Ethylene + GA₃ Sprays for Delaying Flower Bud Initiation in Chrysanthemum (*Dendranthema grandiflora* Tzvelv.) Stock Plants

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Summary. Our objective was to determine whether repeated applications of 2-chloroethylphosphonic acid (ethephon) + gibberellic acid (GA₃) to stock chrysanthemum plants that are day-neutral for flower bud initiation would increase the number of quality cuttings. Across five cultivars, there were no significant differences between controls and plants receiving 250 ppm ethephon in the total number of cuttings per plant. The percentage of cuttings with crown buds was greater for cuttings from controls than for ethephon-treated plants. Applying 500 ppm ethephon significantly reduced the number of cuttings. We conclude that chrysanthemum clones day-neutral for flower bud initiation and development with low long-day leaf number could be selected to form a 4 to 5 week production group.

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Typical year-round glasshouse production of chrysanthemums relies on the autumn-flowering cultivars (Cockshull, 1984), that is, cultivars short day (SD) for flower bud initiation (FBI) and flower bud development (FBD) (Okada, 1963), because applying SD by means of artificial shading allows for timing of the crop. With the general trend toward shortening production time of glasshouse crops, chrysanthemum producers are selecting cultivars fitting the summer-flowering classification, that is, day neutral (DN) for FBI and FBD (Okada, 1963). For example, 'Bright Golden Anne', 'Minngopher', 'Ruby Mound', and 'White Grandchild' initiate flowers rapidly, even under long days (LD) (Cockshull, 1975; Cockshull and Langton, 1984; Cockshull et al., 1979). Cultivars DN for FBI and FBD and with a low LD leaf number (LDLN) could initiate a new production class, yielding flowering plants 4 to 5 weeks from planting of rooted cuttings without using artificial SDs (Anderson et al., 1989).

A major problem in developing a clonally propagated, 4- to 5-week class of truly DN glasshouse chrysanthemums is the difficulty in producing cuttings on a commercial scale from plants with low LDLN that flower under virtually all glasshouse environments. A synthetic plant growth substance, 2-chloroethylphosphonic acid (ethephon) was reported to delay flower initiation in chrysanthemum (Carpenter and Carlson, 1972; Cockshull and Horridge, 1978; Cockshull et al., 1979; Kher et al., 1974). Applying ≥ 200 ppm ethephon to chrysanthemums with low LDLN under LD delayed or prevented FBI, causing Cockshull et al. (1979) to suggest using this technique to delay flower bud formation on stock plants and cuttings. Our objective was to determine whether repeated ethephon applications to stock chrysanthemum plants with low LDLN, which are DN for FBI (and in some cases FBD), would increase the number of quality cuttings by reducing flower bud formation.

Materials and methods

Field-grown plants of five garden chrysanthemum cultivars ('Burnt Copper', 'Grape Glow', 'Minngopher', 'Minnwhite', and 'Ruby Mound') and one breeding selection (MN 83-267-3)

were dug on 27 Oct. and planted in 15-cm pots (2570 ml) in a 1 soil : 2 sphagnum peat : 1 sand (by volume) soil mix. These plants were placed in a greenhouse with a 22 °C day/20 °C night (± 3 °C) regime. High-intensity discharge lights were used to extend the photoperiod to 18 h and as a source of supplemental light. Average irradiance levels were $400 \mu\text{mol}\cdot\text{s}^{-1}\cdot\text{m}^{-2}$. Plants remained under LD (18 h), non-inductive photoperiod, throughout this study. Three weeks after potting, 0, 250, and 500 ppm of 2-chloroethylphosphonic acid was applied as a foliar spray. One week later and 1 week after each flush of cuttings was taken, the three ethephon concentrations were reapplied, with 5 ppm gibberellic acid (GA₃) added to the ethephon. The GA₃ was added to counter the reduction in internode length caused by treating plants with ethephon. Terminal cuttings, 4.0 to 7.5 cm in length, were taken from stock plants in four flushes: 1 Dec., 21 Dec., 20 Jan., and 24 Feb. The number of cuttings were recorded for each flush, and average cutting fresh weight was determined for the third and fourth flushes. Ten cuttings per replicate from each treatment were selected randomly from flush 4 and were scored for flower or crown buds; percentage of cuttings with flower buds was calculated. The experimental design was a completely random design with two replicates. Data were analyzed using repeated measures analysis of variance (Systat, Evansville, Ind.) as this is the most appropriate statistical procedure for analysis of several measurements on the same experimental unit (SAS Institute, Cary, N.C.). Because multivariate and univariate tests for within-subject effects, flush, and its related interactions were the same, only the univariate analysis is presented. However, univariate tests for within-subject effects and interactions require certain patterns of covariance matrices, Type H covariances (Hunyh and Feldt, 1970). The data did not satisfy this assumption. When this occurs, an adjustment to numerator and denominator degrees of freedom can be used for testing significance levels for F tests. Two adjustments based on a degrees-of-freedom adjustment factor known as ϵ (epsilon) were used to determine significance levels for F tests. The first adjustment, Greenhouse-Geisser epsilon (G-G), represents a

