SECTION 8
PROPAGATION

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Split Wounding of Stem Cuttings

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Nature of Work: Wounding of stem cuttings has been thought to increase rooting efficacy in ornamental plant taxa for least the last 50 years (4). When used in combination with rooting hormone formulations, rooting percentages may again be amplified (1,2,4).

The general purpose of wounding is to stimulate the initiation of roots from cortex and phloem cells by exposing a greater surface area of these cells than is provided by the cut base in an unwounded cutting (4). Furthermore, cells exposed by wounding may take up the hormone solution more readily, as well as some water and other growth factors from the rooting medium (4). Wounding treatments vary widely; from removal of basal branchlets or leaves, to smashing the bases of stems, and the various slice or incision wounding systems (2,3,4). Split wounding may be an improvement over other wounding techniques in some taxa. With this technique, more cells that differentiate new root tissues may potentially be exposed, especially in the cortex. MacDonald (4) reported that research done at East Malling Research Station demonstrated that split wounding gave the best results in rooting of dormant hardwood cuttings of rootstocks of Malus. Later the technique was shown to improve rooting percentages in Acer, Castanea, and other cultivars of Malus (4).

Though split wounding is a relatively common practice in England (2), there has been very little published about this technique in American literature. Because of the potential benefits this technique could offer for improving the rooting of difficult-to-root species, the following study was initiated to quantify the effectiveness of split wounding relative to other wounding techniques commonly used in the United States. Hardwood cuttings of five taxa of evergreen woody plants were studied for their responses to split wounding, double slice wounding (scraping), incision wounding, and the unwounded condition.

Rooting hormone concentrations for experimental taxa were formulated as recommended by Dirr & Heuser (1). All cuttings were treated with varying concentrations of indole-3-butyric acid (IBA), amine form, in isopropyl solvent. The proper concentration for each taxon was reached by diluting the IBA/alcohol solution with distilled water. Cuttings were stuck individually in rooting trays containing 24 cells with dimensions of 2 3/4 x 3 1/8 x 4 3/8 inch which tapered to a 3/8 inch orifice suspended 1 inch above the bottom of a frame. The propagation substrate used was 80:20 (by volume) horticultural grade perlite and screened sphagnum peat. The experiment was arranged in random block design, with each treatment being replicated

three times with six cuttings of each taxon per replication, and three trays per taxon.

Terminal cuttings with two or three nodes were taken. Wounding treatments of cuttings were made on the basal one inch (2.5 cm), with the four wounding treatments consisting of: (1) No wound, where only basal leaves (if present) were removed; (2) Double slice wound, where two opposing longitudinal scrapes were made on each side of the stem, removing a strip of bark and exposing the cambium; (3) Double incision wound, where two opposing longitudinal cuts were made along the vertical axis of the stem, with the blade resting on the wood, thus severing the cambium; and (4) Split wound, where stems were split longitudinally in the middle. All wounding was accomplished using standard razor blades with cuttings supported on a table top. All cuttings were dipped in the hormone solution for seven seconds and allowed to "air-dry" before insertion into the rooting substrate. Cuttings were placed under intermittent mist operating at six seconds every six minutes during daylight hours and with bottom heat maintained at 72 degrees F (22 degrees C) by Biotherm hot water tubes.

The five taxa selected for study, hormone concentrations applied and dates inserted and evaluated were: (1) *Illicium parviflorum* (Small anise-tree), 3000 ppm IBA, inserted: February 3,1991, evaluated April 20, 1991; (2) *IlexX* 'Nellie R. Stevens' holly, 8000 ppm IBA, inserted: January 27, evaluated, April 27; (3) *Juniperus horizontalis* 'Blue Horizon'(Blue Horizon juniper), 5000 ppm IBA, inserted: February 2, 1991, evaluated April 7, 1991; (4) *Photinia X fraser,* (Red-tip photinia), 10,000 ppm IBA, inserted: January 27, 1991, evaluated March 17, 1991; (5) *Prunus caroliniana* (Carolina cherrylaurel), 6000 ppm IBA, inserted, February 2, 1991, evaluated April 5, 1991.

The parameters evaluated for all taxa were rooting percentage, root weight and root length. Data were evaluated using General Linear Models Procedure and mean separation between treatments was tested by the Waller-Duncan Mean Separation Test at $p = 0.05$.

Results and Discussion: No statistically significant differences were elucidated from any of the treatments (Data not Shown). There were no differences observed in rooting percentage, root weight or root length. Anise-tree and 'Nellie R. Stevens' holly cuttings rooting 100% in all treatments; 'Blue Horizon' juniper cuttings rooted 100% in all treatments except for double slice wounding (94%); while Carolina Cherrylaurel rooted 100% with incision wounding or without wounding at all, and 94% rooting with double slicing or split wounding. Red Tip photinia rooted 94% when split wounded or double slice wounded, and 89% with incision wounding or the unwounded condition. Because none of the above rooting percentages were significantly different, it would appear that with proper hormone

application, there is no decided benefit in wounding these taxa. It is important to note, however, that the act of stripping the lower branchlets from the bases of juniper cuttings probably provided the essential wounding for the "unwounded" cuttings in this taxon.

Although rooting parameters measured produced no differences some observations of the cuttings not discernable by statistical analysis were made. For example, 'Blue Horizon' juniper cuttings that were drastically wounded by double slice or split wounding procedures appeared to create excessive destruction of tissues in the wounded region. The juniper cuttings often rooted only above the wounded region, which usually rotted. This effect was also noted in cherrylaurel and anise-tree whose thin stems commonly rotted along the entire basal portion when split wounded. In general, it appears that species having wide stems, like photinia and holly, can be expected to tolerate split wounding more readily.

Significance to Industry: The results of this study indicated that hardwood cuttings of *Illicium parviflorum,* (Small anise-tree), *Ilex X 'Nellie* R. Stevens' holly, *Juniperus horizontalis* 'Blue Horizon', *Photinia Xfraseri* (Red-tip photinia), and *Prunus caroliniana* (Carolina cherrylaurel) root well with hormone formulations and concentrations suggested by propagation literature irrespective of wounding treatment. Observations of cuttings demonstrated that wounding procedures which created the greatest destruction of basal stem tissue often resulted in rooting above the wounded portion of the stem and rotting occurred in the wounded zone.

Literature Cited:

1. Dirr, Michael A. and Charles W. Heuser, Jr. 1987. The Reference Manual of Woody Plant Propagation. Varsity Press, Inc. Athens, GA. 239 p.

