AGE AND SIZE COMPARISONS OF REGENERATING SHORTLEAF PINE SEEDLINGS BURNED MULTIPLE TIMES IN ECOSYSTEM RESTORATION AREAS

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Abstract—Shortleaf pine (*Pinus echinata* Mill.) ecosystem restoration has been a major management goal in the Ouachita National Forest since the early to mid-1990s. Restoration efforts have focused on periodic prescribed burning and thinning operations to restore disturbance dependent vegetation communities suitable for the recovery of the endangered red-cockaded woodpecker, as well as other species able to utilize open forests and woodlands. Questions exist about how frequent burning will affect recruitment of shortleaf pine regeneration into larger age and size classes. Determining shortleaf pine's sprouting capabilities in response to repeated periodic prescribed burns could aid managers in recruitment of new age classes, while still promoting fire-dependent vegetation communities. The three stands in this study have undergone restoration activities for the past 6 to 12 years. The objectives of this study were: (1) to compare shortleaf pine root and shoot age, weight, and volume values, as well as root diameter, basal diameter, sprout height growth, live foliage weight, regeneration densities, and sprout production parameters in three analogous stands, and (2) develop regression equations for predicting sprout height following a burn from seedling root characteristics. Root ages were older than stem ages, and there were statistical differences in root ages indicating that the majority of seedlings and saplings regenerated following the same event. Seedlings and saplings in the stand that had been burned fewer times with a lower overstory basal area were characterized by larger morphological characteristics than seedlings in stands with greater burn frequencies for most variables.

INTRODUCTION

Restoration of shortleaf pine (*Pinus echinata* Mill.) communities has gained momentum over the last several years across the Ouachita and Ozark Highlands of Arkansas, Missouri, and Oklahoma. Historically, fire has been an important anthropogenic disturbance in the Ouachita Highlands. Fire return intervals across the area prior to European settlement around 1820 were 7-20 years, from 1820-1920 2 to 4 years, and from 1920-2000 4 years or longer, but this estimate is very area dependent with some locations having 50-year plus return intervals (Guyette and others 2006, Johnson and Schnell 1985, Stambaugh and Guyette 2006). Efforts to restore shortleaf pine-bluestem ecosystems on the Ouachita National Forest (NF) began in the region approximately 20 years ago with a primary goal of restoring the endangered red-cockaded woodpecker, but a variety of species of flora and fauna benefit from this work. Strong timber markets in the region promote restoration treatments such as midstory removals and prescribed burning because a portion of the income from harvests are allowed to fund the restoration treatments through the Knutson-Vandenberg Act of 1933 and the National Forest Management Act of 1976 (Bukenhofer and Hedrick 2013, Bukenhofer and others 1994, Guldin and others 2004, Hedrick and others 2007). This restoration work is scheduled for roughly 268,000 acres, or 20 percent, of the 1.8-million acre Ouachita NF, and it is estimated that roughly 50,000 acres are fully restored to date (Hedrick and others 2007, Zhang and others 2012).

Shortleaf pine seedlings regenerating in many of these restoration areas are damaged by prescribed burning every two to three years on average and by logging activity during thinnings (Lilly 2010). The species' ability to sprout may assist in the survival of a young cohort after a disturbance, but little is known about the number of times seedlings can resprout after repeated burns. One year-old seedlings achieve variable survival rates of about 40-90 percent or better after late dormant season burns, but burn timing, intensity, and seedling age can drastically alter survival rates (Cain and Shelton 2000, Clabo 2014, Little and Somes 1956, Shelton and Cain 2002). Older seedlings should have higher growth and survival rates compared to younger seedlings due to thicker bark, larger, more developed basal crooks that contain more dormant buds, and larger root systems

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based on a collection of past studies and observations (Cain and Shelton 2000, Lilly and others 2012, Little and Somes 1956).

Knowledge of how time since a burn and number of burns affect seedling survival and growth is ambiguous at best. Early in the twentieth century, Mattoon (1915) reported that seedlings can resprout at least three times from three individual disturbances even though the type of disturbance and survival rates following the disturbance(s) were not identified. The same account states that shortleaf pine seedlings can completely make up for height lost in a disturbance within 2 to 4 years. In pine-bluestem restoration areas of Arkansas, seedlings have resprouted 6-8 times in association with each successive burn. Some of these seedlings have had a 3.1 inch diameter root with a stem only 3.2 feet tall, which indicates a much older root system than stem (Lilly 2010). The previous findings were observations and were not carried out with an organized study design in areas with varying frequencies and intervals of burn occurrences. A non-replicated study in New Jersey in the 1930s found that 46 percent of shortleaf seedlings can produce ¼ to ¾ of their pre-disturbance height in less than a year following a late spring burn and 86 percent can produce ¾ of their pre-burn height or more within 2 years (Moore 1936).

