CHAIR'S 'ARMCHAIR' REPORT

Changes are happening at the News Bulletin. I'd like to thank Dr. Ron Smith for his dedication and service as editor of the News Bulletin. Ron has been editor since issue #24 (January 1996), was a consistent contributor long before that, and has helped in organizing several of our workshops. I extend a warm thank you to Ron for all of his efforts for the Tree Seed Working Group.

Taking over as editor, Dale Simpson is no stranger to the working group. Dale manages the National Tree Seed Centre in Fredericton and has played an active role in the Tree Seed Working Group and CTIA activities. Dale also serves as our OECD representative for tree seeds in Canada. Welcome on board Dale!

Dale and I are planning to have the next News Bulletin (#37) distributed electronically. It will be extremely important that we have a complete list of all e-mail contacts. Please forward your e-mail address to Dale at dsimpson@nrcan.gc.ca. If we do not receive your e-mail then you will not receive a copy of the News Bulletin. The theme for the next News Bulletin is "Hardwoods" and I know many people are enthusiastic about the work being performed in this area. Dale and I look forward to your contributions.

The theme of this News Bulletin is "Quality Assurance Monitoring" and although many have an idea what this is, I hope some of the contributions enlighten you on what quality assurance (QA) can be and what it can do for you. I'll introduce the subject here, but read the articles and associated references for a more complete (and practical) picture.

Within the scope of our Working Group's mandate, quality assurance should begin with the monitoring of our seed or pollen crops to determine if harvesting is viable. Pre-collection evaluations focus on predicting yield, identifying pest problems, and determining the appropriate harvest time. These principles are equally applicable to wild stand and orchard crops, although the intensity is usually greater with the latter. Most formal quality assurance or certification schemes begin with ensuring that source or genotype is correctly identified. This allows transfer limits to be placed on the seed based on provenance and other test information. This step is so crucial in forestry - ensuring that the propagules we place on the landscape are adapted to that site for their life-span. These issues are generally handled through legislation (News Bulletin #35) or internationally by certification through the Organization for Economic Cooperation and Development (OECD). The issue of climate change is certainly starting to move up the priority list with the issue of adaptation (and to what?)
It is difficult today to think of QA and not have the subject of certification come to mind. Companies are having their management systems, products, and forest-certified. Certification initiatives are generally focused on either management practices or products. Some good links for those interested are the Forest Certification Watch™ (http://www.sfcw.org) and the Canadian Sustainable Forest Certification Coalition (http://www.sfsc.org).

Most of us who are processing cones and seeds, testing seeds or producing seedlings from seed are looking for more specific information. For agricultural seed the Canadian Seed Institute (http://www.csi-ics.com) is a good link and they also produce a Canadian Seed Quality News newsletter. This is a valuable source of information and good model, but very little of the information is of practical value for forest tree seeds.

In tree seeds, there is a practical demand for uniformity or conformance in attributes such as seed size, germination capacity, purity, and moisture content, especially for container seedling production. Similar to manufacturing, the conformance of these attributes to some known estimates increases the efficiency of operations and ultimately the profit margin. With increases in seed costs, the consumer expectations of seed performance becomes even greater. In tree seeds we have a particular quandary – we are trying to minimize variation in a highly variable crop.

How do we minimize the expression of variation in seed characteristics and still retain the inherent genetic variability? This is firstly a question of how we can accomplish this and secondly if we are minimizing variation (through some type of selection), do we understand the consequences. Selection is not something we should be afraid of. Selection is a part of our business from the natural selection of pollen that will successfully fertilize an ovule, through the seed we select to compose a seed lot, to the seed we sample (select) to test, and eventually to the selection of seedlings that will be planted on our landscape. It is a matter of understanding the key bottlenecks and hopefully being able to quantify what impact they have on the genetic diversity of our crops. Gene conservation can be thought of as the quality assurance of diversity!

In most processing and manufacturing enterprises (that were responsible for the development of current QA theory) significant variation from what is expected makes goods and services undesirable. Determining the amount of variation and how this may change during processing is a large part of the QA development. Many current QA programs are more focused on processes than product. The theory is that this allows one to pinpoint ‘problem phases’ and if all the processes work effectively, the resulting product will be good (Vareed and Jobe 1999 – see Selected References – Quality Assurance).

I’ll shift to one of the other QA bibles (Summers 1999) to expand in a different direction. In any production system the amount of variation can be attributed to controlled variation (due to the process itself) and secondly uncontrolled variation (caused by factors external to the process). An example may help – if we are performing a germination test and do not control the amount of water we add or the temperature we maintain the seeds at we are adding to the variability of the process (controlled variation). An example of uncontrolled variation is the inherent genetic variability in the seed lot we are testing. The whole process of documentation, in QA or certification, addresses the need to identify and control, within limits, the variation due to the process itself.

Does current QA methodology focused on the manufacturing sector have a role in tree seeds? My contention is that they offer many useful tools to examine and track products and processes, but these do not place a value on diversity. This is even true of our seed industries in agriculture and floriculture. Quality assurance will become a larger part of your life if you wish to be competitive and accountable to your clients (I truly believe this). You will need a system that serves the needs of your clients. Instituting an off-the-shelf solution may not be the answer as it may erode what we determine to be our greatest value for the future – the genetic diversity of our forest trees.

Dave Kolotelo
TSWG Chairperson

EDITOR’S NOTES
Thanks Dave for those kind words of introduction. We are all indebted to Ron Smith for editing the News Bulletin for the last seven years. Ron picked up the ball from Hugh Schooley when he retired from the Petawawa National Forestry Institute. Ron’s dedication to this task is much appreciated and he leaves the News Bulletin as being a First Class publication. Thanks again Ron for your years of dedication and please feel free to contribute from time to time.

I plan to pick up where Ron has left off. Two issues of the News Bulletin will be published annually as this seems to be a reasonable number. Our Chairperson, Dave Kolotelo, continues to do an exceptional job at soliciting articles and even finds the time to submit some himself. I like the idea of having ‘theme issues’ and I hope you do too. As always, this is your News Bulletin so please consider submitting articles voluntarily or when asked to do so. What may not seem like ‘news’ to you is probably of interest to others.

Ron is continuing to try to resurrect our TREESEED Discussion Group. There have been a few glitches but hopefully it will be working properly soon.

Dale Simpson
Editor

TREE SEED WORKING GROUP

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PACIFIC REGENERATION TECHNOLOGIES INC.

Pacific Regeneration Technologies Inc. (PRT) provides quality nursery products and services for reforestation. PRT is comprised of 13 nurseries located across Canada in Ontario, Saskatchewan, Alberta, and British Columbia. In 2002, production consisted of 130,000 000 forest seedlings.

Quality control and risk management are an integral part of PRT’s production processes. Since most of the seedlings grow from seed, seed quality is of utmost importance. Determining or verifying the quality of seed for an order, prior to sowing, is essential for successful production. The germination capacity of the seed is used to determine the oversow and sowing rate (seeds per cavity) to be used. Accurate seed weight ensures that the proper amount of seed is ordered and prepared.

PRT Prince Albert, Saskatchewan has a fully equipped seed extractory capable of processing cones of all conifer species. The facility’s seed testing laboratory performs seed quality assurance testing on behalf of the entire company. Seed is sent to PRT Prince Albert to be tested from jurisdictions throughout North America.

Often, seed that is delivered from an extractory or storage facility will include seed quality information for that particular seed lot. This is very useful information, however, PRT’s internal quality control program calls for seed lots to be tested (or retested) prior to sowing so that the information is available for sowing. It is well known that seed lots stored for a long period lose some germination capacity, therefore, quality data for a seed lot tested a number of years ago may not be accurate. Germination capacity tests are carried out in climate-controlled growth chambers and normally involve the testing of both stratified and unstratified seed. PRT Prince Albert’s long-term seed storage facilities are fully alarmed with high and low temperature alarms as well as equipment failure alarms.

