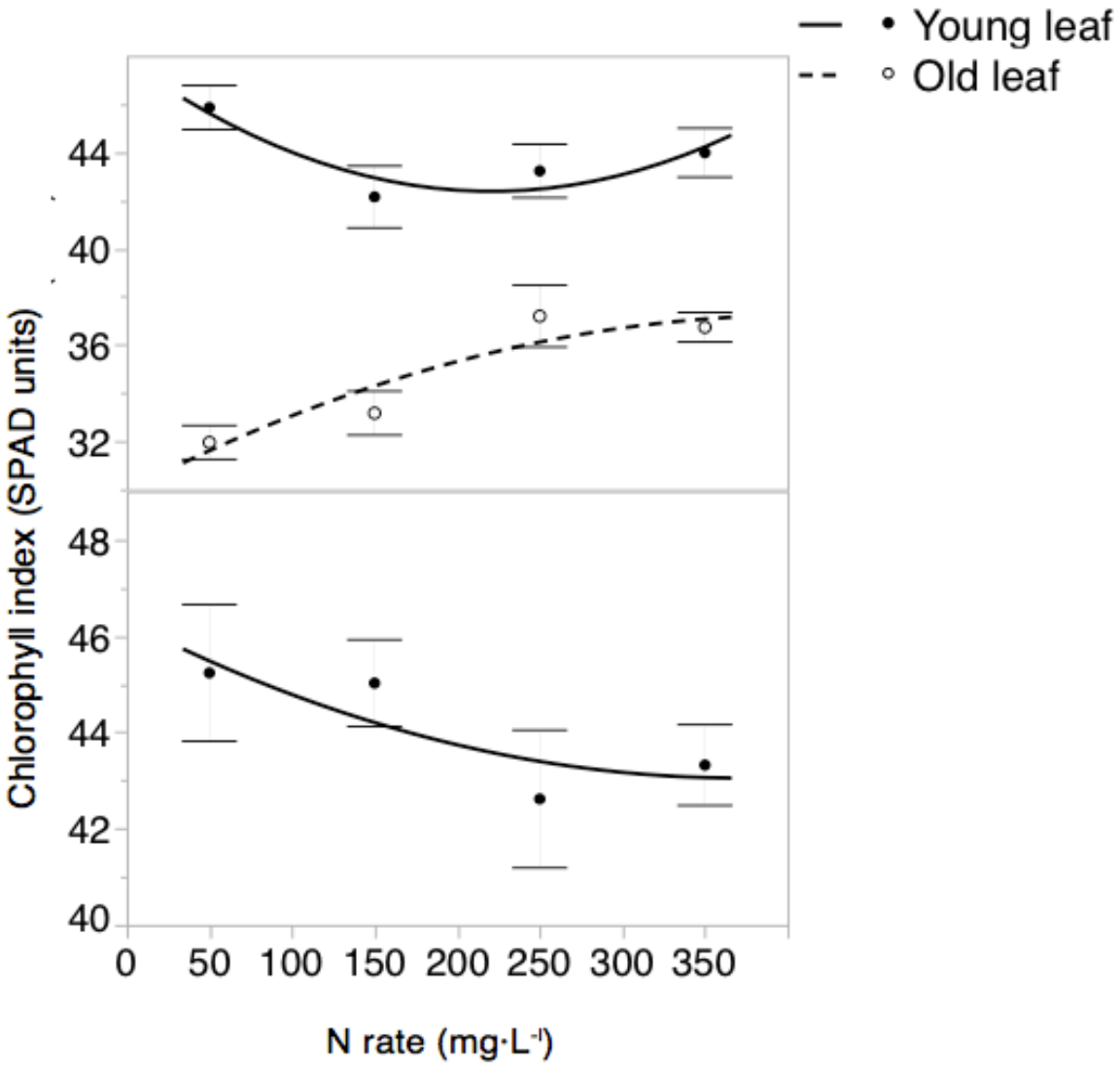


Figure 2.6. Response of media electrical conductivity (EC) for delphinium 'Guardian Mix' to nitrogen fertilization applied via constant liquid feed at four rates: 50, 150, 250, or 350 $mg \cdot L^{-1}$. EC was measured every 2 weeks by the PourThru method. Each point represents the mean substrate EC measured for 4 plants. The point at 0 weeks after treatment represents the mean substrate EC of 4 randomly selected plants from the entire experiment; fertilization had not yet begun at this time. In subsequent weeks, EC was measured for 4 plants at each N rate. Fertilizer rate had a quadratic effect on EC; $R^2 = 0.43$, 0.50, 0.78, 0.80 for N rates 50, 150, 250, and 350 $mg \cdot L^{-1}$, respectively. The arrow indicates time at which all pots were leached with clear water and fertilizer formulation was altered.

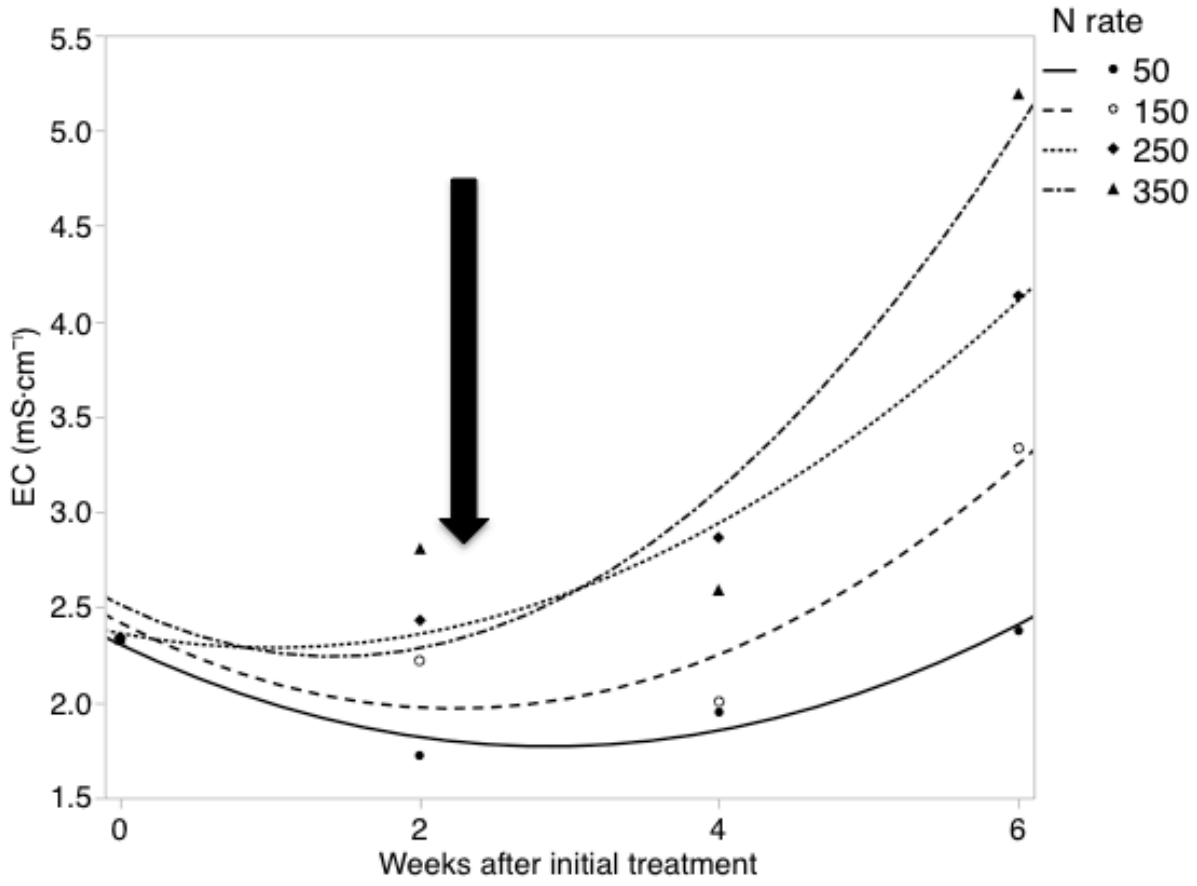


Figure 2.7. Relationship between vase water uptake and vase life for delphinium ‘Guardian Mix’ cut flowers ($r = 0.57$). ‘Guardian Mix’ contains three cultivars distinguished by flower color: blue, lavender, or white.

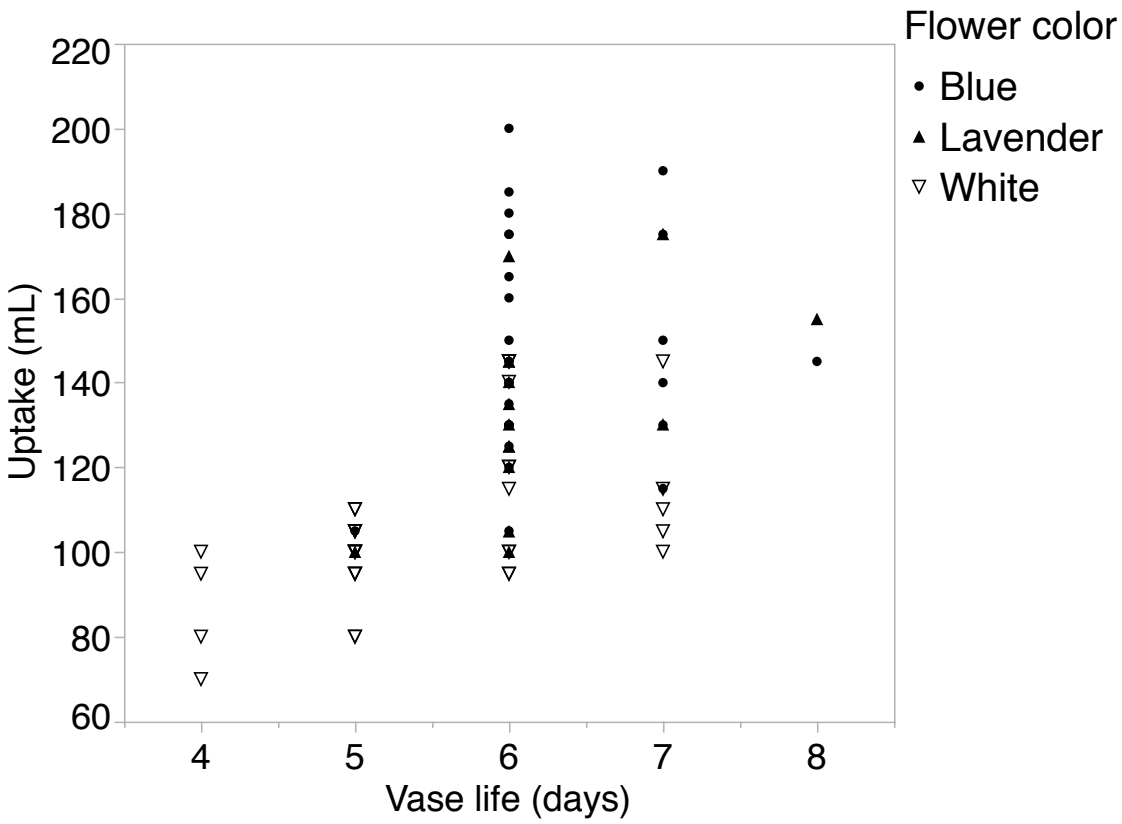


Figure 2.8. Substrate volumetric water content (VWC) of delphinium 'Guardian Blue' plants grown under three irrigation treatments: control (sufficient water), moderate or severe water deficit. VWC was measured daily for 2 plants in each treatment and block (4 plants total) with a Procheck handheld VWC meter (Decagon Devices, Pullman, WA).

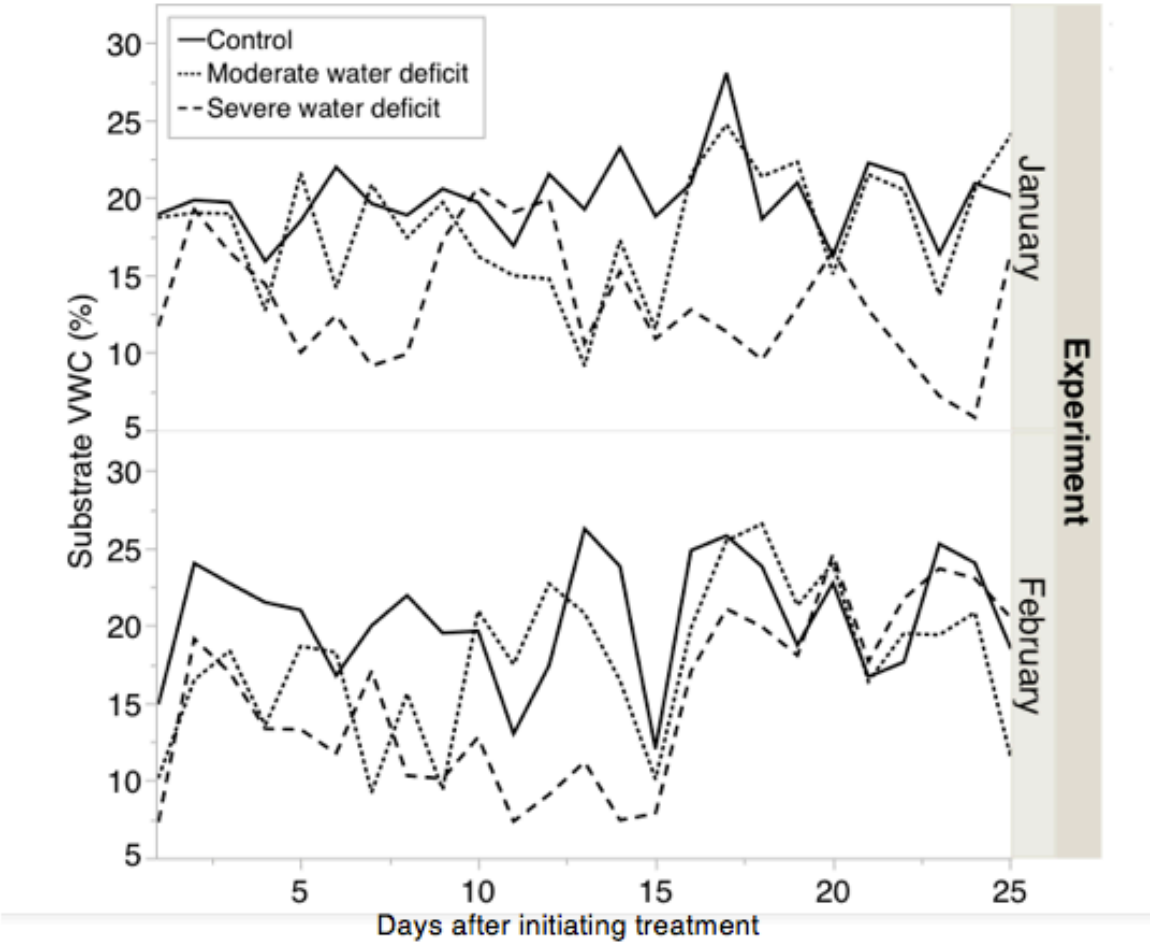
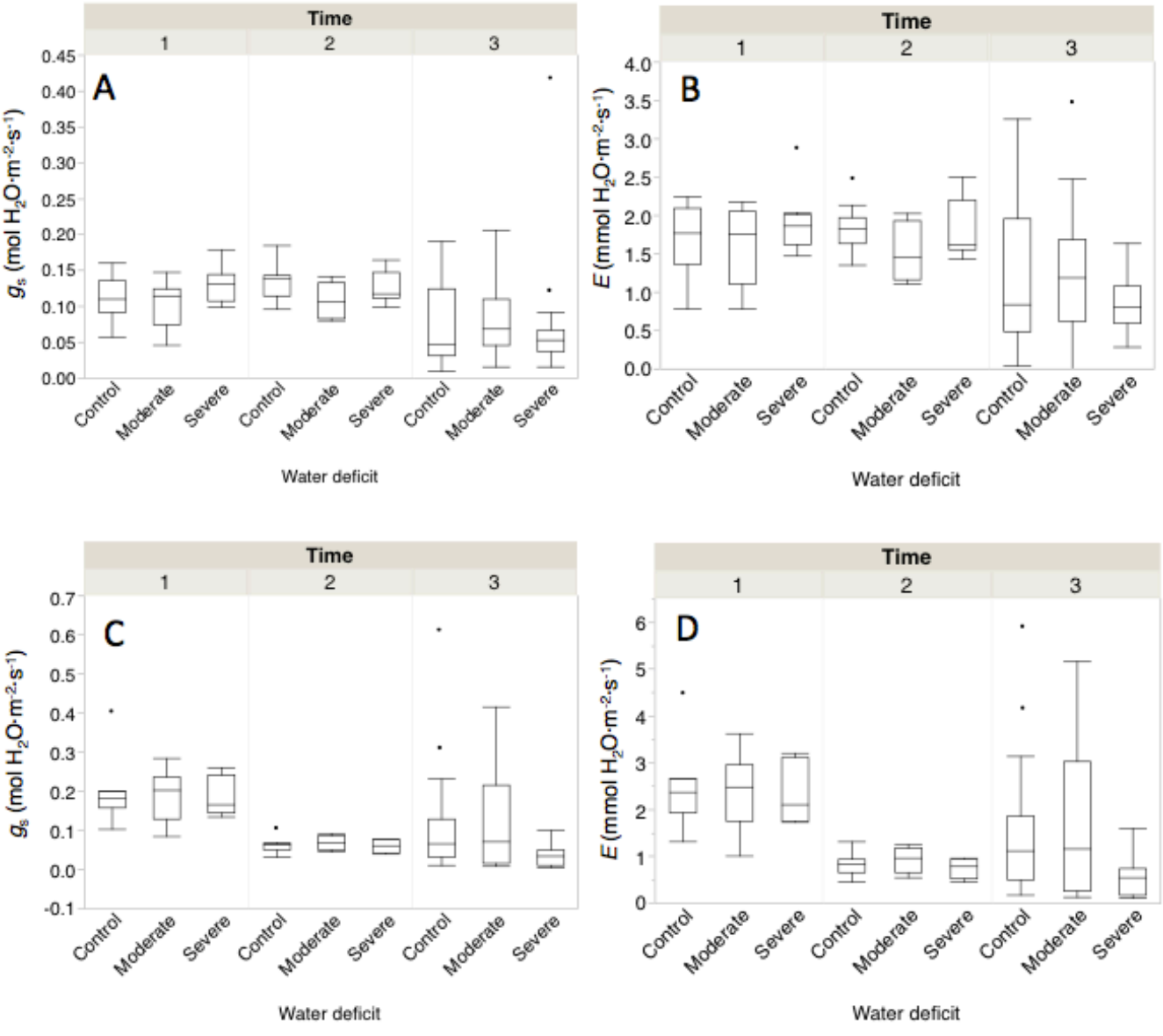


Figure 2.9. Stomatal conductance (g_s) and transpiration rates (E) of delphinium 'Guardian Blue' measured before applying water deficit treatments (time 1), 2 weeks after the start of water deficit treatments (time 2), and 1 day after cut flower harvest (time 3). Stomatal conductance and E were measured with a LI-6400XT (LI-COR, Lincoln, NE). The experiment was conducted twice, with cut flower harvests beginning in January for the first experiment (A, B) and in February for the second experiment (C, D).



