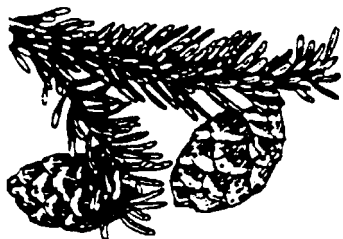

CANADIAN FOREST GENETICS ASSOCIATION
ASSOCIATION CANADIENNE DE GÉNÉTIQUE FORESTIÈRE



Tree Seed Working Group

NEWS BULLETIN

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THIS ISSUE AT A GLANCE

Page	Article
3	Tree Seed Workshop Overview
3	Seed Viability Prediction Options
4	The Practical Applications of Water Activity to Improve the Efficiency of Seed Management Systems
4	Using Water Activity in Tree Seed Banking and Its Implementation in Alberta
6	The Application of Water Activity to the Management of Pollen and Seeds in Quebec
8	The Well-Informed Collector: Tools for Equilibrium Relative Humidity Measurements in the Field
10	Impact of GA on Female Cone Production in Lodgepole Pine Orchards
13	National Tree Seed Centre
13	Upcoming Meetings
14	Recent Publications

CHAIR'S 'ARMCHAIR' REPORT

Hello and Happy holidays to everyone. It seems like it has been such a long year, so it is worthwhile reflecting on 2015. The most recent activities relate to completing the priority processing requests at our facility for upcoming sowing and seed sales for in-demand seed. Much of these priorities focussed on interior lodgepole pine and interior Douglas-fir seed from younger seed orchards. Crop volumes were low to moderate this year and insect damage was considerable for several seedlots. Notable problems were the redcedar cone midge (*Mayetiola thujae*) on the coast and the fir coneworm (*Dioryctria abietivorella*) in most Douglas-fir crops. Bumper natural stand cone crops were found for interior western redcedar and this helps to fill a declining inventory as we have no interior redcedar seed orchards.

It was a pleasure to attend the Canadian Forest Genetics Association conference in Fredericton and although somewhat small I was impressed by the quality of the speakers and tours. It was quite an amazing week and a bit, as I was able to visit four tree seed facilities (National Tree Seed Centre, Atlantic Forest Seed Centre, J.D. Irving Seed Plant, and the Quebec Tree Seed Centre) during my visit back East. It is always rewarding to see the dedication of people who work in our highly specialized niche and how they uniquely approach common activities. You always learn something – try to do that with a conference call!

The workshop theme “Development and Application of Water Activity for Monitoring Seed Moisture Content” focussed on a key issue that has been a hot topic in the News Bulletin as of late. It is unfortunate that all of our speakers were unable to attend in person, but special thanks to Bob Karrfalt and Fabienne Colas for their

presentations, connecting remotely, and dealing with technology limitations. Thank you to Lindsay Robb for discussing the topic from Alberta's perspective. I probably would not have been able to travel without presenting something – so I hope it was useful and/or interesting to some. I encourage you to contact the presenters directly if you want to discuss their presentations beyond what appears in their Abstracts.

We also added a “Cross Canada seed centre check-up” that included visual tours of their respective facilities. These were presented by Fabienne (Quebec), Al Foley (Ontario), Lindsay (Alberta) and myself for British Columbia. I thought that this was a good addition that provided another perspective on how different facilities approach the same task of seed extraction and purification in addition to providing an overview of other activities. I'm already looking forward to the next workshop in Alberta in 2017. Hope you can make it.

I think there was some convergence in our water activity target discussions, but I think we are still a ways off from an accepted universal target. I think that this does slow the rate of adoption of this relatively expensive technology, but cheaper alternatives (i.e., hygrometers) are available if you don't need a large daily throughput of results. It was also passed on to me that the ISTA Moisture Committee is looking to “*develop a new method to determine the equilibrium Relative Humidity (eRH) – i.e. water activity – in seeds. The project will involve different aspects: from general explanation of the method, to the elaboration of scientific results collected during the next two years from several labs. The scope is introducing a new chapter dealing with this topic in the ISTA Rules during the next triennium*”. That's great news, but hopefully they'll also not forget those who have already contributed to try to get the technology integrated in ISTA: <https://www.seedtest.org/upload/cms/user/ISTA-June18-0830-SympSession3-P4-Colas.pdf> <https://www.seedtest.org/upload/cms/user/ISTA-June11-Seminar-1330-McGill.pdf>

Water activity continues to play a key role in non-destructively ensuring that our genetic conservation samples are ready for freezer storage. We continue to chip away at our backlog. Trying to improve and/or better understand the role of kiln RH% and cone scale reflection was an active area of work this past fall with lodgepole pine. Not quite ready for prime time distribution, but quite happy to discuss our trial work and results with those who are interested.

