

# ROOTING OF FRASER FIR CUTTINGS: EFFECTS OF POST-SEVERANCE CHILLING AND OF PHOTOPERIOD DURING ROOTING

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**Abstract.** Terminal stem cuttings from 5-year-old Fraser fir [*Abies fraseri* (Pursh) Poir.] stock plants were collected in early fall, when in a state of rest or winter dormancy. Cuttings were subjected to dark storage at 4°C for 0, 2, 4, 6, 8, 10 or 12 weeks. Following storage, and prior to insertion into a rooting medium, cuttings were subjected to one of two treatments: non-treated, and wounding + indolebutyric acid (IBA). Cuttings received short- or long-days during a 10-week rooting period. Non-chilled cuttings did not root or break bud. High percent rooting occurred after 4 to 6 weeks of chilling, whereas visible terminal bud activity peaked after a 10-week chill. Rooting was primarily contingent upon IBA treatment and chilling, although long days had a strong promotive effect when cuttings were chilled less than 6 weeks.

Tree species indigenous to temperate zones typically become dormant in early fall and resume growth only after a period of low temperatures. For some conifer species, plants or stem cuttings collected in September and October exhibit low capacity for cambial activity, root initiation, or budbreak (1,2,8,9,10). As the chilling requirement is satisfied, rooting capacity and cambial activity increase to a peak between December and April. Long days promote cambial activity, budbreak, and root initiation in dormant Douglas fir (*Pseudotsuga menziesii* (Mirb.) Franco) and balsam fir (*Abies balsamea* L.) plants and stem cuttings collected in early fall, in effect, substituting in part for the chilling requirement (8,10). The compensating effect of long days tends to diminish as the chilling requirement is satisfied.

Preliminary work with Fraser fir (*Abies fraseri* (Pursh) Poir.) stem cuttings collected in early fall and rooted under 10- to 12-hour days indicated rooting percentages were relatively high following 4 or 8 weeks of constant chilling at 4°C, whereas visible terminal bud activity was increasing but still relatively low even after 8 weeks of chilling (5). This suggested different chilling requirements for rooting and budbreak.

The objective of the current study was to examine rooting

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capacity and visible terminal bud activity of Fraser fir stem cuttings as influenced by duration of post-severance chilling and photoperiod during rooting.

### MATERIALS AND METHODS

Terminal Fraser fir stem cuttings, 16 to 25 cm in length, were collected on October 8, 1980 from 5-year-old (3+2) transplants growing under uniform fertility levels at the Joseph A. Gill Nursery (North Carolina Forest Service) in Crossnore (36° 01'N latitude, 81° 56'W longitude; elevation = 980 m). The provenance was Roan Mountain (36° 09'N latitude, 82° 05'W longitude, elevation 1900 m). Cuttings consisted of multiple shoots that resulted from previous frost damage to terminal buds. Orthotropic shoots were used to avoid the plagiotropic growth habit of cuttings taken from lateral branches (5). Only one cutting was taken per stock plant to ensure a broad genetic base in the experiment. Stock plants had experienced 3 frosts and a cumulative total of 60 hours of air temperatures below 5°C during the 3 weeks preceding collection of cuttings. They were assumed to be in a state of rest (3) or winter dormancy (4). The cumulative effect of the low temperatures in satisfying the chilling requirement for budbreak was assumed to be negligible since maximum day temperatures during that period were 18 to 27°C.

As cuttings were collected, they were sealed in polyethylene bags, placed in a cooler with ice, and transported to Raleigh. Prior to cold storage, cuttings were trimmed acropetally to 15.3 cm. A group of cuttings was randomly selected to receive no additional chilling; the others were sealed in polyethylene bags and dark stored at 4°C. Cuttings in each bag were oriented in a vertical position with terminal buds upward and the bases resting on moist paper towels.