Seed upgrading is another service carried out at PRT Prince Albert, as well as other PRT nurseries. This process involves upgrading low germination capacity seed lots by systematically removing extraneous material and non-viable or empty seed, thereby increasing the total germination capacity of the seed lot. Both ‘air separation’ and ‘water density separation’ techniques are used in the upgrading process. PRT Campbell River specializes in upgrading very difficult and low germinating seed lots of Abies species.

Seed quality control also takes place at the sowing line. Quality control personnel remove sown blocks from the sowing line at random and count and record the seeds per cavity in that block. The block is then marked so it can be identified later and set out in the greenhouse where it is monitored and the germination results are recorded for the next several weeks. This provides a ‘field’ germination test, where overall germination and germination rate are determined. For each seed lot, several germination test blocks are set out in the greenhouse at staggered locations. The size of the seed lot determines how many test blocks are set out, however, even the smallest seed lots have at least one test block. These data are used for future reference when growing the same seed lot and allows for a comparison against the laboratory tests.

The importance of a solid quality control program cannot be emphasized enough where seed is concerned (or any facet of the forest nursery business for that matter). At PRT we believe that strict adherence to our quality control program and risk management plans has played a large part in the success of the company.

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NEW RETESTING FREQUENCIES AND DETERIORATION RATE ESTIMATES FROM THE BCMoF TREE SEED CENTRE

Seed lot retesting is considered to be a Quality Assurance (QA) function with the goal of providing accurate, up-to-date information on the germination capacity (GC) of a seed lot. The GC will guide seed requirements and how this seed is allocated into styroblocks (# seeds/cavity) to produce at least one germinant per cavity. The seed testing conditions are ‘considered’ optimum, but operational nursery results may differ. It is my opinion that lab test results cannot always accurately predict the germination of seed under the variety of growing conditions that it may be exposed to. Proactive nurseries should track germination (through their own QA programs) and have a good handle on how lab test results relate to germination under their specific germination environment. The GC of a seed lot will change over time as seed deteriorates in long-term storage. An estimate of a species deterioration rate aids in the decision of how often seed lots should be retested as part of our stewardship function.

This analysis of the germination history of seed lots in storage at the BCMoF Tree Seed Centre (TSC) was performed to:
1) provide individual seed lot linear deterioration rate estimates,
2) provide an average estimate of a species’ linear deterioration rate in storage, and
3) identify seed lots that are deteriorating faster than the species average.
Methodology

To determine the linear deterioration rate of a seed lot only three pieces of information were required: a) initial germination capacity (GC), b) current GC, and c) the time between the two tests. The seed lot deterioration rate was calculated using the following formula:

\[
\text{Deterioration Rate} = \frac{(\text{GC}_{\text{current}} - \text{GC}_{\text{initial}})}{(\text{time interval})} = \Delta \text{GC} / \Delta \text{time}
\]

Results are presented as change in GC per year and species averages were obtained by averaging all seed lots of that species. The standard error of the deterioration rate estimate is also presented to provide a measure of precision. The only conditions placed on seed lots were that they had more than one germination test (of the same type) and were in storage at least 500 days. Deterioration rates calculated using shorter intervals are likely to give high estimates that are based more on sampling variation than deterioration. For example, a 5% difference in germination over 5 years is equivalent to a deterioration of 1% per year, but over 6 months this same GC difference inflates to an estimate of 10% per year.

Although seed lots may not deteriorate in a linear fashion through their lifespan, the construction of non-linear deterioration equations or pooled seed lot regression analysis is not practical with the available data. Some data limitations are that data points per seed lot, irregular testing intervals, and relatively few long-term results. The presented method provides a simple means of estimating and discussing deterioration and provides a biological basis for the recommended germination retesting frequencies. Calculation of seed lot deterioration rates also allows one to identify and test more frequently the seed lots that are deteriorating faster. The working assumption is that estimation of deterioration rates using a linear procedure will not produce significantly different species deterioration rankings than a true average of the non-linear deterioration rate (i.e., Thuja plicata is going to be a priority no matter what the exact deterioration rate is).

Results

It is TSC policy that the germination retesting frequencies will be examined at 5-year intervals with the last review occurring in 1997. The 1997 and 2002 species deterioration estimates are presented in Table 1. Sample sizes for most species have increased considerably. The exceptions are the Abies spp. In 1997 the total sample size was based on several germination test types. The 2002 estimates, for all species, are based solely on one germination test type. For the 2002 analysis the average seed lot age (SAGE), maximum seed lot age (MAGE), and the maximum amount of time used in the calculations of deterioration rate (DAE) are included in Table 1. The DAGE is less than the MAGE when a change in germination test type has occurred during a seed lot's lifetime. This affects the results of Abies amabilis, A. grandis, A. lasiocarpa, Pinus monticola and P. ponderosa.

Some species also have positive estimated rates of deterioration and these are considered to be artifacts of the process and more likely related to sampling variation, lack of appreciable deterioration, and improvements in seed testing rather than increases in GC during storage. The use of only one test type per species, a decreased number of species with positive deterioration rate estimates, and an increase in seed lot information, both in numbers of lots as well as in maximum seed lot age, increase my confidence in the 2002 results.

The deterioration rate played a large role in the recommended retesting frequency, but other factors such as the precision of this estimate, the sample size, degree of change in estimated deterioration rate since 1997, number of seed lots currently in storage, age of seed lots in sample, and the number of problem seed lots per species all influenced the frequency recommendations.

The largest change in estimated deterioration rate was with Chamaecyparis nootkatensis. This species had a 23X increase in the sample size, but it still had the smallest sample size, largest standard error, and youngest seed lots of any species included here. The retesting frequency of this species has therefore been doubled from testing every 36 months to every 18 months.

Abies amabilis and A. lasiocarpa have several unique characteristics that deserve discussion. Standard test types have changed more frequently for these species as research into an optimum dormancy breaking treatment has progressed. Therefore, long-term data with any one test type is limited in these species. Seed lots of these species, and especially A. lasiocarpa, sometimes display highly variable germination results between subsequent tests. This is reflected in the positive deterioration estimates and large standard errors associated with these estimates. This is problematic for assigning seed lot quality. These species also possess resin vesicles and the 'condition' of these will impact seed longevity. A more conservative retesting frequency has been assigned to Abies spp. compared to other species with similar deterioration rates. The other non-Abies species with resin vesicles are Thuja plicata, Tsuga mertensiana and Tsuga heterophylla which have relatively high estimated deterioration rates.

The other species showing a large change in deterioration estimates is Pinus monticola. In 1994 the standard pretreatment for this species was changed and treatment significantly increased the GC of this species. The data available using this test type were extremely limited in 1997. The 2002 estimate is substantially reduced, but there is a large standard error associated with this estimate. A more conservative retesting frequency is recommended for this species compared to its low estimated deterioration rate. The remaining species have reasonably low deterioration rate estimates and standard errors.

Further Considerations

It has been suggested that the species datasets should be broken down into categories based on seed lot age or based on initial seed quality. To investigate the data distribution of initial seed lot quality and seed lot age, these factors are plotted against the seed lot’s estimated linear deterioration rate for Pseudotsuga menziesii var. glauca (Figures 1 and 2). These plots appear fairly typical for the species.

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1 Deterioration rate estimates are available for other species, but sample sizes are quite small and they have therefore not been presented. Contact the author for additional information on a BC tree species that is not presented.
In Figure 1, the data illustrate that most seed lots are initially of high quality, but the deterioration rate does not increase with decreasing initial seed quality. In Figure 2, the range of deterioration estimates is greater for young seed lots and appears to decrease with seed lot age. These observations are initially counterintuitive – let’s look at some possible reasons. The dataset is from an operational facility – it does not represent the results of a controlled experiment. Individual seed lots will differ in basic attributes such as moisture content, seed size, level of dormancy etc. Testing frequencies have fluctuated over the 40-year time period we have data on for some species and for other species the retesting history with one test type is minimal.

