MIDPOINT OF THE RANGE OF GERMINATIONS
FOR EACH COLLECTION DATE AND TREATMENT

*Seedfall Sept. 22, 1983 approx.*
THE EFFECT OF TIME OF CONE COLLECTION, CONE AND SEED MATURATION INDICES AND 
AFTER-RIPENING ON SEED QUALITY OF INTERIOR SPRUCE.

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SUMMARY

Seasonal changes in cone and seed maturity were observed in a limited sample from 
a stand of mature white spruce [Picea glauca (Moench) Voss] in the Star Creek 
drainage, East of Lumby, B. C. Indices of cone and seed maturity were correlated 
with seed germination following the application of three (3) after-ripening treatments. 
No physical cone criteria effectively described seed ripeness, though a general 
relationship was evident: little sound seed was obtained from collections made 
when cone moisture content exceeded 100 percent. After-ripening substantially 
improved laboratory germination, with the cool, moist condition (5°C and 80+ percent 
humidity) providing the greatest enhancement particularly in the mid picking dates. 
The operational aspects of cone collection are discussed in light of these findings 
and regional criteria are suggested.

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INTRODUCTION

Much has been written on cone maturity and the quality of conifer seed. Attention is directed to the excellent review of this subject by Edwards, 1980. Several published works pertain to maturity and field collections of white spruce (Picea glauca (Moench) Voss) (Crossley, 1953; Cram and Warden, 1957; Revel, 1969 [report unpublished]; Zasada, 1973 and Winston and Haddon, 1981). Notwithstanding a conscientious effort on the part of most collectors to harvest ripe cones and seeds, the field forester yearly confronts the question of when to start operational cone collections. Subjective criteria involving embryo development and embryo endosperm shrinkage have been suggested (Dobbs et al., 1976); however, these criteria are not sufficiently quantitative to ensure the collection of a ripe crop. Seed of high germination and vigor is required for nursery container production to maximize the number of filled cavities and be cost effective, and for bareroot production to offset the fairly harsh germination environment often experienced in nursery seedbeds.

The operational aspects of harvesting seed of the required quality is difficult and confounded by a number of factors. Under warm, dry climatic conditions, mature spruce cones open rapidly and disperse seed. Operationally, this has meant scheduling cone collections earlier than the optimum stage of ripeness to avoid seed flight and an uncollected crop. The 10 to 15 year periodicity of good cone crops in spruce, and diminishing seed inventories may require large collections even when medium sized cone crops are identified.

Despite the findings of several researchers on the potential of artificial ripening for increasing germination of immature seed (Winston and Haddon, 1981, Zasada, 1973), the technique has not been used extensively in spruce. In order to increase
Introduction (cont'd)

the viability of spruce seed without reducing the length of the collection season
or risking the loss of a heavy cone crop, an understanding and verification of
the effects of after ripening on seed maturity is necessary.

The study reported here was designed to relate field evaluations of cone ripeness
(embryo-cavity ratio of seed) with laboratory evaluation (moisture content and
specific gravity of cones) and seed germination following cone after-ripening by
the following methods:

1. Immediate extraction.
2. After-ripen at ambient air temperature for 4 weeks.
3. After-ripen at 5°C and 75 to 100 percent relative humidity
   for 4 weeks.

This is not a study of either population or environmental variation, nor does the
trial consider the operational techniques of cone after-ripening now practiced.
The importance of understanding both environmental and population variations,
as well as techniques for after-ripening, is recognized.
METHODS

A. Stand and tree selection:

The stand chosen for this study is a moderately stocked, mature to decadent, mixed aged Spruce-Balsam stand (inventory type SB 831-M) in the upper Star Creek drainage. It is located 35 kilometers North and East of Lumby, B. C. at approximately 1525 meters elevation. The area is moderately sloped (25 to 50%) and rolling with fair drainage and East to South-east aspect.

