INCREASING SURVIVAL PROBABILITY, HEIGHT GROWTH, AND DIAMETER GROWTH OF 1-0 SHORTLEAF PINE SEEDLINGS THROUGH GENETIC SELECTION, SITE PREPARATION, AND VEGETATION CONTROL

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Abstract—Natural regeneration of shortleaf pine (*Pinus echinata* Mill.) has proven difficult due to slow growth, high mortality, and lack of competitive ability. Consequently, artificial regeneration is frequently used for reforestation, although regeneration failures are common. We examine how genetic selection, in conjunction with extensive site preparation and understory vegetation control, can increase reforestation success. We used data from three long-term studies in southeastern Missouri to examine survival of 5,500 1-0 shortleaf pine seedlings. One site contained 2,000 full-sib plus 1,000 orchard-run 1-0 containerized seedlings planted on a mechanically cleared hillslope. Herbicide was applied at initial planting and 1 year after planting to 2,000 seedlings, while 1,000 seedlings received no herbicide. In addition, 2,500 orchard-run bareroot seedlings were planted at two similar sites: one utilizing herbicide understory control for 10 years, and one underplanting in an existing forest with no site preparation or herbicide. Seedling survival after 7 years on sites with no vegetation control was 30 percent. With herbicide treatments before and after planting, survival of orchard-run seedlings was 60 percent 7 years after planting. When pedigreed seedlings were planted on mechanically prepped and herbicide-treated sites, average survival increased to 85 percent after 7 years. Survival of planted shortleaf pine seedlings increased as site preparation intensity and understory competition control increased. Long-term survival was reduced by underplanting seedlings in an existing hardwood stand, where overstory and understory competition was present.

INTRODUCTION

Shortleaf pine (*Pinus echinata* Mill.) has the widest range and distribution of any southern pine in the United States, and is found in at least 22 States from New York and New Jersey in the Northeast, to Oklahoma and Texas toward the Southwest (Lawson 1990). It is the only pine species native to Missouri, naturally occurring throughout the southern and southeastern areas of the State (Brinkman and Rodgers 1967, Liming 1946), where it is commonly found as a component of varying significance with oaks (*Quercus* spp.), hickories (*Carya* spp.), and other hardwoods (Moser and others 2007). Though not generally a commercial species of interest, restoration of shortleaf pine is a priority throughout Missouri to increase wildlife diversity and forest resilience (Eddleman and others 2007, Kabrick and others 2015). Artificial and natural regeneration methods have encountered mixed success due to fire suppression, understory competition, lack of seedling establishment, and slow seedling growth (Gwaze and others 2006a, Jensen and others 2007, Lawson 1990, Stambaugh and others 2007).

There is a need to identify methods for increasing the early survival and growth of shortleaf pine seedlings. Though considerable research into site preparation methods, spacing, herbicide use, and genetics exist for loblolly (*Pinus taeda* L.), slash (*P. elliotti* Engelm.), and longleaf (*P. palustris* Mill.) pines (Baldwin and others 2000, Land and others 2004, Lewis and others 1985, Raley and others 2003, Schubert and others 2004, South and others 2004), there is less current research into these practices for shortleaf pine (Gwaze and others 2005, Gwaze and others 2006b, Smith 2011, Studyvin and Gwaze 2007). Our objectives were to assess the effects of genetic selection, site preparation, and

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vegetation control on first- and seventh-year survival, height growth, and diameter growth of planted 1-0 shortleaf pine seedlings.

METHODS AND DATA ANALYSIS

Site Descriptions

Three datasets were used for this study. The first was from a study conducted at the Wurdack Research Center in Crawford County, Missouri (N 37°47'23.9", W 91°25'12"). The site was a continuous, south-facing slope of 10-30 percent, with two soil series occurring: Reuter (loamy-skeletal, siliceous, active, mesic Typic Paleudalfs) on the upper and lower backslopes, and Goss (clayey-skeletal, mixed, active, mesic Typic Paleudalfs) occurring on lower slope positions (USDA NRCS 2017). The second study was conducted at the Long-Term Soil Productivity site (LTSP) in Shannon County, Missouri (N 37°10'47.9", W 91°6'35.9"). Soils on site were primarily of the rocky, nutrient-poor Clarksville series (loamy-skeletal, siliceous, semi-active, mesic Typic Paleudults), formed from hillslope sediments and cherty residuum from dolomite (USDA NRCS 2017). The third study was conducted at Sinkin Experimental Forest, Reynolds and Dent counties, Missouri (N 37°29'24", W 91°15'36"). Soils included the Clarksville and Coulstone series, (loamy-skeletal, siliceous, semi-active, mesic Typic Paleudults), as well as the Nixa series (loamy-skeletal, siliceous, active, mesic Glossic Fragiudults) (USDA NRCS 2017). At all three sites, the site index (base age 50) for shortleaf pine was between 17.1 m and 18.3 m (California Soil Resource Lab 2017). Thus, height and diameter growth differences among the three sites were likely due to treatments rather than site factors.