Following cold storage for 0, 2, 4, 6, 8, 10 or 12 weeks, cuttings were trimmed to 15 cm and needles removed from the basal 4 cm. Half the cuttings received no treatment; others received wounding + IBA. Wounding consisted of 4 equidistant vertical cuts into the xylem on the basal stem, each cut about 2.5 cm in length and parallel to the longitudinal axis of the cutting. IBA was applied by dipping the basal 2 cm of each cutting into a 5000 ppm IBA solution followed by 15 minutes of drying before insertion into the rooting medium. The solution was prepared by dissolving reagent grade IBA in 50% isopropyl alcohol. Cuttings were inserted to a 4-cm depth in a raised greenhouse bench containing a moist medium of Canadian peat and sand (1:1, v/v). Intermittent mist operated 5 seconds every 5 minutes between 0700 and 1830 hours daily. Day/night maximum/minimum ambient air temperatures



were  $24 \pm 5 / 14 \pm 4^{\circ}\text{C}$ . Day/night maximum/minimum rooting medium temperatures at a 2-cm depth were  $21 \pm 3 / 16 \pm 3^{\circ}\text{C}$ . On warm sunny days, 50% shade cloth was used to maintain the ambient air temperature below  $29^{\circ}\text{C}$ . In addition to rooting and chilling treatments, 2 photoperiod treatments were utilized in the mist bed: short- and long-days.

The experimental design was a split-plot with 4 replications. Photoperiods served as main plots, and chilling x rooting treatments were randomized within each main plot. A single raised greenhouse bench was used. A replication consisted of 2 main plots, each with 14 contiguous sub-plots. Each sub-plot contained 8 cuttings. Short- and long-day treatments were randomly assigned to main plots in each replication. Short-days were imposed by placing lightproof, plywood boxes over the appropriate main plots between 1700 and 0800 hours daily. Temperature and relative humidity inside and outside the boxes were similar. Long days were effected by a 3-hour night light break with incandescent light between 2300 and 0200 hours daily. The light was supplied by 100 W incandescent lamps located 70 cm above the surface of the rooting medium and spaced 100 cm apart. This provided a radiant power density<sup>1</sup> of 1.9 and  $2.1\text{W}/\text{m}^2$  at the center and outer edge of the bed respectively, measured at the terminal buds with a Li-Cor LI-185A quantum/radiometer/photometer. Black cloth covered the bench between 1700 and 0800 hours daily.

The experiment was initiated on October 9, 1980 and terminated April 1, 1981. Each treatment was represented by 32 cuttings which remained in the mist bed for 10 weeks. Data included the number and length of roots  $> 1$  mm in length. Cuttings having one or more roots were classified as rooted. For each plot, the average number and length of roots per rooted cutting were computed, giving equal weight to each cutting. An active terminal bud was defined as one which was visibly swollen or had burst. Percentages were transformed with the angular transformation and data subjected to standard analysis of variance procedures.

## RESULTS

Cuttings collected in early October, which received no artificial chilling, failed to root (Figure 1A). Rooting response to chilling and photoperiod was strongly contingent upon wounding + IBA (Table 1). Percent rooting for IBA-treated cuttings increased early in the chilling cycle and reached a virtual maximum under long days following 4 weeks of chilling; 6 weeks under short days. Non-IBA-treated cuttings first rooted

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<sup>1</sup> Photomorphogenic radiation between 750 and 830 nm.

after 4 to 6 weeks of chilling and reached the maximum after 10 weeks. Following 2 or 4 weeks of chilling, percent rooting of IBA-treated cuttings was about 2× greater under long days, but the difference between photoperiods was negligible for chilling durations  $\geq$  6 weeks. Percent rooting of non-IBA-treated cuttings was not affected by photoperiod at any point.

**Table 1.** Test of significance for the effect of post-severance chilling and photoperiod on rooting and visible terminal bud activity of stem cuttings collected from 5-year-old Fraser fir stock plants in early October.