On each collection date, three trees were randomly selected from among those that had a heavy cone crop and exhibited differences in cone color. The study area was an active cutting permit of Weyerhaeuser Canada Ltd. (C.P. 60). The selected trees were all operationally felled.

A minimum of 650 normal, mostly insect-free cones were picked from each tree. These cones were divided into 2 replications of 3 treatments (6 samples) of approximately 100 cones each. In addition, 50 randomly selected cones from each tree were saved for immediate determination of moisture content, specific gravity, and embryo-cavity ratio.

Cones were picked on the following dates:

- August 17th (1 tree only)
- August 19th
- August 25th
- August 27th
- September 10th
- September 17th
METHODS (cont'd)

A. Stand and tree selection (cont'd)

An additional collection was attempted on September 24th, at which time the cones had mostly opened. Insufficient seed was available to make a viable sample, as most of the heavier (filled) seed had already fallen. From this observation, it was estimated that seedfall had occurred during the previous two days (September 22nd to 24th).

B. After-ripening treatments:

1. Immediate extraction - On each collection date, two samples of cones from each tree were placed in a drying oven to open the cones. Cones remained in the oven (at 35°C to 45°C) until they were dry and cone scales fully reflexed.

2. After-ripen at ambient air temperature for 4 weeks - On each collection date two samples of cones from each tree were put in a shaded but open storage area on a wood pallet, 0.6 meters off the ground for air circulation. The paper bags were left open and the cones agitated periodically for improved ventilation. After 28 days of after-ripening, the bags were placed in the drying oven and the seed extracted.
B. After-ripening treatments (cont'd):

3. After-ripen at 5°C and 75 to 100% relative humidity for 4 weeks - The last two samples from each tree were placed in a controlled environment chamber set at 5°C. Temperature and relative humidity were monitored during the initial seven days for calibration. Temperature ranged from 3°C to 7°C and relative humidity from 75% to 100%. After 28 days of cold after-ripening, the bags were placed in the drying oven and the seed extracted.

C. Determination of Cone and Seed Maturation Indices:

1. Embryo-Cavity ratio - On each collection date, ten insect free cones were randomly selected from each sample tree. Three or four seeds were extracted from each cone. From these 30 to 40 seeds, 10 full seed were randomly selected and sliced longitudinally to expose the largest cut surface. The cut surface was observed under a 10X hand lens, and percentage of endosperm cavity occupied by the developing embryo was estimated visually to the nearest 10%. The average of these ten estimates was taken as the embryo cavity ratio (E/C). Estimation of embryo development was made by the same person for the sake of consistency.
METHODS (cont'd)

C. Determination of Cone and Seed Maturation Indices (cont'd):

2. Specific Gravity - A random sample of five cones from each tree at each collection date was tested for specific gravity (SG) by the volumetric method. Each cone was weighed to the nearest 0.01 gram and, by water displacement in a graduated cylinder, the cone volume determined to the nearest 0.1 ml. Care was taken to establish a reading on the graduated cylinder as soon as the larger bubbles escaped from the cone scales. If the cone was left immersed in water for any length of time, it was found to gradually absorb water, resulting in an artificially high SG.

3. Moisture Content - At each sampling date, five randomly selected cones from each tree were cut in half (to assure thorough drying). The halves were weighed separately (green weight) and then placed in a drying oven at 35°C to 45°C. The cut cones were weighed periodically during drying to determine the length of time required for cone weight to stabilize. After approximately 1½ days in the oven, no further weight reductions occurred. The moisture content of the cones was expressed as the change in cone weight relative to the dry cone weight.
METHODS (cont'd)

D. Extraction and Seed Processing:

When the cones were sufficiently dry, the seed was extracted using the tumbling drum at the Ministry of Forests Kalamalka Research Station. The extracted, unseparated seed was forwarded to the Ministry of Forests Seed Centre at Duncan, B. C. for cleaning, x-ray, and germination testing. The seed was hand screened to remove pitch and fine particles but not dewinged or separated.