2. Hartmann, Hudson J. and Dale E. Kester. 1975. Plant Propagation: Principles and Practices. 4th Ed. Prentice-Hall, Inc., Englewood Cliffs, N.J. 727 p.

3. Humphrey, Brian. Production Director, NotCutts Nursery. England. Personal communlcation.

4. Macdonald, Bruce. 1986. Practical Woody Plant Propagation for Nursery Growers. Timber Press, Portland, OR. 669 p.

Position of Cutting on Rooting and Subsequent Growth of 'GulfRay', an Azalea With a Prostrate Growth Habit

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Nature of Work: 'GulfRay' is an evergreen azalea that grows vigorously with profuse reddish pink flowers similarly to the vigor and flowering of azalea cultivars such as 'Elegans Superba' ('Pride of Mobile') and 'Formosa', which are members of the Southern Indian Hybrid, azalea group. The distinctive feature of 'GulfRay' is its cascading or weeping prostrate growth habit. The origin of 'GulfRay' azalea is not known. It was found in the yard of a residence near Agricola, MS (30 miles north of Pascagoula) by Ray Dean of Sun Ray Nursery, Lucedale, MS. The azalea was planted as part of the foundation planting more than 40 years ago. Dean gave this plant to the Mississippi Agricultural and Forestry Experiment Station for evaluation and release.

'GulfRay' was planted at the South Mississippi Branch Station office as part of the landscape foundation planting in the spring of 1986. The prostrate growth habit was exhibited by the plants. However, in 1989 a few branches in the center of many of the plants exhibited upright growth. The objective of this study was to evaluate the effects of position of cutting on the stock plant and IBA concentration on the rooting of cuttings of Rhododendron 'GulfRay'.

Cuttings from stems growing prostrate (horizontally) and from those that appeared to be growing upright (vertically) were taken on July 11, 1989. Cuttings were inserted without a quick dip treatment and with a 5 second quick dip treatment in a solution of 50% water and 50% ethanol only and with 2,500, 5,000 and 10,000 ppm of IBA in 50% water and 50% ethanol solution, Table 1. This resulted in a two, cutting position x five, quick dip method factorial arrangement of treatments. Treatments were arranged in a randomized complete block design and replicated 6 times with 5 cuttings as an experimental unit. Roots were evaluated using a scale of 6=excellent, 5=good, 4=fair, 3=poor, 2=alive and no roots and l=dead. The cuttings were inserted into a medium of 100% vermiculite and rooted in a greenhouse. The propagation medium was hand irrigated as required and humidity was controlled with a Humidifan, Model 110, manufactured by Jaybird Manufacturing, Center Hall, PA 16828.

Rooting was acceptable regardless of treatment. Best rooting was obtained with cuttings taken from horizontal branches and rooting was not influenced by quick dip method, Table 1. Excellent rooting of cuttings taken from vertical or upright stems occurred only with plants not receiving a quick dip treatment. Also, with vertical cuttings good rooting was obtained with

ethanol and water only and slightly poorer rooting was obtained with ethanol and water plus IBA, quick dip treatment.

The objective of a second study was to evaluate the effects of position of cutting taken from the stock plant and fertilizer rate on the subsequent growth of Rhododendron 'GulfRay'.

Well rooted cuttings from the first experiment were planted in 3 inch diameter pots and grown to liner size in a 55°F minimum green house. The plants were maintained in two groups. One group of plants was from cuttings taken from horizontal branches and one group of plants was from cuttings taken from young vertical branches. Plants from both groups were planted in 3 gallon containers on April 26, 1990. Fertilizer treatment rates were 6, 11 and 16 lbs of 17-7-12 per yd^3. A randomized complete block design was utilized with treatments in a factorial arrangement of position of cuttings and fertilizer rate. Treatments were replicated 6 times with one container plant as an experimental unit.

The plants were grown in a 33% shade house. All plants were transplanted into 7 gallon containers on April 25, 1991 and fertilized at the same rates previously applied. The plants were placed on pedestals (cinder blocks) in the spring of the second season to accommodate the weeping, prostrate growth habit of this azalea.

Plant height after the 1990 and 1991 growing seasons was not affected by position cuttings were taken from the stock plants or fertilizer rate, Table 2. Branches or stems from the center of the plant that initially grew upright become prostrate with time. Plant width increased as fertilizer rate increased after the 1991 growing season, but not after the 1990 growing season. Flowering occurred from mid March to early April, Table 2. Flowering was slightly better at the lowest fertilizer rate.

Table 1. Exp. I. Effects of position of cutting and quick dip method on the rooting of Rhododendron 'GulfRay', 1989.

Cutting position[1]	Quick dip method	Root evaluation[2]
horizontal	none	5.57ab[3]
horizontal	50% ethanol	5.80a
horizontal	50% ethanol + 2500 ppm IBA	5.53ab
horizontal	50% ethanol + 5000 ppm IBA	5.80a
horizontal	50% ethanol + 10000 ppm IBA	5.93a
vertical	none	5.67ab
vertical	50% ethanol	4.87bc

vertical	50% ethanol + 2500 ppm IBA	3.97d
vertical	50% ethanol + 5000 ppm IBA	4.23cd
vertical	50% ethanol + 10000 ppm IBA	4.07cd
LSD		0.814
Significance		
Cutting position (CP)		**
	Quick dip (QD)	*
	CP x QD	**

[1] Horizontal cuttings taken from stems growing horizontally; vertical cuttings taken from stems growing vertically.

[2] Root evaluation - 6=excellent, 5=good, 4=fair, 3=poor, 2=alive and no roots, and l=dead.

[3] Mean separation by Fisher's protected LSD, 0.05 level; means followed by the same letter are not significantly different.

Table 2. Exp. II. Effects of position of cutting and fertilizer rate on the growth and flowerinq of 'GulfRay'.

Cutting position	17-7-12 rate (lb/yd^3)	Height 11/90 (in)	Width 11/90 (in)	Flower rating1 03/18/91	Flower rating1 04/02/91	Height 10/91 (in)	Width 10/91 (in)
horizontal	6	9.4	28.3	0.2b[2]	7.7a	14.2	43.7c
horizontal	11	9.8	28.3	1.3a	4.8c	13.4	46.lbc
horizontal	16	8.3	30.3	0.9ab	5.8bc	12.6	52.4a
vertical	6	9.8	28.0	0.3b	6.5ab	14.6	44.1c
vertical	11	7.7	29.9	0.9ab	5.3bc	14.6	49.2ab
vertical	16	8.9	27.6	l.lab	5.2c	11.8	49.6a
LSD		-	-	1.0	1.3	-	3.7
Significance							
Position(P)		NS	NS	NS	NS	NS	NS
Fertilizer(F)		NS	NS	*	**	NS	**
P x F		NS	NS	NS	NS	NS	NS
F rate, linear		NS	NS	*	**	NS	**

[1] Flower rating - 10=excellent, 0=no flower.

