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Propagation of Fraser Fir¹

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– Abstract –

Fraser fir [Abies fraseri (Pursh) Poir.], the most important Christmas tree species in North Carolina, is rapidly gaining popularity nationwide. It is propagated by seed, but special needs such as genetic improvement will involve use of grafting, air layering, propagation by stem cuttings, and micropropagation (tissue culture). This paper summarizes research conducted to date on Fraser fir concerning these techniques in addition to information regarding seed production and sexual propagation.

Index words: Christmas trees, Abies fraseri, tissue culture, grafting, air layering, asexual propagation, micropropagation.

Significance to the Nursery Industry

Most Fraser fir Christmas trees are of seedling origin. Genetic improvement of the species will involve vegetative propagation. Fraser fir is easy to graft and air layer, and readily produces roots on stem cuttings. However, plagiotropic growth of rooted cuttings is a major problem that limits commercial propagation of Fraser fir by cuttings. Hedges that produce orthotropic shoots are one feasible source of cutting material. Efforts to micropropagate Fraser fir have had little success, but serve as a foundation for future research.

Introduction

Fraser fir [*Abies fraseri* (Pursh) Poir.] is highly prized as a Christmas tree because it has dark green, fragrant foliage, a natural Christmas-tree shape, and good keeping qualities. Approximately 4 to 5 million Christmas trees were cut in North Carolina in 1993, with a farmgate value of 80–100 million dollars. Fraser fir comprises at least 90% of the annual state production. Until the 1950s, natural-grown Fraser firs were harvested from mountain tops (western North Carolina, southwestern Virginia, and eastern Tennessee) and sold mostly in local markets. Fraser fir was practically unknown in the national market in 1970. Today, it is one of the most popular species nationwide, and regularly wins the biennial contest sponsored by the National Christmas Tree Association. President Clinton's tree in 1993 was a Fraser fir from Mitchell Co., N.C.

Distribution. Fraser fir is one of two Abies species indigenous to eastern North America. It is thought to hybridize with balsam fir (Abies balsamea (L.) Mill.], to produce bracted balsam fir (Abies balsamea var. phanerolepis Fern.) (18). Fraser fir, naturally occurs between 1130-2040 m (3700–6700 ft) (Fig. 1A) in a few scattered locations in the Appalachian Mountains of western North Carolina, eastern Tennessee and southwestern Virginia (Fig. 1B) (18). Production in North Carolina is limited to the mountainous, western counties at elevations principally between 915 and 1370 m (3000–4500 ft).

Seed Production and Sexual Propagation of Fraser Fir

Seed production. Trees produce few viable seeds prior to 20 to 30 years of age, and bountiful crops only occur every 3 to 5 years (10). The balsam woolly adelgid (Adelges piceae Ratz.) is destroying mature, native stands where most seed is collected. The North Carolina (N.C.) Forest Service has established several seed orchards to insure a seed source for nursery production. One orchard (established in 1974) in Crossnore, N.C. now provides all the seed used by the N. C. Forest Service for production of 3-year-old seedlings sold to the public.

Other agents are also blamed for the decline and destruction of mature stands. Acid deposition has been implicated (3). In some stands, the loss of mature, seed producing trees is so extensive that reproduction has ceased (J.B. Jett, N.C. State Univ., personal communication). It is possible that Fraser fir could eventually become extinct in the wild, existing only in cultivation.

Seeds and sexual propagation. Seeds of Fraser fir are produced in one growing season. There are approximately 60,000 cleaned seeds per pound (0.45 kg) (10). Germination is inherently low and rarely exceeds 55% (10).

Stratification (moist-prechilling) for 4 to 8 weeks at 4° C (39°F) accelerates germination, broadens the range of temperatures over which germination occurs, and reduces sensitivity of seeds to light (1). Light stimulates germination, but there is no obligate light requirement (1). The light effect is linked to phytochrome (13).

