

ARTIFICIAL RIPENING OF LOBLOLLY PINE CONES AND SEEDS 1/

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Loblolly pine cones are considered to be mature and collectable when they will float in SAE 20 motor. However, there may be as little as 2 weeks from the time that the cones float until they open and shed their seed. The short duration of the collection period becomes increasingly critical in large seed production areas and seed orchards unless large supplies of manpower are available.

A possible solution to this short-period collection problem would be to collect the cones prior to maturity and to artificially ripen the cones and seed. Artificial ripening studies have proven successful with cones of other coniferous species.

The Eastern Tree Seed Laboratory in cooperation with the Georgia Kraft Company initiated a study to determine if loblolly pine cones and seed from early collections could be artificially ripened. Earl Belcher of the Laboratory and Joe Thompson of the Georgia Kraft Company were instrumental in carrying this study to completion.

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- 1/ Belcher, Earl W. and Joe Thompson. 1968. Artificial ripening of loblolly pine cones and seed. Final Office Report. Data on file at the Eastern Tree Seed Laboratory, Macon, Georgia.
 - 2/ Wakeley, P. C. 1954. Planting the southern pines. USDA, Forest Serv., Agr. Mono. 18, 233 pp.

Ten loblolly pine seed trees (left from a shelterwood cutting 5 years previous) were selected for use in this study. These trees, located on Georgia Kraft Company lands near Forsyth, Georgia, were vigorous individuals with large cone crops.

Cone collections were made from each tree starting on September 7, 1967, and continuing every fifth day thereafter as follows:

| | |
|--------------|--------------|
| Collection 1 | September 7 |
| Collection 2 | September 12 |
| Collection 3 | September 18 |
| Collection 4 | September 22 |
| Collection 5 | October 2* |
| Collection 6 | October 11* |
| Collection 7 | October 17* |

(*) - Limited collections made on these dates for the purpose of determining normal cone maturity dates.

Thirty-six cones per tree were collected on each date until maturity was reached (as determined by 90 percent flotation in SAE 20 motor oil). Immediately after collection, the cones of each tree were placed in a kraft paper bag. This bag was then placed in two thicknesses of polyethylene bags and transported immediately to the Eastern Tree Seed Laboratory where all analyses were made.

Upon arrival at the Laboratory, the cones were divided into six groups of six cones each per tree. Each group of cones received one of the following separate treatments. Treatments were:

- A. Immediate germination test of seed.
- B. Cones artificially ripened on racks for 30 days at 72°F.
- C. Cones artificially ripened on racks, covered with damp sphagnum moss, for 15 days at 38°F.
- D. Cones artificially ripened on racks, covered with damp sphagnum moss, for 30 days at 38°F.
- E. Cones artificially ripened on racks, covered with damp sphagnum moss, for 45 days at 38°F.
- F. Cones artificially ripened on racks with only the ° base in a moist medium (Kimpak) for 30 days at 38 F.

Cone specific gravity was determined from the Group A cones, using the water displacement method. The seed were then excised from Group A cones and the following notations made: (a) seed color and appearance, and (b) seed moisture content as determined on a dry weight basis.

Following each of the six ripening treatments, the cones were held at 72 F (room temperature) for 2 days and then placed in a forced-air-draft oven at 110 F for 24 hours. Seed from those cones unopened after the heat treatment were hand-extracted (by cutting) from the cones. After extraction, the seed from each group were divided into two equal samples and germinated as follows:

1. One-half of each sample tested unstratified.
2. One-half of each sample stratified 30 days in polyethylene bags prior to germination test.
3. All seed (unstratified and stratified) were germinated on two layers of Kimpak at 72°F with 16 hours of 160 foot-candles of light. The tests ran a minimum of 28 days.

An analysis of variance was made on the arcsin transformation of the full seed germination percentage at 28 days. Differences were tested at the 1 percent level of probability and Duncan's multiple range test was applied to the means of significant effects.

Specific gravity and seed moisture content were correlated to collection date and a regression was computed.

