PRIMING AND POST-PRODUCTION OF *TAGETES* AND *CAMPANULA* SPECIES

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CHAPTER I

INTRODUCTION

Horticulture products have always been relied upon in every aspect of our society. Some applications are obvious such as fruit and vegetable consumption or the florist industry, but not all applications are apparent to the average consumer. One example is the colorants used by the poultry and dairy industry. Natural sources of these pigments come from a common bedding plant and another from a favorite vegetable, corn. The muli-faceted application of these plants and others ensures their continued profitability, which in turn stimulates further research resulting in practical knowledge and application.

The florist industry is a conspicuous outlet of horticulture products, but many are unaware of the industry's contribution to the national economy. In 1999, the United States cut flower industry grossed $426 million dollars, which was a 3% increase over the previous year (United States Department of Agriculture, 2000). Much of the increase was due to specialty cut flowers, which have been growing in popularity with the industry and consumer for the last several years. However, continued success of specialty cut flowers requires introduction of new species with proper postharvest handling protocols clearly identified.

TAGETES ERECTA COMMERCIAL PRODUCTION

African marigold (*Tagetes erecta* L.) flower pigments can be extracted and used as a natural food colorant by imparting an orange color to egg yolks and poultry skin. Lutein and zeaxanthin are the main xanthophyll pigments present in marigold flower

petals and subsequently egg yolks due to their highly absorptive nature with fatty tissue (Karunajeeva et al., 1984). Currently, *T. erecta* plants are grown in Mexico, Peru, and India, but the quality of extracted pigments is low. Hence, a more efficient program must be developed for harvesting and processing lutein.

COOL GERMINATION TEMPERATURES

Early spring sowing hinders commercial production of *T. erecta*. Spring field emergence is unpredictable due to suboptimal temperatures depending upon the plant species. Suboptimal temperatures have been correlated with a reduction in germination rate, total germination percentage, and seedling emergence (Kondra et al., 1983; Livingston and de Jong, 1990; and Blackshaw, 1991). However, primed perennial ryegrass (*Lolium perenne* L.) seed had significantly greater germination percentage and seedling root growth than control seed at suboptimum germination temperatures of 5, 10, and 15°C (Danneberger et al., 1992).

PRIMED SEED

Priming enhances seed germination by manipulating the seed's hydration level allowing germination to begin but is halted before radicle emergence. Priming improves emergence uniformity and reduces seedling exposure to soil crusting, soil-borne pathogens, and unfavorable temperatures (Bennett et al., 1992). As a result, the time required for germination and field emergence is shortened and stand establishment of direct-seeded plants is improved.

PLANT STAND ESTABLISHMENT

Direct-seeded plants are vulnerable to many factors affecting stand establishment, such as soil crusting, poor seed to soil contact, high or low temperatures, and soil

moisture (Bennett et al., 1992). Consequently, direct-seeded plants often have uneven germination, slow emergence, and poor stand establishment (Wurr and Fellows, 1983). While transplants are initially more expensive to grow and set in the field, they can be planted at the exact space desired for the crop and have the benefit of quickly producing a uniform stand for earlier harvests. Transplanted plants often produce their first harvestable crop of flowers before direct-seeded plants begin to flower.

NITROGEN APPLICATION

Baldwin et al. (1993) conducted an eight year study in which five *T. erecta* cultivars were grown in Virginia and Mexico for pigment extraction and lesion-nematode reduction. They noted plant "vigor" decreased as the season progressed due to nitrogen deficiency. In the following season, the addition of (4.6 kg/ha) of ammonium nitrate after the first two harvests improved the following harvests. Pigment yield increased most after three nitrogen applications in a single season.

TAGETES HARVEST METHOD

Flowers of *T. erecta* plants grown in other countries are hand-picked due to the plentiful and relatively cheap labor. If *T. erecta* plants are to be commercially grown in the United States, mechanical harvesting must be a viable option. The plant must withstand the destruction a mechanical harvester can inflect on plants after multiple harvests and still yield acceptable flower mass.

CAMPANULA MEDIUM 'CHAMPION' CUT FLOWERS

Campanula medium L. is a biennial plant commonly used in gardens in the United States and sold as flowering potted plants in Europe (Dole and Wilkins, 1999). 'Champion' cultivars were bred specifically for cut flower production. They have a short

crop time, typically flowering in 16 to 20 weeks, making it possible to grow and sell this normally biennial plant in a cost effective time period (Cavins, 1999). However, limited research has been done that examines the postharvest life of cut *C. medium* flowers (Sakata Corp., personal communication).

COOLER STORAGE

Storing cut stems in coolers benefits the grower and florist by extending the season, regulating availability during peak production or high demand periods, improving production efficiency, and enabling long-term shipment (Goszczynska and Rudnicki, 1988). Low temperature storage reduces the rate of metabolism, transpiration, endogenous ethylene production, and pathogen growth thus maintaining quality of the stored material.

PRETREATMENTS AND PULSES

Pretreatments and pulses are techniques that greatly enhance the quality and longevity of many cut flowers. Pretreatments and pulses are generally short-term treatments lasting a few hours just after harvest but have shown to increase rate and amount of flowers opening, improved petal coloration, and extended vase life of cut flowers (Nowak and Rudnicki, 1990).

CONTINUOUS SUCROSE

Cut stems of many species benefit from a continuous supply of sucrose in the vase solution. Sucrose replaces photosynthates after the stems are harvested, increasing vase life and enabling buds to properly develop and become larger (Halevy and Mayak, 1979; Sacalis, 1993). Sucrose percentages normally used in vase solutions are lower than those for pulsing.

LIGHT

Light conditions are not known to be a significant postharvest factor for cut flowers. Vase life has shown to be unaffected, but low light conditions after harvest can affect flower color development. Incomplete coloration of thin petals has been correlated with purple cultivars of *Eustoma grandiflorum* Griese. under low light after stem harvest (Griesbach, 1992; Kawabata et al., 1995). Thus, the effect of postharvest light conditions on cut *C. medium* stems should be examined.

OBJECTIVES

This research project had three objectives:

1) to evaluate the effect of cultivar, sowing date, and priming treatments on *T. erecta* field seedling emergence and stand establishment;

2) to determine the *T. erecta* cultivar and production method most suitable for commercial production yielding the greatest quantity of lutein pigment; and

3) to maximize *C.medium* 'Champion' storage life, color development in flowers opening after harvest, and vase life for the retailer and consumer.

Information gained from this research will promote commercial production and mechanical harvest of direct-seeded *T. erecta* plants for pigment recovery. In addition, postharvest care and quality of *C. medium* flower stems will be improved within the florist industry.

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CHAPTER II

PRIMED *TAGETES ERECTA* SEEDS ENHANCE FIELD EMERGENCE

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ABSTRACT

Field seedling emergence of four African marigold (*Tagetes erecta* L.) cultivars, 'A-975', 'E-1236', 'I-822', and 'Orange Lady', was examined over three years using three or four spring sowing dates. For all cultivars, seeds sown on 15 May 1998 were the first to emerge (4.1 days), had the shortest emergence time (5.7 days), and emerged most uniformly (3.9 days). Total emergence percentages were highest for the last sowing date, except for 'A-975', which linearly decreased from the first (88%) to the last sowing date (34%). In 1998, 'I-822' had the slowest emergence time (8.4 days) and greatest number of days to first harvestable flower (88 days) compared with other cultivars. 'Orange Lady' produced the largest fresh flower mass (36,900 $kg.ha^{-1}$). In 1999, seed sown on the third of four planting dates, 29 Apr., had the fewest number of days to first emergence

(5.3) and fastest emergence time (7.3 days). Cultivar and sowing date interacted such that total emergence was highest on 29 Apr. for 'A-975' and 'Orange Lady' but on 15 Apr. for 'I-822'. Total emergence for 'E-1236' was low (≤16%) regardless of sowing date. Sowing on 15 Apr. and 29 Apr. produced the highest flower number (399 and 387) and fresh flower mass (52,500 and 52,900 $kg.ha^{-1}$), respectively. In 2000, wet priming of 'E-1236' and 'I-822' seed shortened emergence time, emergence uniformity, and increased total emergence percentage at early sowing dates compared with both solid matrix primed and unprimed seed.

INTRODUCTION

African marigold (*Tagetes erecta* L.) petals are commercially valuable as a natural source of xanthophyll pigments (yellow-orange pigments) used primarily by the poultry industry to color egg yolks and poultry skin. Currently, marigold plants are grown for pigment production in Mexico, Peru, and India, but the quality of extracted pigments is low. Baldwin et al. (1993) conducted an eight year study in Virginia and Mexico and concluded that the two greatest problems for marigold production were plant stand establishment and weed control. Stand establishment was unsuccessful unless large quantities of de-tailed seed were used in the precision seeder. Otherwise, repeat plantings were necessary to achieve adequate stand establishment.

Stand establishment is a common problem in commercial crop production. Proper stand establishment will lead to early and uniform harvests but can be restricted by soil crusting, poor seed to soil contact, extremely high or low temperatures, pathogen

invasion, and inadequate soil moisture (Bennett et al., 1992). Direct-seeded plants, which are typically slow to emerge, are vulnerable to many of these factors causing uneven germination and non-uniform plant stand (Wurr and Fellows, 1983). Overall, fewer harvest periods may occur with direct seeding, but it is a more economical procedure for the producer than transplanting. If transplants are used, the seedlings are more likely to produce a uniform stand resulting in earlier harvests. Transplanted plants often produce their first crop when direct-seeded plants are just beginning to flower. However, transplant production and planting costs may be prohibitive, especially when required plant densities are high.

Priming is used to enhance germination of direct-seeded plants and improve stand establishment. Priming is defined as a presowing seed treatment to enhance germination and increase seedling emergence uniformity under adverse environmental conditions (Parera and Cantliffe, 1994). By shortening the length of time necessary for initial stand establishment, priming improves seedling uniformity and reduces seedling exposure to soil crusting, soil-borne pathogens, and unfavorable temperatures (Bennett et al., 1992). In priming, the hydration level of the seed is manipulated to allow the first and second stage of germination to begin, but germination is halted before the third stage, radicle emergence, is completed. Once the radicle emerges, the seed will die if removed from favorable hydration conditions. Primed seed may subsequently be stored and then transported to the field for planting. Thus, the time required for germination and emergence in field conditions is shortened. Seedling growth and development of primed seed are indistinguishable from untreated seed except under stressful conditions (Parera and Cantliffe, 1994). Osburn and Schroth (1989) discovered that sugar beet (*Beta*

vulgaris L.) seeds primed in either NaCl or polyethylene glycol (PEG) reduced damping-off disease (*Pythium ultimum)* by 50 and 65%, respectively, compared with unprimed seed.

Inorganic salts, e.g., KNO_3, K_3PO_4, and PEG, are the most commonly used priming treatments, while a chemical soak such as gibberellic acid can also be used in place of stratification with dormant seeds. Inorganic salt solutions are relatively inexpensive, easy to aerate, quickly removed from the seed, and possibly of nutritional benefit (Parera and Cantliffe, 1994). During the priming process, ions from the salt solution accumulate within the seed reducing the osmotic potential and increasing water absorption. Ion accumulation can also have a detrimental effect upon the developing embryo depending on the species and concentrations. Polyethylene glycol is a chemically inert compound, and its large molecules are prevented from damaging seed by penetration. However, PEG is expensive, difficult to aerate, and hard to remove from the seed after treatment due to its viscosity (Parera and Cantliffe, 1994).

Solid matrix priming is another priming method that consists of combining seeds with water and either an organic or inorganic material for a predetermined time period (Parera and Cantliffe, 1994). The solid organic or inorganic material allows water to adsorb to the particles' surfaces regulating the seeds' water uptake. Taylor et al. (1988) stated solid matrix priming materials should not cause phytotoxicity, should retain a high water holding capacity, be friable at different moisture levels, and remain easy to remove from seeds after treatment. Examples of solid matrix priming materials are vermiculite, calcined clay, and sodium polypropionate gel. Compared to other priming techniques solid matrix priming is inexpensive and can be used on large quantities of seed (Khan,

1992).

Factors that affect seed priming effectiveness are aeration, light, treatment duration, temperature, osmotic potential, seed quality, dehydration after priming, and seed storage (Parera and Cantliffe, 1994). Treatment duration may last from 6 hours to 3 weeks depending upon the species and often correlates with temperature. Haigh et al. (1986) found that carrot 'Yates Baby 242' (*Daucus carota* L.) and tomato 'UC 82B' (*Lycopersicon esculentum* Mill.) seed germination was not significantly different after priming for 7 days at 25°C compared to 14 days at 15°C. Rivas et al. (1984) stated that surface-drying primed seed slowed germination rate of jalapeno peppers (*Capsicum annuum* L.) for all temperatures tested (5-35°C) compared to not surface drying. Additionally, Rivas et al. (1984) stated that both petri dish and media experiments were necessary to determine the optimum priming method. Quite often, conflicting results can be obtained from just one set of experimental protocols.

One limiting factor with field emergence is cool temperatures in early spring. Suboptimal temperatures have been correlated with a reduction in germination rate, total germination percentage, and seedling emergence (Kondra et al., 1983; Livingston and de Jong, 1990; Blackshaw, 1991). Danneberger et al. (1992) found that primed perennial ryegrass (*Lolium perenne* L.) seed had a greater germination percentage and seedling root growth than control seed at suboptimum germination temperatures of 5, 10, and 15 °C. Priming was conducted by using PEG 8000 at -1.1 MPa for 48, 76, 96, and 168 hours. Jalapeno seed primed in a 3% KNO_3 solution germinated at 5 and 10 °C while untreated seed did not germinate (Rivas et al., 1984). Polyethylene glycol primed seed of four grass species (*Pseudoroegneria spicata* [Pursh] Löve, *Elymus lanceolatus* [Scribn. and

J.G. Smith] Gould, *Poa sandbergii* Vasey, and *Sitanion hystrix* [Nutt.] J.G. Smith) were unable to rapidly germinate at temperatures lower than optimum for control seed (Hardegree, 1996). Additionally, optimum water potential was determined to be either equal to or less negative than the water potential at which radicle emergence was prevented for the control. Temperature range used was 5-35°C, and water potentials tested were 0 to -2.5 MPa. Sosa-Coronel and Motes (1982) found using 6 $\mu g.mg^{-1}$ GA in aerated water columns improved germination time and uniformity in pepper (*Capsicum annuum* L.) plants. In fact, the maximum percent germination was reached in only 2 to 3 days compared with an average of 10 days for maximum germination percentage.

Commercial production of direct-seeded *T. erecta* plants would benefit from a priming method that enhances emergence and produces quicker and more uniform stand establishment. Objectives for this research were to evaluate the effect of field sowing date on unprimed *T. erecta* seed emergence, identify an effective priming method for *T. erecta* seed, and compare primed seed to unprimed seed in field emergence and stand establishment.

MATERIALS AND METHODS

Seed of three *T. erecta* experimental cultivars, 'A-975', 'E-1236', and 'I-822', (Goldsmith Seeds, Inc., Gilroy, Calif.) and one commercial cultivar, 'Orange Lady' ('OL') were examined for field emergence over a three-year period. Seed viability was determined by placing 50 seeds in a petri dish with a paper towel moistened with deionized water. Two petri dishes per cultivar were used. Room temperature was

maintained at 23-26°C with constant florescent light (80 $\mu mol.m^{-2}.s^{-1}$). Daily germination counts were made. Seeds were considered germinated after the radicle had penetrated the seed coat and was visible to the naked eye. Seeds were planted in raised beds at the Oklahoma Botanical Garden and Arboretum in Stillwater, Okla. (USDA climatic zone 6b-7a). Soil type was Norge Loam (fine-silty, mixed, thermic Udic Paleustolls) with soil pH near 6.5. Plots (replications) were 1.2 m across and 1.5 m long with 0.6 m between plots. Soil was watered as required to maintain field capacity using drip irrigation. Three sowing dates at 14-day intervals were used in 1998: 17 Apr., 1 May, and 15 May; four in 1999: 1 Apr., 15 Apr., 29 Apr., and 13 May; and four in 2000: 6 Apr., 20 Apr., 5 May, and 19 May. Rows were spaced 23 cm apart, and spacing between plants was 23 cm resulting in 20 planting locations per plot. Three seeds were planted by hand in each location. Number of emerged seedlings was marked daily using toothpicks, and the average daily soil temperature (average of 3 measurements) was recorded. Seedlings were considered emerged after plumule emergence and cotyledons were fully opened. Three weeks after the first seedlings emerged, plants were thinned to a 23 cm spacing. Parameters collected were days to first seedling emergence, emergence time [days to 50% emergence (T50)], emergence uniformity [days from 10% to 90% seedling emergence (T10-90)], and total emergence percentage. Emergence time was calculated using the method of Furutani et al. (1985). In 1998 and 1999, days from seeding to first flower harvest (3 or more mature flowers), flower number, and fresh flower mass per replication were recorded at each harvest. Mature flowers with outer petals reflexed were hand-picked at 18 weekly harvests (1998) or 6 bi-weekly harvests (1999). The experimental design was completely randomized with four replications.

