U. S. FOREST SERVICE RESEARCH PAPER NE-36
1965
NORTHEASTERN FOREST EXPERIMENT STATION, UPPER DARBY, PA.
FOREST SERVICE, U. S. DEPARTMENT OF AGRICULTURE
RALPH W. MARQUIS, DIRECTOR

Germination of Eastern White Pine Seed

as influenced by stratification

by Raymond E. Graber

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STRATIFICATION has been a standard practice for many years in conditioning tree seed for testing or for sowing in forest nurseries. Literally, the term means to place the seed in layers, and originally the treatment involved placing layers of seed between layers of a moisture-retaining medium such as peat moss or sand. However, the layering is incidental; the essential conditions are moistness and low temperature, and the term stratification is now applied to any seed treatment that provides those conditions. It is so used in this paper.

Stratification breaks dormancy, insuring rapid and uniform germination as soon as conditions become favorable. In nature this process occurs as seeds over-winter on the forest floor. But

when dormant seeds, such as those of white pine *(Pinus strobus)*, are sown in the spring, stratification is necessary to prepare the seeds for germination. This treatment is particularly necessary for direct seeding, where fast and complete germination may be the deciding factor between adequate stocking or failure.

Direct seeding can be done either in the fall, with untreated seed, or in the spring, with stratified seed. Spring seeding has an advantage: the seed is exposed to predators for the minimum time.

Even though the practice of stratifying white pine seed is widely used and recommended,[1] no detailed statistical evaluation of the effects of stratification on this species has been published. The purpose of this study was to measure the influence of stratification temperature and length of stratification period on the completeness and speed of germination in white pine seed.

MATERIALS AND METHODS

Three white pine seed collections were made in 1961 within a 30-mile radius in southwestern Maine. The cones were air-dried and seed was extracted in a small tumbler. The seeds were dewinged by hand and winnowed to remove inert matter and empty seeds. They were then dried to a moisture content of 5 percent and stored at 36°F. for 14 months.

Seeds, in lots of 50, were placed in petri dishes on water-saturated kimpack (a highly absorbent sterile germination paper) and stratified in refrigerators at three constant temperatures: 36 $\pm$ 1°F., 40 $\pm$ 1°F., and 50 $\pm$ 2°F. Each refrigerator was illuminated continuously with a 15-watt incandescent light.

After stratification the seeds were rinsed in tap water and replaced in the petri dishes on clean saturated kimpack. The seeds

[1]Barton, L. V.: Hastening the Germination of Some Coniferous Seeds; Amer. Jour. Bot. 17: 83-105, 1930.—Baldwin, H. I.: Effect of After-Ripening Treatment or Germination of White Pine Seeds of Different Ages; Bot. Gaz. 96: 372-376, 1934.—United States Forest Service: Woody-Plant Seed Manual; U. S. Dept. Agr. Misc. Pub. 654, 416 pp., illus., 1948.

were incubated 40 days at 68-86°F. (alternating 86°F. for 8 hours with natural light and 68°F. for 16 hours in darkness). Germination counts were made daily for the first 20 days and at 2-day intervals thereafter. A seed was counted when the emerging radical was 2 mm. long or longer.

After 40 days of incubation the ungerminated seeds were cut longitudinally and inspected. All well-filled seeds with fully developed embryos were considered viable. Germination percentage was based on the total number of viable seeds. The number of days required to attain 33 1/3 percent germination of the viable seeds was used as a measure of the rate or speed of germination. This benchmark, 33 1/3 percent was selected because in nearly all cases germination reached or exceeded that amount in the 40-day period, thus allowing statistical comparisons of the treatments to be made.

The study was divided into two parts. In Experiment I, the treatment combinations of stratification period and temperature included the range of those commonly recommended for eastern white pine seed. Seed from the three collection sources was stratified at 36°, 40°, and 50°F. for 10, 30, 60, and 90 days. These treatments were replicated three times with 50 seeds per replicate. A control (no stratification) was added for each seed source; however, the control data were not included in the analysis. Analysis of variance was used to evaluate the effects of the treatments and orthogonal comparisons were made to locate significant differences.