Several important questions remain concerning shortleaf pine regeneration and sprouting in restoration areas. The first is whether managers must interrupt cyclic prescribed burning in order for saplings to grow large enough to withstand burns and develop into merchantable size classes. The second question is whether seedlings can survive successive burns and reliably resprout. Therefore, the objectives of this study are (1) to compare shortleaf pine root and shoot age, weight, and volumes, as well as root diameter, basal diameter, sprout height growth, live foliage weight, regeneration densities, and sprout production parameters in three analogous stands with a differing number of occurrences and time lapses since prescribed burning last occurred, and (2) develop regression prediction equations for each stand for sprout height following a burn from a variety of seedling root characteristics.

METHODS

This study was conducted on the Ouachita NF on the Poteau/Cold Springs Ranger District in Scott County, Arkansas. The Ouachita Mountains are characterized by east to west oriented ridges with broad U-shaped valleys. Elevations range from 500 to 2,700 feet (Hedrick and others 2007). Three stands that had received varying amounts and intensities of restoration treatments, such as midstory removals, regeneration harvests, thinnings, and prescribed burns for the last 6 to 12 years were chosen for this study. All three stands were within 6 miles of one another and on similar landforms. Shortleaf pine was the dominant overstory species in all three stands.

Inventories of the three stands were conducted on July 22-24, 2013. The first stand was located the farthest east of the three stands and was located on the Buffalo Creek Road (34°48'31.44"N 94°02'21.95"W) on a south aspect at an upper slope position. The soils in this stand are typic hapludults and lithic dystrudepts consisting of the Carnasaw Series, Carnasaw-Sherless Complex, and Carnasaw-Sherless-Clebit Complex types. The site index for shortleaf pine on these sites is 60 to 65 feet at base age 50 years (Vodrazka 1998). Prescribed burns were completed on 29 August 2002, 25 March 2006, 30 June 2009, and 22 April 2013. The second stand was located to the west of stand one along the Buffalo Creek Road and had a similar aspect and slope position as stand one. A commercial thinning was conducted in spring 2001 and left 60-70 square feet per acre of basal area. Prescribed burns were completed on 15 March 2003, 27 February 2007, 19 March 2010, and 8 September 2011. Stand 3 was located farthest west of the three stands adjacent to the Boles Motorway (34°48'46.11"N 94°09'42.16"W). This stand had a similar aspect, slope position, and contained similar soils as stands 1 and 2. A shelterwood harvest was completed on stand 3 in July 2007, leaving about 30 square feet of basal area per acre. Prescribed burns were completed on 4 March 2010 and 27 February 2012.

Sampling grids were originated at a random point located at least one chain from a stand boundary. Plots were located every two chains along a predetermined azimuth perpendicular to the prevailing elevation gradient in the stand. In between lines separated plots by two chains resulting in a 12x6 chain grid totaling 18 plots per stand. Each sampling location was a 1/100th acre circular plot. All shortleaf pine seedlings less than or equal to 4.5 feet tall were counted to estimate per acre seedling densities. The closest seedling to plot center (regardless if the seedling was located in the 1/100th acre plot) was measured for height, basal diameter, and the number of sprouts. A note was recorded if the seedling had a dead stem still present alongside the live stem(s). This seedling was then removed from the ground with a shovel in a manner so as to preserve as much of the root system as possible.

The extracted seedlings (N=54), were returned to the lab for additional measurements. All needles were removed from the seedlings in order to facilitate stem and root volume measurements. Needles from three random seedlings in each stand were kept and oven dried to a constant weight in a VWR Scientific 1380 FM forced air oven for 48 hours at 97 degrees F and then weighed. Taproot length and longest lateral root length were measured to determine taproot and lateral root length

thresholds for root volume measurement. A threshold was set because not all of the root system could be extracted in many instances because of the rocky soil conditions at some plot locations. Average length and standard deviation statistics were used to select the threshold lengths. The threshold length was set at 5.1 inches for taproots and lateral roots. Root systems were then severed from the stems for volume determination using the water displacement technique outlined in Burdett (1979). Volume determinations were done only for live stems and roots.

