VASE LIFE DETERMINATION AND POSTHARVEST EVALUATION OF SPECIALTY CUT FLOWERS

BY

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VASE LIFE DETERMINATION AND POSTHARVEST EVALUATION OF SPECIALTY CUT FLOWERS

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The purpose of this study was to determine the postharvest characteristics of nine specialty cut flower species not yet available in the fresh cut flower market. In addition, the possibility of ethanol as a single preservative treatment with anti-ethylene and germicide properties was investigated.

This project was the fifth year of an on-going project funded by the Oklahoma Center for the Advancement of Science and Technology to encourage the production of cut flowers in Oklahoma. I want to thank my principal advisor Dr. John Dole for accepting me as his research assistant and for acting as my mentor and friend.

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CHAPTER I

INTRODUCTION

Wholesale production of cut flowers and greens totaled over \$419 million in 1993 for the 36 states with greatest sales (USDA, 1994). Cut greens comprised \$115 million and major species such as gladiolus, roses, carnations, and chrysanthemums contributed \$147 million. 'Other species', which include specialty cuts, comprised \$157 million. While Oklahoma cut flower sales totaled \$9.3 million for 1989 (Arnett, personal communication), only \$207 thousand in sales were actually produced within the state (USDA, 1994), demonstrating a significant deficit. Estimates for Oklahoma produced specialty cut flower sales in 1993 indicate an increase to approximately \$400 thousand (J. Dole, personal communication). In addition, Oklahoma is ideally situated close to several large metropolitan areas which may serve as potential consumers of Oklahomaproduced specialty cut flowers.

Interest in the production of specialty cut flowers is gaining among producers and consumers. The Association of Specialty Cut Flower Growers, a national organization devoted to the production of unique specialty cut flowers, has observed a 40% membership increase in 1992 (J. Laushman, personal communication). Oklahoma's neighboring state of Kansas had an estimated \$10-12 million of specialty cut flower production in 1992 (A. Stevens, personal communication). Oklahoma has the potential for producing a variety of field-grown specialty cut flowers (Bratcher, 1992). Species of

special interest are listed below:

Echinacea purpurea (L) Moensch (coneflower), is a native, early to mid-season flowering, herbaceous perennial with strong 1.5 to 3 ft stems and several pink, purple, or white daisy-like flowers. Coneflower is currently marketed as a dried flower, but is not common as a fresh cut flower. Helianthus maximilliani Schrad. (Maximillian's sunflower), is a native, late-season flowering, herbaceous perennial with 3 to 6 ft stems scattered with dozens of yellow daisy-like flowers creating a large spike. Penstemon digitalis Nutt. (beard's tongue) is a native, early-season flowering, herbaceous perennial with several tubular white to purple flowers on a spike. The cultivated annual Cosmos bipinnatus Cav. Ann. 'Sensation' (cosmos), flowers mid to late-season with 2 to 3 ft stems and pink, white, red, or purple daisy-like flowers. Celosia plumosa L.(celosia) is a cultivated, mid to late-season flowering annual with 1.5 to 2.5 ft stems and brilliant red, orange, and yellow plumes of tiny flowers. Achillea filipendulina Lam. 'Coronation Gold' (yarrow) a perennial, flowers early to mid-season with strong 2.5 to 3 ft stems terminating with flat clusters of yellow flowers. Cercis canadensis L. (eastern redbud), is a native, early-season flowering tree with pink or white pea-shaped flowers clustered along the stem. An early flowering shrub, Weigela sp. Thunb. (weigela), has loose clusters of tubular red, white, or pink flowers on long, straight stems. For mid to lateseason flowering, Buddleia davidii Franch. (butterfly bush) produces fragrant dense spikes of small blue, pink, red, purple, or white flowers.

A successful cut flower species must have the ability to withstand the various handling procedures throughout the market chain that can decrease vase life. The

marketing chain begins with harvest from the field. The cut flower is sold to the wholesaler and then passed on to local retail markets for sale to the consumer. A number of independent shippers and brokers may be involved at each stage. Throughout this marketing chain, the flower is potentially subjected to a number of abuses such as rough handling, poor quality water, and high temperatures which can accelerate senescence and decrease the quality and sales season duration of the flower.

Cut flowers are usually harvested during the early morning hours while the flower is turgid and there is time to ship it either to the wholesaler or directly to the retail market. Cut flowers must have the capacity to withstand packaging, temperature extremes, and long-distance transport.

Low temperature is the most important factor in the successful storage of cut flowers. A desirable cut flower should demonstrate the ability to undergo periods of cold storage, modified atmosphere storage, or low pressure storage. Low storage temperatures allow extended storage by slowing down floral senescence through a reduction in the rate of metabolic processes and bacterial growth (Nowak and Rudnicki, 1990; van Doorn and de Witte, 1991). By storing cut flowers, the grower, wholesaler, or retailer maximizes the sales season. Cut flower species such as peony (*Paeonia officinalis*), gladiolus (*Gladiolus sp*), and snapdragon (*Antirrhinum majus*) may be stored dry up to four weeks, while tulips (*Tulipa gesneriana*) may be stored for up to eight weeks (Nowak and Rudnicki, 1990). Flowers may be stored dry, such as peony, tulip, or wet, as with freesia (*Freesia sp*.) and gerbera (*Gerbera sp*.) (Nowak and Rudnicki, 1990). Wet storage, the most common practice, is normally only short term (1 to 2 days) and dry storage is for more than 3 days (Halevy and Mayak, 1981).

To further extend the postharvest life, cut flowers may be subjected to various preservative treatments which may contain silver thiosulfate (STS), 8-hydroxyquinoline citrate (8-HOC), and/or a carbohydrate source. Pretreatment or pulsing with STS inhibits the action of ethylene (Beyer, 1976; Joyce, 1988; Nowak, 1981; Nowak and Mynett, 1985; Reid et al., 1980; Staby and Reid, 1980; Tingley and Prince, 1990; Veen and van de Geijn, 1978). Ethylene is produced throughout the life of the plant. Ethylene may affect flower initiation and development, senescence, and fruit development. In some species, once the flower stem is removed from the plant, the synthesis of ethylene may be enhanced, expediting flower senescence. Extensive work has been performed to determine the site of ethylene synthesis, mode of action, site of accumulation and control methods (Bufler et al., 1980; Goh et al., 1985; Reid and Wu, 1992; Whitehead et al., 1984). Trace amounts of ethylene are produced by Cymbidium flowers when pollinia are removed which causes blushing of the labellum (anthocyanin accumulation) and flower Small quantities of ethylene can be responsible for initiating general senescence. senescence of the flower (Reid and Wu, 1992). Ethylene production typically consists of three distinct phases: (1) an initial low steady rate, (2) an accelerated rise to maximum emanation, and (3) a final decline in production (Halevy and Mayak, 1981).

Cut flower species vary in sensitivity to ethylene. Cut flower species such as alstroemeria (*Alstroemeria* spp.), carnation (*Dianthus caryophyllus*), and delphinium (*Delphinium hybrids*) are sensitive while species such as anthurium (*Anthurium andreanum*) and tulip are not sensitive (Nowak and Rudnicki, 1990). Cut flower species

that respond to low exogenous concentrations of ethylene are those in which ethylene is naturally involved in senescence (Reid and Wu, 1992). Ethylene sensitivity may vary among cultivars. Rose cultivars exhibited a variety of responses to exogenous ethylene such as acceleration of flower opening, abscission, distortion, and inhibition of flower opening (Reid et al., 1989). Cut flower species that produce ethylene may affect the longevity of other ethylene-sensitive species during shipping or storage.

Silver has long been recognized as the most effective measure to control ethylene action and limitation of vase life. Nowak (1981) confirmed the anti-ethylene effect of silver by observing that florets from snapdragon spikes treated with silver did not drop, but wilted and dried on the stem. Historically, silver nitrate was initially applied as an ethylene control measure. Researchers determined that the action and mobility of silver nitrate within the flower was greatly enhanced with the addition of sodium thiosulfate (Veen and van de Geijn, 1978). The combination of these two compounds led to the development of the STS complex. Reid et al. (1980) confirmed the efficacy of STS in extending carnation vase life. Staby et al. (1993) demonstrated that STS reduced or completely inhibited the abscission of flowers from delphinium and *Penstemon* spp. (beard's tongue). STS is recognized as the most effective measure of reducing ethylene sensitivity and greatly increasing vase life.

Treatment with 8-HQC prevents the accumulation of bacterial populations in the vase water (van Doorn et al., 1990). Microorganisms may lead to an increase in enzymatic damage, disruption of plant cell membranes, ethylene production, and physical occlusion at the base of or in the stem causing premature flower wilting (Larsen and

romarty, 1967; Marousky, 1971; Marousky, 1980; van Doorn et al., 1990; Zagory and .eid, 1986). 8-HQC may also effect flower longevity by acidifying the water which has een attributed to decreasing enzymatic activity and degradation of the xylem while .nhancing solution uptake (Marousky, 1971).

Carbohydrates are the primary source of nutrition and energy for the flower and are necessary for maintaining biochemical and physiological processes. Sugars support processes that are fundamental in prolonging vase life by maintaining mitochondrial structure and functions, improving water balance by regulating transpiration, and increasing water uptake (Halevy and Mayak, 1979; Nowak and Rudnicki, 1990). Leaf blackening of three *Protea* species during shipping and storage has been attributed to the postharvest inflorescence sink demand (McConchie and Lang, 1993). Postharvest treatments with a carbohydrate source is the most effective means to prevent leaf blackening. Han (1992) demonstrated that pulsing with concentrations of sucrose greater than 10% for 20 hrs improved the postharvest life of cut *Liatris spicata* (L) Willd. by increasing the length of inflorescence showing color and by prolonging the vase life of the spikes.

Alcohols have been studied as a method to reduce ethylene sensitivity (Heins, 1980; Paull and Goo, 1982; Saltveit, 1989; Saltveit and Mencarelli, 1988; Wu et al., 1992). Continuous treatment with 8% ethanol doubled the vase life of 'White Sim' standard carnation flowers and almost no ethylene was produced by ethanol-treated flowers (Wu et al., 1992). However, Wu et al. (1992) did not compare ethanol to STS, the commercially accepted control of preventing ethylene action in extending the vase life

carnations. Other possible characteristics of ethanol such as germicidal properties were t examined.

Nowak and Rudnicki (1990) anticipate that all ethylene sensitive flower species ill require treatment prior to entering international markets. However, STS is an xpensive, harmful heavy-metal environmental contaminant (Altman and Solomos, 1993). Alcohols may provide a single preservative treatment that is environmentally safe, less expensive, and easily made. Alcohols may replace STS and/or 8-HQC as preservatives.

Objectives

The research presented has three objectives:

to determine the effects of storage temperature and duration, shipping duration, and reservatives on the postharvest life of nine field-produced cut flower species;

) to determine the postharvest ethylene production and sensitivity of these cut flower pecies;

3) to determine the effect of ethanol on ethylene synthesis and sensitivity, microbial growth, and postharvest life of cut flowers.

The information provided through this research will provide sufficient information to enable growers to diversify production by offering marketable, unique regionallyproduced specialty cut flowers.

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CHAPTER II

VASE-LIFE DETERMINATION OF NINE SPECIALTY CUT FLOWER SPECIES

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Abbreviations. STS, silver thiosulfate; 8-HQC, 8-hydroxyquinoline citrate

Abstract. Selected postharvest attributes of flowers of three native herbacious, three cultivated herbaceous, and three woody species were studied. *Echinacea purpurea* (purple coneflower) had a vase life in deionized (DI) water of 9.1 days and could be stored for one week at 2 or 4C and shipped up to five days. *Helianthus maximilliani* (Maximillian's sunflower) had a vase life of 6.0 days in DI water and could be stored for one week at 2C and shipped for one day. *Penstemon digitalis* (penstemon) had a vase life of 9.3 days in deionized (DI) water and could be stored at 2C up to three

weeks with no decrease in vase life. Achillea filipendulina 'Coronation Gold' (yarrow) had a vase life of 10.7 days in DI water and could be stored at 2C up to 2 weeks and shipped for two days. Celosia plumosa 'Forest Fire' (celosia) had a vase life of 8.8 days in DI water and could be stored at 2, 4, or 7C for one week. Cosmos bippinatus 'Sensation' (cosmos) had a vase life of 6.2 days in DI water and could be stored at 2C up to two weeks and shipped for one day. Buddleia davidii (butterfly bush) had a vase life of 4.9 days and could be stored at 2C up to 2 weeks. Cercis canadensis (eastern redbud) had a vase life of 5.6 days in DI water and could be stored at 2C up to two weeks. Weigela sp. (weigela) had a vase life of 5.0 days in DI water and did not tolerate cold storage or shipping. Silver thiosulfate (STS) and 8hydroxyquinoline citrate (8-HQC) increased the vase life of Maximillian's sunflower, 'Coronation Gold' yarrow, 'Forest Fire' Celosia, butterfly bush, and weigela. Sucrose increased the vase life of Maximillians sunflower, 'Coronation Gold' yarrow, butterfly bush, and weigela. An additional 50% of the total number of buds opened at vase life termination when butterfly bush and eastern redbud were harvest with $0 \le 25\%$ buds open. Butterfly bush and eastern redbud stems harvested with $25 \le 50\%$ buds open had 50 <. 75% buds open at vase life termination. Butterfly bush and eastern redbud harvested with $50 \le 75\%$ and $75 \le 100\%$ had 100% of buds open at vase life termination.

Introduction

A successful cut flower species must have the ability to withstand the various

handling procedures throughout the market chain that decrease vase life. Low temperature is the most important factor in the successful storage of cut flowers. Storage temperatures just above freezing allow extended storage by slowing down the senescence process of flowers through a reduction in metabolic processes and bacterial growth rate (Nowak and Rudnicki, 1990; van Doorn and de Witte, 1991). Cut flowers may also be subjected to various preservative treatments which may contain STS, 8-HQC, and/or a carbohydrate source to extend postharvest life (Nowak and Mynett, 1985; Reid et al., 1980; Staby and Reid, 1980; Tingley and Prince, 1990; van Doorn et al., 1990).

Harvest at the appropriate stage of flower development influences appearance and longevity. Optimal stage of flower development for harvest depends upon species, cultivar, season, distance to market, and consumer preference (Nowak and Rudnicki, 1990). Cut flowers such as carnation (*Dianthus caryophyllus*) may be harvested at less advanced stages of development in summer than winter. Flowers harvested for direct sale may be harvested at more advanced stages than flowers shipped long distances.

