

# Role of Ethanol in the Vase Life and Ethylene Production in Cut *Lisianthus* (*Eustoma grandiflorum* Mariachii. cv. Blue) Flowers

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## ABSTRACT

Effect of ethanol (2, 4 & 6%) combined with 2.5% sucrose on lisianthus (*Eustoma grandiflorum* Mariachii. cv. blue) cut flowers was studied. The vase were placed in chambers at 25°C, relative humidity about 70% and 14h photoperiod that was maintained using fluorescent lamps (light intensity of 15  $\mu\text{mol m}^{-2} \text{s}^{-1}$ ) at the top of the corolla. Data were recorded for vase life, fresh weight, ethylene production rate, solution uptake over time and analyzed statistically. Results revealed that 2% ethanol along with 2.5% sucrose were the most effective on vase life of lisianthus cut flowers. Use of ethanol inhibited ethylene production and increased water uptake

**Key Words:** Lisianthus cut flower; Ethanol; Vase life; Ethylene production

## INTRODUCTION

*Eustoma grandiflorum* (Raf.) Shinn is native to the southern US and mainly inhabits the moist prairies from Nebraska to Colorado and Texas. *Eustoma* was introduced into Japan more than 60 years ago (Ohkawa *et al.*, 1991). *Eustoma grandiflorum* (lisianthus) hybrids have continued to gain acceptance as new cut flowers, bedding plants, and potted flowering plant since their introduction promoted the floriculture trade in the early 1980s (Halevy & Kofranek, 1984). Short postharvest vase life is one of the most important problems on the cut flowers. However, longevity of vase life is an important factor in consumer preference (Kader, 2003; da Silva, 2003). Senescence of cut flowers is induced by several factors, e.g., water stress (Sankat & Mujaffar, 1994), carbohydrate depletion (Ketsa, 1989), micro-organisms (Witte & van Doorn, 1991) and ethylene effects (Wu *et al.*, 1991; Han & Miller, 2003). In carnations, senescence of the petals is associated with a climacteric-like increase in ethylene production during the final stages. This increase in ethylene production is accompanied by the irreversible inrolling, wilting and abscission of the petals (Nichols, 1979; Kim *et al.*, 2005). Ethylene production of cut *Eustoma* flowers increased with flower senescence and Treatment with silver thiosulphate (STS), an ethylene action inhibitor, extended flower longevity (Ichimura *et al.*, 1998). However, it contains silver which is a possible environmental pollutant. Ethanol has been found to be effective in increasing the vase life of carnation flowers by inhibiting ethylene biosynthesis (Heins & Blakely, 1980; Wu *et al.*, 1992) as well as its action (Wu *et al.*, 1992). Pun *et al.* (1999) found that using 4% ethanol on the vase solutions of carnation increase postharvest longevity. Sucrose is the major form of photosynthetically

assimilated carbon that is transported in plants (Lalonde *et al.*, 1999). Han (2003) found that addition of sugar to vase solution improved the intensity of the petal color but did not improve bud opening, longevity, or size of non-cold-stored Oriental Lily cv. Stargazer cut flower harvested at the commercial marketing stage. However, addition of sugar to the vase solution of defoliated stems not only restored the color on the petals but increased the size of the open flowers (Han, 2003). A floral preservative solution containing aluminum sulfate at 150mg L<sup>-1</sup> under 25°C, extend cut eustoma (*Eustoma grandiflorum* Shinn. cv. Hei Hou) vase life (Liao *et al.*, 2001). The purpose of this work was to find responses of the cut *Eustoma* flowers to ethanol application and its effect on vase life and ethylene production.

## MATERIALS AND METHODS

This study on the effect of ethanol treatments on vase life and some quality attributes of lisianthus cut flowers, in completely randomized design with three replications. Cut flower stems of eustoma (*Eustoma grandiflorum* Mriachii cv. Blue) (40cm in length) were placed in solution containing ethanol at 2, 4 and 6% after cutting. Four cut flowers (each cut stem contained 1-2 flowers) were placed in a 500mL flask with 400mL of solution. Distilled water was used for the controls. In all treatments, except controls, 2.5% sucrose was used and placed in chambers at 25°C. The relative humidity was about 70% while 14h photoperiod was maintained using fluorescent lamps with a light intensity of 15  $\mu\text{mol m}^{-2} \text{s}^{-1}$  at the top of the corolla. Data were statistically analyzed using SAS software. The vase life of cut flowers was completed when the petals or stem below the flower head lost turgidity.

**Ethylene production.** For measurement of ethylene

produce in flowers, one flower was sealed in a 300mL flask vessel. All vessels were sealed and kept at 25°C. After 2h, 1mL gas samples were withdrawn with a hypodermic syringe for ethylene determination. Ethylene content was measured using a Shimadzu Gas Chromatograph equipped with an activated alumina column fitted with a flame ionization detector. Nitrogen was used as a carrier gas. Measurements were repeated three times. Ethylene production was calculated as  $\text{nl g}^{-1} \text{h}^{-1}$ .

