Progress Report

Development of a new method for breaking buds on ornamental flowering plants.

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Report Date: February 2010 (2009-10 Midterm Report)
Funded by the Joseph H. Hill Memorial Foundation, Inc.
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Introduction

We have carried out a considerable amount of research on induction of bud breaking in roses, gerbera and a variety of other plants. Much of this research has focused on using plant growth regulators (PGRs) and cultural methods (e.g. photoperiodic control in gerbera) and was funded in part by the Hill Foundation as well as the national IR4 program. Unfortunately of these methods, the ones that were effective did not allow targeted initiation of one particular dormant bud. At the same time we also tried to find a targeted method that could be used to cause individual specific lateral buds to break. The currently-funded work is aimed at exploiting a discovery by us of such a targeted bud-breaking method.

This method is based on our observation that localized axillary bud break on the flower stem of *Rosa hybrida* 'Kardinal' can be induced to break through mechanical manipulation of the stem by partially compressing the internode above a specific axillary bud. We call this treatment a "Partial Crush" (PC) treatment. It induces a bud break at the proximal node, which will grow to produce a flower stem for subsequent harvest without harming the current stem or successive growth. The effect on cut-flower rose was to generate a specific and timed bud break from 7 to 14 days earlier than stem pruning or flower harvesting. Applying this treatment can potentially increase and better time production of cut flower roses. Furthermore, the method may be applicable in a variety of ornamental plants and it is part of this project to explore which other plants might respond to this treatment and how the treatment might need to be modified to maximize effectiveness.

The scientific basis for the process of bud breaking is that apical dominance inhibits axillary bud breaks and lateral shoot branching due to the inhibitory effects of auxin (IAA), which is biosynthesized in the shoot apex and polar transported in the plant

(Sachs & Thimann 1967; Leyser 2003). Axillary buds lower on a stem have a higher degree of inhibition then apical buds (Le Bris et al 1998). Apical dominance inhibits axillary bud breaks because of the polar transport of auxin through the stem from the growing apex (Kitazawa et al. 2008). Cytokinin encourages cell division and is translocated in the plant from the root upwards (Sachs and Thimann 1967). We speculated that disruption in the translocation of the growth hormones due to partial compression of the rose stem reduces auxin's inhibitory effect on the axillary bud below the compression site; accumulation of cytokinin below the wound probably encourages cell division and release of the bud. Research is needed to confirm this scientific basis or to discover what is actually going on so that we can tailor horticultural methods to it. At this time the most promising method is the Partial Crush method and the results of applying that method is described in this report.

Development of a commercial protocol to induce forced axillary bud breaks using the Partial Crush treatment.

Previous research helped develop a method that induces specific axillary bud breaks by partially crushing the stem above a selected axillary bud (Lieth, Orsi, unpublished). This treatment utilizes mechanical manipulation of the growing stem to force specific and timed axillary bud breaks on the growing flower stem to break before harvest where apical dominance is removed by pruning. The potential application of this treatment include forcing bud break in hard to break plant species, and a reduction in time between harvest periods due to early bud break. A practical protocol of application of the partial crush (PC) treatment needs to be developed in order to make the treatment available to growers to use on their commercial flower crops. The depth of application of the compression to the stem, the plant's carrying capacity of the treatment and effects of the treatment to subsequent generations of stems needs to be analyzed in order to develop an effective protocol for use. The purpose of this trial is to test the depth of compression needed in order to induce axillary bud break before stem harvest.

Hydrangea macrophylla

Previously we found that *Hydrangea macrophylla* is receptive to the PC treatment and buds broke on average 8 days after treatment. However, it was noted that 21 days after the application of PC some of the growing flower stems became necrotic at the crush site with the necrosis progressing up the stem to the terminal flower bud. This caused wilting in the leaves and flower cyme, followed by cell necrosis on the stem and eventual stem death. Since it was noted that the death occurred by day 21 and the PC treatment was applied approximately 6 weeks before harvest when the flower cymes were the size of a quarter, to assuage this issue in trial 2, the PC treatment was applied 3 weeks before harvest in an effort to force bud break before harvest, but before stem death can occur.

Screening of Ornamental and Cut Flower Greenhouse Plants Receptiveness to the PC Treatment.

Our objective is to identify and evaluate the partial crush (PC) treatment's effectiveness on other plant species that are commercially valuable. A variety of plants from different plant families are being tested to determine whether the PC treatment is family specific or generally works on all species and to what extent. We are particularly interested in determining for which ornamental plants and cut flower species the PC treatment represents an effective tool. Once species that are receptive to the treatment are identified a commercial protocol can be established for the utilization of the PC to force axillary bud breaks.

Materials and Methods

Partial Crush Protocol- Depth Compression for Forced Bud Break

Trial 1:

In a block with two rows with 15 *Rosa hybrida* 'Kardinal' plants, for a total of 30 plants, different applications of the partial crush treatment was tested so as to find the most efficient management practice. Plants were grown in 2-gallon containers of UC Mix (1 sand: 1 redwood sawdust: 1 peat) amended with Osmocote® slow release fertilizer and irrigated by drip irrigation (2 gal/hr emitters), with amended half-strength Hoagland's solution (ref.), 4 times a day for 2 minutes each cycle in order to maintain a leaching fraction of 30%.

