FACTORS AFFECTING THE VIABILITY AND GERMINATION BEHAVIOR OF CONIFEROUS SEED. VI. STRATIFICATION AND SUBSEQUENT TREATMENT, PSEUDOTSUGA MENZIESII (MIRB.) FRANCO¹

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ABSTRACT

Stratification effects increase with duration of treatment; short treatments accelerate germination markedly for incubation at 25°C but longer treatments are necessary for rapid germination at 15° or 10°C. Post stratification drying and storage reduce the stratification effect, particularly at low incubation temperatures, but do not adversely affect germinative capacity. Implications for seed testing and nursery practice are discussed.

INTRODUCTION

Stratification has long been used to hasten the germination of seed and has become common practice in tree nurseries of the Pacific Coast. It is an empirical technique based largely upon experience and varying in detail from nursery to nursery. Some use media to hold and supply moisture to the seed; others presoak the seed and then store it without medium at a temperature slightly above freezing.

Stratification not only accelerates germination at the commonly used incubation temperatures but it also makes germination possible at temperatures of 10°C or lower (Allen, 1960). Although growth at low temperature is confined largely, if not wholly, to the radicle, it does enable the root to penetrate the soil and become well established before any obvious top growth begins. This kind of behavior has been observed in March when the upper one-half inch of soil reached approximately 15°C at mid-day. It may explain the superior development of fall-sown seedlings in comparison with late-spring sowings of either untreated or stratified seed. Early sowing of well stratified seed would probably simulate results from fall-sowing.

Stratification has disadvantages: the work involved in mixing seed and moist medium, the cost of cold storage for several weeks, and the difficulty of separating treated seed from medium or of handling moist seed. Most of these can be minimized by using the "naked stratification" technique described by Allen and Bientjes (1954) and by other means. Some seed users are concerned about possible effects of longer treatments, loss of moisture during treatment, and the waste that might occur should some of the treated seed not be used.

This report covers a miscellany of experiences with stratified seed of Douglas fir, taken from a number of studies made during the period 1959-61. Although the studies were not designed specifically for the purpose they do shed some light on the effects of stratification and subsequent storage on seed from a variety of origins. For this reason they have been brought together. Although generalizations are attempted, it is realized that most of the seed

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lots were of exceptionally high quality, having been processed by hand methods, and that the conclusions may not be valid for seed of lower quality.

METHODS AND RESULTS

General

Stratification was accomplished by presoaking for 24 hours in tap water at room temperature, superficial drying, and storing in plastic vials inside loose-lidded sealers at 0-2°C.

All test seeds were incubated on the surface of vermiculite at water-holding capacity in covered Petri dishes. Moisture content was checked frequently and brought to standard as necessary. Germinates were counted as soon as their radicles reached or passed 0.1 inch in length and then removed; abnormal germinates were recorded but not included in the data summaries given here. As usual for Douglas fir, abnormalities were mostly of the reversed embryo type in which the radicle is oriented away from instead of towards the micropyle. Two samples of 25 seeds each were used in all tests.

Test duration for stratified seed was usually 30 days although, with an incubation temperature of 15°C. or 10°C., this was often extended to 40 days. Unstratified seed was incubated for about 50 days when germination had usually ceased. Seeds still ungerminated at the end of the period were cut and classified as **sound**, **deteriorated**, **insect-filled**, or **empty**. **Germination per cent (G)** at the end of the test, and **germinative capacity (GC)**, — germination per cent plus sound seed remaining) are shown for most tests along with a measure of the **rate of germination, R**, the median value for germination time in days based upon germinative capacity.

Since several different studies are covered by this report, each will be described separately as an **experiment**, even though it was part of a larger study.

Experiment 1

Samples from 26 seed lots from a variety of locations in British Columbia, collected in 1959, were stratified for 0-120 days and tested at 25°C., 15°C., and 10°C. All but six seed lots came from individual mother trees; those six lots were area collections included to show the range in behavior among areas for comparison with the range among trees. Six lots are of Interior origin (F15, F16, F17, F20, F21, and F23) and the remainder came from the Coast. Values of **G**, **GC**, and **R** are given in Tables 1, 2, and 3 for the three test temperatures.

