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EFFECTS OF STORAGE TREATMENTS ON THE RIPENING AND VIABILITY OF VIRGINIA PINE SEED

In a study at the Beltsville, Md., Experimental Forest in 1953 and 1954, the seed of Virginia pine (*Pinus virginiana* Mill.) became at least 45 percent viable 8 weeks before the natural opening of cones. Because seedfall in that locality usually starts during the first or second week of November, it was concluded that cone collecting could begin there safely in early September (*1*).

However, the patterns of viability before seedfall differed in the 2 years. In 1953 viability tended to increase between early September and the beginning of seedfall in November, whereas in 1954 viability peaked at 86 percent on September 7 and then declined to 37 percent on October 25.

The 1954 pattern of viability was not only at odds with the 1953 results, but also with data obtained in studies on other species of pine (*2, 3, 4*). Hence it seemed advisable to take another look at the development of seed viability in Virginia pine. This was done with seed of the 1960 crop, again on the Beltsville Forest.

The 1960 study included not only the pattern of viability before seedfall, but also 6 treatments of seed collected at weekly intervals during the ripening period. These treatments involved varying lengths of dry storage at 25° to 28° C. and 5° C., and varying periods of wet storage at 5° C.

Methods

Seed collection.—Seed for the 1960 study was collected from 10 Virginia pines at 7-day intervals between July 26 and November 15, when seedfall began. The trees, growing in an old-field stand, were large-

crowned, about 16 years old, and fruiting abundantly. Five cones were cut from each tree on each collection date. The cones were gathered from different parts of each crown to avoid any possible bias due to effects of cone position.

The 50 cones in each collection were mixed to insure randomness among later subsamples. Average specific gravity was determined within an hour after the cones were collected.

Seed extraction.—The seed in 25 randomly selected cones from each 50-cone sample was hand-extracted within 24 hours after collection for use in 5 of the 6 storage treatments. Extraction was done by pulling back each cone scale and carefully easing out the seeds. The cones were not predried nor preheated.

In the remaining treatment, which called for storage of cones under room conditions, relatively little hand extraction was necessary.

Treatments.—The 6 storage treatments of cones and seeds are tabulated below, in number of days:

	Dry storage of cones	*Storage of seeds*	
Treatment	*at 25°-28° C.*	*Dry at 5° C.*	*Wet at 5° C.*
I	0	0	0
II	0	0	2
III	0	0	5
IV	0	0	14
V	21-154	56	2
VI	0	94-207	2

Dry storage of seeds was in tightly stoppered vials.

Germination tests. — In all treatments, after seeds were extracted, mixed, and subjected to any scheduled dry storage, 100 apparently sound seeds were placed on damp, shredded peat moss in each of 2 or 3 plastic germinating dishes. Dishes of seeds exposed to wet storage were sealed and stored at 5° C. for the proper number of days. After the completion of any scheduled wet storage, seeds of all treatments were exposed for 2 hours to 160 foot-candles of cool-white fluorescent light. Then the dishes were sealed and placed in a dark oven held at 25° ± 1° C., which Toole *et al.* (*6*) had suggested as the temperature most conducive to the germination of Virginia pine seed.

Germination was tallied in each dish at 7, 14, 25, and (usually) 46 days after the seeds were placed in the oven. After the last examination, all ungerminated seeds were cut to determine soundness so that viability could be expressed as percent of sound seed. A seed was counted as germinated if its radicle extended 5 mm. or more, and an ungerminated seed was regarded as sound if endosperm tissue was visible (even though

Table 1. — *Percent germination of sound Virginia pine seeds, by collection dates and storage treatments*[1]

Collection date	Specific gravity of cones	Storage treatment I	II	III	IV	V	VI
July 26	1.06	0	0	0	0	—	0
Aug. 2	1.06	0	0	0	0	—	0
9	1.05	0	0	0	0	—	0
16	1.02	0	0	0	0	4	0
23	1.07	0	0	0	0	35	25
30	1.07	7	2	3	4	73	76
Sept. 6	1.07	5	7	8	7	81	82
13	1.03	29	33	22	39	76	61
20	1.04	65	65	57	75	83	72
27	.99	87	79	86	84	69	70
Oct. 3	.99	83	86	81	65	79	87
11	.99	82	88	66	72	53	84
18	.94	67	79	71	84	60	91
25	.91	53	62	67	78	73	72
Nov. 1	.89	61	79	72	83	75	73
8	.78	86	85	81	78	81	85
15	.60	78	84	82	91	82	89
Average (all values)		41	44	41	45	54[2]	57
Average (September 13 to November 15)		69	74	68	75	73	78

[1]Each value was based on two 100-seed samples, except in treatment V, in which three 100-seed samples were tested.

[2]Missing values for the first three collection dates, for which germination was not tested, were assumed to be zeros.

rotted). In treatments V and VI, observations on germination were terminated after 25 days because rot was becoming prevalent in the ungerminated seeds.