Bratcher (1992) identified nine specialty cut flower species which were suitable for cut flower production in Oklahoma due to rapid, uniform germination and rooting, resistance or tolerance to insect and disease damage, attractive flower color, and strong stems with sufficient length for cut flowers. The objectives of the research reported here were to determine the effects of low temperature storage, ambient temperature shipping, and preservative treatment on nine cut flower species and to determine the

appropriate developmental stage for butterfly bush and eastern redbud harvest.

Material and Methods

Postharvest Experiments. Flower stems of three native herbaceous species: Echinacea purpurea (L) Moensch (coneflower), Helianthus maximilliani Schrad. (Maximillian's sunflower), Penstemon digitalis Nutt. (penstemon); three cultivated herbaceous species: Achillea filipendulina Lam. 'Coronation Gold' (yarrow), Celosia plumosa L. 'Forest Fire' (celosia), Cosmos bipinnatus Cav. Ann. 'Sensation' (cosmos); and three woody species: Buddleia davidii Franch. (butterfly bush), Cercis canadensis L. (eastern redbud), and Weigela sp. Thunb. (weigela) were grown and harvested (Mar.- Oct., 1993-94) from field plots at the OSU Nursery Research Station (Stillwater, OK). Coneflower, Maximillian's sunflower, and cosmos were harvested when the first ring of disk florets were fully open. Penstemon, celosia, weigela, eastern redbud, and butterfly bush were harvested when approximately 50% of the flowers were open. Yarrow flowers were harvested when 100% of florets were newly opened.

Cut flower stems were harvested no later than 11:00 a.m. and were re-cut to 30.0 cm for all species except celosia which was re-cut to 25.0 cm. After recutting, all cut flower stems were randomly assigned to one of 18 postharvest treatments that were grouped into the following 4 experiments: 1) storage duration treatments were held in deionized (DI) water in a dark cooler for 1, 2, or 3 wk at $2 \pm 1C$; 2) storage

temperature treatments were held in DI water in a dark cooler for 1 wk at 2, 4, or $7\pm$ 1C; 3) shipping duration treatments were placed in DI water in a dark room for 1, 2, 3, 4, or 5 days at $24\pm 2C$ and 4) preservative treatments included stems that were or were not pretreated (pulsed) in 1mM silver thiosulfate (STS) for 1 h at $2\pm 1C$ and were then held for 24 h in a dark cooler at $2\pm 1C$ in 200 mg liter⁻¹ hydroxyquinoline + 300 mg liter⁻¹ citric acid (8-HQC) and either 0, 4, or 8% sucrose. Pre-treated stems were transferred to DI water and held at ambient temperature ($23\pm 2C$) under continuous light exposure. STS was prepared and stored as recommended by Cameron et al. (1985). Control stems for all experiments were held in DI water at ambient temperature under continuous light.

The vase life of each cut flower species was calculated as the total number of days from harvest to termination and as post-treatment vase life (PTVL), the number of days after removal from a specific treatment to termination. The vase life of the cut flower stems was terminated when 50% or more of the flowers exhibited visual senescence symptoms which included in-rolling, wilting, shattering (petal drop), drying, or loss of color. Penstemon and weigela were terminated when flowers exhibited 50% or greater in-rolling and shattering. Purple coneflower and eastern redbud were terminated when flowers exhibited 50% or greater wilting and loss of color. In addition, eastern redbud exhibited shattering as a senescence symptom. Yarrow, celosia, Maximillian's sunflower, cosmos, and butterfly bush were terminated when the flowers exhibited 50% or greater wilting.

Ten Maximillian's sunflower and eastern redbud stems per treatment were

placed in a completely randomized design. Ten purple coneflower, penstemon, 'Coronation Gold' yarrow, 'Forest Fire' celosia, 'Sensation' cosmos, butterfly bush, and weigela stems per treatment were blocked by harvest date and were harvested from different plants. Data within each experiment were analyzed by General Linear Model procedure with means separation by trend analysis (SAS Institue, Cary, N.C.) and all means were compared to the control using Dunnett's test (SAS Institute).

Development stage study. Eastern redbud and butterfly bush were grown as in postharvest experiments and harvested at four developmental stages: (1) $0 > to \le 25\%$ of buds open, (2) >25 to $\le 50\%$ of buds open, (3) >50 to $\le 75\%$ of buds open, (4) > 75 to $\le 100\%$ of buds open. After harvest from the field, cut flower stems were re-cut to 30.0 cm and held in DI water at ambient temperature (23±2C) under continuous light until termination of vase life. Developmental stage of buds open at vase life termination was recorded for each stem. Ten stems per development stage of each species were harvested, blocked by harvest date, and placed in a completely randomized design.

Results

Echinacea purpurea. In both years, storage for one week at 2 and 4C did not significantly decrease vase life after treatment (Table 2.1). Stems stored at 7C had a maximum total postharvest life of 14.0 days in 1993. Stems stored at 4C in 1994 had

a maximum total postharvest life of 14.6 days. In 1993, purple coneflower stems may be stored up to three weeks at 2C without significantly effecting vase life after treatment, with a total postharvest storage and vase life of up to 21 days. In 1994, storage of any duration decreased vase life after treatment. Simulated shipping from one to five days did not affect vase life in both years. Stems shipped in 1994 had a maximum total postharvest life of 11.4 days. Purple coneflower showed various responses to preservative treatments. Treatment with STS and 0% sucrose significantly increased total vase life and vase life after treatment in 1993 compared with the control, but in 1994 stems treated with 4 or 8% sucrose, but no STS, had decreased vase life after treatment. Within the preservative treatments, no significant difference was found in either year.

Helianthis maximilliani. Storage for one week at 2C did not significantly decrease vase life after treatment in 1993 (Table 2.1). Storage for one week at 4 and 7C significantly decreased vase life after treatment in 1993. PTVL of stems stored at 2, 4, or 7C were not different from the control in 1994. Storage at 2C beyond one week significantly decreased vase life after treatment in both years. Maximum total postharvest vase life was 11.0 days for stems stored one week at 2C in 1994. Vase life after treatment linearly decreased as the simulated shipping duration increased, while total vase life was not effected in both years. STS, sucrose, and 8-HQC preservatives increased vase life in both years. A maximum total postharvest life of 8.4 days occurred with stems that received STS, 8-HQC, and 4% sucrose in 1993.

Penstemon digitalis. Storage for one week at 4C in 1993 did not significantly

effect vase life after treatment. Storage for one week at 4 or 7C in 1994 did not significantly decrease vase life after treatment (Table 2.3). Maximum total postharvest life was 15.1 days for stems stored at 4C in 1993 and 13.7 days in 1994. Penstemon stems may be stored up to three weeks at 2C without significantly decreasing vase life. Total postharvest storage duration life was 28.4 days in 1993 and 28.0 days in 1994. Vase life after treatment linearly decreased as the simulated shipping duration increased from one to five days. Preservative treatments did not affect the postharvest vase life in 1994.

Achillea filipendulina 'Coronation Gold'. Postharvest life after treatment significantly decreased when stems were stored at 2, 4, or 7C in 1993 compared to the control, while no significant decrease in vase life after treatment was observed in 1994 for stems stored at any temperature (Table 2.4). Maximum total postharvest life was 14.1 days for stems stored at 4C in 1994. Storage duration of any length significantly decreased vase life after treatment in both years. As storage time increased, the vase life linearly decreased in 1994. No data was presented for three weeks storage in 1993 due to a cooler malfunction. As simulated shipping duration increased, vase life after treatment decreased linearly in 1993 and curvilinearly in 1994. In 1993, stems treated with 4 or 8% sucrose had a significantly shorter total vase life as compared to the control.

Celosia plumosa 'Forest Fire'. Storage for one week at 2, 4, or 7C did not significantly affect celosia vase life (Table 2.5). Stems stored at 7C in 1993 had a maximum total postharvest vase life of 11.3 days in 1993 and 11.5 days in 1994. As

storage duration increased, vase life after treatment decreased linearly in 1994 and vase life after treatment was significantly less than the control for all storage durations. Total postharvest vase life was not effected by simulated shipping in both years. As the shipping duration increased from one to five days, vase life after treatment linearly decreased in both years. Celosia stems showed various responses to preservative treatments. Total vase life for stems treated with 0% sucrose, regardless of STS pretreatment, was significantly increased in 1993. Vase life after treatment significantly decreased for stems treated with 4 or 8% sucrose in 1994, regardless of STS pretreatment. A significant quadratic and linear response was observed in 1993 for sucrose treatments while a significant linear STS and sucrose interaction was observed in 1993.

Cosmos bipinnatus 'Sensation'. Data are presented for 1994 only; harvestable stems were not available in 1993. Storage for one week at 7C significantly decreased vase life after treatment while storage at 2 or 4C did not affect vase life after treatment (Table 2.6). Stems stored at 2C had a maximum total postharvest life of 12.5 days. As storage duration at 2C increased, vase life after treatment decreased linearly. Vase life responded linearly to simulated shipping duration. As shipping duration increased from two to five days, vase life after treatment significantly decreased. Preservative treatments had no significant effect on vase life.

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Buddleia davidii. Storage at 7C significantly decreased vase life after treatment in 1994, but no effect of storage temperature was observed in 1993 (Table 2.7). Stems stored at 2C had a total vase life of 10.9 days in both years. Vase life after treatment

for stems stored for three weeks was significantly less in both years than the control. Vase life decreased in 1993 and a cubic response was observed in 1994 as storage duration increased. Vase life after treatment linearly decreased in both years with a cubic response in 1994 as simulated shipping duration increased. Four days of shipping significantly decreased vase life after treatment in 1993 while all shipping durations significantly decreased vase life in 1994 compared to the control. Pretreatment with STS significantly increased vase life in 1993 compared with the control. Stems not pretreated with STS and with 8-HQC and 0% sucrose had a total vase life of 6.5 days in 1993. Vase life after treatment responded quadratically to sucrose concentration and a STS/sucrose interaction existed in 1993. Butterfly bush stems harvested at the $0 > to \le 25$ developmental stage opened significantly more flowers at the time of termination than the other stages (Figure 2.1A). The buds on cut flower stems harvested at >75 to $\le 100\%$ did not open further.

Cercis canadensis. Vase life after treatment significantly decreased for stems stored for one week at 2, 4, or 7C compared to the control in 1993 while no significant response was observed in 1994 (Table 2.8). No significant differences (P=0.05) were found between the storage temperatures in both years. Stems stored one, two, or three weeks in 1993 and stems stored for three weeks in 1994 exhibited a significantly shorter vase life after treatment as compared to the control. Stems stored for two weeks at 2C had a total vase life of 19.3 days and the vase life after treatment was not affected. Vase life after treatment decreased linearly as the simulated shipping time increased in both years. Preservatives did not effect vase life. Eastern

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redbud stems harvested at the $0 > to \le 25\%$ developmental stage opened significantly more flowers at termination than the other stages (Figure 2.1B). Stems harvested at >25 to ≤ 50 , or >50 to ≤ 75 had opened 25-50% more flowers at vase life termination. Eastern redbud stems harvested at >75 to $\le 100\%$ did not open further.

Weigela sp. Stems that were stored at 2, 4, or 7C showed a significant decrease in vase life after treatment for both years (Table 2.9). Vase life after treatment decreased linearly in both years with increasing storage duration. As simulated shipping duration increased, vase life decreased linearly in 1993 and curvilinearly in 1994. Cut flower vase life after treatment increased significantly with sucrose treatment in 1993 while no significant response to sucrose was observed in 1994. Stems receiving only 8-HQC and 0% sucrose had the greatest total vase life of 6.0 days in 1993. STS had no effect in either year.

Discussion

Weigela was the only species to respond negatively to cold storage at 2, 4, or 7C (Table 2.9). Weigela stems subjected to these temperatures exhibited visual symptoms of chilling injury. After removal from 2C, stem leaves exhibited blackening accompanied with abscission of individual flowers which could be due to cell disruption and depletion of carbohydrate reserves during storage. *Protea exima* (protea) cut flower stems exhibit leaf blackening due to depletion of leaf carbohydrate ALISHBAIND BRANKS VALDEVILLA

during storage at 20C regardless of light (Bieleski et al., 1992). However, deleterious effects of chilling temperatures on the quality of *Anthurium andraenum* 'Andre' (anthurium) flowers could not be related directly to sugar content or to respiration (Pritchard et al., 1991). Similarly, celosia stems stored beyond one week at 2C exhibited blackening of the plume one to two days after removal from the cooler (Personal observation). *Gloriosa rothschildiana* O'Brien cut flowers stored at 1C developed chilling injury with black leaves, stems, and flowers within three days (Jones and Truett, 1992).

Storage at 7C significantly decreased the vase life of butterfly bush and cosmos (Tables 2.7 and 2.6). Low storage temperatures just above freezing are thought to delay flower senescence by reducing metabolic processes and bacterial growth, making extended storage feasible (Nowak and Rudnicki, 1990; van Doorn and de Witte, 1991). Increased senescence at 7C probably did not slow metabolic processes which decrease senescence.

Penstemon, celosia, butterfly bush, eastern redbud, and weigela did not tolerate simulated shipping (Tables 2.3, 2.5, 2.7, 2.8, and 2.9). Elevated temperatures during shipping may increase respiration (Halevy and Mayak, 1981). *Protea sp.* exhibit leaf blackening and a decrease in vase life during shipping which may be attributed to postharvest inflorescence sink demand (McConchie and Lang, 1993). During the first 24 h after harvest, 82% of starch is depleted in *Protea neriifolia* (protea) inflorescence, demonstrating a strong sink strength. Cut flowers, excluding tropical species, should be cooled as rapidly as possible after harvest and then shipped at the optimal low ALIGHTNING PREVERSIVELY

temperature (Nowak and Rudnicki, 1990). Species such as penstemon, celosia, butterfly bush, and eastern redbud could be cooled to remove field heat and shipped at cooler temperatures to reach long distance markets. Also, cosmos and yarrow which can be shipped for one and two days, respectively, could possibly be precooled at optimal storage temperatures to increase shipping duration.

STS treatments extended the vase life of 'Forest Fire' celosia (Table 2.5). Pretreatment with STS has been proven to inhibit the action of ethylene in *Dianthus caryophyllus* L. 'White Sim' (carnation) (Reid et al., 1980), *Chamelaucium unicinatum* Schau. (geraldton wax flower) (Joyce, 1988), anthurium (Paull and Goo, 1982), *Antirrhinum majus* L. (snapdragon) (Nowak, 1981), and *Delphinium* sp. (delphinium) (Staby et al., 1993). Species such as penstemon, yarrow, cosmos, eastern redbud, butterfly bush, and weigela possibly did not respond to STS because ethylene may not play a role in the senescence of the flowers (Reid and Wu, 1992) or the pulsing time and temperature may not have been appropriate for the species.