I had also hoped to contribute an article on cheap solutions to damping-off in preparation for the upcoming sowing season. That didn't happen, but I will pass on the recipe that triggered that thought which came from an old (early Spring 2004) edition of our local Gardenwise magazine:

“Here's a great organic preventative (by damping-off), as well as an excellent foliar feed:

- ▶ 500 ml water
- ▶ small handful of chamomile leaves and flowers
- ▶ spray bottle

Boil together water and chamomile for 5 minutes. Cool, then strain. Pour into spray bottle and mist seedlings as they emerge. Repeat until second set of true leaves appears, or every 10 days”

I'm sure there are other home remedies that could be integrated in operations – many of our plants contain amazing chemicals and we don't always need to turn to the chemist for an answer. I'll leave you with one last thought for the year from Jack Johnson “*The wisdom's in the trees, not the glass windows*” Happy New Year to all!

Dave Kolotelo



EDITOR'S NOTES

It has amazed me how quickly this year has flown by! There has certainly been a lot of activities happening to keep us all busy. The Seed Workshop, held in August, was a success and was well attended. Dave and I felt that the theme of water activity was appropriate as this technology is being used by more people and the workshop was an excellent opportunity to introduce this technology to more people. The abstracts of the presentations are provided in the News Bulletin.

Melissa Spearing has provided an interesting article about the practical application of equilibrium relative humidity when collecting and drying seed prior to extraction and cleaning. Jack Woods and Roger Painter summarize results from a Gibberellin application trial conducted in lodgepole pine seed orchards. Dale Simpson summarizes some activities, accomplishments, and future endeavours of the National Tree Seed Centre.

I hope that you all have a great winter (for those of us in the Northern Hemisphere) and I look forward to sending you the next issue in July.

Dale Simpson
Editor

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Comments, suggestions, and contributions for the News Bulletin are welcomed by the Chair and Editor.

All issues of the News Bulletin are available at:
<http://www2.gov.bc.ca/gov/topic.page?id=4E4651B3A01448FAB6F39ACAD1C348C1>

TREE SEED WORKSHOP OVERVIEW

On August 17th the Tree Seed Working Group held a workshop in Fredericton, NB in association with the Canadian Forest Genetics Association conference. The theme was “Development and Application of Water Activity for Monitoring Seed Moisture Content”. The workshop was attended by 36 people and presentations were given by four people (see below) followed by brief presentations on provincial tree seed centres given by: Fabienne Colas (Quebec), Al Foley (Ontario), Lindsay Robb (Alberta), and Dave Kolotelo (British Columbia). In the afternoon participants visited the National Tree Seed Centre and the Atlantic Forest Seed Centre. Abstracts of presentations given by the four invited speakers follow this article.

Welcome Dave Kolotelo

Seed Viability Prediction Options
Dave Kolotelo

The Practical Applications of Water Activity to Improve the Efficiency of Seed Management Systems Bob Karrfalt

Using Water Activity in Tree Seed Banking and its Implementation in Alberta Lindsay Robb

The Application of Water activity to the Management of Pollen and Seeds in Quebec Fabienne Colas

I want to thank everyone who contributed to the workshop and the tours. Bob and Fabienne had an additional challenge by giving their presentations virtually but the technology worked well and I appreciate their willingness to do this and for their participation when answering questions and contributing to the discussion.

Dale Simpson

SEED VIABILITY PREDICTION OPTIONS

The need to be able to predict seed viability is important to collection and processing decisions. For collection decisions it is also necessary to have an understanding of the reproductive biology and anatomical changes taking place within the seed during development. The question of “when” and characteristics of immature and mature seed were reviewed. Emphasis were on Gymnosperm species, but some Angiosperm examples were presented. The differences between germinability and viability were discussed.

The second major question is ‘how’ and a review of available techniques including tetrazolium, hydrogen peroxide, incubation, x-rays and cutting tests was presented. Emphasis was placed on the use of cutting tests and x-rays and a variety of examples were provided discussing the advantages and shortcomings of each. Examples of seed deterioration and possible mechanisms were discussed.

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THE PRACTICAL APPLICATIONS OF WATER ACTIVITY TO IMPROVE THE EFFICIENCY OF SEED MANAGEMENT SYSTEMS

Water activity has great potential as a way to effectively manage seed moisture when storing seeds both short term and long term. It is low cost, easy to use, universally applicable to orthodox seeds, and rapid. In the practical application of water activity to seed management there are several considerations that will make it more understandable and workable for seed managers and their clients. The first point is the name. While water activity has meaning from a more pure scientific viewpoint, in practical application it is a foreign term. Relative humidity on the other hand is used in the daily conversation (e.g., weather report) and is what is actually measured in a water activity test. Therefore, equilibrium relative humidity is proposed as the more practical term. Bringing the seed into equilibrium with the air in the test chamber is critical and is influenced by seed size, seed coat, and rate of drying. Therefore, the effect of these factors is discussed. Adoption of a universal equilibrium relative humidity (ERH) is important both because time and resources are not sufficient to study each species in detail to find its optimum ERH for storage and the limits of the test equipment to measure the true ERH make a highly refined ERH recommendation meaningless.