The data have been grouped by topic and are discussed in one of the following categories: collection trees, specific gravity and cone maturity, cone and seed characteristics, cone opening, and seed germination.

COLLECTION TREES

All trees, except one, showed little diameter growth for the last 5 years. They averaged 29 years in age and 15 inches in diameter. Total height was quite uniform, the average being 67 feet. The amount of crown, as computed by crown ratio, was uniform for 7 of the 10 trees.

No relationships between tree characteristics and seed maturity or germination were found,

SPECIFIC GRAVITY AND CONE MATURITY

Cones from the first collection were from 2 weeks to 6 weeks immature as determined by an oil flotation test of 10 cones per tree. Also, flotation in either oil or water could go from 0 to 90 percent of the cones tested, within the 5-day collection interval. Cones were considered mature when a flotation of 90 percent, or better, in SAE motor oil was reached. Once the cones of a tree had reached this maturity, no further collections were made from that tree.

Although flotation tests provided a reasonable measure of specific gravity, actual specific gravity was easier to correlate with collection date. A linear relationship was noted with an average of **0.30** decrease each **5** days in specific gravity. This indicates that cones will float in SAE 20 motor oil approximately **16** days after floating in water.

CONE AND SEED CHARACTERISTICS

Cone characters.--All cones were bright-green on the first collection date; but, with the exception of one tree, had turned to various shades of brown by the fourth collection date.

A wide range was observed in average cone size of the individual study trees.

In general, the range in cone weight paralleled the range in cone volume. The only measurable cone characteristic of cone ripening was the relationship of cone volume to cone weight (or specific gravity).

Seed characters.--Variation was noted in the seed at each collection date. At the first collection, seed from all trees was a tan-color. The seedcoat was smooth (no ridges) and the wings were generally a light-brown. When the seed were removed from the cone, they had only a wing stub attached.

By the second collection (September 12), the wings and seedcoat had darkened and ridges were noted on the seedcoat. The wings were developed enough to come out intact when the seed were removed from the cone.

The ridges on the seedcoat were more pronounced by the third collection and, in general, the seedcoats were mottled or all black. Wings could easily be removed from the excised seed of this collection as the seed began to dry. All seedcoats were thicker and darker by the fourth collection.

The cone, and also the seed, lost moisture as maturity was approached. The decrease in seed moisture was **3.64** percent every **5** days, as indicated by a regression of seed moisture with collection date.

CONE OPENING

Cones of Treatment A were opened by hand at the time of collection, so will not be considered here. Cone opening increased as the collection date increased with considerable variation from tree to tree. Most of the cones could be opened when the specific gravity

reached 1.00 (point of water flotation) or less, if they were collected after September 18 and were stored for **30** days or more in damp moss at 38°F (Treatments D, E, or F) prior to a heat treatment.

SEED GERMINATION

Average full seed germination is given in table 1 for the four collections. All means of the main effects (collection date, storage treatment, and seed treatment, as well as interactions) were highly significant.

Collection date.--Average germination significantly increased through the third collection, with no increase noted in the fourth collection. The average germination of seed from cones 2 weeks, 3 weeks, 5 weeks, and 6 weeks immature (based on oil flotation) are given in table 2. Although stratification promoted germination in all cases, maximum germination (of stratified samples) increased with later collections.

Storage treatment.--All storage treatments (B, C, D, and E) significantly promoted germination over that of immediate testing (Treatment A). Germination of seed from cones stored at 72°F (35 percent relative humidity) for 30 days (Treatment B) was significantly better than all other treatments.

With cool, damp storage (Treatments C, D, and **E**), **30** days (Treatment D) provided greater germination than that for 15-day. Treatment F (cones stored on ends) was no better, or worse, than Treatments D and **E**.

Seed treatment.--Seed dormancy decreased as cones matured, although the seed remained quite dormant until 5 to 10 days before cone maturity. Stratification significantly promoted germination over that of unstratified seed.

INTERACTIONS

Collection date x storage.--The results indicate that dry storage was significantly better on the first collection than other treatments. As the cones aged, they required less air exchange and thereby could tolerate more coverage and damp storage.