Results

Viability prior to seedfall. — The 1960 results confirmed earlier findings: (1) that Virginia pine seed becomes sufficiently viable 2 months before seedfall to warrant collecting for routine use; and (2) that some seeds mature even earlier (table 1).

The 1960 results also were in accord with the 1953 observations of Church and Sucoff on Virginia pine (*1*) and with reported observations on other pines (*2, 3, 4*) that, once high viability is reached, it tends to remain high until seedfall. After September 20, 1960, there were no consistent variations in the percent of seed that germinated (table 1). The decline in germination observed by Church and Sucoff after September 7 in 1954 seems illogical and is unexplainable.

Table 2. — *Percent of total germination occurring within 7 days, by collection dates and storage treatments*

Collection date		Storage treatment					
		I	II	III	IV	V	VI
Aug.	16	—	—	—	—	11	—
	23	—	—	—	—	41	33
	30	14	40	20	0	71	50
Sept.	6	0	0	0	0	89	59
	13	36	21	17	11	58	57
	20	12	17	23	28	76	79
	27	41	68	79	69	86	69
Oct.	4	70	80	87	81	74	76
	11	85	98	100	96	73	88
	18	80	99	99	99	57	87
	25	73	98	99	98	49	65
Nov.	1	90	97	98	97	79	56
	8	83	97	99	98	84	69
	15	75	90	73	98	77	69

Treatment effects on seed germination.—The amount of germination varied with pregermination treatment, but not always as expected. Toole *et al.* (*6*) had reported that imbibition of moisture at 5° C.—that is, cold wet storage—increased the germination of Virginia pine seed. In our study, cold wet storage (treatment II-IV) increased germination only slightly, if at all, over germination of seeds that did not receive such a treatment (treatment I) (table 1). However, the values in table 1 are for total germination; germination during the first 7 days was, in fact, somewhat increased by a few days of cold wet pregermination treatment.

This quicker germination induced by cold wet storage was especially evident in the samples collected after October 1. For example, the proportion of total germination that occurred within 7 days often approached 100 percent in these later collections in treatments II-IV but fell between 70 and 90 percent in treatment I, which differed only in the omission of cold wet storage (table 2).

Prolonged dry storage had more effect than wet storage on total germination, especially among seeds collected before September 20. For example, in the September 6 collection, treatments I-IV, which provided no dry storage, showed less than 10-percent germination, as compared to 81 or 82 percent under treatments V and VI, which included dry storage (table 1). Prolonged dry storage advanced the collection date at which high seed germination occurred by about 3 weeks.

Prolonged dry storage seemed first to favor and then to hinder rapid germination. For seeds collected prior to October 1, quicker germination usually occurred among those that had undergone such storage (treatments V-VI compared to treatments I-IV), but for seeds collected after October 1 a reverse relationship was apparent (table 2).

Cone maturity and germination. — The cones became physiologically mature during the latter part of September. Maturity was indicated by a change in cone color from green to dark purple during the third week of the month, and by a drop in specific gravity below 1.0 in the cones collected on September 27. Before that date specific gravities had ranged from 1.02 to 1.07—with the high of 1.07 appearing in late August and persisting through early September.

Once the specific gravity had dropped below 1.0, there was no consistent effect of date of collection on the germination of sound seeds (table 1).

Seedling abnormalities.—Schubert (5) has described seedling abnormalities associated with prematurely collected, though viable, seeds of western species of pine, so an attempt was made to determine whether similar abnormalities would develop from prematurely collected seeds of Virginia pine. Extra seeds from treatment VI were used, and 18 seedlings from each of the 13 collections of viable seeds were grown in clay pots. The seedlings were observed for 37 days after germination started. No abnormalities associated with early cone collection appeared.

Discussion

The 1960 study confirmed 1953 and 1954 findings that, at least 2 months before natural seedfall, Virginia pine seeds ripen on the trees sufficiently to be capable of germination.

The 1960 data also indicate that seeds that have been collected early continue to ripen off the trees under certain storage conditions. Prolonged cold dry storage in closed containers, with or without prior storage in cones at room temperatures, apparently favors the ripening and subsequent germination of seed collected between August 30 and September 20. Such storage is easily provided and could be a routine practice with seed collected for use in forest nurseries.

If cold dry storage is not available, cone collections should be delayed until cones are mature. This time can be determined by a change in cone color from green to purple, and by a drop in specific gravity of the cones to 1.00 or less.

No adequate guide other than calendar date is available for determining when cone collections may be started for seeds that are to be afterripened by cold dry storage. Because this date may be affected by location and climate, the results of this study should not be applied outside southern Maryland without allowing for some local verification.

None of the storage treatments tried in the 1960 study gave the absolute maximum in germination of sound seeds. Cutting tests indicated that 89 to 100 percent of all lots were sound. Germination, even when based on *sound* seeds (table 1), usually reached maxima of only 86 to 91 percent. Possibly better control of seed-rotting organisms would have increased these values.

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