[2] Mean separation by Fisher's protected LSD, 0.05 level; means in columns followed by the same letter are not significantly different.

Vegetative Propagation of Atlantic White Cedar by Stem Cuttings

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Nature of Work: Atlantic white cedar [(Chamaecyparis thyoides (L.) BSP], also known as southern white cedar or swamp cedar, grows in a narrow coastal belt from Mississippi to southern Maine (2). It normally occurs on wet sites or in swamps near sea level, usually on very acid peat (3). Its distribution is scattered, owing to exacting site requirements, extreme sensitivity to fire, and the inability to compete on drier sites. In addition to landscape usage, the light-weight, rot-resistant wood of Atlantic white cedar is used for boat construction, waterfowl decoys, and house siding. Natural stands are disappearing at an alarming rate as a consequence of logging, fire, drainage, and forest conversion. This is causing concern about loss of genetic diversity.

Atlantic white cedar also has potential as a Christmas tree. It is similar in appearance to eastern redcedar (Juniperus virginiana L.), another Christmas tree species, but has the advantage of soft foliage compared to the prickly (awl-like) juvenile foliage of redcedar.

Little information has been published concerning vegetative propagation of this species by stem cuttings. The species has been rated as somewhat difficult to root from stem cuttings (1). Therefore, the objective of this research was to examine rooting of stem cuttings in relation to cutting length, growth stage (hardwood vs. softwood), and indolebutyric acid (IBA) treatment.

Hardwood and softwood stem cuttings were collected from a plantation of seedling Atlantic white cedar Christmas trees in Nash County, N.C. on February 6 and August 26, 1992, respectively. Trees were about 4 years old, with heights of approximately 2 m (6.6 ft). Cuttings were trimmed to lengths of 12 or 24 cm (4.7 to 9.4 in), treated with 0 to 15,000 ppm reagent grade IBA dissolved in 50% isopropyl alcohol and rooted in a raised greenhouse bench under intermittent mist.

Results and Discussion: When hardwood cuttings were collected in February, 12-cm (4.7-in) cuttings had 82% rooting, compared to 41% for 24-cm (9.4-in) cuttings. Survival, percent rooting, and root lengths were greatest for 12-cm (4.7-in) cuttings. Rooting response was best without IBA treatment, with one exception: root number on long cuttings was greatly increased by 15,000 ppm IBA. For softwood cuttings collected in late August, survival and rooting were virtually 100%, with or without IBA treatment, and 24-cm (9.4-in) cuttings had longer and more numerous

roots. Roots on softwood cuttings were about three times longer than those on hardwood cuttings.

Significance to Industry: Compared to other tree or landscape species that we have observed, stem cuttings (hardwood and softwood) of Atlantic white cedar root prolifically, making the species easy to propagate vegetatively. This would allow cloning of superior selections. In addition, new vegetative growth that arose from rooted cuttings was orthotropic (exhibited vertical growth). This would be advantageous for production of landscape, timber, and Christmas trees where straight stems are important.

Literature Cited:

1. Dirr, M. A. 1990. Manual of Woody Landscape Plants: Their Identification, Ornamental Characteristics, Culture, Propagation and Uses. 4th ed. Stipes Publishing Co., Champaign, Ill.

2. Korstian, C. F. and W. D. Brush. 1931. Southern white cedar. U. S. Dept. Agr., Forest Serv. Tech. Bul. 251.

3. Little, S. 1950. Ecology and silviculture of white cedar and associated hardwoods in southern New Jersey. Yale Univ. School For. Bul. 56.

Influence of Stock Plant Fertility on Adventitious Rooting of Stem Cuttings of Eastern Redcedar

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North Carolina

Nature of Work: Hardwood stem cuttings of eastern redcedar (Juniperus virginiana L.) were taken from containerized stock plants fertilized weekly with a complete nutrient solution containing 0, 5, 10, 20, 40, 80, 160, 320, or 640 ppm nitrogen. Foliar concentrations of mineral nutrients and carbohydrates within stock plants were measured prior to excision of cuttings. After 3 months in a propagation bed, data were collected on percent rooting, root count, root length, and root dry weight of cuttings (1).

Results and Discussion: Although growth of stock plants of eastern redcedar was optimal at 100-150 ppm nitrogen (2), rooting of stem cuttings was maximized at 20-40 ppm. This same concentration was required to elicit a threshold response, as responses at 20 ppm N were 1.6 (percent

rooting), 2.2 (root count), 1.5 (root length), and 6.3 (root dry weight) times those at 10 ppm. Nitrogen concentrations supraoptimal for growth of stock plants decreased rooting response. At 640 ppm nitrogen, all rooting parameters, except root length, declined significantly compared to maximum response.

Of the mineral nutrient concentrations measured in stock plant foliage (nitrogen, phosphorus, potassium, calcium, magnesium, manganese, and boron), only boron and potassium were significantly correlated with rooting response. Potassium was correlated with rooting percentage ($P=0.01$, $r=0.96$) and root length ($P=0.05$, $r=0.83$) whereas boron was correlated with rooting percentage ($P=0.01$, $r=.98$), root count ($P=0.05$, $r=0.80$), length ($P=0.05$, $r=0.85$), and dry weight ($P=0.05$, $r=0.76$). Boron is known to act synergistically with indoleacetic acid (IAA) to enhance adventitious rooting (4, 5), but potassium has previously been considered of little importance in studies of this type.

Foliar starch and sucrose concentrations within cuttings at time of excision were significantly correlated with percent rooting ($P=0.05$, $r=.84$) and root length ($P=0.05$, $r=.77$), respectively. Foliar concentrations of starch and sucrose fluctuate widely during the year in eastern redcedar (2), and may be associated with the seasonality that the species expresses in rooting response (3).

Although nitrogen fertilization of stock plants affected rooting, there were no significant correlations between foliar nitrogen concentrations and rooting response. This result underscores the limitations of interpreting adventitious rooting data in relation to a single applied nutrient without monitoring the status of other nutrients within stock plants.

