Tree Improvement Branch

Cone and Seed Improvement Program
BCMoF Tree Seed Centre

from **Seed and Seedling Extension Topics**
Volume 13 December 2001

2001 White Pine Quality Assurance Monitoring

Large differences in germination capacity have been documented between dormancy breaking protocols (stratification) performed in operational seed preparation [OSP] and in the lab [LAB] over the last several sowing seasons. We try and minimize procedural differences, but some attributes such as seed volume cannot be made equivalent between the two areas. The current methodology for western white pine (Pw) includes a 14-day running water soak followed by 98 days of cold stratification (germination test type=G55).

A total of twenty western white pine (Pw) 2001 sowing requests (SRQ) had 25 grams added to allow for a direct comparison in germination capacity (GC) (effectiveness in breaking dormancy) between the OSP and LAB methods. This allowed for a direct comparison between OSP and LAB methods:

For the samples obtained from OSP requests moisture content will be estimated i) after draining, just prior to surface drying; ii) after surface drying; iii) after 7 weeks or the half-way point in stratification and iv) at end of stratification = 14 weeks (98 days). The LAB samples will also have similar moisture content estimates performed, but since surface drying does not occur, only i, iii and iv will occur for lab sampling. The results will quantify variability in moisture content after draining and determine whether lab moisture content in stratification is closer to moisture content before or after surface drying in OSP. All moisture content estimates were based on targetting procedures.

Results

Germination

Even with our efforts to mimic the LAB procedures in OSP the results indicate that our OSP techniques are inferior in breaking dormancy to lab procedures. The germination capacity (GC) of the OSP requests averaged 58%, while the LAB and previous SPAR[1] GC both averaged 91% based on 19 samples[2]. **The falldown in germination ranged from 6 % to 60% indicating that the individual seedlot may have a large impact on the way white pine responds to OSP techniques**.

Extended Stratification

[1] SPAR – Seed Planning and Registry System – these GC test results indicate the previous test result on which sowing request sizes are based on (with seeds per gram).
[2] One sowing request was shipped to the nursery prior to testing and no SRQ germination or moisture content during stratification data was obtained.

For three sowing requests, the nursery requested that we hold the seed for an additional two weeks of stratification. Sampling and germination testing were performed after 14 weeks stratification (pretreatment completion) and after 16 weeks stratification. The additional two weeks stratification resulted in a 5% average increase in GC (71 to 76%). An additional request was tested after 15 and 16 weeks stratification with a 3% gain in GC.

Following the testing of operational sowing requests cutting tests were performed on ungerminated seed. Most seed appeared to be viable exhibiting a firm, white megagametophyte and a mature apparently healthy embryo. Nurseries that performed cutting tests on white pine requests also confirmed the apparent viability of ungerminated seeds. **These results indicate that stratification may not have been of a sufficient duration (or moisture content – *covered later*) to totally overcome dormancy.**

Stratification Unit Size

For white pine, sowing requests are divided into stratification units of 1000 grams or less (i.e. a 3200 g request is divided into 4 units of 800 grams each). All other species use a 3000 g minimum stratification unit size. It has been suggested that smaller 'units' may have higher moisture contents and this may increase germination in Pw. The coefficient of determination (r^2) between bag size and the moisture content i) after draining and ii) after surface drying both were 0.60. The moisture contents during stratification were not as well correlated with bag size [≈ 0.10 at 7 and 14 weeks] indicating that bag size has much less influence on stratification moisture content (at least after 7 weeks). The relationship between bag size and nursery germination falldown (Lab minus Nursery GC) was weak (r^2 =0.25), but it did indicate that smaller bag sizes are experiencing smaller falldowns in GC (Figure 1).

