Abstract

Gocke, Matthew Harrison. Production system influences the survival and morphology of rooted stem cuttings of loblolly pine (*Pinus taeda* L.) and sweetgum (*Liquidambar styraciflua* L.) (Under the direction of Barry Goldfarb and Daniel J. Robison.)

Forest planting stock must be capable of high rates of survival and good field performance to justify the expense of reforestation efforts. Seedling grading standards have improved the quality of forest planting stock and have increased expectations for survival and field growth of out-planted forest seedlings. For many tree species, rooting stem cuttings provides an alternative means of producing planting stock to that of conventional seedling propagation. Use of rooted stem cuttings (rooted cuttings) in forestry has many potential advantages for both research and operational applications. However, to realize these benefits, it is important that high quality rooted cuttings are produced to enable field performance on par with seedlings of the same species and similar provenance. Developing specific grading standards for rooted cutting planting stock, therefore, is critical for successful field performance, and, is a topic of increasing interest for clonal forestry of particular species.

Grading standards for rooted cuttings may differ from those of seedlings, because of potential biological differences and increased production costs for rooted cuttings. Furthermore, various production systems exist capable of producing high quality rooted cutting planting stock and may require individual grading standards. Loblolly pine and sweetgum, to a lesser degree, are two commonly out-planted forest tree species in the southeastern United States. Seedling grading standards exist for both species in this region. Increased interest in clonal propagation of loblolly pine and sweetgum requires development of rooted cutting grading standards to ensure high rates of survival and good field performance.

Two studies conducted in 2000 and 2001 investigating rooted cutting production systems for loblolly pine and sweetgum are described in the following two chapters. The effects of a transplant, a containerized, and a direct-stick production system on

morphological characteristics of loblolly pine rooted cuttings were evaluated in the first chapter. Morphological comparisons were made among the various stock types tested. In the second chapter, feasibility and morphological effect of a transplant, a containerized, and a direct-stick rooted cutting production system were evaluated for sweetgum. Semi-hardwood (SH) stem cuttings of sweetgum were tested in all three production systems with special emphasis placed on the presence of new shoot growth following rooting. Hardwood (H) stem cuttings of sweetgum were also rooted in a direct-stick system in an outdoor nursery bed to test the reported ability of this cutting type to produce new shoot growth in the same season as rooting.

Rooted cutting morphology varied among clone and production system for both loblolly pine experiments. By the second loblolly pine experiment, over 90% of the rooted cuttings produced in the systems tested met acceptable seedling grading standards, including the second cycle (May sticking). Results of this study demonstrated that all three production systems evaluated were capable of producing high quality planting stock and that two full production cycles can be obtained in one growing season in the containerized and transplant systems.

All four production systems evaluated in the sweetgum study produced rooted cuttings. Morphological measurements varied among these same rooted cuttings according to production system. The transplant and containerized systems produced a large number of rooted cuttings with high rates of survival and large root systems. The SH direct-stick system produced rooted cuttings with sizeable root systems, but proved more sensitive than the other systems tested. A SH direct-stick system requires a back up irrigation system and a secondary power source to be effective. The H direct-stick production system was the only system to produce rooted cuttings exhibiting substantial shoot growth during the first growing season. Some of these rooted cuttings also developed extensive root systems, but survival was low.

Production system influences the survival and morphology of rooted stem cuttings of loblolly pine (*Pinus taeda* L.) and sweetgum (*Liquidambar styraciflua* L.)

by

Matthew Harrison Gocke

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(Co-Chair of Advisory Committee)	(Co-Chair of Advisory Committee)

Dedication

To Pops and the plants around his pool.

Biography

Matthew Harrison Gocke was born on May 20, 1969 in Morgantown, West Virginia. As the son of a drafted Army Captain, he moved around the country with his family, living in Charlottesville, VA; Junction City, KS; and Wheaton, MD. At the age of five he moved with his family to his mother's home town of Raleigh, NC. Matt graduated from Jesse O. Sanderson High School in Raleigh, NC in 1987. After graduation, he attended the University of North Carolina at Chapel Hill where he received a Bachelor of Arts degree in Education with a major concentration in Secondary Education and Social Studies. Matt played bass guitar and drums, traveled, landscaped, and bussed tables following his first college experience. In the Spring of 1999, Matt returned to academic life, this time at North Carolina State University where he began studying for a Master of Science Degree in Forestry.

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I would also like to thank my parents and my brothers for their endless support, sometimes leading to hours of sticking cuttings, planting trees, or weeding around stock plants. We are all better gardeners.

Finally, and most of all, I would like to thank my lovely and wonderful wife, Dara. She has never known me not as a graduate student. Thank you for your love and understanding.

Table of Contents

Table of Contents (continued)

List of Tables

Loblolly pine rooted cutting experiments

Sweetgum rooted cutting experiments

List of Tables (continued)

Sweetgum rooted cutting experiments

List of Figures

Loblolly pine rooted cutting experiments

Sweetgum rooted cutting experiments

Literature Review

Stock Quality of Seedlings and Rooted Cuttings

Very young trees aged 1 to 4 years, generally, prepared for forest plantings are referred to as planting stock. Stock quality measures the ability of this planting stock to survive and grow rapidly after out-planting (Thompson 1985, Mexal and Landis 1990). Evaluating stock quality offers nurserymen and reforestation managers useful information for predicting the success and reducing the costs of reforestation efforts (Leaf et al. 1978, Dunsworth 1997, Mohammed 1997, Tanaka et al. 1997).

Production Systems

Several production systems have been developed to provide a variety of planting stock types suitable for reforestation. Plants cultured as seedlings, rooted stem cuttings, *in vitro* plantlets, and seedlings derived from somatic embryos are grown typically under one of three production systems; in outdoor nursery beds as bare-root stock, in containers as containerized stock, or initially as containerized stock and then transplanted to an outdoor nursery bed as transplant stock (Stein et al. 1975, Hahn 1984, Menzies and Arnott 1992). Each of these three production systems provides unique advantages and disadvantages for reforestation efforts (Menzies and Arnott 1992).

Bare-root production systems provide a relatively low cost, low intensity means of producing many large seedlings, that are easy to handle and transport, and well suited to overcome problems associated with competing vegetation and animal and insect damage, once out-planted in the field (Barnett 1978, Wilder-Ayers and Toliver 1987, Menzies and Arnott 1992). Bare-root stock, however is also susceptible to physical damage and desiccation during lifting, packaging, storage, and transport to the field; has a more limited planting season (Menzies and Arnott 1992); and has root systems that often require several months to regenerate roots lost during lifting and pruning (Barnett 1978).

Containerized systems provide planting stock with well protected root systems that are less susceptible to physical damage and desiccation during storage and transport, and transplant shock at the time of out-planting. Containerized stock often performs better on low quality sites and in drier conditions and has an extended planting window as compared to bare-root stock (Stein et al. 1975, Barnett 1978, Bayley and Kietzka 1997, Menzies and Arnott 1992). A disadvantage of containerized production systems is the increased cost associated with the need for containers, multiple growing facilities, and skilled labor required to manage a system demanding greater attention to irrigation, fertilization, and disease and insect monitoring (Stein et al. 1975, Menzies and Arnott 1992).

Transplant production systems incorporate advantages offered by bare-root and containerized systems. Some of these advantages include maximizing nursery bed space by transplanting only germinated seedlings and rooted cuttings, shortening nursery time by optimizing the environmental condition at each stage of development, and controlling root growth by pruning roots prior to transplanting to encourage fibrous root development. Transplant systems may also produce larger plants than can be grown in containerized (Menzies and Arnott 1992) and bare-root systems. One disadvantage of a transplant system is the increased costs of production due to containers and media and the labor required for transplanting. Furthermore, because transplant systems result in bare-root stock, many of the disadvantages associated with root system damage apply.

Morphological characteristics used to evaluate stock quality for these three production systems may differ due to morphological variation among stock types (Wilder-Ayers and Toliver 1987, Barnett 1986) and basic differences in development and cultural regimes (McGilvray and Barnett 1982, Krasowski and Owens 2000). Field evaluation is needed to develop appropriate morphological grading standards for these three stock types.

Seedling Morphological Measurements

Numerous plant characteristics affect stock quality and a number of these characteristics have been used to measure and predict seedling performance. Nursery-grading systems typically measure morphological characteristics to evaluate potential seedling survival and growth (Wakeley 1954, Thompson 1985, Wilder-Ayers and Toliver 1987, Mexal and Landis 1990). Several studies support the predictive value of these morphological measurements for a variety of forest tree species including, Douglas-fir [*Pseudotsuga menziesii* (Mirb.) Franc.], European larch (*Larix decidua* Mill), Norway spruce [*Picea abies* (L.) Karst.] (Mason 1991), slash pine (*Pinus elliottii* Engelm.) (Wakeley 1969), white spruce [*Picea glauca* (Moench) Voss] (Mullin and Svaton 1972), loblolly pine (*Pinus taeda* L.)(Wakeley 1954, Sluder 1979, McGilvray and Barnett 1982), and Sitka spruce [*Picea sitchensis* (Bong.) Carr.] (South and Mason 1993)

Specific morphological grading standards often differ among species, provenance, stock type, and operational requirement (Mexal and Landis 1990). These standards are often defined by field observations and should incorporate the morphological requirements of a seedling with practical planting limitations, such as the size of a root system that can be properly planted with ease (Carlson and Miller 1990). Furthermore, because no single morphological characteristic provides a sufficient assessment of seedling stock quality, a combination of characteristics is often used to provide the best predictive results (Cleary et al. 1978, Menzies and Arnott 1992). Measured morphological characteristics include root collar diameter, shoot height, root mass, root fibrosity, number of first order lateral roots, and shoot to root dry weight ratios.

Seedling Shoot Morphology

Two shoot characteristics, root collar diameter (RCD) and shoot height are often measured to evaluate seedling stock quality. Thought to best integrate the morphological response of a plant to its environment, RCD is commonly used as a standard for sorting and grading reforestation stock. RCD is often positively correlated with survival (Thompson 1985, Carlson and Miller 1990, Mexal and Landis 1990, Dunsworth 1997). For example, larger diameter loblolly pine (Wakeley 1969, Blair and Cech 1974, South and Mexal 1984) and Douglas-fir (Blake et al. 1989) seedlings usually survive at higher rates than their smaller diameter counterparts. Minimum RCD measurements are commonly recommended to ensure high rates of survival for those planted. South and Mexal (1984), for instance, recommended planting southern pine seedlings with diameters > 4 mm to achieve survival rates > 80%. Seedlings with larger diameters also tend to have a greater number of primary lateral roots, increased root growth potential, and greater shoot and root weights. Each of these characteristics has been positively correlated with seedling survival (Thompson 1985, Hatchell 1986, Mexal and Landis 1990). Many nurseries cull seedlings below a minimum diameter, because of their lower potential for survival.

RCD can also affect seedling height and volume growth. In two separate studies, seedlings of loblolly pine with larger initial RCD measurements maintained height advantages over seedlings with smaller initial RCD measurements for the first 5 years of growth (Switzer and Nelson 1967, Dierauf 1993). Similarly, for white pine (*Pinus strobus* L.) (Ward et al. 2000) and sweetgum (*Liquidambar styraciflua* L.) (Kaszkurewicz and Keister 1975), larger initial RCD measurements were positively correlated with height growth after 7 and 4 years, respectively. Greater volume gains have likewise been reported for species of southern pine with larger initial RCD measurements (Wakeley 1969, Blair and Cech 1974, South et al. 1988, Dierauf 1993). A review of several southern pine seedling grade studies reporting long-term volume production concluded that average volume production was 25% greater for seedlings with RCD measurement > 4.8 mm, in contrast to seedlings with RCD measurement between 3.2 - 4.8 mm (South and Mexal 1984).

Shoot height, another commonly measured morphological characteristic, is correlated frequently with field growth. Shoot height is highly correlated with needle number and is, therefore, considered a good measure of photosynthetic capability and transpirational demand. Both of these measurements affect growth (Armson and Sadreika 1974, Thompson 1985, Mexal and Landis 1990, Carlson and Miller 1990). Several studies have indicated that initial height is positively correlated with subsequent height growth (Hunt and Gilmore 1967, McGilvray and Barnett 1982, Melburg and Nashbund 1987, Mexal and Landis 1990). Sitka spruce (South and Mason 1993), loblolly pine (Sluder 1979), and sweetgum (Kaszkurewicz and Keister 1975) seedlings with larger initial heights, for example, grew taller than seedlings with shorter initial heights after 6, 3, and 2 years of growth, respectively. Dierauf (1973), however, found that height had little effect on growth of loblolly pine seedlings if the comparison was among seedlings of the same RCD class. Likewise, at a certain point stem height was negatively correlated with survival for loblolly pine seedlings planted on dry sites (Tuttle et al. 1988). Tall seedlings with small or inefficient root systems are often incapable of acquiring enough water to satisfy daily transpirational demands. On dry sites or during years of drought high mortality rates are common for tall seedlings with inadequate root systems.

Seedling Root Morphology

Root morphology is equally important for determining seedling survival and growth. Several characteristics describing root system morphology have been used to evaluate seedling stock quality, including root mass, root system fibrosity, and the number of first order lateral roots (FOLR) (Cleary et al. 1978, Thompson 1985, Carlson and Miller 1990, Mohammed 1997). Root characteristics such as these strongly affect the capacity for a newly planted seedling to take up water and produce new roots (Carlson and Miller 1990). Seedlings with a greater ability to take up water are more capable of minimizing water stress that can negatively impact seedling performance.

Root mass is a common morphological measurement used to evaluate seedling stock quality (Thompson 1985, Mexal and Landis 1990) and can be expressed in terms of root dry weight, root volume, or root area. Positively correlated with RCD, root mass has been associated with increased seedling survival and growth (Thompson 1985). Blake et al. (1989) found that larger root dry weights provided higher survival rates for Douglas-fir seedlings. Larger root dry weights were also associated with increased height growth (Long and Carrier 1993).

Seedling root dry weight has been positively correlated with root growth potential (RGP) (Carlson and Miller 1990). Larsen et al. (1986) demonstrated that loblolly pine seedlings with greater root dry weights had greater RGP. However, RGP is more often thought of as a function of root architecture rather than absolute size (Mexal and Landis 1990). Root dry weight as a morphological measurement, therefore, often fails to account for differences in root fibrosity, number of FOLR, and other aspects of root architecture. Evidence refuting the relationship between root size and seedling performance also exists. For example, Krasowski and Owens (2000) found that root system size was poorly related to root system hydraulic properties and post-planting growth performance in seedlings of white spruce.

Grading root system fibrosity can be another valuable tool for evaluating seedling stock quality (Deans et al. 1990). Like root mass, the number of fibrous roots is positively correlated with RCD and, therefore, seedling survival and growth (Mexal and Landis 1990). In two separate experiments, an increased number of fibrous roots improved survival for seedlings of lodgepole pine (*Pinus contorta* Dougl. ex Loud.) (Hermann 1964) and Douglas-fir (Burdett 1976). Likewise, field mortality increased for loblolly pine when 50-75% of the fibrous root system was removed from seedlings prior to out-planting (Rowan 1983). Root growth potential has also been positively correlated with root fibrosity. More sites for new root growth are thought to exist for root systems with a greater number of fibrous lateral roots (South 1985, Larsen et al. 1986).

Another measurement of root morphology employed to evaluate seedling stock quality is the number of first order lateral roots (FOLR) (Carlson and Miller 1990). Like root mass and root fibrosity, FOLR is also positively correlated with RCD and, therefore, seedling survival and growth (Mexal and Landis 1990). Increased survival has been associated with increased numbers of FOLR for seedlings of sweetgum (Kormanick et al. 1995), northern red oak (*Quercus rubra* L.) (Kormanick et al. 1995, Thompson and Schultz 1995, Ward et al. 2000), and loblolly pine (Wilder-Ayers and Toliver 1987). Height growth for seedlings of northern red oak was also positively correlated with the number of FOLR. Some researchers, however, have disputed the relative importance of FOLR as a morphological attribute (Teclaw and Isebrands 1991, Dey and Parker 1997). In one study, for example, no correlation was found between the number of FOLR and field performance for white pine (Ward et al. 2000). However, some have incorporated this root system measurement into target seedling criteria. One commercial forestry organization established an acceptable minimum of six first order lateral roots as one of their loblolly pine seedling grading standards (Carlson and Miller 1990).

Seedling Shoot to Root Ratio

Seedling survival is ultimately dependent on the ability of a root system to provide enough water to overcome transpirational loss (Carlson and Miller 1990). Shoot weight to root weight ratio (S: R ratio), a morphological index designed to measure this balance is used commonly to evaluate stock quality (Thompson 1985, Mohammed 1997, Tanaka et al. 1997). Several researchers report a close relationship between loblolly pine seedling survival and S: R ratio measurements (Larsen et al. 1986, Rowan 1987) and some have demonstrated increased survival of out-planted loblolly pine seedlings with lower S: R ratios (Hams and Langdon 1977, Wilder-Ayers and Toliver 1987). A negative correlation between S: R ratio and survival has also been reported for seedlings of ponderosa pine (*Pinus ponderosa* Dougl. Ex Laws) (Lopushinsky and Beebe 1976) and Mexican weeping pine (*Pinus patula* Schiede ex Schlecht. & Cham.) (Bayley and Kietzka 1997).

Some controversy exists regarding the usefulness of S: R ratio as a consistent predictor of seedling survival (Thompson 1985). Factors such as planting site environment, stock type, and nursery culture appear to confound this relationship (South 1985, McGilvray and Barnett 1982, South and Mexal 1984, Barnett and Brisstuckte 1986, Van den Driessche 1991). For example, preferred S: R ratios often change according to the environment of the planting site. When soil moisture is limiting for recently out-planted seedlings, lower S: R ratios are associated with higher survival rates. S: R ratio is less closely related with survival when moisture is plentiful (South and Mexal 1984).

S: R ratio can likewise provide a deceptive picture of seedling morphology. A seedling with a low S: R ratio could posses a large root system, comprised of a large taproot, but lacking any lateral and fibrous roots. Caution has, therefore, been expressed for employing S: R ratio as a sole indicator of seedling survival (Cleary et al. 1978, Menzies and Arnott 1992).

Seedling Morphology and the Planting Environment

The relationship between seedling performance and morphology is strongly influenced by the environmental conditions of the planting site (South 1985). Factors such as available soil moisture, competing vegetation, desiccating wind, and soil temperatures can strongly affect seedling performance (South and Mexal 1984, Carlson 1986, Grossnickle 1988, Stape et al. 2001). Successful reforestation efforts take into account the interaction of morphology and environment when matching planting stock with planting site (Zwolinski et al. 1994). For southern pines, the optimal shoot height and S: R ratio often changes according to the environment of the planting site. For example, taller seedlings typically outperform shorter seedlings on sites with heavy vegetation, while the reverse is true in droughty conditions (South and Mexal 1984**).**

Seedling Nursery Bed Density

The density at which seedlings are grown in the nursery can significantly affect seedling morphology and, therefore, resulting stock quality (Shipman 1966, Wynia and McCain 1981). In general, seedlings produced at wider spacings have larger stem diameters, greater shoot and root dry weights, and greater uniformity than seedlings produced at closer spacings. Also, seedlings grown at lower densities often have greater heights and more balanced S: R ratios (Mullin and Bowdery 1977a, Duryea 1984, Van den Dreissche 1984a). These advantages can result in lower cull rates and greater ease of grading (Duryea 1984). Improved field performance is also common for out-planted seedlings grown at lower densities (Mullin and Bowdery 1977a, Duryea 1984).

Seedlings of red pine (*Pinus resinosa* Ait.) (Mullin and Bowdery 1978), longleaf pine (*Pinus palustris* Mill.) (Scarbrough and Allen 1954, Derr 1955), and black spruce [*Picea mariana* (Mill.) B.S.P.] (Wynia and McCain 1981) had greater dry weights and larger root collar diameters when grown at lower densities. Similarly, loblolly pine, slash pine (Shoulders 1960), and sweetgum (South 1975) seedlings were shown to have larger root collar diameters when grown at lower seedbed densities. Based on root collar diameter standards of Wakeley (1954), cull rates for the same loblolly and slash pine seedlings (Shoulders 1960) were inversely correlated with seedling density. Seedlings grown at a density of $93 \cdot m^{-2}$ produced 90% acceptable seedlings, 70% of which were categorized as Grade 1. In contrast, seedlings grown at $372 \cdot m^{-2}$ produced 60-75% acceptable seedlings, only 50% of which were categorized as Grade 1. Cull rates also decreased as density decreased for seedlings of longleaf pine (Scarbrough and Allen 1954).

S: R ratio and height have also been compared among tree seedlings grown at various densities. Armson and Sadreika (1968) and Mullin and Bowdery (1977b) both found that seedlings of white spruce had lower S: R ratios when grown at lower densities. A positive correlation between S: R ratio and seedbed density was also reported for seedlings of white pine (Mullin and Bowdery 1977a). Height differences, however, were less predictable in relation to seedling densities. Wynia and McCain (1981) demonstrated that seedling height of black spruce increased with decreasing density. In contrast, Shoulders (1960) found no difference in height among seedlings of loblolly and slash pine grown at various densities. Shipman (1966) demonstrated that seedlings of loblolly pine grown at densities of $465 \cdot m^{-2}$ and $557 \cdot m^{-2}$ were taller than seedlings grown at $186 \cdot m^{-2}$, $279 \cdot m^{-2}$, or $372 \cdot m^{-2}$.

The morphological impact of seedbed density on seedling growth in the nursery also appears to affect seedling field performance. Increased survival for seedlings grown at lower densities has been reported for a number of species, including longleaf pine (Scarbrough and Allen 1954, Derr 1955), red pine (Mullin and Bowdery 1978), white spruce and Sitka spruce (Van den Dreissche 1984b). However, in many cases, survival can be as dependent on the availability of moisture following out-planting, as the morphological impact of lower seedbed density (Van den Dreissche 1984b). For example, Shoulders (1960) and Shipman (1966) reported that slash and loblolly pine seedlings grown at low densities survived at higher rates when rainfall was below average. However, when rainfall was adequate, there was no difference among seedlings grown at different densities

On the other hand, superior field growth almost always appears to be associated with lower seedbed density. An inverse relationship between seedbed density and height growth has been reported for many species including white pine (Mullin and Bowdery 1977a), longleaf pine (Scarbrough and Allen 1954, Derr 1955), slash pine, loblolly pine (Shipman 1966), red pine (Mullin and Bowdery 1978), white spruce (Mullin and Bowdery 1977b), and radiata pine (*Pinus radiata* D. Don) (Balneaves and McCord 1976, Bowles 1981).

Seedling Physiological Measurements

Morphological characteristics do not account for all of the variation observed in seedling performance and morphological grading often fails to eliminate seedlings with a low capacity for survival (Wakeley 1954, Thompson 1985, Mexal and Landis 1990, Dunsworth 1997). Seedling stock quality is dependent on both morphological and physiological characteristics (Ritchie 1984). A newly planted seedling will only be able to overcome planting stresses if its physiological processes function adequately to ensure morphological development (Grossnickle and Folk 1993). In other words, the viability and vigor of a seedling are directly related to its physiological status (Mexal and Landis 1990, Sampson 1997). For this reason, two morphologically similar plants with different physiological characteristics may perform very differently when planted in similar environments.

Factors affecting the physiological status of a seedling include genotype, production system, nursery culture, and lifting and storage practices (South and Mexal 1984, Mexal and Landis 1990, Mason 1991). These factors impact plant nutrition, carbohydrate reserves, hydration, and the capacity to cope with pests and stress. The response of a sweetgum seedling to droughty conditions, for instance, is affected by genotypic variation within the species. When sweetgum seedlings were exposed to moisture stress, drought tolerant genotypes allocated more dry weight to the roots and less to the leaves (Elhert 1989). Likewise, sweetgum genotypes that exhibited greater stomatal insensitivity during moisture stress provided continued gas exchange and, as a result, had faster, longer, and more efficient growth during droughty conditions (Elhert 1989).

An additional factor affecting the physiological status of seedlings is the density at which they are grown in the nursery (Shipman 1966). When all other requirements are optimized, lower densities provide emerging seedlings with greater access to mineral nutrients, moisture, and light (Van den Dreissche 1984a). As a result, seedlings grow larger and produce more leaf area, allowing for greater photosynthetic capability (Van den Dreissche 1984b, Duryea 1984) and an associated increase in stored carbohydrate reserves. A larger store of reserves may promote more vigorous growth once the seedling is planted in the field (Duryea 1984).

Much research has been conducted to create methods for defining the physiological attributes of seedlings. Though techniques for measuring root growth potential, seedling viability, cold hardiness, drought tolerance, and several other physiologically based indicators of stock quality have been developed, none have provided a single predictor of stock quality (Mexal and Landis 1990, Tanaka et al. 1997). In addition to their predictive limitations, existing physiological tests are expensive, difficult to conduct and interpret, and time consuming (Mason 1991, Mohammed 1997). For these reasons, physiological testing has received limited acceptance for operational application (Mohammed 1997).

Some organizations in the United States, Canada, United Kingdom, and Sweden have developed methods for evaluating stock quality that integrate morphological and physiological characteristics (Dunsworth 1997, Mohammed 1997). For example, since 1985, one commercial forestry organization has incorporated measurements of shoot height, RCD, and S: R ratio with the evaluation of RGP, seedling viability, cold hardiness, and disease screening to provide standards with which to cull seedlings of inferior stock quality (Tanaka et al. 1997). This method of stock quality evaluation and others like it have proven beneficial for improving cultural techniques in the nursery, testing viability, improving plantation success on high risk sites, and reducing costs associated with replanting after unsuccessful attempts at reforestation. However, morphological grading is still currently the most common method for evaluating stock quality in forest nurseries around the world (Thompson 1985, Mexal and Landis 1990, Dunsworth 1997, Mohammed 1997). Along with improving uniformity and performance of planting stock, morphological measurements can be of great comparative value when the physiological status of seedlings is equal (Thompson 1985, Mexal and Landis 1990, Mason 1991, Dunsworth 1997).

Rooted Cuttings

Rooted Cutting Grading Standards

Developing specific grading standards for rooted stem cuttings (rooted cuttings) are critical for successful field performance, and a topic of increasing interest for clonal forestry in some species (Frampton et al. 2002). Like seedlings, rooted cuttings must conform to certain morphological and physiological standards to ensure successful field performance. However, grading standards for rooted cuttings may differ from those applied to seedlings, because of potential biological differences and increased production costs for rooted cuttings (Ritchie et al. 1993, Puttonen 1997).