Significance to Industry: Eastern redcedar is gaining popularity as an ornamental owing to diversity in growth form and tolerance to diverse cultural and environmental conditions. Although generally propagated sexually, seedlings of eastern redcedar display tremendous phenotypic variability. This research should benefit propagators in both the horticultural and forestry industries who wish to reduce this variability and produce the species vegetatively by stem cuttings. In addition, data herein provide insight regarding the relationship of stock plant fertility and adventitious rooting of stem cuttings.

Literature Cited:

1. Henry, P. H., F. A. Blazich, and L. E. Hinesley. 1992. Nitrogen nutrition of containerized eastern redcedar. II. Influence of stock plant fertility on adventitious rooting of stem cuttings. J. Amer. Soc. Hort. Sci. 117:568-570.

2. Henry, P. H., F. A. Blazich, L. E. Hinesley, and R. D. Wright. 1992. Nitrogen nutrition of containerized eastern redcedar. I. Growth, mineral nutrient concentrations, and carbohydrate status. J. Amer. Soc. Hort. Sci. 117:563-567.

3. Henry, P. H., F. A. Blazich, and L. E. Hinesley. 1992. Vegetative propagation of eastern redcedar by stem cuttings. HortScience 27:1272-1274.

4. Weiser, C. J. and L. T. Blaney. 1960. The effects of boron on the rooting of English holly cuttings. Proc. Amer. Soc. Hort. Sci. 75:704-710.

5. Middleton, W., B. C. Jarvis, and A. Booth. 1978. The boron requirement for root development in stem cuttings of Phaseolus aureus Roxb. New Phytol. 81:287-297.

Seed Germination of Rhododendron carolinianum: Influence of Light and Temperature

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Nature of Work: Research was conducted to examine the influence of varying photoperiods and a constant versus an alternating temperature on seed germination of Rhododendron carolinianum Rehd. (Carolina rhododendron) (1).

On November 6, 1989, mature seed capsules were collected from a native population of open-pollinated plants of R. carolinianum growing in Henderson County, North Carolina at an elevation of 700 m (2300 ft). Capsules were stored in a paper bag at 20° C (68°F) for 30 days. Seeds were then removed from the capsules and stored at a moisture content of 6% in a sealed glass bottle at 4° C (39° F). Moisture content of the seeds was determined by calculating the mean moisture content of six, 200-seed samples following drying at 105° C (221°F) for 24 hr.

In July and October 1990, seeds were removed from storage and graded by manual removal of abnormal, damaged, and undersized seeds. Graded seeds [approximately 825,000 pure seeds per 28g (1 oz)] were sown in covered 9-cm (3.5 in) glass petri dishes, each containing two prewashed germination blotters moistened with tap water. Following placement of seeds in the dishes, half were designated for germination at 25 ° C (77° F) and the other half for germination at an 8/16 hr thermoperiod of 25°/15°C (77°/59°F). All dishes were placed in black sateen cloth bags and the seeds allowed to imbibe overnight at 21°C (70°F). The next day, bags were randomized within two growth set at the appropriate temperatures. Cham-

ber temperatures varied within +0.5 °C (0.9°F) of the set point.
Within each temperature regime, seeds were subjected daily to the following nine photoperiods: total darkness, 1/2, two 1/2 hr photoperiods separated by 7 1/2 hr of darkness, 1, 2, 4, 8, 12, or 24 hr. Regardless of temperature, photoperiod treatments were administered the same time each day and for the alternating temperature of 25°/15°C (77°/59°F), all photoperiod treatments with the exception of total darkness and 24 hr irradiation began with the transition to the high temperature portion of the cycle.

Growth chambers were equipped with cool-white fluorescent lamps that provided a photosynthetic photon flux (400-700 nm) of 69 μmol-•m^{-2}• s^{-1} (5.3 klx) as measured at dish level with a cosine corrected LI-COR LI-185 quantum/radiometer/photometer (LICOR, Lincoln, Neb.). All photoperiod treatments, except total darkness and the 24 hr irradiation, were regulated by removal and placement of the petri dishes in black sateen cloth bags. For the 24 hr photoperiod treatment, the petri dishes remained continuously unbagged in open chamber conditions. Regardless of the photoperiod, temperatures within the petri dishes never exceeded ambient by more than 1° C (2° F). Petri dishes representing the total darkness treatment remained in black cloth bags throughout the experiment and all watering and germination counts were performed under a green safelight. Germination blotters were kept moist with tap water throughout the experiment. Seeds showing signs of decay were immediately removed from the dishes.

Each photoperiod was replicated four times within a temperature regime with a replication consisting of a petri dish containing 100 seeds. Germination counts were recorded every 3 days for 30 days. A seed was considered germinated when the emerging radicle was $\geq$ 1 mm (0.04 in). The experiment was conducted two times (July and October 1990).

For each experiment, percent germination was calculated as a mean of four replications per treatment. Data were subjected to analysis of variance procedures and regression analysis (3). Since there were no significant differences between the two experiments, only data from the October 1990 study are presented.

Results and Discussion: For both temperatures, no germination occurred during a 30-day period for seeds not subjected to light. At 25° C (77° F) increasing photoperiods increased germination with germination of 26 to 39% occurring by day 30 for the 12 and 24 hr photoperiods, respectively. The alternating temperature of 25°/15°C (77°/59°F) enhanced germination when light was limiting. At this temperature germination ≥58% occurred by day 30 for photoperiods 2 4 hr. For photoperiods 2 8 hr, 30-day germination ≥70% was realized.

Significance to Industry: Regardless of temperature, seeds of R. carolinianum required light for germination. However, the photoperiod which maximized germination varied, depending on the temperature. Seeds should not be covered during propagation because of their extremely small size [approximately 825,000 pure seeds per 28 g (1 oz)] and light requirement.

Literature Cited:

1. Blazich, F. A., S. L. Warren, M. C. Starrett, and J. R. Acedo. 1993. Seed germination of Rhododendron carolinianum: Influence of light and temperature. J. Environ. Hort. 11: (In press).

2. Downs, R. J. and J. F. Thomas. 1991. Phytotron procedural manual for controlled environment research at the Southeastern Plant Environment Laboratory. N. C. Agr. Res. Serv. Tech. Bul. 244 (Revised).

3. SAS Institute. 1985. SAS User's Guide: Statistics, Version 5 Edition. SAS Institute, Cary, N.C.

Tissue Culture Propagation of 'German Red' Carnation

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Texas

Nature of Work: 'German Red' carnation (Dianthus caryophyllus) is a brilliant red-flowered perennial with excellent environmental tolerances. The origin of this cultivar is unknown, but it has been observed growing in cottage gardens in rural Texas for many years (Welch, 1989) and is believed to have been introduced by German settlers over 100 years ago. Although carnations are generally poorly adapted to the southern U.S., 'German Red' is highly heat tolerant and continues to flower throughout the summer in Texas. In addition, the plant possesses good cold tolerance and has survived winter temperatures as low as -4° F (-20° C). In the San Antonio area, 'German Red' carnation blooms virtually year-round with peak bloom periods in the spring and fall.

