# STRATIFICATION OF SEED OF PINUS STROBIFORMIS

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### Introduction

It was not necessary to pretreat seed of <u>Pinus</u> <u>strobiformis</u> Engelm. from northern Arizona and northern New Mexico in order to increase germination values. In general, sulfuric acid treatments were detrimental; they were especially harmful when combined with stratification in moist sand. Stratification alone for 45 days produced high germination values, but these were not significantly different from those obtained from untreated seed.

The numerous techniques used to hasten and produce uniform. seed germination have been adequately described and summarized (3,4). The present test is related to the part of the "Woody-Plant Seed Manual" (6, p. 272), which reads "Germination [of seeds of <u>Pinus flexilis</u> var. <u>reflexa</u>] only fair without pretreatment." However, a footnote on the same page reads "Treatment only suggested for this species; experimental data not complete." In the present note, evidence shows that scarification, stratification, and other techniques were not required for rapid and uniform germination.

As part of a nursery test within a rangewide progeny study of the <u>Pinus flexilis</u> complex, several seed stratification techniques were used to determine the efficacy of pregermination treatments to induce more rapid and uniform germination rates. The series of tests described under the methods section was applied to the seeds of two stand collections of the southern member of the complex, <u>Pinus</u> <u>strobiformis</u>, which is nomenclaturally synonymous with P. <u>flexilis</u> var. <u>reflexa</u> Engelm.

### Materials and Methods

The two seed lots used for this test grew near Flagstaff, Ariz., and Osha Canyon, N. Mex. When both lots arrived, the seeds were extracted from their cones, separated into sound and malformed seeds by momentary floatation in 95-percent ethyl alcohol, and stored dry in individual scaled jars at  $4^{\circ}$  C. for 75 days.

Eight treatments were applied to both seed lots:

- A. Control (dry storage at 4 ° C.)
- B. 5-minute soak in reagent-grade sul furic acid
- C. 30-minute soak in reagent-grade sul furic acid
- D. Cold soak in distilled water at 3<sup>o</sup> C. for 7 days
- E. Cold soak in distilled ;eater at 3° C. for 14 days
- F. Stratification in moist sand for 45 days
- G. Treatment F superimposed upon treatment B
- H. Treatment F superimposed upon treatment C

Four replicates, with 25 seeds per replicate, were used in each of the 16 treatments.

After treatment, the seeds were placed in random order by replicate upon moist blotting paper disks and covered by the upper shell of a petri dish. The disks rested on several layers of moist Kempac paper laid in large germination trays. Conditions within the 2- by 1.3- by 0.6meter modified germination chamber for the 35 days of the test were cycled at 8 hours at 30° C. with fluorescent illumination, and 16 hours at 20° C. with no illumination. The germination paper was kept moist throughout the test; in addition, the travs were shifted daily to eliminate some of the variability due to position. At the end of the test, all ungerminated seeds were stained with tetrazolium chloride to determine evidence of respiration.

### Results

A series of germination curves was plotted for both seed lots subjected to the eight treatments (fig. 1). The values are expressed as' total percentage of germination on 11 dates. No treatment at all (A) or stratification in moist



Figure 1.--Percentage of germination on 11 dates for <u>Pinus</u> strobiformis from Flagstaff, Ariz., and Osha Canyon, N. Mex.

sand (F) was most conducive to germination. The 5-minute soak in sulfuric acid (B) was not aslethal to the seeds as the 30-minute soak (C). Cold soaking in distilled water (D and E) had the same effect as the 5-minute acid soak, but germination values of the former were somewhat lower. The treatments consisting of a combination of scarification with sulfuric acid followed by stratification in moist sand (treatments G and H) killed most of the seed and delayed germination. Although both seed lots displayed si milar germination patterns in the treatments, there was less total germination and a slight delay in germination in the New Mexico lot. Tests with the tetrazolium indicator revealed a lack of respiration for all ungerminated seed. Virtually all the ungerminated seeds, especially those subjected to treatments C, G, and H, contained blackened, pasty endosperm and gray embryos, indicating chemical injury by the treatment series; untreated embryos were ivory white.

#### Discussion

It was of primary importance that no pretreatment was required to germinate seed successfully from the northern Arizonan and northern New Mexico origins. The treatment of the seed in moist sand for 45 days, which is a modified form of the standard recommendation for most conifer seed (4), accelerated germination slightly but not significantly. The 5minute soak in sulfuric acid (treatment B) apparently scarified the testae of the seeds enough to allow rapid imbibition of water, and thus produced a significantly higher germination value than cold soaking alone (treatments E and D). For most of the seeds, treatments C, G, and H destroyed the embryo-endosperm system, thus denying normal germination, and induced decay of tissue.

Scarification of hard-tested tree seed in sulfuric acid for 1 to 6 hours was suggested Q5), but this treatment was lethal because of the testa permeability of Pinus strobiformis seed. However, in a series of exploratory, tests, R. J. Boyd subjected Pinus monticola seeds to a 45-minute soak in concentrated sulfuric acid, followed by a 15-minute tapwater wash with immediate immersion in hydrogen peroxide, and recorded 53 percent germination at 10 days and 70 percent at 45 days.1 Swofford wrote 2 "We have tried acid scarification with some species and have found that the moisture content of the seed should be less than 10 percent before scarification. Apparently, the higher moisture content of the seed makes the action of the sulfuric acid more violent with the possibility of the destruction of the seed." The

<sup>1</sup>Boyd, R. J. Official correspondence from U.S. Forest Service, Intermountain Forest and Range Experiment Station, Ogden, Utah. 1963.

<sup>2</sup> Swofford. T. F. Official correspondence from U.S. Forest Service, Region 8, Eastern Tree Seed Testing Laboratory, Macon, Ga. 1959. moisture content of the <u>P. strobiformis</u> seeds of the present test was about 10 percent. In relation to the water-soaking treatments (D and E), with no aeration, which gave fair germination values in the present test, Swofford also commented, "Our experience with conifers has been that soaking for that long a period [7 and 14 days] is injurious to the seed. You might possible soak the seed without injury by aerating the water." Both of Mr. Swofford's comments are well taken and suggest further approaches to the numerous seed-germination problems still to be solved with the respective species of the genus <u>Pinus.</u>

Under nursery conditions, samples of 17 origins of <u>Pinus strobiformis</u> from Arizona, New Mexico, and Texas germinated rapidly and uniformly with no pretreatment, but samples of 44 origins of <u>P. flexilis</u> James seeds that also were untreated germinated spasmodically much later. In fact, some of the northern sources from Montana germinated 1 full year after sowing. In this test, as well as one now underway with <u>P. chiapensis</u> (Martinez) Andresen (1 2), <u>P. monticola</u> Dougl., and P. <u>strobus</u> L., there are highly related correlations between (1) latitude and germination date--origins from lower latitudes germinate earlier; and (2) date of collection and germination--early collecting dates result in later germination.

## Literature Cited

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