maximum-likelihood of Box's ϵ factor (Greenhouse and Geisser, 1959) and is a conservative estimate. The second adjustment, Hunyh-Feldt epsilon (H-F), is constructed using unbiased estimators of the numerator and denominator and is a less conservative estimate than G-G (Hunyh and Feldt, 1976).

Results and discussion

Repeated measures analysis of variance revealed that the number of cuttings obtained from a flush was influenced by the number of cuttings taken from the previous flush, and there was a significant flush $\times$ cultivar interaction for number of cuttings per flush (Table 1). The lowest number of cuttings was obtained from the first flush, regardless of cultivar. Harvest from the second flush yielded roughly double the number of cuttings obtained from the first flush for all except 'Minngopher' (Table 2). The number of cuttings increased in each of the subsequent flushes for MN 83-267-3, 'Grape Glow', 'Minnwhite', and 'Ruby Mound', whereas they remained nearly constant for 'Burnt Copper' and 'Minngopher', hence the significant flush $\times$ cultivar interaction. Since repeated-measures analysis of variance revealed no significant interaction be-

tween flush and treatment or flush, cultivar, and treatment, the effects of cultivar and treatment (ethephon applications) were determined from univariate analysis. Cultivar and ethephon application significantly affected average cutting weight and total number of cuttings. The cultivar $\times$ treatment interaction was not significant (Table 1).

Number of cuttings within flushes 1 or 2 did not differ among treatments (Table 2). For flushes 3 and 4, fewer cuttings were obtained from plants treated with 500 ppm compared to the control and the 250-ppm treatment. A major factor contributing to the reduction in cutting counts was injury to the terminals of lateral branches on plants treated with 500 ppm ethephon. Injury appeared after applying ethephon following the second flush of cuttings and increased after the next application. Terminals were scorched and cuttings were brittle. As a result of this damage, many lateral branches did not develop sufficiently to be taken as a cutting for rooting. No physical damage was observed on lateral branches of control or plants treated with 250 ppm.

Total number of cuttings and average fresh weight per cutting were not correlated in this study. Cuttings

from control plants had significantly higher mean fresh weights than those from ethephon-treated plants (Table 2). However, 21% of cuttings from controls had flower buds, whereas only 3% of those from the plants treated with 250 ppm had flower buds. Thus, the number of marketable cuttings was highest from plants treated with 250 ppm ethephon (Fig. 1). The increased weight of floral tissue may account for the differences in cutting fresh weight observed between the control and the 250-ppm ethephon treatment. The reduced weight of cuttings from plants treated with 500 ppm ethephon was the result of damage to the terminal meristems of the shoots. These results indicate that treating stock plants with 250 ppm ethephon + 5 ppm GA₃ increased the number of marketable cuttings by reducing crown-bud formation on lateral branches. The average fresh weight per cutting varied with cultivar. MN 83-267-3, 'Minngopher', and 'Ruby Mound' produced the largest cuttings; the other cultivars produced significantly smaller cuttings.

'Grape Glow' produced the largest number of cuttings from flushes 2, 3, and 4 and produced the largest total number of cuttings per plant. 'Grape Glow' was the only cultivar that did not form terminal flower buds (Table 3). 'Minngopher' control plants produced >155 cuttings, but >21% of these cuttings had formed flower buds. Therefore, the number of usable cuttings was reduced to 122. Similar results were observed for the other cultivars, as terminal flower formation reduced the number of usable cuttings by 5% to 10%. Either 'Grape Glow' is not DN for FBI or more nodes must be formed before initiation occurs. The other cultivars initiated flowers rapidly under LDs. This has been reported for 'Minngopher' and 'Ruby Mound' (Cockshull et al., 1979).