Chapter 3: Foliar applications of calcium chloride and postharvest quality of cut sunflower (*Helianthus annuus* L.) 'Moulin Rouge' and 'Procut Lemon'

Abstract

The vase life of cut sunflowers (*Helianthus annuus* L.) can be limited by early abscission of petals. Calcium can strengthen cell walls and may delay senescence in cut flowers. Calcium chloride was applied as a weekly foliar spray at 0 (control), 500, 1000, or 2000 $mg \cdot L^{-1}$ calcium during the production of sunflower 'Moulin Rouge' and 'Procut Lemon.' Increasing rates of calcium sprays corresponded to increases in fresh and dry weight of 'Moulin Rouge' cut stems and increases in tissue calcium content for this cultivar. Fresh and dry weight of 'Procut Lemon' stems was highest for stems receiving the control spray and tissue calcium content did not correspond to the rate of calcium spray for 'Procut Lemon.' For both cultivars, calcium sprays had no impact on pre- or post-harvest transpiration and stomatal conductance. Postharvest transpiration and stomatal conductance were higher for 'Moulin Rouge' cut flowers. 'Moulin Rouge' flowers had shorter postharvest vase life than 'Procut Lemon' cut flowers. Calcium application had no effect on vase life of cut 'Moulin Rouge' or 'Procut Lemon' flowers. Foliar sprays of calcium are not recommended for improving postharvest longevity of 'Moulin Rouge' or 'Procut Lemon' flowers.

Introduction

Calcium is an essential plant nutrient with many functions within the plant. Calcium acts as a stabilizing component of plant cell walls (Hepler, 2005). Calcium may delay postharvest

senescence of flowers and foliage by preserving the integrity of plant cells. Placing cut roses (*Rosa hybrida* L.) in vase solution containing calcium chloride ($CaCl_2$) extended postharvest longevity of the flowers and petals (Torre et al., 1999). Preharvest foliar sprays of Ca strengthened stems of herbaceous peony (*Paeonia lactiflora* Pall.; Li et al., 2012) and extended vase life of gerbera (*Gerbera jamesonii* Bolus; Gerasopoulos and Chebli, 1999). Calcium sprays also extended the postharvest shelf life of broccoli (*Brassica oleracea* L. var. *italica*) microgreens, including higher visual quality and decreased levels of postharvest microbial growth on the microgreens (Kou et al., 2014).

Calcium ions also play an important role in regulating the opening and closure of stomata (Schwartz, 1985). Direct applications of $CaCl_2$ to leaves have been shown to induce rapid stomatal closure, resulting in decreased transpiration rate (E) and stomatal conductance (g_s; Atkinson et al., 1990; Schwartz, 1985). Cut roses with high E exhibit shorter vase life (Mayak et al., 1974). Thus, foliar Ca applications that induce stomatal closure may be able to extend cut flower vase life by limiting transpiration.

Sunflowers (*Helianthus annuus* L.) grown for the cut flower market can have issues with petal abscission after harvest (Tata and Wien, 2014). Vase life varies between sunflower cultivars; red-flowered cultivars show a high tendency to lose petals and have shorter vase life than cultivars with yellow or orange flowers (Tata, 2013). Preharvest Ca applications by spray or drench at rates up to 500 $mg \cdot L^{-1}$ Ca to sunflower 'Superior Sunset' increased tissue Ca content but did not extend vase life, while postharvest Ca applications applied in the vase solution did prolong vase life of 'Superior Sunset' flowers (Nan, 2007). Supplying sunflowers with Ca at higher rates or more frequently during production may help flowers hold their petals and extend vase life.

Market research indicates that short cut flower longevity results in consumer dissatisfaction and that consumers are willing to pay more for flowers with longevity guarantees (Rihn et al., 2011, 2014). Extending sunflower vase life could increase their value. The objective of this experiment was to determine the postharvest impact of foliar Ca sprays applied at different rates during production of sunflower 'Moulin Rouge' or 'Procut Lemon' cut flowers. Specifically, this experiment investigated the effect of Ca on sunflower vase life, stomatal conductance, and transpiration rates.

Materials and Methods

Plant materials

Helianthus annuus 'Moulin Rouge' and 'Procut Lemon' seeds (Johnny's Selected Seeds, Winslow, ME) were planted directly in trade gallon pots (2.8 L) containing soilless substrate (Fafard 52 mix; Sun Gro Horticulture, Agawam, MA). Plants were grown from June to August 2015 in a glass greenhouse in Blacksburg, VA. Mean temperature was 24.1 ± 0.2°C during the day and 22.6 ± 0.1 at night, mean relative humidity was 74.0 ± 0.2%, and mean daily light integral was 19.5 ± 0.8 $mol \cdot m^{-2} \cdot d^{-1}$. Flower stems were supported using plastic mesh. Plants received 150 $mg \cdot L^{-1}$ N of 15N-2.2P-12.5K-4Ca-2Mg (Jack's Professional LX; J.R. Peters Inc., Allentown, PA) via Constant Liquid Feed (CLF) when soil volumetric water content (VWC) fell to 18% or below, as measured with a Procheck handheld VWC meter (Decagon Devices, Pullman, WA). Electrical conductivity (EC) and pH of the substrate were measured every 2 weeks using the PourThru method. Due to low EC values (0.56 ± 0.09 $mS \cdot cm^{-2}$) CLF was increased to 200 $mg \cdot L^{-1}$ N from July 30 to August 7.

Calcium application

Plants were arranged in a strip plot design to allow for easy application of Ca sprays to each cultivar (Figure 3.1). Beginning 3 weeks after planting, plants received a weekly foliar spray of calcium chloride dihydrate (Fisher Scientific, Pittsburgh, PA) at one of four rates: 0 (control), 500, 1000, or 2000 $mg \cdot L^{-1}$ Ca. At the time of the first Ca application, 'Moulin Rouge' plants had 8 to 10 leaves and 'Procut Lemon' plants had 6 to 8 leaves. Sprays continued once each week until both cultivars had swollen flower buds (6 weeks after seeding). Spray was applied to runoff with spray volume adjusted for plant size each week (from 450 $mL \cdot m^{-2}$ to 830 $mL \cdot m^{-2}$).

Postharvest data collection

Sunflowers were harvested in the morning when bracts had lifted so that when looking directly at the flower head, at least 60% of the disc was visible, colored petals, but before all petals were completely expanded (Figure 3.2). All stems were recut to a length of 50 cm. Leaves below the uppermost three leaves and axillary flower buds more than 30 cm below the main flower head were removed from the stems. For three plants in each subplot, leaves four through nine were dried for nutritional analysis. Leaves for nutritional analysis were dried in an oven at 55°C for at least 48 hours, ground to a powder, and sent to QAL (Panama City, FL) for analysis of tissue calcium content. After recutting stems, diameter of the flower head and fresh weight of the flower stem were measured. Flowers were placed in tap water for transport to the postharvest laboratory.

The postharvest laboratory was maintained to simulate a typical consumer's home. Average temperature in the postharvest laboratory was 22.2 ± 0.0°C and average relative humidity was 48.9 ± 0.7%. In the postharvest lab, plants received ambient daylight through windows and supplemental light from fluorescent bulbs for ~8 $h \cdot d^{-1}$. Average instantaneous light intensity during the day was 127 ± 77 $\mu mol \cdot m^{-2} \cdot s^{-1}$. Stems were held individually in vases containing 600 mL of deionized (DI) water.

Water uptake for all stems was measured 4 days after harvest (DAH) when vase water was changed. A vase containing only DI water was used to quantify evaporation that occurred over these 4 days and the volume of evaporated water was subtracted from uptake for each stem. Flower head diameter was measured at 5 DAH to determine any changes occurring after harvest. Vase life was visually assessed once each day. Vase life was terminated when at least 50% of the petals had wilted or abscised, or when at least two undesirable qualities were observed (Figure 3.3, 3.4). At the end of vase life, fresh weight of the flower stem and reasons for terminating vase life were recorded. Flower stems were dried in an oven at 55°C for at least 48 hours before measuring dry weight.

Measurement of gas exchange

Transpiration rate (E) and stomatal conductance (g_s) were measured with a LI-6400XT (LI-COR, Lincoln, NE). Measurements of the youngest fully expanded leaf were made using the 6400-40 LCF chamber. One day after the final Ca spray was applied and before any flowers had been harvested, E and g_s were measured on three randomly selected plants in each subplot. One day after each flower harvest, E and g_s were measured again for two stems from each subplot. Environmental conditions during these measurements are listed in Appendix B.

Statistical analysis

Data were analyzed using SAS JMP Pro 11 (SAS Institute, Cary, NC). Means for postharvest data were compared using strip plot analysis of variance (ANOVA) with a significance level of 0.05. The effects of increasing Ca rates were analyzed with linear and quadratic regressions.

Results and Discussion

Calcium application did not affect vase life or postharvest quality of sunflower 'Moulin Rouge' and 'Procut Lemon' (Table 3.1). Nan (2007) observed no extension of vase life in sunflower 'Superior Sunset' receiving sprays of Ca at rates up to 500 $mg \cdot L^{-1}$ Ca. Similarly, vase life of 'Moulin Rouge' and 'Procut Lemon' flowers did not benefit from foliar Ca applications. Calcium applied at rates up to 2000 $mg \cdot L^{-1}$ Ca had no impact on vase life of 'Moulin Rouge' or 'Procut Lemon.'

Vase water uptake was not affected by cultivar ($P = 0.15$) or Ca application rate ($P = 0.42$). Reasons for ending vase life of sunflowers varied (Table 3.2). For both cultivars, the most commonly observed reason for ending vase life was wilting of petals. Petal abscission, or loss of petals, affected 24% of 'Moulin Rouge' and 3% of 'Procut Lemon' flowers in this study. Previous research has indicated that petal abscission is an issue that can limit the vase life of cut sunflowers, especially red sunflowers such as 'Moulin Rouge' (Tata, 2013). Tata (2013) harvested flowers after they had completely opened. In our study, as in some other studies of sunflowers (Clark et al., 2010), sunflowers were harvested before petals had opened completely.

When harvested at this earlier flower stage, petal wilting is a more prevalent problem than petal abscission.

Observed vase life of 'Moulin Rouge' flowers was 9.5 days, while 'Procut Lemon' flowers lasted 12 days (Table 3.3). Previous research indicates that vase life varies between sunflower cultivars, with yellow-flowered cultivars such as 'Procut Lemon' outlasting cultivars with red flowers such as 'Moulin Rouge' (Tata, 2013). Market research reveals that consumers are willing to pay a premium for flowers with longevity guarantees (Rihn et al., 2014). Thus, longer lasting 'Procut Lemon' flowers may be more valuable to cut flower growers than 'Moulin Rouge' flowers. However, survey results showed that consumers and florists prefer the appearance of some red sunflowers, including 'Moulin Rouge' (Ferguson et al., 2012). When choosing sunflower cultivars, cut flower growers must consider the needs of their specific market as well as flower longevity.

In addition to differences in postharvest vase life, the cultivars also differed in their flower size and opening of the flowers after harvest (Table 3.1). 'Procut Lemon' flowers had a larger diameter at the time of harvest ($P = 0.09$), but 'Moulin Rouge' flowers opened more after being harvested ($P = 0.04$, Table 3.3). Visual quality of both cultivars was acceptable (Figure 3.5), so differences in flower diameter are unlikely to affect the value of these sunflower cultivars. Growers who want to control the flower size of sunflowers can utilize plant spacing to manipulate flower size (Rice, 2015).

The effect of Ca on flower opening after harvest was slight ($P = 0.08$). There was no clear pattern in the diameter change of 'Procut Lemon' flowers with increasing Ca application rate (data not reported). For 'Moulin Rouge,' control flowers receiving 0 $mg \cdot L^{-1}$ Ca opened 2.5 ± 0.5 cm after harvest, while flowers treated with Ca opened 4.1 ± 0.3 cm. Calcium promoted

greater opening of ‘Moulin Rouge’ flowers after harvest. Growers should consider the labor and costs associated with Ca applications before implementing this practice, as flower opening in ‘Moulin Rouge’ was the only observed benefit of foliar Ca applications. Previous research indicates that foliar Ca application can alter the diameter of other cut flowers. Preharvest Ca fertilization at rates between 100 and 300 $mg \cdot L^{-1}$ Ca resulted in increased flower diameter in gerbera (Albino-Garduño et al., 2008). Rose flower response to Ca sprays differs between cultivars; some flowers increased in diameter (Nabigol, 2012) and others decreased in diameter (Mehran et al., 2008) after Ca application. Responses to Ca treatment vary between cut flower taxa, so the impact of Ca should be assessed before being utilized.

Calcium sprays affected the weight of flower stems of each cultivar differently. There was a significant interaction effect of cultivar and Ca rate on both fresh and dry weights of cut sunflowers, so the main effects of cultivar and Ca rate cannot be evaluated for these responses (Table 3.1). Calcium application led to increased weight in ‘Moulin Rouge’ cut stems, but ‘Procut Lemon’ stems treated with Ca weighed less than controls (Figure 3.6). Kou et al. (2014) found that foliar applications of $CaCl_2$ increased biomass of broccoli microgreens. ‘Moulin Rouge’ flowers in this study showed a similar response to foliar Ca sprays, but the weight of ‘Procut Lemon’ flowers did not increase with Ca application rate. The patterns observed in stem weight of each cultivar corresponded with differences in relative Ca content of stems in each treatment group; in general, flower stems with higher Ca contents were heavier than stems of the same cultivar with lower Ca contents.

For ‘Moulin Rouge’ cut stems, increased rates of Ca application led to higher calcium content of plant tissues (Figure 3.6). However, the Ca content of ‘Procut Lemon’ flowers did not reflect the rate of Ca application. This suggests that foliar sprays of Ca may be an effective

means of increasing Ca content of 'Moulin Rouge' but not 'Procut Lemon' plants. In gerbera, foliar sprays of Ca did not lead to increases in plant tissue Ca content when applied throughout production, but did increase Ca content when applied only before anthesis (Deljou and Gholipour, 2014). Preharvest Ca sprays increased tissue calcium content in gerbera (Gerasopoulos and Chebli, 1999), peony (Li et al., 2012), and roses (Nabigol, 2012).

Regardless of Ca treatment, 'Procut Lemon' plant tissues contained more Ca than 'Moulin Rouge' plant tissues. Calcium is important in stabilizing cell walls (Hawkesford et al., 2012) and extended the vase life of cut roses (Capdeville et al., 2004), alstroemeria (Galati et al., 2015), gerbera (Deljou and Gholipour, 2014), and lisianthus (Saeedi et al., 2015). Higher Ca content of 'Procut Lemon' plants may contribute to the longer vase life of this cultivar.

Before harvest, there was no difference in g_s or E for plants of different cultivars (P = 0.50, P = 0.84, respectively) or for plants receiving different rates of Ca (P = 0.59, P = 0.78, respectively). Postharvest g_s and E were not altered by Ca treatment (Table 3.1). In one cultivar of gerbera g_s was unaffected by Ca fertilization, while a second cultivar showed a reduction in g_s when Ca fertilization was applied at a rate of 240 mg·L^{-1} Ca (Albino-Garduño et al., 2008). Applying solutions containing calcium nitrate directly to leaves of *Commelina communis* induced rapid stomatal closure, however theses effects were no longer evident 1 day after the Ca was applied (Atkinson et al., 1990). In the current study, foliar Ca applications stopped at least 2 days before sunflowers were harvested. Any changes to E or g_s due to Ca application may have no longer been apparent when gas exchange was measured after sunflower harvests.

Postharvest E and g_s were different between the cultivars (Table 3.3). 'Moulin Rouge' flowers had increased E and g_s in comparison to 'Procut Lemon' flowers. High E has been linked to shortened vase life in cut roses (Mayak et al., 1974). High E of 'Moulin Rouge' stems

may lead to greater water loss via transpiration and contribute to the shorter vase life of 'Moulin Rouge' in relation to 'Procut Lemon' flower stems. However, the correlations between vase life and E or g_s for stems in this study were weak (Figure 3.7). While water loss via transpiration plays a role in senescence of cut sunflowers, other factors not measured in this study may help determine the length of vase life.

Based on the results of this experiment, foliar Ca applications are not recommended for improving postharvest qualities of 'Moulin Rouge' or 'Procut Lemon' cut sunflowers. Vase life was not improved by Ca application applied as a foliar spray at rates up to 2000 $mg \cdot L^{-1}$ Ca. Vase life of 'Moulin Rouge' flowers was 3 days shorter than vase life of 'Procut Lemon' flowers. Lower Ca content in 'Moulin Rouge' plant tissues, coupled with increased postharvest g_s and E in this cultivar may contribute to the earlier senescence of this cultivar. Other practices that limit postharvest water loss by transpiration may be more effective at extending the vase life of these cultivars.

