Long-Term Container Effects on Root System Architecture of Longleaf Pine

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Abstract: Longleaf pine (*Pinus palustris* Mill.) seedlings cultured in three container cavity volumes and two cavity types (regular or copper oxychloride coating for root pruning) were excavated three years after planting in 2007 in Louisiana, U.S.A. Copper root pruning did not affect seedling growth. Seedlings from small cavities (60 ml) were smaller than those from medium (93 ml) or large (170 ml) cavities. About 59%, 35%, and 6% of the first-order lateral roots (FOLR) originated from the top, middle, and bottom thirds of the root plug in all treatments. Copper seedlings had more FOLR egress from the top root zone than regular seedlings which had almost even distribution of FOLR egress in all root zones. Copper treatment, however, did not completely ameliorate lateral root or taproot deformity. A few saplings toppled after Hurricane Gustav in 2008 and more saplings toppled in August 2009. Possible causes of sapling toppling are discussed.

Keywords: container planting stock, copper root pruning, *Pinus palustris* Mill., root system architecture, sapling toppling

During the last two decades, public, industrial, and private land managers and owners have been actively restoring longleaf pine ecosystems in the southern United States (Barnett 2002, Boyer 1989, Landers et al. 1995). Of 33 to 69 million longleaf pine seedlings produced annually, 70% are container stock and the rest are bareroot stock (South et al. 2005, McNabb and Enebak 2008). Most studies show that container-grown seedlings usually have a higher survival rate than bare-root stock (South et al. 2005 and references cited therein). However, saplings from container stock, but not bareroot stock, have experienced wind-throw during high wind events (South et al. 2001). One of the attempted improvements in the morphological characteristics of container stock root systems was to coat the interior cavity wall with copper (Cu). Slow release of low concentration Cu stops seedling lateral roots from elongating once they reach the cavity wall (Ruehle 1985). In a root growth potential test, longleaf pine seedlings grown in Cu coated cavities produced more new roots than those grown in non-Cu containers or bareroot seedlings (South et al. 2005). Lodgepole pine (*P. contorta* Douglas ex. Louden) grown in Cu-coated cavities had fewer leaning seedlings three years post planting than those from cavities without a Cu coating (Krasowski 2003).

A study comparing the effects of container cavity volume and copper root pruning on longleaf pine seedling growth, field performance, and sapling stability was implemented in 2004 in central Louisiana. Here, we will focus on the container effects on growth and root system architecture of the seedlings excavated three years after planting. We will also examine the preliminary results of container treatments on sapling toppling.

Materials and Methods

Seedling Culture and Stand Establishment

This study is a part of a long-term study whose nursery culture and field stand establishment protocols were described by Sung et al. (in press) and Sword Sayer et al. (in press). Briefly, mixed seedlots of longleaf pine from Florida were sown into containers in April, 2004. There were 2 by 3 factorial container treatments: two types of cavity coating and three cavity volumes. Cavity volume for the small (S), medium (M) and large (L) cavities were 60 ml (2.8 cm top diameter and 13.3 cm depth), 93 ml (3.6 cm top diameter and 15 cm depth), and 170 ml (4.2 cm top diameter and 15.2 cm depth), respectively. Styroblock ® and Copperblock® containers (Beaver Plastics Ltd, Edmonton, Alberta, Canada) of the above mentioned cavity volumes were used for no copper (R) and copper root pruning (Cu) treatments, respectively. Copper oxychloride was the active ingredient in the coating on the interior cavity surface. Protocols for growing longleaf pine by Barnett and McGilvray (2000) were adapted for this study with some modifications (Sword Sayer et al. in press).

The field study site is located on the Palustris Experimental Forest within the Kisatchie National Forest in Rapides Parish of central Louisiana (31°10’N, 92°41’W). The soil is a moderately well-drained, gently sloping Beauregard silt loam (fine silty, siliceous, superactive, thermic, Plinthaquic Paludults). Mima mounds of Malbis fine sandy loam (fine loamy, siliceous, subactive, thermic, Plinthic Paleudults) are scattered across the study area. The field study is a randomized complete block factorial design with four replications. Blocking was by soil drainage. Twenty-four treatment plots of 0.0576 ha (24 x 24 m) each were established. Seedlings grown in the six cavity treatments (Cu-L, R-L, Cu-M, R-M, Cu-S, and R-S) were randomly assigned to a plot in each block. In early November 2004, 27-week-old container-grown longleaf pine seedlings were lifted and planted on the same day. Seedlings were planted at a 2 x 2 m spacing. Treatment plots are 12 rows of 12 trees. All plots were prescribe burned in February, 2006 (15 months after planting) and again in May 2009 (fifth growing season) as part of the routine management of the site.