The main features of the results are these:

1. In view of the limited sampling, germinative capacity at 25°C was remarkably uniform among unstratified seed and seed stratified for 10, 20, or 40 days. Only one lot, F13, showed a serious drop with increase in stratification period. Extended stratification (80 or 120 days) resulted in a definite decrease in the case of lots F13 and F15, and a possible decrease for lots F7, F12, and F35.
2. The median germination time at 25°C decreased from a range of 5.7—24.0 days for unstratified seed to one of 2.7—6.7 days for seed stratified for 10 days. A relatively quick and reliable test is possible following 10 days' stratification by the technique used; little is gained, from the standpoint of testing, by using a longer period of stratification. This conflicts on the

TABLE 1
G, GC, AND R FOR 26 LOTS OF DOUGLAS FIR SEED FOLLOWING
STRATIFICATION FOR 0-120 DAYS. EXPERIMENT 1. INCUBATION AT 25°C.

Lot No.	Source ¹	Unstrat.			Strat. 10 d.			Strat. 20 d.			Strat. 40 d.			Strat. 80 d.			Strat. 120 d.		
		G	GC	R	G	GC	R	G	GC	R	G	GC	R	G	GC	R	G	GC	R
F1	UBCF	90	90	6.6	86	86	4.3	100	100	4.6	90	90	3.9	94	94	3.5	82	82	2.9
F2	UBCF	96	96	15.0	92	92	3.9	96	96	3.8	98	98	3.8	96	96	3.5	92	92	2.9
*F4	UBCF	82	82	8.7	58	62	5.7	78	78	5.6	72	72	5.5	70	70	5.0	70	70	6.2
*F7	UBCF	92	94	6.4	90	90	4.9	90	90	4.5	92	92	4.1	86	86	4.2	82	82	4.7
*F8	UBCF	76	76	8.7	92	92	4.3	72	72	3.5	84	84	3.6	84	84	4.0	92	92	3.6
*F9	UBCF	72	76	18.3	68	68	5.2	68	68	4.4	76	80	5.0	68	76	6.6	72	72	5.0
*F10	UBCF	92	92	15.5	92	92	3.8	96	96	3.1	100	100	3.6	84	84	3.6	84	84	2.7
*F11	Campbell R.	92	92	14.5	100	100	3.9	96	96	3.6	96	96	2.9	100	100	2.8	98	98	2.6
*F12	Campbell R.	80	80	10.7	78	80	4.9	69	68	5.7	72	76	5.0	72	72	5.6	64	66	4.1
*F13	Campbell R.	42	70	38.0	50	74	14.5	42	48	13.0	34	36	7.5	30	36	9.0	36	36	8.0
*F14	Campbell R.	100	100	8.3	96	98	5.1	94	96	5.0	96	96	5.4	90	90	6.1	94	94	4.0
F15	Solsqua	88	90	18.7	92	94	4.0	92	92	3.0	96	96	3.1	60	60	2.6	14	14	1.0
F16	Alta Lk.	90	90	8.1	80	80	4.3	94	94	3.5	92	92	3.7	90	90	3.1	90	90	2.7
*F17	Falkland	98	98	9.9	98	98	2.7	98	98	1.7	98	98	1.6	98	98	1.5	98	98	0.8
*F20	Falkland	94	94	8.6	94	94	4.7	92	92	3.8	86	86	2.9	100	100	2.2	100	100	1.5
*F21	Falkland	96	98	13.2	98	98	3.8	100	100	2.8	100	100	2.7	100	100	2.6	92	92	2.2
*F23	Falkland	92	92	23.0	88	88	4.8	96	96	4.3	96	96	4.1	96	96	2.9	96	96	2.3
*F25	UBCC	86	86	5.9	90	90	2.8	90	90	2.9	82	82	2.8	96	96	2.7	92	92	2.4
*F27	UBCC	74	78	17.8	84	84	6.7	78	78	4.0	86	86	3.7	84	84	4.3	80	80	2.9
*F30	UBCC	90	90	19.5	86	90	4.3	96	96	3.7	92	92	3.0	94	94	3.3	92	92	3.6
*F32	UBCC	96	96	9.0	100	100	2.8	98	98	2.9	100	100	3.2	96	96	2.6	100	100	2.5
*F33	UBCC	86	86	24.0	98	98	4.5	94	94	4.2	96	96	3.4	98	98	2.6	98	98	2.5
*F34	UBCC	80	80	22.7	96	96	4.3	86	86	4.4	98	98	4.2	98	98	3.4	92	92	2.5
*F35	UBCC	86	90	15.3	88	88	5.3	88	88	4.8	88	88	4.8	74	80	5.8	80	80	4.4
F89	Mt.																		
	Benson, V.I.	88	89	5.7	—	—	—	98	98	4.4	94	94	3.8	94	94	3.4	100	100	2.3
F90	Cedar, V.I.	89	90	14.2	—	—	—	96	96	6.3	92	92	5.4	98	98	3.5	90	90	2.4