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USING WATER ACTIVITY IN TREE SEED BANKING AND ITS IMPLEMENTATION IN ALBERTA

The purpose of this presentation was to explain water activity and seed moisture to seed practitioners at an introductory level that is easy to understand. This explanation leads into the discussion of Alberta's new regulations for seed storage, to be released soon in the revised Forest Genetic Resource Management Standards (FGRMS). Due to the nature of this article, it is difficult to provide sufficient background for beginners without visuals and in the space allowed. If you are interested and would like a pdf of the

presentation with texts for the slides, please email and I would be happy to supply this.

For seed practitioners in Canada, seed moisture has long been measured using moisture content (MC) on a wet weight basis. This method uses an oven and balance, which makes it relatively cheap, accurate, and reproducible. However, the method takes up to 18 hours to complete and is destructive to the seeds. Globally in seed banks over the past 30 years, there has been a move away from using the moisture content method to measuring water in seeds using equilibrium relative humidity as a measurement and standard for seed storage.

Most seeds gain and lose water fairly quickly; however, once the seeds are in equilibrium with the surrounding air, they stop gaining or losing and reach what is called 'equilibrium relative humidity'. This number is then a measurement of the air relative humidity at which the seed is neither gaining nor losing water. To do this we use a water activity meter, which measures the potential energy of water and is usually a non-destructive process that takes approximately 5 minutes. This water activity measurement is easily converted into equilibrium relative humidity (% eRH) for our purposes.

The use of eRH standards essentially puts all orthodox seeds on the same playing field by eliminating moisture differences due to differing oil contents in each species. An isotherm graph (Fig. 1) shows a generalized relationship between eRH and MC for any given species and the line can be divided into three water binding regions (Bewley et al. 2013). The first water binding region, usually near 0-15% eRH, represents the tightly bound water inside the seed that is generally not available for most processes. The second water binding region, generally ~15-80% eRH represents weakly bound water and the third water binding region, generally ~80-100% eRH, represents the very loosely bound 'free' water. The goal for maximizing seed longevity in storage is the apex between regions 1 and 2, where most of the water that could be used for ageing processes has been removed.

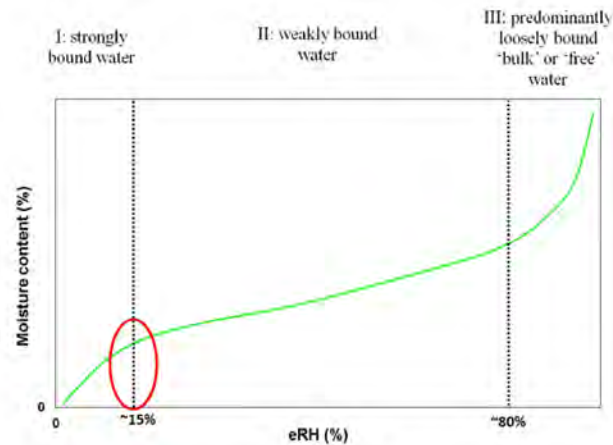


Figure 1. Generalized isotherm showing three water binding regions: strongly bound, weakly bound, and free water. Red circle designates desirable apex between regions I and II, where most of the water involved in the ageing process has been removed.

Figure 2 shows the eRH/MC isotherms for four important commercial tree species, one endangered tree species, and five non-tree species that are important to oil sands reclamation in Alberta. The different vertical positions of the species on the graph are a direct result of the different oil contents of each species, ranging from approximately 5% oil content for sedges to 60% for endangered limber pine (*Pinus flexilis*). The cluster of similarly positioned lines in the middle for lodgepole pine (*Pinus contorta*), Douglas-fir (*Pseudotsuga menziesii*), white spruce (*Picea glauca*), and black spruce (*Picea mariana*) are due to similar oil contents (~35%) for these four economically important species. However, adding the shrubs and other tree seeds to this graph produces the extremely wide spread and, as is shown by the orange bar representing the previous Alberta seed storage standard of 4–8% MC, the use of moisture content standards makes it impossible to store all species at optimal seed moisture levels. In the past, many of these species have been stored with moisture exceeding optimal levels. However, by using an eRH standard, as shown by the green bar, it is easy to ensure that all species will be stored with moisture levels that will ensure maximum longevity.

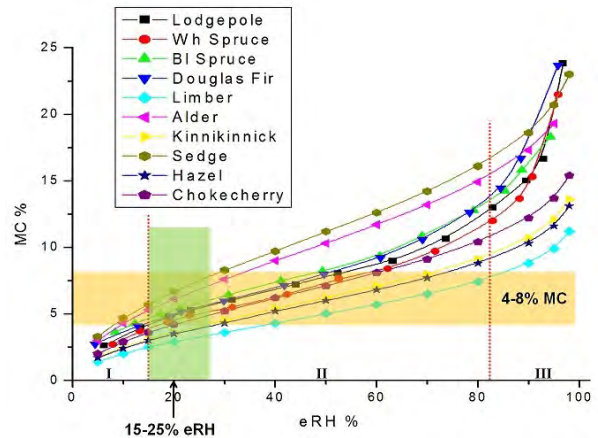


Figure 2. Isotherms of ten Alberta tree and shrub species. Oil contents range from approximately 5–60%. Dotted red vertical lines approximate the apices between the three water binding regions. The orange horizontal bar represents Alberta’s previous 4–8% moisture content seed storage requirement and the green vertical bar represents the new 15–25% eRH standard.