Seedling Excavation and Measurements

In November, 2007 (three years after planting), four seedlings from each of the 24 plots were randomly selected for excavation with shovels. Seedlings were excavated at a 30 cm radius from the stem and to the depth of the end of the deepest root. Soil was removed from roots and seedlings were transported to a greenhouse for evaluation. After height (Ht) and root collar diameter (RCD) were measured, each seedling was cut off at the root collar. Fascicles and stems were oven dried at 70°C for dry weights (DW).

Root system architecture was assessed as follows. Length of the deepest roots, either sinkers or taproot, was measured. Sinkers are the adventitious roots that form at the end of the taproot and extend vertically. Number of first-order lateral roots (FOLR) per seedlings was counted. First-order lateral roots (FOLR) are lateral roots originating from the taproot, sturdy in structure, and with a diameter of at least 3 mm measured 2 cm away from the taproot. A root plug template was drawn for each cavity volume with its original depth and twice the original top diameter because most taproots had grown almost to the fullness of the original cavity diameter. Each seedling root system was placed on a root plug template of the corresponding cavity volume. Three zones of equal plug depths were designated, namely top root zone starting from root collar down and followed by middle zone and bottom zone. Each zone depth was 5 cm, 5 cm and 4.4 cm for L, M, and S cavities, respectively. Number of FOLR originating from and egressing from each root zone on the template was counted. Roots egressing from the bottom of the root plug were grouped as the end zone. After all the parameters for root system architecture were determined, individual FOLRs were cut off right at the originating point on the taproot and were combined based on the root plug zone where they originated. Secondary or tertiary roots remained on the FOLRs. Taproot and FOLR DW were determined.

Sapling Toppling Evaluation

Hurricane Gustav passed through central Louisiana on September 1, 2008. We assessed the study site for wind damage three weeks later. All but one of the toppled saplings (i.e., leaning more than 15° from the vertical) resumed the vertical position by April 2009. One sapling remained down but was still producing new flushes in 2009. This sapling was excavated in April 2009 to examine its root system architecture.

Between August 7 and 17, 2009, quite a few saplings either toppled over or began toppling. These saplings will be monitored continuously and eventually excavated to assess root system architecture in 2010.

Statistical Analysis

ANOVA of seedling variables such as Ht, RCD, DW of seedling components, was conducted using a 2 by 3 factorial randomized complete block design with four blocks. Copper root pruning (Cu and R) and cavity volume (L, M, and S) were the main effects. Number of FOLR originating from each root plug zone and number of FOLR egressing from each root plug zone were tested by ANOVA using a 2 by 3 factorial randomized complete block split plot in space design with four blocks. Whole plot effects were cavity volume and copper coating, and the subplot effect was root plug depth (top, middle, bottom, and end). If significant treatment effects were found, plot mean comparisons were made with Tukey-Kramer Multiple Range Tests ($\alpha = 0.05$).

Results and Discussion

Seedling Growth

Copper root pruning did not significantly affect Ht, RCD, or DW of the excavated seedlings (data not shown). Earlier results from the same study reported by Sword Sayer et al. (in press) indicated that Cu increased S seedling size but not the size of M or L seedlings at the end of nursery production. This Cu effect was not repeated after the first-year in field (Sword Sayer et al. in press). In a study of *P. halepenis* by Tsakaldimi and Ganatsas (2006), Cu root pruning increased seedling size at planting and two years after planting. However, the post-planting differences were mainly due to the differences at planting. Cavity type and volume significantly affected number of FOLR. Number of FOLRs for Cu and R seedlings was 8.6 and 7.1, respectively. Seedlings of L, M, and S cavities had 9.1, 8.3, and 6.2 FOLR. This increasing FOLR number with cavity volume was also observed during nursery production by Sword Sayer et al. (in press). Number of FOLR has been documented as an important factor for seedling and young tree growth (Kormanik et al. 1998). Therefore, Cu root pruning, at worst, has no negative impact on container stock seedling growth in the nursery or field; and it can have a positive effect on seedling growth for several years (Tsakaldimi and Ganatsa 2006).