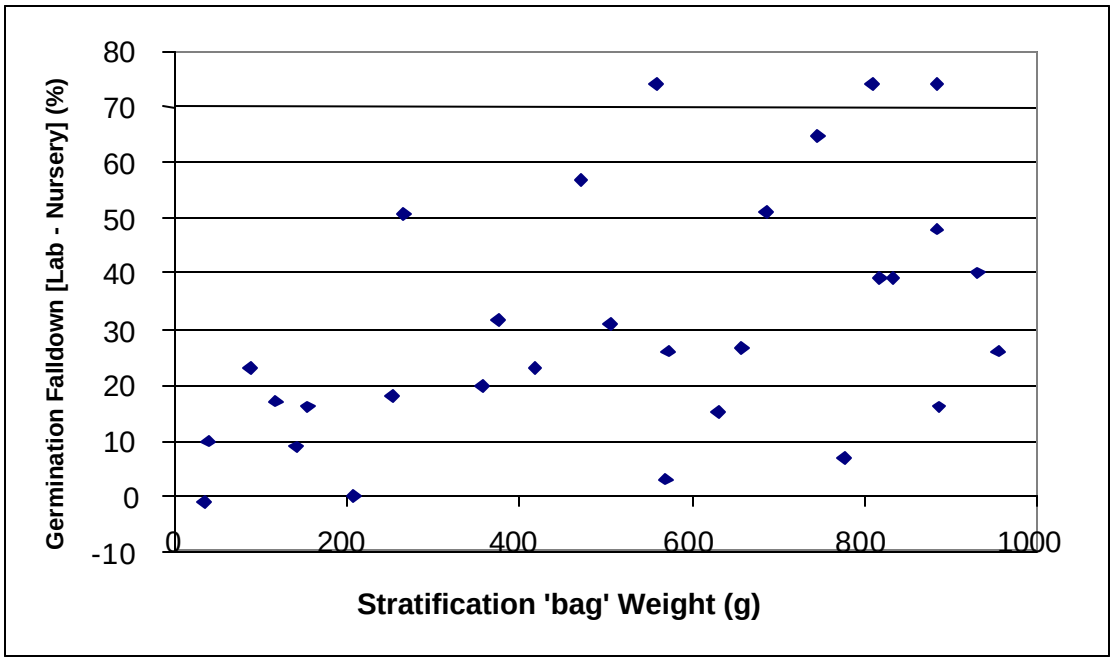

Figure 1. The relationship between sowing request bag size and germination falldown.

Moisture Content

For OSP the average moisture of the seed following a 14-day soak and drainage of excess moisture was 40.6%. After surface drying the moisture content was reduced to 35.5% indicating that surface drying removes 5.1% moisture from the seed. This was fairly consistent across seedlots and the r-squared value between drained and surface dry moisture content was 0.89. After surface drying the OSP samples have a moisture content 1.7% less than LAB samples.

The OSP samples appear to lose moisture during stratification in comparison to lab samples (Figure 2). This appears to be a significant difference in the two treatments : **lab samples are maintained in stratification at a moisture content approximately 3.4% greater than in OSP!** The lab samples appear to maintain fairly consistent moisture content from draining through to shipping (0.9% drop). The lab samples are not surface dried as the small samples (100 seeds) dry very quickly and could result in insufficient moisture within the seeds. The lab samples are also stratified in a closed unit that does not permit air exchange.

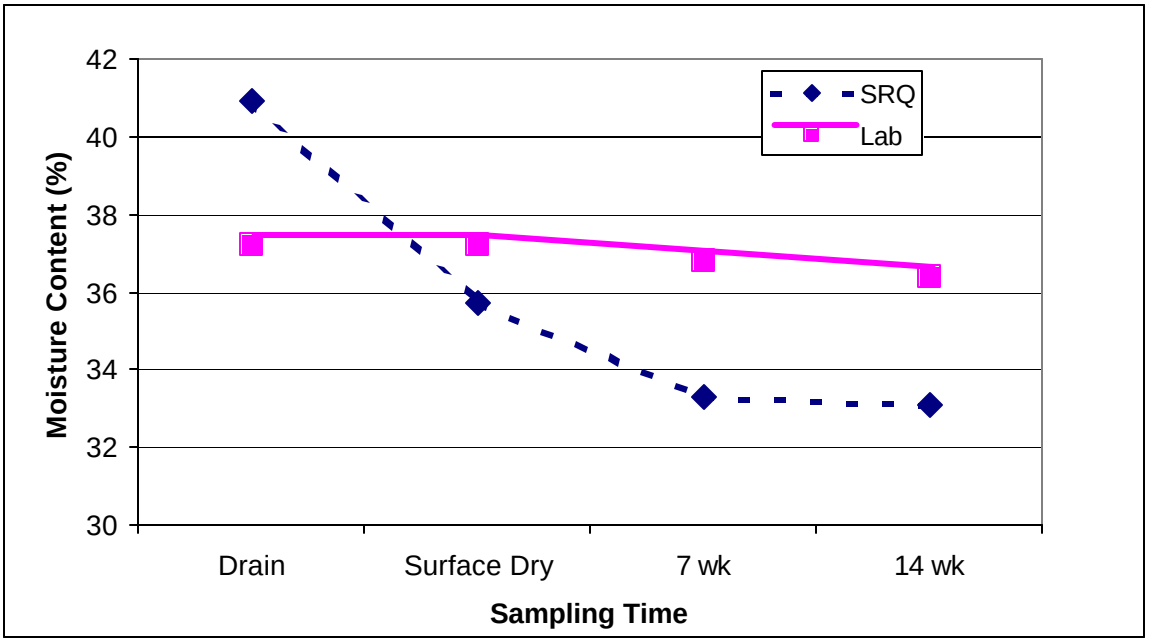

Figure 2. Comparison between moisture content after Drain, Surface Drying and 7 & 14 weeks of stratification for lab samples and operational sowing requests (SRQ) [n=19].