Table 1. The 1997 and 2002 estimates of species deterioration rates (DET) in germination (%/year), standard errors (S.E.) for 2002 estimates, sample sizes (#), recommended retest frequencies in months, average seed lot age (SAge), maximum seed lot age (MAge), and maximum time used in deterioration rate calculations (DAge) in years.

<table>
<thead>
<tr>
<th>Species</th>
<th>1997 DET %/year</th>
<th>#</th>
<th>Retest months</th>
<th>2002 DET %/year</th>
<th>#</th>
<th>S.E.</th>
<th>Retest months</th>
<th>SAge years</th>
<th>MAge years</th>
<th>DAge years</th>
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<tr>
<td>Abies amabilis</td>
<td>-0.78</td>
<td>254</td>
<td></td>
<td>0.06</td>
<td>165</td>
<td>0.15</td>
<td>26</td>
<td>7.8</td>
<td>24.6</td>
<td>8.9</td>
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<td></td>
<td>-0.72</td>
<td>49</td>
<td>0.22</td>
<td>22</td>
<td>11.3</td>
<td>22.4</td>
<td>12.5</td>
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<td>Abies lasiocarpa</td>
<td>0.67</td>
<td>150</td>
<td></td>
<td>0.17</td>
<td>107</td>
<td>0.54</td>
<td>20</td>
<td>8.9</td>
<td>26.7</td>
<td>4.9</td>
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<tr>
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<td>15</td>
<td>0.46</td>
<td>-2.16</td>
<td>35</td>
<td>0.79</td>
<td>18</td>
<td>4.6</td>
<td>10.2</td>
<td>10.2</td>
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<td>95</td>
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<td>-0.67</td>
<td>173</td>
<td>0.07</td>
<td>24</td>
<td>7.8</td>
<td>25.2</td>
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<td>-0.22</td>
<td>1233</td>
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<td>13.3</td>
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<tr>
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<td>-0.15</td>
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<td>349</td>
<td>0.03</td>
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<td>41.8</td>
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<td>609</td>
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<tr>
<td>Tsuga heterophylla</td>
<td></td>
<td>272</td>
<td>-1.22</td>
<td>-11.3</td>
<td>366</td>
<td>0.07</td>
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<td>12.1</td>
<td>34.1</td>
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<tr>
<td>Tsuga mertensiana</td>
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<td>33</td>
<td>-0.36</td>
<td>0.46</td>
<td>47</td>
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</table>

3 In British Columbia, Picea glauca, P. engelmannii and hybrids between these species are not differentiated and commonly referred to as “interior spruce.”

If a low GC is caused mainly by the presence of non-viable seed, then although the GC may be low it doesn’t necessarily mean that the remaining viable seed will deteriorate faster than average. The presence of non-viable seed is generally not significant for our main reforestation species today, but it can be substantial in Abies spp., Chamaecyparis nootkatensis or older seed lots. Are these non-viable seeds empty or deteriorated? And what is the condition of the viable seed? These questions are relevant to seed lot storability, but are not adequately addressed by the GC estimate.

There are much more data available on young (<10 years old) seed lots with generally high GC values. The reasons are of a practical nature as seed lots are consumed to grow seedlings and most do not remain in storage for long lengths of time. For tree seeds, the quality of all steps throughout the seed handling process has dramatically changed for the better in the past 40 years. The deterioration rates of our oldest seed lots are therefore probably greater than current collections.

Initial GC and seed lot age may not be good general predictors of deterioration. Other factors such as timing of collection, post-collection handling, cone and seed processing techniques, and seed moisture content may have a large impact on the deterioration rate. The long-term effects of these factors may not be evident in the initial GC estimate. It has been suggested that since deterioration is generally slow it would be better to
investigate speed of germination that is generally thought to decrease prior to seed death and the resulting drop in GC. This suggestion has merit, but unfortunately germination rate data are only readily available for the past 10 years.

The deterioration estimates are generally quite small on a species basis, but there is a wide range of variability between seed lots. To investigate the proportion of variation attributable to species a random effects model using PROC VARCOMP was used in SAS to determine the proportion of variance that can be accounted for by species. This analysis showed that only 6% of the variability could be accounted for by species with the remaining variability being accounted for within-species or at the individual seed lot level. This emphasizes the point that although it may be operationally useful to look at species differences the greatest amount of variability resides at the individual seed lot level.

![Figure 1](image1.jpg)

**Figure 1.** The relationship between initial germination capacity (GC) and estimated deterioration rate for *Pseudotsuga menziesii* var. *glauca* (n=609).

![Figure 2](image2.jpg)

**Figure 2.** The relationship between seed lot age and estimated deterioration rate for *Pseudotsuga menziesii* var. *glauca* (n=609).
Discussion

The method used for quantifying deterioration in this article has several advantages including providing a simple, easily understood procedure for calculating and discussing seed lot deterioration rates. The method also allows one to identify seed lots that deteriorate significantly faster than the species average enabling one to test these seed lots more frequently and maintain the same level of confidence in the results. Species average deterioration rates and their precision play a key role in determining the retesting frequency, but other factors such as sample size, degree of change in estimated deterioration rate since 1997, number of seed lots currently in storage, age of seed lots, and the number of problem seed lots per species all influenced the frequencies. There is no mathematical formula for this.

The main disadvantage of this method is that it is a linear estimate of the deterioration rate, but we expect that deterioration over time, measured in terms of GC loss, is not linear. The number of tests available on each seed lot is a major obstacle in employing more sophisticated analytical techniques to the data. The expectation is that GC will be relatively stable or decrease linearly until the majority of seeds fail to germinate and a larger decrease in GC will occur. At what age and how quickly individual seed lots enter this rapidly deteriorating phase is not known. Some seed lots (even some species) appear to have a stable GC over a fairly long timespan (decades vs. years).

Species retesting frequencies are used to identify pending retests for a species. The actual decision to perform a retest depends on other factors such as seed lot size, seed lot usage, and genetic class. The retesting frequencies act as a filter, but not a determinant of when retesting of an individual seed lot should be performed. The results clearly show that most species store very well, but the amount of variability within a species dictates that decisions should be made on an individual seed lot basis. As deterioration is generally low with consigns it is at least equally important to have earlier identification of the exceptions (seed lots with high deterioration rates) as it is to predict average seed lot performance. What to measure and how to efficiently integrate this result within a scheduling tool is a current challenge.

The revised deterioration rates and recommended retest frequencies will allow lab staff to focus on retesting rapidly deteriorating species (priorities) and individual seed lots deteriorating faster than average. The more rapid retesting of species that do not store as well provides an accurate estimate of germination and also adds more datapoints to the database to improve the estimate of deterioration in future reviews. These methods are not perfect (and neither is the database) and our quantification of deterioration may change in the future. Maybe feedback on this article will spur that change?

The seed lot and species deterioration estimates are currently our best biological estimates of change in germination capacity in long-term storage. I don’t believe that we will suddenly discover a method of quantifying deterioration at the species level that is better than at the individual seed lot level. There are too many unquantified features of collection, processing, handling, and year-to-year variability to accurately predict the relatively small change in GC over time on a species basis. It is time to recognize the unique characteristics of the individual seed lot. The key to the future of retesting lies in using the information on how an individual seed lot performs to guide how often it should be retested. The species average provides a benchmark for the individual seed lot.

Comments are graciously welcomed.

Dave Kolotelo

OXIDATIVE STRESS, ANTIOXIDANTS AND PROSPECTS FOR SEED CHEMOTHERAPY

Introduction

“It would appear that life originated as a result of free radical reactions, selected free radical reactions and assured ‘evolution’ by employing them for mutation and death” - (D. Harman 1986)

The accumulation of free radicals presents a distinct hazard to all organisms (Miquel 1989). Plants are particularly sensitive to oxidative stress because they cannot move to escape large doses of radiation. Free radicals are highly reactive compounds that are produced in the chloroplasts of leaves. They are formed when an electron, excited by light, is transferred to molecular oxygen, rather than its normal receptor. Visible symptoms can be observed in organisms when detoxification is inadequate. These symptoms develop in diseased, aged, and stressed trees due to the accumulation of singlet oxygen, hydroxyl, and other free radicals.