The commercial production of 'German Red' carnation is severely limited by lack of suitable mass propagation methodology. Although cuttings can be rooted rather easily, the profuse flowering habit of this cultivar results in stock plants which produce predominately flowering shoots that yield poor cutting material. Cuttings taken from flowering shoots result in weak, spindly plants that have few, if any, branches and only a single terminal

flower. Such plants have poor post-propagation performance and consumer appeal. Because other carnations have responded well to tissue culture, the objective of the work reported herein was to determine the feasability of using micropropagation to produce large quantities of 'German Red' carnation. Specifically, we evaluated in vitro shoot multiplication from nodal explants, in vitro and ex vitro rooting of microcuttings, and performance following transplanting to the greenhouse and outdoors.

Explants were taken from both vegetative and flowering shoots of field-grown stock plants and consisted of 1/2 inch long stem segments containing a single node (Fig. 1). The explants were rinsed in distilled water, placed in 70% ethanol for 30 sec., and then rinsed 4-5 times with sterilized distilled water. Following this initial treatment, the explants were surface-sterilized with 5.25% sodium hypochlorite for 15 min. Following surface sterilization, the explants were placed on Murashige and Skoog's revised medium (Murashige and Skoog, 1962) supplemented with 100 ppm each of arginine, alanine, and ascorbic acid plus 2.0 ppm benzylamino purine (BAP) and 0.5 ppm naphthaleneacetic acid (NM) (hereafter referred to as "MS medium"). To minimize the potential deleterious effects of phenolics, 0.25% polyvinylpyrrolidone was added to the medium. After 15 days, shoots were subcultured onto fresh MS medium. At 21-day intervals thereafter, shoots were harvested and subcultured on fresh MS medium. The cultures were maintained at a temperature of 77° ± 2° F (25° ± 1° C) and under a 16 hour photoperiod (photosynthetically active radiation [PAR] of 40-50 μmol m^{-2} s^{-1}). At the end of each subculture period the number of shoots per culture was counted in at least 10 culture tubes. For rooting, shoots produced in vitro on MS medium were placed on 1/2 strength MS medium devoid of BAP and NM but with 0 or 0.1 ppm indole-3-butyric acid (IBA), or rooted ex vitro in 4-inch plastic pots filled with a commercial growing medium containing peat and perlite (Redi-Earth, Grace Hortic. Products, Cambridge, Mass.) The ex vitro rooted microcuttings were either left untreated (control) or dipped in Rootone F (0.2% 1-napthaleneacetamide, 0.1% IBA, and 4.04% tetramethylthiuramdisulfide) prior to placement in the rooting medium. Environmental conditions during rooting were the same as for shoot multiplication. The percentage of shoots with roots was determined daily by visual observation for the in vitro cultures and after 12 days for the plants rooted ex vitro. Following in vitro rooting, plants were transplanted to 4-inch plastic pots filled with Redi-Earth. Clear polyethylene wrap was placed over the pots and plants were maintained for seven days under the same environmental conditions as for shoot multiplication. The plastic wrap was removed after seven days. Seven days after removal of the plastic wrap, plants were transferred to a greenhouse without shading. The plants were maintained in the greenhouse for about one month after which they were transplanted outdoors at the Texas A&M Research and Extension Center in Dallas. Their performance (survival, flowering) outdoors was monitored for a period of three months.

Results and Discussion: After 4-5 days, explants taken from vegetative shoots began to exhibit budbreak at the node (Fig. 1). After 15 days, there were about 4 shoots per culture (Fig. 2). These shoots were then subcultured onto fresh medium and each subsequently produced about 10 additional shoots after 21 days.

About 18 shoots were produced per culture during the subsequent 21-day subculture periods. As long as shoots were subcultured every 21 days, no vitrification was observed and shoots remained healthy. When cultures were not subcultured regularly, however, shoots frequently became vitrified.

Explants taken from flowering shoots likewise exhibhed budbreak 4-5 days after inoculation. Shoots that developed in vitro, however, nearly all terminated in flower buds. These buds developed to anthesis but resulted in poor-qualhy propagules. Thus, for purposes of mass propagation, explants should not be taken from flowering shoots.

Shoots produced from vegetative explants began exhibiting adventitious rooting about five days after placement on 1/2 strength MS medium devoid of growth regulators. By 12 days after inoculation these shoots were well-rooted. The addition of 0.1 ppm IBA to the medium delayed rooting slightly but still resulted in 100% rooting after 12 days. These results indicate that IBA is not necessary for obtaining good in vitro rooting of 'German Red' carnation microcuttings.

One-hundred percent of the ex vitro microcuttings were rooted within 12 days regardless of whether or not they were treated with auxin (i.e. Rootone F). This indicates that in vitro rooting is not necessary for propagation of ‘German Red’ carnation and that ex vitro rooting of microcuttings may offer a more economical alternative. These findings also indicate, as with in vitro rooting, that auxin is of little value in promoting ex vitro rooting of ‘German Red’ carnation.

Almost all (> 98%) of the plants rooted in vitro or ex vitro survived transfer to the greenhouse. Thus, acclimation does not seem to be a problem for micropropagated ‘German Red’ carnation. Plants placed in the greenhouse grew rapidly and, within a month, high quality, well-branched plants were produced. All of these plants survived transplanting to outdoors and continued rapid growth. Within about 4 weeks after transplanting outdoors, these plants began flowering and the flowers were phenotypically identical to those on the original ‘German Red’ carnation stock plants.

Significance to the Industry: These findings suggest that in vitro mass propagation of 'German Red' carnation is feasible. In the present study,

high quality flowering plants were obtained within 5-6 months after taking nodal explants from field-grown stock plants (Fig. 3). Our results further indicate that a single explant can result in the production of about 720 microshoots within about two months (Fig. 4). These shoots served as microcuttings which were rooted in vitro or ex vitro with 100% success. Because virtually all of the rooted microcuttings survived acclimation and transfer to the greenhouse, about 720 flowering plants can be obtained from a single explant within 5-6 months. This of course may vary somewhat from lab to lab, but demonstrates that micropropagation from nodal explants can be a means for producing large numbers of this heat tolerant carnation from limited stock plant material. One caveat is that explants should not be taken from flowering shoots. Also, subculturing should occur every three weeks to avoid problems with vitrification.