¹UBCF—University Research Forest, Haney, B.C. UBCC—University of B.C. campus area, Point Grey, B.C. * Single-tree collection.

TABLE 2
G, GC, AND R FOR 26 LOTS OF DOUGLAS FIR SEED FOLLOWING
STRATIFICATION FOR 0-120 DAYS. EXPERIMENT 1. INCUBATION AT 15°C.

Lot No.	Source ¹	Strat. 20 d.			Strat. 40 d.			Strat. 80 d.			Strat. 120 d.		
		G	GC	R	G	GC	R	G	GC	R	G	GC	R
F1	UBCF	16	54	30 +	56	72	15.7	84	84	7.7	72	72	7.3
F2	UBCF	16	58	30 +	76	86	11.8	82	86	8.8	98	100	7.4
F4	UBCF	2	42	30 +	8	64	30 +	26	66	30 +	62	74	12.3
F7	UBCF	10	46	30 +	26	86	30 +	60	84	11.6	58	86	11.7
F8	UBCF	48	68	9.7	80	80	8.0	72	76	8.9	68	68	8.6
F9	UBCF	12	80	30 +	12	88	30 +	44	80	18.0	72	76	10.2
F10	UBCF	32	92	30 +	88	96	8.6	96	96	8.5	100	100	6.6
F11	Campbell R.	22	80	30 +	80	90	9.1	96	100	7.6	96	96	6.7
F12	Campbell R.	8	82	30 +	42	74	12.7	40	68	14.0	60	72	10.4
F13	Campbell R.	4	66	30 +	2	68	30 +	4	48	30 +	8	38	30 +
F14	Campbell R.	0	94	30 +	10	94	30 +	6	86	30 +	70	96	12.0
F15	Solsqua	60	82	8.0	96	98	6.0	18	70	30 +	8	78	30 +
F16	Alta Lk.	32	58	13.8	74	84	11.2	94	94	8.5	88	88	6.4
F17	Falkland	86	96	5.6	96	96	4.6	98	98	4.4	96	96	3.1
F20	Falkland	86	88	6.5	98	98	5.6	100	100	4.9	100	100	3.5
F21	Falkland	98	98	6.7	100	100	7.1	94	94	5.7	98	98	5.5
F23	Falkland	100	100	7.5	96	96	6.0	88	92	5.7	84	84	5.3
F25	UBCC	22	60	30 +	88	88	10.2	84	84	7.2	90	90	5.7
F27	UBCC	0	74	30 +	38	82	30 +	70	84	8.6	90	90	6.2
F30	UBCC	20	74	30 +	82	92	7.7	100	100	7.7	88	88	8.5
F32	UBCC	4	48	30 +	22	48	30 +	100	100	8.2	100	100	6.4
F33	UBCC	0	40	30 +	42	60	14.5	98	98	6.3	96	96	6.6
F34	UBCC	2	48	30 +	46	66	15.5	92	92	7.9	92	92	5.9
F35	UBCC	18	96	30 +	38	90	30 +	64	88	12.9	76	84	9.5
F89	Mt. Benson, V.I.	—	—	—	78	92	12.2	92	92	8.8	94	96	6.1
F90	Cedar, V.I.	—	—	—	38	88	30 +	82	86	9.6	90	94	7.4

¹ UBCF—University Research Forest, Haney, B.C.