Reactive forms of oxygen are generated by exposure to air pollution, UV light, pathogens, and herbicides. Free radicals, e.g., superoxide dismutase, catalase, and glutathione reductase, interact with membrane components of stressed plants, causing them to ‘leak’. It is the loss of essential metabolites from cells that slows growth and initiates senescence.

Antioxidant Defenses in Plants

Antioxidants combat free radical reactions that cause mutation and death. Since organisms cannot store large amounts of antioxidants, a daily intake can increase mean life span (Harman 1978) and maximize longevity (Miquel 1989).

Plants can mobilize an impressive array of defences, depending on the type of stress. If defence compounds can be extracted and then reapplied, they could provide a cheaper, safer, and easier method of alleviating stress, as compared to chemical fertilizers, herbicides, fungicides, or insecticides. As medicinals, antioxidants have been used for centuries in food and medicine. Commercially, they have preserved food, cosmetics, and pharmaceuticals for decades. Most recently, natural defence compounds in trees have been mobilized to protect plants from heat, frost, drought, and other stress factors. The research involves the use of antioxidants – nature’s pharmacopoeia – to help counter the adverse effects of age, disease, and environmental stress.
Synthetic Antioxidants – Ambiol

Ambiol, (2 methyl-4-[dimethylaminomethyl]-5-hydroxybenzimidazole dihydrochloride), falls into one of 14 classes of synthetic antioxidant (Santrucek and Krepalka 1988). Seed treatment with the antioxidant Ambiol stimulated growth of a wide variety of crop species, including soybean (Glycine max) and canola (Brassica napus) (Darlington et al. 1996), carrots (Daucus carota var. sativus) (Rajasekaran and Blake 2002), jack pine (Pinus banksiana) (Rajasekaran and Blake 1999), and black spruce (Picea mariana) (Borsos-Matovina and Blake 2001). Ambiol increased the stress resistance of seedlings exposed to extreme temperatures (Borsos-Matovina and Blake 2001) and drought (Rajasekaran and Blake 2002). Ambiol treatment reduced genetic damage induced during the radiation leak at Chernobyl and increased radiation resistance of Scots pine (Pinus sylvestris) growing within a 30 km zone of the Chernobyl Atomic Energy Plant (Vishnevetskaya and Roy 1999).

We have tested Ambiol and found that it is a powerful antioxidant. Seed treatment with Ambiol prevented membrane leakage in experiments conducted on black spruce (Borsos-Matovina and Blake 2001), jack pine (Rajasekaran and Blake 1999), white pine (Pinus strobus) (Islam et al. 2003) and carrots. In the latter case, Ambiol-treated carrot seedlings maintained membrane capacitance under drought, indicating they were better able to sustain the electrical properties of membranes (Rajasekaran and Blake 2002). Ambiol appears to donate a H+ to oxygen radicals, which would interrupt the free-radical chain reactions that occur during oxidative stress (Vishnevetskaya and Roy 1999).

Like other antiethylene agents, (aminovinyglycine, AVG, the polyamine, spermine), Ambiol reduced membrane leakage and inhibited stress-ethylene production (Rajasekaran and Blake 1999; Islam et al. 2003). Anti-stress, anti-ethylene and antioxidant properties best explained the membrane-sparing action of Ambiol (Rajasekaran and Blake 1999). Since it is a benzimidazole, Ambiol is also a non-purine cytokinin, which could also explain its ability to stimulate growth.

Natural Antioxidants and Seed Germination

Field, greenhouse, and laboratory studies have confirmed the growth-promoting effects of the natural antioxidant BioProtect. BioProtect has been used to harden seedlings on an operational scale which involved treating millions of seed with antioxidant. These studies were conducted in collaboration with: La Maison Verte, Hearst Forest Management, Irving, Agrium Inc., Cook Lake Nursery, Boreal Nursery, Hills Nursery, and LUSTR CO-OP. Field trials have been conducted near Hearst, Thunder Bay, Dryden, and in Nova Scotia. Currently, BioProtect is being tested in New Brunswick and British Columbia. These studies have confirmed that seed treatment enhanced the speed and totality of germination, seedling growth, and stress tolerance. Positive growth responses were first detected in the greenhouse and these carried over into the field where they were evident for at least several years.

Seed Deterioration in Storage

Although seeds remain viable for many years, their vitality declines during storage. There is an obvious genetic component to seed deterioration since more vigorous seed lots decline more slowly in storage. The seed environment is also important. Cold, dry storage enhances viability, while elevated storage temperatures and humidity result in slow, uneven germination. Seeds deteriorate despite the absence of microorganisms, any other unfavourable environmental factor, or morphological factor. Although the internal changes in aging seed are poorly understood, seed membranes obviously deteriorate. This is because of the loss of essential metabolites from seeds occurs during the decline in seed viability.

Seed decline in storage can be measured in terms of mean germination time, electrical conductivity of seed steep water, and by embryo staining, e.g., with tetrazolium chloride. Since these are costly and time-consuming, they represent a major expense for any seed company. Greenhouse operators prefer to use younger seed. This can result in a high-grading that diminishes the value of older, stored seed.

We considered the possibility that antioxidants could delay seed deterioration, or at least partly reverse some of its negative effects. If so, it might be possible to use antioxidants to enhance seed germination and growth as seeds ‘age’. Review of the literature revealed relatively few compounds that stimulate seed germination. These include strigol, ethylene, ethanol, 2-nonanone, octylthiocyanate, and several anaesthetics, including chloroform and ether. While these compounds affect plant membranes, their mode of action is unknown.

Hydration of seeds also accelerates seed emergence and may promote more uniform germination. Seed priming with hot water initiates earlier germination. Primed, pre-germinated seeds are used where there is low soil moisture. Although soaking, spraying, and dipping with water improved germination of some species (Basu and Mandal 1985; Basu 1990), they failed to enhance confer seed germination. The growth-enhancing effects of BioProtect occurred independently of cold, moist seed stratification.

Ambiol also accelerates seed germination, reduces the time required for emergence, and increases stress tolerance and plant viability. Treated canola seeds were twice as likely to have emerged 10 days after planting, compared to untreated seeds (Vishnevetskaya et al., 1986). At the most effective concentration (10 mg/L), Ambiol caused a four-fold acceleration of the germination rate of cucumber seed (Cucumis sativus) (Kritenko and Blake, unpublished data).

Although germination of 30-year-old jack pine seed remained relatively low, it was significantly enhanced by a 24-hour soak with BioProtect. A 24-hour soak with BioProtect also accelerated germination of young, fresh (2-year-old) white pine seed (Pinus strobus) (Blake 2002), as shown in Figure 1. In the study conducted by the Angus Seed Plant (A. Foley, pers. comm.), a 24-hour seed-soak with BioProtect enhanced: 1) overall germination rate, by 10% (78% vs. 68%), 2) germination energy, by 30%, and 3) germination period was reduced by 14 days (17 days vs. 31 days).

Conclusions

A progressive loss of metabolic constituents through seed membranes occurs while seeds deteriorate in storage. Seed aging is increased by hot, moist storage and can be, at least partly, reversed by antioxidant
Figure 1. Impact on germination of soaking white pine (Pinus strobus) and white spruce (Picea glauca) seed in Bioprotect.

treatment. For example, a single 24-hour seed-soak with Bioprotect enhanced the speed and totality of germination in 30-year-old jack pine seed. Peak germination also increased by up to 50% (Ocran and Blake, unpublished data). Several compounds, including Bioprotect, Ambiol and salicylic acid also accelerated the germination of young, fresh seed.