Experiment II was carried out to further elucidate the effects of stratification period. It was restricted to one seedlot—8-61-L—and to one temperature—40°. The additional stratification periods tested were 1, 5, 20, and 45 days. These treatments were replicated four times. Another 50-seed replicate of seedlot 8-61-L was added to each of the Experiment I periods of 10, 30, 60, and 90 days at 40°, thus providing four replicates of all eight stratification periods for this seedlot and temperature. Since all seeds in both experiments were treated and germinated concurrently, statistical comparison (analysis of variance) of the eight periods was valid.

RESULTS

Experiment I

The effects of seed source and period of stratification on both percentage of germination and rate of germination were significant. Stratification temperature significantly affected only the rate of germination (table 1). All statements of statistical significance in this paper refer to the 1-percent level of probability.

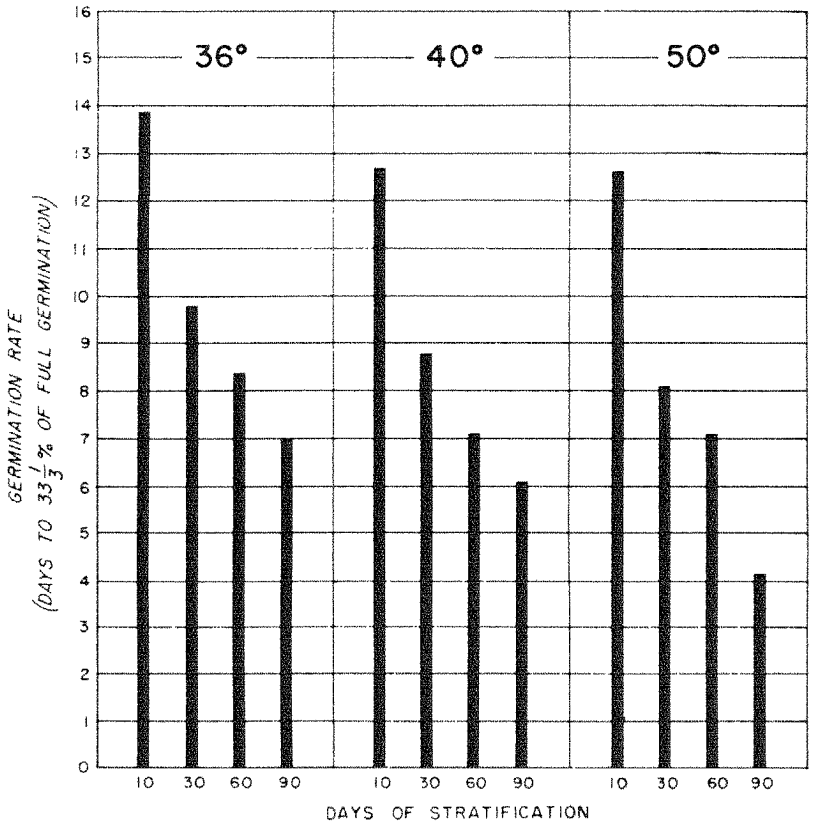
Seed source.—Although germination for seed source 6-61-A was significantly higher, the differences among the three sources were so small that they have little practical importance (table 1). The seed sources also differed in rate of germination: source 4-61-WC was significantly slower than source 6-61-A.

Stratification temperature. — Increasing stratification temperature above 36°F. resulted in a slightly greater (but not significant) percentage of germination (table 1).

Table 1.—Average percent germination and rate of germination as affected by seed source, stratification temperature, and stratification period

Item	Germination (percent)	Rate of germination (days to 33-1/3 percent of full germination)
Seed source:		
4-61-WC	89.9	9.33
6-61-A	94.8	8.25
8-61-I.	91.8	8.72
Stratification temperature:		
36°F.	90.4	9.7
40°F.	93.1	8.2
50°F.	92.9	8.0
Stratification period (days):		
Control	43.2	—
10	70.0	13.0
30	98.9	8.7
60	99.7	7.6
90	99.9	5.7