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Tables and Figures

Table 3.1. Analysis of variance of postharvest parameters for Helianthus annuus 'Moulin Rouge' and 'Procut Lemon' cut flowers receiving foliar calcium sprays during production at 0 (control), 500, 1000, and 2000 $mg \cdot L^{-1}$. Stomatal conductance and instantaneous transpiration rate of the youngest fully expanded leaf were measured with a LI-6400XT (LI-COR, Linclon, NE) 1 day after flowers were harvested.

		Mean square						
Source	df	Vase life	Postharvest diameter change	Fresh weight at harvest	Fresh weight at termination	Dry weight	Stomatal conductance	Instantaneous transpiration rate
Cultivar	1	113*	184*	535 ^{NS}	7224 ^{NS}	60.8 ^{NS}	0.0819***	6.61*
Ca rate	3	1.03 ^{NS}	5.42 ^{NS}	210 ^{NS}	182 ^{NS}	2.88 ^{NS}	0.0383 ^{NS}	3.09 ^{NS}
Cultivar × Ca rate	3	0.407 ^{NS}	4.90 ^{NS}	615*	662*	10.2*	0.0193 ^{NS}	1.48 ^{NS}

^{NS},*,**,*** Nonsignificant or significant at $P \leq 0.05, 0.01$, or 0.001, respectively.

Table 3.2. Reasons for terminating vase life and their observed incidence in 'Procut Lemon' and 'Moulin Rouge' cut flowers.

Reason for termination	% Incidence	
	'Moulin Rouge'[z]	'Procut Lemon'[y]
Wilting of petals or flower head	92	85
Abscission of petals	24	3
Browning of flower and bracts	0	69
Stem breakage	5	0
Closing of flower	0	31
Neck bend	0	67

[z] n=37

[y] n=38

Table 3.3. Postharvest parameters with significant differences between sunflower 'Moulin Rouge' and 'Procut Lemon' cut flowers receiving foliar applications of calcium chloride. Spray applications of 0, 500, 1000, or 2000 $mg \cdot L^{-1}$ Ca did not change vase life, flower diameter, postharvest change in flower diameter, stomatal conductance, or transpiration rate of 'Moulin Rouge' or 'Procut Lemon' flowers. Values represent the mean response measured in at least 33 stems of each cultivar.

Cultivar	Vase life (days)	Flower diameter at harvest (cm)	Postharvest change in flower diameter (cm)	Stomatal conductance (mol $H_2O \cdot m^{-2} \cdot s^{-1}$)	Transpiration rate (mmol $H_2O \cdot m^{-2} \cdot s^{-1}$)
Moulin Rouge	9.5	9.9	3.7	2.31	0.213
Procut Lemon	12.1	11.1	0.6	1.68	0.143
P value	0.0405	0.0961	0.0401	<0.0001	0.0321

Figure 3.1. Diagram of sunflower foliar calcium experimental set up. This was a strip plot experiment with two main plots. Calcium rate was randomly applied at one of four rates: 0 (control), 500, 1000, 2000 mg·L^{-1} to strip plots appearing vertically. Cultivar was randomly applied at two levels: 'Moulin Rouge' (MR) and 'Procut Lemon' (PCL) to strip plots appearing horizontally. Numbers within each subplot indicate individual plants. Six plants were present for subplot; postharvest data was collected for at least three of these six plants.

	2000 mg·L^{-1} Ca			500 mg·L^{-1} Ca			1000 mg·L^{-1} Ca			0 mg·L^{-1} Ca		
MR	1	5	9	13	17	21	25	29	33	37	41	45
	2	6	10	14	18	22	26	30	34	38	42	46
PCL	3	7	11	15	19	23	27	31	35	39	43	47
	4	8	12	16	20	24	28	32	36	40	44	48

	0 mg·L^{-1} Ca			1000 mg·L^{-1} Ca			500 mg·L^{-1} Ca			2000 mg·L^{-1} Ca		
PCL	49	53	57	61	65	69	73	77	81	85	89	93
	50	54	58	62	66	70	74	78	82	86	90	94
MR	51	55	59	63	67	71	75	79	83	87	91	95
	52	56	60	64	68	72	76	80	84	88	92	96

Figure 3.2. Sunflower stage at cut flower harvest. Minimum harvest stage for 'Moulin Rouge' (A) and 'Procut Lemon' (C) flowers occurred when bracts had lifted from the disc so that when looking directly at the flower head, at least 60% of the disc was visible, colored petals. 'Moulin Rouge' flowers opened rapidly, and maximum harvest stage (B) occurred when some, but not all, petals had opened beyond a 90° angle from the disc. For 'Procut Lemon' flowers, maximum harvest stage (D) was when 100% of petals had lifted from the disc, but had not opened beyond a 90° angle from the disc.

Figure 3.3. Reasons observed for terminating vase life of sunflower 'Moulin Rouge.' 'Moulin Rouge' cut stems were terminated when at least 50% of petals exhibited petal wilt (A) or petal abscission (B), or when flower head was at least halfway closed (C). Stem breakage was also observed in two stems (not pictured). Note that some stems exhibited more than one of these qualities.

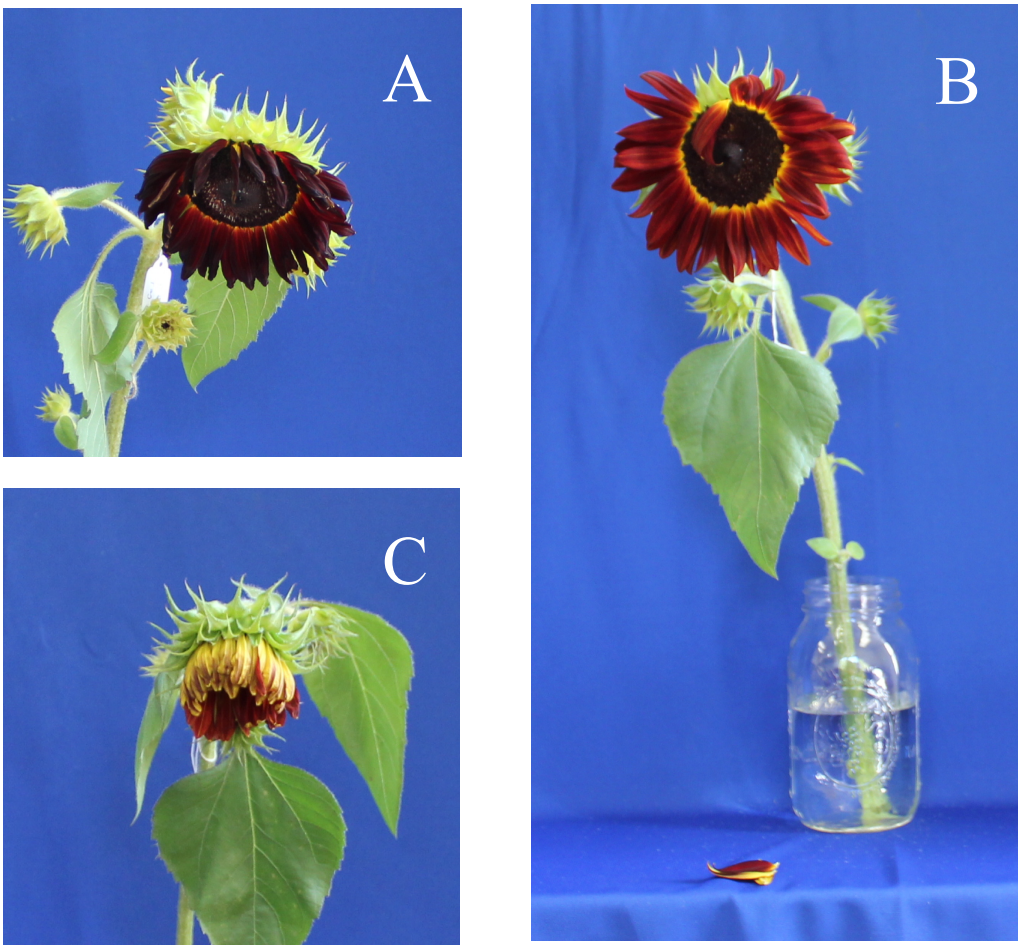


Figure 3.4. Reasons observed for terminating vase life of sunflower 'Procut Lemon.' 'Procut Lemon' vase life was ended when at least 50% of petals exhibited petal wilt (A) or petal abscission (not pictured), or when at least two of the following were present: closing of the flower head (B), neck bend (C), or discoloration of petals and/or bracts (D). Stem breakage was also observed in two 'Procut Lemon' flowers (not pictured). Note that some stems exhibited more than one of these qualities.

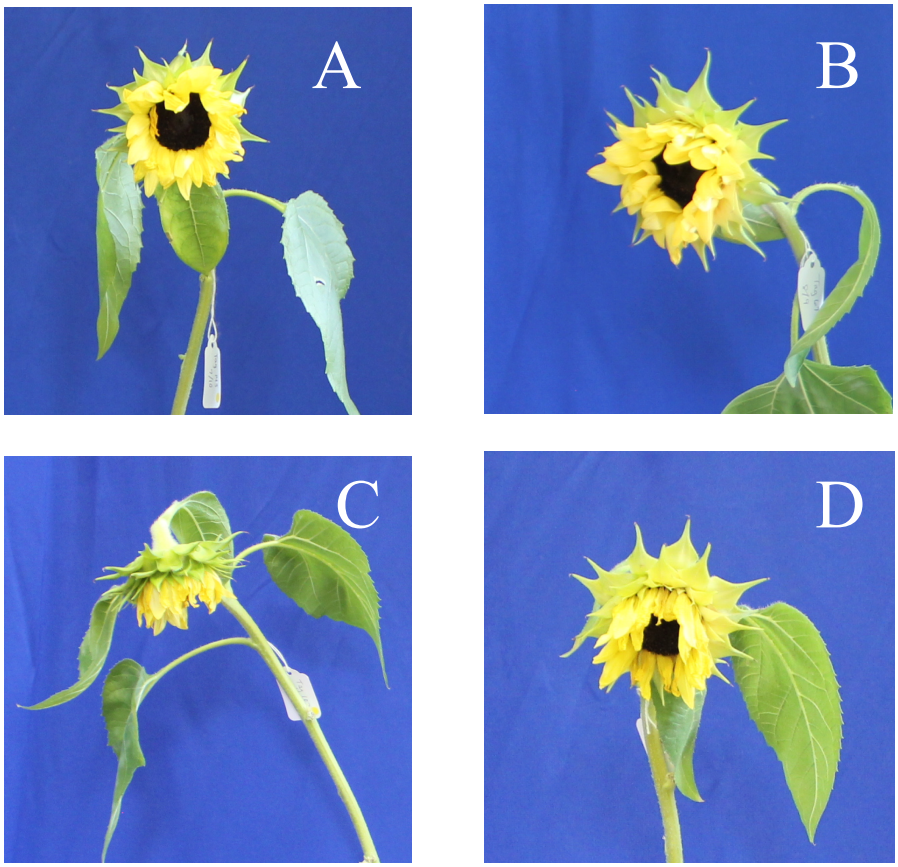


Figure 3.5. Sunflower 'Moulin Rouge' (A) and 'Procut Lemon' (B) flowers pictured 5 days after harvest. Flowers pictured from left to right received foliar sprays of calcium chloride at 0, 500, 1000, and 2000 $mg \cdot L^{-1}$ Ca, respectively.

Figure 3.6. Relationship between calcium application rate and fresh weight at harvest (A), fresh weight at the end of vase life (B), dry weight measured after ending vase life (C), and tissue calcium content (D) for 'Moulin Rouge' (MR) and 'Procut Lemon' (PCL) cut stems receiving calcium foliar sprays at 0, 500, 1000, and 2000 mg·L^{-1}. Tissue calcium content was measured on combined samples of leaves from three plants in each subplot collected at the time of flower harvest.

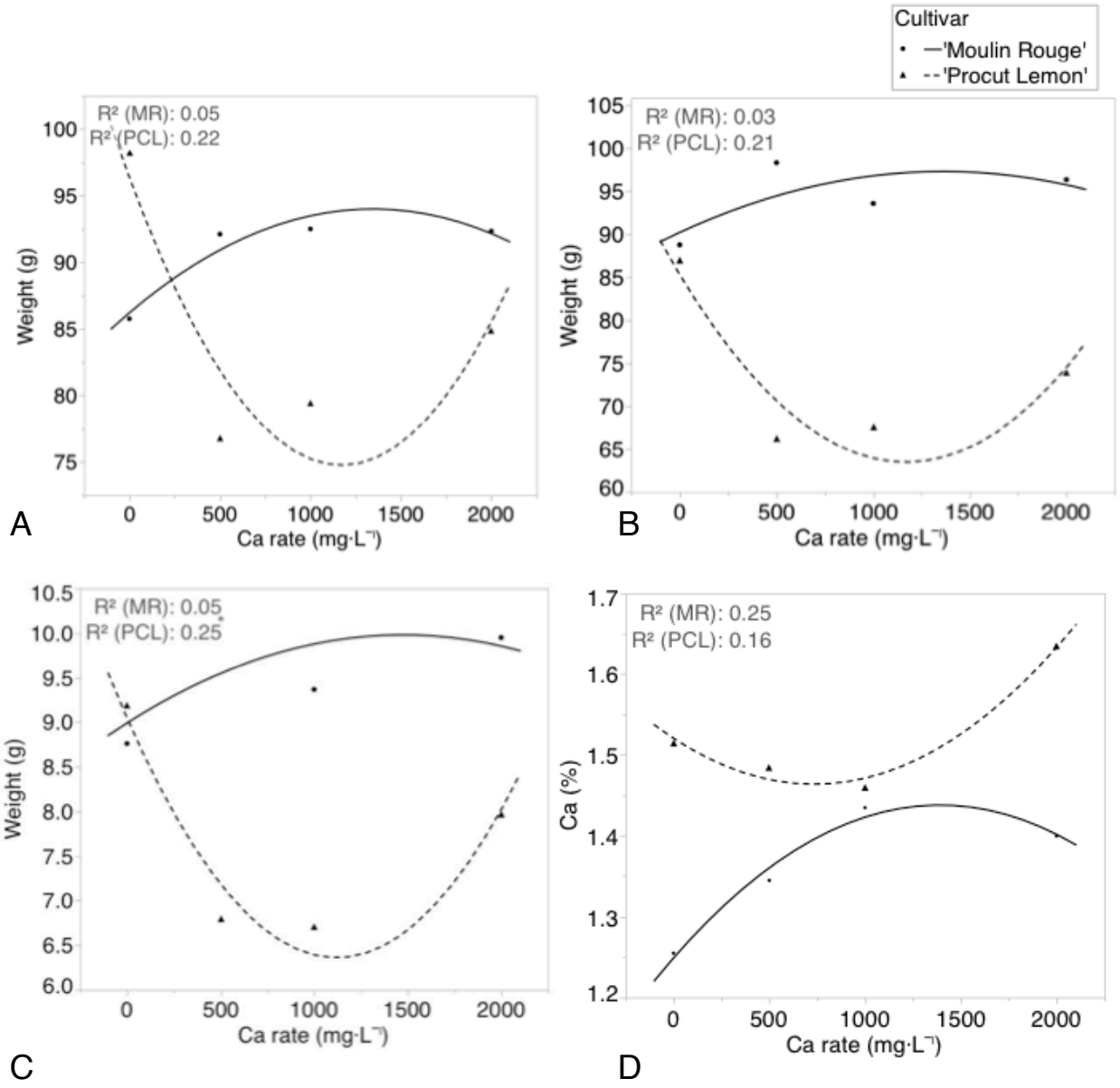
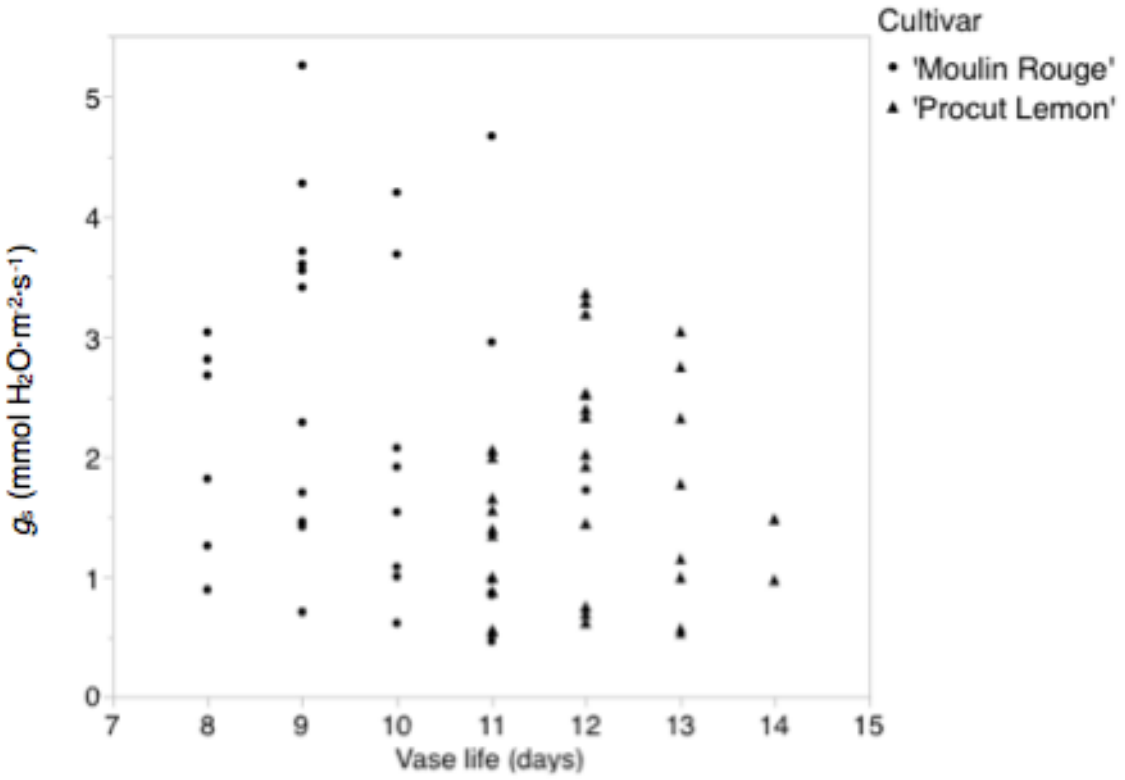
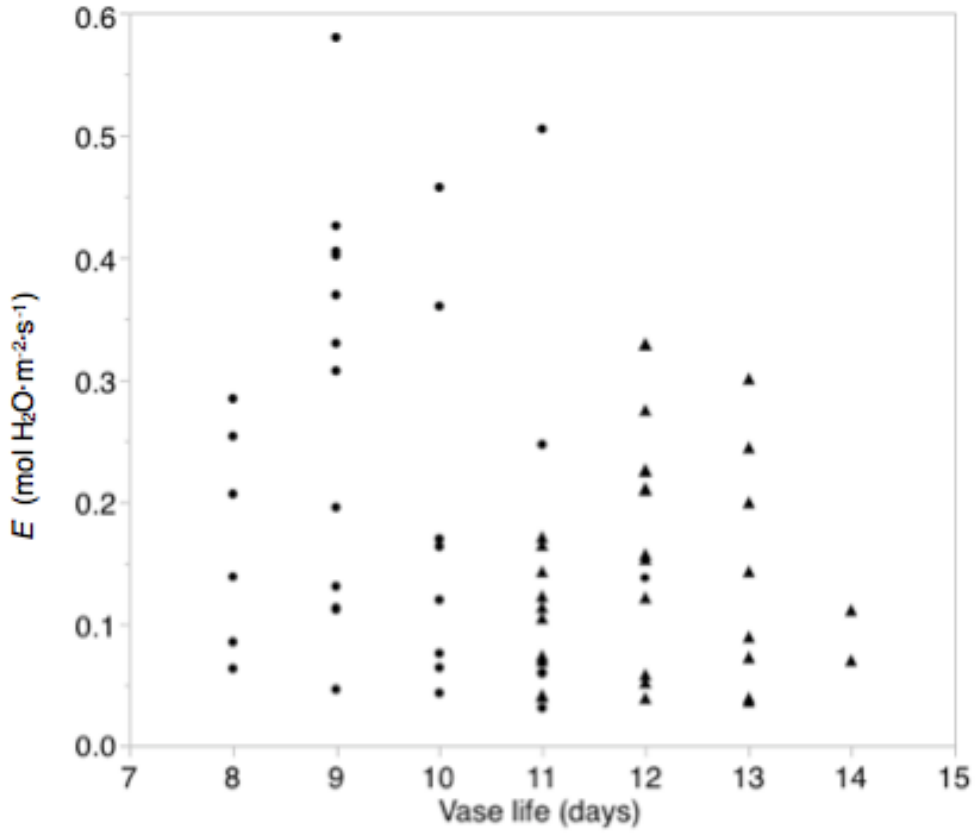