Stems and roots from stands 1 and 2 were oven dried to a constant weight for 48 hours at 124 degrees F. Stems and roots from stand 3 were oven dried to a constant weight for 72 hours at 140 degrees F due to their larger average size. Stems and roots were then weighed to the nearest ounce. Root diameter was measured and marked at the widest point of the basal crook. Roots and stems were aged following volume and weight measurements. All roots were examined prior to cutting for aging to determine how well the basal crook was developed. Well-developed basal crooks were those where the angle of deflection from upright on the stem was 45 to 90 degrees (Will and others 2013). Forty-nine of the 54 seedlings returned to the lab had well-developed basal crooks. The five seedlings that did not have well developed crooks all had nearly vertical taproots, which may indicate intermediate morphological traits that occur when hybridization with loblolly pine occurs (Tauer and others 2012). For seedlings that had well-developed basal crooks, cuts were made at the widest point of the basal crook and at the point below the basal crook where the taproot turns vertical again. For seedlings without a well-developed basal crook, the root was cut at the widest point of the taproot. Stems were aged just above the root collar. Sections were then sanded using a Delta® Shopmaster belt/disc sander with 320 and 400 grit sandpaper until rings were clearly visible. Sections were examined and

Table 1—ANOVA means and standard errors in parentheses are presented for each shortleaf pine seedling variable by stand for the shortleaf pine restoration study on the Ouachita National Forest in Arkansas, 2013. A variable name followed by an "*" indicates significant differences among stands

Stand	Root Volume *(Fluid Ounces)**	Stem Volume *(Fluid Ounces)**	Stem Weight *(Ounces)**	Root Weight *(Ounces)**	Basal Crook Widest Point Average *(Inches)**
One	0.8 (+/-0.21)	0.06 (+/-0.017)	0.05 (+/-0.02)	0.68 (+/-0.17)	0.72 (+/-0.08)
Two	0.62 (+/-0.15)	0.23 (+/-0.06)	0.17 (+/-0.05)	0.45 (+/-0.12)	0.78 (+/-0.09)
Three	3.06 (+/-0.81)	0.77 (+/-0.21)	0.67 (+/-0.2)	2.02 (+/-0.56)	1.26 (+/-0.15)

Table 1 Continued—ANOVA means and standard errors in parentheses are presented for each shortleaf pine seedling variable by stand for the shortleaf pine restoration study on the Ouachita National Forest in Arkansas, 2013. A variable name followed by an "*" indicates significant differences among stands

Stand	Stem Age *(Years)**	Root Age Average *(Years)*	Root Age at Widest Point of Basal Crook *(Inches)*	Root Age Below the Basal Crook *(Inches)*	Sprout Number
One	1 (+/-0.08)	9.82 (+/-0.71)	10.35 (+/-0.74)	10.09 (+/-0.71)	10.89 (+/-2.3)
Two	3.71 (+/-0.27)	7.94 (+/-0.6)	9.06 (+/-0.64)	8.56 (+/-0.57)	3.42 (+/-0.74)
Three	4.58 (+/-0.35)	9.73 (+/-0.76)	10 (+/-0.78)	9.87 (+/-0.76)	10.71 (+/-2.41)

Table 1 Continued—ANOVA means and standard errors in parentheses are presented for each shortleaf pine seedling variable by stand for the shortleaf pine restoration study on the Ouachita National Forest in Arkansas, 2013. A variable name followed by an "*" indicates significant differences among stands

Stand	Tallest Sprout *(Feet)**	Basal Diameter *(Inches)**	Needle Weight *(Ounces)**	Seedlings Per Acre*
One	0.87 (+/-0.1)	0.1 (+/-0.02)	0.21 (+/-0.08)	74.5 (+/-47.6)
Two	1.64 (+/-0.18)	0.24 (+/-0.03)	0.53 (+/-0.2)	11.2 (+/-7.7)
Three	2.43 (+/-0.27)	0.46 (+/-0.06)	0.73 (+/-0.27)	4099.5 (+/-2750.3)

aged using a magnifying glass or a Fisher Scientific binocular microscope.