In 2000, 'E-1236' and 'I-822' seeds were primed by two methods, wet and solid matrix. Unprimed seeds served as the control. Seed Dynamics, Inc. (Salinas, Calif.) developed the wet priming method using restricted methodology unavailable for publication. With the solid matrix primed method, 50 seed were placed in a 5 by 8 cm zip-loc polyethylene bag filled with calcined clay (Super Absorbent, 500 μm, pH 7.0, Balcones Mineral Corp., Flatonia, Texas) at a weight ratio of 1:5 seed:calcined clay. Calcined clay was baked at 77 °C 24 h prior to use to ensure it was not contaminated with pathogens. Bags were filled with deionized water at 10% volume by weight of the seeds plus calcined clay and mixed. Water potential (Ψ) at 10% moisture content was –2.5 MPa. Previous trials revealed priming with 10% moisture for 1 day yielded primed seed without radicle emergence (unpublished data). Bags were stored upright in an environmental growth chamber (Model 1-35LL, Percival Manufacturing Co., Boone, Iowa) for 1 day at 20 °C with cool white florescent lighting (20.7 $\mu mol.m^{-2}.s^{-1}$.). Temperature and light conditions used were specified by Association of Official Seed Analysts (1993) for *T. erecta* seed. After removal from the chamber, seed and calcined clay were sifted, and seeds were weighed. Seeds were surface-dried on paper towels for 24 h at 23 °C and weighed again. Wet and solid matrix primed seed were primed simultaneously and then stored with unprimed seed at 2 °C until planted in the field. Four replications were used for each priming treatment.

The relationship between Ψ and moisture content on a dry weight basis was determined for Super Absorbent. Water potential was measured with a chambered *in situ* pyschrometer (Merrill Specialty Equipment, Logan, Utah) and read with a Wescor HP-115 water potential system (Wescor, Inc., Logan, Utah). Equipment was calibrated

against KCl standards. Super Adsorbent water potential was determined three times with three replicates prior to analysis.

All data were analyzed using the general linear model procedure, trend analysis, Duncan's multiple test range, and an interaction least squares difference where applicable (SAS Institute, Inc., Cary, N.C.). Percent data were transformed using the arcsin procedure prior to statistical analysis. Correlation analysis was used to evaluate the relationship between average daily soil temperature with days to first emergence, T50, T10-90, and total emergence percentage.

RESULTS

Seed viability tests for experiments conducted in 1998, 1999, and 2000 are presented in Table 2.1.

1998 Season

Correlation analysis revealed an inverse relationship between average daily soil temperature and days to first emergence ($R^2 = 0.84$, $P \leq 0.0001$), T50 ($R^2 = 0.89$, $P \leq 0.0001$), and T10-90 ($R^2 = 0.45$, $P = 0.0014$), respectively. No correlation between temperature and total emergence percentage existed.

Days to first emergence. Delayed sowing linearly decreased days to first emergence (Table 2.2). Seeds sown 15 May emerged approximately 3 days faster than those sown on 17 Apr. Cultivar did not affect the number of days to first emergence, and no significant interactions existed.

Emergence time (T50). 'I-822' had the slowest T50 while T50 for the other

cultivars was similar (Table 2.2). Emergence time decreased curvilinearly with delayed sowing. No significant interactions existed.

Emergence uniformity (T10-90). Delayed sowing linearly decreased T10-90 with the 15 May sowing date 2 days shorter than 17 Apr. (Table 2.2). Cultivar did not affect T10-90, and no significant interactions existed.

Total emergence. Cultivar and sowing date interacted such that delayed sowing produced 36% and 29% higher total emergence percentages for 'I-822' and 'OL', respectively, compared with earlier sowing dates (data not presented). Total emergence percentages were the same for the second and third sowing dates with 'I-822' (76%) and 'OL' (86%). Total emergence for 'A-975' linearly decreased from 88% to 34% as sowing date progressed. Sowing date had no effect on 'E-1236' total emergence percentage.

Days to first flower harvest. 'I-822' had the most days (88) from sowing until the first flower harvest compared with the other cultivars (Table 2.3). 'OL' was the next longest with 79 days, and 'A-975' and 'E1236' were statistically similar at 70 days. Delayed sowing linearly decreased the number of days from sowing until the first flower harvest was produced (Table 2.3). No significant interactions existed.

Flower number. Cultivar and sowing date interacted such that 'A-975' total flower number linearly decreased from 21,751,000 to 9,121,000 flowers.ha^{-1} with delayed sowing (data not presented). Total flower numbers for the other treatments were statistically similar and averaged 12,151,000 flowers.ha^{-1} for the entire season.

Fresh flower mass. 'OL' produced the greatest mass of fresh flowers (36,900 kg.ha^{-1}), which is 25% higher than the next highest cultivar (Table 2.3). Values for the

other cultivars were statistically similar. Sowing date did not influence fresh flower mass, and no significant interactions existed.

1999 Season

A freeze occurred on 18 Apr. with the minimum air temperature reaching –1.7 °C. Approximately 20% of the emerged seedlings from the first sowing date, 1 Apr., were visibly affected with a dark, water-soaked appearance indicative of plasma membrane leakage. Dead seedlings were counted, but no difference occurred among the cultivars (data not presented).

Correlation analysis revealed an inverse relationship between average daily soil temperature and days to first emergence ($R^2 = 0.48$, $P \leq 0.0001$) and T50 ($R^2 = 0.44$, $P = 0.0003$), respectively. Correlations between temperature and T10-90 and total emergence percentage did not exist.

Days to first emergence. 'E-1236' had the slowest emergence period (7.9 days) while emergence of the other cultivars was statistically similar (Table 2.4). Sowing date created a curvilinear response with 29 Apr. having the shortest emergence period (5.3 days) and 1 Apr. the longest (8.2 days). No significant interactions existed.

Emergence time (T50). Sowing date had a curvilinear effect on T50 with the shortest T50 (7.3 days) on 29 Apr. and the longest on 1 Apr. (10.3) (Table 2.4). Cultivar did not influence emergence time, and no significant interactions existed.

Emergence uniformity (T10-90). Cultivar and sowing date interacted such that the shortest T10-90 (2.4 days) occurred with 'E-1236' sown on 1 Apr. and the largest (6.5 days) occurred with 'A-975' sown on 1 Apr. (Table 2.5). 'A-975' had a curvilinear response with the shortest T10-90 occurring on 29 Apr. (4.4 days) and the longest on 1

Apr. (6.5 days). Responses for the other treatments were statistically similar.

Total emergence. An interaction between cultivar and sowing date occurred affecting total emergence percentage (Table 2.5). Total emergence for 'OL' linearly increased with delayed sowing, but total emergence for 'A-975' and 'I-822' had curvilinear responses. 'E-1236' emergence was low with 16% emergence or lower.

Days to first flower harvest. Cultivar and sowing date interacted to affect days from sowing to first flower harvest (Table 2.6). Days to first flower harvest decreased curvilinearly with sowing date for 'A-975' and 'E-1236'. 'I-822' also had a curvilinear response such that the number of days to first flower decreased from the first sowing date (100 days) to the third sowing date (78 days) but increased slightly for the last sowing (81 days). 'OL' days to first flower harvest decreased linearly with delayed sowing. The fewest number of days until first flower harvest was produced by the 13 May sowing of 'E-1236' (65 days), and the longest period (104 days) resulted from the 1 Apr. sowing of 'A-975'.

Flower number. Sowing date curvilinearly affected flower number such that the greatest flower numbers were produced from the 15 Apr. sowing date (12,725,000 per ha) and the fewest (7,846,000 per ha) from 1 Apr. (Table 2.7). Cultivar did not affect flower number, and no significant interactions existed.

Fresh flower mass. Sowing date had a curvilinear effect on fresh flower mass such that plants sown on 29 Apr. had the greatest mass (52,900 $kg.ha^{-1}$), and plants sown on 1 Apr. had the least (33,900 $kg.ha^{-1}$) (Table 2.7). Cultivar did not affect fresh flower mass, and no significant interactions existed.

2000 Season

Correlation analysis revealed an inverse relationship between average daily soil temperature and days to first emergence ($R^2 = 0.29$, $P = 0.0051$), T50 ($R^2 = 0.42$, $P \leq 0.0001$), and total emergence percentage ($R^2 = 0.22$, $P = 0.0315$), respectively. A correlation did not exist between temperature and T10-90.

Days to first emergence. Priming method and sowing date interacted to affect the number of days to first emergence (Table 2.8). Seed for each priming method had a curvilinear response such that the smallest number of days for emergence (4.0) occurred on the third sowing date, 5 May, and the largest (4.9 to 5.0) on the first sowing date, 6 Apr. Wet primed seed also had 4.0 days to emergence on the second sowing date, 20 Apr. Cultivar did not influence the number of days to first emergence.

Emergence time (T50). An interaction occurred between priming method and sowing date to affect T50 such that the fastest T50 (4.8 days) occurred with wet primed seed sown on 5 May, and the slowest T50 occurred with solid matrix primed seed sown on 6 Apr. (7.9 days) (Table 2.8). Each priming method had a curvilinear response such that the third sowing date, 5 May, had the smallest T50, but the longest period occurred with the first sowing date, 6 Apr. Cultivar did not influence T50.

Emergence uniformity (T10-90). Priming method interacted with sowing date. Unprimed seed had a curvilinear response with the shortest T10-90 (2.8 days) occurring for the 19 May sowing date, but the longest T10-90 for the 20 Apr. sowing date (Table 2.8). The solid matrix priming method linearly decreased T10-90 with delayed sowing, but T10-90 for wet primed seed did not significantly differ among sowing dates. Cultivar did not affect T10-90.

Total emergence. A three-way interaction occurred among cultivar, priming method, and sowing date for the total emergence percentage (Table 2.9). Wet priming affected both 'E-1236' and 'I-822' seed such that there was a linear and curvilinear decrease, respectively, in the total emergence percentage with delayed sowing. Total emergence percentage for the first sowing date, 6 Apr., was 86% and 94% for 'E-1236' and 'I-822', respectively. No difference existed between solid matrix primed and unprimed seed.

DISCUSSION

1998 Season

Delayed sowing linearly decreased both the number of days to first emergence and T10-90 (Table 2.2). Sowing *T. erecta* seed on 15 May resulted in the shortest period to first emergence (4.1 days) and also provided more uniform emergence (3.9 days) than the other treatments. Cultivar influenced T50; for example, 'I-822' reached peak emergence 8.4 days after sowing. Other cultivars had a quicker emergence time reaching their peak emergence at approximately 7.4 days. While T50 decreased with delayed sowing, the response leveled off at the third sowing date. As would be expected, delayed sowing with warmer soil temperatures had higher total emergence percentages than earlier sowing dates for 'I-822' and 'OL'. Interestingly, total emergence percentage remained the same for both the second and third sowing dates of 'I-822' (76%) and 'OL' (86%). The warmer soil temperatures corresponding with the third sowing date did not result in higher emergence percentages. 'A-975' had the highest emergence percentage (88%) of all treatments on 1 Apr. Subsequent sowing dates decreased emergence percentages from 71% down to 34%. Apparently, 'A-975' may germinate and emerge

better at cooler temperatures, which would be a valuable asset for commercial cultivar development of *T. erecta*.

'I-822' had the longest period from sowing until the first flower harvest (88 days) (Table 2.3). Both 'A-975' and 'E-1236' produced their first flower harvests 18 days earlier than 'I-822', which could result in a substantial difference in commercial production. Sowing on 15 May also produced the first flower harvest 9 days and 18 days earlier than sowing on 1 May and 17 Apr., respectively. Favorable environmental conditions in late spring allowed plants that were sown late to produce a crop in fewer days than those sown early. Flower number over the entire season did not differ among treatments except that 'A-975' had a linear decrease (21,783,000 to 13,427,000 to 9,121,000 flowers.ha^{-1}) with delayed sowing (data not presented), which was probably due to decreased 'A-975' emergence percentage at later sowing dates. 'OL' produced the highest fresh flower mass (36,900 kg.ha^{-1}), which is higher than reported by Baldwin et al. (1993) for their top-producer, 'Toreador' (32,200 kg.ha^{-1}). Interestingly, the latest sowing date, 15 May, produced the earliest flower harvest, but statistical analysis showed no difference among sowing dates for fresh flower mass (Table 2.3).

1999 Season

'E-1236' was slower to emerge (7.9 days) than the other cultivars (approximately 6.3 days) (Table 2.4). The curvilinear response of sowing date for days to first emergence and T50 indicate that 29 Apr. was the optimum sowing date with 5.3 days until first emergence and T50 at 7.3 days. While temperatures for 1 Apr. and 15 Apr. sowing were most likely too cool, 13 May temperatures may have been too warm for optimum emergence. 'E-1236' had the shortest T10-90 on 1 Apr. with 2.4 days, but total

emergence percentage was an unacceptable 4%. 'E-1236' seed quality may have been the problem as the emergence percentage was below 16% for all sowing dates and seed viability was only 51% (Table 2.1). After record examination, Goldsmith Seeds, Inc. reported that 'E-1236' seed supplied for 1999 was from the previous year's seed lot, which also had low viability (53%) (Table 2.1). Total emergence percentages for all cultivars were lower than a desirable 85-90% indicating a need for development of a suitable priming method (Table 2.5). The highest emergence percentages correlated with the last two sowing dates for 'A-975', 'E-1236', and 'OL' when temperatures were warm. Highest emergence percentages for 'I-822' occurred with the second and third sowing dates. In 1998, 'A-975' germinated better at cooler soil temperatures than warm soil temperatures, but this was not apparent in 1999.

As in 1998, 'I-822' and 'OL' had more days to first harvestable flower than 'A-975' and 'E-1236' (Table 2.6), which is most evident from the 13 May sowing date for all cultivars: 'I-822' (81 days), 'OL' (73 days), 'A-975' (68 days) and 'E-1236' (65 days). The last sowing date had the fewest number of days to first harvestable flower for all cultivars, excluding 'I-822', when compared to previous sowing dates. Again, favorable environmental conditions (higher soil and air temperatures and light intensities) allowed plants sown late to reach first harvest in fewer days than plants previously sown. Sowing on 15 and 29 Apr. produced a higher flower number and greater fresh flower mass than sowing on either 1 Apr. or 13 May (Table 2.7). Greater flower quantities and mass indicate 15 and 29 Apr. were the optimum times for direct sowing *T. erecta* for 'E-1236', 'I-822', and 'OL'. Cultivar was not significant for fresh flower mass, and 1999 quantities exceeded those for 1998 and values reported in Baldwin et al. (1993) by 26 to

30%.

2000 Season

Regardless of priming method, the shortest number of days to emergence (4.0 days) occurred with the third sowing date, 5 May (Table 2.8). Wet primed seed also emerged 4.0 days from sowing on 20 Apr., and solid matrix primed seed emerged 4.1 days after sowing on 20 Apr. Neither priming method proved to be advantageous over unprimed seed regarding days to first emergence for any sowing date.

Parera and Cantliffe (1994) stated priming's advantage is most evident when examining emergence uniformity under adverse environmental conditions. In our study, primed seed was advantageous when soil temperatures were cool. The T50 for the 6 Apr. sowing date was 6.8, 7.7, and 7.9 for the wet primed, unprimed, and solid matrix primed seed, respectively (Table 2.8). Under cool temperatures, wet priming resulted in a faster emergence time than either solid matrix primed or unprimed seed. Likewise, on the 20 Apr. sowing date, T50 was 5.8 and 5.9 for wet and solid matrix primed seed while T50 was still 7.2 days for unprimed seed. At slightly warmer temperatures, the solid matrix primed seed reached T50 as fast as the wet primed seed. The T50 was comparable for all methods at the last two sowing dates. The wet primed seed had more uniform emergence than unprimed or solid matrix primed seed for the first three sowing dates, but T10-90 was similar for all treatments for the last sowing date.

Enhanced emergence under adverse conditions was most evident with total emergence percentage. Wet primed seed had approximately 30% higher total emergence at early sowing dates with both cultivars than solid matrix or unprimed seed (Table 2.9). Solid matrix priming was not useful since no difference occurred between solid matrix

primed and unprimed seed. Beckman et al. (1993) found no difference in seedling field emergence between untreated big bluestem (*Andropogon gerardii* Vitman) seed and seeds exposed to 48-h solid matrix priming (-6 MPa, 17 °C) while examining. Tall fescue (*Festuca arundinacea* Schreb.) seed primed in vermiculite (-1.5 MPa, 20 °C, 4 days) increased germination rate by 53% but did not affect emergence uniformity or final emergence percentage compared to unprimed seed (Pill et al., 1997).

CONCLUSIONS

Tagetes erecta cultivar differences were evident in emergence parameters and flower harvest data for each of three years examined, but results were inconsistent from year to year. In 1998, 'I-822' had the slowest emergence time, but in 1999 it was statistically similar to the other cultivars. 'A-975' emergence percentage in 1998 was high for the earliest sowing dates and decreased with later dates. However, this trend was not apparent in 1999. Regardless, 'A-975's apparent affinity for cooler temperatures should be further examined for plant breeding application of direct-seeded *T. erecta* plants.