Figure 3.7. Correlation between stomatal conductance (g_s) and vase life (r = -0.259) or transpiration rate (E) and vase life (r = 0.268) of cut sunflower ‘Moulin Rouge’ and ‘Procut Lemon.’ Stomatal conductance and transpiration rates were measured 1 day after harvesting flowers for 33 ‘Moulin Rouge’ and 34 ‘Procut Lemon’ cut stems.




0.6
0.5
0.4
0.3
0.2
0.1
0.0
E (mol H2O·m-2·s-1)
7
8
9
10
11
12
13
14
15
Vase life (days)

Chapter 4: The effects of benzyladenine and exogenous ethylene exposure on postharvest characteristics of cut sunflowers and dahlias

Abstract

Applying benzyladenine (BA) to cut flowers can delay senescence and extend vase life. BA was applied as a preharvest spray or postharvest dip to sunflower (*Helianthus annuus* L.) 'Moulin Rouge' and 'Procut Lemon' flowers at 0 or 300 $mg \cdot L^{-1}$. Mean vase life of 'Moulin Rouge' flowers was 9.6 days and vase life of 'Procut Lemon' flowers was 12.5 days. BA treatment by either application did not change vase life. Measured rates of postharvest stomatal conductance and instantaneous transpiration rates were similar for all cut sunflowers. Stomatal conductance and transpiration rates showed significant decreases over the first three days after harvest. BA was also applied as a postharvest dip to dahlia (*Dahlia ×hybrida*) 'Karma Yin Yang' and 'Park Princess' flowers. Vase life of dahlia flowers was not extended by BA treatment. Research indicates a connection between plant responses to benzyladenine and ethylene. The reported ethylene sensitivity of dahlia cultivars varies. Dahlia 'Park Princess,' 'Bride to Be,' 'Cherish,' and 'Lollipop' were exposed to ethylene at 0.8 $\mu L \cdot L^{-1}$ for 18 hours. Ethylene exposure did not alter vase life in any of the cultivars studied.

Introduction

Consumers express dissatisfaction with cut flowers that have short longevity and are willing to pay more for flowers with extended vase life (Rihn et al., 2011, 2014). Preharvest environmental conditions play a role in determining the vase life of cut roses (*Rosa hybrida* L.;

In et al., 2016; Marissen, 2005) and freesia (*Freesia hybrida*; Slootweg, 2005). Low transpiration rates (*E*) have led to extended vase life in cut roses (In et al., 2016; Mayak et al, 1974) and lisianthus (*Eustoma grandiflorum*; Chuang and Chang, 2012).

Benzyladenine (BA) is a cytokinin that can delay senescence of flowers. Longer vase life in sunflowers (*Helianthus annuus* L.) corresponded to higher levels of endogenous cytokinins (Tata, 2013). Exogenous cytokinin applications may extend the vase life of some cut flower taxa. BA applications extended vase life of *Alstroemeria hybrida* L. (Hicklenton, 1991), lotus (*Nelumbo nucifera* Gaertn.; Imsabai and van Doorn, 2013), and tulips (*Tulipa* spp.; van Doorn et al., 2011). BA applications made directly to the flowers by dip or spray reduces the need for transport of BA through the flower stem and can be more effective than applying BA in the vase solution (Philosoph-Hadas et al., 1996). Response to treatment with BA varies with species and cultivar, so research is necessary to make specific recommendations (Paull and Chantrachit, 2001).

Plant responses to cytokinins such as BA have been genetically linked to responses to ethylene (Cary et al., 1995). Transformed petunia (*Petunia* × *hybrida*) with elevated endogenous cytokinin levels exhibited delayed flower senescence and a delay or reduction in ethylene biosynthesis (Chang et al., 2003). In some species, cytokinins regulate ethylene biosynthesis (El-Showk et al., 2013). BA application altered postharvest ethylene production in cut flowers of lisianthus (Asil and Karimi, 2010; Huang and Chen, 2002) and carnation (*Dianthus caryophyllus* L.; Mor et al., 1983). Similarly, treating broccoli (*Brassica oleracea* L. var. *italica*) florets with BA decreased ethylene production and extended their postharvest shelf life (Rushing, 1990).

Dahlia and sunflower are two specialty cut flower species with limited vase life. In sunflower, vase life can be limited by the loss of flower petals. Tata (2013) observed that sunflowers with red flowers were shorter-lived and more susceptible to petal drop than cultivars with yellow or orange flowers. Research has shown that sunflowers are insensitive to exogenous ethylene exposure (Tata, 2013; Woltering and van Doorn, 1988) and that the application of BA may help sunflowers hold their petals after harvest (Tata, 2013). In dahlia, BA applications extended the vase life of some cultivars (Shimizu-Yumoto and Ichimura, 2013). Dahlia cultivars that responded to BA also showed sensitivity to CEPA. CEPA is a plant growth regulator that decomposes to produce ethylene (Warner and Leopold, 1969). Previous research has indicated low ethylene sensitivity in other dahlia cultivars (Dole et al., 2009; Woltering and van Doorn, 1988).

The objective of this research was to test the effects of benzyladenine application on the postharvest vase life and quality of cut sunflower and dahlia. The impacts of BA application method on a short-lived cultivar, 'Moulin Rouge,' and a longer-lived cultivar, 'Procut Lemon,' were assessed, including effects on stomatal conductance (g_s) and transpiration rates (E). In dahlia, postharvest responses to BA dip application were measured for 'Karma Yin Yang' and 'Park Princess' flowers. Due to the connection between BA and ethylene responses and differences in the ethylene sensitivity of previously studied dahlia cultivars, further experiments were conducted to assess ethylene sensitivity in dahlia 'Park Princess,' 'Bride to Be,' 'Cherish,' and 'Lollipop.'

Materials and Methods

Sunflower

Helianthus annuus 'Moulin Rouge' and 'Procut Lemon' seeds (Johnny's Selected Seeds, Winslow, ME) were planted directly in trade gallon pots (2.8 L) containing soilless substrate (Fafard 52 mix; Sun Gro Horticulture Canada Ltd., Agawam, MA). Plants were irrigated when average volumetric water content (VWC) of the substrate was 18% or less, as measured with a Procheck handheld VWC meter (Decagon Devices, Pullman, WA). Beginning one week after planting, plants received 150 $mg \cdot L^{-1}$ N of 15N-2.2P-12.5K-4Ca-2Mg (Jack's Professional LX; J.R. Peters Inc., Allentown, PA) via Constant Liquid Feed (CLF). To ensure appropriate growing conditions, substrate pH and electrical conductivity (EC) were monitored every 2 weeks using the PourThru method. Substrate pH measured on July 29 was acidic (5.1 ± 0.1), so liquid lime was applied with irrigations on July 30 and August 5 to elevate substrate pH levels. Plants were grown in a greenhouse covered in a double layer of polyethylene film. The greenhouse temperature throughout the study averaged 26.5 ± 0.4°C during the day and 21.9 ± 0.3°C at night. Mean relative humidity was 53.0 ± 0.4 and mean daily light integral was 14.2 ± 1.3 $mol \cdot m^{-2} \cdot d^{-1}$.

The experimental design for this study was split-split plot (Figure 4.1). Cultivar was applied to whole plots at two levels, 'Moulin Rouge' or 'Procut Lemon.' Benzyladenine (BA; Configure, 10%; Fine Americas, Inc., Walnut Creek, CA) rate was applied to split plots at two levels, 0 or 300 $mg \cdot L^{-1}$. Method of BA application was applied to split-split plots at two levels, pre-harvest spray or postharvest dip of the flower head. BA spray was applied in the morning to 'Procut Lemon' plants on August 5, 2015 and to 'Moulin Rouge' plants on August 10, 2015.

Flowers of 'Procut Lemon' were harvested from August 7 to 11. 'Moulin Rouge' flowers were harvested from August 13 to 18. Sunflowers were harvested in the morning before 9:30 AM when bracts had lifted so that when looking directly at the flower head, at least 60% of the disc was visible, colored petals, but before all petals were completely expanded (Figure 4.2).

After harvest, the three leaves at the top of the stem were left on flower stems and all subsequent leaves were removed. For 'Moulin Rouge' flowers, any secondary flower buds in the top 30 cm of the stem were left on flower stems and subsequent buds were removed. Flower stems were cut to 50 cm and placed in tap water for transport to the postharvest laboratory.

The postharvest laboratory was maintained to simulate a typical consumer's home. Average temperature in the postharvest laboratory was 22.2 ± 0.0°C and average relative humidity was 48.9 ± 0.7%. In the postharvest lab, plants received ambient daylight through windows and supplemental light from fluorescent bulbs for ~8 $h \cdot d^{-1}$. Average instantaneous light intensity during the day was 127 ± 77 $\mu mol \cdot m^{-2} \cdot s^{-1}$. On the day of harvest, fresh weight of stems, stem caliper, and flower head diameter were measured. After recording these measurements, dip treatment was applied before stems were placed individually in vases containing 600 mL of deionized (DI) water.

Water uptake of each flower stem was measured 4 days after harvest (DAH) when vase water was replaced. A jar containing only DI water was used to quantify evaporation over the first 4 days of vase life; the volume of water evaporated from this jar was subtracted from the uptake for all flower stems. Flower diameter was measured again when flowers had completely opened by 5 DAH. Vase life was assessed visually once each day. Vase life was terminated when at least 50% of the flower petals had wilted or abscised, or when two or more undesirable aesthetic qualities were observed (Figure 4.3, 4.4). Observed reasons for terminating vase life

are listed in Table 4.1. After termination, flower stems were dried for at least 48 hours in an oven at 55°C and weighed to determine dry weight.

Before BA treatments were applied, stomatal conductance (g_s) and instantaneous transpiration rate (E) of the youngest, fully expanded leaves of sunflower plants were measured using an LI-6400XT (LI-COR, Linclon, NE) with the 6400-40 LCF chamber head attachment. After each flower harvest, g_s and E were measured for two stems from each split-split plot. Selected stems were measured at three times postharvest: 1 DAH, 3 DAH, and near the end of vase life (8 DAH for 'Procut Lemon' and 10 DAH for 'Moulin Rouge'). Environmental conditions observed during these measurements are listed in Appendix C.

Data were analyzed using SAS (SAS Institute, Cary, NC). Vase life, change in flower head diameter, change in fresh weight, uptake, and dry weight data were analyzed using univariate, split-split plot ANOVA. Stomatal conductance and E were analyzed over time using a repeated measures ANOVA. In all analyses, the time from spray to harvest was analyzed as a covariate of application method.

Dahlia and BA

Dahlia 'Park Princess' and 'Karma Yin Yang' tubers (Brent and Becky's Bulbs, Gloucester, VA) were planted in ground beds on June 10, 2015, at the Urban Horticulture Center in Blacksburg, VA. Average daily temperature during this study was 21.2 ± 0.3°C, average relative humidity was 86.4 ± 0.7%, and average daily solar radiation was 0.154 ± 0.006 $kW \cdot m^{-2}$.

Tubers were fertilized with controlled release fertilizer (Osmocote Plus 15-9-12, 12 to 14 month longevity at 21° C;15N-3.9P-10.0K; Scotts Miracle-Gro, Marysville, OH) at a rate of 4.7 $kg \cdot m^{-3}$ and mulched with bark to deter weeds. To encourage vegetative growth, flower buds

were removed from all plants for the first 8 weeks after planting. Tubers were irrigated immediately after planting. After this initial irrigation, rainfall was the primary source of water with supplemental irrigation only occurring when there was no rain for over 1 week. During this study, average daily rainfall was 2.90 ± 0.75 mm.

This experiment was repeated twice for 'Park Princess' and once for 'Karma Yin Yang.' 'Park Princess' flowers for the first experiment were harvested on September 4, 2015. Harvest for the second experiment with 'Park Princess' and harvest of 'Karma Yin Yang' flowers occurred on September 24, 2015.

Flower harvests occurred in the morning before 9 AM. Flowers were harvested when 50% to 75% of petals were expanded and flowers were placed in tap water for transport to the postharvest laboratory. Average temperature in the postharvest laboratory was 23.0 ± 0.0°C and average relative humidity was 55.6 ± 0.4%. In the postharvest lab, plants received ambient daylight through windows and supplemental light from fluorescent bulbs for ~8 $h \cdot d^{-1}$. Average instantaneous light intensity during the day was 127 ± 77 $\mu mol \cdot m^{-2} \cdot s^{-1}$.

After harvest, stems of each cultivar were randomly assigned to one of three treatments: 5 second dip of flowers into BA at 300 $mg \cdot L^{-1}$, 5 second dip of flowers into water, or no dip treatment (untreated control). Each treatment was replicated on 6 cut flower stems of each cultivar. For each stem, fresh weight and stem caliper were measured before applying dip treatments. After treatment, flower stems were placed individually in vases containing 500 mL of DI water. Water uptake was measured 5 days after the flower harvest on September 24. A jar containing only DI water was used to quantify evaporation over the first 5 days of vase life; the volume of water evaporated from this jar was subtracted from the uptake for all flower stems.

Vase life was assessed visually once each day in the morning. Vase life was defined as the number of days until at least 50% of the flower petals had wilted (Figure 4.5). Fresh weight of each stem was measured on the final day of vase life. At the end of vase life flower stems were dried in an oven at 55°C for at least 48 hours before measuring dry weight.

Dahlia and Ethylene

Vegetative cuttings were collected from dahlia 'Bride to Be', 'Cherish', 'Lollipop', and 'Park Princess' plants growing in ground beds in Blacksburg, VA, in October 2015. Cuttings received a 3-second basal dip of 1500 $mg \cdot L^{-1}$ indole-3-butyric acid (IBA) rooting hormone (Hortus IBA Water Soluable Salts 20% IBA; Hortus USA Corp., New York, NY). Cuttings were rooted in 72 cell trays (cell height 5.7 cm, volume 35.4 mL) in a soilless substrate (Fafard 3B; Sungro Horticulture). Cuttings were grown in a glass greenhouse under mist until roots began to form. Once rooting began, plants were moved to a polyethylene-covered greenhouse. Average temperature was 19.9 ± 0.1°C during the day and 18.4 ± 0.1°C at night. Average relative humidity was 30.1 ± 0.2% and average daily light integral was 6.9 ± 0.4 $mol \cdot m^{-2} \cdot d^{-1}$. After transplanting, dahlias were fertilized at each irrigation via constant liquid feed (CLF) at 150 $mg \cdot L^{-1}$ N of 15N-2.2P-12.5K-4Ca-2Mg (Jack's Professional LX; J.R. Peters Inc.).