At Wurdack, seedlings consisted of genetically improved, 1-0 containerized shortleaf pine stock representing 12 full-sib families, plus standard, orchard-run seedlings from the State nursery at Licking, MO. Families were crossed from elite parents selected in natural stands on the Mark Twain National Forest in the 1970s (Gwaze and others 2005, Smith 2011). The existing hardwood forest was clearcut, and a masticator was used to grind tree stumps and nonmerchantable material. The site was then divided into four blocks. Seedlings were deployed in complete blocks, with 700-1,000 seedlings per block. Three blocks had a silvopasture treatment applied, which involved a double row of trees spaced 3.05 m by 3.05 m followed by a 12.19-m wide alley. A mix of native warm and cool season grasses were broadcast seeded into the alleyways. Herbicide pellets (Velpar®) were applied to the seedling planting rows for 2 years following planting. The fourth block eliminated the alley, leaving rows evenly spaced at 3.05 m. No herbicides were applied in this block and no grasses were seeded. Instead, an additional 12 families (not replicated in the other three blocks) were selected and planted along with the 12 replicated families. Tree heights and diameters were measured in 2010, 2011, and 2017. For purposes of this analysis, trees planted in the no-herbicide treatment will be identified as either genetically improved (G), or orchard-run (O) pedigree. Treatments at Wurdack included VC-O (orchard-run seedlings planted with herbicide application as a vegetation control), No-VC-G (trees belonging to one of the 12 genetic families at Wurdack, planted with no herbicide applied as vegetation control), No-VC-O (orchard-run seedlings planted with no herbicide applied as vegetation control), and the additional 12 pedigreed families each planted with vegetation control.

At the LTSP, a 3 x 3, split-plot factorial study design implemented in three blocks was used to simulate harvest-related soil disturbances including organic matter (biomass) removal, soil compaction, and vegetation control. The study was established in 1995, following a nationally implemented protocol which dictated methodology (Ponder and Mikkelson 1995, Powers and others 1990). Three levels of organic matter removal included removal from site of tree boles only, whole tree (bole, top, limbs), and whole tree plus forest floor (tree bole, top, limbs, and raking of forest floor to remove the organic soil horizon). Three levels of soil (whole-soil) compaction were implemented: no compaction (average bulk density of ~1.30 g/cm^3), moderate compaction (average bulk density ~1.60 g/cm^3), and severe compaction (average bulk density ~1.80 g/cm^3). In addition, half of each treatment plot had chemical herbicide applied annually for 10 years following seedling planting. Bare-root, 1-0 northern red oak (*Quercus rubra* L.), white oak (*Q. alba* L.), and shortleaf pine were planted following the application of the treatments (only shortleaf pine data are included in these analyses). Tree height and diameter (basal diameter or diameter at breast height, depending on tree height) were measured annually from 1995 until 2004, with additional measurements in 2013 and 2016. Treatments at the LTSP included for this analysis were VC (chemical vegetation control) and No-VC (no vegetation control). For additional information on study design, plot layout, and site characteristics, see Ponder and Mikkelson (1995) and Lyczak (2019).

At the Sinkin Experimental Forest, treatments included underplanting 1-0 bareroot shortleaf pine seedlings in 48 treatment units in existing second-growth mixed hardwood stands after manipulating the overstory in each treatment unit to a single stocking level between 0 and 75 percent (Gingrich 1967). A windstorm 1 year after seedling planting caused the blowdown and removal of additional overstory trees in some treatment plots, lowering the overstory density and residual basal area. In order to determine the effects of competition on shortleaf pine seedlings, the nearest hardwood competitor was identified, tagged, and measured and tracked throughout the experiment. Hardwood competitors were from either advanced reproduction or stump sprouts. Pine height and diameter (basal diameter or diameter at breast height, depending on height) were measured in 2008, 2009, 2010, 2013, and 2017. Identified hardwood competitors were measured in 2010, 2013, and 2017. Treatments for the Sinkin Experimental Forest included trees less than 80 percent of the height of the nearest competitor regardless of overstory stocking (<80Ht), trees greater than 80 percent of the nearest competitor in height, with an overstory stocking level of 45 percent or more (>45 percent stocking), trees greater than 80 percent of competitor height, with an overstory stocking between 15 and 45 percent (15-45 percent stocking), and trees within 80 percent of competitor height with an overstory of 15 percent or less (<15 percent stocking). For additional information on study design, plot layout, and site characteristics, see Kabrick and others (2011, 2015).

