CONE CHARACTERISTICS AND SEED QUALITY 10 YEARS AFTER AN UNEVEN-AGED REGENERATION CUT IN SHORTLEAF PINE STANDS

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Abstract—Cone characteristics and seed quality for 16 released (stand density 14 square meters per hectare) and 16 unreleased (stand density 28 square meters per hectare) shortleaf pine (*Pinus echinata* Mill.) trees were described by d.b.h. class (28, 33, 38, 43 centimeters) and crown position (upper south, upper north, lower south, and lower north). The 38-centimeter d.b.h. class produced significantly heavier cones than other classes. Average cone dry weight for released trees did not differ significantly by crown position, but the lower north crown position produced significantly lighter cones for unreleased trees. Total seeds per cone did not differ significantly between released and unreleased trees, by d.b.h. class, or crown position; the overall average was 46 seeds per cone. Upper crown positions produced a higher percentage of sound seeds per cone (61 percent) than the lower crown positions (50 percent) for both released and unreleased trees. The percentage of sound seeds also differed significantly between released and unreleased trees with the 38- and 43-centimeter d.b.h. classes of released trees producing the highest percentage. Both released and unreleased trees produced significantly more sound seeds per cone in the upper south crown position (31 seeds per cone) compared to the other crown positions (averaging 25 seeds per cone). Seeds from released trees averaged 91 percent germination compared to 85 percent for unreleased trees.

INTRODUCTION

Requirements for adequate quantities of viable shortleaf pine (*Pinus echinata* Mill.) seeds to naturally regenerate forests has increased interest in the cone producing ability of natural stands (Baker 1992). Flower induction is influenced by at least five factors: nutrient relationships, induction hormones, light conditions, soil moisture, and temperature (Barnett and Haugen 1995). A thinning or regeneration cut could positively affect three of the above variables: moisture, light, and nutrients. Yocom (1971) reported that removing all trees within 9.1 meters of shortleaf pine seed trees significantly increased the number of sound seeds per cone and doubled the average cone production per tree. Studies have found that pine seed quality is higher when seedfall is greatest (Stephenson 1963, Shelton and Wittwer 1996). A study of seed quantity and quality in shortleaf pine cones from two 15 hectare natural stands found 36 total seeds per cone with an average of 17.5 and 14.5 sound seeds per cone for single-tree selection and seed-tree stands, respectively (Wittwer and others 1997).

Dickmann and Kozlowski (1971) found seeds per cone for red pine (*Pinus resinosa* Ait.) to depend on the number of productive ovules, degree of pollination, and ovule abortion, and they concluded that the number of productive ovules was not highly dependent on the number of scales. A study of table-mountain pine (*Pinus pungens* Lamb.) found cone length did not affect the number of viable seeds, and there was no relationship between tree age, seed viability, or cone size (McIntyre 1929). A study on young open-grown Douglas-fir (*Pseudotsuga menziesii* (Mirb.) Franco) found the outer extremities of branches on the upper and middle south side of the crown had more cones and greater seed contents except for the west quarter of the crown (Winjum and Johnson 1964).

We conducted this study to determine if cone characteristics and seed quality in shortleaf pine vary by crown position, tree diameter, and release treatment. A better understanding of these relationships will be useful for selecting trees to retain for seed production in natural stands and will provide indicators for forecasting future seed crops.

STUDY AREA

The study area was located in the Ouachita National Forest on the Winona Ranger District, Perry County, AR. Before implementation of uneven-aged management, the stand was irregularly-aged with a uniform canopy dominated by shortleaf pine with mixed hardwoods in the mid to lower canopy. Sixteen uneven-aged management plots (0.20 hectare) were established between December 1988 and March 1989 reducing the overstory pine basal area from 27.6 to 13.8 square meters per hectare (Shelton and Murphy 1997). Plots received one of three residual hardwood basal area treatments (0, 3.4, and 6.9 square meters per hectare). Four plots with complete hardwood control were selected for this study. Each of the 0.20-hectare plots was surrounded by an 18 meter buffer zone receiving the same treatment.

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Shortleaf pine site index averaged 17.4 meters at 50 years. Shelton and Murphy (1997) have given a more detailed description of the study area.