Seven weeks after sticking, dahlia were transplanted to trade gallon (2.8 L) containers containing soilless media (Fafard 52 mix; Sungro Horticulture). Stems were supported with plastic mesh. Flower buds produced during the first 4 weeks after transplant were removed to encourage branching. CLF was increased to 200 $mg \cdot L^{-1}$ N beginning 10 weeks after transplant due to low electrical conductivity of the growing media (0.84 ± 0.19 $mS \cdot cm^{-1}$ as measured with the PourThru method); fertilization continued at this rate for the remainder of the experiment.

This experiment was repeated, with harvests occurring on March 9 and March 15, 2016. Flowers were harvested when 70% to 90% of petals had expanded. Harvests occurred before 11:00 AM and flowers were placed in water for transport to the postharvest laboratory. Depending upon availability of flowers, 12 to 16 stems of each cultivar were harvested. In the postharvest laboratory, all stems of each cultivar were cut to the length of the shortest stem. To determine uniformity of each cultivar, fresh weight of each stem was measured and flower quality was assessed. Each stem was placed individually in a vase containing deionized water.

All flowers received one of two postharvest treatments: exposure to ethylene at 0 (control) or 0.8 $\mu L \cdot L^{-1}$ of ethylene for 18 hours. Flower vases were placed in one of two plexiglass chambers at 2:00 pm on the day of harvest. Small fans were placed inside each chamber to encourage air circulation. A dish of activated carbon was placed in the control chamber to remove any exogenous ethylene. Ethylene was injected into the other chamber to reach the desired concentration. Samples of air were collected from each box at 1 and 18 hours after ethylene exposure. Ethylene concentrations of these air samples were verified using gas chromatography.

After 18 hours, flowers were removed from the chambers and placed in the postharvest laboratory for evaluation of vase life. Vase life was determined visually once each day and was defined as the time until at least half of the flower petals had wilted (Figure 4.5). Average temperature in the postharvest laboratory was 21.9 ± 0.1°C and average relative humidity was 15.6 ± 0.4%. In the postharvest lab, plants received ambient daylight through windows and supplemental light from fluorescent bulbs for ~8 $h \cdot d^{-1}$. Average instantaneous light intensity during the day was 5.00 ± 0.66 $\mu mol \cdot m^{-2} \cdot s^{-1}$.

Data for all dahlia experiments were analyzed using SAS JMP Pro 11 (SAS Institute). Data were separated by cultivar and by harvest date for each experiment and analyzed using analysis of variance (ANOVA) with a significance level of 0.05.

Results and Discussion

Sunflower

Cultivar, BA rate, and BA application method did not change the vase life, water uptake, postharvest change in fresh weight, or dry weight of flower stems of cut sunflower in this study (Table 4.2). BA application method and the time between spray application and flower harvest affected the change in flower diameter after harvest. Flowers harvested 6 or more days after the BA spray tended to open more after harvest (Table 4.3). However, only 'Moulin Rouge' flowers were harvested on these days, so this may reflect an effect of cultivar. Visually, there were no observed issues with flowers of either cultivar opening after harvest (Figure 4.6). Due to the lack of other benefits to applying a BA spray and the difficulty of timing spray applications to occur 6 days before harvest, this method is not recommended for promoting the opening of cut sunflowers after harvest.

Mean vase life of 'Moulin Rouge' was 9.6 days. 'Procut Lemon' stems had a mean vase life of 12.5 days. Similarly, Tata (2013) observed shorter vase life and increased susceptibility to petal drop in red-flowered sunflowers such as 'Moulin Rouge' than in cultivars with yellow flowers, including 'Procut Lemon.' Research indicates that cut flower consumers are willing to pay a premium for longer lasting arrangements (Rihn et al., 2014). This suggests that 'Procut Lemon' flowers could be more valuable than 'Moulin Rouge' flowers. 'Procut Lemon' flowers also matured about one week earlier than 'Moulin Rouge' flowers in this study, which could

further increase the value of 'Procut Lemon' for cut flower growers. However, florists and consumers have expressed preferences for some cultivars with red flowers such as 'Moulin Rouge' (Ferguson et al., 2012). Growers should assess the preference of their specific market when choosing sunflower cultivars. Note that vase life in this study may be longer than the vase life observed by customers and florists due to the ideal postharvest environment. Placing sunflowers in arrangements may expose the stems to higher levels of ethylene or microbes and may shorten postharvest longevity.

Petal abscission is a reported issue that can shorten the vase life of cut sunflowers (Tata and Wein, 2014). In this experiment, 26% of 'Moulin Rouge' and 3% of 'Procut Lemon' flowers lost petals by the end of vase life (Table 4.1). Petal wilt was the most frequently observed reason for terminating vase life in both cultivars. In previous studies where petal abscission was a common issue, sunflowers were harvested after petals had completely opened (Tata, 2013). Similar to our study, Clark et al. (2010) harvested several cultivars of sunflower when the petals were perpendicular to the stem axis; petal wilt and browning, not petal loss, were cited as the reasons for ending of vase life. Harvesting sunflowers before petals have opened completely may be a simple strategy for limiting the postharvest loss of petals.

In previous studies, BA has helped delay leaf yellowing in *Alstroemeria* (Hicklenton, 1991) and goldenrod (Philosoph-Hadas et al., 1996) and delayed blackening of cut lotus flowers (Imsabai and van Doorn, 2013). Discoloration of petals or flowers was not observed in 'Moulin Rouge' flowers, but 25% of 'Procut Lemon' flowers exhibited browning or discoloration of flower petals or bracts (Table 4.1). Discoloration was only observed in 'Procut Lemon' flowers that received a spray or dip with 0 $mg \cdot L^{-1}$ BA. BA may have helped delay discoloration in 'Procut Lemon' flowers treated at 300 $mg \cdot L^{-1}$ BA. However, all stems with observed

discoloration exhibited at least one other undesirable characteristic leading to their termination, including petal wilt, neck bend, or closing of the flower head. Thus, delayed discoloration of 'Procut Lemon' cut flowers did not extend vase life.

There was no difference between stomatal conductance (g_s, $P = 0.07$) and instantaneous transpiration rates (E, $P = 0.68$) measured on plants of either cultivar before BA application. After harvest and BA treatments, g_s and E differed between 'Moulin Rouge' and 'Procut Lemon' cut stems (Table 4.4). One day after harvest, g_s of 'Moulin Rouge' cut stems was 0.175 ± 0.014 mol $H_2O \cdot m^{-2} \cdot s^{-1}$ and E was 2.16 ± 0.15 mmol $H_2O \cdot m^{-2} \cdot s^{-1}$. The g_s and E of 'Procut Lemon' stems at the same time were lower at 0.118 ± 0.009 mol $H_2O \cdot m^{-2} \cdot s^{-1}$ and 1.44 ± 0.09 mmol $H_2O \cdot m^{-2} \cdot s^{-1}$, respectively. Decreased rates of postharvest E have been coupled with extended vase life in roses (In et al., 2016) and lisianthus (Chuang and Chang, 2012). Lower rates of E by 'Procut Lemon' flowers help explain the longer vase life of this cultivar.

Stomatal conductance and E decreased with time after harvest (Table 4.5). Both g_s and E decreased from 1 day to 3 days after harvest. However, g_s and E were not changed from 3 days after harvest until the final measurement at 8 and 10 days after harvest for 'Moulin Rouge' and 'Procut Lemon' cut stems, respectively. These observations suggest that stomata close during the first 3 days after harvest of cut 'Moulin Rouge' and 'Procut Lemon' flowers and remain closed thereafter. Roses exhibit a similar decrease in g_s during vase life (In et al., 2010). The time period immediately after harvest is a crucial time for controlling water loss via stomata of cut flowers.

Benzyladenine is not recommended for extending the vase life of sunflower 'Moulin Rouge' or 'Procut Lemon.' Harvesting sunflowers at the appropriate flower stage minimizes postharvest petal abscission. While BA delayed discoloration in 'Procut Lemon' stems, this

effect failed to extend vase life due to the presence of other senescence symptoms on treated stems. Stomatal conductance and *E* of cut sunflowers decreased during the first 3 days of vase life, so controlling water loss during this crucial time period may be an effective means of extending vase life.

Dahlia and BA

Mean vase life for 'Karma Yin Yang' flowers was nearly 7 days (Table 4.6). Vase life of 'Karma Yin Yang' flowers dipped in BA at 0 or 300 $mg \cdot L^{-1}$ was not different from vase life of untreated controls. In addition, 'Karma Yin Yang' cut flowers showed no changes in other measures of postharvest quality when receiving dip treatment with BA at 0 or 300 $mg \cdot L^{-1}$. Water uptake, fresh weight of flowers at harvest, weight change over vase life, and dry weight of stems after termination were similar in all treatment groups for 'Karma Yin Yang.' BA treatment failed to alter these measures of postharvest water relations in 'Karma Yin Yang' flowers.

Postharvest longevity of 'Park Princess' flowers differed between the two experiments. Stems harvested on September 4 had a mean vase life of nearly 9 days (Table 4.6). 'Park Princess' flowers harvested on September 24 had a shorter longevity of nearly 7 days. Environmental factors including temperature, relative humidity, light intensity, and vapor pressure deficit of the production environment affect the vase life of cut roses (*Rosa hybrida* L.; Marissen, 2005) and freesia (*Freesia hybrida*; Slootweg, 2005). At the time of flower harvest on September 4, the temperature was 23.1°C and relative humidity was 68.7%. The morning of September 24 was cooler and less humid, with an observed temperature of 16.6°C and relative humidity of 59.1%. These environmental conditions may have contributed to the observed

differences in vase life. Dahlia 'Park Princess' flowers harvested in a higher-humidity environment may have lost less water via transpiration, thus prolonging their vase life.

In both experiments, vase life of 'Park Princess' flowers receiving dip treatment did not differ from vase life of control flowers. Uptake was only measured for flowers harvested on September 24 and was not significantly altered by BA treatment. For flowers harvested on September 4, fresh weight at harvest, fresh weight change, and dry weight of stems were similar for all treatment groups of 'Park Princess.' For flowers harvested on September 24, 'Park Princess' flowers in the control group were significantly heavier than other flowers at harvest. This weight difference remained significant when dry weight was measured at the end of vase life. However, the change in fresh weight of 'Park Princess' flowers harvested on September 24 was not significantly different between treatment groups. Differences in flower weight did not correlate with water uptake or vase life of 'Park Princess' flowers (data not reported). Fresh weight of flowers at harvest was not a determining factor for postharvest longevity of 'Park Princess' cut flowers. A similar change in fresh weight among all flowers suggests that postharvest water relations were the same for all 'Park Princess' dahlias, regardless of their weight.

Previous research suggests that treatment with BA may extend the vase life of cut dahlia flowers (Shimizu-Yumoto and Ichimura, 2013). However, the results of our experiment do not support the use of BA for extending the vase life of cut dahlia 'Park Princess' or 'Karma Yin Yang.' Treatment with BA failed to extend the vase life of these cultivars. Studies of other cut flowers indicate that BA application is not effective in extending vase life of all taxa (Paull and Chantrachit, 2001; Sui et al., 2015).

Dahlia and Ethylene

Gas chromatography verified a lack of ethylene in the control chamber and ethylene levels of 0.85 ± 0.06 $\mu L \cdot L^{-1}$ in the second chamber. Ethylene levels remained stable from the start to end of the 18 hour duration when flowers were in the chambers.

At the time of harvest, fresh weight and quality of all flower stems of each cultivar was similar (data not reported). Exposure to ethylene had no effect on the vase life of the dahlia cultivars examined (Table 4.7). These results agree with observations of dahlia 'Karma Thalia' by Dole et al. (2009) wherein 'Karma Thalia' vase life was not altered by exposure to exogenous ethylene at 1 $\mu L \cdot L^{-1}$ for 16 hours. In contrast, Shimizu-Yumoto and Ichimura (2013) observed ethylene-sensitivity in dahlia 'Kokucho' when flowers were exposed to exogenous ethylene at 2 or 10 $\mu L \cdot L^{-1}$ throughout the postharvest evaluation. The active threshold level for ethylene to have physiological impacts on plants is generally cited as 0.1 $\mu L \cdot L^{-1}$ (Wills and Golding, 2014). Thus, exposure to ethylene at levels of 1 $\mu L \cdot L^{-1}$ is sufficient to elicit a response in ethylene sensitive cut flower taxa. Ethylene levels in produce markets average about 0.06 $\mu L \cdot L^{-1}$ ethylene (Wills and Golding, 2014). Exposure to ethylene at concentrations as high as 1 $\mu L \cdot L^{-1}$ for extended periods of time is unlikely for cut dahlias. Dahlia vase life is adversely affected by cold storage (Dole et al., 2009). Due to difficulty of storage and their short vase life, dahlia sales are usually limited to local markets, with a limited time period between harvesting and flowers reaching the final consumer.

Previous studies have connected exogenous BA applications to changes in ethylene biosynthesis of cut flowers and extended vase life (Asil and Karimi, 2010; Huang and Chen, 2002; Mor et al., 1983). Dahlia 'Kokucho' exhibited sensitivity to ethylene and the postharvest longevity of this cultivar was improved by BA application (Shimizu-Yumoto and Ichimura,

2013). The longevity of dahlia 'Park Princess' flowers in our experiments did not benefit from treatment with BA and flowers were insensitive to ethylene. These results suggest that ethylene-sensitivity and the effects of exogenous BA on postharvest longevity in cut dahlia may be linked. A linkage between ethylene sensitivity and extended vase life in cut flowers treated with exogenous BA is further supported by the results of our sunflower study. Sunflowers are insensitive to ethylene (Tata, 2013; Woltering and van Doorn, 1988) and the vase life of sunflower 'Moulin Rouge' and 'Procut Lemon' flowers was not extended by exogenous BA application.

Conclusions:

Harvesting sunflowers before petals have completely expanded may be a simple method to reduce the incidence of postharvest petal abscission. Applying BA by preharvest spray or postharvest dip is not recommended for extending the vase life of sunflower 'Moulin Rouge' or 'Procut Lemon.' Length of vase life in these cultivars may be limited by water loss via transpiration. 'Moulin Rouge' flowers exhibited higher postharvest g_s and E, as well as a shorter vase life than 'Procut Lemon' flowers. BA application did not alter g_s or E. Practices that induce stomatal closure and reduce postharvest transpiration, especially at the beginning of vase life, may extend the vase life of these sunflower cultivars.

BA was also ineffective at extending the vase life of dahlia 'Park Princess' and 'Karma Yin Yang.' 'Park Princess' flowers, as well as flowers of dahlia 'Bride to Be,' 'Cherish,' and 'Lollipop' were insensitive to exogenous ethylene exposure. Our results contrast observations of extended vase life in ethylene-sensitive dahlia 'Kokucho' flowers after they were treated with exogenous BA (Shimizu-Yumoto and Ichimura, 2013). These studies support the hypothesis that

the effectiveness of BA in extending vase life of dahlia is tied to ethylene sensitivity. In addition, studies of sunflower 'Moulin Rouge' and 'Procut Lemon' indicate that BA does not extend vase life of other cut flower taxa that are insensitive to ethylene.

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Tables and Figures

Table 4.1. Reasons for terminating vase life of sunflower 'Procut Lemon' and 'Moulin Rouge' flowers. Vase life was determined visually once each day. Some stems exhibited multiple reasons for termination.

Reason for termination of vase life	% incidence	
	'Procut Lemon'	'Moulin Rouge'
Wilting of petals	95	93
Abscission of petals	3	26
Flower head begins to close	40	22
Browning and discoloration of petals or bracts	25	0
Bending at neck of flower stem	73	0
Flower stem breakage	3	0

Table 4.2. Univariate analysis of variance of postharvest parameters for cut sunflowers. Cut sunflowers 'Procut Lemon' and 'Moulin Rouge' received treatment with benzyladenine (BA) at rates of 0 or 300 $mg \cdot L^{-1}$, applied as a preharvest spray or a postharvest dip. Flower harvests occurred 2 to 8 days after the BA spray application, so the time from spray to harvest was analyzed as a covariate of BA application method.

		Mean Square				
Source of variation	df	Vase life	Change in flower diameter	Water uptake	Change in fresh weight	Dry weight
Cultivar	1	26.1 ^{NS}	15.2 ^{NS}	799 ^{NS}	282 ^{NS}	0.241 ^{NS}
BA rate	1	0.260 ^{NS}	0.00269 ^{NS}	1950 ^{NS}	0.256 ^{NS}	5.79 ^{NS}
Cultivar × BA rate	1	0.812 ^{NS}	1.17 ^{NS}	122 ^{NS}	2.99 ^{NS}	0.512 ^{NS}
Application method	1	0.310 ^{NS}	29.3***	152 ^{NS}	4.74 ^{NS}	1.10 ^{NS}
Cultivar × application method	1	0.279 ^{NS}	3.35 ^{NS}	125 ^{NS}	7.80 ^{NS}	11.2***
BA rate × application method	1	2.46 ^{NS}	0.907 ^{NS}	64.3 ^{NS}	3.67 ^{NS}	1.87 ^{NS}
CV × BA rate × application method	1	0.169*	0.000519 ^{NS}	58.4 ^{NS}	0.122 ^{NS}	3.84*
Time spray to harvest	1	1.62 ^{NS}	20.9**	515 ^{NS}	5.43 ^{NS}	0.355 ^{NS}
Time spray to harvest × cultivar	1	2.51*	3.27 ^{NS}	200 ^{NS}	0.104 ^{NS}	12.6***

		Mean Square				
Source of variation	df	Vase life	Change in flower diameter	Water uptake	Change in fresh weight	Dry weight
Time spray to harvest × BA rate	1	1.47 ^{NS}	1.96 ^{NS}	209 ^{NS}	5.38	4.93*

^{NS},*,**,*** Nonsignificant or significant at $P \leq 0.05, 0.01,$ or 0.001, respectively.

Table 4.3 Postharvest change in flower diameter of sunflower 'Moulin Rouge' and 'Procut Lemon' receiving BA at 0 or 300 mg·L^{-1} as a postharvest dip or preharvest spray. BA spray was applied 2 to 8 days before harvesting flowers. Each value represents the mean of measurements from at least four flower stems.