Data Analyses

For each planting site, survival probabilities in relation to pedigree, vegetation control, or overstory treatment were determined using logistic regression for the first- and seventh-year data (table 1). We used a generalized linear mixed model with a binary distribution (survival = 1, mortality = 0), logit link function, and random intercept using PROC GLIMMIX in SAS 9.4 software (SAS Institute Inc., Cary, NC). Treatment effects on the first- and seventh-year tree heights and diameters were analyzed using a generalized linear mixed model with a lognormal distribution, identity link function, and no intercept, using PROC GLIMMIX in SAS 9.4. In these models fixed effects included treatments and random effects included the statistical blocks (Wurdack, LTSP) or treatment units (Sinkin). Least-square means of survival probabilities and tree heights were subjected to pairwise comparisons using the Tukey-Kramer adjustment procedure for multiple comparisons (Kramer 1956) at alpha = 0.05.

RESULTS

First-year survival probability at the LTSP installation was about 60 percent and did not differ significantly ($p = 0.30$) among VC treatment (fig. 1). At the Sinkin installation, first-year survival for seedlings that were shorter than their nearest competitor was about 63 percent and significantly lower ($p < 0.01$) than for seedlings that were similar in height or taller than nearest competitors regardless of overstory stocking level. At Wurdack, seedlings with No-VC had mean survival of probability of 75 percent and was significantly lower ($p < 0.01$) than for seedlings with vegetation control regardless of pedigree.

The same trends among treatments observed during year 1 were also observed at year 7 although the survival probability in the No-VC treatments decreased during the sampling interval (fig. 2). Survival probability from year 1 to year 7 remained above 95 percent for all Wurdack families in the VC treatment, as well as all Sinkin trees taller than their nearest competitor. At Sinkin, the presence of taller competitors reduced survival probability to 35 percent. Several treatment categories appeared to have had an increase in survival probability from year 1 to year 7 from stump sprouts following damage or top kill that had occurred since the first data collection.

Mean height in year 1 was highly variable across studies and treatments (fig. 3). There was no significant difference ($p = 0.99$) in height between VC treatments at LTSP, where first-year height averaged 55 cm. At the Sinkin, underplanted seedlings with an overstory stocking <15 percent had a mean seedling height of 44 cm, but seedlings were not significantly greater ($p > 0.05$) than for other treatments which averaged 29 cm. At Wurdack, mean seedling height was about 70 cm in the No-VC treatment but was nominally (but not

Table 1—Site, primary treatment, vegetation control parameter, and number of trees used in analyses

Site	Primary treatment	Vegetation control	Trees
			n
Wurdack	Genetically improved	Yes	736
Wurdack	Genetically improved	No	756
Wurdack	Orchard-run	Yes	872
Wurdack	Orchard-run	No	408
Sinkin	Stocking level <15%	No	480
Sinkin	Stocking level 15-45%	No	510
Sinkin	Stocking level >45%	No	540
LTSP	-	Yes	384
LTSP	-	No	379

Sites are Wurdack Research Center, Sinkin Experimental Forest, and a Long-Term Soil Productivity (LTSP) site, Missouri.