Early sowing delayed first seedling emergence and emergence time. The last two weeks of Apr. were optimum for direct-seeding *T. erecta* in Oklahoma for both emergence parameters and total flower yield.

Wet priming was superior to solid matrix priming and unprimed seed for emergence time, emergence uniformity, and total emergence percentage. Days to first emergence was similar for all three priming methods and sowing dates. Further research

should be conducted with wet priming methods, i.e., ionic salt solutions or PEG to improve on the promising results obtained here. More trials should also be conducted with solid matrix priming to see if more favorable results can be obtained.

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Table 2.1. *Tagetes erecta* seed viability for experiments conducted in 1998, 1999, and 2000. Values shown are mean of two 50-seed repetitions.

Year	Cultivar	Priming method	Viability (%)
1998	A-975	Unprimed	96
	E-1236	Unprimed	53
	I-822	Unprimed	77
	Orange Lady	Unprimed	73
Significance			
Cultivar			NS
1999	A-975	Unprimed	82 a [z]
	E-1236	Unprimed	51 b
	I-822	Unprimed	88 a
	Orange Lady	Unprimed	73 a
Significance			
Cultivar			0.0348
2000	E-1236	Unprimed	89 b
		Wet	96 b
		Solid matrix	90 b
	I-822	Unprimed	100 a
		Wet	100 a
		Solid matrix	100 a
Significance			
Cultivar (C)			0.0105
Priming (P)			NS

Table 2.1. Continued

Year	Cultivar	Priming method	Viability (%)
C*P			NS

[z] Mean separation within column by Duncan's multiple range test. Means followed by the same letter are not significantly different at $P \geq 0.05$.

Table 2.2. Effect of cultivar and three sowing dates for *Tagetes erecta* seedling emergence. Values shown are means of four repetitions (1998).

Treatment	Days to first emergence	Emergence time (T50) (days)	Emergence uniformity (T10-90) (days)
		Cultivar	
A-975	5.4 [NS]	7.4 b [z]	4.2 [NS]
E-1236	5.8	7.7 b	4.2
I-822	5.9	8.4 a	5.6
Orange Lady	5.3	7.1 b	4.5
		Sowing date	
17 Apr.	7.2	10.3	6.0
1 May	5.6	7.1	4.0
15 May	4.1	5.7	3.9
Significance			
Linear	<0.0001	<0.0001	0.0028
Quadratic	NS	0.0008	NS

[NS] Nonsignificant or significant at $P \geq 0.05$.

[z] Mean separation within columns by Duncan's multiple range test. Means followed by the same letter are not significantly different at $P \geq 0.05$.

Table 2.3. Effect of cultivar and three sowing dates for *Tagetes erecta* flower harvest. Values shown are means of four repetitions for 18 weekly harvests (1998).

Treatment	Sowing to first flower harvest (days)	Fresh flower mass ($kg.ha^{-1}$)
	Cultivar	
A-975	70 c [z]	27,800 b
E-1236	70 c	27,900 b
I-822	88 a	19,900 b
Orange Lady	79 b	36,900 a
	Sowing date	
17 Apr.	86	31,900
1 May	77	28,300
15 May	68	24,200
Significance		
Linear	<0.0001	NS
Quadratic	NS	NS

[NS] Nonsignificant or significant at $P \geq 0.05$.
[z] Mean separation within columns by Duncan's multiple range test. Means followed by the same letter are not significantly different at $P \geq 0.05$.

Table 2.4. Effect of cultivar and four sowing dates for *Tagetes erecta* days to first seedling emergence and emergence time. Values shown are means of four repetitions (1999).

Treatment	Days to first emergence	Emergence time (T50) (days)
	Cultivar	
A-975	6.1 b [z]	8.5 [NS]
E-1236	7.9 a	9.3
I-822	6.4 b	8.6
Orange Lady	6.4 b	8.7
	Sowing date	
1 Apr.	8.2	10.3
15 Apr.	7.8	9.7
29 Apr.	5.3	7.3
13 May	5.6	8.0
Significance		
Linear	<0.0001	<0.0001
Quadratic	NS	0.0173
Cubic	0.0006	0.0001

[NS] Nonsignificant or significant at $P \geq 0.05$.
[z] Mean separation within columns by Duncan's multiple range test. Means followed by the same letter are not significantly different at $P \geq 0.05$.

Table 2.5. Effect of cultivar and four sowing dates for *Tagetes erecta* seedling emergence uniformity and total germination percentage. Values shown are means of four repetitions (1999).

Cultivar	Sowing date	Emergence uniformity (T10-90) (days)	Total emergence (%)
A-975 (A)	1 Apr.	6.5	30
	15 Apr.	4.6	47
	29 Apr.	4.4	63
	13 May	6.0	47
E-1236 (E)	1 Apr.	2.4	4
	15 Apr.	2.9	13
	29 Apr.	3.6	16
	13 May	5.6	13
I-822 (I)	1 Apr.	5.2	27
	15 Apr.	4.9	59
	29 Apr.	5.4	57
	13 May	4.9	44
Orange Lady (OL)	1 Apr.	6.4	24
	15 Apr.	4.1	45
	29 Apr.	5.2	59
	13 May	5.0	68
Significance			
Cultivar		0.0045	<0.0001
Sowing date (S)			
Linear (L)		NS	<0.0001

Table 2.5. Continued.

Cultivar	Sowing date	Emergence uniformity (T10-90) (days)	Total emergence (%)
Quadratic (Q)		0.0103	<0.0001
Cubic (C)		NS	NS
A*SL		NS	**
A*SQ		**	***
A*SC		NS	NS
E*SL		***	NS
E*SQ		NS	NS
E*SC		NS	NS
I*SL		NS	NS
I*SQ		NS	***
I*SC		NS	NS
OL*SL		NS	***
OL*SQ		NS	NS
OL*SC		NS	NS

NS, **, *** Nonsignificant or significant at $P \geq 0.05$ or 0.01, respectively.

Table 2.6. Effect of cultivar and four sowing dates on days from sowing until first flower of *Tagetes erecta*. Values shown are means of four repetitions (1999).

Cultivar	Sowing date	Sowing to first flower harvest (days)
A-975 (A)	1 Apr.	104
	15 Apr.	78
	29 Apr.	71
	13 May	68
E-1236 (E)	1 Apr.	-- [z]
	15 Apr.	87
	29 Apr.	87
	13 May	65
I-822 (I)	1 Apr.	100
	15 Apr.	82
	29 Apr.	78
	13 May	81
Orange Lady (OL)	1 Apr.	102
	15 Apr.	92
	29 Apr.	78
	13 May	73
Significance		
Cultivar		NS
Sowing date (S)		
Linear (L)		<0.0001
Quadratic (Q)		0.0120

Table 2.6. Continued.

Cultivar	Sowing date	Days to first flower
Cubic (C)		NS
A*SL		***
A*SQ		***
A*SC		NS
E*SL		***
E*SQ		**
E*SC		-- [z]
I*SL		***
I*SQ		***
I*SC		NS
OL*SL		***
OL*SQ		NS
OL*SC		NS

[NS], **, *** Nonsignificant or significant at $P \geq 0.05$ or 0.01, respectively.
[z] Only three repetitions had emerged seedlings.

Table 2.7. Effect of four sowing dates on *Tagetes erecta* flower harvest. Values shown are means of four repetitions for six bi-weekly harvests (1999).

Sowing date	Flower no. (flowers.ha^{-1})	Fresh flower mass (kg/ha)
1 Apr.	7,846,000	33,900
15 Apr.	12,725,000	52,500
29 Apr.	12,342,000	52,900
13 May	8,292,000	35,900
Significance		
Linear	NS	NS
Quadratic	0.0002	0.0003
Cubic	NS	NS

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

Table 2.8. Effect of priming method and sowing date on *Tagetes erecta* seedling emergence. Values shown are means of four repetitions (2000).

Priming method	Sowing date	Days to first emergence	Emergence time (T50) (days)	Emergence uniformity (T10-90) (days)
Control (CT)	6 Apr.	5.0	7.7	4.9
	20 Apr.	4.6	7.2	7.0
	5 May	4.0	5.1	3.7
	19 May	4.3	5.6	2.8
Wet (W)	6 Apr.	4.9	6.8	4.1
	20 Apr.	4.0	5.8	3.9
	5 May	4.0	4.8	2.8
	19 May	4.6	5.7	3.0
Solid-matrix (SM)	6 Apr.	5.0	7.9	4.8
	20 Apr.	4.1	5.9	4.0
	5 May	4.0	5.5	4.8
	19 May	4.6	5.9	3.0
Significance				
Priming		NS	<0.0001	0.0090
Sowing date				
Linear (L)		<0.0001	<0.0001	<0.0001
Quadratic (Q)		<0.0001	<0.0001	NS
Cubic (C)		NS	0.0011	NS
CT*SL		***	***	***
CT*SQ		**	**	***

Table 2.8. Continued.

Priming method	Sowing date	Days to first emergence	Emergence time (T50) (days)	Emergence uniformity (T10-90) (days)
CT*SC		NS	***	***
W*SL		NS	***	NS
W*SQ		***	***	NS
W*SC		NS	NS	NS
SM*SL		**	***	**
SM*SQ		***	***	NS
SM*SC		NS	NS	NS

NS, **, *** Nonsignificant or significant at $P \geq 0.05$ or 0.01, respectively.

Table 2.9. Effect of cultivar, priming method, and sowing date on *Tagetes erecta* total seedling emergence percentage. Values shown are means of four repetitions (2000).

Cultivar	Priming method	Sowing date	Total emergence (%)
E-1236 (E)	Control (CT)	6 Apr.	68
		20 Apr.	59
		5 May	64
		19 May	58
	Solid matrix (SM)	6 Apr.	63
		20 Apr.	71
		5 May	66
		19 May	62
	Wet (W)	6 Apr.	86
		20 Apr.	93
		5 May	68
		19 May	60
I-822 (I)	Control	6 Apr.	67
		20 Apr.	70
		5 May	74
		19 May	78
	Solid matrix	6 Apr.	73
		20 Apr.	83
		5 May	74
		19 May	59
	Wet	6 Apr.	94

Table 2.9. Continued.

Cultivar	Priming method	Sowing date	Total emergence (%)
		20 Apr.	72
		5 May	67
		19 May	63
Significance			
Cultivar			0.0152
Priming			0.0092
Sowing date (S)			
Linear (L)			0.0002
Quadratic (Q)			NS
Cubic (C)			NS
E*CT*SL			NS
E*CT*SQ			NS
E*CT*SC			NS
E*SM*SL			NS
E*SM*SQ			NS
E*SM*SC			NS
E*W*SL			***
E*W*SQ			NS
E*W*SC			NS
I*CT*SL			NS
I*CT*SQ			NS

Table 2.9. Continued.

Cultivar	Priming method	Sowing date	Total emergence (%)
I*CT*SC			NS
I*SM*SL			NS
I*SM*SQ			NS
I*SM*SC			NS
I*W*SL			***
I*W*SQ			***
I*W*SC			NS

NS, **, *** Nonsignificant or significant at $P \geq 0.05$ or 0.01, respectively.

CHAPTER III

OPTIMIZING MARIGOLD (*TAGETES ERECTA* L.) PETAL AND PIGMENT YIELD

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Additional index words. African marigold, lutein, xanthophylls, transplants, direct seeded, ammonium nitrate, hand harvest, mechanical harvest

ABSTRACT

African marigold (*Tagetes erecta* L.) flower pigments can be extracted and used as a natural food colorant imparting an orange color to egg yolks and poultry skin. We examined four cultivars of marigold for their ability to be commercially grown and mechanically harvested over a four year period. 'E-1236' was consistently a top producer for three seasons in terms of flower number, flower diameter, plant and flower canopy height, plant stand, and fresh flower, dried flower, and dried petal mass. 'E-1236' produced the greatest quantity of lutein, a carotenoid pigment, in 1998 (22,000 $g.ha^{-1}$) using a spectrophotometer for quantification. In 1999, 'E-1236' and 'Orange Lady' both produced the greatest quantities (21,000 $g.ha^{-1}$). Using transplants rather than direct-seeded plants resulted in two more harvests in a single season and in greater lutein production by transplants. One mid-season ammonium nitrate application (4.6 $kg.ha^{-1}$) in 1998 produced larger flower diameters with direct-seeded plants. In 1999, four monthly

nitrogen applications reduced mass of dried receptacles and interacted with cultivar and establishment method to affect plant stand, flower diameter, and plant canopy. Plants were hedge-trimmed in 1999 to mimic mechanical harvesting resulting in a 45-55% reduction in flower harvest data compared to hand-harvested flowers.

INTRODUCTION

African marigold (*Tagetes erecta* L.) petals are commercially valuable as a natural source of xanthophyll pigments (yellow-orange pigments). The poultry industry uses them primarily as feed additives to impart an orange color to egg yolks and a yellowish tinge to poultry skin. Over the years, consumers have equated orange egg yolks with healthiness; colorless bland products are undesirable to the consumer. These pigments must be included as a feed additive if the final product is to attain the desired color since birds lack the ability to synthesize them. Thus, the pigments are absorbed through a diet that includes xanthophyll supplementation (Marusich and Bauernfeind, 1981). Xanthophyll addition costs $5 to $15 per ton of poultry feed (Williams, 1992). Lutein and zeaxanthin are the main xanthophyll pigments present in egg yolks due to their highly absorptive nature with fatty tissue (Karunajeeva et al., 1984). Yellow corn, alfalfa, and marigold can serve as natural sources of xanthophylls. Marigold has been most commonly used by the poultry industry to augment the xanthophyll present in corn and alfalfa feed standardizing the feed's xanthophyll content (Delgado-Vargas et al., 1998).

Xanthophylls are only one of two classes of carotenoid pigments. Carotenes are the other class of carotenoids and have a general structure of $C_{40}H_{56}$ (Gross, 1987). β-carotene is a well known carotene used as a pigment in milk fat (Williams, 1992). Xanthophylls are the oxygenated derivatives of carotenes and, consequently, are more unstable and sensitive to deterioration. Direct sunlight, high heat, oxygen, and acids may damage these pigments, resulting in low quantitative pigment recovery through loss of particularly unstable pigments or conversion to other carotenoids that would otherwise not be present (Davies, 1976). Lutein ($C_{40}H_{56}O_2$) is the primary xanthophyll pigment that produces the orange color in *T. erecta* flowers. Within the petals, lutein is esterified with one or two fatty acids and composes roughly 90% of the petals' identified pigments (Quackenbush and Miller, 1972).

Carotenoids are present in plastids, particularly chloroplasts and chromoplasts. While chloroplasts are present in all photosynthetic tissue, especially leaves and unripe fruit, chromoplasts are found primarily in flowers and ripe fruit. Lutein is the predominant carotenoid pigment, typically comprising 40 to 50% of the total carotenoids present in chloroplasts (Gross, 1987). Chromoplast structure in flowers consists of multiple concentric membranes and is categorized as "membranous" chromoplast (Gross, 1987).

Carotenoids are beneficial as a natural pigment source and have many commercial applications. Forty carotenoids are known to be Vitamin A precursors in mammals with β-carotene as the most well known example. The medical community has even recognized the benefits of carotenoid pigments in humans for their ability to negate the damaging effects of singlet oxygen. Carotenoid pigments have shown positive benefits

in slowing the growth of induced skin tumors, treating dermatological diseases, and lowering the overall risk of cancer in human beings (Mathews-Roth, 1982). Lutein serves special pharmacological use as an ophthalmologic ointment with the trade name Adaptionol® (Gau et al., 1983). Thus, the potential for broad commercial use of carotenoids should generate further interest in *T. erecta* as an alternative crop.

Baldwin et al. (1993) conducted an eight year study in which five *T. erecta* cultivars were grown in Virginia and Mexico for pigment extraction and lesion-nematode reduction. All plants were direct-seeded, and flowers were hand-harvested. Highest xanthophyll yields were achieved by 'Toreador' (31.20 $kg.ha^{-1}$). They noted plant "vigor" decreased as the season progressed due to nitrogen deficiency. In the following season, the addition of (4.6 $kg.ha^{-1}$) of ammonium nitrate after the first two harvests improved the following harvests. Pigment yield increased most after three nitrogen applications in a single season. The authors concluded that the two greatest problems for marigold production were plant stand establishment and weed control. They felt an active weed control program from the time of planting was necessary because marigolds were unable to compete with invasive plants. Plant stand establishment was unsuccessful unless large quantities of de-tailed seed were used in the precision seeder equipment. Otherwise, repeat plantings were necessary to achieve adequate stand establishment.

