

## Thidiazuron Effects on *Dianthus caryophyllus* 'Lunetta'

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### Abstract

Experiments were conducted to determine the effects of Thidiazuron (TDZ) applied preharvest under glasshouse conditions on *Dianthus caryophyllus* 'Lunetta'. Thidiazuron at 0, 1, 10, 100, and 1000  $\mu\text{M}$  was applied as a foliar spray in two separate experiments. 15 plants were used for each concentration in both experiments. Experiments were arranged in completely randomized designs. Time to flowering was recorded, and relative stem length, total nitrogen, and tissue water content were measured at harvest. Postharvest vase life, relative fresh weight changes, and solution uptake changes were also measured. TDZ treatments sometimes decreased relative stem length compared to the control (0  $\mu\text{M}$ ). TDZ treatment tended to decrease total nitrogen and water content of tissues slightly, but not significantly ( $P > 0.05$ ). In the first experiment, TDZ at 100  $\mu\text{M}$  significantly increased the vase life of cut carnation flowers compared to the control. In the second experiment, TDZ applied preharvest at both 10 and 100  $\mu\text{M}$  significantly increased their vase life. TDZ treated flowers tended to maintain higher relative fresh weight, with positive differences for the 100  $\mu\text{M}$  TDZ treatment being apparent at days 5, 7 and 9 of vase life. Solution uptake was sometimes higher in TDZ treated flowers.

### INTRODUCTION

Various studies have demonstrated improvement of the postharvest life of cut flowers as a result of cytokinin treatments (e.g. Lukaszewska et al., 1994; Paull and Chantrachit, 2001). However, the effects of preharvest treatment effects on postharvest characteristics of cut flowers are largely unexplored. Preharvest variables can be an important determinant of the quality and longevity of cut flowers and foliage (Celikel and Karaaly, 1995). For example, mean relative humidity in the greenhouse is an important variable accounting for vase life differences in the one cultivar produced in different greenhouses (Marissen and Benninga, 2001).

Thidiazuron (N-phenyl-N-1,2,3-thidiazol-5-ylurea; TDZ) is a relatively novel urea-type compound with plant growth regulator-like activity. It has been used commercially at high concentrations as a cotton defoliant (Malik et al., 2002). At low concentrations, it has been used for regeneration in tissue culture (Singh and Simal, 2001). TDZ appears to be stable in plant tissues (Mok et al., 2000; Ferrante et al., 2002a). Although the exact mode of TDZ action is not known, it has been reported to modulate cytokinin biosynthesis and/or metabolism (Mok et al., 2000), with effects on ancillary endogenous hormones, proteins, and enzymes (Murthy et al., 1998). TDZ seems to mimic both auxins and cytokinins (Murthy et al., 1998). TDZ is around 50-100 times more active in inducing cytokinin-like effects than common cytokinins (Genkov and Iordanka, 1995).

Bosse and Van Staden (1989) reported that DHZ (dihydrozeatin) used as a pulse treatment for 6-24 h at a concentration of  $2 \times 10^{-4}$  M significantly delayed carnation flower senescence. An increase in the longevity of carnation flowers by 15 d was found at  $4 \times 10^{-6}$  M. Lukaszewska et al. (1994) showed that exogenous application of zeatin, zeatin

ribiside, 2iP, and 2iPA in holding solutions delayed rose senescence by 34-56%, and thereby prolonged longevity. Zeatin and zeatin ribiside were most effective on roses at  $1 \times 10^{-7}$  M, and prolonged rose longevity by 13 d. Treatments with TDZ effectively prevent leaf yellowing in alstroemeria (Ferrante et al., 2002a). TDZ was most effective as a 10  $\mu$ M pulse treatment or at 1  $\mu$ M as a continuous treatment. TDZ treatment was also useful on phlox inflorescences at  $< 50 \mu$ M (Sankhala et al., 2003a). Treatment with TDZ greatly reduced flower shedding and induced additional flower buds during vase life. The efficacy of TDZ treatments depends upon the genotype and the concentration. TDZ treatment of chrysanthemum and tulip inhibited leaf yellowing, but did not enhance the quality of the flowers (Ferrante et al., 2003). TDZ in comparison with BA had little effect on vase life of cut *Eucalyptus parvifolia* (Ferrante et al., 2002b). Pulse treatments with 10  $\mu$ M TDZ decreased the vase lives of 'Champagne', 'Laser', 'Magnum' and 'Neon' roses, but slightly increased that of 'Tresor 2000' (Chamani et al., 2006). A 10  $\mu$ M TDZ pulse treatment increased the vase life of 'First Red' rose, and TDZ in combination with sucrose had a greater positive effect on vase life than either TDZ or sucrose alone.