Free radical damage accelerates seed ageing and increases membrane permeability. Pre-treatment with anti-ethylene compounds (e.g., hydrobrominolide, spermine, salicylic acid, Ambiol) spares membranes in drought-stressed plants, as shown by the reduction in membrane leakage (Rajasekaran and Blake 1999; Islam et al. 2003). A single (24 h) seed treatment with anti-

oxidant promoted seed germination, increased plant hardiness, and accelerated seedling growth under laboratory and operational conditions for at least several years. Natural antioxidants, therefore, may offer distinct possibilities for both the seed and seedling industries. For further information on how natural therapeutic agents can enhance growth and stress tolerance of plants, the reader can refer to www.ambiolinc.com.

Literature Cited

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SEED QUALITY ASSURANCE AND INTEGRATED PEST MANAGEMENT

In the world of seed procurement and processing, the term quality assurance is a paradigm of the industry. In general terms, quality assurance can be defined as all actions taken to ensure that standards and procedures are adhered to and that delivered products or services meet performance requirements. In our attempts to produce the highest quality seed, many procedures are scrutinized and developed to ensure a high and repeatable level of success. Yet, the formula is not a simple one as there are organisms other than ourselves who readily desire this resource. In essence, insects and fungi are natural integral components of forest ecosystems but acquire the term ‘pest’ because of their impact on our utilisation and management of this fibre resource. These associations range from the general to very intricate depending on the seed species, environment, and the particular pest. Insects and diseases may attack conifer reproductive structures at any time from cone bud initiation through to cone collection and seed extraction.

In agriculture and forestry, the process of mediating their effect on the final commodity comes under the banner of pest management. Within environmental and regulatory considerations, the process of dealing with a pest is to develop an integrated approach. Experience has shown us that reliance on any one method will result in a decline in the effectiveness of the control measure over time. The most obvious example is insect and disease resistance to pesticides. In British Columbia, the term integrated pest management (IPM) is defined as a decision making process that uses a combination of techniques to suppress pests and that must include, but is not limited to, the following elements:

1) planning and managing ecosystems to prevent organisms from becoming pests,
2) identifying potential pest problems,
3) monitoring populations of pests and beneficial organisms, pest damage, and environmental conditions,
4) using injury thresholds in making treatment decisions,
5) reducing pest populations to acceptable levels using strategies that may include a combination of biological, physical, cultural, mechanical, behavioural, and chemical controls, and
6) evaluating the effectiveness of treatments.

Many of the terms and objectives above are integral to seed quality assurance and lead directly into requirements defined by regulatory and certification programs.

All this looks good on paper but the actual process of definition, monitoring, evaluation, and management is not that easy. For one, the hosts, both the parent trees or seed are living biological end products of genetic and environmental interaction and their behaviour cannot be
predicted with absolute certainty. The very same goes for the plasticity of the pests (insects, pathogens, and weeds) as they attempt to maximise their biological footprint. The point and degree of economic impact varies with tree species, life cycle stage and environmental conditions.

In the acquisition and processing of conifer seed for reforestation needs, pest management strategies become an integral part of assuring the quality, quantity, and genetic conservation of any reforestation program. Cone and seed insect pests and pathogens need to be identified, monitored, and various strategies developed to minimize their impact.

- In natural stands, forest health monitoring may dictate cone collection timings and location. In seed orchards, the intensity of production demands strict observations and management alternatives to keep potential insect pests in check or help to identify windows of opportunity to escape the potential impact of a pest. Monitoring programs with pheromone traps may help to identify the phenological links between the host trees and the insect pests.

- Cone collection and post-collection procedures need an IPM component to minimise the impact of inoculating the cones with pathogenic fungi. The placement of tarpaulins on the ground helps to minimize cone contact with the forest dust and consequently reducing the chance of inoculation with _Caloscypha._

- At the cone and seed processing stage, all handling procedures must be closely monitored to reduce the possibility of bulking up pathogenic fungi through the sheer concentration effect of many seed species and sources flowing repeatedly through the same equipment. Regular sanitation procedures at key processing steps will help to reduce the degree of fungal inoculum.

- Seed testing is paramount to quickly identify cryptic insects and pathogens. X-ray radiographs help to identify the degree of seed insect infestation while RT-PCR determine seed-borne pathogen levels. Once identified, then alternative handling strategies or procedures can be initiated to minimize the impact on seed deployment.

- Long-term storage environments must be developed and monitored to exclude or short-circuit insect and/or fungi life cycles.

- Seed pre-treatment procedures can have a parallel IPM plan to ensure seed viability. For example, seed soaking using running water may help to reduce the risk of damping-off in seed lots identified with significant _Fusarium_ levels.

- Constant monitoring during stratification and shipment to identify potential fungal build-up problems. The use of a post-stratification sanitation treatment with hydrogen peroxide will help to reduce the potential risk of _Caloscypha_ spread or _Fusarium_ damping-off in the nursery environment.

In essence, all these procedures have merit but operational considerations will override the best-

engineered plans. At each stage of the process, each seed collector, processor, and user must balance the demand for the best possible seed with logistics. In real terms, the development of a quality assurance / IPM program will be contingent on the process being:

1) technically possible,
2) practically feasible,
3) economically desirable,
4) environmentally acceptable, and
5) politically advantageous.

The continual effort required to identify the intersection point for all these issues is the challenge in developing a quality assurance program. Pest management is definitely an integral part of that process. The practicality of this reminds me of a story about IPM at reforestation nurseries. Every year a field trip is held at a particular reforestation nursery for students enrolled in a post-graduate pest management program. As part of the day, we proceed to various stations around the nursery looking at different aspects of production. We ask the students to comment on what pest management strategies they think have been incorporated into the production process. Before the student's arrival, the nursery manager and I walk to the different stations and review the important points. At the end of the tour, the nursery manager is always amazed at how much of the day-to-day activities dedicated to producing the best possible seedlings are tied to pest management. As one Dutch IPM specialist stated in a lecture on the use of bio-control agents: "If you want...you can."

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**CYBER NEWS BULLETIN**

Dave mentioned in his 'armchair report' that this will be the last News Bulletin that will be printed and mailed. I am sure most of you receive electronic copies of other newsletters and this is an efficient means of distribution. However, in order for this to happen, I need your e-mail address. Even if you know that you have provided your e-mail address before, please re-send it just to be sure. If I do not receive it then I will assume you no longer wish to receive the News Bulletin. There may be some you who do not have access to e-mail. You can still receive a copy of the News Bulletin by post but you will have to inform me that this is your choice. You can contact me at: Dale Simpson, Natural Resources Canada, Canadian Forest Service, P.O. Box 4000, Fredericton, NB, E3B 5P7 or dsimpson@nrcan.gc.ca.
NATIONAL TREE SEED CENTRE

There was a good fruit and seed crop on most all species in the Maritimes this year. The spring weather was cool and moist which resulted in collection being delayed by up to two weeks. This two week delay tended to be maintained through the autumn. Nevertheless, 350 collections were made from 32 species. Most of the effort was made in collecting seed and fruit from single trees. Staff cooperated in making single-tree hemlock (Tsuga canadensis) collections as part of a research project evaluating genetic variation and the mating system in old-growth hemlock populations. The cone harvest portion of the study will continue over a period of years when there are good cone crops in order to collect from all the study populations. There was a heavy seed crop on black ash (Fraxinus nigra) for the first time in at least six years. Single-tree collections were made from three populations. The winter months will be spent processing, extracting, and testing all this seed.

The Seed Centre’s web site became operational the end of August. The number of visits is increasing each month. You can check out the Seed Centre at www.atl.cfs.nrcan.gc.ca.

Developmental research on germination testing has focused on hardwood species. An undergraduate student is conducting a Baccalaureate thesis on beech (Fagus grandifolia). He is evaluating three cold/moist stratification times and three germination temperature regimes. Another student is comparing excised embryo and tetrazolium tests with regular germination tests for American mountain ash (Sorbus americana) and showy mountain ash (S. decora). Other trials have included testing stratification times and germination temperatures on sugar maple (Acer saccharum) and investigating the impact of storing silver maple (A. saccharinum) seed just below 0°C. Results from some of these studies will be reported in the next News Bulletin which will have a “hardwood” theme.