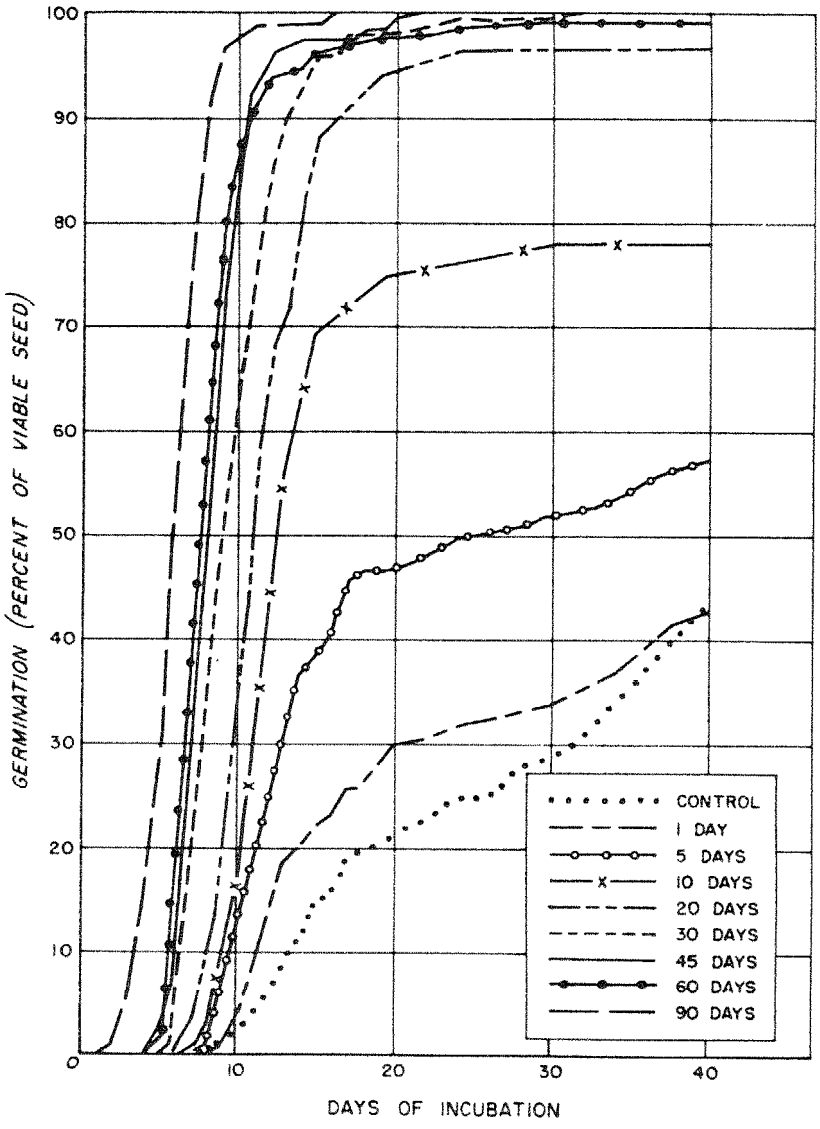
Figure 1.—Rate of germination as influenced by stratification temperature and period.



Speed of germination increased significantly with each increase in stratification temperature (fig. 1). Premature germination occurred at 50° when the stratification period was 60 days or longer. After 60 days' treatment, 1 percent of the seeds had germinated, and after 90 days an average of 32.4 per cent had germinated.

Stratification period.—Length of stratification period was the major factor affecting both amount and speed of germination. The 10-day period was insufficient to fully overcome seed dormancy, yet an average of 70 percent of the stratified seed germinated, compared to 43.2 percent of those in the untreated controls (table 1). Germination after 30 days of stratification was

Figure 2.—Patterns of germination during 40 days' incubation for white pine seed stratified for various periods at 40°F.



significantly lower than after the 60- and 90-day treatments, but the actual differences were only 1 percentage point or less, and thus of no practical importance. Under the test conditions, almost

complete germination occurred after 30 or more days of treatment regardless of stratification temperature or seed source.

The effects of stratification period were more pronounced on the rate of germination than on the ultimate amount (fig. 1). Each increase in length of period significantly increased the rate of germination.