Several studies have evaluated potential rooted cutting grading standards for a number of conifer species including eastern white pine (Struve et al. 1984), western hemlock [*Tsuga heterophylla* (Raf.) Sarg.] (Foster et al. 1985), Douglas-fir (Ritchie 1989) and loblolly pine (Goldfarb et al. 1998, Frampton et al. 1999, Foster et al. 2000, Frampton et al. 2002). Shoot growth was positively correlated with root number for the two studies involving container grown rooted cuttings of eastern white pine (Struve et al. 1984) and western hemlock (Foster et al. 1985). On the other hand, results for container-grown loblolly pine rooted cutting studies were more varied. Foster et al. (2000) found root number was a weak but significant predictor of field height and diameter for rooted cuttings of loblolly pine after 5 years of growth. When root number was increased from one to five in their study, rooted cutting shoot height increased by 7 % and diameter growth increased by 12 %. In contrast, after 1 year in the field, Goldfarb et al. (1998) reported that for loblolly pine rooted cuttings, root number and root symmetry were not significantly correlated with height growth. They suggested that another measure of root morphology, such as root mass, was probably more closely related to shoot growth (Goldfarb et al. 1998).

In another loblolly pine study, root dry weight was associated with increased survival for rooted cuttings, when out-planted under dry field conditions (Frampton et al. 2002). In that study, seeds and stem cuttings were propagated in an outdoor nursery bed. At the time of lifting, the rooted cuttings had root dry weights twice that of the seedlings. After 5 years, the rooted cuttings survived at considerably higher rates than the seedlings (77% versus 42%). Dry conditions immediately following out-planting negatively affected seedling reforestation efforts in the surrounding area of the test site and may have adversely affected survival of the seedlings and rooted cuttings at the test site as well. Frampton et al. (2002) concluded that the larger root systems of the rooted cuttings likely contributed to their higher rates of survival.

In the same study (Frampton et al. 2002), rooted cuttings of loblolly pine with root systems rated at planting as poor, fair, and good were compared. Results suggested that rooted cuttings with only one or a few poorly branched roots should be culled to enhance survival during dry years. Additional recommendations for improving the stock quality of rooted cuttings of loblolly pine include: 1) culling rooted cuttings with reduced foliage coverage; 2) culling or pruning rooted cuttings with multiple leaders to reduce forked trees in the field; and 3) culling smaller caliper rooted cuttings with poor and fair root quality to potentially increase volume production (Frampton et al. 2002). A Douglas-fir field study similarly demonstrated superior performance for rooted cuttings with fair to good root systems as compared to those with poor root systems within the same diameter class (Ritchie et al. 1993).

Rooted Cutting Planting Stock

For many tree species, rooting cuttings provides an alternative means of producing planting stock to that of conventional seedling propagation. Examples of forest tree species operationally deployed as rooted cuttings include Japanese sugi (*Cryptomeria japonica* D. Don), Australian flooded gum (*Eucalyptus grandis* Hill ex. Maid.), Norway spruce, radiata pine and eastern cottonwood (*Populus deltoids* Bartr.) (Frampton and Hodges 1989). Use of rooted cuttings in forestry has many potential advantages for both research and operational applications including: 1) genetic evaluation of plant material; 2) reduction of genetic variability in non-genetic studies (Zobel and Talbert 1984); 3) greater capture of genetic gain by exploiting non-additive as well as additive genetic affects; 4) greater stand uniformity; 5) matching genotypes to specific sites (Frampton and Hodges 1989); and 6) providing a platform from which genetically modified plants can be propagated (Hartmann et al. 2002). To realize these benefits, it is important that high quality rooted cuttings are produced to enable field performance at least on par with seedlings of the same species and similar provenance.

Field Performance: Seedlings versus Rooted Cuttings

Many studies have compared field performance of seedlings and rooted cuttings of the same species. Some studies report greater survival and height growth for seedlings (Sweet and Wells 1974, Struve and McKeand 1990), while others report greater survival (Frampton et al. 2002) and height growth (Fielding 1970, Struve et al. 1984) for rooted cuttings. Furthermore, some studies have reported adult phase growth characteristics for rooted cuttings including; reduced taper, fewer growth cycles, smaller limbs, thinner bark, and sparser branching patterns (Libby and Hood 1976, Zobel and Talbert 1984, Foster et al. 1987, Foster 1988, Struve and McKeand 1990, Frampton et al. 2000). These varied results and observations are largely attributed to genetic potential, the age of the donor plant, morphology and vigor of the rooted cutting root system, and the handling of plant material prior to out-planting (Zobel and Talbert 1984, Foster et al. 1987, Ritchie et al. 1993, Stelzer et al. 1998).

Cuttings collected from older more mature trees commonly exhibit reduced growth rates. Foster et al. (1987) demonstrated that tree age or maturation had a negative effect on the growth of loblolly pine rooted cuttings as compared to seedlings. However, when stem cuttings of loblolly pine were collected from young trees no difference was detected for height, diameter, or volume after 5 (Foster et al. 1987) and 10 (Foster 1988, Stelzer et al. 1998) years of growth. Similar growth trends for rooted cuttings originating from various aged stock plants have been demonstrated for rooted cuttings of slash pine (Franklin 1969), radiata pine (Sweet and Wells 1974), Sitka spruce (Roulund 1981) and Douglas-fir (Ritchie et al. 1992).

Root system morphology often varies between seedlings and cuttings of the same species. For stem cuttings of several species, adventitious roots originate from within the stem of the cutting, whereas seedlings typically develop a single taproot with several lateral roots branching off of it (Goldfarb et al. 1998, Frampton et al.

1999, Frampton et al. 2002). Despite the difference in root form, several studies have demonstrated nearly identical performances of seedlings and rooted cuttings. Mason and Gill (1984), for example, reported similar field performance for normal seedling transplants and good quality rooted cuttings of Sitka spruce. Likewise, Douglas-fir seedlings and rooted cuttings with the same diameters and root system quality performed similarly (Ritchie et al. 1993).

Field performance, however, is not only dependent on the morphological characteristics of a plant but also its physiological characteristics (Ritchie 1984, Mexal and Landis 1990, Sampson 1997). Difference in vigor between seedlings and rooted cuttings can differentially affect the field performance of each stock type. When root growth of yellow-cedar [*Chamaecyparis nootkatensis* (D. Don) Spach] seedlings and rooted cuttings with the same initial root area were compared, the seedlings produced more than twice as much new root area as the rooted cuttings after 21 days (Grossnickle and Russel 1990). Perhaps the seedlings in that study produced root systems with greater root growth capacity than the rooted cuttings due to differences in physiological quality of the two planting stock types. New root development after field planting is crucial for seedling establishment (Stone and Schubert 1959, Burdett et al. 1983, Burdett 1987), and is often used as a measure of physiological health.

Study Justification and Outline

Forest planting stock must be capable of high rates of survival and exhibit good field performance to justify the expense of reforestation efforts. Seedling stock quality grading standards have improved the quality of forest planting stock and have increased expectations for survival and field growth of out-planted forest seedlings.

For many tree species, rooting stem cuttings provides an alternative means of producing planting stock to that of conventional seedling propagation. Use of rooted cuttings in forestry has many potential advantages for both research and operational applications. However, to realize these benefits, it is important that high quality rooted cuttings are produced to enable field performance on par with seedlings of the same species and similar provenance.

Developing specific grading standards for rooted cutting planting stock, therefore, is critical for successful field performance, and a topic of increasing interest for clonal forestry in some species (Frampton et al. 2002). Like seedlings, rooted cuttings must conform to certain morphological and physiological standards to ensure successful field performance. However, the grading standards for rooted cuttings may differ from those applied to seedlings, because of potential biological differences and increased production costs for rooted cuttings (Ritchie et al. 1993, Puttonen 1997). Furthermore, various production systems exist capable of producing high quality rooted cutting forest planting stock. Each production system may produce very different planting stock and planting schedules suitable for very different planting conditions and competition factors. Therefore, research is needed to strengthen the current literature concerning rooted cutting production system effects on subsequent stock quality. With this information rooted cutting grading standards can begin to be developed for a number of forest species.

Loblolly pine and sweetgum, to a smaller degree, are two commonly out-planted forest tree species in the southeastern United States. Seedling grading standards exist for both species in this region and are supported by many years of research, especially for loblolly pine. Increased interest in clonal propagation of these two species in the past 2 decades has stimulated research in this area. Currently, production and deployment of rooted cuttings of loblolly pine is not yet fully operational (Goldfarb et al. 1998), but research is currently being conducted by industrial, governmental, and academic organizations (Goldfarb 1997, Steltzer and Goldfarb 1997, LeBude et al. 2004), and several commercial forestry organizations are also currently pursuing pilot-scale rooted cutting production (Frampton et al. 2000). Rooted cutting research of sweetgum has developed successful small scale rooted cutting production systems. Currently, however, there are few sweetgum rooted cuttings currently out-planted.

The greatest obstacle for producing loblolly pine rooted cuttings is the cost of production. In a region where more than 1.2 billion genetically improved loblolly pine seedlings are planted annually and grown in intensively managed forest plantations (McKeand et al. 2003), labor intensive rooted cutting practices are much less efficient than the almost fully mechanized seedling production systems. For rooted cuttings to be competitive, they must first be adaptable to current seedling production systems. Along with attempts to mechanize portions of rooted cutting production, rooted cutting research is needed to evaluate the feasibility and resulting stock quality of current loblolly pine seedling production systems adapted for rooted cutting production. Though the majority of loblolly pine seedlings are produced as 1-

year-old, bare-root seedlings, loblolly pine seedlings are also produced in 1 year as containerized planting stock and as transplant stock. Transplant stock is propagated initially in plugs or containers in a greenhouse and then transplanted outdoors to a nursery bed for subsequent growth. Morphological comparison of loblolly pine rooted cutting planting stock from these three production systems would offer valuable information concerning production system effects on rooted cutting stock quality. Accompanied by stock quality field tests, rooted cutting stock quality grading standards could begin to be established for loblolly pine.

Rooted cutting production of sweetgum must also overcome the obstacle of cost. Besides the disadvantage of increased labor costs, semi-hardwood (SH) stem cuttings of sweetgum require 2 years of nursery growth to match the stock quality of 1-year-old sweetgum seedlings. The inability to achieve adequate shoot and root growth in the same season as rooting adds an additional year to the sweetgum rooted cutting production cycle and, therefore, increases costs. Production systems capable of producing high quality planting stock in one year are needed for sweetgum rooted cuttings to be a viable reforestation option.

Sweetgum seedlings can be produced in the same three seedling production systems as for loblolly pine. Sweetgum rooted cutting studies are needed to investigate the potential of these production systems for SH stem cuttings. Hardwood (H) sweetgum stem cuttings, reportedly, have the ability to produce shoot growth in the same season as rooting. Evaluating sweetgum rooted cutting production system utilizing H stem cuttings may also provide new opportunities for sweetgum rooted cutting production.

Two studies investigating rooted cutting production systems for loblolly pine and sweetgum are described in the following two chapters. The effects of three production systems on morphological characteristics of rooted cuttings of loblolly pine were evaluated in the first chapter. Morphological comparisons were made among the various stock types. In the second chapter, the feasibility and morphological effect of potential sweetgum rooted cutting production systems were evaluated. SH stem cuttings were evaluated in all three production systems with a special emphasis on the presence of new shoot growth following rooting. H stem cuttings were also rooted in a direct-stick system in an outdoor nursery bed to test the ability of this system to produce rooted cuttings exhibiting new shoot growth in the same season as rooting.

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Production system influences survival and morphology of rooted stem cuttings of loblolly pine (*Pinus taeda* L.)

Matthew H. Gocke[1,] Barry Goldfarb[1], Daniel J. Robison[1], L. J. Frampton[1], and Frank Blazich[2]

[1]Dept. of Forestry and Environmental Resources, NC State University

[2]Dept. of Horticultural Science, NC State University

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Email address: mhgocke@gmail.com

Abstract

Two nursery experiments conducted during 2 consecutive years evaluated the effects of production system on survival, and morphology of rooted stem cuttings (rooted cuttings) of loblolly pine. During these two experiments, dormant hardwood stem cuttings of loblolly pine, representing four clones, were rooted and then grown in one of three production systems: in outdoor beds as bare-root stock (DS); in containers as containerized stock (CT); or rooted in containers and then transplanted to an outdoor nursery bed as transplanted bare-root stock (TP). In the first experiment, treatments within a production system were also designed to evaluate additional factors affecting stock quality including; mist duration and the use of shade during the first 16 weeks of rooting for the DS system and two plug types, Jiffy™ Forestry Peat Pellets (JF) and Grow-Tech™ Rooting Sponges (GT), and three transplant times for the TP system. During the second experiment, two mist levels and the effect of shade were evaluated for the DS production system. The potential for two production cycles within one growing season, February versus May sticking dates, was also evaluated for the CT and TP systems. Rooted cutting morphology varied among clone, production system, and treatment within a production system during both experiments. All production systems and clones tested produced acceptable reforestation stock, including the second production cycle (May sticking) cuttings produced in the CT and TP systems, during the second experiment.

In both experiments, stem cuttings rooted in the GT-TP system produced planting stock with the largest shoot height, shoot dry weight, root collar diameter, root dry weight, and shoot: root ratio. For both the GT and JF transplant production systems, earlier transplant times produced larger size rooted cuttings, while later transplant times resulted in greater survival rates. The TP cuttings rooted in higher percentages and grew to a larger size in the GT rooting sponges than in the JF peat pellets. DS system rooted cuttings receiving a greater duration of mist had greater shoot height, shoot dry weight, and root dry weight. Shade had a minimal effect on DS system survival and morphology. In the second experiment, over 90% of the rooted cuttings produced in the production systems tested attained acceptable seedling grading standards. These results demonstrated that the three rooted cutting production systems evaluated are capable of producing high quality planting stock and that two full production cycles can be obtained in one growing season for the CT and TP systems. Installation of field trials designed to evaluate survival and field performance of rooted cuttings of loblolly pine representing these stock types will permit development of individual stock type grading standards. However, results of these field studies may indicate that new grading criteria are only necessary for containerized and bare-root rooted cutting stock.

Introduction

More than 1.2 billion genetically improved loblolly pine (*Pinus taeda* L.) seedlings are planted annually and grown in forest plantations in the southeastern United States (McKeand et al. 2003). However, there is interest in using vegetative propagation to further improve the genetic quality of loblolly pine planting stock. Production and deployment of loblolly pine rooted stem cuttings (rooted cuttings) is not yet fully operational (Goldfarb et al. 1998). Rooted cutting research is currently being conducted by industrial, governmental, and academic organizations (Goldfarb 1997, Steltzer and Goldfarb 1997, LeBude et al. 2004), and several commercial forestry organizations are also currently pursuing pilot-scale rooted cutting production (Frampton et al. 2000).

One area of research necessary for successful operational deployment of loblolly pine rooted cuttings is identification of production systems capable of producing high quality planting stock. This planting stock must be capable of good initial field survival and growth to ensure the genetic potential of each plant is realized (Carniero 1995, Frampton et al. 2000).

Several production systems have been developed to provide a variety of stock types suitable for reforestation. Plants cultured as seedlings, rooted stem cuttings, *in vitro* plantlets, and seedlings derived from somatic embryos are typically grown under one of three production systems: in outdoor nursery beds as bare-root stock; in containers as containerized stock; or initially as containerized stock and then transplanted to outdoor nursery beds as bare-rooted transplants (Stein et al. 1975, Hahn 1984, Menzies and Arnott 1992). Each of these three production systems provides unique advantages and disadvantages for reforestation efforts (Menzies and Arnott 1992). Stock quality for these three production systems may differ due to morphological variation (Wilder-Ayers and Toliver 1987, Barnett and Brisstuckte 1986) and basic differences in development and cultural regimes (McGilvray and Barnett 1982, Krasowski and Owens 2000) among stock types.

Numerous morphological characteristics affect stock quality and several of these characteristics have been used to establish grading criteria useful for predicting seedling performance of a variety of forest tree species (Wakeley 1954, Thompson 1985, Wilder-Ayers and Toliver 1987, Mexal and Landis 1990) including Douglas-fir [*Pseudotsuga menziesii* (Mirb.) Franc.] (Mason 1991) and loblolly pine (Wakeley 1954, Sluder 1979, McGilvray and Barnett 1982, Rowan 1987). Specific morphological grading standards for tree seedlings often differ among species, provenance, stock type, and operational requirement (Mexal and Landis 1990). Because no single morphological characteristic provides a sufficient assessment of seedling stock quality, a combination of characteristics is often used to provide the best predictive results (Cleary et al. 1978, Menzies and Arnott 1992).

Root collar diameter (RCD) and shoot height are two shoot morphological characteristics often measured to evaluate seedling stock quality. RCD is commonly used as a standard for sorting and grading reforestation stock and is often positively correlated with seedling survival (South and Mexal 1984, Thompson 1985, Carlson and Miller 1990, Mexal and Landis 1990, Dunsworth 1997). Seedlings with larger RCDs also tend to have a greater number of primary lateral roots, increased root growth potential, and greater shoot and root mass; each of which have been positively correlated with seedling survival (Thompson 1985, Hatchell 1986, Mexal and Landis 1990). RCD is also often positively associated with seedling height (Switzer and Nelson 1967, Dierauf 1993) and volume growth (Wakeley 1969, Blair and Cech 1974, South and Mexal 1984, South et al. 1988, and Dierauf 1993). Initial shoot height is frequently positively correlated with field growth (Hunt and Gilmore 1967, McGilvray and Barnett 1982).

Morphological characteristics of root systems used to evaluate seedling stock quality include measurements of root mass, such as root dry weight, and the number of first order lateral roots (FOLR) (Cleary et al. 1978, Thompson 1985, Carlson and Miller 1990, Mohammed 1997). Root dry weight is positively correlated with RCD and has, therefore, been associated with increased seedling survival and growth (Thompson 1985, Blake et al. 1989, Long and Carrier 1993). Seedling root weight has likewise been positively correlated with root growth potential (Larsen et al. 1986, Carlson and Miller 1990). The number of FOLR has also been positively associated with survival (Wilder-Ayers and Toliver 1987).

Shoot dry weight to root dry weight ratio (S: R ratio), a morphological index designed to measure the ability of a root system to provide enough water to overcome transpirational loss is also used to evaluate stock quality (Thompson 1985, Carlson and Miller 1990, Mohammed 1997, Tanaka et al. 1997). Some researchers report a close relationship between seedling survival and S: R ratio measurements (Larsen et al. 1986, Rowan 1987) and have demonstrated increased survival of seedlings with lower S: R ratios (Hams and Langdon 1977, Wilder-Ayers and Toliver 1987). However, other researchers discredit the usefulness of S: R ratio as a consistent predictor of seedling survival (Thompson 1985). Factors such as planting site environment, stock type, and nursery culture appear to confound this relationship (South 1985, South and Mexal 1984, McGilvray and Barnett 1982, Barnett and Brisstuckte 1986, Van den Driessche 1991).

Current grading strategies for seedlings of loblolly pine often adhere to minimum root collar diameter measurements along with other morphological measurements including shoot height, root mass, root fibrosity, S:R ratio, FOLR, needle length, and the presence of secondary needles (South and Mexal 1984, Thompson 1985, Wilder-Ayers and Toliver 1987, Mexal and Landis 1990).

Less effort has been focused on the morphology of rooted cuttings as related to stock quality. Though seedling grading standards may be applicable to rooted cuttings, potential biological differences and increased production costs may create a need for separate rooted cutting grading standards (Ritchie et al. 1993, Puttonen 1997).

Some studies support the predictive value of root morphology measurements, such as root number (Foster et al. 2000) and root mass (Goldfarb et al. 1998, Frampton et al. 2002) on field performance of rooted cuttings of loblolly pine. Foster et al. (2000) reported that root number was a weak, but significant predictor of field height and diameter breast height (dbh) for rooted cuttings of loblolly pine after 5 years of growth (Foster et al. 2000). In contrast, after 1 year in the field, Goldfarb et al. (1998) reported that field shoot height was not significantly correlated with root number, nursery shoot height, or root symmetry for rooted cuttings of loblolly pine and speculated that another measure of root morphology like root mass was probably more important for shoot growth in the field.

Providing additional evidence supporting the potential influence of root mass on field performance, Frampton et al. (2002) reported that larger root dry weights increased survival rates for loblolly pine rooted cuttings when out-planted under dry field conditions. In the same study, rooted cuttings of loblolly pine rated as having poor, fair, and good root systems were compared. Results suggested that rooted cuttings with only one or a few poorly branched roots should be culled to enhance survival during dry years. Additional recommendations for improving stock quality of loblolly pine rooted cuttings included: 1) culling rooted cuttings with reduced foliage area; 2) culling or pruning rooted cuttings with multiple leaders in order to reduce forked trees in the field; and 3) culling smaller caliper rooted cuttings with poor and fair root quality to potentially increase volume production (Frampton et al. 2002).

Study objective

Like loblolly pine seedlings, rooted cuttings of the species can be produced in bare-root, containerized, and transplant production systems. However, additional research is needed to better understand the effect of production system on loblolly pine rooted cutting morphology. Beginning in 2000 a study was conducted to determine the effect of three production systems on morphological characteristics of loblolly pine rooted cuttings. Several treatments were also tested within specific production systems. Four clones were evaluated within these production systems and treatments over a 2 year period.

Materials and Methods

The experiments were conducted at the North Carolina State University Horticultural Field Laboratory, Raleigh, NC. The first experiment was conducted from March to December 2000 and tested the following production systems: 1) a transplant system in which stem cuttings were rooted in a greenhouse in Grow-Tech Rooting Sponges™ (Grow-Tech Inc., San Juan Bautista, CA) and then transplanted at either 7, 9, or 11 weeks to an outdoor nursery bed for subsequent growth (GT); 2) a transplant system in which stem cuttings were rooted in a greenhouse in Jiffy ™ Forestry Peat Pellets (Jiffy Products Ltd., Shippagan, N.B., Canada) and then transplanted at either 7, 9, or 11 weeks to an outdoor nursery bed for subsequent growth (JF); 3) a containerized system, in which stem cuttings were rooted in a greenhouse in Ray Leach SC-10 SuperCells™ (Stuewe and Sons, Inc., Corvallis, OR) for 12 weeks and then transferred outdoors in the same tubes for subsequent growth (CT); and 4) a direct-stick system (DS), in which stem cuttings were stuck directly into an outdoor nursery bed for rooting (in full sun or partial shade) and growth.

The second experiment was conducted from February to December 2001. The production systems tested in the second experiment were the same as those tested in the first experiment, with the following exceptions: 1) the Jiffy peat pellets were not used, 2) two, instead of three, transplant times were evaluated for the GT system; 3) two mist levels and two light levels were tested in a 2 x 2 factorial combination for the DS system; and 4) two sticking dates (February and May) were evaluated for the GT and CT systems. In all, there were five production systems tested in Experiment 2: February GT, February CT, DS, May GT, and May CT.

Rooting and Growth Facilities

A 1.2 m wide x 40 m long raised bed, built with pressure treated lumber, was constructed for use in both experiments. The structure was placed on top of a thin layer of gravel and the inside was lined with weed mat. The bed was filled with soil from the coastal plain of North Carolina with a loamy sand soil texture comprised of 85% sand, 12.2% silt, and 2.8% clay. The nursery bed soil had an initial pH of 5.1 and a pH of 5.5 at the beginning of the second experimental year.

The nursery bed was equipped with overhead NaanDan™ "Water and Sprinkling" nozzles (Kibbutz Naan, Naan, Israel 76829) with a flow rate of 41.6 $L \cdot h^{-1}$. A fixed-cycle control system, a Rainbird ™ Controller Model MIC-12 (Rainbird Corporation, Glendora, CA), controlled the irrigation frequency and duration during the first experiment and was adjusted by manually reprogramming the controller. From April 25 to August 8, 2000, the mist irrigation was applied to the stem cuttings in the outdoor nursery bed, on average, 40 seconds every 10 minutes. The following month, the irrigation regime was slowly reduced to 10 minutes every three days beginning September 9. The irrigation was discontinued October 9 (Appendix I).

For the second experiment, the Rainbird controller was replaced with a variable-cycle control system (Davis Engineering Solar 6A Misting Controller, Davis Engineering, Winnetka, CA). The frequency of irrigation was automatically adjusted according to the amount of sunlight detected by a light sensor connected to the controller. Sunlight was measured in solar units. One solar unit equaled 2,000 foot-candles or 0.02 $mol \cdot m^{-2}$. On a clear sunny day at noon in June, 30 seconds equaled approximately 1 unit, while on a completely overcast day at noon, one unit equaled approximately 3 minutes. No irrigation was applied at night.

The number of solar units programmed between misting intervals increased over time to correspond with the decreasing misting requirements of the stem cuttings as they formed adventitious roots. The irrigation frequency for the DS system was slowly decreased from once every 8 units on April 13 to once every 120 units on July 17, 2001. After July 17, the irrigation regime was further reduced until the rooted cuttings were hand-watered every third day beginning August 8 (Appendix II).

Irrigation for the GT and CT system rooted cuttings in the second experiment was tapered rapidly after transplant or transfer to the nursery bed. These rooted cuttings initially received overhead irrigation once every 32 units for 60 seconds for 4 weeks. After this initial acclimation period, the irrigation was reduced until the CT system cuttings were hand watered every second day and the GT system rooted cuttings were hand watered every third day. The irrigation was discontinued October 1, 2001 (Appendix II). The pH of the nursery bed irrigation water was 5.7 the first year, and 5.9 the second year. Pine bark mulch was also placed on the surface of the nursery bed the second year to retain soil moisture and reduce compaction due to repeated overhead irrigation.

In both experiments, stem cuttings in the GT, JF, and CT systems were rooted in a clear polyethylene-covered greenhouse with ambient light. Heating and cooling systems were stuck to maintain the day/night air temperature at 23-26/20-23^0 C. Stem cuttings were misted intermittently at a variable frequency related inversely to the relative humidity (RH) in the greenhouse. Variable frequencies were defined by designating minimum (60% RH) and maximum (99% RH) off times between misting applications. Off times for intermediate humidity values were calculated based on a linear function. The minimum and maximum off times varied according to the time of day. An environmental management software package (Q-Com, Irvine, CA) calculated mist frequency and triggered a traveling gantry (boom) system (ITS, McConkey, Mt. Puyallup, WA) to apply mist. Misting frequency (number of boom passes) was identical for all treatments within an experiment, but differed (due to environmental conditions) between experimental years. During each experiment, boom traveling speed was altered according to the time during the rooting period. For both experiments, the amount of mist per boom pass was decreased every 4 weeks for the 12 week rooting period. Additional details concerning the frequency and amount of mist applied are summarized in LeBude et al. (2004). The pH of the greenhouse irrigation water was 6.4 during the first experiment, and 6.2 during the second.