For this reason, Alberta is changing its seed storage standards in the newest revision of FGRMS to reflect the increasing involvement with non-economically important and non-tree species. When the revised standards go into effect, they will allow both the old 4–8% MC limits and the new 15–25% eRH limits for nine economically important tree species. All other species will only be accepted for storage by falling within the new eRH limits. In the future, all species will eventually be moved to comply with eRH storage limits as Alberta changes its storage protocols to align with international seed banking standards (FAO 2013).

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THE APPLICATION OF WATER ACTIVITY TO THE MANAGEMENT OF POLLEN AND SEEDS IN QUEBEC

Until now, moisture content (MC) measurements were used to determine the water status of forest reproductive material. This measure is destructive, time consuming, and provides only quantitative information. It does not describe the chemical availability of the water present in a compound, nor the resulting potential negative effects of water on the conservation of stored pollen and seedlots.

Measuring water activity (A_w) allows this distinction. This tool was primarily developed for the agri-food industry (Scott 1953). It describes a product's water status qualitatively. This, in turn, provides information about the product's degree of stability and predicts its conservation potential cited as "shelf life". Indeed, A_w and not MC governs the development of microorganisms and the production of toxins, and thus, the reactions associated with food degradation (Scott 1953).

In a given system, A_w is the ratio between observed water vapour pressure and the one of pure water at the same temperature. In other words, it is the equilibrium relative humidity (eRH) of the air surrounding a system at the same temperature (Rahman and Labuza 2007).

A_w describes the state of water in matrices, whereas eRH qualifies the surrounding environment. Measuring A_w also has the advantage of being rapid, easy, and most importantly, non-destructive. This allows a sample to be measured repeatedly during its conservation.

A Fruitful Collaboration

Thanks to technology transfer and collaboration between France's Irstea (formerly called Cemagref) and Quebec's Direction de la recherche forestière (DRF) of the ministère des Forêts, de la Faune et des Parcs, A_w measurements have been used in Quebec since 2007 for the treatment of forest tree pollen and seeds.

The first step of this work involved the production of sorption isotherms (Brooker et al. 1992) to characterize water relations in seeds of the various tree species used in Quebec's provincial planting program. We characterized a total of 10 conifer and 5 hardwood species, with results similar to those obtained for French forest species. By analysing the isotherms, we were able to determine the optimal A_w for the conservation of each species, and, for each, the resulting MC (Colas et al. 2010). We also determined the mean minimum MC which corresponds to combined water associated with cell membranes (monolayer water). Drying seeds below this minimum MC could impair their long-term conservation (Wang 1974, Sun 2002). The current target A_w for seed conservation in Quebec is 0.35 (Colas et al. 2010).

A water activity-regulated dryer was built from blueprints provided by Irstea (Baldet and Colas 2013). It enables the stabilization of seedlots at the targeted A_w . Its main feature is that it works without heat, on the basis of the 2-pressure principle developed by the National Institute of Standards and Technology (Hasegawa and Little 1977). It is simple and easy to set up and use, and allows the stabilisation of seed and pollen lots to a safe A_w level for their conservation in the best possible conditions.

Water Status During Conservation

The water status of seeds or any other product is in balance with that of the environment in which it is conserved (Freire and Mumford 1986; McDonald 2007). For optimal conservation, each seed or pollen grain should contain only the water necessary for its survival, and no more. A_w is a reliable indicator of changes in the water balance between seeds and their conservation environment (Baldet et al. 2009). Yet, humidity is a risk factor which is incompatible with good-quality conservation. Seeds must be protected from ambient eRH using airtight containers (Wang 1974), or kept under controlled eRH conditions (Freire and Mumford 1986). Unfortunately, no container is completely impervious. Water vapour will enter or exit as long as equilibrium is not reached (Walters 2007).

A trial aimed at developing a frequency table for germination tests at the Berthier Tree Seed Center (BTSC) revealed that A_w increased significantly in preserved seedlots (Colas et al. 2012), thanks to permeability of the polymer and repetitive opening of the containers. The non-destructive nature of A_w measurements was a real advantage in this study. After measurement,

seeds were stabilized to the optimal A_w in the dryer controlled by water activity and returned to conservation. The same lots can be measured repeatedly.

We began work to develop an optimal storage container that would improve the conditions for long-term conservation of forest tree seed in Quebec. This involved developing a new polymer which was less permeable to water vapour. Results are expected by the end of 2015.