Storage treatment x seed treatment.--Stratification increased the germination of all storage treatments. The effect of stratification varied with the storage treatment. It had the least effect on cones which had been stored dry (Treatment B) and the greatest effect on cones (seed) which had not been stored (Treatment A). The effect decreased with the length of cool, damp storage.

Table 1.--Average full seed germination by treatments

| Collection: | Seed treatment: | Cone treatment | | | | | |
|-------------|-----------------|----------------|------|------|------|------|------|
| | | A | B | C | D | E | F |
| | | Percent | | | | | |
| <u>1</u> /1 | Unstratified | 7.3 | 93.1 | 43.5 | 64.3 | 66.8 | 61.2 |
| | Stratified | 46.0 | 97.8 | 81.0 | 91.1 | 88.9 | 86.7 |
| 2 | Unstratified | 13.0 | 93.1 | 63.3 | 76.3 | 69.0 | 82.5 |
| | Stratified | 47.9 | 95.0 | 83.6 | 86.4 | 72.6 | 94.4 |
| 3 | Unstratified | 23.6 | 96.1 | 80.9 | 94.9 | 83.8 | 93.9 |
| | Stratified | 65.1 | 97.2 | 92.7 | 96.9 | 90.0 | 96.7 |
| 4 | Unstratified | 52.0 | 88.1 | 80.5 | 69.8 | 82.6 | 71.3 |
| | Stratified | 86.4 | 97.9 | 97.7 | 82.2 | 93.7 | 83.3 |
| <u>2</u> /5 | Unstratified | 77.7 | | | | | |
| | Stratified | 85.6 | | | | | |
| 6 | Unstratified | 94.7 | | | | | |
| | Stratified | 94.9 | | | | | |
| <u>3</u> /7 | Unstratified | 92.6 | | | | | |
| | Stratified | 94.6 | | | | | |

1/ Average of ten trees for collections 1 through 4.

2/ Average of four trees for collections 5 and 6.

3/ Average of two trees for collection 7.

Table 2.--Germination of Treatment A seed from individual trees with maturity (as determined by oil flotation) at various intervals after initial collection

| Tree number | Seed treatment | Sept. 7 | Sept. 12 | Sept. 18 | Sept. 22 | Oct. 2 | Oct. 11 | Oct. 17 | Remarks |
|-------------|----------------|---------------------|----------|----------|----------|--------|---------|---------|--------------------------------------|
| | | Percent germination | | | | | | | |
| 2 | Unstratified | 15 | 10 | 44 | 82 | -- | -- | -- | 2 weeks immature at first collection |
| | Stratified | 69 | 63 | 84 | 88 | | | | |
| 7 | Unstratified | 8 | 3 | 32 | 84 | 95 | -- | -- | 3 weeks immature at first collection |
| | Stratified | 47 | 18 | 36 | 90 | 94 | | | |
| 9 | Unstratified | 12 | 9 | 49 | 76 | -- | 95 | -- | 5 weeks immature at first collection |
| | Stratified | 72 | 77 | 93 | 98 | -- | 96 | | |
| 8 | Unstratified | 9 | 40 | 24 | 61 | 76 | 94 | 96 | 6 weeks immature at first collection |
| | Stratified | 65 | 74 | 80 | 94 | 94 | 96 | 98 | |

Collection date x seed treatment.--Unstratified germination increased to the third collection, which was the first collection to have an average specific gravity of less than 1.00. Following this collection, unstratified germination decreased.

The effect of stratification decreased as the cones matured. A greater promotion of germination was noted with the last two collections over that of the first two collections.

Storage treatment x seed treatment x collection date.--Germination percentage of all treatments became more uniform (less spread) with later collections, with stratification increasing the uniformity. In the earliest collection, dry storage significantly improved germination over that of other treatments although all storage treatments were better than no treatment. With later collections, other treatments proved to be equal to dry storage in the following progression:

1. Thirty-day cool storage without covering (F).
2. Thirty-day cool storage with covering (D).
3. Fifteen-day cool storage with covering (C).