Yarrow, celosia, butterfly bush, and weigela had various responses to sucrose treatments. Similarly, liatris (*Liatris spicata*) exhibited various responses to sucrose pulsing treatments which was attributed to genetic variability of the plant materials used in the study (Han, 1992). If the optimal pulsing time, temperature, light, and sucrose concentration is not used, little or no effect may be observed (Halevy and Mayak, 1981). Possibly, various responses to sucrose treatments among the cut flowers in this study were due to genetic variability, improper sucrose concentration, and improper pulsing temperature.

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Penstemon, cosmos, and eastern redbud did not respond to sucrose preservative treatments. Carbohydrates, the primary source of nutrition and energy for the flower, are necessary for maintaining biochemical and physiological processes. Cut flower stems in this study were subjected to sucrose concentrations of 2, 4 or 8% for a 20 h period. Some cut flower species required greater concentrations of sucrose to encourage floral opening and vase life extension. *Triteleia laxa* Benth. (Brodiaea) required a 20h 10% sucrose pulse (Han et al., 1990). Generally, cut flowers treated for longer periods require a lower sucrose concentration while high concentrations are used for shorter pulsing treatments (Halevy and Mayak, 1981). Han (1992) found that sucrose primarily affected the vase life of liatris through opening of flower heads and, to a lesser extent, longevity. Also, sucrose had limited influence on the longevity of liatris flower heads once they were fully developed.

Butterfly bush and eastern redbud stems harvested at $0 > to \le 25$ bud stage opened the most additional buds and reached 50-75% buds open at vase life termination (Figures 2.1A and 2.1B). Development of flowers after harvest depends upon carbohydrates and other photosynthates in plant tissues (Nowak and Rudnicki, 1990). Han (1992) found that without sucrose in the vase solution only flower heads nearly fully developed reached anthesis and opened only partially. The addition of sucrose in the vase solution was necessary for complete development and opening of all liatris flower heads. The addition of sucrose to the vase holding solution might have opened more butterfly bush and eastern redbud flowers.

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In conclusion, Echinacea purpurea (purple coneflower) was able to be stored

one week at 2 or 4C, and survived simulated shipping from one to five days. Preservatives did not effect the postharvest life. Helianthus maximilliani (Maximillian's sunflower) could be stored for one week at 2C and tolerated one day of simulated shipping. STS, 8-HQC, and sucrose preservatives extended vase life. Penstemon digitalis (penstemon) was able to be stored at 4 or 7C for one week and up to three weeks at 2C. Penstemon did not tolerate simulated shipping at 24C from one to five days. STS, sucrose, and 8-HQC preservatives did not extend the vase life. Achillea filipendulina 'Coronation Gold' (yarrow) was able to be stored at 4 or 7C for one week and up to two weeks at 2C. 'Coronation Gold' yarrow tolerated up to two days simulated shipping. STS, sucrose, and 8-HOC preservatives had inconclusive effects on the vase life. Celosia plumosa 'Forest Fire' (celosia) was able to be stored at 2, 4, or 7C for one week. Storage at 2C beyond one week decreased the vase life. 'Forest Fire' celosia did not tolerate simulated shipping at 24C for one to five days. Preservative treatments with STS may have extended the vase life. Preservative treatments that include 4 and 8% sucrose decreased the vase life. Cosmos bipinnatus 'Sensation' (cosmos) was able to be stored at 2 or 4C for one week. Storage at 2C beyond two weeks decreased vase life. Cosmos tolerated one day of simulated shipping. STS, sucrose, and 8-HQC preservatives did not extend the vase life. Buddleia davidii (butterfly bush) was able to be stored at 4C for one week and up to two weeks at 2C. Butterfly bush did not tolerate simulated shipping at 24C from one to five days. STS, sucrose, and 8-HQC preservatives had variable effects on vase life. Butterfly bush exhibited greatest percent in bud opening when

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flowers stems were harvested with $0 > to \le 25\%$ buds open. Cercis canadensis (eastern redbud) was able to be stored at 4 or 7C for one week and up to two weeks at 2C. Eastern redbud did not tolerate simulated shipping. STS, sucrose, and 8-HQC preservatives had variable effects on vase life. Eastern redbud exhibited the greatest percent in bud opening when flowers stems were harvested with $0 > to \le 25\%$ buds open. Weigela sp. (weigela) does not tolerate storage at 2, 4, or 7C for one week. Stems did not tolerate simulated shipping at 24C from one to five days. STS, sucrose, and 8-HQC preservatives may or may not have extended the vase life.

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	<u>1993</u> <u>1994</u>				
Treatment	Total (days)	PTVL (days)	Total (days)	PTVL (days)	
Control	4.6	4.6	9.1	9.1	
		Storage Temperat	ture		
2.0C	10.0 ^{*z}	3.0	12.7	5.7	
4.0C	12.1*	6.1	14.6*	7.6	
7.0C	14.0*	7.0	10.8	3.8 *	
Linear	•	*	NS	NS	
Quadratic	NS	NS	*	*	
-		Storage Duration	on		
1-week 2.0C	1 2 .9*	5.9	11.3	3.6*	
2-week 2.0C	18.7 [*]	4.7	18.1*	4 .1 [*]	
3-week 2.0C	21.3*	2.5	22.9*	1.5*	
Linear	***	NS	***	NS	
Quadratic	NS	NS	NS	NS	
		Shipping Durat	ion		
1-day 24.0C	9.3	8.0	9.2	8.2	
2 - day 24.0C	9.1	7.1	9.7	7.5	
3 - day 24.0C	7.1	4.1	8.5	5.4	
4-day 24.0C	9.9	6.7	8.7	4.7	
5-day 24.0C	9.1	4.1	11.4	6.4	
Linear	NS	NS	NS	NS	
Quadratic	NS	NS	NS	NS	
Cubic	NS	NS	NS	NS	

Table 2.1. Total postharvest life and post-treatment vase life (PTVL) in days of *Echinacea purpurea* (purple coneflower) as affected by storage temperature, storage duration, simulated shipping duration and preservatives. Significance ($P \le 0.05$) was determined by comparing each treatment to the control using Dunnett's test. Means are an average of data from ten stems.

			Preservatives		
No STS	0% Suc	13.0 [*]	11.0*	11.4	10.4
No STS	4% Suc	9.8	8.8	6.8	5.8 *
No STS	8% Suc	12.0 [•]	11.0 [•]	8.3	7.3*
STS	0% Suc	13.4*	12.4*	10.0	9.0
STS	4% Suc	13.4*	12.4*	9.7	8.7
STS	8% Suc	10.2	9.2	9.2	8.2
STS		NS	NS	NS	NS
Sucrose	(L)	NS	NS	NS	NS
Sucrose	(Q)	NS	NS	NS	NS
STSxSuc	: (L)	NS	NS	NS	NS
STSxSuc	: (Q)	NS	NS	NS	NS

^{2*}Significant at P \leq 0.05 using Dunnett's Test ^{NS}, *, ***, Nonsignificant or significant at P \leq 0.05, 0.001, respectively

	<u>199</u>	<u>93</u>	<u>19</u>	94
Treatment	Total (days)	PTVL (days)	Total (days)	PTVL (days)
Control	6.0	6.0	5.0	5.0
		Storage Temperature		
2.0C	12.1*	5.1	10.9*	3.9
4.0C	11.7*	4.7*	10.6*	3.6
7.0C	10.3 [•]	3.3*	10.9*	3.7
Linear	***	***	NS	NS
Ouadratic	NS	NS	NS	NS
		Storage Duration		
1-week 2.0C	12.2 [*]	5.2	11.0*	4.0
2-week 2.0C	17.7*	3.7 *	16.8 *	2.8*
3-week 2.0C	22.6*	1.6*	22.0 [*]	1.0*
Linear	***	***	***	***
Quadratic	NS	NS	NS	NS
		Shipping Duration		
1-day 24.0C	6.3	5.3	5.4	4.4
2-day 24.0C	6.0	4.0 [*]	5.3	3.3*
3-day 24.0C	6.1	3.1*	5.6	2.6*
4-day 24.0C	6.7	2.7*	5.3	1.3*
5-day 24.0C	7.1	2.1*	6.9	1.9*
Linear	NS	***	**	***
Ouadratic	NS	NS	*	•
Cubic	NS	NS	NS	NS

Table 2.2. Total postharvest life and post-treatment vase life (PTVL) in days of *Helianthus maximilliani* (Maximilian's sunflower) as affected by storage temperature, storage duration, shipping duration and preservatives. Significance ($P \le 0.05$) was determined by comparing each treatment to the control using Dunnett's test. Means are an average of data from ten stems.

Preservatives

No STS	0% Suc	7.9*	6.9	7.6*	6.6*	
No STS	4% Suc	8.2*	7.2*	7.1*	6.1*	
No STS	8% Suc	7.6*	6.6	7.1*	6.1*	
STS	0% Suc	8.0*	7.0*	7.7*	6.7*	
STS	4% Suc	8.4*	7.4*	7.3*	6.3*	
STS	8% Suc	7.7*	6.7	6.7*	5.7*	
STS Sucrose Sucrose	(L) (Q)	NS NS	NS NS	NS NS	NS NS	
STSxSuc	c (L) c (Q)	NS NS	NS NS	NS	NS	

^{2*}Significant at P \leq 0.05 using Dunnett's Test ^{NS}, *, **, ***, Nonsignificant or significant at P \leq 0.05, 0.01, 0.001, respectively

	<u>199</u>	<u>93</u>	<u>19</u>	<u>94</u>
	Total	PTVL	Total	PTVL
Treatment	(days)	(days)	(days)	(days)
Control	9.3	9.3	8.5	8.5
		Storage Temperatur	e	
2.0C	14.5 ^{*z}	7.5*	11.8*	4.8 *
4.0C	15.1*	8.1	13.7*	6.7
7.0C	_ ^y	-	13.5*	6.5
Linear	-	-	NS	NS
Quadratic	-	-	NS	NS
-		Storage Duration		
1-week 2.0C	13.0 [*]	6.0*	13.6*	6.6
2-week 2.0C	22.2*	8.2	21.1*	7.1
3-week 2.0C	28.4*	7.4	28.0*	7.0
Linear	***	NS	***	NS
Quadratic	NS	NS	NS	NS
		Shipping Duration		
1-day 24.0C	9.0	8.0*	7.9	6.9*
2-day 24.0C	9.2	7.2*	8.5	6.5*
3-day 24.0C	9.2	6.2*	6.5*	3.5*
4-day 24.0C	8.8	4.8*	7.3	3.3*
5-day 24.0C	8.8	3.8*	6.8*	2.0*
Linear	NS	***	*	***
Ouadratic	NS	NS	NS	NS
Cubic	NS	NS	NS	NS

Table 2.3. Total postharvest life and post-treatment vase life (PTVL) in days of *Penstemon digitalis* (penstemon) as affected by storage temperature, storage duration, shipping duration and preservatives. Significance ($P \le 0.05$) was determined by comparing each treatment to the control using Dunnett's test. Means are an average of data from ten stems.

Preservatives

11.86

No STS	0% Suc	-	-	8.7*	7.7
No STS	4% Suc	-	-	9.0 [•]	7.9
No STS	8% Suc	-	-	9.4*	7.8
STS	0% Suc	-	-	9.2	8.2
STS	4% Suc	-	-	8.6	7.6
STS	8% Suc	-	-	8.3	7.3
STS		-	-	NS	NS
Sucrose (L)	-	-	NS	NS
Sucrose (Q)	-	-	NS	NS
STSxSuc	(L)	-	-	NS	NS
STSxSuc	(Q)	-	-	NS	NS

^{z*}Significant at P \leq 0.05 using Dunnett's Test ^{NS}, *, ***, Nonsignificant or significant at P \leq 0.05, 0.001, respectively ^y Data not available

	199	93	<u>19</u>	994
Treatment	Total (days)	PTVL (days)	Total (days)	PTVL (days)
Control	10.7	10.7	8.1	8.1
		Storage Temperature		
2.0C	14.4 ^{*z}	7.4*	12.3*	6.3
4.0C	14.8 [*]	7.8*	14.1 [*]	7.1
7.0C	15.3*	8.3*	13.2*	6.8
Linear	NS	NS	NS	NS
Ouadratic	NS	NS	NS	NS
		Storage Duration		
1-week 2.0C	16.4*	6.0*	13.8*	6.8*
2-week 2.0C	19.3*	5.3*	20.1^{*}	6.1*
3-week 2.0C	_y	-	24.3*	3.3*
Linear	-	-	***	*
Quadratic	-	-	NS	NS
		Shipping Duration		
1-day 24.0C	10.6	9.6	8.3	7.5
2 - day 24.0C	8.9*	6.9	8.3	5.7
3-day 24.0C	9.1*	61	6.5	3.5*
4 - day 24.0C	8 Q*	49	8.1	4.1*
5-day 24 0C	8.6*	36	8.4	3.4*
Joury 27.00	**	**	NS	***
Quadratic	NS	NS	*	*
Cubic	NS	NS	NS	NS

Table 2.4. Total postharvest life and post-treatment vase life (PTVL) in days of Achillea filipendulina 'Coronation Gold' (yarrow) as affected by storage temperature, storage duration, shipping duration and preservatives. Significance ($P \le 0.05$) was determined by comparing each treatment to the control using Dunnett's test. Means are an average of data from ten stems.