Literature Cited:

1. Murashige, T. and F. Skoog. 1962. A revised medium for rapid growth and bioassays with tobacco tissue cultures. Physiol. Plant. 15: 473-497.

2. Welch, W.C.1989. Perennial Garden Color for Texas and the South. Taylor Pub., Dallas, Texas, pp.94-95.

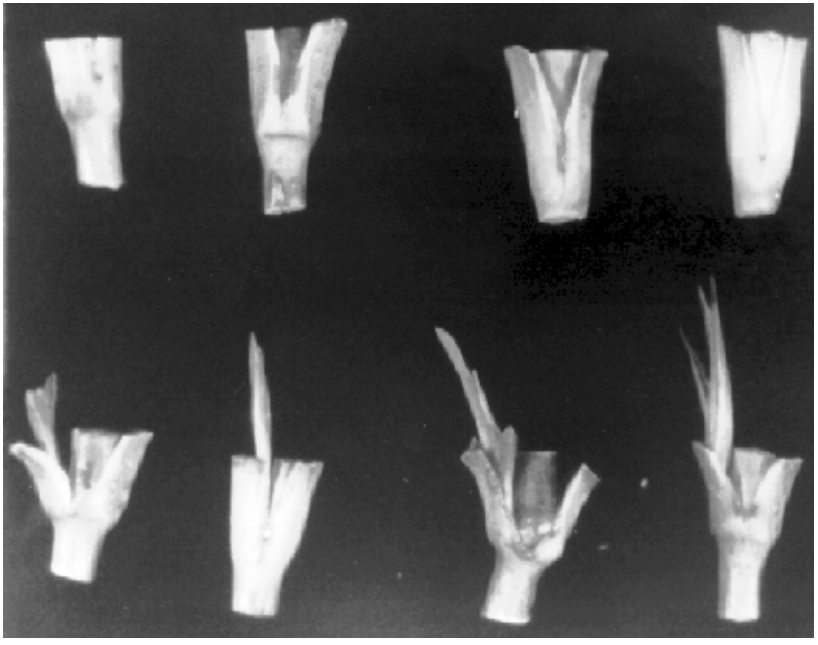
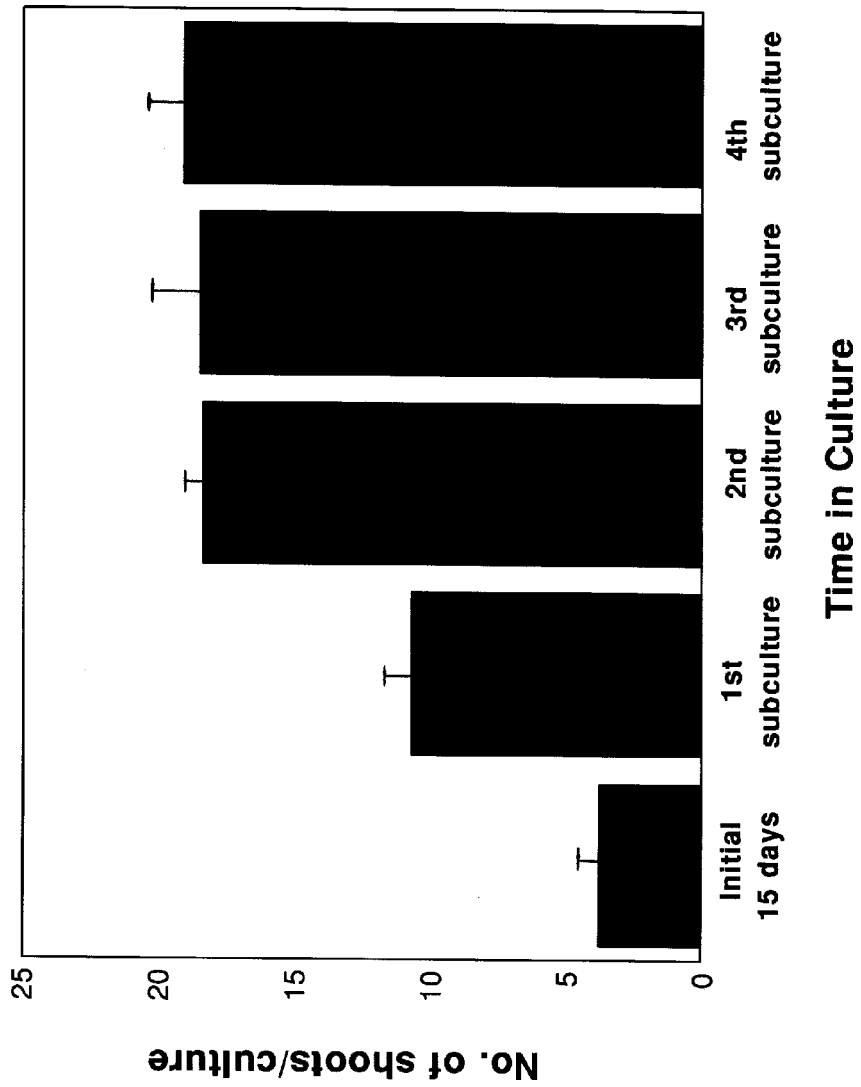
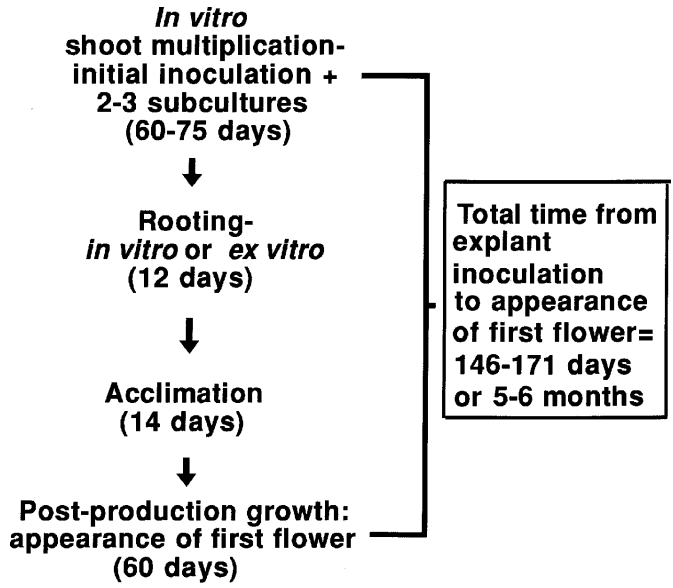


Fig.1. Photograph of 'German Red' carnation nodal explants at the time of inoculation (upper) and 5 days later (lower). Distance between lines



on scale is 1 mm (25.4 mm = 1 inch).