Plant stand establishment is always an important consideration in commercial production. Factors that effect stand establishment are soil crusting, poor seed to soil contact, extremely high or low temperatures, pathogen invasion, and soil moisture (Bennett et al., 1992). Direct-seeded plants are vulnerable to many of these factors and consequently often have uneven germination, slow emergence, and poor stand

establishment (Wurr and Fellows, 1983). Direct-seeding is less expensive initially so most growers use direct-seeding in field operations. However, unless a precision seeder is used to plant the seed, additional labor is required to thin seedlings to the desired spacing. While transplants are initially more expensive to grow and set in the field, they can be planted at the exact space desired for the crop and have the benefit of quickly producing a uniform stand allowing earlier harvests. Transplanted plants often produce their first harvestable crop of flowers before direct-seeded plants begin to flower.

Currently, *T. erecta* plants are grown for pigment production in Mexico, Peru, and India. The flowers are hand-picked, stored, dried, and processed into a pelletized form for pigment extraction. Several problems with this process lower the pigment extraction quality and yield. After harvest, flowers may be stored outside unprotected for days or weeks before drying, exposing the pigment to damaging heat, light, and mold growth. High air temperatures during the drying process also reduce pigment quality. During pelletization, seeds and other green material may be included which lowers the product's purity (Buser, 1997). Hence, a more efficient program must be developed for harvesting and processing lutein.

The key to successful commercial *T. erecta* production in the southern plains will be to select a cultivar that can withstand the environmental conditions typical for the area, including wind, drought, and heat stress. The cultivar should consistently produce a large number of orange flowers that do not vary in the degree of color and be relatively unaffected by pests and diseases. Also, the plant must withstand the destruction a mechanical harvester can inflict on plants after multiple harvests. Although Baldwin et al. (1993) investigated commercial African marigold production, they did not examine

cultivar suitability for mechanical harvest. The objectives of this project are to determine which cultivar and production methods would be most suitable for commercial *T. erecta* production using mechanical harvest and provide the greatest pigment yield.

MATERIALS AND METHODS

Four *T. erecta* cultivars, 'A-975', 'E-1236', 'I-822', and 'X-986', developed by Goldsmith Seeds, Inc. (Gilroy, Calif.) and one commercial cultivar, 'Orange Lady' ('OL') were evaluated over a four-year period. In 1996, plants were grown in raised beds at the Oklahoma Botanical Garden and Arboretum in Stillwater, Okla. (USDA climatic zone 6b-7a). Soil type was Norge Loam (fine-silty, mixed, thermic Udic Paleustolls) with soil pH near 6.5. Plots were watered as required to maintain field capacity using drip irrigation. In 1997, 1998, and 1999, plants were grown in Hinton, Okla. (USDA climatic zone 7a) at S. S. Farms. Soil type was Pond Creek loam (deep, fine sandy loam) with soil pH near 6.5. Plots were watered as required to maintain field capacity using sprinkler irrigation.

1996 Season

'OL' was evaluated both as direct-seeded plants and transplants as establishment methods. Plots (repetitions) were 3.7 m long and 1.2 m wide. Rows were spaced 31 cm apart with three different spacings, 10 cm, 23 cm, and 36 cm, used within the rows. Six harvests were made over the entire season. Mature flowers with outer petals reflexed were hand-picked approximately every two weeks. Data collected included the number of flowers produced per repetition and fresh flower, dried flower, dried petal, and dried

receptacle mass. Fresh flowers were dried for 24 h using a temperature of 49 to 52 °C. Soil was analyzed and amended as needed for nutrient content (Baldwin et al., 1993). Experimental design was a completely randomized design with four repetitions.

1997 Season

'A-975', 'E-1236', 'I-822', 'OL', and 'X-986' were evaluated as transplants only at the 23 cm within-row spacing in rows spaced 31 cm apart. Plots (repetitions) were 1.2 m wide and 1.5 m long allowing 0.6 m between plots. Nine harvests were made over the season. Flowers were dried using air heated to 66 °C and then blown through the flowers suspended on a wire mesh for 3 to 4 h. Flowers were considered dry when the petals were brittle, but receptacles remained flexible. Petal moisture content was determined using the Association of Official Analytical Chemists (AOAC) method (1984). All other procedures were the same as in 1996.

1998 Season

'A-975', 'E-1236', 'I-822', and 'OL' were evaluated both as direct-seeded plants and transplants. Transplant flats were started on 10 April, and seedlings were field-planted on 11 May along with direct-seeded plants (3 seeds per hole). Plots (repetitions) were 1.2 m wide and 1.5 m long. Rows were spaced 23 cm apart, and plants were spaced 23 cm within each row. A pre-plant soil analysis was made, and soil was amended with 4.6 $kg.ha^{-1}$ of ammonium nitrate prior to planting. An additional nitrogen application was made mid-season (20 Aug.) in the form of ammonium nitrate at the rate of 4.6 $kg.ha^{-1}$. Eleven harvests were made throughout the season. The same data were collected as in 1996 with the addition of lutein pigment analysis. The Association of Official Analytical Chemists Method 43.018 (1984) which requires a spectrophotometer (Shimadzu UV

160U, UV-visible recording, Shimadzu Corp., Kyoto, Japan), was used to estimate lutein quantity in the petal material. Appendix A contains detailed information about sample handling, processing, and pigment analysis. Additionally, flower diameter, plant and flower height, and plant stand were recorded each harvest. Plant and flower canopy height were measured as the distance from the ground to the uppermost leaves and the base of the flower receptacles, respectively. The experimental design was a randomized complete block with four repetitions. Plots were blocked according to nitrogen application.

1999 Season

The same cultivars, establishment methods, plot size, spacing, and data were used as in the 1998 season. Transplant flats were started on 16 Apr., and seedlings were field-planted on 13 May along with direct-seeded plants (3 seeds per hole). Nitrogen was applied at 4.6 $kg.ha^{-1}$ monthly for four months on 23 Jun., 26 Jul., 26 Aug., and 22 Sep. Plots were either hand-harvested or cut at the flower canopy height specific for each repetition using a hedge-trimmer, intended to mimic action of a mechanical harvester. Eight harvests were made on hand-harvested plants, and hedge-trimmed plants were harvested five times. Three weeks between harvests were required to produce mature flowers with hedge-trimming while only 2 weeks were required for hand-harvesting. Experimental design was a split-split plot with four repetitions. Nitrogen application was the main plot with harvest method as the sub-plot. Cultivar and establishment method were equally randomized as the sub-sub plots.

Within each repetition, data were collected only from interior plants. Flowers from border plants were harvested but not collected to eliminate edge effect. Data were

subjected to a general linear model procedure, trend analysis, Duncan's multiple range test, and an interaction least squares difference where applicable (SAS Institute, Inc., Cary, N.C.). Percent data were transformed using the arcsin procedure prior to statistical analysis. Correlation analysis was used to evaluate the relationship between pigment concentration and petal moisture content, average daily air temperature, and average daily light intensity (MJ/m^2) in 1998 and 1999. Air temperature and light intensity data were provided courtesy of the Oklahoma Mesonet Project, a cooperative venture between Oklahoma State University (Stillwater, Okla.) and the University of Oklahoma (Norman, Okla.).

RESULTS

1996 Season

Transplants yielded 45 to 55% greater flower numbers and fresh flower, dried flower, dried petal, and dried receptacle mass than direct-seeded 'OL' plants (Table 3.1). Increasing plant spacing decreased flower numbers and fresh flower, dried flower, dried petal, and dried receptacle mass.

1997 Season

Cultivar 'E-1236' consistently yielded the greatest flower numbers and fresh flower, dried flower, and dried petal mass, but yields were not different from 'A-975' (Table 3.2). 'OL' had the largest amount of dried receptacles. 'X-986' was the poorest producer, yielding 47 to 64% less than 'E-1236' (Table 3.2).

1998 Season

Correlation analysis showed a significant relationship ($R^2 = 0.20$, $P = 0.0172$) between dried petal moisture content and pigment and also between average daily air temperature and pigment concentration ($R^2 = 0.26$, $P = 0.0013$). Correlation analysis between dried petal moisture content and pigment concentration was conducted to show whether over-drying of petal material resulted in pigment loss. Light intensity was also significantly correlated with pigment concentration ($R^2 = 0.19$, $P = 0.0197$).

Harvest data. 'E-1236' produced the greatest flower numbers and fresh flower, dried petal, and lutein pigment mass (22,000 $g.ha^{-1}$) (Table 3.3). 'OL' had the largest amount of dried receptacles but did not differ from 'E-1236'. Transplants yielded 59 to 69% higher amounts for these six parameters as compared to direct-seeded plants. Nitrogen application had no effect upon measured parameters, and no significant interactions existed.

Plant and flower canopy. Transplanted 'I-822', 'OL', and 'E-1236' plants had the tallest plant canopies (Table 3.4). Direct-seeded 'A-975' and 'E-1236' plants and transplanted 'A-975' plants had the shortest plant and flower canopies. The tallest flower canopies were produced by 'I-822' and 'OL' transplants. Nitrogen application did not affect plant canopy or flower canopy.

Flower diameter. Nitrogen application and establishment method interacted such that flower diameter was smallest (5.1 cm) for direct-seeded plants that did not receive nitrogen during the season but largest (5.7 cm) for direct-seeded plants that did receive nitrogen ($P = 0.0196$). Cultivar and harvest method did not affect flower diameter.

Plant stand. Cultivar and establishment method interacted such that transplanted 'A-975' and 'E-1236' and direct-seeded 'A-975' had the highest percentages of dead plants at mid-season (29%, 39%, and 37%, respectively) (Table 3.4). Interestingly, 'E-1236' transplants had the highest percentage of dead plants and direct-seeded 'E-1236' plants had the lowest percentage at mid-season. At the end of the season, cultivar was the only variable that affected the percentage of dead plants (data not presented). 'A-975' had a significantly higher percentage (55%) of dead plants than 'OL' (23%). 'E-1236' and 'I-822' produced intermediate percentages of dead plants, 43% and 33%, respectively.

1999 Season

Correlation analysis between pigment concentration and dried petal moisture content was significant ($R^2 = 0.21$, $P = 0.0319$), and pigment concentration and average daily air temperature were also significant ($R^2 = 0.26$, $P = 0.0100$). There was no correlation between pigment concentration and light intensity.

Flower number. A three-way interaction among cultivar, establishment method, and harvest method occurred (Table 3.5). Hand-harvested 'OL' transplants and direct-seeded 'E-1236' plants produced the highest number of flowers for the season (7,048,200 and 7,622,200 flowers/ha, respectively). 'I-822' as both transplants and direct-seeded plants produced the lowest number of flowers regardless of whether hand-harvesting or hedge-trimming was used.

Fresh flower mass. The interaction among cultivar, establishment method, and harvest method also affected fresh flower mass (Table 3.5). Hand-harvested treatments yielded a greater amount of fresh flowers than those that were hedge-trimmed. The

highest yields were achieved by hand-harvesting direct-seeded 'E-1236' plants and transplants of 'E-1236', 'A-975', and 'OL' (22,400; 20,800; 21,400; and 23,000 $kg.ha^{-1}$, respectively). The highest yields for plants that were hedge-trimmed were 10,600; 11,000; and 11,800 $kg.ha^{-1}$ for transplants of 'A-975', 'OL', and 'E-1236', respectively.

Dried petal mass. The same three-way interaction among cultivar, establishment method, and harvest method affected dried petal weight (Table 3.5). Hand-harvested treatments yielded greater dried flower masses than those that were hedge-trimmed. Both transplants and direct-seeded plants of 'E-1236' produced the greatest amount of dried petals (1,800 and 2,100 $kg.ha^{-1}$, respectively) when hand-harvested. For hedge-trimmed plants, 'A-975' (800 $kg.ha^{-1}$) and 'E-1236' (900 $kg.ha^{-1}$) transplants yielded the highest quantity of dried petal material.

Pigment mass. The three-way interaction occurred among cultivar, establishment method, and harvest method also affected pigment concentration (Table 3.5). Hand-harvested 'OL' transplants (21,300 $g.ha^{-1}$) and direct-seeded 'E-1236' plants (20,700 $g.ha^{-1}$) produced the highest yields. The lowest yields were obtained from all hedge-trimmed treatments except for the 'OL' transplants which produced the greatest yield within hedge-trimmed treatments.

Dried flower mass. 'OL' (3,900 $kg.ha^{-1}$) yielded significantly higher amounts of dried flowers as compared to 'I-822' (2,600 $kg.ha^{-1}$). 'E-1236' (3,800 $kg.ha^{-1}$) was similar to 'OL' and to 'A-975' (3,300 $kg.ha^{-1}$). Transplants produced more dried flowers than direct-seeded plants. Hand-harvesting yielded significantly higher amounts of dried flowers than hedge-trimming (Table 3.6). Nitrogen applications did not affect dried flower weight, and no significant interactions existed.

Dried receptacle mass. 'OL' (2,900 $kg.ha^{-1}$) yielded the most dried receptacles, and 'I-822' (1,700 $kg.ha^{-1}$) produced the least among the cultivars (Table 3.6). 'A-975' and 'E-1236' produced amounts that were intermediate to the other cultivars. Plants not receiving nitrogen application produced more dried receptacles than plants receiving nitrogen. Transplants also produced more dried receptacles than direct-seeded plants. Hand-harvesting produced more dried receptacles than hedge-trimming. No significant interactions existed.

Plant stand. At mid-season and the end of the season, transplants had a higher percentage of dead plants than direct-seeded plants (Table 3.6). Nitrogen addition increased the percentage of dead plants, and hedge-trimmed plants had a lower percentage of dead plants than hand-harvested plants. Cultivar did not influence plant stand, and no significant interactions existed.

Flower diameter. Two three-way interactions (cultivar, nitrogen application, and establishment method and nitrogen application, establishment method, and harvest method) occurred affecting flower diameter (Table 3.7). The first three-way interaction revealed that transplanted or direct-seeded 'I-822' plants with or without nitrogen had the largest flower diameters. Establishment method and nitrogen addition significantly affected diameter within a cultivar, but no single treatment caused flowers of one cultivar to be larger than the other. The second three-way interaction revealed that transplants yielded larger flower diameters than direct-seeded plants when hedge-trimming was used on plants that did not receive a nitrogen application. All other comparisons had equal flower diameters for the same nitrogen application and harvest method. Hedge-trimming plants reduced flower diameter compared to hand-harvesting the plants regardless of

establishment method or nitrogen application (Table 3.7). When nitrogen was not added, hedge-trimmed direct-seeded plants had significantly smaller flower diameters compared to the other treatments. Hedge-trimmed transplanted plants had similar flower diameters to hand-harvested plants. For plots that did receive nitrogen, all hand-harvested plants, regardless of establishment method, had larger flower diameters than those that were hedge-trimmed.

Plant canopy. An interaction among cultivar, establishment method, and nitrogen application affected plant canopy (Table 3.7). 'I-822' transplants with or without nitrogen application had the tallest plant canopies of all treatment combinations. 'A-975' transplants with or without nitrogen addition were the shortest.

Flower canopy. A three-way interaction among cultivar, establishment method, and harvest method affected flower canopy (Table 3.8). Direct-seeded 'I-822' plants had the tallest flower canopies of all treatment combinations regardless of harvest method. 'A-975' transplants had the shortest flower canopies regardless of harvest method.

DISCUSSION

In 1996, 'OL' transplants consistently produced approximately 45% to 55% greater amounts of all measured flower parameters than 'OL' direct-seeded plants (Table 3.1). Transplants often produce higher yields within a season than direct-seeded plants. Leskovar and Cantliffe (1993) found that within a single season transplants yielded 78% more in the first planting and 43% more in the second planting of bell pepper fruit (*Capsicum annuum* L.) compared with direct-seeded plants. However, Cooksey et al.

(1994) found that transplanted paprika pepper (*Capsicum annuum* L.) plants had higher fruit yields compared with direct-seeded plants in only one out of three years. Increasing marigold plant spacing decreased all of the measured parameters because plots with tighter spacing contained more plants than those with wide spacing. The number of plants per repetition was 60, 26, and 18 for 10 cm, 23 cm, and 36 cm spacings, respectively. While increased spacing increased flower number per plant from 29 for 10 cm spacing to 49 and 66 for 23 cm and 36 cm spacings, respectively, the increase per plant was not enough to offset the decline in plants per acre. Even though the 10 cm spacing had the highest yields, plants were short-lived and prone to insect damage and disease pressure (data not presented).

The 1997 cultivar comparison demonstrated that 'E-1236' and 'A-975' had high yields for flower number and fresh flower, dried flower, and dried petal mass making them possible candidates for commercial production (Table 3.2). 'I-822' and 'OL' also had acceptable yields. While 'OL' produced the highest dried receptacle mass, dried petal mass was 47% below that of 'E-1236'. 'X-986' had yields well below those for all other cultivars and was dropped in subsequent years (Table 3.2).