Stock plants, cutting material, and rooting procedures

Dormant, hardwood stem cuttings were collected for both experiments from hedged stock plants maintained by the NCSU Loblolly and Slash Pine Rooted Cutting Program. The stock plants originated from 4 clones of loblolly pine, 3 of which were used in each experiment. The clones were derived from individual seedlings from four different full-sib families. Clones A, B, and D originated from seedlings germinated in Spring 1993, while Clone C originated from a seedling germinated in Spring 1996. The seedlings were hedged and subsequent new shoots were collected and rooted to bulk-up the number of stock plants for each clone. Stock plants of clone A, B, and D were rooted initially in February 1997, while stock plants of Clone C were rooted initially in February 1998. Clones A, B, and C were tested in Experiment 1, while clones A, C, and D were tested in Experiment 2.

In both experiments, stem cuttings longer than 8 cm were excised from the stock plants in February and wrapped in moist paper towels in groups of 30. The stem cuttings were then placed into a portable cooler and stored at 5^0 C until sticking. At the time of sticking, the base of each cutting was trimmed to produce an 8 cm stem cutting. The basal 1 cm of the stem cutting was then dipped into a 10 mM solution of 1-naphthalene- acetic acid (NAA) (40% ethanol/60% water) for 3 seconds.

The rooting media, sticking depth, and the stem cutting density used for both experiments varied according to the production system tested (Appendices III & IV). The base of each stem cutting rooted in the GT and JF systems was inserted or "stuck" 1.5 cm into the pre-formed holes of the Grow-Tech rooting sponges or the Jiffy peat pellets. The GT 225 (10 ml) sponges were placed in Winstrip "WS 162" trays (Winstrip, Inc., Fletcher, NC) at a density of 1,259 $\cdot$ m $^{-2}$. The Jiffy 18 mm pellets (15 ml) were prepackaged by the manufacturer into plastic trays at a density of 2,184 $\cdot$ m $^{-2}$. Rooting percentage was evaluated at the time of transplant for these two systems. In Experiment 1, all the GT and JF system stem cuttings (whether rooted or not) were transplanted 7, 9, or 11 weeks after rooting in the greenhouse to the nursery bed. In Experiment 2, rooting percentage was evaluated at the time of transplant and only stem cuttings that had rooted were transplanted to the nursery bed. A stem cutting was considered rooted if at least one root could be observed emerging from the sponge or pellet, or in the case of no visible emerging root, at least one root (> 1 cm) was observed when the sponge or pellet was carefully torn open. The rooted cuttings from the GT and JF systems were transplanted into the nursery bed at a density of 269 $\cdot$ m $^{-2}$. Extra rooted cuttings from the greenhouse comprised the border rows in the nursery bed and were also used to fill any vacancies in treatment plots.

Stem cuttings for the CT system in Experiment 1 were inserted into Ray-Leach Supercells filled with a medium of 1 peat : 1 perlite (v/v). The container medium was modified to 5 peat : 4 perlite : 1 vermiculite (v/v/v) for Experiment 2 to increase the cation exchange capacity (CEC) of the medium. The growing tubes were placed into trays in the greenhouse at a density of 527 $\cdot$ m $^{-2}$. The containers remained in the trays, but the density was reduced to 269 $\cdot$ m $^{-2}$ by placing the growing tubes in every other hole when the cuttings were transferred outside. In Experiment 1, all the containerized stem cuttings (whether rooted or not) were transferred after 12 weeks of rooting in the greenhouse to the nursery bed in spaces without soil. Only rooted cuttings were transferred to the nursery bed in Experiment 2. Rooting percentages were determined by pulling gently on the cutting at the time of the transfer to the nursery bed. The stem cuttings were considered rooted, if the stem cutting remained firmly in the medium.

Stem cuttings stuck in the outdoor nursery bed were inserted half their length, 4 cm, into the nursery bed soil at a density of 269 $\cdot$ m $^{-2}$. PVC structures, supporting shade-cloth (50%), provided shade to the stem cuttings in the shade treatment between 10 a.m. and 4 p.m. during both experiments for the first 16 weeks following sticking.

Fertilizer

The rooted cuttings were fertilized during the study with either a water-soluble all-purpose (20-20-20) (Peters Professional 20-20-20 with micronutrients, J.R. Peters, Inc./J.R. Peters Laboratory, Allentown, PA) or acid (17-6-6) fertilizer (Peter's Acid Greening soluble fertilizer, Scott's Co., Allentown, PA). Medium pH was monitored during Experiment 1 and medium pH and electrical conductivity (EC) were monitored during Experiment 2. During both experiments, acid fertilizer was applied when needed to maintain medium pH below 6.0. In Experiment 1, fertilizer was applied uniformly to all nine treatments tested. From June 11 to July 23, 2000, the rooted cuttings received 50 ppm N/wk. This rate increased to 100 ppm N/wk from July 30 to October 6. The total amount of nitrogen applied during Experiment 1 was 100 kg N/ha (Appendix V).

Fertilizer rates for Experiment 2 were adjusted according to production system. Fertilization for the February GT and CT rooted cuttings began on May 22, the DS rooted cuttings were first fertilized on May 29, and the May GT and CT rooted cuttings were first fertilized on July 3, 2001. Weekly rates of application ranged from 50 – 150 ppm N/wk. The total amount of nitrogen applied during Experiment 2 was 110 kg N/ha for the DS, May GT, and May CT; 155 kg N/ha for the February GT; and 180 kg N/ha for the February CT. EC was maintained between 0.15 and 0.4 during Experiment 2 (Appendix VI).

Other macro- and micronutrients were applied as indicated by periodic foliage nutrient analyses following recommendations for loblolly pine seedlings (C.B. Davey and J.B. Jett, Dept. of Forestry, North Carolina State University, Raleigh, NC, personal communication). The cuttings were treated with chelated iron during both experiments and boron during the second experiment (Appendices V and VI).

Treatments: Experiment 1

A total of nine treatments were tested in Experiment 1. Stem cuttings from the GT, JF, and CT systems were stuck in the greenhouse March 10, while the DS system stem cuttings were stuck outdoors April 25. Rooted cuttings produced under one of six TP system treatments were evaluated for rooting percentage, survival, shoot height, shoot and root dry weight, RCD, primary root number, and S: R ratio. The six treatments evaluated two plug types and three transplant times. An initial group of stem cuttings was transplanted to the nursery bed April 27 (7 weeks), followed by the second group on May 11 (9 weeks), and the third group on May 25 (11 weeks). One CT system treatment (transferred outdoors June 1) and two DS system treatments, testing the effect of shade during rooting, were also evaluated for survival, shoot height, shoot and root dry weight, RCD, primary root number, and S: R ratio.

Treatments: Experiment 2

Ten treatments were tested during the second experiment. Rooted cuttings produced by four TP system treatments were evaluated for rooting percentage, survival, shoot height, shoot and root dry weight, root collar diameter (RCD), primary root number, and shoot to root ratio (S: R ratio). These four treatments evaluated two sticking times, February GT and May GT, and two transplant times. An early set of cuttings was stuck February 22 and a late set was stuck, May 4. Transplant times for the February rooted cuttings were April 26 (9 weeks) and May 17 (12 weeks). For the May rooted cuttings, the first transplant occurred on July 3 (8 weeks) and the second July 24 (10 weeks). Transplant date for the late stick was shortened to prevent further damage to new roots by fungus gnat (*Bradysia coprophila* (Dipt.: Sciaridae) larvae that were observed during rooting.

There were also two sticking time treatments in the CT system, February CT and May CT. The February CT stem cuttings were stuck in the greenhouse February 22 and transferred to the nursery bed after 12 weeks May 17. The May stem cuttings were stuck in the greenhouse May 4 and transferred to the nursery bed July 24. Four DS system treatments were also stuck on April 14 and tested the effects of high (1-minute duration) versus low (30-second duration) misting regimes and full sun versus shade (50%) during the initial 16 week rooting period. The DS system stem cuttings and the early set of GT and CT system stem cuttings tested in Experiment 2 were stuck approximately 2 weeks earlier in the calendar year than similar systems tested in Experiment 1. Additionally, in Experiment 2, the May GT and May CT systems were tested to evaluate the effects of a later production cycle on stock quality.

Rooted cuttings from both experiments were either lifted from the nursery bed or removed from their containers in mid-December to assess RCD, shoot height, and number of primary roots. Survival was determined per plot for each clone tested.

Following these initial measurements, the rooted cuttings were severed at the root collar, placed in paper bags, and dried for 72 h at 70^{0} C. Shoot and root dry weights were then determined for each clone on a plot basis. S: R ratios were calculated by dividing shoot dry weight by root dry weight.

All rooted cuttings in Experiment 1 were measured and destructively harvested in this manner. However, for the second experiment, only six rooted cuttings per plot were selected at random for each clone for destructive harvesting. The remaining rooted cuttings were reserved for a subsequent field test.

Experimental Design

Experiment 1 was arranged in a split-plot design with eight blocks, each containing nine treatments (whole-plots) randomly assigned within each block. The sub-plots were three rows of 10 stem cuttings, each row containing one of the three clones (subplots). GT, JF, and CT system treatments originated in the greenhouse and were arranged in the same split-plot design across the length of two benches. The same block structure was maintained when the rooted stem cuttings were moved to the outdoor nursery bed. The design of the second experiment was similar to the first, with the exception that there were six blocks, 10 treatments, and 15 stem cuttings per clone for each treatment within a replication.

Physical refinements were made to the design of the nursery bed during the second experiment to more successfully separate individual treatment plots. During the first experiment, irrigation intended for one plot could be carried by the wind onto adjacent plots. Clear plastic sheets (4 mil) were placed in between plots to block irrigation drift in Experiment 2. The reduction in the number of replications in the second experiment, also allowed for greater spacing among treatment plots. Finally, the height of the DS system shade cloth frames was lowered and, with the increased spacing, the early morning and late afternoon shading of adjacent treatment plots was eliminated.

Statistical Analysis

Analyses of variance for the traits assessed in both experiments were conducted on plot means using the Mixed Model Procedure in SAS (SAS, Inc. Cary, NC). Sources of variation used in the model were either analyzed as fixed effects (system, clone, system x clone, treatment within system, and clone x treatment within system) or as random effects (block), and were considered significant when the probability of a greater F-value was ≤ 0.05. Least squares means were reported in both experiments due to unbalanced data.

In Experiment 1, the arcsine square root transformation was used for survival, while in both experiments, the log transformation was used for shoot height, shoot dry weight, RCD, and root dry weight, and a square root transformation was used for the number of primary roots. Non-transformed mean values were reported. Pair-wise comparisons and pre-planned contrasts were used to determine significance among means.

Results

Rooting Percentages and Survival Rates

Experiment 1

The overall survival rate for the loblolly pine rooted cuttings produced in Experiment 1 was 58%. Rooting percentage, independent of survival, was not determined during the first experiment. Clonal survival varied according to production system and resulted in a significant system by clone interaction (Table 1.1). Clone C had the highest survival ranking for each of the production systems tested, ranging from 56.4% to 81.8%. Clone A did not differ from the other two clones in the GT system, but was among the lowest ranked clones in the other three systems (Figure 1.1).

Significant survival differences were also detected among the transplant treatments within the GT and JF production systems. Transplanting cuttings at 11 weeks increased survival rates by 17% and 20% for the GT and JF systems, respectively, as compared with transplanting at 7 weeks (Figure 2.1). Rooted cuttings produced in the DS system survived at 62% (Figure1.1).

Experiment 2

Overall rooting percentage and survival for the loblolly pine stem cuttings stuck in Experiment 2 were 55% and 71%, respectively (Figure 3.1). Survival rates for the DS system stem cuttings were considered a measure of rooting percentage and were, therefore, identical. Rooting percentages and survival rates for each clone varied according to production system and resulted in a significant system by clone interaction (Table 2.1). Rooting percentages (65-93%) and survival rates (89-100%) of clone D ranked highest for the February and May GT and CT production systems, while Clone C ranked highest in rooting percentage and survival (66.7%) for the DS system (Figure 3.1). In general, Clones C and D performed relatively well within each production system. In contrast, rooting percentages for Clone A varied substantially among production systems. For example, rooting decreased from 62.0% (February GT) to 20.0% (February CT) and 24.0% (DS) for cuttings of Clone A. Across clones there was a significant difference in rooting percentage between the February and May GT systems (Table 3.1). Rooting percentage for the May GT stem cuttings was adversely affected by an infestation of fungus gnat larvae.

Rooting percentage and survival also varied for each clone according to treatment within a system (Table 2.1). For the six treatments tested within the February and May GT and CT systems, Clone D ranked among the highest for rooting percentage and survival (Figure 4.1). Meanwhile, for the four treatments tested within the DS production system, Clone C had the highest rooting percentage and survival ranking for the HW/S, HW/SH, and LW/SH treatments.

Shoot Morphology

Experiment 1

Production system significantly affected shoot morphology of the rooted cuttings produced in Experiment 1 (Table 1.1). Rooted cuttings produced within the GT system had significantly greater shoot height, shoot dry weight, and RCD than the other three systems (Table 4.1). Rooted cuttings produced with the JF system also had significantly greater shoot height than the CT or DS systems and significantly greater shoot dry weight and RCD than rooted cuttings produced with the DS system. The CT system produced rooted cuttings with greater shoot height and shoot dry weight than the DS production system.

Clones differed significantly for shoot height, shoot dry weight, and RCD (Table 1.1). Clone C produced rooted cuttings with the greatest shoot height (22 cm) and shoot dry weight (33 g), while Clones A (4.4 mm) and C (4.3 mm) had larger RCDs than Clone B (3.8 mm) (Table 5.1).

Shoot height and RCD for each clone also varied among treatment within a production system (Table 1.1). Among the GT and JF production system treatments, Clone C produced the tallest rooted cuttings with the largest RCDs when transplanted at 7 and 11 weeks. Clone A ranked highest for both traits at 9 weeks (Figure 5.1).

Experiment 2

Shoot height, shoot dry weight, and RCD for each clone varied according to production system (Table 2.1). Clone D produced the tallest rooted cuttings (18.5 – 47.7 cm) in the February and May GT and CT systems, while Clone C produced the tallest rooted cuttings (27.6 cm) in the DS system (Figure 3.1). Clone D had the greatest shoot dry weight in the February GT system (79.5 g), while Clone C ranked highest for all of the other systems tested (18 –74 g). Clone C also ranked highest for RCD in the February GT (7.1 mm), February CT (5.4 mm), and DS (6 mm) systems, while clone A ranked highest in the May GT (5.6 mm) and CT (4.9 mm) systems (Figure 3.1).

Significant differences in shoot dry weight were detected among individual treatments tested within production systems (Table 2.1). Transplant time, within the May GT system, significantly affected shoot dry weight (Table 6.1). The rooted cuttings transplanted at 8 weeks had significantly larger shoot dry weight (23.2 g) than those transplanted at 10 weeks (17 g). Similarly, within the DS system, stem cuttings rooted in the HW/S treatment had significantly larger shoot dry weight (26.3 g) than the stem cuttings rooted in the LW/S treatment (18.3 g) (Table 6.1).

There were differences in shoot height and shoot dry weight between the combined HW and LW-DS system treatments, the February and May GT system, and the February and May CT system (Table 3.1). Stem cuttings rooted in the DS system under HW were significantly taller (24.7 vs. 21.6 cm) and had larger shoot dry weight (26.9 vs. 19.2 g) than those rooted in the DS system under LW. The February GT and CT systems produced taller rooted cuttings with greater shoot dry weight than their respective May counterparts. The February GT system also produced rooted cuttings with greater RCD (7 mm) than the May GT system (5.3 mm).

Root Morphology

Experiment 1

Production system significantly affected the root dry weight of loblolly pine rooted cuttings produced in Experiment 1 (Table 1.1). The GT (13.7 g) and CT (9.3 g) systems produced rooted cuttings with significantly larger root dry weight than the JF (6.7 g) and the DS (3.8 g) systems (Table 4.1). The JF system rooted cuttings had significantly larger root dry weight than those produced in the DS system.

Significant differences in root dry weight also existed among the three clones (Table 1.1). Clone C produced rooted cuttings with significantly larger root dry weights (10.5 g) than Clones A (8.0 g) or B (6.6 g) (Table 5.1). Primary root number for each clone varied according to production system (Table 1.1). Clone C produced the greatest number of roots for the JF (3.3) and DS (2.5) systems, while Clone A produced the greatest number for the CT (2.8) system (Figure 1.1). Clones A and C both produced the greatest number of roots for the GT (3.5) system.

Experiment 2

The number of primary roots and root dry weight for each clone varied according to production system (Table 2.1). Clone D produced rooted cuttings with the greatest number of primary roots in the February GT (4.7), February CT (4), and May CT (6.7) systems, while Clone A ranked highest in the May GT (4.8) system. Clone C ranked highest for the DS (2.6) system (Figure 3.1). For root dry weight, Clone D had the highest rank for the February GT (28.8 g), February CT (13.5 g), May GT (8.8 g), and May CT (11.8 g) production systems, while rooted cuttings of Clone C ranked highest in the DS (11.2 g) system.

Treatments within a system significantly affected root dry weight (Table 2.1). In the DS system, the HW/SH treatment produced rooted cuttings with significantly greater root dry weights (9.5 vs. 6.3 g) than the LW/S treatment (Table 6.1). Furthermore, contrasts between the HW and LW treatments revealed significant differences for root dry weight. Stem cuttings rooted under the HW treatments had significantly greater root dry weight than those rooted under the LW treatments (9.3 vs. 7.1 g, respectively) (Table 6.1). GT system contrasts demonstrated that the February GT system produced rooted cuttings with significantly greater root dry weight then rooted cuttings from the May GT system (Table 6.1).

Shoot to Root Ratio

Experiment 1

Production system and clone significantly affected the S: R ratio of loblolly pine rooted cuttings produced in Experiment 1 (Table 1.1). The GT (3.3), JF (3.2), and the DS (2.9) system produced rooted cuttings with significantly greater S: R ratios than the CT system (1.5) (Table 4.1). Clone C produced rooted cuttings with a significantly greater S: R ratio (3.3) than Clones A (2.5) or B (2.4) (Table 5.1).

Experiment 2

There was a significant system by clone interaction for S: R ratio (Table 2.1). Though Clone C produced rooted cuttings with the largest S: R ratio for each of the production systems evaluated, the magnitude of the difference between Clone C and the other clones varied among production systems. For example, the S: R ratio of Clone C was significantly greater than the other clones in the February GT, May GT, and DS systems, but was similar to Clone A in the February and May CT systems (Figure 3.1). Contrasts revealed significant differences in S: R ratio between stem cuttings rooted in the DS/HW (2.9 g) and DS/LW (2.5 g) systems (Table 6.1). In addition, stem cuttings rooted in the May GT (2.8) system had significantly smaller S:

R ratios than those rooted in the February GT system (3.4). Treatments within a system significantly affected S: R ratio (Table 2.1).

Seedling Root Collar Diameter Classes

To rate the rooted cuttings produced in terms of existing seedling quality standards, RCD measurements were used to classify the rooted cuttings into seedling grades developed by Wakeley (1954).

Experiment 1

The GT system produced the highest percentage of rooted cuttings that were classified as Grade 1 and acceptable (Grades 1 and 2 combined) (Figure 6.1). However, only the GT-7 and GT-9 week treatments produced > 50% Grade 1 rooted cuttings. In the GT system, the 7 week transplant treatment (63%) produced the greatest number of Grade 1 rooted cuttings, while the 9 week transplant treatment (97.3%) produced the greatest number of acceptable grade rooted cuttings. The same pattern followed for rooted cuttings produced in the transplant treatments in the JF system. In the CT system, 89% of the rooted cuttings were considered acceptable, but only 10% were classified as Grade 1. The two DS system treatments produced the lowest number of acceptable grade rooted cuttings, with DS/S and DS/SH producing 58% and 66%, respectively.

Experiment 2

The February and May GT system produced the highest percentage of Grade 1 and acceptable rooted cuttings (Figure 6.1). Additionally, all the systems and treatments tested in this experiment produced > 50% Grade 1 rooted cuttings and over 90% acceptable grade rooted cuttings. Within the GT systems, the 9 week February GT transplant treatment produced the greatest proportion of Grade 1 rooted cuttings (80%), only slightly higher than the 9-week May GT system (76 %). For the two CT systems, the February CT system produced a greater number of Grade 1 rooted cuttings (62 %), than the May CT system (50 %). For the DS system treatments, the

LW/SH produced the greatest proportion of Grade 1 rooted cuttings (65%), while HW/S produced the lowest number of Grade 1 rooted cuttings (58%).

Discussion

Variation in morphological measurements is common among reforestation stock (Wilder-Ayers and Toliver 1987). For the two experiments, shoot and root morphology, as well as survival, varied among production systems. (In Experiment 2, rooting among production systems varied according to clone). Clone and treatment within a system also affected rooted cutting stock quality, often with "clone x production system" or "clone x treatment within a system" interactions. Additionally, rooting environment and cultural practices were influential factors affecting rooted cutting stock quality.

For both experiments, rooted cuttings from the GT (Experiment 1) and February GT (Experiment 2) transplant production systems were among those producing the largest rooted cuttings. In each case, the GT production system appeared to offer a consistent means for producing a large number of well developed rooted cuttings. In comparison, the JF and CT system rooted cuttings produced in Experiment 1 were average for most traits, while the DS system showed reasonable survival, but tended to produce the smallest rooted cuttings.

At the end of the second experiment, stem cuttings rooted in the February GT, February CT, and DS systems had greater survival rates and were larger than rooted cuttings produced by similar treatments (GT, CT, and DS systems) in Experiment 1. The February CT and DS systems in Experiment 2 produced rooted cuttings that were comparable for most shoot and root morphology traits. However, there were differences in survival between these two systems in Experiment 2. The February CT system rooted cuttings survived at rates similar to the February GT system rooted cuttings, while the DS system produced rooted cuttings with the lowest survival rates. Lower survival for the DS system rooted cuttings was most likely a result of lower rooting percentages. However, rooting was not evaluated independently from survival for the DS system stem cuttings in this study. Had the DS system stem cuttings rooted in higher percentages, survival of rooted cuttings for this system would probably have been higher.

Differences in clonal performance were also observed in Experiments 1 and 2. Significant “clone x production system” and “clone x treatment within a system” interactions suggest that clones should be tested in particular rooted cutting production systems before implementing operational production. However, because of the limited number of genotypes tested in these two experiments, it is not possible to extend this conclusion to all loblolly pine clones.

GT and JF Transplant Systems

Survival rates for the JF system rooted cuttings in Experiment 1 were lower than expected. In Experiment 1, stem cuttings stuck in the JF, GT, and CT systems were rooted under a mist regime designed to optimize rooting for stem cuttings of loblolly pine in Ray Leach SuperCells (CT system). This regime was also conducive for rooting stem cuttings at high percentages in the GT sponges, but apparently not as conducive for stem cuttings stuck in the JF pellets. LeBude (1999) demonstrated that loblolly pine stem cuttings were capable of rooting in high percentages in JF pellets. In Experiment 1, the JF pellets appeared to be more saturated with water than the GT sponges. Anaerobic conditions inside the JF pellets may have decreased or delayed rooting. Poor rooting conditions may have also been responsible for the smaller size of the JF rooted cuttings, by retarding root development and delaying growth before transplant. Tailored misting regimes may be successful in optimizing rooting conditions and survival for a given plug type. Following transplant to the outdoor nursery bed in Experiment 1, the GT system rooted cuttings and many of the JF rooted cuttings produced a considerable amount of new shoot and root growth. Growth rates among rooted cuttings seemed to reflect differences in root development at the time of transplant. Faster rooting clones were able to gain a competitive advantage and crowd clones with poorly developed root systems. Transplanting rooted cuttings into a nursery bed in clonal blocks, reducing nursery bed densities, and conducting top-pruning to control the height growth of early rooting cuttings might limit competition among rooted cuttings (Frampton and Hodges 1989).

Comparisons among transplant times for the two plug types tested in Experiment 1 revealed significantly lower survival rates, but greater shoot height and RCD measurements for the earliest GT and JF transplant treatments. Rooted cuttings transplanted earlier were afforded a longer growing season at the expense of a less developed root system at the time of transplanting. Decreased survival for the 7 week transplant time may have resulted from the inability of stem cuttings with poorly developed roots to survive transplant to an outdoor nursery bed. The intermediate transplant time of 9 weeks, perhaps, provided the best compromise between survival and growth. In Experiment 2, there were no differences in survival or shoot and root morphology between the February GT 9 weeks and February GT 12 weeks transplant treatments. Clonal differences, for all traits excluding RCD, existed within the February GT system tested in Experiment 2. Variation in clone performance within the February GT system was perhaps caused by differences in root system quality at the time of transplant and clonal growth traits.

The May GT system produced rooted cuttings that survived at similar rates but were smaller than those rooted in the February GT system. Based on the grading criteria of Wakeley (1954), over 90% of the surviving rooted cuttings from the May GT system were classified as Grade 1 or Grade 2. It appears that two GT rooted cutting production cycles are feasible in one growing season. Locating rooted cutting production systems in more southerly latitudes, with longer growing seasons should enhance growth for all the systems tested in this study.

A potential advantage of a transplant system is the ability to provide optimal rooting and post-rooting growing environments (Menzies and Arnott 1992). Following rooting, the rooted cuttings transplanted into an outdoor nursery bed were provided an excellent environment for shoot and root growth. In Experiments 1 and 2, the loamy sand of the nursery bed provided excellent drainage, adequate CEC, good moisture retention, and room for growing roots to expand. Also, the outdoor location of the nursery bed provided full sunlight for photosynthesis. These factors coupled with the advantageous rooting environment in the greenhouse, favored shoot and root growth for the GT transplant rooted cuttings.

Containerized Systems

Survival of the rooted cuttings produced in the CT system in Experiment 1 was lower than expected. In an earlier study rooted cuttings of loblolly pine in Ray Leach SuperCells survived at 83% (LeBude 1999). Most likely, rooting percentage and cultural treatment confounded survival for these rooted cuttings. For some stem cuttings, rooting may have been incomplete or nonexistent, resulting in mortality following transfer to the outdoor nursery bed. Likewise, when placed outdoors, the containerized media dried more quickly than the mineral soil of the nursery bed. Plant growth is the first process to be retarded by insufficient soil moisture (MacDonald 1984) and photosynthesis rates may be drastically reduced by even relatively small soil moisture deficits (Lavender 1984). Insufficient soil moisture, therefore, may have negatively impacted survival and growth of the containerized rooted cuttings.

The containerized medium [1 peat: 1 perlite (v/v)] used in Experiment 1 also had a lower cation exchange capacity (2.4 vs. 3.1 meq/100g) than the mineral soil in the nursery bed. Cuttings from all four systems were fertilized at the same rate throughout Experiment 1. Nutrient deficiencies within a plant typically reduce growth (Lavender 1984). Lower soil fertility, therefore, may have negatively impacted survival rates and morphological measurements of the rooted cuttings produced in the CT system in Experiment 1.