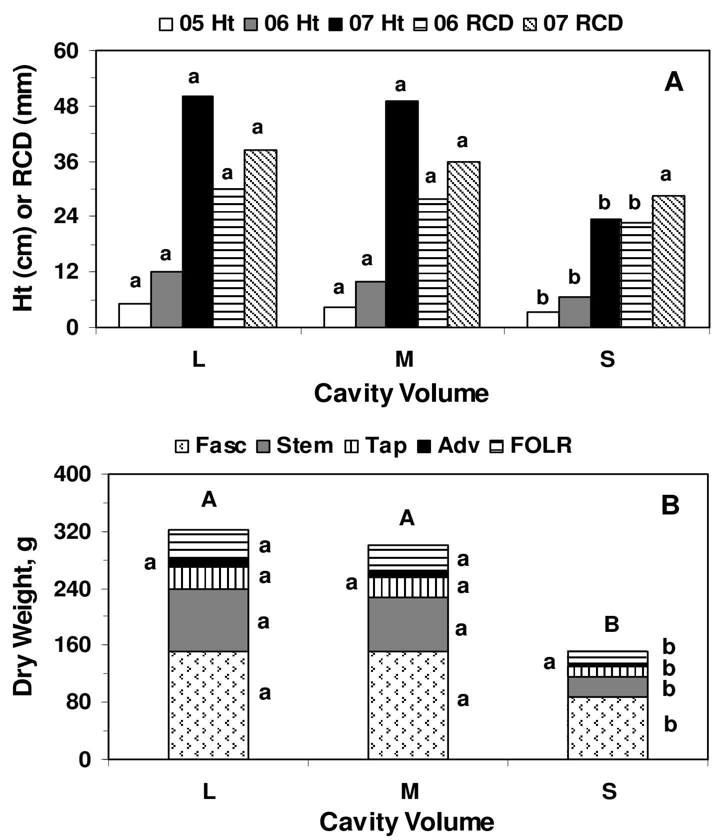

Figure 1. Effects of container cavity volume (large, medium, and small) on height and root collar diameter (A) and dry weights (B) of longleaf pine seedlings excavated three years after planting in Louisiana, U.S.A. Means associated with each variable and year with different letters are significantly different at a 0.05 level by Tukey-Kramer tests. Capital letters in (B) are for total seedling dry weights.

Cavity size had significant effects on all growth parameters except for RCD in 2007 (Figure 1A and B). Seedlings grown in the S cavities were the smallest since one year after planting. During the second year in the field, all L seedlings began accelerated Ht growth (i.e., bolting with a height of > 12 cm) whereas only 28% of M seedlings and none of the S seedlings bolted during the second year. During the third year, 84% and 69% of M and S seedlings bolted, respectively. Early height growth is critical for longleaf pine seedlings to compete with surrounding vegetation for resources such as light, water, and nutrients (Ramsey et al. 2003).

Each of the S seedling components was significantly lower in DW than its corresponding component in the L and M seedlings except that there was no volume effect on the adventitious root DW (Figure 1B). Growth of L and M seedlings was comparable for all parameters evaluated (Figure 1). These growth results were similar to the results reported by Sword Sayer et al. (in press) for seedlings at planting and one year after planting. For producing container longleaf pine seedlings, Barnett and his colleagues (1997, 2000, 2002) recommended a minimum cavity of 98 ml which is the M cavity volume in this study. Indeed, seedlings grown in the S containers in the nursery did not catch up in growth with the L or M seedlings even three years after planting.

Dry weight allocation to each seedling component was not affected by Cu root pruning but was affected by cavity volume. Seedlings from S cavities allocated a significantly higher percentage of DW (60%) to fascicles compared to 54% in M and 50% in L seedlings. Allocation to stem had the opposite trend as that of the fascicles. The S seedlings allocated significantly less DW (16%) to the stems than M (21%) and L (23%) seedlings. No differences were observed in percents of DW allocation to taproots, adventitious roots, or FLORs among treatments.

Root System Architecture

When FOLRs were grouped by their origination zones (top, middle, or bottom) on the taproots, cavity type or volume did not change the number and DW of FOLRs in each root plug zone (Figure 2A). The top root zone had the highest percents, about 60%, of FOLR number and DW followed by the middle root zone (Figure 2A).