		Change in flower diameter (cm)	
Application method	Time spray to harvest (days)	'Moulin Rouge'	'Procut Lemon'
Dip	0	3.42 ± 0.46	1.11 ± 0.27
Spray	2	-[z]	0.60 ± 0.36
Spray	3	3.07 ± 0.43	-0.13 ± 0.20
Spray	4	2.28 ± 0.47	0.84 ± 0.21
Spray	5	1.38 ± 0.66	0.62 ± 0.30
Spray	6	4.50 ± 0.53	-
Spray	7	4.69 ± 0.36	-
Spray	8	4.93 ± 0.32	-

[z]Lack of data indicates that no flowers of the cultivar were harvested on the day indicated.

Table 4.4 Repeated measures analysis of variance between subjects effects for stomatal conductance (g_s) and transpiration rates (E) of sunflower 'Moulin Rouge' and 'Procut Lemon' cut flowers treated with benzyladenine (BA) at 0 or 300 mg·L^{-1} applied as a preharvest spray or a postharvest dip. Stomatal conductance and E were measured 1 day after harvest (DAH), 3 days after harvest, and near the end of vase life (day 8 for 'Moulin Rouge' and day 10 for 'Procut Lemon'). Flower harvests occurred 2 to 8 days after BA spray was applied, so the time from spray to harvest was analyzed as a covariate of application method.

		Mean Square	
Source of variation	df	g_s	E
Cultivar	1	0.0441**	5.15**
BA rate	1	0.00123 ^{NS}	0.278 ^{NS}
Cultivar × BA rate	1	0.000376 ^{NS}	0.0000885 ^{NS}
Application method	1	0.0117 ^{NS}	0.284 ^{NS}
Cultivar × application method	1	0.0114 ^{NS}	0.472 ^{NS}
BA rate × application method	1	0.0107 ^{NS}	2.22 ^{NS}
CV × BA rate × application method	1	0.00105 ^{NS}	0.375 ^{NS}
Time spray to harvest	1	0.00845 ^{NS}	0.314 ^{NS}
Time spray to harvest × cultivar	1	0.0150 ^{NS}	1.15 ^{NS}
Time spray to harvest × BA rate	1	0.00232 ^{NS}	0.403 ^{NS}

^{NS},*,**,*** Nonsignificant or significant at $P \leq 0.05, 0.01$, or 0.001, respectively.

Table 4.5 Repeated measures analysis of variance within subjects effects for stomatal conductance (g_s) and transpiration rates (E) of sunflower 'Moulin Rouge' and 'Procut Lemon' cut flowers treated with benzyladenine (BA) at 0 or 300 mg·L^{-1} applied as a preharvest spray or a postharvest dip. Stomatal conductance and E were measured 1 day after harvest (DAH), 3 DAH, and near the end of vase life (8 DAH for 'Moulin Rouge' and 10 DAH for 'Procut Lemon'). Flower harvests occurred 2 to 8 days after BA spray was applied, so the time from spray to harvest was analyzed as a covariate of application method.

	Mean square			
	g_s		E	
Source	1 to 3 DAH	3 DAH to end	1 to 3 DAH	3 DAH to end
Mean	0.175***	0.000397 ^{NS}	20.7***	0.544 ^{NS}
Cultivar	0.0310 ^{NS}	0.00604 ^{NS}	3.13 ^{NS}	0.593 ^{NS}
BA rate	0.0122 ^{NS}	0.000276 ^{NS}	2.31 ^{NS}	0.0403 ^{NS}
Cultivar × BA rate	0.00575 ^{NS}	0.0000118 ^{NS}	1.82 ^{NS}	0.0463 ^{NS}
Application method	0.000728 ^{NS}	0.000269 ^{NS}	0.00670 ^{NS}	0.589 ^{NS}
Cultivar × application method	0.0138 ^{NS}	0.0143 ^{NS}	3.90 ^{NS}	0.889 ^{NS}
BA rate × application method	0.000471 ^{NS}	0.000340 ^{NS}	0.605 ^{NS}	0.174 ^{NS}
Cultivar × BA rate × application method	0.0162 ^{NS}	0.000358 ^{NS}	4.09*	0.906 ^{NS}

Source	Mean square			
	g_s		E	
	1 to 3 DAH	3 DAH to end	1 to 3 DAH	3 DAH to end
Time spray to harvest	0.00853 ^{NS}	0.00743 ^{NS}	0.907 ^{NS}	2.93*
Time spray to harvest × cultivar	0.000812 ^{NS}	0.0117 ^{NS}	0.876 ^{NS}	1.20 ^{NS}
Time spray to harvest × BA rate	0.000757 ^{NS}	0.000363 ^{NS}	0.0285 ^{NS}	0.0987 ^{NS}

^{NS},*,**,*** Nonsignificant or significant at $P \leq 0.05, 0.01,$ or 0.001, respectively.

Table 4.6. Effect of postharvest treatments on vase life and postharvest parameters of 'Karma Yin Yang' and 'Park Princess' dahlia cut flowers. Postharvest treatments included: dip of flower head in benzyladenine (BA) at 0 or 300 $mg \cdot L^{-1}$, or no dip treatment.

Treatment	Vase life (d)	Uptake (mL)	Fresh weight at harvest (g)	Fresh wt loss over vase life (g)	Dry weight (g)
'Karma Yin Yang'					
Dip: 0 $mg \cdot L^{-1}$ BA	7.0[z]	21.7	22.6	4.0	2.3
Dip: 300 $mg \cdot L^{-1}$ BA	6.5	27.5	21.4	2.4	2.2
No dip	6.5	30.8	23.3	3.1	2.4
P value	0.616	0.242	0.602	0.498	0.222
'Park Princess' harvested September 4					
Dip: 0 $mg \cdot L^{-1}$ BA	8.7	-	16.3	2.3	1.75
Dip: 300 $mg \cdot L^{-1}$ BA	8.7	-	18.8	1.3	1.98
No dip	8.3	-	15.7	1.6	1.63
P value	0.814	-	0.130	0.406	0.0882

Treatment	Vase life (d)	Uptake (mL)	Fresh weight at harvest (g)	Fresh wt loss over vase life (g)	Dry weight (g)
			‘Park Princess’ harvested September 24		
Dip: 0 mg·L^{-1} BA	6.8	23.3	12.1 b[y]	0.5	1.48 b
Dip: 300 mg·L^{-1} BA	6.8	30.8	12.1 b	1.2	1.47 b
No dip	6.3	31.7	16.9 a	1.3	1.88 a
P value	0.574	0.235	0.0035	0.370	0.0079

[z]n=6

[y]Means within a column followed by the same letter are not significantly different (student’s t test, $P < 0.05$).

Table 4.7. Mean vase life of cut dahlia 'Bride to Be,' 'Park Princess,' 'Cherish,' and 'Lollipop' flowers exposed to ethylene at 0 (control) or 0.8 $\mu L \cdot L^{-1}$ for 18 hours. Vase life was determined visually as the number of days until at least 50% of flower petals had wilted.

Ethylene concentration (ppm)	'Bride to Be'	'Park Princess'	'Cherish'	'Lollipop'
		March 9 harvest		
0	5.6	7.9	6.6	6.1
0.8	5.9	7.4	6.0	6.0
P value	0.405	0.347	0.207	0.761
		March 15 harvest		
0	5.3	5.0	5.7	5.4
0.8	5.3	5.7	5.5	5.1
P value	1.00	0.289	0.600	0.483

Figure 4.1. Experimental design for sunflower and benzyladenine experiment. Cultivar was applied to each of 4 whole plots at one of two levels: 'Moulin Rouge' (MR) or 'Procut Lemon' (PCL). Each whole plot contained 2 split plots; BA rate was applied to split plots at 0 or 300 $mg \cdot L^{-1}$. Split plots were divided into 4 split-split plots and BA application method was assigned to each split-split plot as a spray (S) application before harvest or as a postharvest dip (D). To account for differences in time from the preharvest spray application until sunflowers were ready to harvest, more split-split plots were assigned to the spray application than the dip application method. Each split-split plot contained 6 plants; postharvest data was collected for at least 2 plants within each split-split plot.

MR	0 $mg \cdot L^{-1}$	S	S	S	D	D	S	S	S	0 $mg \cdot L^{-1}$	MR
	300 $mg \cdot L^{-1}$	D	S	S	S	S	D	S	S	300 $mg \cdot L^{-1}$	
PCL	300 $mg \cdot L^{-1}$	S	D	S	S	D	S	S	S	300 $mg \cdot L^{-1}$	PCL
	0 $mg \cdot L^{-1}$	S	D	S	S	S	S	S	D	0 $mg \cdot L^{-1}$	

Figure 4.2. Sunflower stage at cut flower harvest. Minimum harvest stage for 'Moulin Rouge' (A) and 'Procut Lemon' (B) flowers occurred when bracts had lifted from the disc so that when looking directly at the flower head, at least 60% of the disc was visible, colored petals. 'Moulin Rouge' flowers opened rapidly, and maximum harvest stage (B) occurred when some, but not all, petals had opened beyond a 90° angle from the disc. For 'Procut Lemon' flowers, maximum harvest stage was when 100% of petals had lifted from the disc, but had not opened beyond a 90° angle from the disc.

Figure 4.3. Reasons observed for terminating vase life of sunflower 'Moulin Rouge.' 'Moulin Rouge' cut stems were terminated when at least 50% of petals exhibited petal wilt (A) or petal abscission (B), or when flower head was at least halfway closed (C). Stem breakage was also observed in two stems (not pictured). Note that some stems exhibited more than one of these qualities.

Figure 4.4. Reasons observed for terminating vase life of sunflower 'Procut Lemon.' 'Procut Lemon' vase life was ended when at least 50% of petals exhibited petal wilt (A) or petal abscission (not pictured), or when at least two of the following were present: closing of the flower head (B), neck bend (C), or discoloration of petals and/or bracts (D). Stem breakage was also observed in two 'Procut Lemon' flowers (not pictured). Note that some stems exhibited more than one of these qualities.

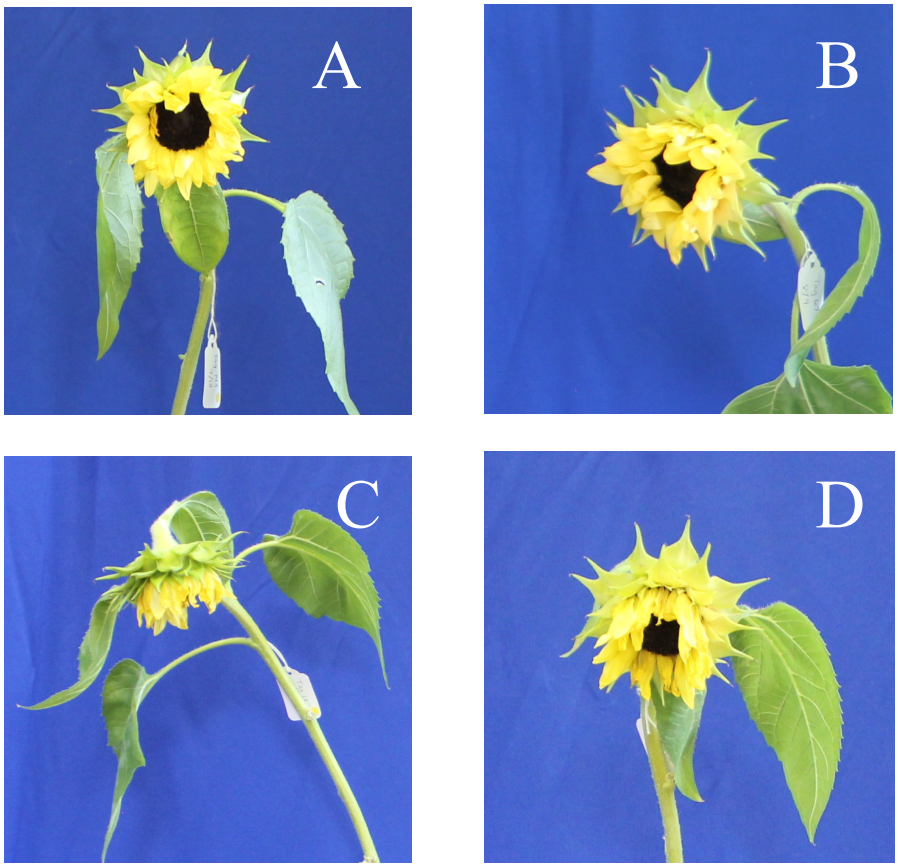

Figure 4.5. Petal wilt in dahlia ‘Park Princess,’ ‘Karma Yin Yang,’ ‘Bride to Be’ and ‘Lollipop’ flowers (clockwise from top left).

Figure 4.6. Cut ‘Procut Lemon’ (A) 4 days after harvest and ‘Moulin Rouge’ flowers 5 days after harvest. For both cultivars, treatments pictured from left to right are as follows: preharvest spray with 0 mg·L^{-1}BA, preharvest spray with 300 mg·L^{-1} BA, postharvest dip in 0 mg·L^{-1} BA, postharvest dip in 300 mg·L^{-1} BA. At the time that these photographs were taken, flowers had completely opened.

Chapter 5: The effect of simple handling and harvesting procedures on the vase life and postharvest quality of dahlia 'Karma Yin Yang' and 'Park Princess' flowers.

Abstract

Dahlia cut flowers often exhibit short vase life. These experiments were conducted to identify simple techniques that could extend vase life of cut dahlia 'Park Princess' and 'Karma Yin Yang' flowers. Dahlia were harvested when flowers were at budbreak, half open, or open. Vase life of 'Park Princess' flowers was extended when flowers were harvested half open or earlier, but 'Park Princess' flowers harvested at budbreak failed to open completely after harvest. 'Karma Yin Yang' vase life was not changed for flowers harvested at different flower stages. Placing 'Karma Yin Yang' and 'Park Princess' flowers in hot or ambient temperature vase water had different effects for flowers harvested on different days. Weather conditions may have altered the water requirements of flowers harvested on different days. However, placing 'Park Princess' or 'Karma Yin Yang' flowers in hot water is not recommended, as it had no effect or a negative effect on vase life. Fungicide (12.8% pyraclostrobin, 25.2% boscalid) was applied a rate of 900 $mg \cdot L^{-1}$ and volume of 75 $mL \cdot m^{-2}$ to 'Park Princess' and 'Karma Yin Yang' flowers three days before flower harvest. Fungicide extended vase life of 'Park Princess' flowers but did not change vase life of 'Karma Yin Yang.' Fungal infection was not observed in flowers receiving fungicide or control sprays, so the use of fungicide is not recommended for dahlia 'Park Princess' or 'Karma Yin Yang.'

Introduction

Market research indicates that consumers of cut flowers are willing to pay extra for flower arrangements with longer vase life and arrangements with a longevity guarantee (Rihn et al., 2014). However, the external appearance of flowers at harvest does not always accurately indicate postharvest longevity of cut flowers (In et al., 2016a, 2016b). To ensure optimal vase life of cut flowers, research is needed to identify the best harvesting and handling practices for specific taxa.

Dahlia are commonly grown as cut flowers but often exhibit short vase life, with cut stems that can last less than one week after harvest. Growers have adopted a variety of methods for harvesting and handling dahlias (Hankins, 2009). Dahlia flowers are commonly harvested when mostly open, but Dole et al. (2004) saw extended vase life in dahlia 'Karma Thalia' harvested at budbreak. Other sources suggest that placing freshly cut dahlias into hot water will encourage water uptake and could improve postharvest longevity (Cathey, 1978; Hankins, 2009). In addition, diseases, including *Botrytis*, can negatively impact dahlia flowers and foliage and should be controlled during dahlia production (Fanelli and Dole, 2006).

The objectives of this research were to identify simple measures that growers can take to extend the vase life of cut dahlia flowers. Specifically, experiments investigated the impact of flower stage, vase water temperature, and fungicide application on the postharvest longevity of dahlia 'Karma Yin Yang' and 'Park Princess.'

Materials and Methods

Dahlia 'Karma Yin Yang' and 'Park Princess' tubers (Brent and Becky's Bulbs, Gloucester, VA) were planted on June 10, 2015, in ground beds at the Urban Horticulture Center

in Blacksburg, VA. During this study, average daily temperature was 21.2 ± 0.3°C, average relative humidity was 86.4 ± 0.7%, and average solar radiation was 0.154 ± 0.006 $kW \cdot m^{-2}$. Tubers were fertilized with controlled release fertilizer (Osmocote Plus 15-9-12, 12 to 14 month longevity at 21°C; 15N-3.9P-10.0K; Scotts Miracle-Gro, Marysville, OH) at a rate of 4.7 $kg \cdot m^{-3}$ and mulched with bark mulch to deter weeds. To encourage vegetative growth, flower buds were removed from all plants for the first 8 weeks after planting. Tubers were irrigated immediately after planting. After this initial irrigation, rainfall was the primary source of water with supplemental irrigation only occurring when there was no rain for over 1 week. During this study, average rainfall was 2.90 ± 0.75 $mm \cdot d^{-1}$.

Flower stems were harvested at least 13 weeks after planting. Harvests occurred in the morning before 10 AM. Stems were placed in tap water for transport to the postharvest evaluation laboratory. In the postharvest laboratory, all stems were recut to a length of 20 cm and placed in individual vases containing 500 mL of deionized (DI) water. In the postharvest laboratory, average temperature was 22.8 ± 0.0°C and average relative humidity was 36.5 ± 0.5%. Plants received ambient daylight through windows and supplemental light from fluorescent bulbs for ~8 $h \cdot d^{-1}$. Average instantaneous light intensity during the day was 127 ± 77 $\mu mol \cdot m^{-2} \cdot s^{-1}$.