Stem cuttings rooted in the February CT system in Experiment 2 exhibited increased survival and growth compared with the CT system stem cuttings rooted in Experiment 1. Survival rates for the February CT rooted cuttings were almost identical to the survival rates determined for the February GT rooted cuttings, as well as survival rates reported for other studies involving containerized rooted cuttings of loblolly pine (LeBude 1999). Likewise, many of the shoot and root morphological traits assessed for the February CT rooted cuttings were considerable larger than the CT rooted cuttings produced in Experiment 1. For instance, mean shoot height increased from 15.8 to 22.6 cm and shoot dry weight increased from 14.1 to 21.5 g

(Table 3.1 and Table 5.1). Improved cultural practices including greater attention to containerized irrigation requirements, increased fertilization rates, pH and EC monitoring, and a modified container medium [1 peat: 1 perlite: 1 vermiculite (v/v/v)], intended to increase soil fertility, were all likely factors affecting the increase in survival and growth for rooted cuttings produced in this system during the second year.

The May CT rooted cuttings were some of the smallest produced in Experiment 2. This system produced rooted cuttings with the lowest shoot height, RCD, and S: R ratio of all the systems tested. Rooting percentage and survival rates for the May CT rooted cuttings, however, were among the highest during the second experiment. Despite the disparity in size and survival, the May CT system produced over 90% acceptable grade rooted cuttings, based on the grading standards of Wakeley (1954). Two CT rooted cutting production cycles are, apparently, feasible in one growing season in Raleigh, NC.

Root growth was limited in both experiments for the containerized rooted cuttings. Average root dry weight for the CT system rooted cuttings (Experiment 1) and the February CT system cuttings (Experiment 2) were 9.3 g vs. 10.5 g, respectively (Table 4.1 and Table 6.1). Root growth is often limited in containerized systems because of the smaller volume of medium the roots have to explore (Barnett and Brisstuckte 1986). Limited root growth for containerized stock, however, is not always considered a disadvantage for producing high quality reforestation stock. Containerized root systems are often compact, stay intact during transplanting to the field, and begin growing more quickly after out-planting than bare-root seedlings, which require more time to regenerate roots lost during lifting (Barnett 1978). Furthermore S: R ratios are often lower for containerized systems. The February CT system produced rooted cuttings with a lower S: R ratio than rooted cuttings produced in the February GT or DS systems. Improved survival has been reported for reforestation stock with lower S: R ratios when planted on dry sites (South and Mexal 1984).

Direct-stick System

Survival rates for the rooted cuttings produced in the DS system in Experiments 1 and 2 were similar to rooting percentages obtained by Frampton and Hodges (1989). These authors reported 64% rooting for dormant loblolly pine stem cuttings collected from pruned hedges and stuck in an outdoor nursery bed. In another study, Frampton et al. (1999) classified rooted cuttings produced in a DS system in terms of RCD seedling grades, with 55% classified as Grade 1. In the present study, only the DS system stem cuttings rooted in Experiment 2 had a similar percentage of Grade 1 cuttings, with approximately 62%. Less than 10% of the DS system rooted cuttings produced in Experiment 1 were classified as Grade 1. Reduced growth of the DS system rooted cuttings in Experiment 1 was likely due to sub-optimal irrigation (in this particular case over-watering during the initial rooting phase).

Rooting environment greatly affects the success of adventitious root formation (Hartmann et al. 2002). Survival rates for the DS system stem cuttings were perhaps lower than the other production systems tested in Experiment 1 and 2 because of the environmental conditions of the outdoor nursery bed. Non-rooted cuttings experience moisture loss as a result of transpiration from the needles. Successful rooting environments limit transpiration rates by providing a humid atmosphere with low transpirational demands, moderate temperatures that reduce heat stress, and maintaining light levels that allow photosynthesis and carbohydrate production, while simultaneously moderating temperature (Hartmann et al. 2002). Enclosed rooting areas, such as greenhouses, are often used to successfully root stem cuttings by increasing levels of humidity and, thus, reducing transpirational demand. Accompanied by intermittent mist, designed to reduce leaf temperatures and to provide moisture to the base of the stem cutting, and shading, provided to moderate light and temperature levels, favorable environmental conditions can be created for successful adventitious root formation.

Outdoor rooting environments generally have lower humidity levels and higher temperatures than those in the more controlled environment of a greenhouse. Furthermore, stem cuttings stuck in an outdoor nursery bed are often subject to desiccating winds not present in a greenhouse. Such environmental conditions can increase water stress in a stem cutting and inhibit or delay root formation (Hartmann et al. 2002). Environmental differences between the greenhouse and the outdoor nursery bed most likely negatively impacted rooting and survival for the loblolly pine stem cuttings stuck in the outdoor nursery bed during both studies. Better control of the rooting environment in and around the nursery bed may increase rooting and survival for this production system.

Applying the appropriate amount of irrigation to encourage root formation is challenging, as was evident in two outdoor loblolly pine rooting trials reported by Frampton and Hodges (1989). These studies demonstrated the negative affect of excessive or insufficient water on root formation. To overcome some of the guesswork associated with DS system rooting, Frampton et al. (1999) developed a system designed to modify irrigation according to measurable levels of environmental stress, including; ambient air temperatures, relative humidity, and wind-speed.

The irrigation controller utilized in Experiment 1 did not control for environmental variation as in Frampton et al. (1999) and, led to excessive irrigation. Though survival rates were still similar to the rooting results reported by Frampton et al. (1999), the resulting irrigation regime might have been a contributing factor affecting shoot and root growth following rooting in Experiment 1. Over-watering can result in saturated soils creating anaerobic conditions that limit root growth (Lavender 1984). Refinements made to the irrigation system during the second year (Appendices I and II) reduced the amount of water applied to the stem cuttings and likely contributed to the increase in shoot and root growth from Experiment 1.

The duration of irrigation applied to the DS system stem cuttings in Experiment 2 also affected shoot and root growth measurements. DS system stem cuttings rooted under HW were significantly taller and had larger root dry weight. However, the duration of irrigation did not affect rooting or survival. The HW treatment apparently provided a more conducive environment for root formation in the nursery bed, allowing for earlier root formation and greater growth following rooting. Secure and consistent watering is crucial for optimal rooting in this system. All of the production systems evaluated in this study, in fact, would benefit from back-up systems designed to protect cutting material from equipment failure during rooting. Useful back-up systems include; auxiliary irrigation systems, a secondary source of power, and an alarm system to notify supervisors of potential problems

Another limiting factor for stem cuttings rooted in the DS system was the time required to form adventitious roots. Based on observation of adventitious root formation on border stem cuttings surrounding the DS system plots, rooting began approximately 3 ½ months into the growing season. In contrast, rooting occurred as early as 5 weeks among some of the GT and CT system stem cuttings. This extended time required to root stem cuttings outdoors limited the amount of time allotted for subsequent shoot and root growth. Additionally, weather conditions in Raleigh, NC prohibited DS system sticking dates until the middle of April. Sticking prior to this date might have exposed the stem cuttings to freezing temperatures and frost heaving in the nursery bed. Perhaps, in a warmer climate, an earlier sticking date would allow for a longer growing season and greater growth for rooted cuttings produced in this system. However, even with the relatively short growing season in Raleigh, a considerable proportion of the DS rooted cuttings produced in Experiment 2 (over 90%) were classified as Grade 1 or Grade 2 stock (Wakeley 1954) (Table 4).

Despite the limitations of rooted cutting production in a DS system, continued work with this system could one day provide an efficient method of loblolly pine rooted cutting production that would conveniently fit into existing bare-root seedling nursery facilities. Advantages of this type of integration include lower production costs, greater ease of handling and transport, and decreased susceptibility of disease and nutritional imbalances (Dirr and Heuser 1987, Wilder-Ayers and Toliver 1987, Menzies and Arnott 1992).

Conclusion

Survival and shoot and root morphology measurements for rooted cuttings of loblolly pine varied among the TP, CT, and DS production systems tested in Experiments 1 and 2. However, by the end of the second year, in the three systems tested, 90% of all the rooted cuttings produced met acceptable grading standards (Wakeley 1954). GT system rooted cuttings were among the largest produced each year and provided a consistent means of producing a large number of well developed rooted cuttings. Transplanting TP system rooted cuttings to the outdoor nursery bed after 9 weeks of rooting in the greenhouse provided the best combination of survival and size of the three transplant times tested. Although the CT system rooted cuttings required greater attention to moisture requirements, more regular pH and electrical conductivity (EC) monitoring, and higher fertility rates, this system provided a high percentage of rooted cuttings with well protected root systems and low S: R ratios.

Rooted cuttings produced in the DS system survived at the lowest rates and were consistently smaller than the February transplant system rooted cuttings. This difference in size was most likely due to slower and lower percentages of rooting for the DS system stem cuttings. By the end of the second experiment, the DS system rooted cuttings were comparable in size to the February CT and May TP system rooted cuttings. Rooting percentage, survival, and growth could increase for stem cuttings stuck in a DS system if environmental conditions in and around outdoor nursery bed were improved during the initial rooting phase, the irrigation regime for this system was optimized, and the location of production was in more southerly latitudes than Raleigh, NC, with longer growing seasons.

A few studies report potential rooted cutting grading standards for loblolly pine (Goldfarb et al. 1998, Frampton et al. 1999, Foster et al. 2000, Frampton et al. 2002). These studies suggest root number (Foster et al. 2000) and measurements of root mass (Goldfarb et al. 1998, Frampton et al. 2002) as potential indicators of loblolly pine rooted cutting stock quality. Additional research involving loblolly pine rooted cuttings could enhance existing grading criteria. Likewise future field test may demonstrate advantages of specific stock types in specific situations. For instance, tall reforestation stock, such as those produced in early season TP production systems, may be favored for use on sites with a large amount of vegetative competition.

Separate grading standards may also be needed for containerized and bare-root rooted cutting stock grown in an outdoor nursery bed, as either TP or DS planting stock. Due to greatly reduced soil volume, shoot and root growth is usually more limited for containerized stock than for nursery grown stock. Likewise, due to the compact and protected nature of containerized root systems, fewer root morphological measurements could be practically applied as stock quality standards. Grading containerized root systems requires removing the container, and as a result, discarding its protective function. Of course, containerized rooted cutting stock with poorly developed root systems could also be identified at the time of planting and culled. Shoot morphology measurements such as shoot height and RCD, combined with a visual check for the presence of roots emerging from the bottom of the container, might provide more appropriate grading criteria for this rooted cutting production system.

On the other hand, both shoot and root morphology measurements would be appropriate for grading bare-root rooted cutting stock. Morphological differences among TP and DS system rooted cuttings could be reduced by optimizing cultural practices for each production system. For instance, top or root pruning could moderate shoot and root growth in the TP system. Efforts should also be made to conduct field tests with various rooted cutting stock types and stock with various shoot and root morphological measurements in years with both average and below average rainfall.

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Table 1.1. Experiment 1: Mixed model, split-plot analysis of variance p-value results for seven loblolly pine rooted cutting morphology traits according to system, clone, system x clone, treat (system), and clone x treat (system). Cuttings from three clones were stuck in each of nine treatments representing four rooted cutting production systems.

Trait	System	Clone	System x Clone	Treat (Systems)	Clone x Treat (System)
*Survival (%)	**< 0.0001**	**< 0.0001**	**0.0188**	**0.0128**	0.2777
*Height (cm)	**< 0.0001**	**< 0.0001**	0.1192	**0.0126**	**0.0181**
*Shoot Dry Weight (g)	**< 0.0001**	**< 0.0001**	0.2575	0.7980	0.2661
*RCD (mm)	**< 0.0001**	**< 0.0001**	0.6412	**0.0010**	**0.0497**
*Primary Root Number	**< 0.0001**	**0.0001**	**0.0259**	0.2616	0.3834
*Root Dry Weight (g)	**< 0.0001**	**0.0001**	0.1650	0.9246	0.1203
S: R Ratio	**0.0002**	**0.0041**	0.2359	0.3993	0.5608

*Analysis of variance was conducted on transformed data.

Table 2.1. Experiment 2: Mixed model, split-plot analysis of variance p-value results for eight loblolly pine rooted cutting morphology traits as influenced by system, clone, system x clone, treat (system), clone x treat (system). Cuttings from three clones were stuck in each of ten treatments representing five rooted cutting production systems.

Trait	System	Clone	System x Clone	Treat (Systems)	Clone x Treat (System)
Rooting Percentage	**0.0025**	< **0.0001**	**0.0004**	**0.0329**	< **0.0001**
Survival (%)	< **0.0001**	< **0.0001**	**0.0011**	0.1980	**0.0059**
*Height (cm)	< **0.0001**	< **0.0001**	< **0.0001**	< **0.0001**	0.6393
*Shoot Dry Weight (g)	< **0.0001**	< **0.0001**	**0.0002**	< **0.0001**	0.0839
*RCD (mm)	< **0.0001**	< **0.0001**	**0.0003**	< **0.0001**	0.9493
*Primary Root Number	< **0.0001**	< **0.0001**	< **0.0001**	0.4724	**0.0031**
*Root Dry Weight (g)	< **0.0001**	< **0.0001**	**0.0011**	**0.0002**	0.0872
S: R Ratio	**0.0003**	< **0.0001**	**0.0385**	< **0.0001**	0.1260

*Analysis of variance was conducted on transformed data.

Table 3. 1. Experiment 2: Effect of treatment on eight loblolly pine rooted cutting morphology traits. P-values were determined using preplanned contrasts. Factors tested within the DS system included two misting durations, high water (HW) and low water (LW), and two light levels, full sun (S) and 50% shade (SH).

Preplanned Contrasts	Rooting (%)	Survival (%)	*Height (cm)	*Shoot Dry Weight (g)	*RCD (mm)	*Primary Root Number	*Root Dry Weight (g)	S: R Ratio
DS/S vs. DS/SH	N/A	0.1674	0.4149	0.0520	0.6648	0.9174	0.0744	0.9097
DS/HW vs. DS/LW	N/A	0.4421	**0.0017**	**0.0007**	0.3795	0.6521	**0.0133**	**0.0174**
Feb. GT vs. May GT	**< 0.0001**	0.3333	**< 0.0001**	**< 0.0001**	**< 0.0001**	0.1065	**< 0.0001**	**< 0.0001**
Feb. CT vs. May CT	0.1162	0.5619	**< 0.0001**	**0.0164**	0.1557	**0.0052**	0.2265	**0.0440**

*Analysis of variance was conducted on transformed data.

Table 4. 1. Experiment 1: Production system means (standard error) and significance for five loblolly pine rooted cutting morphology traits. Production system means (*) with the same letter, within a trait, had values that did not differ significantly ($p<0.05$) using pair-wise comparison of transformed (**) data.

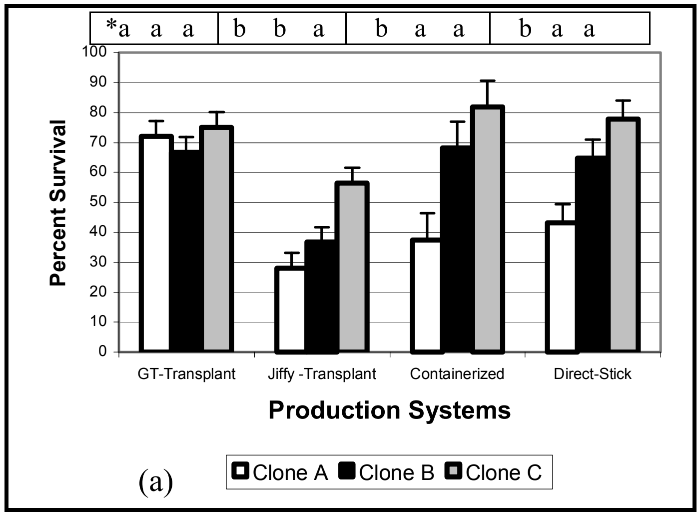
Morphology Traits	GT Transplant Production System	JF Transplant Production System	CT Production System	DS Production System
** Height (cm)	***26.5 a** (0.5998)	**22.14 b** (0.6622)	**15.8 c** (1.061)	**13.62 d** (0.7503)
** Shoot Dry Weight (g)	**44 a** (2.516)	**21.9 b** (2.717)	**14.1 b** (3.949)	**10.17 c** (2.967)
** RCD (mm)	**4.93 a** (0.121)	**4.23 b** (0.1279)	**3.97 b** (0.1821)	**3.52 c** (0.1397)
** Root Dry Weight (g)	**13.7 a** (0.7077)	**6.65 b** (0.7793)	**9.31 a** (1.203)	**3.79 c** (0.8680)
S: R Ratio	**3.33 a** (0.2180)	**3.21 a** (0.2379)	**1.49 c** (0.3576)	**2.89 b** (0.2626)

Table 5. 1. Experiment 1: Clone means (standard error) and significance for five loblolly pine rooted cutting morphology traits. Clone means (*) with the same letter, within a trait, had values that did not significantly differ (p<0.05) using pair-wise comparisons of transformed (**) data.

Trait	Clone A	Clone B	Clone C
** Height (cm)	*18.7 b (0.6738)	17.8 b (0.6511)	22 a (0.6414)
** Shoot Dry Weight (g)	19 b (2.8244)	16 b (2.7515)	32.7 a (2.7208)
** RCD (mm)	4.37 a (0.1257)	3.79 b (0.1230)	4.32 a (0.1219)
** Root Dry Weight (g)	8 b (0.8175)	6.6 b (0.7921)	10.5 a (0.7812)
S: R Ratio	2.46 b (0.2485)	2.44 b (0.2414)	3.29 a (0.2383)

Table 6.1. Experiment 2: Treatment within a system shoot height, shoot dry weight, root collar diameter (RCD), root dry weight, and shoot to root ratio means (standard error) and significance for loblolly pine rooted cuttings. Factors tested within the direct-stick system included two misting durations, high water (HW) and low water (LW), and two light levels, full sun (S) and partial shade (SH). Treatment means (*) with the same letter, within a production system, had values that did not differ significantly (p<0.05) using preplanned contrasts of transformed data (**).

Traits	February GT Trans. System		February Cont. System	Direct-Stick System				May GT Trans. System		May Cont. System
	9 wk	**12 wk**	**12 wk**	**HW/S**	**HW/SH**	**LW/S**	**LW/SH**	**8 wk**	**10 wk**	**10 wks**
****Shoot Height (cm)**	***43.2 a** (1.7531)	**43.3 a** (1.7787)	**22.6** (1.7787)	**25.6 a** (1.7531)	**23.8 ab** (1.7531)	**21.7 b** (1.8164)	**21.5 b** (1.7787)	**27.2 a** (1.731)	**25.0 a** (1.8164)	**17.0** (1.7531)
****Shoot Dry Weight (g)**	**73.9 a** (3.899)	**61.5 a** (4.027)	**21.5** (4.027)	**26.3 ab** (3.899)	**27.5 a** (3.899)	**18.3 c** (4.212)	**20.1 bc** (4.027)	**23.2 a** (3.899)	**17.2 b** (4.212)	**13.9** (3.899)
****RCD (mm)**	**7.15 a** (0.2237)	**6.85 a** (0.2311)	**5.0** (0.2311)	**5.46 a** (0.2237)	**5.31 a** (0.2237)	**5.06 a** (0.2237)	**5.35 a** (0.2311)	**5.5 a** (0.2237)	**5.2 a** (0.2417)	**4.6** (0.2237)
****Root Dry Weight (g)**	**23.3 a** (1.315)	**18.5 a** (1.4)	**10.5** (1.358)	**9.1 ab** (1.315)	**9.5 a** (1.315)	**6.3 b** (1.420)	**7.8 ab** (1.358)	**8.4 a** (1.315)	**6.2 a** (1.420)	**8.6** (1.315)
Shoot to Root Ratio	**3.37 a** (0.1687)	**3.5 a** (0.1738)	**2.0** (0.1714)	**2.82 a** (0.1687)	**2.92 a** (0.1687)	**2.56 a** (0.1753)	**2.52 a** (0.1714)	**2.82 a** (0.1687)	**2.72 a** (0.1753)	**1.65** (0.1687)



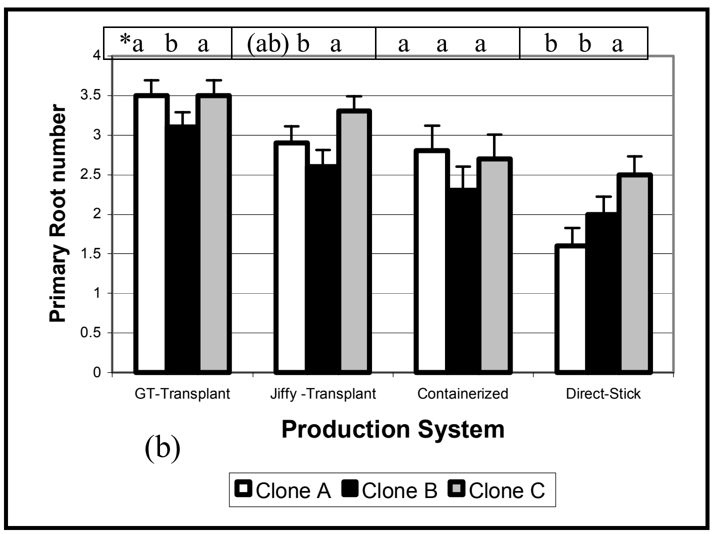


Figure 1.1. Experiment 1: Means, standard error, and significance as influenced by system and clone on (a) percent survival and (b) primary root number for loblolly pine rooted cuttings. Clone means (*) with the same letter, within a system, had values that did not differ significantly (p< 0.05) using pair-wise comparisons of transformed data.

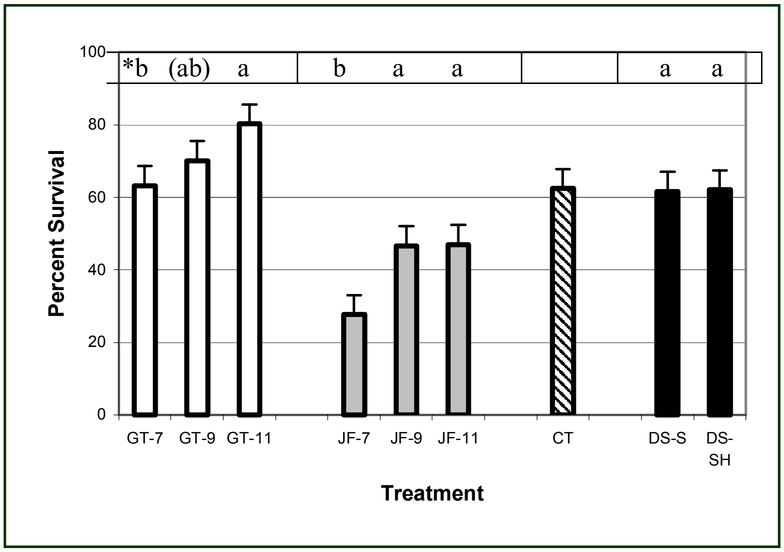


Figure 2.1. Experiment 1: Means, standard error, and significance as influenced by treatment (system) on percent survival for loblolly pine rooted cuttings. Treatment means with the same letter (*), within a system, had values that did not differ significantly (p<0.05) using preplanned contrasts of transformed data.

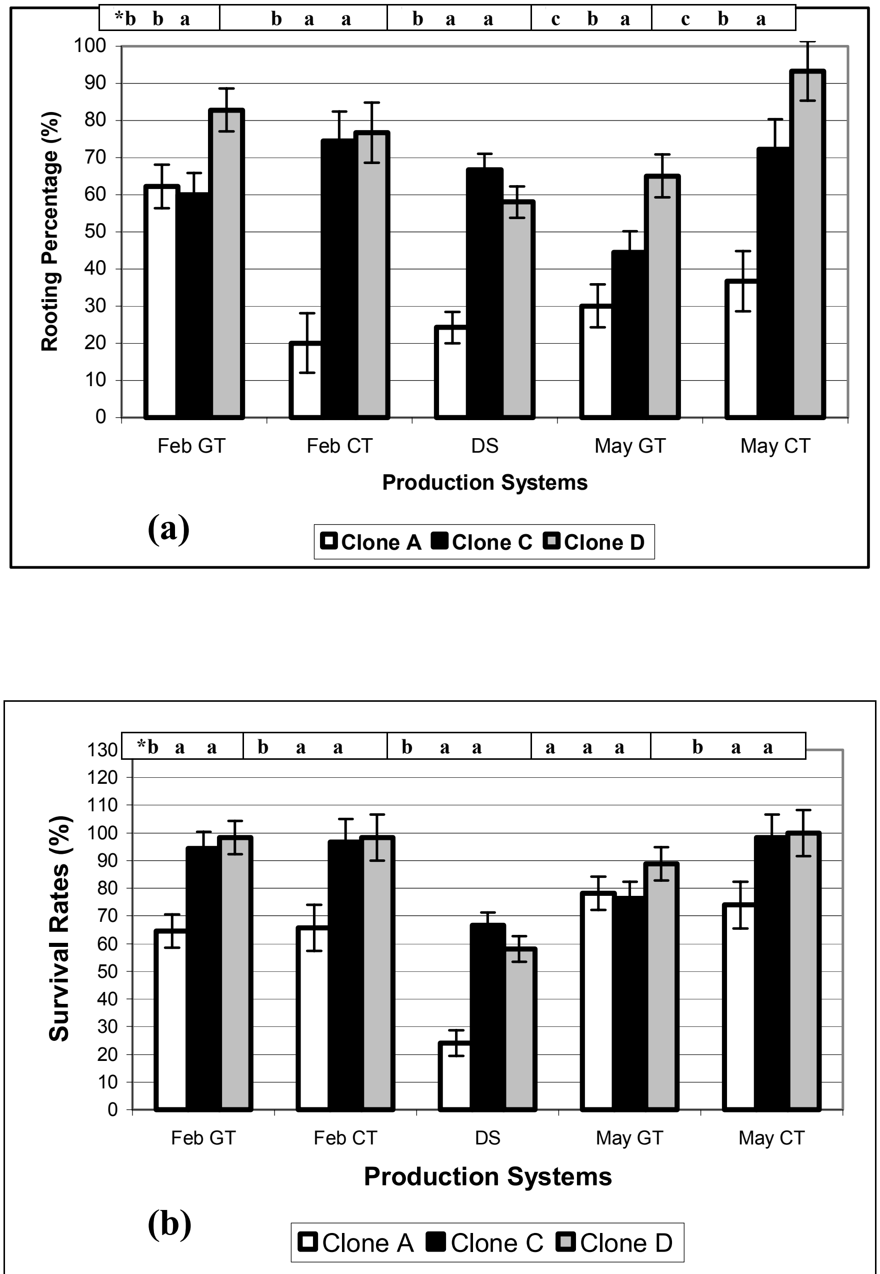
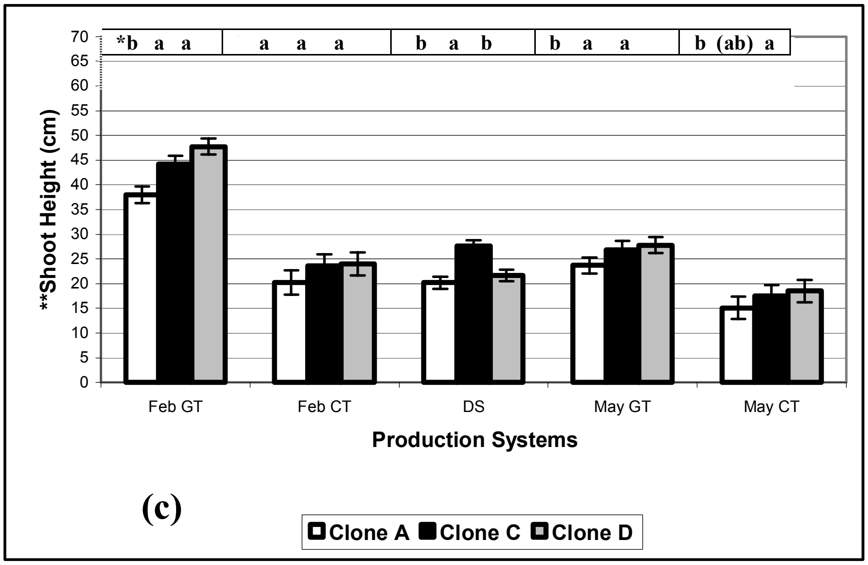


Figure 3.1. Experiment 2: Mean (a) rooting percentage and (b) survival rate for loblolly pine rooted cuttings as influenced by system and clone. Clone means with the same letter (*), within a system, had values that did not differ significantly ($p<0.05$) using pair-wise comparisons.