Under laboratory conditions, the optimum germination temperature for nonstratified seeds of Fraser fir is an 8/16hr cycle of $30^{\circ}/20^{\circ}$ C ($86^{\circ}/68^{\circ}$ F) with light provided during the 8-hr portion of the cycle (10, 12, 17). Germination is maximum when seeds receive about 500 to 600 degree-hours per 24-hr cycle (1). One hour of light during the high temperature portion of the cycle maximizes germination (1). However, the timing of irradiation is critical; to maximize

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germination, seeds must be irradiated during the latter part of the warm portion of the cycle (13). Given sufficient time, germination can occur at 4°C (39°F); at this temperature, seeds subjected to intermittent light germinate faster than seeds maintained in darkness (4).

Vegetative Propagation

Fraser fir is relatively easy to air layer, and graft (11). When grafting, scions are usually collected in early spring before budbreak, and cleft grafted onto 5- or 6-year-old, well established transplants. The graft union is wrapped with a budding rubber band and Parafilm, and no other covering is

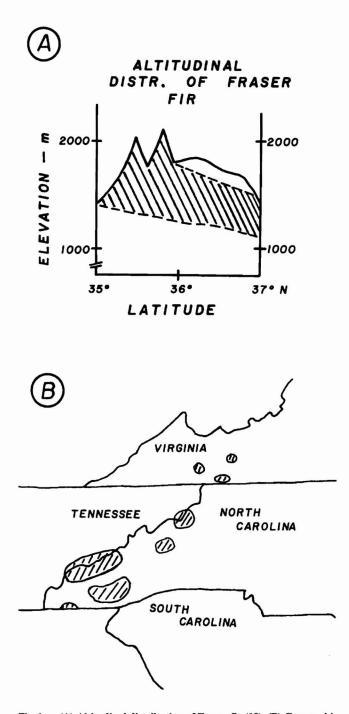


Fig. 1. (A) Altitudinal distribution of Fraser fir (18). (B) Geographical distribution (18).

required. Success is normally very high. These techniques are useful in establishment of seed orchards, but are impractical for producing large numbers of plants. Therefore, other methods of vegetative propagation are needed. Our efforts have concentrated on (a) propagation by stem cuttings and (b) micropropagation.

Propagation by Stem Cuttings

Time of collection. Early experiments initially utilized hardwood cuttings taken from dormant plants during November through February. These cuttings rooted well, but there were some problems. The succulent, elongating shoots were susceptible to gray mold (*Botrytis cinerea* Pers.). Because the chilling requirement was already satisfied, these cuttings tended to break bud quickly. This led to an apparent competition for internal resources between expanding shoots and the developing roots (28). As a result, shoot growth was weak, with poor bud development. Subsequent vigor and growth were also weak. At the end of the rooting phase, these cuttings were out of synchrony with natural growth cycles. This caused problems in moving them outdoors and achieving normal growth afterwards.

Softwood cuttings can be taken when shoot elongation has ceased and the tissue is beginning to mature. These shoots have differentiating terminal buds (21) that continue to de-

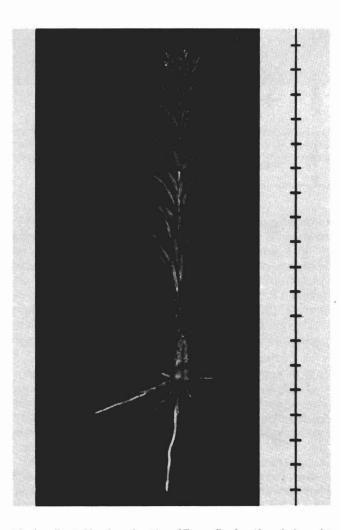
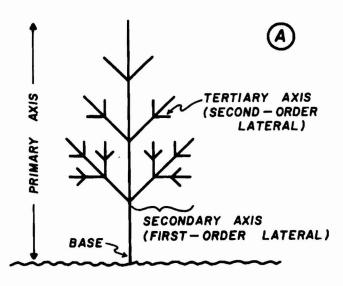


Fig. 2. Rooted hardwood cutting of Fraser fir after 10 weeks in a mist bed. Scale divisions = 1 cm (0.4 in.).

velop but do not break during the rooting phase. For cuttings from 14-year-old stock plants, the best balance of root formation and shoot growth has been achieved with cuttings from the upper third of the crown just prior to lignification (late June in western North Carolina) (28). These cuttings exhibited superior responses when they were wounded and treated with 1500 ppm (0.15% in 50% isopropyl alcohol) indolebutyric acid (IBA) for 1 sec prior to insertion into the rooting medium. Following a 10-week rooting period, cuttings were potted and placed outdoors in the fall. By spring, following natural chilling during the winter, complete budbreak occurred, resulting in plants comparable to 2-year old plants propagated by hardwood cuttings (28).