Results of 1998 were similar to those in 1997. 'E-1236' was the top producing cultivar for flower number and mass of fresh flowers, dried petals, and pigment (Table 3.3). The other cultivars were approximately equal for the measured parameters. 'OL' again had the largest amount of dried receptacles, and 'OL' dried petals composed only 50% of the 'E-1236' value. 'E-1236' pigment mass (22,000 $g.ha^{-1}$) was 31% greater than the next highest cultivar, 'I-822' (15,200 $g.ha^{-1}$). The amount produced by 'E-1236' was 29% less than the top producing *T. erecta* 'Toreador' (31,200 $g.ha^{-1}$) as reported by

Baldwin et al. (1993) but comparable to 20,300 $g.ha^{-1}$ produced by 'Xanthophyll'. Samples were shipped to a laboratory for analysis (methodology not specified).

Transplants yielded 59 to 69% greater quantities of the measured parameters than direct-seeded plants (Table 3.3). In 1998, transplants had two more harvests early in the season than direct-seeded plants (data not presented). Direct-seeded plants did not compensate with larger harvests at the end of the season as might be expected. Leskovar and Cantliffe (1993) also had higher yields with transplanted bell pepper plants and found that direct-seeded plants did not have a significant yield improvement at the end of the season.

Cultivar and establishment method interacted to affect plant and flower canopy height (Table 3.4). Apparently, height of naturally tall cultivars was enhanced by the transplanting treatment as compared to direct-seeded plants of the same cultivars. Plant and flower canopy height is an important consideration for mechanical harvesting as tall cultivars with a uniform flower canopy are more likely to produce more easily harvestable crops. Flower canopies of 'E-1236', 'I-822', and 'OL, were 6.1 cm, 5.9 cm, and 7.4 cm, respectively, above the plant canopy. The greater the disparity between flower and plant canopies, the more likely that a mechanical harvester could collect the flowers without severely damaging the plant's foliage and flower buds allowing more harvests in a single season. Plant and flower canopy heights for 'A-975' (30.0 and 34 cm, respectively) were too low for mechanical harvesting. Plant height of cultivars used in Baldwin et al. (1993) ranged from 35.0 cm to 96.9 cm.

The percentage of dead plants at mid-season was affected by cultivar and establishment method (Table 3.4). Interestingly, 'E-1236' had both the highest

percentage of dead plants (39%) as transplants and the lowest percentage (3%) as direct-seeded plants. This pattern was not evident among the other cultivars. The percentage of dead plants at mid-season was similar between direct-seeded (37%) and transplanted (29%) 'A-975' plants and 'E-1236' transplants. At the end of the season, cultivar was the only variable affecting the percentage of dead plants with 'A-975' having the highest percentage (55%) (data not presented). 'A-975' would not perform well in commercial production due to the small plant stature and since by the end of the season over half of the plants were dead.

Flower diameter was the only data parameter affected by nitrogen application in 1998. Nitrogen application caused direct-seeded plants' flower diameter to be larger (5.7 cm) than direct-seeded plants that did not receive nitrogen (5.1 cm) (data not presented). Flower diameter for transplanted plants was similar regardless of nitrogen application. Fresh flower and dried petal mass were unaffected by nitrogen.

A three-way interaction among cultivar, establishment method, and harvest method affected flower number and fresh flower mass in 1999 (Table 3.5). Hand-harvesting produced the largest flower number compared with hedge-trimming plants, but cultivar selection and establishment method were also critical factors. If transplanted plants were to be used with hand-harvesting, 'OL' would be the better choice, but 'E-1236' is the better cultivar for hand-harvested direct-seeded plants. Hand-harvesting was expected to recover greater flower numbers simply because hedge-trimming will miss flowers that are not in the same flower canopy plane. Regardless of which establishment or harvest method is used, 'I-822' would not be a good choice when maximum flower number is desired. Considering fresh flower mass, hand-harvesting yielded higher values

than hedge-trimming due to more frequent harvests (Table 3.5). For instance, transplanted 'E-1236' plants produced 43% more fresh flower mass with hand-harvesting than hedge-trimming. Direct-seeded 'E-1236' plants that were hand-harvested produced as well as transplants from 'A-975', 'E-1236', and 'OL' that were harvested under both harvest methods.

The interaction among cultivar, establishment method, and harvest method was evident for mass of dried petal and pigment. As previously stated, hand-harvesting yielded higher results than hedge-trimming, and 'E-1236' was a top producer for both harvest methods (Table 3.5). With hedge-trimming, transplants produced a larger mass of dried petals suggesting that transplants would be feasible with mechanical harvesting. Lutein pigment mass was probably the most important data parameter since it determines the feasibility of commercial production and mechanical harvesting. Thus, treatments that generate the greatest pigment mass will outweigh positive aspects of other treatments, such as larger flower number. Highest pigment yields resulted from hand-harvesting transplanted 'OL' and direct-seeded 'E-1236' plants (Table 3.5). All hedge-trimmed plants produced the lowest pigment amounts except for transplanted 'OL' (10,600 $g.ha^{-1}$) plants that were statistically similar with hand-harvested 'A-975' direct-seeded plants (9,500 $g.ha^{-1}$). Hedge-trimmed direct-seeded plants' pigment production was less than half of that reported by Baldwin et al. (1993) for hand-harvested direct-seeded plants.

'OL' produced the largest mass of dried flowers and dried receptacles compared with other cultivars, but as previously noted, dried receptacles composed 75% of the dried flower mass and dried petals only 25% (Table 3.6). Percent composition of the

other cultivars' dried flower mass by dried receptacle mass were 67% for 'A-975', 64% for 'E-1236', and 67% for 'I-822'. 'E-1236' produced a greater percentage of dried petals due to smaller receptacles, which was demonstrated with the dried petal parameter. Surprisingly, nitrogen addition significantly lowered mass of dried receptacles (Table 3.6). A corresponding increase in dried petals would explain this result, but nitrogen was not significant for any flower mass parameter. Transplanted plants produced 25% and 27% more mass of dried flowers and dried receptacles, respectively, than direct-seeded plants (Table 3.6), which is in agreement with Leskovar and Cantliffe (1993). Likewise, hand-harvested plants yielded 55% and 52% more dried flower and dried receptacle mass, respectively, than direct-seeded plants.

Plant stand was affected by establishment, nitrogen application, and harvest method in 1999 and not cultivar as in 1998 (Table 3.6). Interestingly, the treatments that produced the largest quantities of flower number and dried flower mass, hand-harvesting and transplants, had higher percentages of dead plants at mid-season and end of the season compared with hedge-trimmed and direct-seeded plants, respectively. One would assume that hedge-trimming would cause more plant damage leading to higher percentages of dead plants than hand-harvesting, but this did not occur. Plants that produced greater flower, petal, and pigment mass under hand-harvesting were more likely to die than hedge-trimmed plants with less production. Plants that received nitrogen also had higher percentages of dead plants than those that did not receive nitrogen (Table 3.6). Excessive nitrogen fertilization may have resulted in lush growth and subsequently increased insect damage and disease problems (Agrios, 1997). Also, the form of nitrogen, ammonium or nitrate, may either enhance or reduce disease severity

depending upon the disease. Acidic soil pH appears to be correlated with increased disease incidence when ammonium nitrogen is used. However, increased disease occurs more commonly with nitrate nitrogen at neutral or alkaline soil pH (Agrios, 1997). Disease problems were not noted for the 1999 season, even though soil pH was 6.5, but arthropod damage was prevalent and will be discussed later.

Two three-way interactions (cultivar, nitrogen application, and establishment method and nitrogen application, establishment method, and harvest method) affected flower diameter in 1999 (Table 3.7). As in 1998, nitrogen application and establishment method were factors in both interactions. Transplanted plants that received nitrogen had large flower diameters within each cultivar. With the second interaction, harvest method along with nitrogen application and establishment method influenced flower diameter (Table 3.7). Interestingly, hedge-trimming reduced flower diameter compared with hand-harvesting regardless of establishment method or nitrogen application. Hedge-trimming undoubtedly damaged the top part of the plant canopy, which may have directed photosynthates from reproductive structures to foliage. This may partly explain the decreased flower number as well. The addition of nitrogen caused hedge-trimmed direct-seeded plants to have flower diameters equal to hedge-trimmed transplanted plants.

Cultivar, establishment method, and nitrogen application affected plant canopy height in 1999 (Table 3.7). Although these three parameters interacted, they did not alter the naturally occurring height of one cultivar over another. Transplants of 'I-822' were the tallest and transplants of 'A-975' were the shortest regardless of nitrogen application.

Flower canopy was affected by cultivar, establishment method, and harvest method in 1999 (Table 3.8). As in 1998, 'I-822' had the tallest flower canopy but only as

direct-seeded plants and not as transplanted plants. 'A-975' transplants had the shortest flower canopy both in 1998 and 1999.

Spider mites (*Tetranychus urticae*) were a problem in 1998 and 1999 but not in 1996 or 1997. The 1998 and 1999 growing seasons were hotter and drier than in previous years, and these conditions usually correspond with spider mite infestation. Dry and hot environmental conditions limit effectiveness of a fungus (*Amblyseius fallacis*) that parasitizes spider mites and keeps populations in check (Berberet, personal communication). Chemical control of spider mites was achieved by spraying *O,O*-dimethyl-*S*-1,2-di(carboethoxy) ethyl phosphorodithioate (Malathion®, Miller Chemical and Fertilizer Corp., Hanover, Pa.) and 1,1-bis-(Chlorophenyl)-2,2,2-trichloroethanol (Kelthane®, Rohm and Haas Co., Philadelphia, Pa.) at recommended label rates. Corn earworms (*Helicoverpa zea*) were found to be feeding from within the receptacle in 1998 and 1999. Plants were sprayed with *O,O*-diethyl*O*-(2-isopropyl-4-methyl-6-pyrimidinyl) phosphorothioate (Diazanon, Prentiss Drug and Chemical Co., Inc., Newark, N.J.) and (s)-cyano (3-phenoxyphenyl) methyl (s)-4-chloro-alpha-(1-methyl ethyl)benzeneacetate (Asana®, duPont Corp., Wilmington, Del.) in 1998 for control. Interestingly, in 1999, blister beetles (family Meloidae) voraciously fed on the majority of the 'A-975' plants but did not feed on plants from other cultivars. Undoubtedly, this contributed to the increased percentage of dead plants for that cultivar. Baldwin et al. (1993) stated that mites and thrips were a problem only in the early season, and corn earworms were buried within the blooms from mid to late season. Routine insecticide applications were used to control the pests (chemicals not specified). Baldwin et al. (1993) also had problems with *Alternaria* leaf spot.

CONCLUSIONS

The highest amounts of lutein pigment were consistently produced by 'E-1236' for both years that pigment was measured. 'E-1236' was also a top producer for flower number and mass of fresh flower, dried flower, and dried petal for 3 years. Transplanted plants yielded higher amounts for all data parameters as compared to direct-seeded plants for 3 years. Surprisingly, nitrogen application created mixed results by increasing flower diameter, increasing the percentage of dead plants, and reducing dried receptacle mass. However, nitrogen application did not increase pigment yield. Dried flower and pigment yields were greatly increased by hand-harvesting compared with hedge-trimming in the year both were examined. Therefore, based on yield alone, we recommend hand-picking 'E-1236' transplanted plants for commercial production of *T. erecta*. However, hand-harvesting and transplants are not practical for commercial operations due to high labor expenses. Nitrogen fertilizer did not benefit data parameters enough to warrant its recommendation in commercial production. Annual pre-plant soil fertility analysis should be conducted to ensure proper nutrient availability.

If a commercial producer was interested in growing *T. erecta* as direct-seeded plants with hand-harvesting, 'E-1236' was a top producer (pigment mass 20,700 $g.ha^{-1}$) for that treatment at times equal to or better than transplanted 'E-1236' plants. For mechanical harvesting, transplanted 'E-1236' and 'OL' plants performed best with 'OL' producing the highest pigment mass (10,600 $g.ha^{-1}$) compared with 'E-1236' (6,200 $g.ha^{-1}$). If for economic reasons, a commercial producer wished to use mechanical harvesting with direct-seeded plants, 'E-1236' was the best overall producer for that treatment with pigment mass of 5,500 $g.ha^{-1}$ in a single season.

Even though the highest yield was obtained from hand-harvesting of transplants, such practices would be labor intensive and cost prohibitive. Future research should examine practices that would make mechanical *T. erecta* harvesting more profitable if marigolds are to be considered an alternative crop for the southern plains. Hedge-trimming, as conducted in this project, only cut flowers that were within the horizontal plane of the treatments' flower canopy, but all cut flowers were collected. A mechanical harvester might be able to cut more flowers than hedge-trimmers, but, most likely, would not retain as many flowers as we did during our work.

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Table 3.1. Effect of establishment method and spacing on *Tagetes erecta* 'Orange Lady' data collected at Stillwater, Okla. Values shown are means of four repetitions for six harvests (1996).

Treatment	Flower no. (flowers.ha^{-1})	Fresh flower mass (kg.ha^{-1})	Dried flower mass (kg.ha^{-1})	Dried petal mass (kg.ha^{-1})	Dried receptacle mass (kg.ha^{-1})
		Establishment method			
Direct-seeded	27,906,000	800	115	43	72
Transplanted	61,998,000	1,600	225	97	128
		Spacing (cm)			
10	56,226,000	1,500	212	87	126
23	40,949,000	1,100	153	63	91
36	37,696,000	1,000	144	61	83
Significance					
Establishment (E)	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001
Spacing (S)					
Linear	<0.0001	0.0004	0.0002	0.0012	0.0001
Quadratic	NS	NS	NS	NS	NS
E*S	NS	NS	NS	NS	NS

[NS] Nonsignificant

Table 3.2. Effects of five *Tagetes erecta* cultivars on harvest data collected at Hinton, Oklahoma. Values shown are means of four repetitions for nine harvests (1997).

Cultivar	Flower no. (flowers.ha^{-1})	Fresh flower mass (kg.ha^{-1})	Dried flower mass (kg.ha^{-1})	Dried petal mass (kg.ha^{-1})	Dried receptacle mass (kg.ha^{-1})
A-975	37,186,000 ab [z]	4,700 ab	811 ab	464 ab	348 bc
E-1236	42,703,000 a	5,300 a	906 a	530 a	377 b
I-822	22,867,000 c	4,300 b	728 b	418 b	312 bc
Orange Lady	31,190,000 b	4,200 b	741 b	283 c	443 a
X-986	19,486,000 c	2,500 c	482 c	193 d	289 c
Significance	<0.0001	<0.0001	<0.0001	<0.0001	0.0013

[z] Mean separation within columns by Duncan's multiple range test. Means followed by the same letter are not significantly different at $P \geq 0.05$.

Table 3.3. Effect of four *Tagetes erecta* cultivars and establishment method on harvest data collected at Hinton, Oklahoma. Values shown are means of four repetitions for eleven harvests (1998).

Treatment	Flower no. (flowers.ha^{-1})	Fresh flower mass (kg.ha^{-1})	Dried flower mass (kg.ha^{-1})	Dried petal mass (kg.ha^{-1})	Dried receptacle mass (kg.ha^{-1})	Pigment mass (g.ha^{-1})
			Cultivar			
A-975	7,622,000 b [z]	13,400 b	3,300 c	1,300 b	1,900 c	8,900 c
E-1236	11,035,000 a	25,700 a	6,100 a	2,400 a	3,700 ab	22,000 a
I-822	5,677,000 b	16,700 b	4,300 bc	1,600 b	2,700 bc	15,000 b
Orange Lady	7,080,000 b	17,400 b	5,900 ab	1,200 b	4,700 a	11,000 bc
Significance	0.0010	0.0062	0.0032	0.0020	0.0001	0.0002
			Establishment method			
Transplanted	11,?58,000	25,200	6,800	2,300	4,400	20,000
Direct-seeded	3,476,000	9,500	2,500	700	1,800	7,200
Significance	0.0001	0.0001	0.0001	0.0001	0.0001	0.0001

[z] Mean separation within columns by Duncan's multiple range test. Means followed by the same letter are not significantly different at $P \geq 0.05$.

Table 3.4. Effect of four *Tagetes erecta* cultivars and direct-seeded (DS) or transplanted (TR) establishment on plant and flower canopy heights and plant stand for Hinton, Oklahoma. Values shown are means of four repetitions for eleven harvests (1998).

Cultivar	Establishment method	Plant canopy height (cm)	Flower canopy height (cm)	Dead plants at mid-season (%)
A-975	DS	27.3 [z]	35.4	37
	TR	28.4	32.9	29
E-1236	DS	28.2	37.2	3
	TR	41.1	47.2	39
I-822	DS	39.3	47.2	24
	TR	44.9	52.3	22
Orange Lady	DS	36.1	42.8	9
	TR	44.5	50.3	19
Significance				
$LSD_{0.05}$ Different cultivar or establishment method		6.2	9.1	24.6

Table 3.5. Effect of four *Tagetes erecta* cultivars, direct-seeded (DS) or transplanted (TR) establishment, and hand-harvesting (HH) or hedge-trimming (HT) on harvest data for Hinton, Okla. Values shown are means of four repetitions (1999).