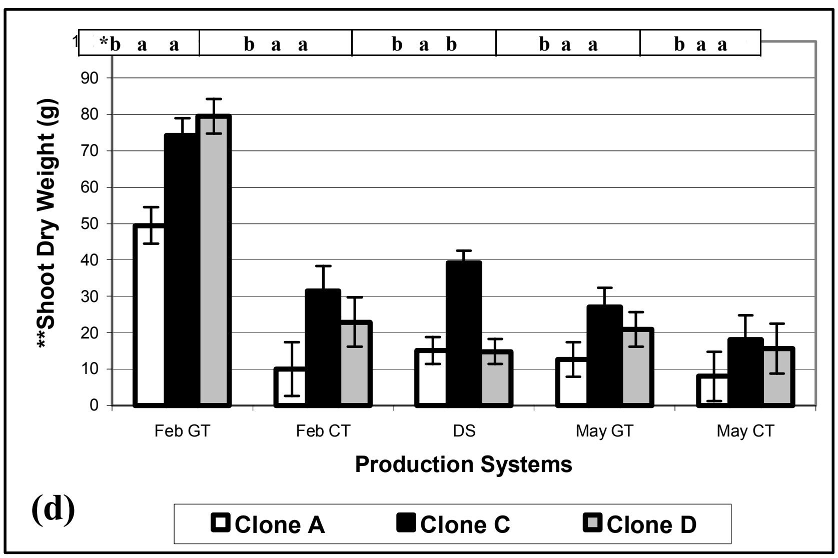
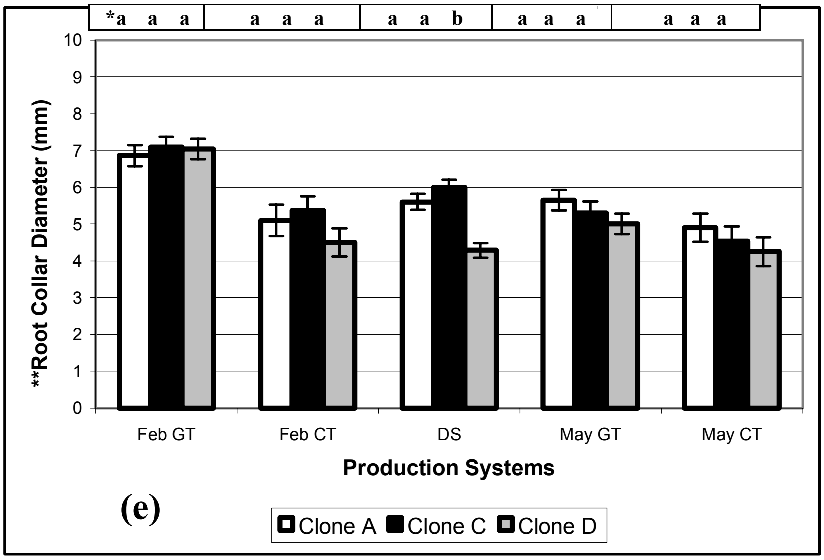


Figure 3.1. Experiment 2: Mean (c) shoot height and (d) shoot dry weight for loblolly pine rooted cuttings as influenced by system and clone. Clone means with the same letter (*), within a system, had values that did not differ significantly ($p<0.05$) using pair-wise comparisons of transformed data (**).



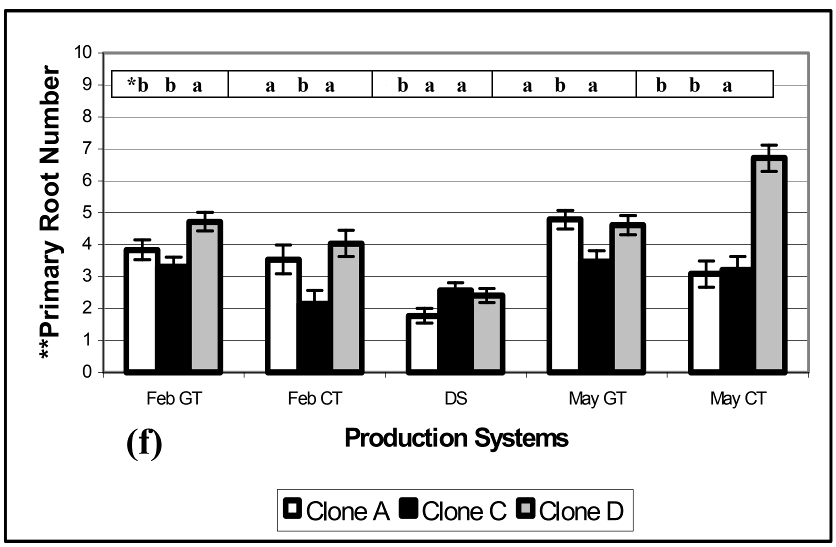


Figure 3.1. Experiment 2: Mean (e) root collar diameter and (f) primary root number for loblolly pine rooted cuttings as influenced by system and clone. Clone means with the same letter (*), within a system, had values that did not differ significantly ($p<0.05$) using pair-wise comparisons of transformed data (**).

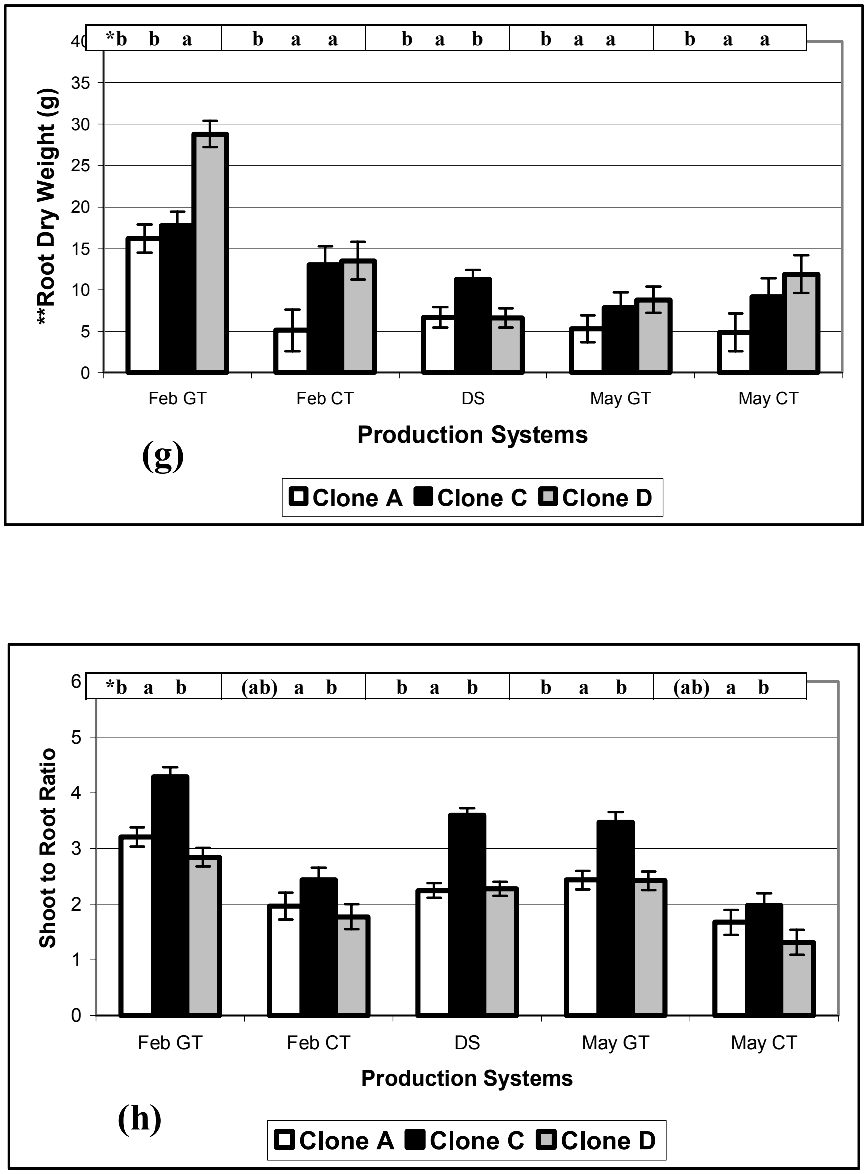


Figure 3.1. Experiment 2: Mean (g) root dry weight and (h) shoot to root ratio for loblolly pine rooted cuttings as influenced by system and clone. Clone means with the same letter (*), within a system, had values that did not differ significantly (p<0.05) using pair-wise comparisons of transformed data (**).

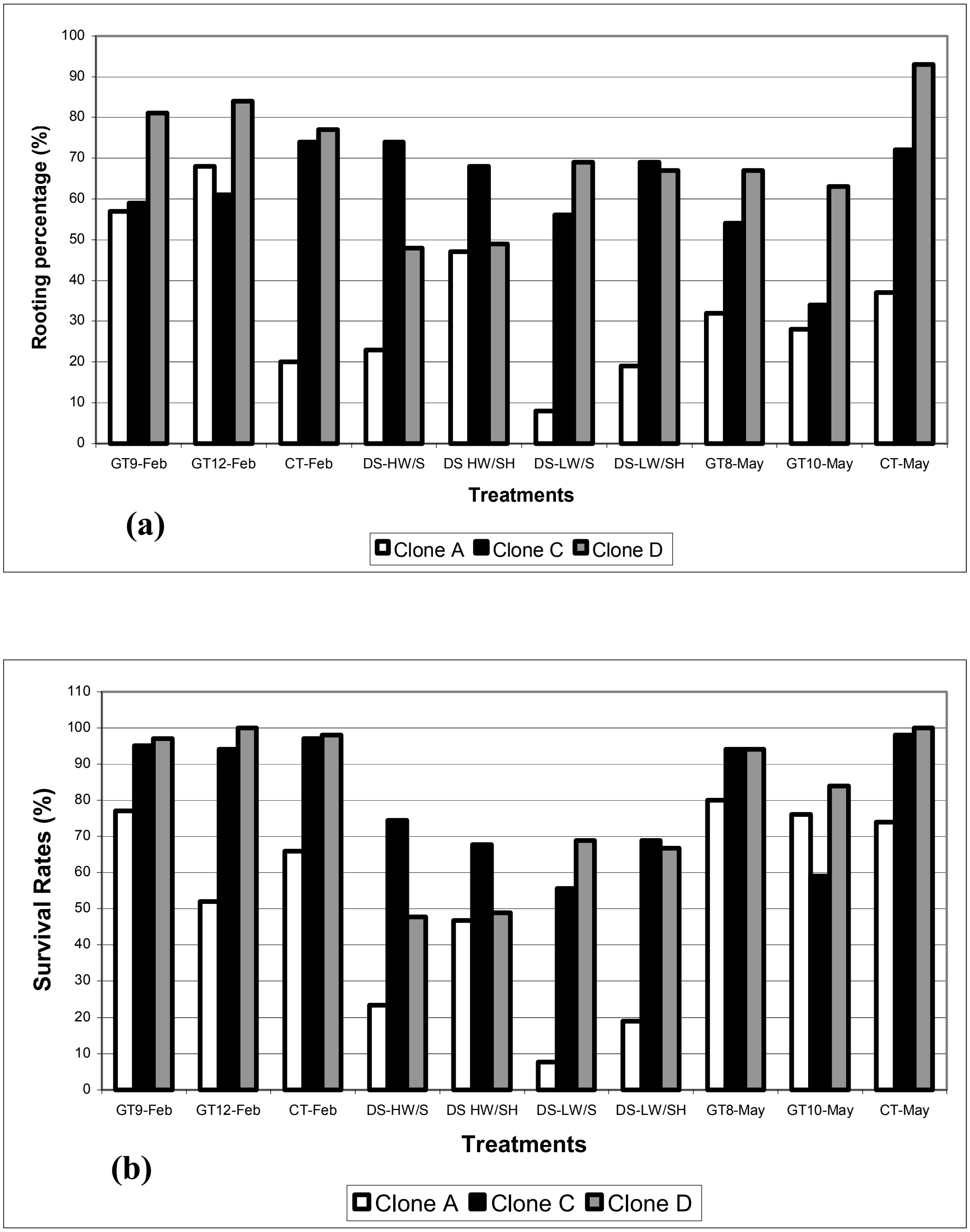


Figure 4.1. Experiment 2: Mean (a) rooting percentage and (b) survival rates for loblolly pine rooted cuttings as influenced by treatment and clone.

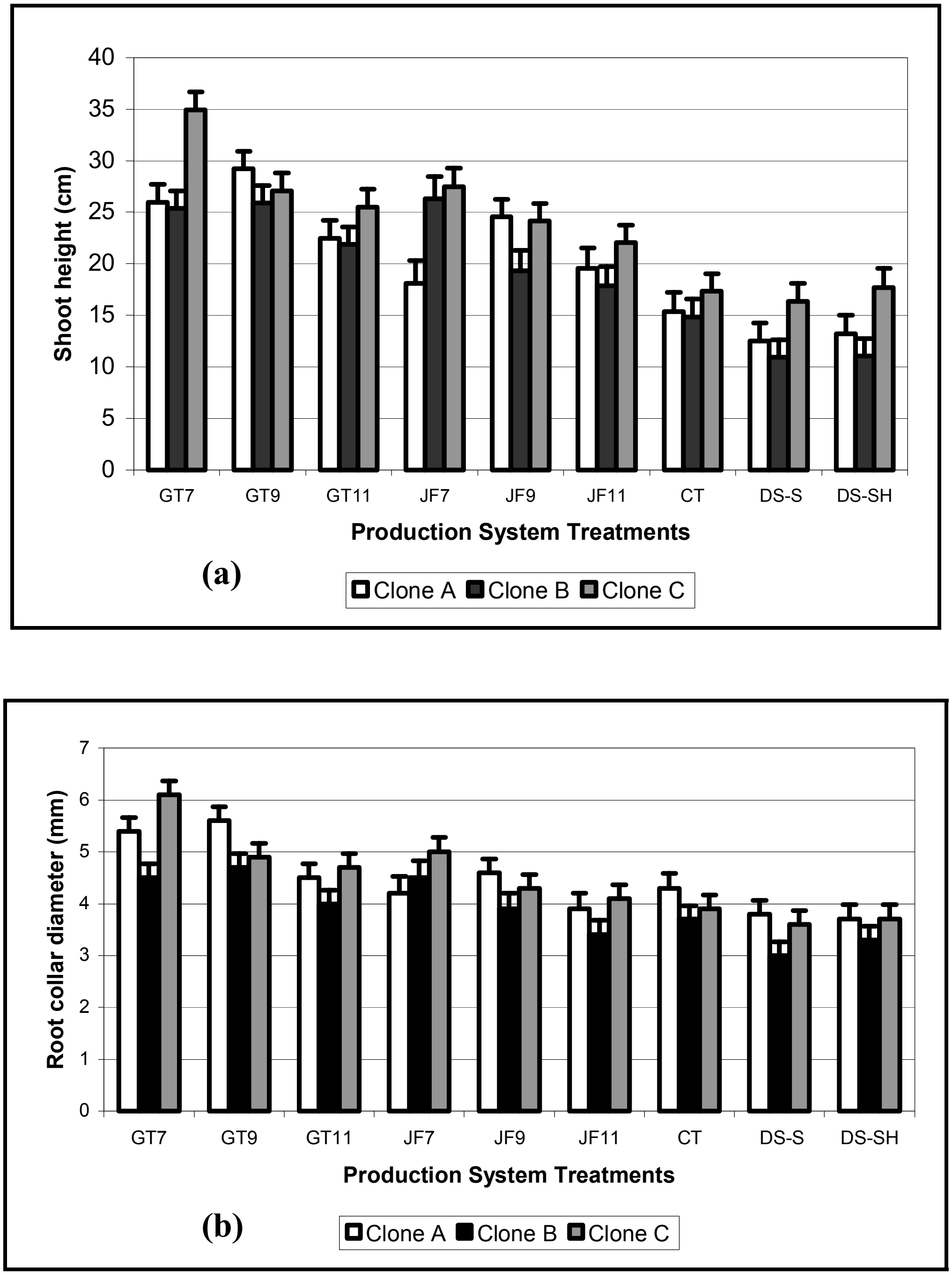


Figure 5.1. Experiment 1: (a) Shoot height and (b) root collar diameter means and standard error for loblolly pine rooted cuttings as influenced by treatment and clone.

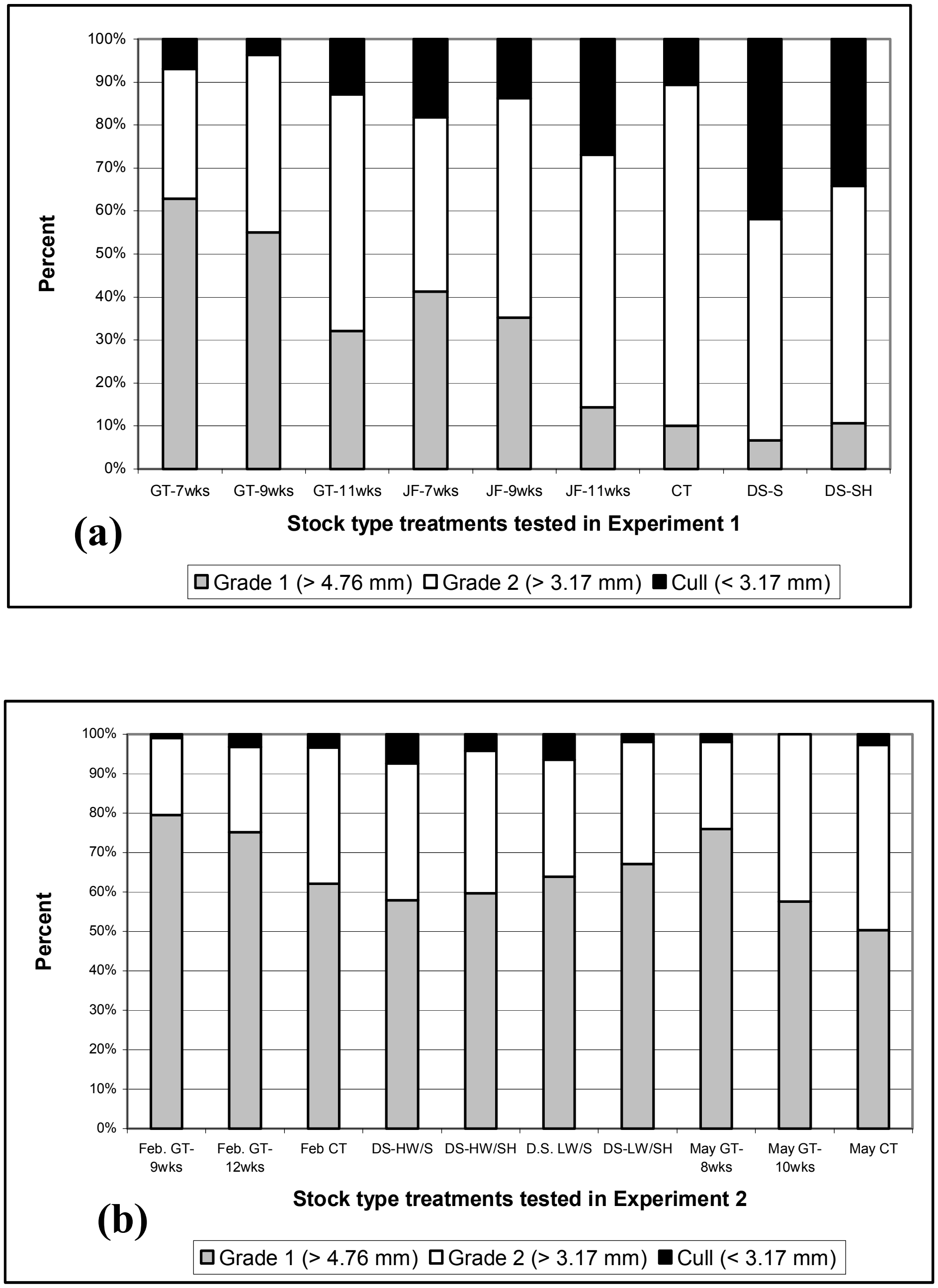


Figure 6.1. Grade of rooted cuttings according to seedling root collar diameter classes (Wakeley 1954). Percentage of each grade shown for rooted cuttings produced within (a) one of nine stock type treatments in Experiment 1 and (b) one of ten stock type treatments in Experiment 2.

Appendices

Appendix I. Experiment 1: The outdoor nursery bed irrigation regime employed in a loblolly pine rooted cutting experiment conducted at the NCSU Horticulture Field Laboratory in Raleigh, NC in 2000.

Date	**<u>Outdoor Nursery Bed Irrigation Regime for the</u>** **<u>GT Transplant, JF Transplant, CT, and DS Systems</u>** (Frequency (seconds) / Duration (seconds) of mist)
4/25 – 5/4	**12 min / 15 sec**
5/5	**12 min / 25 sec**
5/6	**10 min / 25 sec**
5/7 – 5/8	**8 min / 30 sec**
5/9	**10 min / 30 sec**
5/10	**9 min / 45 sec**
5/11 – 5/13	**10 min / 45 sec**
5/14 – 5/15	**8 min / 45 sec**
5/16	**9 min / 45 sec**
5/17 – 8/3 8:00 AM to 11:00 AM	**12 min / 45 sec**
5/17 – 8/3 11:00 AM to 7:00 PM	**8 min / 60 sec**
8/4	**12 min / 60 sec**
8/8	**15 min / 60 sec**
8/11	**20 min / 60 sec**
8/23	**40 min / 60 sec**
8/26	**2 hours / 60 sec**
8/30	**4 hours / 60 sec**
9/1	**6 hours / 60 sec**
9/5	**10 minutes every three days**
10/9	**Water off!**

Appendix II. Experiment 2: Four outdoor nursery bed irrigation regimes employed in a loblolly pine rooted cutting experiment conducted at the NCSU Horticulture Field Laboratory in Raleigh, NC in 2001.

Weeks of study	Date	Outdoor Nursery Bed Irrigation Regimes (Frequency (*solar units) / Duration (seconds) of mist)			
		DS High Irrigation	DS Low Irrigation	February GT and CT	May GT and CT
1	4-13	8 / 60	8 / 30	N/A	N/A
2	4-20	10 / 60	10 / 30	N/A	N/A
3-4	4-27	12 / 60	12 / 30	16 / 60	N/A
5-6	5-10	14 / 60	14 / 30	32 / 60	N/A
7	5-22	18 / 60	18 / 30	32 / 60	N/A
8	5-29	18 / 60	18 / 30	48 / 60	N/A
9	6-5	22 / 60	22 / 30	Rainbird: every 2 days for the containers and every 3 days for the transplants	N/A
10	6-12	30 / 60	30 / 30	2 X day/5 min	N/A
11-12	6-19	45 / 60	45 / 30	2 X day/5 min	N/A
13-14	7-3	60 / 60	60 / 30	Hand water once a day	16
15	7-17	120 / 60	120 / 30	"	32
16	7-24	Rainbird: 3hours/3minutes	Rainbird: 3hours/3minutes	Hand water every 2 days	32
17	8-1	1 day/5 minutes	1 day/5 minutes	Rainbird: every 2 days for the containers and every 3 days for the transplants	32
18	8-8	Hand water every three days	Hand water every three days	"	Rainbird: 2 X day/5 min
19	8-15	"	"	"	Hand water once a day
20	8-22	"	"	"	every 2 days for the containers and every 3 days for the transplants

***The Davis Solar 6A misting controller frequency was based on light measurements. One unit equals 2000 foot-candle or .02 moles/m^2. On a clear sunny day at noon, one unit equals approximately 30 seconds. On a completely overcast day at noon, one unit equals approximately 3 minutes. At night no irrigation was applied. The Rainbird was a fixed-cycle irrigation controller used periodically to supplement irrigation.**

Appendix III. Experiment 1: Production systems, treatments, sticking and transplant dates, sticking depth, and cutting density (greenhouse/nursery bed) for a loblolly pine rooted cutting experiment conducted at the NCSU Horticulture Field Laboratory in Raleigh, NC in 2000.

Propagation system	Treatments	Sticking date	Transplant date	Sticking depth (cm)	Density (ft^2) Greenhouse /Nursery bed
GT Transplant	1. GT 7 weeks	March 10	April 27	1.5	117/25
	2. GT 9 weeks		May 11		
	3. GT 11 weeks		May 25		
Jiffy Transplant	4. Jiffy 7 weeks	March 10	April 27	1.5	117/25
	5. Jiffy 9 weeks		May 11		
	6. Jiffy 11 weeks		May 25		
Containerized	7. Containerized 12 wks	March 10	June 1	2	49/25
Direct-stick system	8. Sun	April 25	N/A	4	NA/25
	9. Shade				

Appendix IV. Experiment 2: Production systems, treatments, sticking and transplant dates, sticking depth, and cutting density (greenhouse/nursery bed) for a loblolly pine rooted cutting experiment conducted at the NCSU Horticulture Field Laboratory in Raleigh, NC in 2001.