	Preservatives				
No STS	0% Suc	10.4	9.4*	8.2	7.2
No STS	4% Suc	8.0 [*]	7.0*	8.2	6.9
No STS	8% Suc	8.4 [•]	7.4*	9.3	7.8
STS	0% Suc	10.2	9.2*	8.9	8.2
STS	4% Suc	9.6*	8.6*	8.5	7.8
STS	8% Suc	11.0	10.0*	8.7	7.9
STS		•	•	NS	NS
Sucrose	(L)	NS	NS	NS	NS
Sucrose	(Q)	NS	NS	NS	NS
STSxSuc	c (L)	NS	NS	NS	NS
STSxSuc	c (Q)	NS	NS	NS	NS

^{z*}Significant at P \leq 0.05 using Dunnett's Test ^{NS}, *, ***, Nonsignificant or significant at P \leq 0.05, 0.001, respectively ^{y-} Data not available

	199	93	<u>19</u>	994
Treatment	Total (days)	PTVL (days)	Total (days)	PTVL (days)
Control	5.6	5.6	8.8	8.8
		Storage Temper	rature	
2.0C	7.8	0.8	11.5	7.8
4.0C	10.0	3.0	10.3	3.0
7.0C	11.3	4.3	11.5	4.5
Linear	NS	NS	NS	NS
Quadratic	NS	NS	NS	NS
-		Storage Dura	ttion	
1-week 2.0C	7.0	0.0 ^{z*}	9.3	2.3*
2-week 2.0C	14.0*	0.0*	14.2 [*]	0.2*
3-week 2.0C	21.0 [*]	0.0*	21.0 [•]	0.0*
Linear	***	NS	***	•
Quadratic	NS	NS	NS	NS
		Shipping Dur	ation	
1-day 24 0C	7.2	6.2 *	6.5	5.5*
2 - day 24.0C	8.5	6.5*	7.5	5.4*
3 -day 24.00	6.0	3.0*	8.3	5.3*
A-day 24.0C	6.8	2.8*	6.6	2.6*
-4 u y 24.00	7.2	2.2*	7.2	2.3*
I inear	NS	**	NS	**
Quadratic	NS	NS	NS	NS
Cubic	NS	NS	NS	NS

Table 2.5. Total postharvest life and post-treatment vase life (PTVL) in days of *Celosia plumosa* 'Forest Fire' (celosia) as affected by storage temperature, storage duration, shipping duration and preservatives. Significance ($P \le 0.05$) was determined by comparing each treatment to the control using Dunnett's test. Means are an average of data from ten stems.

			Preserva	tives	
No STS No STS No STS	0% Suc 4% Suc 8% Suc	7.8* 5.9 5.8*	6.8 4.9 4.8	8.6 6.7 6.5	7.6 5.7* 5.5*
STS STS STS	0% Suc 4% Suc 8% Suc	8.2* 6.8 5.9	7.2 5.8 4.9	9.2 8.3 7.8	8.2 7.3 6.8
STS Sucrose Sucrose STSxSuc	(L) (Q) :: (L)	NS ····	NS 	NS NS NS	NS NS NS
STSXSuc) (V)	NS	NS	NS	NS

^{z*}Significant at P \leq 0.05 using Dunnett's Test ^{NS}, *, **, ***, Nonsignificant or significant at P \leq 0.05, 0.01, 0.001, respectively

Table 2.6. Total postharvest life and post-treatment vase life (PTVL) in days of Cosmos bipinnatus 'Sensation' (cosmos) as affected by storage temperature, storage duration, shipping duration and preservatives. Significane ($P \le 0.05$) was determined by comparing each treatment to the control using Dunnett's test. Means are an average of data from ten stems.

<u>1994</u>					
	Total	PTVI.			
Treatment	(days)	(days)			
	(,)-)	(;)/			
Control	6.2	6.2			
	Storage T	emperature			
2.0C	12.5 ^{*z}	5.5			
4.0C	12.0 [*]	4.8			
7.0C	10.1*	3.1*			
Linear	**	**			
Quadratic	NS	NS			
	Storage	Duration			
1-week 2.0C	11.8*	4.2			
2-week 2.0C	16.1*	2.7*			
3-week 2.0C	21.7*	0.7*			
Linear	***	***			
Quadratic	NS	NS			
	Shipping	g Duration			
1-day 24.0C	6.9	5.9			
2-day 24.0C	5.3	3.4*			
3-day 24.0C	7.0	4.1*			
4-day 24.0C	6.3	2.3*			
5-day 24.0C	6.8	1.3*			
Linear	NS	***			
Quadratic	NS	NS			
Cubic	NS	NS			

Preservatives

No STS0%SucNo STS4%SucNo STS8%Suc	6.9 7.1 7.5	5.8 6.1 6.5	
STS 0% Suc STS 4% Suc STS 8% Suc	7.8 7.5 7.9	6.3 6.5 6.9	
STS Sucrose (L) Sucrose (Q) STSxSuc (L) STSxSuc (Q)	NS NS NS NS	NS NS NS NS NS	

^{2*}Significant at P \leq 0.05 using Dunnett's Test ^{NS}, ^{**}, ^{***}, Nonsignificant or significant at P \leq 0.01, 0.001, respectively

	<u>199</u>	<u>93</u>	<u>19</u>	<u>1994</u>		
Treatment	Total (days)	PTVL (days)	Total (days)	PTVL (days)		
Control	3.8	3.8	4.9	4.9		
		Storage Temperatu	re			
2.0C	10.9* ^z	3.9	10.9*	3.9		
4.0C	10.3 [*]	3.3	10.8*	3.8		
7.0C	9.6*	2.6	10.2*	3.2*		
Linear	NS	NS	NS	NS		
Quadratic	NS	NS	NS	NS		
-		Storage Duration	2			
1-week 2.0C	11.0 [*]	4.0	11.5*	4.3		
2-week 2.0C	17.5*	3.5	19.3*	5.1		
3-week 2.0C	21.0*	0.0*	23.0 [*]	2.0*		
Linear	***	***	***	**		
Quadratic	***	NS	**	**		
		Shipping Duration	n			
1-day 24.0C	4.4	3.4	4.5	3.5*		
2-day 24.0C	5.2	3.2	4.0 *	2.0*		
3-day 24.0C	4.6	1.6	4.4	1. 7 *		
4-day 24.0C	5.2	1.2*	5.2	1.9*		
5-day 24.0C	7.0	2.0	5.0	0.0*		
Linear	**	*	**	***		
Quadratic	NS	NS	NS	NS		
Cubic	NS	NS	•	**		

Table 2.7. Total postharvest life and post-treatment vase life (PTVL) in days of *Buddleia davidii* (butterfly bush) as affected by storage temperature, storage duration, shipping duration and preservatives. Significance ($P \le 0.05$) was determined by comparing each treatment to the control using Dunnett's test. Means are an average of data from ten stems.

			Preservatives		
No STS	0% Suc	6.5*	5.5*	5.6	4.6
No STS	4% Suc	6.4*	5.4*	5.4	4.4
No STS	8% Suc	6.3 *	5.3*	5.7*	4.7
STS	0% Suc	5 9*	4 9*	6.0*	50
STS	4% Suc	6.3*	5.3*	5.8*	4.8
STS	8% Suc	5.4*	4.4	5.8*	4.8
STS		**	**	NS	NS
Sucrose	(L)	NS	NS	NS	NS
Sucrose	(Q)	NS	***	NS	NS
STSxSuc	: (L)	NS	NS	NS	NS
STSxSuc	c (Q)	NS	***	NS	NS

^{2*}Significant at P \leq 0.05 using Dunnett's Test ^{NS}, *, **, ***, Nonsignificant or significant at P \leq 0.05, 0.01, 0.001, respectively

Table 2.8. Total postharvest life and post-treatment vase life (PTVL) in days of *Cercis canadensis* (eastern redbud) as affected by storage temperature, storage duration, shipping duration and preservatives. Significance ($P \le 0.05$) was determined by comparing each treatment to the control using Dunnett's test. Means are an average of data from ten stems.

<u>1993</u>			<u>19</u>	<u>1994</u>		
Treatment	Total (days)	PTVL (days)	Total (days)	PTVL (days)		
Control	9.0	9.0	5.6	5.6		
		Storage Tem	perature			
2.0C	13.4 ^{*z}	6.4*	12.0	5.0		
4.0C	13.2 [•]	6.2 [*]	12.7	5.7		
7.0C	13.4 *	6.4 [*]	12.5	5.5		
Linear	NS	NS	NS	NS		
Ouadratic	NS	NS	NS	NS		
		Storage Du	uration			
1-week 2.0C	13.8	6.8 *	12.5*	5.5		
2-week 2.0C	20.4*	6.4*	19.3*	5.3		
3-week 2.0C	19.4 *	2.0*	25.0*	4.0 [•]		
Linear	**	***	***	**		
Quadratic	**	**	NS	NS		
		Shipping D	uration			
1-day 24.0C	8.6	7.8*	4.6	3.6*		
2-day $24.0C$	8.2	6.2*	5.6	3.6*		
3-day 24.0C	7.8*	4.8*	4.7	1.7*		
4 - day 24.0C	7.3*	3.3*	5.9	1.9*		
5-day 24.0C	7.2*	2.2*	5.4	0.4*		
I inear	**	**	NS	***		
Quadratic	NS	NS	NS	NS		
Cubio	NS	NS	NS	NS		
Cubic	C M T	UT D	143	110		

Preservatives

No STS No STS No STS	0% Suc 4% Suc 8% Suc	9.9 9.4 10.3	8.9 8.4 9.3	6.1 5.6 5.8	5.1 4.6 4.8	
STS	0% Suc	10.1	9.1	5.7	4.7	
STS	4% Suc	9.4	8.4	6.2	5.2	
STS	8% Suc	9.8	8.8	6.1	5.1	
STS		NS	NS	NS	NS	
Sucrose	(L)	NS	NS	NS	NS	
Sucrose	(Q)	NS	NS	NS	NS	
STSxSuc	: (L)	NS	NS	NS	NS	
STSxSuc	c (Q)	NS	NS	NS	NS	

^{z*}Significant at P \leq 0.05 using Dunnett's Test ^{NS}, ^{**}, ^{***}, Nonsignificant or significant at P \leq 0.01, 0.001, respectively

Table 2.9. Total postharvest life and post-treatment vase life (PTVL) in days of *Weigela* sp. (weigela) as affected by storage temperature, storage duration, shipping duration and preservatives. Significance ($P \le 0.05$) was determined by comparing each treatment to the control using Dunnett's test. Means are an average of data from ten stems.

<u>1993</u>				<u>1994</u>		
Treatment	Total (days)	PTVL (days)		Total (days)	PTVL (days)	
Control	3.0	3.0		5.0	5.0	
		Storage T	<i>Semperature</i>			
2.0C	9.0 ^{*z}	2.0 [•]		8.8 [•]	1.8°	
4.0C	8.9 ⁻	1.9		9.1	2.1	
7.0C	8.6	1.6		8.7	1.7	
Linear	NS	NS		NS	NS	
Quadratic	NS	NS	~ ·	NS	NS	
		Storage	Duration			
1-week 2.0C	9.4 *	2.5		8.8*	1.8*	
2-week 2.0C	15.6*	1.6*		15.1 *	1.1*	
3-week 2.0C	21.9 *	0.9*		21.0 [*]	0.1*	
Linear	***	***		***	***	
Quadratic	NS	NS		NS	NS	
Shipping Duration						
1-day 24.0C	2.7	1.7*		4.3	3.3*	
2-day 24.0C	3.6	1.6 *		4.3	2.3*	
3-day 24.0C	3.4	0.6*		3.9	0.9*	
4-day 24.0C	3.4	0.4*		4.2	0.2*	
5-day 24.0C	3.3	0.1*		5.1	0.1*	
Linear	*	***		NS	***	
Ouadratic	**	NS		NS	**	
Cubic	NS	NS		NS	NS	

No STS	0% Suc	6.0 [*]	5.0 [*]	4.8	3.8
No STS	4% Suc	4.3*	3.3	5.3	4.3
No STS	8% Suc	5.0*	4.0 [*]	5.4	4.4
STS	0% Suc	5.4*	4.4*	5.4	4.4
STS	4% Suc	4 .9 [*]	3.9*	6.0	5.0
STS	8% Suc	4.6*	3.6	5.6	4.6
STS		NS	NS	*	NS
Sucrose	(L)	**	**	NS	NS
Sucrose	(Q)	**	**	NS	NS
STSxSuc	c (L)	**	**	NS	NS
STSxSuc	c (Q)	**	**	NS	NS

^{z*}Significant at P \leq 0.05 using Dunnett's Test ^{NS}, *, **, ***, Nonsignificant or significant at P \leq 0.05, 0.01, 0.001, respectively

Figure 2.1 Effect of developmental stage on final number of open flowers at vase life termination for eastern redbud (A) and butterfly bush (B). Developmental stages were determined as bud opening percentage: (1) 0 > to ≤ 25%, (2) >25 to ≤ 50%, (3) >50 to ≤ 75%, and (4) >75 to ≤ 100% of buds open.

Figure 2.1



Chapter III

Postharvest Ethylene Production and Sensitivity of Eight Specialty Cut Flower Species

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<u>Additional index words</u>. Achillea filipendulina 'Coronation Gold', Buddleia davidii, Celosia plumosa 'Forest Fire', Cercis canadensis, Cosmos bipinnatus 'Sensation', Echinacea purpurea, Helianthus maximilliani, Penstemon digitalis, Weigela species.

Abbreviations. AVG, aminoethoxyvinylglycine; AOA, aminooxyacetic acid; STS, silver thiosulfate.

Abstract. Ethylene and CO_2 evolution was measured in headspace gases at 0, 12, 24, and 48 h after harvest and the effects of exogenous applications of 0.0, 0.2, or 1.0 ul·liter⁻¹ ethylene for 20 h on vase life was observed in eight specialty cut flower species.

Helianthus maximilliani (Maximillian's sunflower), Penstemon digitalis (penstemon), Achillea filipendulina 'Coronation Gold' (yarrow), Celosia plumosa 'Forest Fire', Cosmos bippinatus 'Sensation', Buddleia davidii (butterfly bush), and Weigela sp. (weigela) produced a small ethylene peak attributed to wound ethylene followed by a steady rise in CO_2 . Buddleia davidii (butterfly bush) and Weigela sp. (weigela) exhibited a climacteric-like pattern of ethylene evolution followed by a steady rise in CO_2 production. Echinacea purpurea (purple coneflower) ethylene biosynthesis was not significant during the 48 h period after harvest. Vase life of purple coneflower, yarrow, celosia, cosmos, and butterfly bush was not affected by exogenous ethylene. Exogenous ethylene application to Maximillian's sunflower, penstemon and weigela resulted in flower abscission and decreased vase life indicating that Maximillian's sunflower, penstemon and weigela were ethylene-sensitive cut flower species.

Introduction

Ethylene and CO_2 biosynthesis have been observed on numerous cut flower species such as *Gladiolus* sp. (gladiolus) (Serek et al., 1994), *Dianthus caryophyllus* 'Elliot's White' (carnation) (Altman and Solomos, 1994), and *Lathyrus odoratus* (sweet pea) (Mor et al., 1984). Tingley and Prince (1990) characterized ethylene production and sensitivity of 16 evergreen species. Jones and Truett (1992) observed that ethylene within sealed bags containing *Gloriosa rothschildiana* O'Brien (gloriosa lily) increased from 0.02 to 0.3 ug·liter⁻¹. Trace amounts of ethylene are produced by *Cymbidium* flowers when pollinia are removed, causing blushing of the labellum (anthocyanin accumulation) and initiating general senescence of the flower (Reid and Wu, 1992).