This indicates that an air exchange is important before cones reach maturity, but becomes unimportant after natural maturity.

Additional germination.--Germination of both stratified and unstratified seed of Treatments A and C was slow and steady. After 28 days, there was sufficient germination to warrant leaving the tests at germinating conditions. Germination of the stratified and non-stratified seed was finally concluded after 37 days and 59 days, respectively.

Although the germination at 28 days was used for the analysis and discussion, table 3 shows that these treatments produced considerable germination although it was slow, The difference was less with later collections and with stratification. Only the last collection of Treatment C (after stratification) reached that of Treatment B.

In conclusion, the data gathered from this study showed that there was considerable variation between trees. Within the 10 trees sampled, cones varied from 2 to 6 weeks immature at the first collection. Even with this variation, the seed of cones from the first collection were artificially ripened. They increased from an average germination of 7.3 percent to 93.1 percent after a 30-day storage at 72°F and 35 percent relative humidity. Cool damp storage was satisfactory after the specific gravity reached 1.00. Prior to this, the data indicate that air exchange to the cone is essential.

Table 3.--Summary of germination after 28 days

| | | Average germination | | | |
|-------------|---|---------------------|---------|--------------|---------|
| | | Stratified | : | Unstratified | |
| Collection | : | 28 days | 37 days | 28 days | 59 days |
| | | Percent | | | |
| TREATMENT A | | | | | |
| 1 | | 46 | 66 | 7 | 57 |
| 2 | | 48 | 59 | 13 | 54 |
| 3 | | 65 | 75 | 24 | 64 |
| 4 | | 86 | 89 | 52 | 80 |
| TREATMENT C | | | | | |
| 1 | | 81 | 88 | 44 | 80 |
| 2 | | 84 | 89 | 63 | 80 |
| 3 | | 93 | 93 | 81 | 85 |
| 4 | | 98 | 98 | 80 | 87 |

Cones of early collection would not open when removed from storage and placed in the oven. However, cones which were left under trees at these early collections opened normally; therefore, it would seem that the standard procedures used in this study are the reason for lack of cone opening. Most likely, a fluctuating relative humidity would be required to open artificially ripened cones. The necessary fluctuation may even change according to the specific gravity of the cones.

No tree characteristics were found to give any indication of cone maturity. It could, however, be determined by cone specific gravity or seed moisture content. The former is a more practical method than the latter. The determination of specific gravity can be made in the field by an oil flotation test with results comparable to the laboratory. This study also indicates that cones will float in oil approximately 2 weeks after floating in water.

Cone characteristics offered no indication of maturity, although seed characteristics did offer some measure. The seed became mottled or darker in color at least 10 days before oil flotation. Considerable variation was noted, such as one tree having dark seed 20 days before cone maturity. Cones of this particular tree did open at these early collections; therefore seed color may serve as a guide to cone maturity.

The germination data indicated that seed dormancy in loblolly pine decreases with the approach of maturity. Germination of seed of all collections and treatments prior to cone maturity was promoted by stratification.

Germination of seed excised from cones (Treatment A) at the first collection was 7 percent in 28 days and 57 percent in 59 days. Even stratification did not promote germination to the level of later collections, which indicates that a period of artificial ripening is necessary. It also seems that a warm, dry period is best, which might indicate the need for a reduction in seed moisture.

Discussion

Q. Jeffers: Have you run a gas analysis on the bag materials used?

A. Bonner: No, we haven't run a gas analysis on these materials; however, all the packaging materials we used have been used, or are being used, in vegetable seed packaging tests at the Seed Storage laboratory at Fort Collins. So far they haven't had any trouble with gas in their tests. The possibility may exist but we haven't looked into it. If the bag is sealed, the possibility is very unlikely.

Q. Mickelson: With reference to the material, why were you concerned with storing for 18-24 months?

A. Bonner: The crop failure, of course, is my main concern. We had a pretty wide-spread crop failure in 1969 in the mid-South. Two years in a row (although this is not likely) may be possible. I don't worry about 10 to 12 years' storage because if we can successfully store for 3 to 5 years we have the main part licked.