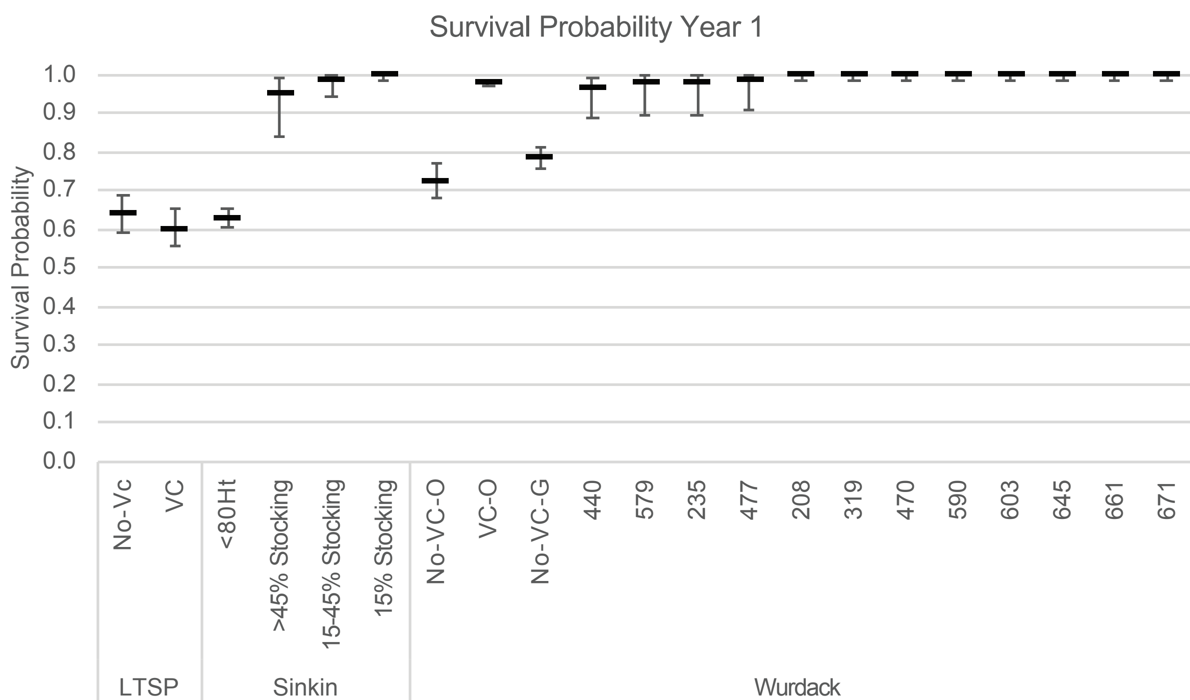

Figure 1—Survival probability 1 year after planting. Error bars represent ± one standard error of the mean. Sites and treatments are defined in the text.

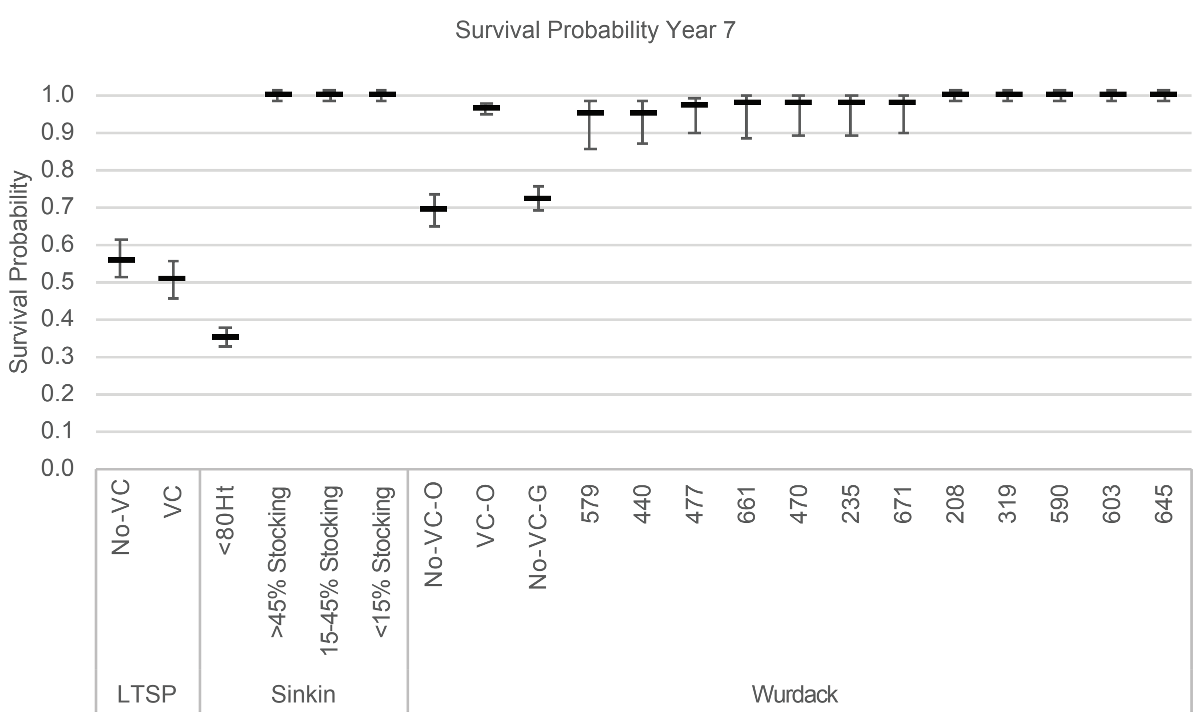

Figure 2—Survival probability 7 years after planting. Error bars represent ± one standard error of the mean. Sites and treatments are defined in the text. Some probabilities may be higher than year 1 due to stump sprouting between measurements.