METHODS

Tree Selection

Sixteen released trees were selected from the buffer zones of four treated plots, and sixteen unreleased trees were selected from the adjacent unharvested pine-hardwood mixed forest. Sample trees were randomly selected from 5-centimeter d.b.h. classes (28, 33, 38, and 43 centimeters). Sample trees with malformed crowns were excluded from selection. The unreleased stand had about 28 square meters per hectare of total basal area with two-thirds in shortleaf pine and the remainder in hardwoods. Sample trees were measured for height, d.b.h., crown length, crown width, and 5-year radial growth increment at stump height.

Cone Evaluation

Sample trees were felled during the middle of October 1998 when cones were mature but before seed fall. After felling, the crown was measured for total length and was equally divided into upper and lower halves. The crown was further divided into north and south faces creating four crown positions: lower north, lower south, upper north, and upper south. All branches were removed and separated by crown position, providing a complete population of cones for sampling. The four crown positions and four tree d.b.h. classes were considered treatments split between the released and unreleased treatments with four replicates of each treatment. Twenty cones with no visible defects were randomly sampled from each crown position for seed extraction. Very few crown positions failed to produce at least 20 normal cones; all cones were sampled when a shortage occurred. An additional 10 cones per crown position were sampled for dry weight determination. Cone measurements included length, diameter, dry weight, and volume. Cone volume was determined by water displacement. Other cone attributes evaluated were potentially productive scales per cone, total and sound seeds per cone, and percent germination of sound seeds.

Cones were allowed to air dry for 6 weeks and were then tumbled for 25 minutes. Cones were then oven-dried at 35 degrees Centigrade for 48 hours and tumbled for an additional 15 minutes. Most seeds were extracted prior to oven drying, which was a secondary measure to recover any additional seeds. The efficiency of seed extraction was tested by dissecting 80 randomly selected cones after processing; 2.3 seeds per cone were not removed with a coefficient of variation of 63 percent. Potentially productive cone scales, defined as being large enough for two enlarged sound or empty ovules, were counted after all seeds had been removed on 10 of the 20 cones sampled per crown position to determine the potential seed production capacity.

Seed Evaluation

Before further seed evaluation, wings and other inert matter were removed. A series of float tests were used to separate the empty seeds from the sound seeds. To test the efficiency of this process, 20 floating seeds were sampled from each crown position and cut to verify that they were empty. After cutting 2,460 floating seeds, only 1 percent appeared to be sound. Sound seeds were allowed to air dry for 3 days before storing in a refrigerator at 3 degrees Centigrade.

Following Association of Official Seed Analysts (1978) guidelines, a germination test was conducted using a sample of 200 sound seeds per crown position with four replicates of 50 sound seeds each when ample seeds were available. All seeds were used for quantities below 200. Seeds were soaked for 24 hours at 21.1 degrees Centigrade; after draining, seeds were placed in polyethylene bags and stratified for 28 days at 4.4 degrees Centigrade. After stratification, all replicates were placed into 4.5 x 4.5-centimeter dishes with a substrate of three layers of filter paper. Seeds were equally spaced to prevent the spread of fungi from infected seeds. Two milliliters of de-ionized water was added to each dish at the start of the germination test, and 0.20 milliliter was added every 7 days. Fungicide was applied on the fifth day of the germination test to contain mold spread. Light was provided 8 hours per day with a temperature of 30 degrees Centigrade. The remaining 16 hours per day coincided with a temperature of 20 degrees Centigrade.

Germination counts began on the fourth day and continued daily. Seedlings with radicles half the size of the seeds or

Table 1—Results of analyses of variance testing the effects of stand density, tree d.b.h. class, and crown position on cone dry weight, productive scales per cone, percent sound seed, sound seeds per cone, and germination percent

Source of variation	Cone dry weight	Productive scales per cone	Percentage sound seed	Sound seeds per cone	Germination percent
			P-value[a]		
D.b.h. class (D)	0.033	0.292	0.521	0.206	0.113
Stand density (S)	0.021	0.187	0.015	0.054	0.022
DxS	0.122	0.678	0.039	0.323	<0.001
Crown position (P)	0.001	<0.001	<0.001	0.010	0.484
DxP	0.303	0.700	0.463	0.201	0.121
SxP	0.044	0.929	0.934	0.677	0.164
DxPxC	0.293	0.506	0.945	0.964	0.363