Multivariate analysis of variance (MANOVA) was used to test for differences among the following dependent variables: stem volume, root volume, stem weight, root weight, basal crook average width, stem age, root age at the widest point of the basal crook, average root age, tallest sprout height, and sprout number. A MANOVA was used to reduce the likelihood of Type I errors that can occur when using many univariate analysis of variance (ANOVA) analyses by themselves. MANOVA also takes into consideration the correlation of closely related variables. ANOVA was used to test for stand differences if the MANOVA found significant differences initially in the dependent variables. In addition, a one-way ANOVA was used to test for differences in needle weights and seedling density per acre among the three stands. Tukey mean separation (alpha=0.05) was used for all ANOVA analyses. Log transformations were completed as necessary for non-normality issues and back transformed estimates were reported. Multiple linear regression was used to test which root variables were the best predictors of sprout height in seedlings that had been topkilled. Tested variables included: taproot length, taproot diameter, largest lateral root diameter, number of lateral roots, and longest lateral root length. All analyses were completed using SAS 9.4.

RESULTS

The four MANOVA tests indicated significant differences among the three stands. Wilk's Lambda ($p<0.0001$), Pillai's Trace ($p<0.0001$), Hotelling-Lawley Trace ($p<0.0001$), and Roy's Greatest Root ($p<0.0001$) were all significant, thus individual variable ANOVA tests were conducted with stand as a fixed factor. Results are presented in Table 1. Stem volume ($p<0.0001$) and root volume ($p=0.0002$) both displayed significant differences across stands, with seedlings in stand three having much greater averages than stands 1 or 2. Stand 2 had the smallest root volume average with 0.62 fluid ounces displacement, whereas stand 1 had the smallest stem volume average with 0.06 fluid ounces displacement. Stem ($p<0.0001$) and root weight ($p=0.0008$) were significantly different as well. Again, stand 3 had the greatest average weights. Stand 1 had the smallest average stem weight (0.05 ounces), and stand 2 had the smallest average root weight (0.45 ounces). There were significant differences among stands for the widest average point of the basal crook ($p=0.0031$). Stand 3, displayed the greatest average width at 1.26 inches, while stands 1 and 2 were statistically similar. Stem age significantly differed among stands ($p<0.0001$). Stand 1 stems averaged one-year-old making them the youngest on average. Stands 2 and 3 were statistically similar and averaged 3.7 and 4.6 years-old on average.

There were no significant differences in average root age ($p=0.1621$), root age at the widest point of the basal crook ($p=0.3644$), or root age below the basal crook ($p=0.059$). Root ages for all three variables ranged from 7.9 to 10.3 years old across stands. Sprout number displayed significant differences among stands ($p<0.0004$). Stands 1 and 3 were statistically similar (10.89 and 10.71 sprouts per seedling), but stand 2 differed in sprout production with 3.42 sprouts per seedling on average. Sprout height was statistically different across the three stands ($p<0.0001$). Stand 3 had an average sprout height of 2.43 feet, stand 2, 1.64 feet, and stand 1, 0.87 feet. Significant differences in average basal diameter were found as well ($p<0.0001$). Stand 1 had the smallest basal diameters on average at 0.1 inches, followed by stand 2 at 0.24 inches, and finally stand 3 at 0.46 inches. There were no statistical differences in average needle weights across stands ($p=0.1161$). There were significant differences in seedling densities across the three stands ($p<0.0001$). Stands 1 and 2 were statistically similar (74.5 and 11.2 seedlings per acre), whereas stand 3 had a much greater density of 4099.5 seedlings per acre.

The multiple linear regression results for predicting sprout height in feet following a burn by taproot length, lateral root number, longest lateral root length, largest lateral root diameter, and taproot diameter had different results for stand 1 as compared to stands 2 and 3 which were very similar. For stand 1, largest lateral root diameter and taproot diameter did not contribute to prediction of sprout height and were thus removed from the model (table 2). These three variables predicted 84 percent of sprout height differences. Root variables from stand 1 predicted the following equation for sprout height:

(1) $\text{Sprout Height} = 0.25 + 0.024a + 0.024b + 0.054c$
where a=taproot length in inches, b=number of lateral roots in inches, and c=longest lateral root length in inches.

Regression parameters for stand 1 are found in Table 2. Largest lateral root diameter was the only significant prediction variable for stand 2. This variable predicted 56 percent of differences in sprout height. This root variable produced the following regression equation for stand 2 for prediction of sprout height:

(2) $\text{Sprout Height} = -0.0409 + 2.63a$
where a=largest lateral root diameter in inches.

Largest lateral root diameter was also the only variable for stand 3 seedlings that contributed to a prediction of sprout height following a burn (table 2). This variable predicted 41 percent of the differences in sprout height.