The biosynthetic pathway of ethylene synthesis, methionine \rightarrow S-adneosyl methionine \rightarrow 1-aminocyclopropane-1-carboxylic acid (ACC) \rightarrow ethylene, has been determined (Yang, 1980). Conversion of ACC into ethylene is inhibited by anaerobiosis, elevated CO₂ concentrations, and temperatures greater than 35 C (Yang, 1980). Wounding of plant tissue induces the synthesis of ACC synthase, leading to ACC accumulation and ethylene production.

Cut flower species vary in sensitivity to ethylene. *Alstroemeria* sp. (alstroemeria), carnation, and *Delphinium* hybrids (delphinium) are sensitive while *Anthurium andreanum* (anthurium) and *Tulipa* sp. (tulip) are not sensitive to ethylene (Nowak and Rudnicki, 1990). Placing flowers in an atmosphere enriched with ethylene accelerates the autocatalytic production of ethylene in wilted petals, growing ovaries and whole flowers. Autocatalytic ethylene production develops gradually with advancement of senescence (Nowak and Rudnicki, 1990). Cut flower species that produce ethylene may effect the longevity of other ethylene-sensitive species during shipping or storage. The objective of this research was to determine postharvest ethylene evolution and sensitivity of eight cut flower species.

Cut flower stems of Echinacea purpurea (L) Moensch (coneflower), Helianthus maximilliani Schrad. (Maximillian's sunflower), Penstemon digitalis Nutt. (penstemon), Cosmos bipinnatus Cav. Ann. 'Sensation' (cosmos), Celosia plumosa L. 'Forest Fire' (celosia), Achillea filipendulina Lam. 'Coronation Gold' (yarrow), Buddleia davidii Franch. (butterfly bush), and Weigela sp. Thunb. (weigela) were grown and harvested from field plots at the OSU Nursery Research Station (Stillwater, Okla). Maximillian's sunflower, and cosmos were harvested when the first ring of disk florets was fully open. Penstemon, celosia, weigela, and butterfly bush were harvested when approximately 50% of the flowers were open. Yarrow flowers were harvested when 100% of florets were open. Additional, untreated coneflower cut flower stems were purchased and shipped overnight from a commercial supplier (Dos Osos Multiflora, Watsonville, Calif).

Cut flower stems were harvested no later than 11:00 a.m. and were re-cut to 20.0 cm in length for all species. After recutting, one stem of each species was placed in a 150-ml glass beaker containing 100-ml deionized (DI) water and sealed in a 3-liter glass jar equipped with flushing and sampling ports. Jars were flushed with ethylene-free air for approximately 3 min. before being sealed. Jars were held at $21 \pm 3C$ for 48 h under continuous light exposure. Two 1-ml head-space gas samples were taken from each jar after 1, 12, 24, and 48 h for CO₂ and ethylene determination. CO₂ analysis was conducted by injecting 1-ml head-space gas into an infrared gas analyzer (IRGA)(Horiba PIR-2000) connected to a chart recorder. Lamp grade nitrogen was used as the carrier

gas. A CO₂ response curve was linearly established by running 2045 and 5000 ul-liter⁻¹ CO₂ standards (Scott Specialty Gases, Plumsteadville, Pa). Ethylene analysis was conducted using a gas-tight syringe to inject 1-ml head-space gas into a gas chromatograph (GC) (model 540, Tracor Instruments, Austin, Texas). The GC was equipped with a 30-m x 0.53-mm column (GS-Q Megabore, J & W Scientific, Folsom, Calif.) and flame ionization detector. Helium carrier gas flowed at 93 cm·s⁻¹. An isothermal run was conducted to determine ethylene gas concentration with an oven temperature of 60C. Injector and detector temperatures were 180 and 200 C, respectively. Mean production rates were calculated from response factors derived from gaseous ethylene standards (Scott Specialty Gases, Plumsteadville, Pa). The experimental design was a completely randomized design with one to three replicates of ten stems blocked by harvest date and randomly harvested from different plants.

The response of coneflower, Maximillian's sunflower, penstemon, cosmos, celosia, yarrow, butterfly bush, and weigela to exogenous applications of ethylene was observed in a flow-through system. After harvest from the field, one cut flower stem was sealed in a 3-liter glass jar and subjected to exogenous ethylene concentrations of 0.0, 0.2, or 1.0 ul liter⁻¹ at a flow rate of approximately 30 liter h⁻¹ for 20 h. Flow-through ethylene gas concentrations were achieved by ten-fold dilution of 2.0 and 10.0 ul liter⁻¹ ethylene standards (Scott Specialty Gases, Plumsteadville, Pa). Controls of 0.0 ul liter⁻¹ contained approximately 50g of Ethysorb (StayFresh Ltd, London, England) in the bottom of the 3-liter glass container to oxidize ethylene produced by the cut stems. Stems were subjected to exogenous ethylene under continuous light exposure at 21.0 ± 3 C. After

treatment, each stem was removed from the sealed container and held at 21 ± 3 C under continuous light exposure. The response of the cut flower stems to exogenous applications of ethylene was observed as the vase life in days after harvest. The vase life of the cut flower stems was terminated when the flowers visually exhibited 50% or greater senescence symptoms. Penstemon and weigela were terminated when flowers exhibited 50% or greater in-rolling and flower abscission. Purple coneflower was terminated when flowers exhibited 50% or greater wilting and loss of color. Yarrow, celosia, Maximillian's sunflower, cosmos, and butterfly bush were terminated when the flowers exhibited 50% or greater wilting. The experimental design was a completely randomized design with one to three replicates of ten stems per treatment blocked by harvest date and randomly harvested from different plants for each treatment. One replicate was harvested of Echinacea purpurea (purple coneflower), Helianthus maximilliani (Maximillian's sunflower), Penstemon digitalis (penstemon), and Weigela sp. (weigela). Three replicates were harvest of Achillea filipendulina 'Coronation Gold' (yarrow), Celosia plumosa 'Forest Fire' (celosia), Cosmos bipinnatus 'Sensation', and Buddleia davidii (butterfly bush).

Results

Echinacea purpurea. The rate of ethylene production from purple coneflower did not vary during the 48 h accumulation period (Table 3.1, Fig. 3.1A). The 1 h accumulation of CO_2 was significantly lower than at 12, 24 and 48. Vase life of purple coneflower was not affected by exposure to exogenous ethylene (Table 3.2).

Helianthus maximilliani. Rate of ethylene production varied during the 48 h accumulation period (Table 3.1, Fig. 3.1B). The 1 h accumulation of ethylene and CO_2 was significantly lower than at 12, 24, and 48 h. Vase life of Maximillian's sunflower decreased significantly when exposed to 1.0 ul·liter⁻¹ ethylene (Table 3.2).

Penstemon digitalis. Rate of ethylene produced did not vary during the 48 h accumulation period (Table 3.1, Fig. 3.1C). CO_2 accumulation increased from 1 h to 24 h and then remained steady. Vase life of penstemon decreased significantly when exposed to 1.0 ul·liter⁻¹ ethylene for 20 hrs compared to the control treated with 0.0 ul·liter⁻¹ ethylene (Table 3.2).

Achillea filipendulina 'Coronation Gold'. Rate of ethylene produced did not vary during the 48 h accumulation period (Table 3.1, Fig. 3.1D). The 1 h accumulation of CO_2 was significantly lower than at 12, 24, and 48 hrs. Means CO_2 production rate was significantly different from 1 to 12 hrs and 48 h was significantly different from 1, 12, and 24 h. CO_2 production rates were not different between 12 and 24 hrs. Vase life of yarrow was not affected by exposure to exogenous ethylene (Table 3.2).

Celosia plumosa 'Forest Fire'. Rate of ethylene production did not vary during the 48 h accumulation period (Table 3.1, Fig. 3.1E). The 1 h accumulation of CO_2 was significantly lower than at 12, 24, and 48 h. CO_2 production peaked at 12 hrs and slowly decreased to hour 48. Vase life of celosia was not affected by exposure to exogenous applications of ethylene (Table 3.2). Cosmos bipinnatus 'Sensation'. Rate of ethylene production did not vary during the 48 h period (Table 3.1, Fig. 3.1F). The 12, 24, and 48 hr accumulation of CO_2 was significantly higher than at 1 h of accumulation. Vase life of cosmos was not affected by exposure to exogenous applications of ethylene (Table 3.2).

Buddleia davidii. The 1 h accumulation of ethylene was significantly higher than at 48 h (Table 3.1, Fig. 3.1G). Ethylene production was greatest at 1 h and declined. The 1 h accumulation of CO_2 was significantly lower than at 12, 24, and 48 h (Table 3.1, Fig. 3.1G). CO_2 production peaked at 24 h and declined. Vase life of butterfly bush was not affected by exposure to exogenous applications of ethylene (Table 3.2).

Weigela species. The rate of ethylene produced during the 48 hr accumulation period was significantly different (Table 3.1, Fig. 3.1H). Ethylene production was greatest at 1 h and declined. The 1 h accumulation of CO_2 was significantly lower than at 12, 24, and 48 h. Vase life of weigela significantly decreased when exposed to 0.2 and 1.0 ul·liter⁻¹ ethylene for 20 h compared to the control treated with 0.0 ul·liter⁻¹ ethylene (Table 3.2).

Discussion

Ethylene production from cut celosia, cosmos, buddleia, and weigela stems was higher one hour after harvest than at 12, 24, and 48 h accumulation periods and was followed by a rise in CO_2 by hour 12 after harvest. Maximillian's sunflower, penstemon and yarrow demonstrated an ethylene peak at 12 h with a concomitant rise in CO_2 followed by a decrease in ethylene and CO_2 production after 12 h. At the time of the rise in CO₂ ethylene production decreased, which is in agreement with previous findings that elevated levels of CO₂ inhibit ethylene production (Yang, 1980). Comparing the time course of ethylene production to the vase life of celosia (7.2 days), cosmos (6.2 days), Maximillian's sunflower (6.0 days), penstemon (8.9 days) and varrow (9.7 days) when held in DI water (Redman and Dole, unpublished data), indicates that the initial ethylene produced may be wound ethylene and not climacteric ethylene. Upon cutting or bruising young Ipomea transcendentia cut flower stems, ethylene production rose then decreased without development of typical senescence symptoms (Halevy and Mayak, 1981). Mechanical wounding such as cutting or abrasion causes orange, banana, tomato, apple and other fruit tissues to produce large amounts of ethylene which in turn accelerates ripening and may cause loss of fruit quality during storage (Yu and Yang, 1980). Standard 'White Sim' carnation flowers typically reach a peak in ethylene production six days after harvest when senescence symptoms are visible followed by a concomitant rise in CO_2 (Whitehead et al., 1984). The CO_2 and ethylene biosynthesis measurement period for celosia, cosmos, Maximillian's sunflower, penstemon, and yarrow may not have been long enough to identify ethylene production associated with senescence. After 48 h, celosia, cosmos, penstemon, and yarrow exhibited no visual senescence symptoms.

Comparing the time course of ethylene and CO_2 production with the vase life of the relatively short-lived butterfly bush (4.4 days) and weigela (3.5 days) (Redman and Dole, unpublished data), the pattern of ethylene and CO_2 biosynthesis resemble the climacteric production of ethylene and CO_2 found in carnation (Brandt and Woodson, 1992). Carnation flowers begin to exhibit a peak in ethylene production at the onset of visual senescence symptoms which is characterized by an in-rolling or sleepiness of the outer-whorl of petals (Reid and Wu, 1992). Ethylene production from *Lathyrus odoratus* L. (sweet pea) followed a typical climacteric pattern 80 h after harvest with flowers visibly wilting and abscising one day after the peak (19 nl/flower/h) (Mor et al., 1984). Both butterfly bush and weigela flowers had begun to abscise at the time of the 48 h head-space gas sample.

Applications of three levels of exogenous ethylene for a 20 h period had no affect on the vase life of purple coneflower, yarrow, celosia, and cosmos. Ethylene may not play a role in reducing vase life of these flowers. Butterfly bush exhibited a climactericlike peak in ethylene production followed by a rise in respiration, but was insensitive to exogenous ethylene. *Gladiolus sp.* (gladiolus) also exhibit a modest climacteric-like peak in ethylene production, but individual florets were insensitive to exogenous ethylene (Serek et al., 1994). Serek et al., (1994) concluded that gladiolus was an ethyleneinsensitive flower and ethylene was not a factor in floret senescence.

Exogenous applications of 0.2 or 1.0 ul·liter⁻¹ ethylene decreased the vase life of penstemon. The senescence symptoms of weigela and penstemon upon 20 h exposure to these ethylene concentrations were flower in-rolling and abscission. Species sensitive to exogenous ethylene typically show petal abscission as the initial senescence symptom (Reid and Wu, 1992). Ethylene is probably naturally involved in development of senescence symptoms of species sensitive to low concentrations of ethylene (Reid and Wu, 1992). During the 48 h biosynthesis study, penstemon did not exhibit a climacteric-like peak of ethylene
production. Considering that penstemon vase life was decreased by exogenous ethylene, increased levels of ethylene may have been detected if the biosynthesis study extended for the duration of the penstemon vase life. In previous studies, weigela and penstemon did not respond to pretreatment with STS (Redman and Dole, unpublished data). However, Staby et al., (1993) found that STS reduced or completely inhibited the abscission of flowers from *Penstemon hartwegii* x *P. cobaea* 'Firebird' (beard's tongue) indicating that some penstemon species may be ethylene sensitive species.

In conclusion, purple coneflower, celosia, cosmos, and yarrow were ethyleneinsensitive cut flower species. Maximillians's sunflower did not produce a climactericlike pattern of ethylene and CO, production and was sensitive to exogenous ethylene. Even though butterfly bush exhibited a modest climacteric-like pattern of ethylene and CO₂ production, it was insensitive to exogenous applications of ethylene and was probably an ethylene-insensitive species also. Weigela exhibited an climacteric-like pattern of ethylene and CO₂ production and was sensitive to exogenous ethylene which accelerated flower abscission. Penstemon did not exhibit a climacteric pattern of ethylene and CO₂ production, but was sensitive to exogenous ethylene and accelerated flower abscission. Maximillian's sunflower, penstemon and weigela were considered ethylene-sensitive cut flower species. However, to determine the true nature of ethylene biosynthesis and sensitivity, further research on ACC synthase activity, and ACC content and the effects of ethylene inhibitors such as aminoethoxyvinylglycine (AVG), aminooxyacetic acid (AOA), and STS on flowers would be required. Postharvest recommendations for Maximillian's sunflower, penstemon and weigela would include treatment with an inhibitor of ethylene biosynthesis and action, plus avoiding shipping and storage near flowers or produce that may produce ethylene.