Plants were pruned hard, with all flower stems or new growth removed to create a several flower stems to be timed for the experiment. Three weeks after pruning when the new flower stems were at the visible bud stage, (flower bud visible to the naked eye; about 10 to 14 days before the harvest date), four different treatments will be applied with the objective of finding the optimal compression depth for forcing axillary bud break. The treatments include:

1.) Control (CTRL) - The stem was flagged where the compression zone would have been, just above the targeted axillary bud. The bud is observed for any changes without any manipulation.
2.) Partial Crush 20% (PC20) - The stem was compressed to a depth of 20% of its caliper above the targeted bud.
3.) Partial Crush 40% (PC40) - The stem was compressed to a depth of 40% of its caliper above the targeted axillary bud.
4.) Partial Crush 60% (PC 60) - The stem was compressed to a depth of 60% of its caliper above the targeted axillary bud.

Each treatment was imposed using needle-nose pliers to crush the stem to a specific desired depth. Digital calipers were used to measure the stem width before the treatment was applied. The caliper of the stem was recorded, as was the depth of compression into the stem for each treatment. As the pliers were used to crush the stem the digital caliper recorded the width of the stem so the pliers did not crush at a greater or lesser depth then needed. The treatments were applied to the internode 0.5 to 1.0 cm above the most basipetal 5-leaflet leave on the flower stem.

Treatments were applied randomly within the planting block. 10 replicates of each treatment for a total of 40 stems treated were applied on December 4, 2009. Initial measurements taken before any treatment were applied include: stem caliper (mm), stem length (cm), number of nodes along stem, and any general observations about the stem. The targeted axillary bud's date of bud break, and any general observation of the stem growth were recorded. Stems were harvested individually when all 5 sepals are fully extended downward. Before the stem was removed its final stem length (cm) was recorded. All harvest dates occurred between 12/18/2009 to 12/28/2009.

All data was analyzed using analysis of variance (GLM, SAS) using t-test at the 0.05 significance level.

Ornamental and Cut Flower Crops Screening Trial

Several plant species were selected to be grown in the greenhouses to receive the PC treatment and to contrast the results with no treatment or other treatments that typically induce bud break. Plant species that were used in these trials consisted of ornamental plants: *Hydrangea macrophylla* 'Angel's Robe', *Rhododendron* sp. 'Florist Azalea', and Bachelor's buttons; and cut flower varieties currently being tested include: *Antirrhuinum majus* (Snapdragon), *Helianthus annuus* (Sunflower). Other plant species are presently being grown or are in current trials in the greenhouse include: *Anemone sp., Delphinium sp., Eustoma sp.* (Lisianthus), *Callistephus chinensis* (Aster), *Dianthus sp.* (Dianthus), *Limmonium sp., Matthiola incana* (Stock), *Tanacetum parthenium* (Matricaria), *Trachelium caeruleum, Verbascum x hybrida,* and *Veronica hybrida* (Veronica).

Rhododendron sp. 'Florist Azalea', Bachelor's Button, Cut Flower Screening Trials

For each of these plant species only stems with an apical floral meristem were used. Stems chosen had flowers that were at the visible bud stage. The following were the measurements taken before any treatments were applied:

- Date of treatment, Plant ID, Replicate number
- Stem length (cm) - measured from the treated axillary bud to the stem apex.
- Node position – treated node position recorded measured from the bottom of the stem to the top
- Nodes per stem

After measurements were taken, the stem were tagged above the targeted node with a tag containing the plant ID, and date; then the treatment was applied. The PC treatment was applied to the stem 0.5cm above the node that is targeted for bud break. The stem was crushed with needle nose pliers to a depth of 40% to 60% of the stem caliper. Control treated stems were flagged above the targeted node intended for bud break. After the stem was treated the date of axillary bud break was recorded, along with the following bud (or stem) length measurements at 5,7,14, 21 and 28 days of any resulting new stem. Any observations about the stem growth were noted.

Hydrangea macrophylla - trial 2

Hydrangea plants were grown in 1 gallon containers in UC Mix (1 sand: 1 redwood sawdust: 1 peat) amended with Osmocote® slow release fertilizer. Two treatments were tested: partial crush (PC) treatment and control (CTRL). The treatments were applied to growing flower stems above the targeted axillary bud when the flower cyme had approximately 20% of its bract color showing. This was estimated to be 3 weeks before the harvest date which was considered to be when the first flower opens. The PC and CTRL treatment were applied and observed the same as the ornamental and cut flower screening trials previously stated.

Results/Discussion

Partial Crush Protocol- Depth Compression for Forced Bud Break

Preliminary data are shown in Table 1. Data are currently still being collected for this trial. After the stems were treated the targeted buds were observed for bud break before harvest. No stems reached bud break before harvest although several of the PC treated stems had swollen buds before harvest. After the treated stem was harvested, the targeted bud are still being observed for bud break date and the bud's subsequent harvest date once it reaches maturity.