[a]*P* level is the probability of obtaining a larger F-statistic

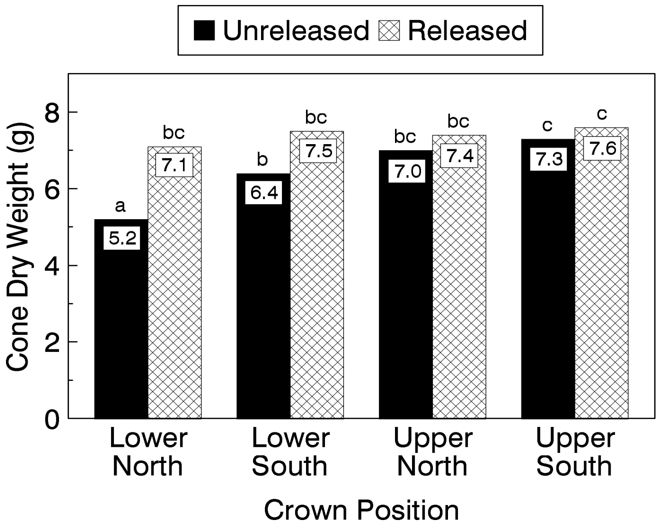

Figure 1—Shortleaf pine cone dry weight for released and unreleased trees by crown position.

longer were evaluated as normal or abnormal according to Association of Official Seed Analysts (1978) standards and removed daily. At the end of testing, seeds that had failed to germinate were cut to determine if they were full or empty. If the percent germination for a replicate deviated by 25 percent or more below the average of all replicates it was omitted from data analysis. Replicates were also omitted when 20 percent or more of the sound seeds were fungi filled. Only 12 replicates out of 450 had to be excluded due to fungi or deviation from the mean percent germination.

Data Analysis

Mean values for cone characteristics were calculated on a per crown position basis. When crown positions lacked enough cones for dry weight determinations, cones already processed for seeds were used; the weight of missing seeds was estimated from the seed weight of the crown position in question. Percent germination was transformed with the

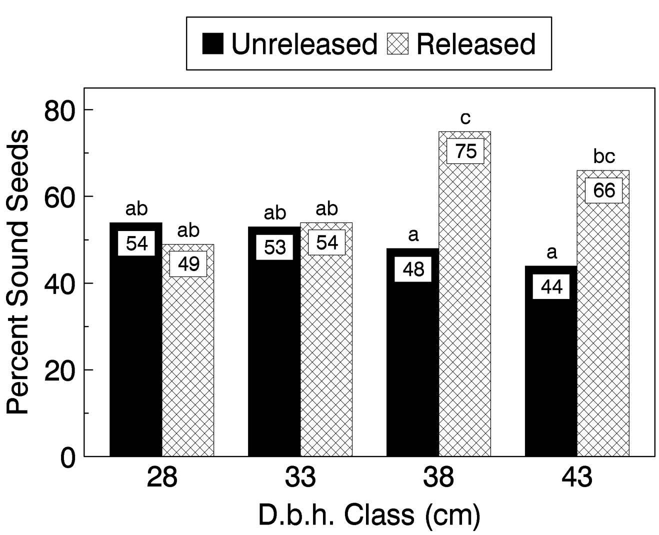

Figure 2—Percentage of sound seed in cones of released and unreleased shortleaf pine by tree d.b.h. class.

arsine square root transformation. The MIXED procedure from the SAS Institute (1997) was used to analyze the data. An analysis of variance for the split-split plot arranged in a randomized complete block design was used to make inferences about cone characteristics by crown positions, d.b.h. class, (split unit treatments) and stand density (main unit treatment). All variables were considered fixed for the mixed model except blocks. Significance was accepted at $\alpha = 0.05$. Multiple mean comparisons were attained by using the LSMEANS statement and DIFF (Fishers Least Significant Difference) and SLICE options (SAS Institute 1997). The SLICE option tests for simple effects; for example, if the interaction of factor A*B is significant, the effect of A for each level of B is tested. Occasionally the relative ranking of all means and their separation may appear contradictory. This may be due to the use of transformed data, multiple standard errors, and missing observations. Multiple standard errors are due to calculations used in the means comparison tests. Means presented in figures are least squares means or estimated means. The only exception is for percent germination, which uses arithmetic means because of the transformation. The arithmetic means and the least squares means will sometimes differ due to an unbalanced design (missing observations).