Conclusion

A_w measurements are now used operationally in Quebec for seed and pollen lot treatment. This qualitative tool for monitoring the water status of stored lots was adopted because of its reliability and ease of operation. At the BTSC, A_w is now used to govern the final drying of seedlots during extraction and has been added to the list of quality control tests. Because of its non-destructive nature, A_w measurement will be an essential tool to verify the reliability of seedlot conservation in a conservation bank for forest genetic diversity. It is noteworthy that the International Seed Testing Association has scheduled the implementation of a standard for water activity in its 2015 timetable.

Acknowledgement

We thank Denise Tousignant (DRF, MFFP) for translating this text.

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**THE WELL-INFORMED COLLECTOR:
TOOLS FOR EQUILIBRIUM RELATIVE
HUMIDITY MEASUREMENTS IN THE
FIELD**

For independent seed collectors, instructive phrases like “mature seeds should be dried in a cool, well-ventilated place” are based on solid seed science but often subject to vague interpretation in the field. In past years of collecting, I went about judging “dryness” as best I could: using colour change indications, a hammer to shatter a sample or spreading out rained-on collections on a cloth with a fan on in my garage. It may be shocking to you in the professional and institutional seed research world where precision protocols and access to digital instruments are necessary for the vital work you do, but independent collectors are often stymied by lack of basic real-time readings on which to judge our actions. Is it even fully mature? Are the seeds dry enough or not? If you get seeds for your research from the National Tree Seed Centre that I collected, how do you or I really know what those seeds experienced at the beginning of their potential longevity?

During my time at Kew’s Millennium Seed Bank (MSB), Wakehurst Place, Ardingly, UK, in September 2014, I was fortunate to be shown very effective and simple ways of maintaining initial seed quality right from the moment of collection, complete with data exact enough to act on. The MSB places a strong emphasis on equilibrium relative humidity (eRH%) as a function of seed longevity, and conversely the impact that poor post-harvest handling activities can have on seed aging. Not until this level of training were the daily fluctuations of eRH% and the usefulness of hygrometers to collectors explained. In plotting Toronto’s climate norms (average monthly temperature and eRH% highs and lows) and comparing them with Israel, South Africa, and Ghana, I realized a double-whammy of circumstances that jeopardize orthodox collections.

In most of Canada, rarely are ambient conditions below 50% eRH, and the only ideal time (January – April) to effectively air-dry fresh collections is completely out of sync with most temperate species’ seed dispersal phenology. As an added bonus, low humidity can halt or greatly slow the growth of insect pests and fungi. Again, many seed collectors don’t fully appreciate that dry seeds collected on a warm September afternoon will reabsorb moisture at night, especially without a dew point hygrometer that would indicate potential for 90–100% RH temperatures to be reached. It is almost impossible without artificial desiccants in a sealed container or an air-conditioned, dehumidified room to maintain a safe level of eRH% for collections at all times regardless of the weather forecast.

In the two seasons since this training, I have invested in, begun using, and promoting an improved seed collection kit for field use similar to MSB’s “Blue Barrel” model. It was not expensive equipment. This kit is most useful for conservation-sized or single-lot nursery use collections (2,000–10,000 seeds) of orthodox angiosperms with dry fruits, depending on volume of non-seed debris. Large-scale conifer collections are typically better organized with immediate shipping to appropriate cone-drying facilities, though a probe hygrometer can be useful for inserting amongst shipping bags to determine if cones are overheating. Forgive me that some of the examples here utilize vegetable garden seed, but orthodox is orthodox and the principles are the same.

The foundation of a simple post-harvest handling decision tool lies in a reasonably good hygrometer with sensors in a probe tip, durable enough for dusty fieldwork. MSB used the Gemini Data Loggers Ltd’s (UK) Tinytag View models for their field kits, with a range of 0–100% RH (+/- 3% eRH) and -30 to 60°C (+/- 0.2°C) for £180 (Jan 2014 price). While my Christmas wish list still includes a Rotronic HygroPalm unit for all psychometric parameters and data logging ability, I was able to find Extech Instruments 445815 Big Digit Hygro-Thermometer and an 18”/45cm long remote probe on www.amazon.ca for \$150 CAN. Accuracy range was suitable: humidity 10–99% RH with +/- 4% RH accuracy, temperature range -10 to 60°C with +/- 1°C accuracy, dew point display and an alarm if a maximum or minimum eRH% threshold is reached. I calibrate mine every few months of use with the manufacturer’s kit, and when spending time at the National Tree Seed Centre (NTSC) this January, compared it to the Rotronic model (Fig 1). It only required minor adjustment to match. For the probe tip, I fitted a snug rubber car gasket (\$7 from

Canadian Tire) into a mason jar lid to sample fresh or dried seeds as the dominant fraction of eRH%. A 125 or 250 ml mason jar isn't too much extra gear to carry around in the woods. Using the decision-making chart from MSB's Technical Information Sheet #4 (http://www.kew.org/sites/default/files/04-Post%20harvest%20handling%20web_0.pdf) I have two simple measurements to take and compare: seed moisture from my sample container and ambient conditions the day (and evening) of collection. I then follow their recommendations for safe drying based on whatever I have available (Fig 2).