Patterns of FOLR egress from different root zones were not affected by cavity volume. However, Cu root pruning treatment changed the FOLR egress pattern significantly from that of the R seedlings (Figure 2B). For seedlings grown in regular cavities, FOLR egress was evenly distributed among the four root plug zones. Within a Cu seedling root system, the top root plug zone had the highest percent of FOLR egress followed by the middle zone. Both the bottom and the end root zones had the lowest percentages of FOLR egress in the Cu seedlings (Figure 2B). The FOLR egress pattern in Cu seedlings reflected the pattern of FOLR origination but the R seedling root system did not. Regular cavity imposed a great impact on the lateral root architecture by vertically displacing at least 33% and 11% of FOLR from egressing from the top and middle root zones to lower root zones. Copper root pruning partially ameliorated this lateral root system deformity with about 13% of the top zone FOLRs egressing from lower zones. These patterns of FOLR egress were also reported by Sword Sayer et al. (in press) on the seedlings one year after planting. However, FOLR origination zone data were not reported in that study. Seedlings with a deeper lateral root system may not acquire nutrients and water in the top soil zone. Furthermore, we have observed more lateral root spiraling and vertically extending within the root plug in R seedlings compared to Cu seedlings. These deformed lateral roots may strangle each other or their taproots and further exacerbate sapling stability problems.

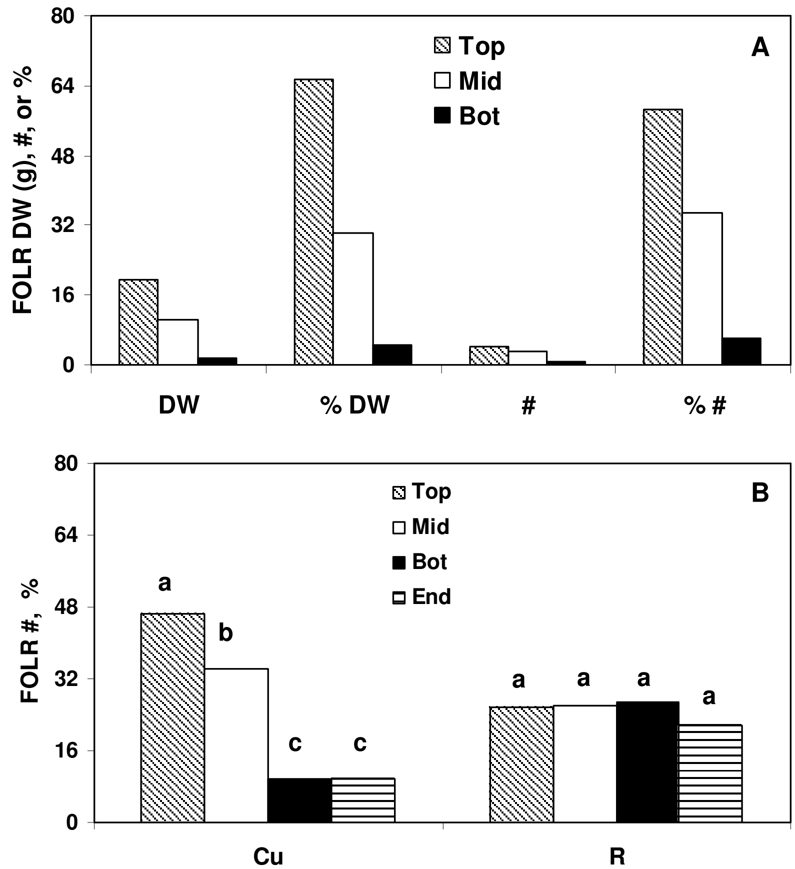

Figure 2. (A) Distribution of dry weights (DW) and number of first-order lateral roots (FOLR) within three root plug zones for all excavated seedlings. (B). Distribution of percentage of the number of FOLR egressing from three root plug zones and the end zone. Within each cavity type (Cu-copper coating and R-regular) means associated with different letters are significantly different at a 0.05 level by the Tukey-Kramer tests.

Sapling Stability and Root System Architecture

Taproots and sinkers can contribute to vertical anchorage and non-deformed lateral roots can contribute to horizontal anchorage for sapling stability (Burdett et al. 1986, Coutts et al. 1999). In this study, cavity type or volume did not affect taproot lengths. The deepest root in each seedling may be a taproot, a sinker, or a vertically trained lateral root. Cavity volume affected the deepest root length. The mean length of the longest root in the L seedlings was 46 cm and was significantly greater than those in M (40 cm) and S (32 cm) seedlings. If the longest root in a seedling was the taproot or a sinker, then it probably contributed to vertical anchorage for that seedling. The vertically trained lateral root may not be of much help to seedling stability if it spirals around the taproot or it zigzags within the root plug as observed in some of the excavated seedlings.