Table 2—Multiple regression results for predicting dominant shortleaf pine sprout height in stands 1, 2, and 3 for the shortleaf pine restoration study on the Ouachita National Forest in Arkansas, 2013

Stand 1: (R^2=0.84)	Parameter Estimate	SE	t-value	p-value	Partial R-Squares
Intercept	0.25182	0.09176	2.74	0.0158	—
Taproot Length	0.02391	0.01119	2.14	0.0507	24.607
Lateral Root Number	0.02422	0.01031	2.35	0.0339	28.293
Lateral Root Length	0.05359	0.01078	4.97	0.0002	63.813
Stand 2: (R^2=0.56)					
Intercept	-0.0409	0.144	-0.34	0.74	—
Largest Lateral Root Diameter	2.63	0.585	4.5	0.0004	—
Stand 3: (R^2=0.41)					
Intercept	1.35	0.46	2.92	0.01	—
Largest Lateral Root Diameter	3.34	0.99	3.37	0.0039	—

This root variable produced the following equation for stand 3 for prediction of sprout height:

(3) Sprout Height = 1.35 + 3.34a
where a=largest lateral root diameter in inches.

MANAGEMENT IMPLICATIONS AND CONCLUSIONS

Past research has shown that prescribed fire is the cheapest and least intensive site preparation or intermediate operation to achieve suitable shortleaf pine regeneration densities (Yocom and Lawson 1977). The fire return interval averaged across the three stands in this study was just over three years. The lack of significant differences among root ages indicate that the majority of seedlings regenerated around the same time before most of the burns associated with restoration activities began. The younger stem ages in comparison to the roots and the presence of a dead, charred stem on 96 percent of the seedlings sampled across stands indicates that prescribed burns are topkilling the majority of seedlings each time they occur. The multiple linear regression equations indicate that fewer root parameters are important for predicting sprout height in stands that have been burned longer ago than more recently. In addition, stand 3, which had received two fewer burns than stands 1 or 2 when this study was conducted, tended to have larger seedling attributes across many of the variables that were measured even though it had the second shortest time lapse since a burn. One conflicting factor that may account for this finding is that stand 3 is past the seed cut of a shelterwood regeneration harvest, whereas the other two stands are still in late-rotation mature stand conditions; the larger size of saplings in stand 3 is thus not an unexpected finding. Those saplings are on their way to becoming a second age cohort in that regenerating stand. These findings indicate that burning intervals in restoration areas will have to be more variable until seedlings and saplings reach minimum age and size thresholds as suggested by Guldin (2007), Stambaugh and others (2007), and Walker and Wiant (1966) to avoid topkill. Walker and Wiant (1966) reported that saplings 2-6 years old can survive a moderate intensity burn if crown scorch is less than 70 percent, basal diameter is greater than or equal to 0.5 inches, and height is greater than or equal to 5 feet. Competition from hardwoods would likely not be a problem with slightly longer burn intervals as shortleaf pine can typically compete successfully with or outcompete hardwoods around ages 5-7 on shortleaf sites in the region (Cain 1991).

The much lower regeneration densities per acre in stands 1 and 2 as compared to stand 3 indicate that more periodic prescribed burns result in fewer regenerating seedlings and saplings per acre, but not necessarily overstory density. Stands 1 and 2 would be considered extremely understocked according to most guidelines (e.g. Blizzard and others 2007), indicating that some type of change in management will be necessary in order to achieve necessary seedling densities and stocking prior to the end of overstory timber rotations. Past research has shown newly established seedlings can persist under a partial or full overstory for a period of time with limited effects on

survival, but height growth will be negatively affected (Kabrick and others 2011, Shelton 1995). Other research by Fan and others (2012) on periodic prescribed burning in mixed pine-hardwood forests in Missouri has shown that shortleaf pine seedling and sapling mortality is high compared to associated species as indicated by the results in this study.

Thus, when the time eventually comes to regenerate stands similar to stands 1 or 2 with the shelterwood method, foresters should inventory the shortleaf advance growth to see if the seedling bank of shortleaf seedlings and saplings is sufficient to regenerate the stand. If not, some degree of reliance on seedfall from the seed trees in the shelterwood will still be needed to fully stock the new age cohort. This study and others indicate that after a good regeneration year burning should stop in restoration areas until seedlings reach adequate sizes and ages to survive burns.

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