Flowers remained in postharvest laboratory for evaluation of vase life. Vase life was visually assessed once each day in the morning. End of vase life occurred on the first day when at least 50% of the flower petals had wilted (Figure 5.1). In each experiment, the water uptake was measured for every stem on the first day that vase life was terminated for any flower of that cultivar. A jar containing only DI water was placed in the postharvest laboratory to quantify water evaporation from the vase and the volume of water evaporated from this jar was subtracted

from the water uptake for each stem. In addition, the following characteristics were measured for all stems: vase life, fresh weight at harvest, and fresh weight at the end of vase life. After ending vase life, each stem was dried in an oven at 55°C for at least 48 hours before measuring dry weight.

Flower stage experiment

Dahlia 'Park Princess' and 'Karma Yin Yang' were harvested on October 9, 2015. Flowers were harvested based on flower stage. Six stems were harvested for each of three flower stages. The flower stages were as follows:

(A) Budbreak: flower head showing color with at least one petal lifted from the bud but less than 5% of flower petals expanded.

(B) Half open: intermediate stage of opening. 10% to 40% of petals expanded for 'Park Princess' and 20% to 75% of petals expanded for 'Karma Yin Yang'.

(C) Open: flower head almost completely open. 80% to 90% of petals expanded for 'Park Princess' and 90% to 95% of petals expanded for 'Karma Yin Yang'.

Vase water temperature experiment

This experiment was conducted twice. Harvests occurred on October 9 and October 16. Flowers of both cultivars were harvested when 50% to 75% of the petals had expanded. In this experiment, treatments were applied immediately after recutting stems. Stems were randomly placed into one of two treatments: hot vase water or ambient temperature vase water. Hot water was heated to 36°C to 43°C. After placing stems in vases, the hot water was allowed to cool to ambient temperatures. Ambient temperature water was 20°C to 23°C.

Fungicide experiment

On October 13, 2015, plants in this experiment received one of two spray treatments: control (tap water) or fungicide (12.8% pyraclostrobin, 25.2% boscalid, Pageant® Intrinsic™ brand fungicide; BASF, Research Triangle Park, NC). Fungicide was applied at a rate of 900 $mg \cdot L^{-1}$ and volume of 75 $mL \cdot m^{-2}$ in order to fully cover foliage and flower buds. Three days later, on October 16, six flowering stems from each treatment were harvested for 'Park Princess'. Due to lack of flowers at the appropriate flower stage on 'Karma Yin Yang' plants, four stems were harvested from control plants and six flowers from plants treated with fungicide.

Statistical analysis

Data were analyzed by cultivar and experiment using analysis of variance (ANOVA) in JMP Pro 11 (SAS Institute, Cary, NC).

Results and Discussion

Weather conditions

Harvests for these experiments occurred on October 9 (flower stage and vase water temperature experiments) and on October 16, 2015 (vase water temperature and fungicide experiments). Immediately prior to these harvests, dahlias received irrigation via rainfall without supplemental irrigation. On the morning of October 9, temperature and relative humidity were higher than on October 16 (Table 5.1). One hour before flowers were harvested on October 9, a small rain event (0.03 mm) occurred. On October 16, no rain events had occurred for over 3 days. Recent rainfall and high relative humidity on October 9 may have led to a more favorable

water status and reduced water loss by transpiration in any flower stems harvested on this date in comparison to flowers harvested on October 16.

Flower stage experiment

For both cultivars, flowers harvested at earlier developmental stages weighed less (Table 5.2). This was reflected in fresh weight at harvest, fresh weight at termination, and dry weights. Differences in dry weights of the flowers indicate that flowers harvested at earlier developmental stages had accumulated less biomass at the time of harvest. Water uptake did not differ between flowers harvested at different developmental stages; all stems showed the same ability to take up water from the vase.

Vase life of 'Karma Yin Yang' flowers was not altered by flower stage. 'Park Princess' flowers harvested when half open or earlier had significantly longer vase life than flowers harvested fully open. The extended vase life seen in 'Park Princess' flowers harvested at budbreak agrees with previous research reporting longer vase life for dahlia 'Karma Thalia' flowers harvested at budbreak (Dole et al, 2004). While cut flower growers generally harvest dahlias when flowers are almost completely open (Hankins, 2009), harvesting flowers at an early stage may be beneficial for the longevity of some cultivars such as 'Park Princess.'

Although the vase life of 'Park Princess' flowers harvested at an earlier flower stage was longer, these flowers failed to open completely in the vase (Figure 5.2). As expected, the percentage of open petals at harvest differed for flowers harvested at each flower stage, with flowers harvested at budbreak having the fewest open petals and open flowers having the most open petals (Table 5.2). Percentage of open petals remained significantly different among the flowers harvested at different stages at both 3 and 5 days after harvest. At 5 days after harvest,

flowers harvested at budbreak were more closed than flowers harvested when open, but flowers harvested when open or half open had a similar amount of open petals. For 'Park Princess,' it may be preferable to harvest flowers when they are half open due to improvements in vase life with minimal impact on postharvest flower opening. Waiting to harvest 'Karma Yin Yang' until flowers have fully opened will ensure flowers are open during vase life and will not be detrimental to postharvest longevity. Due to variations between cultivars, dahlia growers should conduct their own trials to determine the proper flower stage at which to harvest.

Vase water temperature experiment

The vase water temperature experiment was repeated. In both experiments, flower weights (fresh and dry) did not significantly differ between treatments (Table 5.3). In the first experiment harvested on October 9, 'Karma Yin Yang' and 'Park Princess' flowers took up significantly more hot water than ambient temperature water (Table 5.3). 'Park Princess' flowers placed in hot water had shorter vase life than flowers placed in ambient temperature water, while 'Karma Yin Yang' vase life was unaffected by water temperature. For all flowers harvested on October 16, vase water temperature had no impact on water uptake or vase life. Some sources suggest that placing cut dahlia stems in hot water may extend vase life (Cathey, 1978; Hankins, 2009), but the results of this study do not support this practice. In other cut flower taxa, water uptake occurs more rapidly in cold vase water (Slootweg, 1995). Rapid uptake of cold water has been attributed to the negative relationship between the solubility of gases and water temperature; as water temperature decreases, the solubility of gases in water increases. Thus, cold water has a greater capacity to absorb any air inside cut flower stems.

Vase life and water uptake of dahlias harvested on different days may have been impacted by weather conditions. Variations have been observed in the vase life of roses grown in greenhouses with different temperature and relative humidity levels (In et al., 2016a). On the first harvest day, October 9, temperature and relative humidity were increased in comparison to conditions on October 16 (Table 5.1). In addition, rainfall occurred 1 hour before the October 9 harvest. Flowers harvested on October 16 had last received rainfall over 3 days before harvest. Flowers harvested on October 16 tended to take up more water, regardless of treatment. This suggests that the water status of the dahlia flowers was more favorable on October 9 than October 16. The higher relative humidity and recent rainfall on October 9 may have helped minimize water loss of dahlias cut on this day. The effects of vase water temperature may only be important when dahlia stems are harvested under favorable conditions. In order to extend vase life, it may be beneficial to harvest dahlia flowers when environmental conditions, rather than manipulating vase water temperature.

Fungicide experiment

Fungal infection was not observed during postharvest evaluation of 'Karma Yin Yang' or 'Park Princess' flowers. *Botrytis cinerea* can limit vase life of cut gerbera (*Gerbera jamesonii* Bolus; Slootweg and Körner, 2009) and roses (*Rosa hybrid* L., Hammer et al., 1990). However, *Botrytis* infection did not occur during this evaluation of 'Park Princess' and 'Karma Yin Yang' flowers.

Vase life of 'Karma Yin Yang' flowers was about 6 days and was not altered by fungicide application (Table 5.4). Fungicide application extended the vase life of 'Park Princess' flowers by about 2 days. Applying fungicide did not change the water uptake or fresh or dry

weights of stems of either cultivar. The mechanism by which fungicide application extended 'Park Princess' vase life in this experiment is unclear. Pyraclostrobin fungicides have been shown to reduce respiration and improve tolerance of ornamental species to drought stress and cold stress (BASF, 2013). Fungicide applied to dahlia 'Park Princess' may have had a similar impact on the stress tolerance of these plants. 'Park Princess' flowers for this experiment were harvested under lower humidity conditions and had not received rainfall for over 3 days. Fungicide application may have improved the tolerance of flowers to these mild drought conditions in addition to the stress of being harvested. While fungal infection was not observed in this study, applications of pyraclostrobin fungicide may improve the vase life of some dahlia cultivars by increasing their tolerance of stressful conditions.

Conclusions

In general, vase life of dahlia 'Park Princess' and 'Karma Yin Yang' flowers was limited to 1 week or less. 'Park Princess' vase life can be improved without compromising flower opening by harvesting flowers when half open, while 'Karma Yin Yang' flowers should be harvested when flowers are fully opened. Growers should conduct trials to determine which dahlia cultivars benefit from being harvested at an earlier flower stage. Placing cut dahlias in hot vase water is not recommended as it had either a detrimental or no effect on the vase life of these cultivars. Fungicide application was unnecessary for preventing fungal infection in 'Karma Yin Yang' and 'Park Princess' flowers, but fungicide application extended vase life for 'Park Princess' flowers. Vase life differed for dahlias harvested on different days. Harvesting dahlia flowers under ideal weather conditions may be important in determining postharvest longevity of dahlias and deserves further research.

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Tables and Figures

Table 5.1. Environmental conditions at the time of dahlia 'Park Princess' and 'Karma Yin Yang' cut flower harvests. Flowers were harvested from ground beds in Blacksburg, VA, between 9 and 10 AM on October 9 and October 16, 2015.

	October 9	October 16
Air temperature (°C)	16.1	13.1
Relative humidity (%)	82.4	59.3
Last recorded rainfall amount (mm)	0.03	1.14
Time of last recorded rainfall (hours before harvest)	1	78

Table 5.2 Postharvest qualities of dahlia 'Karma Yin Yang' and 'Park Princess' cut flowers harvested at budbreak, half open, or open.

Flower stage at harvest	Weight at harvest (g)	Weight at end of vase life (g)	Dry weight (g)	Uptake (mL)	Vase life (days)	Percentage of open petals		
						0 Days after harvest	3 days after harvest	5 Days after harvest
				'Karma Yin Yang'				
Budbreak	11.2[z] b[y]	10.5 b	1.45 b	55.0	6.83 a	1.7 c	43.3 c	77.5 b
Half open	15.5 b	13.7 ab	1.83 b	27.5	6.33 ab	39.2 b	71.7 b	87.0 ab
Open	24.0 a	17.3 a	2.33 a	27.5	5.33 b	91.7 a	96.7 a	98.8 a
P value	0.0001	0.017	0.002	0.495	0.061	<0.0001	0.001	0.008
				'Park Princess'				
Budbreak	8.5 b	7.9 b	1.21 b	10.8	9.83 a	0.8 c	38.3 c	80.0 b
Half open	8.7 b	7.5 b	1.25 b	11.7	8.83 a	21.7 b	80.8 b	85.8 ab
Open	12.4 a	10.5 a	1.52 a	15.8	5.83 b	86.7 a	92.5 a	95.0 a
P value	0.002	0.002	0.008	0.694	0.0002	<0.0001	<0.0001	0.016

[z]n=6.

[y]Means within a column followed by the same letter are not significantly different (student's t test, $P < 0.05$).

Table 5.3. Postharvest qualities of dahlia 'Karma Yin Yang' and 'Park Princess' cut stems placed in ambient or hot water after harvest. This experiment was conducted twice with harvests occurring on October 9 and October 16, 2015. Ambient water temperature was 20°C to 23°C and hot water was 36°C to 43°C.

Water temperature	Weight at harvest (g)	Weight at end of vase life (g)	Dry weight (g)	Water uptake (mL)	Vase Life (days)
'Karma Yin Yang' harvested October 9					
Ambient	19.3[z]	13.3	1.97	25.0	5.5
Hot	19.9	14.5	2.05	47.5	6.3
P value	0.623	0.354	0.437	<0.0001	0.065
'Karma Yin Yang' harvested October 16					
Ambient	16.8	13.5	1.80	69.2	6.3
Hot	17.2	13.6	1.99	71.7	5.3
P value	0.901	0.969	0.453	0.707	0.125
'Park Princess' harvested October 9					
Ambient	13.0	10.8	1.63	24.2	6.0
Hot	12.1	9.7	1.50	37.5	4.0
P value	0.445	0.351	0.196	0.002	0.010
'Park Princess' harvested October 16					
Ambient	12.2	8.7	1.60	54.2	2.8
Hot	11.5	9.0	1.55	56.7	4.3
P value	0.621	0.830	0.658	0.757	0.153

$^{z}n=6$.

Table 5.4 Postharvest qualities of dahlia 'Karma Yin Yang' and 'Park Princess' cut stems receiving preharvest sprays of water (control) or fungicide (12.8% pyraclostrobin, 25.2% boscalid, Pageant® Intrinsic™ brand fungicide; BASF, Research Triangle Park, NC) at 900 $mg \cdot L^{-1}$. Spray was applied 3 days before harvest at volume of 75 $mL \cdot m^{-2}$ in order to cover foliage and flower buds.

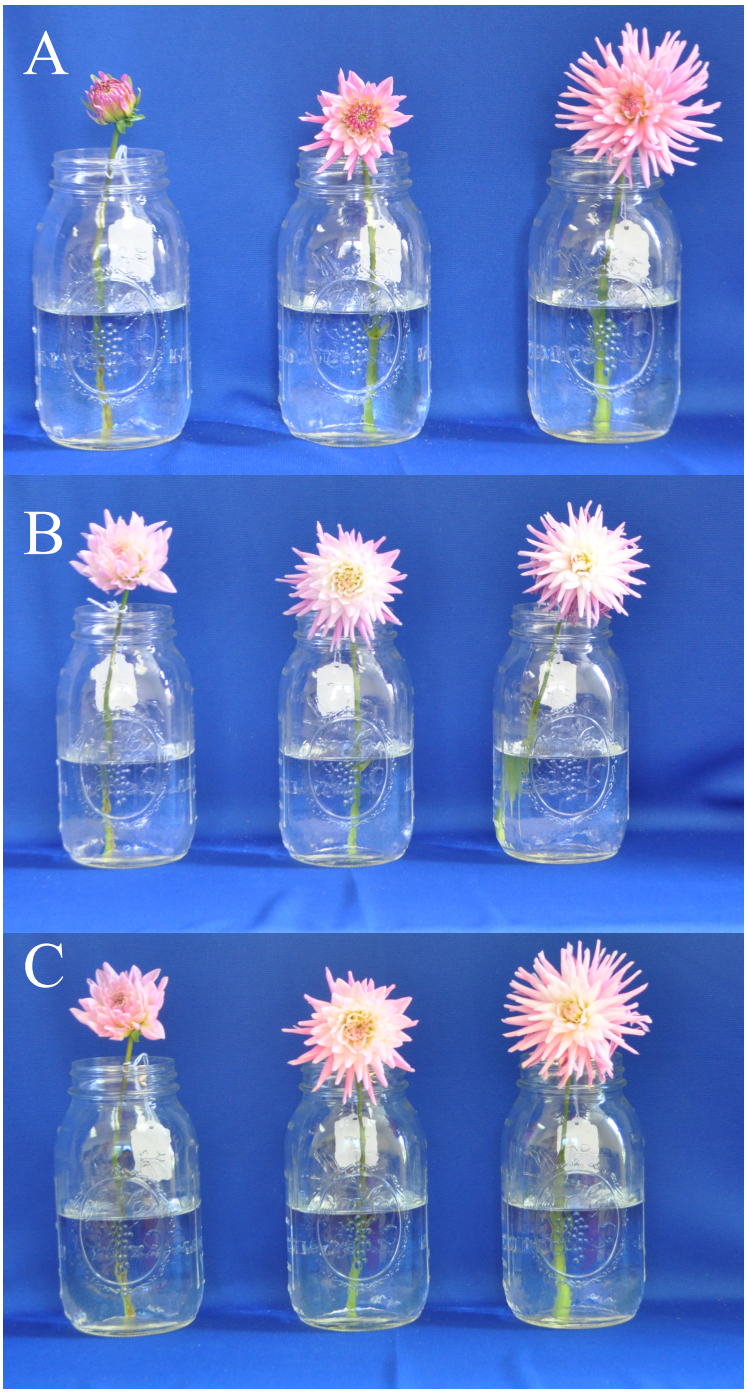
Treatment	Vase life (days)	Uptake (mL)	Weight at harvest (g)	Weight at end of vase life (g)	Dry weight (g)
			'Karma Yin Yang'		
Control[z]	5.8	53.8	12.9	11.5	1.65
Fungicide[y]	6.5	64.2	12.9	11.9	1.66
P value	0.225	0.392	0.992	0.806	0.996
			'Park Princess'		
Control	2.8	47.5	11.2	8.2	1.51
Fungicide	5.5	36.7	11.7	9.4	1.49
P value	0.021	0.294	0.737	0.309	0.926

[z]n=4 for 'Karma Yin Yang stems in the control treatment group.

[y]n=6 for all other treatments.

Figure 5.1. Petal wilt in dahlia ‘Park Princess’ (left) and ‘Karma Yin Yang’ (right).

Figure 5.2. Dahlia ‘Park Princess’ cut flowers at 0 (A), 3 (B), and 5 (C) days after harvest. Flowers were harvested at three different flower stages pictured from left to right: budbreak, half open, and open.



Chapter 6: Summary: Suggested Strategies for Improving Cut Flower Vase Life

Introduction

The majority of cut flowers sold in the United States are imported from other countries where production costs are low. In order to be competitive, flower growers in the United States focus on specialty cut flower varieties, which often have a short vase life. Extending the vase life of specialty cut flowers can increase their value. Recommendations for extending cut flower vase life vary. Production factors, harvest procedures, and postharvest handling can alter vase life. The objective of this research was to identify methods for optimizing the vase life of cut delphinium (*Delphinium elatum* L.), sunflower (*Helianthus annuus* L.), and dahlia (*Dahlia ×hybrida*) flowers. A variety of factors from production through postharvest were studied.