The present study was conducted to examine effects of preharvest TDZ treatments on the physicochemical characteristics of *Dianthus caryophyllus* 'Lunetta' during the subsequent pre- and postharvest periods.

## MATERIALS AND METHODS

*D. caryophyllus* 'Lunetta' plantlets were obtained from a commercial greenhouse (Pakdasht, Tehran) and transported to the research greenhouse of Mohaghegh Ardebil University, Ardebil. Plants were potted-up into media comprised of 25% sand, 25% cattle manure, and 50% farm soil. 3 weeks after planting, the plants were pinched-back to encourage uniformity of flowering. 6 weeks after pinching, plants were treated with foliar sprays of TDZ. TDZ (Sigma Chemical Co.) was dissolved in 2 ml of 1 M KOH and prepared at 0, 1, 10, 100, and 1000  $\mu$ M concentrations. pH was neutralized with 2 mL 1 M HCl. The experiment was duplicated, and 15 plants for each TDZ concentration were used in both experiments. Experiments were arranged in completely randomized designs. The traits measured were: time to flowering; relative stem length (%), total nitrogen, and tissue water content at harvest; and, vase life, relative fresh weight changes (%), and solution uptake changes after harvest. Flowers were harvested at the fully open stage (Eisinger, 1977). Postharvest experiments were carried out under vase life evaluation room conditions of  $22 \pm 2^\circ\text{C}$ , 60-70% relative humidity (RH) and 12 h photoperiod with cool white florescent lamps. The harvested flowers were stood into vases containing distilled water and 10  $\mu$ l L<sup>-1</sup> chlorine (a.i.).

### Preharvest Assessments

Time to flowering was recorded as days from TDZ application to full flower opening (harvest). Relative stem length percentage (RLP, %) for stems was calculated using the formula:  $\{(L_t - L_0) / L_0\} \times 100$ ; where,  $L_t$  = stem length (cm) at time t (week) and  $L_0$  = stem length (cm) at time 0. Time 0 was days of treatment. Water content of tissues (WCT, %) was calculated using the formula:  $\{(W_t - W_d) / W_t\} \times 100$ ; where,  $W_t$  = fresh weight of the stem (g) on day 0 (day 0 was harvest day) and  $W_d$  = dry weight of the stem (g) at the end of vase life. Total N was determined in dried tissue samples of 1 g d.w. by the Kjeldhal method using concentrated H<sub>2</sub>SO<sub>4</sub>, K<sub>2</sub>SO<sub>4</sub> and Selenium to digest the sample.

### Postharvest Assessments

Vase life was recorded as the time (days) after harvest (day 0) that flowers showed petal in-rolling, which was taken as the first sign of flower senescence (Bosse and Van Staden, 1989). Relative fresh weight (RFW, %) for stems was calculated using the formula:  $(W_t / W_{t=0}) \times 100$ ; where  $W_t$  = weights of stems (g) on days 0, 2, 4, 6, etc. and  $W_{t=0}$  = weight of the same stem (g) on day 0. Vase solution usage (ml / day / g f.w.) was determined using the formula:  $(S_{t-1} - S_t) / W_{t=0}$ ; where,  $S_t$  = solution weight (g) at t = days

1, 2, 3, etc.  $S_{t-1}$  = solution weight (g) on the previous day, and  $W_{t=0}$  = fresh weight of the stem (g) on day 0.

### Statistical Analyses

Data were analyzed with Minitab Release 13.1 for Windows (Minitab Inc.). The Duncan's Multiple Range Test (DMRT;  $P = 0.05$ ) was used for comparisons of treatment means. Least significant difference (LSD;  $P = 0.05$ ) values were also calculated.

## RESULTS AND DISCUSSION

In first experiment (data not shown), plants treated with 1000  $\mu\text{M}$  TDZ tended to produce flowers later than in most other treatments, but the minor effect was not significant ( $P > 0.05$ ). No trends were evident for the other TDZ treatments compared to the control. In the second experiment, treatment with TDZ at 1000  $\mu\text{M}$  significantly ( $P < 0.05$ ) delayed flowering compared to 10 and 100  $\mu\text{M}$  TDZ treatments (Fig. 1). Thus, flowers were produced earliest with treatments of 10 and 100  $\mu\text{M}$  TDZ. TDZ treated flowers generally had proportionally lower stem lengths compared to the control (Fig. 2). Significant differences were initially apparent at 2 and 3 weeks after treatment; e.g. control vs. 100  $\mu\text{M}$  TDZ treated flowers. The lowest stem length percentage in the end was for plants treated with 1000  $\mu\text{M}$ . TDZ treatments tended to slightly decrease total carnation stem N and tissue water contents at harvest, but these effects were not significant (Tables 1 and 2).