Cultivar	Establish-ment method	Harvest method [z]	Flower no. (flowers. ha^{-1})	Fresh flower mass ($kg.ha^{-1}$)	Dried petal ($kg.ha^{-1}$)	Pigment mass ($g.ha^{-1}$)
A-975	DS	HH	4,624,000	14,400	1,270	9,500
		HT	1,818,000	6,500	500	4,200
A-975	TR	HH	6,091,000	21,400	1,700	13,500
		HT	3,381,000	10,600	810	6,000
E-1236	DS	HH	7,622,000	22,400	2,140	20,700
		HT	1,850,000	6,800	540	5,600
E-1236	TR	HH	5,645,000	20,800	1,810	14,500
		HT	3,189,000	11,800	910	6,200
I-822	DS	HH	2,838,000	13,300	1,300	13,100
		HT	1,021,000	4,200	310	3,200
I-822	TR	HH	3,157,000	15,600	1,300	14,400
		HT	1,563,000	6,200	440	4,800
Orange Lady	DS	HH	5,804,000	14,500	1,000	11,800
		HT	2,519,000	6,300	370	4,400
Orange Lady	TR	HH	7,080,000	23,000	1,660	21,300
		HT	3,253,000	11,100	760	10,600
Significance						
$LSD_{0.05}$ Different cultivar and establishment method for same harvest method			1,276,000	3900	350	3400

Table 3.5. Continued.

Cultivar	Establish-ment method	Harvest method [z]	Flower no. (flowers. ha^{-1})	Fresh flower mass ($kg.ha^{-1}$)	Dried petal ($kg.ha^{-1}$)	Pigment mass ($g.ha^{-1}$)
$LSD_{0.05}$ Same or different cultivar, establishment, or harvest method			1,435,140	4000	370	3500

[z] HH plants were harvested eight times in the season, and HT plants were harvested five times.

Table 3.6. Effect of cultivar, establishment method, nitrogen application, and harvest method for *Tagetes erecta* harvest at Hinton, Oklahoma. Values shown are means of four repetitions (1999).

Treatment	Dried flower mass ($kg.ha^{-1}$)	Dried receptacle mass ($kg.ha^{-1}$)	Dead plants at mid-season (%)	Dead plants at end of season (%)
		Cultivar		
A-975	3300 b	2200 b	27 ^{NS}	29 ^{NS}
E-1236	3800 ab	2400 b	17	18
I-822	2600 c	1700 c	14	16
Orange Lady	3900 a	2900 a	17	18
		Establishment method		
Direct-seeded	2900	2000	12	12
Transplanted	3800 ***	2700 ***	26 ***	28 ***
		Nitrogen (kg/ha)		
0.0	3500 ^{NS}	2400 **	14	15
4.6	3200	2200	24 **	26 **
		Harvest method [z]		
Hand-harvested	4700 ***	3100 ***	23 **	24 **
Hedgetrimmed	2100	1500	15	16

^{NS}, **, *** Nonsignificant or significant at $P \geq 0.05$ and 0.01, respectively

[z] Hand-harvested plants were harvested eight times in the season, and hedge-trimmed plants were harvested five times.

Table 3.7. Effect of cultivar, direct-seeded (DS) or transplanted (TR) establishment, nitrogen application, and hand-harvested (HH) or hedge-trimmed (HT) harvest on *Tagetes erecta* flower diameter and plant canopy height. Values shown are means of four repetitions (1999).

Cultivar	Establishment method	Nitrogen application ($kg.ha^{-1}$)	Harvest method [z]	Flower diameter (cm)	Plant canopy height (cm)
A-975	DS	0.0	-	6.0	27.3
		4.6	-	6.0	27.3
A-975	TR	0.0	-	6.1	26.9
		4.6	-	5.9	26.4
E-1236	DS	0.0	-	6.2	38.2
		4.6	-	6.4	38.8
E-1236	TR	0.0	-	6.6	43.4
		4.6	-	6.2	40.8
I-822	DS	0.0	-	7.1	44.1
		4.6	-	6.7	40.8
I-822	TR	0.0	-	6.9	46.2
		4.6	-	7.2	46.7
Orange Lady	DS	0.0	-	5.4	39.1
		4.6	-	5.6	38.6
Orange Lady	TR	0.0	-	5.5	40.6
		4.6	-	5.2	41.8
-	DS	0.0	HH	6.5	-
-	DS	0.0	HT	5.9	-
-	DS	4.6	HH	6.3	-

Table 3.7. Continued

Cultivar	Establishment method	Nitrogen application ($kg.ha^{-1}$)	Harvest method	Flower diameter (cm)	Plant canopy height (cm)
-	DS	4.6	HT	6.0	-
-	TR	0.0	HH	6.3	-
-	TR	0.0	HT	6.2	-
-	TR	4.6	HH	6.4	-
-	TR	4.6	HT	5.9	-
Significance					
$LSD_{0.05}$ To compare cultivar and establishment method with same nitrogen application				0.2	0.9
$LSD_{0.05}$ Same or different cultivar, establishment, or nitrogen application				0.2	0.9
$LSD_{0.05}$ To compare establishment method with same nitrogen application and harvest method				0.2	-
$LSD_{0.05}$ To compare harvest method with same nitrogen application and same or different establishment method				0.3	-
$LSD_{0.05}$ To compare nitrogen application with same or different harvest method and establishment method				0.2	-

[z] Hand-harvested plants were harvested eight times in the season, and hedge-trimmed plants were harvested five times.

Table 3.8. Effect of cultivar, direct-seeded (DS) or transplanted (TR) establishment, and hand-harvested (HH) or hedgetrimmed (HT) harvest on *Tagetes erecta* flower canopy height. Values shown are means of four repetitions (1999).

Cultivar	Establishment method	Harvest method [z]	Flower canopy height (cm)
A-975	DS	HH	35.3
		HT	35.3
A-975	TR	HH	32.8
		HT	28.0
E-1236	DS	HH	51.6
		HT	46.1
E-1236	TR	HH	48.6
		HT	46.0
I-822	DS	HH	59.3
		HT	55.9
I-822	TR	HH	57.1
		HT	51.2
Orange Lady	DS	HH	51.8
		HT	46.1
Orange Lady	TR	HH	48.9
		HT	44.6
Significance			
$LSD_{0.05}$ Different cultivar and establishment method for same harvest method			1.1
$LSD_{0.05}$ Same or different cultivar, establishment, or harvest method			1.1

[z] HH plants were harvested eight times in the season, and HT plants were harvested five times.

CHAPTER IV

POSTHARVEST HANDLING OF CUT *CAMPANULA MEDIUM* FLOWERS

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ABSTRACT

Various postharvest treatments were applied to evaluate and improve the longevity and quality of cut *Campanula medium* L. 'Champion Blue' and 'Champion Pink' stems. Stems tolerated 2° C storage for 1 week either wet or dry. Longer storage of 2 or 3 weeks yielded higher percentages of senesced flowers at termination (5.9 days) and significantly shorter vase lives (5.0 days). A 38 °C pretreatment with a 5% sucrose pulse solution produced the longest vase life (10.3 days) and maintained high quality even at 6 days (4.2, with 5.0 as best). Stems had an average vase life of only 3.3 days when placed in floral vase foam but lasted 10.0 days without foam. When stems were placed in a 2.0% sucrose solution without foam, vase life was 12.2 days. High (110 $\mu mol.m^{-2}.s^{-1}$) or low (10 $\mu mol.m^{-2}.s^{-1}$) light levels did not affect postharvest parameters, but did affect opening flower color as measured by a chroma meter. The most recently opened flowers under low light conditions were paler than those under high light conditions.

INTRODUCTION

In 1999, the United States cut flower industry grossed $426 million dollars, which was a 3% increase over the previous year (United States Department of Agriculture [USDA], 2000). Much of the increase was due to "specialty cut flowers", which include species outside of the most commonly used flowers: carnations (*Dianthus caryophyllus* L.), chrysanthemums (*Dendranthema x grandiflorum* Kitam.), gladiolus (*Gladiolus* L.), and roses (*Rosa* L.). Specialty cuts have been growing in popularity with the industry and consumers for the last several years. Roses, carnations, chrysanthemums, and gladiolus, which have been the mainstay of the cut flower industry for years, were "down sharply" in 1999 as part of a continuing trend (USDA, 2000). Continued success of specialty cut flowers requires the continual introduction of new species. *Campanula medium* L. is a biennial plant commonly used in gardens in the United States and sold as flowering potted plants in Europe (Dole and Wilkins, 1999). Flower color is predominately blue or lavender but can be also white or pink. The inflorescence is a raceme of 2.5 cm long cup-shaped flowers creating an unusual overall shape not common in cut flowers. 'Champion Blue' and 'Champion Pink' were bred specifically for cut flower production. They have a short crop time, typically flowering in 16 to 20 weeks, making it possible to grow and sell this normally biennial plant in a cost effective time period (Cavins, 1999). However, limited research has been done to examine the postharvest life of cut *Campanula* flowers (Sakata Seed America, Inc., personal communication).

Cultural conditions and handling of a cut flower after harvest determine the length of its vase life (Nowak and Rudnicki, 1990). Thus, growers and florists can employ

many techniques, such as cooler storage, pretreatments, pulsing, sucrose solutions, and light conditions, to lengthen vase life and maintain quality of cut flowers.

Storing cut stems in coolers benefits the grower and florist by extending the season, regulating availability during peak production or high demand periods, improving production efficiency, and enabling long-term shipment (Goszczynska and Rudnicki, 1988). Low temperature storage reduces the rate of metabolism, transpiration, endogenous ethylene production, and pathogen growth thus maintaining quality of the stored material. Recommended storage temperatures for cut flowers depend on the particular species and cultivar, but generally vary between 0 and 4 °C (Nowak and Rudnicki, 1990). Either wet or dry storage is used for cut flowers. Dry storage consists of wrapping stems in a moisture-absorbing paper, such as newspaper, and then wrapping in polyethylene plastic or storing in a wax-lined box. Dry storage is usually employed for several weeks but can reduce vase life and quality (Nowak and Rudnicki, 1990). With wet storage, flower stems are placed in water or a preservative solution for short durations, such as a few days.

Pretreatments and pulses are other techniques that greatly enhance the quality and longevity of cut flowers. Pretreatments and pulses are generally short-term treatments (a few hours) conducted just after harvest. A pretreatment may consist of placing stems in heated water (approximately 38 °C) to enhance water absorption or applying an anti-ethylene compound, such as silver thiosulfate (STS) or 1-methylcyclopropene (1-MCP). Silver thiosulfate is effective against exogenous ethylene-induced senescence in some species and cultivars, but the health and environmental hazards imposed by handling and subsequent disposal of the compound limit its use. 1-methylcyclopropene has

demonstrated effectiveness equal to STS but has also shown efficacy against endogenous ethylene sources (Serek et al., 1995). The ease of applying 1-MCP as a gas has ensured its broad-scale use in commercial applications.

During pulsing, floral stems are placed in solutions containing high amounts of sucrose and a germicide from a few hours to two days. Silver thiosulfate can also be applied in a pulse solution. Percentages of sucrose ranged from 0.5% to 25% depending upon the species and cultivar (Goszczynsda and Rudnicki, 1988). The sucrose amount and pulsing duration must be closely monitored to avoid leaf and petal damage. Sucrose pulsing increased rate and number of flowers opening, improved petal coloration, and extended vase life of carnations, chrysanthemums, peonies (*Paeonia lactiflora* Pall.), snapdragons (*Antirrhinum majus* L.), and bird-of-paradise (*Strelitzia reginae* Ait.) (Nowak and Rudnicki, 1990).

In addition to pulsing, cut stems also benefit from a continuous supply of sucrose in the vase solution. Sucrose replaces photosynthates after the stems are harvested, increasing vase life and enabling buds to properly develop and become larger (Halevy and Mayak, 1979; Sacalis, 1993). Sucrose percentages normally used in vase solutions are lower than those for pulsing and can range from 0.5% to 7%. With the presence of sucrose, addition of a germicide is necessary to inhibit microbial growth. The most commonly used anti-microbial compound with cut flowers is 8-hydroxyquinoline citrate (8-HQC) (Sacalis, 1993). Not only is 8-HQC effective against microbes, it also is slightly acidic which benefits water absorption. Acidic water (pH 3.0 to 4.0) is more readily absorbed by plants than alkaline water (Sacalis, 1993).

Light conditions are not known to be a significant postharvest factor for most cut flowers. Vase life has shown to be unaffected, but low light conditions during long-distance transport can result in leaf yellowing in some species, such as alstroemerias (*Alstroemeria* L.), chrysanthemums, dahlias (*Dahlia* Cav.), and gladiolus (Nowak and Rudnicki, 1990; Sacalis, 1993). Flower color is often less intense when grown under low light conditions. Halevy and Mayak (1974) reported the pale color of cut roses grown under low light conditions was reversed when sucrose was added to the solution during bud opening. Flowers that remained on the plant under low light conditions continued to be pale. However, low light conditions after harvest can also affect flower color development. Incomplete coloration of thin petals has been correlated with purple cultivars of *Eustoma grandiflorum* Griese. under low light conditions after stem harvest (Griesbach, 1992; Kawabata et al., 1995).

Flower color is best measured objectively using the parameters: value, hue, and chroma, as defined by Hunter (1942), rather than subjectively with the naked eye. Value is a measure of the color's lightness or darkness. Hue represents the actual colors found on a color wheel, i.e., red, blue, green, and yellow. Chroma conveys the vividness or dullness of a certain color (Minolta, 1985). Chroma meters measure and quantify value, hue, and chroma represented as L*, a*, and b*, respectively. With a numerical value, objective comparisons can easily be made among various treatments.

Objectives of this research were to maximize storage life, color development in flowers opening after harvest, and vase life of *C. medium* 'Champion' for the retailer and consumer. Information gained from this project would prove valuable to growers, floral

retailers, and ultimately the consumer by providing more variety in the choice of cut flowers.

Materials and Methods

Cut stems of *C. medium* 'Champion Blue' and 'Champion Pink' were obtained from Burdette and Coward Co., Punta Gorda, Fla. Stems were placed in unamended tap water after harvest and cooled at 2 °C until shipped dry overnight to Stillwater, Okla. on 5 Jan. 2000. 'Champion Blue' stems were used in expts. 1, 3, 4, and 5; 'Champion Pink' stems were used in expt. 2. Upon arrival, boxed stems were held at 2 °C until processed the same day for experiments. All stems were recut under water and then placed in the appropriate treatment. Unless otherwise indicated, the solution (floral solution) used in all experiments was 22 °C deionized water amended to pH 3.5 using citric acid and 200 $mg.L^{-1}$ 8-HQC. Sterile glass vases that held approximately 180 ml floral solution were used in all experiments. Room air temperature averaged 19/24 °C minimum/maximum, and light level averaged 115 $\mu mol.m^{-2}.s^{-1}$. The following parameters were measured in each experiment: number of flowers per stem open prior to treatment, number of flowers per stem open during treatment, vase life after removal from treatment, and number of senesced flowers at termination. Also, quality ratings (1 to 5, with 5 best) were measured daily beginning the first day of treatment.

Flower color (value, hue, and chroma) was measured using a Minolta CR-200 chroma meter (Minolta Corp., Ramsey, N.J.) during expts. 2 and 4 one week after treatment on a flower open prior to treatment (first flower) and the most recently opened flower (second flower).

Prior to placement in the treatments, stems were sorted by the number of opened flowers (4 to 9 flowers/stem), and treatments blocked accordingly. Fifteen repetitions (flower stems) were used for each treatment, except for expt. 2 in which 12 repetitions were used. Stems were terminated when half of the florets had necrotic edges, individual florets were wilted, or the stem collapsed. Data were subjected to the general linear model procedure, and mean separation was accomplished by trend analysis or Duncan's multiple range test (SAS Institute, Cary, N.C.). Percentage data were transformed using the arcsin procedure prior to statistical analysis.

Exp. 1 - Cold storage duration

Cut stems were held 0, 1, 2, or 3 weeks in a 2 °C cooler (RH 86%) either dry or wet. For dry storage, stems were placed in boxes lined with paper and plastic. Wet stems were placed in sterile floral buckets filled with floral solution. The 0 week storage stems were placed directly into floral vases. At weeks 1, 2, and 3, fifteen stems were randomly selected from each treatment, recut under water, and placed in floral vases filled with floral solution. All remaining stems were recut under water and placed back into their respective treatments. Floral solution was changed in buckets for the wet treatment prior to return of the stems. The experimental design was a randomized complete block.

Exp. 2 – Pretreatments and pulses

Cut stems received three different 4 h pretreatment methods: 1) floral solution heated to 38 °C, 2) 1-methylcyclopropene (1-MCP) (Ethylbloc, BioTechnologies for Horticulture, Inc., Burr Ridge, Ill.) and 22 °C floral solution, or 3) 22 °C floral solution. After pretreatments, stems received a 24 h pulse treatment of 0, 5, or 10% sucrose in floral solution. For 1-MCP application, 2.3 g were added to 45 ml buffer agent releasing

1-MCP gas into 5.1 m^3 of enclosed air space. Chroma meter readings were taken one week after the stems had been in the floral vases. Experiment was a completely randomized design.