**Water uptake and fresh weight.** The volume of water uptake was calculated by subtracting the volume of water evaporated from a flask of the same volume without cut flowers. The fresh weight of the cut flowers also measured daily.

## RESULTS

**Vase life.** Flower stems kept in water containing ethanol had significantly increased vase life relative to the water control for all concentration of ethanol (Table I). The use of 2% ethanol along with 2.5% sucrose resulted in a greater extension in vase life than other treatments.

**Water uptake and fresh weight.** From day 3 of vase life, water uptake rate decreased for all solutions tested except treatment of 2% ethanol (Fig. 1). In solution containing 2% ethanol, water uptake increased to 6 day after treatment and then decreased. Uptake rate decreased rapidly in deionized water and solutions containing 4, 6% ethanol while flowers in the solutions containing 2% ethanol showed the minimum decrease in water uptake rate from day 6 (Fig. 1). However, average of water uptake, from beginning treatment to control flower wilting, in solutions containing ethanol except 6% ethanol, were significantly higher than control (Table II). Solution uptake was higher for 2% ethanol than the other concentrations. Fresh weight of cut flowers in all treatments increased during earlier and then declined (Fig. 2) From 3 day of vase life, fresh weight of cut flowers decreased for control and 6% ethanol. In 2 and 4% ethanol, fresh weight of cut flowers increased up to 6 days and decreased for ethanol 4% and constant to 12 day for ethanol 2% and then decreased. Means of dray weight of leaves in various ethanol-containing vase solutions was significantly lower than control (Fig. 3).

**Ethylene production.** Treatment with ethanol decreased ethylene production in all concentrations ( $p < 0.01$ ). Ethylene production rate began to decrease earlier and then increased in during of senescence. Eustoma flowers showed typical climacteric patterns of ethylene production during senescence (Fig. 4). Highest means of ethylene production inhibition was found with 6% ethanol (Table II), but this caused discoloration and breakdown of stems after 10 days as well.

**Percentage of flower opening, wilting and bending.** Percentage of flower and bud bending were lower in ethanol than controls (Table III). The lowest percentage of flower and bud bending were observed at 2% ethanol. Percentage

**Table I. Vase life at 25 °C of Eustoma cut flowers containing immersed in the water containing ethanol.**

Treatments	Flower vase life (days)
Control 0%	9.33 d
Ethanol 2%	15.00 a
Ethanol 4%	13.66 b
Ethanol 6%	12.33 c

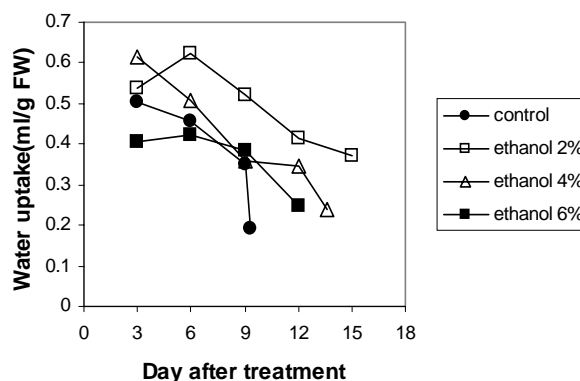
\*Means followed by the same letter are not significantly different at 1% level, using DNMR.

**Table II. Means of water uptake, water loss and ethylene production rate from beginning treatment to control flower wilting, in solutions containing ethanol**

Treatment	Means of water up take ( $\text{ml g}^{-1} \text{FW}$ )	Means of water loss ( $\text{ml g}^{-1} \text{FW}$ )	Means of ethylene production ( $\text{nl g}^{-1} \text{FW h}^{-1}$ )
Control 0%	0.437 c	0.462 b	2.729 a
Ethanol 2%	0.560 a	0.496 a	1.843 b
Ethanol 4%	0.494 b	0.440 c	1.803 bc
Ethanol 6%	0.404 d	0.370 d	1.634 c

\*Means followed by the same letter are not significantly different at 1% level, using DMRT.

**Fig. 1. Effect of various ethanol-containing vase solutions on the average of water uptake during vase life.**



of flower opening and longevity vase life of open flowers on solutions containing ethanol along with 2.5% sucrose were greater than control (Table III).

## DISCUSSION

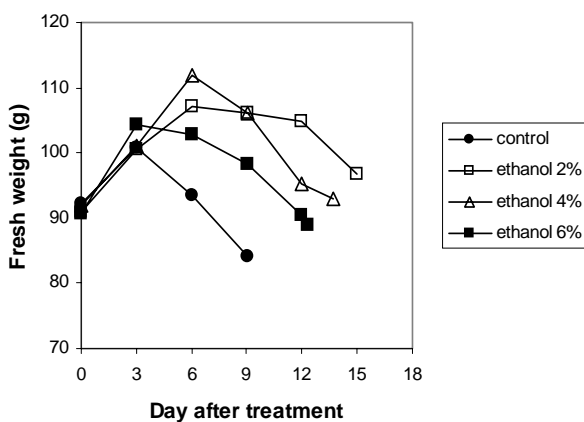
It has been suggested that the presence of microorganisms in water can cause physical plugging of the cut stem, release toxic metabolites and/or enzymes, evolve damaging levels of ethylene and induce a hypersensitive response resulting in PCD (Alvarez, 2000). Use of disinfectants improve water conductance by preventing bacterial growth and reducing occlusions (van Doorn,