Discussion

The results clearly show that the conditions provided to pretreat LAB samples is not being replicated for OSP. The LAB prepared samples had an average germination 33% greater (91% vs. 58%) than that achieved with OSP. This difference is considered accurate as differences in sample time, location and methodology were virtually eliminated.

The most significant pretreatment difference is the size of the sample pretreated. In the Lab 100 seeds are used per replicate and up to a maximum of 1000 grams (approximately 50 000 seeds) are used in OSP. In the lab, seeds are not touching during stratification and there is a large air: seed

ratio. The container is closed during stratification in the lab. In OSP adjacent seeds are touching (from all directions) within the one Kilogram seed mass and the air:seed ratio and the kimpack:seed ratio is reduced relative to the lab. To provide an equivalent air: seed ratio in OSP one would need an air volume of 60 to 75 litres within the stratification unit for one Kg of seed [totally impractical].
The most obvious physiological difference between the Lab and OSP is the moisture content of the seeds during stratification. The average OSP moisture content of 33.1% appears insufficient to efficiently overcome dormancy compared to the 36.5% level experienced in the Lab. This relatively high moisture requirement is considered to be critical and will be the focus for improvements in OSP. For other species (i.e. Pli and Sx) the optimal moisture content for stratification is 30%.

The strategy adopted has been to try and mimic the tray system used successfully in the lab. This may not be appropriate as a container to precisely mimic this environment is probably too large to be practical. The tray system is also problematic as it occupies a large area and makes it difficult to monitor and 'manipulate' seeds. The tray system in OSP has not provided improved germination in the two years in which it was implemented (2000 and 2001.

The most frustrating part of working with Pw has been the failure of the lab-tested procedures to meet our expectations when conducted on operational quantities of seed (up to 1 Kg). It is possible that there is no one pretreatment that will produce equivalent results with small (100 seeds) and large (up to 500 000 seeds) quantities of seed! Feedback from some nurseries stratifying their own requests indicates that our problems are not unique, but some growers are quite successful at stratifying Pw. They emphasize that the seeds must be kept "moist" during stratification and that seed "manipulation" or the movement of seed within the stratification unit is required and is performed by some on a daily basis.

Recommendations

Several changes are being recommended for 2002 sowing of western white pine. The testing of recommendations is problematic due to the need to have operational quantities of seed available for trial purposes and the time required to obtain results with Pw pretreatments (approximately 5 months). These recommendations are directed specifically at OSP and no changes are being recommended for the lab testing of Pw.

1. Operational stratification of **Pw requests should revert to being performed in polyethylene bags**. Smaller sized stratification units (500 g) will be tested on a limited scale in 2002 to evaluate potential benefits.

2. The **moisture content of Pw requests stratified in OSP should be increased** to better correspond to the moisture content experienced in the lab. The increased moisture content can be accomplished by instituting the following:
    a) Eliminate surface drying performed on Pw sowing requests.
    b) Monitor moisture content during stratification. **All sowing requests stratified at the TSC should have fresh weights recorded following draining and after one month of stratification.** This will allow for an adjustment in moisture content**.**

c) I am recommending that the target moisture content range of Pw should be between 35 and 38% during stratification. Moisture content adjustment should occur if the moisture content is less than 34% or greater than 40%.

3. All **Pw sowing requests should be monitored and 'handling' during stratification every Monday, Wednesday and Friday**. This involves a visual inspection for fungal growth and moisture status (excessively wet or dry), and handling of the seed within the bag to break up any fungal colonies and redistribute moisture within the stratification unit.

4. **The TSC should volunteer to extend stratification to our clients up to a maximum of 120 days**. All OSP results indicate that increased stratification will increase GC and 120 days is the standard duration used in the US to stratify Pw. The large amount of viable seeds following standard stratification suggests that the duration may not be long enough. All nurseries growing Pw have agreed to this extension, but it may not always be possible due to the late entry of sowing requests.

5. To improve the reliability of germination estimates for Pw **there should be at least two germination tests performed on a seedlot prior to use for sowing**. This will be impossible for new seedlots, but these seedlots will be retested again prior to the next sowing season. A thorough review of retesting frequencies will occur in the winter of 2002 and will likely result in more frequent Pw testing

6. **All returned select white pine sowing requests should be tested on arrival** at the Tree Seed Centre. This allows for an evaluation of seed quality to determine the fate of the seed (destroy ***vs.*** dryback, store and retest in 6 months). It can also provide valuable information on whether dormancy is re-introduced following drying and storage at -18°C.

7. **Increase the extension and communication** of issues and findings related to operational stratification of western white pine.

This is an abbreviated version of a full ten page report that can be obtained by contacting me directly. Please forward comments to Dave Kolotelo [Dave.Kolotelo@gems7.gov.bc.ca] or call me at (604) 541-1683 extension 228. Thank you.

David Kolotelo, RPF
Cone and Seed Improvement Officer
Dave.Kolotelo@gems7.gov.bc.ca
(604) 541-1683 extension 228