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OECD SEED CERTIFICATION

Canada has participated in the “OECD Scheme for the Control of Forest Reproductive Material Moving in International Trade” since 1970. The tree seed Scheme along with others is administered by the Organization for Economic Cooperation and Development (OECD) headquartered in Paris. The current Scheme was adopted by OECD in 1974. At that time the Government of Canada appointed the Canadian Forest Service as the Designated Authority to implement the Scheme. Since then, certification work has been conducted almost exclusively in British Columbia because of the demand for seed from tree species located there. The Scheme is a means to promote and facilitate international trade and movement of forest tree seed, plants, and propagules. Participation in the Scheme is voluntary. Certified seed carries with it a Certificate guaranteeing the category and provenance and implicit that all rules and regulations were followed in the collection and processing of the seed. Certification does not cover seed quality, i.e. germination. It is the dealer’s responsibility to have seed lots tested for purity, moisture content, and germination.

The Scheme contains four categories of reproductive material: Source Identified, Selected, Untested Seed Orchards, and Tested. All OECD certified seed shipped from Canada has been in the Source Identified category. Guidelines have been written for registration of untested seed orchards but no seed has been certified under this category.

A new Scheme has been written to take into account progress being made in vegetative propagation and new techniques/types of seed plantations and seed orchards. Four categories of forest reproductive material are recognized: Source Identified, Selected, Qualified, and Tested. Under the Tested category, genetically modified propagules were included which unfortunately has resulted in a delay in the unanimous approval of the new Scheme by member countries. At the biannual meeting of member countries held in Paris in early October it was decided that an “Expert Committee” be formed to re-write those portions of the Scheme that refer to genetically modified organisms. It is imperative that the new Scheme be approved as soon as possible because a number of European countries have modified their forestry legislation based on it, the European Union’s Directive (developed in parallel) comes into force 1 January 2003, and as a means of promoting international trade.

For further information on the new Scheme, refer to Nanson (2001) in the Recent Publications section or visit www.oecd.org for further details on the current Scheme.

Dale Simpson

SEED PHYSIOLOGY AND TECHNOLOGY SYMPOSIUM

The International Union of Forestry Research Organizations (IUFRO) Research Group “Seed Physiology and Technology” (RG 2.09.00) held its latest Symposium during September 11-15, 2002, in Chania (pronounced “Hanya”) on the island of Crete. Co-organized by the University of Athens, under the very capable direction of Prof. Costas A. Thanos and his committee, the Mediterranean Agronomic Institute of Chania (MAICh), and the Hellenic Ministry of Agriculture, the symposium was sponsored by the International Society for Seed Science (ISSS). Seventy-one (71) participants from 23 countries made 53 contributions. The latter were collected in a “Programme and Book of Proceedings” presented to each participant upon registration. This is a first for RG 2.09.00: to have the Proceedings available at the start of the symposium. Congratulations to our Greek hosts for this achievement, one that future organizers would do well to emulate!
Contents of the Book of Proceedings include 32 full-text presentations, plus 21 in abstract form, accompanied by a list of some 30 posters. Canadian contributions came from T. Beadmore (with K. Forbes and J. MacDonald) (CFS Fredericton) – none of whom were able to attend; A. Mossier (with J.E. Major and O.P. Rajora (also CFS Fredericton); B.I. Daigle and J.D. Simpson (also CFS Fredericton, but also unable to attend); Y.A. El-Kassaby (UBC, Vancouver); and D.G. Edwards (FTB Forest Tree Beginnings, Victoria – formerly CFS Victoria). A former staffer at UVIC (Victoria), now returned to his native Ireland, C. O’Reilly also participated. A prominent forced-absentee was RG Chairman Jack Vezzo (USFS) whose funding was belatedly cut off to help toward the extreme fire-season experienced in the U.S.A. The presentations were of a uniformly high calibre, a testimony to the organizational activities of Prof. Thanos and his colleagues.

Three excursions/field trips were part of the program. One was an all-day (8-hour) hike down the National Park of Samaria Gorge in the Lefka Ori (White Mountains), the longest gorge in Europe. From the start, to the most-welcome restaurant (where dinner was waiting) at the beach-side community of Ormos Agios Roumelis, we covered 18 km, descending 1 km in the first 16 km. Lunch, courtesy of local Greek Forest Service staff, was laid on about mid-way, and we were introduced to the Cretan aperitif “raki”. Fantastic scenery, but the very short – and infrequent – stretches where the trail went uphill, were a blessed relief. A geologist’s paradise, the lower walls of the gorge show the tremendous land-folding forces of times long past; in the space of about 100 m the limestone strata change from horizontal to sheer vertical. The highlight of the hike was “The Gates” where the gorge is some 3 m (that is not a typo) wide, and the cliffs are over 300 m high. A two-hour ferry ride along the southern shore of Crete, past some idyllic villages many of which can only be approached by sea, brought us to Chora Sfakion where our bus waited to take us back to Chania, some fresh socks, and muscle relief.

The second, brief field trip took us to the small, but excellent Archaeological Museum of Chania, situated in what was the Venetian Franciscan monastery in the old city. While smaller than the museum of Heraklion (Iraklion), the collection and displays focus on finds from the western end of the island, and many of these are exquisite despite their 2000-4000-year antiquity. Finds from the Neolithic era, as well as Byzantine, Roman, Venetian, and other times are still being dug up all over Crete, especially in the Chania region.

A third half-day excursion took us first to the women’s monastery of Chrissopigi on the outskirts of Chania. A walled enclosure, the grounds are a gardener’s dream. From there to the Chania nursery where – among many other plants – were some 4 000, 1.5 m high Cupressus sempervirens shrubs destined to grace the 2003 Olympic Games in Athens. Then to the bay, and one of the many gorgeous beaches, at Agios Onoulfrios, the village of which was the site for the film “Zorba the Greek”.

The last session of the symposium was an RG business meeting at which it was announced that the next symposium will be held in Athens, Georgia, August 2003, sponsored by the USFS. Also, two proposals were presented by D.G. Edwards, one to revive the World Directory of Tree Seed Workers (in an electronic format), and second a Global Bibliography of Tree Seeds.

Immediately following, the International Plant Genetic Resources Institute and the Danish International Development Agency (IPGRI/DANIDA) opened a four-day final workshop on “Effective conservation and use of intermediate and recalcitrant tropical forest tree seeds”. The first session of this meeting was given over to a pre-planned business meeting of the International Tree Seed Testing Association’s (ISTA) Forest Tree and Shrub Seed Committee (FTSSC), of which 16 members had attended the symposium. Many IUFRO members also participated, as did several participants for the IPGRI/DANIDA workshop.

Dr. Zdenka Prochazkova (Czech Republic, FTSSC Chair) and Dr. Hugh Pritchard (UK, Co-Chair) led the half-day ISTA FTSSC meeting held immediately following the closing of the symposium. The agenda covered: 1) Committee Member reports: Hugh Pritchard spoke to germination of tropical palm seeds, Gary Johnson (USA) spoke to paired germination tests and the need to seek clarification from ISTA whether these are to be discontinued, George Edwards spoke to revisions of pure seeds definitions governing gymnosperm seeds. 2) Referee samples and ISTA proficiency tests, and the protocol for sample preparation which needs modification for forest tree seeds. 3) Harmonization of test methods between ISTA and the Association of Official Seed Analysts of North America (AOSA), especially regarding purity sample weights, and germination conditions. 4) The autumn 2003 FTSSC Workshop in Prague. 5) Update of the 1991 Tree and Shrub Seed Handbook, compilation of a multilingual glossary of tree names, and a second visit to the IUFRO proposal for a Global Bibliography of Tree seeds.