Exp. 3 – Vase solutions and substrates

Cut stems were placed in vases containing either floral foam (Aquafoam™ Instant, Syndicate Sales, Inc. Kokomo, Ind.) or floral solution, and one of five solutions containing 0, 0.5, 1, 2, or 4% sucrose was added. Foam was allowed to thoroughly soak in the respective solution before placement in vase. Solution level in the foam vases was monitored to ensure it stayed below the bottom of the cut stem. A completely randomized design was used.

Exp. 4 – Light levels

Cut stems were placed in floral solution and exposed to either 10 or 110 $\mu mol.m^{-2}.s^{-1}$ light. Chroma meter readings were taken after 1 week.

Exp. 5 – Control solutions

Cut stems were placed in one of four control solutions: 1) 22 °C deionized water (pH 4.0), 2) 22 °C deionized water amended to pH 3.5 with citric acid, 3) 22 °C deionized water (pH 4.0) plus 200 $mg.L^{-1}$ 8-HQC, and 4) 22 °C deionized water amended to pH 3.5 with citric acid and 200 $mg.L^{-1}$ 8-HQC. The experiment was a completely randomized design.

RESULTS

Expt. 1 – Cold storage duration

Vase life. Increasing cold storage duration linearly reduced vase life (Table 4.1). Storage method (dry or wet) did not affect vase life.

Flowers opened during storage. A curvilinear response was evident for the percentage of flowers that opened during cold storage, such that weeks 1 and 3 had the largest percentages, while week 2 exhibited the lowest percentage (Table 4.1). Storage method did not affect the percentage of flowers opening.

Senesced flowers at termination. Storage method and time interacted to affect the percentage of senesced flowers at termination (Table 4.1). Dry storage for 3 weeks resulted in the greatest percentage of senesced flowers, but wet storage for 2 or 3 weeks resulted in the lowest percentages of senesced flowers at termination. Treatments with 0 weeks storage did not have any senesced flowers.

Quality at 3 days. A cubic interaction existed between the storage method and time for the quality ratings at 3 days, such that dry or wet storage at 0 weeks resulted in the highest rating (5.0), but dry storage for 3 weeks produced the lowest rating (2.7) (Table 4.1).

Quality at 6 days. Wet storage produced significantly higher quality ratings at 6 days as compared with dry storage (Table 4.1). Quality decreased linearly with increasing storage time (Table 4.1).

Quality at 9 days. Increasing storage time linearly decreased the quality rating at 9 days (Table 4.1). Storage method did not affect rating at 9 days.

Expt. 2 – Pretreatments and pulses

Vase life. Pretreatments and pulses interacted such that 1-MCP or 38 °C floral solution combined with the 5% sucrose pulse resulted in the longest vase life (Table 4.2). The shortest vase life occurred when the 38 °C treatment was used in combination with 10% sucrose pulse.

Flowers opened during treatment. Increasing sucrose concentration in the pulse solution linearly decreased the percentage of flowers opening during treatment (Table 4.2). Treatments with 0% sucrose stimulated the greatest percentage (26.0) of open flowers while 10% sucrose produced the lowest percentage (9.5). Pretreatment methods did not affect the percentage of flowers opening.

Senesced flowers at termination. Pretreatment and pulse methods interacted such that increasing sucrose concentration increased the percent of senesced flowers at termination with all three pretreatments, but the greatest increase with increasing sucrose concentration occurred with the 38 °C pretreatment (Table 4.2).

Quality at 3 days. A curvilinear interaction between pretreatment and pulse solution existed such that increasing sucrose concentrations reduced quality ratings at 3 days with all three pretreatments, but the lowest rating occurred with the 38 °C pretreatment (Table 4.2).

Quality at 6 days. Pretreatments interacted with the pulse solution such that 38 °C with 5% sucrose produced the highest quality rating, but 38 °C with 10% sucrose produced the lowest quality rating (Table 4.2).

Quality at 9 days. Pulse solution displayed a quadratic response for the quality rating at 9 days, while the pretreatments did not have an effect (Table 4.2). The 5% sucrose produced the highest quality rating at 9 days, and the 0% sucrose produced the lowest.

Flower color. Pretreatment methods did not significantly ($P \leq 0.5$) affect flower value, hue, or chroma (Table 4.3). Increasing sucrose concentration linearly increased value and decreased hue and chroma numbers for the first flower measured. Thus,

increasing sucrose concentration resulted in paler, less intensely colored flowers. Increasing sucrose concentration curvilinearly increased value and chroma and decreased hue of the second measured flower. Consequently, increasing sucrose concentration curvilinearly decreased value and hue differences and increased chroma difference between the first and second measured flowers.

Experiment 3 –Vase sucrose and substrates

Vase life. Floral vase substrate and sucrose concentration interacted such that stems in the no-foam treatment with 2.0% sucrose had the longest vase life (12.2 days), but the shortest vase life occurred with foam and 0.5% sucrose (2.3 days) (Table 4.4).

Senesced flowers at termination. Substrate material and sucrose concentration interacted to create the largest percentage of senesced flowers (46.2) in the no-foam treatment with 4.0% sucrose (Table 4.4). The treatment combination of foam with 0.5% sucrose was the only treatment to yield no senesced flowers due to rapid death of the stem (2.3 days).

Quality at 3 days. A curvilinear interaction between the floral vase substrate and sucrose percentage occurred such that a rating of 5.0 was given to the stems in the no-foam treatment with 0.5% sucrose, but foam and 0.5% sucrose had a rating of 3.3 (Table 4.4).

Quality at 6 and 9 days. The majority of stems in the foam treatment did not survive past 3 days (Table 4.4). Stems in the no-foam treatment maintained high quality at 6 (4.7) and 9 (3.8) days, but ratings decreased linearly with increasing sucrose concentration (Table 4.4).

Expt. 4 – Light levels

Light level did not significantly affect vase life, percentage of senesced flowers at termination, or quality at 3, 6, or 9 days (data not presented). High and low light levels did not significantly affect value, hue, or chroma for the first flower that was measured (Table 4.5). Under low light, the second measured flower was paler in color, less intense, and less vivid compared with the second measured flower under high light treatment. The difference in value between first and second flower was unaffected by light. However, hue and chroma differences between first and second flowers were significantly different.

Expt. 5 – Control solutions

Deionized water (DW) plus 200 $mg.L^{-1}$ 8-HQC, regardless of pH, yielded the the highest quality ratings at day 6 (Table 4.6). No significant difference existed among the treatments for vase life, number of senesced flowers at termination, and quality ratings at 3 or 9 days (data not presented).

DISCUSSION

Expt. 1 – Cold storage duration

Stems that were stored either wet or dry for 1 week had similar vase lives, but vase life was about 2 days shorter than for stems that had not been stored (Table 4.1). As dry storage increased to 2 or 3 weeks, however, the percentage of senesced flowers at termination significantly increased compared with wet storage. While wet storage at 3 weeks yielded the shortest vase life (5 days), it had a low percentage of senesced flowers

(0.5%) at termination. Long term storage often results in decreased vase life (Sacalis, 1993). Kelly and Starman (1990) found that cut *Physostegia purpurea* Blake flowers had a vase life of approximately 4 days after 7 days dry storage at 1 °C. Similarly, six cut flower cultivars and species, *Leucadendron* R. Br. 'Silvan Red', *Leucospermum* R. Br. 'Firewheel', *Protea cynaroides* L., *Thryptomene calycina* (Lindl.) Stapf., *Telopea speciosissima* R. Br., and *Verticordia grandiflora* Endl., of the Proteaceae family had vase lives of at least 7 days even after 3 weeks of dry storage at 1 °C (Jones and Faragher, 1991). The Proteaceae stems were given a 24 h pre- and post-treatment in distilled water and held at 1 °C. Little difference existed between wet and dry storage of *Campanula*, except that quality was significantly lower at 3 and 6 days, and the percentage of senesced flowers was greater at termination for dry stored stems. At 9 days, no significant difference existed between quality ratings of stems placed in wet or dry storage. Interestingly, many of the florets (18%) continued to color and open while in storage (Table 4.1).

Expt. 2 – Pretreatments and pulses

The 38 °C pretreatment combined with a 5% sucrose pulse proved to be the best treatment by providing a long vase life (10.3 days), an acceptable percentage of senesced flowers at termination (6.8), and high quality, even at 6 days (4.2) (Table 4.2). The 1-MCP pretreatment with 5% sucrose pulse yielded results similar to 38 °C pretreatment and 5% sucrose except that the senesced flower percentage was higher (12.2) and quality ratings were lower (3.8 at 6 days) in the 1-MCP pretreatment. Stems that received 10% sucrose pulse solutions responded poorly regardless of the pretreatment used, producing the highest percentage of senesced flowers and lowest quality ratings. High sucrose

concentrations have been deleterious to other cut flowers, such as chrysanthemums and roses (Halevy et al., 1978; Kofranek and Halevy, 1980). Treatments that used 0% sucrose yielded the lowest percentage of senesced flowers in *Campanula*. Stems that received 1-MCP had a significantly longer vase life (9 days), but by only 1 to 2 days compared with other treatments. Sisler et al. (1996) also found that potted *Campanula carpatica* Jacq. 'Blue Chips' had a display life of 9.0 days when treated with 1-MCP for 6 h compared with 3.3 days for untreated plants.

Sucrose pulse treatments affected flower color whereas pretreatments had no effect (Table 4.3). The linear response of the first flower value, hue, and chroma to sucrose concentration indicated increasing levels of sucrose resulted in paler, grayish flowers for those flowers open at time of treatment. The curvilinear interaction between the second flower hue and chroma and sucrose pulse treatment indicated that flowers unopened at treatment time responded differently to sucrose levels than already open flowers. Color, as determined by hue and chroma, for the second flower was paler for the 5.0% sucrose pulse treatment than the 0.0% or 10.0% treatments. Apparently, flowers unopened and not colored when placed into the 24 h 5% sucrose treatment failed to develop full color when they did open compared to flowers already colored and opened when placed into the treatment. Kawabata et al. (1995) indicated that sucrose solutions (0, 0.125, 0.25, or 0.5 M) were beneficial for full color development for unopened *Eustoma grandiflorum* flower buds after they were detached at the peduncle. Color was measured objectively using a scanner and image software and scored between 0: white and 256: black. Full color development in unopened *Campanula* flowers would most

likely be achieved with a 24 h pulse containing sucrose concentrations lower than 5.0% but greater than 0%.

Expt. 3 –Vase sucrose and substrates

Overall, stems that received no foam with either 1.0% or 2.0% sucrose solution had the longest vase life, lowest percentage of senesced flowers, and high quality ratings at 3, 6, and 9 days (Table 4.4). Han (1998) found that cut *Heuchera sanguinea* Engelm. flowers had a longer vase life and higher percentages of opening buds at 0.5% or 1.0% sucrose compared to higher sucrose concentrations, but stem collapse increased at greater than 1.0% sucrose. Cut *Triteleia laxa* Benth. stems had a vase life of 12 days when held in solutions containing 2.0% sucrose, but vase life decreased as sucrose concentrations increased (Han et al., 1990). Stems inserted into foam, regardless of the sucrose solution percentage, performed poorly; buds failed to open, and vase life was only 2.3 to 5.2 days. Interestingly, stems inserted into foam with 4.0% sucrose had the longest vase life within the foam treatments, but for the no-foam treatments, stems in 4.0% sucrose had a vase life only 0.5 days greater than the 0% sucrose. Most likely, the *Campanula* stems were unable to take up sufficient solution in the foam to sustain themselves, but when 4.0% sucrose was used, stems were able to absorb enough carbohydrates to increase vase life and decrease the percentage of dead flowers at termination. Longevity and continued development of cut flowers have been positively correlated with an external supply of carbohydrates (Halevy and Mayak, 1979). Stems in the no-foam treatment with 4.0% sucrose were able to absorb sufficient solution, but the high concentration of sucrose was detrimental. Quality ratings at 3, 6, and 9 days were exceptional for the stems not

inserted into foam and treated with either 0.5%, 1.0%, or 2.0% sucrose. Little difference existed in quality ratings for 3 and 6 days among the no-foam treatments.

Expt. 4 – Light levels

Light level did not affect postharvest life, quality, or color of the first measured flower (Table 4.5). However, the second measured flowers under low light conditions were paler and less intense in color than the flowers under high light. Griesbach (1992) also found that *Eustoma grandiflorum* flowers that opened under low light were paler than those opening in high light conditions as measured by a photometer microscope. The pale *Eustoma* flowers contained 70% less total anthocyanin pigments than darker flowers, and Griesbach (1992) postulated that decreased biosynthesis was the cause of this reduction. Kawabata et al. (1995) also found that *Eustoma* cut flowers were paler under low light than high light and that shading the leaves and stem reduced flower color. Shading the flower buds did not change the color, even in low light conditions. Kawabata et al. (1995) added that using 0.5M sucrose at 280 $\mu mol.m^{-2}.s^{-1}$ produced flower color in cut stems similar to potted plants held in high light conditions. The authors suggested that sucrose supply enhanced anthocyanin synthesis and maintained the expected flower color even under low light conditions. Nowak and Rudnicki (1990) also stated that petal color is positively correlated with the supply of carbohydrates since increased carbon dioxide levels negated the negative effects of shading rose buds during anthocyanin formation.

Expt. 5 – Control solutions

The deionized water (DW) plus 200 $mg.L^{-1}$ 8-HQC, regardless of pH, yielded the highest quality ratings at 6 days (Table 4.6). Otherwise, no significant differences existed

among the treatments with or without 8-HQC. Ironically, pH of the unamended deionized water averaged an already low 4.0 (range 3.9 – 4.1), which undoubtedly contributed to the similarity among treatments. Deionized water at a higher pH would most likely yield different results. When comparing several biocides (200 $mg.L^{-1}$ 8-HQC, 50 $mg.L^{-1}$ $AgNO_3$, or 100 $mg.L^{-1}$ Physan [n-alkyl dimethyl benzyl ammonium chloride-n-alkyl dimethylethylbenzyl ammonium chloride]) in floral solution, 200 $mg.L^{-1}$ 8-HQC was equally effective as other biocides but had the advantage of not producing a phytotoxic response in cut *Triteleia laxa* flowers (Han et al., 1990). Woodson (1987) found that bud-cut *Freesia hybrida* Bailey stems had increased vase life after 24 h pulsing with 200 $mg.L^{-1}$ 8-HQC as compared to pulsing with deionized water alone (pH not provided). Han et al. (1990) and Han (1998) used deionized water (pH not provided) with 200 $mg.L^{-1}$ 8-HQC as the control solution in experiments with cut *T. laxa* and *H. sanguinea* stems. In both studies, the control solution had the shortest vase life and lowest percentage of bud opening as compared with other treatments.

CONCLUSION

Campanula medium cut stems responded best to: 1) wet storage at 2 °C for one or two weeks, 2) 38 °C pretreatment combined with a 24 h 5.0% sucrose pulse, 3) continuous 1.0% or 2.0% sucrose solution with no foam in the vase, and 4) high light conditions in the postharvest area. The next step would be to examine combinations of these treatments to develop the best handling method for all aspects of postharvest care. If cut *Campanula* stems are to be successful with the florist industry, they should have a

vase life of at least seven days and acceptable quality after insertion into floral foam. The use of pretreatments and 24 h pulse treatments prior to insertion into floral foam may lengthen vase life to an acceptable range. On the other hand, lower ranges of sucrose may be necessary if foam is not used in vases, as evidenced by the results. In future experiments, the lower temperature limit of *Campanula* stems should be further examined. Flowers continued to color and open on the stem while in 2 °C storage, which is unusual for cut flower stems. Apparently, 2 °C was not low enough to halt metabolic activity and further development; lower temperatures may allow longer storage while still achieving acceptable vase life and quality. Also, a lower range of sucrose concentrations, both for the 24 h pulse treatment and for the continuous supply of sucrose, should be examined. From the results, we can assume that the upper tolerance limit to sucrose was reached in both experiments, and lower sucrose concentrations might result in improved postharvest parameters. Han (1998) found that 2.5%, and 5.0% continuous sucrose concentrations were too high in working with *H. sanguinea*, but 0.5%, and 1.0% concentrations were acceptable.

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Table 4.1. Effect of wet or dry and 0, 1, 2, or 3 weeks cold storage on postharvest quality of *Campanula medium* 'Champion Blue'. Values shown are means of fifteen stems.

Storage type	Storage duration (weeks)	Vase life (days)	Flowers opened during storage (%)	Senesced flowers at termination (%)	Q3[z]	Q6	Q9
Dry (D)	0	10.4	0.0	0.0	5.0	5.0	4.1
Dry	1	8.5	20.2	2.6	4.5	4.0	2.9
Dry	2	6.2	15.4	3.6	3.1	3.2	2.7
Dry	3	7.2	17.0	5.9	2.7	2.5	1.9
Wet (W)	0	10.4	0.0	0.0	5.0	5.0	4.1
Wet	1	8.3	18.9	1.1	4.6	4.3	3.1
Wet	2	6.9	13.9	0.5	4.9	3.9	2.6
Wet	3	5.0	22.6	0.5	4.3	2.3	1.0
Dry	-	8.1	13.2	3.0	3.8	3.8	3.1
Wet	-	7.7	13.8	0.5	4.7	4.2	3.4
-	0	10.4	0.0	0.0	5.0	5.0	4.1
-	1	8.4	19.6	1.8	4.6	4.2	3.0

Table 4.1. Continued.