The fact that the genotypes (cultivars) differ in response to removal of the terminal stem cuttings is an important consideration in selecting cultivars for a 4- to 5-week production period. Because the plants have been selected for low LDLN, the number of axillary buds available for the next flush of cuttings is limited. Genotypes exhibiting an increase in number of cuttings in consecutive flushes, such as, 'Grape Glow' and 'Minnwhite', may outyield genotypes selected for large

Table 1. Repeated measure analysis of variance for the effects of cultivar and ethephon treatment on cutting count per flush and analysis of variance for the effects of genotype and ethephon treatment on average cutting weight and total number of chrysanthemum cuttings

Source	df	Mean square	F	Adjusted <i>P</i> value	
				G-G ²	H-F ³
<i>No. cuttings/flush</i>					
Flush	3	3898.64	51.698	<0.001	<0.001
Flush × cultivar	15	224.23	2.973	0.006	0.002
Flush × treatment	6	128.84	1.708	0.163	0.138
Flush × cultivar × treatment	30	45.09	0.598	0.900	0.933
Error	51	75.41			
<i>Avg cutting wt</i>					
Cultivar	5	0.848	10.471	<0.001 ^x	
Treatment	2	1.351	16.691	<0.001 ^x	
Cultivar × treatment	10	0.129	1.592	0.192 ^x	
Error	17	0.081			
<i>Total cuttings (no.)</i>					
Cultivar	5	5089.12	10.023	<0.001 ^x	
Treatment	2	4395.33	8.656	0.003 ^x	
Cultivar × treatment	10	576.78	1.136	0.393 ^x	
Error	17	507.77			

²G-G is the Greenhouse-Geisser epsilon estimator that represents the maximum likelihood estimate of Box's ϵ factor, a multivariate test.

³H-F is the Hunyh-Feldt estimator that represents a less conservative estimate of Box's ϵ factor.

^xP value.

numbers of cuttings from the initial flush, such as 'Minngopher', when total productivity of the stock plant is considered (Table 3).

Cultivar responses to ethephon varied but not significantly. Cultivar

had a significant influence on cutting fresh weight and total numbers of cuttings obtained per stock plant, suggesting that chrysanthemum clones with low LDLN that are DN for FBI and FBD should be selected for cut-

ting production. Using ethephon on stock plants can be beneficial in that the reduction of crown-bud formation will yield a significant increase in the number of usable cuttings. We conclude that, in conjunction with the ethephon + GA₃ sprays, chrysanthemum clones DN for FBI and FBD with low LDLN could be selected to form a 4- to 5-week production group.

Table 2. Number of cuttings and average fresh weight (in grams) of cuttings taken from chrysanthemum stock plants treated with three concentrations (0, 250, 500 ppm) of ethephon.

Ethephon (ppm)	Cuttings (no.)					Fresh wt (g/cutting)	Budded (%)
	Flush I	Flush II	Flush III	Flush IV	Total		
0	23.1 a ^z	35.8 a	46.8 a	51.1 a	156.8 a	1.50 a	21.67 a
250	21.7 a	38.1 a	47.2 a	50.4 a	158.1 a	1.00 b	3.33 b
500	19.6 a	32.2 a	32.6 b	38.1 b	122.4 b	0.87 b	0.00 b

^zMean comparisons were performed using Fisher's protected least significant difference. Numbers within a column with different letters are significantly different at P=0.05.

Fig. 1. The production of marketable cuttings (by flush) from stock chrysanthemum plants treated with 0, 250, or 500 ppm ethephon. Bars within a flush with different letters are significantly different at P = 0.05. Mean separation was performed using Fisher's protected least significant difference.

Table 3. Number of cuttings and average fresh weight (in grams) per cutting taken from stock plants of one breeding selection and five cultivars of garden chrysanthemum.

Genotype	Cuttings (no.)					Fresh wt (g/cutting)	Budded (%)
	Flush 1	Flush 2	Flush 3	Flush 4	Total		
83-267-3	15.0 a ^z	31.7 ab	37.8 ac	44.2 ad	128.7 a	1.36 a	6.33 b
Burnt Copper	17.3 ab	30.8 ab	35.4 a	31.3 b	110.4 ab	0.93 b	6.33 b
Grape Glow	24.8 b	43.2 a	56.7 bc	65.2 c	189.8 d	0.89 b	0.00 a
Minngopher	33.5 c	41.7 a	36.0 a	44.2 ad	155.3 c	1.38 a	21.67 c
Minwhite	20.3 ab	40.0 a	48.5 bc	55.0 ac	163.8 cd	0.58 b	10.00 b
Ruby Mound	17.7 ab	24.8 b	36.8 ac	39.3 bd	118.7 ab	1.57 a	5.00 b

^zMean comparisons were performed using Fisher's protected least significant difference. Numbers within a column with different letters are significantly different at P=0.05.

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