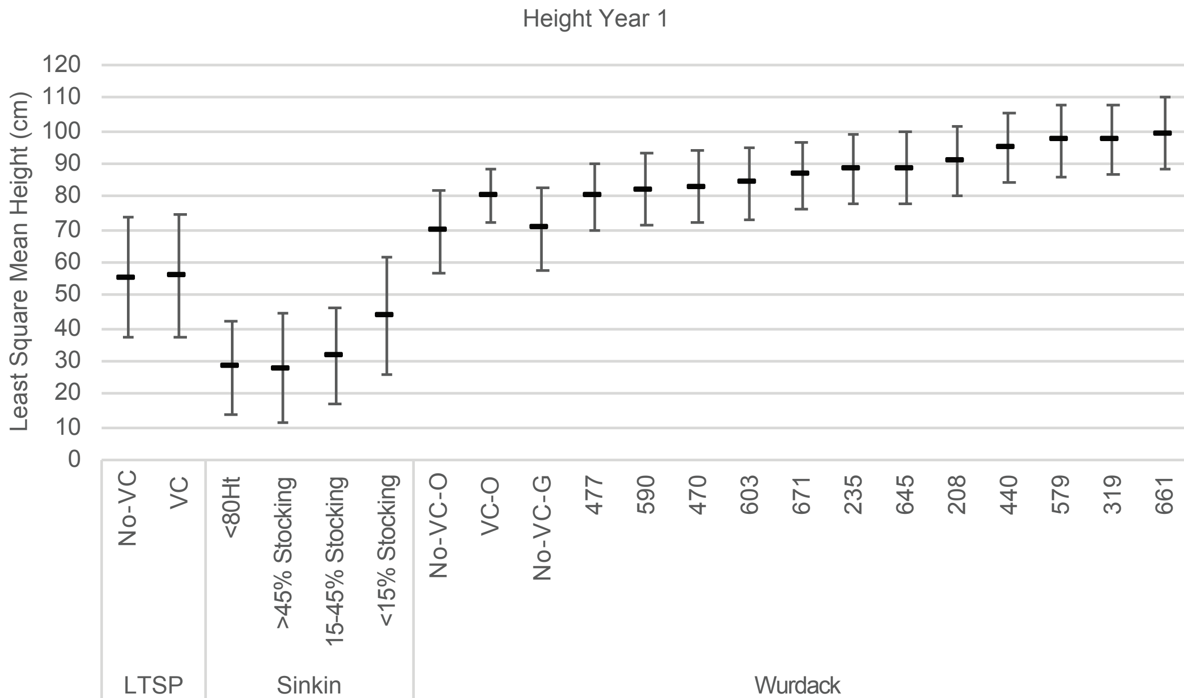

Figure 3—Mean tree height 1 year after planting. Error bars represent ± one standard error of the mean. Sites and treatments are defined in the text.

significantly) greater where VC treatments were applied. At Wurdack, the top five families had a mean height of 96 cm.

At year 7 there were no significant differences ($p > 0.05$) in seedling heights by VC treatment at LTSP and at Wurdack (fig. 4). However, at the Sinkin study, seedlings that were shorter than their nearest competitor were also significantly shorter ($p = 0.01$) than seedlings in the other groups, with a mean height of 84 cm. For seedlings that were as tall as or taller than their nearest competitor, seedling height increased significantly ($p < 0.01$) with decreasing overstory stocking. When the overstory was eliminated, mean seedling height was similar to all but the top five pedigreed families, with an average of 463 cm and the top five families had an average height of 503 cm after 7 years.

At year 1 there were no significant differences ($p > 0.05$) in basal diameter among treatments at each site but there were notable trends (fig. 5). For underplanted seedlings at the Sinkin, the mean basal diameter was 4.5 mm and it appeared to be greatly reduced by the presence of a partial overstory. For pedigree stock at Wurdack, No-VC had a mean of 14 mm and the addition of VC nominally increased mean basal diameter to > 18 mm. Two families had a nominally greater basal diameter, with a mean of 24 mm. Basal diameter was not recorded in year 1 at the LTSP sites.

By year 7, diameter at breast height (d.b.h.) was significantly increased by VC treatment at LTSP ($p < 0.01$) but only nominally increased ($p < 0.05$) at Wurdack where seedling diameter was greater than 80 mm (fig. 6). At Sinkin, underplanted trees with overstory stocking <15 percent averaged 73 mm d.b.h., and had significantly greater d.b.h. ($p > 0.01$) than seedlings having a greater overstory stocking or seedlings that were shorter than understory competitors.