Storage type	Storage duration (weeks)	Vase life (days)	Flowers opened during treatment (%)	Senesced flowers at termination (%)	Q 3[z]	Q 6	Q 9
-	2	6.5	14.7	2.0	4.1	3.6	2.7
-	3	6.1	19.8	3.2	3.5	2.4	1.8
Significance							
Storage type		NS	NS	0.0046	0.0001	0.0043	NS
Storage duration (S)							
Linear (L)		0.0001	0.0001	0.0119	0.0001	0.0001	0.0001
Quadratic (Q)		NS	0.0001	NS	NS	NS	NS
Cubic (C)		NS	0.0001	NS	NS	NS	NS
D*SL		NS	NS	***	***	NS	NS
D*SQ		NS	NS	NS	NS	NS	NS
D*SC		NS	NS	NS	**	NS	NS
W*SL		NS	NS	NS	**	NS	NS

Table 4.1. Continued.

Storage type	Storage duration (weeks)	Vase life (days)	Flowers opened during treatment (%)	Senesced flowers at termination (%)	Q 3[z]	Q 6	Q 9
W*SQ		NS	NS	NS	NS	NS	NS
W*SC		NS	NS	NS	**	NS	NS

NS, **, *** Nonsignificant or significant at $P \geq 0.05$ or 0.01, respectively

[z] Quality ratings (0 to 5, with 5 the best) determined at 3 (Q3), 6 (Q6), and 9 (Q9) days after end of treatment

Table 4.2. Effect of 24-h unheated or heated water or 1-methylcyclopropene (1-MCP) pretreatments and 0.0, 5.0, or 10.0% sucrose pulses on postharvest quality for *Campanula medium* 'Champion Pink'. Values shown are means of twelve stems.

Pre-treatment	Sucrose pulse (%)	Vase life (days)	Flowers opened during treatment (%)	Senesced flowers at termination (%)	Q 3[z]	Q 6	Q 9
Unheated (U)	0.0	9.1	34.1	5.4	4.9	4.1	2.7
Unheated	5.0	7.8	19.4	14.4	4.2	3.7	2.8
Unheated	10.0	8.8	11.6	19.0	4.2	3.9	2.7
38 °C (38)	0.0	7.3	25.3	1.1	4.8	3.9	2.1
38 °C	5.0	10.3	30.7	6.8	4.5	4.2	3.2
38 °C	10.0	3.6	10.2	43.2	3.3	2.9	2.8
1-MCP (M)	0.0	7.8	18.9	1.4	5.0	4.1	2.5
1-MCP	5.0	10.4	18.2	12.2	4.4	3.8	3.1
1-MCP	10.0	9.7	6.8	24.4	4.0	3.4	2.7
Unheated	-	8.6	21.7	12.9	4.5	3.9	2.7
38 °C	-	7.1	22.1	17.0	4.4	3.9	2.8
1-MCP	-	9.3	14.7	12.7	4.5	3.8	2.8

Table 4.2. Continued.

Pre-treatment	Sucrose pulse (%)	Vase life (days)	Flowers opened during treatment (%)	Senesced flowers at termination (%)	Q 3[z]	Q 6	Q 9
-	0.0	8.1	26.0	2.6	4.9	4.0	2.4
-	5.0	9.5	22.8	11.1	4.4	3.9	3.1
-	10.0	7.3	9.5	28.8	3.9	3.5	2.7
Significance							
Pretreatment		0.0230	NS	NS	NS	NS	NS
Pulse (S)							
Linear (L)		NS	0.0006	0.0001	0.0001	0.0121	NS
Quadratic (Q)		0.0111	NS	NS	NS	NS	0.0135
U*SL		NS	NS	**	***	NS	NS
U*SQ		NS	NS	NS	NS	NS	NS
38*SL		***	NS	***	***	**	NS
38*SQ		***	NS	***	***	**	NS

Table 4.2. Continued.

Pre-treatment	Sucrose pulse (%)	Vase life (days)	Flowers opened during treatment (%)	Senesced flowers at termination (%)	Q 3[z]	Q 6	Q 9
M*SL		NS	NS	NS	0.01	0.05	NS
M*SQ		NS	NS	NS	NS	NS	NS

[NS], **, *** Nonsignificant or significant at $P \geq 0.05$ or 0.01, respectively

[z] Quality ratings (0 to 5, with 5 the best) determined at 3 (Q3), 6 (Q6), and 9 (Q9) days after end of treatment

Table 4.3. Effect of 24-h unheated or heated water or 1-methylcyclopropene (1-MCP) pretreatments and 0.0, 5.0, or 10.0% sucrose pulses on chroma meter readings for *Campanula medium* 'Champion Pink'. Values shown are means of twelve stems.

Pretreatment	Sucrose pulse (%)	1st flower value	2nd flower value	Value difference	1st flower hue	2nd flower hue	Hue difference	1st flower chroma	2nd flower chroma	Chroma difference
Unheated	0.0	59.1	66.9	-7.9	13.3	3.8	9.5	-5.2	1.3	-6.5
Unheated	5.0	59.1	68.0	-9.0	11.7	7.0	4.7	-4.1	-2.0	-2.2
Unheated	10.0	60.1	64.0	-3.9	10.5	7.4	3.1	-4.3	-2.4	-1.9
38 °C	0.0	56.9	64.0	-7.1	14.6	3.7	10.9	-4.8	1.3	-6.1
38 °C	5.0	60.8	64.8	-4.0	11.8	8.0	3.7	-4.4	-2.0	-2.4
38 °C	10.0	62.4	67.9	-5.5	10.1	3.7	6.4	-3.8	-0.9	-2.9
1-MCP	0.0	56.7	64.7	-8.0	14.1	5.3	8.8	-5.0	-0.7	-4.5
1-MCP	5.0	60.5	65.7	-5.2	11.5	7.5	4.0	-4.5	-2.3	-2.2
1-MCP	10.0	62.5	67.4	-4.9	10.6	5.9	4.7	-4.2	-1.1	-3.2
Unheated	-	59.4	66.5	-7.2	12.0	5.9	6.1	-4.6	-0.8	-3.9
38 °C	-	59.3	64.8	-5.5	12.8	5.7	7.0	-4.5	-0.5	-4.1
1-MCP	-	59.8	65.9	-6.1	12.1	6.2	5.8	-4.6	-1.4	-3.3

Table 4.3. Continued.

Pretreatment	Sucrose pulse (%)	1st flower value	2nd flower value	Value difference	1st flower hue	2nd flower hue	Hue difference	1st flower chroma	2nd flower chroma	Chroma difference
-	0.0	57.6	65.3	-7.7	14.0	4.3	9.6	-5.1	0.6	-5.6
-	5.0	60.2	66.1	-6.0	11.7	7.5	4.1	-4.4	-2.1	-2.3
-	10.0	61.6	66.1	-4.6	10.5	6.3	4.2	-4.2	-1.6	-2.7
Significance										
Pretreatment (P)		NS	NS	NS	NS	NS	NS	NS	NS	NS
Pulse (S)										
Linear (L)		0.0020	NS	0.0172	0.0016	NS	0.0001	0.0493	0.0135	0.0012
Quadratic (Q)		NS	NS	NS	NS	0.0346	0.0050	NS	0.0308	0.0172
P*SL		NS	0.0320	NS	NS	NS	NS	NS	NS	NS
P*SQ		NS	NS	NS	NS	NS	NS	NS	NS	NS

[NS] Nonsignificant or significant at $P \geq 0.05$.

Table 4.4. Effect of floral foam substrate and 0, 0.5, 1.0, 2.0, or 4.0% sucrose solution on postharvest quality of *Campanula medium* 'Champion Blue'. Values shown are means of fifteen stems.

Substrate	Sucrose (%)	Vase life (days)	Senesced flowers at termination (%)	Q 3[z]	Q 6	Q 9
Foam (F)	0.0	2.8	0.6	3.7	--[y]	--
Foam	0.5	2.3	0.0	3.3	--	--
Foam	1.0	2.4	1.2	3.5	--	--
Foam	2.0	3.7	3.9	4.0	3.2	--
Foam	4.0	5.2	2.4	4.4	3.3	--
No-foam (NF)	0.0	8.1	0.7	4.9	4.7	3.4
No-foam	0.5	9.4	1.5	5.0	4.9	4.0
No-foam	1.0	11.7	1.9	4.9	4.7	4.1
No-foam	2.0	12.2	6.0	4.9	4.8	4.0
No-foam	4.0	8.6	46.2	4.7	4.3	3.4
Significance						

Table 4.4. Continued.

Substrate	Sucrose (%)	Vase life (days)	Senesced flowers at termination (%)	Q 3[z]	Q 6	Q 9
Substrate		0.0001	0.0010	0.0001	0.0001	--[y]
Solution (S)						
Linear (L)		0.0152	0.0001	0.0007	0.0014	NS
Quadratic (Q)		0.0017	0.0157	NS	NS	0.0004
Cubic (C)		NS	NS	0.0176	NS	NS
F*SL		***	NS	***	--	--
F*SQ		NS	NS	NS	--	--
F*SC		NS	NS	***	--	--
NF*SL		NS	***	NS	--	--
NF*SQ		***	***	NS	--	--
NF*SC		NS	NS	NS	--	--

[NS], **, *** Nonsignificant or significant at $P \geq 0.05$ or 0.01, respectively

[z] Quality ratings (0 to 5, with 5 the best) determined at 3 (Q3), 6 (Q6), and 9 (Q9) days after end of treatment

[y] Insufficient number of stems were available to provide a quality rating or statistical analysis.

Table 4.5. Effect of high (110 μmol.m^{-2}.s^{-1}) or low (10 μmol.m^{-2}.s^{-1}) light intensity on chroma meter readings of *Campanula medium* 'Champion Blue'. Values shown are means of fifteen stems.

Light intensity	1st flower value	2nd flower value	Value difference	1st flower hue	2nd flower hue	Hue difference	1st flower chroma	2nd flower chroma	Chroma difference
High	32.9	51.0	-18.1	19.4	10.2	9.3	-23.7	-13.9	10.2
Low	34.4	60.0	-25.6	21.7	5.7	16.0	-25.5	-7.7	2.2
Significance									
Light	NS	0.0338	NS	NS	0.0373	0.0050	NS	0.0278	0.0126

NS Nonsignificant or significant at $P \geq 0.05$.

Table 4.6. Comparison of deionized water (DW) control solutions on flower quality for *Campanula medium* 'Champion Blue'. Values shown are means of fifteen stems.

Solution	Q 6 [z]
DW, pH 4.0	4.2 b [y]
DW, pH 3.5	4.0 b
DW, pH 4.0 + 200 mg.L^{-1} 8-HQC	4.7 a
DW, pH 3.5 + 200 mg.L^{-1} 8-HQC	4.8 a
Significance	
Solution	0.0019

[y] Mean separation within columns by Duncan's, $P \geq 0.05$.
[z] Quality ratings (0 to 5, with 5 the best) determined at 6 (Q6) days after end of treatment

CHAPTER V

SUMMARY

For *Tagetes erecta* seedling field emergence, cultivar differences were evident in measured emergence parameters and flower harvest data for each of three years examined, but results were inconsistent from year to year. Early sowing delayed days to first seedling emergence and mean emergence rate. Results from 1999 indicated 29 Apr. as an optimum sowing date in Oklahoma for direct-seeded *T. erecta* plants for both emergence parameters and total flower yield. Wet priming was superior to solid matrix primed and unprimed seed for mean emergence rate, emergence uniformity, and total emergence percentage.

'E-1236' was the top producer for flower number and mass of fresh flower, dried flower, dried petal, and pigment for *T. erecta* commercial production. Transplanted plants yielded higher quantities for all data parameters as compared to direct-seeded plants for three years. Surprisingly, nitrogen application created mixed results by increasing flower diameter and reducing dried receptacle mass. However, nitrogen application did not increase pigment yield but did increase the percentage of dead plants. Hand-harvesting flowers greatly increased yields compared with hedge-trimming. We would recommend hand-picking 'E-1236' transplanted plants for commercial *T. erecta* production based upon these results. Supplemental nitrogen fertilization did not increase pigment harvest and is not recommended for commercial production.

Optimum postharvest quality and color of cut *Campanula medium* 'Champion' stems was obtained with each of the following individual treatments. Wet storage at 2°C for one or two weeks, a 4-h 38°C floral solution (deionized water, amended to pH 3.5 using citric acid and 200 ppm 8-HQC) pretreatment combined with 24-h 5.0% sucrose pulse, no-foam with either continuous 1.0% or 2.0% sucrose vase solution, and high light (110 $\mu mol.s^{-1}.m^{-2}$) conditions in the postharvest area had the best results of the treatments examined. Combinations of these treatments should be further tested to develop the best handling method for *C. medium* cut flower postharvest care.

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APPENDIX A

Sample Processing

1. After flowers were completely dry and crisp to the touch, dried flower mass was recorded.

2. Dried petals were removed from the flower by hand, and dried petal and receptacle mass were separately recorded. Dried petals were placed in a polyethylene bag and frozen for further analysis.

Petal Moisture Content

1. Dry aluminum pans in 120°C oven for 30-min. Use 3 replications per sample.

2. Remove pans from oven to dessicator and allow to cool for 15-min.

3. Weigh pan. Add sample petals, record weight, and place back inside dessicator.

4. Place pans inside 120°C oven for 24-h. Remove pans from oven to dessicator and allow to cool for 15-min.

5. Weigh pan with petal. Use masses to calculate the petal moisture content percentage.

Sample Handling

1. Randomized samples were removed from the freezer and placed in the dark until warmed to room temperature (approximately 30-min). Total petal sample mass was weighed, and placed in a dessicator for moisture content and pigment analysis. Samples remained in the dessicator for about 16-h.

2. Samples were weighed again and ground into a fine powder using a UD cyclone sample mill (Part # 3010-030, UD Corp., Boulder, CO).

Powder Moisture Content

1. Dry aluminum pans in 70°C oven for 30-min. Use 3 replications per sample.

2. Remove pans from oven to dessicator and allow to cool for 15-min.

3. Weigh pan. Add sample powder, record weight, and place back inside dessicator.

4. Place pans inside 70°C oven for 2-h. Remove pans from oven to dessicator and allow to cool for 15-min.

5. Weigh pan with powder. Use masses to calculate the moisture content percentage.

Pigment Analysis

1. Three 50-mg repetitions from each powder sample were placed into separate 100-ml volumetric flasks. Thirty-ml of extractant (10-ml hexane, 7-ml acetone, 6-ml absolute alcohol [200 proof ethyl alcohol], and 7-ml toluene) was pipetted into each flask, stoppered, and swirled for 1-min.

3. 25-ml deionized water was pipetted into each flask, stoppered, and swirled for 1-min.

4. 2-ml 40% methanolic potassium hydroxide was pipetted into each flask, swirled 1-min, stoppered, and placed in a 56°C water bath for 20-min.

5. Sample was cooled on ice in the dark for approximately 5-min. It was then removed from the ice and allowed to stand in the dark for 1-h.

6. 30-ml hexane was pipetted into the flask, swirled 1-min, diluted to volume with 10% sodium sulfate solution, and shaken for 1-min.

7. Sample stood in the dark for 1 more h.

Chromatography

1. Chromatographic tube (Kontes 10.5 x 300 mm., Corning, Inc., Corning, NY) was fitted on a vacuum flask with a small quantity of glass wool in the bottom. Adsorbent (equal amounts of silica gel G and diatomaceous earth) was added until approximately 12-cm layer had formed. Full vacuum was applied and more adsorbent added until a 7-cm layer formed.

2. Anhydrous sodium sulfate was added until a 2-cm layer formed.

3. 2-ml eluant (80 hexane:10 acetone:10 methanol) was added to the chromatographic tube as last of the solution was entering adsorbent. 2-ml of sample upper phase was added.

4. 5-ml eluant was added to tube after sample. Vacuum was turned on, and flow adjusted to 2 or 3 drops per second.

5. After drops slowed to 2 or 3 per minute, vacuum was turned off and flask removed. Contents were swirled and poured into 25-ml volumetric flask. Vacuum flask was rinsed with 6-ml eluant and added to 25-ml volumetric flask.

6. 25-ml volumetric flask was brought up to volume with eluant, inverted several times, and placed in dark to attain room temperature.

Spectrophotometer

1. Sample was put into a 1-ml quartz cuvette, and absorbance measured using a Shimadzu UV 160U, UV-visible recording, spectrophotometer (Shimadzu Corp, Kyoto, Japan) set at 474 nm.

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