Summary of recommendations for effective post-harvest handling of seeds

Seed Maturity Stage	Seed Moisture Status	Ambient Conditions	
		"Dry" (daytime RH < 50%)	"Humid" (daytime RH > 50%)
Immature	85 - 100% eRH	Hold intact fruits under shaded, ambient conditions for 1-2 weeks*	
Natural dispersal	"Dry" < 50% eRH	Hold in loosely packed bags in a well ventilated, shady location. Minimise moisture absorption at night.	Transfer to seed bank as soon as possible OR Dry with desiccant OR Place in air-conditioned room
	"Wet" > 50% eRH	Dry in thin layer, in well ventilated location. Minimise moisture absorption at night.	

* Remove seeds from fleshy fruits as soon as morphological signs (e.g. fruit colour) indicate that they are fully ripe. Allow to dry slowly under ambient conditions before transferring to a cool dry-room.



Figure 1. Comparing my Extech hygrometer to the Rotronic HydroPalm 23-AW at the National Tree Seed Centre, top. Testing a batch of garden *Phaseolus* for long-term potential. Adjusted eRH% at 20°C is 33.7%, bottom.

Figure 2. Excerpt from “Post-Harvest Handling of Seed Collections” by Kate Gold, Millennium Seed Bank Partnership, Wakehurst Place, Ardingly, UK.

Many times this past year, I have been surprised to feel seed that is “dry” coming off the parent plant, only to put it in the sample container and it still reads 50–60% eRH. In speaking with Lindsay Robb at the Tree Seed Working Group Workshop this past summer, I also realize the caveat of adjusting eRH% readings to 20°C for standard comparison. Her recommendation to use the conversion tool at www.cactus2000.de/uk/unit/masshum.shtml makes this a breeze. Using my bucket example below, 29% eRH at 17.5°C converts to 24.8% eRH at 20°C (approximately 6% mc). If I were collecting for Lindsay using the 2015 Alberta standards, this would be a good starting point for their 15–25% eRH target and helpful for the seed arriving to the seed bank in a partially conditioned state for further testing. For the USDA National Seed Laboratory, 30% eRH is their target (Karrfalt 2014). With a measurable number in hand, I am more confident communicating with buyers about the potential quality of this collection and have flexibility in shipping around adverse weather conditions.

“Save for a rainy day” takes on a new meaning when you have silica gel. It is supremely useful both as an emergency drying agent when field conditions are above 50% RH and as an indication of moisture leakage in a hermetic vessel once seeds are safely dried. And, as I have found out, capable of maintaining stable humidity levels within a gamma-lid sealed 5-gallon pail (\$11 lid ULINE, \$5 Home Depot pail) for those times when you are too busy still collecting to check, clean or ship earlier collections (Fig 3). Once they are closer to 30% eRH, seed longevity is greatly extended and cellular damage minimized. I have also

purchased a 30-gallon airtight HDPE plastic drum with metal-locking lid and rubber gasket for larger collections with lots of or similarly weighted debris; crispy dry chaff separates much easier with my limited low-tech cleaning equipment. From www.silicagel.ca, I ordered both loose bulk indicating silica gel in 2 kg bags (\$14.10 CAN per kg when ordering 10 kg or more, a lifetime supply for a collector!) and 300 x 1 gram indicating packets at a non-discounted price of \$0.18 each (volume discounts as low as \$0.094 cents each) for placement inside 2 l mason jars of fully dried seeds of valuable or short-lived collections.



Figure 3. My 5-gallon pail with a screw-top gamma-seal lid and silica gel in the bottom. The hygrometer probe monitors eRH% around the paper seed bags.

Regarding the amount of silica gel used in my kit, MSB advises a general rule of a seed : desiccant ratio of 1:1 and a more exacting formula is given on page 601 of *Seed conservation: turning science into practice* (Smith et al. 2003). However, given that I use this pail as a temporary repository for many variations of bag sizes and species, and am aware of the dangers of drying too rapidly, I use the conservative manufacturer's recommendation of 1–2 g/l of volume. I change or add more silica gel as the hygrometer indicates after 7–10 days. I maintain fresh and recharged silica in mason jars and another airlock pail. This system works best with paper and cloth bags (not poly), and more can be put into the pail with a chicken wire cage column in the center to hang bags from.

To conclude, I suggest:

- eRH% is not just a tool for seed processors and seed banks; collectors should be made aware of and use it for high quality collections in humid climates with daily means over 50% RH.

- Instrumentation to do quick field measurements with suitable accuracy is cheap and easily accessible, and decision-making charts freely available online.
- Air-tight pails/drums and rechargeable silica gel are a practical field solution to begin drying seeds to a suitable eRH% prior to cleaning and storage.