**Table III. Effect of 2% ethanol-containing vase solutions on the percentage of flower and bud bending, flower and bud opening, semi open flowers and flower wilting of lisianthus cut flowers.**

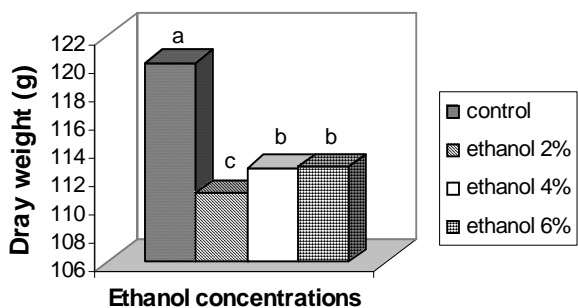
Day after treatment	percentage of flower and bud bending		percentage of flower and bud opening		percentage of semi open flowers		percentage of flowers wilting	
	Control 0%	Ethanol 2%	Control 0%	Ethanol 2%	Control 0%	Ethanol 2%	Control 0%	Ethanol 2%
0	0	0	21.02	20.27	1.66	2.62	0	0
3	1.63	0	22.68	20.27	3.27	2.62	0	0
6	18.03	0	14.72	21.60	6.45	6.37	7.96	0
9	24.59	5.05	6.6	19.17	6.45	10.08	16.08	5.05
10	63.93	-----	3.42	-----	3.27	-----	22.44	-----
12	-----	6.33	-----	16.50	-----	8.8	-----	9
15	-----	16.41	-----	7.41	-----	7.56	-----	19.33

1998). We found that the Eustoma cut flowers treated with 2 and 4% ethanol indicated more water than those of controls. This allowed more increase in fresh weight, mainly due to better corolla development (Table II & III, Fig. 1 & 2). However, water uptake in cut flowers treated with 6% ethanol continued for longer time than those of control. These results were in agreement with reports by van Doorn (1997) who indicated that vascular blockage causes a state of water deficit and thereby shortens the vase life.

**Fig. 2. Effect of various ethanol-containing vase solutions on the average of fresh weight during vase life**

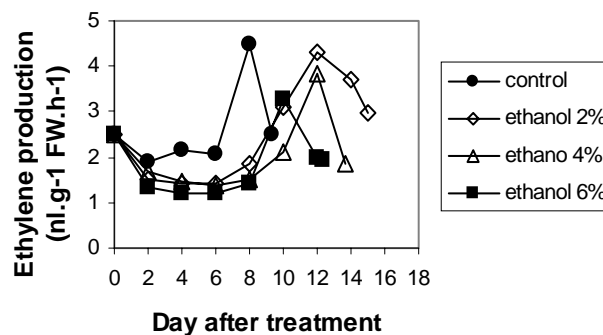


**Fig. 3. Effect of various ethanol-containing vase solutions on the average of dray weight of leaves from beginning of treatment to control wilting**



The present studies have reconfirmed the role of ethanol for increasing the vase life of carnation flowers by inhibiting ethylene production. Treatment with 4% ethanol inhibited ethylene production in carnation flowers as well as improved sensitivity against ethylene (Pun *et al.*, 1999 2001). It was found that treatment Eustoma cut flowers with solutions containing ethanol can inhibit ethylene production, which was greatest at 6%. However, it caused discoloration and breakdown of stems after 10 days and reduced vase life. Ichimura and Hiraya (1999) reported that treatment with sucrose extends the vase life of florets harvested at a bud stage. In addition, sucrose promotes pigmentation of petal colors in some cut flowers including Eustoma. Keeping the flowers in vase solutions containing sucrose has been shown to extend their vase-life (Han, 2003; Yamane *et al.*, 2005). Cut flowers treatment with sugars increases the availability of respirable substrates (Paulin, 1986; Huang & Chen, 2002; da Silva, 2003), delay the onset of hydrolysis of structural cell components (Parups & Chan, 1973; Donoghue *et al.*, 2002), decrease ethylene production and sensitivity (Huang & Chen, 2002; Pun *et al.*, 2005). Sucrose can act as a source of nutrition for tissues approaching carbohydrate starvation it may also act as an osmotically active molecule thereby having a role in flower opening and subsequent water relations (Kuiper *et al.*, 1995). These data showed that using solution containing 2% ethanol combined with 2.5%

**Fig. 4. Effect of various ethanol-containing vase solutions on the average of ethylene production rate during vase life**



sucrose substantially extended the vase life. Role of ethanol as a disinfectant, in improved water conductance and reducing occlusions is plausible.

In conclusion, all concentration of ethanol along with 2.5% sucrose improved significantly the vase life of *Lisianthus* cut flowers. Treatment with solutions containing 2% ethanol along with 2.5% sucrose increased vase life by delayed senescence.

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