Contraction of the

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Source of Variation	df	Mean squares	
	Echinacea p	ourpurea	
Stem C ₄ H	9	0.007 ^{NS}	
Hour CH	3	0.027*	
Stem CO ₂	9	2.87×10^{6NS}	
Hour CO ₂	3	4.39x10 ^{7***}	
	Helianthus m	aximilliani	
Stem C ₂ H	9	0.018 ^{NS}	
Hour C.H.	3	0.032*	
Rep x Hr C_2H_4	3	0.001 ^{NS}	
Stem CO ₂	9	2.21x10 ^{8NS}	
Hour CO ₂	3	3.73x10 ^{8NS}	
Rep x Hr CO ₂	3	1.75x10 ^{8NS}	
	Penstemon	digitalis	
Stem C ₂ H₄	9	1.29 ^{NS}	
Hour C,H ₄	3	1.40 ^{NS}	
Stem CO ₂	9	5.72x10 ^{6***}	
Hour CO ₂	3	$2.20 \times 10^{7***}$	
Ac	hillea filipendulina	'Coronation Gold'	
Rep C.H.	1	0.017 ^{NS}	
Stem C _H	9	3.239 ^{NS}	
Hour C.H.	3	3.252 ^{NS}	
Rep x Hr C_2H_4	3	1.497 ^{NS}	
Ren CO.	1	7.08x10 ^{6***}	
Stem CO.	9	3.08x10 ^{6***}	
\mathcal{O}_2	-	• • • •	

Table 3.1. Analysis of variance for 1-ml ethylene and CO_2 head-space gas samples taken at 1, 12, 24, and 48 h after harvest. Means are an average of data from 10-30 stems/species.

Hour CO ₂	3	2.93x10 ^{7***}
Rep x Hr CO_2	3	5.35x10 ^{4NS}

Celosia plumosa 'Forest Fire'

Rep C_2H_4	2	33.53 ^{NS}
Stem C_2H_4	9	10.99 ^{NS}
Hour C_2H_4	3	31.53 ^{NS}
Rep x Hr C_2H_4	6	8.9 ^{NS}
Rep CO ₂	2	2.41x10 ^{8***}
Stem CO ₂	9	2.14x10 ^{7***}
Hour CO ₂	3	2.38x10 ^{8***}
Rep x Hr CO ₂	6	$2.32 \times 10^{4***}$

Cosmos bippinatus 'Sensation'

Rep C_2H_4	2	25.92 ^{NS}
Stem C_2H_4	9	18.71 ^{NS}
Hour C_2H_4	3	11.43 ^{NS}
Rep x Hr C_2H_4	6	14.17 ^{NS}
D 00	•	1 00-106***
$\operatorname{Rep} \operatorname{CO}_2$	2	1.02X10 ²
Stem CO_2	2 9	$3.24 \times 10^{5***}$
$\begin{array}{c} \text{Rep } CO_2 \\ \text{Stem } CO_2 \\ \text{Hour } CO_2 \end{array}$	2 9 3	3.24x10 ⁵ *** 8.18x10 ^{6***}

Buddleia davidii

Rep C_2H_4	2	10.81 ^{NS}
Stem C_2H_4	9	7.63 ^{NS}
Hour C_2H_4	3	15.89 ^{NS}
Rep x Hr C_2H_4	6	5.58 ^{NS}
Rep CO ₂	2	1.48x10 ^{7NS}
Stem CO ₂	9	4.96x10 ^{6NS}
Hour CO ₂	3	6.71x10 ^{7****}
Rep x Hr CO ₂	6	7.49×10^{6NS}

Weigela sp.

Rep C ₂ H ₄	2	46.87 ^{NS}	
Stem C_2H_4	9	29.77 ^{NS}	
Hour C_2H_4	3	80.20 ^{NS}	
Rep x Hr C_2H_4	6	10.67 ^{NS}	
Rep CO ₂	2	9.44x10 ^{7***}	
Stem CO ₂	9	3.31x10 ^{6**}	
Hour CO ₂	3	8.61x10 ^{7***}	
Rep x Hr CO ₂	6	9.27x10 ^{6***}	

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

Stem Number	Treatment (ul·liter ⁻¹)	Vase life (days)
	Echinacea p	urpurea
10	0.0	3.1 <u>+</u> 0.7
10 10	0.2 1.0	2.6 ± 1.1 1.1 ± 1.3
	Treament	NS
	Helianthus mo	ıximilliani
10	0.0	5.4 <u>+</u> 0.8
10 10	0.2 1.0	5.5 ± 0.7 $4.5^{*z} \pm 1.3$
	Treatment	NS
	Penstemon	digitalis
10	0.0	6.5 <u>+</u> 2.5
10 10	0.2 1.0	4.6 <u>+</u> 2.4 4.6* <u>+</u> 1.0
	Treatment	•
	Achillea filipendulina	'Coronation Gold'
20	0.0	8.4 <u>+</u> 2.4
20	0.2	7.9 ± 2.4
20	1.0	7.7 <u>+</u> 2.8
	Treatment Rep	NS NS

Table 3.2. The effect of 0.0, 0.2, and 1.0 ul·liter⁻¹ exogenous ethylene applied for 20 h at 30-liters h^{-1} on vase life (days) of cut flowers. Means are an average of data from 10-30 stems/species <u>+</u> SE.

Celosia plumosa 'Forest Fire'

30	0.0	5.3 + 2.5
30	0.2	4.6 + 1.8
30	1.0	4.5 <u>+</u> 1.6
	Treatment	NS
	Rep	NS
	Treatment x Rep	NS

Cosmos bipinnatus 'Sensation'

30 30 30	0.0 0.2 1.0	6.3 ± 2.2 6.3 ± 2.1 5.9 ± 1.8
	Treatment	NS
	Rep	NS
	Treatment x Rep	NS

Buddleia davidii

30	0.0	3.1 ± 1.0
30	0.2	3.1 + 1.2
30	1.0	3.1 ± 1.4

Treatment	NS
Rep	NS
Treatment x Rep	NS

NS

	Weigela	sp.	
30	0.0	3.9 <u>+</u> 1.2	
30	0.2	2.2^{+} 0.6	
30	1.0	1.2° <u>+</u> 0.5	
- <u></u>	Treatment	***	

^{z*} Significant using Dunnett's t-tests ^{NS}, ^{*}, ^{***} Nonsignificant or significant at P<0.05 or 0.001, respectively.

Figure 3.1. Ethylene (-•-) (nmol ethylene/liter/hr) and CO₂ (-• -) (umol CO₂/liter/hr) production as measured with 1-ml ethylene and CO2 head-space gas samples taken at 1, 12, 24, and 48 h after harvest for *Echinacea purpurea* (A), *Helianthus maximilliani* (B), *Penstemon digitalis* (C), Achillea filipendulina 'Coronation Gold' (D), Celosia plumosa 'Forest Fire' (E), Cosmos bipinnatus 'Sensation' (F), Buddleia davidii (G), and Weigela species (H). Means are an average of data from 10-30 stems. Bars represent ± SE.







Chapter IV

Comparison of Ethanol as a Floral Preservative with Silver Thiosulfate and 8-Hydroxyquinoline Citrate

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Additional index words. Dianthus caryophyllus 'Atlantis', D. caryophyllus 'White Sim', Rosa hybrida, R. hybrida 'Better Times', silver thiosulfate (STS), 8-hydroxyquinoline citrate (8-HQC).

Abbreviations. ACC, 1-amino-cyclopropane-1-carboxylic acid; AA, acetaldehyde; ADH, alcohol dehydrogenase; CFU, colony forming unit; EFE, ethylene forming enzyme; 8-HQC, 8-hydroxyquinoline citrate; NA, nutrient agar; NB, nutrient broth; SDW, sterile deionized water; STS, silver thiosulfate.

Abstract. The postharvest evolution of ethylene and CO₂ measured from headspace gas samples was not different in Dianthus caryophyllus 'Atlantis' cut flower stems continually treated with 8% ethanol or silver thiosulfate (STS). The respiration peak was suppressed in ethanol and STS treated stems. Ethanol and STS treated stems did not provide protection from the action of ethylene when exposed to 1.0 ul·liter⁻¹ ethylene. Exposure to 0.0 or 0.2 ul·liter⁻¹ ethylene did not affect the vase life of ethanol or STS treated stems. STS significantly increased the vase life of carnation flowers as compared to ethanol treated stems. Flowers continually treated in 8% ethanol exhibited the greatest number of bacterial colony forming units (cfu) in the vase solution but had the longest postharvest life which was attributed to the inhibitory effect upon ethylene action. Five bacteria isolates were identified as carnation isolate (CI) 10, CI 11, CI 12, CI 13 and CI 14. The isolates CI 11, CI 12, and CI 13 decreased the vase life of 'Atlantis' carnations. The isolates CI 10 and CI 14 did not affect the vase life of 'Atlanatis' carnation flowers. Growth of all five isolates was inhibited in nutrient broth (NB) supplemented with 8% ethanol, 8-HOC, or a combination of 8% ethanol + 8-HQC. Three of the five bacteria isolates were identified as Xanthomonas maltophila (CI 10), Flavomonas oryzihabitans (CI 12), and Xanthomonas species (CI 14). The isolates CI 11 and CI 13 have not been identified.

Introduction

Alcohols have been studied as a method to reduce ethylene sensitivity in carnation and tomato (Heins, 1980; Paull and Goo, 1982; Saltveit, 1989; Saltveit and Mencarelli, 1988; Wu et al., 1992). Continuous treatment with 8% ethanol doubled the vase life of 'White Sim' carnation flowers (*Dianthus caryophyllus* 'White Sim') (Wu et al., 1992). Silver thiosulfate (STS) also doubles the vase life of 'White Sim' carnation cut flowers when pulsed for 10 minutes (Reid et al., 1980). STS is the commercially accepted control of ethylene action. Nowak and Rudnicki (1990) anticipate that all ethylene sensitive flower species will require treatment with STS prior to entering international markets. However, STS is an expensive and harmful heavy metal environmental contaminant (Altman and Solomos, 1993). Alcohols may provide an alternative preservative treatment that is environmentally safe, less expensive, and readily available from renewable natural resources.

Vase solution bacteria may decrease the postharvest life of cut flowers by increasing enzymatic damage, disrupting plant cell membranes, stimulating plant wound ethylene, and causing physical occlusion at the base of or in the stem (Larsen and Cromarty, 1967; Marousky, 1971; Marousky, 1980; van Doorn et al., 1990; Zagory and Reid, 1986). Ethanol inhibits bacterial growth and is often used as a sterilant (Ingram, 1990). Ethanol in the vase solution of cut flowers may exhibit germicidal properties that contribute to vase life. Treatment with 8-HQC, a commercially accepted control of bacterial growth, prevents the accumulation of bacterial populations in the vase water (van

Doorn et al., 1990). Ethanol will be compared to 8-HQC in terms of germicidal properties. The objectives of this research were to determine the effects of ethanol on ethylene synthesis and sensitivity, microbial growth, and postharvest life of carnation.

Materials and Methods

Untreated standard white carnations (*Dianthus caryophyllus* L. 'Atlantis') stems were purchased from a commercial grower (Matsui Wholesale Flowers, Salinas, Calif.), shipped dry and recut to 20.0 cm.

Ethanol vs. STS - Exogenous Ethylene Sensitivity. Carnation stems were either pulsed or continually treated with 8% ethanol or 1 mM STS, then exposed to exogenous ethylene in a flow-through system. The carnation stems were continually treated or pulsed for 1 h in 8% ethanol or 1 mM STS, then transferred to deionized water (DI). Individual cut flower stems were sealed in a 3-liter glass jar and subjected to ethylene concentrations of 0.0, 0.2, or 1.0 ul·liter⁻¹ at a flow rate of 30-liters h⁻¹ for 20 h. A control of 0.0 ul·liter⁻¹ ethylene contained 50.0 g Ethysorb (StayFresh Ltd., London England) inside the jar to oxidize ethylene produced by the cut flower.

After ethylene treatment, flowers were removed from the containers and held at room temperature until no longer commercially acceptable. Senescence date for each cut flower was recorded and the vase life was calculated in days from the start of the ethylene treatment. The experimental design was a $3 \times 2 \times 2$ factorial with 10 stems

randomly assigned to each treatment as in a completly randomized design. Data within each experiments were analyzed by general linear model procedure with means separation by trend analysis (SAS Institute, Cary, N.C.) and all means were compared by least significant difference (LSD) (SAS Institute).

Ethanol vs. STS - Endogenous Ethylene Production. Carnation stems were placed in 8% ethanol or 1 mM STS and sealed in 3-liter glass jars equipped with flushing and sampling ports. Jars were flushed with ethylene-free air for approximately 3 min before being sealed. Jars were held at $21 \pm 3C$ for twelve days under continuous light exposure. Two 1-ml head-space gas samples were taken each day for CO_2 and ethylene accumulation until the carnation flowers were no longer commercially acceptable. Jars were sealed 2 h prior to each head-space gas sample. CO₂ analysis was conducted by injecting 1-ml head-space gas into an infrared gas analyzer (IRGA)(Horiba PIR-2000). Lamp grade nitrogen was used as carrier gas. A linear CO₂ response curve was established with gaseous standards of 5000 ul·liter⁻¹ CO₂ standard (Scott Specialty Gases, Plumsteadville, Pa). Ethylene analysis was conducted using a gas-tight syringe to inject 1-ml head-space gas into a gas chromatograph (GC) (Model 540, Tracor Instruments, Austin, Texas). The GC was equipped with a 30-m x 0.53-mm column (GS-Q Megabore, J & W Scientific, Folsom, Calif.) and flame ionization detector. Helium was used as Runs were conducted isothermally with an oven temperature of 60C. carrier gas. Injection and detection termperatures were 180 and 200 C, respectively. Mean production rates were calculated from response factors derived from gaseous standards (Scott Specialty Gases, Plumsteadville, Pa). The experimental design was a completely

randomized design with 5 stems randomly assigned to each treatment. Data were analyzed by General Linear Model procedure with means seperation by trend analysis (SAS Institute, Cary, N.C.) and mean production rates were compared by least significant difference (SAS Institute).