RESULTS AND DISCUSSION

Sample Tree Description

Average age (76 vs. 78 years) and height (20.4 vs. 20.1 meters) of released and unreleased shortleaf pine trees were very comparable (Grayson 2000). Crown width (8.2 vs. 7.3 meters) and length (10.1 vs. 9.1 meters) of released trees averaged slightly greater than the unreleased trees. Released trees contained four more branches per tree with a basal diameter of 2.5 centimeters and greater. Released trees grew 0.51 centimeter more than unreleased trees in radial increment at stump height over the last 5 years.

Cone Characteristics

Analyses of variance indicated no significant main effects or interactions for cone length, diameter, and volume, which averaged 4.8 centimeters, 2.2 centimeters, and 11.9 cubic centimeters, respectively. Dry weight of cones differed significantly by d.b.h. class, stand density, and crown position, and a significant interaction occurred between stand density and crown position (table 1). Cone dry weight averaged 7.5 grams per cone for released trees and 6.5 grams for unreleased trees. Trees in the 38-centimeter d.b.h. class produced significantly heavier cones at 8.0 grams compared to all other classes. Cone dry weight on released trees did not differ significantly by crown position, but this was not the case for unreleased trees (figure 1). For unreleased trees, the lower north crown position differed significantly from all other crown positions including the released trees.

The number of potentially productive scales indicates potential seed production with each scale capable of containing two ovules. An analysis of variance indicated a significant effect for crown position (table 1); means were as follows: lower north (53 scales per cone), lower south

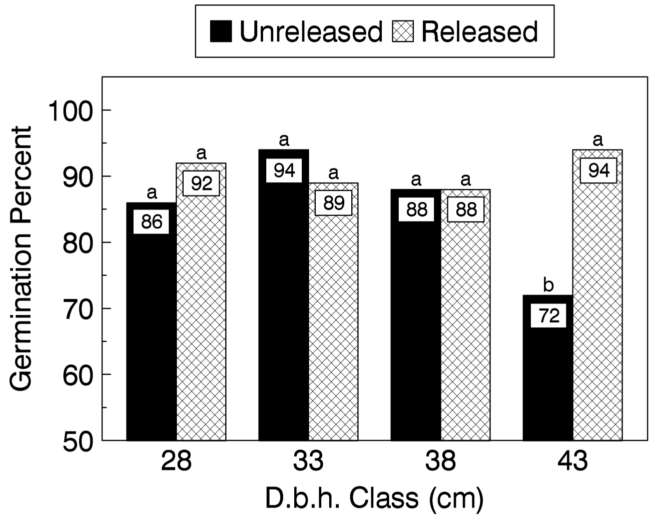

Figure 3—Germination percent of seed from released and unreleased shortleaf pine by tree d.b.h. class.

(55), upper south (56), and upper north (56). The small differences were probably related to differences in cone size by crown position. The total number of seeds per cone was not significantly influenced by tree d.b.h., stand density, or crown position; the overall average was 46 seeds per cone. Despite silvicultural manipulation in the released stand, a gain in total seeds per cone was not apparent.

Seed Characteristics

The percentage of sound seeds was significantly different for stand density, the interaction of stand density and d.b.h. class, and crown position (table 1). Trees in the released stand produced cones with a significantly higher percentage of sound seeds (61 vs. 51 percent of total). There was a trend for the larger trees to produce a higher percentage of sound seeds in the released stand (figure 2). The ranking and means for percent sound seeds by crown position were as follows: lower north (48 percent) = lower south (51) < upper north (59) = upper south (63).