Cuttings collected in early October (designated semi-dormant) require less chilling for rooting than for budbreak (20). In one experiment, stem cuttings were collected from 14-year-old stock plants the first week in October, and chilled in a dark 4°C (39°F) coldroom for 0, 4, or 8 weeks (27). After a 135-day rooting period, rooted cuttings from the 0-, 4- and 8-week chilling durations were placed in a 4°C (39°F)



MULTIPLE TERMINALS

Fig. 3. Location on stock plants from which cuttings were taken. (A) Schematic of branch order, (B) Multiple terminals used as a source of cuttings.

coldroom under an 8-hr photoperiod for 11, 7 or 3 weeks, respectively, so that all plants received a total of 11 weeks chilling during the experiment. In April, plants were placed outdoors under 50% lath shade in a gravel-based nursery production area. Vegetative growth was measured in August. Best overall responses were achieved with upper-crown cuttings chilled 4 weeks and then treated with 4500 ppm (0.45%) IBA, or lower crown cuttings dipped in 1500 ppm (0.15%) IBA after 4 weeks of chilling (27). Similar to results with softwood cuttings, the separation of rooting and budbreak for semi-dormant hardwood cuttings produced rooted cuttings comparable in size to 2-year old plants from traditional hardwood cuttings.

Using softwood and semi-dormant hardwood cuttings eliminates incomplete budbreak and weak shoot growth, but plagiotropic growth still occurs. Plagiotropism varies by collection date as well as origin of cuttings on the tree (27, 28, 29, 30). Staking softwood cuttings during rooting reduces the severity of plagiotropic growth (30).

Auxin treatment. Treatment with auxin is essential (14, 15). With hardwood cuttings, rooting is good with the free acid of IBA applied at a concentration of 5000 ppm (0.5% in 50% isopropyl alcohol) for 1 sec to the basal 2 cm (0.8 in) of a cutting (14, 15). Similar results can be obtained with Hormodin 3 [8000 ppm (0.8%) IBA in talc] (15). However, applying IBA as a concentrated solution gives more consistent results. IBA concentrations of 1500–4500 ppm should be used for softwood and semi-dormant hardwood cuttings.

Wounding. When used in combination with auxin treatment, wounding improves rooting (15). An effective wounding treatment is four equally spaced vertical cuts (light wounds) into the base of the cutting, each about 2.5 cm (l in) in length and parallel to the long axis of the cutting. Extensive swelling of the bases occurs in auxin-treated cuttings in the area of the wounds (Fig. 2). Roots normally emerge near the cutting base; rarely along margins of wounds.

Length of cuttings. We have tested cuttings ranging in length from 9 to 25 cm (3.5 to 10 in). Generally, rooting increases with cutting length, but budbreak is better on shorter cuttings (19). We suggest a length of 13 to 17 cm (5 to 6.5 in). During preparation of cuttings, needles are removed from the basal 4 cm (1.5 in), followed by wounding and treatment with auxin, as described above.

Crown position and branch order. Branch order influences rooting of Fraser fir cuttings. Most of our rooting studies have utilized tips from secondary axes (first-order laterals, Fig. 3). First- and second-order laterals root similarly, and generally root better than primary axes (19). Cuttings from tips of primary axes grow orthotropically and produce symmetrical trees (Fig. 4A). Virtually all lateral cuttings are plagiotropic the first year following rooting (Fig. 4B, plant on the right). Plagiotropism is more pronounced with cuttings from older trees.

Stock plant age. Rooting capacity of Fraser fir decreases with stock plant age. Terminal cuttings from 4-to 5-year old plants of seedling origin usually root in high percentages. In one study, rooting percentages of first-order laterals of



Fig. 4. (A) Fraser fir Christmas tree propagated by a rooted cutting and ready for harvest. Initially, the cutting was a primary axis (orthotropic shoot) taken from a 3-2 transplant (5-year old plant) having multiple terminals. Photograph was taken after 10 growing seasons in the field. (B) Rooted, lateral hardwood cutting of Fraser fir exhibiting plagiotropic growth (plant on the right); normal, orthotropic transplant of seedling origin (plant on the left).