Duration of chilling (weeks)	Parameter												
	Active terminal buds (percent)			Rooting (percent)			Root length			Roots/rooted cutting			
	T <sup>z</sup>	P	T×P	T	P	T×P	T	P	T×P	T	P	T×P	
0	(-)y	-	-	-	-	-	-	-	-	-	-	-	-
2	-	-	-	*	NS	NS	*	NS	NS	*	NS	NS	NS
4	NS	NS	NS	*	*	*	*	NS	NS	*	NS	NS	NS
6	NS	NS	NS	*	NS	NS	*	NS	NS	*	NS	NS	NS
8	*	NS	NS	*	NS	NS	*	NS	NS	*	NS	NS	NS
10	*	NS	NS	*	NS	NS	*	NS	NS	*	NS	NS	NS
12	*	*	NS	*	NS	NS	*	NS	NS	*	NS	NS	NS

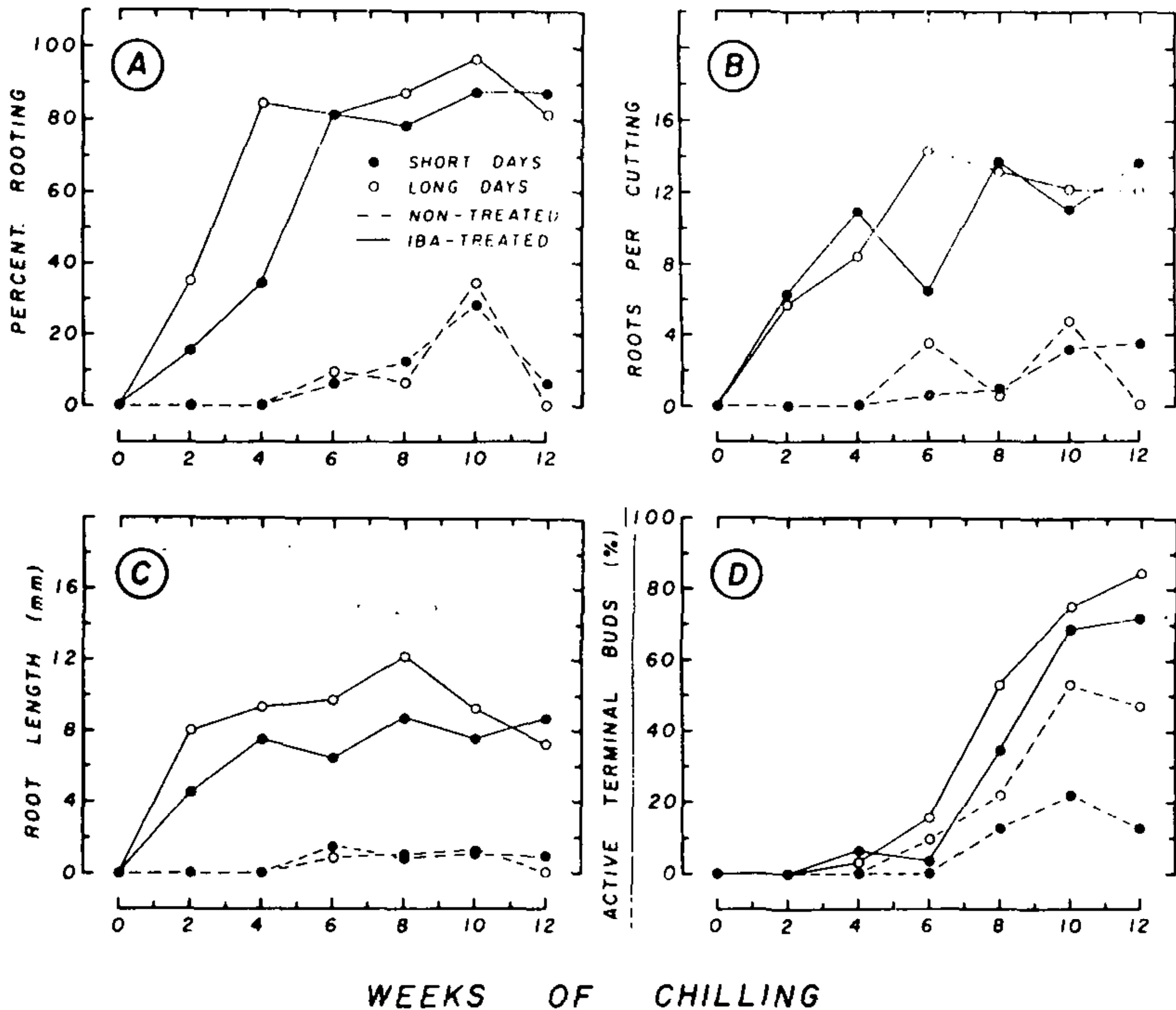
T<sup>z</sup> = IBA treatment, P = photoperiod, T×P = IBA treatment × photoperiod interaction. (-)y, values for all treatments were zero; (\*), significant at 5% level; (NS), not significant.