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IMPACT OF GA ON FEMALE CONE PRODUCTION IN LODGEPOLE PINE ORCHARDS

Introduction

Operational stem injections of GA_{4/7} (GA) were applied in seven lodgepole pine (*Pinus contorta* var. *latifolia*) orchards managed in cooperation with SelectSeed Ltd. in mid-July 2014. This report presents the results of sampling done to evaluate the impact of the GA applications on 2015 female cone production.

Methods

GA applications followed protocols set out in Table 1. Applications were done in mid-July, 2014, following the recommendation of Painter (2014). Approximately 20 to 30% of the ramets

in each orchard were treated. Evaluations of GA impact were done in one orchard on each of four sites (Table 2). For each sample orchard a pair of ramets from each of 24 or 25 parental clones was chosen. One of the ramets in the pair received GA in 2014 and the other ramet did not receive GA. Ramets for each pair were chosen without bias to cone number and based solely on proximity in the orchard, similar crown size, and health (only healthy ramets were assessed). New female cones (2016 crop) were counted on all sample ramets.

Table 1. GA application protocols used for mid-July, 2014 GA treatments. GA injected per ramet was between the lower and higher rates shown. GA solution concentration was 80 grams GA_{4/7} per liter of ethanol.

Stem diam. (cm)	Lower rate		Higher rate	
	Total ml soln. per ramet	Total GA delivered (mg)	Total ml soln. per ramet	Total GA delivered (mg)
7	0.5	36	0.5	36
8	0.5	36	1.0	72
10	1.0	72	1.5	108
12	1.0	72	2.0	144
15	1.5	108	3.0	216
17	3.0	216	4.0	288
20	4.0	288	6.0	432
22	4.0	288	7.0	504

Results

GA application increased cone production between 24 and 41% across the four sample orchards, with an average increase across all orchards of 32% (Table 2). Variability in production among ramets from the same clone was substantial, however, across the four orchards between 60 and 88% of the paired samples had more cones on the GA-treated ramet than on the control ramet (Table 2).

A trend was evident for less GA impact in orchards with higher overall cone production (Fig. 1). A scatter plot of the 98 sample pairs showed a trend for higher cone production on GA-treated ramets relative to non-treated ramets from the same clone (Fig. 2). Most points in the scatter plot are above the red line that indicates equal production for treated and non-treated ramets. The trend line in this scatter plot is flatter than the red line, indicating more successful GA response in clones that tend to produce fewer cones ($R^2 = 0.44$). Similarly, a plot of the difference in cone production between the treated and non-treated ramets in each sample pair relative to the cone production on non-treated ramets shows a non-significant trend ($R^2 = 0.09$) to less GA success in clones with a greater propensity to flower, however, this trend is strongly influenced by a small sample of ramet pairs where the non-treated ramet had a large crop (Fig. 3).

Table 2. Cone count results by orchard for GA treated and non-treated ramets

Orchard	Site	Zone	Average number of cones per ramet		Percent difference	Percent clones with more cones on GA-treated ramets
			GA treatment	no GA		
238	Kettle R.	CP	62	44	41	76
339	Tolko	TO high	84	62	36	67
240	Sorrento	BV	127	94	36	88
338	PRT	TO low	156	125	24	60
	All sites		107	81	32	73

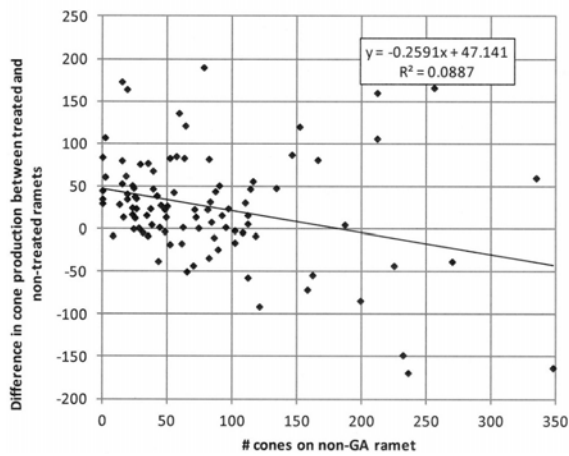


Figure 1. Percentage increase in cone production relative to the average number of cones on non-GA treated ramets, by orchard.

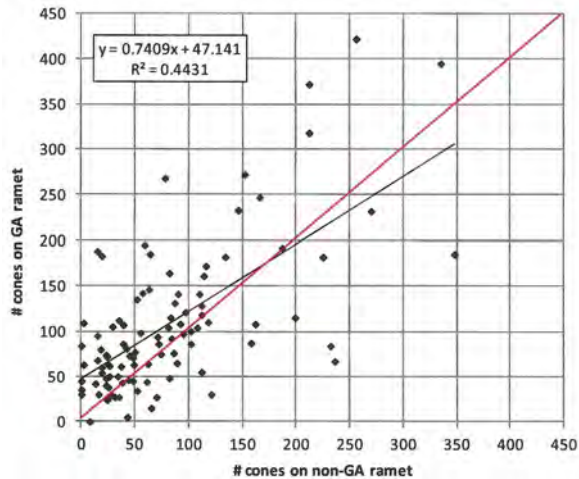


Figure 2. Relationship between the number of cones on treated ramets vs. non-treated ramets for 98 clonal paired samples. The diagonal red line indicates equal production for each of the ramets in a clonal pair.