Materials and Methods

Vase life of cut delphinium flowers was investigated in two experiments. In the first experiment, delphinium 'Guardian Mix' plants were grown with fertilizer delivered as constant liquid feed at nitrogen rates of 50, 150, 250, or 350 $mg \cdot L^{-1}$ N. A second experiment investigated the impacts of drought stress on cut delphinium 'Guardian Blue' flowers. Deficit irrigation was applied at moderate or severe levels, while control plants received sufficient irrigation. Irrigation was applied based on volumetric water content (VWC) of the substrate.

The response of cut sunflowers to calcium fertilization or benzyladenine (BA) application was investigated. Calcium chloride was applied as a weekly foliar spray at 0, 500, 1000, or 2000 $mg \cdot L^{-1}$ calcium during production of 'Moulin Rouge' and 'Procut Lemon' flowers. In a separate

experiment, BA was applied to the same cultivars at 300 $mg \cdot L^{-1}$ as a preharvest spray or a postharvest dip.

Dahlia vase life was observed under a variety of conditions. For dahlia 'Karma Yin Yang' and 'Park Princess,' experiments studied the impact of preharvest fungicide application, the effect of flower stage at harvest, postharvest BA application, or vase water temperature. Ethylene sensitivity was determined for dahlia 'Park Princess,' 'Bride to Be,' 'Cherish,' and 'Lollipop' cut flowers.

Results and Discussion

Observed vase life of cut delphinium and dahlia was generally less than 1 week. Cut sunflower vase life was longer, with 'Moulin Rouge' flowers lasting 9 days and 'Procut Lemon' flowers lasting 12 days.

Maintaining a favorable water balance after harvest is essential to prolonging the vase life of cut flowers. For cut delphinium 'Guardian Mix' flowers, vase life was positively correlated with water uptake. Encouraging stems to take up more vase water may be the key to extending vase life of delphinium. In addition, minimizing water loss from stomata via transpiration may extend the vase life of cut flowers. 'Moulin Rouge' sunflowers with high rates of postharvest stomatal conductance and transpiration exhibited shorter vase life than 'Procut Lemon' sunflowers with lower stomatal conductance and transpiration rates.

Practices that encourage water uptake and minimize transpiration of cut flowers may extend their vase life. For cut delphinium 'Guardian Mix', nitrogen fertilization rate did not change water uptake. In addition, drought stress applied before harvest to delphinium 'Guardian Blue' did not alter water uptake or water loss by transpiration. For cut sunflower 'Moulin

Rouge' and 'Procut Lemon,' water uptake or water loss from stomata was not changed by foliar calcium applications or by BA applied before or after harvest. Postharvest BA application also did not affect water uptake of cut dahlia 'Karma Yin Yang' and 'Park Princess.' Placing these dahlia cultivars in hot vase water can encourage increased water uptake, but the impact of hot vase water on vase life was variable. Preharvest fungicide applications to dahlia 'Karma Yin Yang' and 'Park Princess' flowers also did not change water uptake. For these taxa, other practices should be tested to identify better methods for improving postharvest water relations.

Cut flowers should be harvested at the appropriate stage of flower development. For sunflower 'Moulin Rouge' and 'Procut Lemon,' harvesting flowers before the petals have completely opened can reduce petal abscission. Petal wilt was observed more frequently than petal abscission on flowers harvested at this stage. Dahlia 'Karma Yin Yang' should be harvested when fully open, as harvesting at earlier stages does not increase vase life. Dahlia 'Park Princess' vase life is extended by 3 days for flowers harvested when half open or earlier. However, harvesting 'Park Princess' flowers before they are half open is not recommended as flowers harvested at budbreak did not open completely after harvest. Growers should conduct individual tests for each cultivar to ensure the flowers are harvested at the appropriate stage.

Knowing the ethylene sensitivity of cut flower taxa may inform production and handling practices. Dahlia 'Park Princess,' 'Bride to Be,' 'Cherish,' and 'Lollipop' were not sensitive to exogenous ethylene exposure. 'Park Princess' flowers did not respond to BA treatment. In contrast, previous research showed extended vase life in dahlia 'Kokucho' when treated with BA (Shimizu-Yumoto and Ichimura, 2013). Dahlia 'Kokucho' was also found to be sensitive to ethylene. These results suggest a link between ethylene sensitivity and the impact of BA on cut flower senescence. This relationship is further supported by the results seen in sunflower

'Moulin Rouge' and 'Procut Lemon.' Sunflower vase life was not extended by BA application. Previous research has shown a lack of sensitivity to ethylene in sunflowers (Woltering and van Doorn, 1988). Senescence processes may differ between taxa that are ethylene sensitive and ethylene insensitive. Thus, factors affecting vase life will also be different.

The results of our research confirm that the vase life of specialty cut delphinium, dahlia, and sunflower can be short. In all taxa the observed vase life was less than 13 days, with delphinium and dahlia vase life limited to less than 1 week. Senescence symptoms that lead to the end of vase life are variable across taxa and differ for plants grown in different environments. Handling practices are taxa-specific. Growers can use this research in combination with other published cut flower research to better inform their production and handling methods.

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Appendix A: Environmental conditions during gas exchange measurements of delphinium 'Guardian Blue'

Stomatal conductance and transpiration rates were measured on the youngest, fully expanded leaves of Delphinium 'Guardian Blue' plants using a LI-6400XT (LI-COR, Linclon, NE). Delphinium 'Guardian Blue' plants in this experiment were subjected to irrigation treatments. Measurements were taken before applying the irrigation treatments, 2 weeks after the treatment began, and 1 day after harvest (DAH) of cut flowers. The experiment was repeated twice, with flower harvests beginning in January and February 2016 for the first and second experiments, respectively. The measurements on January 7 and 20 were made using the 6400-02 LED chamber attachment; all other measurements were made using the 6400-40 leaf chamber fluorometer chamber attachment. For all measurements, the stomatal ratio was set to 0.5. The stability factors were as follows:

(1) Photo
    a. Time: 15 seconds
    b. CV <3
    c. Slope <1

(2) Cond
    a. Time: 15 seconds
    b. CV <3
    c. Slope <1

(3) CO2R
    a. Time: 15 seconds

b. Slope <1

The required number of stability required for each measurement is included in the table.

Date	Measurement	Start time	End time	Stability required	Relative Humidity	Leaf temperature	Leaf vapor pressure deficit	PAR[z]	CO_2 sample	Flow
					January Experiment					
1/7/16	Pre-treatment	10:40 AM	11:40 AM	3/3	49.5 ±0.4	22.5±0.1	1.29±0.03	400.1±0.1	384.5±0.4	400.2±0.01
1/20/16	Post-treatment	8:55 AM	9:45 AM	3/3	49.0±0.4	21.3±0.1	1.22±0.02	200.6±0.1	389.9±0.3	500.2±0.4
1/25/16	1 DAH#1	9:35 AM	9:50 AM	2/3	44.3±0.7	21.1±0.1	1.36±0.04	62.6±0.0	399.7±0.4	400.3±0.01
1/27/16	1 DAH#2	10:20 AM	10:50 AM	2/3	42.0±0.6	22.2±0.3	1.48±0.05	98.9±0.2	398.3±0.1	400.3±0.01
1/29/16	1 DAH#3	2:25 PM	3:25 PM	2/3	35.5±0.5	21.8±0.5	1.65±0.07	35.5±4.1	399.4±0.1	400.3±0.01
1/31/16	1 DAH#4	9:00 AM	9:25 AM	2/3	36.0±0.5	23.9±0.1	1.83±0.03	57.5±5.3	400.1±0.0	400.3±0.01
2/2/16	1 DAH#5	10:30 AM	11:25 AM	2/3	35.3±0.3	21.8±0.5	1.66±0.07	75.9±9.0	398.9±0.1	400.3±0.01
2/4/16	1 DAH#6	8:50 AM	8:55 AM	2/3	51.2±0.1	22.1±0.1	1.21±0.00	99.2±0.2	398.9±0.0	400.3±0.05
2/6/16	1 DAH#7	9:15 AM	9:25 AM	2/3	51.4±1.1	19.5±0.2	1.05±0.05	101.2±0.2	399.6±0.8	400.4±0.02

February Experiment										
2/4/16	Pre-treatment	9:00 AM	10:00 AM	3/3	50.3±0.3	22.0±0.1	1.19±0.01	149.2±0.1	397.1±0.1	400.3±0.00
2/18/16	Post-treatment	10:10 AM	1:05 PM	3/3	55.5±0.5	24.8±0.1	1.32±0.02	499.8±0.4	397.6±0.3	400.2±0.00
2/21/16	1 DAH#1	10:00 AM	10:20 AM	2/3	54.5±0.4	22.2±0.1	1.15±0.02	100.1±0.1	399.3±0.3	400.3±0.01
2/23/16	1 DAH#2	8:30 AM	9:00 AM	2/3	32.8±0.2	23.5±0.1	1.92±0.02	99.5±0.1	399.2±0.2	400.3±0.01
2/25/16	1 DAH#3	9:20 AM	10:00 AM	2/3	40.2±0.2	21.5±0.1	1.50±0.02	101.5±0.2	399.8±0.2	400.3±0.01
2/29/16	1 DAH#4	9:50 AM	10:30 AM	2/3	40.6±0.6	22.2±0.1	1.52±0.03	100.6±0.1	398.9±0.4	400.3±0.01
3/2/16	1 DAH#5	9:05 AM	9:25 AM	2/3	36.3±0.5	19.6±0.1	1.40±0.01	102.7±0.1	398.9±0.2	400.4±0.01
3/5/16	1 DAH#6	10:05 AM	10:15 AM	2/3	37.6±2.0	19.9±0.1	1.39±0.07	102.3±0.3	399.2±0.6	400.4±0.01

[z]Photosynthetically active radiation

Appendix B: Environmental conditions during gas exchange measurements of sunflower 'Moulin Rouge' and 'Procut Lemon' receiving foliar calcium sprays

Stomatal conductance and transpiration rates were measured on the youngest, fully expanded leaves of sunflower 'Moulin Rouge' (MR) and 'Procut Lemon' (PCL) plants using a LI-6400XT (LI-COR, Linclon, NE) with the 6400-40 leaf chamber fluorometer chamber attachment. Sunflower plants in this experiment received foliar sprays of calcium chloride at 0 (control), 500, 1000, or 2000 $mg \cdot L^{-1}$ Ca. Measurements were taken on plants before flower harvests and on stems one day after harvest (DAH). For all measurements, the stomatal ratio was set to 0.5. For each measurement, the following stability factors were met:

(1) Photo
    a. Time: 15 seconds
    b. CV <3
    c. Slope <1
(2) Cond
    a. Time: 15 seconds
    b. CV <3
    c. Slope <1
(3) CO2R
    a. Time: 15 seconds
    b. Slope <1

Date	Measurement	Cultivar	Start time	End time	Relative Humidity	Leaf temperature	Leaf vapor pressure deficit	PAR[z]	CO_2 sample	Flow
8/6/15	Preharvest	MR, PCL	7:45 AM	10:00 AM	63.9±1.3	23.7±0.4	0.78±0.03	151.5±0.4	393.0±0.2	208.5±5.8
8/8/15	1DAH	PCL	10:40 AM	11:40 AM	58.7±0.3	24.2±0.0	1.17±0.02	198.3±0.1	393.9±0.4	200.2±0.0
8/9/15	1DAH	PCL	10:50 AM	12:00 PM	57.7±0.4	23.7±0.1	1.14±0.02	198.4±0.0	394.8±0.3	300.3±0.0
8/10/15	1DAH	MR, PCL	10:30 AM	2:00 PM	57.4±0.3	24.1±0.1	1.20±0.02	198.3±0.0	396.2±0.3	300.2±0.0
8/11/15	1DAH	MR, PCL	10:10 AM	12:15 PM	56.9±0.3	23.7±0.1	1.19±0.01	198.9±0.0	396.0±0.2	300.2±0.0
8/12/15	1DAH	MR, PCL	9:50 AM	12:10 PM	53.3±0.6	23.6±0.1	1.27±0.03	198.6±0.0	394.7±0.3	276.2±8.7
8/13/15	1DAH	MR	9:55 AM	10:45 AM	53.9±0.5	24.0±0.1	1.31±0.04	198.4±0.1	396.6±0.3	300.2±0.0
8/14/15	1DAH	MR	10:15 AM	11:15 AM	53.3±0.6	23.2±0.1	1.25±0.04	199.0±0.1	395.9±0.4	300.2±0.0
8/15/15	1DAH	MR	9:45 AM	10:25 AM	56.5±0.8	23.2±0.1	1.14±0.05	198.8±0.1	394.9±0.5	300.2±0.0

[z]Photosynthetically active radiation

Appendix C: Environmental conditions during gas exchange measurements of sunflower 'Moulin Rouge' and 'Procut Lemon' receiving treatment with benzyladenine

Stomatal conductance and transpiration rates were measured on the youngest, fully expanded leaves of sunflower 'Moulin Rouge' (MR) and 'Procut Lemon' (PCL) plants using a LI-6400XT (LI-COR, Linclon, NE) with the 6400-40 leaf chamber fluorometer chamber attachment. Sunflower plants in this experiment received benzyladenine (BA) at 0 or 300 $mg \cdot L^{-1}$ by a preharvest spray or postharvest dip application. Measurements were taken on plants before flower harvests and on stems 1 day after harvest (DAH), 3 DAH, and near the end of vase life (8 DAH for 'Moulin Rouge' and 10 DAH for PCL). For all measurements, the stomatal ratio was set to 0.5. For each measurement, the following stability factors were met:

(1) Photo
    a. Time: 15 seconds
    b. CV <3
    c. Slope <1

(2) Cond
    a. Time: 15 seconds
    b. CV <3
    c. Slope <1

(3) CO2R
    a. Time: 15 seconds
    b. Slope <1

Date	Measurement	Start time	End time	Relative Humidity	Leaf temperature	Leaf vapor pressure deficit	PAR[z]	CO_2 sample	Flow
8/4/15	Preharvest	7:50 AM	1:45 PM	63.5±0.7	28.0±0.2	1.15±0.03	340.8±11.1	392.8±0.3	441.7±11.2
8/8/15	1DAH (PCL)	10:40 AM	11:40 AM	58.7±0.3	24.2±0.0	1.17±0.02	198.3±0.1	393.9±0.4	200.2±0.0
8/9/15	1DAH (PCL)	10:50 AM	12:00 PM	57.7±0.4	23.7±0.1	1.14±0.02	198.4±0.0	394.8±0.3	300.3±0.0
8/10/15	1DAH (PCL), 3DAH (PCL)	10:30 AM	2:00 PM	57.4±0.3	24.1±0.1	1.20±0.02	198.3±0.0	396.2±0.3	300.2±0.0
8/11/15	1DAH (PCL), 3DAH (PCL)	10:10 AM	12:15 PM	56.9±0.3	23.7±0.1	1.19±0.01	198.9±0.0	396.0±0.2	300.2±0.0
8/12/15	1DAH (PCL), 3DAH (PCL)	9:50 AM	12:10 PM	53.3±0.6	23.6±0.1	1.27±0.03	198.6±0.0	394.7±0.3	276.2±8.7
8/13/15	3DAH (PCL)	9:55 AM	10:45 AM	53.9±0.5	24.0±0.1	1.31±0.04	198.4±0.1	396.6±0.3	300.2±0.0

8/14/15	1DAH (MR), 3DAH (PCL)	10:15 AM	11:15 AM	53.3±0.6	23.2±0.1	1.25±0.04	199.0±0.1	395.9±0.4	300.2±0.0
8/15/15	1DAH (MR)	9:45 AM	10:25 AM	56.5±0.8	23.2±0.1	1.14±0.05	198.8±0.1	394.9±0.5	300.2±0.0
8/16/15	1DAH (MR), 3DAH (MR)	8:40 AM	9:30 AM	56.7±0.3	24.0±0.1	1.21±0.02	198.3±0.1	395.7±0.3	300.2±0.0
8/17/15	1DAH (MR) 3DAH (MR) 10DAH (PCL)	9:45 AM	11:00 AM	55.8±0.6	23.5±0.1	1.20±0.03	198.9±0.1	396.2±0.3	300.2±0.0
8/18/15	1DAH (MR), 3DAH (MR) 10DAH (PCL)	10:15 AM	1:15 PM	40.6±0.6	23.6±0.1	1.67±0.03	198.4±0.1	395.9±0.3	300.2±0.0
8/19/15	1DAH (MR), 3DAH (MR) 10DAH (PCL)	9:55 AM	12:00 PM	51.6±0.5	23.7±0.1	1.36±0.03	198.4±0.1	396.9±0.3	300.2±0.0
8/20/15	3DAH (MR), 10DAH, (PCL)	9:25 AM	11:30 AM	46.7±0.3	23.9±0.1	1.54±0.02	199.0±0.1	397.4±0.2	252.4±10.6

8/21/15	3DAH (MR), 8DAH (MR), 10DAH (PCL)	9:40 AM	11:30 AM	44.5±0.4	23.9±0.1	1.61±0.02	198.6±0.1	396.7±0.3	300.2±0.0
8/22/15	8DAH (MR)	8:45 AM	9:20 AM	48.1±0.3	24.5±0.1	1.56±0.02	197.8±0.2	397.6±0.2	300.2±0.0
8/24/15	8DAH (MR)	9:35 AM	10:15 AM	48.4±0.1	24.4±0.1	1.54±0.01	198.3±0.1	397.3±0.3	300.2±0.0
8/25/15	8DAH (MR)	8:25 AM	9:25 AM	46.8±0.4	24.5±0.1	1.60±0.02	198.2±0.1	397.6±0.3	300.2±0.0
8/26/15	8DAH (MR)	10:15 AM	10:50 AM	45.2±0.4	24.1±0.1	1.61±0.02	198.2±0.1	397.5±0.3	300.2±0.0

[z]Photosynthetically active radiation