Production system	Treatments	Sticking date	Transplant date	Sticking depth (cm)	Density (/ft^2) Greenhouse / Nursery bed
GT Transplant	1. February - 9 wks	February 22	April 26	1.5	117/25
	2. February - 12 wks		May 17		
	3. May - 8 wks	May 4	July 3		
	4. May - 10 wks		July 14		
Containerized	5. February - 12 wks	February 22	May 17	2	49/25
	6. May - 10 wks	May 4	July 14		
Direct-stick	7. High water/sun	February 14	N/A	4	25
	8. High water/shade				
	9. Low water/sun				
	10. Low water/shade				

Appendix V. Experiment 1: Fertilizer regime employed during a loblolly pine rooted cutting experiment conducted at the NCSU Horticulture Field Laboratory in Raleigh, NC in 2000.

Week	Date	Systems
		GT, CT, and DS
8	6-11	50 ppm N/wk
9	6-18	50 ppm N/wk
10	6-25	50 ppm N/wk
11	7-2	50 ppm N/wk
12	7-9	50 ppm N/wk
13	7-16	50 ppm N/wk, Iron chelate
14	7-23	50 ppm N/wk
15	7-30	100 ppm N/wk
16	8-5	100 ppm N/wk
17	8-12	100 ppm N/wk
18	8-19	100 ppm N/wk
19	8-26	100 ppm N/wk
20	9-2	100 ppm N/wk
21	9-9	100 ppm N/wk
22	9-16	100 ppm N/wk
23	9-23	100 ppm N/wk
24	9-30	100 ppm N/wk
25	10-6	100 ppm N/wk
Total Amount of N / ha		100 kg N / ha

Appendix VI. Experiment 2: Four fertilizer regimes employed during a loblolly pine rooted cutting experiment conducted at the NCSU Horticulture Field Laboratory in Raleigh, NC in 2001.

Week	Date	Systems			
		DS	February GT	February CT	May GT and CT
7	5-22		50 ppm N / wk	50 ppm N / wk	N/A
8	5-29	50 ppm N / wk	50 ppm N / wk	50 ppm N / wk	N/A
9	6-5		50 ppm N / wk .1lb Solubor (10 kg/ha)	50 ppm N / 2X wk .1lb Solubor (10 kg/ha)	N/A
10 -12	6-12	50 ppm N / wk	50 ppm N / wk	50 ppm N / 2X wk	N/A
13-14	7-3	50 ppm N / wk	50 ppm N / wk	50 ppm N / 2X wk	50 ppm N / wk
15	7-17	100 ppm N / wk	100 ppm N / wk	100 ppm N / 2X wk	100 ppm N / wk
16	7-24	100 ppm N / wk	100 ppm N / 2X wk	100 ppm N / 2X wk	100 ppm N /2X wk
17	8-1	100 ppm N / wk	100 ppm N / 2X wk Iron Chelate	100 ppm N / 2X wk Iron Chelate	150 ppm N / wk
18-19	8-8	150 ppm N / wk	150 ppm N / 2X wk	150 ppm N / 2X wk	150 ppm N / wk
20	8-22	150 ppm N / wk Iron Chelate .1lb Solubor (10 lbs/acre)	150 ppm N / 2X wk Iron Chelate	150 ppm N / 2X wk Iron Chelate	150 ppm N / wk Iron Chelate .1lb Solubor (10 lbs/acre)
21	8-29	150 ppm N / wk Until 10/1	150 ppm N / wk Until 10/1	150 ppm N / wk until 10/1	150 ppm N / wk until 10/1
Total amount of N / ha		110 kg N / ha	155 kg N / ha	180 kg N / ha	110 kg N / ha

Supplementary Sweetgum Literature Review

Species overview with special emphasis on propagation of sweetgum by stem cuttings and the challenges associated with rooting this species

Liquidambar styraciflua L. (sweetgum), a member of the Hamamelidaceae, is an important commercial hardwood species in the southern United States. Sweetgum is used extensively for a variety of wood products including furniture, cabinets, particle-board, boxes, crates, pallets, and plywood (Sutter 1989). It is also valued by the paper industry due to its extensive range and abundance (Harlow et al. 1996). Sweetgum is often used as a street tree because of its unique star shaped leaves, bright fall color, rounded crown, tolerance to a variety of sites (Sutter 1989, Plotnik 2000), and general pest resistance (Harlow et al. 1996).

The only species in the genus *Liquidambar* indigenous to the new world (Harlow et al. 1996), the native range of sweetgum extends from Connecticut (41^{0} N) south to central Florida, and west to eastern Texas, Oklahoma, Arkansas, and Missouri. Sweetgum also extends further south into Central America with concentrations in Mexico, Belize, Guatemala, and Honduras. The southern limit of sweetgum is central Nicaragua (13^{0} N) (Sutter 1989, Harlow et al. 1996). Sweetgum occurs at elevations ranging from sea level to approximately 900 m in the southern Appalachians (Harlow et al. 1996).Sweetgum has been planted as an ornamental, and experimentally in the subtropics as well.

Sweetgum tolerates a variety of soil types, ranging from heavy clay to poorly drained alluvial sites (Sutter 1989) and is commonly found in mesic upland forests, on slopes, and in ravines (Harlow et al. 1996). However, it performs best on well-drained flood plains and moist, acidic alluvial clay and loam soils of river bottoms (Harlow et al. 1996).

The species is intolerant of low light levels. It is an aggressive pioneer species especially in disturbed areas and old fields. Sweetgum is a prolific stump sprouter and is also capable of producing an abundance of fast growing root suckers. Some sweetgum stands are of stump sprout or root sucker origin (Harlow et al. 1996). Sweetgum can live between 200 and 300 years of age, reach 24 to 36 m in height, and

0.9 to 1.2 m at diameter breast height (dbh). The largest recorded sweetgum was 60 m tall and had a dbh of 1.8 m (Harlow et al. 1996).

Sweetgum has been propagated by seed (Sutter 1989) and used in forest plantings for several decades. Seeds of the species are commonly germinated and grown outdoors in nursery beds to obtain 1-0 bare-root planting stock (Kaszkurewicz and Keister 1975, McNabb 2001). Seedlings are also produced in containers as 1-0 containerized planting stock.

In plantations, sweetgum has preformed variably. On good sites with weed control it can grow rapidly in plantation systems, while under dry and weedy conditions the growth rate can be slow. A common observation in plantations is the great variability among individual trees, perhaps, reflecting high genetic diversity (Daniel J. Robison, personal communication). Growth of sweetgum is highly genetically controlled (Cunningham 1989). Genetic improvement and clonally propagation may be important in deploying sweetgum as a plantation species. One promising method for clonally propagating sweetgum is by rooting juvenile stem cuttings.

Rooted Cuttings of Sweetgum

Several researchers have demonstrated the potential for rooting sweetgum stem cuttings. Sweetgum stem cuttings have been collected and rooted from root suckers (Johnson 1964, Farmer 1966), epicormic branches (Kormanik and Brown 1973, Kormanik and Brown 1974), and stump sprouts (Johnson 1964, Cunningham 1989). In each case, the cuttings rooted represented juvenile material.

For many tree species, stem cuttings collected from juvenile shoots generally root more easily than from shoots in an adult phase of growth and exhibit less abnormal growth habits (Teclaw and Isebrands 1987, Hamann 1998, Hartmann et al. 2002). Unfortunately, a limited number of stem cuttings can be obtained annually from root suckers, epicormic branches or stump sprouts. Each of these three cutting types provides a means of producing a limited number of juvenile stem cuttings from individual, mature trees. If large-scale clonal propagation of selected sweetgum genotypes is desired, techniques for providing a large number of juvenile stem cuttings for rooted cutting propagation will be required (Hamann 1998).

Pruning slows down the process of maturation in several tree species by stimulating juvenile shoot growth (Hartmann et al. 2002). Together pruning and serial propagation have been successfully used to produce stock plant hedges that are capable of providing numerous juvenile stem cuttings from a single sweetgum genotype (Cunningham 1989, Rieckermann 1994). These two authors also succeeded in rooting juvenile stem cuttings in high percentages.

Limited attention and research has been focused on identifying stock quality characteristics for seedlings of sweetgum and for that matter, seedlings of hardwood species, in general. However, it is commonly accepted that hardwood seedlings planted as reforestation stock should be large enough to compete with native vegetation (South 1975). Likewise, it has been noted that sweetgum seedlings with large root collars and well developed roots improve survival and early growth (Kaszkurewicz and Keister 1975). In one sweetgum study, Johnson and McElwee (1967) reported that seedlings with root collar diameters $\geq$ 1cm were desired because of a related increase in survival and height growth after planting.

Rooted Cutting Challenges

Sweetgum presents challenges to developing rooted cutting production systems capable of producing high quality planting stock in one growing season. Rooted, semi-hardwood (SH) stem cuttings of sweetgum often fail to produce new shoot growth immediately following rooting. Shoot growth of these rooted cuttings occurs predominately in the second year, after the new plants are subjected to an over-wintering period. This delay in new shoot growth creates problems for foresters. Lacking new shoot growth, a 1-year-old rooted cutting would have difficulty out-competing other vegetation during plantation establishment due to its small size (Rieckermann 1994).

Sweetgum rooted cuttings with high quality root systems are also difficult to produce in one growing season (Cunningham 1989). Rooted cuttings with poorly developed root systems are at a disadvantage when out-planted and create difficulties during plantation establishment (Thompson 1985, Carlson and Miller 1990, Mexal and Landis 1990, Rieckermann 1994). A second season of nursery growth is often required to produce rooted cuttings large enough to successfully compete in the field. Unfortunately, the cost of a 2-year production cycle currently outweighs the genetic gain derived from the rooted cuttings (Lambeth et al. 1993), greatly reducing their usefulness in operational forestry.

Studies have investigated potential factors affecting shoot and root growth of newly rooted SH sweetgum stem cuttings. These factors include time of cutting collection (Cunningham 1989), origin of stem cutting material (terminal versus sub-terminal stem cuttings) (Rieckermann 1994), and nitrogen fertilizer following rooting (Rieckermann 1994).

Cunningham (1989) demonstrated that juvenile SH stem cuttings harvested from 4-year-old hedges produced more shoot growth and had more developed root systems when collected and rooted earlier in the growing season. Cunningham (1989) also compared field performance of sweetgum seedlings to rooted cuttings and concluded that smaller root volumes were potentially responsible for the lower survival rates of the rooted cuttings at the end of the first year.

Stem cutting material origin (terminal versus sub-terminal stem cuttings) significantly affected rooting percentage, shoot growth, and root growth for newly rooted sweetgum cuttings (Rieckermann 1994). Terminal SH stem cuttings rooted at higher percentages, while the sub-terminal stem cuttings produced rooted cuttings with greater shoot and root dry weights, shoot length, and percentage of cuttings with new shoot growth.

Application of nitrogen fertilizer (0 – 200 mg N/liter) also affected shoot and root growth for newly rooted sweetgum SH stem cuttings (Rieckermann 1994). Shoot dry weight increased with increasing rates of nitrogen. However, the duration of shoot growth was not affected by the nitrogen fertilizer rate applied. In the same study root growth decreased with application rates > 50 mg N/liter.

Hardwood Stem Cuttings

Successful rooting of hardwood (H) stem cuttings of deciduous species is common for a smaller range of species then for either softwood or semi-hardwood cuttings. Sweetgum stem cuttings in this particular stage of growth are characterized by bark that has a brownish, woody appearance. Deciduous tree species rooted successfully at this stage of growth include *Betula* spp. (Vaclav 1974), *Platanus* x *Acerifolia* (London Plane Tree) (MacDonald 1984)**,** *Platanus occidentalis* L. (American sycamore), *Salix* spp. (Hook et al. 1974), *Populus deltoids* Bartr. (eastern cottonwood) (McKnight 1970), and *Quercus rubra* L. (northern red oak) (Thinmann and DeLisle 1939). Rooting H stem cuttings can reduce the summer propagation workload and provide a "back-up" in the event of failure during summer propagation (MacDonald 1986). H stem cuttings can also be stuck directly in the field for a few species, including poplar (*Populus)* and willow (*Salix)* (Wilkinson et al. 1981). Shoot growth sometimes occurs following rooting of H stem cuttings. This could be of particular importance for woody tree species, such as sweetgum, that have difficulty producing new shoots following rooting when propagated from softwood or semi-hardwood stem cuttings.

Results of rooting trials with H stem cuttings of sweetgum have varied. In one rooted cutting trial, sweetgum H stem cuttings collected from the tops of 1-year-old seedlings were rooted successfully and produced considerable new shoot growth (John Frampton, NC State Univ. Christmas Tree Gen. Progr., personal communication). However, Cunningham (1989) rooted only 11 out of 720 H stem cuttings collected in November from hedged stock plants. None of the H stem cuttings Cunningham collected in March rooted.

Several factors affect success rooting of H stem cuttings including time of collection, handling and storage practices, and the rooting environment (MacDonald 1986). The best time for H stem cutting collection varies and optimal collection time should be investigated for each species and clone. Handling H stem cuttings correctly, following collection, is also crucial for high rooting percentages. H stem cuttings not processed immediately are typically stored at 3^0C in vertically oriented bundles of 20 to 30 cuttings (MacDonald 1986). Slightly moistened peat or some other material is placed at the bottom of the storage container to limit cutting desiccation by providing a source of humidity.

After sticking, every effort should be employed to limit budbreak prior to root formation. Following budbreak, carbohydrate reserves are usually redirected to the emerging leaves as opposed to the basal portion of the stem where rooting is to occur (Dirr and Heuser 1987). When leaves emerge prior to root formation the cutting often dies. Maintaining warm temperatures below ground and cool temperatures above ground may encourage root formation without promoting new shoot growth. Time of sticking, shade, ventilation, and bottom heat are sometimes successfully used to simulate these environmental conditions (MacDonald 1986, Dirr and Heuser 1987). Irrigation is also utilized to prevent excessive moisture loss from the cuttings. Depth of sticking can further effect cutting desiccation. The greatest need for irrigation is when budbreak occurs on cuttings with little or no roots (MacDonald 1986).

Semi-Hardwood Stem Cuttings

Sweetgum stem cuttings collected in a semi-hardwood (SH) transitional stage of development typically root at high percentages than sweetgum stem cuttings collected in other stages of development (Cunningham 1989). Sweetgum stem cuttings in a semi-hardwood stage of growth are characterized by bark that is in transition from a greenish succulent appearance, to a brownish, woody appearance. As this change in appearance occurs, the stem of the cutting also becomes less elastic. Sweetgum shoots typically reach this stage of growth in the middle of June in Raleigh, North Carolina (35.8^0 N, 110 m).

Handling and preparation of SH sweetgum stem cuttings following collection is critical for obtaining optimal rooting. When cuttings cannot be stuck immediately following collection, they are stored at low temperatures (above freezing) in a humid environment to reduce moisture stress (MacDonald 1986). Rieckermann (1999) stored SH sweetgum stem cuttings for 30 days in a cooler at 4^0C without adversely impacting rooting. Cutting length can also affect rooting percentages. In one study, terminal 15-cm sweetgum stem cuttings collected from hedged stock plants in late

May to early June rooted at significantly higher percentages than terminal 5- and 10-cm stem cuttings collected at the same time (Robison et al. 1999).

Leaf trimming, after stem cutting excision, is practiced commonly to reduce transpirational loss, facilitate sticking, conserve space required for propagation (MacDonald 1986) and increase air flow among stem cuttings to reduce the likelihood of disease (Rieckermann 1999). Leaf trimming has also been shown to increase rooting percentages for other hardwood tree species, including, *Eucalyptus camaldulensis* (Geary and Harding 1984). Though leaf trimming did not increase rooting percentages among sweetgum cuttings, Rieckermann (1999) found that rooted cuttings with trimmed leaves produced less root growth and more shoot growth than non-trimmed cuttings.

Exogenously applied rooting hormones and plant growth regulators, such as indole-3-butyric acid (IBA) and naphthalene acetic acid (NAA), respectively, are commonly applied to the bases of stem cuttings prior to sticking to increase overall rooting percentages, hasten root initiation, increase the number and quality of roots, and increase uniformity in rooting (MacDonald 1986, Hartmann et al. 2002). Though rooting hormones are not required for rooting excised sweetgum stem cuttings collected in May, those treated with up to a 1% concentration of IBA rooted slightly better than those left untreated (Cunningham 1989). Further, Cunningham found that excised sweetgum stem cuttings treated with IBA in talc produced many more roots than when treated with IBA in ethanol.

Choosing the correct rooting medium is another important aspect of a successful rooted cutting production system. There is no ideal rooting medium for all cuttings, species, and environmental conditions. However, rooting medium should be clean, moist, well aerated, and well drained (Hartmann et al. 2002). Rooting medium provides support for the cutting during rooting, moisture to the cutting, allows for air exchange at the base of the cutting, and creates a dark opaque environment for the inserted portion of the cutting to generate roots (Hartmann et al. 2002). Cost and availability of the medium components are additional considerations. Sweetgum rooted cuttings performed equally well when rooted in a medium mixture (by volume) of 1 peat: 2 perlite: 2 vermiculite, 1 peat: 1 perlite, 1 peat: 1 sand, and 1 peat: 1 bark (Rieckermann 1999).

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Feasibility and morphological effect of potential production systems for rooted stem cuttings of sweetgum (*Liquidambar styraciflua* L).

[1] Matthew H.Gocke, [1] Daniel J. Robison, [1] Barry Goldfarb, [1] L. J. Frampton, and [2] Frank Blazich

[1] Dept. of Forestry and Environmental Resources, North Carolina State University, Raleigh, NC 27695, USA
[2] Dept. of Horticultural Science, North Carolina State University, Raleigh, NC 27695, USA

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Email address: mhgocke@gmail.com

Abstract

Production systems resulting in high quality reforestation stock are needed to realize the benefits of sweetgum rooted cuttings representing select genotypes. Five sweetgum rooted cutting production systems were evaluated with three clones. The first experiment was conducted from June 2000 to April 2001 and evaluated four rooted cutting production systems for semi-hardwood cuttings: two transplant plug systems, one containerized system (CT) and one direct-stick system, in which stem cuttings were stuck directly into an outdoor nursery bed for rooting and growth. The second experiment was conducted from January 2001 to April 2002 and evaluated hardwood sweetgum stem cuttings in an outdoor direct-stick system. Cuttings used in this experiment were collected in January or March to investigate the effect of collection time on rooting and subsequent growth. The third experiment was conducted from June 2001 to April 2002 and evaluated rooted cutting production systems for semi-hardwood cuttings in a transplant and a containerized system. The plug systems used Jiffy Forestry Peat Pellets (JF) and Grow-Tech Rooting Sponges (GT).

Sweetgum semi-hardwood stem cuttings stuck in the JF system in Experiment 1 rooted at significantly higher percentages (97 vs. 82%) than the GT system and survived at significantly higher rates (82%) than the GT (59%), the CT (59%), and the direct-stick (35.5%) production systems. Significant differences were also present between the two clones tested in Experiment 1. Root dry weight was significantly

greater for the stem cuttings rooted in the CT (0.33 g) system than the direct-stick (0.23 g) and JF (0.17 g) production systems. In Experiment 2, hardwood stem cuttings stuck directly into an outdoor nursery bed survived at approximately 16%, averaging all treatments and clones. Clone survival rates changed according to collection time and resulted in significant collection time by clone interactions, with the highest survival for one clone collected in January (55.5%). Root dry weight and the percentage of rooted cuttings with newly elongated shoots were significantly greater among those stem cuttings collected in January (1.48 g and 6.3%) as opposed to March (0.33 g and 1.1%). In Experiment 3, the CT system semi-hardwood stem cuttings rooted at significantly higher percentages (88.5 vs. 63%) and survived at significantly higher rates (84.1 vs. 53.3%) than those stuck in the JF system.

All the production systems evaluated in this study demonstrated potential for producing sweetgum rooted cuttings. Morphological measurements varied among the different planting stock produced by each production system. The JF transplant, the GT transplant, and the CT production systems demonstrated the potential for producing a large number of rooted cuttings with high rates of survival and large root systems. The semi-hardwood direct-stick system proved more sensitive than the other systems tested, and requires a back-up irrigation system and a secondary power source to be effective. The hardwood direct-stick production system was the only system to produce rooted cuttings exhibiting substantial shoot growth during the first growing season. Some of these cuttings also developed extensive root systems, but survival was low.

Introduction

Liquidambar styraciflua L. (sweetgum), a member of the Hamamelidaceae, is an important commercial hardwood species in the United States and is used for a variety of wood and paper products. Though most commonly harvested from naturally regenerated stands, a smaller percentage of this species is harvested from artificially regenerated stands. Sweetgum reforestation stock is almost always propagated by seed (Sutter 1989) and has been used in forest plantings for several decades. Seed of the species are commonly germinated and grown outdoors in nursery beds to obtain 1-0 bare-root planting stock (Kaszkurewicz and Keister 1975, McNabb 2001). Seedlings are also produced in containers as 1-0 containerized planting stock.

In plantations, sweetgum has preformed variably. On good sites with weed control it can grow rapidly in plantation systems, while, under drier conditions, its growth rate can be very slow. Sweetgum growth is highly genetically controlled (Cunningham 1989). Genetic improvement and clonally propagation may be important in deploying it as a plantation species. One promising method for clonally propagating sweetgum is by means of rooting juvenile stem cuttings.

For many tree species, stem cuttings collected from juvenile shoots root more easily than from shoots in an adult phase of growth and exhibit less abnormal subsequent growth habits (Teclaw and Isebrands 1987, Hamann 1998, Hartmann et al. 2002). Root suckers (Johnson 1964, Farmer 1966), epicormic branches (Kormanik and Brown 1973, Kormanik and Brown 1974), and stump sprouts (Johnson 1964, Cunningham 1989) represent forms of juvenile shoot growth in sweetgum. Stem cuttings can be collected from these shoots and successfully rooted. Unfortunately, a limited number of rooted cuttings can be obtained annually from each of these approaches. If large-scale clonal propagation of selected sweetgum genotypes is desired, techniques for providing a large number of juvenile stem cuttings for rooted cutting propagation is required (Hamann 1998).

Pruning slows down the process of maturation in several tree species by stimulating juvenile shoot growth (Hartmann et al. 2002). Pruning and serial propagation have been used successfully to produce stock plant hedges capable of providing numerous juvenile stem cuttings from a single sweetgum genotype (Cunningham 1989, Rieckermann 1994).

High quality rooted cuttings are required to realize genetic gain and provide uniform response following out-planting. Like seedlings, rooted cuttings must conform to certain morphological and physiological standards to ensure successful field performance (Ritchie et al. 1993). Additionally, production of high quality rooted cuttings must be accomplished in an economically viable manner, applicable to a scale appropriate for large field plantings.

Limited attention and research have been focused on identifying stock quality characteristics for sweetgum seedlings and, for that matter, seedlings of hardwood species, in general. However, it is commonly accepted that hardwood seedlings planted as reforestation stock should be large enough to compete with native vegetation (South 1975). Likewise, it has been noted that sweetgum seedlings with large root collars and well developed roots improve survival and early growth (Kaszkurewicz and Keister 1975). In one sweetgum study, Johnson and McElwee (1967) reported that seedlings with root collar diameters ≥ 1 cm were desired because of a related increase in survival and height growth after planting.

Nursery grading systems measure seedling morphology to predict field survival and growth (Wakeley 1954, Thompson 1985, Wilder-Ayers and Toliver 1987, Mexal and Landis 1990). Several studies support the predictive value of these morphological measurements for a variety of forest tree species including Douglas-fir [*Pseudotsuga menziesii* (Mirb.) Franc.], European larch (*Larix decidua* Mill), [Norway spruce (*Picea abies* (L.) Karst.] (Mason 1991); slash pine (*Pinus elliottii* Engelm.) (Wakeley 1969); white spruce [*Picea glauca* (Moench) Voss] (Mullin and Svaton 1972); loblolly pine (Wakeley 1954, Sluder 1979, McGilvray and Barnett 1982, Rowan 1987); and Sitka spruce [*Picea sitchensis* (Bong.) Carr.] (South and Mason 1993)

Measured morphological characteristics include root collar diameter (RCD) (South and Mexal 1984, Thompson 1985, Mexal and Landis 1990), shoot height (Kaszkurewicz and Keister 1975, Sluder 1979, McGilvray and Barnett 1982, South and Mason 1993), root weight (Thompson 1985, Blake et al. 1989, Long and Carrier 1993, Carlson and Miller 1990), and shoot to root weight ratio (S: R ratio) (Thompson 1985, Mohammed 1997, Tanaka 1997). Specific morphological grading standards often differ among species, provenance, stock type, and operational requirement (Mexal and Landis 1990). These standards are often defined by field observations and incorporate morphological requirements with practical planting limitations, such as the size of a root system that can be properly planted with ease

(Carlson and Miller 1990). Furthermore, because no single morphological characteristic provides a sufficient assessment of stock quality, a combination of characteristics is often used to provide the best predictive results (Cleary et al. 1978, Menzies and Arnott 1992).

The relationship between seedling performance and morphology is also strongly influenced by the planting site (South 1985). Factors such as available soil moisture, competing vegetation, desiccating wind, and soil temperatures can strongly affect seedling performance (Mexal and South 1984, Carlson 1986, Grossnickle 1988, Stape et al. 2001). Successful reforestation efforts take into account the interaction of morphology and environment when matching planting stock with planting site (Zwolinski et al. 1994).

Sweetgum growth habits following rooting present challenges to the development of a propagation system capable of producing a large number of high quality planting stock in one growing season. Rooted, semi-hardwood sweetgum stem cuttings often fail to produce new shoot growth immediately following rooting. Shoot growth for these cuttings occurs predominately in the second year, after the new plants are subjected to an over-wintering dormancy period. This delay in new shoot growth creates problems for foresters. Lacking new shoot growth, a 1-year-old rooted cutting would have difficulty out-competing other vegetation during plantation establishment due to its smaller size (Rieckermann 1994).

Sweetgum rooted cuttings with high quality root systems are also difficult to produce in one growing season (Cunningham 1989). Rooted cuttings with poorly developed root systems are at a disadvantage when out-planted and create difficulties during plantation establishment (Thompson 1985, Carlson and Miller 1990, Mexal and Landis 1990, Rieckermann 1994). A second season of nursery growth is often required to produce rooted cuttings large enough to successfully compete in the field. Unfortunately, the cost of a 2-year production cycle currently outweighs the genetic gain derived from the rooted cuttings (Lambeth et al. 1993), greatly reducing their usefulness in operational forestry.

Rooting hardwood cuttings of sweetgum, however, may offer the potential to produce rooted cuttings of sufficient size to be out-planted after one growing season in the nursery. Shoot growth may occur following rooting for hardwood sweetgum stem cuttings, allowing for additional vegetative growth. Successful rooting of hardwood stem cuttings is common for a smaller range of species than for either softwood or semi-hardwood cuttings (MacDonald 1986).