The number of roots per rooted cutting (Figure 1B) was greatly enhanced by wounding + IBA, and maximum root lengths (Figure 1C) were realized after 4 to 8 weeks of chilling. Root formation in non-IBA-treated cuttings was too erratic to identify the chilling requirement for maximum rooting. Also, there was no significant relationship between photoperiod during rooting and the number and length of roots per rooted cutting (Table 1).

Non-IBA-treated cuttings produced few roots (Figure 1B) at any duration of chilling, and where present, the roots were very short (Figure 1C). IBA-treated cuttings, which produced numerous roots even following short durations of chilling, had much longer roots. Long days increased root length of IBA-treated cuttings chilled 10 weeks or less (Figure 1C). Root length under long days was virtually maximum on IBA-treated cuttings after only 2 weeks chilling; 4 weeks under short days.

Time course of visible terminal bud activity (Figure 1D) contrasted with that of percent rooting (Figure 1A). Visible bud activity was negligible through 4 weeks of chilling under long days, 6 weeks under short days. All cuttings, regardless of treatment, exhibited a sharp increase in visible bud activity for the interval between 6 and 10 weeks of chilling, and addi-

tional chilling beyond 10 weeks was of little benefit. IBA-treated cuttings subjected to long days and chilled 6 weeks or longer consistently displayed more visible terminal bud activity than those receiving short days, the absolute difference being 10 to 20%. The capacity of long days to promote bud activity was most apparent, however, for non-IBA-treated cuttings chilled 10 to 12 weeks, the absolute difference being 30 to 35% (Figure 1D, Table 1).



**Figure 1.** Rooting and visible terminal bud activity of Fraser fir stem cuttings collected from 5-year-old stock plants on October 9, 1980, artificially chilled for 0 to 12 weeks at 4°C, and rooted 10 weeks in a mist bed. (A) percent rooting, (B) average number of roots per rooted cutting, (C) mean root length per rooted cutting, (D) percent visibly active terminal buds. An active bud was defined as one which had broken bud or was visibly swollen. Legend in (A) applies to all figures.

### DISCUSSION

Chilling was clearly a prerequisite for rooting of early fall-collected cuttings (Table 1A), and neither IBA nor extended photoperiods, either singly or in combination, induced rooting in non-chilled cuttings. Many non-chilled cuttings (time 0) died or partly defoliated in the mist bed, whereas those chilled 2 weeks or longer did not. Stock plants apparently must



achieve a certain tissue maturity, or attain a certain phase of dormancy before cuttings can be successfully transferred to a mist bed for rooting. Since cuttings chilled 2 weeks or longer did not die or defoliate in the mist bed, the necessary physiological changes evidently occurred during the first 2 weeks inside the 4°C, dark cold room, and were not dependent on the presence of light or a root system. These changes had probably proceeded to an unknown extent in response to shorter days and decreasing temperatures during the month prior to collection of cuttings, and continued in the cold room.

The principal effect of long days was to increase percent rooting for IBA-treated cuttings early in the dormancy cycle; those chilled 2 to 4 weeks had 2- to 2.5-fold higher rooting percentages under long days. Thereafter, the effect of photoperiod on percent rooting was not significant (Table 1). This poses a question why long days compensated for chilling in IBA-treated cuttings, but not in non-treated cuttings. Long days might have caused the formation of substances in the buds or foliage, which subsequently interacted with auxin to promote rooting. In this regard, Lavender and Hermann (6) demonstrated that resumption of root, bud, and cambial activity in quiescent Douglas-fir seedlings was dependent upon the presence of foliage, and foliage exposed to long days appeared to export substances (not identified) which stimulated meristematic activity.