TDZ applied at 100  $\mu\text{M}$  in the first experiment (data not shown) and at 10 and 100  $\mu\text{M}$  in the second experiment (Fig. 3) increased the vase life of cut carnation flowers compared to the 0  $\mu\text{M}$  control. TDZ treated flowers sometimes maintained a higher relative fresh weight, with significant differences at 5, 7, and 9 d after treatment between 100  $\mu\text{M}$  TDZ treated flowers and the control (Fig. 4). Highest relative fresh weight was specifically associated with plants treated with 100  $\mu\text{M}$  TDZ. Vase solution usage tended to be higher in TDZ treated flowers, although no significant differences were found between TDZ treated flowers and control (data not shown).

The vase life of cut 'Lunetta' carnation flower can be increased by preharvest application of around 10-100  $\mu\text{M}$  TDZ. Increasing the postharvest life of carnations with applied cytokinins has been reported previously (e.g. Eisinger, 1970; Shibuya et al., 2000). Bosse and Van Staden (1989) used DHZ as a pulse treatment for 6-24 h at  $2 \times 10^{-4}$  M to delay carnation flower senescence. Similarly, a 10  $\mu\text{M}$  TDZ pulse treatment increased the vase life of 'First Red' rose, which exhibited greater water uptake and higher relative fresh weight (Chamani et al., 2006). Increased vase life of cut 'Lunetta' carnations may be associated with solution uptake maintenance by TDZ and/or inherently low levels of cytokinins in their tissues. BA treatments can maintain water uptake (Paull and Chantarachit, 2001). In anthurium, a decline in water uptake was related to reduced vase life (Paull and Goo, 1985). Paull and Chantrachit (2001) reported that various *Anthurium* cultivars showed different vase life responses to BA treatment, from a 20% reduction to a 2.5-fold increase. They suggested that a lack of response to BA may be due to high natural levels of cytokinins already in the tissue. Sankhala et al. (2003b) observed that treatment with 50  $\mu\text{mol/L}$  TDZ increased the proportion of new flowers opening on cut phlox flowers after 7 days.

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## **Tables**

Table 1. Effect of different TDZ concentrations on the N content at harvest of ‘Lunetta’ carnation flower stems (LSD = 0.21) (experiment 2).

Treatment	Total Nitrogen (%)
Control	2.28
TDZ (1 $\mu$ M)	2.04
TDZ (10 $\mu$ M)	2.05
TDZ (100 $\mu$ M)	2.06
TDZ (1000 $\mu$ M)	2.15

Table 2. Effect of different TDZ concentrations on the water content of ‘Lunetta’ carnation flower stems (LSD = 9.27) (experiment 2).

Treatment	Water content (%)
Control	82.7
TDZ (1 $\mu\text{M}$ )	82.5
TDZ (10 $\mu\text{M}$ )	81.9
TDZ (100 $\mu\text{M}$ )	81.7
TDZ (1000 $\mu\text{M}$ )	82.2

### Figures

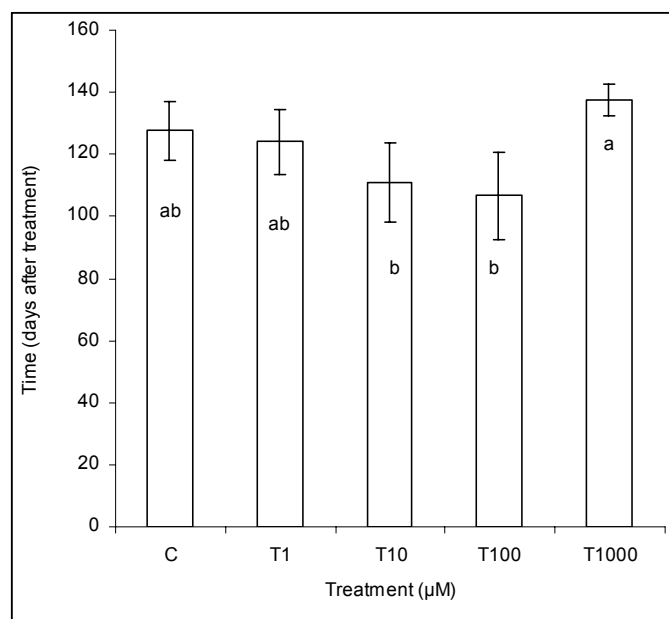


Fig. 1. Effects of different TDZ concentrations on the time to flowering of cut ‘Lunetta’ carnation plants (experiment 2).