5-, 12- and 22-year old trees (14) were 75%, 13%, and 4%, respectively, for nontreated cuttings; and 92%, 50%, and 29%, respectively, for cuttings that received wounding and auxin treatment (14).

Genotype (intraspecific) variation. There is great variation in rootability among genotypes of Fraser fir. In one experiment using 12-year-old trees, rooting ranged from 3% to 78% (19). This problem could hinder attempts to root cuttings from individual trees with desirable Christmas tree traits.

Rooting environment.

Bottom heat. Rooting is usually best when a cool to moderate air temperature is used with a warm rooting medium. Most of our experiments with hardwood cuttings have been conducted in heated greenhouses where ambient day/night air temperatures were $24^{\circ} \pm 5^{\circ}/14^{\circ} \pm 4^{\circ}C$ ($75^{\circ} \pm 9^{\circ}/57^{\circ} \pm 7^{\circ}F$); day/night rooting medium temperatures $21^{\circ} \pm 3^{\circ}/16^{\circ} \pm 3^{\circ}C$ ($70^{\circ} \pm 5^{\circ}/61^{\circ} \pm 5^{\circ}F$). Under these conditions bottom heat [18° to $24^{\circ}C$ (65° to $75^{\circ}F$)] promotes rooting (15). Rooting medium temperatures $\leq 16^{\circ}C$ ($60^{\circ}F$) inhibit or slow rooting and should be avoided. *Photoperiod.* In fall-collected cuttings that have received a limited amount of chilling (3 weeks), rooting is improved by interrupting the night with a 3-hr light break [100 W incandescent bulbs about 70 cm (28 in) above the surface of the rooting medium and spaced 1 m (39 in) apart] (20). After exposure to longer durations of chilling (9 or 12 weeks), photoperiod does not affect percent rooting (20). There is no significant relationship between photoperiod during rooting and the number and length of roots per rooted cutting.

Intermittent mist. All our rooting research has utilized intermittent mist; beds employ in-bed installation of deflection type mist nozzles. Timing (regulation) of mist is controlled by a 24-hr timer and a short-interval timer, and operates only during daylight hours. Rooting media and cuttings should be kept moist but not overly wet.

Much of our research has involved rooting cuttings of Fraser fir in raised greenhouse benches containing rooting medium. We have also rooted cuttings in small individual containers, ground beds, flats, and flats composed of individual cells. Containers and media should be well drained. Two good rooting media are 1 peat:1 perlite (by vol) or 1 peat:1 sand (by vol).

Orthotropic shoots and hedging

If stem cuttings from lateral branches of Fraser fir did not exhibit plagiotropic growth following rooting, or if persistence of such growth lasted only 1 or 2 years, stem cuttings could be used to clone superior selections. This approach is currently utilized to propagate elite Douglas-fir [*Pseudotsuga menziesii* (Mirb.) Franco] Christmas trees in the Pacific Northwest (22). With Fraser fir, plagiotropic growth of rooted cuttings persists several years, which would seriously limit the production of Christmas trees from lateral branches.

In general, rooted cuttings often show a slower growth rate with increasing age of the stock plant (8). Thus, rooted cuttings from 5-year-old trees would likely grow faster than similar cuttings from 14-year-old trees. This would tend to lengthen rotations and increase costs. We have not systematically studied growth rates. However, on the basis of limited observations, rooted orthotropic shoots from seedlings and transplants appear to require more time to produce a Christmas tree, compared to plants of seedling origin.

Because orthotropic shoots produce normal, upright trees (Fig. 4A), we carried out experiments involving decapitating or hedging stock plants to maximize the yield of orthotropic shoots (29). Various percentages of top growth were removed from 4- and 14-year-old stock plants. The greatest yield occured when most of the aerial part of the plant was removed. This resulted when decapitation removed half of the 2-year-old wood from 4-year-old plants and when 14-year-old trees were cut off 3 cm (1.2 in) above the lowest whorl of branches. Dikegulac [as Atrimmec; sodium salt of $2,3:4,6-bis-O-(1-methylethylidene)-\alpha-L-xylo-2$ hexulofuranosonic acid] sprays at 1000 or 3000 ppm were of no benefit.