UBCC—University of B.C. campus area, Point Grey, B.C.

TABLE 3
G, GC, AND R FOR 26 LOTS OF DOUGLAS FIR SEED FOLLOWING
STRATIFICATION FOR 0-120 DAYS. EXPERIMENT 1. INCUBATION AT 15°C.

Lot No.	Source ¹	Strat. 40 d.			Strat. 80 d.			Strat. 120 d.		
		G	GC	R	G	GC	R	G	GC	R
F1	UBCF	12	74	30+	94	94	18.1	66	66	11.9
F2	UBCF	6	74	30+	76	90	17.8	94	100	12.4
*F4	UBCF	2	74	30+	8	72	30+	24	62	30+
*F7	UBCF	4	78	30+	14	88	30+	38	76	27.0
*F8	UBCF	44	76	23.5	72	76	15.6	52	60	14.5
*F9	UBCF	4	92	30+	8	68	30+	48	68	18.2
*F10	UBCF	4	96	30+	60	92	15.8	88	92	11.1
*F11	Campbell R.	12	96	30+	98	98	12.8	100	100	10.6
*F12	Campbell R.	0	90	30+	14	80	30+	32	70	30+
*F13	Campbell R.	0	68	30+	4	52	30+	14	46	30+
*F14	Campbell R.	0	92	30+	2	88	30+	44	96	30+
F15	Solsqua	74	78	12.6	22	84	30+	4	88	30+
F16	Alta Lk.	20	64	30+	94	94	14.3	74	74	9.4
*F17	Falkland	92	94	8.3	98	98	7.0	92	92	4.8
*F20	Falkland	98	98	9.0	90	92	8.2	100	100	5.6
*F21	Falkland	94	94	10.9	96	96	10.4	98	98	8.7
*F23	Falkland	72	78	10.8	92	92	10.4	100	100	9.4
*F25	UBCC	22	76	30+	94	94	12.8	96	96	9.4
*F27	UBCC	0	72	30+	54	78	14.5	82	86	9.4
*F30	UBCC	8	76	30+	74	92	13.2	76	86	15.5
*F32	UBCC	0	50	30+	96	96	15.1	96	96	9.8
*F33	UBCC	2	88	30+	100	100	12.2	100	100	10.6
*F34	UBCC	4	70	30+	96	96	13.9	96	96	9.2
*F35	UBCC	6	84	30+	2	94	30+	60	78	15.2
F89	Mt. Benson, V.I.	—	—	—	92	94	13.2	98	100	10.3
F90	Cedar, V.I.	—	—	—	90	100	17.2	88	96	10.3

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* Single-tree collection.

- surface with previous recommendations based upon attaining maximum rate of germination following treatment (Allen, 1941; Allen and Bientjes, 1954).
3. Extended stratification for 80 days or longer was of value when incubation temperatures were low. Relatively rapid germination occurred at 15°C following 80-120 days' stratification. Since this is of the order of surface-soil temperature during the natural outdoor germination period for Douglas fir, long stratification which simulates overwintering outdoors may have value in nursery practice when fall sowing is impossible or inconvenient. Extended treatment and early spring sowing should encourage early root establishment prior to top development.
 4. Most seed lots showed their full germinative capacity at all three incubation

temperatures provided that the seed was adequately stratified (80 days or longer) in the case of the two lower temperatures. To this extent, the germinative capacity indicated by the tests was the same regardless of the duration of stratification and the incubation temperature.

Experiment 2

Seed from seven lots, after 120 days' stratification, was treated in one of three ways.

- (a) dried at room temperature for 7 days in open dishes, soaked 16 hours at room temperature, and incubated at 25°C.
- (b) dried as in (a), then stored in capped vials at room temperature for an additional 7 days, soaked, incubated at 25°C.
- (c) dried as in (a), then stored in capped vials for an additional 525 days at room temperature, soaked, incubated at 25°C.