E. Germination Testing:

From each sample, two 100-seed subsamples were randomly selected. Each subsample was placed in a plastic germination box on blotting paper over wet perlite. The seed was not pre-soaked or stratified. Humidity during germination was controlled by adding a measured amount of water to each box. The germination boxes were placed in Conviron incubators for 25 days. Germination counts were recorded on the 4th, 7th, 9th, 11th, 14th, 16th, 18th, 21st, 23rd, and 25th days for each subsample. Environmental conditions in the incubators were as follows:

Day 0 to Day 7 : 16 hrs./day dark @ 20°C and 8 hrs./day light @ 30°C.

Day 8 to Day 25 : 16 hrs./day dark @ 20°C and 8 hrs./day light @ 20°C.

It is important to note that because the seed in this study was not separated, pre-soaked, or stratified prior to testing, the germination results are not directly comparable to the results from operationally collected seed.
RESULTS

A. Germination of Extracted Seed:

Results of this study are based on total germination after 25 days. Seed germination exhibited a seasonal increase with the number of days elapsing from August 17th, 1982 to seed flight, which occurred between September 22nd and 24th, 1982. Fresh extraction of cones, with no after-ripening, yielded few germinants prior to September 10th, 1982, only two weeks ahead of natural seed flight (Figure 1). A sigmoid curve may have fit the data better and facilitated interpretation but the mathematical complexity was beyond the capability of our computation equipment.

\[ g = 0.02 e^{0.15t} \quad (r^2 = 0.51) \]

\[ \text{DATE OF COLLECTION} \]

\[ \text{TIME OF COLLECTION (days)} \]

\[ \text{GERMINATION (\%)} \]

**FIGURE 1.** Immediate Extraction. Percent laboratory germination of seed extracted from cones immediately after collection.
RESULTS (cont'd)

A. Germination of Extracted Seed (cont'd):

After-ripening cones, either by ambient air temperature storage (Figure 2) or by the cool, moist storage reported by Winston and Haddon (1981) (Figure 3), significantly improved germination totals of the earlier collections.

\[ g = 20.561n - 9.88 \quad (r^2 = 0.60) \]

**FIGURE 2.** Ambient Temperature, Percent Laboratory Germination of Seed, extracted from cones following four weeks of cone after-ripening at air temperature.
Germination of sample lots exposed to cool, moist after-ripening conditions was consistently greater than the traditional dry after-ripening. This conclusion was statistically verified by an analysis of variance performed on the germination results obtained with the September 17th, 1982 collection (Appendix I) and with Kruskal-Wallis rank tests performed on August 19th, and 30th and September 17th, 1982 collections (Appendix II).
RESULTS (cont'd)

A. Germination of Extracted Seed (cont'd):

The effect of after-ripening was significant at each collection period. In addition, while only 3 trees were collected and included at each sampling time, there were significant differences between the trees. The interaction between trees and after-ripening was significant. Referring specifically to the September 17th, 1982 collection (Appendix I), after-ripening and trees were significantly different at the 99% confidence level, and the tree x after-ripening interaction significant at the 75% level (a level assumed in this case to represent an event of biological significance).

The Kruskal-Wallis rank test performed for the ambient and controlled environment after-ripening treatments for each tree showed that two trees were significantly different. However, seed and cones from one tree were not enhanced by the cool after-ripening (Appendix II).

No conjecture is presented to explain the significance of the after-ripening by tree interaction, or why Tree 3 might not be expected to respond to the cool, after-ripening condition.

It is probable that greater total germination for all treatments would have occurred if the seed had been pre-soaked and stratified prior to testing.
RESULTS (cont'd)

B. Cone Maturation Indicies:

The general pattern of cone maturation indices measured for the key ripening period from August 17th to September 17th, 1982 are displayed in Figure 4.

\[ y = 54.81 + 14.791x \quad (r^2 = 0.6) \]

\[ y = 173.3 - 23.791x \quad (r^2 = 0.43) \]

**Figure 4.** A description of cone and seed maturation indices in spruce as they change during the 1982 cone collection period at Star Creek, east of Lumby, B.C.
RESULTS (cont'd)

B. Cone Maturation Indicies (cont'd):

Field calls of the embryo-cavity ratio increased from an average of 60 percent elongation in August 17th, 1982 and reached 100 percent between August 27th and 30th, 1982. This was maintained through the remainder of the study period. No discernable trend could be firmly established between specific gravity and time of collection. In the period up to 5 days before seed flight, this value was not useful as an indicator of cone ripeness.