Three weeks after Hurricane Gustav passed through central Louisiana in September 2008, we surveyed the study site and recorded sapling toppling. Each container treatment had 1 to 4 toppled saplings. Most of the toppled sapling stems became somewhat sinuous by May, 2009. One toppled sapling remained down yet was flushing in Spring 2009. This sapling was excavated in April 2009. Its taproot was damaged about 7 cm from root collar. Two adventitious roots were formed from the damaged taproot end. One grew downward as a sinker and the other one grew outward (Figure 3).

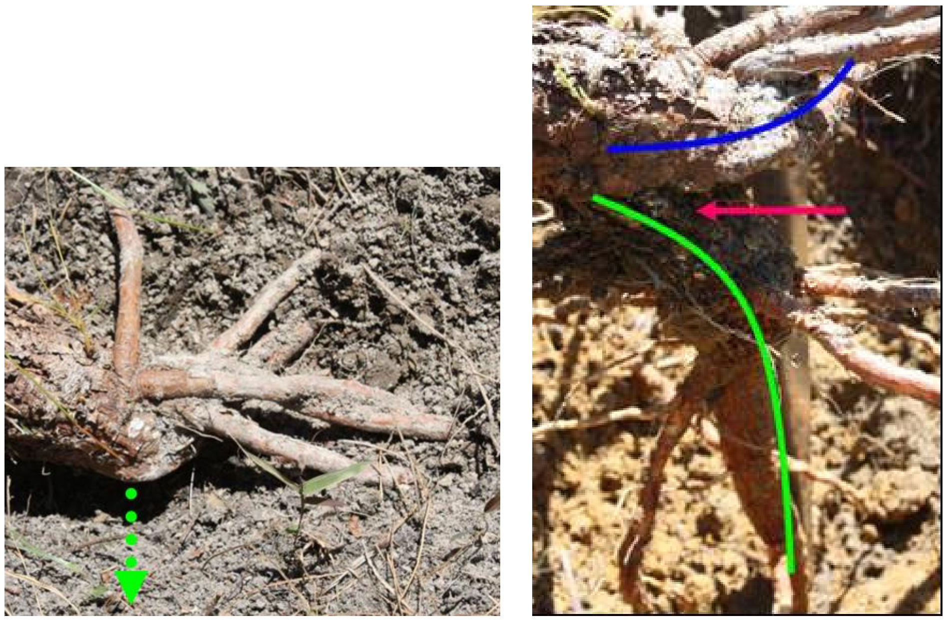

Figure 3. A longleaf pine sapling toppled over after Hurricane Gustav in September, 2008. Pictures were taken in April 2009 before (left) and after (right) soil was removed. Dotted green arrow in the left picture indicates the position of a sinker root which was delineated in green in the right picture. Magenta arrow points at past damage the taproot sustained. Blue line shows the outward extension of the other adventitious root from the end of the damaged taproot.

Sometime between August 7 and 17 2009, the current study site may have sustained a mild wind storm which resulted in sapling toppling in all treatments. A total of 16, 8, 11, 11, 6 and 4 seedlings either toppled over or toppling (with > 15° of leaning) in Cu-L, R-L, Cu-M, R-M, Cu-S, and S plots, respectively. Our preliminary survey did not find any specific factor or combination of factors (e.g., height, diameter, sapling location within the plot, plot location, and container treatment) that could be attributed to this incident. A sapling excavation study will be conducted in 2010 to determine the cause. In a tree excavation study by Harrington and Gatch (1999), stem sinuosity of young *P. taeda* L. was highly associated with bent taproots as opposed to straight taproots. Based on the information gathered from Figure 3 and the results by Harrington and Gatch (1999), we speculate that the toppling longleaf saplings in our study have short, damaged, deformed taproot or sinker or both or lack a taproot or a strong sinker. They may also have an uneven distribution of lateral roots around the taproot circumference or tangled lateral roots. It is also possible that the saplings may be planted over a layer of dense soil which restricts vertical extension of roots.

Summary

Our results support the use of container cavities with copper coating for chemical root pruning during seedling production to reduce the extent of lateral root deformity imposed by hard cavity walls. Even after three years in the field, seedlings from the regular cavities still have a greater extent of lateral root deformity than the Cu seedlings. Furthermore, seedlings grown in cavities of 60 ml remained smaller in size and dry weight than seedlings produced in larger cavities three years after planting. Although sapling stability may be improved with seedlings grown in copper cavities, the vertical anchorage provided by taproots or sinkers probably has to come from additional improvements to containers or changes in planting methods.