Several production systems have been developed to produce a variety of stock suitable for reforestation. Plants cultured as seedlings, rooted cuttings, *in vitro* plantlets, and seedlings derived from somatic embryos are grown typically under one of three production systems; 1) in outdoor nursery beds as bare-root stock, 2) in containers as containerized stock, or 3) initially in containers or plugs, and then later transplanted to an outdoor nursery bed as transplant stock (Stein et al. 1975, Hahn 1984, Menzies and Arnott 1992). Each production system offers advantages and disadvantages to reforestation efforts (Menzies and Arnott 1992).

Study Objectives

The objective of this study was to identify rooted cutting production systems that produce high quality sweetgum reforestation stock. Five potential production systems and three sweetgum clones were evaluated in three rooted cutting experiments conducted between 2000 and 2002 at North Carolina State University in Raleigh, NC.

The first experiment was conducted from June 2000 to April 2001 and evaluated four rooted cutting production systems for semi-hardwood stem (SH) cuttings. The second experiment was conducted from January 2001 to April 2002 and evaluated hardwood (H) sweetgum stem cuttings in a direct-stick (DS) production system (H-DS). The third experiment was conducted from June 2001 to April 2002 and as in the first experiment evaluated rooted cutting production systems for SH stem cuttings.

Materials and Methods

Stock Plants, Cutting Material and Rooting Procedures

For all three experiments, sweetgum stem cuttings were collected from annually hedged stock plants maintained by the NC State University Hardwood Research Cooperative (HRC). Stem cuttings collected for the first two experiments represented shoot growth during the third year. Cuttings collected for the third experiment originated from the same hedges, but from shoot growth during the fourth year.

The stock plants were obtained from a Union Camp Corporation mother tree study and progeny test in Virginia (Mike Cunningham, personal communication). The seedlings were pruned 10 cm above the root collar with bypass pruners. New shoots growing from these pruned hedges were collected and rooted in May 1998 for the purpose of bulking-up the number of stock plants of each clone. Ramets of each clone were planted at the HRC - Reedy Creek field site in Raleigh, NC, on April 25-31, 2000 and organized into rows according to clone. The planting site was ripped, amended with topsoil and hardwood mulch, fertilized with diammonium phosphate (equivalent to 560 kg/ha), and tilled the previous winter. The hedges were spaced 1.2 m between rows (clones) and 0.3 m among individual ramets of the same clone. The stock plants were pruned 10 cm above the root collar following planting. The clones were designated as UC1, UC2, and 2074. Clones UC1 and 2074 were tested in Experiment 1, all three clones were tested in Experiment 2, and Clones UC1 and UC2 were tested in Experiment 3. Following each collection, the stock plants were re-pruned 2 cm above the base of the current season's growth.

Outdoor Rooting and Growth Facilities

A 38 m long x 1.2 m wide x 0.5 m tall raised nursery bed, built with pressure treated lumber, was constructed for use in all three experiments. The rectangular structure was placed on top of a thin layer of gravel and the inside was lined with weed mat. The bed was filled with soil from the coastal plain of North Carolina with a loamy sand soil texture comprised of 85% sand, 12.2% silt, and 2.8% clay. The loamy sand had an initial pH of 5.1. The pH of the nursery bed irrigation water was 5.7 during the first experiment and 5.9 during the second and third experiments. A shade cloth, providing 50% ambient light, covered the SH-DS cuttings during the initial 16 weeks of rooting. Pine bark mulch was placed on the surface of the nursery bed during Experiments 2 and 3 to better retain soil moisture and to reduce compaction caused by repeated overhead irrigation as observed during Experiment 1.

The nursery bed was equipped with an overhead sprinkler system constructed of PVC piping equipped with overhead NaanDan™ "Water and Sprinkling" nozzles (Kibbutz Naan, Naan, Israel 76829) with a misting flow rate of $41.6\ L \cdot h^{-1}$. During the first, and at the end of the second experiment, a fixed-cycle Rainbird ™ Controller (Model MIC-12 (H0), Rainbird Corporation, Glendora, CA), regulated misting frequency and duration (Appendices I and II). The fixed-cycle controller was replaced with a variable-cycle control system from Davis Engineering (Solar 6A Misting Controller, Davis Engineering, Winnetka, CA), during the rooting phase of the second experiment. The frequency of irrigation was automatically adjusted according to the amount of sunlight detected by a light sensor connected to the controller. Sunlight was measured in solar units. One solar unit equaled 0.02 moles m^{-2}. On a clear sunny day at noon in June, 30 seconds of full sun equaled approximately one unit, while on a completely overcast day at noon, one unit equaled approximately three-minutes. The number of solar units required to accumulate before triggering the misting regime increased over time to correspond with the decreasing misting requirements of the cuttings as they formed adventitious roots (Appendix II). No irrigation was applied at night.

The same Davis Engineering controller was used during the rooting phase of the third experiment. However, instead of regulating misting frequency and duration by accumulating solar units, the machine regulated misting according to manually programmed periods of time (minutes and seconds) (Appendix II).

Indoor Rooting Facilities

Cuttings stuck in the GT, JF, and CT systems were rooted in a clear polyethylene-covered greenhouse in natural light prior to being moved to the outdoor bed. Heating and cooling systems were set to maintain the day/night air temperature at 23-26/20-23 ^{0} C. Cuttings were intermittently misted at a variable frequency related inversely to the relative humidity (RH) in the greenhouse. Variable frequencies were defined by designating minimum (60% RH) and maximum (99% RH) off times between misting applications. Off times for intermediate humidity values were calculated based on a linear function. The minimum and maximum off times varied according to the time of day. For the periods from 0600 to 0900 h, the minimum and maximum off times were 10 and 35 min, respectively. For the periods from 0900 to 1800 h, 1800 to 2100 h, and 2100 to 0600 h, minimum and maximum off times were 8 and 24 min, 10 and 40 min, and 60 and 240 min, respectively.

An environmental management software package (Q-Com, Irvine, CA) calculated mist frequency and triggered a traveling gantry (boom) system (ITS, McConkey, Mt. Puyallup, WA) to apply mist. Misting frequency (number of boom passes) was identical for each treatment within an experimental year, but may have differed due to environmental conditions between experimental years. During each experiment, boom traveling speed was altered according to the length of the rooting period. For the two experiments that included indoor rooting, the amount of mist per boom pass was decreased every four weeks for the three-month rooting period. During the first four weeks the cuttings received 102 ml m^{-2}. During the second and third four week time periods the cuttings received 75 and 61 ml m^{-2}, respectively (LeBude et al. 2004). The pH of the greenhouse irrigation water was 6.4 during the first experiment and 6.2 during the third experiment.

Fertilizer

For all three experiments, fertilization was initiated eight weeks after sticking with a water-soluble, all-purpose 20-20-20 fertilizer, (Peters Professional 20-20-20 with micronutrients, J.R. Peters, Inc./J.R. Peters Laboratory, Allentown, PA) (Appendices III and IV). The fertilizer was applied over the cuttings in this form with an injection sprayer.

Experiment 1

Stem cuttings for the first experiment were collected from shoots transitioning from softwood to semi-hardwood (SH). For sweetgum growing in Raleigh, NC ($35^0$5’ N, 78^0 4’W, 110 m) this typically occurs in June. The bark begins to change from green to brown as it transitions to a woody appearance and the stem becomes less elastic. Stem cuttings, 16-cm in length, were collected June 8-9, 2000. Stem cutting sections represented both the shoot’s terminal 16-cm (including the terminal bud) and its second 16-cm or sub-terminal section.

Immediately following collection, the leaves and petioles present on the lower half of each stem cutting were removed with bypass pruners. The stem cuttings were then oriented vertically in a plastic storage container with the basal portion submerged in approximately 5 cm of water and placed in a cooler at 5^0 C for approximately 1 week until the time of sticking. Between June 16 - 22, 2000, these SH stem cuttings were removed from storage, the base of each was re-cut and dipped 2 cm into 0.8% IBA talc (Hormodin #3, Olympic Horticultural Products Company, P.O. Box 230, Mainland, PA), and stuck in one of four production systems.

The four production systems evaluated in Experiment 1 were; 1) a transplant system in which SH stem cuttings were rooted in a greenhouse in “72S Flexiplug™” Grow-Tech™ (GT) Rooting Sponges (Grow-Tech Inc., San Juan Bautista, CA) and then transplanted at either 11 or 13 weeks to an outdoor nursery bed for subsequent growth (GT-11 and GT-13); 2) a transplant system in which SH stem cuttings were rooted in a greenhouse in Jiffy™ “30-mm” Forestry Peat Pellets (JF) (Jiffy Products Ltd., Shippagan, N.B., Canada) and then transplanted at either 11 or 13 weeks to an outdoor nursery bed for subsequent growth (JF-11 and JF-13); 3) a containerized system (CT), in which SH stem cuttings were rooted in a greenhouse in medium [1 peat: 1 perlite (v/v)] filled growing tubes (Ray Leach SuperCells™ , Stuewe and Sons, Inc. Corvallis, OR) for 12 weeks and then moved outdoors, but retained in the same tubes for subsequent growth (CT-12); and 4) a direct-stick system (DS), in which SH stem cuttings were stuck directly into the outdoor nursery bed for rooting and growth (SH-DS).

Rooting media, sticking depth, and the cutting density varied according to the production system tested. The base of each stem cutting was inserted 2 cm into the pre-formed holes of either the GT rooting sponges or the JF peat pellets. The volumes of the JF pellet and GT sponge were approximately 60 and 74 ml, respectively. The GT rooting sponges and JF peat pellets were placed in Winstrip “WS 72” trays (Winstrip, Inc., 1856 Jeffrey Rd., Fletcher, NC 28732) at a density of 570 m^{-2}.

The GT and JF system rooted cuttings were transplanted by hand into the nursery bed (roots intact in the plugs/pellets) on September 1 and 15, 2000, following 11 and 13 weeks of rooting, respectively, at a density of 172 m^{-2}. Rooting percentage was evaluated at the time of transplant for both systems. A stem cutting was considered rooted if at least one root could be observed emerging from the rooting sponge or peat pellet, or in the case of no visible emerging root, at least one root (> 1 cm) was observed when the sponge or pellet was partly torn open.

SH stem cuttings rooted in the CT system were inserted 2 cm deep into medium in the growing tubes. The tubes were placed in trays in the greenhouse at a density of 527 m^{-2}. The containers remained in the trays but the density was reduced to 269 m^{-2}. This spacing was achieved by placing the tubes in every other hole when the cuttings were moved outside, September 8, 2000, to the nursery bed in spaces without soil after 12 weeks of rooting. Stem cuttings rooted in the outdoor nursery bed were inserted 5 cm into the nursery bed soil at a density of 172 m^{-2}. Extra rooted cuttings from the greenhouse comprised the border rows in the outdoor nursery bed and were also used to fill any vacancies in treatment plots.

The SH-DS stem cuttings were arranged in a split-plot design with 6 blocks, each containing 5 treatments organized in random order (whole plots). Within each treatment plot, there were two rows of ten stem cuttings. Each row represented either Clone UC1 or 2074 (subplots). During rooting in the greenhouse, the GT, JF, and CT system stem cuttings were arranged in a complete block design with 6 blocks across the length of one bench. Fifteen stem cuttings per clone per block were stuck in each of the five treatments rooted in the greenhouse in this experiment. When the rooted cuttings were moved outdoors, they were organized in a manner identical to the SH-DS stem cuttings outlined above. The stem cuttings were fertilized at a rate of 50 ppm

N at the beginning of weeks 8, 10, and 12, and at a rate of 100 ppm N at the beginning of weeks 13, 15, and 17. The total amount of nitrogen applied during this experiment was equivalent to 55 kg N/ha (Appendix III). The SH sweetgum rooted cuttings were destructively harvested on April 1, 2001 and evaluated for over-wintering survival, first year shoot growth, and root dry weight. Survival rate was considered a partial indication of rooting for the SH- DS and CT system stem cuttings due to the difficulty of conducting non-destructive observations of adventitious root formation.

Experiment 2

Hardwood (H) stem cuttings were collected on January 18 and March 20, 2001. Only terminal shoots > 16-cm were selected. The H stem cuttings were then bundled by clone and stored vertically in a plastic storage container on top of saturated Oasis™ floriculture blocks (Oasis, Smithers-Oasis, Kent, OH) to prevent desiccation. Holes in the bottom of the storage container allowed drainage to discourage excess moisture. The cuttings and container were kept in a cooler at 2^{0} C until time of sticking.

At the time of sticking, April 20, 2001, the basal portion of each H stem cutting was re-cut and dipped 2 cm into a solution of indole-3-butyric-acid (IBA) and naphthalene acetic acid (NAA) at a dilution rate of 5 parts water to 1 part Dip'N Grow™ (Astoria-Pacific, Inc., Clackamas, OR) rooting compound (1% IBA and 0.5% NAA) for 5 seconds. The stem cuttings were then inserted 5 cm into the outdoor nursery bed soil at a density of 172 m^{-2}. The H stem cuttings were arranged in the outdoor nursery bed in a split-plot design with 6 blocks, each containing two treatments organized randomly (whole plots). Within each treatment plot, there were 5 rows of 10 stem cuttings. Fifteen stem cuttings (subplots) represented each of the three clones tested (UC1, UC2, and 2074). Fertilizer applied during Experiment 2 was 50 ppm N at the beginning of weeks 9, 11, and 13, and 100 ppm N at the beginning of weeks 15, 17, 19, 21, and 23. The total amount of nitrogen applied during Experiment 2 was the equivalent of 105 kg N/ha (Apendix IV). The H rooted cuttings were destructively harvested in April 2002 and evaluated for over-wintering survival, percent of newly elongated shoots (NES) following rooting during the first year, and root dry weight.

Experiment 3

The semi-hardwood (SH) stem cuttings evaluated in the third experiment were collected during the transition from softwood to semi-hardwood, as in Experiment 1. Terminal 16 cm stem cuttings were collected and stuck on June 18, 2001. The stem cuttings were prepared and stuck in the same manner as described in Experiment 1 for the JF and CT systems, with the following exceptions: 1) stem cuttings in Experiment 3 were stuck on the same day as collection; 2) only Jiffy Forestry Peat Pellets (JF) were utilized in the transplant treatments; and 3) no direct-stick (DS) system was tested. Both the JF and CT rooted cuttings were moved on August 27, 2001, to the outdoor nursery bed, 9 weeks after initial sticking.

During rooting in the greenhouse, the JF and CT system stem cuttings were arranged in a complete block design with 6 blocks across the length of one bench. Fifteen stem cuttings per clone per block were stuck in both of the treatments tested in this experiment. When the rooted cuttings were moved outdoors to the nursery bed, they were organized into a split-plot design with 5 blocks; each containing two treatments organized randomly (whole plots). Within each treatment plot there were four rows of five rooted cuttings. Ten rooted cuttings (subplots) represented each of the clones (UC1 and UC2) tested.

The JF system stem cuttings were fertilized at 50 ppm N at the beginning of weeks 8 and 10, and at 100 ppm N at the beginning of weeks 12 and 14. The total amount of nitrogen applied to the JF stem cuttings during this experiment was the equivalent of 55 kg N/ha. The CT stem cuttings were fertilized at 50 ppm N at the beginning of weeks 8 and 10, and 50 ppm N twice during weeks 12 and 14. The total amount of nitrogen applied to the CT stem cuttings in Experiment 3 was the equivalent of 55 kg N/ha (Appendix IV).

Rooting percentage was determined for both production systems evaluated in Experiment 3. Rooting in the CT system was determined by gently pulling on the stem cuttings. If the stem cuttings remained firmly in the medium they were considered rooted. Rooting percentages for JF stem cuttings were determined by visual means or by carefully pulling open the pellet or sponge to determine the presence or absence of newly formed roots. All rooted cuttings produced in Experiment 3 were destructively harvested in April 2002 and evaluated for over-wintering survival, first year shoot growth, and root dry weight.

Statistical Analysis

Analyses of variance for the traits assessed in Experiments 1, 2, and 3 were conducted on plot means using the GLM procedure of the Statistical Analysis System (SAS, Inc., Cary, NC). All sources of variation were analyzed as fixed effects, and were considered significant when the probability of a greater calculated F-value was less than or equal to 0.05. Type III p-values and non-transformed least squares means are reported.

To improve variance homogeneity in Experiment 1, the arcsine (square root) transformation was used for survival, and the square root transformation for root dry weight in both models (Tables 1.2 and 2.2). In the second experiment, no transformations were necessary. For the third experiment, the arcsine transformation was used for rooting percentage and survival, while the square root transformation was used for root dry weight. Main effect means and interactions were evaluated for significance by conducting pair-wise comparisons.

Results

Rooting and Survival: Experiment 1

Sweetgum SH stem cuttings stuck in the JF transplant system rooted at significantly ($P=<0.0010$) higher percentages (97 vs. 82%) than the SH stem cuttings stuck in the GT transplant system (Table 1.2). Rooting percentage was not directly measured for the CT and SH-DS system stem cuttings. Survival was, therefore, considered a relative measure of rooting percentage for these two systems.

Mean survival for CT system rooted cuttings was 59%, for the SH-DS system 35.5%, for the JF system 82%, and for the GT system 59% (Table 1.2). Among these four production systems, rooted cuttings originating from the JF system survived at significantly (P=<0.0001) higher rates than the rooted cuttings produced in the other three systems (Table 1.2). Survival also differed significantly (P=<0.0001) for the two clones tested in Experiment 1 (Table 1.2). Across all production systems, rooted cuttings from Clone UC1 survived at significantly higher rates than rooted cuttings representing Clone 2074 (71 vs. 47%) (Table 1.2).

Within the JF and GT production systems, clone survival rates varied according to transplant time and resulted in significant (P=0.0378) transplant time by clone interactions (Table 2.2). When rooted cuttings were transplanted at the later time of 13 weeks, survival rates for Clone UC1 increased significantly, while no significant difference in survival between the two transplant times existed for Clone 2074.

Root Dry Weight: Experiment 1

Significant differences (P=0.0003) in root dry weight were detected among the SH-DS, JF, and CT production systems (Table 1.2). Root dry weight was significantly greater among the SH stem cuttings rooted in the CT system (0.33 g) than the DS (0.23 g), and the JF (0.17 g) systems (Table 1.2). There was no difference in root dry weight among the SH-DS and JF systems.

Clonal differences were also detected for root dry weight (Table 1.2). Rooted cuttings of Clone UC1 had a significantly (P=0.0077) greater root dry weight than rooted cuttings representing Clone 2074 (0.26 vs. 0.19 g) (Table 1.2). Clone UC1 also produced rooted cuttings in the JF system with greater average root dry weight (0.20 g) than Clone 2074 (0.15 g) (Table 2.2).

Survival: Experiment 2

The mean survival rate for all H stem cuttings stuck in Experiment 2 was 16%. Rooting percentage was not determined in this experiment and therefore, survival was considered a relative measure of rooting. Clone survival rates varied according to collection time and resulted in significant (P<0.0001) collection time by clone interactions (Table 3.2). The survival rate for Clone UC2 was significantly greater when H stem cuttings were collected in January. Survival rates for H stem cuttings of Clones UC1 and 2074 were not significantly altered by collection time (Table 3.2). Survival for each clone decreased steadily until approximately July 5. After this date survival rates stabilized and remained largely unchanged to the end of the rooting season (Figure 1.2).

Root Dry Weight and First Year Shoot Growth: Experiment 2

Mean root dry weight for H cuttings collected in January was significantly greater than for H cuttings collected in March (1.38 vs. 0.31 g) (Table 3.2). The percentage of rooted cuttings, per clone, that produced newly elongated shoots (NES) varied according to collection time and resulted in significant (P=0.0123) clone x collection time interactions. Percent NES was significantly greater for rooted H cuttings of Clone UC2 when collected in January. No similar significant decrease in NES was observed for Clones UC1 and 2074. In Experiment 2, new shoot growth averaged 18.4 cm and ranged from 1 to 60 cm.

Rooting and Survival: Experiment 3

Sweetgum SH stem cuttings stuck in the CT system rooted at significantly (P<0.0001) higher percentages (88.5 vs. 61%) and survived at significantly (P<0.0001) higher rates (85.5 vs. 53.5%) than those stuck in the JF system (Table 4.2).

Root Dry Weight: Experiment 3

Clone root dry weight measurements varied according to production system and resulted in significant (P=0.0002) system by clone interactions (Table 4.2). Rooted SH stem cuttings representing Clone UC1 produced significantly larger root dry weights when stuck in the JF system, while Clone UC2 produced rooted cuttings with significantly larger root dry weights in the CT system (Table 4.2).

Discussion

Rooting percentages, Survival rates, and Root Growth: Experiment 1

Rooted SH stem cuttings produced in the JF system survived at significantly higher rates than rooted SH cuttings produced in the other three systems tested. Rooted cuttings in the GT and CT system survived at intermediate rates, while those in the SH-DS system survived at the lowest rate. Since SH stem cuttings were able to root and survive in all of the systems tested, modifying environmental conditions for each system may result in higher survival for each of the systems. The underlying reason why survival varied among the systems tested was not investigated. However, several possibilities, supported by observation, may explain the differences in survival and provide clues as to how survival might be enhanced in the various systems.

For instance, differences in the moisture content of the JF peat pellets and GT rooting sponges may have been responsible for the difference in rooting between these two plug types. Rooting medium moisture content can significantly affect rooting percentages (Copes 1977). Excessive amounts of water can cause anaerobic conditions (Dirr and Heuser 1987) which can lead to poor rooting for stem cuttings of most woody plants. Both transplant systems received the same amount of mist throughout the rooting period. However, the GT sponges were observed to hold more water and to be more saturated than the JF pellets. Reducing mist frequency for the GT sponges might lower moisture content and encouraged greater root formation. Furthermore, the individual spaces in the Winstrip™ trays used to hold the two plug types during rooting were approximately the same size and shape as the GT sponges. This resulted in a tight fit. The rounded shape of the JF pellets created a looser fit and may have provided greater air flow and subsequently less saturated conditions within the pellets.

Plug size can also affect adventitious root formation. LeBude et al. (1999) observed variation in rooting for loblolly pine stem cuttings stuck in various sizes of JF pellets. Differences were also observed between two JF pellet sizes evaluated in an earlier sweetgum rooted cutting experiment (Gocke 2000). Only one size for each plug type, JF 30 cm and GT 72R, was tested in Experiment 1 (60ml and 74 ml), respectively. Perhaps the optimal plug size for sweetgum SH stem cuttings stuck in the JF and GT system differ.

Two clones were evaluated in Experiment 1. Results suggest variation in rooting ability among genotypes of sweetgum. However, because only two genotypes were tested, it is difficult to interpret these results in terms the entire sweetgum population. Even so, variation in rooting ability has been reported among clones of both angiosperms and gymnosperms (Hassig and Riemenschneider 1988), and Cunningham (1989) and Rieckermann (1999) have previously reported clonal differences in sweetgum rooting ability. Cunningham (1989) found sweetgum clone rooting averages ranging from 41 to 100%. Selecting clones based on their ability to form adventitious roots may help optimize rooting and survival for sweetgum stem cuttings.

Over-wintering survival differed significantly for the two clones in Experiment 1. For all four of the production systems evaluated, Clone UC1 survived at significantly higher rates than Clone 2074. Clone survival within the JF and GT transplant systems, was further affected by transplant time. However, because clone did not significantly affect rooting percentage within these transplant systems (P=0.1134) (Table 1.2), there must have been other factors, such as root growth, responsible for differences in survival reported among clones. Clone UC1 had significantly larger root dry weights than Clone 2074 and may be genetically predisposed to early root formation, more vigorous root growth following adventitious root formation, or have some of the traits which promote good root development and survival.

CT system survival, 59%, was lower than expected in Experiment 1 given that in a previous study SH sweetgum stem cuttings stuck in growing tubes and placed in greenhouse conditions similar to those in Experiment 1, rooted at 79% (Cunningham 1989). CT system rooting percentages were not determined during Experiment 1. However, after a 12 week rooting period, few roots had appeared at the base of the containers. The appearance of roots growing out of the base of rooting containers is usually a fail-safe indication of rooting. Perhaps a rooting period longer than 12 weeks and/or an extended acclimation period in a low stress environment might encourage additional root development and ensure higher rates of survival.

Survival rates for the CT system rooted cuttings were also potential influenced by cultural treatment upon transfer to the nursery bed. Outside, in full sun, the containerized medium dried more quickly than the mineral soil of the nursery bed. Although the CT system rooted cuttings were watered every other day, insufficient soil moisture may have affected the survival of these rooted cuttings; particularly those cuttings with poor root development. Plant growth is the first process to be hindered by insufficient soil moisture (MacDonald 1984), and photosynthesis rates may be drastically reduced by even relatively small soil moisture deficits (Lavender 1984). However, the CT system rooted cuttings produced greater root dry weight than rooted cuttings from the SH-DS and JF systems. Modifying the post-rooting irrigation regime to prevent periodically dry conditions in the CT system should increase survival, and/or root dry weight, as well as rate of development.

Poor rooted cutting survival rates for the SH-DS system were likely the result of a single irrigation failure. On August 7, 2000 the watering system was inadvertently shut off from 10:00 AM to 4:30 PM. The high temperature that day was 35 ^{0}C. Although the shade cloth (50%) prevented some damage, a large portion of the stem cuttings exhibited wilting and/or scorching the following day. Wide-scale mortality was evident by the following week. Rooting in this system was not apparent when visually evaluated on August 1st. The 35.5% of SH-DS system cuttings that did survive probably had already formed some adventitious roots and were able to survive the relatively long interruption in mist. The DS system, and other systems, would benefit from several back-up systems to protect sensitive SH stem cuttings. Useful back-up systems include; auxiliary irrigation systems, a secondary source of power, and an alarm system to notify supervisors of potential problems.

Shoot Growth Following Rooting; Experiment 1

No substantial shoot growth occurred following rooting for any of the rooted SH cuttings produced during Experiment 1, and root growth was generally less than optimal. Rooted cuttings with poorly developed roots often survive at lower rates when out-planted (Thompson 1985, Blake et al. 1989, Carlson and Miller 1990). For success in field planting, rooted cuttings must attain root and height growth at least equivalent to the standards set by bare-root forest tree nurseries. For bare-root sweetgum seedlings produced at the North Carolina Division of Forest Resources' seedling nursery located at the Goldsboro Forest Center (GFC) in Goldsboro, NC, given conventional planting techniques, those standards include RCD > 6.35 mm and root systems longer than 12.7 cm (Greg Pate, GFC, personal communication). Because of poor root and shoot development in the first year, two growing seasons are required to produce high quality sweetgum rooted cutting stock from SH stem cuttings.