The results are given in Table 4 together with the pertinent data from Table 1 for comparison. The highlights are as follows:

TABLE 4
G, GC, AND R FOR SIX LOTS OF DOUGLAS FIR SEED STRATIFIED 0-120 DAYS
FOLLOWED BY DRYING AND STORAGE. EXPERIMENT 2. INCUBATION AT 25°C.

		G	GC	R	G	GC	R	G	GC	R
		Lot F1			Lot F2			Lot F11		
Strat.	0	90	90	6.6	96	96	15.0	92	92	14.5
	10	86	86	4.3	92	92	3.9	100	100	3.9
	20	100	100	4.6	96	96	3.8	96	96	3.6
	40	90	90	3.9	98	98	3.8	96	96	2.9
	80	94	94	3.5	96	96	3.5	100	100	2.8
	120	82	82	2.9	92	92	2.9	98	98	2.6
	120 - a ¹	72	72	2.6	92	92	2.8	100	100	2.3
	120 - b ²	60	60	2.5	100	100	3.0	96	96	2.6
	120 - c ³	68	68	5.1	—	—	—	92	92	7.9
		Lot F25			Lot F32			Lot F33		
Strat.	0	86	86	5.9	96	96	9.0	86	86	24.0
	10	90	90	2.8	100	100	2.8	98	98	4.5
	20	90	90	2.9	98	98	2.9	94	94	4.2
	40	82	82	2.8	100	100	3.2	96	96	3.4
	80	96	96	2.7	96	96	2.6	98	98	2.6
	120	92	92	2.4	100	100	2.5	98	98	2.5
	120 - a	92	92	2.3	100	100	1.7	100	100	2.4
	120 - b	80	80	2.0	100	100	1.7	100	100	2.4
	120 - c	60	60	7.2	88	88	5.1	92	92	6.9

¹ 120-a = residue from 120 days strat., dried R.T. 7 days open vials; soaked 16 hrs. R.T., incub. 25°C.

² 120-b = as "a" but stored 1 week after drying, in capped vials at R.T.

³ 120-c = as "b" but stored in capped vials 525 days R.T.

Treatment (a) produced no loss in germinative capacity, and either no change or a slight increase in rate of germination. Drying at room temperature for 7 days to approximately 10 per cent moisture content, following stratification, had no measurable harmful effects.

Treatment (b) generally resulted in no unfavorable effects.

Treatment (c), with the seed dried to about 10 per cent moisture content and held there for about 18 months, generally had little effect upon germinative capacity but did decrease the rate of germination markedly. Nevertheless, only in one case definitely (F25) of six tested, did the rate slow down to that of the original unstratified seed. To this extent, under the conditions of the experiment, stratification effects were only partially nullified by low moisture content brought about by extended storage.

Experiment 3

Seed of a very slow lot, F34, was stratified for 64 days at a moisture content of about 80%. Samples were tested at 10°, 15°, and 25°C after the following treatments;

- A. no further treatment.
- B. dried at room temperature 4 days to moisture content of about 10 per cent.
 - (a) stored in capped vials at room temperature for 16 days.
 - (b) no further storage.
 - (c) stored open at room temperature for 16 days.
- C. As in B but dried at 0-2°C to about 40 per cent moisture content:
 - (a) stored in capped vials at 0-2°C for 16 days.
 - (b) no further storage.
 - (c) stored open at 0-2°C for 16 days.
- D. Residue from 64 days' stratification stored open at 0-2°C for 274 days.
- E. Unstratified control.

The results are given in Table 5 with highlights as follows:

TABLE 5
G, GC, AND R FOR SEED LOT F34 STRATIFIED 64 DAYS FOLLOWED BY VARIOUS STORAGE TREATMENTS. EXPERIMENT 3. INCUBATION AT 25°, 15°, AND 10°C.