Table 1. Partial Crush (PC) protocol trial 1.

Treatment	Mean Actual % Crush	Mean Change in Stem Length (cm)	Days to bud break after treatment	n of stem to reach budbreak to date
PC60	58%	9.1 a	24.7 a	7
PC40	41%	10.1 a	27.5 a	6
PC20	22%	11.8 a	31.5 a	8
CTRL	0.0%	10.3 a	29.4 a	5

*Mean separations determined by standard t-test (0.05)

For the first time since the PC treatment trials have started there were no bud breaks prior to harvest on *Rosa hybrida* 'Kardinal'. This is found to be out of the norm for this variety of cut flower rose because in several previous trials, the growing flower stems and bottom canes have all shown to significantly be effected by forced bud break prior to harvest.

There are some differences in this trial which leads us to conclude that they are affecting the forced bud breaks. First, the time of year for application of the PC treatment has never been done to growing flower stems in the month of December. This suggests that carbohydrates might be a major player in forced bud break due to reduced photosynthesis in the winter months.

Secondly, when the treatment was applied the crush zone was on the stem 0.5-1.0 cm above the targeted node, but the crush site was only about 3mm tall, the width of the needle nose pliers and was applied in one clean motion or compression. The PC treatment was applied in this way because of the need to measure the depth of crush with the digital calipers at the time of application. Previous studies with the PC treatment were applied by the touch at an estimate of 40-60% of the stem caliper at 0.5-1.0cm above the targeted node, but the zone of the stem that was crushed was approximately 1.0cm in height on the stem and was crushed in a slow motion, first with the stem tissue being softened and then the crush was applied until the pliers could not be squeezed further. In the next trial the zone of crush will be a greater width on the stem and will not be just the width of needle nose pliers.

Ornamental and Cut Flower Crops Screening Trial

Out of the 4 species that have currently been analyzed, there was no significant difference between the PC and CTRL stem and the effect of forced bud break before harvest. All data indicates that the PC treatment did not affect the mean stem length (Table 2), or increase the number of bud breaks prior to harvest. Data are still being collected for the other eleven cut flower species that are being screened for their receptiveness to the PC treatment.

Table 2. Ornamental and Cut flower species screened for receptiveness to PC treatment

Species	Treatment	n	number of plants to break before harvest	Mean Stem Length (cm)		Mean Days to Bud Break	
A. majus (Snapdragon)	PC	10	4	108.7	a	30.0	a
	CTRL	10	1	112.6	a	36.0	a
H. annuus (Sunflower)	PC	10	0	89.3	a	------	
	CTRL	10	0	105.4	a	------	
Rhododendron sp. (Azalea)	PC	9	1	9.8	a	14.0	a
	CTRL	9	1	10.0	a	7.0	a
Bachelor's Buttons	PC	9	6	59.3	a	5.0	a
	CTRL	9	2	60.4	a	5.0	a

*Mean separations determined by standard t-test (0.05)

More screening and research is needed to discover valuable cut flower or ornamental species that are receptive to the PC treatment. This will also help us determine if there is a way to determine if a plant might be receptive to this treatment either because that plant family is sensitive to the PC treatment or if it is due to the physiology of the plants (i.e. C3, C4 photosynthesis).

Hydrangea macrophylla - trial 2

For this trial the PC treatment was applied at a later stage in the flower growth to induce forced bud break but the stem was removed for harvest before death could occur. Of the 7 stems that were treated with the PC treatment all of them had bud break before harvest (Table 3). Of the 8 Control treatment stems only 2 had bud break prior to harvest. The days to bud break show the time from treatment to bud break for those buds that broke prior to harvest.

Table 3. Hydrangea trial 2. Efficacy of later application of PC treatment on reducing stem death prior to harvest

Species	Treatment	n	number of plants to break before harvest	Mean Stem Length (cm)		Days to Bud Break	
Hydrangea macrophylla 'Angel's Robe'	PC	7	7	29.5	a	24.6	a
	CTRL	8	2	26.2	a	29.5	a

*Mean separations determined by standard t-test (0.05)

Applying the treatment at the first color stage instead of when the buds were the size of a quarter prevented any stem death prior to harvest in stage two. A new trial with a larger sample size will be conducted in the spring time to provide better statistical data on the late application of the PC treatment.

Conclusion

While it is not surprising that some species are not likely to benefit from this method, others, including *Hydrangea* and Rose, show great potential. It would be beneficial to continue the research in developing a PC method protocol for commercial application and screening of other high value cut flower species. Hydrangea needs further testing to better refine a commercial application protocol, as well as Rose. Further testing on cut flower Hydrangea varieties, could either prolong the growing season or harvestable yields per year. The next step in the project is to identify other cut flower species that are receptive to the PC forcing method, further refine protocols for commercial application of the PC treatment on Roses and *Hydrangea*, and determine whether it is plant growth hormones that are causing the forced bud breaks due to the PC treatment or if the forced bud break is due to carbon partitioning and photosynthesis.

Literature Citied

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