Fig. 2. Number of shoots per culture 15 days after inoculation and at the end of each subsequent subculture period. Each subculture period



was 21 days long. Bars indicate standard error of the mean.

Fig. 3. Flow chart summarizing the various stages of 'German Red' carnation micropropagation from nodal explants.

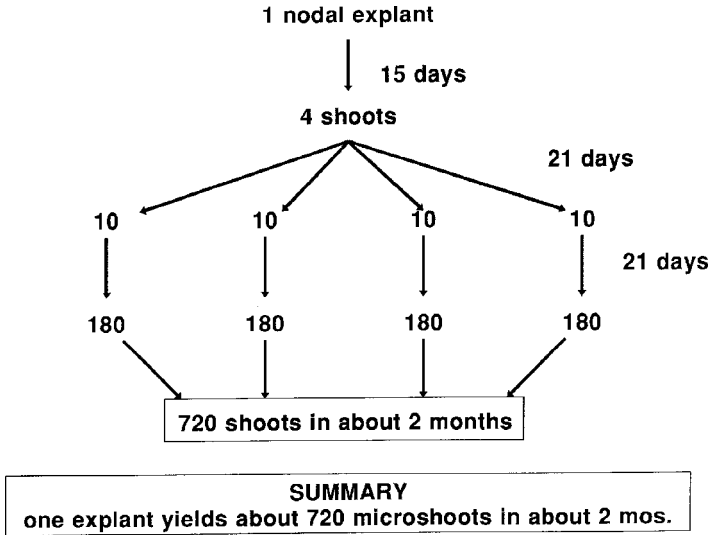


Fig. 4. Flow chart illustrating shoot multiplication from a single 'German Red' carnation nodal explant.

Evaluation of Four Fungicides on the Rooting of Three Ornamental Plant Species

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Nature of Work: Cuttings from propagation beds are often attacked by fungi such as Rhizoctonia, Pythium, Sclerotinia and Sclerotium. Fungicide recommendations are given for management of these fungi without knowledge of their effect on rooting.

Researchers have reported that certain fungicides can influence the rooting of certain herbaceous and woody plant species (1,2,3,4). Yet, some growers are currently using fungicide dips and drenches in propagation areas without clear evidence of their effect on rooting.

The purpose of this study was to evaluate the effect of four fungicides on the rooting of three plant species: Ilex x 'Nellie R. Stevens', Taxus x media 'Brownii', Juniperus chinensis 'Pfitzeriana'.

Cuttings were collected and stuck on January 12, 1993. Five inch cuttings were wounded on one side, immersed in a 5 second dip of Dip N' Grow (1:5 aqueous dilution) and stuck in a medium containing 70% (v/v) perlite and 30% Promix BX. Cuttings were placed under mist (12 sec/12 minutes). Twenty-four hours after sticking, cuttings were drenched with one of the following fungicides: Aliette 80WP, Chipco 26019 50WP, Cleary's 3336 4F or Subdue 2E.

Results and Discussion: All fungicide treatments resulted in increased rooting and root quality as compared to the untreated control, except for Cleary's 3336 on Ilex (figures 1 and 2). In the Ilex cuttings the Chipco 26019 and Subdue treatments had highest percentage rooting and best root quality. All fungicide treatments increased percentage rooting and root quality of both the yew and juniper cuttings compared to the untreated control.

In a similar unreported study involving the rooting of Taxus X media cuttings, Cleary's 3336 or Cleary's 3336 in combination with Subdue or Aliette improved the rooting and root quality.

Significance to Industry: Any benefit to applying fungicides drenches shortly after sticking cuttings appears to be dependent on the specific fungicide and plant species. It is yet to be determined if the numbers of cuttings rooting increased due to disease suppression or some plant growth regulator effect inherent to the fungicides tested. Future research will investigate the effect of fungicidal drenches on the rooting of a greater

number of woody plant species.

Literature Cited:

1. Hausbeck, M.K., C.T. Stephens and R.D. Heins. 1990. Size and flowering of seedpropagated geraniums in response to fungicide drenching schedules. HortScience 25 (6) :644-646.

2. Lee, Leland W., K.C. Sanderson and J.G. Williams. 1983. Effect of fungicides applied to polyurethane propagation blocks or rooting of poinsettia cuttings. HortScience 18 (3) :359-360.

3. Morgan, D.L. and P.F. Colbaugh. 1983. Influence of chemical sanitation treatments on propagation of Buxus microphvlla and Peperomia caperata. Proc. Inter. Plant Prop. Soc. 33:600-607.

4. Powell, C.C., Jr., 1989. The safety and efficacy of fungicides for use in Rhizoctonia crown rot control of directly potted unrooted poinsettia cuttings. Plant Disease 72:693-695.

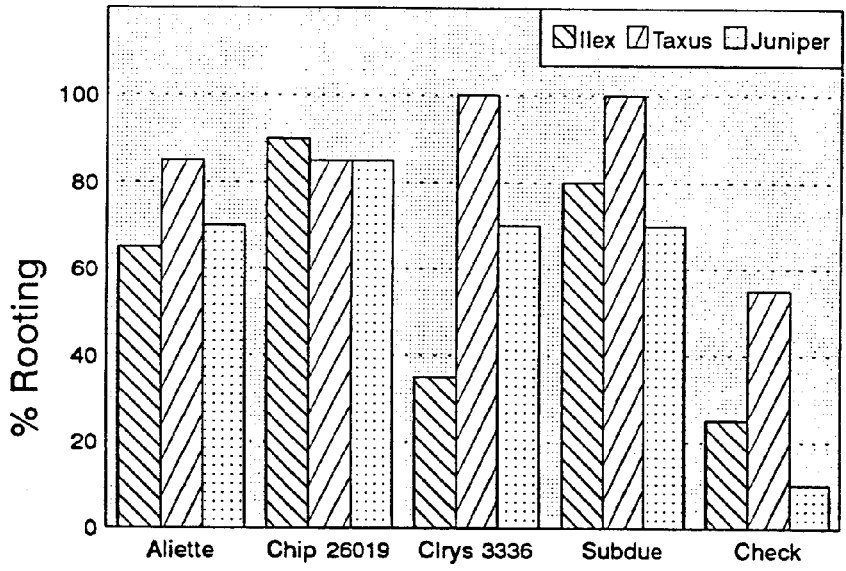

Figure 1. Effect of fungicide drenches on rooting of Ilex x 'Nellie R. Stevens', Taxus x media 'Brownii', Juniperus chinensis 'Pfitzeriana'.