Ethanol vs. 8-HQC - Bacterial Population and Vase Life Determination. Carnation flowers were either pulsed or continuously incubated in sterile vases containing 200 ml of 8% ethanol or 200 mg·liter⁻¹ hydroxyquinoline + 300 mg·liter⁻¹ citric acid (Sigma Chemical Co.; St. Louis, MO) or 8% ethanol + 200 mg·liter⁻¹ hydroxyquinoline + 300 mg·liter⁻¹ citric acid. Pulsed stems were treated with chemical solution for 24 h in a dark cooler at 2 ± 1 C, transferred to sterile DI water (SDW), and incubated at $21 \pm$ 2C under continuous light exposure. Continually treated and control stems were incubated in solution or SDW, respectively, at room temperature under continuous light exposure.

Bacterial populations of the vase solutions were determined by dilution plating at 48 h intervals from the initiation of the experiment until the carnation flowers were no longer commercially acceptable. Aliquots (0.1 ml) from serial dilutions of the vase solutions were plated on nutrient agar (NA) (Difco, Detroit, MI) plates containing 100 ug ml⁻¹ cyclohexamide (NAc) to inhibit fungal growth. Bacterial counts were recorded following incubation for 5 days at 25C. Fifteen bacterial isolates, representing the most common colony morphology types among the treatments, were purified and stored in 15% glycerol at -70 C for subsequent analysis. Senescence date for each cut flower was recorded and the vase life was calculated in days from the start of treatment. The experimental design was a 2 x 4 factorial arrangement. Ten stems were placed in a completely randomized design and blocked by harvest date. Data was analyzed by General Linear Model procedure with means separation by least significant difference (SAS Institute).

Ethanol vs. 8-HQC - Bacteria Identification and Inoculation. Five isolates, designated carnation isolate (CI) CI3, CI10, CI11, CI12, and CI14, were utilized in further studies. The Gram reaction of the isolates was determined using a stain procedure, and three of the isolates were tentatively identified using Biolog GN microplates (Biolog Inc., Hayward, CA). Bacterial isolates were grown on Trypticase soy agar for 24-48 h. Bacterial cells were removed with a sterile cotton swab, suspended in sterile 0.85% NaCl to an OD₆₀₀ of 0.15. 150 ul of the bacterial suspension was loaded into each well of the GN microplates. The plates were incubated for 24-48 h at 28 C in the dark. Two replicate GN microplates were utilized for each isolate. Plates were read at 24, 48 and 72 h with a 590 nm filter on a microplate reader. Results were analyzed with Biolog GN database version 3.50 to determine the identity of each isolate. The growth and survival of the bacterial isolates in the germicidal solutions was monitored in nutrient broth (NB) and NB amended with 8% ethanol, 8-HQC, or 8% ethanol + 8-HQC. Following a 24-48 h incubation at 25 C on NA, the bacteria were suspended in 0.85% NaCl to an OD₆₀₀ of 0.15 using a Bausch and Lomb Spectronic 20 spectrophotometer. Five µl of the bacterial suspension was inoculated into NB or amended NB in standard culture tubes. The tubes were incubated at 280 rpm on a rotary shaker for 24-48 h at 28 C. The experimental design was a 5 X 4 factorial arrangement in a completely ransomized design. Four replicate tubes were utilized for each experiment, and the experiment was repeated once. Bacterial counts from each tube were determined by dilution plating on NA.

The bacterial isolates CI3, CI10, CI11, CI12, and CI14 were then examined to determine their effect on vase life of 'Atlantis' carnation flowers incubated in SDW. Five carnation stems were placed in a completely randomized design and individually assigned to each treatment. Bacterial suspensions in SDW were prepared as described above. Two-ml of inoculum was diluted into 198 ml SDW. One carnation flower was incubated in each vase; the top of each vase was covered with Parafilm (American Can Co., Greenwich, CT) to reduce bacterial contamination. The experiments were performed under continuous light exposure at 25C, and were terminated when individual flowers were no longer commercially acceptable. At termination, bacterial counts from vase solutions and flower vase life were determined as described above.

Results

Ethanol vs. STS. Rate of ethylene production was not different between 'Atlantis' carnation stems held in STS or ethanol (Table 4.1, Fig. 4.1). A peak in ethylene production of 26.0 nmol·g. fr. wt⁻¹·h⁻¹ was reached by day nine for ethanol-treated stems while a smaller peak of 13.4 nmol·g. fr. wt.⁻¹·h⁻¹ was exhibited by day eight for STS-treated stems (Table 4.1, Fig 4.1). Respiration rate decreased significantly as incubation period increased. STS-treated stems had a significantly higher CO₂ production rate as

compared to ethanol-treated stems (Table 4.1, Fig. 4.2). Vase life of 'Atlantis' carnation stems continually treated in STS was significantly higher than ethanol-treated stems when exposed to exogenous ethylene (Table 4.2). Vase life of carnation stems decreased significantly when exposed to 1.0 ul·liter⁻¹ ethylene (Table 4.2). Exposure to 0.2 ul·liter⁻¹ exogenous ethylene did not affect the vase life as compared to the control of STS or ethanol-treated stems.

Ethanol vs. 8-HQC. Carnation stems continually treated with 8% ethanol had the highest bacterial count at vase life termination $(1.15 \times 10^7 \text{ cfu} \text{ ml}^{-1} \text{ vase solution})$ and vase life (10.4 days) (Tables 4.4 and 4.5, respectively). Stems continually treated with 8-HQC or a combination of 8% ethanol + 8-HQC exhibited the least number of bacteria and a vase life of 8.7 and 7.3 days, respectively (Tables 4.4 and 4.5). Stems held in DI water had a significantly lower mean bacterial count than stems continually held in 8% ethanol after day 4. Vase life of carnation stems was not different between control stems or stems continuously held or pulsed in 8% ethanol, 8-HQC, or 8% ethanol + 8-HQC (Table 4.3).

Growth and survival of CI 10 was significantly higher in tubes containing NB only than tubes amended with germicide (Table 4.6). Bacterial growth in NB treated with 8% ethanol, 8-HQC, or a combination of 8% ethanol + 8-HQC was less than the untreated control (Table 4.6). CI 11 and CI 12 growth was completely inhibited in NB treated with a combination of 8% ethanol + 8-HQC (Table 4.5). CI 13 growth was not inhibited in NB treated with 8% ethanol, 8-HQC, or a combination of 8% ethanol + 8-HQC, but was significantly lower than the control. CI 14 growth and survival in NB was 8.2 log cfuml⁻¹ which was significantly higher than in tubes containing 8% ethanol, 8-HQC, or a combination of 8% ethanol + 8-HQC. Following the inoculation of a SDW vase solution with the five isolates, CI 11, CI 12, and CI 14 resulted in a significantly shorter carnation vase life as compared to the control (Table 4.7). The vase life of flowers inoculated with CI 10 and CI 14 were not different from the control (Table 4.7). Three of the isolates were tentatively identified using Biolog GN microplates (Biolog Inc., Hayward, CA) as *Xanthomonas maltophila* (CI 10), *Flavomonas oryzihabitans* (CI 12), and *Xanthomonas species* (CI 14). The remaining isolates were not identifiable by use of the Biolog GN microplates.

Discussion

Carnation stems continuously held in 8% ethanol or STS produced less than 50 nmol·g fr. wt.⁻¹·h⁻¹ ethylene (Figure 4.1). 8% ethanol and STS treated 'Atlantis' stems did not exhibit a respiratory peak. Standard 'White Sim' carnation stems had a peak in ethylene production of 230 nl/flower/h (Heins, 1980). Similar to findings by Wu et al. (1992) and Heins (1980), ethylene production of 'Atlantis' carnation stems was strongly suppressed by continuous treatment in 8% ethanol. Ethylene peak reached by continually-treated STS stems was one-half that of the ethylene peak produced by continually-treated ethanol stems.

Wu et al. (1992) suggested that the primary effect of ethanol is preventing the

induction of increased ethylene biosynthesis. Treatment with ethanol vapor inhibited lycopene and ethylene synthesis in ripening tomato fruit (Saltveit and Mencarelli, 1988). Pulse treatment as short as 10 min with 1.0 mM STS increased the vase life of 'White Sim' carnation (Dianthus caryophyllus 'White Sim') cut flowers (Reid, et al., 1980). STS inhibites ethylene action by binding to the physiological receptor (Reid and Wu, 1992). Wu et al. (1992) suggested that ethanol interferes with ethylene action other than competing with the ethylene binding site. Once ethylene production begins, ethanol was not effective in inhibiting ethylene synthesis (Heins, 1980). Continuous treatment with ethanol reduced the accumulation of the immediate precursor of ethylene, 1aminocyclopropane-1-carboxylic acid (ACC), and completely inhibited the activity of the ethylene forming enzyme (EFE). Treatment of grape berries (Vitis vinifera L. cv. Sultanina) with acetaldehyde (AA) caused a reduction in ethylene evolution (Pesis and Marinansky, 1992). Pesis and Marinansky (1992) suggested that AA is the reactive compound in the inhibition of ethylene biosynthesis of carnation flowers because ethanol is converted into AA by alcohol dehydrogenase which is present in carnations.

Exposure to exogenous applications of 1.0 ul·liter⁻¹ ethylene significantly decreased the vase life of Atlantis carnation cut flowers (Table 4.2, Fig. 4.3). Treatment of 'White Sim' carnation cut flowers treated with 8% ethanol reduced the senescence-promoting effects of exogenous ethylene at concentrations less than 0.6 ul·liter⁻¹ (Wu et al., 1992). Inhibition of ethylene action in carnation by ethanol was noncompetitive. Application of STS to 'White Sim' carnation flowers reduced ethylene binding activity suggesting that silver competitively inhibits ethylene action (Reid and Wu, 1992). Mean

vase life of 'Atlantis' stems exposed to exogenous applications of 0.0, 0.2, or 1.0 ul·liter⁻¹ ethylene and treated with STS was significantly longer (6.5 days) than ethanol-treated stems (5.3 days) (Table 4.2, Fig. 4.3). 'Atlantis' stems exposed to 0.2 ul·liter⁻¹ ethylene and continually-treated with STS had a significantly longer vase life than continual and pulsed ethanol-treated stems.

Continuous treatment with 8% ethanol caused phytotoxicity along the stem which appeared as white to brown streaks. In addition, continual treatment with 8% ethanol caused stem topple of 'Atlantis' carnation cut flowers. Similar to findings by Heins (1980) and Wu et al. (1992), the stem collapsed at the node immediately above the vase solution.

The vase life of 'Atlantis' carnation stems continually treated with 8% ethanol was significantly longer than stems continually treated with 8-HQC or 8-HQC + 8% ethanol (Table 4.3). However, the bacterial count of continually ethanol-treated stems was significantly higher than 8-HQC or 8-HQC + ethanol. High molar concentrations of ethanol were required to inhibit growth, kill cells, or block metabolism of bacteria (Ingram, 1990). Some bacteria were sensitive to ethanol while others are not. *Escheria coli* was sensitive to ethanol concentrations above 6% while *Lactobacillus heterohiochii* and *L. homohiohnii* were capable of growth in ethanol concentrations above 18% by volume (Ingram, 1990). Addition of ethanol to growing cultures of *E. coli* caused a change in fatty acid composition with the replacement of 16:0 with 18:1 fatty acid. The change is immediate, dose dependent, and reversible (Ingram, 1990). When *E. coli* cells were grown in the presence of 16:0 fatty acid, the result was a change in membrane fatty

acid composition which was accompanied by death in the presence of ethanol. The importance of fatty acid composition was not within the degree of saturation, but the length of the chain. The adaptive response of *E. coli* to increase fatty acid chain length by shifting from 16 to 18 carbon fatty acids increased the number of CH_2 units that interact and strengthen the primary membrane barrier which determines permeability (Ingram, 1990). The bacteria present in the ethanol-treated carnation stems may have been ethanol-tolerant bacteria.

Bacteria may produce ethylene in the vase solution (Zagory and Reid, 1986) and bacteria may cause xylem blockage (van Doorn et al., 1989). Silver nitrate reduced the number of bacteria and prevents xylem blockage but STS did not reduce bacteria number or affect hydraulic conductance (van Doorn et al., 1989). When two thirds of rose xylem vessels were occluded with a razor blade water uptake was not reduced because the loss of vessels was counteracted by an increased flow rate in the remaining non-occluded vessels (van Doorn et al., 1989). A mean bacterial count of 1.2 x 10⁷ cfurml⁻¹ was observed in continually ethanol-treated stems terminated at day 16 (Table 4.4). Stems terminated at days 6, 8, and 10 exhibited bacterial counts of 10⁶ cfu^{-ml⁻¹} which was sufficient to decrease hydraulic conductance as reported by van Doorn et al. (1989). However, the vase life of continually ethanol-treated stems was not decreased. 8-HOC inhibited ethylene production in rose (Rosa hybrida) (van Doorn et al., 1989) and reduced vascular blockage in 'Better Times' rose (R. hybrida 'Better Times') (Marousky, 1971). 8-HQC and the 8-HQC + ethanol continually-treated stems demonstrated a significantly lower mean bacterial count. However, the vase life of both treatments was significantly

lower than the continually ethanol-treated stems which demonstrates the primary importance in extending the postharvest life of carnation cut flowers by inhibiting the production and action of ethylene.

Vase life of 'Atlantis' carnation stems held in SDW inoculated with carnation isolate (CI) 11, CI 12, and CI 13 was significantly shorter as compared to the control (Table 4.7). CI 12 was identified as Flavomonas oryzihabitans which has not been reported in previous bacteria evaluations of cut flower vase solutions. CI 10 and CI 14 were identified as Xanthomonas sp. which have also not been reported in previous research. Bacteria have been isolated and identified as Pseudomonas sp., Flavobacterium lutescens, and Alcaligenes faecalis from vase solutions containing 'Sonia' rose (Rosa hybrida 'Sonia') which were not inhibited by 600 mg liter⁻¹ hydroxyquinoline citrate (van Doorn and de Witte, 1990). Pseudomonas sp. has been the most widely used to observe the effect of bacteria in the vase solution on the postharvest life of cut flowers (Jones and Hill, 1993; van Doorn and de Witte, 1990; van Doorn and de Witte, 1994). Both Flavobacterium sp. and Alcaligenes sp. were present for identification in the Biolog data base indicating that the isolates CI 11 and CI 13 are bacteria species that have not been identified previously or are not available in the Biolog data base. Vase life of 'Atlantis' carnation stems inoculated with CI 12 (Flavomonas oryzihabitans) was significantly shorter than the control. CI 12 should possibly be included in future studies to observe the effect of bacteria on the postharvest life of cut flowers. Growth of CI 13 in NB treated with 8% ethanol, 8-HQC, or 8% ethanol + 8-HQC was less than the control but exceeded 10⁵ cfu⁻¹¹ and decreased the vase life of 'Atlantis' carnations demonstrating a deleterious effect on the vase life of carnation flowers.