While the IPGRI/DANIDA meeting was billed as the “final workshop”, in reality it was the occasion on which participants presented their findings to date, so that the next phase of the project could be more effectively planned. Approximately two dozen participants attended and this provided a rare opportunity for north temperate tree seed workers to get a glimpse of the problems confronting their colleagues working with subtropical and tropical tree seeds. Dr. E. Dullio (IPGRI) and Dr. D. Joeker (DANIDA) were the main individuals behind this meeting.

As a symposium/meeting/workshop venue, Chania will be hard to beat. The weather was in the high 20’s to mid 30’s (Celsius) the entire time, it rained heavily once during a session – so hard that speech was almost drowned out, and it sprinkled (conveniently) two or three nights during the wee hours. The local Cretans were very friendly, the food and beverages (hic!) were excellent, and the scenery, especially Chania harbour and the old city, a place to revisit. Of course, no visit to Crete would be complete without going to Knossos (a two-hour bus ride to the capital, Heraklion, then a 15 minute bus to the archaeological site), which is the 4000-year-old centre of the Minoan civilization. So much for a poor tourist to do!

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Editors Note: You can visit the web site www.cc.iao.gr/biolog/bb/Seeds2003.html/ to view pictures of the symposium and to download the proceedings.
GERMINATION TESTS: HOW PRECISE ARE THEY?

How many people have thought of how precise the germination capacity (GC) estimate of a seed lot is? If you determine that a seed lot has a GC of 85% and immediately repeat the test, how close do you think you should be? Many decisions are made based upon a seed lots’ GC without any regard to the precision or variability in that estimate. To be perfectly clear on precision (some call it efficiency), I mean how consistent or how much variation is present in repeated measurements. In seed testing, we generally use multiple replications (usually 4 replications of 100 seeds) and average the results of the 4 replications to obtain a more precise estimate of the GC.

The standard deviation conveys the degree of variation in GC among the replicates. If we wish to provide an estimate of the precision of an overall mean (of the 4 replications) the standard error is appropriate (Zar 1974). Most basic statistics textbooks can provide more details on these statistics. The sample standard error of a mean is estimated as the standard deviation of the four replications divided by the square root of the sample size. Another way to think of the standard error is the variation in the estimate (i.e., overall mean GC) after repeating the same sampling process and point estimation over and over again.

This article will focus on the precision of germination tests, but more importantly open up discussion of what would be the most useful format for its presentation? The intent is to be able to allow for some quantification of the precision of a GC value on an individual seed lot basis. In Table 1, the results of five Tsuga heterophylla germination tests (not extremes) are presented to illustrate germination test precision. The four replicates are averaged to produce the GC and the standard error is calculated as the standard deviation divided by 2 (square root of 4). Knowing the standard error and the t-value (3.182 in all our examples here with 4 replicates), the 95% confidence intervals are calculated. These indicate the range of values between which we are 95% confident the true population mean falls based on sample data provided.

Seed lot A and B both have a GC of 84.8%, but much greater variation is present in seed lot B with a much wider (less precise) confidence interval. One may think that replicate 2 is abnormal and should be removed to improve the precision of the estimate. The International Seed Testing Association (ISTA 1999) does have tolerances for the maximum tolerated range between replicates, but the replicates in this example all fall within that range. We therefore have a less precise estimate of GC in seed lot B. Is there a practical need to quantify this?

In seed lot C the GC is 95.2% and a nursery may single-sow this seed in containers. If the germination is actually 92% will this compromise the crop? Seed lot D is a fairly poor seed lot with a confidence interval that spans over 20 percentage points. How do we integrate this large variability into our sowing decisions? Probability models have generally been used to determine sowing requirements, but none, to my knowledge, include the GC precision as an input variable.

There is a large range in the observed precision among the seed lot tests. Are there similar differences at the species level? Average statistics have been generated for each of the species with the most commonly used germination test (Table 2). The average mean and average standard error represent what would be expected after a single test of 4 replicates. This corresponds to the variation among all of the test means for each species.

Mean GC and mean standard error are the average of individual germination test results. The number of seed lots is provided as a reference to indicate the number of distinct genetic populations. In discussing species, the mean standard error can be used as a good reference point in comparing species as all germination tests contained four replicates. The two Pinus contorta varieties displayed the greatest precision and this comes as no surprise to those familiar with the species. What might be surprising is that the contorta variety with an average GC of 92% can be expected to have a true seed lot mean of between 88.2% and 95.9% with 95% confidence. At the other end of the spectrum one has Chamaecyparis nootkatensis with a GC of 36.8% and a confidence interval of between 29.9% and 43.7%. These values give some sense of species differences, but generally the individual seed lot will be the unit to investigate and quantify germination test precision.

Discussion

An investigation of germination test precision is something that should be addressed. I am too often amazed at how people assume that there is no error in the estimate of a seed lots’ GC. I am also amazed to hear that the same confidence would be placed in a Pinus contorta GC and an Abies spp. GC. I am not sure if a quantification of germination test precision is something useful to nurseries, but all sources of variability should be explored as we fine-tune our seedling production systems. There has also been international interest in this subject as ISTA begins to address the question of uncertainty of measurement in all tests.

I have advocated the use of the standard error as it indicates the precision of a germination test. The standard error can also be used to calculate confidence intervals for estimated mean GC values. The standard 95% probability has been used, although one may be content with 90% confidence that the GC will fall within a narrower range? Knowing the standard error,

<table>
<thead>
<tr>
<th>Seed lot</th>
<th>Rep 1</th>
<th>Rep 2</th>
<th>Rep 3</th>
<th>Rep 4</th>
<th>Mean GC</th>
<th>Standard Error</th>
<th>95% confidence interval</th>
</tr>
</thead>
<tbody>
<tr>
<td>A</td>
<td>84</td>
<td>84</td>
<td>88</td>
<td>83</td>
<td>84.8</td>
<td>1.11</td>
<td>82.4 - 87.2</td>
</tr>
<tr>
<td>B</td>
<td>87</td>
<td>77</td>
<td>88</td>
<td>87</td>
<td>84.8</td>
<td>2.59</td>
<td>76.6 - 93.0</td>
</tr>
<tr>
<td>C</td>
<td>98</td>
<td>96</td>
<td>94</td>
<td>93</td>
<td>95.2</td>
<td>1.11</td>
<td>91.7 - 98.9</td>
</tr>
<tr>
<td>D</td>
<td>79</td>
<td>73</td>
<td>64</td>
<td>77</td>
<td>73.2</td>
<td>3.33</td>
<td>62.6 - 83.8</td>
</tr>
<tr>
<td>E</td>
<td>90</td>
<td>93</td>
<td>88</td>
<td>86</td>
<td>89.2</td>
<td>1.49</td>
<td>84.5 - 93.9</td>
</tr>
</tbody>
</table>
### Table 2. Precision of germination tests by species including sample size, average estimated mean germination capacity (GC), average estimated standard error, and average 95% confidence limits