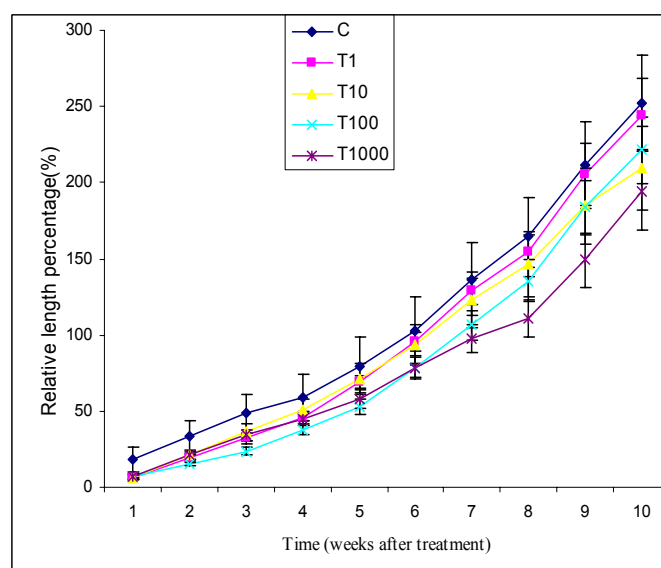


Fig. 2. Effects of different TDZ concentrations on preharvest changes in relative stem length percentage of cut ‘Lunetta’ carnation flowers (experiment 1).

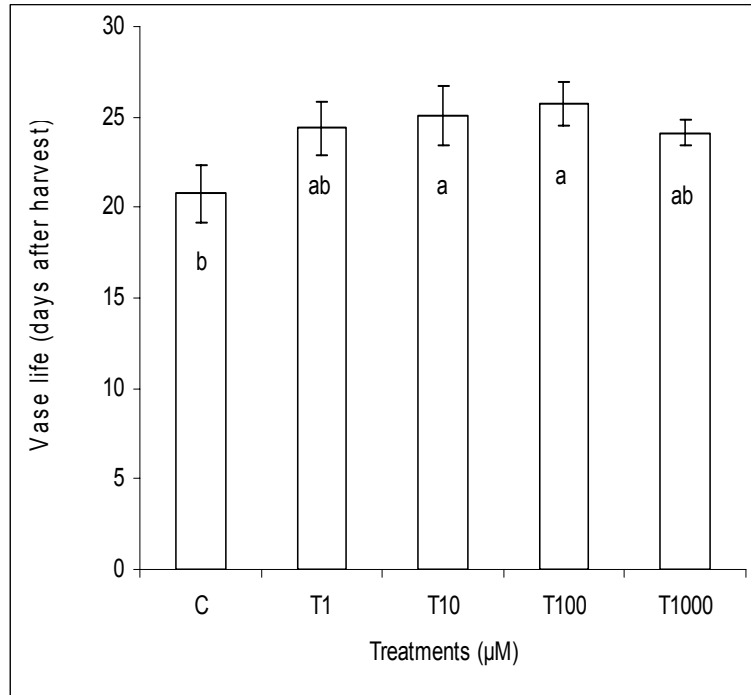


Fig. 3. Effects of different TDZ concentrations on the vase life of cut ‘Lunetta’ carnation flowers (experiment 2).

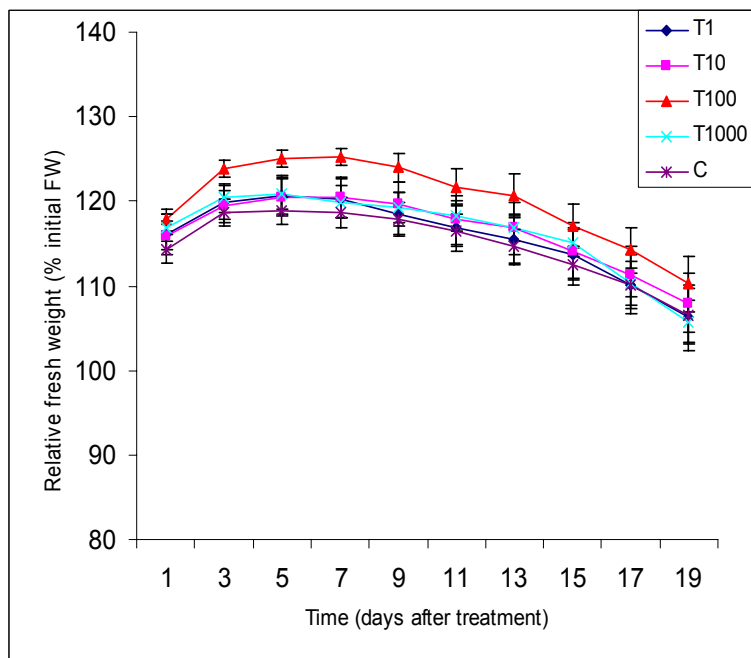


Fig. 4. Effects of different TDZ concentrations on postharvest changes in relative fresh weight of cut ‘Lunetta’ carnation flowers (experiment 2).

# **Genetic and New Varieties**