DISCUSSION

Survival

Shortleaf pine seedling survival at 1 and 7 years following planting was primarily affected by the presence of understory competition (figs. 1 and 2). This finding is similar to Blizzard and others (2007), who reported that understory competition density had a greater effect on naturally regenerated shortleaf pine seedling survival than did overstory density. Similarly, Kabrick and others (2015) reported overstory stocking had no significant effect on shortleaf pine survival during the first five years after planting. Genetic selection and site preparation do not appear to have increased survival in this study, as underplanted seedlings within 80 percent of their competitor's height exhibited similar survival probabilities at the Sinkin study regardless of overstory stocking, while seedlings planted in prepared plots at LTSP had significantly lower survival than prepared plots at Wurdack (Lyczak 2019). Further, when pedigreed

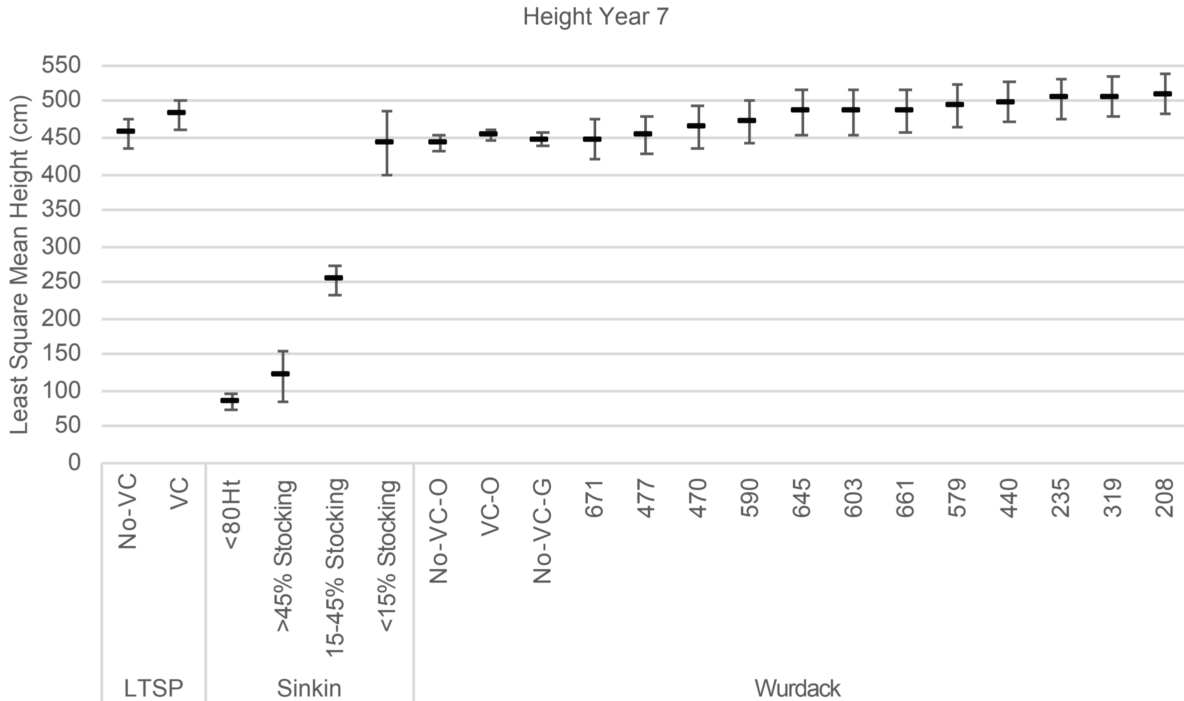

Figure 4—Mean tree height 7 years after planting. Error bars represent ± one standard error of the mean. Sites and treatments are defined in the text.