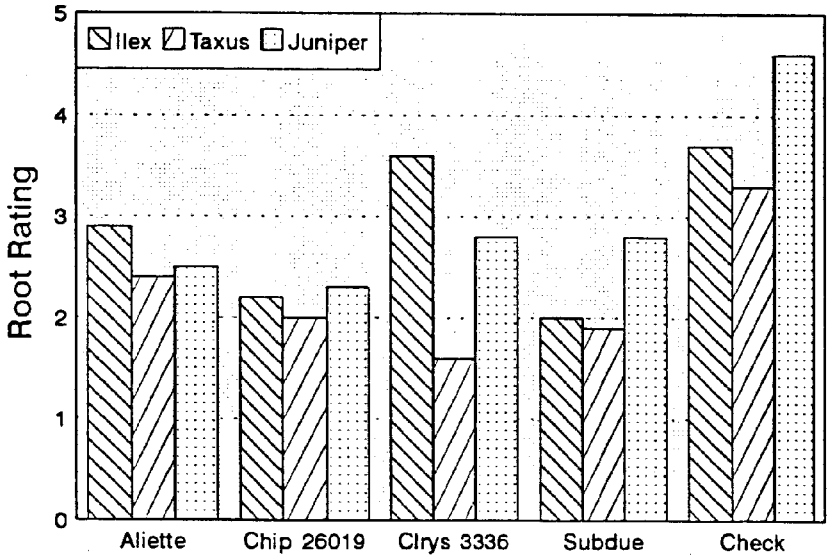

Figure 2. Effect of fungicide drenches on root quality of Ilex x 'Nellie R. Stevens', Taxus x media 'Brownii', JuniDerus chinensis 'Pfitzeriana'. Root Rating Scale: 1 = large root system, 2 = medium root system, 3 = small root system, 4 = callus tissue only, 5 = no callus tissue.

Root Initiation and Development for Chinese Fringetree Stem Cuttings as Influenced by Auxin Application

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Nature of Work: Stem cuttings of Chinese Fringetree (Chionanthus retusus Lindl. & Paxt.) are generally considered difficult to root. Dirr (1) reported about 50% of stem cuttings treated with a 1.0% IBA quick dip solution formed roots. Although not published, propagators in Tennessee and Alabama have reported higher rooting percentages, 85 to 97%, when cuttings were treated with 0.8 to 2.0% IBA in talc (1). Regardless of auxin treatment, recently hardened, juvenile cutting wood appears to be essential for success in rooting. The objective of this study was to evaluate the influence of auxin type, concentration and method of application on root initiation and development of Chinese Fringetree stem cuttings.

Recently hardened-off, terminal, 6.0 inch (15.2 cm) singlestem cuttings from 2 year old container grown plants of Chinese Fringetree were prepared on September 24, 1992. Prior to sticking, basal leaves were stripped from the stem, and cuttings were submerged in a Daconil (chlorothalonil) fungicide solution and allowed to drain. The basal end of the cuttings was cut at an angle and treated with one of 8 auxin treatments before being stuck in 78.8 in^3 (1291.3 cm^3) plastic pots containing a 100% vermiculite medium. Auxin
treatments were: 3,000, 8,000 and 16,000 ppm IBA as K-IBA liquid quick dips and commercial talc preparations (Hormex Nos. 3, 8 and 16, respectively; Brooker Chemical Corp., North Hollywood, CA), 10,000 ppm NAA as a liquid quick dip, and an untreated control. Quick dip treatments consisted of dipping the basal end of cuttings into respective auxin solutions for 5 seconds and allowing to dry for 60 sec before sticking.

Cuttings were placed in an intermittent mist bed with a polyethylene covering. Initial mist settings were 4 sec every 4 min from 8:00 AM to 4:30 PM for the first 2 weeks. Thereafter, mist settings were adjusted based on day-length and stage of root development. The experimental design was a randomized complete block design with 5 blocks of 5 cuttings per treatment. Cuttings were harvested and evaluated on December 10, 1992. Data included: percent rooted, root quality, total root number and average root length (3 longest roots per cutting/3).

Results and Discussion: All auxin treatments increased rooting %, root quality rating, root length and root number when compared to the untreated control (Table 1). Cuttings treated with 1.0% NAA as a quick dip had the highest root rating, longest average root length and greatest root number compared to the other auxin treatments, with the exception of the 1.6% K-IBA quick dip treatment which was similar. The 1.6% K-IBA quick dip treatment was more effective than the 1.6% IBA talc treatment having greater rooting %, root quality rating, average root length and root number. In general, as quick dip K-IBA concentration increased rooting %, root quality rating, average root length and root number increased, while root evaluation parameters were similar among the IBA talc treatments.

Significance to Industry: The greatest number of stem cuttings to root (72 to 84%) and the highest quality cuttings of Chinese Fringetree were produced with 1.0% NAA or 1.6% K-IBA as quick dips for the treatments evaluated in this study. Further research is required to determine if higher concentrations of NAA or K-IBA would improve these results.

Literature Cited:

1. Dirr, M.A. and C.W. Heuser, Jr. 1987. The reference manual of woody plant propagation. ISt ed. Varsity Press, Athens, GA.

Table 1. Effect of auxin application on root initiation and development for Chinese Fringetree stem cuttings.[z]

Auxin treatment	Rooting (%)	Root quality rating[y]	Root length (in)[x]	Root number
Quick dip				
1.0%, NAA	84a	2.8a	3.0a	26.4a
0.3%, K-IBA	56bc	1.9bc	2.Obc	4.8bc
0.8%, K-IBA	56bc	2.Obc	1.8bc	8.2b
1.6%, K-IBA	72ab	2.5ab	2.7ab	21.3a
Talc				
0.3%, IBA	60abc	1.9bc	1.7bc	1.8c
0.8%, IBA	40c	1.5c	1.3c	3.2bc
1.6%, IBA	44c	1.8c	1.6c	4.3bc
Untreated control	12d	0.8d	0.3d	0.2c

[z] Cuttings were stuck September 24, 1992 and harvested December 10, 1992.

[y] Root quality was rated on a scale from 0-5 where 0 = no roots or callus and 5 = heavy root system with secondary branching.

[x] Root length was the 3 longest roots per cutting/3.

[w] Mean separation in columns by LSD, p = 0.05.