Literature Cited

BARNETT, J.P., and J.M. MCGILVRAY. 1997. *Practical guidelines for producing longleaf pine seedlings in containers*. US Forest Service General Technical Report SRS-14. 28 p. US Forest Service, Asheville, NC.

BARNETT, J.P., and J.M. MCGILVRAY. 2000. Growing longleaf pine seedlings in containers. Native Plant Journal 1(1): 54-58.

BARNETT, J.P. 2002. Longleaf pine: Why plant it? Why use containers? P. 5-7 in *Proceedings of Workshops on growing longleaf pine in containers – 1999 and 2001*, Barnett, J.P., R. K. Dumroese, and D.J. Moorehead (eds.). General Technical Report SRS-56. US Forest Service, Asheville, NC.

BOYER, W.D. 1989. Response of planted longleaf pine bare-root and container stock to site preparation and release: fifth-year results. P. 165-168 in Miller, J.H. (comp.). *Proceedings of the fifth biennial southern silvicultural conference.* General Technical Report SO-74. US Forest Service, New Orleans, LA.

BURDETT, A.N., H. COATES, R. EREMKO, and P.A.F. MARTIN. 1986. Toppling in British Columbia's lodgepole pine plantations: Significance, cause and prevention. The Forest Chronicle 62: 433-439.

COUTTS, M.P., C.C.N. NIELSEN, and B.C. NICOLL. 1999. The development of symmetry, ridigdity and ahcorage in the structural root system of conifers. Plant and Soil 217:1-15.

HARRINGTON, T.B., and J.A. GATCH. 1999. Stem sinuosity, tree size, and pine injury of machine-planted loblolly pine with bent versus straight taproots. Southern Journal of Applied Forestry 23: 197-202.

KORMANIK, P.P., S.S. SUNG, and S.J. ZARNOCH. 1998. Immature loblolly pine growth and biomass accumulation: Correlations with seedlings initial first-order lateral roots. Southern Journal of Applied Forestry 22: 117-123.

KRASOWSKI, M.J. 2003. Root system modifications by nursery culture reflect on post-planting growth and development of coniferous seedlings. The Forest Chronicle 79: 882-891.

LANDERS, J.L., D.H. VAN LEAR, and W.D. BOYER. 1995. The longleaf pine forests of the southeast: Requiem or renaissance? Journal of Forestry 93: 39-44.

MCNABB, K., and S. ENEBAK. 2008. Forest tree seedling production in the southern United States: the 2005- 2006 planting season. Tree Planters' Notes. 53(1): 47-56.

RAMSEY, C.L., S. JOSE, B.J. BRECKE, and S. MERROTT. 2003. Growth response of longleaf pine (Pinus palustris Mill.) seedlings to fertilization and herbaceous weed control in an old field in southern USA. Forest Ecology and Management. 172: 281-289.

RUEHLE, J.L. 1985. The effect of cupric carbonate on root morphology of containerized mycorrhizal pine seedlings. Canadian Journal of Forest Research. 15: 586-692.

SAS Institute Inc. 2004. SAS/STAT® 9.1 User's Guide. Cary, NC: SAS Institute Inc. 5121 p.

SOUTH, D.B., S.W. HARRIS, J.P. BARNETT, M.J. HAINDS, and D.H. GJERSTAD. 2005. Effect of container type and seedling size on survival and early height growth of *Pinus palustris* seedlings in Alabama, U.S.A. Forest Ecology and Management 204: 385-398.

SOUTH, D.B., J. SHELTON, and S.A. ENEBAK. 2001. Geotropic lateral roots of container-grown longleaf pine seedlings. Native Plants Journal 2:126-130.

SUNG, S.S., J.D. HAYWOOD, M.A. SWORD-SAYER, K.F. CONNOR, and D.A. SCOTT. D.A. (in press). Effects of container cavity size and copper coating on field performance of container-grown longleaf pine seedlings. in *Proceedings of the 14th biennial southern silvicultural research conference*. Stanturt, J. A. (ed.). US Forest Service, Asheville, NC.

SWORD SAYER, M.A., J.D. HAYWOOD, and S.S. SUNG. (in press). Cavity size and copper root pruning affect production and establishment of container-grown longleaf pine seedlings. Forest Science

TSAKALDIMI, M.N., and P.P. GANATSAS. 2006. Effect of chemical root pruning on stem growth, root morphology and field performance of the Mediterranean pine *Pinus halepensis* Mill. Scientia Horticulturae 109: 183-189.