<table>
<thead>
<tr>
<th>Species</th>
<th>No. tests (no. seed lots)</th>
<th>Average mean GC</th>
<th>Average standard error</th>
<th>Average 95% confidence interval</th>
</tr>
</thead>
<tbody>
<tr>
<td>Abies amabilis</td>
<td>474 (236)</td>
<td>63.1</td>
<td>2.16</td>
<td>56.2 - 70.0</td>
</tr>
<tr>
<td>Abies grandis</td>
<td>137 (65)</td>
<td>66.7</td>
<td>2.05</td>
<td>60.2 - 73.2</td>
</tr>
<tr>
<td>Abies lasiocarpa</td>
<td>383 (188)</td>
<td>56.2</td>
<td>1.98</td>
<td>49.9 - 62.5</td>
</tr>
<tr>
<td>Abies procera</td>
<td>38 (20)</td>
<td>63.2</td>
<td>2.13</td>
<td>56.4 - 70.0</td>
</tr>
<tr>
<td>Chamaecyparis nootkatensis</td>
<td>138 (95)</td>
<td>36.8</td>
<td>2.17</td>
<td>29.9 - 43.7</td>
</tr>
<tr>
<td>Larix occidentalis</td>
<td>556 (201)</td>
<td>77.2</td>
<td>1.80</td>
<td>71.4 - 82.9</td>
</tr>
<tr>
<td>Picea glauca / engelmannii complex</td>
<td>2332 (1225)</td>
<td>82.0</td>
<td>1.76</td>
<td>76.4 - 87.7</td>
</tr>
<tr>
<td>Picea lutzii</td>
<td>124 (50)</td>
<td>84.6</td>
<td>1.67</td>
<td>79.3 - 89.9</td>
</tr>
<tr>
<td>Picea sitchensis</td>
<td>322 (197)</td>
<td>90.3</td>
<td>1.35</td>
<td>86.0 - 94.6</td>
</tr>
<tr>
<td>Pinus contorta var. contorta</td>
<td>120 (66)</td>
<td>90.8</td>
<td>1.34</td>
<td>86.5 - 95.0</td>
</tr>
<tr>
<td>Pinus contorta var. latifolia</td>
<td>3173 (1798)</td>
<td>92.0</td>
<td>1.20</td>
<td>88.2 - 95.9</td>
</tr>
<tr>
<td>Pinus monticola</td>
<td>251 (158)</td>
<td>82.4</td>
<td>1.79</td>
<td>76.7 - 88.1</td>
</tr>
<tr>
<td>Pinus ponderosa</td>
<td>437 (224)</td>
<td>86.4</td>
<td>1.52</td>
<td>81.6 - 91.3</td>
</tr>
<tr>
<td>Pseudotsuga menziesii var. menziesii</td>
<td>583 (372)</td>
<td>90.0</td>
<td>1.39</td>
<td>85.6 - 94.5</td>
</tr>
<tr>
<td>Pseudotsuga menziesii var. glauca</td>
<td>1091 (615)</td>
<td>87.7</td>
<td>1.48</td>
<td>83.0 - 92.5</td>
</tr>
<tr>
<td>Thuja plicata</td>
<td>1058 (400)</td>
<td>73.1</td>
<td>1.93</td>
<td>67.0 - 79.3</td>
</tr>
<tr>
<td>Tsuga heterophylla</td>
<td>931 (365)</td>
<td>78.3</td>
<td>1.81</td>
<td>72.6 - 84.1</td>
</tr>
<tr>
<td>Tsuga mertensiana</td>
<td>129 (50)</td>
<td>86.9</td>
<td>1.64</td>
<td>81.7 - 92.1</td>
</tr>
</tbody>
</table>

1 In British Columbia, Picea glauca, P. engelmannii and hybrids between these species are not differentiated and commonly referred to as “interior spruce”

One can calculate the confidence interval for any probability. If one wants a higher confidence in the GC estimate, the size of the confidence interval will increase. Is there another way to increase our confidence? The simple, not so cost efficient, solution is to simply increase the number of replicates in our sample. For highly valuable crops, with appreciable genetic variation, this may be a realistic solution.

This is my first attempt at looking at the precision of germination testing, but not my last. I would appreciate feedback on the validity and practicality of the methods examined. There are certainly some issues and questions that I will continue to investigate, but this is the current status. Some of these issues are the normality of the GC estimates within a species distribution. Do transformations improve or change the results? Are our newer germination tests showing greater precision? How do unstratified and stratified germination tests of the same seed lot compare in precision? Are seed orchard crops showing more precision or does the recombination that supposedly gives us more genetic variation also give us less precision in our germination tests? There are still many questions to be asked – stay tuned.

**INTERNATIONAL WORKSHOP ON SEED BIOLOGY**

The 7<sup>th</sup> International Workshop on Seed Biology, organized by the International Society for Science (ISSS), was held in Salamanca, Spain, May 12-16, 2002. About 300 people working on seed biology, from many countries, attended. The four days of conferences were divided into five sessions: seed development, seed germination and dormancy, desiccation and other stress tolerance and conservation, seed ecology, and seed biotechnology. The great majority of the conference was dedicated to fundamental research. Many presentations were on biochemical functions of seed. Analysis of receptors, proteins, enzymes, ribosomes, etc. were some of the major topics covered. We learned a lot on the function of seed, metabolism, and mechanisms and roles of hormones on dormancy and the breaking of dormancy. There were only a few presentations and posters directly related to forest tree seed. The proceedings are not available yet, but you can contact the Chairman for further information; M. Gregorio Nicolas at gnf@guu.usal.es

**References**


Dave Kolotelo

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E-mail: michele.bettez@mnr.gouv.qe.ca
WOODY PLANT SEED MANUAL

Publication of the first comprehensive handbook on the seeds of trees and shrubs produced by the USDA Forest Service, the Woody-Plant Seed Manual (USDA Misc. Publ. 654), was delayed by World War II until 1948 even though the manuscript was completed in 1941. This first manual included 420 taxa in 140 genera and was invaluable in theboom of tree planting that occurred in the 1950's and 60's. An updated and expanded manual, Seeds of Woody Plants in the United States (USDA Agric. Handbook, 450) was published in 1974. This volume contained data for approximately 800 taxa in 188 genera. Its popularity led to multiple printings, and portions of it were translated into several other languages.

The numerous advances in tree seed technology in the last quarter-century have now dictated the publication of a new revision, again named the Woody Plant Seed Manual. The revision process by a team of Forest Service scientists and cooperators from universities and other government agencies has now been underway for over 6 years. The book will contain almost 1,300 taxa in 230 genera, a considerable increase in the scope of the 1974 handbook. Because such a large undertaking requires so much time and effort by so many people, the finished product is still many months away from release. In order to make the new technology available as soon as possible, a decision was made to post completed portions of the book on this interim website. Here, genus chapters containing text and tables (along with some of the photos and drawings) are presented in preliminary html format. As editing of additional genus chapters is completed, they will appear here.

The above was copied verbatim from the web site. I urge you to visit http://wpsm.net. It is a work in progress and we are fortunate that it is so readily available.

Dale Simpson

UPCOMING MEETINGS

AOSA Annual Meeting
Seattle, Washington, USA
June 5-11, 2003
Contact: Nancy Hartshorn 509-225-2630

Western Forest Genetics Association
Whistler, BC
July 28-31, 2003
www.genetics.forestry.ubc.ca/wfga2003

IUFRO Research Group 2.09.00
Seed Physiology and Technology
Athens, Georgia, USA
August 11-14, 2003
Contact: Dr. Gary Johnson wjohnson03@fs.fed.us

XII World Forestry Congress
Québec City, Québec
September 21-28, 2003
www.wfc2003.org

Forest Nursery Association of British Columbia
Comox Valley, Vancouver Island
September 23-25, 2003
Contact: Siritol Paquet sven@telus.net

International Seed Testing Association
Forest Tree and Shrub Seed Committee, Workshop
Prague, Czech Republic
October 20-25, 2003
Contact: Zdenka Procházková
prochazkova@vulnhuh.cz
Topics include: purity, germination, tetrazolium, health testing, and referee tests.

If you know of a meeting that would interest the readership, please forward the information to the Chairperson or Editor.

SELECTED REFERENCES – QUALITY ASSURANCE

As I usually indicate, but feel compelled to again, this is not meant to be an exhaustive review of our theme. It is a sampling of some of the more interesting articles or texts on the subject from my filing cabinet.

General


Seed Certification


Seed Quality


Statistics


Dave Kolotebo

(RECENT PUBLICATIONS)


Schroder, T., R. Kehr, and A. Hutterman. 2002. First report of the seed-pathogen Geniculodendron pyriforme, the imperfect state of the ascomycete Caloscypha fulgens, on imported conifer seeds in Germany. For. Pathol. 32: 225-230.