In summary, STS was a more effective postharvest treatment as compared to continual treatment with 8% ethanol by reducing the production of ethylene and suppressing the climacteric respiratory peak. STS and 8% ethanol did not prevent the senescence promoting effects upon exposure to 1.0 ul·liter⁻¹ ethylene. STS extended the vase life and prevented the action of 0.2 ul·liter⁻¹ ethylene as compared to stems continually treated with ethanol. Continual treatment with 8% ethanol inhibited ethylene production and action and did not provide germicidal properties.

Ethanol is desirable as a floral preservative because it is environmentally safe. The practical value of ethanol as a single postharvest preservative treatment is limited by the need to continually treat flowers (Wu et al., 1992). In addition, the effects of high ethanol levels (phytotoxicity and stem topple) are not desirable to the cut flower industry. The film processing industry safely uses and disposes of more silver than the floral industry each year (Staby et al., 1993). Handlers of cut flowers should be able to safely use STS by implementing proper silver recovery procedures (Staby et al., 1993). Research efforts to identify other environmentally safe postharvest floral preservatives is encouraged.

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Table 4.1. Analysis of variance for ethylene and CO_2 biosynthesis of standard white 'Atlantis' carnations continually treated in 1 mM STS or 8% ethanol.

Source of Variation	df	Mean square
	Ethylene	
Treatment	1	164.77 ^{NS}
Day	11	155.87 ^{NS}
Stem	9	92.57 ^{NS}
Treatment x day	11	212.61*
	CO2	
Treatment	1	1.23x10 ^{6***}
Day	11	6.20x10 ^{6***}
Stem	9	3.57x10 ^{6***}
Treatment x day	11	$9.72 \times 10^{4 \text{NS}}$

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

Table 4.2. Analysis of variance for the effects of exogenous ethylene on the vase life of standard white 'Atlantis' carnations continuously held or pulsed (treatment) in 8% ethanol or 1mM STS (chemical).

Source of Variation	df	Mean Square
Chemical	1	37.93***
Treatment	1	286.74***
Chemical x Treatment	1	1.11 ^{NS}
Ethylene	2	515.95***
Chemical x Ethylene	2	15.73**
Treatment x Ethylene	2	38.46***
Chemical x Trmt x Ethylene	2	2.23 ^{NS}

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

Table 4.3. Analysis of variance for the effects of pulse or continual treatment of 'Atlantis' carnation stems in 8% ethanol, 200 mg·liter⁻¹ + 300 mg·liter⁻¹ citric acid (8-HQC), a combination of 8% ethanol + 8-HQC, or deionized water on mean bacteria colony forming units per ml (cfu·ml⁻¹).

Source of Variation	df	Mean square
Treatment	6	88.82***
Rep	9	4.68 ^{NS}
Day	8	155.96***
Treatment x Day	34	11.38***

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

Table 4.4. Mean bacterial colony forming units (cfu) per 1-ml vase solution samples taken every two days until termination of vase life. Control stems were continuously held in sterilized deionized water (SDW) at $21\pm 2C$.

Treatment	Day	Number of Stems	Bacterial Count (cfu/ml vase solution)
Control	0	10	0.0
Control	2	10	5.15×10^{1}
Control	4	9	8.45 x 10 ⁴
Control	6	8	1.58×10^{5}
Control	8	4	1.34×10^{5}
Pulse 8% Ethanol	0	10	0.0
Pulse 8% Ethanol	2	10	1.77 x 10
Pulse 8% Ethanol	4	10	1.82×10^3
Pulse 8% Ethanol	6	10	2.81×10^{5}
Pulse 8% Ethanol	8	8	5.38 x 10 ⁵
Pulse 8% Ethanol	10	1	1.10 x 10 ⁶
Pulse 8-HQC	0	10	0.0
Pulse 8-HQC	2	10	0.0
Pulse 8-HQC	4	10	2.14×10^3
Pulse 8-HQC	6	10	1.75×10^{5}
Pulse 8-HQC	8	6	2.89×10^5
Pulse Ethanol + 8-HQC	0	10	0.0
Pulse Ethanol + 8-HQC	2	10	3.16 x 10
Pulse Ethanol + 8-HQC	4	10	5.94×10^3
Pulse Ethanol + 8-HQC	6	8	9.23 x 10 ⁴
Pulse Ethanol + 8-HQC	8	6	9.3×10^{5}
Pulse Ethanol + 8-HQC	10	3	1.48 x 10 ⁶
Continuous 8% Ethanol	0	10	0.0
Continuous 8% Ethanol	2	10	0.0
Continuous 8% Ethanol	4	10	4.17×10^2
Continuous 8% Ethanol	6	10	2.55 x 10°
Continuous 8% Ethanol	8	9	3.20 x 10°
Continuous 8% Ethanol	10	6	5.45 x 10°
Continuous 8% Ethanol	12	3	$5.41 \times 10^{\circ}$
Continuous 8% Ethanol	14	3	2.03 x 10'

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Continuous 8% Ethanol	16	1	1.15 x 10 ⁷
Continuous 8-HQC	0	10	0.0
Continuous 8-HQC	2	10	1.26 x 10
Continuous 8-HQC	4	9	2.44 x 10
Continuous 8-HQC	6	9	1.91×10^{1}
Continuous 8-HQC	8	5	1.55×10^{1}
Continuous 8-HQC	10	3	1.17×10^2
Continuous 8-HQC	12	2	0.0
Continuous 8-HQC	14	2	0.0
Continuous 8-HQC	16	2	0.0
Continuous ETOH+8-HQC	0	10	0.0
Continuous ETOH+8-HQC	2	10	1.70 x 10
Continuous ETOH+8-HQC	4	7	0.0
Continuous ETOH+8-HQC	6	6	2.82 x 10
Continuous ETOH+8-HQC	8	4	0.0
Continuous ETOH+8-HQC	10	3	0.0
Continuous ETOH+8-HQC	12	1	0.0
Continuous ETOH+8-HQC	14	1	0.0
Continuous ETOH+8-HQC	16	1	1.50×10^2

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Table 4.5. Mean vase life of 'Atlantis' carnation stems pulsed or continually treated in 8% ethanol, 200 mg·liter⁻¹ hydroxyquinoline + 300 mg·liter⁻¹citric acid (8-HQC), or a combination of 8% ethanol + 8-HQC. Control stems were continuously held in sterile deionized water at $21 \pm 2C$. Means are an average of data from 10 stems/trmt.

Treatment	Vase life (days)	
Control	6.6	
Pulse 8% Ethanol Pulse 8-HQC Pulse Ethanol + 8-HQC	8.3 ^{NS} 7.8 ^{NS} 7.8 ^{NS}	
Continuous 8% Ethanol Continuous 8-HQC Continuous ETOH + 8-HQC	10.4*** 8.7 ^{NS} 7.3 ^{NS}	

^{NS}, *** Nonsignificant or significant as compared to the control at P \leq 0.001, respectively.

Table 4.6. Mean log colony forming units (log cfu ml⁻¹ vase solution) of carnation bacteria isolates (CI) CI 10, CI 11, CI 12, CI 13, and CI 14 incubated for 24-48 hr on a rotary shaker in sterilized, deionized water (SDW), 8% ethanol, 200 mg liter⁻¹ hydroxyquinoline + 300 mg liter⁻¹ citric acid (8-HQC), or a combination of 8% ethanol + 8-HQC. Means are an average of 8 samples/treatment.

 Treatment	log cfu/ml	
CI 10		
SDW	10.6	
8% ethanol	5.74	
8-HQC	5.78	
8% ethanol+8-HQC	5.60	
LSD	0.44	
CI 11		
SDW	6.86	
8% ethanol	0.09	
8-HQC	0.09	
8% ethanol+8-HQC	0.0	
LSD	0.29	
CI 12		
SDW	9.94	
8% ethanol	0.38	
8-HQC	5.21	
8% ethanol+8-HQC	0.0	
LSD	0.31	

CI 13	
SDW	7.94
8% ethanol	4.98
8-HQC	5.15
8% ethanol+8-HQC	4.98
LSD	0.65

- 10.004

CI 14

SDW	8.20
8% ethanol	1.24
8-HQC	1.10
8% ethanol+8-HQC	1.15
LSD	1.09

Table 4.7. Mean vase life of 'Atlantis' carnation stems held in 200-ml sterilized deionized water (SDW) or SDW inoculated with carnation isolates (CI), 10, CI 11, CI 12, CI 13, or CI 14. Means are an average of five stems/inoculum treatement.

Treatment	Vase life (days)	
Control	6.0	
CI 10 CI 11 CI 12 CI 13 CI 14	6.2 ^{NS} 5.0** 5.2** 5.4** 6.0 ^{NS}	

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

List of Figures

Figure 4.1. Ethylene biosynthesis of 'Atlantis' carnation stems continually treated with 8% ethanol (--) or STS (--). Head-space gas samples were taken daily until the flower was no longer commercially acceptable. Means are an average of data from 5 stems/treatment. Bars represent \pm SE.

Figure 4.2. CO_2 production of 'Atlantis' carnation stems continually treated with 8% ethanol (-•-) or STS (--). Head-space gas samples were taken daily until the flower was no longer commercially acceptable. Means are an average of data from 5 stems/treatment. Bars represent <u>+</u> SE.

Figure 4.3. Vase life in days of 'Atlantis' carnation stems pulsed in 8% ethanol (- \blacklozenge -) or STS (- \blacktriangle -), or continually treated in 8% ethanol (- \blacksquare -), or STS (- \bullet -). Means are an average of 10 stems/treatment.

Figure 4.4. Mean number of bacterial colony forming units/ml vase solution (cfu/ml) for 'Atlantis' carnation stems pulsed in 8% ethanol (--), 200 mg·liter⁻¹ hydroxyquinoline + 300 mg·liter⁻¹ citric acid (8-HQC) (--), or a combination of 8% ethanol + 8-HQC (--). Continually-treated stems were held in 8% ethanol (--), 8-HQC (--), or a combination of 8% ethanol + 8-HQC (--). Control stems (--) were continuously held in sterilized deionized water (SDW) at 21 <u>+</u> 2C.

Figure 4.1.



Figure 4.2.







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Figure 4.4.



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Chapter V

Summary

Interest in the production of specialty cut flowers is gaining among producers and consumers. The quest to find a unique cut flower that is easily produced, having attractive flower color with strong stems that are of sufficient length is demanding. A cut flower must also exhibit an extended postharvest life to be able to withstand the handling during marketing and meet consumer expectations for longevity after purchase.

Purple coneflower, Maximillian's sunflower, penstemon, 'Coronation Gold' yarrow, 'Forest Fire' celosia, 'Sensation' cosmos, eastern redbud, butterfly bush, and weigela exhibit suitable production characteristics for the cut flower market. Postharvest attributes of these species were examined.

Purple coneflower could be stored for one week at 2 or 4C and shipped up to five days. Maximillian's sunflower can be stored for one week at 2C and shipped up to one day and STS, sucrose, and 8-HQC preservatives extend the vase life. Penstemon could be stored at 4 or 7C for one week and up to three weeks at 2C. Coronation Gold yarrow could be stored at 2C up to two weeks and shipped for two days and STS, sucrose, and 8-HQC preservatives extend the vase life. Forest Fire celosia could be stored at 2, 4, or 7C for one week and STS and 8-HQC preservatives extend the vase life. Sensation cosmos could be stored at 2C up to two weeks and shipped for one day. Butterfly bush

could be stored at 2C up to 2 weeks and STS, sucrose, and 8-HQC preservatives extend vase life. Eastern redbud could be stored at 2C up to two weeks. Weigela did not tolerate cold storage or shipping and STS, sucrose, and 8-HQC preservatives may extend the vase life.

During shipping or handling, the postharvest life of cut flowers may decrease upon exposure to ethylene. Cut flowers that are ethylene sensitive should not be shipped or stored near other cut flowers or produce that evolve ethylene. Yarrow, celosia, cosmos, and butterfly bush produced a small ethylene peak attributed to wound ethylene and did not respond to exogenous ethylene. Purple coneflower did not produce ethylene and did not respond to exogenous ethylene. Maximillian's sunflower did not produce an ethylene peak and was sensitive to exogenous ethylene. Penstemon and weigela produced a small ethylene peak attributed to wound ethylene and responded to exogenous ethylene by exhibiting severe flower abscission and decreased vase life. Maximillian's sunflower, penstemon and weigela should not be shipped or stored near ethylene producing plant material and should be treated with anti-ethylene preservatives.

Ethanol was confirmed as a postharvest floral treatment that can inhibit ethylene biosynthesis and action. Ethanol is a desirable postharvest treatment because it is readily available, easily made, and environmentally safe. Current commercial practices to prevent ethylene sensitivity include the use of STS which contains the heavy metal, silver, and is not environmentally safe. STS-treated carnation cut flowers produced less ethylene and lasted longer than ethanol-treated flowers.

Additional postharvest attributes such as germicide properties were investigated

by comparing ethanol to 8-hydroxyquinoline citrate (8-HQC). Ethanol treated stems had the greatest number of bacterial colony forming units (cfu) and longer vase life than 8-HQC-treated stems. The anti-ethylene properties of ethanol were of greater importance to the postharvest life of carnation flowers than its germicidal properties.

Five bacteria isolates were identified as carnation isolate (CI) 10, CI 11, CI 12, CI 13, and CI 14. The isolates CI 11, CI 12, and CI 13 decreased the vase life of 'Atlantis' carnations. Isolates CI 10 and CI 14 did not affect the vase life of 'Atlantis' carnation flowers. Growth of all five isolates were inhibited in nutrient broth (NB) amended with 8% ethanol, 8-HQC, or a combination of 8% ethanol + 8-HQC. Three of the five bacteria isolates were identified as *Xanthomonas maltophila* (CI 10), *Flavomonas oryzihabitans* (CI 12), and *Xanthomonas* species (CI 14). The isolates CI 11 and CI 13 have not been identified.

The continued search for new cut flower species that have an extended postharvest life should be continued to diversify cut flower production and to maintain consumer interest. Even though current postharvest practices include compounds that are not environmentally safe, these compounds were the most effective in extending the vase life of cut flowers. Attention to proper use and disposal of used floral preservatives should be a common practice of cut flower handlers. Further research to discover environmentally safe floral preservatives is recommended.

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