Experiment II

The additional stratification periods included in Experiment II resulted in highly significant differential responses in germination. Germination patterns for the eight stratification periods and the control are shown in figure 2. Nearly complete germination (96.5 per cent) occurred after only 20 days of stratification. The 5-day treatment, with 57 percent germination, was roughly intermediate between the control (43 percent) and the 10-day treatment (78 percent). One day of treatment had no effect: germination was identical with that for the control. An almost linear relationship was evident between length of stratification period and total germination for 1, 5, 10, and 20 days of treatment (fig. 3).

As expected, rate of germination increased with length of stratification period (table 2). The control and the 1-day treatment are omitted from this table because some replicates did not attain 33 1/3 percent germination during the 40-day incubation period.

Table 2.—Rate of germination of white pine seedlot 8-61-L for different periods of stratification at 40°F.

Period of stratification (days)	Rate of germination (days to 33 1/3 percent of full germination)
5	14.50
10	11.50
20	10.50
30	8.75
45	7.75
60	7.25
90	5.75

DISCUSSION and RECOMMENDATIONS

Length of the stratification period was the most important variable influencing speed and amount of germination. Although the effects of seed source and stratification temperature were statistically significant, the differences were relatively small and of little practical importance.

The test results indicate that nearly complete germination may be expected after 20 to 30 days' stratification. The main advantage of longer treatment is faster germination. In practice, particularly in field sowing, speed of germination can be as important as percentage of germination, perhaps even more important. Growing conditions (heat and moisture) usually are near optimum for germination and seedling establishment only for a short time in the spring. As the season advances, high temperatures and rapid drying of the surface layers of the soil may cause severe losses of newly germinated seedlings.

When seed is sown in the fall, exposure for 6 or 7 months to natural conditions of moisture and low temperature prepares

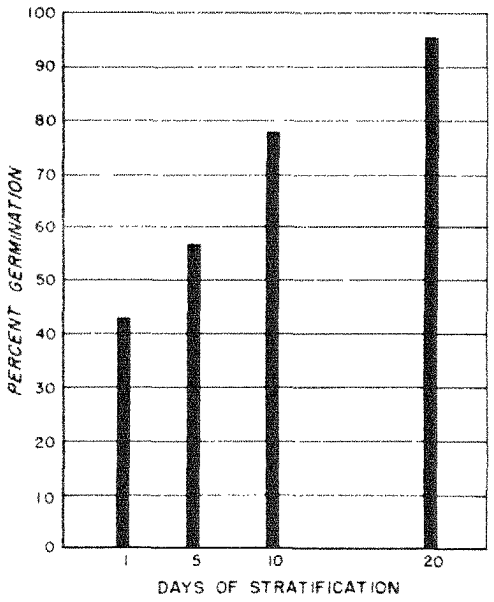


Figure 3.—Percent germination of white pine seed after 1 to 20 days' stratification at 40°F., showing the almost linear effect of length of stratification period.

the seed for rapid germination at the earliest favorable time. But when sown in the spring the seed must be prepared artificially for quick germination when conditions become suitable. To accomplish this objective a relatively long period of stratification is most effective. It is quite possible that a period in excess of 90 days may be optimum, at least for certain seed sources.

Also, it must be remembered that seed of the same species from different parts of its geographic range may have very different stratification requirements. For example, Mergen[2] found that white pine seed collected in North Carolina required a long period of stratification (in excess of 84 days), while seed collected in New Brunswick germinated almost completely without any stratification.

The data presented here are probably valid for seed from the white pine region of New England. Based on this assumption, 30 days' stratification will result in virtually complete germination of seed from this region. Where maximum speed of germination is desired, longer periods—in excess of 60 days—are recommended.

The stratification temperature had a very minor influence on rate of germination. If 30-day or shorter periods of stratification are planned, a temperature near 50°F. may give a slight advantage. For longer periods of stratification, however, lower temperatures are recommended to avoid premature germination. In our tests, seed germinated during both the 60- and 90-day treatments at 50°. Forty degrees appeared to be near optimum; seed treated at this temperature germinated more rapidly than seed treated at 36°, and no germination occurred during stratification.

■

[2]Mergen, F. ECOTYPIC VARIATION IN PINUS STROBUS L. Ecology 44: 716-727, 1963.