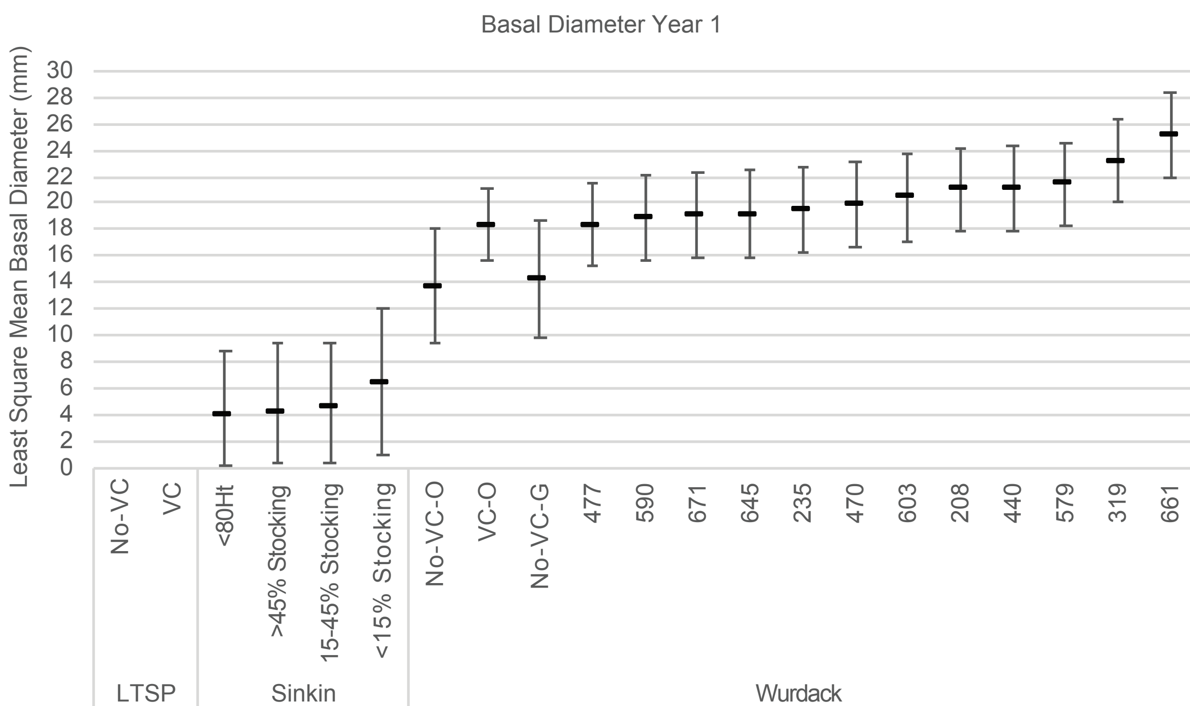

Figure 5—Year 1 mean basal diameter. Error bars represent ± one standard error of the mean. Sites and treatments are defined in the text. Diameter readings were not recorded at LTSP until year three.

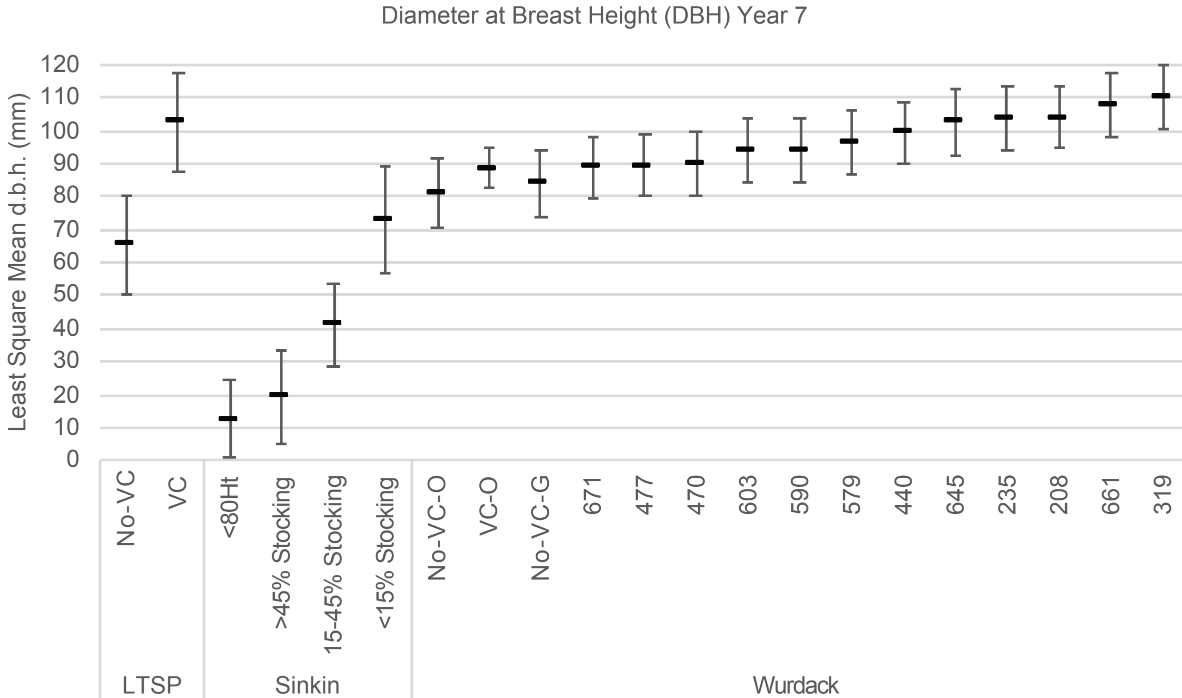

Figure 6—Year 7 diameter at breast height (d.b.h.). Error bars represent ± one standard error of the mean. Sites and treatments are defined in the text.