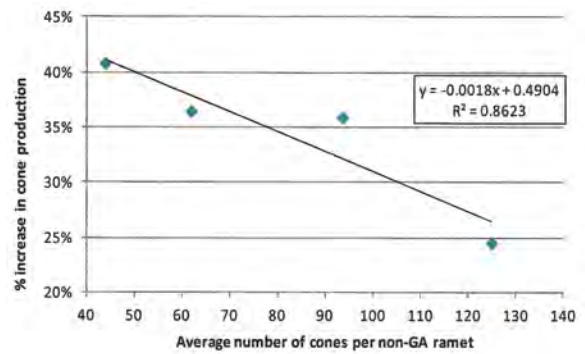


Figure 3. Relationship between the difference in cone number between GA-treated and non-treated ramets from each of 98 clonal pairs relative to the number of cones on the non-treated ramet of each pair.

Conclusion

Operational GA treatment in lodgepole pine orchards successfully increased cone production by approximately 32%. Orchards and clones with lower production appear to benefit more on a relative basis than orchards and clones that have higher cone production without GA treatment.

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Orchard managers and operational staff at PRT Armstrong, Tolko (Eaglerock), Kettle River Seed Orchards, and Sorrento Nurseries are thanked for their cooperation and dedication in the application of GA. Funding through the OTIP component of the Provincial LBIS Tree Improvement Program partially supported the operational GA applications in 2014. Funding for this assessment and report was provided by SelectSeed Ltd.

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National Tree Seed Centre

The National Tree Seed Centre will be 50 years old in 2017! It was established at the former Canadian Forest Service's Petawawa Forest Experiment Station at a time when reforestation programs were expanding in Canada and there was a need for technology development and transfer for activities such as seed testing and storage. Another role that the Seed Centre filled was the acquisition, collection, and provision of seed of known origin and quality for research; a role it continues to play today with about 80% of the requests coming from Canadian research organizations. The Seed Centre was transferred to Fredericton, New Brunswick in 1996 where the inventory of accessions from indigenous tree and shrub species continues to increase.

Ex situ conservation has become an increasingly important activity and builds on the thousands of collections already in storage. For example, there are over one thousand white spruce (*Picea glauca*) seedlots stored, most of which were collected from throughout the species range for a series of provenance trials planted in the early 1980's. Another ongoing project is the storage of ash seed in response to the threat imposed by emerald ash borer (*Agrilus planipennis*). The Seed Centre has a cryogenic facility where butternut (*Juglans cinerea*) germplasm, in the form of embryonic axes, is stored because seed cannot be stored under conventional conditions. Butternut is an endangered species so it is critical to be able to conserve its genome.

Staff have been active over the years at improving, modifying, and developing seed processing methods and techniques. Many of the methods used are similar to those used at commercial seed extraction facilities but are scaled down due to the small lot sizes. One method typically used by the Seed Centre for *Picea* and *Pinus* using absolute ethanol to separate empty from full seed was adopted for *Alnus* and *Betula*. It is impossible to separate winged seed of these two species and often germination was very low. By first de-winging the seed then placing the seed in ethanol so that filled seed sank and empty seed floated resulted in a substantial improvement in germination and no apparent damage to the seed (Simpson and Daigle 2013). An additional benefit is a decrease in the volume of seed for storage. The last several years have seen projects to optimize seed treatments for *Prunus* species to maximize and synchronize germination for nurseries in Alberta growing planting stock for oil sand site reclamation.

The Global Tree Seed Bank Initiative was developed in 2014 and funded by the Garfield Weston Foundation. It is a four-year project involving the Global Trees Campaign and the Millennium Seed Bank Partnership (MSBP). The initiative is working with existing and new MSB partners across the world to target seed collection from specific tree species. Canada will be participating in this project and activities will complement existing national and provincial efforts to conserve genetic resources. Seventy-nine tree and shrub species have been identified. The Seed Centre will be coordinating the activities and will be storing a sample from each of the collections. Seed collection will begin in 2016 and will continue until 2018.

The Seed Centre will continue to play an active role in conserving Canadian forest genetic resources and providing seed for research. The Centre is always interested in collaborating with researchers, universities, government agencies, and others to locate additional sources of seed and to participate in seed research projects.

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UPCOMING MEETINGS

31st ISTA Congress

June 15–21, 2016

Tallinn, Estonia

<http://ista-tallinn2016.ee/>

BC Seed Orchard Association

June 20–21, 2016

Langley, BC

<http://www2.gov.bc.ca/gov/content/industry/forestry/managing-our-forest-resources/tree-seed/events>

Seed Ecology V - Seeds in the Web of Life

August 21–25, 2016

Caeté, Brazil

<http://seedecology.wix.com/seedecologyv>



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