Treatment	Incubation Temperature								
	25°C			15°C			10°C		
	G	GC	R	G	GC	R	G	GC	R
A.	92	92	3.3	100	100	7.5	100	100	13.1
B.(a)	94	96	5.7	28	96	50 +	0	100	50 +
(b)	100	100	4.2	88	100	9.6	40	92	50 +
(c)	84	88	5.7	7	92	50 +	0	92	50 +
C.(a)	94	94	2.8	96	96	6.2	92	92	9.2
(b)	96	96	3.8	96	100	8.5	96	100	14.2
(c)	94	94	3.8	98	98	9.4	100	100	14.7
D.	96	96	7.0	—	—	—	—	—	—
E.	72	90	21.7	—	—	—	—	—	—

Germinative capacity held up under all treatments with the possible exception of B(c). Rate of germination was increased by C(a) but in all other treatments the rate was lower than for A, incubated after 64 days' stratification. At 25°C, no treatment nullified the stratification effect completely. Drying at room temperature slowed germination more than did drying for the same period of time at 0-2°C. But further storage at 0-2°C at 40 per cent moisture content, for a total stratification of 80 days, had a further stratification effect, nearly equivalent in total of that provided by 120 days' stratification (see Tables 1, 2 and 3,—F34).

In general, no ill effects resulted from drying the seed after 64 days' stratification but the effects of stratification were partially nullified by drying to 10 per cent moisture and holding for 16 days. Drying to 40 per cent and further storage at 0-2°C, however, were beneficial; this is consistent with the view expressed previously (Allen, 1962) that 40 per cent moisture content permits effective stratification.

Experiment 4

Seed lot F34 (see Tables 1-3), a relatively "dormant" lot, was treated as follows:

- A. Stratified 34 days.
- B. As A, but dried open at room temperature for 7 days to approximately 10 per cent moisture content.
- C. As B, but room storage continued for a total of 14 days.
- D. Unstratified.

The seeds were tested at 15°C and 25°C for 40 days, with the following results:

TABLE 6
G, GC, AND R FOR SEED LOT F34 UNSTRATIFIED AND STRATIFIED FOR 34 DAYS
FOLLOWED BY DRYING AND STORAGE. EXPERIMENT 4. INCUBATION AT
25° AND 15°C.

Treatment	Incubation Temperature					
	15°C			25°C		
	G	GC	R	G	GC	R
A. Stratified	78	98	11.3	98	98	3.8
B. Stratified, stored 7 days	6	100	40 +	96	98	4.0
C. Stratified, stored 14 days	—	—	—	82	96	6.5
D. Unstratified	—	—	—	56	94	24.0

These data indicate that the 7-day drying had little effect on behavior at 25°C but a marked effect on that at 15°C. Extension of room-temperature storage to 14 days, on the other hand, slowed germination at 25°C but had no effect upon germinative capacity. C still retained a considerable stratification effect compared with D, but less than that shown after 10 days' stratification (see Table 1, F34).

Experiment 5

Collections were made from two adjacent trees, 10/34 and 11/34 in September, 1959, when cones were browning (lots F27 and F34, respectively,

Table 1) and again in January, 1960, when about two-thirds of the seed had been disseminated.

The four lots were tested shortly after collection at 25°C unstratified, and at 15°C and 25°C after 20 days' stratification, and again in September-November, 1960, at 25°C unstratified. The unused residue from the January, 1960, collection was left in stratification for a total of 288 days and then tested at 25°C.

TABLE 7
G, GC, AND R FOR SEED FROM TWO TREES, COLLECTED IN SEPTEMBER, 1959,
AND JANUARY, 1960. EXPERIMENT 5.