Chilling alone does not appear to have the capacity to induce 100% terminal budbreak in Fraser fir cuttings. Maximum rooting and visible terminal bud activity for non-IBA-treated cuttings was 35 and 53%, respectively, following a 10-week chill. Much more visible terminal bud activity would have been expected late in the dormancy cycle, particularly if resumption of bud growth was due to the reduction or elimination of inhibitors within the bud as a consequence of low temperatures. Based on Douglas-fir studies, Roberts and Fuchigami (9) suggest that budbreak is tied to the internal balance of inhibitors and promoters, which is affected not solely by chilling, but other factors as well. For example, given equivalent periods of chilling, e.g., 8 to 10 weeks, IBA-treated cuttings had much higher bud activity, even under short days, than non-treated cuttings subjected to long or short days (Figure 1D). Treated cuttings also rooted in higher percentages (Figure 1A), and had greater root numbers and lengths (Figures 1C, 1D). Since auxins move primarily in a basipetal direction, the addition of auxin to the distal end of a cutting would not in itself be expected to directly affect budbreak. Obviously, however, exogenous application of IBA dramatically increased terminal bud activity as well as rooting response (Figure 1A, 1D). Per-

haps the effect of auxin on bud activity was an indirect effect exercised through root formation, but this was not investigated. Once roots are present and actively growing, acropetal transport of hormones, e.g., gibberellins (7) and other substances, could promote bud activity in terminal buds predisposed to break bud following a sufficient period of chilling. Also supporting this hypothesis is the observation (unpublished data) that visible bud activity during a 10-week rooting period decreases with increasing cutting length. This suggests that materials moving acropetally from the roots might require a longer time to reach the terminal bud in quantities sufficient to cause a noticeable change in visible bud activity.

Results (Figure 1) confirm observations that the chilling requirement for rooting is less than that for budbreak (5,9). The rooting response of IBA-treated cuttings was essentially maximum following 6 weeks of chilling, whereas visible terminal bud activity increased sharply thru 10 weeks of chilling. For non-treated cuttings, both maxima occurred following 10-weeks of chilling. Currently, it cannot be explained why the response differed for treated and non-treated cuttings. Since visible terminal bud activity was greater for all cuttings following 10 weeks of chilling, it appears the capacity to break bud hinges upon physiological changes brought about by the cumulative effects of chilling over a long period of time. In contrast, rooting occurs in high percentages following brief chilling if sufficient levels of auxin are applied. Generally, auxin and other substance(s) which promote cambial activity and rooting originate in increasing amounts from buds and foliage as the chilling requirement for budbreak is satisfied (6,8,10). In the current studies, endogenous levels of these substances in non-IBA-treated cuttings were apparently too low at all points in the chilling cycle to promote a strong rooting response. Even though applied auxin overcame the restriction on rooting, it is suspected that chilling dependent factor(s), e.g., inhibitors, prevented budbreak early in the dormancy cycle. Following a sufficient period of chilling, and reduction or elimination of the inhibitors, budbreak would be enhanced by hormones or other substances arising from the cambium, leaves, or roots. The results reported herein are consistent with the hypothesis that resumption of growth is controlled by a balance of promoters/inhibitors (9) which is affected by numerous factors, e.g., duration of chilling, and the presence or absence of a developing root system.

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JOERG LEISS: Did you compare lateral with apical cuttings?

FRED MILLER: Yes. Lateral cuttings were a problem because they continued to grow as lateral branches.

## **UTILIZING CAPILLARY IRRIGATION FROM THE PROPAGATION BENCH TO HARVEST**

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The concept of capillary watering is an age old practice but its application to irrigation of container-grown nursery stock on ground beds outside is a new use in the U.S. This method of watering nursery stock had its beginning at the Efford Experiment Station in England and is now in widespread use in Europe (12) and New Zealand (7). Capillary watering has several advantages to overhead watering including reduced water consumption, water run-off, weeds, root (5) and foliar diseases. This procedure has been evaluated at The Ohio State University for the past several years and the following report summarizes several of these studies (10,11).