Differences in sound seeds per cone were nearly significant for stand density ($P = 0.054$); averages were 22 seeds per cone for unreleased trees and 31 seeds per cone for released trees. Wakeley (1954) reported that cones average between 25 and 35 sound seeds during a good seed year for shortleaf pine, which is comparable to our results. Lyons (1956) reported that ovule abortion within the cones of red pine may be associated with nutritional factors. If this is the case, released trees in this study could have received a short-term increase in available nutrients, which should have increased overall tree vigor. The reduction in sound seeds for unreleased trees could be due to carbohydrate deficiencies or self-pollination, since both released and unreleased trees produced approximately the same number of total seeds per cone. Reduced air movement in unthinned stands might hamper pollen cloud dispersal and lead to increased self-pollination and embryo abortion.

Crown position was also significant for sound seeds per cone, and means were ranked as follows: lower north (23 sound seeds per cone) = lower south (26) = upper north (26) < upper south (31). According to Perry and Coover (1933), shortleaf pine cones from the top of the crown produced the most viable seeds (24 seeds per cone) followed by the middle crown (20), and finally the crown base (18). The greater sound seed yield in the upper south crown position is probably due to greater carbohydrate production where light levels are higher and growth is more vigorous when compared to other crown positions.

No significant differences in seed weight were found in our study. Dewinged sound seeds per gram ranged from 68 to 132 for released trees and 66 to 147 for unreleased trees, averaging 99 and 95 sound seeds per gram, respectively. Wakeley (1954) reported cleaned and de-winged shortleaf pine seeds average 106 seeds per gram with a range from 80 to 138.

Analysis of variance for germination of sound seeds indicated a significant main effect for stand density and a significant stand density by d.b.h. class interaction (table 1). Seeds from released trees averaged 91 percent germination compared to 85 percent for unreleased trees. Unreleased trees in the largest d.b.h. class (43 Centimeter) exhibited significantly lower seed germination than other size classes for both released and unreleased trees (figure 3). Cone production of trees in this class was low (Grayson 2000), and a positive correlation between cone and seed production and germination percent has been reported (Shelton and Wittwer 1996).

CONCLUSIONS

Cone length, diameter, and volume did not vary significantly by stand density, tree d.b.h., or crown position. Cone dry weight differed significantly between released and unreleased trees. Cone weight also differed significantly by tree d.b.h. class with the 38 centimeter d.b.h. class trees producing heavier cones compared to all other classes. The lower north crown position produced significantly smaller cones by weight compared to all other crown positions. The number of potentially productive scales per cone varied significantly by crown position with the upper north position produced significantly greater numbers. The lower north crown position produced significantly fewer potentially productive scales than all other crown positions. Overall, the differences in cone scale numbers by crown position were small and may not have silvicultural importance.

Percentage of sound seeds per cone differed significantly by stand density, the interaction of density and tree d.b.h., and crown position. The released trees in the 38-centimeter d.b.h. class and larger produced significantly greater percentages of sound seeds when compared to the lower diameter classes and unreleased trees. No significant difference was detected between diameter classes for unreleased trees. For released trees, the general trend was for higher sound seed percentages in the upper crown, with increasing tree diameter. Percentage of sound seed tended to decrease with increasing diameter for unreleased trees. Results for percentage of sound seed suggest that selecting larger diameter released trees, at least 36 centimeters in d.b.h. or greater, will increase seed quality. In addition, released trees produced on average 9 more sound seeds per cone than unreleased trees. The upper south crown position produced significantly more sound seeds per cone than other crown positions by 5 to 8 seeds per cone. Germination of seeds was not significantly affected by crown position or stand density, indicating that germination is consistent within the crowns of released and unreleased shortleaf pine.

The reduction in stand basal area to 14 square meters per hectare, the recommended stocking level in uneven-aged stands, had a pronounced effect on seed production within the stand 10 years later. Enhanced seed production is important to the success of uneven-aged management because regeneration depends on seeds produced by retained trees. Most seeds produced in this stand were on trees with d.b.h. greater than 35 centimeters. Thus, we recommend that maximum diameters used for regulating the stocking and structure of uneven-aged stands be over 35 centimeters. More seeds were produced in the upper canopy. Thus, forecasting systems relying on cone counts or ratings should focus on this portion of the canopy.

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