A. Tested shortly after collection, 25°C, unstratified.								
Incubation Temperature	Tree No.	Collected Sept./59			Collected Jan./60			R
		G	GC		G	GC		
25°C:	10/34	74	78	17.8	92	92		7.4
	11/34	96	96	22.5	94	94		12.7
B. Tested March-April, 1960, at 15° and 25°C, stratified 20 days.								
25°C:	10/34	78	78	4.0	—	—		—
	11/34	86	86	4.4	—	—		—
15°C:	10/34	0	74	40 +	76	90		8.8
	11/34	2	48	40 +	74	95		8.7
C. Tested in September-November, 1960, after sealed cold storage, unstratified.								
25°C:	10/34	—	—	—	100	100		13.0
	11/34	96	98	30.7	96	100		17.0
15°C:	10/34	—	—	—	4	90		40 +
	11/34	—	—	—	4	100		40 +

D. Tested at 25°C after 288 days' stratification, January, 1960, collection only:

10/34. All germinated at 0-2°C; G = 85.7%.

11/34. 30% germinated at 0-2°C; G of remainder = 96%; R = 0.67.

The data under A indicate that the equivalent of stratification took place on the tree within the cones between September and January and resulted in faster germination. The data under B show a marked effect of "storage" on the tree on germination rate at 15°C following 20 days' stratification; the late collection behaved as if it had had between 40 and 80 days' stratification (see Table 2, F27 and F34) rather than 20. The results under C show that approximately six months' dry cold storage slowed germination of both the early and the late collections. The data under D are of interest only because of the germination obtained at 0-2°C and the maintenance of full germinative capacity during 9 months' stratification.

Experiment 6

Two U.B.C. Forest collections, U21 and U22, were stratified in the normal way (A), and open in the refrigerator at 0-2°C (B) for 43 days and 266 days. Moisture contents of U21 and U22 at the beginning of stratification were 80% and 86%, respectively. At the end of 43 days no change had taken

place in A whereas, in B, moisture content had dropped to 24-25. By the end of 288 days of stratification, A had dropped to 6-7% and B to 11-13% moisture content. Tests were conducted at 15°C and 25°C with the following results:

TABLE 8
G, GC, AND R FOR TWO SEED LOTS STRATIFIED IN TWO WAYS FOR 43 AND 266 DAYS. EXPERIMENT 6. INCUBATION AT 25°C AND 15°C.

Treatment Period	Incub. Temp.	Treatment A						Treatment B					
		G	U21 GC	R	G	U22 GC	R	G	U21 GC	R	G	U22 GC	R
43 days	25°C	92	92	3.4	88	88	2.8	84	84	3.8	90	90	4.3
	15°C	94	94	7.5	98	98	6.3	90	92	9.8	92	96	9.9
266 days	25°C	78	78	8.3	86	90	7.3	92	92	7.5	92	92	7.6
	15°C	72	76	21.7	88	90	17.5	76	90	20.3	90	90	18.2

With stratification for 43 days, treatment B was somewhat less effective than A, particularly when incubation was at 15°C. During extended stratification with steady drying of seed, the effectiveness of treatment gradually lessened; some loss of germinative capacity was indicated for U21 under treatment A.

Experiment 7

Seeds from reciprocal crosses were collected in September, 1958, and tested at 25°C the following summer after 51 days' stratification. The residue was left in capped vials for a total storage of 275 days at 0-2°C during which the moisture content dropped from about 60% to an estimated 20%. These seeds were tested at 25°C. The following show the results of the original tests made in 1959 and the final made in 1960.

TABLE 9
G, GC, AND R FOR DOUGLAS FIR SEED FROM RECIPROCAL CROSSES UNTREATED AND STRATIFIED FOR 51 OR 275 DAYS. EXPERIMENT 7. INCUBATION AT 25°C.

Cross	1959 Test						1960 Test		
	Untreated			Strat. 51 days			Strat. 275 days		
	G	GC	R	G	GC	R	G	GC	R
E _o x A	87	97	17.4	94	94	4.8	90	90	5.6
E _o x B	90	90	10.0	—	—	—	100	100	5.6
B _o x E	87	97	13.9	88	88	3.7	100	100	4.2
B _o x A	27	85	30+	87	87	3.4	100	100	3.5

The most striking aspect of these data is the ability of carefully processed seed to endure, without loss, up to 9 months' stratification. The untreated seed varied widely in "dormancy" at 25°C but much less after either 51 days' or 275 days' stratification.