Survival and Root dry weight; Experiment 2

Time of collection of hardwood stem cutting influences rooting percentages and survival rates, and is ultimately influenced itself by the environmental conditions and physiological status of the plant (MacDonald 1986, Dirr and Heuser 1987). Although collection time was an important factor in the survival of Clone UC2 in Experiment 2, it had little effect on the survival of the remaining two clones. A similar interaction between clone and collection time was reported by Wally et al. (1980) for H cuttings of pecan [*Carya illinoinensis* (Wangenh.) K. Koch.] Collection time also affected root dry weight for rooted H stem cuttings. H stem cuttings collected in January had significantly larger root dry weights than stem cuttings collected in March.

Collection of H stem cuttings generally corresponds with the time at which the stock plants are experiencing dormancy. Dormancy refers to the stage of growth, following active vegetative and reproductive growth, typified by reduced cellular activity (Capon 1990). Plants may continue to appear dormant, but in fact have entered a new physiological stage, quiescence, when all of its chilling hour requirements have been met but budbreak and active growth have not been stimulated by warm temperatures (Barry Goldfarb, personal communication). The January collection time for Clone UC2 may have represented a more fully dormant physiological state. H stem cuttings collected in late March were more likely to have received most of its chilling-hour requirements and may have been in a quiescent physiological state.

Perhaps collecting stem cuttings in a dormant state facilitates production system controls designed to delay budbreak, while encouraging root formation. Warm temperatures below ground and cool temperatures above ground can stimulate rooting while deterring budbreak. Budbreak prior to adequate root formation often results in mortality (MacDonald 1986, Dirr and Heuser 1987) as was observed, most consistently, for H stem cuttings collected in March in Experiment 2. However, others have observed that H stem cuttings root in higher percentages when collected in a quiescent state (John Frampton, personal communication).

Regardless of collection time and physiological state of growth, several approaches have been used to foster rooting of H stem cuttings of difficult to root species (MacDonald 1986). Heating devices designed to increase below ground temperatures, while cool temperatures are maintained above ground, have been successful for rooting cuttings stuck inside and outdoors. Altering the sticking date, shading the stem cuttings, and providing ventilation have also been used to control above ground temperatures during the initial rooting period (MacDonald 1986). Cunningham (1989) stuck H stem cuttings in containers placed in a greenhouse. Warm air temperatures in the greenhouse may have stimulated budbreak prior to adequate root formation, causing the large-scale mortality reported.

Storing H stem cuttings in a cold, humid environment, until the time of sticking, can also increase rooting percentages. A humid environment reduces the risk of desiccation during storage, while cold temperatures (just above freezing) allow the stem cuttings to receive an appropriate number of chilling hours without the chance of warm temperatures to stimulate respiration and new shoot growth. Experimenting with various rooting hormones, hormone concentrations, and the time of hormone application may also increase rooting and survival for H sweetgum stem cuttings.

A rooting hormone was applied to the base of the H stem cuttings at the time of sticking, which was up to three months following collection for some of the stem cuttings. Applying rooting hormones immediately following collection increased rooting percentages for H stem cuttings of several woody plant species including;

Cornus alba 'Elagantissima', *C. stolinifera* 'Flaveiramea', *Philadelphus* x *virginalis*, and *Sambucus canadensis* 'Aurea' (MacDonald 1986), and this may have been helpful for sweetgum.

Shoot Growth Following Rooting; Experiment 2

The percentage of rooted H cuttings with newly elongated shoot growth (NES) was significantly affected by collection time, with a significant decline in the percentage of rooted cuttings with NES from January to March. H stem cuttings collected in March resumed shoot growth prior to adventitious root formation. Die-back of early shoot growth on H stem cuttings was a good indication of rooting failure in Experiment 2.

NES length was highly variable for the rooted cuttings producing new shoot growth the first growing season. While most shoot growth was moderate for these rooted cuttings, a few cuttings exhibited exceptional growth. One such cutting grew 49 cm tall in the first growing season. This same cutting produced 11 primary roots and had a root collar diameter of 9.5 cm.

Experiment 3

Two rooted SH stem cutting production systems tested in Experiment 1 were repeated in Experiment 3. The Jiffy peat pellets (JF) were selected to represent a transplant production system, and were selected over the Grow-tech rooting sponges because of superior performance in rooting and survival in Experiment 1. However, no comparison has been made concerning subsequent growth between these two transplant medium types. The CT system was also selected to be repeated in Experiment 3, to compare a "pellet" type system to a more conventional containerized production system. Irrigation following transplant and transfer to the outdoor nursery bed was monitored closely in Experiment 3, with some adjustments specific to the requirements of each rooted cutting stock type.

Rooting and Survival Percentages: Experiment 3

Rooting percentages for semi-hardwood (SH) stem cuttings stuck in the JF system decreased by 35% between Experiments 1 and 3. Rooting percentages often vary from year to year for individual species and among genotypes within a species, and often depends on a multitude of factors. Clone 2074 was replaced with Clone UC2 in Experiment 3 and may have created a source of variation between experiments. A shorter amount of time allotted to rooting, prior to transplant, may have also been responsible for decreased rooting for the SH stem cuttings stuck in the JF system during Experiment 3.

Survival rates for rooted SH cuttings decreased by 28.5% between Experiments 1 and 3 in the JF transplant system. Although clonal selection in Experiment 3 may have influenced survival, differences in the length of the rooting period between the two experiments also likely influenced survival. SH stem cuttings stuck into the JF pellets in Experiment 3 were transplanted into the outdoor nursery bed after 9 weeks of rooting. This rooting period was 2 to 4 weeks shorter than for those transplanted in Experiment 1. The shorter transplant time impacted survival in Experiment 3, most likely, by limiting development of the root system prior to transplant. Survival rates for loblolly pine rooted cuttings likewise declined with shortened transplant times (Gocke 2006, Chapter 2).

Survival rates for rooted cuttings stuck in the CT system increased by 25% between Experiment 1 and 3. Rooting percentage for the CT system stem cuttings in Experiment 3 were only 4% larger than their final survival rates, suggesting that conditions in the outdoor rooting bed were nearly optimal for survival following rooting.

Root Dry Weight and Shoot Production; Experiment 3

Root dry weight measurements increased more than 100% between Experiment 1 to 3 for the JF transplant and CT system rooted cuttings. These results suggest that growing conditions were improved for these two systems in Experiment 3. Yet, even with a large increase in root growth, no significant shoot growth was observed.

Conclusions

All five production systems evaluated in the three experiments conducted in this study demonstrated potential for producing sweetgum rooted cuttings. However, post-sticking stock quality measurements differed among production systems, years, and clones tested. The JF and the GT transplant, and the CT production systems each demonstrated the potential for producing a large number of rooted cuttings with high survival rates and large root systems. Optimizing mist frequency and duration for each individual medium (plug) type during rooting, optimizing plug size, and optimizing post-rooting cultural practices (water and fertility management) should further improve stock quality for the JF and GT system rooted cuttings. Optimizing post-rooting cultural practices should also improve stock quality for the CT production system.

The SH-DS production system proved more sensitive than the other systems tested. Although the SH-DS system produced rooted cuttings with moderately large sized roots, survival was low. Secure and consistent watering is crucial for optimal rooting in this system, as well as, in the TP and CT systems. All of the production systems evaluated in this study would benefit from back-up systems designed to protect cutting material from equipment failure during rooting. Useful back-up systems include; auxiliary irrigation systems, a secondary source of power, and an alarm system to notify supervisors of potential problems. Continued focus on post-rooting cultural practices could also improve survival rates and root system size for SH-DS system rooted cuttings.

The H-DS production system was the only system to produce rooted cuttings exhibiting substantial shoot growth during the first growing season. Some of the rooted H cuttings also developed extensive root systems. Unfortunately, survival in this system was low. Collection time affected root dry weight and the number of rooted cuttings exhibiting new shoot growth. Altering the hormone concentration and time of application (eg. at the time of collection) may result in higher rooting percentages for this system.

The inability to obtain consistent and substantial shoot growth immediately following rooting from semi-hardwood and hardwood stem cuttings in the systems and conditions tested, severely limits the efficiency of these systems and suggests a critical research area.

A number of future scenarios could be tested to overcome shoot growth inhibition for SH stem cuttings and increase the success rates for H stem cuttings. These scenarios include testing: 1) potential post-rooting techniques to encourage shoot growth for rooted SH stem cuttings such as the application of shoot growth hormones following rooting, pruning terminal buds to promote growth of lateral buds, or placing the stem cuttings in conditions of extended day length and warm temperatures (26^0 C) following rooting to stimulate shoot growth, 2) the potential for rooting SH stem cuttings longer than 15-cm, out-planting them after one growing season (regardless of shoot growth), and managing them under intensive silvicultural practices to control competing vegetation and provide adequate moisture, and 3) the potential to improve on the H stem cutting results obtained from Experiment 2, in terms of survival, root dry weight, and new shoot growth, by optimizing collection time, growth, and environmental conditions. The advantage of this last scenario may be to produce high quality sweetgum rooted cutting stock in the same season as rooting.

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Table 1.2. Rooting percentage, survival, and root dry weights means (standard error) and significance for semi-hardwood sweetgum stem cuttings as influenced by production system and clone in Experiment 1. Production system and clone means (*) with the same letter, within a trait, had values that did not significantly differ (p<0.05) using pair-wise comparisons of transformed (**) data.

Dependent Variables	**Production System**				**Clone**	
	JF "inclusive"	GT "inclusive"	DS "inclusive"	CT	UC1 "inclusive"	2074 "inclusive"
Rooting (%)	*97 a (2.9)	82 b (2.9)	--	--	92.5 (2.9)	86 (2.9)
	P=0.0010				P=0.1153	
****Survival (%)**	82 a (3.84)	59 b (3.84)	35.5 c (5.42)	59 b (5.42)	71 x (3.32)	47 y (3.32)
	P<0.0001				P<0.0001	
****Root Dry Weight (g)**	0.17 b (0.018)	--	0.23 b (0.0257)	0.33 a (0.0257)	0.26 x (0.0192)	0.19 y (0.0192)
	P=0.0003				P=0.0077	

Note: "Inclusive" implies that the values presented in this table were obtained by averaging means from all treatments evaluated within a production system or for a specific clone.

Table 2.2. Rooting percentage, survival, and root dry weight means (standard error) and significance for semi-hardwood sweetgum stem cuttings as influenced by production system (Grow-Tech™ rooting sponge (GT) and Jiffy peat pellet (JF) transplant systems), transplant time, clone, and clone x transplant time in Experiment 1. Transplant system, transplant time, clone, and clone x transplant time means (*) with the same letter, within a trait, had values that did not significantly differ ($p<0.05$) using pair-wise comparisons.

Dependent Variables	Transplant System		Transplant Time		Clone		Clone x Transplant time			
	JF	**GT**	**11 wks**	**13 wks**	**UC1**	**2074**	**UC1 11wks**	**UC1 13wks**	**2074 11wks**	**2074 13wks**
Rooting %	*97 a (2.6)	82 b (2.6)	86 a (2.6)	92.5 a (2.6)	92.5 a (2.6)	86 a (2.6)	90 (3.7)	95 (3.7)	82 (3.7)	90 (3.7)
	p=0.0002		p=0.0754		p=0.0754		p=0.6505			
***Survival %**	82 a (3.9)	59 b (3.9)	67 a (3.9)	74 a (3.9)	83 a (3.9)	58 b (3.9)	73 b (5.6)	92.5 a (5.6)	61 bc (5.6)	56 c (5.6)
	p=0.0004		p=0.1109		p<0.0001		p=0.0378			
***Root Dry Weight (g)**	0.17 (0.008)	--	0.165 a (0.0115)	0.178 a. (0.0115)	0.195 a (0.0115)	0.148 b (0.0115)	0.177 (0.016)	0.21 (0.016)	0.15 (0.016)	0.144 (0.016)
	N/A		p=0.5084		p=0.0132		p=0.1897			

Table 3.2. Survival, root dry weight, and percent of newly elongated shoots (NES) means (standard error) and significance for hardwood stem cuttings of sweetgum as influenced by collection time, clone, and clone x collection time in Experiment 2. Collection time, clone, and clone x collection time means (*) with the same letter, within a trait, had values that did not significantly differ (p<0.05) using pair-wise comparisons.

Dependent Variables	**Collection Time**		**Clone**			**Clone x Collection Time**					
	January 20, 2001	March 18, 2001	UC1	UC2	2074	UC1-Jan	UC1 - March	UC2-Jan	UC2 - March	2074-Jan	2074 - March
Survival %	*23.7 a (1.8)	7.8 b (1.8)	4.4 b (2.2)	34.4 a (2.2)	8.3 b (2.2)	8.9 b (3.1)	0 c (3.1)	56.7 a (3.1)	12.2 b (3.1)	5.6 bc (3.1)	11.1 b (3.1)
	p<0.0001		p<0.0001			p<0.0001					
Root Dry Weight (g)	1.38 a (0.033)	0.31 b (0.033)	0.76 (0.041)	0.92 (0.041)	0.85 (0.041)	1.5 (0.058)	0 (0.058)	1.6 (0.058)	0.29 (0.058)	1.08 (0.058)	0.63 (0.058)
	p=0.0277		p=0.3219			p=0.1024					
% NES (newly elongated shoots)	6.3 a (1.1)	1.1 (1.1)	2.2 (1.3)	6.6 (1.3)	2.2 (1.3)	4.4 b (1.9)	0 b (1.9)	12.2 a (1.9)	1.1 b (1.9)	2.2 b (1.9)	2.2 b (1.9)
	p=0.0008		p=0.0246			p=0.0123					

Table 4.2. Rooting percentage, survival, and root dry weight means (standard error) and significance for semi-hardwood sweetgum stem cuttings as influenced by production system, clone, and production system x clone in Experiment 3. Production system, clone, and production system x clone means (*) with the same letter, within a trait, had values that did not significantly differ (p<0.05) using pair-wise comparisons. Transformed data was analyzed for all three dependent variables.

Dependent Variables	**Production System**		**Clone**		**Production System x Clone**			
	JF-9 wks	CT-9wks	UC1 "inclusive"	UC2 "inclusive"	JF - UC1	JF-UC2	CT-UC1	CT-UC2
Rooting %	*61 b (3.1)	88.5 a (3.5)	70 b (3.3)	79 a (3.3)	57 (4.4)	65 (4.4)	83 (4.9)	94 (4.9)
	p<0.0001		P=0.0465		P=0.8241			
Survival %	53 x (3.4)	85.5 y (3.4)	65 a (3.6)	74 a (3.6)	50 (4.8)	57 (4.8)	79 (5.4)	92 (5.4)
	p<0.0001		P=0.0635		p=0.5707			
Root Dry Weight (g)	0.45 a (0.24)	0.5 a (0.23)	0.48 a (0.23)	0.47 a (0.22)	0.41 c (0.23)	0.49 b (0.28)	0.55 a (0.24)	0.44 bc (0.18)
	p=0.6587		P=0.7688		p=0.0002			

Note: "Inclusive" implies that the values presented in that column were obtained by averaging the means of all treatments tested for each clone.

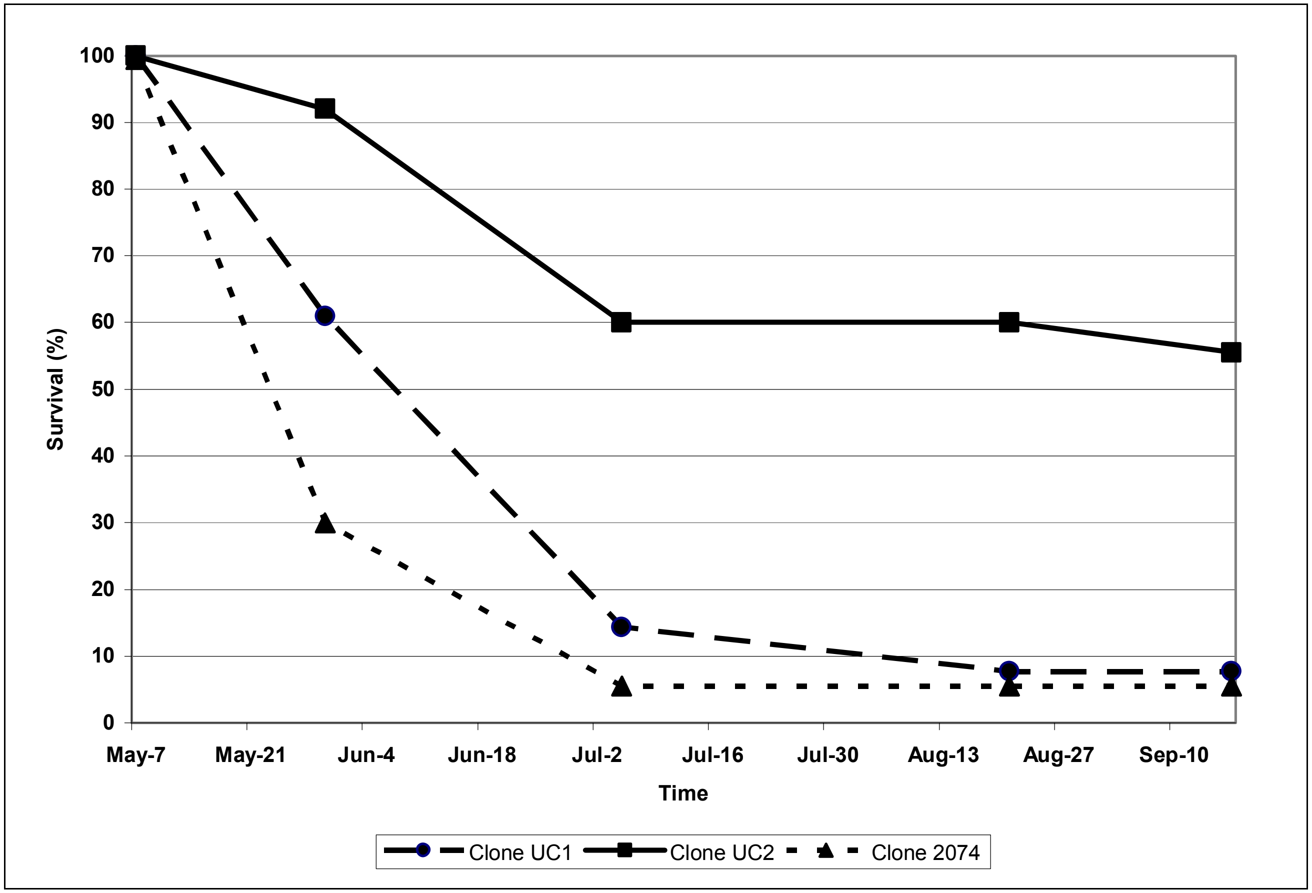

Figure 1.2. Experiment 2: Rate of survival for sweetgum hardwood stem cuttings collected from three clones on January 20, 2001 and stored in a cooler at 2^0 C until stuck in an outdoor nursery bed on April 14, 2001

Appendices

Appendix I. Experiment 1: The indoor and outdoor irrigation regimes employed in a sweetgum rooted cutting experiment conducted in 2000 at NC State University in Raleigh, NC.

<table>
<tr><th rowspan="2">Date</th><th colspan="3">Indoor and Outdoor Irrigation Regime for Experiment 1</th></tr>
<tr><th>SH-DS
(frequency/duration)</th><th>GT/JF (11- and 13-weeks)
(frequency/duration)</th><th>CT (12-weeks)
(frequency/duration)</th></tr>
<tr><td>6/16/00</td><td>N/A</td><td rowspan="8">Details in the Materials and Methods section</td><td rowspan="11">Details in the Materials and Methods section</td></tr>
<tr><td>6/22 – 7/13</td><td>60 sec / 5 min</td></tr>
<tr><td>7/14 – 7/21</td><td>60 sec / 6 min</td></tr>
<tr><td>7/21 – 7/31</td><td>60 sec / 7 min</td></tr>
<tr><td>*8/1</td><td>60 sec / 8 min</td></tr>
<tr><td>8/4</td><td>60 sec / 9 min</td></tr>
<tr><td>8/7</td><td>**Water off event!</td></tr>
<tr><td>8/17</td><td>60 sec / 10 min</td></tr>
<tr><td>8/18</td><td>60 sec / 15 min</td><td rowspan="5">GT- 11 and JF-11 weeks placed outside mist in propagation greenhouse, watered once a day</td></tr>
<tr><td>8/23</td><td>60 sec / 20 min</td></tr>
<tr><td>8/24</td><td>60 sec / 25 min</td></tr>
<tr><td>8/25</td><td>60 sec / 30 min</td><td rowspan="4">CT-12 placed outside mist in propagation greenhouse, watered twice a day</td></tr>
<tr><td>8/30</td><td>60 sec / 1 hour</td></tr>
<tr><td>9/1</td><td>60 sec / 2 hours</td><td rowspan="2">GT- and JF-11 transplanted to nursery bed, and hand watered once a day for the first week

GT- 13 and JF-13 weeks placed outside mist in propagation greenhouse, watered once a day</td></tr>
<tr><td>9/5</td><td rowspan="2">60 sec / 6 hours</td></tr>
<tr><td>9/8</td><td rowspan="2">GT-11 and JF-11 hand watered once every three days for remainder of study</td><td rowspan="2">CT-12 transferred outside and hand watered once a day for the first week</td></tr>
<tr><td>9/10</td><td rowspan="3">3 min / 6 hours</td></tr>
<tr><td>9/15</td><td>GT- and JF-13 transplanted to nursery bed,
Hand watered once a day for the first week</td><td rowspan="3">CT-12 hand watered once every two to three days</td></tr>
<tr><td>9/22</td><td rowspan="2">GT-13 and JF-13 hand watered once every three days</td></tr>
<tr><td>9/26</td><td>5 min / 10 hours</td></tr>
<tr><td>10/9</td><td colspan="3">End of growing season, water turned off.</td></tr>
</table>

* First sign of rooting in a border cutting. Many cuttings appear to have calloused

** Water was interrupted from 10 am to 4:30 pm. The high temperature that day was 95^0 F

Appendix II. The indoor and outdoor irrigation regimes employed in two sweetgum rooted cutting experiments conducted in 2001 at the NC State University in Raleigh, NC. Experiment 2 evaluated hardwood stem cuttings in a direct-stick production system (H-DS), while Experiment 3 evaluated semi-hardwood cuttings in Jiffy peat pellet (JF) transplant and containerized (CT) production systems.

<table>
<tr><th rowspan="2">Date</th><th colspan="2">Indoor and Outdoor Irrigation Regime 2001</th></tr>
<tr><th>H-DS – Experiment 2
(frequency / duration)</th><th>JF/CT (9-weeks) – Experiment 3
(frequency/duration)</th></tr>
<tr><td>4/13</td><td>8 units* / 60 sec</td><td rowspan="16">Details in the Materials and Methods section</td></tr>
<tr><td>4/20</td><td>10 units / 60 sec</td></tr>
<tr><td>4/27</td><td>12 units / 60 sec</td></tr>
<tr><td>5/3</td><td>12 units / 60 sec</td></tr>
<tr><td>5/10</td><td>12 units / 60 sec</td></tr>
<tr><td>5/17</td><td>12 units / 60 sec</td></tr>
<tr><td>5/22</td><td>12 units / 60 sec</td></tr>
<tr><td>5/29</td><td>12 units / 60 sec</td></tr>
<tr><td>6/5</td><td>14 units / 60 sec</td></tr>
<tr><td>6/7</td><td>20 units / 60 sec</td></tr>
<tr><td>6/24</td><td>25 units / 60 sec</td></tr>
<tr><td>7/4</td><td>30 units / 60 sec</td></tr>
<tr><td>7/17</td><td>45 units / 60 sec</td></tr>
<tr><td>7/24</td><td>**2 min / 1 hour</td></tr>
<tr><td>8/1</td><td>5 min / 1 day</td></tr>
<tr><td>8/8</td><td rowspan="3">Hand watered once every three days for remainder of season</td></tr>
<tr><td>8/27</td><td>JF and CT moved to the outdoor nursery bed after 9-weeks of rooting and hand watered once a day for the first week</td></tr>
<tr><td>9/3</td><td>CT hand watered once every two days, while JF hand watered once every three days for the remainder of the growing season</td></tr>
<tr><td>10/1</td><td colspan="2">End of growing season, water off!</td></tr>
</table>

* Irrigation frequency was controlled by a variable-cycle control system, the Davis Solar 6A misting controller. Frequency was automatically adjusted by this controller according to the amount of sunlight detected by a light sensor connected to the controller. Light was measured in solar units. One solar unit equaled 2000 foot-candle or 0.02 moles/m^2. On a clear sunny day at noon, one solar unit equaled approximately 30 seconds. On a completely overcast day at noon, one unit equaled approximately 3 minutes.
At night no irrigation was applied.

** Irrigation frequency was controlled by a fixed-cycle control system, the Rainbird MIC-12. Irrigation and frequency was controlled by manually reprogramming the controller.

Appendix III. The fertilization schedule employed in a sweetgum rooted cutting experiment conducted in 2000 at NC State University in Raleigh, NC.

Date	Fertilization Schedule Experiment 1		
	SH-DS (rate/frequency)	GT/JF (rate/frequency)	CT (rate/frequency)
8/11	--	50 ppm N / wk	50 ppm N / wk
8/18	50 ppm N / wk	--	--
8/25	--	50 ppm N / wk	50 ppm N / wk
9/1	50 ppm N / wk	--	--
9/8	--	50 ppm N / wk	50 ppm N / wk
9/15	50 ppm N / wk	100 ppm N/ wk	100 ppm N/ wk
9/22	100 ppm N/ wk	--	--
9/29	--	100 ppm N/ wk	100 ppm N/ wk
10/6	100 ppm N/ wk	--	--
Total	55 kg N/ ha	55 kg N/ ha	55 kg N/ ha

Appendix IV. The fertilization schedules employed in two sweetgum rooted cutting experiments conducted in 2001 at NC State University in Raleigh, NC.

Date	Fertilization Schedule Experiment 2	Fertilization Schedule Experiment 3	
	H-DS (rate/frequency)	JF-9 weeks (rate/frequency)	CT-9 weeks (rate/frequency)
6/22	50 ppm N/ wk	--	--
7/6	50 ppm N/ wk	--	--
7/20	50 ppm N/ wk	--	--
8/3	100 ppm N/ wk	--	--
8/13	--	50 ppm N/wk	50 ppm N/wk
8/17	100 ppm N/ wk	--	--
8/27	--	50 ppm N/wk	50 ppm N/wk
8/31	100 ppm N/ wk	--	--
9/10	--	50 ppm N/wk	50 ppm N/wk
9/14	100 ppm N/ wk	--	--
9/24	--	100 ppm N/wk	50 ppm N/ 2X wk
9/28	100 ppm N/ wk	--	--
10/8	--	100 ppm N/wk	50 ppm N/ 2X wk
Total	105 kg N/ ha	55 kg N/ ha	55 kg N/ ha