In hedging studies, 4-year-old stock plants produced less than five shoots per year, and 14-year-old plants produced less than 20 shoots (29). Rooting of cuttings varied according to their location of origin on stock plants. Vertical leaders from 4-year-old plants rooted slightly better (77%) than lateral branches (66%). For plants that were partly decapitated, rooting of plagiotropic and orthotropic shoots was 92% and 30%, respectively (29).

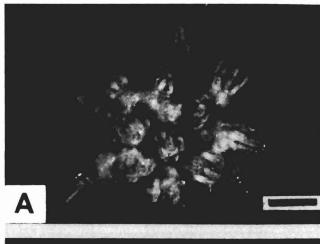
Rooting differences were more pronounced in older trees. Orthotropic shoots that developed on decapitated, 14-yearold plants had 46% rooting, compared to 99% for lateral branches (29). For those shoots which rooted, the percentage that survived the next season (31%) and remained orthotropic (23%) was much lower. At the end of the growing season following rooting, the greatest number of orthotropic shoots per original stock plant resulted from treatments which removed the greatest amount of top growth. Future efforts should probably concentrate on increasing yield, rooting percentage, growth rate, and survival of orthotropic shoots from trees of Christmas tree age (12 to 14 years from seed).

Micropropagation

Micropropagation can be used to propagate many conifers including economically important species such as Douglas fir, loblolly pine (*Pinus taeda* L.), Monterey pine (*Pinus radiata* D. Don) and Norway spruce (*Picea abies* L.) (7). Procedures normally utilize embryonic tissues (explants from zygotic embryos). These embryos are genetically different from either parent tree. Efforts to directly clone select trees have had limited success (2, 6, 9, 16). However, research with embryonic explants has laid the groundwork for current and future research, which hopefully, one day, will permit operational vegetative propagation from mature trees. Preliminary attempts to micropropagate Fraser fir by utilizing explants from vegetative tissues were unsuccessful, and led the authors to conduct research with embryonic explants.

Results thus far regarding tissue culture propagation of Fraser fir from embryonic explants have been disappointing. Cotyledons produce few adventitious buds in vitro (25). Buds form mainly on hypocotyls, and numbers are low (average of two to three buds per hypocotyl) (Fig. 5). Bud elongation is poor due to bud dormancy. Budbreak has been difficult to accomplish, but we have been able to achieve limited elongation and to root some of the shoots, resulting in plantlets. Rooting of shoots is also problematic, with room for improvement (24, 25). We have been unable to acclimate the plantlets to greenhouse conditions following transfer to a growing medium. Research is continuing to increase in vitro bud production on hypocotyls and to determine why buds do not form on cotyledons.

Attempts by others to induce shoot elongation by culturing vegetative buds in vitro, followed by rooting, have largely failed owing to the inability to form roots (26). Shoot elon-



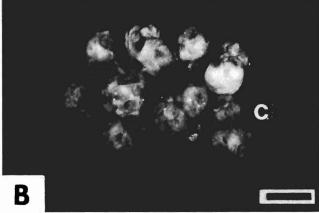


Fig. 5. In vitro adventitious bud development on embryonic explants of Fraser fir (The response of these particular explants was greater than average.) Scale bars = 2 mm (0.08 in). (A) Explant, consisting of a hypocotyl. (B) Explant consisting of a hypocotyl with attached cotyledons (c). The radicle has been removed. Note the unresponsive cotyledons.

gation is also a problem (26). In addition, bombardment of buds with microprojectiles coated with DNA has succeeded only in short-term, transient gene expression (26).

The difficulties encountered with Fraser fir and balsam fir (5, 24, 25, 26, 31) suggest that cloning elite Fraser fir by micropropagation is probably years away. Therefore, vegetative propagation with stem cuttings from hedging-derived, orthotropic shoots appears to have more short-term potential for mass propagation of select trees.

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