seedlings were planted without VC, survival was greater by more than 20 percent in year 1, and 30 percent by year seven when compared to the same families in herbicide treatments at Wurdack. These survival numbers are surprisingly and substantially lower than those reported by Brissette and Barnett (2003), who observed 90 percent survival 10 years after planting on half-sib shortleaf pine in northern Arkansas. Other site preparation methods, including ripping and prescribed fire, also result in mean first-year survival rates over 90 percent, suggesting a possible alternative to herbicide application (Cain and Shelton 2000, Gwaze and others 2006a). Though overall survival probability at LTSP was 60 percent, irrespective of VC, we believe this may be due to unique conditions encountered at planting, which included drought and rodent damage (Ponder 1997), rather than a result of treatments.

Height Growth

Height growth is a highly heritable trait in shortleaf pine, according to a study of progeny selected from phenotypically superior wild trees at a first-generation seed orchard in Missouri (Gwaze and others 2005). It was therefore surprising that we found only 2 of 12 full-sib families outperformed standard seedlings after 7 growing seasons. Pedigreed seedlings were only nominally taller after the first year compared to orchard-run seedlings (fig. 3). By the seventh growing season, however, only two families (319 and 208) were nominally taller than orchard-run seedlings (fig. 4). Underplanted seedlings that were shorter than competitors or that had moderate to high levels of overstory stocking appeared to have the greatest height reductions compared to seedlings grown in the open. Unlike survival, the presence of overstory with greater than 45 percent stocking significantly reduced mean height by nearly half, while seedlings that were shorter than a nearby competitors, regardless of overstory density, had a mean height reduction of almost 85 percent. In other studies, site preparation and herbicide application provided the most significant height growth gains for other southern pines (Land and others 2004, Lewis and others 1985, Schubert and others 2004, South and others 2004). As these stands continue to develop, early height growth, especially that above competing vegetation, is an important element in evaluating whether shortleaf pine will recruit to the overstory upon canopy closure (Raley and others 2003).

Diameter

Diameter growth followed a similar trend to that of height (figs. 5 and 6). Greater differences in diameter among treatments suggests that seedling diameter growth may be more sensitive to competition than is height. Year 1 basal diameter was nominally greater in pedigreed seedlings than other treatments, although seedling production and planting methods may account for these differences. Diameter was not measured on the LTSP site until year 3, so we were unable to compare herbicide and site preparation treatments. However, pedigreed seedlings planted without VC at Wurdack had nominally smaller basal diameters (14 mm) than pedigreed seedlings with VC (18 mm) at year 1. When the Sinkin trees are compared to those at Wurdack, it appears that seedlings with any level of overstory will have substantially reduced diameter growth during the first year. By year 7, diameter at breast height (d.b.h.) remained low where a partial overstory was present (Sinkin) but did not vary by pedigree where trees were grown in the open (Wurdack). Heritability estimates for diameter are lower than those for height, which is supported by these results (Gwaze and others 2005). The presence of understory competition significantly reduced d.b.h., even when trees were taller than their nearest competitor (Sinkin) or as tall as site trees in VC plots (LTSP). Diameter growth is considered to be less important than height growth during the stem exclusion stage of stand development (see Oliver and Larson 1996). However, larger diameters are associated with higher survival probabilities, increased resistance to fire and pest damage, as well as greater resilience to the effects of competition (Brinkman and Rodgers 1967, Brinkman and Smith 1968, Kabrick and others 2015, Lawson 1990, Stambaugh and others 2007).

CONCLUSIONS

We found that competition remains the most important factor reducing survival and growth of planted shortleaf pine seedlings. While orchard-run seedlings free from understory competition have similar survival and height growth to genetically improved seedlings, they have nominally lower overall diameter growth. The use of orchard-run stock, in conjunction with understory vegetation competition control during the first several growing seasons, should allow for successful shortleaf pine establishment under most site conditions encountered in Missouri.

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