Cone moisture content decreased from August 19th through September 17th, 1982 to a low of approximately 95 percent of oven-dryweight. While this value per se is not highly correlated with germination values and hence cone ripeness (Figure 5), 95 percent cone moisture content appears to be an operationally useful index at least in terms of defining the earliest "safe" collection date.

The germination quality of spruce seed is shown to be a function of cone ripeness at the time of collection (a factor primarily influenced by date of cone collection and the inter-tree variability in ripeness) and cone after-ripening treatments. After-ripening cones in a cool (5°C), moist environment for a 4 week period may enhance quality of seed from cones collected green and up to 6 weeks before natural seed flight. Mature seed that does not require cone after ripening was collected only in a narrow time frame within 1 week of natural seed flight. None of the commonly used cone ripeness criteria evaluated -- specific gravity, moisture content or field embryo-cavity ratio...
RESULTS (cont'd)

B. Cone Maturation Indicies (cont'd):

-- proved effective indicators of ripe seed yield. However, it was clear that even with after-ripening, little quality seed might be expected from collected cones whose moisture content at the time of collection exceeded 100 percent, or whose field embryo-cavity ratio was less than 100 percent.

FIGURE 5. SEED QUALITY AS A FUNCTION OF FIELD EMBRYO-CAVITY RATIO CALLS AND CONE MOISTURE CONTENT.
OPERATIONAL IMPLICATIONS

Operational cone collections in the Kamloops Region generally follow an established routine. Stands identified as having collectable cone crops are monitored for cone ripeness and seed maturity beginning in early August. During the latter part of August, cone samples are taken and the seed cutting test is used to estimate seed maturity. When the majority of the seed embryos exceed 75 percent of the length of the cavity within the endosperm, then falling of trees and collecting of cones commences. This usually occurs in the last week of August or the first week of September, depending upon the weather patterns in that year.

Following cone collection, full sacks are usually delivered to Ministry of Forests District Offices for checking, storage, and shipment to processing facilities. During good cone seasons, storage facilities are often at a premium. Cone collectors are not usually aware of the details of cone handling after seedlots are delivered to the District Office.

In 1982, eleven spruce seedlots were operationally picked by Weyerhaeuser Canada Ltd. between August 24th and September 6th, from Blue River in the North to Penticton in the South. Cone and seed maturation indicies were measured and recorded for each seedlot as picking commenced. These are summarized below.

<table>
<thead>
<tr>
<th></th>
<th>Range</th>
<th>( \bar{x} )</th>
</tr>
</thead>
<tbody>
<tr>
<td>Embryo cavity (E/C) ratio:</td>
<td>70% - 100%</td>
<td>90%</td>
</tr>
<tr>
<td>Moisture Content</td>
<td>66% - 130%</td>
<td>93%</td>
</tr>
<tr>
<td>Specific Gravity:</td>
<td>.70 - .98</td>
<td>.81</td>
</tr>
</tbody>
</table>
OPERATIONAL IMPLICATIONS (cont'd):

If the results of this trial are applicable throughout the Region, then picking of all operational lots should have been delayed by as much as 10 days (especially for those lots with E/C ratios below 100%) to produce seed of the highest quality.

The timing of our collections in 1982 was driven by two primary factors:

1. The latter part of August and early part of September were warm and dry.

2. Embryo development was rapidly approaching full elongation, averaging 90% at the time of picking, which exceeded our minimum guideline of 75%.