Experiment 8

The residue of Interior seed lots F17, F18, F20, F21 and F22 from the study reported in part as Experiment 1, after 120 days' stratification, was treated

as follows:

- A. no further treatment.
- B. dried 7 days open at room temperature to moisture content of about 10%.
- C. as B but at room temperature 14 days.
- D. dried and stored open in refrigerator at 0-2°C for 14 days.

TABLE 10
G, GC, AND R FOR FIVE INTERIOR SEED LOTS STRATIFIED FOR 120 DAYS
FOLLOWED BY DRYING AND STORAGE. EXPERIMENT 8.
INCUBATION AT 25°, 15°, AND 10°C.

Lot No.	Treatment	Incubation Temperature								
		25°C			15°C			10°C		
		G	GC	R	G	GC	R	G	GC	R
F17	A	98	98	0.8	96	96	3.1	92	92	4.8
	B	96	96	1.6	100	100	2.4	96	96	3.7
	C	100	100	1.2	96	96	3.1	92	96	3.9
	D	92	92	1.3	96	96	2.7	100	100	3.5
F18	A	92	92	3.0	80	88	7.1	82	88	11.6
	B	72	72	2.7	80	80	6.2	76	76	7.6
	C	72	80	3.3	56	92	7.5	24	80	21+
	D	80	80	2.8	88	88	6.2	43	74	12.7
F20	A	100	100	1.5	100	100	3.5	100	100	5.6
	B	92	92	2.0	92	92	2.8	100	100	4.9
	C	90	90	2.1	76	84	3.7	92	100	6.1
	D	78	78	1.8	92	92	3.0	92	96	4.7
F21	A	92	92	2.2	98	98	5.5	98	98	8.7
	B	96	96	2.8	96	96	5.2	100	100	6.6
	C	—	—	—	—	—	—	—	—	—
	D	100	100	2.6	100	100	4.9	100	100	7.0
F22	A	94	94	2.8	94	94	6.4	88	88	10.2
	B	80	80	3.0	88	88	4.2	88	88	6.0
	C	86	90	3.2	76	94	6.1	68	88	10.5
	D	90	90	4.2	80	90	6.0	80	92	7.8

Using A for each seed lot as the basis for comparison, it is apparent that treatment B has generally resulted in faster germination at 15°C and 10°C but not at 25°C. This behavior may be accounted for by the effect of higher temperature during drying at room temperature (25-25°C) while the seed was still moist enough to react. On the other hand, the effect of treatment D was to extend stratification by 14 days and increase the rate of germination of some lots at the lower temperatures. In very few cases did any significant drop in germinative capacity result from treatments B or D compared with A.

Treatment C, however, resulted either in slower germination than A or in about the same rate, suggesting that any advantage of higher temperature during initial drying was offset by the extended period at low moisture content during which the stratification effect was gradually lost.

CONCLUSIONS

Stratification nearly always accelerates germination without affecting germinative capacity. Its effect is increased with duration of treatment, particularly at low incubation temperatures. At 25°C the first few days of stratification have the greatest relative effect upon rate of germination.

Stratification effects are slowly reversible as moisture content is dropped below about 40%. Even prolonged storage following stratification and drying to about 10% rarely offset the stratification effect completely and have little if any effect upon germinative capacity in the case of high quality seed. Reversal of stratification effects is more pronounced at low incubation temperatures than at high; "secondary dormancy" at low temperatures results quickly from drying but germinative capacity is retained.

The results suggest that good quality seed can be dried after stratification, if not needed, and stored at 0-2°C for at least one year with little loss. It can be stratified for 120 days or longer without loss of viability in most cases and then either used, continued in cold storage with gradual drying, or dried down to 10 per cent quickly and re-stored at 0-2°C. Even limited room-temperature storage at 10 per cent moisture is satisfactory for period of a few months.

One advantage of extended stratification is its hastening effect on germination at low temperatures, enabling protrusion of the radicle and root establishment early in the growing season. Short stratification, or long stratification followed by drying and further storage, do not provide this advantage. On the other hand, for seed testing a short period of stratification is adequate since incubation temperatures are near optimal.

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