A period of cool, wet weather commenced about September 5th, and the rest of the month remained cooler than average. These conditions appeared to have slowed final seed maturation and hence, date of seedfall. Although weather trends in any given season are difficult to predict, it would still seem that full embryo elongation is a reasonable target for commencing collections.

Optimum cone collection timing is only one of the important steps in providing quality seed. Post-collection cone storage and handling techniques are equally important in enhancing seed quality. The results of this study indicate that there may be more flexibility in picking date, if controlled cone storage and after-ripening techniques are applied. Average germination appears to be greatly increased on a range of early collection dates and moderately increased on seed picked at the optimum date. Consistency of seed germination which is of great importance to the nurseryman, may also increase.
OPERATIONAL IMPLICATIONS (cont'd):

A number of questions should be examined as a follow-up to this study. They include:

1) What after-ripening techniques produce the best average germination?

2) What, if any, are the effects of after-ripening treatments on seed physiology particularly as it relates to vigor and storage capability?

3) Will the observed lifts in average germination or increased seed consistency be realized in nursery beds or cavities?

Introduction of alternate after-ripening techniques such as the controlled atmosphere regime should not be delayed while these questions are answered.
LITERATURE

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No. 3: 98 p.

Edwards, D.G.W. 1980. Maturity and quality of tree seeds -- a state-of-the- 

Winston, D.A. and B. D. Haddon 1981. Effects of early cone collection and 
artificial ripening in white spruce and red pine germination. Can. J. 
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Zasada, J.C. 1973. Effect of cone storage method and collection date on 
Alaskan white spruce (Picea glauca) seed quality. In Proceedings of 
the I.U.F.R.O. International Symposium on Seed Problems. 
APPENDIX I. Analysis of variance of germination test results from cones collected on September 17th, 1982.

<table>
<thead>
<tr>
<th>Source</th>
<th>df</th>
<th>SS</th>
<th>MS</th>
<th>F</th>
</tr>
</thead>
<tbody>
<tr>
<td>Replications</td>
<td>3</td>
<td>94.08</td>
<td>31.36</td>
<td>1.32</td>
</tr>
<tr>
<td>Treatments</td>
<td>8</td>
<td>4804.39</td>
<td>600.55</td>
<td>25.23**</td>
</tr>
<tr>
<td>Ripening</td>
<td>2</td>
<td>1668.22</td>
<td>834.11</td>
<td>35.05**</td>
</tr>
<tr>
<td>Trees</td>
<td>2</td>
<td>2939.56</td>
<td>1469.78</td>
<td>61.76**</td>
</tr>
<tr>
<td>Trees x Ripening</td>
<td>4</td>
<td>196.61</td>
<td>49.15</td>
<td>2.07*</td>
</tr>
<tr>
<td>Error</td>
<td>24</td>
<td>571.17</td>
<td>23.80</td>
<td></td>
</tr>
<tr>
<td>Total</td>
<td>35</td>
<td>5469.64</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

1/ ns - not significant.
2/ ** - significant at the 99% confidence level.
3/ * - significant at the 75% confidence level.
APPENDIX II. Kruskal-Wallis rank test values summarized by after-ripening treatment and by tree for the September 17th, 1982 cone collection.

<table>
<thead>
<tr>
<th>Tree Number</th>
<th>After-ripening treatment</th>
<th>Immediate Extraction</th>
<th>After Ripened at Air Temperature</th>
<th>After Ripened at 0°C</th>
</tr>
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<tbody>
<tr>
<td>1</td>
<td></td>
<td></td>
<td>5.87(^1/)</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>6.84*</td>
<td></td>
</tr>
<tr>
<td>2</td>
<td></td>
<td></td>
<td>9.85*</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>5.33*</td>
<td></td>
</tr>
<tr>
<td>3</td>
<td></td>
<td></td>
<td>7.09*</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>1.04(^{ns})</td>
<td></td>
</tr>
</tbody>
</table>

\(^1/\) * significant at the 90% confidence level.